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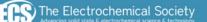
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### Microplastics evaluation in edible tissues of flying fish (Parexocoetus mento) from the Bintaro fish market, Lombok, Indonesia

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Abstract. Microplastics (MPs) are plastic fragments that are degraded into small particles with a diameter of <5 mm. Pollution in the ocean by MPs has become a global threat which damages aquatic and marine ecosystems. There are several types of MPs detected in the digestive tract and in edible fish tissue. However, such report remains limited in Indonesia. Despite the high rate of plastic pollution in Indonesian coastal areas. This study aims to determine the presence, abundance and characteristics of MPs including type, shape, and color in the edible tissue of commercially important pelagic fish species from the Bintaro fish market, Lombok, West Nusa Tenggara. Flying Fish (Parexocoetus mento) was evaluated for MPs content in their edible tissue. Isolation of MPs in fish meat was conducted by firstly adding 10% KOH solution followed by incubation for 24 h. Furthermore, the solution was incubated in the oven for 48 h at a temperature of 90°C. The WPO (Wet Peroxide Oxidation) reaction was carried out by adding 30 mL of 0.05 M Fe(II) oxide and 30 mL of 20% Hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) continued by heating on a hotplate at 75°C at 350 rpm for 45 minutes to remove organic matter from fish. The remaining filtrateswere treated by further heating, followed by filtration with stainless steel sieves (45  $\mu$ m). Current results show five types of MPs detected in fish sample were Fragment, Filament/fiber, Film, Foam and Pellet. The most abundant type of MPs was fragment type (368.67 particle/fish). Current results show a significant amount of MPs were found in edible tissue of flying fish in Bintaro market. However further characterization of MPs would be needed to confirm the MPs types. Nevertheless, current study show the potential contamination of MPs in P. mento fish commercially sold in fish market at Lombok, Indonesia. Hence, awareness should be raised in local communities to reduce plastic pollution in marine and coastal areas.

#### 1. Introduction

Marine debris such as plastic waste in the ocean will be further fragmented into smaller particles due to UV exposure, climate change, physical abrasion such as tidal waves, tides, wind so that it is fragmented into smaller sizes. The plastic fragments will be degraded until the molecular weight of the plastic is reduced and the plastic will become brittle and turn into smaller particles with a size less than 5 mm, this kind of plastic we would call microplastics (MPs) [1]. MPs are divided into two groups based on the process of formation, namely primary and secondary MPs. Primary MPs are MPs that are intentionally shaped by the industry with the size of a micro-sized microbead from the beginning of its manufacture [2]. While, secondary MPs are formed from large plastics that experience fragmentation during use or size reduction due to degradation [3]. These pieces of plastic can come from fish nets, industrial raw materials, household appliances, plastic bags, synthetic fibers for washing clothes or weathering plastic products [4].



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reduction due to degradation [3]. These pieces of plastic can come from fish nets, industrial raw materials, household appliances, plastic bags, synthetic fibers for washing clothes or weathering plastic products [4].

Water that have been contaminated with MPs can threaten ecosystem and interfere food chain of marine biota such as marine mammals, fish, and seabirds which will accumulate in large gyres in the middle of the ocean because MPs like transporters, have a tendency to bind other materials such as waste, heavy metals, detergents, pesticides and toxins, which contain some of the chemicals produced and which can get into the tissues of marine organisms, including species that consumed by humans [5].

Disposal of this plastic waste will disrupt the ecosystem of the waters. Research conducted by Jambeck et al. (2015) [6], they stated that Indonesia was in second place with the largest amount of marine plastic waste in the world. Based on the accumulated population in Indonesia, the pile of plastic waste that ends up in the waters reaches 0.48-1.29 million tons/year. It is estimated that 60-80% of the waste in the sea comes from plastic waste [5].

Based on statistical data released by the Central Statistics Agency (2017) [7], fishery production in Indonesia from 2011 to 2016 showed an increase every year with a total production of 23.26 million tons. In particular, fish production in Mataram city reached 1,797 tons in 2015. However, the increase in MPs from the greater fragmented of plastic debris in marine ecosystems has led to uptake by various marine organisms [8]. The ecological risk of plastic particles absorbed by fish will cause a decrease in the fitness level of the organism due to physical blockage, inflammation and decreased fecundity that affects fish populations [9].

The amount of plastic waste in Indonesia's oceans will threaten the marine life in it. Data on the presence of MPs in pelagic fish from Indonesian waters is still very limited, despite the level of plastic pollution in Indonesia is high. Therefore, it is very important to conduct research on the content and characteristics of MPs in flying fish (*Parexocoetus mento*) as an important commercial food in the wider community at the Bintaro Fish Market, Lombok, West Nusa Tenggara.

#### 2. Material and Methods

#### 2.1 Sample collection and preparation

The samples taken were of important commercial fish species that are often consumed in the region. Flying fish (*Parexocoetus mento*) used in this study were collected at Bintaro Fish Market, Lombok (8°33'30.9"S 116°04'30.1"E). The type of fish in this study was carried out 3 replications. *P. mento* was cleaned from impurities and washed under running water. The fish samples obtained were then identified and recorded thecharacteristic morphometric and meristic of the fish. The references used for the identification of fish species in this study refer to the books of William T. White et al (2013) [10] and the Food and Agriculture Organization of the United Nations (FAO, 1997) [11].

#### 2.2 Isolation of MPs

The Isolation of MPs in fish meat method were processed using a modified National Oceanic and Atmospheric Administration (NOAA, 2015) [12]. MPs isolation was carried out with 50 gr of fish that had been filleted and by adding 10% KOH solution followed by incubation for 24 hours. The WPO (Wet Peroxide Oxidation) was carried out by adding 30 mL of 0.05 M Fe(II) oxide and 30 mL of 20% Hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) continued by heating on a hotplate at 75°C at 350 rpm for 45 minutes to remove organic matter from fish. The remaining filtrates were treated by further heating, followed by filtration with stainless steel sieves (45  $\mu$ m). Observe with a brightfield microscope (Nikon Eclipse, China).

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#### 2.4 Statistical analysis

Analyze the data using imageJ software program. The results were than statiscally analyzed using one-way ANOVA and Tukey's with a 95% of confidence level. A significant value of <0.05 was considered statistically different. Statistical analysis was performed using GraphPad Prism version 9.2.0 (GraphPad software, Inc).

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#### 3. Results and Discussion

The sampling location in this study was carried out at the Bintaro Fish Market, Ampenan, Lombok (Fig. 1). Ampenan is one of three districts in the city of Mataram and is located in a coastal area with most of the general profession as fishermen. The location selection was carried out because this area is one of the main suppliers of marine fish in Lombok, which will be further distributed throughout smaller markets.

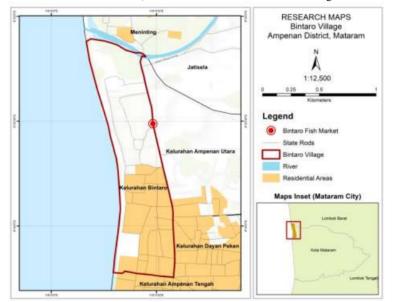


Figure 1. Sampling Site at the Bintaro Fish Market, Ampenan, Lombok.

The classification for flying fish spesies from phylum chordata, family exocoetidae genus parexocoetus and spesies *P. mento*. In this study, morphometric and meristic measurements of flying fish were carried out.

<b>Table 1.</b> Morphometric of Parexocoetus mento.				
Species (Weight = gram)*	Code	Measures	Range (cm)	Mean ± SD
Parexocoetus mento	TL	Total Length	15.50-17.00	$16.17\pm0.76$
$(29.90 \pm 1.35)$	SL	Standard Length	12.50-13.50	$13.00\pm0.50$
	HL	Head Length	4.00-4.50	$4.33 \pm 0.29$
	BL	Body Length	8.50-9.00	$8.67 \pm 0.29$
	CL	Caudal Length	3.00-3.50	$3.17 \pm 0.29$
	ED	Eye Diameter	1.00-1.52	$1.20\pm0.28$

\*Value represent Mean ± SEM of 3 replications.

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Table 2. Meristic of Parexocoetus mento.							
Species	Mouth Type	Body Shape	Scale Type	Number of Predorsal	Number of Transverse Scales	Number of Linea Lateral Scales	Fish Fin Formula
Parexocoetus mento	Terminal	Fusiform	Cycloid	22-31	22-29	39-56	D 8-10

\*D = Dorsal fin

#### Table 3. The abundance of MPs in P. mento, majority found the most was fragments

Types of Microplastic	Abundance (Particle/fish)	Percentage (%)
Fiber	62.67	0.02
Film	263.67	0.08
Foam	219.33	0.07
Fragment	368.67	0.11
Pellet	173.00	0.05

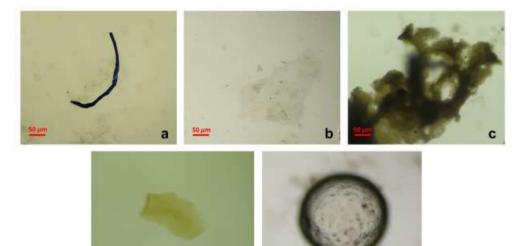


Figure 2. Identification of five MPs particles found in *P. mento* tissue with a magnification of 10x (a) Fiber/filament, (b) Film, (c) Foam, (d) Fragment, and (e) Pellet

e

d

The measured characteristic morphometric on *P. mento* is done by measuring TL (Total length), SL (Standard Length), HL (Head Length), BL (Body Length), CL (Caudal Length) and ED (Eye Diameter. The results obtained how the total length on *P. mento* averaged 16.17 cm with a weight of 29.90 gr. Based on research conducted by Mishra et al. (2010) [13, 14]. The morphometric data of *P. mento* showed results that were not significantly different from those obtained in this study (Table 1). Table 2. shows the meristic characteristic *P. mento* has a fusiform body type with a very slender (stream-line), propulsion which is very helpful in fish movement [15]. Cycloid scales are shaped like circles, the edges of the scales

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are smooth and flat, generally found in fish with weak fin rays (Malacopterygii) [16]. The character of meristic of *P. mento* has the formula of dorsal rays (D 8-10). This means that from some samples of flying fish there are those that have a dorsal weak fin rays of 8-10 and *P.mento* does not have hard fin rays [17, 14]. Morphometric measurements were carried out to determine the identification of morphological variations based on morphometric and meristic characters in *P. mento* [18].

We present the first study to evaluate MPs pollution in flying fish in the fish market in Lombok Indonesia as an important commercial fish species frequently consumed in the region. Flying fish is an important commercial commodity in certain areas in the country, such as South Sulawesi, Maluku, and North Sulawesi and is one type of marine resource that has economic value [19]. Such special structures in certain areas in the higher MPs contamination of the organism. In addition, some fish species such as northern and ovies show increased aggregation into plastic waste, which increases their consumption of MPs [20]. Such species specific structural or behavioral differences considered as the reason for highest MPs contamination found in *P. mento* in this study. In contrast, visual predators attack a single individual prey they select visually from the water column. They are still susceptible to ingesting MPs attached to food or misidentified as prey [21] and secondary consumption through prey items [22].

There are 5 types of MPs found in flying fish, including fiber/filament, film, foam, fragment and pellet (Fig 2) [23]. The shape of microplastics is one of the factors that affect the potential to be eaten by fish. for example, round shapes (pellets or microbeads) are more likely to be avoided than irregular, colored shapes like Fragments [24]. This supports the results of the study by finding more types of fragments in fish than other types of MPs. Studies by MPs accumulation was found to be abundant in sieve size 45  $\mu$ m [25]. The fragment type was the most abundant and significant MPs of the four MPs types obtained in the edible tissues of *P. mento*. As could be seen in table 3. we can conclude that the most abundant MPs types found in flying fiel dominate by fragment type. The data shows a total of 368.67 particles/fish with a percentage of 0.11%. Fragments are reported to be a major component in floating MPs in the coastal waters of the southwest coast of India [26, 27]. The average quantity of MPs in edible tissue was 13.04  $\pm$  0.82 items/gr. this is comparable to the edible tissue **1** the pelagic dried fish species reported by Karami et al. (2017) [28]. However, the results of the study by Su et al. (2019) [29] and Akoueson et al. (2020) [30] differed, where significant MPs contamination was only found in inedible tissues (intestines and gills) and not in edible fish tissues. This contrasts with the findings of Karami et al. (2017) [28] where MPs in the removed flesh (muscle and skin) were higher (approximately four times) compared to the excised organs (offal and gills) in dried fish.

#### 4. Conclusion

In conclusion, a significant amount of MPs were found in edible tissue of flying fish in Bintaro market. The highest amount of MPs found in pelagic at Bintaro fish market were fragments type. However chemical characterization of MPs would be needed for a better confirm. Further study of MPs in different fish species would be interesting to provide information regarding MPs contamination in West Nusa Tenggara or other regions in Indonesia.

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