



Structural composition of phytoplankton communities in the tropical Thamirabarani estuary, Southeast coast of India

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ABSTRACT

This study investigated the seasonal effects on phytoplankton populations in the Thamirabarani estuary on India's southeast coast. 49 species from 31 genera, 24 families, and three phyla (bacillariophyte, miozoa, and cyanobacteria) were identified. The number of species, density, and Margalef richness (d') were high in the pre-monsoon season (43 ± 1 cells/L, 8463 ± 72 ind./m³, 4.64 ± 0.12 ind./m³) and low in the post-monsoon season (35 ± 1 cells/L, 6448 ± 262 ind./m³, 3.84 ± 0.10 ind./m³) with significant serial changes ($\chi=0.43$) in species composition between the seasons. However, these parameters did not show any significant variations in station-wise. The phytoplankton relationship with environmental factors (especially temperature, pH, phosphate, ammonia, salinity, silicate, and chloride) indicated the influence of the nutrient dynamics on plankton communities due to anthropogenic discharges and vegetations (agricultural land and mangroves) in the study area. Hence, suitable improvement strategies have been implemented in this area, and its computation demonstrates the link between phytoplankton abundance and the most effective environmental parameters.

KEYWORDS: Phytoplankton, Thamirabarani estuary, Nutrients, Community structure, Diversity, Environmental parameters

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INTRODUCTION

The dynamics of phytoplankton and their recurrence throughout time are driven by a complex combination of the aquatic ecosystem's physical, chemical, and biological processes. Since its inception, the idea of phytoplankton functional groups has been widely used in ecological studies of many marine environments around the world (Latinopoulos *et al.*, 2020; Nagy-László *et al.*, 2020). Estuary ecosystems are hence prime locations for phytoplankton development (Niu *et al.*, 2015). Phytoplankton is an important food chain link and a major primary producer in aquatic ecosystems (Zhang *et al.*, 2016; Srichandan *et al.*, 2019). The productivity of higher trophic-level species is largely regulated by the biomass and productivity of phytoplankton by a range of parameters. National Aeronautic and Space Administration reports that 50% to 90% of the oxygen in the atmosphere is produced by phytoplankton, depending on the season (Thangaradjou *et al.*, 2013). Increasing nutrient levels, together with other hydrological restrictions, control phytoplankton dynamics in aquatic environments (Srichandan

et al., 2019; Jia *et al.*, 2019). Phytoplankton biomass, or chlorophyll-a, is employed as an excellent indicator of water quality and eutrophication since it offers good insights into that specific location (McQuatters-Gollop *et al.*, 2009; Ninčević-Gladan *et al.*, 2015). Aquatic ecosystem productivity and health are indicators of the plankton population (Prabhakar *et al.*, 2011). Further, the physicochemical and accessibility factors significantly impact phytoplankton species composition, density, and diversity (Chattopadhyay *et al.*, 2003).

Phytoplankton biomass in estuaries varied greatly depending on freshwater intake, tidal movement and water turbidity. Researchers have traditionally focused primarily on the dynamic interaction between phytoplankton and nutrients in order to explain experimental ecology (Chattopadhyay *et al.*, 2003). Anthropogenic activities have grown recently, increasing the concentration of nutrients and resulting in high productivity in coastal environments (Rakesh *et al.*, 2013), and increasing eutrophication to affecting biological processes due to increased levels of nutrients in estuarine environment (Jia *et al.*, 2019). Although the main source of coastal eutrophication

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is agricultural fertilizers, the health of the ecosystem is also impacted by livestock, wastewater, urban runoff, river flow, and aquaculture. Thus, both natural and human processes pose a hazard to estuarine ecosystems (Jin *et al.*, 2016; Yi *et al.*, 2018). Significant variations in physicochemical parameters show varying effects on the population and distribution of several phytoplankton species, which in turn reveal the water's quality (Liu *et al.*, 2004; Shekhar *et al.*, 2008). Numerous hydrochemical and physical parameters, including temperature, salinity, pH, nitrate, nitrite, ammonia, and silicate, have a significant impact on the spatial and temporal fluctuations in phytoplankton dispersion. The range and diversity of species in the estuary environment are changed by the impact of these variables on the phytoplankton community (Durate *et al.*, 2006; Madhu *et al.*, 2007).

The Thamirabarani River (Porunai), the only perennial river in South India, rises from the Agastyar Koodam peak of the Pothigai hills of the Western Ghats. It flows through Tamil Nadu's Tirunelveli and Tuticorin districts, containing nutrients that are mixed from estuaries and discharged into the Bay of Bengal in the Gulf of Mannar region. The Gulf of Mannar, located between the west coast of Sri Lanka and the southeast coast of India, is a unique marine environment rich in biodiversity, home to over 3600 species of aquatic plants and animals, and is appropriately known as a biologist's paradise (Pitchaikani & Lipton, 2016). Because it combines areas of intensive socio-economic activity and developing areas, this significantly bio-diversified environment is economically significant. During the northeast monsoon (NEM) season, a large amount of nutrients is discharged into this region via freshwater input from rivers such as Malvathu in Sri Lanka and Thamirabarani in South India (Kumaraguru *et al.*, 2006). It is highlighted that the average rainfall of this area is 75 cm (Suresh *et al.*, 2023) and the highest rainfall in the year 2023 was 93 cm.

Even though, several studies conducted on the distribution and density of phytoplankton in Indian estuaries (Perumal *et al.*, 2009; Singh & Chaturvedi, 2010; Sarangi & Devi, 2017; Roshith *et al.*, 2018; Pramanik *et al.*, 2020), the study of phytoplankton community structure in response to environmental factors in the Thamirabarani estuary is still rare. Thus, the purpose of this study is to learn more about the structure and variety of the plankton community and its function in the Thamirabarani Estuary with the objectives (i) to study the community structure and density of phytoplankton (ii) to analyse the key determinant for the seasonal dynamics of phytoplankton density (iii) distribution and investigate the relationship between environmental parameters and phytoplankton community.

MATERIALS AND METHODS

Study Area

The study area covered three different zones at Thamirabarani Estuary (Palayakayal) including the Mouth Zone (st. 1 - 8°39'49.34" N; 78°07'45.83" E), Estuary zone (st. 2 - 8°39'46.96 N; 78°06'11.05" E), and Creek Zone (st. 3 - 8°38'39.77 N;

78°05'15.94" E). The study was conducted during the pre-monsoon, monsoon and post-monsoon (July 2019 - May 2020) (Figure 1). This estuary is surrounded by sparse mangrove vegetation and receives freshwater during the Southwest and Northeast monsoons, as well as seawater from the Bay of Bengal. High amounts of nutrients are discharged by riverine sources (Thamirabarani, Gundar, Vembar, and Vaipar) during the Northeast monsoon season (Kumaraguru *et al.*, 2006). Mangrove ecosystems and mud flats are found in this estuary's coastal surroundings.

Environmental parameters

The environmental parameters were measured in all the seasons from three distinct zones. Water samples were immediately transported to the laboratory for analyses of physicochemical, nutrient, and biological characteristics after being stored in an ice box. A Water temperature (°C) was measured with great accuracy by a laboratory thermometer. Salinity (ppt) was analysed by Salino-refractometer (PSU), pH of the water was measured with a calibrated pH pen (pH Scan1 Tester-Eutech Instruments, Singapore) and dissolved oxygen (mg/L) content was calculated using the Winkler's method (Winkler, 1888). The dark and light bottle technique was used to estimate primary production. The chemical parameters of water samples were used to examine water nutrients such as nitrite (NO₂), nitrate (NO₃), ammonia (NH₄), inorganic phosphate (PO₄), chloride (Cl), reactive silicate (SiO₄) following the standard methodology described (Strickland & Parsons, 1972).

Sampling and Analyses of Plankton

To assess the phytoplankton composition, in each sampling point, a bucket sampler was used to manually collect 3 L of superficial (0–30 cm) water samples from the estuary. Following the collection of each sample, water samples were immediately separated into three subsamples (each at 500 mL), water sample was fixed in a 0.5% Lugols iodine solution (Thronsdon, 1978). Samples of phytoplankton were given one full day to settle before were focused to 10 milliliters using a greater capacity water decanter. Using a BX51 OLYMPUS microscope (Olympus, Japan), the phytoplankton composition of this concentrated sample (100 mL) was examined. The quantity of phytoplankton was assessed at a 400x magnification. The phytoplankton was analyzed using a Sedzwick rafter counting chamber, and the abundance was expressed in cells/L (Aktan *et al.*, 2005). To find out the different species, the traditional identification keys (Hasle & Syversten, 1997) were also employed. Phytoplankton abundance is assessed in cells per milliliter (cells/L) (Baliarsingh *et al.*, 2016) and identified using standard manual methods (Smith & Whitley, 1977; Santhanam *et al.*, 1987; Perumal *et al.*, 1998; Al-Kandari *et al.*, 2009). Cells of recognized species in a 1ml sample were counted using a chamber in triplicate, and the density was calculated using the formula $N = n \times v/V$ (Asha *et al.*, 2018; Varghese *et al.*, 2022). Where, N is the mean cell number in 1ml of sample, v is the volume of concentrate (mL), and V is the volume of seawater filtered (L). The relative density of phytoplankton groups was then estimated as a percentage



Figure 1: Map showing the sampling stations in the Thamirabarani estuary

of the total number of plankton cells using the overall density and the density of each group.

Statistical Analysis

The biodiversity indices for the phytoplankton (Margalef's species richness-d, Shannon Wiener diversity- H' \log^2 , and Pielou's evenness- J') were measured using PRIMER 6.0. Multivariate Analyses were done on square-root transformed density data to detect possible assemblage differences between the stations and seasons. PERMANOVA was done to test seasonal (pre-monsoon, monsoon, and post-monsoon) and stations (st. 1, st. 2 and st. 3) differences in density. The distribution pattern in relation to factors of season and station (fixed ones) was tested using 999 permutations. Bray-Curtis similarity was used for cluster analyses, and the serial change in the distribution of phytoplankton was tested in relation to seasons. SIMPER was used for finding species characterizing the composition in all the seasons and stations. Initially, the average dissimilarity between all pairs of inter-group samples was calculated. The environmental parameters were log-transformed and normalized before calculating the resemblance using Euclidean distance to match the biota. BIOENV was employed to relate plankton distribution with environmental parameters such as salinity, temperature, pH, chloride, dissolved oxygen, ammonia, nitrite, nitrate, phosphate and silicate.

Using R programming, the pairwise cross correlation between each environmental variable and the structural parameters (density, number of species, d , $H' \log^2$, and J') of phytoplankton was evaluated using Pearson's correlation (Wei *et al.*, 2017). The Pearson coefficient's magnitude was used to interpret the correlation values; a correlation of less than 0.5 was considered

low, a correlation of 0.01 was considered moderate, and a correlation of 0.001 was considered strong.

Pearson's correlation (Wei *et al.*, 2017) analysis was conducted using the R programming to assess pair-wise cross-correlation between each environmental variable and structural parameters (density, number of species, d , $H' \log^2$, and J') of phytoplankton. The correlation values were interpreted based on the magnitude of the Pearson coefficient, with values less than 0.5 indicating a low correlation, values of 0.01 indicating a moderate correlation, and values of 0.001 indicating a high correlation.

RESULTS

Hydrographical Parameters

Temporal changes

The sampling locations mean variance in temperature ranges are (Mean \pm SD in pre-monsoon (Pr): 29.00 ± 0.0 , monsoon (M): 27.90 ± 0.37 , and post-monsoon (Ps): 26.36 ± 0.64 °C) and salinity (Pr: 32.29 ± 0.17 , M: 31.47 ± 0.14 , and Ps: 31.29 ± 0.18 PSU), ammonia (Pr: 3.70 ± 0.21 , M: 3.39 ± 0.29 , and Ps: 2.13 ± 0.33 mg/L), silicate (Pr: 3.94 ± 0.37 , M: 3.63 ± 0.11 , and Ps: 2.59 ± 0.28 mg/L), and Chloride (Pr: 5.47 ± 1.65 , M: 3.99 ± 0.48 , and Ps: 3.70 ± 0.48 mg/L) generally decreased from pre-monsoon to post-monsoon seasons, whereas nitrate (Pr: 5.82 ± 0.24 , M: 5.82 ± 0.41 , and Ps: 6.38 ± 1.65 mg/L) showed the opposite trend as it increased from pre-monsoon to post-monsoon seasons (Figure 2). The nitrite (Pr: 0.52 ± 0.19 , M: 0.69 ± 0.13 , and Ps: 0.54 ± 0.27 mg/L) and phosphate (Pr: 0.22 ± 0.02 , M: 0.33 ± 0.04 , and Ps: 0.16 ± 0.01 mg/L) increased from pre-monsoon to monsoon then decreased towards post-monsoon

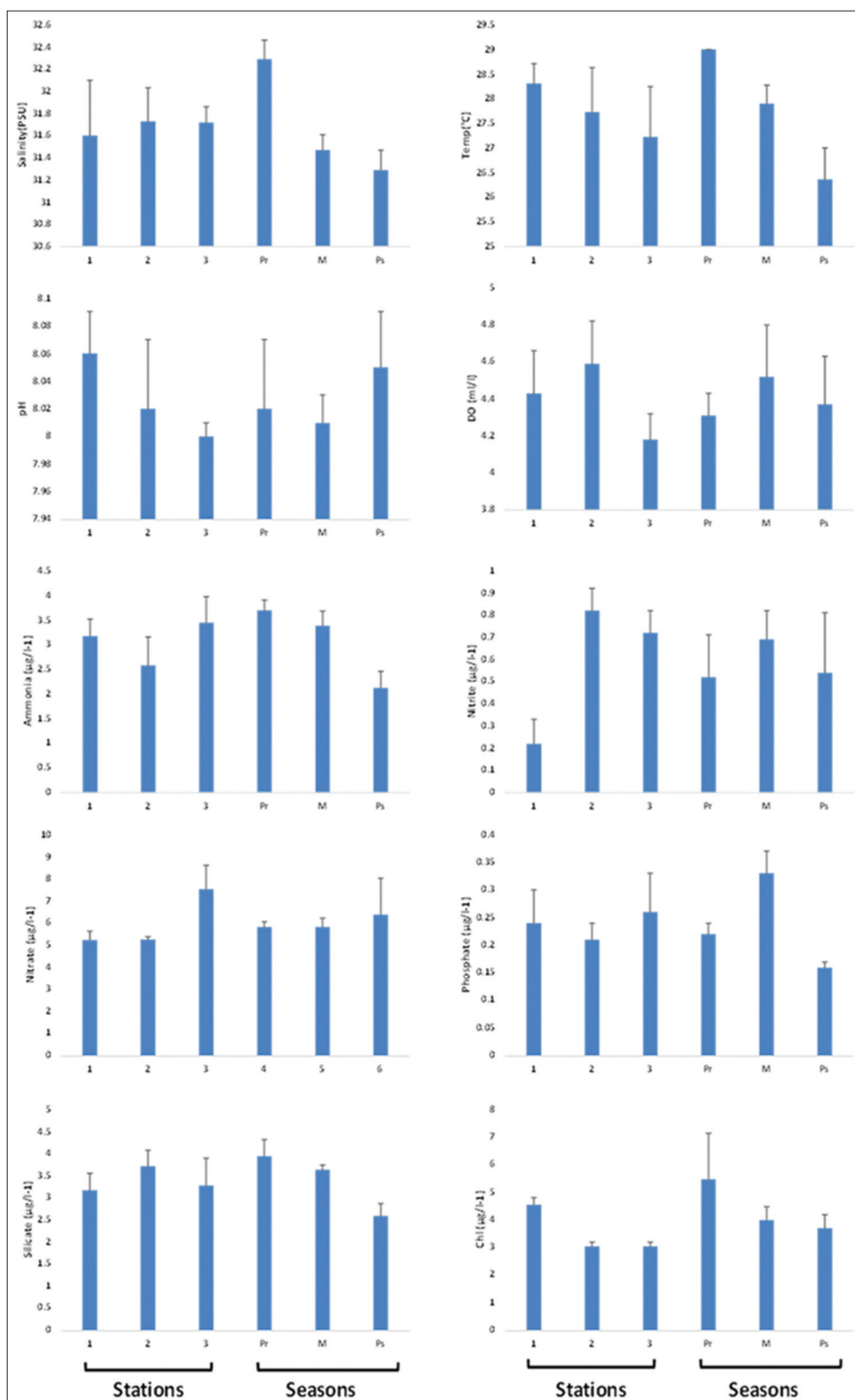


Figure 2: Hydrographical parameters (salinity, temperature, pH, dissolved oxygen), nutrients (ammonia, nitrite, nitrate, phosphate, silicate and chlorophyll (Chl)) in Thamirabarani estuary

(Figure 2). The pH (Pr: 8.02 ± 0.05 , M: 8.01 ± 0.02 , and Ps: 8.05 ± 0.04 mg/L) decreased from pre-monsoon to monsoon and then increased towards post-monsoon. However, dissolved oxygen (Pr: 4.31 ± 0.12 , M: 4.52 ± 0.28 , and Ps: 4.37 ± 0.26 mg/L)

did not vary significantly between the seasons (Figure 2). PERMANOVA analysis showed significant differences only in ammonia ($F=7.55$, $P<0.05$) and phosphate ($F=18.747$, $P<0.05$) with seasons.

Spatial changes

The temperature's range of mean values are Mean \pm SD (st. 1: 27.73 ± 0.90 , st. 2: 28.31 ± 0.40 and st. 3: 27.22 ± 1.02 °C), pH (st. 1: 8.06 ± 0.03 , st. 2: 8.02 ± 0.05 , and st. 3: 8 ± 0.01), and Chl (st. 1: 4.55 ± 0.25 , st. 2: 3.04 ± 0.15 , and st. 3: 3.04 ± 0.15 mg/L) decreased from station 1 to stations 3 (Figure 2). However, nitrate (st. 1: 5.26 ± 0.12 , st. 2: 5.24 ± 0.40 and st. 3: 7.53 ± 1.08 mg/L) increased from station 1 to station 3. Ammonia (st. 1: 3.18 ± 0.34 , st. 2: 2.58 ± 0.58 , and st. 3: 3.45 ± 0.53 mg/L) and phosphate (st. 1: 0.21 ± 0.03 , st. 2: 0.24 ± 0.06 , and st. 3: 0.26 ± 0.07 mg/L) decreased from station 1 to station 2 then increased towards station 3, whereas nitrite (st. 1: 0.82 ± 0.10 , st. 2: 0.22 ± 0.11 , and st. 3: 0.72 ± 0.10 mg/L) silicate (st. 1: 3.71 ± 0.36 , st. 2: 3.17 ± 0.38 , and st. 3: 3.28 ± 0.61 mg/L), and dissolved oxygen (st. 1: 4.43 ± 0.23 , st. 2: 4.59 ± 0.23 , and st. 3: 4.18 ± 0.14 mg/L) increased from station 1 to station 2 then decreased towards station 3 (Figure 2). Generally, the spatial variations in salinity (st. 1: 31.6 ± 0.50 , st. 2: 31.73 ± 0.30 , and st. 3: 31.72 ± 0.14 (PSU) were similar. PERMANOVA analysis indicated significant differences only in ammonia ($F=25.72$, $P<0.05$), nitrite ($F=9.4183$, $P<0.05$), salinity ($F=7.32$, $P<0.05$), and temperature ($F=14.03$, $P<0.05$) with stations.

Principal Component Analysis

The Principal component analysis (PCA) was calculated using the rotated matrix analysis. The first three axes explained more than 80% of the variability (Table 1). The first axis (44.9% of variations) generally divided the seasons according to the natural gradient from pre-monsoon, monsoon and post-monsoon; the variation was explained by the salinity due to the rainfall, significant changes in ammonia (0.475) and Chloride (0.819). Variability in the second axis (24.7%) was explained by change in nitrate (0.824). However, the third axis (18.5%) was related to the nitrite (0.824) and silicate (0.461).

Species composition and distribution of phytoplankton

A total of 65819 individuals representing 49 taxa were recorded from the three stations sampled in three seasons as Pre-monsoon (Pr), monsoon (M) and post-monsoon (Ps) (Table 2). Bacillariophyceae contributed more to the total density (66.73%) followed by dinophyceae (21.71%) and mediophyceae (6.64%). The contribution of cyanophyceae (2.47%) and oscillatoriohyceae (2.44%) was comparatively less. The mean number of species (S) (Pr: Mean \pm SD 43 ± 1 , M: 37 ± 1 , and Ps: 35 ± 1 cells/L), mean density (N) (Pr: 8463 ± 72 , M: 7094 ± 181 , and Ps: 6448 ± 262 ind./m³) decreased from pre-monsoon to post-monsoon seasons. In station-wise, the mean number of species (1: Mean \pm SD 40 ± 3 , 2: 38 ± 2 , and 3: 38 ± 3 cells/L) decreased from station 1 to station 2 and 3, however, mean density was similar in the three stations (1: 7272 ± 751 , 2: 7358 ± 535 , and 3: 7376 ± 558 ind./m³). PERMANOVA revealed significant seasonal differences in the number of species ($F=22.02$, $P<0.05$) and density ($F=16.8$, $P<0.05$), whereas in station-

Table 1: The result of PCA for analysis of environmental parameters distribution in the study area

Axis	1	2	3	4
Eigenvalues	0.192	0.105	0.079	0.0377
%Variation	44.9	24.7	18.5	8.8
Cum. %Variation	44.9	69.7	88.2	97
Loading of Variable				
Salinity (ppt)	0.041	-0.01	0.029	0.101
Temp (°C)	0.165	-0.282	0.007	0.163
pH	-0.005	0.002	-0.012	-0.021
DO (mg/L)	-0.076	-0.141	-0.048	0.099
Ammonia ($\mu\text{g/L}^{-1}$)	0.475	-0.179	0.161	0.656
Nitrite ($\mu\text{g/L}^{-1}$)	-0.072	0.128	0.824	-0.336
Nitrate ($\mu\text{g/L}^{-1}$)	0.136	0.824	0.157	0.356
Phosphate ($\mu\text{g/L}^{-1}$)	0.07	-0.045	0.15	0.147
Silicate ($\mu\text{g/L}^{-1}$)	0.201	-0.395	0.461	0.004
Chl ($\mu\text{g/L}^{-1}$)	0.819	0.124	-0.18	-0.512

wise, the number of species and density homogenously distributed ($P>0.05$).

Among the plankton, bacillariophyceae was dominant, with consistently being the most dominant group at all stations and seasons (Table 2). Altogether, 32 species of bacillariophyceae belonging to 19 genera and 14 families were identified. The mean \pm SE of average density was high in the pre-monsoon season (5479 ± 283 ind./m³) and lower in the post-monsoon season (4405 ± 145) and the density was 4757 ± 202 in the monsoon season. Station-wise, the lowest average density was recorded at station 1 (4562 ± 182) and the highest at station 3 (5099 ± 294) and the density was 4981 ± 493 ind./m³ at station 2. The dominant bacillariophyceae encountered were *Biddulphia obtuse*, *Chaetoceros diversus*, *Coscinodiscus sp.*, *Skeletonema costatum*, and *Odontella mobiliensis*.

The taxa Dinophyceae are with ten species belonging to 6 genera and 5 families. The dominant species were *Prorocentrum micans* and *Prorocentrum sp.* (Table 2). The highest density was in pre-monsoon (1968 ± 69 ind./m³) and the lowest in post-monsoon season (1234 ± 168 ind./m³) and the density was 1755 ± 165 ind./m³ in monsoon season. Among the stations, a higher density was recorded at station 1 (1585 ± 263 ind./m³) and a lower density at station 3 (1487 ± 263 ind./m³) and the density was 1690 ± 148 ind./m³ at station 2.

Besides Dinophyceae, Mediophyceae (three species belonging to two genera and two families), Cyanophyceae (two species belonging to two genera and two families), and Oscillatoriohyceae (two species belonging to two genera and two family) were recorded in very low numbers (Table 2).

Diversity

The Margalef richness (d) index (Pr: Mean \pm SD 4.64 ± 0.12 , M: 4.10 ± 0.09 , and Ps: 3.84 ± 0.10 cells/l) decreased from premonsoon to post-monsoon seasons. While the Pielou's evenness index (J') (Pr: 0.98, M: 0.99, and Ps: 0.99) increased from premonsoon to post-monsoon seasons (Table 3). However,

Table 2: Density (individuals/unit) of phytoplankton species in the study area

Species	Family	PRM1	MON1	POM1	PRM2	MON2	POM2	PRM3	MON3	POM3
<i>Nitzschia</i> sp.	Bacillariaceae	160	150	168	150	180	180	168	170	180
<i>Nitzschia longissima</i>	Bacillariaceae	160	0	150	0	0	195	180	0	170
<i>Nitzschia seriata</i>	Bacillariaceae	280	298	0	290	280	0	0	0	0
<i>Bacillaria paradoxa</i>	Bacillariaceae	150	150	0	0	0	0	138	130	0
<i>Bellerochea</i> sp.	Bellerocheaceae	180	160	150	130	130	160	150	158	180
<i>Bellerochea malleus</i>	Bellerocheaceae	135	135	130	128	130	180	168	150	150
<i>Gyrosigma</i> sp.	Pleurosigmataceae	164	0	198	166	0	146	288	0	187
<i>Gyrosigma balticum</i>	Pleurosigmataceae	182	182	138	158	141	141	150	160	154
<i>Pleurosigma normanii</i>	Pleurosigmataceae	165	168	156	168	168	178	157	157	155
<i>Navicula gracilis</i>	Naviculaceae	160	145	160	0	0	0	298	284	287
<i>Biddulphia</i> sp.	Biddulphiaceae	180	280	260	0	0	0	268	258	255
<i>Biddulphia obtuse</i>	Biddulphiaceae	260	245	255	287	266	289	298	282	295
<i>Odontella sinensis</i>	Biddulphiaceae	160	168	184	157	155	165	132	157	184
<i>Bacteriastrium</i> sp.	Chaetocerotaceae	380	350	0	0	0	0	276	288	0
<i>Bacteriastrium comosum</i>	Chaetocerotaceae	168	120	168	158	120	148	144	130	176
<i>Chaetoceros</i> sp.	Chaetocerotaceae	158	168	110	120	157	130	138	169	160
<i>Chaetoceros affinis</i>	Chaetocerotaceae	110	190	100	158	110	150	156	122	122
<i>Chaetoceros curvisetus</i>	Chaetocerotaceae	180	180	172	190	193	165	155	183	165
<i>Chaetoceros diversus</i>	Chaetocerotaceae	290	278	287	270	255	266	0	284	0
<i>Coscinodiscus</i> sp.	Coscinodiscaceae	280	272	280	290	168	160	246	255	246
<i>Coscinodiscus centralis</i>	Coscinodiscaceae	160	0	0	158	0	0	147	0	0
<i>Planktonella</i> sp.	Coscinodiscaceae	160	160	140	190	190	182	142	150	160
<i>Lauderia borealis</i>	Lauderiaceae	360	128	0	0	155	0	290	118	0
<i>Skeletonema costatum</i>	Skeletonemaceae	168	198	164	284	268	256	290	284	290
<i>Eucampia</i> sp.	Hemiaulaceae	182	156	157	168	184	190	198	188	187
<i>Astrionella</i> sp.	Fragilariaceae	0	0	0	168	160	178	148	180	168
<i>Astrionella glacialis</i>	Fragilariaceae	180	152	160	146	180	180	167	167	175
<i>Rhizosolenia</i> sp.	Rhizosoleniaceae	180	0	170	160	0	180	157	0	160
<i>Rhizosolenia alata</i>	Rhizosoleniaceae	197	138	138	187	157	180	155	142	160
<i>Ditylum</i> sp.	Lithodesmiaceae	0	0	0	150	120	120	0	0	0
<i>Ditylum brightwelli</i>	Lithodesmiaceae	160	160	0	280	298	0	260	240	0
<i>Odontella mobiliensis</i>	Triceratiaceae	210	198	160	214	190	286	190	180	290
<i>Eucampia zoodiacus</i>	Hemiaulaceae	139	157	126	190	184	210	168	138	157
<i>Leptocylindrus</i> sp.	Leptocylindraceae	156	156	189	188	150	148	198	130	179
<i>Leptocylindrus danicus</i>	Leptocylindraceae	0	0	0	590	0	280	270	0	270
<i>Ceratium</i> sp.	Ceratiaceae	180	147	124	160	154	159	0	0	0
<i>Ceratium furca</i>	Ceratiaceae	180	0	190	180	0	178	165	0	164
<i>Ceratium macroceros</i>	Ceratiaceae	280	234	0	0	0	0	346	346	0
<i>Peridinium</i> sp.	Peridiniaceae	138	112	112	180	158	180	160	159	160
<i>Protoperidinium</i> sp.	Protoperidiniaceae	290	260	0	280	250	0	210	259	0
<i>Prorocentrum</i> sp.	Prorocentraceae	180	130	160	360	340	340	310	250	250
<i>Prorocentrum micans</i>	Prorocentraceae	310	298	284	290	257	280	250	260	262
<i>Dinophysissp.</i>	Dinophysaceae	180	0	162	190	0	240	0	0	0
<i>Dinophysis punctata</i>	Dinophysaceae	180	130	150	168	168	170	159	136	136
<i>Dinophysis caudata</i>	Dinophysaceae	160	184	0	178	210	0	240	240	0
<i>Microcystis</i> sp.	Microcystaceae	160	148	168	180	165	152	0	0	0
<i>Anabaena</i> sp.	Aphanizomenonaceae	0	180	0	0	182	0	1	290	0
<i>Oscillatoria</i> sp.	Oscillatoriaceae	210	0	182	168	0	168	230	0	198
<i>Trichodesmium</i> sp.	Microcoleaceae	1	146	0	0	166	0	2	138	0

PRM1-Premonsoon 1; MON1-Monsoon 1; POM1- Postmonsoon 1; PRM2- Premonsoon 2; MON2- Monsoon 2; POM2- Postmonsoon 2; PRM3- Premonsoon 3; MON3- Monsoon 3; POM3-Postmonsoon 3

the Shannon–Wiener ($H'(\log^2)$) (Pr: 5.32 ± 0.04 , M: 5.16 ± 0.03 , and Ps: 5.07 ± 0.04) index showed similar values between the seasons. PERMANOVA analysis showed significant differences in Margalef richness ($F=20.84$, $P < 0.05$), Pielou's evenness ($F=17.487$, $P < 0.05$), and Shannon–Wiener ($F= 17.18$, $P < 0.05$) indices with seasons. In station-wise, the Margalef richness (1: 4.12 ± 0.15 , 2: 4.35 ± 0.28 , and 3: 4.11 ± 0.29), Pielou's evenness (1: 0.99, 2: 0.99 and 3: 0.98), and Shannon–Wiener (1: 5.17 ± 0.05 , 2: 5.23 ± 0.09 , and 3: 5.15 ± 0.09) (Table 3) indices did not show such significant variations and PERMANOVA results also indicated homogenous distribution ($P > 0.05$).

Multivariate analysis of community structure

The dendrogram showed four groups (one each in each season and another one is nearby seasons) (Figure 3). The Pearson's correlation (r) was 0.43 having the sample statistic of 0.2% shows significant serial changes in species composition between the seasons.

Principal Coordinate (PCO) and SIMPER

The output derived from the analysis of the Principal Coordinate indicated that the first axis explained 58.6% of

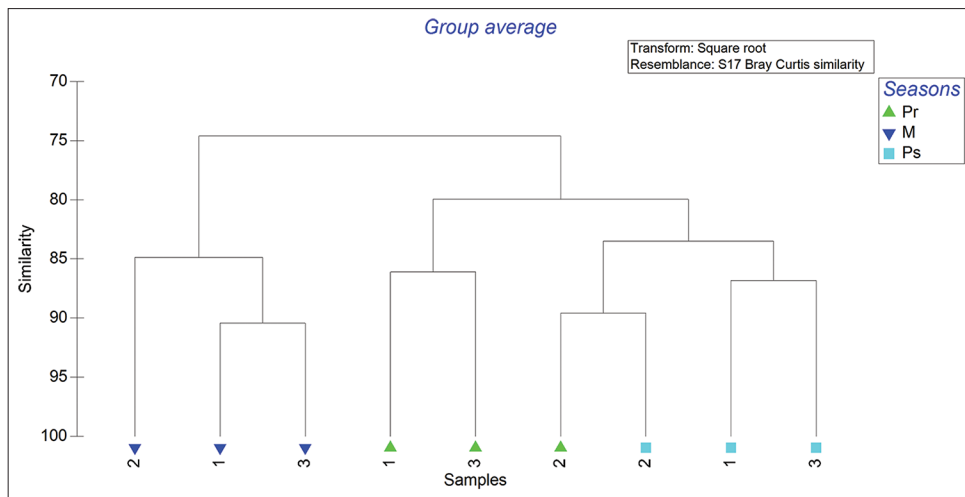


Figure 3: Dendrogram drawn for plankton distribution in the Thamirabarani Estuary. (Pr-premonsoon; M-monsoon; Ps-postmonsoon; 1-station 1; 2-station 2; 3-station 3)

the total variations and the second axis 30.1%. The first axis separated clearly all the seasons and the second axis separated all the stations (Figure 4). As per the SIMPER analysis, characteristic species of premonsoon were *Anabaena* sp., *Gyrosigma* sp., *Oscillatoria* sp., *Leptocylindrus danicus*, *Nitzschia longissimi*, *Ceratium furca*, *Rhizosolenia* sp., *Coscinodiscus centralis*, in monsoon were *Protoperidinium* sp., *Ditylum brightwelli*, *Anabaena* sp., *Dinophysis caudata*, *Trichodesmium* sp., *Bacteriastrium* sp., *Lauderia borealis*, *Ceratium macroceros*, *Nitzschia seriata*, and *Bacillaria paradoxa*, and in postmonsoon were *Oscillatoria* sp., *Ceratium furca*, *Gyrosigma* sp., *Nitzschia longissimi*, *Rhizosolenia* sp., *Leptocylindrus danicus*, and *Dinophysis* sp. in station-wise, while *Biddulphia* sp., *Navicula gracilis*, *Bacteriastrium* sp., *Ceratium macroceros*, *Bacillaria paradoxa*, *Microcystis* sp., *Ceratium* sp., *Nitzschia seriata*, *Dinophysis* sp. clearly showed affinity towards station 1, *Leptocylindrus danicus*, *Astrionella* sp., *Ditylum* sp., *Microcystis* sp., *Ceratium* sp., *Nitzschia seriata*, *Dinophysis* sp. showed affinity towards station 2, and *Astrionella* sp., *Leptocylindrus danicus*, *Navicula gracilis*, *Biddulphia* sp., *Ceratium macroceros*, *Bacteriastrium* sp., *Bacillaria paradoxa* indicated affinity towards station 3.

Relationships between the Plankton Composition and Environmental Parameters

Pearson’s correlation

Pearson’s correlation values for temperature, salinity, pH, dissolved oxygen, ammonia, silicate, Chloride, nitrate, nitrite, phosphate, number of species, density, Margalef richness (d), Pielou’s evenness index (J’), and Shannon–Wiener (H’(log2)) for the study period. Temperature showed a weak positive correlation with silicate (r2=0.75), ammonia (r2=0.73), density (r2=0.70), and positive correlation with Margalef richness (d) (r2=0.83), number of species (r2=0.82), Shannon–Wiener (H’(log2)) (r2=0.82), and weak negative correlation with Pielou’s evenness index (j) (r2=-0.75). Salinity had a weak positive correlation with Margalef richness (d) (r2=0.74),

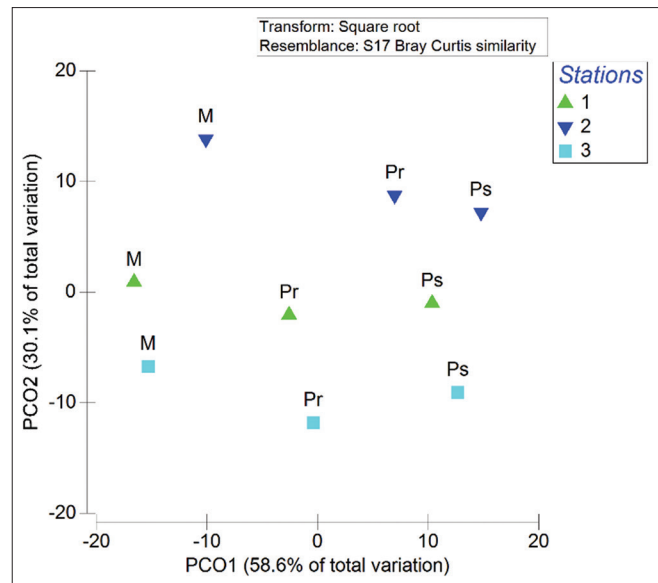


Figure 4: Principal Coordinate (PCO) for plankton density in Thamirabarani Estuary. (Pr-premonsoon; M- monsoon; Ps- postmonsoon; 1-station 1; 2-station 2; 3-station 3)

Table 3: Mean, standard deviation and range in density of phytoplankton (unit) at different stations and seasons in the study area

Seasons and stations	S	N	d	J'	H'(log2)
PRM	43±1	8463±72	4.64±0.12	0.98	5.32±0.04
MON	37±1	7094±181	4.10±0.09	0.99	5.16±0.03
POM	35±1	6448±262	3.84±0.10	0.99	5.07±0.04
St-1	40±3	7272±751	4.35±0.28	0.99	5.23±0.09
St-2	38±2	7358±535	4.12±0.15	0.99	5.17±0.05
St-3	38±3	7376±558	4.11±0.29	0.98	5.15±0.09

S-The mean number of Species; N-The mean Density; d-Margalef’s species richness; J’-Pielou’s evenness; H’-Shannon Wiener diversity (log2); PRM-Premonsoon; MON-Monsoon; POM-Postmonsoon; St. 1- Station -1; St. 2- Station -2; St. 3- Station -3

number of species (r2=0.76), Shannon–Wiener (H’(log2)) (r2=0.73), and negative correlation with Pielou’s evenness

index (j) ($r^2=-0.68$). Ammonia displayed a weak positive correlation with Margalef richness (d) ($r^2=0.67$), number of species ($r^2=0.68$), density ($r^2=0.67$), and negative correlation with Pielou's evenness index (j) ($r^2=-0.78$). Silicate showed a weak positive relationship with density ($r^2=0.71$) and a negative relationship with Pielou's evenness index (j) ($r^2=-0.72$). Chloride showed only a negative correlation with Pielou's evenness index (j) ($r^2=-0.72$).

Bio-env

The τ value of 0.683 fell away from the histogram, showing a significant relationship between changes in environmental parameters and plankton (Table 4). Pearson's correlation values in the best combinations varied from 0.683 to 0.413. An environmental variable manifested in the highest combination (0.683) was phosphate, beside this, combinedly pH and phosphate influenced the community in next level at 0.642. In addition to this environmental parameters, ammonia, phosphate and silicate were manifested in the combination with lowest correlation (0.413).

DISCUSSION

Environmental Parameters

The rate of seasonal rainfall and lower atmospheric temperatures can dominate water temperature declines (Vajravelu *et al.*, 2018). The water temperature in the study area varied substantially from seasonal cyclic changes in air temperature and precipitation in certain areas which was most likely related to the time of sampling since it was reliant on the presence of sunshine and heating impact (Romin *et al.*, 2021). The water temperature fluctuated insignificantly across sample sites and did not appear to constitute a hazard to the aquatic system (Aknaf *et al.*, 2017). The station's proximity to the river's mouth opening means that the river's replenishment of fresh water has an effect on ST 3's lowest temperature during the post-monsoon season (Romin *et al.*, 2021).

During the monsoon season, the estuary is directly linked to the sea which progress to declining salinity, and the variation of salinity in estuaries, backwaters, and mangrove waterways is guide by the entry of freshwater through the land flowoff administrate by monsoon or mangrove waterways (Srinivasan & Natesan, 2013). The minimal vertical mixing of water by the

tidal cycles modified the salinity of the estuary (Xia *et al.*, 2011). Seasonally, a low pH value was recorded in the post-monsoon period, whereas a slightly higher value was observed in the monsoon period which indicated that the coastal environment is beneficial to aquatic ecosystems (Santhanam & Perumal, 2003). The concentration of Dissolved Oxygen (DO) is quite inversely related to the temperature of an aquatic body. The high dissolved oxygen levels are most likely due to significant oxygenation of the surface water caused by freshwater input, while the lower DO levels reported in summer were the result of slowing development and mortality of algae caused by plankton development arrest and mass death, while the monsoon arrives, both of which need dissolved oxygen (Aknaf *et al.*, 2017). The concentrations of DO and pH in water alter over time and are frequently impacted by other variables, including temperature, salinity and all of which are important for successful organisms and water resource management (Araoye, 2009).

Nutrients

Ammonia alone showed a significant change with seasonal fluctuation. During the monsoon season, ammonia concentrations increase due to runoff excess rainwater and phytoplankton breakdown (Senthilkumar *et al.*, 2008; Thangaradjou *et al.*, 2013). In the present observation st.3 shows a favorable correlation between ammonia with temperature, whereas in St.2, a negative correlation exists between DO and nitrate. Anthropogenic activities are the major source of ammonia and nitrate in the present study area. The estuaries shallower parts have higher nutrient concentrations because their reduced water mass makes them more prone to human activity and enhanced river impact. Nutrient enrichment has become a societal concern as a result of inadequate protection measures and rising human demands on the environment (Howarth & Marino, 2006; Boyer *et al.*, 2009; Srichandan *et al.*, 2019). Eutrophic water quality in the estuary promoted phytoplankton growth while causes the estuarine environment (Yi *et al.*, 2018). Silicate concentration was greater in the pre-monsoon than post-monsoon season. This was caused by terrestrial runoff water to reach the estuary ecosystem (Vajravelu *et al.*, 2018). The nitrate concentration varied seasonally, lower nitrate concentrations may be due to increased nitrate consumption by photosynthetic organisms and the invasion of neritic water, which contains very little nitrate (Govindasamy *et al.*, 2000).

Phosphate alone reveals a significant seasonal shift ($p<0.05$, $F=7.55$) with seasonal inconsistency. Higher levels of inorganic phosphate may be connected to heavy rain interference generated by human activity and terrestrial excess water (Satpathy *et al.*, 2009), whereas the low value in summer may be attributable to photoautotrophic phosphate utilization and sediment buffering under contrary environmental conditions (Perumal *et al.*, 2009).

Composition, Population, and Diversity of Phytoplankton

In three stations, about 65,819 individuals were recorded in all seasons throughout the present research study. Bacillariophyceae

Table 4: BEST analysis matching phytoplankton and with environmental variables

Number of variables	Correlation	Variable selections
1	0.683	Phosphate
2	0.642	pH, Phosphate
2	0.561	Salinity, Phosphate
3	0.537	Salinity, pH, Phosphate
2	0.485	Do, Phosphate
2	0.461	Temperature , Phosphate
3	0.459	Temperature, pH, Phosphate
3	0.458	pH, Do, Phosphate
3	0.421	Salinity, Do, Phosphate
3	0.413	Ammonia, Phosphate, 9

dominated by 32 species, followed by Dinophyceae 10, Mediophyceae 3, and 2 species representing Cyanophyceae and Oscillatoriophyceae. However, our findings were consistent with earlier studies on Santragachi lake in West Bengal (Ghosh *et al.*, 2012), Parangipettai coastal water in India (Vajravelu *et al.*, 2018), Pasur river estuary in Bangladesh (Zinat *et al.*, 2021), China seas (Cui *et al.*, 2018; Zhong *et al.*, 2021; Wang *et al.*, 2022; Wei *et al.*, 2022) and Mekong delta in Vietnam (Truong *et al.*, 2018). Diatoms, dinoflagellates, haptophytes, and tiny prasinophytes are the most diverse and ecologically significant eukaryotic phytoplankton taxa in modern oceans, with some frequently forming huge blooms visible in satellite photos (Not *et al.*, 2022). Do Rosário Gomes *et al.* (2014) studied phytoplankton species distribution, and Lotliker *et al.* (2018), Xiang *et al.* (2019) and Goes *et al.* (2020) demonstrated that the transition from diatoms, In Chabahar Bay in the Gulf of Oman, the northeast monsoon had the highest phytoplankton density ($1,168,112 \pm 136,189$ Cells/L) during vertical mixing of the water column caused by evaporative cooling resulted in a high concentration of nutrients in the euphotic layer (Jalili *et al.*, 2022).

The dominance of Bacillariophyceae come across with *Biddulphia obtuse*, *Chaetoceros diversus*, *Coscinodiscus* sp., *Skeletonema costatum*, and *Odontella mobiliensis* which is due to intermixing of nutrients during pre-monsoon and monsoon seasons and release of nutrients by decomposition to resulted phytoplankton density, correspondingly (Palleyi *et al.*, 2008; Sasamal *et al.*, 2005). Similarly, *Biddulphia obtuse* was predominantly observed in all the stations and all seasons. *Ceratium* spp. was identified as the most frequent dinoflagellate found in the Gulf of Mannar region (Jayasiri & Priyadharshini, 2007). Hilaluddin *et al.* (2020) reported certain diatoms *Biddulphia* sp., *Skeletonema costatum* and *Coscinodiscus* sp. react quickly to environmental changes by altering their community composition.

Total phytoplankton quantity and diatom density have varied significantly in season-wise. El-Gindy and Dorghan (1992) demonstrate a considerable seasonal change on phytoplankton diversity to environment disparities at different stations. Choudhary and Pal (2010) found that diatoms predominate in Indian coastal water. The productivity of the estuarine ecosystem primarily depends on phytoplankton, which accounts for approximately 90% of total estuarine primary production (Srinivasan & Natesan, 2013). A decreased level of phytoplankton diversity was recorded by various researchers by both seasonal and spatial variations in different estuaries. Canini *et al.* (2013) at Kudat coastal areas by 37 genera in the Philippine mangrove estuary; 30 genera in the Sungai Brunei estuary (Majewska *et al.*, 2017); 24 species in the Kota Kinabalu wetland region (Azad & Jinau, 2020) and 24 genera in Malaysia Kudat, Subah's coastal region (Romin *et al.*, 2021).

Different factors can affect changes in plankton density depending on local climatic conditions (Al-Yamani *et al.*, 2010). Several studies found that the phytoplankton density was decreased during the post-monsoon season (Hassan *et al.*, 2010),

A reduction in phytoplankton density may have been caused by temperature drop, strong water currents, inflow of fresh water, and limited nutritional (Saraji *et al.*, 2014; Mirzaei *et al.*, 2017). The physical and chemical factors of the estuarine environment exhibit its influence on the phytoplankton composition, density, and growth (Vajravelu *et al.*, 2018). Salinity, which is mostly caused by evaporation and dilution, influences phytoplankton density (Khomayis, 2002; Schumann *et al.*, 2006).

Numerous studies have demonstrated how pH and water temperature affect photosynthesis, respiration, phytoplankton development, and community succession in aquatic environments (Williamson *et al.*, 2010; Moore, 2010). During the pre-monsoon season, the average plankton density was superior; however, in the post-monsoon and monsoon season, its density was inferior. The least value for phosphate and nitrate observed during monsoon seasons of the present study was similar to the results of Rahaman *et al.* (2013). The relentless utilization of phosphate by phytoplankton and other primary producers may reduce its level during monsoon.

Phytoplankton was shown significantly positively linked with nitrite in the stream area. The highest phytoplankton density at S3 is most likely due to the outflow of metabolic waste products (domestic sewage), which contain high nutrients and stimulate phytoplankton growth (manmade activity). An increased Chloride concentration may alter osmoregulatory activities in various phytoplankton species. At low chloride concentrations, an elevated Chlorophyceae population was observed, while at higher chloride concentrations enhanced the cyanobacteria population. Because high chloride concentration nutrient acceptance by rivalry cyanobacteria, declines chlorophyceae diversity in the present study was in consistent with the findings of Pilkaitytė *et al.* (2004) and McGowan *et al.* (2020).

The higher Marglef's species richness (d) in pre-monsoon (4.64 ± 0.12) and lower in post-monsoon (3.84 ± 0.10) were recorded respectively, that exhibited a considerable fluctuation throughout all seasons. Choudhury and Pal (2010) found a low value of species richness in post-monsoon (0.705-2.914) and pre-monsoon seasons (0.787-1.446) in West Bengal coastal waters. The highest diversity index was observed during the post-monsoon season, which corresponds with the data of Baliarsingh *et al.* (2015) from coastal waters off the Rushikulya estuary in India. Dayala *et al.* (2014) stated that changes in phytoplankton density and diversity in different months/seasons may be related to variable water conditions. Because phytoplanktons are the key link in any aquatic ecosystem's food chain, this information on species availability in different months can be utilized to connect with the fishing in this ecosystem (Ghosh *et al.*, 2012). Pre-monsoon had unlimited species richness (d) than other seasons. The Shannon Weiner index (H') showed a higher value in pre-monsoon and lower in post-monsoon, however, the degree of modification was fairly unsure. When Shannon index (H) was larger than 3, it was at its peak in the pre-monsoon season, suggesting that the water in this estuary is favorable for phytoplankton growth and production (Zinat, 2021).

Pielou's Evenness (J') highest value in monsoon and post-monsoon, exhibited a little fluctuation among seasons, indicating that the ecosystem's species distribution is homogeneous. The post-monsoon season may have the highest value of diversity indices because the high phytoplankton species make up the favorable environmental circumstances with a higher concentration of nutrients in the water column (Dupuis & Hann, 2009). A high positive correlation was observed between the diversity indices (species richness, evenness, dominance & Shannon Weiner). A positive correlation has existed between the diversity indices with temperature and salinity ($r=0.76$ to 0.70). The species diversity index values reported for all seasons were greater than those observed by Gharib *et al.* (2011) and Choudhury and Pal (2010) in the southeastern Mediterranean Sea. Dendrogram analysis indicated that the monsoon season is deviated from pre-monsoon and post-monsoon. Among the monsoon, stations 1 and 3 was deviated from station 2. The plankton distribution in station 2 was completely deviated from stations 1 and 3 for both and post-monsoon. The current study found a large number of diatoms and dinoflagellates, which agrees with the findings of Saraji *et al.* (2014) and Mirzaei *et al.* (2017). According to SIMPER study, the species contribute most to the average similarity within groups (various seasons and stations) were found by cluster analysis utilizing the similarities (Jalili *et al.*, 2022).

CONCLUSION

The present study indicates that the temporal and spatial distribution of water quality varies greatly across stations and seasons in the Thamirabarani estuary. During the northeast monsoon, the water current flows from north to south, with nutrient-rich freshwater discharged from the Thamirabarani river altering nutrient dynamics in the fishing grounds, ultimately increasing nutrient concentration. Bacillariophyceae were consistently the most dominant group at all stations and seasons. Due to tidal action and anthropogenic activities, phytoplankton diversity/density was minimal in S1 and S2 when compared to S3. Phosphate, and pH conditions are significantly essential elements that can influence variations in phytoplankton compositions of temporal and spatial distributions. Monsoon rainwater brings large amount of minerals/nutrients which significantly enhances the phytoplankton density by access them. This finding shows that the estuarine environmental factors have ecological significance. Comprehensive monitoring of qualitative and quantitative characteristics of various phytoplankton communities is warranted for a better understanding of their potential repercussions.

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