

## Microplastics in Sediments and Antique Ark Cockles (*Anadara antiquata*) from Surabaya Coastline, East Java

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### Abstract

Microplastic contaminating the environment has become a global issue and is known to be found in marine biota. Surabaya coastline is the main area of community activities, thus contributing to the release of microplastics into the environment. Antique Ark cockle (*Anadara antiquata*) as bivalve is susceptible to microplastic contamination from the surrounding environment. This study aims to determine the abundance and characteristics of microplastics in *A. antiquata* and sediments collected from mangrove-covered area of Gunung Anyar (MGA) and coastal area of Bulak District (BLK). The number of microplastics in cockles was found higher in BLK (5,1 particles/individual) compared to MGA (2,3 particles/individual). Fiber (BLK = 64.13%, MGA = 54.64%) and fragment (BLK = 35.87%, MGA = 45.36%) were found in cockles, fiber was found to be dominating in cockles from both locations. The number of microplastics in sediment was also higher in BLK (361, 31 particles/kg dry weight) compared to MGA (170,82 particles/kg). Fragments (52.45%), fibers (47.19%), and pellets (0.25%) were found in BLK. Meanwhile, the microplastics found in MGA are fragments (51.10%), fibers (46.26%), filaments (2.20%), and pellets (0.44%). Fragments are dominating in both locations. Mann-Whitney test shows a significant abundance between microplastics found at BLK and MGA from sediment and cockles.

**Keywords:** Coastal; Cockles; Mangrove; Microplastic

### 1. Introduction

Microplastics are synthetic polymer particles measuring  $\leq 5$  mm and act as pollutants to the environment and become a threat to marine biotas for decades, becoming a global issue [1]–[3]. It is known that the amount of plastic produced in 2016 reached over 335 million tons per year [1], and between 5-13 million tons were discarded into the sea, with the number continuously increasing and becoming a globally recognized issue [1]–[3]. Approximately 80% of microplastic particles released into the environment originate from anthropogenic activities in terrestrial areas [4]–[8], including household, industrial, and agricultural waste [9] such as fabric fibers, fertilizers, detergents, and other cleaning chemicals [10]; and the remaining smaller amount comes from coastal areas such as port activities, fishing, and recreational activities [1], including fishing nets and lines, ship mooring ropes, degraded plastic bags, ship and boat surface paints, etc. [9]. The waste generated will enter freshwater streams, which will subsequently flow into marine waters [2], [11].

Surabaya is one of the largest cities in Indonesia, with a high population of around 3.1 million people, and facilitated many anthropogenic activities such as fish markets, marine aquacultures, marine transportation, tourism, river discharge, and fishermen's residence [12], [13]. The high population and land usage, along with improper waste management are related to the abundance of microplastics released in the area from terrestrial sources [12], [14], [15].

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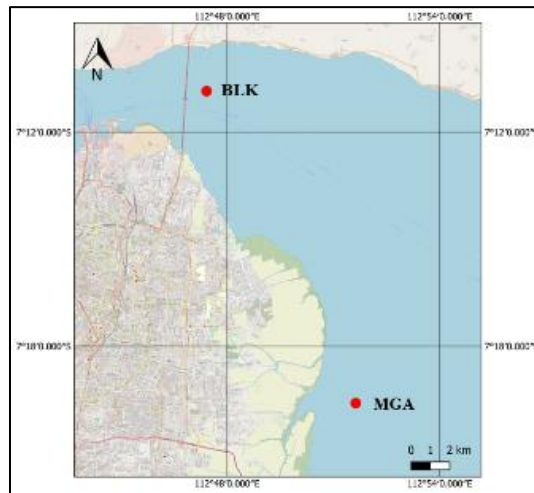
The presence of microplastics in the environment increases microplastic bioavailability towards marine biota, such as bivalves [16]. Bivalves are filter-feeding biota, by filtering large amounts of water, actively capturing small suspended food particles found on the seabed substrate such as detritus [16], [17]. Filter feeder sometimes allows the entry of non-food foreign particles present in their environment, such as microplastics [10], [18]–[20]. The small size of microplastics increases the possibility of accidental ingestion by marine biota [10]. Microplastics have been found in various marine biota, such as cockles [16], [21].

Inhabitant organisms in coastal areas such as Bivalvia are greatly influenced by their surrounding environmental conditions [18]. *Anadara antiquata* can be found dwelling in sediments [22] alongside east coast of Surabaya and feeding as a filter-feeder [16], [22] can be exposed to microplastics present in their surroundings, such as in water bodies or sediments [18]. The small size of microplastics (<5 mm) increases the potential for unintentional ingestion by Bivalvia because of their similar size to their food sources [10], [19]. The abundance of microplastics found in bivalves shows a positive linear correlation to the abundance of microplastics in the water [23] and sediment [16], [24], [25]. Microplastics found in organisms tend to vary and depend on the spatial conditions of their living environment [12], [26], making it important to conduct observations of microplastics in locations with different characteristics. Surabaya coastal area has mangrove-covered areas such as Gunung Anyar (MGA) and non-mangrove-covered areas such as Bulak district (BLK). *Anadara antiquata* can be found alongside the coast both in MGA and BLK. Therefore, this research aims to determine the abundance and types of microplastics found in the Antique Ark (*A. antiquata*) from MGA and BLK.

## 2. Materials and Methods

### 2.1. Study Area and Sample Collection

The coastal area of Surabaya is a border area with the Sunda Strait with diverse coastal activities and high population densities. The study was conducted in two main coastal areas with different characteristics, namely in Bulak sub-district (BLK), which is not covered by mangroves, and Gunung Anyar mangrove area (MGA).



**Figure 1** Study area and sampling location

Sampling was conducted in May 2024. Sediment and shell samples were taken from both locations with three replicates at each site. Each sampling point was determined based on the presence of cockles. Sediment was collected using a bottom grab sampler and placed into containers. Cockles were hand-collected with a total of 45 individuals per location. The cockles used in this study are approximately 5 cm, which is the size of adult cockles. All samples were then placed in a cooling box at a temperature of 4 °C before being transferred to the laboratory. The sediment and cockles that have been obtained will subsequently be stored in a freezer at -20 °C before proceeding to the preparation stage.

During the sample collection, transfer, and preparation, stainless steel and glass tools were used to avoid contamination.

### 2.2. Sample Preparation

The sediment is filtered using a sieve with a mesh size of 4,25 mm to separate it from larger particles prior to drying with an oven at 60 °C until a constant weight is achieved [16], [23]. Each 15-20 grams of dry sediment is placed into a

beaker glass and treated with 20 ml of 30%  $\text{H}_2\text{O}_2$  solution, covered with aluminum foil, due to organic matter destruction process, and incubated for 24 hours at 50 °C [16].

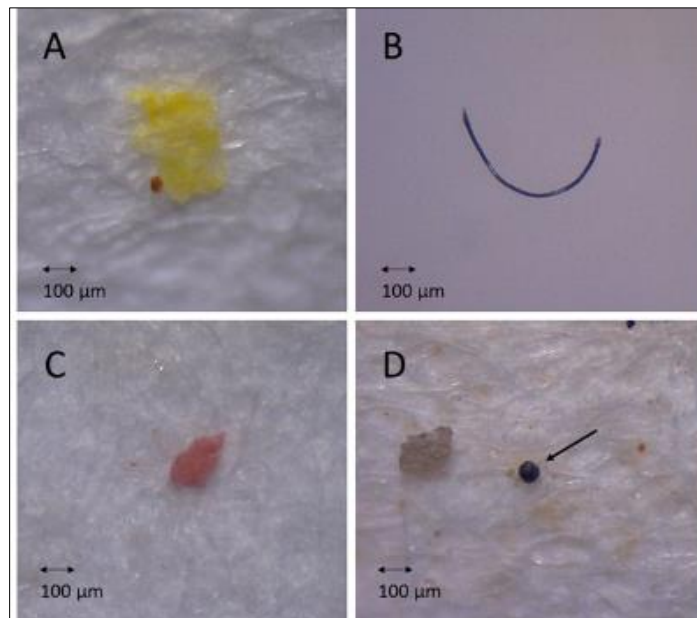
The selected cockles will be separated from their cockles, rinsed with distilled water, and their wet weight will be measured. All the individual tissues of the cockles were placed into a beaker glass and treated with a 30%  $\text{H}_2\text{O}_2$  solution in a 2:1 volume ratio of cockle tissue for organic matter destruction. The glass was then covered with aluminum foil and incubated at 50 °C for 72 hours [23], [27].

### 2.3. Floatation Methode and Filtration Using NaCl Solution

The digestate was dissolved using a NaCl solution of 1.2 g/ml at a ratio of 3:1, then homogenized and allowed to settle for 24 hours [16], [27]. NaCl 1.2 g/ml solution is the most commonly used salt solution in the flotation method because it is environmentally friendly and cost-effective [23]. The supernatant will settle at the bottom of the glass while the microplastic particles will float in the solution [16], [23], allowing for the filtration process using Whatman grade 42 filter paper with a pore size of 2.5  $\mu\text{m}$  under vacuum using a Buchner funnel filtering kit [28]. Thus, each filter paper obtained is stored in a petri dish and dried in an oven for 1 hour at 60 °C [25].

### 2.4. Microplastic Identification and Analysis

Visual observation was conducted using an Olympus stereo microscope (magnification x40) and Optilab viewer 2.2 software to facilitate visual observation, while microplastic measurement was performed using Image Raster 3 software. The shapes observed are categorized into fiber, fragment, foam, pellet, and filament. In addition to observing the characteristics of microplastics, visual observations also include counting the number of microplastics obtained from each individual.



**Figure 2** Variation in microplastic types obtained from both sediment and cockles at both observation locations. A) Filament, B) Fiber, C) Fragment, D) Pellet

During the laboratory work process to prevent the risk of microplastic contamination not originating from cockle meat, all activities are carried out based on widely used standards in microplastic extraction. All testing equipment used is made of glass and aluminum. All solutions are covered with aluminum foil.

### 2.5. Data Analysis and Visualization

The abundance of microplastic characteristics in cockles is presented in the form of the number of particles per gram of meat weight, while microplastic data in sediments will be presented in the form of the number of particles per kilogram of dry weight. Types of microplastics are presented in percentage (%) bar graphs. The obtained data were subsequently tested for normality using the Shapiro-Wilk test. The data were not normally distributed, so the Mann-Whitney test was

conducted to see the difference in values between the Bulak and Gunung Anyar mangrove data groups. Data analysis was conducted using SPSS software version 30.0.0 (SPSS, Inc., Chicago, IL, USA).

### 3. Results

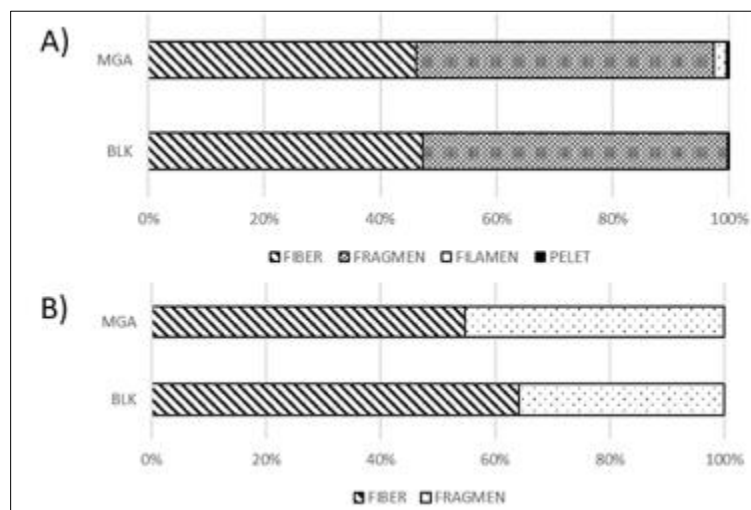
#### 3.1. Microplastics Abundances from Sediments and *Anadara antiquata*

Microplastics were found at all sediment sampling locations. BLK has the highest number of microplastic particles with an average of 361.31 particles/kg. Meanwhile, the number of microplastic particles found in MGA has an average of 170.82 particles/kg. The abundance of microplastics in the sediments at both locations shows a significant difference ( $p\text{-value} = 2,206.10^{-9}$ ,  $p \leq 0.05$ ).

The cockles obtained in the Bulak area (BLK) had an average of 5.1 microplastic particles per individual, with a range of 2-8 microplastic particles per individual. Meanwhile, the cockle obtained in the Gunung Anyar mangrove area (MGA) had an average of 2.3 microplastic particles per individual, with a range of 0-4 microplastic particles per individual. The abundance of microplastics in clams at both locations showed a significant difference ( $p\text{-value} = 1.949.10^{-12}$ ,  $p \leq 0.05$ ).

#### 3.2. Microplastics Based on Types

The types of microplastics found in BLK include fragments (52.45%), fibers (47.19%), and pellets (0.25%). Meanwhile, the microplastics found in MGA are fragments (51.10%), followed by fibers (46.26%), filaments (2.20%), and pellets (0.44%) (Figure 2a).



**Figure 3** Microplastics percentages from *A. antiquata* based on types. A) Sediments, B) Cockles

The microplastic types in the cockles were only found in the form of fibers and fragments (Figure 2b). The fiber form dominated in both locations (BLK = 64.13%, MGA = 54.64%) followed by the fragment form (BLK = 35.87%, MGA = 45.36%).

### 4. Discussion

Microplastics found in the BLK area, an urban region without mangroves, show significantly higher quantities compared to those found in the MGA area, which is covered with mangroves. Based on the prominent pneumatophore root system, the mangrove ecosystem acts as an important sink for various types of contaminants [29], [30]. Plastic waste can spread across ocean areas through wave currents and tidal currents [1], [31]. The plastic waste can be trapped in the stems and roots of mangroves that are above ground, preventing it from being carried away by water currents to open waters [29], [30]. Plastic waste can eventually be trapped in mangrove areas, and with its exposure to environmental conditions, macroplastics will eventually degrade into microplastic particles over time [29]. There is no correlation between the amount of microplastic being trapped by the mangrove to microplastic being released into water body as a result of fragmentation [32]. However, it is known that the amount of microplastics in the environment has a positive correlation to the amount of microplastics found in the bodies of biota [16], [23]–[25]. This is because the amount of

microplastics that can be ingested by biota depends on the availability of microplastics in their environment. The more microplastics present in the environment, the higher the possibility of biota ingesting microplastics [33].

The types of microplastics can explain the origin of the plastic debris that produces these microplastics. Fragment shapes are types produced from the fragmentation or fragments of macroplastics [34] made of hard and thick materials [32] such as bottles, jars, plastic bags, and PVC materials [35]. The diverse and irregular shapes of microplastics can affect the density of microplastics in aquatic environments. Microplastics in the form of fibers and filaments have a higher buoyancy, making them more difficult to settle in water bodies and harder to sink, thus they are more easily dispersed to various locations and more likely to interact with various biota in the water body [18].

Fiber form comes from macroplastics that have a fiber-like shape [36], generally originating from fishing nets, rope fibers, and even fabric fiber residues that have been washed away and carried by river currents [25], [29]. Its fiber-like shape allows the fiber to fold into smaller sizes, and it can also adhere to or bind with food particles, making it easier for fiber-shaped microplastics to be ingested. Additionally, because the fragmentation process of fiber-shaped microplastics is longer compared to other types, it leads to the accumulation of fibers over an extended period. Therefore, fiber-shaped particles pose a greater threat to aquatic organisms compared to other types, especially to organisms that feed through filter-feeding, which are morphologically adapted to avoid ingesting large particles such as fragments, etc. [37].

Filament-shaped microplastics have a characteristic thin, film-like shape and low density. Filament-shaped microplastics originate from the fragmentation of plastics such as thin plastic bags or food wrappers, as well as agricultural films [29], [38]. This shape causes filament particles to float more easily in the water column, resulting in a lower quantity in the sediment, leading to a reduced number of filaments ingested by cockles [38].

Pellet-shaped microplastics have a characteristic round shape like spherules. This form is generally produced in small sizes and originates from cleaning products such as scrubs in facial cleansers, toothpaste, and other chemical cleaning liquids [25]. Like filaments, pellet-shaped particles have a low density, making them easy to float in the water column and be carried by water flow [35]. The presence of pellet-shaped microplastics ingested by cockles is difficult to digest, causing them to get stuck or remain in the digestive tract [39].

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## 5. Conclusion

Microplastics were found in sediments and cockles at all locations. The number of microplastics in cockles was found higher in BLK (5,1 particles/individual) compared to MGA (2,3 particles/individual). Fiber (BLK = 64.13%, MGA = 54.64%) and fragment (BLK = 35.87%, MGA = 45.36%) were found in cockles, fiber was found to be dominating in cockles from both locations. The number of microplastics in sediment was also higher in BLK (361, 31 particles/kg dry weight) compared to MGA (170,82 particles/kg). Fragments (52.45%), fibers (47.19%), and pellets (0.25%) were found in BLK. Meanwhile, the microplastics found in MGA are fragments (51.10%), fibers (46.26%), filaments (2.20%), and pellets (0.44%). Fragments are dominating in both locations. This research is the result of observations under current conditions, and with the increasing release of microplastics into the environment over time, ongoing monitoring is necessary. This study also found that microplastics contaminate cockles, which are one of the food sources, thus posing a potential risk to human health.

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## Compliance with ethical standards

### *Disclosure of conflict of interest*

No conflict of interest is to be disclosed.

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