QUANTIFICATION OF PHOTOSYNTHETIC PIGMENTS OF PLANTS, WATER AND SEDIMENT SAMPLES IN CHIRACKAL AND KATTIPARAMBU OF ERNAKULAM DISTRICT, KERALA

ABSTRACT

Aims : The present study intended to investigate the pigment composition of four selected mangrove plants viz., *Avicennia officinalis, Excoecaria agallocha, Rhizophora mucranata and Sonnaratia alba*, water and sediment samples.
Place and Duration of Study: Chirackal and Kattiparambu of Ernakulam District, Kerala. Duration from 2013 December to 2015 December.
Methodology: The pigment concentration of the biotic samples, water and sediments were estimated using standard method in Spectrophotometer.
Results: Plants showed high pigment concentration compared to water and sediments. High Chlorophyll ‘a’ (2%), Chlorophyll ‘b’ (0.8%) and Total Chlorophyll (2.74%) were observed in *Excoecaria agallocha* of Kattiparambu and Carotenoids (0.72%) observed in *Rhizophora mucranata,* Chirackal. In sediment samples, high Chlorophyll ‘a’(0.85%), Total Chlorophylls (1.31%) and Carotenoids (0.725%) were reported in Chirackal area and Chlorophyll ‘b’ (0.595%) reported in Kattiparambu. Chlorophyll ‘b’ (0.6%) and Carotenoids (0.86%) were reported high in the water samples of Kattiparambu region and Chlorophyll ‘a’ (0.61%) and Total Chlorophylls (0.86%) in Chirackal. Plants showed strong positive correlation (1,0.999,0.998 and 0.997) among themselves, with water and sediment and also very strong positive correlation (0.998 and 0.997) with sediment samples between the two media implying common source of plants and sediments.
Conclusion: Seasonal changes and local geological conditions are the major factors for variations in pigment concentrations in plants, water and sediment samples.

Keywords: Chlorophyll, Carotenoids, Pigments, Sediments, Mangroves, Correlation.

1. INTRODUCTION

Total leaf pigment includes chlorophyll-a, chlorophyll-b and carotenoids that are necessary for photosynthesis process. Variation in leaf pigments (chlorophylls and carotenoids) and its relation can be due to internal factors and environmental conditions. Chlorophyll and carotenoids content varied with microclimatic conditions in species (1). The ratio of chlorophyll-a and chlorophyll-b in terrestrial plants has been used as an indicator of response to light shade conditions (2). The small proportion of chlorophyll a/b is considered as sensitive biomarker of pollution and environmental stress (3). Acetone gives very sharp chlorophyll absorption peaks and has great merit as the solvent for assay of chlorophylls (4). Chlorophyll is a pigment that has a clear impact on the spectral responses of plants, mainly in the visible spectrum portion. N is a key element in chlorophyll, therefore is usually a high correlation between them (5).

Previous studies indicated that chlorophyll pigments have antioxidant, anti inflammatory and wound healing properties. It has been observed that chlorophyll pigments contain chlorophyllin which is responsible for increasing the number and activity of dominant immune cells like Bcells, T- cells and macrophages essential to human health (6,7). Quantification of pigments in extracts from heavy metal-stressed plants. It has been found that under conditions of heavy metal stress the central ion of Chlorophylls and Mg_2, can be exchanged by heavy metals, which causes inhibition of photosynthesis and thus constitutes an important damage mechanism in stressed plants.

2. MATERIAL AND METHODS

Collection of Samples : Fresh leaf samples were washed thoroughly first in tap water followed by distilled water in the laboratory, kept to dry in room temperature and ground in an electric mixer (8). Then analyzed for the determination of chlorophylls (Chl-a and Chl-b) and carotenoids content. Water sample collected from three locations of Kattiparambu and Chirackal area in clean sampling bottles. For chlorophyll estimation, sample was collected from the sub surface water in sampling bottle and add 1 ml saturated $MgCl_2$ per litre of sample and keep in chill condition, then used for analysis. Sediment also collected in polythene bags from three locations of these two areas, dried, powdered and used for the analysis.

Estimation of pigments

The amount of chlorophyll present in the leaves was estimated by the standard method (9). 500 mg of leaf tissue was kept in a pestle and mortar with 10 ml of 80% acetone and it was ground well and the homogenate was centrifuged at 3000 rpm for 15 minutes and the supernatant was stored for analysis. In water and finely powdered sediment sample, 90% acetone was added, mixed and kept for overnight at low temperature under dark for extraction. The supernatant extract was centrifuged at 2000 to 3000 rpm to get clear solution and the solution was used for analysis. Absorbance of the samples were measured at 645nm, 663nm and 480nm in a spectrophotometer. The chlorophyll content was determined by using the following formula,

$$\text{Chlorophyll a (mg/g.fr.wt)} = \frac{12.7 \times A663 - 2.69 \times A645}{a \times 1000 \times W} \times V$$

$$\text{Chlorophyll b (mg/g.fr.wt)} = \frac{22.9 \times A645 - 4.68 \times A663}{a \times 1000 \times W} \times V$$

$$\text{Total Chlorophylls (mg/g.fr.wt)} = \frac{20.2 \times A645 + 8.02 \times A663}{a \times 1000 \times W} \times V$$

$$\text{Carotenoids (mg/g.fr.wt)} = A480 + (0.114 \times A663) - 0.638 \times A645$$

Where,

A - Absorbance at respective wave length
a - Length of path in the cell
W - Fresh weight of sample (g)
V - Vol. of extract (ml)

3. RESULTS AND DISCUSSION

Chloropyll a (Chl a) is a ubiquitous pigment and can be used as a global biomass indicator. In the present study, pigment level of plants gave good results when compared to water and sediments. *E. agallocha* in Kattiparambu showed high range of Chl.a and Chl.b (2.01% and 0.804%). Total chlorophylls were found to be higher in *E. agallocha* (2.74%) of Kattiparambu, and lower in *E. agallocha* (1.09) of Chirackal. Similarly, carotenoids were measured to be higher in leaves of *A. officinalis* (0.72%) and *E. agallocha* (0.76%) of Chirackal and Kattiparambu respectively, minimum levels of caretenoid was present in *R. mucranata* (0.48%) of Chirackal compared to other plants (Figure-1). The change in the carotenoids and tocopherols during seed maturation of *Cassia* species is studied (10). Water and sediment samples of Chirackal showed high chl.a (0.61% and 0.83%) and total chlorophyll (1.074% and1.31%) contents. High range of chl.b in water (0.61%) and sediment (0.85) was reported from Kattiparambu. High range of carotenoids (0.86%) reported in Kattiparambu water and sediment carotenoids (0.73%) from Chirackal (Figures 2 &3). As a whole, this study revealed the influence of abiotic factors on the growth, reproduction and development of plants. Carotene pigments were the most important photosynthetic pigments, and they prevented chlorophyll and thylakoid membrane from the damage of absorbed energy by peroxidation (11). Chlorophyll is the most indispensable class of primary compounds as they are the only substances that capture sunlight and make it available to plant system for its cultivation on photosynthesis (12).

Figure 1 :Pigment content in Plant Samples (% value)

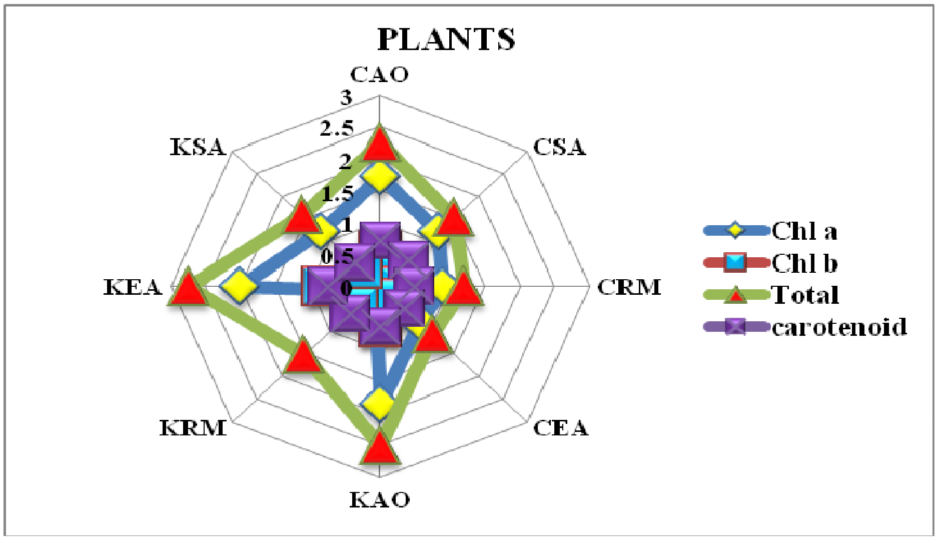


Figure 2 :Pigment content in Water Samples (% value)

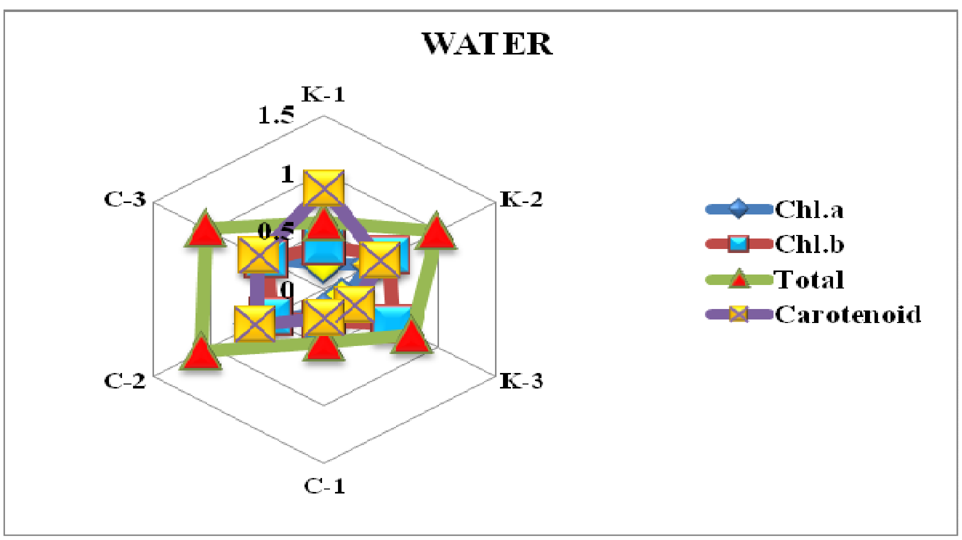
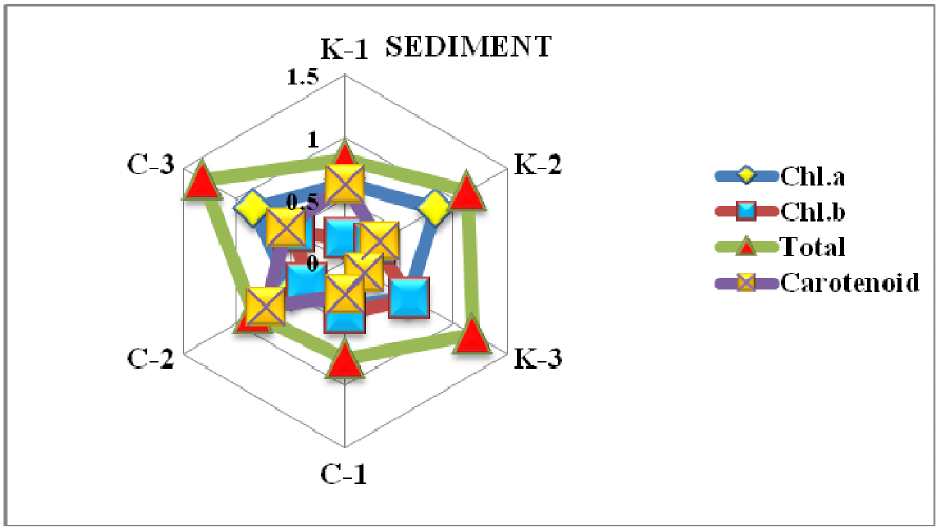


Figure 3 :Pigment content in Sediment Samples (% value)



Correlation studies

The result of Pearson's correlation coefficient studies conducted between the pigment contents in Plants, Water and Sediments in Table 1. Plants showed strong positive correlation (1,0.999,0.998 and 0.997) among themselves, with water and sediment and also very strong correlation (0.998 and 0.997) with sediment between the two media implying common source of plants and sediments. There was a strong negative correlation between water and plants (-0.224,-0.221 and -0.123). This suggests that in plants there is less production of pigments in the presence of certain sediments and water or vice versa in a particular condition.

Table 1: Correlation Analysis of plants, water and sediments

	CAO	CSA	CRM	CEA	KAO	KRM	KEA	KSA	SK1	SK2	SK3	SC1	SC2	SC3	WK1	WK2	WK3	WC1	WC2	WC3
CAO	1																			
CSA	0.994**	1																		
CRM	0.994**	0.998**	1																	
CEA	0.984*	0.994**	0.997**	1																
KAO	0.995**	0.978*	0.981*	0.964*	1															
KRM	0.536	0.518	0.575	0.588	0.545	1														
KEA	0.993**	0.975*	0.979*	0.962*	1.000***	0.562	1													
KSA	1.000***	0.994**	0.995**	0.985*	0.995**	0.536	0.993**	1												
SK1	0.793	0.851	0.851	0.886	0.731	0.522	0.726	0.795	1											

SK	0.999	0.98	0.98	0.97	0.999	0.54	0.99	0.99	0.7											
2	**	7*	9*	5*	**	5	8**	8**	63	1										
SK		0.72	0.75	0.71		0.71	0.85	0.78	0.3	0.81										
3	0.787	0	1	6	0.838	3	0	5	57	5	1									
SC		0.68	0.73	0.71		0.86	0.80	0.74	0.4	0.76	0.97									
1	0.745	9	2	0	0.786	1	1	3	25	9	0*	1								
SC		0.61	0.64	0.69		0.79	0.51	0.56	0.8	0.53	0.34	0.51								
2	0.563	3	4	2	0.510	2	4	5	65	9	2	3	1							
SC	0.970	0.95	0.97	0.96	0.970	0.72	0.97	0.97	0.7	0.97	0.85	0.86	0.6							
3	*	8*	5*	9*	*	4	4	0*	87	2*	5	1	77	1						
			-					-												
W	-	0.15	0.12	0.05		0.45	0.27	0.22	0.3	0.25	0.29	0.05	0.6	0.06						
K1	0.224	9	3	8	0.282	8	5	1	16	1	5	6	80	3	1					
W		0.56	0.62	0.61		0.96	0.66	0.61	0.4	0.63	0.86	0.95	0.6	0.78	0.2					
K2	0.612	9	4	8	0.643	6*	0	1	31	1	2	9*	45	0	17	1				
W		0.16	0.23	0.20		0.78	0.33	0.24	0.0	0.28	0.75	0.83	0.2	0.43	0.0	0.87				
K3	0.247	9	0	5	0.314	9	6	4	61	4	8	0	52	7	78	4	1			
W		0.55	0.60	0.60		0.98	0.63	0.58	0.4	0.60	0.82	0.93	0.6	0.76	0.2	0.99	0.8			
C1	0.589	2	9	7	0.615	1*	3	9	50	6	7	9*	84	5	83	8**	62	1		
W		0.84	0.87	0.89		0.89	0.84	0.84	0.8	0.84	0.78	0.86	0.8	0.94	0.2	0.88	0.5	0.88		
C2	0.846	5	9	0	0.837	2	6	7	02	5	3	9	48	5*	49	2	43	6	1	
W		0.74	0.79	0.79		0.95	0.78	0.76	0.6	0.76	0.82	0.92	0.7	0.89	0.2	0.95	0.6	0.96	0.98	
C3	0.763	5	0	5	0.769	5*	1	3	72	9	9	6*	98	7	64	8*	98	1*	0*	1

4. CONCLUSION

Results from the above analysis clearly indicate that extraction of photosynthetic pigments depend on chemical nature of bio-molecules (cholorophyll-a, chlorophyll-b and carotenoids). Temporal and seasonal changes and local geological conditions are the reasons for variations in pigment concentrations in plants, water and sediment samples. Sediment pigments proved to be good indicators of lake-ecosystem response to climate change and long-term variability in the photo trophic community, which is needed for predicting possible effects of future climate change. It was also recognized that the quality of the pigment record is highly dependent on the preservation regime in the sediment and water. Therefore further study in this context is recommended.

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