

# Community structure of epilithic microalgae in the Upper Setail River, Banyuwangi District, East Java, Indonesia

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**Abstract.** Arfiati D, Zakiyah U, Rusydi AN, Al Athor MZ, Orchida K, Inayah ZN, Ulya WH, Sahal AA, Rachmawati R, Andhani TD, Pratiwi RK. 2025. Community structure of epilithic microalgae in the Upper Setail River, Banyuwangi District, East Java, Indonesia. *Biodiversitas* 26: 2309-2319. Setail River is one of the freshwater ecosystems in Banyumas District, East Java, Indonesia, which contains a lot of rocky substrates. This substrate can serve as a site for epilithic microalgae to attach, which act as natural food sources for aquatic organisms and serve as a bioindicator of the aquatic environment. The purpose of this study was to analyze the community structure of epilithic microalgae in the Setail River. This study was carried out from December 2023 to September 2024 using a survey method at 5 sampling points in the Upper Setail River, Banyuwangi District. Epilithic microalgae samples were taken using the quadrant transect method (5x5 cm<sup>2</sup>) and identified using the Modified Lackey Drop Micro Transect Counting Method. The identified results included 35 genera from 4 divisions of epilithic microalgae. The microalgae divisions are Bacillariophyta (27 genera), Chlorophyta (2 genera), Cyanophyta (3 genera), and Charophyta (3 genera). The abundance of epilithic microalgae is 8,169-21,240 cells/cm<sup>2</sup>. The Bacillariophyta division is the most frequently found division, with a percentage ranging from 83 to 95%. *Navicula* is the most frequently found genus, with the highest average abundance of 6,225 cells per cm<sup>2</sup>, making it a key indicator of organic pollution in waters. The diversity index ranges from 1.65-2.17 (moderate, H': 1-3), the evenness index ranges from (0.75-0.80) (e>6 high), and the dominance index ranges from 0.17-0.33 (low, C approaching 0). The Saprobic Index analysis obtained in the Setail River waters ranges from 0.60 to 1, indicating that the waters are lightly polluted ( $\beta$ -mesosaprobic), which suggests that only a few pollutants enter from organic and inorganic materials. The lowest saprobic value was found at station 5, indicating that further management of waste input into the river is necessary to prevent severe pollution due to organic and inorganic waste in the Setail River waters.

**Keywords:** Bioindicator, community structure, diversity, epilithic microalgae, water quality

## INTRODUCTION

A river is an area of flowing fresh water (lotic waters) from upstream to downstream (Harvey and Schmadel 2021). Rivers provide numerous resources that are beneficial to both humanity and ecology, including supporting aquatic biodiversity, which encompasses flora and fauna (Dutta et al. 2017). As an ecosystem, rivers are home to various types of aquatic life and play an important role in the water cycle on Earth (Wang and He 2022). Nevertheless, river ecosystems are considered one of the most dynamic ecosystems in the world (Stubbington et al. 2022). This ecosystem is vulnerable to pollution due to both natural and human-induced interference, resulting in significant pressure on the river's biota and ecology that exceeds its assimilation and regeneration capacity (Angeles et al. 2018; Anh et al. 2023).

The Setail River is one of the large rivers in Banyuwangi District, East Java Province, Indonesia. The source of Setail River originates from Mount Raung and flows to the southern and eastern regions of Banyuwangi District. This river is a source of raw water for local

community activities. The Setail River empties directly into the sea, specifically in the Bali Strait or the Indian Ocean, in Muncar Sub-district, Banyuwangi District. Setail River Estuary, Banyuwangi, is an area with great potential in the fisheries sector. Moreover, the Setail River has quite high fishery biological resources (Arifki and Erwanto 2023).

Monitoring and management of the Setail River are necessary to maintain its quality and prevent pollution. Water quality monitoring can use biological indicator methods such as benthic macroinvertebrates, fish, and periphyton (including epilithic microalgae) (Vermaat et al. 2022; Sevindik et al. 2024). The use of biological indicators has various advantages, including reflecting overall ecological quality, as they provide accurate measurement information on environmental changes (Zaghloul et al. 2020; Samudra et al. 2022). Microalgae are widely used as indicators to detect disturbances in aquatic ecosystems because they are the primary producers and are sensitive to environmental changes (Huang et al. 2018; Ogi et al. 2021; O'Neill and Rowan 2022). Changes in microalgae biomass can serve as a key indicator of how human activities are altering river ecosystems (Dunck et al.

2015; Trottier et al. 2022).

Epilithic microalgae are microalgae that attach to a substrate in the rock surface (benthic algae or periphyton) (Saputra et al. 2018). The use of epilithic microalgae as bioindicators has several advantages over other organisms, making it more effective and economical. The distribution of epilithic microalgae in waters is very wide (González-Hourcade et al. 2023); they have limited movement or are sessile (settled) and can migrate or other changes to escape pollution, tolerate the surrounding environment or die (Arsad et al. 2021), have a short life cycle and reproduce quickly (Stancheva and Sheath 2016), are able to attach to rock substrates and are resistant to strong currents (Ugya and Meguellati 2022). Additionally, this organism is easier to sample in riverbeds that are quite shallow, as they are typically located upstream, and the substrate conditions are rocky. Community structure studies are crucial for understanding the health of the aquatic environment and its ecological function (Letáková et al. 2018).

Dynamic changes in epilithic microalgae communities can reflect the effects of environmental change (Peng et al. 2020; Zhong et al. 2022). Information regarding the diversity and composition of epilithic microalgae in the Setail River remains limited due to a lack of attention. The existence of the Setail River is crucial for the community, as it serves as a primary source of raw water for the City of Banyuwangi. Various community activities carried out in

the Setail River, especially in the upstream section, will result in changes to the water quality status, affect water availability, and impact the survival and diversity of aquatic biota. This research aims to analyze the community structure (identifying the type, diversity, and abundance) of epilithic microalgae and assess the health status of rivers using the saprobic index. We anticipate that this research will be beneficial for the management and conservation of the Setail River to protect and balance the ecosystem.

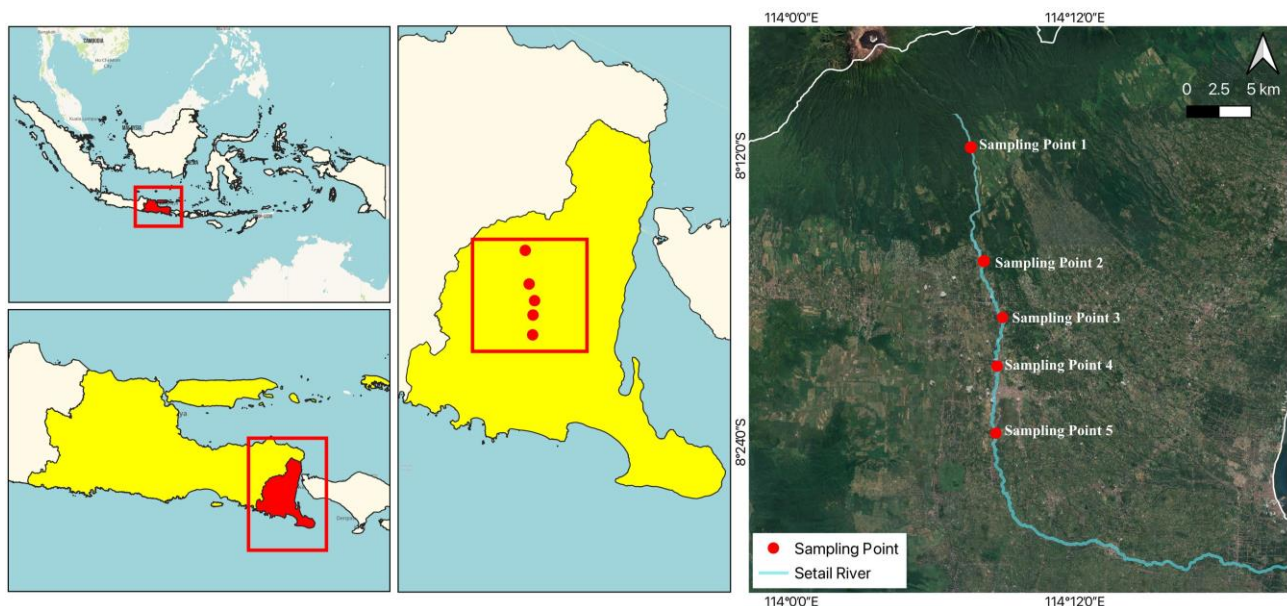
## MATERIALS AND METHODS

### Study area

This research was conducted from November 2023 to September 2024 in the Setail River, Banyuwangi District, East Java, Indonesia. This research is a field survey, while laboratory analysis was conducted at the Hydrobiology Laboratory of the Aquatic Environment Division and the Sand Source Freshwater Fisheries Laboratory, Faculty of Fisheries and Marine Sciences, Universitas Brawijaya, East Java. The selection of sampling locations was conducted using purposive sampling. There are 5 Sampling Points (SP), which can be seen in Table 1 and Figure 1. Sampling was carried out 3 times with an interval of 14 days (2 weeks).

**Table 1.** Sampling point in Setail River, Banyuwangi, East Java, Indonesia

| Sampling point | Coordinate                      | Positioning and characteristics   |
|----------------|---------------------------------|---|
| 1              | 8°11'38.1" S and 114°07'36.5" E | Upstream area, near the waterfall, located at 712 meters above sea level              |
| 2              | 8°16'24.1" S and 114°08'09.6" E | Around the agricultural area and location at 355 meters above sea level               |
| 3              | 8°18'45.9" S and 114°08'56.4" E | Neighboring residential areas and location at 243 meters above sea level              |
| 4              | 8°20'48.3" S and 114°08'42.8" E | City areas and around cafe areas and location at 195 meters above sea level           |
| 5              | 8°23'36.9" S and 114°08'39.8" E | Areas around residents' gardens (moorland) and location at 138 meters above sea level |



**Figure 1.** Research location map showing sampling points in Setail River, Banyuwangi District, East Java, Indonesia

## Research procedure

### Sample collection

Samples of epilithic microalgae were taken by taking river substrate in the form of rocks, whose surface area was then measured. Next, the sample is removed from the stone by brushing in one direction and temporarily placed in a petri dish. After all the samples have been removed from the media and the brush has been cleaned with a small amount of distilled water, ensure that any remaining samples are completely removed and place them in a sample bottle. The bottle is then filled with 1% Lugol's solution and 2% formalin. Next, the organisms that make up the periphyton are identified using a microscope.

### Laboratory analysis

Laboratory analysis was conducted to identify and count the genus of epilithic microalgae found. Observations were made using an Olympus binocular microscope with 40x ocular magnification. The epilithic microalgae found were counted using the Modified Lackey Drop Micro transects Counting Method and documented (Utama et al. 2019). The identification of the genus of epilithic microalgae was based on the identification guidebooks by Prescott (1962) and Bellinger and Sigeo (2010).

### Measurement of water quality

Measurement of physical and chemical parameters of water was conducted in situ and ex situ. In situ water quality measurements include temperature, measured using a thermometer; current velocity, measured with a modified Current meter; Dissolved Oxygen (DO) levels, measured with a DO meter; and pH, measured with pH paper. At the same time, organic matter was measured ex situ using the  $\text{KMnO}_4$  titration method, and nitrate, phosphate, and ammonia were measured using a spectrophotometric method.

## Data analysis

### Epilithic microalgae abundance

The abundance value of epilithic microalgae can be calculated using the modified formula of the Lackey Drop Micro Transect Method (Utama et al. 2019) as follows:

$$K (\text{cell}/\text{cm}^2) = \frac{N \times At \times Vt}{Ac \times Vs \times As} \quad (1)$$

Where,

- K : Epilithic microalgae abundance (cells/cm<sup>2</sup>)
- N : Number of epilithic microalgae observed
- As : Leaf substrate area (cm<sup>2</sup>)
- At : Cover glass area
- Ac : Field of view
- Vt : Epilithic microalgae sample volume
- Vs : Sample volume observed

### Diversity index

The Shannon-Weaver Diversity Index was used to determine the epilithic microalgae diversity (Shannon and Weaver 1949; Masithah and Islamy 2023). The calculation is as follows:

$$H' = \sum_{i=1}^S p_i \ln p_i \quad (2)$$

Where,

- H' : Diversity index
- S : Number of species
- $p_i$  :  $n_i/N$
- $n_i$  : Number of individuals of species i
- N : Total number of individuals

Diversity index criteria:  $H < 1$ : Low diversity;  $1 < H < 3$ : Medium diversity;  $H > 3$ : High diversity.

### Evenness index

The evenness index indicates the evenness of the species (Ogi et al. 2021). The evenness index was calculated using the following formula:

$$E = \frac{H'}{H_{\max}} \quad (3)$$

Where,

- E : Evenness index
- $H_{\max}$  : Maximum value of diversity/  $\ln(S)$
- S : Number of species.

The evenness index category: 0-0.4: Low diversity; 0.4-0.6: Medium diversity; 0.6-1.0: High diversity

### Dominance index

The dominance index is calculated to determine the type of epilithic microalgae that dominates (Samudra et al. 2022). The dominance index uses the Simpson Dominance Index formula as follows:

$$c = \sum_{i=1}^N \left(\frac{n_i}{N}\right)^2 \quad (4)$$

Where,

- C : Dominance index
- $n_i$  : Number of individuals-i
- N : Total number of individuals in the sample

The dominance index value ranges from 0 to 1. If the C value is close to 0, there are no dominant species; conversely, a C value closer to 1 indicates that a dominant species is present. The greater the dominance index value, the lower the diversity.

### Saprobic index

The saprobic index accorded to Dresscher and van der Mark (1976) and was calculated by the formula:

$$X = \frac{C + 3D - B - 3A}{A + B + C + D} \quad (5)$$

Where,

- X : Saprobic Coefficient, between -3 to +3
- A : Number of species groups Ciliates (polysaprobic)
- B : Number of species groups Cyanophyta (class Cyanophyceae), Euglenophyta ( $\alpha$ -mesosaprobic)
- C : Number of species groups Chlorophyta (order Chlorococcales), Chrysophyta (class Bacillariophyceae/Diatoms) ( $\beta$ -mesosaprobic)
- D : Number of species groups Peridinians + Chrysophyceae + Conjugate (oligosaprobic)

**Table 2.** Categories of water pollution level based on saprobic index

| Pollutant source                     | Pollution level | Saprobic phase               | Saprobic index value |
|--------------------------------------|-----------------|------------------------------|----------------------|
| Organic matter                       | Very heavy      | Polysaprobic                 | -3 to -2             |
|                                      |                 | Poly/ $\alpha$ -mesosaprobic | -2 to -1.5           |
| Organic and inorganic substances     | Medium          | $\alpha$ -meso/polysaprobic  | -1.5 to -1           |
|                                      |                 | $\alpha$ -mesosaprobic       | -1 to -0.5           |
|                                      |                 | $\alpha/\beta$ -mesosaprobic | -0.5 to 0            |
| Organic and inorganic substances     | Light           | $\beta/\alpha$ -mesosaprobic | 0 to 0.5             |
|                                      |                 | $\beta$ -mesosaprobic        | 0.5 to 1             |
|                                      |                 | $\beta$ -meso/oligosaprobic  | 1 to 1.5             |
| Few organic and inorganic substances | Very light      | Oligo/ $\beta$ -mesosaprobic | 1.5 to 2             |
|                                      |                 | Oligosaprobic                | 2 to 3               |

Source: Dresscher and van der Mark (1976)

## RESULTS AND DISCUSSION

### Epilithic microalgae diversity

The results of the epilithic microalgae community in the Setail River, obtained at 5 sampling points, revealed 4 divisions. The four divisions include Bacillariophyta, Cyanophyta, Chlorophyta, and Charophyta. The Bacillariophyta division consists of 27 genera including *Achnanthes*, *Achnantheidium*, *Amphora*, *Aulacoseira*, *Coconeis*, *Coscinodiscus*, *Cyclotella*, *Cymbella*, *Diatoma*, *Diploneis*, *Encyonopsis*, *Ephitemia*, *Fragilaria*, *Frustulia*, *Gomphonema*, *Gyrosigma*, *Isthmia*, *Melosira*, *Navicula*, *Nitzschia*, *Pinnularia*, *Rhoicosphenia*, *Sellaphora*, *Stauroneis*, *Surirella*, *Tabellaria* and *Tryblionella*. The division Cyanophyta consists of 2 genera, including *Anabaena* and *Oscillatoria*. The Chlorophyta division consists of 2 genera, including *Chlorella* and *Sphaerocystis*. The Charophyta division consists of 3 genera, including *Chlosterium*, *Cosmarium*, and *Hyaloteca*. An overview of the epilithic microalgae identified in this study is presented in Figure 2.

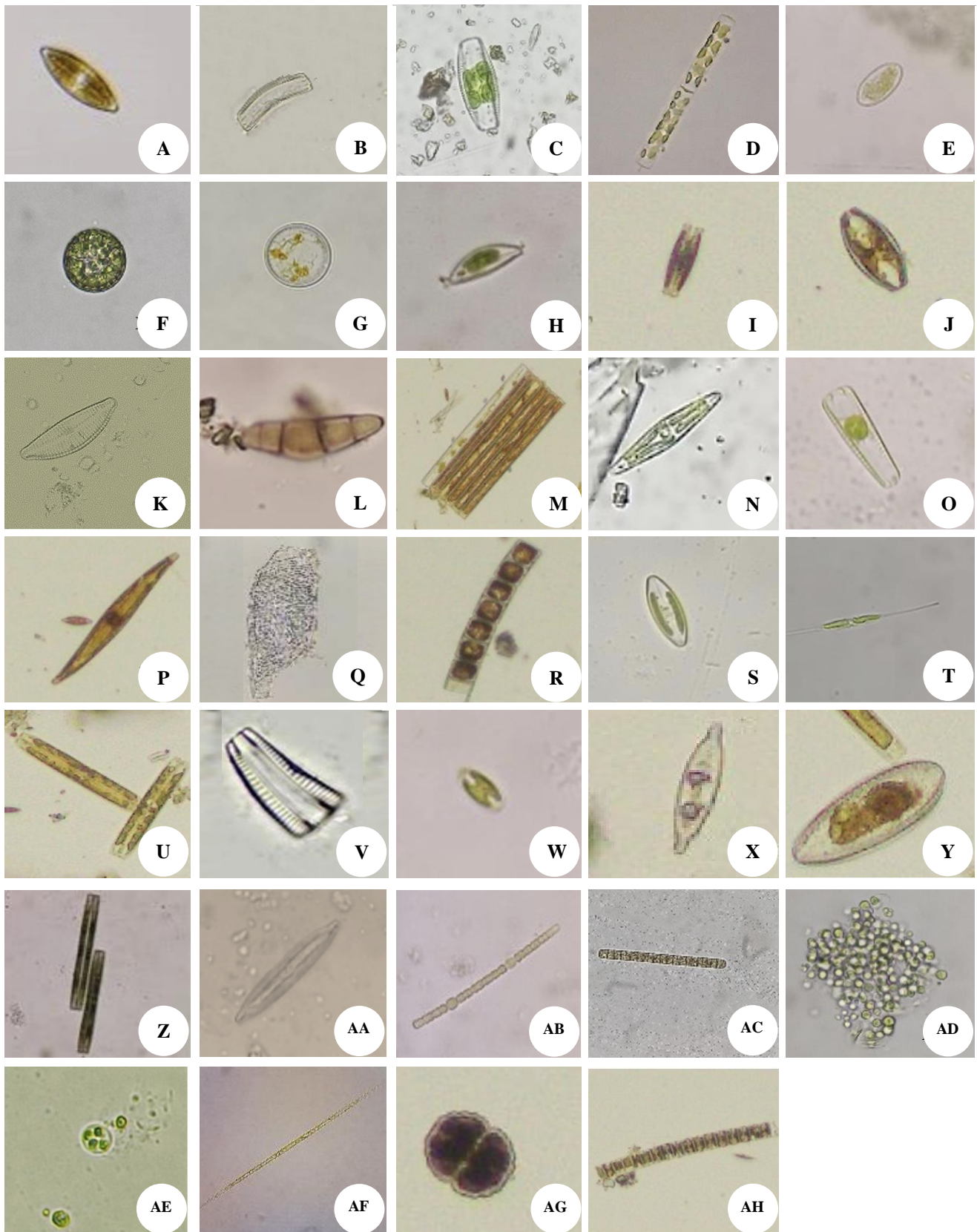
### Abundance of epilithic microalgae

The abundance of epilithic microalgae ranged from 8,169 to 21,240 cells/cm<sup>2</sup> (Figure 3). The highest abundance was found in SP 4, while the lowest was found in SP 5. Water currents could be a factor influencing the highest abundance in SP 4, as the currents in SP 4 tended to be lower than those in SP 1 to SP 3. The composition of periphyton species, including epilithic microalgae, changes in response to varying flow speeds, water level fluctuations, and wind-driven waves in different river waters, resulting in significant differences in cell density and biomass (Zhao et al. 2022). The condition of shade can also be influenced because it is related to the intensity of sunlight entering the water. Shade is one of the main factors controlling periphyton biomass (Kiffer et al. 2018). Warm water temperatures in these streams may be related to changes in riparian vegetation that allow more sunlight to reach the stream channels. The rapid growth of microalgae can be attributed to higher water temperatures,

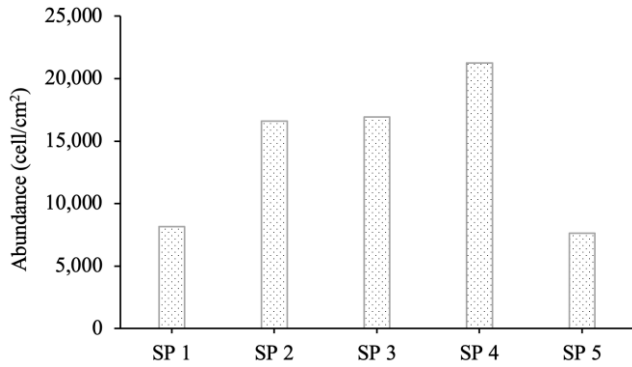
increased light intensity, and elevated dissolved nutrients in the water column. Temperature is a key factor in algal biomass development, as it regulates cellular metabolism and thereby optimizes growth rates. Highlight availability is a consequence of the loss of riparian canopy along streams, which not only drives higher biomass but also changes in composition (Pacheco et al. 2022).

Sampling Point (SP) 5 has a low abundance, which can be attributed to the higher water depth compared to other sampling points and the lower brightness of the water, which tends to be more turbid. The water depth may influence light availability for periphyton growth. Total Suspended Solid (TSS) and high turbidity also further reduce light availability (Baure et al. 2022). In addition, the rock substrate in SP 5 differs from that in other SPs, which is thought to cause variations in the abundance of periphyton. The type of substrate plays a crucial role in determining the abundance, composition, and distribution of periphytic algal communities. The morphology (e.g., overall body architecture, shape) and anatomy (e.g., surface microtopographic smoothness or roughness) of the substrate are the primary reasons for the abundance of periphytic algae (Peng et al. 2023). The high number of periphyton-eating organisms can also cause abundance. Aquatic herbivorous organisms, such as aquatic insects, crustaceans, and fish, can significantly reduce algal biomass in rivers by utilizing periphyton communities as their natural food source (Kiffer et al. 2018).

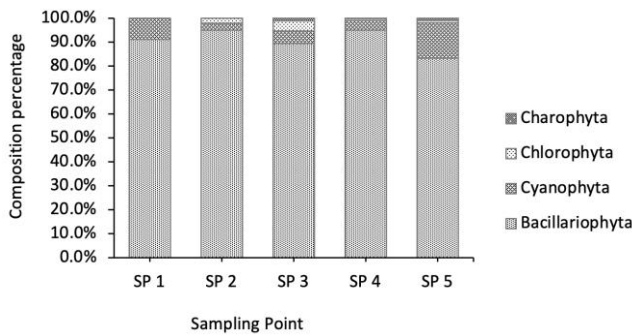
Based on the research results, the highest composition of epilithic microalgae at all sampling points was from the Bacillariophyta division, ranging from 83 to 95% (Figure 4). The high composition of Bacillariophyta is most likely due to its adaptation to many environmental conditions and ecological tolerance (Hu et al. 2022). Bacillariophyta is a group of periphytic algae that is dominant in shallow-water ecosystems. There are two reasons why this type of periphytic algae composition is abundant in waters. First, most diatom species possess specialized, pillow-like, neck-like, or tubular structures that enable them to attach to substrates, such as those in the genera *Gomphonema*, *Frustulia*, and *Navicula*, which confer a competitive advantage over other species in stressful environmental conditions. Therefore, Bacillariophyta (diatoms) are a relatively common group in periphytic algal communities (Peng et al. 2023). Bacillariophyta is a cosmopolitan group distributed across various aquatic environments, and it exhibits a remarkable ability to adapt quickly, displaying a wide tolerance (Arfiati et al. 2023). The Bacillariophyta division can survive in the worst environmental conditions by producing more mucus as a form of adaptation. Bacillariophyta possesses tools such as stalks, which enable them to attach to specific substrates (Ogi et al. 2022). The lowest percentage of epilithic microalgae abundance from the 5 sampling points, specifically the Charophyta division, was 0-1.1%. This is because the Charophyta is only able to live in unpolluted waters and is less able to adapt to environments that have high environmental pressure. In general, Charophyta is recognized as a reliable indicator of water quality (Bučas et al. 2019).



**Figure 2.** A genus of epilithic microalgae found in Setail River, Banyuwangi District, East Java, Indonesia. A. *Achnanthes*; B. *Achnanthidium*; C. *Amphora*; D. *Aulacoseira*; E. *Coconeis*; F. *Coscinodiscus*; G. *Cyclotella*; H. *Cymbella*; I. *Diatoma*; J. *Diploneis*; K. *Encyonopsis*; L. *Ephitemia*; M. *Fragilaria*; N. *Frustulia*; O. *Gomphonema*; P. *Gyrosigma*; Q. *Isthmia*; R. *Melosira*; S. *Navicula*; T. *Nitzschia*; U. *Pinnularia*; V. *Rhoicosphenia*; W. *Sellaphora*; X. *Stauroneis*; Y. *Surirella*; Z. *Tabellaria*; AA. *Tryblionella*; AB. *Anabaena*; AC. *Oscillatoria*; AD. *Chlorella*; AE. *Sphaerocystis*; AF. *Chlosterium*; AG. *Cosmarium*; AH. *Hyaloteca*

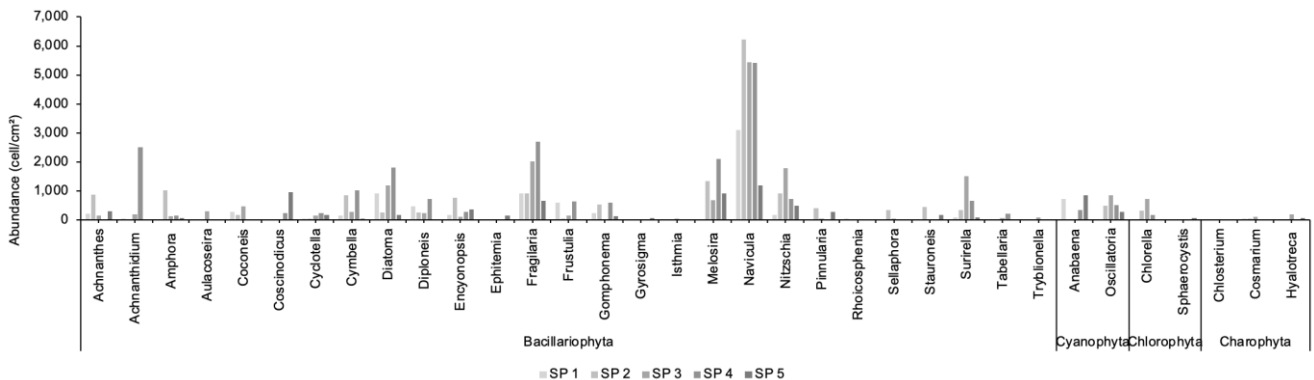


**Figure 3.** Mean relative abundance of microalgae epiphytic based on division in Setail River, Banyuwangi District, East Java, Indonesia



**Figure 4.** Composition percentage of epilithic microalgae based on division in Setail River, Banyuwangi District, East Java, Indonesia

The abundance of epilithic microalgae by genus is listed in Figure 5. *Navicula* was found to be the dominant genus at all sampling points except at SP 5. Analysis of the abundance of the *Navicula* genus, which was the dominant genus at all sampling points, ranged from 1,195-6,225



**Figure 5.** Mean relative abundance of epilithic microalgae based on division in Setail River, Banyuwangi District, East Java, Indonesia

cells/cm<sup>2</sup>, followed by *Fragillaria* (662-2,709 cells/cm<sup>2</sup>, *Melosira* (688-2,103 cells/cm<sup>2</sup>), *Diatoma* (183-1,812 cells/cm<sup>2</sup>) and *Nitzschia* (166-1,793 cells/cm<sup>2</sup>). This genus demonstrates the ability to adapt to various environmental conditions, indicating that this microalgae species has a high tolerance for changes in water quality, including pollution (Arumugham et al. 2023). Several microalgae genera used as indicators of pollution include *Navicula*, *Nitzschia*, *Cymbella*, and *Melosira*. These genera serve as indicators of organic pollution, where reliance on a single species or genus can lead to increased complexity (Palmer 1996; Arumugham et al. 2023). The genera *Cocconeis* and *Cylotella* are dominant around the sides of rivers where the water flow is slow. *Navicula*, *Achnanthes*, and *Nitzschia* are found to be dominant in waters with fast water flow, such as rivers (Everest and Aslan 2016). Microalgae of the genus *Pinnularia*, *Sirurella*, and *Navicula* have a high tolerance and thrive in acidic waters. Diatoms generally dominate most epilithic algae because highly motile diatom species, such as *Navicula*, can adhere strongly to hydrophobic surfaces (Chen et al. 2019).

**Diversity, evenness, and dominance index**

Epilithic microalgae at the five sampling points had a diversity index ranging from 1.65 to 2.17 (medium, H' 1-3), a uniformity index of 0.75-0.80 (e>6, high), and a dominance index ranging from 0.17 to 0.33 (low, C close to 0). The indices of diversity, uniformity, and dominance of Setail River epilithic microalgae are presented in Table 3.

Based on diversity criteria, the results of the epilithic microalgae diversity index at all sampling points have biota stability classified as moderate, as indicated by an H' value of 1-3 (Odum 1971). The diversity index value typically indicates a balanced ecosystem and moderate ecological pressure, suggesting that neither dominant nor minority species were observed in the study. Meanwhile, a low diversity index value indicates that the environment is experiencing disturbance, and the structure of the organisms within it is stressed (Shabrina et al. 2021).

**Table 3.** Diversity, evenness, and dominance index of epilithic microalgae in Setail River, Banyuwangi, East Java, Indonesia

| Sampling point | Diversity index (H') | Evenness index (E) | Dominance index (C) |
|----------------|----------------------|--------------------|---------------------|
| 1              | 1.70                 | 0.78               | 0.27                |
| 2              | 1.65                 | 0.75               | 0.33                |
| 3              | 2.17                 | 0.79               | 0.17                |
| 4              | 2.00                 | 0.80               | 0.20                |
| 5              | 1.91                 | 0.78               | 0.23                |

The results of calculating the uniformity index (E) of epilithic microalgae in the Setail River yielded values ranging from 0.75 to 0.80. The results of the uniformity index at all sampling points are classified as high species uniformity as they have an E value  $>0.6$ , which indicates high species uniformity (Odum 1971). If the uniformity index value is relatively high at each station, then the presence of each type of biota in the waters is evenly distributed. A uniformity index that is small or close to zero tends to indicate an unstable community, meaning that the distribution of individuals across species is not uniform, and a particular species tends to dominate the population (Arfiati et al. 2024). The low evenness value of epilithic microalgae populations reflects the uneven distribution of individuals with low dominance, such that only organisms that can survive are found, and vice versa (Yunandar et al. 2023).

The results of calculating the dominance index (C) of epilithic microalgae in the Setail River obtained values ranging from 0.17-0.33. The dominance index at all sampling points has a value of  $0.5 < C < 1$ , indicating that a single genus dominates (Odum 1971). The low value of the dominance index is due to the even number of species obtained, with no single species being dominant (Meisaroh et al. 2019). A low dominance index value indicates that the aquatic ecosystem is in good condition (Martsenyuk et al. 2016). The dominance of each sampling point is inconsistent, which may be related to environmental, physical, and chemical factors, such as current conditions, nutrients, and light (Ren et al. 2021).

### Saprobic index

Analysis of the Saprobic Index of epilithic microalgae in the upper Setail River yields values ranging from 0.60 to 1 at the five observation points. In general, the five sampling points have water saprobic index values that tend to be the same. There were no significant differences in the saprobic index values at each sampling point (Table 4). The saprobic index value in the Setail River indicates that the waters are lightly polluted ( $\beta$ -mesosaprobic), with a value ranging from 0.6 to 1. Water bodies can be categorized as lightly polluted ( $\beta$ -meso saprobic) if the saprobic index value ranges from 0.5 to 1.5 (Drescher and van der Mark, 1976). The  $\beta$ -mesosaprobic category indicates that only small amounts of pollutants enter from organic and inorganic materials. Organic matter pollution is related to the content of nitrates and phosphates, as these nutrients are essential for microalgae reproduction (Taher et al. 2022; Asmarani et al. 2024). Until now, the Setail River has been used for various human activities, including washing and

bathing, which may have contributed to light pollution at several sampling points. The use of rivers as a venue for human activities can lead to the pollution of lake water by anthropogenic waste (Ramadhan et al. 2016).

Based on table 4 to obtain the value of the periphyton saprobic index, it is grouped into 4 groups. The results of observations and identification only obtained 2 groups, namely group B: Number of Cyanophyta species groups (Cyanophyceae class), Euglenophyta ( $\alpha$ -mesosaprobic) and C: Number of Chlorophyta species groups (Chlorococcales order), Chrysophyta (Bacillariophyceae/Diatoms class). While group A: Number of Ciliates species groups and group D: Number of Peridinians + Chrysophyceae + Conjugate species groups were not found in samples taken during the study. The absence of these groups can be caused by many factors, including the unsuitability of aquatic environmental conditions.

The lowest saprobic value was found at the SP 5; this was caused by a large amount of organic and inorganic pollutant input originating from previous (upstream) streams, such as washing, bathing, rice field activities, plantations, and other activities that entered the river waters and are carried to the central part, causing anthropogenic pollution and altering the structure of periphytic microalgae communities, as well as the types of microalgae that are resistant to environmental changes and will survive in these waters. Saprobic levels are strongly influenced by the proximity of water bodies to residential areas, sedimentation, and the presence of organic and inorganic pollutants that enter the waters (Lutfiana 2022). Sampling point SP5 and SP1 have low saprobic index value, may be attributed to the high phosphate content, which can originate from rock weathering. Weathering and erosion of rocks gradually release phosphorus as water-soluble phosphate ions. Most phosphate is washed into natural water from the leaching process. Plants and algae use phosphate as a nutrient for growth. Stunted and excessive growth of plants and algae has been linked to deficiencies and excesses of phosphate ions (Tafesse 2012). The main source of phosphorus is phosphate rock or other rock deposits (Mabagala and Mng'ong'o 2022).

### Environmental condition

The results of temperature measurements in Upper Setail River ranged from 23°C to 29.1°C. Water temperature is another major environmental factor in aquatic ecosystems, and it can have a significant impact on aquatic communities. This is because water temperature influences the productivity and metabolism of river microorganisms, invertebrates, and fish (Bondar-Kunze et al. 2021). Regarding the influence of temperature, most algal species adapt their photosynthetic mechanisms. An increase in water temperature increases the rate of photosynthesis, while a decrease in temperature causes changes in enzyme activity and reduces the concentration of chlorophyll-a (Bondar-Kunze et al. 2021). In general, algal species diversity increases from 0°C to 25°C and decreases at temperatures  $>30^\circ\text{C}$ , whereas biomass increases with temperature from approximately 0 to 30°C and then decreases from 30-40°C. Algal growth is limited

to below 55°C, while the upper-temperature limit for algal growth is 68°C (Jonker et al. 2013).

The current speed in the upper Setail River at SP 1-SP4 has a fast current speed (0.66-0.77 m/s), while SP 5 has a slow current speed (0.06±0.04 m/s) (Table 5). Current speeds between 0 to 25 cm/s are classified as slow, 25-50 cm/s are classified as moderate, 50-100 cm/s are classified

as fast, and more than 100 cm/s are classified as very fast (Sumarsih et al. 2016; Amatullah et al. 2024). Flow velocities above 30 cm/s increase shear stress and influence periphytic organisms, taxonomic composition, and nutrient cycling. Increased flow velocity also reduces the potential for colonization and increases exfoliation and abrasion of newly growing cells (Bondar-Kunze et al. 2021).

**Table 4.** Saprobic index of microalgae epilithic in Setail River, Banyuwangi, East Java, Indonesia

| Genus                 | SP 1         | SP 2          | SP 3          | SP 4          | SP 5         |
|-----------------------|--------------|---------------|---------------|---------------|--------------|
| Group A               |              |               |               |               |              |
| -                     |              |               |               |               |              |
| Group B               |              |               |               |               |              |
| <i>Anabaena</i>       | 719          | 0             | 0             | 344           | 850          |
| <b>Total</b>          |              |               |               |               |              |
| Group C               |              |               |               |               |              |
| <i>Achnanthes</i>     | 217          | 885           | 160           | 31            | 299          |
| <i>Achnantheidium</i> | 51           | 0             | 198           | 2,514         | 0            |
| <i>Amphora</i>        | 0            | 1,014         | 139           | 157           | 78           |
| <i>Aulacoseira</i>    | 0            | 0             | 299           | 34            | 0            |
| <i>Coconeis</i>       | 279          | 166           | 481           | 0             | 0            |
| <i>Coscinodiscus</i>  | 0            | 0             | 0             | 242           | 956          |
| <i>Cyclotella</i>     | 51           | 55            | 161           | 241           | 168          |
| <i>Cymbella</i>       | 152          | 846           | 280           | 1,027         | 56           |
| <i>Diatoma</i>        | 909          | 253           | 1,189         | 1,812         | 183          |
| <i>Diploneis</i>      | 471          | 267           | 239           | 737           | 31           |
| <i>Encyonopsis</i>    | 166          | 760           | 120           | 275           | 370          |
| <i>Ephitemia</i>      | 0            | 0             | 0             | 0             | 162          |
| <i>Fragilaria</i>     | 912          | 908           | 2,028         | 2,709         | 662          |
| <i>Frustulia</i>      | 595          | 54            | 162           | 647           | 26           |
| <i>Gomphonema</i>     | 242          | 535           | 0             | 608           | 130          |
| <i>Gyrosigma</i>      | 0            | 0             | 0             | 0             | 78           |
| <i>Isthmia</i>        | 0            | 0             | 40            | 0             | 0            |
| <i>Melosira</i>       | 0            | 1,344         | 688           | 2,103         | 915          |
| <i>Navicula</i>       | 3,101        | 6,225         | 5,429         | 5,422         | 1,195        |
| <i>Nitzschia</i>      | 166          | 919           | 1,793         | 728           | 495          |
| <i>Pinnularia</i>     | 0            | 410           | 41            | 0             | 282          |
| <i>Rhoicosphenia</i>  | 55           | 0             | 0             | 0             | 0            |
| <i>Sellaphora</i>     | 0            | 353           | 41            | 34            | 0            |
| <i>Stauroneis</i>     | 0            | 442           | 0             | 0             | 173          |
| <i>Surirella</i>      | 84           | 334           | 1,516         | 654           | 99           |
| <i>Tabellaria</i>     | 0            | 0             | 60            | 207           | 0            |
| <i>Tryblionella</i>   | 0            | 0             | 79            | 0             | 0            |
| <b>Total</b>          | <b>7,450</b> | <b>15,770</b> | <b>15,142</b> | <b>20,185</b> | <b>6,359</b> |
| Group D               |              |               |               |               |              |
| -                     |              |               |               |               |              |
| <b>Saprobic Index</b> | <b>0.71</b>  | <b>1.00</b>   | <b>1.00</b>   | <b>0.95</b>   | <b>0.60</b>  |

**Table 5.** Environmental conditions of Setail River, Banyuwangi, East Java, Indonesia

| Parameters                        | SP 1       | SP 2       | SP 3       | SP 4       | SP 5       |
|-----------------------------------|------------|------------|------------|------------|------------|
| Temperature (°C)                  | 23±1.55    | 25.1±2.69  | 26.3±1.60  | 26.9±1.35  | 29.1±1.91  |
| Current velocity (m/s)            | 0.71±0.01  | 0.77±0.00  | 0.77±0.00  | 0.66±0.14  | 0.06±0.04  |
| Brightness                        | 100%       | 100%       | 100%       | 100%       | 30-70 cm   |
| pH                                | 7±0.00     | 7±0.00     | 7±0.00     | 7±0.00     | 7±0.00     |
| Dissolved Oxygen (DO) (mg/L)      | 7.05±1.24  | 6.63±1.49  | 6.73±1.13  | 6.50±1.32  | 6.00±0.72  |
| CO <sub>2</sub> (mg/L)            | 25.57±1.11 | 47.94±1.02 | 41.55±1.34 | 28.77±1.24 | 51.14±0.82 |
| Nitrate (mg/L)                    | 0.014±0.01 | 0.019±0.01 | 0.016±0.01 | 0.015±0.01 | 0.021±0.01 |
| Phosphate (mg/ L)                 | 0.086±0.06 | 0.085±0.06 | 0.059±0.04 | 0.068±0.04 | 0.107±0.05 |
| Ammonia (mg/L)                    | 0.063±0.02 | 0.071±0.03 | 0.099±0.08 | 0.067±0.05 | 0.142±0.18 |
| Total Organic Matter (TOM) (mg/L) | 17.20±7.53 | 20.96±9.49 | 19.90±9.61 | 13.87±7.67 | 9.55±4.18  |

The results of pH measurements in the Setail River at all sampling points were 7. The pH value obtained did not fluctuate; this result is considered good for water and the growth of aquatic organisms, such as microalgae. The optimal pH of each algal species varies depending on the specific physiological reactions of the microalgae to variations in environmental pH. A cell's ability to survive these pH variations depends on its ability to adapt its physiology to the pH gradient. Several studies have shown that the optimal pH for maximum growth of microalgae is located around pH 7.0-7.6 (Filali et al. 2021)

Dissolved Oxygen (DO) in the Upper Setail River has values ranging from 6-7.05 mg/L. The dissolved oxygen (DO) value that can support the life of aquatic organisms is  $\geq 5$  mg/L (Bianingrum et al. 2020). According to Iswanto et al. (2015), dissolved oxygen levels also vary seasonally and daily due to mixing and movement (turbulence) of water masses, photosynthetic activity, respiration, and waste (effluent) entering water bodies. The dissolved oxygen obtained in this research is considered optimal for the growth of aquatic organisms.

Nitrate values at all sampling points ranged from 0.014-0.021 mg/L. A suitable nitrate concentration level for microalgae growth is typically between 0.01 and 5 mg/L (Sibarani et al. 2020; Santoso et al. 2024). If nitrate levels in water exceed optimal levels, it can have a negative impact on organisms, such as microalgae and aquatic life, potentially leading to algal blooms. The phosphate value in the Setail River ranges from 0.068 to 0.107 mg/L. The range of orthophosphate that is good for microalgae growth is 0.09 to 1.8 mg/L. Levels below 0.004 mg/L of orthophosphate compounds function as a barrier; however, at levels above 1.0 mg/L, phosphate can cause blooming (Iswanto et al. 2015).

The ammonia value at all sampling points ranged from 0.063-0.142 mg/L. Ammonia concentration, according to PP RI No. 22 of 2021, is 0.3 mg/L (Muhaemin et al. 2023). Ammonia is a compound produced from the metabolism of aquatic organisms and utilized by aquatic autotrophic organisms (microalgae) as a source of nitrogen. Total Organic Matter (TOM) values at 5 sampling points ranged from 9.55 to 20.96 mg/L. The TOM value in this study remains optimal for the life of epilithic microalgae, with a range of 5.43-29.83 mg/L. The high TOM is caused by high human activity (Dianto et al. 2020).

In conclusion, the Setail River headwaters are characterized by an epilithic microalgae community structure and a saprobic index, which is classified as  $\beta$ -mesosaprobic in lightly polluted waters. The highest abundance of epilithic microalgae, specifically the genus *Navicula* (Bacillariophyta), exhibits high tolerance to variations in water quality, including pollution. However, overall, the water quality in the upper Setail River can support the aquatic ecosystem. These rivers require regular maintenance and monitoring to maintain their water quality, ensuring they can be used sustainably.

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