Recoletos Multidisciplinary Research Journal Vol. 13 no. 1 (June 2025) DOI: <u>https://doi.org/10.32871/rmrj2513.01.12</u>



Orginal Article

# New Records of Subaerial Epilithic Algae from Elevated Rock Surfaces in Mt. Makiling Forest Reserve Laguna, Philippines

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#### Abstract

**Background:** Subaerial microalgal communities, particularly those inhabiting elevated rock surfaces, remain understudied in the Philippines. This study presents the first floristic survey of microalgae from such unique habitats within the Mt. Makiling Forest Reserve.

**Methods:** Rock surface samples were collected from various elevations in the reserve and subjected to morphological examination using light microscopy. Species were identified using standard morphological techniques and taxonomic references.

**Results:** A total of 18 species of microalgae were described taxonomically, of which seven species belonged to Cyanophyceae, five species to Bacillariophyceae, three species to Chlorophyceae, two species to Trebouxiophyceae, and one species to Mediophyceae. Notably, Synechococcus elongatus (Nägeli) Nägeli, a rare cyanobacterium, and Stephanocyclus meneghinianus (Kützing) Kulikovskiy, Genkal & Kociolek, a diatom, are reported for the first time in the Philippines. All documented taxa represent new distributional records for this habitat type. This survey significantly expands the known diversity of subaerial microalgae in the country and underscores the ecological importance of rock surface habitats as reservoirs of algal biodiversity.

**Conclusion:** These findings provide critical taxonomic and biogeographic data that can support further ecological and conservation studies.

Keywords

Microalgae, Philippines, subaerial habitat, taxonomy, terrestrial, tropical

# INTRODUCTION

Subaerial algae are common yet underexplored inhabitants of solid substrates across terrestrial ecosystems, often forming gelatinous, greenish, or dark patches on exposed rock, bark, or soil surfaces. These algae absorb water and minerals directly from substrates and survive extreme desiccation, making them well-adapted to fluctuating environmental conditions (Arguelles, 2019a, 2019b, 2019c; Bhakta et al., 2014). Their diversity and distribution are shaped by both anthropogenic and natural factors such as substrate type, vegetation cover, topography, and climate (Nováková & Neustupa, 2015; Sophia et al., 2016). Diatoms, green algae, and cyanobacteria are among the most commonly reported taxa in subaerial habitats due to their ecological plasticity and adaptive traits (Davis & Karpook, 1999; Sophia et al., 2016).



Despite growing global interest, studies on subaerial algal communities remain limited—especially in tropical regions. Much of the existing literature focuses on temperate regions, with algae surveyed from tree bark, historic monuments, and stone surfaces (Nienow, 1996; Nováková & Neustupa, 2015; Sophia et al., 2016). In the Philippines, published work on subaerial algae is extremely scarce, with only one corticolous algal survey previously reported in Mt. Makiling Forest Reserve (Arguelles, 2019c). Leaving a significant gap in our understanding of the diversity, ecology, and distribution of subaerial algae in the country, particularly from ecologically rich forested habitats.

Mt. Makiling Forest Reserve, a designated ASEAN Heritage Park, is one of the most biologically diverse forest ecosystems in the Philippines, known for its complex terrain and rich assemblage of endemic flora and fauna. While numerous biodiversity assessments have focused on the area's plants and animals (Gonzalez et al., 2020), microalgal diversity—especially that of subaerial habitats—remains unstudied. This study addresses this knowledge gap by conducting the first taxonomic survey of subaerial epilithic microalgae from elevated rock surfaces in Mt. Makiling. It contributes novel baseline data to the national algal species inventory, enriches our understanding of regional biogeographic patterns, and lays the groundwork for future comparative phycological and ecological research in tropical forest environments.

## **METHODS**

#### Study Site and Sampling of Subaerial Epilithic Algae

The sampling was conducted in August 2020 within the Mt. Makiling Forest Reserve (14°08'N, 121°14'E), a protected area in Luzon Island, Philippines, known for its rich biodiversity and complex geological formations (Figures 1 and 2). Specifically, the focus was on subaerial epilithic algae growing on elevated limestone outcrops. A total of 36 samples from exposed rock surfaces found within forested zones of the reserve. The study selected two randomly chosen elevated limestone rocks in each designated sampling area. Six distinct rock surface areas were sampled from each rock, resulting in a representative collection across microhabitats. The sampling height was standardized at 150–160 cm above the soil, minimizing contamination from soil splash and ensuring the algal community was exclusively subaerial.

Fragments of rock with visible algal biofilms were chipped off carefully using a sterilized geological hammer. These biofilm-bearing rock fragments were immediately placed into sterile Tarson conical tubes to prevent contamination. Samples were kept in a cooler box during field collection to maintain integrity and transported promptly to the laboratory for further analysis. This handling protocol ensured minimal disturbance to the algal cells and preserved their morphological features for taxonomic observation. All safety and sterilization procedures were followed to prevent cross-contamination between sites and preserve the authenticity of each sample.

#### **Microscopic Observation and Taxonomic Identification**

In the laboratory, algal specimens were first observed under an Olympus CX31 binocular microscope equipped with an Infinity X digital camera, allowing high-resolution imaging for initial morphological assessment (Arguelles, 2019c; Arguelles & Monsalud, 2018). A subset of the samples was subjected to chemical digestion following standard protocols described by Round et al. (1990) to prepare clean frustules for diatom identification. All processed and unprocessed specimens were deposited at the Philippine National Collection of Microorganisms (PNCM) for long-term preservation and future reference. Despite being collected in 2020, the specimens were maintained under appropriate preservation conditions, ensuring taxonomic integrity and reproducibility of subsequent analyses.

Importantly, this study represents the first documented effort to characterize the diversity of microalgae, particularly subaerial epilithic taxa, from Mt. Makiling's rock surfaces. This lack of prior records emphasizes the novelty and ecological significance of the survey, providing a critical baseline for future ecological, taxonomic, and conservation-related studies in tropical subaerial microhabitats.



## **Taxonomic Identification and Enumeration**

Identification of each microalgal taxa was done up to the lowest practical unit using all possible and accessible taxonomic monographs, literature, and information (Desikachary, 1959; John & Tsarenko, 2011; Martinez, 1984; Prescott, 1962; Velasquez, 1962; Wehr & Sheath, 2003; Zafaralla, 1998). The morphological features used in microalgae identification, such as color and the size and shape of specialized and vegetative cells; features of the sheath and cellular envelopes; cellular constrictions and sheath; trichome characteristics and cellular filaments, and cell number in a coenobium were considered in the taxonomic identification of the different microalgal species. The preserved diatom slides (voucher specimens) were kept at the Philippine National Collection of Microorganisms (PNCM), BIOTECH-UPLB, College, Laguna, Philippines (Arguelles, 2020b; Arguelles & Monsalud, 2018). The abundance of microalgal species was assessed based on the number of specimens observed per slide, following the five-point scale developed by Wołowski (1998):

- [+] Species observed as a single specimen on at least one slide;
- [1] Species observed as up to six specimens on most slides (sparse);
- [2] Species present on every slide but not in all microscopic fields (frequent);
- [3] Species present in every slide and all visible fields (persistent);
- [4] Species formed visible blooms on the water surface (abundant/in masses).



Figure 1. Location map of Mt. Makiling Forest Reserves in Laguna (Philippines)



Figure 2. Subaerial rock surfaces from sampling areas in Mt. Makiling Forest Reserves



# RESULTS

Overall, 18 subaerial microalgal species were identified in scraped algal samples collected from subaerial rock surfaces found in Mt. Makiling Forest Reserves (Table 1). Of these microalgal taxa, seven species identified belonged to Cyanophyceae, five species to Bacillariophyceae, three species to Chlorophyceae, two species to Trebouxiophyceae, and one species to Mediophyceae. The survey represents five classes, 15 orders, 16 families, 16 genera, and 18 species based on a current microalgal taxonomic system of classification. The survey reports Cyanophyceae (38.89%) and Bacillariophyceae (27.78%) as the major groups of microalgae in the subaerial rock surfaces found in Mt. Makiling Forest Reserves followed by Chlorophyceae (16.67%), Trebouxiophyceae (11.11%), and Mediophyceae (5.56%). These algal species are documented for the first time in this unique habitat and regarded as new distributional records for the Philippines. Taxonomic literature used in microalgae identification of all described species are listed under the morpho-taxonomic description of each taxon. In addition, currently accepted taxonomic names based on Algaebase (Guiry & Guiry, 2022) are used in this floristic survey.

## **Dichotomous Key**

1. Cells with a typical nucleus and organelles bounded by membranes21. Cells lacking a typical nucleus and pigments scattered in the periphery of the cell12
2. Cells characterized with cell walls made of silica32. Cells characterized with cell walls made of cellulose8
<ul><li>3. Valves are elongated and linear</li></ul>
<ul><li>4. Valves are linear-lanceolate</li></ul>
<ol> <li>5. Valves are lanceolate with broadly rounded rostrate apices</li></ol>
6. Valves linear and lanceolate; apices broadly rounded; 14.5–20.5 μm long, 4.0–4.5 μm wide Nitzschia palea 6. Valves linear-lanceolate with subcapitate or rostrate apices; 14.0–49.0 μm long, 3.5–5.0 μm wide Fragilaria vaucheriae
<ul> <li>7. Valves are elliptic or linear-elliptic and flat</li></ul>
8. Cells are fusiform or sickle-shaped
<ol> <li>9. Cells are sickle-shaped or cylindrical</li></ol>
10. Cells are coccoidal or spherical and capable of forming sarcinoid colonies <i>Desmococcus olivaceous</i> 10. Cells are coccoidal or spherical and not capable of forming sarcinoid colonies
11. Cells are 9.0 – 16.0 μm in diameter
12. Cells organized into filaments or trichomes, either solitary or grouped
13. Filaments without heterocyte       14         13. Filaments with heterocytes       Nostoc sp.



14. Filaments are straight or bend slightly (sometimes) towards the anterior end cells, with slightly constructe crosswalls
14. Filaments are short, isopolar, and without a sheath; cross walls are constricted and not attenuated Pseudanabaena galea
15. Cells are spherical or hemispherical in shape 15. Cells are cylindrical or long oval in shape
16. Colonies usually exist in groups of 2–4 cells, with a thick mucilaginous sheath covering the entire ce
16. Cells are spherical, usually in groups of 2-4 cells confined in a thin mucilage Chroococcus minut
17. Cells are cylindrical and solitary, sometimes occurring in groups, but do not form mucilaginous coloni
17. Cells are solitary and cylindrical, sometimes occurring in groups with a thin layer of mucilage

Table 1. Occurrence of epilithic microalgae from subaerial rock surfaces found in Mt. Makiling Forest Reserves

Microalgae –	Sampling Sites		
	Site 1	Site 2	Site 3
Stephanocyclus meneghinianus	1	1	-
Diatoma vulgaris	+	+	+
Nitzschia palea	-	-	+
Fragilaria vaucheriae	-	+	-
Cocconeis sp.	2	1	+
Encyonema montanum	1	1	+
Kirchneriella lunaris	+	+	+
Tetradesmus obliquus	1	-	2
Desmococcus olivaceous	3	3	3
Chlorococcum infusionum	3	3	3
Chlorella sp.	3	2	3
Nostoc sp.	4	4	2
Oscillatoria sp.	3	3	3
Pseudanabaena galeata	-	1	2
Chroococcus schizodermaticus	2	2	1
Synechococcus nidulans	2	1	2
Synechococcus elongatus	2	2	1

[-] absent; [+] species occurred only as a single specimen at least on one slide; [1] sparse; [2] frequent; [3] very frequent; [4] in masses

#### **Phylum: BACILLARIOPHYTA**

Class: Medioiophyceae Order: Thalassiosirales Family: Thalassiosiraceae Genus: *Stephanocyclus* Skabitschevsky 1. *Stephanocyclus* meneghinianus (Kützing) Kulikovskiy, Genkal & Kociolek (Figure 3A) Basionym: *Cyclotella meneghiniana* Kützing Valves are radially symmetrical and small with a diameter of 5.0 – 20.0 μm; striae located at the marginal area of the valve surface (6 – 9 rows for every 10 μm); the central area of the valve is wide, flat, smooth



striated by grooves and radially disposed while the marginal striae radiated. Specimen: LUZON, Laguna, Los Baños, (Mt. Makiling Forest Reserve), E. DLR. Arguelles *s.n*. Photomicrograph prepared from the mounted specimen.

(Arguelles, 2021a, p. 190; Arguelles, 2021d, p. 132; Arguelles, 2020b, p. 252; Arguelles, 2019a, p.19; Arguelles & Monsalud, 2018, p. 5; Kulikovskiy et al., 2022, p. 12)

Class: Bacillariophyceae Order: Fragilariales Family: Fragilariaceae Genus: *Fragilaria* Lyngbye

1. Fragilaria vaucheriae (Kützing) J.B.Petersen (Figure 3B)

Valves linear-lanceolate with subcapitate or rostrate ends; 14.0 – 49.0 µm in length and 3.5 – 5.0 µm in width. The central area of the valve is large, quadrangular, and unilateral, which extends to the valve margin; ghost striae are observable. Striae are mostly parallel at the center of the valve while getting slightly radiate as it approaches the posterior and anterior ends, with 11-14 rows of striae for every 10 µm. Specimen: LUZON, Laguna, Los Baños, (Mt. Makiling Forest Reserve), E. DLR. Arguelles *s.n.* Photomicrograph prepared from the mounted specimen.

(Kociolek et al., 2015, p. 685; Wetzel & Ector, 2015, p. 276)

Class: Bacillariophyceae Order: Achnanthales Family: Cocconeidaceae

Genus: Cocconeis Ehrenberg

1. *Cocconeis* sp. (Figure 3C)

Valves are linear-elliptic or elliptic and flat,  $9.5 - 61.0 \mu m$  in length and  $7.0 - 27.5 \mu m$  in width. The raphe is straight and filiform. Striae are radiated (in the surface of the valve) and interrupted by the presence of a hyaline ring near the margin of the valve. Areolae are 15 - 25 in  $10 \mu m$  along a stria.

Specimen: LUZON, Laguna, Los Baños, (Mt. Makiling Forest Reserve), E. DLR. Arguelles *s.n.* Photomicrograph prepared from the mounted specimen.

(Jahn et al., 2009, p. 281; Romero & Jahn, 2013, p.5)

Class: Bacillariophyceae Order: Cymbellales Family: Gomphonemataceae Genus: Encyonema Kützing

1. Encyonema montanum Bahls (Figure 3D)

Synonym: Encyonema montana Bahls

Valves are semicircular with a slightly (weak) convex ventral margin and a strongly arched dorsal margin. The valve length is 16.0-21.5  $\mu$ m, and the width is 7.0-8.0  $\mu$ m. The raphe is filiform. Raphe fissures at the terminal end deflect ventrally, while the proximal raphe ends dorsally deflect. Ventral striae are parallel (weakly radiate), while dorsal striae radiate. The striae number is 11-12 in 10  $\mu$ m.

Specimen: LUZON, Laguna, Los Baños, (Mt. Makiling Forest Reserve), E. DLR. Arguelles *s.n*. Photomicrograph prepared from the mounted specimen.

(Bahls, 2017, p. 22)

Class: Bacillariophyceae Order: Rhabdonematales Family: Tabellariaceae Genus: *Diatoma* Bory

1. Diatoma vulgaris Bory (Figure 3E)

Valves are lanceolate (isopolar and bilaterally symmetrical) with broadly rounded rostrate apices; 9.2-12.5  $\mu$ m in width and 16.0-60.5  $\mu$ m in length; striae are very fine (40.0-51.0 in 10  $\mu$ m); cingulum with few bands (two or up to three types) open at both opposite ends of the frustule.

Specimen: LUZON, Laguna, Los Baños, (Mt. Makiling Forest Reserve), E. DLR. Arguelles *s.n.* Photomicrograph prepared from the mounted specimen.

(Kociolek et al., 2015, p. 691)



Class: Bacillariophyceae Order: Bacillariales Family: Bacillariaceae Genus: *Nitzschia* Hassall 1. *Nitzschia palea* (Kützing) W. Smith (Figure 3F) Basionym: *Synedra palea* Kützing Valves are lanceolate in shape, and linear, rapidly narrowing at the end poles with rostrate or rounded apices. Valves are 14.5 – 20.5 μm in length and 4.0 – 4.5 μm in width. Fibulae are equidistant and distinct with distinguishable striae (10 - 13 rows of striae for every 10 μm). Specimen: LUZON, Laguna, Los Baños, (Mt. Makiling Forest Reserve), E. DLR. Arguelles *s.n.* Photomicrograph prepared from the mounted specimen. (Arguelles, 2021d, p. 132; Arguelles, 2020b, p. 253; Bahls & Luna, 2018, p. 51; Bahls et al., 2018, p. 53) **Phylum: CHLOROPHYTA** Class: Chlorophyceae

Class: Chlorophyceae Order: Sphaeropleales Family: Scenedesmaceae Genus: Tetradesmus G.M. Smith

1. Tetradesmus obliquus (Turpin) M. J. Wynne (Figure 3G)

Synonym: Scenedesmus obliguus Turpin (Kützing)

Basionym: Achnanthes obliqua Turpin

Coenobium is arranged linearly with 4-8 or (less frequently) 10 cells attached adjacent to each other. Cells are fusiform with pointed apices (11.5 - 25.0 µm in length and 6.0 - 8.5 µm in width). The cell wall is without spines (smooth); the parietal chloroplast is present with a pyrenoid.

Specimen: LUZON, Laguna, Los Baños, (Mt. Makiling Forest Reserve), E. DLR. Arguelles s.n. Photomicrograph prepared from the mounted specimen.

(Arguelles, 2019b, p. 820; Arguelles, 2019d, p. 9)

Class: Chlorophyceae

Order: Sphaeropleales

Family: Selenestraceae

Genus: Kirchneriella Schimidle

1. Kirchneriella lunaris (Kirchner) Möbius (Figure 3H)

Basionym: Rhapidium convolatum var. lunare Kirchner

Kirchneriella lunata Schmidle

The colony is composed of 2-8 or up to 64 clusters of cells; cells are sickle-shaped or cylindrical with a length of 8.0–12.0  $\mu$ m and width of 3.0-6.0  $\mu$ m; parietal chloroplasts with 1–4 pyrenoids (per cell) usually observed near the cell wall.

Specimen: LUZON, Laguna, Los Baños, (Mt. Makiling Forest Reserve), E. DLR. Arguelles s.n. Photomicrograph prepared from the mounted specimen.

(Arguelles, 2021b, p. 839; Arguelles, 2020a, p. 593)

Class: Chlorophyceae

Order: Chlamydomonodales

Family: Chlorococcaceae

Genus: Chlorococcum Meneghini

1. Chlorococcum infusionum (Schrank) Meneghini (Figure 3I)

Basionym: Cystococcus humicola Nägeli

Lepra infusionum Schrank

Cells possess a parietal, cup-shaped chloroplast with a single prominent pyrenoid. Cells are solitary or sometimes in clusters of cells (9.0 – 16.0 (up to 30.0)  $\mu$ m in diameter).

Specimen: LUZON, Laguna, Los Baños, (Mt. Makiling Forest Reserve), E. DLR. Arguelles *s.n.* Photomicrograph prepared from the mounted specimen.

(Arguelles, 2021c, p. 175; Pantastico, 1977, p. 76)





**Figure 3.** Photomicrographs of (A) Stephanocyclus meneghinianus (Kützing) Kulikovskiy, Genkal & Kociolek, (B) Fragilaria vaucheriae (Kützing) J.B.Petersen, (C) Cocconeis sp., (D) Encyonema montanum Bahls, (E) Diatoma vulgaris Bory, (F) Nitzschia palea (Kützing) W. Smith, (G) Tetradesmus obliquus (Turpin) M. J. Wynne, (H) Kirchneriella lunaris (Kirchner) Möbius, (I) Chlorococcum infusionum (Schrank) Meneghini. All scale bars = 10 μm unless specified otherwise.



Class: Trebouxiophyceae Order: Prasiolales Family: Prasiolaceae Genus: *Desmococcus* F. Brand 1. *Desmococcus olivaceus* (Persoon ex Archerson) J. R. Laundon (Figure 4A) Basionym: *Lepraria olivacea* Persoon ex Archerson Cells aggregate to form 2-4 celled colonies (sarcinoid colonies) with a diameter of 4.0-8.5 μm. Cells are coccoidal, spherical, or hemispherical, 2.5 - 4.0 μm in diameter. The chloroplast is parietal with a single pyrenoid (sometimes not distinguishable). Specimen: LUZON, Laguna, Los Baños, (Mt. Makiling Forest Reserve), E. DLR. Arguelles *s.n.* Photomicrograph prepared from the mounted specimen. (Arguelles, 2019c, p. 5; Rindi & Guiry, 2003, p. 259) Class: Trebouxiophyceae Order: Chlorellales

Family: Chlorellaceae Genus: Chlorella Beyerinck [Beijerinck]

1. Chlorella sp. (Figure 4B)

Cells are coccoidal with a diameter of 2.5 - 3.5  $\mu$ m. The chloroplast is cup-shaped with a pyrenoid. Cells form 2-4 hemispherical spores (autospores) as a means of reproduction and released by bursting out the cell wall of the mother cell.

Specimen: LUZON, Laguna, Los Baños, (Mt. Makiling Forest Reserve), E. DLR. Arguelles *s.n.* Photomicrograph prepared from the mounted specimen.

(Arguelles, 2019a, p. 19; Prescott, 1962, p. 237)

#### Phylum: CYANOBACTERIA

Class Cyanophyceae Order: Synechococcales Family: Synechococcaceae Genus: *Synechococcus* Nägeli 1. *Synechococcus elongatus* (Nägeli) Nägeli (Figure 4C) Basionym: *Protocococcus elongatus* Nägeli Cells are bluish-green with homogenous protoplast, solitary, cylindrical, or short oval (in shape), and rounded apical ends. Cells are 2.0 – 9.0 μm in length and 1.0 – 3.0 μm in width, occasionally existing in groups of cells and with a thin layer of mucilage. **A new record for the Philippines.** 

Specimen: LUZON, Laguna, Los Baños, (Mt. Makiling Forest Reserve), E. DLR. Arguelles s. n. Photomicrograph prepared from the mounted specimen.

(Czerwik-Marcinkowska & Mrozińska, 2011, p. 219; Roy et al., 2014, p. 7)

**Class Cyanophyceae** 

Order: Synechococcales

Family: Synechococcaceae

Genus: Synechococcus Nägeli

1. Synechococcus nidulans (Pringsheim) Komárek (Figure 4D)

Basionym: Lauterbania nidulans Pringsheim

Cells are solitary and cylindrical, sometimes occurring in groups, but do not form mucilaginous colonies. Cells are 1.0 – 2.0 µm wide, 3.0 – 8.0 µm long with rounded apices and homogenous protoplast.

Specimen: LUZON, Laguna, Los Baños, (Mt. Makiling Forest Reserve), E. DLR. Arguelles *s.n.* Photomicrograph prepared from the mounted specimen.

(Arguelles, 2019d, p. 6; McGregor, 2013, p. 21)



Class Cyanophyceae Order: Chroococcales Family: Chroococcaceae Genus: Chroococcus Nägeli

1. Chroococcus schizodermaticus West (Figure 4E)

Colonies has a diameter of 26.0–40.0  $\mu$ m, which is composed of groups of 2–4 cells. Mucilaginous sheaths are delimited and wide, occupying the whole algal cells with enlarged outer layers. Vegetative cells are bluish-green, or spherical or hemispherical with a diameter of 3.5–8.5  $\mu$ m.

Specimen: LUZON, Laguna, Los Baños, (Mt. Makiling Forest Reserve), E. DLR. Arguelles *s.n*. Photomicrograph prepared from the mounted specimen.

(Arguelles, 2021d, p. 132; Desikachary, 1959, p.103; McGregor, 2013, p. 53; Tavera & Komárek, 1996, p. 527)

Class Cyanophyceae

Order: Chroococcales

Family: Chroococcaceae

Genus: Chroococcus Nägeli

1. Chroococcus minutus (Kützing) Nägeli (Figure 4F)

Basionym: Protococcus minutus Kützing

Cells are spherical or ovoid, bluish-green in color, and bound in a thin mucilaginous sheath. Cells with homogenous protoplasts are usually in groups of 2-4, 6.5–7.5  $\mu$ m in diameter with mucilaginous sheath and 3.5–5.5  $\mu$ m without mucilaginous sheath.

Specimen: LUZON, Laguna, Los Baños, (Mt. Makiling Forest Reserve), E. DLR. Arguelles *s.n.* Photomicrograph prepared from the mounted specimen.

(Arguelles, 2021c, p. 178; Desikachary, 1959, pp. 104-105; Martinez, 1984, p. 31)

Class Cyanophyceae

Order: Oscillatoriales

Family: Oscillatoriaceae

Genus: Oscillatoria Vaucher ex Gomont

1. Oscillatoria sp. (Figure 4G)

Filaments are non-branching, straight or bend (slightly) at the apical end cells, 2.0 – 4.0 µm in width, and slightly constricted crosswalls. Cells bluish-green in color with homogenous protoplasm, 1.0 - 1.5 µm long and 2.5 – 4.0 µm wide; posterior end cell is hemispherical or rounded. Anterior end cells are not attenuated and rounded, not capitate (absence of calyptra); akinetes and heterocytes are not present. Specimen: LUZON, Laguna, Los Baños, (Mt. Makiling Forest Reserve), E. DLR. Arguelles *s.n.* Photomicrograph prepared from the mounted specimen.

(Desikachary, 1959, pp. 204-214; Martinez-Goss et al., 2019, p. 20; Martinez, 1984, p. 65)

Class Cyanophyceae

Order: Synechococcales

Family: Pseudanabaenaceae

Genus: Pseudanabaena Lauterborn

1. Pseudanabaena galeata Böcher (Figure 4H)

Trichomes are short, bluish-green, isopolar, and without a mucilaginous sheath, constricted cross walls, and not attenuated. Cells are cylindrical with 2.5-  $3.0 \mu m$  in length and 0.5- $1.5 \mu m$  in width; terminal cells are rounded.

Specimen: LUZON, Laguna, Los Baños, (Mt. Makiling Forest Reserve), E. DLR. Arguelles *s.n.* Photomicrograph prepared from the mounted specimen.

(Arguelles, 2019c, p. 3; Komárek & Anagnostidis, 2005)

Class Cyanophyceae Order: Nostocales Family: Nostocaceae Genus: *Nostoc* Lauterborn Vaucher ex Bornet & Flahault 1. *Nostoc* sp. (Figure 4I) Filaments with bluish-green cells that are not attenuated in both the apical and posterior end; cells are



barrel or spherical in shape and are usually shorter than broad (6.0-7.5 μm long); constricted crosswalls are observable in the trichomes; heterocytes are spherical in shape with a diameter of 7.0-8.0 μm. Specimen: LUZON, Laguna, Los Baños, (Mt. Makiling Forest Reserve), E. DLR. Arguelles *s.n.* Photomicrograph prepared from the mounted specimen.

(Arguelles, 2021b, p. 836; Arguelles & Monsalud, 2018, p. 31; Desikachary, 1959, p. 387; Martinez, 1984, p. 52; Velasquez, 1962, p. 342)



**Figure 4.** Photomicrographs of (A) Desmococcus olivaceus (Persoon ex Archerson) J. R. Laundon, (B) Chlorella sp., (C) Synechococcus elongatus (Nägeli) Nägeli, (D) Synechococcus nidulans (Pringsheim) Komárek, (E) Chroococcus schizodermaticus West, (F) Chroococcus minutus (Kützing) Nägeli, (G) Oscillatoria sp., (H) Pseudanabaena galeata Böcher, (I) Nostoc sp. All scale bars = 10 μm unless specified otherwise.



## DISCUSSION

The subaerial habitats found in terrestrial ecosystems provide a unique ecological niche that supports the growth of the rare and diverse collection of terrestrial microalgal species. However, only a few studies have been documented in these environments, and to date, no detailed floristic survey of the subaerial algal flora in forest areas in the Philippines has been published (Arguelles, 2019c; Martinez, 1984; Velasquez, 1962; Zafaralla, 1998). This investigation is the first floristic survey of microalgal communities in elevated subaerial rocks found in Mt. Makiling Forest Reserves. Eighteen subaerial microalgal species were identified from scraped algal samples on rock surfaces, spanning five classes: Cyanophyceae (7 species), Bacillariophyceae (5), Chlorophyceae (3), Trebouxiophyceae (2), and Mediophyceae (1). These algal species are documented for the first time in this unique habitat and are considered new distributional records for the Philippines. In addition, the study documented the existence of a rare microalgae, *Synechococcus elongatus* (Nägeli) Nägeli, which is taxonomically described as a new record in the Philippines. *Stephanocyclus meneghinianus* (Kützing) Kulikovskiy, Genkal & Kociolek is also reported here for the first time in the Philippines.

The population of subaerial algal taxa (18 species) identified and described in this survey is comparable with a previous floristic survey done by Neustupa and Skaloud (2008) as well as Rindi & Guiry (2003), where they documented a total of 28 and 51 algal taxa, respectively. Interestingly, only four species of subaerial algae identified in this study (*Desmococcus olivaceus*, *Nostoc* sp., *Chlorella* sp., and *Chroococcus* sp.) were also observed from previous studies. Previous studies documented green microalgae, cyanobacteria, and diatoms as the main and dominant class of algae in a subaerial environment, which conforms to the results obtained in this study (Davis & Karpook, 1999; Sophia et al., 2016). Also, some of the microalgal species documented in the current survey were not reported from other previous taxonomic surveys of microalgae from subaerial habitats, which proves the unique diversity of the algal flora in Mt. Makiling Forest Reserves (Liu et al., 2023; Lopez-Bautista et al., 2006; Neustupa & Skaloud, 2008; Rindi & Guiry, 2003). Generally, variations in the microalgal diversity in subaerial habitats are common, with some cosmopolitan species and others regarded as rare and temporary.

In previous studies, substratum was considered one of the important factors that affect the distribution and diversity of subaerial algae in rock surfaces (Davis & Karpook, 1999; Rindi & Guiry, 2003; Rifón-Lastra & Noguerol-Seoane, 2001). Substratum characteristics such as hardiness, pH, porosity, texture, and chemical composition play a critical role in dictating the group of microalgae that will colonize the rock substrate. Generally, cyanobacteria prefer alkaline rock surfaces more than the acidic ones (Rindi & Guiry, 2003). This explains the dominance of filamentous and coccoidal species of cyanobacteria in rock surfaces of limestones from the sampling area. In addition, proliferous growth of cyanobacteria in subaerial rock surfaces can also be attributed to the ability of these organisms to form mucilage and sheaths as a form of adaptation to extreme desiccation. Subaerial algae are well-adapted species to varying environmental conditions where they exist. These microalgal taxa form a dense and diverse community even when exposed to extreme dryness and heavy rains. The adaptive strategy of microalgae to counter extreme dry conditions includes the formation of zygospores, sporopollenin-like compounds, autospores, akinete, and cysts formation (Arguelles, 2021c).

These ecological adaptations allow subaerial microalgae to endure situations when a long duration of dryness in tropical rainforests occurs. The occurrence of *D. olivaceus* was observed in almost all the samples collected on the subaerial rock surfaces. This algal species seems to be one of the most tolerant and common subaerial algal species. Previous studies showed that *D. olivaceous* has a wide ecological tolerance since this organism can grow in extreme conditions such as very high nitrogen concentrations, permanent shade (limited sunlight), polluted air, and extreme dryness (Arguelles, 2019c; John & Rindi 2015; Lopez-Bautista et al., 2006; Neustupa & Skaloud, 2008; Rindi & Guiry, 2003). The proportion of new and undescribed species of microalgae is high in subaerial tropical habitats, considering that several cryptic and pseudocryptic species occur in different groups of terrestrial algae.

In this study, some species were identified up to the genus level since morphology-based taxonomy of the algal strain requires a more detailed analysis (such as scanning electron microscopy imaging and molecular analysis) to have a more accurate taxonomic delimitation. Therefore, taxonomic conclusions on



the identification of novel species of subaerial algae should be made using a combination of phenotypic and molecular data (Arguelles, 2019c; John & Rindi 2015; Neustupa & Skaloud, 2008).

Due to limited documented floristic studies on subaerial algae in rainforest areas in the Philippines, the algal records of this study are regarded as an important contribution to the increasing knowledge on the diversity of microalgae in the country. It is important to emphasize that little is known about the subaerial algae of tropical rainforests. Therefore, additional taxonomic surveys in Philippine forests, as well as ecological studies (such as evaluation of the effect of light, pH, and substratum in diversity and distribution pattern of subaerial algae), should be the topic for future studies. The results of these studies will help us to fill the gap in understanding the occurrence and diversity of subaerial algae that can be found in several unexplored natural subaerial habitats in the Philippines.

## CONCLUSION

This study represents the first floristic survey of subaerial epilithic microalgal communities from elevated rock surfaces in Mt. Makiling Forest Reserve, contributing 18 newly documented species to the Philippine algal inventory for subaerial habitats. The discovery of *Synechococcus elongatus* (Nägeli) Nägeli and *Stephanocyclus meneghinianus* (Kützing) Kulikovskiy, Genkal & Kociolek as new records in the Philippines underscores the untapped algal diversity present in tropical rainforest environments. Beyond taxonomic value, the findings have broader ecological implications, as subaerial algae play essential roles in primary production, biofilm formation, and nutrient cycling on terrestrial surfaces—especially in forest ecosystems where microhabitat diversity supports unique microbial communities.

From a conservation standpoint, documenting microbial biodiversity in protected areas like Mt. Makiling reinforces the importance of preserving microhabitats that are often overlooked in biodiversity assessments. While morphology-based identification provided an initial taxonomic baseline, future studies should integrate complementary approaches such as scanning electron microscopy (SEM) for high-resolution analysis of diatom frustule structures, DNA barcoding to resolve cryptic species, and pigment profiling (e.g., via HPLC or TLC) to support biochemical differentiation of algal groups. These advanced tools can refine species delineation, reveal hidden diversity, and strengthen the ecological and phylogenetic understanding of subaerial microalgal communities in tropical environments.

#### Funding

The research was funded by the University of the Philippines Los Baños (UPLB Core Project Fund Code: 4700004).

**Ethical Approval** 

Not applicable

#### **Competing interest**

The authors declare no conflicts of interest.

#### **Data Availability**

Data will be made available by the corresponding author on request.

#### **Declaration of Artificial Intelligence Use**

In this work, the author did not use generative AI or AI-assisted technologies in the preparation, analysis, or writing process.

#### Acknowledgement

The author gratefully acknowledges the Philippine National Collection of Microorganisms at the National Institute of Molecular Biology and Biotechnology (BIOTECH), University of the Philippines Los Baños, for their



invaluable support throughout this study. Sincere appreciation is also extended to the anonymous reviewers whose thoughtful comments and suggestions greatly improved the quality of this manuscript.

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## How to cite this article:

Arguelles, E. DLR. (2025). New Records of Subaerial Epilithic Algae from Elevated Rock Surfaces in Mt. Makiling Forest Reserve Laguna, Philippines. Recoletos Multidisciplinary Research Journal 13(1), 149-163. <u>https://doi.org/10.32871/rmrj2513.01.12</u>