

# Shape Characterization and Organ-Specific Distribution of Microplastics in Indian Squid (*Uroteuthis duvaucelii*) from Dumaguete City Public Market

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## Article Details:

Received: 5 April 2026

Revised: 15 April 2026

Accepted: 22 April 2026

Published: 12 May 2026

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## Recommended Citation:

Cadiz, S. B., Soldivillo, J. S. (2026). Shape Characterization and Organ-Specific Distribution of Microplastics in Indian Squid (*Uroteuthis duvaucelii*) from Dumaguete City Public Market. The International Review of Multidisciplinary Research. 1 (5), 439-445.  
<https://doi.org/10.5281/zenodo.20144964>

## Index Terms:

cephalopods; microplastic morphology;  
microplastic pollution; seafood contamination;  
*Uroteuthis duvaucelii*

**Abstract.** Microplastic contamination in marine food resources has emerged as an increasing environmental and food safety concern; however, data on organ-specific microplastic distribution in commercially sold cephalopods in the Philippines remain limited. This study investigated the occurrence, morphology, and organ-specific distribution of suspected microplastics in Indian squid (*Uroteuthis duvaucelii*) sold in the Dumaguete City Public Market. Using a quantitative descriptive-analytical design, fifteen squid specimens were collected from three market stalls through convenience sampling. Gill and stomach tissues were dissected, chemically digested using 10% potassium hydroxide (KOH), and examined under stereomicroscopy for microplastic identification and morphological classification. A total of 489 suspected microplastic particles were detected across all sampled squid, indicating contamination in 100% of specimens. Of these, 278 particles were recovered from stomach tissues and 211 from gill tissues. Four microplastic morphologies were identified: fragment, fiber, film, and bead. Fragment-shaped particles were the most abundant, comprising 318 particles (65.03%), followed by fibers with 152 particles (31.08%), films with 16 particles (3.27%), and beads with 3 particles (0.61%). Fisher's Exact Test revealed a statistically significant association between microplastic morphology and organ location ( $p = 0.005$ ), indicating that the distribution of microplastic shapes differed between gill and stomach tissues. These findings suggest that Indian squid sold in Dumaguete City may serve as a potential vector of dietary microplastic exposure and provide baseline evidence of microplastic contamination in a commercially important cephalopod species in the Philippines. Continued monitoring using polymer-confirmatory analytical techniques is recommended to strengthen future assessments of seafood contamination and associated food safety implications.

## Introduction

Plastic has become an indispensable material in modern society because of its low cost, durability, and versatility. However, the increasing production and consumption of plastic have also intensified the problem of plastic pollution in aquatic environments. An estimated 19–23 million tonnes of plastic waste enter aquatic ecosystems annually (UNEP, 2025). Through physical, chemical, and biological degradation, larger plastic debris fragments into smaller particles known as microplastics (MPs), commonly defined as plastic particles measuring less than 5 mm in diameter. Microplastics may be classified according to origin as either primary or secondary, with primary microplastics intentionally manufactured at microscopic sizes and secondary microplastics resulting from the fragmentation of larger plastic materials (Shwe, 2022).

Microplastics have emerged as a pervasive environmental contaminant and have been detected in rivers, coastal waters, soil, and atmospheric systems. Their prevalence is expected to increase alongside global plastic production, which rose

from 234 million tonnes in 2000 to 460 million tonnes in 2019 and is projected to reach 736 million tonnes by 2040 (OECD, 2024). Beyond environmental persistence, microplastics present biological concerns due to their capacity to disrupt physiological processes and transport toxic substances such as bisphenol A, phthalates, and heavy metals. Exposure to microplastics has been associated with inflammation, cellular damage, altered metabolism, gut microbiome disruption, and enhanced toxicity of co-occurring contaminants (Ziani, 2023; Dutchen, 2023).

Marine ecosystems are particularly vulnerable to microplastic contamination because land-based and coastal plastic waste is readily transported into the ocean, where it becomes available for ingestion by aquatic organisms (Li et al., 2021). Consequently, commercially important seafood species have become a growing focus of microplastic research. In fish from the southern Baltic Sea, microplastics were detected in the gills, digestive tract, and liver at concentrations ranging from 1 to 12 items per fish (Piskula & Astel, 2023). Similarly, Gong et al. (2021) reported microplastic contamination in the gills, intestine, and stomach of pelagic squid, with concentrations ranging from 4.0 to 7.4 items per individual. These findings indicate that both digestive and respiratory organs may serve as important sites of microplastic accumulation in marine organisms.

The issue is of particular concern in the Philippines, which has been identified as a major contributor to marine plastic leakage. In 2019, the country accounted for 36.38% of global plastic waste emitted to the ocean (Share of Global Plastic Waste Emitted to the Ocean, n.d.). This has been linked in part to the country's widespread reliance on single-use plastic packaging under the so-called "sachet economy." As a result, concerns regarding microplastic contamination have extended beyond environmental systems to food products intended for human consumption. In the Philippines, microplastics have been detected in consumer food items such as sea salt, sugar, fish sauce, and cooked rice (Espiritu et al., n.d.), indicating that dietary exposure may already be occurring through multiple food pathways.

The Philippines is also highly dependent on seafood because of its archipelagic geography and fish-centered dietary culture. In 2022, the country ranked 11th globally in fish production, and seafood remains a major component of the Filipino diet (Canada, 2025). Previous Philippine studies have documented widespread microplastic contamination in marine food organisms. Pustadan (2022) reported that 100% of sampled mussels from selected Philippine sites tested positive for microplastics, while substantial proportions of extracted particles from sampled bangus and mud clams were likewise confirmed as microplastics. These findings highlight the need for further investigation of commercially consumed marine organisms in local market settings.

Among seafood species, cephalopods are of particular interest because of their ecological importance and high consumption rates. Previous studies have shown that microplastic contamination in marine organisms varies according to both particle morphology and organ location. Fibers, fragments, beads, and films are among the most commonly reported microplastic morphologies in biological samples (D'Hont et al., 2021; Singh et al., 2022; Mariano et al., 2021). Organ-specific studies have likewise demonstrated variation in microplastic accumulation across tissues, with some reports indicating greater concentrations in digestive organs and others identifying gill-associated exposure pathways (Cordeiro, 2023; Prata et al., 2022). Such findings suggest that evaluating both microplastic shape and anatomical distribution may provide important insights into contamination pathways and organismal exposure.

The Indian squid, *Uroteuthis duvaucelii*, is a fast-growing cephalopod commonly found in Philippine waters and widely sold for human consumption. Its commercial importance, ecological relevance, and prevalence in local seafood markets make it a suitable species for investigating microplastic contamination in marine food resources. Previous work involving Indian squid and related cephalopod species has demonstrated the presence of fibrous microplastics in digestive and edible tissues, suggesting possible implications for food safety and contaminant transfer to consumers (Chen, 2021).

Despite increasing concern regarding microplastic contamination in Philippine seafood, limited studies have examined the shape-based and organ-specific distribution of microplastics in Indian squid sold in local public markets. Addressing this gap is important for establishing baseline contamination data and improving understanding of potential food safety risks associated with seafood consumption. Accordingly, this study aims to investigate the occurrence and shape-based characterization of microplastics in Indian squid collected from the Dumaguete City Public Market. Specifically, it seeks to answer the following questions: (1) What microplastic shapes are present in the gills and stomach of sampled Indian squid? and (2) Is there an association between microplastic shape and organ location (gills versus stomach) in sampled Indian squid? Through the analysis of microplastic shape and organ-specific occurrence, this study contributes to the growing body of literature on microplastic contamination in Philippine marine food resources and provides baseline evidence relevant to food safety and marine pollution monitoring.

## Methodology

This section describes the research design, sampling procedures, specimen processing, microplastic extraction methods, and statistical analyses employed to investigate the occurrence, morphology, and organ-specific distribution of suspected microplastics in sampled Indian squid.

### *Research Design*

This study employed a quantitative descriptive-analytical research design to determine the occurrence, morphological characteristics, and organ-specific distribution of microplastics in Indian squid samples obtained from the Dumaguete City Public Market. The descriptive component involved the quantification and classification of detected microplastics according to morphological shape and anatomical location. The analytical component involved examining the association between microplastic shape and organ location (gills versus stomach) using inferential statistical analysis. No experimental manipulation was performed, and all observations were based on laboratory examination of collected biological samples.

### *Participants and Sampling Technique*

The study samples consisted of fifteen (15) market-available Indian squid, *Uroteuthis duvaucelii*, purchased from the Dumaguete City Public Market, Negros Oriental, Philippines. Specimens were sourced from three separate fish stalls within the market, with five squid obtained from each stall. The sampled stalls were distributed across different sections of the market to capture variation in vendor sources within the available retail area.

Convenience sampling was employed in the selection of specimens. Fish stalls were included based on their active sale of Indian squid during the sampling period, and the first five available squid from each participating vendor were selected for purchase and inclusion in the study.

Included in the study were whole, market-ready squid specimens morphologically identified as *Uroteuthis duvaucelii* and suitable for dissection and organ extraction. Specimens that were visibly damaged, incomplete, or unsuitable for organ isolation were excluded from the study.

To provide contextual information regarding specimen origin, vendors were informally asked about the sourcing of their squid products. However, most vendors were unable to provide reliable information regarding catch location or supplier origin. Consequently, the geographic origin of the sampled specimens could not be verified and was treated as a study limitation.

Morphometric and biological characteristics of the sampled squid, including weight, body size, and sex, were not recorded and were therefore not included in the analysis.

### *Data Gathering Procedure*

Sampled squid specimens were morphologically identified as *Uroteuthis duvaucelii* based on established diagnostic characteristics described in taxonomic references, including body shape, fin morphology, tentacle structure, sucker arrangement, and presence of photophores (SeaLifeBase, 2025; Marine Biodiversity of Bangladesh, 2025). Identification was further reviewed through consultation with a qualified biologist prior to analysis.

Following collection, specimens were stored under frozen conditions and dissected within approximately five hours of purchase to preserve tissue integrity and minimize gut evacuation, consistent with recommendations for microplastic ingestion studies (Lusher et al., 2017).

Prior to dissection, all work surfaces and instruments were disinfected using ethanol. To minimize external contamination, the researcher wore surgical gloves and a face mask throughout specimen handling, and plastic materials were avoided in the dissection area whenever possible.

Each squid specimen was dissected individually. The mantle was opened using surgical scissors to expose the internal organs. The gills and stomach were selected for analysis because they represent likely routes of microplastic exposure

through respiration and ingestion, respectively. Both gills and the stomach were carefully excised and transferred into individually labeled glass containers. Labeling followed a structured coding system indicating stall number, organ type, and specimen number (e.g., S1G1 = Stall 1, Gill, Specimen 1; S1S1 = Stall 1, Stomach, Specimen 1). A total of 30 organ samples were collected, consisting of 15 gill samples and 15 stomach samples.

Microplastic extraction was performed through alkaline digestion using 10% potassium hydroxide (KOH) solution. Potassium hydroxide is recommended for biological samples with moderate to high lipid content because it digests organic tissues while preserving most plastic polymers and facilitates extraction through triglyceride saponification (Lopes et al., 2022).

Each organ sample was immersed in 20 mL of 10% KOH solution in its respective glass container and allowed to digest at room temperature for 24 hours. Following digestion, the resulting solution was transferred to glass Petri dishes for microscopic examination.

Digested samples were examined under a stereomicroscope for the detection and characterization of suspected microplastic particles. Stereomicroscopy was selected due to its suitability for visual identification of microplastics based on physical morphology and particle appearance (Mariano et al., 2021), although it is acknowledged to have lower confirmatory capability compared with spectroscopic methods such as Fourier-transform infrared (FTIR) spectroscopy.

Suspected microplastic particles were visually classified according to morphological shape categories adapted from established microplastic characterization frameworks (Mariano et al., 2021), namely: fiber, bead, fragment, and film. All identified particles were recorded according to specimen source, organ location, and morphological classification.

#### *Data Analysis Procedure*

Descriptive statistics were used to summarize the occurrence and distribution of microplastics detected in the sampled Indian squid. Frequency counts and percentages were computed to describe: (a) the proportion of squid samples containing suspected microplastics, (b) the total number and percentage distribution of microplastics by organ location (gills and stomach), and (c) the frequency and percentage of each microplastic morphological category (fiber, fragment, bead, and film).

To examine the association between microplastic morphology and organ location, a contingency table was constructed using microplastic shape categories and anatomical location (gills versus stomach). Because some expected cell counts were below the minimum assumptions required for chi-square analysis, Fisher's Exact Test was employed to determine whether a statistically significant association existed between microplastic shape and organ location.

All statistical analyses were conducted using an alpha level of 0.05 for significance. Results were interpreted such that a p-value less than 0.05 indicated a statistically significant association between microplastic shape and organ location.

#### *Ethical Considerations*

This study utilized commercially available squid specimens purchased from a public market, all of which were already deceased prior to acquisition; thus, no live animal handling, experimentation, or euthanasia procedures were involved in the conduct of the study.

All laboratory procedures involving chemical digestion and specimen processing were performed in accordance with standard laboratory safety protocols. Potassium hydroxide (KOH) and other laboratory materials were used solely for research purposes and handled with appropriate personal protective equipment, including gloves and face masks, to minimize occupational exposure and laboratory hazards.

Work surfaces and instruments were disinfected before and after specimen processing to maintain laboratory hygiene and reduce contamination risk. Biological and chemical wastes generated during the study were managed and disposed of in accordance with institutional laboratory waste disposal and safety guidelines.

## Results and Discussion

This section presents the findings of the study in relation to the identified research questions and discusses the observed patterns of microplastic occurrence, morphology, and organ-specific distribution in sampled Indian squid in the context of existing literature.

### *Microplastic Shapes Present in the Gills and Stomach of Sampled Indian Squid*

A total of 489 suspected microplastic particles were identified across all sampled Indian squid specimens, with all squid samples testing positive for microplastic contamination. Of the total detected particles, 211 were recovered from gill tissues and 278 were recovered from stomach tissues. Four microplastic morphologies were observed: fiber, fragment, bead, and film (Table 1).

| Location | Shape        | Gills      | Stomach    | Total      |
|----------|--------------|------------|------------|------------|
|          | Fiber        | 76         | 76         | 152        |
|          | Fragment     | 122        | 196        | 318        |
|          | Bead         | 3          | 0          | 3          |
|          | Film         | 10         | 6          | 16         |
|          | <b>Total</b> | <b>211</b> | <b>278</b> | <b>489</b> |

*Table 1. Distribution of microplastic morphologies in the gills and stomach of sampled Indian squid*

Fragment-shaped particles were the most abundant morphology, comprising 318 particles or 65.03% of all detected microplastics. Of these, 122 were identified in the gills and 196 in the stomach. Fibers were the second most prevalent morphology, accounting for 152 particles (31.08%), with equal occurrence in both organs (76 particles each). Film-type microplastics accounted for 16 particles (3.27%), with 10 found in the gills and 6 in the stomach. Bead-shaped particles were the least frequently observed morphology, comprising only 3 particles (0.61%), all of which were detected in gill samples.

The predominance of fragment-type microplastics suggests that secondary microplastic pollution may be a major contributor to contamination in the sampled squid, as fragments commonly arise from the degradation of larger plastic debris in marine environments (Shwe, 2022). This finding contrasts with several previous studies on marine fish and cephalopods wherein fibers were reported as the dominant microplastic morphology (Sambolino, 2023; Chen, 2021), indicating possible variation in local pollution sources, environmental conditions, or contamination pathways.

The greater abundance of microplastics in stomach tissues relative to gills may indicate that ingestion through feeding represents a major pathway of microplastic exposure in Indian squid. Digestive organs are commonly reported as primary sites of microplastic accumulation due to direct ingestion and trophic transfer of contaminated prey (Gong et al., 2021).

### *Association Between Microplastic Shape and Organ Location*

To determine whether microplastic morphology differed according to organ location, the association between particle shape and anatomical location (gills versus stomach) was examined using Fisher's Exact Test (Table 2).

| Test                | N   | p    |
|---------------------|-----|------|
| Fisher's Exact Test | 489 | .005 |

*Table 2. Fisher's Exact Test for the Association Between Microplastic Morphology and Organ Location*

Results indicated a statistically significant association between microplastic morphology and organ location,  $p = 0.005$ , suggesting that the distribution of microplastic shapes varied significantly between the gills and stomach of sampled Indian squid.

Fragment particles were more frequently observed in stomach tissues ( $n = 196$ ) than in gills ( $n = 122$ ), while bead-shaped microplastics were detected exclusively in gill samples. Fiber occurrence was equal across both organs. These findings

suggest that microplastic morphology may influence organ-specific accumulation patterns within squid tissues. Differences in particle distribution between organs may be explained by distinct uptake pathways and retention mechanisms. Stomach-associated microplastics likely reflect ingestion during feeding, whereas gill-associated particles may represent exposure through respiration and direct water contact (Prata et al., 2022). Particle morphology may affect hydrodynamic behavior, retention, and tissue deposition, thereby influencing anatomical distribution.

However, the association analysis should be interpreted with caution because individual detected microplastic particles were treated as observational units in the contingency analysis. As multiple particles may originate from the same squid specimen, the assumption of statistical independence may not be fully satisfied, potentially introducing pseudoreplication into the inferential analysis. Future studies are encouraged to employ specimen-level analytical approaches or hierarchical statistical models that account for clustering of particles within individual organisms.

## Conclusion and Recommendations

This study demonstrated that Indian squid (*Uroteuthis duvaucelii*) sold in the Dumaguete City Public Market contained suspected microplastics in all examined specimens, indicating widespread contamination among sampled market-available squid. Four microplastic morphologies were identified in the analyzed organs: fiber, fragment, bead, and film. Among these, fragment-shaped microplastics were the most prevalent, followed by fibers, films, and beads.

Organ-specific analysis showed that stomach tissues contained a greater number of suspected microplastic particles than gill tissues, suggesting that ingestion may represent a primary route of microplastic exposure in the sampled squid. Statistical analysis further indicated a significant association between microplastic morphology and organ location, suggesting that particle shape may influence anatomical distribution within squid tissues.

Overall, the findings provide baseline evidence of microplastic contamination in a commercially consumed cephalopod species sold in Dumaguete City and contribute to the growing body of literature on microplastic pollution in Philippine marine food resources. These results highlight the potential for dietary exposure to microplastics through seafood consumption and underscore the need for continued monitoring of microplastic contamination in locally marketed marine organisms. Future studies should incorporate polymer confirmation techniques and specimen-level analytical approaches to strengthen the characterization and interpretation of microplastic contamination patterns.

## Acknowledgement

The authors express their gratitude to the Institute of Environmental and Marine Sciences of Silliman University for providing access to the laboratory equipment used in this study.

## Funding

This research received no external funding from any public, commercial, or not-for-profit funding agency, and no organization provided financial support for the conduct of the study, authorship, or publication of this article.

## Competing Interests Statement

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this article.

## Data Availability Statement

The data supporting the findings of this study are available from the corresponding author upon reasonable request.

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## Appendices

No appendices are attached to this study.