

Chapter 2

Diatoms of the benthic community

2A.1 Introduction

Diatoms constitute an important part of the microphytobenthic community at intertidal sand flats. The diatom population here is usually composed of pennate diatoms, which are either epipsammic (attached to sand grains) or epipellic (motile forms within sediments). Intertidal sand flats are dynamic environments, where the tidal generated water movement and the associated processes of deposition and resuspension of sediment affect the composition and distribution of diatoms. In addition, hydrodynamic processes carry planktonic diatoms present in the ambient water to the intertidal sediment. These planktonic forms can be in either their vegetative or their resting stage, and can contribute to population dynamics. So far, studies on diatom populations have been restricted to the epipellic pennate diatoms, and those forms which reside, although temporarily, on or within the sediment grains have not received due attention. Diatom populations could, however, be better understood by taking into consideration both permanent residents and temporary visitors.

In many studies, diatoms were only investigated in the top few centimeters of the sediment (Riznyk et al. 1978; Colijn and Dijkema 1981; Varela and Penas 1985; Lukatelich and McComb 1986). However, the presence of diatoms at a depth of 20 cm has also been reported (Steele and Baird 1968; Colijn and Dijkema 1981; de Jonge and Colijn 1994), based on chlorophyll *a* estimations. In intertidal sand flats, a number of factors may be responsible for displacing the diatoms from the surface sediment layers to the deeper layers. Diatoms can retain photosynthetic capacity in the dark, deeper sediments and thus form an important pool of potential primary producers, which can resume photosynthesis if resurfaced (Fielding et al. 1988).

In the present study, sediment at the low, mid and high tide zones was investigated for the temporal variation in diatom abundance up to a depth of 15 cm. Chlorophyll *a* content and physico-chemical parameters such as wind speed, sediment characteristics, nutrients and suspended load were correlated with diatom abundance. Such a study will help gain insight into the dynamics of diatom populations, which form an important component of the microphytobenthic community, responsible for the littoral production and thus forming a basic link in intertidal food webs.

2A.2 Materials and methods

2A.2.1 Study area and sampling strategy

Sediment sampling was carried out on a sand flat at Dias Beach ($15^{\circ} 27' N$; $73^{\circ} 48' E$), located near Dona Paula Bay and surrounded by the Mandovi and Zuari estuaries (Fig. 2A.1). This beach is about 200 m in length. The tides are semi-diurnal with an

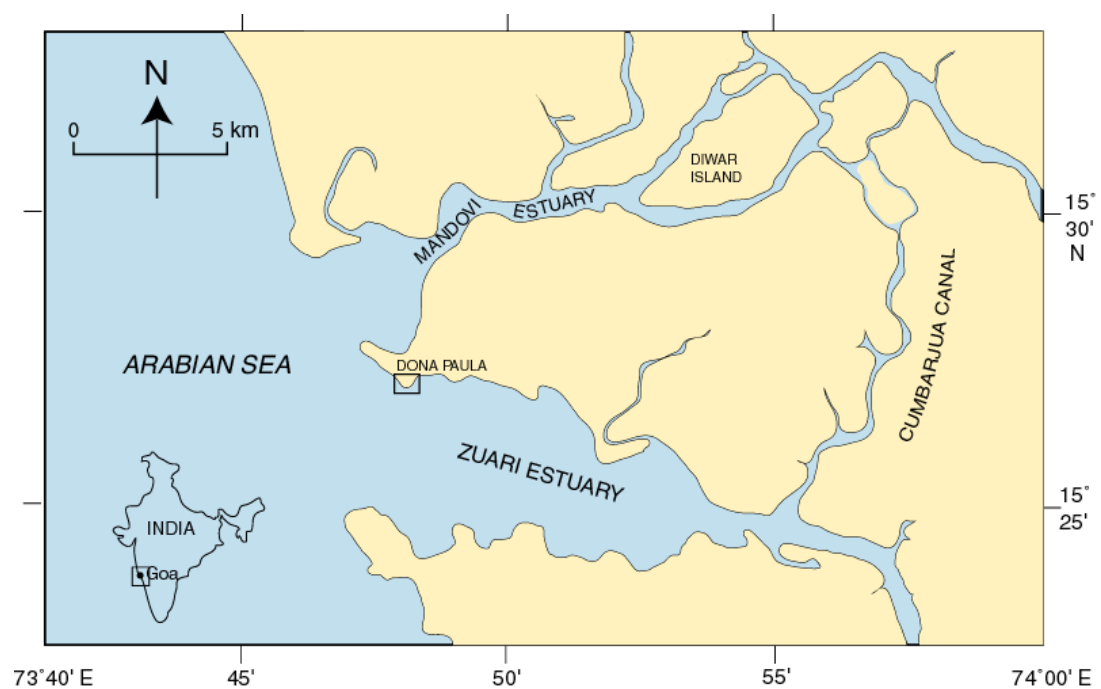


Fig. 2A.1 Geographical location of study

average spring tide range of ~ 2 m and neap tide range of ~ 0.7 m. In this area, the wave heights are > 1.5 m during June-August and low (< 0.7 m) during October-April (Chandramohan et al. 1997). Dias Beach is sheltered and protected on both sides by rocky cliffs. This locality experiences three seasons: the pre-monsoon (February-May), the southwest monsoon (June-September) and the post-monsoon (October-January). The sediment sampling at the low tide zone was carried out on a monthly basis for 17 months (May 1998-September 1999) whereas at the mid and high tide zones was carried out every alternate month. Sediment samples were collected in triplicates during the lowest low tide, using a hand-corer with an inner diameter of 4.5 cm. The core length collected was 15 cm. Each core was divided into three sections of 5 cm intervals (0-5, 5-10 and 10-15 cm). Simultaneously, a separate sediment core of the same dimensions was collected for chlorophyll *a* and grain size analysis.

Interstitial water samples were collected by digging the sediment (~ 15 cm) with a shovel and allowing the water to collect. The water samples were allowed to stand for a few seconds while sand particles settled. Temperature of the surface sediment was recorded at the study site, and analysis of salinity (Mohr-Knudsen titration method) (Strickland and Parsons 1965) and nutrients such as nitrate ($\text{NO}_3\text{-N}$), nitrite ($\text{NO}_2\text{-N}$), phosphate ($\text{PO}_4\text{-P}$) and silicate (SiO_3) along with chlorophyll *a* (corrected for phaeopigments) were carried out by standard procedures (Parsons et al. 1984). Salinity and nutrient values were obtained for the entire 15 cm core, whereas chlorophyll *a* was estimated for each of the three core sections (0-5, 5-10 and 10-15 cm). Simultaneously, surf water samples were also collected from the surface at about 3 m from the low tide zone for the analysis of water parameters as mentioned above. A known volume of surf water (500 ml) was transferred to PVC bottles

(triplicates) and preserved with Lugol's iodine solution to estimate the diatom population following the method described by Hasle (1978).

2A.2.2 Enumeration of diatoms in the sediment

Since this study was carried out on a sandy beach and included the observation of attached forms, direct observation and quantification of samples posed a problem. Even if diatoms in their attached state could be observed directly, there was always a chance of missing cells attached to the other side of the grain. The ultrasonication technique was not of much help either, since some forms did not detach from the substrata and fragile forms were at risk of being ruptured, thus leading to false quantification results. Therefore, the extinction dilution method (most-probable-number method, MPN) (Imai et al. 1984; Yamochi 1989; Imai et al. 1990; Ishikawa and Tamaguchi 1994; Itakura et al. 1997) was employed for the quantification of the diatom flora in each section of the sediment core at the low, mid and high tide zones. Subsequent to incubation of the sediment samples, the appearance of centric and pennate diatoms was observed. The appearance of both these forms may have been from two sources; either they were the products of multiplication of vegetative cells or germination of resting stages.

An appropriate amount (1 to 2 g wet wt) of sediment sample was suspended in f/2 medium (Guillard and Ryther 1962) at a concentration of 0.1 g wet wt ml⁻¹. This stock was then subjected to a serial ten-fold dilution (10⁻¹ - 10⁻⁵) with the culture medium, and then 1 ml aliquots of diluted suspensions were inoculated into five replicate culture wells. Incubation was carried out at 27 ± 1 °C with a 12 h light: 12 h dark photcycle. The growth of diatoms in each culture well was examined microscopically after an incubation period of 6-8 days. The wells in which growth was observed were scored as positive. The MPN (for a series of 3 ten fold dilutions)

of diatoms in the sediment sample (MPN g^{-1} wet sediment) was then calculated according to a statistical table (Thronsen 1978). This table covers a range of five dilution steps, and a set of three dilutions have to be chosen out of the five cultured to get the MPN number. The diatom density per cubic centimeter wet sediment was obtained by multiplying the MPN value with the apparent specific gravity of the wet sediment (Imai and Itakura 1999). The diatoms were identified based on the keys provided by Heurck (1896), Subrahmanyam (1946), Desikachary (1977,1987) and Tomas (1997).

2A.2.3 Grain size analysis

Sediment grain size at the low, mid and high tide zones was analyzed by dry sieving (Folk 1968). Categories of grain size included: $>1,000 \mu\text{m}$ (very coarse sand); 500-999 μm (coarse sand), 250-499 μm (medium sand), 125-249 μm (fine sand), 63-124 μm (very fine sand) and $< 63 \mu\text{m}$ (silt and clay).

2A.2.4 Data analysis

Univariate statistical methods

Univariate techniques included the calculation of the Shannon-Wiener diversity index (H'), richness ($H' \text{ max.} = \log_2 S$, where S is the number of taxa) and evenness ($J' = H'/H' \text{ max.}$) of the total diatom population (both benthic and planktonic) (Pielou 1969) at the low, mid and high tide zones.

Multivariate statistical methods

The $\log(X + 1)$ - transformed data on the relative abundance of diatoms in the intertidal sediment at the low, mid and high tide zones was used for the construction of a lower triangular dissimilarity matrix using Bray-Curtis coefficients (Bray and Curtis 1957). This dissimilarity matrix was then subjected to clustering and ordination analysis. Clustering was performed using the weighted pair group average method

(Pielou 1984). Ordination was by non-metric multidimensional scaling (NMDS) (Kruskal and Wish 1978). There are advantages in applying more than one method, since each is based on different assumptions and may give different insights (Gray et al. 1988). In case of the cluster analysis: (1) the individual, once placed in a group, loses its identity (2) the sequence of individuals is arbitrary and (3) only the inter-group relationships are shown. In view of these disadvantages, it is advisable to employ an additional method of presentation to show individual relationships such as NMDS (ordination).

The $\log(X + 1)$ -transformed data on diatom abundance in the water column was subjected to clustering by squared Euclidean distance and the group average method (Pielou 1984).

A two-way analysis of variance (ANOVA) (Sokal and Rohlf 1981) was conducted on the $\log(X + 1)$ transformed relative abundance data of each diatom, at the low, mid and high tide zones, in order to test the significance of month and depth. The diversity, richness and evenness values of the diatom assemblages were also subjected to two-way ANOVA to evaluate the temporal (month) and spatial (depth) variations.

Multiway ANOVA (MANOVA) without replication (Sokal and Rohlf 1981) was performed on the arcsine-transformed grain size values (sediment composition) at the low, mid and high tide zones, in order to assess whether any significant variance existed with respect to different sampling periods (months) and depths.

Diatom relative abundance and the arcsine-transformed grain size data for the different depths and tidal zones as well as the $\log(X + 1)$ -transformed data on water parameters (temperature, salinity and nutrients) were subjected to multiple regression analysis.

Regression was performed on the log (X+1)- transformed data on sediment chlorophyll *a* (linear function), wind speed and suspended load (power function) versus diatom abundance.

2A.3 Results

2A.3.1 Environment

Surf water temperature ranged between 20 °C and 31 °C whereas that of sediment surface ranged between 21 °C and 30 °C respectively. Salinity was as low as 10 psu in the monsoon, while during the other seasons it ranged between 28 and 35 psu. The nutrient concentrations in the surface and interstitial waters showed a similar trend. Nitrate concentration ranged between 0.08 and 12 μM and was maximum in monsoon

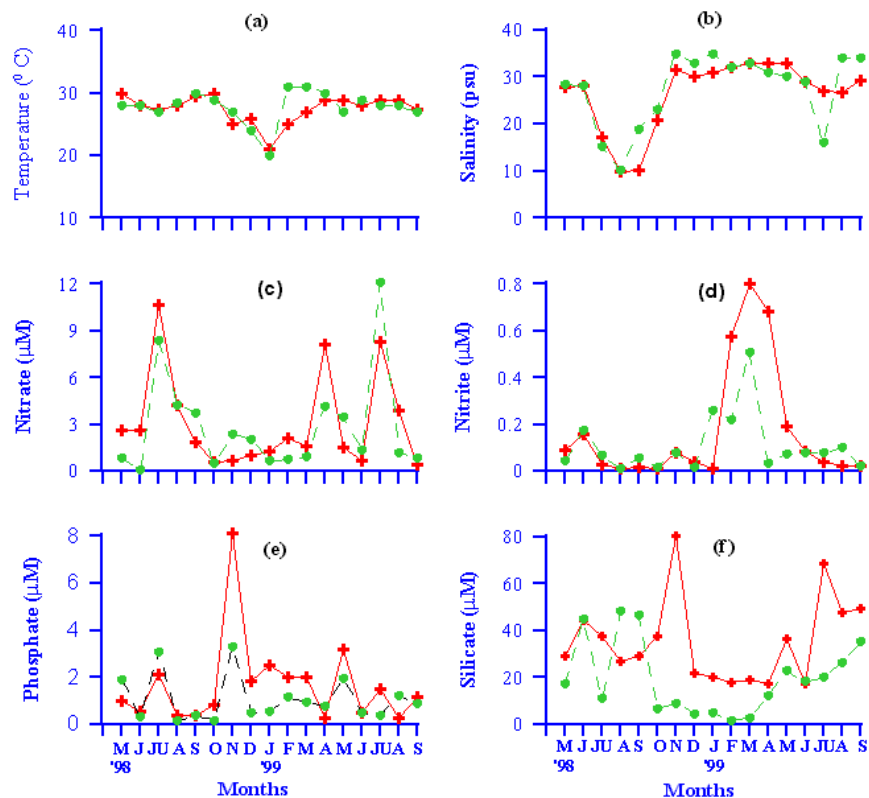


Fig. 2A.2 Temporal variations in environmental parameters of surf waters and interstitial waters

(a) Temperature; (b) Salinity; (c) Nitrate (NO₃-N); (d) Nitrite (NO₂-N); (e) Phosphate (PO₄-P) and (f) Silicate (SiO₃)

—+— Interstitial waters - - - ● - - - Surf waters

with a peak in July, which was followed by a bloom of *Skeletonema costatum* in the ambient waters. Nitrite ranged between 0.013 and 0.8 μM being highest in premonsoon (March). Premonsoon and postmonsoon seasons experienced a rise in the phosphate concentration in the surf and interstitial waters. The surf water silicate concentrations were generally highest during the monsoon whereas in interstitial waters it was found to be above 17 μM throughout the investigation period (Fig 2A.2). The observed low salinity, high nitrate and silicate concentrations were due to high land runoff during the monsoon season.

2A.3.2 Diatoms in surf waters

In the water column, 34 species of diatoms belonging to 27 genera were encountered (Table 2A.1a). Of these, 19 were centric and 15 pennate. *Navicula transitans* var. *derasa* f. *delicatula* was found to be the most abundant diatom species.

Clustering of this diatom community at 50% dissimilarity level revealed *Navicula delicatula* to be the most dissimilar diatom in terms of distribution.

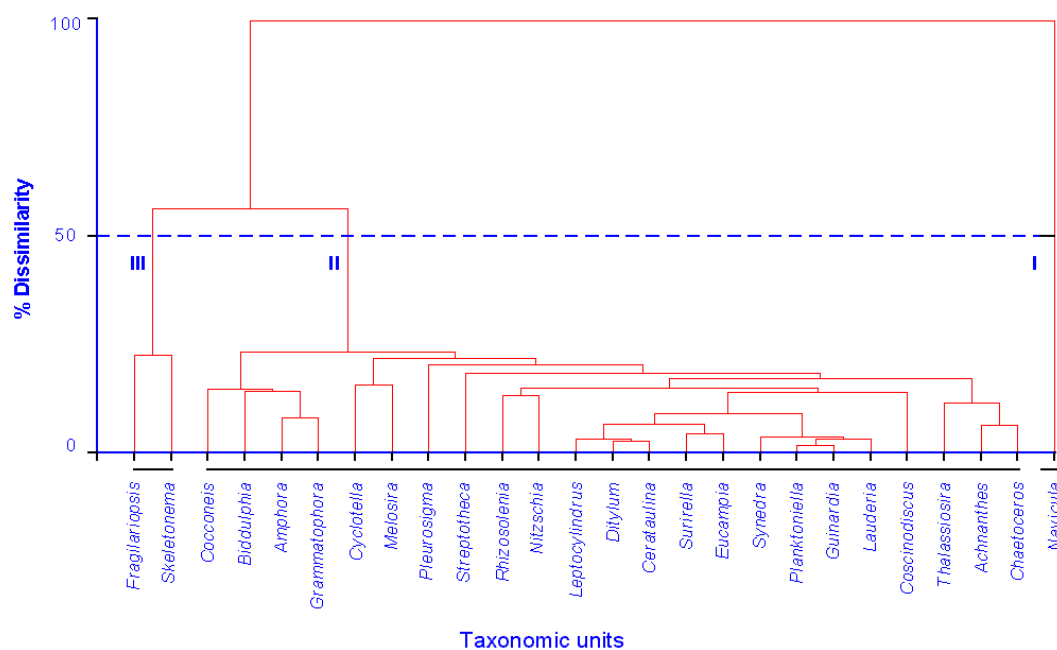


Fig. 2A.3 Cluster dendrogram of the surf water diatom community, constructed by the group average method

Table 2A.1. Diatoms recorded in (a) surf waters, (b) low tide zone, (c) mid tide zone, and (d) high tide zone of intertidal sediment (0-15 cm) during the study period

	a	b	c	d
Centric diatoms				
<i>Bellerochea malleus</i> (Brightwell) Van Heurck		+	+	+
<i>Biddulphia mobiliensis</i> (Bailey) Grunow	+	+	+	+
<i>Biddulphia sinensis</i> (Greville) Grunow	+	+	+	+
<i>Cerataulina pelagica</i> H. Peragallo	+			
<i>Chaetoceros curvisetus</i> Ehrenberg	+	+		
<i>Chaetoceros diversus</i> Ehrenberg	+	+		
<i>Coscinodiscus concinnus</i> Wm. Smith			+	
<i>Coscinodiscus marginatus</i> Ehrenberg	+	+	+	+
<i>Cyclotella</i> sp.	+		+	+
<i>Ditylum brightwellii</i> (West) Grunow	+			
<i>Eucampia zodiacus</i> Ehrenberg	+			
<i>Guinardia flaccida</i> (Castracane) Peragallo	+			
<i>Lauderia borealis</i> Cleve	+			
<i>Leptocylindrus danicus</i> Cleve	+			
<i>Melosira nummuloides</i> C.A. Agardh	+	+	+	+
<i>Planktoniella sol</i> (Wallich) Schutt Peragallo	+			
<i>Rhizosolenia alata</i> Brightwell	+			
<i>Rhizosolenia</i> sp.	+			
<i>Skeletonema costatum</i> Greville	+	+		
<i>Helicotheca tamesis</i> Shrebs	+	+	+	+
<i>Thalassiosira eccentrica</i> (Ehrenberg) Cleve	+	+	+	+
<i>Thalassiosira</i> sp1		+	+	+
<i>Thalassiosira</i> sp2		+	+	+
Pennate diatoms				
<i>Achnanthes longipes</i> Agardh	+			
<i>Achnanthes subsessilis</i> Kutzing			+	+
<i>Amphora coffeaeformis</i> (Agardh) Kutzing	+	+	+	+
<i>Amphora rostrata</i> Wm. Smith	+	+	+	+
<i>Amphora</i> sp.	+	+	+	+
<i>Cocconeis scutellum</i> Ehrenberg	+	+	+	+
<i>Cymbella</i> sp.			+	+
<i>Fragilariopsis cylindrus</i> (Grunow) Cleve	+	+	+	+
<i>Grammatophora marina</i> Kutzing	+	+	+	+
<i>Navicula crucicula</i> (Wm. Smith) Donkin			+	
<i>Navicula transitans</i> var. <i>derasa</i> f. <i>delicatula</i> Heimdal	+	+	+	+
<i>Navicula subinflata</i> Grunow	+	+	+	+
<i>Navicula</i> sp.	+	+	+	+
<i>Nitzschia closterium</i> (Ehrenberg)			+	+
<i>Pinnularia</i> sp.			+	+
<i>Pleurosigma angulatum</i> Sensu W. Smith	+	+	+	
<i>Pseudonitzschia seriata</i> Cleve	+	+	+	+
<i>Raphoneis</i> sp.			+	+
<i>Surirella</i> sp.	+			
<i>Synedropsis gaillonii</i> Grunow	+			
<i>Tabellaria</i> sp.			+	+
<i>Thalassiothrix nitzschoides</i> (Grunow) Mereschkowsky	+	+	+	+

Clustering revealed 3 main groups (Fig. 2A.3). The first group comprised of *Navicula*. The third group was represented by *Skeletonema costatum* and *Fragilariopsis cylindrus*, which dominated blooms in the ambient waters during

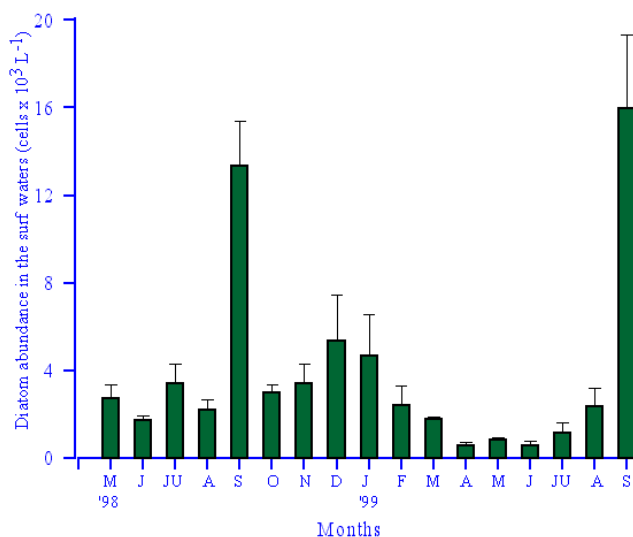


Fig. 2A.4 Temporal variations in total diatom abundance of the surf water

August and September (Fig. 2A.4). The rest of the forms formed the members of the second group.

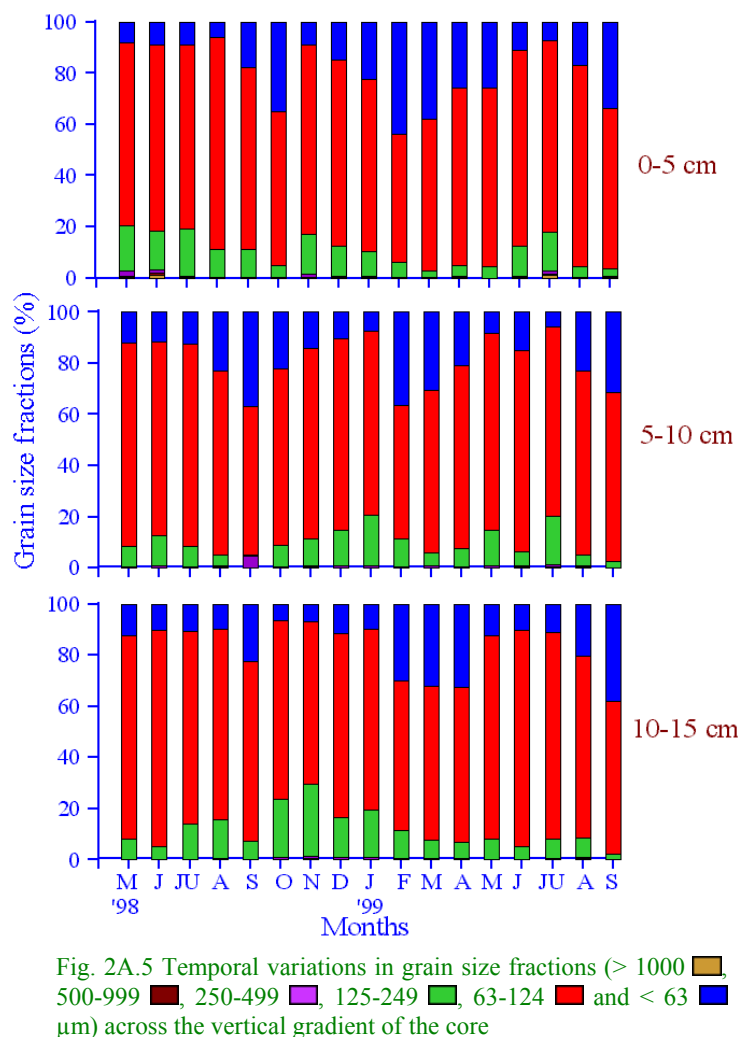
2A.3.3 Low tide zone

Sediment characteristics: Very fine sand (63-124 μm - silt/clay) was found to be the most abundant grain size fraction of the sediment throughout the study period across the 15 cm core (Fig. 2A.5). The contribution of sediment particles > 250 μm was negligible.

MANOVA of different months versus grain size and depth versus grain size revealed

Table 2A.2. MANOVA (Without replication) for the temporal variation in the sediment characteristics along the vertical gradients at the low tide zone (0-5, 5-10 and 10-15 cm sediment core sections) (* $p \leq 0.025$; NS = not significant)

Source of variation	df	SS	MS	Fs
Months	16.00	27.76	1.73	
Core sections	2.00	106.72	53.35	
% Grain size fractions	5.00	70930.88	14186.18	
Months X Core sections	32.00	914.22	28.57	0.787 ^{NS}
Months X % Grain size fractions	80.00	5361.15	67.01	1.845*
Core sections X % Grain size fractions	10.00	749.10	74.91	2.063*
Months X Core sections X % Grain size fractions	160.00	5810.90	36.32	



a significant variation (Table 2A.2). Across the three sections, the percentage of < 63 μm grain size fraction peaked on two occasions. One peak was observed towards the end of monsoon (September to October) while the other was evident during February to April (pre-monsoon). During these periods, a simultaneous decrease in the percentage of 63-124 μm grain size fractions was observed.

Diatoms in sediment: In the sediment, 24 species of diatoms (12 centric, 12 pennate) belonging to 16 (8 each of pennates and centrics) genera were recorded (Table 2A.1b). Pennate diatoms were dominant in terms of abundance (Fig. 2A.6). The most abundant pennate diatoms were *Amphora* and *Navicula*; *Thalassiosira* was the abundant centric diatom.

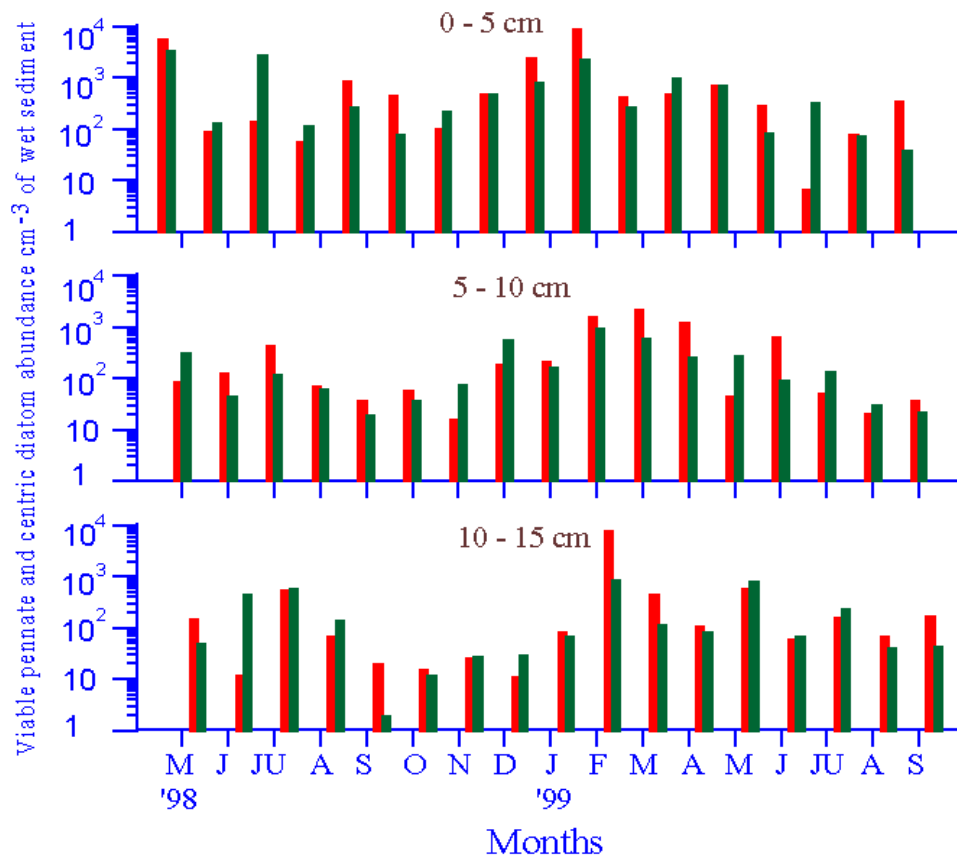


Fig. 2A.6 Temporal variations in pennate and centric diatom abundance across the vertical gradient of the core
■ Pennate diatoms; ■ Centric diatoms

Diatoms encountered in this area can be divided into three groups based on their abundance; *Amphora*, *Navicula* and *Thalassiosira*, which were present throughout the period of investigation, formed the first group (Fig 2A.7a). The second group comprised forms that were frequently encountered in the sediment and included eight genera, of which four were centric diatoms (*Biddulphia*, *Melosira*, *Streptothecha* and *Coscinodiscus*) and four were pennates (*Grammatophora*, *Cocconeis*, *Nitzschia* and *Thalassiothrix*) (Fig. 2A.7b). The diatoms appearing rarely formed the third group and included *Pleurosigma*, *Fragilariopsis*, *Skeletonema*, *Chaetoceros* and *Bellerocha*.

Temporal and Vertical distribution of diatoms in the sediment: The total diatom abundance varied from one sampling period (month) to another, but was found to peak during the pre-monsoon season (Fig. 2A.8). Diatoms belonging to group I (*Amphora*, *Navicula* and *Thalassiosira*) and group II (*Biddulphia*, *Melosira*, *Coscinodiscus*, *Streptotheca*, *Cocconeis*, *Thalassiothrix*, *Nitzschia* and *Grammatophora*) were abundant in the pre-monsoon period, whereas diatoms of group III (*Pleurosigma*, *Fragilariopsis*, *Skeletonema*, *Chaetoceros* and *Bellerochea*) were abundant during the monsoon and post-monsoon seasons.

The temporal variation in abundance of the diatoms across the three individual core sections did not reveal any particular trend. The group I diatoms (*Amphora*, *Navicula* and *Thalassiosira*) were found to peak in February across the three core sections. The

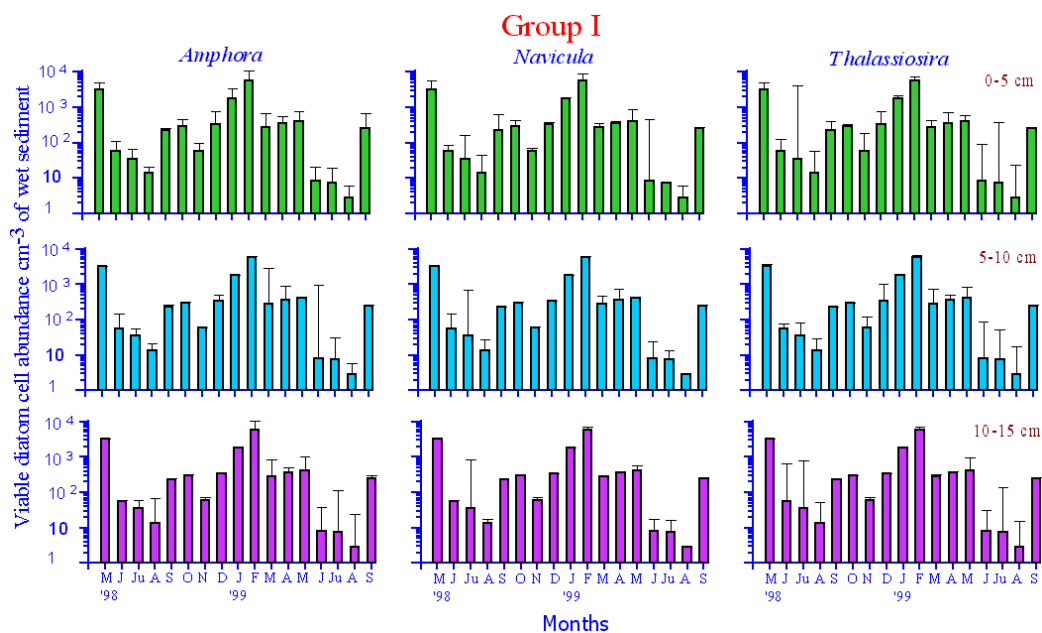


Fig. 2A.7a Temporal variations in diatom abundance across the vertical gradient of the core for group I

distribution of *Amphora* kept a lower profile during the monsoon period, while *Thalassiosira* formed a trough from August to October. *Biddulphia* (group II) peaked during February-March and was not encountered in September down to 15 cm depth.

Group II

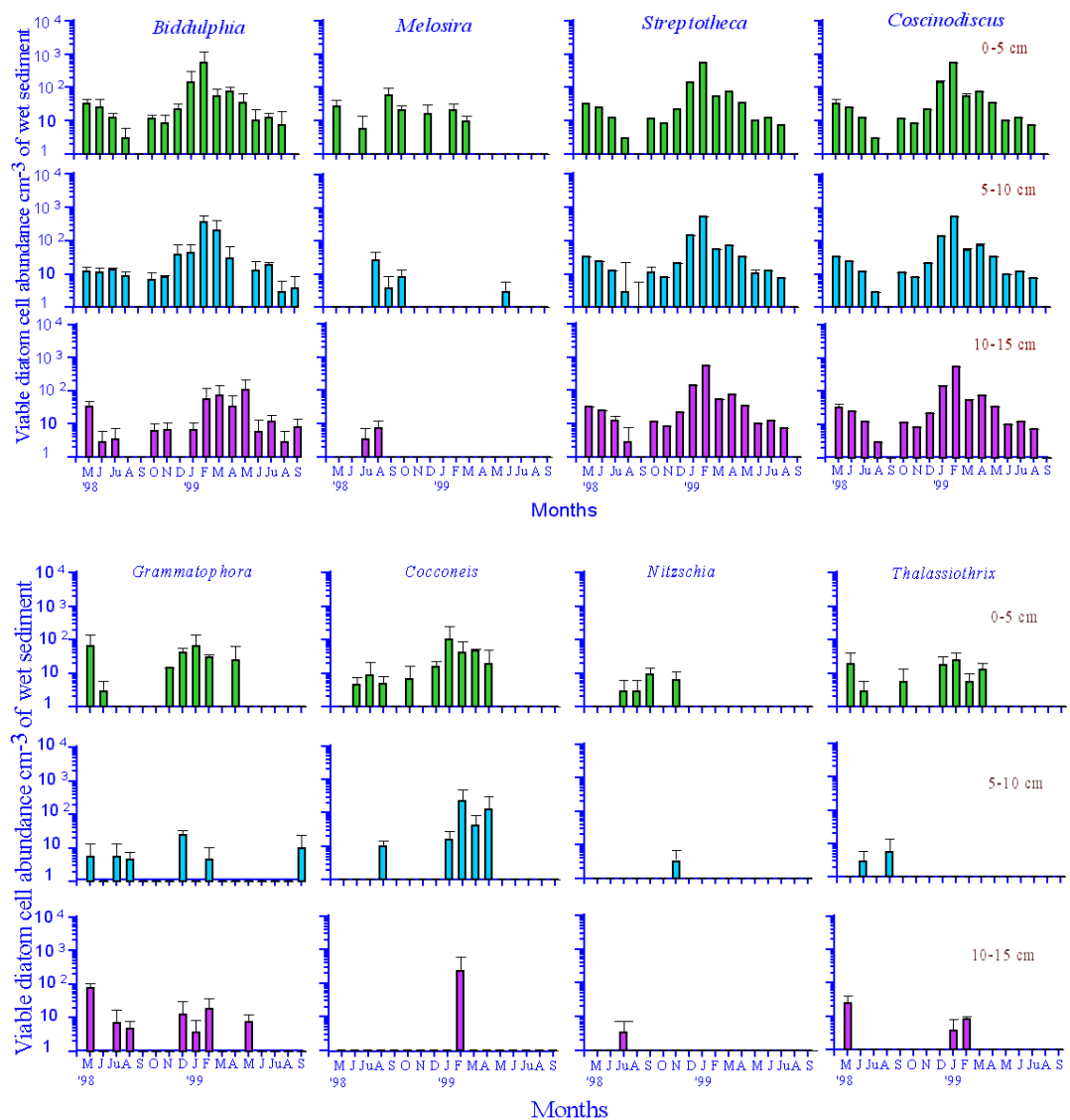


Fig. 2A.7b Temporal variations in diatom abundance across the vertical gradient of the core for group II

Amongst the group III diatoms, *Bellerocha* was present only in the 0-5 cm stratum (May and June), while *Chaetoceros* was found at 0-5 cm (May and August) and at 10-15 cm (August) depths. *Pleurosigma* was encountered on only one occasion (January) in the 0-5 cm stratum, while at 5-10 cm it was observed during January and August.

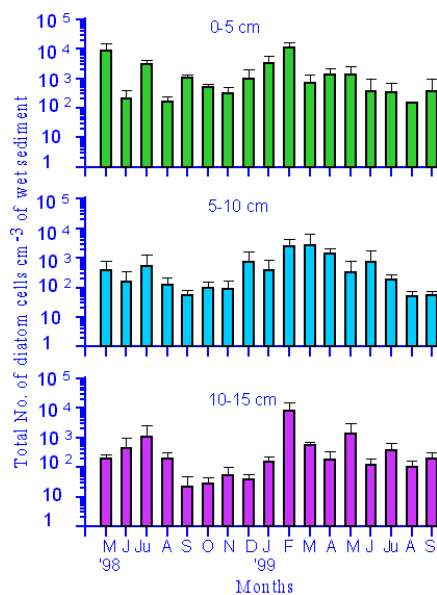


Fig. 2A.8 Temporal variations in the total diatom abundance across the vertical gradient of the core at low tide

Thus, the diatom data when subjected to two-way ANOVA revealed significant variation with respect to months ($P \leq 0.001$), depths (except for *Skeletonema* and *Streptotheca*) and also the month X depth interaction (except for *Skeletonema*) (Table 2A.3).

Table 2A.3 Significance values of the two way ANOVA evaluating the variations in diatom abundance with respect to months and core sections (0-15 cm) (NS = not significant)

Diatoms in the intertidal sediment	Month $p \leq$	Core section $p \leq$	Month x Core section $p \leq$
<i>Navicula</i>	0.001	0.005	0.05
<i>Amphora</i>	0.001	0.005	0.001
<i>Grammatophora</i>	0.001	0.005	0.001
<i>Pleurosigma</i>	0.001	0.025	0.001
<i>Nitzschia</i>	0.001	0.01	0.001
<i>Thalassiothrix</i>	0.001	0.001	0.001
<i>Cocconeis</i>	0.001	0.001	0.001
<i>Fragilaria</i>	0.001	0.005	0.001
<i>Bellerochea</i>	0.001	0.001	0.001
<i>Thalassiosira</i>	0.001	0.001	0.005
<i>Biddulphia</i>	0.001	0.001	0.001
<i>Coscinodiscus</i>	0.001	0.005	0.001
<i>Melosira</i>	0.001	0.001	0.001
<i>Skeletonema</i>	0.001	NS	NS
<i>Streptotheca</i>	0.001	NS	0.001
<i>Chaetoceros</i>	0.001	0.005	0.001

Diatom diversity was found to decrease drastically with a low evenness value in July at the 0-5 cm depth, as *Thalassiosira* dominated the community. During the other

Table 2A.4 Two way ANOVA for the diatom (a) diversity (b) generic richness and (c) evenness of the sediment with respect to months and core section (* = $p \leq 0.001$; ** = $p \leq 0.005$; *** = $p \leq 0.01$)

	df	SS	MS	Fs
(a)				
Months	16	23.58	1.47	5.28*
Core Section	2	3.29	1.64	5.88**
Months X Core Section	32	24.15	0.75	2.7*
Within subgroup error	102	28.5	0.28	
Total	152	79.52		
(b)				
Months	16	21.04	1.31	11.41*
Core Section	2	6.15	3.07	26.7*
Months X Core Section	32	24.6	0.77	6.67*
Within subgroup error	102	11.76	0.11	
Total	152	63.54		
(c)				
Months	16	1.39	0.087	2.81*
Core Section	2	0.3	0.15	4.81***
Months X Core Section	32	2.07	0.065	2.10**
Within subgroup error	102	3.15	0.03	
Total	152	6.9		

Table 2A. 5 Values of species diversity (H'), species richness ($H' \max.$) and evenness (J') of the diatom community at the low tide zone (Shannon-Wieners diversity index)

MONTHS	0-5 cm			5-10 cm			10-15 cm		
	H'	$H' \max.$	J'	H'	$H' \max.$	J'	H'	$H' \max.$	J'
May '98	1.71	3.32	0.52	1.15	2.32	0.49	2.4	2.81	0.85
June	2.12	3	0.71	1.85	2.32	0.8	0.24	2	0.12
July	0.41	3	0.14	1.24	2.58	0.48	1.29	2.81	0.46
August	2.22	3	0.74	3.03	3.32	0.91	2.49	3	0.83
September	1.76	2.81	0.62	1.87	2.32	0.8	1.07	1.58	0.67
October	1.7	2.81	0.6	2.29	2.58	0.88	1.89	2	0.94
November	1.51	2.58	0.58	1.26	2.32	0.55	1.85	2	0.92
December	2.1	3.17	0.66	1.31	2.32	0.56	0.85	1	0.85
January '99	2.02	3.17	0.64	2.7	2.81	0.96	2.1	3	0.71
February	1.74	3.32	0.52	2.37	3	0.79	1.31	3	0.44
March	2.3	3.17	0.72	1.33	2.81	0.47	1.09	2.32	0.47
April	1.78	2.81	0.63	2.07	3	0.7	1.39	1.58	0.88
May	1.75	2.32	0.75	0.7	1.58	0.44	2.11	2.58	0.82
June	1.1	2.32	0.48	0.8	2.58	0.31	1.9	2.32	0.82
July	0.35	1.58	0.22	1.76	2.32	0.76	1.7	2.32	0.73
August	1.54	2.58	0.6	1.73	2.32	0.74	1.82	2.32	0.78
September	1.51	2.58	0.58	2.41	2.81	0.86	1.93	2.8	0.69

months the diversity in the 0-5 cm stratum ranged between 1.5 and 2.3, while at 5-10 and 10-15 cm depths it fluctuated between 0.2 and 2.5. Thus, diatom diversity (H'), richness ($S = H' \max.$) and evenness ($J' = H'/H' \max.$) revealed significant variation

with respect to months ($P \leq 0.001$), depths, as well as months X depths (Table 2A.4 and Table 2A.5).

The dendrogram at 50% dissimilarity level revealed 5 groups of diatoms in the three

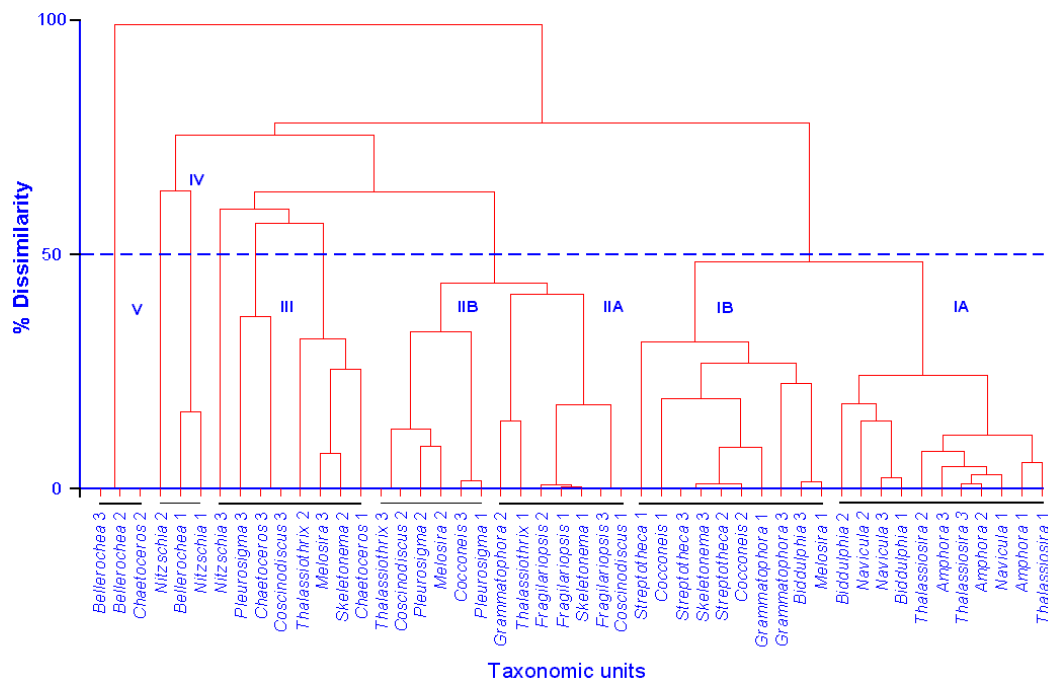


Fig. 2A.9a Cluster dendrogram of diatoms across the vertical gradient of the core using the Bray-Curtis coefficient and group average method.

sediment core sections (Fig. 2A.9a). Group I comprised two sub-groups, IA and IB. Sub-group IA was represented by the most abundant diatoms, i.e. *Thalassiosira*, *Amphora* and *Navicula* for the entire core. *Biddulphia* from the 0-5 and 10-15 cm core sections also formed a member of this sub-group. The abundance of these diatoms ranged up to 10^4 cells cm^{-3} wet sediment. The second sub-group (IB) included those forms present at particular depths up to an abundance of 10^2 cells cm^{-3} wet sediment. Group IIA comprised forms occurring rarely and with abundance below 10^2 cells cm^{-3} wet sediment at particular depths during the study period, and group IIB included forms such as *Cocconeis* (10-15 cm) and *Pleurosigma* (0-5 cm) with

abundance ranging between 10^2 and 10^3 cells cm^{-3} wet sediment. The abundance of rest of the forms belonging to this group was below 10^2 cells cm^{-3} wet sediment.

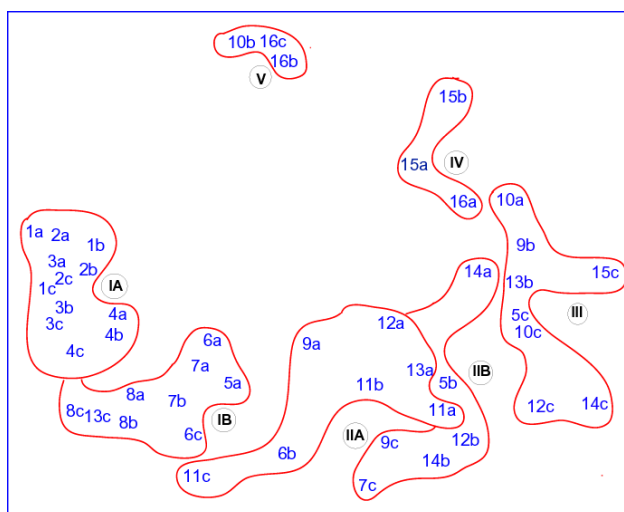


Fig. 2A.9b Non-metric Multidimensional Scaling ordination of diatoms across the vertical gradient of the core using the Bray-Curtis coefficient and group average method.

1. *Thalassiosira*; 2. *Amphora*; 3. *Navicula*; 4. *Biddulphia*; 5. *Melosira*; 6. *Grammatophora*; 7. *Cocconeis*; 8. *Streptothecha*; 9. *Thalassiothrix*; 10. *Chaetoceros*; 11. *Fragilaria*; 12. *Coscinodiscus*; 13. *Skeletonema*; 14. *Pleurosigma*; 15. *Nitzschia*; 16. *Bellerochea*

Group III included the depths at which the respective forms occurred only once or twice during the study period. Forms with an abundance ranging between 1 and 10 cells cm^{-3} wet sediment clustered to form group IV. Group V represented the forms, at particular depths, that were absent throughout the study period. The diatom distribution portrayed a similar pattern as that of the cluster analysis when subjected to two-dimensional NMDS ordinations (Fig. 2A.9b).

Relationship with observed interstitial water parameters: Multiple regressions were performed on the nutrient (nitrogen, phosphorus, silicate), temperature and salinity

Table 2A.6 Partial regression coefficients (Beta values) for the dependent variable (viable diatoms cm^{-3} wet sediment) and independent variable (water parameters)

Diatoms	Intercept	R	Temperature($^{\circ}\text{C}$)	Salinity(psu)	Nitrate ($\text{NO}_3\text{-N}$)	Nitrite($\text{NO}_2\text{-N}$)	Phosphate($\text{PO}_4\text{-P}$)	Silicate(SiO_3)
Pennate	1.12	0.8	0.186	0.135	0.086	0.057	0.41	-0.84*
Centric	-2.55	0.87	0.307	0.212	0.69*	-0.18	0.83*	-0.8*

values (obtained for the entire 15 cm core) versus the total count of pennate and centric diatoms (0-15 cm). It was observed that, for pennates, which mainly reside in the sediments, nutrients did not play an important role, whereas, for centric diatoms, nitrate and phosphate positively influenced their abundance (Table 2A.6).

Relationship with grain size: Distribution of different grain size fractions was found to differ with the different depths, as well as the different months (Fig. 2A.5).

Table 2A.7 Partial regression coefficients (Beta values) for the dependent variable (viable diatoms cm⁻³ wet sediment) and independent variable (grain size) (I = 0-5; II = 5-10; III = 10-15 cm)

Depth	Diatoms	Intercept	R	1000	500	250	125	63	<63
I	<i>Navicula</i>	-13.7	0.84	-1.02 *	0.45	0.38	1.71	1.71	2.97
	<i>Amphora</i>	9.74	0.88	-0.29	-0.28	0.68 *	-0.29	-0.99	-0.43
	<i>Grammatophora</i>	-4.48	0.71	-0.36	-0.21	0.96 *	0.42	0.49	1.11
II	<i>Pleurosigma</i>	-0.025	0.74	0.222	-0.42	-0.154	0.87*	-0.456	0.681 *
	<i>Thalassiothrix</i>	-0.004	0.87	-0.028	-0.23	0.8 *	-0.39	0.28	-0.19
III	<i>Fragilaria</i>	2.07	0.85	-0.11	0.074	-0.253	-0.235	-0.668*	-0.082

However, subsection of this data to multiple regression analysis indicated that the grain size could serve as a predictor of only some of the pennate diatoms, e.g. *Amphora*, *Grammatophora*, *Pleurosigma* and *Thalassiothrix* (Table 2A.7). Amongst these, the 250µm grain size fraction served as a predictor of the abundance of *Amphora* and *Grammatophora* at 0-5 cm depth. *Pleurosigma* was influenced by 125 and < 63 µm grain size at 5-10 cm, whereas, for *Thalassiothrix*, the choice of substratum was the 250 µm grain size at the same depth. However, a similar substratum preference of these diatoms at the other depths was not evident. *Navicula*, which was abundant throughout the year, was negatively influenced by the 1000 µm grain size fraction at 0-5 cm. However, the percentage composition of this grain size was negligible. *Fragilariopsis* was negatively influenced by 63µm grain size at the lowermost depth (10-15 cm).

Wind speed and suspended load: Wind speed in the study area ranged from 2.0 to 4.7 m s^{-1} . Regression analysis between suspended matter and wind speed was statistically

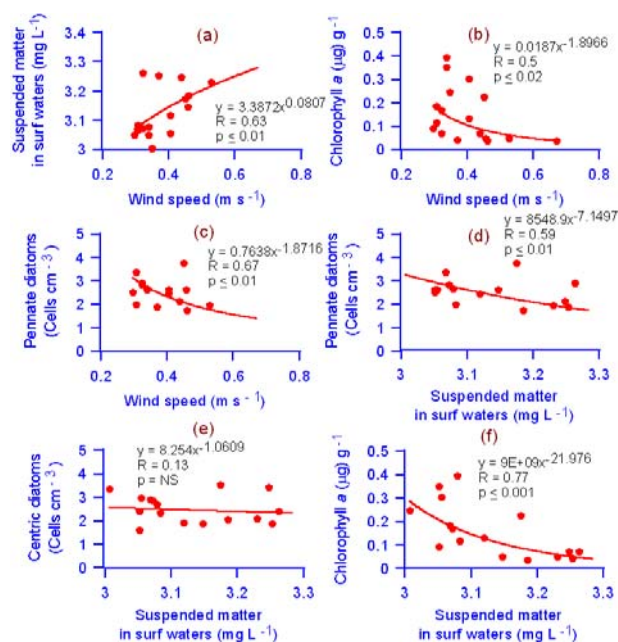


Fig. 2A.10 Polynomial regression between wind speed and suspended matter (a), chlorophyll *a* (b), and pennate diatom abundance (c); and between suspended matter and pennate diatom abundance (d), centric diatom abundance (e) and chlorophyll *a* (f) (NS = not significant)

significant. Chlorophyll *a* concentrations and pennate diatom abundance in the 0-5 cm stratum revealed a significant negative correlation between wind speed and suspended load (Fig. 2A.10).

Chlorophyll *a* and diatoms: During monsoons, chlorophyll *a* concentrations were lower irrespective of depth; the highest concentration in this season was observed in the 10-15 cm stratum. During late post-monsoon and pre-monsoon seasons, the 0-5 cm stratum had higher concentrations of chlorophyll *a*; these peaked during December and March (Fig. 2A.11). Regression analysis revealed that the chlorophyll *a* concentrations at 0-5 and 5-10 cm depths were good predictors of pennate as well as centric diatoms, whereas, for the third depth (10-15 cm), this was not the case (Fig. 2A.12). Although the regression analysis showed a negative correlation of diatom

abundance with chlorophyll *a* at 10-15 cm depth, the appearance of diatoms in the culture wells provided with growth media and light (prerequisite factors for photosynthesis) confirms the presence of functional chlorophyll *a* at this depth.

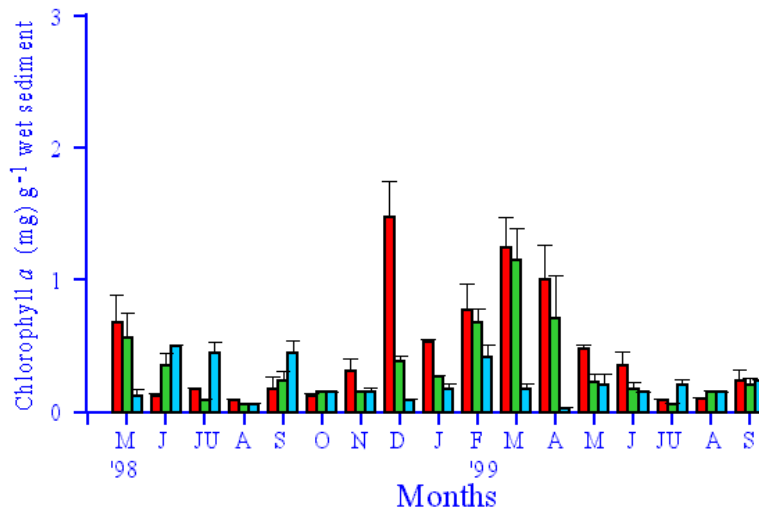


Fig. 2A.11 Concentrations of chlorophyll *a* across the three vertical sections of the sediment core at low tide zone
 ■ 0 – 5 cm; ■ 5 – 10 cm; ■ 10 – 15 cm

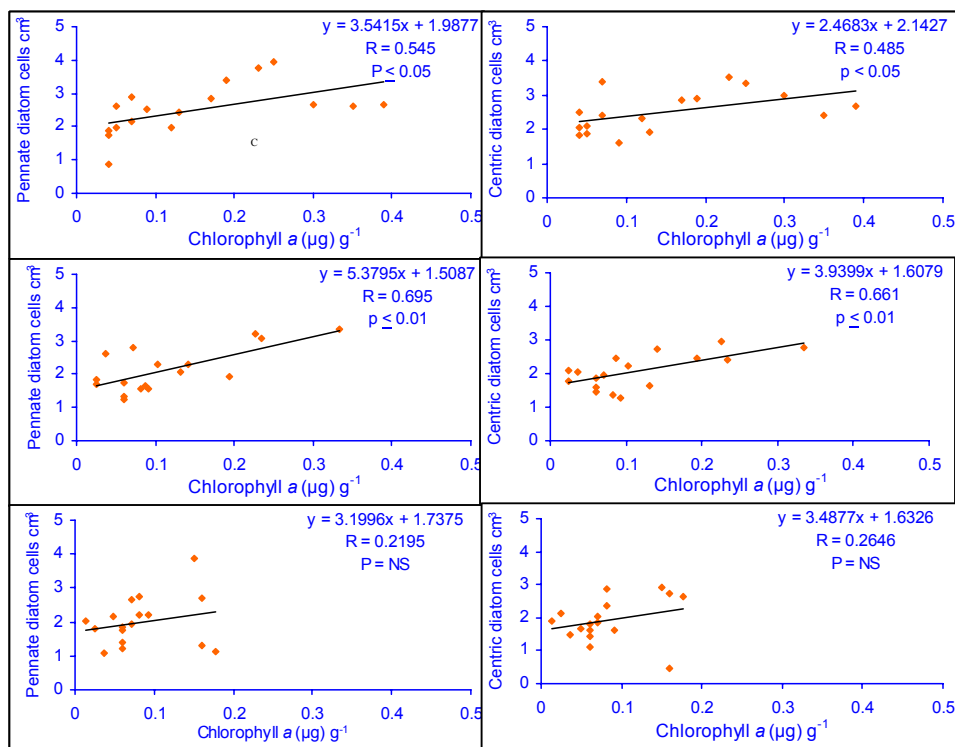


Fig. 2A.12 Linear regression between chlorophyll *a* and pennate and centric diatom abundance across the three vertical sections of the core (NS = not significant)

2A.3.4 Mid tide zone

Sediment characteristics: Fine sand (125-249 μm) was found to be the most abundant grain size fraction of the sediment throughout the study period across the 15 cm core (Fig. 2A.13b). The contribution of sediment particles $> 250 \mu\text{m}$ was negligible. MANOVA of different depth versus grain size revealed a significant variation (Table 2A.8b).

Diatoms in sediment: The sediment harbored 30 species of diatoms (11 centric, 19 pennate) belonging to 20 (7 centric, 13 pennate) genera (Table 2A.1c). Pennate diatoms were dominant in terms of abundance (Fig. 2A.14b). The most abundant pennate diatoms were *Amphora* and *Navicula*, whereas, *Thalassiosira* was the abundant centric diatom. Centric diatoms such as *Skeletonema* and *Chaetoceros*, recorded at the low tide zone were not observed at this zone.

Based on their abundance diatoms encountered in this area can be divided into three groups as given in Table 2A.9 and Figs. 2A.15a-c.

Temporal and Vertical distribution of diatoms in the sediment: At the 0-5 cm stratum the total diatom abundance peaked in November, January and March whereas at the 5-15 cm stratum not much variation was observed. Amongst the group I diatoms, *Navicula* peaked from November to March across the three core sections; *Amphora* and *Thalassiosira* peaked only in the 0-5 cm depth, during the same period. Their distribution in the 5-15 cm depth did not show any particular trend. Amongst the Group II diatoms, in the 0-5 cm, *Melosira* was most abundant from July to March, *Biddulphia* and *Nitzschia* from September to March and *Pleurosigma*, *Thalassiothrix* and *Cyclotella* from November to March. In the group III diatoms, *Bellerochea* peaked from November to March, *Fragilariopsis* in September, *Grammatophora* and *Coscinodiscus* in July and *Streptothecca* in September.

Thus, all but *Bellerochea*, *Raphoneis* (depth and month x depth) and *Thalassiothrix* (depth) showed significant variation when subjected to two-way ANOVA (Table 2A.10b).

Diatom diversity peaked in July with a high evenness due to an evenly distributed population (Table 2A.11a). Significant variation with respect to month ($P \leq 0.001$), depth as well as month x depth was observed for diatom diversity (H'), richness ($S = H' \max.$; except with respect to depth) and evenness ($J' = H'/H' \max.$) (Table 2A.12b).

The dendrogram at 50% dissimilarity level revealed 7 groups of diatoms (Fig. 2A.16b). The first group comprised two pennate diatoms, *Amphora* and *Navicula*, and a centric diatom *Thalassiosira*, which were the most abundant forms. Abundance of these diatoms ranged up to 10^4 cells cm^{-3} wet sediment. Group II was represented by forms occurring 4-7 times, with an abundance ranging between 10^1 and 10^2 cells cm^{-3} wet sediment. Group III comprised forms with an abundance ranging between 10 and 10^3 cells cm^{-3} wet sediment and which mostly occurred 3 times during the sampling period. The forms belonging to group IV occurred once during the sampling period with an abundance ranging between 10^1 and 10^2 cells cm^{-3} wet sediment. Forms, which occurred twice and thrice during the sampling period comprised group V and group VI respectively. Group VII was represented by forms, which had an abundance ranging between 10 and 10^2 cells cm^{-3} wet sediment.

The diatom distribution portrayed a similar pattern as that of the cluster analysis when subjected to 2D NMDS ordinations (Fig. 2A.17b).

Relationship with grain size: Distribution of different grain size fractions was found to differ with the different depths (Fig. 2A.13b). Multiple regression analysis indicated that the grain size could serve as a predictor of diatoms such as

Grammatophora, *Coscinodiscus*, *Biddulphia*, *Bellerochea*, *Navicula*, *Cyclotella*, *Thalassiothrix*, *Raphoneis*, *Streptothea* and *Pleurosigma* (Table 2A.13a). Amongst these, the < 63 µm grain size fraction served as a predictor of the abundance of *Grammatophora* and *Coscinodiscus* at 0-5 cm depth. *Biddulphia* and *Bellerochea* were influenced by 500 and < 63 µm grain size at 5-10 cm. At the 10-15 cm depth for *Navicula*, the choice of substratum was the 125, 63 and < 63 µm grain size whereas for *Cyclotella*, *Thalassiothrix*, *Bellerochea*, *Raphoneis* and *Pleurosigma* 500 µm grain size served as a predictor.

Wind speed: Regression analysis between wind speed and parameters such as chlorophyll *a*, pennate and centric diatom abundance did not reveal any significant relation.

Chlorophyll *a* and diatoms: Chlorophyll *a* concentrations were higher at the 0-5 cm stratum with peaks from September through March (Fig. 2A.18b). Regression analysis revealed that the chlorophyll *a* concentrations only at 0-5 cm depth were good predictors of pennate as well as centric diatoms (Fig. 2A.19b).

2A.3.5 High tide zone

Sediment characteristics: Fine sand (125-249 µm) was found to be the most abundant grain size fraction of the sediment throughout the study period across the 15 cm core (Fig. 2A.13c). The contribution of sediment particles > 250 µm was negligible. MANOVA of different months versus grain size revealed a significant variation (Table 2A.8c).

Diatoms in sediment: 27 species of diatoms (10 centric, 17 pennate) belonging to 19 (7 centric, 12 pennate) genera were recorded in the sediment (Table 2A.1d). Pennate diatoms were dominant in terms of abundance (Fig. 2A.14c). The most abundant pennate diatoms were *Amphora* and *Navicula*, whereas, *Thalassiosira* was the

abundant centric diatom. Centric diatoms such as *Skeletonema* and *Chaetoceros*, recorded at the low tide zone were not observed at this location.

Based on their abundance, diatoms encountered in this area can be divided into three groups as given in Table 2A.9 and Figs. 2A.15a-c.

Temporal and Vertical distribution of diatoms in the sediment: The total diatom abundance peaked in September, November, January and March at the 0-5 cm stratum whereas the 5-10 cm stratum did not exhibit much variation. Amongst the Group I diatoms, *Amphora* and *Thalassiosira* peaked from September through March whereas *Navicula* peaked from November through March in the 0-5 cm depth. Amongst the Group II diatoms, peaks were observed for *Melosira* from July through September, *Cocconeis* and *Thalassiothrix* from November through March and *Streptothecca* from September through March. Group III diatoms such as *Nitzschia*, *Grammatophora* and *Achnanthes* peaked in September, January and July respectively.

Thus, the diatom data when subjected to two-way ANOVA revealed a significant variation with respect to months ($p \leq 0.001$) and depths for all forms except *Pinnularia*, which did not reveal significant relationship with respect to month x depth (Table 2A.10c).

The diatom diversity was lower as compared to the low and mid tide zones (Table 2A.11b). Significant variation with respect to months ($p \leq 0.001$), depths as well as months x depths was observed for diatom diversity (H'), richness ($S = H' \text{ max.}$) and evenness ($J' = H'/H' \text{ max.}$) (Table 2A.12c).

The dendrogram at 50% dissimilarity level revealed 5 groups of diatoms respectively (Fig. 2A.16c). The first group comprised two pennate diatoms, *Amphora* and *Navicula*, and a centric diatom *Thalassiosira*, which were the most abundant forms, except for *Thalassiosira* in the 0-5 cm section. Abundance of these diatoms ranged up

to 10^3 cells cm^{-3} wet sediment. Group II comprised forms, which occurred 3-7 times during the sampling period. Forms, which occurred once represented group III whereas those which, occurred 1-3 times during the sampling period, represented group IV. Group V comprised forms occurring 1-2 times during the sampling period with an abundance ranging between 10 and 10^2 cells cm^{-3} wet sediment.

The diatom distribution portrayed a similar pattern as that of the cluster analysis when subjected to 2D NMDS ordinations (Fig. 2A.17c).

Relationship with grain size: Distribution of different grain size fractions was found to differ with the different months (Fig. 2A.13c). Subjection of this data to multiple regression analysis indicated that the grain size could serve as a predictor of *Grammatophora* (500 μm grain size fraction) at 0-5 cm, and *Achnanthes*, *Biddulphia*, *Melosira* and *Streptotheca* (250 μm grain size fraction), *Cyclotella* (500 and 125 μm grain size fraction), *Pinnularia* (63 μm grain size fraction) and *Coscinodiscus* (500, 125 and 63 μm grain size fraction) at the 10-15 cm (Table 2A.13b).

Wind speed: Regression analysis between wind speed and parameters such as chlorophyll *a*, pennate and centric diatom abundance did not reveal any significant relation.

Chlorophyll *a* and diatoms: Chlorophyll *a* concentrations generally followed a low profile except in the 10-15 cm stratum during July (Fig.2A.18c). Regression analysis did not reveal significant relationship between chlorophyll *a* concentrations and pennate and centric diatoms (Fig. 2A.19c).

2A.3.6 Comparison between the low, mid and high tide zones

Sediment characteristics: While very fine sand (63-124 μm - silt/clay) was found to be the most abundant grain size fraction of the sediment at the low tide zone, it was fine sand (125-249 μm) which dominated at both, the mid and high tide zones throughout the study period across the 15 cm core. Exception was in July at the mid tide zone at 10-15 cm where 63-124 μm grain size fraction dominated and at high tide zone during May at 5-15 cm depth (Table 2A.8a-c and Fig. 2A.13a-c).

Table 2A.8a. MANOVA (Without replication) for the temporal variation in the sediment characteristics along the vertical gradients at the low tide mark (0-5, 5-10 and 10-15 cm sediment core sections) (* $p \leq 0.05$; NS = not significant)

Source of variation	df	SS	MS	Fs
Months	8	7.62	0.95	-
Core sections	2	90.3	45.15	-
% Grain size fractions	5	34989.15	6997.83	-
Months X Core sections	16	684.8	42.8	0.83 ^{NS}
Months X % Grain size fractions	40	3062.23	76.5	1.5*
Core sections X % Grain size fractions	10	833.97	83.39	1.63 ^{NS}
Months X Core sections X % Grain size fractions	80	4085.62	51.07	-

Table 2A.8b. MANOVA (Without replication) for the temporal variation in the sediment characteristics along the vertical gradients at the mid tide mark (0-5, 5-10 and 10-15 cm sediment core sections) (* $p \leq 0.05$; NS = not significant)

Source of variation	df	SS	MS	Fs
Months	8	4.68	0.58	-
Core sections	2	5.56	2.78	-
% Grain size fractions	5	71,107.24	14,221.45	-
Months X Core sections	16	22.97	1.44	0.061 ^{NS}
Months X % Grain size fractions	40	745.02	18.63	0.79 ^{NS}
Core sections X % Grain size fractions	10	458.84	45.88	1.94*
Months X Core sections X % Grain size fractions	80	1,891.74	23.65	-

Table 2A.8c MANOVA (Without replication) for the temporal variation in the sediment characteristics along the vertical gradients at the high tide mark (0-5, 5-10 and 10-15 cm sediment core sections) (***) $p \leq 0.001$; NS = not significant)

Source of variation	df	SS	MS	Fs
Months	8	43.87	5.48	-
Core sections	2	0.65	0.32	-
% Grain size fractions	5	75,960.36	15,192.07	-
Months X Core sections	16	41.8	2.61	0.097 ^{NS}
Months X % Grain size fractions	40	2,773.48	69.33	2.56***
Core sections X % Grain size fractions	10	187.59	18.76	0.69 ^{NS}
Months X Core sections X % Grain size fractions	80	2164.14	27.05	-

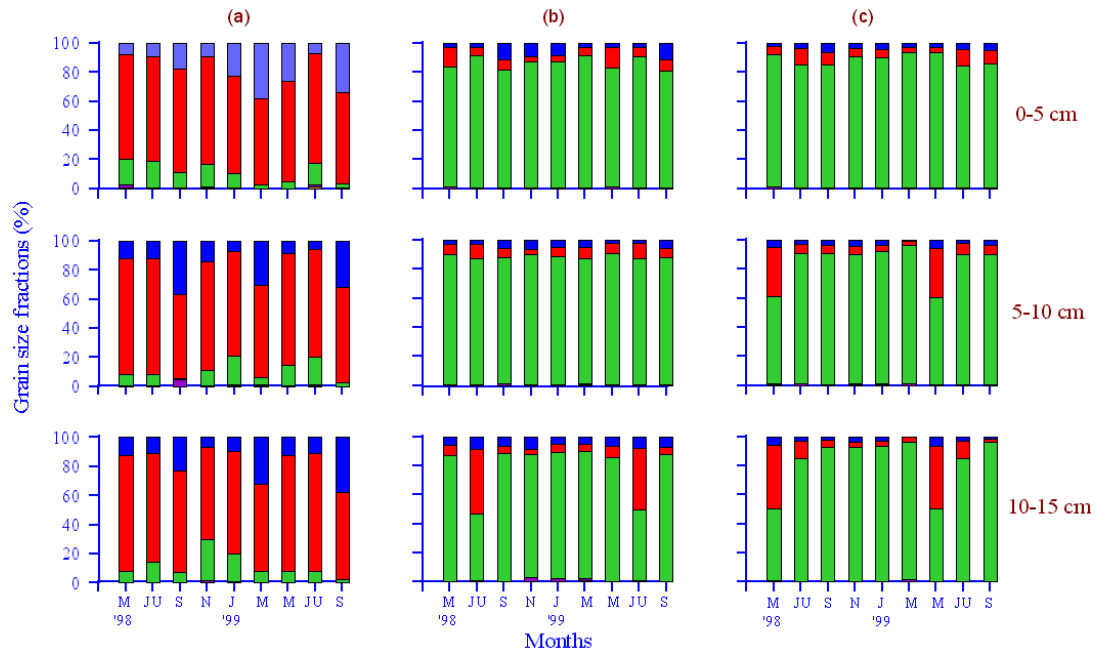


Fig. 2A.13 Temporal variations in grain size fractions (> 1000 μm , 500-999 μm , 250-499 μm , 125-249 μm , 63-124 μm and < 63 μm) across the vertical gradient of the core at the low tide zone (a); mid tide zone (b) and high tide zone (c)

Diatoms in intertidal sediments: Diatoms were encountered up to 15 cm depth throughout the intertidal zone. The pennates dominated over the centrics throughout

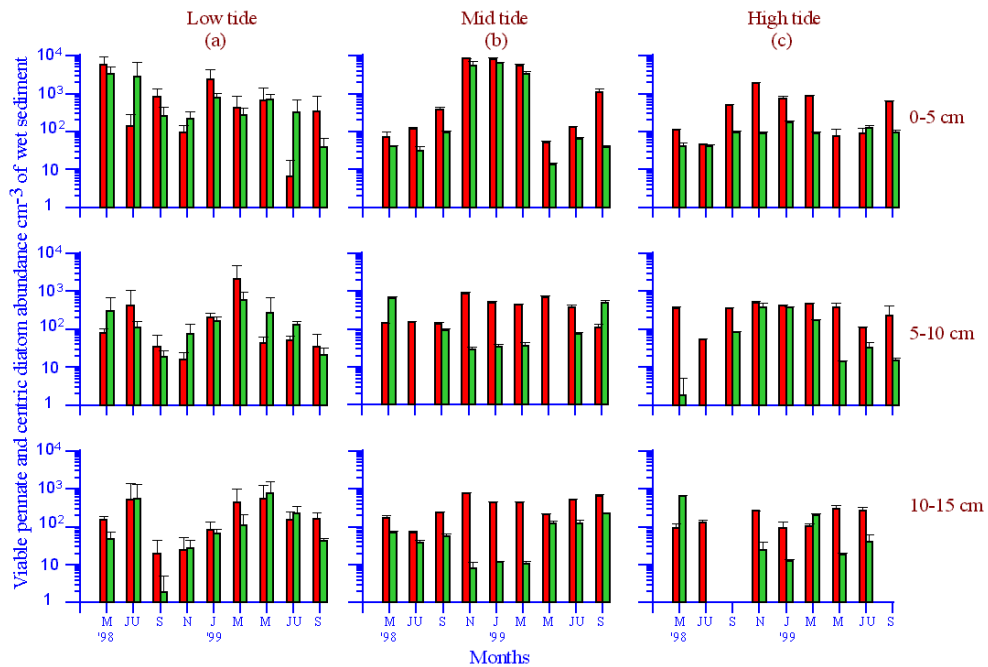


Fig. 2A.14 Temporal variations in pennate and centric diatom abundance across the vertical gradient of the core
■ Pennate diatoms; ■ Centric diatoms

the intertidal zone (Fig. 2A.14a-c). The dominant diatoms which comprised group I were common to all the three tidal zones whereas those which comprised group II and III differed with the tidal zone. The highest number of species was encountered at the mid tide zone followed by the high and low tide zones (Figs. 2A.7a-b and 2A.15a-c). The abundance, diversity, richness and evenness of diatom cells showed spatial and

Table 2A.9 Diatom groups encountered at the low, mid and high tide zones
(a = low tide, b =mid-tide and c =high tide)

Diatoms	Group I			Group II			Group III		
	a	b	c	a	b	c	a	b	c
<i>Amphora</i>	+	+	+						
<i>Navicula</i>	+	+	+						
<i>Thalassiosira</i>	+	+	+						
<i>Biddulphia</i>				+	+				+
<i>Melosira</i>				+	+	+			
<i>Streptotheca</i>				+		+			
<i>Coscinodiscus</i>				+				+	+
<i>Cyclotella</i>					+				+
<i>Grammatophora</i>				+				+	+
<i>Cocconeis</i>				+	+	+			
<i>Nitzschia</i>				+	+				+
<i>Thalassiothrix</i>				+	+	+			
<i>Pleurosigma</i>					+		+		
<i>Fragilariopsis</i>							+	+	+
<i>Skeletonema</i>							+		
<i>Chaetoceros</i>							+		
<i>Bellerochea</i>							+	+	+
<i>Cymbella</i>					+				+
<i>Achnanthes</i>								+	+
<i>Streptotheca</i>								+	
<i>Raphoneis</i>								+	+
<i>Pinnularia</i>									+
<i>Tabellaria</i>					+	+			

temporal variations throughout the intertidal zone (Tables 2A.10, 2A.11 and 2A.12).

Grain size fractions, which served as predictors of some diatoms differed with the tidal zones and also depth (Table 2A.8a-c and Fig. 2A.13a-c). Wind stimulated the resuspension of the sediment, along with the pennate diatoms down to 5 cm depth only at the low tide zone (Fig. 2A.10). The depth up to which chlorophyll *a* concentrations could serve as predictors of diatom abundance differed with the tidal

level, from 10 cm depth at the low tide zone, to 5 cm depth at the mid tide zone to 0 cm at the high tide zone (Fig. 2A.19a-c).

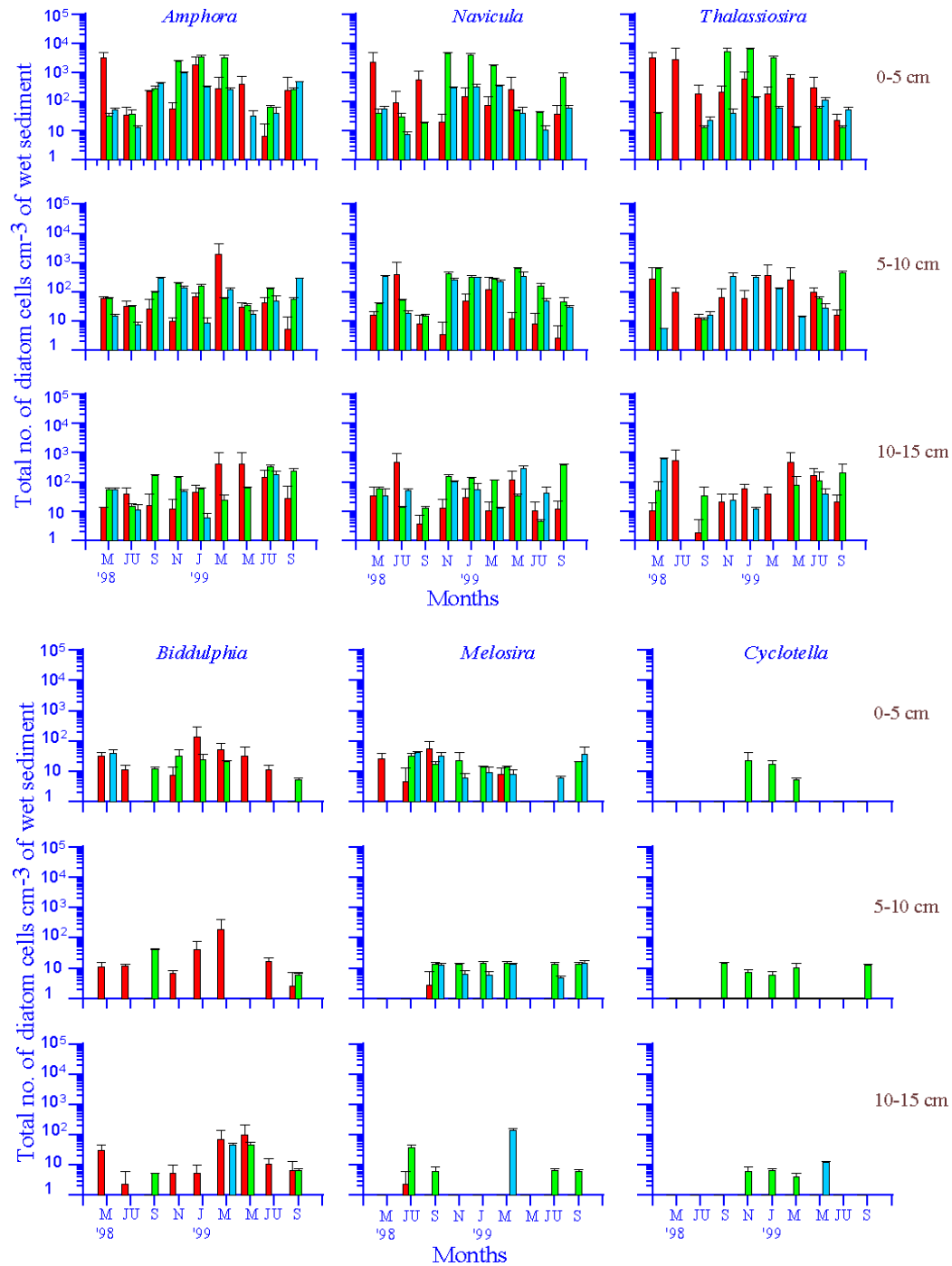


Fig. 2A.15a Temporal variations in diatom abundance across the vertical gradient of the core

■ Low tide zone; ■ Mid tide zone; ■ High tide zone

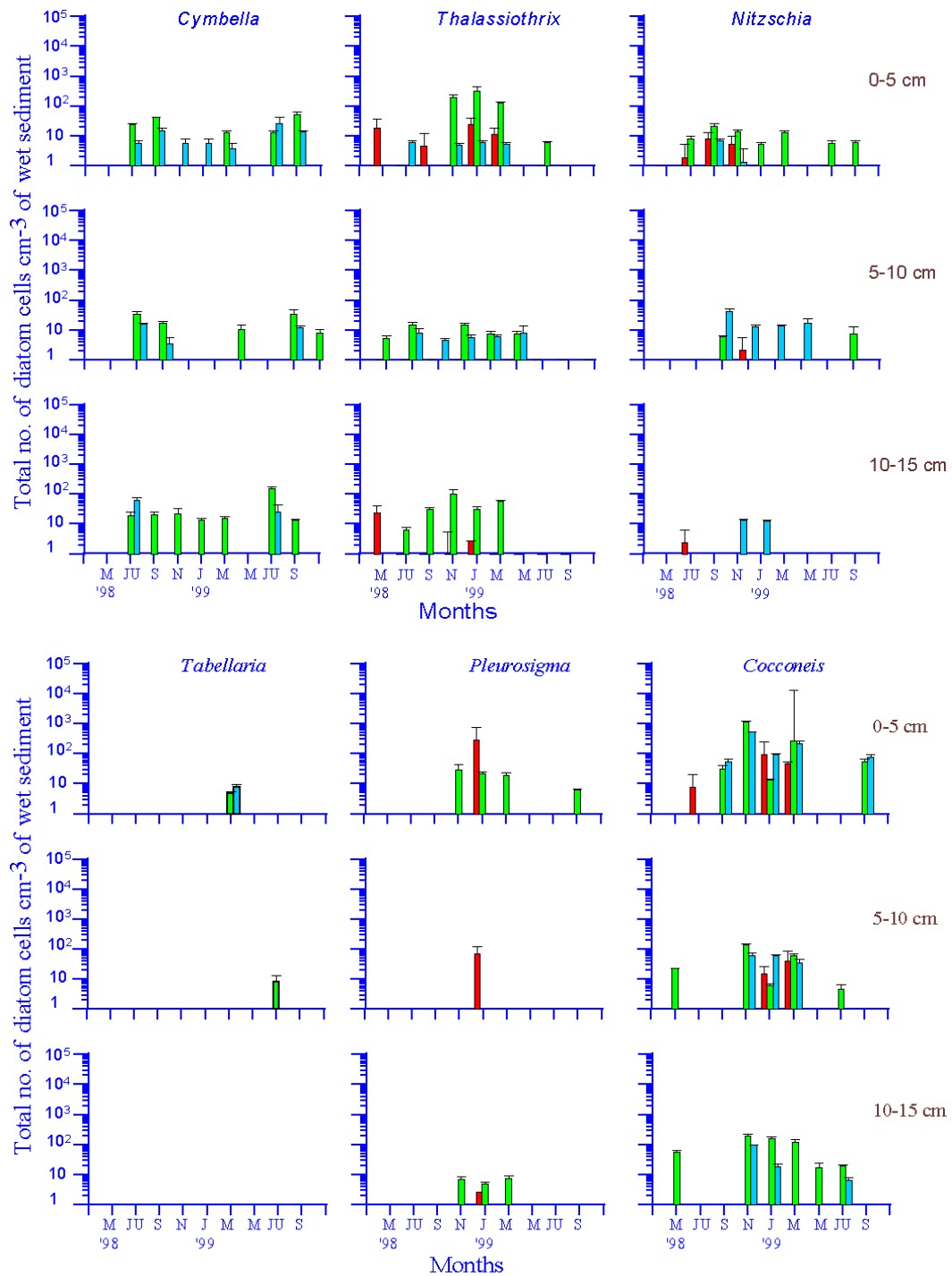


Fig. 2A.15b Temporal variation in diatom abundance across the vertical gradient of the core

■ Low tide zone; ■ Mid tide zone; ■ High tide zone

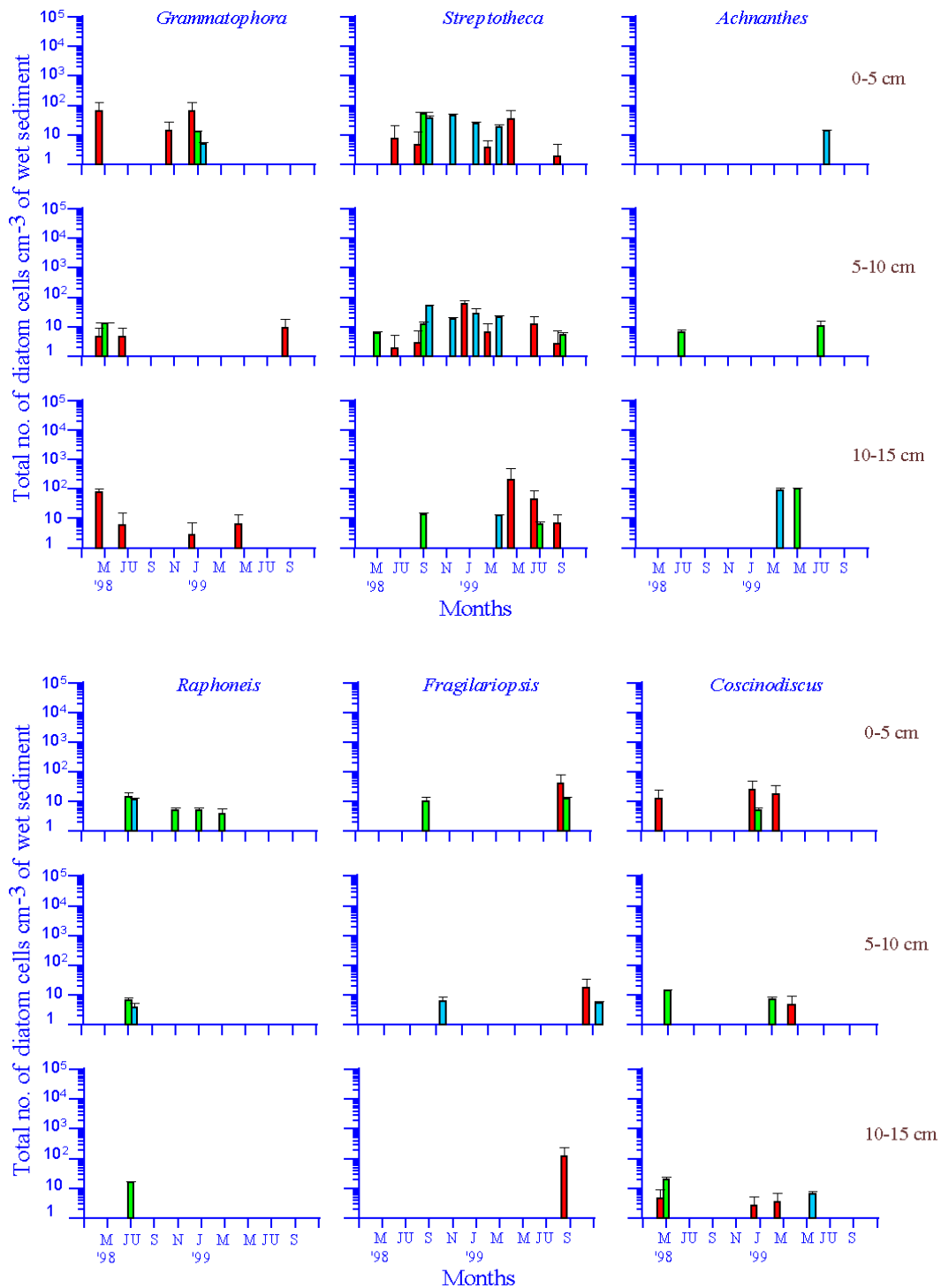


Fig. 2A.15c Temporal variations in diatom abundance across the vertical gradient of the core

■ Low tide zone; ■ Mid tide zone; ■ High tide zone

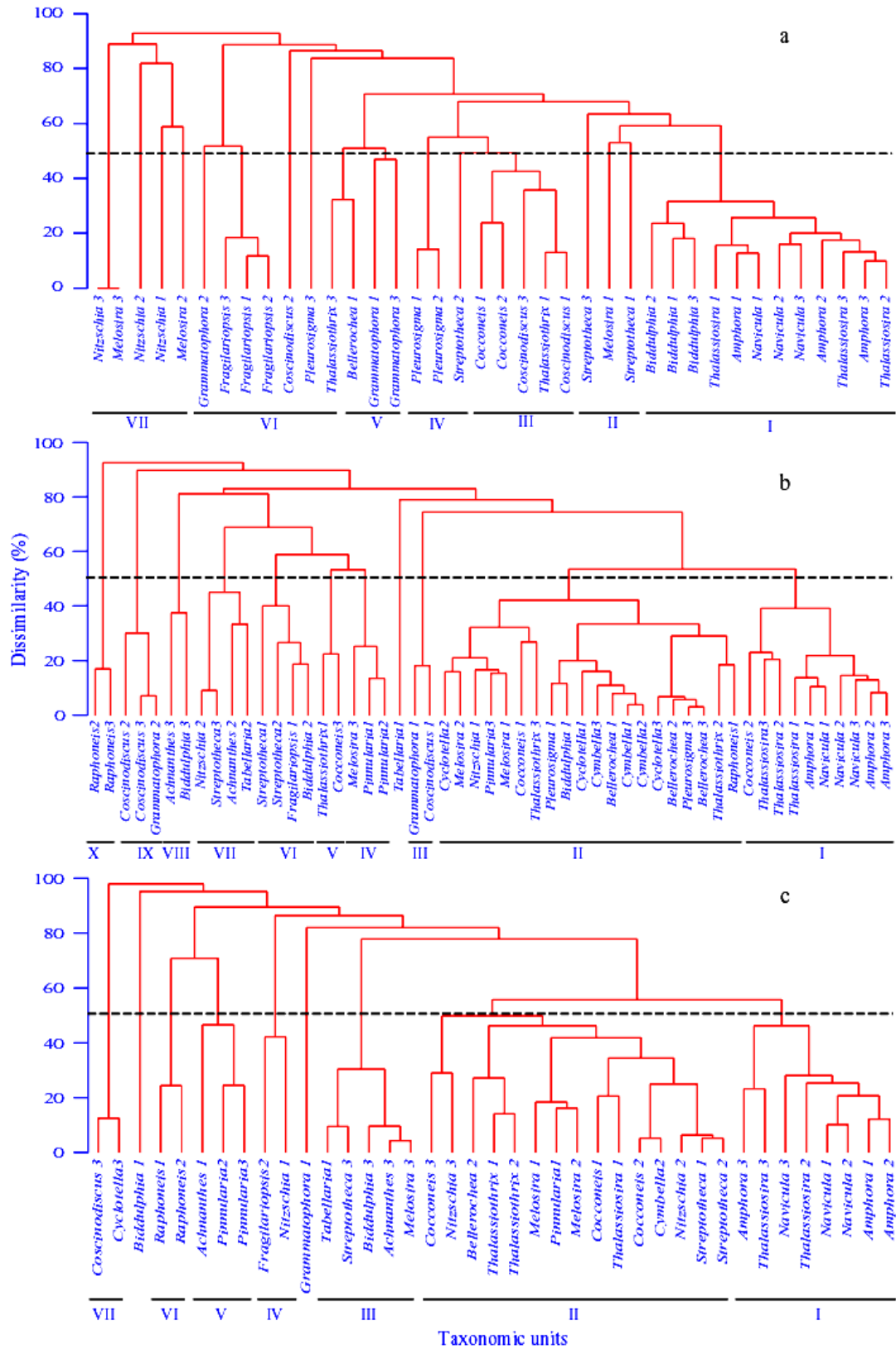


Fig. 2A.16 Cluster dendrogram of diatoms across the vertical gradient of the core using Bray Curtis coefficient and group average method a) low tide, b) mid tide and c) high tide

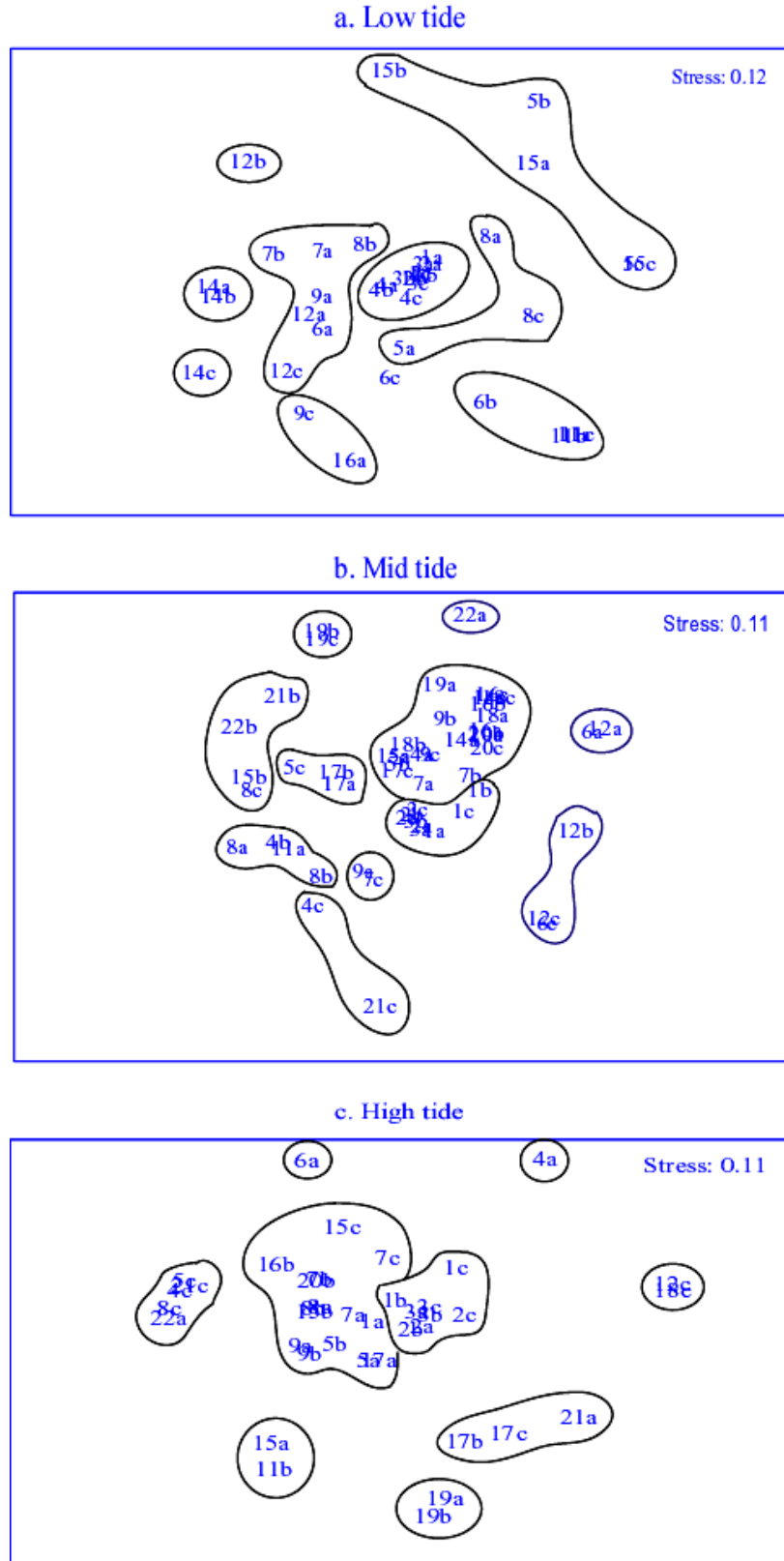


Fig. 2A.17 Multidimensional scaling ordination of diatoms across the vertical gradient based on the Bray Curtis dissimilarity coefficient

1. *Thalassiosira* 2. *Amphora* 3. *Navicula* 4. *Biddulphia* 5. *Melosira* 6. *Grammatophora* 7. *Cocconeis* 8. *Streptothecha* 9. *Thalassiothrix* 10. *Chaetoceros* 11. *Fragilariopsis* 12. *Coscinodiscus* 13. *Skeletonema* 14. *Pleurosigma* 15. *Nitzschia* 16. *Bellerochea* 17. *Pinnularia* 18. *Cyclotella* 19. *Raphoneis* 20. *Cymbella* 21. *Achnanthes* 22. *Tabellaria*

Table 2A.10. Significance values of the two-way ANOVA evaluating the variations in diatom abundance with respect to months and core sections (0-15 cm) at the low tide (a), mid tide (b) and high tide marks (c) (NS = not significant).

Diatoms in the intertidal sediment	Month			Core section			MonthXCore section		
	p ≤			p ≤			p ≤		
	a	b	c	a	b	c	a	b	c
<i>Navicula</i>	0.001	0.005	0.001	0.005	0.001	0.025	0.05	0.025	0.005
<i>Amphora</i>	0.001	0.005	0.005	0.005	0.05	0.001	0.001	0.001	0.001
<i>Grammatophora</i>	0.001	0.001	0.001	0.005	0.001	0.001	0.001	0.001	0.001
<i>Pleurosigma</i>	0.001	0.001	0.001	0.025	0.001	0.001	0.001	0.001	0.001
<i>Nitzschia</i>	0.001	0.001	0.001	0.01	0.001	0.001	0.001	0.001	0.001
<i>Thalassiothrix</i>	0.001	0.001	0.001	0.001	NS	0.001	0.001	0.001	0.001
<i>Cocconeis</i>	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001
<i>Fragilariopsis</i>	0.001	0.001	0.001	0.005	0.001	0.001	0.001	0.001	0.001
<i>Bellerochea</i>	0.001	0.001	0.001	0.001	NS	0.001	0.001	NS	0.001
<i>Thalassiosira</i>	0.001	0.001	0.001	0.001	0.001	0.001	0.005	0.001	0.001
<i>Biddulphia</i>	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001
<i>Coscinodiscus</i>	0.001	0.001	0.001	0.005	0.005	0.001	0.001	0.001	0.001
<i>Melosira</i>	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001
<i>Skeletonema</i>	0.001	-	-	NS	-	-	NS	-	-
<i>Streptotheca</i>	0.001	0.001	0.001	NS	0.005	0.001	0.001	0.001	0.001
<i>Chaetoceros</i>	0.001	-	-	0.005	-	-	0.001	-	-
<i>Cyclotella</i>	-	0.001	0.001	-	0.01	0.001	-	0.001	0.001
<i>Tabellaria</i>	-	0.001	0.001	-	0.001	0.001	-	0.001	0.001
<i>Pinnularia</i>	-	0.001	0.001	-	0.001	0.05	-	0.001	NS
<i>Cymbella</i>	-	0.001	0.001	-	0.001	0.001	-	0.001	0.001
<i>Raphoneis</i>	-	0.001	0.001	-	NS	0.001	-	NS	0.001
<i>Achnanthes</i>	-	0.001	0.001	-	0.001	0.001	-	0.001	0.001

Table 2A. 11a. Values of species diversity (H'), species richness (H' max.) and evenness (J') of the diatom community at the mid tide zone (Shannon-Wieners diversity index)

MONTHS	0-5 cm			5-10 cm			10-15 cm		
	H'	H'max	J'	H'	H'max	J'	H'	H'max	J'
May '98	1.58	1.28	1.00	2.89	2.86	0.96	2.31	1.89	1.00
July	2.56	2.38	0.99	2.54	2.44	0.98	2.56	2.50	0.99
September	3.27	3.36	0.99	3.13	3.27	0.99	2.93	2.97	0.98
November	3.54	3.65	0.96	2.90	2.72	0.96	3.06	2.98	0.97
January '99	3.69	4.28	0.94	3.04	3.25	0.96	3.07	3.15	0.97
March	3.71	4.29	0.95	3.09	3.16	0.98	3.08	3.11	0.97
May	0.97	0.95	0.97	0.94	0.68	0.94	2.57	2.15	0.99
July	2.51	2.39	0.97	3.07	3.16	0.97	2.66	2.54	0.95
September	3.19	3.33	0.96	2.88	2.92	0.96	2.44	2.15	0.94

Table 2A. 11b. Values of species diversity (H'), species richness (H' max.) and evenness (J') of the diatom community at the high tide zone (Shannon-Wieners diversity index)

MONTHS	0-5 cm			5-10 cm			10-15 cm		
	H'	H'max	J'	H'	H'max	J'	H'	H'max	J'
May '98	1.58	1.22	1.00	1.42	1.32	0.90	1.54	1.11	0.97
July	2.54	2.67	0.98	2.28	2.46	0.98	1.55	1.30	0.98
September	2.74	2.51	0.98	2.87	2.98	0.96	-	-	-
November	2.93	3.01	0.92	3.20	3.30	0.96	2.29	1.90	0.99
January '99	3.03	3.03	0.96	3.19	3.38	0.96	2.28	2.20	0.98
March	3.01	3.05	0.95	3.24	3.30	0.98	2.27	1.92	0.98
May	1.00	0.87	1.00	1.49	1.24	0.94	1.43	1.33	0.90
July	2.53	2.37	0.98	2.27	2.10	0.98	2.26	1.94	0.97
September	2.54	2.09	0.98	1.88	1.67	0.94	-	-	-

Table 2A.12. Two-way ANOVA for diatom diversity, richness and evenness with respect to months, and coresctions at the (a) low tide, (b) mid tide and (c) high tide marks

a	<i>df</i>	SS	MS	F _s
Diversity				
Months	16	23.58	1.47	5.28
Core sections	2	3.29	1.64	5.88
MonthsXCore section	32	24.15	0.75	2.7
Within subgroup error	102	28.5	0.28	
Total	152	79.52		
Generic richness				
Months	16	21.04	1.31	11.41
Core section	2	6.15	3.07	26.7
MonthsXCore section	32	24.6	0.77	6.67
Within subgroup error	102	11.76	0.11	
Total	152	63.54		
Evenness				
Months	16	1.39	0.087	2.81
Core section	2	0.3	0.15	4.81
MonthsXCore section	32	2.07	0.065	2.1
Within subgroup error	102	3.15	0.03	
Total	152	6.9		
<hr/>				
b	<i>df</i>	SS	MS	F _s
Diversity				
Months	8	10.66	1.33	12.17
Core sections	2	1.47	0.73	6.7
MonthsXCore section	16	10.75	0.67	6.14
Within subgroup error	54	5.91	0.11	
Total	80	28.78		
Generic richness				
Months	8	6.81	0.85	31.29
Core section	2	0.14	0.07	2.6
MonthsXCore section	16	2.18	0.14	5.01
Within subgroup error	54	1.47	0.03	
Total	80	10.61		
Evenness				
Months	8	0.84	0.11	8.57
Core section	2	0.3	0.15	12.34
MonthsXCore section	16	1	0.06	5.08
Within subgroup error	54	0.66	0.01	
Total	80	2.8		
<hr/>				
c	<i>df</i>	SS	MS	F _s
Diversity				
Months	8	24.72	3.09	33.54
Core sections	2	7.36	3.68	39.93
MonthsXCore section	16	8.35	0.52	5.67
Within subgroup error	54	4.97	0.09	
Total	80	45.4		
Generic richness				
Months	8	7.09	0.89	23.2
Core section	2	3.64	1.82	47.61
MonthsXCore section	16	2.87	0.18	4.7
Within subgroup error	54	2.06	0.04	
Total	80	15.67		
Evenness				
Months	8	2.38	0.3	10.85
Core section	2	0.72	0.36	13.08
MonthsXCore section	16	2.62	0.16	5.96
Within subgroup error	54	1.48	0.03	
Total	80	7.2		

Table 2A.13a. Partial regression coefficients (Beta values) for the dependent variable (viable diatoms cm⁻³ wet sediment) and independent variable (grain size) (I = 0-5 cm; II = 5-10 cm; III = 10-15 cm) at mid tide zone (* p < 0.05; **p < 0.01; ***p < 0.005; ****p < 0.001)

Depth	Diatoms	Intercept	R	1000	500	250	125	63	< 63
I	<i>Navicula</i>	-1.14	0.94	-	-0.99*	-1.29	0.43	1.23	0.11
	<i>Biddulphia</i>	2.87	0.99	-	-0.62**	0.43	-0.08	-0.98*	-0.007
	<i>Pleurosigma</i>	-2.42	0.98	-	-1.01**	-0.86	0.51	0.78	0.057
	<i>Grammatophora</i>	-2.73	0.98	-	0.09	0.76	0.48	-0.46	1.12***
	<i>Coscinodiscus</i>	-1.98	0.99	-	0.09	0.76	0.48	-0.46	1.12***
	<i>Cocconeis</i>	8.06	0.99	-	-0.92***	-0.58	-0.19	-0.1	-0.59**
	<i>Streptothea</i>	8.56	0.92	-	0.63*	2.29	-1.07	-3.14*	-0.29
II	<i>Biddulphia</i>	-3.69	0.95	-	0.86*	-0.69	0.32	-0.17	0.29*
	<i>Bellerochea</i>	-3.69	0.95	-	0.86*	-0.69	0.32	-0.17	0.29*
	<i>Raphoneis</i>	-8.43	0.98	-	0.86*	-0.69*	0.38	-0.21	0.29
III	<i>Navicula</i>	-30.7	0.95	-	-0.21	0.56	1.27*	1.73*	2.19*
	<i>Cyclotella</i>	-3.68	0.95	-	0.86*	-0.69	0.32	-0.17	0.29
	<i>Thalassiothrix</i>	21.49	0.98	-	0.95**	-0.93**	-0.54	-1.39*	-0.65
	<i>Bellerochea</i>	-3.69	0.95	-	0.86*	-0.69	0.32	-0.17	0.29
	<i>Raphoneis</i>	-8.43	0.98	-	0.86*	-0.69*	0.39	-0.21*	0.29
	<i>Pleurosigma</i>	-3.69	0.95	-	0.86*	-0.69	0.32	-0.17	0.29

Table 2A.13b. Partial regression coefficients (Beta values) for the dependent variable (viable diatoms cm⁻³ wet sediment) and independent variable (grain size) (I = 0-5 cm; II = 5-10 cm; III = 10-15 cm) at high tide zone (* p < 0.05; **p < 0.005; ***p < 0.01; ****p < 0.001)

Depth	Diatoms	Intercept	R	1,000	500	250	125	63	< 63
I	<i>Grammatophora</i>	-54.04	0.96		1.56*	-1.53*	11.95	9.48	2.95
III	<i>Achnanthes</i>	-0.97	0.99	-0.38**	0.26	0.72*	0.22	0.49	-0.35
	<i>Biddulphia</i>	-0.87	0.99	-0.38**	0.26	0.72*	0.22	0.48	-0.35
	<i>Melosira</i>	-1.08	0.99	-0.38**	0.26	0.72*	0.22	0.48	-0.35
	<i>Cyclotella</i>	-3.63	0.99	-1.02****	0.12*	-0.04***	2.16**	3.28	0.25
	<i>Pinnularia</i>	17	0.99	0.011	-1.01	1.01	-5.44*	0.54*	0.044
	<i>Coscinodiscus</i>	-2.74	0.99	-1.02****	0.12*	-0.044	2.16***	3.28****	0.25
	<i>Streptothea</i>	-0.55	0.99	-0.38**	0.26	0.72*	0.22	0.49	-0.35

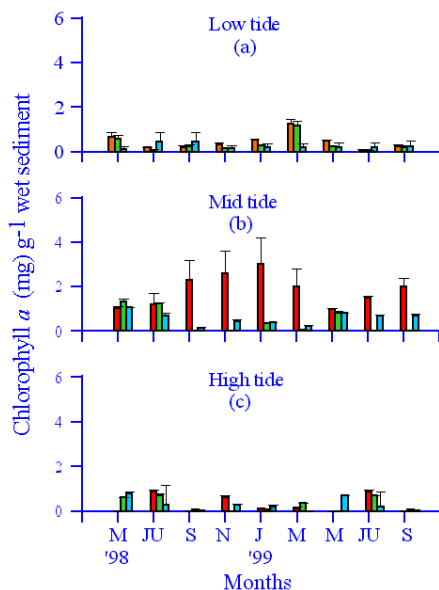
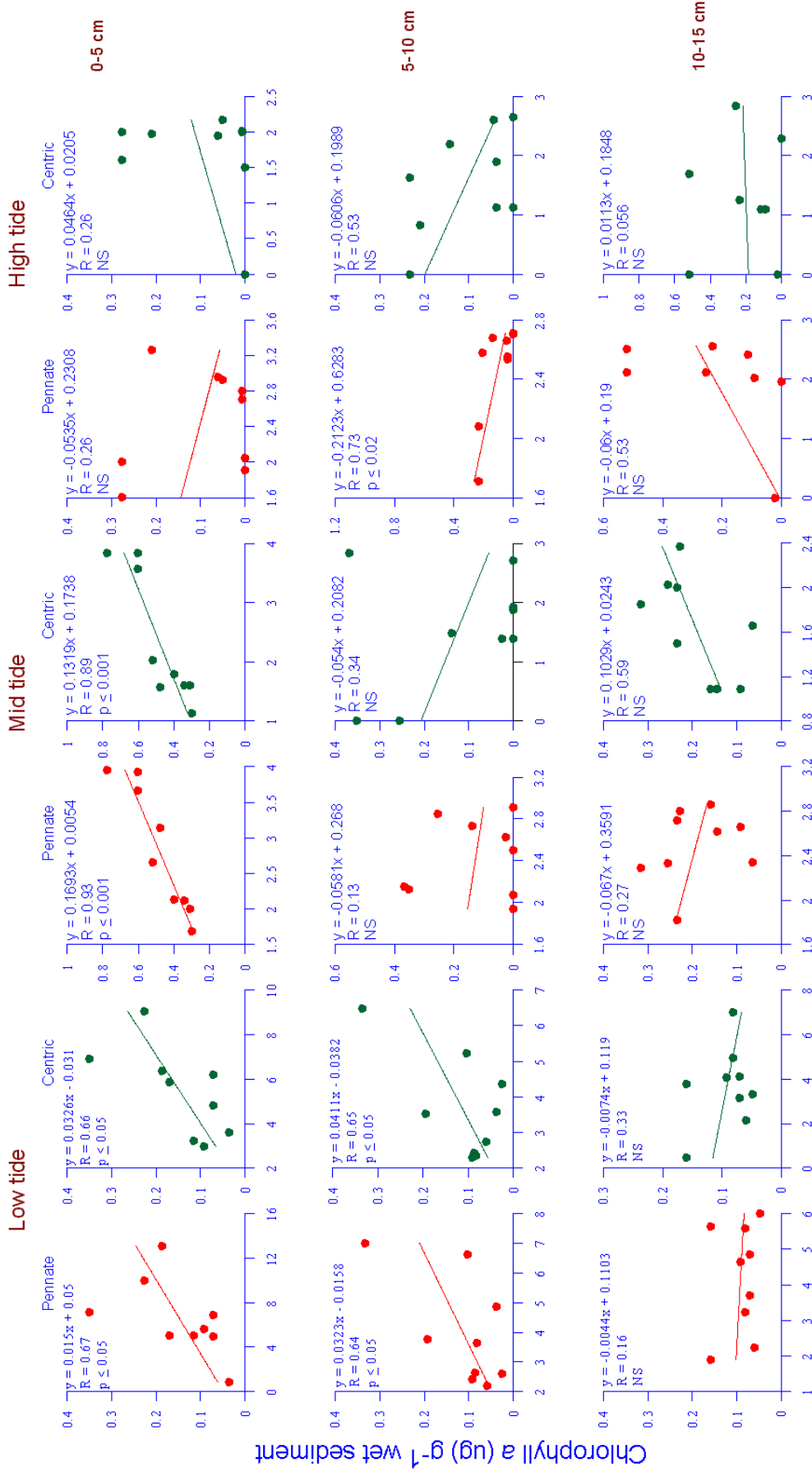


Fig. 2A.18 Concentrations of chlorophyll *a* across the three vertical sections of the sediment core at the low, mid and high tide zones

2A.4 Discussion

The sediment-dwelling diatoms form a major component of the microphytobenthic community in the intertidal region (Meadows and Anderson 1968; Round 1979a). This diatom community is generally classified as the epipsammon (particle attached diatoms) and the epipelon (free living diatoms). However, a third group of diatoms encountered in surf water, i.e. the phytoplankton, may also get incorporated into this diatom community (MacIntyre et al. 1996), some of whose members form a permanent component of this community throughout the year. So far, investigations on diatoms of the microphytobenthic community have been focused on epipsammic and epipellic pennate diatoms (Fenchel and Staarup 1971; Admiraal et al. 1982; de Jonge 1985). Steele and Baird (1968) have reported that the distribution of these epipsammic and epipellic autotrophic flora may extend to a depth of 20 cm. Observation under microscope usually reveals either quite clean-looking grains or grains with a film of mucilage in which bacteria and organic particles are prolific (Round 1981). Due to the difficulties of microscopic observation, the labor involved



Diatom cell abundance g^{-1} wet sediment

Fig. 2A.19 Linear regression analysis between chlorophyll *a* and pennate and centric diatom abundance across the three vertical sections of the core at the low (a), mid (b) and high tide (c) zones

and the time required, findings have been based on either chlorophyll estimation (Steele and Baird 1968) or epifluorescence microscopic techniques (Grontved 1960; Admiraal et al. 1982; Baillie 1986; Delgado 1989; Cheng et al. 1993). Another widely used method of harvesting diatom cells by using the lens tissue is not purposeful here, since it harvests only the motile pennate diatoms, without providing any information on the sub-surface diatom population (Round and Palmer 1966). The Ludox-TM density separation method described by de Jonge (1979) gives quantitative results but is laborious. The MPN method employed in this investigation includes incubation of the sample, which allows the diatoms to grow and multiply and thus enables easy observation (Imai et al. 1984, 1990; Yamochi 1989; Ishikawa and Tamaguchi 1994; Itakura et al. 1997). Although this method does not give the exact number of diatom cells, since it is based on the presence or absence of the diatoms (both vegetative and resting), it qualitates the relative abundance of different taxa. In the present study, f/2 medium was employed for the growth of diatoms, as it has been extensively used in the culture of phytoplankton (French and Hargraves 1980; Eilertsen et al. 1995). However, differences in experimental conditions and the composition of the medium could result in variations that need to be evaluated in detail.

2A.4.1 Intertidal horizontal distribution

The presence of diatoms from the low to the high tide zone reveals their constant redistribution over intertidal areas by tidal action. The diatom population was abundant in the mid tide zone followed by the low and the high tide zone. This pattern of intertidal distribution is the result of one or a combination of several environmental factors. The high tide zone is the most terrestrial of the beach environments. Sand in the low tide zone dries out only at the surface during exposure, whilst in the high tide

zone, sand dries for longer periods and to a greater depth. This will also be enhanced due to the comparatively coarser grain size at this zone. Since, the high tide zone is exposed for a longer period of time, the effects of desiccation in this zone may keep the population low (Bush 1966; McIntire and Wulff 1969). A decreasing oxygen concentration from the waterline landwards (Brafield 1964) can also explain a lower abundance in the high tide zone. Sediment temperatures are more variable with increasing elevation on the beach coincident with a longer period of exposure to incident radiation (Johnson 1965). This greatly fluctuating environment may be a hindrance to the growth of diatoms at the high tide zone. In the low and mid tide zone, the diatom population is exposed to nutrient-laden seawater for longer periods of time than those of the high tide zone. These factors may also enhance the growth of diatoms of the low and mid tide zones.

2A.4.2 Intertidal Vertical distribution

The appearance of diatoms down to a depth of 15 cm from the low to the high tide zones revealed that their viability was not affected by the conditions prevailing at this depth. It was observed that this depth harbored, not only the pennate diatoms, some of which are permanent residents of this area, but also centric forms, which are usually found in the plankton. The occurrence of group I diatoms in the sediment throughout the year indicated that the two pennate forms were natives of this area. The centric diatom *Thalassiosira*, which was present in surf water but whose vegetative cells were not observed in the sediment, appeared after incubation; this must have occurred through the germination of resting stages, which are regularly brought in with coastal sediment and redeposited in the intertidal sediment. Most of the pennate diatoms such as *Navicula* and *Amphora*, which are primarily encountered on bottom sediments, are also often found in the water column (de Jonge and Van Beusekom 1992; Tomas

1997) through resuspension, and it seems reasonable to assume that their primary production in the water column is as effective as it is on the tidal flats (de Jonge and Van Beusekom 1995). Their presence in the water column is also reflected in the microfouling population of different types of substrata immersed in the sub-surface estuarine waters of the present study area (Smita et al. 1997).

A number of physical, biological and chemical factors may be responsible for the temporal and spatial variation in the diatom abundance, diversity, richness and evenness.

2A.4.3 Physical factors

Wind speed and suspended load: Gabrielson and Lukatelich (1985) and de Jonge (1995) showed that chlorophyll *a* increase in the water column was related to resuspension. Resuspension is dependent on the sediment composition of the tidal flats and wind speed (de Jonge 1995). In a study on the resuspension of microphytobenthos and mud, a statistically significant linear relationship was found between wind speed and the fraction of suspended matter < 55 μm in the water above tidal flats in EMS estuary (de Jonge and Van Beusekom 1992). The wind speed ranged between 2 to 14 m s^{-1} in the EMS estuary (de Jonge 1995). This is much higher than the speeds encountered in the present study area, which range from 2.0 to 4.7 m s^{-1} . However, even at such a comparatively low wind speed, a significant relationship was observed between wind speed and suspended load at the low tide zone, suggesting that the wind speed contributes to resuspension of the sediment only at this zone. Also, the relation of both wind speed and suspended load with chlorophyll *a* and relative abundance of pennate diatoms at the low tide zone, in the 0-5 cm stratum was observed to be negative (Fig. 2A.10). This shows that wind

stimulates resuspension of the sediment along with pennate diatoms and, thus, chlorophyll *a* in the 0-5 cm stratum, only at the low tide zone.

Grain size: The dynamic habitat of the sandy shore encompasses grains of different size groups, and their proportion was found to alter with month and depth, only depth and only months for the low, mid and high tide zone respectively (Table 2A.2 and Table 2A.8a-c; Fig. 2A.5 and Fig. 2A.13a-c). It was observed that although a particular grain size at one depth served as a predictor of some diatoms, a similar observation was not evident at other depths and tidal zones (Table 2A.7 and Table 2A.13). This indicates that factors other than grain size play a role in the temporal and vertical distribution of diatoms.

Suspension and deposition: Diatoms such as *Amphora*, *Navicula* and *Thalassiosira*, which were dominant in the surface sediment layers, were also the dominant diatoms throughout the 15 cm core of the low, mid and high tide zones; this included both pennate and centric diatoms. In sandy sediments, light irradiance can decrease to 1% at a depth of 2-3mm (Rasmussen et al. 1983). Therefore, the dominance of the above-mentioned species beyond the photic zone cannot be attributed to cell division. Moreover, since centric diatoms do not possess the ability to move on their own, a migratory mode of transport can also be ruled out for these forms. Hydrodynamic processes such as the waves are responsible for deposition and resuspension of the sediment (Baillie and Welsh 1980, Delgado et al. 1991; de Jonge and Van Beusekom 1992, 1995; de Jonge and Van den Bergs 1987, de Jonge 1992). The churning of water caused by these processes leads to the suspension of both the diatom-laden sediment particles and the free-living diatom cells in the surface waters. After redistribution, these suspended particles are again deposited on or within the

sediment. During this process, an exchange of diatom cells can take place between the surface and deeper layers of the sediment.

Advection: Advective transport of the diatom cells can occur through the sediment column via percolation of surface water, when the sand is loosened during flood tide. The degree of permeability of the sediment depends on the grain size, being higher for sand than for silty sand (Rusch and Huettel 2000). This vector will be more effective in case of planktonic diatoms, present in the overlying waters or left behind on the sediment surface by the receding tides (Huettel and Rusch 2000). In the case of pennate diatoms, only those detached will be affected. Although, all the forms dominant in surface layers were found throughout the 15 cm core in this investigation, the centric diatom *Thalassiosira* dominated the entire community up to 15 cm (Fig. 2A.9a-b and Fig. 2A.15a). It can be assumed that in sandy sediments, oscillatory wave motions (Reid and Kajiura 1957) will cause water exchange to a sediment depth of at least 20 cm (Steele and Munro 1970). Thus, this vector may act as a selective carrier mostly for the loosely attached or the free-living diatom cells, especially the centric forms.

2A.4.4 Biological factors

Grazing: Since diatoms are the major autotrophic organisms of this region, they also form the major food for consumers, e.g. for meio- and macrobenthos. An earlier study revealed that macrobenthic organisms are higher in the food chain than meiobenthic and are represented by more detritus feeders than suspension feeders [amphipoda (40.73 %) and polychaetes (32.52 %)] (Rodrigues 1984). It was also observed that the energy increase caused by the wave action and currents at Dona Paula beach is initiated during the pre-monsoon and peaks during the monsoon period, resulting in erosion of the beach (Rodrigues 1984). This results in an increase

in the population of these grazers during the post-monsoon and a decrease during the monsoon and pre-monsoon seasons, which may be one of the factors responsible for the higher abundance of group I and group II diatoms during the pre-monsoon period.

Vertical migration: In a dynamic environment, such as the intertidal region, permanent residents will be adapted to the changing environmental conditions and possess mechanisms to overcome difficult situations. The free-living epipellic pennate diatoms are known to exhibit migratory behavior, whereby they move in the sediment, away from or towards light, the former when the tide rises and the latter when the tide recedes. Migration is said to occur in order: (1) to escape the bright light conditions and reach a place where the conditions are feasible for primary production, (2) to reduce predation by surface feeders and (3) to avoid transport due to resuspension by the advancing tide (Faure-Fremiet 1951; Ganapati et al. 1959; Palmer and Round 1965; Round 1979a; Heckman 1985). In this investigation, two major pennate diatoms (*Navicula* and *Amphora*) were observed up to 15 cm sediment depth (Fig. 6a). Vertical migratory behavior in different sediment types has been related to physical factors such as tidal rhythms and light intensity (Hopkins 1966; Round and Palmer 1966). But many of these field studies were based on a millimeter scale (Hopkins 1963) except one, which reported four diatom species migrating down to 1 cm (Joint et al. 1982). However, work conducted in shallow, outdoor tidal tanks equipped with wave generators showed that vertical migration of a diatom, *Hantzschia virgata* extends beyond 8 cm during high tide (Kingston 1999). Hence, such migratory behavior of these diatoms can result in the transportation of the surface diatoms to deeper layers, much beyond the photic zone. However, this self-initiated mode of transportation is only possible for epipellic pennate diatoms, and hence this cannot be the only factor responsible for diatom transport.

Bioturbation: During the processes of feeding, defecation and locomotion, deposit feeders such as polychaetes, bivalves, amphipods, gastropods and crustaceans continuously rework the sediment. Burrowing forms such as polychaetes (e.g. *Scoloplus kerguelensis*) and amphipods (e.g. *Talorchestia*) dominate the macrofauna of this area (Rodrigues 1984). Many field and laboratory studies have demonstrated that continuous sediment reworking by burrowing animals generally stimulates sediment transportation, carrying along with it the attached and adhered microflora and thus acting as another biological factor in the regulation of species diversity and benthic community structure (Pamatmat 1968; Lukatelich and McComb 1986; de Jonge and Van den Bergs 1987). The fact that diatoms were observed up to 15 cm depth indicates that burrowing organisms play an active role even at this depth. However, in the present investigation, even though the diatom numbers did not reveal any particular trend in depth-wise distribution and were encountered up to 15 cm depth in appreciable numbers, the correlation between chlorophyll *a* and diatom numbers was found to be highest in the surface (0-5 cm) stratum, followed by the 5-10 cm core section and negligible at 10-15 cm depth at the low tide zone. This positive correlation reduced to a depth of 5 cm at the mid tide zone, to 0 cm at the high tide zone. Hence, this indicates that even if bioturbation activity reached beyond the 10 cm zone, its effects must have been overridden by dormancy or desiccation in case of the high tide zone.

2A.4.5 Chemical factors

Nutrients can play an important role in the distribution of some of the diatoms, especially during the late monsoon period, which is characterized by the renewal of nutrients to the surface waters by upwelling and land run off. *Fragilariopsis* and *Skeletonema* (group III) appeared in the intertidal sediment at the low tide zone

during August-September (monsoon). The centric diatom *Skeletonema* blooms during August-September in these waters (Smita et al. 1997) and is carried to the intertidal sediment by advective transport. *Fragilariopsis*, a pennate diatom occurring in chains, was found in high numbers during the same period, both in the water column and in the intertidal sediment. Since the subsiding bloom of *Skeletonema* undergoes resting stage formation and *Fragilariopsis* is a resident of the coastal sediment, carried by water masses to the intertidal sediment or the water column (Fig. 2A.4, 2A.7a-b and 2A.15c). The abundance of centric diatoms at the low tide zone was influenced by nitrate and phosphate, whereas in the case of pennates, which mainly reside in the sediments, nutrients did not play an important role throughout the intertidal zone (Table 2A.6).

2A.4.6 Survival strategies

Once the diatoms have reached a depth of 15 cm, either voluntarily or forcefully, they have to adapt to some life saving strategies, at least till they are brought back to the surface by some mechanism. One of the possibilities for survival is that they adapt to spending long periods in the dark, at low metabolic rates with negligible degradation of pigments (Steele and Baird 1968; French and Hargraves 1980) and uptake of dissolved organic compounds (Admiraal and Peletier 1979).

Laboratory observations have shown that cells of *Hantzschia virgata* exhibit migration up to 8 cm during high tide (Kingston 1999). Taking this fact into consideration, it is obvious that in an area at the lowest low tide zone, the cells cannot migrate up and down during the short period of exposure at every tidal interval. If these vegetative cells are subjected to high sedimentation rates, their presence at such depths without photosynthesizing, for a period beyond their tolerance limit will expose them to unfavorable conditions, such as darkness, under which photosynthesis

will be hindered. Thus, the cell will either undergo transformation from the vegetative to a resting stage, triggered by the surrounding conditions, in order to survive the unfavorable environmental condition(s), or face death and degradation. Experimental work has shown that permanent darkness and low nutrients may result in resting stage formation (Anderson 1975; Hargraves and French 1983).

Although chlorophyll *a* in itself is insufficient to describe fluctuations in benthic diatom standing stock (de Jonge 1980), this pigment provides a useful index for the photosynthesis potential of a population (McIntyre et al. 1996). A significant relation between diatom cell numbers and chlorophyll *a* to a depth of 10 cm and 5 cm at the low tide and mid tide zones respectively revealed that both pennate and centric diatoms contributed to the total pool of chlorophyll *a* down to this depth (Fig. 2A.12 and Fig. 2A.19). In the 0-5 cm stratum, the ability of pennate diatoms to remain attached to the substrate may play an important role in this contribution, whereas centric diatoms may be brought to the sediments by incoming tides. In the 5-10 cm stratum, the positive correlation with chlorophyll *a* revealed that resuspension was not effective down to this depth and that the stock was securely placed. Although viable cells did occur at 15 cm, the reason for the lack of relationship between chlorophyll *a* and diatom cell number at 10-15 cm depth at low tide zone and 5-15 cm at the mid tide zone could be either that there was no sufficient renewal of cells even when advection occurred or that the cells were in dormant stage. The negative correlation at the high tide zone throughout the 15 cm depth may be due to the effects of desiccation when exposed for longer durations. However, they would remain viable as primary producers throughout the intertidal zone down to 15 cm depth, and when they resurfaced could play an important role in the benthic community.

2B.1 Introduction

The intertidal areas are dynamic habitats, subjected to harsh hydrodynamic processes such as waves, currents and tidal exposure. These areas are inhabited by benthic diatoms, which have been divided into two categories. Some of the diatoms move actively in the surface layers and form a component of the epipelon, whilst others are relatively, although not completely immobile, being attached to sand grains contributing to the epipsammon component of the diatom community (Round 1965; 1979b). While both the assemblages are capable of movement, a necessity in such an unstable habitat, only the epipellic species move sufficiently fast (1 to $25 \mu\text{m s}^{-1}$) to undergo diurnal rhythmic migrations (Round 1971). The epipsammic species move only very slowly from one sand grain to the other, although even here low-level diurnal rhythmic activity has been reported (Harper and Harper 1967; Harper 1969).

Depending on the grain size and organic content, irradiance in sandy sediment decreases to 1% at a depth of 2 to 3 mm (Rasmussen et al. 1983) during low tide. So, only the diatoms in that sediment layer play a role in photosynthesis. But, during high tide turbulence and shear stresses generated by tidal currents or wind waves results in the resuspension of the sediment surface along with the associated diatoms, thus disturbing the microphytobenthic community at the sediment surface. However, as a result of this a substantial part of the biomass of these algae may be found in deeper layers of the sediment. In such conditions, the ecology of the residing microphytobenthos gets highly complex. In order to overcome these difficulties and survive in such an environment, certain mechanisms have been adopted. The survival strategies include adaptation to spending long periods in the dark at low metabolic rates and with negligible degradation of pigments (Steele and Baird 1968; French and Hargraves 1980), uptake of dissolved organic compounds (Admiraal and Peletier

1979) or to undergo transformation from vegetative to a resting stage (Anderson 1975; Hargraves and French 1983). On the other hand, the self-initiated vertical migration helps diatoms to move to the surface during low tides for photosynthesis and move down during high tides (Round and Palmer 1966; Round 1979b; Joint et al. 1982; Paterson 1989). There exists a controversy regarding the reason for the vertical migration and the factors affecting it. Light and tidal rhythm are the two variables, which seem to induce this migratory behavior (Round and Palmer 1966).

Much of the literature concerning vertical migration of benthic microalgae has centered on species inhabiting mud flats, where the amplitude of migration is generally 3mm for diatoms (Aleem 1950; Hopkins 1963; Round and Haphey 1965; Round and Eaton 1966; Round and Palmer 1966; Palmer and Round 1967; Round 1978; Paterson 1986; Pinckney et al. 1994). In case of the sand flat community, Round (1979a) observed *Hantzschia virgata* migrate up to a depth of 3 mm whereas Joint et al. (1982) found three diatom genera (*Navicula*, *Diploneis* and *Nitzschia*) migrate down to 1cm depth. Several other studies confirmed that most of the living diatoms on sand flats occur in the uppermost centimeter of sand (Meadows and Anderson 1968; Harper 1969; Riznyk and Phinney 1972; Round 1979a, b). However, work conducted in shallow outdoor tidal tanks equipped with wave generators showed that vertical migration of *Hantzschia virgata* could extend beyond 8 cm during high tide (Kingston 1999).

Vertical migration can have important consequences for measurements of both diatom abundance and photosynthesis. Moreover, the effort and expense of counting diatoms and the difficulty of calculating biomass from cell abundance preclude the use of cell counts in most investigations. Instead, the photosynthetic pigment, chlorophyll *a* is used as an index of microphytobenthic biomass.

This investigation explored the behavior of diatoms with reference to tidal cycles in the field, as well as when removed from the effects of tides in the laboratory and incubated under 12 h light: 12 h dark, continuous dark, or continuous light.

2B.2 Materials and methods

2B.2.1 Sampling site

Sediment sampling was carried out on a sand flat at Dias Beach (15° 27' N; 73° 48' E), located near Dona Paula Bay and surrounded by the Mandovi and Zuari estuaries. This beach is 200 m long. The tides are semi-diurnal, with an average spring tide range of ~ 2 m and neap tide range of ~ 0.7 m.

2B.2.2 Sampling period

The first sampling was carried out on 30 - 31 March 2001 (Summer season; case I) and the second sampling on 14 - 15 January 2002 (Winter season; case II). The sampling program is given in Table 2B.1.

Table 2B.1. Details of the tidal time, tidal amplitude, temperature, day length, total sunshine and solar radiation of the sampling days

Sampling date	Sampling period	Tidal Time (H)	Tidal amplitude (m)	Temperature (°C)	Day length (H)	Total Sunshine (H)	Solar radiation (mw cm ⁻²)
30 th March 2001	Morning Low Tide	08:02	0.33	27	12.17	8.2	10.4 - 27.4
	Noon High Tide	14:50	1.94				54.1 - 73.5
	Evening Low Tide	20:25	1.11				1.1 - 1.2
31 st March 2001	Night High Tide	01:51	1.9	12.2	9.5	1 - 1.1	
	Morning Low Tide	08:49	0.38			27.2 - 47.1	
14 th January 2002	Mid-Morning High Tide	10:48	1.83	21	11.04	9.9	33.4 - 61.4
	Evening Low Tide	17:25	0.27				4.5 - 20.7
	Night High Tide	00:32	2.27				1.0 - 1.1
15 th January 2002	Early Morning Low Tide	06:27	1.09	11.05	9.5	1.1 - 1.4	
	Mid Morning High Tide	11:25	1.82			60.7 - 70.2	

2B.2.3 Sediment characteristics

Very fine sand (63 to 124 μm) is the most abundant grain size fraction of the sediment. The sediment was moderate to well sorted, with a sorting coefficient ranging from 0.32 to 0.58 (Mitbavkar and Anil 2002).

2B.2.4 Sediment core collection

Sediment cores (1 cm length) were collected in five replicates using small poly-vinyl chloride (PVC) corers with an inner diameter of 3 cm.

2B.2.5 Sampling program

During each of the morning and noon tides of case I and mid-morning and evening tides of case II (using a particular reference point), 10 cores were collected for immediate observation whereas for laboratory incubation 80 cores were collected in summer and 120 cores in winter (Fig. 2B.1). The cores were sealed at the lower ends with PVC caps and transferred to the laboratory. Of the total cores collected for laboratory incubation, 40 cores were incubated in 12 h light: 12 h dark, 40 cores in continuous dark, and the additional 40 cores collected during winter were incubated in continuous light. Incubation was carried out at 27 ± 1 $^{\circ}\text{C}$ under a light intensity of 32.3 mw cm^{-2} . Simultaneously, the set of cores collected for immediate observation were sectioned immediately and out of the 10, 5 were used for diatom enumeration and 5 for chlorophyll *a* analyses.

Out of the 40 cores from each of the incubated conditions, 10 were removed after every 6-hour. Five cores were used for chlorophyll *a* analyses and 5 were used for diatom enumeration. Simultaneously during the same period, cores were collected from field for immediate observation.

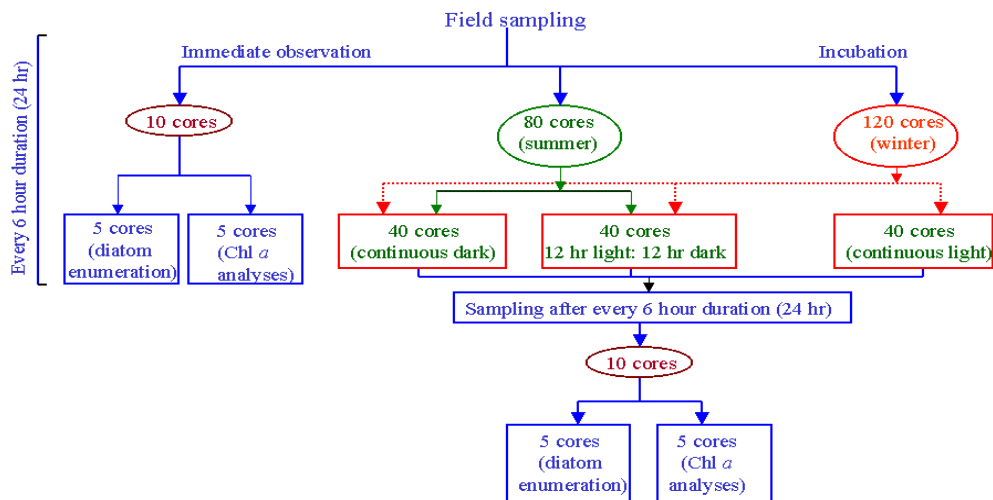


Fig. 2B.1 Schematic representation of the experimental protocol

2B.2.6 Core sectioning

All sediment cores were sectioned at 1mm intervals. Out of these, the 1st, 3rd, 5th and 7th mm sections were used for diatom enumeration and chlorophyll *a* analyses.

2B.2.7 Diatom enumeration

Each of the sediment core sections was suspended in autoclaved seawater. As epipellic diatoms dominated the summer samples, the sediment was washed thoroughly five times to suspend the free and loosely adhered epipellic diatom cells from sand grains. This supernatant was separated and used for epipellic diatom quantification and identification. In the winter samples, epipellic as well as epipsammic diatoms dominated the population, so the washed sediment, after the separation of epipellic diatoms was further subjected to ultrasonication to separate the attached epipsammic diatoms. The diatoms were identified based on the keys provided by Heurck (1896), Subrahmanyam (1946), Desikachary (1987) and Tomas (1997).

2B.2.8 Chlorophyll *a* analyses

A known amount of the sediment sample from the respective depth interval was extracted in 90% acetone for 18 h (corrected for phaeopigments) (Parsons et al. 1984).

2B.2.9 Data analysis

The log (X+1) transformed data on diatom abundance of the low tide and high tide incubated cores under different conditions were subjected to multiple analyses of variance (MANOVA) (Sokal and Rohlf 1981).

2B.3 Results

2B.3.1 Case I

On this occasion, low tide occurred in the morning after sunrise. The values of solar radiation and tidal amplitude are given in Table 2B.1. During the morning exposure period, randomly distributed brownish patches were observed. Immediate microscopic observation of this sediment revealed dominance of two epipellic diatom species, *Navicula transitans* var. *derasa* f. *delicatula* Heimdal and *Amphora coffeaeformis* (Agardh) Kutzing. Hence, case I study was restricted only to these two dominant forms.

Temporal variation in the field

Cell abundance of epipellic diatoms and chlorophyll *a* concentrations were high in the surface 1mm only during the morning low tide (Fig. 2B.2a and 2B.3a). Of the two dominant diatoms, *A. coffeaeformis* was abundant (Fig. 2B.4a and 2B.5a). Cell density at the 3, 5 and 7mm sediment core depths was negligible as compared to that of the surface. The fact that the period of decline in surface population was not reflected as an increase at the lower depths sampled, suggests that either the cells have been suspended into the surface waters or migrated beyond the 7mm depth.

Observation of the laboratory incubated cores sampled during (a) Morning low tide

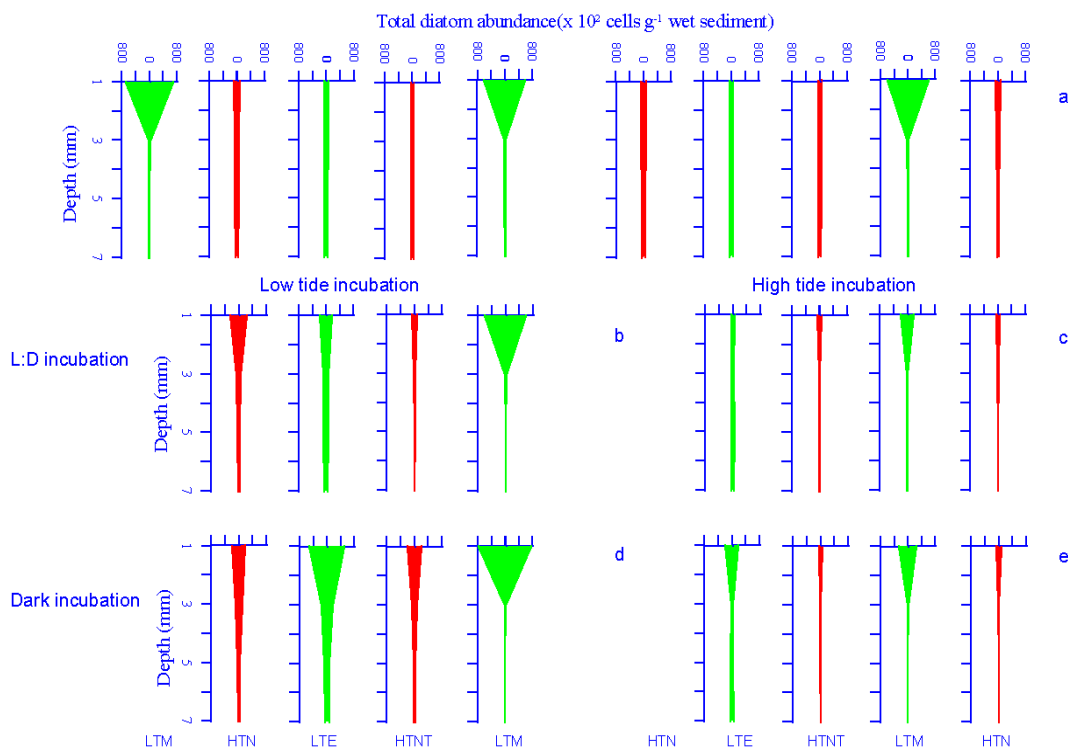


Fig. 2B.2 Depthwise distribution of mean total diatom (10^2 cells g^{-1} wet sediment) of case I sediment cores for the different tidal periods in the field (a), under controlled conditions for the “low tide” [12 h light: 12 h dark (b), continuous dark (d)] and high tide incubated cores [12 h light: 12 h dark (c), continuous dark (e)]

LTM=Low tide morning; HTN=High tide noon; LTE=Low tide evening; HTNT=High tide night

12 h light: 12 h dark incubation: When incubated under this condition, the diatom population and chlorophyll *a* concentrations were found to follow a trend similar to that observed in the field (Fig. 2B.2b and 2B.3b).

Continuous dark incubation: When incubated in continuous darkness, the surface cell abundance and chlorophyll *a* concentrations revealed a tidal rhythm, wherein the period of field emersion and high tide cover coincided with surface maxima and subsequent fall respectively (Fig. 2B.2d and 2B.3d).

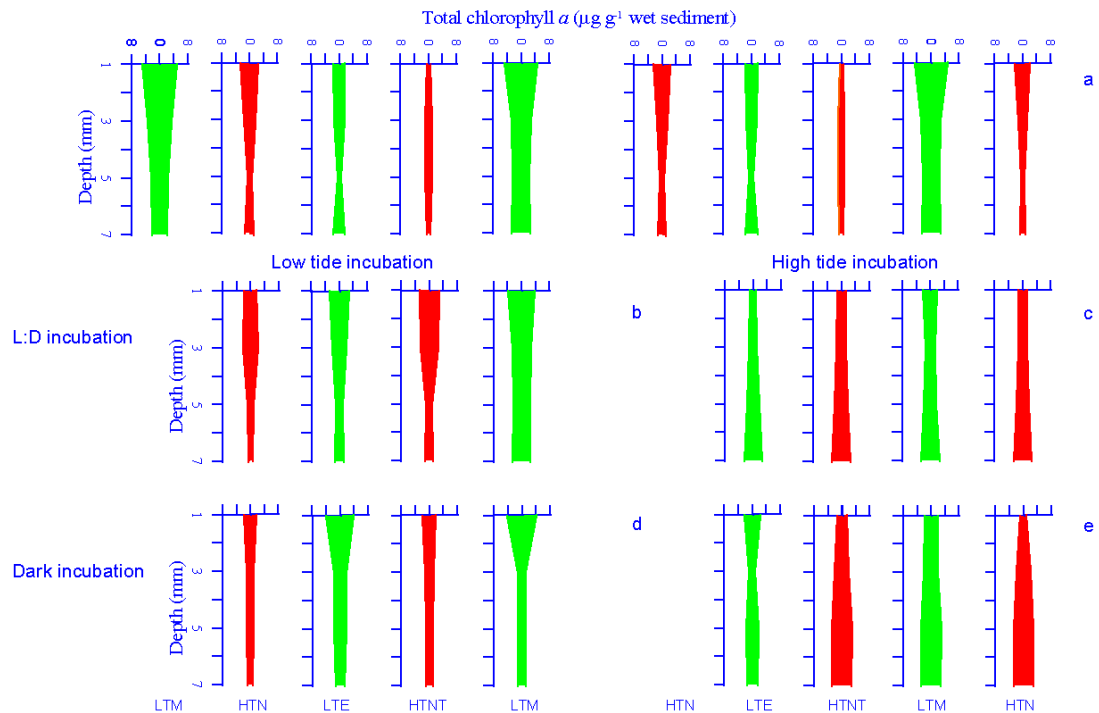


Fig. 2B.3 Depthwise distribution of chlorophyll *a* ($\mu\text{g g}^{-1}$ wet sediment) of case I sediment cores for the different tidal periods in the field (a), under controlled conditions for the “low tide” [12 h light: 12 h dark (b), continuous dark (d)] and high tide incubated cores [12 h light: 12 h dark (c), continuous dark (e)]
 LTM=Low tide morning; HTN=High tide noon; LTE=Low tide evening; HTNT=High tide night

(b) Noon high tide

As the cores were collected during high tide, the surface diatom population was comparatively lower than that collected during low tide. Resuspension into surface waters or migration beyond the sampled depth are the probable factors responsible for the low cell abundance. On incubation in the 12 h light: 12 h dark and continuous dark condition, this population exhibited the same rhythm as the cores collected during the morning low tide (Fig. 2B.2c and 2B.2e).

Comparison between low tide and high tide incubated cores

MANOVA did not reveal any significant variation between the total diatom abundance of the low tide and high tide incubated cores with incubation time and depth when subjected to 12 h light: 12 h dark incubation (Fig. 2B.2b and 2B.2c). *A.*

coffeaformis and *N. delicatula* population behaved similarly (Fig. 2B.4b, 2B.4c, 2B.5b and 2B.5c). However, in the dark incubated cores, variation in diatom abundance between the tides with incubation time (Fig. 2B.2d and 2B.2e) is due to the

Table 2B.2 Significance values of the MANOVA evaluating the temporal variation in diatom abundance of case I along the vertical gradient (1, 3, 5 and 7mm sediment core sections) with respect to incubation conditions. (NS- Not-Significant) (a=Tides; b=Incubation time; c=Depth)

	12 hr light: 12 hr dark incubation			Continuous dark incubation		
	axb	axc	bxc	axb	axc	bxc
Total diatom abundance	NS	NS	NS	0.025	NS	NS
<i>Amphora coffeaformis</i> abundance	NS	NS	NS	NS	NS	NS
<i>Navicula delicatula</i> abundance	NS	NS	NS	0.025	NS	0.05
Chlorophyll <i>a</i>	0.005	0.001	NS	NS	0.025	NS

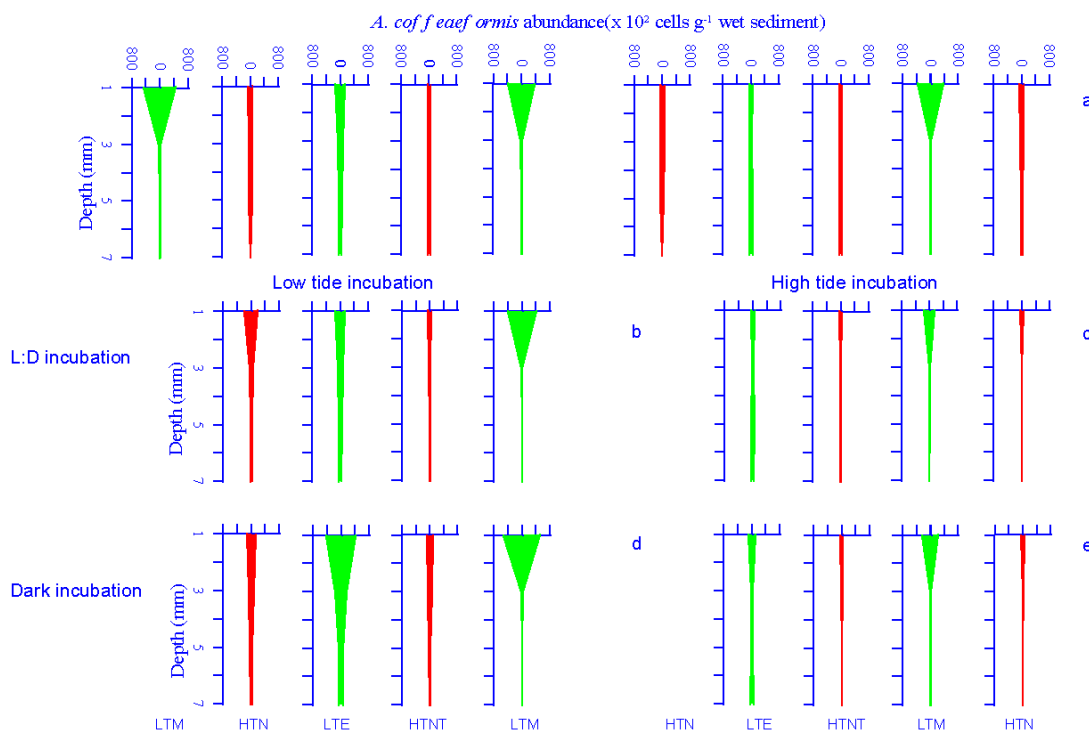


Fig. 2B.4 Depthwise distribution of *A. coffeaformis* (10^2 cells g^{-1} wet sediment) of case I sediment cores for the different tidal periods in the field (a), under controlled conditions for the “low tide” [12 h light: 12 h dark (b), continuous dark (d)] and high tide incubated cores [12 h light: 12 h dark (c), continuous dark (e)]

LTM=Low tide morning; HTN=High tide noon; LTE=Low tide evening; HTNT=High tide

comparatively lower initial number of cells in the high tide incubated cores which resulted in a lower abundance during the surface migration as compared to the low tide incubated core (Table 2B.2).

Comparison between 12 h light: 12 h dark and continuous dark incubations

Table 2B.3 Significance values of the MANOVA evaluating the temporal variation in diatom abundance of case I along the vertical gradient (1, 3, 5 and 7mm sediment core sections) with respect to low and high tide incubations. (NS- Not-Significant) (b=Incubation time; c=Depth; d=Conditions)

	Low tide incubation			High tide incubation		
	dxb	dxc	bx c	dxb	dx c	bx c
Total diatom abundance	0.005	NS	0.001	NS	NS	0.001
<i>Amphora coffeaeformis</i> abundance	0.05	0.025	0.05	NS	0.025	0.05
<i>Navicula delicatula</i> abundance	0.05	NS	0.001	0.001	NS	0.01
Chlorophyll <i>a</i>	NS	NS	NS	NS	NS	NS

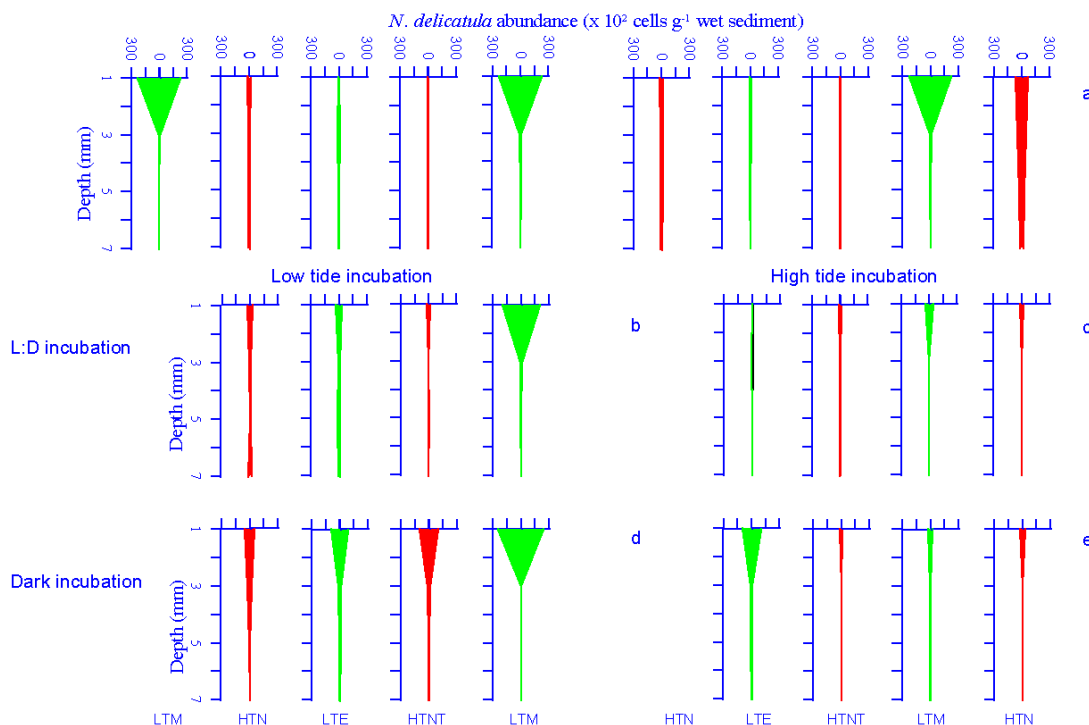


Fig. 2B.5 Depthwise distribution of *N. delicatula* (10^2 cells g^{-1} wet sediment) of case I sediment cores for the different tidal periods in the field (a), under controlled conditions for the “low tide” [12 h light: 12 h dark (b), continuous dark (d)] and high tide incubated cores [12 h light: 12 h dark (c), continuous dark (e)]

LTM=Low tide morning; HTN=High tide noon; LTE=Low tide evening; HTNT=High tide

When the diatom abundance of the low tide incubated cores were subjected to MANOVA, the total epipelagic diatom abundance revealed a significant variation between the conditions (12 h light: 12 h dark and continuous dark) with time (Fig. 2B.2b and 2B.2d). Both *A. coffeaeformis* and *N. delicatula* populations exhibited this variation (Fig. 2B.4b, 2B.4d, 2B.5b and 2B.5d). Each of these conditions also revealed depthwise variation in diatom abundance with incubation time. In the high

tide incubated cores, in each of the conditions the total diatom abundance revealed a significant depthwise variation with incubation time (Fig. 2B.2c, 2B.2e, 2B.4c, 2B.4e, 2B.5c and 2B.5e). Chlorophyll *a* concentrations did not show any significant variation between the two conditions with time as well as with depth (Table 2B.3).

2B.3.2 Case II

On this occasion, low tide occurred before sunrise. The values of solar radiation and tidal amplitude are given in Table 2B.1. First sampling was carried out during mid-

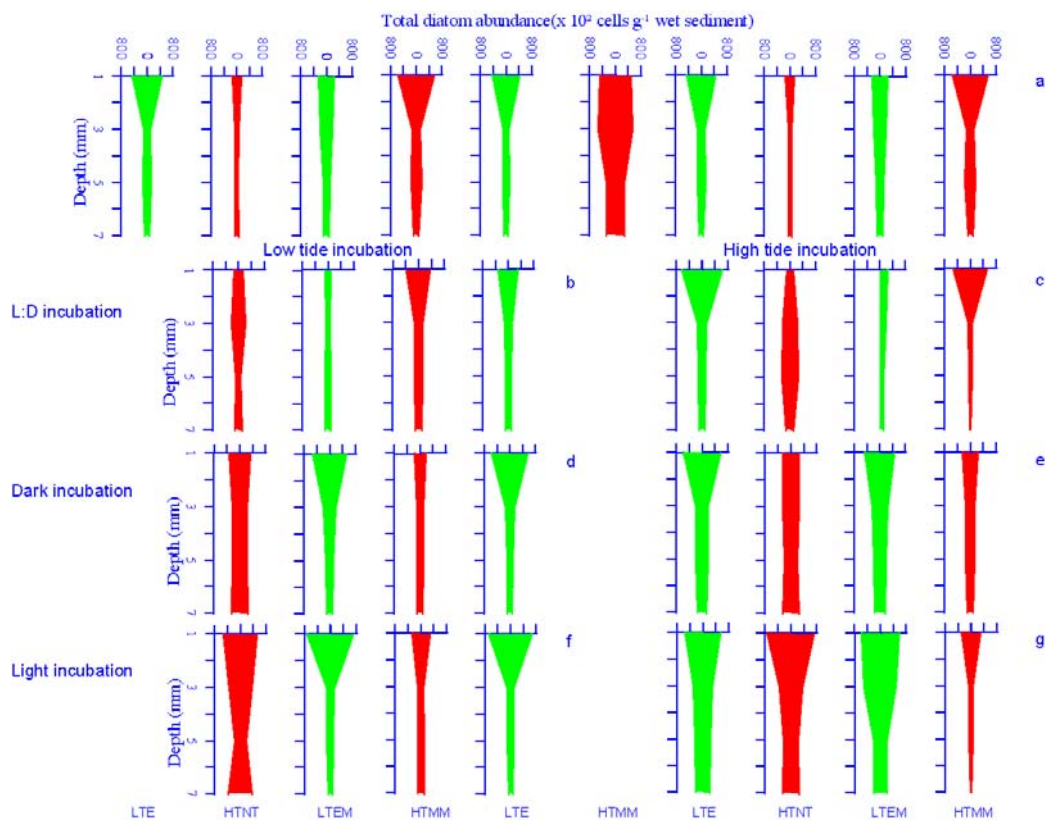


Fig. 2B.6 Depthwise distribution of mean total diatom (10^2 cells g^{-1} wet sediment) of case II sediment cores for the different tidal periods in the field (a), under controlled conditions for the “low tide” [12 h light: 12 h dark (b), continuous dark (d), continuous light (f)] and high tide incubated cores [12 h light: 12 h dark (c), continuous dark (e), continuous light (g)] LTE=Low tide evening; HTHT=High tide night; LTEM=Low tide early morning; HTMM=High tide mid-morning

morning high tide subsequent to the early morning low tide. Immediate observation of the field-collected cores revealed a mixture of epipellic (*N. delicatula* and *A. coffeaeformis*) and epipsammic diatoms (*Cocconeis scutellum* Ehrenberg, *Cymbella*

sp., *Amphora costata* Wm. Smith, *Rhaphoneis ampiceros* (Ehrenberg) Ehrenberg and *Grammatophora marina* (Lyngbye) Kutzing), which were found in almost equal abundance.

Temporal variation in the field

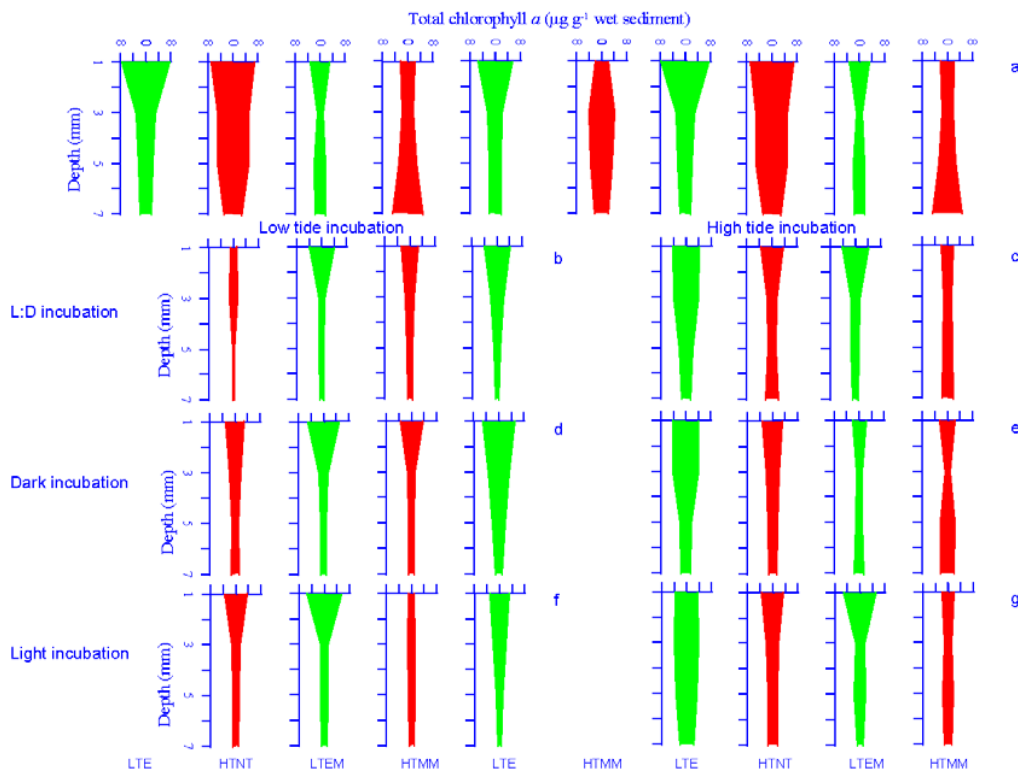


Fig. 2B.7 Depthwise distribution of chlorophyll *a* ($\mu\text{g g}^{-1}$ wet sediment) of case II sediment cores for the different tidal periods in the field (a), under controlled conditions for the “low tide” [12 h light: 12 h dark (b), continuous dark (d), continuous light (f)] and high tide incubated cores [12 h light: 12 h dark (c), continuous dark (e), continuous light (g)]
 LTE=Low tide evening; HTHT=High tide night; LTEM=Low tide early morning; HTMM=High tide mid-morning

Cell abundance of epipellic diatoms was high in the surface 1mm during the mid-morning high tide. This maximum continued even during the subsequent evening low tide. The following night high tide and early morning low tide revealed a decrease in the surface epipellic cell abundance (Fig. 2B.6a). Thus in this winter season, diatom surface migration was observed from mid-morning to evening on a diel basis. The surface cell abundance of epipellic and epipsammic diatom population was not reflected in the chlorophyll *a* concentrations during the mid-morning high tide (Fig.

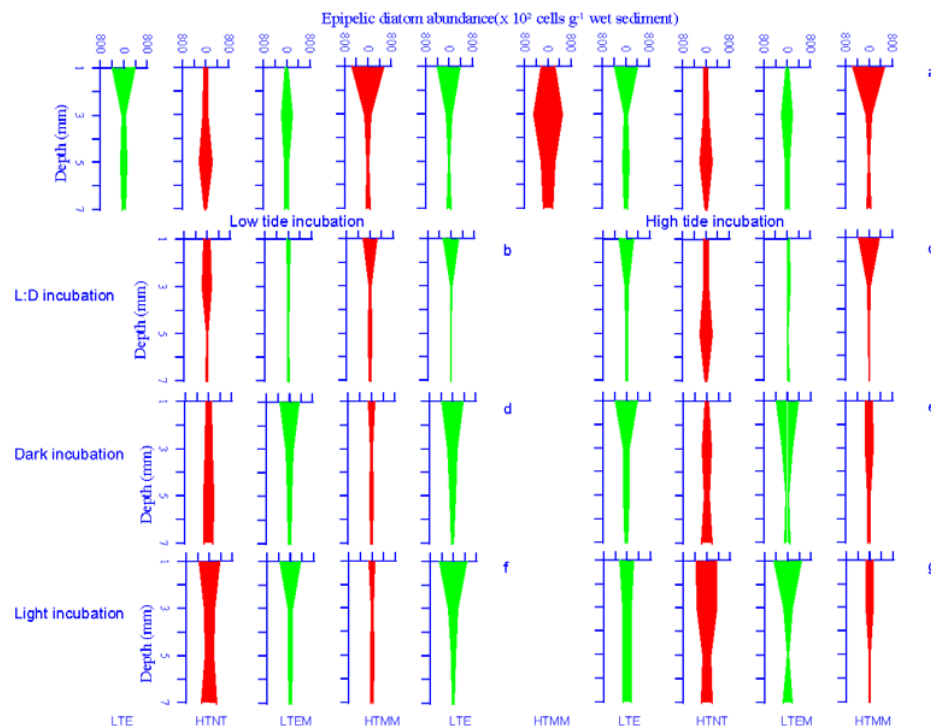


Fig. 2B.8 Depthwise distribution of mean epipelagic diatom (10^2 cells g^{-1} wet sediment) of case II sediment cores for the different tidal periods in the field (a), under controlled conditions for the “low tide” [12 h light: 12 h dark (b), continuous dark (d), continuous light (f)] and high tide incubated cores [12 h light: 12 h dark (c), continuous dark (e), continuous light (g)]

LTE=Low tide evening; HTHT=High tide night; LTEM=Low tide early morning; HTMM=High tide mid-morning

2B.7a, 2B.8a and 2B.9a). However, in the evening low tide exposure, the surface abundance of epipelagic diatoms was reflected in chlorophyll *a* concentrations (Fig. 2B.7a and 2B.8a).

Observation of the laboratory incubated cores sampled during (a) Mid-morning high tide

12 h light: 12 h dark incubation: When incubated under this condition, similar results were obtained as that during case I incubation wherein the surface epipelagic diatom abundance (*N. delicatula* and *A. coffeaeformis*) was found to follow a trend similar to that observed in the field (Fig. 2B.8c, 2B.10c and 2B.11c). Such a behavior was not observed in the epipsammic diatom population (Fig. 2B.9c).

Continuous dark incubation: When incubated in continuous darkness, epipelagic diatom population exhibited a tidal rhythm (Fig. 2B.8e, 2B.10e and 2B.11e).

Continuous light incubation: When incubated in continuous light, the epipelagic diatom population was seen to remain at the surface for a period of 18 h, i.e., from the time of collection, mid-morning high tide to the corresponding evening low tide to night high tide to early morning low tide of field (Fig. 2B.8g). Chlorophyll *a* concentrations also revealed similar results (Fig. 2B.7g).

(b) Evening low tide

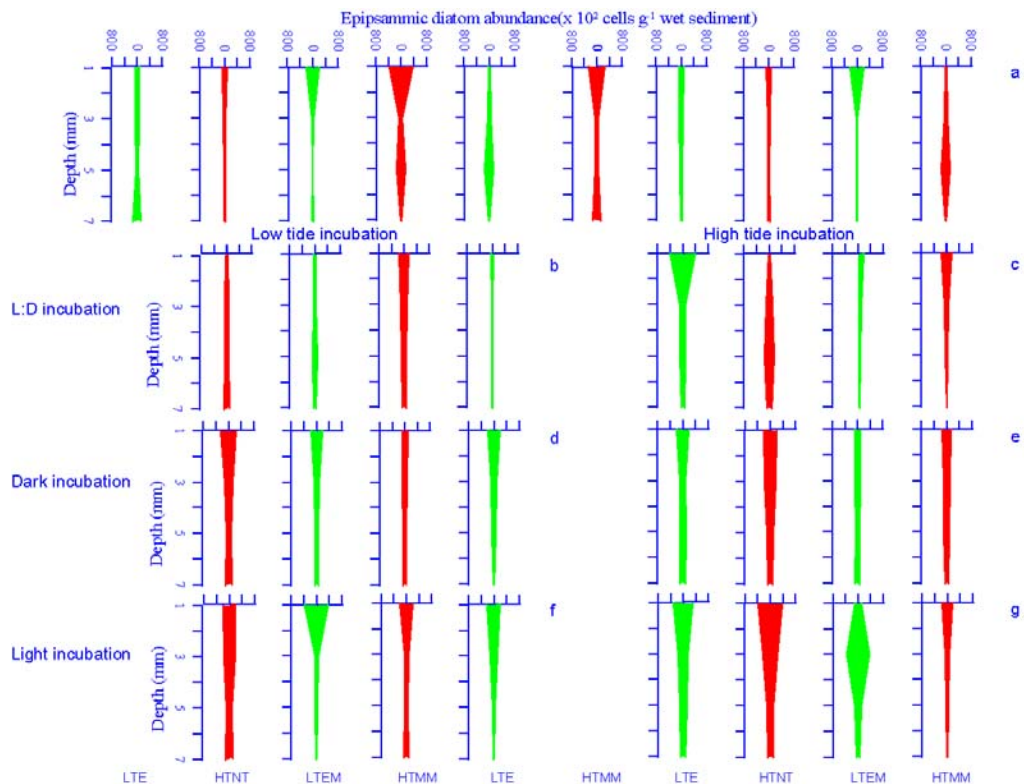


Fig. 2B.9 Depthwise distribution of mean epipsammic diatom (10^2 cells g^{-1} wet sediment) of case II sediment cores for the different tidal periods in the field (a), under controlled conditions for the “low tide” [12 h light: 12 h dark (b), continuous dark (d), continuous light (f)] and high tide incubated cores [12 h light: 12 h dark (c), continuous dark (e), continuous light (g)]

LTE=Low tide evening; HTHT=High tide night; LTEM=Low tide early morning; HTMM=High tide mid-morning

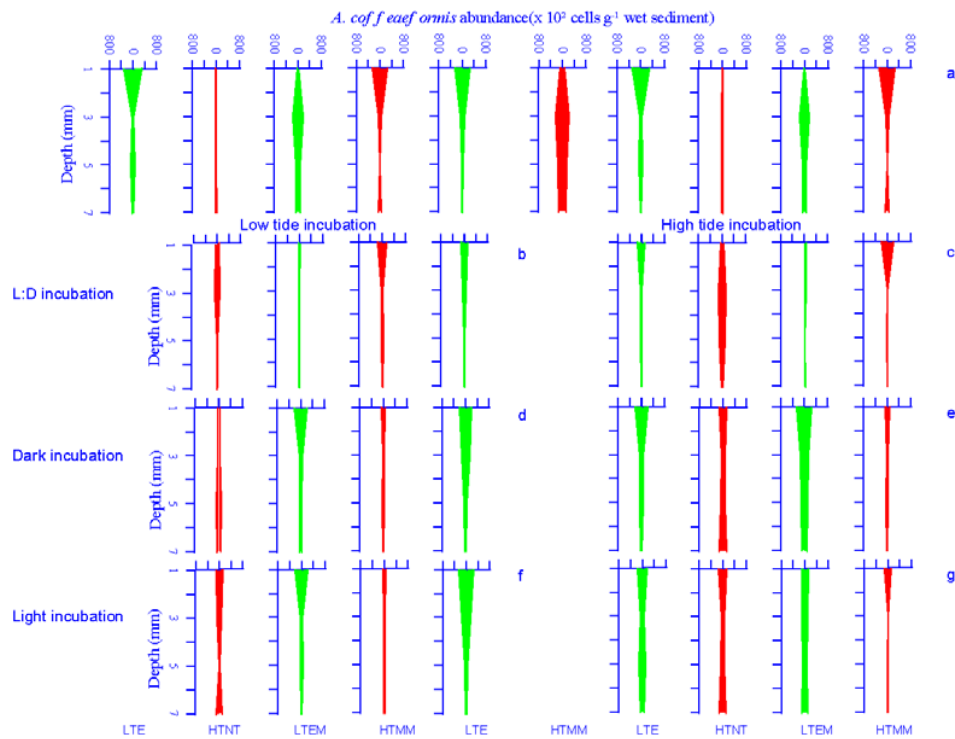


Fig. 2B.11 Depthwise distribution of *A. coffeaeformis* (10^2 cells g^{-1} wet sediment) of case II sediment cores for the different tidal periods in the field (a), under controlled conditions for the “low tide” [12 h light: 12 h dark (b), continuous dark (d), continuous light (f)] and high tide incubated cores [12 h light: 12 h dark (c), continuous dark (e), continuous light (g)]
 LTE=Low tide evening; HTHT=High tide night; LTEM=Low tide early morning;
 HTMM=High tide mid-morning

In this case, the 12 h light: 12 h dark and continuous dark incubation trend was similar as that observed for the high tide incubated cores (Fig. 2B.8b and 2B.8d). Chlorophyll *a* concentrations did not reveal a similar trend (Fig. 2B.7b and 2B.7d).

Continuous light incubation: When incubated in continuous light, the diatom population remained at the surface for a period of 18 h, i.e., mid-morning high tide to the evening low tide field exposure, to the corresponding night high tide to early morning low tide of field (Fig. 2B.8f). Chlorophyll *a* concentrations were high up to 12 h, beyond which it showed a decline (Fig. 2B.7f).

Comparison between low and high tide incubated cores

Table 2B.4. Significance values of the MANOVA evaluating the temporal variation in diatom abundance of case II along the vertical gradient (1, 3, 5 and 7mm sediment core sections) with respect to incubation conditions. (NS- Not-Significant) (a=Tides; b=Incubation time; c=Depth; d=Condition)

	12 hr light: 12 hr dark incubation			Continuous dark incubation			Continuous light incubation		
	axb	axc	bx c	axb	axc	bx c	axb	axc	bx c
	p <			p <			p <		
Total diatom abundance	NS	NS	NS	NS	NS	NS	NS	NS	NS
Epipellic diatom abundance	NS	NS	NS	0.01	NS	NS	0.005	NS	NS
<i>Amphora coffeaeformis</i> abundance	NS	NS	NS	0.025	NS	NS	0.025	NS	NS
<i>Navicula delicatula</i> abundance	NS	NS	NS	0.05	NS	NS	0.005	NS	NS
Epipsammic diatom abundance	NS	NS	NS	NS	NS	NS	NS	NS	NS
Chlorophyll <i>a</i>	NS	NS	0.05	NS	NS	0.025	NS	NS	0.025

MANOVA did not reveal any significant variation between the diatom abundance (for epipellic i.e., *N. delicatula* and *A. coffeaeformis* and epipsammic diatoms) of the low and high tide incubated cores when subjected to 12 h light: 12 h dark incubation (Fig.

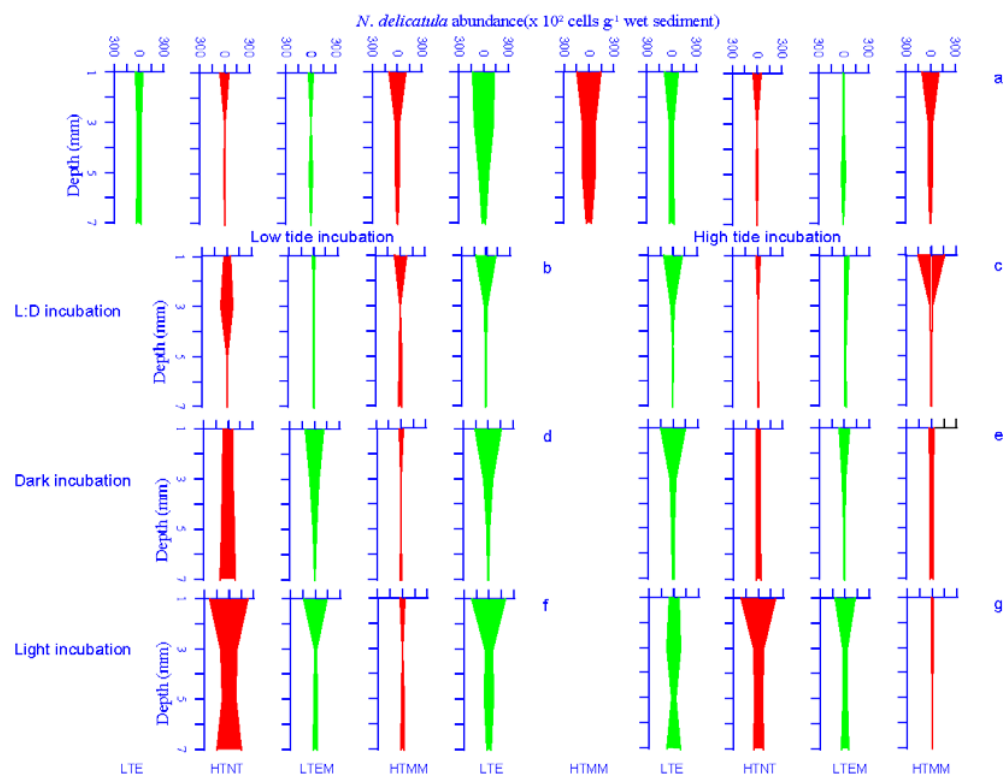


Fig. 2B.11 Depthwise distribution of *N. delicatula* (10^2 cells g^{-1} wet sediment) of case II sediment cores for the different tidal periods in the field (a), under controlled conditions for the “low tide” [12 h light: 12 h dark (b), continuous dark (d), continuous light (f)] and high tide incubated cores [12 h light: 12 h dark (c), continuous dark (e), continuous light (g)]
 LTE=Low tide evening; HTHT=High tide night; LTEM=Low tide early morning;
 HTMM=High tide mid-morning

2B.8b, 2B.8c, 2B.9b, 2B.9c, 2B.10b, 2B.10c, 2B.11b and 2B.11c). In the continuous dark and continuous light condition, epipellic diatoms (*N. delicatula* and *A. coffeaeformis*) showed significant variation between the tides and incubation time (Fig. 2B.8d-g, 2B.10d-g, 2B.12d-g; Table 2B.4).

Comparison between 12 h light: 12 h dark, continuous dark and continuous light incubations

Table 2B.5. Significance values of the MANOVA evaluating the temporal variation in diatom abundance of case II along the vertical gradient (1, 3, 5 and 7mm sediment core sections) with respect to low and high tide incubations. (NS- Not-Significant) (a=Tides; b=Incubation time; c=Depth; d=Condition)

	Low tide incubation			High tide incubation		
	dx _b	dx _c	bx _c	dx _b	dx _c	bx _c
	$p \leq$			$p \leq$		
Total diatom abundance	NS	NS	0.05	NS	NS	0.001
Epipellic diatom abundance	NS	NS	0.01	NS	NS	0.005
<i>Amphora coffeaeformis</i> abundance	NS	NS	0.01	NS	NS	0.001
<i>Navicula delicatula</i> abundance	NS	NS	0.05	0.025	NS	0.05
Epipsammic diatom abundance	0.001	NS	NS	NS	NS	0.001
Chlorophyll <i>a</i>	NS	NS	NS	NS	NS	0.001

When the diatom abundance values of the low tide and high tide incubated cores were subjected to MANOVA, the epipellic diatoms (*N. delicatula* and *A. coffeaeformis*) revealed a significant variation between incubation time and depths (Fig. 2B.8, 2B.10 and 2B.11). This variation was not significant for the epipsammic diatoms (Fig. 2B.9; Table 2B.5).

2B.4 DISCUSSION

During the morning low tide exposure, comparatively higher irradiance and lower tidal amplitude triggered the upward migration of diatoms in case I, in contrast to case II. Subsequently, availability of sufficient light for photosynthesis during the morning exposure and also, increasing light intensity and tidal amplitude, triggered the diatoms to migrate downwards just prior to the noon high tide. Round (1979b) and Admiraal (1984) have proposed that benthic microalgae are adapted to respond to excessive

irradiance levels by migrating downwards into darker layers of sediment. Nielsen et al. (1995) and Nielsen and Ekelund (1995) has demonstrated that exposure to solar ultraviolet radiation significantly affects phytoplankton photosynthesis and cell motility. The ability of microphytobenthos to migrate vertically within the surface sediment may be considered as a form of behavioral photoacclimation, allowing cells to avoid potentially damaging irradiance and temperature conditions (Kromkamp et al. 1998; Perkins et al. 2001). There is also a possibility of some part of the population being suspended into the overlying waters due to the rising tides. Case II, where the diatoms exhibited upward migration during the high tide coverage reveals the overriding influence of light over tides. During the high tide, the overlying water and turbidity reduces the amount of light available for photosynthesis, as a result of which the diatoms continued to remain at the surface even during the evening low tide. Perkins (1960), Round and Palmer (1966), Haphey-Wood and Jones (1988) and Janssen et al. (1999) have reported a similar behavior for diatoms inhabiting muddy sediments. So, migratory behavior of diatoms can persist at the sediment-water interface in the evening low tides, provided the light intensity and tidal amplitudes are favorable, as in case II. In case I, the lowered light intensity and higher tidal amplitude did not favor surface migration. The subsequent rise in tidal amplitude and absence of light triggered the diatoms towards positive geotrophy, in both case I and case II. Strickland (1958) has reported that optimal irradiance for photosynthesis varies with latitude and season, which may explain the behavioral differences between the two cases.

In this entire process of vertical migration, an important factor that needs consideration is the impact of physical forcing on the sediment caused by wind or tidal currents. Turbulence and shear stress generated by the incoming tide lead to

suspension of the diatom cells into the overlying waters (Baillie and Welsh 1980; Delgado et al. 1991; de Jonge and Van Beusekom 1992; 1995; de Jonge and Van den Bergs 1987). This may be an additional factor responsible for the lowering of the surface diatom population, other than their positive geotrophic movement during immersion. However, the entire suspended population will not be lost to water column, since a part of it will start resettling at the beginning of emersion. This process will in turn contribute to the rise in cell numbers at the sediment surface during subsequent exposure, along with the upward migration from the deeper sediment layers.

2B.4.1 Chlorophyll *a*

The high concentrations of chlorophyll *a* with a significant relation with diatom abundance (Table 2B.2) confirms the upward migration during the morning low tide in case I (Fig. 2B.2a). In case II, lack of sufficient amount of light for photosynthesis due to the water coverage during the mid-morning high tide resulted in reduced chlorophyll *a* concentrations (Fig. 2B.7a). This is evident from the non-significant relation between the chlorophyll *a* and diatom abundance (Table 2B.3). Variation in cellular chlorophyll *a* as a function of irradiance is well documented for diatoms (Durbin 1974; Paasche 1968; Brown and Richardson 1968; Beardall and Morris 1976). In the temporal conditions, Hitchcock (1980) has reported that *Skeletonema costatum* can potentially alter its chlorophyll *a* content on an hourly time scale, thereby maximizing its light harvesting ability during relatively “dark” periods. Probably, the benthic diatoms also adopt similar mechanisms to cope with the low light intensities, which are reflected in the increased chlorophyll *a* concentrations during the evening low tide (Fig. 2B.7a). Both these cases reveal the importance of

light in terms of intensity, depending on which the diatoms will decide their exposure duration.

2B.4.2 Laboratory incubations

The daily light:dark cycle is one of the strongest and most predictable environmental fluctuations that phytoplankton experience (Brand and Guillard 1981). Accordingly they must have evolved a variety of adaptations to this cycle as well as to the daily fluctuations of irradiance. Behavior of benthic diatom assemblages, similar to that observed in field, when incubated under the 12 h light: 12 h dark condition, with the tidal effects removed, confirmed the presence of an endogenous biological clock. Round and Palmer (1966) have observed such a behavior for the diatom population from muddy intertidal sediments. In the present study, the absence of suspension under controlled conditions in contrast to the natural environment was responsible for a higher density of diatom cells as compared to the field.

In continuous darkness, expression of a tidal rhythm may be attributed to an innate behavior. This behavior is otherwise superimposed by the diel rhythm in the presence of light. Odum (1970) stated that the biological clock not only couples internal and external rhythms, it enables individuals to anticipate changes. Thus, it is possible that when light is present, this tidal endogenous rhythm is manipulated according to the light intensity. Harper (1960) has reported daily migrations in continuous darkness for diatoms inhabiting muddy sediments. Round and Palmer (1966) observed that suppression of migratory cycle by pre-treatment in continuous dark had little or no effect on its subsequent expression on re-exposure to favorable conditions (i.e., when light intensity is sufficient). They suggested that the clock continued to run in the dark in spite of the fact that the overt rhythm was not displayed. Such tidal rhythms can also be an attempt to search for light, which is of prime requirement for

photosynthesis. In this search, the diatoms probably tend to produce more of chlorophyll *a*, which is a light harvesting pigment. Several workers have recorded increasing concentrations of chlorophyll *a* in the dark (or night) (Yentsch and Reichert 1963; Steeman Nielsen and Jørgensen 1968; Taguchi 1976). In the present study, this is evident from the chlorophyll *a* concentrations in case I and case II, which is in par with that produced during the other incubation conditions. This is confirmed from the non-significant variation shown by chlorophyll *a* concentrations between incubation conditions and time as well as depth (Table 2B.2 and 2B.3).

On exposure to continuous light, capability of the diatom population to remain at the surface for a period of 18 h followed by a subsequent downward migration shows that there is an optimum duration up to which the diatoms can tolerate light exposure. In both the high tide and low tide incubated cores, the cells started migrating downwards after 18 h of light exposure. But in case of low tide incubation, reduction of chlorophyll *a* after 12 hours of continuous exposure to light (Fig. 2B.7f), followed by a subsequent downward migration of epipelagic diatom cells (Fig. 2B.8f) reveals the impact of continuous light exposure on the physiology of the cells. However, this was not observed in the high tide incubation. The difference probably might be a result of the entire days exposure to natural light before the population was sampled. Whereas in case of the high tide incubated core, the population was exposed for a longer duration to a fixed irradiance in the laboratory. The physiological effect (i.e., degradation of chlorophyll *a*) might act as a trigger for the cells to become positively geotrophic. Nelson and Brand (1979) made the assumption regarding the temporal separation of metabolic processes, which has resulted in the evolution of one or more vital processes that can occur only during the dark phase and are unable to operate in continuous light.

In the natural environment, a high degree of hourly and fortnightly variability in benthic microalgal productivity has been reported by Pinckney and Zingmark (1991) and Serodio and Catarino (2000). These workers have attributed this variability to vertical migrations by epipelagic diatoms within the upper layers of the intertidal sediments. The present study suggests that vertical migration in diatoms is not strictly a tidal or a diel behavior and differs with different seasons depending on the environmental conditions. Also, the migratory behavior of the epipelagic diatoms is not related to the time of the day but to the duration of exposure to light, which is in turn dependent on the irradiance. This behavior has also been suggested by Verduin (1957) in case of phytoplankton, which after being exposed to dim light during the day can carry on high rates of photosynthesis during late afternoon hours. It is also evident that for the migratory behavior investigations, chlorophyll *a* alone cannot be used as a predictor because it is subject to change depending on the physiology of the cells, which is controlled by the prevailing environmental conditions such as light availability. Light availability is in turn controlled by the season or temporal variations. For e.g., in case II, during the mid-morning high tide the cell numbers were high but not chlorophyll *a*, since sufficient light was not available. Thus, it can give erroneous results that upward migration has not occurred. Also, unlike cell abundance, chlorophyll *a* concentrations did not show significant variations with depth. By taking, both cell abundance as well as the chlorophyll *a*, physiology of the cells can be inferred and such information can be important for primary productivity studies.

Resurfacing of diatom population, after their homogenous distribution through tidal resuspension, is observed in the field as well as in controlled conditions when subjected to 12 h light: 12 h dark, continuous dark and continuous light conditions

even in the absence of tidal action. So these benthic diatoms via vertical migration are responsible for a considerable input of organic carbon into the sediment thereby supporting various communities of heterotrophic organisms in the intertidal areas.