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Anthropogenic particles in the muscle, gill, and gastrointestinal tract of marine fish sold for human consumption

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ABSTRACT

Contamination of marine fish with the widespread distribution of anthropogenic particles (APs) becomes increasingly severe, however, related research on the assessment of the occurrence of APs in the edible tissue of commercial fish is scarce. The objective of this study was to evaluate the features of APs pollution based on seven species of commercial marine fish (n = 12 per species) and investigate the accumulation of APs in different tissues of fish namely gill and gastrointestinal tract (GIT), and muscle. The results show that a total of 62 APs were detected in 33 out of 84 (39.3%) fresh fish samples using a micro-Raman spectrometer which in particular is characterized by a blue color, shape-like fiber, and size smaller than 0.5 mm. Among them, 47 (75.8%) particles were identified as pigments such as indigo, chrome yellow-orange, disperse yellow, and pigment black. The other 11 (17.7%) particles were plastic including polypropylene (PP), polyethylene terephthalate (PET), and polyacrylonitrile (PAN). And the rest 4 (6.5%) particles were anthropogenic cellulose fibers. Muscle tissue from six species of fish was detected to contain a total of 15 APs. Based on the total mean of APs found in fish muscle (0.018 AP items/g tissue) and on the consumption of fish in Malaysia (59 kg/capita/year), the estimated human intake of APs through fish consumption was 1062 AP items/year/capita. Considering that food consumption is an important route of human exposure to APs, it is suggested to add APs testing into the guidelines of food safety management systems and adopt mitigation measures to reduce the APs pollution in food.

1. Introduction

Anthropogenic particles (APs) are a variety of particles produced directly or indirectly by human activities, which is mainly consisting of plastic, pigments, and textile fibers. The ubiquity of APs in the marine environment and the toxicity of the chemicals associated with APs have begun to raise concerns about the negative effect on human health caused by marine products with accumulated APs. These concerns have prompted a concerted effort by the global scientific community, environmental activists, and the general public to assess the impact of marine APs on human and environmental health, including organizations such as the European

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Food Safety Authority (EFSA), National Center for Ecological Analysis and Synthesis (NCEAS), United States Environmental Protection Agency (US EPA), and National Oceanic and Atmospheric Administration (NOAA). These groups acknowledged the lack of legislation regarding APs as contaminants in foods and reported that there is a necessity to conduct research to elucidate how marine APs pose risks to human health through the food chain [1–3].

Among all the APs that pose a threat to the environment, plastic gains a leading position due to its widespread distribution and difficulty in degrading. Plastic has been found worldwide in the marine environment, extending from the coast to the deep sea, and to the Arctic Sea ice from remote locations [4–6]. It is estimated that there are more than 5 trillion plastic debris (over 250,000 tons) floating at the sea [7] and nearly 800 marine species are affected by plastic debris through entanglement or ingestion [8,9]. Marine organisms such as fish are most likely to incidentally take up microplastics (MPs, ≤ 5 mm) due to their small size, either by swallowing because they confuse them with food or by other processes [5]. Among 25 species that contributed the most to global marine fisheries, 11 were detected to contain MPs [10]. Some MPs are composed of hazardous monomers (e.g., bisphenol-A, polyvinyl chloride, styrene) and chemical additives (e.g., nonylphenol, phthalates), and can sorb various environmental contaminations (e.g., polychlorinated biphenyls, petroleum hydrocarbons, organochlorine pesticides, polybrominated diphenyl ethers, alkylphenols, metals) [11–13]. These hazardous substances could be released in the organisms which swallowed the particles [14].

Pigments are widely used in colorful paintings and coatings to increase the attractiveness of the products, such as wooden products, metal products and many lightweight products made of textile fibers, plastic, or fiberglass [14]. Heavy abrasion could occur in the process of sandblasting buildings or ship maintenance and repair facilities, resulting in the generation of dust and large debris which could subsequently enter the marine environments directly or indirectly via wind and water, likely with the formation of small particles through mechanical weathering and abrasion [15,16]. Metals are usually contained in pigments and paints and might be leached out from particles after swallowing by organisms [17,18].

All textile fibers are APs since all of them have been handled by humans, such as dyed, even though their basic composition is natural [19]. Although synthetic fibers as a form of plastics are difficult to degrade in the environment and have attracted more attention from researchers, anthropogenic cellulose fibers are also not easily degraded and could be accumulated in the environment [20,21]. Moreover, even textiles made from natural fibers may contain up to 27 % added chemicals by weight and are also not harmless [20,22,23]. Therefore, all kinds of textile fibers that appeared in organisms should not be overlooked as they could also contribute to water pollution.

APs ingested by organisms have been proved by some experiments to have the potential to be transferred to higher trophic levels. And through bioaccumulation and biomagnification, eventually, APs and toxic pollutants will have the potential to pose a threat to human health [24]. To understand the potential impacts of marine APs on human health, the first step is to determine whether APs are present in seafood such as fish that are caught and sold for human consumption.

Although some studies have reported the presence of APs in wild-caught fish commonly consumed by humans, there are limited studies demonstrating the presence of APs in fish sold directly in wet markets for human consumption. In this study, seven commonly commercial marine fish were collected directly from a wet market in Kota Kinabalu, Malaysia, to measure the presence of APs in fish. In addition, at present, only a few studies emphasize the content of APs in the muscle of fish especially in Southeast Asia, while most studies focus on detecting the APs loads in the gastrointestinal tract (GIT, including intestine and stomach) [25]. Unfortunately, APs that are retained in the digestive tract may be further transferred to other tissues, even the edible tissues like the muscle of fish, and pose health risks to consumers even if they only consume eviscerated fish [1,8,26]. In this study, we not only detected AP levels in the gill and GIT of fish but also determined whether APs were transferred to the muscle of these fish species. Meanwhile, the abundance, color, size, and morphotype of all isolated APs were recorded and their chemical composition was analyzed using micro-Raman spectroscopy. The results of this study can help people understand the level of APs contained in commercially available edible fish, thereby inferring the possible health risks to consumers, and providing reference data for the development of relevant food safety regulations.

2. Materials and methods

2.1. Sample collection and preparation

From September 2020 to December 2021, samples of 7 types of marine fish species Orange-spotted grouper (*Epinephelus coioides),*

Table 1
The fork length and wet weight of the analyzed fish species (n = 12 per species examined).

Common name	Species	Fork length (cm)		Wet weight (g)	
		Mean	Range	Mean	Range
Orange-spotted grouper	*Epinephelus coioides*	26.4 ± 2.8	20.1–30.0	290.0 ± 91.3	123.8–390.4
Malabar trevally	*Carangoides malabaricus*	26.5 ± 4.6	21.9–34.6	446.8 ± 215.9	225.6–904.4
Yellowtail Scad	*Atule mate*	22.8 ± 2.2	18.3–27.2	204.3 ± 56.62	111.45–331.9
Mackerel tuna	*Euthynnus affinis*	29.4 ± 6.4	25.0–45.5	471.2 ± 349.1	256.1–1425.0
Delagoa threadfin bream	*Nemipterus bipunctatus*	21.6 ± 1.7	18.5–24.0	199.0 ± 59.9	116.5–306.9
Red Bigeye	*Priacanthus macracanthus*	23.1 ± 2.7	18.0–26.0	193.1 ± 60.5	85.8–279.4
Pickhandle barracuda	*Saurida tumbil*	32.1 ± 5.8	22.9–40.5	255.2 ± 104.2	87.3–404.5

Malabar trevally *(Carangoides malabaricus),* Yellowtail Scad *(Atule mate),* Mackerel tuna *(Euthynnus affinis),* Delagoa threadfin bream *(Nemipterus bipunctatus),* Red bigeye *(Priacanthus macracanthus),* Pickhandle barracuda *(Saurida tumbil)* (12 per species) were purchased from the wet market in Kota Kinabalu, Malaysia, with a total purchase of 84 marine fish. All fish were caught from fishing areas off the west coast of Sabah. The samples were kept in pre-cleaned cold boxes with ice to keep the temperature below 0 °C and transferred to the laboratory. The fork length from the anterior part of fish to the median point of caudal fin rays and the wet weight of each fish were recorded to the nearest 0.1 cm and 0.1 g, respectively [27] (Table 1).

2.2. Extraction of particles

The muscle of each fish was weighed 10 g, and the whole gill and GIT were taken and weighed, respectively, using an electronic weighing balance (PS3500.R1, RADWAG, Poland). Then their weights were recorded.

The extraction of particles from fish was carried out according to the method of Jabeen et al. (2017) and Abbasi et al. (2018) [1,27]. The tissues of fish were separately placed into 500 mL glass bottles and then approximately 100 mL of 30% H_2O_2 (SYSTERM) was added to each bottle to digest organic matter. These bottles were placed in the oscillation incubator (WIS-20, Wise Cube, Korea) at 60 °C with 80 rpm for 48–72 h. To stop the digestion and maintain the integrity of particles, the solution was diluted with 200 mL of ultrapure (Milli-Q) water. Then the particles were separated from the tissue residue by stirring the diluent using a glass rod and centrifuging for 5 min at 4000 rpm. The supernatant was filtered through an 11-μm pore size, 42.5-mm cellulose filter (Whatman Grade 1), and then these filter papers were placed into clean Petri dishes.

2.3. Observation and identification of anthropogenic particles

The particles retained on the filters were observed under a microscope (ECLIPSE E400 POL, Nikon, Japan), and images (40–100 magnification) were taken. A visual assessment was made according to the physical characteristics of particles, such as size, color, and morphology [8,28]. Particles were categorized into four morphotypes (fragment, fiber, sphere, and flake), five sizes ($\leq$200, 200–500, 500–1000, 1000–5000, and >5000 μm), and seven colors (blue, red, yellow, black, green, brown, and white).

All visually identified APs are analyzed over a range of 100–3000 cm^{-1} using a micro-Raman spectrometer (XploRA Plus, Horiba, Japan) coupled with an EM-CCD detector (Jobin Yvon, Horiba, Japan) and a confocal microscope using a laser beam excitation of $\lambda =$ 785 nm with 100× objective and 5 accumulations. To increase the quality of spectra, the baseline correction (Horiba Labspec 6) was applied to particles before the library search. The identification of polymers was carried out using the reference spectral libraries from Bio-Rad.

2.4. Contamination control

To prevent the contamination of APs, all processing of fish occurred inside a clean bench with particle filtration, and the following steps were taken in the laboratory due to micro-APs being ubiquitous in the indoor environment [29]. Before using the filter paper, it was observed with a microscope to ensure that no suspected APs were attached to it. All chemical reagents were filtered before use. White cotton lab coats and nitrile gloves were worn throughout the experiment. The surfaces and apparatus such as glassware and tools were thoroughly cleaned with 70% ethanol [1]. In the laboratory, fish samples were rinsed with ultrapure water before being dissected and the samples that were not in use would be covered immediately. To assess the possible presence of AP contamination in the air, three clean Petri dishes were placed directly next to the workplace in all procedures and analyzed as procedural blanks. In addition, to assess the potential AP contamination from apparatus and chemicals, three procedural blanks (without tissues, replacing fish samples with ultrapure water) were processed, filtered, and analyzed in parallel with fish samples. No AP was found in the blanks.

2.5. Estimated human exposure to anthropogenic particles through fish consumption

The approach for estimating human exposure to APs from fish consumption is based on the data from Food and Agriculture Organization of the United Nations (FAO) on per capita fish consumption in Malaysia (59 kg/year/capita) in 2016 [30] and on the mean of the number of APs in muscle considering the seven fish species:

Human AP intake per year per capita (AP items/year/capita): mean of AP items in the muscle tissues (AP items/g) × fish consumption per year per capita in Malaysia (g/year/capita)

2.6. Data analysis

Statistical analyses were carried out using IBM SPSS Statistics 25.0 software. The one-way analysis of variance (ANOVA) was used to evaluate the significance of the experiment results at 95% confidence intervals. Significant differences were recorded at $p < 0.05$.

3. Results

3.1. Chemical characteristics of anthropogenic particles

A total of 81 visually identified APs were detected in all 7 types of fish species and 62 particles were confirmed as APs using a micro-

Raman spectrometer. The microscopic images of some APs are shown in Fig. 1 (A-F). As shown in Fig. 2A, these 62 APs included 47 pigment particles (75.8%), 11 plastic particles (i.e., particles confirmed as plastic or plastic plus pigment) (17.7%), and 4 anthropogenic cellulose fibers plus pigment (6.5%). There were significant differences ($p < 0.05$) in the concentration of these three different types of particles in fish samples (Fig. 3A). The most abundant pigments were indigo (42.6%), followed by chrome yellow-orange (14.9%), disperse yellow (12.8%), pigment black 9 (8.5%), cadmium orange medium (6.4%), pigment brown 7 (4.3%), copper phthalocyanine (2.1%), acid yellow 76 (2.1%), deorlin blue 5 (2.1%), permanent bordo (Violet, 2.1%), and pigmosol green (2.1%) (Fig. 2B). And there were significant differences ($p < 0.05$) among the concentration of different kinds of pigment particles in fish samples (Fig. 3B). Particles identified as plastics were polypropylene (PP, 45.5%), polyethylene terephthalate (PET, 45.5%), and polyacrylonitrile (PAN, 9.1 %) (Fig. 2C). There was no significant difference ($p > 0.05$) among the concentration of plastic particles with different chemical compositions in fish samples (Fig. 3C). Meanwhile, all particles identified as anthropogenic cellulose fibers contained pigments, which included modal plus reactive red 4 (25.0%) and cellulose acetate sorbate plus acid orange 74 (75.0%) (Fig. 2D). There was no significant difference ($p > 0.05$) between the concentration of modal particles and cellulose acetate sorbate particles (Fig. 3D). Fig. 4 (A-E) shows the Raman spectra and photographs of a few representative APs and spectra of their corresponding reference materials, including indigo, disperse yellow 9, polypropylene (PP), PP plus pigmosol green, and modal plus reactive red 4. And the Raman spectra of more identified APs are shown in Supplementary Fig. 1.

3.2. *Physical characteristics of anthropogenic particles*

The results showed that seven colors of APs were found in investigated fish samples and blue color (38.7%) was the dominant color of APs followed by red (30.6%), yellow (11.3%), black (9.7%), green (4.8%), brown (3.2%), and white color (1.6%) (Fig. 5A). There were significant differences ($p < 0.05$) among the concentration of different color particles in fish samples (Fig. 6A). Different shapes of APs (e.g., fragment, fiber, flake, and sphere) were recorded. About 56.5% of collected particles was fiber in shape, followed by fragment (35.5%). The number of flakes (6.5%) and spheres (1.6%) was relatively lower compared with the former shapes (Fig. 5B). And there were significant differences ($p < 0.05$) among the concentration of particles with different shapes in fish samples (Fig. 6B). Meanwhile, the size of APs ranged from 40 μm to 3 cm, and APs smaller than 500 μm were dominant and accounting for 51.6% of all identified APs (Fig. 5C). However, there was no significant difference among the concentration of particles in different size groups (Fig. 6C).

3.3. *Contents of anthropogenic particles in fish tissues*

APs were present in all 7 fish species and the number of APs in different species was no significant difference ($p > 0.05$). Of all 84 fresh fish samples, 33 (39.3%) fish samples were found to contain APs. With each species of fish, we found APs in 50.0% of *Epinephelus coioides,* 33.3% of *Carangoides malabaricus,* 50% of *Atule mate,* 83.3% of *Euthynnus affinis,* 25% of *Nemipterus bipunctatus,* 16.7% of *Priacanthus macracanthus,* 25% of *Saurida tumbil.* Meanwhile, different tissues (gill, GIT, and muscle) of fish were investigated and APs were observed in all of these tissues. The number of APs found inside the gill counts as 40.3% of the total number, while GIT (35.5%)

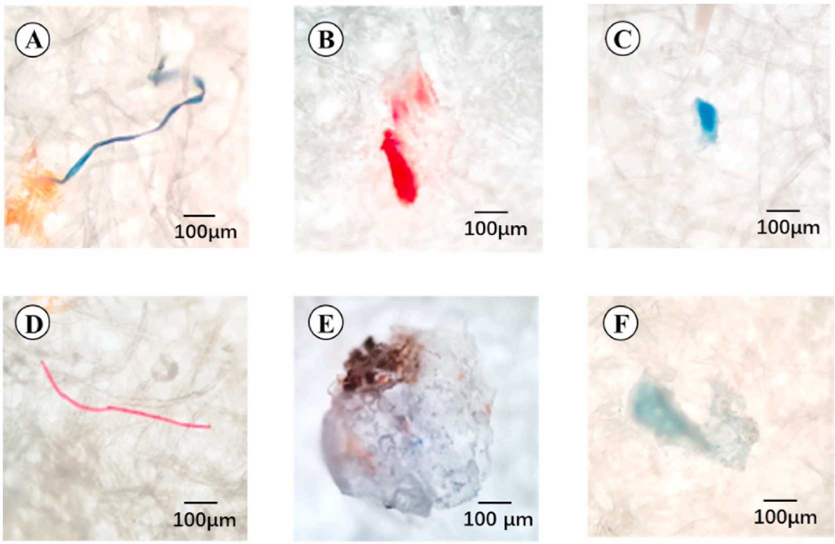


Fig. 1. Photographs of anthropogenic particles in fish from Malaysia. The morphotypes included fiber (A, D), flake (B), fragment (C, F), and sphere (E). Scale bar = 100 μm. The magnification time using a microscope = 100 × . These particles were identified by micro-Raman spectroscopy as (A) Indigo, (B) Chrome yellow-orange, (C) Copper phthalocyanine (pigment blue 15), (D) Polyethylene terephthalate, (E) Polypropylene, (F) Polypropylene + Copper phthalocyanine.

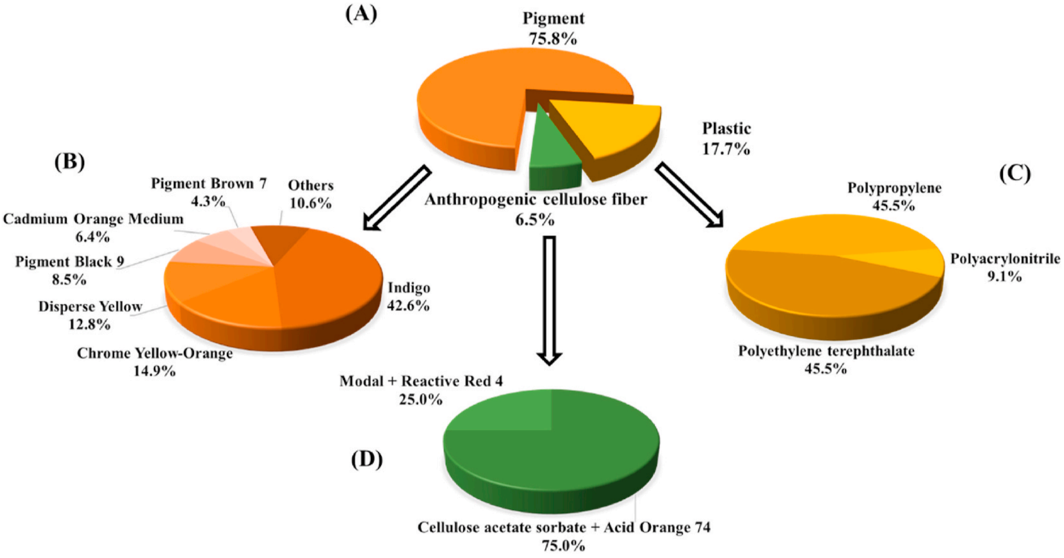


Fig. 2. The compositions of the extracted anthropogenic particles from fresh fish samples. (A) The percentage of different types of anthropogenic particles and their corresponding proportion of (B) pigments, (C) plastics, and (D) anthropogenic cellulose fibers.

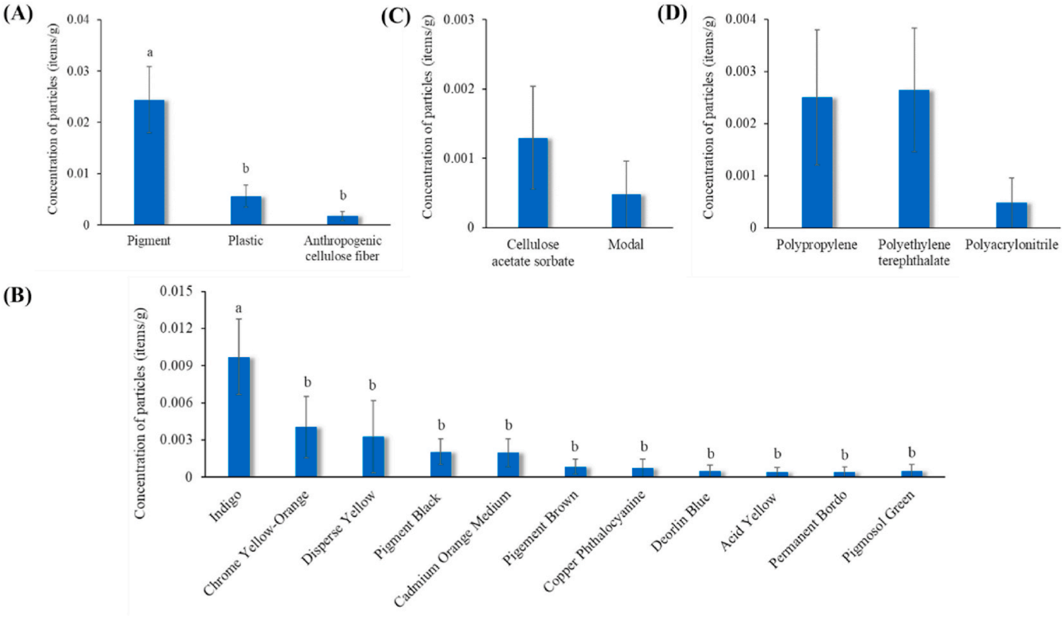
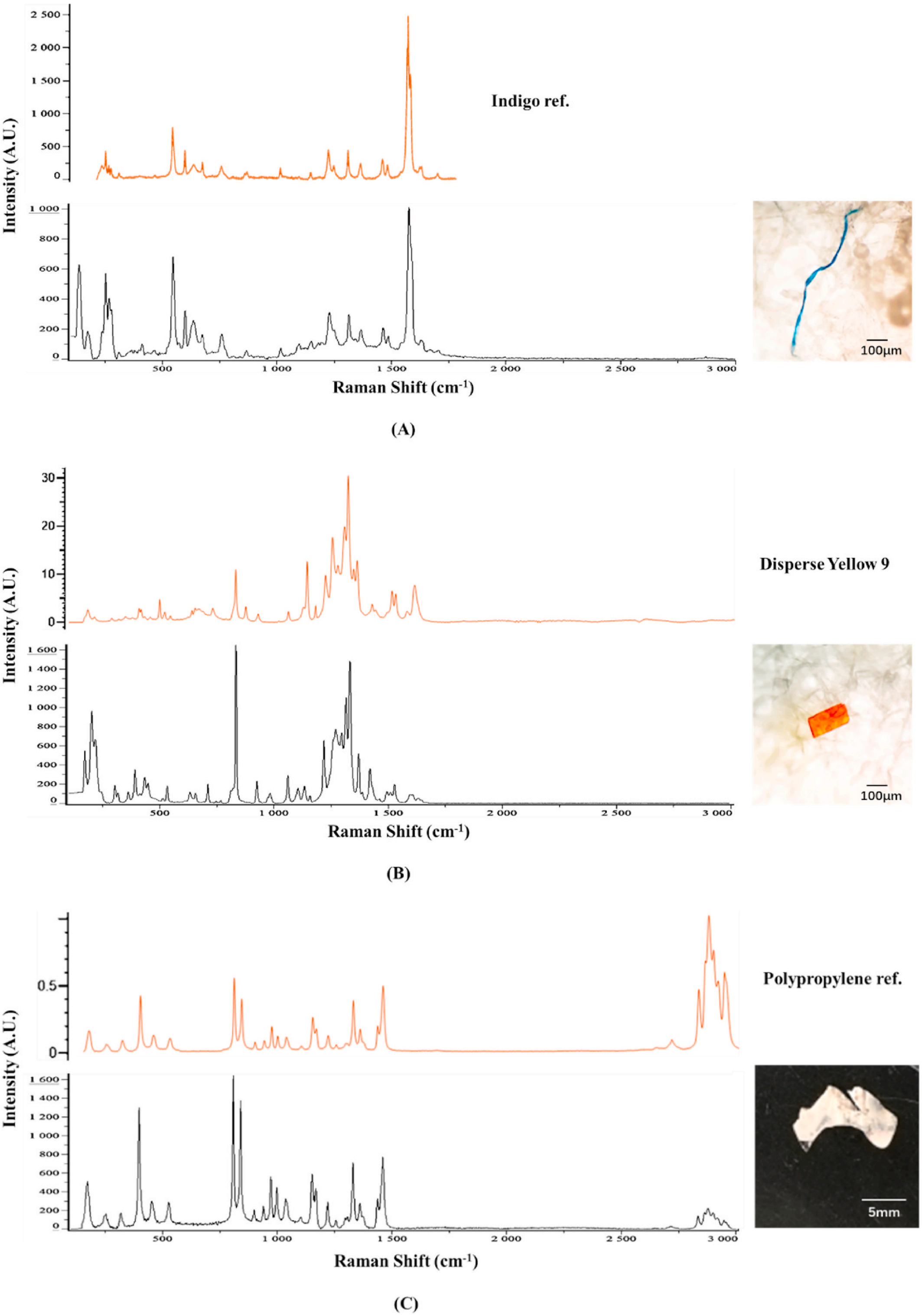


Fig. 3. The concentration of particles in fish samples (mean ± SEM). (A) The concentration of different types of anthropogenic particles and their corresponding concentration of (B) pigments, (C) plastics, and (D) anthropogenic cellulose fibers. The superscript letters indicate significant differences between groups ($p < 0.05$).

and muscle (24.2%) showed less APs as compared to gill (Fig. 5D). The concentration of particles in different tissues was no significant difference ($p > 0.05$) (Fig. 6D). Fig. 7 is a stacked bar chart of the number of APs isolated from the gill, GIT, and muscle of each fresh fish species. The APs observed in the muscle tissue of fish included 12 pigments, 2 anthropogenic cellulose fibers, and 1 plastic. Among muscle of all fish species, only muscle samples of *Priacanthus macracanthus* didn't contain APs.

3.4. *Estimated intake of anthropogenic particles by human consuming fish*

Based on the total mean of APs found in fish muscle (0.018 AP items/g tissue, N = 84) and on the consumption of fish per capita in Malaysia [30], the estimated human intake of APs through fish consumption was 1062 AP items/year/capita.



(A)

(B)

(C)

Fig. 4. Raman spectrum of some anthropogenic particles and spectra of their corresponding reference materials. These particles were identified as (A) indigo, (B) disperse yellow 9, (C) polypropylene, (D) polypropylene + pigmosol green, and (E) modal + reactive red 4.

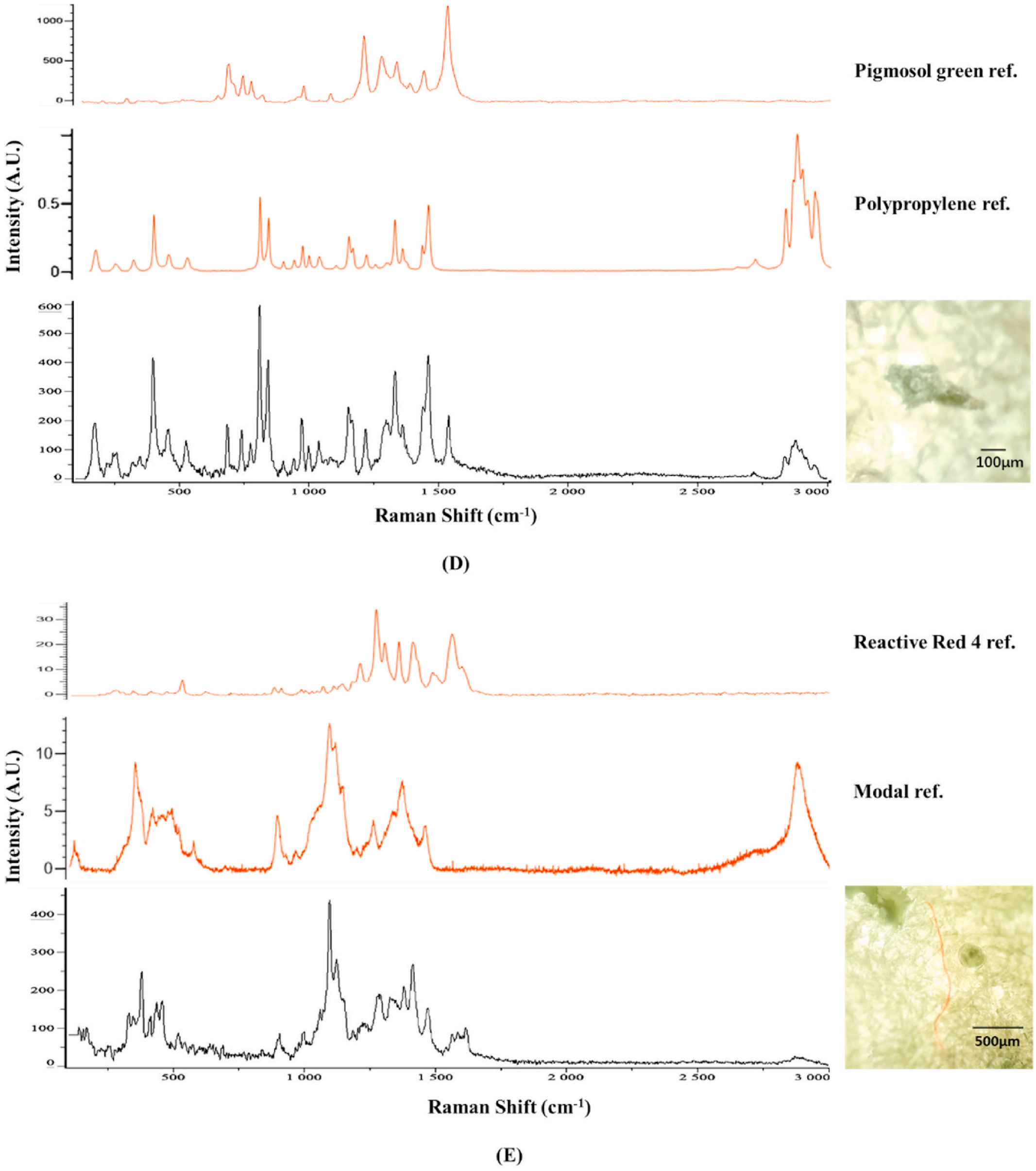


Fig. 4. (*continued*).

4. Discussion

4.1. Contents of anthropogenic particles in fish tissues

In this study, it is observed that the highest abundance of particles was detected in the gill. The gills of aquatic organisms are their first organ exposed to APs during respiration, which increases the possibility of these particles getting stuck among the gill filaments. The result also showed that the muscle of fish as the important edible part was also detected to contain APs. Similarly, Karami et al. (2017) also found MPs in eviscerated flesh of fish, and in some fish species, their MP loads were even higher than the excised organs [31]. Additionally, several studies on fish have confirmed that micro-APs can be transferred from the GIT or gill to other organs including the muscle tissue, which has raised consumers' concerns about the safety of fish products [19,32,33]. These results indicated that removing the gill and GIT of fish during cooking could reduce the risk of ingesting APs for consumers but cannot eliminate it.

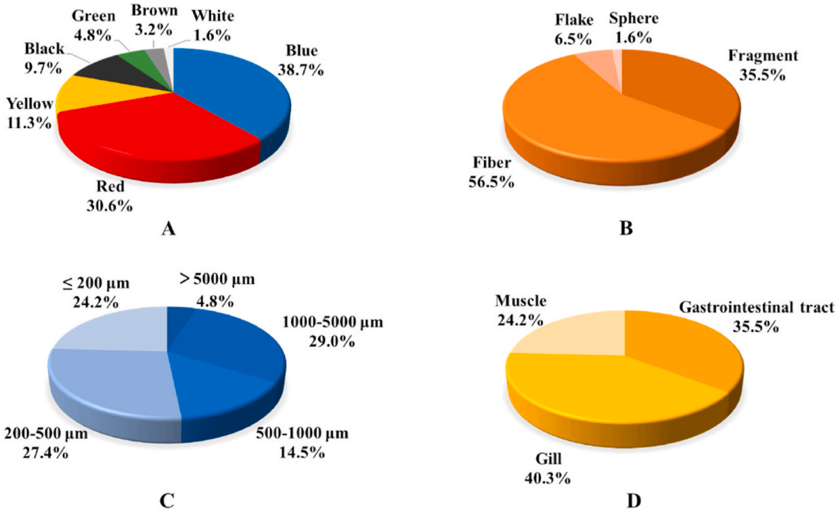


Fig. 5. The percentages of different colors (A), shapes (B), sizes (C), and tissue distribution (D) of anthropogenic particles in fish.

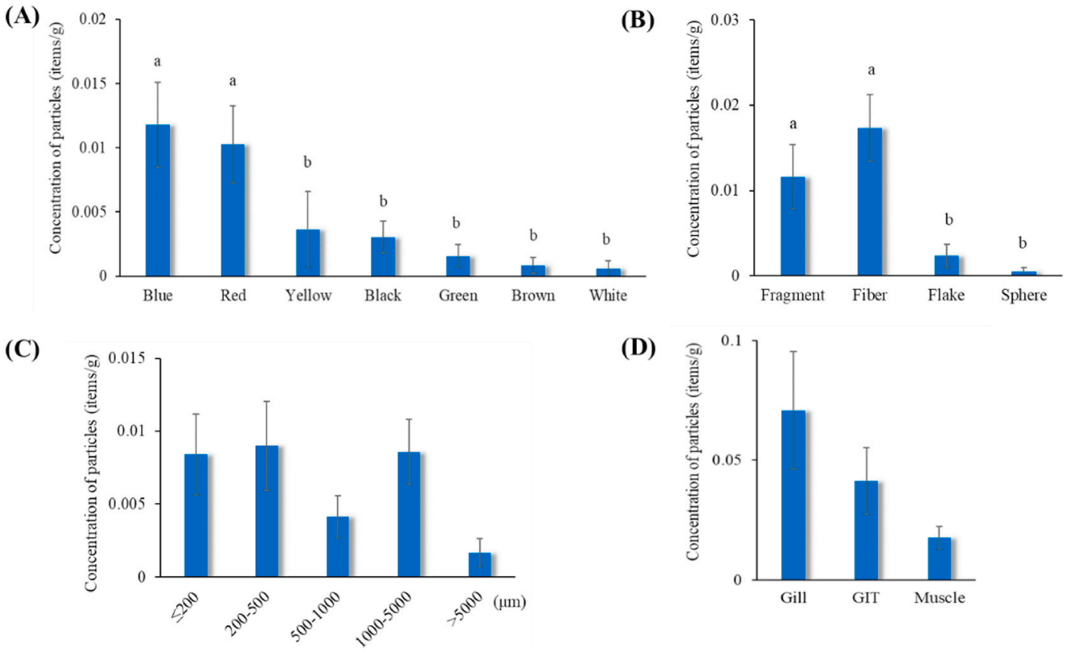


Fig. 6. The concentrations of different colors (A), shapes (B), size (C), and tissue distribution (D) of anthropogenic particles in fish samples (mean ± SEM). The superscript letters indicate significant differences between groups ($p < 0.05$).

4.2. Chemical characteristics of anthropogenic particles

4.2.1. The pigment particles

Pigments used in coatings, paints or to colorize plastic products usually contain metal components (e.g., arsenic yellow, cadmium red, chrome yellow, and white lead) [14]. Among all inorganic pigments, lead chromates are the most versatile due to their low cost and excellent lightfastness [31,34]. However, there is no safe level of exposure to lead for humans, especially for young children, which is the current consensus in the scientific community [35,36].

The results of this study show that the dominant pigment is indigo and the shape of indigo particles is mostly fiber. Indigo is most commonly associated with the production of blue jeans and denim cloth. Athey et al. (2020) found that one pair of used jeans could release 56000 ± 4100 microfibers per wash, which resulted in a lot of indigo-dyed fibers entering the environment [37]. In this study, most indigo particles are fiber in shape. Hence, it is inferred that the indigo-dyed fibers in fish samples may have originated from blue

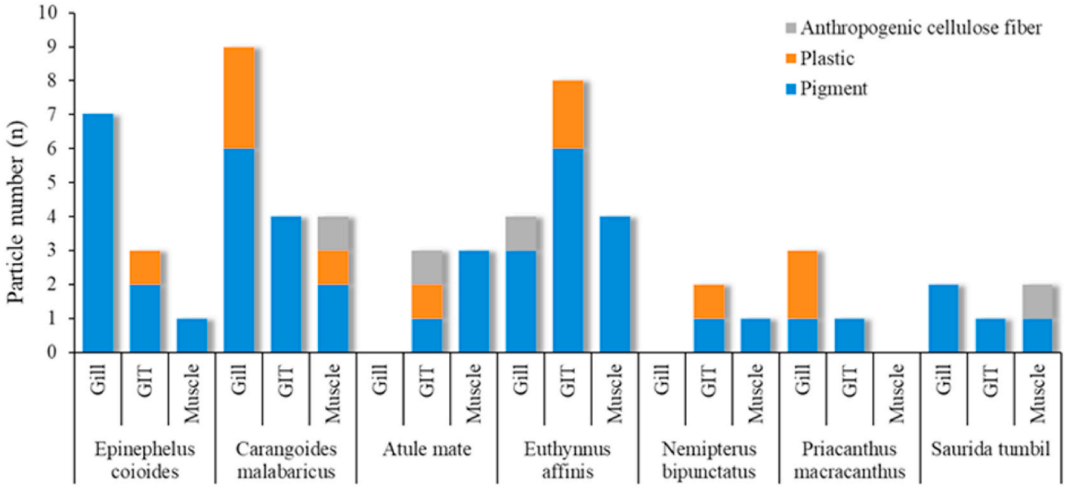


Fig. 7. The number of anthropogenic particles isolated from different tissues (gill, gastrointestinal tract, and muscle) of seven tested fish species.

jeans and denim cloth. However, the Raman spectrums of these particles correspond only to those of indigo and do not contain any Raman spectrum of textile fibers. It can be understood that pigments can interfere with strong Raman signals, which hinders the identification of plastic polymers and textile fibers. Therefore, it remains to be possible that these identified pigment particles are plastic or textile fibers covered with pigments [31]. Moreover, it is inferred that the true amounts of plastics and textile fibers isolated from fish samples are greater than the measured value.

4.2.2. The plastic particles and textile fibers

The abundance of plastic shown in this study was compared with other recent studies on fish species collected from Southeast Asia (Table 2). In this study, 84 individual marine fish were examined, and out of 62 identified APs, only 11 were plastic polymers resulting in a 0.13 items/individual plastic, which was lower than that in the other two Malaysian studies, which were 0.30 items/individual and 0.39 items/individual, respectively (Table 2). Meanwhile, according to these references in Table 2, the average plastic number in each fish collected from Malaysia is lower than the other three Southeast Asian countries including Indonesia, Philippines, and Thailand. However, due to the absence of sufficient published literature on the plastic abundance of fish in Southeast Asian countries, a more in-depth study should be made before any conclusive conclusion can be made. From Table 2, the plastic abundance in fish in four studies only using a microscope was reported to be higher than in other studies using rigorous chemical analyses such as μ-Raman and ATR-FTIR analyses. One of the possible reasons is that some non-plastic particles might be erroneously characterized as plastic when observed only using a microscope since various shapes and colors of plastic are available, and visually, many non-plastic particles such as sand, chitin fragments, and diatom shell fragments are similar to plastic particles [38]. Also, as the size of particles decreases, the possibility of misidentification using the microscope is increased [39]. Previous studies have shown that the occurrence of misidentification of plastic-like particles using a microscope is usually more than 20 % and sometimes even more than 70% [8,31,40]. Löder & Gerdts (2015) found that plastic particles made up only 1.4% of all plastic-like particles [38]. Therefore, after using a microscope to quickly and economically screen out plastic-like particles based on the characteristics of the particles, further analysis by utilizing spectroscopy, such as Raman spectroscopy and FT-IR spectroscopy, is strongly recommended.

In the present study, the plastic polymers identified by micro-Raman spectroscopy were PP, PET, and PAN, which are the common plastic types found in fish [21,31,41]. These plastic polymers have been widely used in many areas, such as fishing gear, toys, packaging, construction films, and the manufacture of electronic components. FAO estimates that 640,000 t of fishing gear are lost to the marine environment every year and their fragmentation will result in the release of large amounts of plastic particles [42]. Moreover, these plastic polymers as the components of synthetic fibers are widely used in the textile industry.

In addition to synthetic fibers, textile fibers also include anthropogenic cellulose fibers and natural fibers. In this study, 4 particles were confirmed as modal or cellulose acetate sorbate which is anthropogenic cellulose fibers. Although anthropogenic cellulose fibers are rarely reported, a few studies found that they are served as the dominant microfiber type, which is ubiquitous in the environment and has even been found to accumulate in high concentrations in Arctic marine sediment [20,22,43,44].

Although the major sources of fiber in the aquatic environment have not been comprehensively reviewed, wastewater from household washing is generally considered a significant point source of fiber in environments [45]. According to Napper & Thompson (2016), 6 kg of synthetic materials (polyester, polyester-cotton blend, and acrylic) could release 137,951–728,789 fibers per wash, resulting in high levels of MP fiber pollution in the environment [46].

In the past two decades, the average annual consumption of textiles has tripled, from 7 kg per capita to 13 kg, and has reached 100 million tons. Correspondingly, the demand for textile fibers in the textile industry continues to grow and is expected to increase to 130 million tons by 2025 [47]. which will likely lead to further aggravation of textile fiber pollution in the environment. Meanwhile, the COVID-19 pandemic drives explosive growth in the use of masks. It is estimated that more than 129 billion masks are consumed

Table 2
Summary of recent studies reporting the occurrence of plastics in fish in Southeast Asia.

Species name	Number of individuals examined	Number of isolated plastic-like particles	Number of plastics or percentage of plastics in all particles (%)	Identification	Average plastic number per individual	Dominant plastic polymer	Size range	Parts	References
Malaysia									
[a] **7 species of fish**	**84**	**81**	**11(13.58 %)**	**μ-Raman**	**0.13**	**PP and PET**	**0.5–30 mm**	**Muscle, gill, and GIT**	**This study**
Chelon subviridis *Johnius belangerii* *Rastrelliger kanagurta* *Stolephorus waitei*	120	61	59%	μ-Raman and FESEM-EDX	0.30	PP and PE	0.001–1 mm	Eviscerated flesh and excised organs (viscera and gills)	Karami et al., 2017
[b] 11 species of fish	110	56	76.8%	μ-Raman and FESEM-EDX	0.39	PE	0.2–34.9 mm	Viscera and gills	Karbalaei et al., 2019
Indonesia									
[c] 9 species of fish	174	–	2063	Microscope	11.86	–	<1 mm	Stomach and gut	Hastuti, Lumbanbatu, & Wardiatno, 2019
Decapterus macrosoma *Rastrelliger kanagurta* *Siganus canaliculatus* *Spratelloides gracilis*	76	–	105	Microscope	1.38	–	0.5–5 mm	Gastrointestinal tract	Rochman et al., 2015
Philippines									
Auxis rochei *Rastrelliger kanagurta* *Chanos chanos*	81	–	635	Hot needle	7.84	–	<4 mm	Gastrointestinal tracts	Abiñon et al., 2020
Siganus fuscescens	120	–	72	ATR-FTIR	0.6	PP	<2 mm	Guts	Bucol, 2020
Thailand									
[d] 24 species of fish	165	–	258	Microscope	1.56	–	0.13–17.16 mm	Stomach	Azad et al., 2018
[e] 8 species of fish	107	–	–	Microscope	1.76	–	0.03–3.84 mm	Stomach and intestine	Kasamesiri & Thaimuangphol, 2020

Notes.
-: Not reported.
ATR-FTIR: attenuated total reflectance Fourier transform infrared spectroscopy.
EDX:energy-dispersive X-ray spectroscopy.
FESEM:field emission scanning electron microscopy.
PE: Polyethyl.
PP: Polypropylene.

[a] 7 species of fish: *Epinephelus coioides, Carangoides malabaricus, Atule mate, Euthynnus affinis, Nemipterus bipunctatus, Priacanthus macracanthus, Saurida tumbil.*

[b] 11 species of fish: *Megalaspis cordyla, Epinephelus coioides, Rastrelliger kanagurta, Euthynnus affinis, Thunnus tonggol, Eleutheronema tridactylum, Clarias gariepinus, Colossoma macropomum, Nemipterus bipunctatus*, *Ctenopharyngodon Idella, Selar boops.*

[c] 9 species of fish: *Oreochromis mossambicus, Scatophagus argus, Siganus canaliculatus, Crenimugil seheli, Mugil cephalus, Chanos chanos, Anodontostoma chacunda, Sardinella fimbriata, Abalistes stellaris.*

[d] 24 species of fish: *Alepes apercna, Dasyatis zugei, Dendrophysa russellii, Leiognathus berbis, Leiognathus fasciatus, Leiognathus splendens, Alepes melanoptera, Alepes vari, Anodontostoma chacunda, Johnius borneensis, Johnius carouna, Opisthopterus tardoore, Rastrelliger brachysoma, Sardinella gibbose, Sardinella jussieu, Scomberomorus commerson, Scomberomorus guttatus, Alepes kleinii, Drepane longimana, Megalaspis cordyla, Sardinella albella, Scomberoides tala, Scomberoides tol, Terapon theraps.*

[e] 8 species of fish: *Labiobarbus siamensis*, *Puntioplites proctozyson*, *Cyclochelichthy repasson*, *Henicorhynchus siamensis*, *Labeo chrysophekadion*, *Mystus bocourti*, *Hemibagrus spilopterus*, *Laides longibarbis*.

globally every month [48]. Due to the lack of environmental awareness and inappropriate waste management, a large amount of mask waste is discarded without any treatment, which further increases the number of textile fibers released into the environment.

4.3. Intake of anthropogenic particles by humans consuming fish

The consumption of fish in Malaysia was among the world's highest [30]. Compared to other countries with low fish consumption, people in Malaysia are more vulnerable to fish-related food safety hazards, including APs, in terms of their health. However, due to the scarcity of relevant research, the safe limit of APs that can be ingested by humans through food is still ambiguous. Thus, it remains a necessity to determine the concentration of APs in the edible tissues of fish and understand the potential risks to human health. The estimate made in the present study indicated that the intake of APs through fish consumption by people in Malaysia was 1062 AP items/year/capita. And a previous study by Barboza et al. (2020) estimated the intake of MPs through fish consumption by people in Portugal, Spain, Italy, United States, and Brazil, with results of 3078 MP items/year/capita, 2576 MP items/year/capita, 1679 MP items/year/capita, 1156 MP items/year/capita, and 518 MP items/year/capita, respectively [49].

In addition to fish, plenty of food resources are validated to contain APs (e.g., beer, salt, honey, sugar, and oyster) [50–52]. Due to the wide distribution of APs in food, it seems unlikely to avoid the ingestion of APs, even for newborn babies fed only on breast milk [53,54]. To maximumly prevent the potential risks of APs to humans, it is encouraged to conduct more researches concerning human exposure to APs through ingestion of food and the toxicity of APs to humans. Based on the endeavors, the safety limits of APs in food for humans might be determined and relevant food safety regulations could be established to protect consumer health.

In addition to ingestion, inhalation is also a major exposure route for APs. Humans can inhale APs suspended in the air caused by daily human activities, such as the wear of vehicle tires, microfiber detachment from clothes, or even opening a plastic package [55–57]. It is estimated that each person inhales 2.16×10^3 MPs per year [58]. Moreover, during the COVID-19 pandemic, the prolonged use of masks may further increase the amounts of APs inhaled by humans [58]. Meanwhile, dermal contact is another route of AP exposure. Consumer products such as face creams, cleansers, toothpastes and cosmetics that people use on a daily basis may contain APs [59]. Some studies suspected that nanoscale particles may penetrate human skin [60]. Even so, there are no studies that quantify dermal exposure to APs and their potential effects.

5. Conclusion

In this work, a thorough study has been carried out on the presence of APs in gill, GIT, and muscle of seven commonly consumed marine fish species from Kota Kinabalu wet market, Malaysia. The results show that the dominant color and shapes of APs isolated from fish samples are blue and fiber, respectively, and the size is mostly smaller than 0.5 mm. By employing the micro-Raman spectrometer, the type of APs could be classified into plastic, pigment, and anthropogenic cellulose fibers. In addition, the muscle of fish is detected to contain some APs, indicating that removing the gill and GIT of fish could reduce the risk of ingesting APs for consumers but cannot eliminate it. And the estimated human intake of APs through the consumption of fish muscle in Malaysia was 1062 AP items/year/capita. Thus, it is highly recommended to perform future investigations of APs pollution in the edible tissue of fish in order to evaluate potential APs pollution in human food. Concerning the current load of APs in fish and the growing consumption of fish, it is suggested to add APs testing into the guidelines of food safety management systems and adopt mitigation measures to reduce the APs pollution in the aquatic environment.

This study would update the data on the distribution of APs in the marine ecosystem and contributes to a better understanding of the potential human exposure to APs. To fully estimate the implications for the marine food web and human health, more *in vitro* studies are needed in the future since they could help to find the underlying reason for APs translocation among different fish tissues and evaluate possible effects on the organism.

Ethics statement

All experiments were performed in accordance with relevant local guidelines and regulations. The slaughtering of marine fishes was conducted according to established animal welfare guidelines.

Data availability statement

Data included in article/supp. material/referenced in article.

CRediT authorship contribution statement

Peiru Gao: Conceptualization, Investigation, Methodology, Validation, Writing – original draft. **Nor Qhairul Izzreen Mohd Noor:** Funding acquisition, Resources, Supervision, Writing – review & editing. **Umi Hartina Mohamad Razali:** Writing – review & editing. **Mohd Hazim Mohd Yusop:** Writing – review & editing. **Sharifudin Md Shaarani:** Formal analysis, Project administration, Writing – review & editing.

Declaration of competing interest

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 paper.

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Appendix A. Supplementary data

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