

MICROPLASTIC CHARACTERISTICS IN *Amphibalanus amphitrite* AS A POTENTIAL BIOINDICATOR IN THE WATERS AROUND THE SUROBOYO BRIDGE

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ABSTRACT

Microplastics are small particles resulting from plastic degradation, typically ranging from 0.001 mm to less than 5 mm in size. This study aimed to determine the visual and polymer or chemical characteristics of microplastics found in barnacles *Amphibalanus amphitrite* collected from the waters surrounding the Suroboyo Bridge, as well as to evaluate their potential as a bioindicator species for microplastic pollution. A quantitative descriptive approach was applied, including visual identification of microplastics using a stereo microscope and chemical characterization using *Attenuated Total Reflectance Fourier Transform Infrared (ATR-FTIR)*. Barnacle samples were categorized into two size groups, namely small and large, to examine the relationship between barnacle size and microplastic characteristics. The average shell size of the large group was 2.88 cm, while the small group averaged 1.24 cm. The results showed that the average abundance of microplastics in the soft tissues of large barnacles was 61.93 particles per individual, while small barnacles contained 26.3 particles per individual. The dominant visual characteristics of microplastics in both groups were fragment shapes, blue coloration, and particle sizes ranging from 10 to 50 μm . Polymer analysis identified *polypropylene* and *nylon* as the main polymer types. Furthermore, this study highlights a clear relationship between barnacle size and microplastic accumulation, where larger individuals tend to accumulate higher quantities of microplastics. However, the size distribution of microplastics, ranging from 10 to 50 μm , was relatively consistent across both barnacle size groups, indicating that particle size is not strongly influenced by barnacle size but rather by environmental availability. These findings support the potential of *Amphibalanus amphitrite* as a bioindicator species for microplastic pollution due to its sessile nature and filter-feeding mechanism, which enable the accumulation of microplastics over time.

Keyword: *A. amphitrite*, ATR-FTIR, Bioindicator, Microplastic, Suroboyo Bridge

Introduction

Plastic has become one of the most widely used materials since the 1950s, particularly for packaging purposes. The increasing demand for plastic has led to a significant rise in global plastic production [1,2]. Due to its durability and resistance to degradation, plastic waste persists in the environment and accumulates along coastlines, on the water surface, within the water column, and in marine sediments [3]. Over time, plastic debris undergoes fragmentation into smaller particles, known as microplastics [4]. Microplastics are defined as plastic particles ranging from 0.001 mm to

less than 5 mm in size and are classified into primary and secondary types based on their origin [5–7].

Microplastics are widely distributed in marine environments, including coastal areas, open oceans, deep-sea sediments, and even remote islands [8–10]. Rivers act as major transport pathways, carrying microplastics from terrestrial sources to coastal waters. In addition, intensive human activities in coastal regions significantly increase the input of microplastics into marine ecosystems [11]. One area with high potential for microplastic contamination is the waters surrounding the Suroboyo Bridge. This area is influenced by various anthropogenic activities, including domestic discharge, coastal tourism, and

industrial waste inputs, which contribute to the accumulation of plastic debris and its subsequent fragmentation into microplastics [12,13]. Therefore, this location represents a relevant site for assessing microplastic contamination and its potential impacts on marine organisms. The presence of microplastics in marine environments poses potential risks to aquatic biota [14]. Microplastics can be ingested and accumulated by various organisms, including crustaceans, mollusks, echinoderms, and other macrobenthic groups [2]. Among these, barnacles (*Amphibalanus amphitrite*) are considered particularly suitable for microplastic studies due to their sessile lifestyle, filter-feeding mechanism, and ability to accumulate particles from the surrounding environment. As sessile organisms permanently attached to hard substrates, barnacles can reflect site-specific environmental conditions over extended periods. In addition, their feeding strategy increases the likelihood of microplastic uptake, making them effective organisms for both bioindicator and biomonitoring purposes [15,16].

Previous studies have shown that barnacles tend to accumulate higher amounts of microplastics compared to other coexisting organisms such as gastropods, crabs, and bivalves [18]. This further supports their suitability as model organisms for assessing microplastic pollution. However, studies focusing on both the physical characteristics and polymer composition of microplastics in barnacles remain limited, particularly in relation to their accumulation patterns across different size classes. Therefore, this study aims to analyze the visual characteristics and polymer types of microplastics in *A. amphitrite* collected from the waters around the Suroboyo Bridge, as well as to evaluate their potential as bioindicator organisms. In addition, barnacles were classified into different size groups to examine the relationship between organism size, microplastic accumulation, and polymer characteristics, providing a more comprehensive understanding of microplastic exposure and its ecological implications.

Methodology

Time and Place

This research was conducted from January 2025 to July 2025. Barnacle (*A. amphitrite*) samples were collected in the waters around the Suroboyo Bridge using a handsorting sampling mechanism. Barnacle sample preparation and visual observations of

microplastic characteristics were conducted in the Ecology Laboratory, Department of Biology, Faculty of Science and Data Analytics, Sepuluh Nopember Institute of Technology, Surabaya. Microplastic chemical analysis was conducted in the Industrial Biotechnology Laboratory, Department of Industrial Chemical Engineering, Faculty of Vocational Studies, Sepuluh Nopember Institute of Technology, Surabaya. The following is a map of the sampling area.

Tools and Materials

The tools used in this research were a cool box, beaker glass, Buchner funnel, digital caliper, digital balance, oven, petri dish, pipette, measuring cylinder, stereo microscope, Optilab, Fourier transform infrared (FTIR), SPSS software, Optilab Viewer 2.2 software, Image Raster 3 software, Microsoft Excel 365 software, and Tableau Public 2025.2 software. The materials used were barnacles (*A. amphitrite*), aluminum foil, Whatman filter paper number 42 with a pore size of 2.5 μ m, 30% H₂O₂ solution, and distilled water.

Work Procedure

Barnacle samples were collected in the waters around the Suroboyo Bridge. Barnacle samples were collected by hand sorting and using a putty knife. The samples were then placed in ziplock bags. The ziplock bags were then labeled with the sampling location and date of collection. The samples were stored in a cool box for temporary storage, before being transported to the laboratory.

Morphometric measurements were taken, including the diameter, height, and weight of the barnacle's soft body using digital calipers and an analytical balance. Sample preparation was then carried out by separating the tissue from the shell, followed by tissue digestion with 30% H₂O₂, and oven drying at 60°C. The dissolved samples were filtered using Whatman paper number 42.

Visual character analysis of microplastics was performed using a 4.5X stereo microscope connected to an optilab to identify shape, color, and size, and calculate microplastic abundance using the formula:

$$K = Ni / N$$

where K represents microplastic abundance, Ni represents the number of microplastics, and N represents sample weight.

Polymer character analysis was then performed using ATR-FTIR to identify the type of plastic polymer based on its infrared absorption spectrum.

Microplastic abundance data was analyzed descriptively and quantitatively. Statistical analysis was performed using SPSS software.

Result and Discussion

Abundance of Microplastics in Barnacles

The results of the calculation of the total abundance of microplastics in the soft body of barnacles are divided into two categories of samples, namely small group barnacles (*A. amphitrite*) and large groups where each sample contains 30 individual barnacles. The abundance of microplastics of these 30 individual barnacles and the average abundance of microplastics in each individual found in each sample category can be seen in table 1.

Table 1 Table of Microplastic Abundance in Barnacles (*A. amphitrite*)

| | Teritip (<i>A. amphitrite</i>) | |
|---|----------------------------------|--------------|
| | Small | Big |
| Total abundance microplastics on 30 individual (partikel) | 1.858 | 789 |
| Flat abundance Rata Microplastics (particle/individual) | 61.93 ± 42.88 | 26.3 ± 21.11 |

The table above shows the abundance of microplastics in 30 individuals for each sample category that the abundance of microplastics in large group barnacles (*A. amphitrite*) is 1,858 with the average abundance of microplastics per individual of 61.93 ± 42.88. Meanwhile, the abundance of microplastics in small group barnacles (*A. amphitrite*) was 789 with the average abundance of microplastics per individual 26.3 ± 21.11.

The abundance of microplastics in barnacles is affected by the level of water pollution, especially in coastal areas with high human activity. As a filter feeder, *Amphibalanus amphitrite* is more susceptible to microplastic accumulation because it filters seawater continuously using cirri. Microplastics that resemble natural foods can be ingested and settle in the digestive tract because they cannot be digested. The sessile nature of barnacles increases the chances of accumulation over time of stay in contaminated locations [3].

This study found that *A. amphitrite* from around the Suroboyo Bridge contained microplastics, with an average of 61.93 particles/individual in the large group and 26.3 in the small group. The dominant microplastics are fragments of 10–50 µm, blue in

color, and composed of *polypropylene* and *nylon* polymers. The differences in polymer types with other studies are due to variations in waste sources and anthropogenic activity. The accumulation of microplastics correlates with barnacle populations due to the feeding mechanism of *filter feeders*. The presence of barnacles reflects the level of microplastic pollution in the coastal environment exposed to human activities [19].

Character Visual

From the observations that have been made, the visual character of microplastics is obtained starting from the shape of microplastics, the color of microplastics and the size of microplastics as follows:

Microplastic Shape

The results of observations show that there are two forms of microplastics found in barnacles, namely fragment and fiber form. Fragment-type microplastics have thick physical characteristics, have irregular shapes, and sharp edges and have a smooth single end [20]. Fiber microplastics have the characteristics of a thin, long, and synthetic fiber-like shape. This type of microplastics comes from the degradation of fishermen's fishing gear waste such as fishing gear and nets [21]. The abundance of each form in each species can be seen in figure 1.

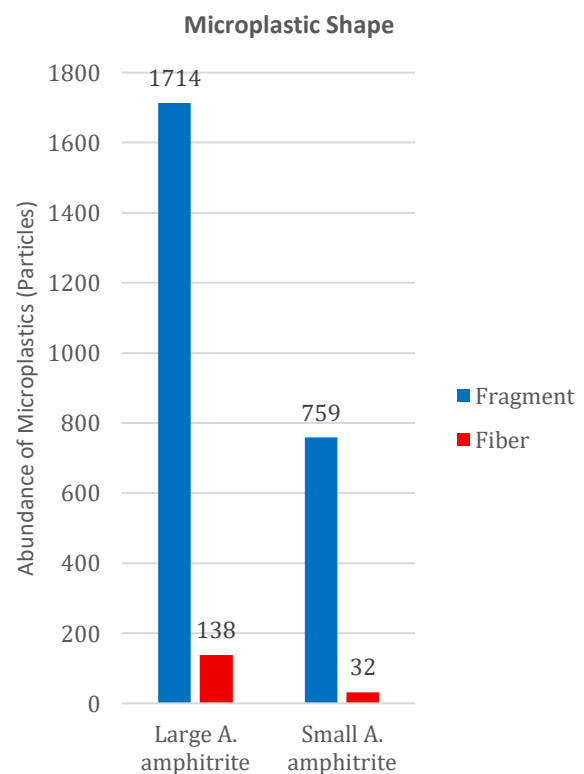


Figure 1. Diagram of the total abundance of microplastics based on their shape in the Suroboyo Bridge barnacle species.

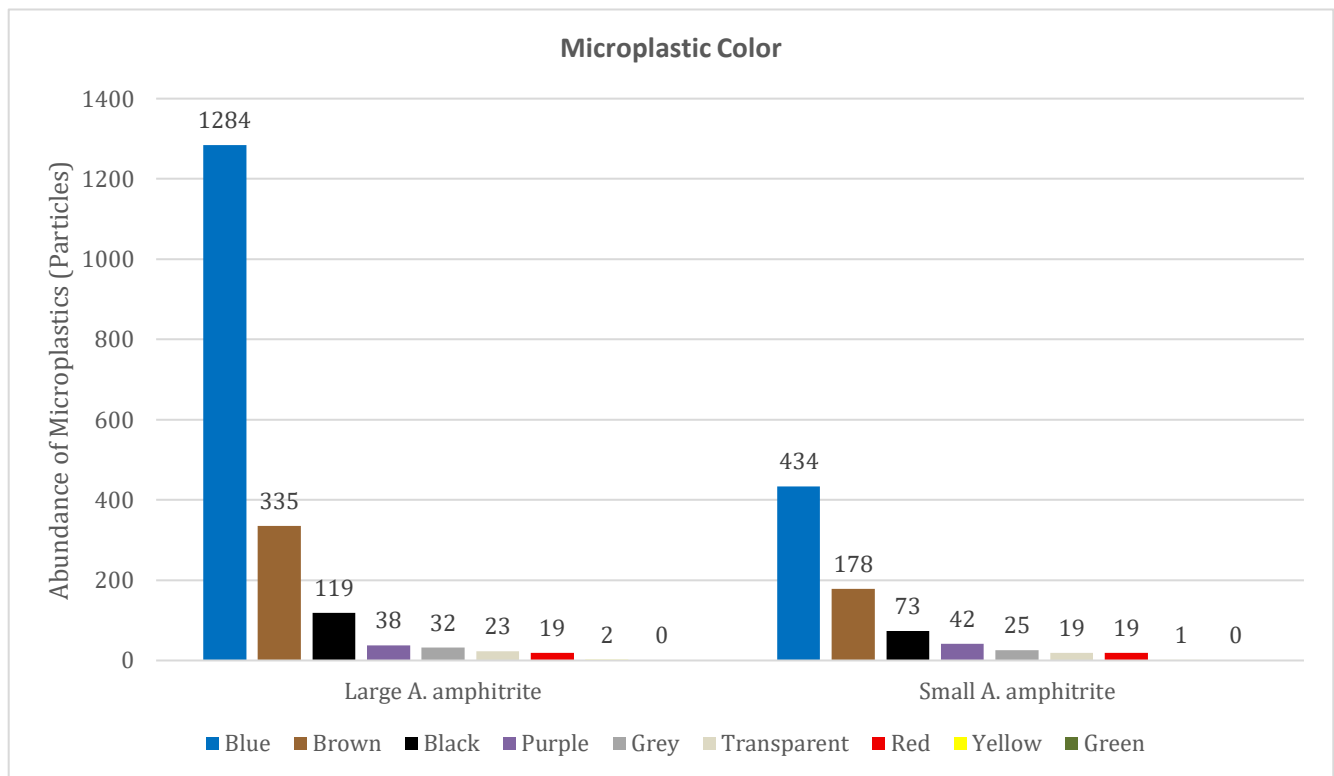


Figure 2. Diagram of the total abundance of microplastics based on their color in the Suroboyo Bridge barnacle species.

Based on the results of observation and calculation of microplastic forms in the two categories of barnacle samples above, the most dominant forms of microplastics were found in each of the 30 individuals barnacles (*A. amphitrite*) large groups and small group barnacles (*A. amphitrite*) were fragments with a percentage of 69% (1,714 particles) and 31% (759 particles) respectively. Then the next form found is fiber with consecutive percentages in large group barnacles (*A. amphitrite*) and small group barnacles (*A. amphitrite*) are fiber with a percentage of 81% (138 particles) and 19% (32 particles) respectively. The research located at the Suramadu Bridge showed similar results to this study, namely *fragments*, *fibers*, and *pellets* were found in barnacle-type microplastics (*A. amphitrite*) with the most dominant forms found were fragments and fibers with a percentage of 67% and 30% [19]. Microplastics *fragments* and *fibers* are also the most dominant forms of microplastics found in barnacles in Hong Kong waters with percentages of 3.4% and 95.7% [18]. Microplastics of fragments and *fibers* are most commonly found in barnacles because fragments have dense textured characteristics and can also come from the fragmentation of plastic packaging, such as beverage bottles, plastic bags, and PVC pipes. The small fragment form is easier to enter the body of crustacean group species because it is about the same size as organic particles or other

microorganisms that are food in the crustacean group that are about 1–40 μm in size, such as plankton [22,23]. Meanwhile, the type of fiber has the characteristics of length and color found in the sea, it can also come from household waste, including synthetic fabrics, fishing boat waste, fishing gear such as nets and fishing gear [19]. The dominant forms of microplastic particles found are fragments and fibers that come from secondary sources, such as plastic waste that degrades in the marine environment [24].

Microplastic Color

Visual observations of microplastics identified based on their physical characteristics are color. Microplastic color observation was carried out using optilab viewer 2.2. The abundance of each color in each species can be seen in figure 2.

Based on the results of observation and calculation of microplastic color in both categories of barnacle samples, the dominant microplastic color was found in each of the 30 individuals (*A. amphitrite*) large group and (*A. amphitrite*) small group was blue with percentages of 55% (434 particles) and 69% (1,284 particles) respectively. Then the next dominant color is brown with consecutive percentages in the large group barnacle (*A.amphitrite*) and small group barnacles (*A.amphitrite*) are 18% (335 particles) and 23% (178 particles). The study located at the

Suramadu Bridge showed similar results to this study, namely the dominant color of the microplastics found in the barnacles was blue with a percentage of 78% and brown with a percentage of 14% [19].

The color of microplastics can help identify the source of waste and distinguish it from non-plastic particles. Conspicuous colored microplastics are easier to recognize. Exposure to sunlight triggers oxidation that causes discoloration over time. Microplastics with sharp colors show no significant discoloration due to oxidation [1]. Blue microplastics are the most dominant color found in barnacles, which can come from domestic wastewater waste, such as washing water from residential areas and wastewater management plants [19]. Microplastics with dark colors tend to have the ability to absorb higher pollutants such as PAHs and PCBs, and can also come from PS and PP plastics. Grey or clear microplastics indicate the length of time required in the process of photodegradation by UV rays, this color is generally derived from PE and PP plastics. The yellow color indicates the length of time microplastics have been in the water and are oxidized [25]. Microplastics with green and red colors are suspected to come from packaging bags, detergents, and soaps used by the public. Microplastics with a purple color can come from various sources of plastics that undergo environmental degradation and fragmentation. Purple color is usually obtained from types of plastics such as PS and PP [2].

Microplastic Size

Based on the observation results, in the Figure there are 4 categories of microplastic size found in barnacles (*A. amphitrite*), namely the 0-10 µm category, the 10-50 µm category, the 50-100 µm category, and the >100 µm category [19]. The different types of microplastic sizes are due to the time of fragmentation in the waters, the longer the fragmentation process, the smaller the size of the microplastics. UV radiation and strong ocean waves are some of the factors that can affect the size of microplastics through the fragmentation process [26]. The abundance of each size in each species can be seen in figure 3.

Based on the results of observation and calculation of microplastic size in the two categories of barnacles (*A. amphitrite*) samples above, the most dominant microplastic sizes were found in each of the 30 individuals barnacles (*A. amphitrite*) large groups and small groups barnacles (*A. amphitrite*) were 10-50 µm with percentages of 75% (1,389 particles) and 69% (543 particles) respectively.

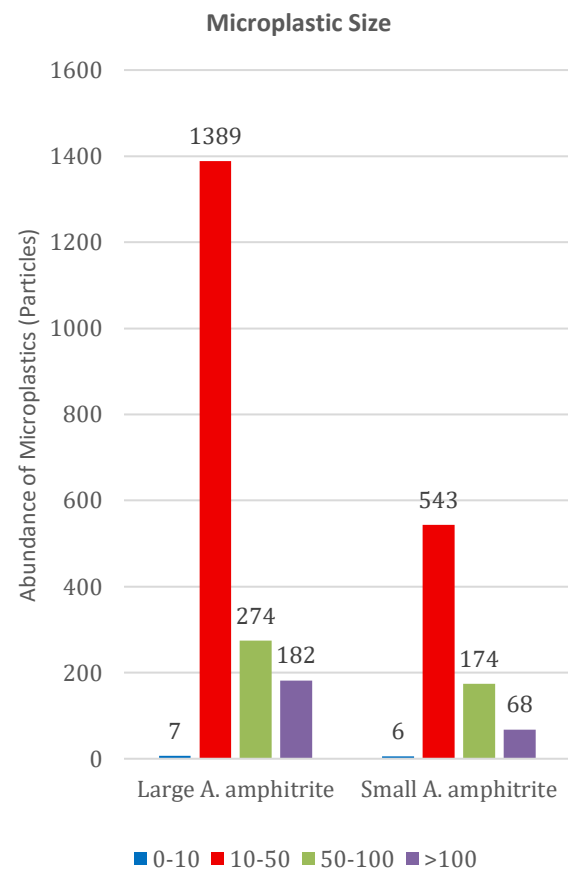


Figure 3. Diagram of the total abundance of microplastics based on their size in Suroboyo Bridge barnacle species.

Furthermore, the most commonly found sizes were 50-100 µm with the percentages of large group barnacles (*A. amphitrite*) and small group barnacles (*A. amphitrite*) being 15% (274 particles) and 22% (174 particles).

The size range of microplastics is significant because it determines their potential impact on biota and ecosystems. Microplastics that are below 40 µm in size will have the same size range as most microplankton and nanoplankton and have a greater potential to be digested by various organisms. In general, the size limits of microplastics can be optionally defined by their sampling and analysis methods [27].

Polymer Characteristics of Microplastics

Attenuated Total Reflectance–Fourier Transform Infrared (ATR-FTIR) is a widely used spectroscopic method for the detection and identification of microplastic particles, particularly for small particle sizes down to 10 µm, providing high resolution and reliable analytical results [28]. In this study, microplastics isolated from the soft tissues of barnacles *Amphibalanus amphitrite* were first

characterized based on their physical properties, including shape, color, and size, as described in the previous section. Subsequently, chemical characterization was performed on representative particles selected from the dominant shape and size categories identified in both small and large barnacle groups to ensure consistency in comparative analysis. The FTIR spectral analysis identified two dominant polymer types, namely *polypropylene* and *nylon*, in all barnacle samples. These results are consistent with the visual characteristics observed, where fragment-shaped microplastics with sizes ranging from 10 to 50 μm were predominant across both size groups. The consistent detection of similar polymer types and particle size ranges in both small and large barnacles indicates that microplastic characteristics are largely influenced by environmental availability rather than barnacle size. However, in line with the findings presented in the abstract, larger barnacles exhibited a higher abundance of microplastics compared to smaller individuals, suggesting that accumulation is more strongly related to organism size and exposure duration rather than differences in microplastic type or size. Overall, these findings reinforce the role of *Amphibalanus amphitrite* as a potential bioindicator species, as it not only accumulates microplastics in significant quantities but also reflects the dominant polymer composition present in the surrounding environment.

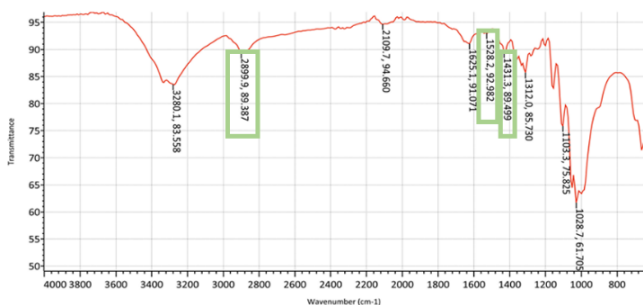


Figure 4. Infrared spectrum FTIR test results on fiber-shaped microplastics on barnacles.

The results of the chemical analysis of microplastics using the FTIR method were translated as the spectrum waves to determine the type of microplastic polymer. The picture above is the result of the analysis of microplastics in the form of fibers on barnacles (*A. amphitrite*) whose polymer is *nylon*. Nylon polymers have chemical compounds (functional groups) O-H and C=C. Nylon polymers can be used for fishing nets and lines [31]. To indicate the presence of nylon polymers, it is with a wavenumber range of 3400-3200 cm^{-1} for N-H and 1680-1620 cm^{-1} for C=C. Fiber microplastics are thought to come from nylon polymers due to the degradation of cigarette filters,

synthetic fibers and fishing nets belonging to fishermen in an area [29].

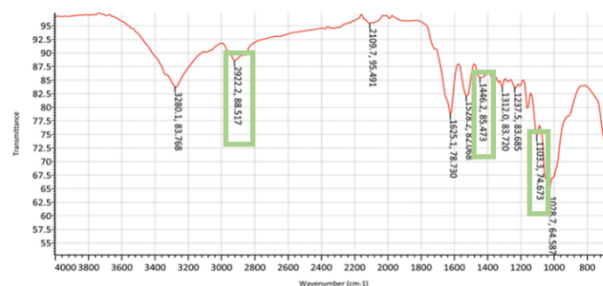


Figure 5. The infrared spectrum of FTIR test results on microplastics in the form of fragments in barnacles.

The results of the chemical analysis of microplastics using the FTIR method were translated as the spectrum waves to determine the type of microplastic polymer. The image above is the result of the analysis of fragment-shaped microplastics on barnacles (*A. amphitrite*) whose polymer is *Polypropylene* (PP). *Polypropylene* (PP) is a type of microplastic found in all parts of the Asian continent. This polymer is shown by the presence of peak absorption of the CH_3 rocking, CH_3 bending, C-H stretching and C-C stretching function groups [30]. The wavelength range of 1105-1107 cm^{-1} indicates the presence of a strain between the C-C or C-H bonds that are part of the PP structure [31]. This type of polymer is usually in the form of fragments, films, fibers, pellets, or foam in environmental samples. *Polypropylene* (PP) has rigid properties, is strong, lightweight, resistant to grease, and is stable at high temperatures, so it is commonly used for food packaging, household appliances, and automotive components [32].

Bioindicators of Microplastics in Barnacles

The results of this study indicate that *Amphibalanus amphitrite* has strong potential as a bioindicator of microplastic pollution in the waters surrounding the Suroboyo Bridge. This potential is supported by its biological characteristics, including a sessile lifestyle, filter-feeding mechanism, and ability to accumulate particles from the surrounding aquatic environment. As a sessile organism attached to hard substrates in coastal areas, barnacles are able to reflect environmental conditions over extended periods. Furthermore, their filter-feeding behavior enables the uptake of suspended particles, allowing microplastics to enter and accumulate in their bodies [19]. The findings also demonstrate an increase in microplastic accumulation with increasing body size of *A. amphitrite*. The larger group exhibited an average abundance of 61.93 ± 42.88 particles per individual,

while the smaller group showed 26.3 ± 21.11 particles per individual. This pattern suggests that microplastic accumulation is more closely related to organism size and exposure duration rather than variations in microplastic characteristics. Consequently, *A. amphitrite* can effectively reflect long-term exposure to microplastic pollution in its habitat [33]. Chemical analysis using the ATR-FTIR method identified two dominant polymer types, namely *polypropylene* in fragment form and nylon in fiber form, which are commonly associated with domestic and industrial waste in coastal areas of Surabaya. *Polypropylene*, with a density of $0.91\text{--}0.94 \text{ g/cm}^3$, tends to remain suspended in the water column, increasing its availability to filter-feeding organisms. Although *nylon* has a higher density of $1.13\text{--}1.15 \text{ g/cm}^3$, smaller particles may still remain in suspension due to hydrodynamic processes, allowing them to be ingested by barnacles [34]. In addition to their role as bioindicators, the accumulation of microplastics may also influence the physiological condition and resilience of barnacles. The presence of microplastic particles in soft tissues can potentially interfere with filtration processes and reduce feeding efficiency, which may lead to physiological stress. Over time, these effects could impact growth, reproductive performance, and the ability of *barnacles* to withstand environmental pressures. Compared to other crustaceans such as shrimp and crabs, which are motile, *A. amphitrite* provides a more reliable representation of local environmental conditions. The mobility of shrimp and crabs introduces variability in habitat exposure, making it more difficult to interpret spatial patterns of microplastic contamination. In contrast, the sessile nature of barnacles allows for a more accurate assessment of site-specific microplastic pollution [35].

Conclusion

This study shows that the average abundance of microplastics in the soft tissues of *Amphibalanus amphitrite* is higher in larger individuals, with 61.93 ± 42.88 particles per individual, compared to 26.3 ± 21.11 particles per individual in smaller individuals. The dominant physical characteristics of microplastics in both groups include fragment shapes, blue coloration, and particle sizes ranging from 10 to 50 μm , while the identified polymer types consist of polypropylene and nylon. These findings confirm that *A. amphitrite* functions not only as a bioindicator, reflecting the presence and characteristics of microplastics in the environment, but also as a

biomonitor, demonstrating the accumulation of microplastics over time in relation to organism size and exposure duration. The presence of microplastics in barnacle tissues also suggests a potential pathway for microplastics to enter the marine food web. Furthermore, the accumulation of microplastics may have negative biological implications, as it can interfere with feeding processes and physiological functions, potentially affecting growth, reproduction, and overall organism resilience. Therefore, this study highlights that microplastic contamination is not only a matter of particle abundance but also a potential ecological concern, as prolonged accumulation in barnacles may influence their lifespan and contribute to broader impacts on coastal ecosystem stability.

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