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Benthic and Planktonic Microalgae Community in Probolinggo Beach

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Abstract

Microalgae, micro-sized plant organisms, play a crucial role in aquatic ecosystems. While many microalgae inhabit substrates or the bottom of water bodies, several types are planktonic. This study aimed to identify the types and abundance of microalgae in both sediment and water column habitats, as well as to analyze the environmental factors influencing their abundance. The research encompasses observations of water quality factors, microalgae abundance, relative abundance, diversity index, evenness index, and dominance index. Statistical analyses were using non-metric multidimensional scaling (NMDS) and Canonical Correspondence Analysis (CCA). The study was conducted in May-June 2022, with bi-weekly sampling at three points within each location for two months. Microalgae identified in the coastal area of Probolinggo belong to the Bacillariophyceae, Cyanophyceae, and Chlorophyceae classes. The highest microalgae abundance in the sediment habitat was 58,472 ind/cm^2, while in the water column was 4,118 ind/l. Diversity, evenness, and dominance indices in both sediment and water column habitats ranged from 1.93 to 2.61, 0.88 to 0.98, and 0.09 to 0.10, respectively. NMDS and CCA analyses indicate a graphical representation of the Bacillariophyceae class, demonstrating its prevalence across all sites.

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INTRODUCTION

Microalgae, microscopic organisms capable of efficient photosynthesis (Rafaelina *et al*., 2016), play a vital role in the environment, particularly in marine ecosystems. Serving as primary producers, they support marine ecosystem life by generating oxygen, constructing food webs, and providing habitats for various organisms. These organisms have similar characteristics to plants in general, as they possess chlorophyll pigments and can conduct photosynthesis, akin to general plant biology (Purbani *et al*., 2019).

Microalgae are frequently observed in coastal areas characterized by diverse ecosystems, including mangroves, macroalgae, and coral reefs (Risjani *et al*., 2021). Beyond these ecosystems, microalgae can also live in the water columns (planktonic) and sediments (epiphytic). According to Nirmalasari (2018), the abundance of planktonic microalgae in waters serves as a vital food source for other biotas. Planktonic microalgae, equipped with chlorophyll, play an important role in photosynthesis, generating organic matter and dissolved oxygen essential for the marine food cycle. Epiphytic microalgae, described by Lestari *et al*. (2020), are benthic and planktonic microalgae that live attached to various substrates, such as macroalgae.

The abundance of epiphytic microalgae serves as an indicator of water quality pollution. Varied habitat characteristics will influence microalgae diversity, offering insights for predicting water quality in diverse ecosystems. Several types of microalgae exhibit distinct levels of adaptation to environmental conditions, including physical, chemical, and biological parameters (Apriansyah *et al*., 2021; Arsad *et al*., 2022).

The coastal area of Probolinggo Beach harbors a substantial population of microalgae. Obtaining information on the distribution of microalgae-based on substrate characteristics is of paramount importance. This study aimed to identify the types and abundance of microalgae in sediment and water column habitats, with a concurrent analysis of environmental factors influencing microalgal abundance.

METHODOLOGY

Ethical Approval

In this study, it is noteworthy that no animals were employed. Ethical approval for this research aligns with established guidelines for humane and responsible scientific inquiry, ensuring the welfare and ethical treatment of living organisms are upheld under applicable standards.

Place and Time

The research was conducted in the coastal area of Probolinggo Beach, consisting three distinct sites: site 1 (Bentar Beach, coordinates -8°13'16.7" South Latitude 113°16'36.7" East Longitude), site 2 (Pesona Beach, coordinates -8°14'45.1" South Latitude 113°15'23.8" East Longitude), and site 3 (Tambak Bahak Beach, coordinates -8°16'28.2" South Latitude 113°7'8.8' ' East Longitude). The research was conducted in May-June 2022. Sampling was carried out two times every two weeks with three sampling points at each designated location.

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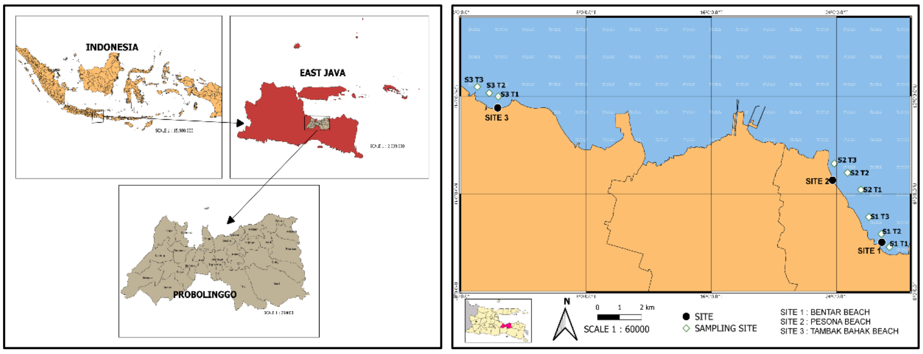

Figure 1. Map of microalgae diversity research location.

Research Materials

The sampling of microalgae in sediments adhered to the methodology outlined by Benni *et al*. (2020). Sediment samples were extracted at a depth of 1 cm during the lowest tide, and collected from the surface of sediments positioned along a 5x5 cm transect. Subsequently, the samples were placed into 100 ml bottles filled with seawater and homogenized to facilitate the dissolution of microalgae from the substrate. Following homogenization, 30 ml of the sample was extracted and treated with 2 ml of 1% Lugol's solution.

The sampling of microalgae in the water columns involved the extraction of 25 l of water samples at various distinct points. Subsequently, the water samples underwent filtration using a plankton net with a mesh size of 150 microns. The filtered samples were then transferred to 30 ml sample bottles and treated with 2 ml of a 1% Lugol solution.

Water quality measurements, including temperature, were conducted using a portable pH/Mv/Oc meter. Transparency was assessed with a Secchi disk, while current velocity was measured using a Current Meter Tatonas TH-02. pH levels were determined with a Krisbow kw06-744 pH meter, dissolved oxygen (DO) with a PDO-520 DO meter, and salinity with a refractometer. Additionally, the concentrations of orthophosphate, nitrate, and chlorophyll-a were quantified using a spectrophotometric method following the Boyd Nessler protocol.

Research Design

The employed research methodology was a quantitative descriptive approach, wherein data were collected directly at the research location using a purposive sampling technique (Etikan *et al.*, 2016). The sampling process commenced with the measurement of water quality, comprehensive observation, and recording of the surrounding environmental conditions. Additionally, samples were collected for subsequent analysis at the Laboratory of Hydrobiology, Universitas Brawijaya, Malang, together with the identification of microalga types and quantitative calculations.

Work Procedure

Observation and Calculation of Microalgae

The quantification of microalgae abundance in the sediment habitat referred to the APHA formula (2012), while the assessment of microalgae abundance in the water column habitat followed the APHA formula (1989). Water quality indices included the diversity index (Winahyu *et al*., 2013), the evenness index (Winahyu *et al*., 2013), and the dominance index (Lestari *et al*., 2020).

Data Analysis

The acquired data were processed using Microsoft Excel 2013. Statistical analysis employed to determine the correlation between water quality parameters and microalgae abundance encompassed non-metric multidimensional scaling

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(NMDS) and canonical correspondence analysis (CCA) using the Past 4.10 software.

RESULTS AND DISCUSSION

Microalgae Composition

The microalgae composition identified in sediments comprised 16 genera, consisting of three classes including Bacillariophyceae (12 genera), Chlorophyceae (1 genus), and Cyanophyceae (3 genera). Similarly, microalgae observed in the water columns exhibited 15 genera, involving three classes-Bacillariophyceae (11 genera), Chlorophyceae (1 genus), and Cyanophyceae (3 genera). The prevalence of the Bacillariophyceae class in Probolinggo Beach is noteworthy, aligning with Everest and Aslan's (2016) statement that Bacillariophyceae is a common class in various aquatic environments, attributed to its adaptability to diverse conditions owing to the silica composition of their cell walls, conferring resistance to environmental challenges.

The genera identified in sediments include *Amphora, Guinardia, Isthmia, Nitzechia, Gyrosigma, Pleurosigma, Rhizosolenia, Diploneis, Pinnularia, Navicula, Rhopalodia,* and *Synedra*, from the Bacillariophyceae class; *Chladophora* genus from the Cyanophyceae class; and *Oscillatoria, Bacularia and Phormidium* genera from the Cyanophyceae class. Moreover, the genera identified in the water columns include *Navicula, Guinardia, Hemiaulus, Isthmia, and Synedra. Nitzschia, Achnanthes, Rhizosolenia, Gyrosigma, Amphora,* and *Pleurosigma* from the Bacillariophyceae class; *Chladophora* genus from the Chlorophyceae class; and *Oscilatoria, Bacularia* and *Phormidium* genera from the Cyanophyceae class.

Microalgae Abundance

The observed variations in microalgae across each site indicate a consistent pattern. Genera identified in the water column exhibit diversity, likely influenced by frequent tides and sea waves at Probolinggo Beach. Jannah (2020) stated that the dynamic nature of tides and sea waves significantly impacts microalgae presence. Furthermore, the influence of high currents is suggested to contribute to the presence of epiphytic microalgae in water columns, resulting in increased microalgal diversity.

Table 1. Average microalgae abundance on Probolinggo Beach.

Habitat	Water Column (Ind/L)		Sediment (Ind/cm^2)	
	Measurement 1	Measurement 2	Measurement 1	Measurement 2
Bentar Beach	4118	3157	51060	44472
Pesona Beach	3843	2951	45295	37059
Tambak Bahak Beach	4049	3637	58472	50237

Source: Research documentation, 2022.

The highest recorded microalgae abundance during measurement 1 was observed in Bentar Beach, registering at 4,118 ind/L, while during measurement 2, Tambak Bahak Beach exhibited the highest abundance at 3,637 ind/L. Regarding sediment samples, Tambak Bahak Beach displayed the highest microalgae abundance during both measurements, with values of 58,472 ind/cm^2 and 50,237 ind/cm^2, consecutively. Disparities in microalgae abundance can be attributed to factors such as rainfall, nutrient levels, and currents. Benni *et al.* (2020) highlight that sediment type plays a crucial role in influencing organic matter content. Sandy sediments, characterized by larger pore water, facilitate efficient oxidation, resulting in lower organic matter content. Conversely, muddy clay sediments, with a finer texture, exhibit higher organic matter content. Notably, Tambak Bahak Beach manifested a higher microalgae abundance in sediments compared to other sites.

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Relative Abundance

The relative abundance of microalgae in both sediments and water columns at the research location in Probolinggo Beach exhibited substantial variation. In sediment samples, the Bacillariophyceae class, specifically the *Synedra* genus, demonstrated the highest relative abundance during measurement 1, constituting 42%. This abundance was further detailed across the three sampling sites: 16% at site 1, 15% at site 2, and 11% at site 3. In measurement 2, the relative abundance was 32%, with site-specific proportions of 11%, 13%, and 8% for sites 1, 2, and 3, respectively. The prevalence of the *Synedra* genus in sediments is attributed to its diatom form, characterized by layered wrapping cells, enabling adaptation to low dissolved oxygen environments, resulting in its abundant presence in water bodies (Lobban *et al*., 2019). Conversely, in water columns, the Bacillariophyceae class, particularly the *Rhizosolenia* genus, exhibited the highest relative abundance during measurement 1, amounting to 47%. Site-specific proportions were 17%, 11%, and 19% for sites 1, 2, and 3, respectively. In measurement 2, the relative abundance was 27%, with site-specific proportions of 11%, 5%, and 11% for sites 1, 2, and 3. The consistently high abundance of the Bacillariophyceae class is consistent with findings in various studies, highlighting its resilience to extreme environmental conditions. Microalgae genera with cosmopolitan characteristics, such as the *Synedra* and *Rhizosolenia*, are known to thrive in diverse habitats. According to Adinugroho *et al*. (2014), *Rhizosolenia* serves as a natural food source for aquatic organisms, particularly fish in marine environments.

Biological Index

Diversity Index

The determination of the diversity index for microalgae in sediments produced a range of values. In measurement 1, the diversity index values in sediments ranged from 2.47 to 2.60, while in measurement 2, they ranged from 2.56 to 2.61. Examination of the diversity index values revealed that, in measurement 1, the highest value occurred at site 3, registering at 2.60, whereas the lowest was recorded at site 2, with a value of 2.47. In measurement 2, the diversity index exhibited its highest value at site 3, measuring 2.61, and its lowest at site 2, with a value of 2.55.

The diversity index measurements for microalgae in water columns exhibited a range of values. In measurement 1, the diversity index values in water columns ranged from 2.2 to 2.51, while in measurement 2, they ranged from 1.93 to 2.52. Analysis of the diversity index values revealed that, in measurement 1, the highest value was recorded at site 1, reaching 2.51, while the lowest was observed at site 2, with a value of 2.20. Conversely, in measurement 2, the diversity index demonstrated its highest value at site 3, measuring 2.52, and its lowest at site 2, with a value of 1.93.

Analysis of the diversity index for microalgae in sediments and water columns across three sites, which exhibited values within the range of less than 3 and more than 1, suggest that the area maintains a biota community with a moderate level of stability. The quality of the water columns appears to be relatively conducive to the growth of aquatic biota. According to Inyang and Wang (2020), a high diversity index implies an even distribution of species, which can contribute to the control of physico-chemical parameters within the area.

Evenness Index

The evenness index of microalgae for sediments in measurement 1 exhibited values ranging from 0.94 to 0.96, with the highest value recorded at site 3, accounting for 0.96. Concurrently, the evenness index shared identical values at sites 1 and 2, both registering at 0.94. In measurement 2, the evenness index ranged from 0.96 to 0.98, with the highest value observed at site 1, accounting for 0.98, and

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the lowest value recorded at site 3, measuring 0.96.

The evenness index for water columns in measurement 1 demonstrated values ranging from 0.89 to 0.95, with the highest value recorded at site 1, accounting for 0.95, and the lowest value observed at site 2, accounting for 0.89. In measurement 2, the evenness index exhibited a range of values from 0.88 to 0.98 with the highest value noted at site 3, measuring 0.98, and the lowest value recorded at site 2, accounting for 0.88.

The findings of the evenness index values observed in both in sediments and water columns indicated values approximating 1. Consequently, it can be inferred that the uniformity within water columns is in a balanced state, suggestive of the absence of competition for resources or habitat. Conversely, evenness value nearing 0 signifies competitive dynamics and the presence of species dominance within these aquatic environments (Fan *et al*., 2021).

Dominance Index

The dominance index for sediments in measurement 1 showed a range of values between 0.06-0.10 with the highest dominance index value found at site 2, accounting for 0.10, and the lowest value at site 3, accounting for 0.06. Meanwhile, measurement 2 showed a range of values between 0.08-0.09 with the highest dominance index value at site 2, accounting for 0.09 and the domination index value at sites 1 and 3 shared an identical value of 0.08. Regarding water columns in measurement 1, the dominance index ranged between 0.09-0.10. The dominance index in water columns in measurement 2 showed a value in the range between 0.09-0.12.

The outcomes of the measurements for the dominance index of microalgae in both sediments and water columns revealed a value close to 0 at each site. This suggests the absence of any single dominating species within the area. As indicated by Marsela *et al*. (2021), the domination index ranges from 0 to 1, and a smaller value signifies the absence of any dominating species, whereas a larger value indicates the prevalence of a dominant species.

Water Quality Parameters

The following table (Table 2) presents the water quality parameters recorded during measurements 1 and 2 at three sites. These parameters encompass various crucial indicators contributing to our understanding of the aquatic environment. The measured values include temperature (°C), transparency (cm), depth (cm), currents (m/s), salinity (ppt), dissolved oxygen (DO, mg/L), pH, nitrate (mg/l), orthophosphate (g/l), and chlorophyll-a (mg/m^3). This comprehensive dataset provides valuable insights into the environmental conditions.

Based on Table 2, the water quality parameters measured in this study exhibited values within a normal range conducive to microalgae growth. According to Lauritano *et al*. (2020), variations in water temperature inversely correlate with the quantity and diversity of microalgae. This phenomenon arises due to the impact of temperature fluctuations on the metabolic activity and developmental processes of microalgae. Furthermore, the transparency of water also affects microalgae density, with higher abundances observed at a depth of 1 m compared to a depth of 5 m. This observation aligns with established trends indicating a positive correlation between transparency and microalgae abundance. Additionally, the study indicates that increased water current velocity can impede the deposition of organic matter, thereby stimulating higher concentrations of benthic microalgae.

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Table 2. Water quality parameters.

Parameter	Measurement 1			Measurement 2			Literature
	Site 1	Site 2	Site 3	Site 1	Site 2	Site 3	
Temperature (⁰C)	30.5 - 31.4	31 – 31.8	33.5-33.8	28.7-30.9	31.5-32.7	30.8-33	25–35°C (Maulana *et al.*, 2017)
Transparency (cm)	40 – 43.5	39.5 – 43	35 -39	42.5 – 47.5	37 – 38. 5	32 – 36	>45 cm (Herawati *et al.*, 2021)
Depth (cm)	89 -93	89-94	75 - 86	99- 121	93- 104	84 – 91	-
Currents (m/s)	0.052 – 0.057	0.043 0.049	0.077 – 0.081	0.061 – 0.069	0.052 – 0.057	0.045 – 0.051	Slow current< 0.1 m/s medium current 0,1–1 m/s (Padang *et al.*, 2020)
Salinity (ppt)	26 - 30	30-31	23 - 26	26 - 30	29 - 31	23 – 24	15–32 ppt (Nurjijar *et al.*, 2022)
DO (mg/L)	8.1 – 8.3	8 – 8.2	7.3 – 7.6	6.9–7.3	7.2–7.7	7 – 7.7	>5 mg/ L (Arofah *et al.*, 2021)
pH	7.4 – 7.7	7.3– 7.8	7.3 – 7.6	7.5–7.8	7.8–8.1	7.7–7.8	6–9 (Kumar and Saramma, 2018)
Nitrate (mg/l)	0.22 – 0.35	0.23 – 0.36	0.33 – 0.40	0.22 – 0.61	0.16 – 0.27	0.27 – 0.81	Oligotrophic 0–1 mg/ L (Adriani *et al.*, 2019)
Orthophosphate (g/l)	0.02 – 0.1	0.04 – 0.05	0.05 – 0.09	0.05 – 0.07	0.03 – 0.04	0.09 – 0.12	Oligotrophic <0.015 to mesotrophic 0.015-0.13 mg/ L (Nurjijar *et al.*, 2022)
Chlorophyll-a (mg/m^3)	0.52 – 1.01	0.79 – 1.13	0.42 – 0.97	1.8 – 3.1	0.69-0.87	0.57 – 1.41	15-30 mg/m^3 (moderate) (Gunawan *et al.*, 2019)

NMDS (Non-metric Multidimensional Scaling)

The statistical analysis used in this study was the NMDS. This method grouped the number of microalgae found at each site based on class, specifically Bacillariophyceae, Chlorophyceae, and Cyanophyceae (Figure 2).

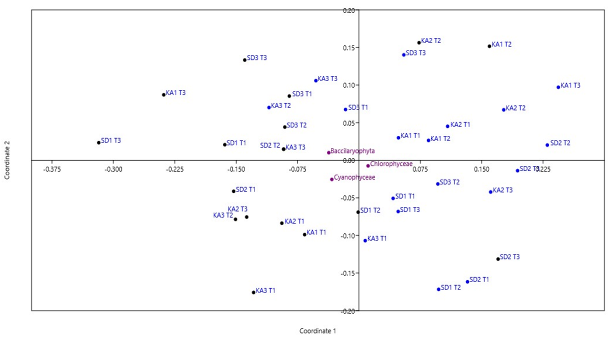

Figure 2. Sketch of statistical analysis NMDS; (SD) sediments, (KA) water columns.

Concurrently, environmental parameters, including temperature, transparency, currents, depth, pH, DO, nitrate, orthophosphate, salinity, and chlorophyll-a were quantified. The NMDS analysis yielded a stress value of 0.09703 and an R^2 value of 0.9149 on axis 1 and 0.001836 on axis 2. These values falling below 0.15, are considered indicative of a robust analysis with a low error rate (Rahman *et al.*, 2020). Statistical analysis revealed a proximity of points representing sediments and water columns, suggesting

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a similarity and even distribution of microalgae between these two habitats. This observed resemblance is attributed to the uniform sampling process conducted at the same geographical location.

CCA (Canonical Correspondence Analysis)

The statistical analysis resulting from the environmental parameter approach used in the CCA method yielded a strong positive correlation among specific parameters. Bacillariophyceae exhibited positive correlations with orthophosphate, DO, temperature, and nitrate. Chlorophyceae demonstrated positive correlations with pH, salinity, chlorophyll-a, currents, depth, and transparency. Meanwhile, Cyanophyceae displayed positive correlations with transparency and depth. According to Yolanda *et al*. (2016), there is a positive relationship between DO and nitrate. A higher amount of dissolved oxygen (DO) in water helps break down organic matter. This process increases nitrate levels, which, in turn, leads to higher chlorophyll-a content. The concentration of nutrients in water plays a role in regulating microalgae growth, as insufficient nitrate content can result in microalgae mortality. The results of statistical analysis, illustrating the correlations between microalgae with water quality parameters are depicted in Figure 3.

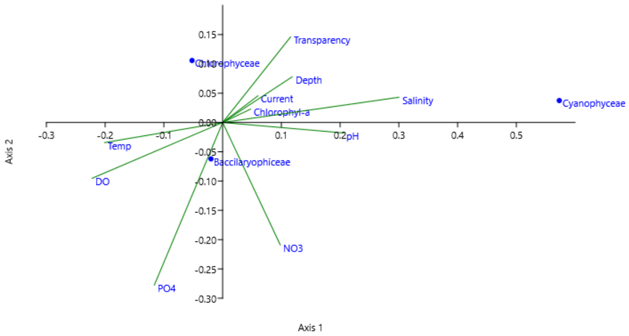

Figure 3. Sketch of CCA statistical analysis.

The statistical analysis of the environmental parameters of microalgae generates a vector line on the environmental parameter scale leading to the microalgae point. This alignment signifies elevated environmental parameters within the area. Conversely, if the point diverges from the direction of the environmental parameter vector line, it suggests lower environmental parameters. This discrepancy implies that specific parameters, such as dissolved oxygen (DO) and nitrate, exhibit higher values. This observation suggests the presence of factors, such as mangroves, known to enhance DO and nitrate content. According to Indrawati *et al.* (2013), plant-derived litter contributes to heightened nutrient levels and is utilized by microalgae. Additionally, plants will increase DO levels through the process of photosynthesis.

CONCLUSION

The findings from the microalgae identification indicate that the Bacillariophyceae class is predominant across all locations at Probolinggo Beach. The measured water quality parameters were within a typical range conducive to microalgae growth. The microalgae at Probolinggo Beach exhibit a normal growth pattern, as supported by the evenness, diversity, and dominance indices values. These values collectively suggest the absence of significant dominance by a particular type of microalgae. Therefore, it can be inferred that the waters of Probolinggo Beach maintain a relatively favorable ecological condition.

CONFLICT OF INTEREST

There is no conflict of interest among all authors upon writing and publishing the manuscript.

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AUTHOR CONTRIBUTION

The authors' contributions to the study or experiment are as follows: Sulastri Arsad conceptualized the study, did data analyses, wrote the articles, and conducted revisions. Rut Suharni P. Sihombing undertook sampling, laboratory work, and data analysis, and contributed to article writing. Mohammad Mahmudi contributed to the study's conceptualization and performed statistical and data analyses. Oktiyas Muzaky Luthfi provided corrections and constructive input for the manuscript. Ikha Safitri offered critical insights during the manuscript's development. Fika Dewi Pratiwi provided constructive criticism for the manuscript's enhancement.

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