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Occurrence of Microdebris in Muscle of Round Scad *(Decapterus maruadsi)* Collected from Central Vietnam

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Abstract

Round scad is the common dish often consumed in Central Vietnam because of its low price, easy-catching, and high nutritional value. In this study, microdebris includes all synthetic, semi-synthetic, and naturally-derived items extracted from the tissues. The accuracy of the analytical procedure including digestion and polymer flotation methods was tested through the analysis of standardized polymers. The recoveries of polypropylene, polyethylene, polyethylene terephthalate, polyamide and polycarbonate ranged from 94 to 102%. The degree of digestion efficiency in this study was very high, reaching 95 to 99 %. The numbers of microdebris determined in round scad from Phu Yen province and Thua Thien Hue province were 0.9 ± 0.4 and 1.1 ± 0.5 items/g-wet weight and 87 ± 43 and 71 ± 32 items/individual respectively. Only fibers and fragments were found as microdebris in the samples, in which the microfibers were the most commonly-found type with 63% and 89% in samples collected from Thua Thien Hue and Phu Yen provinces, respectively. The level of microdebris accumulated in round scad was relatively high compared with the ranges reported in the literature, suggesting a potentially high risk of humans consuming round scad as daily food.

Keywords: Microdebris; Round scad; *Decapterus maruadsi*; Central Vietnam

1. Introduction

Marine debris is defined as "any persistent, manufactured or processed solid material discarded, disposed of or abandoned in the marine and coastal environment" (UNEP, 2009). In the study of Kroon *et al.* (2018), microdebris sized between 0.1 μm and smaller than 5 mm represented synthetic, semi-synthetic and naturally-derived items. These objects are in different forms of manufactured and modified materials, in which microplastics (MPs) are the common debris. MPs are small, non-biodegradable, and persistent polymers in the environment. They are ubiquitous in the environment and raise many questions to the public because of their adverse impacts on health, biodiversity, and ecosystem (Bellas *et al.,* 2016; Avio *et al.,* 2017; Bessa *et al.,* 2018). Large amounts of continental-origin plastic debris enter the marine environment primarily through rivers (Lebreton *et al.,* 2017), industrial and urban wastewater, and runoff from beach sediments and neighbor fields. The other sources of plastic debris in the marine environment are direct sources, including offshore industrial activities (such as oil and gas exploration), aquaculture activities including the wear and tear of fishnets, and tourism activities that may increase littering at sea (Barboza *et al.,* 2018). In recent years, the pollution of MPs has drawn many scientists' interests since their small size leads to difficulties in quantifying and evaluating their impacts. The influences of MP on the marine ecosystem and human life remain unanswered.

Vietnam is one of the top countries releasing plastic waste into the environment (Jambeck *et al.,* 2015), which poses serious environmental problems (Chau *et al.,* 2020). Therefore, microplastic pollution in Vietnam is an important issue that needs more public attention. In fact, microplastics were found in different environmental matrices in Vietnam such as solid waste (Lahens *et al.,* 2018), coastal sands (Hien *et al.,* 2020), surface water, and sediment (Strady *et al.,* 2021). In addition, MPs were found in green mussels from a central province in Vietnam (Phuong *et al.,* 2019). However, the information on MPs in other common seafood in Vietnam has limited or has not been reported.

Round scad (*Decapterus maruadsi*) belongs to the family of mackerel, has high nutritional and economic value, and is exploited and used in many parts of the world. In Vietnam, round scad is caught mainly in the central provinces. They can be caught all year round, but the main fishing season is around May to July. Round scad hydrolysate has been reported to have strong free radical scavenging and antioxidant (Jiang *et al.,* 2014; Zhang *et al.* 2020). According to Jiang *et al.* (2014), two novel peptides isolated from the protein of round scad can be developed into antioxidative ingredients in supplementary foods. Investigation on the occurrence of microdebris in the edible muscle of round scad provides useful information for understanding the health risk caused by marine pollution.

In this study, the accumulation of microdebris in the edible fish muscle of round scad was evaluated. The aims of the study include 1) to evaluate and compare the occurrence of microdebris in the muscle of round scad in Thua Thien Hue and Phu Yen provinces and 2) to investigate the characteristics of microdebris, including shape, color, and size distribution, accumulating in the collected round scad. The rationale of this approach will provide useful information to raise public awareness about the pollution caused by microdebris and call for actions of reducing microdebris discharge into the environment.

2. Materials and Methods

2.1 Sample collection

Thirty-two round scads (*Decapterus maruadsi*) were sampled in Central Vietnam in July 2020. In Thua Thien Hue province, samples were collected from the biggest seashore market in Hue City, while samples in Phu Yen were collected at Tuy Hoa market as presented in Figure 1. All fish samples were wrapped in aluminum foil and kept at 4 ^{0}C in cool containers filled with ice during transportation to the laboratory. The length, width, and weight of each fish were recorded prior to dissection as shown in Table 1. The fish muscle samples were homogenized after removal of skin, bones, and viscera before being digested by 10% KOH.

2.2 Samples preparation, digestion and observation

The procedures for digestion, extraction, and separation used in this study were adapted from Karami *et al.* (2017). Briefly, 60 mL of 10% KOH was added into each flask containing 6 g of homogenized fish muscle. After being covered by aluminum foil, the flasks were placed in an oven at 50 °C for 48 hours, and then at ambient temperature for 24 hours before proceeding to the next steps. Approximately 15 mL of 4 M NaI solution was added to each flask.

Table 1. Characteristics of round scad collected in Central Vietnam

Location	n	Length (cm)	Width (cm)	Whole weight (g)	Muscle weight (g)	Habitat
Thua Thien Hue	16	21 ± 0.9	3.8 ± 0.4	87 ± 12	59 ± 10	Pelagic, reef-associated
Phu Yen	16	24 ± 1.2	4.3 ± 0.2	111 ± 5.0	99 ± 6.0	

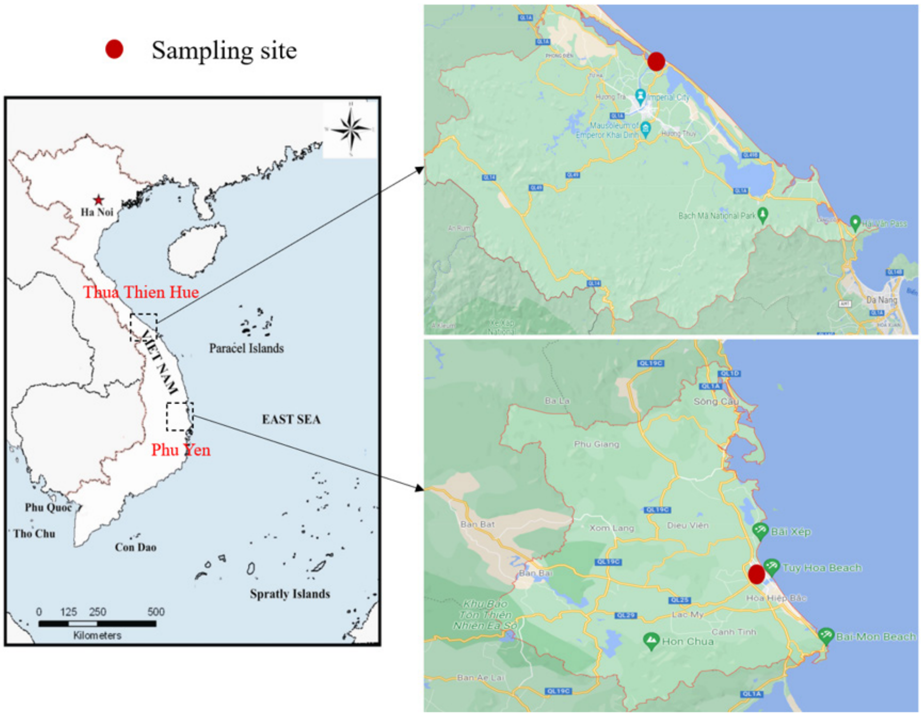

Figure 1. Sampling position in Thua Thien Hue and Phu Yen provinces

The liquid was mixed by an ultrasonic bath (Power-Sonic 420, Korea) and kept stable overnight. The supernatant was separated and directly filtered over a microfiber filter (Whatman GF/B) using a vacuum system. The remaining parts were continued to dissolve with NaI to separate the supernatant through the filter membrane. This process was repeated three times. Finally, the filter was placed into a clean petri dish with a cover for further measurement.

10% KOH was diluted from 45% (w/v) KOH ultrapure solution (Sigma-Aldrich, USA) and 4 M NaI was prepared from NaI powder ultrapure grade (Sigma-Aldrich, USA). Whatman glass fiber filter papers (GF/B, No.1821-047) with 47 mm diameter and 1.0 µm particle retention rate were used during the sample processing procedure.

The filter paper containing extracted microdebris was observed under a YM0745-RT3L Trinocular Stereo Microscope (Shodensha, Japan) and photographed with an HDCT-500DN3 camera. A visual assessment was conducted to identify the particles according to their physical characteristics. Microdebris was classified as fibers (including line) and fragments (including film). In order to differentiate microdebris and other items (sand or undissolved fish tissues), a hot needle was used as described by previous studies (Witte *et al.,* 2014; Khuyen *et al.,* 2021). When a hot needle pointed directly at the extracted item, the microdebris would produce odor of smelting and get curl especially fibers, while no reaction is observed on the sandy item is examined.

2.3 Quality control and quality assurance (QA/QC)

A procedural blank extraction without muscle was carried out in parallel with the fish muscle samples to control contamination during the analysis. All equipment and containers used for the experiments were made of glass and metal to prevent microplastic contamination during the experiments.

The degree of digestion efficiency, % *H*, was calculated to test the amount of undigested organic and/or inorganic materials remaining on the filter according to Eq. (1).

$$\% H = \frac{m_S - (m_a - m_0)}{m_S} \times 100 \quad (1)$$

where: m_S is the initial weight of the muscle sample; m_a is the weight of dry filter paper after filtration, and m_0 is the weight of dry filter paper before filtration.

The accuracy of the analytical procedure including digestion and polymer flotation methods was tested through the analysis of standardized polymers and the recoveries (% Rev) of polypropylene (PP), polyethylene (PE), polyethylene terephthalate (PET), polyamide (PA), polycarbonate (PC) were calculated according to Eq. 2 as follows:

$$\% Rev = \frac{(m_a - m_0)}{m_{Spiked\ MPs}} \times 100 \quad (2)$$

where: m_a and m_0 are above description and $m_{spiked\ MPs}$ is mass of polymer standards added (PP, PE, PET, PA, PC).

3. Results And Discussion

3.1 Abundance of microplastics in fish

The degree of digestion efficiency, % *H*, in this study was very high, reaching 95 to 99 %, within the range of optimum digestion efficiency reported in previous studies (Cole *et al.,* 2014; Karami *et al.,* 2017). The recoveries (% Rev) of PP, PE, PET, PA, and PC ranged from 94 to 102%, indicating that the methods of sample decomposition and polymer flotation achieved high efficiency and were suitable for microdebris analysis in the tissue samples.

Microdebris was found in all round scad muscles collected from Phu Yen and Thua Thien Hue provinces with the number of 87 ± 41 and 65 ± 30 items/individual, respectively. The average numbers of microdebris found in fish muscles by wet weight were 0.9 ± 0.4 and 1.1 ± 0.5 items/g-ww in Phu Yen and Thua Thien Hue, respectively (Figure 2). A procedural blank extraction without muscle showed that no microplastics were detected in these blank samples. Statistically with Tukey test, it revealed that there is no significant difference in the number of microdebris in round scads, based on mass or individual, between samples collected from Phu Yen and Thua Thien Hue ($p > 0.05$). In the work of Ory and colleagues (2017), there was high detection frequency of MPs in amberstripe scad caught from Chile with 80% individual ingested at least one MP item.

In comparison with other studies, levels of microdebris found in our study are significantly higher than those reported worldwide. As shown in Table 2, the levels of MPs found in the round scad samples are higher than that in Indonesia with 0 – 21 MPs/individual (Rochman *et al.,* 2015) or that in the south China Sea with 0.222 MPs/individual (Koongolla *et al.,* 2020). Besides, levels of microdebris in our study are significantly higher than those in other species reported in Iran, UK and China

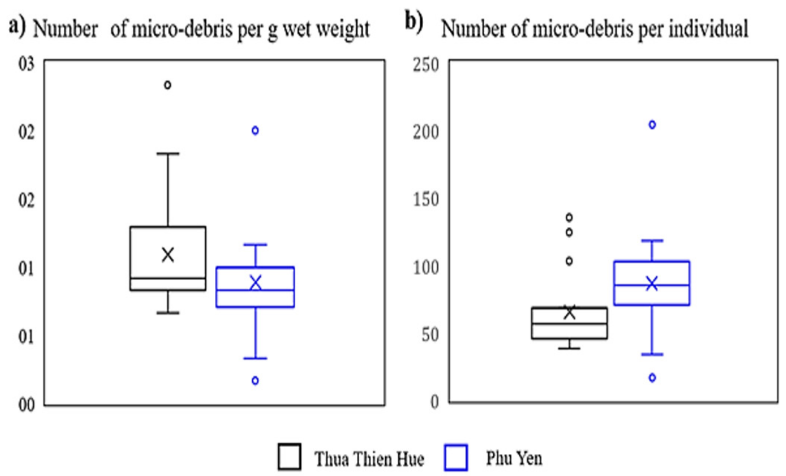

Figure 2. Abundance of microdebris in fish: (a) items/g-ww and (b) items/individual (n = 16).

(Abbasi *et al.,* 2018; Barboza *et al.,* 2020; Li *et al.,* 2021). However, the number of microdebris in this study is similar to those in Shrimp scad (*Alepes djedaba*) and Orange-spotted Grouper (*Epinephelus coioides*) collected from Khark Island, Iran (Akhbarizadeh *et al.,* 2018). The high abundance of microdebris accumulated in round scad muscle suggests that microdebris could be accumulated through the food web and a higher level of microdebris contamination might occur.

3.2 Shape of microdebris

Fibers and fragments were the two main shapes of microdebris found in the round scad samples from Central Vietnam as presented in Figure 3. In which, microfibers were mainly observed as The predominant shape of microdebris found in fish samples both from Thua Thien Hue and Phu Yen was microfibers with 63% and 89%, respectively. Microfibers found in fish muscles included single fibers and tangled fibers, while fragments included amorphous shapes as presented in Figure 4. The results are in line with those observed in fish muscles reported in literature with the high proportion of fibers, followed by fragments (Mathalon and Hill, 2014; Li *et al.,* 2015; Abbasi *et al.,* 2018; Hosseinpour *et al.,* 2021). MPs found in fiber shape were in the range of 70 – 100% in varying fish species, which were reported by various studies (McGoran *et al.,* 2017, Peters *et al.,* 2017, Sathish *et al.,* 2020; Hosseinpour *et al.,* 2021). The origin of microfibers in fish muscle coming from various sources that includes laundry discharges (Bessa *et al.,* 2018), breaking of fishing gear (e.g., ropes and nets), or plastic decomposed from larger pieces. However, some studies indicated that the MP fragments were found as the dominant shape in fish (Ory *et al.,* 2017), water, and sediment (Zhang *et al.,* 2017; Part *et al.,* 2020). The major conformational differences of MP shapes in fish might be influenced by various factors including living environment, properties of primary materials, fragmentation processes, digestion, and accumulation mechanisms.

Table 2. Comparison of microplastic pollution in fish muscle in the present study with those in previous studies

Country	Species	Part of body	Microdebris (items/individual)	Microdebirs (items/g-ww)	Size (μm)	Reference
Vietnam	Round scad	Muscle	17 – 204	0.2 – 2.3	100 – 500 (mainly)	This study
China	Pond fish	Muscle	3–92	n.a.	n.a.	Li *et al*., 2021
South China Sea	Round scad	Gastrointestinal tracts and gills	0.222	n.a	n.a	Koongolla *et al*., 2020
UK	*D. labrax;*	Muscle	n.a.	0.4 ± 0.7	501 – 1500	Barboza *et al*., 2020
	T. trachurus;		n.a.	0.7 ± 1.3		
	S. colias		n.a.	0.6 ± 0.8		
Khark Island (Iran)	*A.djedaba*	Muscle	n.a.	0.80 ± 0.12	n.a.	Akhbarizadeh *et al*., 2018
	E.coioides		n.a.	0.775 ± 0.22	n.a.	
Iran	*S. sihama*	Muscle	14.1	0.25	100 – 500 (mainly)	Abbasi *et al*., 2018
	C. abbreviatus		12.0	0.16	250 – 500 (mainly)	
Chile	Amberstripe scads	Guts	2.5 ± 0.4	n.a.	1.3 ± 0.1 (mm)	Ory *et al*., 2017
Indonesia	Round scad	Gastrointestinal tracts and gills	0-21	n.a.	n.a.	Rochman *et al*., 2015

n.a. = not available

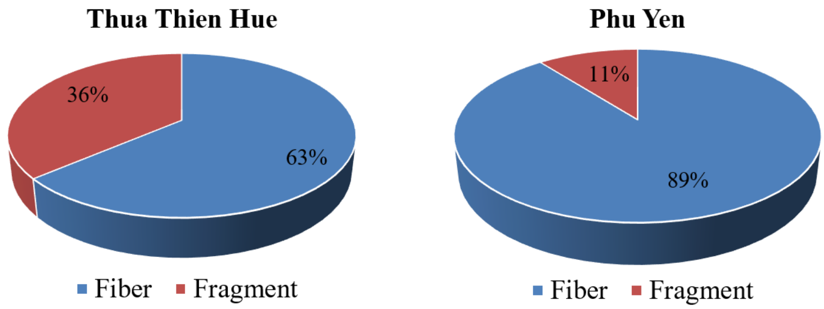

Figure 3. The chart of the composition of microdebris shapes as fiber or fragment found in the round scad samples.

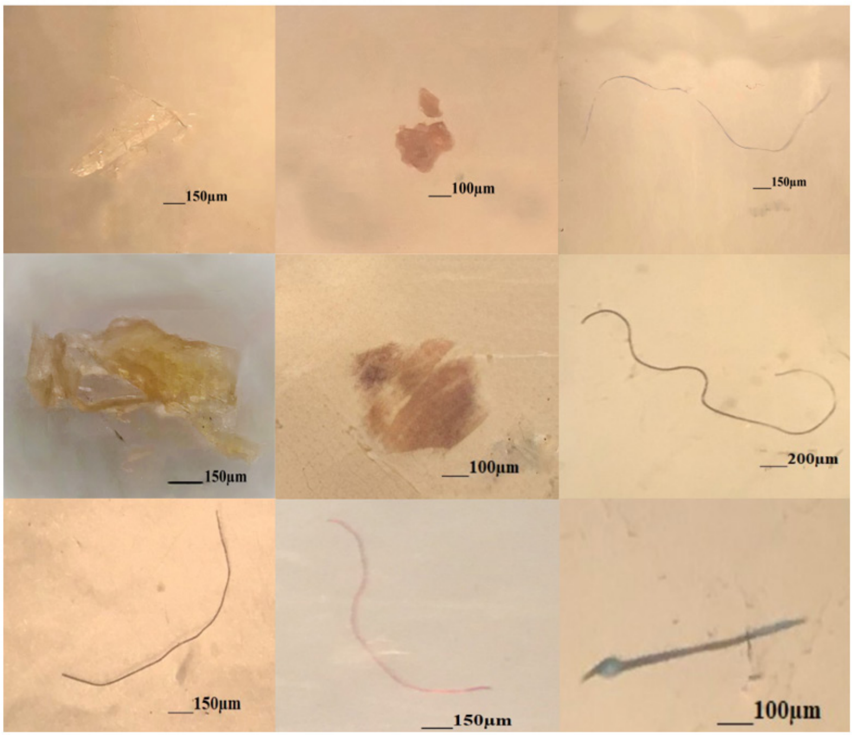

Figure 4. Photographs of microdebris in the muscle of round scad showing different shapes and colors (The images were taken directly on the filter paper)

3.3 Colour distribution of microdebris

The colour distribution of microdebris visibly observed and recorded is shown in Figure 5. Overall, for samples collected from Thua Thien Hue province, white-transparent was the most popular colour found in all samples, accounting for 41%, followed by blue-green with 18%. Black-grey and red-pink of microdebris contributed approximately 17% and 15%, respectively and the yellow-orange was the least common colour with about 9% (Figure 5a). Similarly, Koongolla *et al.* (2020) reported that the transparent (83%) was also the most common colour of microdebris in fish samples collected in the territorial waters of Vietnam (South China Sea), implying the similarity in colour of microdebris found in fish caught in the nearby area.

In contrast, for samples from Phu Yen province, black-grey was the most popular colour found in all samples, accounting for 33%, followed by white-transparent with 27%. Yellow-orange was not the least common colour with about 15%, equivalent to red-pink. Blue-green was the least common colour with about 9% (Figure 5b). This result is similar to that reported by Abbasi *et al.* (2015) and Hosseinpour et al. (2021), which revealed that black-grey MPs in fish samples were the highest at 71.1% and 64%, respectively.

3.4 Size distribution of microdebris

The size distributions of microdebris found in the scad muscle samples are shown in Figure 6, which indicate increasing abundance of microdebris with decrease in sizes. In this study, the sizes of microfiber debris were a wide range of ≤ 100 µm in diameter and several hundred µm to a few mm in length. The diameter of fragments found in fish muscle ranged from ≤ 100 µm to several hundred µm. In general, the sizes of < 100, 100 – 200, 200 – 500 µm dominated in the round scad muscle samples in Thua Thien Hue accounting for 35%, 21%, and 23%, respectively. Meanwhile, these ratios in the round scad muscle samples in Phu Yen were respective 66%, 11%, and 8% (Figure 6).

Therefore, the common size found in fish muscles was < 500 µm with the proportion of 80% of total microdebris. The results are in line with previous studies which reported that the size of MPs in other fish species is smaller than 500 µm (Abbasi *et al.*, 2018; Klangnurak and Chunniyom, 2020; Hosseinpour *et al.* 2021). Plastic and non-plastic materials enter the ocean and undergo mechanical changes that break larger debris into smaller ones. These tiny pieces are often mistaken as food and consumed by many different aquatic organisms.

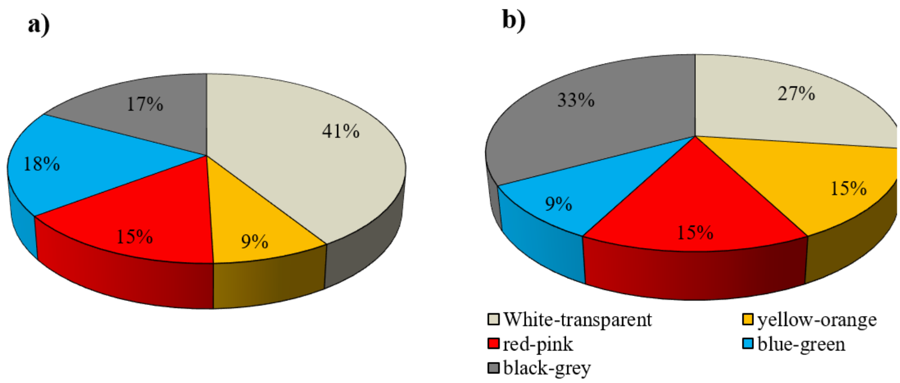

Figure 5. The chart with the overall colour distribution of microdebris observed in round scad collected from a) Thua Thien Hue and b) Phu Yen

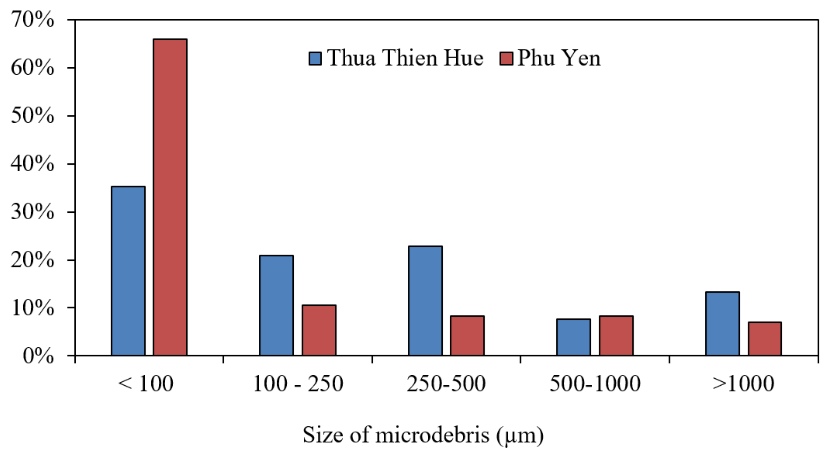

Figure 6. The size distribution of microdebris in the round scad samples

4. Conclusion

As the first investigation on microdebris in round scad muscle samples in Phu Yen and Thua Thien Hue provinces, this study successfully separated, evaluated the occurrences, and characterized morphology of microdebris in muscles of the round scad – a common seafood in Vietnam. The results reveal that a quite high level of microdebris was found in the round scad samples collected from Central Vietnam. This suggests that the occurrence of microdebris in marine fish is unneglectable, and posed a potentially high risk of humans consuming round scad in particularly, and marine fish in general as daily food. Microfiber was the dominant shape, accounting for 63 and 89% for samples collected from Thua Thien Hue and Phu Yen provinces, respectively. While the most commonly-found color of microdebris was white-transparent, followed by blue-green, black-grey, red-pink, and yellow-orange. Most microdebris were observed in the size range of < 500 μm with the dominance of size < 100 μm. The different characteristics of microdebris, including level, colour, shape and size distributions, found in Thua Thien Hue and Phu Yen provinces suggest that microdebris pollution (levels, sources, composition of original debris) in these areas might be different. Therefore, further studies on characterizing microdebris pollution in surrounding marine environment are deemed necessary. The most popular shape of microdebris as fibers, indicating some possible sources of microdebris in the fish muscles. Further studies should be conducted to identify the chemical compositions of microdebris to understand and predict their origins. Any form of accumulation, the presence of microdebris in round scads in Central Vietnam raises concerns about the potential risks for the transfer of the synthetic material into humans.

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Conflict of interest

The authors declare no conflict of interest.

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