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Assessment of Epilithic Microalgae along Longitudinal Gradient in Freshwater Bodies of South India

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Abstract: Epilithic microalgae are associated with hard substrates of freshwater environments and they play a vital role in primary production, nutrient cycling and contributing habitat for other organisms in aquatic environments. Thus, the present study was focused to assess the diversity and distributional pattern of epilithic microalgae along longitudinal gradient in lentic freshwater bodies, Tamil Nadu, India. Nine freshwater bodies (lakes, ponds and temple tanks) were examined in Madurai district. Microalgae were collected from submerged substrates of bedrock, boulders and pebbles in lentic waters. In total, 14 genera of microalgae were collected. *Fragilaria* was the highest percentage. The high diversity was observed in site 9 revealed by Shannon diversity index. Abundance of microalgae was high in lakes when compared to temple tanks and ponds. Of twelve environmental variables, total dissolved solids, conductivity and salinity were significant factors for the distribution of microalgae. This finding suggests that microalgae in lentic freshwater bodies are not influenced with longitudinal gradient whereas chemical parameters of water such as total dissolved solids, dissolved oxygen, electrical conductivity and salinity are the determining factors.

Keywords: Diversity, Distribution, Environmental variables, Longitude, Freshwater

Microalgae are unicellular aquatic organisms and they exist in variety of environments ranging from ponds, lakes, brackish water and oceans. Microalgae perform photosynthesis in aquatic environment and participate in the role of energy transmission in the ecosystem (Thore et al., 2023). In addition, microalgae used for pollution indicators, nutrition dynamics, sewage treatment, food industry, and synthesis for novel compounds. About eight lakhs species of microalgae are distributed in the world, of which 50,000 species described (Ampofo and Abbey 2022). Several types of microalgae are commonly found in hard substrates (bed rocks, boulders, pebbles) of freshwater environments known as epilithic, including Bacillariophyta, Chlorophyta, Cyanophyta and Chrysophyta) (Arulraj et al., 2022). Epilithic microalgae play an important role in primary production, nutrient cycling and habitat for other organisms in aquatic environments.

Studies showed the epilithic microalgae research that diversity of microalgae from building surfaces during monsoonal period (Samad and Adhikary 2008), biodiversity of microalgae in Western and Eastern Ghats rivers, India (Suresh et al. 2012), microalgae diversity in unexplored freshwater bodies associated with industry region (Severes et al. 2018), periphytic microalgae colonization and litter decomposition in an intermittent stream (Arulraj et al. 2019), seasonal variation of microalgae and cyanobacteria in Komuki dam, Tamil Nadu (Selvaraj et al., 2021) and periphytic microalgae colonization in mosquito breeding stream puddles of Western Ghats (Arulraj et al., 2022). Due to increasing anthropogenic impacts in freshwater bodies like waste disposal, urbanization, habitat destruction and sewage integration, necessity to study the ecology of freshwater environments, in particular urban regions. Therefore, the present study focused to study the diversity and distribution of epilithic microalgae along a longitudinal gradient in freshwater bodies of Tamil Nadu, South India.

MATERIAL AND METHODS

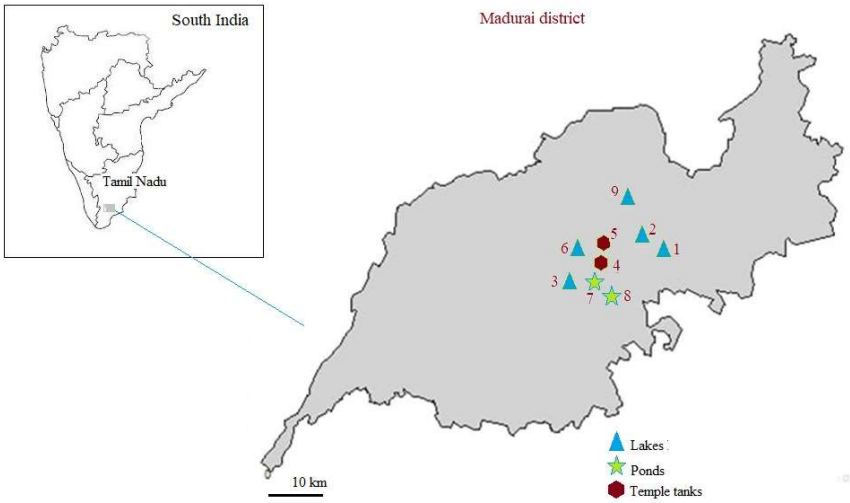
Sampling was collected in three seasons September 2023 (South-west monsoon), November 2023 (North-east monsoon) and pre-summer (February 2024) in the submerged substrates of boulders, gravels and pebbles from nine freshwater bodies in triplicates from Madurai district of Tamil Nadu, India (5 lakes, 2 ponds and 2 temple tanks, Fig. 1) Measurements of water temperature, pH, electrical conductivity, total dissolved solids and salinity were done with portable digital tester (PCS Testr 35, Eutech Instruments, India). Dissolved oxygen was measured by Winkler method in the laboratory. Water circumference and average water depth were measured with the help of meter tape. Water color is also recorded by visual observation. The canopy cover is measured with Densiometer. The submerged substrates were taken in a plastic tray and they were rinsed with filtered pond water. The surface biofilms were scrapped from the substrates by using a hard tooth brush. The scrapped biofilm was immediately stored in 25 ml

plastic vial. Then, it brought to the laboratory and preserved at−4°C until analysis. In the laboratory, 1ml aliquot of algal sample taken each site was counted by zig zag slide observation method and identified with standard identification manual up to genus level.

Data analyses: All data were sorted out in MS-Excel. All statistical analyses including diversity indices and principal component analysis were calculated by using statistical software Biodiversity Pro and PAST 4.0. Diversity indices for microalgae distributed different sites were calculated with Biodiversity Pro 2.0 software.

RESULTS AND DISCUSSION

The physical and chemical parameters differed with sampling sites (Table 1). The high dissolved oxygen, pH, conductivity and salinity were observed in site 1 and low total dissolved solids were at site 1 (lake), while the low dissolved oxygen, pH, conductivity and salinity were at site 7 (pond). A



Vandiyur-L1, Kannenthal-L2, Seventhikulam-L3, Pannaikudi-L4 and Madakulam-L5; two ponds: Piramalai nagar A-P6 and B-P7; two temple tanks: Saravanapoikai-T8 and Tiruparankundram-T9

Fig. 1. Sampling sites of Madurai district, Tamil Nadu province, India

Table 1. Physico-chemical analysis (mean) of different water bodies of sampling sites for three seasons

Site	L1	L2	L3	L4	L5	P6	P7	T8	T9
Latitude (N)	9.933	9.963	9.872	8.319	9.885	9.522	9.522	9.877	9.525
Longitude (E)	78.15	78.14	78.07	77.57	78.07	78.04	78.04	78.74	78.04
Water temperature (°C)	32	27.7	33.4	32	32.5	35.6	34.2	28.7	36.3
Atmosphere temperature (°C)	35.1	31.3	31.4	38.0	33.4	40.0	32.8	32.6	39.3
Salinity (ppm)	850	742	560	97	502	115	98.5	401	621
Total dissolved solids (ppt)	128	110	850	625	743	177	146	599	926
Conductivity (µmhos)	1757	1559	1180	216	1054	248	208	830	1342
pH	7.6	8.2	8.2	6.4	6.8	7.2	6.2	7.5	7.4
Dissolved oxygen (mgL^{-1})	12	8	8	8	8	4	4	8	8
Water circumference (ft)	2077	800	150	800	3150	100	80	1020	120
Water depth (ft)	15	7	5	40	45	52	20	15	30
Number of riparian species	3	4	3	3	4	3	3	2	2
Riparian cover	Open	Open	Open	Open	Open	Open	Open	Open	Open
Water colour	Light green	Light green	Dark green	Light green	Green	Light green	Light green	Green	Light green

Five lakes: Vandiyur-L1, Kannenthal-L2, Seventhikulam-L3, Pannaikudi-L4 and Madakulam-L5; two ponds: Piramalai nagar A-P6 and B-P7; two temple tanks: Saravanapoikai-T8 and Tiruparankundram-T9

total of 2,271 individuals of microalgae were collected under 14 genera in 11 orders and 6 phyla for three seasons of sampling sites (Table 2). The sites 1 and 9 recorded higher number of taxa whereas other sites 11 to 12 species. Microalgae are influenced with several environmental factors and anthropogenic impact. They have the capability of producing and storing desired products as cell metabolites, and adapting themselves when there is a change in the environmental conditions (pH, temperature, light, carbon dioxide, salinity, and nutrients (Gatamaneni et al., 2018). Of 14 microalgal genera collected, *Fragilaria* was occupied the highest percentage (25%) followed by *Chlorococcum* and *Scenedesmus*. The higher number of *Fragilaria* was at site 1 followed by sites 4 and 6. The site 1 had the high numbers of *Chlorococcum* whereas *Scenedesmus* was high at site 4 (Table 2). The freshwater araphid diatom of *Fragilaria* is a dominant species of the cosmopolitan planktonic diatom, widely distributed in oligotrophic and mesotrophic water reservoirs across the northern hemisphere (Galachyants et al., 2019) and they distributed in all seasons (Deb et al., 2024). *Scenedesmus* and *Chlorococcum* are a dominant genus of algae commonly found in wastewater ponds and it adaptability allows for direct cultivation in wastewater environments (Lyon et al., 2015, Hashmi et al., 2024).

The site 1 and 9 had the higher number of taxa (14) whereas other sites hold 11 to 12 numbers of taxa (Table 3). Shannon and Margalef indices showed that the high diversity was in site 9. Simpson diversity index revealed that the high diversity value was occurred at site 8 and low diversity at site 2. Evenness index was high at site 8 and low at site 2. Microalgae sampled in three freshwater bodies of lakes, ponds and temple tanks showed that 14 taxa found in lakes and temple tanks and 12 taxa in ponds were observed. Abundance of microalgae constituted the higher percentage (63.5%) in lakes, followed by 23.6% in temple tanks and 12.7% in ponds. When compared to generic-wise distribution among sampling sites, temple tanks and lakes had the higher percentage of taxa rather than ponds (Fig. 2) and generic composition had high similarity revealed by ternary plot analysis (Fig. 3).

Table 2. Distribution of epilithic microalgae collected from sampling sites during three seasons

Order	Class	Family	Genus	Sampling site								
				S1	S2	S3	S4	S5	S6	S7	S8	S9
Desmidiales	Zygnematophyceae	Closteriaceae	*Closterium*	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	-	-	ǂ
Zygnematales	Zygnematophyceae	Peniaceae	*Penium*	ǂ	-	-	-	-	-	-	-	ǂ
Zygnematales	Conjugatophyceae	Mesotaeniaceae	*Roya*	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ
Chlamydomonadales	Chlorophyceae	Chlamydomonadaceae	*Chlamydomonas*	ǂ	-	-	-	-	-	-	-	ǂ
Chlamydomonadales	Chlorophyceae	Chlorococcaceae	*Chlorococcum*	Φ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ
Sphaeropleales	Chlorophyceae	Scenedesmaceae	*Scenedesmus*	ǂ	ǂ	ǂ	Φ	ǂ	ǂ	ǂ	ǂ	ǂ
Ulotrichales	Ulvophyceae	Ulotrichaceae	*Ulothrix*	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ
Oscillatoriales	Cyanophyceae	Microcoleaceae	*Platensis*	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ
Chroococcidiopsidales	Cyanophyceae	Aliterellaceae	*Gloeocapasa*	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ
Euglenida	Euglenoidea	Euglenaceae	*Euglena*	ǂ	Φ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ
Fragilariales	Bacillariophyceae	Fragilariaceae	*Fragilaria*	Φ	Φ	Φ	Φ	Φ	Φ	ǂ	ǂ	Φ
Cymbellales	Bacillariophyceae	Cymbellaceae	*Cymbella*	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ
Naviculales	Bacillariophyceae	Amphipleuraceae	*Frustula*	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ
Naviculales	Bacillariophyceae	Naviculaceae	*Navicula*	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ	ǂ

-:No individuals; ǂ: 1 to 100 individuals; Φ: >100 individuals

Table 3. Diversity analyses for epilithic microalgae collected from sampling sites for three seasons

	S1	S2	S3	S4	S5	S6	S7	S8	S9
No. of taxa	14	12	12	12	12	12	11	11	14
Simpson index	0.8752	0.8458	0.8858	0.8704	0.8637	0.8587	0.8883	0.8926	0.8824
Shannon index	2.371	2.103	2.303	2.26	2.205	2.213	2.269	2.305	2.403
Evennes index	0.7649	0.6827	0.8339	0.7983	0.7562	0.7622	0.8795	0.9115	0.7897
Margalef index	2.73	2.483	2.503	2.359	2.581	2.457	2.473	2.473	2.848

The eigen value and percentage of variance were1.215 and 71.66 for PC1 and 35061 and 20.65 for PC2. The scores of principal component Analysis are given table 4. Result of the PCA indicates that total dissolved solids, conductivity and salinity were significantly correlated with microalgae distribution rather than other environmental variables (Fig. 4). Among sampling sites, the lake sites of L1 and L2 were important sites for the distribution of microalgae (Fig. 4). The different longitudinal and lateral gradients found in the aquatic system underpins the composition of biological communities, given that species tends to occur predominantly in the patches that most favor their development. Total dissolved solids are an important factor for determining the abundance and growth of phytoplanktons (Li et al., 2013). Environmental variables of pH, conductivity and nutrient concentrations are related to

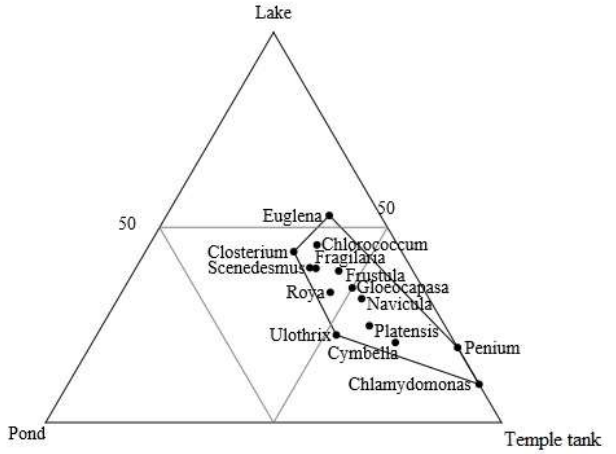


Fig. 3. Distribution of taxa among sampling sites in three freshwater bodies

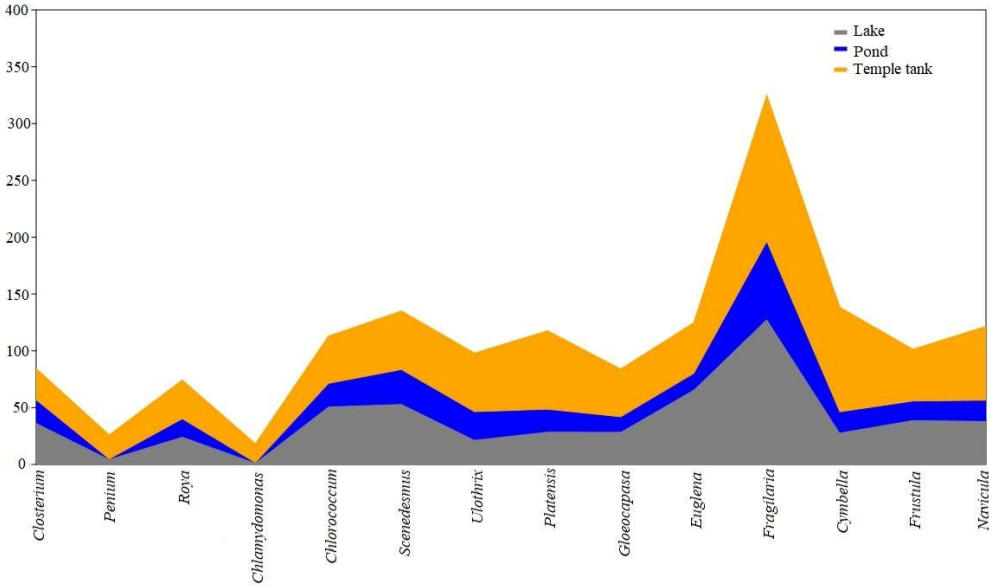


Fig. 2. Generic composition of microalgae (no. of individuals) collected from three freshwater bodies

Table 4. Loading score and loading values of Principal component analysis

	PC 1	PC 2	Site	PC 1	PC 2
LAT -Latitude	0.00017	0.00044	L1	1391	480.76
LON -Longitude	0.000042	0.00012	L2	115.45	690.9
WAT-Water temperature	-0.0009	-0.0007	L3	-647.06	512.1
ATT-Atmosphere temperature	-0.0008	-0.0012	L4	-356.19	-685.24
SAL-Salinity	0.14168	0.40293	L5	2153.1	-590.18
TDS-Total dissolved solids	0.01613	0.04509	P6	-1039.3	-474.21
CON-Conductivity	0.28955	0.84822	P7	-1074.7	-511.46
pH	0.000043	0.001	T8	54.874	-135.36
DO-Dissolved oxygen	0.00144	0.00209	T9	-597.21	712.69
WC-Water circumference	0.94363	-0.3284			
WD-Water depth	0.00087	-0.0196	Eigen	1.215	71.66
NR-No. of Riparian species	0.00029	-0.0002	Covariance	35061	20.65
Microalgae	0.07342	0.08872			

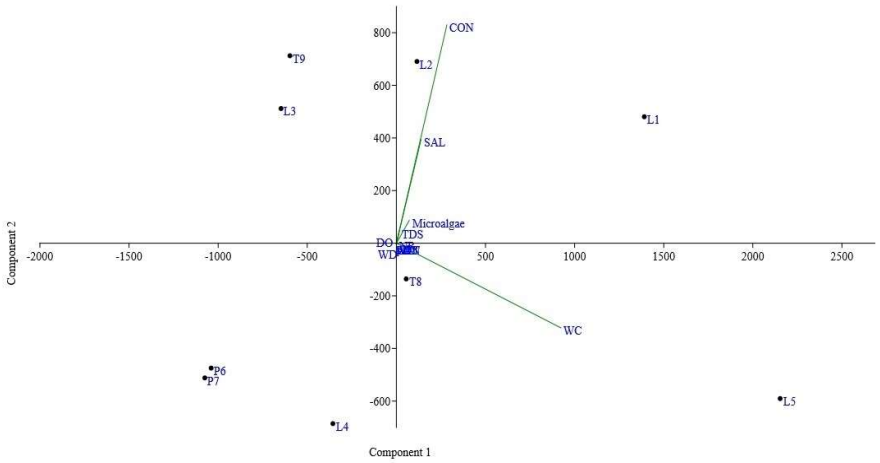


Fig. 4. Principal component analysis (PCA) shows the relationship between environmental variables and microalgae distribution

the richness and abundance of phytoplanktons (Moura et al., 2021).

CONCLUSION

The present study was carried out the distribution of microalgae with longitudinal gradient in lentic freshwater bodies. In total, 14 genera of microalgae were collected. Among genera, *Fragilaria* had the highest percentage followed by *Chlorococcum* and *Scenedesmus*. The highest diversity value was observed in both lakes and temple tanks sites and ponds had low diversity. Abundance of microalgae was high in lakes when compared to temple tanks and ponds. Total dissolved solids, conductivity and salinity were significant factors for the distribution of microalgae rather than other environmental variables. Overall, microalgae in lentic freshwater bodies are determining by chemical characters of water (total dissolved solids, dissolved oxygen, electrical conductivity and salinity) and not be influenced with longitude.

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