

STUDY OF DIATOM DYNAMICS IN FRESH WATER HABITS OF KODAGU DISTRICT, KARNATAKA, INDIA

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ABSTRACT:

The study was conducted to analyze the diatom composition of three lakes in Kodagu district for the first time. 3 freshwater habitats namely, Honnamana kere, Dodda kere, and Tavarekere were selected for the study based on the influence of anthropogenic activities. A total of 38 species belonging to 15 genera were reported. The genera with the largest number were *Navicula*, *Pinnularia*, *Cymbella*, and *Eunotia* with 9,4,3, and 3 species respectively. Further, diversity indexes like Shannon-Weiner, Pielou's evenness index, and Soren sons's index were used to determine the trophic status of the lake. Site 1 showed a higher population count, while site 2 recorded a higher species count. It was understood from the study that nutrient parameters like TDS and nitrate content will play a major role in diatom diversity. The CCA plot revealed the relationship of the diatom taxa with the environmental parameters. *Gomophena acuminatum* was found to be grown in high nitrate conditions, and taxa like *Pinnularia ovata*, *P.gibba*, *Nitzschia acicularis*, and *Navicula disjuncta* showed a positive relation to EC and TDS, inferring their preference to oligotrophic habitats. Since this study is the baseline study of the area, it can be further used as a reference to understand the effects of anthropogenic activity on freshwater habitats in the area.

Keywords: Biomonitor, Diversity indices, Canonical Correspondence Analysis, water quality, diatom diversity.

Abbreviations:

EC- Electrical Conductance

TDS- Total Dissolved Solids

CCA -Canonical Correspondence Analysis

PAST-Paleontological statistics software

Tk- Tavarekere

DK -Dodda Kere

HK- Honamma Kere

1. Introduction:

Freshwater habitat is one of the essential natural resources on the planet. With the increasing world population, the demand and need for freshwater habitats are always on the rise. However, many anthropogenic activities have led to the degradation of this, and the rate is alarming. To curb these effects, many water analysis



parameters, indexes, and regulatory bodies are in the picture. In the conventional route, the quality of water is examined by the physicochemical parameters of organic and inorganic constituents. This analysis restricts the information up to the quantification of Physicochemical parameters and does not provide any insight into the ecological impact of the pollutants. In such cases, monitoring and assessing the resident biota may provide us with a much clearer picture of the effect of pollutants (Coesel, 2013.). It is known that water bodies are characterized by their spatial and temporal variations and only analyzing the physical and chemical parameters of water is not sufficient in that regard(Hwang et al., 2014).

In such situations the biota of the habitats come into the picture, the measurement and monitoring of the biotic factors help in understanding the effect of anthropogenic influence and can be used as a signal to understand the trophic level of the water bodies (Srivatsava *et al.*, 2017). Various biotic factors like Fishes, Macrophytes, zooplanktons, periphytons, phytoplanktons have been reported as biomonitors (Nazneen et al., 2022, Plessel et al., 2017, Kazmi et al., 2023,Xiong et al., 2020). Among periphytons, diatoms have been established as potential biomonitors to assess the effect of environmental pollutants in freshwater habitats (Stevenson *et al.*, 2010).

Diatoms are known for their rapid response to environmental changes (Venkatachalapthy and Karthikeyan, 2014), and are reported to be excellent biomonitors of nutrient enrichment, heavy metal contamination, and acidification of water sources(Ali, Jabbar & Hassan, 2018, Tokatli et al., 2020). Diatoms are microscopic, single-celled organisms, with silica frustules. They have the potential to act as warning signs for increasing pollution. They are also used as a parameter to check the success rate of habitat restoration, because of their shorter life cycle. This also gives them an edge over other bioindicators like fish and macro-invertebrates. The cost borne for their analysis and sampling is relatively low, and they can be collected and stored much more easily.

Diatom samples can be collected easily for long periods for analysis. As a result, research on diatoms is becoming a crucial component of monitoring and evaluation initiatives in many nations like Brazil (Bere et al., 2011), Zimbabwe (Bere et al., 2015), China (Yandg et al., 2015, Lei et al., 2017), Bangladesh (Hasan et al., 2023) However, In India, the diatom study is majorly restricted to the documentation from various sampling sites (Vijayan D., 2016, Pradhan et al., 2020). The works of Nautiyal et al., (1996, 2000, 2009) majorly focuses on the diatoms of Himalayan regions specifically. However, few workers have tried to interlink the diatom diversity with environmental parameters(Venkatachalapthi and Karthikeyan, 2012, Nautiyal and Verma, 2015). There is still a huge lacuna to be explored to establish a uniform system of sampling and species counting for the diatoms to be considered as biomonitors.

The present study was considered with the objective of reiterating the importance of diatoms as a biomonitoring tool in India. The objectives were

- 1) To prepare a baseline data of diatom diversity in the sampling sites.
- 2) To study the distribution of diatoms with reference to the environmental variables of sampling sites, and to identify the bio-indicator diatoms of the sites.

2. Materials and Methods:

2.1 Study area:

Samples were collected from 3 lakes (Table1) located in Kodagu district, Karnataka state, India (Fig1). The sample sites were selected based on the influence of anthropogenic activity in and around the region.

Further, the water levels and the nutrient sources of the lakes were a parameter of consideration. The geographical location of the sites was noted with GPS and the depth of the lakes was measured using a weighted line.

2.2 Physicochemical analysis of water:

The water samples were collected monthly in the morning period of 7 AM to 9 AM from June 2020 to May 2022. The parameters like water temperature were measured on-site. The parameters like pH, TDS, and EC were conducted immediately upon reaching the laboratory. The parameters like D.O. and nitrates were carried out as per the guidelines of APHA(1998).

2.3 Diatom Collection, Identification, and Analysis:

The diatoms were collected using two methods a) By using plankton nets of mesh size of 10 μ m. b) collecting the water samples by squeezing the roots and rhizoids of aquatic macrophytes into sterile specimen bottles in duplicates.

The sample was treated with 1% lugol's iodine and 4% formalin(John, 2000). The sample was deposited in the herbarium of Dept. Of Botany, Jnana Kaveri Campus, Kodagu with a voucher number and date of collection.

Temporary mountings were prepared and photographed to document the diversity of diatoms in the sample. The identification was done using standard literature such as Krammer and LangeBertalot (1986),Gandhi (1998), Metzeltin and Lange-Bertalot (2007), and Krammer (2003). The valid names and author citations were documented using the algaebase (Guiry and Guiry, 2019). The quantitative analysis of samples was done, by centrifugation of the desired amount, and counting the number of organisms using a Sedgewick rafter (Karthick et al., 2010). The diatom quantification was measured as organisms/L.

Statistical Analysis:

The diversity of diatoms distribution in the sample sites was determined using diversity indices like the Shannon diversity index, Pielou's evenness index, and Sorensens's index using the PAST software. Furthermore, based on the diversity indices value, the lake's trophic state was determined. The CCA plot was constructed to understand the influence of water parameters on the diversity of diatoms (PAST 4.03 version).

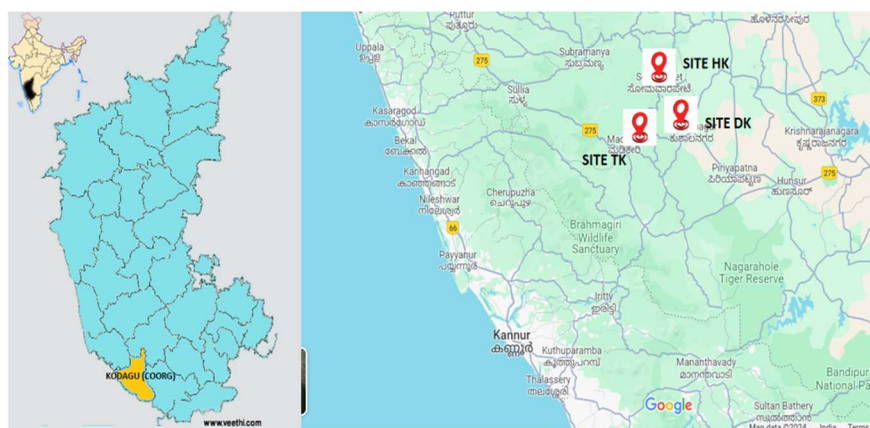


Fig 1: Study sites (Source : Google maps)

Table 1 :

Site	Latitude	Longitude	Depth (m)	Level of anthropogenic activity	Habitat description
Honnamana kere (HK)	12.6208 ⁰ N	75.8806 ⁰ E	5	high	The lake is adjacent to a temple, where religious activities are carried out regularly with offerings being thrown into the lakes. They have, introduced fish communities.
Dodda kere (DK)	14.73901 ⁰ N	76.3702 ⁰ E	8	moderate	The water body is situated amid the forest and agriculture lands. The lake is used as an water source for irrigation in summer and fishing ground for locals.
Tavare kere (TK)	12.4555° N	75.9570° E.	4	High	The lake is situated near to a industrial sewage rivulet. (about 200 meter). The lake is populated with and <i>Azolla</i> sp, <i>Lemna</i> sp., <i>Ceratophyllum</i> sp. and <i>Salvinia</i> . sp.

Description of sampling sites.

Results

The results of the Physicochemical analysis of water samples are shown in Table 1. PreMonsoon. The surface temperature of water varied from 24 ± 1.63 °C in pre-monsoon to 26.7 ± 4.27 °C post-monsoon. (graph 2) The pH of lakes varied from 7.3 ± 0.26 to 8.4 ± 0.16 . The lowest pH was recorded pre-monsoon of 2020 and the highest was recorded in pre monsoon of 2021 (graph 1)

The TDS was measured in mg/L. The TDS values varied from 55.075 ± 14.01 in the Monsoon of 2021 to 429.65 ± 42.9 in Pre monsoon of 2022. The TDS was minimal in monsoon and highest in Pre-monsoon (graph 3). The EC was measured in $\mu\text{S}/\text{cm}$. The EC varied from 12.3 ± 3.37 to 65.5 ± 5.7 $\mu\text{S}/\text{cm}$. The highest conductance was observed in Lake TK in the monsoon season (graph 4). The D.O. level varied from 3.22 ± 0.44 mg/L to 9.1 ± 0.39 mg/L (graph 5). The level was high in the winter season. The values of nitrate were, 0.12 ± 0.1 and the maximum was 1.22 ± 0.7 (graph 6)

The diatom diversity of the three lakes varied along with their population. The diatom diversity was highest at Lake DK with 16 taxa in 2020-21 and 18 taxa in 2021-22. This was followed by lake TK with 15 taxa in both study periods. However, the population count viz, organisms/L was higher in lake TK. The diatom diversity of *Naviculla*, *Pinnularia*, *Cymbella*, and *Eunotia* with 9,4,3,3 species respectively. The taxon number varied seasonally with the least in pre-monsoon and maximum in post-monsoon.

The diversity indices like Shannon Diversity [$H'(\log e)$], Simpson diversity index [D],

Pielou's Evenness (J') and the trophic status of lakes are mentioned in Table 2. The

The diversity index of Shannon, Simpson, and evenness in lakes is mentioned in Table 2.

The highest Shannon index was 2.62 in lake DK (2021-2022), highest Simpson index was in lake DK 0.9141 (2021-2022). However, the evenness was found to be inversely proportional to the diversity indices value.

The CCA plot elucidating the relationship of diatom distribution in response to environmental parameters like pH, DO, TDS, EC, and Nitrate is depicted in Fig 2 and Fig 3. Plot 1 (fig 2) showed 54% similarity, with an Eigen value on Axis 1 and an Eigen value of 0.50 with 45% similarity on Axis 2. Plot 2 (fig 3) showed an Eigen value of 0.57 with 55.3% similarity in Axis 1 and an Eigen value of 0.48 with 45.9% similarity in Axis 2. In the CCA plot, the length of the environmental parameters corresponds to the importance of the parameter. The position of the species shows their preferred habitat about the environmental parameter. Out of the 33 species, 22 species showed a normal distribution.

In season 1 (2020-21), taxa like *Pinnularia ovata*, *Nitzschia acicularis*, *Navucula germanii* showed a positive relation with EC and TDS. Few other taxa like *Navicula trivalis*, *Melosira juergensivini*, *Melosira granulata* and *Fragillaria crotensis* showed a moderately higher D.O, TDS, and EC.

In season 2 (2021-22), *Gomophonema truncatum*, showed positive relation with Higher nitrates, *Pinnularia gibba*, *P.ovata*, *Nitzschia acicularis*, *Navicula germanii*, showed a positive relation with EC and TDS. Few other taxa like *Navicula trivalis*, *Melosira juergensivini* and *Melosira granulata* showed a moderately higher D.O, TDS, and EC.

	Shannon Diversity /E[H'(log e)		Simpson diversity index [D]		Pielou's Evenness (J')		Trophic status
	2020-21	2021-2022	2020-21	2021-2022	2020-21	2021-2022	
TK	2.526	2.479	0.9102	0.9017	0.8339	0.795	Mesotrophic
DK	2.471	2.62	0.8896	0.9141	0.6958	0.7633	Mesotrophic
HK	2.128	2.155	0.8598	0.8696	0.8396	0.8632	Mesotrophic

Table 2: The values of different diversity indices of the lakes .

CCA PLOT

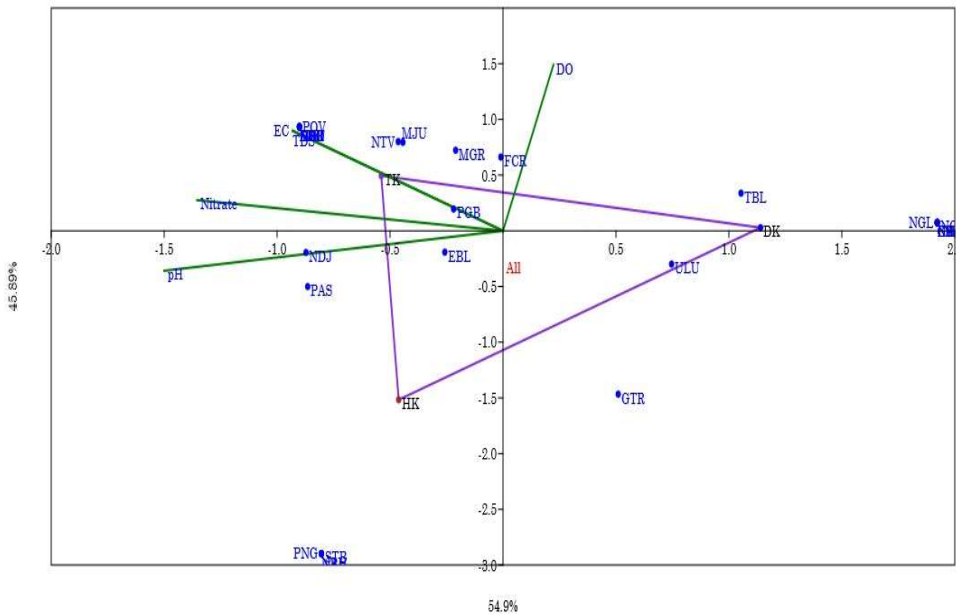


Fig 2 : Canonical correspondence analysis (CCA) graph depicts the distribution of diatom study sites TK, DK and HK and its correlation with 5 environmental parameters(2020-21).The vector line is the correlation of

physical parameter with the ordination axis. The taxon are indicated as acronyms (Appendix II).

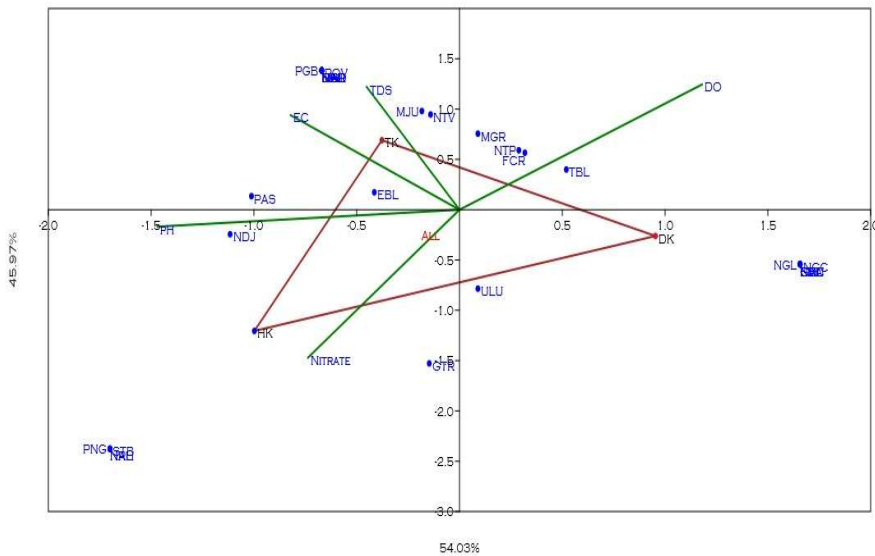


Fig 3: Canonical correspondence analysis (CCA) graph depicts the distribution of diatom study sites TK, DK and HK and its correlation with 5 environmental parameters(2020-21).The vector line is representing the correlation of the physical parameter with the ordination axis. The taxon are indicated as acronyms (Appendix II).

4. Discussion:

Natural events like rainfall and seasonal variations of atmospheric temperature contribute to changes in water quality (Gay & Ferguson, 2012). Anthropogenic activity is also a major contributor in impacting the water quality. In some months not many effects caused due to anthropogenic activity were observed in two seasons, due to the reduced activity because of Covid -19 lockdown.

The diversity indices of diatoms in any water habitat are usually studied with the use of multiple indices. Here we chose three indices as it is frequently used (Renuka, 2014). The value of the Shannon diversity index varies from 1-4. If the value is less than 1, it indicates the trophic status to be eutrophic, if the value is greater than 1, it is Oligotrophic, and if the value lies between 1to 3, the sample sites are Eutrophic (Shetty & Krishnakumar, 2022).

In the Shannon diversity index, the higher the value, the higher the diversity, Which co-relates to our sampling site DK in season 2. The Simpson diversity index increases with an increase in the diversity and species richness. The evenness is inversely proportional to the Shannon diversity index, which correlates to our study in the sampling site of DK in seasons 1 and 2.

Diatoms play an important role as bioindicators in fresh water systems, and they are known to show seasonal variations (Srivatsa et al., 2017). We found that with variation in seasons, the diversity and abundance of the

diatom taxa varied, with a reduction in dry season. This was also reported by many workers (Martinez, et al., 2003, Nahar et al., 2010, Hu et al., 2020).

On performing the CCA plot, a few of the species were found to be potential bioindicators, like *Gomphonema truncatum*, showed positive relation with Higher nitrates, *Pinnularia gibba*, *P.ovata*, *Nitzschia acicularis*, *Naviucula germanii*, showed a positive relation with EC and TDS. Some of these taxa were reported as bioindicators in sampling sites by a few workers(Halder &Sinha, 2015, Martin & Fernandes, 2012). As per Lange Bertalot (1979), species are indicative of their upper limit of pollution tolerance, and not their lower limits. Thus few species, even though being reported as indicators in eutrophic habitats, can be observed in mesotrophic habitats also.

Our results showed that parameters like Tot. Nitrate, EC, and TDS are good parameters to be considered for evaluating the prediction of biomonitors. It is also reported that along with these parameters, pH, Temp, and vegetation can also be factors to be considered (Bere, 2014). The vegetation cover of the sample site affects the absorption and intensity of light reaching the water surface, which in turn affects the rate of photosynthesis(Pan et al., 1996).

Eventhough our results didnot correlate to this, as the pH wasn't showing a strong correlation with any of the diatoms, It has been found relevant in other similar studies, (Round, 2004). However, it is to be noted that the abundance and diversity of diatom taxa varies from one region to another, and previously reported indicator organisms may not be found in the current sampling site.

Conclusion:

The use of diatoms as bioindicators is a well-developed procedure in European countries. However, it is underdeveloped in India with very little available literature. This study shows that few taxa of diatoms have the potential to be considered for indicators of water quality. The data obtained through the study shows a clear influence of ecological parameters on diatom diversity. However, more spatial and temporal data needs to be evaluated to come to a firm conclusion on the species mentioned above. That being said, this study only focuses on one class of phytoplankton, while other classes, macrophytes and zooplankton are not taken into consideration. The application and significance of the studies majorly depend on the sampling sites and the technique followed for the counting. There should be an approach to study and standardize the protocols and develop a universal technique for diatom biomonitoring.

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Declarations:

The authors declare no conflict of competing interests.

Ethics approval and consent to participate

Not applicable

Availability of data and material:

All data generated and analyzed during this study are included in this article.

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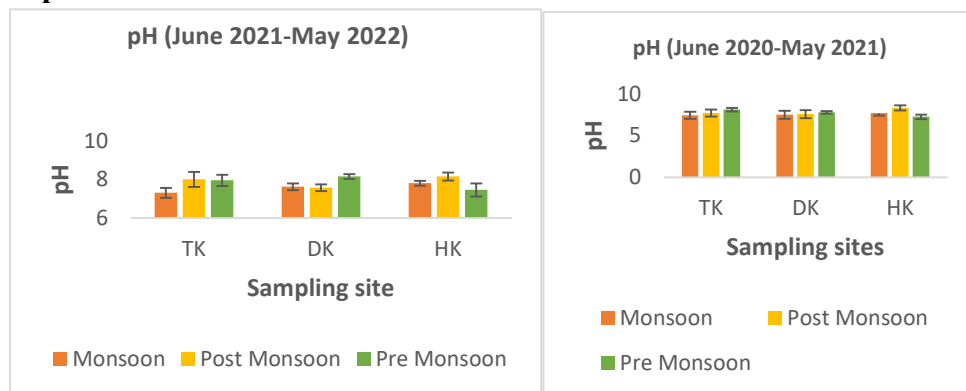
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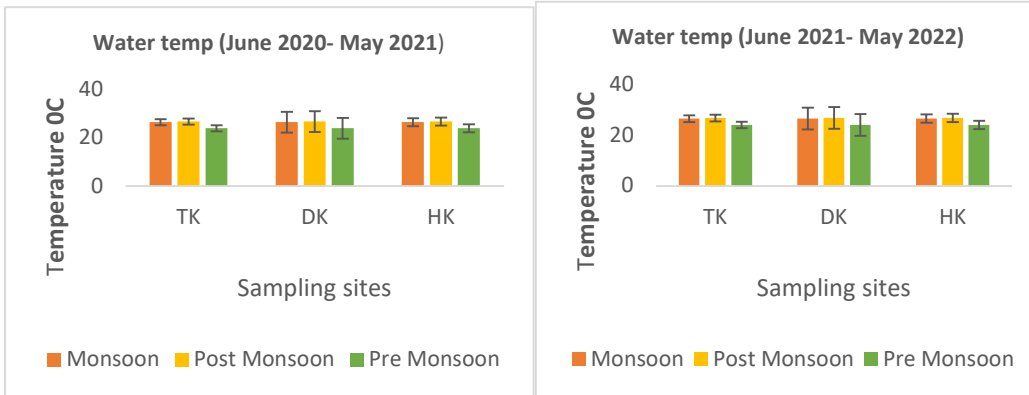
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Annexure 1; The Physico-chemical parameters of the water samples

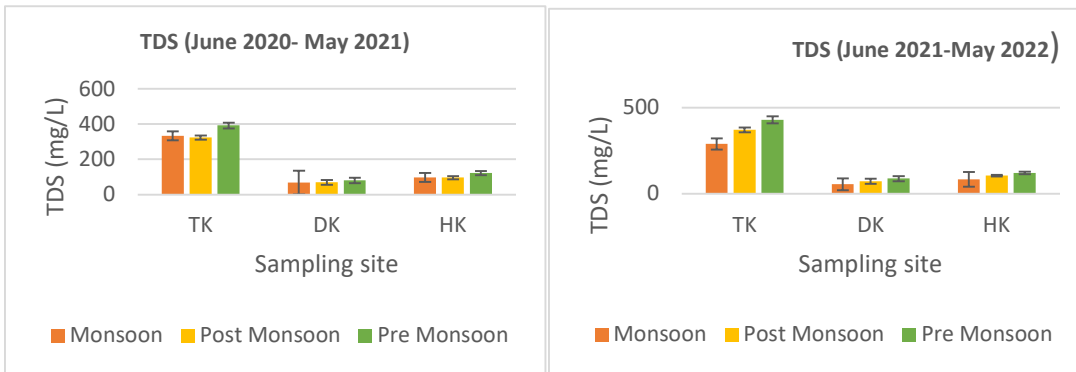
1. pH



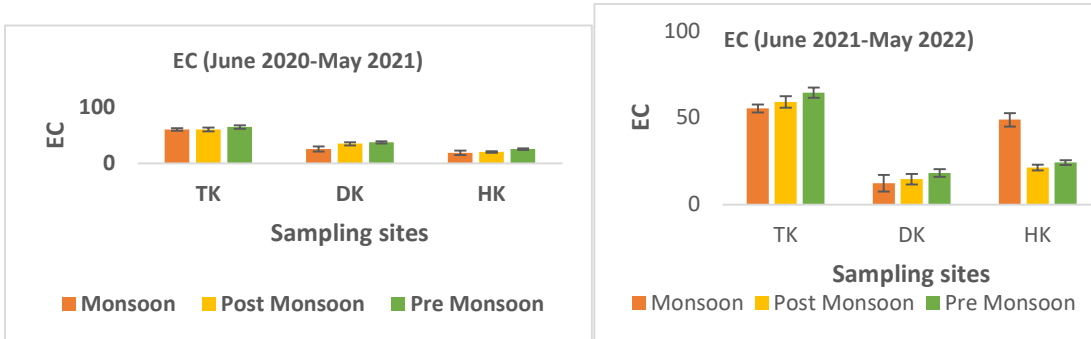
2. Temperature



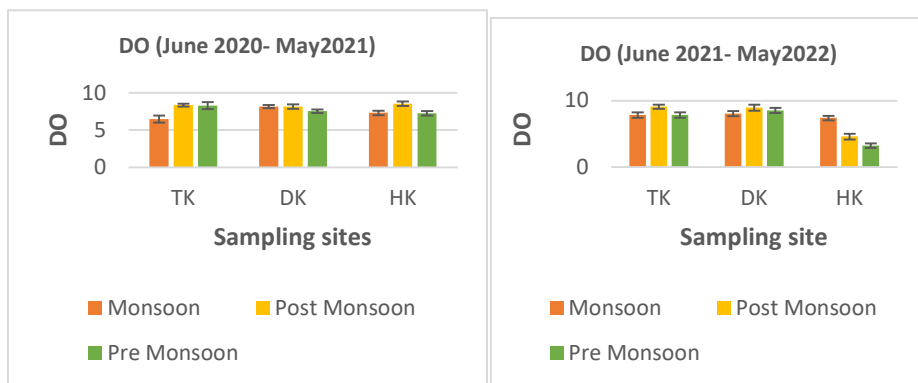
2. TDS



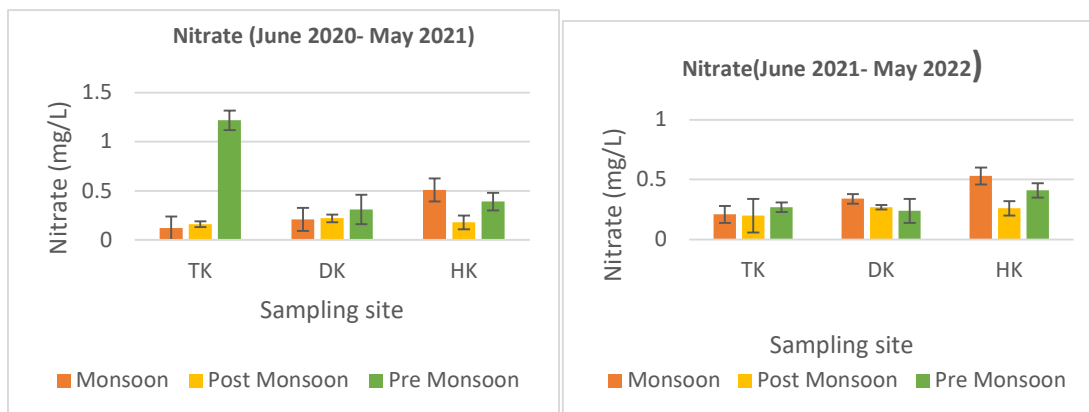
4. Electrical Conductance



5. Dissolved Oxygen



6. Nitrate



Annexure 2: The distribution of diatoms in three sampling sites.

Srl No	Abbr v.	Diatom name	TK		DK		HK	
			20-21	21-22	20-21	21-22	20-21	21-22
1	CLA	<i>Cymbella lanceolata</i> (C.Agardh)Kirchner	Red	Red	Yellow	Green	Red	Red
2	CHL	<i>Cymbella helvetica</i> Kutzing	Red	Red	Yellow	Yellow	Red	Red
3	CVL	<i>Cymbella ventricosa</i> Cleve-Euler	Red	Red	Green	Green	Red	Red
4	EBL	<i>Eunotia bilunaris</i> (Ehrenberg) Schaarschmidt	Green	Green	Yellow	Yellow	Yellow	Yellow
5	EM U	<i>Eunotia mucophila</i> (Lange-Bertalot, Norpel-Schempp & Alles).	Red	Red	Yellow	Yellow	Red	Red

6	EIN	<i>Eunotia incisa</i> W.Smith ex W.Gregory						
7	DCR	<i>Diploneis crabro</i> (Ehrenberg) Ehrenberg						
8	FCR	<i>Fragilaria crotonensis</i> Kitton						
9	GTR	<i>Gomphonematruncatum</i> Ehrenberg						
10	GAC	<i>Gomphonema</i> <i>acuminatum</i> Ehrenberg						
11	MJU	<i>Melosira juergensivii</i> C.Agardh						
12	MG R	<i>Melosira granulate</i> (Ehrenberg) Ralfs						
13	MA T	<i>Mayamaea atomus</i> (Kutzing) Lange- Bertalot						
14	NTP	<i>Navicula tripunctata</i> (O.F.Müller) Bory						
15	NTV	<i>Navicula trivialis</i> Lange- Bertalot						
16	NFC	<i>Nitzschia fonticola</i> (Grunow) Grunow						
17	NDJ	<i>Navicula disjuncta</i> Hustedt.						
18	NGR	<i>Navicula germanii</i> Wallace. Kutz						
20	NRO	<i>Navicula radiosa</i>						
23	NSL	<i>Navicula salinarum</i> Var. intermedia						
24	NGL	<i>Navicula gibbula</i> Cleve						
25	NCC	<i>Navicula cryptocephala</i> Kutzing						
26	NRH	<i>Navicula reinhardtii</i> Grunow.						
27	NAL	<i>Nitzschia acicularis</i> (Kutzing) W.Smith						
27	PAS	<i>Pinnularia</i> <i>acrosphaeria</i> .Breb						

			Yellow	Green	Red	Red	Yellow	Yellow
28	PGB	<i>Pinnularia gibba</i> Ehr	Green	Green	Yellow	Yellow	Yellow	Yellow
29	PNG	<i>Pinnularia neglecta</i> Mayer	Red	Red	Red	Red	Green	Green
30	POV	<i>Pinnularia ovata</i> Krammer	Green	Green	Red	Red	Red	Red
31	STB	<i>Synedra tabulata</i> (C.Agardh)Kutzing.	Red	Red	Red	Red	Yellow	Yellow
32	TBL	<i>Tabellaria</i> sp.	Yellow	Green	Green	Green	Red	Red
33	ULU	<i>Ulnaria ulna</i> (Nitzsch) Compère	Green	Yellow	Green	Green	Yellow	Green

