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Original Research Article

Identification of diatoms from different rivers in Chhattisgarh

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ABSTRACT

Diatoms are unique microalgae found in almost all water bodies. They are the only single-celled organisms with a siliceous cell wall or frustules and are ubiquitous in water bodies. Their presence and diversity make them valuable in forensic investigations, particularly in cases of drowning. Diatoms can serve as trace evidence by linking suspects to crime scene. The diversity of diatom species varies across different areas influenced by environmental factors and seasonal variations. Extensive research has been conducted on diatoms in various states of India, but the diversity of diatoms was not studied in the major rivers of the Chhattisgarh region. In this study, we aimed to address this gap by conducting a taxonomic analysis of diatoms and adding them to a new database. Samples were collected from different rivers in Chhattisgarh resulting in the identification of 34 diatom species. This study contributes to understanding diatom diversity in the region and enhances the forensic potential of diatoms in future investigations.

1. Introduction

Diatoms are unique microscopic algae containing silica and having distinct geometrical shapes. They are unicellular, eukaryotic and photosynthetic organisms. Their cell size ranges between 2-200um. They occur in the wet or moist places where photosynthesis is possible, diatoms are either planktonic (free-floating) or benthic (attached to a substratum) in nature.[1] They occur either in solitary cells or in colonies, which can take the shape of ribbons, fans, zigzags or stars.[2] Taxonomists classify diatoms into two major groups on the basis of symmetry i.e., Centric- circular and Pennate- elongate. Diatom taxonomy is based on morphological characters like size, shape and patterns of frustules.[1] In the presence of adequate nutrients and sunlight, an assemblage of living diatoms doubles approximately every 24 hours by asexual multiple fission, the maximum life span of individual cell is about six days.[3] Diatoms are major group of organisms in terms of diversity, abundance and productivity of marine and freshwater ecosystems. They are solely responsible for about 20-25% of global oxygen production i.e.; approximately every fourth breathe of oxygen we inhale.[1] They also play a significant role in drowning cases.

Every year in India, drowning is a significant contributor to causes of death, including accidental, suicidal, or homicidal drowning. Drowning is a form of asphyxia that occurs when respiration is impeded by submersion in a fluid, regardless of whether the fluid enters the lungs.[4] Diagnosing drowning is a challenging aspect of forensic pathology, especially in cases where the bodies have undergone decomposition.[5] In such circumstances, the diatom test serves as a crucial and reliable forensic investigation method for detecting drowning. Whether, it occurred before or after death.[4]

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Solving drowning cases in forensic pathology present significance challenges. Drowning is defined as the immersion or submersion of a person in any fluid, typically water. Determining the manner and location of drowning is a complex task for forensic pathologists in order to establish the cause of death. The manner of death in drowning cases can vary, including suicide, accidental, or homicide. Additionally, there are instances where the body is discovered in a decomposed state due to prolonged immersion in water. Furthermore, the presence of aquatic organisms on the victim's organ or tissues can sometimes confuse the investigation.[6]

From a historical perspective, a crucial aspect in the investigation of drowning has been the quest for a reliable, specific, and easily applicable test. Consequently, the diatom test has emerged as the primary laboratory procedure for the detection for detecting drowning incidents. In order to ascertain the manner of death, it is emphasized the collaboration between forensic pathologists and specialist-biologists, who could work together to generate the diatom database of the water bodies of different regions. This collaborative effort would facilitate the identification and localization of rare diatom species, which could greatly assist in accurately determining the specific site of drowning.[4]

Hence, the present study was to record and disseminate the information on biodiversity of diatoms exist within rivers of Chhattisgarh state in order to serve as an updated and contemporary biodiversity database for future preferences. The study area covered rivers from different regions of Chhattisgarh. Doodhnadi, this river is one of the tributaries of Mahanadi. Its origin is about 15 km from Kanker district. Malajkundam is the waterfall of the river Doodh, which flow towards east and joins the Mahanadi. Kharun nadi, its origin is about 120 km from Petchua hill south-east of Chorhanala in Balod. This river merges with Shivnath River in Somnath area near Simga. Mahanadi, it is the largest river of Chhattisgarh and Orissa region. Its length is 851 km. its length in Chhattisgarh is 286 km. the delta of this river begins about 7 miles east of the city of Cuttack in Orissa. From here it divides into many streams and joins the Bay of Bengal. Nandigaon in Raigarh, Chhattisgarh has the largest road bridge in the state of Chhattisgarh. Shivnathnadi, it is the longest tributary of the Mahanadi River, which joins Changori in the Janjgir-Champa district in Chhattisgarh, India.[7] Its origin is about 290 km. the name comes from the God Shiva in Hinduism. Hasdeo nadi, it is the main river of Chhattisgarh. It flows through many districts of Chhattisgarh and irrigates their land. It originates from Guru Ghasidas National Park in Koriya district. The length of Hasdeo River is 210 km. it forms two waterfalls in Koriya district- the first is the Gauri Ghat waterfall, and the second is the Amritdhara waterfall. Both waterfalls are situated in the middle of the dense forest.

2. Materials and Methods

2.1. Study area

The study area covered rivers from different regions of Chhattisgarh known as the Rice Bowl of Central India carved out of Madhya Pradesh to become the 26^{th} state of the Indian Union in 2000.[8] The state has tropical climate with hot summer and cold winters. Most of the precipitation occurs during monsoon (July to mid-September). Day temperature during the summers (mid-April to June end) can touch a high of around 45°C while in the winters the temperature may fall well below 10°C.[9]

2.2. Sample collection

Water samples were collected from various locations in Chhattisgarh,[10] specifically from five sites mentioned in Table 1, during the period of 1 July 2022- 20 August 2022. The samples were stored in biochemical oxygen demand (BOD) bottles and treated with 5% neutralized formaldehyde for diatom analysis. Prior to examination, the diatoms needed to be cleaned by removing unwanted materials that could hinder microscopic observation, such as cell contents, pigments, sand and mud.[11] To achieve this, 10ml of water, including sediment, was taken from each sample and centrifuged for five minutes at 5000 rpm. The settled portion was mixed with concentrated acids (HNO3+HCL) in a 3:1 ratio and digested in a water bath for 3-4 hours before cooling.[12] After digestion, the dried diatoms were diluted with distilled water and observed under a microscope.[13] A digital binocular microscope was utilized to preserve images of the diatoms seen on the slide as photographs. DPX solution was used for permanent mounting of the slides, enabling future observations. The diatom species on the slides were examined using a digital binocular microscope at 10x magnification with DPX mount. The identification of diatom species was based on their morphological characteristics, with taxonomic identification supported by available literature (Spaulding et al., 2021).[14] Analyzing diatom species in the water samples facilitated the creation of a database that aids experts and researchers in forensic investigations and further research analysis. It should be noted that diatoms exhibit significant diversification among different water sources, despite their prevalence in water bodies.[10]

3. Results

The current study focuses on the biodiversity of diatoms, and a total of 34 genera of diatoms were identified from five collection sites. These diatom genera were categorized into seven different morphology categories. Notably, Actinella was found to be abundant in Doodhnadi, Encyonema in Hasdeo River, Nitzschia in Kharun River, and Navicula in Mahanadi. These findings highlight the presence of

Table 1:

S. No	Sample type	Rivers of Chhattisgarh	Location	Genera by morphology
01	Freshwater	Mahanadi	Arang	Asymmetric Biraphid, Monoraphid, Eunotioid
02	Freshwater	Kharun	Raipur	Asymmetric Biraphid, Centric, Araphid, Epithemiod
03	Freshwater	Shivnathnadi	Bhilai-Durg	Centric, Araphid
04	Freshwater	Doodhnadi	Kanker	Centric, Araphid
05	Freshwater	Hasdeo	Korba	Asymmetric Birapid, Araphid

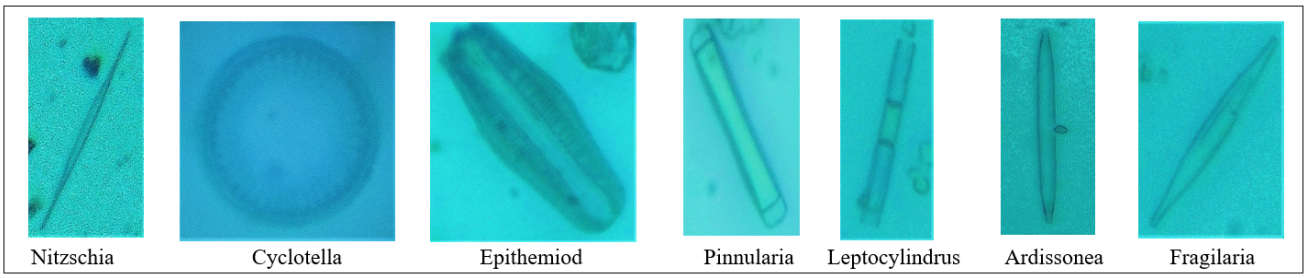

Fig. 1: Diatoms of Kharun river

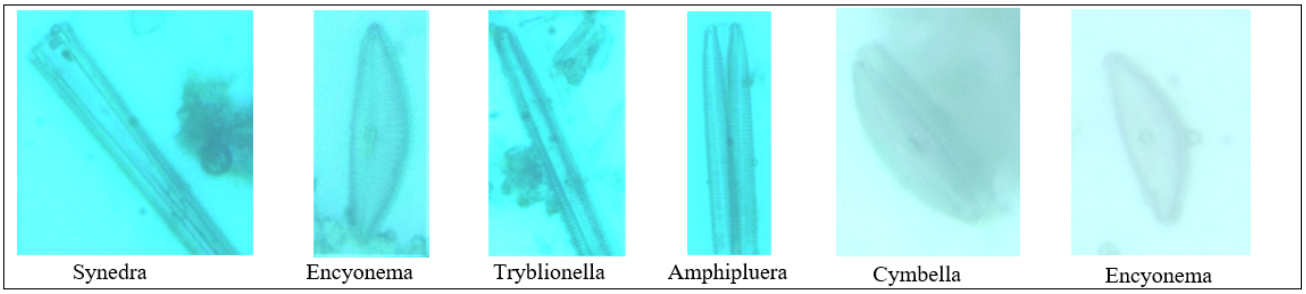

Fig. 2: Diatoms of Hasdeo river

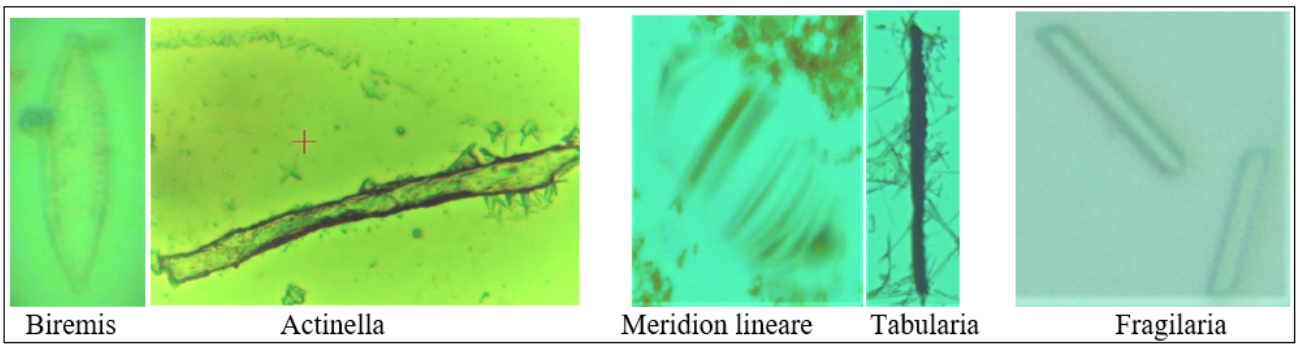

Fig. 3: Diatoms of Doodh river

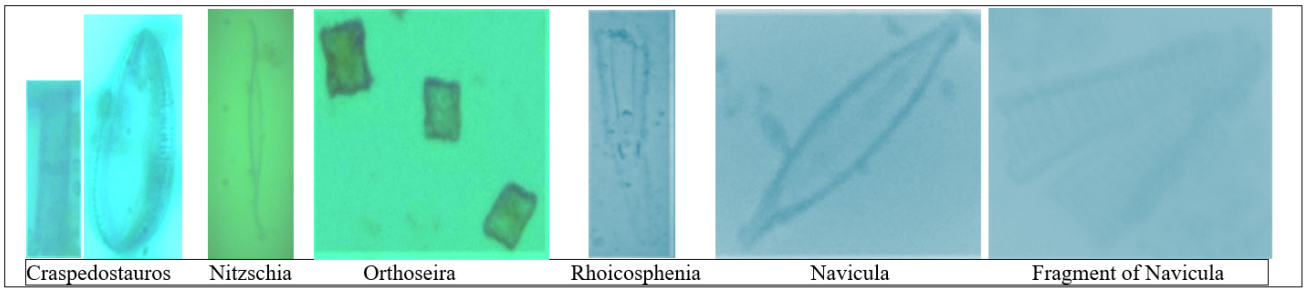

Fig. 4: Diatoms of Mahanadi river (Arang)

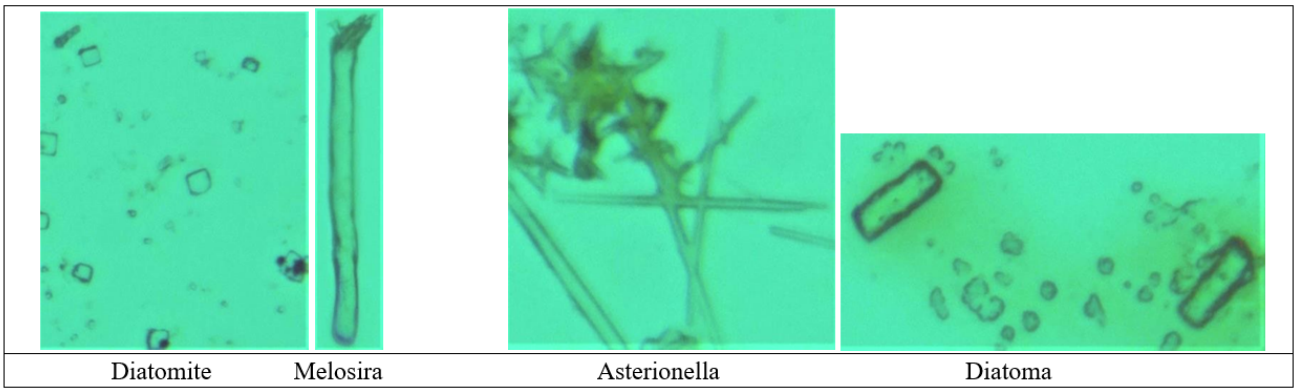

Fig. 5: Diatoms of Shivnath river

Table 2: Total species of diatoms found from different regions of Chhattisgarh

S.No	Arang (Mahanadi)	Korba (Hasdeo river)	Raipur (Kharun river)	Kanker (Doodhnadi)	Durg-Bhilai (Shivnath river)	Raipur (Pondwater)	Koria
1	Nitzschia	Encyonema	Navicula	Tabularia	Melosira	Pronia	Achnanthes
2	Navicula	Synedra	Grammatophora	Bacillaria	Asterionella	Fragilaria	Amphora
3	Microstatus	Tryblionella	Acanthoceras	Fragilaria	Diatoma	Synedra	Cymbella
4	Rhoicosphenia	Hantzschia	Fragilaria	Actinella	Diatomite	Achanthes Cocconeis	Eunotia
5	Diatoma	Cymbella	Pinnularia	Synedra		Rhoicosphenia	Fragilaria
6	Pinnularia	Amphipleura	Ulnaria	Eunotia		Cymbella	Gomphonema
7	Halamphora	Pinnularia	Cyclotella			Amphora	Hantizschia
8			Leptocylindrus			Navicula Pinnularia	Navicula
9			Ardissonea			Pinnularia	Nitzschia
10			Nitzschia			Eunotia	Stauroneis
11						Asterionella	Synedra
12						Nitzschia	

different diatom species in different locations within the region. The examination revealed variations in the structure of diatoms, which could be attributed to factors such as climate changes, water supply, pH levels, and geographical conditions. Analyzing diatom species in water samples not only contributes to understanding the diversity of diatoms but also assists experts and researchers in building a comprehensive database. The whole detail of total diatom species is mentioned in Table 1.[10]

4. Discussion

The presented findings show a significant level of compatibility and similarity between the diatom genera observed in the present study and those reported in several previous studies. A total of 34 genera were identified in the study area, and out of these, 20 genera were found to be common in the studies conducted by Silmour et al.,[11] Gautam et al.,[9] Keshari et al.,[2] Pal et al.,[15] Kaushik et al.,[16]

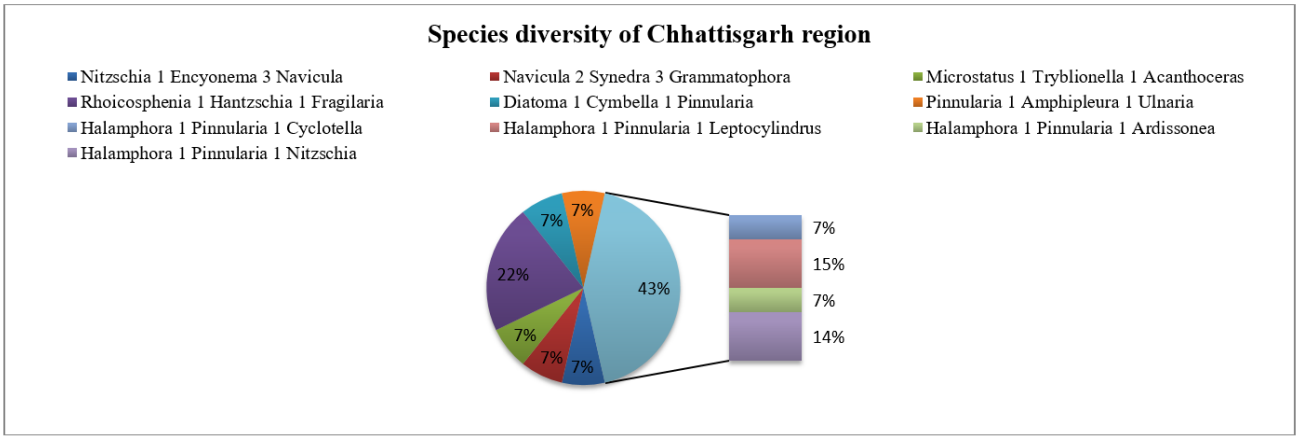

Chart 1: Presenting diatom species of different rivers of Chhattisgarh region, India

Shrivastava et al.,[17] Bharati et al.,[1] Verma et al.,[13] Sharma et al.,[6] Kumar et al.,[3] Dahiya et al.,[18] Singh et al.[19] and Bhardwaj et al.[20]

The study conducted by Silmour et al.[11] reported 7 common genera with the present study, namely Nitzschia, Navicula, Synedra, Hantzschia, Achanthoceras, Fragillaria and Enotia. These genera were observed in 5 water sample from different places in Koria, Chhattisgarh, showing accuracy and similarity with the present study.

Similarly, Gautam et al.[9] observed 12 diatoms from water bodies in the Haryana region, with 3 genera (Melosira, Nitzschia, and Pinnularia) found to be similar to the present study. Keshari et al.[2] reported 4 genera from samples collected in the Kotulpur area of West Bengal, with 2 genera (Eunotia and Navicula) found to be similar to the present study.

Pal et al.[15] examined drowning cases in Himachal Pradesh and found Navicula, Astrionella, Nitzschia, Cyclotella, and Cymbella as the most frequent genera, which were also observed in the present study. Kaushik et al.[16] examined drowning cases in Himachal Pradesh and reported 7 common genera with the present study (Cymbella, Cyclotella, Diatoma, Navicula, Tabullaria, Rhoicosphenia, and Synedra).

Shrivastava et al.[17] reported 4 common genera (Cymbella, Fragillaria, Navicula, and Synedra) from Bundelkhand, Uttar Pradesh. Bharati et al.[1] observed 22 species belonging to 13 genera, with 9 genera (Cyclotella, Synedra, Fragillaria, Eunotia, Navicula, Pinnulatia, Cymbella, Nitzschia, and Hantzschia) found to be most common in the present study.

Verma et al.[13] conducted a survey of diatoms flora in Punjab and found 13 genera, all of which showed close similarity to the present study. Sharma et al.[6] reported 70 diatom species from Hamirpur, Himachal Pradesh, with 7 genera (Fragillaria, Cymbella, Navicula, Diatoma, Nitzschia, Encyonema, and Ulnaria) found in common with the present study. Prashant et al.[3] observed 12 genera (Fragillaria, Synedra, Achanthes, Rhoicosphenia, Cymbella, Navicula, Pinnularia, Eunotia, Astrionella, and Nitzschia) of diatoms from ponds in Raipur, Chhattisgarh, with 10 genera found to be similar to the present study.

Dahiya et al.[18] identified 66 genera of diatoms from Gujarat, with 5 genera (Pinnularia, Ulnaria, Bacillaria, Tabullaria, and Navicula) found to be common in the present study. Singh et al.[19] reported 20 diatom genera from different regions of Delhi, with 6 genera (Fragillaria, Synedra, Nitzschia, Encyonema, and Ulnaria) showing similarity with the present study. Nitika et al.[20] observed 55 genera of diatoms from Himachal Pradesh, with 10 genera (Synedra, Diatoma, Nitzschia, Navicula, Fragillaria, Melosira, Cymbella, Cyclotella, Encyonema, and Amphipluera) found to be similar to the present study.

Overall, the consistency and compatibility in the diversity and distribution of diatom genera across multiple studies and different regions provide a strong foundation for discussion and further investigations in the field of diatom research.

5. Conclusion

Diatoms are most frequently used in forensic science to identify drowning as the cause of death. In the current study, diatoms were taxonomically analysed using water samples from five rivers and added to a new database. It is crucial to continuously examine water bodies and their taxonomy every year. Diatom flora variety brought on by climate change makes it possible to accurately detect drowning deaths. Diatom tests were very helpful in solving drowning cases that involved bodies that were skeletonized and in advanced stages of decomposition. They can also be useful in medico-legal investigations, especially in situations where autopsy and spot test results do not support drowning as the cause of death.[21] In such

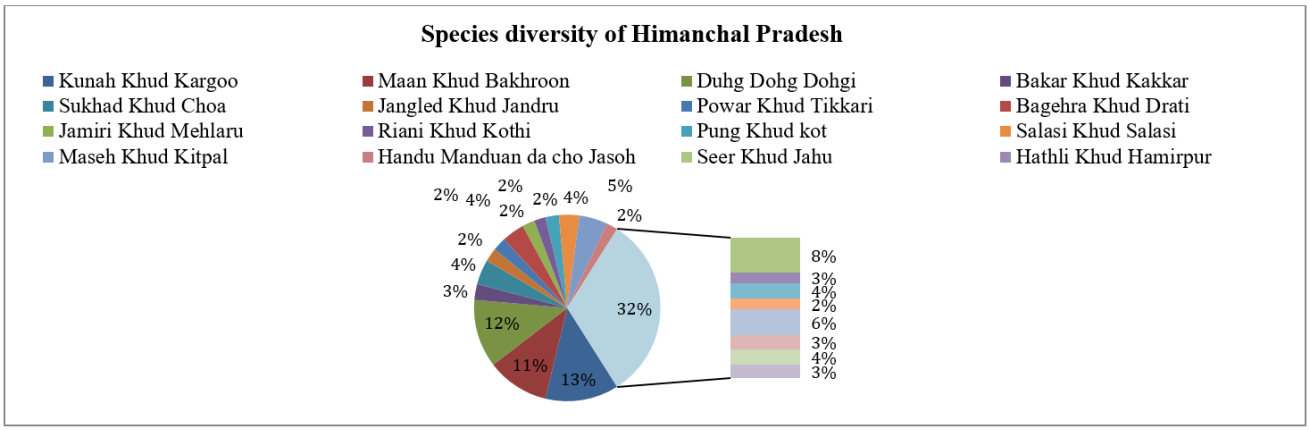

Chart 2: Representing diatom species of Himanchal Pradesh, India

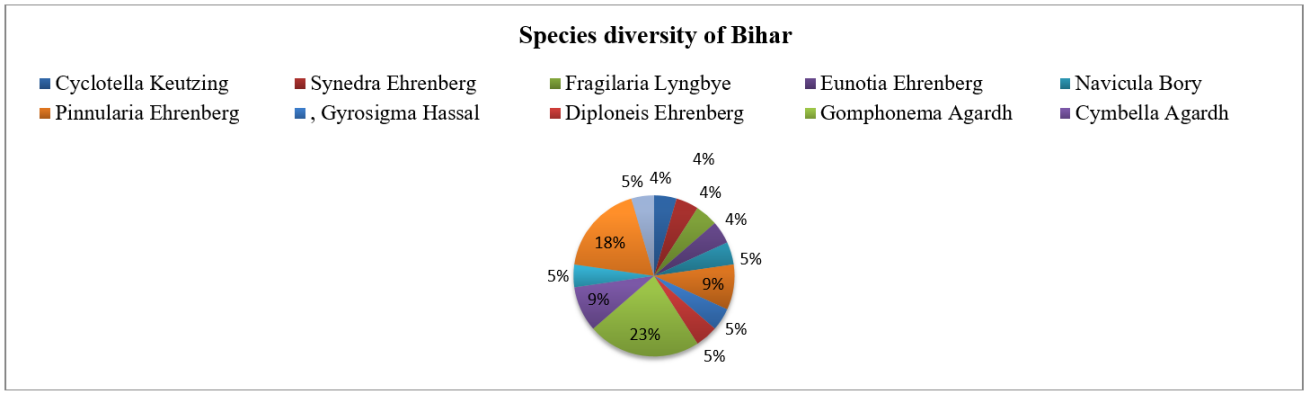

Chart 3: Representing diatom species of Patna, Bihar, India

circumstances, the location and season of drowning can be determined if a careful examination of the diatom flora of a specific body is kept on file. This led to the creation of a fingerprinting method for diatoms using a database of the most frequently occurring site-specific and seasonal diatoms, together with their shape and size.[4] The results of this study demonstrate the innovative applications of diatom biodiversity in forensic, environmental, and future biotechnological developments. Due to their ability to secrete oil, diatoms are useful in the manufacturing of petrol. The use of diatoms in the manufacture of solar panels, cosmetics, antibiotics, soaps, biofuels, and anti-proliferative compounds is also growing in popularity.[19]

6. Source of Funding

None

7. Conflict of Interest

The authors declare no conflict of interest.

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