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RESEARCH ARTICLE

# Medical Document Clustering using Similarity Measures

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## ABSTRACT

There is an incredible development in the data rate in biotechnology research field. Scientists are trying to predict the diseases by finding the links between molecules or life mechanisms. Text mining systems can be helpful for researchers who are in search of precise answers to their big data analysis. Proposed system uses Similarity measures algorithms that are used in the data mining process to give solution to the unanswered queries. When a user post a query in a database or web pages, a long list of results will be displayed. It is the job of the user to read and analyze the results to find relevant answer. It is a time consuming process which can be replaced by using text mining system. For significant tasks such as information retrieval and web search, text processing plays a vital role where the system itself analyze the results and provides the appropriate solution to the queries posted by the user. The future for Big Data analysis is the emerging text mining technique.

**Keywords:** medical data mining; similarity measure; classification; clustering

## INTRODUCTION

This template, Medical data mining has an improvising potential for exploring the patterns which are hidden in the data sets of medical field. In order to proceed clinical diagnosis process, the patterns that have been discovered are used. Heterogeneous and voluminous medical data are widely available in a raw and unstructured format. Medical data should be collected and organized in a structured format. In healthcare centers and hospital, doctors and analyst used to test the patients and provides the personal and medical report. This data will be collected and can be integrated to form a hospital information system. User-oriented approach is required to search for the hidden patterns in the textual data.

The WHO (World Health Organization) had estimated that more than 12 million deaths occur every year worldwide due to heart diseases. Medical diagnosis is a complicated task that should be executed accurately for proceeding an efficient medical data mining task.

Text mining is a tool used to improve the quality of searching process in scientific information and discovering pertinent insights. The prospective for the Biotech industry is considerable in terms of efficiency and finance in research area. The progress of text mining process leads to the development in biotechnology field and benefits the patient sooner. For example in Figure 1, Text-mining tool can show us the genes which are associated with breast cancer:

Following are some of the limitations of current text mining tools. Some of the mining process are limited to abstracts or summary of the content and if the researcher is not subscribed to the text content, they cannot mine the full text content. Importing our own article within the TM tool could be hard because of technical challenges.
Document conversion( from PDF to XML Format) can be complex. Information could be searched within text database by using keywords and huge volume of data will be mined. It results in producing a list of results which are not always relevant to the query. Ever since the Internet development, tools for searching have improved and powerful solutions could be given to all the queries. On the other hand, there are some limitations in the results. When we are in need of some specific answers, we may obtain an wide-ranging record of sources in which we need to analyse and obtain the relevant answer.

## LITERATURE SURVEY

Introducing semantic information from WordNet ontology has been used to improvise text clustering quality. lexical chains concept have been introduced to extract semantically related textual words, which represents the textual content [1]. Different similarity measures and the issues related to it are discussed in this paper. Features are taken in to account to predict the patterns in advance [2]. Knowledge could be discovered from scientific articles that were stored in huge repositories. Classification and clustering algorithms have been used in order to segregate the data [3]. Explicit model called as Markov model is useful for classification. Presently support vector machine (SVM) and machine learning technologies have been used widely in text classification [4]. D - Fault dependency matrix is a methodical effusive model [5] which is used to catch the level of the system deficiency indicative data comprising of dependencies between observable symptoms within the framework.

Automated determination of free text phrases mapping to pre-defined concepts could aid in the annotation of notes that have been obtained from clinical reports. It increase the speed of processing systems that deals with natural language. Open text phrases are mapped to predefined concepts by using automated and rapid methods. The rate of the speed of natural language processing systems have been increased by predicting the associations between phrase and concepts[6]. Precise clinical decision making in medical monitoring depends on the fusion of multi-parameter signals. In order to deliberately manage the workloads on the medical monitoring device and to transfer the complicated training data, cloud computing technologies have been used[7]. Assistive gadgets such as wireless devices have been used to provide support to the elderly people and various methods are discussed[8]. Newspapers are the main source of news but unfortunately some of us need others help to gain the knowledge of News Reading Voice Activated Multilingual Newspaper Reading System. The technologies like speech synthesis, speech recognition and the web are integrated together in the system [9]. An important consideration is to make the project entirely self-sustainable through entrepreneurial means and to impart enough knowledge base to the participants to preserve the momentum even as the initial promoters' active involvement gradually ceases [10].

Sustainability of a healthcare tracking management system is required while implementing in rural communities. This is a survey paper which conducts survey on slum communities in India and proposes appropriate services system. The paper includes initial analysis and proposes ideas for value-added services [11]. In this paper, a robust approach for transmission of medical images with concealed patient information as watermark is evinced. In this approach, spatial domain digital watermarking technique is adapted wherein the patient information is embedded as watermark into the lower order bits of the medical image pixels. The watermark, which comprises of text data, is encrypted to prevent unauthorized access of data [12]. In-depth analysis of meaning is required to understand the medical patterns that have been selected in the form of arteries images. Several methods have been proposed for reasoning in a semantic way and it is based on resonance analysis which is cognitive [13]. Comparison of RVM and SVM Classifier Performance in Analysing the Tuberclosis in Chest X Ray is discussed [14].

### Automated Health Care and Diagnosis

People uses cloud computing features and can communicate between handheld devices and wearables through Bluetooth and cloud using wi-fi or data services. Visualization techniques are used to communicate the important data concerning the person's health and it is sent to cloud for obtaining feedback and processing. The current condition of the person diagnose report will be noted by the doctor and stored in cloud. Monitoring controlling process requires time and text mining mechanism in order to mine the dataset. Consider an example where feedback concept is used. If a data regarding diabetic person indicates that the sugar level in blood is high, then the cloud will process the feedback and sent the data to the gadgets or wearables on the victim to release an additional insulin dose. If the data indicates lesser blood sugar level, an SMS may be sent to the device which is handheld mobile phones informing the person to eat food regularly.

The process of diagnosis involves identifying the symptoms and based on the physician knowledge, symptoms should be matched. Figure 3 shows the text mining mechanism in which a given set of discharge sheets are analysed and the suitable match to the symptoms should be identified.

Number of discharge sheets are stored in the cloud server and instead of manual intervention, automated diagnoses is done by the text mining and information retrieval process. The Text mining software finds a discharge sheet with almost similar symptoms and return the diagnosis listed on the discharge sheet. Software would identify the symptoms with the discharge sheet in Table I which lists "Asthma" as the analysis. By analyzing different set of discharge sheets belongs to same location, we can find the major issue prevails in that particular area.

## PROPOSED METHODOLOGY

### Data set

The most sensitive data is medical data that is used for diagnosing and monitoring the health of an individual regularly and data should be private. US Law insist to get patient's consent for using their medical data without mentioning the personal details. For machine processing, accurate medical information is required but it should not be identifiable and it should be available for automated healthcare needs.

### Labeling

Each report should be precisely labeled with the name of the disease. Same disease could be mentioned using different word with similar meaning and henceforth, Synonym word replacement methodology can be used as a preprocessing step.

Each discharge sheet or report should be precisely named with disease name. Semantic web techniques can be used in connecting the written diagnosis to a typical ontology. Labeling is essential for proceeding the similarity measure task.

## Algorithm used

Discovering the discharge sheet that accurate matches the given symptoms is the main goal. K-nearest neighbor algorithm is used for this process. All the documents should be considered as vectors and it can be visualized as points in a multi-dimensional space (Vector space model). Figure 3 shows the two dimensional visualization of documents represented as vectors.

Each point should be labeled and if the points are closer, it is more similar. The point's location is based on the significance of the words the contains. Functions presented in 'R' language can be used to plot the vectors.

## Similarity Measure for Medical Data Processing

Medical Dataset is used as input information for the system. Necessary pre-processing steps such as stop word removal, stemming and synonym word replacement should be done. K-nearest neighbor algorithm should be used for medical document clustering. Similarity measure for medical data processing(SMMDP) is the technique used for similarity measures. The system architecture is displayed in figure 4.

Following are the three cases that is to be considered.

Features are presented commonly in both the document group.
Features are presented in either one of the document group.
Features are not presented in both of the document group.
Lexical Pattern Clustering of Medical data

A semantic relation should be considered for effective pattern clustering. For obtaining patterns that are semantically related in medical data, Lexical pattern clustering algorithm is be used. The word co-occurrence refers to occurrence of two words at the same time was obtained. Threshold value is required to group the documents in cluster format.

## Lexical Skip Pattern Clustering Algorithm

**Input:** patterns P ($p_1$, … ,$p_n$), threshold $\theta$
**Output**: Clusters C
SORT (P)
C ← {}
For pattern $p_i \in P$
do
max ← - ∞
$c^*$ ← null
for cluster $c_J \in C$ do
sim ← cosine ($p_i$, $c_J$)
if sim > max then
max ← sim
c ← $c_J$
end if end for

if max > θ then
c* ← c* ⊕ $p_i$
else
C ← C ∪ {$p_i$}
end if
end for
return C

### Text Mining in Health Care Industry

The healthcare industry is a enormous spender on technology with the propagation of hospital management systems. To log patient statistics, devices which are low cost are used. Since there is an increase in the patient data abruptly, it is necessary to mine the comments in doctor's discharge sheets. Outputs of the mining process can yield information which benefits the healthcare industry in following numerous ways and displayed in figure 5.

The high risk diseases are isolated by frequencies of keyword per region. The mix of medicines to stock is leveraged to optimize on the inadequate outlets store. It should be noted that the frequency of disease related keywords will change. An early warning system can be built by mining the medical data. For example, if the keyword frequency of lungs or breathing exceeds 70 appearances, that too specifically in the last month for a given region, it can be a hint to excessive environmental conditions that results in respiratory problems.

## CONCLUSION

Text mining is the recent research development area and it is widely used for mining medical data. An enhanced similarity measure SMMDP is proposed to gauge the similarity involving in the document sets. Lexical pattern derivation technique is used for extracting semantic relations between the words in the medical documents. When a user enters the query in the form of keyword, similar documents will be extracted and clustered.

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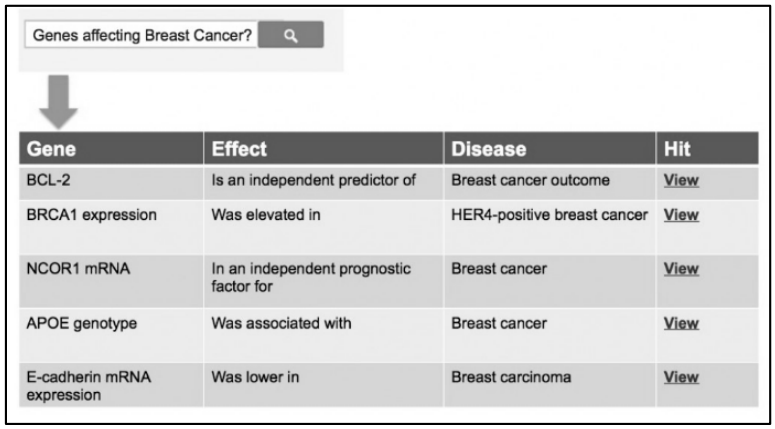


| Gene | Effect | Disease | Hit |
|---|---|---|---|
| BCL-2 | Is an independent predictor of | Breast cancer outcome | View |
| BRCA1 expression | Was elevated in | HER4-positive breast cancer | View |
| NCOR1 mRNA | In an independent prognostic factor for | Breast cancer | View |
| APOE genotype | Was associated with | Breast cancer | View |
| E-cadherin mRNA expression | Was lower in | Breast carcinoma | View |

**Fig.1Results of Text mining tool displying genes associated with cancer**

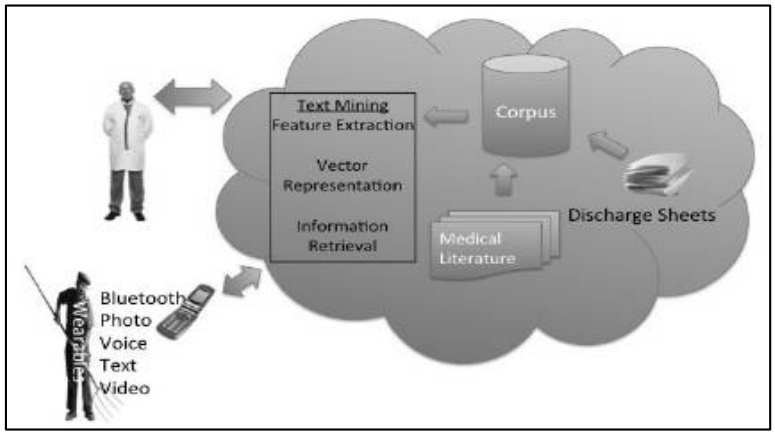


**Fig.2.Automated Health Monitoring and Diagnosis process**

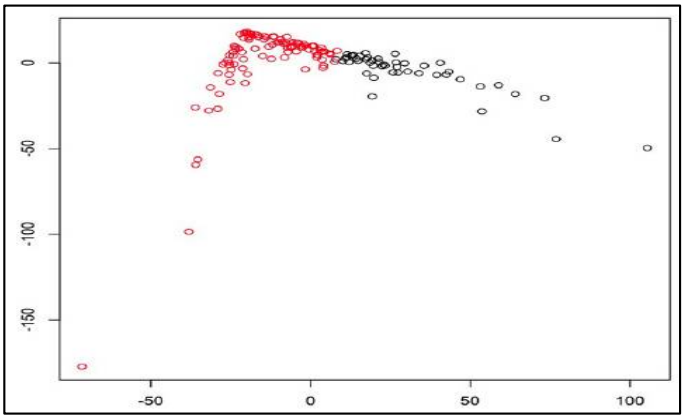


**Fig.3.2D Visualization of documents**

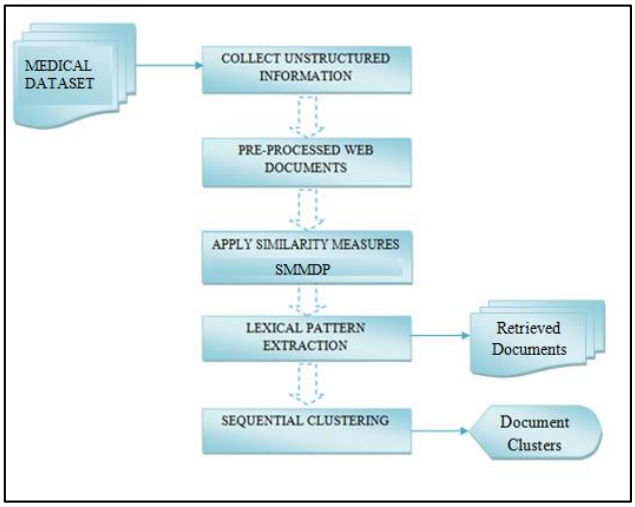


**Fig.4. a.System Architecture**

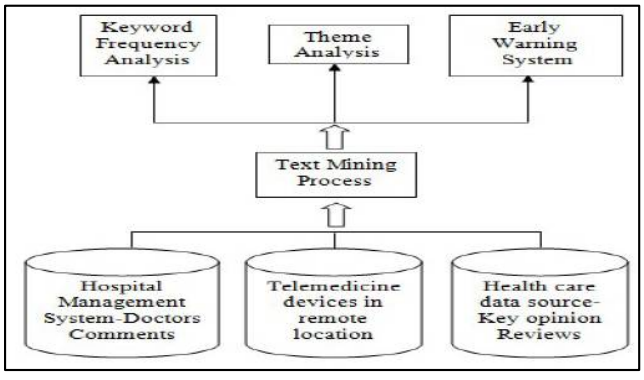


**Fig.4. b.System Architecture**

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RESEARCH ARTICLE

# Image Seclusion Trespassing in OSN Using Watermarking Techniques

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## ABSTRACT

An interpersonal interaction between users (moreover informal communication site on the web, SNS or web-based social networking) is an online stage that is used by people to develop interpersonal organizations or social relations with various people who extent comparable non-open or profession interests, exercises, foundations or genuine presence associations. Social networking sites are various and that they comprise a selection of latest information and communique tools which include availability on computer and laptops, cell gadgets inclusive of tablet computers and smart phones, digital photograph/video/sharing and "web logging" diary entries on-line (blogging). While Online Social Networks (OSNs) permit users to share snap shots without difficulty, they also divulge customers to numerous confinementthreats from each the OSNs and external entities. Image over the social network is transferred or transmitted among servers and a couple of customers. Privacy of that statistics could be very important as it belongs to personal touchy facts. In present gadget, textual content primarily based encryption can be implemented in social networks. There are many extraordinary approached of storing data securely over the social networks, using large records together with give up-to-stop encrypted facts transmission, dynamic credential era best for textual content data. In this paper, introduce a novel watermarking scheme with wavelet set of rules named as discrete wavelet rework in actual time social community utility. In this scheme can use images and saved in server in cozy format. And also make bigger the paper, categorize the image as sensitive or normal. If it's miles touchy manner, perform copyrights algorithms. Then offer the permission to the receiver end for download the images in comfortable manner. And also implement safety controls to dam mouse operations and print display options. Experimental result may be suggests that during real time social community environments and comparative observe of existing algorithms based on computational time and confinement rate.

**Keywords:** Online social networks, Image Acquisition, Privacy violations, Digital watermarking techniques, Wavelet co-efficient values

## INTRODUCTION

In today's years, online Social Networks (OSNs) have attracted many hundreds of thousands of customers worldwide. Even though Social Networks have perpetually been a primary part of day-to-day life, now that more and more oldsters are connected to the internet, their on-line counterparts are enjoyable a more and more essential feature. OSNs have also grown to be a scorching difficulty in regions of research ranging from sociology to computer technological know-how and arithmetic. Except for enabling customers to create a community to symbolize their social ties, many OSNs facilitate importing of multimedia content material cloth, pretty quite a few techniques of communiqué and sharing many factors of everyday lifestyles with friends. Humans can keep in contact with (bodily some distance off) acquaintances, pretty certainly percentage content cloth and experiences and hold up-to-the-minute within the alleviation in their own domestic or whilst at the flow. Social network systems offer an easy human computing device interface for internet clients, making it clean to proportion unlimited-shape statistics (equal to photographs and films) with buddies wherever and whenever. Additionally, clients can revel in real-time and unfastened chats with others, post the brand new status updates/affirm-ins, and categorical critiques about present social sizzling spots. On the grounds that social networking's introduction, we've substantive a few massively advantageous structures emerge (including fb, Twitter, and Instagram). When surfing on such systems, most clients are blind to the platform's privacy troubles; however truly, users' social community privacy is primary. Some touchy information equal to a personal alternative, profile, and shared photos will be leaked to others who aren't granted entry rights, if the social media carrier supplier doesn't take first-class precautions to protect entry manipulate. It's plain that most of the people social community platforms intention to preserve their consumers' privacy as loads as they may be capable of. Nevertheless, blessings aside, advantage threats to user privacy are extra frequently than no longer underestimated. For example, due to the general public nature of many OSNs and the internet itself, content material can easily be disclosed to plenty broader viewers than the individual intended. Users extra frequently than no longer have drawback revoking or deleting understanding and information some customers would possibly also be posted by way of using others without their consent. Confinement in OSNs is a complicated remember and isn't at all times intuitive to clients, specially whilst you take into account that it's on no account instances similar to how privacy works in real-existence interactions. Ideally, clients need to be successful to alternate a few confinements for functionality, without their know-how becoming handy past the scope they intend. For instance, a customer of self-help OSN would like to satisfy parents with the same medical scenario however does not want every person to recognize about his ailment. Even in a great deal much less extreme times, the price of confinement is most of the time underestimated. In this paintings, we spotlight that the essence of the scenario is that present mechanisms for outlining entry to pics in OSNs, cannot honestly control instances in which the involved parties have conflicting settings. First, the photo uploaded is regarded the owner of the snapshot and is granted full rights, whereas the oldsters displaying in the photo must no longer seemed co-homeowners and have to no longer granted any rights. On high of this well-known coarse-grained procedure, OSN companies put into effect additional insurance policies, a number of so that it will significantly complicate problems. Moreover, any users which can be tagged have an effect at the visibility of the image, because the picture will likely be viewable by way of all their contacts (default privacy environment). Therefore, even when the users tagged within the photo have confined its visibility, if the uploaded has now not confined entry the photograph will in all likelihood be publicly available, something which the remainder users isn't always going to even be conscious of. Generally, these events may also be characterized as cases of conflicts of hobby, the location they want of the content material cloth author goes in the direction of the choice of the depicted users, or the privacy settings of consumer override those of 1 other. Note that although the access manipulate mechanisms may want to range at some stage in OSNs, conflicts of hobby are a normal impediment, as they stand up from the content material fabric of the snap shots. The various varieties of social networks are proven in figure 1.

### Related Works

H. Cheng, X. Zhang, et.al, [1] Proposed a remarkable plan for encoded JPEG illustrations, where intra-piece, between square, and between viewpoint conditions among DCT coefficients are presented. With this plan, the scrambled

JPEG depictions additionally can be sold through a mix of the float figure and change encryption and outsourced to a server. Furthermore, further, with the given encoded question picture and the scrambled database previews, it's miles advantageous for the server to figure their likenesses in scrambled area by means of utilizing the procedures of a Markov technique and multi-classification manage vector pc (SVM). As the reason for the plan is to manage the test of picture recovery in scrambled space even as keeping the record length and structure consistence for JPEG photographs, appropriate ideal here, we first adopt an incomplete picture encryption strategy underneath thought to encode JPEG photographs. The bind is intricate to clear up for the basic cryptography. Presumably the most current incomplete encryption structures for JPEG pictures are frequently arranged on squares rearrange, DCT coefficient stage, and scrambling the indications of DCT coefficients. The proposed encryption strategy aren't ready to least difficult meet the necessities of format consistence and report length upkeep yet what's more allow precious data with respect to the time of every factor length whole number (VLI) code for DCT coefficients. It implies that it is anything but difficult to in any case aggregate the credible size of any VLI code concerning DCT coefficients from a scrambled JPEG picture. On account of the conditions of DCT coefficients in everything, their comparing VLI code size ought to have same connections, which may likewise be abused to create trademark for photo recovery.

A. Rial, M. Deng,et.al,... [2] Combining encryption with virtual watermarking, a purchaser– merchant watermarking (BSW) convention is effectively an uneven fingerprinting convention the area the unique mark is inserted by method for the utilization of watermarking in the scrambled locale. The essential motivation is that each customer acquires a scarcely particular imitation of the computerized material gave by utilizing the seller. This kind of trade, the watermark (or unique finger impression), does now not hurt the perceptual best of the computerized and can't be easily expelled through the purchaser. Because of the last resources, when a malevolent purchaser redistributes a pilfered propagation, the vender can relate the pilfered proliferation to its purchaser through its installed watermark. On the elective hand, a noxious merchant aren't ready to outline a genuine customer because of reality the customer's watermark and the conveyed watermarked material are obscure to the dealer. The principle commitment of work is a formal insurance assessment of BSW conventions. Furthermore, enlist the brilliant world/real worldwide worldview to characterize security of anonymous BSW conventions. With acknowledge to traditional rough fingerprinting plans, which diagram each wellbeing property each one in turn, this definition results inside the improvement of conventions that are secure beneath sythesis. The definition is regular in the surface that it catches the security homes required for any copyright insurance convention that offers clients with obscurity. Also, we diagram security for visually impaired and intelligible watermarking plans, and examine the homes that watermarking plans need to accommodate the advancement of loose BSW conventions.

J. Zhang, Y. Xiang,et.al,..[3] Implemented substance based photograph recovery (CBIR) is a beguiling programming that can be accomplished all the more adequately on distributed computing. CBIR objectives to go looking virtual pics from full-measure photograph records contraptions introduced on their unmistakable substance material depicted with the helpful asset of components alongside shade, surface and frame. On distributed computing, CBIR frameworks can serve additional easily by methods for sparing the calculation time of photo assessment and purchasing. The unbalanced productivity and flexibility of distributed computing may furthermore change the organization of CBIR structures. In the first place, the patrons and their parts in a photo recovery watermarking convention are sublime from the ones out of a purchaser– merchant watermarking convention. In a customer–provider watermarking convention, the dealer is the proprietor of a virtual substance material fabric, who directs the watermark inclusion, and the client can get a watermarked advanced substance material texture. In assessment, in a photo recovery watermarking convention, the purchaser is the proprietor of an inquiry picture, who should embed a watermark to defend its legitimate, and the backer supplier of CBIR will look photographs in accordance with the watermarked question picture procured from the client. The other makes some present security alternatives inapplicable; e.g. the appropriate response of the unbinding issue for a customer– dealer watermarking convention is inapplicable in a photograph recovery watermarking convention.

T. Bianchi and A. Piva, et.al,... [4] Has been tended to presenting calm watermark implanting, it's components the region the watermark installing is done in a way that the substance fabric proprietor does now not have get section to the last watermarked variety, even as never again uncovering the ordinary substance material. Alternatives exist to securely and accurately implant a watermark each at the server's side and at the customer's angle. Loose server-side implanting might be connected as a building obstruct in unbalanced unique mark conventions, providing a cryptographically comfortable strategy to the buyer's rights obstacle, at the equivalent time secure shopper factor installing gives a terribly effective alternative to the technique adaptability wander. The nearness of endowed verifiers can likewise be understood by turning to quiet watermark recognition, i.e., to an intuitive proof plan the area the substance material proprietor persuades a further entranced social affair that his/her substance texture conveys a given watermark without revealing unstable data that could encourage the watermark expulsion, much the same as the key of the watermarking calculation or the real watermark. In the resulting segments, we can show the previously mentioned strategies, searching for to outfit the peruser with a conspicuous running out in their benefits and their acclaim boundaries. The paper will stop with an exchange about conceivable new examinations charges, spend significant time in investigate challenges which can be particularly interesting for the sign preparing neighborhood.

A. Piva, T. Bianchi,et.al,... [5] Represented through the customer viewpoint watermark inserting: on this case, a server– supporter structure is once more embraced; despite the fact that, in this illustration, the server is allowed to convey a nitty gritty copy of the substance material to every one of the customers with the guide of broadcasting methods, without the need to create one-of-a-type watermarked duplicates (thus evacuating the bottlenecks remunerate in the server-factor watermark installing process); rather, every buyer should be responsible for implanting an individual watermark deciding the obtained copy. On this case, nonetheless, considering that the customers are endowed, fitting options should be conceived never again to enable pernicious clients to have right of passage to the normal substance material texture or to the watermark to be embedded. Another approach, said as agreeable watermark inserting, has been proposed for going through this sort of issue: here, the server transmits the indistinguishable scrambled variant of the long-set up work to the majority of the buyers, however a client unmistakable puzzle makes it feasible for decoding of the substance material and at the indistinguishable time understood implanting of a redid watermark, procuring an interestingly watermarked adaptation of the work. In unmistakable, we securely planned a LUT-set up quiet shopper side implanting strategy empowering us to insert a spread wind up dither tweak (ST-DM) watermark. As it's far going to be checked inside the accompanying segments, this adjustment shouldn't be simple; while you review that the buyer perspective inserting system forces a couple of imperatives that don't enable us to implant a characteristic ST-DM watermark. Regardless, the exploratory impacts will check that the pervasiveness of ST-DM versus SS watermarking displayed inside the established installing plans is kept up furthermore inside the buyer angle inserting way.

## Existing work

Social networking web sites might be a new international to create social family individuals amongst human beings that proportion facts like text, picture, videos, activities, pursuits, backgrounds or day-to-day-life connections. A social community issuer consists of an instance of every user (commonly a profile), his or her social links, and number further offerings. Social network internet web sites are an internet-primarily based provider that lets in humans to make a public profile, to make a list of users with whom to proportion connections, and evaluate and move the connections in the method. Probably the most nicely-preferred social networking websites are face-eBook, Gmail, yahoo, LinkedIn, Google plus, Twitter, and plenty of others. Communications over the Social Networks don't appear to be secure. Many attacks and violation of privacy are lately confronted in our most elegant networking web sites. We use the social networking web sites for speaking to our buddies and sharing digital know-how like textual content, snap shots, video and so forth. After we percentage a digital statistics to our friends; the know-how may just face a few attacks from the attackers and/or unauthorized customers. In the direction of this verbal exchange replacement legal users or third parties shouldn't be involved. Any unauthorized customers make a try to attack a

communication that is attempting to get entry to the picture for modifying or misusing. The attacker's best reason is to make crime utilizing the personal digital facts from social networking websites. The attacker tries to assault the conversation in masses of approaches i.e., violates the privacy, records attacking from the servers, etc.

## RDH established method

The present process is to guard extraordinarily non-public, different or secret understanding from unauthorized customers. Here, privacy safety is a maximum essential limitation of many social networking websites. And paintings utilizing Reversible statistics Hiding (RDH) techniques go to gather its significance as a consequence of the exponential growth and mystery verbal exchange of capabilities man or woman over the net. All social networking web sites' architectures include range of servers, databases, web site, records like text, image, and video and so on. In existing paintings, consumer try to upload an image, the frontend program embed some confinement facts into the photograph using Reversible statistics Hiding (RDH) gadget utilizing and additionally retailer encrypted snapshot into database. To showcase this photograph on friend's wall, the frontend software exams the pix embed privacy knowledge suit with buddy's confinement knowledge. If each confinement information's are equal, then best the snapshot is visible to the acquaintances. Or else, the person is not a pal so the photo simply is not apparent. Right here, first gadget is embedding and second one is to maintain the encrypted image into database. Ordinarily of know-how hiding, the picture will expertise a few distortions because of understanding hiding and cannot invert again to the original photograph item. That's, some parameter distortion has befell to the duvet object even after the hidden records were extracted out. Within the Reversible expertise hiding, each photo and information are equally principal. The Reversible records hiding gadget, the customary cover item lossless recovered after the message is extracted. The prevailing framework is shown in fig 2.

## Broadcast encryption scheme

The improvement of confinement-keeping information is accomplished with the aid of the data owner on every occasion man or woman records want be shared. The essential concept is that the info owner has proper manage over get right of entry to his/her non-public records, notably the ones revealing identification knowledge and personal lifestyles (e.g., photographs, motion pictures, copyrighted materials). More typically, the facts proprietor could act as a group manager who classifies contacts consistent with their roles (e.g., family, coworkers, and excessive college classmates, wearing activities membership members) and materials them the corresponding memberships. Every function defines a subgroup, the individuals of which can be restrained to targeted understanding classes. A knowledge category is created by using the information owner describing the set of statistics files that can be accessed as an entire with the aid of way of 1 or more subgroups. The granularity of information instructions is adjustable depending at the fineness of preferred access manage. For example, while the types are coarsely mentioned as track, films, pix, my testimonies, and so on, a subgroup of contributors who're permitted to a class can entry the complete information in that class. This is typically undesirable because the data proprietor can also want to free up awesome information only to related men and women (e.g., loved ones pix or videos handiest to be had to cherish ones people). The records owners could have the liberty to create their own lessons situated on the quantity and type of their subgroups, which is a design dilemma and will not be elaborated extra. Broadcast encryption permits for a applicable transmitter to send encrypted knowledge to a set of customers such that high-quality a privileged subset of users can decrypt the facts. Broadcast encryption is designed for and in large part implemented in the at ease distribution of copyrighted media over the net. The posted encryption steps will also be defined in fig 3. Other features of broadcast encryption contain encrypted document structures (e.g., home windows EFS) for constrained record sharing, mailing file programs for sending exceptional emails, and so forth. This requirement states that facts privacy is preserved within the presence of collusion attacks the location two or greater entities collude to obtain extra data at the sufferer than what is available to each colluding man or woman.

**Proposed framework**

Images at the social networks, execute 3 main protection traits: Confidentiality, Integrity and Authenticity. Confidentiality means that simplest the entitled humans have the access to the particular images, as a result tagging. Integrity way the picture has not been modified through non-accredited man or woman. Authenticity is the evidence that picture has indeed the extraordinary individuals as proven, or is a modified variation using the quite a number photograph processing programs.

The increment within the development and use of software image editors has accompanied the increase in the tampering of these normal traits. Specifically, the flourishing use of social networks has made the sharing and distribution of photos quite handy. The integrity and authenticity is the compelling question as, amongst exclusive fields, those pictures are also being used as proof in the courts of law. It is extraordinarily crucial to verify the integrity of these photos and is most often captivating to establish if a photo has been manipulated from the time of recording. To have knowledge of, how things cross inside the history of a jpeg photograph, we can put into effect watermarking process to cover default pattern into image. Watermarking also can be accomplished making use of discrete wavelet turn out to be. In numerical evaluation and beneficial analysis, a discrete wavelet change into (DWT) is any wavelet remodel for which the wavelets are discretely sampled. As with exceptional wavelet transforms, a key capabilities it has over Fourier transforms is temporal resolution: it captures every frequency and region understanding (vicinity in time). Water mark bits are embedded into photo. The discrete wavelet grows to be algorithm is outlined in fig 4.

Based in inverse DWT, we will get the scene water mark that can be restored into customary image. In the interface aspect, we will exchange the color of textual content pixels into color of photograph pixels. So photo may also be considered as undeniable content. Headquartered on this atmosphere, hacker complicated to grasp in regards to the photograph safety. Person can set confinement settings to dam the pictures to down load by way of third parties. So unauthorized users most effective get watermark information handiest. Then utilizing disable options in mouse right click on and print reveal options. Snapshot confinementis maintained in social networks. Moreover, the idea of boycotts and their organization are not accepted by any of these entrance control models. The utilization of substance construct separating in light of messages posted on OSN client dividers represents extra difficulties given the short length of these messages other than the extensive variety of themes that can be talked about. Short classification has recognized up to now couple of considerations in established researchers. This classifier will be utilized as a part of various leveled methodology. The primary level assignment will be ordered with positive and negative marks. The second level go about as a negative, it will create progressive participation. This review will be utilized as succeeding stages for sifting process. Short classifier incorporates portrayal, machine learning based grouping. The proposed structure is appeared in fig 5.

## METHODOLOGIES

SNS have emerged as very famous considering that they have many attracting capabilities for the customers. Most social networking web sites allow member to design their own profiles so as to design their profile web page so that it will specific themselves and to mirror their character. Users can customize the profile layout, add applications and may upload pix and different kind of information. SNS also carries Friends listing, containing different customers of SNS. Through SNSs users can preserve in contact with friends and circle of relatives, they could locate antique pals; contact buddies of friends, and even can touch humans they didn't previously known at all. Some SNSs also assist customers to find a activity or set up enterprise contacts, together with connecting with customers, partners and in locating out jobs and commercial enterprise possibilities

### Social network creation

Social community refers to interplay among human beings wherein they create, percentage, and/or change information and ideas in virtual groups and networks. A social community supervisor is the character in an organization depended on with monitoring, contributing to, and filtering, measuring and otherwise guiding the social media presence of a logo, product, individual or corporation. In face e book, GUIis a sort of person interface that allows customers to engage with customers through graphical icons and visible indicators inclusive of secondary notation, instead of textual content-based interfaces, typed command labels or textual content navigation. In this module, we are able to have 3 kinds of users consisting of image owner, picture users and image server. Image proprietor may be adding the photo into system and photograph server shops the pictures in database. Image customers use pics which might be shared by photo owner.

### Upload image

The first level of any sharing gadget is the photo acquisition level. After the image has been obtained, diverse techniques of processing can be carried out to the image to carry out the various exceptional imaginative and prescient obligations required these days. However, if the image has now not been acquired satisfactorily then the meant tasks might not be practicable, despite the useful resource of a few form of photo enhancement. The basic -dimensional photo is a monochrome (greyscale) picture which has been digitized. Describe picture as a -dimensional light intensity characteristic f(x,y) wherein x and y are spatial coordinates and the cost of f at any point (x, y) is proportional to the brightness or gray cost of the picture at that point. In this module, we will upload various photographs which include herbal pics, face pictures and other pix. Uploaded pics can by way of any type and any size.

### Embed the watermark

In this module, we can embed the watermark text into pictures. Digital media can be saved correctly and can be manipulated very without difficulty the use of computers, resulting in numerous safety troubles. The hassle of defensive the copyright of digital media may be solved by virtual watermark. Digital watermarking is an idea of hiding possession statistics into the multimedia facts, which can be extracted in a while to prove the authenticated proprietor of the media. Watermarking guarantees authenticating possession, protecting hidden records, prevents unauthorized copying and distribution of pics over the internet and guarantees that a virtual picture has now not been altered. We can implement Discrete Wavelet Transform (DWT) area image watermarking system for real time photograph. In the embedding method, the watermark may be encoded into the cover photograph using a particular area. This location values is used to shield the photos. The output of the embedding technique, the watermarked picture, is then transmitted to the OSN home web page.

### Privacy settings

Each user photos are first labeled into confinement coverage. Then confinement rules of every pix may be categorized and analyzed for predict the coverage. So we adopting levels approach for policy recommendation than applying the common one-degree facts mining tactics to mine both image capabilities and regulations together. The two-stage approach permits the system to appoint the primary stage to classify the policy as with privacy or without privacy. In the second level, we can set without privacy manner, pick the person list information.

**Protection system**

In this module, we are able to set the safety or blocking gadget to avoid 1/3 celebration aces without knowledge of photograph owners. This module is used to set the photograph with privacy. If consumer set with confinement settings approach, all users are taken into consideration as 0.33 events. Based in these putting, unauthorized consumer most effective perspectives the photo and can't be used. If he downloads approach, best get water mark values. Finally provide hardware control system consisting of mouse controls and keyboard controls. Then disable the mouse operations and gadget print display screen alternatives. Mouse code and print display controls values are extracted and to provide coding implementation to disable the coding as fake settings. We can enforce this idea in all browsers and to put into effect in all pics which are shared via social users.

## CONCLUSION

The appearance of famous on line social networking has precipitated in the compromise of conventional notions of privacy, really in visible media. With a view to facilitate useful and principled protection of picture privacy online, we've were given furnished the layout, implementation, and evaluation of picture protect machine that correctly and effectively protects purchaser's image privacy throughout well-known OSNs. The virtual watermarking method primarily based fully on DWT coefficients amendment for social networking services has been presented on this paper. In the embedding manner, the coefficients in LL sub-band had been used to embed watermark. Within the extraction process, everyday coefficient prediction primarily based on mean clear out is used to boom the accuracy of the extracted watermark. On extending the Machine Learning (ML) textual content categorization techniques to automatically assign with each quick textual content message a fixed of classes based on its content material. Then exploiting a bendy language to specify Filtering Rules (FRs), through which customers can kingdom what contents, should now not be displayed on their partitions. FRs can assist a ramification of various filtering criteria that can be combined and customized in keeping with the person desires. As part of destiny paintings, to implement cryptographic techniques and various were filtering strategies to comfy OSN domestic web page. And also enlarge the paintings in confinement primarily based uploaded video content sharing web sites. The experimental final results showed a bigger universal performance in unique time software.

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**Fig 1: Social Networks**

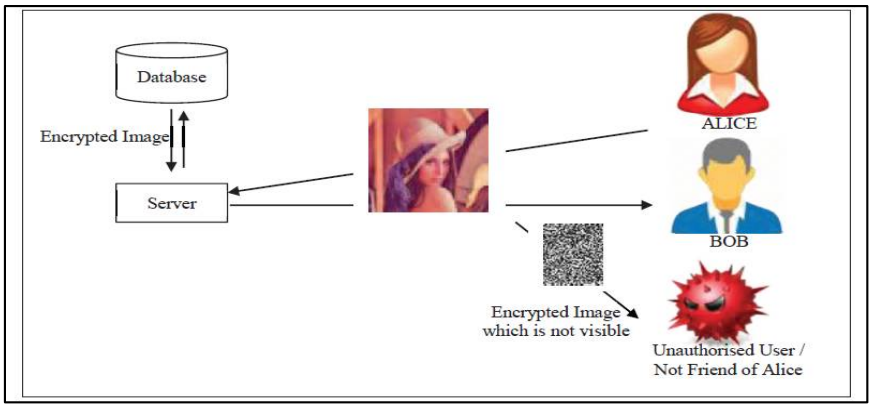


**Fig 2. RDH framework**

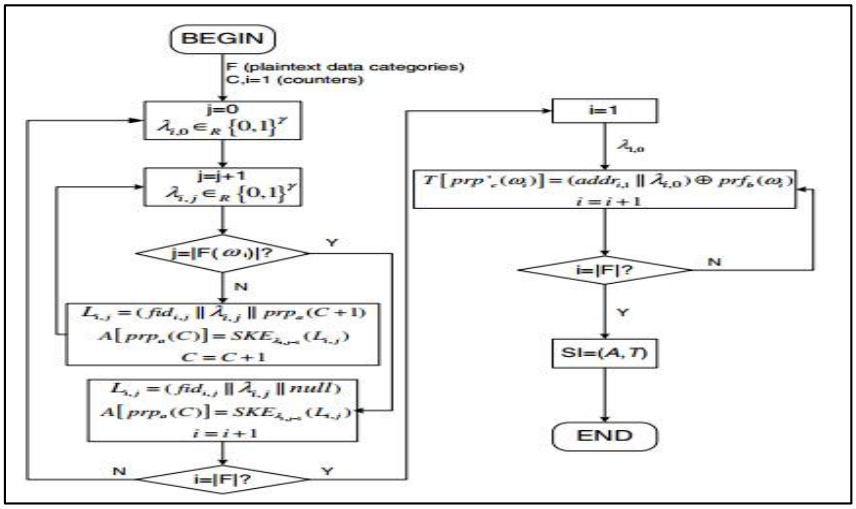


**Fig 3: Broadcast encryptio**

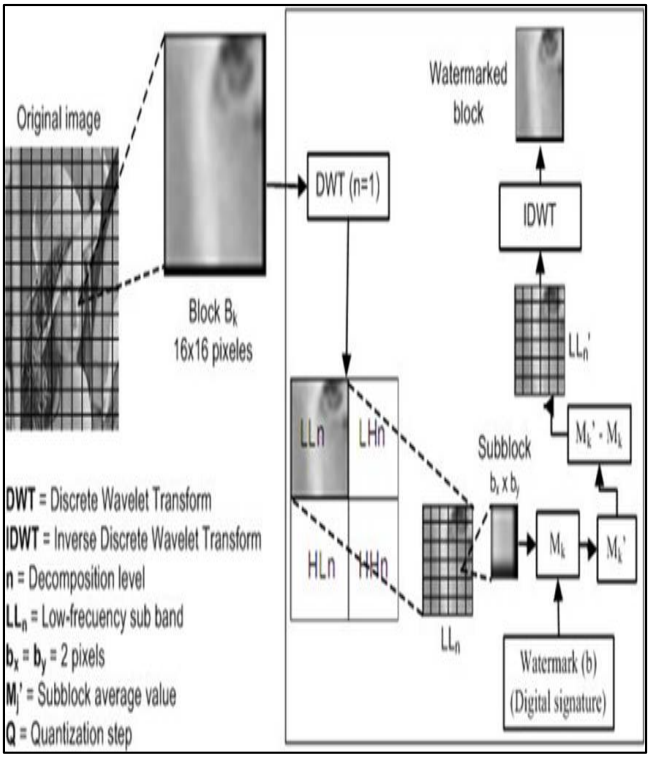


**Fig 4: Discrete wavelet steps**

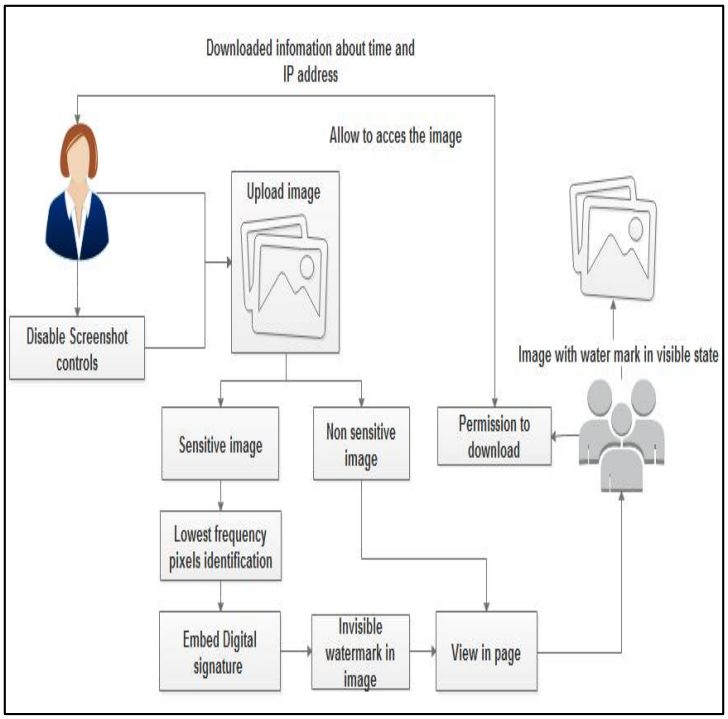


**Fig 5: Proposed Framework**

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RESEARCH ARTICLE

# Privacy Conservation and Integrity Affirmation in Cloud Computing

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## ABSTRACT

To secure the data from unauthorized users such as attackers, hackers and to ensure privacy, integrity, confidentiality and availability of data, we can go with DNA cryptography. It is one of the emerging techniques in cryptographical system. By using this technique the data is being encrypted and decrypted during the data transmission. Encryption and decryption of data is done by using same keys. In this project, a new algorithm is proposed using DNA structure for the purpose of security in cloud environment. DNA cryptography is a technique to encrypt and decrypt the data using DNA sequence.Encryption and decryption of data is done by using same keys. In cryptography, a key is a snippet of data that decides the utilitarian yield of a cryptographic calculation. The key is used to convert the plaintext to cipher text in an encryption and to convert the cipher text to plain text in decryption algorithm.

**Keywords:** Cryptography; DNA; Third Party Auditing;

## INTRODUCTION

Distributed computing comprises of 3 fundamental conveyance models wherever frameworks offer Software as a Service (SaaS), Platform as a Service (PaaS), and Infrastructure as a Service (IaaS). In SaaS, the administration provider gives the hard-ware framework, the item and associates with the client through a front-end entry. In PaaS, the administration provider gives an products development devices for the host to build up the apparatus. In IaaS, the administration provider gives virtual serversunmistakable informatics locations and blocks of capacity on request. Frameworks act over the cloud should think about security and uprightness issues wherever the information ought to be non-open previously (i.e., creation), amid (i.e., transport), and when (i.e., capacity) transmission over the cloud. A client needs to put stock in remote frameworks in taking care of his/her delicate information. there's no assurance that a cloud provider can keep the information non-open and protect its honesty unless explicitly gave.

Uprightness, regarding data and system security, is that the affirmation that information will exclusively be gotten to or changed by those authorized to attempt and do in this way. Measures taken to affirm honesty typify predominant the physical setting of arranged terminals and servers, constraining access to information, and keeping up thorough confirmation hones. learning uprightness likewise can be powerless by ecological dangers, similar to warmth, clean, and electrical surges. Practices took after to protect learning trustworthiness inside the physical setting include: making servers available exclusively to arrange executives, keeping transmission media lined and monitored to affirm that they cannot be suggested.

## Existing System

In the existing framework, it characterizes the strategies for enhancing the effectiveness and security of compacted DNA arrangement correlation calculations, under different questioning situations. For example, one situation includes a querier, Bob, who needs to test if his DNA string, Q, is near a DNA string, Y, possessed by an information proprietor, Alice, however Bob does not have any desire to uncover Q to Alice and Alice will uncover Y to Bob \emph{only if} it is near Q. It depicts a protection upgraded strategy for looking at two packed DNA groupings, which can be utilized to accomplish the objectives of such a situation. This strategy includes a minimization to set differencing, and depicting a security upgraded convention for set differencing that produces supreme protection for Bob (in the data theoretic sense), and a quantifiable level of security insurance for Alice. One of the imperative highlights of this convention, which makes them in a perfect world suited to protection upgraded DNA arrangement examination issues, is that the correspondence intricacy of arrangements is relative to a limit that limits the cardinality of the set contrasts that are of intrigue, as opposed to the cardinality of the sets included (which connects to the length of the DNA groupings). Also, in the conventions, the querier, Bob, can undoubtedly process the set distinction just if its cardinality is near or underneath a predetermined edge.

## Proposed System

A secure cloud storage framework is intended with the options of each privacy and integrity. By the framework the storage security is accrued. the method starts with the privacy preservation. a brand new cryptography rule supported the ideas of desoxyribonucleic acid organic process comes. during this rule the key worth used each sender and receiver is public. The rule is especially accustomed inscribe and decode each string and document in any format like .txt, .doc, .docx. In beginning key generation is performed. when key generation the coding method can manufacture the cipher content. Then mix each the cipher content and key by mistreatment XOR operation to come up with ending. The reverse of the coding method is that the coding method. consequent method is integrity verification. In beginning get the computer file from the user. Generate the hash worth for the computer file. Then it displays the scale of every file. In second step the file is spitted into ten chunks supported the file computer memory unit size. After that, notice the hash worth for the every file mistreatment SHA-1 rule and construct Merkle Hash Tree mistreatment chunks. Then it'll show the basis node and leaf node. At the time of verification the node hash values square measure verified. If the result's true then it show the verification is dead with success. Otherwise every chunk are checked with the hash values.

In particular, to accomplish temperate learning elements, the overall evidence of capacity models by controlling the exemplary Merkle Hash Tree development for square label verification. To help sparing treatment of various evaluating errands, it extra investigates the method of straight blend mark to expand fundamental outcome into a multiuser setting, wherever TPA will play out different inspecting assignments in the meantime. Inside and out security and execution examination demonstrate that the anticipated plans square measure to a great degree sparing and obviously secure.

**Module Explanation**

**Confidentiality- DNA cryptography algorithm:**

**Key generation:**

Step-1:Enter a name then find its length.
Step-2:
A)If length = 16 take the string and proceed to next step.
B)If length > 16 take 16 character from right to left of the word and proceed to next step.
c) If length <16,
i)If the length >10 find the some of digits and insert in first and last positions
ii)If the length < 10 find the some of the length by add the length again and again and find the single digit of the sum. Then, insert value in first and last position.
Step-3: Split the string based on string length %3.
Step-4: Reverse the substrings.
Step-5: Convert it into ASCII and ASCII to binary.
Step-6:Find the value of length % sum of length of digits.
i)If the value is < 2 then, take the characters in odd and even positions and merge them.
ii)If it is > 2 then, take the character at the position of multiplies of that value and merge them.

**Encryption**
**:**
Step-1: Get the text file from the user.
Step-2: Read the content of the file and convert into ASCII and ASCII to binary.
Step-3: Take 1's complement of binary value and reverse it.
Step-4: Save the result.
Step-5: Perform XOR operation of the step 4 result and key generation of the result.
Step-6: Apply the following conditions,
**00-A,10-G,01-C,11-T**
Step-7: Transcription:
**A->T,G->C,T->A,C->G.**
Step-8: Translation:
**A->U**
Step-9: Write the above result in another file.

**Decryption:**

Step-1: Take the input as output of the encryption.
Step-2: Read the file.
Step-3: **U->A.**
Step-4: **T->A,C->G,A->T,G->C**.
Step-5:Apply the following conditions,
**A-00,G-10-G,C-01,T-11**
Step-6: Reverse and take 1's complement.
Step-7: Convert into ASCII and ASCII to character.
Step-8: Write the result into file.

**Integrity Verification Algorithm**

Step-1: Get the input file from the user.
Example- file name: zyx.txt (78kb)
Then find the hash value for that particular file.
Step-2: Divide the input file in equal parts or user choice.
Example- zyx.txt split into equal parts
Step-3: Calculate the hash value for the spitted file and apply the SHA-1 algorithm to calculate the hash value for each spitted file.
Step-4: Each files having separate hash value. Those values must store in one excel file.
Step-5:78KB file Hash values are spitted by using the merkle hash tree to display the hash value for every node.
Step-6: To display the root node and the leaf node.
Step-7: Finally it displays the original file hash.
Step-8: Compare the hash value which is stored in the excel file with merkle tree hash value.
Step-9: Finally it displays the output file.

## CONCLUSION

In the existing system, there are some problems in privacy preservation and integrity verification. Those problems lead to major security issues in cloud data storage. To overcome the issues, a new Secure Cloud Storage architecture is defined. In the architecture each and every steps are well defined and secured with emerging algorithms. The results shown that the architecture is secure and it will give an effective security features while implementing in an organization.

### FUTURE WORK

In future the secure cloud storage architecture will enhanced to secure multimedia files like image, audio, video and graphical format files.

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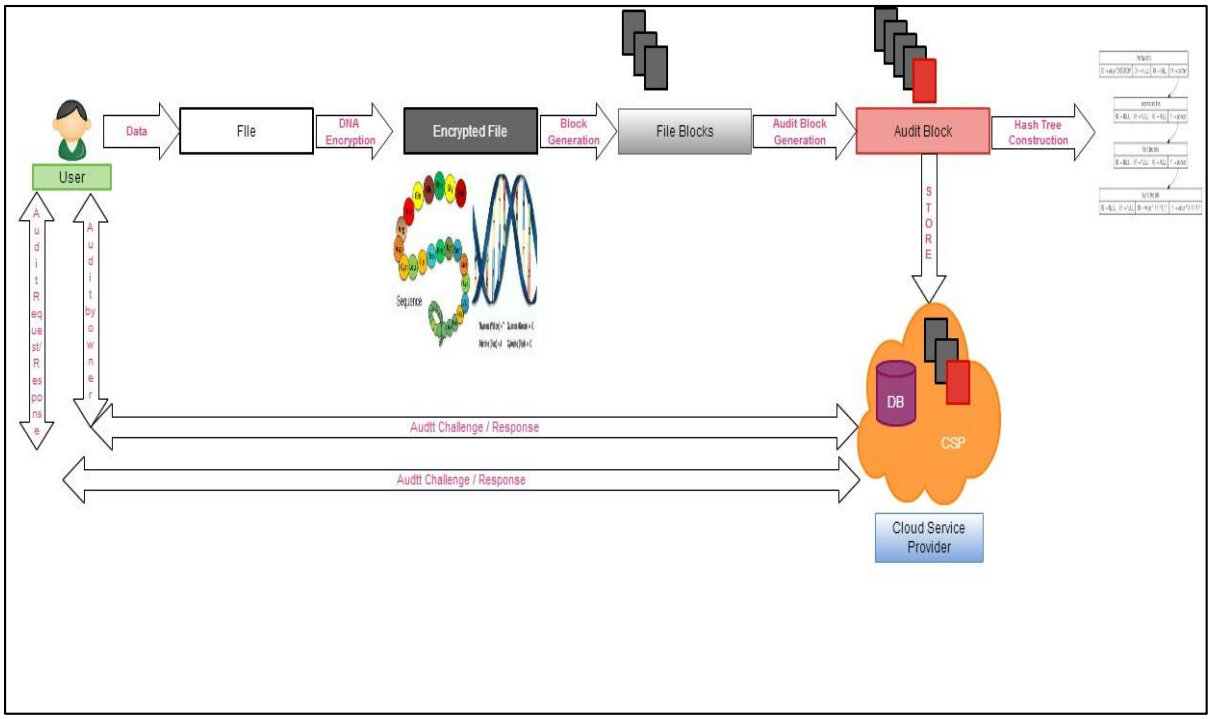


**Fig.1.System Architecture**

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RESEARCH ARTICLE

# Smart Home Big Data: Human Activity and Behavior Based Prediction Model for Medical Applications

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## ABSTRACT

These days, there is a consistently expanding movement of individuals to urban regions and because of this present individuals' wellbeing gets influenced by action changes. In such change, a huge number of homes are being furnished with smart gadgets which produce enormous volumes of fine-grained and indexical information that can be broke down to help smart city administrations. In this paper, propose a model that uses smart home huge information as methods for learning and finding human activity designs for healthcare applications. We propose the utilization of successive pattern mining, cluster analysis and forecast to quantify and analyze vitality use changes started by inhabitants' conduct. Since individuals' routines are generally distinguished by ordinary schedules, finding these schedules enables us to perceive a typical exercises that may demonstrate individuals' challenges in taking look after themselves, for example, not planning food preparation or not utilizing shower, to address the need to inspect fleeting vitality utilization designs at the machine level, which is specifically identified with human exercises. The information from smart meters are recursively mined and the outcomes are kept up crosswise over progressive mining works out. The aftereffects of distinguishing human action designs from machine use are introduced in points of interest in this paper alongside precision of short and long period expectations

**Keywords:** healthcare applications, successive pattern mining, cluster analysis

## INTRODUCTION

The request for medicinal services assets will be extraordinarily influenced by this immense inundation of individuals to downtown areas. This phenomenal statistic change places huge weight on urban areas to reexamine the conventional methodologies of giving well-being administrations to inhabitants [1]. In reacting to the new needs and difficulties, urban communities are at present grasping monstrous computerized change with an end goal to

help practical urban groups, furthermore, give more advantageous condition [2]. In such change, many homes are being furnished with smart gadgets (e.g. smart meters, sensors and so on.) which create monstrous volumes of fine-grained and indexical information that can be investigated to help healthcare applications [3]. Since each person habits are for the most part distinguished by ordinary schedules, finding these schedules enables us to perceive peculiar exercises that may demonstrate individuals' troubles in taking care to themselves, for example, not getting ready sustenance or not utilizing bath [4]. The fundamental relationship between's machine utilization inside the smart home and routine exercises can be utilized by human services applications to identify potential medical issues [5]. This paperproposes the utilization of vitality information from shrewd meters introduced at homes to reveal vital exercises of tenants and watches and breaks down readings from smart meters to perceive exercises and changes in conduct. Disaggregated control utilization readings are specifically related to the exercises performed at home. For ex: If the person is watching television and meanwhile washing machine is in ON mode, then we will be able to analyze that person is simultaneously using more than one gadgets and can easily predict the pattern and the duration of the used gadgets. The model which uses FP-development for design acknowledgment and k-means clustering calculations is equipped for distinguishing apparatus to-machine and apparatus to-time relationship through incremental mining of energy consumption information. Bayesian system for action forecast in light of individual and different apparatus use. This is noteworthy for wellbeing applications that join updates for patients to play out specific exercises in light of chronicled information. For included exactness of the framework, the expectation display coordinates probabilities of machine to-apparatus and apparatus to time affiliations, in this way perceiving exercises that happen .

## Releated Work

Recently, there has been a developing enthusiasm for utilizing keen home innovations for identifying human movement designs for good/ comfort checking applications. The fundamental objective is to take in tenants' behavioural qualities as an approach to comprehend and anticipate their exercises that could show medical problems. In this area, we audit existing work in the writing, which utilize brilliant homes information to investigate clients' conduct. Identifying human exercises in savvy homes by implies of investigating savvy meters information is considered in [10]. In the paper, Detecting Household Activity Patterns from Smart Meter Data [4] a calculation is proposed for distinguishing residential exercises from the total information gathered by the savvy meter . There are two kinds of exercises: Type I exercises are those that can be distinguished by the savvy meter information and Type II exercises can be distinguished from the savvy meter information and ecological detecting (temperature and dampness). Smart meter profiling for wellbeing applications [5] in which the brilliant meters is utilized to screen power use and examination for savvy meters to distinguish and anticipate behavioral anomaly for remote perceive sudden changes in the conduct of people inside brilliant homes. Utilizing information wellbeing observing is talked about in [17] and [18]. The creators in [17] utilize regular apparatuses use from shrewd meter and keen attachment information to follow general exercises and learn one of a kind time section gatherings of apparatus' vitality utilization. Author in [19], utilizes Bayesian systems to anticipate tenant conduct from gathered acute meters information. The investigation proposes conduct as an administration in light of a solitary machine, yet does not give a model to be connected for true situations. The authors in [22] recommend a grouping approach to recognize the circulation of customers' transient utilization designs, be that as it may, the investigation does not consider apparatus level use points of interest. This won't not be material for human action acknowledgment since particular exercises require individual and various apparatus to machine and time affiliation.

## Architecture

Figure speaks to the proposed demonstrate. It begins by cleaning also, setting up the information and afterward applying regular design digging for finding apparatus to-machine affiliations, i.e., figuring out which apparatuses are working together. At that point, it utilizes bunch investigation to decide appliance-to- time affiliations. With these two procedures, the framework is ready to separate the example of apparatus utilization which is at that point

utilized as contribution to the Bayesian system for here and now and long haul exercises forecast.

The yield of the framework is used by particular healthcare applications relying upon the expected utilize. For instance, a healthcare provider may just inspired by knowing exercises identified with intellectual weakness where following the grouping of day by day exercises is essential for reminding the patient when irregular conduct is recognized.

### Approach/implementation

### Cleaning/Pre-processing Phase

In the primary phase of the cleaning procedure we created modified systems to expel noises from the information and set it up for mining. In the wake of cleaning and planning, the dataset is decreased.

### Human activity extraction phase

The point is to find human movement designs from smart meters . For instance, exercises for example, "Sitting in front of the TV, Cooking, Using Computer Preparing Sustenance and Cleaning Dishes or Clothes" are typically standard schedules. In seeking after such process, all machines that are enrolled dynamic amid the 30-minute time interim are incorporated into the source database for visit design information mining.

### Clustering Analysis Phase

A cluster analysis instrument is utilized to find apparatus use time regarding hour of day (00:00 - 23:59), time of day (Morning, Afternoon, Night, Night), weekday, week and additionally month of the year. Appliance to-time affiliations are hidden data in the savvy meter time arrangement information which incorporate adequately close time-stamps, when pertinent apparatus has been recorded as dynamic or operational. Utilizing this data we can bunch a class or group of machines that are in operation at the same time or covering.

### Activity Prediction Phase

We incorporate the regular examples and machine to-time relationship to find out about the utilization of various machines and construct the action expectation show. The mechanism comprises Bayesian network which is a directed acyclic graph, where hubs speak to irregular factors what's more, edges show probabilistic conditions.

## CONCLUSION

Proposing Prediction Model for perceiving human exercises designs from low determination savvy meters information. Inhabitants' propensities and conduct take after an example that could be utilized as a part of wellbeing applications to track the prosperity of people living alone or those with self-restricting conditions. The appropriateness of the proposed model to accurately recognize various machine use and make short and long haul expectation at high precision. The human propensities and conduct take after an example that could be utilized as a part of wellbeing applications to track the wellbeing issues of people living alone or those with self-restricting conditions. These human exercises can be construed from machine to-apparatus and apparatus to-time affiliations. The materialness of the proposed display was to effectively distinguish numerous machine uses and make short and long haul forecast. The yield of the framework is used by particular human services applications relying upon the planned utilize.

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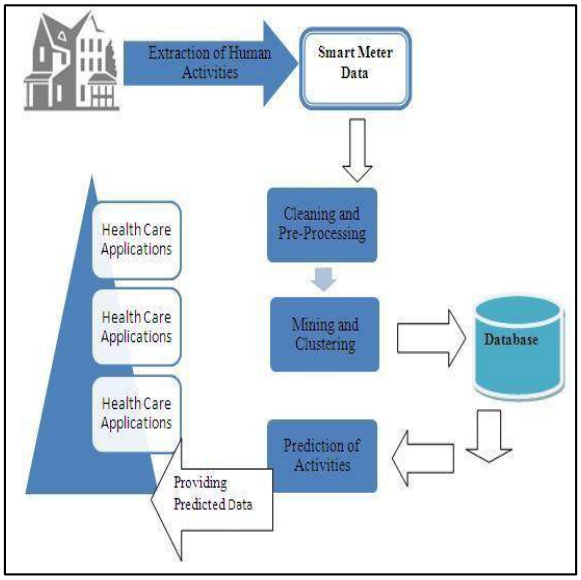


**Fig.1.Architecture of the proposed system.**

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RESEARCH ARTICLE

# Social Voting Recommendation Using Hierarchical Agglomerative Clustering

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## ABSTRACT

The most recent decade has seen a huge development of online administrations as an outstanding modernization for distributing information, figuring resources, and projects over the Web. With expanding appropriation and nearness of Online administrations, planning novel methodologies for productive and compelling suggestion has happened to vital significance. In existing administrations disclosure and proposal approaches concentrate on watchword predominant Web benefit web crawlers, which have numerous confinements, for example, poor suggestion execution and overwhelming reliance on right and complex inquiries from clients. Late research endeavors on online administration suggestion focus on two conspicuous methodologies: communitarian sifting and substance-based proposal. Lamentably, both methodologies have a few disadvantages, which limit their materialness in Web benefit suggestion. In proposed framework for suggestion we will utilize Agglomerative Hierarchal Clustering or Hierarchal Agglomerative Clustering for powerful proposal in this system. Our approach considers at the same time both rating information and linguistic substance information utilizing a probabilistic generative model.

**Keywords:** online social networks (OSNs), recommender systems (RSs), social voting, preprocessing

## INTRODUCTION

Vast information has developed as a widely perceived model, drawing in examination from government, industries and the erudite world. As a rule, Big Data deals with enormous amount of content with tremendous efficiency to solve the data set related problems. Enormous applications were developed to utilize the process inside a "mediocre slipped by time" is on the ascent. The basic check for Big Data applications is to concentrate on large data to convert it into valuable data. In attitude of this test, a Clustering-primarily based Collaborative Filtering technique is proposed on this paper, which goes for enrolling comparative administrations over similar groups to recommend advantages cooperatively.

Group based proposal is best idea of as a variation on client based suggestion. Rather than prescribing things to clients, things are prescribed to bunches of comparative clients. This involves a pre-preparing stage, in which all clients are divided into bunches. Suggestions are then delivered for each bunch, to such an extent that the prescribed things are most intriguing to the biggest number of clients. The upside of this approach is that proposal is quick at runtime in light of the fact that practically everything is pre registered.

The most recent decade has seen a gigantic development of Web benefits as an outstanding newness for sharing statistics, figuring property, and initiatives at the internet. With expanding reception and nearness of the Web administrations, outlining novel methodologies for productive and successful Web benefit proposal has happened to principal significance. In existing web administrations revelation and proposal approaches concentrate on watchword overwhelming Web benefit web indexes, which have numerous impediments, for example, poor suggestion execution and substantial reliance on right and complex questions from clients. Late research endeavours on Web benefit suggestion fixate on two conspicuous methodologies: collective separating and substance based proposal. Lamentably, both methodologies have a few downsides, which confine their relevance in Web benefit suggestion. In proposed framework for suggestion we will utilize Agglomerative Hierarchal Clustering or Hierarchal Agglomerative Clustering for viable proposal in web-administrations. Our approach considers all the while both rating information linguistic substance information of Web administrations utilizing a probabilistic generative version.

## Related Works

Gediminas Adomavicius, Alexander Tuzhilin in 2005. The business and the scholarly community on growing new ways to deal with recommender frameworks in the course of the most recent decade. The enthusiasm for this range still stays high since it constitutes an issue rich research zone and due to the wealth of functional applications that assistance clients to manage data over-burdens and give customized proposals, substance, and administrations to them. Cases of those applications incorporate prescribing books, CDs, and different items at Amazon films. Besides, a portion of the merchants have consolidated suggestion abilities into their business servers. These techniques join community oriented and content-based strategies. Great measurements to gauge the viability of suggestions has been widely tended to in the recommender frameworks the execution assessment of proposal calculations is generally done as far as scope and exactness measurements another client, having not very many appraisals, would not have the capacity to get precise proposals. Proposal process, supporting multicriteria appraisals, and giving more adaptable and less nosy sorts of suggestions.

[2] Memory based community separating has been examined widely in the writing and has ended up being fruitful in different sorts of customized recommender frameworks. It is utilized as a part of recommender frameworks. CBF frameworks examine the substance of an arrangement of things together with the evaluations gave by singular clients to deduce which nonrated things may be of enthusiasm for a particular client through acquisition of [24] relevant data partitioning. Synergistic separating does not depend on the substance portrayals of things, but rather simply relies upon inclinations communicated by an arrangement of users.CF gives an intense approach to conquer these challenges. The data on individual inclinations, tastes, and quality are altogether conveyed in (express or certain) client appraisals. Utilize PMCF to determine answers for two especially critical issues in CF, memory and model-based CF probabilistically quick reactions of model-based CF. Built profile space does to be sure permit both a precise and quick expectation of client appraisals.

[3] Recommender frameworks have turned out to be progressively vital instruments to help clients effectively deal with an expansive type of provided things and administrations and concentrate on generally intrigue. Extensively, recommender. Frameworks are ordinarily in light of one among two option methodologies. The substance-based approach makes portraits that catch the trademark highlights of clients and things, and utilizations these highlights that distinguish likely partiality amid a client and a thing. The fundamental trouble with this approach is that it is

regularly difficult and now and then difficult to gather the outer data expected to make the profiles. Another procedure that stays away from either extraordinary by methods for express advancement. We treat the related client thing pair as an improvement variable. The idle elements and these distinct mark factors are determined at the same time. Transductive SVMs actualize the low-thickness detachment. ZAN performs generously superior to anything ZAM and Popularity oriented plans. The execution of ZAN has a tendency to enhance with client arranged weighting yet winds up noticeably practically identical with thing focused and uniform weightings. It has enhancement way to deal with one-class lattice finishing and synergistic sifting issues.

[4] Recommender frameworks can experience the ill effects of information sparsity and cool begin issues. Be that as it may, informal organizations, which empower clients to manufacture connections and make distinctive sorts of things, exhibit an extraordinary chance to lighten these issues. a star-organized half breed chart fixated on a social space, which associates with other thing areas. The colossal volume of things produces an issue of data over-burden. Conventional web post proposal approaches experience the ill effects of information scantily. One regular kind of way to deal with suggestions, known as synergistic separating procedures, portrays clients' idle highlights autonomously with client thing communications in a solitary thing area. The clients read web posts made by their group and may embrace comparative client marks to their companions. an extensive genuine interpersonal organization dataset and demonstrated that the proposed technique significantly supports the social suggestion execution. 27.6% of the accessible data in the objective area, our strategy accomplishes tantamount execution with techniques that utilization all accessible data in the objective space without exchange learning. HRW strategy on a star-organized diagram, which is a general technique to consolidate mind boggling and heterogeneous connection structures. It takes care of the information sparsity issue. Exchange learning point of view and reduced the information sparsity issue. Transferable names, performs much superior to HRW.

Yehuda Korean in 2006**.** Recommender frameworks give clients customized recommendations for items or administrations. These frameworks frequently depend on Collaborating Filtering (CF), where past exchanges are examined to build up associations amongst clients and items. The techniques are tried on the Netflix information. Results are superior to those already distributed on that dataset. Electronic retailers and substance suppliers offer a colossal choice of items, with uncommon chances to meet an assortment of extraordinary needs and tastes. CF methods require no area learning and dodge the requirement for broad information gathering. Netflix discharged a dataset consist of 100 million motion picture appraisals and tested the examination group to create calculations that could beat the exactness of its suggestion framework, Cinematic. Models are lack at distinguishing solid relationship among a little arrangement of firmly related things, correctly where neighborhood models do best. It is simple mix with different strategies that depend on comparatively organized worldwide cost capacities. It generally viewed as having a place with neighborhood models, in particular, a capacity to disclose suggestions and to deal with new clients flawlessly. SVD does not parameterize client we enhanced suggestion quality relies upon effectively tending to various parts of the information

## Proposed System

A key piece of a recommender framework is a synergistic separating calculation foreseeing clients' inclination for things. To depict diverse effective community oriented separating procedures and a system for consolidating them to acquire great forecast. Recommender frameworks are vital for internet business. In the event that an organization offers numerous items to numerous customers, it can profit considerably from displaying customized suggestions. It numerous business utilizations of recommender frameworks for items like books, motion pictures, music and others. Likewise, numerous applications not specifically business have risen: customized suggestions for sites, add diverse effective indicators to the gathering. Great competitors are techniques effectively connected with progress to communitarian. The objective of the challenge is to create a decent forecast of clients' inclinations for films. System for joining expectations and portrayed techniques that consolidated together give a decent forecast for the Netflix

Prize dataset. A troublesome piece of building a recommender framework, knows inclinations of clients for a few things, to precisely anticipate which different things they will like.

Community oriented separating is a compelling proposal approach in which the inclination of a client on a thing is anticipated in light of the inclinations of different clients with comparative interests. A major test in utilizing communitarian separating techniques is the information sparsity issue which regularly emerges in light of the fact that every client ordinarily just rates not very many things and subsequently the rating framework is to a great degree meager. The MCF issue is especially appropriate for huge scale web based business and interpersonal interaction administrations which frequently give a differing scope of items or administrations. CF methods abuse past exercises of the clients, for example, their exchange history or item fulfillment communicated in appraisals, to anticipate the future exercises of the clients. CF-based proposal frameworks have turned out to be progressively prevalent in light of the fact that it is for the most part significantly less demanding to gather the past exercises of clients than their profiles, halfway because of protection contemplations. The MCF issue is especially reasonable for vast scale online business and informal communication administrations which frequently give a differing scope of items or administrations. The adequacy of abusing the connections between various areas. A multi-space CF (MCF) issue which mutually models an accumulation of rating expectation undertakings emerging from different areas.

[8] Clients regularly rate just a little portion of every single accessible thing. The nonattendance of evaluations conveys helpful data for enhancing the best k hit rate concerning all things, under mellow presumptions, without predisposition from information notwithstanding when appraisals are leaving not indiscriminately. The possibility of recommender frameworks is to consequently propose things to every client that s/he may get engaging. The nature of recommender frameworks can be surveyed regarding different criteria, including exactness, assorted variety, amaze/good fortune, and clarify capacity of proposals. Enhancing suggestion exactness regarding all things when the accessible preparing and test information are MNAR. Include the advancement of more refined surrogate target capacities for preparing, and all the more effective models.

## Working Model

Community filtering and recommender frameworks are basically worried about two related issues: rating prediction and positioning. The objective of the rating forecast errand is to precisely foresee the rating a client would relegate to an individual thing. Collaborative filtering strategies endeavor to tackle the two issues by utilizing rating information gathered from an extensive group of clients. It basically expresses that the likelihood that a rating is lagging does not rely upon the estimation of that rating, or the estimation of some other lagging rating. The elect of non-arbitrary missing information by testing two extra benchmark techniques: closest neighbor relapse and framework While Proposal precision can be enhanced by joining trust connections got from informal organizations. Latest work on informal community construct suggestion is engaged in light of limiting the root mean square mistake (RMSE). Informal community based best k proposal, that prescribes to a client few things at any given moment. To enhancing the exactness of best k proposal utilizing informal communities. There is some change on the normal suggestion exactness over all clients, the change is especially huge for the purported cold users, to survey the execution of the proposed interpersonal organization based best k. Enhancing the exactness of best k proposal utilizing trust data got from social networks. To utilize voting-based calculation as a straightforward way to deal with consider both watched and missing ratings. It expands the calculation multifaceted nature, which is different key execution metric.supply evaluations for things that they do like. It more averse to supply evaluations for things that they don't care for. While missing information isn't absent aimlessly, this essential property neglects to hold, and it isn't conceivable to disentangle the probability be-yond. The motion pictures are [25] classified and the [26] objects from the images are clustered for the better recommendations. utilizing straightforward. It is effortlessly disregarded in recommender frameworks, for instance, clients will probably supply evaluations for things that they do like. It more averse to supply evaluations for things that they don't care for. While missing information isn't absent aimlessly, this essential property neglects to hold, and it isn't conceivable to disentangle the probability be-yond.

Missing information isn't absent aimlessly, this critical property neglects to hold, and it isn't conceivable to improve. Intrigued by is would we be able to get better appraising forecast and positioning execution on rating expectation Social proposal is pervasive in genuine, yet top k suggestion utilizing on the web informal organizations has been inadequately considered in the proposal writing.

### Algorithms/Techniques

Agglomerative various leveled bunching is a base up grouping strategy where bunches have sub-groups, which thusly have sub-groups, and so on. The great case of this is species scientific categorization. Quality articulation information may likewise show this various leveled quality (e.g. neurotransmitter quality families). Agglomerative various leveled bunching begins with each and every protest (quality or test) in a solitary group. Then, in each progressive emphasis, it agglomerates (consolidates) the nearest combine of bunches by fulfilling some likeness criteria, until the point when the greater part of the information is in one group.

Properties of the hierarchy:

- Clusters generated in early stages are nested in those generated in later stages.
- Clusters with different sizes in the tree can be valuable for discovery.

## CONCLUSION

We display a Club CF approach for huge information applications significant to benefit proposal. Before applying CF system, administrations are converged into a few groups by means of an AHC calculation. At that point the rating likenesses between administrations inside a similar group are registered. As the quantity of administrations in a bunch is substantially less, Club Collaborative Filtering needs less calculation time. In addition, as the appraisals of administrations in a similar group were more applicable with in different bunches. On the off chance that we need, include the new motion pictures with name, classification and year

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RESEARCH ARTICLE

# The Social Network of AI Chatbots

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## ABSTRACT

The proposed system helps to develop and implement an online social network platform of artificial chat bots. The human user can only interact or chat with the other chat bot within the network. There is no option to chat with the other human. These chat bots are created by individual users who signed up in the platform. However, the bots itself could be created at any other existing platform supported by Machine Learning. They are integrated into this Social Network by appropriate embed code.The bots may work for a specific purpose or as a conversational bot interacting with what had it learned from its administrator. The admin could teach the chat bot in his own way through Machine Learning options supported by the chosen platform.The user could transfer the skills from one chat bot to another without making efforts to impart the same skill again. The education portal is made to support transaction of skills more easily between bots within the network. The terms and conditions to deal with a bot include that they must not be treated with abusive languages since they are in the development stage.

**Keywords:** artificial chat bots, Social Network, Machine Learning

## DOMAIN

**Machine Learning**

The artificial chat bots are taught by the administrator who create them and they tend to learn from their own experiences through conversation in the platform. The bots are further expanded to accept queries from the other user and manipulate replies based on the data from the databases. “A computer program is said to learn from experience E with respect to some class of tasks T and performance measure P, if its performance at tasks in T, as measured by P, improves with experience E”

The two major types of Machine Learning are,

- Supervised learning
- Unsupervised learning

**Supervised Learning**

Several pre-classified data inputs are provided to the system for the machine to learn its behaviour. With the learned data, it could classify any new input as expected.

## CLASSIFICATION

"The output variable takes class labels"
The category problem deals with discrete goals with finite set of feasible values. inside the most not unusual kind of type issues, the goal variable will have simplest two feasible values, which includes actual/fake, applicable/not applicable, replica/now not duplicate, Cat/canine and so forth. every so often the target variable could have greater than two outcomes.

**Major examples of Classification:**

- Identifying Cancer based on the size and type of Tumour
- Identifying the type of fruit based on colour, size and shape

**Regression:**

"The output variable takes continuous values" Plotting a variety of inputs in a co-ordinate system results in a curve. To predict the output for a given new set of inputs, the lines must arise from both the co-ordinates.

**Major examples of Regression:**

- Predicting the price of land/home with some pre-defined values
- Predicting the weather forecast measures for a specific month/year

**Major fields of Application:**

- Handwriting recognition
- Learning to rank
- Object recognition in computer vision
- Optical character recognition
- Spam detection
- Pattern recognition
- Speech recognition
- Bioinformatics

**Unsupervised Learning:**

No pre-classified data are passed to the system. The system accepts a bunch of raw data and finds a hidden pattern within them. Through the identified pattern, it divides the whole data into several classes.

The two major types of unsupervised learning are,

- Clustering
- Association

### Clustering

A clustering trouble is wherein you want to discover the inherent groupings in the records. Those organization of data are continually much like each different belonging to a selected magnificence.

### Major examples of Clustering

- Groups news stories into cohesive groups (Google News)
- Classifying human genomes with microscopic data

### Association

An affiliation rule learning problem is wherein you need to find out guidelines that describe massive portions of your data.

### Major examples of Association

- Market Basket analysis to find items bought together
- Auto suggestion of items in E-Commerce portals
- Point of Sale Systems
- Web usage mining

### Major Algorithms used

- k-means for clustering problems
- Apriori algorithm
- Hidden Markov models
- Hierarchical clustering

### Natural Language Processing

The bots converse with no need of any syntactical form of sentence construction. NLP is integrated as part of them to support conversation like a human from the other end. Hence, the bots could undergo Turing Test to validate its performance over the course of time to distinguish between a more natural and artificial conversation.

NLP is a way for computer systems to analyze, recognize, and derive meaning from human language in a clever and useful manner. "Apart from common word processor operations that treat text like a mere sequence of symbols, NLP considers the hierarchical structure of language: several words make a phrase, several phrases make a sentence and, ultimately, sentences convey ideas,"

NLP algorithms are generally based on machine mastering algorithms. In preference to hand-coding big units of policies, NLP can rely on machine mastering to automatically learn those regulations by analyzing a hard and fast of examples (i.e. a massive corpus, like a e book, down to a group of sentences), and creating a statical inference.

### General procedure followed in NLP Systems:

- Summarize blocks of text using Summarizer
- Identify the type of entity extracted

### Major examples of Natural Language Processing:

- Track trending topics and popular hashtags
- RSS Reader imparted with Machine Learning
- Understanding Human language in standalone chat bots

**Steps followed in NLP:**

**From Linguistics to Natural Language Processing**
**Linguistics:**

Linguistics is the way to observing of language, along with its grammar, semantics, and phonetics take a look at of laptop structure

**Computational Linguistics**

Computational linguistics is the s for knowledge and generating herbal language. One natural feature for computational linguistics will be the checking out of grammars proposed with the aid of theoretical linguists. For that reason, it is the current take a look at of linguistics using the gear of pc science.

**Statistical Natural Language Processing**

The statistical dominance of the field also regularly leads to NLP being defined as Statistical herbal Language Processing, possibly to distance it from the classical computational linguistics methods.

**Challenges in Natural Language Processing**

*Lexical ambiguity* – it's miles at very primitive stage inclusive of phrase-level.
for instance, treating the phrase "board" as noun or verb?
Syntax level ambiguity – A sentence can be parsed in one-of-a-kind ways.
as an example, "He lifted the beetle with pink cap." – Did he use cap to lift the beetle or he lifted a beetle that had pink cap?
*Referential ambiguity* – regarding something using pronouns.
as an instance, Rima went to Gauri. She stated, "i am tired." – exactly who is tired?

**Simple Algorithms used in NLP**

- Context-Free Grammar
- Top-Down Parser

**Natural Language Processing APIs**

- IBM Watson
- Amazon Lex
- Microsoft Cognitive Services
- Facebook's DeepText

**Front End:**
**ReactJS:**

React is a front-stop library evolved by using fb. it's miles used for dealing with the view layer for web and mobile apps. ReactJS permits us to create reusable UI additives. it is currently one of the maximum famous JavaScript libraries and has a strong foundation and large network in the back of it.



It goals basically to offer velocity, simplicity, and scalability. React procedures best person interfaces in applications. This corresponds to View inside the model-View-Controller (MVC) pattern, and may be utilized in aggregate with other JavaScript libraries or frameworks in MVC, consisting of AngularJS.

**Notable features:**

- One-way data flow
- Virtual DOM
- JSX

**Back End:**

**Django:**

Django is a unfastened and open-supply web framework, written in Python, which follows the model-view-template (MVT) architectural pattern. it's miles maintained via the Django software foundation (DSF), an unbiased agency established as a non-earnings.

Django's primary purpose is to ease the creation of complicated, database-pushed web sites. Django emphasizes reusability and "pluggability" of components, speedy improvement, and the precept of don't repeat yourself. Python is used throughout, even for settings documents and statistics fashions. Django additionally offers an elective administrative create, read, update and delete interface.

**Contrib Package Applications:**

- Tools for generating news and Atom syndication feeds
- Tools for generating Google Sitemaps
- An extentable authentication system
- The adaptable administrative interface
- Default mitigation for cross-web page request forgery, move website scripting, sql injection, password cracking

**Django implemented sites:**

- Instagram
- Mozilla

**Proposed System**

The proposed system helps to develop and implement an online social network platform of artificial chat bots. The human user can only interact or chat with the other chat bot within the network. There is no option to chat with the other human. These chat bots are created by individual users who signed up in the platform. However, the bots itself could be created at any other existing platform supported by Machine Learning. They are integrated into this Social Network by appropriate embed code.

**Advantages of Proposed System**

1. Registering a chat bot for every user and teaching them to learn encourages research over Machine Learning domain
2. Friendly access to advanced technologies with less effort
3. Ability to introduce artificial intelligence into the Social network that evolves on its own

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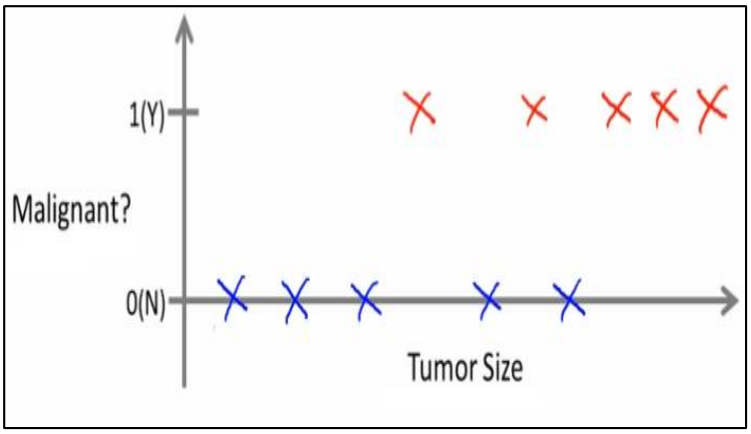


**Fig. 1.Identifying malignant tumor**


**Fig. 2.House price prediction**

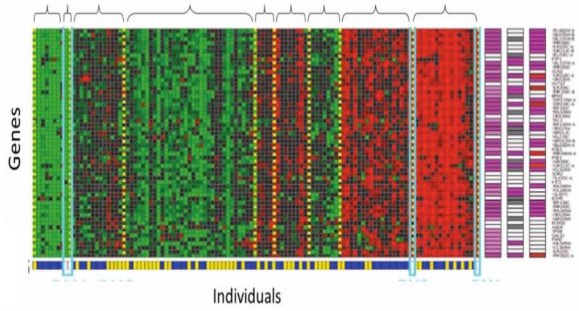


**Fig.3.Clustering of genes**

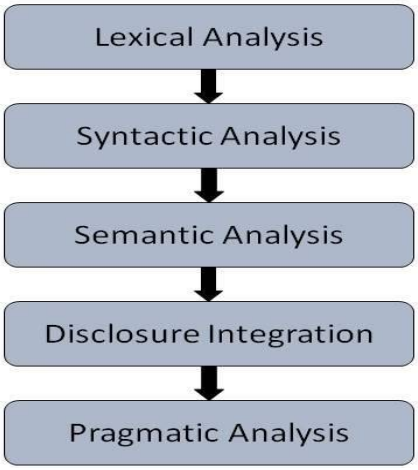


**Fig.4. Steps involved in NLP**

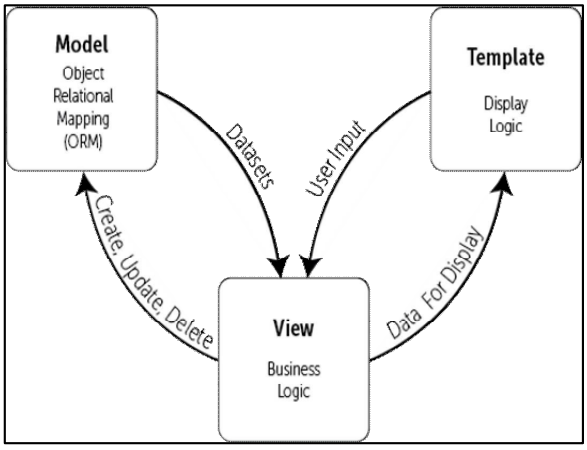


**Fig.5. MVT architecture in Django**

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**RESEARCH ARTICLE**

# MORPHEUS: Generating Structured Music with Constrained Patterns and Tension

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## ABSTRACT

Extraction of discriminative highlights from remarkable facial patches assumes a crucial part in viable outward appearance acknowledgment. The exact discovery of facial historic points enhances the confinement of the striking patches on confront pictures. This paper proposes a novel system for demeanor acknowledgment by utilizing appearance highlights of chose facial patches. A couple of conspicuous facial patches, contingent upon the situation of facial points of interest, are extricated which are dynamic amid feeling elicitation. These dynamic patches are additionally handled to get the notable patches which contain discriminative highlights for arrangement of each combine of articulations, in this way choosing distinctive facial fixes as notable for various matches of demeanor classes. One-against-one order technique is received utilizing these highlights. What's more, a mechanized realizing free facial historic point identification system has been proposed, which accomplishes comparable exhibitions as that of other condition of-craftsmanship milestone recognition strategies, yet requires fundamentally less execution time. The proposed technique is found to perform well reliably in various resolutions, consequently, giving an answer for articulation acknowledgment in low determination pictures. Tests on CK+ and JAFFE outward appearance databases demonstrate the viability of the proposed framework.

**Keywords:** Facial Emotion Detection, Facial Expression Recognition, Audio Feature Recognition, Emotion Based Music player.

## INTRODUCTION

PCs are rapidly turning into a pervasive piece of our lives. We invest a lot of energy collaborating with PCs of one kind or another. Right now the gadgets we utilize are unconcerned with our emotional states. They are candidly visually impaired. Be that as it may, effective human-human correspondence depends on the capacity to peruse full of feeling and passionate signs. Human-PC communication (HCI) which does not consider theaffective conditions of its clients loses a vast piece of the data accessible in the interactionWhen talking or singing inwardly, passionate outward appearances and motions areoften joined with sound-related signs of feeling, making an effective multimodalaffective experience.Music assumes an essential part in upgrading a person's life as it is a vital medium of diversion for music sweethearts and audience members and now and again even imparts a therapeutic approach. Inthe present world, with regularly expanding headways in the field of interactive media and innovation, different music players have been produced with highlights likefast forward, invert, variable playback speed (look for and time compression),local playback, gushing playback with multicast streams. In spite of the fact that these highlights fulfil the client's fundamental prerequisites, yet the client needs to confront the assignment of physically perusing through the playlist of melodies and select tunes in view of his present temperament and conduct. The presentation of Audio Emotion Recognition (AER) and Music Information Retrieval (MIR) in the conventional music players gave naturally parsing the playlist in view of different classes of feelings and mood.

## LITERATURE SURVEY

Different methods and methodologies have been proposed and created to group human passionate state of behaviour. The proposed approaches have concentrated just on the portion of the fundamental feelings. With the end goal of feature recognition, facial highlights have been classified into two noteworthy classifications, for example, Appearance-based feature extraction and geometric based component extraction by zheng. Geometric based element extraction method considered only the shape or major conspicuous purposes of some critical facial highlights, for example, mouth and eyes. There is another scheme that is naturally section an info picture, and to perceive facial feeling utilizing identification of shading based facial highlight guide and arrangement of feeling with straight forward bend and separation measure is proposed and implemented. In other plan there is programmed technique for constant feeling acknowledgment utilizing outward appearance utilizing a new anthropometric display for facial element extraction.

[1] Anagha S. Dhavalikar and Dr. R. K. Kulkarni Proposed Automatic Facial Expression acknowledgment framework. In Thissystem there are three stage 1.Face identification

[2] Feature Extraction and 3.Expression acknowledgment. The First Phase FaceDetection are finished by YCbCrColour demonstrate, lighting pay for getting face and morphological activities forretaining required face i.e. eyes and mouth of the face. This System is additionally utilized AAM i.e. Active Appearance Model Technique for facial element extraction In this strategy the point on the face likeeye, eyebrows and mouth are found andit make an information record which gives data about model focuses identified and distinguish the face the a demean or are givenas input AAM Model changes as indicated by expression.

[3] Yong-Hwan Lee ,Woori Han and Youngseop Kim proposed framework in view of Bezier bend fitting. This framework used twoventure for outward appearance and feeling initial one is location and examination of facial zone from input unique picture Furthermore, next stage is confirmation of facial feeling of qualities include in the area of interest.

[4]ArtoLehtiniemi and Jukka Holm proposed framework in view of enlivened state of mind picture in music suggestion. Onthis framework the client interface with a gathering of pictures to get music proposal as for kind ofpicture. this music proposal framework is produced by Nokia looked into centre. This framework utilizes literary meta tagsfor portraying the class and sound flag preparing.

[5]F. Abdat, C. Maaoui and A. Pruski proposed framework completely programmed outward appearance and acknowledgment framework basedon three stage confront identification, facial characteristics extraction and outward appearance arrangement. This framework proposed anthropometric model to distinguish the face include direct consolidated toward shi and Thomasi strategy. In this strategy the variationof 21 separations which portray the facial element from nonpartisan face and the characterization base on SVM (Support Vector Machine).

## METHDOLOGY

### Facial Landmark Detection

The position of the face is often not centered in the detected sub-image. Since the detection of the eyes and mouth will be done on detected face regions of a predefined sizeproposed a robust, learning-free, light-weight generic face model fitting method for localization ofthe facial organs.

Exploratory outcomes have demonstrated that the time required for sound element extraction is irrelevant (around 0.0006 sec) and melodies are put away prehanded the aggregate estimation time of the proposed framework is corresponding to the time required for extraction of facial highlights (around 0.9994 sec).Also the different classes of feeling yield a superior exactness rate when contrasted with past existing framework The computational time taken is 1.000sec which is less along these lines helping in accomplishing a superior ongoing execution and efficiency.The framework in this way goes for furnishing the Windows working framework clients with a less expensive, extra equipment free and precise feeling based music framework. The Emotion Based Music System will be of awesome favourable position to clients searching for music in light of their state of mind and enthusiastic behaviour. It will help decrease the hunting time down music in this manner diminishing the superfluous computational time and in this way expanding the general precision and productivity of the framework.The framework won't just diminish physical pressure yet will likewise go about as a help for the musical treatment frameworks and may likewise help the music advisor to therapies a patient. Likewise with its extra highlights said above,it will be a total framework for music darlings and audience.

## CONCLUSION

We have introduced an entire and completely robotized approach for outward appearance recognizable proof by all the while using the face surface and face subsurface highlights. We displayed another calculation for the face recognizable proof and acknowledgment, which would more be able to dependably extricate the face, includes and accomplishes considerably Higher exactness than beforehand proposed facial ID approaches. The proposed approach shows a low level of multifaceted nature, which makes it reasonable for ongoing applications, the element focuses or key focuses recognized and mapped alongside the face picture. Contingent on the chose highlights and the deliberate area properties of the human face, the distinctive demeanor of the human was additionally characterized utilizing SVM. The proposed strategy is unrivaled contrasted and other best in class approaches and that the examination of the general picture nature of the face pictures uncovers exceptionally important data that might be proficiently used to segregate them from counterfeit attributes.

**Future Scope for Implementation**

- Facial recognition can be used for authentication purpose.
- Android Development.
- Can detect sleepy mood while driving.
- Can be used to determine mood of physically challenged & mentally challenged people

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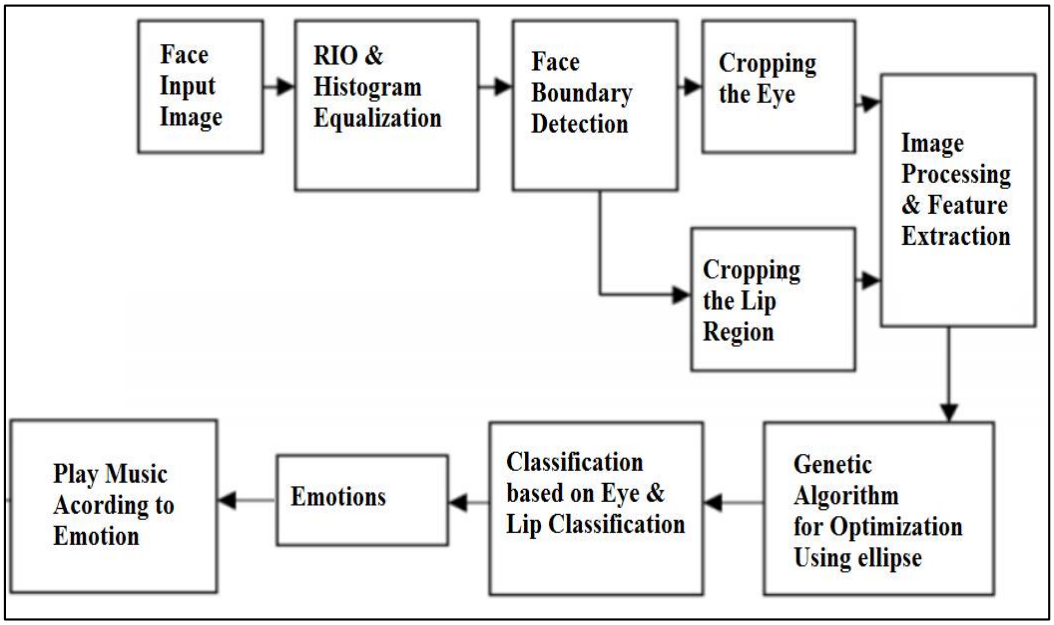


**Fig.1.Architecture**

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RESEARCH ARTICLE

# Ariyalur, Cement City of India its Present Status and Grave Impact of Dust Pollution on Some Important Agricultural Crops

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## ABSTRACT

Our Country forests are transforming into concrete jungles, Ariyalur, Cement city of India its present contribution is significant in this transformation survey is conducted to derail its importance and impacts on agricultural crops. Certain morphological parameters fresh weight dry weight etc is studied in correlation with the dust pollution.

**Keywords:** Ariyalur cement city, Agricultural crop, pollution and pollutant.

## INTRODUCTION

Air, land and water are the most basic amenities for living organisms. Once, these amenities were pure, virgin, undisturbed, uncontaminated and basically most hospitable for living organisms. But nowadays, the environment has become undesirable and contaminated by environmental pollution and therefore harmful for the health of fauna and flora. Pollution is an undesirable change in the physical, chemical and biological characteristics of water, air or land that will be or may be, harmful to human and other life, industrial processes, living condition and cultural assets (Odum, 1975). Human population explosion, rapid industrialization, increased deforestation, unplanned urbanization, scientific and technological advancement etc. are main causes for all kinds of pollution. The first Indian cement factory was established in 1914 at Porbundar in Gujarat. At present, there are 155 major cement industries (160 million tonnes per annum) and 300 mini cement plants (9 million tonnes per annum) in India. There are nearly 20 major cement industries located in Tamil Nadu. The main industries such as Alagappa cements (Pudupalayam), Ariyalur Cement Works (Kallankurichi), Chettinad cements (Keelapazhur), Dalmia cements (Thamaraikulam), Dharani

cements (Veenakaikatti), Grasim cements (Reddiarpalayam), Ramco cements (Govindapuram) and Sankar cements (Thalavai) are located in and around ariyalur. They produce enormous amount of dust and pollute the surrounding environment. Among them, Ariyalur Cement Works, Ariyalur was established in 1979. It produces as much as 1500 MT/day employing dry process method. During production, it releases a stalk emission of 1,60,000 MT/hr. The emitted dust is carried out to surrounding areas and deposited over the vegetation and soil.The accompanying growth of cement industries in India has consequently magnified the pollution problem. Cement industries are regarded to be highly-pollution prone, especially with regard to particulate emission. They also play a vital role in the imbalance of the environment and produces air pollution hazard. The main objective of this research is to study agricultural crop diversity in and around cement industries and assess the impact of the dust given out by a cement factory on the physico chemical characteristics of the important agricultural crops

## STUDY AREA

Ariyalur is one of the district head-quarters of Tamil Nadu. The area lies at 11° latitude and 79° longitude extending about 7.26 Sq. Km. The soil is black clay loamy with calcareous deposits. Due to the presence of enormous amount of calcium carbonate in the soil, the area suits for the installation and working of large number of cement industries. The list and location of major cement industries in Ariyalur district of Tamil Nadu is presented in Table. 1. Ariyalur cement works, a unit of TANCEM and one of the oldest cement industries in Ariyalur, established in the year of 1979. The factory produces nearly 1500MT/day to meet the demand of the society and it has employed dry process method. During its production, a considerable amount of dust is produced from various processes of the industry. Finally, it releases a stack emission of 1,65,000 $m^3$/hr to the atmosphere. The dust particles are carried over to surrounding areas and deposited into the soil and affects the vegetation.

### Manufacturing of cement

Cement results from the grinding of the clinker with other additive (including gypsum) which are intended to improve its setting qualities. The clinker is produced by burning the mixture of limestone, clay and sometimes irons ore or bauxite, to higher temperatures such as (1450 – 1600°C).Air pollution has long been known to have an adverse effect on plants. Initially, sulphur dioxide was considered as a dangerous pollutant. Now, with the advent of various new industrial processes, the range of harmful pollutants, mainly suspended particulate matter (SPM), has multiplied tremendously. Presence of any other gas or solid in the air degrades its quality and when the concentration of the extraneous gases and solids exceed certain limit, they adversely affect not only the living beings but also the plants.

Plant injury caused by air pollution is most common near large cities, industries such as refineries, power plants as well as cement industries, if pollution is not controlled. Damage in isolated areas occurs when pollutants are spread through longer distances by wind currents. Factors that govern the extent of damage and the region where air pollution is a problem are type and concentration of pollutants, distance from the source, length of exposure and meteorological conditions.

### Scenario of Cement Manufacturing in India

The first cement plant in India was commissioned in 1914 with installed capacity of 10,000 tonne per annum at Porbandar, Gujarat by Indian Cement Company Limited. Since then industry has come a long way. At present there are 128 large cement plants in India having installed capacity of 152.14 million tonnes and actual production of 125.56 million tonnes. India ranks world's second largest cement plants are comparable to the world best in technology absorption, energy conservation as well as environment pollution control.

In the above context, let us take the case of the Gujarat Ambuja Cements Limited (GACL). A cement plant has been set up by this Company at Darlaghat, Dist Solan Himachal Pradesh in 1994 with the corporate philosophy of 'sustainable Development'. The plant covers approximately 50 has of non-prime land with a mining area of about 3 km away from the plant site in village Kashlog. The mines are open cast captive mines and limestone is brought to plant by overland belt conveyor passing through tunnels. A Glass Bag House is attached to the Raw Mill and Kiln section. The uniqueness of this plant is that the bag filters have been installed at each loading and transfer points to minimize dust emission. A Sewage Water Reclamation Plant for an average flow of 220 cu. m/day treats domestic wastewater and 100% treated water is recycled for cooling and other purposes. An Environmental Management Division has been set up with Lab to monitor air and water emissions and water emissions and meteorological conditions. Nurseries have been set up in the plant and mining area to improve the vegetation in the vicinity of plant and mines. The emissions from the plant are below 50µg/ m3 and noise and vibration in the mines are maintained much below the stipulated standards.

**Cement Manufacturing Process**

There are three distinct process for manufacturing of cement, mainly wet process, semi-dry process and dry process. Today with the advancement of technology, dry process has been adopted with precalciner. Cement is manufactured (Saxena, 1990) in four basic stages:-

Quarrying and crushing,

- Grinding and blending of raw materials.
- Clinker production, and
- Finished grinding

Cement manufacturing due to dry process contributes significantly to air pollution, not only in the vicinity of the works but also in the surrounding area, from mining raw material to the end of process. Large quantities of pulverized material to final packing of cement, resulting emission of dust and in turn causing air crushing of raw material to final packing of cement, resulting emission of dust and in turn causing air pollution.

## MATERIALS AND METHODS

A survey was made to record the list of various agricultural crop cultivated in and around the cement factory area, Ariyalur, Tamil Nadu. All plant species were collected from cement dust polluted area and were identified with the help of Flora of Presidency of Madras (Gamble, 2005), in addition to that, they were categorized into herbs, shrubs and trees and they are presented in the Table.2 and Plate I and II.

**Survey of Agricultural crops**

In the area surveyed, it has been reported that the agricultural crops such as chillies (*Capsicum frutescens*), groundnut (*Arachis hypogaea*), greengram (*Vigna radiata*), rice (*Oryza sativa*) and sorghum (*Sorghum bicolor*) are extensively cultivated so a study was conduted to know the impact of cement dust on this agricultural crops cultivated in villages located around cement factory area.

Five plant samples of each crop were collected from the cement dust polluted area (3 km distance for the emission source towards wind direction) at the time of harvest. Similar crop plants collected from the non-polluted area (10 km distance away from the emission source) treated as control. The morphological growth parameters such as plant height (cm/plant), total leaf area ($cm^{-2}$/plant), dry weight (g/plant) and yield (kg/hectare) of crop plants taken from both control and polluted sites were recorded and presented in Table 2.

**Plant height was recorded** by using cm scale; the total leaf area was calculated by measuring the length and width of the leaf as described by Yoshida *et al.* (1972).

$$\text{Leaf area } (cm^2) = K \times \text{length} \times \text{breadth}$$

Where

K = Kemp's constant (for dicot leaves 0.66)

**Fresh weight and Dry weight**

The plant samples taken for morphometric studies were used for determination of fresh weight and dry weight of plant material. They were dried in a hot air oven at 80°C for 24 hrs and their dry weight was determined by using an electrical single pan balance.

**Yield**

The yield was obtained by weighing the harvested consumable parts like fruit in Chilly (*Capsicum frutescens*) fresh weight, seeds in Groundnut (*Arachis hypogaea*) and Greengram (*Vigna radiata*) dry weight, grains in Rice (*Oryza sativa*) and Sorghum (*Sorghum bicolor*) dry weight and expressed in terms of kg/hectare. All the morphological growth parameters such as plant height (cm/plant), total leaf area ($cm^{-2}$/plant), dry weight (g/plant) and yield (kg/hectare) of crop plants taken from both control and polluted sites are recorded and presented in Table 2.

## RESULTS AND DISCUSSION

The survey conducted for the plant bio-diversity in the study area prominently reviled 41 different plant species belonging to 26.

The Earth's biological diversity is being lost at a rate that impoverishes our quality of life and threatens our future. We threaten nature by our activities; the consequences inturn threatens us An increase in quarrying, open-cast mining and road traffic suggest that dust deposition onto vegetation may be increasing. In this the cement industries play a major role causing imbalance of the environment and produces air pollution hazards. In recent years, air pollution due to emission of cement dust is assuming a mammoth proportion due to installation of more and more cement plants to meet the ever-growing needs of the society. They pollute the environment in the form of dust to the surrounding area of cement factories (Uma *et al*., 1994 and Ayanbamiji and Ogundipe, 2010). Due to high energy consumption and dust emission potential, cement industry falls under red category (high polluting industries) by Ministry of Environment and Forest, Goverment of India.

The presence of air pollutants affects the distribution of plant diversity which is highly dependent on the presence of pollutants in the ambient air (Dwivedi and Tripathi, 2007).The amount of cement dust deposited on leaves are found to vary from species to species. The differences in the amount of foliar dust deposition may be due to texture of leaves as well as their angularity with the stem. The leaves that are pubescent, fleshy, larger in size offer greater chance for dust deposition, whereas the leaves with shining and smooth surface, small in size offer lesser chance for the deposition.The rough and hairy leaves collect particulates upto seven times efficiently than the smooth leaves (Little, 1977). Air pollution plays important functional role in many types of vegetation. It is clear that the cement dust pollution is an operative ecological factor causing deterioration in the quality of environment (Shah *et al*., 1989).

The impact of environmental pollution due to cement dust on natural vegetation is well documented by earlier workers (Agarwal *et al*., 1987; Gupta and Mishra, 1994 and Hegazy, 1996). The distribution of species around a cement works was studied and alterations in the composition and frequency of plant ecotypes were reported by Sree Rangaswamy *et al*. (1973). The chemical nature of pollutants and the amount of dust produced by cement factories has an adverse effect on local vegetation (Lerman and Darley, 1975). Topic (1999) indicated that the cement industry was the main cause of destruction of vegetation cover and environmental degradation in various countries. The cement dust spread over large area affect vegetation and natural resources (Singh and Srivastava, 2002). The changes in growth of Olive trees grown under cement dust polluted area were reported. A reduction in number of plant species during monsoon near cement factory area due to crust formation on leaf surface in the presence of moisture in rainy season was reported (Pathbanaban *et al*., 1979).

Shafig and Iqbal (1987) and Abdullah and Iqbal (1991) recorded less number of floras around cement factory area when compared to other industries. Cement dust caused removal of a number of raindeer Lichens (*Cladonia rangifera*) in a zone around factory (Kortesharju *et al*., 1990). Grantz *et al*. (2003) reported that the direct physical effects of mineral dusts on vegetation became apparent only at relatively high surface loads (>7 g $m^{-2}$). The impact of cement dust accumulation on vegetation and seed banks in Egypt were reported by Adamson *et al*. (1994). Cement dust contains heavy metal which are hazardous to the biotic environment and causes impact on ecosystem, man and animal health (Kumar *et al*., 2008; Cipurkovic *et al*., 2011). Fakhry and Migahid (2011) reported the decrease in vegetation diversity in response to cement dust pollution. Cement dust can spread over a wide area and accumulated on lichen, plants, animals and soils in and around cement factory (Schuhmacher *et al*., 2009 and Al-Saleh *et al*., 2011).

Cement dust coming from the cement factories creates serious pollution problem and cause enormous damage to biotic and abiotic components. The crust is formed over the surfaces of soil and leaves because of the settling dust. It consists of calcium silicates which are typical of the clinkers (Burned lime stone) from which cement is made. In the presence of free moisture, calcium silicate ($CaOSio_2$) and calcium aluminate ($CaOAl_2O_2$) undergoes slow hydration, form a colloidal gel, which crystallize and solidify to form an impervious hard crust over the surface of plant and soil. Cement dust has been reported to be harmful to plants, the dust forms thick crust, which interfere with light absorption that leads to stunted growth and growth of fewer leaves in plants (Borka, 1986). The summative effect places a very important role. In the study area all the plants showed stunted growth, reduction in the total leaf area and fresh, dry weight all this contributing to the significant reduction in the yield. In a habitat all the habits of the living organism contribute to its establishment increased pollution leading to alteration of natural chemical symphony of the environment leading to various physical changes like change in temperature, and many more attributes affecting the metabolic pathways leading to destruction of the organisms along with its habitat. The Earth's biological diversity is being lost at a rate that impoverishes our quality of life and threatens our future.

Dust pollution is often ignored in, our country street food, eateries serves not only food but pollutants along with the food. This pollutants includes heavy metals, carcinogens and many other fine particles of chemical residues for this we not only pay heavy price but also we become prey for ultimately leading to severe fatal health problems further impact of this pollutant and human health need to be investigated.

## CONCLUSION

Cement dust pollution places a key factor in altering the ecosystem and it as greater negative implication on the local vegetation which can't be ignored and in long term it would drastically affect the ecosystem linkages by altering nature of soil, absorption of light, photosynthesis which would be a serious peril to biotic community.

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***Musa paradisiaca***

***Cucurbita maxima***

***Lycopersicum esculentum***

**Cement on Agricultural Crops**

**PLATE-I Some important agricultural crop**

**Table. 1 List of cement industries located in Ariyalur District of Tamil Nadu**

| S.No. | Name of the cement industry | Location | Annual production (lakh tonnes) |
|---|---|---|---|
| 1. | Dalmia cement | ThamaraiKulam | 5.25 |
| 2. | India cement | Dalavoi | 9.13 |
| 3. | Ramco Cement | Govindapuram | 5.25 |
| 4. | Chettinad Cement | Keelapalur | 4.00 |
| 5. | Tamil Nadu cement | Kallan Kuruchi | 5.00 |
| 6. | Grasim Industries | Reddipalayam | 4.00 |
| 7. | Alagappa cement | Pudupalayam | Not available |
| 8. | Vijay cement | Melmathur | ,, |
| 9. | Sankar cement | Dalavoi | ,, |
| 10. | Dharani cement | VeenakaiKatti | ,, |

**Table.2.Growth and yield response of some economically important Agricultural crops grown in cement dust polluted sites and control sites**

| S. No | Name of the plant | Plant height (cm/plant) | | Total leaf area ($cm^2$/plant) | | Dry weight (g/plant) | | Yield (kg/hectare) | |
|---|---|---|---|---|---|---|---|---|---|
| | | Control | Polluted | Control | Polluted | Control | Polluted | Control | Polluted |
| 1. | Chilly (*Capsicum frutescens*) | 68.67 ±3.431 | 63.10 ±3.155 | 462.56 ±23.128 | 390.30 ±9.515 | 4.50 ±0.225 | 3.20 ±0.160 | 850.53 ±42.526 | 724.32 ±36.216 |
| 2. | Groundnut (*Arachis hypogaea*) | 36.89 ±1.844 | 31.21 ±1.560 | 231.98 ±11.599 | 187.45 ±9.372 | 9.14 ±0.457 | 7.65 ±0.382 | 840.51 ±42.025 | 746.34 ±37.317 |
| 3. | Greengram (*Vigna radiata*) | 48.43 ±2.421 | 42.95 ±2.147 | 301.23 ±15.061 | 265.01 ±13.250 | 9.43 ±0.471 | 6.92 ±0.346 | 345.12 ±17.256 | 216.32 ±10.816 |
| 4. | Rice (*Oryza sativa*) | 81.53 ±4.076 | 73.12 ±3.656 | 670.12 ±33.506 | 580.27 ±29.013 | 3.19 ±0.159 | 2.80 ±0.140 | 2515.43 ±125.771 | 1946.12 ±97.306 |
| 5. | Sorghum (*Sorghum bicolor*) | 137.29 ±6.864 | 119.67 ±5.983 | 823.45 ±41.172 | 693.28 ±34.664 | 4.78 ±0.239 | 3.01 ±0.150 | 1210.43 ±60.521 | 987.45 ±49.372 |

±Standard deviation

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RESEARCH ARTICLE

# Foliar Application of Seaweed Bio Formulation Enhances Growth and Yield of Banana cv. Grand Naine (AAA)

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## ABSTRACT

Seaweed extracts have been reported to improve the yield and quality of different fruits and vegetables. In this study, a field experiment was conducted at National Research Center for Banana, Trichy, and Tamil Nadu, to evaluate the effect of different seaweed bio-formulations developed by Sea6Energy Pvt Ltd. Bangalore, on growth, yield and quality of Banana cv.Grand Naine. Four different seaweed formulations (LBS) were applied as foliar spray at vegetative and early flowering stages of the plant. It was observed that foliar application of LBS6S@1ml/L improved the bunch weight significantly by 25.24% over control followed by LBS3@5ml/L with 12.62% over water control. The number of hands and fingers per bunch also increased to 5.78% and 6.6% respectively with LBS6S @1ml/l treatment over control.

**Keywords:** Banana, cv. Grand Naine, Sea weed Bio formulations, Yield.

## INTRODUCTION

Banana is an economically important fruit crop. It is the second most important fruit crop in India next to mango. Banana is very popular fruit due to its year round availability, taste, low price and high nutritive value. India is the largest producer of Banana in the world, followed by China, Philippines, Brazil and Ecuador. As per NHB estimate (National Horticulture Board), India produces 29.895 million tons on an area of 0.837 million ha [1]. Around 87% of all the bananas grown worldwide are produced by small-scale farmers for domestic use, while the remaining 13%, mainly dessert bananas, are traded internationally. Production of Banana is highest in Maharashtra (3924.1 mt), followed by Tamilnadu (3543.8 mt). It accounts for 13% of the total area and 33% of the production of fruits.

The banana cv. Grand Naine (AAA) is a Cavendish group of banana and an internationally accepted dessert banana cultivar. It is also extensively used for processing purposes and has occupied the premier export market. Grand Naine is gaining popularity due to its tolerance to abiotic stresses and its fruit quality such as size, colour, well-spaced hands, shelf life etc. as compared to other cultivars. Food security and climate change is one of the major challenges across the world. Sustainable agriculture has the unique potential to mitigate climate change and strengthen resilience to the impacts of climate change. Vision for sustainable agriculture is therefore important for the world. The use of eco-friendly bio-products such as seaweed extracts has emerged as one of the important strategies for sustainable agriculture. Use of seaweed extracts in agriculture could deliver an important contribution to sustainable food production.

Seaweeds are macroscopic, multicellular marine algae that commonly inhabit the coastal regions of the world's oceans. There are more than 9,000 species of macroalgae which are broadly classified into three main groups viz., Rhodophyta (red algae), Phaeophyta (Brown algae) and Chlorophyta (green algae), based on pigments present in them. The scientific benefits of applying seaweed extracts in agriculture have been extensively reported and well-reviewed in peer-reviewed scientific publications and more broadly in the plant biostimulant [2, 3, and 4]. Seaweed extracts have been used to increase crop yield, improve growth and induce resistance to biotic an abiotic stress and increase nutrient uptake from soil.Around 15 million metric tonnes of seaweed products are produced annually (FAO 2006), a considerable portion of which is used for nutrient supplements and as biostimulants or biofertilizers to increase plant growth and yield. Extract of *Ascophyllum nodosum*, a cold water brown macroalage is well documented for its use in agriculture. Its application has been shown to increase yield and productivity of different crops such as lettuce, cauliflower, spinach etc. [5, 6, 7, 8]. However, this seaweed is harvested from the wild stocks from the natural bed of the ocean. This limits its scalability in agriculture with the growing demand of food around the world.

*Kappaphycus alvarezii* is tropical seaweed and it is being cultivated in India since more than a decade for extraction of thickening agent called kappa-carrageenan using traditional farming systems. Extracts from Kappaphycus have also been reported to improve crop productivity. Foliar application of *K. alvarezii* extract on okra significantly increased the yield (20.47%) [9]. Similarly, application of *K. alvarezii* extract improved yield of wheat (*Triticum aestivum*). The nutritional quality of grain such as carbohydrate, protein and minerals was also improved under the influence of treatment [10]. Sea6Energy Pvt. Ltd., Bangalore has developed mechanised farming system to cultivate *Kappaphycus alvarezii* in rough sea water using patented technologies [11]. The company has also prepared different liquid bio-formulations from *Kappaphycus alvarezii* for use in agriculture using patented technology of seaweed processing [12].

The present study is to investigate the potential of different seaweed derived formulations on improving the yield and fruit quality of banana (var. Grand Naine) under field condition. The field trial was conducted at National Research Center for Banana (ICAR), Trichy, and Tamil Nadu.

## MATERIALS AND METHODS

### Preparation of seaweed extract

*Kappaphycus* species of seaweed biomass was cultivated in the tropical waters of southeast coastline of the Indian states. The seaweed biomass thus obtained was crushed to separate solid and liquid fractions. These fractions were further processed to prepare different extracts using the patented methods of extraction by Sea6 Energy Pvt Ltd [12].

**Preparation of trial plot**

The experiment was conducted at Research and Development plot of National Research Centre for Banana, Tamilnadu farm located at 10° 50' N latitude 74° 50' E longitude. Prior to planting banana, land was ploughed and levelled. During soil preparation basal dose of FYM is added and thoroughly mixed into the soil. A pit size of 45cm x 45cm x 45cm is dug out. Prepared pits were left to solar radiation helps in killing the harmful insects, are effective against soil borne diseases and aids aeration. The pits were refilled with topsoil mixed with 5 kg of FYM (well decomposed), 300g of single super phosphate, and 150 gm of Neem cake and 10 gm of Carbofuran. After that the pits were irrigated well a day before planting and planted next day in the pit after removing the plastic bags of tissue cultured plants. The experimental plot had silty clay loam soil type with pH of 7.9- 8.1.

**Variety selection and seedling transplantation**

A triploid banana cultivar 'Grand Naine, (AAA) was used for this study. The disease-free tissue cultured plants of this cultivars was obtained from Blossom Tissue Culture Nurseries, Hosur, Tamil Nadu. The plants were laid out in a simple randomized design in the field and planted with a distance of 1.8 m x 1.8 m. Each plant was considered as one replication. Minimum of 12 plants were maintained for each treatment.

**Treatment details**

All the seedlings were foliar sprayed before planting as per the dosage of different treatments given in Table 1. Subsequently, the various bioformulations were applied as foliar spray at 60, 120, 180 days after planting i.e during vegetative stage and at flowering stage.Standard package of practice as recommended by NRCB was followed for irrigation, pest control and fertilizer application. The Banana Sakthi (micro nutrients from NRCB) was also given at 3$^{rd}$, 5$^{th}$ and 7$^{th}$ months after planting. The experimental plot was maintained as weed free with manual weeding and frequent loosening of soil.

**Propping of banana plants**

Casuarinas poles were provided as support to the developing bunch to avoid lodging and uprooting of plants owing to the heavy banana bunches and to protect the banana plants from wind damage.

**Bunch care**

For enhancing the bunch grade in terms finger size and bunch weight bunch care is important. After full opening of banana bunches, the male bud was removed and sprayed with potassium sulphate (1.5%) thoroughly drenching the bunch first one week after the opening of the last hand and a second spray one month later.

**Collection of data**

Physical parameters such as plant height, plant girth, no. of green leaves, no. of days to flowering, bunch weight, no. of hands, no. of fingers, finger length, finger girth, unripened fruit to pulp ratio were measured. For post-harvest analysis, the harvested fruits were artificially ripened with ethylene and the ripened fruits were analysed for their TSS (Total Soluble Solids), acidity and pulp to peel ratio.

## RESULTS AND DISCUSSION

Foliar application of LBS6S@1ml/l improved the bunch weight significantly by 25.24% over water control followed LBS3@5ml/l with 12.62% over water control (Table 2, Fig 1). The number of hands and fingers per bunch also recorded an increase of 5.78% and 6.6% respectively with LBS6S @1ml/l treatment over water control (Table 3). However, no significant difference was observed in the vegetative parameters i.e., number of leaves, plant height

and girth due to the application of seaweed bioformulations (Table 2). The ratio of pulp to peel in the unripened fruits from plants treated with the bioformulations was higher as compared to water control. LBS6S@1ml/l treatment enhanced this parameter significantly by 28.77% over water control followed by 18.7% with LBS6@1ml/l. In addition, low TSS was recorded in unripened fruits from plants treated with LBS6@1ml/l as compared to water control. Higher pulps to peel ratio and low TSS in unripened fruits are highly desirable traits (Table 4). However, no significant difference in finger length and girth was observed due to the treatments.

Overall, the application of seaweed derived bioformulations has improved several crop parameters and yield in banana. Tropical red seaweed bioformulations used in the present study have shown enhanced root development, improved photosynthetic efficiency and better nutrient uptake in several plant model systems such as mung bean, cucumber, tomato, rice in lab conditions and field (data unpublished).The seaweed bioformulations used in this study are rich in potassium and functional saccharides from *Kappaphycus alvarezii* and these components are known to have plant growth promoting effect. Potassium is one of the principle plant nutrients and it is involved in many physiological processes such as stomatal movement, photosynthesis, enzyme activity etc. [13, 14]. Application of potash has also been shown to increase the yield and quality of maize, wheat, soybean and cotton [15]. Thalooth et al. (2006) showed that foliar application of potassium enhances the vegetative growth of the plants and improves crop yield. It is also well established that seaweed cell wall oligosaccharides can stimulate or inhibit growth and development in plants [16]. Gonzalez et al. (2013) also showed that oligosaccharide from red seaweed increase growth of tobacco plants by enhancing photosynthesis, basal metabolism, and cell division as well as metabolic pathways involved in nitrogen and sulfur assimilation [17, 18].

## CONCLUSION

It can be concluded from the present study that the yield of Banana Cavendish cv. Grand Naine, (AAA) is enhanced by foliar application of seaweed formulations. Based on the yield & yield components and quality parameters, the seaweed bio-formulations LBS6S@1 ml/L has performed the best followed by LBS3 @5ml /L.

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**Table 1: List of treatments**

| S. No. | Chemical | Dosage |
|---|---|---|
| T1 | LBS3 | @ 5 ml/L |
| T2 | LBS4 | @ 2 ml/L |
| T3 | LBS6 | @0.5 ml/L |
| T4 | LBS6 | @ 1 ml/L, |
| T5 | LBS6S | @ 1 ml/L |
| T6 | Control | Water spray |

**Table 2: Effect of sea weed bio-formulations on Plant Growth parameters at flowering in cv. Grand Naine**

| Treatment | Height | Girth | No. of Green Leaves | No. of days taken for flowering |
|---|---|---|---|---|
| Control (Water Spray) | 218.71 | 66.53 | 14.11 | 231.54 |
| LBS 3 @ 5 ml/L | 214.04 | 65.36 | 13.78 | 238.21 |
| LBS 4@ 2 ml/L | 213.54 | 65.19 | 14.28 | 237.13 |
| LBS 6@ 0.5 ml/L | 216.44 | 69.89 | 14.22 | 241.39 |
| LBS 6@ 1ml/L | 216.22 | 68.17 | 14.00 | 238.78 |
| LBS 6S @ 1 ml/L | 215.04 | 64.53 | 13.86 | 252.46 |
| General Mean * | 210.65 | 64.58 | 13.97 | 249.30 |
| CD at 5% * | 6.55 | 6.63 | NS | 32.39 |
| CV (%) * | 5.03 | 8.79 | 3.86 | 9.42 |

*General Mean, CD and CV values provided here is for the complete study conducted.

**Table 3 : Effect of sea weed bio-formulations on Plant Growth parameters at flowering in cv. Grand Naine**

| Treatment Name | Bunch wt.(Kg) | No. of hands | No. of fingers | Finger Length (cm) | Finger girth (cm) |
|---|---|---|---|---|---|
| LBS 3 @ 5 ml/L | 20.88 | 8.65 | 147.94 | 19.67 | 13.00 |
| LBS 4@ 2 ml/L | 18.13 | 8.32 | 141.53 | 20.33 | 12.67 |
| LBS 6@ 0.5 ml/L | 19.88 | 8.49 | 140.69 | 22.00 | 13.50 |
| LBS 6@ 1ml/L | 20.44 | 9.22 | 151.89 | 21.67 | 13.33 |
| LBS 6S @ 1 ml/L | 23.22 | 9.33 | 158.33 | 21.07 | 12.71 |
| Control (Water Spray) | 18.54 | 8.82 | 148.44 | 21.00 | 13.83 |
| General Mean* | 20.55 | 8.99 | 153.89 | 20.91 | 13.18 |
| CD at 5%* | 2.93 | 0.76 | 14.42 | NS | NS |
| CV(%)* | 14.69 | 8.36 | 5.30 | 5.32 | 3.85 |

*General Mean, CD and CV values provided here is for the complete study conducted.

**Table 4: Effect of sea weed bio-formulations Post-harvest quality parameters of un-ripened green banana fruits**

| Treatment Name | Un-ripened Fruit Pulp/peel ratio | Un-ripened Fruit TSS* | Ripened fruit TSS* | Ripened fruit Acidity |
|---|---|---|---|---|
| Control (Water Spray) | 1.39 | 3.06 | 20.00 | 0.30 |
| LBS 3 @ 5 ml/L | 1.43 | 3.25 | 19.62 | 0.30 |
| LBS 4@ 2 ml/L | 1.55 | 2.82 | 19.54 | 0.37 |
| LBS 6@ 0.5 ml/L | 1.63 | 3.85 | 18.84 | 0.30 |
| LBS 6@ 1ml/L | 1.65 | 2.94 | 19.74 | 0.31 |
| LBS 6S@ 1 ml/L | 1.79 | 2.91 | 20.47 | 0.32 |
| General Mean** | 1.56 | 3.08 | 19.93 | 0.31 |
| CD at 5%** | 0.32 | 1.13 | NS | 0.06 |
| CV (%)** | 4.67 | 1.97 | 2.58 | 2.39 |

* Total soluble solids

** General Mean, CD and CV values provided here is for the complete study conducted.


**Fig-1: Harvesting of fully matured banana and weighing in the field.**
**(A) Control (Water spray) (B) Foliar Spray with LBS 6**

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RESEARCH ARTICLE

# Ariyalur, Cement City of India its Present Status and Momentous Dust Pollution

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## ABSTRACT

Ariyalur, Cement city of India constitute for maximum production of cement and this not only affects the life style economy of people in the country but also their health. Pollutants modify air thus life .In the study ambient air quality of ariyalur region in terms of particulate matter and a biotic factors have been enumerated.

**Keywords:** Particulate matter, Meteorological, Dust pollution, Pollution control

## INTRODUCTION

Even thou Ariyalur is called as Cement city of India significant data on the a biotic factors are not available there are several source of pollution at Ariyalur, which once had beautiful natural ambiance pollution studies in such rural places are highly ignored as this place itself is ignored among different pollution air pollution as major and direct impact on all life forms in the habitat. Air pollutants can be harmful to human health if their concentrations exceed certain acceptable levels. Cement manufacturing industries have contributed substantially to the air pollution problem as point source of emission. Fallout of cement factory emission is determined by several factors such as variation in cement manufacturing processes, efficiency of emission control devices, meteorological and topographical conditions. Understanding the current status of ambient air quality in and around Arasu cement Industry, Ariyalurwill help to identify the probable sources of pollutants to facilitate the effective control measures. The aim of the present work is to determine the ambient air quality of ariyalur region in terms of particulate matter and also to enlighten on a biotic factors.

## MATERIALS AND METHODS

### Study area

Ariyalur has witnessed an unusual phenomenon of increasing of the sea level about 120 million years ago. The area located in the vicinity of a cement factory is approximately 4 km in the North east direction from the main town of Ariyalur district. About 65 km away from Tiruchirapalli city (North west direction) and is located 250 km south west of Chennai city. The area lies at $11^{o}$ latitude and $79^{o}$ longitude extending over 120 acres of land.The first cement factory in this area was started by 1978 and the cement manufacturing type happens to be dry process. There are three pairs of smoke stacks at different heights operating throughout the year. The first pair is the tallest smoke stack is Electrostatic precipitator dust (ESP), the second one is umbrella shaped, slightly lower than the first. The concentration of PM was greater within the radius of 2km and lesser at about to 5 km radius depending upon the wind direction and wind velocity.

The area around the factory is cultivatable with dry and wetland farming. The soil is black, clay loamy with calcareous deposits. The average annual rainfall is about 2.89 cm. Rainfall is maximum during October and November, mainly due to depression of North east monsoon. The hottest months of Ariyalur commences from January to July, due to the radiation of the calcareous soil.The study area has hot and continental monsoon climate with occasional maritime effects. The major sources of rainfall in this area are northeast monsoon and the area is categorized as the semi-arid zone. Climatically the area witness the following four seasons.

- Pre monsoon – March to may
- Southwest monsoon– June to September
- Northeast monsoon –October to December
- Winter– January to February

### Selection of sampling sites

The sampling sites were chosen with the following criteria as guide lines. It should provide a true representation of the receptor sites. Sampling site should be two to four kilometers around the cement factory. It should not be behind a tall building or structure and should not isolate itself frombeing exposed to the specific localized sources. The sites may be on concentric circular lines with the area of interest in the center.To study the extent and effect of pollutants on common people, five residential and sensitive areas were selected. The selections of stations are such that they covered all the four directions.

### Sampling sites

1. Anna Nagar(AN) / Colloge Campus
2. Maruthi Nagar(MN)
3. Manaleri(ML)
4. Kallenkuruchi(KK)
5. Chettithirukkonam.(CT)

### Meteorological parameters

The data for the climatological factors of Ariyalur were analysed for the period of five years during January 2002 – December 2006.A comparison of the data was made with that of the data obtained from the Ariyalur meteorological

centre, Ariyalur district. The various factors studied are temperature, relative humidity, rainfall, rainy days, wind direction and wind velocity. The data were statistically analysed.

## Methods for measurement of meteorological parameters

Themeteorological parameters play an important role in the distribution of pollutants. Hence the present study aims at the detailed study of meteorological parameters, pollutant concentration and also their effect on the vegetation.

## Rain fall

Rain gauge measures the precipitation in millimeter. The type of rain gauge used is standard rain gauge. It was kept away from trees and structures, so that the instrument is not damaged.

### Atmospheric pressure

The atmospheric pressure was measured using the aneroid barometer. It has a light metal case with ring suspension. It has reliable sensor. These barometers are of great accuracy, easy to use and affordable.

### Humidity

The Humidity was measured using round type Hygrometer. It was made up of light weight acrylic. Easily mountable, initially it was calibrated.

### Wind direction recorder

It is operated mechanically without any power supply. The wind vane is an instrument used to measure the wind direction. It is mounted on a roof top about 10 mts above the ground away from building, trees and other objects which interfere with the wind direction. The wind vane used for the present investigation was Hydromat instrument and has been manufactured as per IS 5799; 1970.

### Wind speed recorder

The wind speed was recorded using non mechanical Anemometer which records the number of revolution per minute electronically. This type of anemometer is less sensitive than the cup anemometer. Further using the same instrument the temperature was also recorded.

### Construction of wind rose

The wind rose was constructed based on the meteorological data collected for 5 years as per software, www.environmentallake.com and version 5.9.0.

### Suspended particulate matter (SPM) determination(NEERI, 1991)

The gravimetric method was employed for the quantification of dust fall that is a suitable method for the particles of one micron or larger size. Dust fall samples were collected at four polluted sites and control site from the emission sources during 3 times a day for every month during January 2006-December 2006.

### Dedusting of the equipment

Before mounting the fiber glass filter paper on the sampler, the sample was dedusted carefully with a duster cloth.

### Filter paper preparation

The glass micro fiber filters (what man GF/A) of size 20.3x25.4cm were exposed to the light source and inspected for pinholes, particles or any other imperfection.

The filter sample was placed for drying in a hot air oven (105ºc) for an hour without unfolding the filter. The sampling filter was removed from the oven and cooled in desiccators for 15 minutes. The filter sample was than weighed accurately nearest to the 4the decimal of a gram. The RPM was calculated as follows.

### Determination of respirable particulate matter (RPM)

$$\text{Concentration of RPM} = \frac{W \times 10^6}{V \times T} \ \mu g/m^3$$

Where
V = Flow rate in cubic meter per minutes
T = Total period of sampling in minutes
W = Difference in final and initial weight of filter paper in grams.

### Determination of Non respirable particulate matter (NRPM)

$$\text{Concentration of NRPM} = \frac{W \times 10^6}{V \times T} \mu g/m^3$$

Where
V = Flow rate in cubic meter per minutes
T = Total period of sampling in minutes
W = Difference in final and initial weight of hopper in grams.

### Determination of suspended particulate matter (SPM)

The SPM comprises RPM & NRPM. RPM was calculated using the relation. Concentration of SPM ($\mu g/m^3$) = concentration of NRPM + concentration of RPM.Thus the RPM, NRPM & SPM were calculated.

### Chemical and glass wares

Analar grade chemicals of BDH and Merck (India) and glass wares from Borosil were employed for all the experiments.

### Air quality index

Air quality index is calculated using the formula given as follows

$$AQI = \frac{100 \times Vobs}{Vstd}$$

Where
Vobs – Observed values (μg/m$^3$)
Vstd– Standard values (μg/m$^3$)

The following table explains the AQI values along with the condition of the ambient air in terms of pollution.

### Statistical analysis

The data thus obtained during experiments were tabulated, analysed and interpreted with the help of statistical tools like correlation co-efficient to verify the validity of the data and inferences (Palanismy and Manoharan,).

## RESULTS

The meteorological parameters in the vicinity of cement plants were estimated for a period of 5 years starting from Janauary-2002 to December 2006. The parameters analysed include temperature, rainfall, rainy days, relative humidity, average wind speed and the predominant wind direction. With the data obtained five wind rose have been constructed. Based on these meteorological conditions the study areas were selected representing different direction, with respect to the cement factory. At the selected sites the RPM, NRPM and SPM levels have been analysed for a period of one year (Jan.2006-December 2006) during day and night time.On the basis of SPM value an polluted site and a control site has been selected and the plant of economical importance namely *Helianthus annuus L.* was cultivated.The effect of cement dust on growth parameters, bio-chemical properties and phyto chemical components were assessed. Finally the traffic census of the selected polluted site and control site made. The results are summarized as follows.

### Meteorological conditions of Ariyalur

### Temperature

The meteorological condition of Ariyalur were assessed and tabulated in Table 2 & 6. During the year 2002, the maximum temperatures have been recorded during May and June (39$^o$C) and a minimum has been recorded during September (26$^o$C).For the year 2003, highest temperature 39.8$^o$C has been recorded during April and a minimum21.16$^o$C have been recorded during November.For the year 2004, similar higher temperature, 39.12$^o$C was recorded during April and the lower temperature around 24.0$^o$C was recorded during September.The same trend has been noticed for the year 2005. A maximum of 40.0$^o$C was noticed during April and a minimum value has been noticed during October, (26.19$^o$C.)But for the year 2006, the month of April and June has shown 39.0$^o$C as maximum and the month of November (20.83$^o$C) has shown the minimum temperature.

### Rainfall

For the year 2002, the highest rainfall recorded was 5.13mm during September and for about 3 months there has not been any rainfall recorded. But for the whole year the number of rainy days was just 46 during 2002.During the year 2003 the highest rainfall recorded during October was 12.03 mm and a minimum of 0.03 mm was recorded during January 2004. For the year 2004,the number of rainy days happens to be 62.During 2005, the month of November had a good spell of rain (15.54mm) when compared with June, when the rain has been just 0.3mm. Similarly the number of rainy days has been higher during September, October, and November 2005. (i.e., 12-13 days)

For the year 2006, lesser rainfall has been noticed similar to 2002 and 2003. It is 8.33mm during September and a minimum was 0.26mm during July 2006, and the number of rainy days recorded during 2006 was about 59, similar to that of 2003.

### Relative humidity

The relative humidity was found to be a maximum during the early hours of the day (92%) in the month of November, and minimum value obtained was 54.78% during June 2002 and this value correspond to the sun set period.And for the year 2003, the same month of November has experienced high relative humidity of 96.18% and during the hottest months of the year, April and May the value of about 55% has been recorded.But for the year 2004, the highest relative humidity has been recorded during January (93.8%) and the minimum was noticed during August and the value of 53.4%.During 2005 and 2006 as for the previous years the relative humidity was higher in the month of November (around 91-92%). Though there was a reduction of relative humidity during the hotter months the humidity has not reduced more than 63% during 2005 and not more than 56% during 2006 respectively.

### Average wind speed

As the wind speed is of higher importance with respect to pollution, it was recorded continuously for 24 hours a day and the average has been represented at 8.30 am and 5.30 pm of every day. Finally the monthly average has been tabulated. The highest wind speed recorded was 29.84 km/h during July 2002. Where as the lowest value recorded was 11 km/h during November 2002.For the year 2003, the average wind speed was higher during June and July (29km/h) and the minimum was during the month of December (12.16 km/h). Similar result has been noticed during the year 2004.And during 2005, the maximum wind speed of 29.19 km/h was noticed during July and a minimum has been noticed during August 2005 (11 km/h).Similarly the average wind speed of 27.13 km/h was recorded during July 2006. And a lower value has been recorded during October 2006 (14.72%).

### Wind Direction

The predominant wind direction recorded during 2002-2006 happens to be north east and at times the dominant direction was ENE and toward west.

### Wind Rose

With the acquired meterological data wind rose were constructed for the period of five years (2002-2006).(Fig.1-5).

### Summary of RPM, NRPM and SPM Levels in and around Ariyalur during January 2006 – December 2006.

The level of RPM, NRPM and SPM recorded at various sampling stations has been tabulated in table 4.6- 4.41.(Fig.4.6 – 4.41)

During January 2006, the highest RPM level [$0.80x10^3\mu g/m^3$) has been recorded at MI. Similarly the NRPM level and the SPM level the highest the same region ($0.48x10^3\mu g/m^3$) respectively. The values cited were the day values, and the highest values happen to be 0.78, 0.26, and $1.04x10^3\mu g/m^3$respectively.

During February 2006, the highest RPM level [$0.90x10^3\mu g/m^3$) has been recorded at MR. Similarly the NRPM level and the SPM level the highest the same region ($1.16x10^3\mu g/m^3$) respectively. The values cited were the day values, and the highest values happen to be 0.76, 0.17, and $0.92x10^3\mu g/m^3$respectively.

During March 2006, the highest RPM level [1.06x10$^3$µg/m$^3$) has been recorded at MR. Similarly the NRPM level and the SPM level the highest the same region (1.98x10$^3$µg/m$^3$) respectively. The values cited were the day values, and the highest values happen to be 0.92, 0.78, and 1.70x10$^3$µg/m$^3$respectively.

During April 2006, the highest RPM level [0.41x10$^3$µg/m$^3$) has been recorded at K I. Similarly the NRPM level and the SPM level the highest the same region (0.72x10$^3$µg/m$^3$) respectively. The values cited were the day values, and the highest values happen to be 0.36, 0.22, and 0.58x10$^3$µg/m$^3$respectively.

During May 2006, the highest RPM level [0.64x10$^3$µg/m$^3$) has been recorded at KI. Similarly the NRPM level and the SPM level the highest the same region (1.16x10$^3$µg/m$^3$) respectively. The values cited were the day values, and the highest values happen to be 0.48, 0.42, and 0.85x10$^3$µg/m$^3$respectively.

During June 2006, the highest RPM level [0.61x10$^3$µg/m$^3$) has been recorded at KI. Similarly the NRPM level and the SPM level the highest the same region (1.04x10$^3$µg/m$^3$) respectively. The values cited were the day values, and the highest values happen to be 0.54, 0.38, and 0.92x10$^3$µg/m$^3$respectively.

During July 2006, the highest RPM level [0.62x10$^3$µg/m$^3$) has been recorded at KI. Similarly the NRPM level and the SPM level the highest the same region (1.31x10$^3$µg/m$^3$) respectively. The values cited were the day values, and the highest values happen to be 0.58, 0.41, and 0.99x10$^3$µg/m$^3$respectively.

During August 2006, the highest RPM level [0.33x10$^3$µg/m$^3$) has been recorded at KI. Similarly the NRPM level and the SPM level the highest the same region (0.51x10$^3$µg/m$^3$) respectively. The values cited were the day values, and the highest values happen to be 0.41, 0.17, and 0.58x10$^3$µg/m$^3$respectively.

During September 2006, the highest RPM level [0.81x10$^3$µg/m$^3$) has been recorded at MR. Similarly the NRPM level and the SPM level the highest the same region (1.23x10$^3$µg/m$^3$) respectively. The values cited were the day values, and the highest values happen to be 0.62, 0.38, and 1.00x10$^3$µg/m$^3$respectively.

During October 2006, the highest RPM level [0.32x10$^3$µg/m$^3$) has been recorded at MR. Similarly the NRPM level and the SPM level the highest the same region (0.53x10$^3$µg/m$^3$) respectively. The values cited were the day values, and the highest values happen to be 0.28, 0.25, and 0.53x10$^3$µg/m$^3$respectively.

During November 2006, the highest RPM level [0.41x10$^3$µg/m$^3$) has been recorded at MI. Similarly the NRPM level and the SPM level the highest the same region (0.62x10$^3$µg/m$^3$) respectively. The values cited were the day values, and the highest values happen to be 0.52, 0.18, and 0.70x10$^3$µg/m$^3$respectively.

During March 2006, the highest RPM level [0.35x10$^3$µg/m$^3$) has been recorded at MI. Similarly the NRPM level and the SPM level the highest the same region (0.51x10$^3$µg/m$^3$) respectively. The values cited were the day values, and the highest values happen to be 0.26, 0.17, and 0.35x10$^3$µg/m$^3$respectively.

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**Plate-1.Arasu cement factory –Ariyalur.**

**Plate-2. Experimental set up**

**Table1: Air quality categories based on AQI**

| Category | AQI of ambient air | Description of ambient air quality |
|---|---|---|
| 1 | Below 10 | Very clean |
| 2 | Between 10 and 25 | Clean |
| 3 | Between 25 and 50 | Fairly clean |
| 4 | Between 50 and 75 | Moderately Polluted |
| 5 | Between 75 and 100 | Polluted |
| 6 | Between 100 and 125 | Heavily polluted |
| 7 | Beyond 125 | Severely polluted |

(Mohanty, 1999)

**Table 2:Meteorological conditions of Ariyalur, in the vicinity of a cement plant associated with wind - rose during - January 2002 - December 2002**

| Month (2002) | Temperature | | Rain fall (24 hrs) (mm) | Rainy Days (24 hrs) (mm) | Relative humidity | | Average wind speed (24hrs) | | Predominant wind direction | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Max °C | Min °C | | | 08.30 hrs (%) | 17.30 Hrs (%) | AM ($kmh^{-1}$) | PM ($kmh^{-1}$) | AM | PM |
| January 31days | 27.06 | 26.81 | Nil | Nil | 91.00 | 72.88 | 16.74 | 15.31 | NE | NE |
| February 28days | 39.61 | 32.14 | 3.18 | 5 | 89.81 | 64.57 | 14.17 | 21.61 | ENE | ENE |
| March 31days | 36.67 | 27.59 | Nil | Nil | 79.12 | 56.97 | 19.67 | 18.19 | NE | NE |
| April 30days | 38.86 | 31.64 | Nil | Nil | 75.61 | 58.17 | 12.06 | 20.78 | ENE | ENE |
| May 31days | 39.61 | 28.87 | 2.87 | 4 | 73.63 | 64.58 | 20.69 | 21.70 | WSW | WSW |
| June 30days | 39.83 | 33.00 | 3.50 | 4 | 69.31 | 54.78 | 29.71 | 21.83 | W | W |
| July 31days | 37.69 | 28.41 | 0.65 | 3 | 70.53 | 65.00 | 29.84 | 30.63 | W | WNW |
| August 31 days | 36.59 | 28.87 | 1.26 | 3 | 86.14 | 75.60 | 20.17 | 18.47 | W | W |
| Septem-ber 30days | 35.17 | 26.00 | 5.13 | 7 | 82.19 | 79.37 | 23.51 | 17.42 | WNW | WNW |
| October 31days | 33.61 | 29.33 | 4.53 | 11 | 86.19 | 80.11 | 15.61 | 14.10 | WNW | WSW |
| Novem-ber 30days | 32.11 | 28.11 | 1.75 | 7 | 92.00 | 84.20 | 13.26 | 11.00 | ENE | NE |
| Decem-ber 31days | 28.11 | 29.19 | 0.35 | 2 | 86.00 | 81.81 | 11.49 | 12.68 | NE | ENE |

Values are mean of total number of days of a month.

**Table 3: Meteorological conditions of Ariyalur, in the vicinity of a cement plant associated with wind - rose during - January 2003 - December 2003**

| Month (2003) | Temperature | | Rainfall (24 hrs) (mm) | Rainy Days (24hrs) (mm) | Relative humidity | | Average wind speed (24hrs) | | Predominant wind direction | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Max °C | Min °C | | | 08.30hrs (%) | 17.30 Hrs (%) | AM ($kmh^{-1}$) | PM ($kmh^{-1}$) | AM | PM |
| January 31days | 29.40 | 27.64 | 0.39 | 1 | 87.66 | 81.07 | 18.41 | 20.49 | NE | NE |
| February 28days | 37.61 | 28.17 | NIL | NIL | 79.80 | 63.17 | 16.80 | 19.29 | NE | ENE |
| March 31days | 33.33 | 29.42 | NIL | NIL | 86.50 | 68.63 | 19.77 | 16.39 | ENE | NE |
| April 30days | 39.80 | 26.17 | 0.10 | 1 | 65.46 | 55.90 | 13.08 | 19.66 | ENE | ENE |
| May 31days | 38.00 | 29.63 | 0.58 | 4 | 55.40 | 64.32 | 28.01 | 19.90 | WSW | WSW |
| June 30days | 37.65 | 28.16 | NIL | NIL | 71.12 | 59.44 | 29.61 | 20.19 | W | W |
| July 31days | 38.68 | 35.54 | 2.45 | 4 | 72.30 | 69.67 | 29.00 | 30.74 | W | WNW |
| August 31 days | 27.54 | 31.13 | 3.03 | 9 | 84.90 | 95.30 | 23.48 | 16.39 | W | W |
| September 30days | 33.49 | 29.11 | 2.28 | 7 | 80.35 | 89.09 | 24.17 | 20.10 | WNW | WNW |
| October 31days | 33.07 | 27.80 | 5.60 | 13 | 80.13 | 78.90 | 22.15 | 19.16 | WNW | WNW |
| November 30days | 30.21 | 21.16 | 7.52 | 15 | 94.05 | 96.18 | 17.11 | 13.20 | ENE | NW |
| December 31days | 20.13 | 22.06 | 0.32 | 2 | 72.09 | 67.80 | 12.16 | 14.65 | NE | NW |

Values are mean of total number of days of a month.

**Table 4: Meteorological conditions of Ariyalur, in the vicinity of a cement plant associated with wind - rose during - January 2004 - December 2004**

| Month (2004) | Temperature | | Rainfall (24 hrs) (mm) | Rainy Days (24hrs) (mm) | Relative humidity | | Average wind speed (24hrs) | | Predominant wind direction | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Max °C | Min °C | | | 08.30hrs (%) | 17.30 Hrs (%) | AM ($kmh^{-1}$) | PM ($kmh^{-1}$) | AM | PM |
| January 31days | 29.14 | 26.01 | 0.03 | 1 | 93.81 | 90.11 | 16.91 | 18.23 | WE | NE |
| February 28days | 38.08 | 36.70 | Nil | NIL | 86.12 | 60.90 | 14.36 | 20.70 | NE | ENE |
| March 31days | 38.87 | 34.60 | Nil | NIL | 80.36 | 62.11 | 18.11 | 20.32 | ENE | ENE |

| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| April 30days | 39.12 | 33.61 | 1.33 | 1 | 79.81 | 62.70 | 14.21 | 16.30 | WSE | ENE |
| May 31days | 34.53 | 30.50 | 11.69 | 13 | 75.01 | 67.51 | 20.31 | 21.36 | W | WSW |
| June 30days | 29.76 | 28.11 | 0.66 | 1 | 63.66 | 68.15 | 28.86 | 30.80 | WSW | W |
| July 31days | 28.92 | 27.19 | 2.61 | 4 | 83.14 | 65.60 | 24.72 | 29.11 | W | SW |
| August 31 days | 32.81 | 30.14 | 1.10 | 2 | 53.40 | 67.90 | 20.65 | 18.14 | WSW | WSW |
| September 30days | 28.17 | 24.00 | 6.10 | 11 | 86.11 | 65.17 | 21.43 | 22.37 | WNW | NW |
| October 31days | 29.11 | 26.79 | 12.03 | 16 | 86.80 | 67.78 | 13.26 | 15.47 | WNW | WNW |
| November 30days | 30.70 | 28.16 | 3.66 | 12 | 90.43 | 71.54 | 15.30 | 12.39 | WNW | ENE |
| December 31days | 31.71 | 30.00 | 0.06 | 1 | 87.00 | 80.41 | 12.60 | 15.76 | NE | ENE |

Values are mean of total number of days of a month.

**Table5: Meteorological conditions of Ariyalur, in the vicinity of a cement plant associated with wind - rose during - January 2005 - December 2005**

| Month (2005) | Temperature | | Rainfall (24 hrs) (mm) | Rainy Days (24hrs) (mm) | Relative humidity | | Average wind speed (24hrs) | | Predominant wind direction | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Max °C | Min °C | | | 08.30hrs (%) | 17.30 Hrs (%) | AM ($kmh^{-1}$) | PM ($kmh^{-1}$) | AM | PM |
| January 31days | 30.28 | 28.14 | Nil | Nil | 90.11 | 88.17 | 18.31 | 16.32 | NE | ENE |
| February 28days | 34.65 | 38.43 | Nil | Nil | 85.87 | 80.91 | 15.62 | 20.22 | NE | ENE |
| March 31days | 36.17 | 38.73 | 0.90 | 2 | 83.19 | 72.12 | 17.36 | 19.25 | NE | NE |
| April 30days | 40.08 | 38.14 | 2.53 | 5 | 79.98 | 69.61 | 15.81 | 20.11 | ENE | ENE |
| May 31days | 37.54 | 36.16 | 6.06 | 5 | 76.67 | 69.81 | 20.91 | 22.36 | WSW | WSW |
| June 30days | 38.63 | 29.76 | 0.30 | 1 | 69.71 | 67.00 | 24.31 | 20.19 | W | WSW |
| July 31days | 38.93 | 27.57 | 4.58 | 2 | 84.21 | 67.19 | 28.80 | 29.19 | WSW | W |
| August 31 days | 34.80 | 26.43 | 5.10 | 5 | 63.29 | 66.14 | 20.56 | 11.00 | WNW | W |
| September 30days | 30.15 | 28.61 | 4.53 | 12 | 80.90 | 63.25 | 20.39 | 19.41 | WNW | WNW |
| October 31days | 24.91 | 26.19 | 2.85 | 13 | 85.13 | 66.17 | 15.48 | 22.01 | W | WNW |
| November 30days | 28.24 | 26.42 | 15.54 | 12 | 92.16 | 72.38 | 13.76 | 12.81 | NE | NE |
| December 31days | 28.98 | 27.15 | 9.26 | 9 | 89.08 | 80.91 | 13.91 | 16.81 | NE | ENE |

Values are mean of total number of days of a month.

**Table 6**: **Meteorological conditions of Ariyalur, in the vicinity of a cement plant associated with wind - rose during - January 2006 - December 2006**

| Month (2006) | Temperature | | Rainfall (24 hrs) (mm) | Rainy Days (24hrs) (mm) | Relative humidity | | Average wind speed (24hrs) | | Predominant wind direction | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Max °C | Min °C | | | 08.30 hrs (%) | 17.30 Hrs (%) | AM ($kmh^{-1}$) | PM ($kmh^{-1}$) | AM | PM |
| January 31days | 31.12 | 23.60 | 0.35 | 2 | 91.10 | 76.00 | 17.70 | 19.11 | NE | NE |
| February 28days | 32.43 | 24.18 | NIL | NIL | 89.70 | 64.13 | 13.22 | 18.14 | ENE | ENE |
| March 31days | 35.62 | 27.33 | 0.48 | 4 | 77.78 | 56.18 | 15.63 | 17.18 | NE | NE |
| April 30days | 39.13 | 30.11 | 2.60 | 3 | 75.13 | 58.10 | 20.32 | 22.18 | ENE | ENE |
| May 31days | 38.91 | 30.14 | 4.87 | 5 | 73.41 | 64.80 | 26.17 | 29.12 | WSW | WSW |
| June 30days | 39.70 | 29.18 | 2.90 | 2 | 68.61 | 58.18 | 19.91 | 23.78 | WSW | W |
| July 31days | 38.12 | 27.16 | 0.26 | 1 | 76.33 | 64.16 | 27.13 | 23.72 | W | W |
| August 31 days | 36.14 | 28.13 | 2.10 | 5 | 80.73 | 74.63 | 19.36 | 24.31 | WNW | W |
| September 30days | 35.19 | 28.17 | 8.33 | 10 | 82.47 | 72.13 | 16.90 | 19.82 | WNW | WNW |
| October 31days | 31.17 | 26.03 | 5.00 | 12 | 84.63 | 73.18 | 14.72 | 16.73 | WNW | WSW |
| November 30days | 28.14 | 20.83 | 4.35 | 12 | 91.31 | 70.10 | 18.43 | 19.21 | ENE | NE |
| December 31days | 28.91 | 21.17 | 0.45 | 3 | 86.34 | 68.38 | 15.63 | 17.22 | NE | ENE |

Values are mean of total number of days of a month.

**Table 7: Summary of RPM levels in and around ariyalur during January 2006 (mean values)**

| S.NO | AREA | DAY VALUES $\mu g/m^3$ | NIGHT VALUES $\mu g/m^3$ |
|---|---|---|---|
| 1 | ANNA NAGAR | $0.07 \times 10^3$ | $0.19 \times 10^3$ |
| 2 | MARUTHI NAGAR | $0.09 \times 10^3$ | $0.16 \times 10^3$ |
| 3 | MANALERI | $0.80 \times 10^3$ | $0.78 \times 10^3$ |
| 4 | KALLENKURICHI | $0.32 \times 10^3$ | $0.24 \times 10^3$ |
| 5 | CHETTITHIRUKONAM | $0.05 \times 10^3$ | $0.27 \times 10^3$ |

**Table 8:Summary of NRPM levels in and around ariyalur during January 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.17 X 10³ | 0.24 X 10³ |
| 2 | MARUTHI NAGAR | 0.27 X 10³ | 0.19 X 10³ |
| 3 | MANALERI | 0.48 X 10³ | 0.26 X 10³ |
| 4 | KALLENKURICHI | 0.24 X 10³ | 0.14 X 10³ |
| 5 | CHETTITHIRUKONAM | 0.16 X 10³ | 0.21 X 10³ |

**Table9:Summary of SPM levels in and around ariyalur during January 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.24 X 10³ | 0.43 X 10³ |
| 2 | MARUTHI NAGAR | 0.36 X 10³ | 0.35 X 10³ |
| 3 | MANALERI | 1.28 X 10³ | 1.04 X 10³ |
| 4 | KALLENKURICHI | 0.56 X 10³ | 0.38 X 10³ |
| 5 | CHETTITHIRUKONAM | 0.21 X 10³ | 0.48 X 10³ |

**Table10:Summary of RPM levels in and around ariyalur during February 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.08 X 10³ | 0.19 X 10³ |
| 2 | MARUTHI NAGAR | 0.90 X 10³ | 0.38 X 10³ |
| 3 | MANALERI | 0.34 X 10³ | 0.76 X 10³ |
| 4 | KALLENKURICHI | 0.24 X 10³ | 0.35 X 10³ |
| 5 | CHETTITHIRUKONAM | 0.04 X 10³ | 0.16 X 10³ |

**Table11:Summary of NRPM levels in and around ariyalur during February 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.02 X 10³ | 0.04 X 10³ |
| 2 | MARUTHI NAGAR | 0.26 X 10³ | 0.17 X 10³ |
| 3 | MANALERI | 0.63 X 10³ | 0.16 X 10³ |
| 4 | KALLENKURICHI | 0.13 X 10³ | 0.13 X 10³ |
| 5 | CHETTITHIRUKONAM | 0.22 X 10³ | 0.08 X 10³ |

**Table 12:Summary of SPM levels in and around ariyalur during February 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.10 X $10^3$ | 0.23 X $10^3$ |
| 2 | MARUTHI NAGAR | 1.16 X $10^3$ | 0.55 X $10^3$ |
| 3 | MANALERI | 0.97 X $10^3$ | 0.92 X $10^3$ |
| 4 | KALLENKURICHI | 0.37 X $10^3$ | 0.48 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.26 X $10^3$ | 0.24 X $10^3$ |

**Table 13:Summary of RPM levels in and around ariyalur during March 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.81 X $10^3$ | 0.52 X $10^3$ |
| 2 | MARUTHI NAGAR | 1.06 X $10^3$ | 0.92 X $10^3$ |
| 3 | MANALERI | 0.67 X $10^3$ | 0.58 X $10^3$ |
| 4 | KALLENKURICHI | 0.23 X $10^3$ | 0.41 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.06 X $10^3$ | 0.06 X $10^3$ |

**Table14:Summary of NRPM levels in and around ariyalur during March 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.72 X $10^3$ | 0.65 X $10^3$ |
| 2 | MARUTHI NAGAR | 0.92 X $10^3$ | 0.78 X $10^3$ |
| 3 | MANALERI | 0.48 X $10^3$ | 0.52 X $10^3$ |
| 4 | KALLENKURICHI | 0.26 X $10^3$ | 0.44 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.32 X $10^3$ | 0.23 X $10^3$ |

**Table 15:Summary of SPM levels in and around ariyalur during March 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 1.53 X $10^3$ | 1.17 X $10^3$ |
| 2 | MARUTHI NAGAR | 1.98 X $10^3$ | 1.70 X $10^3$ |
| 3 | MANALERI | 1.15 X $10^3$ | 1.10 X $10^3$ |
| 4 | KALLENKURICHI | 0.49 X $10^3$ | 0.85 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.38 X $10^3$ | 0.44 X $10^3$ |

**Table 16:Summary of RPM levels in and around ariyalur during April 2006 (mean values)**

| S. No. | AREA | DAY VALUES µg/m³ | NIGHT VALUES µg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.20 X $10^3$ | 0.11 X $10^3$ |
| 2 | MARUTHI NAGAR | 0.31 X $10^3$ | 0.28 X $10^3$ |
| 3 | MANALERI | 0.12 X $10^3$ | 0.14 X $10^3$ |
| 4 | KALLENKURICHI | 0.41 X $10^3$ | 0.36 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.09 X $10^3$ | 0.07 X $10^3$ |

**Table 17:Summary of NRPM levels in and around ariyalur during April 2006 (mean values)**

| S. No. | AREA | DAY VALUES µg/m³ | NIGHT VALUES µg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.24 X $10^3$ | 0.31 X $10^3$ |
| 2 | MARUTHI NAGAR | 0.18 X $10^3$ | 0.17 X $10^3$ |
| 3 | MANALERI | 0.06 X $10^3$ | 0.08 X $10^3$ |
| 4 | KALLENKURICHI | 0.31 X $10^3$ | 0.22 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.12 X $10^3$ | 0.11 X $10^3$ |

**Table 18:Summary of SPM levels in and around ariyalur during April 2006 (mean values)**

| S. No. | AREA | DAY VALUES µg/m³ | NIGHT VALUES µg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.44 X $10^3$ | 0.42 X $10^3$ |
| 2 | MARUTHI NAGAR | 0.49 X $10^3$ | 0.45 X $10^3$ |
| 3 | MANALERI | 0.18 X $10^3$ | 0.22 X $10^3$ |
| 4 | KALLENKURICHI | 0.72 X $10^3$ | 0.58 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.21 X $10^3$ | 0.18 X $10^3$ |

**Table19: Summary of RPM levels in and around ariyalur during May 2006 (mean values)**

| S. No. | AREA | DAY VALUES µg/m³ | NIGHT VALUES µg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.41 X $10^3$ | 0.36 X $10^3$ |
| 2 | MARUTHI NAGAR | 0.38 X $10^3$ | 0.18 X $10^3$ |
| 3 | MANALERI | 0.19 X $10^3$ | 0.28 X $10^3$ |
| 4 | KALLENKURICHI | 0.64 X $10^3$ | 0.48 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.17 X $10^3$ | 0.19 X $10^3$ |

**Table20:Summary of NRPM levels in and around ariyalur during May 2006 (mean values)**

| S. No. | AREA | DAY VALUES µg/m³ | NIGHT VALUES µg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.49 X $10^3$ | 0.42 X $10^3$ |
| 2 | MARUTHI NAGAR | 0.21 X $10^3$ | 0.28 X $10^3$ |
| 3 | MANALERI | 0.23 X $10^3$ | 0.24 X $10^3$ |
| 4 | KALLENKURICHI | 0.52 X $10^3$ | 0.37 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.11 X $10^3$ | 0.14 X $10^3$ |

**Table21:Summary of SPM levels in and around ariyalur during May2006 (mean values)**

| S. No. | AREA | DAY VALUES µg/m³ | NIGHT VALUES µg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.90 X $10^3$ | 0.72 X $10^3$ |
| 2 | MARUTHI NAGAR | 0.59 X $10^3$ | 0.46 X $10^3$ |
| 3 | MANALERI | 0.42 X $10^3$ | 0.52 X $10^3$ |
| 4 | KALLENKURICHI | 1.16 X $10^3$ | 0.85 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.28 X $10^3$ | 0.33 X $10^3$ |

**Table22:Summary of RPM levels in and around ariyalur during June 2006 (mean values)**

| S. No. | AREA | DAY VALUES µg/m³ | NIGHT VALUES µg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.19 X $10^3$ | 0.24 X $10^3$ |
| 2 | MARUTHI NAGAR | 0.28 X $10^3$ | 0.36 X $10^3$ |
| 3 | MANALERI | 0.17 X $10^3$ | 0.13 X $10^3$ |
| 4 | KALLENKURICHI | 0.61 X $10^3$ | 0.54 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.09 X $10^3$ | 0.10 X $10^3$ |

**Table 23:Summary of NRPM levels in and around ariyalur during June 2006 (mean values)**

| S. No. | AREA | DAY VALUES µg/m³ | NIGHT VALUES µg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.17 X $10^3$ | 0.27 X $10^3$ |
| 2 | MARUTHI NAGAR | 0.31 X $10^3$ | 0.42 X $10^3$ |
| 3 | MANALERI | 0.19 X $10^3$ | 0.18 X $10^3$ |
| 4 | KALLENKURICHI | 0.43 X $10^3$ | 0.38 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.11 X $10^3$ | 0.13 X $10^3$ |

**Table 24:Summary of SPM levels in and around ariyalur during June 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.36 X 10³ | 0.51 X 10³ |
| 2 | MARUTHI NAGAR | 0.59 X 10³ | 0.78 X 10³ |
| 3 | MANALERI | 0.36 X 10³ | 0.31 X 10³ |
| 4 | KALLENKURICHI | 1.04 X 10³ | 0.92 X 10³ |
| 5 | CHETTITHIRUKONAM | 0.20 X 10³ | 0.23 X 10³ |

**Table25:Summary of RPM levels in and around ariyalur during July 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.24 X 10³ | 0.21 X 10³ |
| 2 | MARUTHI NAGAR | 0.18 X 10³ | 0.32 X 10³ |
| 3 | MANALERI | 0.33 X 10³ | 0.30 X 10³ |
| 4 | KALLENKURICHI | 0.62 X 10³ | 0.58 X 10³ |
| 5 | CHETTITHIRUKONAM | 0.09 X 10³ | 0.11 X 10³ |

**Table26: Summary of NRPM levels in and around ariyalurduring July 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.18 X 10³ | 0.21 X 10³ |
| 2 | MARUTHI NAGAR | 0.11 X 10³ | 0.27 X 10³ |
| 3 | MANALERI | 0.27 X 10³ | 0.24 X 10³ |
| 4 | KALLENKURICHI | 0.69 X 10³ | 0.41 X 10³ |
| 5 | CHETTITHIRUKONAM | 0.17 X 10³ | 0.16 X 10³ |

**Table27:Summary of SPM levels in and around ariyalur during July 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.42 X 10³ | 0.42 X 10³ |
| 2 | MARUTHI NAGAR | 0.29 X 10³ | 0.59 X 10³ |
| 3 | MANALERI | 0.60 X 10³ | 0.54 X 10³ |
| 4 | KALLENKURICHI | 1.31 X 10³ | 0.99 X 10³ |
| 5 | CHETTITHIRUKONAM | 0.26 X 10³ | 0.27 X 10³ |

**Table28:Summary of RPM levels in and around ariyalur during August 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.08 X $10^3$ | 0.09 X $10^3$ |
| 2 | MARUTHI NAGAR | 0.11 X $10^3$ | 0.08 X $10^3$ |
| 3 | MANALERI | 0.17 X $10^3$ | 0.20 X $10^3$ |
| 4 | KALLENKURICHI | 0.33 X $10^3$ | 0.41 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.09 X $10^3$ | 0.11 X $10^3$ |

**Table29:Summary of NRPM levels in and around ariyalur during August 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.11 X $10^3$ | 0.13 X $10^3$ |
| 2 | MARUTHI NAGAR | 0.13 X $10^3$ | 0.09 X $10^3$ |
| 3 | MANALERI | 0.21 X $10^3$ | 0.21 X $10^3$ |
| 4 | KALLENKURICHI | 0.18 X $10^3$ | 0.17 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.07 X $10^3$ | 0.08 X $10^3$ |

**Table30:Summary of SPM levels in and around ariyalur during August 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.19 X $10^3$ | 0.22 X $10^3$ |
| 2 | MARUTHI NAGAR | 0.24 X $10^3$ | 0.17 X $10^3$ |
| 3 | MANALERI | 0.38 X $10^3$ | 0.41 X $10^3$ |
| 4 | KALLENKURICHI | 0.51 X $10^3$ | 0.58 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.16 X $10^3$ | 0.19 X $10^3$ |

**Table 31:Summary of RPM levels in and around ariyalur during September 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.39 X $10^3$ | 0.42 X $10^3$ |
| 2 | MARUTHI NAGAR | 0.81 X $10^3$ | 0.62 X $10^3$ |
| 3 | MANALERI | 0.17 X $10^3$ | 0.21 X $10^3$ |
| 4 | KALLENKURICHI | 0.06 X $10^3$ | 0.09 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.09 X $10^3$ | 0.12 X $10^3$ |

**Table32:Summary of NRPM levels in and around ariyalur during September 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.21 X 10³ | 0.32 X 10³ |
| 2 | MARUTHI NAGAR | 0.42 X 10³ | 0.38 X 10³ |
| 3 | MANALERI | 0.09 X 10³ | 0.08 X 10³ |
| 4 | KALLENKURICHI | 0.02 X 10³ | 0.03 X 10³ |
| 5 | CHETTITHIRUKONAM | 0.03 X 10³ | 0.04 X 10³ |

**Table33:Summary of SPM levels in and around ariyalur during September 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.60 X 10³ | 0.74 X 10³ |
| 2 | MARUTHI NAGAR | 1.23 X 10³ | 1.00X 10³ |
| 3 | MANALERI | 0.26 X 10³ | 0.29 X 10³ |
| 4 | KALLENKURICHI | 0.08X 10³ | 0.12 X 10³ |
| 5 | CHETTITHIRUKONAM | 0.12 X 10³ | 0.16 X 10³ |

**Table 34:Summary of RPM Levels in and around ariyalur during October 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.09 X 10³ | 0.07 X 10³ |
| 2 | MARUTHI NAGAR | 0.32 X 10³ | 0.28 X 10³ |
| 3 | MANALERI | 0.28 X 10³ | 0.19 X 10³ |
| 4 | KALLENKURICHI | 0.07 X 10³ | 0.08 X 10³ |
| 5 | CHETTITHIRUKONAM | 0.06 X 10³ | 0.07 X 10³ |

**Table35: Summary of NRPM levels in and around ariyalur during October 2006 (mean values)**

| S. No. | AREA | DAY VALUES μg/m³ | NIGHT VALUES μg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.11 X 10³ | 0.13 X 10³ |
| 2 | MARUTHI NAGAR | 0.21 X 10³ | 0.25 X 10³ |
| 3 | MANALERI | 0.19 X 10³ | 0.18 X 10³ |
| 4 | KALLENKURICHI | 0.02 X 10³ | 0.03 X 10³ |
| 5 | CHETTITHIRUKONAM | 0.02 X 10³ | 0.02 X 10³ |

**Table36:Summary of SPM levels in and around ariyalur during October 2006 (mean values)**

| S. No. | AREA | DAY VALUES µg/m³ | NIGHT VALUES µg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.20 X $10^3$ | 0.20 X $10^3$ |
| 2 | MARUTHI NAGAR | 0.53 X $10^3$ | 0.53X $10^3$ |
| 3 | MANALERI | 0.47 X $10^3$ | 0.37 X $10^3$ |
| 4 | KALLENKURICHI | 0.09 X $10^3$ | 0.11 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.08 X $10^3$ | 0.09 X $10^3$ |

**Table37:Summary of RPM levels in and around ariyalur during November 2006 (mean values)**

| S. No. | AREA | DAY VALUES µg/m³ | NIGHT VALUES µg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.22 X $10^3$ | 0.20 X $10^3$ |
| 2 | MARUTHI NAGAR | 0.31 X $10^3$ | 0.28 X $10^3$ |
| 3 | MANALERI | 0.41 X $10^3$ | 0.52 X $10^3$ |
| 4 | KALLENKURICHI | 0.19 X $10^3$ | 0.21 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.06 X $10^3$ | 0.07 X $10^3$ |

**Table 38:Summary of NRPM levels in and around ariyalurduring November 2006 (mean values)**

| S. No. | AREA | DAY VALUES µg/m³ | NIGHT VALUES µg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.19 X $10^3$ | 0.17 X $10^3$ |
| 2 | MARUTHI NAGAR | 0.17 X $10^3$ | 0.21 X $10^3$ |
| 3 | MANALERI | 0.21X $10^3$ | 0.18 X $10^3$ |
| 4 | KALLENKURICHI | 0.12 X $10^3$ | 0.11 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.02 X $10^3$ | 0.03 X $10^3$ |

**Table 39:Summary of SPM levels in and around ariyalur during November 2006 (mean values)**

| S. No. | AREA | DAY VALUES µg/m³ | NIGHT VALUES µg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.41 X $10^3$ | 0.37 X $10^3$ |
| 2 | MARUTHI NAGAR | 0.48 X $10^3$ | 0.49 X $10^3$ |
| 3 | MANALERI | 0.62 X $10^3$ | 0.70 X $10^3$ |
| 4 | KALLENKURICHI | 0.31 X $10^3$ | 0.32 X $10^3$ |
| 5 | CHETTITHIRUKONAM | 0.08 X $10^3$ | 0.10 X $10^3$ |

**Table40:Summary of RPM levels in and around ariyalur during December 2006 (mean values)**

| S. No. | AREA | DAY VALUES µg/m³ | NIGHT VALUES µg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.21 X 10³ | 0.20 X 10³ |
| 2 | MARUTHI NAGAR | 0.24 X 10³ | 0.21 X 10³ |
| 3 | MANALERI | 0.35 X 10³ | 0.26 X 10³ |
| 4 | KALLENKURICHI | 0.18 X 10³ | 0.11 X 10³ |
| 5 | CHETTITHIRUKONAM | 0.11 X 10³ | 0.13 X 10³ |

**Table 41: Summary of NRPM levels in and around ariyalur during December 2006 (mean values)**

| S. No. | AREA | DAY VALUES µg/m³ | NIGHT VALUES µg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.17 X 10³ | 0.17 X 10³ |
| 2 | MARUTHI NAGAR | 0.12 X 10³ | 0.06 X 10³ |
| 3 | MANALERI | 0.16 X 10³ | 0.09 X 10³ |
| 4 | KALLENKURICHI | 0.02 X 10³ | 0.05 X 10³ |
| 5 | CHETTITHIRUKONAM | 0.04 X 10³ | 0.03 X 10³ |

**Table 42:Summary of SPM levels in and around ariyalur during December 2006 (mean values)**

| S. No. | AREA | DAY VALUES µg/m³ | NIGHT VALUES µg/m³ |
|---|---|---|---|
| 1 | ANNA NAGAR | 0.38 X 10³ | 0.37 X 10³ |
| 2 | MARUTHI NAGAR | 0.36 X 10³ | 0.27 X 10³ |
| 3 | MANALERI | 0.51 X 10³ | 0.35 X 10³ |
| 4 | KALLENKURICHI | 0.20 X 10³ | 0.16 X 10³ |
| 5 | CHETTITHIRUKONAM | 0.15 X 10³ | 0.16 X 10³ |

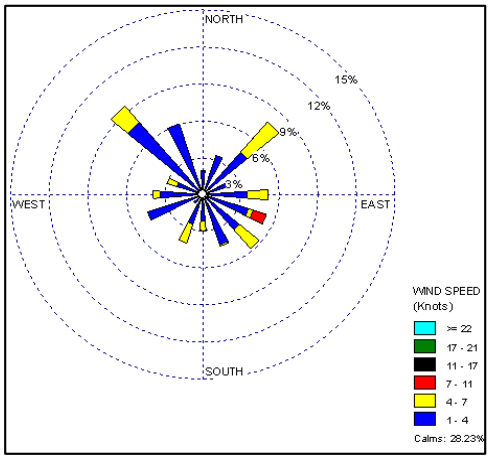


**Fig. 1: Wind – rose constructed for the period of January 2002 - December 2002**

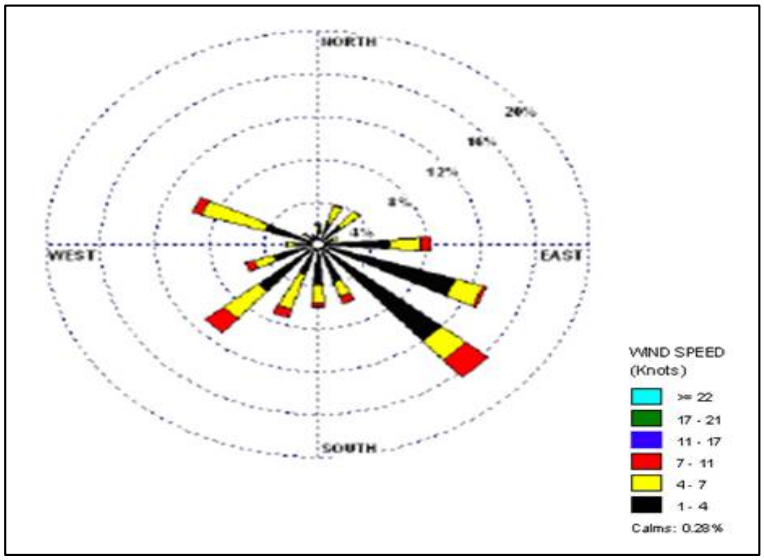


**Fig. 2: Wind – rose constructed for the period of January 2003 - December 2003**

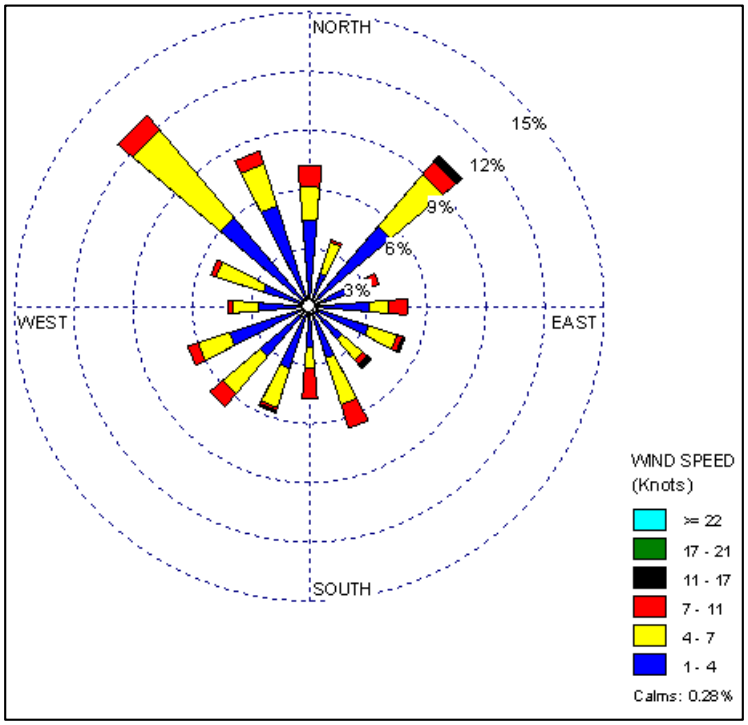


**Fig. 3: Wind - rose constructed for the period of January 2004 - December 2004**

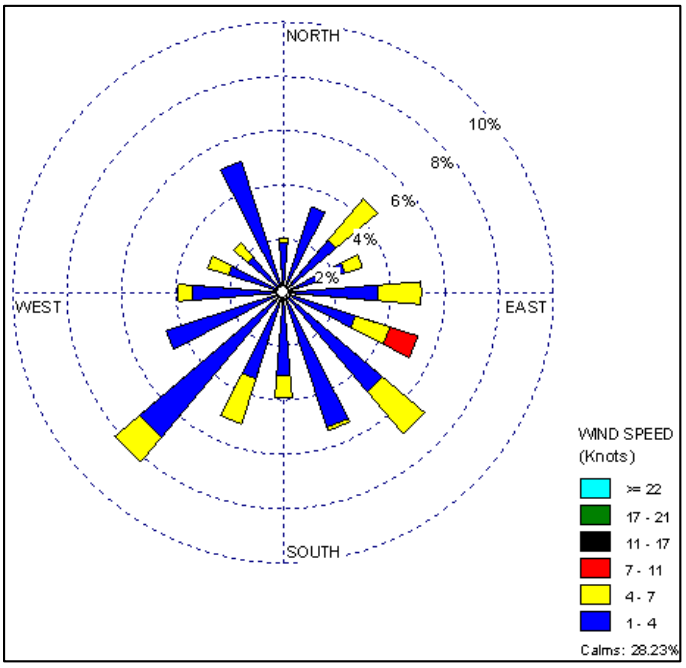


**Fig. 4: wind - rose constructed for the period of January 2005 - December 2005**

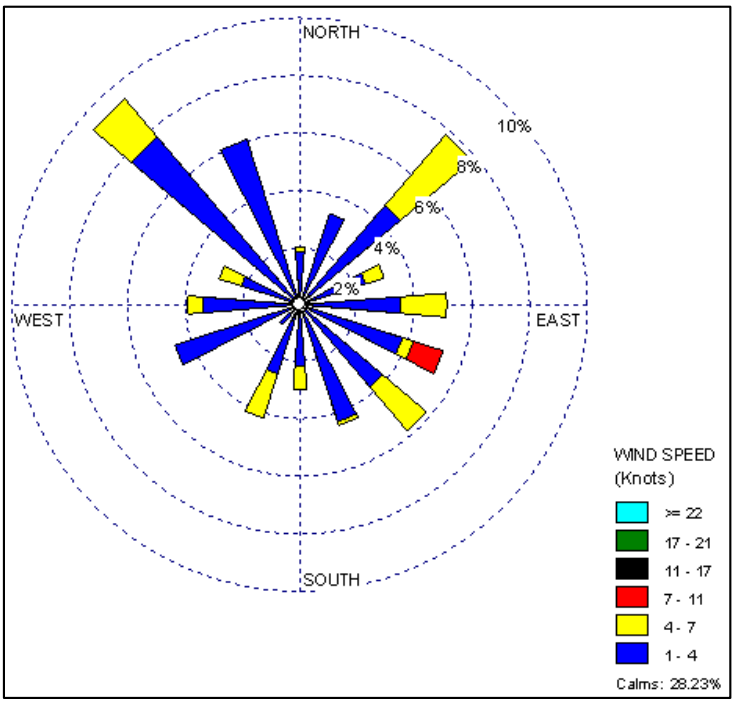


**Fig .5: wind - rose constructed for the period of January 2006 - December 2006**

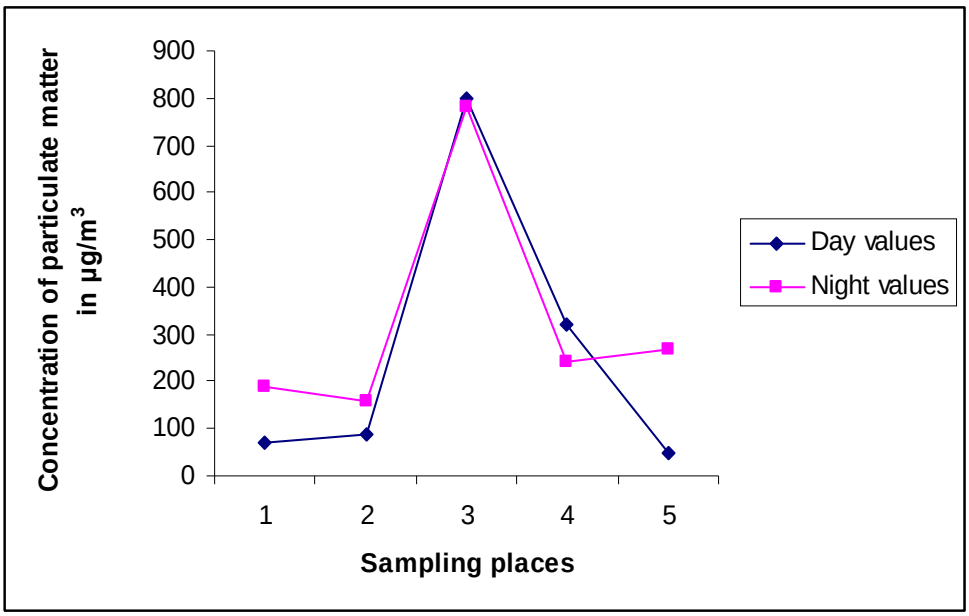


**Fig .6: Diagrammatic representation of RPM levels of different sampling stations during January 2006**

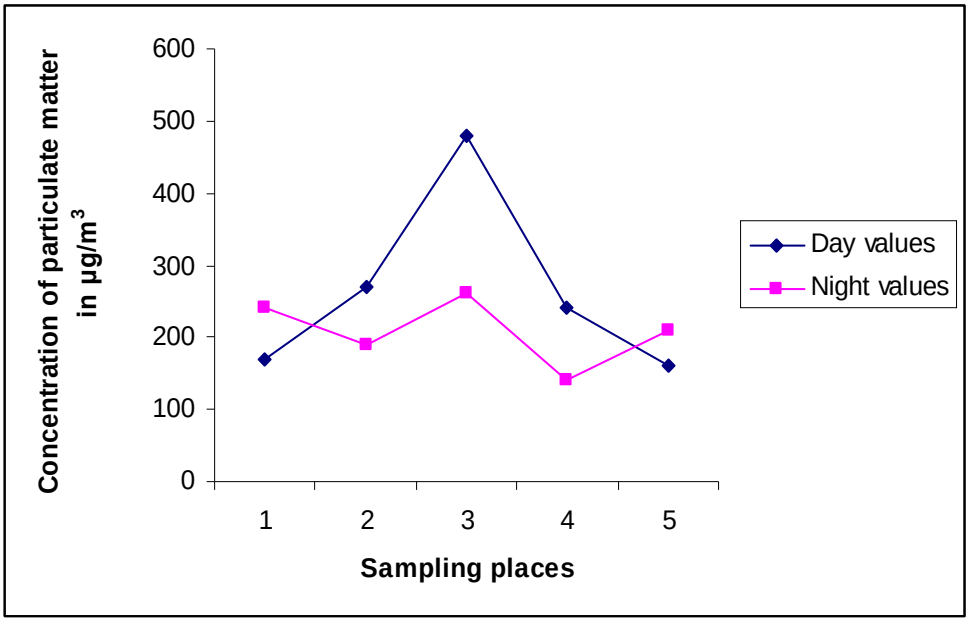


**Fig. 7: Diagrammatic representation of NRPM levels of different sampling stations during January 2006**

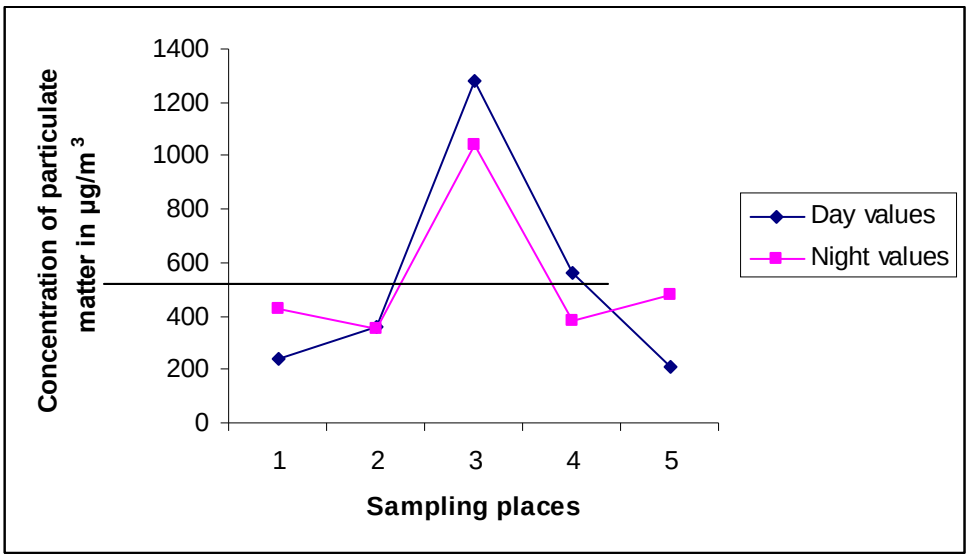


**Fig.8: Diagrammatic representation of SPM levels of different sampling stations during January 2006**

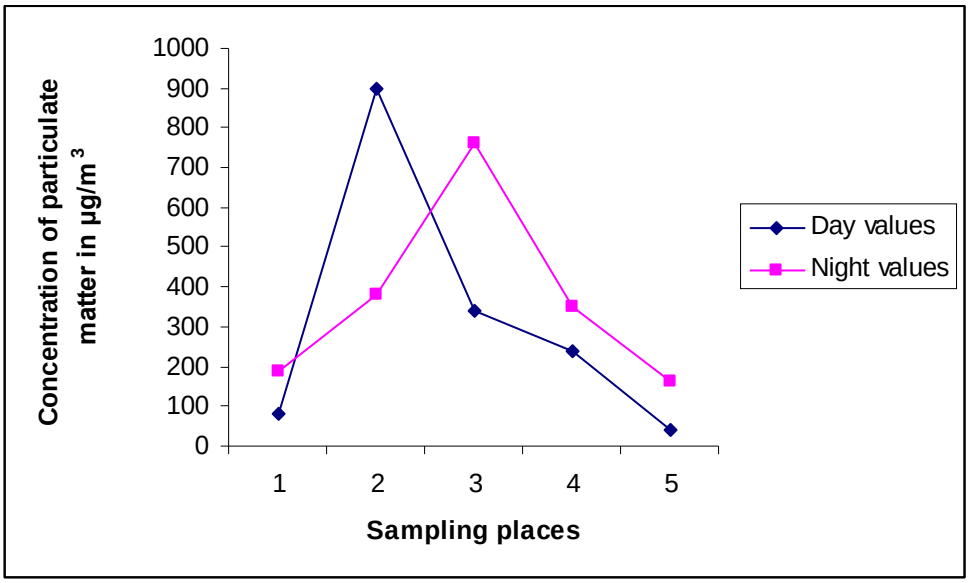


**Fig. 9: Diagrammatic representation of RPM levels of different sampling stations during February2006**

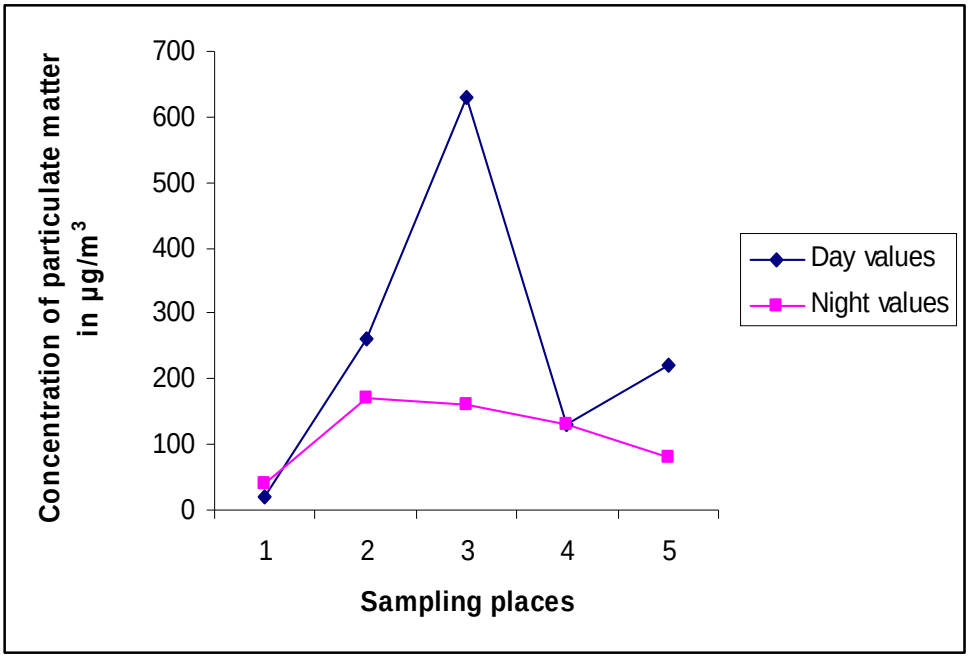


**Fig. 10: Diagrammatic representation of NRPM levels of different sampling stations during February2006**

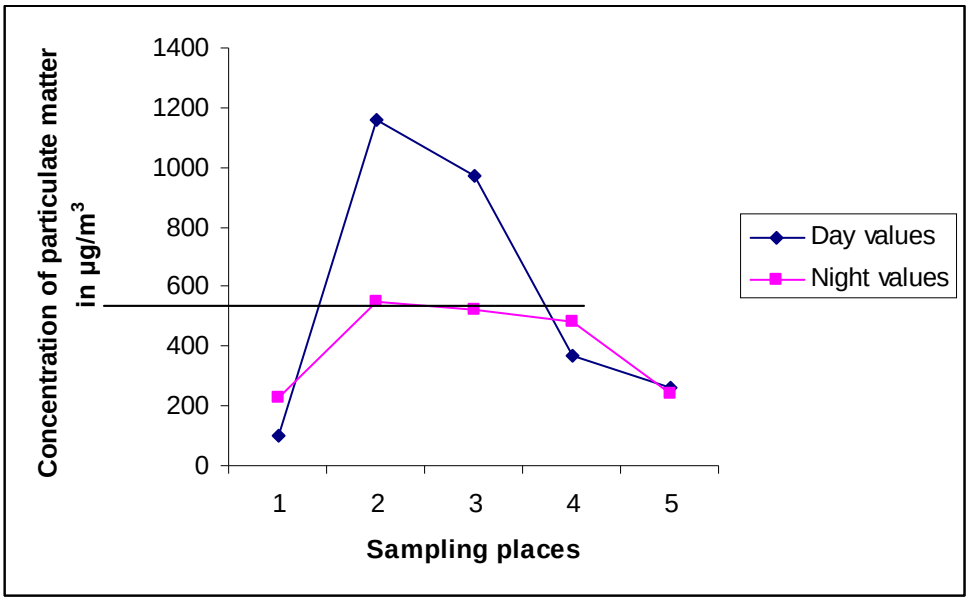


**Fig.11: Diagrammatic representation of SPM levels of different sampling stations during February2006**

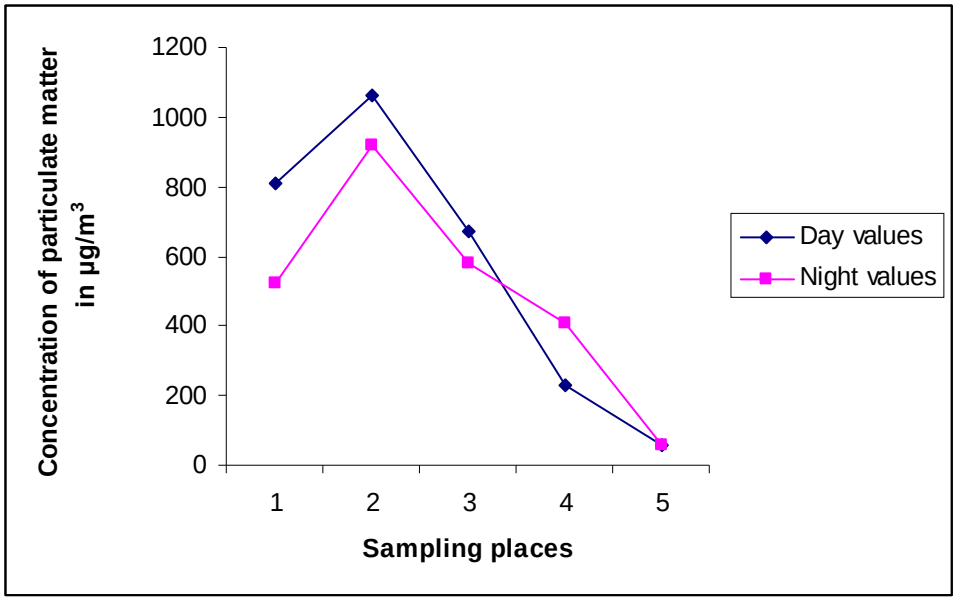


**Fig 12: Diagrammatic representation of RPM levels of different sampling stations during March 2006**

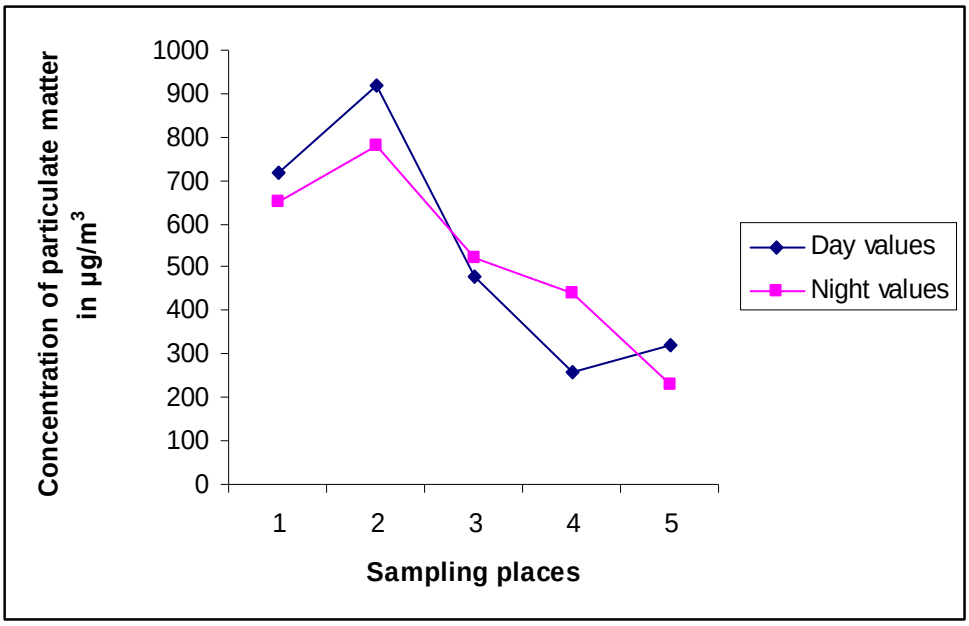


**Fig .13: Diagrammatic representation of NRPM levels of different sampling stations during March 2006**

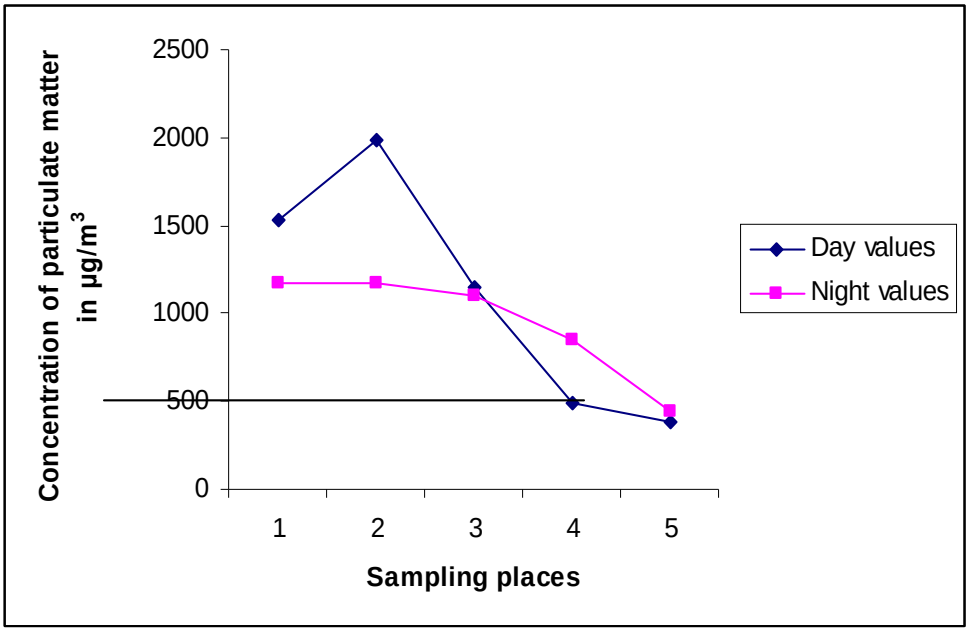


**Fig. 14: Diagrammatic representation of SPM levels of different sampling stations during March 2006**

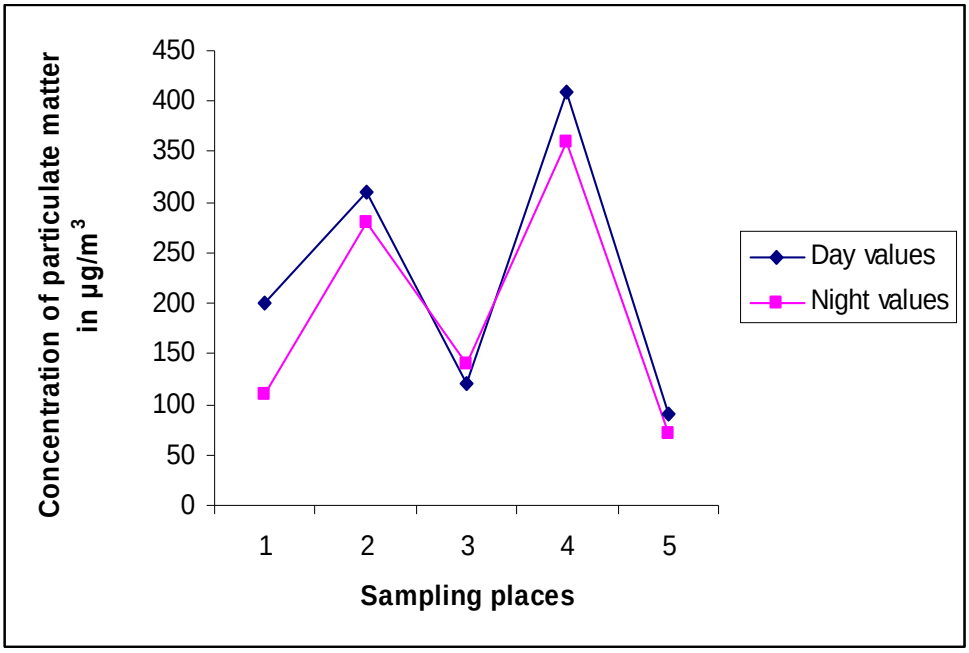


**Fig. 15: Diagrammatic representation of RPM levels of different sampling stations during April 2006**

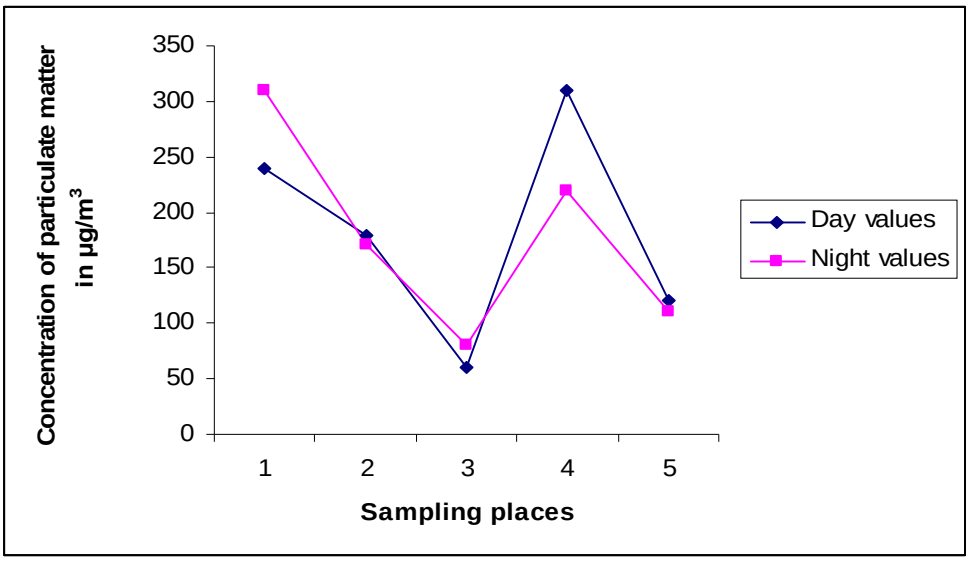


**Fig .16 : Diagrammatic representation of NRPM levels of different sampling stations during April 2006**

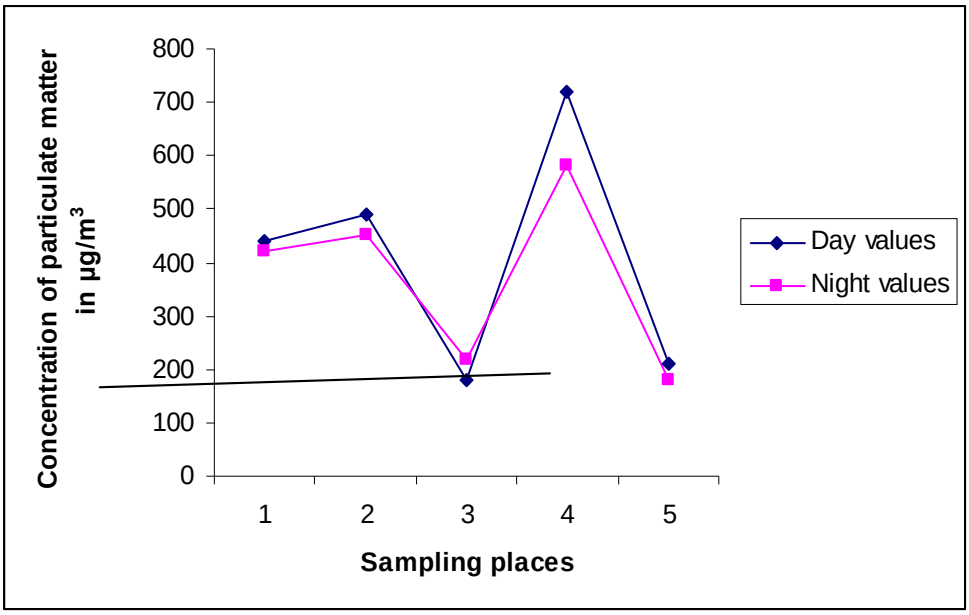


**Fig. 17: Diagrammatic representation of SPM levels of different sampling stations during April 2006**

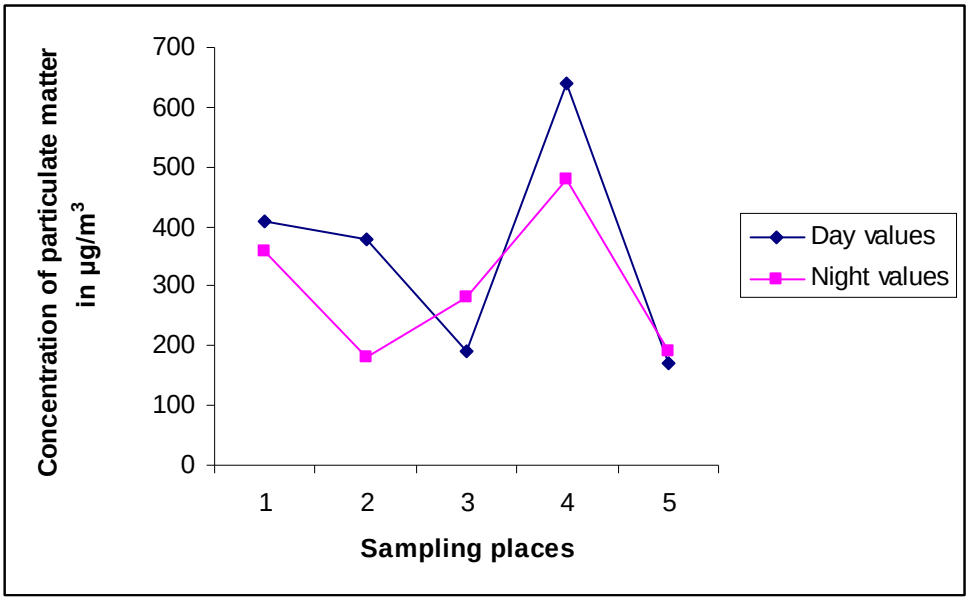


**Fig. 18: Diagrammatic representation of RPM levels of different sampling stations during May 2006**

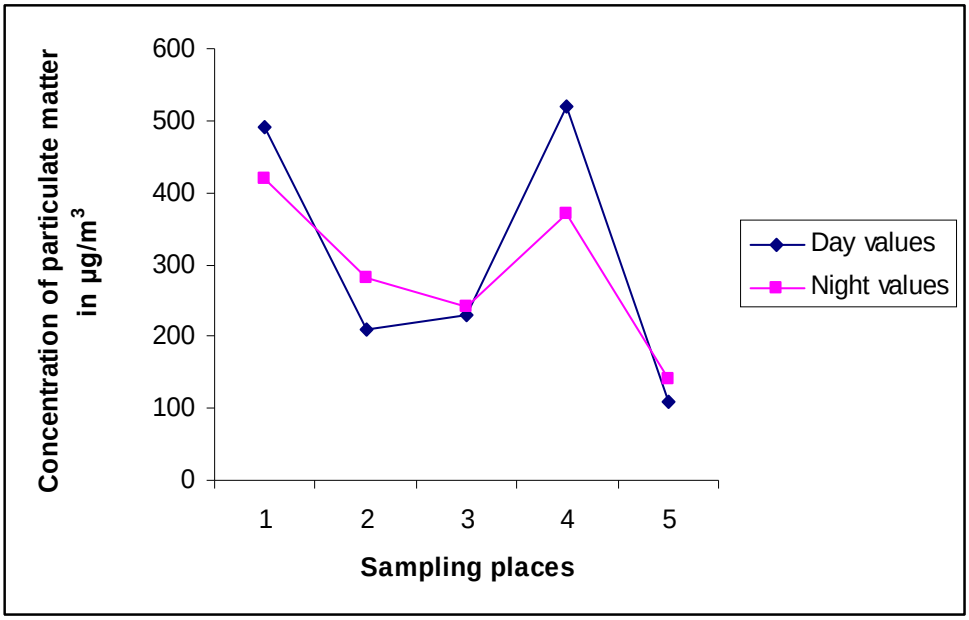


**Fig .19: Diagrammatic representation of NRPM levels of different sampling stations during May 2006**

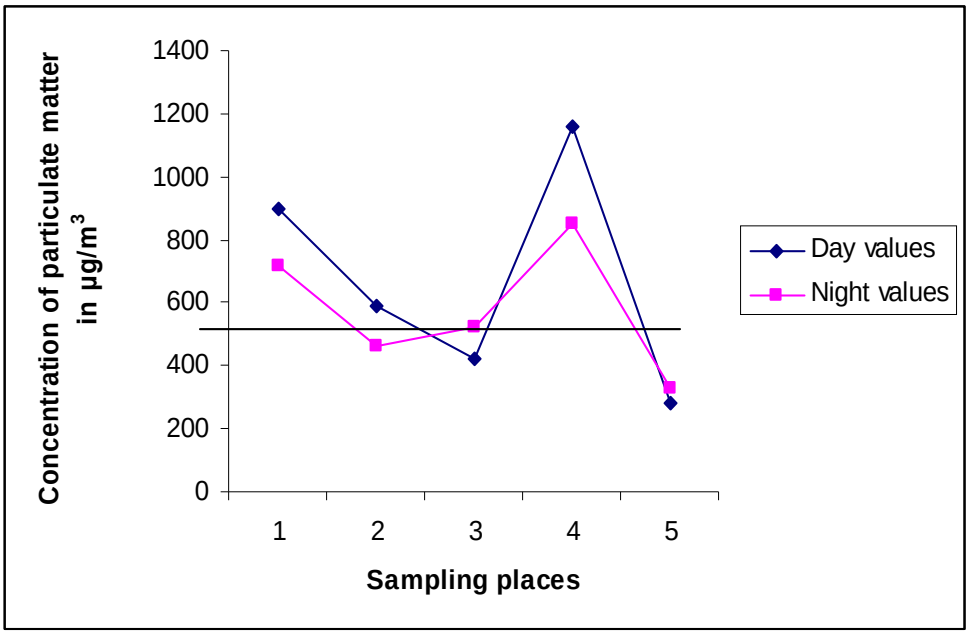


**Fig. 20: Diagrammatic representation of SPM levels of different sampling stations during May 2006**

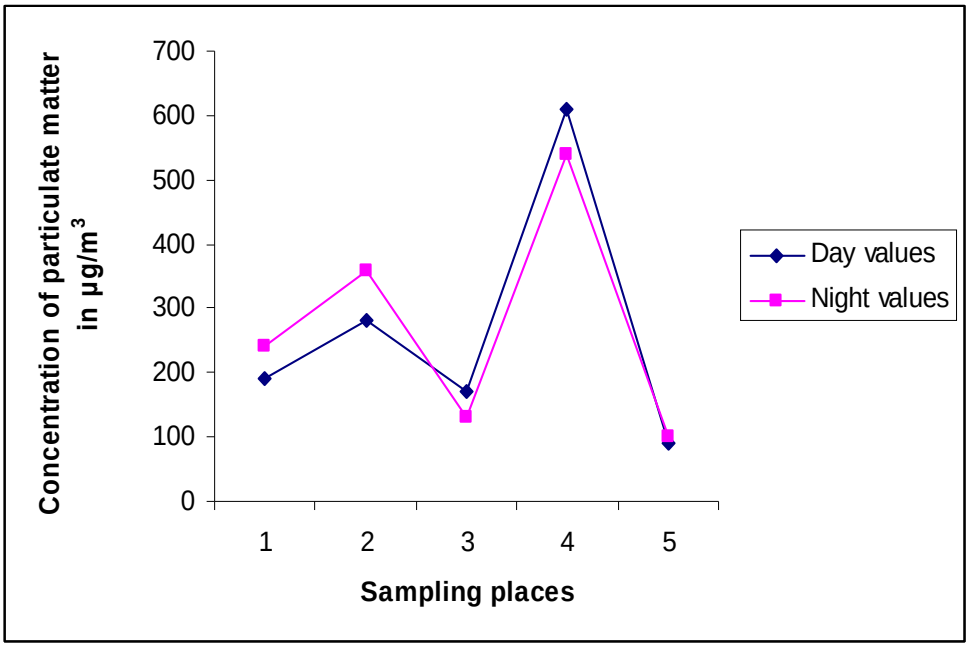


**Fig. 21: Diagrammatic representation of RPM levels of different sampling stations during June 2006**

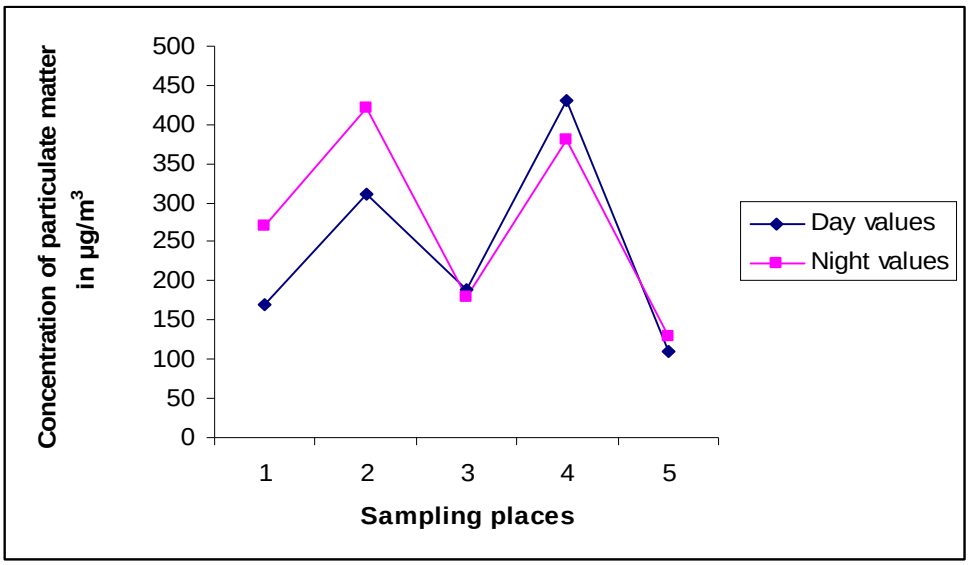


**Fig. 22 :Diagrammatic representation of NRPM levels of different sampling stations during June 2006**

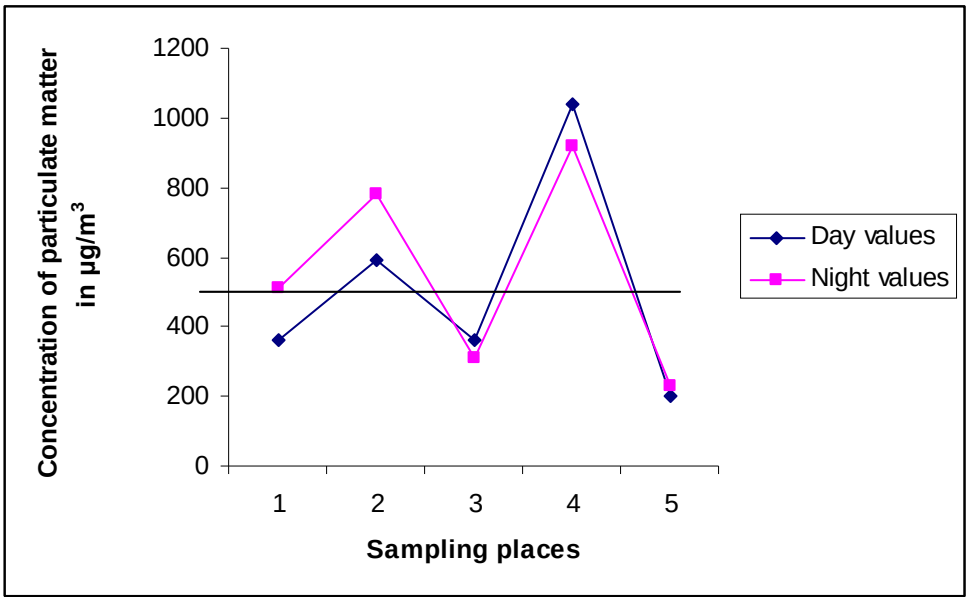


**Fig. 23: Diagrammatic representation of SPM levels of different sampling stations during June 2006**

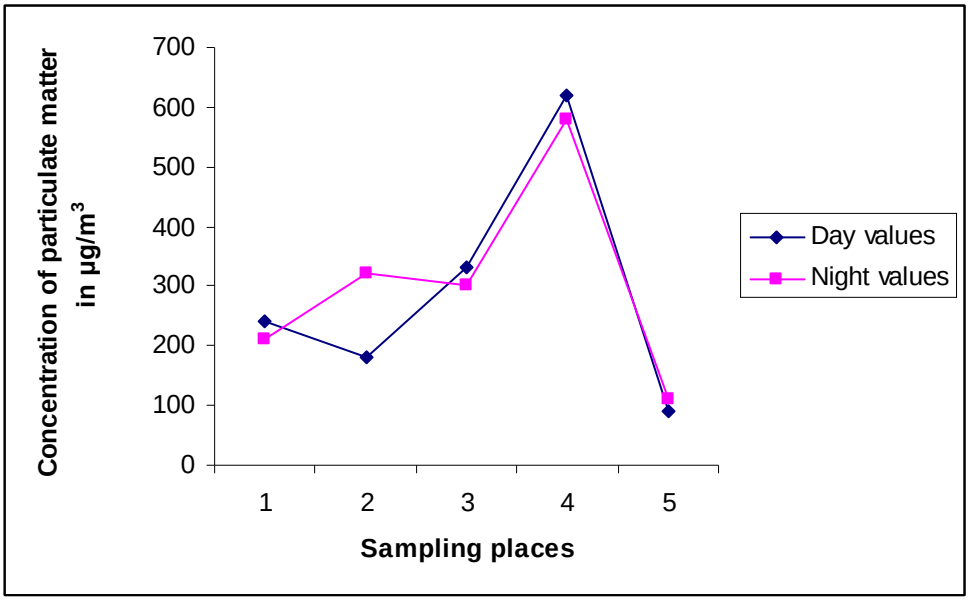


**Fig .24: Diagrammatic representation of RPM levels of different sampling stations during July 2006**

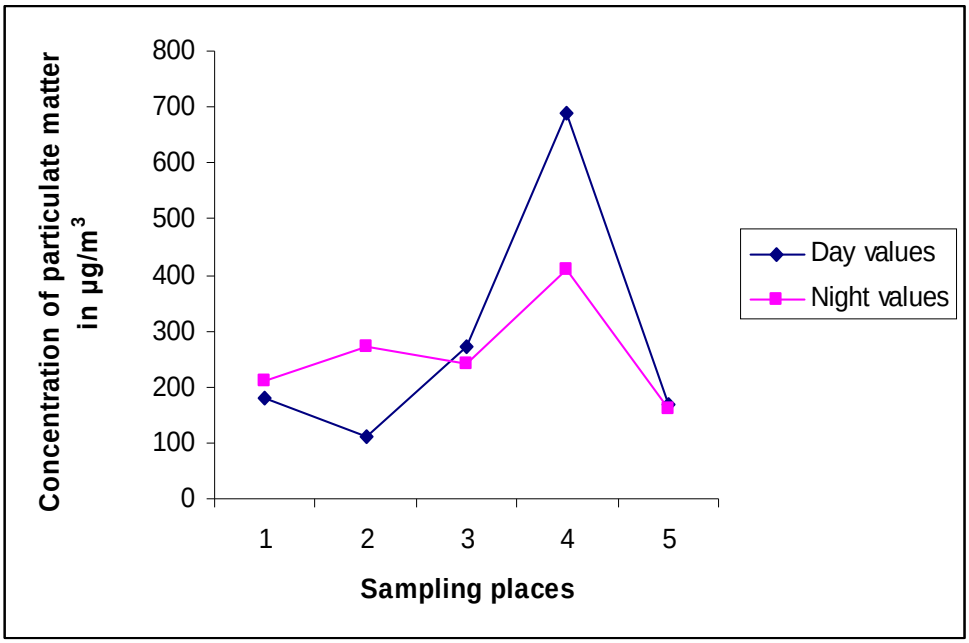


**Fig .25 :Diagrammatic representation of NRPM levels of different sampling stations during July 2006**

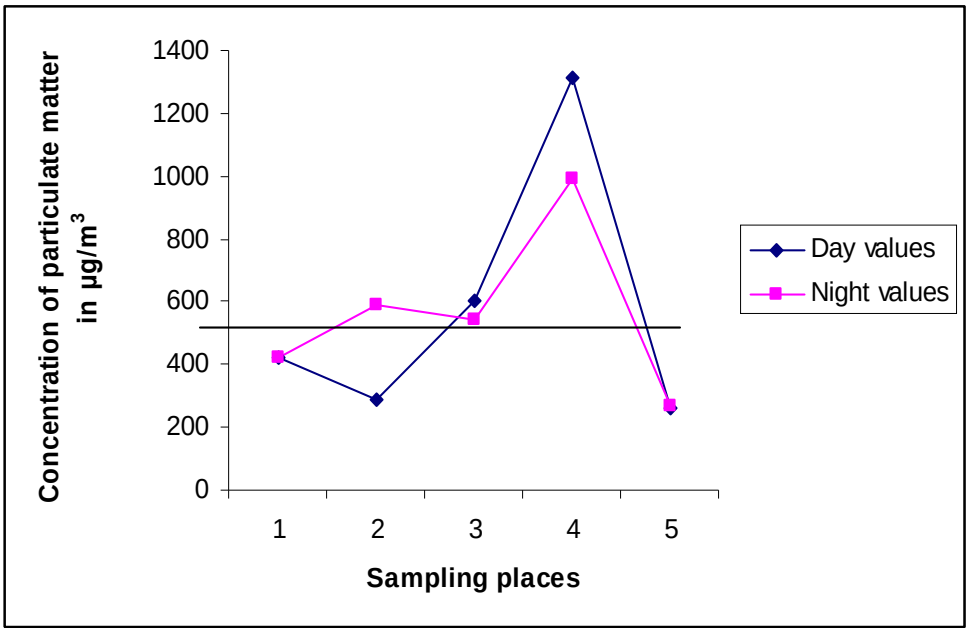


**Fig. 26: Diagrammatic representation of SPM levels of different sampling stations during July 2006**

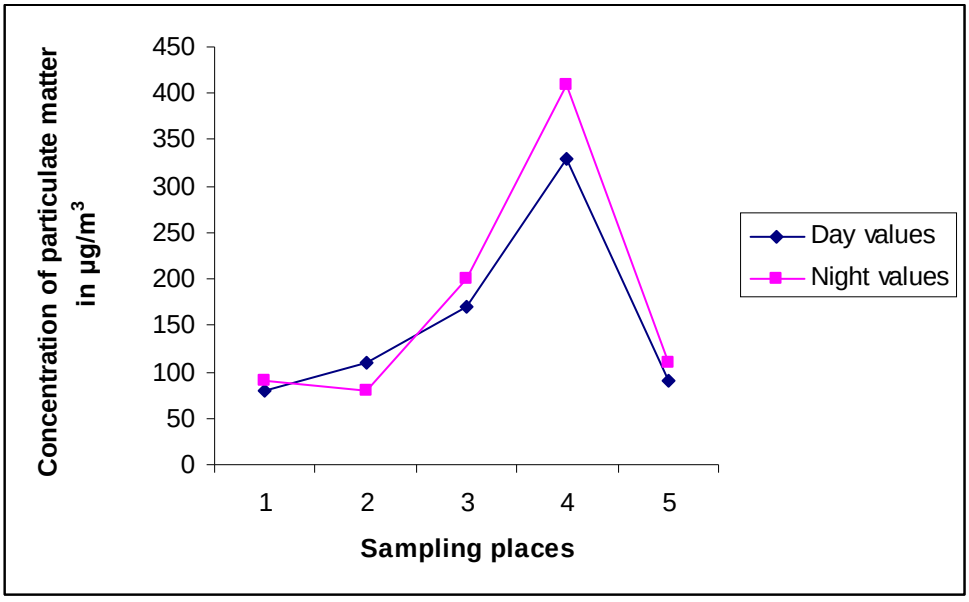


**Fig.27: Diagrammatic representation of RPM levels of different sampling stations during August 2006**

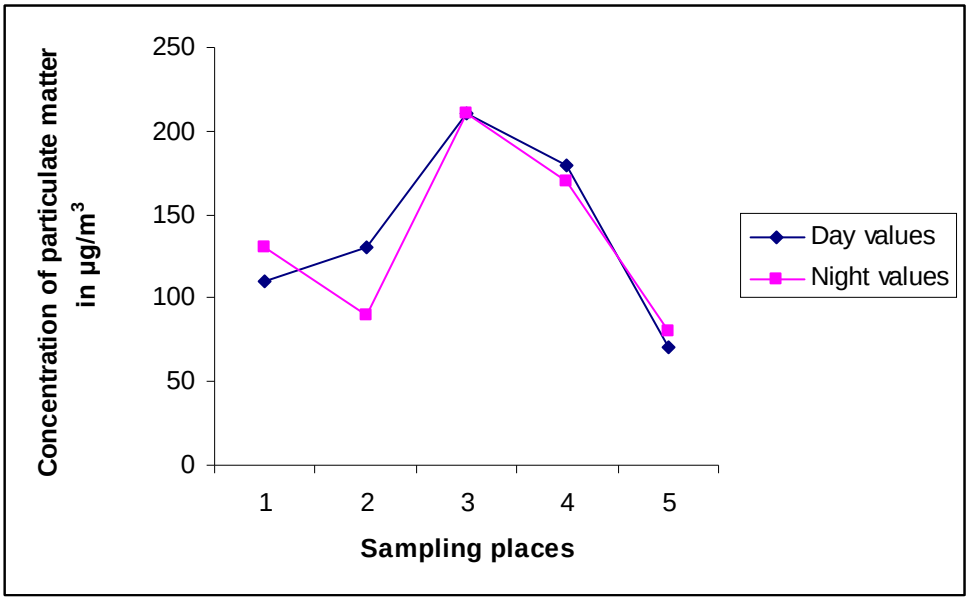


**Fig. 28: Diagrammatic representation of NRPM levels of different sampling stations during August 2006**

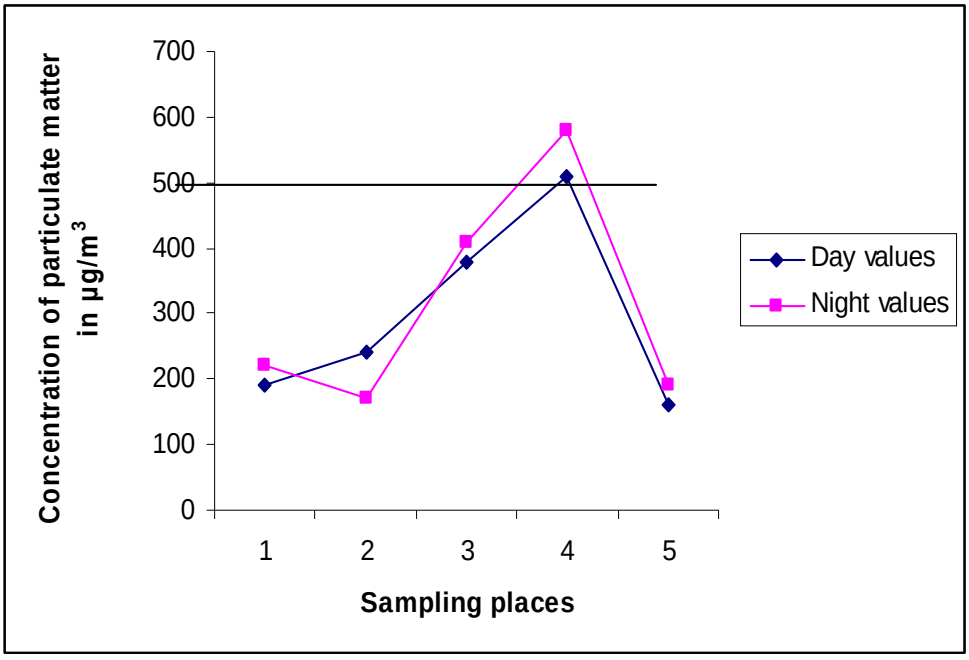


**Fig. 29: Diagrammatic representation of SPM levels of different sampling stations during August 2006**

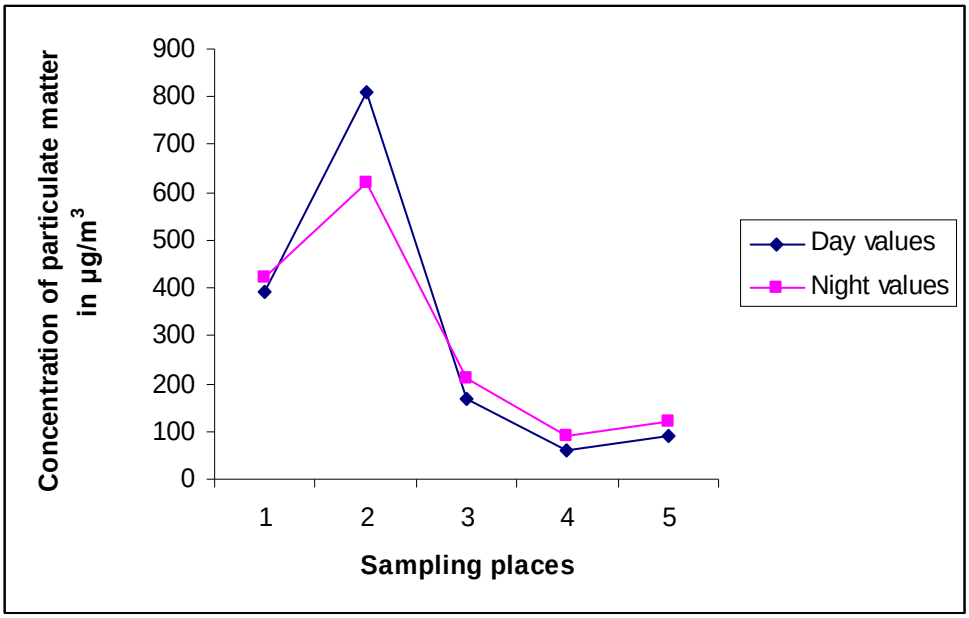


**Fig. 30: Diagrammatic representation of RPM levels of different sampling stations during September 2006**

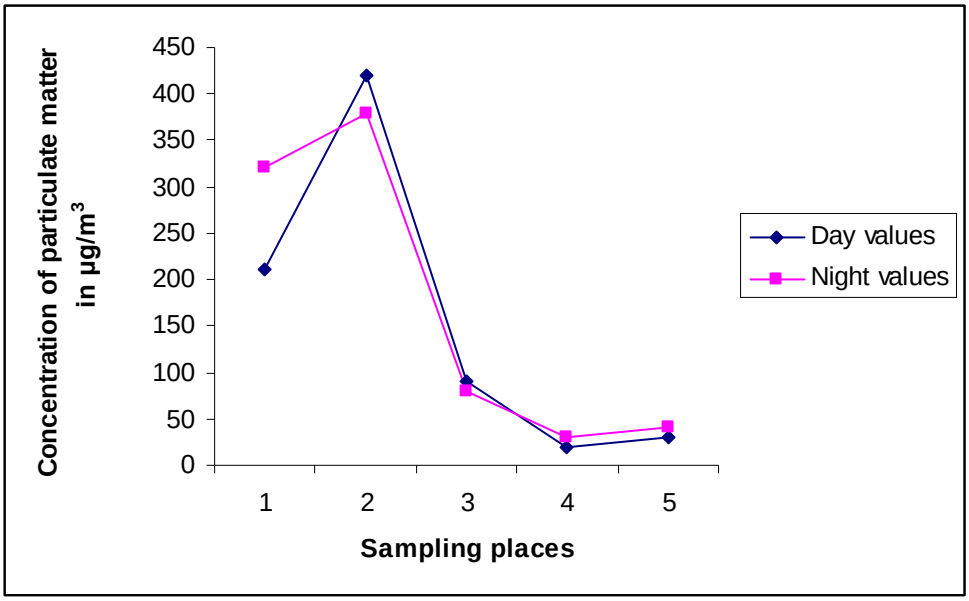


**Fig.31: Diagrammatic representation of NRPM levels of different sampling stations during September 2006**

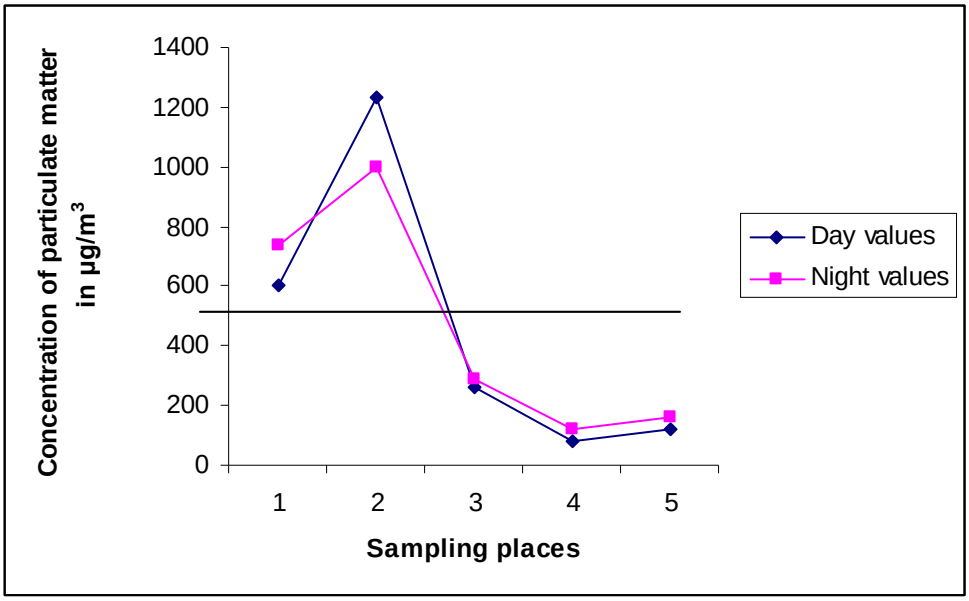


**Fig.32: Diagrammatic representation of SPM levels of different sampling stations during September 2006**

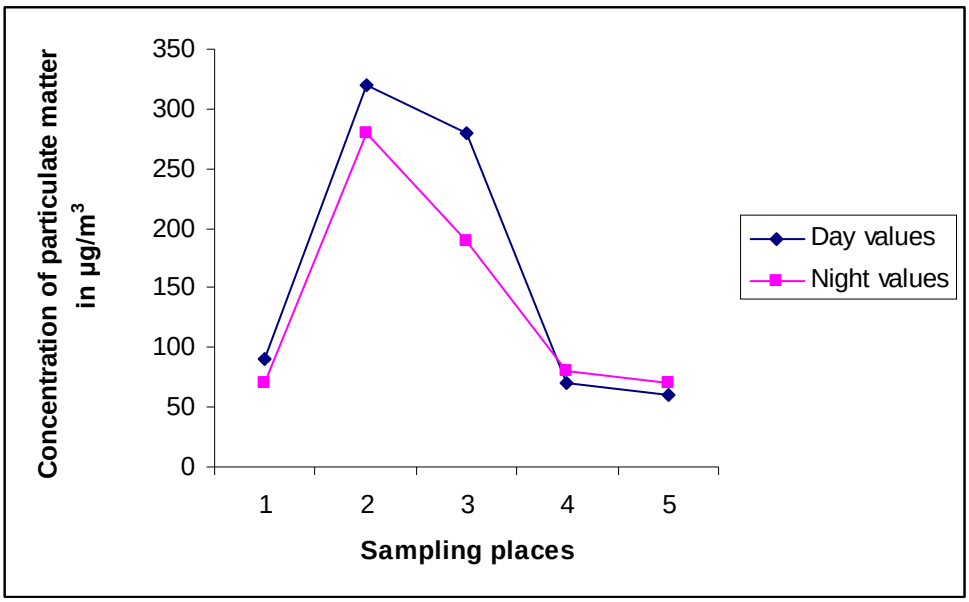


**Fig .33: Diagrammatic representation of RPM levels of different sampling stations during October 2006**

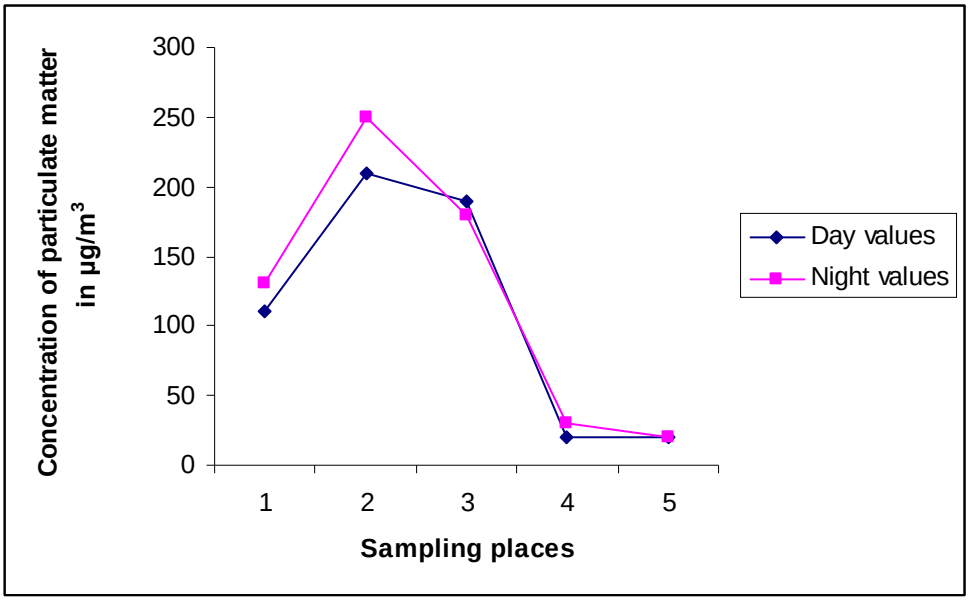


**Fig. 34: Diagrammatic representation of NRPM levels of different sampling stations during October 2006**

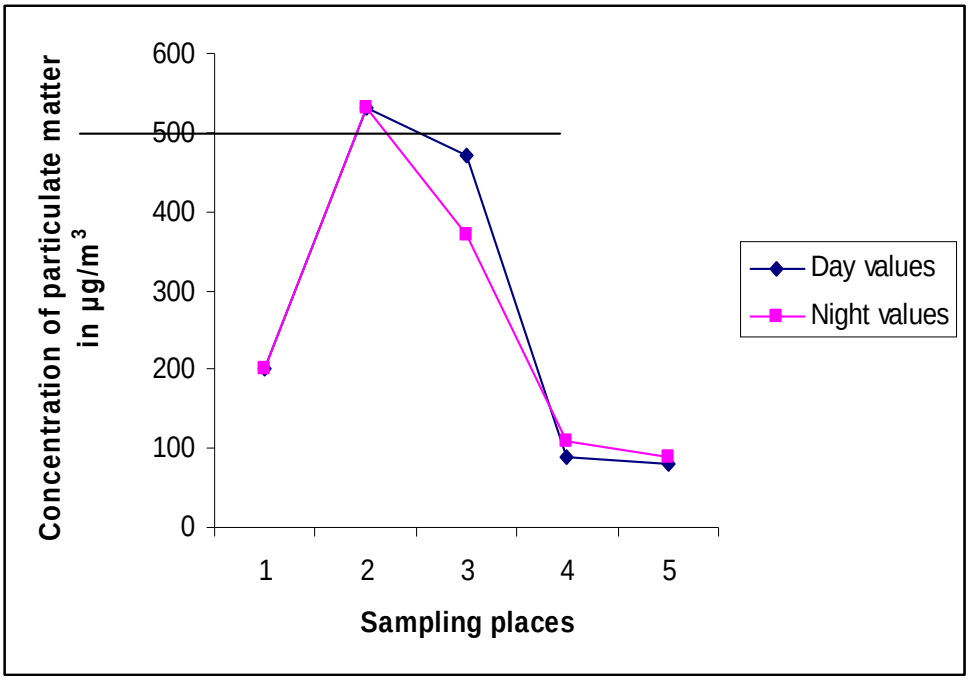


**Fig. 35: Diagrammatic representation of SPM levels of different sampling stations during October 2006**

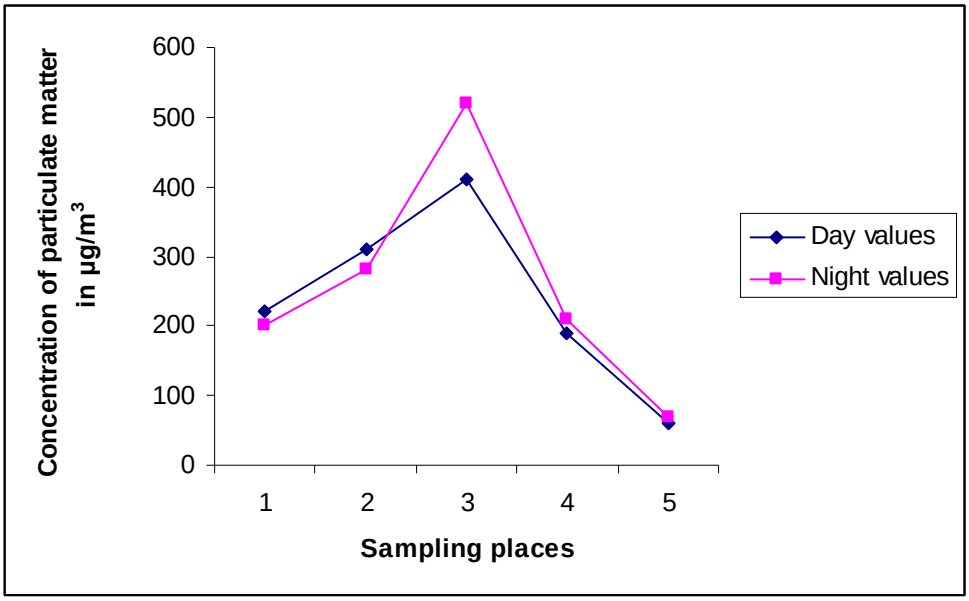


**Fig. 36: Diagrammatic representation of RPM levels of different sampling stations during November 2006**

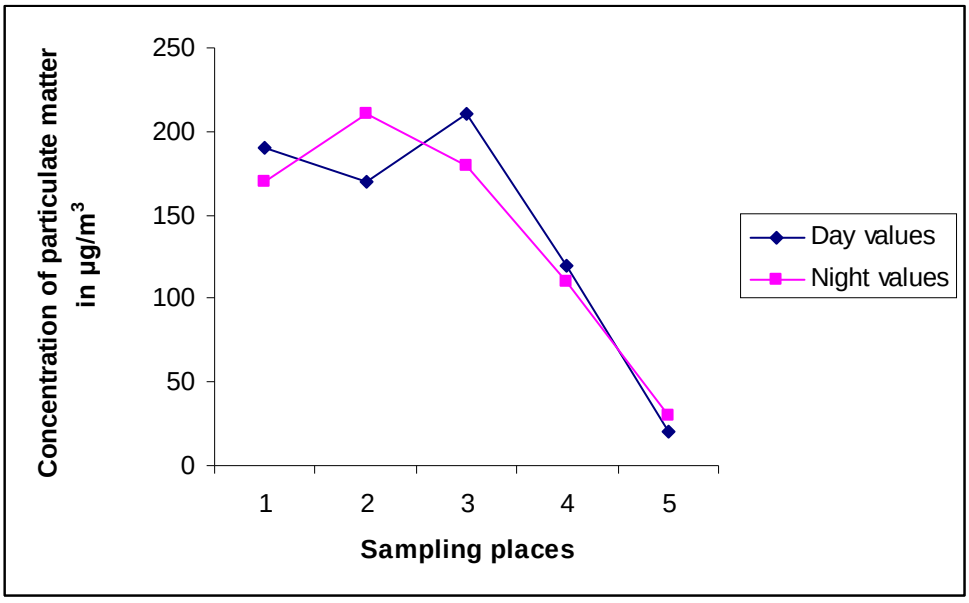


**Fig. 37: Diagrammatic representation of NRPM levels of different sampling stations during November 2006**

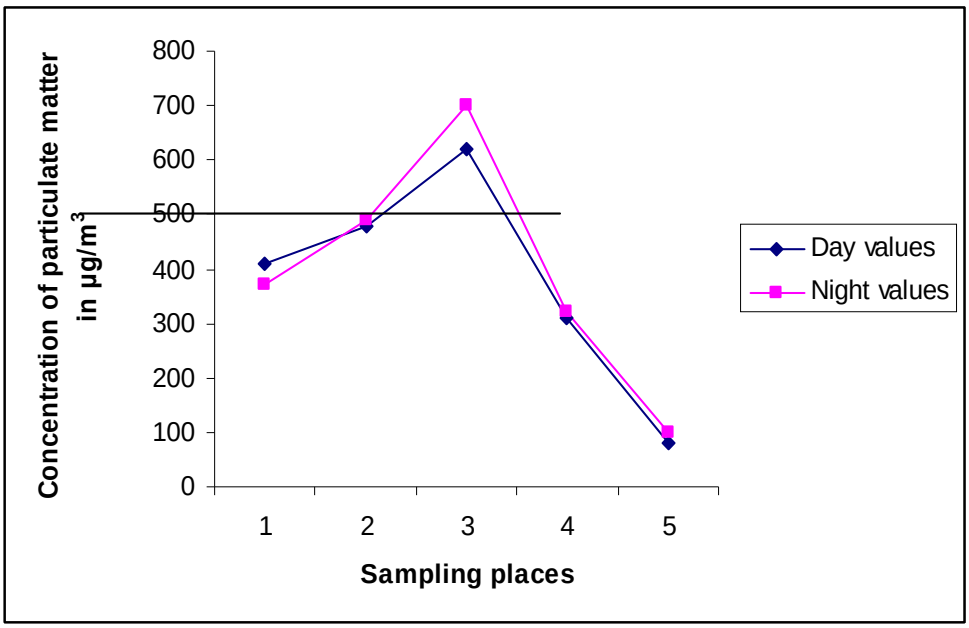


**Fig. 38: Diagrammatic representation of SPM levels of different sampling stations during November 2006**

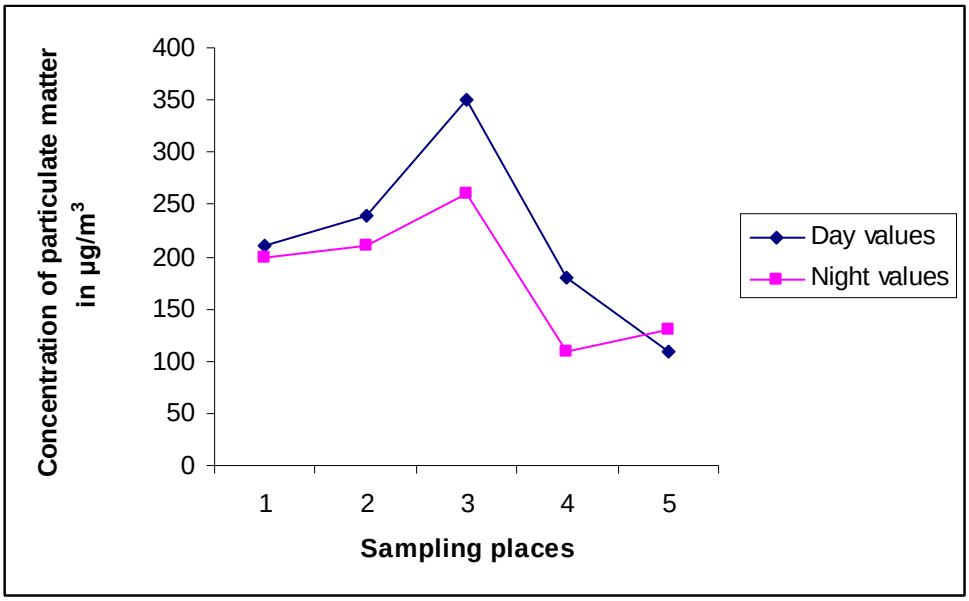


**Fig. 39: Diagrammatic representation of RPM levels of different sampling stations during December 2006**

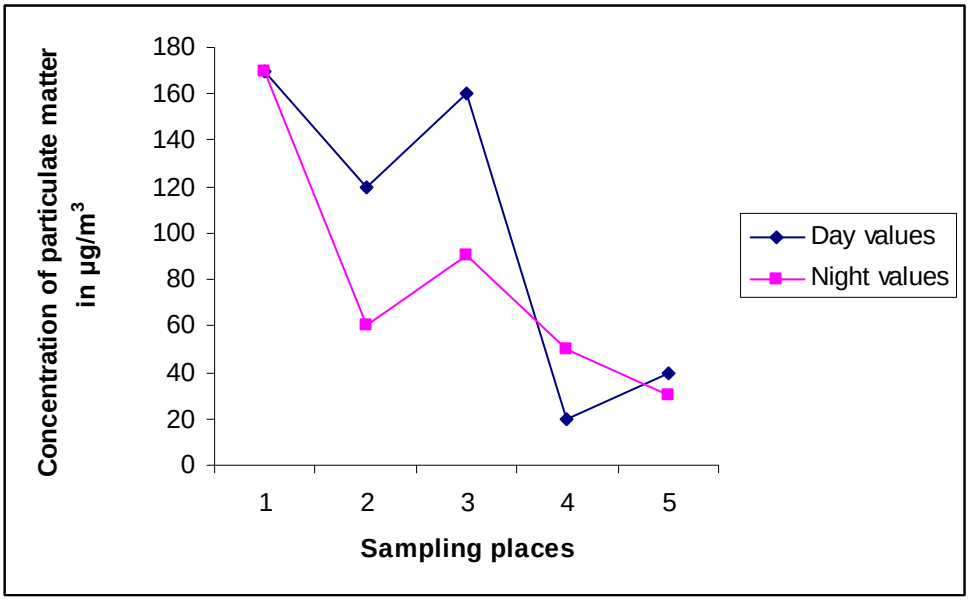


**Fig. 40: Diagrammatic representation of NRPM levels of different sampling stations during December 2006**

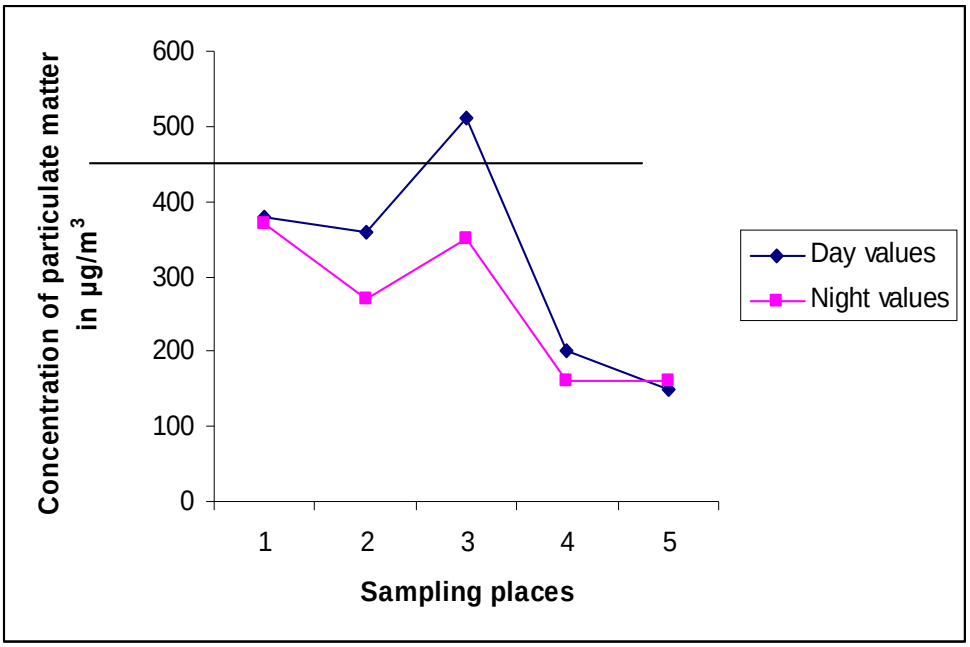


**Fig.41: Diagrammatic representation of SPM levels of different sampling stations during December 2006**

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RESEARCH ARTICLE

# Isolation of *Staphaloccoccus aureus* from House Mice and Study Sensitivity to Antibiotic and Extract of Plants in Iraq

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## ABSTRACT

The aim of study was conducted to isolate *Staphylococcus aureus* from intestine of house mice, one hundred samples of feces were collected from intestine of ( 100 ) house mice during period from (1-1 - 2017 to 1-8-2017)in AL-Huriya city in Baghdad the samples were cultured on selective media incubated at 37 C° for 24hr then gram stain were done, The results showed that (30%) of samples were positive for *S. aureus* ,the sensitivity of these isolates against antibiotics Vancomycin (30mg), Imipenem (10mg), Tetracycline (30mg), Amoxillin Clavulanicacid (30mg) ,Cefixime(5mg) Ciprofloxacin(10mg) ,Azithromycin(15mg) , Levofloxacin (5m) were studied the results showed that *S. aureus* were sensitive to Vancomycin (93.33%)Imipenem, (93.33%) Tetracycline ,(73.33%) Amoxillinclavulanicacid, (60%) Ciprofloxacin, (96.66%) Azithromycin, (83.33%),Levofloxacin,(93.33%) and Cefixime (66.66%) and *S.aureus* were resistant to Vancomycin (3.33%), Imipenem (3.33%) , Tetracycline(23.33%), Amoxillinclavulanicacid (36.66%) ,Azithromycin (13.33%), Levofloxacin (3.33%), and Cefixime (33.33%) while that *S. aureus* were intermediate in(33.33%)to all antibiotic except Cefixime , the sensitivity of *S. aureus* to hot watery extracts of plants in concentration (200mg) which include (Sumac, Pomegranate, *Crocus sativus,* Linum usitatissimum, *Lawsonia inermis)* were range from (23.33%-6.66%) to Linum usitatissimum, (23.33%-3.33%) Pomegranate, (16.66%-33.33%)Sumac, (43.33-3.33) *Lawsonia inermis, and (36.66%-23.33 %*) *Crocus sativus* the present study is the first study in Iraq for isolation *S.aureus* from house mice .

**Keywords:** house mice, bacteria, sensitivity, antibiotic ,plant, extract.

## INTRODUCTION

house mouse are dangerous rodent cause multiple damages to human resources by feeding on crops and stored commodities and by their faecal and urine contamination and colonization of rodents and wild mice with *S. aureus* is of limited scope [1]. *S. aureus* is carried asymptomaticlly on human body and responsible for numerous infections, food poisoning endocarditis, septic arthritis, abscesses, pneumonia and sepsis, has been found in various animal species, which is in close contact with humans [2] it is a opportunistic dangerous pathogen causing bacterial infection community and in hospitals with antibiotic resistant, susceptible to various disinfectants like alcohols, chlorhexidine, formaldehyde ,sodium hypochlorite,iodophors,sodium hypochlorite ,combination of alcohol and iodine. and can also be destroyed by heat [3].Antibiotic therapy involving Ampicillin, Oxacillin or Cephalosporin, has traditionally been used to treat *S.aurues* but the recent emergence of bacterial resistance to these antibiotics has lessened their effectiveness, Bacteria may become resistant by antibiotic , metabolites of plant like polypeptide, flavones,quinines,lectins,alkaloids,, tannins, coumarin, terpenoids, and flavonoids have potential antimicrobials to bacteria,and the role of plants metabolite against bacteria play effective effect in resistant bacteria [4] Return to nature is becoming important idea, because of increasing side effects of drugs. The variation in phytochemicals compounds in plants lead to use this materials in different application, like industrial, economic and medical application ,in developing countries the used plants for treat different diseases instead of chemical drugs [5] Sumac (*Rhus typhina*.) is aperennial belongs to the genus Rhus.it used for food and medicinal purposes,it has came originally from united states ,Canada and northern hemispheres [6]. Pomegranates belong to *Punicaceae* family and are the used as food and treatment, The crude extracts of plant leaves for the phytochemical and medical purposes have been tested frequently has a vital role in the prevention of cancer, viral diseases, diabetes**,**bacterial infections, ultraviolet radiation-induced skin damage and infant brain ischemia due to synthesizing putative active molecules such as Gallic acid, Ellagic acid, Punicalin, Punicalagin, Anthocyanins, and Flavanols compounds [7]. *Crocus sativus*. is member of the Iridaceae family, commonly known as saffron, is a perennial stemless herb of the Iridaceae family it is found in france,Greece,Italy,S outhern Europe, Iran, Spain, Morocco, Egypt, Azerbaiijan, .India,turkey ,it contain about 150 non volatile and volatile compounds and the studies had indicated that saffron has anti genotoxic ,anti-ersclerotic ,anti inflammatory,,antitussive and cytotoxic activites uantispasmodic ,aphrodisiac, sedative, stomachic, expectorant, emmenagogue [8] Linum usitatissimum is a functional food has polysaccharide ,lignan, and fatty acid shows evidence of digestibility, bioactive peptides, antimicrobial, anti-Parkinson's, anti-proliferative, antihypertensive, anticancer, immune enhancing, antiulcer and antioxidant activities, [9] *Lawsonia inermis* (Sudanese Henna) a perennial plant had antimicrobial agents antibacterial, antiviral, antimycotic and antiparasitic activities ,dyeing properties, and the presences of hypericin ,quinine,and anthraquninone had antimicrobial activity against methicillin -sensitive.and methicillin resistant Staphylococcus and can used in treatment [10].

## MATERIALS AND METHODS

### Feces Sampling

One hundred mice had been collected in Al- huria city.The samples of feces were taken from intestine of mice **.**Identification of *Staphalococus aureus* Cultural identification. The samples of feces were diluted inPBS and were inoculated in nutrient broth at37 C° for 24 hr ,aloopful from broth cultured on blood agar,incubation at 37 C° for 24h Mannnitol Salt Agar at 37 C° for 24h, the growth of yellow colonies and hemolysis give indication for positive result , and gram staining was done, coagulase and catalase tests for as descibed [11].

### Antibiotics susceptibility test

To study the susceptibility of the isolates staph against eight antibiotics include Imipenem (10mg) ,tetracycline (30mg), Amoxillinclavulanicacid (30mg), Vancomycin (30mg Cefixime (5mg) ,Ciprofloxacin (10mg), Azithromycin (15mg), Levofloxacin (5mg) the trial was accomplished using a disc diffusion method on Mueller hinton Agar according to, measurement of diameter of the inhibition zone12].

### Plants extract preparation

The plant materials *Lawsonia inermis, Linum usitatissimum*, *Crocus sativus*, Pomegranate ,Sumac used in this study was collected from local marked and verified by Iraq national herb agricultural minisitry. Extraction of plants were first dried and ground to powder,then( 50)gm of plant soaked in 250 ml of hot disteled water, The mixture is uniformed by electric blender for 30 minutes in 24hr at room temperature then separated in centrifuge for 3000 cycle in 15 min then the solution filter by whatman No.1 then the solution deried in Rotary vacuum evaporator in temp 40 c then dried in incubator for 3-4 days then we prepare stock solution from the extract in concentration ( 200mg) for ml then filterd in mellipor filter 0.2 according to the methods [13].

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## RESULTS AND DISCUSSION

House mice play an important roles in bacterial infection in human and *S. aureus* is a apportunisic pathogen in mice, the widely distributed of house mice is in Al- Huriya city in Baghdad may lead to infect human with different type of bacteria like *S. aureus* ,in our study results of bacterial isolation of (100) samples of feces from house mice showed that(30%) were belong to *S. aureus* these results agree with the results of other study that (15.3%) of S.aureus in wild rodent and demonstrated that shrews and rodents had show the signs of adaptation for S. aureus [14]. In study reported an inC57BL/6J mice Several faecal species were recorded including S. aureus (¼ 6), *S. aureus* bacteria,[ 15] In anthor study demonstrated off spring retrospective data on the S. aureus prevalence [16 ]also other study confirm that *S. aureus* is considered an opportunistic pathogen in mice[17]. In this study the results showed that. *S.aureus* were sensitive to vancomycin (93.33%) ,(93.33% )Imipenem ,(73.33%) Tetracycline ,(60% )Amoxillin ,(96.66%) Ciprofloxacin ,(83.33% )Azithromycin,(93.33%)Levofloxacin,(66.66% ) cefixime and that S.aureus were resistant (3.33%) vancomycin,(3.33%) Imipenem,(23.33%) tetracycline ,(36.66%) Amoxillinclavulanic ,(13.33%) Azithromycin,(3.33%) Levofloxacin ,(33.33%) Cefixime while that S.aureus were intermediate in (33.33%) to all antibiotic except Cefixime Table[1].

Respectively in our study, these findings correlate with other studies where these antibiotics had high sensitivity [18,19]. in their guidelines for prophylaxis and treatment of MRSA recommended use of Tetracyclines for skin and soft tissue infections[20]. In other study showed sensitivity of peniciilin and Amoxicillin-clavulanate to be 7.1% and 63% [21]. The antimicrobial sensitivity was was found in 16 strains of S.aureus resistance to Ampicillin (69.5%),and susceptible to Vancomycin [22].

Also other study high level of resistance to Amoxacillin (80%), penicillin (87%), Oxacillin (87%), and were susceptible to Vancomycin (100%).Ceftriaxone (97%), Ciprofloxacin (91%) [23] the sensitivity of *S.aureus* to hot watery extracts of plants (sumac, Pomegranate*, Crocus sativus,* Linum usitatissimum*, Lawsonia inermis* were range from (23.33%-6.66%)to Linum usitatissimum, (23.33%-3.33%) to Pomegranate, (16.66%-33.33%)to sumac, (43.33%-3.33%)to *Lawsonia inermis, (36.66%-26.66%)* Table[2].

Scientific experiments since the late 19th century have documented the antimicrobial properties of some spices, herbs, and their components. [6] the use of natural products as treatment in healing and treatment of various diseases has been on the rise in the last few decades and plants extract containing active antimicrobials compounds [7]. The antimicrobial properties of some spices theirs compounds have been documented, sumac extracts have

possess strong antimicrobial and antioxidant activities [24].other study found The pomegranate extracts effect against *S. aureus* isolates[25]. The antibacterial activity of different extracts of pomegranate fruit against *S. aureus* It has been found that the crude and purified peel extracts have high antibacterial activity, [26,27].Results obtained from their study clearly demonstrate broad spectrum antimicrobial activity of pomegranate against seven species of bacteria, It is proven that all parts of a plant can be used as a starting material for medicinal purposes. Medically useful compounds can be obtained from the peel, seeds, flower, bark, juice and roots of pomegranate. The pomegranate peels with antimicrobial activity against methicillin-resistant*S. aureus* (MRSA) has been confirmed recently, indicating its potential to control microbes. The peels of pomegranate can also be used as an antibacterial agent against dental plaque causing bacteria [28]. Reported the antimicrobial activity of flaxseed extract against*S. aureus*. According to the report. In study of [29]extract flaxseed had higher antimicrobial activity against *S. aureus* among the pathogens tested It was also reported that lignans inhibit microbial growth by destabilizing the cell membranes. [30] The studied extracts of *L. inermis* antibacterial activities against antibacterial activity against *S. aureus*[31] Water extract of *L. inermis* was effective against *S. aureus*,.with inhibition zone it was disagreed with [32] in Sudan who reported the water extract most effective one followed by methanol extract was used agar disc diffusion method while results in Sudia Arabia were found that the aqueous extracthad the best inhibitory zone on 8 out of 9 tested bacteria include *S. aureus,*. The variation in the results of previous and present study may be due to the variation in the method of antibacterial activity of henna[33]. extraction investigate the antibacterial of *Crocus sativus Staphylococci*. The aqueous extract showed maximum inhibition zone against one bacteria *Staphylococci*- and the zone was 11 mm and the reported that *C. sativus* aqueous extracts inhibited S . aureus and the inhibition zone is11mm.[34]Other study reported that *C. sativus* is more for different antimicrobial agents and this is clinically important and the resistance development[35].In our study we find that plant extract was effective against S.aureus in comparism with antibiotic. We concluded that house mice act as reservoir for *S. aureus* and the hot watery extract of plants can be used for inhibit growth of *S. aureus*.

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**Table 1: Results of antibiotic sensitivity test**

| antibiotic | Number senstitive | Senstitive % | Number intermediate | Intermediate % | Number resistant | Resistant % |
|---|---|---|---|---|---|---|
| Vancomycin | 28 | 93.33 | 1 | 3.33 | 1 | 3.33 |
| Imipenem | 28 | 93.33 | 1 | 3.33 | 1 | 3.33 |
| Tetracycline | 22 | 73.33 | 1 | 3.33 | 7 | 23.33 |
| Amoxillin clavulanic acid | 18 | 60 | 1 | 3.33 | 11 | 36.66 |
| Ciprofloxacin | 29 | 96.66 | 1 | 3.33 | 0 | o.oo |
| Azithromycin | 26 | 83.33 | 1 | 3.33 | 4 | 13.33 |
| Levofloxacin | 28 | 93.33 | 1 | 3.33 | 1 | 3.33 |
| cefixime | 20 | 66.66 | 1 | 0.00 | 10 | 33.33 |

**Table 2. Sensitivity of *S.aureus* to hot watery extracts of plants in mm**

| Plant | 10mm | % | 15mm | % | 20mm | % | 25mm | % | 30mm | % |
|---|---|---|---|---|---|---|---|---|---|---|
| *Linum usitatissimum* | 7 | 23.33 | 7 | 23.33 | 5 | 16.66 | 9 | 30 | 2 | 6.66 |
| Pomegranate | 7 | 23.33 | 9 | 30 | 4 | 13.33 | 9 | 30 | 1 | 3.33 |
| sumac | 5 | 16.66 | 9 | 3o | 5 | 16.66 | 10 | 33.33 | 0 | 0 |
| *Lawsonia inermis,* | 13 | 43.33 | 7 | 23.33 | 6 | 20 | 1 | 33.33 | 0 | 0 |
| *Crocus sativus* | 11 | 36.66 | 4 | 13.33 | 7 | 23.33 | 8 | 26.66 | 0 | 0 |

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RESEARCH ARTICLE

# The Short Laser Pulse to Create Micro-Plasma for Identification of Kidney Stone Elements

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## ABSTRACT

Laser induced breakdown spectroscopy (LIBS) is a high resolution spectroscopy which used for assignment of kidney stone elements is a chemometric method. This method is a rapid spectro-chemical analysis technique that uses a short laser pulse of Q-switching Nd-YAG laser with 1064 nm wavelength with energy (200,300 and 400) mJ and pulse duration (9ns). The important experimental parameters such as the laser energy distance between the lens and sample and angle of the optical fiber from the target were optimized.The short laser pulse to create a micro-plasma on the sample surface.The emission spectrum of kidney stone elements were analyzed by using spectrum analyzer (Thorlabs) model CCS 100/M made in Germany with spectrum range from (320-720) nm , optical resolution about (0.5)nm and time integration 800μs.

**Keywords:** Kidney stone, High resolution spectroscopy, LIBS.

## INTRODUCTION

Laser induced breakdown spectroscopy (LIBS) technique based on analyzing of atomic emission spectroscopy for excited atoms and ions of elements in plasma formed on the surface of the samples by means of focused pulsed laser. There are several new developments of the LIBS method that are reported elsewhere[1]. The main benefit of the LIBS techniques is that it can be used for direct chemical analysis without difficult chemical preparation provided for the samples. It has a very high sensitivity, reach part per million [2]. This method is used to detect bones, skin and human's fossils. It is also used to detect background characteristic likes age, sex and statues of bodies[3]. A kidney stone can be as small as a grain of sand or large as golf ball about 80% of stone made of calcium oxalate, calcium

phosphate or combination of both. A bout 10-15% is made of magnesium ammonium phosphate. A bout 5% are made of Uric acid less than 1% of stone made of Cysteine. There are five major categories as shown in Fig.(1)

## Plasma parameters

In LIBS experiments, after the initial plasma decay and during the entire observation interval the local thermodynamic equilibrium (LTE) conditions are assumed to hold. For optically thin plasma the re-absorption (self-absorption) effect of plasma emission are negligible. So, the emitted spectral line intensity (I) is measure of the population of the corresponding energy level of this element in the plasma[4].

The main parameters of laser induced breakdown spectroscopy (LIBS) are electron density and plasma temperature. The obtained spectra are helpful in extracting the electron density and plasma temperature. The factors that influence the emitted intensity of the plasma are the number density of the emitting species (ions and neutral), electron density and temperature. These parameters are also responsible for different excitation and ionization processes occurring in LIBS. Many methods have been described for determining the plasma temperature based on the absolute or relative line intensity (line pair ratio or Boltzmann plot), the ratio of lines to continuum intensity. etc. depending on the experimental condition.

One of these methods may be more suitable than others. Provided that the local thermodynamic equilibrium (LTE) hypothesis is fulfilled, the plasma temperature can be calculated by using the Boltzmann equation to generate a Boltzmann plot is given by:

$$I = \frac{hc}{4\pi\lambda} \cdot N(T) \frac{A_{Ki} g_K}{U(T)} \exp^{-\frac{E_K}{KT_e}} \quad \text{.....(1)}$$

where $\lambda$ is the wavelength , Aki is the transition probability, $g_K$ is the excited level energy, $T_e$ is the temperature , K is the Boltzmann constant, U(T) is the partition function. The emitted spectral line intensity from a given state of excitation can be used to evaluate the plasma temperature. The lines must be well resolved for accurately evaluating their wavelength intensity (I), and their probabilities $A_{Ki}$ must be known. Reformulating Saha Boltzmann equation (2)gives:

$$\ln\left(\frac{I\lambda}{A_{Ki} g_K}\right) = -\frac{E_K}{KT} - \ln\frac{U_T}{F_i} \quad \text{.....(2)}$$

where F is an experimental factor and C is the species concentration. By plotting the left hand side of equ. (2) Vs. the excited level energy $E_K$, the plasma temperature can be obtained from the slope of the obtained straight line[5].

The emitted spectral line intensity is measure of the population of the corresponding energy level of a certain species in the plasma. Under the assumptions that plasma is both in LTE and optically thin, if we have information on the intensity emitted from several excited levels. We can then determine the plasma temperature which is responsible for the observed population distribution. The condition that the atomic and ionic states should be populated and depopulated predominantly by electron collisions, rather than by radiation requires an electron density which is sufficient to ensure the high collision rate. The corresponding lower limit of the electron density is given by Mc whirter criterion by below equation (3):

$$Ne \geq 1.6 * 10^{12} T^{1/2} (\Delta E)^3 \quad \text{.....(3)}$$

where ΔE (eV) is the highest energy transition for which the condition holds, and T is the plasma temperature. This criterion is a necessary, though insufficient condition for LTE, and is typically fulfilled during the first stage of plasma lifetime. It is, however, difficult to satisfy for the low-lying states, where ΔE is large. However for any Ne, it

is possible to find high excitation level where the states are close enough for equation (3) to hold. In this case the plasma is said to be in partial LTE[6-8].

## MATERIALS AND METHODS

**Experimental part**

The optical emission spectra for plasma ablation from kidney stone samples surfaces were recorded using LIBS experimental system which is shown in figure(2).The system consists of Q-switching Nd-YAG laser wavelength 1064nm with 9ns pulse duration and 10 Hz pulse repetition frequency, with. Different energies (200,300,400)mJ were used laser beam is focused on the surface of the sample located at the focal length of the converging lens (f=10cm). Optical fiber adjusted at 45$^{o}$ with beam directed at 10 cm distance from the sample where plasma was generated. Spectroscopic information was obtained from laser targets plasma spectra in air at atmospheric pressure. The spectrum was recorded over a (320-720)nm wavelength range using spectrum analyzer (Thorlabs) model CCS 100/M resolution < 0.5 nm supplied with thorlab.Program to record the spectrum in PC- computer.National institute for stander and testing (NIST) data base were used to identify the emission spectrum of kidney stone elements as well as to obtain some of atomic a ionic constant necessary for calculate electron temperature and plasma density.

**Sample preparation**

The kidney stone samples were supplied by medicine city hospital in Baghdad, Iraq for different size, age and sex and used as it is.

## RESULTS AND DISCUSSION

Figure (3) shows plasma emission spectra for kidney stone sample of sex-male for age 23year in air at atmospheric pressure in the spectral range (320-750)nm and laser irradiance at wavelength 1064 nm at different laser pulse energy from (200-400)mJ. It has found that the spectral emission lines for elements CaI, PII, C II, N II, Mg II, S II, O I at different wavelength which identified using (NIST) atomic spectroscopic data base. The intensity of the plasma emission spectrum will be increase with crease laser pulse energy irradiance.The plasma temperature (Te) at 400mJ laser pulsed energy irradiance was determined for the neutral calcium lines by using saha-Boltzmann equation (2) as shown in figure (5)**.**The electron temperature for Neutral CaI as calculated from Boltzmann plots is Te = (28000 k) and the electron density was calculated by Mc whirtr criterion using equation (3)is Ne = (2.6051 x $10^{16}$ $Cm^{-3}$). Similar way the electron temperature and plasma electron density were calculated for the rest of elements of kidney stone and tabulated in Table(2).An important phenomena where a dip in the center of the spectral line profile was noted for neutral calcium due to the $3p^{6}3d4p \rightarrow 3p^{6}3d4s$ at 526.79 nm. As the laser energy is increased, a dip in the center of line profiles. Known self-absorption begins to appear which is shown in figure(6).

## CONCLUSION

LIBS technique can be used for direct chemical analysis for kidney stone samples without performing a sample preparation. The method has a high resolution spectroscopy to identify the elements in kidney stone such as (Ca, P, C, N, O, H , S, and Mg) in a very short time. It is also observed that the relative intensities of the spectral line of each elements vary with the change in laser irradiance. It is observed that the number density increases with increase in the laser irradiance, which is linked to the reflection and the absorption of the laser photons by the plasma and depends upon the plasma frequency and plasma shielding.

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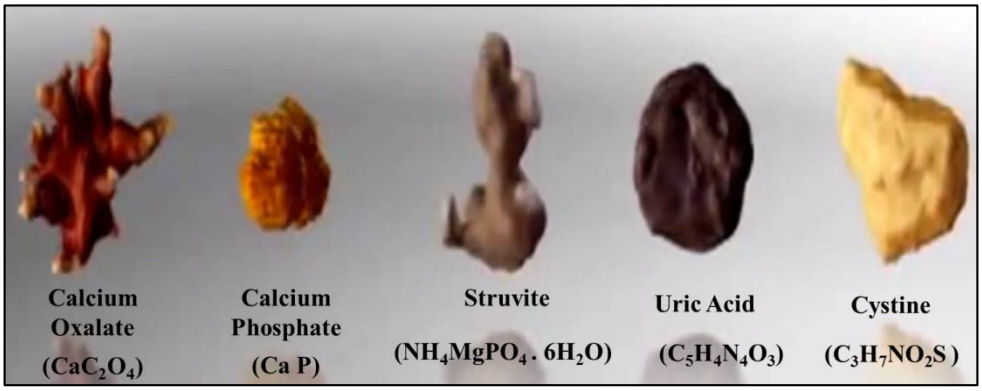


**Fig.1 : Shows a kidney stone with the five major categories as follows**

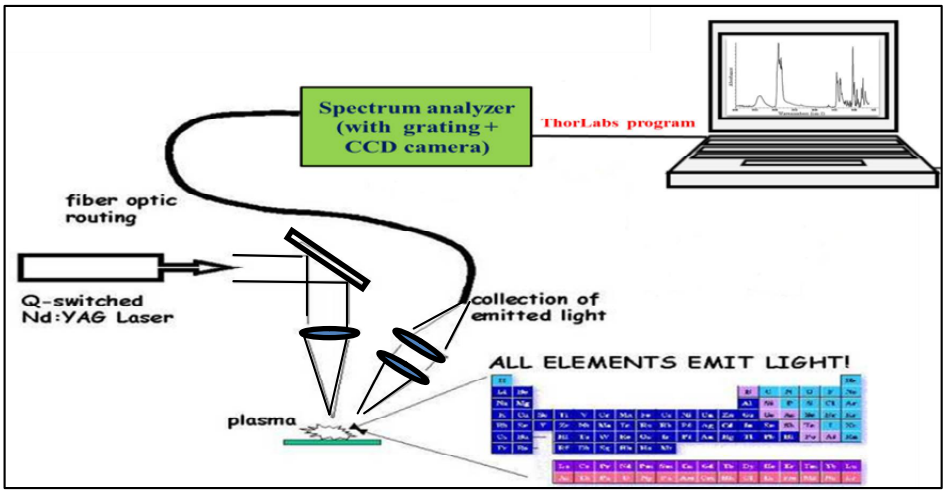


**Fig.2: Schematic diagram of the experimental setup for LIBS.**

**Fig.3 : Intensity Vs. wavelength for kidney stone sample (Male,23year) with variation in laser pulse energy irradiance.**

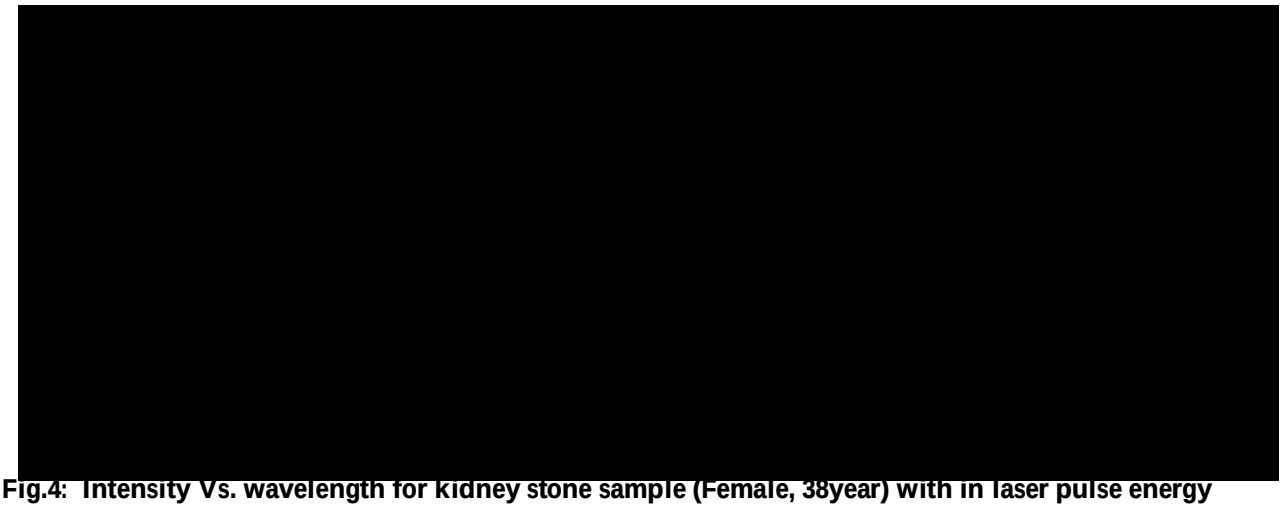

**Fig.4: Intensity Vs. wavelength for kidney stone sample (Female, 38year) with in laser pulse energy (180mJ).**

**Fig.5.Boltzmann plots for neutral calcium**

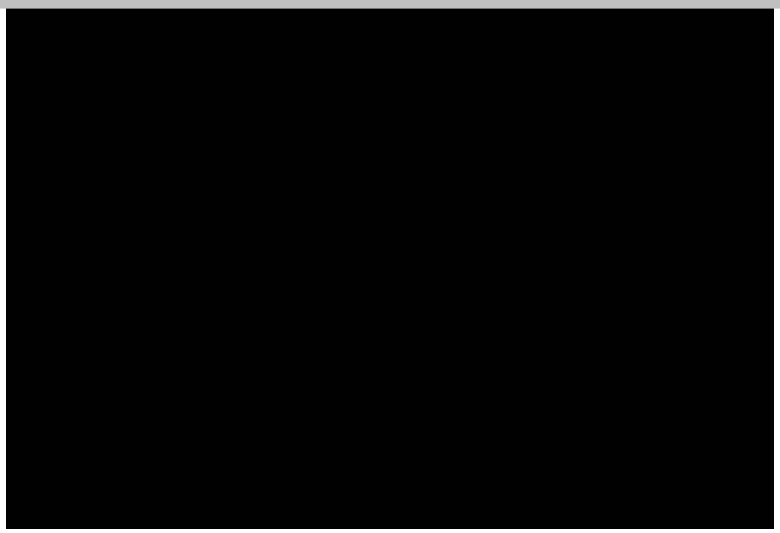

**Fig.6: Self-absorption phenomenon for neutral calcium lines at 526.79 nm.**

**Table 1: Atomic spectroscopic data of the neutral Calcium lines (Male - Ca I).**

| Element | λ | I | $A_{ki}$ | J | $g_k$ | El_eV | Eu_eV | Configuration |
|---|---|---|---|---|---|---|---|---|
| (Name) | Nm | (A.U) | ($10^8$ $s^{-1}$) | Upper | Upper | $Cm^{-1}$ | $Cm^{-1}$ | Upper→Lower |
| Ca I | 386.90 | 0.03 | 0.072 | 2 | 5 | 20335.360 | 46 164.644 | $3p^6 4s6f \rightarrow 3p^6 3d4s$ |
| Ca I | 422.73 | 0.08 | 2.20 | 1 | 3 | 0.000 | 23652.304 | $4s4p \rightarrow 4s^2$ |
| Ca I | 428.99 | 0.02 | 0.60 | 1 | 3 | 15 157.901 | 38 464.808 | $3p^6 4p^2 \rightarrow 3p^6 4s4p$ |
| Ca I | 430.24 | 0.04 | 1.36 | 2 | 5 | 15 315.943 | 38 551.558 | $3p^6 4p^2 \rightarrow 3p^6 4s4p$ |
| Ca I | 442.81 | 0.02 | 0.498 | 1 | 3 | 15157.901 | 37748.197 | 4s4d→4s4p |
| Ca I | 445.44 | 0.09 | 0.87 | 3 | 7 | 15315.943 | 37757.449 | 4s4d→4s4p |
| Ca I | 487.75 | 0.08 | 0.188 | 3 | 7 | 21 849.634 | 42 343.587 | $3p^6 4s4f \rightarrow 3p^6 3d4s$ |
| Ca I | 526.79 | 0.17 | 0.15 | 1 | 3 | 20 335.360 | 39 335.322 | $3p^6 3d4p \rightarrow 3p^6 3d4s$ |
| Ca I | 558.63 | 0.24 | 0.49 | 3 | 7 | 20371.000 | 38259.124 | 3d4p→3d4s |
| Ca I | 610.01 | 0.04 | 0.096 | 1 | 3 | 15157.901 | 31539.495 | 4s5s→4s4p |
| Ca I | 643.62 | 0.14 | 0.53 | 4 | 9 | 20 371.000 | 35 896.889 | $3p^6 3d4p \rightarrow 3p^6 3d4s$ |
| Ca I | 646.36 | 0.10 | 0.47 | 3 | 7 | 20 349.260 | 35 818.713 | $3p^6 3d4p \rightarrow 3p^6 3d4s$ |
| Ca I | 671.48 | 0.06 | 0.12 | 1 | 3 | 21 849.634 | 36 731.615 | $3p^6 4s5p \rightarrow 3p^6 3d4s$ |

**Table 2 : Electron temperature and electron density of kidney stone for different elements at different laser pulsed energy irradiance**

| A | Laser pulse Energy (mJ) | Plasma Temperature $T_e$ (K | Electron Density $N_e$ ($cm^{-3}$) |
|---|---|---|---|
| Ca II | 400 | $7.2 * 10^4$ | $12.5581 * 10^{16}$ |
| C II | 180 | $14.41 * 10^3$ | $156.2110 * 10^{16}$ |
| C I | 200 | $7.2 * 10^3$ | $67.0636 * 10^{16}$ |
| C (I,II) | 400 | $4.8 * 10^4$ | $214.4792 * 10^{16}$ |
| O I | 180 | $7.2 * 10^4$ | $48.6912 * 10^{16}$ |

| | | | |
|---|---|---|---|
| O II | 300 | $2.88 * 10^4$ | $646.0529 * 10^{16}$ |
| O II | 400 | $2.88 * 10^4$ | $643.0052 * 10^{16}$ |
| N II | 180 | $3.6 * 10^4$ | $324.3468 * 10^{16}$ |
| N II | 200 | $7.2 * 10^4$ | $112.8508 * 10^{16}$ |
| N II | 400 | $3.6 * 10^4$ | $276.9900 * 10^{16}$ |
| P II | 200 | $2.4 * 10^4$ | $64.9897 * 10^{16}$ |
| P (I,II) | 300 | $48 * 10^4$ | $82.6506 * 10^{16}$ |
| P II | 400 | $4.8 * 10^4$ | $113.2381 * 10^{16}$ |
| H I | 400 | $7.2 * 10^3$ | $29.7068 * 10^{16}$ |
| Mg I | 180 | $2.4 * 10^4$ | $4.3901 * 10^{16}$ |
| Mg ( I , II ) | 300 | $20 * 10^4$ | $92.1599 * 10^{16}$ |
| Mg ( I , II ) | 400 | $7.2 * 10^4$ | $76.5408 * 10^{16}$ |
| S II | 180 | $7.2 * 10^3$ | $60.1850 * 10^{16}$ |

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**RESEARCH ARTICLE**

# Antibacterial, Mechanical, Barrier and Color Properties of Plastizer Thymol in Nanocomposites Polylactic acid , Nano clay

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## ABSTRACT

Polylactic acid (PLA) is biodegradable aliphatic polyester well suited for packaging applications. Antibacterial active films based on poly(lactic acid) (PLA) were prepared with thymol and nano clay (1,2,4, and 6 wt%) by solvent casting methods. The films were characterized structural,by( X-ray diffraction , FTIR) , mechanical, gas barrier, color and antibacterial properties. X-ray diffraction analysis show that all peaks of thymol are presented intense crystalline peaks at 2θ= 15°, 17°,20°,23º, and 30 º that mean the crystalline nature of the thymol and appear mixed with PLA and only 6° , 20°, 30° for nano clay .FTIR analysis show bonds of represent the ring aromatic group of thymol and hydroxyl groups . Mechanical properties (Tensile strength, Young Modulus, Elongation and Tear resistance) show improvement in flexibility because that thymol acted as plasticizers, which reduce the intermolecular forces of polymer chains, thus improving the flexibility and extensibility of the films. Barrier properties (oxygen transmission rate and water vapor permeability) show slightly decreased with the increase of thymol loading and increased when adds nano clay. Color test show that the PLA/thymol/nanoclay composites doesn't have any significant effects on films transparency, that the key research challenges in the development of nanocomposites based on biodegradable polymer used in packaging food application.The antimicrobial properties of thymol containing films showed a significant activity against *Escherichia coli* and increased improvement inhibition zone . The results indicated the potential of PLA/thymol /nano clay nanoomposites for applications in antimicrobial packaging and helping to create synergism of the biopolymers.

**Keywords:** Polylactic acid , thymol , nano clay , antibacterial , barrier , color

## INTRODUCTION

Some material polymer derived from environmentally friendly materials (biopolymers) has generated a lot of Poly Hydroxyl alkanoic e.g., PHA (PHA), poly hydroxy butyrate (PHB), and Poly Lactid Acid (PLA). However, one of the emerging biopolymers examined are PLA (Poly Lactid Acid) due biodegradation capabilities so high that classified as environmentally friendly polymers. Unlike synthetic polymers are generally derived from petroleum materials, PLA is derived from renewable sources, namely from a variety of plants that have a high starch content such as cassava, sweet potato, banana, corn and so on and Some characteristic properties of pure PLA such as weak thermal stability, low gas barrier properties, and low ductility and toughness are inadequate for food packaging applications [1].In recent times, several PLA-based technologies have emerged with an emphasis on achieving chemical, mechanical, and biological properties equivalent or superior to conventional polymers. The frequent need for a chemical or physical modification of PLA to achieve suitable properties for its intended consumer and biomedical applications, however, has demanded significant attention in the last decade [2]. PLA is brittle and exhibits low thermal stability, medium gas barrier, and low solvent resistance against water. As pointed out earlier, the intercalation of nanoplatelets with high aspect ratio such as layered silicate montmorillonite clay can lead to an enhancement in the polymer matrix properties in terms of mechanical properties and barrier [3].

PLA is linear aliphatic thermoplastic polyester derived from lactic acid, which is obtained from the fermentation of 100% renewable and biodegradable plant sources, such as corn or rice starches and sugar feed stocks. It can be produced by chemical conversion of corn or other carbohydrate sources into dextrose. Dextrose is fermented to lactic acid followed by polycondensation of lactic acid monomers or lactide. However, the most common way to produce PLA is the Ring Opening Polymerization (ROP) of lactide monomer formed from lactic acid [1-3].PLA has excellent properties compared to other biopolymers, including:

a) Environment friendly, PLA is biodegradable, recyclable and compostable.
b) Biocompatibility, PLA should not produce toxic or carcinogenic effects in local tissues
c) Processibility, PLA has better thermal processibility
d) Energy savings, PLA requires 25–55% less energy to produce

The synthesis of PLA is a multistep process that starts from the production of lactic acid and ends with its polymerization. An intermediate step is often the formation of the lactide. Figure 1 shows that the synthesis of PLA can follow three main routes. Lactic acid is condensation polymerized to yield a low molecular weight, brittle polymer, which, for the most part, is unusable, unless external coupling agents are employed to increase its chains length. Second route is the azeotropicdehydrative condensation of lactic acid. It can yield high Monomers, Polymers and Composites from Renewable Resources, molecular weight PLA without the use of chain extenders or special adjuvents. The third and main process is ring-opening polymerization (ROP) of lactide to obtain high molecular weight PLA, patented by Cargill (US) in 1992. Finally, lactic acid units can be part of a more complex macromolecular architecture as in copolymers [4].

Nano Clay minerals are the basic constituents of clay raw materials and platy structure is the dominant morphology. Depending on the clay type, the individual layers could be composed of two, three or four sheets of either $[SiO4]^{-4}$ tetrahedra or $[AlO3(OH)3]^{6}$ octahedra. Nanoclays are easily available, environment friendly, low cost chemical substances and a large volume [5]. Clay minerals are hydrous silicates and may simply be described as fine-grained particles with sheet like structure stacked over one another. Owing to this geometry, they are commonly known as phyllosilicates, sheet-structured silicates. Phyllosilicates are mainly composed of fine grained aluminosilicates and are formed as a result of chemical weathering of silicate minerals at the surface of the earth. Nanoclays are commonly dominated by phyllosilicates and may be separated from the clay fraction or the bulk clay material by different approaches. Methods used for extracting and processing of nanoclays include energetic stirring followed by centrifugation and freeze-drying; centrifugation and cross-flow filtration; and ultracentrifugation [6,7]

Nano clay is fine-grained crystalline materials. A layer is the basic structural unit of nanoclays and these layers are prone to arrange themselves. On this score layered silicate mineral nanofillers, particularly montmorillonite nanoclays, offer great advantages due to both economical (availability of resources and reasonable price) and technological (nanodimensions, high aspect ratio, excellent exploitation properties such as mechanical stiffness/strength, flame retardancy and barrier characteristics) factors. Greatly due to relatively low costs of this type of nanofiller, improvement of intercalation/exfoliation technologies as well as high cost-performance ratio of the nanocomposites[8].

Thymol is a plastizers principle component of thyme (Thymus vulgaris) and oregano (Origanum vulgare) essential oil, has received extensive interest as effective antimicrobial and antifungal agent. Thyme oil was incorporated into several films including soy edible film and chitosan film. Thymol is only slightly soluble in water at neutral pH, but it is extremely soluble in alcohols and other organic solvents. It is also soluble in strongly alkaline aqueous solutions due to deprotonation of the phenol[9]. Thymol is a plasticizers that low molecular weight material added to polymeric materials such as paints, plastics, or adhesives to improve their flexibility and lowers Tg makes material more flexible ( increase chain mobility). In packaging plasticizer is a substance added to materials to impart flexibility, workability, and elongation .Thymol has a refractive index of 1.5208 and an experimental dissociation exponent (pKa) of 10.59±0.10. Thymol absorbs maximum UV radiation at 274 nm.Thymol displays strong antimicrobial activity against natural spoilage bacteria and food borne pathogens, e.g. Salmonella typhimurium, Escherichia coli, Listeria monocytogenes, Bacillus cereus, and Staphylococcus aureus [ 10 ].

There are a number of natural antibacterial bio-active agents such as essential oils (e.g. thymol, carvacrol and linalool), plant extracts (e.g. olive leaf extract), enzymes (lysozyme and peroxidase), chelating agents (e.g. amino acids), metals (e.g. silver) that have been incorporated into packaging materials to provide excellent activity. A wide range of essential oil (EO) extracts including thymol have been incorporated directly into packaging materials , that has the potential to inhibit Gram-positive bacteria and to a lesser extent, Gram-negative bacteria, including the pathogenic strains of Escherichia coli, Staphylococcus aureus and Bacillus cereus and activity of thymol has been attributed to its hydrophobic nature whereby it is most likely to partition in the lipid layer of bacteria cell membranes and mitochondria resulting in the leakage of cell contents and by minimizing the growth of microorganisms due to the inherent antimicrobial properties. Thymol and menthol are naturally occurring plant derived compounds, which have excellent pharmaceutical and antimicrobial application [11].

The objective of this study was to develop a new thymol incorporated antimicrobial and with PLA/nano clay films which can be used to packaging application and to evaluate the effect of thymol on the barrier , structural, mechanical, color and antimicrobial properties of PLA/thymol / nano clay films over a range of different concentrations (1, 2, 4, and 6 wt%) . This can potentially extend the shelf life of some food products and can be further developed as a rigid packaging material and/or coating.Ray et al. [5] studied the properties of PLA nanoclay composite films. They observed an intercalation of the layered silicates that resulted in an improvement in modulus of the nanocomposites and a noticeable reduction in oxygen permeability.

## MATERIALS AND METHODS

Poly (lactic acid), Pure grade PLA AI - 1001, with density 1.25 g/ cm3, was supplied by (Shenzhen Esun Industrial Co., Ltd. Chain).Nano clay supplied by Nanoshel LLC (USA)with average diameter of the particles (as recorded by the company) was about < 80 nm. Chloroform Solution was purchased from Applied Chem. (Darmstadt, Germany).

### Preparation of samples

Pure PLA prepared with weighted grade (3wt%) and then dissolved in chloroform to obtain 30 wt% solution of PLA grade by slowly in 60°C for 2 hours warming until the solution become viscous using magnetic stirrer hot plate, then

cast into petri dish with 20cm in diameter at room temperature for 24 hour to ensure complete solvent removal. The thickness of the cast solution was approximately 105 µm.PLA/ thymol films were prepared at percentages (90/10) (wt. /wt.) polylactic acid dissolved in chloroform for one hour to have good distribution and thymol also dissolved in chloroform and two solutions were mixed for an hour , then cast on the glass plates and then kept at 60°C to ensure complete solvent removal.PLA/thymol/ nanoclay nanocomposite films were prepared by many steps a weight percent of nanoparticles were used with different weight percentages (1, 2,4 and 6 wt. %) and swollen in chloroform by mixing for 2 hours while shearing mixer at 800 rpm mixed with solution of polylactic acid /thymol at 90/10 (wt./wt.) .After two solutions were mixed for an hour and sonication process was again introduced in order to improve dispersion of nano filler in the polymer matrix. Then cast on the glass plates and then kept in vacuum oven for 24 hour.

**Characterizations**

**Thickness**

Determination of thickness of PLA and PLA/thymol /nanoclay films calculated by electronic digital micrometer (293-821, Mitutoyo) sensitivity was used to measure the thickness of PLA and PLA/nano clay films and find that is 105µm.

## X-Ray Diffraction (XRD) Analysis of pure PLA, PLA/thymol and PLA/thymol /nano clay

The structure of PLA and PLA/thymol /nano clay films were characterized by Phillips X'Pert Pro MRD (Cu Kα radiation ($\lambda$=1.54 nm),40 kV,40 mA) between 5° and 60°.

**Fourier Transform Infrared (FTIR) Analysis**

The infrared spectra were recorded with the help of Shimadzu FTIR -7600 in range 400 to 4000 $cm^{-1}$.

**Mechanical Properties**

**Tensile Strength**

According to ASTM D-882[12] standard modulus of elasticity, tensile strength, and percent elongation equipped with a 5 kg load cell in tensile mode. Tested films were cut in 10 mm width and 80 mm in length and the initial gauge length and testing speed were fixed at 10 mm/min. Two metallic grips were attached for griping both ends of the test specimen of the film. Tensile strength ($\sigma s$)**,** Young's modulus ($E$) were determined according to the following equation:

$\sigma s = F/(A)$ ……1

$E = F L_0/A \Delta L$ ……..2

**Where:** ***F***: force exerted on an object under tension, $L_0$: original length, ***A*:** cross section area, $\Delta L$: length of the object changes.

**Tear Strength**

Tear strength of films was determined on the same Universal Electronic Dinamometer and carried out according to ASTM D-1922[13] standard, using the trouser tear method. The sample size was 8cm long and 6.3 cm wide were cut from composite blown film in machine direction and Samples for tear resistance were cut from middle of the width. A pendulum impact tester is used to measure the force required to propagate slit a fixed distance to the edge of the test sample.

**Optical Properties**

**Color test and Brightness**

Color properties were evaluated measuring color coordinates in the CIELAB color space L* (lightness), a*(redness and greenness) and b* (yellowness and blueness) were analyzed using a Konica CM-3600d color flexdiff. The instrument was calibrated with a white standard and measurements were carried out in quintuplicate at random positions over the film surface. Average values for samples were calculated. Total color differences (ΔE) was evaluated by Equation (3)

$$\Delta E=\sqrt{\Delta a2 + \Delta b2 + \Delta L2} \quad \text{.....3}$$

Where: ΔL = L stander*-L sample , Δa = a stander*– a sample , Δb = b stander*-b sample , Stander values for white plate (L*: 93.49, a*: −0.25, b*: −0.09)respectively . Five measurements were taken on each film, one at the center and four around the perimeter, and the mean values were used.

**Barrier properties**

**Oxygen Transmission Rate (OTR)**

Oxygen Transmission Rate (OTR) of the films was measured according to the ASTM D-3985[14] using gas permeation instrument Qualities (Canada). The diffusing oxygen is measured by a detector that is sensitive only to oxygen, after 12 to 16 hours test was completed .Before each measurement, the samples were kept dry in vacuum oven. The measured time interval is then transformed into the gas permeability rate expressed in $cm^3/m^2$.day. Gas permeability of the films was determined at constant temperature (23°C) and relative humidity (0% RH) conditions and tests Area 50$cm^2$and thickness ≤ 3 mm.

**Water Vapor Transmission Rate**

The water vapor transmission rate WVTR of the films was measured according to the ASTM E-96[15] using water permeation instrument Qualities (Canada).Film specimens were mounted horizontally on poly (methylmethacrylate) cups filled with distilled water up to 1 cm under the film. The cups were weighed every hour for a period of value was noted usually after 6-8 hours. The slopes of the steady state (linear) portion of weight loss versus time curves were used to calculate WVTR at conditions of 37°C and 90% relative humidity with 50$cm^2$ area of a body and thickness is ≤ 3 mm, with unit gm /$m^2$·24h.

**Antimicrobial activity**

The E.coli was selected to demonstrate antibacterial activity via an agar disc diffusion assay. The bacteria culture was stored in TSB broth containing 30% v/v glycerol at -80°C and was sub-cultured in broth twice before being used. Samples of 0.1 mL of E. coli suspensions containing approximately 106 - 107 CFU mL-1 were spread over a prepared agar surface in a Petri dish. Film discs of 6 mm diameter were placed in triplicate on the Petri dish containing the agar. Plates were incubated at 37°C for 24 h and the diameter of the resulting inhibition zone was measured directly after the incubation period using a digital Vernier calliper. The presence of a clear zone of inhibition around the test films was taken as indication of antibacterial activity in the films and the test was performed in triplicate.

## RESULTS AND DISCUSSION

### X-Ray Diffraction (XRD) of nano clay

XRD patterns of the clay nano powder is given in Fig (2A) shows the XRD of the clay nano powder typical peaks around 2Θ= 5°, 20°, 25°,30°, and 35 ° of the (001), (002) (111), (003) (005),reflections corresponding to Na – montmorillonite.In Fig (2B) shows the XRD of the PLA/thymol /nano clay nanocomposites peaks at 2θ= 6°,15°,17°,20°,23°,and 30° where 2θ =17°, 20° of pure PLA that mean is semicrystalline it was shown by Nadia A. [16]. thymol are presented intense crystalline peaks at 2θ= 15°, 17°,20°,23º, and 30 º .All these peaks demonstrated the crystalline nature of the thymol and appear mixed with PLA and only 6° , 20°, 30° for nano clay .

### Fourier Transform Infrared (FTIR) Analysis of pure PLA and PLA/thymol /nano clay

Figure (3A) shows the FTIR spectra of the pure PLA, PLA/thymol and PLA/thymol/nanoclay. For pure PLA the peak at 3000-2850 $cm^{-1}$ were assigned to the –C–H asymmetric and symmetric vibration of $CH_3$ groups in the side chains, peak at 3424 $cm^{-1}$ of -OH . The peak at 2921$cm^{-1}$ asymmetric stretching –$CH_2$-, peak at 1730 $cm^{-1}$ corresponding to the stretching vibration carbonyl group (C-O) from the repeated ester units (is due to the carbonyl group in the lactic acid) , peaks at (1300-1500 ) $cm^{-1}$ of the deformational vibrations of methyl group of PLA are appear at peal at 1414 $cm^{-1}$ of –$CH_3$ bending vibration and peak at 1150 $cm^{-1}$ of –C–O– stretching vibration from the ester units, and peak at 934 – 851 $cm^{-1}$ of C-C single . These statements are similar those described by Nadia A. [16 ] and Auras et.al [17 ] , where shows characteristic stretching frequencies for C=O, –$CH_3$ asymmetric, –$CH_3$ symmetric, and C–O, at 1746, 2995, 2946 and 1080 $cm^{-1}$ respectively.

Figure (3B) shows that a small amount of thymol is present on the film surface with a higher concentration of thymol and shows the intensity of the FTIR band mapped of PLA containing 10% w/w thymol film a band due to the hydroxyl group stretching vibration appears in the range of 3400–3500 $cm^{-1}$ and 2991 cm-1 also of hydroxyl group , , band at 2941 $cm^{-1}$ of C-H methyl group stretching , band at 1610-1504 $cm^{-1}$ that represent the ring aromatic group of thymol , band at 807 $cm^{-1}$ of ring aromatic bending OH .Moreover, the band at 3500-3400 $cm^{-1}$ with the expected absence of a sharp, carbonyl group absorption at approximately 1610 $cm^{-1}$ .

In Fig (3C) for PLA/thymol /nano clay at 6% the nanocomposite spectrum presented the presence of bands in the zone of absorption of the silicate groups between 900 - 1100 $cm^{-1}$ confirming the incorporation of the nanoclays in PLA E matrix. Exactly the bands found were at 460 $cm^{-1}$(Si-O-Si bending), 520 $cm^{-1}$(Si-O-Al bending), 1540 $cm^{-1}$(Si-O stretching), and at 1675 $cm^{-1}$ (-OH bending from absorbed water).In the case of PLA/thymol /nano clay, it is possible to observe the characteristic bands for thymol, at 730 and 805 $cm^{-1}$owing to ring vibrations of aromatic groups[36]. Presence of thymol evidenced by the presence of phenolic groups bands from 3650 to 3200 $cm^{-1}$, and at 1000-970 $cm^{-1}$ corresponding to C-O bending. Some displacement of thymol peaks occurred with the presence of organoclay, revealing some interactions between thymol and modified organoclays.

### Mechanical properties of pure PLA and PLA/nano clay/thymol

The films intended for food packaging applications required sufficient flexibility to avoid breaking during the packaging procedure that mean need materials have high flexibility, the goal of blending with thymol with PLA was to enhance plastic elongation and reduce brittleness of PLA.The mechanical properties (Tensile Strength, Elongation, tear resistance and Young modulus) of PLA, PLA/thymol , and PLA/thymol /nano clay at (1,2,4, and 6% wt) films were shown in Table 2.

It could be seen that blending PLA with thymol decrease in TS from (9.43 to 7.12 ) MPa , Young modulus from (2.83 to 2.02 )GPa and Tear Resistance from (12.5 to 10.4) m N/mm but improved and increased in Elongation between (8-134 )% at break of PLA film upon blending with thymol . Thymol is a plastizers is able to decrease interaction forces between PLA chains and increase distance between Chains. The main goal of plasticizer addition into PLA matrix was to decrease rigidity of PLA and increase ductility of PLA because that PLA a brittle material while the addition of thymol increases the ability of PLA to plastic deformation which is reflected in the decrease of yield stress and an increase of % elongation that appear in Table (2) , showing increase flexibility to 134% in (90/10) wt % thymol that reduces the intermolecular force and increases the mobility of the polymeric chains, thereby improving the flexibility and plastic deformation because exhibited to reduce the cohesive force between the molecular chains that vary important in packaging application because need to flexibility, workability and increased elongation which would also result in changes in the crystallinity of polymer resulting in an increase in ductile properties.

In PLA/thymol /nano at (1,2,4, and 6wt%) nanocomposites , increased in tensile strength, Young Modulus and Tear Resistance values was detected , the combined action of thymol and nano clay on the PLA mechanical behaviour was evaluated and shows increase of the nanocomposites was observed resulting in more flexible and stretchable materials because the presence of nano clay contributed to the thymol ability to increase the PLA chain mobility, which also promoted a more effective dispersion of nanoparticles. These combined effects could be related to the presence of Van der Waals interactions between the hydroxyl groups of thymol appear in FTIR

When add thymol and nano clay increased in Tensile Strength , Young Modulus between (10.3-20.5)MPa , (2.09-2.99)GPa and Tear Resistance between (11.7 to 19.7) m N/mm respectively because that due to the hydrogen bonding interaction between PLA and nano clay and the structure of PLA consists of two hydroxyl groups at the end of its polymer chains these prepared PLA/nano clay composites embedded in a network of PLA polymeric chains and the origin hydrogen bonds formed between the PLA molecules and the nano clay and these new hydrogen bonds would improved the mechanical properties. The improvement of mechanical properties due to reinforcement effect of the rigid inorganic nano clay, which constrains the molecular motion of PLA chains. The main reason for this behavior may be attributed to the resistance exerted not only by the clay itself with high surface area high aspect ratio and vary high elastic modulus , but also by the stronger interfacial interaction between the matrix and layered silicate due to the vast surface exposed to the clay layers [18]. Good dispersion of the clay platelets in the polymer reduced tensile ductility and increased tensile strength compared with neat polymer. Theoretically, the complete dispersion of clay nanolayers in a polymer optimizes the number of available reinforcing elements for carrying an applied load and deflecting cracks. The coupling between the tremendous surface area of the clay and the polymer matrix facilitates stress transfer to the reinforcement phase, allowing for such tensile and toughening improvements [19,20 ]. Tear resistance improves dramatically by the incorporation of the clay particles. This can be explained by considering that the silicate layers are able to inhibit or at least to slow down crack propagation by deviating their tear path .

However, brittleness might limit the application of PLA/nano/thymol films in the packaging industry where elasticity is required that is very useful for packaging applications. Ranging in elongation between (134-921) % with increase in nano clay concentration that because the nanocomposites of non-polar polymers such as polylactic acid with clays exhibit weak interfacial interactions and low degree of exfoliation and consequently show improved stress at break values because effect of plastizers thymol in nanocomposites [9 ].

## Optical properties of pure PLA and PLA/nano clay/thymol

Color is important factors to be considered in food packaging since it could influence consumer acceptance and commercial success of a food product. Table 3 shows Color parameter and brightness of pure PLA , PLA/thymol and PLA/thymol/ nano clay/ Nanocomposites.

Color of PLA/thymol blend film was a little higher than that of pure PLA. This might be because that thymol was colorless semi-transparent crystals. In Table (3) the differences among samples were not perceptible to the human eyes. In PLA/thymol /nano and brightness of the films had high transparency even at high thymol and nano clay concentrations. This result suggested high transparency for PLA/ thymol /nano clay nanocomposite films was suitable for food packaging application. Because the aspect of packaging could influence the decision of consumers, high transparency in food packaging is required when used thymol and nano clay mean good distribution appear in XRD and FTIR.

**Barrier Properties (OTR, WVTR) of pure PLA and PLA/nano clay /thymol**

Generally biobased polymers, like PLA, show higher , OTR , WVTR values than other polymers .Barrier properties are very important in the main functions of food packaging is to avoid or minimize moisture transfer between the food and the surrounding atmosphere. The effect of thymol amount on (OTR,WVTR) of PLA/thymol blends was shown in Table (5)that are increased to (1812.8 $cm^3.mm/m^2.day.bar$ and 196.3 respectively) because that addition of a plasticizer thymol caused reduce intermolecular forces and increases free volume between polymer chains, which not only increases flexibility, but also oxygen permeability and water vapour transmission rate containing to be higher compare to PLA because that thymol is hydrophilic and functional groups ends , therefore when addition to the PLA polymer does bring a negative effect on the water permeability of the blend due to the higher free volume attained and the corresponding changes in diffusion and also in solubility of water in this blend and that lead to increased in OTR and WVTR to 199.3 $g.mm /m^2 .day$ compare with PLA is 181.8 $g.mm / m^2 .day$ that mean the addition of plasticisers adversely acts the barrier properties and increases mass transfer through the film , due to the higher mobility of the polymer chain and higher free volume that mean thymol acted as a plasticizer and rearranged the polymer chains layers subsequently lower the degree of crystallinity and increased the free volume in the polymer matrix [9, 10 ].

The main limitations of PLA as a packaging material are a high gas permeability ($O_2$, and water vapor) that show in Table (4) .Higher oxygen permeability coefficients and water vapour transmission rate are an indication of lower barrier protection that appear in pure PLA . The high molecular weight glassy polymers (PLA) with rigid chains have very high oxygen permeability. Polylactic acid is showing that (OTR) and (WVTR) of PLA taking into account of (488.96 $cm^3.mm/m^2.day.atm$) and (181.818 $g.mm /m^2 .day$) respectively that mean pure PLA is biodegradable nature it suffers from some serious drawbacks such as its great sensitivity towards moisture and its poor water vapour barrier property [19] . PLA is a relatively poor barrier to water and PLA is hydrophobic significantly tend to be non-polar , that mean exhibit a high contact angle but is sensitive to water vapour that appear in Table (4).In PLA/clay nanocomposites/thymol shown in Table (4) it is found that there is a significant reduction in OTR and WVTR with increasing clay content, which starts to level off at clay additions at 6 weight %, the reduction in permeability is due to the effect of the nanoclay due to effect of nanoclay on the barrier properties of polymers can be quantified by the 'tortuous path' due to the maximum improvement in barrier properties, assuming that the clay platelets are 100% intercalated or exfoliated and perfectly aligned. Polymer nanocomposites are constructed by dispersing a filler material into nanoparticles that form flat platelets [20].

These platelets are then distributed into a polymer matrix creating multiple parallel layers which force gases to flow through the polymer in a "torturous path", forming complex barriers to gases and water vapor. As more tortuosity is present in a polymer structure, higher barrier properties will result in 1-6% of nano clay to polymer matrix due to the impermeable clay layers distributed in polymer matrix consequence increasing the effective diffusion path length. The tortuous path theory is based on the premise that a molecule must fellow a more complicated path when nano clay is dispered throughout the polymer matrix than when the matrix consists of the homopolymer alone[19-21] .

**Antibacterial activity**

The antibacterial effects of the films evaluated by agar diffusion method because of its ability to simulate wrapping foods and to be used as a quantitative test. This method is based on the release of active agents from composite film disc into the agar, producing a clear zone of bacterial growth inhibition around the Gram negative bacteria E. coli and when addition of higher concentrations of nano clay expanded the inhibition zone widths that appear in Fig (5) and Table (5). It was not surprising and no inhibition of E. coli was evident for the pure PLA films containing no thymo but full growth of PLA/thymol and PLA/thymol /nano clay 6% because that thymol is a volatile organic compound extracted from thyme and it has excellent antibacterial properties[22]. Thymol is able to inhibit growth of gram -positive microbes and gram-negative and also nano clay is very effective nanoparticles have been studied in different applications, including food packaging, due to their strong antibacterial properties.

## CONCLUSION

Films based on PLA were prepared with thymol and nano clay by solvent casting methods. The addition of thymol decreased tensile strength, young modulus, tear resistance and increased elongation and the flexibility and extensibility of the films with thymol.Barrier properties increased when addition thymol and decrease at PLA/thymol and nano clay because that tortuous path of platelets of nano clay .Optical properties did not change with the addition of thymol but increased when addition nanoclay. PLA/thymol films containing 10wt% thymol exhibited significant antibacterial effect against E. coli increased when addition nano clay composite films might be supplied commercially for antibacterial packaging applications activity during food storage.

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**Table 1: Chemical structure of commonly used smectite type clays, M: monovalent cation, x: degree of cations isomorphous substitution in octahedral sheets, [8].**

| Smectites | Chemical formula |
|---|---|
| Montmorillonite | Mx(Al4-xMgx)Si8O20(OH)4 |
| Hectorite | Mx(Mg6-xLix)Si8O20(OH)4 |
| Saponite | MxMg6(Si8-xAlx)O20(OH)4 |

**Table 2 : Mechanical properties of pure PLA , PLA/thymol and PLA/thymol /nano clay**

| Samples | Tensile Strength MPa | Elongation % | Young Modulus GPa | Tear Resistance mN/mm |
|---|---|---|---|---|
| **Pure PLA** | **9.34** | **8.00** | **2.83** | **12.5** |
| **PLA/Thymol 90/10 %** | **7.12** | **134** | **2.02** | **10.4** |
| **PLA/Thymol/CLAY %1** | **10.3** | **388** | **2.09** | **11.7** |
| **PLA/Thymol/CLAY %2** | **13.4** | **394** | **2.39** | **13.5** |
| **PLA/Thymol/ CLAY%4** | **15.1** | **428** | **2.55** | **17.6** |
| **PLA/Thymol/CLAY %6** | **20.5** | **921** | **2.99** | **19.7** |

**Table 3: Color Properties of pure PLA and PLA/nanocomposite/thymol**

| Sample | $L^x$ | $a^x$ | $b^x$ | Brightness |
|---|---|---|---|---|
| Pure PLA | 90.05 | 14.38 | -2.76 | 80.88 |
| PLA/Thymol 90/10 | 90.37 | -22.08 | 2.49 | 83.66 |
| PLA/ Thymol/CLAY %1 | 93.37 | -22.17 | 2.51 | 84.97 |
| PLA/ Thymol/ CLAY %2 | 93.80 | -21.27 | 2.77 | 88.89 |
| PLA/ Thymol / CLAY%4 | 94.25 | -21.13 | 34.31 | 94.46 |
| PLA/ Thymol/ CLAY %6 | 98.89 | -21.54 | 4.73 | 104.89 |

**Table 4: Oxygen Permeability Coefficients and Water Vapour Transmission Rate of PLA and nanocomposites/thymol**

| Samples | Oxygen Permeability Coefficients ($cm^3.mm/m^2.day.bar$) | Water Vapour Transmission Rate( $g.mm/m^2.day$) |
|---|---|---|
| Pure PLA | 488.9 | 181.8 |
| PLA/Thymol | 1812.8 | 199.3 |
| PLA/ Thymol/nanoclay %1 | 332.9 | 101.7 |
| PLA/ Thymol/nano clay %2 | 325.5 | 116.9 |
| PLA/ Thymol / nano clay %4 | 211.2 | 101.3 |
| PLA/ Thymol/ nano clay %6 | 124.6 | 83.8 |

**Table 5: Inhibition zone of E.coli for pure PLA , PLA/thymol , and PLA/thymol /nano clay**

| Sample | samples | Inhibition zone diameter (mm ) of E. coli |
|---|---|---|
| A | PLA | - |
| B | PLA/thymol | 8 |
| C | PLA/nano clay 6% | 10 |
| D | PLA/thymol /nano clay 6% | 12 |

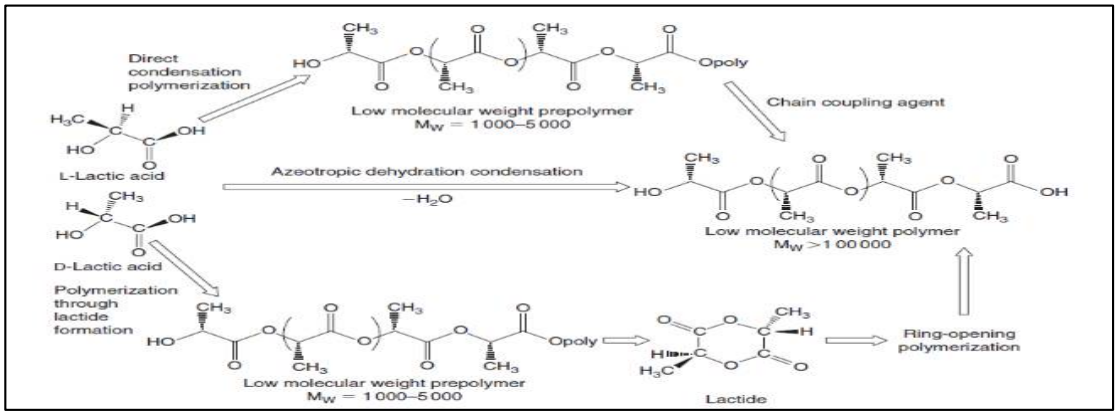


**Fig.1: Synthesis methods for obtaining high molecular weight of PLA[4]**

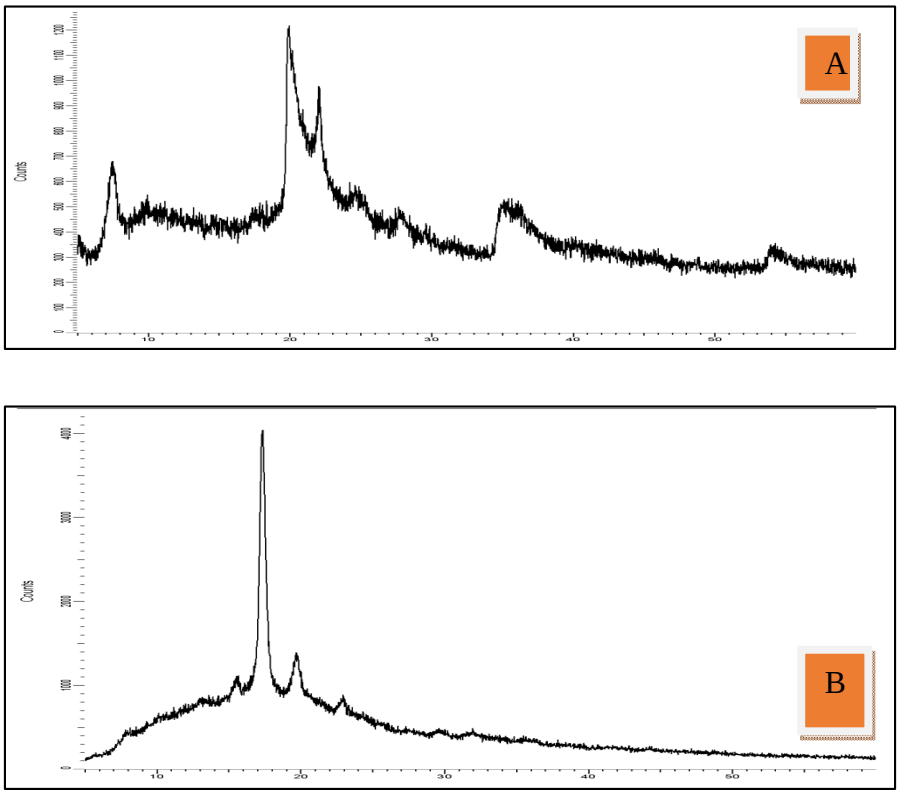


**Fig.2 : X-ray diffraction of nano clay and PLA/thymol /nanocomposites at 6%**

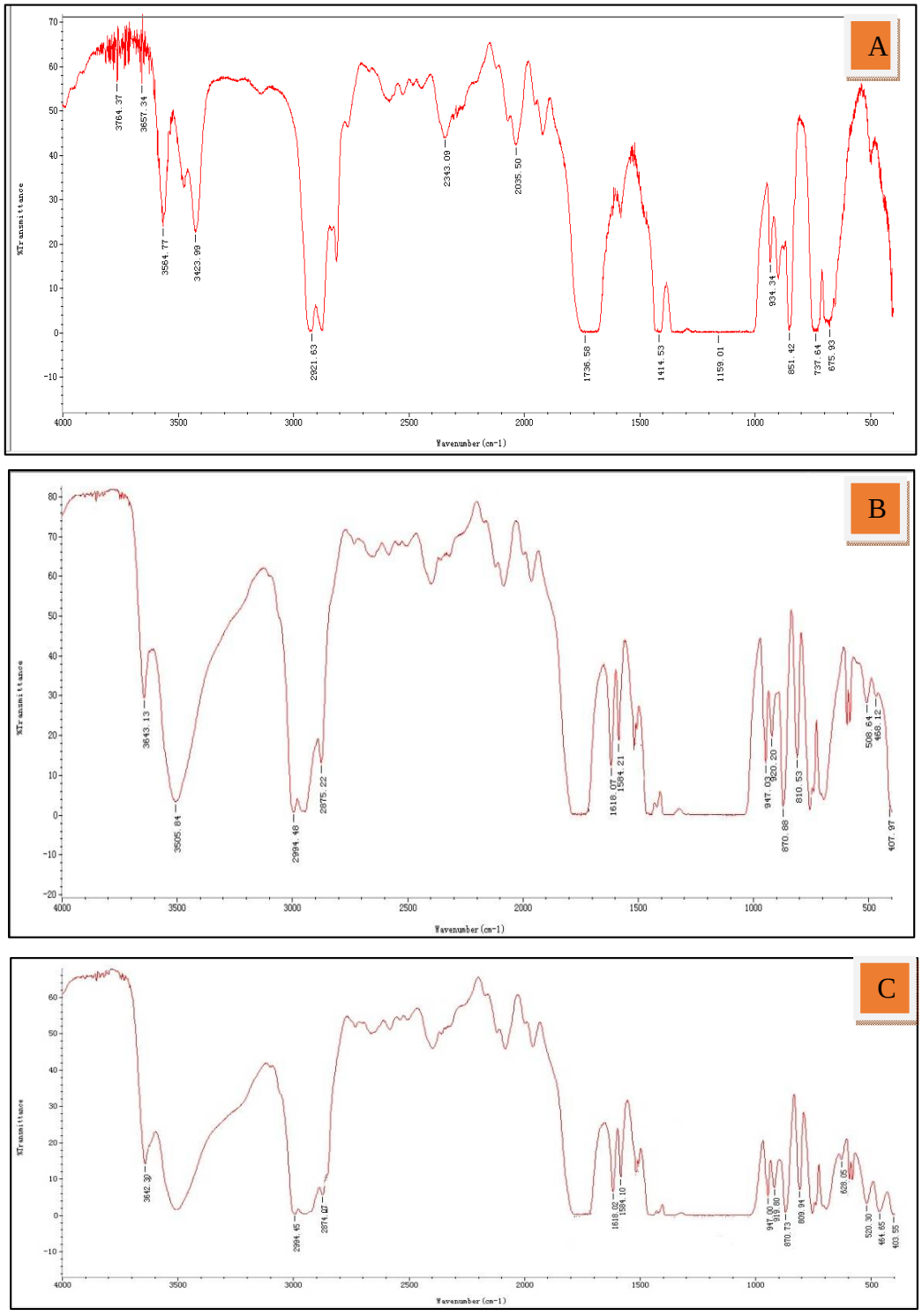


**Fig.3A.shows the FTIR spectra of the pure PLA, 3B. shows that a small amount of thymol is present on the film surface, 3C.for PLA/thymol /nano clay at 6% the nanocomposite spectrum.**

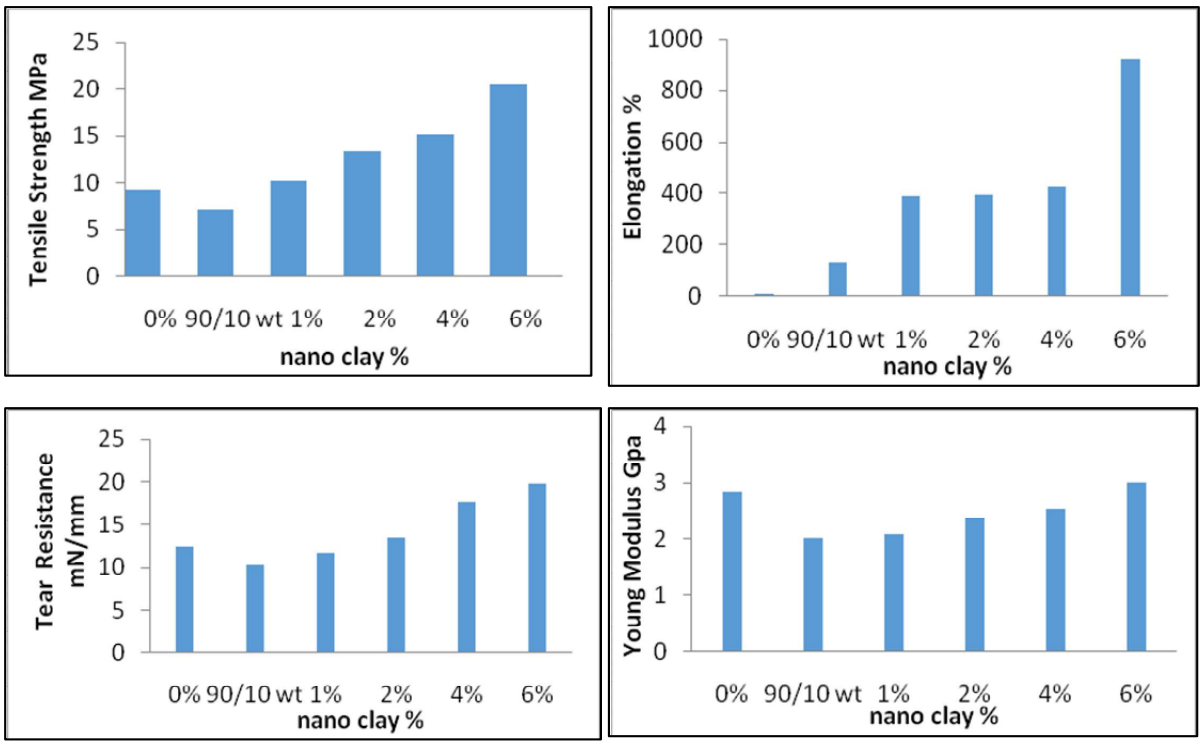


**Fig.4: Mechanical properties of pure PLA , PLA/thymol (90/10) wt%, PLA/thymol /nano clay (1,2,4,6wt%)**

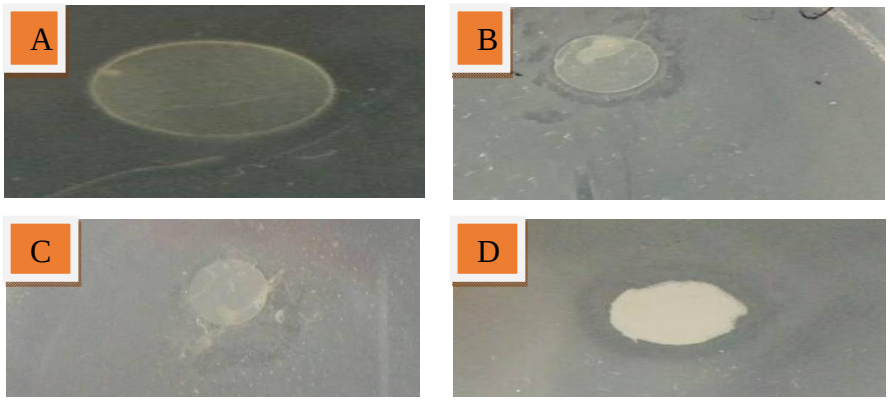


**Fig.5: Antibacterial activity of films against E. coli after 24 h of incubation at 37°C: (a) PLA control, (b) PLA with 90/10 wt% thymol , (c) PLA /nano clay at 6 % and (d) PLA /thymol/nano clay**

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RESEARCH ARTICLE

# Antimicrobial Activities of Cranberry Extract to *Ecoli* O157 by using Molecular Techniques.

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## ABSTRACT

The plants are used as alternative medicineto treatment bacterial infections.In this study previously cultured isolates *E.coli* O157 from urine clinical sample from hospital of Baghdad city.Our study showedantimicrobial activity of hot aqueous extract cranberry with different concentration (0.5,1,3,6 %) by using agar diffusion methods and give high diameter inhibition zone when using concentration 6% .This extract of cranberry were prepared to study plasmid curing against *E.coli* O157.The UV transilluminatorof agarose electrophoresis were used to observed the plasmid DNA bands of *E.coli* show ability of extract of cranberry to induce the cleavage in cells (DNA fragmentation) of *E.coli* which show disappear of plasmid in 6% concentration.Many antibiotics as (Erythromycine(E)Chloromphenicol(C) Cephotaxime(CTX)Trimetheprime(TMP)Gentamycin(CN)Ciprofloxacin(CIP)Cephlaxine(CL)Oxytetracy line(T)Amoxicillin(AX)Cefixime(CFM)Nitrofurantion(F) were tested. It revealed the microbial E.coli resistance to nine antibiotics and intermediate resistant to ciprofloxacine and nitrofurantoin.This study revealed that hot aqueous extracts of cranberry better plasmid curing activity ,so to prevent the development of urinary tract infection can combinationthe cranberry extract with antibiotics.

**Keywords:** E.coli, cranberry , Aqueous extracts, plasmid DNA, curing .

## INTRODUCTION

Herbal medicines are used of since ancient times, as antiviral, antifungal and antibacterial activities[1]. Medicinal plants consist of many kind of active composition such as terpenoids, alkaloids, sulphur compounds, phenols,esters, aldehydes, alcohols, glycosides, thymol are responsible for ant inflammatory activities[2].Urinary tract infections were common and costly medical problem affecting the people, *Escherichia coli* are most bacteria responsible for most Urinary tract infections[3].Cranberry consist proanthrocynadins and fructosewhich like *E.coli* structure so, they enterin the receptors of the uroepithelial cells and blocks theireceptors of uroepithelial cell. *E. coli* will notable to

enter in the receptors of uroepithilial cells andthey are killed by the antibiotics and flush out whenpatient consume the water[4].Genes accountable forresistance to antibiotics arefound inplasmid DNA. R plasmids consist kindsof genes encoding resistance to wide spectrum antimicrobial agents asantibiotics, heavy metals, disinfectantasformaldehyde , mutagenic agents asethidium bromide ,therefore the ways to little plasmidmove of antibiotic resistance is to remove the plasmids. This process is known as plasmid curing[5].Cranberriesfruite are wealthy in benzoic acid, which is then excreted in urine as hippuric acid, Therefor the advantage of cranberry juice was based on the urinary excretion of hippuric acid, which is a bacteriostatic agent and has the potential to acidify urine [6].

## MATERIALS AND METHODS

### Bacterial samples

The pathogenic *E. coli* O157 were isolated from UTI patients in Baghdad hospital, Iraq. Then transported with sterilized condition to zoonotic laboratory Veterinary collage of Baghdad university ,then cultured on blood agar were incubated for 24h at 37C. The isolates were identified in laboratory by bacteriological methods such as gram stain, colony morphology, and biochemical tests and cultured on sensitive media for *E.coli* o157 (Hi Chrom media) which give purple colure[7].

### Preparation of cranberry Extract

### ExtractPreparation

cranberry fruit used in this study were collected from local shops imported from United Arab Emirates.The fruits washed , dried, pulverized and then sieved.

### Aqueous Extraction

The aqueous extraction will accorded to[8]. By taking 10 gram dried powder put in 100 ml water in beakers and heated for 10 minute on a magnetic stirrer untiltemperature reached to 95C°, allowed the mixture to cool for 10 min ,filtered through filter paper to remove smaller particles .Then dissolved Extracts in 10 ml DMSOto make extract 0.5%, 1% , 3% and 6% solutions ( Table 1).

### Antimicrobial activity

### Disc diffusion assay

According to[9] (Grove and Randall, )1955[10] and Kavanagh, 1972),byinoculated clinical isolates in nutrient broth and incubated at 37 C overnight then taking 5 ml suspensions of bacterial (1.5 ×$10^8$ CFU/ml) (density 0.5 on the McFarland scale)Plates of muller-hinton agar were prepared and 0.1 ml of culture broth were added and spread with a sterile spreader. Wells were made in the plates with a crock borer (0.65 cm).The wells were filled with 0.1ml of each concentration of 0.5%, 1%, 3%, and 6%, mg of extract and the plates incubated overnight at 37 C. The antimicrobial activities were test by measuring the diameter of the zone of inhibition in mm. The same technique was used for determination of antibiotic (erythromycin , amoxicillin, ceftxime,oxytetracycline,cloromphenicol, cefotaxime, trimetheprime, gentamycin, ciprofloxacin ,nitrofurantion and cephlaxin).

**Curing of plasmid mediated**

According for [11] the Isolation of plasmid DNA content for plasmid profile (fermentas).Agarose electrophoresis technique.

## RESULTS AND DISCUSSION

The present study show the appearance of *E. coli* O157 is bacillus, gram-negative. Pink colonies on MacConkey agar due to lactose fermentation and give purple colure when cultivation on sensitive media for *E.coli* o157 (Hi Chrom media) while produces a zone of hemolysis on blood agar and it appearance motile in microscopic. Givepositive result in Indol test and when added to hydrogen peroxide produce the enzyme catalase.This study examined activities of antimicrobial of aqueous extract of cranberry against the *E.coli O157* and its force was assessed by the diameter inhibition zone when increased concentration Table(1). Which agreed with [12] who foundthe Different concentrations of Cranberry extract caused different degrees of zones of inhibition against *E. coli* O157:H7? This refers the sizes of inhibition zones were according to different concentration.

Also this study contract with [13]who studied the activitycranberry extract against different pathogens including *E. coli*, *Salmonella* spp, *Listeria monocytogenes P. aeruginosa and S. aureus,*and agreed with[14]who declared that *E. coli* O157:H7 was dominant from urinary tract infection patients *E. coli* isolates revealed different results against the antimicrobial disc. *E. coli* (O157) showed resistance to nine antimicrobial disc( erythromycin , amoxicillin, ceftxime,oxytetracycline,clorомphenicol, cefotaxime, trimetheprime, gentamycin and cephlaxin) ,while the isolates give intermediate resistant to ciprofloxacine and nitrofurantion Table(2). That similar to study of [15] who record the isolated of *E. coli* give highest resistance to nearly all antibiotics except Rifampicin, Oflaxacin, Gentamicin, Tetracyclin.

In present study using aqueous extracts of cranberry showing *E.coli* plasmid DNA curing gradually by using different concentration, this show the complete curing getting by increase the concentration of extracts of cranberry 6% (Figure 1).Is agreement with[16] who found The ability of both genera E.coli and protues adherence to epithelial cells was decreased when exposed to cranberry during in vitro experiment and similar to [15]find the plasmid DNA present in the *Acinetobacter spp.,E.coli* and *Proteus spp*.Completelycured by Using methanolic extracts of seeds of *Cuminumcyminum, Coriandrumsativum* and *Myristicafragrans* [17] reported curing of plasmid by *Plumbago. Auriculata* root extracts is higher frequency as 13%, *E. coli* with 15%, *P. vulgaris* with 32% and *K. pneumoniae* with 30%

## CONCLUSION

The aqueous cranberry extractgive highly potency of plasmid curing of isolates *E.coli* bacteria by using agarose gel electrophoresis.

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**Table 1:- Zone of inhibition (mm) of cranberry against *E.coli*O157**

| Isolates | Inhibition Zone (mm)in 0.5%. | Inhibition Zone (mm)in 1% | Inhibition Zone (mm)in 3% | Inhibition Zone (mm)in 6% |
|---|---|---|---|---|
| **E.coli** | **5** | **5.5** | **8** | **10.3** |

**Table 2:Zone of inhibition (mm) of Antimicrobial agents against *E.coli*O157**

| Antimicrobial agents | *E.coli* ( O157) | | |
|---|---|---|---|
| | S | I | R |
| Erythromycin | (0)mm | (0)mm | (0)mm |
| Cloromphenicol | (0)mm | (0)mm | (0)mm |
| Cefotaxime (CTX ) | (0)mm | (0)mm | (0)mm |
| Trimetheprime (TMP) | (0)mm | (0)mm | (0)mm |
| Gentamycin (CN) | (0)mm | (0)mm | (0)mm |
| Ciprofloxacine (CIP) | | (18)mm | |

| | | | | |
|---|---|---|---|---|
| Oxytetracycline | (T) | | | |
| Nitrofurantion | (F) | | (17)mm | |
| Amoxicillin | (AX) | (0)mm | (0)mm | (0)mm |
| Ceftxime | (CFM) | (0)mm | (0)mm | (0)mm |
| Cephlaxin | (CL) | (0)mm | (0)mm | (0)mm |

R: resistance, I: intermediate, S: sensitive

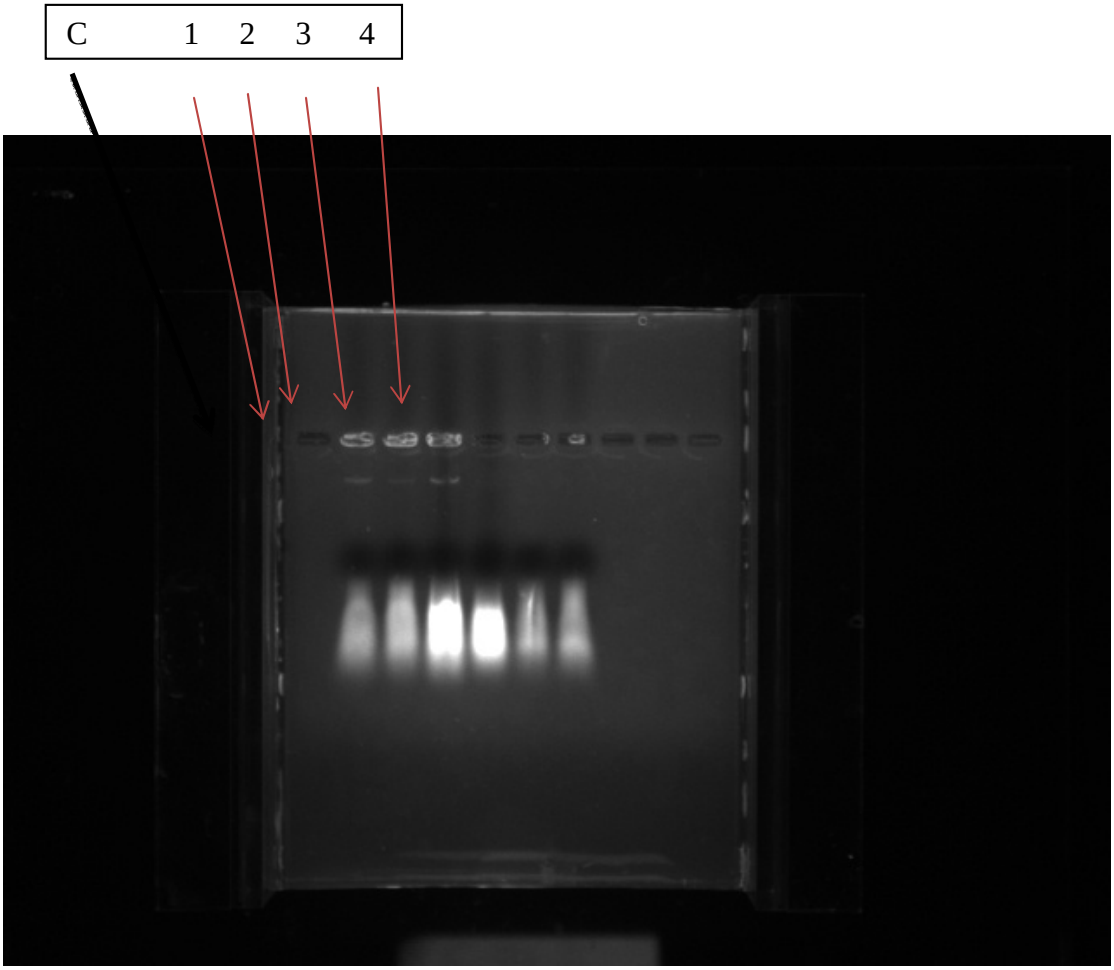


**Figure 1**: Agarose gel electrophoresis analysis of plasmid DNA from *E.coli* O157 arrow black control untreated after treatment show DNA fragmentation induced by different concentrations of the cranberry aqueous extract , whereas; treated *E.coli* with 1-treated with 0.5% , 2- treated with 1% , 3- treated with 3% and 4- treat treated 6% (Plasmid disappearance) C- Control.

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**RESEARCH ARTICLE**

# Cost Analysis of Water Purifying Techniques at Household Level

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**ABSTRACT**

This study investigates the economical treatment to purify the contaminated water. Various techniques are used to remove toxins to make it suitable for human health. The cost of most economical treatment to remove toxins from drinking water is Rs. 1.25 for open surface water while for storage tanks water it costs only Rs. 0.15. After following different techniques water is risk-free for drinking and cooking. Boiling and reverse osmosis treatment methods are not economical because they cost per liter as Rs. 1.00 to Rs. 2.50 and Rs. 3.472 respectively. The cost bored on treating the impure water is most suitable than expenses on medical treatments after illness. Using pure water of different brands available in the market is costly. In case of different types of impure water, this paper recommends different treatments methods. Research also guides different treatment methods of impure water.

**Keywords:** Drinking Water, Reverse Osmosis, Activated Carbon, Boiling, Cost Analysis

## INTRODUCTION

Water is absolutely imperative for the survival, development and assurance for human life; these reasons make it a novel item in this world. In any case, roughly one billion people in this world not have a direct access with consumable water, particularly those individuals living in underdeveloped nations. Water crisis has forced on these areas where threatening the standard of living in the developing countries. As indicated by explanation of the United Nations on Human Development Report, roughly in one year, 1.4 million youngsters expire because of inaccessible and unclean drinking water, and 3.6 million individuals lapse each year because of water-borne diseases, including the 84% are kids and 98% are grown-ups, the individuals who breathing in the developing countries. Anderson and Eikebrokk (2012) are of the view that it is above board, it is a key health problem in this world. It must be required

some extraordinary consideration to save the life of the millions individuals because of coming up short of survival from preventable illnesses [1].Devlin RA (2011) argues that waterborne diseases are the main cause of sicknesses that is reason of misery and demise in under developed countries. In developing countries enhanced arrangement of water and sanitation can serve to enhance health of half population of the world because the incalculable diseases are the result from hazardous drinking water. Studies concerned with water, health and cleanliness predict that roughly 2.4 million deaths could be saved every year if each individual with great honed on cleanliness framework and had crisp water for drinking. The diseases that are the result of unhygienic water include typhoid, malaria, cholera, gunie worm, amoeba, giardia and other pests [2].Many pathogenic microbes like protozoa and bacteria in contaminated water are transferred in human body by drinking as well as through washing and cooking food by using that contaminated water. Men are severely harmed by such pathogenic microorganisms and also grow many waterborne diseases in their body. Along with waterborne diseases millions of people in this world are also affected by water-washed diseases. Water-washed diseases mean those illnesses which can easily be removed by washing with water [3].Proper hygiene process has significant importance for healthy life. About 50% of common diarrhoea and other respiratory and skin diseases can easily be removed simply by washing hands with soap. Dubois.M et.al (2010) is of the opinion that neontal deaths can be reduced if hands are washed pos-delivery. Impure drinking water sometimes has faeces contamination of eggs, which results in form diseases like fever, vomiting, nausea and diarrhea [4].

In this paper, we want to analyze the costs of common water purifying techniques and available costs of pure drinking water selling under different brand names in the market of Pakistan with the justify a procedure to get pure water is feasible for common man.

## MATERIALS AND METHODS

It is stated that various methods have been introduced to extract impurities from contaminated water. Drinking water testing standard method are used to detect the quality of potable water, the standard parameters consider as pH, TDS, EC, TSS, etc. [5, 6]. Following methods on digital equipments are used for assessing quality control parameters that can assure drinking water quality as make it safe drinking water.

### Parameters to check Drinking Water Purity

According to Fux E (2011) methods used to check drinking purity are as follows [7]

- Spectrophotometer was used to check the turbidity and colour of water.
- pH was analysed by the use of pH meter.
- Utermohl method was used to examine the romoval of cyantoxins and cyanobacteria.
-

### Coagulants Solutios Preparation

### Aluminium Sulphate

$Al_2(SO_4)_3.14H_2O$ is the chemical formula of Aluminium Sulphate. In this study different concentration solutions were used as (5, 10, 15, 20, 25, 30 (mg/l)).Two jar test equipment was used to perform coagulation experiment at normal room temprature i.e 27$^{O}$C. Performance efficiency of Aluminium Sulphate is checked in different concentrations, which are mentioned above, to eliminate toxins from drinking water samples [8].

#### Boiling

5 litre volume vessels of 2 Kg made of steel were taken. Different samples of drinking as mentioned above were put in these vesses. Thermometers were put inside to measure temprature on different time periods as 2 minutes, 3 minutes and 5 minutes [9].

# RESULTS

## Cost-Benefit Analysis by Using Aluminium Sulphate

Aluminium Sulphate is an economical treatment. We can use it for the canal water treatment. Canal water comprises of 5% to 10% of toxins. For the treatment of ground water and storage water tanks, boiling is recommended. Toxins and organic materials vaporize in case of boiling. In this way low toxicity is removed and water is available for drinking.Figure-1 shows that by using coagulants the treatment cost per liter for canal water is Rs. 1.25 while for the storage water tanks and ground water is Rs. 0.50 and 0.15 respectively. After treatment water is safe for drinking purpose certainly.

## Using chemical Chlorination and Cost-Benefit Analysis

Figure-2 predicts that by using Ferric Chloride for canal drinking water is less than Rs. 1 per liter and Rs. 0.05 and Rs. 0.04 is for storage water tanks treatment and ground water treatment respectively.Microbial contamination in water is a genuine risk to every living creature, including plants, animals and human. It must need purification process appropriately. We test here two distinctive economical techniques. To start with utilize chlorine and second utilize boiling, both are reliable however ensure more economical and method depends on situations [10].

## Hot Boiling Process and Cost-Benefit Analysis

Coagulation treatment is more economical treatment as compared to boiling treatment. Boiling treatment costs from Rs. 2.50 to Rs. 1.00 (Figure-3). Water quality also influences treatment process. In boiling method 5 minutes are needed to successfully remove microbes disinfection. Chlorine treatment requires specific dose to disinfect microbes because excess usage of chlorine causes toxicity.

## Water Filtration Technology as Reverse Osmosis for Water Filtration

Reverse osmosis is a technique in which inorganic solids present in impure water are removed. In this technique impure water passes through semi-permiable membrane with pressure. In this way chlorine, lead, fluoride and dissolved salts and others are eliminated and water is available for drinking and making food. The cost analysis of drinking water is shown in Figure-4.

## Drinking Water Brands in the Market

In Pakistan different national and international companies are selling pure drinking water under different brand names. These companies follow different treatment methods to remove toxins from unhealthy water. The prices of different companies are as under Fig. 5.

## Cost comparison among different drinking water treatment techniques

Different methods are available for the purification of drinking water like chemical chlorination, using aluminium sulphate, boiling process and reverse osmosis etc. many companies are also selling pure drinking water in the market. Above table depicts that for the treatment of canal drinking water chemical chlorination is suitable treatment having cost just 0.15 per liter Rupee. For treatment of storage tank water again chemical chlorination is the best choice bearing cost 0.05 only. The economical method for the ground water is again chemical chlorination having cost 0.04 Rupee only as given in table 1.

However boiling technique is suitable for ground water treatment as compared to aluminium sulphate. While treating canal water and storage tank water, aluminium sulphate is economical choice. The most expensive drinking water is of different brands available in the market.

## DISCUSSION

Wang GS et al. (2007) is of the view that everyone in this world is anxious about water quality and wants to avail it as cheaper as possible [11]. That's why all the treatment methods can be used to remove all the harmful impurities from the drinking water. All the traditional methods should be improved to remove toxins from the drinking water so as to avail it economically. Chlorination is a common technique to remove impurities from drinking water but it should be used in cautious way. Its excess usage is the cause of cancer in humans. 3mg/l chlorine is enough for the treatment of canal water and 1.5 mg/l of chlorine is sufficient to remove toxins from storage tank water [11, 12].It is the high time to let people know about the use of pure water and its storage procedure so as to improve the health standard of the community. It is the only way to reduce water borne diseases like cholera and diarrhea etc. Chlorination and aluminium sulphate are very effective techniques to eliminate toxins like shiga toxin, botulinum toxin and microcystins etc. from the dirty water. Many researchers revealed that 90-98% toxins can easily be removed from impure water through chlorination and aluminium sulphate [13,14]. Trachoma which is the eye infection disease which leads to blindness can be prevented by just face washing habit. Trachoma is caused by bacteria (Chlamydia trachomatis). Its symptoms are cloudy cornea and swollen eye-lids etc. It multiplies through direct contact with used objects of the infected person like towel etc. the simple way to avoid it is face and hand wash. Trachoma is the cause of 8 millions blindness around the world mostly in Africa and Asia. Currently 55 countries are affected by this bacterial disease. Only pure water can avoid countless diseases [15,16].

## CONCLUSION

Above discussion depicts that the removal of impurities of different contaminated samples has different treatments. Above study reveals that use of chlorine and aluminium is suitable for different situations but their excess use can cause diseases. Boiling technique can also be recommended because it is easy, simple and economical. Reverse osmosis technique is also better and easily available in the market. At the end pure drinking water selling under various brands is costly one.

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**Table 1: Cost comparison among different drinking water treatment techniques**

| Water Samples | Treatment Cost per Liter in Pak Rupees (Rs.) | | | | |
|---|---|---|---|---|---|
| | Aluminium Sulphate | Chemical Chlorination | Boiling Process | Reverse Osmosis Water Filtration | Drinking Water Brands in the Market (Rs.) |
| Ground Water Treatment | 0.15 | 0.04 | 1 | 3.472 | 8.33 (Gourmet)<br>22 (Nestle & Acquafina) |

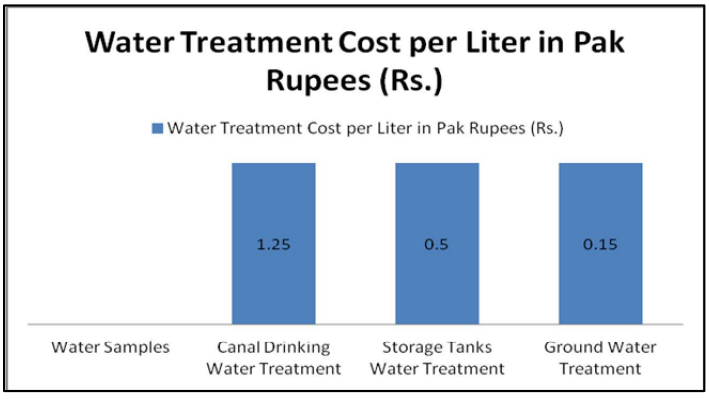


**Figure 1: Cost-Benefit Analysis by Using Aluminium Sulphate**

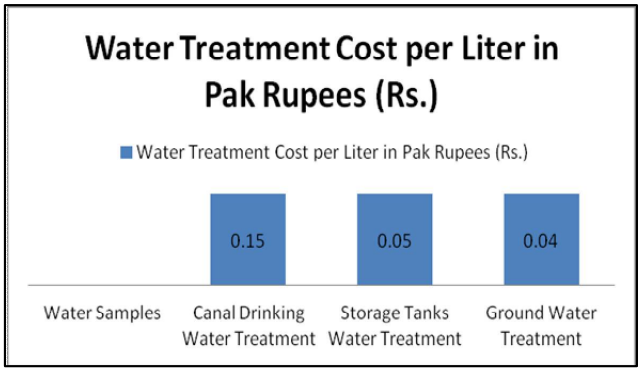


**Fig. 2: Chemical Chlorination and Cost-Benefit Analysis**

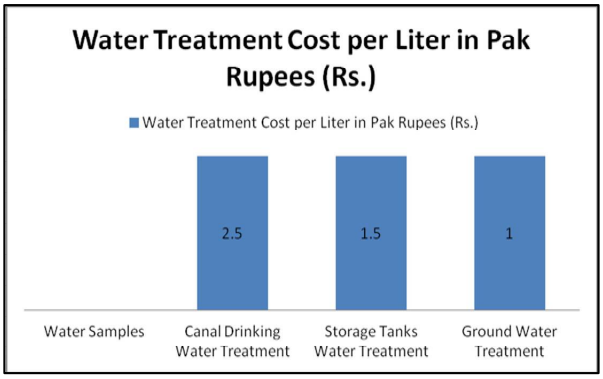


**Fig. 3: Cost Analysis method using Boiling Process**

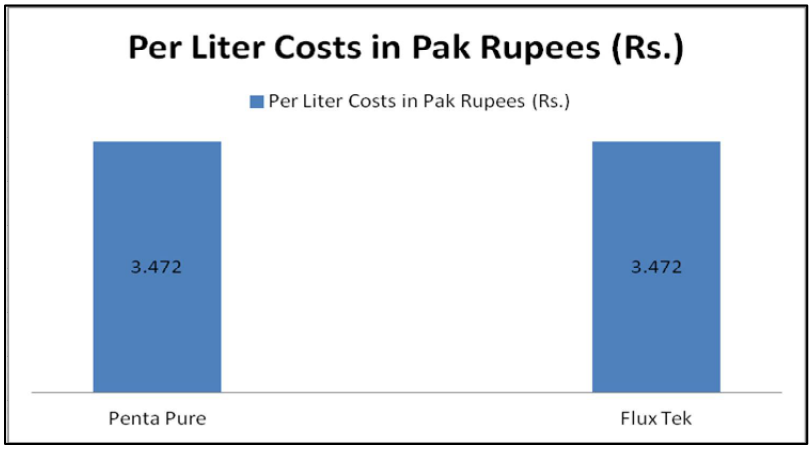


**Fig. 4: Water Filtration Technology as Reverse Osmosis for Water Filtration**
***assuming the life of machine one year and average use is 16 Liters**

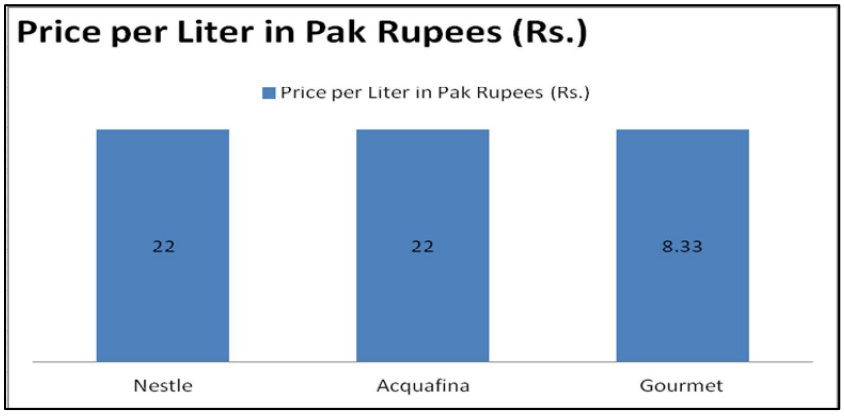


**Figure 5: Drinking Water Brands in the Market of Pakistan**

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RESEARCH ARTICLE

# Effects of Different Levels of DL-Methionine Supplementation on Low Hatch Weight Broiler Chicken Carcass Characteristics

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## ABSTRACT

The present experiment was carried out to study the effects of two hatching weight and three different dietary concentrations of methionine on carcass characteristics of broiler chicken. Totally one hundred sixty day old Vencobb -400 chicks were purchased, out of that forty chicks with hatch weight of 42.0- 45.0 g were placed in to G-I as average hatch weight (AHW) and another one hundred and twenty chicks with hatch weight of 35.0-38.0 g were placed into G-II, GIII and G-IV groups as low hatch weight (LHW) with four replicates of 10 chicks in each group. The AHW chicks were supplemented with BIS (2007) recommended methionine level in pre-starter, starter and finisher diet considered as 100 per cent. The diet of GII, GIII and G IV groups were supplemented with 100,110 and 120 per cent of BIS (2007) methionine recommendations respectively. The live weight of broiler birds at 42nd day at its market age were 2327.60 g, 2298 g, 2267.60 and 2262.80 g in G-I,G-II,G-III and G-IV respectively and evidenced not significant increase between groups. The broiler chicks on methionine higher than BIS recommendations showed non-significant increase in carcass weight, breast weight, carcass yield and breast yield.

**Keywords:** methionine, low hatch weight broilers, carcass yield, breast yield.

## INTRODUCTION

The researchers conducted number of studies to determine requirements of methionine for broiler chicksbecause they are considered as first limiting amino acid in practical corn-soybean meal based diets, andoften supplemented in synthetic crystalline forms. The recent research has suggested that levels of methionine in excess of NRC (1994) recommendations may result in enhanced performance, especially in regard to breast meat yield (Hickling *et al*., 1990). In India, the commercial broiler chicks (vencobb 430 Y) had average day-old hatch weight of 36 to 48 g. The variation in day-old hatch might be due to difference in egg weight and hatching process. Ulmer *et al.* (2010) studied that egg weight of 39.9, 43.0 and 46.5 g hatched out 34.1, 37.0 and 40.1 g of day old chick weight in 29$^{th}$ week flock age when compared against 59$^{th}$ week flock age of Vencobb 500 broiler breeders. The researchers reported that market weight of commercial broilers was 1.1 times higher than broilers chicks had hatch weight of 53.1 g compared against day-old hatching weight of 43.5 g (Sklan*et al*., 2003). The regulatory effects of methionine on growth performance and breast meat yield have been demonstrated in broilers (Ahmed and Abbas, 2011). However, there was no study reported onmethionine requirement for different hatch weight chicks. The present study envisaged to evaluate the recommendations of BIS (2007) for methionine and effect on carcass characteristics in broilers.

## MATERIALS AND METHODS

### Experimental design and diets

Totally one hundred sixty day old Vencobb -400 chicks were purchased, out of that forty chicks with hatch weight of 42.0- 45.0 g were placed in to G-I as average hatch weight (AHW) and another one hundred and twenty chicks with hatch weight of 35.0-38.0 g were placed into G-II, GIII and G-IV groups as low hatch weight (LHW) with four replicates with 10 chicks in each group. The experimental rations were prepared as per BIS (IS: 1374; 2007) recommendations for all the nutrients except variation in the DL methionine content in pre-starter, starter and finisher ration. Experimental ration (R1) was prepared with BIS recommended DL-methionine (0.50, 0.50 and 0.45 per cent for broiler pre-starter, starter and finisher respectively). The experimental ration (R2) was prepared with 110 per cent of BIS (2007) recommended methionine (0.55, 0.55 and 0.50 per cent for broiler pre- starter, starter and finisher respectively). The experimental ration (R3) was prepared with 120 per cent of BIS (2007) recommended methionine (0.60, 0.60 and 0.54 per cent for broiler pre- starter, starter and finisher respectively). The group G1 and G2 were fed with ration R1. The groups G3 and G4 were fed with rations R2 and R3 respectively.The pre-starter feed was fed from day one to 7$^{th}$ day and from 8$^{th}$ to 21$^{st}$ day starter feed and 22$^{nd}$ to 42$^{nd}$ day finisher feed was given in all groups of birds.

### Sample collectionand Carcass study

On 42$^{nd}$ day four birds from each group were randomly selected, weighed and starved for 12 hours and slaughtered.The carcass weight was calculated by removing the feathers, blood, head, feet, and organs, except the lungs and kidneys. The carcass yield (carcass weight relative to live body weight) and yield of breast muscle (breast muscle weight relative to carcass weight) was calculated (Choo*et al*., 2014).

### Statistical Analysis

All data were analyzed by one-way ANOVA using SPSS statistical software (ver.16.0 for Windows, SPSS Inc., Chicago, IL). The differences were considered to be significant at $P < 0.05$. The data are presented as means and SEM.

## RESULTS

### Carcass and Breast Yield

Broiler chicks with low hatch weight (LHW) (35.0-38.0 g) also achieved live body weight compared to average hatch weight chicks (AHW) body weight at 42nd of its age. The higher per cent of carcass yield of 77.58 was recorded in LHW chicks supplemented with 110 per cent compared against 74.81 per cent in 120 per cent supplemented group.

## DISCUSSION

Methionine is an essential amino acid for broilers which involves in protein synthesisand dietary methionine supplementation show improvements in growth performance have been widely observed in chicken.The day-old hatch weight also one of the factor to influence the growth performance of broilers.However, the level of supplementation differs as per recommendations of NRC (1994) or BIS (2007). The results of this study is in agreement with findings of Ahmed and Abbas (2011) who reported that methionine at 0, 100, 120 and 130 per cent of NRC recommendations proved there was an increase in absolute weight and relative weight of breast, but no significance difference between groups.

Wen *et al*. (2014) reported supplementation of methionine at 0.60 and 0.53 per cent in broiler starter and finisher with increased level of methionine diets respectively,better the relative weightof breast muscle among the organs tested. In our study, the level of methionine in broiler pre starter, starter and finisher rations were 0.50, 0.50 and 0.45 per cent respectively shown increased breast muscle in LHW chicks compared to AHW chicks and increase in methionine level not shown significant improved in breast muscle yield. Methionine promotes broiler growth by regulating the developmentof breast muscle. The finding observed in this study is consistent withthe data of Zhai*et al*. (2012), who reported that breastmeat yield was greater in 42nd day broilers fed diets with0.51 per cent Methionine than that of broilers fed diets with 0.41 per cent Methionine from 22 to 42 day of its of age. This may be due to increasedmuscle protein deposition induced by Methionine (Nagao *et al*.,2011; Zhai*et al*., 2012).Jianlin*et al*. (2004) reported that, on carcass traits at different ages had no significant interactions were observed between lysine and methionine for any parameter when both lysine and methionine were fed equal or in excess of NRC (1994) recommendations.

## CONCLUSION

The present study results suggest nosignificant interactions between methionine and low hatch weight chicks when they were supplemented with equal or in excess of BIS (2007) recommendations. The results imply that peopleformulating diets on the “Ideal Protein” basis need not elevate the level of methionine even the day old-chicks weightare less than average weight. The BIS (2007) recommendations considered minimum requirements of methionine.

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**Table 1. Effect of dietary concentration of methionine on carcass characteristics with different hatch weight broiler chicks**

| Attribute | GROUP | | | | SEM | P Value |
|---|---|---|---|---|---|---|
| | G1 | G2 | G3 | G4 | | |
| Live weight (g) | 2327.60 ± 77.24 | 2298.00 ± 70.03 | 2267.60 ± 106.89 | 2262.80 ± 39.57 | 172.70 | 0.927 |
| Carcass Weight (g) | 1773.80 ± 82.46 | 1768.20 ± 88.68 | 1756.00 ± 70.25 | 1694.00 ± 53.36 | 167.51 | 0.867 |
| Carcass yield (%) | 76.09 ± 1.41 | 76.76 ± 1.76 | 77.58 ± 1.28 | 74.81 ± 1.32 | 3.26 | 0.596 |
| Breast weight (g) | 494.20 ± 16.77 | 442.80 ± 24.77 | 500.40 ± 16.35 | 473.00 ± 26.09 | 47.99 | 0.261 |
| Breast Yield (%) | 28.03 ± 1.28 | 25.62 ± 2.94 | 28.76 ± 1.75 | 28.03 ± 1.73 | 4.519 | 0.715 |
| Giblet weight (g) | 106.00 ± 5.54 | 98.00 ± 3.35 | 98.20 ± 6.35 | 94.00 ± 3.58 | 10.899 | 0.393 |
| Giblet Percent (%) | 6.07 ± 0.59 | 5.65 ± 0.53 | 5.61 ± 0.34 | 5.57 ± 0.26 | 1.003 | 0.844 |

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RESEARCH ARTICLE

# Detection of the Main Mycotoxins in Feed of Horses in Al-Zawra'a Park and Study their Effects on Hematological Feature

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## ABSTRACT

The aim of the current study is to detect fungi and mycotoxins presence in horse's feed and to determine the effect of these mycotoxins on haematological feature in these animals. Twenty five samples of horse's grains were obtained from Al-Zawra'a Park in Baghdad during the period of November 2016 to April 2017 for isolation of fungi, by using Sabouraud dextrose Agar and incubated at 28±2 ºC for (3-7) days. Then, detection of mycotoxin by Elisa in two seasons, winter and summer. Blood samples were collected from 20 horses in Al-Zawraa Park in Baghdad for determination of Red blood cell count, Hemoglobin concentration, packed cell volume determination, Total leukocyte count and differential leukocyte count for lymphocytes and neutrophils by using device for haematological analysis. The present study reported 92% of fungal contamination in the grains of horses that collected from Al-Zawra'a Park. and demonstrated highly significant variation between seasons with ($P<0.01$) by expression the mycotoxin contaminated feed of horse is very common in Summer than in Winter particularly with Ochratoxin A. Heamatologically, the study showed highly significant difference in 20% of horses suffering from anemia (6.38 ± 0.72, 8.25 ± 0.59, 24.70 ± 2.07 for RBCs,Hb and PCV respectively) and 100% suffering from leukopenia with ($P<0.01$). In conclusion, routine inspection of horse feeds should be done for fungal growth. Fungi identification procedures and tools in addition to valid testing can be used to detect the potential of ochratoxin A, besides, monitoring of the presence of fungi and mycotoxins in foods of animal is very important as considerations of their hazards for health.

**Keywords:** Mycotoxins, horses feed, Hematological paramaters, Ochratoxin A, Iraq.

## INTRODUCTION

Mycotoxins are considered to be secondary metabolites that are structurally have diverse toxic ability and usually produced by a wide range of moulds which infesting different agricultural commodities [1, 2]. It was found that moulds that contaminate food grains not only cause spoilage, but also they can turn them to be toxic as a result of mycotoxins elaboration that are known to be important bio-agents and are responsible for variety hazards of man and animal health [3,4].Mycotoxins, as it is known, are present in environments where they are developed from. Such environments are feed, cereals and food that are based on grain. It was found that high substrate humidity which is usually associated with the processing, ambient temperature, type of food and production or storage are considered to be the main environmental factors that contribute to the mycotoxins occurrence [5,6].Bacelar *et al*., (2016)[7] mentioned that the most significant mycotoxins in food industry and agriculture are from the six classes that are widely spread in human food and animal feed. These classes are: ochratoxins (OTs), aflatoxins (AFs), zearalenone (ZEN), T2 toxin, deoxynivalenol (DON) fumonisins (FBs) and Patulin .These mycotoxins have various effects that are rendered them to be hepatotoxic, carcinogenic, neurotoxic, nephrotoxic, and teratogenic. Besides, in according to the type of mycotoxin involved, they can impair immune system [8,9,10].

Furthermore, different mycotoxins or more than one mycotoxin can contaminate the typical feed of animal that usually formed from several sources. These mixed feeds may possess all the mycotoxins that found in several individual ingredients [11].liver physiological, biochemical and haematological changes, in addition to animals growth depression may be induced by the consumption of diet that is contaminated with multiple mycotoxins [12].Many studies had demonstrated that swine, cattle and poultry feeds had been extensively screened for mycotoxins [13]. On the other hand, few researches had been focused on mycotoxins in the foods of horse because of the rare interest of the productivity of such animals. So, this research was performed to detect fungi and mycotoxins presence in horses feed and to determine the effect of these mycotoxins on haematological features in these animals.

## MATERIALS AND METHODS

### Source of samples

The samples of horses' grains had been collected from Al-Zawraa Park in Baghdad during the period of November 2016 to April 2017. Twenty five samples with 150 gram were taken and each one was divided into 3 parts, so the total cultivated samples were 75. Each sample subjected to surface sterilization by 2% sodium hypochloride for isolation of fungi by using Sabouraud dextrose Agar (BDH England) then incubated at 28±2 ºC for (3-7) days according to [14].

### Detection of mycotoxins in the samples of horses' grains

The other parts of sample were used for detection of mycotoxin by Elisa technique in Veterinary Directorate - Central Veterinary Laboratories and researches Department- Public health and food safety Laboratory in two seasons, winter and summer.

### Hematological parameters analysis

Blood samples were collected aseptically from jugular vein of 20 horses in Al-Zawra'a Park in Baghdad and using EDTA-contained tubes for determination of haematological parameters.Red blood cell count, packed cell volume, Hemoglobin concentration, Total leukocyte count and differential leukocyte count for lymphocytes and neutrophils were determined by using device for haematological analysis called Human Count 60 VET(Germany).

### Statistical Analysis

The Statistical Analysis System- SAS [15] program was used to effect of difference factors in study parameters. Chi-square test was used to significant compare between percentage and T-test was used to significant compare between means in this study.

## RESULTS

### Results of fungal isolation

The present study reported high percentage of fungal contamination in the grains of horses that collected from Al-Zawraa Park in Baghdad which reached to 92% represented by isolation of 69 fungal isolates from the total cultivated samples were 75. These isolates were classified into 6 types of important molds as shown in table (1).

### Detection of mycotoxins in the samples of horses' grains

All samples of commercial horse feed were analysed quantitatively by Elisa technique for detection three different mycotoxins which are Aflatoxin B1, Ochratoxin A and T2 toxin in two seasons, winter and summer. The results demonstrated highly significant variation between seasons with ($P<0.01$) by expression the mycotoxin contaminated feed of horse is very common in Summer than in Winter (Table 2) particularly with Ochratoxin A, While the levels of other two mycotoxin were lower than the critical or even toxic level.

### Hematological parameters analysis

Twenty horses were taken to detect the effect of mycotoxins on their haematological feature. These animals were divided according to the sex as in table 3.The current study expressed highly significant difference ($P<0.01$) in 20% of horses suffering from anemia (table 4) through detection the number of circulating erythrocytes, packed cell volume and hemoglobin concentration which revealed to decrease significantly($P < 0.05$) when compared with control animals(table 5).On the other hand, There was a marked significant reduction in total leukocytes count ($P<0.01$)which was observed in all animals that fed on contaminated grains with 100% (table 6), Whereas there was a significant changes observed between differential leukocyte count in neutrophil and lymphocyte which appeared as increase in neutrophil and a decrease in lymphocyte counts ($P<0.05$)when compared with control group (table 7).

## DISCUSSION

The main two fungi groups of the major concern in the equine world are field fungus *Fusarium*, that has the ability to produces zearalenone, fumonisin and T2, and the storage fungus *Aspergillus*, that is responsible for aflatoxin and ochratoxin production [16]. Aflatoxicosis with its deleterious toxicological effects in horses had been focused on in many studies according to their risk [17, 18]. While [7] highlighted on the Equine leukoencephalomalacia (ELEM) which is demolished neurologic system of equidae that is attributed to contaminated corn consumption with one or more types.According to our knowledge, there is little or no studies were carried out to detect the impact of food-contaminated mycotoxins on the health and particularly haematological features in Iraqi horses in Baghdad. So the current study was performed to detect 3 types of mycotoxins in feed of horses but the result express only Ochratoxin A (OTA) has significant value when compared with limited value (2ppb) in the summer season whereas Aflatoxin B1 and T2 toxin expressed below the limited level (20ppb and 150ppb) respectively inspite of the Aflatoxin showed in near with limited value. This result may be attributed to environmental conditions efficacious and nutritional

necessities for growth and production of OTA which in constant with [19] how investigated the impact of temperature and relative humidity on production of ochratoxin A and found that the relative humidity has less significant role than temperature in OTA production. which elucidate the finding of the present study about OTA production in summer increase significantly. Morever, Ali *et al*., (2013)[20] studied the effect of varies degrees of temperature on OTA production in common cereals by Aspergillus spp. and the results showed the production is temperature dependent and 30ºC was the most favorable temperature to produce significant quantity of OTA as compared to other temperatures.

On the other hand, Hashem *et al*., (2015)[21] reported the formation of OTA from *Aspergillus carbonarius* had relationship with nitrogen, carbon sources and water activity. Dhanasekaran *et al.,* (2011)[22] pointed to the maximum temperatures for Aflatoxin production have been reported to be 40-42$^0$C that may be explain the finding of this study in which the Aflatoxin B1appear lower than the embarrassing level, Also, it was found that when Aflatoxin B1 mixed with OTA in feed contamination, OTA prevents the major impact of Aflatoxin then decrease the chance diagnosis of aflatoxicosis in feed.Concerning to the sex of animal, the present study showed that the dealing with 60% of animals were mares. This subject has major importance because it was found that teratogenicity caused by natural occurrence of OTA and it's transferring through horse placenta as firstly discovered by [23] who mentioned that the more sensitive animals were monogastric species. This return to the OTA kinetic which vary between horses and ruminant in which the OTA degraded in rumen by enzymes while in monogastric like horse, OTA is absorbed from the gastrointestinal tract without, or with little, prior degradation as mentioned by [24, 25].

The current study aimed to investigate the effect of OTA on haematological feature in horses and the results showed 20% of animals have anemia due to marked decrease in number of circulating erythrocytes, packed cell volume and hemoglobin concentration significantly. This finding is in line with [26] who found that the ochratoxin A induced Iron deficiency anemia and does not cause syndrome of hemorrhagic anemia, whereas the anemia usually caused by deficiency of nutrition that resulted from mycotoxin. While in poultry, it is documented that the OTA cause anemia as reported by [2].Regarding to leukopenia in this study was observed in all tested animals that fed on contaminated grains with 100%, this result agree with [27] who revealed that white blood cells number was decreased when mice fed on OTA due to spleen and lymph nodes necrosis which in turn, led to decrease lymphocytes proliferation and production of cytokines.In addition, feeding OTA contaminated diets may increase the severity of secondary infections in animals causing increase in neutrophil count [28] which in constant with the present result.

## CONCLUSION

As a conclusion, routine inspection of horse feeds should be done for fungal growth. Fungi identification procedures and tools in addition to valid testing can be used to detect the potential of ochratoxin A, besides, monitoring of the presence of fungi and mycotoxins in foods of animal is very important as considerations of their hazards for health.

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**Table 1: No. of samples that give +ve result for fungal contamination**

| Spp. | No. | Percentage (%) |
|---|---|---|
| *Penicillium spp.* | 31 | 44.93 |
| *Alternaria spp.* | 16 | 23.19 |
| *A. ochraceus* | 10 | 14.49 |
| *A. flavus* | 8 | 11.59 |
| *Mucor spp.* | 2 | 2.90 |
| *Rhizopus spp.* | 2 | 2.90 |
| Total | 69 | 100% |
| Chi-Square ($\chi^2$) | | 12.438 ** |
| ** (P<0.01). | | |

**Table 2. Detection of Ochratoxin A in winter and summer season (PPb)**

| Season | No. | Mean ± SE |
|---|---|---|
| Winter | 4 | 2.55 ± 0.19 |
| Summer | 14 | 19.62 ± 1.07 |
| T-Test | --- | 4.217 * |
| * (P<0.01). | | |

**Table 3. Division of animals according to sex.**

| Sex | No . | Percentage (%) |
|---|---|---|
| Male | 8 | 40.00 |
| Female | 12 | 60.00 |
| Total | 20 | 100% |
| Chi-Square ($\chi^2$) | --- | 8.250 ** |
| ** (P<0.01). | | |

**Table 4. Distribution of cases according to Anemia**

| Cases of Anemia | No . | Percentage (%) |
|---|---|---|
| +ve | 4 | 20.00 |
| -ve | 16 | 80.00 |
| Total | 20 | 100% |
| Chi-Square ($\chi^2$) | -b-- | 13.00 ** |
| ** (P<0.01). | | |

**Table 5. Effect of ochratoxin A on parameters associated with anemia**

| The Group | Mean ± SE | | |
|---|---|---|---|
| | RBCs (x $10^{12}$) | Hb (g/dl) | HCT (%) |
| Cases | 6.38 ± 0.72 | 8.25 ± 0.59 | 24.70 ± 2.07 |
| Control | 8.70 ± 0.50 | 13.80 ± 1.60 | 37.10 ± 4.20 |
| T-Test | 1.273 * | 2.884 * | 6.603 * |
| * (P<0.05). | | | |

**Table 6: No. and percentage of cases suffering changing in WBC, Lymphocyte and Neutrophil**

| Parameters | Total No. | Decreased No. (%) | Increased No. (%) | Chi-Square ($\chi^2$) |
|---|---|---|---|---|
| WBCs | 20 | 20 (100%) | 0 (0.00%) | 15.00 ** |
| Lymphocyte | 20 | 14 (70.00%) | 6 (30.00%) | 10.75 ** |
| Neutrophil | 20 | 3 (15.00%) | 17 (85.00%) | 13.63 ** |
| ** (P<0.01). | | | | |

**Table 7. Compare between differential count of Lymphocytes and Neutrophill with control**

| The Group | Mean ± SE | |
|---|---|---|
| | Lymphocytes (%) | Neutrophill (%) |
| Cases | 18.56 ± 4.17 | 72.08 ± 6.22 |
| Control | 61.00 ± 5.00 | 33.00 ± 2.94 |
| T-Test | 12.461 * | 17.935 * |
| * (P<0.05). | | |

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RESEARCH ARTICLE

# Soil Water and Salt Movement and the Effect of Desalination on Saline Soil under Different Irrigation Amount

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## ABSTRACT

In view of the fact that the soil salinity in the coastal reclamation area of Jiangsu province is too high, the desalting time is long, and the desalting effect is not obvious, this article analyzes the effect of different irrigation treatments on soil temperature, moisture, and salinity, and analyzes the salt rejection rate. The results showed that the regularity of the changes in soil moisture treated with different amounts of irrigation has some similarities and differences. When the amounts of irrigation were 240L, 200L and 160L, the soil moisture content in each soil fluctuated among 18.0~48.9%, 16.09~48.26% and 13.37~47.99% respectively. The salinity content of 0~55cm soil layers dropped below 1.0 $g \cdot kg^{-1}$ after the treatment of 240L irrigation. Excluding the negative desalination rates of 170cm and 200cm soil layers, the desalination rates of other irrigation treatments were all above 41.16%, especially the plowing layer reached more than 87.31%; After the treatment of 200L irrigation, the soil salinity content in 0-25cm soil layer was dropped below 1 $g \cdot kg^{-1}$, the salt rejection rate was over 95.81%, and the soil salinity content in other soil layers were above 1.0 $g \cdot kg^{-1}$. After the treatment of 160L irrigation, the soil salinity content in 0~25cm soil layers dropped below 1 $g \cdot kg^{-1}$, and the soil salinity content in other layers were above 1 $g \cdot kg^{-1}$. Excluding the negative desalination rates of 170cm and 200cm soil layers, the soil desalination rates of the remaining layers reached between 17.26% and 94.19%. It provides a certain basis and support for the irrigation and desalination technology of saline-alkali soil in the coastal area of Jiangsu Province.

**Keywords:** Coastal area; Irrigation amount; Saline-alkali soil; Salinity content; Desalination

## INTRODUCTION

The coastal shoal is generally refers to the muddy coast intertidal zone (Xiong Wanying, 2004), mainly distributed in areas where land and sea meet,and it is mainly produced in areas where coastal areas are smooth, where sediments accumulate, and where the impact of waves is less active (Wang Ying, 1983). The intertidal zone is the core area along the beach and is mainly divided into two parts, the upper and lower subtidal zone. The area of tidal flats in China is also relatively wide. The coastal line of the beach is up to 4000 km from north to south and from Liaoning to the coastal areas of Guangxi province. The area of coastal beach resources in Jiangsu is about 6520.6 $km^2$, accounting for 1/4 of the total area of China's beach, which is the first in coastal provinces of China (Chen Fang, 1998). The total area of Yancheng City of Jiangsu Province is 16973 $km^2$ and the area of the beach is 6830000 mu. The rational and effective development and utilization of coastal beach resources can not only increase the cultivated land area, but also increase the balance of cultivated land requisition compensation, ease the contradiction between less land occupations per capita and promote the development of agricultural economy in Jiangsu province (Hao Shurong, 2009). However, the soil salinization in China's coastal areas is increasingly severe, with heavy salt content in the soil, shallow groundwater levels, and the infrastructure of water conservancy facilities, soil and water conservation and other engineering measures are not well-enhanced, the silting of river channels is serious, irrigation and drainage facilities are not complete, which exacerbates the soil salinization and further affects the development of coastal beaches.Faced with so many uncertainties, how to effectively develop coastal tidal flat, reasonable saline alkali land and make coastal tidal flat become fertile fields is a problem that we need to solve urgently.Therefore, we have conducted research on the effect of irrigation and desalination of saline-alkali soil in coastal reclamation areas in Jiangsu Province.

At present, there are a large number of studies on irrigation and salt washing at home and abroad, such as Bresler (E. Bresler, 1971) study showed that when the soil salt drip irrigation to drip irrigation belt on both sides and bottom gradually increased, and the distribution of soil salinity changes with the dripper discharge and irrigation amount. Ayars (Ayars J E, 1991) and other studies showed that soil salt content of daily irrigation is much lower than irrigation in 3 to 4 days. Yakirevich (A Yakirevich, 2013) studied the effect of leaching of salt in aquifers and the important effect of irrigation water reflux on the salinization of groundwater in the irrigated area. Bveese (Bveese F, 1985) showed that saline water drip irrigation can effectively save water compared with other irrigation methods, and under the same irrigation volume, the degree of salinization of drip irrigation is significantly lighter than that of sprinkler irrigation. Yang Jianjun (2004) showed that salt washing with irrigation can effectively reduce the accumulation of nitrate nitrogen and reduce the accumulation of salt in the soil (Yuan Qiaoxia, 2008; Cao Hongxia, 2003).Li Wei (Li Wei, 2011) showed that after irrigation, $K^+$ showed a "single peak" characteristic in profile distribution. $Ca^{2+}$ was well coupled with moisture and temperature in all soil layers, and both had synergistic effect on migration of $Ca^{2+}$. Du (Du Lei, 2014) also showed that the water and salt transport in 0-20cm soil layer varied greatly after irrigation, and the soil salt content and soil moisture and temperature in 0-5cm and 40-50cm soil layers were well coupled. Zhao (Zhao Chengyi, 2009) found that the peak value of soil salinity decreased with the increase of irrigation volume, and the effect of desalination was directly proportional to the amount of irrigation. Li (2014) carried out a numerical simulation study of soil water and salt transport based on Hydrus-1D. The results showed that the salt accumulation areas treated with irrigation volumes of 1.4 L, 2.8 L and 4.2 L were located at 20 cm , 30 cm and 40 cm respectively. The salinity rates in the salt accumulation areas were 49.2%, 57.6%, and 100% respectively and the desalination rates of surface soil were 87.2%, 93.7%, 96.8% respectively. Yin Jiandao (2002) showed that with the increase of irrigation volume, the soil salt content experienced a sharp decline and slowly declined until a relatively stable dynamic process. The desalting trajectory conformed to the power function equation.The study of Wang (Wang Ruoshui, 2014) shows that the high frequency irrigation can effectively enhance the leaching of soil salt in the plough layer, and effectively inhibit the soil salt back.

This study aims at the status of severe salinization in the coastal area of Dongtai, Jiangsu Province, by carrying out the test of saline soil desalting under different irrigation amount, analysis of the effect of irrigation on soil temperature, moisture, salinity and desalination efficiency, predict the desalination rate and duration which the soil reach the normal growth of crops, define the correlation between the amount of irrigation and irrigation calendar time, analysis of effects of soil water salt dynamic factors and influence mechanism, put forward irrigation techniques and methods suitable for coastal reclamation area of Jiangsu heavy saline soil desalting, the result have a great significance to our coastal mudflat development.

## MATERIALS AND METHODS

### Experimental site

The experiments are initiated in Jiangsu Province Hydraulic Research Institute coastal test base(latitude 32°51'11″N, longitude 120°53'45″E), a test base close to west coastal highway, about 5-6 km from the coastline. The test belonged to the subtropical and warm temperate monsoon with significant four seasons, rainfall concentrated in hot rainy season, spring temperature changes, an invigorating autumn climate sunshine. The annual average temperature is 15.0 □, rainfall is 1061.2 mm, and 2130.5 hours of sunshine.

### Experimental design

The experiment set 3 vapour permeability testing pits, each testing pits of about (length×width×depth is 2m×2m×2m), the dry density of the soil in surface(0-100cm) is 1.48g/cm$^3$, the bottom(101-200cm) is 1.5g/cm$^3$, and the 0.3m gravel filter layer is laid at the bottom of the testing pits. In addition, the computer control room, the water table control room and the underground observation corridor are also set up to monitor the transmission data. Each test pits in 10cm, 25cm, 55cm, 80cm, 110cm, 140cm, 170cm and 200cm were buried sensors, respectively, among them, pit buried temperature, salinity and moisture sensors were used to measure the value of real-time monitoring, each test pit soil layers of temperature, moisture and salinity and irrigation. The soil in the test area is coastal saline soil, the soil is mainly composed of light loam or loam. The physico-chemical properties of the soil are shown in Table 1.The experiment set up 3 vapour permeability test pits, that is, 3 treatments and 1 variable factors. Among them, running water was irrigated and irrigation volume was set at 3 levels, 240L, 200L and 160L, respectively, and 1 sets of groundwater level were set up, is 1.5m, irrigation frequency is 4 days, and irrigation with small water frequency. The experimental treatment is shown in Table 2.

### Main tested indices and methods

The study was adopted real-time data acquisition system to measured the data, it included a Campball TDR soil moisture acquisition system, soil temperature monitoring system, testing-pit underground water level monitoring system, underground water level automatic water supply and drainage system. The automatic soil moisture acquisition system adopts the Campbell company's water automatic monitoring equipment. The soil salinity and electrical conductivity are monitored and transmitted by the corresponding sensors embedded at various levels. Among them, the soil temperature is monitored by sensors buried at various levels, the soil moisture content is monitored by sensors buried at various levels, and the soil salinity is monitored by sensors buried at various levels.

#### Statistical analysis

Data analysis and drawing are carried out by Excel 2007.

## RESULTS AND DISCUSSION

**The change of soil water vertical distribution under different irrigation amount**

Figure 1 is the characteristics of soil water change in different depths under the 240L(a), 200L(b) and 160L(c) irrigation amount. It can be seen from Figure 1, the change of soil moisture content varies under different irrigation conditions. Under different treatments, both changes of shallow soil moisture content is high, the amplitude is maximum, it mainly because of the impact of the soil is not only affected by irrigation, and is also affected by climate; The deep soil moisture content is relatively shallow changes slowly, because the layer is less affected by climate, mainly due to the effect of irrigation. It also can be seen from Figure 1, under the different irrigation condition, the moisture is mainly concentrated in the 0-110 cm soil layer, which is mainly affected by the combined action of the surface irrigation water and the simulated groundwater. After irrigation, the soil moisture moves downward due to the action of gravity gradient and matrix potential gradient, the upper layer irrigated the downward layer and the groundwater moved upward through the capillary rising action. At the same time, the soil water content in the surface layer decreased, forming a clear crust, and the water loss rate decreased.

Figure 2 is the changes in the water content of 0~200cm with different irrigation amount which is before irrigation, 1d after irrigation, and one irrigation period. The characteristic curves of water change showed that 0~80cm had the highest soil water content with 240L irrigation amount, then the 200L irrigation amount, and the 160L irrigation amount was the lowest, especially after irrigation 1d. However, the soil moisture content of 80~200cm layer fluctuated greatly, after irrigation, the 200L treatment was the highest, then 160L, and 240L were the lowest, which is because the deep soil was mainly affected by groundwater level. It also can be seen from figure 2, water redistributed across 1d after irrigation, the moisture content increased again with the increase of soil depth. After 1d irrigation, the soil moisture content of 0-80cm showed 240L> 200L> 160L. After 4d irrigation, the soil water content of different treatments showed an increasing trend, and the difference between them was relatively small, the change was mainly in 110~200 cm, and 160L treatment increased first and then decreased, and all of them finally showed 200L> 160L> 240L. The irrigation amount is the basis for determined the desalination system. Therefore, it is a great importance to study soil water movement under different irrigation conditions for designing desalination system of saline alkali soil rationally.

**The change of soil salt vertical dynamic under different irrigation amount**

Figure 3 is the vertical change of soil salt under the irrigation amount of 240L (a), 200L (b) and 160L (c) respectively. It can be seen from the overall trend, after 6 times irrigation leaching, the coastal saline soil in the desalination process, the overall trend is similar: in the leaching process, soil salt content of 0~140cm was decreased, have a obvious desalinization; But 170cm and 200cm soil layers in the whole the soil salt content increased, and it has a clear salt accumulation phenomenon. The soil salt content showed a similar trend, shallow soil salinity showed 0~25cm sustained and rapid decline, salt has been leaching down, showing irrigation pressure salt phenomenon, part of salt with water discharge; 110~140cm deep soil salinity increases slowly at first, then decreased; 170cm and 200cm soil salt content due to the impact of underground water level, resulting in a large number of salt, salt content not declined, but increased. During the days after drainage to the next irrigation, the soil salinity will rise to the surface with the capillary force of soil under evaporation, and it will accumulate to the surface of the soil with time and return to salt. However, with the increase of the irrigation times, the salinity of the surface and the lower layer decreased synchronously.

It also can be seen from figure 3, the salt distribution in the soil profile after irrigation is similar to the water distribution, and the change is mainly concentrated in the 0-140 cm soil layer. Before irrigation, the soil salt content in different treatments were at a relatively high level, among them, the irrigation amount of 240L and 200L treatment

was the most obvious, that has the obvious effect of salt leaching irrigation is 240L and 200L, while the amount of irrigation 160L the soil salinity fluctuations, this may be because the smaller amount of irrigation, in the period of time after irrigation, there was a phenomenon of returning salt. After the first irrigation, the soil salinity dropped sharply, the irrigation of 240L and 200L was the most obvious, decreased -0.87~28.78% and -14.03~31.62%, the shallow layer is larger, 80~140cm decline is small, and 170cm and 200cm appeared a large amount of salt, salt drop is negative, compared with the initial value content, the salt content is increased. When the irrigation amount was 160L, the 0-10cm decreased the largest, reached 34.62%, and due to the irrigation amount is small, 55cm, 80cm, 140cm, 170cm and 200cm all returned to salt, and the salt content increased, the other layer had no salt accumulation due to the accumulation of salt in the deep soil. After the second to fourth irrigation, 0~140cm soil continued to decline, 170cm and 200cm soil salt content was still slowly rose, as 240L and 200L is obvious, there have been significant fluctuations in irrigation amount was 160L with different irrigation cycle, mainly because of the amount of irrigation is smaller, the soil salt by evaporation is larger. After the fifth irrigation, soil salinity in the 0~55cm soil layer of 240L irrigation amount has been reduced to 1.0 $g \cdot kg^{-1}$, the soil desalination rate reached more than 89.79%, 80cm, 110cm soil desalination rate has reached more than 70.88%, while 140cm, 170cm and 200cm soil desalination rate is relatively small, only between 10.78%~40.49%. 200L irrigation the soil salt content decreased above 1.0 $g \cdot kg^{-1}$, and 160L irrigation the soil salt content were between 2~3 $g \cdot kg^{-1}$, that indicated that under the same irrigation times, the optimal desalination effect is 240L treatment, and the worst desalination effect is 160L treatment. After the sixth irrigation, 0~140cm soil salt content continued to decline of 240L irrigation treatment, in addition to the 170~200cm soil layer, each layer of both the desalination rate has reached more than 50%; Just 0~25cm soil salt content decreased to 1 $g \cdot kg^{-1}$ of 200L irrigation treatment, the rest of the soil is still above 1 $g \cdot kg^{-1}$; Only 0~10cm soil layer soil salt content decreased to 1 $g \cdot kg^{-1}$ of 160L irrigation treatment, and other layers of soil salinity is between 1~3 $g \cdot kg^{-1}$. After irrigation, the soil salt content in soil 0~140cm relative to the sixth irrigation was continuing to decline of 240L irrigation treatment, while 170cm and 200cm was increased, the 0~55cm soil salt content decreased to 1 $g \cdot kg^{-1}$, and other layers of soil salt content in 1 $g \cdot kg^{-1}$ is still the best, shallow the soil salt has the best leaching effect, desalination rate is relatively high, reached more than 87.31%, while the deeper soil desalination rate is negative, appeared in the soil salt accumulation, salt content increased significantly.

The 0~25cm soil salt content is reduced to 1 $g \cdot kg^{-1}$ of 200L in irrigation treatments,, the rest of the soil was above 1 $g \cdot kg^{-1}$, among them, 0~25 cm soil desalination rate reached more than 95.81%, 25~140cm soil desalination rate between 27.59%~78.89%, 170cm and 200cm soil salt content as a whole the rise, the desalination rate is negative, the soil showed shallow desalting rate is higher, while the deep desalting rate is low, the desalination rate is negative, the salt content increased, which is mainly affected by the amount of irrigation amount and underground water influence. The soil salt content of soil in 0~25 cm decreased to 1 $g \cdot kg^{-1}$ of 160L irrigation treatment, the other soil salt content was above 1 $g \cdot kg^{-1}$, the desalination rate between -3.58%-94.19%, in addition to the shallow soil, the desalination rate is not high, indicated that the irrigation amount of 160L although salt can achieve certain effect, but the overall poor effect of desalination.

### Effect of different irrigation on soil salt-leaching

In order to achieve both water and soil irrigation desalting can achieve good effect, according to the formula (1) will be calculated under different irrigation treatments of different soil desalination rate were summarized, and then calculate the normal salt content at different soil and different soil depth under irrigation water, to provide some theoretical guidance for the actual production.

$$L = \frac{S_1 - S_2}{S_1} \times 100\% \qquad (1)$$

Where, L is rate of desalination (%), $S_1$ is the initial salt content ($g \cdot kg^{-1}$); $S_2$ is teh final salt content ($g \cdot kg^{-1}$).

It can be seen from Table 3, the irrigation of 240L can make the soil salt content falls below 1.0 $g \cdot kg^{-1}$ of 0~55cm, reduced non saline soil, the rate of desalination reached more than 87.31%; the strength of saline soil 80cm and

110cm falls below the light salinized soil, the rate of desalination reached more than 63.39%, indicating that the irrigation of 240L has better irrigation desalting the effect of 0~110cm soil, and the rate of desalination is high, and the 140cm soil desalination rate was 41.16%, also played a certain effect of desalination, and for 170cm and 200cm bottom soil desalination rate is negative, which is mainly in the underground water level below soil produced salt accumulation, and is affected by the underground water level, salt is difficult to discharge, so the bottom soil salt content increased, the desalting effect is poor. The irrigation of 240L can make the soil salt content falls below 1.0 $g \cdot kg^{-1}$ of 0~25cm, reduced non saline soil, the rate of desalination reached more than 95.81%, among them, 55cm, 80cm and 110cm soil salt content is still above 1.0 $g \cdot kg^{-1}$, the desalting rate between 45.82%~78.89%, but the effect of soil desalting is poor, still belongs to light or moderate saline soil salinization; For the deep layer of 140cm desalination rate is only 27.59%, the desalting effect is poor, soil salt content is still at a high level; For 170cm and 200cm soil salt content as a rising trend, the desalination rate is negative, there is an obvious phenomenon of salt accumulation. The irrigation of 160L can can make the soil salt content falls below 1.0 $g \cdot kg^{-1}$ of 0~25cm, the rate of desalination reached more than 89.13%, while the 55cm, 80cm and 110cm in soil, the the rate of desalination is only 30.99%~65.31%, compared with 240L and 200L irrigation compared to desalination effect is even worse; For the 140cm soil desalination rate is only 17.26% that small changes in soil salt content; For 170cm and 200cm soil layer soil desalination rate is still negative, after irrigation of salt is the initial value of influence is still on the rise, 240L and 200L may also have a small amount of irrigation.

It can be seen from Table 3, the surface salt of irrigation is very vulnerable to irrigation. During the irrigation period, the surface soil desalted, the desalination rate of the surface layer is over 89.13% before and after washing, and the salt accumulation phenomenon can be well alleviated. With the deepening of the soil layer, the soil desalination rate was significantly reduced, the soil desalination effect was significant in the 0-80 cm soil after leaching, and the salt rejection was low at 110 cm and 140 cm. The soil desalination rate in the whole 0~140cm soil layer was positive, but the soil salinity content in 170cm and 200cm soil layers increased, the salt rejection rate was negative, and the soil salinity was higher in the bottom soil. Irrigation did not effectively reduce the accumulation of salt in the underlying soil. According to the law of water and salt transport, the salt in the soil mainly moves with the water. Under the conditions of water leaching, the surface soil salinity enters the deep soil and flows out through the drainage system. As the amount of irrigation decreases, the soil washes the salt, the ability to reduce, so 160L irrigation treatment salt worst. Therefore, to control the amount of irrigation water and groundwater depth can significantly control leaching soil salinity reduction, due to the groundwater can not be controlled in actual agricultural production, so we should control the amount of irrigation amount, accelerate the downward movement of surface water, leaching when water infiltration increased, enhanced salt leaching and thus slow down the accumulation of salt in soil.

In conclusion, under the different irrigation amount conditions, the greater the amount of irrigation, water and soil contact more sufficient,so that the leaching depth is deeper, and the more filtrate volume, a lot of soil salt with water discharged from the soil, so the desalting effect is better; when the irrigation amount is small, in the influence of evaporation and groundwater, to filter out the liquid volume less or even no filtrate discharge, easy to cause the upper soil layer lower poly salt phenomenon, the desalting effect is poor.

## The relationship between the soil desalination rate, the duration of irrigation and the amount of irrigation

Figure 4 is the relationship between the desalination rate and the duration of irrigation. When the irrigation lasts is the abscissa, except for 170cm and 200cm soil layer, the leaching efficiency increased rapidly first, then slowly with the increase of irrigation lasts, the 170cm and 200cm soil layer was mainly affected by groundwater level, and the overall decline trend. Irrigation of 240L treatment, 10cm, 25cm and 55cm soil desalination rate was significantly higher than other soil layers, the desalination rate was above 87.31%, and by the early irrigation affected, in continuous irrigation after 24d desalination rate tends to slow, mainly is the soil salt content has been reduced to 1.0 $g \cdot kg^{-1}$, therefor, after irrigation 24d, 0~55cm soil desalination rate tends to slow, and for the deep soil desalination

rate is relatively large variation, mainly was effected by evaporation, soil salt leaching, the accumulation and return of salt and other phenomena, caused by the large changes in salinity. For the 170cm and 200cm soil desalination rate was increased first and then decreased, early irrigation of salt with irrigation infiltration, the salt content had a brief declined, but due to the influence of underground water level, surface soil salinity infiltration to the bottom after difficult to discharge, so have accumulated a lot of salt, the salt content increased, and the soil desalination rate decreased; For 200L and 160L irrigation, 10cm and 25cm soil desalination rate is relatively obvious, the overall upward trend with irrigation duration, 55cm and 80cm also showed an upward trend, but the relative trend was relatively small, while 110cm and 140cm fluctuated greatly, mainly because their salinity decreased with irrigation and increased with the increase of evaporated salt. As for the 170cm and 200cm soil layers also affected by the groundwater level, the desalination rate was negative and the salt content increased. In conclusion, both the irrigation amount larger or smaller, desalination rate in a certain range of soil and irrigation duration is proportional to the duration as the irrigation continues, desalination rate increased, and the impact on the underlying soil mainly depends on the level of underground water, so it the have no significant relationship with irrigation duration.

Figure 5 is the relationship between the rate of desalination and the amount of irrigation. It can be seen from the figure, when the amount of irrigation as the abscissa, under different irrigation amount the soil desalination rate changes significantly, the 0~140cm soil desalination rate increases with the increase of irrigation amount, 170cm and 200cm soil desalination rate decreased with the increase of irrigation amount, among them, the increase of 10cm and 25cm soil layer trend the relatively slow, mainly shallow soil leaching not directly affected by the irrigation of salt accumulation, and different irrigation in shallow soil have the same desalination efficiency, so the desalination rate changed little, and for 55cm, 80cm, 110cm and 140cm soil layer soil desalination rate with irrigation amount increased relatively large, because of it has a larger impact by accumulation and return salt of deep soi, therefore, the amount of irrigation has obvious influence on it, and for 170cm and 200cm soil layer soil desalination rate decreased with the increase of irrigation amount, this is mainly because the underground water level in 150cm, the greater the amount of irrigation, the more salt leaching, and is affected by the underground water level, the bottom soil salt solution and difficult to discharge, so the salt accumulation is more serious, so the greater the amount of irrigation, and 170cm and 200cm soil desalination rate is small.

## CONCLUSION

The effects of different irrigation on soil moisture with the irrigation cycle changed gradually, and wavy slowly decreased, deep soil moisture is greater than the shallow soil, the irrigation amount was 240L, 200 L and 160 L, the soil water content fluctuates between 18.8~58.57%, 16.09~58.76% and 13.37~55.94%. Before irrigation, the average water content of each treatment was low. After irrigation, the 0-80cm soil profile was the highest with 41.79% irrigation amount, and reached the maximum of 240L. The lowest irrigation amount of 160L was 13.37%, it indicated that soil water content was directly proportional to irrigation volume. The different irrigation treatments of the soil salinity showed a similar trend, shallow soil salinity showed 0~25cm sustained and rapid decline, salt has been leaching down, showed irrigation pressure salt phenomenon, part of salt with water discharge; 110~140cm deep soil salinity increased first, then decreased. 170cm and 200cm soil salt content in the whole period increased. The greater the amount of irrigation water, the more complete the contact between the water and the soil, the deeper the leaching layer, the more the volume of the filtrate, the large amount of soil salt with the water discharge, the effect of desalination is better; The smaller the amount of irrigation, under the influence of evaporation and groundwater level, the depth of the leaching soil layer is limited, the volume of the filtrate is not even filtered out, it is easy to cause poly-salt in the lower layer and desalination in the upper soil, so the desalination effect is poor. With the increase of irrigation amount, the upper layer of soil quickly desalted, and the middle and lower layers enter the desalination stage, the formation, downward movement and disappearance of salt peaks in the profile reflect the dynamic characteristics of soil salt transfer from top to bottom. The deeper of the soil layer, the slower the salt peak descends and the longer the duration, so the more difficult it is to desalt.

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**Table 1.Physico-chemical properties of the soil at the experiment site**

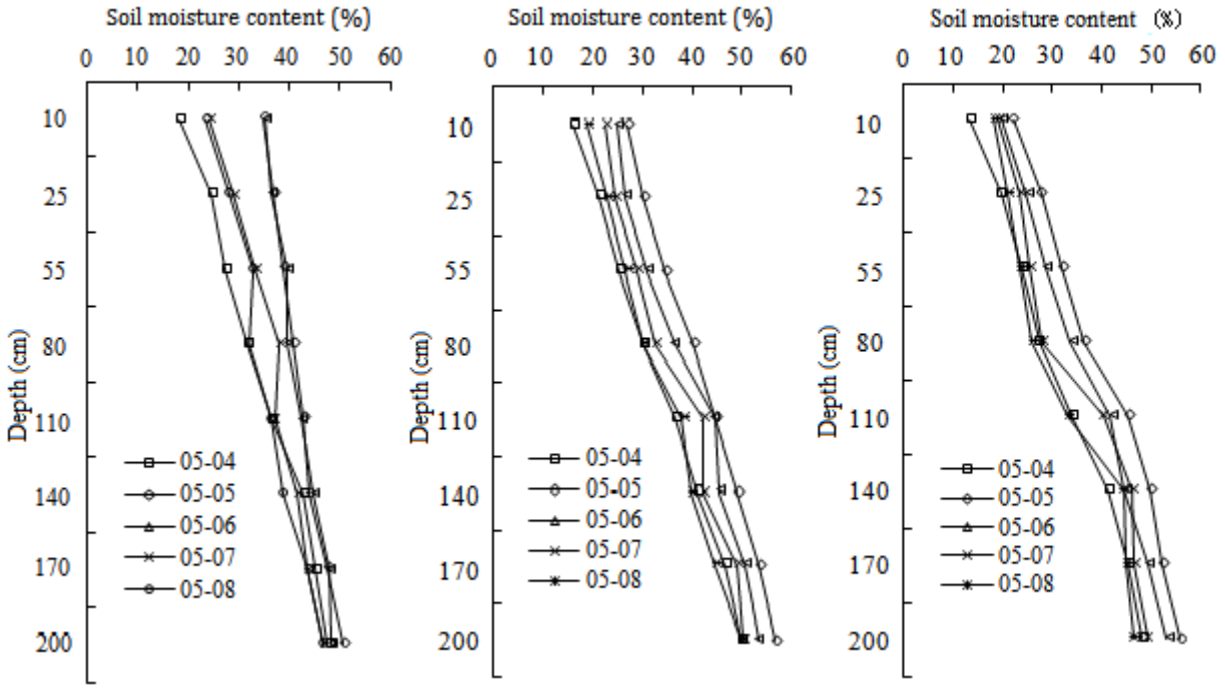
| Moisture content (%) | Volumetric weight ($g/cm^3$) | Soil total salt (g/kg) | Organic matter (g/kg) | Electrical conductance ($\mu s/cm$) | Grain composition (%, mm) | | |
|---|---|---|---|---|---|---|---|
| | | | | | >0.05 | 0.05-0.001 | <0.001 |
| 25.9 | 1.46 | 11.19 | 1.64 | $0.438\times10^4$ | 34.8 | 60.2 | 5 |

**Table 2.Experimental design**

| Testing pits | Irrigation amount (L) | Underground water level (m) | Irrigation frequency (d) |
|---|---|---|---|
| C1 | 240 | 1.5 | 4 |
| C2 | 200 | 1.5 | 4 |
| C3 | 160 | 1.5 | 4 |

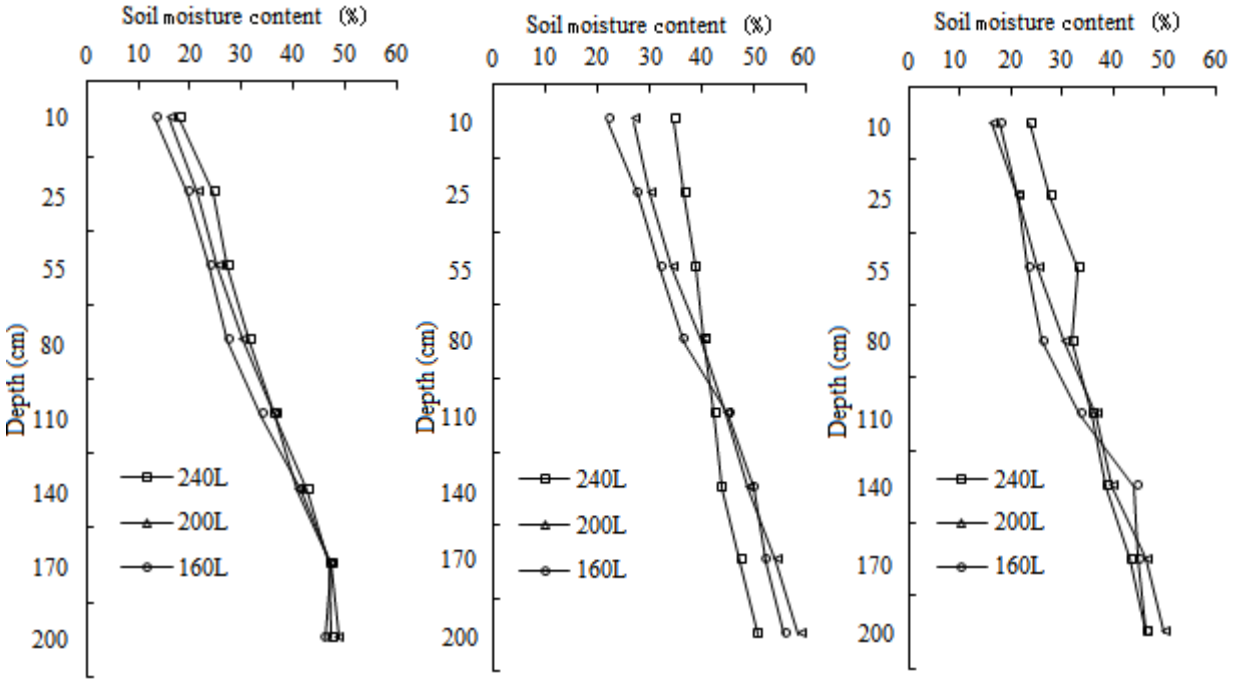
**Table 3.The rate of desalination before and after irrigation of 0~200cm soil layer**

| Soil layer (cm) | Irrigation amount (240L) | | | Irrigation amount (200L) | | | Irrigation amount (160L) | | |
|---|---|---|---|---|---|---|---|---|---|
| | Initial salt content ($g\cdot kg^{-1}$) | Final salt content ($g\cdot kg^{-1}$) | Rate of desalination (%) | Initial salt content ($g\cdot kg^{-1}$) | Final salt content ($g\cdot kg^{-1}$) | Rate of desalination (%) | Initial salt content ($g\cdot kg^{-1}$) | Final salt content ($g\cdot kg^{-1}$) | Rate of desalination (%) |
| 10 | 11.126 | 0.036 | 99.68 | 11.139 | 0.096 | 99.14 | 11.533 | 0.669 | 94.19 |
| 25 | 8.717 | 0.025 | 99.72 | 8.698 | 0.365 | 95.81 | 8.033 | 0.873 | 89.13 |
| 55 | 6.976 | 0.885 | 87.31 | 6.611 | 1.395 | 78.89 | 4.074 | 1.413 | 65.31 |
| 80 | 5.931 | 1.558 | 73.74 | 5.730 | 1.965 | 65.70 | 5.390 | 2.397 | 55.53 |
| 110 | 4.539 | 1.661 | 63.39 | 4.288 | 2.323 | 45.82 | 4.381 | 3.023 | 30.99 |
| 140 | 4.869 | 2.865 | 41.16 | 4.099 | 2.968 | 27.59 | 4.077 | 3.373 | 17.26 |
| 170 | 3.647 | 4.225 | -15.86 | 3.868 | 4.139 | -7.01 | 3.637 | 3.768 | -3.58 |
| 200 | 2.076 | 2.337 | -12.56 | 2.258 | 2.397 | -6.16 | 2.419 | 2.490 | -2.91 |



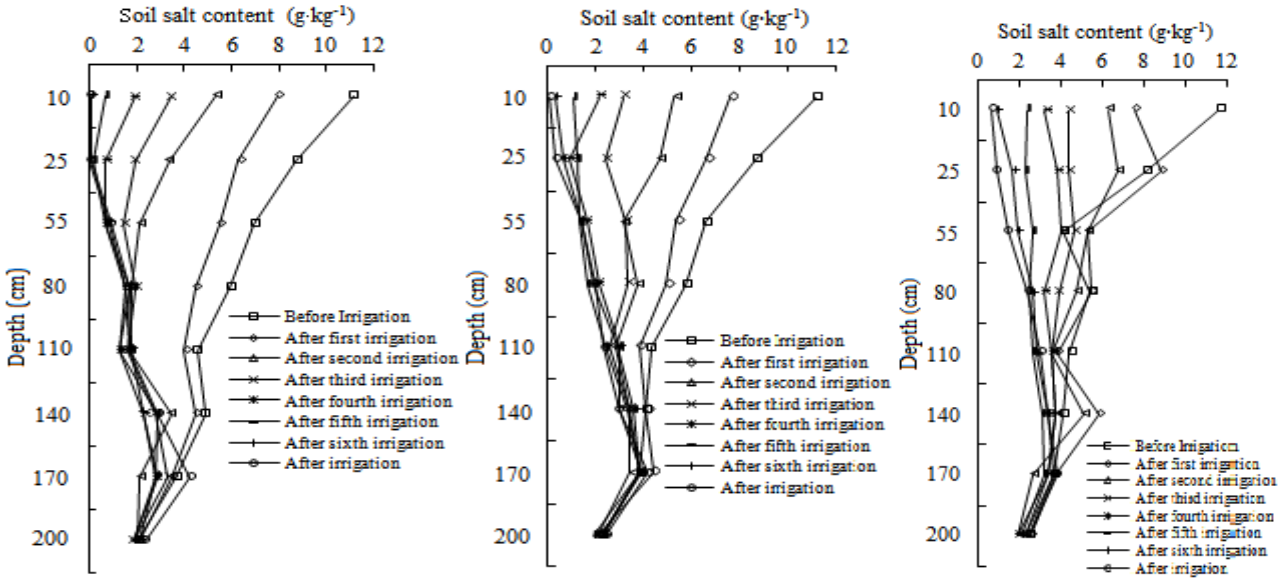

□a□ 240L □b□ 200L □c□160L

**Figure 1.The characteristics of soil water change under the different irrigation amount**



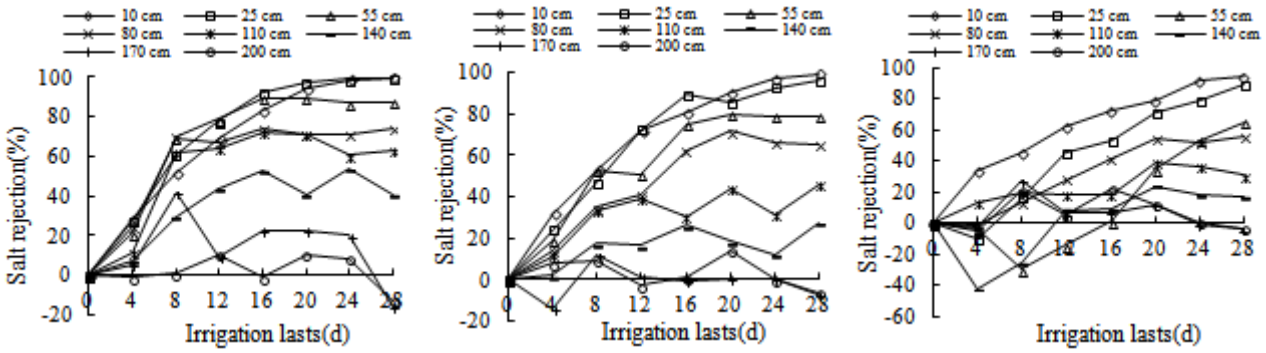

□a□Before irrigation □b□1d after irrigation □c□One irrigation period

**Figure 2.The changes of soil moisture under different irrigation amount**




**(a) 240L** **(b) 200L** **(c) 160L**

**Figure 3. The vertical changes of each layer soil salinity of different irrigation amount**




**□a□240 L** **□b□200 L** **□c□160 L**

**Figure 4.The relationship between the desalination rate and the time of the irrigation calendar**

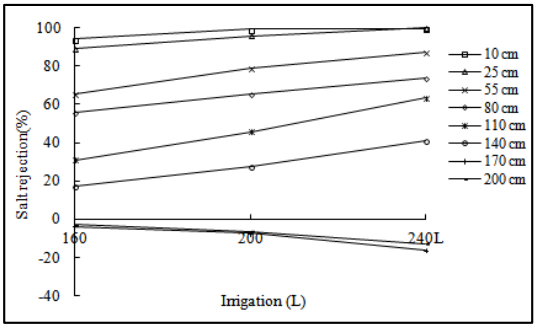



**Figure 5.The relationship between desalination rate and irrigation amount**

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**RESEARCH ARTICLE**

# Study on Au -Ni Core-Shell Nanowires for Optical Applications

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**ABSTRACT**

The optical properties of Au-Ni core-shell nanowire were simulated using the Mie-Lorentz scattering approach. The simulation fixed the core diameter of Au at approximately 20nm and the thickness of the Ni shell at 20nm. The optical absorbance spectrum, optical transmittance spectrum, absorption coefficient and the optical energy gap of Au and Au-Ni coaxial nanowire as a function of wavelength were calculated. The study reveals an excellent enhancement in the values of optical absorbance spectrum and absorption coefficient for Au-Ni core-shell nanowires compared to Au nanowires. The results of energy gap calculations demonstrated that Au-Ni core-shell nanowires have an optical application as ultra-violet photo-detectors.

**Keywords:** Mie scattering, Absorption efficiency, optical transmittance spectrum, Au-Ni core-shell nanowires

## INTRODUCTION

A nanowire can be defined as a nanostructure with a diameter in the order of a nanometer [1]. Also, it is known as a structure that has a thickness of tens of nanometers with an unconstrained length. The study of semiconductor nanowires has developed rapidly over the last few years. Various types of nanowire have been utilized in novel scientific applications [2–6]. In the last decades, an advanced application of gold nanostructures has emerged [7]. As a result of its extraordinary properties, such as low reactivity and malleability, gold occupies a special position in electrochemical analyses [8]. Although gold is a distinguished bulk material, various surprises appear when one considers the nanoscale structures. For example, in the transition from gold crystalline (fcc) to nanowires, there appears a remarkable organization of matter [9]. Gold nanowires (Au NWs) are a key research subject in the field of nanotechnology due to their unique physical properties and potential applications in nano-photonic devices and sensors [10,11]. To investigate the photonic applications of Au-core/Ni-shell NWs, optical properties of both Au NW and Au-core/Ni-shell NWs were calculated. A nanowire-based core/shell structure is used in this work to combine the advantages of Au and the Ni materials and thus to improve the performance of the pattern. Optical properties play an important role in the study of the synthesis of semiconductor materials. The optical properties investigated

in this study are the optical absorbance spectrum, optical transmittance spectrum, absorption coefficient, and optical energy gap.

## METHODOLOGY

### Computational Details

In this study, we studied the optical properties of such coaxial nanowires in the framework of Mie- Lorenz scattering theory. It was developed to evaluate the circular cylinders with core and core-shell forms. This formalism enables the calculation of the absorption and extinction efficiency of individual layers. The calculation of un-polarized light absorption of core-shell nanowires used the simulator "Optical Properties of Single Coaxial Nanowires" [12]. The coaxial NWs are evaluated as infinitely long cylinders along the z-axis, normally lighted by an electromagnetic plane wave with an incident propagation wave vector $\vec{k}$ , as shown in Figure 1. The radii of the core-shell inside coaxial nanowires are given by $a$ and $b$ , respectively. The angle of incidence propagation wave is denoted by $\alpha$. The simulation considers that transverse magnetic (TM) wave and the transverse electric (TE) wave are both perpendicular to the nanowire axis.

The absorption efficiency $Q_{abs}$ refers to the ratio of the absorption cross-section and geometrical cross section of a single wire, and is given by:

$$Q_{abs} = C_{abs}/C_{geom} \qquad (1)$$

where $C_{abs}$ and $C_{geom}$ are the absorption and geometrical cross sections respectively [13].

The absorption cross section is

$$C_{abs} = C_{ext} - C_{scat} \qquad (2)$$

Where $C_{ext}$ is the extinction cross section and $C_{scat}$ is scattering cross section.
In case of un-polarized light such as sunlight, we can represent the absorption cross section $C_{abs}$ of coaxial wires as [14]:

$$C_{abs} = (C_{ext}^{TE} + C_{ext}^{TM})/2 - (C_{scat}^{TE} + C_{scat}^{TM})/2 \qquad (3)$$

In order to calculate the Mie coefficients of the system:

- Determining the solutions of the 2D vector wave equation in various spatial regions to express the electromagnetic fields.
- Interchanging the magnetic field into the electric field of Maxwell's equations.
- These solutions can be written in form of a summation for Bessel functions, whose coefficients have been calculated by solving the boundary conditions.
- Applying the boundary conditions for continuity at interface case.
- The results are then symbolized in a matrix form.
- Computing the unidentified Mie coefficients.

## RESULTS AND DISCUSSION

### Results of optical absorbance spectrum

The absorption spectrum (A), using absorbance unit (a.u.) as a function of the wavelength ($\lambda$) for (Au, Ni) nanowires and Au/Ni-Coaxial NWs is shown in Figure 2. The results show the behavior of absorbance spectra is completely opposite to that of transmission spectra. High values of absorbance in the UV region makes this material useful in photovoltaic applications [15], for Coaxial NWs, there was a dramatic decrease in the absorption with increasing wavelength. This can be attributed to that the incident photon could not be the excited electron and move it from the valance band to the conduction band, because the incident photon energy is below the optical energy gap of the metal. Absorbance is defined as the ratio between the intensity of the radiation absorbed by the material and the intensity of the radiation incidence on it [16]. The absorbance spectrum increases when the shell enfolds the core, especially within the spectrum region of UV-visible to the end of the spectrum range due to increase of the surface roughness.

### Results of optical transmittance spectrum

The spectrum of optical transmittance is associated with thickness, chemical composition, crystal structure and surface morphology of samples [17]. The optical transmittance spectra (T%) as a function to the wavelength were calculated within the range (300-1000) nm for (Au, Ni) NW and Au/Ni-Coaxial NWs using the relation (4) [18].

$$T = e^{-2.303A} \quad \ldots\ldots\ldots\ldots (4)$$

Where $A$ is the absorbance spectrum. Figure 3 illustrates the transmittance as a function to the wavelength. The transmittance values were found to be the lowest in the wavelength region corresponding to the optical energy gap for Au NWs. For example, in the ultraviolet region of the spectrum within the range 300-400 nm, there is high transparency in the visible region with transparency reaching about 88%. Low ranges of transmittance recorded for Ni NW along spectrum range, while the transmittance curve of Au- core NW shows a sudden and sharp increase at wavelengths above 600-650 nm then a slight decrease above wavelength of 700 nm. This indicates that these wires can using in various solar cell applications due to the valuable spectral region of the solar cells lying in the visible section of the electromagnetic spectrum [19]. Transmittance was reduced to 26% when the core was covered with the shell. The differences in transparency values are likely due to the differences in core/shell thickness. This is in consistent with the increase of the surface roughness promoting the increase of the surface scattering of light. In addition, the absorption edge shifted toward a longer wavelength (red shift) as a result of an increase in the particle size of the crystal [20]. Red shift in the absorption edge is useful for optical applications where it prevents unfavorable absorption in the luminous range of the spectrum [21].

### Results of absorption coefficient

The absorption coefficient ($\alpha$) was verified from the region of high absorption at the fundamental absorption edge of the nanowire according to the following relation (5) [18].

$$\alpha = 2.303 \left(\frac{A}{t}\right) \quad \ldots\ldots\ldots\ldots\ldots (5)$$

Where $t$ is the thickness of nanowires,Figure 4 shows $\alpha$ for Au, Ni and Au/Ni NWs as a function to the wavelength. It decreases with increasing wavelength; this means that direct electronic transition occurs. In addition, we can observe from this figure that $\alpha$ generally rises with expanding coaxial NW thickness, where in the case of thicker

NWs, more atoms are present in the atomic structure, so farther states will be available for the photons to be absorbed. It is noticeable that the edge of optical absorption is moving towards the high wavelengths, while a sharp reduction in the absorption coefficient was observed with increasing wavelength within the range 300-550 nm. This is due to the increase in the transmittance spectral within this spectral range. Additionally, there is a shift in the absorption edge towards high wavelengths where the absorption edge represents the wavelength at which the material is transparent to the higher wavelengths and absorbed to the shorter wavelengths, such as at the end of the absorption process, and the beginning of transmittance.

### Results of optical energy gap ($E_g^{opt}$)

In general, values of energy gap ($E_g$) depend on several factors such as crystal structure, distribution, and arrangement of atoms in the crystal lattice and crystal regularity [17]. The direct optical energy gap values have been verified using the Tauc equation (6) [16].

$$\alpha h\upsilon = \beta\left(h\upsilon - E_g^{opt}\right)^r \ldots\ldots\ldots\ldots\ldots\ldots (6)$$

Where $h\upsilon$ is the incident energy, h is plank constant, r is a constant which takes the values (1/2, 3/2, 2, 3) depending on the material and the nature of the optical transition whether it is direct or indirect, and $E_g^{opt}$: is optical energy gap. The Tauc equation is benefited to find the type of optical transition. A plot of $(\alpha h\upsilon)^2$ versus $h\upsilon$ for Au, Ni and Au/Ni NWs is illustrated in Figure 5. The plot is linear showing the direct band gap of the structure. Extrapolation of the linear of the line to the $h\upsilon$ axis gives the values of energy band gap, which found to be approximately 2.5 eV for Au NWs. This result is close to those obtained by H. Bakkali et al [22] and M. Bagheri et al [23] while 2.3 eV for Ni NWs, this is in agreement with F. I. Ezema et al [24]. The band gap decreases slightly to 2.4 eV for Coaxial-Au/Ni NWs as compared to Au NWs. This might be due to an increase in thickness and in the localized state density near the band edges causes a decrease in the value of optical energy gap [18] [24].

## CONCLUSION

The optical properties of Au-Ni core-shell nanowires were studied by theoretical simulation using the Mie scattering framework. It was shown that Au-Ni core-shell NWs demonstrate an excellent improvement in the values of both optical absorbance spectrum and absorption coefficient. The values of Au-Ni coaxial NW energy gap were slightly shifted down due to the increase in shell thickness. From the discussed optical properties of coaxial- Au/Ni nanowires, it is evident that nanowires can act an extremely important role in the development of optoelectronic devices.

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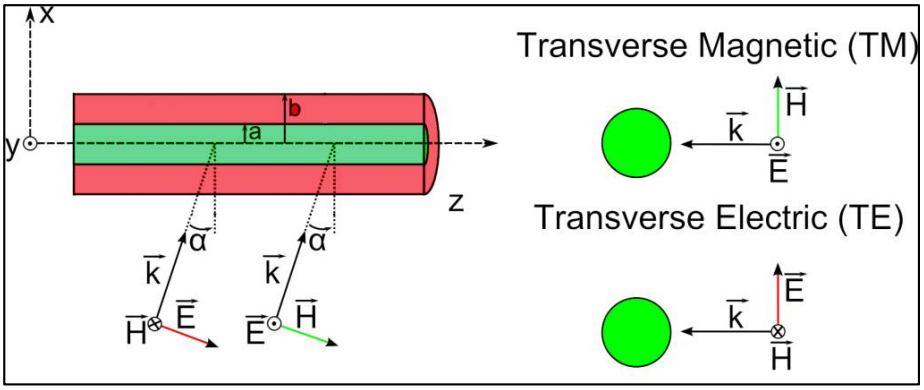


**Figure 1: Schematic diagram of the coaxial nanowire with the direction of TM and TE propagation**

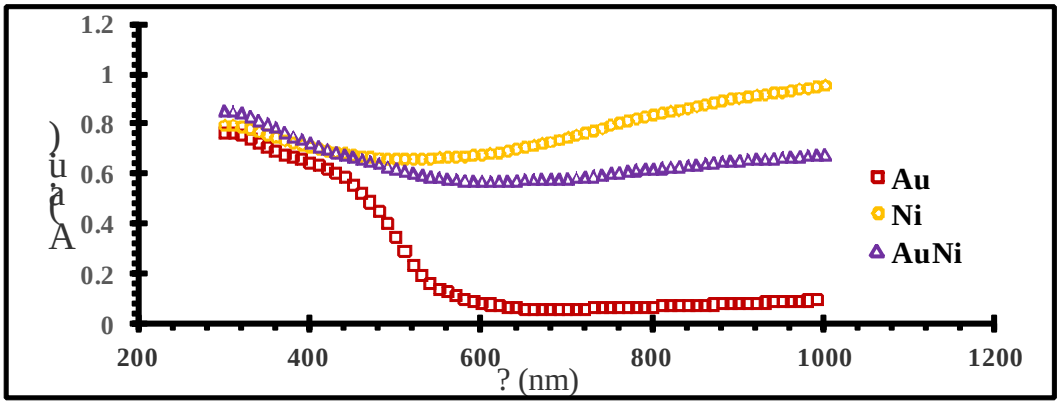


**Figure 2: Absorbance as a function to the wavelength for Au, Ni and Au/Ni NWs**

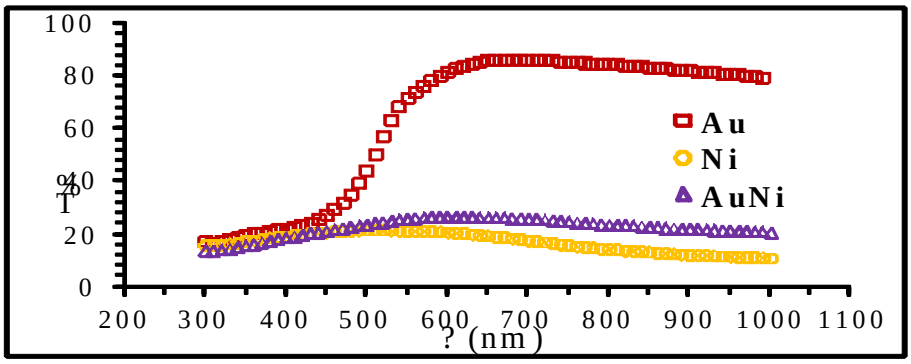


**Figure 3: Transmittance as a function to the wavelength for Au, Ni and Au/Ni NWs.**

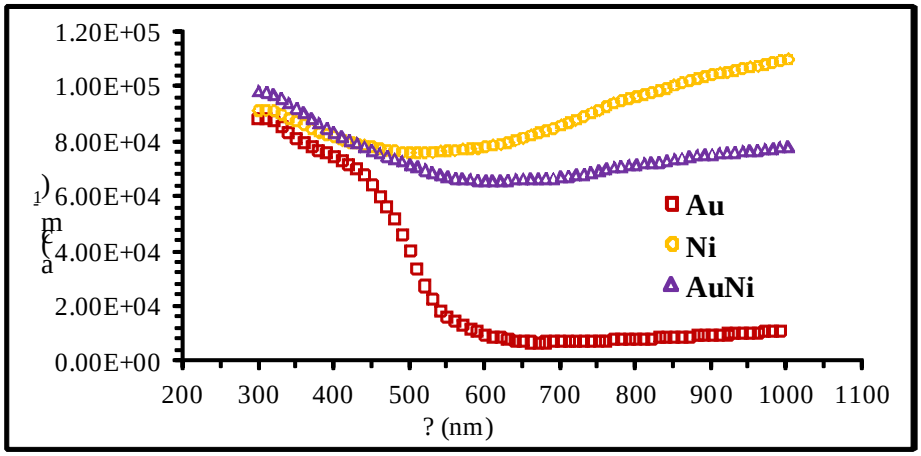


**Figure 4: Absorption coefficient of Au, Ni and Au/Ni NWs versus the wavelength**

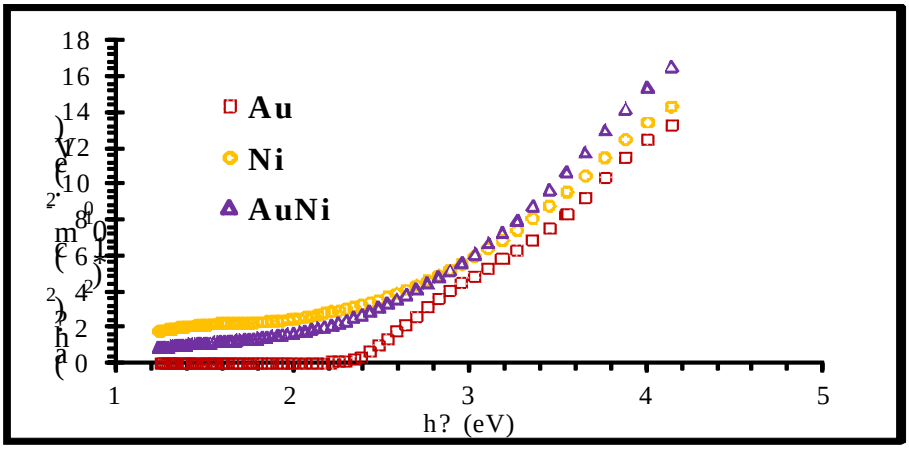


**Figure 5:Tauc plot for Au, Ni and Au/Ni NWs**

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**RESEARCH ARTICLE**

# Molecular Detection, Gross and Histopathological Studies of Infectious Bronchitis in Chicken in Wayanad District

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**ABSTRACT**

A comprehensive study was conducted to detect the presence of Infectious bronchitis virus (IBV) in chickens affected with respiratory infection in Wayanad district of Kerala. The tissue samples were examined for gross lesions as well as histopathological lesions after formalin fixation and further tissue processing. Molecular detection was also carried out after RNA/ DNA extraction. For IBV detection, nested reverse transcription polymerase chain reaction (RT-PCR) targeting N gene region was carried out and eight samples were detected as positive giving a percentage positivity of 16 per cent. By sequencing and BLAST analysis, it was found that IBV isolate has similarity with Chinese isolates. The study could successfully detect the presence of Infectious Bronchitis in the poultry population in Wayanad district of Kerala.

**Keywords:** Infectious bronchitis, gross lesions, histopathology lesions, molecular detection

## INTRODUCTION

Poultry rearing is one of the fastest growing and the most organised agricultural sectors in India. Chickens are prone to a number of respiratory infectious diseases which can cause massive mortality in a short period. Many poultry farms in the district of Wayanad in Northern Kerala witnessed considerable increase in mortality in the recent past predominantly due to respiratory dysfunctions leading to wiping out of entire flock in some cases. The clinical symptoms and post mortem observations were suggestive of viral aetiology and major bacterial causes were ruled

out. Infectious bronchitis virus (IBV) is capable of producing respiratory, reproductive as well as nephropathogenic lesions even though respiratory infection being the major one. There are reports of occurrence of this viral infection from other parts of the country, and only a few cases have been reported from Kerala. So far no detailed study was conducted on this disease in chicken in Wayanad district. Early detection of these viral agents and rapid adoption of effective control methods are obligatory to reduce economic loss to the poultry farmers.

## MATERIALS AND METHODS

Samples for the study were collected from 50 birds died with respiratory affections which were brought from organised poultry farms in and around Wayanad district to the Department of Veterinary Pathology, College of Veterinary and Animal Sciences, Pookode for necropsy. Clinical samples collected included lung, trachea and spleen from dead birds. The tissue samples were collected in sterile screw capped polypropylene vials using sterile scissors and forceps and stored at -80°C after proper labelling to prevent RNA degradation till further processing. The tissue samples for histopathological examination were preserved in 10 per cent formalin and embedded in paraffin for histopathological examination. Sections were cut at four micron thickness and stained by routine Haematoxylin and Eosin (H&E) stain (Bancroft and Gamble, 2008).

### Detection of IBV by nested RT-PCR

### Extraction of total RNA from tissues

Total RNA was isolated from the tissue samples by using Trizol reagent as per the manufacturer's protocol. Briefly, 50-100 mg tissue was homogenized thoroughly in 1 mL of TRIzol reagent and incubated for 5 min at room temperature after homogenization. Subsequently, 0.2 mL chloroform was added. The microcentrifuge tube was shaken vigorously by hand for 15 sec and incubated for 2-3 min at room temperature. The mixture was centrifuged at 12,000 rpm for 15 min at 4°C. Aqueous phase was collected in a new microcentrifuge tube and 0.5 mL 100 per cent isopropanol was added. After 10 min incubation at room temperature the mixture was centrifuged at 12,000 rpm for 10 min at 4°C. Supernatant was removed leaving only the RNA pellet. RNA pellet was washed with 1 mL 75 per cent ethanol. After vortexing, the mixture was centrifuged at 7500 rpm for 5 min at 4°C. Wash was discarded and pellet was air dried for 10 min at 37°C. Further RNA pellet was resuspended in nuclease free water and stored at -80°C until used.

### Synthesis of complementary DNA (cDNA) by reverse transcription of total RNA

Complementary DNA (cDNA) was synthesized using Thermo Scientific RevertAid H Minus First Strand cDNA Synthesis Kit (Thermo Scientific, USA) by following manufacturer's protocol. To a DEPC treated PCR tube kept on ice, added 1 µg of total RNA, 1 µL (0.2 µg/µL) of random hexamer primer and rest nuclease free water (NFW) to make the volume up to 12 µL. The mixture was incubated at 65°C for 5 min and was snap chilled on ice. To the above mixture, 4 µL of 5X reaction buffer, 1 µL (20 U/µL) Ribolock RNase inhibitor, 2 µL of 10 mM dNTP mix and 1 µL (200U/µl) RevertAid H minus M-MuLV reverse transcriptase were added and the mixture was incubated at 25°C for 5 min followed by 60 min at 42°C. The reaction was terminated by heating at 70°C for 5 min. The cDNA was stored at -20°C for further use.

### Nested RT-PCR

Presence of IBV genome in the tissue samples was detected by nested RT-PCR using primers specific to N gene as described by Farsang *et al*. (2002) with slight modifications. The details of the outer set and inner set primers are given in Table 1. The 25 µL reaction mixture of the first round PCR comprised of 12.5 µL of 2X EmeraldAmp GT PCR Mastermix containing *Taq* polymerase (TaKaRa), 1 µL each of 10 pmol outer forward and reverse primers, 1 µL

of cDNA and rest NFW to make up the volume. Along with this, a positive control (cDNA prepared from Bronki-L vaccine ie, Newcastle (LaSota strain) and Infectious bronchitis (Massachusetts type) combined vaccine and no template control (NTC) was included replacing template with NFW. The cycling conditions were 95°C for 3 min (initial denaturation), 35 cycles of 95°C for 30 sec (denaturation), 60°C for 1 min (annealing) and 72°C for 1 min (polymerization) followed by a single cycle at 72°C for 5 min (final extension). For the nested PCR the product of the first PCR was taken as template. The 25 µL reaction mix consisted of 12.5 µL PCR 2X master mix, 1 µL each of 10 pmol inner forward and reverse primers, 1 µL PCR product and rest NFW. The cycling conditions for amplification were 95°C for 3 min (initial denaturation), 38 cycles of 95°C for 30 sec (denaturation), 53°C for 45 sec (annealing) and 72°C for 1 min (polymerization) followed by final extension at 72°C for 5 min.

### Visualization of the PCR product

Agarose gel of one per cent concentration was prepared with 0.5X TBE buffer. Agarose was weighed and dissolved in 0.5X TBE buffer by heating and was cooled to 50°C. Ethidium bromide was added to a final concentration of 0.5 µg/ml to the melted agarose. The gel was poured into the gel tray after placing the comb. After the gel has set, the comb was removed and gel was placed in the electrophoresis apparatus. The six µL of the PCR product was loaded into the wells. One microliter of 100 bp DNA ladder (Thermo Scientific, India) was loaded in one of the wells as the molecular weight marker. Electrophoresis was carried out at 80V until bromothymol blue dye has migrated to more than two-thirds of the gel. The gel was visualized to identify the size of the product and photograph was taken in a gel documentation system under UV illumination.

## RESULTS

### Sample Collection

In this study, clinical samples from 50 birds belonging to different regions of Wayanad district were collected. The tissue samples were obtained from dead birds which showed post-mortem lesions like pulmonary oedema, pneumonia, cloudy airsacs, sinusitis, pulmonary haemorrhage and tracheal haemorrhage.

### Gross Lesions

The important gross lesions observed were pulmonary congestion, pulmonary oedema, haemorrhagic streaks on trachea, mucus in trachea, frothy exudate in lungs, pneumonic changes and haemorrhages on proventriculus and caecal tonsil (Fig.1-3).

### Histopathological lesions in respiratory system

Lungs showed severe bronchial lesions with marked pulmonary congestion, haemorrhage and thrombi in the pulmonary artery, mural and channelled thrombi with fibrinoid degeneration of blood vessel wall.Trachea showed marked desquamation and mild or very minimal hyperplastic reaction of mucosal epithelia, heavy infiltration of mononuclear cells, marked congestion and haemorrhage, loss of cilia, severe infiltration of submucosa with lymphocytes and macrophages, degeneration and desquamation of mucosa (Fig. 4-9).

### Detection of IBV by nested RT-PCR

Of the 50 samples tested, eight were found positive for IBV by nested RT-PCR targeting the N (nucleocapsid protein) gene region. Percentage positivity was calculated to be 16 per cent. Amplicons of 380 bp generated by the nested PCR reaction were obtained on agarose gel electrophoresis (Fig.10) and the non-template control (NTC) exhibited no band.Representative positive samples obtained for IBV were sent for sequencing to AgriGenome Labs Private

Limited, Cochin. When the chromatograms obtained after sequencing were analysed, it was observed that there were no errors. On BLAST analysis, IBV isolate showed 95 per cent similarity to Chinese isolates (Fig. 11).

## DISCUSSION

Respiratory infections are quite common in the poultry flocks all over the world. The causes of these infections could be ranging from temperature variations to different infectious agents such as viral, bacterial and fungal. An important emerging poultry pathogen is IBV that affects the respiratory, reproductive as well as renal system of infected birds.In recent years in Wayanad, there has been a rise in poultry mortality due to respiratory illness. Hence, the study was conducted to detect and identify the respiratory viral disease, IB. This study also included detailed investigations on clinical pathology and molecular characterization of viral respiratory infection. The clinical signs observed in the infected birds were dyspnoea, respiratory rales, coughing and sinusitis. Major pathological lesions observed were pneumonia, pulmonary oedema, congested lung, liver and kidney, thick and slimy airsacs, thick exudates from sinus, haemorrhagic tracheitis, petechial haemorrhage on proventriculus, haemorrhagic caecal tonsillitis, haemorrhagic enteritis, cardiomegaly, hepatomegaly and spleenomegaly. Similar lesions were reported by Khataby *et al*. (2016).The histopathological lesions observed in the eight IB detected cases were pulmonary congestion, haemorrhage and thrombus. Trachea showed loss of cilia of epithelial cells, marked desquamation of mucosal epithelia, heavy infiltration of mononuclear cells, marked congestion and haemorrhage. Similar lesions like loss of cilia, oedema, rounding and sloughing of epithelial cells and infiltration by lymphocytes were described by Bande *et al*. (2016).

In this study, eight cases were detected as positive with a percentage positivity of 16 per cent. Many experiments had substantiated that nested PCR assays are more sensitive than the single-run PCR. The higher sensitivity is needed for a reliable diagnosis and nested assay provides a potent, novel tool in IB detection (Ballagi-Pordany *et al*., 1992). This justifies the use of nested RT-PCR targeting N gene in this study.

## CONCLUSION

Among various livestock rearing sectors, poultry rearing is the most acceptable and congenial one for all categories. It contributes a crucial part of the income to small households and furnishes livelihood of two-third of rural community. In the recent past, many cases of massive poultry death were reported owing to respiratory affections in Wayanad district of Kerala, the accurate diagnosis of which could not be carried out. This scenario claimed testing for considering emerging as well as established viral pathogen like IBV as causative of major respiratory infection. Hence, the present study was undertaken with the objective of detecting IBV and to ascertain the gross and histopathological alterations due to these respiratory viral diseases. This is essential for accurate identification of cases of poultry mortality and also to add to knowledge about this disease of poultry prevalent in Wayanad region. The study could successfully detect the presence of IBV in the poultry population of Wayanad district of Kerala by molecular methods and the histopathological lesions in affected birds were analysed in detail.

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**Table 1. Details of Pcr primer (outer and inner set) used for detection of IBV**

| Set | Primer Name | Sequence 5'-3' | Target region/ Gene | Product size (bp) | Reference |
|---|---|---|---|---|---|
| Inner | NIBF1 | AAT TTT GGT GAT GAC AAG ATG A | N gene | 402 | Farsang *et al*., 2002 |
| | NIBR1 | CAT TGT TCC TCT CCT CAT CTG | | | |
| Outer | NIBF2 | GTG ATG ACA AGA TGA ATG AGG A | | 380 | |
| | NIBR2 | CAG CTG AGG TCA ATG CTT TAT C | | | |

**GROSS LESIONS**

**Fig.1. Lung- Infectious bronchitis- severe pneumonic changes- frothy exudate from lungs**

**Fig. 2. Lung- Infectious bronchitis- congestion and oedema**

**Fig. 3. Trachea– Infectious bronchitis-lumen lined with red streaks of haemorrhage**

**HISTOPATHOLOGICAL LESIONS**

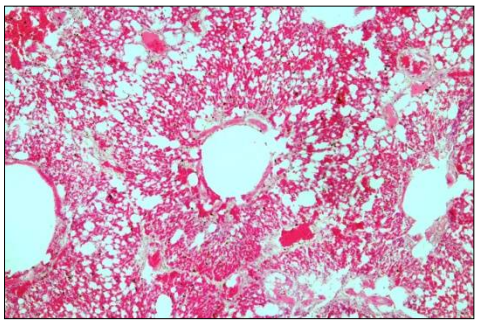

**Fig.4. Lung- Infectious bronchitis- severe multifocal vascular congestion (H&E, 100X)**

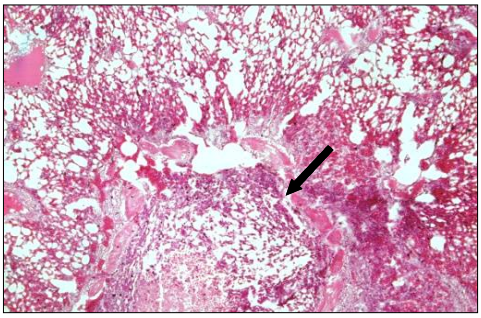

**Fig.5. Lung- Infectious bronchitis-severe bronchial epithelial destruction (H&E, 100X)**

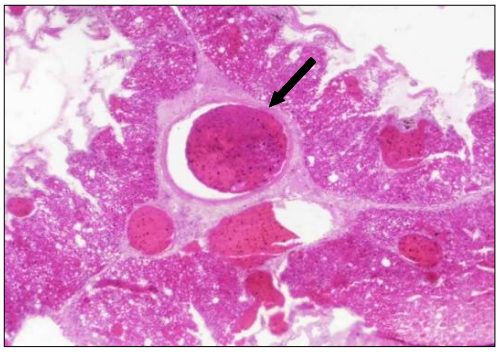

**Fig.6. Lung- Infectious bronchitis-lumen of pulmonary artery occluded with thrombus (H&E, 100X)**

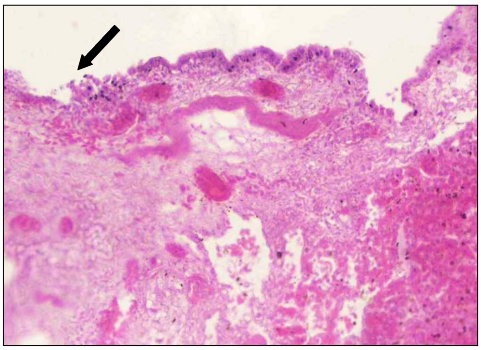

**Fig.7. Lung- Infectious bronchitis- bronchial epithelial desquamation (H&E, 200X)**

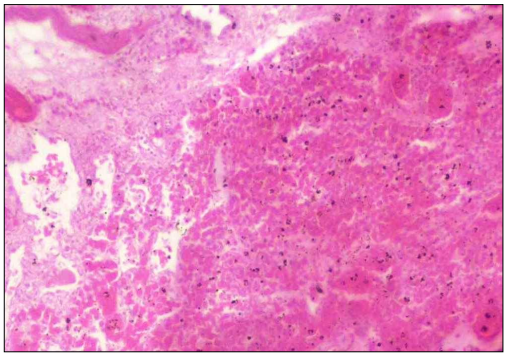

**Fig.8. Lung- Infectious bronchitis- extensive pulmonary haemorrhage (H&E, 200X)**

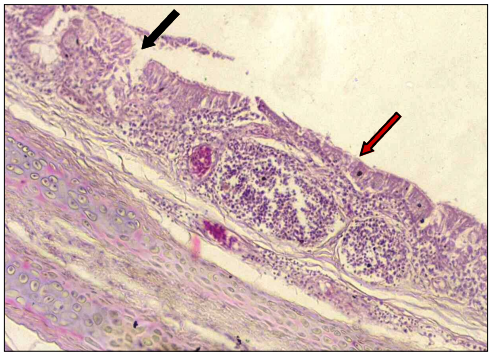

**Fig.9. Trachea- Infectious bronchitis- tracheal epithelial desquamation (black arrow) with mild to moderate infiltration of mononuclear cells (red arrow) (H&E, 200X)**

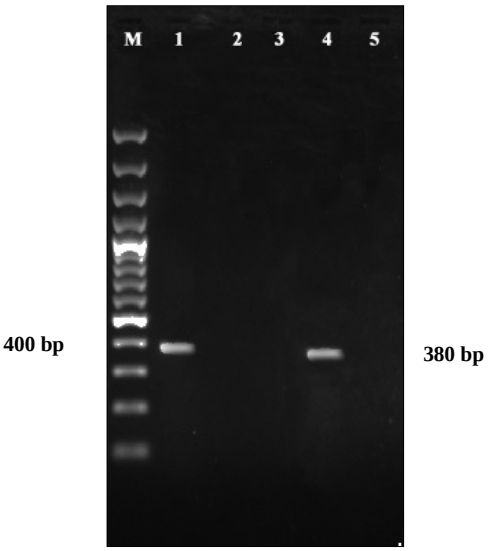


**Fig.10: Agarose gel showing 380 bp amplicons generated by targeting N gene of IBV**

Lane M : 100 bp DNA Ladder
Lane 1 : Positive control
Lane 2-4 : Samples
Lane 5 : No template control

| Description | Max score | Total score | Query cover | E value | Ident | Accession |
|---|---|---|---|---|---|---|
| Infectious bronchitis virus strain GX-YL130806200 spike protein (S), 3a protein (3a), 3b protein (3b), envelope protein (E), membrane protein (M), 5a pro | 488 | 488 | 100% | 4e-134 | 95% | KR265091.1 |
| Infectious bronchitis virus strain GX-YL130806200 N protein (N) gene, complete cds | 488 | 488 | 100% | 4e-134 | 95% | KJ940501.1 |
| Infectious bronchitis virus spike protein (S), 3a protein (3a), 3b protein (3b), envelope protein (E), membrane glycoprotein (M), 5a protein (5a), 5b (5b), ar | 488 | 488 | 100% | 4e-134 | 95% | KJ669722.1 |
| Infectious bronchitis virus strain GX-YL13080630 nucleocapsid protein (N) gene, complete cds | 488 | 488 | 100% | 4e-134 | 95% | KF996286.1 |
| Infectious bronchitis virus strain ck/CH/LSD/110410, complete genome | 488 | 488 | 100% | 4e-134 | 95% | KP118893.1 |
| Infectious bronchitis virus strain ck/CH/LLN/130101, complete genome | 488 | 488 | 100% | 4e-134 | 95% | KP118892.1 |
| Infectious bronchitis virus strain ck/CH/LHLJ/111246, complete genome | 488 | 488 | 100% | 4e-134 | 95% | KP118891.1 |
| Infectious bronchitis virus strain ck/CH/LLN/130102, complete genome | 488 | 488 | 100% | 4e-134 | 95% | KP118888.1 |
| Infectious bronchitis virus strain ck/CH/LHB/140532, complete genome | 488 | 488 | 100% | 4e-134 | 95% | KP118887.1 |
| Infectious bronchitis virus strain ck/CH/LSD/110851, complete genome | 488 | 488 | 100% | 4e-134 | 95% | KP118884.1 |
| Infectious bronchitis virus strain ck/CH/LBJ/140402, complete genome | 488 | 488 | 100% | 4e-134 | 95% | KP118882.1 |
| Infectious bronchitis virus strain ck/CH/LHB/130927, complete genome | 488 | 488 | 100% | 4e-134 | 95% | KP118880.1 |
| Infectious bronchitis virus strain ck/CH/LHB/121010, complete genome | 488 | 488 | 100% | 4e-134 | 95% | KP036503.1 |
| Infectious bronchitis virus strain CK/CH/LLN/111169, complete genome | 488 | 488 | 100% | 4e-134 | 95% | KF411040.1 |
| Infectious bronchitis virus strain 4/91 vaccine, complete genome | 488 | 488 | 100% | 4e-134 | 95% | KF377577.1 |
| Infectious bronchitis virus isolate IBV01 5b protein gene, partial cds; and nucleocapsid protein (N) gene, complete cds | 488 | 488 | 100% | 4e-134 | 95% | EU889030.1 |

**Fig. 11. BLAST analysis of nucleotide sequence of the N gene of IBV isolate**

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**RESEARCH ARTICLE**

# The Role of Pumpkin Seed Oil in Healing of Wounds in Diabetic Mice

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## ABSTRACT

This study conducted to explore the role of pumpkin seed oil of Cucurbita ***pepo*** ,as alternative of traditional medicinal treatment for management wound healing in diabetic mice. Firstly ,the oil of ***Cucurbita Pepo*** (pumpkin seed oil) obtained by hot extraction method with n-hexane . Forty male Balb-c mice ,positively induced diabetes by single intra peritoneal dose of alloxan 150mg/kg BW, and surgical bilateral skin wounds performed on the back region ,after they were provide diabetic. The mice received daily insulin therapy 14 Iu/kg.bw along the period of the experiment . Clotting time ,platelet counts, blood glucose, insulin concentration ,white blood cells count, red blood cells count, differential white blood cells count ,pro thrombin time ,thrombin time and fibrinogen level ,were assessed after one day of incision and after complete wound healing in all experimental animals . The wounds were followed up through measuring their diameters, photographically and histologically (through obtaining skin biopsy after 4,6,8,10 day and so on of incision till healing) . The result showed, the extraction yield of ***C. pepo*** seed is 58% oil (pumpkin seed oil), and the phytochemical analysis of both pumpkin seed oil and powder proved presence of saponin ,flavonoid and steroid. There were no significant $p<0.05$ changes in clotting time ,platelet count ,red blood cells count and pro thrombin time between their values after one day of incision and after healing of all experimental groups . There were significant $p<0.05$ decrease in serum insulin concentration ,blood glucose level , white blood cells count ,lymphocytes count and thrombin time of groups treated with PSO and PSO plus MEBO when compared between their values after healing to those after one day of incision , but the fibrinogen level was significantly $p<0.05$ increase in the group treated with pumpkin seed oil only .The diameter of wounds of all experimental

groups that treated with PSO,MEBO and PSO plus MEBO , showed significant p<0.05 decrease, but complete healing involved most number of treated animal with PSO,MEBO,PSO+MEBO along the period of treatment . Histologically , the all experimental treated groups showed clearly progression of wound healing represented by epithlization and collagen formation which were specially too early in PSO+MEBO and PSO alone groups almost complete healing .

**Keywords:** Alloxan, *Cucurbita pepo* , MEBO, PSO

## INTRODUCTION

Wounds are physical injuries that result in an opening or break of the skin that cause disturbance in normal skin anatomy and function[1] .The process of wound healing occurs in different phases such as coagulation, epithelization, granulation, college nation and tissue remodeling[2]. Collagen, the major component which strengthens and supports extracellular tissue, contains substantial amounts of hydroxyproline, which has been used as a biochemical marker for tissue collagen[3]. Decreases in growth factors responsible for tissue repair such as platelet derived growth factor (PDGF) and transforming growth factor-b (TGF-b) documented in diabetic wounds .

In patients with diabetes, alterations in MMP (matrix metallo proteinases) expression and activation are consistent with a pattern associated with increased degradation of newly formed ECM (extracellular matrix) . Many factors can influence on wounds healing are essential for developing better therapeutic option for wound treatment [4] . Improper diet and infection at wound site ,drugs, elderly age ,insufficient oxygen supply and tissue perfusion to wound area ,diabetes and other disease condition .The use of herbal remedies individually or in combination with standard medicines has been used in various medical treatises for the cure of different diseases . Pumpkin is one of the well-known edible plants and has substantial medicinal properties due to the presence of unique natural edible substances. It contains several phytoconstituents belonging to the categories of alkaloids, flavonoids, and palmitic, oleic and linoleic acids. Our study designated to Investigating the effect of pumpkin seeds oil for treatment wounds in diabetic mice. Hopefully, it will facilitate the development of strategies to improve the rate of tissue repair in diabetes.

## MATERIALS AND METHODS

**The Drugs**

**Alloxan**: obtained from Labor ,India and dosed IP 150 mg /kg. Bw single dose for inducing diabetes in mice . **Insulin**: vial contains 100 IU supplied by Eli Lilly , USA. **MEBO ointment** : produced by julphar **,**U.A.E. **Pumpkin** (***cucurbita . pepo***) fruit purchased from ALshorga market-Baghdad –Iraq during October 2016. And extracted by n-hexane .

**The Animals:** forty male Balb-C mice , with body weight ranged 20-30 grams and age 8-12 week, purchased from animal house - Ministry of Technology. They were bred under optimum condition of ventilation, temperature 25 °C and dark and light cycle 12/12. The animals were fed on special formula feed pellets and given water ***ad libitum***. Throughout the experiment, each 6 mice were housed in a plastic cage 20x15x15 cm dimensions containing hard-wood chip as bedding. The bedding was changed continuously to ensure a clean environment. They were left for two weeks to acclimatization before starting of the experiment.

**Expermental design.**The experiment is conducted as a approval by the scientific committee of physiology, biochemistry and pharmacology department, college of veterrinary medicine, university of Baghdad, take inconsideration the welfare of animals**.**

**Clinical study**

Forty(40) Balb –c male mice, positively induced diabetic by alloxan single dose 150mg/kg intrapertinally , and daily single dose of insulin 14 Iu/kg.bw S/C , they were divided equally into four groups, and assigned as. Group 1, treated with pumpkin seed oil topically , and mice equally subdivide into two sub groups one of them left till healing but the other mice subjected to biopsy . Group2, treated daily with derma beta® cream topically, Group 3.treated with combination of derma beta cream plus pumpkin seed oil topically , Group 4: was not treated with any materials and considered control group .The mice of G2,G3 and control also divided equally and subjected to biopsy as in G1.

Red blood cells count ,White blood cells (WBCs) count and Platelets count Carried out according to[5]. Differential count of WBCs , Determination of clotting time (per minute) as prescribed by [5].Plasma Insulin Concentration (Iu/ml) , pro thrombin time ( PPT) , thrombin time and fibrinogen level were assessed according to procedures recommended by , [6] [7] respectively .Blood glucose estimated by glucose meter ,clinical observation, measurement the diameter and histopathological observation of wounds along the period of the study were performed .

**Statistical Analysis**

Data were analyzed by using SPSS ,version 21 . Statistical analysis of data based analysis of variance (ANOVA) two ways and significances P< 0.05.Least square differences (LSD) used for comparing between means of all parameters of all experimental groups[8] .

## RESULTS AND DISCUSSION

**The yield Extract of *Cucurbita pepo:*** The yield extract was 58% oil.The oil showed no changes in color, consistency, no smell odor and there were no fungal and bacterial growth. Pumpkin seed oil exhibited highly physical and biological stability ., since there were no undesirable changes occurs upon it till now. They contained the following phytochemicals; steroids, flavonoids and saponin , while alkaloids, phenol, terpenoids, resins, tannins , glycosides and coumarins didn't appear.

**Clotting time (second):** Clotting time of all experimental animals in different groups involved in the study, showed no significance P<0.05 differences, as table (1).

**Platelets count:** After one day of incision the animals of treated groups, 1(PSO), 2 (MEBO) and 3(PSO+MEBO) ,showed significant $p < 0.05$ decrease in their platelet count when compared to G4 (control) . After healing , the treated geoups,1and 2showed no significant $p < 0.05$ changes in platelet count , when compared to G4(control) but only treated group 3(PSO+MEBO)showed significant $p < 0.05$ decrease in platelet count in comparison to G4(control).But when we compared the after one day of incision of the platelet count of all treated group and after healing showed no significant p<0.05 changes ,as table (1).

**The Serum Insulin concentration :** After one day of induced incision ,the serum insulin activity of groups(G1,G2,G3,G4) which where proofed diabetic and received insulin therapy showed no significant $p<0.05$ difference . After wound healing, groups one ,two and three ,showed significant $p<0.05$ decrease in serum insulin concentration ,when compared with G4(control), table 2.

**Blood Glucose Level :**After one day and after complete healing of incision all treated group 1,2and 3 showed significant $p<0.05$ decrease in glucose level in comparison with control group, also the glucose level of post healing showed significant $p<0.05$ decrease in glucose level of all experimental groups in comparison with their glucose level after one day of incision ,table 2.

**Pro thrombin Time** :of all treat groups showed no significant $p<0.05$ difference after one day of incision in comprise to control group ,while pro thrombin time after wound healing show significant $p<0.05$ decrease in all treat group when compared with control group .The only pro thrombin time of healing of group one showed significant $p<0.05$ decrease when compared to it pro thrombin time after one of incision while the other experimental group do this ,table 3.

**Thrombin Time :**The only G3(PSO+MEBO) showed significant $p<0.05$ decrease in the thrombin time after one day of incision in comprise with control group ,but after wound healing all treat group showed significant $p<0.05$ decrease in thrombin time in comprise with control group .The thrombin time after healing of all treat groups showed significant $p<0.05$ decrease when compared to their value after one day of incision ,table 3

.

**Fibrinogen Level** : After one day of incision G1(PSO) showed significant $p<0.05$ decrease in fibrinogen level after one day of incision ,while G3(PSO+MEBO) showed significant $p<0.05$ increase in fibrinogen level when compared with control ,while after wound healing all treat groups showed significant $p<0.05$ increase in fibrinogen level in comprise with control group .When we compared the fibrinogen level between the fibrinogen after wound healing with fibrinogen level after one day of incision of all treat group we noticed the only G1(PSO) showed significant $p<0.05$ increase, table 3.

**The visual observation and diameter measure assessment of wound**

The wound healing of the experimental animals groups , was observed through measuring the diameter of wound and flowed up photographically and histological. Bilateral Circular wounds with diameter 8 millimeter were incised initially in the skin (back region) of all experimental animals, figure 1,table 4.

**Wound after four days of treatment:** The all experimental animals groups, showed no significant difference $p<0.05$ in the wound diameter, figure 2 ,table 4.

**Wound after fourteen days of treatment:** the G3 (PSO+MEBO),still showed significant $p<0.05$ decrease in wound diameter when compared to G4(control ) but not with both G1( PSO)and G2(MEBO), and we also observed there are four animals of each of G1(PSO) and G3 (PSO +MEBO) showed complete wound healing, figure 3, table 4.

**But after sixteen and eighteen days of treatment** also only G3 showed significant $p<0.05$ decrease in wound diameter in comprise with control group and not wealthy that all animals of G3 showed complete wound healing .after 16,18,30, the other G1,G2showed no significant $p<0.05$ change in wound diameter comprise with control group, figure 4, table 4. Finally , after 30 days of experimental the only one animal of each group 1,2 and control had failed wound healing , as figure 5, table 4.

This study has revealed that oil from pumpkin seeds extracted by n-hexane is an important source of many healthy components such as antioxidant and antimicrobial agents. Furthermore, the presence of sterols and polyunsaturated fatty acids in pumpkin oil make it an excellent drug in pharmaceutics and cosmetics which would provide potential protection against skin problem, e.g. dermatological wound. In fact, our findings revealed also that cutaneous wound healing in mice treated with pumpkin oil extract was better than untreated or reference groups by the means of morphometric and histological data. Pumpkin seed oil exhibited highly physical and biological stability since extraction at early 2017 till now , The thought , may be due to the environment of the storage in liquid nitrogen[9] . Pumpkin Flavonoid anti-inflammatory activity, anti-microbial..Pumpkin polysaccharide exhibiter higher cytoprotective effect, indicating that could enhance the cytoprotective effect[11]. Antibacterial characteristics of pumpkin seed oil when applied on infected wounds with pathogenic bacteria ***S.aureus*** that treated infected wounds completely within the course time of treatment. Flavonoids, one of the phytoconstituent were found in pumpkin , and it was thought contribute as antibacterial effects as recorded in different studies that flavonoids possess antibacterial activity[12].

Out of the two types of diabetes, the incidence of non-insulin dependent diabetes mellitus (NIDDM) is much higher than the insulin dependent diabetes mellitus (IDDM). Sulphonyl ureas and few biaguanides are valuable treatment for hyperglycemia in NIDDM, but they are unable to lower glucose concentration to within normal range and reinstate a normal pattern of glucose homeostasis permanently. Use of these therapies is restricted by their pharmacokinetic properties, secondary failure rates and accompanying side effects[13] . onion bulb *(A//ium cepa)* and cloves of garlic *(A//ium sativum)* have long been used as dietary supplement for traditional treatment of diabetes. Former is used as stimulant, diuretic and expectorant[14] . In some plants hypoglycemic effect was demonstrated with extracts only but the active principles were not isolated or studied. Ethanolic extract of seeds of *Zizyphus zujuba* and *Tichosanthus diocea,* were 'reported, to exhibit hypoglycaemic activity .Ethanolic extract of stem bark of *Michelia champaca* (Hindi Champa) showed hypoglycemic effect in rats .Ethanolic extract of root of *Hedychium spicatum* was found effective in rats.Extract of root of *Panax bipinntifidum* and *Hamiltonia scaveolenswere* found to be effective in diabetic mice and rabbits respectively.The research knowledge generated also would be foundational for other researchers to develop novel wound care strategies using essential oils and other botanicals.So we are carried out our research in mice as model for humane and or of we have get promising response for health the wounds in diabetic animal ,this may be encourage for say question for closed trials. Pumpkin works as an antiseptic, astringent and anti-inflammatory and provides detoxifying, soothing and toning support. *Cucurbita Pepo,* pumpkin seed oil a powerful antioxidant and mild retinoic acid substitute [15].

## CONCLUSION

Pumpkin seed oil extracted by n-hexane, where characterized by both microbial and physical stability under storage condition at -170°c and refrigerator temperature.The result of this study showed the pumpkin seed oil contains some effective biological ingredients that have a role in reducing wound inflammation and accelerate healing in both diabetic and non-diabetic mice. when compared to MEBO cream which is used clinically to treat wounds and burns. Pumpkin seed oil, showed no substantial changes in some blood picture parameters (RBC, WBC, platelets) such as blood coagulation criteria ,that play a vital role in wound healing in diabetic mice

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**Table1: Clotting Time/ second and platelet count/ mm³ of diabetic Mice treated with PSO,MEBO and PSO+MEBO compared with diabetic control.**

| groups | Clotting time after day one of wound S.E ±M | Clotting time after wound healing M± S.E | Platelet count after day one of wound M± S.E | Platelet count after wound healing M±S.E |
|---|---|---|---|---|
| **G1 diabetic PSO n=5** | 60 | 60 | $240.00\times10^6\pm105.65\times10^3$ CB a | $252.00\times10^6\pm470.00\times10^3$ A a |
| **G2 diabetic MEBO n=5** | 60 | 60 | $262.66\times10^6\pm333.33\times10^3$ B a | $263.33\times10^6\pm566.6\times10^3$ A a |
| **G3 diabetic PSO+MEBO n=5** | 60 | 60 | $181.00\times10^6\pm110.00\times10^3$ DBC a | $130.66\times10^6\pm933.3\times10^3$ B a |
| **G4 diabetic Control n=5** | 60 | 60 | $379.33\times10^6\pm593.33\times10^3$ A a | $340.33\times10^6\pm393.34\times10^3$ A a |

-LSD of platelet count = $101.36\times10^3$ - capital letters denote significant $p < 0.05$ differences among groups .

-small letters denote significant $p < 0.05$ differences within groups .

**Table 2:Serum Insulin Activity/ IU And Blood Glucose level (mg/dl) of diabetic mice treated with PSO,MEBO and PSO+MEBO.**

| groups | Insulin After one Day M±S.E | Insulin after healing M±S.E | Glucose after one day M±S.E | Glucose after wound healing M±S.E |
|---|---|---|---|---|
| **G1 diabetic PSO n=5** | 87.80±3.27 A a | 78.80 ± 0.58 CB b | 221.40±4.92 DBC a | 114.80±24.23 D b |
| **G2 diabetic MEBO n=5** | 86.40±2.50 A a | 79.60±2.20 B a | 225.00±8.66 CB a | 173.80±17.49 CB b |
| **G3 diabetic PSO+MEBO n=5** | 86.80± 7.46 A a | 65.40± 0.50 D b | 230.00±20.18 B a | 179.60±5.87 B b |
| **G4 diabetic Control n=5** | 85.00± 3.56 A a | 91.00± 0.70 A a | 294.00± 4.01 A a | 206.00±22.45 A b |

-LSD of insulin =7.199 , LSD of glucose=26.80 -capital letters denote significant p <0.05 differences among groups . -small letters denote significant p <0.05 differences within groups .

**Table3. Pro thrombin Time / second , Thrombin Time/second, Fibrinogen /(mg/d L) Level of diabetic mice treated with PSO,MEBO and PSO+MEBO compared with diabetic and control.**

| groups | Pro thrombin after one Day M±S.E | Pro thrombin After Healing M±S.E | Thrombin after one Day M±S.E | Thrombin After healing M±S.E | Fibrinogen after one Day M±S.E | Fibrinogen After healing M±S.E |
|---|---|---|---|---|---|---|
| **G1 diabetic PSO n=5** | 16.63±0.56 A a | 13.36±0.27 DC b | 40.46±1.25 A a | 33.50±0.28 B b | 190±2.72 C b | 284.33±2.33 A a |
| **G2 diabetic MEBO n=5** | 15.46±0.64 A a | 15.20±0.47 B a | 40.93±2.03 A a | 32.00±0.05 CB b | 219± 16.80 ABC a | 220.00±16.37 B a |
| **G3 diabetic PSO+MEBO n=5** | 15.70 ±0.75 A a | 14.46±0.26 CB a | 37.00±4.06 B a | 28.40±0.04 D b | 249.33±31.63 A a | 267.33±33.67 A a |
| **G4 diabetic Control n=5** | 15.63±0.67 A a | 17.26±0.37 A a | 41.13±0.55 A a | 42.10±1.24 A a | 195.33±6.48 B a | 169.00±5.85 C a |

LSD of pro thrombin=1.77, LSD of thrombin = 3.42, LSD of fibrinogen =43.09- capital letters denote significant p < 0.05 differences among groups.- small letters denote significant p < 0.05 differences within groups.

**Figure 1: Circular wounds with diameter 8mm incised in all experimental groups**

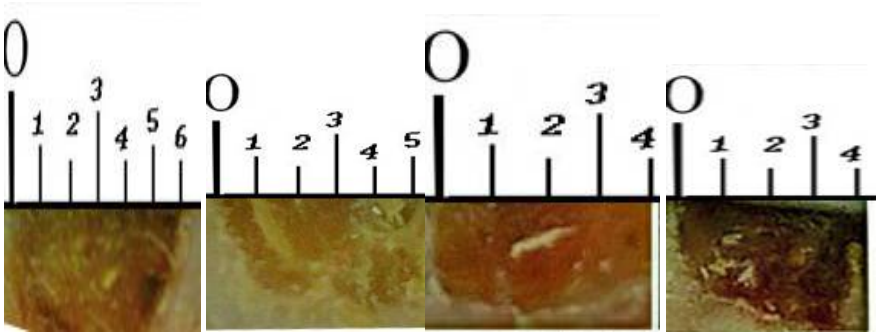

**Figure ,2 : wounds after four days of treatment , the only G3 (PSO+MEBO) showed significant p<0.05 decrease in wound diameter when compared to the G4(control )G1(treated with PSO)and G2 (treated with MEBO)**

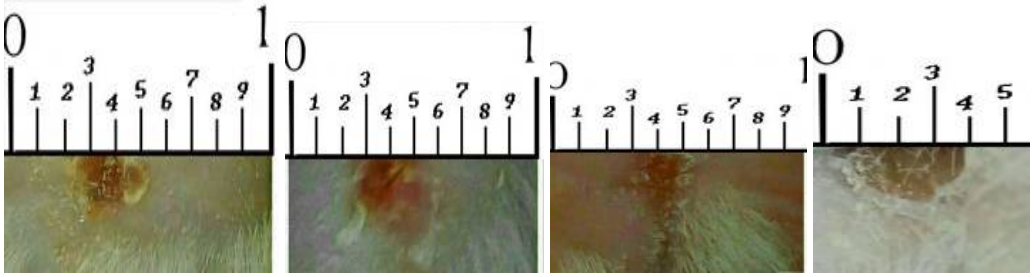

**Figure.3: Wounds of experimental groups after 14 days of treatment .G1 (PSO). G2 diabetic (MEBO). G3 (PSO+MEBO). G4 (control).**

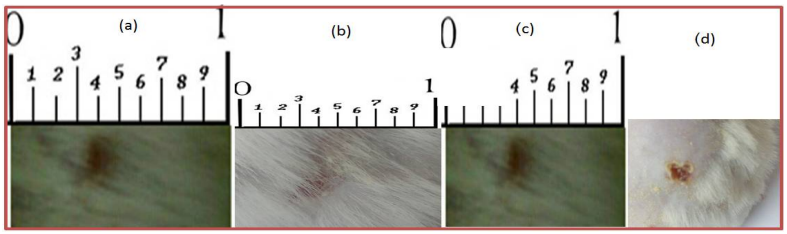


**Figure4: Wounds of experimental groups after 16-18 days of treatment (a) G1 (PSO). (b)G2 (MEBO).(c) G3 (PSO+MEBO). (d) G4 not treated (control).**

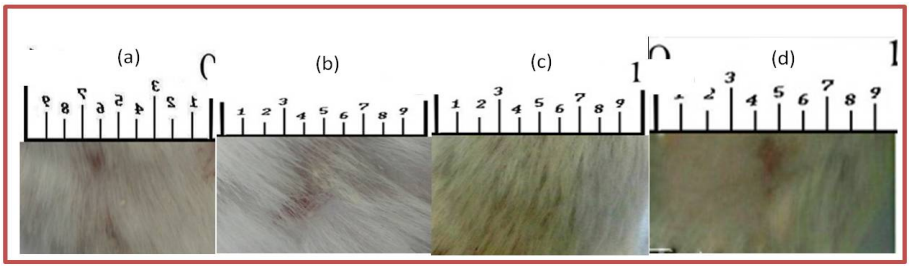


**Figure5: Wounds of experimental groups after 20,30 days of treatment (a) G1 (PSO). (b)G2 (MEBO).(c) G3 (PSO+MEBO). (d) G4 not treated (control).**

**Table 4.Wounds diameter / millimeter of diabetic mice treated with PSO,MEBO and PSO+MEBO.**

| groups | 1day | 4day | 6 day | 8 day | 10day | 12 day | 14day | 16day | 18day | 30day |
|---|---|---|---|---|---|---|---|---|---|---|
| **G1 diabetic PSO n=5** | 8mm A a | 7.20±0.3 A a | 7.20±0.3 A | 5.60±0.7 A b | 5.20±0.5 A | 4.40±0.5 A dbc | 3.20 A | 3.00 A fe | 2.40 A f | 1.00 A h |
| **G2 diabetic MEBO n=5** | 8mm A a | 6.80±0.5 A ab | 6.80±0.5 A | 5.60±0.6 A b | 5.40±0.6 A | 3.80±1.1 AB | 2.60±0.9 AB | 1.80±0.8 AB | 1.20 AB gf | 1.00 A hg |
| **G3 diabetic PSO+MEBO n=5** | 8mm A a | 7.00 .44 A | 6.60±0.5 A b | 3.20±0.7 B c | 3.20±0.5 B | 2.20 ± 0 B ce | 1.40±0.4 B ed | 1.20±0.3 B fe | 0.80±0.3 B gf | …… |
| **G4 diabetic Control n=5** | 8mm A a | 7.20 .37 A a | 7.20±0.4 A a | 5.60±0.4 A | 5.60±0.5 A | 4.40± 50 AB cb | 3.20 AB | 3.0 AB | 2.40 AB e | 1.0 A g |

-LSD of wound healing=1.33 -capital letters denote significant p <0.05 differences among groups . small letters denote significant p <0.05 differences within groups .

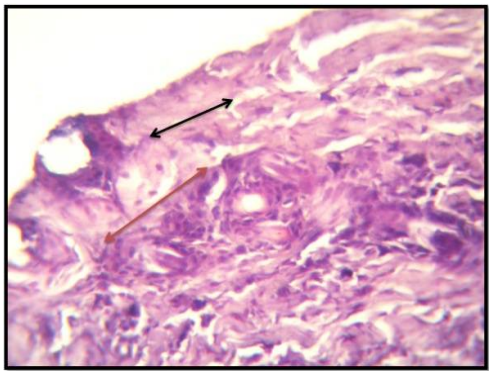

**Figure 1: Histological section in the injured skin of mouse (diabetic treated with PSO), after four days observe , infiltration of MNCs (⟷), Fibrous threads took place and new epidermal layer formed the marginal ends started to thicken (⟷) , (H &E X400).**

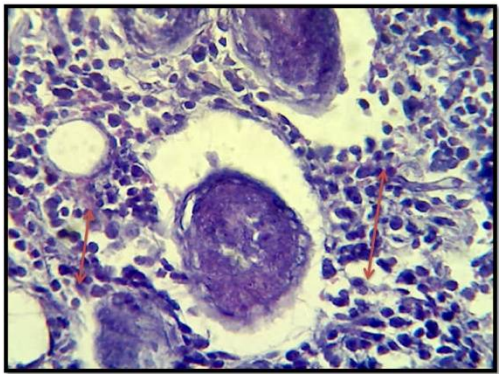

**Figure 2: Histological Section in the injured skin of diabetic mouse post incision and treatment with MEBO+ PSO after four days , observe severe infiltration of neutrophils and MNCs (⟷), (H & E stain 400X).**

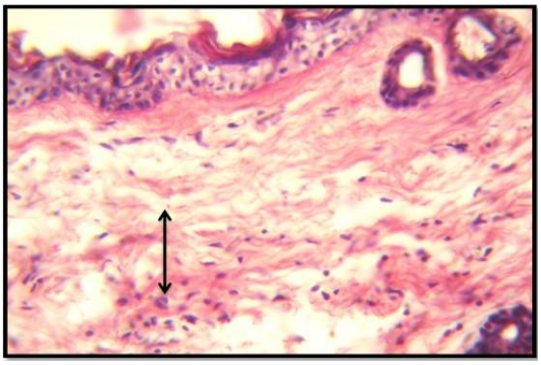

**Figure3: Histological Section in the injured skin of diabetic mouse (control group diabetic) after four days, observe severe inflammatory cells infiltration in dermis with decrease in healing. (⟷), (H & E stain 400X).**

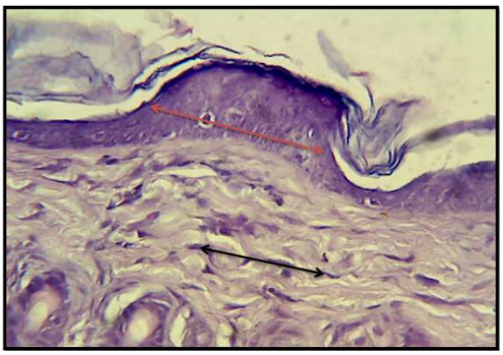

**Figure4: Histological Section in the injured skin of diabetic mouse PSO group after twelve days , observe increase in collagen fiber proliferation (⟷ ) ,(H & E stain 400X).**

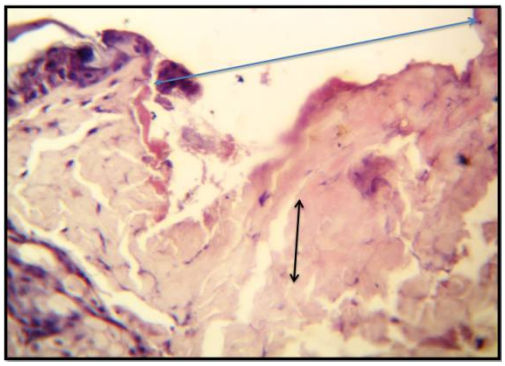

**Figure5: Histological Section in the injured skin of diabetic mouse control group after twelve days, observe failure of wound healing, loss of the epithelial layer of the epidermis ( ⟷) and necrosis of the connective tissue in the dermis (⟷ ), (H & E stain 400X).**

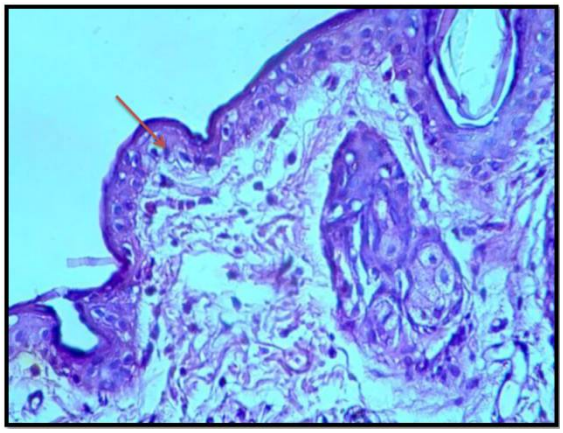

**Figure6: Histological Section in the injured skin of diabetic mouse PSO group after thirty days , observe full thickness of epidermal (⟷) , (H & E stain 400X).**

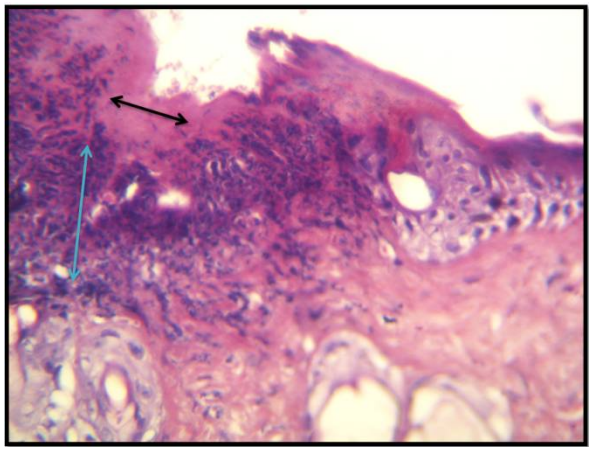

**Figure7: Histological Section in the injured skin of diabetic mouse control group after thirty days , observe loss of epithelial cells and necrosis (⟷) and neutrophil in the epidermis and dermis (⟷ )(H & E stain 400X).**

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**RESEARCH ARTICLE**

# Effect of Ginger Alcoholic Extract on the Glucose Level and Lipid Profile in Diabetic Mice

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## ABSTRACT

Hyperlipidemia is an important risk factor contributing to atherosclerosis in diabetes mellitus. *Zingiber officinale* (ginger) is known for its hypoglycemic and hypochlosteremic actions. The present study was designed to evaluate anti-hyperlipidemic and hypoglycemic activity of alcoholic extract of ginger in experimentally induced diabetic mice. Forty male mice with weight 25-30gm, fed on diet and water ad libitum were divided into 4 groups (10 mice in each group): group I non-diabetic non-treated, group II diabetic non-treated, group III diabetic treated with ethanol extract (300 mg/kg b.w.) group IV diabetic treated with metformin (100 mg/kg b.w.) orally.Diabetes was induced byintraperitoneal Injection of alloxan 100 mg/ Kg b.w.Animals were sacrificed after 7days of induction.The samples of blood were collected by cardiac puncture and serum separated for lipids profile, glucose and liver enzymeestimation. The results suggest a significant anti-hyperlipidemic and hypoglycemic action of *Zingiber officinale* (ginger) alcoholic extract in alloxan induced diabetic mice and may be exploited clinically.

**Keywords:** ginger alcoholic extract, diabetic mice.

## INTRODUCTION

Since ancient times, ginger has been model source of medicines as it contains chemical agents with therapeutic properties. The general population is increasingly using herbal medicines in the form of dietary supplements for the treatment of many disorders [1].Ginger has been used as a medicinal plant in treat stomach upset, diarrhea and nausea for more than 2000 years [2].It is used to relieve throat inflammation and asthma. It is a valued remedy for treatment of coughs and bronchitis. Ginger has also been reported to be effective for the treatment of various abnormalities like inflammation, rheumatism, cold, heat cramps, microbes and diabetes [3].

The mature ginger roots are fibrous and nearly dry. Plants are rich in a wide variety of secondary metabolites including such as terpenoids, alkaloids flavonoids, and tannins etc.The characteristics flavor of ginger is because of mixture of gingerone, shoagoles, gingerols, and volatile oils. The gingerols have analgesic, sedative and antibacterial properties [4]. Pharmacologically, the plant (fresh rhizome) or its extracts have been investigated for its hypoglycemic [5,6]hypocholesteromi[7,8,9] anti-inflammatory [10,11] anti-microbial[11] and anticancer activities[12].
The objective of the study was to evaluate antihyperlipidmic and hypoglycemic properties of ginger alcoholic extracts in diabetic mice.

## MATERIALS AND METHODS

### Collection of plant

The fresh dried ginger rhizomes that collected from local markets in Baghdad were ground to fine powder by electrical grinder. This powder was stored in airtight bottles and subjected to various extraction procedures.

### Extraction

To prepare alcoholic (ethanol) extract 10 g powder of ginger was soaked in 100 ml ofethanol(70%). The flasks were put on magnetic stirrer for 72 hours with shaking at 45°C. The solution was then filtered using muslin cloth. The supernatant was again filtered using Whatman filter under strict aseptic conditions and the filtrate was collected in fresh sterilized bottles and stored at 4°C until further use.The crude extractswere centrifuged at 3000 rpm for 10 minutes at 25°C.The ethanol extracts was evaporated at 50°C. All dried extract samples were dissolved in distilled water to the final concentration of 100 mg/ml and centrifuged again at 10,000 rpm to remove the undissolved residues. The extract solutions were stored at 4°C [13].

### Animals

Male mice weighing between 25-30gm were housed in air-conditioned animal room at 22±2$^0$C and were fed on standard mice pellet diet and allowed to drink water ad libitum.

### Experimental induction of diabetes in mice

After 24 hrs fasting, micewere injected alloxan 100 mg/ Kg b.w.ip. Blood glucose was estimated after 72 hrs from alloxan injection by placing a test strip in the glucometer. A drop of blood was collected by aseptically cutting the tail at the tip (0.1 cm) with sterile blade and then applying the drop of blood to the test area of the strip. Mice with blood glucose of higher than 11.5mmol/l (72 hrs after alloxan injection) were considered diabetic and selected for experimentation [14].

### Experimental design

Forty male micewere divided into 4 groups (10mice in each group): group I non-diabetic non-treated, group II diabetic non-treated, group III diabetic treated with extract (300 mg/kg b.w. orally) andgroup IV diabetic treated with metformin (100 mg/kg b.w. orally).

### Estimation effect of *Zingiber officinale* (ginger) alcoholic extract on blood glucose level, lipid profile and liver enzyme activity in diabetic mice

After 72 hours, blood glucose levels were measured from tail-vein blood of all groups by glucometer, level higher than 11.5 mmol/l considered as diabetic. Standard drug metformin(100 mg/kg) and extracts (300 mg/kg) were administered once daily for seven days to diabtic mice groups. Blood glucose levels were measured after 7 days by glucometer. After completing antihyperglycemic test all mice were sacrificed and about 2 ml of blood was collected directly from aorta of heart by syringe. Serum samples were obtained by centrifugation of blood at 3000 rpm for 15 minutes. The concentration of serum lipid total cholesterol (TC), triglyceride (TG), low density lipoprotein (LDL),

high density lipoprotein (HDL), aspartate aminotransferase (AST), alanine aminotransferase (ALT) and alkaline phosphatase (ALP) levels of mice were detected[14].

### Statistical Analysis

Statistical analysis of data was performed using SAS (Statistical Analysis System - version 9.1). One-way ANOVA and Least significant differences (LSD) post hoc test was performed to assess significant difference among means.. $P < 0.05$ was considered statistically significant (SAS.2010.SAS/STAT Users Guide for Personal Computer. Release 9.1.SAS Institute, Inc., Cary, N.C., USA.

## RESULTS

### A.Effect of *Zingiber officinale* (ginger) alcoholic extract on blood glucose level and lipid profile in diabetic mice

From the data presented in table 1, it is observed that the administration of aloxan induced hyperglycemia and hyperlipidemia in mice. Administration of extract at 300mg/kg body weight showed a significant reduction in the levels of blood glucose, serum total cholesterol, LDL, triglycerides and increasing HDL levels in comparison with metformin treated group.

### B.Effects of *Zingiber officinale* (ginger) alcoholic extract on liver enzyme in diabetic mice

In this study, serum alanine aminotransferase (ALT), aspartate aminotransferase (AST) and alkaline phosphatase (ALP) activities were significantly high in diabetic than in normal mice. On the other hands, the extract revealed a significant reduction in liver enzymes (table 2).

## DISCUSSION

Diabetes mellitus is a chronic disorder produces inadequate blood glucose control and leads to acute and chronic complications, overt symptoms of diabetes mellitus include hyperglycemia, and hyperlipidemia[15].The present study was undertaken to investigate the effects of *Zingiber officinal*(ginger) alcoholic extract on glucose level, lipid profile and liver enzyme activity in alloxan induced diabetic mice compared to normal non-diabetic controls. Injection of alloxan(100 mg/ Kg b.w,i.p) produced marked hyperglycemia and hyperlipidemia (increased total cholesterol, LDL& TG and decreased HDL). Treatment with *Zingiberofficinale*(ginger) alcoholic extract (300 mg/ Kg b.w, p.o) for 7 days to alloxan induced diabetic mice produced significant blood glucose and lipid lowering (decreased total cholesterol, LDL & TG and increased HDL) effects. Thus suggesting a significantanti-hyperlipidemic and hypoglycemic action for *Zingiber officinale*(ginger) alcoholic extract in alloxan induced diabetic mice.A similar result was reported by [6]in their study on the effects of ginger juice in diabetic rats.Theresults are in agreement with those of previousstudies[7,8,11,16]who showed similar lipid lowering effectsof *Z. officinale* in different experimental animal models.also in agreement with [17] and [18] who reported that ginger reduce the liver enzyme which followed the hyperglycemia and hyperlipidemia.

The major pungent component of ginger is gingerol [19].Gingerols can be converted to shogoals and zingeroneby dehydration and retro-aldol reaction, respectively. These ginger components havebeen shown to have a variety of pharmacological effects In addition,Akhani*et al.,* [6]reported that ginger extract partiallyalleviated the hypoinsulinaemia observed in induced diabetic animals.

This hypoglycaemic action of ginger maybe due to effects involving serotonin receptors, an increase inpancreatic secretion of insulin from b cells or release of boundinsulin.The plasma lipid lowering effect of GE is possibly associated with several processes, including disruption ofcholesterol absorption from the GI tract [20] and interference with cholesterol biosynthesis inliver [8,21]. Several lines of evidence revealed that ginger containsantioxidant properties which have a hypocholesterolemic effect and anti-atherogenic, and these activities mightbe attributed to the inhibition of LDL oxidation and the suppression on the activity of HMG-CoA (3-hydroxy-3methyglutaryl co-enzyme A) reductase[14,22,23]. This also might occur due tothe elevation of hepatic cholesterol 7-alpha-hydroxylase activity, which is a rate-limiting enzyme in thebiosynthesis of the bile acids and stimulates the conversion of cholesterol to bile acids leading to the excretion ofcholesterol from the body[24]. A significant decline in serum lipid profile observed in ginger extract treated mice suggests the atheroprotective potential of this herb. Ginger is known to retard thedevelopment of atherosclerosis [25].

The serum ALT, AST and ALP levels were extensively elevated in diabetic than in normal mice. This may due to the disturbance of lipid metabolism resulting in accumulation of TG in liver and an increased increment of the liver index, ginger flavonoids may have an additive effect to the endogenous scavenging compounds as they can increase the function of the endogenous antioxidants. In addition, flavonoids and terpenoids may reduce total cholesterol, TG, and LDL through inhibition of pancreatic lipase which responsible of liberation of triglyceride into fatty acids and glycerol [26].Further studies are suggested for investigating possible mechanisms of action and the active components of ginger responsible for the observed beneficial effects in the diabetic condition. However the findings itself are of great interest and significance; and can be clinically exploited.

## CONCLUSION

The results suggest a significant anti-hyperlipidemic and hypoglycemic action of *Zingiber officinale* (ginger) alcoholic extract in alloxan induced diabetic mice and may be exploited clinically.

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**Table-1: Effects of *Zingiber officinale* (ginger) alcoholic extract on blood glucose level andlipid profilein diabetic mice.**

| Groups / Test | Non diabetic non treated | Diabetic non treated | Diabetic treated with ginger alcoholic extract (300 mg/kg b.w.) | Diabetic treated with metformin 100mg/kg |
|---|---|---|---|---|
| Blood glucose(mmol/l) | 6.52±0.10 D | 17.54±0.09 A | 7.60±0.07 B | 6.80±0.08 C |
| Total cholesterol(mg/dl) | 106.60±2.0 B | 254.70±6.38 A | 116.20±2.91 B | 108.70±1.21 B |
| HDL (mg/dl) | 38.30±0.22 A | 30.60±0.37 D | 34.50±0.40 C | 36.50±0.55 B |
| LDL | 46.50±1.19 | 200.00±2.63 | 56.00±1.74 | 51.00±2.03 |

| (mg/dl) | C | A | B | BC |
|---|---|---|---|---|
| TG (mg/dl) | 92.00±1.44 C | 226.00±2.46 A | 108.00±1.56 B | 96.50±1.79 C |

The different capital letters refer to significant differences between different groups at (P<0.05).

**Table-2: Effects of *Zingiber officinale* (ginger) alcoholic extract on liver enzymes in diabetic mice**

| Groups / Test | non diabetic non treated | Diabetic non treated | Diabetic treated with ginger alcoholic extract (300 mg/kg b.w.) | Diabetic treated with metformin 100mg/kg |
|---|---|---|---|---|
| AST(U/l) | 143.70±2.33 C | 185.20±2.16 A | 165.00±3.38 B | 159.00±6.09 B |
| ALT(U/l) | 58.20±2.23 C | 82.70±2.57 A | 66.80±2.58 B | 61.00±1.78 BC |
| ALP(U/l) | 54.60±1.77 D | 84.20±2.28 A | 70.00±1.67 B | 64.00±2.24 C |

The different capital letters refer to significant differences between different groups at (P<0.05).

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RESEARCH ARTICLE

# Heat Wave & Sun Stroke: Mechanism & Management

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## ABSTRACT

Heat wave is an ecological phenomena, meteorologically synoptic in scale. There are no medicines. Sun stroke kills. It is Heat wave dependent. There is need to understand the (complex) nuances. Data is wanting.Indian subcontinent's unique geographical location, geospatial architecture, latitudinal extent, geomorphology and orography because annual-seasonal meteorological variations (of variable contrasts) that swings extensively & variedly over such large geography; a Billion population and high breed rate; sole civilization on earth in continuum for 5000yrs. Hence, the parameters are studied from known positions/schools of the sciences and correlated with her deep heritage (backward synchronization) ranging two decade, sub-continental wide, also covers 2 millennia of traditional/classical literature. Multi-disciplinary study. Multi-lateral initiative shall be more beneficial. Fall in soil moisture (ecological phenomena) causes heat wave (meteorological phenomena). Jointly, becauses sun stroke (physiological phenomena). Mother nature annually auto reverses heat wave & soil moisture. Sun stroke has thus far remained non-reversible, associated with hemodynamics failure cum gut collapse (original 1st time findings) apart hypothalamus impotence (thermal shock- pseudo anaphylaxis). Anabolic stage endure stress well. Menopause & geriatric stages more & acutely susceptible, respectively. Meteorological mechanics and pathophysiology of sun stroke; home based remedies; clinical anti-dotes have been discussed. Scientifically evaluable & co-relable. India has a plethora of native heritage practices vis-à-vis caption domain. Also has an enviable range vis-à-vis heat wave resistant home stead design and dress code. Very little is known about heat wave-sunstroke causes, processes and effects. Less is known about mankind's heritage in this regards. Multi-lateral initiative shall be more beneficial.

**Keywords:** Heat wave & Natural signs; Sun Stroke & Symptoms; Mechanics of Heat wave; Pathophysiology of Sun stroke. Soil moisture Loss; Indian Native home stead designs; Indian heritage food;

## INTRODUCTION

Heat wave and sun stroke have been established as couples. Yet, no in-depth understanding exists about the mechanics of this couple. Further more, there is also a need to understand the mechanics of heat wave, soil moisture

loss and the mechanics of sun stroke separately. It is said 60% of the Indian sub-continent experiences varied periods of heat wave cum sun stroke conditions between March to July, annually. This is also the field work period for all national and macro level projects. Therefore, the public (specially field engineers) should know as much as possible.

**What is Heat Wave ?**

Heat Wave (HW) is a meteorological phenomena. The India Meteorology Department (IMD) defines HW on the basis of recordings done at two contiguous met stations. For stations that have a normal summer temperature range of 40-45$^0$C wherein an excess by an order of 5-6$^0$ C is classed as 'Normal Heat Wave', while an excess by an order of >7$^0$ C is classed as 'Severe Heat Wave'. For stations that have a normal summer temperature range of >45$^0$C, an excess by an order of 4-6$^0$ C and >6$^0$ C are classed as 'Normal Heat Wave' and 'Severe Heat Wave' respectively **[1]**. Thus, the Indian official scheme relate HW solely to temperature anomaly in relation to place and time. In fact, not so simple.

HW is a borderless meteorological phenomena. It effects large geo-spatial domains and is a severe ecological stress creator. High population density coupled with industrialized anthropogenic lifestyle in hot regions (additionally) fold multiplies the anthropogenic factors of the stress on to the ecology, raising such stress to 'acute levels'. Sun stroke (SS) is its signature on biological beings only. HW affects the soil moisture flora, fauna and the anthropomorphs (humans). Primarily HW is catalysed by adiabetic pull i.e., inversion (inspite heavy updraft of moisture). Such condition connotes loss of moisture at surface. In other words, SS is the earliest visual signature of the (i) 'acute levels' of impending ecological stress (ii) biological stress. Unmanaged, it can keep auto intensifying, geographic expansion, positing as a obstinate met phenomena i.e. generation of 'low work done potential ecology', *alias* (iii) aridification. It is the signature of the point of 'no-return' **[2]**. This more creates human security problem**.**

**Heat Wave Mechanism**

What is the effect of heat wave ? The effect of heat wave results first in drying of water bodies progressing from shallow to deep sources; fall in water table; loss of soil moisture; caking and pulverization of top soil; pulverization; dust making; gets air borne; onsets wind erosion cycle (apart massive flow out even with moderate rains). Intensification of such cycle with every passing year. As soil moisture gets low, flora and fauna (animals & humans i.e. biological life) also start loosing physiological fluid resulting in wilting and drying of flora and fatality of the fauna (blood, lymph, serum in anthropomorphs & theriopomorphs. Water & sap in fauna constitute physiological fluids. Most vital.). Historically floral cover acts as the anti dote. However, flora cannot survive sans soil moisture. Moisture i.e., relative humidity (RH) experiences osmosis efficiently. Soil moisture (SM) cannot remain sans a supporting atmospheric RH. Thus HW is a cascade. The end product is draught. Sun stroke (SS) is a constituent part of HW. In the Indian sub-continent, HW is triggered when the seas around have warm SST conditions with the Sun being aligned to the N of 10$^0$ Lat. (as in 2016). Such combination results in intense and as well in long period episodes of HW and SS conditions encompassing large swaths of the Indian peninsula. Hence, HW and SS conditions are preceded by soil moisture loss. Thus a multi-disciplinary approach is warranted.

Fig.1 schematic explains HW phenomena over domain Odisa. We can see the Sun is aligned (noon time India) at a location which has one side facing the sea and three sides land locked. The incident sun rays inject astronomical energy in form of heat into the top soil at location of loss of soil moisture (SM). The region to its east is marked 1 (is of relative higher soil moisture and atmospheric RH). The region to the west of location SM is marked as 2 (is of relative low soil moisture and atmospheric RH). Depending upon, geomorphology, physiography and floral surface cover such separation occur. This is our field observation based theory. The theoretical rebound of the astronomical heat is projected at point X. Line SM-X is west to east moving phenomena, i.e. region-2 keeps on expanding while region-1 gets on constricting. Heating generates 'low' alters the architecture and the spacing of the barometers, additionally with reducing variations between day and night. From direction east (sea side) two vectors marked W1

& W2 having variable RH and speed are flowing in towards SM. These are easterlies/sea breeze colloquially better known as *Chaiti* (breeze that blows in the month of *Chaitra*). Inland RH is logically to be low. The kinetic component of W1 & W2 vector the RH from sea towards SM. However SM cannot be imputed into the soil as because the central and western Odisa regions (and further inland) have lower RH. Thus, ever increasing sea sourced moisture is designated for central Indian peninsula and not for western Odisa *per say*. This includes SM (from every point) that is expressed via evapotranspiration is eventually vectored inland (region marked 2) at angles $\theta 1$ & $\theta 2$. No. 3 is the meteorological boundary layer. No. 4 (semi-parabolic arcs) shows the gradual lowering of the theoretical T-$\Phi$ line (therm-fi) and its migration towards the west and south west. Wherever, SS. By juxtaposing Fig.1 and Fig.3, we come to know, that the T-$\Phi$ line is dependent on SM loss and its ingression towards the eastern coast. Geographically, the horizontal line represents an average of the Tel-Mahanadi-Brahmani drainage systems. The western coast has the coastal mountain range. It is a natural barrier. The eastern coast is aligned to the latitudes and logically to the natural angulation of the geostrophic winds (Fig.-1 is a $1^0$ map). Therefore, the eastern shore board is the supplier of moisture, while the west looses it to the desert regions that are in its beyond. Therefore, SM can be the most effective anti-dote. Non else. Furthermore, the *chaiti pabana* (wind, 20-30Knots) has a connection with the moon rise, set and zenith pass timings and alignment. In 2016 the moon had extreme northerly swing resulting in very strong *chaiti* **[3]**. We have used Odisa as because we have conducted our experiments and validated them in such region (over 2 decades).

In Fig.2. we present a Satellite image [**4]** from geo-stationary position dt. 21-12-2008 at 06UTC (Capricornian solstice-India noon time). The incident Sun rays refract towards the central line as it enters the dense atmosphere which has a concavo-convex architecture as is done while 'focusing'. The constricted focused region of the sun is well reflected (only) on the ocean water. Meteorologically this is the effective sun's size based coverage on the terrestrial surface (post lens effect). We have marked the place in the Indian ocean with a circle. We have then drawn an identical circle with central India as the centre (for on 21st June of every year the focus region gets to be aligned also with India). We note that whole of the Indian nation comes virtually within the swath of the focal region. Focus means intensive injection of solar energy. This cannot happen sans the 'lens effect' of the atmosphere. 'Lens effect' is a blessing for biological evolution, mitosis and plurality (*Veda*). Most of the regions that come under the glare of the 'lens effect', are home to lush green equatorial or salubrious tropical flora, fauna and ancient civilizations. Therefore, temperature is not the villain. It is lack of RH which is the real villain. The IMD has eminently used a simple classification based on field work, hand collected, long period average data (temp. & RH). It has not considered the laws of physics neither the principles of fluid mechanics nor physiology, geography, geomorphology or soil moisture or the SST anomaly. It has neither adopted our type of methodology to explain even to the select quarters of the administration the 'why & how' of heat wave. We have herein taken these parameters, because an understanding is paramount and it is a national requirement. Our efforts confirm that it is the astronomical energy that is the cause of heat, while low RH is the cause of its wave like propagation, and lack of SM is the cause of Sun stroke i.e. heat wave and sun stroke has to be related to SM, RH, SST, Physiography, Flora cover, direction of ground breeze and the coefficient of the evapo-transpiration on macro-to-regional geo-spatial scale. And, IMD only talks about HW and its's model is plaintive. It does not talk about ecological i.e. phyto, agro, veterinary and human stress or security (sun stroke), management, etc. The fact is HW may or may not generate SS conditions (socio-economic shocks). Planning cannot afford (to factor in) such large scale natural loop holes. Millions cannot any further be left in the lurch. The nation has to go beyond the term 'heat wave' for micro-macro-regional planning for obtaining assured better results thereof from investments. Our study posits in such directions, also.

**Soil Moisture Loss**

What is the mechanism of SM loss ? In brief, the principles of thermodynamics states that heat is energy. It comprises of particles which in compressible media propagates in wave form due to (thermal) gradient. In atmosphere, it is vectored by wind and is guided by boundaries (solid/liquid), with which it interacts via convection and forms couples by the process of absorption and radiation via an delayed action mechanism (specially

when incident). Energy absorption and retention is better in the presence of liquids (dissolved or suspended). Clay is such an candidate. Therm absorption in soil medias is always in form of small wavelets and the return radiation is in the form of outgoing long wave radiation (OLR). It is expressed in units of Watts/$m^2$, an is read as (cloud) top temperature. It has an property to warm the moisture. Energised moisture become buoyant and shear away. Thus, post noon the room interiors have higher trapped moisture as expressed by anthropogenic activity. Higher RH down regulates the adiabatic process and destabilizes its architectural field. Low RH enlarges and stabilizes. Hence, post noon humans and animals experience heightened levels of perspiration. It is also felt outdoors in breezeless conditions (atmospheric stall). Therefore fatigue is experienced at dusk. At this juncture, exercise mechanically assists evapotranspiration and resultant heightened oxygenation of the tissues dispel fatigue. Hence open field games are played in the afternoons, while in-auditorium afternoon play is tiring.

Fig.-3 is that of the mean OLR for Indian post noon for Capricornia solstice 2017 **[5]**. We can see that as early as Mar., the whole region of Gujarat, Maharastra, parts of MP & Karnataka are yielding high OLR (300w/$m^2$), with one branch poised towards western Odisa. This means the atmosphere of this region is relatively breezeless and the lithosphere of these regions are expressing moisture into the atmosphere and that it has a tele-connection with the Somalian desert (bridging the Arabian sea at 1.5Km above the MSL). We are of the opinion that this region is the mother bed of soil desiccation and HW in India. It eventually expands till July through April-June). OLR is always a vortical phenomena and is followed by vectoring of the moisture specially during the nocturnal hours. It results in irretrievable loss of moisture from a given place. OLR contour change least during waxing phase of the moon, because lunar gravity imparts heightened buoyancy to moisture and makes it 'stand still'. A stalled moisture field does not allow column pressure to alter from pocket to pocket. This phenomena down-regulates surface breeze. The day next HW is more intense. HW is observed to be associated with diurnal moon pass ($4^{th}$ quarter to 72hrs post new moon phase).

Moisture is the voluminous component of the atmosphere. Alteration in moisture content inflicts change in volume, convection, condensation, collation, trapping and release of radiative therm, etc., processes (in the\of the atmosphere). Therefore, SM acts as a (delayed action) regulator. It thwarts abrupt extreme swivels in RH and temperature. It up-regulates precipitation. Due to such mechanics, therm and volume also form a natural preferred pair and in turn also inflict variability in barometric pressure (hPa). It is associated with convection and non-uniformity in the temperature & vortical conditions within the atmospheric column height. Such unbalanced state of energy is called *enthalpy*. It is a state of dynamic motion which imparts higher 'work done' abilities. Dynamic motion, makes more moisture available (Fermions that behave alike Bosons). This is a loop. Therefore *Enthalpy* is associated with absence of HW and or SS (even in *loo* periods in core *loo* domains). Opposite of *Enthalpy* is baroclinic conditions, when the uniformity in the column height of the atmospheric temperature rises to higher altitudes i.e. it becomes more uniform (always being associated with low RH) with a reduction of work done ability. It is a sort of 'inversion'. Low RH occur due to a non-participating soil (low SM). Therefore, only baroclinic conditions is conducive for HW. Which in turn induces SS and it cannot germinate if SM is normal.

**Urban Heat Island**

In India, urban heat island is a phenomena of the post 1970. It means that the temperature profiles of the urban regions stand out in contrast to the neighbouring rural regions i.e. the urban centre will be recording high diurnal temperature, while there will no change (say)100 kms away from its periphery. Hence it is an island. Bhubaneswar is Smart City Candidate No.1. The twin city of Bhubaneswar-Cuttack is such a case. Large scale use of reinforced concrete cement (RCC) have been used to quick build with comparative economy, large urban conglomerates and industry centers. The structures include (i) buildings (ii) roads. The surface area exposed to the incident solar radiation is many times more. The thermal conduction and 'heat sink' property of RCC is much more than soil. There is a commensurate reduction of soil surface area with heightened anthropogenic use of water table resources. Used water becomes surface run off (mostly in covered drains) and does not seep underground. All these were never

there prior to urbanization/industrialisation. Urban centers built primarily of RCC act as alike 'heat radiators', only during tropical summer conditions. Due to fold increase of surface area and therm entrapment, it creates a local pocket of heightened heat between the local hours of 9AM to 1PM. During these 4 hours, a strong up-draft mechanism is triggered. If there be at the same time (a) strong surface breeze extending over a synoptic domain (b) or if it be 'low' RH period then up-drafted moisture from local/regional scale will be vectored away in a no immediate return mechanism (lateral transportation and adiabetic loss), as that happens in region(s) marked as 4 in Fig.1., during *Chaitra* (Mar-Apr.) in Odisa.

Our Fig.4. schematically explains urban heat island phenomena in and around Bhubaneswar-Cuttack twin city region. We can understand that RH from the twin city region will more efficiently be expressed towards the central India via the mechanism explained in our Fig.1. Long period repetitive occurrence of such a mechanism inflicts lowering of water table even in annually long period flood prone regions like the Brahmani-Mahanadi delta. Due to water training hydraulic devices (quick flood less flow back to the sea), such mechanics gets robustised. Atmospheric and soil trapped fluid flow is thence towards the Urban Heat Island. In the immediate vicinity it causes severe and occasionally acute water shortage at surface level. Erstwhile arable lands tend to get fallow. Crops wilt, productivity drops, tolerance to fertilizers and insecticides diminishes. Moisture stress heightens, window widens. The moisture in the hay and live stock feed reduces, so do the hay staple size and thickness. Incidence of large scale swift spreading village fires are reported. Yield from domesticated milching cattle also takes a down turn. Incidence of skin diseases take a up-turn. Over decadal periods, top soil becomes powdery and is efficiently transported away over long distances even by small episodes of rain or made air borne by coastal wind (land breeze in particular) and anthro activity or cattle graze (*godhuli*). Thus, Bhubaneswar is getting more dusty by the year. During the no-rain period the dust gets air borne and traps radiative heat, creating green house conditions around urban centers. All these because of (modern plan period) anthropogenic induced localised heightened seasonal evapo-transpiration. Such a phenomena occurring in central and western Orissa, will draw out much (additional) moisture from the neighbouring deciduous (even reserve) forests and subsequently also from the coastal regions and will inflict much damage to the flora (same along west coast of India) i.e. such a heat island phenomena is already occurring along the Rayagada-Titlagada-Balangir-Bargada-Sambalpur-Jharsuguda-Bilaspur-Raipur-Kantabanji axis (circular & enlarging); includes urban centers. This is triggeing irreversible aridification mechanism with disastrous consequences on coastal fragile ecology having wider domain ramification **[6]**. On the Indian sub-continent aridification has been progressing from across Paki into Indo; into Paki from Arabia & into Arab from Sahara ~ The maintainer of humidity (else there would be too much humidity as because moisture induces/assist dynamic motion. This makes more moisture available i.e., Fermions that behave alike Bosons (Our Personal communication to President IMS, as part of project VLRF-2017).

Heat islands up-regulate SM loss. Its genetic bed is Dharwad-Vidharbh region. It is synoptic in scale. Man made water bodies are thin films of liquid. They act as anti-dote **[7]**. Therefore, creation of water bodies (has to be part of the techno-socio-economic plan of India) in and around every urban centers having a surface area calculated as per $m^3$ of the aggregate of the structures in a given urban centre. Roughly (for planning purposes) a range of 1:16-20$m^2$ is advocated as an effective counter measure. The water bodies (thin film liquids at surface) preferably be located to the NE or SE (best either) of urban/industrial centers that are to the west of $78^0$ E meridian, and to the NW or SW (best either) of urban/ industrial centers that are to the east of $78^0$ E meridian. Presently, storm & flood water (which is sweet & fertile silt laden) is trained back to the saline sea via the shortest and swiftest path). Instead, flood water storage is strongly recommended – longest circuitous path. A paradigm shift in mind set (towards this all encompassing problem) and in engineering priority practices is needed. If we juxtapose our Fig.-1 with Fig.-3. we then note that it mimics an all India scenario and is helpful in understanding the mechanics that is atypical as well as region specific.

**Natural Indicator**

Human beings i.e., antrhropomorphs are homeotherms (can control body temperatures) and are also categorized as warm blooded creatures. Snails; fishes; snakes; lizards, fleas; flies and birds are Poikilotherms (cannot control body temperature) and are also categorized as cold blooded creatures. HW effect on homeotherms is preceded by noticeable effect on poikilotherms such as absence of fleas, flies, dying birds, while the non climber reptiles often come on-ground (abandon Burroughs) or trying to climb trees, total absence of butterflies, juvenile sized leafs falling off from trees (off season *patjhar*); budding failure; wide spread viral attacks on domesticated animals and pets, many similar signs - a series, pre human fatality.

**Sun Stroke Aspect**

It is a clinical condition, albeit, the clinicians (not fundamental scientists) have vague explanations. Clinicians variously describe often with classical Greek & Latin literary phones (assume them to be scientific terms). The used clinical terms do not describe the mechanics nor the kinetics. Only narrate the observed. SS is marked by high pyrexia, manifestation of a varied series of individual specific clinical symptoms leading normally to morbidity and consequent fatality all along associated with hypovolemia (physiological fluid depletion). In general, it is attributed to expouser of the human body to extreme heat wave for prolonged periods. SS is an infliction by the earth science processes on to the biological systems. In the previous centuries SS was related to sun light i.e., lumen (not heat) and to the eyes and nape **[8]**. This is pseudo comfort, permits the body extended expouser which portends ill.

It is said that, SS can happen to all. This is a fallacy. SS transpires out of evapo-transpiration management failure at individual level during freak or seasonal HW episodes. In other words, SS need not affect the flora or the fauna before striking anthropomorphs (can be reverse). SS can strike an individual who is not accustomed to summer heat and or an individual who has impaired gut; damaged brain; underfed; wrong diet; the geriatric in general. As a rule, annually, when soil moisture gets to be very low, the other biologicals (flora & fauna) start loosing moisture (via osmosis). This is meteorologically termed as 'HW' and medico-clinically termed as 'SS', whereas it is directly related to soil-moisture loss. SS kills, for it depletes physiological fluid balance hypovolemia (tissue perfusion failure)→neuro-muscular morbidity → shock → seizure → demise. Hypovolemia is also a major cause of *myocardial infraction* (heart attack), prior to that cerebral black outs and necrosis occur (heart is spared). HW conditions up-regulates each and every pathology thereby heightening & hastening fatal conditions. The real cause of demise may be some other underlying cause and not HW or SS.

**Six Symptoms of Sun Stroke**

As the (human/animal) body loses fluid, at first the gut starts loosing fluid with constriction of lumen's cross section. Food starts drying up, the gut goes into a override with heightened peristalsis, it registers as stomach cramp (1st symptom), lumen villis are severely adversely effected causing failure of food/drug uptake mechanism (salt, ion & minerals in particular from gut) which has long term debilitating effect (reverses very slow). It is followed by genesis of a gradient from the lymph towards the vasculatures (a surreal situation), for the lymph nodes are 'one way lock gates' (lymphatic drainage stops). They shut down in an effort to restrict lymph loss. The heart initially goes into an over ride eventually lapsing into low stroke volume and ectopic syndrome. Deep seated numb pain embalms the body. Thereafter, the exposed muscles start contributing salt, ion and fluid. Whole body musculature starts acutely aching known as *myalgia* (2nd symptom). The ion mechanism pathways in the body gets askew, with head ache (3rd symptom), when not attended to effectively, palor (fixed eye balls) and neck slumping occur as in cerebral malariasis/kala azar, etc.(one of the terminal symptoms). Thereafter, follows failure of signal transduction pathways and collapse of hemodynamics with concurrent renal shut down. To overcome, the brain first marginally raises the body perspiration which soon fails (because of progressing hypovelemia), in response to 'sweat failure', it whips the heart (by expressing *aldarin*) into a override, sans any hyper tension of the systole or the diastole eventually lapsing

into acute hypotension (non traceable vein). At this juncture having Beetle nut in mouth spells irreversible doom (in non stroke state it is good). The brain seizes not, it continues to senses danger to itself, therefore, it then extensively relaxes the autonomic muscles initially of the lower limbs resulting first in 'unsteady steps' ($4^{th}$ symptom) followed by 'sleepy feeling' / black out ($5^{th}$ symptom). With the fall, public attention is drawn towards the stricken and casualty is reported. Process fatality (cascade) has long set in by then (almost too late). Immediately post the $4^{th}$-$5^{th}$ symptoms, something very extraordinary 'reaction' manifests, the brain shuts down the temp regulator sub-organ (*hypothalamus*), which makes the human physiology a '*homeotherm'* (warm blooded creature seem to change phase to that of a cold blooded creature-*poikilotherm*). It results in soaring of peripheral temperature clinically articulated as 'pyrexia' ($6^{th}$ symptom). All this happens in a cascade mechanism as part of 'homeothermic effect' (physiological systems that can regulate own body temperature).

SS is termed as 'stroke', for it is much alike 'anaphylactic shock', yet there is often no seizure nor any hyponatraemia. Even, there is a draw back of fluid from the lymphatic network (spell grave prognosis). Thus, SS is a state of failure of the innate homeothermic mechanism and the associated tissue perfusion force. It also portends a state of sub-normal hemodynamics (low BP and low cardiac output). During such stage oral infusion of potable water and or intravenous infusion of plasma expanders (saline) and or ice pack application is bound to fail and fatality ensues. There are no therapy as yet. One is wanted. Plasma expanders and exogenous cooling is alike beating around the bush. Therefore, in status SS, the aphorism 'treated extensively, the patient died', holds teasingly good. The treating clinician do not understand all this, he and his lot are cue less, stops running from pillar to post, reminds self about God, gives up (becomes a believer !). Hope is the tether. Metaphysics replaces bio-physics. It is nursing that delivers the most. An old experienced nurse (often) makes the difference.

One has to appreciate, that clinicians are mere technician of the biological bodies. Even an eminent clinic-surgeon of great standing is a mere fire fighter *par excellence,* and not a design engineer. Researchers have yet to arm them. This transaction is a mini step in such direction.

**Treatment**

Sun shade and standard/moderate gut loading (solid + fluid + ions + glucose) is the best anti-dote (prophylaxis\preventive). In clinics and hospitals ice pack are used. We are of the considered view that use of ice packs further complicates the faltering hemodynamics. In place of ice pack, cool (10-$20^0$C) 2%KCL+5% NaCl +5% CaOH mixed water should be used as spung dissolved into cold potable mineralized water. $O_3$ (ozone) therapy may also prove beneficial. Gut to periphery pathway route ~ is the sole systemic working route of ion injection available ~ needs to be utilized as SOS. Therefore, 2%KCL+5% NaCl +2% CaOH should be orally slow fed as the sole internal medicine. Preferably cow milk diluted or goat milk (1:4 v/v $H_2O$ & milk) be administered orally, for rapid induced mechanical quenching. 2%KCL +CaOH2 2% may also repeatedly be scrubbed gently on the gums as well for rapid sub-lingual up take (shortest route to heart & brain). Phosphate intra muscular injectibles (or sub-lingual) should also be considered. All these constitute Medical Meteorology [9-12].

**Meteorological History**

The term Sanatan means 'hydespas' (perennial) and the term Hindu (Lama-Mongol phone) means non-migrating\river valley\settled\unflinching.When we look up the History of India, we note that it has been enjoying a settlement type of well cultured life since very early times (whence barbarians roamed most of Europe & the Americas). From every conservative perspective it is clear that the Indian sub-continent has the oldest living culture (has been in continuum since 5000yrs before present). All other peer civilization and cultures seem to have vanished leaving back deserts. Anthropologically, human migration has happened towards the Indian sub-continent. Counter immigration tantamounting to full abdication has never been. The IMD data says that there has virtually been NO change in annual rainfall or temperature pattern over the Indian peninsula (only annual variability,

including 2008). In Fig.5. we have produced a graph pertaining to RF during the last 100 yrs [13]. Meanwhile, India has gone on to house 1.2 Billion stomachs and an enviable scientific heritage. This means, native Indians may have designed devices, mechanisms and methodologies to avert/overcome/ down regulate the effect of aridification mechanics, heat wave and sun stroke. In this segment we consider as to how the native has been coping with the heat wave and how we have been inspired i.e. few tangible (native/national) and intangible heritage aspects are discussed. Monsoon is the dominant met phenomena of the Indian sub-continent. And, it is marked by gross variability. HW is one of the signature products of such variability. The ancient mind had abdicated domain even in face of climate change (e.g., Nile; Euphrates, Indus, etc.). Monsoon is the Emperor of variability. It seems that it fails climate change (also). Thus, the same period mind native to monsoon climate had devised intelligent means and methods to overcome weather variability. Climate change in domain India thus stand discounted (Geological scale).

**Native Heritage Aspects**

**Soil & Moisture Matters**

Thus India (along with China) have 'permanent agriculture' & top soil conservation [14]. Indian farmers practice mono crop. They leave the stubble behind. The stubble provides micro level shed, traps moisture at night and translocates it back to the soil and finally thwart cake formation and soil denudation. Finally they allow the stubble to convert to fertile soil cum binder. It is a nocturnal-diurnal action. Thus she has a rich heritage in top soil conservation and SM retention. She also has unique practice of holding sweet water at surface she has a rich heritage in related engineering [15] as thin film water bodies [7].

**Home Stead Design**

Coastal India is Cyclone prone [16,17] and also land Tornados [18]. She therefore also has heritage in severe weather resistant homestead design & engineering [19]. However, (rural india's) heat wave resistant native-ethnic designs aspects have not been presented. Central and western Odisa (representative candidate of all India *loo* regions, a province of india, eastern shore board) we have noted that traditional dwellings have something called '*nia verandha'* (fire vestibule). Fig.-6 schematically explains the same. We may note that the passages of the *nia verandha* have bends at right angles. The desiccated, dry, hot breeze/wind fails to barge onto the doorway due to principles of cancellation that come into play in tune with Newton's 3$^{rd}$ law of motion. The passage width vary, which causes constriction and expansion of the air mass before it reaches the (closed) door. Because of such compression, a boundary layer of compressed air develops around the hutment. Village hutment door shutters are normally very ordinary, hence are not effective in shutting out the direct incident 'loo' (hot summer blast). Therefore, the *nia verandha* has a significant role. In c.18$^{th}$ A.D., constriction followed by expansion of manually blown air was used to be resorted to cool VIP room interiors and even surgery rooms. In process barocline compression inflicts heat into the system. In design *nia verandha* expansion inflicts cooling.

This apart, the Odisa village hutments (roofs) have *attoo* (inner ceiling). *Attoo*(s) are either horizontal or slanting. They are made of timber planks or a combination of bamboo and clay, wood and tile interlay, sand & lime plaster, etc. The walls of the hutments are thick ranging between 12-24 inches wide. Fig.7 schematically explains the concept of cooling in a rural conical thatched hutment (well to person). No.1 & 2 marks the surface level air and dry-desiccated/humid air. We know near the walls (solid state boundary/vector) a compressed boundary layer of air always exits. Outside breeze gets mildly compressed and climbs (cools also), is allowed into the room at wall height level through a constricted opening No.3, where it gains velocity and cools further. The impressed air expands within the room and cools further. As it gains moisture and warmth from the anthropogenic activity inside, mass gain occurs and it experiences gravity wave type phenomena = cool feeling. The non moisturized dry air rises along the inclined inner syncline of the roof from all sides. Structures that have *aatoo* may also have opening for access to the upper ceiling No.4. This further helps in air mass separation. At the apex a region of higher RH and heat is built

up No.6. On the outer, high contrast exists with surface area more than 1:1 (*aatoo* : outer). Top heating takes place here and enstrophy type phenomena gets located in and around the apex (roof-divider), No.7. This causes an ever present feel good factor inside the hutment. In western Orissa, the apex region of the tiled and the *Khapara ghara*, normally have small room as part of dual storied plan. The mono or the multi storey's roofed with *khaparas* (glyptic tiles). They are laid out in rows in an interlocking manner, often 2 or 4 layers thick. The layers have mini windows (due use of *khaparas* of variable cross section) between the upper & the lower tile spaces, thus heat escape (Fig\drawing not produced). Very versatile, unique and effective. The walls are thick, made of admixtured material (having varying thermal character). These act as roof atop *attoo* cum two way ventilator. It is entirely local industry & labour based also.

**Dress Heritage**

In the semi-arid and in the bone dry deserts, the Banzaras (forest wanderers), the Bedouins (desert wanderers) and the natives use layers of long staple make cotton thin textile, loosely slung over the head, neck and whole body. The numerous folds and the interlayer gaps act as conduction failure zones. The conduction route via the cotton fabric is enlarged many times in terms of length dimension while the depth dimension is marked by discontinuity (inter-folds). Moisture from the body is also trapped in these layers which results is an ever present macro area, mini convective current(s), in a reducing manner towards the outer. Convection cools. The inter-layer gaps keep on altering (squeeze & expanse) with even slight movements of the body. As a result, evolution of uniformity of pressure, moisture and thermodynamic conjugation constantly keeps on failing. Between the long and the short, the long staple cotton allows close and tight knit of the fabric (traps outgoing body moisture), while the loose drape attire assists micro area breeze, which assists cooling. Two contrasting environment domains are maintained in close proximity. The white mono-chrome textile (*al-khalla* ?) head drapery of the desert regions also efficiently reflects cum cancels the incident light, assists better vision inspite of the glowing Sun. The headband acts as a circular lock, upto which point internal RH is higher [20].

**Food Heritage**

Rice, Chuda, Mudhi do not pass hypovolemic stress test. Dal (lintel) fares better. The tropical (water inundated crops) staple food products have relatively low moisture retention property. They loose moisture swiftly and self semi-bake into hard, filamentous, crystalline cakes, that do not reform (well or to original) on regain of moisture/fluid balance. Gluttonous food like wheat, dates (*khajur*), major and minor millets (*jowar, bazra, maize, rasi*….) dry fruits (like almonds, apricots) resists rapid fluid loss, cake formation and crystallisation within the gut. Post drying they (comparatively) re-form very well on re-gain of moisture/fluid. When wheat is intermixed with minor millets or lintel, the exposed surface takes a (dense) glossy coat comprising of organic polymers. Evapotraspiration slows down further and cake formation mechanics is down regulated if not thwarted. No crystalline cake is formed incase of mixed lintel gruel (*tadkaa, khechedi*). Hence, a Banzara or a Bedouin is not as much thirsty nor need to carry a large ration of potable water. A coastal native even when with belly full *pakhala* (watered rice), on initial expouser to *loo* conditions experiences a feeling of insatiable thirst. This is initial signature of a rudimentary 'hypovolemia'. In spite of this, Sun-stroke is the single largest killer in semi-arid and in the bone dry deserts world wide. Onion, cucumber and all species of water melon also helps to retain fluid and maintain gut lumen. We raise the question apart from semi-arid region cultivated gluttonous staple food and wheat, what else helps the homeotherm in systemic self assistance to avert or down-regulate body fluid loss and in body temperature control ? We note it is intake of curd (lacto-bacillus) and citrus fruit (Vit.C with fiber) and ascorbates. Excessive intake of NaCl leads to opening up of the renal irrigation channel and consequent onset of insatiable thirst cycle. Reducing sugars (as in dates & canes)are better. Intake of multi-vitamins (admixed high Dalton proteins) are potentially risky and toxic during heat wave episodes. Nicotine is better – for it moistens the broncheols and the alveoiles via a direct and reflux mechanism. Caffeine not, for it dilates the vasculature, assists heightened hypotension askews hemodynamics further. Tea assists vascular constriction may be of limited help in therapy.

Interestingly, Cow, Camel, Goat and last of all Buffalo milk positions as potent anti-Sun stroke (prophylactic cum therapeutic) food agents. This is because of the biogenic elemental Calcium, which is a efficient, most important and largest biochemical component of the cerebrospinal fluid (CSF) and neuromuscular signal transducer. Very recently, researchers have shown that civilization may have benefited from extreme natural events such as meteorite strike [21].

## DISCUSSION

The administration can do precious enough by monitoring soil moisture loss over extensive region (earliest signature). India has a good data bank in agro-met domains. The data should be opened upto the public. The nation can then pre-calculate with near precision about likely period, and intensity of any impending 'Heat wave' conditions in relation to time and place, and its impact on flora and fauna. HW & humidity in particularly has been related to medical health hazard [22]. Youth (hormonal anabolic stage) endure HW well. They should be encouraged to go into the mid-day sun and come back with solutions in sciences. For humanity to march, medicines are a must. For treating sun stroke patients there is not even a single specific medicine nor even a protocol as pan global \ national \ regional standard operating procedure (SOP). This communication suggests a few for the first time and also indicates a backbone for any SOP.

Fall in soil moisture (ecological phenomena) causes heat wave (meteorological phenomena). Jointly, becauses sun stroke (physiological phenomena). Insurmountable. Pan globally the geographical swath is enlarging even extending into Rhine land to Danube Europe (well beyond), which were otherwise considered as 'not prone to heat wave'. Technology is assisting, longer life cycles, extended hours of anthropogenic activity mostly involving greater amounts of energy conservation. And, most of such energy being derived from isotopes ~ which inflict long scope alterations at environment surface & inter-faces (soil & atmosphere). Mother nature annually auto reverses heat wave & soil moisture. Isotope based energy production and its conservations requires longer scope for recovery – is the suggestive natural indication (by such time the next annual dry season cycle heralds). Sun stroke has thus far remained non-reversible, associated with hemodynamics failure cum gut collapse (original 1st time findings) apart hypothalamus impotence (thermal shock- pseudo anaphylaxis). It heralds fatality. Internal infusion of salts and electrolytes via the oral route is advocated. Glutonic food that do not crystallize on fluid loss and whereas regain slimy form on contact with liquids (hand made, pan baked Indian roti). Bristling brine loaded with salts (mono & divalent cations), ascorbates, alkaloids and flavinoids (*Toranee* as in Odisa) are indicated as the anti-dote cum preventives. Even rice ferments (fluids called handia) in earthen utensils as made by various tribes of India indicates excellent physiological stress bearing ability (evapo-transpiration body fluid loss) and venal-lymphatic stenosis (personal data). Lymphatic pathway failure (due draw back of lymph fluid) is also clearly indicated in SS (2017 summer data). Draw back of lymph fluid causes muscle failure (specially the involuntary) and griping intestinal collapse conditions due to crystalising food in the lumen (An reverse {un-natural} inter connect is established which cannot be restored clinically). Therefore, poly layered, loose fitting cotton fabric body wear & and head gear avert body fluid loss and thermal shocks. In its absence, Bristling brine (& or handia) substitutes effectively. Native home stead designs as to how they cool interiors and heat wave failing vestibules (physics of the mechanics) are discussed with drawings. Heat wave & sun strokes are here to stay. There is a need to fight back. The WHO may need to have a protocol in place supported by the UN.

## CONCLUSION

Heat wave & Sun stroke are natural phenomena. Pan globally, the geographical swath is alleged to be on the rise due global warming & climate change. Demographic demand of the geo-space is on the rise, exponentially so also are HW and SS related casualties (which is often in group numbers). HW is dependent on geographical location, geospatial architecture, latitudinal extent, geomorphology, orography and annual-seasonal meteorological

variations, etc. It is a ecological\environmental crisis. SS is a medical emergency. They are inter-connected. The Indian civilization has been in continuum since at least 5000yrs before present. And whereas, all other ancient civilization rose and have withered away (mostly to desertification mechanics). Rural india has a plethora of native practices vis-à-vis sun stroke and food based prophylaxis. She also has an enviable range vis-à-vis heat wave resistant home stead design and dress code. The mechanics of meteorological process & environmental response in the genesis of heat wave has been discussed in an illuminating manner for the first time. The pathophysiology od sun stroke and the food and drug based (clinical & home based) anti-dotes have been presented succinctly. All these scientifically can be evaluated. Multi-lateral initiative shall be more beneficial. Heritage practices are always local employment friendly. Very little is known even within India. India administration is also quite ill-informed about her native heritage. This long period, multi-disciplinary communication is ground breaking and opens new vistas of inquest for socio-cultural benefits & for the comfort-life style industry. It is nascent & is not exhaustive.

## ACKNOWLEDGMENTS

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**In Memoriam**

These presents are in the memory of Prof. Bijoy Kishore Misra (Co-Author ), who left us for his Heavenly abode during preparation of this paper.We had made oral presentation at the NATIONAL SEMINAR on Disaster Management : Towards Ecological & Human Security, Jamshedji Tata Centre for Disaster Management, Tata Institute of Social Sciences, Deonar, Mumbai, India. April 24-25, 2009. This work is dedicated to the rural communities in his memory.

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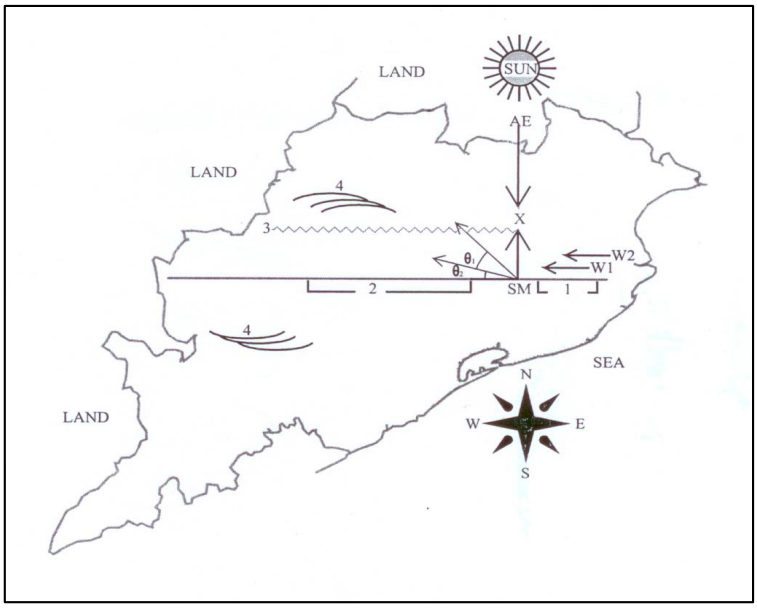


**Fig.1 : Schematically explains HW phenomena over domain Odisa (eastern sea board, India).**

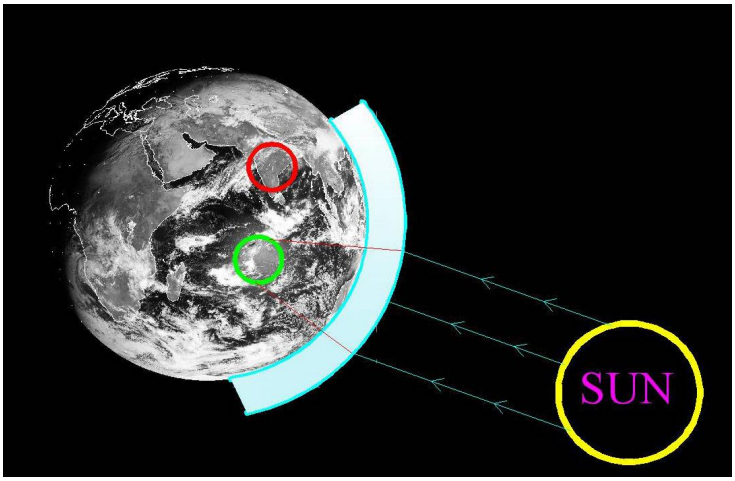


**Fig.2 : The incident Sun rays refract towards the central line as it enters the dense atmosphere (concentration effect). The photosphere is reflected on the earth's curvature in circular form covering nealy 250 of latitudinal & longitudinal space – which is equivalent of the length & breadth of the Indian sub-continent.**

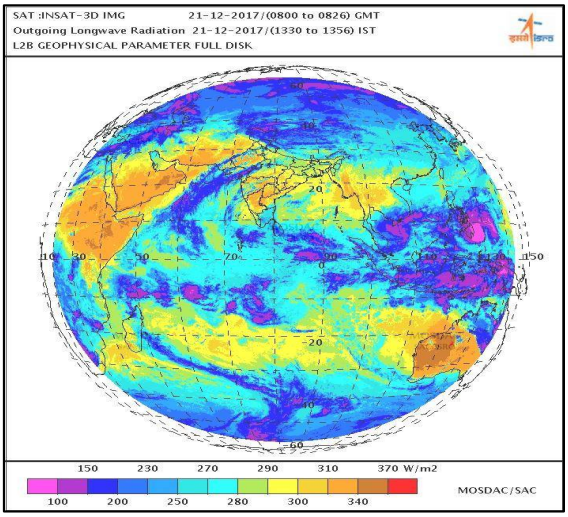


**Fig.3 : Shows the mean OLR at 300w/m2 band over sub-continent India, on dt. 21-12-2017 (08 to 08-26 UTC). Light colour represents the highest radiation. While deep regions indicate low OLR activity. OLR also indicates out-bound (from soil) moisture transmission, low altitude of the adiabatic line, cool surface breeze, high evapotranspiration, crop & seed drying.**

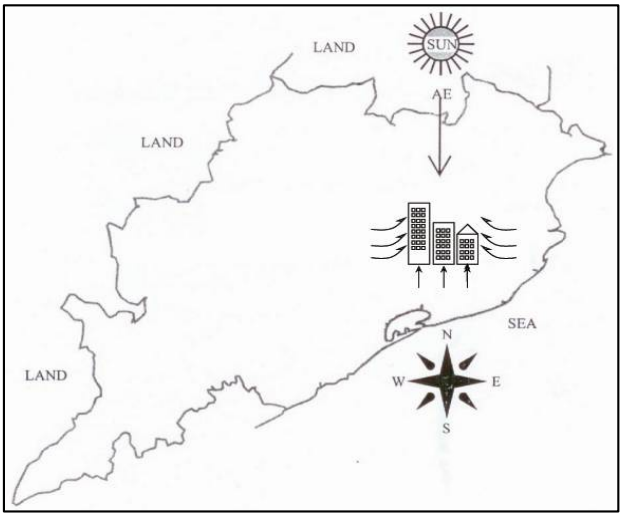


**Fig.4 :Schematically explains 'Urban Heat Island Phenomena' in and around Bhubaneswar-Cuttack twin city regions (20.2961° N, 85.8245° E & 20.4625° N, 85.8830° E, respectively), Odisa, eastern sea board of India.**

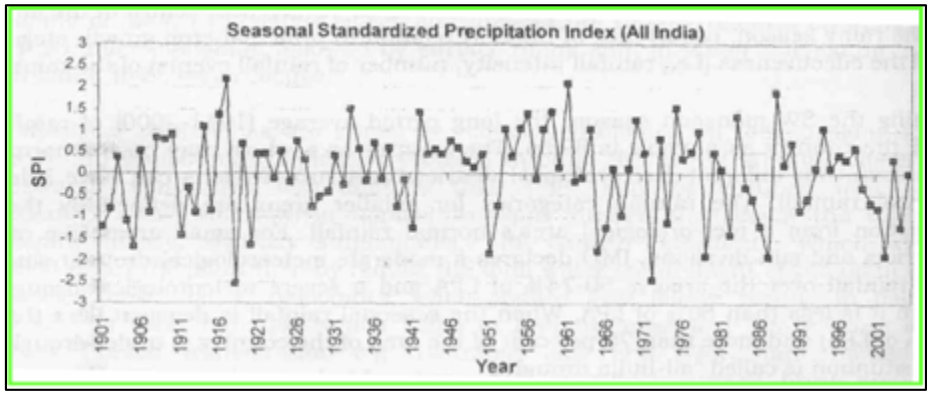


**Fig.5. Govt of India record showing the Standard Precipitation Indies for all India during the century period of 1901-2001 (industrial revolution & world war period shows consistency of RF). There is no noteworthy deviation over the century period.**

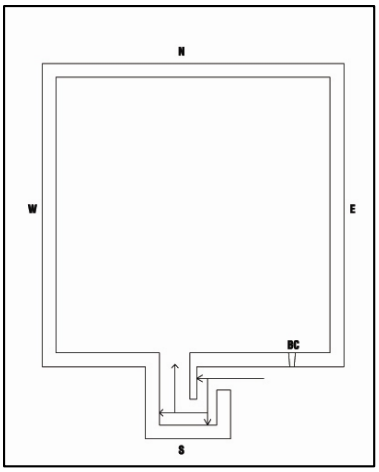


**Fig.6. Schematically explains 'Nia Varanda' heat wave baffle vestibule design and the fluid flow aspects.**

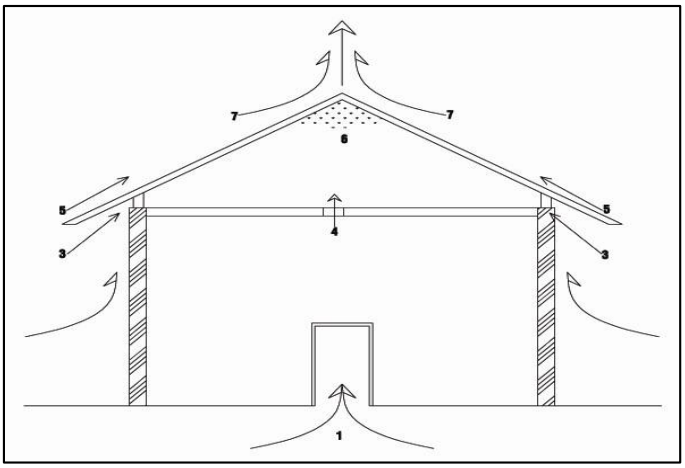

**Fig.7. Schematically explains 'rural thatched conical hutment' in rural India Arrows indicate the fluid flow during hot hours and the result of such mechanics translates in an down regulation of the lapse rate resulting in cooling governed also by Bernoulli's principle.**

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RESEARCH ARTICLE

# Isobolographic Toxic Study on P-glycoprotein Inhibitor –Verapamil with Colchicine in Acute and Chronic Administration in Mice

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## ABSTRACT

Toxicological study was performed in mice to compare the effect of interaction of colchicine and verapamil given orally alone and in combination by using isobolographic technique to compare LD50 of acute administration of drugs alone and in combination with that after chronic administration of low therapeutics doses for colchicine and verapamil (50µg\kg and 1.5mg\kg B.W) and high therapeutic doses of 70µg\kg and 3mg \kg B.W)respectively .The combined doses of both drugs of low and high dose were given to different allocated treated groups in mice dosed orally for 90 days ,then LD50 were performed in these animals.Chronicity index and isobolographic results were indicative that the sort of interaction of both drugs most possibly at P-glycoprotein cellular transporter system since verapamil was substrate inhibitor while colchicine was p-gp substrate .The results showed increase in accumulation of verapamil after alone and was less after combined dosing while colchicine showed no accumulation but less tissue concentration recorded in combined dosing .The results of hematological effect and clinical observed symptoms in animals of treated groups were in consistence with the results of chronicity index and isobolography since they were more toxicity symptoms appeared with hematological change manifested by decrease in red blood cells count ,packed cell volume and platelet count blood values as the course of experiment increase. These changes were more in combined dosing groups accordingly with the doses of both drugs causing the death of some of their animals during the course of experiment .

**Keywords:** p-glycoprotein , LD50, chronicity index , isobolography , colchicine ,verapamil.

## INTRODUCTION

P-glycoprotein (P-gp) belongs to the ATP-Binding Cassette (ABC) transporter family and it is expressed in various body tissues, such as liver, kidney, intestine, testes,and brain ( Colabufo et al .,2009 , Staud et al ., 2010).In the brain,

P-gp is localized at the luminal membrane of endothelial cells of blood capillaries where it actively modulates the permeation of xenobiotics ( Ohtsuki, and Terasaki ,2007) . P-gp overexpression has been observed in several tumours causing Multi Drug Resistance (MDR) due to the treatment with chemotherapeutics because of active transport of the drugs out of the cells (Pérez-Tomás, 2006) . Although compounds that interact with P-gp have various unrelated structures, they have recently been classified into four categories: substrates, inhibitors, modulators, and inducers .In studies performed in recent years, compounds transported by P-gp were considered as substrates, while compounds that compromise the function of the transporter were classified as inhibitors. Moreover, the term inhibitor was often used synonymously with modulator (Colabufo et al ., 2010).Colchicine is a medication most commonly used to treat gout,familial Mediterranean fever, pericarditis (National Prescribing Service 2010).Colchicine inhibits microtubule polymerization by binding to tubulin, one of the main constituents of microtubules. Availability of tubulin is essential to mitosis. The toxicity Long-term exposure to colchicine can lead to toxicity, particularly of the bone marrow, kidney, and nerves. Effects of long-term colchicine toxicity include agranulocytosis, thrombocytopenia (Van Echteld et al ., 2014).

Verapamil is a medication used for the treatment of high blood pressure, chest pain from not enough blood flow to the heart, and supraventricular tachycardia. It may also be used for the prevention of migraines and cluster headaches. Verapamil's mechanism in all cases is to block voltage-dependent calcium channels.in the smooth muscle lining blood vessels. By relaxing the tone of this smooth muscle, calcium channel blockers dilate the blood vessels. This has led to their use in treating high blood pressure and angina pectoris (The American Society of Health-System Pharmacists,2016).

## MATERIALS AND METHODS

**Materials:** In this experiment ,colchicine 1 mg and verapamil 40 mg were used,then prepared the concentration (50µg\ml and 70µg/ml for low and high dose for colchicine , 1.5 mg\ml and 3mg\ml for low and high dose for verapamil)according to body weight of the animals ,also using stomach tube for oral administration , EDTA tube for blood samples for RBC count , haemocytomter and microscope for counting.

**Animals :** Ninety three Adult Webster Albino mice obtained from Al-Razi center\ Baghdad , aged over 3 months and weighted 25-30 gram were used to perform different studies . They were fed standard pellet diet and drink tap water. The animals were left in special cages with normal conditions two week for adaptation in the animal house in veterinary medicine \university of Baghdad. Thirty animals for acute study while sixty three were used in chronic study. This study was performed under the rules of ethics for manegment labratory animals submitted by university of sided committee in the college of veterinary medicine.

**Parameters of study**

**Clinical and physiological study:**Animals behavior daily were measured and monitored through all experiment period for all groups and subgroups.

**Isobolographic analysis study and chronicity index.**

**Hematological test**: Red blood cell count(RBC), packed cell volume(PCV),platelet count

**Acute toxicity study (single lethal dose LD50)**

Thirty mice were divided into three groups : first group ( 10 mice) used to study acute toxicity of colchicine which administered at different oral lethal doses ranged between(50-70 ug/kg/ b.w) , the second group (10 mice ) were used to study acute toxicity of verapamil which administered at different oral lethal doses ranged between(1.5-3

mg\kg\b.w) while ,the third group (10 mice) were administered with different oral lethal doses of colchicine and verapamil in combination and their LD50 were calculated according to up & down method (Dixon, 1980).

### Dixon method

This test calls for dosing individual animals in sequence singly at 24-hour intervals, with the initial dose set at "the toxicologist's best estimate of the LD50." Following each death (or moribund state) the dose is lowered; following each survival, it is increased, according to a prespecified dose progression factor. If a death follows an initial direction of increasing doses, of 10-20 % or a survival follows an initial direction of decreasing dose with the same ratio, three additional animals are tested following the same dose adjustment pattern and then testing is ended. The LD50 is calculated using the following equation (Appendex-1): (Dixon , 1980).

Where: $LD50 = Xf + K d$

Xf = last dose administered , K = value from Table , D = difference between dose levels

### Chronic administration study

Sixty three adult male mice were divided into four groups; first group of 9 mice was act as control(C) and receive distilled water orally , the second group (18 mice ) administered combined therapeutic doses of colchicine and verapamil twice per a day and divided into two subgroups low therapeutic doses of colchicine and verapamil (CVL)and high therapeutic doses of colchicine and verapamil (CVH) . CVL ( 9 mice ) were given orally (50ug + 1.5mg )/ kg while CVH (9 mice ) were given orally ( 70ug + 3 mg) /kg respectively twice time a day for 3 months. The third group ( 18 mice ) received colchicine twice a day \two time weekly and divided equally into two subgroups ,Colchicine low dose(CL) and Colchicine high dose (CH) twice per a day orally with a minimum and maximum therapeutic dose 50 ug/kg and 70 ug /kg respectively for 3 months. The forth group (18 mice ) received verapamil twice per a day and divided equally into two subgroups ,Verapamil low dose( VL ) and Verapamil high dose(VH) given orally 1.5 mg /kg and 3 mg \ kg respectively represented the low and high therapeutic doses of verapamil .

### Isobolographic analysis study and chronicity index

This analysis usually used to determine the sort of interaction of two drugs, in which a line draw to join the LD50 of each of the two drugs studied in isobolograph and an intersection point determined by lines drawed vertically from the LD50 values of their interaction. The conclusion of drugs interaction were determined accordingly to the position of this intersection point to the two drugs joined LD50 line. If the point is to the right, it mean there is an antagonistic effect(□1), to the left, mean potentiation or synergistic effect(□1), while on the line(=1), it mean there is an additive effect between the two drugs.

#### Chronicity index

It is a measure of the possibility of a drug accumulation after its chronic administration based on comparison the ratio of LD50 values after acute and chronic exposure to the drugs according to this equation:

$$\text{Chronicity index} = \frac{\text{LD50 of Acute dose}}{\text{LD50 after 3 months dose with sublethal dose}}$$

If the result more than 1 , there is possibility of accumulation **( Klaassen, 1998).**

**Red blood cell count(RBC)**

RBC count is measured by using Hemocytometer .Count the number of cells in the small 5 squares in the center of hemocytometer , according to (Johnson, et al.2002).

**Packaged cell volume (PCV)**

Hematocrit is also sometimes called packed cell volume (PCV), can be determined by centrifuging heparinized blood in a capillary tube (also known as a microhematocrit tube) .Since a tube is used, this can be calculated by measuring the lengths of the layers by using the PCV ruler as percentage (John et al ,2009).

**Platelet count**

Platelets, are a component of blood whose function (along with the coagulation factors) is to stop bleeding by clumping and clotting blood vessel injuries.Platelet concentration is measured either manually using a hemocytometer , and then calculate in 5 square like as in RBC count or by placing blood in an automated platelet analyzer (Michelson,2013).

## RESULTS

**Experiment Part One**

**1. Acute toxicity study (single lethal dose LD50)**

Acute toxicity studies showed that LD50 for verapamil and colchicine according to Dixon method were 134.8& 18.9 mg/ kg .B.W respectively that reduced after their interaction to 67.5+ 8.8 mg/kg.B.W (Table 1 ).

**Isobolographic** analysis showed that the intersected lines of LD50 levels of verapamil and colchicine after their administration together was near the line that joined LD50 values of both drugs alone (Figure 1) estimated as the following equation**(Ronald ,2006 )** :

**Where:** **a\A + b\B** ⟶ **67.5\134.8+8.8\18.9=0.9**

a: acute or chronic combined administrated drug1

A: acute or chronic alone administrated drug 1

b : acute or chronic combined administrated drug2

B : acute or chronic alone administrated drug2

the result showed that there was a low synergistic toxic effect after the combination of colchicine and verapamil after their acute administration .

**Experiment Part Two**

**2.LD50 after chronic administration**

Chronic administratin of both drugs showed that LD50 for verapamil and colchicine after the end of chronic theraputic dosing for 90 days according to Dixon method were 85.2& 18.6 mg/ kg .B.W respectively that reduced after their interaction to 50.1+ 14.6 mg/kg.B.W (Table 2 ) Isobolographic analysis showed that the intersected lines of LD50 levels of verapamil and colchicine after their administration together was near the line that joined LD50 values of both drugs alone (Figure 2) estimated as the following equation (Ronald ,2006 ):

**a\A + b\B** ⟶ **50.1\85+14.6\18.6 =1.2**

the result of interaction between the combined verapamil and colchicine were antaoganist effect(above the line).

**Chronicity index (CI)**

Chronicity index was measured according to LD50 after acute and chronic administration of different treatment by using the following equation:

$$CI = \frac{\text{LD50 of acute administration of alone drugs or combined one}}{\text{LD50after chronic therapeutic administration of two drugs alone or combined one}}$$

The results of chronicity index for alone drugs and that after combination were :

CI (Verapamil alone) =134.8/85.2 = 1.58, CI (Colchicine alone) =18.9/18.6 = 1.01
CI (Verapamil combined) =67.5/50.1 = 1.3, CI (Colchicine combined) =8.8 /14.6 = 0.6

**Clinical observation and physiological study:** During 45 day of experiment ,the results of animal groups clinical observation of the colchicine and verapamil in the low and high doses showed slightly weakness, Corner aggregation and apathy as compared with the same groups at zero time ,whereas the combination group showed nervous behavior and aggression with some mortility as compared with the combination group at zero time.The results of clinical observation at 90 day showed that there were noticeable changes in the behavior and some abnormal clinical symptom like nervous behavior and aggrasivness with incoordination movement, Hair falling specially in the high dose combination of colchicine and verapamil groups as compared with zero time that showed normal behavior and normal clinical symptoms ,and there were some mortility in the low and high dose groups of combination and in the high dose of colchicine alone and high dose of verapamil alone .

**Hematological test**: **1. Red blood cell count (RBC)**:The results of (RBC) count between periods after 45 and 90 days of treatment with the tested drugs showed there were significant decrease in RBC of all groups at 90 days as compared with groups of 45 days . The results of (RBC) test between groups showed significant decrease($p \leq 0.05$) in (CVL and CVH) as compared with CL ,CH ,VL and control groups . (VH ) showed significant decrease ($p \leq 0.05$) in RBC count as compared with CL , CH and control groups . At 90 day of treatment with the tested drugs , the results between groups of (CVL and CVH) groups showed significant decrease ($p \leq 0.05$) in RBC count as compared with control group and other treated groups. Also, the (CL,CH ,VL and VH) showed significant decrease in RBC count as compared with control . The result of combined group CVL showed significant decrease($p \leq 0.05$) in RBC count as compared with that of CVH ,also the high dosing groups of colchicine and verapamil (CH ,VH) recorded significant decrease in RBC count as compared with low dosing groups(CL , VL ) (table 3 ).

**Packed cell volume (PCV)%:**The results of (PCV) test between periods after 45 and 90 days of treatment with the tested drugs showed there were significant decrease in levels in all groups in 90 days as compared with groups in 45 days . The results between groups in 45 day showed significant decrease($p \leq 0.05$) in PCV of group ( CVH) as compared with CL ,CH ,VL, VH ,CVL and control groups , and there was no significant decrease in (PCV) in the all remaining groups (CL ,CH,VL,VH ) as compared with control one . At 90 day of treatment with the tested drugs , the results between groups of ( CVH) group showed significant decrease ($p \square 0.05$) as compared with (CL , CH ,VL , VH ,CVL and control group. The high dosing groups CH and VH showed significant decline in PCV level when compared with low dosing groups CL and VL as shown in table ( 3).

**Platelet count\mm³:** The results of platelet test between periods after 45 and 90 days of treatment with the tested drugs showed there were significant decrease ($p \leq 0.05$) in platelet count in all groups in 90 days as compared with these groups in 45 days . The results of platelet test between groups at 45 day treatment showed the following significant ($p \leq 0.05$) reduction order of (VL, control , CL ,CH ,VH , CVL and CVH) . At 90 day of treatment,CVH group showed the highest significant decrease ($p \square 0.05$). in platelet count as compared with (CL , CH ,VL , VH ,CVL and control group .Also, the CVL showed significant decrease in platelet count as compared with CL ,CH ,VL ,VH

and control groups ,while the high dosing alone groups CH and VH showed significant decline in platelet count as compared with that of low dosing groups CL and VL,table ( 3).

## DISCUSSION

Drug-drug interactions (DDIs) have been attributed to the modulation of P-gp function following concomitant administration of drugs that fall into one or more categories. The co-administration of a P-glycoprotein inhibitor causes a much greater increase in drug concentration in brain than in plasma. A major problem that may confound attempts to use P-glycoprotein modulators in the clinical setting. Although the use of effective P-glycoprotein modulators (P-glycoprotein inhibitors), such as (verapamil) may improve the treatment of cancers, these P-glycoprotein modulators also inhibit P-glycoprotein function in normal cells, resulting in increased toxicity (Konig and Muller 2013). The results of isobolographic study and chronicity index of alone and combined administration possibly indicative the sort of interaction at different organ or system which attributed to the effect of p-gp on the absorption and transport of both drugs thus may affect their concentration levels in tissue or blood( Kim 2002) .Verapamil was known as substrate p-gp inhibitor (Gao ,et al.,2001) which mean inhibit the ATP transporter function of the drug outside cell, thus cause increase in its accumulation and resulted in increase of chronicity index (1.58) for LD50 of acute administration. Colchicine alone results of isobolographic chronicity index recorded of no change in LD50 ratio after acute and chronic administration which indicative no accumulation of drug in tissue possibly because colchicine not affect p-gp and so its kinetic was so efficient that complete drug was excreted after each day of administration of therapeutic colchicine doses .The results of administration of combined Verapamil and colchicine at low therapeutic doses were indicative there were competition between verapamil and colchicine for p-gp transporter system from the cell because they are both substrate to p-gp transporter that was inhibited by verapamil effect which make the concentration of verapamil less than that in alone administration that resulted in less chronicity index (1.3)but still there was accumulation effect of the drug in tissue. Chronicity index results of colchicine combined administration with verapamil showed there was less concentration of colchicine possibly in tissue after administration with p-gp inhibitor that possibly affect their level in tissue due to the competition on available p-gp sites which might effect the outside and inside transport to the cell since, p-gp is cell membrane transporter system act in bidirection pathway (Szewczyk et al., 2015).

These results were supported by the symptoms that observed through the period of experiment in chronic administration for both drugs alone and combined. Toxicity symptoms that developed through chronic administration of verapamil was indicative of accumulation since, there is many toxicity symptoms were developed throughout the experiment .While no clear symptoms appeared in colchicine group.After combined chronic administration there were noticeable side effect symptoms developed for both drugs at low and high doses that caused also death of 2 animals in verapamil alone high dose and 4 animals in combined verapamil and colchicine low and high doses which are inconsistency with the results of chronicity index that indicated accumulation specially for verapamil drug.

Verapamil may improve the treatment of cancers, these P-glycoprotein modulators also inhibit P-glycoprotein function in normal cells, resulting in increased toxicity. the toxic side effect of both drugs (verapamil and colchicine )on the haematological event as reported for colchicine caused aplastic anemia, low white blood cell counts that can last for several days; bone marrow suppression; thrombocytopenia; and ascending paralysis leading to potentially fatal respiratory failure, also the side effect of Verapamil specially in high dose is erythrocyte hemolysis(Konig and Muller ,2013).The mechanism of reduction of RBC count ,PCV and platelet possibly caused by using the both drug together in a combined dosesthat change their concentration in blood specially for colchicine as it recorded its low accumulation in tissue . Verapamil cause increase in concentration of colchicine in blood and cause hemolysis of RBC in predictable time and concentration dependent manner and increase the fragility of erythrocyte specially during

hypotonic osmotic stress and Ca+2 free condition . This is continuous with its inhibitory properties to the platelet aggregation by inhibit epinephrine –induced platelet aggregation ( Katz,1985 ).

## CONCLUSION

From the results obtained and discussion in this study, we can conclude that:

1. The p-glycoprotein(p-gp) inhibitor –verapamil was more affected on the results by its accumulation effect after the period of the experiment than colchicine drug showed in chronicity index.
2. The isobolographic study showed antaoganist effect due to verapamil competition effect on p-gp receptors site after combined administration of drugs(colchicine and verapamil) .
3. A significant decrease in red blood cell count, packed cell volume and platelet.

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**Table 1. Estimate of LD50 Dose for Verapamil, Colchicine and their interaction in Acute Toxicity Study**

| Treatment Groups | Initial dose mg/kg body weight | Final dose mg/kg body weight | Difference of doses | Result after 24 hours | LD50 Mg/kg |
|---|---|---|---|---|---|
| Verapamil | 80 | 120 | 20% | OOOXOXO* | 134.8 |
| Colchicine | 12 | 16 | 15% | OOXOXO | 18.9 |
| Verapamil + Colchicine | 95<br>13 | 75<br>10 | 20%<br>15% | XXOOXX** | 67.5<br>8.8 |

*O=live of animals **X=dead of animals

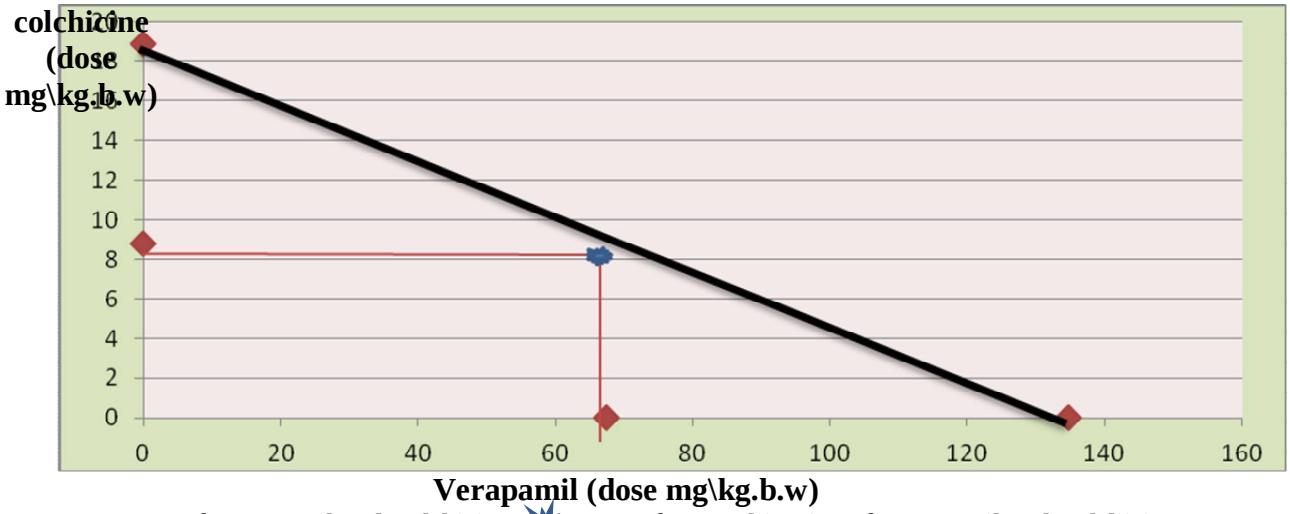


**Figure.1. Isobolographic analysis for Acute Toxicity interaction between Verapamil and Colchicine**

**Table.2.Estimate of LD50 Dose for Verapamil, Colchicine and their interaction in chronic administration**

| Treatment Groups | Initial dose mg/kg body weight | Final dose mg/kg body weight | Differences of doses | Result after 24 hours | LD50 Mg/kg |
|---|---|---|---|---|---|
| Verapamil | 140 | 100 | 20 % | XXXOXOX* | 85.2 |
| Colchicine | 18 | 18 | 15% | OXXOX | 18.6 |
| Verapamil<br>+<br>Colchicine | 85<br>+<br>18 | 76<br>+<br>16 | 20%<br>+<br>15% | XXOXOX** | 50.1<br>+<br>14.6 |

Verapamil (dose mg\kg.b.w)

*O=live of animals **X=dead of animals

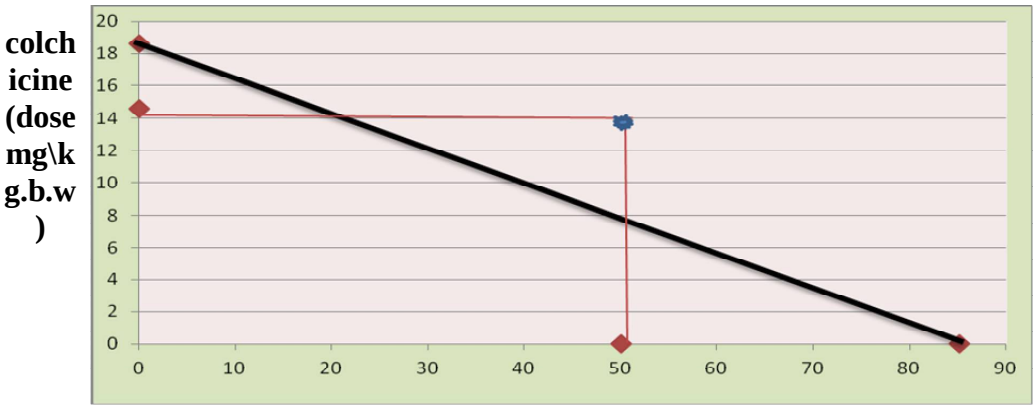


**Figure. 2 .Isobolographic analysis for chronic Toxicity interaction between Verapamil andd Colchicine**

**Table ( 4 ) Red blood cell (RBC) count ×10⁶cell/µl , Packed cell volume (PCV) count% , Platelet count \mm³ after chronic administration of colchicine and verapamil as alone and combined different doses in mice.**

| Period G | 45 days RBC | 90 days | 45days PCV | 90days | 45days Platelet | 90 days |
|---|---|---|---|---|---|---|
| Control | 6.19±0.12 A a | 6.19±0.15 A a | 39.57±0.68 A a | 39.4±0.92 A a | 420.71±2.34 B b | 421.2±3.36 A a |
| CL | 6.02±0.12 A a | 5.58±0.13 B b | 38.71±0.61 AB a | 35.4±0.93 B b | 416.71±2.33 C a | 390±6.16 C b |
| CH | 5.84±0.09 AB a | 4.98±0.04 C b | 37.57±0.64 AB a | 31.4±0.5 C b | 413.37±2.52 D a | 321.4±13.16 E b |
| VL | 5.63±0.09 BC a | 5.34±0.06 B a | 38±0.57 AB a | 37.2±0.96 B b | 423±1.75 A a | 394.8±4.05 B b |
| VH | 5.38±0.09 CD a | 4.91±0.04 C b | 37.71±0.42 AB a | 30.8±0.86 C b | 413.71±3.43 E a | 328±5.6 b D |
| CVL | 5.25±0.11 D a | 4.47±0.03 D b | 37±0.69 B a | 29.2±0.58 C b | 408.85±2.55 F a | 230±7.72 b F |
| CVH | 5.11±0.05 D a | 4.02±0.04 E a | 33±0.87 C a | 24±0.37 D b | 395.28±1.84 G a | 187.6±1.86 b G |
| | LSD=0.31 | | LSD=2.21 | | LSD=1.07 | |

**N=7 , CL: colchicine low dose (50µg\kg) , CH: colchicine high dose (70µg\kg), VL: verapamil low dose (1.5mg\kg) , VH: verapamil high dose (3mg\kg) ,CVL: colchicine - verapamil low dose (50µg\kg , 1.5mg\kg), CVH: colchicine - verapamil high dose (70µg\kg , 3mg\kg). capital letters donate differences between groups p≤0.05 ,small letters donate differences within groups p≤0.05.**

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RESEARCH ARTICLE

# Study of the Protective Role of the Aqueous Extract of the Black Caraway (*Nigella sativa*) on the Toxicity of Kidney Induced by the Pesticide 2,4-D in the Albino Mice

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## ABSTRACT

The aim of this paper was to investigate the protective effect of aqueous extraction for black caraway (*Nigella sativa*) on the toxicity of kidney induced by the pesticide 2,4-D in the albino rats. The study involved the use of (20) male mice of the Swiss white type (*Mus musculus*). The animals were divided into three groups, the first one was control group included the mice that dosage with physiological saline solution (0.09 NaCl), while the other group included the mice that treated with 150 mg/kg 2,4-D. the final group was consist of mice treated with 4 ml/kg of black caraway (*N. sativa*) aqueous extraction. then the animals were dissected after 30 days for the removal of kidneys, then it was fixed by fixative solutions after that a serial of histological sections preparations were conducted.. The results show that histological changes represented by sores in glomeruli and renal tubules with debris in some glomeruli and shrinking in the others and thicken in the basement membrane and some urinary tubules, in addition to expansion in Bowman's space beside hypertrophy and congestion in Blood vessels. The results also show that there were degenerative changes represented by many swelling in the cells lining of the renal tubules In addition to nuclear changes, especially the pyknotic nuclei and the emergence of edema between the interstitial tissue of renal tubules and the occurrence of vaculation among the renal tubules. It was also observed that there was Degeneration in renal tubule cells and expansion in the cavity. It was also observed that there was necrosis and vaculation between the proximal and distal tubular tubules. The results of the study show histological changes of the kidneys treated with 2,4-D pesticide with concentration of 150 mg / kg and that treated with the aqueous extract of *N. Sativa* seeds at a concentration of 4 ml / kg for 30 days. The use of aqueous extract helped to cure renal tissue and minimize changes and the return of tissue to a similar state original structure of the control group with congestion in some blood vessels

**Keywords:** kidneys, Bowman's capsule, 2,4-D pesticide, renal tubules, glomeruli

## INTRODUCTION

Herbicides are potentially toxic to humans, including many pesticides such as 2,4-D (2,4-dichlorophenoxyacetic acid) which considered as one of the most successful systemic pesticides used in modern agriculture. This pesticide was used with high concentrations to control many broad-leafed grass species in grasslands, gardens, agricultural fields and forests (1) . The chemical composition of 2,4-D is similar to that of indole acetic acid (IAA), which is a plant hormone acts as an inhibitor to plant growth. The toxic effects of 2,4-D pesticide have been documented where it was observed that exposure to this pesticide arises from a variety of damage to rodents, such as genetic (2) **,** hepatic (3) and neurological damage (4) . It has been shown that the herbicide 2,4-D increases fat oxidation in both animal and human cells in vitro and also causes cellular mutations that can lead to cancer (5) . It contains dioxin compounds and a group of known chemicals that can be hazardous to human health and the environment (6) . In rodents, the 2,4-D pesticide causes an increase in levels of progesterone and prolactin hormone, causing abnormalities in the rumen cycle (7) as well as hypoglycaemia effect (8) . High birth defect rates were observed in areas that use 2,4-D pesticide and other herbicides of the same category. This increase in defects was more pronounced among infants (9). Despite the apparent effect of this widespread pesticide, few attempts have been made to monitor the effect of the 2,4-D pesticide on the urinary system, which is highly sensible to many chemicals in the environment that can function either directly or indirectly on this sensitive organ In the body (10) including pesticides. From another hand black caraway (*Nigella sativa*) is considered as a medicinal herbal plant that belongs to the Ranunculaceae family and includes 15 species. Black caraway has many pharmaceutical benefits and uses including anti-inflammatory drugs and diabetes, as well as anti-bacterial activity(11) **,** anti-cough anti-anxiety (12) effects and Anti-asthma (13) . It is also used to treat high blood pressure due to its containment of quinone; therefore it is widely used as a medical treatment(11) . *N. sativa* has an important and vital role in combat viruses that cause cell hypertrophy and are used as a promoter of liver cells (14). Black caraway contains many types of phenolic compounds like thymol and thymoquinone, which enhances its ability to inhibit, kill or prevent the growth of microorganisms that infect humans and animals without causing any side-effects (15) . So the goal of this search is determination the preservative effect of the aqueous extract of the *Nigella sativa* on the poisoning of the Kidney induced with the 2,4-D pesticide in the albino mouse (*Mus musculus*).

## METHODOLOGY

The pesticide dose used was prepared depending on half lethal dose ($LD_{50}$) which is 370 mg/kg in mice according to(16) .Concentration of 150 mg/kg was selected and the weights of mice were ranged between 20-30 g. The treated doses were prepared according to the following relation:

$$\frac{1000}{W_m} = \frac{D}{x}$$

Where: $D$: The selected dose in (mg/kg), $x$: The dose according to mouse weight and $W_m$ : Weight of mouse in (g).

The mice were treated with the used dosage of 2,4-D pesticide once daily for one month for each treated concentration.

### Preparation of Black Caraway Aqueous Extract

The aqueous extractor of *Nigella sativa* was prepared according to (17) via weighing 50 g of black caraway seeds and gridding by electric grinder to obtain a black caraway powder. The powder then dissolved in 1L of distilled water in 37°C and leaves the blend overnight. Finally the mixture was filtered using What man filter paper 41 to remove the impurities. The filtrate was kept in sealed bottles until using.

**Laboratory Animals and Histological Study**

Twenty male mice of Swiss mice (*Mus musculus*) were obtained from animal house in Biology Department-College of Education for Pure Science/Diyala University. The average weights and ages were between 20-30 g and 8-10 weeks respectively. The experiments animals were divided randomly into three groups as follow: 1st group (CG) is control group and has 8 male mice and 2nd group (EG) is experimental group and has 16 male mice. The second group is sub divided into two groups of 8 male mice for each one. The animals in one of these sub-group (EG1) were treated with 150 mg/kg of 2,4-D pesticide while the mice in the other sub-group (EG2) were treated with 4 ml/kg of *N. sativa* aqueous solution. The animals were treated once daily for 30 days. In the end of the last day of dosage, all mice were anesthetized with chloroform, post-mortem to eradication of kidney, fixed with formalin for 24 h, washed with tap water and transferred to 70% alcoholic solution for keeping. Histological sections were prepared depending on (18) **.** The sections were (Dehydration) with an ascending chain of ethyl alcohol, then immersed in xylene for (clearing) and embedding in paraffin wax. The wax molds were cut using rotary microtome with 7µm thickness. The sections was stain used hematoxylin and eosin stain according to (19) and finally loaded with Canada balsam, tested and photographed using light microscope supplied by digital camera.

## RESULTS AND DISCUSSION

The urinary system in the vertebrae plays a sensitive role in the body, and the function of kidney -*which represents one organ of urinary system*-, is a part of many complex processes by which maintain the stability of the internal environment of the vertebrate bodies via acid-base balancing. The kidney has many functions including its role in removing many harmful and excess substances of body needed as well as the importance of sustaining animal and human life. This organ (kidney) filters the blood passing through the glomeruli into the body tissues in addition to its role in re-absorption of the electrolytes that needed by the body (20) . In particular, the kidney is exposed to the effects of toxins and drugs due to the large amounts of blood reached to its comparing to its size and to the concentration of the substance in its tubules as a result of the absorption of water in the tubules cells on the basis of the absorption of the substance itself or its secretion by these cells and about one third of the volume of blood pumped by the heart every time it goes to the kidney and about 10 % of the blood is filtered in the glomerulus and in the ) Bowman's capsule in the form of a leaky glomerular and approximately 1% is put out and the rest is re-absorbed from renal tubules (21) . Most of the damage effects due to expose to toxins may be caused by tubules or tissue cells, and poisoning may be ACUTE or chronic(22) . The results of this study show that the mice that treated with concentration of 150 mg/kg of 2,4-D for 30 days were suffering from obvious changes in their tissues, which were caused by inflammatory changes in the glomeruli and renal tubules with the debris of some glomeruli and shrinkage of others and the thickening and thickening of the basement membrane of the glomeruli and some urinary tubules and the expansion of Bowman's space and hypertrophy, and congestion of blood vessels as explained in Figure (1). This result is agreed with (23) . This congestion can be attributed to the inflammatory response of the body to the toxic substance, which is characterized by its beginning by increasing blood flow to the lesion area as indicated by a study of ( 24) . While the study of (25) showed that the accumulation of monocytes inflammatory cells around the Degeneration in the cells of the renal tubules after chronic exposure to a 0.01 mg/kg dose of phenyl rate was associated with enzymatic activity of these renal tubules cells. The current study also showed that degenerative changes were caused by the swelling of the lining cells of many of the tubules in addition to nuclear changes, especially the pyknotic nuclei and the occurrence of necrosis in the Parietal layer of glomeruli and the emergence vaculation of cells in the Parietal layer glomeruli as illustrated in Figure (2), while the apparent detachment of some of the epithelial cells of the tubules from the basal membrane and the emergence of edema between the interstitial tissue of the renal tubules and the occurrence of vaculation between the tubules and also observed cell debris in the lumen of the renal tubules and expansion of the cavity compared to the control group as in Figure (1). pyknotic nuclei of renal tubules and acute cellular Degeneration and vaculation were also observed in the lining of the epithelial cells of the renal tubules. The study also showed the occurrence of necrosis and vaculation between the

proximal and distal tubular endothelial cells And the appearance of edema between cells Figure (3). The results of this study are agreed with the results of (26) and (27) . The cause of this renal degeneration can be attributed to the disruption of the toxic process of the manufacture of certain enzymes cytochrome due to the removal of mitochondrial proteins as well as interference with the work of many other enzymes in cells lining the urinary tubules (28) . While the study of (29) indicated that renal degeneration was due to the accumulation of toxic substances in kidney tissues. The researchers(30) noted that renal tubular epithelial swelling means an increase in the number of endothelial cells within the tubules without proliferation outside the basal membrane. Previous studies have shown that herbicides, phenoxyacetic acid, accumulate at high levels in the liver and kidney through the organic acid transport system. This accumulation reduces oxygen consumption in the part nephrotic of the cortex, thus separating the renal mitochondrial association, which increases the potential for toxicity of the organ (31) . The changes in the renal cortex are due to the harmful effect of 2,4-D and the association of chlorophenoxy compounds to the kidneys .The results of the present study showed that the histological changes of the kidneys treated with pesticide with 2,4-D at a concentration of 150 mg/kg and treatment with the water extract of the seeds of the ring at a concentration of 4 ml/kg and for 30 days were characterized by the tissue sections of this group that the kidney tissue in general has returned to The structural structure is similar to the control group with almost congestion in some blood vessels and capillaries of the glomerulus appear more or less similar to control as explained in Figure (4). Figure (5) showed the presence of a small partial swelling in the lining of the renal tubular cells and the majority of the cells of the tubes showed normal tissue structure but most of them still show destructive brush boundaries. These results are consistent with previous studies where it was shown that the water extract of the *N. sativa* has a wide range of prevention as the presence of thymocinone acts as an anise to remove the oxygen roots. A helpful effect may be helpful in improving renal function(32) . Many of the active antioxidants have been isolated from some natural herbs, thus, it is assumed that the beneficial effect of black seeds and other herbs is most likely due to their protection from cellular damage caused by oxidative stress (33) . The *N. sativa* seed extract ingredients have special properties to remove free radicals (34) . Cells exposed to oxidative stress suffer from extensive cell damage due to the oxygen root that stimulates DNA cracking, lipid oxidation and extensive protein damage(35) . The current study assumes that the black bean provides protection to the cell membrane from the oxidative stress effect of pesticide 2-4-D. This is due to the antioxidant components contained in the black bean seed extract and the Thymoquinone effect, which has the potential to stimulate the hydrogenated glucose (6-phosphate), and eventually converted into active form of intracellular antioxidant glutathione (36).

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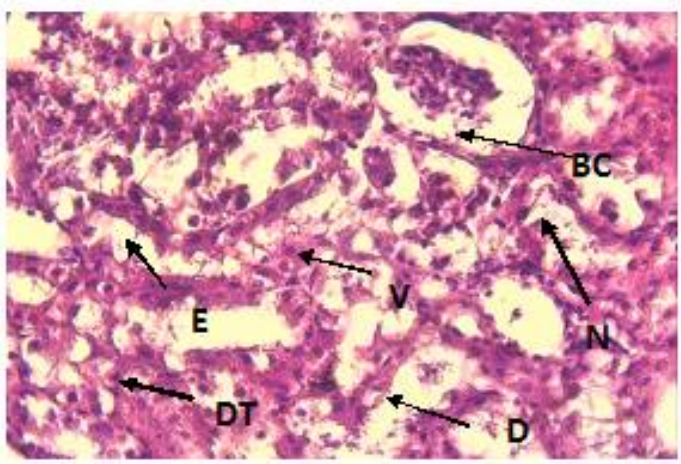


**Figure 1.Parasagittal section of the kidney of mice receiving 2,4-D for one month showing Degeneration and vaculation in the cells of the renal tubules . BC space of Bowman V vaculation E Edema D cell debris in the lumen of the renal tubules DT Degeneration in the cells of the renal tubules N necrosis H&E x 400).**

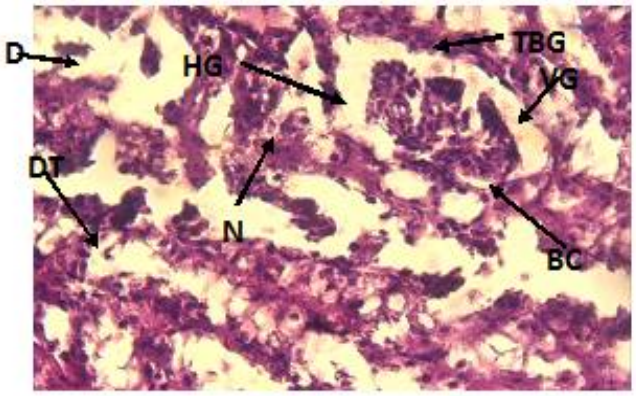


**Figure 2.Parasagittal section of the kidney of mice receiving 2,4-D for one month showing Shrinkage of some glomeruli And expansion the space of Bowman. BC space of Bowman V vaculation E Edema D cell debris in the lumen of the renal tubules DT Degeneration in the cells of the renal tubules N necrosis HG hypertrophy of the glomerular capillaries , TBG thickness of the basal membrane of the glomerulus H&E x 400).**

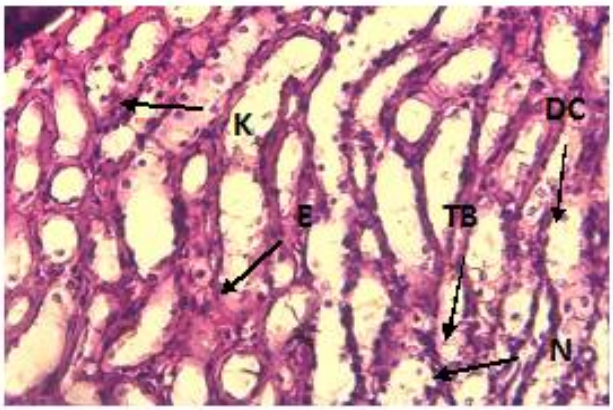


**Figure 3 Parasagittal section of the kidney of mice receiving 2,4-D for one month showing K pyknotic nuclei, E Edema N necrosis TB thickness of the basal membrane DC detachment of cells from the basement membrane H&E x 400).**

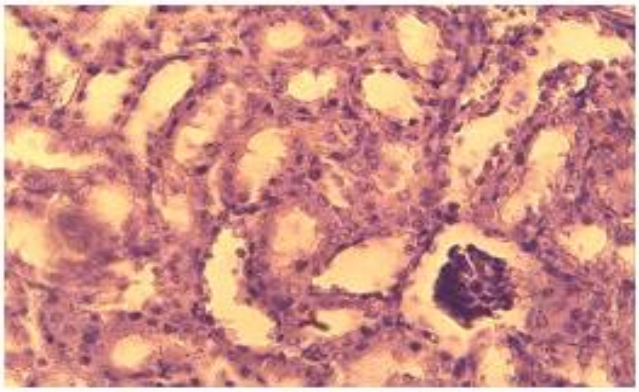

**Figure4.Parasagittal section of the cortex of kidney of mice receiving 2,4-D plus Aqueous extract of the black caraway (*Nigella sativa*) for one month showing that , the renal corpuscle and most of the renal tubules appeared more or less similar to the control (H&E x 400).**

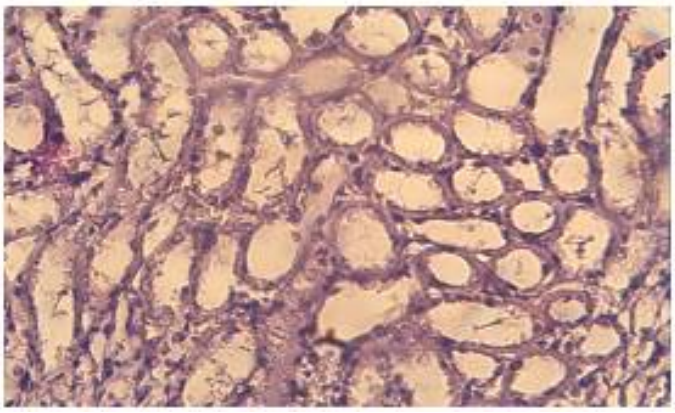

**Figure 5. Parasagittal section of the medulla of kidney of mice receiving 2,4-D plus Aqueous extract of the black caraway (*Nigella sativa*) for one month showed no clear lesion appearance except slightly cloudy swelling of laying cell of renal tubules H&E x 400).**

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RESEARCH ARTICLE

# Alchemic (*Spagyric*) Method of Solid Fixed Dose Therapy for Electro-Homeopathy: Natural Medicine

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## ABSTRACT

From c.1500 to c.1700 century in London, barbers and surgeons were in the same guild known as Company of Barber Surgeon (building still stands in London). According to Oxford University medical historian Margaret Pelling [1], barber-surgeons were effectively the General Practitioners of their day. Mainland Europe was consistently at war and the surgeon barber was more ever present, both in peace time and battle field location [2]. Regimental barbers also doubled up as emergency surgeon and thus grew the technique of surgery. It is relevant to state that the famous anatomist Rudolf Hoernale [3] has proven that the science of anatomy and invasive medicine was not known in Greece. That the Greek medicine experts use to visit India to learn. Indians (Ayoorveda specialists) never went anywhere to learn anything [4].Between c.1550-c.1700 the Fellowship of Surgeons merged with the Barbers' Company (Building existing at Monkwell Square, London) which in c.1800 was issued a Royal Charter [5] and got to be known as the *Royal College of Surgeons*, which till date holds the right to issue FRCS etc., practitioner's certifications (these till date be non-graduate {deemed} certificates as alike. 'Bar-at-Law'). The moot point in relation to caption (for this paragraph) is that at inception everything is simple, and retrospectively quite laughafable. Because, in the then Europe i.e., London-Berlin-Rome Paris axis (England specially), the barber surgeons were referred to as 'worshipful company of barbers' [6], now appalling indeed.Thus, there was no systematic observation (historical scale) based medicine what-so-ever as is connoted by the term Ayoorveda (plural health) and or any evidence based medicine as in connoted by the term Allopathy (antagonises symptoms).

**Key words;** Ayoorveda, Allopathy,Electro homeopathy.

## INTRODUCTION

Christian Friedrich Samuel Hahnemann (c. 1755 – 1843) was a educated German translator (humanist polyglot who became a physician i.e., Qualified MD. Leipzig Uni., c.1779) [6], He had invented, innovated and implemented a method of suspending plant and mineral or their whole crude extracts (tincture) in 100% ethyl alcohol and using them to treat the ailing on the fundamental principle of "like-cures-like" i.e., 'eliciting equal/identical symptoms' via internal medicine. This came to be known as 'Homeo-pathy". It arose out of homsapiens/ homeotherms and their pathology. Quite scientific indeed as compared to the historical Surgeon-barbers. Homeopathy swept Europe. While the principles of homeopathic medicine making was (and remains) quite scientific the practice of "like-cures-like" got loaded with frailties. Science in Europe was marching on brains. The maxim "dilution increases potency" failed homeopathy on the touch stone of experimental science. By & by Europe started looking for something better. 100% ethyl alcohol is acutely hepato-billary; neuro and nephro toxic.

As against this, Count Cesare Mattei (c.1809 - 1896) was an Italian who studied natural science, anatomy, physiology, pathology and then exclusively chemistry and botany ending up inventing a phyto-extraction process (spagyric) for therapeutic purposes **[7].** In short, he was a full blown scientist (who did focused work) in the domain of health care research in the then pan Europe. In his time he was famous also as a scientist. He gave to the world 'Electro-Homeopathy' using the spagyric method (Italian phone for alchemy). The term alchemy can be related serially as inorganic chemistry-organic chemistry. Spagyric denotes the phyto-chem part of organic chemistry for therapeutic purposes. With rise of scientific English diction\s the phone spagar fell into disuse and finally got discounted. The process of phyto-extraction for human service and family welfare has since swept the world.

Count Mattei could extract in water and then refine such extracts via separation/isolation and then re-inject the same in the inert base of the original plant mass used. Thus off due diligence in relation to then times he was convinced that he was using atomic levels of purification and hence optimistically termed it as 'electro'. Because isolation and inert base solid phase medicine could be used in incremental doses and exhibited electric fast response to doses.

## MATERIALS

Medicinal herbs are used as a material for medicinal preparation.

## METHODS

Electro homeopathy bulk drugs are made via the following steps.

(a) Distillation (b) Fermentation (c) Incineration*

*Calcinations being the old term of incineration

### Distillation

The herbs are washed and then boiled and distilled via a distiller. The herbs are kept carefully and aseptically aside. The distilled material contain essential & non-essential oils comprised of amino acids. These being in oil form are poor hydro-phylic and hence separation (or harvesting is convenient). Thus the therapeutic component of the fluid mass (volative & non-volatile; the medicinal flavinoids are made available) via such preliminary step. Electrically negative. Now they get to be as packed mass (a).

### Fermentation

The set aside wet herb masses (molasses) is then put into flask and fermented. It is a time scale process. Sugar & alcohol, etc are got post fermentation. Electrically negative yet assists ion-mechanics and help in indentifying NfKb gateways in various measures (signaling and cytokines) for major physiological pathways. The sugars that such post distillation molasses yield are non reducing in nature and whereas are efficient in ATPs cycles and energy conversion cycles in-vivo. The alcohol are phyto ethanol and are physiologically essential for gut functioning and are compatible with food and drugs. When they come in contact with toxic foreign bodies (bacteria\virus), they enter the pathogens and antagonize via precipitation of nucleic acids i.e., the DNA & RNA break down and do not support mutation. Availablity of drug moiety delivers the kill. All these post fermentation by-products are the very ones in which and around which the pre-extracted oils/amino acids & the flavinoids (drug moieties) were naturally bound/available (when the herb was standing free in dirty soil). These be the synergic support vessels; vehicles and efficacy up-regulators. Now they get to be as packed mass, sterile state (b).

### Incineration

The post fermentation (post extract of the fermented alcohol & sugar) molasses are then put into an oven and incinerated. Ash is got. The ash is mostly buff white **[8]** while it is as alike gray & black bodies as in carbon black. Burning of cellulose that has undergone the insult of extensive extraction (that is bereft of any residual compound) turns gray (1-20% gray, visible spectrum) and not black. In EH this is what happens. It is the counter-availing signature that distillation and fermentation and post ferment extraction cum washing has been proper. The burnt mass reduces by an order of 1/100 vol., of the original plant mass (pre-distillation), has a moisture content of <2% (dry & desiccated). It is full of micro-pores and are clatherin. Low energy. Electrically semi-conductor property. Trace elements (mostly) & minerals if any remain in the molasses and finally get shifted to the incenerated mass. Per gram having a surface area ranging between 7 - 11 $m^2$ . Particle size being mean dia 240 - 320 nm. It is therapeutically sterile. When the ash is added back a solid base is got for the natural extracts. Thus a solid drug is got from a green plant. This is ( c ).

All the three are added back and mixed to a homogenized state (slow & controlled). It makes the Electro-homeo product (d) = Bulk Drug.

Indeed it is 'electro' by pre-Ist World War technical terminology. In both the wars Italy lost. Count Mattie's genius got side lined. Not the efficacy nor the relevance.

### SOLID FIXED DOSE

Fixed dose from such Bulk Drug is numerically calculated as follows.

A + B + C = lower limit of the potency of the bulk drug (label expiry datum).
A x B x C = upper limit of the potency of the bulk drug (making datum).
√ (abc) = standard, replicable mean potency of the bulk drug (label potency of the solid fixed dose at prescription datum).

By physically dividing the potent Bulk Drug we arrive at fixed dose form (pills) which can be administered on the basis of pathological acuity; body mass index; age; sex; etc., clinical considerations.This is as good as present day allopathic medicine making. Quite unlike homeopathy (opposite of 'dilution increases potency'). In EH if the incinerated gray mass be added in more quantity then dilution happens = less potent. However, it is natural medicine; less to nil toxic; low potency and systemic. As a thumb rule tinctures are more toxic. EH is not tincture.

Researchers and specially altruistic administrations are looking at all school of medicines as part of Social Contract [10].

## DISCUSSION

The objective herein is to bring to light that EH is a genuine invention in the domain of health care of the industrial revolution period of Europe. It is sterling in many an aspects (subsequent communication). Also employment, investment & cottage industry friendly. European Commission has possibly per-chance missed focusing on it. EH is loaded with opportunity. Multi-lateral initiative is warranted.

## CONCLUSION

EH is a natural medicine. Making process makes it sterile and QC enabled.Potency calculation is possible. Fixed solid dose based therapeutics. Uses one base OR starting material. Not external additive driven Nor confounded. Synergic with other therapies; soft tissue targeting. Useful as Complementary & Supplementary therapy; neo natal, cancer, geriatric, etc. Very useful in chronic, drug resistant & metabolic diseases. EH is scientific; Original & independent. Employment friendly (hugely). Patient centric care focused. Gives lot of prescription room to the treating clinician.

## ACKNOWLEDGEMENTS

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7. http://homeoint.org/photo/m/matteic.htm
8. when a metal\wood (cellulose) surface having poly coats of multi colours (varying burning; peeling; foaming points) is exposed to hot flame various coats indicate various colours and at end leave a white buff streak as the burn edge line.
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RESEARCH ARTICLE

# Study of the Effect of Ribavirin on the Histological Structure of Kidneys in the Albino Mouse (*Mus musculus*)

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## ABSTRACT

This study was designed to know the effect of ribavirin drug on the histological composition of kidney in albino mouse. Thirty adult males with average weights and ages were between 22-29 g and 10-12 weeks respectively.. The experiments animals were divided randomly into three groups as follow: 1st group is the control group and has 10 male mice and 2nd group is experimental group and has 20 male mice. The second group is subdivided into two groups of 10 male mice for each one. The animals in one of this sub-group were treated with 100 mg/kg of Ribavirin while the mice in the other sub-group were treated with 200 mg/kg of Ribavirin .The treatment with Ribavirin showed pathological cases in tissues and cells of kidney including Inflammatory changes in the glomeruli and renal tubules necrosis, ,congestion in blood vessels edema.Also epithelial separation in kidney tubules The results also show that there were degenerative changes represented by any swelling in the cells lining of the renal tubules In addition to nuclear changes, especially the pyknotic nuclei.

**Key words:**kidneys, Bowman's capsule, Ribavirin, renal tubules, glomeruli

## INTRODUCTION

Ribavirin (1-~-D-ribofuranosyl-1, 2, 4triazole- 3-Carboxamide) is purine nucleoside analog widely used as an antiviral drug (1) Ribavirin is a drug that inhibits the inhibitors of an enzyme inosine monophosphate dehydrogenase (IMPd) activity which catalyzes the conversion of inosinate to xanthate (2). Ribavirin inhibit types of DNA and RNA for viruses such as influenza virus, herpes, measles, chickenpox, virus, hepatitis (3) It has been used to treat many diseases such as haemorrhagic fever and Crimean-congo haemorrhagic fever And hemorrhagic fever with kidney syndrome The drug Ribavirin is also active against viruses from the Arenaviridae family, Bunyaviridae, and Lassa fever It is also effective if used within 6 days of the onset of fever Other indicators include ribavirin

treatment for RSV, HCV, and Hantavirus infection. More than this, ribavirin is a very well-tolerated drug against RSV and viral hepatitis (4) The combination of this drug with interferon alpha is considered as a treatment for viral hepatitis (5) Due to its clinical effectiveness, many studies have been carried out to evaluate the toxicity effect. Despite the therapeutic benefits of this drug, its use is not free of side effects and toxicity. These effects have effects on the tissues and organs of the body, Ribavirin is reported to produce abnormalities in the cranial and limb joints of rats and hamsters (6). In addition to these anomalies, it also inhibits DNA synthesis and stimulates necrosis of dead cells in mice (7 ). The Ribavirin is toxic to bone marrow cells in the monkey which causes anemia dependent on the dose amount (8). aerosol therapy produces dyspnoea, Deterioration of the respiratory system, bacterial pneumonia and deterioration of lung function (9) However, its toxicity to the tissue structure of the kidney is unclear, So the goal of this search is Study of the effect of ribavirin on the tissue structure of kidneys in white mice in the albino mouse (Mus musculus).

## METHODOLOGY

Thirty male mice of Swiss mice (*Mus musculus*) were obtained from an animal house in Biology Department-College of Education for Pure Science/Diyala University. The average weights and ages were between 22-29 g and 10-12 weeks respectively. The experiments animals were divided randomly into three groups as follow: 1st group (CG) is the control group and has 10 male mice and 2nd group (EG) is experimental group and has 20 male mice. The second group is subdivided into two groups of 10 male mice for each one. The animals in one of this sub-group (EG1) were treated with 100 mg/kg of Ribavirin while the mice in the other sub-group (EG2) were treated with 200 mg/kg of Ribavirin. The animals were treated once daily for 30 days. At the end of the last day of dosage, all mice were anesthetized with chloroform, post-mortem to the eradication of kidney, fixed with formalin for 24 h, washed with tap water and transferred to 70% alcoholic solution for keeping. Histological sections were prepared depending on (10). The sections were (Dehydration) with an ascending chain of ethyl alcohol, then immersed in xylene for (clearing) and embedding in paraffin wax. The wax molds were cut using rotary microtome with 7µm thickness. The sections were stain used hematoxylin and eosin stain according to (11) and finally loaded with Canada balsam, tested and photographed using light microscope supplied by a digital camera.

## RESULTS AND DISCUSSION

The kidneys play an important role in the body by filtering the blood passing through glomeruli into the tissues of the body, as well as by re-absorbing the electrolytes needed by the body (12) The kidney is constantly affected by many toxins and drugs for the large amount of blood to its size on the one hand and the concentration of the material in the tubules as a result of water absorption or in the tubules cells as a result of absorption of the substance itself or highlighted by these cells (13). Damage caused by exposure to toxins may be concentrated in the tubules or tissues of the cells. Poisoning may be acute or chronic. (14). The results of this study show that the mice that treated with concentration of 100 mg/kg of ribavirin for 30 days were suffering from obvious changes in their tissues, which were caused by inflammatory changes in the glomeruli and renal tubules with hugeness of some glomeruli and the thickening of the basement membrane of the glomeruli and some urinary tubules and the smallness of Bowman's space and Bleeding and congestion and blood clots in the blood vessels as explained in Figure (1) This result is agreed with (15). This congestion can be attributed to the inflammatory response of the body to the toxic substance, which is characterized by its beginning by increasing blood flow to the lesion area as indicated by a study of (16). The current study also showed that degenerative changes were caused by the swelling of the lining cells of many of the tubules and the emergence of edema between the interstitial tissue of the renal tubules and the occurrence of vacuolation between the tubules and pyknotic nuclei of renal tubules and Degeneration was also observed in the lining of the epithelial cells of the renal tubules. The study showed the occurrence of necrosis and pyknotic in the proximal and distal tubular endothelial cells And the appearance of edema between cells Figure (1,2). The cause of this renal degeneration can be attributed to the disruption of the toxic process of the manufacture of certain enzymes

cytochrome due to the removal of mitochondrial proteins as well as interference with the work of many other enzymes in cells lining the urinary tubules (17). While the study of (18) indicated that renal degeneration was due to the accumulation of toxic substances in kidney tissues. The researchers (19) noted that renal tubular epithelial swelling means an increase in the number of endothelial cells within the tubules without proliferation outside the basal membrane.

The adverse effects of the drug increased with increased concentration of the drug, as mice that were injected with ribavirin at a concentration of 200 mg / kg resulted in more negative effects from the previous concentration Which was characterized by the occurrence of more inflammatory changes in the glomeruli and renal tubules and the emergence of vaculation and necrosis in the glomeruli and renal tubules and thicken and intensify the basement membrane of glomeruli and some urinary tubules and the expansion of Bowman's space and congestion and hemorrhage of blood vessels and the occurrence of necrosis in the area of the wall of glomeruli and the emergence of cells vaculation in the parietal area of the glomerular Figure (3). The present study showed that degenerative changes were caused by the swelling of the lining cells of many of the tubules, in addition to nuclear changes, especially nucleic pyknotic, as well as the apparent detachment of some of the epithelial epithelial cells of the tubules from their basal membranes, the emergence of edema between the interstitial tissue of the renal tubules and the occurrence of vaculation between the tubules Also, it was observed debris in the renal tubule cells and expansion of the cavity and the separation of some cells and collected in the cavity of the tubes Figure (4). pyknotic nuclei were also observed in the renal tubules and acute degeneration in the lining of the epithelial cells of the renal tubules, as well as the occurrence of necrosis and vaculation between the proximal and distal tubular and the appearance of between cells edema Figure (5). The emergence of the case of separation of the cells of the renal tubules from the basal membranes, especially the cells of the tubules descending and ascending in the area of the medulla of the kidney with the emergence of the case of necrosis of some cells may be due to the effect of ribavirin inhibitory prostacendin in the cells of endothelial lining of the arteries that feed the renal cells if the work of prostacendin stimulating the arteries On the expansion and that this inhibition leads to the reduction of these arteries, which reduces the supply of blood, which in turn affects the feeding of cells and thus the death of cellular This study came in line with the findings of the researcher (20) The results have shown the emergence of a case of inflammation in treated animals. Which may be due to the effect of ribavirin, which inhibits the action of COX enzyme in the manufacture of prostaglandins in the cells lining the blood vessels, which play a role in regulating the response of blood vessels and inflammation (21)The study showed the presence of edema in the spaces between the cells forming renal tubules. Which may be caused by the effect of the drug on cell membrane regulation and this result is consistent with (21) which indicated the effect of reflection on the kidney cells on the membrane regulation, which leads to the collection of fluid edema in the spaces between cells.

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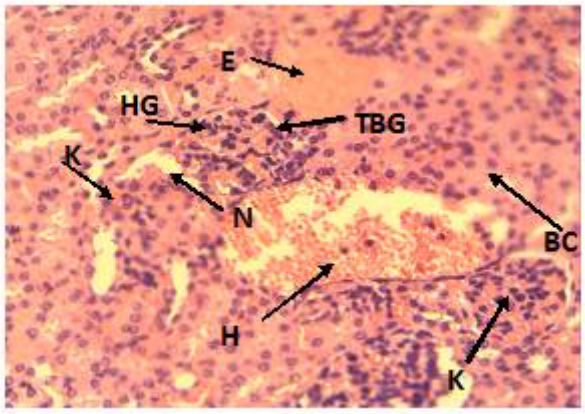


**Figure 1.Parasagittal section of the kidney of mice treated with 100 mg/kg of ribavirin for one month showing Degeneration and vaculation in the cells of the renal tubules . HG hypertrophy of the glomerular capillaries TBG thickness of the basal membrane of the glomerulus E Edema ,N necrosis .K pyknotic nuclei. H hemorrhage H&E x 400).**

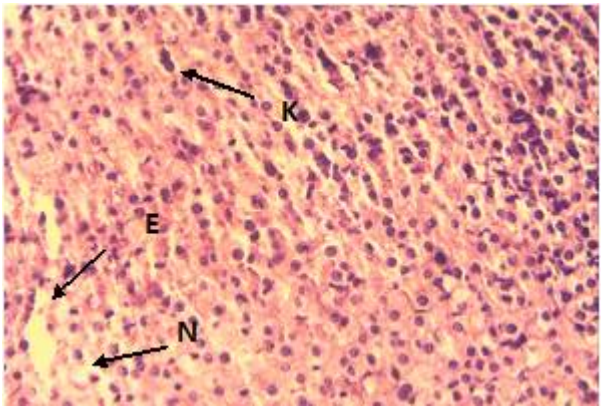


**Figure 2 Parasagittal section of the kidney of mice treated with 100 mg/kg of ribavirin for one month showing. TBG thickness of the basal membrane E Edema ,N necrosis .K pyknotic nuclei. H&E x 400).**

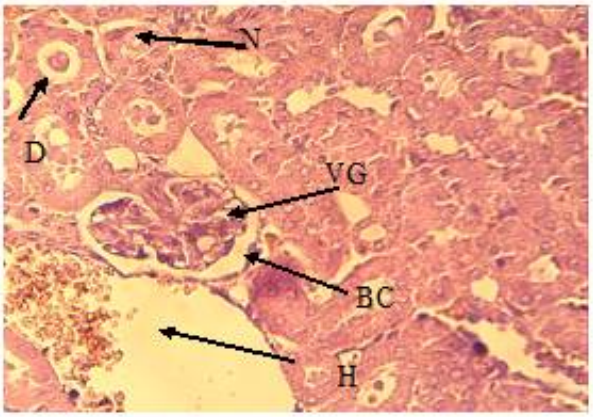


**Figure 3.Parasagittal section of the kidney of mice treated with 200 mg/kg of ribavirin for one month showing Degeneration and vaculation in the cells of the glomeruli and renal tubules . BC space of Bowman V vaculation of the glomerular capillaries cell debris in the lumen of the renal tubules,N necrosis .K pyknotic nuclei. H hemorrhage H&E x 400).**

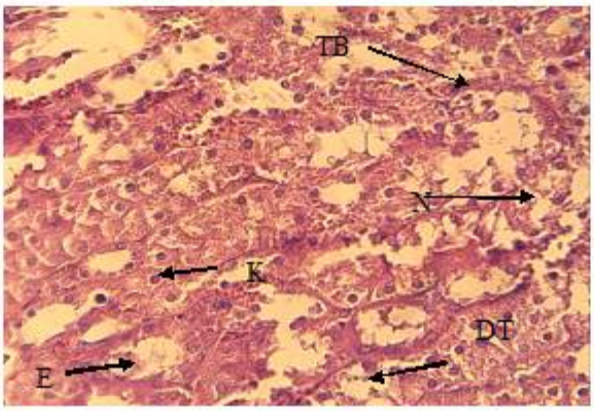


**Figure 4. Parasagittal section of the kidney of mice treated with 200 mg/kg of ribavirin for one month showing Degeneration and vaculation in the cells of the renal tubules . D cell debris in the lumen of the renal tubules TB thickness of the basal membrane E Edema ,N necrosis .K pyknotic nuclei. DT Degeneration in the cells of the renal tubules H&E x 400).**

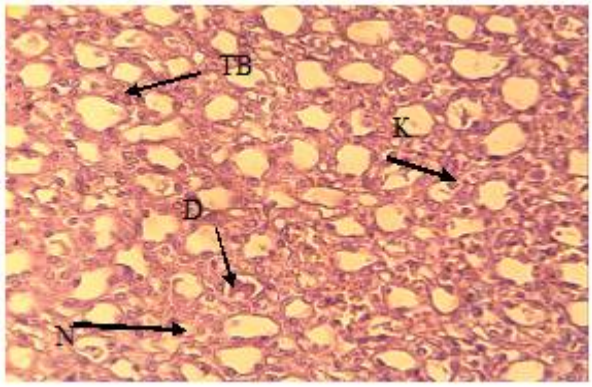


**Figure 5.Parasagittal section of the kidney of mice treated with 200 mg/kg of ribavirin for one month showing. D cell debris in the lumen of the renal tubules TB thickness of the basal membrane,N necrosis .K pyknotic nuclei H&E x 400).**

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RESEARCH ARTICLE

# Hazards Eruption of Taftan Volcano (Tephra, Lava Flow) and Its Risk Management, SE Iran

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## ABSTRACT

Taftan volcano is located at a height of about 4000 meters 45 kilometers north of Khash city in Sistan and Baluchestan province. This stratovolcano is one of the numerous volcanoes forming the volcanic arc of the Makran subduction zone, in which the Oman Sea oceanic crust leads to the north. Previous studies have shown that lava exits, ash erosion, and the production of pyroclastic streams in the past. Due to the low morphology of the volcano, the presence of spa springs and sulfate as well as the subduction process, there is a probability of the resurrection of the volcano at this time, in which case the eruption of the Stromboli, volcano to sub-pliny may occur.Taftan Stratovolcano is a semi-active volcano that can erupts again and be active, In the past, the volcano has been accompanied by activities such as lava flows, ash eruption and clouds eruption. Its possible eruption is estimated at 3 to 5 and is equivalent to Stromboli, volcano to sub-pliny types.Taftan is one of three active or semi-active volcanoes in Iran. The last volcano eruption is unknown, but sulfur gases are released from the crater, indicating that the volcano's mountain is not extinguished. After a terrible earthquake of Saravan, which was 7.5 magnitude, there were reports of Taftan volcano activity. This earthquake can affect the behavior of this volcano, along with its ability to activate it, can also reduce its current activity. According to the study, the ashes from the eruption will move toward the east of the volcano and will threaten a number of villages in the area, but the lava flows to the north and south and can reach the villages of Tamin, Sangan, and Tamandan.

**Key words:** Hazard zonation, Tephra, Lava, Taftan volcano, SE Iran

## INTRODUCTION

Taftan is a young and semi-active volcano in the age of Pliocene-Quaternary in Balochestan and 50 km from Khash. The altitude of this volcano is 4050 meters above sea level and about 2000 meters wide. This volcano is built on the Upper Cretaceous and the Eocene. The first Taftan eruption, including lava and pyroclastic rocks, with dacite and

riodacite from 20km west - northwest of the present peak (Ganser 1966). One of the interesting features in Taftan is the mineralogy and the inverted transformation of minerals in the quaternary andesites of this volcano. No gems rocks are observed in Taftan lava. Potassium rocks and subtropical tuffs cover a large part of the east and southwest Taftan volcano, which consists mainly of pumice. The first volcanic eruption is 20 kilometers northwest of the current peak, and then other centers in the eastern part of this point have been activated. The activities of these centers are explosive eruptions, resulting in ductile and agglomerate slices. The last blast of the Taftan explosive is two explosive phases. According to geologists, volcanic activity is associated with an earthquake, but no earthquake is indicative of volcanic activity.In the event of an explosion of supersonic type, they will be propagated on some marginal levels to the north, east and south of the volcano, and will threaten the villages of these areas. Together, the maps map out areas of vulnerable volcanic activity and can be used for management actions such as land use change, prediction and warning, preparedness for rescue and relief.

From a geological point of view, this volcano is located at the end of the structural zone of the Flysch Nehbandan-Khash and the northern zone of Makran. So far volcanic stratigraphic studies have not been carried out on this volcano. In general, Taftan stratigraphy can be divided into three parts, older rocks of activity, activity rocks and rocks after Taftan volcanic activity.Effective communication is essential in managing ballistic hazard and risk (Barclay et al. 2008; Leonard et al. 2014). Science needs to be communicated to decision-makers, stakeholders, and the public and understood and absorbed by them so they can make informed decisions. Similarly, the public, stakeholders, and decision-makers should communicate to scientists what type of information they need to make decisions relevant to their situations.There is a steep slope on the southern slope of the volcano, partly ruined by the explosion and subsequent erosion. From the steep slopes, yellow and white fumaroles rise above the summit, with a number of fumaroles coming out from the highest peaks of the mountain, a white cloud that is visible at a distance of 100 kilometers and shows the magnificence of an active volcano. The Taftan lava covers an area of 1,300 square kilometers.Several studies have been done on the volcano Taftan volcano usually aimed at better understanding and therefore have not paid the assessment and hazard zonation. The most important of these studies were by Ganser (1953-1962, 1966), Zareians and colleagues (1968), Stucklin (1968), Ganser (1971), Giroux and Conrad (1976), Moeein Vaziri and Amin Sobhani (1978), Darwishzadeh (1360ᵖ), Ghazaban (2004), Gansar 2008), Beyabangard (1979) and Moradian (2008), and Beyabangard and Meradian (2009).Considering the desert evidence and calculating the correlation coefficient of different volcanic phases by Mohammad 2008, Taftan has different eruptions from Stromboli to volcano and in some cases pliny. In total, although humankind did not consider its eruption during the historical period, However, due to the intense activity of fumaroles, sulfates, hot springs and the last eruption and severe erosion of the volcano, there is still a continuation of the process of induction of the Omani oceanic plate, which is the cause of the formation of the volcano, It can be seen that there is a possibility of its re-eruption, and in this regard it is necessary to identify and assess the threats posed by it and the threatened areas.

It is estimated that in case of its new activity, the eruption will be volcano to sub-pliny with intensity of 3 to 5 in the magnitude VEI scale. Considering the type of previous eruptions and the petrological profile of this volcano, there is the possibility of activity of lava flow, pyroclastic and ash (tephra) in this volcano, which requires zoning for them. In addition, it is worth noting that due to the low rainfall of the area and the low snowfall at the level of the cone and the lack of lake crater, the likelihood of Lahara being damaged in the small part is low.

## Area of Study

The Taftan volcanic mountain is located in the southeast of Zahedan and northwest of Khash, at an altitude of 4,110 meters above sea level. Taftan is part of a volcanic zone extending to northern Pakistan and belonging to the Mount Sultan's volcano, which is located in Pakistan and seems to be dormant volcano. Taftan is formed inside the Flysch zone of the east of Iran.

The Taftan volcanic cone consists mainly of pyroclastic and lava flows, its activity has continued beyond the water and within the Pliocene to Pleistocene range. Taftan currently has sulfate and fumarole activity, which shows itself as a volcanic peak with the evaporation of steam, sulfur and sulfuric acid. In the Taftan area, there are also some hot springs, The most important hot springs are: Band Ghalo , Teng Bolboli , Jan Panah , Dareh Gol and etc. Plio-quaternary stratovolcanoes of Iran are considered as potential volcanic threats in Iran. Hence, it is helpful to evaluate the probability of their reactivation, their kind of threat as well as to determine locations at threat.

## Hazard Zoning

### A.Tephra hazard (Volcanic ash)

As noted above, there is a risk of the production and distribution of ash due to the possible eruption of Taftan. The distribution of ash in the area is mainly influenced by atmospheric conditions of the region. In this regard, the velocity, direction and puff of 5 different geopotentials were obtained from the base of the NCER/NCAR Center. Using the VORIS software, the area of distribution of ash in the region was obtained. Based on this figure. Due to the dominance of the western winds in the region, the ash moves westward to the east, and its plume forms in the east of the volcano.Fortunately, there are no towns or villages in the area of high ash deposits, but the village of Sangan and other eastern villages of the slopes are within a thickness of less than 10 cm.

#### B.Lava flow hazard

Probability of lava flow due to eruption of the possibility of Taftan volcano, it is necessary to identify areas suitable for the propagation of lava around the volcano. It was assumed that the lava would erupts from the main crater of Taftan volcano and it flows through the suitable slopes.Although this location may be the most probable location of the lava exit, it may not be in practice.Maximum distance covered for each direction is about 5000 meters, the lava flow rate was used to frustrate the natural dams used for each flow path 3-meter and the probability of lava flow from a various of 5000 directions was considered.In the figure number 5 is shown the possible range of lava deposits in and around Taftan volcano. According to it, villages in the Kharestan district including Tamin in the northeast of Taftan, Sangan in southern Taftan, Tamendan and Goosheh in the west of Taftan, are at high risk lava flow. A possible north-eastern flow will stop in the north-east plains but the flows of southern and southeast after passing the villages of Tamendan and Goosheh standstill in the southern (north Khash) plains.

#### C. pyroclastic flow hazard

Pyroclastic flows are high density mixtures of hot, dry rock fragments and hot gases that move away from the vent that erupted them at high speeds.Pyroclastic flows vary considerably in size and speed, but even relatively small flows that move <5 km from a volcano can destroy buildings, forests, and farmland. On the margins of pyroclastic flows, death and serious injury to people and animals may result from burns and inhalation of hot ash and gases.Considering the history of Taftan and its eventual eruptions, there is a risk of developing pyroclastic flows. The probable flows are shown in below prepared map which is almost covering most sides of Taftan volcano and nearby towns and villages.

These flows can be disseminated at levels in the north, east and south of the volcano and threatens the villages of southern Sangan (east of Taftan) and Kharestan (north of Taftan) district. Pyroclastic flows generally follow valleys or other low-lying areas and, depending on the volume of rock debris carried by the flow, they can deposit layers of loose rock fragments to depths ranging from less than one meter to more than 200 m.

**Risk Management**

Effective risk management is built on communication, hazard education and engagement with the at-risk communities (Johnston et al. 1999, 2000; Paton et al. 2001; Twigg 2002; Gregg et al. 2004; Leonard et al. 2008; Dohaney et al. 2015). Appropriate risk management actions by stakeholders, emergency managers and the public require an adequate perception of the risk and the correct actions to take in a crisis, with perception dependent on the hazard information received and exposure to impacts (Johnston et al. 1999; Leonard et al. 2014).Therefore, it is necessary to install seismic devices around the Taftan volcano in order to see if the new seismic activity is observed by matching other phenomena such as temperature and composition of hot springs, temperature and combustion gases from the volcano crater, changing the elevation and slope of the probability of a volcanic eruption.

Volcanic Hazards fall under Geological and Geomorphological Hazards. Although volcanic eruptions are more predictable than earthquakes, there is very little if anything that man can do to prevent or alter the hazardous events happening. The only events people can occasionally have control over are the diversion of lava flows, lahars and lake gas. Ashfall, pyroclastic flows, fountaining, lava eruptions and lahars cannot be prevented from happening. It is how the risk is decreased to people that is the main strategy of volcanic hazard management.The intensity of volcanic phenomena decreases with the distance from the eruptive center (crater or fissure). Topographic or meteorological factors may modify the progression of the phenomenon, such as the diversion of flows by the morphology.

Measures that must be taken in residential areas near the volcano. The main Strategy for preserving lives in the event of an eruption is Public Information.

1. Public Preparation: making sure the public are prepared and know what to do before, during and after and eruption.
2. Establish evacuation centers, routes and logistics
3. Put out information on TV, radio or if need be personal contact
4. Drills and Sirens
5. Clear up teams
6. Co-ordination of emergency Services
7. Exclusion Zones
8. Hazard and risk maps and land use planning.

## CONCLUSION

Taftan Stratovolcano is a semi-active volcano that can erupts again and be active, In the past, the volcano has been accompanied by activities such as lava flows, ash eruption and clouds eruption. Its possible eruption is estimated at 3 to 5 and is equivalent to Stromboli, volcano to sub-pliny types.Therefore, it is necessary to install seismic devices around the Taftan volcano in order to see if the new seismic activity is observed by matching other phenomena such as temperature and composition of hot springs, temperature and combustion gases from the volcano crater, changing the elevation and slope of the probability of a volcanic eruption.

Considering the type of previous eruptions and the petrological profile of this volcano, there is the possibility of activity of lava flow, pyroclastic and ash (tephra) in this volcano, which requires zoning for them. In addition, it is worth noting that due to the low rainfall of the area and the low snowfall at the level of the cone and the lack of lake crater, the likelihood of Lahara being damaged in the small part is low. Lava and pyroclastic streams can threaten some villages around the cone. Ashes moved to east and some of eastern villages of volcano will be threaten. But the lava flows to the north and south and they can reach the villages of Tamin, Sangan, goosheh and Tamendan. The types of eruptions and hazard zonation maps presented in this study indicate the areas at risk, which can be timely evacuated residents in these areas.

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**Taftan is formed inside the Flysch zone of the east of Iran.**

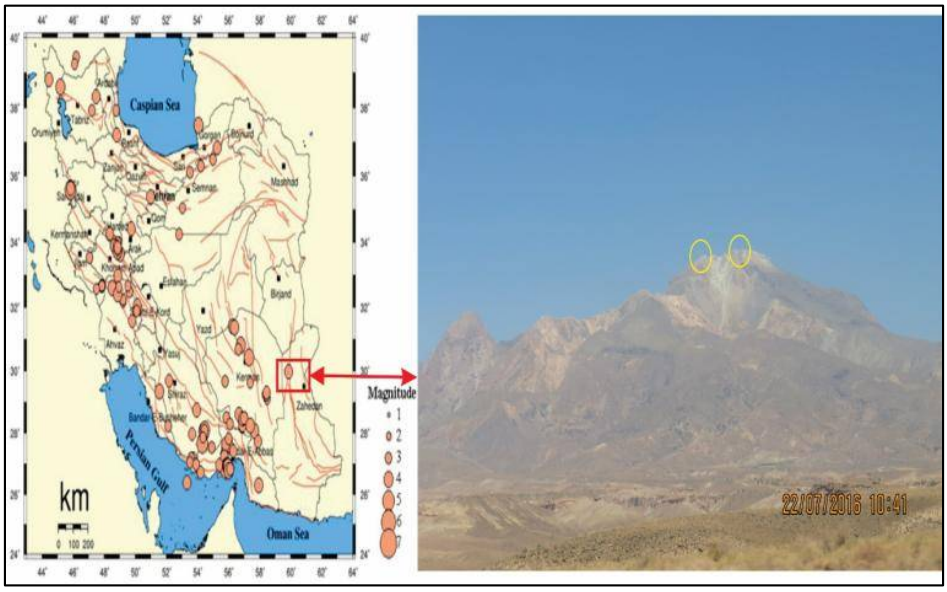


**Fig1. A view of the two cones of Nar Kooh and the mother Kooh of Taftan volcano.**

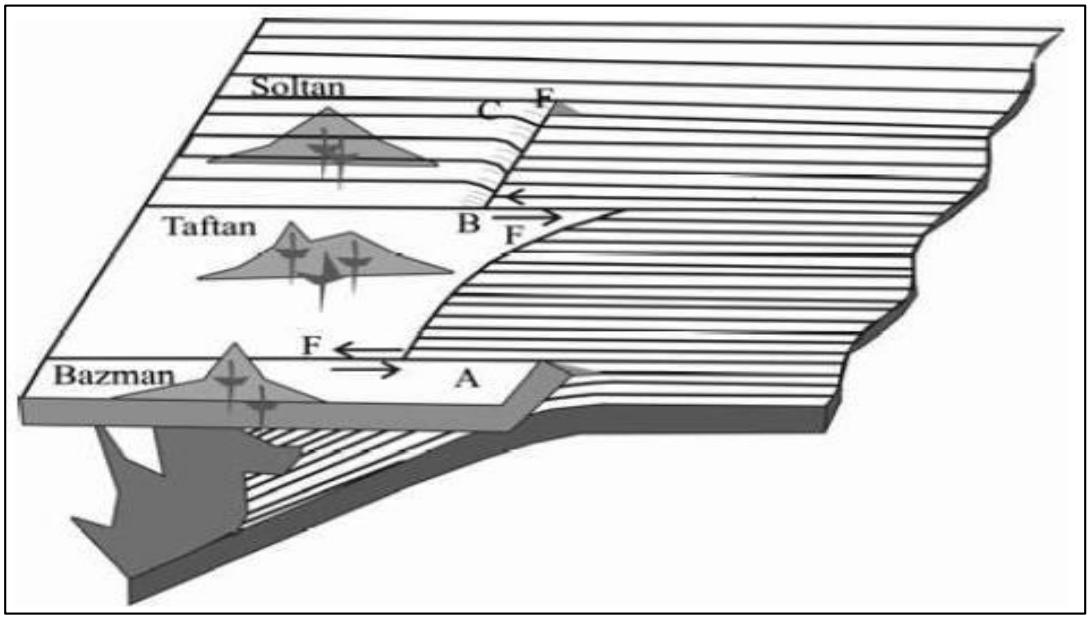


**Fig.2.Schematic image of the Oman crater depletion zone under the continental crust of Iran**

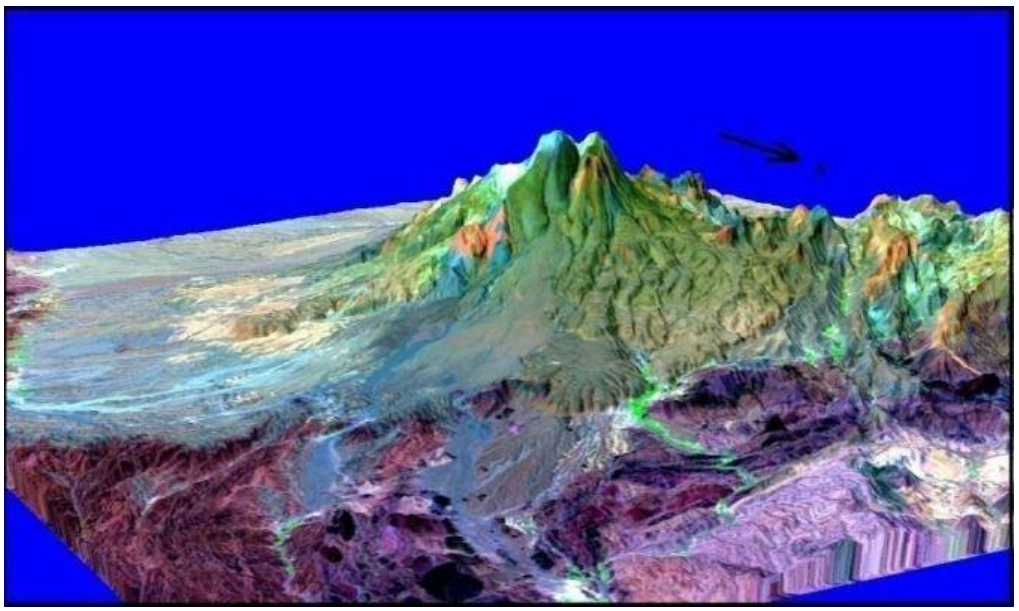

**Fig.3.Topographic image of Taftan volcano (DEM = 30 m + ETM$^{+}$ = 7-4-1 Bands)**

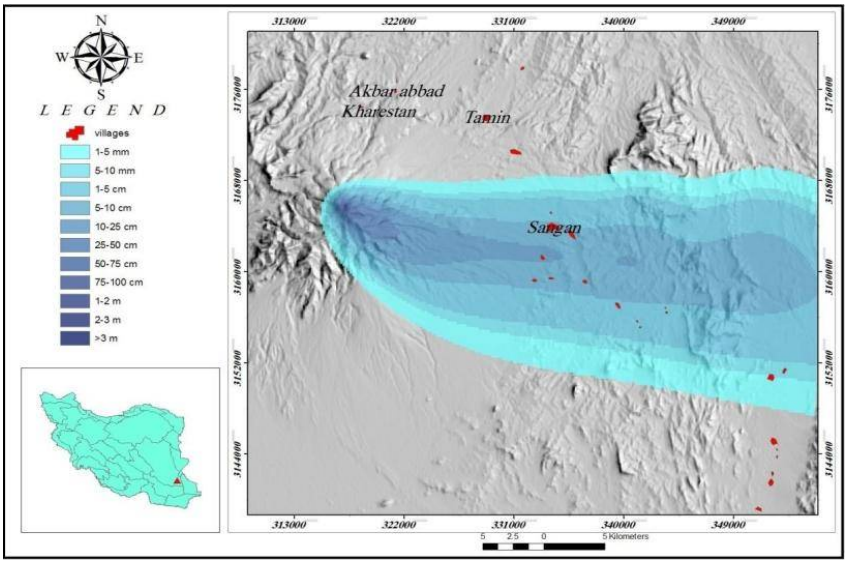


**Fig.4. Map of hazard ash zonation in the possible eruption of Taftan volcanoes and villages under threat of this phenomenon.**

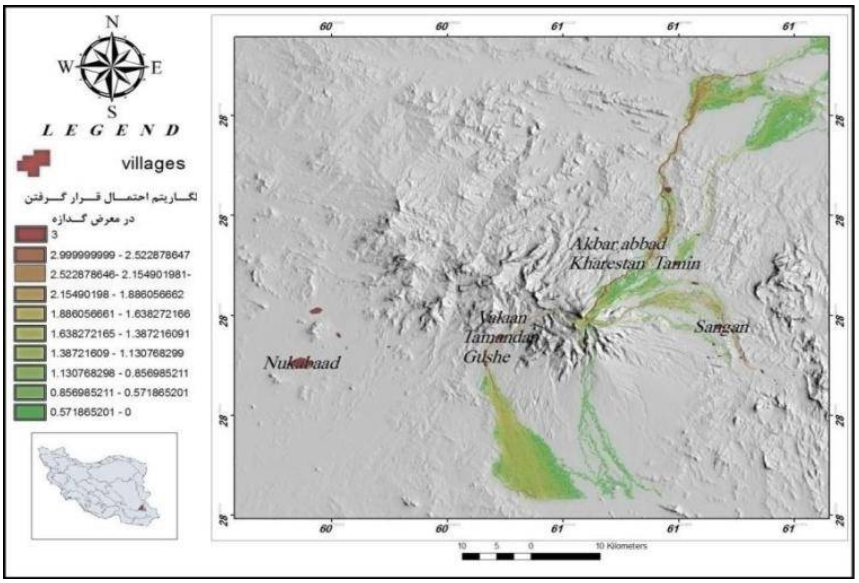


**Fig.5. Map of lava flows zoning in the possible eruption of Taftan volcanoes and villages under the threat of this phenomenon**

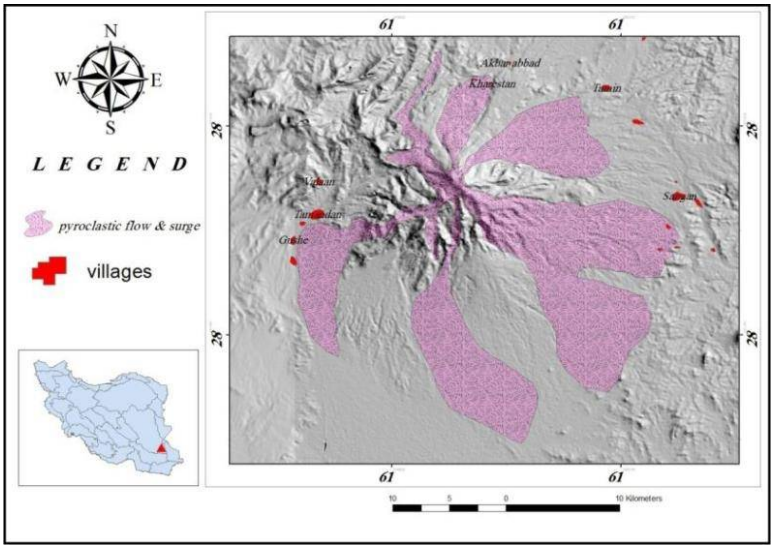


**Fig.6.Pyroclastic zonation map in the possible eruption of Taftan volcanoes and vulnerable villages**

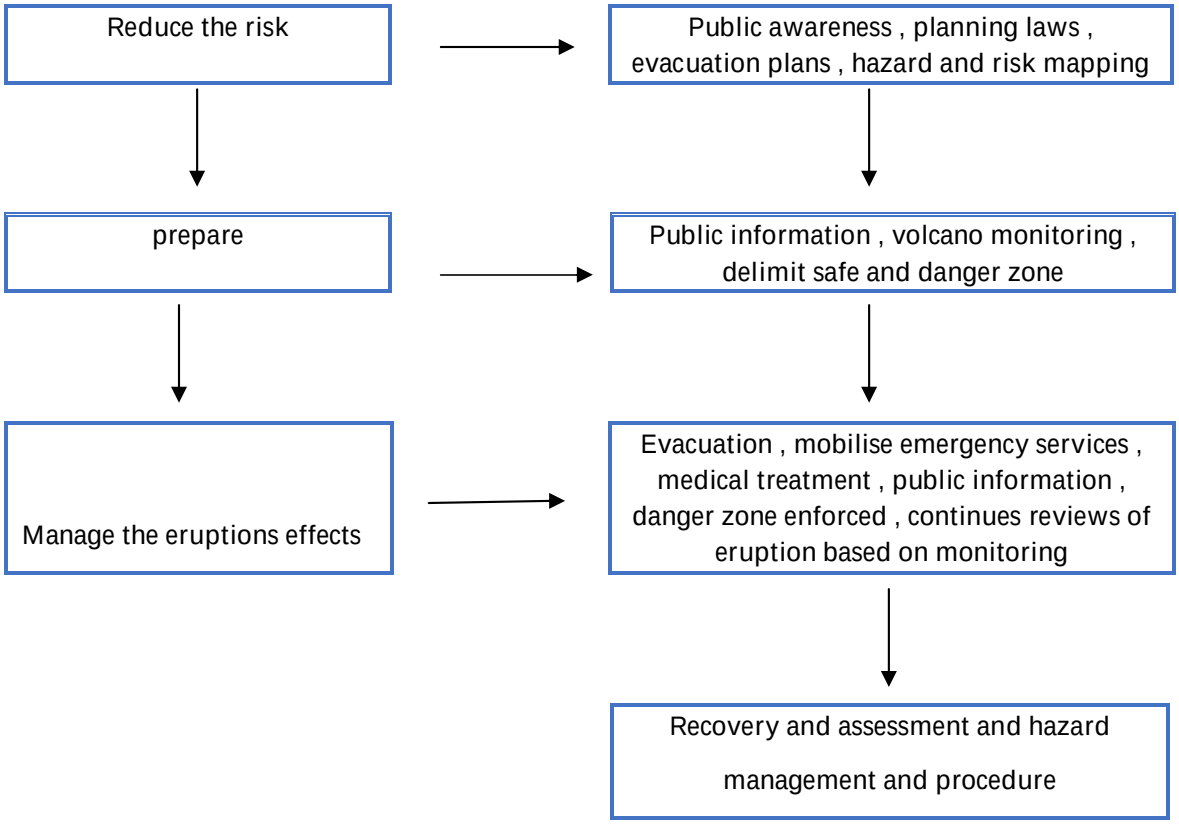


**Table1.volcanic hazard management.**

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RESEARCH ARTICLE

# The Study of Topography and Lakes Diversity in Bangaluru Silicon City of India

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## ABSTRACT

The present study niceties the topography and lakes diversity in Bangaluru. Bangaluru being an IT city and formerly known as garden city as seen different faces, from garden city to a garbage city and presently silicon city. This IT hub as a diver's topography pleasing climate and splendid habitat during this transformation lakes, pond, and puddles have become breeding ground for mosquitoes this paper derails topological study of silicon city and lake diversity.

**Key words:** topography, mosquitoes, silicon city, splendid habitat.

## INTRODUCTION

Nature is wonderful engineer and its system works flawlessly until and unless we indulge we use, utilize nature in recent times we have been exploiting nature rather than using it. (Raajasubramanian Devarajan *etal* 2015). Soil is nature's gift to nurture the plants, which intern nourishes the biotic community thus ecosystem. Now a day's this soil as become pool for various, toxins, syntactic non degradable chemicals, heavy metals etc. Soil as been polluted in all possible means, polluted water being reservoirs of chemicals heavy metals lets this chemical percolate into the soil. Polluted air caring all the toxin dust depositing on the soil, Human population explosion, rapid industrialization, increased deforestation, unplanned urbanization, scientific and technological advancement etc. have further hyped all kinds of pollution (Raajasubramanian Devarajan *etal* 2015).We have indulged is spoiling, polluting natural habitat either we have desirably added undesirable pollutant into water or altered the natural flow of water effecting the habitat and creating unhealthy competition among the habitant of the ecosystem.So this research paper attempts to derails preliminary data leading to significant research, even in this advanced scientific scenario certain preliminary data are of greater dearth this research paper give data on topology and lakes diversity in Bangalore.

### Study Area

Bangaluru City in Karnataka State of India lies between 12°44' and 13°14'N latitude and 77°25' to 77°47'E longitude and at a mean altitude of 921m above mean sea level (MSL)(Fig.1,2).

The climate of the district enjoys an agreeable temperature range from the highest mean maximum of 33°C in April to lowest of 14°C in January. It has two rainy seasons from June to September and October to November coming one after the other but with opposite wind regime, corresponding to south-west and north-east monsoons. The mean monthly relative humidity is lowest in the month of March (44%) and high during the month of June to October, being between 80 to 85% on an average. The mean annual rainfall is 859.6 mm and the mean number of rainy days is about 57. Bangaluru receives 54% of the total rainfall in the Southwest monsoon period with a rainfall of 496 mm and 34 rainy days, whereas the Northeast monsoon contributes a mean rainfall of 241 mm and mean rainy days being 14 (Kamath, 1990).

## MATERIALS AND METHODS

A detailed Survey was condected, Bangaluru was divided into North, South,East and West and the Bangaluru city map was obtained from Bangaluru corporation and survey was conducted to understand the topography and lakes diversity in Bangaluru silicon city of India. Further to study pollution level and physico-chemical analysis of possible mosquito breeding habitat in Bangalore city.

## RESULTS AND DISCUSSION

Lakes of Bangaluru occupy about 4.8% of city's geographical area (640 sq.km.) covering both urban and rural areas (Krishna et al., 1996). Bangaluru has many man-made lakes but no natural lakes. They were built for various hydrological purposes and to serve the needs of drinking water and irrigation.About 120 lakes are located in Bangaluru city, out of which three lakes were selected for the present study. The three lakes selected are, 1. Yelahanka lake, 2. Hebbal lake and 3. Muninagara lake (Fig. 5,6).

### Topography of Bangalore city lakes

**Hebbal Lake** is located about 10 kms towards North of Bangaluru and covers a water spread area of about 64.5 hectares. The geographic location details of the Lake Are13°02.569N latitude and 77°35.418E longitude at a mean altitude of 917m above MSL (Figs.3 & 4).

**Muninagara Lake** is located in Bangaluru rural district, an extension which is about 20 kms towards South of Bangalore and covers an area about 150 acres. The geographical location details of the lake are 12°44.94°N latitude and 77°32.263E longitude at a mean altitude of 921m above MSL (Figs.5 & 6).

**Yelahanka Lake** is situated in Yelahanka town, an extension which is about 15 kms towards North of Bangaluru and covers an area of about 200 acres. The geographic location details of the lake are 13°0.6382N latitude and 77°35.783 longitude at a mean altitude of 917m above MSL (Figs.7&8).

Present investigation paves way adds to preliminary data's for conducting any kind of vital studies on Bangaluru the capital city of Karnataka and IT capital of India.

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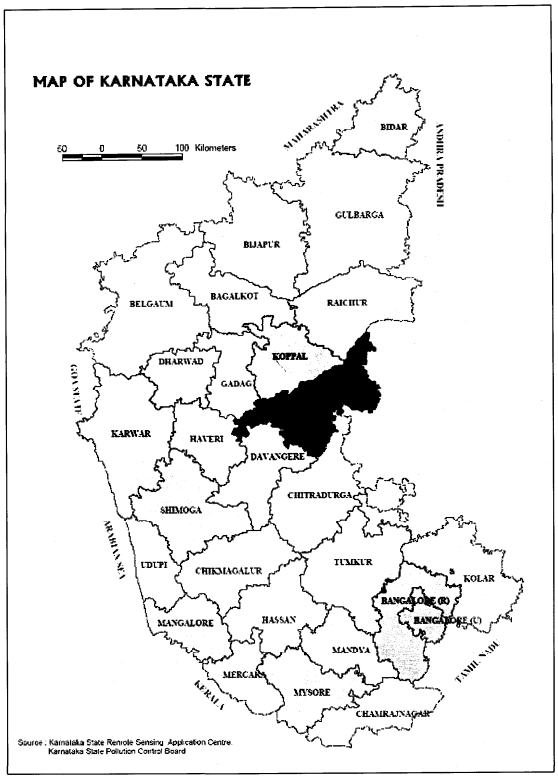


**Fig.1. Study Area**

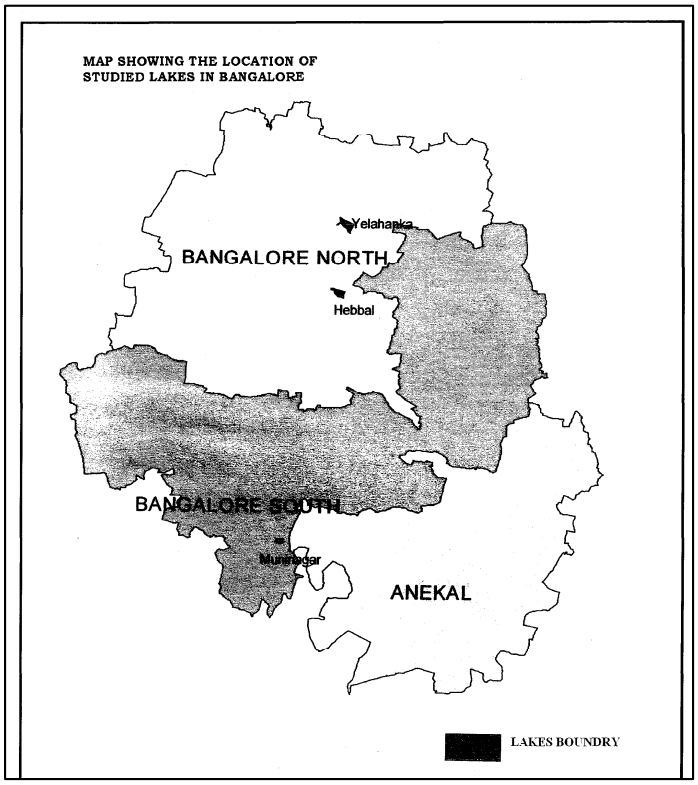


**Fig.2.Location of Studied Lakes in Bangalore**

**Fig.3.Hebbal Lake – Collection Site A**

**Fig.4.Hebbal Lake – Collection Site B**

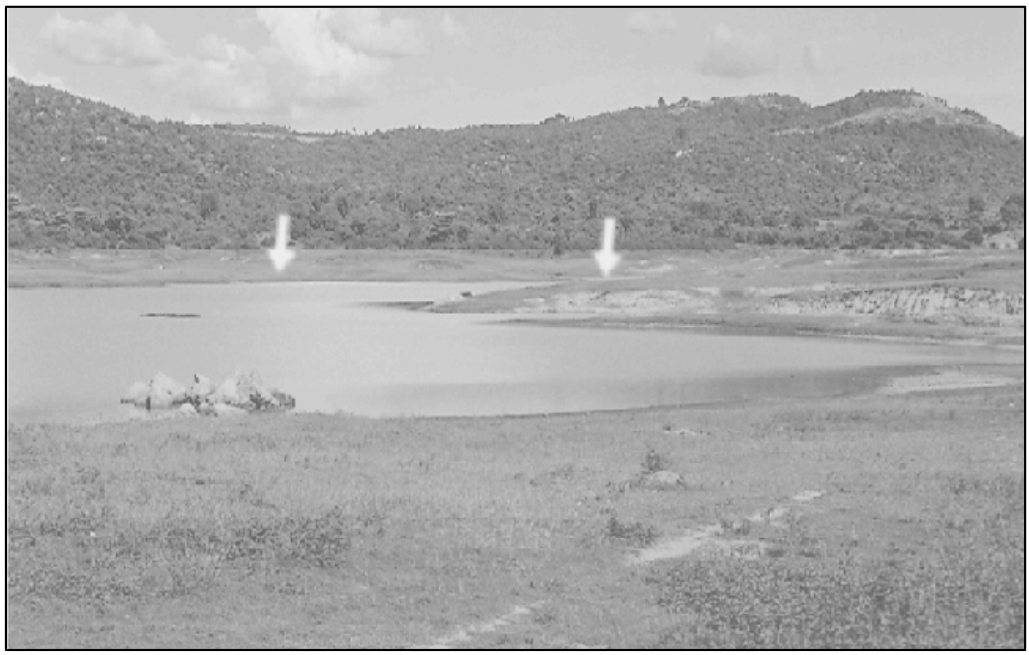

**Fig.5.Muninakara Lake – Collection Site A**

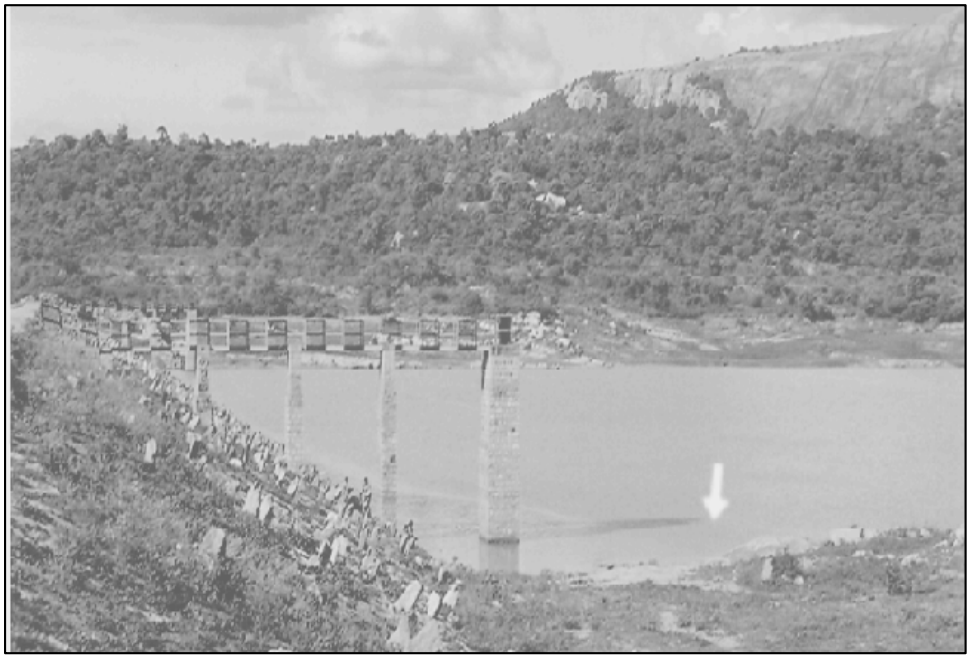

**Fig.6.Muninakara Lake – Collection Site B**

**Fig.7.Yelahanka Lake – Collection Site A**

**Fig.8.Yelahanka Lake – Collection Site B**

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RESEARCH ARTICLE

# The Study of Physico-Chemical Parameters Suspended and Dissolved Solids in Lakes of Bangaluru Garden City of India.

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## ABSTRACT

Physico-chemical parameters suspended and dissolved solids in lakes of Bangaluru are studied all this parameter is very much vital in maintaining the eco system balance this helps in steady maintenance of food chain and food web. Slight chance can have yet significant influence. Three study area lakes were chosen and several parameters like $P^H$, Conductivity (uS/cm), Turbidity (NTU), Suspended solids (mg/L) and Dissolved solids (mg/L) there was significant change in all the 3 habitat which resulted in diverse flora and fauna

**Key words:** Suspended solids, Bangaluru, Physico-chemical, Conductivity and Dissolved solids

## INTRODUCTION

Water is the most common yet the most precious resource on earth without which there would be no life on earth (Kumar and Ravindranath, 1998). It is an important factor which controls the limits of distribution of terrestrial animals (Allee et al., 1949). It is the most vital resource for the life molecule to survive. It possess a number of physical and chemical properties that help the molecule to act as best suited medium for the life activities. Most of the bio-chemical reactions that occur in the metabolism and growth of the living cells involves water, hence it has been referred to as a universal solvent. It has played a predominant role in governing the distribution of animals.

The quality of water bodies is influenced by physico-chemical environment. These factors determine the ecological conditions and also predict the nutrient status of the water. In India, water pollution is a major problem. Most of the water bodies around industrial and urban centers receive large amount of effluents either partially treated or untreated, thus affecting the quality of water. Most of the lentic water bodies are polluted due to urbanization, industrial growth and other man made problems.

Evaluation of effects due to water pollution involves physical, chemical and biological analysis. Physical and chemical parameters give information related to the time sampling, indicating levels and sources of various pollutants. The biotic and abiotic factors of aquatic ecosystem affect the life.

The physical factors are pH, light, temperature, turbidity etc. The chemical factors help in maintaining the salinity of water. The electrical conductance is mainly caused by the ions of calcium, magnesium, sodium, potassium, carbonate, sulphate, chlorides and heavy metals (Sudha Rani, 2004).The pH is a measure of the intensity of acidity or alkalinity and measures the concentration of hydrogen ions in water. The hydrogen ion concentration of the water is influenced by the biological activities, besides the addition of the chemical substances. The ions, besides maintaining the concentration of water, also acts as a buffer and thereby preserve the natural alkalinity.This research paper attempts to brief Physico-Chemical Factors of water bodies of Bangalore

### Study Area

Bangalore City in Karnataka State of India lies between 12°44' and 13°14'N latitude and 77°25' to 77°47'E longitude and at a mean altitude of 921m above mean sea level (MSL).

The climate of the district enjoys an agreeable temperature range from the highest mean maximum of 33°C in April to lowest of 14°C in January. It has two rainy seasons from June to September and October to November coming one after the other but with opposite wind regime, corresponding to south-west and north-east monsoons. The mean monthly relative humidity is lowest in the month of March (44%) and high during the month of June to October, being between 80 to 85% on an average. The mean annual rainfall is 859.6 mm and the mean number of rainy days is about 57. Bangalore receives 54% of the total rainfall in the Southwest monsoon period with a rainfall of 496 mm and 34 rainy days, whereas the Northeast monsoon contributes a mean rainfall of 241 mm and mean rainy days being 14 (Kamath, 1990).

## MATERIAL AND METHODS

Hydrological samplings were conducted in the selected three lakes at monthly intervals throughout the period of study (Nov 2003 to Oct 2005). The monthly rainfall data was obtained from Indian Meteorological Department, Bangalore and calculated for average values. The atmospheric and ambient water temperature were recorded at the time of sampling. Water samples were collected from surface at monthly intervals in polythene cans (size 2 lts.) between 7 and 9 a.m. for a period of two years from Nov 03 to Oct 05. The samples were analysed in the laboratory on the same day for various physico-chemical parameters.

The water quality analysis of the lake gives the exact nature, cause and levels of the pollutants. The physical parameters such as temperature and turbidity play an important role in lake productivity. The methods adopted for water quality analyses are listed in the table below:

**Temperature**

Measurement of temperature is an important parameter required to get an idea of self purification of reservoirs and lakes. Water temperature plays an important role in aquatic ecosystem health. The temperature of drinking water has an influence on its taste.Temperature of the atmosphere and water samples were recorded at the time of water collection with the help of a centigrade thermometer.

**Conductivity**

Conductivity is a measure of the ability of an aqueous solution to carry an electric current. This ability depends on the presence of ions, on their total concentration, mobility and valence and on the temperature of measurement. It was recorded in the laboratory with a conductivity meter (APHA, 2000). Electrical conductivity (EC) of the sample indicates the concentration of ionisable constituents of the solution.

**Turbidity**

Suspension of particles in water interfering the passage of light is called turbidity. Turbidity is caused by wide variety of suspended matter which range in size from colloidal to coarse dispersion depending upon the degree of turbulence.Turbidity was estimated by Nephelometric method (AHPA, 2000). This method is based on a comparison of the intensity of light scattered by the sample under defined conditions with the intensity of light scattered by a standard reference suspension under the same conditions. Higher the intensity of scattered light, higher the turbidity.The digital Nepheloturbidity meter was adjusted to 40NTU using standard turbidity suspension. Then the sample was taken in a cuvet and readings were noted down directly.

**Suspended Solids**

The suspended solids refer to matters suspended or dissolved in water on waste water. Suspended solid analysis is important in the control of biological and physical waste water treatment processes and for assessing effluent limitations. The total suspended solids are the portion of total solids retained by a filter of 2.0◎m (or smaller) pore size under specified conditions.Suspended solids were estimated by Gravimetric method (APHA, 2000). Analysis of water sample was done as soon as possible after reaching laboratory.Glass fiber filter papers were weighed and kept in oven at 103-105°C. 50ml or aliquot quantity of sample was transferred to filtration unit and passed with suction pump. After completion of filtration, the residue was washed with 50ml of distilled water and the filter paper was removed carefully with the help of a needle. The filter paper was weighed after drying in oven at 103 to 105°C. The difference in the weight gives the amount of suspended solids.

**Dissolved Solids**

Dissolved solids is the portion of solids that passes through a filter of 2.0◎m (or smaller) pore size under specified conditions. It was estimated by Gravimetric method (APHA, 2000).A well mixed sample was filtered through a standard glass fiber filter and the filtrate was evaporated to dryness in a pre-weighed dish and dried at 180°C. The difference in the weight gives the amount of total c . solids.

**pH**

Measurement of pH is one of the most important and frequently used tests in water chemistry. Practically every phase of water supply and waste water treatment such as acid base neutralization, water softening, precipitation, coagulation, disinfection and corrosion control depend on hydrogen ion. At a given temperature the intensity of the acidic or basic character of a solution is indicated by pH or hydrogen ion activity. It is estimated by electrometrical method (APHA, 2000). pH measurement for the sample was done immediately within 2 hrs of collection in the laboratory with the help of digital pH meter (Electrometric method).The basic principle lies in the determination of activity of hydrogen ions by potentiometric measurement using combination of glass and reference electrodes with temperature correction which responds selectively to hydrogen ions.

## RESULTS AND DISCUSSION

The pollution of lakes by industrial wastes and domestic sewage has increased enormously leading to most unsanitary conditions in the environment. This is due to rapid growth of population and industrialisation in the country (Purandara et al., 2003). In India, 80% of the surface water is exposed to pollution. More than 95% of the sewage in the country is not treated. The amount of raw sewage entering the rivers is still increasing. Industrialisation causes huge environmental problems (Zwart and Trivedi, 1995; Nirmal, 1997; Jain et al., 1996; Narain and Chauhan, 2000). In the present study, average values of physico-chemical parameters studied for two years of all the three water lake bodies (having 2 stations each) have been described in detail and also in relation to their seasonal fluctuations, yearly average, interrelationships with plankton and total number of mosquito breeding.

Nature is wonderful engineer and its system works flawlessly until and unless we indulge we use, utilize nature in recent times we have been exploiting nature rather than using it (RaajasubramanianDevarajan2015).All the physical and chemical parameters play a significant role on flora and fauna and helps to study the complex food chains and food web in the eco system. Change in $P^H$, Conductivity (uS/cm), Turbidity (NTU), Suspended solids (mg/L) and Dissolved solids (mg/L) there was significant change in the all the habitat which resulted in diverse flora and fauna study in relation with different habitat at different geographical area is necessary to place in record a detail hypothesis on flora and fauna.

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| PARAMETERS | METHOD USED |
|---|---|
| **Physical** | |
| Water temperature (°C) | Centigrade thermometer |
| Conductivity (uS/cm) | Electrometric method |
| Turbidity (NTU) | Nephelometric method |
| Suspended solids (mg/L) | Gravimetric method |
| Dissolved solids (mg/L) | Gravimetric method |

**Table 1 : Variations in Temperature at the studied lakes**

| | LAKES | | | | | |
|---|---|---|---|---|---|---|
| **MONTH &** | **HEBBAL** | | **MUNINAGARA** | | **YELAHANKA** | |
| **YEAR** | **Station** | | **Station** | | **Station** | |
| | **'A'** | **R** | **'A'** | **'B'** | **'A'** | **'B'** |
| Nov 03 | 22 | 22 | 22 | 22 | 22 | 22 |
| Dec 03 | 21 | 21 | 22 | 21 | 21 | 21 |
| Jan 04 | 23 | 23 | 23 | 23 | 23 | 23 |
| Feb 04 | 22 | 22 | 22 | 22 | 22 | 22 |
| Mar 04 | 24 | 24 | 24 | 24 | 24 | 24 |
| Apr 04 | 24 | 24 | 24 | 24 | 24 | 24 |
| May 04 | 24 | 24 | 24 | 24 | 24 | 24 |
| Jun 04 | 24 | 24 | 24 | 24 | 24 | 24 |
| Jul 04 | 23 | 23 | 23 | 23 | 23 | 23 |
| Aug 04 | 22 | 22 | 22 | 22 | 22 | 22 |
| Sep 04 | 22 | 22 | 22 | 22 | 22 | 22 |
| Oct 04 | 22 | 22 | 22 | 22 | 22 | 22 |
| Year Average | 22.75± 1.01 | 22.75± 1.01 | 22.75± 1.01 | 22.75± 1.01 | 22.75± 1.01 | 22.75± 1.011 |
| Nov 04 | 21 | 21 | 22 | 22 | 22 | 22 |
| Dec 04 | 21 | 21 | 21 | 21 | 21 | 21 |
| Jan 05 | 23 | 23 | 23 | 23 | 23 | 23 |
| Feb 05 | 22 | 22 | 22 | 22 | 22 | 22 |
| Mar 05 | 24 | 24 | 24 | 24 | 24 | 24 |
| Apr 05 | 24 | 24 | 24 | 24 | 24 | 24 |
| May 05 | 24 | 24 | 24 | 24 | 24 | 24 |
| Jun 05 | 23 | 23 | 23 | 23 | 23 | 23 |
| Jul 05 | 23 | 23 | 23 | 23 | 23 | 23 |
| Aug 05 | 22 | 22 | 22 | 22 | 22 | 22 |
| Sep 05 | 22 | 22 | 22 | 22 | 22 | 22 |
| Oct 05 | 22 | 22 | 22 | 22 | 22 | 22 |
| Year Average | — | — | 22.75± 1.01 | 22.75+ 1.01 | 22.75± 0.1 | 22.75± 0.1 |

**Table 2 : Variations in Conductivity at the studied lakes**

| MONTH & YEAR | LAKES | | | | | |
|---|---|---|---|---|---|---|
| | HEBBAL | | MUNINAGARA | | YELAHANKA | |
| | Station | | Station | | Station | |
| | 'A' | 'B' | 'A[1] | 'B' | 'A' | 'B' |
| Nov 03 | 1352 | 1150 | 440 | 390 | 2750 | 2420 |
| Dec 03 | 1392 | 1162 | 295 | 380 | 2770 | 2440 |
| Jan 04 | 1442 | 1162 | 298 | 285 | 2850 | 2520 |
| Feb 04 | 1400 | 1195 | 302 | 292 | 2900 | 2510 |
| Mar 04 | 1575 | 1300 | 322 | 310 | 2950 | 2540 |
| Apr 04 | 1589 | 1305 | 382 | 341 | 3020 | 2552 |
| May 04 | 1650 | 1339 | 410 | 385 | 3115 | 2575 |
| Jun 04 | 1648 | 1335 | 425 | 390 | 3116 | 2576 |
| Jul 04 | 1458 | 1108 | 392 | 330 | 3023 | 2549 |
| Aug 04 | 1225 | 952 | 298 | 248 | 2715 | 1900 |
| Sep 04 | 824 | 750 | 271 | 221 | 2210 | 1285 |
| Oct 04 | 832 | 762 | 280 | 228 | 2215 | 1290 |
| Year Average | 1365.5 ±256.2 | 1126.6 ±247.4 | 342.9 ±59.3 | 308.3 ±57.8 | 2802.8 ±293.4 | 2581.5 ±293.4 |
| Nov 04 | 1055 | 863 | 300 | 250 | 2529 | 2199 |
| Dec 04 | 1105 | 875 | 155 | 140 | 2549 | 2219 |
| Jan 05 | 1155 | 875 | 158 | 145 | 2629 | 2299 |
| Feb 05 | 113 | 908 | 162 | 152 | 2679 | 2289 |
| Mar 05 | 1288 | 1013 | 182 | 170 | 2729 | 2319 |
| Apr 05 | 1302 | 1018 | 242 | 201 | 2799 | 2331 |
| May 05 | 1363 | 1052 | 270 | 245 | 2894 | 2354 |
| Jun 05 | 1361 | 1048 | 285 | 250 | 2895 | 2355 |
| Jul 05 | 1171 | 821 | 252 | 190 | 2802 | 2328 |
| Aug 05 | 938 | 665 | 158 | 108 | 2494 | 1679 |
| Sep 05 | 537 | 463 | 131 | 81 | 1989 | 1064 |
| Oct 05 | 545 | 475 | 140 | 88 | 1994 | 1069 |
| Year Average | 1077.7 ±269.1 | 839.7 ±195.7 | 202.9 ±59.3 | 168.3 ±57.8 | 2263.1 ±470.4 | 2042.0 +470.4 |

**Table 3 : Variations in Turbidity at the studied lakes**

| MONTH & YEAR | LAKES | | | | | |
|---|---|---|---|---|---|---|
| | HEBBAL | | MUNINAGARA | | YELAHANKA | |
| | Station | | Station | | Station | |
| | 'A' | B[1] | 'A' | 'B' | 'A' | 'B' |
| Nov 03 | 31 | 23 | 80 | 75 | 58 | 35 |
| Dec 03 | 28 | 21 | 78 | 72 | 55 | 33 |
| Jan 04 | 28 | 20 | 66 | 66 | 54 | 31 |
| Feb 04 | 24 | 18 | 64 | 64 | 50 | 28 |
| Mar 04 | 21 | 15 | 58 | 56 | 48 | 25 |
| Apr 04 | 21 | 16 | 50 | 54 | 45 | 27 |
| May 04 | 18 | 12 | 45 | 45 | 42 | 23 |
| Jun 04 | 16 | 12 | 32 | 32 | 39 | 21 |
| Jul 04 | 22 | 18 | 40 | 38 | 46 | 24 |
| Aug 04 | 30 | 25 | 69 | 55 | 58 | 38 |
| Sep 04 | 45 | 36 | 78 | 62 | 62 | 62 |
| Oct 04 | 45 | 36 | 76 | 62 | 60 | 62 |
| Year Average | 27.4± 9.0 | 21.0±7.7 | 61.3± 15.6 | 56.8± 12.5 | 52.3± 6.2 | 34.1± 13.4 |
| Nov 04 | 26 | 18 | 70 | 65 | 53 | 30 |
| Dec 04 | 23 | 16 | 68 | 62 | 50 | 28 |
| Jan 05 | 23 | 15 | 56 | 51 | 49 | 26 |
| Feb 05 | 19 | 13 | 54 | 48 | 45 | 23 |
| Mar 05 | 16 | 10 | 48 | 46 | 43 | 20 |
| Apr 05 | 16 | 11 | 40 | 44 | 40 | 22 |
| May 05 | 13 | 07 | 35 | 29 | 37 | 18 |
| Jun 05 | 11 | 07 | 22 | 18 | 34 | 16 |
| Jul 05 | 17 | 13 | 30 | 28 | 41 | 19 |
| Aug 05 | 25 | 20 | 59 | 45 | 53 | 33 |
| Sep 05 | 40 | 31 | 68 | 52 | 57 | 57 |
| Oct 05 | 40 | 31 | 6 | 52 | 55 | 57 |
| Year Average | 22.4± 9.0 | 16.0±7.3 | 51.3± 15.6 | 45.0± 13.3 | 46.4± 7.2 | 29.0± 13.4 |

**Table 4 : Variations in Suspended Solids at the studied lakes**

| | LAKES | | | | | |
|---|---|---|---|---|---|---|
| **MONTH &** | **HEBBAL** | | **MUNINAGARA** | | **YELAHANKA** | |
| **YEAR** | **Station** | | **Station** | | **Station** | |
| | **'A'** | **'B'** | **'A'** | **'B'** | **'A'** | **'B'** |
| Nov 03 | 85 | 39 | 26 | 24 | 95 | 80 |
| Dec 03 | 72 | 35 | 25 | 22 | 94 | 80 |
| Jan 04 | 65 | 32 | 23 | 20 | 94 | 78 |
| Feb 04 | 58 | 32 | 23 | 20 | 94 | 76 |
| Mar 04 | 45 | 29 | 21 | 18 | 88 | 68 |
| Apr 04 | 38 | 25 | 18 | 16 | 81 | 68 |
| May 04 | 28 | 20 | 18 | 14 | 78 | 63 |
| Jun 04 | 28 | 20 | 12 | 10 | 74 | 59 |
| Jul 04 | 38 | 26 | 25 | 18 | 85 | 68 |
| Aug 04 | 52 | 38 | 34 | 25 | 88 | 75 |
| Sep 04 | 72 | 52 | 46 | 32 | 102 | 92 |
| Oct 04 | 70 | 48 | 46 | 32 | 98 | 92 |
| Year Average | 54.3± 18.1 | 33.0± 9.7 | 26.4± 10.1 | 20.9± 6.3 | 89.3± 8.1 | 75.2± 9.8 |
| Nov 04 | 75 | 29 | 16 | 14 | 85 | 70 |
| Dec 04 | 62 | 25 | 15 | 12 | 84 | 70 |
| Jan 05 | 55 | 22 | 13 | 10 | 84 | 68 |
| Feb 05 | 48 | 22 | 13 | 10 | 84 | 66 |
| Mar 05 | 35 | 19 | 11 | 08 | 78 | 61 |
| Apr 05 | 28 | 5 | 08 | 06 | 71 | 58 |
| May 05 | 18 | 10 | 08 | 04 | 68 | 53 |
| Jun 05 | 18 | 10 | 06 | 04 | 64 | 49 |
| Jul 05 | 28 | 16 | 15 | 08 | 75 | 58 |
| Aug 05 | 42 | 28 | 24 | 15 | 78 | 65 |
| Sep 05 | 62 | 42 | 36 | 22 | 92 | 82 |
| Oct 05 | 60 | 38 | 36 | 22 | 88 | 82 |
| Year Average | 44.3± 18.3 | 25.8± 18.3 | 16.7± 9.7 | 11.2± 5.8 | 79.2± 8.1 | 65.1± 9.8 |

**Table 5 : Variations in Dissolved Solids at the studied lakes**

| MONTH & YEAR | LAKES | | | | | |
|---|---|---|---|---|---|---|
| | HEBBAL | | MUNINAGARA | | YELAHANKA | |
| | Station | | Station | | Station | |
| | 'A' | 'B' | 'A' | 'B' | 'A' | 'B' |
| Nov 03 | 872 | 725 | 250 | 200 | 1720 | 1500 |
| Dec 03 | 883 | 730 | 185 | 170 | 1780 | 1515 |
| Jan 04 | 932 | 750 | 187 | 175 | 1852 | 1588 |
| Feb 04 | 915 | 760 | 190 | 179 | 1883 | 1596 |
| Mar 04 | 995 | 790 | 215 | 200 | 1920 | 1610 |
| Apr 04 | 1050 | 805 | 242 | 220 | 1960 | 1615 |
| May 04 | 1096 | 851 | 269 | 242 | 2020 | 1635 |
| Jun 04 | 1092 | 848 | 277 | 248 | 2018 | 1640 |
| Jul 04 | 955 | 725 | 255 | 210 | 1966 | 1614 |
| Aug 04 | 820 | 620 | 192 | 155 | 1730 | 1210 |
| Sep 04 | 525 | 415 | 172 | 130 | 1300 | 810 |
| Oct 04 | 532 | 425 | 179 | 140 | 1320 | 815 |
| Year Average | 888.4+ 180.4 | 702+ 139.1 | 277+ 36.7 | 189.0+ 36.0 | 1789+ 235.2 | 1429+ 297.5 |
| Nov 04 | 692 | 545 | 190 | 140 | 1540 | 1320 |
| Dec 04 | 703 | 550 | 125 | 110 | 1600 | 1335 |
| Jan 05 | 752 | 570 | 127 | 115 | 1672 | 1408 |
| Feb 05 | 735 | 580 | 130 | 119 | 1703 | 1416 |
| Mar 05 | 815 | 610 | 155 | 140 | 1740 | 1430 |
| Apr 05 | 870 | 625 | 182 | 160 | 1780 | 1435 |
| May 05 | 916 | 671 | 209 | 182 | 1840 | 1455 |
| Jun 05 | 912 | 668 | 217 | 188 | 1838 | 1460 |
| Jul 05 | 775 | 545 | 195 | 150 | 1786 | 1434 |
| Aug 05 | 640 | 440 | 132 | 95 | 1550 | 1030 |
| Sep 05 | 345 | 235 | 112 | 70 | 1120 | 630 |
| Oct 05 | 352 | 245 | 119 | 80 | 1140 | 635 |
| Year Average | 708.9 ±180.9 | 520.3 ±139.8 | 157.7 ±36.7 | 129.0 ±36.0 | 1617.4 ±242.9 | 1249 ±297.5 |

**Table 6 : Variations in pH at the studied lakes**

| MONTH & YEAR | LAKES | | | | | |
|---|---|---|---|---|---|---|
| | HEBBAL | | MUNINAGARA | | YELAHANKA | |
| | Station | | Station | | Station | |
| | 'A' | 'B' | 'A' | 'B' | 'A' | 'B' |
| Nov 03 | 6.9 | 7.3 | 6.9 | 7.2 | 7.1 | 7.4 |
| Dec 03 | 6.8 | 7.2 | 7.0 | 7.2 | 7.0 | 7.4 |
| Jan 04 | 6.9 | 7.3 | 6.9 | 7.2 | 7.0 | 7.3 |
| Feb 04 | 6.9 | 7.3 | 6.8 | 7.1 | 7.1 | 7.4 |
| Mar 04 | 6.8 | 7.1 | 6.8 | 7.2 | 6.9 | 7.3 |
| Apr 04 | 6.6 | 7.0 | 6.7 | 7.1 | 6.9 | 7.2 |
| May 04 | 6.5 | 6.9 | 6.6 | 7.0 | 6.8 | 7.1 |
| Jun 04 | 6.5 | 6.9 | 6.6 | 6.9 | 6.8 | 7.1 |
| Jul 04 | 6.8 | 7.2 | 7.0 | 7.2 | 6.0 | 7.2 |
| Aug 04 | 7.0 | 7.4 | 7.2 | 7.5 | 7.2 | 7.4 |
| Sep 04 | 7.2 | 7.6 | 7.3 | 7.8 | 7.3 | 7.5 |
| Oct 04 | 7.2 | 7.5 | 7.3 | 7.8 | 7.2 | 7.5 |
| Year Average | 6.8± 0.22 | 7.23± 0.21 | 6.92± 0.235 | 7.26± 0.27 | 7.03± 0.153 | 7.32± 0.134 |
| Nov 04 | 7.0 | 7.4 | 7.1 | 7.4 | 7.2 | 7.5 |
| Dec 04 | 6.9 | 7.3 | 7.2 | 7.4 | 7.2 | 7.6 |
| Jan 05 | 7.1 | 7.4 | 7.0 | 7.3 | 7.2 | 7.5 |
| Feb 05 | 7.0 | 7.4 | 7.0 | 7.3 | 7.2 | 7.5 |
| Mar 05 | 6.1 | 7.2 | 6.9 | 7.3 | 7.1 | 7.5 |
| Apr 05 | 6.8 | 7.2 | 6.8 | 7.2 | 7.0 | 7.2 |
| May 05 | 6.7 | 7.0 | 6.8 | 7.2 | 6.9 | 7.1 |
| Jun 05 | 6.7 | 7.1 | 6.8 | 7.1 | 6.8 | 7.1 |
| Jul 05 | 6.9 | 7.2 | 7.2 | 7.4 | 7.2 | 7.4 |
| Aug 05 | 7.2 | 7.6 | 7.4 | 7.7 | 7.4 | 7.6 |
| Sep 05 | 7.4 | 7.8 | 7.5 | 8.0 | 7.4 | 7.5 |
| Oct 05 | 7.3 | 7.6 | 7.5 | 8.0 | 7.2 | 7.5 |
| Year Average | 6.9± 0.23 | 7.35± 0.220 | 7.1± 0.26 | 7.4± 0.28 | **7.1**5± 0.171 | **7.41±** 0.173 |

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RESEARCH ARTICLE

# Rural Credits Cooperatives Financial System and its Contribution for Rural Development in China

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## ABSTRACT

The paper elaborated the Rural Credits Cooperatives financial system and its contribution for rural development in China. Rural Credit Cooperatives as cooperative financial establishments and improves the performance consistently every year and constituted about 12.3 % all financial loans in China. Rural Credit Cooperatives was constituted an integral and vital part of rural reforms for the development in China.

**Key words:** Development of rural people, utilizing local talent, loans and development of agriculture sector.

## INTRODUCTION

The Period of 1950s, a channel of Rural Credit Cooperatives was created. During this period, they weren't commercially enterprises same as to the banks, but some other functional departments credit between the official and the people's communes in countryside areas (Tsai, 2004).During late 1970s, when economic reforms after they facilitate certain separate entrepreneurialism and the design of collective enterprises, the Rural Credit Cooperatives starts to activate as bases banks that deliver praise and savings accounts to people and mutual enterprises. During reform stage, the Rural Credit Cooperatives were intentional to perform as a means for the Agricultural Bank of China to direct credit into rural parts. However, the Rural Credit Cooperatives were lacking to fulfill the high requirement for credit in those parts (Tsai, 2002).

Distinctive and enterprises often try to other ways of credit, which variety from Rotating Savings and Credit Associations to pawn shops. Though there are some different practices are all unlawful (while some were accepted to a larger degree instead of others, and were accepted in few places more than in others). Commercial banks are lawful, but from the time when rarer than one percent of loans from state banks goes to self-contained entrepreneurs, they can not fulfill all credits which the rural areas need (Tsai, 2004).Till 1996, the Rural Credit Cooperatives are

organized by the Agricultural Bank of China. In 1996, they were supervised by the People's Bank of China (Tsai, 2004).In July 1998, the rural credit foundations were disqualified when the State Council of the People's Republic of China published the "Provisions on the Cancellation of Illegal Financial Institutions and Activities." This departed the Rural Credit Cooperatives as the only lawful financial institution (other than banks) which helped rural enterprises and entities. In 1998, when the Rural Credit Foundations were disqualified, there were almost 44,000 township-level Rural Credit Cooperatives and 280,000 village-level Rural Credit Cooperatives branches (Tsai, 2002).From the time when 1998, several of the Rural Credit Cooperatives have steadily been transformed into rural commercial banks (RCBs). The firstly foreign investment in an Rural Commercial Bank was permitted in 2006, when Rabobank and the International Finance Corporation (the private sector arm of the World Bank) assimilated stakes in the United Rural Cooperative Bank of Hangzhou (Tsai, 2004).

The definition of Rural Credit Cooperatives is cooperative financial establishment. As from the owner's evenhandedness in the credit union's balance sheet, no countries in the owner's evenhandedness. The paid-in capital is collected of the stock of cooperative associates and the stock of enterprises. According to that, the land truths of the credit cooperatives are well-defined, owned enterprises, according to this reason; credit cooperatives should goes to private finance. In existence, some credit unions goes to state-owned finance (still coordinated by the government), others goes to private finance (actually taken over by citizens), and some collapse in between. RCCs in rural financial exercise in the fundamental situation and the role of the main force are beyond hesitation. The problem of whether it can truly be converted into a machine that complies with worldwide acknowledged cooperative financial principles has been discussed for some time. For a long period, as the inauguration of its formation, China's RCCs have been jointly kept and progressed closer to the state-owned. The "government-run" awareness and functioning are very solid and are trustworthy for the shareholders. Their cooperative finance is "voluntary, mutual aid, common benefit, equality and low productivity. The type of financial and financial services can not redirect much. Partners of cooperatives can not only like the significance and special well of the loans but also extra rights of stocks and extras, but also receive loans that are much higher than the benchmark interest rate. The self-governing management system such as the general assembly, the board of directors, unit appointments system is quite better than the electoral system. China's Cooperative Financial exercise has always been connected with RCCs. While several improvements have taken place, "RCCs have never accurately become Cooperative Financial Organization, which have begun people's suspicion of the basis for the being of cooperative finance. It should be celebrated that in our country, Rural Credit Cooperatives depart from the principle of cooperation for a long period and has objectives motives for their presence. Government involvements, legislative gap, defect in property right systems and heavy historical burden.

## Hubei Provincial Rural Credit Cooperative Union

Hubei Provincial Rural Credit Cooperative Union (hereinafter referred to as "Provincial Rural Credit Cooperative") was started on July 28, 2005, supposed the management of RCCs throughout the province, managerial, directing and service roles.

In July 2012 end of province Agricultural letter social system has one home city equal Agricultural firm (Wuhan Agricultural Firm), and 7 home city equal Association (which Ezhou city Association for combined corporate social), and 4 are city State Office, 20 home Agricultural Firm, 12 home Agricultural collection line, 45 home County Association, 2,103 are business points, further 27,000 more employed personnel, is Hubei Province within resources scale highest, and business points up, and services ranges best inclusive of local financial institution. As the establishing of provincial Rural Credit Cooperative, the "base community oriented to Agriculture, SME-oriented, county Economic-oriented" market situation, concentrative changes and try to systematize core management, and constantly expand services stages, to hasten business development. From July 2012, the provincial Rural Credit Union credits 333.1 billion Yuan and wonderful loan of 211.8 billion Yuan, individually four times and 4.77 times the Associations were firstly founded, with relevant parts of financial organizations throughout the province, 12.53%, total Yuan loans 1th of banking organizations within the province.

**Rural Credit Cooperatives in Anhui Province**

Rural Credit Cooperative in Anhui Province, was formed in 1951, passing through some modifications in 1996 and after the developing of Agricultural Bank, controlled by the people's Bank, in 2003 and was located under the management of banking officials. In October 9, 2004, the Standing Committee of the provincial group Committee meeting definite to set up Anhui Provincial Rural Credit Cooperative Union, Entrance equal beside the management in civic organizations.On November 2004, positively achieved formation agreement and start of the China Banking Regulatory Commission accepted, accomplishment of constitutional processes such like business registering, achieved financial license, and the inaugural ceremony was held in December 18, procedure officially, firstly in the second set of test provinces to inaugurate provincial association, applied variations of supervision system in Rural Credit Cooperatives through the province. Anhui Provincial RCCs Union Association (hereinafter Association) was established in December 18, 2004, by the 83 regions of the province (municipal and district) equal cooperatives combined undertaking, local financial institutions with self-governing lawful ranks, recorded capital of 50 millions Yuan.

Key functions of the provincial association is authorized by the provincial Government, rural cooperative financial organizations through the province (including rural commercial banks and rural cooperative banks, RCCs) for the supervision, assistance, proficiency, and services. Association of RCCs in Anhui Province has 83 Firms parts throughout the province, nearly 30,000 employees, concealing 17 cities and counties across the province. At present, the internal institutions at the provincial members societies overall contains of 8, 2 straight agencies and 14 offices.System-wide overall of 83 bodies of lawful persons, in which five rural commercial banks, rural banks, 22 County-level Associations 56, under the authority of 3,048 branches, totally employed staff of 29,054 peoples, number of channels, and around 40% of employees of banking organizations within the province. For 5 years, the province's rural cooperative financial organizations continuously claim based on Agriculture, farmers and rural regions, service-oriented SMEs, developing changes, consolidation inner governor constructions, identical operations and managements, grown-up for the maximum broad province-wide exposure of local, communities and selling banking institution. In the end of 2009, the province's RCCs financial institution deposit of 206.95 billions Yuan, rising by 1.9 times above by the end of 2004, loan of 144.93 billions Yuan, rising by 1.8 times above by the end of 2004.

**Shandong Provincial Rural Credit Cooperative Union Association**

Shandong provincial RCCs Union Association was established in June 2004, comprised by RCCs and Rural Banks within the province parts. Rural Cooperatives banks within province to workout managements, direction, skillful ideas, and services for function of the financial institution, financed by Shandong Provincial People's Government, the State-owned properties supervisions and Administrative Commission to fulfil the qualifications (Xie 2001).

Shandong Province RCCs was established on 1951 and now has 132 lawful persons at the county level (with rural cooperative banks 19, county cooperatives 113), the organization has 5,564, 62,000 employees, highest channels within China and throughout the province, the largest variety of service, capital of the biggest unified multifunctional native financial institution. On August 2003, the test change of Shandong RCCs by State Council to develop the province.In June 6, 2004, the Shandong provincial RCCs Union Association was established, noticeable the increasing the reforms of RCCs throughout the province has accomplished early result, exposed the reforms and improvement of the new phase. Province Association established HOU, in provincial and provincial Government of perfectly led Xia, in level the apprehensive of monitor support Xia, thorough studying practices science development opinions, effective development operations national economic financial policy, strongly across province economic development whole, started has "12346" of work consideration and development objectives, insisted to deepening reforms for power, to business development towards central line, to facilitate "three Agricultural" for reasons, key functioning indicator produce history top level, additional associating the key force of rural financial situation,

convincingly upkeep the province's economic and social development. In the end of 2008, overall deposit reached to 490 billions Yuan, with the growth of 93.4 billion Yuan, with the growth of 23.6%; the total of terrific loans extended 368.9 billions Yuan, with growth of 51.5 billions Yuan, with growth of 16.2%. Agriculture loans 245.7 billions Yuan, with growth of 36.8 billions Yuan, with of increase of 17.6%.

## Current State of Rural Credit Cooperatives Reforms

With earlier functioning, Rural Credit Cooperatives experienced with remaining profits of 23.3 billions Yuan and non-performing loan ratio correct 5.6% in 2010. With improvements in their resources characteristic can be mostly credited and bailouts from the fundamental Government. During 1990s Rural Credit Cooperatives procedure was basically unsound. The money capability percentage and remaining equity of Rural Credit Cooperatives were in undesirable territory; that means their disadvantages were so massive that they were eating equity of investors. Rural Credit Cooperatives are together the key owners of rural family's investments and the key contributors of domestic credits. During 2007, the People Bank of China provided two systems of financial support 168 billions Yuan debt-for-bonds exchanges and 830 millions Yuan in saved loans to help Rural Credit Cooperatives in removing bad moneys and writing off historical losses. RCCs are the best as a myriad of locality based credit organizations with unreliable financial functioning and asset class. Reforms have also converted several credits cooperative into rural commercial banks and rural cooperative banks. Although RCCs are mostly profit-oriented organizations, they are highly given confident to offer to rural families and Agricultural debtors.

## Situations of Loans and Interest Rate in China

**1. Legal Interest Rate**

By the authorization of State Council and approved prepared by the People's Bank of China level for all types of lawful significance rates.

**2. Benchmark Interest rate**

The People's Bank of China, profitable banks and other economic organizations of credits and loans significance rates are the Benchmark interest rate.

**3. Contract Rate**

Investor giving to lawful loans interest rate fixed by the People's Bank of China and the moving opportunity of involvement, settled with the nonpayers and the constant be identified in the loan deal with specific loan interest rate.

## Loan Situations

To applying loan it has a next situation for nonpayers;

1. Nonpayers must capacity to debt-services costs on program.
2. By adding to the normal person and not accepted by the section of industry and commerce process of enterprise lawful individual.
3. Rural loan has elementary accounts or a common deposit account and keep it within the definite boundary.
4. The assurance and debt it must be match to the supplies of the loan sponsor and loan pledgee.
5. The Rural properties and obligations of the nonpayers shall not be greater than 70 percent.
6. The Rural nonpayers must allow to the rules of the People's Bank of China credit card.

## RCCs Motivations

Key reasons of Rural Credit Cooperatives contain altogether powers to expand the regional financial infrastructures to inspire and take an effective interest in the microfinance. It is not the easy job and Rural Credit Cooperatives may not practice any source of refinancing for the microcredits operation. Solid motivations are compulsory to assure Rural Credit Cooperatives to amendment the mode of operating, but growing them attracted is mostly challenging given their present situations. Crises in the financial division have convinced People's Bank of China to increase control under which Rural Credit Cooperatives and further financial institution operation. Rural Credit Cooperatives needs to be offered with fund at attractive charges and conditions associated with those of new source of refinancing but can not decrease the motivation to activate deposit. The enslavement to donors funding should be prevented. It can be applicable that the Rural Credit Cooperatives collect funding at a ratio not beyond the ratio at which People's Bank of china loans to Rural Credit Cooperatives.

The intermediation of financial at the native levels expands then complementarities between the linked elements of the native financial infrastructures were increased. The demand for the several civil-society institutes and other performers were cooperated with Rural Credit Cooperatives with delivering credits in rural regions. But, the current financial situations of the Rural Credit Cooperatives does not permit them to capitalize in increasing outreach skill and ability building, so Rural Credit Cooperatives needs funding for particular aims.

## Difficulties in Rural Credit Cooperatives

RCCs facing some difficulties in China are rural finance. The following are various difficulties;

### Operational Losses

RCC of China has operational losses of 23 percent of the entire numbers of credits cooperative during 1987. After establishment of China's markets of economy in 1993, RCCs boosted their losses and it touched 19 percent to 52.19percent. Again during 1997, losses happened about to 64.23 percent in Rural Credit Cooperatives.

## Bad Credits Administration

Bad Credits Administrations are also the drawback of Rural Credit Cooperatives. The trust violation elements and the continuance of the credits order ultimately must depends on the restriction forces of laws, which are same to the constructions of the Chinese rural credits structure.

## Bad Situations of Asset Quality

The third problematic situation in RCCs is that the bad situation of assets quality and it is not well. Rural credit Cooperatives has bad loans rates of whole loans during 1999.

## Lack of modern Services

RCCs has continually engage with the loans business and usual deposits saving, out of tables and transitional business is blank because to shortage in modern service and credits instrument. During 1999, Rural Credit Cooperatives loan interests incomes of 642 percent by the income of 35.7 percent and in-between incomes are accounted for only 0.1 percent.

**Inadequate Existing Rural Credits Guarantee Form**

Credits connection and the country credits connection, large number of farmers assurance form must be calculated and tested by several sectors which check and remove the risks of rural credits to some levels. But with the expansion of the supervision, credits need, and capital, this guarantee form might not fulfill the rural financing need and stop the credits risks clearly.

## CONCLUSION

Rural Credit Cooperatives in China is indeed the key head and due to his strong and important role the financial system directly depends on it for rural families investments.To develop the rural areas it running different Agriculture projects for the betterment of Rural Agriculture sector. Rural Credits Cooperatives are the greatest as a countless of region founded credits organizations with variable financial functioning and assets feature. Amendments have also converted several credits associations into Rural Commercial banks and Rural Cooperative Banks. Though RCCs have some difficulties and matters to supervise, but Rural Credit Cooperatives are mostly profit-oriented organizations, they are trying their best encouraging the loan to rural families and Agriculture nonpayers. Rural Credit Cooperatives are very beneficial for the development of rural regions in different forms but specially to boost up the Agriculture sector in China.

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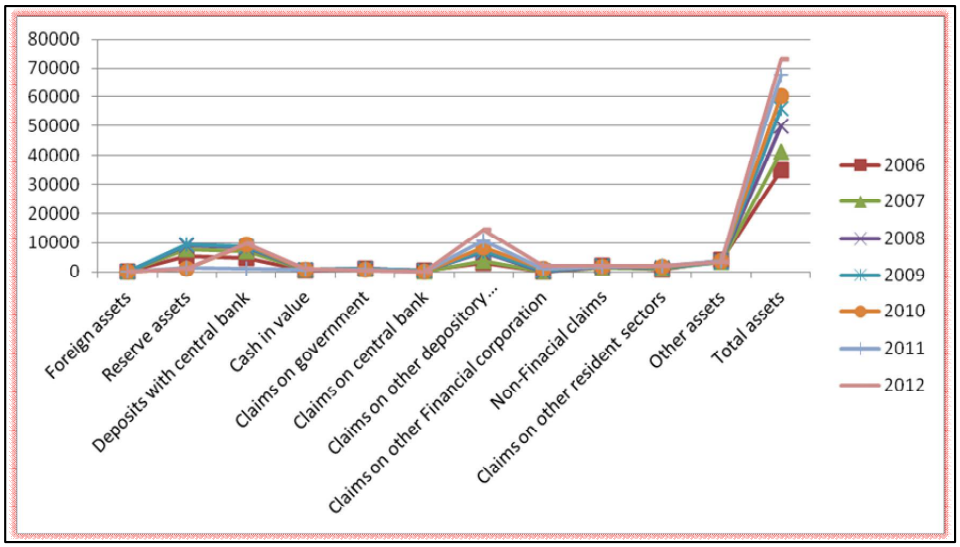
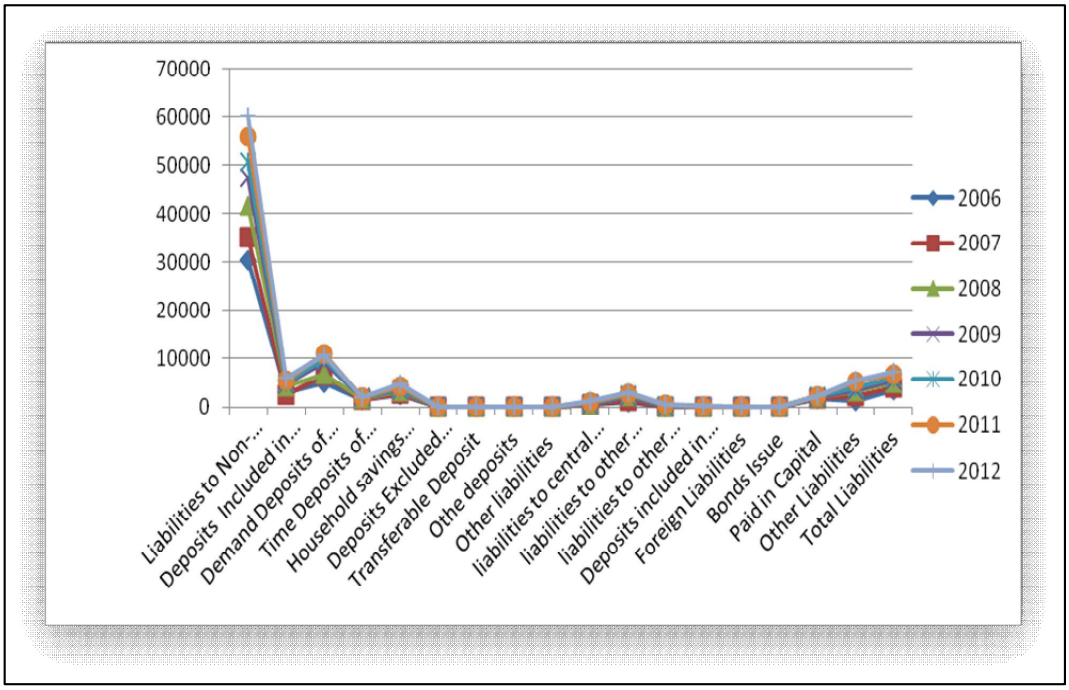



**Fig. 1 Rural Credit Cooperatives Assets from 2006 to 2012 at the end of December**



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**CONFERENCE REPORT**

# National Symposium on "Sustainable Disease Management: Approaches and Applications"

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## REPORT

In the backdrop of the majestic Himalayas the National Symposium on "**Sustainable disease management: Approaches and applications**" as held successfully during 21st to 23rd December 2017 at Department of Plant Pathology, College of Agriculture, G B Pant University of Agriculture & Technology, Pantnagar, Udham Singh Nagar, Uttarakhand. Approximately 248 delegates from more than 12 states (Delhi, J&K, Himachal Pradesh, Uttarakhand, Uttar Pradesh, Bihar, Meghalaya, Orissa, Madhya Pradesh, Gujarat, Maharastra, Rajasthan, Kerala, Nagaland) participated in the symposium, despite the remoteness of the Pantnagar. The symposium was inaugurated by Dr. U S Singh, Director, IRRI South Asia Regional Centre (Varanasi)/ Principal Scientist, Division of Plant Breeding, South Asia Regional Project Coordinator, Stress Tolerant Rice Program, International Rice Research Institute (IRRI), IRRI-India Office (IIO), New Delhi 110012, India and Hon'ble Vice Chancellor of the Gobind Ballabh Pant University of Agriculture and Technology (GBPUAT, Pantnagar. The inaugural session started with the university song and invocation of the deities by lighting the lamp on the stage.

The proceedings were initiated by Dr. K. Vishunavat, Co-Chairman of the National Symposium and Head of Department of Plant Pathology, College of Agriculture, Pantnagar by delivering the Welcome Address. Welcoming the delegates and dignitaries participating from across the country, she gave a brief account of the genesis and current activities at the Department of Plant Pathology of College of Agriculture, GBPUAT. She told the gathering that the department has many notable achievements to its credit. Deartment and stalwarts like Dr. R S Singh, Dr. A N Mukhopadhyay, Dr. R K Tripathi, Dr. Amerika Singh, Dr. S J Kolte etc. and further expanded by many dedicated faculty members who lead the department as recognized leader in the country. She highlighted the fact that the scientists of the department were the first to detect the Khara and apple scab disease in the Uttarakhand state and standardized its management strategies including disease forecasting system periodically broadcast through All India Radio and Television programmes. He placed on record that apart from developing several Integrated Pest Management (IPM) techniques for major hilly and plain crops the Plant Pathology department has a major role in the development of several bio-villages. The department attains excellence in teaching and research in Plant Pathology and the services rendered to the farming community. Out of the 11 Departments of the College of Agriculture at the

University, the department received an "Excellence award for the year 2016". At the end of the welcome address, formal welcome of the dignitaries on dais was done by presenting them with bouquets and shawls.


On the occasion of the inaugural session Souvenir and Abstract book of the Symposium were released on dais by the Hon'ble Vice Chancellor. The Presidential address was then delivered by Dr. K.P. Singh, Zonal President of Indian Phytopathological Society-Mid-east zone. He welcomed all the delegates and the dignitaries to the national symposium and detailed the status of apple diseases in Uttarakhand and the strategies to manage them. He emphasized on the role of disease forecasting and integrated pest management strategies to curb the losses in apple production in Uttarakhand. He said that spraying efficiency can be improved with reduced fungicides use with reliable scab warnings are helpful as during recent years, warning systems for plant protection have increased in importance especially in the context of integrated plant protection. Apple growers use Mills-based predictive systems to determine the risks of primary infection by ascospores. Several workers suggested to changes in the Mills table and developed a new equation, based on the published data, to describe the minimum infection times for conidia. Weather data were recorded using microprocessor-based orchard environment monitor, µMETOS scab warning device and RSS – 412 apple scab predictor. The data provided hourly records of precipitation, temperature, relative humidity, and leaf wetness. At present, the PAD technique is the most frequently used method in Western countries for best forecast of ascospore production in an orchard in order to predict the danger of epidemic in early spring. The concept of potential ascospore dose (PAD) developed by Gadoury and MacHardy have worked with the calculation of inoculum levels of *V. inaequalis*. PAD gave the estimated production of ascospores per $m^2$ of orchard floor and the estimated number of ascospores produced per fertile lesion. Dr. Singh told the audience that based on the random surveys for apple scab disease in Uttarakhand Himalayas it was revealed that there was an increasing trend in disease in Gangotri fruit valley of Uttarakhand. However, in general, the disease intensity remained low. The Chamba-Mussoorie, Almora, Pauri, Tehri, Nainital and Bageshwar fruit belts were apparently free of scab.

In the context of the theme of the National Symposium Dr. D S Pandey, Dean, College of Agriculture, cautioned that the hill and plain region of the state being an extremely fragile and ecologically sensitive region and since most of the water resources originate in the Himalayas, extreme care has to be taken before application of agro-chemicals and pesticides as these may pollute the entire water system of the country. He further added that the green revolution technology with it's over-reliance on high resource use has little relevance for the Himalayan region. The adoption of intensive crop production practices with faulty application of agro-chemicals has resulted in the development of new and emerging plant disease problems and new pathogenic races. The impending climatic change may also bring about sea-change in the disease development pattern and pathogenic diversity compelling the plant pathologists to come out with appropriate management strategies to minimize crop failures in future. He also emphasized that Uttarakhand state being declared as an Organic State, use of harmful agro-chemicals and pesticides should be minimized so as to add value to the organically produced commodities for export purposes. The Himalayan region is rich in bio-diversity of various medicinal and aromatic plants including many wild aromatic species. Such locally

available plants should be explored for their potential use as botanical pesticides for effective management of diseases and pests of the region, requiring low cost, eco- friendly agro-technologies suitable for the largely resource poor mountain farmers. In the end, he hoped that the deliberations in this national symposium will bring out meaningful crop management strategies and appropriate policy issues that may be useful not only to the scientists and planners but also to the farming community of the country as a whole and the Himalayan region in particular.

Indian Phytopathological Society President remarks were presented by the Dr **B. N. Chakraborty,** Department of Botany, University of North Bengal, Siliguri 734013, West Bengal. He stressed that more emphasis has to be given on the study of plant diseases and their management because of the direct losses they cause to the grower. The losses can occur in the field or in the store and at any time between sowing and consumption of the harvest. Standing crop in the field is attacked by disease and plants start drying or their capacity to yield satisfactorily gets reduced. Thus, the farmer gets only a portion of the estimated yield. The resources spent on field preparation, seeds, fertilizers and irrigation are also lost. The harvest is transported to its destination and fruits and vegetables may start rotting during this period. Similarly, there is loss during storage as well. He called upon the participants of the national symposium to lay emphasis on utilizing the modern tools of science to monitor and manage the diseases. The use of new safer plant protection molecules, marker assisted selection for host resistance, transgenic crops and stain improvement in biocontrol agents are some of the areas which need to be discussed thoroughly. Disease management in organic farming and integrated disease management in the perspective of sustainable crop production and environmental safety also need to be addressed. He hoped that the deliberations in this symposium and exchange of ideas on diverse issues relating to microbial diversity and plant disease management for sustainable crop production would ensure greater food availability, nutrition and environmental security in the years to come.

Special Invitee Dr. Dinesh Singh, IPS Secretary, Department of Plant Pathology, Indian Agriculture Research Institute, New Delhi, threw light on the role of Indian Phytopathological Society in the development and dissemination of scientific know how in the country and all around the world. He informed the audience about various awards and fellowships given by the Society. Inaugural address was delivered by the Dr. A P Sharma, Hon'ble Vice Chancellor, GBPUAT, Pantnagar. He began his address with introducing the host University to the delegates participating from different parts of the country. He told that Pantnagar University was the first Agricultural University of the country, which was established on land grant pattern of United States of America Universities and continues to be the role model for a number of State Agricultural Universities established afterwards. Nobel Laureate Dr. Norman E. Borlaug rightly called this university as `*Harbinger of Green Revolution in India*'. He also informed that the G.B. Pant University of Agriculture and Technology was established on the basis of MOU between the Government of India and Government of Uttar Pradesh in 1972 (formally inaugurated in 1976) has responsibility of generating and disseminating technologies in agriculture, forestry and allied field for the sustainable development of the Himalayan region of Uttarakhand.

Inaugural address was delivered by the Chief Guest of the session Dr. Uma Shankar Singh, *Director*, IRRI South Asia Regional Centre (Varanasi), *South Asia Regional Project Coordinator*, Stress Tolerant Rice Program, International Rice Research Institute (IRRI), IRRI-India Office (IIO), NASC Complex, New Delhi 110012, India. He stressed the importance of plant diseases as they are the major constraints to production of high quality and high yielding crops and cause severe deleterious effects in natural environments. In the history of mankind, plant diseases have been highlighted with a number of important events. They have affected course of the human history several times. Plant diseases in particular have been of great concern to Indian agriculture. Although, country has made a great stride in field of plant disease management particularly in cereals, we are always faced with new problems and challenges. He said that the global estimates of loss indicate that 42% of the total global attainable production of eight major crops (rice, wheat maize cotton, potato, soybean, barley and coffee) is lost due to pests, of which 16% is due to insects, 13% due to diseases and 13% due to weeds which in monetary terms was estimated to be an annual loss of about $250 billion worldwide. If this huge loss could be prevented there should be no food shortage for years to come. The effect of this loss is not only economic. Many political and social problems originate from shortage of food.

This increased demand for food will have to be met by developing high yielding crop varieties, ensured fertilizers and irrigation with provision for disease management. He acknowledged that the science of Plant Pathology has an important role in the future success of programmes and policies designed to increase and sustain food production. He appreciated the organizers and scientists involved in plant protection for timely recognition of this challenge by addressing the issue of advances made in microbial diversity and plant disease management for sustainable crop production, through the national symposium. He hoped that the National Symposium would provide an opportunity for exchange of important information data base on this vital subject between the states of India and the deliberations would help the application of cutting edge science not only in the improvement of strategies for plant disease management thereby ensuring nutritional security and sustainable agriculture in the country.

In inaugural keynote address, Dr. Uma Shankar Singh, Director, IRRI South Asia Regional Centre (Varanasi), said that the mission of plant pathology to develop methods for plant disease control as an applied science, and to contribute to plant protection as a basic science through elucidation of principles underlying the expression of pathogenicity of microbes and resistance of host plants. He elaborated his study on enhancing and stabilizing rice productivity under changing climate in collaboration of IRRI-India and took the reference of our ancient knowledge on the use of Gene(s)/ QTLs and their markers identified the tolerance to abiotic stresses, i.e. submergence (*SUB*1), drought (DTYs), salt (*SALTOL*), anaerobic germination, stagnant flood (SF), low P and resistance to biotic stresses (BLB, Blast). He also established several Sub1 (Flood tolerant) varieties released in S. Asia (IR05F102 (Swarna), IR07F102 (IR64) and CR1009), drought tolerant varieties (Sahbhagi dhan (2010), DRR dhan 42 (2014), DRR dhan 43 (2014) and DRR dhan 44 (2016), salt tolerant varieties (India: CR dhan 405 (2013), CR Dhan 406 (2013), Gozaba (WB) (2014), CSR43 (2013), DRR dhan 39 (2014), Binadhan 8 (2015), Binadhan 10 (2015) and CSR 46 (2016). He informed the audience that stress tolerant rice varieties have potential to usher Green Revolution in South Asia by enhancing and stabilizing rice productivity and production in stress prone areas. The technical sessions of NSSDM were covered following 5 sections and two award sessions comprising over 250 delegates from 12 states. Several oral presentations with 176 poster contributions by the prospective young scientist awardees were also made. Every day each session covered plenary lectures, symposia, general poster sessions, special lectures and debates.

## Host Plant Resistance

The session was chaired by **Dr. K P Singh Ex-Professor Plant Pathology/ Director Extension,** Chairman and Dr. U B Singh, NBAIM, supported as Co-Chairman**.** The lead lecture was delivered by Dr. O K Sinha, Ex-Director, IISR, Lucknow, on "Strategies of plant disease management under organic farming conditions". He emphasized on the challenges faced in the course of managing crop diseases. He pointed out that endophytic microorganisms occurring naturally within the plant system can be manipulated for sustainable protection of crop Rhizosphere is the main source of endophytes. The importance and success of bacterial entophytes for the management of crop disease has been discussed. Dr. Sinha focused on traditional method of disease management like changing planting date, vector control, biofumigation, plastic mulch, use of biocontrol agent against pathogenic fungi, bacteria and nematode etc. The second lead lecture was delivered by Dr. Dinesh Singh, IARI, New Delhi on "Diversity, Diagnosis and Management of *Xanthomonas* causing bacterial leaf spot of pepper and tomato in India". He emphasized complete diagnosis of *Xantomonas* spp. causing bacterial leaf spot in pepper and tomato and emphasized that seed is the primary source of inoculums and in soil it can survive for two weeks to 18 months. Plant debris and wild host plants may be the important source of pathogens and act as reservoir. Low doses of streptomycin along with copper sulphate may be used for disease management. Dr. Vaibhav Singh, IARI, New Delhi, elaborated his study on Wheat blast: A new threat in south Asia and our preparedness. He reported that in 2016-17, eight district of Bangladesh were affected by blast fungus in wheat. This disease is disseminated by wheat seed and secondary infection is by air borne conidia. Seed treatment is effective against this disease. He suggested gene pyramiding for rust, blast and blight pathogen in wheat may be initiated to develop resistance cultivars. Dr. Manjunath, IIPR, Kanpur, dealt with potent resistant donors and phylloplane microbes against *Ascochyta rabiei* infecting chickpea. These microbes were characterized using 16S rDNA and ITS sequence analyses for bacteria and fungi. Dr. Faheem Ahamad, AMU,

Aligarh explained the relative tolerance/resistance in rice germplasm against rice root knot nematode, *Meloidogyne graminicola.* Dr. U. B. Singh, ICAR-NBAIM spoke on the microbial modulation of physiological and biochemical responses and systemic defense mechanism against *Rhizoctonia solani* Kuhn causing banded leaf and sheath blight in maize. This lecture focused on novel insights into the underlying mechanisms of priming by beneficial microbes and underscores their capacity to trigger defense responses under biotic stress.

## Prof. M J Narasimhan Academic Merit Award

M J Narasimhan Academic Merit Award zonal contest was held during the last session of the first day. Seven contestants participated in the competition with the eight expert panels, Drs. U S Singh, Director, IRRI-SARC, O K Sinha, Ex-Director, IISR, Rakesh Pandey, HOD Nematology & Pl Prot., CIMAP, Ramji Singh, HOD, SBPUAT, Sunil Kumar, NBIM, Usha Chakraborti, UNWB, B N Chakraborti, IPS President, UNWB and Vinita Singh, BHU, Varanasi. Mr. Devanshu Dev from GBPUAT, presented primary study on studies on anthracnose of pomegranate caused by *Colletotrichum gloeosporioides,* a important diseases causing large revenue losses in term of exchange in tropical and sub-tropical regions. Second paper was presented by Miss Kalpana Gairola, GBPUAT, explained about early detection and management of white rust disease (*Albugo candida*) in Rapeseed mustard. In her presentation she made a point that early detection of *A. candida* was done by PCR-based assay and light microscopy was more helpful for evaluation of some new fungicides for the effective management of the disease. Meenakshi Dwivedi, GBPUAT, Pantnagar, gave the occurrence of crop specificity in growth promoting effect of native *Trichoderma* species isolated from rhizosphere and rhizoplane of three economically important crops viz. rice, wheat and chickpea. However, native *Trichoderma* isolates had pronounced growth promoting effect in their respective crop as compared to other crops. So far, crop specificity in the growth promoting potential of native Trichoderma spp. Has been least exploited. Manish Tripathi, Kumaun University, Almora, presented his new reports on the evaluation of secondary metabolites isolated from endolichenic fungi against crop infecting bacteria. In his research he studied the use of endolichenic fungi may not only help to reduce the dependency on chemically synthesized antimicrobials but will also overcome the loss of lichen diversity for their bactericidal use. P. K. Sajeesh, GBPUAT, Pantnagar, emphasized the combination of copper-chitosan-trichoderma mediated defense induction in potato against late blight pathogen. These combination involves copper (fungicide), chitosan (plant strengthner) and trichoderma (a well known biocontrol agent), suggesting that the components of the triple combination will have different mechanisms for disease management. He reported that due to multiple sites of action this combination may provide a durable and long lasting management strategy for late blight pathogen. U. B. Singh, NBAIM, spoke on the conjunctive impact of root associated bioagents, methyl jasmonate and salicylic acid induced resistance through enhanced phenylpropanoid activities and plant growth promotion in wheat. This study apparently provides the evidence of biochemical cross-talk and physiological responses in wheat following MeJA, SA and bioagent treatment during the bio-trophic infection. The talk by S. K. Modak (GBPUAT, Pantnagar) gave an overview of recent progress of biofumigant for the management of damping off in tomato. He highlighted the significance of biofumigation with Kranti and seed biopriming with PBAT-3 could be a handy option to manage damping off disease of tomato in nursery.

The presentations were evaluated by a panel of judges/experts. **Mr. P. K. Sajeesh and S. K. Modak were adjudged as the better one of the other and stood first and second in Mid-East Zone.** As both the participants secured more than 47 per centage of marks, both of them have been recommended for M J Narasimhan National Level competition. At the end of the day cultural programmes were presented by the graduate and post- graduate students of College of Agriculture and the students of Plant Pathology department. The event included more than 10 programmes by the students. In the end some of the delegates also performed. Delegates were also invited on the dais to give their feedback on the national symposium and the cultural programmes. Delegates were highly appreciative about all the events and proceedings of the national symposium.

**PGPR, Bio-inoculants, Nano-technology and Bio-technology approaches**

The session was chaired by **Dr. Rakesh Pandey, Head Nematology and Plant Protection**, CIMAP Lucknow and **Dr. Vaibhav Singh, Division of Plant Pathology, IARI, New Delhi acted as Co-chairman** of the session. Dr. B N Chakraborti, HOD Botany, UNB, WB, presented the first lead lecture on potential uses of beneficial microorganism in crop protection and entrepreneurship development. During the deliberation he emphasized on the exploitation of soil microbiota for better management of plant diseases. *Trichoderma* can be utilized for rapid composting of tea waste can be used. 410 biopesticides are being produced in India out of which 130 fall under private companies. He appraised the audience about the usage of beneficial microgranism and non chemical methods for the disease management of most of the crops. He also suggested that disease suppression and health improvement in these plants can lead to development of bioformulations using such potential microbial resources. The second invited lecture of the session was presented by Dr. Ramji Singh, HOD Plant Pathology, SVPUAT on potential uses of beneficial microorganism in crop protection and entrepreneurship development. He talked in detail about the status and hazards of disease management through the use of chemical pesticides, while emphasizing the need for safer and effective bio-pesticides based management strategies. He stressed upon the need for educating the farmers for the usefulness, preparation and application of bio-pesticides. He called the attention to the need for quality assurance and effective production and delivery mechanism required for making bio pesticides available to the farmers. He appealed the plant pathologists to devise effective management strategies against major plant disease through bio-pesticides. He gave economically viable methods to use the de-oiled cakes of TBOs viz., Neem, Jatropha, Mahua and Karanja for mass multiplication and longevity of *T. harzianum* and *P. flourescens*. He also provided information on combined application of both bioagents grown on any of these cakes to the sterilized soil resulted in three time increase in root length, 1.5 times increase in shoot length and five time increase in fruit yield of tomato crop. The talk by Faheem Ahmad, AMU, Aligarh, gave an overview of proteomic approach to identify *Meloidogyne incognita* and nematicidal potential of oil cake. He proved that oil cake were significantly killing the juveniles ($J_2$) and also control the penetration in tomato roots. Ahmad also talked the fast and reliable technique to identify Meloidogyne species and give opportunity to use ecofriendly control approach to minimize the crop loss. Dr. S. K. Goswami, ICAR-NBAIM, Mau talked about candidate pathogenicity genes based novel microsatellite markers from *Rhizoctonia solani*. Dr. P. L. Kashyap, ICAR-IIWBR, Karnal, referred to the microsatellite based diagnostic assay for rapid and sensitive detection of brown rust of wheat. He also pointed out that the quantitative real-time PCR method proved to be rapid, sensitive, specific, cost-effective and reliable for the identification and quantification of *Pt* in plant hosts. Sunaina Bist, ICAR-IARI, shared her experiences on rhamnolipids biosurfactant against post flowering stalk rot of maize. So many scientists presented their research work on poster session. The posters were evaluated by senior scientists and were ranked.

**APS Student Award**

APS Student Award zonal contest was held during the forenoon session of the second day. Three contestants participated in the competition. Mr. Devanshu Dev, GBPUAT, Pantnagar presented on pathogenic characterization of *Albugo candida* isolates the cause of white rust disease in Rapeseed mustard. He explained the re-designation of *A. candida* races on the basis of infectivity of different isolates on different genotype of different *Brassica* species. He also provided information among the isolates Bangalore, Simour and Karnal were found to be more virulent and showed different disease reaction with different genotypes. The lecture by Gagan Kumar, I.A.Sc., BHU, Varanasi, dealt with the identification and characterization of biotic (*Fusarium udam*) and abiotic (NaCl) stress responsive WRKY transcription factors in Pigeonpea. He emphasized the transcriptional profiles of these 35 CeWRKY genes in root tissue after 24 h of biotic and abiotic stresses inoculation were systematically investigated using qRT-PCR analysis in wilt resistant and susceptible cultivars of pigeonpea in presence of *Pseudomonas fluorescence* strain OKC. The presentation highlighted new study to identify the organization and transcriptional profiles of CcWRKY genes, which not only facilitates the functional analysis of the CcWRKY genes, and also lays the foundation to reveal the molecular mechanism of stress tolerance in this important crop. Ambika Rautela, GBPUAT, Pantnagar, spoke on

interaction between chitosen and copper for effective and eco-friendly management of plant diseases. She also gave the use of Chitosan + CuOH (TG) complex could effectively be used for management of various plant diseases since copper is a broad spectrum protectant fungicide. Chitosan on integration with copper ions may prolong protection on plant surface by chelating copper ions and thus could prevent washing off of the copper. While also the anti-microbial and plant defense inducing properties of chitosan could lower the amount of copper dose needed for disease management and thus would result into ecofriendly management of plant diseases.

The presentations were evaluated by a panel of four judges/experts, Drs. Basant Ram, Ex-VC RAU Pusa & NDUAT, Faizabad, K P Singh, Ex-Director, Extension Education, GBPUAT, Pantnagar, Usha Chakraborti, Professor & HOD, UNWB, Siligudi, K. A. Batt, SUKAST, Kashmir. **Mis. Ambika Rautela and Devanshu Dev, GBPUAT, Pantnagar** were adjudged as the better one of the other and stood first and second in Mid-East Zone. As both the participants secured more than 45 percentage of marks and recommended for APS student travel award competition.

## Disease management in medicinal and horticultural crops

The third technical session was chaired by Dr. (Mrs.) K. Vishunavat, HOD Plant Pathology, GBPUAT and Dr. B. K. Sharma, Professor, BHU, Varanasi, acted as Co-chairman of the session. The lead lecture was presented Dr. Basant Ram, Former VC of RAU & NDUAT, on integrated management of wheat rust. In his elaborate lecture he emphasized that in order to meet the growing demand of its population. India has to increase the production of wheat. However, many a diseases are threatening this goal. He informed the audience that in wheat stem, leaf and stripe rust's were main causes of yield loss though introduction of resistant varieties were quite helpful in reducing the loss. But the challenges are not yet over. Development of new races like Sr-9 of 'Stripe rust' pathogen originated else in other countries reached India and Ug 99 of 'Stem rust' originated in Uganda has reached up to Iran in 2007. It is quite likely that may reach India any time. Other diseases like Karnal bunt, loose smut etc. also affects the wheat production in the country. He pointed out that integrated disease management using resistant/high yielding varieties, cultural practices and effective biological control measures have to be developed for controlling wheat diseases, in addition to the ongoing efforts for identification of rust resistant genes and their deployment for increasing productivity. He also told that the breakdown of Y9 resistance in mega wheat variety PBW 343 has become a matter of concern. There has been frequent breakdown of rust resistance in other popular wheat varieties in India in recent years. In another keynote lecture, Dr Rakesh Pandey, DAAD Fellow, FNAAS, CSIR-CIMAP, Lucknow provided an overview of the role of root knot nematode in disease complexes with fungi and their management in medicinal and aromatic plants and improve production of plant secondary metabolites. He also talked about the useful microbes like PGPR, mutualistic fungi, and other nematode antagonists disfavor the multiplication and development of root-knot nematode population in soil, enhancing the growth/ yield as well as plant secondary metabolites production in MAPs. The presentations also lead to very good interaction and it was emphasized that the attention has to be given for the profitable marketing of the medicine plants. It was also stressed that timely monitoring for probable diseases in these medicinal plants also should be taken up. The invited lecture was presented by Dr. J P Jaiswal, GBPUAT, Pantnagar, on marker assisted transfer of major and APR genes for durable rust resistance in bread wheat (*Triticum aestivum* L.), highlighting the importance of rust in wheat. He stressed upon the usage of recommended varieties to be cultivated as per the time schedule to reduce the havoc caused by the disease in different parts of the country. He advocated the pyramiding of these race non-specific genes is expected to confer durable rust resistance in wheat. He also pointed out that near isogenic lines (NILs) thus developed in the next three crop seasons will not only be ready for the release as a variety possessing durable rust resistance but also these could be used as mapping population for the detection of QTLs for rust resistance. Dr. Sunil Kumar, ICAR-NBAIM, Mau, while emphasizing the AIMDb: database for agriculturally important microorganisms (AIMs). He also suggested that AIMDb will be helpful for researchers working on agriculturally important microorganism as they can have all the information at a single plateform. Dr. Shilpi Rawat, GBPUAT, Pantnagar, explained about *Ganoderma lucidum*, is one of the most popular medicinal mushrooms. She also talked about the most of the spices have favoured MEA media with a temperature of 20-25°C and pH of 5-6 for optimum mycelia growth.

The role of fungicides, bioagents and botanicals against anthracnose of pomegranate was shown by the work of Devanshu Dev, UAS, GKVK, Bangaluru, Karnataka. So many scientists presented their research work on poster session. The posters were evaluated by senior scientists and were ranked.

## Biotic and abiotic stress management

The fourth technical session was on biotic and abiotic stress management strategies under changing climate scenario, disease dynamics, epidemiology and disease forecasting which was chaired by Dr. Usha Chakraborti, Professor, North Bengal University, and Dr. Ramji Singh, HOD Plant Pathology, SVBPUAT, Meerut acted as Co-chairman of the session. The lead lecture was presented by Dr. D. P. Singh, Head Plant Protection, ICAR-IIWBR, Karnal, Haryana on keeping wheat crop health sound over decades in India: A success story. He also gave an account of the transboundary diseases like stem rust pathotype Ug 99, new pathotypes of stripe rust and news diseases like wheat blast in Bangladesh, posed threats to wheat production in India. In his detailed talk he emphasized that production and productivity has to be increased for which the pathologist and breeders should work together. The breeder seed is produced in sufficient quantity and given to seed producing agencies. On an average 40 resistant genotypes with all passport data on disease resistance are shared with breaders for use in breading programme since 1999-2000 till date. The presentation highlighted to keep it going, we must work to collectively to cope up the effect of climate change, changing cropping system, agronomic practices, conservation agriculture and new pathotypes of rusts and new threats of trans boundary diseases like wheat blast which is present in Bangladesh since 2016. Dr. B. K. Sarma, I.A.Sc, B.H.U., Varanasi, spoke on the host responses to fungal biotroph pathogen challenge: a molecular insight into *Erysiphe pisi*-pea interaction. This lecture focused particularly on the signal transduction was possibly mediated through JA in pea under the stimulus of the microbes as evident from the activities of *LOX1* and *CO11* genes and the cumulative effect of the co-inoculated microbes had a suppressive effect on *E. pisi* conidial development on pea leaves. Dr. Ankita Jha, ICAR-IIWBR, Karnal, Haryana, dealt with current status of yellow rust in wheat using remote sensing (NDVI) based spectrometeorological model was developed using SPSS software in order to estimate/ foretell the possibility of yellow rust and its possible impacts on wheat. Dr. K. A. Bhatt, SKUAST, Jammu & Kashmir, focused on management of European mistletoe (*Viscum album*) infesting Walnut (*Juglans regia*) in Kashmir valley. Dr. Vineeta Singh, BHU, Varanasi, referred to the population structure of *Rhizoctonia solani* AG1IA causing sheath blight of Rice. Her presentation included the molecular variance (AMOVA) based on $\phi$PT (fixation indices) values indicated that most of the genetic diversity occurred within populations, while the variability among populations and among regions contributed 25 per cent and 15 per cent. R. N. Yadav, I.A.Sc., B.H.U., Varanasi, presented scientific evidence on the role of *Trichoderma harzianum* mediated alleviation of drought stress in *Oryza sativa* L. DRR-44. He indicated the exciting possibilities of management of drought stress in plants by application of drought tolerant plant beneficial rhizospheric microorganisms. Large number of scientists presented their research work on poster session. The posters were evaluated by senior scientists and were categorized according to their presentation.

## Integrated management of Phytopathogens

The fifth technical session on integrated management of phytopathogens, plant clinics and extension plant pathology was chaired by Dr. J. Kumar, Prof. Plant Pathology, Ex-VC, GBPUAT & Dean, College of Agriculture, Pantnagar and Dr. Faheem Ahmad, SUKAST, Kashmir acted as Co-chairman of the session. The lead lecture was presented by Dr. K. P. Singh, Prof. Plant Pathology & Ex-Director, Extension, GBPUAT, Pantnagar on Soil suppresiveness-an ecological approach to plant disease management and several contributory papers consisted of several presentations covering diverse area. Several hot topics were discussed which consisted of a talk by Dr. Singh brought out the impressive progress made in the area of disease suppressive soil. He also talked about the recent advances in the area of microbial communities to induce a disease suppressive soil environment does possess potential as a tool in the management of soilborne plant diseases. These microorganisms probably resided in equilibrium with pathogens and plants before the intervention of agriculture. Dr. Nandani Shukla, GBPUAT, Pantnagar, told that tolerant *Trichoderma* strain and copper synergy for efficient management of late blight in potato. She also pointed out that

copper tolerant *Trichoderma* with lower doses Cu- based fungicides could be employed for developing combination products for safe and effective management of late blight disease. Evaluation of three *Trichoderma* spp. From three districts with local strain was shown by the work of Dr. Shashi Tiwari, SHUATS, Naini, Allahabad. Her presentation included the all three spp. of *Trichoderma* was very effective against *Fusarium oxysporum* f sp. *lycopersici* under in vitro and field both condition. Dr. Mohit Kumar, LPU, Punjab shared his experiences on success and sustainability of IDM strategy depends on involvement in generating local specific techniques and solutions for their particular farming systems and integrating control components that are readily available to them. Success requires appropriate policies such as plant protection, private sector investment, trade and export, food safety, land use, education and awareness, and agriculture extension. His experience over the last few decades clearly showed that adoption and support for using participatory approaches help farmers improve their overall field management, including disease management, reducing cost and improving production efficacy. The last speaker of the session was Dr. Roopali Sharma, GBPUAT, Pantnagar, highlighted that large scale implementation of bio-intensive disease management technologies at farmers field. In her talk she pointed out that the capacity building and awareness creation amongst the farmers was done by demonstrating the method of application of bio-pesticides in crops, distribution of inputs, organizing trainings and gosthies, and regular visit at farmers' fields. She was able to find the average yield of rice using bio-intensive technologies was 60q/ha as compared to conventional farmers practices (53q/ha) with an yield increase of 13.5 per cent, the yield obtained in pea was 72q/ha as compared to farmers practices (58q/ha) with an yield increase of 23.5 per cent, in tomato it was 209q/ha as compared to farmers practices (167q/ha) with an yield increase of 24.7 per cent. The presentation was followed by a discussion on different aspects including the shelf life of this compound which was explained as very long, the mode of action of these antagonists on the plant, its significance as an eco-friendly approach for the management of fungal infection and on host specificity of the compound. Posters presentation related on the above themes of the symposium were displayed in the Agriculture Pavilion of the college. In total 17 posters were displayed in this sessions.

On the last day of the symposium plenary session was held. The session was chaired by the organizing chairman of the symposium **Dr. Ravi Khetarpal, Executive Secretary, Asia-Pacific Association of Agricutural Research Institutions (APAARI),** Bangkok 10100, Thailand. The eminent plant pathologist Dr. Basant Ram, Ex-VC of RAU, Pusa and NDUAT, Kumarganj, Faizabad was the guest of honour of the session. The session was initiated by the university song by the students of the college. Dr. A. K Tiwari, Zonal Councilor of the National Symposium formally welcomed the dignitaries on the dais by presenting flower bouquets to them. The dais was also decorated with the presence of two eminent plant pathologists Dr. J. Kumar, Ex-VC of GBPUAT and Dean, College of Agriculture, Dr. K. P. Singh, Ex-Director Extension, GBPUAT, Pantnagar and Dr. J. P. Mishra, Ex-ADG, ICAR.

Co-Convener Dr. R. P. Singh, presented the report of the general body meeting of the members of Mid-east zone chapter of the Indian Phytopathological society. The major emphasis of the decisions was laid on the issue of high fee structure of the IPS sponsored International Symposium, timely supply of the journal published by the society, *Indian Phytopathology,* and to bring more transparency in the election of the office bearers of the national executive body. He also presented the recommendations emanating from the proceedings and deliberations of the National Symposium. He put forward the summarized the recommendations on the basis of the observations made by the Chairpersons and Co-chairpersons of respective sessions.

## Recommendations

1. It was recommended that more emphasis has to be given in microbial exploration, exploitation and conservation for their future use in eco-friendly disease management.
2. Usages of safer and non chemical method to be explored for the effective disease management, both in field and post harvest disease that should include biocontrol agents, botanicals and use of safer and less toxic new plant protection molecules.

3. Bio-pesticide formulations should be carefully studied prior to recommending them to farmers for the disease management. Thorough studies and multi-locational field efficacy trials should be carried out and the product/formulation should be registered.
4. In view of the chronic dependency of the farmers on the chemical pesticides, it was recommended that integrated disease management modules should be worked out on a crop to crop basis for effective disease management.
5. It was appealed that novel non-chemical interventions such as cultural practices, should be given priority as they are more effective and due to their cost effectiveness and farmer's involvement, are easily adopted by the farmers.
6. In cases where alternative management options are not available fungicides should be applied judiciously and on need based only.
7. There was an exclusive stress on developing mutants of effective bioagents which could be compatible to other chemicals, tolerant to adverse climatic conditions like high temperature.
8. It was suggested that the senior highly experienced researchers should guide the juniors in research planning and execution.
9. Need for innovative research by the young scientists of the nation rather the copying the works carried out in other nations/ Indian laboratories, was also emphasized.
10. It was suggested that the innovative/ good quality research article to be submit to the journal Indian Phytopathology for publication through Springer

Organizing chairman of the symposium Dr. J. Kumar, Dean of the host college of agriculture, Pantnagar, presented the Chairman's remarks. He took stock of all the proceedings of the symposium and appreciated the pains taken by the delegates coming from all over the nation, in preparation and presentation of both oral and poster presentation of the results of the scientific research carried out by them. He appreciated the organizing secretary and the co-convener for the immense efforts made by them for successfully organizing the symposium of this magnitude in a harbinger of Green Revolution, the G B Pant University of Agriculture and Technology, Pantnagar. He expressed his gratitude to all the students, scientific and non scientific staff of the college who rendered their services at some point or other in the successful organization of the national symposium. At the end of his remarks, he was felicitated by Dr. K. P. Singh, by presenting him with a shawl and memento.

Delegates Dr. Basant Ram and Dr. K. P. Singh, also presented their views on the present status of research in plant pathology in India and on the relevance of organization of National Symposium on Sustainable disease management: Approaches and applications. They also congratulated the organizers on the successful completion of the symposium. At the end of his remarks, he was felicitated by Dr. J. Kumar, by presenting him with a shawl and memento.Chief Guest of the session **Dr. Ravi Khetarpal**, then presented trophies to the delegates having the three best presentations in each poster session of the symposium. Dr. J. Kumar, Chairman of the plenary session, presented the certificates to the awardees. Dr. Khetarpal, Dr. Basant Ram, Dr. J. Kumar, Dr. K. P. Singh, and Dr. J. P. Mishra remembered the work of **Professors R. S. Singh (GBPUAT), U. P. Singh (BHU) and S. J Kolte (GBPUAT) and presented Lifetime Achievement Award for his exemplary contributions in the field of Plant Pathology. His contributions to the science of Plant Pathology will be remembered for ever.**

In the end of the session Chief guest Dr. Ravi Khetarpal made his observations on the organization of the symposium and the issues raised during the deliberations and discussions. He advised the senior scientists and researchers to guide their juniors in the quest of achieving new heights in the plant pathological research in India. Lastly he announced the end of the proceedings of the symposium and wish the delegates a happy journey back their home. Chairman of the session Dr. J. Kumar, felicitated the Chief Guest with a Memento and shawl. Organizing Secretary Dr. K. P. Singh proposed vote of thanks to the dignitaries, delegates, participants, volunteers, organizers, governmental and private sector sponsors.

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**RESEARCH ARTICLE**

# Monitoring Al-Hammar Marsh Topography and Climatic Applied Satellied Modis Imagery

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## ABSTRACT

The object of the presented study was to monitor the changes that had happened in topography (water, over grass or vegetation, shallow, terra, and soil, etc.) of Al-Hammar Marsh region, by applied remote sensing techniques. To fulfill this goal in search, applied satellite MODIS (RGB) 12-3-2009 image used in study. The supervise and unsupervised classification was be used to classify the satellite image, which is the methodology trace and detect the topology effects and expose, depended on the dataset of satellite image, which performed cell in grid of raster (contains pixel is smallest object in imagery).Moreover, use cell algorithms, called statistics cell, statistics zonal, and statistics neighbors. After then, applied reclassification (super classification) to merge and isolated some region, which is secondary considering features or hybrid such as building, hills, and salt region.Additionally, to achieve calculate the area of each class for features of topography, by application advanced analysis tools of information geographic systems (GIS) to obtain result easy comparison and monitoring Al Hammar Marsh contain with another future studies. These improvements are likely benefits for land-cover mapping and change detection applications**,** also, knowledge that are spectrally bands observer and detector features good application. Study the change of weather factors variables on earth surface lead us understanding ecology phenomena by change in the radiation values received to instrument sensors.

**Keywords:** satellite MODIS Imagery, statistical method, weather factors (temperature, Relative Humidity and precipitation), RS and GIS.

## INTRODUCTION

The many complimentary space borne land remote sensing systems that are now actively monitoring the Earth. The recent launch of Terra, with the Landsat-7 ETM+, ASTER, and MISR sensors, combined with Satellite MODIS observations, comprise a major US commitment to study potential changes in the Earth system at the beginning of the 21st century see [1], [2], [3], and [4].Image data of satellite MODIS raw is using to study the land cover changes, shown figure 1. Through overlaying and buffering analysis. in the GIS platform, With the development of Remote sensing techniques and satellites, satellite images become an important source of information which can be used for monitoring in an efficient and of low cost techniques. Visible, near-infrared, thermal and microwave spectral have been used in studying different environmental phenomena, such as pollution of water bodies, Soil erosion, Weather prediction, flood monitoring desertification and land management.

The Mesopotamian Marshlands were one of the world's greatest wetlands and the largest such ecosystem climatic in the Southwest Asia. This wetlands ecosystem has been fundamentally change because of upstream dam's construction starting in the late 1950s, and the extensive deliberately draining and drying activities that started in the mid 1980s. In the end of the 20th Century, the Mesopotamian Marshlands were almost regions vanished. Disappearance of such globally important ecosystem had significant impacts on the environmental equilibrium and the biodiversity on global scale, see [5].The problem that rose as the whole marshlands recreation management is concerned was the lack of regular, efficient, precise and cost effective data collection program that supply the information to the decision makers. The project of the recreation of Al-Hammar marsh imply many changing processes and varied parameters; waters start flowing to the previously drained marshes, extent of these reflow waters are continuously changing, development of fauna and flora species are closely related to the water depth and and its area coverage many human settlements are rebuild again. In general, the marshlands are in accelerating high rate of changing state, see [6].

These dramatic properly manage such dynamically active processes; a sustainable remote sensing data collection program is required. To select the adequate and most cost effective imaging system, it is required to determine its spatial, spectral and temporal aspects that capable of delineating the different marsh cover types and choosing the proper time for image acquisition that corresponds to the change stage of the each material cover class. These elements are the main tasks in the present research**,** which attempt to study the important change, have been happened on the AL-Hammar marsh for research analogous. These changes include contain chief features topography, classification technique (supervised and unsupervised) have been used to achieve this study by applied cell algorithms of dataset satellite images**.**

In this study we estimated the surface temperature using NASA model. The digital numbers were transformed into absolute radiance, Surface temperature of wide areas can be extracted under assumption that satellite sensor should have proximity to the black body.For Landsat Thermal infrared (TIR) remote sensing provides a unique method for obtaining LST (Land surface temperature) information at the regional and global scales since most of the energy detected by the sensor in this spectral region is directly emitted by the land surface, see [7].

### Study area

The AL-Hammar Marsh is a large complex of wetlands in Iraq, that are part of the Tigris-Euphrates river system, it is representing region election lies in upper top left point (610875.639 Left- 3449974.571Top) and down bottom rights (765403.931 Left- 3381335.374 Top) x,y-axis (Cartesian coordinate system), shown figure 2. Projection coordinate system is WGS1984_UTM_Zone_38N, measurement unite is meters (m). Its surrounding marshes and neighboring marshes and areas of temporary inundation comprise some 3,500 km2 of almost contiguous wetland habitat. The

survey area dimensions of satellite MODIS imagery raw is 848 km column and 770 km row, which chosen study area election from scene source, it is 606 km column and 282 km row. It bordered in the north by the River Euphrates, in the west by the Southern Desert and in the east by the Shatt Al Arab. Cover types, non urban for land use andland cover types include water bodies, green vegetation (usually non irrigated), shallow, and bare soil (saline soil).Generally, The Lake is shallow with a maximum depth of about 1.8 m at low-water levels in early winter and about 3.0 m at high-water levels in late spring.Large parts of the littoral zone dry out during periods of low water banks and islands appear in many places separates spread, see [5], and [6].

### Satellite MODIS Characteristics

The Moderate Resolution Imaging Spectroradiometer (MODIS) are 36-band spectroradiometer measuring visible and infrared radiation and obtaining data that are being using to derive products ranging from vary types of physics research. The first MODIS instrument was launching on board flight on the Terra satellite in December 1999 and Aqua, the second was launching flight on Aqua in May 2002, see table 1.

### The Instrument Characteristics are:

- Medium-resolution, multi-spectral, cross-track scanning radiometer.
- Measures physical properties of the atmosphere, and biological and physical properties of the oceans and land.
- 36 spectral bands 21 within 0.4-3.0 μm and 15 within 3.0-14.5 μm.
- Continuous global coverage every 1 to 2 days.
- Signal-to-noise ratios from 900 to 1300 for 1 km ocean color bands at 70° solar zenith angle.
- NEDT's typically < 0.05 K at 300K.
- Absolute irradiance accuracy of 5% for <3 μm and 1% for >3 μm.
- Daylight reflection and day/night emission spectral imaging.
- Polarization Sensitivity, 2% from 0.43 μm to 2.2 μm and ±45° scan.
- Swath, 2300 km at 110° (±55°) from 705 km altitude.
- Field of View (FOV), ±49.5°.
- Instrument Instantaneous FOV, 250 m (2 bands), 500 m (5 bands), 1000 m (29 bands).

The many data products derived from MODIS observations describe features of the land, oceans and the atmosphere that can be using for studies of processes and trends on local to global scales. As just noted, MODIS products are available from several sources (i,e. MODIS Level-1 and atmosphere products, Land products, Crysphere data products, Ocean color and sea surface temperature products), see table 2 and see [8].

## METHODOLOGY

### Climatology and Meteorology

The main objectives of the present work consist of three stage; first, define The work area from source image for scene (Al Hammar Marsh location south east of Iraq), and geometry matching will be doing with correct coordinate system (WGS_1984_UTM_Zone_38N). Second, apply by work on band individually to see track and detect the changes spectral band on Al Hammar Marsh Topography such as classify.

Third, Spatial Analyst, many processes are represents cell or pixels from all scenes of images, such us, cell statistics, zonal statistics and Neighborhood statistics. Forth, The observation surfaces for each climatic variable such as temperature, Relative Humidity and Total precipitation, which is useful for predicting the climate at every region selecting using regression by apply inverse distance weight (IDW) for validation dataset of European Centre for Medium-Range Weather Forecasts – Official (ecmwf)...www.ecmwf.int. And finally, reflectance the result and illustration in graphs and tables, in order to find Spectrally bands of satellite MODIS imagery trace and detect features Al Hammar Marsh topography, and determine the cover area of each object (water, grass or vegetation, shallow and soil). Classification process, this technique depends on cell algorithms, by satellite image (RGB) separated for individual band one, two, and three. Supervised manner Automatic color extraction, extracts the main features by reclassification, use unsupervised manner, and Notice in works subsequent.

### Statistics process and analysis

In order to obtain the classification of study area cover types. This methodology allowed the creation of thematic maps use classes, which can be using as a cartographic background in topography studies. The results confirm that the classification of MODIS image is a powerful tool for region development, environmental protection, policymaking, risk assessment and planning. Therefore, Classification of single pixel (smallest object in image) is in a multi-dimensional feature space, the spatial context plays a modest role in pixel-based analysis. Consequently, classical algorithms of pixel based image analysis are becoming important for high-resolution classification, Alternatives to a pixel-based classification are developing currently for instance the object oriented approach that takes into account the form, textures and spectral information. Object based classification starts with the crucial initial step of grouping neighboring pixels into meaningful are as. This means that the object (topology) generation must be set according to the resolution and the scale of the expected objects, see [6], [9] and [10]. Most of all process and analysis image satellite depended on pixel, which allowed image processing and determined to find statistics on all cell (pixel), neighbor cell, zonal include cell, and histogram. In this study, assume statistics of all cells to find difference of standard deviations mean for each band of satellite MODIS imagery. This study represents, determine work area and clipping data frame from source imagery shown figures 3 and 4. Monitoring and Protecting of AL-Hammar marsh topography (vegetation, water, shallow and soil) is major concerns for many local and state agencies. High-resolution

## RESULTS

Satellite remote sensing has important tools that can be apply potentially to gather information needed for water, vegetation, shallow and soil area assessments in the marshes. The presented classification results show the ability of the so far developed rule-sets to distinguish between objects and to generate semantic classes. This is demonstration with the high value of the overall result accuracy of the classification; Advantages of object-oriented analysis are meaningful statistic and texture calculation, see [11].The classification technique of this study depends on extract accurately value of each object cell, recently type of satellites MODIS imagery used, and very good results had been gotten overland stage. As show in figure 3, by use band 1, 2, 3, and classification calculate cell algorithms, alternatively, the main features of contain objects, by reclassification. Assumptions, the area of each object calculate to compare with each band, to see optimize bands work, and with any satellite recently, as show in figure 4. To find the band one, water and soil objective observations optimized from band 2, 3. In addition, the band three find optimize from band 2, 3 in observation vegetation, shown [10], and see tables 3, 4 and 5. To find the shallow regions are capture in study only band3 articulately, the region of shallow widely can be able water full future, shown figure 4 and see table 5.

For climatic of the study area, The dataset coverage were obtained from website of the European Centre for Medium-Range Weather Forecasts – Official (ecmwf), All aiming research to continuously improve may be Integrated

Forecasting System (IFS), see [12]. The dataset comprise points location represented by regular grid and for each point include average of temperature (C deg), relative humidity(%) and total precipitation(mm) for each monthly, the distance between two pint location in regular grid is 0.733 decimal minute degree (dm) that is measured equally about 11.7 kilo meter (km), show n figure 5. The regular grid of point location is limited coverage from upper top left (46 14.962, 31 15.323) and down bottom right (47 59.806, 30 0,268) Degrees Minutes, that is covered the study area because, the weather factors are variables instability and movement. The dataset of weather factors are converted from decimal degree to Universal Transverse Mercator (UTM) coordinate system and tabulated into table, see table 6.

### Inverse Distance Weighted (IDW) Inter [olation

The Inverse Distance Weighting interpolator assumes that each input point has a local influence that diminishes with distance. It weights the points closer to the processing cell greater than those do further away. A specified number of points or all points within a specified radius can be used to determine the output value of each location. Use of this method assumes the variable being mapped decreases in influence with distance from its sampled location, see [13].
The Inverse Distance Weighting (IDW) algorithm effectively is a moving average interpolator that is applied usually to highly variable data. For certain data types it is possible to return to the collection site and record a new value that is statistically different from the original reading but within the general trend for the area. The interpolated surface, estimated using a moving average technique, is less than the local maximum value and greater than the local minimum value. IDW will use the measured values surrounding the prediction location. Those measured values closest to the prediction location will have more influence on the predicted value than those farther away. Thus, IDW assumes that each measured point has a local influence that diminishes with distance. The IDW function should been used when the set of points is dense enough to capture the extent of local surface variation needed for analysis. IDW determines cell values using a linear-weighted combination set of sample points. It weights the points closer to the prediction location greater than these farther do away, hence the name inverse distance weighted.

The IDW technique calculates a value for each grid node by examining surrounding data points that lie within study area. The node value is calculating by averaging the weighted sum of all the points, see [13]. Apply spatial analysis at the region by using interpolation techniques inverse distance weighted (IDW) on weather factors individual variable such as temperatures, shown figure 6. The useful techniques of IDW are obtained best results which samples (weather variable) are suited dense with relate study area variation to achieve simulate processing. The linear on surface continuity called barriers are used to specify the location of linear features. These features characteristic are not have z-value, faults, cliffs, and embankments which are resembling barriers.Barriers limit the selected set of the input sample points used to interpolate output z-values to those samples on the same side of the barrier as the current processing cell. Separation by a barrier is determined by line-of-sight analysis between each pair of points. This means that topological separation is not required for two points to be excluded from each other's region of influence. Input sample points that lie exactly on the barrier line will be included in the selected sample set for both sides of the barrier, see [14].

## DISCUSSION AND CONCLUSION

Monitoring and Protecting of AL-Hammar marsh topography such as vegetation, water, shallow and soil are major concerns for many local, state agencies and organizations. High-resolution satellite remote sensing has important tools to gather data captures needed for topography and climatology assessments in the marshes comprehension, and weather factors effective.The presented classification results the ability of developed rule-sets to distinguish between objects and classes. This is demonstration with the high value of the overall result accuracy of the classification**;** these are meaningful statistic and extract topology texture calculation. The classification technique in this study depends on extract accurately value of each object cell (pixel), recently type of satellites MODIS imagery used, and very good results had been gotten overland word stage, and classification calculate cell algorithms,

alternatively, the main features of contain objects, by some procedure process. Assumptions, the area of each object calculate to compare with each band, to see optimize bands work, and with any satellite recently at future. In this search may be found shallow regions observations great useful widely which can be able water full future, show previously figures. They can monitoring and observation the weather factors coverage earth surface which movement effective on the basin of Alhammar marsh and comparison with any months or days data deduction and derivate relation water stockpile, and increasing the green area in order to ecology improvement.

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[13] http://www.gisresources.com/types-interpolation-methods_3/.

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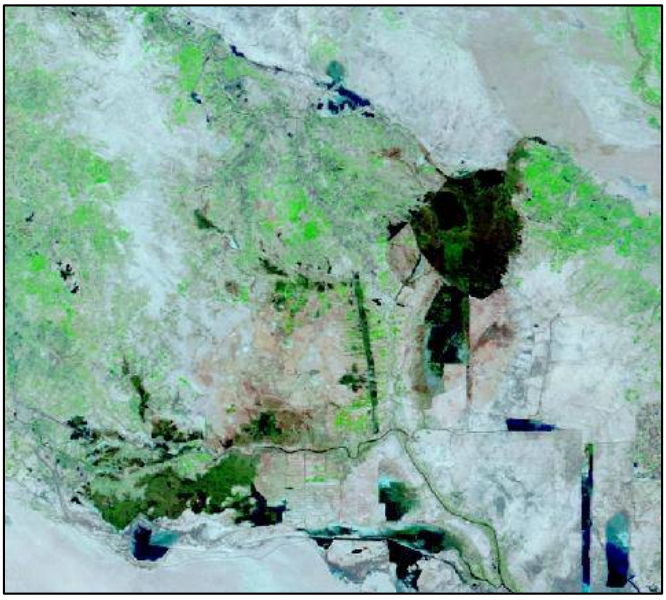


**Fig 1: Source Satellite MODIS Imagery (RGB) raw, production 12-3-2009, which comprises the study area.**

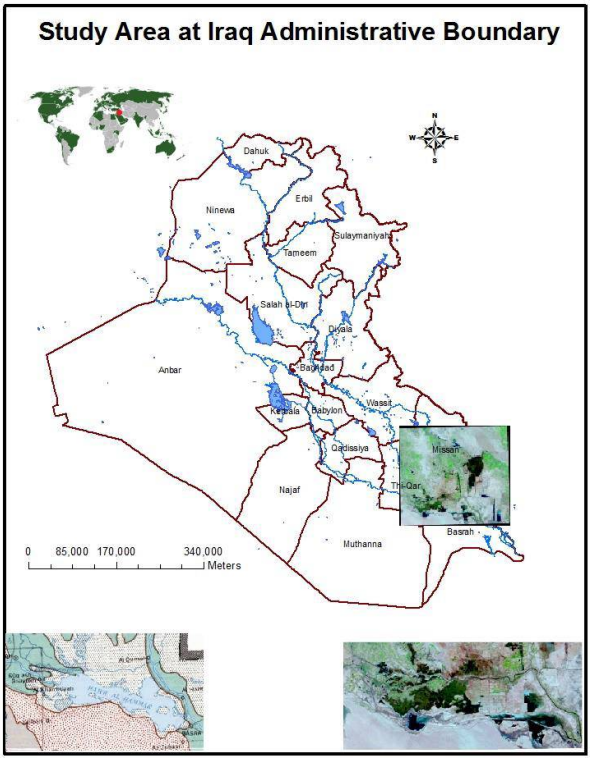



**Fig 2: Represent the study area. Illustration satellite image of Al Hammar Marsh location in south east of IRAQ Map.**

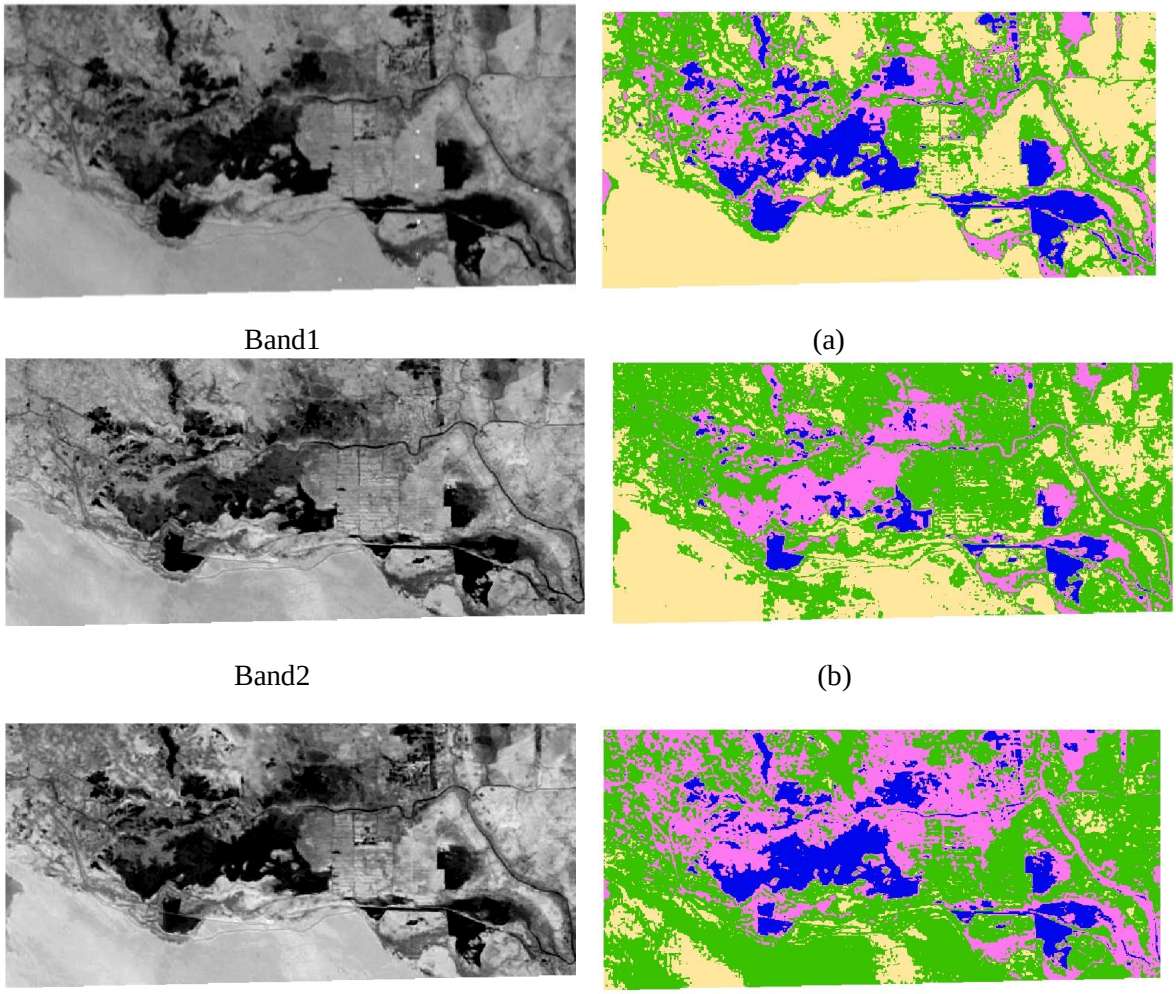
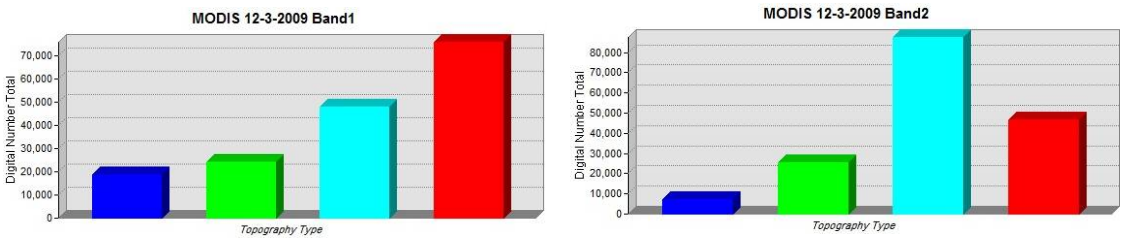



**Fig 3: Presentation, The Satellite MODIS Imagery Band 1, Band 2 and Band 3, and applied supervise and unsupervised process for Classification a,b, and c respectively..**



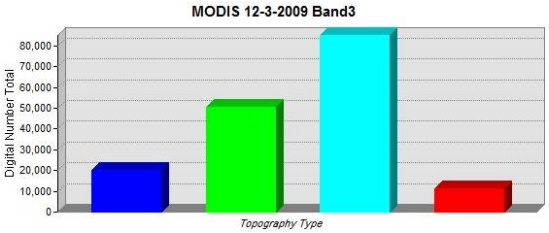



**Fig 4: Illustration of graph, which are representation AL Hammar Marsh Topography area for Band 1, 2, 3 classifications.**

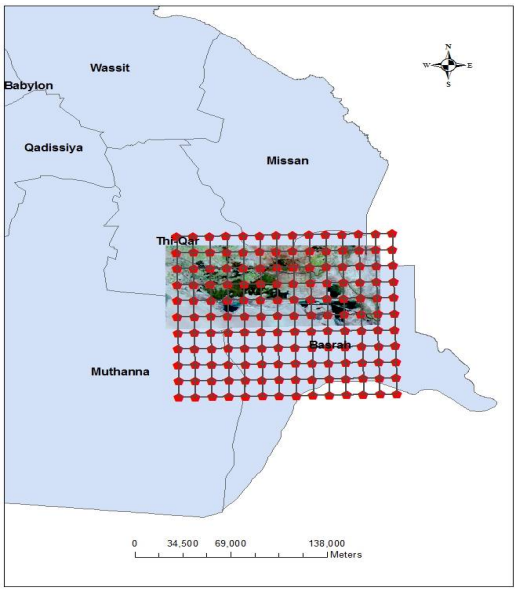



**Fig 5: Representation, the study area coverage by location point gride of weather factor variable.**

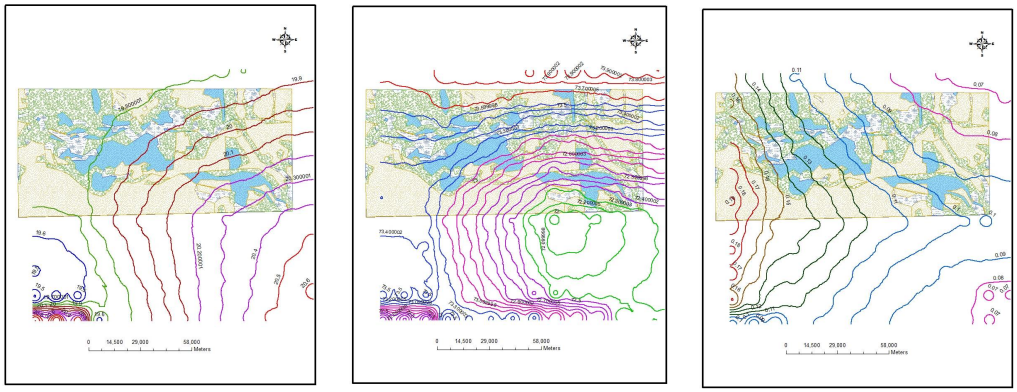


**Fig 6: Illustration, the movement of the weather factors variable on study area are temperature (C deg), relative humidity(%) and total precipitation(mm) respectively.**

**Table 1: Characteristic of satellite MODIS imagery (RGB).**

| Satellite Sensor | Spectral Band No | Description | Mean (μ) | Standard Deviation (σ) | Maxim | Minim |
|---|---|---|---|---|---|---|
| **MODIS** | Band 1 | Blue | 140.12273 | 57.95348 | 255 | 0 |
| | Band 2 | Green | 144.65425 | 40.30133 | 203 | 0 |
| | Band 3 | Red | 126.03875 | 43.82532 | 190 | 0 |

**Table 2: The Instrument Characteristics of satellite MODIS imagery (RGB), NASA agency.**

| Band | Bandwidth (nm) | Spectral Radiance (W/m2 -μm-sr) | Required SNR (Signal-to-noise ratio) | Key Use |
|---|---|---|---|---|
| 1 | 620 - 670 | 21.8 | 128 | Absolute Land Cover Transformation, Vegetation Chlorophyll |
| 2 | 841 - 876 | 24.7 | 201 | Cloud Amount, Vegetation Land Cover Transformation |
| 3 | 459 - 479 | 35.3 | 243 | Soil/Vegetation Differences |

**Table 3: Characteristics of Topography AL Hamar Marsh when Apply Satellite MODIS band 1.**

| ***BAND1*** | | | | | | | |
|---|---|---|---|---|---|---|---|
| **NO** | **Topography Type** | **DN Range** | **Digital Number** | **Topography Area** | **MEAN** | **STD** | **MEDIAN** |
| **1** | Water | 70 | 18693 | 1215510000 | 31.4956 | 23.2678 | 33 |
| **2** | Shallow | 61 | 24288 | 1579330000 | 109.864 | 17.5772 | 115 |
| **3** | Grass | 38 | 48030 | 3123150100 | 155.194 | 11.1629 | 157 |
| **4** | Soil | 83 | 75942 | 4938129900 | 187.792 | 8.64027 | 188 |
| | | | | | | | |

**Table 4: Characteristics of Topography AL Hamar Marsh when Apply Satellite MODIS Band 2.**

| ***BAND2*** | | | | | | | |
|---|---|---|---|---|---|---|---|
| **NO** | **Topography Type** | **DN Range** | **Digital Number** | **Topography Area** | **MEAN** | **STD** | **MEDIAN** |
| **1** | Water | 158 | 6987 | 454329980 | 10.6492 | 14.3948 | 5 |
| **2** | Shallow | 255 | 25717 | 1672250000 | 80.2404 | 38.23 | 79 |
| **3** | Grass | 255 | 87504 | 5689950200 | 159.0379 | 23.9165 | 164 |
| **4** | Soil | 216 | 46745 | 3039589900 | 190.78 | 11.2122 | 192 |

**Table 5: Characteristics of Topography AL Hamar Marsh when Apply Satellite MODIS Band 3.**

| *BAND3* | | | | | | | |
|---|---|---|---|---|---|---|---|
| **NO** | **Topography Type** | **DN Range** | **Digital Number** | **Topography Area** | **MEAN** | **STD** | **MEDIAN** |
| **1** | Water | 158 | 19921 | 1295360000 | 42.1148 | 32.5713 | 42 |
| **2** | Shallow | 255 | 50717 | 3297870100 | 131.162 | 31.6460 | 137 |
| **3** | Grass | 255 | 85088 | 5532850200 | 179.386 | 15.1475 | 183 |
| **4** | Soil | 243 | 11227 | 730035970 | 197.532 | 6.8043 | 199 |

**Table 6: The converter geographic coordinates system (decimal degree) points for projection coordinate system (UTM), and for any point is taken average of temperature (C deg), relative humidity(%) and total precipitation(mm) for march month period, uropean Centre for Medium-Range Weather Forecasts – Official (ecmwf)...www.ecmwf.int.**

| Stations index Number | Longitude ($\lambda$) Degree | Latitude ($\phi$) Degree | X-axis (pixels) | Y-axis (pixels) | Temp Averg | Relativ Humidit | Total Rainfall |
|---|---|---|---|---|---|---|---|
| 1 | 46.25 | 31.25 | 619024.5 | 3457982.4 | 34.14242 | 65.67833 | 0.000396 |
| 2 | 46.375 | 31.25 | 630928 | 3458123.9 | 34.15971 | 65.70467 | 0.000396 |
| 3 | 46.5 | 31.25 | 642831.7 | 3458278.8 | 34.17697 | 65.73232 | 0.000532 |
| 4 | 46.625 | 31.25 | 654735.8 | 3458447.3 | 34.19725 | 65.77684 | 0.000538 |
| 5 | 46.75 | 31.25 | 666640.2 | 3458629.2 | 34.21754 | 65.82274 | 0.000544 |
| 6 | 46.875 | 31.25 | 678545 | 3458824.6 | 34.23777 | 65.87011 | 0.000552 |
| 7 | 47 | 31.25 | 690450.2 | 3459033.6 | 34.25806 | 65.91879 | 0.000558 |
| 8 | 47.125 | 31.25 | 702355.8 | 3459256 | 34.27836 | 65.96892 | 0.000564 |
| --- | --------- | ---------- | ---------- | ---------- | ---------- | ---------- | ---------- |
| 162 | 47.5 | 30 | 741157.8 | 3321417 | 34.71636 | 65.40973 | 0 |
| 163 | 47.625 | 30 | 753219.8 | 3321686.9 | 34.85315 | 65.37846 | 0 |
| 164 | 47.75 | 30 | 765282.4 | 3321970.1 | 34.98995 | 65.3504 | 0 |
| 165 | 47.875 | 30 | 777345.7 | 3322266.4 | 35.12674 | 65.32551 | 0 |
| 166 | 48 | 30 | 210590.3 | 3322575.9 | 35.26352 | 65.30381 | 0 |
| 167 | 46.25 | 30 | 620564.4 | 3319443 | 34.92817 | 65.35754 | 0 |
| 168 | 46.375 | 30 | 632622 | 3319581.1 | 34.43819 | 65.34448 | 0 |
| 169 | 46.5 | 30 | 644679.9 | 3319732.4 | 34.51622 | 65.33437 | 0 |

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RESEARCH ARTICLE

# Synthesis of ZnO Nanostructured using Simple Technique

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## ABSTRACT

Zinc oxide (ZnO) nanostructure was successfully synthesized on p-type Si substrate by using the microwave-assisted chemical bath deposition technique. Si substrate was seeded by a polyvinyl alcohol – Zinc Hydroxide nanocomposites layer earlier to nanostructure growing. Morphology and structural quality of ZnO nanostructure were verified using scanning electron microscopy (SEM) and X-ray diffraction (XRD) which reveals that the hexagonal wurtzite structures in the direction of (0 0 2) plane are vertical to the direction of substrate all along with the z-axis. The investigations showed that the ZnO films present nano sheet-like morphology. Photoluminescence (PL) spectra indicated the higher ultraviolet (UV) intensity located at 383.25 nm. The PL peak of ZnO/Si was approximately near to the value of exact optical band gap of pure Zinc Oxide.

**Keywords:** Zinc oxide, Nanosheet, Optical properties.

## INTRODUCTION

The semiconductor zinc oxide (ZnO) has obtained a significant research attention as a result of its exclusive properties and physical characteristics. It is a direct band gap and has a fixed wurtzite structure. It has fascinated by intensive scientific researches due to its novel properties and useful applications in transparent and spin electronics devices, and piezoelectric applications [1-4]. ZnO nanostructures in different types have been successfully grown via a various techniques including thermal evaporation, chemical vapor deposition, and electrodeposition. The last applications are extremely promising due to the exhibit relevant properties of ZnO nanostructures including high-level catalytic efficiency and effective adsorption ability. Nowadays, researchers have been focused on biosensors applications of ZnO due to its biocompatibility, high isoelectric point (IEP) of ~9.5, and abundance in nature. The high value (IEP) of ZnO causes to a unique ability to immobilize an enzyme with a low (IEP) through electrostatic interaction. Likewise, nontoxicity, high chemical immovability and high electron transfer ability make ZnO a favourable material for immobilization of biomolecules without an electron mediator and can be utilized for improving implantable biosensors [2].The nanoscale science and engineering have discovered a high capacity in the fabrication of many applications like; gas sensors[5],field-effect tansistors [6 ], solar cells [7] ,antibacterial activities

[8], LED [9], laser systems [10], and photocatalysts [11].This study aims specifically to synthesis ZnO nanostructure using chemical bath deposition (CBD).

## MATERIALS AND METHODS

The experimental chemical materials that used in this technique were an analytical mark and applied without any extra purification. The substrates were carefully cleaned with the suitable procedure prior to the growth of the ZnO nanostructure. The process of growing the ZnO nanostructure can be summarized as following: (0.1 mol/L) watery solution of $ZnCl_2$ was strongly stirred for about 10 mins at 70 ◦C. Consistently, 1.5 gm watery solution of polyvinyl alcohol was blended for 30 mins at 80 ◦C. These two solutions were collectively mixed via a blender and then located on a hot plate within 70 ◦C for about 2 hours. A microwave oven with 80 ◦C for 15 mins has utilized to smooth the complexation of zinc ions with polyvinyl alcohol (PVA). The nanocomposites of polyvinyl alcohol – Zinc hydroxide solution was produced by adding Ammonia solution into the blend till the value of pH touch 8.The polyvinyl alcohol – Zinc hydroxide nanocomposites solution was totally spin- covered all substrates as a seed layer. The substrate was annealed with 210 ◦C for approximately 1 hour to decay the Zinc hydroxide into ZnO. Microwave's temperature was subsequently raised to 380 ◦C for about 2 hours. After annealing, the substrate was vertically placed in a beaker including 0.1 mol/L of zinc nitrate hexahydrate ($Zn(NO3)_2{\cdot}6H_2O$) and an equivalent molar concentration of Methenamine or Hexamethylenetetramine ($C_6H_{12}N_4$) melted in DI water .Then, the beaker was positioned in a microwave within 85 ◦C for approximately 2 hours. Conclusively, it is important to wash the substrates of grown ZnO nanostructure with a hot ethanol to eliminate the remained salt and dust.

### Characterisations

The structural properties of the prepared ZnO nanostructure films were investigated using high-resolution X-ray diffraction via X'Pert Pro MRD diffractometer (PANalytical Company). System source is Cu K-alpha (wavelength $\lambda$ = 1.5418 $A^0$) and running with 20 mA and 40 kV. The surface structure of ZnO nanostructure films were examined by SEM via Jeol JSM-6460 LV microscope working with 10 kV. Photoluminescence (PL) spectra were investigated by HR 800 UV, HORIBA Jobin Yvon, and Edison, NJ, USA at wavelengths of 325 and 514.5 nm respectively.

## RESULTS AND DISCUSSION

### Structural Characterizations

The crystal qualities of ZnO thin film deposited on Si (111) were studied with XRD and measured as shown in Fig.1. It is obvious that all the films are polycrystalline with valuable crystallinity. X-ray diffraction spectra show the presence of four peaks , all the diffraction peaks were totally matched to the standard card JCPDS No. 01-079-0205 at 2θ = 31.909, 34.586, 36.398, 47.702, and 56.276° along (100), (002),(101),(102),and (111). The diffracted intensities were calculated in the angles from 30° to 60° at 2θ. The highest value of intensity for the crystal growth orientation has showed up on (101) plane, which was placed at 36. 398°. The X-ray diffraction spectra obviously specified that the diffraction peaks satisfactory match with the hexagonal wurzite structure of the Zinc oxide [11]. The narrow and sharp diffraction peaks expressed that the ZnO nanostructure was synthesized with high crystallinity. Table (1) presents the XRD data of Zinc Oxide from JCPDS card evaluated with the experimental XRD results of ZnO .A favored orientation of the deposited films alongside (002) or (101) directions was detected depending on the substrate position. The Lotgering orientation factor was utilized to estimate the degree of orientation [12].

$$f = \frac{(P - Po)}{(1 - P)} \cdots\cdots\cdots (1)$$

where P is the peak intensity ratio of directions for oriented samples and $P_o$ is peak intensity for powder as standard specimen. In case of the orientation factor f is 1; it indicates that the degree of the orientation is complete; while a completely random state is shown for f < 1. Thus the degree of orientation for (101) plane is complete .The value of *f* for other orientations was found to be less than 1 .This result proves that the ZnO nanostructures dominantly grow along the c-axis at high solution temperature as discussed [13].

The lattice parameters of nanostructure ZnO thin films was verified by fitting the more intense diffraction peak using a Gaussian function and is given by [14]:

$$\frac{1}{d^2} = \frac{4}{3}\frac{(h^2 + hk^2 + k^2)}{a^2} + \frac{l^2}{c^2} \text{.....( 2)}$$

Where *h,k* and *l* are the Miller indices; *d* is the interplanar space; and *a* , *c* are the lattice parameters. The values of ***c*** was found to be 5.1876 $A^o$ for the ZnO nanostructured .

The average crystallite size ($C_s$) of ZnO nanostructured can be estimated from the famous Scherrer's formula [2 ]:

$$C_S = \frac{k\lambda}{\beta \cos\theta} \text{.....(3)}$$

Where *k* is a constant (0.94), $\lambda$ is the X-ray wavelength (1.5418$A^0$), and $\beta$ is the full width at half maximum (FWHM) of a defined XRD peak. It is found that the crystallite (grains) size of the ZnO Nanostructured films 20 nm.

### Microstructural Analysis: Scanning Electron Microscopy

Figure 2 presents SEM of as-deposited nanostructured ZnO film. It can be noticed that ZnO nanostructure films completely cover the substrate, without detecting pinholes or cracks.The formed ZnO have nano sheet-like morphology .The films thickness was found to vary slightly with substrate position.

### Photoluminescence (PL)

Photoluminescence spectra at room temperature in the wavelengths from 350 to 650 nm were obtained via the He–Cd laser (325 nm) as an excitation source. Figure 3 shows the characteristic Photoluminescence spectrum of a ZnO at room temperature where nanostructure samples deposited on Si(111) using MACBD. The ZnO nano-sheet showed higher ultraviolet (UV) intensity located at 383.25 nm. The PL peaks of ZnO/Si were close to the optical band gap of pure ZnO [15,16,17,18]. The PL broad band appearing in the visible region is related to the defective states in the ZnO lattice. Since surface defects profusely exist in Zinc oxide nanostructures [19, 20] according to their high surface to-volume ratio (sa/vol) [21, 22], and the chemical bath deposition growth is often increase the oxygen vacancies [23, 24]. It is predictable that a strong visible light emission can be detected in ZnO nanosheet [25, 26]. Consequently, the detected Photoluminescence peak may be attached to deep level defects, like surface defects and separately ionized oxygen vacancies (V0+) [25, 27]. The defective Photoluminescence bands of ZnO/Si are located at (509 and 760) nm. Native defects in the lattice strongly influenced the optical and electrical properties of a semiconductor. These defects strongly affected on doping and minority carrier lifetime as well as luminescence activity. The native defects play an important role in ZnO conductivity because of the high level of unintentional n-type conductivity where O vacancies and Zn interstitials are regarded as sources of n-type conductivity [28].

## CONCLUSION

Znic Oxide nanostructures were effectively deposited on Si-substrates using simple method without using any kind of metal catalyst. Nanostructures ZnO thin films exhibit a hexagonal wurzite structure as confirmed by XRD analysis, and show very good crystalline quality. Both lattice parameter and crystallite (grains) sizes were calculated.

SEM indicates that ZnO thin films entirely cover the substrate without detecting any pinholes or cracks. The formed ZnO have nano sheet-like morphology.The Photoluminescence spectra of Zinc Oxide nanostructures demonstrate a powerful UV excitonic peak at low temperatures which remains to room temperature.

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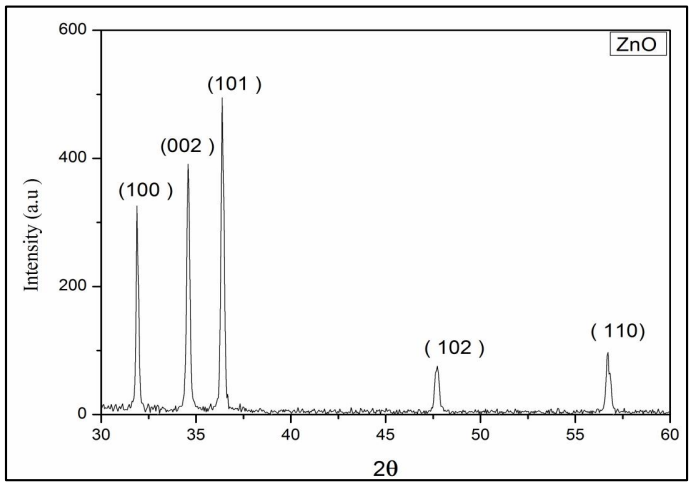


**Figure 1. X-ray diffraction pattern of ZnO nano-sheet**

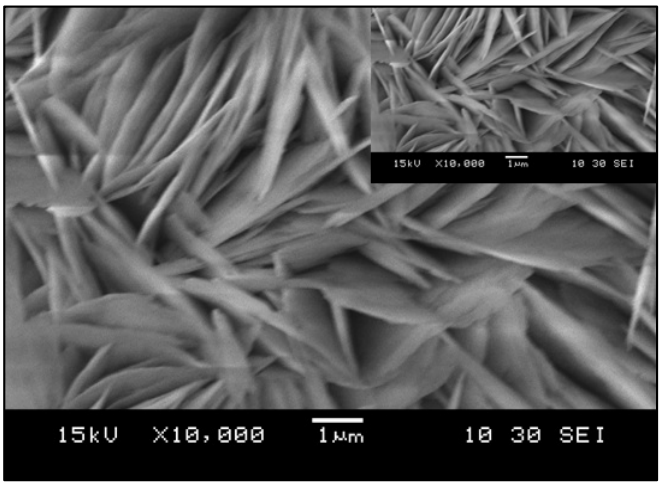


**Figure 2. SEM images nano-sheet deposited on silicon substrate.**

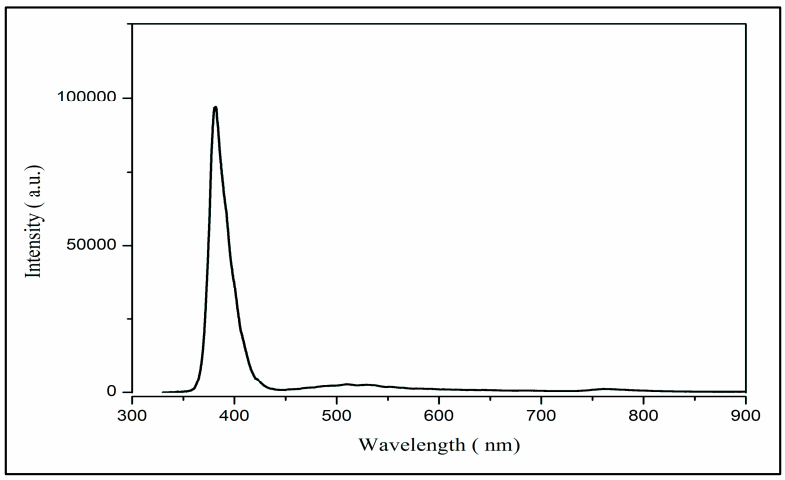


**Figure 3. PL of ZnO nano-sheet deposited on silicon substrate.**

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RESEARCH ARTICLE

# Histological Features of Penis in Indigenous Tom Cat

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## ABSTRACT

The present study involved five adult male cats to investigate the histological features of penis in indigenous tom cat. The samples of penis were fixed in 10% buffered formalin saline and there parts processed with routine paraffin technique and tissue sections were stained by hematoxylin and eosin stain. At the level of the glans penis, the glans was covered with stratified squamous epithelium modified into horny spines and the core of glans penis composed of single corpus cavernosum that supported via Os penis and core of adipose connective tissue and showed no retractor muscle. At the level of body of penis the tunic albuginea was composed of three layers that housed single corpus cavernosum and showed presence of retractor muscle and no presence of Os penis and no ventral groove. At root the two crura was housed the secretory units of bulbourethral glands. The histometrical measurements revealed significant differences at (P>0.05) between tunica albuginea and cavernous tissue of corpus cavernosum. The study concluded that the penis of tom cat has belonged the mixed type of both fibro muscular & Ischia cavernous. Our conclusion, this study described the histological structures of the penis of the tom cat in Iraq. The results also showed the differences between the penis of cat and other animals and provided acknowledge of reproductive system in carnivorous.

**Keywords:** Tom Cat, Penis, Histology, bulbourethral glands.

## INTRODUCTION

The male reproductive function in the domestic cat is very vital for acknowledgment because this species are useful for biomedical research [1, 2] and may become a valuable animal model for examining the physiology of reproduction of felines [2, 3]. Variety in the genital anatomy of male mammals is differentiated by great Dissimilarity in penile morphology, including unexplained differences between closely related species in the relative development of character such as the Os penis, and keratinized penile spines [4, 5, 6, 7]. The glans penis of the male cat is covered by relatively large, pointed, horny spines which are sensitive to androgens, and they are the only known external indicators of the level of male hormone in cats [8]. The penis and its associated structures have provided functional classification among many groups of mammals while also functioning as a shared passage for excretion of urine and semen ejaculation [9]. The penis of the stallion is classified as a vascular penis due to importance of caverns in the

corpus cavernosum , much blood space and delicate tunica alboginea, in ruminant and boar the caverns less wide and connective tissue overcome, small blood space and thick tunica alboginea, thus differentiate as a fibroelastic, however, in cat and dog are intermediate[10,11] . Review of literature revealed rare studies about the microscopical structure of the penis of the local tom cat in Iraq. Therefore, the current study aimed to investigate the histological structures of the penis of the local tom cat in Iraq.

## MATERIALS AND METHODS

Five adult male cats were obtained from the Department of Surgery and Obstetrics at the Faculty of Veterinary Medicine, University of Baghdad after her death due to unsatisfactory accidents were used for this study. The study was carried out at department of Anatomy & Histology, University of Baghdad. The male genitalia were removed immediately and fixed in 10% buffered formalin saline and then the penis was dissected for three parts (Root, Body & glans penis). The Samples were processed with routine histological procedures with upgrading concentration of ethanol (70, 80, 90 & 100%), cleared with xylen and finally embedded in paraffin bath at (58C), then sectioned at 5-6 micrometer with rotary microtome. The tissue sections were stained with hematoxylin and eosin stain. The sections examined by light microscope and images captured for all histological structures of the penis and testes of the cat [12] and Gamble 2008).Statistical analysis has been done by using The SPSS statist version [13] to analyze the data. The results were displayed in Means and Standard Error. One way analysis of Variance (ANOVA) has been used to detect age related variation. The value ($P<0.05$) was considered to be significant.

## RESULTS

Histologically, the glans penis of the tom cat was covered by relatively huge, pointed, horny spines, which covered by keratinized stratified squamous epithelium which supported by well vascular and innervated connective tissue (fig.1 & 2). The glans penis showed corpus cavernosum which has surrounded by dense connective tissue that called tunica albuginea (fig.2). The tunica albuginea has sent trabeculi to corpus cavernosum formed a network of cavernous tissue that lined with endothelial cells, the trabeculi showed a branches of pudendal artery (fig.3&4). The core of glans penis has supported by Os penis which is represent as compact bone and core of adipose connective tissue (fig.1&5).The corpus spongiosum has surrounded the urethra and consisted of plexus of large caverns tissue that surrounded by connective tissue , the urethra lined by stratified squamous epithelium (Fig.6). At the level of glans penis the section showed no retractor muscle (Fig.1 & 6). The cross section of the body penis showed three well distinct layers of connective tissue (fig.7); the firs layer under the skin was thick tunica albuginea, the second layer was well vascular and innervated connective tissue which contained the three main branches of pudendal artery (fig7 & 8). The third layer represented thin layer of dense irregular connective tissue which sent trabeculi to form the network of cavernous tissue of single corpus cavernosum (fig.7&8) the corpus cavernosum composed of cavernous tissue intermingled by adipose connective tissue and supported by collagen bundles (fig.8). Within body part the section showed no ventral groove present at the ventral surface of the penis, so the urethra located ventrally to the corpus cavernosum and surrounded by thin distinct layer of corpus spongiosum, the urethra was lined by transitional epithelium and supported by vascular layer of sub epithelial connective tissue (fig.7&9). Ventrally to the urethra there was a thick bundle of smooth muscle of retractor muscle (fig.10).The skin that covers the penis of the cat showed no hair follicles (fig.7). The root of penis was composed of two crura, each has composed of thick bundles of skeletal muscle housed the secretory units of bulbourethral glands (fig.11&12). Histometrical measurement showed that the ratio of tunica albuginea was 59.8% while the ratio of corpus cavernosum was 40.2%, statistically significant differences between the ratio of tunica albuginea and corpus cavernosum.

## DISCUSSION

The results of the microscopical features of the present study revealed that the glans penis of the tom cat was covered by a horny spines, which covered by keratinized stratified squamous epithelium this result was coincided with [14,15] in chimpanzee and mice and [16] in cane rat, these cornified papillae considered as secondary sex, men while [17,18,19] in camel showed the same keratinized stratified squamous epithelium covered the organ. But [20] showed non keratinized stratified squamous epithelium in buffalo .The glans penis has a cone shape and showed a single corpus cavernosum surrounded by dense connective tissue of tunica albuginea which is sent trabeculi to form a net work of cavernous tissue intermingled by adipose tissue and supported with collagen bundles that lined by endothelium, on the other hand there's no hear follicles were presented, this results were alignment with[21] in dog[22] in Gazella but they are decleared a paired instead of single corpus cavernoum without adipose tissue, in addition[17] showed that the tunica alboginea in camel consisted of cartilaginous cells inside the collagen fibers of tunica albuginea and are around C.C.P and C.S.P and their distribution is dissimilar in different part of penis the same results were mention by[23] in ram and buck when showed that the crural septum of tunica alboginea that separated the copus cavernosum of the two crura will disappear at the level of the proximal part of body and then united and became one erectile body. But [24] in monkey mention un incompatible results when described the penis, said, this organ has a triangular buttom shape glans penis. The core of glans penis has supported by Os penis which is represent as compact bone, this results was in contrast with [21] in dog whom stated that the Os penis was a spongy bone and[25,16] whom described the porcupine Os penis as mixed structure, the outer was compact bone and the inner was spongy bone, while[26] reported that the mice Os penis consisted of compact bone as the present study. The other histological section revealed that the corpus spongiosum has surrounded the urethra and consisted of plexus of large caverns tissue that surrounded by connective tissue , the urethra lined by stratified squamous epithelium at the level of the distal part of the penis at the level of glans penis the section showed no retractor muscle, while the part of the body section showed no ventral groove present at the ventral surface of the penis, so the urethra located ventrally to the corpus cavernosum and surrounded by thin distinct layer of corpus spongiosum and was lined by transitional epithelium and supported by vascular layer of sub epithelial connective tissue. Ventrally to the urethra there was a thick bundle of smooth muscle of retractor muscle, [27,28] stated that the urethra of the Bull, Ram, Stallion and human formed by stratified cuboidal to columnar epithelium instead of stratified squamous and transitional epithelium [18] in camel and[16] in cane rat have the same opinion about the presence of transitional epithelium and stratified squamous epithelium. Mean while [21,29] in dog stated that the corpus spongiosum consisted of plexus of large cavernous surrounded by connective tissue and the retractor muscle observed as smooth muscles similar the current study.The histometrical measurement showed that the ratio of tunica albuginea was 59.8% while the ratio of corpus cavernosum was 40.2%, statistically a significant differences between the ratio of tunica albuginea and corpus cavernosum, according to this result our opinion suggested that the penis of tom cat classified as intermediate type, tends to fibroelastic than cavernous type.

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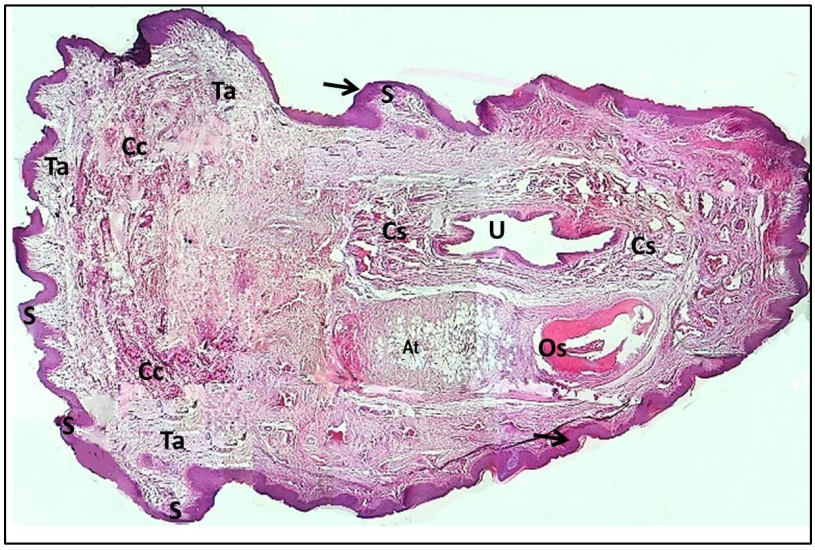


**Figure 1:Transverse section of glans penis (Tom cat) shows: spines (S), tunica albuginea (ta), corpus cavernosum (Cc), corpus spongiosum (Cs), urethral space (U) Adipose tissue (At) Os penis (Os) and epithelium(E). H&E stain. 40x**

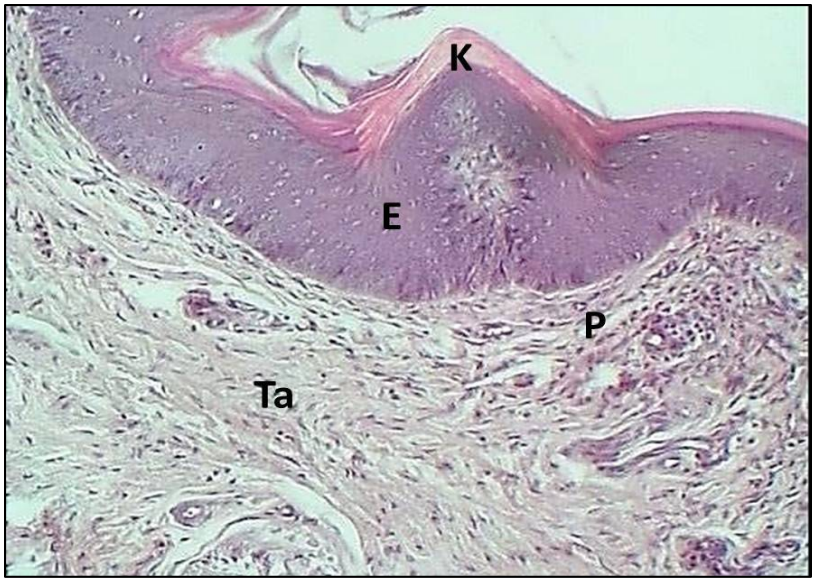


**Figure 2: Magnified section of spine shows: keratin layer (k), stratified squamous epithelium (E) proprial subepithelial connective tissue (P) & tunica albuginea (Ta). H&E stain. 100x.**

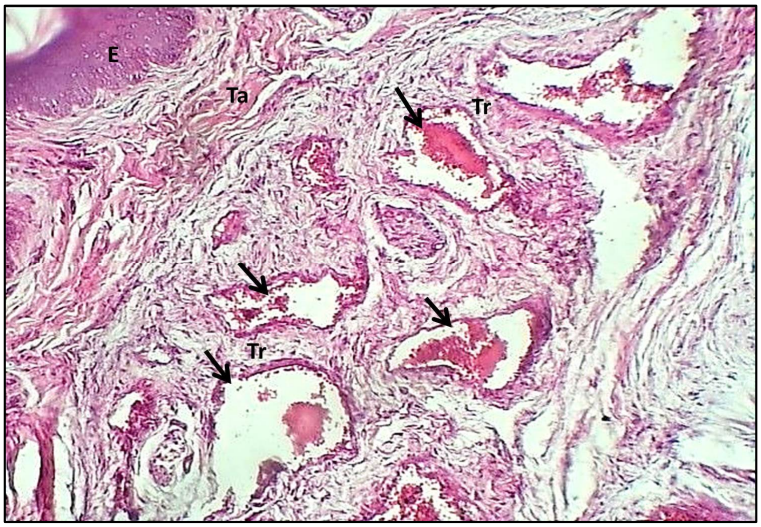


**Figure 3: Magnified section of glans penis shows: epithelium (E), tunica albuginea (Ta), trabeculi (Tr) & erectile tissue filled with blood (arrows) H&E stain. 100 x.**

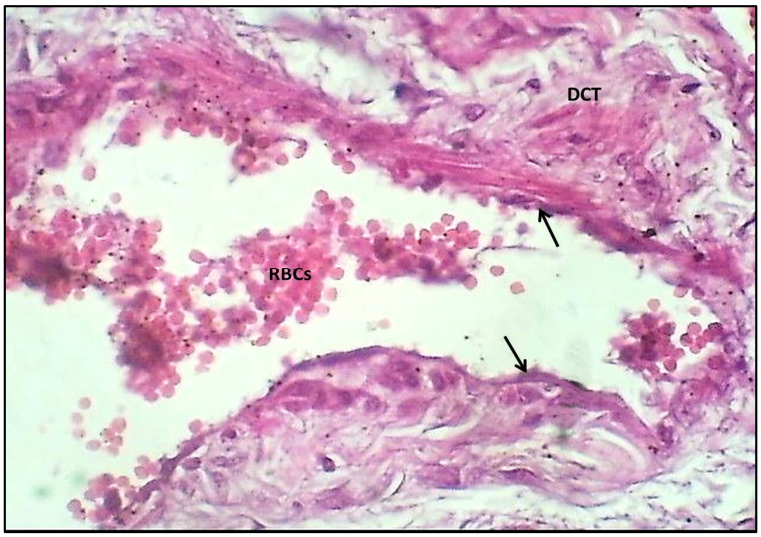


**Figure 4: Magnified section of erectile tissue shows: endothelial cells (arrows), Red Blood Cells (RBCs) & dense irregular connective tissue (DCT) H&E stain. 400 x.**


**Figure 5: Magnified section of glans penis shows: Os penis (B) and periosteum (P). H&E stain. 400 x.**


**Figure 6: Magnified section of urethra shows: urethral lumen (Ul) , epithelium (E), corpus spongiosum (Cs) & cavernous tissue (arrows). H&E stain. 100 x**

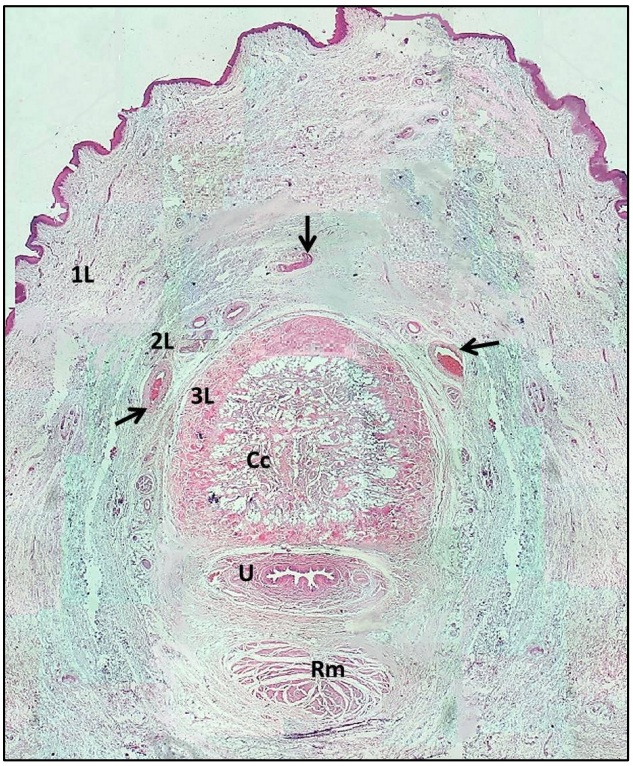


**Figure 7: transverse section of body shows: tunica albuginea (1L), vascular layer (2L), third layer (3L), corpus cavernosum (Cc), urethral (U) , (E), retractor muscle (Rm). H&E stain. 40 x.**


**Figure 8: section at the body shows: artery (a), nerve (n), vascular layer (2L) , denes connective tissue of third layer (3L) trabeculi (black arrows)& adipose tissue (Red arrows). H&E stain. 100 x.**

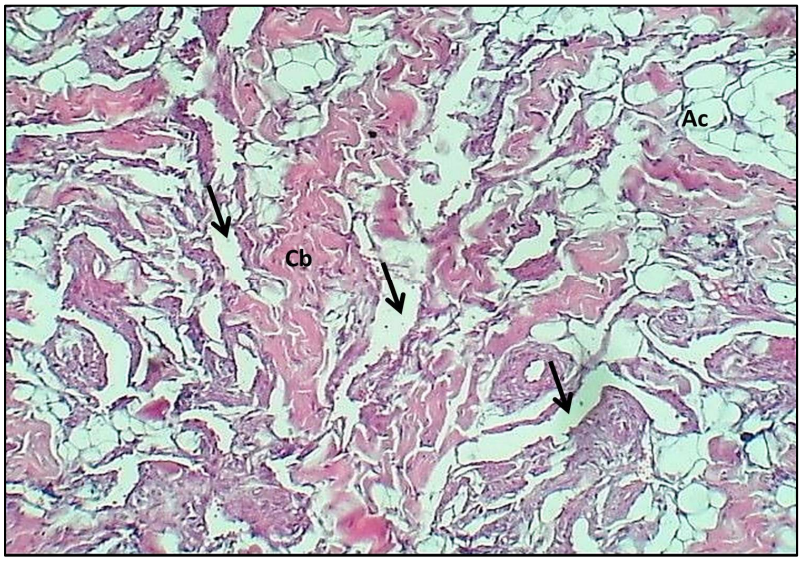


**Figure 8 A: section of corpus cavernosum shows: cavernous tissue (arrows), collagen bundles (Cb)& adipocytes (Ac). H&E stain. 400 x.**

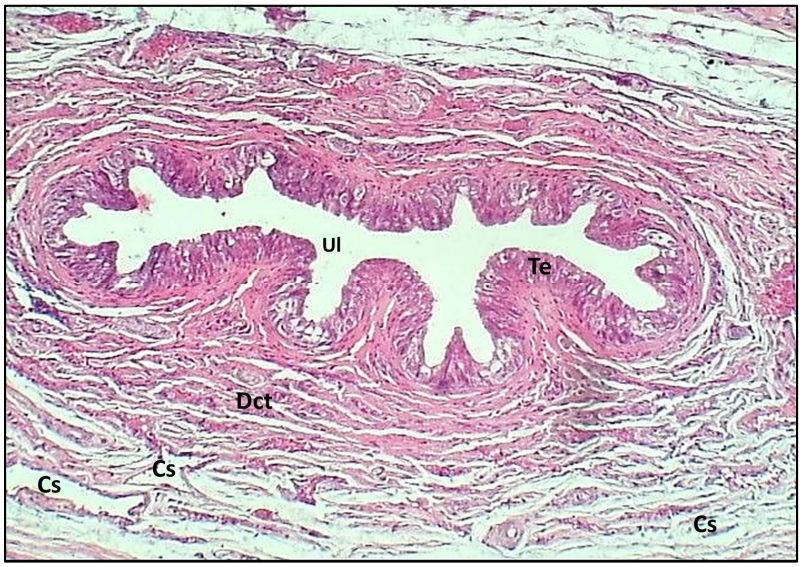


**Figure 9: section of urethra at body part shows: urethral lumen (Ul), transitional epithelium (Te), sub epithelial connective tissue (Dct), corpus spongiosum (Cs). H&E stain. 100 x.**

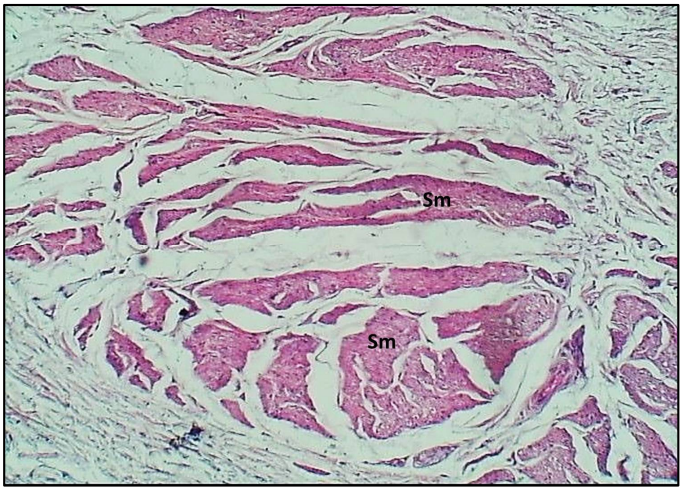


**Figure 10: section at the retractor muscle (body part) shows: bundles of smooth muscle fibers (Sm) . H&E stain. 100 x.**

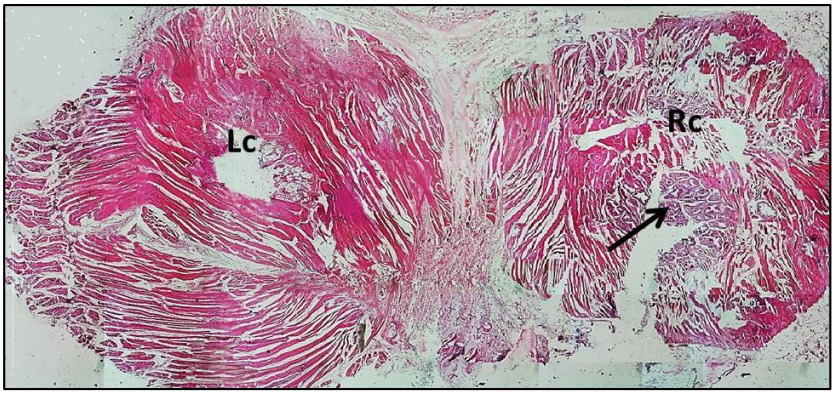


**Figure 11: section at the root of penis shows: left (Lc) & right crura (Rc) & secretory unites of bulbourethral gland (arrow). H&E stain. 40 x.**

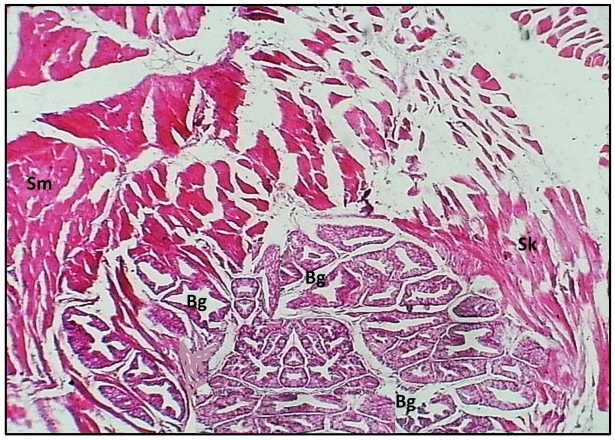


**Figure 12: magnified section at the root of penis shows: skeletal muscle (Sm) & secretory unites of bulbourethral gland (Bg). H&E stain. 100 x.**

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**RESEARCH ARTICLE**

# Influence of Etching Time and Annealing Temperature on Structural, Morphology and Gas Sensitivity Properties of $In_2O_3$/PSi Nanostructured by Laser Induced Plasma

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## ABSTRACT

In this research, nanostructures of $In_2O_3$ were fabricated by pulse laser deposition using Nd: YAG laser with $\lambda$=1064 nm which used as gas sensor. Thin films were deposited on glass, p-type Si and porous silicon (PSi) substrates using 500 mJ laser pulse energy using different shots number (200, 300, 400, 500 and 600) and annealed at different temperatures (273, 523 and 623 K) the physical properties of $In_2O_3$ thin films and then to study characterization of $In_2O_3$ /PSi as $NO_2$ gas sensor study thin films properties deposited on glass substrates.The structural properties of films were studied before and after annealing by X-ray measurements. It was found that pure $In_2O_3$films pattern, in the case (RT), has amorphous structure. When the films annealed at 523 and 623 K temperatures the films crystallinity was improved and became polycrystalline structure with rhombohedral type and has preferred orientation along (222) direction.The AFM data indicate Nano-structured grain growth for $In_2O_3$ thin films.synthesis porous silicon by electrochemical etching for Si wafer in diluted HF acid in water (1:4) at different etching times and different currents. The morphology forPS wafers by AFM show that the average pore diameter varies from 48.63 to 72.54 nm with increasing etching time from 5 to 15min and from 72.54 to 51.37nm with increasing current from 10 to 30 mA.high sensitivity for $NO_2$ gas was (155%) for $In_2O_3$/PSi at 15 min etching

**Keywords:** Indium oxide ($In_2O_3$) Nanocrystalline materials Thin films, gas sensor, metal oxide semiconductor, AFM, porous silicon.

## INTRODUCTION

Indium oxide ($In_2O_3$) is an important n-type semiconductor with its wide direct band-gaps ranged between 3.55-3.75 eV, it's also found to have an indirect band gap of about 2.6 eV [1, 2]. It considered as an important transparent conducting oxide (TCO) material. It has interesting properties such as high transparency to visible light, high electrical conductance, and strong interaction between certain gas sensor molecules and its surfaces [3, 4]. These properties make $In_2O_3$ an interesting material for a variety of applications, including solar cells, panel displays, organic light emitting diodes, photo catalysts, architectural glasses, field emission. [5, 6] Moreover, $In_2O_3$ is an important material for semiconductor gas sensors.$In_2O_3$ have been synthesized by several techniques including sol–gel technique, pulse laser deposition, thermal decomposition, thermal hydrolysis, micro emulsion, spray pyrolysis, chemical, chemical processing, of indium oxide thin films on glass substrates by ablation of a pure indium target in an oxygen reactive atmosphere. This fabrication technique allows control of the structural characteristics of the films by changing the substrate temperatures [7]. $In_2O_3$ with high optical transparency and makes it an important material for different applications. It has been widely used in window heaters, solar cells, and liquid crystal displays [8]. Properties as high transparency in the visible region and high electrical conductivity make $In_2O_3$ a suitable material for application in solar cells [9, 10], optoelectronic devices [11], liquid crystal displays [12], light emitting diodes [13, 14], photo voltaic devices etc.

The porous silicon described was fabricated by the electrochemical anodization of silicon in a hydrofluoric acid (HF) based electrolyte. This is the most common method of fabricating porous silicon though the use of an ammonium fluoride-based electrolyte has also been reported [15-17]. Porous silicon (PSi) is a crystallinesilicon, having within it a branch of nano-sized pores, it has a direct wide energy gap and high resistivity. One of the most important characteristics of PS is the layers and its vast and reactive internal surface. One would expect, that the internal surface would play a significant role in the specific properties of PS layers enabling this material to be entirely different from the bulk one [18]. Electrochemical etching is a more recurrently applied technique for the fabrication of porous silicon [19]. In this study, the morphology, crystalline structure, sizes of PS and gas sensing properties using a static gas sensing system were examined. Nanostructured of $In_2O_3$films have been tested and for several gases. These were observed to be more selective than $NO_2$ at different operating temperatures. There was a strong connection between the surface and the bulk in semiconductors, explaining why these materials are used as gas sensors in addition to many other applications. The practical form of appearance of semiconductor gas sensors is different for a thin film with a comparable thickness of the space charge layer to the total layer thickness (simple, or with the doped surface) [20,1]. The sensing mechanism is based on changes in the film's resistance, which is controlled by gas species. The sensitivity (S) of the film is defined as the ratio of the film resistance in air ($R_{air}$) and gas ($R_{gas}$).

## MATERIALS AND METHODS

$In_2O_3$thin filmswere synthesised using a pulsed laser deposition technique using Nd: YAG laser. The thin films were grown in a vacuum chamber with a background pressure of ~2.5 x $10^{-2}$ mbar. The Nd: YAG laser has a wavelength of $\lambda = 1064$ nm with 6 Hz frequency. Morphologically characteristics of the porous silicon (PSi) structure were fabricated using an electrochemical etching process in HF acid. $In_2O_3$/PSi was produced via the deposition of Electrochemical etching formed the PS. $F^-$ ions are previously obtained available in sufficient amounts at the surface which agrees with [21]. Porous silicon (PSi) having distinctive physical and chemical properties which differ from single crystal Si properties. Some of these properties are significant in gas sensor technology field. In the PSi some part silicon crystals etched away by the electrochemical process make the silicon surface contains many pores. As a result, numerous and unusual physical phenomena can be observed [22]. The morphological features and microstructures of the films were analysed using Atomic Force Microscopy (AFM-Digital Instruments, Nan Scope) operating in tapping mode

Heater consists of a hot plate and a K-type thermocouple located inside the chamber to control the operating temperature of the sensor, and a bias voltage of 6 V between the two sides of the electrode is applied. A switch on the rotary pump clears the test chamber for approximately 1 mbar, and the gas sensor is set to the desired operating temperature.Needle valves are used to control the gas flow, wherein the gas is tested under a different volumetric concentration of 3 % gas to air ratio,Resistance variation is measured using a computer interfaced to a digital multimeter. Initially, the biasing air flow resistance records by the digital multimeter records. After turning on the multimeter for several seconds, the testing gas ($NO_2$) shows a low variation of resistance. After switching off the device, the test gas is used to record the recovery time to give the real sensitivitycorresponding toAli Ahmad Yousif, [23]. The sensitivity is calculated by applying Equation (1), as given in [24]:

$$S = \left| \frac{Rg - Ra}{NRa} \right| \times 100\% \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (1)$$

where (*Ra*) and (*Rg*) represent the electrical resistance of the sensor in air, and the presence of gas respectively and (*N*) is the gas concentration. This may also be calculated from conductance as in Equation (2), [25]:

$$s = \frac{\sigma g}{\sigma a} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (2)$$

$\sigma g$ is the film conductivity in gas, while $\sigma a$ is represented the film conductivity in air.

A layer of porous silicon is doped with $In_2O_3$, on wafer silicon (P-type) and with an orientation of 100, resistively 0.01 - 0.02 and a thickness of 525 ± 25 µA. The PS samples were prepared by the electrochemical etching (ECE) method in hydrofluoric (HF) acid 17 %, and the concentration and the etching cell are made from Teflon as this does not interact with HF acid, and a rubber O-ring is used before the upper part of the cell. The electrochemical cell used has two electrode configurations with a platinum (stainless steel) and electrode as a cathode, and a silicon wafer as an anode. A digital millimetre device was connected to the silicon, and the other side was connected to the power supply. After cutting the silicon samples into 2.25 × 2.25 $cm^2$ pieces, the wafer is washes with acetone and methanol to eject any oxide layer. The samples were put in 48 % concentration of acid in a mixing ratio 4:1 HF.Using chemical ethanol ($C_2H_5OH$) with a purity of 99.9 % and a concentration of 17 %, different etching times of 5, 10, 15 min, at a constant current of 20 mA. plus, the addition of the mixing motor, helped the electrical conduction process. This led to the acceleration of the emptying of bubbled hydrogen ($H_2$) as a catalyst and alcohol ethanol that was used for the evacuation process. The porous silicon samples were next washed in ethanol after their removal from the solution. The samples were left to dry at ambient temperature for several minutes and then stored in containers consisting of methanol to prevent the formation of an oxide layer on the prepared samples.

## RESULTS AND DISCUSSION

### X-Ray diffractionfor thin film

Fig. (1) shows the X-ray diffraction patterns for the pure $In_2O_3$ thin films deposited on a glass substrate by pulse laser deposition (PLD) technique, for as deposited and annealed films at 523, 623 K. It can be noticed from the X-ray that the pattern, in the case (RT), has amorphous structure. When the films annealed at 532 and 632 K temperatures the properties of the films improved and the films became polycrystalline structure with peaks located at (30.6607°, 35.5400°, 51.0928° and 60.6989°), referred to (222), (400), (440) and (622) direction, respectively. In the case annealing at 623 K the peaks at (21.5121°, 30.5845°, 35.4638°, 51.0928° and 60.9276°) referred to (211), (222), (400), (440) and (622), which matched with the $In_2O_3$ reference of rhombohedral according to (card No. 96-101-0589). These results agree with the other researches (S. Hammon*et al.,* 2015) [26]. It can be also seen that the intensities of peaks increase with the increase of the heat treatment. The full widths at half-maximum (FWHM) of the (222) peaks in 523K and 623 K

were 0.5337° and 0.4574°, respectively. The average crystalline size evaluated from this peak, which increased from 15.44 to 18.01 nm with the increase of substrate temperature from 523 to 623 K, due to improvement in the crystallinity. This agrees with other researches [27]. Table (1) shows all XRD parameters for these samples.

### Atomic Force Microscopy for $In_2O_3$ films

Fig. (2) Shows the AFM images for $In_2O_3$ thin films deposited at energy of (500mJ). This figure indicates on formation of Nano-structured grain growth. The root means square value of surface roughness (RMS) derived from the AFM image is (2.01nm) and roughness average (1.76 nm). The grain size for $In_2O_3$ thin films deposited on glass substrates increased from 92.61 nm to 100.93 nm with increasing annealing temperature from RT to 623 K. The increase in grain size was associated with a change in shape and distribution. The maximum root means square value of 4.01 nm at 523 K annealing temperature. It is clearly evident from the images shown that there is a change in the growth as well as arrangement of grains.

### Atomic Force Microscopy for porous silicon wafer

show the Atomic force microscopic image of the oxidized PSi layer surface. The figure (3)illustrates entirely on the nanoscale characterization of PSi films when the etching time increased, a part of pores formed and be with larger structures. Irregular and randomly distributed nanocrystalline silicon pillars and the holes over the entire surface can be seen. Pore morphology when current flowed in the electrochemical cell, the dissociation reaction localized on the particular side of a silicon surface, thus the etching shaped of an array of pores in the silicon wafer. At the high etching time, a highly branched, randomly directed and highly interconnected mesh-work of the pore was obtained. AFM parameters (average diameter, average roughness) for these samples. Various affecting parameters were studied such as the current density and etching time. We have studied the morphologically properties parameters forporous silicon wafer at constant current 20mA and different etching time (5, 10, 15) min. Images showed PS layer had sponge like structure, and average diameter of porosity and thickness of PS layer increased with increasing etching time, average diameter pore range between (48.63-72.54) nm.

### Gas sensor
### $NO_2$ Sensing Mechanism for$In_2O_3$

The thin films specimens were examined for gas sensing using $NO_2$ mixed with air at a concentration of 25 ppm, at a different operation temperature beginning from room temperature 30 °C to 300 °C.The sensing mechanism is related to the desorption of gas species over the surface, leading to charge transfer between the gas and surface molecules and changes in the electrical conductance [28].These figures show increasing in the resistance value when their films exposure to NO2 gas, (Gas ON), then the resistance value back downward at the closure of the gas (Gas OFF). The reason for this behavior can be attributed to the following: Adsorb NO2 gas undergoes an ionic reaction with the surface, electron extracted from the semiconductor and causes the conductivity of the materials to decrease, thus causing the resistance to increase [22].Changing on the resistance value according to the metal oxide semiconductor sensor dissociates to form $O^{-2}$, where the negative charge on the oxygen extracted from the semiconductor. This electron extraction tends to decrease electrons concentration of the surface decreases, thus increase the resistance for the kind of n-type semiconductor where the majority charge carriers are electrons [29].

### Determination of Operation Temperature of the Sensor (15 min etching time)

The changing of resistance is influenced by the presence of the amount of interacted gases and these variations effected by operating temperature [30]. The sensitivity factor (S%) at various temperatures in the range (RT-350

˚C).which deposited on the porous silicon substrate (PS) wafer (100) for $NO_2$ gas. The PS substrate formed at constant etching current (20) mA and 15 min etching time. The gas sensitivity tests at 3 % $NO_2$: air mixing ratio. The sensitivity of the tested films increases as the temperature increases from room temperature reaches a maximum value corresponding to an optimum operating temperature at 200°C then decrease at 300°C operating temperature for all samples.the optimum sensitivity was obtained for pure $In_2O_3$ is 155%.The higher sensitivity may return to the optimum porosity, largest surface area, larger rate of oxidation and the optimum surface roughness.

## CONCLUSION

X-ray diffraction analysis showed polycrystalline structure for all thin films, the preferred peak appears along (222) plane for $In_2O_3$ sample with rhombohedral structure, the increasing annealing temperature leads to enhance the films crystallinity.Morphologically properties using (AFM)forporous silicon showed PS layer with sponge like structure, and average diameter of porosity and thickness of PS layer increased from 48.63 to 72.54 nm with increasing etching time from 5 to 15 min,AFM measurements for $In_2O_3$ films deposited at laser energy of (500 mJ) show that the average diameter increase from 92.61 to 100.93 nm and the root means square value of surface roughness from 2.01 to 2.12 nm with increasing annealing to 623 K. It is clear evident from the AFM images there is a change in the growth as well as arrangement of grains. The gas sensor measurements for samples deposited on PS prepared at 15 min etching time against $NO_2$ gas show that the best operating temperature at 200°C and the highest sensitivity is 155% for $In_2O_3$/PSi.

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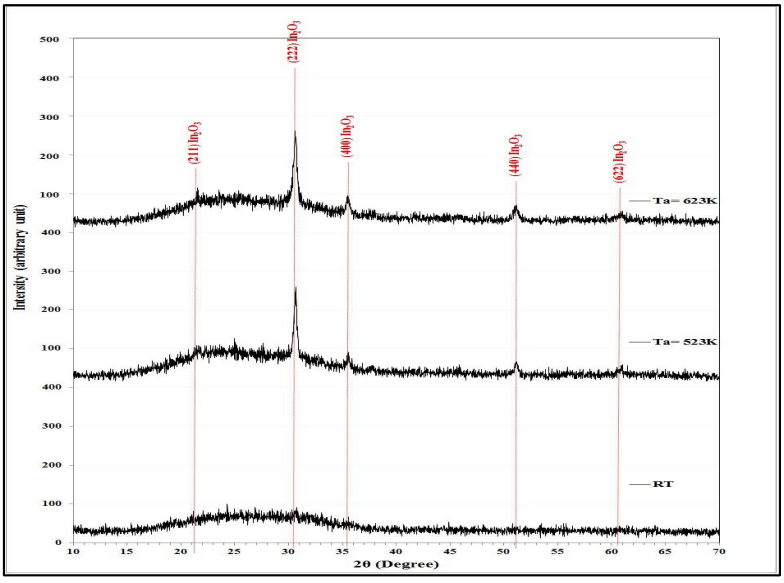


**Fig.1: X-ray diffraction patterns of $In_2O_3$ pure in three cases of temperatures RT, 523,623 K**

**Table 1 : Structural parameters: 2θ, $d_{hkl}$, (hkl), FWHM ad G.S of $In_2O_3$ films at three cases of temperatures RT, 523,623 K**

| Ta (K) | 2θ (Deg.) | FWHM (Deg.) | $d_{hkl}$ Exp.(Å) | G.S (nm) | hkl | $d_{hkl}$ Std.(Å) | Phase | Card No. |
|---|---|---|---|---|---|---|---|---|
| RT | Amorphous | | | | | | | |
| | 30.6607 | 0.5337 | 2.9136 | 15.44 | (222) | 2.9214 | $In_2O_3$ | 96-101-0589 |
| 523 | 35.5400 | 0.6099 | 2.5239 | 13.68 | (400) | 2.5300 | $In_2O_3$ | 96-101-0589 |
| | 51.0928 | 0.7624 | 1.7862 | 11.55 | (440) | 1.7890 | $In_2O_3$ | 96-101-0589 |
| | 60.6989 | 0.7624 | 1.5245 | 12.08 | (622) | 1.5256 | $In_2O_3$ | 96-101-0589 |
| | 21.5121 | 0.4574 | 4.1275 | 17.69 | (211) | 4.1315 | $In_2O_3$ | 96-101-0589 |
| | 30.5845 | 0.4574 | 2.9207 | 18.01 | (222) | 2.9214 | $In_2O_3$ | 96-101-0589 |
| 623 | 35.4638 | 0.4574 | 2.5292 | 18.24 | (400) | 2.5300 | $In_2O_3$ | 96-101-0589 |
| | 51.0928 | 0.8386 | 1.7862 | 10.50 | (440) | 1.7890 | $In_2O_3$ | 96-101-0589 |
| | 60.9276 | 0.8387 | 1.5193 | 10.99 | (622) | 1.5256 | $In_2O_3$ | 96-101-0589 |

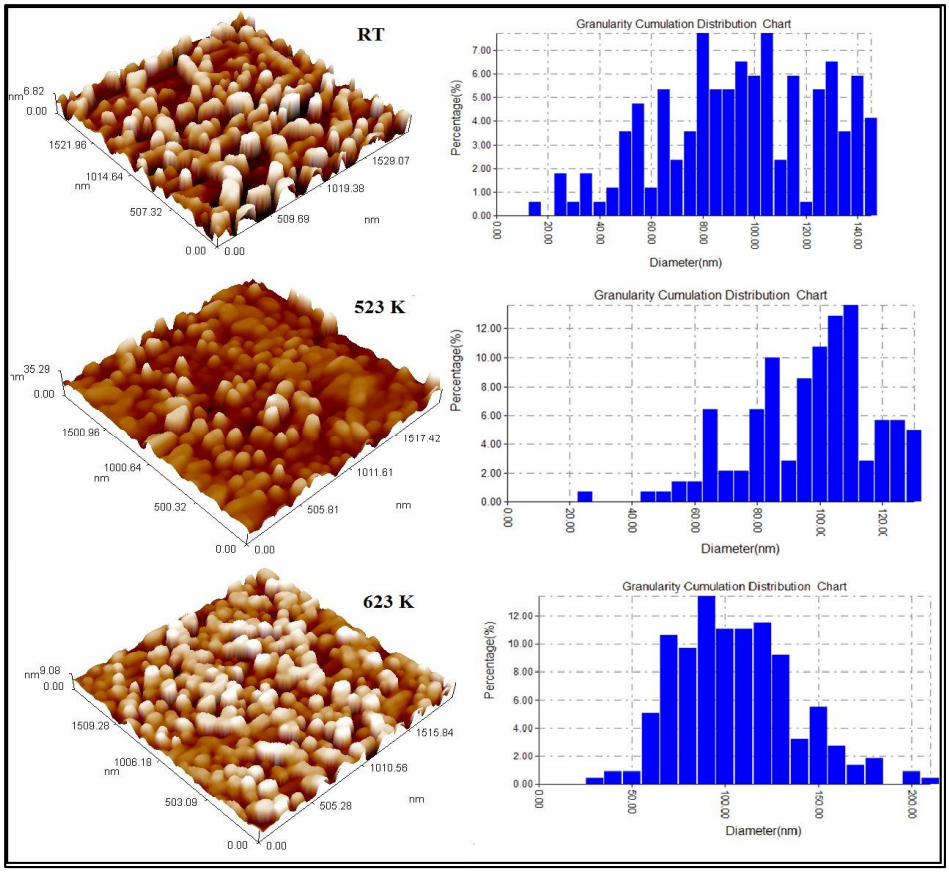


**Fig. 2: Atomic force microscopy images and the granulate distribution for $In_2O_3$ thin films annealed at different temperatures (RT,523 and 623) K.**

**Table 2 : Atomic force microscopy parameters for $In_2O_3$ thin films annealed at different temperatures (RT,523 and 623) K.**

| $T_a$ (K) | Ave. diameter (nm) | Roughness average (nm) | Root mean square (nm) |
|---|---|---|---|
| **RT** | 92.61 | 1.76 | 2.01 |
| **523** | 94.83 | 4.01 | 5.04 |
| **623** | 100.93 | 1.77 | 2.12 |

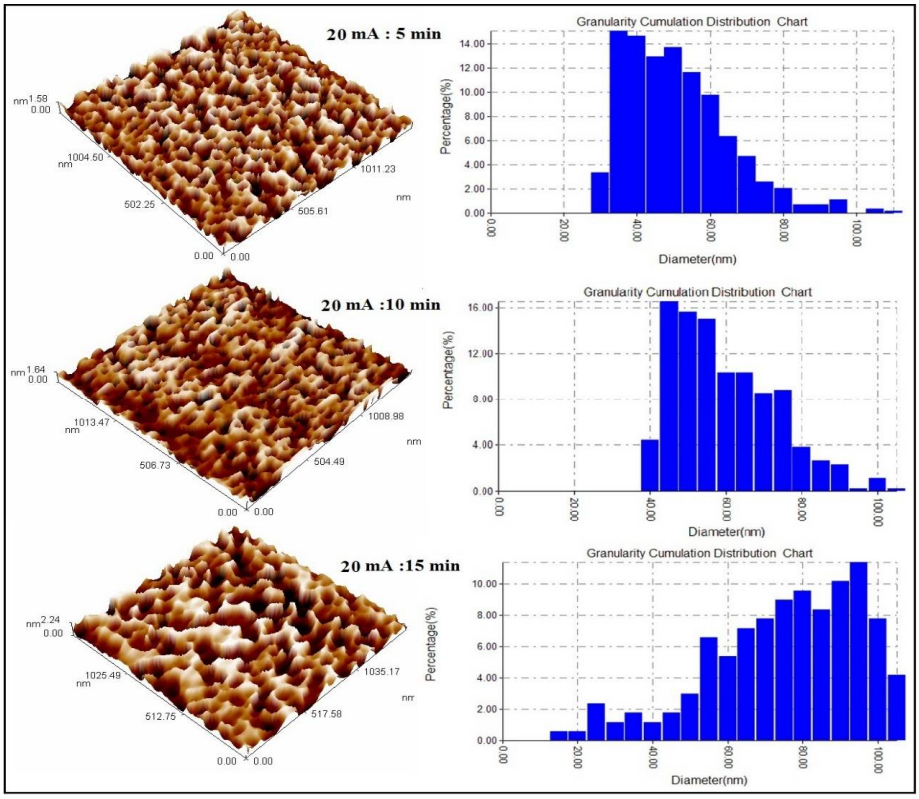


**Fig. 3: Atomic Force Microscopy of porous silicon at current 20 mA and different etching time (5, 10 and 15) min**

**Table 3: AFM parameters for porous silicon wafer at constant current 20mA and different etching time (5, 10, 15) min.**

| Time (min) | Ave. diameter (nm) | Roughness average (nm) | Root mean square (nm) |
|---|---|---|---|
| **5** | 48.63 | 0.402 | 0.464 |
| **10** | 57.55 | 0.386 | 0.445 |
| **15** | 72.54 | 0.581 | 0.665 |

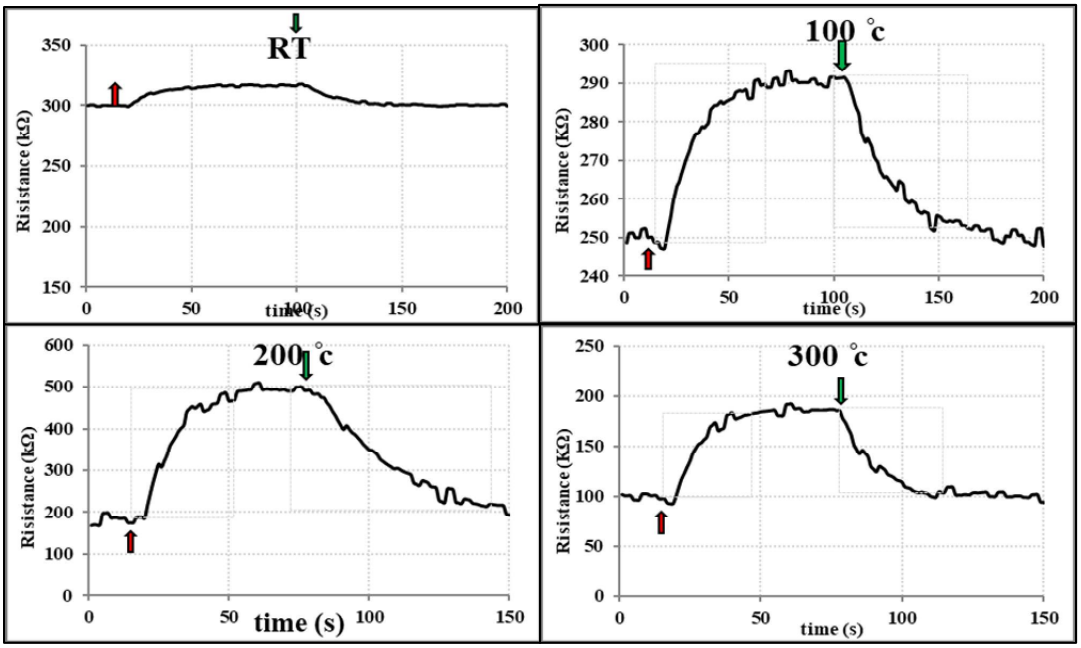


**Fig. 4: Resistance as a function of time at different operating temperatures for $In_2O_3$ /PS at etching time 15 min for $NO_2$ gas.**

**Table 4: Response time, recovery time and sensitivity%$In_2O_3$/Si at etching time of 15min for $NO_2$ gas.**

| Sample | Operating Temp.(°C) | S% | Res. time(s) | Rec. time(s) |
|---|---|---|---|---|
| **$In_2O_3$/Si** | 30 | 5.6 | 30 | 30 |
| | 100 | 16.25 | 35 | 70 |
| | **200** | **155** | **32** | **70** |
| | 300 | 86.6 | 30 | 35 |

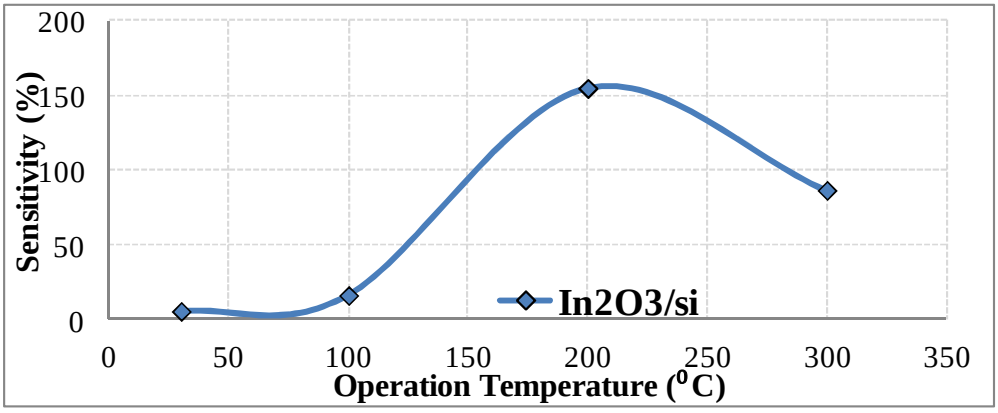


**Fig.5.Sensitivity as a function of operating temperature for $In_2O_3$ /PS at etching current 15min for $NO_2$gas**

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RESEARCH ARTICLE

# Self Health Care Model of Monks with Hypertension in Bangkok, Thailand

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## ABSTRACT

This research has the objective of studying the behavior in health self-care of the monks with hypertension, in Bangkok. The study is done from documents, in-depth interviews with 40 monks of hypertension and those who tend to the monks.There are 20 main informants.The findings from the studies are the Self-care behavior consisted of 2 main components: Self-care in Promoting Health which is exercise, nutritional health care, abstains from addictive things, emotional control and environmental health. Self-care component of Protection from Illness is self-protection and protection in cooperation with the others. The model of self-care of the hypertensive monks is the model of power driving force of self-care quality, consisted of 4 components: Assessment of the conditions, the changing of the behavior, leading to serious practice. Power of self-care component is analysis of cause and the effect of the actions, having the goal and motivation, to have good practice guidelines from the learning and to have self-control.Foundational capabilities for self-care is, awareness of having good health, managing time effectively.Reducing the risk factors in failing to control oneself component is, to have self-discipline, the avoid, denial the negative effect to the health, and protecting through cooperation with the others.

**Keywords:** Self-Care Behavior, monks with Hypertension

## INTRODUCTION

The unlimited increasing technology advancement which provides conveniences for the living of today, came with the silent threat to the health of the people in the society (The Support and Coordination Committee of Good Buddhist Village Project Project, 2013) and also having, unavoidably impact to the monks (Mintra Sararak, Wararat Sanwalee and Wilas Khampaengsee, 2017: Pattama Suphunnakul, Watcharee Srithong, Srithong, 2015) That the

worst silent threat of illness to the monks is the hypertension illness (Prakrusuvithanpatthanabandit, Thawalrat Daengharn, Sarunya Vapuchavitee, 2015; Mintra Sararak, Wararat Sanwalee and Wilas Khampaengsee, 2017)

Hypertension is caused by the blood pressure from the heart into the artery. In the condition of narrow artery or due to the contraction of small blood vessels throughout the body, the blood that flows through these blood vessels is slow and less, causing the heart to pump harder to have more blood to flow, to feed the body. The patient with hypertension is either with Primary Hypertension or Essential Hypertension or Secondary Hypertension. Hypertension is a catalyst for the formation of arteriosclerosis by weakening arterial walls which is due to the increased blood pressure passing through blood vessels. This is also a risk factor of having a Stroke (Janjiraporn Wichai, Saisamorn Poldongnok, Kittisak Sawanyavisut, 2015).The monks are in the most risk groups of having hypertension. This is due to the cause of behavior, such as, drinking coffee more than 6 cups per day, drinking more than 2 bottles per day of the energy drink, smoking cigarettes, in general, at a high rate, eating high fat food or very salty while the rate of exercise is low, including lack of proper health care (Chantima Ruekruenrit and group, 2010: Sawatsri S and Kaewpan W., 2007; Kungskulniti N, Charoenca N, Kengganpanich T, et al., 2012; Mintra Sararak, Wararat Sanwalee and Wilas Khampaengsee, 2017).

The best solution of good health is to adjust the health balance to be at the center of the people, the family, community and the society. The best guideline is to give the importance of health self-care which is food control, exercise, correct use of the medicine and self-care (Orem, 2001; Suriya Sang, 2013). Especially the monks must give the importance of self-care, because from the study it is found that after the monks have been discharged from the hospital and continued to have health care attended to, by other people, it is found that the health condition did not improve (Institution of Health System Research, 2010). But from the study it is also found (Sritisan, Patsarawalai et al, 2013) that the levels of education have an effect to the self-care uncontrollable behavior of the hypertensive patient. Therefore, in solving the problem of hypertensive monks, it should also be changed to self-care method (Patayaporn Laloon, 2011). The Department of Medical Services has campaigned for the monks to have health care for sometimes already (Office of the Crown Property, 2017), together with encouraging the practices of physical therapy and exercises for the monks (Department of Medicine, 2017). Therefore, the researcher is of the opinion that there should be a study of a model of self-care for the hypertensive monks in Bangkok, Thailand. To be useful in developing the model of self-care of hypertensive monks in the other provinces further. Apart from this, the findings from the study will be beneficial to the concerned offices, using the information obtained in planning to develop the solving of health problems of the monks, in respond to the expectation of the society and the needs of Thailand.

### Objectives

The objectives of the research are as followed:

- To study the self-care behavior of the hypertensive monks in Bangkok, according to the context of the Thai society, in respond to the campaign to keep monks healthy and to support the practices of physical therapy and exercises.

- To study the model of self-care of hypertensive monks in Bangkok, according to the best guideline in solving the health problems, which is the guideline on adjusting the health balance to be at the center of the people, in order to have healthy Thai monks for a longer period of living time and sustainable.

## METHODOLOGY

This is a qualitative research. The research was conduct by the study of documents and in-depth interviews with 40 persons: 1) Hypertensive monks who have been admitted and have gone through the prescribed health-care

activities in a campaign by the Medical Department, the Ministry of Public Health for 1 year already 2) With the people who tend to the hypertensive monks 3) With the people involving in the campaign of health-care activities for the monks.The 20 main informants are from Snowball method, tools used in collecting the information are analysis papers, interview forms, voice recorder and the researcher self. The information obtained is by triangular validation and further analyzed. The research was done during June 2017-March 2018.

## RESULTS

### 1.Self-care behavior of hypertensive monks in Bangkok

The 3 main groups of informants are the hypertensive monks, people involved in tending to the monks and those involved in the health-care activity campaign for the monks. All, are of the consistent opinion that in the past before the implementation of the campaign of health-care activities for the monks, the monks did not have any motivation in self-care, did not see the importance of health-care, lack of regularity medication, problems with physical movements, no exercise, and not being careful concerning food intake. As for the care takers who are relatives, living near the temples, would only inquire of the welfare more than would be really taking care of the health and there is little health-care among the monks who tend to one another,

With the concept of campaigning health-care activities for the monks, there are a number of hypertensive monks in Bangkok who joined the activities of self-health-care.This group of monks have a self-care behavior change to lessen the hypertension. Having the monks who look after one another, relatives and other non-relative persons who live nearby the temple, joining in the health promotion activities. By encouraging the matter concerning health-care, together with the people, society in the communities, have cooperated by not giving alms of food or things, which would be harmful to the health such as, cigarettes, energy drinks, to the hypertensive monks.

The self-care behavior of hypertensive monks, being able to control the level of blood pressure not to be higher than the normal, consisted of 2 main components of Self-Care for Health Promotion and Self-Care to Protect from Illness.

### Self-Care for Health Promotion

The important points in Self-Care for Health Promotion are:

1. Exercise, according to the monk's hood duties, such as, walk to receive alms, meditated walks, sweeping the temple courtyard, cleaning the room, etc. The suitable exercise according to the precepts, focusing on slow movements such as, swinging the arms, the traditional hermit gymnastic movements, and muscle stretching.

2. Nutrition, is to practice according to the teaching of Buddha which is to have the balance of consumption, no special foods, eating only sufficiently to survive, sufficiently for the physical need to prevent illness and sufficiently to live the monkhood life. Apart from this, must select to eat food which is not harmful to the health, eating less sweet-fatty-salty food or meat. Instead, should eat fish and fruit, and to eat at regular meal times.

3. Abstain from addicting is to avoid or abstain from any addictive things which is the cause of a decline in physical wellbeing among the monks such as, direct harm and effect to the body is cigarette or soda drinks, energy drinks where there are high amount of caffeine and sugar, and coffee where there is a high amount of sugar, fat and caffeine.

4. Controlling the emotion or to have a peaceful mind, to be calm, is to relief the suffering from illness and strengthening the mind to help in health-care, by the practices of the monkhood such as, praying, meditation, meditated walks.

5. Creating Environmental Health is to have suitable environment around the temples and the living area. This is an important basis of having a good, healthy environment for the healthy living, everything is in order, not keeping unnecessary things in the living quarters.

### Self-Care to Protect from Illness

The important self-care to protect from illness is as followed:

1. Self-care to protect from illness is to be able to control oneself, protecting from the risk factors of increasing the blood pressure and it is also a basis to lower the high blood pressure to the normal condition. The important control is to be able to control the food consumption, control the weight, control emotions, control from becoming addictive, and control oneself in doing the exercises, together with controlling the regularity of taking the medication as prescribed. Also, to control oneself in having sufficient sleep and to protect oneself from contacting other illness.

2. Protecting illness through cooperation with the others. is the protection which needs the aid from the other people such as, in physical therapy, checking the blood pressure in details from the medical officer, including providing information for those who are responsible to the donations and alms receiving things, especially food which is sweet-fatty-salty, additive drinks and cigarettes. However, even though there are such harmful things given in alms, after receiving alms, must have the discipline to select the health hazards and take them out of reach immediately.

Therefore, the important points in the 2 components of self-care behaviors are in self-care of promoting health by exercise and control nutritious food, abstain from being addictive, control emotion and having healthy environment, which will lead to a good self-care in protecting oneself from illness.

**2.Model of self-care of hypertensive monks**

The 3 groups of main informants are hypertensive monks, those tending to the monks, and those involved in the campaign of health-care activities for the monks. They are all of a consistent opinion that the hypertensive monks participated in the campaign of self-care activities, have practiced self-care in the same direction or having the same model of self-care.

The model of self-care of hypertensive monks is the model of driving force of self-care quality, consisted of 4 components: Capabilities for self-care operation, Power for self-care, Foundational capabilities for self-care and Capabilities in lower the level of risk in failing to control oneself, would lead to a success in sustainable self-care.

1. Capabilities for self-care operation, the hypertensive monks must have 3 important capabilities for self-care operation: 1) Assessment of the condition, abnormality of the body during and after exercise, assessment of the capabilities in operating activities in different conditions 2) Changing of Behavior, is the living behavior, health assault behavior, exercise behavior 3) Adherence to serious practice which is the practice in the normal condition and during the hypertensive condition, living style which does not harm the health and regular exercises.

2. Power of self-care of hypertensive monks must have 4 important power capabilities: 1) Analysis the reasons and results of the actions concerning the condition of having hypertension, factors or the cause of higher blood pressure or food which effects the high blood pressure 2) Having a goal and motivation concerning lowering blood pressure, exchanging the knowledge with those who have achieved, and to become a good role model 3) Having a good guideline from the knowledge learned concerning the correct use of the medication or the methods of taking medication, resulted from the self-development and the changes in behavior 4) Controlling oneself concerning correct use of prescribed amount of medication and at stated time, regular practice of health-care to have a good health.

3. Foundational capabilities for self-care, the hypertensive monks must have at least 2 important capabilities: 1) Awareness in obtaining good health by taking an interest in the safety of food, assessing information on health, learning of self-care, learning about self-health, obtaining advises or inquiries on health-care 2) Managing the time efficiently concerning operating activities within the time limited. Moving oneself around instead of staying still in the temple, having the skills of practicing self-care activities, or pursuit for what is beneficial to the health.

4. Lower the risk of failing to control oneself, hypertensive monks must be able to reduce at least 3 risk factors: 1) Having self-discipline concerning medication, food, exercise, sleep 2) Avoid/denial negative effect to health concerning type of food, usage of addictive things, reduce stress from illness 3) Protecting health by cooperating with the others concerning keeping doctor's appointment, control over nutrition food, control of emotion and to have a good environmental health.

## DISCUSSION

From the study of self-care behavior of hypertensive monks and the model of self-care of hypertensive monks in Bangkok, it is found that they are in consistent with the Self-Care Agency, theory of Orem (2001). That a person can develop self-care in 3 levels: Capabilities for self-care operation, Power capabilities for self-care, Foundational capabilities for self-care and in consistent with the concept of Good Health Component of Ministry of Public Health, Department of Health, Bureau of Health Promotion, 2010.Having the feature of integrated concept and theory, from the basic knowledge concerning the self-care needs, having good attitude of good health and having the skills in performing the self-care activities (Tanasak Potranan, Taweesak Kasipol, Chadapa Prasertson, 2017).

## CONCLUSION

Self-care behavior of hypertensive monks in Bangkok consisted of 2 main components: Self-Care by Promoting Health and Self-Care in Protecting from Illness where they are related to one another with continuous practices, that it becomes the supporting behavior from the people, communities and society. Together with having the regular improvement in self-care which is the reason for hypertensive monks to be healthy, they are able to control the blood pressure to be at a correct level. The details are shown in Diagram 1.

Model of self-care of hypertensive monks is the model of driving force of self-care quality, consisted of 4 components: Environment, communities and society, supporting the monks to be aware of the harm from hypertension, which is the basis characteristic of self-care and the component of reducing the risk of failing to control oneself by not giving things which could be harmful to the healthy condition of the monks. If the 2 components are to be practiced as behaviors, they will result in becoming a capability force in self-care and the capabilities in self-care lead to having a good health which in turns, becomes the quality of self-care. The details are shown in Diagram 2.

**Recommendations**

From the study it is found that the success of self-care behavior of hypertensive monks and the success of self-care model of hypertensive monks are from the supports from the people sectors, communities and society.Therefore, the concerned units in planning the development, to solve the problems of the monks' health sustainably, a basis should be created for the people's sector, together with a campaign of health-care for the monks.

From the study it is found that the power- drive model of quality of self-care of hypertensive monks, consisted of 4 components: Capabilities in performing self-care, power of self-care, basic characteristic should be a further study of other factors in a quantitative research, to be able to confirm the suitable model.

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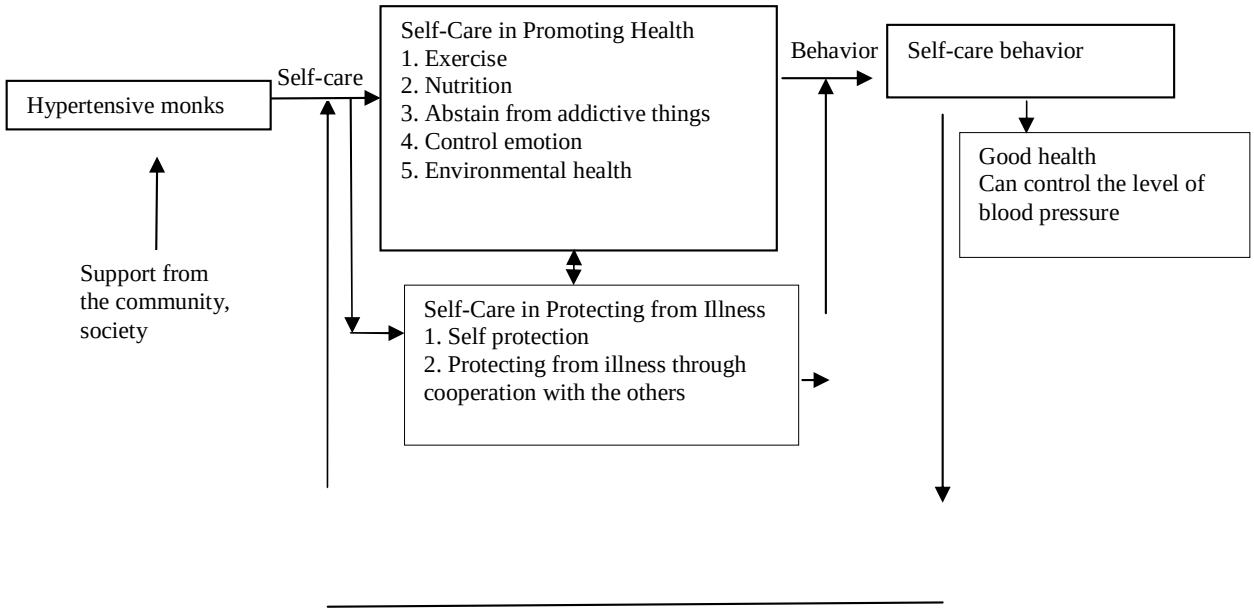


**Diagram 1.Shows the Self-Care behavior of Hypertensive Monks**

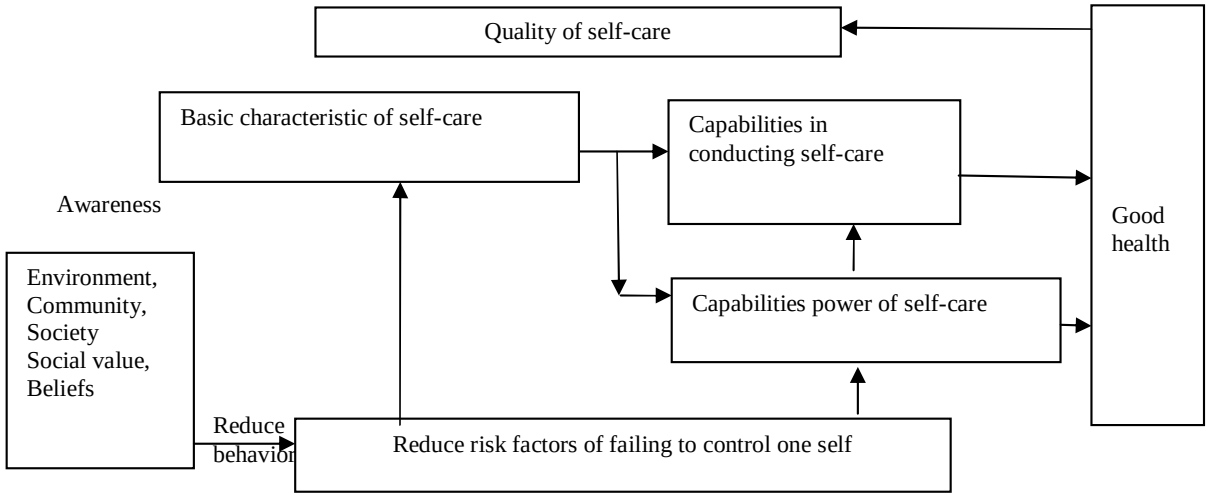


**Diagram 2.Shows the Model of quality force in self-care**

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RESEARCH ARTICLE

# The Development of a Rabies Free Area Model in Bangkok, Thailand

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## ABSTRACT

This research has an objective to study the human's behavior controlling the dogs and the controlling of the spreading of rabies.To study the problematic conditions, raised by declaration of temporarily Rabies Free Area and to study the development of Rabies Free Area model in Bangkok. The study was done by in-depth interviews with 78 dog-owners, stray dog controllers, victims of dog's bites and those involved in setting up the Rabies Free Area in Bangkok and 45 main informants. It is found from the study that 1) Human's behavior on controlling the numbers of dogs depends on the behavior of dog's owner, the responsibilities of taking care of the dogs, the habit of feeding the dogs in public areas. While the behavior of being able to control rabies depends on the vaccination of the dogs, providing information when bitten by dogs, insufficient vaccine and cooperation from the society sectors.2) Problematic conditions from the declaration of temporarily Rabies Free Area are: Problems of implementing the measures controlling dog's care, allowing the non-vaccinated dogs into the area, controlling new born dogs, reducing the risk area of stray-dog's bites and the panic of the rabies outbreak 3) The development of Bangkok Rabies Free Area model, to be a concrete model of safe area locally, as a community base, which is knowledge and consciousness of being responsible, being able to access the information, ownership of the safe area, reducing the risk if being bitten by stray dogs and enforcing the law strictly with the offenders.

**Keywords:** Controlling the spread of rabies, Rabies Free Area

## INTRODUCTION

Rabies caused by contacting rabies virus, transmitted into the human or animal's body, the virus affects the nerves and the brain that it is not curable and eventually followed by death. It is one of the fetal viruses (Department of

Disease Control, 2011) and the number one disease with the highest rate of death (Mongkol Srijun, Somphorn Phornwisetsirikun, Phrutthiphon Sukporn, Pranee Rodtian, Adulsak Wijitra, 2014). It is also found that in Thailand, 53% of the patients with rabies are bitten by the dogs with owners and 47% are by the stray dogs (Prakit Srisai, Jongkon Thamiganont, Haruethai Rungrueang, Wan SingKham, 2014; Kienzle, 2007) and ended with deaths.

However, rabies could be prevented by vaccination ((Prakit Srisai, Jongkon Thamiganont, Haruethai Rungrueang, Wan SingKham, 2014; Kienzle, 2007). Therefore, those who suspect that they have been bitten by dogs or other animals with rabies, must get the vaccine immediately. From the report it is found that there is an increase of vaccinated persons not less than 500,000 cases each year, costing not less than thousand million Baht each year (Department of Disease Control, 2011). Rabies is the disease which causes enormous amount of economic loss (Wirongrong Hoonsuwan, Apirom Puanghat, 2005).

The Ministry of Public Health and the Ministry of Agriculture and Cooperatives have been the leading units in controlling the rabies (Chirapol Sintunawa, Supaporn Wacharapluesadee, Henry Wilde, Thiravat Hemachudha, 2004). That the said issues have been continuously improved and solved (Sritisan, Patsarawalai et al, 2013). In 1980, the death rate has decreased from 370 persons to 30 persons, in 2001 there were only 8 deaths and in 2014, only 7 deaths. Thus, Thailand has set up a strategic plan of eradicating rabies from Thailand by the year 2020, leading to being able to declare Rabies Free Area, according to the goals of the World Health Organization (World Health Organization: WHO) and the Office International des Epizooties(OIE). With the cooperation from the Department of Livestock, Disease Control Department, Department of Local Administration, Association of Sub-district Administration of Thailand, Association of Provincial Administration of Thailand and Municipal League Association of Thailand (Bureau of Disease Control and Prevention, Department of Livestock. 2015).

The past operation, Rabies Free Area has been satisfactory controllable, through communication with the people, providing knowledge concerning rabies (Lanthip Hearabut, Natthisa Booncharoen, 2016), and the operation of surveillance, prevention and control with participations from the locals in the communities has also been done (Warangkana Sriphuwong, etc. al., 2017). However, it is found that at the beginning of 2018, there has been an epidemic of rabies and spreading to many areas with an increasing number of deaths (Department of Livestock, 2018a). That the temporarily epidemic areas, were declared in many provinces (Department of Livestock, 2018b).Even though, the recurrence of the rabies epidemic in 2018 is controllable at a level, but if there is no improvement in the protection, there would be an impact to the operation according to the strategy plan of rabies eradication from Thailand by the year 2020. To be able to achieve the goal, it is of the researcher's opinion that there should be a study of the development of the Bangkok Rabies Free Area model, to be useful in conducting the operation according to the strategy plan of eradicating rabies from Thailand by the year 2020, and to be useful to the life and safeties of the local people in the area. Apart from this, the findings will be useful for the concerned departments and the country's need of developing the solving of the problem sustainably.

## Objectives

1. To study the human's behaviors of the people in controlling the number of dogs and controlling the spread of rabies, and the behaviors of the concerned government's officers in the Bangkok area, including the dogs with owners and stray dogs.

2. To study the problematic conditions, raised by declaration of temporarily Rabies Free Area which has an impact to setting up the local Bangkok Rabies Free Area.

3. To study the development of Bangkok Rabies Free Area model to be as a community base, in order for the Thai people to have a long, healthy life.

## METHODOLOGY

This research is a qualitative research. The study was done by in-depth interviews with 78 persons who are: 1) dog's owners and those who have been taking care of stray dogs in their own houses for at least 1 year. 2) Government officers who are controllers of the number of stray dogs and the performance of the communities 3) Victims of dog's bites within 1 year 4) Those involved in setting up of Rabies Free Area in Bangkok. And 46 main informants by Snowball method. Tools used in collecting information are interview forms, recorder and the researcher self. The information received are by triangular data validation and to be further analyzed. The research was done in March 2018.

## RESULTS

### 1. Human's behavior on controlling the number of dogs and controlling the spreading of rabies

The study of the human's behaviors on controlling the numbers of dogs and controlling the rabies, is the study which covers the dogs with owners and stray dogs, from the in-depth interviews with all the 4 groups of concerned people are: 1) dog's owners and those who have been taking care of stray dogs in their own homes for at least 1 year. 2) Victims of dog's bites 3) Those involved in setting up Bangkok Rabies Free Area 4) Government officers who are controllers of the numbers of stray dogs and controller of the operation of the concerned communities in the Bangkok area.

All the main informants are of the consistent opinion that the people in the area have a good controlling behavior concerning the numbers of the dogs. Together with having the awareness in receiving information and follow the news concerning the controlling of rabies. The details on the development are as followed:

**Human's behavior on controlling the numbers of dogs**

1. Behavior on dog's care, it is found that there is an increasing number of people with moderate or more income, who owns more purebred dogs and keeping the dogs inside the residential area with protection fencings, that there is less opportunity of the dogs being mated with the other dogs, enabling the controlling of the number of the dogs. But among the small income earning group of people, the dogs under their care, are more for watching the houses or the offices, not to be bred. Therefore, dogs are more independent, especially in the government places, educational institutions, the security guards would raise the dogs under their care loosely and freely. This way, the chance of the dogs mating among one another is high and the number of dogs cannot be controlled.

2. The responsibility in taking care of the dogs, it is found that in most high income group of people, are responsive in taking care of their dogs until they die. But with the other group of people, it is found that with the undisciplined dogs, grown unattractive dogs, ill or injured dogs, or wanting new breed of dogs or not able to look after a dog, the dogs would be taken to be abandoned, left, at public places such as, by the side of the roads, at the temples or at schools. Increasing the numbers of stray dogs and high chance of being mated among themselves, that the number of stray dogs is uncontrollable.

3. Feeding the stray dogs, lacking the responsibility, it is found that in general, the people of every levels of income, have compassion for the stray dogs. Upon seeing a stray dog begging for food, it will be fed, in the public places and those stray dogs would be protected by collaring them but not taking the responsibility of declaring ownership of the dogs. Thus, it is not possible for the government controllers or officers of the communities, to control the number of the dogs or achieve any operation concerning stray dogs.

**Behavior on controlling the spread of rabies**

1. Serious vaccination for the dogs, it is found that in the case of dogs with owners, the owners' behavior is of both, having their dogs vaccinated or vaccinated but not on time scheduled or in some cases, having their dogs revaccinated before the scheduled time. As for the stray dogs under the care of some people, they are both with being vaccinated and no vaccination, while with the other stray dogs, there is no record of being vaccinated.

2. Providing information when bitten by dogs and information on being vaccinated, it is found that the information received is not completed because when a person is bitten, either by his/her own dog or by a stray dog, he/she will go to be vaccinated at a private clinic or hospital and the information of having received the vaccine, the information about the dog is not given nor passed on to the rabies epidemic control units, except those who are treated at the government hospitals.

3. Lack of vaccine which could be from not correctly storing, problems on the quality of the vaccine or there is a case of embezzlement that there is no vaccine available at the point of service for the immediate action.

4. The main role on controlling the rabies is still under the responsibility of the government sector while the civil society, has not yet been a part of true surveillance group, which is the cause of ineffective surveillance.

## 2.Problematic conditions raised by declaration of temporarily Rabies Free Area

The study of problematic conditions raised by declaration of temporarily Rabies Free Area which has an impact to the setting up of Rabies Free Area in Bangkok. All the informants are of the consistent opinion that the problems derive from 5 important points:

1. The decision to own a dog without being aware of the responsibility of owning a suitable number of dogs which would not become a burden to oneself. Including the responsibilities of taking care of the dog all its life, the regular vaccinations or health care when injured. These are the important issues of abandoning the dogs that they become stray dogs with increasing numbers, becoming the burden for the Thai society in the near future.

2. Neglecting the vaccination of the dogs in the area, including bringing the dogs from neighboring countries to be in Bangkok area where there is already a safety controlled area and neglecting to account for the non-vaccinated dogs the people bring into the area.

3. Failure to control new born dogs, it is found that there has been an increase in the problem of accounting the new born dogs of those with owners and stray dogs which most of them are not vaccinated.

4. Failure in reducing the risk area of being bitten by stray dogs, it is found that the increasing numbers of stray dog in the market area, government places, in schools, temples or sidewalks in front of convenient stores, is due to many kind hearted people feeding those dogs. Therefore, those who frequent those areas, market, government offices, schools, temples or shop at convenient stores, are exposed much more to the risk of being bitten by those stray dogs.

5. Failure to enforce the law with the people who did not take the responsibility in controlling the numbers of the dogs and controlling the spreading of rabies, it is found that the people who is with inappropriate habit of, such as, does not take the new born dog to be vaccinated, neglecting the scheduled vaccination time for the dogs, abandoning the dogs in public places, feeding stray dogs in the public places, etc., are not

punished according to the law, that controlling the numbers of dogs and controlling the spread of rabies do not reach the planned goal.

The panic from epidemic news, forwarding misleading information or distorted information irresponsibly, it is found that uncontrolled information through media is the cause of doubtfulness, unsure, in most of the safety controlled areas.

### 3. The development of Bangkok Rabies Free Area model

The study of the development of Bangkok Rabies Free Area model, all groups of main informants are of a consistent opinion that the operation on setting up the Rabies Free Area has been successful in many issues, but there is an additional proposal of setting up the Rabies Free Area as a community base. In order to sustain the long-term health of the Thai people, setting up the Rabies Free Area as a community base for the people consisted of 5 components:

1. Providing the knowledge and create consciousness on social responsibility, must be given to the people, to be ready before taking care of pets. That there must be an understanding of the nature of the pets and must be able to take care of them all of their lives. This is the important basis of building up the readiness to participate in protecting from rabies and from becoming contagious to human beings and other animals.

2. Linking the information network on the control of contagious disease between human beings and animals, is to create a way of providing the information quickly and conveniently, such as, registration of new born dogs, information on vaccinated dogs, actions taken when bitten by dogs, seeing stray dogs, or seeing dogs being abandoned, etc. , to the local Control Unit of Contagious Disease between Human-beings and Animals. Also, enabling the Control Unit of Contagious Disease between Human-beings and Animals, to obtain the information from the other areas quickly also.

3. Creating ownership of safety area for the local people in the area is to authorize the local people to have the duties of creating their own Rabies Free Area in their own communities, such as, record the information received of any new dogs entering the area or seeing any abandoning dogs in the public area, checking and follow up on the stray dogs in the area or any unvaccinated dogs. Inform the concerned sectors if found that there is a risky action of spreading rabies in the area. This would be a part of being the owner of the community base safety area, ensuring an effective surveillance.

4. Reducing the risk of being bitten by stray dogs is the correct control of the numbers of the dogs, such as, taking the stray dogs living in public places such as at the market, government places, schools, temples or around the convenient stores, to be vaccinated, spayed and taken care of in the controlled area. Giving correct information concerning feeding the dogs in the public area, and to build a convenient, simple place to take in abandoned dogs.

5. Enforcing the law, is to be implemented strictly with the offenders such as, neglecting to take new born dogs for vaccination, not vaccinating the dogs on schedule, abandoning dogs in public places, feeding stray dogs in public places and bringing in unvaccinated dogs into the safety area, etc.

## DISCUSSION

From the study it is found that, the model of creating Bangkok Rabies Free Area as a community base, is in consistent with the concept of World Health Organization (WHO) (2013) where it focused on solving the problems of rabies by creating knowledge and having the people's consciousness of having the responsibilities. It is in consistent with the research of Noparat Inthongkam and Ing-On Preechanvinich, (2009), where it is found that, to be able to solve the

rabies problem sustainably is to have the cooperation from the civil society sector. That the people must accept that the rabies and the troubles with dogs are the problems and the responsibilities of every one.

## CONCLUSION

1. From the study of the human's behavior in controlling the numbers of dogs and the controlling of rabies, it is found that the behavior in controlling the numbers of the dogs depends on the responsibility of taking care of dogs as pets and feeding dogs in public places, which are related to the daily income.Behavior in controlling the rabies depends on the seriousness of vaccinating dogs, giving the information on being bitten and information on the vaccination, shortage of vaccine and actual participation of the civil society.

2. The study of the problematic conditions, raised from the declaration of temporarily Rabies Free Area, it is found that the problems came from the wrong decision to take care of a dog. Being unable to take such responsibility, carelessly letting the unvaccinated dogs into the area, failure in controlling the numbers of new born dogs, failure in reducing the risk area of being bitten by stray dogs, failure in enforcing the law and the panic from the epidemic news.

3. The development of Bangkok Rabies Free Area model as a community base, it is found that there are 5 factors: 1) Providing knowledge and creating consciousness in having responsibility to the society 2) Linking the information network on the contagious disease control between human beings and animals 3) Creating ownership of safety area for the local people in the area 4) Reducing the risk of being bitten by the stray dogs 5) Enforcing the law with the offenders strictly. The details are shown in Diagram 1.

### Recommendations

From the study where it is found that there should be a community base of Rabies Free Area in Bangkok, in order to be able to implement the strategy of eradicating rabies from Thailand by the year 2020 and beneficial to the safety living of the local people and the concerned units/department, there should be: 1) Providing knowledge and creating responsibility consciousness to the society 2) Linking the information network, controlling the contagious disease between human beings and animals 3) Creating ownership of the safety area for the local people 4) Reducing the risk of being bitten by stray dogs 5) Enforcing the law with the offenders strictly which will be the sustainable way of developing the problem solving.

It is found from the study that the model of setting up Rabies Frees Area as a community base in Bangkok, consisted of 5 components: Proving knowledge and creating responsibility consciousness to the people, linking the information network of the government's sector with the people, giving safety area ownership to the people, reducing the risk and enforcing the law with the offenders strictly, are all of the qualitative research, other studies of various factors should be a quantitative research, to confirm the suitable model further.

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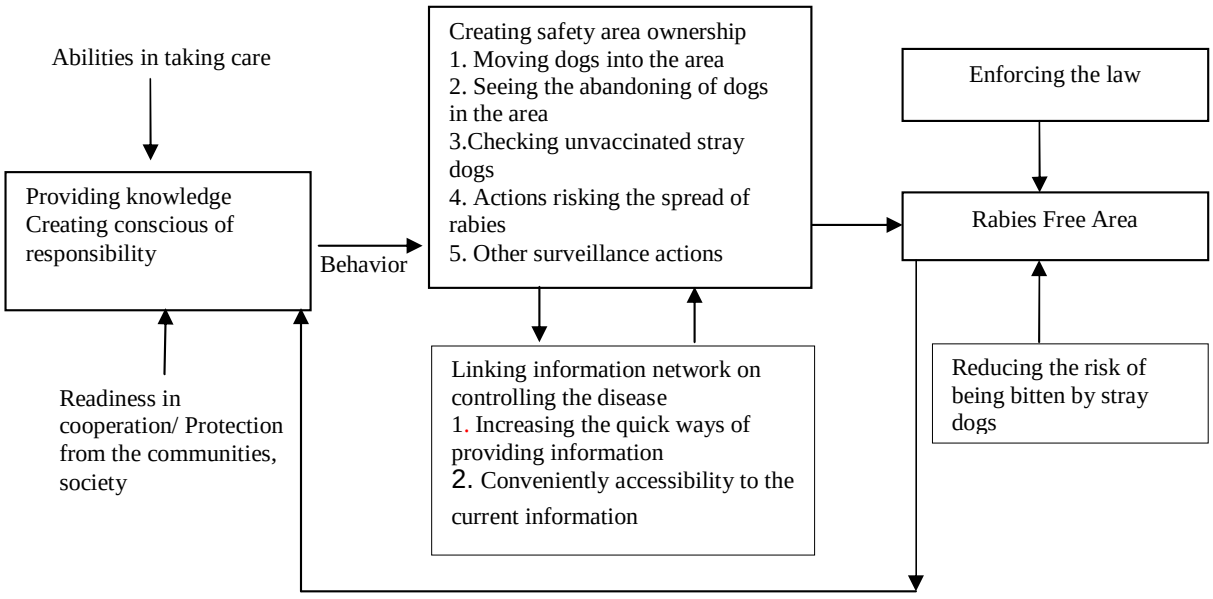


**Diagram 1.Creating Rabies Free Area model as a community base**

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RESEARCH ARTICLE

# Study on Prevalence of *Campylobacter jejuni* and *Escherichia coli* in Pork Meat Samples of Sananrak Municipality, Thailand

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## ABSTRACT

In Thailand most of the population and travellers habituated to eat more pork meat due its taste, availability. However, it may potential to contaminate with *Salmonella, Campylobacter Escherichia coli, Staphylococcus aureus*, and *Listeria* are the most common food born pathogenic microorganisms. E. coli and *Staphylococcus aureus* are routinely assessed to determine safety of food. This study aimed to assess the quality of pork meat in the study area and inculcate results of the food safety in the local community. The results, suggests that the consumption of contaminated pork meat with pathogenic organisms may pose serious threat to consumers. E. coli was observed at higher prevalences in pork meat samples collected from raw meat sellers of study area. The results of the study provided concrete evidence on transmission of the pathogenic organisms from animal to human. Those who are working in poultry farms and occupationally involved in the meat selling, transporters are involving in transmission of pathogenic organisms.

**Keywords:** Prevalence, Pork meat, Food Safety, *E. coli* and *Campylobacter jejuni*

## INTRODUCTION

Thai-population consumes meat especially pork meat due its taste, availability. Undercooked or uncooked raw pork meat prone to be contaminated with wide range organisms such as Campylobacter, Escherichia coli. Campylobacter, Escherichia coli, Staphylococcus aureus, and Listeria are the most common food poisoning pathogens (Mulder RWAW, Schlundt J 1999). Usually contamination occurs at any of the slaughtering process (Mead GC, Hudson WR and. Hiton MH 1994). Poultry products such as pork meat, and chicken meat plays an important role in poisoning

(Mulder RWAW 1999). Hence, poultry meat especially pork meat remains a challenging concern for producers, suppliers, retailers, consumers and public health professionals. pork meat and its products are often prone to expose different pathogenic organisms such as *Campylobacter, Escherichia coli, Salmonellae* these organisms transmitted to its consumers. *Campylobacter, Escherichia coli* transferred mainly carcasses or consumption of undercooked meat. Campylobacteriosis mainly occurs due to animal origins, especially pork meat plays a big role in spreading of campylobacteriosis in humans. In United States of America, *Campylobacter jejuni* found and reported as common causative of food borne infections and has been emerged as most common cause for bacterial gastroenteritis in humans (Tauxe RV 1992). Campylobacter, Escherichia coli are routinely assessed to determine microbial safety, improper hygiene in carcasses.

Globally, foodborne pathogens are the origin of diseases and may leads death in consumers. *Campylobacter*, *Escherichia coli* are the most common origin for foodborne diseases in consumers. Resistant strains infect all the age groups of consumers and wide ranges of infections are the severe consequences. Prevalence of E. coli could cause mild and severe bloody diarrhea, hemolytic uremic syndrome hemorrhagic colitis finally lead to kidney failure (Ferens WA, Hovde CJ 2011, Smith JL, Fratamico PM, Gunther NWT 2014). Prime causative of food borne diseases in humans are livestock animals and acts as reservoirs for food borne pathogens. Contamination occurs mainly through skin-to-carcass or faecal-to-carcass transfer of the pathogen during slaughter process at processing plants (Arthur TM 2010, Brichta-Harhay DM 2008, Elder RO 2000). Hence, these stages of processing of pork meat are the major concern for infection. However, during the slaughtering pork meat contaminates directly or indirectly. While processing or washing of skin and carcass.Therefore, this study aimed to investigate the prevalence of *E. coli* and *Campylobacter* microorganisms, which causes food poisoning in broiler meat as well as discusses with reference to public health importance.

## MATERIALS AND METHODS

**Samples Collection:** A total of 56 samples were collected from retail shops in Sananrak, Thanyaburi, Thailand. Samples were wrapped in a sterile polyethylene bag and identified. The collected carcase samples were transported immediately to laboratory under controlled conditions.

**Sample Preparation:** Skin and muscle samples of neck, breast and thigh skin and muscle samples include breast and thigh muscle. 10 gm samples were collected aseptically from each category followed by ICMSF (1978). Bacterial count was carried out according APHA (1992). Bacterial Isolation, *E. coli* isolated according to APHA and *campylobacter* isolation according to ISO 10272-1: 2006 (1992).

## RESULTS

Out of 56 samples collected from retail open air shops and health centers E. coli found in all the samples with various ranges. Samples from retail shops such as carcass (60%,), fresh meat (75%), hands (37.5%), knife (33.33%), cutting board (50%), and health centers (40%) respectively (table 1).The highest prevalence of E. coli strains in the study area suggests that the high concern for the public health and community health officers. Environmental sampling attributed as useful method to check whether it has any impact on the persistence and dissemination of E. coli strains. Prevalence of E. coli in meat and environmental samples reveals the detrimental effects of E. coli and importance of hygiene of food products.

Campylobacter, isolated from retail shops such as carcass (70%), fresh meat (83.33), hands (62.5%), knife (77.77%), cutting board (58.33%), and health centers (60%) of prevalence in samples collected from retail shops (table 2). The higher levels of *campylobacter* in the study area suggest that the high concern for the community and public health officers. Need to take initiatives to prevent the food poisoning pathogens and provide awareness on the importance

of hygiene of food products.Table 3 reveals the distribution of local consumers according to their symptoms and infections due to Campylobacter such as such as diarrhea (22.85%), diarrhea with blood (8.57), fever (10.48), abdominal cramps (18.10), nausea and vomiting (12.38), and unaffected (27.62%). The study indicates that 77.38% of the local consumers have been affected with different symptoms and 27.62% unaffected respectively.Table 4 reveals the distribution of local consumers according to their symptoms and infections due to E. coli such as such as diarrhea (14.29%), diarrhea with blood (5.71%), fever (12.38%), abdominal cramps, loss of appetite and mild dehydration (22.86%), malaise, nausea and vomiting (20.95%), and unaffected (23.81%). The study indicates that 76.19% of the local consumers have been affected with different symptoms and 23.81% unaffected respectively.

## DISCUSSION

The results of the study provided a concrete evidence in food pathogenic organisms (table 1, 2, 3 and 4) similar result was reported (Northcutt JK, McNeal WD, Ingram KD, Buhr RJ and Fletcher DL, 2008), whereas higher prevalence was reported (Bhandari N, Nepali DB and Paudyal S, 2013). On the other hand, lower levels of coliform counts were reported (Buhr RJ, Berrang ME and Cason JA, 2003), in breast skin, neck skin (Abu-Ruwaida, AS, Sawaya WN, Dashti BH, Murad M, 1994), breast (Gad M, 2004) and thigh muscles of chickens (Daoud JR., Farghaly RM and Maky M, 2012).

Detrimental impact of food poisoning on surrounding could be due heterogenous activities. Quantitative study is carried out by using survey method and questionnaire as instrument to collect research data. A questionnaire-based interview conducted with locals, and the local community were fabricated to comprehend the repercussions of e. coli and campylobacter infections owing to heterogenous activities of poultry producers, meat sellers, which has been revealed and evidenced that majority of the local communities were deteriorating from various health issues. Elevated levels of E. coli and Campylobacter attributed as they live in poultry products and meat. These microorganisms reside on their skin, or in the alimentary tract. Bacterial contamination occurs mainly slaughtering activities and subsequent contaminations may takes place at any of the processing stages such as production process, and washing, environment, equipment, freezing, and workers hands can cause contamination of meat and its products. Similar findings have been reported (Kotula KL, andPandya Y 1995, Georgnaras I, de Jesus A, van Zyl E and van Holy A 1997).

E. coli, is a natural inhabitant in warm blooded animals and ruminants. E. coli mainly lives intestinal tracts of ruminants and mammals. Prevalence of both the organisms reflects contamination and indicates a possible contamination of enteric pathogen. Raw or uncooked foodstuffs get contaminated either production, slaughtering or handling, processing, cross contamination (human-to-food contamination) (Russell SM and Walker JM 1997). After, slaughtering poultry carcasses scalded in a common scaling tank in retail poultry meat shops, under poor hygienic conditions such as stagnant water, excreta and or non-bactericidal temperatures etc, these conditions serve as transmitter for contamination and finally contamination disseminates to all the birds (Berrang ME, Ladely SR and Buhr RJ 2001).

The higher prevalence rates of E. coli and Campylobacter was found in the pork meat. The higher prevalence rates of Campylobacter and E. coli could be attributed to the poor hygienic conditions and improper usage of techniques for cutting, packing, and storage and predominantly affects the hygiene of meat. High prevalence rates of E. coli and Campylobacter microorganisms connected with the poor hygiene. The results provided concrete evidence on how poor hygienic practices in meat product storage, selling practice and lack of experience of butchers could affect the hygiene of raw pork meat. Hence, E. coli and Campylobacter can be spread through work environment, skin, carcasses, intestinal contents, and knives. Poultry meat often infected with Campylobacter and E. coli in the study area. Presence of pathogenic Campylobacter and E. coli microorganisms in meat shows the alarming situations in the

surroundings. Hence, this can be used as indicator organisms to detect the pathogenicity of food especially in meat and meat products (Reddy M 2017).

*Campylobacter* and *E. coli*, are most inculpated food poisoning pathogens which often found in poultry meat attributed as increasing risks of health issues. Because it can act as symbiotic organism as well as a disease producing pathogen.The prevalence of *Campylobacter* and *E. coli* in pork meat predominant due to intestinal cross-contamination, poor hygienic procedures, unorganized retail marketing and water which used for washing of hands, containers and carcasses as a whole all these could be contaminated through faeces or cross contamination.
The study revealed that raw pork meat was often contaminated with Campylobacter and E. coli in retail markets of Sananrak, Thailand. Pork meat often infected with pathogenic microorganisms through poor hygiene of carcasses, production premises, surrounding environment and consumption of undercooked poultry products.

The findings of this study suggest that the consumption of contaminated pork meat with *Campylobacter* and E. coli may pose serious threat to local residents and surrounding communities. Campylobacter and E. coli were found at higher prevalences in raw pork meat samples collected from retail and open-air market sellers of study area. It indicates transmission of the pathogenic organisms by the animal to human. Results suggests that the high prevalence of Campylobacter and E. coli in pork meat may be due to improper processing technics, poor hygienic conditions in retail shops. Further studies need to be undertake on hygiene, awareness on production and processing systems for the production of good quality pork meat to substantiate the results of the study.

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**Table1: Prevalence of E. coli from samples of retail markets and health centers in Sananrak, Thailand.**

| Source of the sample | Type of sample | Samples examined (n) | Positives (n) | Frequency (%) |
|---|---|---|---|---|
| Retail shops and open-air markets | Carcass | 10 | 6 | 60 |
| | Fresh meat | 12 | 9 | 75 |
| | Hands | 8 | 3 | 37.5 |
| | Knife | 9 | 3 | 33.33 |
| | Cutting board | 12 | 6 | 50 |
| | Health centers | 5 | 2 | 40 |

**Table 2: Prevalence of Campylobacter from samples of retail markets and health centers in Sananrak Thailand.**

| Source of the sample | Type of sample | Samples examined (n) | Positives (n) | Frequency (%) |
|---|---|---|---|---|
| Retail shops and open-air markets | Carcass | 10 | 7 | 70 |
| | Fresh meat | 12 | 10 | 83.33 |
| | Hands | 8 | 5 | 62.5 |
| | Knife | 9 | 7 | 77.77 |
| | Cutting board | 12 | 7 | 58.33 |
| | Health centers | 5 | 3 | 60 |

**Table 3: Distribution of quarry workers according to hearing problems**

| | | Frequency | Percent |
|---|---|---|---|
| Valid | Diarrhea | 24 | 22.85 |
| | Diarrhoea with blood | 9 | 8.57 |
| | Fever | 11 | 10.48 |
| | Abdominal cramps | 19 | 18.10 |
| | Nausea and vomiting | 13 | 12.38 |
| | Unaffected | 29 | 27.62 |
| Total | | 105 | 100 |

**Table 4: Distribution of local consumers according to E. coli infections and symptoms**

| | | Frequency | Percent |
|---|---|---|---|
| Valid | Diarrhea | 15 | 14.29 |
| | Diarrhoea with blood | 6 | 5.71 |
| | Fever | 13 | 12.38 |
| | Abdominal cramps, loss of appetite, mild dehydration | 24 | 22.86 |
| | Malaise, nausea and vomiting | 22 | 20.95 |
| | Unaffected | 25 | 23.81 |
| Total | | 105 | 100 |

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RESEARCH ARTICLE

# Study on Behavioral Change of Farmers in using Pesticide for Rice farming in Nakhon Nayok Province, Thailand

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## ABSTRACT

The study aimed to investigate the behavior and symptoms of farmers that grow paddy rice, to analyze factors that have relationship with behavior and work performances while using pesticide and to compare paddy rice farmers' knowledge before and after the program.The research is Quasi – Experimental Research done by Questionnaire and interview of paddy rice farmers group with sample of 52 respondents. The study results found that farmers have high risk and mostly have rash skin. Thus, when analyzed with Pearson's Correlation Coefficient, risk level that effect to health has positive relationship in the statistical significant level of 0.05 (p-value <0.05) in the behavior of using sprayed pesticide.Behavior in spraying pesticide and rinsing vegetables and fruits before eating have high positive relationship with health hazard.Thus, the behavior of chemical stained clothes changing immediately after spraying, the behavior of eating/drinking water around the working areas, the behavior of wearing chemical preventive mask while working with chemical and the behavior of hands washing before drinking water or eating food have positive relationship in high levels with health hazard. With the analysis of Paired t-test, found that after training, farmers have better behavioral changes in chemical usage that different in the statistical significant level of 0.05 (p-value <0.05) by the following training topics as fallows: Farmers must know about pests. Farmers must get acquaint to atomizer. Farmers must know about sprayer and equipment. Farmers must know techniques and proper environment in spraying chemical. Farmers must know about products. And Farmers must know about the safety topic of chemical using.

**Keywords:** Pesticide, Behavioral Change, Rice, Farmers.

## INTRODUCTION

Today, the using of pesticide by agriculturist in Thailand has increased more and more every year. Particular, in 2017, there were 148,979 tons of herbicides, 21,601 tons of insecticide, 19,923 tons of fungicide and 7,814 tons of other products, totaling 198,317 tons (Office of Agricultural Economics 2017). and the chemical hazards that effect to users and environment is also increasingly crucial and is the severe problems (Greennet 2017). Thus, due to the fact that vast chemical has been brought to use in agriculture, therefore, both governmental sectors and private have gave precedence to solve the aforementioned problem. Thailand is the country that most population do agricultures. However, in the past, agriculturists aimed to grow just for family consumption only, but due to population growth, agricultural products have been accelerated to develop for business. Pesticide, then, came to be the roles of maintaining and increasing agricultural products (Suwannakoot A 2016).

Pesticide can be dangerous to both human's health and animal. In other words, it could demolish inner organs, such as liver, kidney, lung, brain, skin, nerve system, reproductive system and eyes, depending on which way it exposes into the body and how much chemical quantity human consumed. More often that when inner organs have accumulated chemical to the limit and cannot hold on to it any more, it will expose into many form of symptoms, for example, cancer, endocrine disease, blood diseases and immune disease (Kunstadter P, Prapamontol T, Sirirojn B, Sontirat A, Tansuhaj A, Khamboonruang C 2001).

Therefore, the promoting of pesticide knowledge and prevention is very important. There are many local departments that have entered into the roles, such as District Agriculture Office, District Agricultural Officer and District Health Promotion Hospital, especially the District Health Promotion Hospital has processed the role of occupational health to community's labor group (Bureau of Occupational and Environmental Diseases 2013). Thus, the hospital has promoted for primary health care service unit to stay alert, to prevent, to control disease and health hazard that will affect to agriculturists and other labors in the community. The District Health Promotion Hospital of Baan Klong 23, North Shore, Moo 3, Baan Klong 23, Si Sa Krabue Sub-district, Ongkharak District, Nakhorn Nayok province has participated in this process too. From primary survey, found that most population earned their living by agricultures, especially 95% of the populations are doing farms. The total of 243 sample group farmers are doing farms that consisted of 2 types of farmed area, they are in-season-rice field (1farming per year) and the out-of-season (3 farming per year) rice field. Thus, from the statistical collection indicated that the using of chemical in insects controlling and protection of farmers is in hazardous levels and found that in the year 2014 and 2015 respectively, the total of 213 and 109 farmers are in the danger levels (Baan Klong health promoting hospital (2017). However, from the statistical data, showed that numbers of paddy rice farmers still using vast quantity of pesticide and the levels of their chemical contamination are at risk and unsafe and that is directly affected to health (Plungkamol S 2015).

Therefore, researcher realized to the danger in using chemical of farmers and interested to arrange for farmers' Behavioral Change in Using Pesticide Program. The program will advise farmers of the correct methods in using chemical pesticide and will create awareness and protection of pesticide exposure to body including its danger that will affect to health.

### Objectives

- To study Behaviors, Symptoms of paddy rice farmers from the contacting of pesticide.
- To analyze factors that related to Relationship of Behavior and Working Performances while Using Pesticide.
- To Compare Paddy Rice Farmers' Knowledge before and after program.

## METHODOLOGY

The research topic of Behavioral Change in Using Pesticide of Farmers of Moo 3, Baan Klong 23, Si Sa Krabue Sub-district, Ongkharak District, Nakhon Nayok province used Quasi - Experimental Research approach with the One Group Pre-test and Post-test Design that emphasized on the tested process of only one group of farmers that had been screened for chemical blood. The total of 52 farmers has been reported from public health academician of the District Health Promotion Hospital, Baan Klong 23, North Shore that they had received Cholinesterase Enzyme level, tested and found with risk and unsafe levels and the contaminated farmers are willing to participate in the Behavioral Change Program that researcher invented. Instruments used in research are Questionnaire that applied with Risk in Pesticide Contact Evaluation in Farmers' Work from the Occupational and Environmental Diseases, Ministry of Public Health (Bureau of Occupational and Environmental Diseases 2017) for the first round. Next round is to promote awareness of danger from chemical using, will demonstrate individually of the correct methods of how to behave oneself toward the using of chemical by showing the awareness video from the Plant Protection Research and Development Office, Ministry of Agriculture (The Plant Protection Research and Development Office 2017) and Cooperatives including the processing of Behavioral Evaluation in Chemical Using by Questionnaire repeatedly after using the chemical

.

**Analysis from Questionnaire** is divided into 4 parts as follows:

**Part 1** is the paddy rice farmers' personal data? General farmers' explanation is done by Descriptive Analysis, Average Value, Percentage and Standard Deviation.

**Part 2.** The Information of Pesticide Application and the Behavior while performing work is done by Descriptive Analysis, Average Value, Percentage, Standard Deviation and Pearson's correlation.

Evaluation of Farmers' Risk in Contacting of Pesticide While Working, scores and outcomes interpretation from the Questionnaire is divided into 2 descriptions.

**Description 1.** Item 16 – 21 are questions related to Risk Behavior or Unsafe Behavior.

**NO** means Risk Behavior in the Items are very little or Never, indicated Low Risk which is equal to 1 score.

**YES/OCCASIONALLY** means performed occasionally, equal to Moderate Risk, will get 2 scores.

**YES/EVERY TIME** means have risk behavior every time or most of the time, equal to High Risk, will get 3 scores.

**Description 2** . Item 22 – 29 are the questions related to Safety Behavior in order to reduce risk or prevent danger from pesticide contact.

**NO** means Safety Behavior is very little or Never performs, indicated High Risk, equal to 3 scores.

**YES/OCCASIONALLY** means perform Safety Behavior only for sometimes, indicated Moderate Risk, equal to 2 scores.

**YES /EVERYTIME** means always perform Safety Behavior or most of the times, indicated Low Risk, equal to 1 score.

**Part 3** Sickness data or disorder symptoms that happened after using or contacting with pesticide by Descriptive Analysis, Average and Percentage. Disorder symptoms are consisted of the followings:

**1) Minor Symptom Group**

Minor Symptom Group is consisted of weary, cough, breathe jam, dizzy, headache, itchy skin/dry skin/broken skin, rash skin/blister, burning pain, red eyes/irritated eyes/itchy eyes, numb, heart palpitation, half-awake sleep, irritated nose, sore throat, thirsty, sweating, tears flow, salivated and running nose.

**2) Moderate Symptom Group**

Moderate Symptom Group is exposed by blepharospasm, blurry vision, chest pain/chest feel tight, feel queasy, vomit, stomachache, diarrhea, muscular weakness, cramp, hands shake and totter.

**3) Severe Symptom Group** is exposed by epilepsy, unconscious and loses consciousness.

**Summary of Basic Health Risk Evaluation**

Outcomes interpretation is from the point of intersection of the answer from the scores of Behavioral in Chemical Work and the Sickness information or the disorder that happened after using or contacting with pesticide, details as shown in Table No 1.

**Part 4** Comparison of Behavioral Change from the Chemical Contact before and after Promotional Program of Knowledge to Farmers.Tested in Paired t-test.

## RESULTS AND DISCUSSION

Researcher collected general personal data of paddy rice farmers by using the sample group that used to have screened chemical blood test from the District Health Promotion Hospital in cholinesterase enzyme quantity check that have risk levels and unsafe levels in the total of 52 farmers. Thus, most farmers are females 57%, male 43%, most are ages between 50-59 years which is equal to 32% and have primary education of 47%. Yet, main occupation of farmers is growing paddy rice and lands are owned by farmers in the area size of 16-30 rais which is equal to 47%. Farmers' average income per year is 105,394 Baht and mostly farmers have been using chemical during 16-30 years which is equal to 55%. Expenditure on pesticide is in the average of 4,508 Baht per year depending on how severe of diseases and pests, which it is in line with Krathiya Saengpakdee's research that farmers use chemicals because pests are very disturbing (Bureau of Occupational and Environmental Diseases 2017, The Plant Protection Research and Development Office 2017, Sangpakdee K, Silprasit k, Peangthai D, Khwaiphan W, Siriyan S, & Kroeksakul P 2014). Most pesticide usage are Glyphosate-Isopropylammonuim, Butachlor and Bispyribac-Sodium which is equal to 77%, followed by Cyhalofop-Butyl+Penoxsulam which is equal to 23%. Mostly, each of pesticide brands will be used in rotation, such as Carbosulfal, Cypermethrin, Abamectin, Chlopyrifos.Pesticides that used when there is epidemic are Carbendazim and Mancozeb. Frequency in chemical usages, mostly will be using 4 times per 1 production which is consisted of pesticide that kill insect and plant diseases. Thus, the average duration in spraying chemical is 3 hours per each time and every farmer uses sprayer in the backpack style. Besides, research found that every farmer that uses pesticide, sprayed the chemical by themselves. Farmers learnt about chemical information by friends who worked as farmers and followed by the District Agricultural Officers which is equal to 51% and 43% respectively.

From the study of the above mentioned general information, researcher has evaluated Risk of Unsafe Behavior while farmers contacted with chemical, found that every time when all farmers use chemical to eliminate insects, their behavior is categorized as High Unsafe Behavior. Thus, for other aspects, such as the using of leakage chemical container to spray, smoking while working, eating at the working areas and drink alcohol at working areas are categorized in the low level of Unsafe Behavior as shown in Table No. 2.

Still, the study found that farmers that have High Safety Behavior while contacting with chemical are farmers who have behavior of washing hands each time before break for water drinking or eating food. Thus, Moderate Safety Behavior are the behavioral characteristic of wearing rubber gloves and chemical protective boots, the changing of clothes that have chemical stains immediately after spraying chemical, cleaning the body by taking bath right away

after spraying chemical and rinse vegetables and fruits each time before eating. The Low Safety of Behavior is to take bath or washes skin that contacted with chemical immediately each time when clothing is soaked with chemical, read the new chemical labels before using and wearing mask to protect chemical respectively as shown in Table No. 3.Mostly, symptom that is found in minor symptom which is equal to 67.3%, symptoms are expressed by the followings, dizzy, itchy skin, rash skin, burning pain, red eyes or itchy eyes, numb, irritate nose, sore throat and thirsty. The symptom that found most is the itchy skin which is equal to 31.4% of the minor symptom.

Followed by, the moderate symptom group which is equal to 26.9%. It showed by the following symptoms, such as blepharospasm, blurry eyes, chest pain/feel chest tight, queasy, vomit, stomachache, cramp and hands shake. The most symptom found is queasy and vomit which is equal to 35.7% of the moderate symptom. Lastly, is the severe symptom group which is equal to 5.8% that consisted of unconscious symptom and lose consciousness. The symptom of unconscious is found the most in 66.7% of the severe symptom. When analyzed the levels of Health Risk from the using or contact with the herbicide, found that most farmers are having high risk which is equal to 71.2%, followed by quite high risk which is equal to 15.4% and have very high risk which is equal to 13.5%.

In addition, when analyzed for Behavioral Risk Values Factor of the using or contact of pesticide that related to Risk Level that effect to Health with the Pearson's Correlation Coefficient, found that Risk Level that effect to Health has positive relationship in the statistical significant level of 0.05 with the sprayed chemical behavior of r=0.771, the sprayed chemical of insecticide behavior of r=0.771, the behavior of clothes with chemical stains change immediately after finish spraying of r=0.826, the behavior of eating food/drinking water around working areas of r=0.842, the behavior of using mask to protect from chemical while working with chemical of r=0.853, the behavior of hands washing before break for drinking water or eating food of r=0.874 and the behavior of rinsing vegetables and fruits before eating of r=0.784 as shown by Table No. 4.

When interpreted the coefficient that obtained from the calculation, found that the Behavior of Using Insecticide Spray, the Behavior of Using Pesticide Spray and the Behavior of Rinsing Vegetables and Fruits before Eating have Relationship Levels quite high. On the contrary, the Behavior of stained Chemical Clothes Changing Immediately After Spray, the Behavior of Eating Food/Drinking Water, the Behavior of Using Protective Mask while Working with Chemical and the Behavior of Hands Washing before Break for Water Drinking or Eating Food have the relationship in high levels.

When researcher acknowledged about Risk Behavior of Farmers, therefore, researcher arranged the Program of Behavioral Change in Using of Pesticide in order to create awareness and to reduce risk that may affect to farmers' health. Thus, researcher arranged for training program of the Plant Protection Research and Development Office in the technical topics of using pesticide that has the followings issues, they are 1. Farmers must know about insects. 2. Farmers must get acquaint to atomizer. 3. Farmers must know about sprayer and equipment. 4. Farmers must know about techniques and proper environment in spraying chemical. 5. Farmers must know about products. And 6. Farmers must know about the safety topic of chemical using. After the training, researcher will measure the behavior in using pesticide of farmers by the next spray period which will be specified by farmers. Measurement will be done by Questionnaire and Interview of Risk Evaluation in Farmers' Work and Pesticide Contact.

When compare before and after the knowledge promotional training to farmers in using of pesticide, found that after training, farmers' behavior in using chemical is better and different in the statistical significant level of 0.05 (p-value <0.05) as shown by Table No. 5.

## CONCLUSION

The study found that most farmers that grow paddy rice is females that have ages between 50-59 years, have primary educational level, their major occupation is growing paddy rice and owned their own land in the size around 16-30 rais. Thus, they have average incomes of 105,394 baht per year. Thus, all famers are using pesticide to eliminate insects, pests, weeds and plant diseases. Mostly, duration in using chemical is between 16-30 years with the chemical expenditures averagely of 4,508 baht per year, depending on the diseases severity and insects. Yet, the popular pesticide is Glyphosate-Isopropylammonuim, Butachlor, Bispyribac-Sodium and Cyhalofop-Butyl+Penoxsulam. However, mostly the using of pesticide brands, will use each chemical brand in rotation, such as the using of Carbosulfal, Cypermethrin, Abamectin and Chlopyrifos. Moreover, there are chemicals such as, Carbendazim and Mancozeb to eliminate plant diseases whenever find that there is epidemic. Frequency in using chemical is 4 times per one round of production. All farmers are the ones who spray pesticide and the duration of spraying chemical is averagely 3 hours per each time depending on manpower. Most chemical knowledge and information are from friends who are farmers, followed by the District Cultural Officers.

When evaluated Risk Level towards Health from the Using or the Contact of Pesticide, found that most farmers are in high risk. Behavioral factors that have relationship with Farmers' Risk Level are the Behavior in Using Insecticide Spray, the Behavior in Using Pesticide Spray and the Behavior of Rinsing Vegetables and Fruits before eating have the relationship quite high. For the Behavior of Chemical Stained Clothes Changing immediately after spray, the Behavior of Eating Food/Drinking Water around the working areas, the Behavior in Using the Protective Mask while working and the Behavior of Hands Washing before Break to drink water or eat food, have relationship in the high levels. When consider sickness data or disorders, found that most are having minor symptoms, such as dizzy, itchy skin, rash skin, burning pain, red eyes or itchy eyes, numb, irritated nose, sore throat and thirsty.

The arrangement of Promotional Program for knowledge dissemination to farmers is to reduce Risk Behavior of Farmers in the Using or Contacting Pesticide safely. And with the guidance from the Plant Protection Research and Development Office that consisted of the following topics, they are 1. Farmers should know about insects. 2. Farmers must get acquaint with atomizer. 3. Farmers must know about the sprayer and equipment. 4. Farmers must know about proper techniques and environment when spraying. 5. Farmers must know about the products, and 6. Farmers must know about the topic of safety in using chemical. Eventually, found that after the training, farmers have better behavior in using chemical.

### Recommendations

This research has received cooperation of academic aspects from the District Health Promotion Hospital of Baan Klong 23, Si Sa Krabue Sub-District, Ongkharak District, Nakhon Nayok Province. The studied results found that, the Behavioral Change Program in Chemical Contact educated farmers with the correct knowledge in the topic of chemical handling and farmers should screen for cholinesterase enzyme level for the next farming year. Last but not least, District Agricultural Officers should support for the acknowledgement of this successful program in order to be able to advise and give suggestion of the topic of chemical operation to new generation of farmers in future continually.

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**Table 1. Summary of Basic Health Risk Evaluation**

| **Symptoms After Contacting with Herbicide** | **Risk Levels to Health of the Using & Contacting of Herbicide** | | |
|---|---|---|---|
| | **14-20 Scores** | **21-28 Scores** | **29-42 Scores** |
| **No Symptom** | Low Risk | Moderate Risk | Risk Quite High |
| **One Level of Minor Symptom** | Moderate Risk | Risk Quite High | High Risk |
| **More than 1 Symptom of Moderate Symptom** | Risk Quite High | High Risk | High Risk |
| **More than 1 Symptom of Severe Symptom** | High Risk | High Risk | High Risk |

**Table 2. Illustrates Unsafe Behavioral Characteristics while Contacting Chemical**

| **Unsafe Behavioral Characteristics while Contacting Chemical** | **Total of Farmers (Risk in Percentage)** | | | **Scores Level** | | **Interpretation** |
|---|---|---|---|---|---|---|
| | **Not Using at All N (Percentage)** | **Occasiona-lly Use N (Percentage)** | **Use Every Time N (Percentage)** | Mean | S.D. | |
| Using Insecticide Spray | - | 6(11.5) | 46(88.5) | 2.88 | 0.32 | High Unsafe Behavior[3] |
| Using Pesticide Spray | - | 6(11.5) | 46(88.5) | 2.88 | 0.32 | High Unsafe Behavior[3] |
| Leakage Container in Spray | 52(100) | - | - | 1 | 0 | Low Unsafe Behavior[1] |

| | | | | | | |
|---|---|---|---|---|---|---|
| Cigarette/Tobacco Smoking while Working | 52(100) | - | - | 1 | 0 | Low Unsafe Behavior[1] |
| Eating Food/Drinking Water at Working Areas | 42(80.8) | 10(19.2) | - | 1.19 | 0.40 | Low Unsafe Behavior[1] |
| Drink Liquor/Beer/Alcohol at Working Areas | 52(100) | - | - | 1 | 0 | Low Unsafe Behavior[1] |

Remark: N is the Sample Group

1. Number to the Power of 3, is the average between 2.34-3.00, means farmers have High Unsafe Behavior.
2. Number to the Power of 2, is the average between 1.67-2.33, means farmers have Moderate Unsafe Behavior.
3. Number to the Power of 1, is the average value between 1.00-1.66, means farmers have Low Unsafe Behavior.

**Table 3.Illustrates the Safety Behavioral Characteristics while Contacting Chemical**

| **Safety Behavioral Characteristic While Contacting Chemical** | **Total of Farmers (Risk Percentage)** | | | **Scores Level** | | **Interpretation** |
|---|---|---|---|---|---|---|
| | **Not Performing at all N (Percentage)** | **Occasionally Perform N (Percentage)** | **Always Perform N (Percentage)** | Mean | S.D. | |
| Read new container's label before using the chemical. | - | 13(25.00) | 39(75.00) | 2.75 | 0.44 | Low Safety Behavior[3] |
| Wearing rubber gloves, boots and protective shoes from chemical. | - | 41(78.80) | 11(21.20) | 2.21 | 0.41 | Moderate Safety Behavior[2] |
| Wearing mask to protect chemical while working with chemical. | - | 19(36.50) | 33(63.50) | 2.63 | 0.49 | Low Safety Behavior[3] |
| Taking bath or washe that contacted with c immediately each tin clothes are soaked w chemical. | - | 4(7.70) | 48(92.30) | 2.92 | 0.27 | Low Safety Behavior[3] |
| Always wash hand before drinking water or eating food. | 3(5.8) | 24(46.2) | 25((48.1) | 1.58 | 0.61 | High Safety Behavior[1] |

| Safety Behavioral Characteristic While Contacting Chemical | Total of Farmers (Risk Percentage) | | | Scores Level | | Interpretation |
|---|---|---|---|---|---|---|
| | Not Performing at all N (Percentage) | Occasionally Perform N (Percentage) | Always Perform N (Percentage) | Mean | S.D. | |
| Always rinse vegetables and fruits before eating. | 7(13.5) | 29(55.8) | 16(30.8) | 1.83 | 0.65 | Moderate Safety Behavior[2] |
| After spraying chemical, change clothing that has chemical stains immediately. | - | 43(82.7) | 9(17.3) | 2.17 | 0.38 | Moderate Safety Behavior[2] |
| Immediately clean up body by taking bath after finish spray. | - | 45(86.5) | 7(13.5) | 2.13 | 0.35 | Moderate Safety Behavior[2] |

Remark: N is the Sample Group.

1.Number to the Power of 3, is the average between 2.34-3.00, means farmers have low safety behavior.

2.Number to the Power of 2, is the average between 1.67-2.33, means farmers have moderate safety behavior.

3.Number to the Power of 1, is the average between 1.00-1.66, means farmers have high safety behavior.

**Table 4.Illustrates Relationship Values between Risk Factors Behavior of the Using or Contacting of Pesticide with the Risk Level that Effect to Health (n=52).**

| Factor | Pearson Correlation | Sig. (2-tailed) |
|---|---|---|
| | Risk Level that Effect to Health | |
| 1. Behavior in Using Insecticide Spray. | r=0.771* | 0.010 |
| 2. Behavior in Using Pesticide Spray. | r=0.771* | 0.010 |
| 3. Behavior in Changing Clothes that stained with chemical immediately after spray. | r=0.826* | 0.050 |
| 4. Behavior in eating food /drinking water around the working areas. | r=0.842* | 0.023 |
| 5. Behavior in Using Protective Mask while working with chemical work. | r=0.853* | 0.049 |

| | | |
|---|---|---|
| 6. Behavior in Washing Hands before break for water drinking or eating food. | r=0.874* | 0.031 |
| 7. Behavior in Rinsing vegetables and fruits before eating. | r=0.784* | 0.000 |

*p<0.05

**Table 5.Compare Difference of Behavioral Average Scores of Farmers in Using or Contacting Pesticide Before and After the Training**

| Factor | N | $\overline{X}$ | S.D. | t | df | p-value |
|---|---|---|---|---|---|---|
| **Risk Behavior of Farmers in Using or Contact with Herbicide.** | | | | | | |
| Before Training | 52 | 28.19 | 1.53 | 28.34 | 51 | 0.00* |
| After Training | 52 | 19.13 | 1.56 | | | |

*p<0.05