HTTPS://DOI.ORG/ 10.5281/zenodo.23214458



Purposeful scientific research as a pillar of sustainable development



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

Gamal M. A. Lashin[1], Yassin M. AL-Ayoti, Mohamed F. Azzazy[2], Gehad A. Hammouda and Nourhan O. A. Zaid

[1] *Botany & Microbiology Department, Faculty of Science, Zagazig University, Egypt.*

[2]*Surveys of Natural Resources Department, Environmental Studies and Research Institute University of Sadat City, Egypt.*

gslashin@zu.edu.eg

ABSTRACT

At Botany and microbiology department, Faculty of Scince, 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, *Chrococcus 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, hlorophyta, 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 Sigee (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), a**nd **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.	*Barbula convolute*	-	+	+	-	19-21
2.	*Barbula ehrenbergii*	-	+	+	-	19
3.	*Barbula indica*	+	+	+	-	10-17
4.	*Barbula javanica*	+	+	+	-	13-35
5.	*Barbula* sp.1	+	+	+	-	12-14
6.	*Barbula* sp,2	+	+	+	+	15-35
7.	*Barbula* sp,3	+	+	+	-	10
8.	*Bryoerthrophyllum recurvirostre*	+	+	+	-	17-30
9.	*Bryum bicolor*	+	+	+	-	10-24
10.	*Bryum kunzei*	+	+	+	-	15-38
11.	*Bryum* sp,1	+	+	+	+	15-25
12.	*Bryum* sp,2	+	+	+	+	13-21
13.	*Bryum* sp,3	+	+	+	+	13-20
14.	*Didymodon luridus*	+	+	+	+	9-25
15.	*Didymodon tophaceus var. tophaceus*	-	+	+	-	23
16.	*Didymodon vinealis*	+	+	+	-	17-21
17.	*Eucladium* sp.	-	+	-	-	14
18.	*Fissidens* sp.	+	+	+	-	10-25
19.	*Funaria hygrometrica*	+	+	+	+	7-19
20.	*Funaria* sp.1	-	+	+	+	6-14
Imbribryum alpinum		+	+	+	-	13-16
21.	*Philonotis evanidinervis*	+	-	+	-	11
22.	*Philonotis hastate*	+	+	+	-	18-21
23.	*Philonotis obtusata*	+	-	+	-	21-27
24.	*Splachnobryum obtusm*	+	+	+	-	19-30
25.	*Trichostomum* sp.1	-	+	+	-	20
26.	*Trichostomum* 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 ygrometrica* ere 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	*Barbula javanica*	395.67	3.05	2.1	0.037
2	14	*Barbula sp1*	467.20	20.48	1.8	0.019
5	132	*Barbula sp1*	310.16	0.48	2.4	0.033
6	145	*Barbula sp2*	854.50	16.81	1.7	0.134
8	179	*Barbula sp2*	419.12	16.99	8.3	0.197
3	22	*Bryum sp2*	779.50	13.69	2.1	0.118
1	13	*Didymodon luridus*	396.70	0.07	1.5	0.023
4	112	*Philonotis hastate*	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

*Chrococcus sp.*and *Oscillatoria sp.* in navicula, in watanabe media were associated to *Chlorella* and *Oscillatoria* and in blue green media were associated to *Chrococcus 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 0f individuals	Shannon's Diversity (H)	Simpson's Diversity (D)
Anaphaxapsa sp.	*Bryum sp2*	54	0.187	4.91×10^{-03}
Chrococcus sp.	*Barbula* sp.2, *Barbula javanica*, *Barbula* sp.1, *Didymodon luridus*, *Bryum sp3*, *Funaria hygrometrica* , *Barbula* sp.2 and *Didymodon luridus*	544.4	0.241	0.507
Oscillatoria sp.	*Bryum* sp.2, *Didymodon luridus*, *Didymodon luridus* and *Didymodon luridus*	3.32	0.024	1.32×10^{-05}
Anabaena sp.	*Funaria hygrometrica* and *Bryum* sp.3	6	0.038	5.15×10^{-05}
Nostoc sp.	*Barbula sp1, Barbula sp2* and *Philonotis hastate*	60.88	0.202	6.25×10^{-03}
Chlorella sp.	*Didymodon luridus*	81.4	0.239	0.011
Scenedsmus sp.	*Barbula* sp.	6.2	0.039	5.53×10^{-05}
Mougeotia sp.	*Barbula* 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 Andrea*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).**

References

Abo-Salama U. Y., (1985). Morphological studies on some Egyptian Bryophytes in Southern Sinai. M. Sc. Thesis, Ain Shams University. 162 p.

Abu-Ziada., 2014. Practical Notes On Soil, Water and Plant Analysis. Botany Department Faculty of science.Mansoura Uni.38pp.

Andrea G. A., Lalalne G. S. and Melanie P. P. (2011). Some Medicinal Bryophytes: their Ethnobotanical Uses and Morphology/ Central Mindanao University, Musuan, Bukidnon, Philippines. Asian Journal of Biodiversity Art. #92, pp.49-80Vol.2December2011.

Andersen, R.A.; Jacobson, D.M. and Sexton, J.P. (1991). Provasoli-Guillard Center for Culture of Marine Phytoplankton. Catalogue of Strains. 98pp. West Boothbay Harbor, Maine, USA, 1991.

Bellinger, E. G. and D. C. Sigee. (2015). Freshwater Algae, Second Edition. Identification, Enumeration, and Use as Bioindicators. John Wiley & Sons, LTD., West Sussex, UK.

Broady, P.A. 1979. The Signy Islands terrestrial reference sites: IX. The ecology of the algae of site 2, a moss carpet. *British Antarctic Survey Bulletin* 47: 13-29.

Broady, P. A. and Ingerfeld, M. 1993. Three new species and a new record of Chaetophoracean (Chlorophyta) algae from terrestrial habitats in Antartica. *European Journal of Phycology*. 28:

Duckett, J.G., Prasad, A.K.S.K., Davies, D.A., and Walker, S. 1977. A cytological analysis of the Nostoc-bryophyte relationship. *New Phytologist* 79: 349-362.

Gignac, L. D. (2001). New frontiers in bryology and lichenology: Bryophytes as indicators of climate change. The Bryologist 104: 410-420.

Granhall, U. and van Hofsten, A. 1976. Nitrogenase activity in relation to intracellular organisms in *Sphagnum* mosses. *Physiologia Planiarum* 36: 88-94.

Hallingback T. and Hodgets N. (2000). Status survey and conservation action plan for bryophytes: mosses, liverworts and hornworts. IUCN/SSC Bryophyte Specialist Group. IUCN, Gland, Switzerland and Cambridge, U.K. 106 pp.

Hallingback, T. and Tan B.C. (2010) Past and present activities and future strategy of bryophytes conservation. Phytotaxa 9: 266-274.

Harlod, E. and Schlichting, Jr. 1975. Some subaerial algae from Ireland. *British Phycological Journal.* 10. Lemes-Da-Silva. N. M., Zanini Branco, L. H. and Necchi-

John, D. M., Whitton B. A., and Brook A. J. (2011). The Freshwater Algal Flora of the British Isles: An Identification Guide to Freshwater and Terre.

Lubos, L. C. (2000). Taxonomy, species richness and distribution of mosses in selected mountains in Mindanao. Dissertation. Central Mindanao University.

Metting, B. 1981. The systematics and ecology of soil algae. *The Botanical Review.* 47. Mikter, S. S. a

Morris J.L., Puttick M.N., Clark, J.W., EdwardsD., Kenrick P. Pressel S., Wellman, C.H, Yang Z., Schneider H. and Don oghue P. C. J. (2018). The timescale of early land plant evolution. Proc. Natl. Acad. Sci. USA 115: E2274–E2283. doi:10.1073/pnas.1719588115.

Rippka, R. and Herdman, M. (1992). Pasteur Culture Collection of Cyanobacterial Strains in Axenic Culture. Vol.1, Catalogue. of strains. 103pp. Institute Pasteur, Paris, France, 1992.

Scheirer, D.C. and Brasell, H.M. 1984. Epifluorescence microscopy for the study of nitrogen fixing blue-green algae associated with *Funaria hygrometrica* (Bryophyta). *American Journal of Botany* 71: 461-465.

Solheim, B., Endal, A., and Vigstad, H. 1996. Nitrogen fixation in arctic vegetation and soils from Svalbard, Norway. *Polar Biology* 16: 35-40.

Solheim, B. and Zielke, M. 2002. Associations between cyanobacteria and mosses. In: *Cyanobacteria in Symbiosis.* B. Bergman, AN. Rai, and U. Rasmussen, eds. B. Kluwer Academic Publishers, Dordrecht, pp. 137-152.

Starr, R.C. and Zeikus, J.A. (1993). UTEX – The Culture Collection of Algae at the University of Texas at Austin. J. Phycol. Suppl. 29 (1993).

Thompson, A.S., Rhodes, J.C. and Pettman, I.. (1988). Culture Collection of Algae and Protozoa. Catalogue of strains. 164pp., Natural Environment Research and Council, England, 5th edit., 1988.

Tuomisto, H (2010). "A diversity of beta diversities: straightening up a concept gone awry. Part 1. Defining beta diversity as a function of alpha and gamma diversity". Ecography. 33 (1): 2–22. Bibcode:2010Ecogr..33....2T. doi:10.1111/j.1600-0587.2009.05880.x

Vincent, W.F. 2000. Cyanobacterial dominance in the polar regions. In: *The Ecology of Cyanobacteria. Their Diversity in Time and Space.* B.A. Witton, and M. Potts, eds. Kluwer Academic Publishers, Dordrecht, pp. 321-340.

Von-Konrat M., Shaw A. J. and Renzaglia, K.S. (2010). A special issue of Phytotaxa dedicated to Bryophytes: the closest living relatives of early land plants. Phytotaxa 9: 5–10.

Watanabe M.M. and Nozaki, H. (1994). NIES-Collection. List of strains, microalgae and protozoa. 4th edit., 127pp. The National Institute for Environmental Studies, Japan, 1994.

Wilson, P. (1990). How to make thin free section, Evansia, 7 : 16.