

Study of epiphytic diatoms on seaweeds and assessing their potential as pollution indicators

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Abstract: Epiphytic floras of marine macrophytes prominently constitute diatoms, cyanobacteria and fungi. Epiphytic diatoms are of a special interest because of their attachment to living substrate and interactions among different components of habitats. The limited mobility of epiphytic diatoms makes them likely to reflect long term environmental conditions for a particular area. Therefore, they potentially can be used as bioindicators of environmental quality, and may be more effective than other conventional bio-indicators. Earlier research on epiphytic diatoms was focused on riverine and estuarine angiosperms. Understanding ecology of epiphytic diatoms on marine algae would enable exploring their bio-indicator potentials in polluted marine habitats.

Keywords: Seaweeds, Epiphytes, Diatoms, Ecology, Bio-indicators.

1. INTRODUCTION

Growing social concern about environmental quality could be observed in recent years, both on a global and local level. This is connected with more and more convincing evidences that environmental pollution results in degradation of particular ecosystems. Emission of harmful substances has negative effects on the natural environment, human health and agricultural production efficiency. When the consequences of environmental pollution become visible, it is often too late to prevent them. Chronic toxic effects, impossible to notice at the initial stage of the process, may manifest themselves after many years.

Due to constant technological progress the natural environment undergoes numerous changes, deteriorating its quality, which often results in negative interactions between particular ecosystem components. During the biological evolution living organisms needed complex defense and adaptation mechanisms to survive under changing environmental conditions. Most of them managed to adapt to specific environments, but when their adaptability threshold is crossed they cannot survive.

Diatoms are extremely reliable ecological indicators for a variety of reasons. As living organisms, they reflect the overall health of the ecosystems they inhabit, as opposed to one-dimensional physical and chemical parameters that can change dramatically over a short period of time depending on the time of year, weather conditions, and other environmental factors [Cairns and Dickson, 1971]. Diatoms are more suitable for biological monitoring than many other organisms because of their seeming ubiquity, short generation time, sensitivity to changes in nutrient levels, and diverse assemblages [Fore and Grafe, 2002].

In view of the above epiphytic diatom communities on seaweeds from Malvan, Anjuna and Colaba were assessed for their potential as bioindicators.

2. MATERIALS AND METHODS

2.1. Description of study area

The study locations were selected along the central west coast of India, between latitude 15° - 17° N and longitude 73° 15' - 74° 30' E, a stretch of about 683 km. Based on the rich algal biodiversity and varying water quality, three Sampling stations were selected in Mumbai, Malvan and Anjuna [Figure.1]. Mumbai [18° 54' N 72° 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. Malvan [16° 03' N 73° 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° 35' N 73° 44' E] has large rocky cliffs, moderate intertidal expanse, and numerous tidal pools which form the best suitable habitat for algal growth.

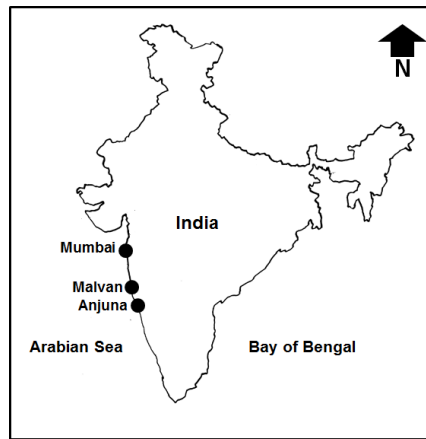


Figure 1: Study Locations along Central West Coast of India

2.2. Sampling

Hydrological and biological samples were collected monthly at selected stations during November 2016 – October 2017. pH, temperature, salinity, suspended solids and nutrient concentrations [NO_3^- , NO_2^- and PO_4^-] in ambient waters were estimated using standard oceanographic techniques [Strickland & Parson 1972]. Thalli of nine seaweed species namely *Ulva sp.*, *Chaetomorpha sp.*, *Caulerpa sp.*, *Padina sp.*, *Dictyota sp.*, *Sargassum sp.*, *Jania sp.*, *Hypnea sp.* and *Gracilaria sp.* along with holdfasts 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 $\sim 0 - 4^\circ \text{C}$.

2.3. Isolation and identification of diatoms

Epiphytic diatoms on seaweeds were isolated by adopting and slightly modifying the HCl digestion methods described for aquatic angiosperms [Shamsudin and Sleigh 1995]. Thalli collected 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.

Cell counts were made using 1 ml of preserved samples, and 700 – 900 cells were counted from each sample. Total abundance was estimated as $\text{No.} \times 10^5 \text{ g}^{-1}$ Wet Weight of thallus. Diatom samples were identified as described by Desikachary [1987] and Tomas [1997].

2.4. Statistical analysis

Correlation matrix was plotted to visualize level of similarity between various epiphytic diatom populations.

3. RESULTS

3.1. Water Quality in Study Area

In general, it was observed that seawater quality at Malvan and Anjuna was almost similar. Water temperature and pH did not show much variations at these two locations. Salinity values were highest during summer and lowest during monsoon, which is natural phenomenon. Higher values of DO and lower BOD at these two locations showed that water is comparatively free from organic pollution. On other side, seawater quality at Colaba showed very high BOD and nutrient values. Figure 2 shows comparative analysis of water qualities at Malvan, Anjuna and Colaba.

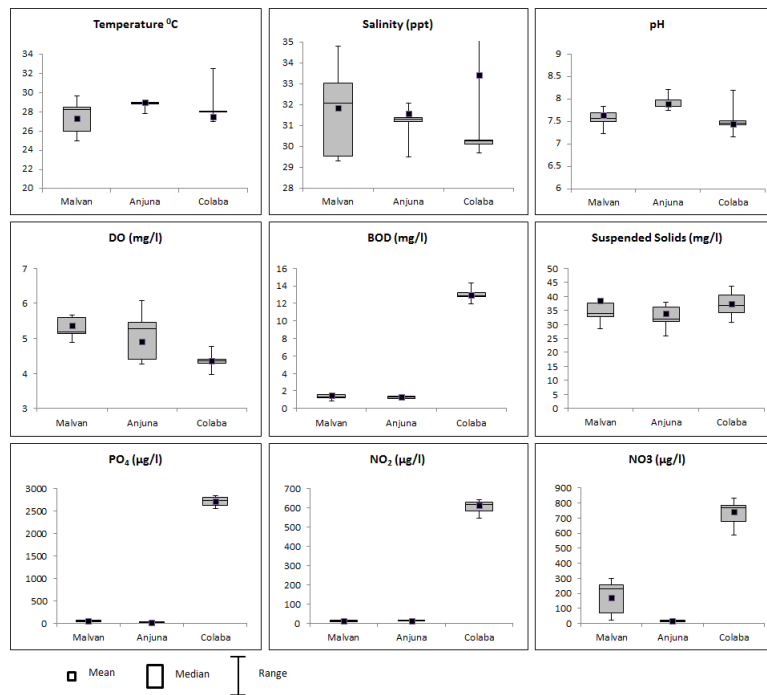


Figure 2: Water Quality in Study Area

3.2. Epiphytic Diatoms on Seaweeds

In present study *Rhizosolenia curvata* was observed to be the most dominant epiphytic species at Malvan and Anjuna. However, its distribution was principally detected in mid and lower intertidal zones. Host seaweeds like *Padina*, *Hypnea*, *Dictyota* and *Jania*, holded *Rhizosolenia curvata* as dominant epiphyte. Genera like *Navicula*, *Nitzschia* and *Licmophora* were also dominant on seaweeds from mid intertidal zone. Other genera like *Grammatophora*, *Caloneis*, *Diploneis* and *Amphora* were commonly observed. Few genera like *Cocconeis*, *Mastogloia*, *Climacosphenia* and *Synedra* exhibited seasonal dominance over few hosts.

It was observed that epiphytic diatom species prefer tranquil conditions to get attached with host. Maximum epiphytic diversity was observed on hosts like *Caulerpa*, *Hypnea* and *Dictyota* which grow in tide pools. Even total epiphytic cell count was considerably higher on these hosts. Figure 3 and Figure 4 explains comparative analysis of epiphyte richness and cell counts, respectively on all host seaweeds at Malvan, Anjuna and Colaba.

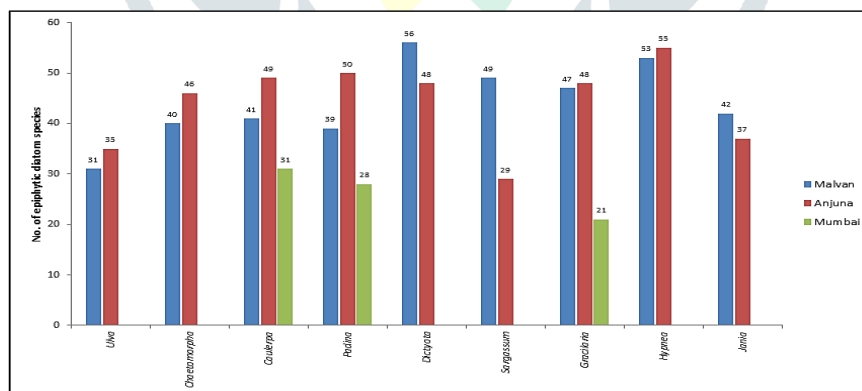


Figure 3: Number of Species of Epiphytic Diatoms on Seaweeds

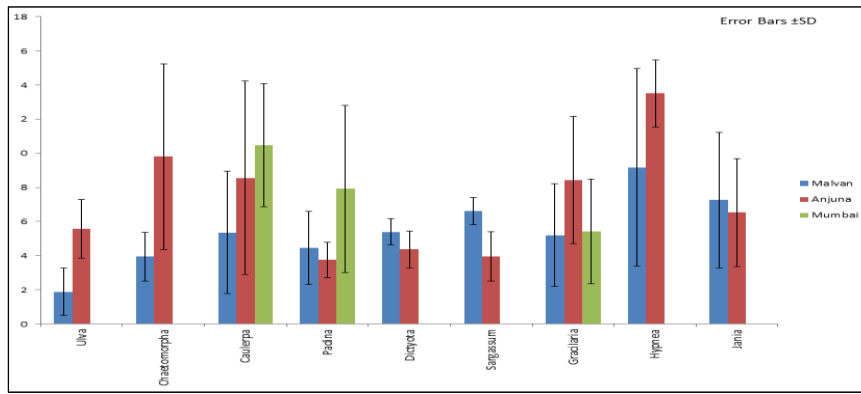


Figure 4: Total abundance of Epiphytic Diatoms as No. X 10⁵ g⁻¹ Wet Weight of thallus

Host species such as *Ulva* and *Chaetomorpha* from upper intertidal zone demonstrated dominance of *Navicula*, *Mastogloia*, *Melosira* and *Cocconeis* genera. However, *Ulva* and *Chaetomorpha* showed less epiphytic richness as well as low cell count. This could be attributed to prolonged exposure as these seaweeds occur in upper intertidal zone. Due to less submergence of upper intertidal zone, abundance of benthic diatoms could be less. Hence, number of benthic diatoms attaching to *Ulva* and *Chaetomorpha* as epiphytes was observed to be low.

Host seaweeds like *Sargassum*, *Gracilaria* and *Jania* mainly occur in lower intertidal zone where nature of water is relatively turbulent. Richness and abundance of epiphytes was relatively less on these hosts as that of hosts from tide pools. *Mastogloia*, *Navicula*, *Diatoma* and *Plagiogramma* were dominant epiphytic diatoms on *Sargassum*. *Gracilaria* and *Jania* showed dominance of *Rhizosolenia curvata*. It was observed that diatom species have a preference for host and different intertidal zones. This encouraged systematic studies of host and zone specificities of epiphytic diatoms.

Epiphytic diatom communities at Malvan and Anjuna exhibited nearly similar characteristics however, epiphytes at Colaba showed entirely different structure. *Biddulphia biddulphiana* was dominant on *Caulerpa* and *Gracilaria* while *Licmophora* spp. and *Nitzschia macilenta* was dominant on *Padina*. Distinct variation between epiphyte species composition was observed in all months. Also total cell count was considerably higher on *Caulerpa* and *Padina* as compared to Malvan and Anjuna. Epiphytic diatoms like *Biddulphia biddulphiana* and *Nitzschia macilenta* were observed only at Colaba and hence could be referred as opportunistic species, hence detailed study of epiphytic diatoms from Colaba promoted to assess their potential as bio-indicator of water pollution.

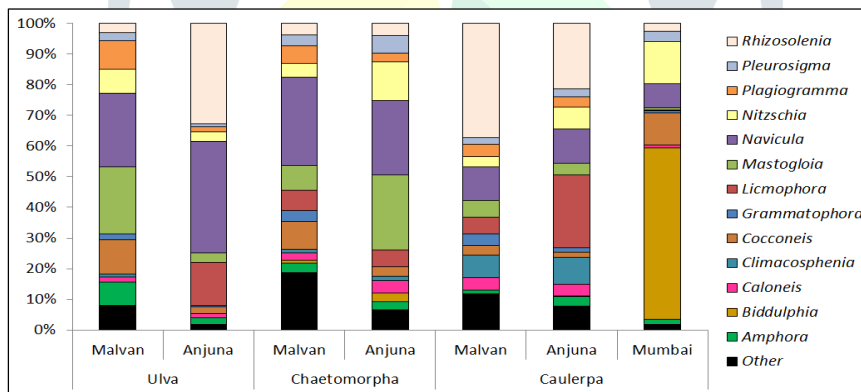


Figure 5: Composition of Epiphytic Diatoms on *Ulva*, *Chaetomorpha* and *Caulerpa*

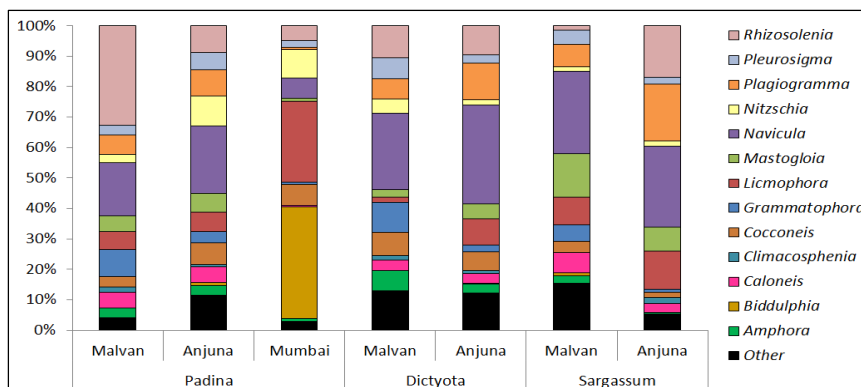


Figure 6: Composition of Epiphytic Diatoms on *Padina*, *Dictyota* and *Sargassum*

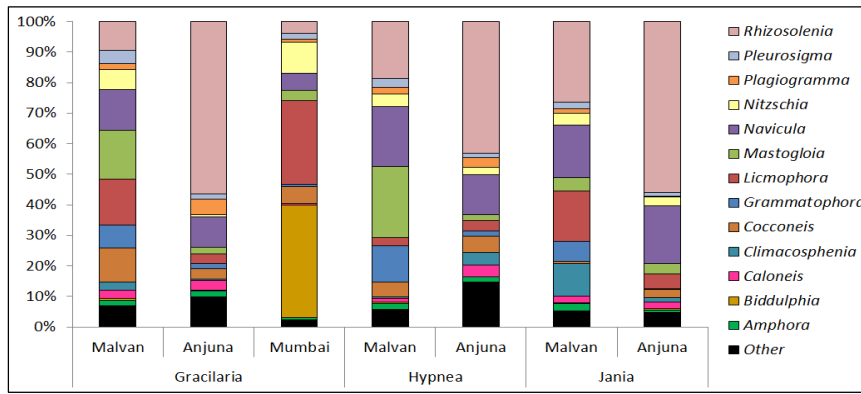


Figure 7: Composition of Epiphytic Diatoms on *Gracilaria*, *Hypnea* and *Jania*

3.3. Epiphytic Diatoms as Bio-indicators

The correlation matrix between water quality and epiphytic diatom populations is given in Table 1. The results of correlation matrix show that diatom species observed at Colaba had strong and positive correlation with nutrient parameters. *Biddulphia biddulphiana* showed positive correlation with PO₄, NO₃ and BOD [R values 0.663, 0.489 and 0.64, respectively]. It also showed negative correlation with DO [R value -0.314]. *Licmophora abbreviata* showed positive correlation with PO₄, NO₃ and BOD [R values 0.598, 0.541 and 0.596, respectively] and negative correlation with DO [R value -0.225]. *Nitzschia macilenta* showed positive correlation with PO₄, NO₃ and BOD [R values 0.378, 0.332 and 0.403, respectively] and negative correlation with DO [R value -0.231].

Table 1: Correlation Matrix of Pollutants and Dominant Epiphytic Diatom Species

L. abb	0.096											
N. mac	0.214	-0.028										
N. hal	-0.128	-0.148	0.035									
R. cur	-0.195	-0.122	-0.249	-0.195								
pH	-0.270	-0.263	-0.043	0.054	0.051							
Sal	-0.096	-0.162	0.060	0.138	0.051	0.189						
Tmp	-0.156	-0.132	0.005	-0.042	0.131	0.177	-0.004					
DO	-0.314	-0.225	-0.231	0.090	-0.034	0.008	-0.442	0.043				
BOD	0.640	0.596	0.403	-0.139	-0.254	-0.352	-0.201	-0.146	-0.394			
NO ₃	0.489	0.541	0.332	-0.186	-0.149	-0.516	-0.462	-0.097	-0.081	0.828		
PO ₄	0.663	0.598	0.378	-0.153	-0.264	-0.314	-0.185	-0.209	-0.414	0.989	0.791	
	B. bid	L. abb	N. mac	N. hal	R. cur	pH	Sal	Tmp	DO	BOD	NO ₃	

Legends: B. bid – *Biddulphia biddulphiana*, L. abb – *Licmophora abbreviata*, N. mac – *Nitzschia macilenta*, N. hal – *Navicula halophila*, R. cur – *Rhizosolenia curvata*, Sal – Salinity, Tmp – Temperature, DO – Dissolved Oxygen, BOD – Biological Oxygen Demand

4. DISCUSSIONS

Biological monitoring is valuable method used in ecological studies to protect and preserve the biological integrity of natural ecosystem, which includes preventive measures. Bioindicators of pollutants are useful in predicting the level and sources of pollution before the effects of the pollutants starts [Pai, 2002; Verma, 2002]. These organisms are generally linked to the use of mathematical distribution of these organisms in the communities to which the bioindicator species belong [Poulickova et al. 2004].

Although not new, the use of bioindicators is an innovative approach for assessing various types of environmental mismanagement, including pollution, high input farming, inappropriate disposal of wastes, contamination, etc. This approach uses biological organisms and biodiversity as tools to assess ongoing situations in the environment [Chang et al. 2014].

The organisms and organism associations are monitored for changes that may indicate a problem within their ecosystem. The changes can be chemical, physiological or behavioral. Bioindicators are relevant for ecological health. Ecological health can be viewed in terms of ecosystems, whereby structural and functional characteristics are maintained. Ecological health can be expanded to include many aspects of human health and well-being. Each organism within an ecosystem has the ability to report on the health of its environment.

Rhizosolenia curvata and *Navicula halophila* showed negative correlation with PO₄, NO₃ and BOD, indicating their less tolerance towards pollution. Hence, from correlation matrix it can be concluded that *Biddulphia biddulphiana*, *Licmophora abbreviata* and *Nitzschia macilenta* can be identified as bioindicators of pollution. These species showed high tolerance towards increased PO₄, NO₃ and BOD concentrations. Correlation matrix also showed linear and positive relationship between diatom species *Biddulphia biddulphiana*,

Licmophora abbreviata and *Nitzschia macilenta* with physicochemical parameters such as PO_4 , NO_3 and BOD. Very less scattering was observed within these parameters. Similarly linear but weakly negative relationship was observed between these diatoms and DO.

Linear and negative relationship was observed between diatom species *Rhizosolenia curvata* and *Navicula halophila* with physicochemical parameters such as PO_4 , NO_3 and BOD. Relationship between salinity, pH and all diatom species was highly scattered. The cluster analysis of epiphytic diatom communities revealed that diatom assemblages at Mumbai are significantly different from other two locations. Though composition of diatoms assemblages displayed wide temporal variations at Malvan and Anjuna, specimens from Mumbai were found to be supportive of one species community hypothesis.

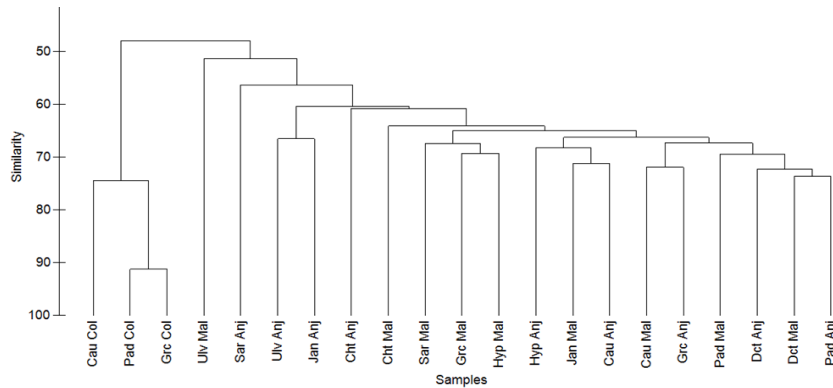


Figure 8: Dendrogram showing linkages between diatom populations

Non linear and negative relationship was observed between *Rhizosolenia curvata* with *Biddulphia biddulphiana*, *Licmophora abbreviata* and *Nitzschia macilenta*. This indicates *Rhizosolenia curvata* cannot tolerate the environmental conditions in which abundance of *Biddulphia biddulphiana*, *Licmophora abbreviata* and *Nitzschia macilenta* found to be high. Hence *Rhizosolenia curvata* could act as bioindicator for cleaner water. *Licmophora abbreviata* and *Nitzschia macilenta* though showed positive relationship with pollutants, they were occasionally observed at Malvan and Anjuna. However, *Biddulphia biddulphiana* exclusively occurred at Colaba and dominated most of the epiphytic population. Hence, it could act as bioindicator for polluted water.

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