TẠP CHÍ KHOA HỌC
TRƯỜNG ĐẠI HỌC QUY NHƠN

Tập tính ăn và lượng vi nhựa ăn vào của một số loài cá bống phân bố ở đầm Thị Nại, tỉnh Bình Định

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TÓM TẮT

Nghiên cứu này nhằm đánh giá tập tính ăn và lượng vi nhựa ăn vào của 3 loài cá bống phân bố ở đầm Thị Nại, tỉnh Bình Định. Mỗi loài được thu 30 cá thể từ ngư dân để nghiên cứu. Trong đó, 20 cá thể của mỗi loài được giải phẫu để xác định thức ăn tự nhiên và 10 cá thể còn lại được tách lấy ống tiêu hóa và phân tích sự tích tụ vi nhựa. Kết quả cho thấy, mặc dù tính ăn của 3 loài cá là khác nhau nhưng tần suất xuất hiện của mùn bã hữu cơ trong dạ dày của các loài đều cao hơn so với các loại thức ăn khác. Lượng vi nhựa ăn vào không khác nhau có ý nghĩa thống kê giữa 3 loài, dao động từ 6,50 đến 9,20 vi nhựa/cá thể; từ 0,58 đến 1,16 vi nhựa/g khối lượng cơ thể; và từ 31,47 đến 57,55 vi nhựa/g khối lượng ống tiêu hóa. Cả 3 loài cá đều ăn vào phần lớn là vi nhựa dạng sợi (75,38 - 82,50%) có kích thước từ 500-2000 µm (59,09 – 65,75%). Từ những kết quả thu được có thể nhận định rằng, tập tính ăn khác nhau của 3 loài cá bống trong nghiên cứu này không ảnh hưởng đến lượng vi nhựa ăn vào và vi nhựa trong mùn bã hữu cơ là nguồn vi nhựa tích lũy chính trong hệ tiêu hóa của 3 loài này. Cần có nhiều nghiên cứu thêm để xác định rõ hơn các tác động sinh học và sinh thái của các vi nhựa đối với cá.

Từ khóa: *Vi nhựa, đầm Thị Nại, loài cá bống, tập tính ăn, tích tụ vi nhựa.*

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Feeding habits and quantity of ingested microplastics of some goby species distributed in Thi Nai lagoon, Binh Dinh province

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ABSTRACT

This study aimed to evaluate feeding habits and microplastic ingestion of three goby species distributed in Thi Nai lagoon, Binh Dinh province. Thirty individuals of each species from fishermen for study. Twenty were dissected to identify natural food and the other ten were used to determine microplastic accumulation in digestive system. The results showed that feeding habits of these three fish species were different, but the occurrence frequency of organic matter in the digestive system of fish was higher than that of other natural food. The concentration of ingested microplastics was from 6.50 to 9.20 particles/fish, from 0.58 to 1.16 particles/g wet body weight and from 31.47 to 57.55 particles/g digestive tract weight and did not significantly differ among these fish species. These three fish species mostly ingested microplastic fibers (75.38 – 82.50%) with most size of 500-2000 µm (59.09 – 65.75%). Our findings indicate that the different feeding habits of the three goby species in this study do not affect the amount of ingested microplastics and the microplastics in organic matter are the main source of microplastics accumulated in the digestive system of these three species. Three studies are needed to better characterize the biological and ecological impacts of microplastics on fish.

Keywords: *Microplastic, Thi Nai lagoon, goby species, feeding habits, microplastic accumulation.*

1. INTRODUCTION

Plastic pollution has been paid attention to worldwide. Human awareness and concerns about microplastic pollution are increasing, especially microplastic pollution in aquatic environment. Microplastic accumulation in aquatic organisms has been reported by many researchers around the world for a long time as well as recently by scientists in Vietnam. Microplastics has been found in many organisms at different nutritional levels such as zooplankton,[1] invertebrates,[1,3] fish.[4] There are many studies conducted and found microplastics in aquatic animals such as in clam *Mytilus galloprovincialis* inhabiting in estuaries of Tagus, Portugal and Po river, Italy,[5] crab *Neohelice granulata* in estuaries in Southwest Atlantic Ocean,[6] or in 24 fish species in estuaries in Brazil,[7] oyster *Crassostrea virginica* and crab *Panopeus herbstii* in estuaries in Florida.[8] In Vietnam, some researchers have recorded microplastic contamination in molluscs such as *Anadara granosa*, *Anadara subcrenata* in Thi Nai lagoon, Binh Dinh province,[9,10] *Mactra grandis*, *Callista lilacina*,

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Marcia hiantina in Cu Mong lagoon, Phu Yen province,[11,12] small fish species in coastal areas of Binh Dinh province[13] or *Meretrix lyrata* in Ho Chi Minh city.[14] In general, these authors mostly focused on determining microplastic contamination in studied animals, but did not assess relationships between feeding habits and microplastics ingestion of these animals. Therefore, understanding of such relationships is still limited.

A large amount of plastics and microplastics discharged from human activities in both lands and seas are accumulated in coastal areas in the world.[14] In the same situation, coastal areas or lagoons (including Thi Nai) in Vietnam are also accumulated microplastics with different concentrations.[9,15]

Thi Nai in Binh Dinh province is the second largest lagoon in Vietnam after Tam Giang – Cau Hai, Thua Thien Hue province. Thi Nai has a diversity of ecosystems such as mangroves, seagrass, etc., where many organisms including fish inhabit, feed and reproduce. Fish species composition in this lagoon recently increase, but they diverse relatively, with 95 species including 7 goby species (Vo and Nguyen).[16] Among goby species in Thi Nai lagoon, tropical sand goby (*Acentrogobius caninus*), maned goby (*Oxyurichthys microlepis*) and redspot arrowfin goby (*Oxyurichthys tentacularis*) are 3 species that commonly appear and are collected to supply local consumers. This study focused on assessing feeding habits and microplastic ingestion, and examined microplastic characteristics accumulated in the digestive tracts of these 3 goby species to contribute to microplastic source identification.

2. METHODS

2.1. Fish collection and treatment

Tropical sand goby (*Acentrogobius caninus*), maned goby (*Oxyurichthys microlepis*) and redspot arrowfin goby (*Oxyurichthys tentacularis*) were collected from fishermen in Thi Nai lagoon, with 20 individuals of each species to be examined for feeding habits and 10 for microplastic accumulation.

Fish were frozen at the collection sites and brought to lab. In the laboratory, fish used for microplastic examination (10 individuals/ species) were treated within the day or frozen in the fridge for later analysis. Fish for examination of feeding habits (20 individuals/species) were treated within the day or fixed in 5% formol for later analysis.

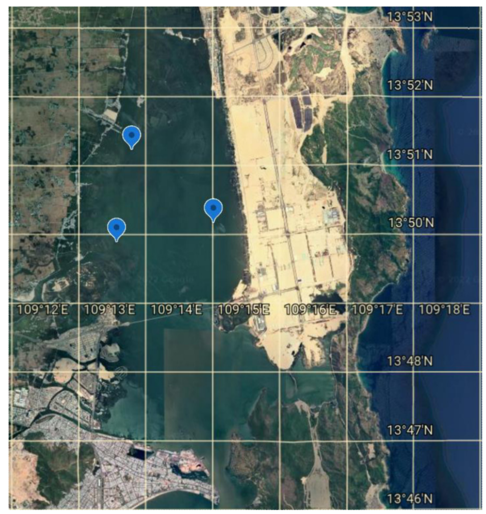

Figure 1. Sample collection sites in Thi Nai lagoon

2.2. Feeding habit examination

Fish individuals were dissected to examine natural food in their digestive tracts. Stereomicroscope Meiji EMTR-3 and binocular microscope Olympus CX21 were used to analyse and identify the food. Based on the analysis results, occurence frequency of food items (Oi%) in the digestive tracts of fish was calculated according to the method of Hyslop.[18]

Oi% = (Number of digestive tracts containing food item i/Total number of digestive tracts containing food) x 100.

2.3. Microplastic contamination examination

Each fish individual was weighed, measured and washed using water filtered through the glass fiber filters GF/A with pore sizes of 1.6 µm, then dissected to remove the digestive tract. Next, the digestive tract was put into a glass beaker containing 20 ml 10% KOH, covered by aluminum foil and placed in an incubator at 60 °C for 24 hours according to the method of Alexandre et al.[18] The samples after incubated were filtered through 1 mm mesh sieve to collect microplastic particles 1- 5 mm on the sieve (if any). The samples under the sieve were then filtered through a 250 µm mesh sieve, and the material on the sieve was rinsed into a beaker to perform the overflow technique using a saturated NaCl solution with 3 replicates. Finally, the sample solution was filtered through a 1.6 µm GF/A filters according to the method of Emilie et al.[15] to remove microplastics. The filters were stored in clean Petri dishes and put in room condition for later stereoscopic observation.

Put the filters under the Leica S9i stereomicroscope to observe and identify microplastics using LASX software. Microplastics were identified with 3 shapes as described by Emilie et al.[15] such ase fibers, fragments and pellets. Microplastics on the filters were measured and photographed.

Microplastic concentration was considered as number of microplastics per individual, number of microplastics per gram of wet body weight and number of microplastics per gram of digestive tract weight of fish.

To control microplastic contamination from the surrounding environment during sample treatment and observation, we followed to suggestions of GESAMP[19] such as cleaning the working area with alcohol before observating and treating samples, wearing cotton clothes and rubber gloves, etc. In addition, at each period of sample observation and treatment, we placed a new filter in a Petri dish nearby the place we were working to check microplastic contamination. During the steps on sample, we did not find any microplastics on the control filter papers.

2.4. Data analysis

Microsoft Excel 2013 was used to calculate the necessary parameters and make diagrams. Anova single factor in Microsoft Excel 2013 was applied to compare some parameters between studied fish species.

3. RESULTS AND DISCUSSION

3.1. Results

3.1.1. Feeding habits of fish

Feeding habits of tropical sand goby, maned goby and redspot arrowfin goby was different (Table 1). The food spectrum of tropical sand goby was most diverse, with 6 groups of prey found in the digestive tracts of fish, of which occurrence frequency of organic matters, fish, seaweeds and shrimps was higher than that of others. Natural food of maned goby only comprised two groups of prey which are organic matters and fish, with the dominance of organic matters (92.86% in occurrence frequency), and natural food of redspot arrowfin goby included 3 groups which are organic matters, fish and seaweeds with the predominance of organic matters. Thus, it can be seen that organic matters are dominant in the natural food spectrum of 3 studied goby species.

Table 1. Natural food of three goby species

Kinds of food	Occurrence frequency of food (%)		
	Maned goby (n=20)	Redspot arrowfin goby (n=20)	Tropical sand goby (n=20)
Organic matters	92.86	45.00	40.00
Fish	7.14	35.00	26.67
Seaweeds	0.00	20.00	26.67
Snails	0.00	0.00	6.67
Shrimps	0.00	0.00	20.00
Zooplanktons	0.00	0.00	6.67

3.1.2. Microplastic accumulation in fish

Microplastic concentration in 3 goby species is expressed as number of microplastics per individual, number of microplastics per gram of

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wet body weight and number of microplastics per gram of digestive tract weight of fish (Table 2). The results showed that the concentration of micropastic ingested by 3 goby species varied from 6.50 to 9.20 microplastics/individual, 0.58 to 1.16 microplastics/g wet body weight and 31.47 to 57.55 microplastics/g weight of digestive tract. There was not significant difference of microplastic concentration between three goby species (p > 0,05).

Table 2. Concentration of ingested microplastic in three goby species

Fish species	Number of microplastics/ individual	Number of microplastics/g wet body weight	Number of microplastics/g digestive tract weight
Maned goby (n=10)	9.20 ± 3.44[a]	0.87 ± 0.38[a]	57.55 ± 29.45[a]
Redspot arrowfin goby (n=10)	6.50 ± 3.12[a]	1.16 ± 0.58[a]	46.76 ± 27.77[a]
Tropical sand goby (n=10)	8.00 ± 4.06[a]	0.58 ± 0.29[a]	31.47 ± 22.50[a]

Note: The different letters indicate significant differences for each parameter (p < 0.05).

The weight of tropical sand goby was highest (13.79 g/fish), followed by maned goby (10.81 g/fish) and redspot arrowfin goby (5.70 g/fish) (p < 0,05) (Table 3). However, the amount of ingested microplastics per gram of wet body weight was not significantly different between three studied species (Table 2). Similarly, the weight of digestive tracts of tropical sand goby was different from others, but the amount of ingested microplastics per gram of weight of digestive tracts was not significantly different between three goby species. Therefore, it can be said that fish size and weight of digestive tract of fish are not the main factors governing amount of microplastics ingested by fish. This is clearly indicated through the correlation between the number of microplastics ingested and wet body weight or digestive tract weight of fish (Table 3). Specifically, the correlation between the number of microplastics and wet body weight of fish was negative for maned goby and redspot arrowfin goby with the coefficient from -0.49 to -0.24 while there was no correlation between these two parameters for tropical sand goby (r = 0.08); the negative correlation was recorded between number of microplastics and weight of digestive tract of fish (coefficient from -0.40 to -0.27).

Table 3. The correlation between wet body weight, digestive tract weight of fish and amount of microplastics ingested in three goby species

Fish species	Wet body weight of fish (g)	Digestive tract weight of fish (g)	Correlation between number of microplastics and wet body weight of fish (r)	Correlation between number of microplastics and digestive tract weight of fish (r)
Maned goby (n=10)	10.81 ± 1.59[b]	0.18 ± 0.07[b]	-0.49	-0.27
Redspot arrowfin goby (n=10)	5.70 ± 0.74[c]	0.15 ± 0.04[b]	-0.24	-0.40
Tropical sand goby (n=10)	13.79 ± 1.86[a]	0.28 ± 0.11[a]	0.08	-0.33

Note: The different letters indicate significant differences for each parameter (p < 0.05).

The shape data of microplastics were pooled for 10 fish of each species. The results showed that there were two shapes of microplastics found in the digestive tracts of three studied fish species, that were fibers and fragments, of which fibers were dominant in all goby species (Figure 2). Specifically, microplastic fibers accounted for from 79.03% to 82.50% while the ratio of microplastic fragments was from 17.50% to 20.97%.

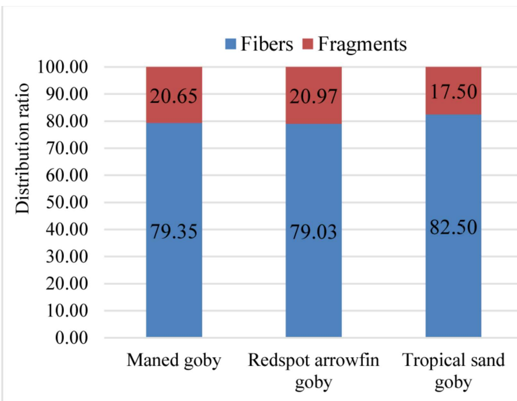

Figure 2. Shapes of microplastics found in three goby species

The fibers found in the digestive tracts of three studied fish species mostly had the length of 300 to 2500 µm, especially the size range of 500 to 2000µm, excepting a high ratio of fibers with the length of 2500-3000 µm (13,64%) observed in tropical sand goby (Figure 3). Specifically, fibers with the length of 500-2000 µm accounted for 65.75% in maned goby, 63.27% in redspot arrowfin goby and 59.09% in tropical sand goby while other fibers only accounted for 34.25%, 36.73% and 40.49% in these three goby species, respectively (Figure 4). Fragments observed in the digestive tracts of three goby species had the area of 45000 to 600000 µm^2 but dominant in range of 45000 - 200000 µm^2, with 77.78%, 69.23% and 71.43% in total in maned goby, redspot arrowfin goby and tropical sand goby, respectively (Figure 5). Thus, it can be seen that the studied fish species ingested microplastics in small size rather than big size range.

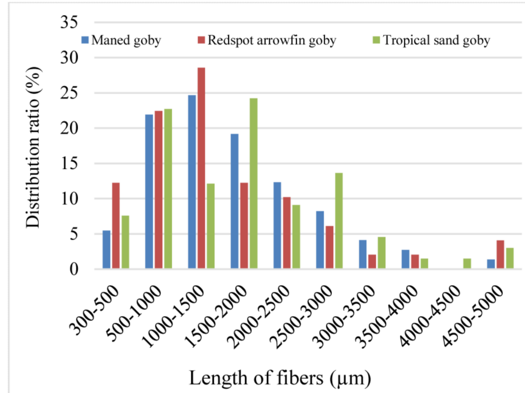

Figure 3. The length distribution of microplastic fibers

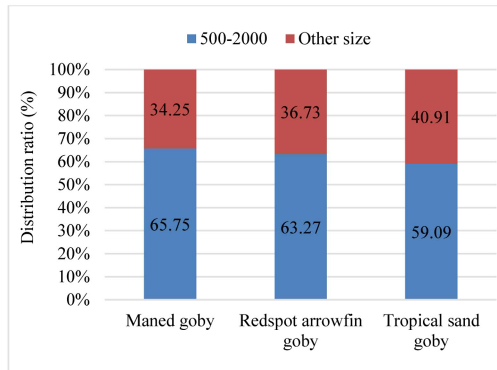

Figure 4. Percentage of microplastic fibers with length of 500-2000µm compared to others

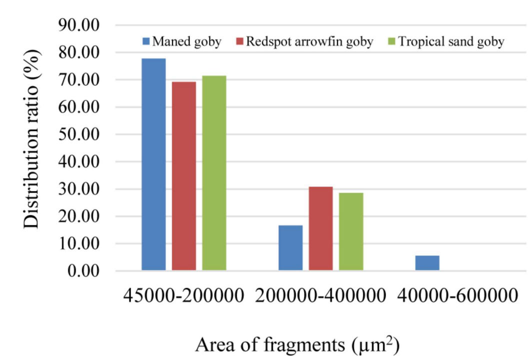

Figure 5. The area distribution of microplastic fragments

3.2. DISCUSSION

3.2.1. Feeding habits and microplastic ingestion of fish

Although the average microplastic concentration in each fish species is different, perhaps this difference is not large enough to lead to significant difference. Natural food spectrum of three studied goby species is different, but organic matters are the dominant food of these species. Hence, it can be speculated that the main source of microplastics accumulated in the digestive tract of these goby species is from organic matters. It can be seen that occurrence frequency of organic matters in the digestive tracts of maned goby is highest (92,86%) and microplastic concentration in this species is high (9.20 microplastics/individual) while occurrence frequency of organic matters in the digestive tracts of tropical sand goby is just 40% but weight of digestive tract of this species 1.5 times significant higher than that maned goby, so the amount of organic matters ingested by tropical sand goby may be nearly similar to maned goby and results in a relatively high microplastic intake in tropical sand goby (8.0 microplastics/individual). The amount of microplastics accumulated in redspot arowfin goby is low (6.5 microplastics/individual) because occurrence frequency of organic matters in this species is just 45% while its digestive tract weight is not different from maned goby, that means the amount of organic matters ingested by redspot arowfin goby is less than maned goby. In addition to main source of microplastics from organic matters, fish can also indirectly ingest microplastics accumulated in their other foods such as fish, shrimps, snails, seaweeds. Walkinshaw et al.[21] suggested that fish species having a narrow food spectrum would be more likely to directly ingest microplastics from environment than indirectly through prey such as in fish species with large food spectrum. This is somewhat reasonable with our results when comparing the food spectrum of maned goby and other 2 species in consideration of the corresponding amount of microplastic ingested. However, redspot arrowfin goby also has a natural food spectrum narrower than that of tropical sand goby, but the concentration of microplastics ingested by this species is lower than that of maned goby. This may be due to the dominance of organic matters in the diet of fish as mentioned above.

3.2.2. Characteristics of microplastics accumulated in the digestive tracts of fish

Concentration of microplastics accumulated in fish species in our study varied from 6.50 to 9.20 microplastics/individual, 0.58 to 1.16 microplastics/g wet body weight and 31.47 to 57.55 microplastics/g digestive tract weight, that is much higher than records of Gopal et al.[22] in 10 marine fish species in Bengal bay - Bangladesh (1.0-3.8 microplastics/individual, 0.02-0.15 microplastics/g wet body weight and 0.63-6.45 microplastics/g digestive tract weight) or the study results of Vendel et al.[23] in 69 fish species in estuaries in Northeast Brazil (vary 1 to 4 and 1.06 microplastics/individual in average), Wanlada and Suwaree[24] in 7 fish species in coastal areas of Thailand Gulf and Adaman sea (0 – 0.4 microplastics/individual), Sukree et al.[25] in *Rastrelliger brachysoma* in Thailand Gulf (2.70 ± 16.62 microplastics/individual). However, microplastic concentration in our study is lower than that reported by Ayu et al.[26] in 9 fish species in coastal areas of Pantai Indah Kapuk, Indonesia (varies from 4.9 to 20 and average of 12.21 microplastics/individual). Therefore, microplastic accumulation level in fish species mentioned in different studies is not similar and that can be due to different characteristics of these fish species but may be also governed by environment at study locations.

The dominance of fibers compared to fragments is also observed in our study. The similar results are reported by Gopal et al.[22] (fibers accounted for 53.4%), Vendel et al.[23] (fibers accounted for 90%), Ayu et al.[26] (89.63% of fibers in total), or Wanlada and Suwaree[24] (fibers accounted for 57.14% in pelagic fish and 82.76% in benthic fish). Likewise, such tendency is also found in molluscs.[26,28] Fish or aquatic animals ingesting amount of fibers higher than other shapes can be due to dominance of fibers in their environment compared to others. This is supported by the study results of Vo[9] and Le[11] with predominance of fibers in surface waters and benthic sediments.

In this study, we found that the studied fish species mostly ingested microplastics in the small size range (500-2000 µm for fibers and 45000 - 200000 µm^2 fro fragments). Similar results are also recorded by Vo and Vo[10] in blood cockle distributed in Thi Nai lagoon, with most fibers long from 500 to 2100 µm. However, the area of fragments observed by these authors is bigger than that in our study. Gopal et al.[22] found that 85% of microplastics obtained from 10 studied fish species had the size of less than 500 µm while ones from 500 – 5000 µm were found with low percentage. Thus, it can be seen that aquatic creatures often ingest a large proportion of microplastics in small size range. This may be due to accumulation concentration of small-sized microplastics in their habitats is higher than that of large-sized microplastics. The evidences of this can be found in the study results of Vo[9] and Le[11]. However, despite ingesting small-sized microplastics, size range together with corresponding percentage of microplastics accumulated in different fish species differ, which can be governed by their feeding habits.

4. CONCLUSION

Feeding habits of maned goby, redspot arrowfin goby and tropical sand goby are different but occurrence frequency of organic matters is higher than that of other food in the diets of these species.

The concentration of microplastics accumulated in three studied goby species is not significantly different and vary from 6.50 to 9.20 microplastics/individual, 0.58 to 1.16 microplastics/g wet body weight, and 31.47 to 57.55 microplastics/g digestive tract weight.

Three studied goby fish species ingest a large amount of 500 - 2000 µm fibers.

Feeding habits does not affect amount of ingested microplastics and the main source of microplastics accumulated in three studied goby species is mostly from organic matters.

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