



Abundance and Distribution of Microplastics in Fish by Trophic Level in Kupang Bay, Indonesia

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ABSTRACT

Kupang Bay is home to a diverse array of fish species and other vital marine resources. However, increasing human activities—including industrial development, fisheries, and tourism—have contributed to elevated levels of microplastic pollution. This study aimed to assess the abundance and distribution of microplastics in fish, categorized by trophic levels (herbivores and carnivores), as well as by the shape, size, and color of microplastics found in different fish organs: the gastrointestinal tract, gills, and muscle, in the waters of Kupang Bay. The research process involved sampling fish from three different locations, identifying microplastics using a microscope, describing the characteristics of identified microplastics, and analyzing their abundance in the fish samples. Microplastics were detected in all organ types of both herbivorous and carnivorous fish. The microplastics appeared in various forms, including fibers, fragments, films, and pellets. Among these, the most dominant type was blue-colored fibers measuring less than 0.25mm. Carnivorous fish exhibited the highest abundance of microplastics, particularly fibers found in the gastrointestinal tract. Notably, there were differences in microplastic abundance across the three sampling sites. However, no significant difference was observed in the average microplastic abundance between carnivorous and herbivorous fish. The presence of microplastics in multiple organs of fish across different trophic levels poses a potential threat to marine biodiversity and fisheries sustainability. Furthermore, the risk of microplastic accumulation through the food chain could have implications for human health.

INTRODUCTION

Plastics in the ocean degrade into microplastics primarily through photooxidation caused by ultraviolet (UV) exposure, especially in surface and coastal waters (Andrady, 2015). Microplastics, ranging in size from 25 micrometers to 5 millimeters, are widely distributed across ecosystems and are considered major pollutants (McHale & Sheehan, 2024). These particles enter aquatic environments and can be reintroduced to humans through ingestion, inhalation, or skin contact (Kye *et al.*, 2023). Microplastics originate

from both primary sources, such as industrial microfibers and cosmetic microbeads, and secondary sources, including the breakdown of larger plastic debris like bags and bottles (Koirala *et al.*, 2025).

Kupang Bay, located in East Nusa Tenggara Province, Indonesia, is a rich habitat for diverse fish species and other important marine resources. However, the expansion of human activities—such as industrial operations, fisheries, and tourism—has contributed to significant environmental degradation. In fact, plastics constitute up to 88.91% of the total waste in the Kupang City mangrove ecosystem, indicating a high potential for microplastic pollution (Toruan *et al.*, 2022). Previous studies have confirmed the presence of microplastic contamination in Kupang Bay, including in the water surface column (Kapo *et al.*, 2020), the intestines of saltwater fish (Widyantoro *et al.*, 2022), and the gills and intestines of the red snappers (Ngai *et al.*, 2024).

Microplastics can easily enter the aquatic food chain, contaminating various marine organisms including fish (McHale & Sheehan, 2024). These particles are readily ingested by marine life either directly from the environment or indirectly through trophic transfer within the food web (Geng, 2024). The ingestion, accumulation, and elimination of microplastics in fish are species-specific and are largely influenced by feeding behavior (Zhang *et al.*, 2023).

The problem intensifies as microplastics transfer through the food web, moving from one trophic level to the next. Carnivorous fish, occupying higher trophic levels, consume smaller, contaminated fish, leading to increased microplastic accumulation (Gao *et al.*, 2024; Habumugisha *et al.*, 2024; Liaqat *et al.*, 2024). Several studies have shown that carnivorous and omnivorous fish tend to accumulate more microplastics than herbivorous species. For instance, Hastuti *et al.* (2019) detected microplastics in the digestive tracts of commercial fish from the coast of Pantai Indah Kapuk, Indonesia. Similarly, Khan and Setu (2022) reported higher microplastic abundance in carnivorous and omnivorous freshwater fish from the Jamuna River, although the correlation with trophic level was not statistically significant. In contrast, Canon-Bastidas *et al.* (2025) observed a significant positive correlation between trophic level and microplastic ingestion among fish from the Ariidae and Sciaenidae families in the Gulf of Tumaco and Buenaventura, suggesting that higher trophic-level fish accumulate more microplastics through biomagnification. This conclusion is further supported by Li *et al.* (2023), who demonstrated that microplastic concentrations increase significantly via trophic transfer from contaminated prey.

Beyond harming aquatic organisms, microplastics serve as carriers for toxic pollutants, facilitating their movement from sediments into fish tissues and ultimately into humans through seafood consumption (Jinadasa *et al.*, 2023; Pourebrahimi & Pirooz, 2023; Mchale & Sheehan, 2024).

Although extensive research has examined microplastic accumulation in the gastrointestinal tracts and gills of fish, studies investigating accumulation in muscle

tissue—especially in relation to trophic level and geographic distribution within Kupang Bay—remain limited. This study, therefore, aimed to assess microplastic accumulation in fish based on two trophic levels (herbivores and carnivores), focusing on the type, size, and color of microplastics found in the gastrointestinal tract, gills, and muscle. Additionally, it seeks to compare microplastic abundance in herbivorous and carnivorous fish sampled from various locations within Kupang Bay.

The findings of this study are expected to serve as an early warning regarding environmental degradation and contribute to greater awareness of ecosystem sustainability. Ultimately, the results may support efforts to ensure the quality and safety of fishery products from the waters of Kupang Bay.

MATERIALS AND METHODS

Research site

This study was conducted in three locations in Kupang Bay. Location 1 is the waters around Lasiana Beach, representing Kupang's tourist attractions and residential areas. Location 2 represents the area around PT Tom, a pearl oyster cultivation area and a tourism area with less dense settlements. Location 3 stands for the waters around Oeba, including most of its coastal areas converted into residential areas, public and fish markets, fishing ports, and densely populated areas (Fig. 1).

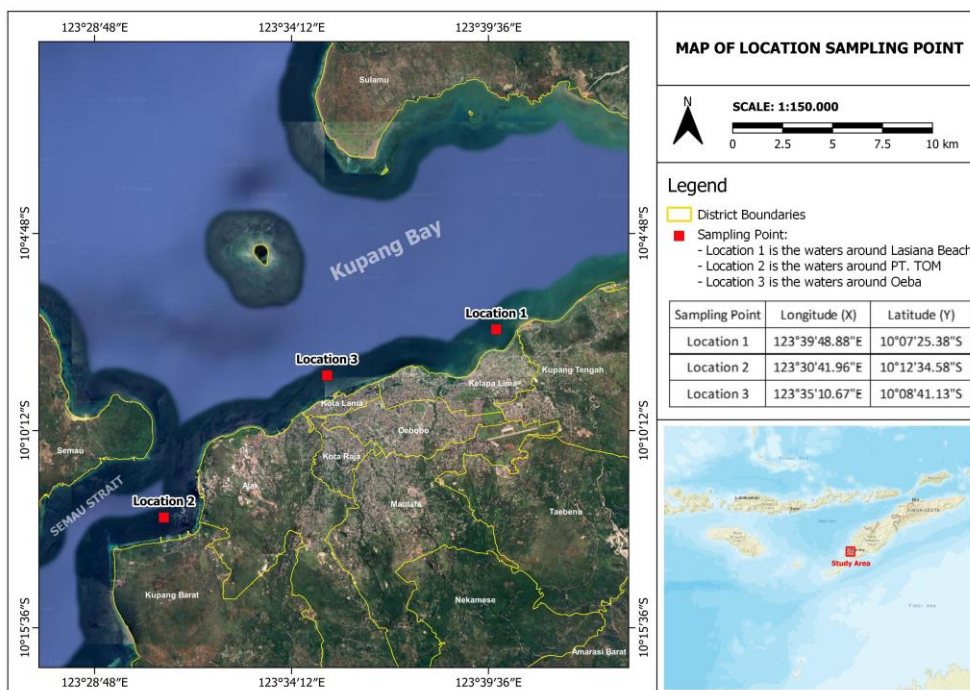


Fig. 1. Map of the waters of Kupang Bay showing the sampling location of East Nusa Tenggara, Indonesia. The red dot represents the location of the carnivorous fish sampling.

Procedures

Sampling

A total of 36 fish samples were collected from three stations within Kupang Bay. Each station provided 12 samples, representing both herbivorous and carnivorous groups. The fish were caught by local fishermen using handlines from outboard motorboats during approximately five hours of fishing activity. Immediately after capture, the samples were placed in a cooling box filled with ice to maintain freshness prior to laboratory analysis.

Dissection procedure

Dissection was conducted to obtain target organs, including the gills, gastrointestinal tract, and muscle tissue, for microplastic analysis. Each fish was dissected using surgical scissors, with an incision made from the anus to the anterior section near the gill opening. The digestive tract (stomach to intestines), gills, and muscle tissue (from the mid-abdomen to the dorsal area) were collected. All dissected parts were temporarily stored in covered beaker glasses to minimize contamination from airborne microplastics.

Microplastic extraction and drying process

For each sample, a 10% potassium hydroxide (KOH) solution—prepared with ultrapure water—was added in a volume three times that of the sample. The tissue samples were placed in chemical glassware and covered with aluminum foil to prevent contamination. Samples were then incubated in an oven at 60°C for 48 hours to digest organic material (Dehaut *et al.*, 2016; Barboza *et al.*, 2020; Singh *et al.*, 2024).

After digestion, the contents were filtered using a vacuum pump and Whatman filter paper with a 2.5µm pore size. The filtered residues were then dried again in an oven at 40°C for 24 hours. Once dried, the samples were ready for microplastic identification.

Microplastic identification

Microplastics were identified under a microscope at 20x magnification using an OptiLab system connected to a laptop for ease of observation and documentation. White and UV lamps were used to enhance visibility during the identification process. Visible microplastic particles were photographed, and their dimensions were measured using ImageJ software. Data were further processed and categorized using Microsoft Excel 2010.

Microplastics were classified based on their shape, size, and color. The observed shapes included fibers, fragments, films, pellets, and granules (Jiang *et al.*, 2018; Zhao *et al.*, 2018). Size categories were adapted from Abbasi *et al.* (2018) and included: <0.25 mm, 0.25–0.50 mm, 0.51–2 mm, and >2 mm. Colors were visually identified and categorized as transparent, blue, yellow, red, black, and green (Abidli *et al.*, 2019).

Data analysis

The abundance and type of microplastic particles were manually counted and recorded. Data were visualized in the form of graphs, tables, and diagrams illustrating the color, shape, size, and abundance (particles per individual) of the microplastics detected.

Statistical analysis was conducted at a significance level of $P=0.05$. The one-way ANOVA was used to assess differences in the average number of microplastics across the three sampling sites. A Tukey post hoc test was performed to compare microplastic accumulation between carnivorous and herbivorous fish species. All statistical tests were carried out using SPSS version 25.

RESULTS

Distribution of microplastics in each research location

The average number of microplastic particles found in fish varied across the three sampling locations: Location 1 (Lasiana Beach), Location 2 (around PT. Tom), and Location 3 (Oeba Beach). Among these, fish from Location 3 exhibited the highest microplastic concentration, with an average of 159.67 particles per individual. This was followed by fish from Lasiana (Location 1), with an average of 119.33 particles, while the lowest average was recorded at PT. Tom (Location 2), with 98.25 particles per individual.

A one-way ANOVA revealed a statistically significant difference in the average number of microplastics among the three locations ($P < 0.05$). Further analysis using Duncan's Multiple Range Test indicated that the microplastic count in fish from Location 3 was significantly higher than that in fish from locations 1 and 2 (Fig. 2).

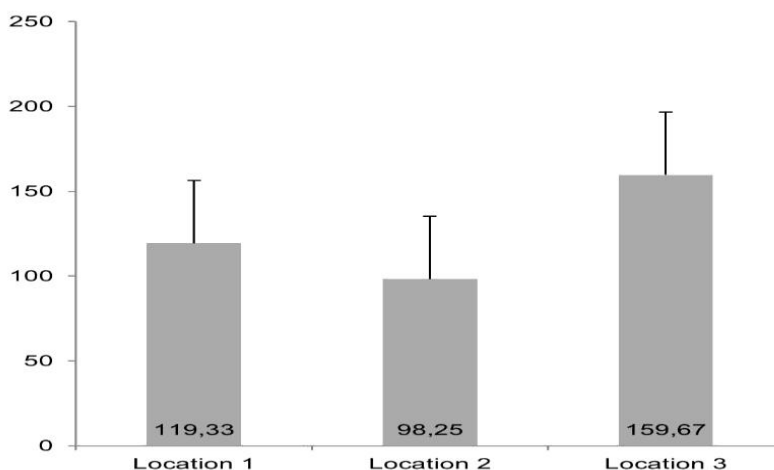


Fig. 2. Average microplastics at each research site

Microplastics by shape

The distribution of microplastics by shape in fish from carnivorous and herbivorous trophic levels revealed that fibers had the highest average abundance across both groups, while pellets—which were detected only in herbivorous fish—showed the lowest average count (Fig. 3).

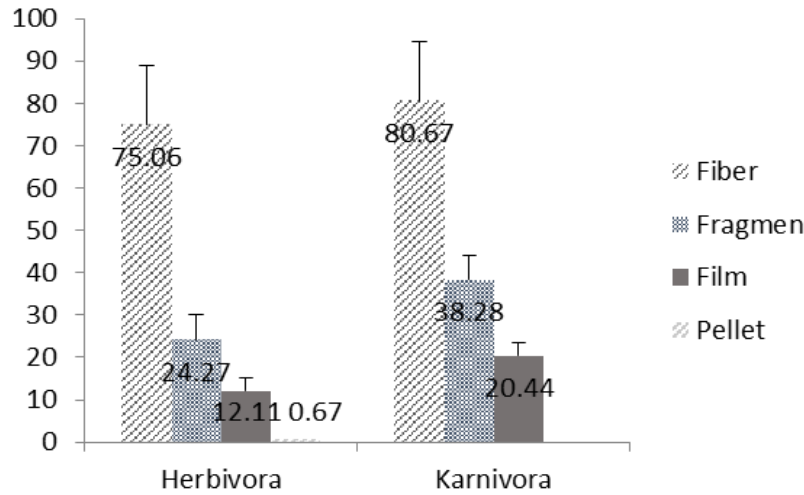


Fig. 3. Average microplastics in herbivore and carnivore fish

Microplastics by color

The distribution of microplastics by color across the three sampling sites showed that blue particles were the most prevalent, accounting for 26% of the total, while green particles were the least common, representing only 2% (Fig. 4).

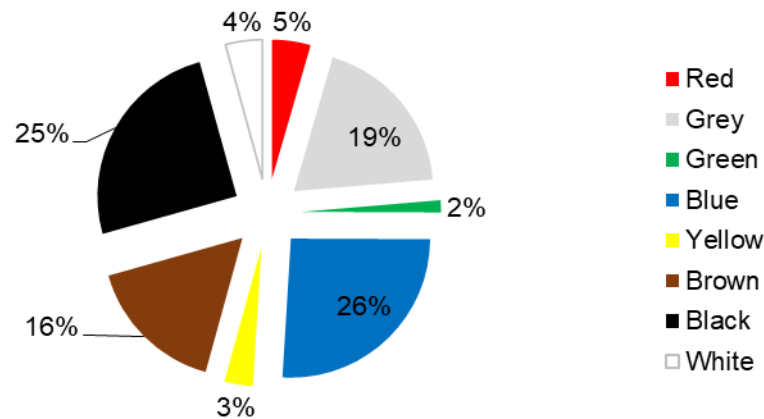


Fig. 4. Percentage of microplastic colors found in three locations

Microplastics by size

Microplastics based on measurements found at three research sites on all fish samples in each organ. Measurements of the length of microplastics found in each organ in all fish samples showed that the most common microplastic size was <0.25 mm in the gastrointestinal tract, followed by gills and meat (Fig. 5).

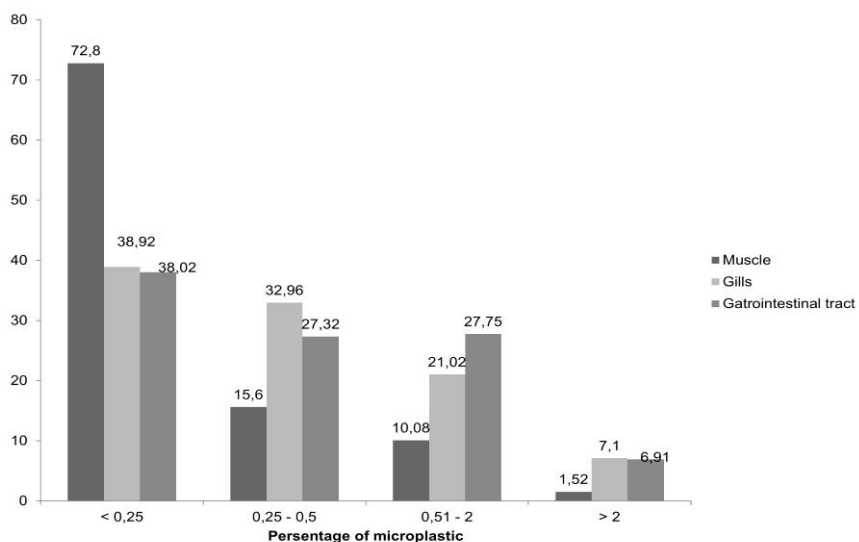


Fig. 5. Percentage of microplastic size of all fish found

Abundance of microplastics

The average abundance of microplastics was analyzed across different fish species representing carnivorous and herbivorous trophic levels, as well as within specific organs. A total of 10 fish species were examined. Among them, *Eucinostomus gula* exhibited the highest average microplastic abundance, with 225 particles per individual. In contrast, *Dascyllus trimaculatus* had the lowest, with an average of 99.67 particles per individual (Fig. 6). The average abundance of microplastics in the gastrointestinal tract, gills, and muscle tissue was also compared between carnivorous and herbivorous groups (Fig. 7).

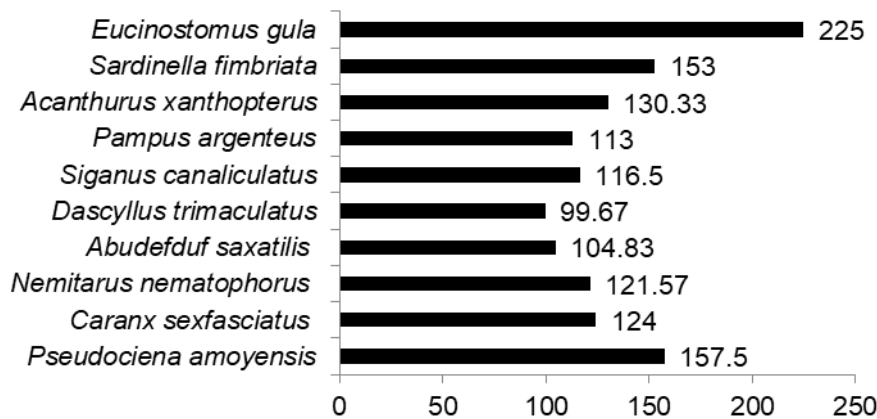


Fig. 6. Average amount of microplastics in each type of fish

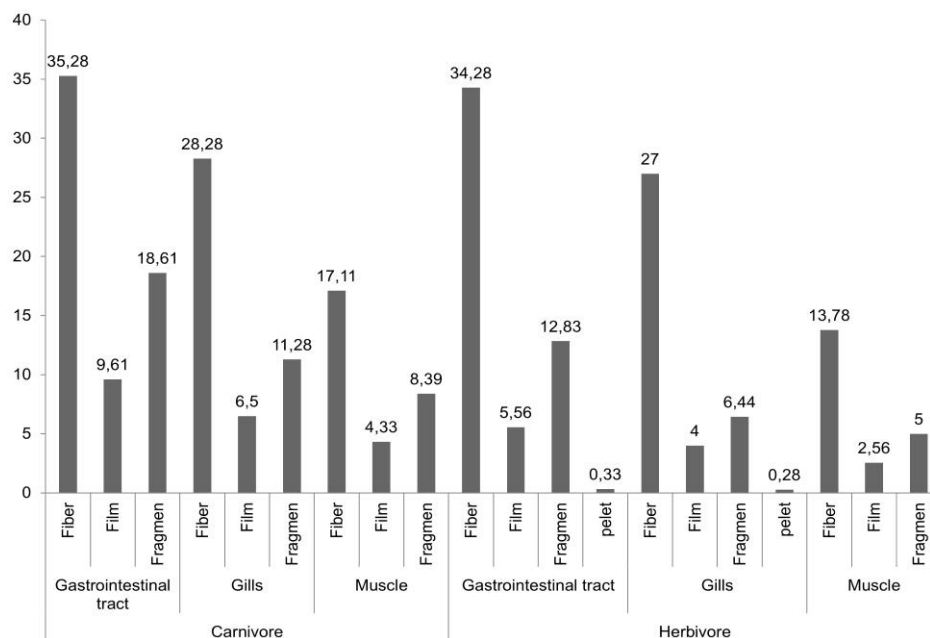


Fig. 7. Average abundance of microplastics in carnivorous and herbivorous fish groups on body parts of microplastic forms

The average microplastic content in carnivorous fish (278.78 ± 188.29 particles/individual) was higher than that found in herbivorous fish (168.17 ± 198.02 particles/individual). Fibers were the most predominant form of microplastics found across all organs, particularly in the gastrointestinal tract and gills. Muscle tissue also showed evidence of contamination, though to a lesser extent. The results indicate a trend suggesting that trophic level may influence microplastic accumulation. However, statistical analysis using an independent samples t-test showed no significant difference between the two trophic groups ($\text{sig} = 0.216$; $P > 0.05$), indicating that the variation in

microplastic abundance between carnivorous and herbivorous fish was not statistically significant. Despite this, descriptive data revealed differences in both the quantity and type of microplastic particles between the groups.

DISCUSSION

Distribution of microplastics at each research location

The content of microplastics in fish is strongly influenced by the sampling location. Several studies have shown that environmental context—such as whether an area is coastal or freshwater—can lead to differences in both the quantity and types of microplastics accumulated. These variations are closely tied to human activities and local environmental conditions (Wardlaw *et al.*, 2022; Putri *et al.*, 2023; Dwiyoitno *et al.*, 2024). Microplastic concentrations tend to be higher in areas with dense human activity, including urban centers, ports, and drainage zones, due to inputs from fishing, domestic waste, and industrial effluents (Bae & Yoo, 2022; Díaz-Jaramillo *et al.*, 2023; Noor *et al.*, 2025).

The results of this study confirm these findings. ANOVA analysis showed a statistically significant difference in microplastic abundance across the three locations ($P < 0.05$). Duncan's post hoc test revealed that Location 3 had a significantly higher microplastic count compared to locations 1 and 2 (Fig. 2). Location 3, situated around Oeba Beach, is a densely populated area with a nearby fish market and traditional shipping activity. Poor waste management practices, combined with reduced water circulation due to natural bay structures and port facilities, contribute to microplastic accumulation in this area.

Kupang Bay is recognized as a coastal region susceptible to microplastic contamination. For example, Widiantoro *et al.* (2023) reported microplastic filaments in these waters, and Toruan *et al.* (2022) attributed the increase in plastic waste to anthropogenic sources and runoff from rivers and drainage systems—factors that promote pollutant buildup in coastal zones.

Shape, size, and color of microplastics

The forms of microplastics identified in this study included fibers, fragments, films, and pellets (Fig. 3), consistent with prior findings by Tubagus *et al.* (2020). Pellet-shaped microplastics were observed only in herbivorous fish. Their presence may be attributed to biofouling, where microbial colonization alters buoyancy and causes particles to sink. Sinking plastics are then more likely to be ingested by benthic or herbivorous fish that feed along the substrate. For instance, Kaiser *et al.* (2017) reported that biofouling increases the density of polyethylene microplastics, causing pellets to settle after about six weeks in seawater. Similarly, fish like *Acanthurus chirurgus* exhibit scraping behavior that increases their likelihood of ingesting embedded particles (Cardozo-Ferreira *et al.*, 2021).

Fibers were the most dominant microplastic type in all organs (gastrointestinal tract, gills, and muscle), as shown in Fig. (4). These are characterized by elongated,

flexible structures and are typically derived from synthetic textiles and degraded fishing gear. Their physical traits enhance dispersion in water and increase ingestion potential (Frost *et al.*, 2022; Weis & De Falco, 2022; Biswal, 2024; Chan *et al.*, 2024; Chen *et al.*, 2024; Samal *et al.*, 2024; Sharma *et al.*, 2024).

The physical characteristics of microplastics—shape, size, and color—strongly influence ingestion by aquatic organisms, often due to visual similarity to food (Benson *et al.*, 2022). In this study, blue and black were the most frequently observed colors (Fig. 5), and the most common size was < 0.25mm (Fig. 6). Blue and black particles are commonly associated with fishing nets and ropes and are frequently ingested by fish due to their abundance and resemblance to prey (Shu *et al.*, 2023; Mutuku *et al.*, 2024; Sacco *et al.*, 2024; Vellore Mohan *et al.*, 2024). Fish appear to have a visual affinity for blue particles (Ory *et al.*, 2018; Neves *et al.*, 2015), and Barboza *et al.* (2020) observed large fibers and fragments in dorsal muscle tissues up to 2363 and 490µm, respectively. Oza *et al.* (2024) also reported frequent detection of black and blue microplastics, particularly < 500µm to 1mm in size, in omnivorous and carnivorous fish in Asia.

Abundance and trophic influence

Among herbivorous fish, the highest microplastic abundance was recorded in *Acanthurus xanthopterus* (130.33 particles/individual), while the carnivorous species *Eucinostomus gula* showed the highest overall abundance. On average, carnivorous fish had a higher microplastic load (278.78 ± 188.29 particles/individual) than herbivorous fish (168.17 ± 198.02), suggesting a trend related to trophic level. However, the difference was not statistically significant (t-test, sig = 0.216). This is likely due to the unequal number of herbivorous and carnivorous fish groups analyzed, which may have affected the statistical power to detect significant differences between the two trophic groups. A similar pattern was also reported by Khan and Setu (2022), who found that carnivorous and omnivorous freshwater fish from the Jamuna River exhibited higher microplastic abundance compared to herbivorous species; however, the correlation between microplastic ingestion and trophic level was not statistically significant.

Microplastic accumulation is influenced by various factors beyond trophic level, including habitat, feeding behavior, and local environmental conditions. Canon-Bastidas *et al.* (2025) reported a positive correlation between trophic level and microplastic ingestion, consistent with Gao *et al.* (2024), who demonstrated biomagnification through trophic transfer. Carnivorous fish are more prone to accumulation due to predation on already contaminated prey (Oza *et al.*, 2024). Supporting this, Khan and Setu (2022) found higher ingestion rates in carnivorous and omnivorous freshwater fish than in herbivores. Similarly, Hastuti *et al.* (2019) observed interspecific variation, with *Sardinella fimbriata* showing the highest average microplastic count. Furthermore, Zhang *et al.* (2023) reported distinct immunological responses in herbivorous fish like the grass carp, likely influenced by their digestive systems and feeding modes.

Organ-specific distribution

This study examined microplastic accumulation in the digestive tract, gills, and muscle tissue. The highest abundance was found in the gastrointestinal tract, especially in carnivorous fish, and predominantly in the form of fibers (Fig. 8). The digestive system is most frequently contaminated due to ingestion of plastic mistaken for food (Bessa *et al.*, 2018; Franzellitti *et al.*, 2019). Gills also showed notable contamination, as their constant exposure to the aquatic environment makes them susceptible to particle absorption (Rasta *et al.*, 2023). Fine fibers can embed in gill lamellae, increasing tissue exposure and risk of damage (Yona *et al.*, 2022, Li *et al.*, 2023).

Microplastic particles that persist in the gastrointestinal tract may translocate to other tissues via the bloodstream. This systemic distribution can reach the liver, muscles, and even the brain (McIlwraith *et al.*, 2021; Putri *et al.*, 2023; Mondal *et al.*, 2024 Ghosh, 2025). For instance, Hossain *et al.* (2024) found 2.60 ± 1.65 microplastic particles per muscle sample, while Singh *et al.* (2024) reported 16 ± 1.4 and 18 ± 1.4 particles/g in the muscle tissue of *Pampus argenteus* from Versova and Bhaucha Dhakka, respectively.

The presence of microplastics smaller than $150\mu\text{m}$ in muscle tissue suggests potential translocation from the gastrointestinal tract via persorption, a passive mechanism allowing particles to cross the intestinal wall. Particles $< 21\mu\text{m}$ may even be taken up by phagocytic cells through active phagocytosis, reaching systemic circulation (Pitt *et al.*, 2024). Earlier studies confirm that small microplastics can migrate to organs like the liver and gills via endocytosis and blood transport (Carr *et al.*, 2012; Franzellitti *et al.*, 2019; Messinetti *et al.*, 2019; Su *et al.*, 2019). Moreover, microplastics may enter through non-dietary pathways—such as wounds or lesions—allowing direct entry into tissues and the bloodstream (Su *et al.*, 2019; Barboza *et al.*, 2020).

CONCLUSION

All fish samples analyzed in this study were found to contain microplastics in the form of fibers, fragments, films, and pellets. The most dominant type of microplastic observed was blue and black fibers, particularly those smaller than 0.25mm , with the highest concentrations found in the digestive tract. Microplastic accumulation varied significantly across sampling locations, with the highest abundance recorded in Location 3, around Oeba Beach. Carnivorous fish exhibited greater microplastic accumulation compared to herbivorous species, indicating a potential correlation between trophic level and the degree of microplastic contamination. This supports the hypothesis that microplastic accumulation increases with trophic level, likely due to trophic transfer through the food web. Further research is recommended to analyze the polymer composition of the microplastics to identify potential pollution sources and better understand their impacts across different trophic levels. In addition, future studies should

investigate the toxicological effects of microplastic exposure on the physiological health of fish. These findings highlight the widespread presence of microplastics across multiple fish species and trophic levels in Kupang Bay. Given that fibers—primarily originating from fishing activities—were the most prevalent form, the results emphasize the need for environmentally friendly, biodegradable fishing gear to reduce plastic input into marine ecosystems.

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