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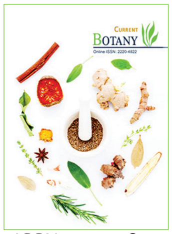

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Comparison of phytoplankton community structure in two tropical estuaries of East Coast of India

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ABSTRACT

Dissolved nutrients concentrations in estuarine waters play important role in shaping phytoplankton community of two Indian river estuaries. The first study site was Saptamukhi river estuary which is a part of Hooghly-Matla estuarine system is located in the Indian Sundarban and second study site was Mahanadi estuary located in Orissa coast (adjacent to industrial and coastal fishing zone). The Saptamukhi estuary received less anthropogenic waste compared to Mahanadi estuary and the main source of nutrient is autochthonous due to huge litter from surrounding mangrove forest. Mahanadi estuarine water received nutrients from industrial effluent and fishing waste. The Saptamukhi estuarine water had less dissolved inorganic phosphorus concentration but huge dissolved inorganic nitrogen concentration. High dissolved silicate concentration might encourage the diatom growth over other phytoplankton group here. In Mahanadi estuarine water, high amount of dissolved inorganic phosphate coming from the phosphate industries supported the growth of Cyanophyceae, Chlorophyceae and Dinophyceae. Chlorophycean genus *Pediastrum* bloom observed in the post-monsoon during this study. In Saptamukhi Estuarine water, in spite of high phytoplankton population in post-monsoon season no individual species reached bloom condition during the study period. In both the estuarine water Gross Primary Productivity was high but the community respiration was higher. Monsoonal runoff from land considerably changed the community in both estuarine water. Dissolve inorganic nitrogen and dissolved inorganic phosphate ratio played major role for the community change of phytoplankton in two estuarine waters.

KEYWORDS: Phytoplankton; diatom; estuary; nutrients

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INTRODUCTION

Phytoplankton, can be categorized as prokaryotic cyanobacteria and eukaroytic algal groups, is responsible for primary production, supporting food webs and play a central role in essential elements, nutrient, and oxygen cycling in estuaries. They respond to changes in different environmental conditions [1,2]. Eutrophication or change in transparency of water column [3,4] alter the structural characteristics of phytoplankton communities (i.e., diversity, richness, and dominant species groups) in estuaries and coastal waters. Changes in essential nutrients concentrations like nitrogen, phosphorus, silicate etc and the ratio of these nutrients have great impacts on controlling phytoplankton species diversity [5, 6]. In tropical estuaries like Mandovi-Zuari, [7] and Cochin estuary [8] nitrogen limits primary productivity. High anthropogenic Phosphorus inputs may also shift ecosystem to N limited [9]. Study of phytoplankton community and controlling factors responsible for changes in community can give the researchers a clue about larger-scale, long-term changes in ecosystem function, including shifts in nutrient cycles, food webs, and fisheries [10]. Apart from elementary Carbon, Nitrogen and Phosphorus are two very important elements for algal growth. Phytoplankton growth flourishes when ratio of nutrients in the ambient water is C: N: P = 166:20:1 [11]. According to the modified Redfield ratio, the optimum dissolved nitrogen and dissolved phosphorus ratio is 20 for phytoplankton but alteration is quite common in this ratio throughout the world oceans and other aquatic bodies due to changes in atmospheric deposition, fertilizer use, waste waters discharge and reduced or increased rate of biologically controlled addition or removal of the nutrients [9]. In-spite of altered redfield ratio many of the phytoplankton species are adapted to that water and form a compact community [12]. But the species composition of community can shift greatly from place to place or time to time in a same place in response to that change [13, 14, 15, 16, 17, 18]. So dissolved inorganic nitrogen and dissolved inorganic phosphate ratio in the water is expected to play a crucial role in phytoplankton growth and community composition. Change in dissolved inorganic nitrogen concentration led to shift of phytoplankton community from one chain forming diatom species to another in the Harima-Nada, eastern Seto Inland Sea, Japan [19]. In estuaries and coastal areas phytoplankton variability is the result of many integrated processes of land, water and lower atmospheric phenomenon. It is important to find out the key factors responsible behind the temporal and spatial changes of community. The controlling factors which play supportive role for phytoplankton growth can be different in estuaries than open ocean and again can be different in different estuaries depending on various physico-chemical and biological interactions. Fresh water runoff enriched with land-derived nitrogen and phosphorus can promote phytoplankton growth in large confined aquatic systems [20]. In coastal oceans upwelling can help in phytoplankton growth and nutrient enrichment and that nutrient may fuel bloom condition of phytoplankton in the estuaries [21]. The bloom condition may arise in the nutrient rich estuaries as a result of thermal stratification [22]. In Estuaries, presence of benthos at bottom sediment support strong benthic pelagic system. As a result of grazing, phytoplankton biomass sink at the bottom of the water body. Phytoplankton resting stages or spores present in sediments after sinking develop into vegetative cells that form seasonal blooms [23]. In a study by Prabhahar et al [24] in Palar river, Tamil Nadu, it was observed that phytoplankton community in 3 different study stations in a same river stretch was different. According to authors the diversity of coastal marine species may have been associated with the allochthonous phytoplankton species from the estuary. The Bay of Bengal and the associated river estuaries drain to coastal area of Bay of Bengal are characterized by dynamic environment [25]. The two study sites of the present investigation represent two different estuarine environments on the basis of natural input and anthropogenic stress. The Saptamukhi estuary has dense mangrove forest along its banks and numerous creeks from the forest ended in the river. The ecosystem is subdivided as forest sub-ecosystem and aquatic sub-ecosystem. Some of the land has been reclaimed for human settlement and agricultural purposes as patches inside the forest [26]. The river banks are periodically inundated and exposed with tide. In general, nutrients rich sediments are characteristic of this ecosystem and nutrient enrichment occurs from plant residues and Litter-fall which has significance impact in p input [27]. The Mahanadi estuary receives effluent from several industries, such as Paradeep Phosphates Ltd. (3264 m3.day^{-1}), Oswal Fertiliser Ltd., Paradeep (4085 m3.day^{-1}) and the East Coast Breweries and Distilleries Ltd., Paradeep (376 m3.day^{-1}) [28]. Both the study sites of present investigation attracted the researchers for decades with their unique ecosystem. Study of Biswas et al [29] in Sunderban mangrove estuaries (study sites with less anthropogenic effects) revealed that the phytoplankton population became 3 fold higher in a decade and 2 diatom species (*Coscinodiscus radiatus* and *Coscinodiscus eccentricus*) dominated consistently from 1990 to 2007 along with increase of N:P molar ratio 10 to 18 from 1990 to 2007. But in the same study site Chowdhury et al [25] did not record any single species bloom or mixed phytoplankton bloom with more or less similar N:P ratio as 2007. In another study by Choudhury et al [30] in anthropogenically influenced Creek and estuary of Sunderban mangrove ecosystem, dominance of diatoms with intermittent appearance of chlorophytes and dinoflagellates with huge nutrient load (nitrate concentration ranged from 31.34 to 65.08μM and ortho-phosphate concentration ranged from 2.05 to 5.17μM) was recorded. According to them, high N:P value restricted the presence of Cyanobacteria in the study sites. In a study along the Mahanadi river stretch by Das sarkar et al [31], they have tried to establish a relation of chlorophyll-a concentration with flow velocity and found that, higher phytoplankton biomass in post monsoon season was observed which coincided with low water velocity. Naik et al [32] concluded from their study that Mahanadi river water phytoplankton community was dominated by diatom followed by Dinophyceae and Cyanophyceae and positive correlation of dissolved oxygen and nutrients with phytoplankton population revealed that DO and nutrients were the main controlling factors. According to Dey et al [9] the presence of *Merismopedia* and *Microcystis* colonies in water column indicated nitrogen deficiency as the two species are non nitrogen- fixing and grow in relatively high phosphate levels. From the other previous studies it is clear that phytoplankton community has a inseparable relation with physical factors, nutrient dynamics and hydrography of the estuarine water and vice versa. So it is important to investigate the dynamics of phytoplankton community in estuarine waters and identify major controlling factor of phytoplankton community to understand the future changes and the vulnerability of the estuarine water to respond to major climatic events. So this study aimed to relate the changes in phytoplankton population in two estuaries with the status of nutrients and other ancillary parameters.

STUDY AREA

The study was conducted from 2015 to 2016. The first study site is located in the Indian Sundarbans (two stations: St. 1. 21°45.22´ N and 88°20.45´ E; St. 2. 21^{0} 45.23´N, 88^{0} 18.48´E, Figure 1a.[25]), which is a part of the Hooghly-Matla estuarine system, on the northeast coast of the Bay of Bengal. This study site is under the influence of dense mangrove forest ecosystem. Tide in the study site 1 is semidiurnal and the tidal amplitude is meso-macrotidal in nature with a range of 2.5–7 m [26].

The second study site (three stations: Stn.1, 20°17´ 38.1” N; 86°42´ 30.3” E, Stn.2, 20°18’29.5” N; 86°41´ 4.4” E and Stn.3, 20°20´ 22.9” N; 86°36´ 50.8” E) is located in the Mahanadi estuary, formed at the mouth of one of the 14 major rivers of India (Figure 1b [9]). The river originates from the Baster Hills and meets the Bay of Bengal near Paradeep. The tide in the estuary is predominantly semidiurnal. The vertical tide range at the mouth varies from 1.45 to 2.20 m. Climate in the region consists of the southwest monsoon (June–September), the northeast monsoon or post-monsoon (October–January), and the pre-monsoon (February–May); 70–80% of annual rainfall occurs during the summer monsoon (southwest monsoon), resulting in high river discharge, which then gradually diminishes during the non-monsoonal months.

MATERIALS AND METHODS

Water sampling was performed from February 2015 to January 2016 in every month covering three seasons, pre-monsoon (February to May), monsoon (June to September) and

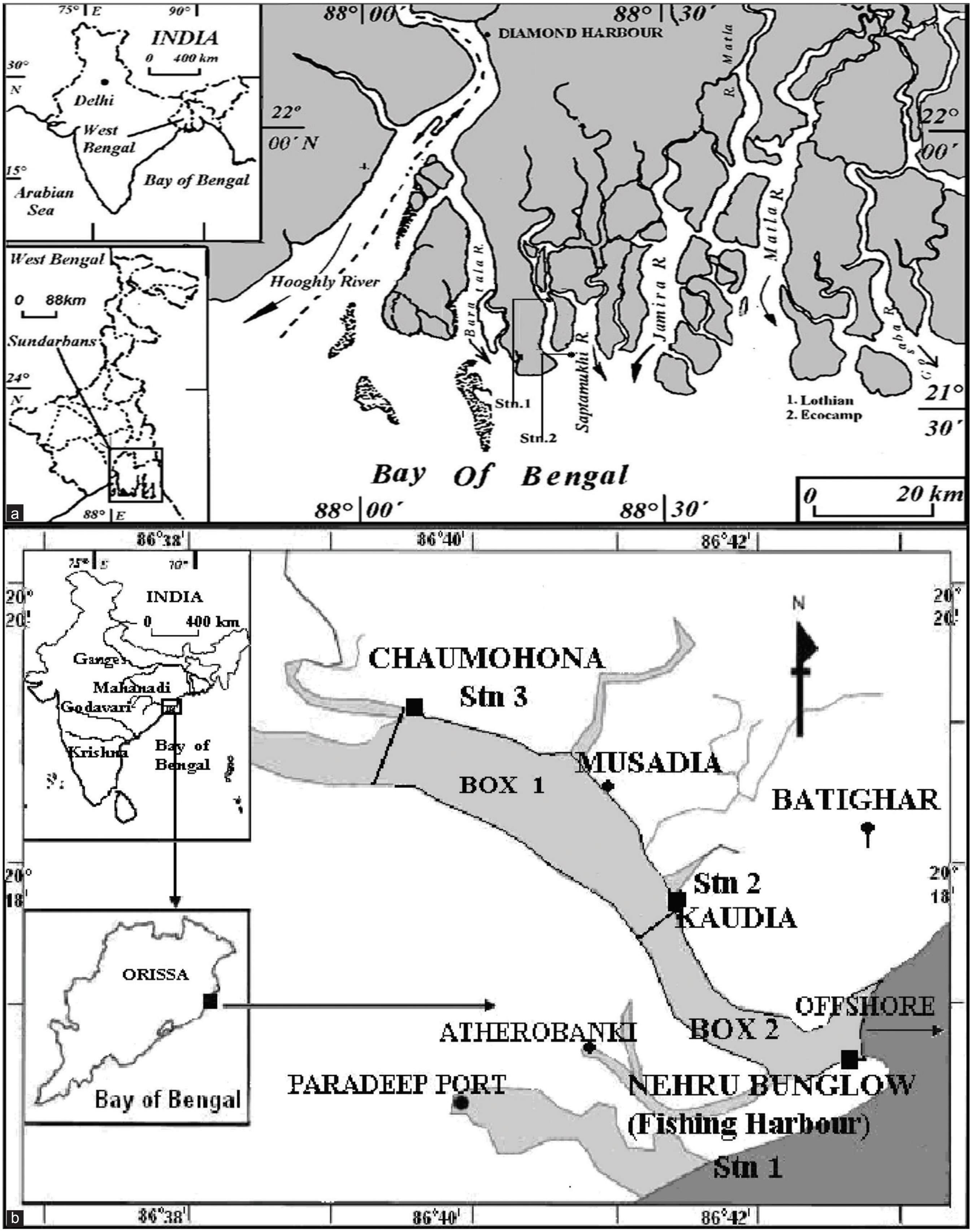

Figure 1: (a) Map of study site 1 in Indian Sundarbans [25], (b) Map of study site 2 in Mahanadi, Paradeep port adjacent area [9]

post-monsoon (October to January). The water samples were collected by using Niskin sampler (Hydro Bios, 436 305, single plastic water Sampler PWS, capacity-5.0 L) Following the sample collection, addition of saturated $HgCl_2$ solution in the field, all samples were transported to a field lab in an ice-box and 1 L of sample was filtered through pre-weighed Millipore filters (GF / F, 0.45 μm) for analysis of the nutrient content. Analyses were completed on the same day of collection of water samples. Air temperatures were recorded using centigrade thermometer. Salinity was measured by Mohr- Knudsen titration method. Transparency of water column was calculated from the secchi disc depth value.

Dissolved inorganic nitrogen present in water available to phytoplankton was as nitrate and ammonia. Nitrate, ammonia, phosphate and silicate in the water samples were measured following standard procedures [33] by using spectrophotometer (UV-VIS Spectrophotometer, 117; Systronics Limited, India, Ahmedabad, Gujrat). N:P molar ratio was derived from dissolved inorganic nitrogen (DIN) and dissolved inorganic phosphate concentration.

Analysis of chlorophyll sample was done by spectrophotometric method according to Strickland & Parson [34].

Phytoplankton samples were collected by net collection and sedimentation process side by side. Net collection was performed by using plankton net made of bolting silk of pore size 20 μm fitted with flowmeter (Hydro Bios, 438110, Hydraulic Pitch =0.3 m per revolution, range of counter = 99999 revolutions, impeller diameter = 75 mm, material = plastic, titanium). Then the total net collection was concentrated into 250 ml and 15ml of formalin (strength=4%) was added for preservation. For sedimentation process of collection, water samples were collected in 1L broad mouth sedimentation tank and preserved with 5 ml of acidified Lugol's iodine (strength=5%) solution. Then water samples were collected in 1L sedimentation tank and were kept undisturbed on the bench top for 5 day [35]. After that, supernatant was siphoned out using a rubber tube and the final sample volume was made to 10ml. The concentrated samples of both net collection and sedimentation collection were analyzed for phytoplankton counts and identification by Sedgwick rafter counting chamber [25]. Measurement of cell size was performed by using calibrated Ocular and stage micrometer under bifocal microscope (Leica 13395H2X, Leica, Buffalo, NY, USA) at 400 X magnification. Randomly selected 30 cells of each species dimension was measured to the nearest ocular unit (1 ocular division = 0.01 mm) [25].

Calculation of % of relative abundance

(R.A.) = n / N [where, n = Total number of individual of species in all samples, N = Total number of individual of all species]

Estimation of dissolved oxygen in estuarine water was based on the Winkler's method [36].

Gross primary production (GPP) and community respiration (R) rates were measured in situ using light and dark bottles following dissolved oxygen method [33]. Water samples collected in BOD bottles and were fixed immediately with Winkler's reagents for estimating initial dissolved oxygen concentration, and remaining bottles were fixed after incubation period. Changes of oxygen were converted to carbon assimilated/respired assuming a photosynthetic quotient of 1.2 and respiratory quotient of 1. GPP and community respiration R were calculated by using formulae 1 and 2

$$\text{Gross primary productivity (GPP) (mg C m}^{-3}\text{ hr}^{-1}) = \frac{12 \times (LB - DB) \times 1000}{32 \text{ x } PQ \text{ x } t} \quad (1)$$

$$\text{And Respiration (mg C m}^{-3}\text{ hr}^{-1}) = \frac{12 \times (IB - DB) \times 1000}{32 \times RQ \times t} \quad (2)$$

Where,

IB = Dissolved oxygen in the initial bottle in mg L^{-1}
DB = Dissolved oxygen in the dark bottle in mg L^{-1}
LB = Dissolved oxygen in the light bottle in mg L^{-1}
PQ = Photosynthetic quotient (1.2)
RQ = Respiration quotient (1.0)
t = Time of incubation in hours

Statistical analyses were done by using MINITAB statistical software (19.2.0).

RESULT

Meteoreology

In Saptamukhi estuary, total rainfall occurred was 1800 mm (Indian Meteorological Department, http://www.imd.Gov.in.) during this study. Air temperature varied annually from 21ºC to 35 ºC. Highest air temperature was recorded during the month of May with a mean value of 34.7 ± 0.28 ºC while the lowest temperature was recorded in the month of January with the average value of 22.4 ± 1.21 ºC . The annual rainfall at Paradeep was 1919 mm (Indian Meteorological Department, http://www.imd.Gov.in.) of which about 83% occurred during the summer monsoon period (June–September). The average air temperature was 30.2 ± 2.39 °C in the pre-monsoon and 24.3 ± 4.2 °C in the post-monsoon. Temperature decreased progressively from May to January and showed lower values in January.

Physico-chemical Parameters

In the Saptamukhi estuarine water, water temperature ranged from 19.6 ºC to 33.5 ºC during the study period. Salinity of estuarine water decreased considerably during the monsoon when huge amount of freshwater flux into the estuary caused considerable dilution of seawater. Lowest salinity (20.3 ± 2.86 psu) was recorded during monsoon period while highest salinity (25.4± 1.40 psu) was recorded during pre-monsoon season when the upstream freshwater flux was minimum. In the Saptamukhi estuarine water, Dissolved oxygen concentration ranged from 5.49 ± 0.73 mg.L^{-1}(Monsoon) to 6.53 mg.L^{-1} (Post monsoon) with the intermediate value of 6.12 ± 0.45 mg.L^{-1} in premonsoon.

Dissolved nitrogen concentration included Nitrate and Ammonia. Ammonia concentration ranged from 4.6 μM to 5.8 μM and Nitrate concentration ranged from 18.8 μM to 24.2 μM in the Saptamukhi estuarine water during the study period. Highest DIN concentration (26.3± 0.04 μM) was recorded during the monsoon. During the post monsoon, its concentration was gradually decreased to 22.9 ± 1.00 μM and in the pre-monsoon the concentration was further decreased to the lowest value of 19.3 ± 0.31 μM. Highest concentration of DIP was observed during the monsoon (1.00 ± 0.07 μM) which was decreased to 0.8 ± 0.03 μM during the post-monsoon. Lowest concentration (0.6 ± 0.01 μM) was recorded during the pre-monsoon. Due to the high river discharge during monsoon season, silicate concentration was higher with a range of value 40.5 to 47.0 μM from June to September. In the post monsoon and pre-monsoon season the concentrations were 33.8 ± 1.63 μM and 31.8 ± 6.60 μM respectively and during monsoon the concentration reached 47.3 ± 6.29 μM .

High transparency (78.6 ± 45.5 cm) was observed during the post-monsoon season when river discharge was minimum. During the monsoon, water showed maximum turbidity (24.7 ± 11.1 cm) due to high river discharge. During the pre-monsoon time, intermediate value of water column transparency was found to be 53.9 ± 24.5 cm.

Water temperature in the Mahanadi estuarine water varied from 22.3 to 31.5 ºC during the study period. Highest surface water temperature was observed in the month of May having the temperature range 29.7 to 31.5 ºC. Lowest water temperature was recorded during the month of January when the temperature varied from 22.3 to 23 ºC. Dissolved oxygen concentration ranged from 4.98 ± 0.05 mg.L^{-1}(Monsoon) to 6.53 ± 0.02 mg.L^{-1} (Post monsoon) with the intermediate value of 5.44 ± 0.05 mg.L^{-1} in premonsoon.

During the monsoon months the salinity decreased from highest value of 24.8 psu (non-monsoonal months) to very low value of 5.0 psu in this estuary. In the month of September, during heavy monsoonal precipitation time, only in the upstream station (st. 3) water, the lowest salinity of less than 1 psu was recorded. Comparatively high salinity was recorded during post-monsoon season (13.5 psu). Highest salinity was recorded during the non-discharge period (pre-monsoon) with a range of 18.0 to 24.8 psu. Dissolved oxygen concentration varied from 5.0 to7.0 mg. L^{-1} during study period. Lowest concentration (5.0 ±0.02 mg. L^{-1}) was recorded in the monsoon months. Highest value was recorded in the post-monsoon months (6.7 ± 0.03 mg. L^{-1}). Comparatively lower values (5.4 ± 0.1 mg. L^{-1}) were observed during pre-monsoon months. DIN concentration was much lower in the estuarine water during post-monsoon (7.6 ± 1.25 μM, Nitrate = 4.12 ± 2.11 μM; Ammonia=3.5 ± 1.56 μM) and pre-monsoon season (6.2 ± 1.78 μM; Nitrate=3.5 ± 0.57 μM; Ammonia= 2.7 ± 1.0 μM) during this study. DIN concentration was generally higher during the monsoon period (30.8 ± 2.32 μM; Nitrate=26.7 ± 1.03 μM; Ammonia= 4.1 ± 0.82 μM). In this estuarine water the DIP concentration was high throughout the year except in the month of May (0.58 to 0.65 μM). In the monsoon period, the DIP concentration was much higher (3.48 ± 1.60 μM) than post monsoon (1.84 ± 0.68 μM) and pre monsoon (0.71 ± 0.66 μM) season. Dissolved silicate concentration was highest during monsoon (137.1±69.6 μM) and become lower during post monsoon (112.3 ± 35.7 μM) reached the lowest value during pre-monsoon (59.4 ± 45.1 μM).

Secchi disc transparency of the water column gradually increased from 23 cm during the monsoon period to 99 cm in the post-monsoon period, and then to 133 cm during the pre-monsoon period.

Primary Productivity and Respiration

In Saptamukhi estuary, gross primary productivity in the photic zone of water column showed seasonal variation with a minimum of 27.1 ± 9.18 mmol C $m^{-2}d^{-1}$ (monsoon), a maximum of 106.0 ± 16.98 mmol C m^{-2} d^{-1} (post monsoon) and intermediate of 75.7 ± 27.7 mmol C m-2 d-1 (pre-monsoon) respectively. Photic zone respiration was higher than the productivity (Pre-monsoon = 164.8 ± 60.63 mmol C m^{-2} d^{-1}; Monsoon = 67.8 ± 21.5 mmol C m^{-2} d^{-1}; Post-monsoon = 113.5 ± 20.10 mmol C m^{-2} d^{-1}). Chlorophyll-a values varied with season with highest value of 11.4 ± 2.4 μg. L^{-1} in post-monsoon season, lowest value of 3.1 ± 1.1 μg. L^{-1} in monsoon season and medium value 4.9 ± 1.2 μg. L^{-1} in pre-monsoon season.

Gross primary productivity in the photic zone of water column was highest in the post-monsoon season in the Mahanadi estuary. The seasonal variation was observed in the GPP values (Pre-monsoon = 108.01 ± 21.02 mmol C m^{-2} d^{-1}; Monsoon = 32.46 ± 12.28 mmol C m^{-2} d^{-1}; Post-monsoon = 184.07 ± 65.93 mmol C m^{-2} d^{-1}). The Chlorophyll-a values followed the seasonal pattern like the Site 1. Highest value of 8.19 ± 2.9 μg. L^{-1} was recorded during post-monsoon season and lowest value of 3.83 ± 0.74 μg. L^{-1} was recorded in monsoon season with intermediate value of 5.1 ± 0.22 μg. L^{-1} during pre-monsoon season.

Comparison of Phytoplankton Community

The composition and total population of phytoplankton for Saptamukhi and Mahanadi estuarine water are given in the Table 1 and Table 2 respectively. Phytoplankton community of Saptamukhi estuary was dominated by diatom with the presence of few dinoflagellate (dinophyceae) species. In this study, in Saptamukhi estuary, total number of microphytoplankton genera was found to be 39 with 7.39 -9.06% abundance of pennate (*Cylindrotheca closterium*,, *Pseudo-nitzschia seriata*, *Gyrosigma acuminatum*, *Rhizosolenia setigera*, *Pinnularia viridis* etc). Out of the centric diatom, dominant species were found to be *Azpeitia biradiata* , *Ditylum brightwellii*, *Melosira granulata*, *Odentella mobilensis*, *Odentella sinensis*, *Denticella regia* etc. Among diatoms, smaller sized genus dominated the population (53.71 – 64.4%) with major relative abundance of

Table 1: Seasonal patterns of phytoplankton average number (No/l), in Saptamukhi estuarine water during study period

SAPTAMUKHI	Pre-Monsoon	Monsoon	Post-Monsoon
Bacillariophyceae	No/l	No/l	No/l
Thalassiosira eccentricus	-	-	435
Coscinodiscus gigas	-	-	152
Coscinodiscus excentricus var. lineatus (Ehrenberg) Cleve	-	-	1179
Azpeitia biradiata	454	278	814
Chaetoceros willei Gran 1897	-	-	825
Chaetoceros bulbosus (Ehrenberg) Heiden 1928	479	362	500
Chetoceros compressum	-	9	250
Chaetoceros pseudocurvisetus Mangin 1910	-	49	150
Chaetoceros decipiens Cleve	-	-	100
Chaetoceros didymus	-	-	179
Cyclotella affinis Grunow	276	515	633
Ditylum brightwellii Grunow	447	607	212
Eucampia zodiacus Ehrenberg	413	-	352
Halosphera minor ostenfeldd	447	347	373
Laudaria annulata	401	135	960
Leptocylindrus danicus	887	-	-
Aulacoseira granulate (Ehrenberg) Simonsen	1275	370	1776
Coscinodiscus blandus A. Schimdt	187	19	745
Coscinodiscus sol C. G. Wallich	164	9	186
Triceratium annulatum Wallich	11	-	1315
Skeletonema costatumn(Greville) Cleve	7458	3312	8924
Surirella ovata Kutzing	2335	702	500
Thalassiosira decipiens	2793	775	-
Podosira subtilis Ostenfeld	-	659	437
Odentella mobilensis Grunow	-	-	250
Odentella sinensis	-	-	250
Denticella regia Schultz	600	400	250
Paralia sulcata (Ehrenberg)Cleve	200	-	250
Asterionellopsis japonica Cleve	500	200	1533
Amphora constricta (Ehrenberg)	350	600	719
Amphipleura pellucida Kutzing	279	-	629
Amphora hyalina f. parvula Grunow	-	-	373
Halamphora coffeaeformis (C. Agardh)	-	-	620
Bacillaria paxillifera (O. F. Muller)T. Marsson	500	18	453
Bellerochea mellitus Brightwell	357	-	774
Cylindrotheca closterium	328	360	586
Pseudo-nitzschia seriata (Cleve) H.Peragallo	301	303	795
Nitzschia habirshawii Febiger ex Cleve & Möller	87	11	31
Parlibellus rhombicus (W.Gregory) E.J.Cox	-	473	-
Gyrosigma acuminatum (Kützing) Rabenhorst	34	90	39
Pinnularia viridis (Nitzsch) Ehrenberg	196	-	-
Proboscia alata (Brightwell) Sundström	-	-	72
Rhizosolenia hebetata f. semispina (Hensen) Gran	93	-	114
Guinardia delicatula (Cleve) Hasle in Hasle & Syvertsen	750	32	106
Rhizosolenia setigera Brightwell	185	66	238
Toxarium hennedyanum (W.Gregory) Pelletan	48	23	54
Thalassionema frauenfeldii (Grunow) Tempère & Peragallo	210	21	199
Thalassiothrix longissima Cleve & Grunow	137	13	179
Dinophyceae			
Tripos fusus (Ehrenberg) F.Gómez	196	60	-
Tripos furca (Ehrenberg) F.Gómez	171	85	-
Tripos muelleri Bory	30	-	-
Dinophysis caudata W.S.Kent	128	59	145
Protoperidinium pentagonum (Gran) Balech	112	-	88
Prorocentrum micans Ehrenberg	-	-	67
Total	23819	10962	29811

the genus *Skeletonema* and *Chaetoceros*. Harrison et al [37] observed phytoplankton community comprising mostly of centric diatom in Indus River delta, while Dham et al. [38] recorded nano-plankton dominated diatom bloom in the Achara River estuary, India.

Abundance of phytoplankton was greater in the post-monsoon than the monsoon and the pre- monsoon seasons in Mahanadi estuary. The highest cell count ($7.21 x 10^5$ cells L^{-1}) was observed during the post-monsoon period when phytoplankton was dominated by two species of *Pediastrum*,

Table 2: Seasonal patterns of phytoplankton average number (No/l) in the Mahanadi estuarine water during study period

MAHANADI	Pre-Monsoon	Monsoon	Post-Monsoon
Bacillariophyceae	**No/l**	**No/l**	**No/l**
Amphiprora constricta	1050	-	600
Amphora hyalina f. parvula Grunow	-	665	1333
Amphipleura pellucida Kutzing	775	202	248
Asterionellopsis japonica Cleve	166	600	30677
Bacillaria paxillifera (O. F. Muller)T. Marsson	-	-	849
Bacteriastrum hyalinum	150	131	901
Odontella mobiliensis Grunow	291	131	461
Odontella sinensis	-	65	843
Denticella regia Schultz	-	329	506
Caloneis bacillum (Grunow) Cleve	-	-	2000
Thalassiosira eccentricus	50	48	3839
Coscinodiscus excentricus var. lineatus (Ehrenberg) Cleve	568	-	1253
Azpeitia biradiata	4108	491	4517
Coscinodiscus gigas	216	65	-
Chaetoceros pseudocurvisetus Mangin	600	111	497
Chaetoceros bulbosus (Ehrenberg) Heiden 1928	300	271	58
Chaetoceros compressum	-	-	2200
Chaetoceros decipiens Cleve	986	632	2859
Chaetoceros lorenzianus Grunow	975	-	2321
Chaetoceros sp.	466	-	1129
Cyclotella affinis Grunow	3204	595	2678
Ditylum brightwellii Grunow	206	395	155
Fragilaria pinnata Ehrenberg	450	71	-
Eucampia zodiacusm Ehrenberg	1650	197	747
Gyrosigma acuminatum (Kützing) Rabenhorst	-	365	143
Hyalodiscus subtilis Bailey	200	-	-
Lauderia annulata	1500	-	-
Melosira granulate	-	575	351
Melosira nummuloides C. agardh	870	942	3400
Cylindrotheca closterium	-	114	-
Navicula pseudocomoides Hendey	175	-	-
Nitzschia brevirostris	-	1371	-
Pseudo-nitzschia australis Frenguelli	-	197	1145
Cylindrotheca closterium	655	636	3844
Nitzschia habirshawii Febiger ex Cleve & Möller	150	432	666
Nitzschia palea	-	1085	-
Nitzschia sp.	500	35	24000
Parlibellus rhombicus (W.Gregory) E.J.Cox	-	1374	481
Planktoniella sol	158	818	87
Plantoniella blanda (A.W.F.Schmidt)Syvertsen&Hasle	-	-	771
Paralia sulcata (Ehrenberg)Cleve	-	800	-
Gyrosigma acuminatum (Kützing) Rabenhorst	100	71	443
Pinnularia viridis (Nitzsch) Ehrenberg	-	35	695
Rhizosolenia stolterfothii H.Peragallo	-	197	-
Rhizosolenia setigera Brightwell	-	658	-
Rhizosolenia hebetata f. semispina (Hensen) Gran	425	30	-
Skeletonema cf. costatum	-	1863	16811
Surirella ovata Kutzing	200	-	3050
Toxarium hennedyanum (W.Gregory) Pelletan	831	544	41736

(Contd...)

Table 2: *Continued*

MAHANADI	Pre-Monsoon	Monsoon	Post-Monsoon
Bacillariophyceae	No/l	No/l	No/l
Thalassionema frauenfeldii (Grunow) Tempère & Peragallo	-	592	860
Thalassiosira sp.	-	114	24548
Triceratium favus Ehrenberg	250	-	-
Thalassiosira hyaline	-	-	2203
Thalassiothrix sp.	800	10	348
Thalassiosira subtilis	-	-	29
Dinophyceae			
Protoperidinium pentagonum (Gran) Balech	150	-	380
Tripos fusus (Ehrenberg) F.Gómez	225	15	75600
Tripos furca (Ehrenberg) F.Gómez	-	-	87
Dinophysis caudata W.S.Kent	-	-	58
Alexandrium minutum Halim	100	-	143
Cyanophyceae			
Anabaena planctonica Brunnthaler	-	-	2000
Microcystis aeruginosa (Kützing) Kützing	600	203	59333
Gloeocapsa aeruginosa Kützing	6000	331	26916
Nodularia chain (with heretocyst)	-	-	9333
Nostoc chain	200	-	55333
Trichodesmium erythraeum Ehrenberg	-	42683	75600
Oscillatoria chain	900	258	68100
Merismopedia colony	-	-	12000
Chlorophyceae			
Actinastrum sp.	-	4800	9333
Ankistrodesmus fusiformis Corda.	-	-	19500
Scenedesmus sp.	-	263	666
Pediastrum simplex	12750	1448	32811
Pediastrum duplex	-	-	40788
Pediastrum young cell	5750	-	-
Spirogyra sp.	100	25	1500
Spirulina sp.	-	35	-
Sphaerocystis colony	-	-	46001
Dictyosphaerium colony	2100	-	-
Total	51900	67918	721764

Trichodesmium erythraeum, two species of Dinophyceae genus *Tripos* and two Cyanophyceae genus *Oscillatoria* and *Nostoc*. Diatoms were most abundant in the post-monsoon period with an overall contribution of some species like *Toxarium hennedyanum*, *Melosira granulata*, *Thalassiosira decipiens* were significant. Contributions of different species of genus *Coscinodiscus* and *Chaetoceros* Were less. The abundance of phytoplankton was low ($0.68x10^5$ cells L^{-1}) during the monsoon period when the genus *Trichodesmium erythraeum* dominated (~ 45%) followed by different species of the genus *Chaetoceros, Skeletonema costatum*, *Parlibellus rhombicus*, and *Pseudo- nitzschia seriata* During the pre-monsoon, the lowest phytoplankton densities (0.52×10^5 cells L^{-1}) was recorded and composition was dominated by *Pediastrum simplex* and *Pediastrum duplex* followed by *Gloeocapsa aeruginosa*. The potentially toxic dinophyte, *Alexandrium minutum* was observed in low densities (0.02-0.19%). Overall, dinophyceae and cyanophyceae population declined from 10.6 to 0.91% and from 49.0 to 18.8%, of the total abundance respectively during the pre-monsoon period, whereas Bacillariophyceae and Chlorophyceae increased from 29.6 to 44.4% and from 18.9 to 39.9%, of the total abundance, respectively, during this season. 57 species of Bacillariophyceae, 6 species of Dinophyceae, 8 species of Cyanophyceae and 9 species of Chlorophyceae were recorded. During the post-monsoon *Pediastrum duplex* was the dominant species of Chlorophyceae group. Dominance of bigger sized Bacillariophycean species was recorded in Mahanadi estuarine water.

The one way ANOVA study of physico-chemical parameters (Table 3) and phytoplankton population showed the variation of N:P ratio and numerical abundance of phytoplankton for both Saptamukhi estuary data and Mahanadi estuary . For the analysis, alpha value was 0.05 and critical F- value was 4.11.

F-value of phytoplankton abundance and N:P ratio in both the estuaries were greater than Critical F-value and p values were lower than alpha value so the null hypothesis was rejected and the analysis of variance was significant.

DISCUSSION

The water and air temperature did not differ much in the two study sites and follow the same seasonal pattern. Lower salinity perhaps triggered the Chlorophycean growth in the Mahanadi estuarine water. In a study by Park *et al* [39], they investigated the growth of chlorophyte Scenedesmus quadricauda in the experiments designed to have the salinity level 0, 3, 6, 12 psu with manipulated N:P ratio of 2, 5, 10, 20 at constant N and P concentrations alternately.. The experimental set up with highest salinity ended up with inhibited growth of the alga. The major finding of the study was the tolerance of salt stress of *Scenedesmus* at highest N : P ratio with highest N by synthesizing intracellular proline. In present investigation, monsoon period was significantly different than other times of the year in terms of nutrients enrichment and turbidity in both the two study sites which brought subsequent changes in other physicochemical characters of estuarine waters. In a study by Biswas et al [26] in 3 distributaries of river Ganges, highest water column transparency was recorded during post-monsoon and out of 10 bloom forming diatom species of those estuarine waters, 7 species bloomed during post-monsoon season. In a study by Naik et al [32] in Mahanadi estuary, inspite of high transparency, high Chlorophyll a concentration was reported during the summer of the year 2006-2007. Monsoonal rain caused high turbidity in the estuarine water and reduced water column transparency. Low transparency limited the light penetration of water column and resulted in reduced gross primary productivity in the water by limiting photosynthesis [40]. Increased nutrient load might fuel the bacterial activity in the water and respiration in water column was increased during monsoon. Instead of being a stratified estuary, Mahanadi estuarine water showed a good oxygen saturation of 78% to 89% throughout the year [9]. Oxygen concentration in both the estuarine waters were above the hypoxia (DO $< 3 mgL^{-1}$, Schimdt et al, [41]). High turbidity probably helped the small celled chain forming species *Skeletonema costatum* to flourish in Saptamukhi estuarine water in monsoon. *Skeletonema costatum* was the 93% of total contributor of phytoplankton cell in the October when the river water nutrients were highest in concentration and turbidity was very high due to upwelling in estuary in Matang Mangrove Forest Reserve (MMFR), Malaysia [42]. In a study by Chu et al [43] it was found that, *Skeletonema costatum* population increased steadily while the total phytoplankton population decreased in a highly turbid estuary of South East Asia. They found the *Skeletonema costatum* population was strongly positively correlated with DIN:DIP, turbidity and DSi:DIN ration than with the nutrients like Nitrate, Phosphate or Silicate separately. Moreover, *Skeletonema costatum* population was negatively correlated with salinity. In Saptamukhi estuarine water, more or less consistent dissolved Silicate concentration throughout the year might have not contributed to the monsoonal increase of *Skeletonema costatum* population. Almost 7 fold increase in dissolved nitrate concentration in Mahanadi estuarine water during monsoon might have fuelled the *Pediastrum* growth. Chu et al [43] found strong positive correlation of *Pediastrum sp* with dissolved nitrate concentration. Stepwise multiple regression with *Pediastrum* sp population with DIN, DIP, dissolved silicate, salinity, water column transparency and DO, DIN was found to be the main factor for variability in the present study. Higher dissolved phosphorus concentration might be the result of factory run-off coming from the Paradeep fertilizer factories [32]. Fluctuation in N:P ratio may change the species dominance in a mixed phytoplankton community. According to Christopher et al [44] decadal variation from 8.2 to 45.0 alter phytoplankton growth and competition among the different species in the community. The line diagram depict the present study result of variation of phytoplankton abundance and N:P molar ratio in two study sites (Figure 2).

Shallow, intertidal estuaries or estuaries having high fresh water flow tend to have strong vertical mixing or short flushing time, though localised bloom can appear in deeper areas with lack of proper vertical mixing [45]. High vertical mixing and short flushing time might be less compared to phytoplankton doubling time or turnover time in estuaries. So in shallow and strong vertically mixed estuaries, effect of high N:P ratio could be overshadowed by short flushing time. That can lead to low phytoplankton population and can resist bloom formation. In some of the estuaries in East Coast of India, removal of phosphate through adsorption along with sedimentation of suspended particle could alter N/P ratios and leave an impact of that on diatom population [29]. In Saptamukhi estuary, during the non-monsoonal months, sediment deposition in the river bed may result in phosphate adsorption and reduction in N:P raito as dissolved inorganic nitrogen concentration did not change much throughout the year. The phosphate may become limiting for phytoplankton growth which resulted into opposite trend with N:P ratio in Saptamukhi estuarine water. On the other hand, effluents received from the phosphate industries help to maintain high phosphate concentration throughout the year in Mahanadi estuarine water. But dissolved nitrogen concentration reduced drastically from monsoon to post-monsoon and GPP decreased gradually in pre-monsoon may indicate the nitrogen limitation [44, 46], as high transparency in water column ensured the light availability for photosynthesis. In a mesocosm experiment in Lake Donghu [47] lower TN: TP ratio was the result of Phytoplankton (*Microcystis* sp) bloom and at the depletion of bloom the ratio became double of previous value. In a study , low TN:TP (Total nitrogen concentration was 5380 $mg.L^{-1}$ and total phosphate concentration was 490 $mg.L^{-1}$) ratio was negatively correlated with phytoplankton biomass and the investigators concluded that nitrogen and phosphorus ration in shallow eutrophic water bodies not always indicate properly the enhancement of limitation of growth of phytoplankton population [12].

In Saptamukhi estuarine water, the phytoplankton community was mostly dominated by Bacillariophyceae probably as a result of dissolved nitrogen availability and a stable N:P ratio near to modified redfield ratio. Investigation of elemental composition of some Bacillariophyceae species (*Thalassiosira*

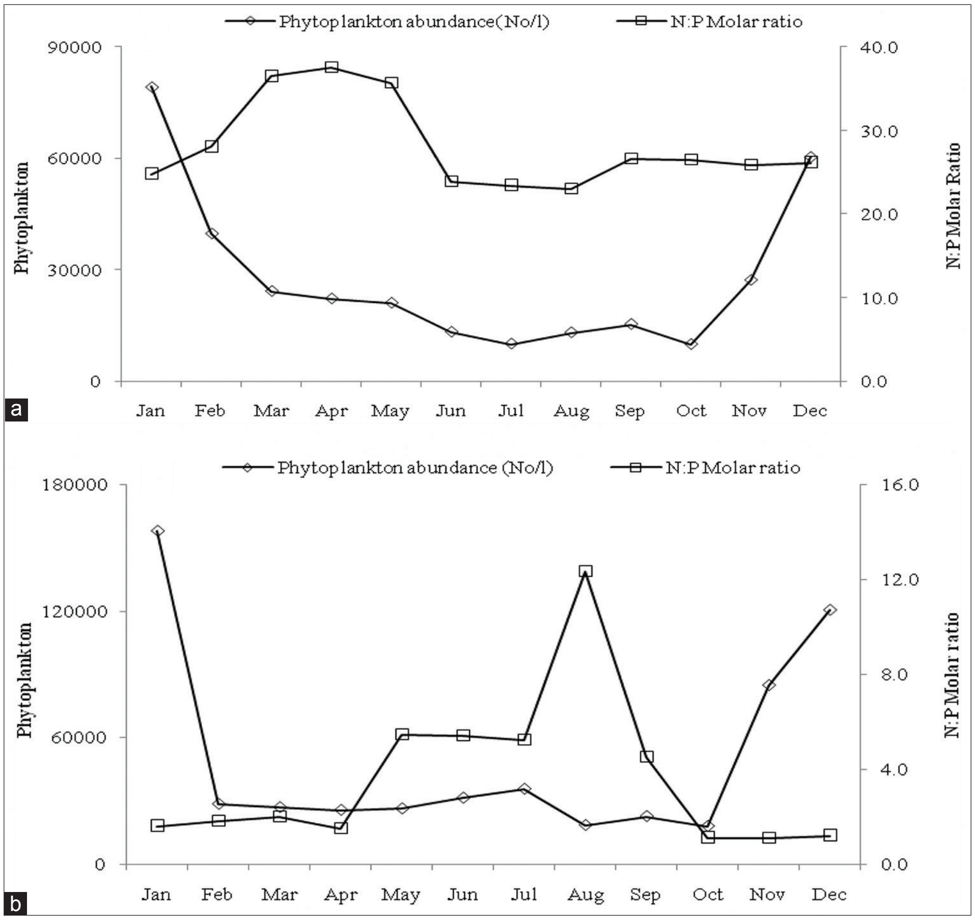

Figure 2: Monthly distribution of average number of Phytoplankton (No/l) and N:P Molar ratio in (a) Saptmukhi estuarine water (b) Mahanadi estuarine water

Table 3: One way ANOVA result of related parameters and phytoplankton numerical abundance in two study sites

SL No	Parameters	Saptamukhi		Mahanadi		N
		F	P	F	P	
1	Rainfall	0.37	0.591	0.52	0.116	36
2	Transparency	0.02	0.889	0.28	0.456	36
3	Temperature	0.3	0.767	0.8	0.743	36
4	Salinity	0.34	0.578	0.67	0.342	36
5	DIN	0.12	0.834	0.25	0.236	36
6	DIP	3.12	0.025	1.7	0.54	36
7	N:P	11.34	0.001	13.45	0.005	36
8	Phytoplankton	7.9	0.004	8.22	0.007	36

sp., *Pseudo-nitzschia* sp etc) revealed that N:P ratio was very close to redfield ratio in a study by Garcia et al [48]. They also observed changes in cellular N:P ratio of Bacillariophyceae changed with temperature but the value ranged between 15 to 30. Lowering of the salinity in monsoon season in Mahanadi estuarine water might cause the presence of fresh-water phytoplankton species which were mostly found in upstream station throughout the year. Among the different study stations, in Ria-Marine Bay system, spain, Chlorophycean species bloom was recorded in the fresh water influenced stations [49]. During peak time of phytoplankton growth i.e. post-monsoon season the Mahanadi estuary showed four-fold greater phytoplankton stock than in the Saptamukhi estuary. Silicate concentration was sufficient in both the estuarine water and supported Bacillariophyceae growth. Previous study found Bacillariophyceae was dominating among phytoplankton when silicate concentration was more than 2 μM[50].

The difference in the N: P ratio in the two estuaries could play vital role in the composition of phytoplankton population in water column. Changes in nutrient supply are often accompanied by alterations in nutrient ratios [51] and as the relative availability of nutrients plays a major role in structuring phytoplankton communities[52], alterations in nutrient ratios can change phytoplankton biomass and species composition especially if there is an increase in N and P availability, but not Si [53]. Availability of nitrogen and phosphorus in the water column played major role for the composition of phytoplankton community. As per the ANOVA result, variation in N:P ratio and phytoplankton abundance for both the estuaries were significant.

CONCLUSION

Difference in N:P ratio might control the composition of phytoplankton community in tropical estuaries though the abundance and occurrence of bloom were the compound result of many other factors which were not considered in this study.

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