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Temporal and Spatial Distribution of Epiphyton with Diversity Indices in a Lotic Ecosystem

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ABSTRACT

Lotic ecosystem monitoring is important to avoid pollution risks, safeguard biodiversity, and maintain ecosystem health. In this essence, it is impossible to ignore the involvement of epiphytic microalgae in riverine monitoring. Two host plants, such as the common reed, *Phragmites australis*, which is primarily found in marsh grasses, and the submerged, free-floating hornwort, *Ceratophyllum demersum*, were chosen for this study. The estimation of various biodiversity metrics, including Brillouin, Dominance (D), Equitability (J), Evenness (e^H/S), Margalef, Menhinick, Shannon (H), and Simpson (1-D), were conducted in Diyala River, Diyala province, Iraq, using data covering the four seasons. One hundred and forty-four taxa of epiphytic algae on both host plants were recorded. Among them, diatoms (Bacillariophyta) were the predominant group of algae on both plants comprising 84% and 92% of *C. demersum* and *P. australis*, respectively, followed by Cyanophyceae (3- 25%) and Chlorophyceae (3- 42%). Preferences of algae on particular host plants were observed. A spatial and temporal of epiphyton algae was observed on the plants. Some epiphyton species, such as *Gomphonema paravalum*, *Ulnaria acus*, *Cyclotella meneghiniana*, *Navicula gregaria*, and *Nitzschia amphibian* arerecorded in the river and are considered bioindicators for determining the water quality. By applying the recommended indices as mentioned in the text you have to recognize the trophic, saprobic, and halobic status of the investigated area based on the identified species. Based on the overall outputs of this study it is anticipated that the Diyala River is getting a pollution load; the level of pollution must be determined (beta-mesosaprobic, alpha mesosaprobic, polysaprobic) that needs to be overcome.

INTRODUCTION

Benthic producers are generally categorized as macroalgae, and microalgae collectively known as micro-phyto-benthos. Both sorts of aquatic organisms primarily support food-web dynamics and play a critical role in an ecosystem functioning **(UNEP, 2004)**. These tiny structures are vulnerable to various environmental factors, especially the light availability in which aquatic plants' growth and survival appeared to be susceptive for instance diatoms

(Laugaste & Reunanen, 2005, Ali *et al*., 2019). Diatoms, regarded as bioindicators, are more specific in their preference and tolerance of environmental conditions than most other aquatic biota. They are the first group of biotas used for detecting organic pollution. Both drought and flooding effects on diatomic community in rivers serve as a climate change impacts, and cause alteration in water quality **(Hassan *et al*., 2010; Hassan & Shaawiat 2016; Hassan *et al***. **2020)**. **Feio *et al***. **(2020)** considered diatoms as river elements of the environmental assessment. **Okito *et al***. **(2021)** reported that epiphyton algae (diatoms) are considered bio-indicted to the environmental conditions of aquatic ecosystems, both in the present and the past. Similarly, macrophytes play a vital role in regulating carbon and nutrient turnover in food web dynamics of shallow-aquatic environments **(Steinman *et al*., 2016)**. Nevertheless, algae in the lotic environment tend to attach to different types of objects. The selected host plants in this study are worldwide considered as nuisance aquatic plants and are distributed in many Iraqi aquatic ecosystems **(Wersal & Madsen, 2012)**. Moreover, algae are considered important bioindicators due to their broad geographical distribution and well-studied ecology. They react rapidly to environmental changes whether physical, chemical، or biological changes in water. Diatoms, in particular, are invaluable in water quality assessment and monitoring (**Chen *et al*., 2016; Elshobary *et al*., 2020)**. Diversity indices are commonly used to determine the environmental impact of the growth and survival of such tiny structures. The change in diversity indices is analarm of the impact of pollution **(Hosokawa *et al*., 2021)**. The indices reflected the response of benthic communities to the environmental change, and species density was most sensitive to the alteration in the environment **(Hosokawa *et al*., 2021)**. The diversity and richness of epiphytic algae were highly correlated with the growing and metabolic state of the host macrophyte **(Kassim *et al*., 2000)**, and many factors influence species diversity, such as light density, temperature, and nutrient concentration **(Dehghan *et al*., 2022)**. Different diversity assessment tools are useful for figuring out water quality in a lotic ecosystem **(Hassan & Shaawiat, 2016, Ali *et al*., 2018)**.

Park *et al*. **(2020)** found that the composition of a community of algae (benthic and periphytic) is reflected in the environmental conditions. Subsequently, **Carvalho *et al***. **(2020)** explained that morphological differences between the aquatic plants as a substrate for epiphyton caused different colonization of species on the host plant. Furthermore, **Laugaste and Reunanen (2005)** emphasized that the morphology of the host plant is the most important factor of epiphyton diversity.

Toporowska *et al*. **(2008)** reported different biomass of epiphyton on four selected host plants and observed preferences of periphyton species on particulate host. Moreover, the temporal variation was noted, with high values recorded in spring and autumn. This study was designed to ascertain the species composition of epiphyton (algal) inhabiting Diyala River and figure out the underlying effects of the city discharges on the algal composition and water quality of the river.

MATERIALS AND METHODS

Description of Diyala River

Diyala River is one of the main tributaries of the Tigris River and is considered as freshwater artery of Iraq. Out of the 386Km length of the river, 300km falls in Iraqi territory which originates from Iran. This 100- 500m broad river possesses a slope of 1.85km **(Al-Ansari, 2013)**. The Diyala River is a unique water resource being utilized for various purposes, such as drinking, industrial, irrigation, fishing, and tourism **(Hassan *et al*., 2018)**.

Sampling strategy

In this study, we designed three locations to cover the entire length of the Diyala River (Fig. 1). Location (I) N44.22″ 39 ’ 34°, E54.92″58’44° is located in Jalawlaa City, after Himreen impoundment, and the river is wider than other studied sites. Besides, it is surrounded by an agricultural land. Location (II) N41.87″ 44 ’ 33°, E54.29″37’44° near Baquba City, and the river receives different discharges from different domestic and industrial activities. Both riverbanks are supported by concrete covering, while location (III) N34.69″61’33° E25.05″32’44° exists in the south of Baghdad City, near Rustumia sewage treatment plant; it is impacted by different domestic and agri-industrial activities.

Physicochemical parameters

Seventeenth physicochemical parameters were measured, according to the outlines of **APHA (2005)**. The parameters are air and water temperature (°C), pH, water flow (WF), light penetration (LP), total dissolved solids (TDS), total suspended solids (TSS), turbidity (TU), electrical conductivity (EC), salinity (SA), total hardness (TH), calcium (Ca), magnesium (Mg), dissolved oxygen (DO), total alkalinity (TA), total nitrogen (TN), and total phosphorus (TP).

Algal samples

Epiphytic algal samples were monthly collected from two aquatic macrophytes throughout October 2016 to July 2017. The months were further categorized into four respective seasons around the year as follows: fall (October and November), winter (December and January), spring (February and March), and summer (April, May, June, and July).

The macrophytes were collected from each site and kept in nylone bags with little river water, and preserved with buffer formalin (4%) in the field. Both macrophytes were present in all sites during the study period, and three replicated samples of host plants were collected from each site. *P. australis* was cut into small parts (2- 3cm long), and the algae were removed using a toothbrush, collected in a vial, while the attached algae on *C. demersum* were removed by shaking vigorously with distilled water. Each algal sample was preserved in Lugol’s iodine solution, and then each sample was divided into two parts. The first part (I) was used for diatom identification by clearing the frustules using Patrick and Reimer methods **(APHA, 2005)**, and enumeration was conducted according to **Hötzel and Croome (1999**). The second part (II) involved identifying the rest of the groups by mounting permanent slides with glycerin jelly method **(APHA, 2005)**. The algae count was conducted using the micro-transect method for diatoms and the hemocytometer method for non-diatomic algae, following the methods outlined by **Martinez *et al*. (1975)**. The taxonomic references of **Round *et al*. (1999)** were used for identification. In addition to species composition, the Chlorophyll-a (C) and Phaeophytin-a (P) were also measured.

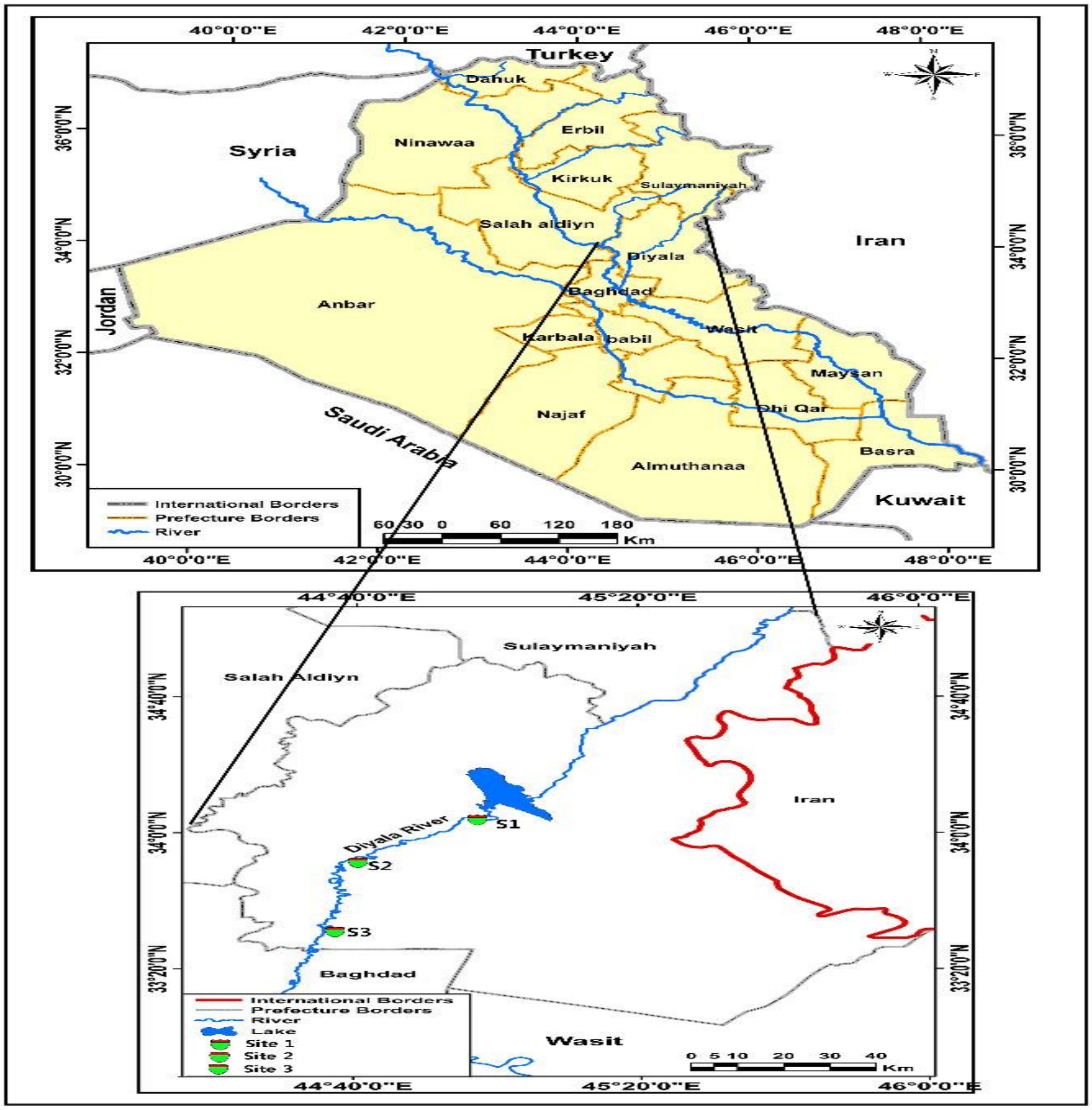


Fig. 1. Three sampling locations are highlighted with zoom-in map in the bottom

Biodiversity indices

In this study, eight indices were calculated as follows: Brillouin, Dominance (D), Equitability (J), Evenness (e^H/S), Margalef, Menhinick, Shannon (H), Simpson (1-D). The estimates of biodiversity indices are based on the references of **Magurran (2004)** and **Salman *et al*. (2017)**.

Statistical analysis

The statistical analyses were performed using R-package, following the guidelines outlined by **(Love *et al*., 2019)**, with JASP used for correlation plots. Student’s T-test was applied to test the relation between the sites and seasons. Analysis of variance (ANOVA) was applied to estimate multi-variance interaction between seasons, sites، and host plants. Post Hoc tests and Pearson’s correlations were used for parametric comparisons. Canonical correlation analysis (CAA) was used by the performance of the CANOCO software to examine the relationship between algal species and environmental parameters, according to the method outlined by **Ter Braak and Smilauer (2002)**.

RESULTS

Physicochemical parameters

The results of physicochemical parameters showed that the parameters values varied among the studied seasons. The river water was alkaline and very hard. Additionally, the DO was higher than 4mg/ l, while the BOD recorded a higher value at site 3. The river was classified as brackish water according to **Team (2004)**. The trophic status of the river (in terms of TN and TP), was classified as oligotrophic according to **Bayram and Kenanoğlu (2017)**.

Table 1. Range of physiochemical and biological parameters during the study period in Diyala River

Parameter	Code	Range
Air temperature (°C)	air °C	12.2- 46.3
Water temperature (°C)	water °C	10.2- 30.4
pH	pH	7.3- 8.8
Water flow(m.Sec^{-1})	WF	0.2- 1.2
Light penetration (cm)	LP	29.8- 77.2
Total dissolved solids (mg.l^{-1})	TDS	395.5- 908.6
Total suspended solids (mg.l^{-1})	TSS	0.1- 0.24
Turbidity (NTU)	TU	8.1- 43.5
Electrical conductivity(µs.cm^{-1})	EC	810.3- 2335
Salinity ‰	SA	0.53- 1.4
Total hardness (mg CaCo3.L^{-1})	TH	295.9- 704.6
Calcium (mg.L^{-1})	Ca	70.8- 137.8

Magnesium (mg.L^{-1})	Mg	20.3- 79.7
Dissolved oxygen (mg.L^{-1})	DO	4.5- 11.1
BOD (mg.L^{-1})	BOD	1.3- 34.3
Total alkalinity (mg CaCo3.L^{-1})	TA	54.3- 122.3
Total nitrogen (µg/l-1)	TN	0.1- 16.2
Total phosphorus (µg/l^{-1})	TP	0.01- 0.08
Total silicate (µg.L^{-1})	TS	226.3- 446.1
P. australis Chlorophyll-a µg. Cm $^{-2}$	C1	7.161- 26.2
phaeophytin-a µg. Cm $^{-2}$	P	0.03- 18.2
C. demersum Chlorophyll-a µg. Cm $^{-2}$	C	10.5- 34.9
phaeophytin-a µg. Cm $^{-2}$	P2	4.0- 25.3

Floristic analysis

A total of 163 taxa of epiphyton were identified and enumerated (Tables 2, 3). A total of 150 taxa were attached on *P. australis*, while 163 taxa were on *C. demersum*. The dominant group was Bacillariophyta (diatoms), followed by Chlorophyceae and Cyanophyceae. *P. australis* comprised 118 species of diatoms and was the predominant group. While on *C. demersum*, there were 99 spp., with chlorophyceae forming a higher percentage on *C. demersum* (38spp.), while on *P. australis*, there were 16 species. In the same trend, the Cyanophyceae were 25spp. on *C. demersum* and 16 spp. on *P. australis*. A temporal variation was noted in algal composition on both host plants (Fig. 2). One species of Euglenophyceae was obsereved on *C. demersum*. Overall, population was dominated by *Navicula* (15 and 26spp), *Cymbella* (8 and 11spp.), *Nitzschia* (10 and 14), and *Gomphonma* (10 and 6spp) on *C. demersum* and *P. australis*, respectively (for each genus, the number are mentioned in parenthesis).

Table 2. The identified epiphyton algae on the two-host plant in Diyala River

Phragmites australis

Anabaena sp.
Aphanocapsa sp.
Chroococcus limneticus lemmerman
Chroococcus turigdus (kutz.) Nageli
Merismopedia convulata de Brebisson
Nostoc sp.
Oscillatoria limentica lemmermann
O. limosa (Rath.) Agardh
O. tenuis Agardh
O.prolifca (Crev.) Gomont

Rhicosphenia curvata (kutz.) Gtunow
R. marina (W. smith) M. schmidt
Stauroneis amphioxys var. *amphioxys* Gregory
A. exigua var . *Heterovalvata krasske*
A. minutissima kutzing
Amphipora paludosa w. smith
A. commutate Grunow
A .veneta Kützing
A.normani Rabhenhorst
Bacillaria paradoxa Gmelin

O. princeps voucher
Phormidium tenue Gomont
Spirulina laxa G. M. Smith
S. subsalsa Oersted
S. major (wittr.) kirchner
S. nordestrdtii Goment
Actinastrum gracilium G.M. Smith
Cladophora glomerata (l.) kutzing
Chlamydomonas sp.
Cloelastrum microporum Nageli
Coelastrum Diana Ehernberg
Coleochaete pulvinata A. Braunin
Mougeotia scalaris Hassal
Oedogonium sp.
Oocystis elliptica west
O. solitaria Wittrock
Pediastrum boryanum (Trup.) meneghini
Scendesmus abundens (Lag.) chodat
S. bijuga (Turp.) Lagerheim
S. arcuatus lemmermann
S. quadricauda (Turp.) de Brebisson
Spirogyra sp.
EUGLENOPHYCEAE
Euglena acus Ehrenberg
COSCINODISCOPHYCEAE

Cyclotella catenata (A. Braun.)
C. meneghinana kutzing
C. ocellata pantocsek
C. operculata (Ag.) kutzing
C. glometra Bachmann
Melosira granulate (EHr.) Ralfs
M. varians Agardn
M. italica (Her.) kutzing
M. roesana Rabenhorst
Achnanthes effinis Grunow
A. *exigua var* . constricata Torka
A. microcephala kutzing
A. *veneta* kutzing
G. spencerii (W.Sm.) Cleve
Hantzschia amphioxys (Ehr.) Grunow
Mastogloia smithii var. amphicephala Grunow
Navicula anglica Ralfs
N. cari Ehrenberg

Caloneis amphisbaena (Bory.) Cleve
Centrorella reichelteii voigt
Cocconeis pediculus Ehrenberg
C. placetula var. *Euglypta* (Her.) Cleve
C. placentula var. *lineata*
Cymatopleura solea (Brab.) w. smith
Cymbella affinis kutzing
C. cistula (Hemp.) Grunow
C. cymbeformis (kutz.) var. Heurck
C. prostrate (Berk.) Cleve
C. gracilis (Rabh.) Cleve
C. lanceolata (Ehr.) van. Heurck
C. lanceolata (Her.) var. Heurck
C. prostrate (Berk.) Cleve
C. tumidula Grunow
C. microcephlo Grunow
C. tumida (Breb.) var. Heurck
C. obtusiscula (kutz.)
C. parava (W.smith) Cleve
C. turgid (Greg.) Cieve
C. ventricosa kutzing
Denticula tenius kutzing
Diatoma anceps (Her.) Grunow
D. vulgare Bory
D. vulgare var. *berve* Grunow
D. vulgare var. *product* Grunow
Diploneis elliptica (kutz.) cleve
D. ovalis var. *oblongella* Nageli
D. puella (schum.) cleve

F. rumpenes kutzing
F. capucina Desmazieres
F. rumpenes Kützing
F. intermedia Grunow
Gomphonma constrictum var. *capitata* (Ehe.) Cleve
G. angustatum(Kütz .) Rabenhorst
G. olivaceum (lyng.) kutzing
G. paravalum (kutz.)Grunow
G. tergestinum (Grun.) Fricke
G. ventricosum Gregoty
Gyrosigma acuminatum (kutz.) Rabenhost
S. anceps var. *anceps* Ehrenberg
Stenopterbia intermedia (lewis) Brebisson

N. cincta(Ehr.) Kützing
N. cincta var. heufleri Grunow
N. cryptocephile var. *veneta* (kutz.) Grunow
N. cryptocephile var. *intermedia* Grunow
N. gregaria Donkin
N. gracilis Ehrenberg
N. exilissima Grunow
N. gastrum(Ehr.) Kützing
N. gothlandica Grunow
N. gracilis Ehrenberg
N. *falaisiensis* Grunow
N. falaisiensis var. lanceola Grunow
N. balophila (Grun.) cleve
N. inflate Donkin
N. lanceolata (A.C.Ag.) kutzing
N. menisculus schumann
N. minuscula Grunow
N. pygmaea kutzing
N. radiosa kutzing
N. spicula cleve
N. salinarum Grunow
N. schroeteri Meister
N. spicula Cleve
N. viridula var. rostellata (Kütz .) Cleve
Nitzschia acuta Hantzsch
N. apiculata (Greg.) Grunow
N. amphinia Grunow
N. closterium (her.) w. smith
N. dissipata (kutz.) Grunow
N. commutate Grunow
N. filiformis (W.smth)
N. frustulum kutzing
N. gracilis Hantzsch
N. hyngarica Grunow
N. paleacea (kutz.) w. smith
N. paleacea Grunow
N. sigma (kutz.) w. smith
N. vermicularis (kutz.) Grunow
Peronia erinacea Brebisson
Pinnularia leptosome Grunow
P. vriidis (Nitzs.) Ehrenberg
Pleurosigma elongyatum smith

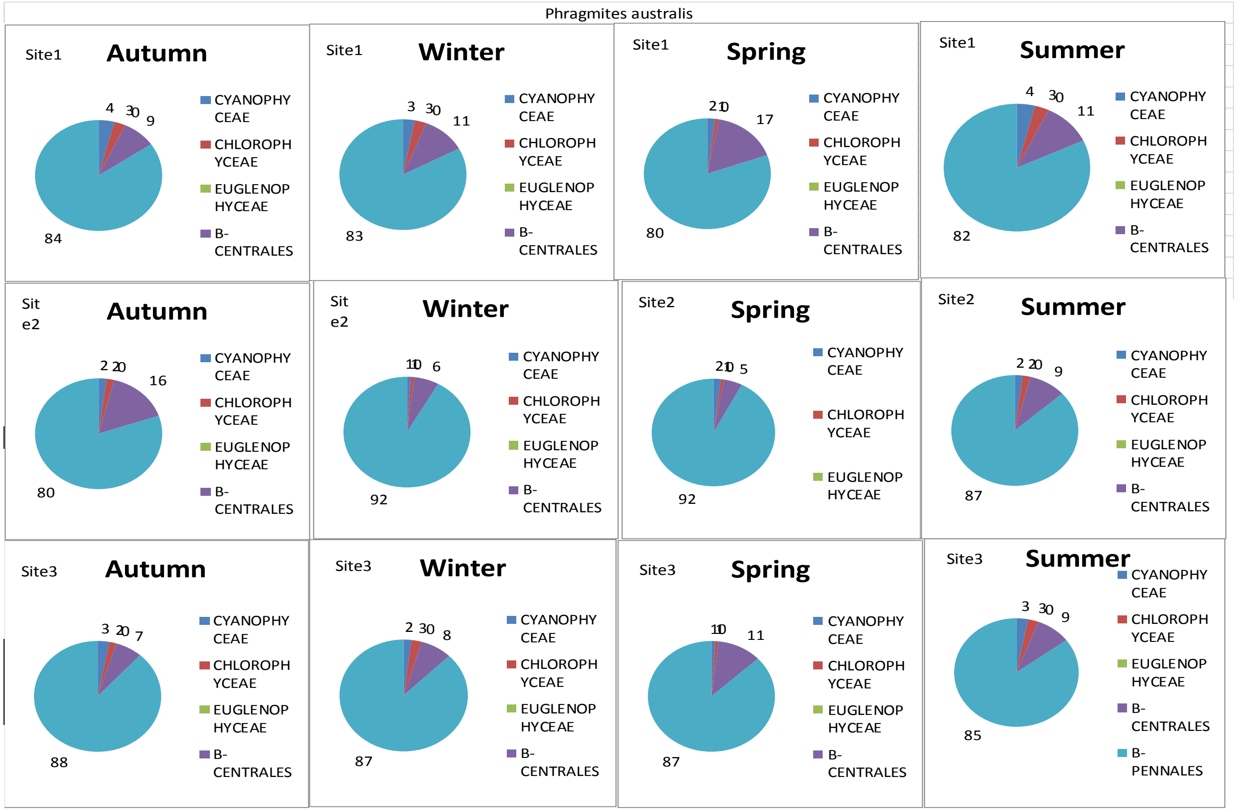
Surirella lineais W. smith
Syneba capitata Ehrenberg
S. linearis var. *constricta* (Ehr.) Grunow
S. ulna (Nitzs.) ehrenberg
C. demersum
Anabaena sp.
Aphanotheca microscopica Nageli
Aphanocapsa rivularia (Carm.) Rbenhors
Chroococcus dispersesus(Keis.) Lemmermann
C. limneticus lemmermnn
Gomphsphaeria lacustris chodt
Lyngbya major meneghinii
L. limnetic
L. majuscule Harvey
Merismopedia convulta de Brebisson
M.glauca Nageli
Nostoc sp.
Osillatoria amphibian Agardh
O. acutissma Gardner
O. agrdii Gomont
O. curvepes Agardh
O. chalybae (mertens) Gomont
O. Formosa Boey
O. limnetica lemmermann
O. limosa (Roth.) Agardh
O. prolifca (Crev.)Gomont
O. princeps vauher
Phormidium tenue Gomont
Spirulina major (Witt.) Kirchner
S. subsalsa kutzing
Ankistrodesmus falcatus (Corda) Ralfa
Cladophora sp.
C. glomerata (lemm.) kutzing
Closterium dianae Ehreberg
C. comarium botrytis meneghinii
C. venus Kutzing
C. subtumidium nordstedt
C. jemnri Ralfs
Cosmarium
C. jennri Ralfs
Eudorina sp.
Oedogonium sp.

Ceratophyllum demersum

Pediasstrum boryanum (Turp.) Memeghinii
C. placentula Ehrenberg

P. simplx (meyen) lemmermann
P. tetras var. tetradron (Corda.) Rabenhorst
P. tetras (Ehr.) Ralfs
Scenedesmus arcuatus lemmermann
S. abundens (lag.) chodat
S.abundens var. *brevieande* (Kirch)
S. bijuga (Turp.) Lagerheim
S. bijuga var. alterna (Turp.) Lagerheim
S. opoliensis Richter
S. quadricauda (Turp.) de Brebisson
S. quadricauda var. longispina smith
S. quadricauda var. maxima (west & west)
S.brasiliensis Bohlin
S. dimorphus (Turp.) Kützing
S. denticulatus lagerheim
S. intermedius chodat
S. opoliensis richter
S. qudricauda (turp.)debrebisson
S. qudricauda var. *longispina* smith
S. qudricauda var. *maxima* (wst&west)
Spirulina major (witt.) kirchner
Spirogyr sp.
Staurastrum gracile Ralfs
Tetraedron minimum (A. Braun) Hansgirg
Nitella sp.
Euglena minuta Prescott
Aulacoseira varians Agardh
Cyclotella meneghiniana kutzing
C. striata (Kütz.) Grunow
C. Bachmann
C. Bachmann
C. meneghiniana Kützing
Stephaenodicus sp.
S. hantzschiiGrunow
Achnanthes sp.
A. brevipes var. intermedia (kutz.) cleve
A. hungarica Husted
A. line W. Smith
A. plonenesis Hust
Amphora coffeaformis Agardh
A.ovalis var. pediculus Kützing
A. coffeaeformis Agardh
A. commutata Grunow
Bacillaria paxillifer (Mul.) Hendy
Cocconeis pediculus Ehrenberg
C.placental Ehrenberg
C. placentula var. lineata (Her.) Cleve
C. placentula var. euglypta (Her.) Ceve
Cymataptopleura Ellipitea
Cymbell affinis Kutzing
C. cistul (Hemp.) Grunow
C. aspera Ehrenberg
C. Helvetica kutzing
C. microcephala Grunow
C. turgida (Greg.) Cleve
C. tumida (Breb.) var. Heurck
C. ventricosa Kutzing
Cymatoplora sola de Brebisson
C. Ellipitca (Breb.) smih
Diatom vulgare Bory
D. vulgare var. brevis Grunow
D. vulgare var. lineais V. H.
D. elongatum. (Lyngb) Agardh
Diploneis puella Schumann
D. ovalis (Hisle) Cleve
D. elliptica kutzing
Epithemia sorex kutzing
E. zebra (Her.) kutzing
Fragilaria brevistriata Grunow
Fragilaria bicapitata Mayer
F. construens Ehrenberg
F. intermedia
Gomphonema acuminattum Ehreberg
G. olivaaceum (Horne.) Dawson
G. acumonatum var. turri Ehrenbery
G. ougur Ehrenbergv
G. constrictum var. capitata (Her.) Cleve
G. gracile Ehrenberg
G. lanceolatum Ehrenberg
G.intricatum var. pumida Grunow
G. olivaceum (lyng.) kutzing
G.angustatum var.producta
G. parvulum (kutz.)
Gyrosigma spenceri var. nodifera Grunow
G.tenuirostrum (Grun.) Cleve-Euler
Mastigola Elliphca var. *dansei* Agardh
Masrtigola smithii Thwaites
Navicule cryptocephala kutzing
N. inflate Donkin
N. vadiosa kutzing
N.parava ralfa
N. phyllepta

C.placental var.euglypta (Her) cleve
N. radiosa Kützing
N. apiculata (Greg.) Grunow
N. sigma (Kütz) W.Smith
N.cuspitata kutzing
N. spicula (Dick.)cleve
N. vadiosa var. *tenella* (Breb.)Grunow
N. mutica kutzing
N. parva Ralfs
N. rhyncocephala kutzing
Nitzchia amphilia Grunow
N. Filiformis (w. Smith) Husted
N. cuspitata kutzing
N. apiculata (Greg.)
N. clausii Hantzsch
N. dissipata (kutz.)Grunow
N. granulate Grunow
N. frustulum var. *perminuta* Grunow
N. sigma (Ehr.) W.smith
N.sigmaoids (Ehr.) W. SMITH
Pleurosigma delicatalum W. smith
Surirella ovalis de Brebisson
S. robusta Ehrenberg
Synedra acus kutzing



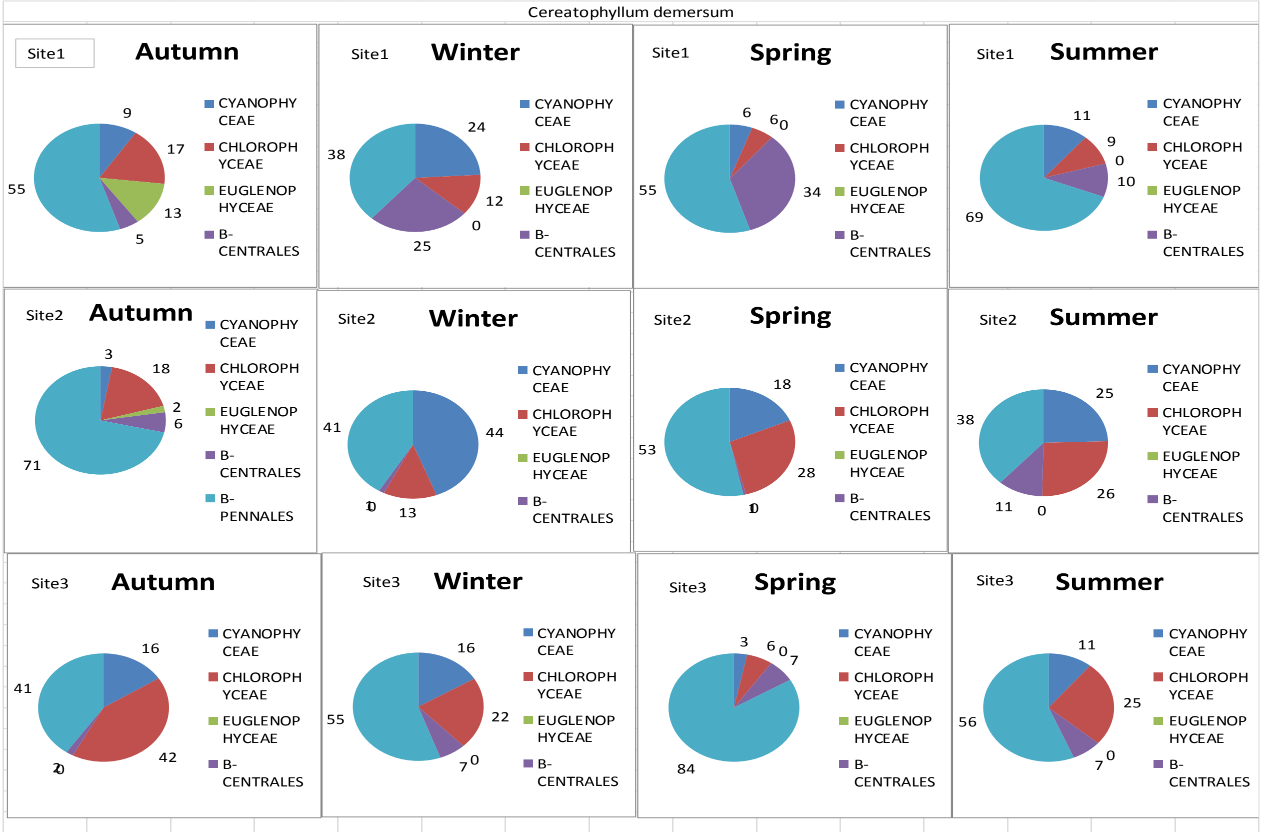


Fig. 2. Percentage of algal genera on the two host plants during four seasons

Table 3. Preference and presence of some epiphyton algae on specific host plant or sites

Group		Site	Hostplant
1	*Cymatopleura solea* (Brab.) *w. smith*	All	*P.australis*
	Navicula viridula var. *rostrata* Skvortsov	2	*P.australis*
	Cymataptopleura ellipitea Krammer	1	*C. desmersum*
2	*Achnanthidium brevipes* var. *intermedia* (kutz.) cleve	1	*C. desmersum*
3	*Encyonopsis microcephala* (Grunow) Krammer	All	*P. australis*
	Snowella lacustris (Chodat) Komárek & Hindák	1&2	*C. desmersum*
	Cymbella aspera Ehrenberg	2	*C. desmersum*
	Gomphonema acuminattum Ehreberg	All	*C. desmersum*

Diversity indices

The index results showed the highest values at site 3 for Brillouin, Equitability, Shannon and Simpson indices (3.621, 0.871, 3.724, and 0.969, respectively), while the lowest values were recorded at site 1 for Brillouin, Dominance, Equitability, Evenness, Shannon and Simpson (3.04, 0.07, 0.785, 0.424, 3.04, and 0.93, respectively) on *Phragmites australis* (Fig. 3). On the other hand, the lowest values for Brillouin (2.426), Equitability (0.787), Margalef (3.582), Menhinick (0.727), Shannon (2.5) and Simpson (0.898) indices on *Cereatophyllum demersum* were recorded at site 2 in spring and summer. Additionally, the lowest values were observed at site 3 in winter (0.026) and autumn (0.48). While, the highest values were at site 3 in the summer (3.644, 10.32, 3.792، and 0.974) for Brillouin, Margalef, Shannon، and Simpson indices, respectively. Additionally, the highest values were recorded at site 2 in spring for Dominance (0.102), Equitability (0.964), and Evenness (0.879); while at site 1, the Menhinick (3.366) showed the highest value among other indices.

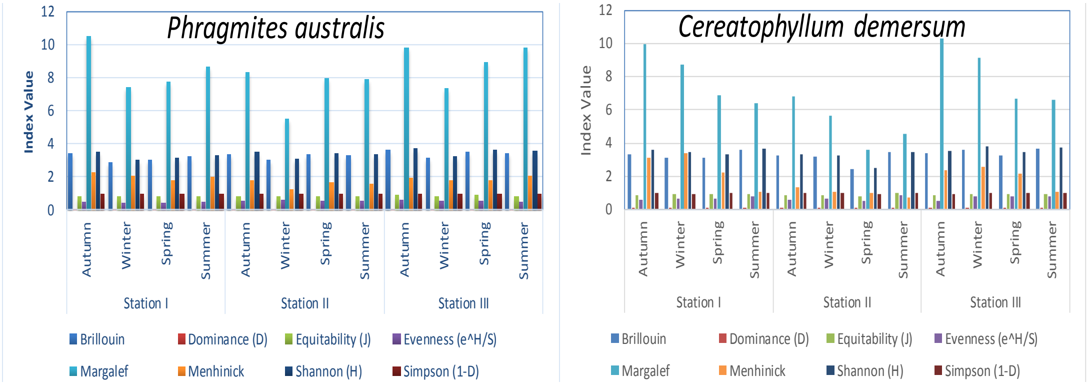


Fig. 3. The seasonal variation of diversity indices during the study period

Table 4. ANOVA analysis for indices

Case	Sum of squares	df	Mean square	F	p
			Ceratophyllum demersum		
Index	445.745	7	63.678	132.546	1.38E-35
Seasons	5.265	3	1.755	3.653	0.017
Index ✻ seasons	23.059	21	1.098	2.286	0.006
Residuals	30.747	64	0.48		
			Phragmites australis		
Index	610.577	7	87.225	656.796	4.89E-57
Seasons	3.197	3	1.066	8.026	1.28E-04
Index ✻ Seasons	10.071	21	0.48	3.611	3.88E-05
Residuals	8.499	64	0.133		

Note. Type III sum of squares.

The statistical analysis showed a significant difference between indices temporally and between them for both host plants (Table 4 & Fig. 4).

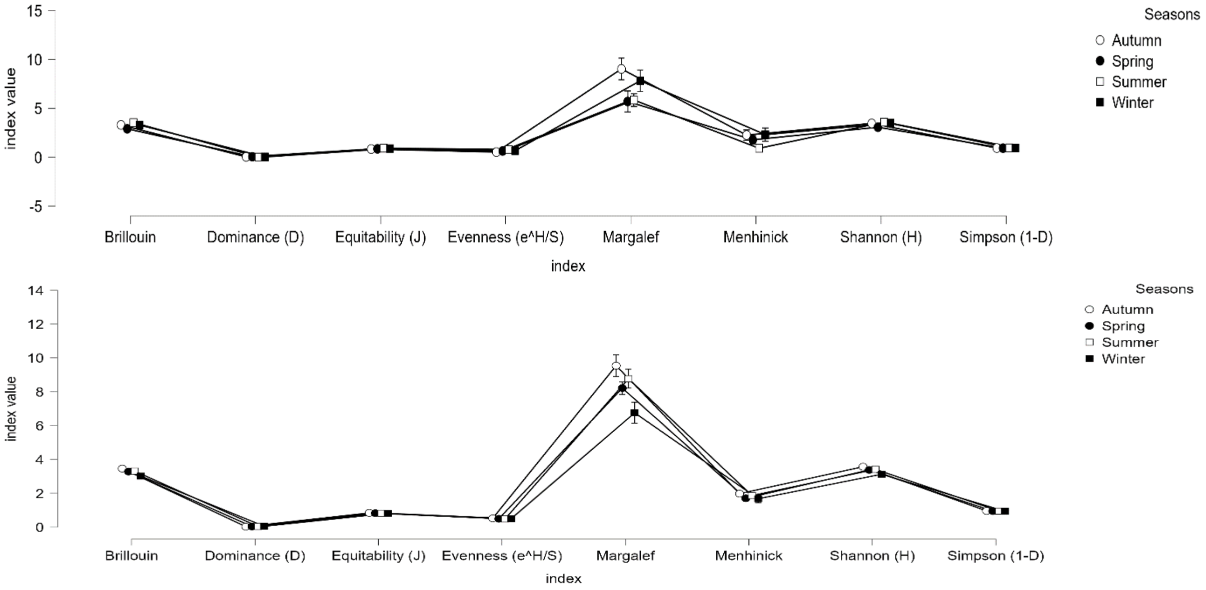


Fig. 4. Descriptive plots for indices of the two host plants

Fig. (5) illustrates the raincloud plots of indices in each season. The Margalef index was significantly different from other indices, while both Brillouin and Shannon indices resembled each other. The results of post hoc comparison between seasons showed a significant correlation between autumn and spring for *C. demersum (P*< .05). The temporal variation in seasons and *P. australis* showed a significant difference *(P*< .05) of epiphytic algae on *P. australis* between autumn and winter and between summer and winter (Table 4), while on *P. australis*, a significant correlation was obtained between autumn & winter as well as between summer and winter (Table 4).

Shannon–Weaver index recorded the lowest values at site 2 and high values at site 3. These values ranged from 1.3 in spring at site 2 to 3.7 in autumn at site 3 (*P. australis*), in addition to 1.2- 3.9 in spring and autumn at sites 2 and 3 (*C. demersum*), respectively. The results of the index did not show any spatial variation of epiphytic algae on both host plants, while only for *C. demersum* a temporal significant variation was noticed between autumn and spring (Table 5).

CAA analysis showed four groups of epiphytic algae on the two host plants; for group 1, algal species (Fig. 6) were related to air and water temperature, while group 2 of algal species was related to DO, LP, and P. Moreover, group 3 showed a relation with EC, TDS, TS, TH, Mg, and Ca, while group 4 was related to TP, TSS, TU, SA، and TN.

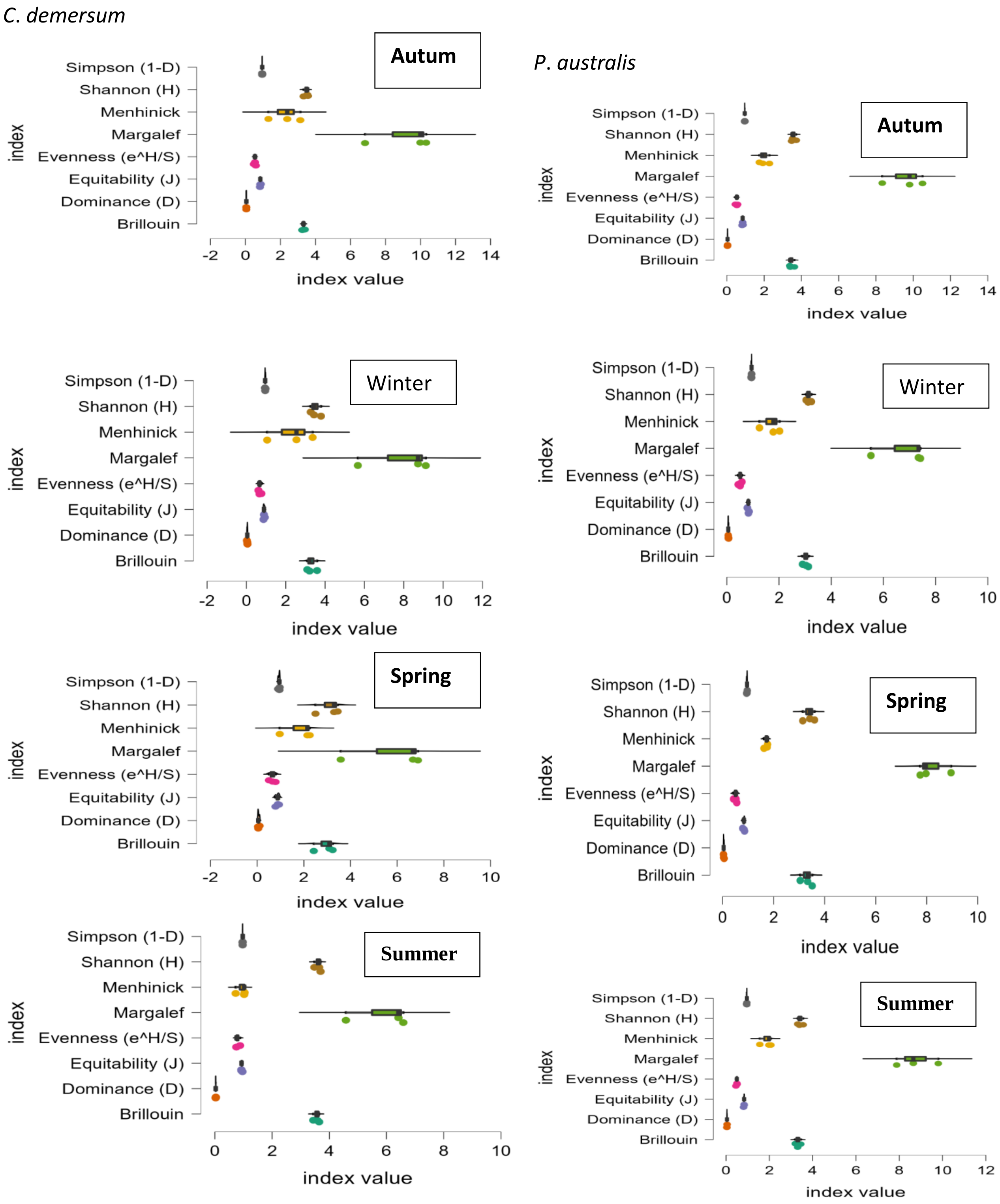


Fig. 5. Raincloud plots for indices and seasons throughout the study period

Table 5. ANOVA analysis for indices

		Mean difference		t	p-Tukey
		Phragmites australis			
Autumn	Spring	0.558	0.2	2.79	0.034*
	Summer	0.476	0.2	2.377	0.092
	Winter	0.12	0.2	0.601	0.931
Spring	Summer	-0.083	0.2	-0.413	0.976
	Winter	-0.438	0.2	-2.189	0.137
Summer	Winter	-0.356	0.2	-1.777	0.294
		Ceratophyllum demersum			
Autumn	Spring	0.25	0.105	2.38	0.091
	Summer	0.155	0.105	1.473	0.459
	Winter	0.503	0.105	4.778	0.00006246***
Spring	Summer	-0.095	0.105	-0.907	0.801
	Winter	0.252	0.105	2.398	0.088
Summer	Winter	0.348	0.105	3.304	0.008**

* $P < .05$, ** $P < .01$, *** $P < .001$.

Note. P-value adjusted for comparing a family of 4.

Note. Results are averaged over the levels of index.

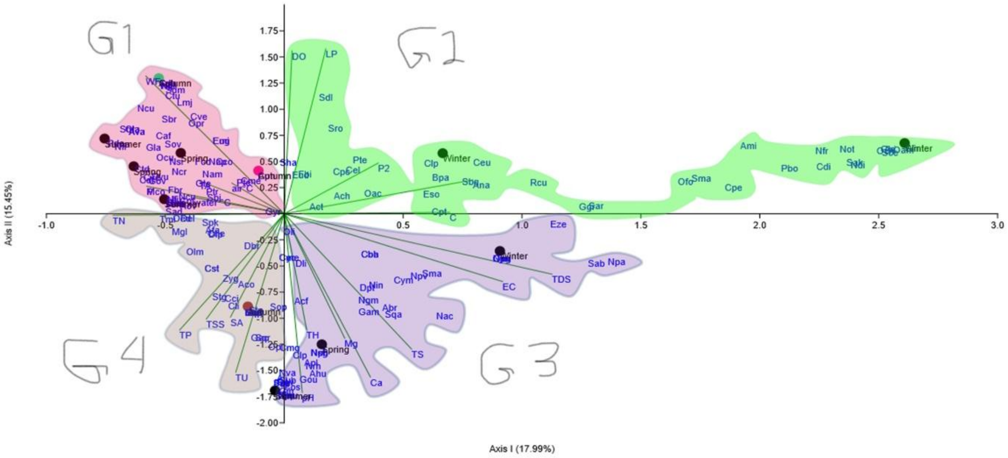


Fig. 6. CCA of algal species on the *C. demersum*

The CAA also revealed four groups of algal species on *P. australis* as follows: LP recorded a correlation with group 1 of algal species. In Group 2, TSS, TH, P2, TP, and TS were correlated with the group algal species. Mg, Ca, SA, and TDS showed a correlation with group 3, whereas group 4 of algal species was correlated with WF, TN, and TA (Fig. 7).

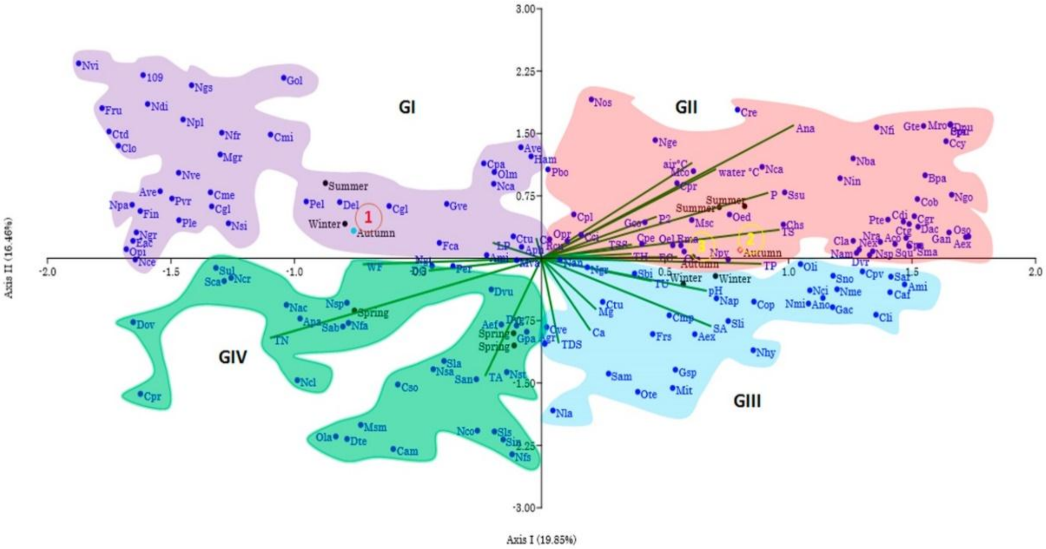


Fig. 7. CCA of algal species on the *P. australis*

DISCUSSION

This study has established a stance that epiphyte plankton presence on different host-plants indicates water quality problems in one of the important freshwater river (Diyala) of Iraq. The overall results of the study reveal that the diatoms in terms of the number of species were decreased on *C. demersum* compared to *P. australis*. This indicates an unhealthy state of the aquatic plants that does not allow being dense to provide a shade for diatoms. A study by **Shaawiat and Hassan (2017)** noted a high number of diatoms on the *C. demersum* (142 diatom taxa), while *P. australis* had 136 epiphytic diatom taxa. In addition, the different nature of the host plants under study (*C. demersum* being a wholly submerged plants, while *P. australis* was an emergent plant) led to competition among these algae for light and other requirements in the assemblage of epiphyte on host plants **(Hassan *et al*., 2007)**. The non-diatomic groups attached to *C. demersum* were more than thaose to *P. australis*, the attachment ability of diatoms on aquatic plants may be due to the specificity of host plant morphology and the mechanism that mediated the attachment of algal groups to stabilize their mobilization **(Pettit *et al*., 2015)**. The Cyanophyceae species preferred C. *demersum* as a host plant (25spp) in contrast with 16spp on *P. australis*.

Burak (2018) reported a different epiphytic algae composition on different host plants and interpreted the results as the preference of algae to biotope. In this study, some species were also attached to a particular host (Table 3), while other species were found to be on both host plants.

Iraqi aquatic ecosystems are characterized by high silicate concentrations, hence the dominancy of diatoms is well-known in these ecosystems **(Ali *et al*., 2018; Maulood & Hassan, 2021)**. Both host plants recorded a high number of diatomic species during this study. The previous studies of epiphytic algae on the *C. demersum* recorded a higher number of taxa in comparison with *P. australis* in lotic ecosystems **(Al-Hassany *et al*., 2014; Shaawiat & Hassan, 2017)**. These results revealed that *C. demersum* was under the impact of pollution (less tolerant) and it is considered unhealthy in contrast with *P. australis*, which is tolerant to pollution **(Hassan *et al*. 2018; Milke *et al*., 2020)**. The percentage of Cyanophyceae species was higher than Chlorophyceae on both host plants in this study. The shift from these groups reflected the alteration in the aquatic environment **(Li *et al*., 2013; Sin & Lee, 2020)**. These results indicate that anthropogenic pollution leads to an increase in the percentage of Cyanophyceae with the shortage of water supply dilemma in the river. In addition, the nature's differences of the host plants under this study (*C. demersum* is wholly submerged plants, while *P. australis* was emergent plant) lead these algae to compete the light and other requirements for assembling the epiphyton on the host plants **(Hassan *et al*., 2007)**.

Benthic algae have recently been used as a tool for water quality monitoring and bioindicators **(Florescu *et al*., 2015; CEN, 2018)**. The *Gomphonema parvulum* and *Ulnaria acus* were found on both host plants, and these species are considered as indicators of organic pollution **(Maishale & Ulavi, 2015)**. *Cyclotella meneghiniana* (on both plants), *Navicula gregaria*, and *Niztschia amphibia* (only on *P. australis*) were indicated as pollutant species and increasing in nutrients **(Potapova & Carlisle, 2011)**. Many authors have reported that the *Cyclotella* is indicated for euotrophic status, specifically *Cyclotella meneghiniana* and *Ulnaria acus*, are indicators of euotrophic status. However, the results of total nitrogen and Chlorophyll a concentrations are extremely high, as indicated in Table (2), which means the water is hypereutrophic resulting from the discharging from different sources **(Tas *et al*. 2002; Stoermer & Julius 2003; Hassan *et al*., 2010)**. Authors mentioned that the diversity and richness indices reflected the relation between epiphyton and themetabolic state of the host plant **(Hassan *et al*., 2007)**.

The high values of indices recorded in autumn and spring are caused by suitable conditions in these seasons for epiphyton growth and its migration activity on host plants **(Burak, 2018)**. **Stevenson and Bahls (2003)** mentioned that the diversity indices applied to water quality assessment and the high species richness values related to nutrient enrichment. Furthermore, **Wilhm (1975**) proposed the relationship between Shannon–Weaver index and pollution status.

The different values of the richness index revealed the variety of sites according to impact of pollution. There was a significant difference between the upstream site and other sites due to the impact of pollution **(Bere & Tundisi, 2011)**.

The values of the Shannon–Weaver index on both host plants were <4 and thus indicate a slight pollution **(Wilhm, 1975)**. **Dreslik (1999)** explained that this index relies on the algal species number, their distribution، and their relationship with water pollution, and for these reasons, only a temporal difference is noted for this index, revealing an increase in epiphyton algae in autumn compared to spring.

Hassan and Shaawiat (2015) reported that, if the value of the Evenness index was <0.5, it represents the homogenization of species. The study results of this index of epiphyton algae were <0.5 on both host plants. **Green (1993**) explained that the low value of Evenness index was an indication of environmental stress.

The low Jaccard similarity index values >0.6 were reflected in the variation of epiphyton growth on the different host plants, in addition to the temporal variation of environmental factors **(Toporowska *et al*., 2008)**. The index values for *P. australis* were higher than the values recorded on *C. demersum*; these results are related to different types of host plants and their architecture for each plant, moreover the allelopathic effect of C. *demersum* against some epiphyton algae **(Gross *et al*., 2003; Toporowska *et al*., 2008; El-Sheekh *et al*., 2010)**. Authors reported the effect of biotic (such as grazing and competition on the resources) and abiotic factors (light, temperature, and nutrient availability) on the values of indices **(Hassan *et al*., 2007)**.

CONCLUSION

this study offers insights into the preference of epiphytic algae for specific hostplants. *P. australis* proved to be favorable habitat for diatoms, where non-diatom algae are attached to *C. demersum*. The complexity of the interaction of environmental and biological factors that coincides with the hydrological state of the river has affected the periphytic algal diversity. The results of the indices reflected the status of river water quality, serving as an indicator for pollution and providing a good tool for water quality assessment. Therefore, we suggest conducting more water quality studies considering Iraqi aquatic ecosystems.

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