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Seasonal Variation and Diversity of *Tripos* (Dinoflagellata) in Relation to Environmental Drivers in the Hooghly River Estuary, Bay of Bengal

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***Abstract*:** Planktonic dinoflagellate *Tripos* spp. were sampled and documented from three selected sites within the Hooghly Estuary, Bay of Bengal. The study focused on their diversity, abundance patterns, and the environmental factors regulating them. 14 species of *Tripos* were identified, with *Tripos furca* and *Tripos fusus* being the dominant. Seasonal patterns indicated that post-monsoon (POM) and pre-monsoon (PRM) exhibited the highest diversity, characterized by low dominance (D = 0.18 to 0.23), high Simpson diversity index (1–D = 0.78 to 0.82), and elevated Shannon diversity index values (H' = 1.75 to 1.89). In contrast, monsoon (MON) displayed the highest evenness (E = 0.90 to 0.94) but reduced diversity. *Tripos fusus* showed maximum abundance (highest cell density 1.92×10^3 Cells/L) during the post-monsoon (POM) season, when nutrient levels and salinity were elevated and the water temperature was relatively lower. Multivariate Non-metric Multidimensional Scaling (NMDS) ordination with environmental vectors displayed a clear separation of seasonal groups, primarily influenced by temperature, salinity, and nutrient gradients. Similarity Percentage (SIMPER) analysis identified *Tripos fusus*, *Tripos furca*, and *Tripos lineatus* are the major contributors to seasonal dissimilarity in the assemblages. Individual rarefaction analyses applied to the abundance further revealed higher species accumulation during late POM and early PRM months. Overall monsoon-influenced hydrological changes are the dominant force regulating *Tripos* diversity and community composition in the Hooghly Estuary.

Keywords: Abundance, Diversity, Salinity, Nutrients, NMDS, post-monsoon

1. INTRODUCTION

Dinoflagellates are a diverse group of single-celled myzozoan protists and a major component of the marine phytoplankton community. They play essential roles in primary production and nutrient cycling, acting as a link between microbial food webs and higher trophic levels (Taylor et al., 2008). Genus *Tripos* (formerly known as *Ceratium*) under family Ceratiacae, a group of cosmopolitan thecate dinoflagellate is of particular ecological interest due to its distribution, high morphological variability, and marked seasonal abundance in estuarine and coastal waters (Gómez, 2012). Owing to their distinct cell structure and morphological features, these dinoflagellates are easily distinguished from others. Different *Tripos* spp. possess variable-sized short or long apical and antapical horns. *Tripos* spp. are highly sensitive to environmental change, and their occurrence and diversity are commonly linked to changes in hydrographic conditions, nutrient inputs, and anthropogenic pressures (Shin et al., 2016; Hallegraeff, 2020).

Estuarine areas serve as high productive transition zones that connect freshwater with marine ecosystems. The Hooghly River estuary is one of the important estuarine systems on the northeastern shore of the Bay of Bengal. This westernmost part of the Gangetic river delta has a lot of mixing of fresh water and plenty of human activity, like fishing boats, trawlers, and the disposal of urban wastewater (Chatterjee et al., 2013; Henderson et al., 2021). Salinity gradients, tidal flows, and nutrient fluxes create a unique biological niche that harbours a wide range of phytoplankton communities in this area. Dinoflagellates are ecologically significant group of phytoplankton, though they receive less attention compared to diatoms. Specifically, thorough studies on *Tripos* distribution, seasonal abundance, and diversity in response to environmental factors are lacking (Naik et al., 2011; Rath et al., 2021). The present study tries to address this gap by integrating seasonal field observations with multivariate statistical analyses to evaluate species-environment relationships in the Hooghly estuary.

Understanding the ecology of *Tripos* in the Hooghly estuary can provide the knowledge of estuarine ecosystem productivity and functioning. Their seasonal abundance

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affects primary production and energy transfer, which affects higher trophic levels such as zooplankton and fish population. The current investigation examines seasonal variation, diversity, and abundance of *Tripos* species in the Hooghly estuary in response to environmental factors that could enhance the understanding of dinoflagellate ecology.

2. MATERIAL AND METHODS

2.1. Study Area

The investigation was conducted from October 2021 to September 2023 in the lower estuarine stretch of the Hooghly River near Namkhana, West Bengal. Three seasonal observations were considered: the pre-monsoon season (PRM: February–May), the monsoon (MON: June to September) and the post-monsoon season (POM: October–January). Sampling was carried out at three locations: Station 1 (21.76056°N, 88.23628°E) adjacent to Namkhana Bridge, Station 2 (21.754776°N, 88.267445°E) near Madanganj, and Station 3 (21.727693°N, 88.266245°E) close to Dwariknagar Ferry Ghat. The study sites receive the impact of freshwater inflow coming from the Hooghly River and saltwater entering from the Bay of Bengal (Fig. 1).

2.2. Sampling Methods

We followed standard procedures to collect water samples. Surface water temperature, pH, salinity, total dissolved solids (TDS), and electrical conductivity (EC) were recorded using a portable multi-parameter probe of HANNA. Dissolved oxygen (DO) and nutrient

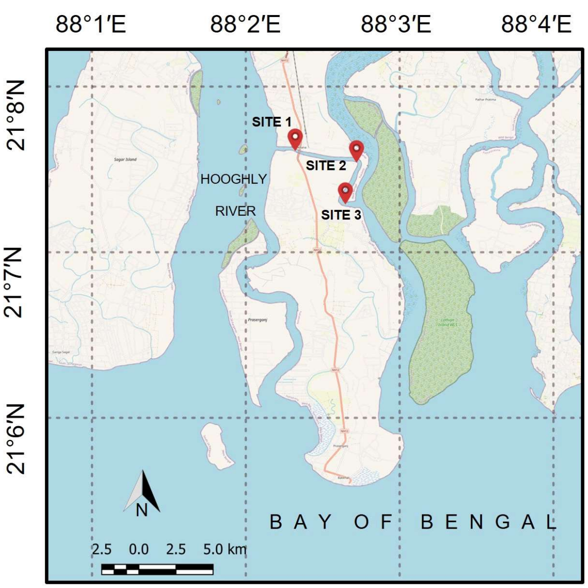

Figure 1. Map of the study area

concentrations were estimated by laboratory analysis using standard methods. Nitrate, phosphate, and silicate content of water were determined spectrophotometrically after filtration of samples, following the methods of Grasshoff et al. (2009). Dinoflagellate samples were collected through sieving 50 liters of water through bolting silk and converted to plankton concentrate, then preserved with 10% Lugol's iodine (Williams et al., 2016). Dinoflagellate cells were counted using a Sedgewick Rafter chamber under a Dewinter Educator Plus and Leica DMi8 microscope. Species identification and nomenclature were done using available literature (Hasle et al., 1996; Gómez, 2021).

2.3. Statistical Analysis

The Shapiro–Wilk test was applied to test for normality in the data of *Tripos* species. If data did not follow a normal distribution, the non-parametric Kruskal-Wallis test was employed. Post-hoc pairwise comparisons with Bonferroni-adjusted *p*-values were done when the Kruskal-Wallis test showed significance. Dominance (D), Simpson's index (1−D), Shannon-Wiener diversity index (H'), Brillouin index (B), Pielou's Equitability index (J), and Margalef richness, Menhinick index were employed to quantify diversity and species richness. Species rarefaction curve was constructed using pooled seasonal data to visualize species richness across different months. To visualize the seasonal patterns of species assemblage, species counts were converted to relative abundance (%). Then Similarity Percentage (SIMPER) analysis (based on Bray–Curtis dissimilarity) was conducted to see which *Tripos* species contributed the most to the differences among seasons, summarizing their average dissimilarity, individual contributions, and cumulative influence. For a broader view of community structure, multivariate statistics Non-metric Multidimensional Scaling (NMDS) with environmental variables as vectors were performed using log (x+1) transformed abundance data to show the alignment with the species and seasonal patterns. Different statistical analyses were carried out in MINITAB, PAST (version 5.0), and Microsoft Excel (2010).

3. RESULTS AND DISCUSSION

3.1. Abundance Structure and Variation of *Tripos*

Test statistics for the Shapiro-Wilk test (W) involving monthly mean abundance of all *Tripos* spp. ranged between 0.642 and 0.771 (N = 14, $p < 0.001$) in different seasons, revealing their non-parametric nature. The Kruskal–Wallis test (tie-corrected) revealed significant seasonal variation in the abundance of *Tripos* ($Hc = 34.01$, $p < 0.0001$), indicating marked variability in community structure within the estuarine system. Dunn's post hoc analysis with Bonferroni correction indicated significant differences ($p < 0.05$) between MON samples and both PRM and POM, demonstrating significant seasonal variation in dinoflagellate abundance across the sites. The study documented 14 species of *Tripos* belonging to several infrageneric sections, like *eugrammus*, *fusiformia*, and *macroceros* (Table 1). *Tripos fusus* was the most observed species with maximum cell density (1932 cells/L) in POM, followed by *Tripos furca* (963 cells/L) and *Tripos trichoceros* (733 cells/L) all exhibiting peak abundance during POM. These three taxa were the most abundant among all and contributed the majority of the *Tripos* assemblage (Baek et al., 2008). Observed seasonal patterns of relative abundance of all *Tripos* and their SIMPER outputs given in Table 1. *T. fusus* exhibited the 36.77% of the average Bray-Curtis dissimilarity, with notably greater abundances during POM and PRM. *T. furca* and *T. trichoceros* were the next two important contributors, each accounting for more than 16% of the dissimilarity, suggesting their rapid shifts in relative abundance in response to fluctuations in salinity and nutrient concentration across seasons. A limited number of species predominated the inter-seasonal variations. *T. mulleri* and *T. lineatus*, added moderate contributions with their fluctuating relative abundances, while all other species have contributed less than 5%. *T. fusus*, *T. furca*, and *T. trichoceros* (cumulatively contributing 69.5%) suggest that these taxa serve as dominant class in the estuary. The impact of rare species like *T. inflatus*, *T. azoricum*, *T. gibberum*, and *T. minutus* (< 0.5%) underscores their limited contribution to overall community turnover (Haque et al., 2021). The combined patterns of abundance, relative contribution, and SIMPER results indicate significant population shifts, especially during POM, highlighting the suitable growth conditions for dominant *Tripos* spp. These seasonal changes are ecologically important and can have strong effect on the estuarine productivity, grazing activity, and the overall trophic structure.

3.2. Variation in Environmental Parameters

Surface water physicochemical properties showed strong seasonal variation, typical of tropical estuarine systems driven by the monsoon (Fig. 2). Water temperature ranging from a minimum of 18.7°C (POM) to a maximum of 33.6°C (MON). Dissolved oxygen (DO) ranging from 4.1 mg/L during August (MON) to a maximum of 7.46 mg/L recorded in January (POM) while the average value 5.0–6.5 mg/L, with higher concentrations consistently observed during the colder PRM and early POM months. pH exhibited moderate spatial-seasonal variation, generally oscillating between 7.0 and 7.8. TDS, EC, and salinity values followed a more or less symmetrical increase and decrease pattern. The nutrient concentrations exhibited marked seasonal variability, with the highest values recorded during the

Table 1. Relative abundance of all documented Tripos along with their SIMPER contribution

Species	Relative abundance (%)			SIMPER contribution		
	POM	PRM	MON	Average dissimilarity	Contribution (%)	Cumulative (%)
Tripos fusus	34.18	34.84	21.19	19.8	36.77	36.77
Tripos furca	20.77	16.52	28.07	8.812	16.37	53.14
Tripos trichoceros	15.4	20.18	20.94	8.81	16.36	69.51
Tripos mulleri	9.53	8.62	7.98	5.205	9.668	79.17
Tripos lineatus	10.13	8.25	20.22	4.22	7.84	87.01
Tripos brevis	3.19	3.09	1.6	1.936	3.596	90.61
Tripos longipes	3.78	2.23	0	1.915	3.557	94.17
Tripos macroceros	0.54	4.11	0	1.782	3.309	97.48
Tripos falcatus	0.57	0.42	0	0.3066	0.5695	98.05
Tripos declinatus	0.44	0.62	0	0.2952	0.5484	98.59
Tripos inflatus	0.56	0.27	0	0.248	0.4607	99.05
Tripos azoricum	0.29	0.41	0	0.1935	0.3595	99.41
Tripos gibberum	0.38	0.18	0	0.1721	0.3197	99.73
Tripos minutus	0.25	0.24	0	0.1436	0.2667	100

monsoon (MON) months. Nitrate from 26 to 47 µM, and silicate levels between 53 and 114 µM. Phosphate exhibited a similar trend, reaching its highest during MON (3.6–3.8 µM) and declining gradually in POM and becoming the lowest during PRM (1.3 µM). Temperature is the key component affecting numerous aquatic organisms, especially the marine phytoplankton group (Hays et al., 2005). A large number of *Tripos* species were observed from December to March, when temperatures ranged between 20 and 26°C. This seasonal enhancement is attributable to reduced temperature and enhanced mixing, while the marked decline during the MON aligns with elevated temperature, increased organic loading, and intensified microbial decomposition, lowered salinity, and enhanced freshwater inflow, which together favour oxygen enrichment and elevated phytoplankton productivity (Kibler et al., 2012). The similar pattern of pH and DO suggests a common regulatory control by temperature. This kind of negative association with pH and DO is characteristic of productive estuarine waters where photosynthesis decreases carbonate chemistry. Fluctuations in EC, TDS, and salinity levels are affected by the temperature, quantity of freshwater inputs, tidal power, and rapid rate of evaporation. In PRM, low freshwater influx, rising temperature, and a higher rate of evaporation caused a lowering of salinity level that was further dropped in MON due to the large flow of freshwater and gradually increased in late POM (Manna et al., 2010). The elevated concentrations of nitrate and phosphate during MON are ecologically significant for dinoflagellates, many of which possess efficient nitrate uptake systems (Abbasi and Ki, 2022). The nutrient-rich, stratified MON environment therefore provides favourable conditions for opportunistic growth, whereas the more oligotrophic PRM restricts productivity and may influence the seasonal succession of *Tripos* species (Baek et al., 2009).

3.3. Dominance Patterns and Species Richness

Based on the abundance pattern of each *Tripos*, an alluvial diagram was constructed (Fig. 3), which revealed different species classes in the assemblage. *T. fusus* and *T. furca* were dominant with their wider bandwidth in POM and PRM. Contributions made by *T. lineatus*, *T. trichoceros*, and *T. mulleri* during POM and PRM, primarily in the frequent classes and occasionally dominant classes. This indicates their moderate abundance but significant ecological significance. On the other hand, species such as *T. minutus*, *T. azoricum*, *T. gibberum*, and *T. inflatus* are always few and compose the rare category. During PRM and POM, their bands become smaller and decline dramatically with a total absence in MON. The figure illustrates that POM and PRM are distinguished by the expansion of species while MON restricts the community to a state of low abundance, with just a few taxa that are able to withstand harsh conditions (Naik et al., 2020). The seasonal succession demonstrates the significant impact of hydrological

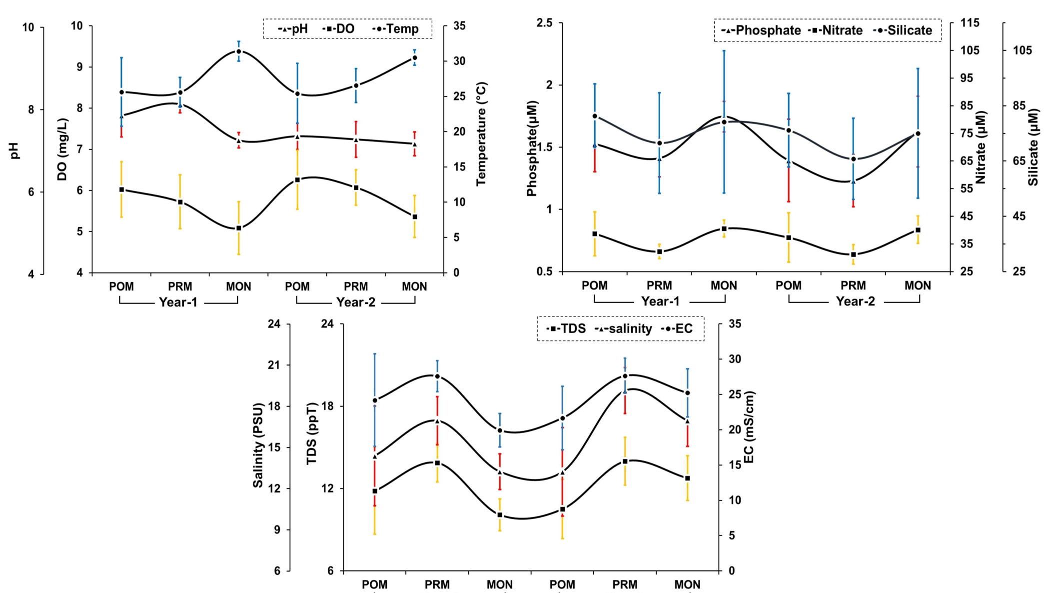

Figure 2. Fluctuation of physicochemical parameters of water during different season

fluctuations on *Tripos* dynamics in the estuary, with POM conditions beneficial to proliferation and growth before the onset of monsoon dilution.

Individual rarefaction curves (Fig. 4) constructed from monthly mean abundance show distinct variation in species accumulation across the sampling period. The curves for January, February, and March demonstrate higher asymptotes, suggesting an increased estimated richness during the late POM and early PRM periods. In contrast, the curves for June–September level off at much lower values, reflecting very few species during peak MON. Intermediate patterns in October–November suggest transitional richness as favourable environmental conditions shift during POM phases. The monthly differences in rarefaction patterns reflect the strong seasonal influence on *Tripos* community structure. Higher richness in POM and early PRM likely corresponds to stable hydrographic conditions, enhanced salinity, nutrient availability, and reduced turbidity, which collectively favour greater species coexistence. The monsoon-related drops in richness (June to September) are due to the inflow of freshwater, the lowering of salinity, and the high turbidity load, all of which are known to reduce dinoflagellate diversity. The gradual recovery observed during the post-monsoon months (October, November) indicates reduced turbidity and re-stabilization of nutrient loads as hydrological conditions become more stable (Patil & Anil, 2011; Bharathi & Sarma, 2019).

3.4. Seasonal Diversity Indices

Changes in diversity and richness indices also showed significant seasonal shifts (Fig. 5). Simpson's diversity

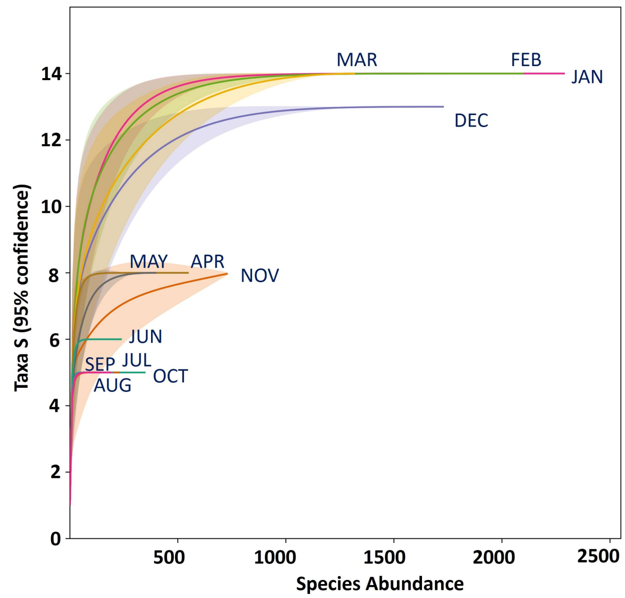

Figure 4. Individual rarefaction curve illustrating temporal variation in species accumulation pattern

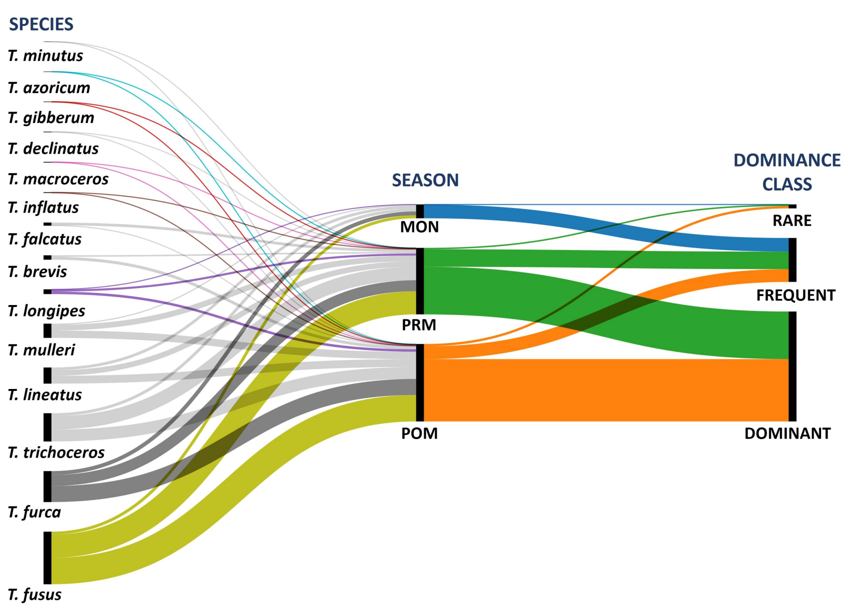

Figure 3. Alluvial diagram of all Tripos spp. redistribution by absolute abundance, band width proportionate to species contribution to the seasonal assemblage (Identified dominance classes as Dominant > 1000, Frequent 100-1000, and Rare < 100)

(1–D) stayed more or less same all year, with values for POM between 0.74 and 0.80, for PRM between 0.72 and 0.82, and for MON between 0.76 and 0.80. Shannon's index (H′) showed clear separation, with values of 1.58–1.89 in POM and PRM and 1.51–1.69 in MON. The Brillouin index (B) showed a clear pattern, with the highest values in the POM and PRM and the lowest values in the MON (1.39–1.55). Interestingly, Pielou's evenness (E) and Equitability index (J) was always higher in MON, ranging respectively from 0.47 to 0.97 and 0.71 to 0.97. The highest values of Margalef richness were in PRM (1.02–1.82), and the lower was in MON (0.71–0.92). Menhinick's index showed similar patterns, with higher values in POM and PRM (0.26–0.42) and lower values in MON (0.30–0.39). The highest value for dominance (D) was in PRM (up to 0.2831), and the lowest value was in MON (about 0.198). The higher Shannon, Simpson, and Brillouin indices observed during POM and PRM suggest more diverse and intricate assemblages characterized by low dominance, suggesting suitable hydrographic conditions that facilitate the coexistence of various taxa. The decrease in diversity and species richness observed during MON can be connected with significant physical stress associated with rainfall, freshwater influx, lowering salinity, turbidity, and hydrodynamic disturbance (Sahu *et al.*, 2014). Though MON displayed very few taxa, but showed the highest evenness, suggesting a uniform distribution along with low abundance assemblage. These marked increase in diversity from late POM (December-January) to its peak in early PRM (February-March) reflects the transition to more favourable water conditions that support the growth and proliferation of *Tripos* populations. The seasonal rise in abundance and diversity is consistent with previous studies that relates phytoplankton growth with positive changes in salinity, stability, and nutrient availability (Sathish et al., 2022; Pradhan et al., 2023; Huang et al., 2024). The investigation reveals that the increase in *Tripos* abundance and diversity during the late post-monsoonal months reflects the specific adaptive response of these dinoflagellate cells to varying hydrological conditions, demonstrating their unique ecology in the relatively unexplored Hooghly estuarine system.

3.5. Species Ordination and Correlation

The NMDS ordination showed that the *Tripos* assemblages were clearly separated into three distinct seasonal clusters along the two NMDS axes (Fig. 7). The distribution of samples reveals significant changes in community structure over time due to variations in nutrient levels and water conditions. Environmental vectors applied to the ordination space elucidated the gradients that support the observed patterns. Axis 1 exhibited strong positive correlations with DO (0.84), EC (0.67), TDS (0.65), salinity (0.55), and pH (0.55), and strong negative correlations with temperature (–0.94), nitrate (–0.78), and phosphate (–0.73).

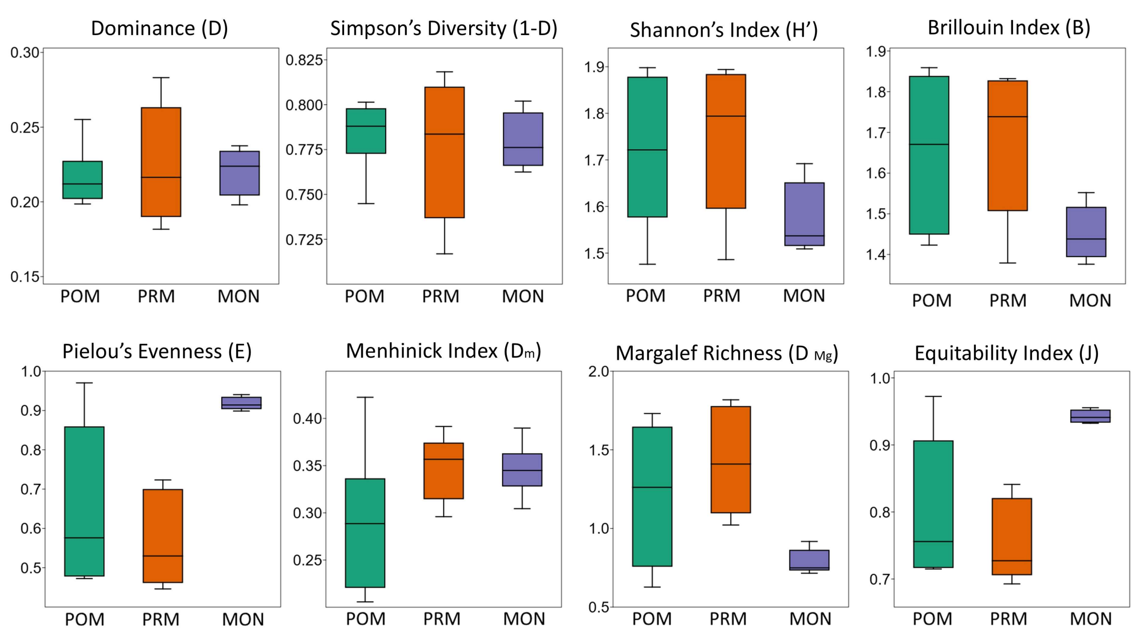

Figure 5. Seasonal variation in diversity and richness indices

This axis shows a gradient of salinity and dissolved oxygen, highlighting the differences between POM saline, oxygenated water, and the nutrient-rich, lower-salinity conditions observed during the MON. Axis 2 exhibited a positive correlation with silicate (0.49) and nitrate (0.47), while showing a negative correlation with salinity (–0.51), EC (–0.43), and TDS (–0.44) suggesting the presence of a secondary nutrient-ion concentration gradient (Kim et al., 2023). The rank of the different environmental vectors in the NMDS ordination along with axis correlations and significance was given in Table 2. The POM samples clustered toward the positive side of Axis 1 and the negative side of Axis 2, corresponding closely with vectors for salinity, DO, EC, and TDS. This indicates that POM assemblages are primarily influenced by high salinity, increased ionic concentration, and enhanced oxygen availability. Species such as *T. fusus*, *T. furca*, *T. brevis*, *T. mulleri*, and *T. trichoceros* were located near these vectors,

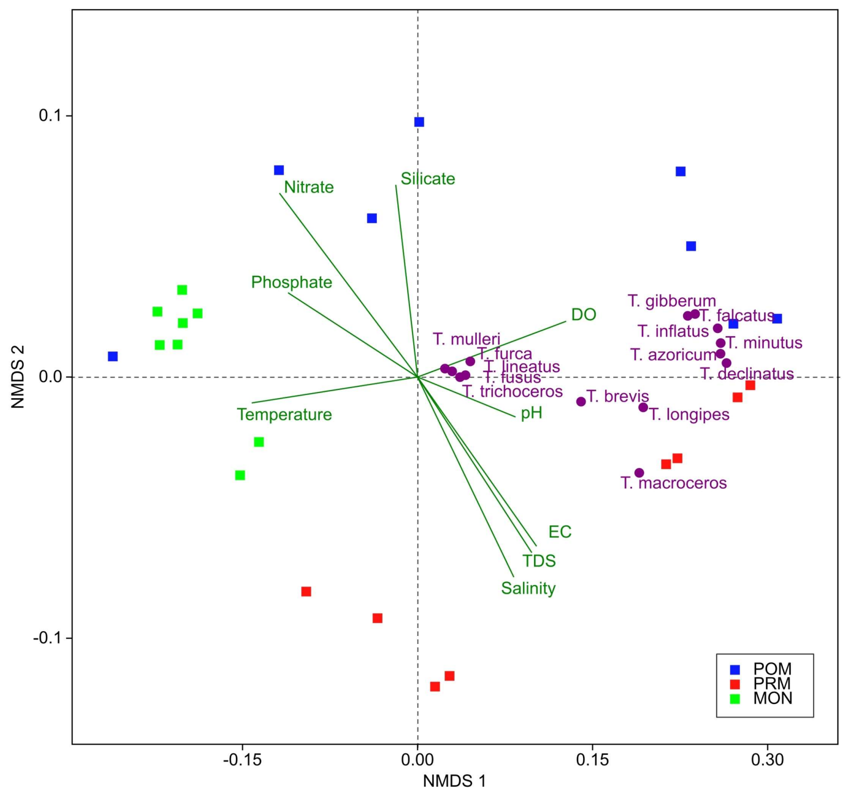

Figure 6. NMDS plot with environmental vectors showing abundance of all *Tripos* spp. in different seasons

Table 2. Environmental vectors fitted to the NMDS ordination showing axis correlations, r², significance

Rank	Environmental variable	NMDS1 (r)	NMDS2 (r)	r²	*p*-value
1	Temperature	−0.93919	−0.06548	0.88637	0.001
2	Nitrate	−0.78213	0.46531	0.82824	0.001
3	Dissolved oxygen (DO)	0.8415	0.1411	0.72803	0.001
4	Electrical Conductivity (EC)	0.67391	−0.42809	0.63742	0.001
5	Total Dissolved solids (TDS)	0.64681	−0.44438	0.61584	0.001
6	Phosphate	−0.73361	0.21327	0.58367	0.001
7	Salinity	0.54506	−0.50626	0.55339	0.001
8	pH	0.55485	−0.10104	0.31807	0.001
9	Silicate	−0.12352	0.48635	0.25179	0.001

indicating a strong preference for POM hydrographic conditions (Bharathi et al., 2022).

POM samples showed a transition from the MON conditions to PRM, and characterized by increasing salinity and intermediate nutrient concentrations. MON samples were grouped together on the negative side of Axis 1 and the positive side of Axis 2, right next to the nitrate and silicate vectors. This indicates that MON assemblages are shaped primarily by nutrient enrichment, reduced salinity, and cooler temperatures characteristic of heavy freshwater inflow. The NMDS data collectively illustrate that the composition of the *Tripos* community is significantly influenced by seasonal fluctuations in hydrographic parameters, including salinity, temperature, dissolved oxygen, and nutrient concentrations (Rothenberger et al., 2014). The recognized seasonal patterns validate the changes in abundance data and the SIMPER analysis, emphasizing the unique ecology of each Tripos throughout the annual cycle (Sidik et al., 2008). By integrating multivariate analyses, the study identifies temperature, salinity, and nutrient gradients as key drivers of species restructuring. These findings are especially relevant under current conditions of increasing pollution and climate-driven changes in freshwater flow and temperature, providing an updated baseline for future ecological assessments.

As phytoplankton form the primary trophic base of estuarine ecosystems and play a fundamental role in supporting zooplankton populations and higher trophic levels. Seasonal variations in *Tripos* abundance may influence food availability for grazers and contribute to dissolved oxygen dynamics through photosynthetic activity. These shifts in abundance can also affect zooplankton and juvenile fish through changes in natural food supply, which in turn supports open estuarine capture fisheries. While such trophic assessments are more straightforward in pond aquaculture, phytoplankton-based insights from this study can still help in understanding natural feeding conditions and may provide useful guidance for fisheries management in dynamic estuarine systems. Although zooplankton and fish productivity were not assessed in the present study, the documented phytoplankton patterns provide important insight into the potential trophic functioning of the open dynamic estuarine ecosystem regulated by tidal exchange, freshwater discharge, and hydrodynamic variability, making direct extrapolation more complex

Present study focuses on the seasonal dynamics of *Tripos,* direct evaluation of higher trophic linkages and fisheries implications requires further dedicated studies incorporating multi-trophic datasets and long-term monitoring. Future research integrating long-term datasets with AI-based predictive modelling and remote sensing approaches could forecast bloom dynamics and improve understanding of estuarine phytoplankton responses under changing climatic and environmental conditions.

4. CONCLUSION

Tripos spp. exhibit clear seasonal pattern in abundance and community structure as due to changing hydrographic conditions. The post-monsoon period supports higher species richness, diversity, and abundance, indicating favourable physicochemical conditions for the growth of these dinoflagellates. Bloom-forming taxa such as *T. furca* and *T. fusus* contributed substantially to seasonal dissimilarity, highlighting their ecological significance in nutrient-sensitive estuarine environments facing anthropogenic pressure. Since phytoplankton form the base of the aquatic food web, understanding the ecology of *Tripos* enhance the understanding of estuarine productivity, trophic functioning and the potential support to fish populations. The study also highlights the need for long-term ecological monitoring that can enhance our knowledge of estuarine phytoplankton dynamics in the tropical monsoon-influenced Hooghly estuary under increasing stress due to climatic change.

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Authors'Contributions

GB: Investigation; Data curation; Formal analysis; Validation; Visualization; Writing draft, SKS: Conceptualization; Methodology; Review & editing; Supervision

Conflict of Interest

The authors declare that they have no known competing financial or personal relationships that could have influenced the work reported in this paper.

Data Availability

Data will be made available by the corresponding author upon reasonable request.

Declaration about use of AI Tools

The authors declare that no generative AI or AI-assisted tools were employed in the writing of the text or in the creation or alteration of any figures, photos, or artwork.

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