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Algal colonization on an insectivorous plant, *Utricularia aurea* Lour. in a freshwater marsh of southern Assam, India

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Abstract

Algal epiphytons on *Utricularia aurea*, an insectivorous macrophyte growing in a freshwater marsh of southern Assam, India was explored. Variation of algal composition and biomass in relation to physico-chemical parameters of the marshy ecosystem were studied. A total of 80 algal epiphytons belonging to 36 genera and 4 different classes were observed to be colonizingon the macrophyte. The species, *Spirogyra crassa*, *S. gracilis*, *S. rivularis* and *Scenedesmus dimorphus* were observed to be the dominant colonizers on *U. aurea*. Inter-relationship between the environmental factors and epiphyton diversity was evaluated by Principal Component Analysis (PCA) and Canonical Correspondence Analysis (CCA). Water temperature, pH and nutrients like nitrate-nitrogen, soluble reactive phosphorus, dissolved-silicon were found to be the regulating factors for epiphytic algal diversity on the macrophyte in the freshwater marsh.

Key words: Epiphytic algae, Diversity, *Utricularia aurea*, Freshwater marsh

Introduction

Freshwater marshes harbor wide variety of life forms. Most important among them are macrophytes that grow profusely in such ecosystems and are considered to play a vital role in providing food and shelter for aquatic living beings (Engelhardt and Richie, 2001). Subtle yet complicated interactions are operative among various organisms in within such aquatic ecosystems in respect of prey relationships, competition for nutrient, living space and light (Carpenter and Lodge, 1986; Gopal and Goel, 1993). As primary producers, macrophytes occupy the base of herbivorous and detritivorous food chains. The water movement and sediment dynamics in such aquatic ecosystem are also profoundly influenced by macrophytes. In addition to offering variety of ecosystem services furnishing food, medicine, biomass and building materials (Costanza *et al*., 1997; Engelhardt and Ritchie, 2001; Egertson *et al.*, 2004; Bornette and Puijalon, 2011, Meena and Rout, 2016), their relevance in wetland restoration, wastewater treatment and invasive species management are well recognized (Lavoie, 2010; Casanova, 2011). Different parts of macrophytes such as stem, root and leave provides shelter for numerous fishes, invertebrates, amphibians and reptiles (Timms and Moss, 1984; Dvořák, 1996). Epiphytic alga which grows and flourishes on such macrophytic host comprise an important autotrophic community of an aquatic ecosystem (Wetzel, 1990; Toporowska *et al*., 2008; Hassan *et al*., 2012; Hafner and Jasprica, 2013; Salman *et al*., 2014; Tunca *et al*., 2014; Al-Hassany and Hassan, 2015; Azam *et al*., 2016; Das and Baruah, 2016). The epiphyton serving as primary producers play a significant role in carbon fixation (Cattaneo and Kalff, 1979).The morphology and the type of macrophytic species governs the distribution and abundance of epiphytic algae species (Messyasz *et al*., 2009; Pomazkina *et al.,* 2012). Inhibition of algal colonisation by allelopathic chemicals released by macrophytes are on record (van Donkand van de Bund, 2002). Though southern Assam (India) is endowed with rather wide variety of wetlands, studies of epiphytons on macrophytes are very few (Rout and Meena, 2012; Meena and Rout, 2018).The macrophyte species *Utricularia aurea* Lour**.** is an insectivorous perennial plant found to grow in abundance in this region's freshwater bodies including the marshes. As a part of extensive ongoing program on documenting epiphytons from different macrophytic species, we present herein, an account of the seasonal variability of epiphytic algae in relation to water quality parameters on a macrophyte, *Utricularia aurea* Lour inhabiting a freshwater marsh in southern Assam.

Materials and Methods

The study was carried out in a freshwater marsh located at Irangmara area of Cachar district, southern Assam, North-East India. This area lies about 22.3 km away from Silchar town, the district headquarter at latitude 24˚41'47.8"N and longitude 92˚43'56.5"E (Fig. 1) with an area of 2624.27m^2.

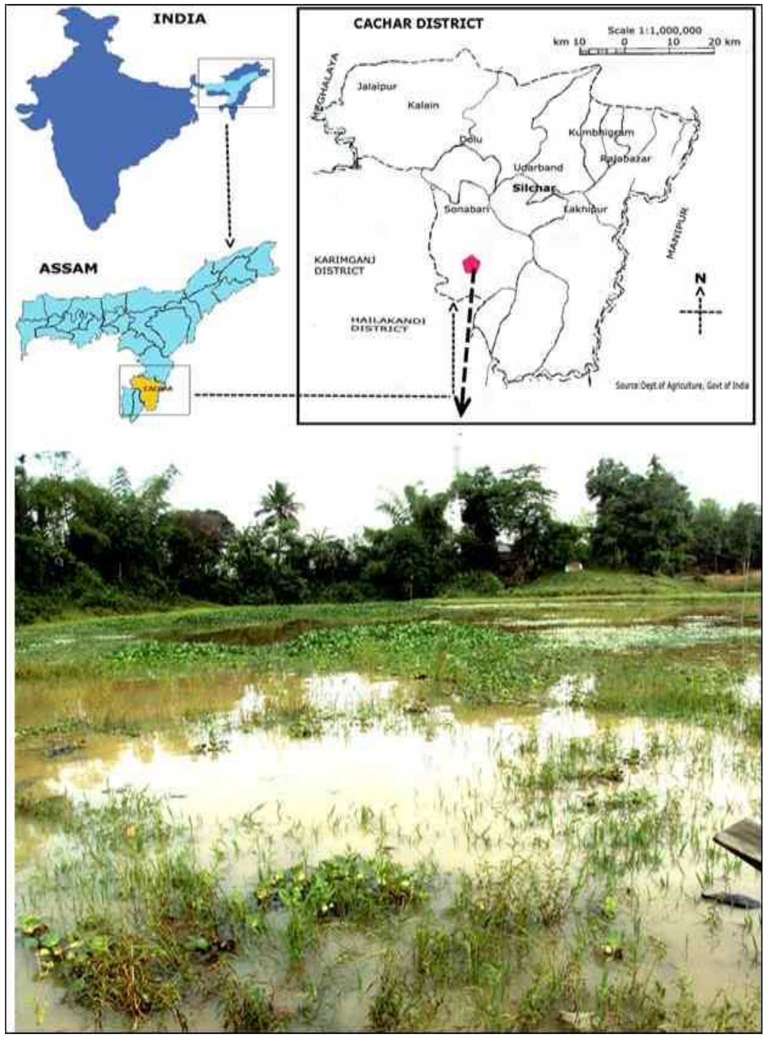

Fig. 1 Map showing the study site.

Analysis of physico-chemical parameters of water

Water samples were collected in PVC bottles from the subsurface and their analyses were performed in triplicate following standard procedures (Wetzel and Likens, 1979; APHA, 2005). Water temperature (WT) was measured on spot using a thermometer. The pH and electrical Conductivity (EC) were measured by using Systronics pH system 362 and by Systronics Conductivity TDS Meter 308 separately. Dissolved Oxygen (DO) was determined by Winkler's method, Free CO_2 (F-CO_2) and Total alkalinity (Talk)were determined by titrimtric method (APHA, 2005), Soluble Reactive Phosphorus (SRP) was analysed by ascorbic acid method (APHA, 2005), dissolved silica by molybdate blue method (Wetzel and Likens, 1979) and nitrate by Brucine method (Suess, 1982).

Collection and identification of epiphytic algae colonized on the macrophyte

The insectivorous plant, *Utricularia aurea* Lour. growing abundantly in a freshwater marsh has been identified using standard literature (Cook, 1996). Monthly sampling of *U. aurea* and water were carried out for 12 months from May 2012 to April 2013. The macrophyte was carefully collected from five different spots and brought to laboratory for further analysis in clean poly bags. For epiphytic algal study, 1g of the freshly collected macrophyte was weighed and preserved in 4.5% formalin (APHA, 2005). Algal enumeration was carried out by Lackey's drop method (Lackey, 1938). For biomass, chlorophyll *a* was estimated by cold extraction method (freezing and thawing) in 90% acetone by grinding the scraped off algae from the weighed sample. The pigment was estimated spectrophotometrically (Strickland and Parsons, 1968). For morphological identification and enumeration of epiphytic algae colonized on *U. aurea*, 0.1 ml of the sample was taken on a clean glass slide held by coverslip. Observations were taken five times. Microphotographs of the algal samples were taken in Leica application suit (LM1000 LED). Morphological details such as size (length and breadth) and shape of the cells, colour, zygospores, branch characteristics and heterocysts etc. were taken into account. Identification of algae were performed following standard keys (Prescott, 1952; Desikachary, 1959; Sarode and Kamat, 1984; Anand, 1998).

Statistical analysis

Shannon-Wiener Diversity Index (H), Evenness index ($J^{/}$) and Berger-Parker Dominance index (d) and Canonical Correspondence Analysis were calculated using Paleontological STatistics (PAST) V-2.13. Principal Component Analysis was performed with Statistical Package for the Social Science (SPSS) V-15.

Results and Discussion

Physico-chemical properties of water in the freshwater marsh

The physico-chemical properties of water and algal biomass (chlorophyll *a*) are shown in Table 1. The water temperature ranged from 17 to 33°C with an annual mean of 27.53°C. The parameter WT is an important factor for limnological studies (Sheela *et al*., 2011). Expectedly, WT of the freshwater marsh was the highest in summer and lowest in winter, which was similar to our previous observations (Rout and Borah, 2009; Rout and Meena, 2012; Devi *et al*., 2013; Meena and Rout, 2018). Temperature is a crucial factor for regulating different physiological processes like respiration, metabolic activities and behavior of the biota. pH was found to be slightly basic in all the months except March and April 2013. An acidic pH of the marsh might be due to the decomposition of organic matters including macrophytes (Das *et al*., 2012). The EC ranged from 39.33 to 87.30 µS/cm with registering a relatively lower value during the rainy season (May, June and July 2012). This might be due to the dilution effect in contrast to dry season when water level decreases and ions increase due to evaporation. Increase in EC due to evapo-transpiration and decomposition of organic matter was reported by Devi *et al.,* (2013, 2016) in Banskandi anua of Cachar district. The DO was found highest in July 2012, whereas it dropped to lowest in the month of August 2012. Higher DO value might be attributed to the turbulence due to rain augmenting oxygenation of the freshwater marsh. A low DO might be due to rise in temperature and decrease in water level due to less rainfall in some month. The BOD ranged from 0.33 to 5.05 mg/l. Increase in BOD might be due to higher organic load in the marsh. The F-CO_2 ranged from 0.88 to 22.29 mg/l with an annual mean of 4.40 mg /l. Highest F-CO_2 was observed in March 2013 and the lowest was experienced in June 2012. This is believed to be due to the interaction among ecosystem respiration rate, net and gross ecosystem productivity (Suzuki *et al.,* 2012). Total alkalinity was found highest in February 2013 (32.53 mg/l) and lowest in September 2012 (11.39 mg /l) with an annual mean of 19.93 mg/l. Pandey *et al.,* (2014) reported highest value of total alkalinity assigning to higher organic load. The D-Si ranged from 2.24 to 14.63 mg/l with an annual mean of 7.55 mg/l. Highest D-Si was observed in July 2012 which might be due to the rain affected weathering of silicate materials, mixing of sediments with water, aeration, reactivity of bedrock and stability of minerals (Dariusz, 2005; Pradeep *et al*., 2016). The NO_3-N ranged from 0.06 to 0.18 mg/l with an annual mean of 0.10mg/l. The SRP ranged from 0.11 to 0.25 mg/ l with an annual mean of 0.15 mg/l. The primary nutrient status (NO_3-N and SRP) in the freshwater marsh could be of both organic and inorganic origin that might be associated with soil, vegetation and landuse practices (Reddy *et al*., 1999).The Chl-*a* was found highest in April 2013 (2.98 mg/ l). The macrophyte, *U. aurea* was not observed in March 2013.

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Table 1: Monthly variation of physico-chemical parameters of water and chlorophyll *a* (May 2012 to April 2013).

Months	WT (°C)	pH	EC (µS/ cm)	DO (mg /l)	BOD (mg /l)	F-CO_2 (mg /l)	TAlk (mg /l)	D-Si (mg /l)	NO_3-N (mg /l)	SRP (mg/ l)	Chl-*a* (mg /l)
May-12	29.67±0.58	7.49±0.04	39.33±0.25	4.23±0.29	2.03±0.51	2.35±0.51	25.21±3.73	7.83±1.36	0.081±0.034	0.247±0.184	0.19±0.05
Jun-12	29.67±0.58	7.39±0.03	39.40±0.44	4.40±0.29	1.86±0.29	0.88±0.01	21.96±2.44	9.30±0.48	0.119±0.001	0.193±0.168	1.00±0.01
Jul-12	29.67±0.58	7.43±0.04	43.50±2.84	6.60±1.02	1.52±1.02	1.76±0.01	16.27±1.41	14.63±2.66	0.181±0.001	0.133±0.001	0.79±0.02
Aug-12	33.00±0.01	7.40±0.17	50.77±1.72	2.54±0.01	1.19±0.29	2.64±0.01	18.71±2.82	11.94±1.01	0.137±0.034	0.189±0.042	1.09±0.01
Sep-12	27.67±1.53	7.10±0.19	51.20±1.40	5.25±0.78	5.05±0.78	1.17±0.51	11.39±1.41	11.74±0.12	0.117±0.001	0.152±0.018	0.59±0.02
Oct-12	30.67±0.58	7.25±0.14	44.47±4.56	4.74±0.29	1.19±0.29	2.05±0.51	13.01±1.41	10.43±1.23	0.081±0.034	0.129±0.021	0.46±0.22
Nov-12	19.33±0.58	7.71±0.06	42.23±1.40	4.40±0.29	1.35±0.29	3.23±0.51	18.71±1.41	6.83±0.64	0.064±0.001	0.173±0.001	0.33±0.01
Dec-12	17.00±0.01	7.55±0.15	45.10±2.42	3.89±0.78	1.35±0.29	2.93±0.51	17.08±0.01	4.65±0.31	0.083±0.034	0.119±0.001	0.48±0.20
Jan-13	18.33±1.15	7.39±0.02	45.74±0.47	6.14±0.09	0.33±0.16	3.23±1.02	21.15±2.82	3.02±0.31	0.079±0.034	0.125±0.002	0.75±0.03
Feb-13	30.67±0.58	7.45±0.06	87.30±0.90	5.49±0.19	2.39±0.77	6.45±0.51	32.53±2.82	4.52±0.14	0.084±0.034	0.105±0.023	2.55±0.42
Mar-13	32.67±0.58	5.69±0.07	86.21±1.52	4.23±0.59	2.20±1.06	22.29±2.21	20.33±1.41	2.24±0.06	0.105±0.034	0.134±0.021	0.00±0.00
Apr-13	32.00±0.01	5.58±0.08	69.57±0.24	5.42±0.29	1.35±0.29	3.81±0.51	22.77±3.73	3.44±0.10	0.078±0.034	0.129±0.009	2.89±0.01
Annual mean	27.53±5.82	7.12±0.71	53.74±17.38	4.78±1.09	1.82±1.16	4.40±5.82	19.93±5.61	7.55±4.07	0.10±0.03	0.152±0.42	0.93±0.13

Epiphytic algal communities on U. aurea

The study on epiphytic algal colonization on *U. aurea* from a freshwater marsh at Irangmara has revealed a total of 80 species belonging to four different classes, viz., Chlorophyceae (47), Bacillariophyceae (24), Cyanophyceae (8) and Euglenophyceae (1) (Table 2; Plate 1,2). In a similar study, a total of 259 algal species belonging to 63 genera with five groups had been reported from two ponds at Silcoorie area of Cachar district (Meena and Rout, 2018).The study on distribution of epiphytic algae on *U. aurea* revealed that *Spirogyra crassa*, *S. gracilis* and *S. rivularis* were found throughout the study period. The species *Closterium gracile* was found only in April 2013.The Table 3 show the variation in the number of algal species found in the four classes during the study period. The Cyanophyceae (5) was found to be colonized during the months of December (2012), January, February and April (2013) presumably favoured by low level of water in the marsh. Greater abundance of cyanobacteria in older parts of *U. aurea* and its role in nitrogen fixation has been reported by Wagner and Mshigeni (1996). While Chlorophyceae (28) was observed to be highest during the months of May, June and July 2012, the Bacillariophyceae was most dominant in December 2012. Euglenophyceae with only 1 species viz. *Phacus chloroplastis* was observed during the month of May, June, July 2012, February and April 2013. The lesser number of Cyanophyceae and Euglenophyceae may be due to the periodic changes in the water content in the marsh resembling previous findings of Rout and Meena, (2012). The dominance of Chlorophyceae family on *U. aurea* coincides with our previous observations elsewhere from pond ecosystem (Meena and Rout, 2018). The macrophyte in the month of March was not encountered possibly due to sudden onset of rain causing submergence of the macrophyte. In the present study, Bacillariophyceae was found to be the second most abundant group which resembled the observation in a natural mid-altitude lake in Himachal Pradesh of (Jindal *et al*., 2014). Diatoms and green algae (mostly desmids) were reported to be the most dominant epiphytic algal colonizer on *Azolla*, *Pistia*, *Nymphaea* and *Ipomoea* (Mpawenayo and Cocquyt, 2007).Inorganic nutrients, anthropogenic activities and lack of water flow might be the reason for distribution of epiphytic algae on the chosen macrophyte (George *et al*., 2014). Extensive growth of green algae in summer and the effect of light, nutrient, host plant and depth of sampling sites has been also documented (Karosienė and Kasperovičiene, 2008). When compared with our previous study involving epiphytons on *U. aurea* from a pond ecosystem (Meena and Rout, 2018) a number of algal species were found to be common epiphytes on the selected macrophyte. The species *Oscillatoria sancta*, *Merismopedia glauca*, *Oscillatoria subbrevis*, *Oscillatoria limosa*, *Chlorobotrys regularis*, *Closterium acerosum*, *C. cynthia*, *C. dianae*, *C. gracile*, *C.*

navicula, *C. parvulum*, *Cosmarium blytii*, *C. polygonum*, *Micrasterias foliacea*, *Oedogonium pusillum*, *Onychonema leave*, *Staurastrum alternans*, *Caloneis bacillum*, *Eunotia lunaris*, *Gomphonema parvulum*, *Navicula cryptocephala*, *Pinnularia subcapitata* and *Stauroneis anceps* were found to be common epiphytic colonizers on *U. aurea* in both the habitats (pond and marsh). However, a few species like *Aphanothece* sp., *Ankistrodesmus falcatus*, *A. spiralis*, *Arthrodesmus curvatus*, *A. gibberelus*, *Closteriopsis longissima*, *Closterium kuetzingii*, *C. leibleinii*, *C. turgidum*, *Cosmarium bioculatum*, *C. impressulum*, *Dictyosphaerium pulchellum*, *Euastruman golense*, *E. turneri*, *Gonatozygon aceleatum*, *Mougeotia genuflexa*, *Nephrocytium lemniticum*, *Pediastrum duplex*, *Pleurotaenium ehrenbergii*, *Scenedesmus armatus*, *S. bijuga*, *S. dimorphus*, *S. quadricuada*, *Spirogyra crassa*, *S. gracilis*, *S. rivularis*, *Staurastrum crenulatum*, *S. sub-rotula*, *C. ventricosa*, *Eunotia bilunaris*, *E. vanheurkii*, *Phacus chloroplastis* etc. were found to be specific to the macrophyte of freshwater marsh. No representative of Dinophyceae could be observed on *U. aurea* from the marsh. This clearly shows a host and habitat specific epiphytic algal colonization on *U. aurea*. The Table 4 shows the diversity indices, Shannon-Wiener Diversity Index (H'), Evenness index ($J^{/}$) and Berger-Parker Dominance index (d) of algae. Shannon-Wiener diversity index (H') ranged from 3.241 to 3.800, while Evenness index ($J^{/}$) ranged from 0.820 to 0.893. Berger-Parker Dominance index (d) ranged from 0.051 to 0.083. Shannon-Wiener diversity index (H') was found highest during the month of June 2012 and lowest during September 2012. Evenness index ($J^{/}$) was found highest during the month of November 2012 and lowest during the month of December 2012. Berger-Parker Dominance index (d) was found highest in October 2012 and lowest in December 2012.

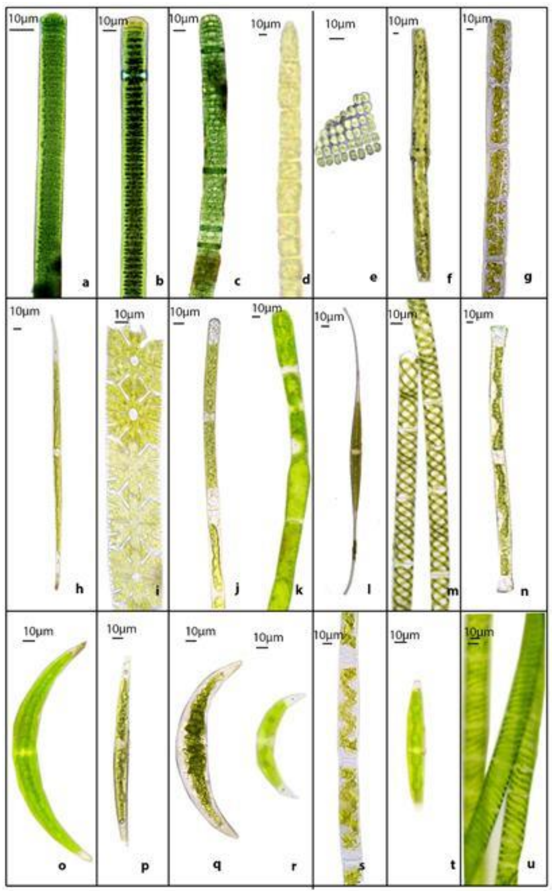

Plate 1: **a**. *Oscillatoria limosa*. **b.** *Oscillatoria subbrevis*. **c.** *Oscillatoria sancta*. **d.** *Anabaena* sp. **e.** *Merismopedia glauca*. **f.** *Pleurotaenium ehrenbergii*. **g.** *Spirogyra* sp. **h.** *Closteriopsis longissima*. **i.** *Micrasterias foliacea*. **j.** *Mougeotia genuflexa*. **k.** *Oedogonium pusillum*. **l.** *Closterium kuetzingii*. **m.** *Spirogyra rivularis*. **n.** *Gonatozygon aculeatum*. **o.** *Closterium parvulum*. **p.** *Closterium acerosum*. **q.** *Closterium turgidum*. **r.** *Closterium cynthia*. **s.** *Spirogyra gracilis*. **t.** *Closterium navicula*. **u.** *Spirogyra crassa.*

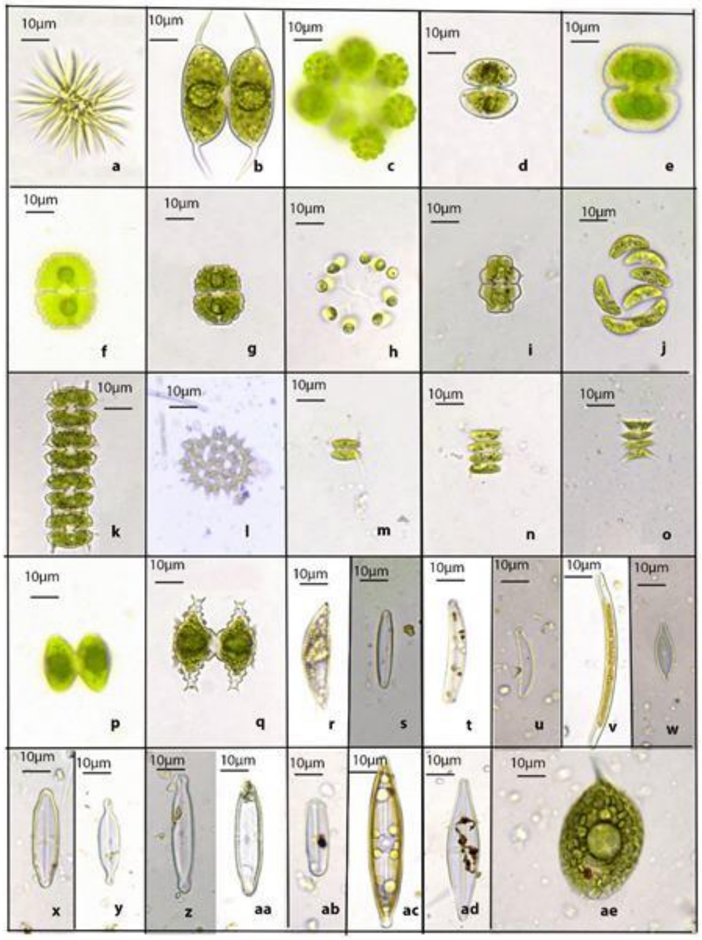

Plate 2: a. *Ankistrodesmus spiralis***. b.** *Arthrodesmus curvatus***. c.** *Coelastrum cambricum***. d.** *Cosmarium bioculatum***. e.** *Cosmarium blytii***. f.** *Cosmarium impressulum***. g.** *Cosmarium polygonum***. h.** *Dictyosphaerium pulchellum***. i.** *Euastrum angolense***. j.** *Nephrocytium limneticum.* **k.** *Onychonema leave***. l.** *Pediastrum duplex***. m.** *Scenedesmus armatus***. n.** *Scenedesmus armatus* **o.** *Scenedesmus dimorphus***. p.** *Staurastrum alternans***. q.** *Staurastrum crenulatum***. r.** *Cymbella ventricosa***. s.** *Caloneis bacillum***. t.** *Eunotia bilunaris***. u.** *Eunotia lunaris***. v.** *Eunotia bilunaris***. w.***Gomphonema parvulum***. x.** *Neidium dubium***. y.** *Pinnularia interrupta***. z.** *Pinnularia subcapitata***. aa.** *Pinnularia microstauron***. ab.** *Sellaphora stroemii***. ac.** *Stauroneis* sp.**ad.** *Stauroneis anceps***. ae.** *Phacus chloroplastis.*

Table 2: Distribution of algae during the study period (May 2012 to April 2013).

	Name of the species	May -12	Jun -12	Jul-12	Aug-12	Sep-12	Oct -12	Nov-12	Dec-12	Jan -13	Feb -13	Mar -13	Apr -13
	CYANOPHYCEAE												
1	*Anabaena* sp.	+	+	+	+	+	-	-	-	-	-	-	-
2	*Aphanocapsa* sp.	-	-	-	-	-	+	+	+	+	+	-	+
3	*Aphanothece* sp.	+	+	+	-	-	-	-	+	+	+	-	+
4	*Chroococcus* sp.	+	-	-	-	-	+	+	+	+	-	-	-
5	*Oscillatoria limosa* Ag. ex Gomont	-	-	-	-	-	-	+	+	+	+	-	+
6	*Oscillatoria sancta* (Kuetz.) Gomont	-	-	-	-	-	+	+	+	+	+	-	+
7	*Oscillatoria subbrevis* Schmidle	+	+	+	+	+	-	-	-	-	+	-	+

8	*Merismopedia glauca* (Ehrenberg) Näg.	-	-	+	+	+	-	-	-	-	-	-	-
	CHLOROPHYCEAE												
9	*Actinastrum* sp.	+	+	+	+	-	-	-	-	-	-	-	-
10	*Ankistrodesmus falcatus* (Corda) Ralfs.	-	-	+	+	+	+	+	+	+	+	-	-
11	*Ankistrodesmus spiralis* (Turner) Lemm	+	+	+	-	-	-	-	-	-	-	-	-
12	*Arthrodesmus curvatus* Turner	-	-	-	-	-	-	-	+	+	+	-	+
13	*Arthrodesmus gibberulus* Joshua	+	+	+	+	+	+	+	-	-	-	-	-
14	*Chlorobotrys regularis* (W. West) Bohlin	+	+	+	+	+	+	+	+	-	-	-	-
15	*Closteriopsis longissima* (Lemm.) Lemm.	-	-	-	-	-	-	-	+	+	+	-	+
16	*Closterium acerosum* Ehrenberg Ralfs.	+	+	+	+	+	+	+	-	-	-	-	-
17	*Closterium Cynthia* De Notaris	-	-	-	-	-	-	-	+	+	+	-	+
18	*Closterium dianae* Ehrenberg.	+	+	+	-	-	-	-	-	-	-	-	-
19	*Closterium gracile* Bréb	-	-	-	-	-	-	-	-	-	-	-	+
20	*Closterium kuetzingii* Bréb	+	+	+	+	-	-	-	-	-	-	-	-
21	*Closterium leibleinii* Kützing ex Ralfs.	+	+	+	-	-	+	+	+	-	-	-	-
22	*Closterium navicula* (Bréb) Lutkem	-	-	-	+	+	+	+	-	-	-	-	-
23	*Closterium parvulum* Nägeli	-	-	-	-	+	+	+	-	-	+	-	+
24	*Closterium turgidum* Ehrenberg ex Ralfs	+	+	+	+	-	-	-	+	+	+	-	-
25	*Cosmarium bioculatum* Brebisson ex Ralfs.	+	-	-	-	-	+	+	-	-	-	-	-
26	*Cosmarium blytii* Wille	-	+	+	+	+	-	-	-	-	-	-	+
27	*Cosmarium impressulum* Elfv.	-	-	-	-	-	-	-	-	+	+	-	+
28	*Cosmarium polygonum* (Naeg.) Arch.	-	-	-	-	-	-	-	-	-	+	-	+
29	*Cosmarium*sp.2	+	+	+	+	-	-	-	-	+	+	-	+
30	*Cosmarium*sp.3	-	+	+	+	+	+	+	+	-	-	-	-
31	*Dictyosphaerium pulchellum* Wood	+	+	+	+	+	-	-	-	-	-	-	+
32	*Euastrum angolense* (West & G.S.West) Willi Krieger	-	+	+	-	-	-	-	+	+	+	-	+
33	*Euastrum* sp.1	-	+	-	-	-	+	+	+	-	-	-	-
34	*Euastrum turneri* West	-	-	-	-	-	-	-	-	-	+	-	+
35	*Gonatozygon aceleatum* Hastings	+	+	+	+	-	-	-	+	+	+	-	-
36	*Micrasterias foliacea* Bail	+	-	-	-	-	-	-	+	+	+	-	+
37	*Mougeotia genuflexa* (Dillw.) C. A. Agardh.	+	+	-	-	+	+	+	+	-	-	-	-
38	*Nephrocytium lemniticum* (G.M.Smith) G.M.Smith	-	-	-	-	-	-	-	-	+	+	-	+
39	*Oedogonium pusillum* Kirchen ex Hirn.	-	-	+	+	+	+	+	+	-	-	-	-
40	*Onychonema laeve* var. *latum* West and West	+	+	+	-	-	-	-	-	-	+	-	+
41	*Pediastrum duplex* Meyen	+	-	-	-	-	-	+	+	+	+	-	-
42	*Pleurotaenium ehrenbergii* (Breb.)	-	-	+	+	+	+	-	-	-	-	-	-
43	*Scenedesmus armatus*(Chodat) G. M. Smith	+	-	-	-	-	-	-	-	-	+	-	+
44	*Scenedesmus armatus* (Chodat) G. M. Smith var. *bicaudatus* (Guglielmetti) Chodat	+	+	-	-	-	-	+	+	+	+	-	-

45	*Scenedesmus bijuga*(Turp.) Lagerheim	-	-	-	+	+	+	+	+	+	+	-	-
46	*Scenedesmus dimorphus* (Turpin) Kuetzing	+	+	+	+	+	+	+	+	+	+	-	-
47	*Scenedesmus quadricuada* (Turpin) Brebissonvar.*longispina*(Chodat) G. M. Smith	+	+	+	+	+	+	+	+	-	-	-	-
48	*Spirogyra crassa* Kuetzing	+	+	+	+	+	+	+	+	+	+	-	+
49	*Spirogyra gracilis* (Hassall) Keutzing	+	+	+	+	+	+	+	+	+	+	-	+
50	*Spirogyra rivularis* (Haesal) Rabenhorst	+	+	+	+	+	+	+	+	+	+	-	+
51	*Staurastrum alternans* Brebisson	-	-	-	-	-	+	+	+	-	-	-	-
52	*Staurastrum crenulatum* (Nägeli) Delponte	+	+	+	-	-	+	+	+	-	-	-	-
53	*Staurastrum recurvatum* Turner	+	+	+	+	-	-	-	-	-	+	-	+
54	*Staurastrum* sp.2	+	+	+	-	-	-	-	-	-	-	-	-
55	*Zygnema* sp.	+	+	+	-	-	-	-	+	+	+	-	-
	BACILLARIOPHYCEAE												
56	*Amphora costata* W. Smith	-	-	+	+	-	-	-	-	+	+	-	+
57	*Caloneis bacillum* (Grunow) Cleve	+	+	+	+	+	-	-	-	-	-	-	-
58	*Cymbella ventricosa* Kuetz.	-	+	+	+	+	+	+	+	-	-	-	-
59	*Eunotia bilunaris* (Ehrenberg) Mills	+	+	-	-	-	-	-	+	+	+	-	+
60	*Eunotia* sp.	+	+	-	-	-	-	+	+	+	+	-	+
61	*Eunotia lunaris* (Ehr.) Grun. v. *subarcuata* (Naeg.) Grun	+	+	+	+	+	+	+	+	-	-	-	-
62	*Eunotia* sp.1	-	+	+	+	+	+	+	+	-	-	-	-
63	*Eunotia vanheurkii var. intermedia*	-	+	+	+	+	+	+	+	-	-	-	+
64	*Gomphonema affine Kützing*	+	+	-	-	-	-	-	+	+	+	-	+
65	*Gomphonema parvulum* (Kützing) Kützing *sensu stricto*	+	+	-	-	-	-	-	+	+	+	-	+
66	*Navicula cryptocephala* Kuetz.	-	+	+	+	+	+	+	+	-	-	-	-
67	*Navicula* sp.1	+	+	-	+	+	+	+	+	-	-	-	-
68	*Navicula* sp.2	+	+	-	-	-	-	-	+	+	+	-	+
69	*Navicula* sp.3	-	+	-	-	-	-	+	+	+	+	-	-
70	*Navicula* sp.4	+	+	-	-	-	-	-	+	+	+	-	-
71	*Navicula* sp.5	-	+	-	-	-	-	-	+	-	-	-	-
72	*Neidium dubium* (Ehr.) Cleve	-	-	-	-	-	+	+	+	+	-	-	-
73	*Neidium* sp.2	-	+	+	+	+	+	+	+	-	-	-	+
74	*Pinnularia interrupta* W. Smith f. *minor* Boye Pet.	-	-	-	-	-	-	-	+	+	+	-	+
75	*Pinnularia microstauron* (Ehrenberg) Cleve	+	+	-	-	-	-	-	+	+	+	-	-
76	*Pinnularia subcapitata* Greg. v. *lapponica* A. Cl.	-	-	-	-	-	-	-	-	-	+	-	+
77	*Sellaphora stroemii* (Hustedt.)	+	+	-	-	-	+	+	+	-	-	-	-
78	*Stauroneis anceps* Ehr.	+	+	-	-	-	-	-	+	-	-	-	-
79	*Stauroneis* sp.	-	-	-	-	-	-	-	-	-	+	-	+
	EUGLENOPHYCEAE												
80	*Phacus chloroplastis* Prescott	+	+	+	-	-	-	-	-	-	+	-	+

Table 3: Variation of algal class found in *U. aurea* during the study period (May 2012 to April 2013).

Algal class	May-12	Jun-12	Jul-12	Aug-12	Sep-12	Oct-12	Nov-12	Dec-12	Jan-13	Feb-13	Mar-13	Apr-13
Cyanophyceae	4	3	4	3	3	3	4	5	5	5	0	5
Chlorophyceae	28	28	28	22	18	21	22	25	19	25	0	20
Bacillariophyceae	12	19	8	9	8	9	11	20	11	12	0	11
Euglenophyceae	1	1	1	0	0	0	0	0	0	1	0	1

Table 4: Diversity indices of the epiphytic algae found in *U. aurea* during the study period (May 2012 to April 2013).

	May-12	Jun-12	Jul-12	Aug-12	Sep-12	Oct-12	Nov-12	Dec-12	Jan-13	Feb-13	Apr-13
Shannon_H	3.6690	3.8000	3.5770	3.3840	3.2410	3.3480	3.4970	3.7130	3.4230	3.6300	3.4880
Evenness (J$^{/}$)	0.8716	0.8764	0.8513	0.8675	0.8810	0.8624	0.8925	0.8195	0.8764	0.8772	0.8841
Berger-Parker	0.0518	0.0557	0.0791	0.0695	0.0759	0.0872	0.0603	0.0507	0.0683	0.0627	0.0596

Interrelationship between physico-chemical properties and epiphytic algal communities in the marsh

Principal Component Analysis (PCA) was performed to evaluate the important factors responsible for the epiphytic algal colonization on *U*. *aurea* (Fig.2)*.* The analysis revealed that first four components with eigenvalue >1 were the most important components in regulating the epiphytic algal colonization in the ecosystem. Eigenvalue is index of variance associated or extracted for each of the linear component (factor) and represents the importance of the components in the ecosystem. In PCA analysis, the first four components PC1 (31.89%), PC2 (19.79%), PC3 (15.35%) and PC4 (12.96%) explained 79.97% of total variance. The rotated component matrix estimated that PC1 was having a very strong correlation among F-CO_2, algal abundance, EC, and pH. The PC1 also revealed that increase of epiphytic algal abundance (0.868) was correlated with decrease in F-CO_2 (-0.946), EC (-0.792) and pH (-0.789). This shows that F-CO_2 declined due to epiphytic algal colonization, which might be due to higher rate of photosynthesis compared to a low decomposition rate. In the study, it was found that epiphytic algal abundance was mostly contributed by the chlorophycean diversity. In PC1, similar to the findings of Brock, (1973), the acidic pH of the freshwater marsh might have facilitated the chlorophycean algal colonization, whereas it limited the growth of cyanobacteria. Thus, pH was found to be an important factor in determining selective epiphytic algal abundance on *U. aurea*. PC2 included NO_3-N (0.830), D-Si (0.790), WT (0.691) and BOD (0.519). It reveals that increase of nutrients like NO_3-N and D-Si increased BOD of the marsh. In PCA, increase of BOD with the increase of WT might be due to the labile organic matter of macrophyte origin in the freshwater marsh (Carpenter *et al*., 1979). A monotonous increase of NO_3-N with WT was also reported by Zheng *et al*., (2016). PC3 represents Chlorophyll *a* (0.875) and Total alkalinity (0.819). Thus algal productivity increased with the increase of Talk of the ecosystem. PC4 was having higher loading of SRP (0.847) and DO (-0.836), which indicates decrease in DO due to increase in SRP. This might be due to the excessive algal and macrophyte growth due to high SRP content that ultimately reduced DO in the marsh.

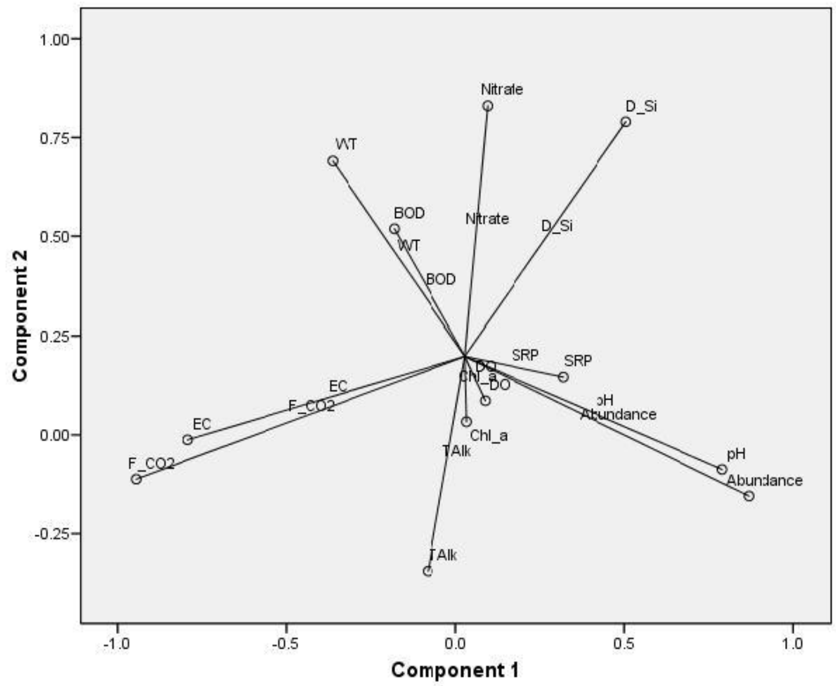

Fig. 2 Biplot of Principal Component Analysis of the variable

The effect of environmental factors on epiphytic algal colonization with respect to species abundance on *U. aurea* was also evaluated by Canonical Correspondence Analysis (CCA). Each orthogonal ordination axis (eigenvector) was associated with an eigenvalue which is the inertia extracted or correlation coefficients. CCA analysis included 10 environmental factors and 80 epiphytic algal species (Fig. 3). The first axis (Axis 1) of the ordination plot accounted for 33.41%, (eigenvalue: 0.358), whereas second axis (Axis 2) accounted for 19.09 % (eigenvalue: 0.205) of total variance. The Axis 1 of the CCA triplot was found to be the most significant canonical axis ($p < 0.05$) to correlate in between the environmental factors and the abundance of each algal species. In a CCA triplot, the relative importance of a particular parameter is represented by the corresponding centrifugal lines (Bodaghabadi *et al*., 2011). Therefore, as revealed also in PCA, D-Si, pH and SRP were the most important factors as observed towards the right ordination of the plot. Towards the left ordination of the plot, F-CO_2, EC and Talk were found to be the most important factors. Comparatively, a very high F-CO_2 was observed in the month of March-2013. F-CO_2 was found to be correlated with *Aphanocapsa* sp., *Merismopedia glauca*, *Closterium parvulum* and *Neidium dubium*. This indicates the adaptability of these species to the elevated CO_2 thus contributing to carbon sequestration in the marsh. Towards the left ordination of the plot, the lowest water temperature was observed in December 2012, and was correlated with *Spirogyra crassa*. Preference of *S. crassa* to grow in low temperature was also reported by Berry and Lembi, (2000). The EC was found to be correlated with *Amphora costata*. *Oscillatoria limosa*, *Closterium cynthia*, *Closteriopsis longissima*, *Arthrodesmus curvatus*, *Closterium gracile*, *Cosmarium impressulum*, *C. polygonum*, *Nephrocytium limneticum*, *Pinnularia interrupta* and *P. subcapitata*. A close association was observed among *Closterium gracile*, *Cosmarium polygonum*, *Euastrum turneri*, *Nephrocytium limneticum*, *Pinnularia subcapitata* and *Stauroneis* sp. *Oscillatoria subbrevis*, *Euastrum angolense* and *Eunotia* sp. were highly correlated with Talk and DO. These two environmental factors were also moderately correlated with *Closterium turgidum*, *Micrasterias foliacea*, *Cosmarium* sp.2, *Scenedesmus armatus*, *Gonatozygon aceleatum*, *Scenedesmus armatus*, *Staurastrum* sp.2, *Zygnema* sp., *Eunotia bilunaris*, *Navicula cryptocephala*, *Navicula* sp.2 and *Pinnularia microstauron*. The BOD, pH, D-Si, SRP and NO_3-N were found to be correlated with algal species like *Anabaena* sp., *Chroococcus* sp., *Ankistrodesmus falcatus*, *Closterium acerosum*, *Chlorobotrys regularis*, *Arthrodesmus gibberelus*, *Closterium acerosum*, *Oedogonium pusillum* etc. A positive interaction among NO_3-N, SRP and algal growth was also reported by Fried *et al*., (2003). The BOD was found to be highly correlated with *Eunotia vanheurkii* and *Scenedesmus dimorphus*. A moderate to low correlation was found in between *Spirogyra gracilis*, *Spirogyra rivularis*, *Scenedesmus bijuga*, *Ankistrodesmus falcatus* and BOD. A significant positive relation between green algal community and BOD was also revealed by Chia *et al.,* (2011). The D-Si was correlated with *Eunotia lunaris*. SRP was closely correlated with *Dictyosphaerium pulchellum*, *Caloneis bacillum* and *Sellaphora stroemii*. A close correlation was obtained among *Anabaena* sp., *Actinastrum* sp., *Closterium kuetzingii*, *Ankistrodesmus spiralis*, *Closterium dianae*, *Staurastrum sub-rotula*, *Navicula* sp.5, *Stauroneis anceps* and SRP. Broadly, we can divide the epiphytic algal community structure into four groups. Towards the right ordination (positive) of the plot, Group 1 include the months of August, September, October and November 2012, mostly effected by the BOD. To the same ordination, Group 2 include the months of May, June and July 2012, correlated with pH, D-Si, SRP and NO_3-N. There was no epiphytic algal colonization on *Utricularia* in the month of March 2013. This month was positioned single towards the left ordination (negative) of the plot, whereas January, February and April 2013 were grouped together.

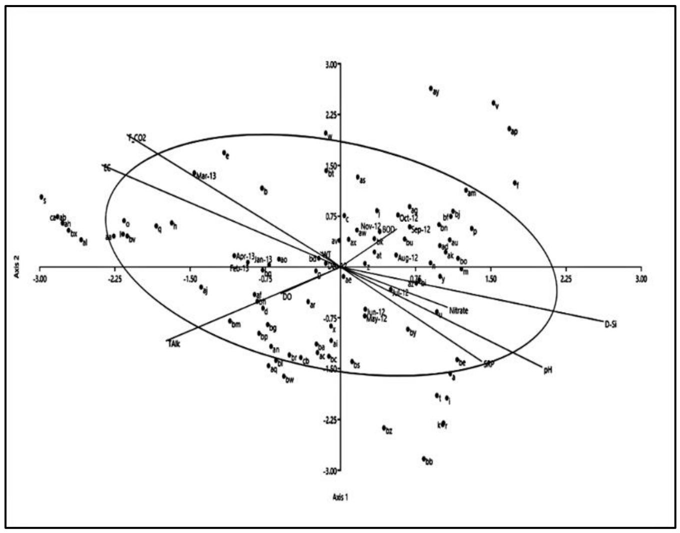

Fig. 3 Ordination diagram of Canonical Correspondence Analysis (CCA). Epiphytic algae are coded as: a. *Anabaena* sp. **b.** *Aphanocapsa* sp. **c.** *Chroococcus* sp. **d.** *Aphanothece* sp. **e.** *Oscillatoria sancta***. f.** *Merismopedia glauca***. g.** *Oscillatoria subbrevis***. h***. O. limosa***. i.** *Actinastrum* sp. **j.** *Ankistrodesmus falcatus***. k.** *A. spiralis***. l.** *Arthrodesmus curvatus***. m***. A. gibberelus***. n.** *Chlorobotrys regularis***. o.** *Closteriopsis longissima***. p.** *Closterium acerosum***. q.** *Closterium cynthia***. r.** *C. dianae***. s.** *C. gracile***. t.** *C. kuetzingii***. u.** *C. leibleinii***. v.** *C. navicula***. w.** *C. parvulum***. x.** *C. turgidum***. y.** *Cosmarium bioculatum***. z.** *C. blytii***. aa.** *C. impressulum***. ab.** *C. polygonum***. ac.** *Cosmarium* sp.2.**ad.** *Cosmarium* sp.3. **ae.** *Dictyosphaerium pulchellum***. af.** *Euastrum angolense***. ag.** *Euastrum* sp.1. **ah.** *E. turneri.* **ai.** *Gonatozygon aceleatum***. aj.** *Micrasterias foliacea***. ak.** *Mougeotia genuflexa***. al.** *Nephrocytium limneticum***. am.** *Oedogonium pusillum***. an.** *Onychonema leave***. ao.** *Pediastrum duplex***. ap.** *Pleurotaenium ehrenbergii***. aq.** *Scenedesmus armatus***. ar.** *S. armatusv. caudatus***. as.** *S. bijuga***. at.** *S. dimorphus***. au.** *S. quadricuada***. av.** *S. crassa.***aw.** *S. gracilis.***ax.** *S. rivularis***. ay.** *Staurastrum alternans***. az.** *S. crenulatum***. ba.** *Staurastrum* sp. 2. **bb.** *S. sub-rotula***. bc.** *Zygnema* **sp. bd.** *Amphora costata***. be.** *Caloneis bacillum***. bf.** *Cymbella ventricosa***. bg.** *Eunotia bilunaris***. bh.** *Eunotia* sp. **bi.** *Eunotia lunaris***. bj.** *Eunotia* sp.1. **bk.** *E. vanheurkii***. bl.** *Gomphonema affine***. bm.** *G. parvulum.* **bn.** *Navicula cryptocephala***. bo.** *Navicula* sp.1. **bp.** *Navicula* sp.2. **bq.** *Navicula* sp.3.**br.** *Navicula* sp. 4. **bs.** *Navicula* sp. 5. **bt.** *Neidium dubium***. bu.** *Neidium* sp.2. **bv.** *Pinnularia interrupta***. bw.** *P. microstauron***. bx.** *P. subcapitata***. by.** *Sellaphora stroemii.* **bz.** *Stauroneis anceps***. ca.** *Stauroneis* sp. **cb.** *Phacus chloroplastis***.**

Conclusion

The macrophyte, *U. aurea* was found to be a suitable host for colonizing diverse algal species. A total of 80 epiphytic algal species representing four different classes (Cyanophyceae, Chlorophyceae, Bacillariophyceae and Euglenophyceae) belonging to 36 genera were found to colonize on the macrophytic host. Chlorophyceae (47) was found to be the dominant colonizer followed by Bacillariophyceae(24), Cyanophyceae(8) and Euglenophyceae (1). Epiphytic algal abundance was found to be effected by different physico-chemical properties of water in the marsh. The WT, pH, EC, F-CO_2, NO_3-N, D-Si were found to be the most important factors regulating epiphytic algal colonization on the macrophytic host. The study provides insights into the diversity and primary productivity of epiphytons on *U. aurea* in the aquatic biocoenosis of a freshwater marsh.

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