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Morpho anatomical and elemental composition of certain seaweeds on the intertidal rocks at Nochiyurani coast of Gulf of Mannar

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ABSTRACT

The present study deals with the morphological and anatomical observations made on three species of seaweeds *viz., Sargassum polycystum*, *Padina gymnospora* and *Dictyota dichotoma* occurring on the intertidal rocks at Nochiyurani coast. Further, deep inventerization of vegetative part, were subjected to SEM-Energy Dispersive Spectroscopic analysis and quantified the following minerals *viz.,* Na, Mg, Si, S, Cl, K, Ca, Mn, P, Fe, Zn and Cr during summer and pre monsoon seasons between April 2016 to September 2016. Significant seasonal variation in the elemental accumulation of the algae was observed. Seasonal distribution of elemental composition in the seaweeds showed that most of the minerals were peak during the summer followed by pre monsoon season. This could perhaps be due to the ambient concentration of these minerals, which were high during these seasons thereby facilitating their uptake by seaweeds. The elemental composition studies of the three species showed better result in *S. polycystum* and *Padina gymnospora*.

Introduction

Seaweeds generally grow in the intertidal and subtidal regions of the sea up to a depth where sufficient light intensity is available for photosynthetic activity. The seaweed ecosystem provides habitat for a variety of invertebrate and vertebrate animals. Hence, from ecological and economical points of view it is an important marine realm. Nochiyurani coast (09°16.016¢N; 78°02.043¢E) is located near Mandapam coast in the Gulf of Mannar. The intertidal region of Nochiyurani coast is dominated by beach rocks which were formed from lithification by calcium carbonate sediment in the interdial and spray zone. The hard substratum of the rocks favours for the growth of diverse marine macro algal species. Seaweeds have rich source of minerals, especially macro and microelements necessary for human nutrition. The nutritional properties of seaweeds are usually determined from their biochemical composition *viz*., protein, carbohydrates, vitamins and amino acids etc. (Darcy-Vrillon, 1993; Mabeau and Fleurence, 1993). Seaweeds are used as regular components of diet and consumed regularly by the coastal people. Significant amount of seaweeds are harvested world wide for the production of phycocolloids. In the present study an attempt was made on the morphology and elemental composition of the seaweeds were subjected to scanning electron microscopy with energy dispersive spectroscopic analysis and the results of these studies are discussed in this paper.

Materials and Methods

Preparation for morphological studies

Seasonal changes in elemental composition has been studied for a period of six months from April 2016 to September 2016. Physico-chemical characteristics of seawater samples collected during different seasons were analysed. Marine algal samples of *Sargassum polycystum*, *Padina gymnospora* and *Dictyota dichotoma* were collected from Nochiyurani coast during low tide at 0.5 m depth. They were sorted out species wise, placed in polythene bags containing seawater and transported to the laboratory. Then they were fixed in 4% formaldehyde solution for taxonomic studies. The seaweeds were identified using the taxonomic keys provided by Srinivasan (1973) and the nomenclature was updated using the website of Appeltans *et al.* (2012) and Guiry and Guiry (2012).

Preparation for anatomical studies

The algal specimens were carefully examined before processing for anatomical studies. All the specimens were examined under a dissection microscope and the external morphology of the thallus was studied. Free thin hand sections

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were taken. The sections were mounted on the slide with glycerin and observe under the microscope.

Mounting

The slide were taken out and mounted using the DPX mountant and 25 x 50 mm cover glasses. The excess of DPX mountant was removed and the slides were carefully indexed. Sometimes glycerine mounted stain using safranin. *Camera lucida* drawing on the anatomical features of the thallus, stipe, air bladder, receptacle and important stages of various reproductive structure were made photomicrographs.

Materials preparation for SEM-EDS study

Thalli of *Sargassum polycystum, Padina gymnosppora* and *Dictyota dichotoma* algal species of 2-3 mm were fixed in 3% glutaraldehyde for scanning electron microscopic studies. Then they were dehydrated through a graded series of alcohol with 12-15 min interval at 4°C upto 70%. They were further dehydrated in 90% and 100% of alcohol and kept at room temperature for 2-3 hours. Finally, the dehydrated samples were treated with critical point drier (CPD). They were then mounted on a stub and the specimens were coated with gold. They were examined using JOEL JSM-56010 LV with INSA-EDS and photomicrographs were taken selectively from computer screen at the central sophisticated Instrumentation Laboratory, Department of Physics, Annamalai University, Annamalainagar, Tamil Nadu.

Estimation of physico-chemical parameters

Physico-chemical parameters of seawaters were analysed for the estimation of temperature, salinity, pH and DO using water and soil analysis kit model 1160-E. Further statistical tools of correlation matrix between different physico-chemical parameters of sweater samples during summer, and pre monsoon period were analyzed.

Results and Discussion

Sargassum polycystum

Morphological characters

Thallus 35 cm tall, with yellowish brown colour, attached with discoid holdfast. Main axis cylindrical and rough due to the presence of numerous outgrowth, supporting alternately arranged branches bearing leaves and vesicles.

Anatomy

Meristoderm

It is a peripheral layer of columnar cells without any air spaces between them. The cells of this layer contain abundant plastids and the fucosan granules. This plastid layer of cells constitutes the meristoderm. It is concerned with photosynthesis.

Cortex

Within the hypodermal layer is the cortex consisting of thick walled polygonal cells. The cortex may be several layer thick. It probably functions as the storage region and plays a mechanical role.

Medulla

The central zone of the thallus is called the medulla. The cells in this region are loosely arranged. They are narrow and elongated the outer cells of the medulla have thick walls whereas the inner have thin walls (Plate-1).

Padina gymnospora

Morphological characters

Plants brown, darken at the base, robust, upto 20 mm across, fan-shaped lobes that split into wedge-shaped pieces. Blade edges are in rolled upwards and inwards. Concentric bands of hairs and sporangia prominent (Plate-2).

Anatomy

The zones of hairs occur alternately on the two surfaces but are more strongly developed on the upper and are stated to be specially prominent in plant exposed to strong light. The cells are more are less isodiametric and with iodine. The longitudinal section of the thallus from the apex to the interior shows circinate coiled vernation. These cells are in the protection of the apical cells. On the thallus surface, trichomes and sproangia or gametangia of two kinds are found on the older parts.

Dictyota dichotoma

Morphological characters

Dictyota is the genus of the family with a flat, ribbon like dichotomously branched thallus. It also occurs along the Indian coast, usually in the rocky tide pools submerged and attached to rocks with the help of discoid holdfast (Plate-3). It is Yellow brown in colour andt attains a height of about 30 cm and show differentiation into three parts namely the frond, the stipe and the hold fast.

Anatomy

It is the concern the sexual or gametophyte plants. They are dioecious (or) heterothallic. The oogonial sori and deep brown in colour.

Antherdia

The antheridial sori look like white, shining spots on the surface of the male plants. The number of antheridia in a sorus varies from 100-300 mm. The marginal vegetative cells of the antheridial sorus grow upwards to form a cup-shaped involcure around it. The mature antheridium is a large colourless structure. It consist of a small basal stalk cells and a body. The body of antheridium is a multicelular structure compound of small, cubical cells called the locules.

Physico-chemical parameter such as atmospheric temperature, surface seawater temperature, salinity, pH and

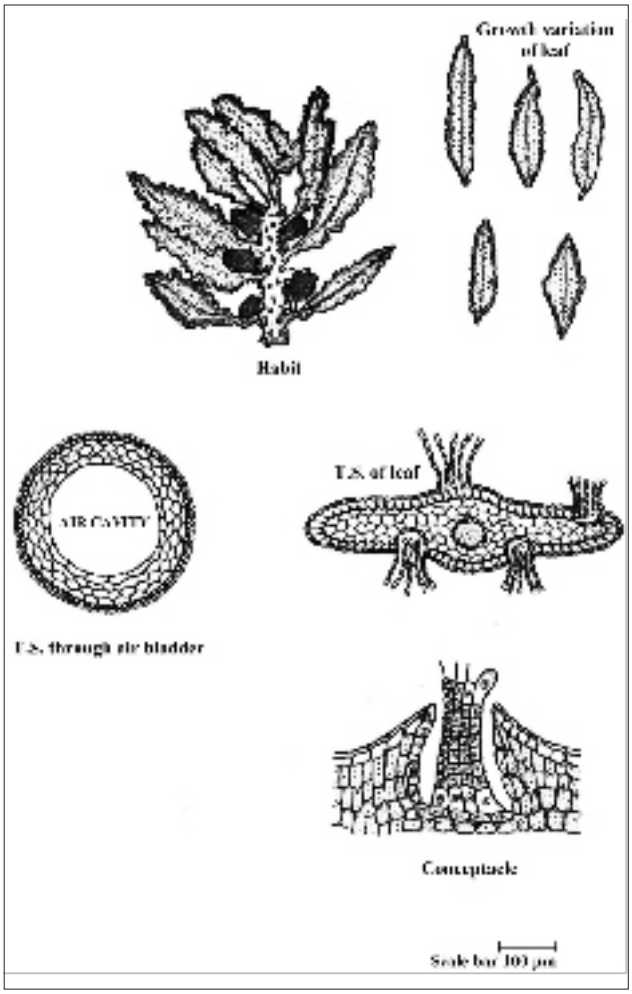

Plate-1. *Morphoanatomical features of* Sargassum polycystum

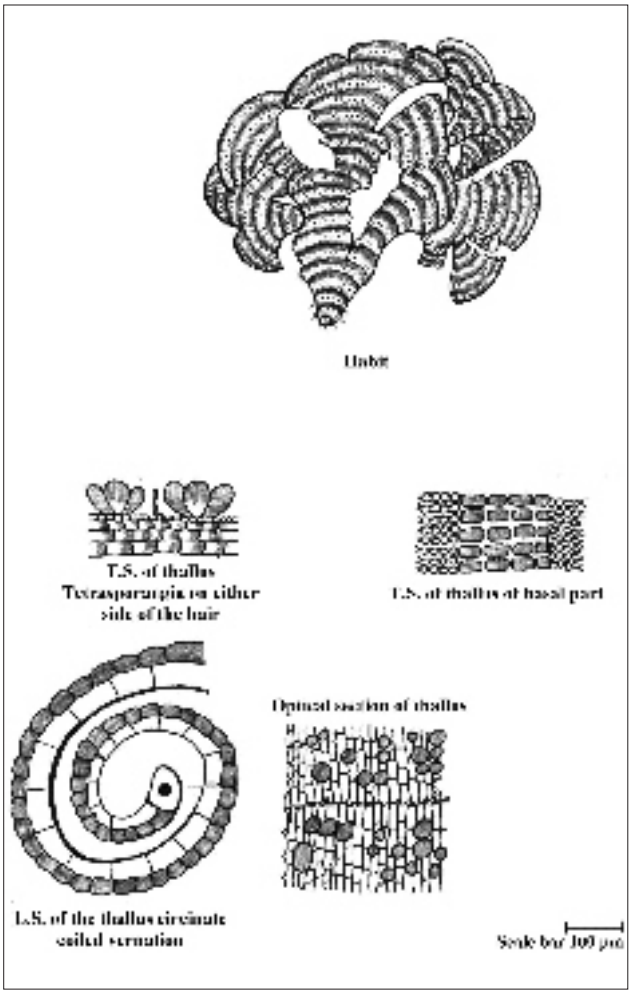

Plate-2. *Morphoanatomical features of* Padina gymnospora

DO are presented in Table-1 and 2. The seasonal variation in surface water temperature and air temperature invariably showed high values during summer and low in pre monsoon. The air temperature varied from 33.6 to 36.0°C. The surface seawater temperature varied from 33.1 to 34°C. The salinity of seawater showed high values during summer and low values in monsoon period. The salinity varied from 33 to 34.2‰. The pH of seawater was high during summer and pre monsoon seasons. The pH varied from 7.1 to 7.5. The DO varied from 4.1 to 4.7 ppm with high value during summer season.

Analysis of the different elemental composition of the three seaweeds using SEM-EDS was carried during the different seasons from April 2016 to September 2016. The experiment was carried out for two seasons *i.e.* summer and pre monsoon. The result of the study showed distinct variation in the elemental composition of the seaweeds analysed. The percentage composition of various elements in the following order in the three seaweeds.

Sargassum polycystum - Ca > Si > Mg > S > Mn > P > Na; *Padina gymnospora* - Ca > Cl > Mg > Cr > Zn > Na > Fe > P > Mn > S > K and *Dictyota dichotoma* - Ca > Cl > P > Si > Cr > Na > Mg > K (Table-3 and 4).

Seaweeds collected during summer season showed maximum contribution of calcium. They were ranging between 41.70 ± 0.20 to 32.85 ± 0.22 total weight in *Sargassum polycystum* and *Dictyota dichotoma* respectively. Minimum values varied among species during pre monsoon period. In *Sargassum polycystum* least values were obtained for K (1.39 ± 0.08), Mn (1.30 ± 0.07), *Dictyota dichotoma* K (1.82 ± 0.07) (Table-3 and 4).

The station selected for the present study is located in the Tamil Nadu coast. The atmospheric temperature fluctuations are suggested to be Ocean's thermal which changes the lay between absorption and release of solar energy to the atmosphere (Varadachari *et al.,* 1987). Atmospheric temperature at Nochiyurani coast had positive correlation with surface seawater temperature, pH and salinity but negatively correlated with dissolved oxygen (Table-1). The maximum surface water temperature at Nochiyurani coast (34 °C) was recorded during the summer season in April 2016 and

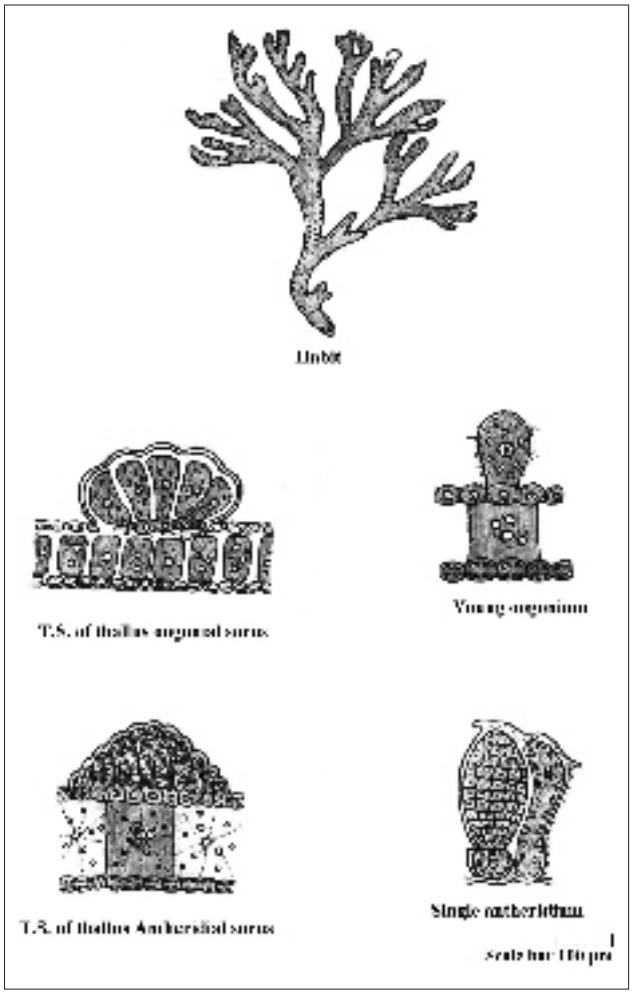

Plate-3. *Morphoanatomical features of* Dictyota dichotoma

the minimum temperature 33.1 °C was observed at the end of monsoon season in September 2016. Difference in surface water temperature pattern is quite evident as surface seawater temperature depends on the solar energy input and it seems to be influenced by several environmental conditions and factors such as inflow of freshwater, solar radiation, warming, evaporation, cooling, wind pattern and mix up with flow from adjoining neurotic waters. Similar seasonal variations were reported from east coast of India by Thangaraj *et al.* (1979).

The monthly surface seawater temperature fluctuation are different for the west coast of India as compared to that of the east coast of India (Murthy *et al.,* 1978; Untawale *et al.,* 1989; Stella Roslin and Lazarus, 2001). However, there is a significant positive correlation between atmospheric temperature and surface seawater temperature in the east and west coasts of India (Ramesh Kumar and Fernandes, 1994). pH remained alkaline throughout the study period at Nochiyurani with maximum values during the summer season. These observations supported the earlier report of Gouda and Panigrahy (1993) from Rushikulya estuary. The uptake of CO_2 by the photosynthesizing organism especially phytoplankton from the estuarine water could have increased the pH levels during the summer season. In general, pH was low during the pre monsoon season and this was associated with lesser salinity regimes.

Distribution of minerals such as Ca, Mg, Na and Cl is high in the selected species of seaweeds in the study area. Significantly higher concentration of elements such as Ca, Mg and K is encountered in the various type of seaweeds during summer and pre-monsoon periods which reflect the capacity of these seaweed to accumulate more amount of elements during

Table 1. *Correlation matrix between different physico-chemical parameters of seawater samples during summer (April-June 2016)*

	Air temperature (°C)	Surface water temperature (°C)	Salinity (‰)	pH	Dissolved oxygen (ppm)
Air temperature (°C)	1.000**				
Surface water temperature (°C)	1.000**	1.000**			
Salinity (‰)	0.800*	-0.500*	1.000**		
pH	1.000**	1.000**	-0.500*	1.000**	
Dissolved oxygen (ppm)	0.500*	0.500*	-1.000**	0.500*	1.000**

**Significant at 1% level; * Significant at 5% level

Table-2. *Correlation matrix between different physico-chemical parameters of seawater samples during pre monsoon (July-September 2016)*

	Air temperature (°C)	Surface water temperature (°C)	Salinity (‰)	pH	Dissolved oxygen (ppm)
Air temperature (°C)	1.000**				
Surface water temperature (°C)	0.970*	1.000**			
Salinity (‰)	-0.866*	1.000**	1.000**		
pH	1.000**	0.866*	0.866*	1.000**	
Dissolved oxygen (ppm)	1.000**	0.866*	0.866*	1.000**	1.000**

**Significant at 1% level; * Significant at 5% level

Table-3. *Elemental composition of seaweeds using SEM-EDS during summer (April-June 2016)*

Seaweeds	Minerals (wt%)												
	Na	Mg	Si	S	Cl	K	Ca	Mn	P	Fe	Zn	Cr	Total
Sargassum polycystum	5.40 ± 0.14	12.71 ± 0.10	19.20 ± 0.08	11.43 ± 0.13	-	-	41.70 ± 0.20	6.82 ± 0.08	2.73 ± 0.07	-	-	-	99.99 ± 0.23
Padina gymnospora	8.47 ± 0.11	15.27 ± 0.19	14.84 ± 0.16	5.43 ± 0.07	17.42 ± 0.19	1.48 ± 0.06	25.13 ± 0.20	3.81 ± 0.08	5.64 ± 0.12	2.47 ± 0.75	-	-	99.96 ± 0.12
Dictyota dichotoma	7.94 ± 0.09	4.65 ± 0.07	11.0 ± 0.09	-	20.93 ± 0.14	3.67 ± 0.08	32.85 ± 0.22	-	13.05 ± 0.10	-	-	5.88 ± 0.07	99.97 ± 0.020

Values are expressed as the mean SD; n = 3

Table-4. *Elemental composition of seaweeds using SEM-EDS during monsoon (July-September 2016)*

Seaweeds	Minerals (wt%)												
	Na	Mg	Si	S	Cl	K	Ca	Mn	P	Fe	Zn	Cr	Total
Sargassum polycystum	2.14 ± 0.10	9.53 ± 0.08	17.98 ± 0.13	8.66 ± 0.12	3.82 ± 0.07	1.39 ± 0.08	29.26 ± 0.31	4.34 ± 0.08	2.64 ± 0.09	1.44 ± 0.06	12.46 ± 0.11	6.31 ± 0.04	99.97 ± 0.23
Padina gymnospora	6.11 ± 0.13	11.32 ± 0.13	10.81 ± 0.09	2.93 ± 0.08	16.41 ± 0.19	2.09 ± 0.05	21.16 ± 0.17	4.50 ± 0.08	4.93 ± 0.07	5.15 ± 0.06	6.61 ± 0.08	7.94 ± 0.12	99.96 ± 0.33
Dictyota dichotoma	5.10 ± 0.11	3.87 ± 0.11	9.14 ± 0.12	2.62 ± 0.06	19.36 ± 0.17	2.80 ± 0.09	25.77 ± 0.23	1.30 ± 0.07	9.87 ± 0.12	8.97 ± 0.07	5.45 ± 0.11	5.71 ± 0.08	99.96 ± 0.19

Values are expressed as the mean SD; n = 3

these seasons. The concentration of Na was also found to be high during summer and pre-monsoon seasons which coincides with peak period of growth (Jayasankar and Kulandaivelu, 1999). Moreover, differences in element concentration of seaweeds in the study area, during the various seasons might be related not only to different mineral level in ambient water but also due to different ecological conditions such as tidal range, temperature and salinity (Munda and Hudnik, 1991).

Generally, bioaccumulation of the elements in the seaweeds depends upon the pH, salinity, dissolved O_2 and the osmotic potential of the system (Styron *et al.,* 1976). Marine algae exhibit high content of ash (De Boer, 1981) mainly due to the presence of Na, K, Ca and Mg cations (Fe and S will be of minor importance). Higher accumulation of Mg and Fe was mostly observed in seaweeds during summer season. It may be explained that the accumulation of elements *in situ* was more due to the reduction in osmoregulation activities usually affected by the increase in salinity. Though the silicon and chloride concentrations in ambient water exceeded those of other elements, the accumulation in seaweeds was very low and the most preferred being Ca, Mg and Na. This probably is due to the fact that Ca and Mg are the predominant occupants of the uptake binding sites of the seaweeds which would inhibit accumulation of silicon and chloride by their competition for binding sites (Foster, 1976). The elements constituents of the cell wall, plastid and Floridean starch of *Hypnea musciformis* were reported by Sivakumar and Rengasamy (1999) using SEM-EDS.

The enhanced bioaccumulation of most of the elements in seaweeds during summer and pre-monsoon seasons could perhaps be due to the following reasons: (i)ambient concentration of these elements was high during these seasons thereby facilitating their uptake by the seaweeds, (ii) seasonal variation in mineral content in seaweed may be related to growth rates and metabolic process (Munda and Hudnik, 1991). Myklestad *et al.* (1978) and Eide *et al.* (1980) reported quick uptake of elements during summer and slow uptake during winter and (iii) ecological implications are important in metal uptake by seaweeds (Kremer and Munda,

1982). This was evident as dissolved oxygen and pH of the water samples during various seasons in the present study showed the variations of correlation between various elements in the seaweeds. The present findings showed maximum values of oxygen during summer and minimum during the winter. The higher values of oxygen during summer (April-June) are associated with the rise in seaweed population (Bhatt and Negi, 1985). These observations are in agreement with those of Chan (1989) that the seasonal variations of the mineral concentration in aquatic biota may be due to seasonal fluctuation in tissue mass and changes in physico-chemical characteristics of the surrounding water. The present study reveals that the rocky formation occurring along the entire intertidal region of Nochiyurani offers good substratum for the rich growth of different seaweeds. The lack of anthropogenic activity along this coast allows the luxuriant growth of many marine algal species and provide rich source of elemental composition.

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