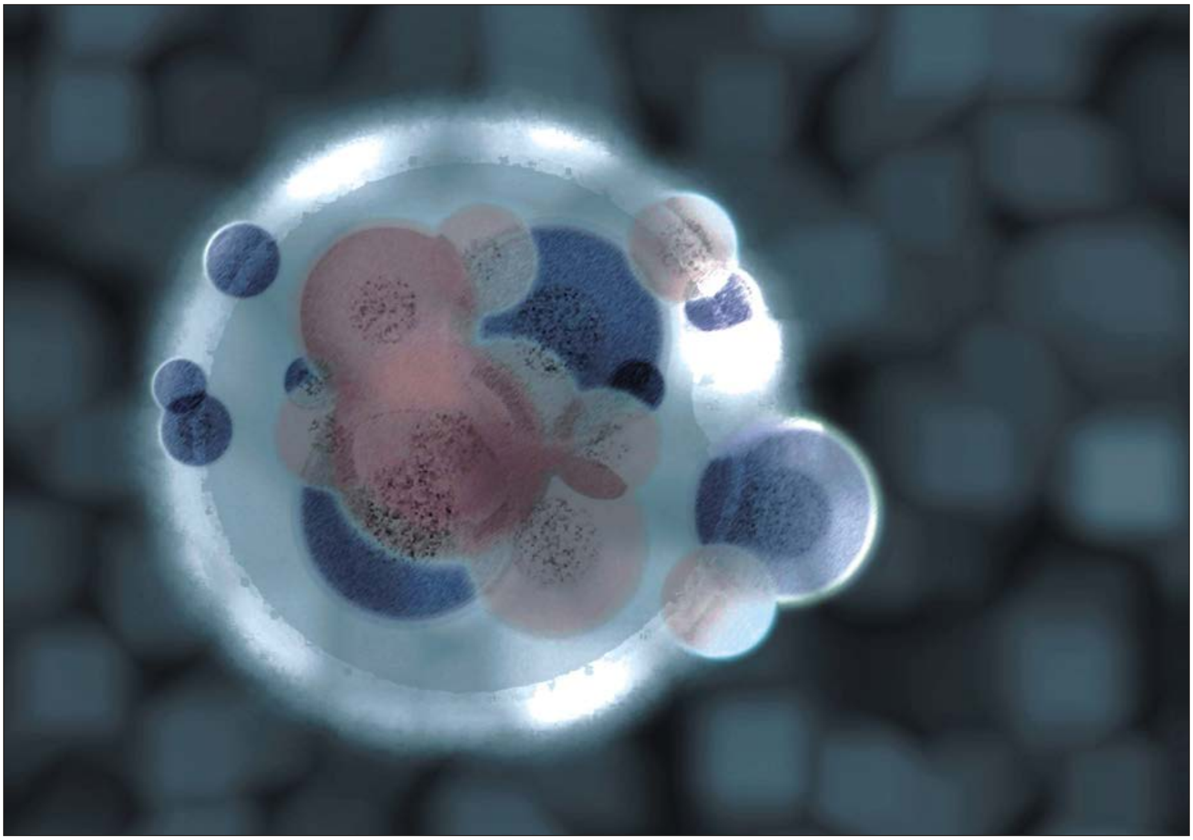

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Composition and Diversity of Phytoplankton from Mangrove Estuaries in Sarawak, Malaysia

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Abstract: The composition and diversity of phytoplankton were studied along with physico-chemical parameters of water of two mangrove-dominated estuaries i.e., Kuala Sibuti (KS) and Kuala Nyalau (KN), Sarawak, Malaysia. A total of 46 species of phytoplankton with the mean density of 147000 cells L^{-1} were recorded from KS estuary i.e., 3 species of Cyanophyceae; 22 species of Bacillariophyceae; 20 species of Dinophyceae and 1 species of Chlorophyceae. The recorded mean density of phytoplankton was 113000 cells L^{-1} with 33 species from 19 genera from KN estuary, in which 19 species were from Bacillariophyceae; 12 species were from Dinophyceae; 1 species was from Cyanophyceae and 1 species was from Chlorophyceae. In both the estuaries, the species composition was found to be in an order of Diatom>Dinoflagellate>Cyanophyceae>Chlorophyceae. Canonical Correspondence Analysis (CCA) revealed that the abundance of Bacillariophyceae and Dinoflagellates was influenced by salinity and conductivity along with ammonium and phosphate while the abundance of Chlorophyceae was influenced by temperature, TDS, DO and pH in KS. The influence of salinity and conductivity along with PO_4 and NH_4 on the abundance of Bacillariophyceae, Dinoflagellates and Chlorophyceae were observed in KN.

Key words: Phytoplankton, composition, diversity, mangrove estuary, Sarawak

INTRODUCTION

In estuarine ecosystems, generally, mangrove habitats are referred to as being nutrient traps. These nutrients support high primary productivity which in turn promote high levels of secondary production (Badarudeen *et al*., 1996). Plankton, particularly phytoplankton plays important role to make such region more productive. Phytoplankton is vital and important organism and act as producer to the primary food supply in any aquatic ecosystem. They are the initial biological components from which the energy is transferred to higher organisms through food chain (Ananthan *et al*., 2004; Tiwari and Chauhan, 2006). Consequently, estuaries support high biomass of secondary consumers and provide economic opportunities in terms of fishery yields.

Data on abundance, distribution and species composition of phytoplankton are essential to know the status of an estuarine ecosystem. Phytoplankton species undergoes spatio-temporal changes in their distribution due to the different effects of hydrographical factors on individual species (Rajkumar *et al*., 2009). They serve as bio-indicators with reference to water quality and also serve as tool for assessing water quality. According to Robertson and Blabber (1992) larval retention and high productivity in mangrove-lined estuaries have generally been attributed to the abundant of planktonic food supply.

The observation on phytoplankton abundance and distribution were conducted in the Gulf of Thailand and east coast of Peninsular Malaysia (Rose, 1926; Boonyapiwat, 1983; Boonyapiwat *et al*., 1984; Suvapepun, 1979; Wongrat, 1982; Piromnim, 1984). However, the study on phytoplankton composition and density in Sarawak coastal and mangrove estuarine environment is scanty. Therefore, observation of phytoplankton abundance, distribution and productivity are essential to assess the overall aquatic health and status in the region. It is expecting that observing phytoplankton status in the estuarine mangrove area of Sarawak is necessary to understand the hydrographic feature and its influence on the organisms around the region. The findings of this study would represent the characteristics of tropical estuary and would help to know the health of mangrove-dominated estuary.

MATERIALS AND METHODS

This study was conducted in the mangrove dominated river estuary in Kuala Sibuti (KS), Miri and Kuala Nyalau (KN), Binutlu, Sarawak, Malaysia and located at 113°23′13.4″E to 3°37′ 2.7″N and

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113°25′13.5"E to 3°25′5"N, respectively in tropical climatic condition. Sampling was done from three stations named stations 1, 2 and 3 from April to May 2013 (Fig. 1a-d). The distance within two stations was approximately 1 km. The station 1 for the both locations was situated near the mouth of river and received neritic water from the adjacent South China Sea with the depth of 3-4 meter. The bank of river of Kuala Nyalau (KN) was lined with *Nypa fruticans* while the river and estuarine area of Kuala Sibuti (KS) was covered by mangrove *Rhizophora apiculata* and *Nypa fruticans*.

Sampling and identification of phytoplankton: *In-situ* data for water pH, temperature, salinity, turbidity, conductivity and Dissolved Oxygen (DO) were detected using water quality meter (WQC-24). For analysis of water nutrients, three surface water samples were collected in clean polyethylene bottles and kept in the ice box and transported to the laboratory. The water samples were filtered using Millipore Filtering System (MFS) and analyzed for dissolved inorganic phosphate, nitrate and ammonia following Parsons *et al.* (1984), Kitamura *et al*. (1982) and Weatherburn (1967), respectively.

Phytoplankton samples collection was carried out from the surface water by horizontal towing of phytoplankton net (0.35 m mouth diameter), made up of bolting silk (mesh size 20 μm) and three replicates were collected. The collected samples were preserved in 5% neutralized formalin for further analysis. Quantitative analysis of phytoplankton was done using settlement method of Sukhanova (1978). Phytoplankton was identified using the standard works of Taylor (1976), Hustedt (1930) and Cupp (1943). Taxonomy of phytoplankton was controlled with http://www.algaebase.org website (Guiry and Dhoncha, 2009). Numerical analysis of phytoplankton was done using sedgewick rafter cell under contrast

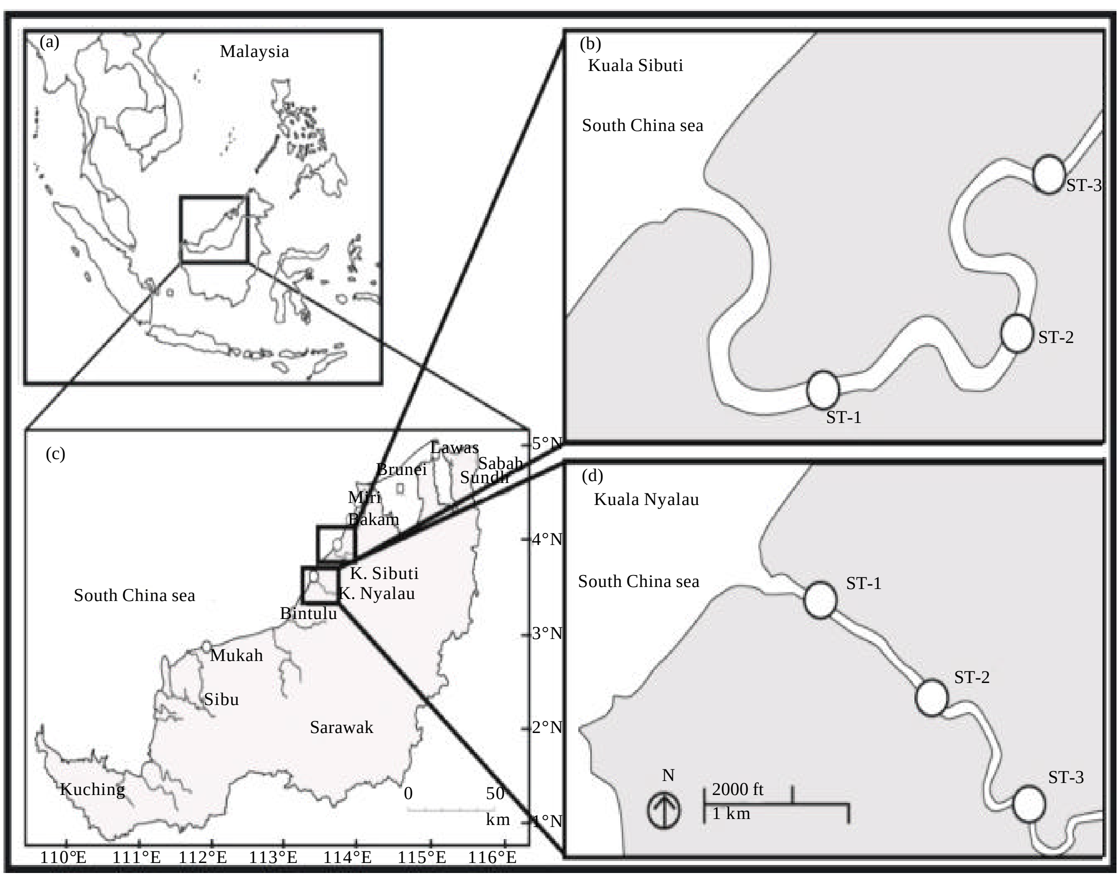

Fig. 1(a-d): Map of study location (a) Sarawak region, (b) Station of Kuala Sibuti, (c) Study locations and (d) Station at Kuala Nyalau in Malaysia compound microscope. Chlorophyll a concentration was estimated following the method described by Coombs and Hall (1982).

Univariate diversity indices such as species diversity (H') and evenness (J') of phytoplankton at different stations of the studied locations were calculated using PRIMER 5 software. Canoco for windows 4.5 was used to observe the distribution of phytoplankton communities in relation to environmental parameters. Simple correlation (r) was calculated for phytoplankton density and physico-chemical parameters using SPSS 10.1 for windows.

RESULTS AND DISCUSSION

Physico-chemical characteristics of estuarine water: The pH in surface water of KS ranged from 5.34 to 7.24 and showed an increasing trend from station 1 to station 3, where the hydrogen ion concentration changed from acidic to light alkaline from river mouth to the upstream (Table 1). In case of KN, the value ranged from 6.02 to 7.15 where the water remained little bit acidic in station 3 and alkaline in station 1 (Table 1). Therefore, water for both the study locations possess mixed characteristics in terms of pH. Generally, fluctuations in pH values can be attributed to factors like removal of CO_2 by photosynthesis through bicarbonate degradation, dilution of seawater by freshwater influx, reduction of salinity and temperature and decomposition of organic matter (Paramasivam and Kannan, 2005). The Dissolved Oxygen (DO) values were found ranging from 2.05 to 3.57 mg L^{-1} in Kuala Sibuti (KS) and highest value was found in station 1 which is in the river mouth and lowest was found in the station 3 which is in the upstream of the river. In case of Kuala Nyalau (KN) the value of DO was higher than that of KS which ranged from 3.93 to 4.45 mg L^{-1}. Salinity at KS showed variations in the ranges of 1.33 to 5.08 PSU, whether in case of KN it showed wider variations ranged from 12.33 to 19.86 PSU (Table 1). The salinity is the main physical parameter that can be attributed to the plankton diversity act as a limiting factor which influences the distribution of plankton community (Sridhar *et al.,* 2006). Salinity was found higher in station 1 for both the locations of KS and KN, it is due to that this station was located at river mouth and influenced by seawater. Salinity gradually decreased at stations 2 and 3. Generally, changes in the salinity of brackish water habitats such as estuaries, backwaters and mangrove are due to the influx of freshwater from land run off, caused by monsoon or by tidal variations. Salinity showed a positive correlation (r^2 = 0.98) with abundance of phytoplankton in KS and the same trend was observed for KN (r^2 = 0.99). Surface water temperature varied from 28.60 to 29.30° C in KS and 27.96 to 29.23° C in KN during the sampling period. Generally, surface water temperature is influenced by the intensity of solar radiation, evaporation and insulation and the low temperature during monsoon could be due to strong sea breeze and cloudy sky (Govindasamy and Kannan, 1996). The total dissolved solids in KS ranged from 2.16 to 9.36 mg L^{-1} and maximum value was recorded in station 2 and lowest in the station 3 where flow of water was low. In KN the total dissolved solid was found higher than that of KS, ranged from 2.16 to 30.56 mg $L^{-1}$and showed the same pattern in KS where lowest value was found in station 3 and highest in station 2 (Table1). Moreover, total dissolved solids also influenced distribution of phytoplankton. Hammer *et al.* (1983) conducted an investigation in Saskatchewan saline lakes, where with a Total Dissolved Solid (TDS) concentration greater than 10 g L^{-1} was typically dominated by chlorophytes in June and/or July and in some cases, diatoms co-dominated during this period. cyanophytes typically dominated lakes with a TDS concentration less than 10 g L^{-1}.

The value of nitrate was found lower at station 1 and higher at station 3 in both the KS and KN. The value of nitrate ranged from 0.34 to 0.68 mg L^{-1} in KS and it was

Table 1: Average values of some physico-chemical parameters, nutrients and chlorophyll *a* in Kuala Sibuti (KS) and Kuala Nyalau (KN) river estuary during the study period

	Station 1		Station 2		Station 3	
Parameters	KS	KN	KS	KN	KS	KN
Water pH	5.340	7.050	6.570	7.150	7.240	6.020
TDS (g L^{-1})	3.600	25.300	9.360	30.560	2.160	12.160
Salinity (PSU)	5.060	19.860	2.360	13.160	1.330	12.330
Temperature (°C)	29.300	29.000	28.760	29.230	28.600	27.960
Turbidity (NTU)	42.200	50.100	55.580	30.330	55.860	79.630
Conductivity (mS m^{-1})	0.930	3.120	0.480	1.480	0.270	0.230
Dissolve oxygen (mg L^{-1})	5.570	4.450	2.980	4.420	2.050	3.930
Nitrate (mg L^{-1})	0.341	0.410	0.675	0.480	0.645	0.990
Phosphate (mg L^{-1}))	0.038	0.062	0.079	0.043	0.041	0.048
Ammonium (mg L^{-1})	1.105	0.984	0.990	0.762	1.002	0.722
Chlorophyll *a* (mg m^{-3})	0.230	0.131	0.115	0.085	0.209	0.183

found ranging from 0.41 to 0.99 mg L^{-1} in KN (Table 1). The possible way of entering nitrate into the estuarine water is through oxidation of ammonia from nitrate to nitrite formation (Rajasegar, 2003). The value for phosphate was also found varying in different stations in KS and ranged from 0.04 to 0.08 mg L^{-1} and the lowest value was found in station 1. In KN, the value of phosphate ranged from 0.04 to 0.06 mg L^{-1} and highest concentration was found in station 1 and the lowest in station 2 (Table 1). High concentration of inorganic phosphate is usually observed during monsoon might possibly be due to intrusion of upwelling seawater into the creek that increased the level of phosphate (Nair *et al*., 1984). Inorganic phosphate showed positive correlation with phytoplankton abundance both for in KS (r^2 = 0.028) and KN (r^2 = 0.435).Concentration of ammonium was found higher in station 1 for both the locations i.e., KS (1.11 mg L^{-1}) and KN (0.98 mg L^{-1}) followed by station 2 (0.99 mg L^{-1}) and 3 (0.76 mg L^{-1}) in KN and station 2 (1.00 mg L^{-1}) and station 3 (0.18 mg L^{-1}) for KS. Recorded higher concentration of ammonium could be partially due to the death and subsequent decomposition of phytoplankton or other detritus matter. Nutrients are considered as one of the most important parameters in the estuarine environment influencing growth, reproduction and metabolic activities of living organisms. Distributional pattern of nutrient depends on the surface runoff and seasonal fluctuation. Since the estuary transitions from nearly oceanic conditions at the mouth to freshwater at its extreme upstream limit, nutrient limitations also show a spatial trend with phosphorus more limiting at the freshwater extreme and nitrogen and silica more limiting near the mouth (Glibert *et al*., 1995). Nutrient concentration and availability are obvious factors controlling phytoplankton biomass (Ferguson *et al.,* 2004; Jacquet *et al*., 2006) particularly in estuaries.

Chlorophyll *a* concentration in water indicates the biomass of phytoplankton. The concentration of Chlorophyll a was found ranging from 0.12- 0.23 mg m^{-3} (Table 1) in KS and highest value was found in station 1. For KN, the concentration of chlorophyll a ranged from 0.085-0.18 mg m^{-3} and the higher concentration was found in station 3 which was in the upstream. The value of concentration of Chlorophyll *a* may be influenced by anthropogenic effects and evidenced by its positive correlation with salinity and may also be due to the fresh water discharges from the rivers, causing turbidity and less availability of light (Kawabata *et al*., 1993; Rajasekar *et al*., 2005). The concentration of Chlorophyll *a* did not show strong correlation with abundance of phytoplankton in both the locations of KN (r^2 = 0.473) and KS (r^2 = 0.122).

Composition and abundance of phytoplankton: A total of 46 species, comprising of 25 genera under 3 divisions i.e., Cyanophyta (Blue-green algae), Diatom (Bacillariophyceae), Dinoflagellate (Dinophyceae) and Chlorophyta (Chlorophyceae) were recorded from KS river estuary (Table 2). Out of 46 species, 22 taxa were from Diatoms; 20 taxa were from Dinoflagellate; 3 taxa were from Cyanophyta and 1 taxon was from Chlorophyta. Thirty three (33) taxa of phytoplankton from 19 genera were recorded from KN river estuary which comprised of 19 taxa from Bacillariophyceae (Diatom); 12 taxa from Dinophyceae (Dinoflagellate); 1 taxa from Cyanophyceae (Cyanophyta) and 1 taxa from Chlorophyceae (Chlorophyta). Diatom was the dominating group of phytoplankton in both the sampling location at KS and KN followed by Dinoflagellates, Cyanophyta and Chlorophyta (Fig. 2a, b). Generally, the distribution and abundance of phytoplankton in tropical waters varied remarkably due to the environmental fluctuations during season and these variations are well pronounced in the sheltered costal systems like mangroves (Rajkumar *et al*., 2009). The present findings agreed with the above-mentioned statement.The percent composition of each group of phytoplankton was found in the following order Diatom>Dinoflagellate>Cyanophyceae >Chlorophyceae. The dominance of diatoms in estuarine and marine aquatic environment was also agreed by Boonyapiwat (1997), Sevindik (2010) and Al-Hashmi *et al*. (2013).

The higher composition and diversity of phytoplankton were found in station 1 for both the locations. Generally, diatoms were found dominant in station 1 followed by stations 2 and 3. It is due to the fact that station 1 was situated at the mouth of the estuary where wide range of salinity was probably suitable for diatoms. In KS, the dominated 6 diatoms were *Rhizosolenia styliformes, Rhizosolenia alata, Rhizosolenia curvata, Rhizosolenia polydactyla, Rhizosolenia clevei* and *Rhizosolenia imbricata* followed by 4 species of *Coscinodiscus* comprising *Coscinodiscus* sp.*, Coscinodiscus centralis, Coscinodiscus thorii* and *Coscinodiscus granii.* In KN, *Coscinodiscus* was the dominant group of phytoplankton comprising *Coscinodiscus* sp., *Coscinodiscus centralis*, *Coscinodiscus thorii*, *Coscinodiscus jonesianus* and *Coscinodiscus granii.*

Dinoflagellate was dominated by genus *Ceratium* in both the locations and recorded species were *Ceratium carriense*, *Ceratium horridum*, *Ceratium paradoxides*, *Ceratium fusus*, *Ceratium furca* and *Ceratium tripos.* The single species of Dinoflagellate entitled *Ceratium lineatum* was found dominated (26000 cells L^{-1})

Table 2: List of phytoplankton found in Kuala Sibuti (KS) and Kuala Nyalau (KN) mangrove estuary during the study period

Taxa	Kuala Sibuti	Kuala Nyalau
Cyanophyceae (Blue-green algae)		
Oscillatoria sp.	+	-
Oscillatoria erythraea	+	-
Anabena sp.	+	-
Spirogyra sp.	-	+
Diatoms (Bacillariophyceae)		
Actinoptychus senarius (Ehrenbarg) Ehrenbarg	-	+
Bacteriastrum comosum J. Pavillard	-	+
Coscinodiscus sp.	+	+
Coscinodiscus centralis Ehrenbarg	+	+
Coscinodiscus thorii	+	+
Coscinodiscus granii Gough	+	+
Coscinodiscus jonesianus (Greville) Ostenfeld	-	+
Chaetoceorspseudo curvisetus Manginn	+	-
Chaetoceors affinis Lauder	+	-
Ditylum brightwelli (West) Grunow	-	+
Eucampia zodiacus Ehrenberg	+	-
Hyalodiscus sp.	+	+
Nitzschia sp.	+	+
Nitzschia sigma (Kutzing) W. Smith	+	-
Navicula sp.	-	+
Odontella sp.	-	+
Pleurosigma elongatum W. Smith	+	-
Pleurosigma normanii Ralfs	+	+
Rhizosolenia styliformes Brightwell	+	-
Rhizosolenia alata (Cleve) Grunow	+	+
Rhizosolenia curvata Zacharias	+	+
Rhizosolenia polydactyla f.squamosa Sundstrom	+	+
Rhizosolenia clevei Ostenfeld	+	+
Rhizosolenia imbrcata Brightwell	+	-
Skeletonema costatum (Greville) Cleve	+	-
Thalassiothrix longissima Cleve and Grunow	+	-
Thalassiothrix sp.	+	+
Thalassionema frauenfeldi (Grunow) Hallegraeff	+	-
Thalassionema sp.	-	+
Dinoflagellates (Dynophyceae)		
Amphisolenia bidentata Schroder	+	-
Ceratium furca (Ehrenberg) Claparede and Lachmann	+	+
Ceratium carriense (Cleve) Jorgesen	+	+
Ceratium lineatum (Ehrenberg) Cleve	+	+
Ceratium fusus(Ehrenberg) Dujardin	+	+
Ceratium paradoxides Cleve	+	+
Ceratium tripos (O.F. Muller) Nitzsch	-	+
Ceratium horridum (Cleve) Gran	-	+
Dinophysis sp.	+	-
Dinophysis caudata Saville-Kent	+	-
Dinophysis miles Cleve	+	+
Protoperidinium divergents (Ehrenberg) Balech	+	-
Protoperidimium oceanicum (VnanHoffen) Balech	+	-
Protoperidinium sp.	+	-
Protoperidinium depressum (Bailey) Balech	+	-
Prorocentrum gracile Schutt	+	-
Gonyaulux sp.	+	-
Gonyaulux spinifera Clparede and Lacchmann	+	-
Phalacroma sp.	+	-
Pyrophacus sp.	+	+
Pyrocystis sp.	-	+
Triceratium sp.	+	+
Triceratium favus	+	+
Chlorophyceae		
Closterium sp.	+	-
Chlorella sp.	-	+

+: Present, -: Absent

in KS while *Dinophysis caudata* in KN (28000 cells L^{-1}). The phytoplankton species, *Ceratium lineatum* is known to be toxic, particularly when it forms in large number and called red tide. Moreover, studies revealed that

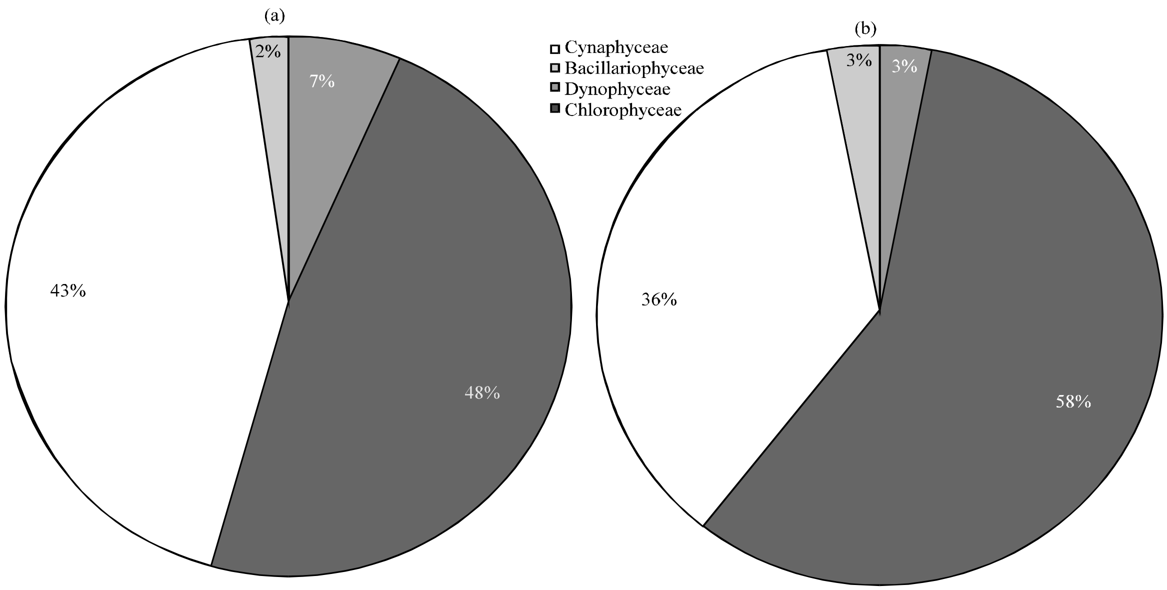

Fig. 2 (a-b): Percentage distribution of phytoplankton in (a) Kuala Sibuti (KS) and (b) Kuala Nyalau (KN) river estuary

dinoflagellates are usually dominated in the aquatic systems with high amounts of humic and fulvic acid (Prakash and Rashid, 1968) and degraded mangrove forests are the great source of those compounds.

Phytoplankton density was observed higher in station 1 followed by stations 2 and 3 for both the locations KS and KN. Comparatively, highest cell density was found in station 1 for both the locations (221000 cells L^{-1} for KS and 162000 cells L^{-1} for KN). The most abundant species was *Ceraium lineatum* (26000 cells L^{-1}) and *Dinophysis caudata* (28000 cells L^{-1}) for KS and KN, respectively. The range of phytoplankton density for stations 2 and 3 were 126000-133000 cells L^{-1} and 51000-88000 cells L^{-1} for both the locations, respectively. The present values showed lower abundance when compared with the studies of Perumal *et al.* (2009), Boonyapiwat *et al.* (1984) and Rajkumar *et al.* (2009), probably due to the short duration of study. In this regard, further long term investigation may record the higher abundance of phytoplankton in these locations. Studies by Boonyapiwat (1997) recorded 214-33520 cells L^{-1} and 178-14223 cells L^{-1} of phytoplankton in the coasts of Gulf of Thailand and east coast of Peninsular Malaysia. These values are lower than the findings of the present study, probably may be influenced by variation of mesh size and different geographical locations. However, another study by Boonyapiwat *et al.* (1984) was also recorded lower (2800-4380 cells L^{-1}) density of phytoplankton using 80 μm mesh size of net.

Canonical Correspondence Analysis (CCA) was applied to explore the phytoplankton communities' distribution in relation to environmental parameters. The statistical significance of the CCA was determined using permutation and level of significance was set at $p<0.05$. Occurrence frequency of the group more than 33% is shown in the plot (Fig. 3a, b). For the case of KN, among the variables included salinity, temperature, pH and nutrients viz., phosphate and ammonium was found strongly related to distribution of phytoplankton (Fig. 3a). In KS, the distribution of the groups of phytoplankton was found mainly influenced by conductivity, salinity and DO and the influence was high in station 1 (Fig. 3b).

Univariate diversity indices including species diversity (H') and evenness (J') were calculated which showed fluctuation in different stations. The ranges of species diversity (H') in KS and KN at stations 1, 2 and 3 were 3.45-3.60 and 2.9-3.27, respectively (Fig. 4a). The ranges of species evenness in KS and KN at station 1, 2 and 3 were 0.92-0.98 and 0.84-0.98, respectively (Fig. 4b). Station 2 showed highest species diversity followed by stations 3 and 1, though highest species abundance was observed in station 1 in both the locations KS and KN. Species diversity and evenness were found higher in KS than that of KN. The findings indicate good species diversity of phytoplankton in the study locations.

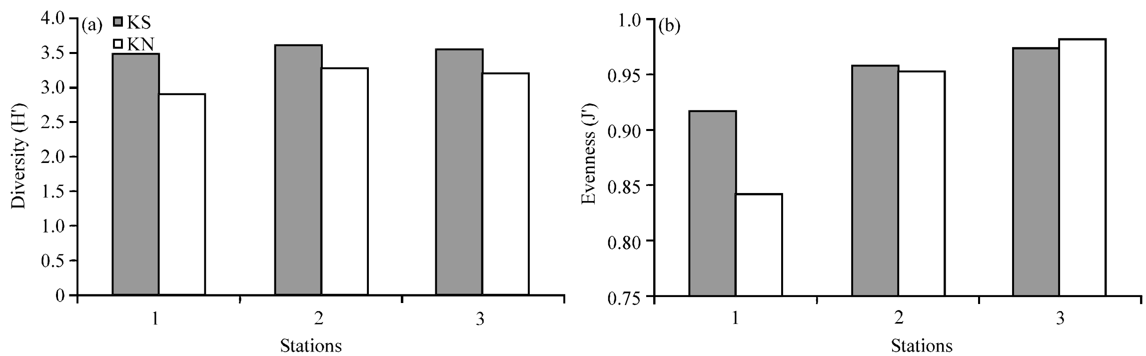

Fig. 3(a-b): Species (a) Diversity and (b) Evenness of phytoplankton at different stations in Kuala Sibuti (KS) and Kuala Nyalau (KN)

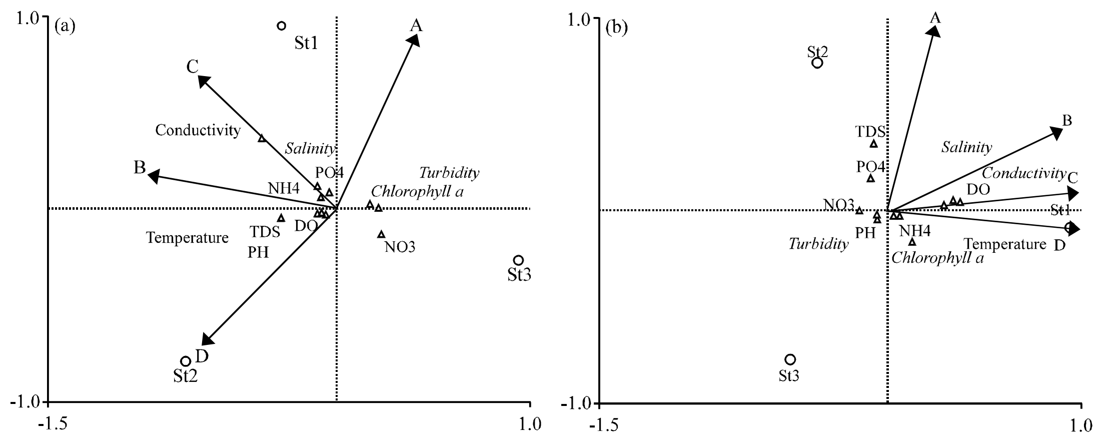

Fig. 4(a-b): Canonical correspondence ordination of the phytoplankton samples and physico-chemical parameters collected at 3 different stations and related environmental parameters at KS. Biplots of the groups (Occurrence frequency 33%) and the environmental parameters, A: Cyanophyceae, B: Bacillariophyceae, C: Dinoflagellates and D: Chlorophyceae

Species diversity and evenness in Kaduviyar estuary in India was found ranging from 2.55-3.46 and 0.82-0.96, respectively (Perumal *et al*., 2009). The evenness index of a bar-built estuary (Thengapattanam estuary) along west coast ranged from 1.4-2.96 (Vareethiah and Haniffa, 1997); 0-0.93 and 0-0.98 for Vellar estuary (Chandran and Ramamoorthi, 1984); 0.65 to 0.92 at Hugli estuary (De *et al*., 1994). A close relationship between diversity and evenness had been reported by DeJong (1975) and Eloranta (1976).

CONCLUSION

Studied mangrove estuaries are located in tropical region and mostly characterized by wet and dry season. Both the estuaries are enriched with variety of mangrove species where KS was dominated by mangrove *Rhizophor aapiculata* and *Nypa fruticans* while *Nypa fruticans* was the dominant species for KN. This study revealed that *Rhizophora* and *Nypa* dominated estuary (KS) shows more species composition (46 species) than that of single stand of *Nypa* dominated estuary, KN (33 species). Diatoms were found most dominating group of phytoplankton in both the locations. Some common phytoplankton species were found in both the estuaries though there was difference in physico-chemical characteristics of water. The abundance and diversity of phytoplankton were influenced by water quality parameters. However, the observed phytoplankton may have the ability to thrive in different environmental condition in these tropical mangrove ecosystems.

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