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Diversity and Association of Algae and mosses in Nile Delta, Egypt

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ABSTRACT

At Botany and microbiology department, Faculty of Science, Zagazig University, Egypt. Association between mosses and algae, may be affected by several factors such as the specificity of epiphytic relationships, intracellular colonization, and the overall biodiversity of the environment. Among the observed organisms, the alga *Mougeotia* sp. and the moss *Barbula* sp.² demonstrated a notably high relative abundance within the moss-algal community, as indicated by Simpson's diversity index. Likewise, *Chroococcus* sp, alongside *Bryum* sp.² and *Didymodon luridus*, recorded the highest values of species richness based on the Shannon index. The overall association between mosses and algae positively contributes to moss diversity observed in the field. The red brick wall was identified as the most favorable microhabitat supporting moss proliferation, while limestone surfaces, cement walls, and red bricks collectively serve as natural substrates conducive to algal growth in association with mosses. This study offers deeper insight into the ecology of aerial algae and highlights the environmental factors shaping algal and moss community structure and their roles in enhancing soil fertility.

Keywords: Association Algae, Chlorophyta, Bryophyta, Mosses, microhabitat.

Introduction

Bryophytes represent one of the earliest groups of green plants to have successfully colonized terrestrial environments approximately 470–551 million years ago, having evolved from aquatic algal ancestors (Morris et al., 2018). The majority of taxonomic and ecological studies on algae have been conducted in temperate regions, with particular focus on species inhabiting soils, building surfaces, and mountainous terrains (Harload and Schlichting, 1975; Metting, 1981; Broady and Ingerfeld, 1993). Mosses (Musci), although relatively limited in number, possess the ability to establish epiphytic or endophytic associations with cyanobacteria and various algal species. In most cases, cyanobacteria grow epiphytically on mosses, with the notable exception of two Sphagnum species wherein cyanobacteria inhabit specialized water-filled hyaline cells believed to offer protection from the acidic conditions typical of bog ecosystems (Solheim and Zielke, 2002). While associations between mosses (Bryophyta) and cyanobacteria have been observed, they

do not involve specialized symbiotic structures analogous to those present in liverworts or hornworts. Instead, these interactions span a continuum ranging from incidental associations to more defined epiphytic relationships and even intracellular colonization of senescent moss cells.

Algae and mosses often coexist in habitats characterized by shared environmental preferences, most notably high moisture levels, as both groups thrive in damp and shaded conditions. These unique and often overlooked organisms play vital roles across diverse ecosystems on every continent (**Von Konrat et al., 2010**). The use of spore traps and culture techniques has revealed that cyanobacterial propagules are frequently produced from a variety of vegetation types—including mosses and lichens—particularly at the end of the growing season as vegetation begins to desiccate. Dispersal mechanisms for these propagules include both wind and meltwater streams (**Broady, 1979**).

Despite these observations, our understanding of how such associations are initiated and maintained remains limited. Critical knowledge gaps persist regarding the mechanisms by which cyanobacteria colonize moss tissues, the potential involvement of specific genes, and the nature of any nutrient exchange between the symbiotic partners. In this study, we employ the terms "epiphytic association" and "intracellular colonization" to describe these interactions between mosses and cyanobacteria. The primary objectives of the present research are to investigate the factors that enable certain mosses to host greater populations of nitrogen-fixing cyanobacteria compared to others inhabiting similar environments, and to characterize the diversity of the associated cyanobacterial taxa.

Materials and Methods

Moss specimens were randomly collected from the provinces of El-Daqahlia and El-Sharkyia in Egypt (winter 2019-autum 2020), and subsequently prepared for further analysis. Identification of moss species was carried out based on the references of **Abo-Salama (1985)**, **Wilson (1990)**, and **Andrea et al. (2011)**. Associated algal taxa were identified according to **Bellinger and Sige (2015)** and **John et al. (2011)**. Diversity indices, including Simpson's and Shannon's, were applied to both mosses and associated algal taxa to determine dominance and richness **Tuomisto, (2010)**.

Growth media for the cultivation of algal species included both liquid and semi-solid forms of Blue Green, Watanabe, and Navicula (NAV) media, following the protocols of **Watanabe and Nozaki, (1994)**.; **El-Nawawy et al. (1958)**; **Stanier et al. (1971)**, and **Starr & Zeikus (1993)**. Chemical characteristics of soil as ; Total Phosphorus, Nitrogen, Potassium, sodium and soil PH were determined according to **Abu. Ziada, (2014)**. .Data analysis, including correlation matrix construction, was conducted using Microsoft Excel 2010 to determine the relationship between environmental parameters and the association between algae and mosses.

Results

As diversity and associations were affected to each other, each moss taxa could be grow better in a certain habitat in which environmental favorable are present. Table (1) below explains the relation between each studied moss taxa to four microhabitats if presence or absence which lately reflect the most favorable habitat for each muss.

Table 1. Diversity of the collected mosses species in different Microhabitats

Mosses species	Microhabitats				Elevation Range in Feet.
	Lime Stone Wall	Cement Wall	Red brick wall	Clay Soil Or wall	
1. <i>Barbula convolute</i>	-	+	+	-	19-21
2. <i>Barbula ehrenbergii</i>	-	+	+	-	19
3. <i>Barbula indica</i>	+	+	+	-	10-17
4. <i>Barbula javanica</i>	+	+	+	-	13-35
5. <i>Barbula</i> sp. ₁	+	+	+	-	12-14
6. <i>Barbula</i> sp. ₂	+	+	+	+	15-35
7. <i>Barbula</i> sp. ₃	+	+	+	-	10
8. <i>Bryoerthrophyllum recurvirostre</i>	+	+	+	-	17-30
9. <i>Bryum bicolor</i>	+	+	+	-	10-24
10. <i>Bryum kunzei</i>	+	+	+	-	15-38
11. <i>Bryum</i> sp. ₁	+	+	+	+	15-25
12. <i>Bryum</i> sp. ₂	+	+	+	+	13-21
13. <i>Bryum</i> sp. ₃	+	+	+	+	13-20
14. <i>Didymodon luridus</i>	+	+	+	+	9-25
15. <i>Didymodon tophaceus</i> var. <i>tophaceus</i>	-	+	+	-	23
16. <i>Didymodon vinealis</i>	+	+	+	-	17-21
17. <i>Eucladium</i> sp.	-	+	-	-	14
18. <i>Fissidens</i> sp.	+	+	+	-	10-25
19. <i>Funaria hygrometrica</i>	+	+	+	+	7-19
20. <i>Funaria</i> sp. ₁	-	+	+	+	6-14
<i>Imbribryum alpinum</i>	+	+	+	-	13-16
21. <i>Philonotis evanidinervis</i>	+	-	+	-	11
22. <i>Philonotis hastate</i>	+	+	+	-	18-21
23. <i>Philonotis obtusata</i>	+	-	+	-	21-27
24. <i>Splachnobryum obtusm</i>	+	+	+	-	19-30
25. <i>Trichostomum</i> sp. ₁	-	+	+	-	20
26. <i>Trichostomum</i> sp. ₂	-	+	+	-	13-17

Red brick walls were identified as the most prevalent microhabitat, supporting 26 of the 27 recorded moss species. In contrast, clay soil or wall habitats supported only 7 species, making it the least favorable habitat for growth. Mosses such as *Barbula* sp.₂, *Bryum* sp., *Didymodon luridus*, and *Funaria hygrometrica* were found in multiple microhabitats, while others like *Eucladium* sp. and *Fissidens* sp. were restricted to cement walls. *Philonotis* species preferred limestone habitats, whereas *Trichostomum* adapted on cement and brick wall habitats. According to elevation range the genus *Philonotis* grows in high elevation (21-27 feet.) and *Funaria* spp. grow in lowest elevation (6-14 feet.).

Table 2. Chemical analysis of the soil of some mosses taxa:

Sample No.	Collection No.	Moss sample	Total phosphorous (mg/L)	Total nitrogen (mg/L)	K ⁺ (ppm)	Na ⁺ (ppm)
7	146	<i>Barbula javanica</i>	395.67	3.05	2.1	0.037
2	14	<i>Barbula sp₁</i>	467.20	20.48	1.8	0.019
5	132	<i>Barbula sp₁</i>	310.16	0.48	2.4	0.033
6	145	<i>Barbula sp₂</i>	854.50	16.81	1.7	0.134
8	179	<i>Barbula sp₂</i>	419.12	16.99	8.3	0.197
3	22	<i>Bryum sp₂</i>	779.50	13.69	2.1	0.118
1	13	<i>Didymodon luridus</i>	396.70	0.07	1.5	0.023
4	112	<i>Philonotis hastate</i>	265.80	34.42	3.4	0.017

Table 2 showing the result of soil analysis of some mosses taxa recorded in different localities. The Soil sample of *Barbula javanica* contains 3.05 mg\L of Total nitrogen, 395.67 mg\L of total phosphours, 2.1 ppm of Potasium and 0.037 ppm of Sodium. This sample was collected from Mit Ghamr - Al Dakahlia Governorate, On a cement wall of water besin - shaded area, at latitudes of 00 '45 030" north 18 '17 031" east and 10 F above the sea level. The soil sample of *Barbula sp₁* having a collection number of (14) contains 20.48 mg\L of Nitrogen, 467.20 mg\L of total phosphours, 1.8 ppm of Potasium and 0.019 ppm of Sodium. This sample was collected from Al Zagazig - Al-Sharkia Governorate On a cement wall of water besin in a sunny area, at latitudes of 19.57 '30 035 north " 31.19 '31 032" east and 12 F above the sea level. The soil sample of *Barbula sp₁* having a collection number of (132) contains 0.48 mg\L of total Nitrogen, 310.16 mg\L of total phosphours, 2.4 ppm of Potassium and 0.033 ppm of Sodium. This sample was collected from Mit Ghamr - Al Dakahlia Governorate, On a lime stone besides of a water canal in a shaded area, at latitudes of 10 '43 030 "north 58 '16 031" east and 19 F above the sea level. The Soil sample of *Barbula sp₂* having a collection number of (145) contains 16.81 mg\L of Nitrogen 854.50 mg\L of total phosphours, 1.7 ppm of Potasium and 0.134 ppm of Sodium . This sample was collected from Mit Ghamr - Al Dakahlia Governorate, On a cement wall of water besin in a shaded area, at latitudes of 00 '45 0 30" north 18 '17 031" east and 10 F above the sea level. The soil sample of *Barbula sp₂* having a collection number of (179) contains 16.99 mg\L of Nitrogen, 419.12 mg\L of total phosphours, 8.3 ppm of Potasium and 0.197 ppm of Sodium. This sample was collected from Sandub - Al Dakahlia Governorate, on a wall of water canal from inside in a shaded area, at latitudes of "49 '42 030 north "08 '17 031 east and 13 F above the sea level. The soil sample of *Bryum sp₂* contains 13.69 mg\L of Nitrogen 779.50 mg\L of total phosphours, 2.1 ppm of Potasium and 0.118 ppm of Sodium. This sample was collected from Bilbeis - Al-sharkia Governorate, On a red brick wall of water besin- shaded area, at latitudes of "42.75 '30 029" north 33.26 '31032 east and 11 F above the sea level. The soil sample of *Didymodon luridus* contains 0.07 mg\L of Nitrogen 396.70 mg\L of total phosphours, 1.5 ppm of Potasium and 0.023 ppm of Sodium. This sample was collected from Mit Ghamr - Al Dakahlia Governorate , on cement wall in a shaded area, at "45.70 '30 043 north "13.55 '31021east and 7 F above the sea level. The soil sample of *Philonotis hastate* contains 34.42 of Nitrogen 265.80 mg\L of total phosphours, 3.4 ppm of Potasium and 0.017 ppm of Sodium. This sample was collected from Mit Ghamr- AL Dakahlia Governorate, on a red brick wall of water besin in a shaded area , at latitudes of 10'43030" north "47 '16 037 east and 18 F above the sea level. According to table 2 *Barbula sp₂* soil have the highest Total phosphorous, sodium and potassium and the lowest content over all the other soils and according to tables 17,18 and 19 *Barbula sp₂* were associated to *Scandesmus sp.* and *Chrococcus sp.* in navicula, in watanabe media were associated to *Nostoc sp.* and *Mougeotia sp.* and in blue green media were associated to *Chrococcus sp.* *Didymodon luridus* soil have the highest lithium content and the lowest total nitrogen and potassium contents over all the other soils and according to tables 17,18 and 19 *Didymodon luridus* were associated to

Chroococcus sp. and *Oscillatoria sp.* in navicula, in watanabe media were associated to *Chlorella* and *Oscillatoria* and in blue green media were associated to *Chroococcus sp.* and *Oscillatoria*. *Philonotis hastate* soil have the highest content of total nitrogen and the lowest Total phosphorous, sodium and lithium content and according to tables 17,18 and 19 *Philonotis hastate* were associated to *Nostoc sp.* in watanabe media.

The main aim of a correlation matrix is to summarize and visualize the strength and direction of an relationships between multiple variables in a dataset.

Table 3. Correlation matrix between Total Phosphorus, Nitrogen, K, Na, Ph. Value, Elevation, Richness, Total phytoplankton and Mosses individuals

	Total Phosphorus	Total nitrogen	K	Na	PH Value	Elevation	Richness	Total phytoplankton	Mosses individuals
Total Phosphorus	1								
Total nitrogen	0.058893	1							
Potassium	-0.49531	0.592033	1						
Sodium	0.949896	0.034222	-0.30638	1					
PH Value	0.349351	-0.38789	-0.85129	0.130898	1				
Elevation	-0.48841	0.3524	0.811303	-0.30727	-0.44921	1			
Richness	-0.02328	0.074651	-0.45477	-0.32026	0.679189	-0.32341	1		
Total phytoplankton	-0.14043	-0.37741	-0.51798	-0.28848	0.368948	-0.58421	0.45807	1	
Mosses individuals	-0.35003	0.762799	0.8789	-0.23715	-0.81794	0.515898	-0.28865	-0.21157	1

Correlation analysis indicated that elevation had a strong positive correlation with the number of individuals, while richness exhibited a strong positive correlation with total phytoplankton counts. These findings highlight the environmental variables most influential in enhancing moss-algae associations.

The aims of the Shannon and Simpson's Diversity indices are to quantify biodiversity within a community by measuring both number of species) and distribution of individuals among species. The Shannon index provides a high-level overview, emphasizing both richness and evenness, while the Simpson index focuses more on the dominance of common species and is less sensitive to rare species.

Table 4. Dominance and Association between Mosses and algae

Algal Taxa	Dominant related mosses Taxa	Number of individuals	Shannon's Diversity (H)	Simpson's Diversity (D)
<i>Anaphaxapsa</i> sp.	<i>Bryum</i> sp ₂	54	0.187	4.91×10 ⁻⁰³
<i>Chrococcus</i> sp.	<i>Barbula</i> sp. ₂ , <i>Barbula javanica</i> , <i>Barbula</i> sp. ₁ , <i>Didymodon luridus</i> , <i>Bryum</i> sp ₃ , <i>Funaria hygrometrica</i> , <i>Barbula</i> sp. ₂ and <i>Didymodon luridus</i>	544.4	0.241	0.507
<i>Oscillatoria</i> sp.	<i>Bryum</i> sp. ₂ , <i>Didymodon luridus</i> , <i>Didymodon luridus</i> and <i>Didymodon luridus</i>	3.32	0.024	1.32×10 ⁻⁰⁵
<i>Anabaena</i> sp.	<i>Funaria hygrometrica</i> and <i>Bryum</i> sp. ₃	6	0.038	5.15×10 ⁻⁰⁵
<i>Nostoc</i> sp.	<i>Barbula</i> sp. ₁ , <i>Barbula</i> sp. ₂ and <i>Philonotis hastate</i>	60.88	0.202	6.25×10 ⁻⁰³
<i>Chlorella</i> sp.	<i>Didymodon luridus</i>	81.4	0.239	0.011
<i>Scenedsmus</i> sp.	<i>Barbula</i> sp.	6.2	0.039	5.53×10 ⁻⁰⁵
<i>Mougeotia</i> sp.	<i>Barbula</i> sp. ₂	0.2	2.159	2.74×10 ⁻⁰⁷

Simpson's index highlighted the highest relative abundance in the association between *Mougeotia* sp. and *Barbula* sp.₂. Shannon's index identified the greatest species richness in associations involving *Chrococcus* sp. with mosses such as *Bryum* sp.₂, *Didymodon luridus*, and *Funaria hygrometrica*.

Discussion

The leaf morphology of mosses plays a critical role in maintaining high humidity around cyanobacteria, thereby supporting extended growth periods during warmer seasons. Cyanobacteria were typically observed on the inner surfaces of moss leaves or nestled between leaf and stem, while outward-facing surfaces were not colonized. (Solheim, and Zielke, 2002.).

The structural features of *Barbula* and *Bryum* leaves, particularly their groove formations, appear well-suited for cyanobacterial colonization, potentially offering protection from environmental stress. However, contrary to the hyaline cells of *Sphagnum* mosses (Granhall and van Hofsten, 1976) these moss cells seem not to be in contact with living neighboring cells.

Red brick walls were identified as the optimal habitat for moss-algae associations, possibly due to their nutrient composition favoring the growth of both groups. Hallingback and Hotsgets 2000; Gignac 2001 and Andreae et al., 2011). Watanabe medium, owing to its nutrient-rich formulation, supported the growth of a wide variety of cyanobacteria and green algae. Additionally, environmental factors such as elevation and nutrient availability (e.g., N, P, K) significantly influenced species richness and the prevalence of phytoplankton. (Thompson et al., 1988, Andersen et al., 1991, Rippka & Herdman, 1992, Starr & Zeikus, 1993, Watanabe & Nozaki, 1994).

Diversity indices provide insights into the distribution and commonness of species within a community. High Simpson values indicate evenness in distribution, while high Shannon values reflect species richness. Cyanobacteria associated with live moss tissues were observed to exhibit higher nitrogen-fixing activity compared to those associated with dead tissues. Certain moss species, such as *Barbula* and *Bryum*, form tight tubular leaf structures that encapsulate cyanobacteria, enhancing their protection (Hallingback and Tan 2010; Scheirer and Brasell, 1984) .

The dispersal of cyanobacterial propagules is facilitated primarily by wind and water, with the exception of some species like *Nodularia* sp. *Nostoc*-type cyanobacteria were the most commonly observed in association with mosses. Additional genera, including *Microchaete*, *Gloeocapsa*, and *Asterocapsa*, along with heterotrophic bacteria and fungi, were also detected on moss surfaces. Epiphytic green algae, notably *Mougeotia* sp. and *Chlorella*, were present on moss species such as *Bryum* and *Didymodon*. (Duckett, *et al.* 1977; Vincent, 2000).

Cyanobacteria play a central role as primary nitrogen fixers and phototrophs in many terrestrial ecosystems. They can exist freely or in symbiosis with bryophytes. Studies have confirmed that cyanobacteria in moss associations are crucial for nitrogen input into vegetated soils, thereby enhancing fertility. This mutualistic interaction is especially well-documented in species like *Sphagnum*, which possess empty hyaline cells facilitating colonization. The presence of cyanobacteria in moss leaf axils are documented by Lubos, (2000).

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