Characteristics of Diatom Communities Epiphytic on Brown Algae *Padinagymnospora* (Kutzing) Sonder, from Central West Coast of India

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Abstract: Epiphytic diatoms among marine microalgae are of special interest for their environmental, biochemical and biotechnological applications. However, seaweeds particularly from the tropics have remained unexplored for their epiphytic flora. Diatom floras associated with Padinagymnospora were evaluated for their community structure. Diatom communities were highly diversified and were represented by 53 species, observing dominant genera Navicula, Rhizosolenia, Nitzschia, Grammatophora and Caloneis from relatively less or non-polluted localities. Composition from polluted areas typically supported one community hypothesis, with dominance of Biddulphia and Licmophora. Species diversity was found to be inversely related with nutrient concentrations in ambient waters. The present study proved that epiphytic diatoms on seaweeds could serve as reliable ecological indicators.

Keywords: Epiphytic Diatoms, Seaweeds, Padinagymnospora, Ecology, Bioindicators

1. Introduction

Seaweeds form a major source of organic matter in near shore waters, and harbor various kinds of associated and epiphytic biota (Chemello and Milazzo, 2002; Ranjitham et al., 2008). Epiphytic floras of marine macrophytes prominently constitute diatoms, cyanobacteria and fungi (Dere et al., 2002; Shamsudin and Sleigh, 1995). Diatoms are of a special interest because of their attachment to living substrate and interactions among different components of habitats. Being oxygen synthesizers, they form very important constituents of aquatic habitats, contributing to about 40% of the biomass (Falkowski et al., 1986). Most of the earlier work done on epiphytic diatoms dealt with freshwater and estuarine environments (Main and McIntire, 1974; Penhale, 1977; Sullivan and Currin, 2000). A few attempts have been made to report association of diatoms with the seaweeds from temperate regions (Ulanova and Snoeijs, 2006), however such efforts have not been made from the tropics.

The limited mobility of epiphytic diatoms makes them likely to reflect long term environmental conditions for a particular area (Fore and Grafe, 2002). Therefore, they potentially can be use as bioindicators of environmental quality, and may be more effective than other conventional bio-indicators (Dixit et al., 1992; Fisher and Dunbar, 2007; Frankovich et al., 2006; Sudhakar et al., 1994). Understanding dynamics of diatom community structure would enable exploring ecological, environmental as well as biotechnological potentials of such forms.

In view of the above, community structures of diatoms associated with *Padinagymnospora* (Kutzing) Sonder, were evaluated along the central west coast of India.

Padinagymnospora, one of the predominant Phaeophyta species from the tropics, is particularly dominant along Indian coast and has an extended growing season of about 7 - 8 months (Dhargalkar et al., 2001). It is generally found in mid-intertidal zones, along rocky shores and rock pools. The erected thallus is lobed, broad and 10 - 15 cm long. The present efforts could enhance knowledge on seaweed ecology with respect to host epiphytic interrelation and productivity, and contribute towards marine biodiversity.

2. Materials and Methods

2.1. Description of study area

The study locations were along the central west coast of India, between latitude $15^{\circ} - 17^{\circ}$ N and longitude $73^{\circ} 15' - 74^{\circ} 30'$ E, a stretch of about 683 km that is indented with numerous river mouths, creeks, beaches, head lands and cliffs. Most of the coastal region is of Deccan lava, composed mostly of basalt and laterite boulders.

Based on the rich algal biodiversity and varying water quality, three Sampling stations were selected in Mumbai, Malvan and Anjuna (Goa) (Figure 1). Mumbai (18^0 54' N 72^0 48' E), a cosmopolitan mega city, has a number of sources of pollution; both industrial and sewage effluents are freely discharged in the marine waters. The Colabacoast of Mumbai represents a moderate rocky intertidal exposure. Malvan (16^0 03' N 73^0 27' E) represents a relatively better formation of rocky intertidal coast with no obvious sources for pollution, and hence the near shore waters remain relatively clear. Anjuna (15^0 35' N 73^0 44' E) in Goa has large rocky cliffs, moderate intertidal expanse, and numerous tidal poolswhich form the best suitable habitat for algal growth.

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Figure 1: Study Locations

2.2. Sampling Methodology

Hydrological and biological samples were collected monthly at selected stations (Figure 1) during November 2016 – May 2017. pH, temperature, salinity, suspended solids and nutrient concentrations (NO₃⁻, NO₂⁻ and PO₄⁻) in ambient waters were estimated using standard oceanographic techniques (Strickland & Parson, 1972). Thalli of *P. gymnospora* along with hold fasts were collected from rocky shores and tide pools in mid intertidal zones, gently transferred in polythene bags so as to minimize the loss of epiphytes, and were immediately stored at ~ $0 - 4^{\circ}$ C.

2.3. Isolation and identification of diatoms

Epiphytic diatoms on *P. gymnospora* were isolated by adopting and slightly modifying the HCl digestion methods described for aquatic angiosperms (Shamsudin and Sleigh, 1995; Tanaka, 1986). Thalli collected for identification were weighed and added to 1.5% HCl at 30° C, and rotated on shaker at 120 rpm for 20 minutes. Isolated epiphytes suspended in HCl solutions were then centrifuged at 4000 rpm for 15 minutes. Supernatants were discarded and pellets were re-suspended in 47 mm GFF filtered seawater. This procedure was repeated until most of the adhered diatoms were removed from the thalli. The final volume was adjusted to 100 ml with acidified formalin (Tomas, 1997). Thalli of *P. gymnospora* were then washed and dried at 60° C to obtain constant weight.

Cell counts were made using 1 ml of preserved samples, and 700 – 900 cells were counted from each sample. Total abundance was estimated as No. X 10^5 g⁻¹ Dry Weight (DW) of *P. gymnospora* thallus. Diatom samples were identified as described by Desikachary (1987), and Tomas (1997).

2.4. Statistical analysis

Diversity indices for each sampling station were estimated using standard formulae (Shannon, 1948). Cluster analysis based on Bray-Curtis similarity was done for total abundance data to estimate the similarities between diatom communities at study sites. The similarity of diatom assemblages, in terms of the relative abundance of the epiphytic diatom taxa on *P. gymnospora*, was estimated for each sampling date by Similarity Index (SIMI) (Stander, 1970). PRIMER 6.0 software was used for statistical analysis.

3. Results

3.1. Hydrological parameters

Box and Whisker plots were examined for nine water quality parameters to assess spatio-temporal variations in water quality (Figure 2). pH, temperature, salinity, dissolved oxygen and suspended solids were observed to be in normal range and nearly similar at all three locations. However, biological oxygen demand and nutrient values were much higher at Mumbai than that of Malvan and Anjuna.



3.2. Species richness and diversity

A total of 53 diatom species were recorded to be epiphytic on *P. gymnospora* during its growing season (November – May). Host specimens from Malvan exhibited maximum species richness (39spp), followed by Anjuna (37spp) and least (28spp) at Mumbai (Table 1). Species richness was observed to be maximum in January and February at Malvan (26spp), while from Anjuna it was observed in November (23 spp).

Licmophora and Biddulphia dominated the epiphytic diatom community from Mumbai with average percentage composition of 57.06% and 10.62%, respectively. *Rhizosoleniacurvata* was observed to be dominant on *P. gymnospora* from Malvan (22.86%) and Anjuna (41.38%). In Malvan and Anjuna, *Navicula, Nitzschia, Grammatophora* and *Caloneis* spp were also found to be dominant during the sampling period. Epiphytic diatom community at Mumbai showed totally different distribution and abundance patterns. Significant temporal variations were observed within epiphytic diatom populations from Malvan and Anjuna. However, epiphytic

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community from Mumbai was dominated by *Licmophora* and *Biddulphia* throughout the study period.

Diversity indices for epiphytic diatom species at respective stations have been presented in Table 2.Shannon Index values at Malvan and Anjuna showed rich diversity of epiphytes while at Mumbai diversity was poor.

3.3. Abundance

Maximum abundance in terms of Diatom Cells X 10^5 g⁻¹ DW of *P. gymnospora* was observed at Mumbai (19.3 X 10^5 g⁻¹ DW) during April. While, at Malvan maximum abundance was observed during November (9.04 X 10^5 g⁻¹ DW).Epiphytic diatom population at Anjuna was maximum during May (11.48 X 10^5 g⁻¹ DW).

Epiphyte abundance decreased gradually from November to May, while at Mumbai it showed sudden growth from February to April. Total cell counts of diatoms on *P. gymnospora* from Mumbai were almost double than that ofMalvan and Anjuna (Figure 3).

This could be attributed to higher nutrient concentrations in Mumbai waters.



Figure 3: Abundance of Epiphytic Diatoms

3.4. Statistical analysis

The similarity of diatom assemblages, in terms of the relative abundance of the diatom taxa on host *P. gymnospora*, was estimated for month wise samples at three locations by calculating Similarity Index (SIMI) (Stander, 1970). Very low SIMI values obtained from diatom populations studied at Malvan (Table 3). Values close to 0 indicated diatom assemblages observed on *P. gymnospora* varied significantly in each month. Whereas at Anjuna and Mumbai higher SIMI values were observed, ranging between 0.3 to 0.7. Highest similarity was observed at Anjuna between samples collected during February and March. Higher SIMI values from Anjuna is attributed to occurrence of *P. gymnospora* in tide pools.

In present study, the Bray–Curtis similarity (Bray and Curtis, 1957) was used to quantify the compositional similarity between epiphytic diatom communities on *P. gymnospora* during different months at Malvan, Anjuna and Mumbai. The analysis of epiphytic diatom communities revealed that diatom assemblages at Mumbai are significantly different from other two locations (Figure

4). Epiphytes from Malvan as well as Anjuna formed one group indicating higher degree of similarity between epiphytic diatoms.

4. Discussion

Padinagymnospora, a brown algae, is the prominent form along the Indian coast. The best season for its growth from the study region has been reported during November -May (Dhargalkar et al., 2001). The present data revealed 53 epiphytic diatoms on P. gymnospora, which is comparatively less than the aquatic macrophytes from freshwater and brackish water habitats (Armitage et al. 2006). This could be attributed to the dynamic environment along the coast, compared to those of river and estuarine environments. Salinity, temperature and nutrient concentrations play an important role in the distribution and abundance of diatoms (Schiebel et al., 2004). Nutrients, particularly nitrate, nitrite, and phosphate, greatly influence the community structure of diatoms. Less diversity was observed at Mumbai, where nutrient values were observed to be very high. However, the maximum number of diatom species and the highest biodiversity indices were estimated for host specimens from Malvan. Composition and distribution trends of diatom diversity from Malvan and Goa were more or less similar.

Diversity index is a measure of the biodiversity of an ecosystem, and such indices facilitate understanding the quality of the environment (Colinvaux, 1973), and enable conservation and utilization of living resources by creating a single annotated index of biological collections. In general, the Shannon index falls in the range of 1.5 - 3.5 for a good diversified area, and it is considered to be zero when there is no diversity. Shannon index values of Malvan and Goa were more than three, indicating rich diversity. Similarly these two stations exhibited highly even distribution of species as evenness values of these stations were very close to one (Table 2). In contrast, though abundance on *P. gymnospora* from Mumbai was found to be relatively high, the species diversity exhibited was very low.

This could be attributed to very high levels of nutrients at Mumbai (Kulkarni et al., 2010). Though higher levels of nutrients favor diatom growth, a few opportunistic species can tolerate such ambience (deJong and Admiraal, 1984). Hence, they could serve as ecological indicators. *Biddulphia* and *Licmophora* spp showed such potentials, however, further investigations are required for explicit conclusion.

Though composition of diatoms assemblages displayed wide spatial variations within all stations, well diverse communities were observed from Malvan and Goa. Specimens from Mumbai were found to be supportive of one species community hypothesis (Sullivan, 1977).

However, entire epiphytic population at Mumbai was observed to be different from Malvan and Anjuna and this could be attributed to polluted water and occurrence of pollution tolerant species at Mumbai (Dhage et al. 2006;

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Sawant and Bhave, 2014). Understanding dynamics of epiphytic diatom community structure would enable exploring ecological, environmental as well as bio-indicator potentials of such forms.



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| Diatom Species | Malvan | Anjuna | Mumbai |
|----------------------------|--------|--------|--------|
| Acnanthesbrevipes | 2.26 | 0.52 | 0.00 |
| Amphora bigibba | 0.37 | 0.00 | 0.00 |
| Amphora coffeaeformis | 0.48 | 0.09 | 0.00 |
| Amphora costata | 0.68 | 0.00 | 0.00 |
| Amphora turgida | 2.41 | 0.98 | 0.00 |
| Biddulphiaaurita | 0.00 | 0.00 | 0.68 |
| Biddulphiabiddulphiana | 0.00 | 0.39 | 9.94 |
| Caloneiscrassa | 0.57 | 0.20 | 0.00 |
| Caloneis liber | 2.58 | 0.00 | 0.00 |
| Caloneiswestii | 3.49 | 1.84 | 0.00 |
| Climacospheniamoniligera | 3.08 | 1.51 | 0.00 |
| Cocconeispellucida | 1.03 | 1.48 | 0.45 |
| Cocconeispseudomarginata | 0.60 | 1.40 | 0.70 |
| Coscinodiscusasteromphalus | 0.61 | 0.72 | 0.45 |
| Cylindrothecaclosterium | 0.54 | 0.21 | 0.77 |
| Cymbellayarensis | 0.74 | 0.00 | 0.50 |
| Diatomavulgare | 0.00 | 0.00 | 0.66 |
| Diploneisnitescens | 0.00 | 0.14 | 0.00 |
| Diploneissubovalis | 0.48 | 1.92 | 1.19 |
| Fragillariaoceanica | 0.00 | 0.00 | 1.77 |
| Fragillariopsis sp. | 0.00 | 0.00 | 1.76 |
| Grammatophorahamulifera | 2.45 | 0.00 | 0.00 |
| Grammatophora marina | 6.33 | 0.26 | 0.24 |
| Licmophoraabbreviata | 2.87 | 1.63 | 41.42 |
| Licmophoraflabellata | 2.42 | 5.42 | 15.64 |
| Mastogloiafimbriata | 0.50 | 0.59 | 0.00 |
| Mastogloiaovata | 0.25 | 3.79 | 0.00 |
| Mastogloiarostrata | 0.00 | 0.72 | 0.00 |
| Melosiranummuloides | 3.62 | 0.00 | 1.83 |
| Naviculabacillum | 4.28 | 2.79 | 0.00 |
| Naviculadirecta | 3.24 | 4.26 | 1.71 |
| Naviculadisclusa | 2.04 | 3.78 | 0.00 |
| Naviculadistans | 7.22 | 8.69 | 2.34 |
| Naviculahalophila | 3.72 | 7.01 | 2.30 |
| Naviculalyra | 0.69 | 0.00 | 0.00 |
| Navicularamossisima | 0.50 | 0.00 | 0.00 |
| Nitzschiacapuluspalae | 0.65 | 0.00 | 0.00 |
| Nitzschialongissima | 0.00 | 0.61 | 0.93 |
| Nitzschiamacilenta | 0.00 | 1.62 | 2.76 |
| Nitzschiaseriata | 2.45 | 1.55 | 0.00 |
| Pinnulariaviridis | 1.82 | 0.00 | 1.68 |
| Plagiogrammainterruptum | 2.11 | 0.00 | 0.00 |
| Plagiogrammastaurophorum | 3.68 | 0.19 | 0.00 |
| Plagiotropisvitrae | 0.97 | 0.09 | 0.11 |
| Pleurosigmaangulatum | 0.00 | 0.34 | 0.00 |
| Pleurosigmaformosum | 0.00 | 0.00 | 0.56 |
| Pleurosigmagalapagense | 0.62 | 1.06 | 0.27 |
| Pseudoeunotiadoliolus | 2.86 | 0.19 | 0.46 |
| Rhabdonemapunctatum | 0.00 | 0.39 | 0.00 |
| Rhizosoleniacurvata | 22.86 | 41.38 | 8.42 |
| Synedratabulata | 0.00 | 0.44 | 0.00 |
| Trachyneisaspera | 1.92 | 1.61 | 0.35 |
| Trachyspheniaaustralis | 0.00 | 0.19 | 0.11 |

Table 1: Percentage composition of diatom species found epiphytic on *P. gymnospora*

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| | | | 117 | 1 1 | | |
|----------|--------|----------|----------|----------|---------|---------|
| Location | Sample | Richness | Margalef | Evenness | Shannon | Simpson |
| | | | Index | Evenuess | Index | Index |
| Malvan | Nov-12 | 18 | 1.87 | 0.69 | 2.01 | 0.75 |
| | Dec-12 | 18 | 1.96 | 0.41 | 1.19 | 0.41 |
| | Jan-13 | 26 | 3.03 | 0.91 | 2.96 | 0.93 |
| | Feb-13 | 26 | 3.08 | 0.95 | 3.10 | 0.95 |
| | Mar-13 | 20 | 2.39 | 0.92 | 2.76 | 0.93 |
| | Apr-13 | 24 | 2.85 | 0.96 | 3.05 | 0.95 |
| Anjuna | May-13 | 22 | 2.57 | 0.91 | 2.82 | 0.92 |
| | Nov-12 | 23 | 2.58 | 0.85 | 2.66 | 0.90 |
| | Dec-12 | 20 | 2.28 | 0.89 | 2.65 | 0.90 |
| | Jan-13 | 20 | 2.19 | 0.49 | 1.51 | 0.52 |
| | Feb-13 | 17 | 1.84 | 0.33 | 0.95 | 0.32 |
| | Mar-13 | 10 | 0.96 | 0.21 | 0.48 | 0.17 |
| | Apr-13 | 16 | 1.68 | 0.45 | 1.25 | 0.47 |
| | May-13 | 15 | 1.76 | 0.81 | 2.20 | 0.85 |
| Mumbai | Nov-12 | 13 | 1.40 | 0.65 | 1.68 | 0.70 |
| | Dec-12 | 16 | 1.71 | 0.66 | 1.83 | 0.76 |
| | Jan-13 | 22 | 2.42 | 0.62 | 1.90 | 0.75 |
| | Feb-13 | 16 | 1.71 | 0.73 | 2.03 | 0.80 |
| | Mar-13 | 26 | 2.67 | 0.47 | 1.54 | 0.57 |
| | Apr-13 | 11 | 1.01 | 0.46 | 1.11 | 0.51 |
| | May-13 | 20 | 2.10 | 0.63 | 1.89 | 0.71 |

 Table 2: Diversity Indices of epiphytic diatom population

 Table 3: Similarity Index values between epiphytic diatom population

| | | | Malvan | | | | | |
|--------|--------|-------|--------|-------|----------|-------|--|--|
| | Nov | Dec | Jan | Feb | Mar | Apr | | |
| Dec | 0.368 | | | | <u> </u> | | | |
| Jan | 0.068 | 0.097 |] | | | | | |
| Feb | 0.043 | 0.056 | 0.050 | 7 | | | | |
| Mar | 0.027 | 0.029 | 0.027 | 0.037 |] | | | |
| Apr | 0.032 | 0.039 | 0.042 | 0.044 | 0.041 |] | | |
| May | 0.041 | 0.035 | 0.042 | 0.040 | 0.036 | 0.038 | | |
| | Anjuna | | | | | | | |
| | Nov | Dec | Jan | Feb | Mar | Apr | | |
| Dec | 0.049 | | | | | | | |
| Jan | 0.039 | 0.164 | | | | | | |
| Feb | 0.045 | 0.186 | 0.565 |] | | | | |
| Mar | 0.047 | 0.203 | 0.626 | 0.750 |] | | | |
| Apr | 0.049 | 0.167 | 0.498 | 0.596 | 0.663 | | | |
| May | 0.055 | 0.055 | 0.094 | 0.110 | 0.121 | 0.123 | | |
| Mumbai | | | | | | | | |
| | Nov | Dec | Jan | Feb | Mar | Apr | | |
| Dec | 0.241 | | | | <u> </u> | | | |
| Jan | 0.265 | 0.240 |] | | | | | |
| Feb | 0.181 | 0.189 | 0.186 |] | | | | |
| Mar | 0.320 | 0.286 | 0.301 | 0.264 |] | | | |
| Apr | 0.318 | 0.271 | 0.292 | 0.274 | 0.442 |] | | |
| May | 0.027 | 0.030 | 0.029 | 0.101 | 0.058 | 0.137 | | |

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