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Identification and Analysis of Microplastic Abundance in Water, Sediment, and Benthic Gastropods in the Upper Reach of the Brugan River, Batu City

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ABSTRACT
Escalating plastic waste generation in Indonesia threatens aquatic ecosystems as macroplastics degrade into persistent microplastics that contaminate water, sediment, and aquatic biota, ultimately entering the trophic food web. This study aimed to determine the abundance and characteristics of microplastics in water, sediment, and gastropods in the upper reaches of the Brugan River, Batu City, and to examine their relationship with water quality. Quantitative descriptive assessment was conducted across three sampling stations representing distinct land-use regimes. Surface water, benthic sediment, and gastropod samples were collected in-situ. Microplastic were extracted by density separation and organic matter oxidation and analyzed using optical microscopy to determine the number, size, shape, and color of the particles. Water quality parameters were also assessed such as temperature, total suspended solids (TSS), turbidity, flow rate, and flow velocity. Results indicated that microplastics were present in every compartment, with the highest concentrations in sediment at 221 particles/kg dry weight (dw), then gastropods at 187 particles/kg dw, and finally water at 3 particles/L. Microplastic particles with fragments, particles less than one millimeter in diameter, and transparent colors were the most prevalent characteristics. The Bioaccumulation Factor (BAF) was 62.33 L/kg and the Transfer Factor (TF) was equal 0.85, confirming substantial microplastic accumulation within benthic biota.

1. INTRODUCTION

The development of the plastic industry has accelerated rapidly in recent years. Global plastic consumption expanded approximately 180-fold between 1950 and 2018. Global plastic production reached approximately 400.3 million metric tons in 2022 (Plastics Europe, 2024), up from roughly 390 million tons in 2021. Following this period, global plastic manufacturing is projected to expand substantially (Pilapitiya & Ratnayake, 2024). Due to the lengthy process required for decomposition, plastic waste presents many problems related to the environment. Because plastic does not decompose rapidly, it accumulates on land and especially in aquatic areas. When plastics enter an aquatic environment, they decompose over long periods of time through various physical, chemical, and biological mechanisms. During this decomposition process, plastics ultimately break down into smaller pieces that are formally referred to as microplastics.

Microplastics are defined as solid plastic particles whose largest dimension is below 5 mm, with a lower bound of about 1 µm in environmental studies (GESAMP, 2019). Within this range, particles are usually separated into small (1 µm to 1 mm) and large (1 to 5 mm) microplastics, a division that also underlies the size classes used here. As a component of aquatic debris, microplastics are considered a greater concern than macroplastics because of their potential to cause serious environmental damage (Suripatty et al., 2023). Microplastics have a high surface area, which favors the sorption of hydrophobic organic pollutants and heavy metals (Hg, Pb, Cr, Cu, Cd, and Zn). This capacity increases as particles weather.

Photo-oxidation introduces oxygen-containing functional groups that make the surface more electronegative (Chang et al., 2024) and microbial biofilm colonization further enhances metal uptake. Weathered microplastics therefore bind divalent metal cations such as PB^{2+}, Cu^{2+}, Cd^{2+}, and Zn^{2+} more strongly than virgin plastics (Mao et al., 2020). Once loaded, these particles can act as vectors that carry the contaminants into the food chain. Microplastic adsorption of heavy metals depends primarily on the physical characteristics of the particles (i.e., pore size, surface area, and polymer type). Microplastics enter aquatic ecosystems through wind currents and human activity nearby, including irrigation networks for agriculture, municipal drinking water intakes, recreation, and illegal dumping (Sugandi et al., 2021). Local populations discard more plastic waste than any other form of waste along riverbanks. Most of this waste from land includes plastic bottles, plastic bags, packaging material, and plastic cups; over time, these items degrade through photo degradation (UV exposure), oxidation, and mechanical abrasion, eventually degrading into microplastics.

Microplastics, which can be found in aquatic systems, do not simply float around in the water; they also sink and accumulate on the bottom of the river. The way microplastics are distributed throughout the aquatic system depends on many different things, such as how dense they are, if they have an electrical charge, how much they tend to clump together, how they interact with other organisms, and how they react to physical changes in the environment (Pradiptaadi & Fallahian, 2022). Microplastics accumulating in the sediments of rivers affect the way the organisms that live in those sediments obtain energy. One example is the organisms that feed on the sediments themselves, such as Gastropods (Fitri & Patria, 2019). Gastropods make up a large portion of the animal community that lives in river environments. These animals use the sediment as their home, where they find food and reproduce (Candri et al., 2020). Gastropods are also important for decomposition of plants and other organic material in the river, and they contribute to the carbon cycle in aquatic systems (Isnaningsih & Patria, 2018). Freshwater gastropods exhibit diverse feeding modes, from scraping and grazing to filtering and detritivory, and Melanoides tuberculata, the taxon recovered in this study, feeds mainly by scraping biofilm and organic detritus from the sediment surface with its radula. This scraping action takes up sedimented particles together with the biofilm, so microplastics deposited in the riverbed are ingested along with food, as reported grazing freshwater snails. Therefore, Gastropods may serve as excellent biological indicators of microplastics sinking into the sediment. When Gastropods eat microplastics, it allows for a way for contaminants to move up the food chain to larger animals and potentially people who consume those animals.

The Brugan River is a sub-watershed located upstream and connected directly to the primary Brantas River sub-watershed; it passes through many different villages in Batu City, East Java. The headwaters of the Brugan River originate at Coban Rondo Waterfall, representing an initially pristine lotic ecosystem that rapidly transitions into zones characterized by intensive anthropogenic activity. In addition to being a localized research site, the Brugan River is also a good example of an ecological system to examine the impact of rapid changes in land use (from highly protected ecotourism upstream to densely populated residential areas and agricultural operations) on microplastic accumulation ratios and spatial patterns in benthic organisms. Microplastic pollution may occur as a result of various intensive anthropogenic activities occurring along the Brugan River, such as agricultural irrigation, livestock farming, domestic use of water resources, and ecotourism.

Based on this rationale, this study is designed to assess the concentrations of microplastics in surface waters and sediments, as well as the morphology of microplastics found in macrozoobenthos gastropods. In addition to quantifying microplastic concentrations and morphology, this study delineated the spatial distribution of plastic contamination across environmental compartments. Synthetic textile fibers are shed in large numbers during domestic laundry and enter rivers through greywater discharge (Napper & Thompson, 2016). We therefore hypothesized that fibrous microplastic whould show a higher relative abundance at Station 2 (residential) than at Station 1 (tourism) and Station 3 (agriculture). We further hypothesized that microplastic accumulation profiles in benthic gastropods would reflect the contaminant characteristics of their host sedimentary substrates. Therefore, if our hypotheses are correct, then gastropods may serve as effective bioindicators of benthic contamination. There were three different sampling sites chosen along an upstream gradient: Station 1 (7°51'47.84" S, 112°30'00.96" E, tourist/lodging zone); Station 2 (7°51'38.84" S, 112°30'17.84" E, residential zone); Station 3 (7°51'33.66" S, 112°30'23.20" E, agricultural/livestock zone).

2. METHOD

2.1 Time and Place

Field sampling was conducted in January 2026 along the upper reaches of the Brugan River (Batu District, Batu City, East Java Province). There were three sampling locations used to represent the different levels of human influence. Location 1 (7°51'47.84" S, 112°30'00.96" E) is primarily characterized by ecotourism and lodging facilities. This location is directly downstream from the very popular Coban Rondo Waterfall tourism area. Location 2 (7°51'38.84" S, 112°30'17.84" E) represents a heavily populated residential area. Location 3 (7°51'33.66" S, 112°30'23.20" E) represents a transition zone that is largely made up of agricultural lands, commercial plantations, and heavy livestock farming.

2.2 Research Methodology

The research design for this study utilizes a combination of descriptive methodology and a quantitative analytical framework. This research design provides a systematic integration of primary and secondary data sets in order to allow for the identification and assessment of empirical evidence. The primary empirical data collected include the exact location coordinates of each sample station, the types of benthic substrate found at each sample station, the chemical composition of both surface water and sediment samples taken from each sample station, macrozoobenthos gastropod samples collected from each sample station, as well as the concentration levels of the microplastics that were extracted from the samples collected from each sample station, along

with their physical characteristics. On the other hand, the secondary data set consisted of geospatial maps that illustrated the designated study area.

2.3 Sampling Methodology

The surface water sampling protocol and identification of exact sampling locations followed all relevant standards for sampling rivers and streams as outlined in the ISO 5667-6:2014 standard. Before beginning the sampling process, the sampling equipment was conditioned to the conditions of the targeted water body. Sampling equipment was rinsed three times with ambient river water before collection. Because this step can retain suspended microplastics on container walls, current guidance recommends pre-rinsing with filtered or reagent water instead (Brander et al., 2020) . Once the sampling equipment had been equilibrated with the surrounding environment, the appropriate volume of water was collected and initially contained in a collection container. The collected water was mixed thoroughly before being transferred into specialized, pre-cleaned container(s) specified by the analytical laboratory performing the testing. At the time of sampling, several on-site environmental parameters (including temperature, turbidity, electrical conductivity, and dissolved oxygen) were determined. These types of environmental parameters cannot be preserved chemically; therefore, they must be determined in-situ. Each of the in-situ determinations made during the sampling event was recorded systematically in a separate field log as supporting information. In addition to recording in-situ determinations, the remaining portions of the homogenized liquid samples were transported back to the laboratory for further analysis.

The sediment sampling was conducted in accordance with the United States Environmental Protection Agency (USEPA, 2001) standard operating procedures. Sampling of the sediment began by deploying either a grab sampler or specialized spade. Only chemically inert devices were used during the sampling procedure to avoid contaminating the sediment matrix. Spades were used as an alternative method of sampling in shallow lotic environments when it was mechanically impossible to deploy the grab sampler. The sampling device was then lowered into the benthic riverbed and pulled up to the surface. After the sampler reached the surface, the top layer of water above the sediment was decanted using a precision siphoning device to minimize loss of fine particulate material. Once the siphoning step was completed, the sediment was placed in chemically inert containers and sealed tightly. Samples were placed in a dark, cold container to prevent premature degradation of the samples and/or contamination from external sources prior to performing laboratory analyses.

The macrozoobenthos gastropod sampling methodology used the USEPA Rapid Bioassessment Protocols developed by Barbour et al. (1998) for single habitat assessment. Manual handpicking was used to remove specimens that were attached to submerged epilithic surfaces, riparian vegetation and the primary benthic substrate in a 1-meter square quadrant. For soft substrates such as mud or sand, mild mechanical agitation was employed to encourage gastropods to become exposed and visible. Biological samples were placed in separate containers containing 100 mL of 70% ethanol, which serves as a universal biological preservative. All containers were labeled with relevant information and were placed in a cooler to keep them structurally sound during transportation. Specimens were rehydrated with water prior to morphological identification for 10-15 minutes. Biological matrixes were viewed under optical magnification and then taxonomically classified using existing macrozoobenthos dichotomous identification keys. Taxonomic classifications were recorded photographically.

2.4 Sample Processing Methodology

Surface water sampling was performed at three different locations on the upstream Brugan River using a non-probability sampling approach that was determined by both the variation in the environment and the proximity of human activity. All sampling procedures adhered to the guidelines of ISO 5667-6:2014. Before collecting the water sample, each bottle was rinsed three times with the surrounding water. Once the water samples were collected, they were placed in a cooler until they could be analyzed in a laboratory setting. First, a wet sieving process was utilized with a 5.6 mm stainless steel sieve to separate and eliminate large debris from the sample. Next, the samples were thermally dried for 24 hours at 90°C (Masura, Baker, Foster, & Arthur, 2015) . A wet peroxide oxidation was used next. A 0.05 M Fe (II) solution and 30% H_2O_2 reagent were added to the sample in a glass beaker placed in an ice bath, and the temperature was monitored to keep the reaction at or below 75°C for 30 minutes (Masura, Baker, Foster, Arthur, et al., 2015). Density separation was employed as well. A saturated NaCl solution (density ≈ 1.2 g/cm^3) was used for density separation, and the mixture was allowed to settle for 24 hours. After incubation, the supernatant was vacuum filtered through Whatman Number 42 filter paper. After filtration, the samples were completely dried. Finally, each sample was analyzed under an Olympus CX33 optical microscope at 40x magnification to count suspected microplastic particles and to categorize them by shape, color, and size. Particle were counted as suspected microplastics only if they showed no cellular or organic structures, had a homogeneous color, and, for fibers, had equal thickness along their entire length (Hidalgo-Ruz et al., 2012).

The process of collecting sediment samples from the three specified locations was accomplished through a purposive sampling approach that considered the unique habitat types and the immediate vicinity of anthropogenic activity. At each station, three separate sediment grabs of approximately 500 g (wet weight) each were collected in shallow water using a stainless-steel shovel. Each sample was placed in an identified ziplock bag and placed inside a cooling container. The wet sedimentary matrices were dried under heat (90°C) for 24 hours (Masura, Baker, Foster, & Arthur, 2015) , crushed into fine particles, and then passed through a mesh size 4- or 5-mm sieve to ensure a uniform mixture of the sediment matrix (Arbabi et al., 2018) . After being dried and processed as described above, a portion (100 grams) of the dried sample was subjected to density separation using a saturated sodium chloride (NaCl) solution. The density separated sample was allowed to settle out of the solution for 24 hours. Once the settled material had fallen out of the solution, the remaining liquid (supernatant) was passed through a 100-mesh sieve. At this point, the samples were subjected to a wet peroxide oxidation treatment. This involved adding 0.05 M iron (II) and

30% hydrogen peroxide (H_2O_2) to the samples and subjecting them to a temperature of 75°C for 30 minutes. After completing the wet peroxide oxidation treatment, the samples were filtered through Whatman No. 42 filter paper under vacuum. When completely dry, the samples were examined microscopically to determine the specific microplastic characteristics present in the sample.

The collection of macrozoobenthos gastropod samples took place in all three sampling coordinates. This process used a purposive methodology that based its sampling design on the different types of anthropogenic activity taking place around each station. Gastropod specimens were collected by hand within a 1 m² area. Collection was accomplished through systematic hand-picking of individual gastropods attached to epilithic surfaces (i.e., rock surfaces), aquatic vegetation, or the primary substrate. To help reveal hidden gastropods in soft benthic substrates, mild mechanical agitation was performed. Once collected, biological samples were immediately preserved in 70% ethanol, systematically labeled, and placed into a cool box for storage. After collection, soft biological tissue was removed from its calcareous shell and then thoroughly rinsed with distilled water. Soft biological tissue was then thermally desiccated at 80 to 105°C for 24 hours until dry mass remained consistent (Masura, Baker, Foster, & Arthur, 2015). After desiccation, soft tissue was digested with 10% KOH at 60°C for 24 hours, a protocol shown to digest biological tissue without significant degradation of most common polymers (Dehaut et al., 2016). Slight surface erosion of alkaline-sensitive polymers such as PET and polycarbonate cannot be excluded, and semi-synthetic fibers such as rayon and cellulose acetate may be degraded (Thiele et al., 2019), so gastropod counts may be conservative. Immediately after digestion, soft biological tissue underwent density separation using a saturated NaCl solution for 24 hours. After completing the separation process, soft biological tissue samples were vacuum filtered through Whatman No. 42 filter paper. Samples were then completely dried and examined under an Olympus CX33 microscope at 40x magnification. Examination included quantifying absolute abundance and morphologically classifying the specific shape, color, and size of the microplastic polymers present in the samples.

To limit laboratory contamination, analysts wore 100% cotton laboratory coats, plastic labware was replaced with glass and stainless-steel equipment wherever possible, and all reagent solutions (saturated NaCl, 0.05 M Fe (II), 30% H2O2, and 10% KOH) were filtered through Whatman No. 42 filters before use. Procedural blanks, consisting or filtered ultrapure water processed alongside the samples through every step, were used to quantify background contamination from laboratory air and reagents, and the mean blank particle count was subtracted from the sample counts. These measures follow the contamination-control principles of ISO 24187:2023 and published recommendations (Brander et al., 2020).

2.5 Bioaccumulation and Transfer Factor Analysis

After the microplastic concentrations were accurately measured, the BAF was calculated to determine how much of the microplastics in the aquatic environment could be accumulated by an organism directly from its surroundings (Assas et al., 2020). The equation for determining the BAF is given below (Equation 1), where C_{biota} is the microplastic concentration in the biological sample (particles/kg dw) and C_{water} is the microplastics concentration in the ambient water sample (particles/L). BAF is therefore expressed in L/kg.

$$BAF\ (L/kg) = \frac{(C_{biota}\ (particle/kg\ dw))}{C_{water}\ (particle/L)} \quad \textbf{(1)}$$

The sediment to biota TF was then determined to assess the rate of movement of microplastics from the benthic sediment matrix to the gastropod samples (Nikhil et al., 2024). A mathematical derivation was made to obtain this ecotoxicological parameter by using Equation 2. C_{biota} is the microplastic concentration in the biological sample (particles/kg dw), and $C_{sediment}$ is the microplastics concentration in the benthic sediment sample (particles/kg dw). TF is dimensionless and was calculated for each station (i = 1, 2, 3) as well as for the river using the pooted values.

$$TF_i = \frac{C_{biota,i}\ (particle/kg\ dw)}{C_{sediment,i}\ (particle/kg\ dw)} \quad \textbf{(2)}$$

3. RESULTS AND DISCUSSION

3.1 Water Quality Characteristics and River Hydrodynamics

In-situ physical and hydrodynamic profiling established baseline environmental conditions across the upstream Brugan River. The environmental data set can be used to explain how the anthropogenic pollutants are dispersed, transported, and precipitated down the river gradient (APHA, AWWA, WEF, 2017). Table 1 provides a complete summary of the water quality measurements taken at each of the three sampling sites.

Table 1. Surface Water Quality Parameters in the Upstream Brugan River

Parameter	Station 1	Station 2	Station 3
Temperature (°C)	22	24	23
Flow Velocity (m/s)	0.3	0.5	0.2
Discharge (L/s)	156.24	332.5	67.8
TSS (mg/L)	6	4	4
Turbidity (NTU)	10	8	5

All three sampling stations demonstrate different levels of water quality based upon the local environment surrounding each station. Station 1 is in a very dark (shaded) area with high velocity water flowing through it and receives little sunlight from above and has cooler groundwater entering it (22 degrees Celsius). On the other hand, Station 2 is the warmest (24 degrees Celsius) due to being in a more open space that is either shallow or slow-moving and exposed directly to the sun, allowing the surface water to heat up rapidly. Station 3 is in between the other two stations with a temperature of 23 degrees Celsius and is indicative of both a mixture of environments. These variations in microclimates and thermal conditions are major abiotic factors influencing the metabolism and feeding habits of benthic animals (Nakkir et al., 2023). In terms of ecohydrology, the differences in

hydrodynamics among the three stations such as the increased discharge of 332.5 L/s at Station 2 versus the decreased flow rate of 0.2 meters/second at Station 3 determine how well the water can transport particles mechanically and apply shear force to the stream bed, thereby determining whether microplastic particles will stay suspended in the water column or fall to the bottom of the stream as part of the benthic matrix (Iqbal & Faisal, 2018; Kumar et al., 2021). The higher TSS (6 mg/L) and turbidity (10 NTU) at Station 1 are attributable to natural fine mineral particles (silt and clay) and organic detritus resuspended by turbulence from the upstream waterfall (Hersyah, 2017; Nuzula & Endarko, 2013). This suspended load did not coincide with elevated microplastic concentrations in the water (5 particles/L), suggesting that TSS and turbidity at this station were not driven by microplastic and that particles were retained mainly in the sediment.

3.2 Microplastic Concentration and Bioaccumulation Dynamics

The mean microplastic concentration recorded within the surface water samples reached 3 particles/L, whereas the benthic sediment and gastropod specimens exhibited accumulation levels of 221 particles/kg dw and 187 particles/kg dw, respectively. Furthermore, the BAF was calculated at 62.33, definitively indicating that the microplastic concentration accumulated within the biological matrices in the study area significantly exceeds the ambient aqueous concentration. Conversely, the sediment to biota TF yielded a value of 0.85. This specific metric signifies that the microplastic concentration within the gastropods remains marginally lower than that of the surrounding sediment. This ratio confirms the occurrence of active pollutant translocation from the benthic sedimentary matrix into the biological tissues, although it has not yet reached a state of hyperaccumulation within the living organisms relative to their primary habitat medium.

Based on the graphical visualization in Figure 1, a massive spatial divergence is clear regarding microplastic abundance across the different environmental compartments. The benthic sediment at Station 1 recorded the highest accumulation peak reaching 328 particles/kg dw, which contrasts sharply with the surface water column that contained only 5 particles/L. This pronounced concentration gradient confirms that benthic sediment serve as the primary depositional reservoir for microplastics in lotic ecosystems (Yang et al., 2021). Synthetic polymer particles undergoing morphological degradation, biofouling, and subsequent density alterations will continuously precipitate and accumulate within the benthic matrix, fundamentally driven by hyporheic exchange mechanisms and hydrodynamic turbulence across the riverbed (Drummond et al., 2022; He et al., 2021).

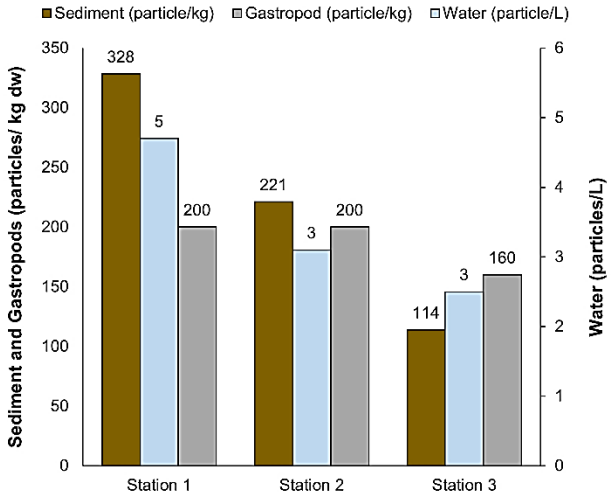


Figure 1. Microplastic concentration distribution across the three sampling stations.

Furthermore, the spatial analysis exhibits a definitive attenuation trend in sedimentary microplastics from the extreme upstream (Station 1) toward the downstream transition segment (Station 3). Sediment contamination was highest at Station 1 (328 particles/kg dw), consistent with tourism-related litter from the Coban Rondo Waterfall area combined with local hydrodynamics. Flow velocity in the calm pool below the waterfall (0.3 m/s) was lower than at Station 2 (0.5 m/s), which favors the settling of particles to the riverbed, whereas faster flow at Station 2 is more likely to carry particles downstream. Extensive recreational activities consistently introduce massive influxes of primary and secondary macroplastic debris, which subsequently degrade into microplastics. This spatial distribution robustly reinforces established ecological paradigms indicating that local land use typologies heavily dictate aquatic pollution trajectories; wherein upstream zones exposed to intensive tourism frequently discharge pollutant loads far exceeding those of domestic settlements or agricultural catchments, as evidenced by Station 3 holding a significantly lower concentration of 114 particles/kg dw in its sediment (Gutiérrez-Rial et al., 2024; Kunz et al., 2023).

From an ecotoxicological perspective, the microplastic bioaccumulation within the gastropod specimens projected an alarming biological stability. The biological matrices maintained a persistent contamination load of 200 particles/kg dw across both Station 1 and Station 2, and sustained a highly elevated count of 160 particles/kg dw at Station 3. This specific metric conclusively validates that freshwater gastropods actively assimilate ambient microplastics, a dynamic directly driven by their nonselective benthic deposit feeding behavior. Although the absolute sedimentary concentration attenuated drastically across the spatial gradient, the biological assimilation remained persistently high, highlighting a profound ecotoxicological vulnerability and suggesting a potential for chronic physiological toxicity induced by synthetic polymer ingestion within benthic habitats (Nikhil et al., 2024; Weber et al., 2021).

3.3 Microplastic Morphological Characteristics

Overall, fragment shapes dominated with a composition of 56%, followed by fibers at 32%, and films at 12% (**Figure 2**).

The pattern of fragment-type microplastics is more prominent in sediment and gastropods, while fibers and films appear more in water, in accordance with the general character of microplastics in aquatic environments. Fragments are generally the result of the breakdown of larger plastics, fibers are mostly associated with the release of synthetic fibers from domestic activities and textile washing, while films often come from thin packaging or plastic sheets that are easily fragmented. Hydrodynamically, fibers and films tend to be suspended longer in the water column because their large surface area relative to mass increases drag and slows settling, whereas more compact fragments settle to the riverbed, where they become available for benthic organisms to ingest (Duis & Coors, 2016).

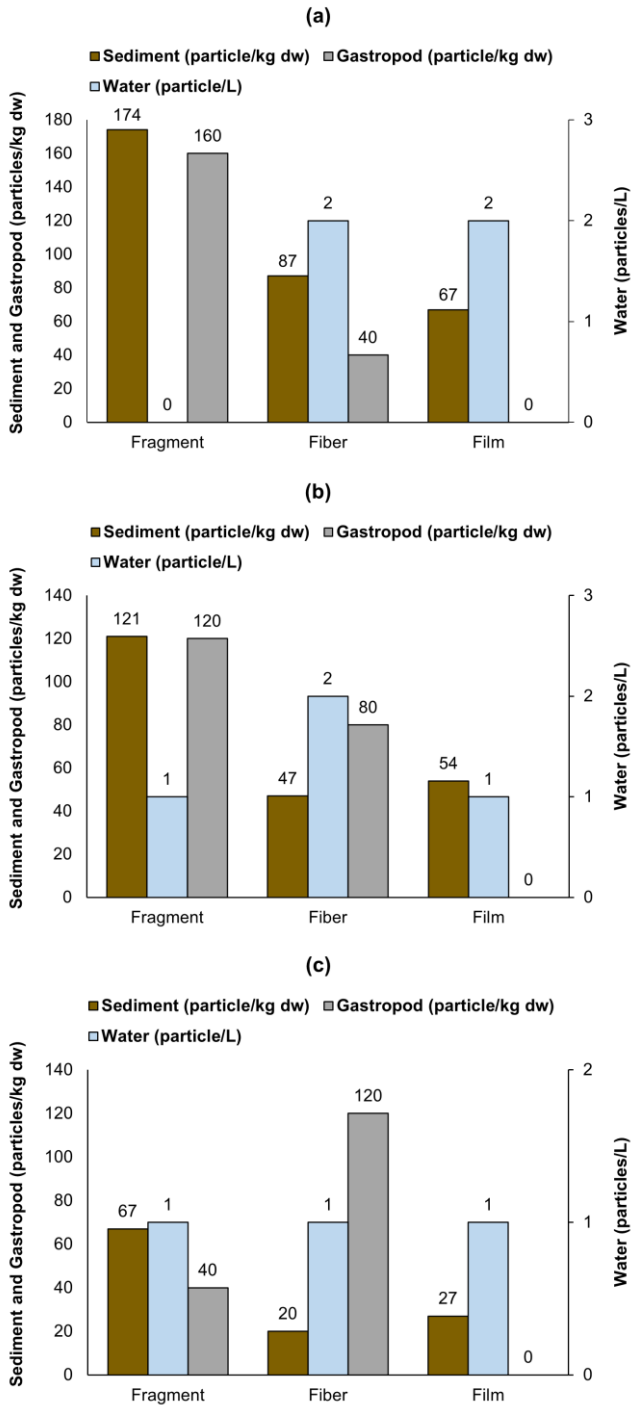


Figure 2. Microplastic particle morphology distribution across: (a) Station 1, (b) Station 2, and (c) Station 3.

The compositional disparity across the normalized matrices indicates that sediment acts as a primary accumulation medium, especially for denser, rapidly precipitating fragments. Within gastropods, fragment dominance signifies that benthic organisms experience primary exposure to matrix-associated microplastics within their sedimentary habitat and feeding grounds, causing their internal morphological profile to closely mirror sedimentary accumulation characteristics (Zaki et al., 2021). This morphological correspondence reinforces the validity of utilizing benthic gastropods as sentinel organisms for localized sedimentary microplastic pollution (Fitri & Patria, 2019). The persistence of fibers in the water column, particularly at Station 2, reflects continuous localized inputs from domestic graywater discharges associated with nearby residential settlements. Conversely, the striking shift toward fiber dominance inside gastropods at Station 3 suggests selective ingestion of organic-rich or biofilm-coated fibrous microparticles originating from agricultural runoff and livestock effluents prevalent in this zone (Kunz et al., 2023). The complete absence of films within the gastropod matrix further indicates that benthic organisms actively ingest particulate debris associated with consumed detritus, whereas films are excluded due to ingestion, morphological constraints or rapid physiological elimination (Weber et al., 2021). This systematic compartmentalization underlines the intricate physical-biological sorting mechanisms operating across lotic ecosystem boundaries (Queiroz et al., 2024).

3.4 Microplastic Size Distribution

Overall, microplastics measuring less than 1 mm dominated with a proportion of 67%, while the 1–5 mm size accounted for 33%. The overall difference in size distribution across the matrices indicates the pathway of microplastic transfer from the water column to the sediment as an accumulation site, after which some small fractions enter benthic organisms as an initial trophic exposure route.

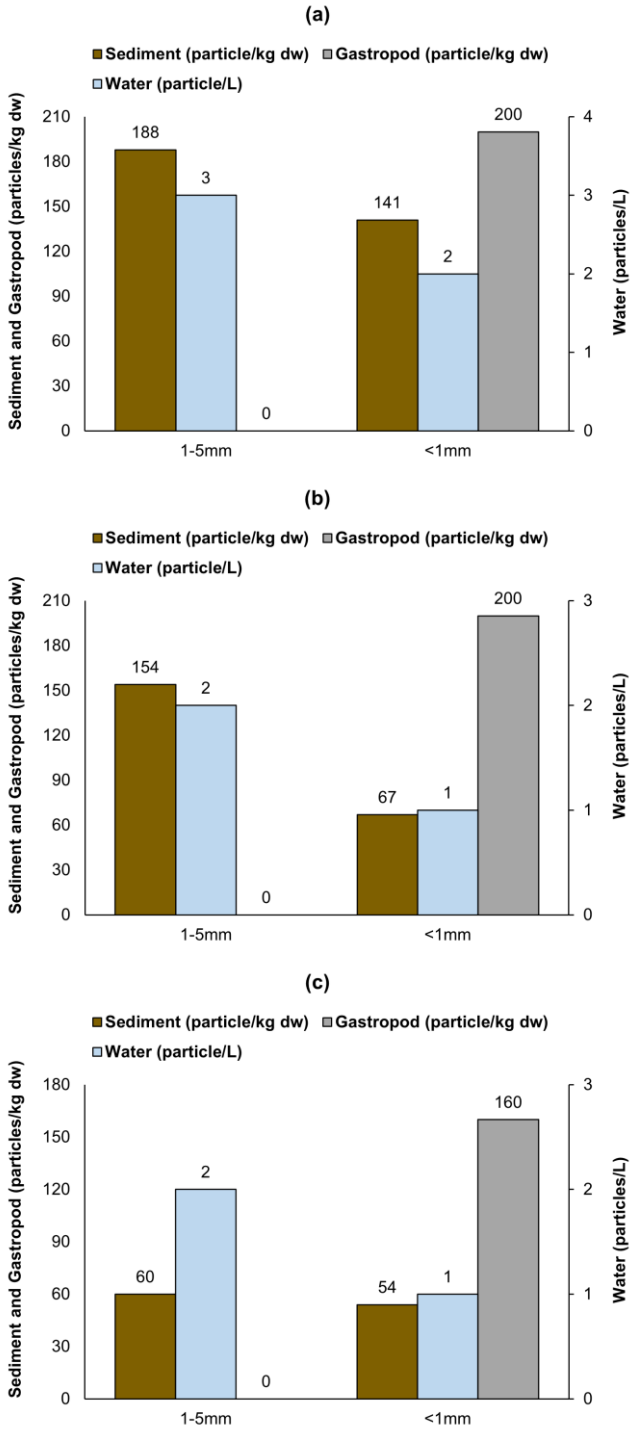


Figure 3. Microplastic particle size distribution across: (a) Station 1, (b) Station 2, and (c) Station 3.

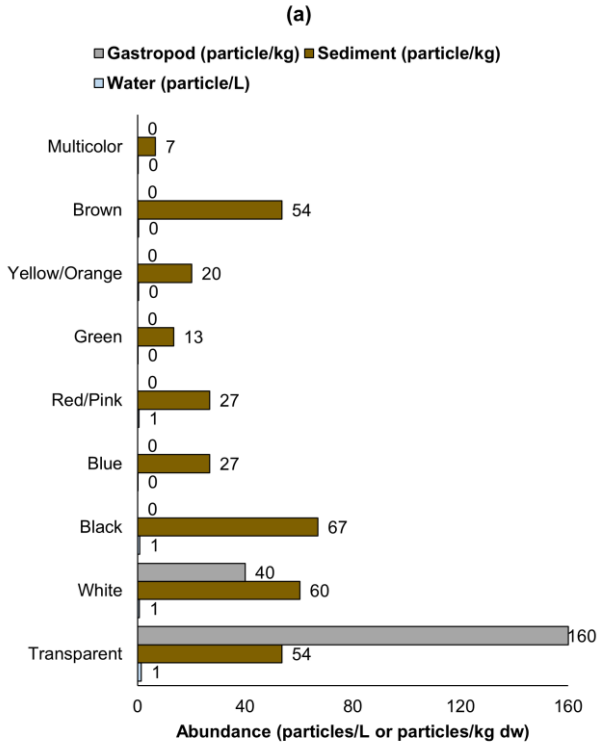
Following data normalization to equivalent quantitative units, the spatial distribution of these size fractions starkly elucidates the functional role of each environmental compartment. In the aqueous matrix across all stations, the 1-5 mm fraction surprisingly constituted the majority (62%), while the <1 mm fraction represented 38%. From a hydrodynamic perspective, larger fragments can remain suspended during high flow conditions but possess a strong propensity to ultimately precipitate, whereas submillimeter particles persist longer in suspension and readily disperse into the trophic chain (Rushdi et al., 2023). Consequently, the water column strictly functions as a transient suspension phase featuring inherently low absolute volumetric concentrations (Kumar et al., 2021).

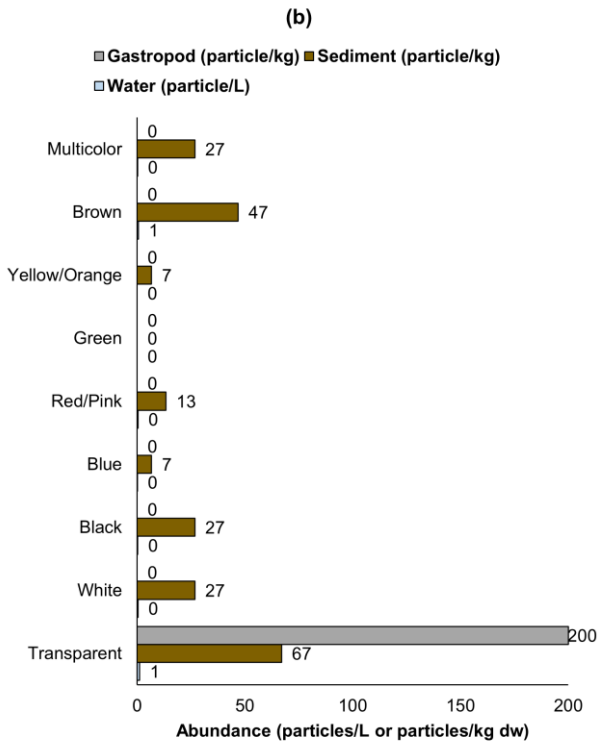
Upon precipitating into the benthic zone, the sedimentary matrix emerges as the primary accumulative reservoir, heavily dominated by the 1-5 mm macro-fraction (61%) alongside a substantial <1 mm sub-fraction (39%). This depositional bias occurs because larger particles exhibit higher specific gravities and rapidly bind with benthic organic matter and minerals, firmly anchoring them within the riverbed (Drummond et al., 2022; He et al., 2021). The concurrent presence of the submillimeter fraction within the sediment signifies continuous in situ mechanical fragmentation, localized environmental weathering, and sustained exposure potential for benthic communities (Zha et al., 2022).

Most critically, the biological assimilation profile within the gastropod specimens reveals an extreme morphological filtering mechanism. Across all sampling stations, the gastropod matrices exhibited an absolute 100% accumulation of the <1 mm submillimeter fraction, while the 1–5 mm size fraction was entirely absent. This selective accumulation aligns with anatomical limits. The mouth aperture and esophageal lumen of small freshwater snails physically restrict particle intake to <1 mm, rendering larger synthetic debris non-ingestible. The benthic foragers such as Melanoides tuberculata are morphologically constrained by their radular feeding apparatus and digestive tract capacities, physically restricting them to ingesting only micro-sized particulates (Weber et al., 2021). Furthermore, submillimeter particles boast a significantly higher bioavailability as they easily entangle within the benthic biofilms and detritus that these organisms graze upon (Gutow et al., 2016; Zaki et al., 2021). This phenomenon reflects a robust and highly selective biological filtration process operating at the sediment boundary layer, definitively tracing the exact ecotoxicological pathway from abiotic accumulation to primary trophic ingestion (Nikhil et al., 2024).

3.5 Microplastic Color Composition

The color composition of microplastics in the study area revealed that transparent, white, black, and brown particles were the most frequently observed, while brighter colors appeared in smaller quantities. This pattern generally indicates a combination of environmentally aged microplastics and fresh anthropogenic inputs from daily sources such as domestic waste, synthetic textile laundering, and plastic packaging. In recent studies, microplastics exposed to ultraviolet radiation, oxidation, mechanical abrasion, and biodegradation tend to experience color fading, surface alteration, and biofilm colonization, causing the particles to become more neutral or dark, whereas bright or striking colors are often associated with fresher inputs from human activities (Zha et al., 2022). Ecologically, the dominance of neutral and dark colors also aligns with river conditions that receive runoff loads and subsequently undergo sedimentation, so that older particles coated with organic material are more likely to persist at the bottom of the water and are then exposed to benthic organisms such as gastropod (Kye et al., 2023).





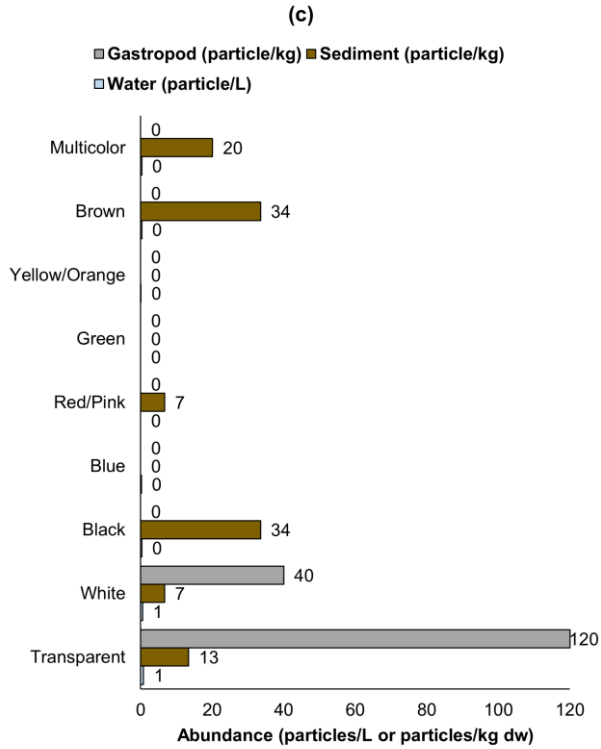


Figure 4. Microplastic particle color across: (a) Station 1, (b) Station 2, and (c) Station 3.

A detailed spatial analysis across the sampling coordinates further elucidates the functional chromatic partitioning of these synthetic microparticles. Within the aqueous matrix, visual observations recorded negligible concentrations across all color spectrums, reaffirming the water column's role as a transient suspension phase where particles rapidly relocate to the benthic boundary layer (Kumar et al., 2021). Conversely, the sedimentary matrices across Station 1, Station 2, and Station 3 functioned as profound long-term accumulative reservoirs, sequestering a highly diverse and darker spectrum predominantly composed of black, brown, and multicolor fragments. This localized accumulation of dark and diverse particles originates from terrestrial runoff and extensive environmental weathering, wherein heavier, biofouled, or mineral-bound plastics precipitate and become permanently trapped within the riverbed (Yang et al., 2021; Zha et al., 2022).

Most notably, the biological assimilation within the gastropod specimens exhibited an extreme chromatic selectivity, overwhelmingly dominated by transparent and white particles. At Station 2, for instance, gastropod tissues accumulated a peak of 200 transparent particles/kg, with all other colors remaining undetected. Rather than relying on visual cues, freshwater snails forage via chemoreception, these transparent particles become coated with microbial biofilms, which relrease chemical signals mimicking natural organic matter and triggering accidental ingestion during grazing. This distinct biological reflection signifies a strong localized exposure; benthic detritivores predominantly ingest faded, biofilm-coated transparent fragments that are either heavily integrated into the fine sediment matrix or visually and texturally indistinguishable from their natural organic food sources during foraging (Romaskila et al., 2023; Zaki et al., 2021). Consequently, the color distribution across these

matrices definitively traces the microplastic lifecycle from fresh diverse inputs to darkened sedimentary sinks, culminating in the selective trophic ingestion of faded sub-fractions by benthic macroinvertebrates (Polt et al., 2023).

3.6 Anthropogenic Influence on Microplastic Abundance

Spatial variations in microplastic burdens reflect coupled interactions between localized anthropogenic pressures and fluvial hydrodynamics. Local land-use typologies heavily dictate the intensity and composition of synthetic waste entering the aquatic system, while in-stream hydrodynamic variables, such as flow velocity and localized turbulence, control the subsequent dispersion and precipitation mechanisms of these microparticles (Kumar et al., 2021; Kunz et al., 2023). Station 1 exhibits the highest concentration in sediment and gastropods owing to its proximity to anthropogenic point sources at the Coban Rondo Waterfall tourist zone, including hospitality fasilities and food stalls. High visitor density generates abundant disposable drink cups and snack packaging, which easily collapse into the aquatic system due to a lack of covered waste receptacles along the riverbanks. Extensive recreational activities and intensive hospitality operations consistently introduce massive influxes of primary and secondary macroplastic debris, predominantly derived from single-use packaging and discarded consumer goods, which rapidly degrade into microplastics under mechanical and environmental stressors (Barnes, 2019; Suripatty et al., 2023).

Station 2 is located in a residential area, while Station 3 is dominated by agricultural land, plantations, and livestock. These varying land-use patterns generate distinct pollutant signatures; residential zones typically discharge high volumes of synthetic textile fibers via domestic graywater effluents, whereas agricultural and plantation sectors often release degraded polymeric fragments originating from agricultural netting, ropes, and mulch films (Nurhasanah et al., 2021; Zhou et al., 2018). This pattern confirms that rivers are not only microplastic transport routes but also accumulation sites, positioning sediment as the primary reservoir from which benthic gastropods actively internalize pollutants. By continuously ingesting riverbed mud and algae for organic nourishment, these bottom-dwelling snails persistently entrapand accumulate microplastics within their digestive tracts. The continuous precipitation of synthetic polymers transforms the benthic boundary layer into a long-term toxicological sink, dramatically escalating the bioavailability of these micropollutants to deposit-feeding fauna, thereby establishing a critical pathway for microplastic integration into the fundamental levels of the freshwater food web (Drummond et al., 2022; He et al., 2021).

The results of this study are in line with the literature that microplastics in rivers are highly influenced by human activity density and are generally higher in sediment than in water and can accumulate in benthic organisms (Drummond et al., 2022; Gutiérrez-Rial et al., 2024; Talbot & Chang, 2022). Consequently, robust spatial monitoring that couples' sedimentary matrices with benthic biomonitors is highly imperative for accurately assessing the ecological risks and managing the localized impacts of anthropogenic point-source pollution within lotic highland ecosystems (Blettler et al., 2018; Frank et al., 2023).

4. CONCLUSION

Sediment is the matrix with the highest concentration with an average amount of 221 particles/kg dw, followed by gastropods at 187 particles/kg dw, while water has the lowest concentration of 3 particles/L. A bioaccumulation factor value of 62.33 L/kg was obtained, which indicates that the microplastic concentration in the biota in the study area is much higher than the concentration in the water, while the transfer factor value of 0.85 can be interpreted as an indication of a real but still limited sediment to biota transfer, so gastropods act as recipients of exposure from the sediment, but have not yet shown enrichment that exceeds the concentration in the sediment itself. These metrics conclusively establish that lotic riverbeds function as the ultimate sinks for anthropogenic polymer pollutants, transforming the benthic boundary layer into a zone of chronic ecotoxicological exposure, while solidifying the utility of gastropods as reliable, non-hyperaccumulating bioindicators of localized pollution.

Overall, this study establishes that benthic sediment acts as the primary microplastic trap, with gastropod ingestion strictly constrained to <1 mm fractions due to anatomical limits. Consequently, freshwater gastropods provide a reliable and practical bioindicator for assessing localized microplastic burdens in riverine enviroments. This specific morphological signature highlights the advanced state of environmental weathering and confirms a severe biological filtration constraint, emphasizing that the most degraded microparticles pose the most immediate ingestion risk to benthic primary consumers. Furthermore, the research data display that microplastic concentrations do not follow a simple geospatial pattern from upstream to downstream; rather, the microplastic load is more likely controlled by a combination of anthropogenic factors and sediment retention processes at each station. In terms of river basin management, localized tourism activities at high-altitude river sources serve as the dominant driver of microplastic distribution in the upper reach of the Brugan River. This mechanism generates hevay pollution loads directly at the headwaters directly challenging the conventional model of downstream urban microplastic accumulation. Nature tourism, food stalls, and lodging in the upstream Brugan River area contribute greatly to producing microplastics originating from product packaging. This finding decisively refutes the conventional paradigm of linear downstream accumulation, proving instead that point-source anthropogenic pressures, act as the primary drivers of aquatic microplastic contamination. Practical environmental management should focus on targeted engineering interventions directly downstream of the tourist area. Key recommendations include planting riparian buffer strips along riverbanks to filter land-based runoff, installing floating debris traps to catch floating plastic wastes, and implementing routine biomonitoring using benthic gastropod as a cost-effective indicator of river health. Advanced spectroscopic characterization, such as Fourier Transform Infrared (FTIR) or Raman spectroscopy, is essential to decode the exact chemical signatures of these polymers, which will transition the findings from a purely physical assessment to a comprehensive ecotoxicological risk evaluation. Additionally, a more specific grouping of microplastic sizes is needed to clarify the data and determine the average size of the identified microplastic particles. Implementing high-resolution, narrow-band size

fractionation will critically unravel the exact tipping point of bioavailability across different benthic species, ultimately providing robust empirical data for establishing stringent environmental quality standards in freshwater ecosystems.

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