MICROPLASTIC FIBER ACCUMULATION IN SOME WILD AND CULTURED SHRIMP SPECIES

Vo Van Chi^{a*}, Tran Thi Bich Hang^{a, b}

^aThe Faculty of Natural Sciences, Quy Nhon University, Binh Dinh, Vietnam ^bTam Quan Secondary School, Binh Dinh, Vietnam ^{*}Corresponding author: Email: vovanchi@qnu.edu.vn

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Abstract

This study was conducted to examine the microplastic fiber abundance and its characteristics in two wild and two cultured shrimp species collected at coastal areas in Hoai Nhon district, Binh Dinh Province. A total of 84 individuals of four shrimp species, Litopenaeus vannamei, Penaeus monodon, Metapenaeus ensis, and Penaeus semisulcatus were collected for analysis. All shrimp specimens were dissected to remove the digestive tracts, which were then treated with 10% KOH to collect microplastic fibers by filtering. The microplastic fiber concentrations varied from 1.96 ± 0.09 to 19.33 ± 10.82 fibers/specimen or 0.20 ± 0.12 to 2.26 ± 1.26 fibers/g of wet body weight. The abundance of fibers in the wild shrimps (Penaeus semisulcatus and Metapenaeus ensis) was higher than in the cultured shrimps (Penaeus monodon and Litopenaeus vannamei). Most fibers observed in the four shrimp species had lengths between 300 and 1500 µm, accounting for 78.72% to 92.82% of the total). White fibers were dominant (30.38%), followed by gray (11.87%), and green (10.60%).

Keywords: Aquatic environment; Microplastic; Microplastic fibers; Pollution; Shrimp.

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1. INTRODUCTION

Microplastics, defined as plastic items from 1 to 5000 μ m in size (Arthur et al., 2009), have received increasing attention from many researchers worldwide. Microplastics were first found in 1972 by Carpenter and Smith (1972) in the pelagic surface water of the Sargasso Sea. To date, microplastics have been found in different environments, such as waters, from rivers to coastal waters (Alomar et al., 2016; Wang et al., 2017; Zhao et al., 2014) and sediments (Mao et al., 2021; Strady et al., 2021), and in aquatic animals, such as molluscs (Abidli et al., 2019; Li et al., 2015; Li et al., 2016; Naji et al., 2018), fish (Lusher et al., 2012; Wu et al., 2020), and crustaceans (Carreras-Colom et al., 2020; Murray & Cowie, 2011). Although microplastics are found in different shapes, including fibers, fragments, pellets, films, and foams, fibers are the most abundant (Bikker et al., 2020; Fu et al., 2020; Mao et al., 2021; Napper et al., 2021; Strady et al., 2021). Many aquatic animals ingest microplastic fibers because they mistake them for food, resulting in a negative impact on their health and the health of other organisms via the food chains (Wright et al., 2013).

Many studies have been conducted worldwide to indicate microplastic contamination status in environments and organisms, but such studies have been rare in Vietnam, although Vietnam ranks fourth in the world in emitting plastic waste into oceans (Jambeck et al., 2015). The first several research studies on microplastics in Vietnam mostly focused on aquatic environments and sediments (Nguyen et al., 2020; Strady et al., 2021). Recent studies in Vietnam have found microplastic contamination in aquatic organisms, including bivalves (Kieu-Le et al., 2022; Le & Vo, 2022; Vo & Vo, 2022) and fish (Nguyen, 2021), but scientific data on other organisms are still lacking. Many aquatic animals are known to play an important role as seafood in the economic development of Vietnam. Among these organisms, shrimp are a favorite food of local residents in different areas of the country and an important national export product. Therefore, examining the microplastic accumulation in shrimps is necessary.

Binh Dinh Province has a long coastline with coastal waters convenient for fishery development. And Hoai Nhon is one of the provincial districts that have developed fishing and aquaculture, with an emphasis on shrimp products. However, the coastal areas of Hoai Nhon have also accumulated quantities of microplastics that can be ingested by aquatic organisms via the food web. The first study to indicate that some fish species collected from these areas ingested microplastics (Nguyen, 2021) found an average concentration of 11.03 ± 7.1 particles per individual, but such information on shrimp is not yet known. Therefore, we selected four shrimp species for this study, *Litopenaeus vannamei*, *Penaeus monodon*, *Metapenaeus ensis*, and *Penaeus semisulcatus*, that are commonly cultured or harvested from the wild and examined the contamination levels and the characteristics of the microplastic fibers. This study will provide scientific data on microplastic contamination in shrimp that can help local authorities develop suitable strategies to reduce the potential risks to human health from microplastics in seafood such as shrimp.

2. METHODOLOGY

2.1. Sample collection

A total of 84 individuals of four shrimp species, *Litopenaeus vannamei, Penaeus monodon, Metapenaeus ensis*, and *Penaeus semisulcatus*, were collected from the wild waters and shrimp ponds in coastal areas of Hoai Nhon district, Binh Dinh Province. The collected shrimps were placed in zip lock bags, frozen, and brought to the lab for later analysis.

Species	Weight (g)	Total length (cm)	Habitat
Litopenaeus vannamei (n=24)	$10.35\pm2.11^{\rm c}$	11.69 ± 0.62^{b}	Shrimp ponds
Penaeus monodon (n=18)	$26.92\pm6.40^{\mathrm{a}}$	$15.28\pm2.11^{\mathtt{a}}$	Shrimp ponds
Metapenaeus ensis (n=24)	2.35 ± 0.38^{d}	$6.93\pm0.36^{\text{c}}$	Wild waters
Penaeus semisulcatus (n=18)	$19.52\pm5.04^{\text{b}}$	$14.67 \pm 1.30^{\text{a}}$	Wild waters

Table 1. Size and habitat of shrimp species

Notes: For each parameter, the different letters indicate the significant difference (p<0.05).

2.2. Sample treatment and observation

After measuring lengths and weighing, the shrimps were thoroughly rinsed with filtered tap water and carefully dissected to remove the digestive tracts. The digestive tracts were placed in glass beakers, treated with 10% KOH, covered with aluminum foil, and placed in an incubator at 60° C for 24 hours according to the method of Dehaut et al. (2016). The samples were then filtered through a 250- μ m mesh sieve, and the material on the sieve was rinsed into a beaker to perform the overflow technique using a saturated NaCl solution. Finally, the sample solution was filtered through a 1.6- μ m GF/A filter using a glass filtration unit connected to a vacuum pump. The filters were stored in clean Petri dishes for stereoscopic observation.

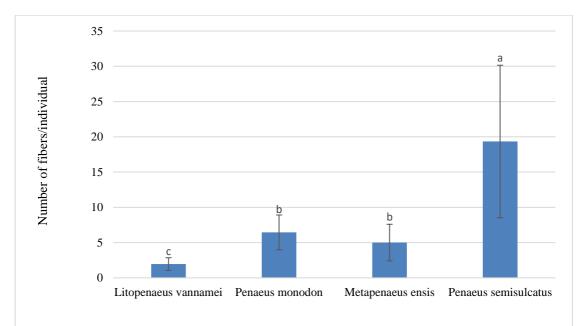
The microplastic fibers on the filters were observed with a Leica S9i stereomicroscope and measured with LASX software. The lengths and colors of the fibers were recorded. We selected a size range of 300 to 5000 μ m for observations of the fibers based on the recommendation of GESAMP (2019) for cases in which μ FTIR or other similar equipment is unavailable.

2.3. Contamination control

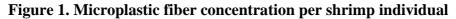
We followed the recommendations of GESAMP (2019) to control the microplastic contamination from the surrounding environment during sample treatment and observation. We cleaned the working area with alcohol, wore cotton clothes and sterile hand gloves, and rinsed the equipment with water filtered through a GF/A glass fiber filter (1.6- μ m pore size) before use. In addition, we carried out the sample treatment and observation in a closed, air-conditioned room to reduce air circulation. At each step of the sample treatment and observation, we placed a new filter in a sterile Petri dish near

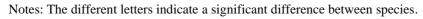
where we were working as a control to check for microplastic contamination. This filter was then also observed under the Leica S9i stereomicroscope to look for contamination. Only three control filters were found to be contaminated, each by one microplastic fiber.

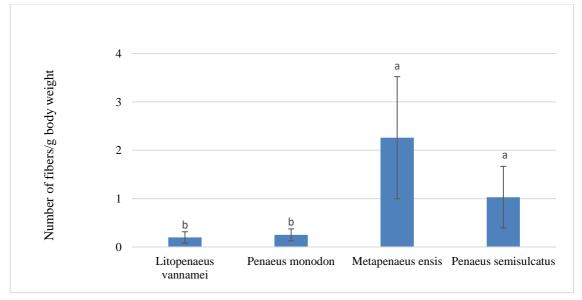
3. **RESULTS AND DISCUSSION**

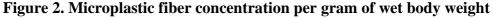


3.1. Microplastic abundance









Notes: The different letters indicate a significant difference between species.

The microplastic fiber concentration was expressed as the number of fibers per shrimp individual and the number of fibers per gram of wet body weight. The average fiber concentration per individual in the four shrimp species varied from 1.96 ± 0.09 to 19.33 ± 10.82 . This value was highest in *Penaeus semisulcatus*, followed by *Metapenaeus ensis* and *Penaeus monodon*, and was lowest in *Litopenaeus vannamei* (p < 0.05, Figure 1). However, the number of fibers per gram of wet body weight of *Penaeus monodon* (0.25 ± 0.12) was not significantly different from *Litopenaeus vannamei* (0.20 ± 0.12) and was significantly lower than that of *Penaeus semisulcatus* (1.03 ± 0.64) and *Metapenaeus ensis* (2.26 ± 1.26) (Figure 2).

Penaeus monodon had the highest wet body weight (Table 1), but not the highest number of fibers per individual or the highest number per gram of wet body weight. In contrast, Metapenaeus ensis ingested a large number of fibers despite its small body size. This means that the weight and size of shrimp are not the major factors governing the level of microplastic contamination. In general, the wild shrimps (Penaeus semisulcatus and Metapenaeus ensis) have high levels of microplastic accumulation compared to the cultured shrimps (Penaeus monodon and Litopenaeus vannamei). Therefore, habitat can be one of the main factors directly affecting microplastic ingestion of these shrimp species. Clearly, the wild habitats in coastal areas can be affected by microplastics released daily from various sources, such as domestic wastewater, industrial activity, waterway activity, and fishing, while the microplastic source in shrimp ponds is mostly from water used during the culture cycle. Moreover, researchers have found a large number of microplastics accumulated in coastal sediments (Le, 2021; Strady et al., 2021; Vo, 2021). Shrimp are benthic organisms that feed on debris, leading to a high microplastic intake. In contrast, mud layers of shrimp ponds are regularly removed after a culture cycle. Therefore, microplastic accumulation in cultivated shrimp will be low.

Our study shows that all four shrimp species ingested microplastic fibers in concentrations from 1.96 to 19.33 fibers/shrimp or 0.2 to 2.26 fibers/g of wet body weight. Microplastic contamination in shrimp has also been reported in other studies. Devriese et al. (2015) showed that Crangon crangon specimens collected from the English Channel and the southern part of the North Sea off the coasts of France, Belgium, the Netherlands, and the UK have accumulated 0.68 \pm 0.55 microplastics/g wet weight or 1.23 \pm 0.99 microplastics/shrimp. A higher concentration of microplastics was observed in L. vannamei from Malaysia, with 1.20 ± 1.06 fibers/g wet weight. Very high levels of contamination were recorded in Pleoticus muelleri from the southwest Atlantic near Argentina and *Fenneropenaeus indicus* from the Indian Ocean, with 468 ± 104 fibers/g wet weight and 1100 ± 51.5 fibers/g wet weight, respectively (Curren et al., 2020). The level of microplastic accumulation in the shrimps in our study is a little bit higher than that found in Crangon crangon by Devriese et al. (2015) or in L. vannamei from Malaysia but much lower than that in *Pleoticus muelleri* and *Fenneropenaeus indicus*, as found by Curren et al. (2020). Compared to the results of the study carried out in the same province (Vo & Vo, 2022), the microplastic abundance in shrimps in our study is also lower than in the mollusc species Anadara granosa (3.26–13.00 microplastics/individual) distributed in Thi Nai lagoon. Another study in Vietnam by Kieu-Le et al. (2022) also

shows a higher fiber concentration per gram of wet weight for the clam *Meretrix lyrata* (2.7 ± 2.4) compared to our results.

3.2. Length distribution of microplastic fibers

The length distribution of the fibers differed between shrimp species. The length of fibers found in *Litopenaeus vannamei* (302–3282 µm) and *Penaeus monodon* (300–2979 µm) were shorter than those in *Metapenaeus ensis* and *Penaeus semisulcatus* (300–5000 µm). However, the median length of fibers observed in *Litopenaeus vannamei* (988 µm) was the highest, followed by *Metapenaeus ensis* (881 µm), *Penaeus monodon* (879 µm), and *Penaeus semisulcatus* (718 µm) (Figure 3). In general, the number of 300–1500-µm fibers was greater than the number of fibers longer than 1500 µm. Shorter fibers, from 300 to 1500 µm in length, accounted for 78.72% in *Litopenaeus vannamei* to 92.82% in *Penaeus semisulcatus*, whereas the percentage of longer fibers varied from 7.18 to 21.28% (Figure 4). Other researchers have also found small fibers to be more abundant than long fibers. For example, microscopic fibers 200 to 1000 µm in length accounted for 63% of total microplastics observed in *Crangon crangon* (Devriese et al., 2015). Similar results have also been reported in other studies (Cabernard et al., 2018; Wang et al., 2017; Zhao et al., 2014). Hebner and Maurer-Jones (2020) identified water currents and ultraviolet radiation as factors that break microplastic size.

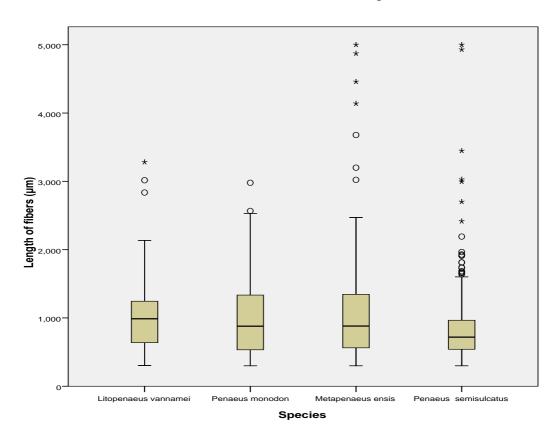
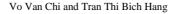


Figure 3. Microplastic fiber lengths (µm) of four shrimp species



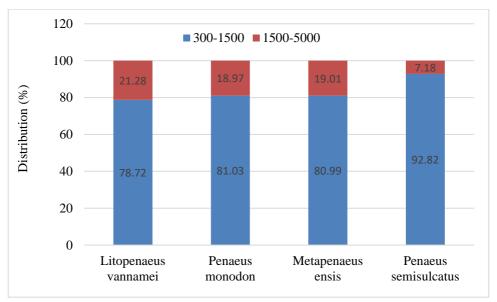
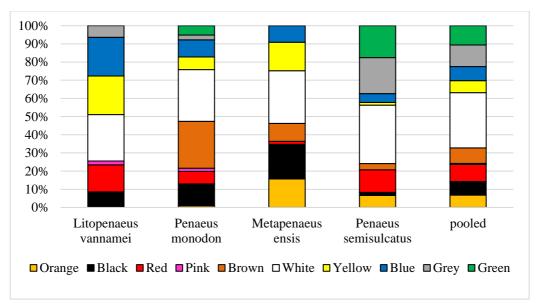


Figure 4. The dominance of fibers with lengths of 300 to 1500 μ m



3.3. Microplastic fiber colors

Figure 5. The distribution of fiber colors observed in four shrimp species

The fiber colors differed between the shrimp species. White was the most common fiber color in all four species (from 25.52 to 32.18% of the total) but the distribution of the other colors was not similar between these species. The other common colors were blue, yellow, and red in *Litopenaeus vannamei*; brown in *Penaeus monodon*; black, yellow, and orange in *Metapenaeus ensis*; and gray and green in *Penaeus semisulcatus*. When pooled for the four species, white was also the most common fiber color (30.38%), followed by gray (11.87%) and green (10.60%) (Figure 5). Vo and Vo (2022) found white microplastics, followed by yellow, to dominate in *Anadara granosa*. White microplastics

were also dominant in the digestive tracts of fish, together with blue (Hastuti et al., 2019). Our results are very different from the observation of Devriese et al. (2015) in *Crangon crangon*, yellow-greenish (50%), purple-blue (43%), translucent (15%) and orange (12%) are common colors. Thus, the color of microplastics observed in organisms differs and can be governed by different sources of microplastics at the different study locations (Gallagher et al., 2016).

4. CONCLUSION

The microplastic fiber concentrations in the four shrimp species varied from 1.96 ± 0.09 to 19.33 ± 10.82 per individual or from 0.20 ± 0.12 to 2.26 ± 1.26 per gram of wet body weight. The wild shrimps (*Penaeus semisulcatus* and *Metapenaeus ensis*) had higher levels of microplastic accumulation than the cultured shrimps (*Penaeus monodon* and *Litopenaeus vannamei*). The percentage of shorter fibers, with lengths of 300 to 1500 µm, varied from 78.72% to 92.82% and was greater than the percentage of fibers longer than 1500 µm (7.18 to 21.28%). White was the dominant fiber color (30.38%) in the four shrimp species, followed by gray (11.87%) and green (10.60%).

In summary, this study provides important scientific information on microplastic accumulation levels and their characteristics in wild and cultured shrimps in the coastal areas of Hoai Nhon district, Binh Dinh. Although this study was conducted in only a small area of the province and on four shrimp species, it is hoped that these results, together with the results of future studies, can contribute to developing suitable strategies to reduce the potential risks to human health from microplastics in seafood. However, to do this, future studies need to prove or clarify the real health risks from microplastic contamination in organisms in Vietnam, which may take much effort and time.

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