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Multivariate Analysis of Phytoplankton in Relation to Physicochemical Parameters Disparity in Parangipettai Waters, Southeast Coast of India

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ABSTRACT
Phytoplankton are the major source of primary production in water bodies. But, due to increased anthropogenic pressures in the coastal waters, there is a gradual decrease in the population of this biological resource. In the present study, multivariate statistical methods including principal component and cluster analysis were applied to surface water quality data sets obtained from Vellar Estuary, Parangipettai, Southeast coast of India. Studies on physicochemical and biological parameters were made during August 2008 to July 2009. Totally 45 species representing different classes namely Bacillariophyceae (36); Dinophyceae (6); Chlorophyceae (1) and Cyanophyceae (2) were recorded. Among the four classes, Bacillariophyceae appeared as the dominant group in terms of total species and cell numbers. The multivariate statistical analysis revealed that the surface water quality and phytoplankton density of the Parangipettai waters and its tributaries is being affected by increased aquaculture practices, agriculture discharges and high level of organic load. These data on the phytoplankton distribution and abundance with reference to hydrography will be of immense help for further ecological assessment in Parangipettai waters.

Key words: Phytoplankton, physicochemical parameters, multivariate analysis, factor analysis, principal component analysis

INTRODUCTION
Estuaries are highly productive ecosystem enriched with nutrients and are known to be extremely abundant in biological resources especially phytoplankton. Compared to marine and freshwater ecosystems, these ecosystems are environmentally unstable (Mclusky and Elliott, 2004). Anthropogenic activities negatively influence the water quality and aquatic ecosystem functions and have a great pressure on these ecosystems, leading to the decrease in water quality and biodiversity, loss of critical habitats and ultimately the quality of life of local inhabitants is affected (Herrera-Silveira and Morales-Ojeda, 2009). Continuous water quality measurements and analyses are necessary for effective management of coastal water quality. Phytoplankton are also known as the "Pastures of sea" which are the major source of primary production in water bodies and it is responsible for one quarter of the world's plant photosynthesis (Mishke *et al.*, 1970). Since, phytoplankton communities are very sensitive to environmental changes these species are used as indicators of water quality (Reynolds, 1997; Reynolds *et al.*, 2002; Brettum and Andersen, 2005).

Phytoplankton is the major biological component of food chain through which the energy is transferred to the higher organisms (Rajesh *et al*., 2002; Ananthan *et al*., 2004; Tas and Gonulol, 2007; Shekhar *et al*., 2008). Biomass and production of phytoplankton are important in regulating the diversity of organisms at higher tropic levels (Kathiresan, 2000). 33-51% of total chlorophyll-a is contributed by the phytoplankton of 5-10 μm size and 20-22% of total gross production in mangrove waters (Kawabata *et al*., 1993). Due to the differential effect of hydrographical factors on individual species, phytoplankton species undergoes spatial-temporal changes in their distribution. They serve as bio-indicators with reference to water quality and thus serve as a tool for assessing the health of the aquatic ecosystems. Further, the measurement of primary productivity of aquatic ecosystem is required to forecast fishery potential of an area (Rajkumar *et al*., 2009).

Due to varied physical and chemical requirements for their population and growth, seasonal variation in phytoplankton abundance, physicochemical parameters and species composition has been studied in Indian coastal waters. (Rajkumar *et al*., 2009; Perumal *et al*., 1999, 2009; Saravanakumar *et al*., 2008; Mathivanan *et al*., 2007; Sridhar *et al*., 2006; Tiwari and Chauhan, 2006; Rajasekar *et al*., 2005; Madhav and Kondalarao, 2004; Rajasegar *et al*., 2000). However, water monitoring management of a long-term period and many sampling sites produces large and complicated data sets consisting of all kinds of water parameters which are difficult to analyze, interpret and extract comprehensive information from them.

In recent years, multivariate statistical analysis, such as Cluster Analysis (CA), Principal Component Analysis (PCA), Discriminant Analysis (DA) and Factor Analysis (FA) have been successfully employed to evaluate the chronological and spatial character of coastal water quality and river water (Kuppusamy and Giridhar, 2006; Wu and Wang, 2007; Chau and Muttil, 2007; Singh *et al*., 2005; Chen and Mynett, 2006; Alkarkhi *et al*., 2009). PCA and CA potentially allow us to simplify the description of observations by finding the structure or patterns in the presence of bewildering data (Ragno *et al*., 2007). In the present study we report the statistical analysis of water quality parameters and phytoplankton diversity in three sites of Vellar estuary (sea mouth, mangrove and interior river). The analysis was carried out to explore the extent of resemblance among the sampling sites, to identify the variables responsible for spatial variations in estuaries water quality and to quantify the influence of possible natural and anthropogenic sources on the water parameters on the three selected sites of the estuary and Bay of Bengal Sea.

MATERIALS AND METHODS

Study area: The river Vellar situated in the southeast coast of India (11°29'50"N and 79°46'24"E) originates in the Shervaryan Hills of Salem District. After meandering through a distance of 480 km, it forms the estuarine system (true estuary) before it merges with the Bay of Bengal at Parangipettai coast. The estuary is subjected to semi-diurnal tides with maximum tidal amplitude of about 1 m. The estuary is about 600 m wide at its mouth (Fig. 1). There is a perfect exchange of both biotic and abiotic variations due the influence of neritic water with estuarine environment. Average depth of the estuary is 2.5 m and the maximum depth at high tide is 5.3 m. There were as many as 42 shrimp farms with water spread area of 150 ha in the surrounding of Vellar (Rajasegar *et al*., 2002). In order to evaluate the water quality and phytoplankton diversity in the estuary, the surveys were conducted during the August 2008 to July 2009 and sites were as follows:

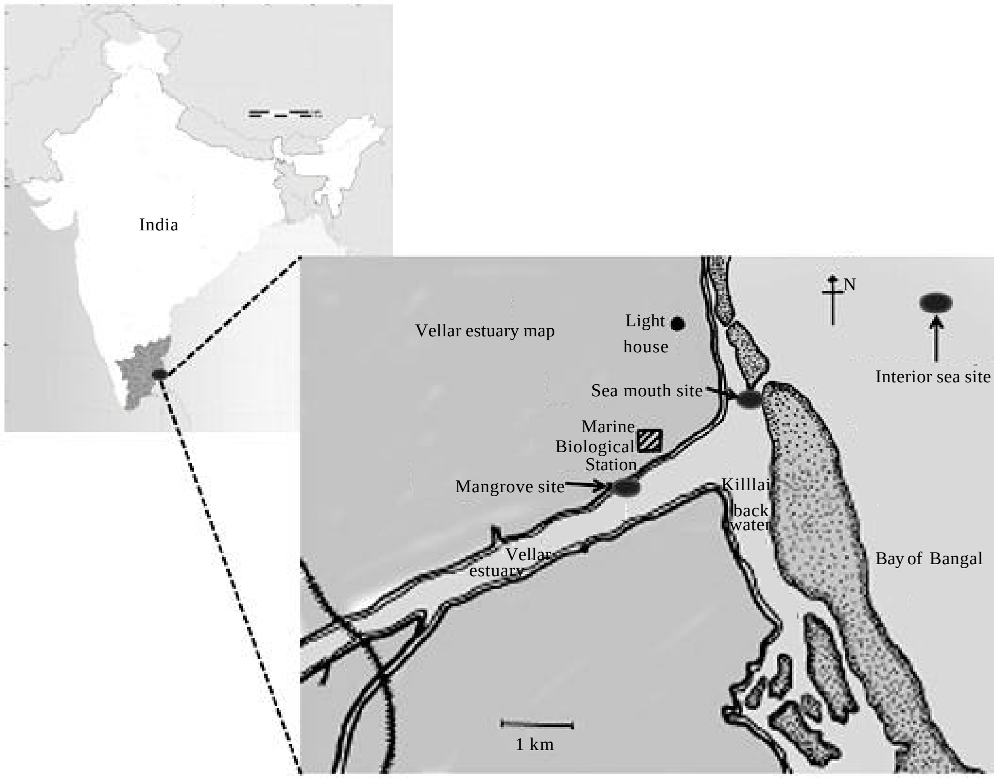

Fig. 1: Map showing the study area

Site 1: Monitoring stations located in interior of the Bay of Bengal Sea

Site 2: Located in Vellar estuary river mouth. The hydrological environment can elucidate important information about water exchange between Vellar estuary and Bay of Bengal Sea

Site 3: Near the mangrove ecosystem of the Vellar estuary which is an aquaculture practice area

Water quality analysis: Physicochemical parameters were directly analyzed at the collection sites. Temperature was measured using a standard Celsius Thermometer. Salinity was estimated with the help of a hand-held Refractometer (ATAGO, Japan). pH was measured by using an ELICO Grip pH meter. Dissolved Oxygen was estimated by the modified Winkler's method and Chlorophyll-a (90% acetone method) measurement were carried out spectrophotometrically (Strickland and Parsons, 1972) and was expressed as mg L^{-1}. Surface water samples were collected in clean polyethylene bottles for the analysis of nutrients which were kept immediately in an ice box and then transported to the laboratory. The collected water samples were filtered by using a Millipore filtering system and then analyzed for dissolved inorganic nitrate, nitrite, ammonia, reactive silicate, inorganic phosphate, by following the standard procedures described by (Strickland and Parsons, 1972) and were expressed in μg L^{-1}. Multivariate statistical methods have been widely applied to investigate environmental phenomena in recent years (Laaksoharju *et al*., 1999; Guler and Thyne, 2004; Anazawa and Ohmori, 2005).

Phytoplankton analysis: Plankton samples were collected by using plankton net (mesh size: 40 μm) at the surface layers and immediately fixed in 5% formaldehyde solution. For the quantitative analysis of phytoplankton, the settlement method described by (Sukhanova, 1978) was adopted. Numerical plankton analysis was carried out using Utermohl's inverted microscope. Phytoplankton was identified using the standard works of (Venkataraman, 1939; Subrahmanyan, 1946; Prescott, 1954; Steidinger and Williams, 1970). The analyses were carried out at CAS in Marine Biology, Annamalai University, India. Biodiversity indices were calculated following the standard formulae of Shannon and Weaver (1949), Gleason (1922) and Pielou (1966).

Multivariate statistical analyses

Visual data mining using box plots: Box plots provide an excellent visual summary of a set of data and are especially useful when comparing two or more sets of data. Visual data mining refers to the visual presentation of data to extract useful information which allows users to summarize, extract and grasp more complex patterns and results than mathematical or text type descriptions of the same. For the spatial and temporal analysis of physicochemical parameters and ANOVA (Analysis of Variance), statistical package SPSS (version 16.0) were used, respectively. In a box plot, the line across the box represents the median, the bottom of the box is at the first quartile (Q) and the top is at the third quartile (Q31). The whiskers are the lines that extend from the bottom and top of the box to the lowest and highest observations inside the range defined by a lower.

Cluster analysis (CA): Cluster analysis was done in the statistical software package Primer 6.1. CA is a group of multivariate techniques to assemble objects based on the characteristics they possess and it classifies objects in such a way that each object is similar to the others in the cluster with respect to a predetermined selection criterion (Shrestha and Kazama, 2007; Iscen *et al*., 2008). Hierarchical agglomerative clustering is the most common approach and provides intuitive similarity relationships between any one sample and the entire data set; it is typically illustrated by a dendrogram (tree diagram). The dendrogram provides a visual summary of the clustering processes, presenting a picture of the groups and their proximity, with a dramatic reduction in the dimensionality of the original data (Alberto *et al*., 2001).

Principal component analysis (PCA) and factor analysis (FA): PCA and FA were analyzed using XLSTAT Version 2012.1.01. PCA is designed to transform the original variables into new, uncorrelated variables (axes), called the principal components which are linear combinations of the original variables. PCA and Factor Analysis (FA) are both variable reduction techniques. The FA suggests important variates to explain the observed variances in the data (Shrestha and Kazama, 2007). It reduces the dimensionality of the data set by explaining the correlation amongst a large number of variables in terms of a smaller number of underlying factors Principal Components (PCs), without losing much information (Vega *et al*., 1998; Alberto *et al*., 2001; Helena *et al*., 2000).

RESULTS

Box plots analysis for water quality parameters: The box plots for the temporal variations of water quality parameters namely rainfall, surface water temperature, salinity, pH and dissolved oxygen are presented in Fig. 2. In all the sites total rainfall recorded was 2042 mm from August 2008 to July 2009, varying from 20-969 mm. The surface water temperature ranged between 25.8-33.3°C. The salinity was in the range of 8-35%. pH ranged from 7.8-8.2. Dissolved

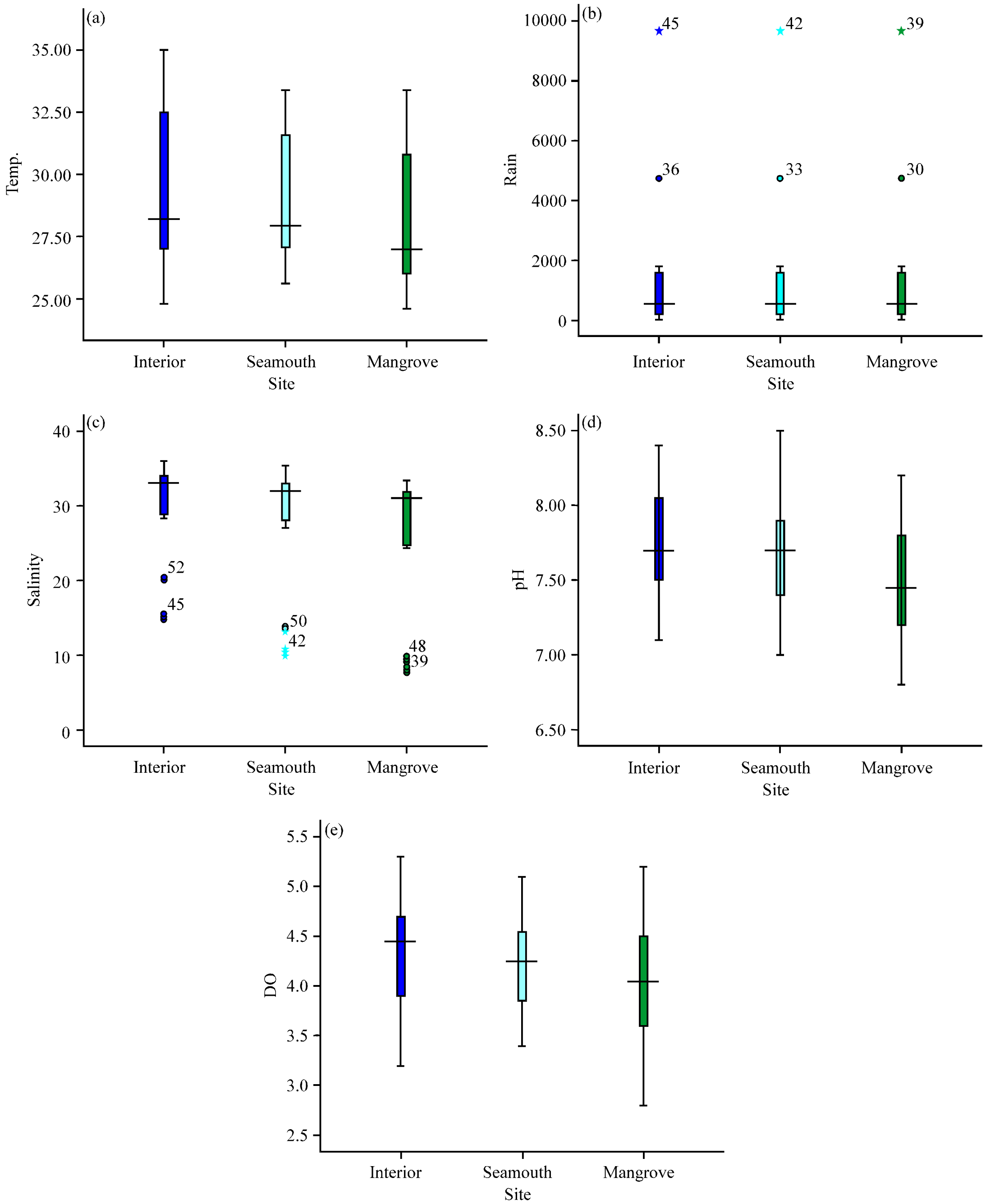

Fig. 2(a-e): Box plot analysis of physical properties (a) Rain, (b) Temperature, (c) salinity, (d) pH and (e) Dissolved oxygen (DO)

oxygen (mL L^{-1}) varied from 3.4-5.1. As expected temperature, salinity and dissolved oxygen showed gradual increase in concentration as we moved from mangrove station to interior sea. The box plots for the inorganic nutrients are presented in (Fig. 3). Nitrites (µg L^{-1}) varied from 0.442-0.912. Nitrates values (µg L^{-1}) were between 2.74-5.6. Phosphates (µg L^{-1}) values ranged between 0.21 and 1.24. The reactive silicate (µg L^{-1}) values ranged from 21.24-62.14. The

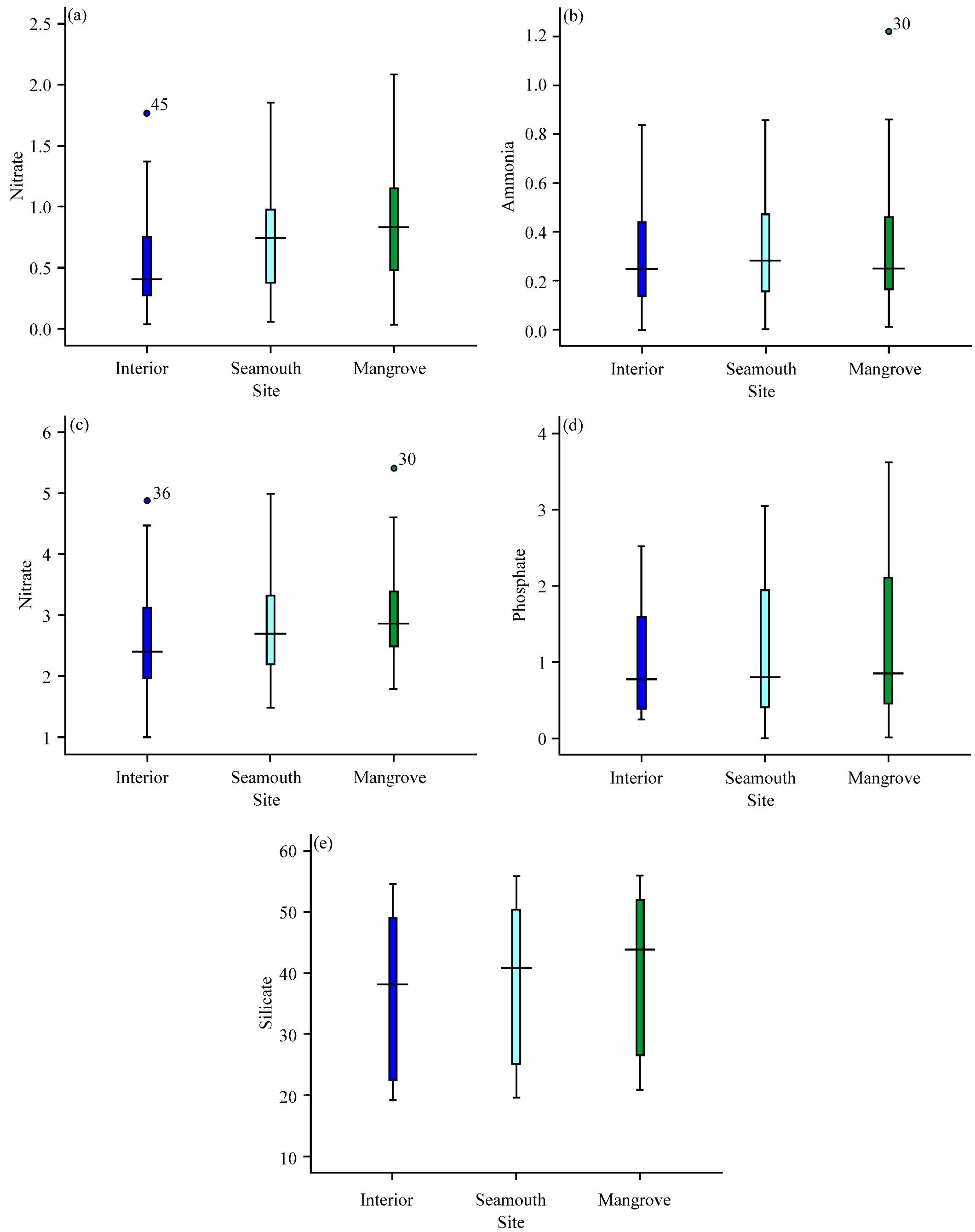

Fig. 3(a-e): Box plot analysis of nutrients (a) Rain, (b) Temperature, (c) Salinity, (d) pH and (e) Dissolved oxygen (DO)

concentrations of ammonia (μg L^{-1}) ranged from 0.062-0.34. Except ammonia all other inorganic nutrients viz., nitrite, nitrate, phosphate and silicate showed a gradual decrease in concentration as we move towards interior to the sea, starting from mangrove station.

Analysis of biological parameters and phytoplankton diversity: The annual variability of primary productivity values (mg cm 3 h^{-1}) ranged between 13-124 during monsoon to summer

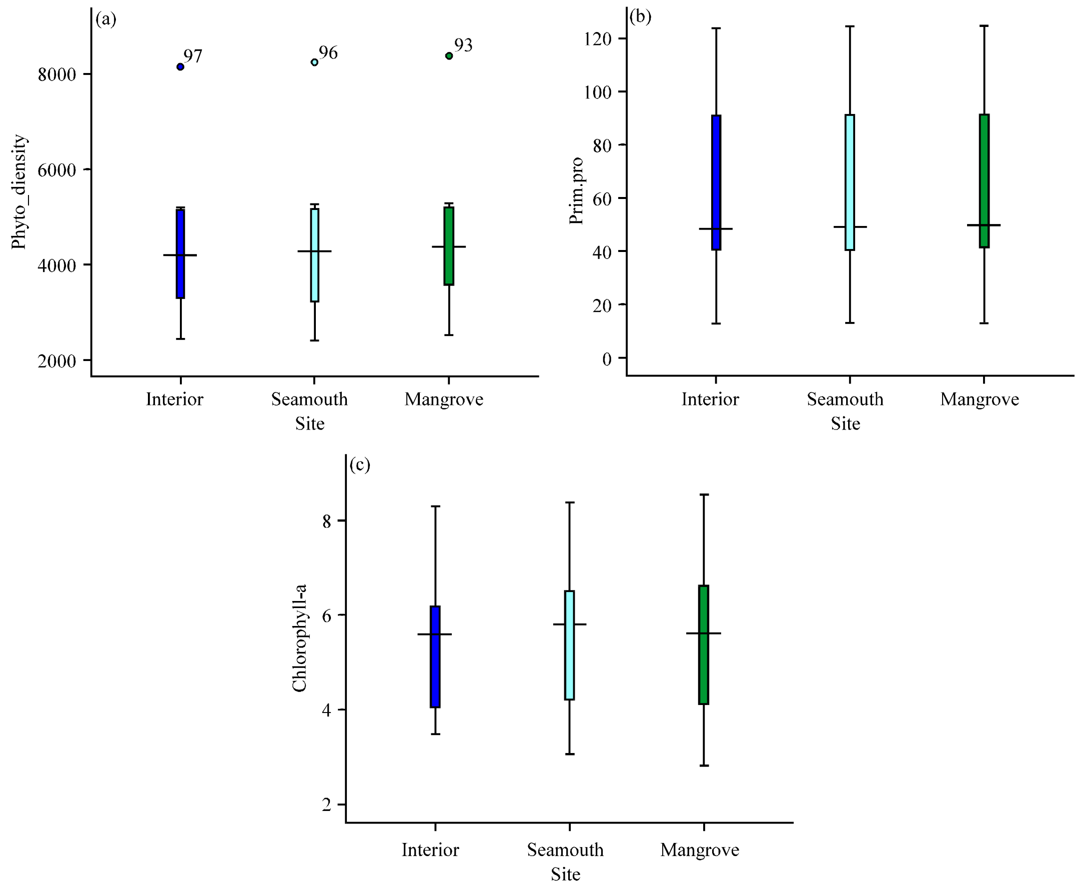

Fig. 4(a-c): Box plot analysis of (a) Phytoplankton density (Phyto_density), (b) Primary production (Prim.pro) and (c) Chlorophyll-a

seasons, respectively. The chlorophyll a ranged between 3.22-8.14 mg m^3. The quantitative ranges of phytoplankton population density (cells L^{-1}) were 3025-8379. Clearly box plot indicate that primary productivity, Chlorophyll-a and phytoplankton density maximum concentration was near mangrove site with respect to sea mouth and interior Fig. 4. The qualitative data of phytoplankton from during the study period list is shown in Table 1. Totally 45 species representing the class of Bacillariophyceae (36), Dinophyceae (6), Chlorophyceae (1) and Cyanophyceae (2) were recorded from three sites. Among the four classes, Bacillariophyceae appeared as the dominant group in terms of total species and cell numbers. The maximum phytoplankton qualitative diversity was found at site three followed by site one and site two, respectively.

Biodiversity indices: Shannon-wiener's diversity index (H') were ranged from 4.326-4.925 bits ind^{-1}. Gleason richness (D') was ranged from 0.926-0.989 and Pielou's evenness (J') were ranged from 0.923-1.002. (Fig. 5).

Multivariate statistical analyses: In the present study, physicochemical parameter and phytoplankton density data were taken for one year from three sites of Parangipettai, India. ANOVA results revealed that the rain was significant ($p<0.01$) with respect to months but was not

Table 1: Check list of phytoplankton species from Vellar estuary during 2008-2009

Phytoplankton	Site 1	Site 2	Site 3
Bacillariophyceae			
Asterionella japonica	+	-	+
Bacteriastrum comosum	+	+	-
Bacteriastrum furcatum	-	+	-
Bacteriastrum sp.,	+	+	-
Bacillaria paradoxa	+	-	+
Bacteriastrum hyalinum	+	-	+
Odontella reticulata	-	+	+
Odontella heteroceros	+	+	+
Coscinodiscus centralis	+	+	+
Coscinodiscus gigas	+	+	+
Coscinodiscus radiatus	+	+	+
Cyclotella striata	-	-	+
Chaetoceros affinis	+	-	+
Chaetoceros curvisetus	+	-	+
Chaetoceros decipiens	+	-	+
Chaetoceros diversus	-	-	+
Chaetoceros furcatus	+	+	+
Ditylum brightwellii	+	-	+
Hemidiscus hardmanianus	-	+	+
Isthmia nervosa	+	-	-
Nitzschia longissima	+	-	+
Nitzschia seriata	+	-	-
Odontella sinensis	+	-	+
Odontella mobiliensis	+	+	-
Navicula henneydii	+	+	+
Pleurosigma elongatum	+	+	+
Pleurosigma normanii	+	+	+
Rhizosolenia styliformis	+	+	-
Rhizosolenia imbricata	+	-	+
Rhizosolenia robusta	+	-	-
Skeletonema costatum	+	+	+
Stephanophysisalmariana	-	-	+
Thalassionema nitzschoides	+	+	-
Thalassiothrix fraunfeldii	+	-	+
Triceratium reticulatum	-	+	+
Triceratium favus	+	+	+
Dinophyceae (dinoflagellates)			
Ceratium macroceros	+	+	+
Ceratium furca	+	-	+
Ceratium trichoceros	+	-	+
Ceratium extensum	-	+	+
Dinophysis caudata	-	+	-
Protoperidinium oceanicum	+	-	+
Chlorophyte			
Chlorella sp.	-	+	+
Cyanophyte			
Oscillatoria sp.	-	+	+
Trichodesmium erythraeum	-	+	+

+: Present and -: Absent

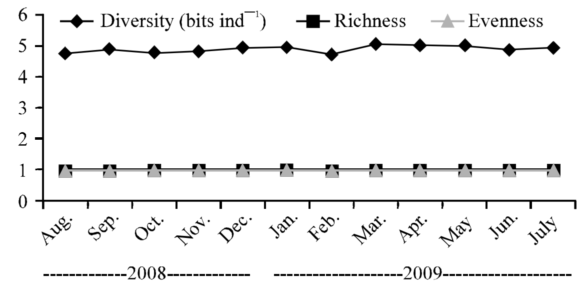

Fig. 5: Biodiversity indices during the studied season

significant in the case of collection sites (Table 2). The temperature significantly varied between the sampling site and months. Similarly salinity, pH, DO, Nitrite, phosphate, primary productivity, silicate and phytoplankton density showed significant variation ($p<0.05$) between sampling sites and months. Concentration of ammonia was not significant ($p<0.01$) with respect to month and sites ($p>0.05$). Moreover, chlorophyll-a was highly significant between months but it was statistically not significant between sampling sites ($p>0.05$).

Cluster analysis: Cluster Analysis (CA) was used to detect similar groups between the three sampling sites in four seasons during 2008-2009. The sampling stations and seasons formed the two major groups. Group A included October to November (Monsoon season) and Group B included December to September, but it was internally further grouped into two groups viz., Group C and Group D which included January to June (post monsoon and summer) and July, August and December (pre-monsoon and monsoon), respectively. Hence, it is clear from the CA analysis that each season formed the group individually but not the stations. It revealed that the sampling station and seasons are significant but not within the group (Fig. 6).

Principal component analysis (PCA): Before applying PCA, Pearson correlation analysis was carried out (Table 3). This was utilized to find an internal structure which ultimately helps in the identification of major sources not accessible at first glance. PCA was applied to the 13 variables collected during four seasons throughout the year (temperature, salinity, pH, rain, dissolved oxygen, nitrate, nitrite, phosphate, silicate, ammonia, primary productivity and Chlorophyll “a”). The highest correlation existed between temperature and chlorophyll-a; temperature and phytoplankton; pH and dissolved oxygen; primary productivity and phytoplankton density and chlorophyll a and phytoplankton density (Fig. 7). Due to the complexity of the relationships, it was difficult to draw more clear conclusions directly. However, principal component analysis can extract the latent information and explain the structure of the data in detail. PCA rendered three significant PCs (eigenvalue >1) that explained 80.73% of the total variance of the dataset. The principal component cannot only interpret the temporal characteristics by clustering the samples, but also can describe their different characteristics and help to elucidate the relationship between different variables by the variable lines. The variable lines were obtained from the factor loadings of the original variables. They stand for the contribution of the variables to the samples. More closer the two variable lines, the stronger is the mutual correlation (Qu and Kelderman, 2001). The highest positive correlation coefficient was observed between phytoplankton density and temperature (Fig. 6).

Table 2: Analysis of variance (ANOVA) tested for physicochemical parameters presented p and F- value of sampling seasons, site and month and site together

Parameters	Terms	df	F-values	p-values	Significance
	Month	11	5.486E8	0.000	**
	Site	2	0.146	0.864	ns
Rain	Month and Site	22	0.244	1.000	ns
	Month	11	505.572	0.000	**
	Site	2	89.476	0.000	**
Temp	Month and Site	22	7.555	0.000	**
	Month	11	3.655E3	0.000	**
	Site	2	917.813	0.000	**
Salinity	Month and Site	22	46.188	0.000	**
	Month	11	2.905	0.003	**
	Site	2	5.247	0.007	*
pH	Month and Site	22	0.321	0.998	ns
	Month	11	10.687	0.000	**
	Site	2	5.865	0.004	*
DO	Month and Site	22	0.611	0.903	ns
	Month	11	7.614E4	0.000	**
	Site	2	37.769	0.000	**
Prim.pro	Month and Site	22	3.010	0.000	**
	Month	11	122.255	0.000	**
	Site	2	2.989	0.057	ns
Chlorophyll a	Month and Site	22	2.631	0.001	*
	Month	11	39.595	0.000	**
	Site	2	12.604	0.000	**
Nitrate	Month and Site	22	0.933	0.555	ns
	Month	11	6.011	0.000	**
	Site	2	6.126	0.003	*
Nitrite	Month and Site	22	0.433	0.985	ns
	Month	11	52.246	0.000	**
	Site	2	3.918	0.024	*
Phosphate	Month and Site	22	0.937	0.549	ns
Silicate	Month	11	1.121E4	0.000	**
	Site	2	728.744	0.000	**
	Month and Site	22	19.701	0.000	**
Ammonia	Month	11	0.827	0.614	ns
	Site	2	0.141	0.869	ns
	Month and Site	22	0.170	1.000	ns
Density	Month	11	1.449E8	0.000	**
	Site	2	1.481E6	0.000	**
	Month and Site	22	1.382E5	0.000	**

*$p<0.005$, **$p<0.01$, **Significant 99%, ns: Not significant

Factor analysis: In this study, water quality variables were grouped using Factor Analysis (FA). The correlation matrix of variables was generated and factors were extracted by the Centroid method. Results of factor analysis including factor-loading matrix, eigenvalues, total and cumulative variance values are presented in Table 4. The factor analysis generated three significant factors which explained 80.73% of the variance in data sets. Parameters were grouped into three factor based on the factor loadings namely factor 1: Salinity, Temperature, Primary

Table 3: Correlation matrix (Pearson (n) of the physicochemical parameters

Variables	Rain	Temp.	Salinity	pH	DO	PP	Chl-a	Nitrate	Nitrite	Phos.	Silicate	Amm.	p_density
Rain	1												
T	-0.458	1											
Salinity	-0.741	0.596	1										
pH	-0.252	0.506	0.464	1									
DO	-0.461	0.548	0.475	0.784	1								
PP	-0.537	0.774	0.444	0.385	0.424	1							
Chl-a	-0.452	0.809	0.526	0.529	0.733	0.680	1						
Nitrate	0.593	-0.176	-0.533	0.106	-0.076	-0.261	-0.190	1					
Nitrite	0.597	-0.136	-0.499	0.359	0.161	-0.182	0.032	0.698	1				
Phosphate	0.680	-0.518	-0.718	-0.069	-0.118	-0.529	-0.328	0.761	0.700	1			
Silicate	0.531	-0.341	-0.613	-0.191	-0.058	-0.317	-0.091	0.523	0.432	0.771	1		
Ammonia	0.254	-0.130	-0.181	0.452	0.284	-0.198	-0.028	0.483	0.661	0.506	0.171	1	
p_density	-0.449	0.727	0.415	0.312	0.523	0.785	0.715	-0.233	-0.104	-0.346	-0.028	-0.160	1

Values in bold are different from 0 with a significance level alpha: 0.05, T: Temperature (°C), PP: Primary productivity, Chl-a: Chlorophyll-a, Amm: NH_3, p_density: Phytoplankton density

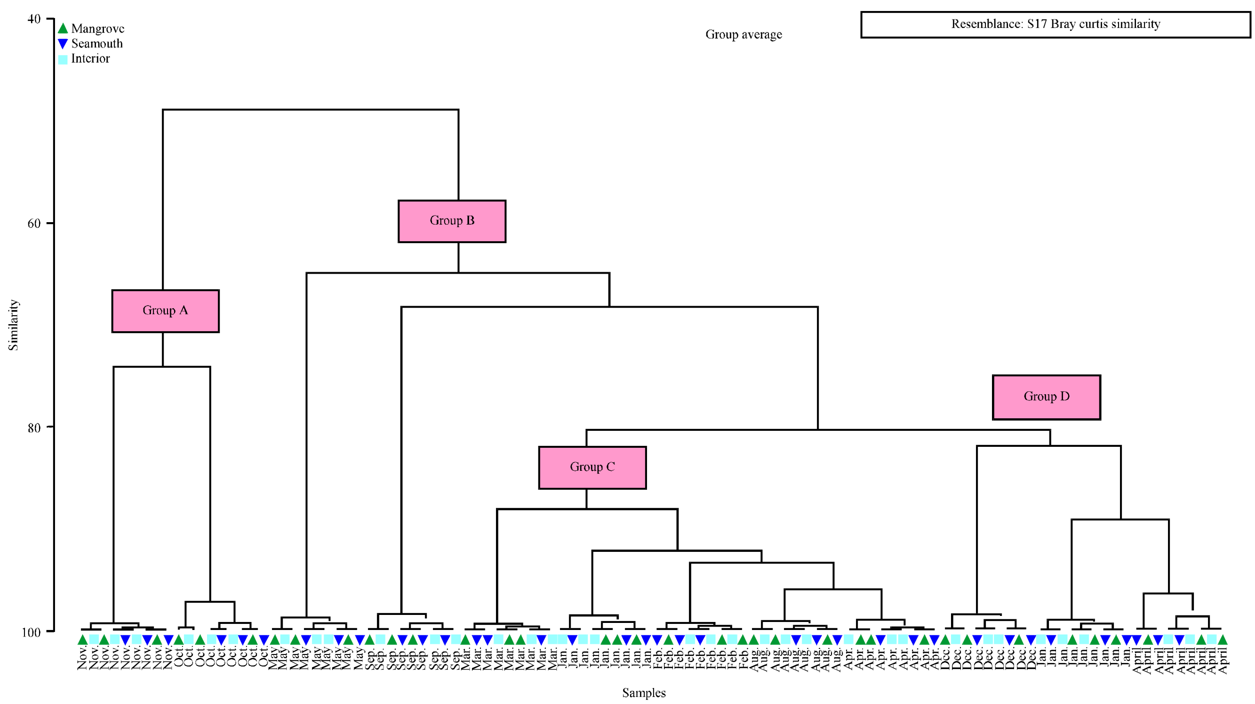

Fig. 6: Dendrogram showing clustering of sampling sites according to water quality parameters of selected sites

productivity and Chlorophyll-a; factor 2: Nitrite, Hydrogen ion concentration (pH), Ammonia and Dissolved oxygen and factor 3: Silicate and Phytoplankton density. Factor 1 (F1) explained 45.44% of the variance. The F1 revealed a high positive loading with respect to salinity, temperature, primary productivity and chlorophyll-a which were 0.847, 0.805, 0.779 and 0.729, respectively.

Confirmation of FA results by PCA analysis: Confirmation of FA results by PCA analysis Principal component analysis (PCA) was applied to data set to confirm results of FA. A scree plot

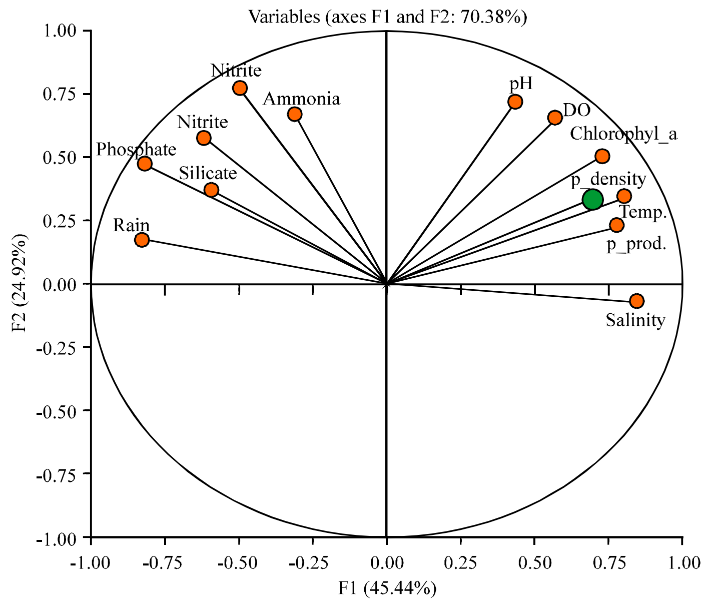

Fig. 7: PCA of physicochemical parameters

Table 4: Factor-loading matrix, eigenvalues and total and cumulative variance values

Variables	F1	F2	F3
Rain	-0.822	0.176	0.164
Temperature	0.805	0.342	0.209
Salinity	0.847	-0.067	-0.326
pH	0.437	0.717	-0.419
DO	0.571	0.656	-0.170
PP	0.779	0.229	0.340
Chlorophyll a	0.729	0.503	0.239
Nitrate	-0.612	0.573	0.086
Nitrite	-0.494	0.770	-0.053
Phosphate	-0.815	0.472	0.105
Silicate	-0.590	0.368	0.523
Ammonia	-0.311	0.668	-0.492
Phytoplankton density	0.696	0.336	0.504
Eigenvalue	5.908	3.242	1.345
Variability (%)	45.444	24.937	10.350
Cumulative %	45.444	70.381	80.730

*F: Factor

presented in Fig. 8 shows the sorted eigenvalues from large to small as a function of the principal components number. As is seen in this figure PCA generated three significant components (number of components of which the eigenvalues are greater than “1” was three). The components weights are presented in Table 5. PCA analysis results also revealed that the first component was associated with Salinity, Temperature, Primary productivity and Chlorophyll-a. The second component comprised Nitrite, Hydrogen ion concentration (pH), Ammonia and Dissolved oxygen and third component Silicate and Phytoplankton density.

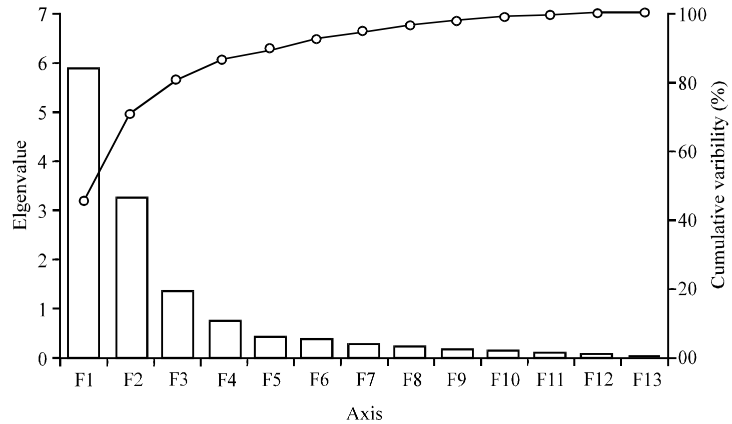

Fig. 8: Scree plot of the eigenvalues

Table 5: Principal component weights

Variables	PC1	PC2	PC3
Rain	-0.822	0.176	0.164
Temperature	0.805	0.342	0.209
Salinity	0.847	-0.067	-0.326
pH	0.437	0.717	-0.419
DO	0.571	0.656	-0.170
PP	0.779	0.229	0.340
Chlorophyll a	0.729	0.503	0.239
Nitrate	-0.612	0.573	0.086
Nitrite	-0.494	0.770	-0.053
Phosphate	-0.815	0.472	0.105
Silicate	-0.590	0.368	0.523
Ammonia	-0.311	0.668	-0.492
p_density	0.696	0.336	0.504

*PC: Principal component

DISCUSSION

In the present study, physicochemical parameters showed a marked difference with respect to the season. Rainfall is the most important cyclic phenomenon in tropical countries as it brings important changes in the hydrological characteristics of the coastal marine environments. The peak values of rainfall were recorded during monsoon in November. The observed high value in March and low value in August could be due to strong land sea breeze and precipitation (Senthilkumar *et al.*, 2002; Santhanam and Perumal, 2003). But there was no difference among rainfall with respect to three stations since the sites were in close vicinity (2 km). The salinity were high during summer season and low during the monsoon season which is in accordance with the findings (Sundaramanickam *et al.*, 2008). Higher summer values (35.0%) may also be attributed to high degree of evaporation and the low amount of rainfall (Sampathkumar and Kannan, 1998; Govindasamy *et al.*, 2000; Rajasegar, 2003). Salinity is a major limiting factor in the distribution of living organisms and variation in salinity by dilution and evaporation is most likely to influence the flora and fauna of the coastal ecosystems (Balasubramanian and Kannan, 2005; Sridhar *et al.*, 2006). pH was recorded highest in summer which might be due to the influence of seawater penetration and high biological activity (Das *et al*., 1997). Generally, temporal fluctuations in pH could be attributed to factors like removal of CO_2 by photosynthesis through bicarbonate degradation, dilution of seawater by freshwater influx, low primary productivity, reduction of salinity and temperature besides decomposition of organic matter (Paramasivam and Kannan, 2005; Prabu *et al*., 2008). Higher values of dissolved oxygen were recorded during monsoon. The observed high monsoonal values might be due to the cumulative effect of higher wind velocity coupled with heavy rainfall and the resultant freshwater mixing (Das *et al*., 1997). It is well known that the temperature and salinity affect the dissolution of oxygen (Vijayakumar *et al*., 2000). Mitra *et al*. (1990) mainly attributed seasonal variation of dissolved oxygen to freshwater flow and terrigenous impact of sediments.

The low nitrite values recorded during summer season may be due to less freshwater inflow and high salinity (Mani and Krishnamurthy, 1989; Murugan and Ayyakkannu, 1991). Similarly, the high level of nitrate could be attributed to fresh water in flow, mangrove leaves (litter fall) decomposition and terrestrial run-off during the monsoon season (Karuppasamy and Perumal, 2000; Santhanam and Perumal, 2003). The recorded low summer values might be due to the limited flow of freshwater, high salinity and utilization of phosphate by phytoplankton (Senthilkumar *et al*., 2002). The low value of silicate recorded during post-monsoonal season could be attributed to uptake of silicates by phytoplankton for their biological activity (Mishra *et al*., 1993; Rajasegar, 2003).

Nutrients are considered as one of the most important parameters in the mangrove environment influencing the distribution of phytoplankton. Distribution of nutrients is mainly based on the season, tidal conditions and freshwater flow from land sources. Higher concentration of ammonia was observed during monsoon season which could be possibly due to the mortality and subsequent decomposition of phytoplankton and also due to the excretion of ammonia by planktonic organisms (Segar and Hariharan, 1989). High concentration of inorganic phosphate observed during monsoon season might possibly be due to intrusion of upwelling seawater into the creek that increased the level of phosphate (Nair *et al*., 1984).

Higher primary productivity was observed during the summer season, because of high population density of phytoplankton which could also be due to neritic element domination, higher salinity and surface water temperature, clear water conditions besides availability of nutrients (Rajasekar *et al*., 2005). The chlorophyll-a (mg m^{-3}) was highest during summer and the lowest during monsoon which could be due to anthropogenic effects as evidenced by its positive correlation with salinity and may also be due to freshwater discharges from the rivers (dilution), causing turbidity and less availability of light (Godhantaraman, 2002; Rajasekar *et al*., 2005).

The abundance of phytoplankton was lowest during monsoon season. The population density was found to be higher in summer season as reported in other coastal regions of Tamil Nadu (Senthilkumar *et al*., 2002). The number of phytoplankton species increased consistently towards the sea mouth, where the salinity was higher (Rajasekar *et al*., 2005). Biodiversity of phytoplankton index values are comparable to those reported earlier (Ignatiades *et al*., 1985) in Saronicos bay (1.53-4.08 bits ind^{-1}). In general, an increase in population density is directly proportional to the diversity index. In present study high index values were observed during summer season in the month of May, due to the upwelling of the nutrients in the coastal waters and species evenness was recorded maximum in the month of November (monsoon season) due to the high nutrients inflow. The observed maximum phytoplankton richness values were recorded during the post-monsoon season. Maximum species diversity observed during summer season (Rajasegar *et al*., 2000).

Statistical techniques often find significant relationships in large datasets (Luoma and Bryan, 1982). While studying the inter-relating variables, correlations between two or more parameters are not easy to establish at a glance (Luoma and Bryan, 1982). Therefore, multivariate analyses for the interpretation of large environmental and biological datasets have been used in plankton research to identify relationships between abiotic and biotic factors and community interpretation (Matta and Marshall, 1984; Pagou and Ignatiades, 1988; Varis, 1991; Marshall and Alden, 1995; Del Giorgio *et al.*, 1997). Results from temporal cluster analysis showed that, based on water quality parameters and phytoplankton data of the Vellar estuary, Parangipettai waters is discriminated into two major groups which clearly indicate that the differences among seasons are significant, indicating seasonal variability during the sampling periods. Study revealed that the sampling station and seasons are significant but not with in the group of the sampling station. It is apparent that most parameters vary with seasons.

PCA results displayed a good correlation for the temperature, primary productivity, Chlorophyll-a, Dissolved oxygen and pH in relation to the phytoplankton density which clearly indicates the immense role of these parameters in the higher growth and production of phytoplankton. Based on the highly correlated values of the parameter as evident by the PCA, these physicochemical and biological parameters could be used effectively and efficiently for enhancing the growth of the phytoplankton while culturing in the laboratory conditions. Hence, once again PCA proves its utility as a powerful pattern recognition tool which attempts to explain the variance of a large data set of intercorrelated variables with a smaller set of independent variables (Simeonov *et al.*, 2003). However, further optimization of the parameters need to be done before implementing for the mass scale production.

High positive loadings indicated strong linear correlation between the factor and parameters. A number of sources could be responsible for high nitrite concentrations in Parangipettai waters: natural organic matter decomposition and deep percolation of nitrate resulting from aquaculture and agricultural waste discharge (Rajasegar *et al.*, 2002). Both point and non-point sources could be the source of ammonia in the study sites. Point sources include municipal waste, industrial operations and large confined livestock operations. Non-point sources comprise soil erosion and water runoff from aquaculture farm and cropland (Devlin *et al.*, 2000).

Based on the results of the factor analysis and also hydrochemical aspects of the water, it was concluded that, F1 can be denoted as "agricultural pollution" factor with presence of Salinity, Temperature, Primary productivity and Chlorophyll-a. F2 which is highly correlated with Nitrite, Hydrogen ion concentration (pH), Ammonia and Dissolved oxygen saturation can be denoted as "Aquaculture farm discharges" factor. Silicate and Phytoplankton density included in F3 are the indicator of organic pollution in water. Therefore, F3 factor represents "organic substances" derived from the litter of coastal mangroves and dead remains of aquatic organisms. These findings of Factor Analysis (FA) were confirmed by the results of PCA.

As mentioned earlier, aquaculture practices and agriculture is the primary land uses (78% of the total catchment area) in the Parangipettai waters and still this percentage is increasing due to more demand of highly nutritive shrimps. The result of the factor analysis supports and confirms that the surface water quality and phytoplankton density of the Parangipettai waters and its tributaries is being affected by aquaculture discharges. On the other hand, uncontrolled agriculture land run-off is causing threat to the surface water quality (Boyacioglu, 2006). This is the alarming sign for the future.

CONCLUSIONS

This study shows that multivariate statistical methods are useful tool for understanding of complex nature of water quality issues by identifying groupings in the set of data. The Parangipettai waters is subjected to seasonal fluctuations in physicochemical parameters depending upon the seasonal tidal amplitude and fresh water influx resulting in a continuous exchange of organic, inorganic, plant and animal matters. This coastal water was a resourceful place for phytoplankton cell abundance and diversity. It is a good tool to comprehend the phytoplankton spatial distribution in marine ecosystem if the species and environment matrix data are suitable. The collected information data on the phytoplankton distribution and abundance with reference to hydrography would form a useful tool for further ecological assessment of Parangipettai waters, southeast coast of India. Thus, multivariate statistical methods including factor, principal component and cluster analysis can be used to understand complex nature of physicochemical parameters and determine priorities to improve the water quality and phytoplankton density. If the current scenario continues, this may lead to severe consequences to the coastal waters of Parangipettai, where thousands of fishermen are directly dependent on these resources as it is the only source of income for their livelihood. In this aspect a proper and effective monitoring of the aquaculture practices along the Parangipettai coast and estuary is the need of the hour.

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