ORIGINAL PAPER

Occurrence of microplastics in two edible seaweeds from local aquaculture in Thailand

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Received: 08 August 2021 / Revised: 30 August 2021 / Accepted: 30 August 2021

Abstract. Microplastics have been found to adhere on seaweeds, which serve as a route for microplastic transfer into the marine food web and human consumption. Because there is a lack of understanding about microplastics in edible seaweeds, this study focused on the occurrence of microplastics in two edible seaweeds from Thai aquaculture. The edible red seaweed *Gracilaria fisheri* from aquaculture in Songkla Province and the edible green seaweed *Caulerpa lentillifera* from aquaculture in Samut Sakhon and Phetchaburi Provinces were used in this study. Microplastics were found in seaweeds in concentrations ranging from 16.46±2.56 to 181.73±86.42 particles per 100g wet weight. The highest abundance of microplastic were observed in the red seaweed *G. fisheri* with 181.73±86.42 particles per 100g wet weight, while the lowest one was observed in the green seaweed *C. lentillifera* with 16.46±2.56 particles per 100g wet weight. Microplastics in the size classes 100-500 and 501-1,000 µm were abundantly found in this study. Black and blue fibrous microplastics were frequently found in the green seaweed *C. lentillifera* with 63.16% of microplastic found at Phetchaburi Province and 42.59% of microplastics found at Samut Sakhon Province. Most microplastics found in this study was polypropylene (PP). Because buoyant microplastics were abundant in the first meter water surface where *G. fisheri* were planted in a shallow water pond, the total accumulation of microplastics on *G. fisheri* was considerably higher than *C. lentillifera* (p<0.05). The presence of microplastic pollution on seaweeds was highlighted in our study. Consequently, water management is essential to reduce microplastic contamination in edible seaweeds. Further research is also needed to precisely detect their presence and measure their abundance.

Keywords: *C. lentillifera*, *G. fisheri*, marine debris, contamination, macroalgae

1. Introduction

Several studies have been conducted to address the global challenges of food shortage and prospective food security, with human population growth identified as one of the primary drivers (Grafton et al., 2015, Bazerghi et al., 2016). Under these conditions, the oceans are a vast source of resources that can play an important role in increasing and sustaining food production. However, an estimated 268,940 tonnes of plastics are currently floating in the world's oceans, with a portion of them forming microscopic plastic fragments known as microplastics (Eriksen et al., 2014). Plastic pollution is being blamed on an increase in the use of plastic, and poor waste management. This plastic pollution raises concerns about human exposure through food, despite the fact that the effects on human health are largely unknown and potential toxicity mechanisms are not well understood (Jambeck, 2015; Neufeld et al., 2016; Wright and Kelly, 2017; Chinfak et al., 2021; Sutthacheep et al., 2021).

Seaweeds (macroalgae) are high in polysaccharides, minerals, vitamins, and bioactive substances such as fucoxanthin, phenolic compounds, and fucoidan, which act as antioxidants, anti-viral, anti-hepatitis, and anti-obesity agents, they are frequently referred to as superfoods (Kumar et al., 2008; Brown et al., 2014). The edible green seaweed *Caulerpa lentillifera* and the edible red seaweed *Gracilaria fisheri* are the two seaweed species found in Thailand. Due to its high rate of growth and nutrient and antioxidant content, *Caulerpa lentillifera* is cultured as a commercial effort in Thailand, and *Gracilaria fisheri* has been served fresh or used in dried products for both humans and animals (Benjama & Masniyom, 2011; Benjama and Masniyom, 2012; Wichachucherd et al., 2019). Several studies have been reported that seaweeds may represent an efficient pathway for microplastics from the water to marine animals (Gutow et al., 2015; Seng et al., 2020). Moreover, a previous study has been conducted to investigate the direct human exposure to microplastics from seaweed consumption (Li et al., 2020). An assessment of current microplastics exposure levels in humans has been identified as a critical step toward determining the potential for adverse effects on human health (Wright & Kelly, 2017). The knowledge on microplastics in edible seaweeds also must be considered. Therefore the aim of this study was to investigate the abundance of microplastics on the edible seaweeds from aquaculture in Thailand.

2. Materials and Methods

Two edible seaweeds, *C. lentillifera* and *G. fisheri*, were purchased from local farmers in Thailand during January-February 2020. The green seaweed *C. lentillifera* were harvested from the hanging cage in soil pond at Samut Sakhon and Phetchaburi provinces, and the red seaweed *G. fisheri* were harvested from a shallow water pond with approximately 50 cm in depth in Songkla Province. The green seaweed *C. lentillifera* is well-known in Thailand's middle path, whereas the red seaweed *G. fisheri* is well-known in Thailand's southern path.

2.1 Microplastics isolation

Under a stereo microscope, the microplastics on a 30g wet weight of *C. lentillifera* and a 10g wet weight of *G. fisheri* were observed and counted in three replicates, and the method was described in Seng et al. (2020) method. Photographs of microplastics on both seaweeds were taken with a digital camera (Olympus, TG-6), and the microplastics were then removed with a needle. Under a compound microscope, the length and colors of microplastics samples were measured. Microplastics size classes were divided into four classes (100-500, 501-1,000, 1,001-2,000, and 2,001-5,000 µm). The microplastics types were identified by using a Fourier transform infraredspectroscopy (FTIR).

2.2 Statistical analysis

The abundance of microplastics in seaweed were expressed in particles per 100 wet weight. Duncan's new multiple range test in R program version 3.6.2 was used to compare microplastics in seaweeds across sites. The difference of total microplastics in seaweed was tested by using the student's t-test in R program.

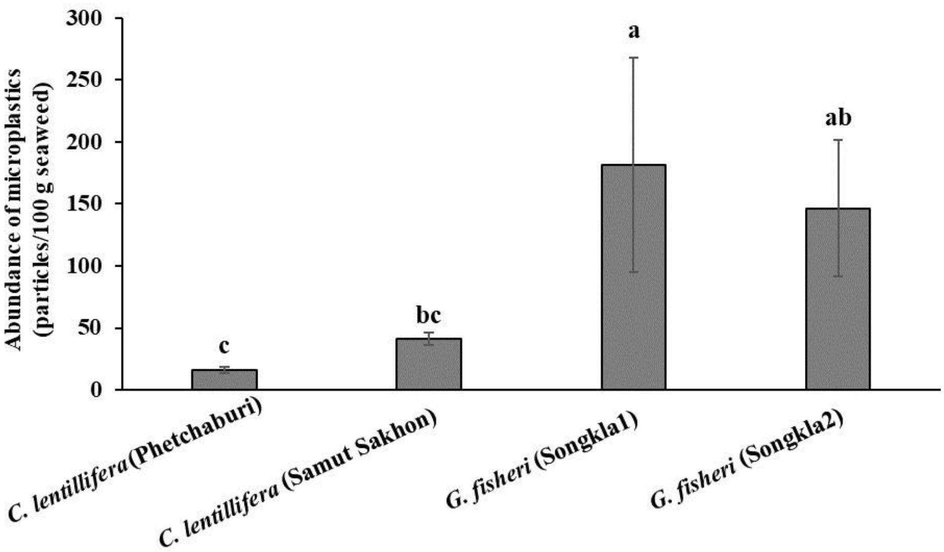

Figure 1. Abundance of microplastics in edible seaweeds from aquaculture

3. Results

The abundance of microplastics in seaweeds ranged from 16.46±2.56 to 181.73±86.42 particles per 100g wet weight. The highest abundance of microplastic in seaweed was found in *G. fisheri* (Songkla 1), while the green seaweed *C. lentillifera* (Phetchaburi) exhibited significantly lower than other sites (p=0.05) (Figure 1). The average of total microplastics in *G. fisheri* were significantly higher than those green seaweed *C. lentillifera* (t = -4.004, df = 5.3105, p-value = 0.0091).

Microplastics were mostly found with lengths ranging from 100 to 2,000 µm. *C.lentillifera* (Phetchaburi) and *G. fisheri* (Songkla2) did not contain microplastics with sizes ranging from 2,001 to 5,000 µm. Microplastics with size class of 100-500 µm were frequently found in *C. lentillifera* (Samut Sakhon) with an average length of 474.86±320.59 µm, while microplastics with a size class of 501-1,000 µm were frequently found in *G. fisheri* (Songkla1) with an average length of 826.65±338.81 µm (Figure 2 and Table 1). All seaweed samples exhibited 100% of fibrous microplastics, which is the red fibrous microplastics that were commonly found in the red seaweed *G. fisheri*, whereas the black and blue fibrous microplastics were frequently found in the green seaweed *C. lentillifera* (Table 1 and Figure 3). The microplastics types identified by using an FTIR showed that most microplastics were polypropylene (PP).

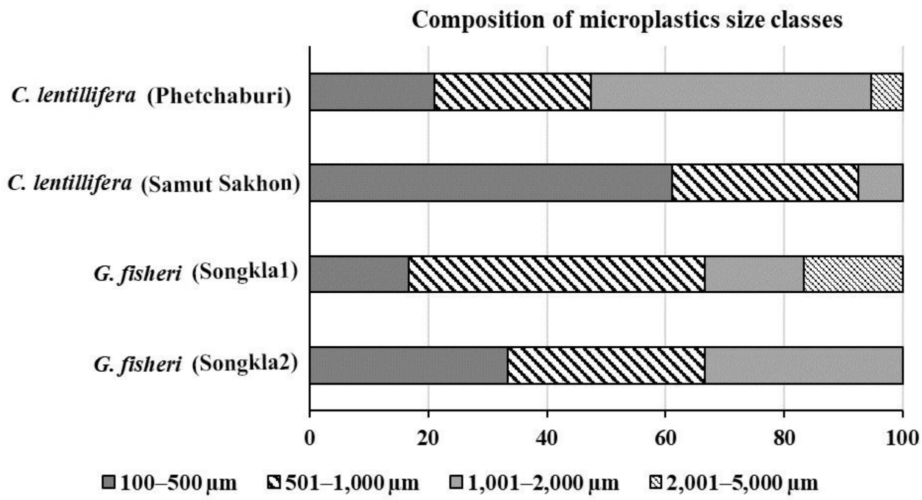

Figure 2. Size class composition of microplastics found in edible seaweeds

Table 1. Characteristics of microplastics found in two edible seaweeds

Microplastics characteristics	Seaweed sources			
	C. lentillifera (Phetchaburi)	*C. lentillifera* (Samut Sakhon)	*G. fisheri* (Songkla1)	*G. fisheri* (Songkla2)
Average size (µm)	1,071.07±584.89	474.86±320.59	1,058.15±707.90	826.65±338.81
Forms (%)				
Fibre	100	100	100	100
Fragments	-	-	-	-
Colors (%)				
Black	63.16	42.59	-	-
Blue	26.32	42.59	-	50.00
Green	-	1.85	-	-
Red	5.26	12.96	83.33	50.00
Brown	-	-	16.67	-
Violet	5.26	-	-	-

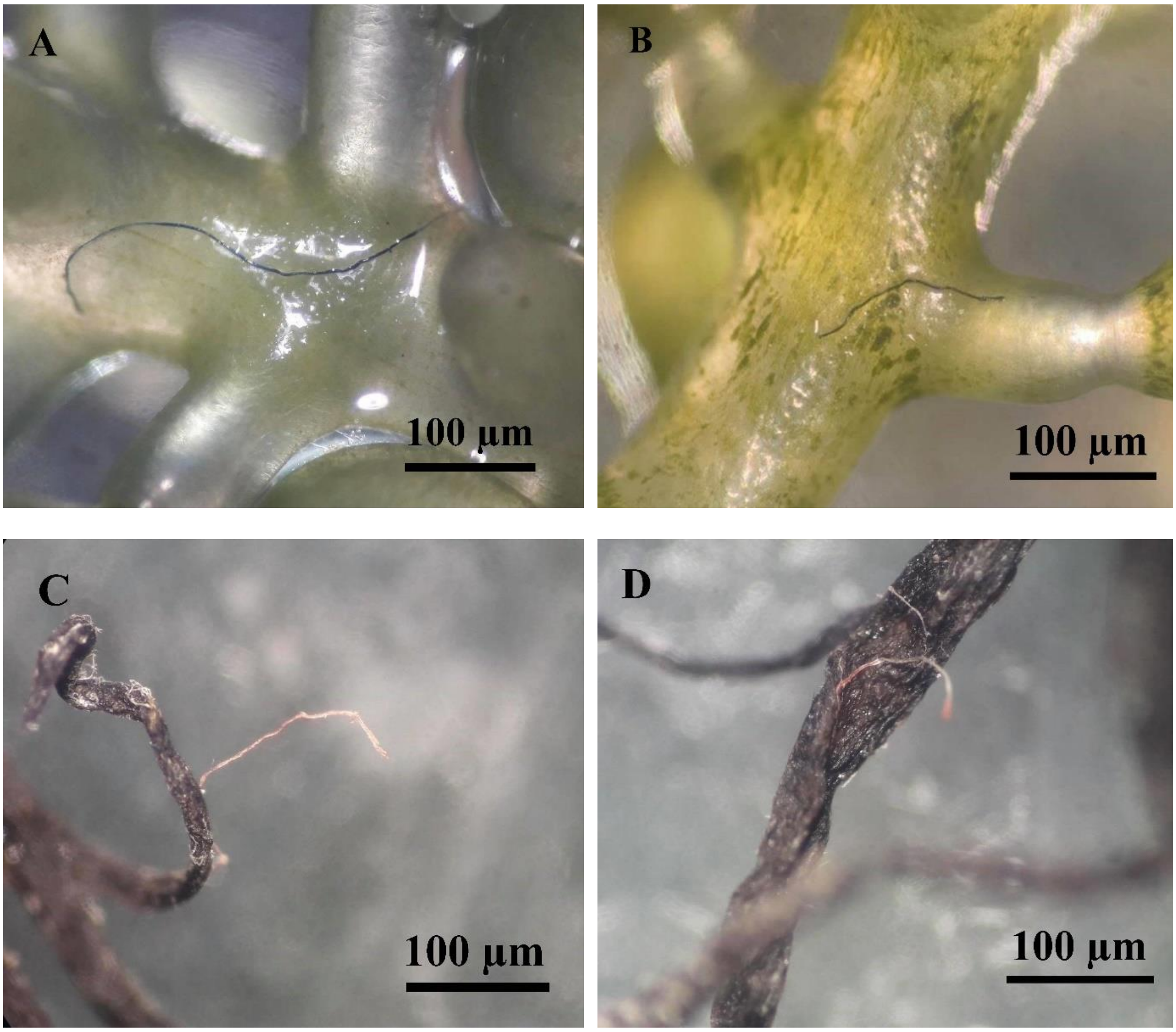

Figure 3. Microplastics particles on *C. lentillifera* (A, B) and *G. fisheri* (C, D)

4. Discussion

The deposit of microplastics on marine macroalgae is a potentially harmful warning, but the route by which microplastics reach the human body has been overlooked (Seng et al., 2020). In a previous study, Li et al. (2020) discovered the first evidence of microplastics on commercial seaweed nori (*Pyropia* spp.). Our study further confirmed the presence of microplastics on two macroalgal species from aquaculture in Thailand. Our results revealed significant differences in microplastic densities among macroalgae taxa, with red seaweed *G. fisheri* having higher than that green seaweed *C. lentillifera*. Moreover, our results showed that the accumulation of microplastics on *C. lentillifera* (ranged from 2.57 to 9.55 particles/g dry weight (dw)) and *G.fisheri* (ranged from 3.81 to 13.87 particles/g dw) was significantly high when compared to commercial seaweed nori (*Pyropia* spp.) ranged from 0.9 to 3.0 particles/g dw (Li et al., 2020). This evidence was also discovered in similar sea agricultural locations for other commercial species, including fish, mussels, and oysters (Mathalon and Hill, 2014; Su et al., 2019; Severini et al., 2019).

The majority of microplastic size classes from *C. lentillifera* (Samut Sakhon) found in this study ranged from 100-1,000µ m, which is similar to the size class of microplastics found near the Chao Phraya River Estuary (Oo et al., 2020). Furthermore, Vibhatabandhu and Srithongouthai (2021) reported that microplastics in the 125-1,000 µm range are abundant in the inner Gulf of Thailand, with an average total abundance of 9.97 pieces/L. In Songkla Province, the abundance of microplastics was only discovered in beach sediment (Thepwilai et al., 2021). However, Amin et al. (2020) discovered that the majority of microplastic particles found in the coastal waters of Terengganu State, Malaysia, which borders the southern region of Thailand, were fibers and fragments including the total concentration of microplastics was the highest at the nearshore station (9 particles/L). Several studies have been reported that abundant types of microplastics in the Gulf of Thailand were fibrous microplastics (Azad et al., 2018; Pradit et al., 2020; Sukhsangchan et al., 2020; Wang et al., 2020).

The abundance of microplastic in seaweed may have been influenced by water depth and sampling site location (Seng et al., 2020). The highest concentrations of microplastics are known to accumulate within the first metre beneath the sea surface from buoyant microplastics (Kooi et al., 2016). Because the red seaweed *G. fisheri* was sampled from shallow waters, while the green seaweed *C. lentillifera* was harvested from a hanging cage, differences in depth-associated microplastic concentrations in seawater may have contributed to the microplastic abundance patterns. Therefore, further research is needed to determine their presence and quantify their abundance more intensively to better understand their potential sources and fates in various environments.

Acknowledgements

This study was funded partly by a budget for research promotion from the Thai Government awarded to Ramkhamhaeng University. We are most grateful to the staff of Marine Biodiversity Research Group, Faculty of Science, Ramkhamhaeng University for their support and assistance in the laboratory and field surveys. This paper is the one of the Thailand's project activities under the V2V Global Partnership.

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