iJRASET
International Journal For Research in Applied Science and Engineering Technology



INTERNATIONAL JOURNAL FOR RESEARCH

IN APPLIED SCIENCE & ENGINEERING TECHNOLOGY

Volume: 13 **Issue:** IV **Month of publication:** April 2025

DOI: https://doi.org/10.22214/ijraset.2025.67727

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International Journal for Research in Applied Science & Engineering Technology (IJRASET)
ISSN: 2321-9653; IC Value: 45.98; SJ Impact Factor: 7.538
Volume 13 Issue IV Apr 2025- Available at www.ijraset.com

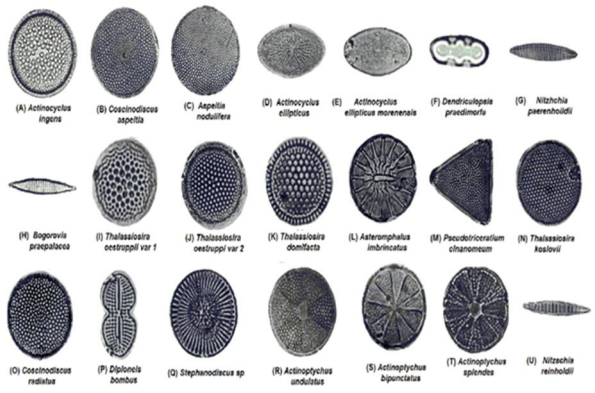
Forensic Analysis of Diatoms with Respect to Drowning Cases in the Western Ghats of Maharashtra

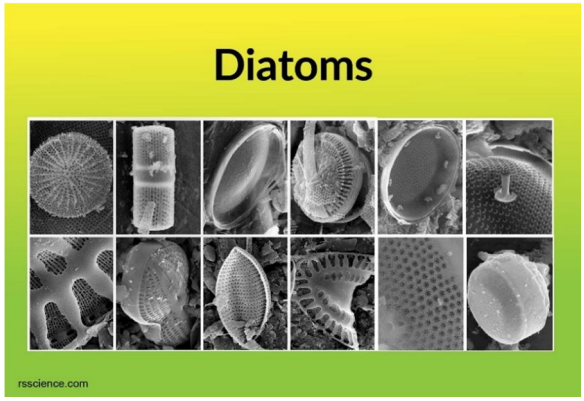
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Abstract: ***Diatom analysis is a valuable tool in forensic science and it is helpful in diagnosis of drowning cases. For extraction and analysis of diatoms, the acid digestion test and centrifugation method is used mostly. Apart from this nitric acid digestion is a widely used method for extraction of diatoms. Other extraction technique like ultrasonic radiation, enzymatic digestion and physical method such as centrifugation and gradient centrifugation method are also employed for this purpose. The presence of diatoms in lungs and other vital organ can indicate the cause of death in drowning case. This topic explores the forensic analysis of diatoms in drowning incident in Western Ghats of Maharashtra. This region has its diverse aquatic environment, carrying a variety of diatoms species, making an ideal area for the studies. By providing a database of local diatoms forensic expert can give an idea of particular case investigation. This study includes the value of diatoms biodiversity in forensic science, especially in the drowning cases of unique ecological context of Maharashtra's Western Ghats.***
Keywords: Forensic diatom analysis, drowning investigation, Western Ghats of Maharashtra, Drowning Cases.

I. INTRODUCTION

Diatoms, a diverse group of algae that perform photosynthesis, are found in various water habitats, and are becoming increasingly important evidence in forensic science. The diatoms are ecologically significant and diverse group of algae. Unlike the Rhodophyta and Phaeophyta, which are primary producer only along ocean coastlines beach or Haptophyta which are significant only in the marine phytoplankton, diatoms are major ingredients of oceanographic and planktonic algal communities worldwide, in terrestrial, brackish and marine environment [1]. These tiny creatures have distinct silica cell walls, named frustules, which exhibit variety of patterns and structures unique to each species [2]. Diatoms comprise approximately 200 genera and more than 10,000 species, with 92 genera and roughly 569 species found in India. Diatoms typically between 20-200 microns in diameter or length, although some species can up to 2 millimeters in length. Diatoms constitute a major part of the algae kingdom and most of them are unicellular organisms.





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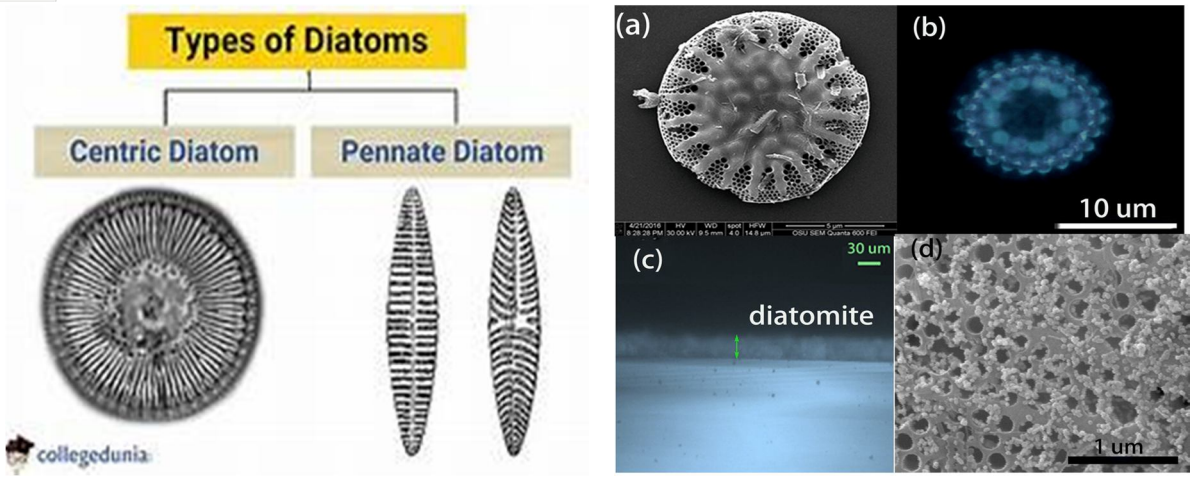


Fig:1

They occur in various forms, including filamentous, fan-shaped, zigzag, and ribbon-shaped colonies, and can be easily noticed in the freshwater and marine environments.

Some important functions of diatoms are:

- Oxygen production
- Food source
- Carbon and nitrogen cycle
- Biomass
- Silicon uptake
- Fertilization
- Insecticides

The primary function of diatoms is to convert dissolved carbon dioxide in the water into oxygen [3] Major examples of diatoms are, Fragilaria, Meridion, Tabellaria. Diatoms can be found in freshwater, brackish, and marine environments, which is crucial for forensic investigations related to bodies of water [4]. The use of diatom analysis in forensic science, especially in cases of drowning, has gained popularity in recent years. In case of drowning cases a person may inhale or ingest water containing diatoms, which then enter the lungs and other organs, providing a basis for forensic investigation [3]. Forensic diatomology, a method used to investigate drowning cases, provides a scientific approach to solving such cases [5]. Besides drowning incidents, diatom analysis can also be used in various other forensic situations, including estimating the time of submersion after death, confirming water-related crime scenes, and distinguishing between freshwater and seawater drownings. Diatoms found in the body can provide valuable insights into the time since death, location, and post-mortem movement in water, helping to differentiates a death by submersion and immersion of a body [6]. Laboratory tests may reveal the presence of diatoms in the body. In forensic practice, it is challenging to determine whether a body in water resulted from drowning or disposal after death. Diatom testing is currently an important supporting technique for determining death by drowning and identifying drowning sites [7]. The Western Ghats of Maharashtra, including areas like Pune and Ahmednagar, offer a unique opportunity for the application of forensic diatom analysis. This region is known for its diverse aquatic settings, including rivers, lakes, reservoirs, and temporary water bodies. This study aims to investigate how studying diatom assemblages in the Western Ghats of Maharashtra can lead to the creation of region-specific diatom databases, improving the credibility and precision of forensic investigations.

II. REVIEW OF LITERATURE

The forensic significance of diatoms emphasized, highlighting their role in determining drowning as a cause of death. The presence of these algae in the victim can provide ideal evidence in forensic investigations, especially when traditional signs of drowning are not apparent. The authors discuss the historical development of the diatoms test, its reliability and acceptance in forensic world [7].

The paper highlights the need of standardization protocols and advanced technologies to increase the accuracy and reliability of diatoms testing. New technologies and refining existing techniques can significantly enhance the forensic application of diatom analysis, leading to more accurate investigation of drowning as a cause of death. Freshwater diatom community structure was examined in relation to environmental and spatial gradients, and the results in ecological context. Diatoms literature stresses the importance of ion concentration and trophic status as major environmental drivers of diatom distribution in lakes and streams, while physical factors effect community structure. Freshwater diatom communities appear to be strongly spatially structured. According to studies from both lakes and streams, pure spatial factors account for 20-30% of total community variation. This suggest that diatoms lack strict ubiquitous dispersal. These results suggests that bioassessment programs utilizing diatoms should consider spatial factors, as diatom communities are strongly spatially structured [8]. A complete autopsy, histopathological examination and full toxicological screening are essential to determine whether death indeed followed submersion in the water, or if any natural disease or substance use to contributed or to caused death. In ambiguous situations, the co-estimation of circumstantial evidence may be of the invaluable importance in concluding the cause and manner of death. Thorough forensic investigation of submersion deaths not only serve the justice administration, but also presents considerable benefits for public health [9]. In this study, the author attempted to extract diatoms from cotton clothing using different methods. Diatoms in clothing can be used to determine contact with surface water and a specific water source, which can help link suspects to Crime scenes. Three methods were performed to obtain the highest average yield: rinsing with ethanol (RE) rinsing with water(RW) and dissolving cotton with nitric and sulfuric acid. Rinsing with ethanol is an effective extraction method for the qualitative and quantitative analysis of diatoms in cotton clothing [10]. Made research on Fresh water diversity in Deepor Beel, a Ramsar site, to investigate freshwater diatom diversity in Deeper Beel, a Ramsar site in Assam, India. The samples were collected from six different sites based on habitat stratification. The results showed that there were 65 species of diatoms, belonging to 26 genera, of which 53 were pennate and 12 were centric types. Abundance of diatom species provides evidence for their important role as primary producers in this wetland ecosystem of Deepor Beel of Assam [11]. Studied the role of diatoms in the world of forensic science. This article reviews the forensic aspects of diatoms analyses and acid digestion method for extracting diatoms. A body recovered from water does not necessarily mean that death was due to drowning. In drowning-related death case, a correlation between the diatoms extracted from bone marrow and samples obtained from drowning medium must be established for the successful determination of drowning site in forensic laboratories. Diatom analysis should be considered positive when number of diatoms is about a minimum established limit; 20 diatoms of pellet obtained from 10g of lung samples and 50 diatoms from bone marrow and drowning sites can strengthen this evidence and positive conclusion can be drawn whether a person was living or not when drowned [12]. Studied the detection of diatoms in water and tissues by combination of microwave digestion, vacuum filtration and scanning electron microscopy. The detection of diatoms has been proposed to be useful in the diagnosis of drowning. In this presented paper, they describe a novel method for qualitative and quantitative analysis of diatoms in water and organs. The further scientific research in the field of automatic diatom identification using SEM images has to be done in order to automate the process of detection and identification of diatoms in water and tissues for the diagnosis of drowning [13]. In the present report, 38 drowning cases are studied from Haryana (India) were examined and solved by "diatom test" in Forensic Science Laboratory Madhuban, Karnal (Haryana). After the medico legal examination, 34 cases were found to be positive for the diatoms test as the crime exhibits (sternum, clavicle, liver, lungs, spleen, kidney, intestine, heart and putrefied viscera) had the same diatoms types as were present in water samples from the drowning sites of the respective cases. Therefore, the diatoms can used as an important tool in the diagnosis of drowning cases [14]. Diatoms plays an important role in Forensic Science and in the studies of water quality. Diatom acts as supportive evidence for ascertaining the cause of death as well as the place of drowning. In present study 10 water bodies located in various part of Mumbai were sampled for study. Isolation of diatoms were carried out using acid digestion method. Total 35 species of diatoms were isolated and characterized. The information obtained helped to create a reference database on diatoms which can be utilized in future for ecological assessment and ready reference in drowning cases [15]. He assessed the water quality in small austral temperate river system in south Africa. Application of multivariate analysis and diatom indices. Canonical correspondence analysis indicated difference in diatom community assemblages explained by dissolved oxygen, temperature, phosphate, nitrate and conductivity. Several foreign indices like TDI (the trophic diatom index), SADI (south African diatom index) were used in the study. Therefore, recommend wider use of SADI as Indicator of water quality conditions in South African lotic systems [16]. Introduced a topic titled Digital whole-slide image analysis for automated diatom test in forensic cases of drowning using a convolutional neural network algorithm, which states that diatom examinations have been widely used to perform drowning diagnosis in forensic practice. However, current methods for discussing the cause of death are time consuming.

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The organs of 35 rats were extracted “0.5 h, 1 h, 6 h, 12 h, 24 h and 48 h” after drowning and those of sham-drowning group killed by mechanical asphyxia were extracted 1 h after body immersed in water. The organs were digested by acid and the diatoms were analyzed by statistics. Result shows the detection rate was 100% 6 hours after drowning except the drowning group, where only one case was positive in the lung. So, it is concluded that the detection rate of diatoms could be considered as important evidence drowning determination [17]. Introduced a topic titled - A Systematic review on various Diatom species associated with drowning. When a person gets drowned in water containing diatoms, due to forceful aspirations they enter into lungs and from lungs get lodged into the body distant organs of body. Various tests have been developed for diagnosis of drowning deaths. The extraction methods of diatoms from tissue samples is based on digestive capability, reclaiming ratio of diatoms, destruction of diatom samples and time consumed for complete diatom digestion. This paper reviewed recent year’ progress in diatom test, samples used and species of diatoms associated with drowning [18]. The literature regarding homicidal drowning. Homicide of a child by drowning occurs occasionally, but it is much rarer that an adult is murdered in this way. When the victim is a child, there will often be few or no signs of violence due to the difference in size between the assailant and the victim, unlike the evidence of a struggle that is often found in adult victims of homicidal drowning, unless the victim was incapacitated by alcohol, drugs or physical weakness, or was unexpectedly pushed into or dragged under water. The incidence of cases in which the manner of death is undetermined is high in bodies retrieved from water. Therefore, a thorough investigation of such cases is needed [19]. Diatoms, also called as the ' jewels of sea' are microorganisms mostly found in aquatic systems. The current review paper is based on the different extraction method and microscopic examination technology used in the area of forensics for analysis of drowning cases. A significant number of techniques are established by different studies to compare the efficiency of various extraction methods for forensic examination. The primary aim of each method is to extract the diatoms from dead bodies. Acid digestion, Enzymatic digestion, Soluene-350, and microwave digestion are the extraction methods of diatoms. For the identification and characterization of diatoms the microscopic methods are Light Microscopy, SEM, TEM, AFM, generally used. The extraction and examination of diatoms play a vital role to identify the location, time since death, postmortem and premortem death of the individual. Diatoms are a boon to the identification of death due to drowning [20]. In the present study, samples were collected from the 12 different water bodies in Aurangabad region. By using acid digested method sample were prepared and analysis was performed. Drowning is considered the third leading unnatural cause of death in India. To investigate the pre-mortem or post- mortem drowning death the site-specific diatoms play a vital role. The actual specification of diatoms is solved by studying the diversity and the Diatom map (D-map) which may help to reduce the investigation time and confirmation of the drowning location. The present study focuses on the regional diatomic distribution map to identify the site of drowning and increase the rate of Investigation which may be helpful in future for further investigation (Shaikh, Ghughe, Sangle, Suryawanshi,2021) [21]. By describing the diatom diversity and tropic status and the connection between diatoms and physiochemical parameters, the present study able to assess the tropic level status and quality of water of Rampur Hamesha Lake and the Kurhda Lake Bhandara district, Maharashtra. To analyze the water, they used different techniques such as pH meters, conductivity meter, TDS meter, etc. In this study Diatoms used as a potential indicator of the water quality and its composition, to evaluate the diatoms present into it. N. palea and N. viridula are the two species considered as an indicator of organic pollution [22]. Due to the beautiful ornamentations present on their frustules wall diatoms called as "jewels of the sea". They considered as golden standard in solving drowning related crimes in the area of forensic science. In this study, we have discussed the efficiency of molecular tool, this phenomenon helps in the making of database which helpful in future. The taxonomists preserve the species and generate records for study. By using diatom DNA barcoding can successfully help in solving drowning related cases with more accurately and lower the chance of contamination. From the past many years, these microalgal species remains very helpful for forensic pathologist in deciphering complex cases of drowning related crimes [5]. This paper evaluates the criteria for diatom testing in forensic investigations, particularly in drowning cases. The presence of diatoms, helps determine whether the cause of death is drowning or not. To improve the sensitivity and specificity of the diatom testing, researchers discuss methods, including microwave digestion, vacuum filtration, and automated scanning electron microscopy. The paper highlights the importance of accurately identifying diatoms and comparing them based on unit weight. It also emphasizes the need for standardized protocols to ensure consistent and reliable diatom testing. Therefore, by incorporating the new technology and refining the existing methods, the forensic application of diatoms testing can significantly improve, providing more reliable and accurate results in drowning investigations [23]. Drowning is a form of asphyxia which is caused by submersion of the body in water or any other fluid. If dead body found is found water is does not necessarily mean, that this person has drowned. To determine the actual cause of death is difficult in drowning cases. While the forensic autopsy is necessary to determine external and internal findings during investigation.

This paper is focuses on the internal and external investigation of dead bodies and the confirmatory test to investigate sample collected by forensic expert. The present study highlights specific points to draw conclusive results in drowning death investigation [24]. Forensic diatomology is a field that deals specifically study with diatoms. They have wide applications in fields other than forensics. Traditional diatom identification methods are time consuming and often vary in efficiency and lack of standardization. To address this issue, advances in computer vision and image analysis have led to the development of automated methods using computer algorithms and machine learning to identify ad classify diatoms. This review highlights the main principles, methodologies, and application used by forensic professional, researches and scientists to have an easy understanding of the potential and significance of diatoms in forensic science [25].

III. CONCLUSION

Diatoms are unicellular, photosynthetic, eukaryotic organism that present in almost all water resources .They are mostly found in river, pond, lakes, ditches and in freshwater, brackish and marine waters as well as in well water, tap water, borewell water. Diatoms also occurs in the terrestrial habitat such as rocks, mosses, and soils, and even in caves. They can be either are planktonic or benthic. These particular qualities of diatoms are used in forensic science in a different way to identify and investigate crimes. There are approximately 15,000 known species of diatoms , but many of them remained undescribed. The diatom cell is enclosed in silica case with different shapes and ornamentations. The ornamentation is often species-specific, providing important information for identification and classification. Due to their small size and hard silica cell wall, they are easily found inside the body of organs of victims and settle down. During conducting the autopsy of the drowned dead body, the presence of diatoms in various tissues reveals whether it's the case of antemortem or the postmortem. Diatoms analysis is crucial in drowning cases , especially in the regions with diverse aquatic environments like the western ghats of Maharashtra state. The Western ghats is an important biogeographic zone of India and one of the thirty-four global hotspots. Variation in the diversified group of the diatoms is observed here. some common species of the diatoms found in the western ghats include Stauroneis datarii, Stauroneis bahlsii, Stauroneis hamiltonii, Gomphonema rajaguruii, Luticola densistriata, Luticola indicensis, Luticola pseudofungiformis,Luticola bhimashankarensis,Luticola kaensis,Luticola yanensis, Luticola maharashtrensis. These are commonly found in the western region of Maharashtra, belonging to various locations within the northern western ghats, such as kass plateau and varandha ghat. Recently, a new freshwater diatom genus named Indiconema has been discovered in the Eastern and western ghats Other newly found diatom species includes Gomphonema rajagurujii, Stauroneis datarii, Stauroneis bahlsii, Saturoneis hamiltonii, luticola species.
Therefore, diatom analysis is significant in determine the location, time since death, and whether it was ante-mortem or postmortem submersion. Various methods such as acid digestion, enzymatic digestion, and microwave digestion can be used for diatom extraction and analysis, aiding in forensic investigation related to drowning cases.

A. Forensic Significance of Diatoms

The study of diatom crucial in forensic science particularly, in in drowning case. Since different varieties of diatoms are found in different geographical areas, they helps determine the location of submersion, time since death and cause of death. As a person drowns, diatoms can be found in his lungs and other organs (such as femur bone), helping to determine whether it was postmortem or antemortem drowning. In case of postmortem drowning diatoms may not be found as significantly as in antemortem drowning. Diatoms can also link a suspect to crime scene, if the diatoms found in clothing, shoes, or object used by the suspect. These can be compared with the diatoms from the crime scene to determine whether they originated from the same location. Therefore, the unique properties of diatoms from different geographical area, help significantly to solve the water related criminal cases.

B. Limitations

The limitation related to diatoms, particularly in drowning cases, are:

1) Diatoms may not be present in abundance in certain water bodies, such as ponds, wells, lakes, or springs, making detection difficult.
2) In some cases, the victims may die due shock, cardiac dysfunction, or other factors, resulting as lack of diatoms in body.
3) Contamination during sample collection and preservation can lead to false result.
4) Different geographical area have unique diatom species, and in some cases, these may not have been discovered, making it challenging to interpret results.
5) Extraction and analysis techniques can damage diatoms, leading to false result.

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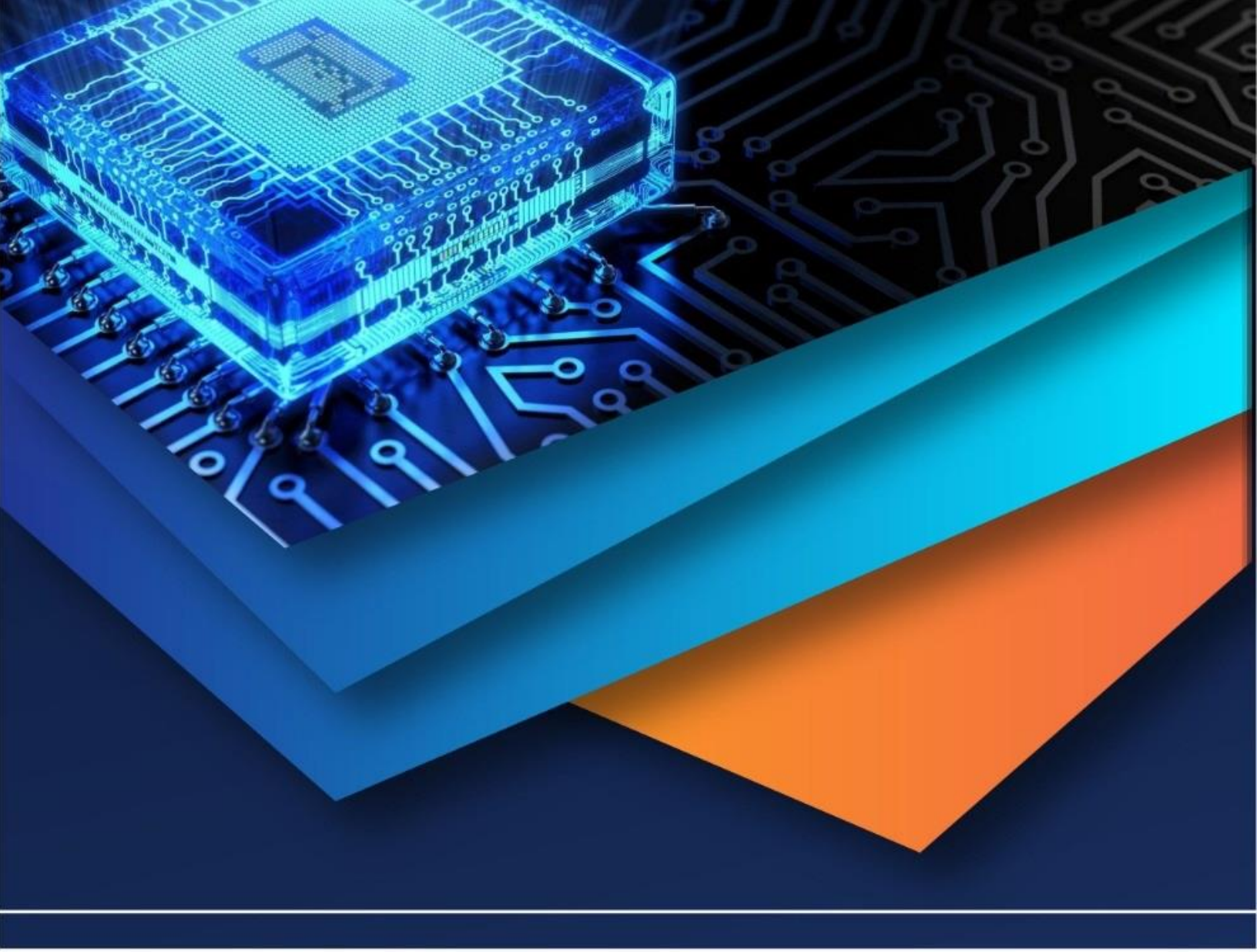
C. Future Implication or Research Gap

There is limited researches carried out on diatoms in Western Ghats of Maharashtra. Further research on diatoms in region can significantly enhance the accuracy of forensic analysis of diatoms aiding in the investigation of drowning cases. The study of diatoms also helps determine water quality, aiding in the assessment of environmental pollution, water pollution and aquatic ecosystems.

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