Diatoms of the microphytobenthic community: population structure in a tropical intertidal sand flat

Abstract Temporal and spatial variations were investigated in the viable diatom population of the microphytobenthic community from an intertidal sand flat of a tropical environment. The presence of diatoms to a sediment depth of 15 cm and their rejuvenation in culture revealed that their viability was not affected by the conditions prevailing at this depth. This depth harbored not only pennate (epipsammic and epipelagic) diatoms, some of which are permanent residents of this area, but also centric diatoms of planktonic origin. The occurrence of diatoms such as *Amphora* and *Navicula* throughout the year in the sediments indicated that the two pennate forms were natives of this area. The centric diatom *Thalassiosira*, the vegetative cells of which were not observed in the sediments, must have appeared in culture through the germination of resting stages, which are regularly carried to the study area with coastal sediments and redeposited in intertidal sediments. Nutrients did not play an important role in the case of the pennates, which mainly reside in the sediments; whereas, in centric diatoms, nitrate and phosphate positively influenced their abundance. Multiple regression analysis revealed that some of the grain size fractions served as predictors of diatoms such as *Amphora*, *Grammatophora*, *Pleurosigma* and *Thalassiothrix*. Wind stimulated the resuspension of the sediment, along with pennate diatoms, down to 5 cm depth. Correlation of chlorophyll *a* with diatom cell numbers, which has been generally used as an indicator of diatom abundance, revealed that chlorophyll *a* concentrations were good predictors of both pennate and centric diatom abundance, to 10 cm depth. However, a negative correlation between chlorophyll *a* and diatoms at the 10–15 cm depth, even when viable diatoms were found in appreciable numbers, suggests survival of these diatoms below the physical disturbance level through the adoption of survival strategies such as resting stage formation.

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 epipsamnic (attached to sand grains) or epipelagic (motile forms within sediments). Intertidal sand flats are dynamic environments, where the tidally 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 epipelagic 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 re-surfaced (Fielding et al. 1988).

In the present study, the sediment at the lowest low tide mark was investigated for the temporal variation in diatom abundance 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.

Materials and methods

Study area and sampling strategy

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 (Fig. 1). This beach is about 200 m in length. The tides are semi-diurnal, with an 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, 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 was carried out on a monthly basis for 17 months (May 1998–September 1999) so that a full cycle of the three seasons and a repetition of one season was considered. Sediment samples were collected in triplicate at the low tide mark 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 (NO$_3$-N), nitrite (NO$_2$-N), phosphate (PO$_4$-P) and silicate (Si) 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 mark for the analysis of the water parameters as mentioned above. A known volume of the 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).

Enumeration of diatoms in the sediment

Since this study was carried out on a sandy beach and included the observation of attached forms, direct examination 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 ultrasonification 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, 1990; Yamochi 1989; Ishikawa and Tamaguchi 1994; Itakura et al. 1997) was employed for quantification of the diatom flora in each section of the sediment core. Subsequent to incubation of the sediment sample, 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 (1–2 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 tenfold serial 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 photocycle. 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 tenfold dilutions) of diatoms in the sediment sample (MPN g$^{-1}$ wet sediment) was then calculated according to a statistical table (Thronsden 1978). This table covers a range of five dilution steps; 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), Subrahmanyan (1946), Desikachary (1987) and Tomas (1997).

Grain size analysis

Sediment grain size was analyzed by dry sieving (Folk 1968). Categories of grain size included: >1,000 μm (very coarse sand);

![Fig. 1 Location of the study area](image-url)