



RESEARCH ARTICLE

Phytoplankton dynamics and pollution impacts in the Sundarbans estuarine ecosystem

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Abstract

Sundarbans belong exclusively to the largest mangrove ecosystem in the world. This work aims to track the spatiotemporal succession of phytoplankton relying upon microscopic study and hydrological parameters circumscribing five different locations of the Indian Sundarbans. Phytoplankton levels, the major contributors to aquatic productivity, should be kept under check to maintain a balanced ecosystem. Statistically validated environmental parameters and phytoplankton dissemination were thoroughly examined on a seasonal and spatial basis from April 2022 to February 2024. With regard to biomass, Bacillariophyceae exhibited prominence with maximum species richness (3.96), diversity (3.67) and assemblage observed during summer (4.7×10^3 ind/l). However, as winter commenced, the immense tourist load on 3 of our sampling stations, Gadkhali, Kumirmari and Dobanki, serving as the main tourist attraction spots, started facing pollution pressure attributable to garbage disposal, poor sanitation and littering from mechanized tourist vessels. This was further reflected in the phytoplankton community as a sudden surge in the Dinoflagellate population owing to blooms of *Ceratium* sp. (15.6×10^3 ind/l) was observed, indicating pollution pressure in the waterbodies, further validated by the increased heavy metal concentration (Lead, Cadmium and Chromium) and COD levels (47.34 ± 3.09 mg/l) during this season. A multidimensional scaling map further validated the similarity trend across these stations, pointing towards the implications of uncontrolled tourism as the probable cause. Therefore, it is essential to impose ecotourism practices keeping in mind the tourist carrying capacity of the ecosystem.

Keywords: diatom; Dinophyceae; phytoplankton; pollution

Introduction

Sundarbans, constituting a massive block of halophytic mangroves, stretch across 4,260 km² circumscribing both the North and South 24 Parganas, spanning from the Hooghly River in West Bengal to Bangladesh's Baleshwar River (1). This deltaic ecosystem is rich in species biodiversity involving a congregation of planktons (phytoplankton as well as zooplankton), amphibians, microorganisms and mammals along with some benthos species (2). Innumerable estuaries, tidal rivers and creeks intersect along the Sundarbans making it encounter an annual precipitation of about 1600-1800 mm. Furthermore, this region exhibits as a nature's barrier against various cyclonic storms (3). UNESCO in 1974 and IUCN in 1989 declared the Sundarbans as a "World Heritage Site"; and as a "Ramsar site" in 2019 because of its unparalleled floral and faunal aggregation (4, 5). In the course of time, unfortunately, this biodiversity-rich zone is facing threats due to enhanced levels of industrialization, climate change, oil spillage, pollution and sea level rise (6). These, in turn, further start eroding the land alongside increasing the waterbodies' particulate matter load, which directly disrupts the aquatic ecosystem balance, posing a deadly impact on the phytoplankton dynamics (7).

Phytoplankton, being the fundamental autotrophic drifters of the aquatic ecosystem, act as an indicator species as per environmental changes and their availability reflects alterations in the estuary's physicochemical conditions (8). They act as the predominant biotic organisms in the aquatic food webs, considering their multifunctionality in producing oxygen and sequestering carbon (9). Enhanced pollution levels of Sundarbans directly get reflected in the plankton community as they absorb the available nutrients for their growth and multiplication, further triggering algal blooms and eutrophication (2). Hence, it is essential to estimate phytoplankton as it poses great economic benefits, at the same time maintaining ecosystem balance (8). With each passing year, enhanced tourist pressure poses a threat to this ecosystem attributable to pollution induced by direct garbage disposal, improper sanitation and wastes released from mechanized vessels that elicit the levels of eutrophication (10). Tourism in a mangrove ecosystem impacts phytoplankton dynamics negatively by enhancing the water pollution levels caused due to improper litter disposal, food scraps, fuel leaks from boats, sewage runoff and other wastes which decompose in the water and alter the nutrient levels outcompeting phytoplankton and causing algal bloom (11, 12). This disrupts the ecosystem balance,

which is otherwise necessary for healthy phytoplankton growth. Alongside, boat traffic and human interference increase the levels of turbidity in water directly impacting the available light for the growth of phytoplankton (11, 13). The primary focus of this study is to achieve a comprehensive understanding of the dynamics of phytoplankton communities in selected less explored regions of the Indian Sundarbans. Additionally, it aims to investigate how these dynamics vary in response to seasonal changes and environmental parameters, thereby providing valuable insights into their intricate relationships within this ecosystem. The sampling sites have been strategically selected to encompass estuaries that experience low and high tourist activity, which will disclose the effect of pollution caused mainly due to anthropogenic interferences on this deltaic waterbody, making our study significant. This can be efficacious as a reference for more detailed plankton research from this arena, including analyzing how changes in phytoplankton communities influence zooplankton populations and their grazing, thereby affecting the marine food web. Additionally, exploring the genetic diversity of phytoplankton would enhance the understanding of their adaptability and evolutionary responses to changing environments.

Materials and Methods

Study sites

The study was implemented through a period of two years (from April 2022 to February 2024) across three main seasons: Pre-monsoon (summer), Monsoon and post-monsoon (winter). A total five sampling spots, mainly estuaries, which were at least 5 km apart from each other in the north-to-south direction spanning through the stretch of the Indian Sundarbans, referred to as Frasersganj (at the junction of the Matla River with the Bay of Bengal), Gadkhali (on the Matla and Bidyadhari river), Kumirmari (on Bidyadhari and

Raimangal), Dobanki (at the junction of Bidyadhari River with the Bay of Bengal near the five river points) and Canning (near the margin of the Matla River) (Fig. 1) were chosen. The selection was made maintaining an ecological gradient starting from Frasersganj as the lowest point of Indian Sundarbans and closest to the Bay of Bengal with low anthropogenic interferences including tourism to Canning as the highest point and far away from the Bay of Bengal. In contrast, Gadkhali, Kumirmari and Dobanki lie midway in the heart of the Indian Sundarbans and act as the main tourist-attracting sites (Table 1). Samples were collected in triplicates and processed from each spot per season.

Hydrological parameter analysis

The mouth of the estuary for each sampling station has been considered as the sampling point and the GPS coordinates for the same have been recorded. From each of the five chosen spots, several hydrological parameters of water, including the pH, Temperature, Dissolved oxygen (DO), Salinity and Total Dissolved Solids (TDS), were analyzed by deploying a Hanna multiparameter probe. The probe was calibrated using a quick calibration liquid provided by the manufacturer and then from the boat, the probe was dipped into the water under standstill conditions to avoid major fluctuations. Triplicate readings for each hydrological parameter were recorded to minimize error. Nutrients such as the inorganic nitrate (NO_3^-), inorganic silicate (SiO_3^{2-}) and inorganic phosphate (PO_4^{3-}) were measured initially by filtering the water samples using a GF/F filter paper followed by storing the filtrate on ice. Later, the samples were returned to the lab and kept in a -20°

Table 1. Sampling points with their respective GPS coordinates

Sampling station	Latitude	Longitude
Fraserganj	21°34.880'N	088°14.192'E
Dobanki	22°00.857'N	088°45.567'E
Gadkhali	22°10.070' N	088°47.586'E
Kumirmari	22°10.576'N	088°55.684'E
Canning	22°18.986'N	088°40.462'E

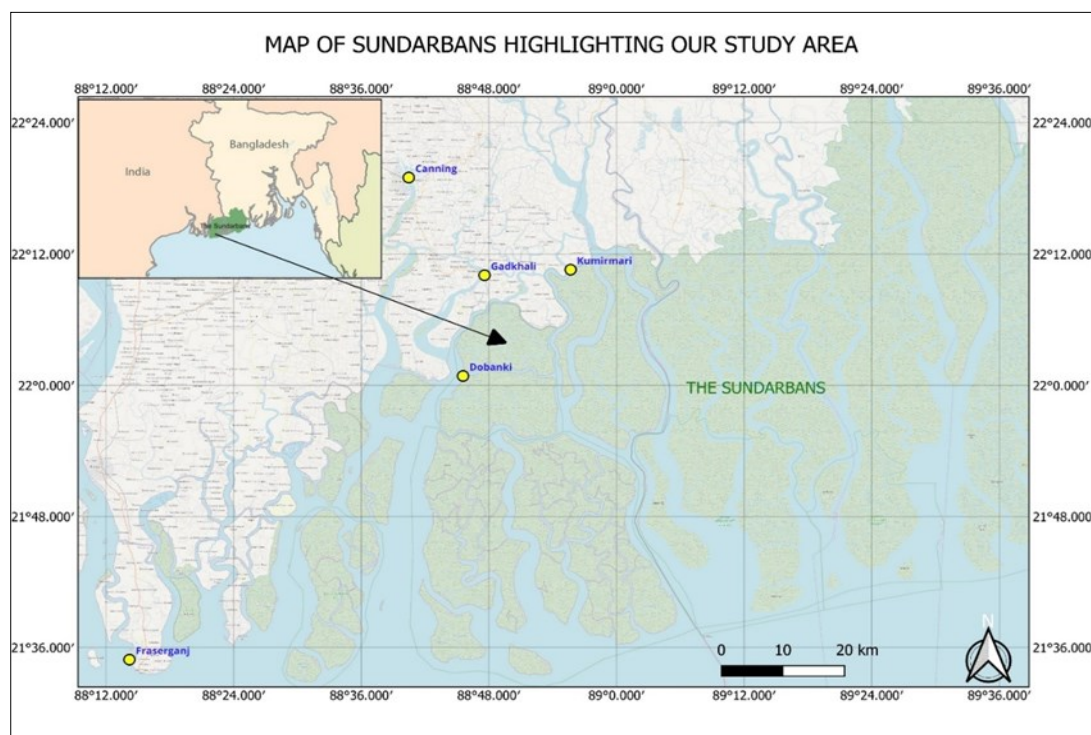


Fig. 1. Map of Sundarbans showing the sampling stations with an arrow indicating lesser to higher anthropological interference.

freezer until further analysis. Nutrient content was estimated following the methods described in (14).

Water pollution analysis

Water samples were collected from a depth of 50 cm and filtered using GF/F filter paper for heavy metal analysis (Lead, Cadmium and Chromium). The samples were returned to the lab under chilled conditions and kept in a -20° freezer until further analysis. Winkler's titrimetric method, as illustrated in (15, 16), also estimated the chemical oxygen demand (COD) and biological oxygen demand (BOD) of water.

Phytoplankton study

For sampling phytoplankton, revised methods, as illustrated by (17), were followed. A plankton net of 20 µ mesh size and 28.5 cm diameter attached with a mechanical flowmeter (Hydro-Bios) was towed from the boat for 3 min (Eqn. 1) for each round of plankton sampling. The initial and final readings displayed in the flowmeter, before and after towing the net, were recorded for future calculation purposes. From each spot, triplicate samples were collected, followed by immediate preservation of the same using 4 % formalin (2). In the case of diatom-rich samples, frustule cleaning with 3 % chromic acid (18) followed by repeated centrifugation to eliminate the extra acid from the sample and washing with distilled water becomes mandatory as it eliminates the organic components from the sample, keeping behind only the silicified cell wall (7).

Taxonomic enumeration

The preserved phytoplankton sample was analyzed under Carl Zeiss Primostar 3 microscope. Sedgewick-Rafter counting chamber (Eqn. 2) was employed for enumerating their abundance (19) which gets expressed in terms of individuals per liter (ind. L⁻¹) (8). A minimum of 3 rafter readings per sample were taken. Before rafter analysis, flowmeter readings were analyzed to get an idea regarding the amount of water that has passed through the net during each round of towing.

Amount of water passed through the net (in liters) =
(Final reading – Initial reading) × 0.3 × πr² × 1000

(Eqn.1)

[where 0.3 is the flowmeter constant; πr² indicates the mouth area of phytoplankton net; r is the radius of the mouth of phytoplankton net (in our case, r = 30 cm)]

Rafter abundance calculation (in individuals/liter) =

$$\left(\frac{\text{Final sample volume}}{\text{Amount of water passed through the net}} \right) \times 1000$$

Eqn.2

Shannon-Weiner diversity index (H) (20), species richness (d) (21) and Pielou's species evenness (22) were also calculated. Later, scanning electron microscopy of the frustule-cleaned samples was carried out and they were identified based on several predefined taxonomic keys (23, 24, 25, 26).

Statistical analysis

The data were statistically validated using SPSS software by specific analytical tests: Two - way ANOVA to analyze if the two independent variables (different study sites and different seasons) simultaneously affect a dependent variable (hydrological and nutrient parameters), this will allow us to not only determine the effect of each variable but also understand whether there is significant interaction between them. After ANOVA showed significant differences among the variables, the Post Hoc Tukey test was performed to identify exactly where the differences lie to gain more detailed insights into our data. Next, Karl Pearson's correlation coefficient was analyzed as it specifically measures the linear relationship between the variables, meaning as one variable increases, the other should also increase (positive correlation) or decrease (negative correlation) at a consistent rate, highlighting the interdependency among two variables. Furthermore, Pearson's chi-square test of association was estimated to authenticate whether the abundance of the phytoplankton species varies with each passing season and lastly, Multidimensional Scaling Map (MDS) to visually represent the relationship between our study sites based on their pairwise similarities revealing the structure of a high-dimensional complex data by plotting them in a lower two-dimensional interface.

Results

Hydrological parameters

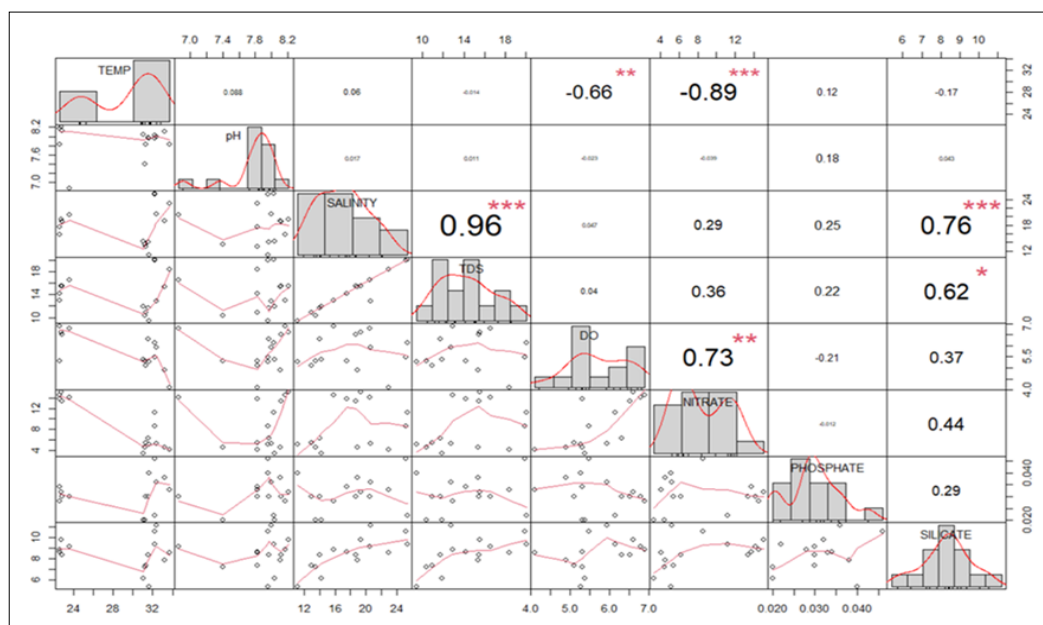
Spatiotemporal variability of all the hydrological parameters (sample number = 45; for five spots three replicate values were taken per season) has been delineated in Table 2. Two-way ANOVA with three readings per parameter; furthermore, the post hoc Tukey test was performed to test the significance based on two variables- seasonal and spatial. The tests revealed statistical seasonal significance i.e. p-value < 0.05 in case of temperature (F value 861.864, p-value 4.55e⁻¹⁰), salinity (F value 24.788, p-value 0.000373), TDS (F value 13.783, p-value 0.00256), nitrate (F value 30.286, p-value 0.000185) and silicate (F value 7.358, p-value 0.0154). Post hoc Tukey test showed a significant difference in TDS of post-monsoon with the monsoon and pre-monsoon with the monsoon. In the case of Nitrate, Post-monsoon with monsoon values and pre-monsoon with post-monsoon values differed significantly. Alongside, significant change in silicate levels for post-monsoon with monsoon and pre-monsoon with monsoon was detected. Karl Pearson's correlation coefficient analysis (Fig. 2) highlighted a positive correlation of salinity with TDS (r = 0.96; p < 0.05) and silicate (r = 0.76; p < 0.05) besides DO with nitrate (r = 0.73; p < 0.05). Negative correlation was documented for temperature with DO (r = -0.66; p < 0.05) and Temperature with Nitrate (r = -0.89; p < 0.05) (Fig. 2).

Delving into the water pollution analysis, it was observed that the concentration of all the three heavy metals, Lead (2.56 mg/l), Cadmium (0.07 mg/l) and Chromium (1.52 mg/l) escalated during the post-monsoon season, especially from 3 of our sampling sites Gadkhali, Kumirmari and Dobanki (Fig. 3). Two-way ANOVA revealed a significant statistical difference of Lead (F value 31.818, p-value 0.000156), Cadmium (F value 7.766, p-value 0.0134) and

Table 2. Cumulative variability of hydrological parameters across all the 5 sampling stations of Sundarbans along 3 different seasons

SEASON	SITE	TEMP (°C)	pH	SALINITY (psu)	TDS (ppt)	DEPTH (m)	TRANSPAR-ENCY (inch)	DO (mg/L)	NO ³⁻ (mg/l)	PO ₄ ³⁻ (mg/l)	SiO ₃ ²⁻ (mg/l)
Pre-M (Pre-Monsoon)	FRASERGANJ	33.21 ± 0.44	8.11 ± 0.12	18.91 ± 0.11	15.39 ± 0.13	1.4 ± 0.2	6.5 ± 0.14	4.90 ± 0.2	4.5 ± 0.15	0.038 ± 0.3	7.9 ± 0.06
	GADKHALI	32.21 ± 0.31	8.03 ± 0.28	25.41 ± 0.15	20.05 ± 0.55	7.3 ± 0.25	18 ± 0.1	6.15 ± 0.12	11.2 ± 0.1	0.022 ± 0.22	9.4 ± 0.1
	KUMIRMARI	32.38 ± 0.21	8 ± 0.29	20.58 ± 0.24	12.80 ± 0.20	6.3 ± 0.3	22 ± 0.06	5.93 ± 0.18	5.2 ± 0.26	0.036 ± 0.15	11.2 ± 0.15
	DOBANKI	32.25 ± 0.188	7.96 ± 0.17	25.22 ± 0.29	19.91 ± 0.36	8.1 ± 0.2	24 ± 0.02	5.48 ± 0.18	8.6 ± 0.15	0.046 ± 0.15	10.6 ± 0.3
	CANNING	33.74 ± 0.41	7.83 ± 0.32	23.01 ± 0.34	18.39 ± 0.19	1 ± 0.5	10 ± 0.3	4.11 ± 0.36	4.1 ± 0.19	0.033 ± 0.2	8.6 ± 0.2
Mon (Monsoon)	FRASERGANJ	31.57 ± 0.075	7.97 ± 0.54	14.03 ± 0.22	11.7 ± 0.4	1.5 ± 0.2	10.3 ± 0.01	6.31 ± 0.07	6.22 ± 0.3	0.03 ± 0.2	8.40 ± 0.1
	KUMIRMARI	31.28 ± 0.075	7.83 ± 0.15	12.99 ± 0.2	10.9 ± 0.4	9.8 ± 0.2	12 ± 0.5	5.1 ± 0.1	5.39 ± 0.2	0.03 ± 0.2	7.40 ± 0.08
	DOBANKI	31.03 ± 0.3	8.04 ± 0.11	14.25 ± 0.12	11.87 ± 0.3	2.8 ± 0.1	17.5 ± 0.1	5.38 ± 0.1	3.28 ± 0.2	0.02 ± 0.09	6.20 ± 0.06
	GADKHALI	31.6 ± 0.11	7.96 ± 0.29	11.12 ± 0.04	9.39 ± 0.5	4.2 ± 0.47	18 ± 0.04	5.26 ± 0.1	5.16 ± 0.14	0.04 ± 0.01	5.40 ± 0.08
	CANNING	31.2 ± 0.37	7.4 ± 0.16	13.46 ± 0.14	10.36 ± 0.3	1.6 ± 0.25	11 ± 0.09	5.3 ± 0.4	4.45 ± 0.18	0.02 ± 0.01	7.23 ± 0.07
Post-M (Post-Monsoon)	FRASERGANJ	23.54 ± 0.05	6.86 ± 0.3	20.47 ± 0.1	16.58 ± 0.14	1.6 ± 0.15	14 ± 0.03	6.79 ± 0.02	14.2 ± 0.6	0.03 ± 0.03	9.2 ± 0.03
	CANNING	22.8 ± 0.1	8.12 ± 0.1	18.73 ± 0.1	15.43 ± 0.15	2.1 ± 0.2	13 ± 0.8	6.52 ± 0.01	13.4 ± 0.04	0.03 ± 0.1	8.4 ± 0.07
	GADKHALI	22.6 ± 0.37	7.83 ± 0.02	17.52 ± 0.06	14.2 ± 0.15	1.8 ± 0.1	15.8 ± 0.06	5.3 ± 0.4	13.8 ± 0.5	0.034 ± 0.2	8.7 ± 0.1
	KUMIRMARI	22.6 ± 0.15	8.17 ± 0.01	15.82 ± 0.07	12.95 ± 0.22	6.2 ± 0.1	12.24 ± 0.01	6.9 ± 0.02	14.6 ± 0.08	0.028 ± 0.05	8.9 ± 0.1
	DOBANKI	22.7 ± 0.11	8.21 ± 0.17	19.35 ± 0.1	15.52 ± 0.25	6.9 ± 0.14	24 ± 0.5	6.61 ± 0.08	15.1 ± 0.02	0.032 ± 0.03	9.8 ± 0.09

Data represented as Mean ± S.E. [DO = Dissolved Oxygen, NO³⁻ = Total Nitrate, PO₄³⁻ = Total Phosphate, SiO₃²⁻ = Total Silicate].



** Correlation is significant at the 0.05 level (2-tailed), *** Correlation is significant at the 0.01 level (2-tailed)

Fig. 2. Karl Pearson Correlation chart showing the scatterplot and correlation coefficients for all the hydrological parameters.

Chromium (F value 77.61, p-value 5.77e⁻⁰⁶**) varying across seasons. Additionally, the calculated COD values ranged between 14.2–16.2 mg/l during monsoon and 18.5–21.2 mg/l during Pre-Monsoon across all the stations. COD values their peak during the winter season, displaying 47.34 ± 3.09 mg/l in Dobanki, 24.56 ± 1.79 mg/l in Kumirmari and 35.83 ± 3.51 mg/l in Gadkhali. The water pollution levels exceeded the permissible range (less than 10 mg/l) set by the World Health Organization (24). Additionally, the BOD values ranged from 3.82 to 9.32 mg/l, with the highest and lowest values recorded at Dobanki and Fraserganj, respectively.

Phytoplankton dissemination

Phytoplankton belonged to 6 major classes comprising 37 different species from our study. The dominant category throughout the study period was Bacillariophyceae (Fig. 4C, 4D, 5J, 5K, 5L, 5M, 5N, 5P, 5Q) with limited appearance of Cyanophyceae members, especially *Dolichospermum* sp. (Fig. 4E), *Pseudanabaena* sp. during the monsoon season. Dinophyceae members summed up to 10 % of the population (Fig. 4F) during post-monsoon comprising four genera: *Ceratium* sp., *Pyrocystis* sp., *Prorocentrum* and *Boreadinium*. Similar findings were observed during the winter season (2). Hence, the dominance hierarchy reveals

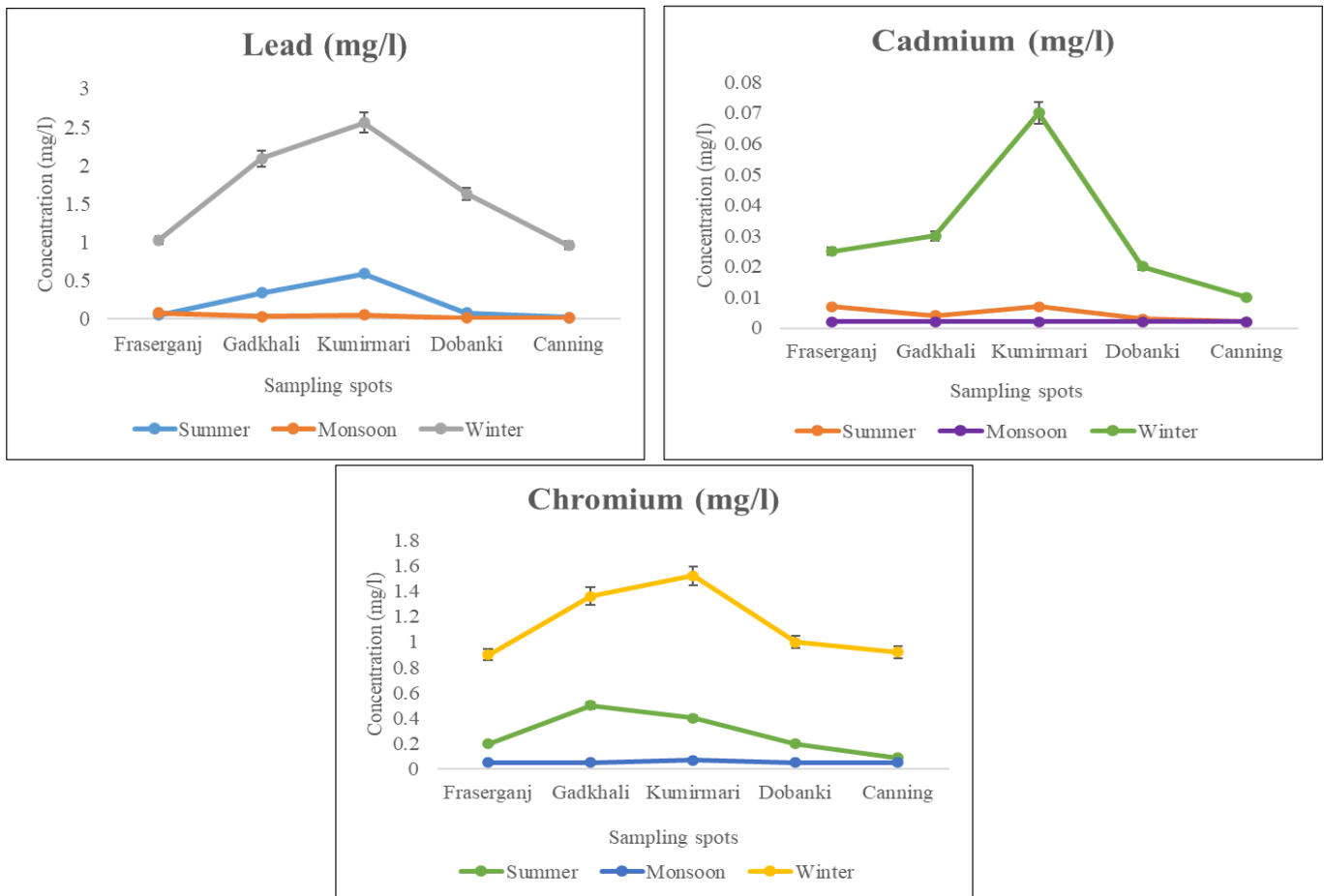


Fig. 3. Estimation of Lead (Pb), Cadmium (Cd) and Chromium (Cr) (mg/l) of water across different seasons by APHA 24th Edition, 2023; 3111B method.

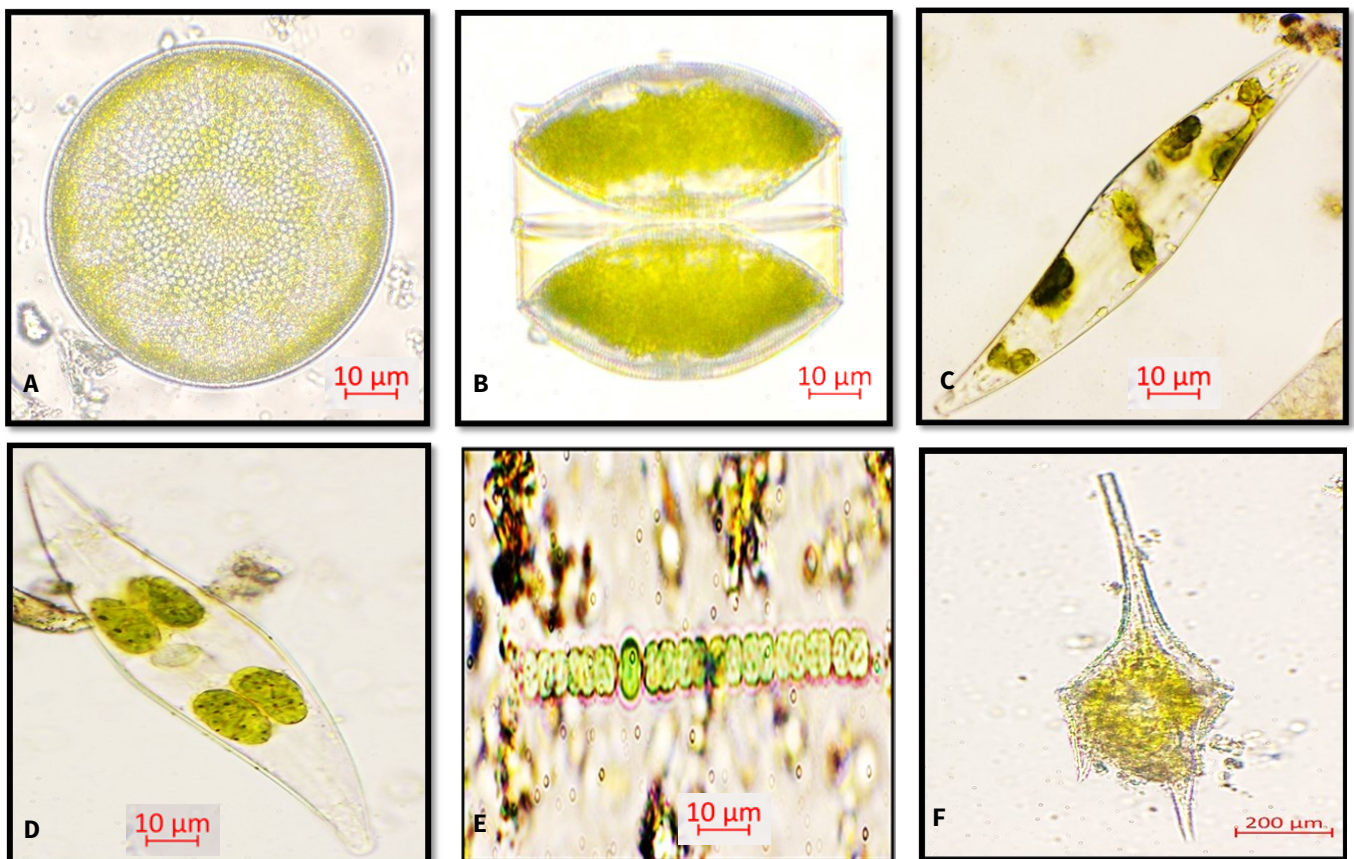


Fig. 4. (A-F) Compound microscope images of few Phytoplankton species.

(A) *Coscinodiscus* sp.; (B) *Coscinodiscus* sp. (girdle view); (C) *Pleurosigma* sp.; (D) *Gyrosigma* sp.; (E) *Dolichospermum* sp.; (F) *Ceratium* sp. (21).

Bacillariophyceae > Dinophyceae > Cyanophyceae (Fig. 6) (7). Blooms of *Ceratium* sp. (biomass 15.6×10^3 ind/l) (COD 47.34 ± 3.09 mg/l) during winter (Table 4) were observed from 3 of our sampling stations: Gadkhali, Kumirmari and Dobanki (27). 18 taxa belonged to Centrales, among which most abundant were *Coscinodiscus*, *Thalassiosira*, *Chaetoceros*, *Proboscia* and *Cylindrotheca*. On the other hand, 19 taxa belonged to Pennates, the most abundant being *Pleurosigma*, *Gyrosigma*, *Nitzschia* and *Navicula* (Table 3) (cumulative abundance of each > 5000 individuals L^{-1}). In contrast, *Skabitschewskia* sp., *Boreadinium* sp., *Surirella* sp., *Eucampia* sp., etc. were limited to their native waterbody, implying their low adaptability to altering environmental conditions.

The phytoplankton population in the Sundarbans halophytic ecosystem flourishes during Pre-Monsoon (June),

with maximum abundance observed from Dobanki (mean, 4.7×10^3 ind/l) and minimum from Fraserganj (mean, 2.4×10^3 ind/l) (Fig. 7). A sharp decline in the species diversity during the monsoon season (September) was recorded across all the stations (mean, Fraserganj = 1.15×10^2 ind/l; Gadkhali = 2.09×10^3 ind/l, Kumirmari = 1.8×10^3 ind/l, Dobanki = 2.12×10^3 ind/l and Canning = 1.4×10^2 ind/l). On the contrary, a noticeable increase in one particular group of phytoplankton, i.e., the Dinophyceae members, was noted during the Post-Monsoon (February) season. This data thus unveils the seasonal variability in phytoplankton dynamics across all our sampling stations of the Indian Sundarbans (Table 3). Throughout our study, both species richness (d) and species diversity (H) were quite high. Coming to species richness, it reached its peak at Dobanki (3.96) during Pre-Monsoon (June). The Shannon-Weiner Diversity index also maximised during this time from

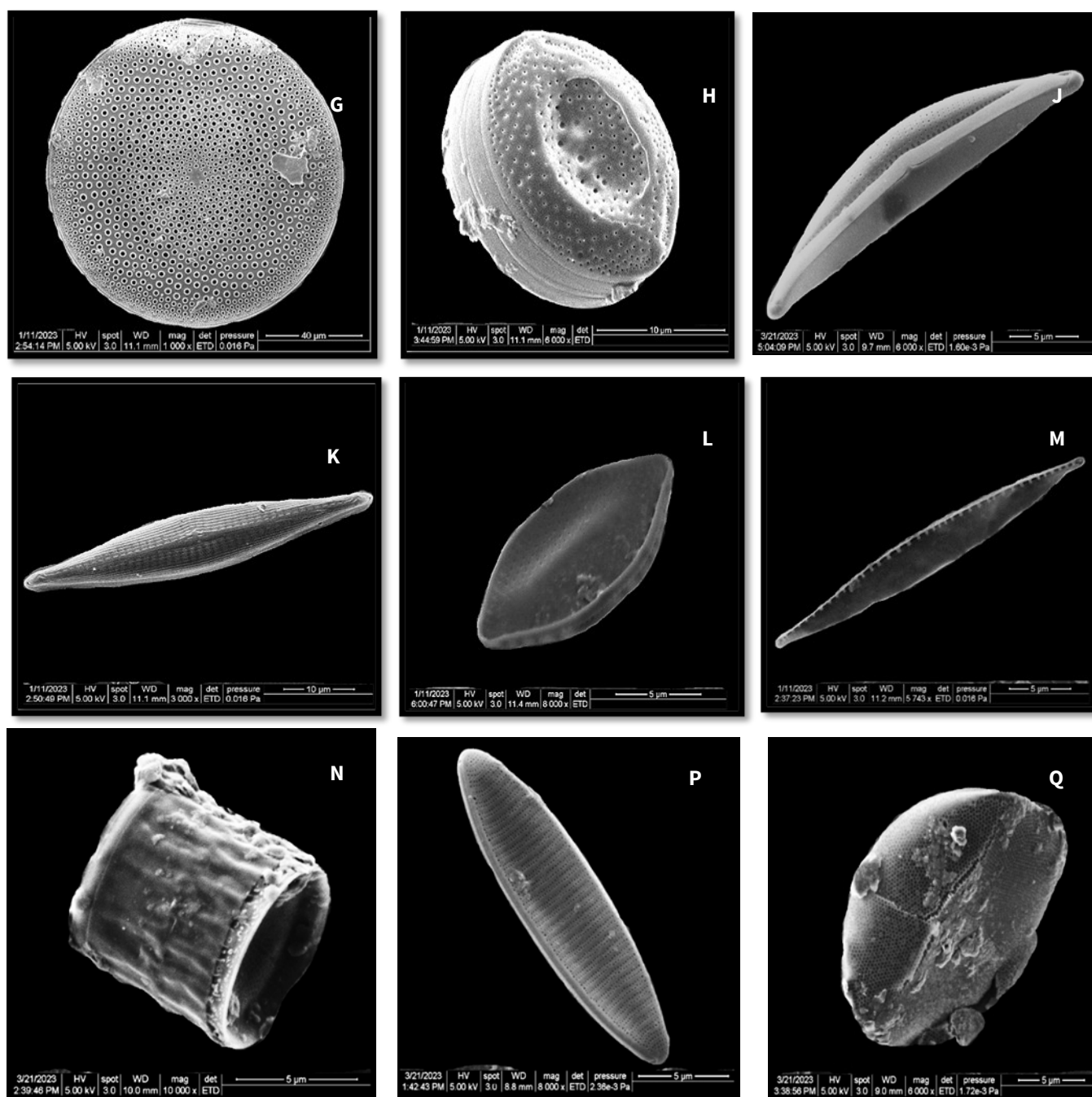


Fig. 5. (G-Q): SEM pictures of few Phytoplankton species observed.

(G) *Coscinodiscus* sp.; (H) *Coscinodiscus* sp. (21); (J) *Cymbella* sp. (22); (K) *Navicula* sp. (identified by Botanical Survey of India); (L) *Skabitschewskia* sp. (22); (M) *Nitzschia* sp. (23); (N) *Skeletonema* sp. (24); (P) *Nitzschia* sp. (24); (Q) *Cocconeis* sp. (24).

Table 3. Seasonal as well as spatial abundance of several Phytoplankton species

SAMPLING STATION	CLASS	FAMILY	SPECIES	CUMULATIVE ABUNDANCE		
				PRE-MONSOON	MONSOON	POST-MONSOON
FRASERGANJ	Bacillariophyceae	1. Leptocylindraceae	1. <i>Leptocylindrus</i> sp.	+++	-	++
		2. Coscinodiscaceae	2. <i>Coscinodiscus</i> sp.	++++	++	+++
		3. Bacillariaceae	3. <i>Pleurosigma</i> sp.	++	+	+
		4. Bacillariaceae	4. <i>Cylindrotheca</i> sp.	+++	-	++
		5. Bacillariaceae	5. <i>Nitzschia</i> sp.	++	+	++
		6. Coscinodiscaceae	6. <i>Coscinodiscus radiatus</i>	+++	+	++
		8. Bacillariaceae	8. <i>Gyrosigma</i> sp.	++	+	++
		9. Naviculaceae	9. <i>Navicula canalis</i>	+++	+	+
		10. Achnanthidiaceae	10. <i>Achnanthidium</i> sp.	+	-	+
		11. Chaetocerotaceae	11. <i>Chaetoceros</i> sp.	-	+	+
		12. Cocconeidaceae	12. <i>Cocconeis</i> sp.	-	-	++
	Cyanophyceae	13. Nostocaceae	13. <i>Dolichospermum</i> sp.	+	++	+
			Unidentified species	+++	+	+
GADKHALI	Bacillariophyceae	1. Coscinodiscaceae	1. <i>Coscinodiscus radiatus</i>	++++	++	++
		2. Bacillariaceae	2. <i>Cylindrotheca</i> sp.	+++	++	-
		3. Bacillariaceae	3. <i>Pleurosigma</i> sp.	++	+	++
		4. Bacillariaceae	4. <i>Gyrosigma</i> sp.	++	+	++
		5. Chaetocerotaceae	5. <i>Chaetoceros</i> sp.	+++	+++	-
		6. Thalassiosiraceae	6. <i>Thalassiosira</i> sp.	++	+	++
		7. Bacillariaceae	7. <i>Nitzschia</i> sp.	+++	+	++
		8. Leptocylindraceae	8. <i>Leptocylindrus</i> sp.	++	-	++
		9. Rhizosolenaceae	9. <i>Proboscia</i> sp.	++	-	+++
		10. Naviculaceae	10. <i>Tropidoneis</i> sp.	-	+	++
		11. Lithodesmiaceae	11. <i>Ditylum</i> sp.	++	+	-
		12. Naviculaceae	12. <i>Navicula canalis</i>	+++	+	++
		13. Coscinodiscaceae	13. <i>Coscinodiscus</i> sp.	++++	++	+++
		14. Thalassiosiraceae	14. <i>Lauderia</i> sp.	-	-	+
		15. Stephanodiscaceae	15. <i>Stephanodiscus</i> sp.	+	-	-
		16. Cocconeidaceae	16. <i>Cocconeis</i> sp.	++	+	+
		17. Gomphonemataceae	17. <i>Gomphonema</i> sp.	++	-	-
		18. Bacillariaceae	18. <i>Bacillaria</i> sp.	-	+	+
		19. Stephanodiscaceae	19. <i>Cyclotella</i> sp.	+	-	++
			Unidentified species	+++	+	+
	Dinophyceae	20. Ceratiaceae	20. <i>Ceratium</i> sp.	-	+	+++
		21. Diplosaliaceae	21. <i>Boreadinium</i> sp.	+	-	++
		22. Pyrocystaceae	22. <i>Pyrocystis</i> sp.	+	-	++
	Cyanophyceae	23. Pseudanabaenaceae	23. <i>Pseudanabaena</i> sp.	+	++	+
KUMIRMARI	Bacillariophyceae	1. Bacillariaceae	1. <i>Nitzschia</i> sp.	+++	+	++
		2. Naviculaceae	2. <i>Navicula canalis</i>	++	+	++
		3. Bacillariaceae	3. <i>Pleurosigma</i> sp.	+++	+	++
		4. Coscinodiscaceae	4. <i>Coscinodiscus radiatus</i>	++++	++	++
		5. Bacillariaceae	5. <i>Gyrosigma</i> sp.	+++	+	++
		6. Coscinodiscaceae	6. <i>Cymbella</i> sp.	++	++	-
		7. Bacillariaceae	7. <i>Cylindrotheca</i> sp.	++	++	+
		8. Lithodesmiaceae	8. <i>Ditylum</i> sp.	+	-	+
		9. Thalassionemataceae	9. <i>Thalassiothrix</i> sp.	+	++	-
		10. Coscinodiscaceae	10. <i>Coscinodiscus</i> sp.	+++	++	+++
		11. Naviculaceae	11. <i>Tropidoneis</i> sp.	++	+	-
		12. Leptocylindraceae	12. <i>Leptocylindrus</i> sp.	+++	-	++
		13. Hemiaulaceae	13. <i>Eucampia</i> sp.	+	+	+
		14. Rhizosolenaceae	14. <i>Proboscia</i> sp.	-	-	+++
		15. Thalassiosiraceae	15. <i>Thalassiosira</i> sp.	-	-	++
		16. Skeletonemataceae	16. <i>Skeletonema</i> sp.	++	-	+
		17. Stephanodiscaceae	17. <i>Stephanodiscus</i> sp.	++	+	-
		18. Fragilariaceae	18. <i>Fragilaria</i> sp.	+	-	++
		19. Achnanthidiaceae	19. <i>Achnanthidium</i> sp.	+	-	-
			Unidentified species	++	+	+
	Dinophyceae	20. Ceratiaceae	<i>Ceratium</i> sp.	-	+	+++
		21. Prorocentraceae	<i>Prorocentrum</i> sp.	+	+	++
		22. Pyrocystaceae	<i>Pyrocystis</i> sp.	++	-	++

DOBANKI	Bacillariophyceae	1. Bacillariaceae	1. <i>Cylindrotheca</i> sp.	+++	-	++
		2. Leptocylindraceae	2. <i>Leptocylindrus</i> sp.	+++	-	++
		3. Lithodesmiaceae	3. <i>Ditylum</i> sp.	+	-	++
		4. Thalassiosiraceae	4. <i>Lauderia</i> sp.	+	-	++
		5. Rhizosolenaceae	5. <i>Proboscia</i> sp.	+++	-	++
		6. Coscinodiscaceae	6. <i>Coscinodiscus</i> sp.	++++	++	+++
		7. Amphipleuraceae	7. <i>Amphipleura</i> sp.	+++	++	-
		8. Coscinodiscaceae	8. <i>Cymbella</i> sp.	++	+	-
		9. Bacillariaceae	9. <i>Gyrosigma</i> sp.	+++	+	++
		10. Coscinodiscaceae	10. <i>Coscinodiscus radiatus</i>	++++	++	++
		11. Bacillariaceae	11. <i>Nitzschia</i> sp.	+++	+	++
		12. Melosiraceae	12. <i>Melosira</i> sp.	+	+	-
		13. Bacillariaceae	13. <i>Pleurosira</i> sp.	+++	+	++
		14. Thalassiosiraceae	14. <i>Thalassiosira</i> sp.	++	++	-
		15. Naviculaceae	15. <i>Navicula canalis</i>	++	+	++
		16. Bacillariaceae	16. <i>Surirella</i> sp.	-	++	-
		17. Achnanthidiaceae	17. <i>Skabitschewskia</i> sp.	+	-	+
		18. Rhizosoleniaceae	18. <i>Rhizosolenia</i> sp.	-	-	+
		19. Stephanodiscaceae	19. <i>Stephanodiscus</i> sp.	+	+	+
		20. Rhizosoleniaceae	20. <i>Guinardia</i> sp.	++	-	+
		21. Thalassionemataceae	21. <i>Thalassionema</i> sp.	-	+	++
			Unidentified species	++	+	+
	Dinophyceae	22. Ceratiaceae	22. <i>Ceratium</i> sp.	-	-	+++
	Cyanophyceae	23. Nostocaceae	23. <i>Dolichospermum</i> sp.	-	++	+
Canning	Bacillariophyceae	1. Thalassionemataceae	1. <i>Thalassiothrix</i> sp.	-	+	-
		2. Naviculaceae	2. <i>Navicula</i> sp.	+++	+	++
		3. Coscinodiscaceae	3. <i>Coscinodiscus</i> sp.	+++	++	+++
		4. Bacillariaceae	4. <i>Nitzschia</i> sp.	+++	+	++
		5. Bacillariaceae	5. <i>Pleurosira</i> sp.	++	+	+
		6. Bacillariaceae	6. <i>Gyrosigma</i> sp.	++	-	++
		7. Bacillariaceae	7. <i>Cylindrotheca</i> sp.	++	-	+++
		8. Rhizosolenaceae	8. <i>Proboscia</i> sp.	-	-	+
		9. Amphipleuraceae	9. <i>Amphipleura</i> sp.	-	-	++
		10. Catenulaceae	10. <i>Amphora</i> sp.	-	+	-
		11. Coscinodiscaceae	11. <i>Coscinodiscus radiatus</i>	+	++	+
		12. Achnanthidiaceae	12. <i>Planothidium</i> sp.	+	+	-
		13. Coscinodiscaceae	13. <i>Cymbella</i> sp.	++	+	++
		14. Cocconeidaceae	14. <i>Cocconeis</i> sp.	++	++	-
			Unidentified species	+	+	++
	Dinophyceae	15. Ceratiaceae	15. <i>Ceratium</i> sp.	-	+	++
	Cyanophyceae	16. Nostocaceae	16. <i>Dolichospermum</i> sp.	+	++	+

'+' indicates presence of a particular phytoplankton species, with a greater number of '+' signs such as '++' and '+++' indicating their abundance in that particular waterbody; whereas '-' indicates absence of a species in that regime

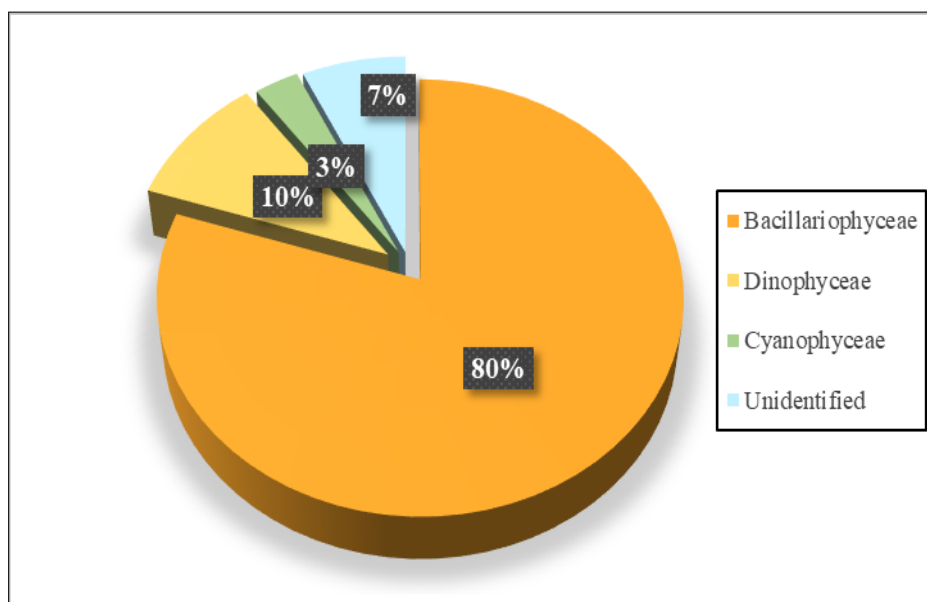


Fig. 6. Phytoplankton members belonging to different classes.

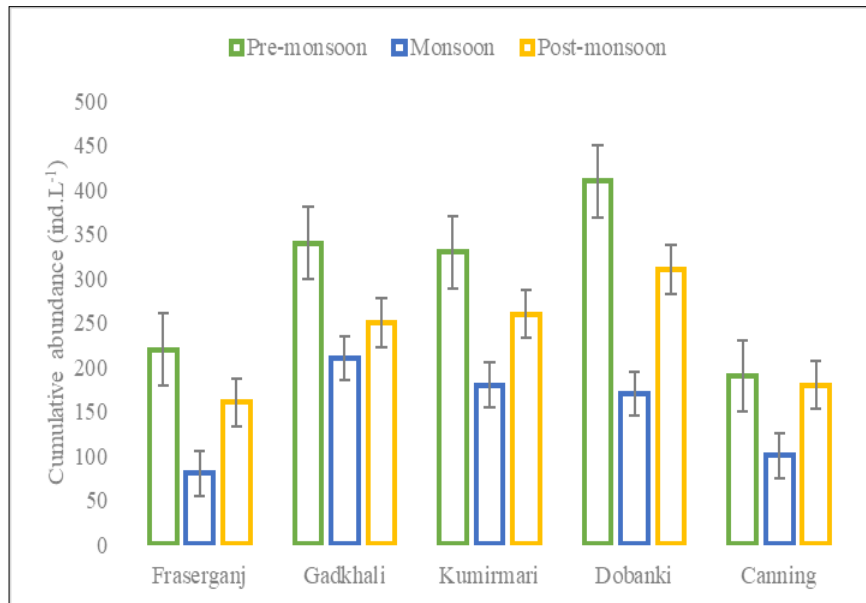


Fig. 7. Seasonal variability of the cumulative abundance (ind. L⁻¹) of phytoplankton community across 5 sampling stations.

Table 4. Abundance of Dinoflagellates across all seasons throughout our study period

Species	Abundance (individuals/litre)		
	Pre-Monsoon	Monsoon	Post-Monsoon
<i>Ceratium</i> sp.	-	7.8×10^2	15.6×10^3
<i>Prorocentrum</i> sp.	10.8×10^2	5.9×10^2	4.7×10^3
<i>Pyrocystis</i> sp.	23.8×10^2	-	5.1×10^3
<i>Boreadinium</i> sp.	97	-	1.8×10^3

Gadkhali (2.91) and Dobanki (3.67). In contrast, the lowest diversity index was displayed from Fraserganj (1.82), declining towards the onset of the monsoon. Moving forward to species evenness (J), the highest value was achieved from Kumirmari (2.58) during Post-Monsoon (February); also, the species evenness values invariably remained greater than 1.

Next, we carried out a Pearson's chi-square association test to authenticate whether the abundance of the

phytoplankton species varies with each passing season. The p-value < 0.05 (X-squared = 51383, p-value < $2.2e^{-16}$) suggested a distinct association between them. Interestingly, in the sampling stations Gadkhali, Kumirmari and Dobanki, the species composition was more or less similar (Table 3).

To deduce the degree of similarity among the phytoplankton species collected from different sampling sites, a Multidimensional scaling map (MDS) was constructed (Fig. 8). The map further validated that the stations Fraserganj and Canning formed two separate groups. In contrast, Gadkhali and Kumirmari were more similar to each other than Dobanki. The stress value was found to be 0.00024 (< 0.01) [(Stress I 0.015, Stress II 0.034 with an optimal scaling factor of 1), (Dispersion Accounted for, i.e., D.A.F 0.99)] which implied outstanding ordinance pattern extrapolating perfect distance among our samples as a good representation of data.

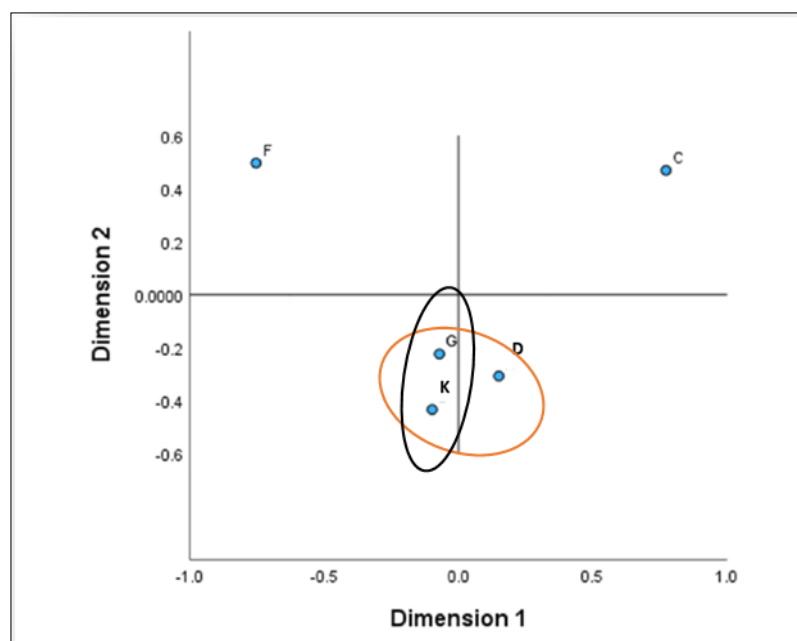


Fig. 8. Multidimensional scaling map performed based on the similarity in phytoplankton dynamics observed from each study site [F: Fraserganj; G: Gadkhali; K: Kumirmari; D: Dobanki; C: Canning].

Discussion

Intensive research on different locations of the Sundarbans has been carried out, focusing on the diversity and assemblage patterns of the phytoplankton community; diatoms were most abundant among the lot. Pennales (represented by 19 taxa) were profusely found during summer, whereas Centrales (represented by 18 taxa) during winter. The otherwise dominant pennate diatoms might have been replaced during the monsoon season owing to intense tidal fluctuations as they settle more slowly than their centric counterparts (27). A similar trend was recorded in previous studies (1, 2). Among the hydrological parameters, temperature varied proportionally per the amount of incident light on the water surface (7). Since Pre-Monsoon results in more light intensity, the temperature was maximum during this season. Similarly, as winter approached, the temperature dropped due to low incident light. The pH of water across all the stations was ascertained to be neutral to moderately alkaline (6.8 - 8.2); which was also reported (28, 29) in their study highlighting other locations of the Indian Sundarbans. A probable reason for this might be the degradation of bicarbonate during the process of photosynthesis that helps in removing carbon dioxide from the system (3) attributable to the decomposition of organic wastes (30). During monsoons, low levels of salinity were observed as an impact of freshwater influx, rainfall and tidal variations (7). The concentration of Total Dissolved Solids (TDS) was in accordance with the levels of salinity, as the leaching of salt poses an impact on the levels of dissolved solids in water (31). Thus, during monsoon, due to tidal encroachment, salinity decreased which in turn lowered the number of dissolved solids present in the waterbody (31). This interrelationship was further statistically justified by the Karl Pearson correlation coefficient, where a positive correlation of salinity with TDS (+0.96) was observed, i.e., as salinity increases, TDS also increases (Fig. 2). The lower values of Dissolved Oxygen (DO) during Pre-Monsoon were the outcome of a surge in temperature during this season, that increased the energy among gas and water molecules further breaking the weak bond between the two. Thus, oxygen dissipates into the atmosphere, lowering its levels in the waterbody (28). Data was again statistically validated by indicating a negative correlation among these two (-0.66) in Pearson's correlation coefficient i.e., as temperature increases, DO decreases. Similar findings were observed (32). Thus, it can be inferred as light intensity increases, DO decreases and temperature increases along with an increase in the salinity and TDS levels. Primary productivity of the Sundarbans ecoregion is related to nutrient concentrations. In our study, nutrient levels in the waterbody have developed a non-uniformed seasonal trend (28), influenced by tidal surges. Levels of total Nitrate accelerated during the winter season as low temperatures, which result in high DO in the waterbody, increase the levels of nitrification (7). Observation confirmed by the distinct negative correlation of Nitrate with Temperature (-0.89) and that of positive correlation with DO (+0.73) (Fig. 2). Total Phosphate and total Silicate levels accelerated during Pre-Monsoon as high-temperature levels during summer increased phosphate decomposition from

the river sediments and increases silicate solubility, owing to aquaculture farm runoff adjacent to the creeks that again fall into the estuary (2). A similar trend was reported by (33, 34). As the phytoplankton backbone is mainly composed of nutrients like nitrate, silicate and phosphate, a surge in their respective levels would pose a direct impact on their community structure as these nutrients get absorbed by them for their growth (7, 35).

Abiotic parameters of water exert influence on the phytoplankton community as during Pre-Monsoon, when the temperature, salinity, nutrient concentration and other factors elevated, abundance of phytoplankton successively escalated (Table 3), observation coincided with that of (7) where they observed maximum phytoplankton abundance in the Sagar Island during the Summer season and lowest during monsoon. Monsoon, on the other hand, when experiencing stressful environmental conditions due to rainfall, most of the phytoplankton members got flushed out into the Bay of Bengal, justifying their low abundance (Fig. 7) during that period (7); observation coinciding with that of (36) and (37). Whereas winter showed an abundance of only one particular group of phytoplankton, i.e., the Dinoflagellates, which act as indicators of eutrophication. In the coastal Yellow Sea, scientists observed that among phytoplankton, diatoms were the most dominant species that varied largely during different seasons and this variation was attributable to the physical parameters, especially those related to water temperature and dissolved inorganic nitrogen during the three seasons with maximum abundance during the summer season (38). Maximum phytoplankton density in the freshwater tank of Talsande, Maharashtra, was observed during summer and minimum during monsoon as rainfall during monsoon lowers their abundance (39). Bio-indication experiments in the Santragachi Lake of West Bengal displayed a low diverse phytoplankton community during monsoon with better water quality than in pre - and post-monsoon seasons (40). This indicated that the density of phytoplankton was higher when temperature and nutrients were high. Phytoplankton communities of the Tibetan plateau had significant remarkable seasonal variations. Their density, biomass and diversity were distinctly lower in the flood (monsoon) than in the non-flood period, which suggested that the flood flow regime plays a key role in balancing and shaping phytoplankton communities (41). All these observations collectively imply the influence of changing environmental parameters across each season on the phytoplankton dynamics.

Diversity measurements act as vital indicators that reflect the health and balance of an ecosystem (28). Throughout our study, the species diversity and richness in the estuary exceeded 1.5, implying a moderately rich phytoplankton assemblage. Though, dilution due to precipitation during monsoon might be the reason for their declined values during this time (2). A high diversity index insinuates a healthy ecosystem, whereas a low value hints towards a degraded one (42). Pielou's evenness index value >1 across all the seasons demonstrated even distribution of phytoplankton. Pearson's chi-square test of association ($p < 0.05$) implied a correlation between the phytoplankton

members with varying seasonal conditions. This variation could be attributed to ecological discrepancy due to climatic and topographical factors (28).

Upon analyzing the data collected across an ecological gradient with Fraserganj having the lowest elevation and Canning having the highest, it came into notice that Gadkhali, Kumirmari and Dobanki exhibited immense similarity in the phytoplankton species composition with a surge in the levels of Dinophyceae members especially during post-monsoon (blooms of *Ceratium*, *Pyrocystis*, *Prorocentrum*) (Table 4) that enact eutrophic conditions in the waterbody (43). Not only that, the concentration of all three heavy metals, as well as the COD levels during winter (47.34 ± 3.09 mg/l), increased way beyond the safety levels declared by WHO (< 10 mg/l), suggesting pollution in the aquatic ecosystem from natural and human-induced factors. Previous studies in the Jambu Island of Sundarbans reported higher COD values (44) and similar BOD values reported in estuarine waters, indicating high organic load that promotes microorganism growth in mangrove patches (45). Intensive industrial activities and various human factors have led to pollution in the Sundarbans delta complex, primarily from domestic sewage and industrial discharge, significantly affecting the local biota. The Haldia port and industrial complex in the lower Gangetic delta have exacerbated this pollution issue. The organic and inorganic waste released by industries and urban areas contains high levels of heavy metals such as zinc, copper, lead, etc. Introducing these heavy metals due to industrialization, unregulated tourism and rapid urban development in the mangrove ecosystem disrupts the biogeochemical cycle, adversely impacting the biotic community. These toxic substances can disturb the ecological balance of an environment and when they enter the food chain, pose serious health hazards, particularly to humans (46). Probable reasons for such pollution could be industrial wastes, sewage and urban effluents, oil spillage, etc. Industrial activities in the Sundarbans have led to untreated effluents being discharged into water bodies, introducing heavy metals and hazardous substances that harm the aquatic ecosystem. Agricultural runoff from pesticides and fertilizers adds to water pollution, as runoff carries these chemicals into the river channels, further contaminating the delta. At the same time, oil spills from shipping pose significant threats to marine life and mangroves (47). The shrimp aquaculture industry also exacerbates the situation, as massive use of chemicals and antibiotics not only endangers mangrove forests but also pollutes surrounding waters (13). Though these causal agents act upon irrespective of any season, then what might have resulted in such escalated levels of water pollution only during one particular season of the year? Further investigation into the same revealed the role of tourism during the winter season.

In the last few years, the tourism pressure has increased immensely in the Indian Sundarbans (48). Favourable tourist season here spans from November to March, i.e., during the post-monsoon season. This poses a threat to the waterbodies as the only mode of transportation in Sundarbans is via waterway. Tourism is a major source of pollution in the Sundarbans, particularly due to improper waste disposal,

noise pollution and the introduction of non-native species that threaten the fragile ecosystem of this mangrove forest. Visitors frequently abandon plastic bottles, food wrappers and other trash, contaminating the waterways and accumulating in the sensitive mangrove environment. Additionally, the rise in mechanized tourist vessels moving through the Sundarbans results in oil spillage, which negatively impacts water quality and disrupts phytoplankton patterns (49). A common issue resulting from this is eutrophication, which is often linked to low oxygen levels and a surge in algae. Global warming and environmental pollution have intensified the occurrence and severity of harmful algal blooms (HABs), affecting both freshwater and marine environments globally. For instance, in North America's Lake Erie, satellite imaging has revealed an expansion in areas impacted by these blooms. The increased frequency of these outbreaks has been associated with human activities like agricultural practices and tourism, which elevate nutrient levels in natural water bodies, leading to imbalances that can promote harmful algal growth (50). Throughout the tourist season in the Sundarbans, the motorized tourist vessels, while sailing through the river channels, cause immense alterations in the substrate structure and species composition along with peat bank erosion; as a repercussion of uncontrolled tourism (10) which also involves littering the waterways with garbage and plastics, discharging bilge and ballast water as well as spilling oil in the waterbody, in turn accelerating the levels of pollution (51). Among our chosen stations, Gadkhali, Kumirmari and Dobanki, being the main tourist attraction spots lying in the heart of the Indian Sundarbans, are thus more exposed to anthropogenic interferences, which directly gets highlighted in the pollution levels of the waterbody during winter. Besides, this season also recorded the sudden blooming of *Ceratium* sp. (15.6×10^3 ind/l), which is a dinoflagellate, only from Gadkhali, Kumirmari and Dobanki, not observed otherwise in ample quantity during the remaining time of the year. Whereas during summer, when favourable environmental conditions prevailed, the phytoplankton assemblage as a whole escalated (52) without focussing on one particular group of indicator species (53). The MDS plot further justified similarity attributes of Gadkhali and Kumirmari with Dobanki by clustering them together based on the abiotic parameters of the waterbody as well as phytoplankton distribution. A probable reason justifying this trend could be the proximity of these spots, as tourists are more likely to divert toward Dobanki after exploring Gadkhali and Kumirmari. This validates pollution pressure due to uncontrolled tourism as Fraserganj and Canning, aloof of such immense tourist load, remained unclustered across the MDS coordinates (Fig. 8), indicating even lesser attributes of similarity. These are the main concerns related to the sustainability of this ecosystem.

Conclusion

The study concludes that, on a seasonal basis, phytoplankton abundance, species richness and diversity were observed during summer. In contrast, winter showed escalation in the population of pollution indicator species, Dinoflagellates due

to uncontrolled tourism that decomposes excess organic wastes on the waterbody. Hence, it can be interpreted from this work that spatiotemporal variation of phytoplankton is dependent on the environmental parameters. Also, improper garbage disposal and vegetation dismantling are likely to increase the harm caused by tourists in this mangrove ecosystem; hence it is extremely crucial to assess the tourist carrying capacity here. Shrimp ponds ooze out pollutants into the river system; so, instead of farming, indigenous fishing can be practiced that will protect the otherwise threatened biodiversity, in turn acting as a source of livelihood for the locals. So, the first step towards conserving this ecosystem would be developing policies for eco-tourism in the Sundarbans, followed by organizing awareness programs for the tour operators, tourists as well as locals. Alongside this, enforcing strict environmental regulations is vital for reducing industrial pollution. Industries should be required to treat their wastewater before it is released, with ongoing monitoring to ensure they comply with these standards. Promoting organic farming techniques and decreasing reliance on chemical fertilizers and pesticides can substantially decrease agricultural runoff. It is essential to create and implement effective waste management strategies by the government authorities, particularly for plastic waste. They can also oversee stormwater runoff to limit excessive water entering the river. Initiatives that encourage recycling, proper disposal methods and community education are important for alleviating the pollution issue. Setting up rapid response teams and developing contingency plans for oil spills can help lessen their effects. Additionally, training local communities and authorities to manage such emergencies, along with actively participating in mangrove replanting initiatives to safeguard the delicate ecosystem, is crucial.

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Authors' contributions

DR, the corresponding author, designed the work outline, RM executed it and SS supervised it. All authors read and approved the final manuscript.

Compliance with ethical standards

Conflict of interest: The corresponding author is a reviewer of Plant Science Today, assigned to review manuscripts belonging to separate disciplines.

Ethical issues: None

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