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REVIEW PAPER

Microplastics ingestions by wild and aquaculture marine bivalves: A systematic review on field investigation study

Ni Luh Gede Rai Ayu Saraswati*
Department of Aquatic Resources Management, Faculty of Marine Science and Fisheries, Udayana University, Bali-Indonesia

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Abstract. Plastics degradation has resulted in a major threat to marine organisms, including bivalves. Thirty-three peer-reviewed papers have been reviewed to understand the geographical spread of microplastics ingestion by marine bivalves, characteristics of microplastics ingested, and limitation of microplastics analysis globallly. Only studies on microplastics investigation in marine bivalves from wild and aquaculture area were selected. Marine bivalves are reported to accumulate microplastics from all marine environment compartments. High proximity area with intensive human activities is suggested to increase the uptake of microplastics by the bivalves. Microfibers and fragments are the common types of microplastics ingested by the bivalves around the world, with various sizes (0.45µm – <45mm) and number of particles per individual (20 – ~175 particles/individual). However, there is uncertainty when comparing the findings from one study to another due to the absence of international standard protocol and microplastics data base. Therefore, this limitation should be addressed prior to monitoring microplastics accumulation in marine bivalves.

Keywords: Microplastics ingestion; wild and aquaculture bivalve; biomonitoring

1. Introduction

Plastic is undoubtely a versatile material which is highly used in today's modern society. About 90% of plastic material is fossil-based and produced to different categories of common plastic types, such as polypropylene (PP), polyethylene (PE), polyethylene terephthalate (PET), and polyvinyl chloride (PVC) (Plastics Europe, 2022). Due to the high demand for plastic products, plastics production globally have increased by more than 50% since the production from the 1950s (1.7 MMT) to 2021 (390.7 MMT) (Plastics Europe, 2011, 2022). However, due to a poor waste management on land, plastics waste leaks into the environment, including the ocean (Jambeck et al., 2015).

About 79% of plastic waste is either found in natural environment or landfill (Geyer et al., 2017). The waste reaches the marine ecosystems mainly through river (Meijer et al., 2021). Based on the recent OECD report, in the ocean, plastic waste is found as macroplastics (88%) and microplastics (12%) (OECD, 2022). Many have reported the impact of macroplastic waste in the

*Corresponding author. E-mail: ayusaraswati@unud.ac.id
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ocean to marine life. Macroplastics have been reported to increase the spread of disease to coral reef (Lamb et al., 2018). Moreover, many of marine animals, such as turtle, seabirds, fish, and marine mamals are found to be entangled and mistakenly ingested with plastic (Gall & Thompson, 2015). As plastics are non-biodegradable material, macroplastic particles will break to tiny particles of microplastics through physical or mechanical degradation process. The degradation then adds up to the current amount of microplastics in the ocean.

Microplastics are defined as plastic particles with the size of less than 5 mm and irregular or regular in shape (Frias & Nash, 2018). Microplastics have spread to all compartments of marine ecosystems (Galgani et al., 2015). Due to its small size, microplastics have been ingested by various marine organisms, from the basic level organisms on food chain, such as zooplankton (Desforges et al., 2015) to higher level organisms, such as bivalves and fish (Markic et al., 2020; Scott et al., 2019). As the particle enter the marine food chain, it is possible that the amount of microplastics magnify on the top tropic level of marine organisms. The ability of microplastics to biomagnify gain a serious concern to the health of both marine organisms and human, as seafood is one of natural source of protein for human.

Marine bivalve is one of the most popular seafood consumed by humans. Bivalves are rich in nutrients for humans, such as vitamin B12, choline, omega-3 fatty acids, and esentials minerals (Fe, Se, Zn) (Wright et al., 2018). About 14.6 million tons of bivalves have been produced from aquaculture globally (Olivier et al., 2020). Different from fish, the entire bivalve soft tissue is consumed by humans. This placed humans at risk on multiple microplastics intake through consumption. As a result, research on microplastics ingestion by marine organisms, including marine bivalve, has gained a serious concern globally due to its potential risk, both to the organisms and humans.

Bivalve is a filter feeder species that has can cleanse water by filtering the organic material, such as phytoplankton and zooplankton, in the water column. Sessile behavior of bivalves makes it prone to environmental contamination, including to microplastics pollution. There are three main factors influencing accumulation of microplastics in bivalves: location of bivalves cultivation, seasonal variations and, proximity of bivalves habitat to contaminant sources (Baechler et al., 2020; Phuong et al., 2018; Reguera et al., 2019). Moreover, due to the global widespread distribution of bivalves, it is possible to compare the microplastic pollution between areas. Therefore, it is suggested that bivalves can be suitable species for microplastics biomonitoring (Li et al., 2019).

Regular microplastics monitoring in bivalves is important to conduct, particularly the wild and cultured bivalves. The practice of bivalves aquaculculture is often in the natural marine environment, which allows the cultured organisms to be exposed with microplastics pollution. The monitoring will help to understand the current state of concentration and contamination level of microplastics ingested by the organisms. It is also suggested that the microplastics in bivalves can indicate the contamination level in the surrounding water (Li et al., 2019). This review aims to understand the microplastics characteristics and geographical spread in wild and cultured marine bivalves globally. The review will also highlight the limitations of conducting microplastics investigation in bivalves for further study. The information from this review will help managers and stakeholders make worldwide decisions about waste management and plastics production.

2. Methods

The flowchart of Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) statement was used in this systematic literature review (Moher et al., 2009) (Figure 1). The papers used in this review were all English papers searched in electronic database of environmental pollution and aquatics journal, including ScienceDirect, Springer, ACS Publication, Google Scholar, and OneSearch. The keywords used were ‘microplastics ingestion’ OR ‘microplastic accumulation’, ‘microplastics mussel’, and ‘microplastics bivalve’. Paper releases range from 2008 to early of 2020. This study did not look at microplastics study in other benthic organisms, such as gastropods and worms, even though they might have similar pattern of microplastics contamination. Only papers published in primary publications from peer-reviewed journal were included. Other grey literature, including books, reports, and news articles were excluded for the review. The reason for this is that the issue around microplastics contamination has recently been widely publicized.

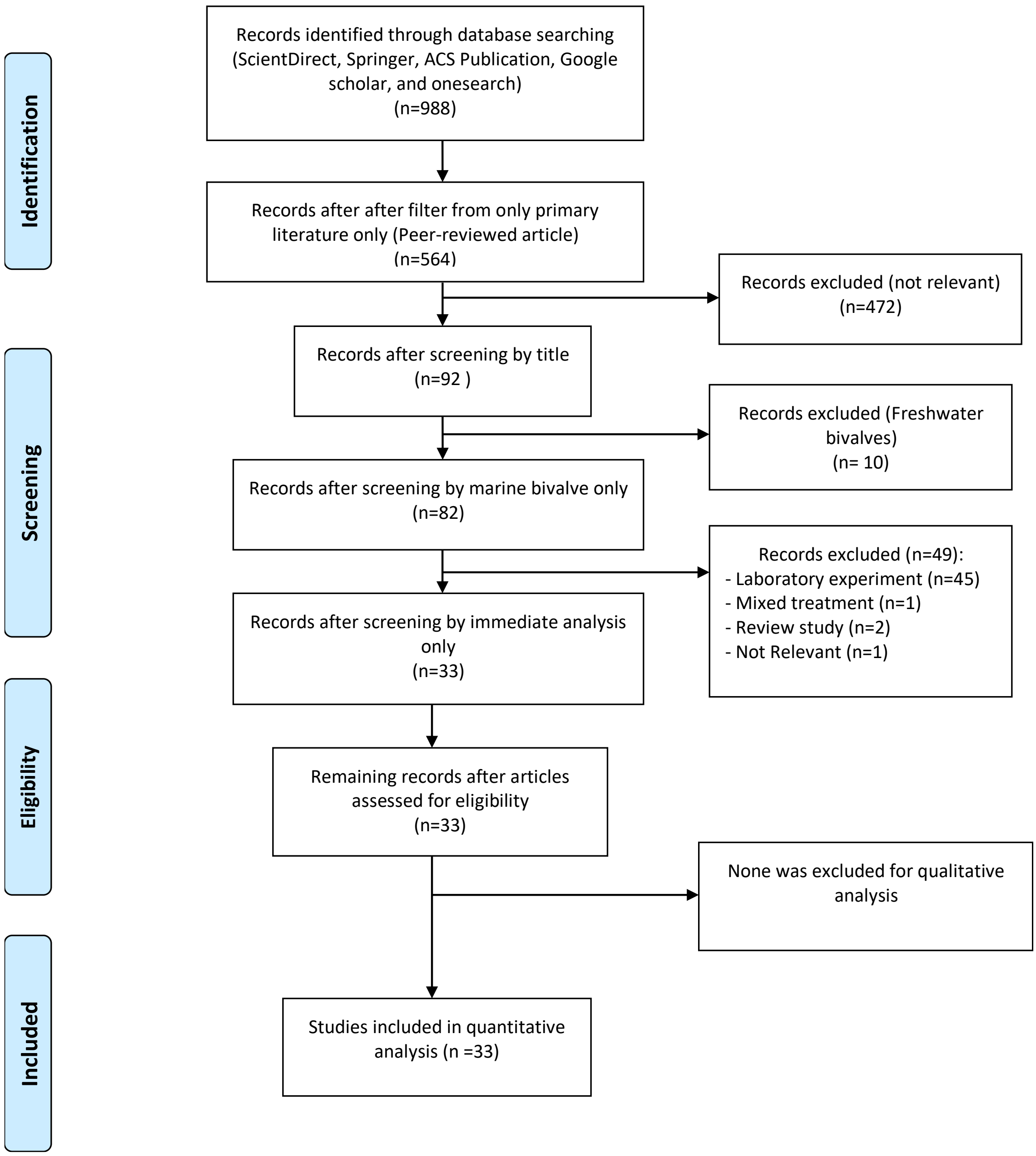

Figure 1. The systematic literature review flowchart adopted from PRISMA statement

There were three screening steps in filtering the papers: matching the title to the keyword, identifying the bivalve habitat (marine species only), and analyzing treatment (Figure 1). The bivalve habitat was divided into two categories: marine and freshwater, while the bivalve sample sources were categorized as wild, aquaculture, market wild and aquaculture, wild and market, and aquaculture and market. The analysis treatment of microplastics in bivalves was categorized as experiment in laboratory, experiment in natural environment, immediate analysis and mixed between experiment in laboratory and immediate analysis. Immediate analysis treatment is when the sample being collected from the environment and immediately analysed or processed for microplastics analysis in the lab. The treatment procedure is completely different with laboratory experiments that require several steps, such as organisms' acclimatization and depuration process prior to executing the microplastics ingestion and exposing the organisms with predetermined contaminant. Only research in marine species of bivalves with immediate analysis (non-laboratory experimental) was eligible and included for this study. Therefore, the laboratory experiment paper was excluded. A simple descriptive method was applied to identify the pattern of microplastics threat to wild and aquaculture marine bivalves. In each included paper, additional information, including the authors, journal source, country, year of publication, database origin, analysis treatment, bivalve habitat, types of microplastics and polymer were recorded.

3. Results and discussions

3.1 PRISMA screening results

Ninety-two peer-reviewed journals were screened by title that examined the study of microplastics accumulation in bivalves. A great number of publications in this issue were published in 2018 and 2019 (28 and 33 papers, respectively), and only eight papers recorded in 2020 (Figure 2a). The majority of papers were obtained from ScienceDirect, accounting for 62 papers, and less than 10 papers were accessed from ACS Publications and Springer. Other databases were a compilation of smaller databases, including IOP Publishing, and Open Journal System (Figure 2b). Based on the analysis treatment (Figure 2c), most of the analysis was conducted by laboratory experiment (45 papers). There were 33 papers conducted using immediate analysis, ranging from estuary to open ocean They were eligible and included for this review study. Only 10 out of 92 papers were using freshwater bivalves in all analysis treatments. This problem appears to have been common in marine bivalves (Figure 2d).

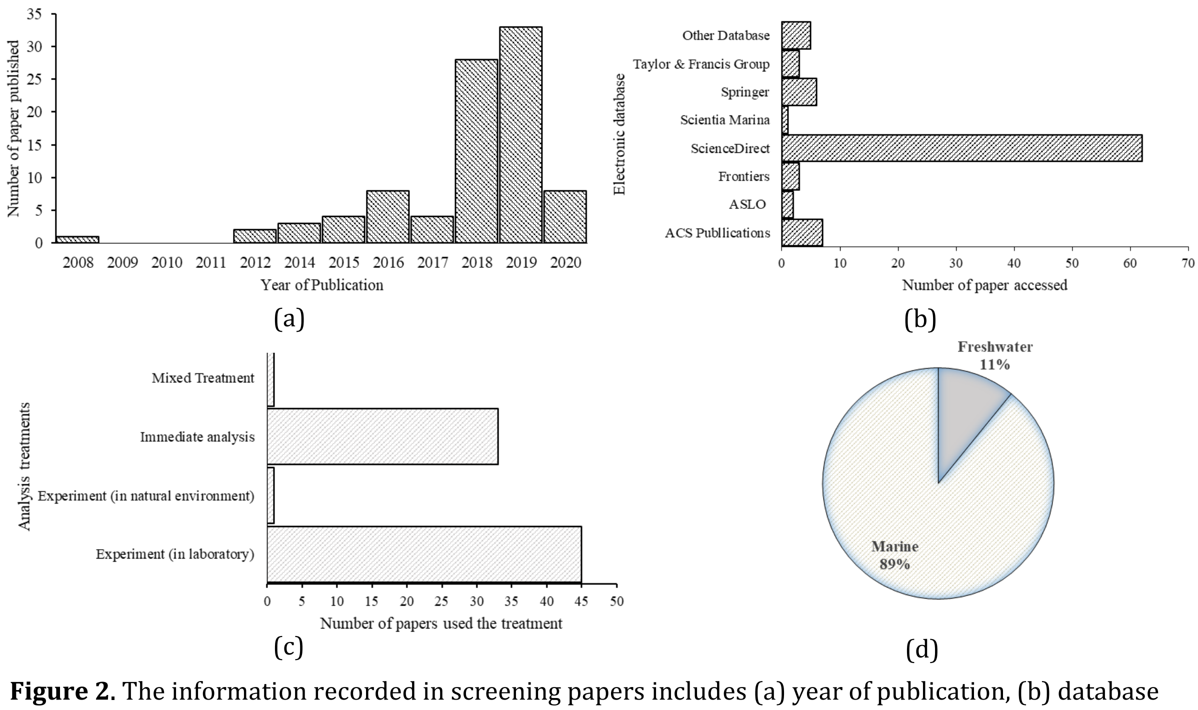

Figure 2. The information recorded in screening papers includes (a) year of publication, (b) database origin, (c) analysis treatments, and (d) bivalve habitats.

3.2. Summary of result from selected papers

A total of 33 papers was eligible for this review. Most of the study were published between 2018 and 2019, in marine and environmental pollution journals. The studies were conducted globally, including in European countries (14 papers), non-European countries (10 papers), Asian countries (7 papers), and the open ocean (2 papers). In terms of the extraction method, nearly all papers applied digestion methods using acid solution, primarily H_2O_2, HNO_3, and KOH at varying concentration. Different reagents were also used in some papers such as using enzyme. Visual identification under the microscope was a compulsory step done by all papers after the digestion of organic material from bivalve samples (Table 1, Appendix 1).

The common type of marine bivalve sampled from the selected papers was mussels (61%), oyster (3%), and mixed of bivalve's species (36%) (Figure 3). The dominant species being investigated was *Mytilus edulis, Mytilus galloprovincialis* and *Crassostrea gigas* (Table 2, Appendix 2). Fifty-three percent of the study sampled bivalves from the wild environment, including from estuaries, coastal beaches and open ocean and other papers sampled the specimen on aquaculture area (13%), market (5%) and a mixed of the three sources (29%).

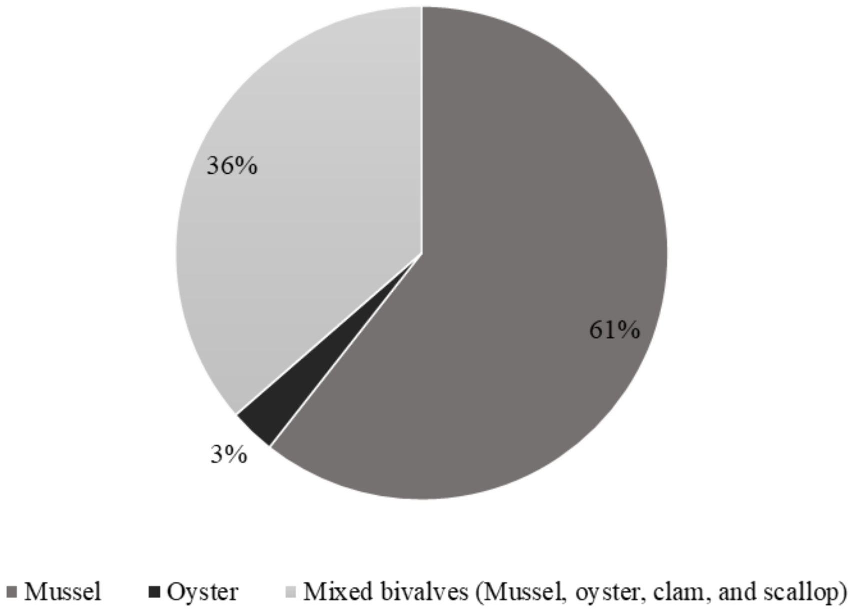

Figure 3. The types of bivalves sampled for microplastics analysis from selected papers

About 66% of papers only investigated the shape of microplastics, while 12.12% only investigated the polymer types of microplastics in their findings. Only 27.27% of the papers managed to conduct both microplastics identifications (shape and polymer identification). Regarding the microplastics shape, microfibers and fragments were the common microplastics ingested by the bivalve (38% and 31% of the papers respectively, Figure 5). Meanwhile, polypropylene (PP) and polyester (PE) were the frequently identified polymer in bivalves (22% for each polymer), followed by polystyrene (10%) and polyethylene (9%) (Figure 6).

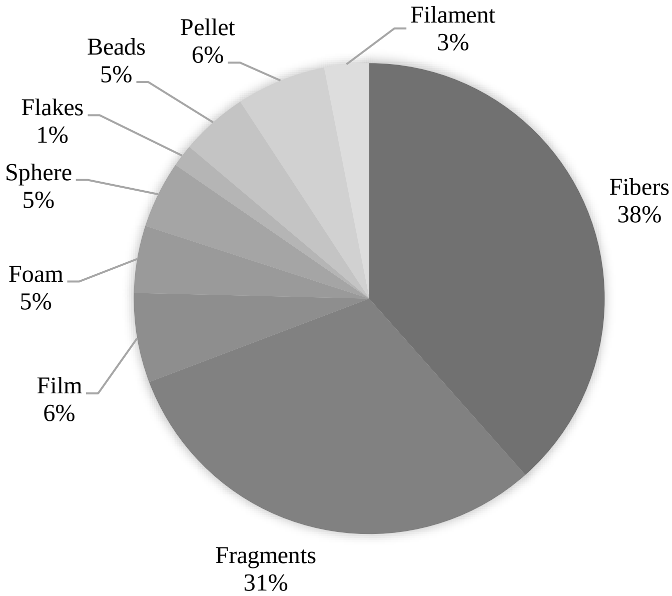

Figure 5. The identification of microplastics shapes from selected papers

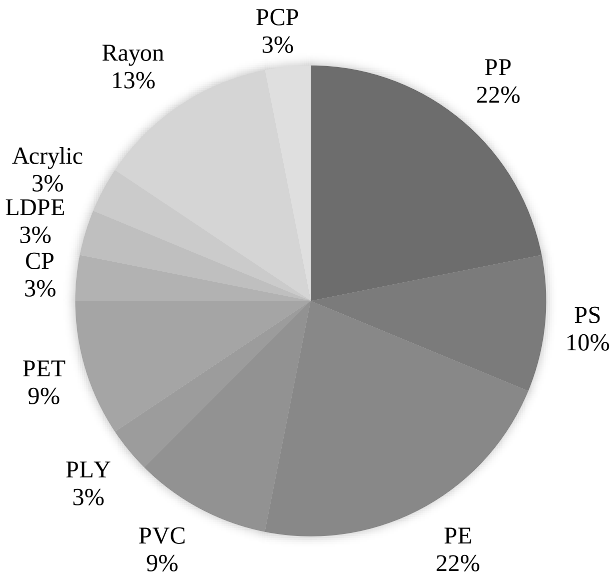

Figure 6. The polymer types of microplastics from selected papers

The size and number of microplastics particles ingested are varied across the location (Table 2, Appendix 2). Most of microplastics identified by the papers was less than 250 μm, with the range of size found globally was from 0.11 μm to more than 45 mm. It is clear that a bivalve ingested at least one particle of microplastics. However, two papers reported a great number of microplastics found in bivalve, ranging from 20 to about 175 particles per individual.

3.3 Discussions

Microplastics investigation in bivalves have been conducted globally from Europe to Asia since 2008. A rapid increase on microplastics investigation in bivalves between the year 2018 and 2019 shows that the contamination becomes a serious concern among the field study (Figure 2a).

All bivalves, both commercial and non-commercial species, reported ingesting at least one microplastic particle from their environment. The most common types of microplastics ingested were mostly fibers and fragments. However, the analysis methods used were varied, which might affect the accuracy of the results.

3.3.1 Global study on microplastics accumulation in marine bivalves

The investigation of microplastics accumulations in wild and cultured bivalve had been conducted in 17 countries globally during the period between 2018 and early 2020. Most of studies were done in European countries, including Belgium, France, Germany, Italy, Norway, Spain, and the UK, as well as in the open ocean. To date, bivalves are the third popular marine organism sampled for microplastics investigations, after fish and crustacean (de Sá et al., 2018). This is parallel with the popularity of fish as food, including bivalves for human consumption (Lusher et al., 2017). Globally, there are four main groups of bivalves that is popular among the seafood consumers: mussels, oysters, clams, and cockles (Wijsman et al., 2018). In relation to the microplastics analysis, it is no doubt that those four main groups of bivalve are chosen to be studied globally (Covernton et al., 2019; Hermabessiere et al., 2019; Li et al., 2019).

The record of sample origin provides information related to the level of ingested microplastics by bivalves. It shows that microplastics have distributed across the habitat of marine bivalves globally, for both wild and aquaculture areas. For example, there is no difference in microplastics ingestion in wild and farmed areas along the coasts of China and Brazil, despite the intensive anthropogenic pressure (Castro et al., 2016; Li et al., 2016). Studies have also confirmed that there is a positive correlation between the amount of microplastics, mainly fibers, in the surrounding environment and the uptake concentration by the bivalve, both water and sediments (Qu et al., 2018; Scott et al., 2019). This indicates that for both wild and aquaculture species are equally being threaten by the microplastics pollution, depending on the contamination input and occurrence on their environment around the world. However, some studies reported that the uptake by the cultured species is higher (Birnstiel et al., 2019; Covernton et al., 2019; Davidson & Dudas, 2016; Phuong et al., 2018). There are several possibilities to explain why cultured marine bivalves ingest more microplastics, such as the use of polypropylene line as media growth, mariculture area's proximity to dense urban area, and contamination during the distribution to market (Mathalon & Hill, 2014). In addition, the dynamic process of the marine environment including current, wave, wind, and tides also influence the distribution of microplastics in the marine environment. Although the intake appears to change between wild and mariculture, the difference appears to be insignificant (Renzi et al., 2018).

3.3.2 Microplastics characteristics

A total of eight microplastics shapes reported to be ingested by marine bivalves globally (Figure 5). Microfibers (38%) and fragments (31%) had the highest proportion of microplastics types found by marine bivalves globally. Fragment ingestion was found a bit later, in 2015, from a fish market in China (Li et al., 2016). Particles microfibers were then started to be frequently found in the following years. A similar result was also mentioned in a review study by Li et al. (2019), where microfibers and fragments being the most microplastics ingested by mussels. This indicates that microfibers and fragments are common microplastics particle in the marine environment.

The main source of microfibers in the coastal environment is primarily from the washing activity of textiles (de Falco et al., 2019), accounting for around 13 million tons of synthetic fibers entering the ocean annually (Mishra et al., 2019). Microfibers have surely spread acrross the marine environment, including the Northern Ionian Sea, Adriatic Sea, and North Pacific Ocean (Digka et al., 2018; Gomiero et al., 2019). Microfibers have also been documented to contaminate marine sediment near densely populated areas (Browne et al., 2011). Meanwhile, the origin of fragments is normally from the breakdown of larger plastic particle. The degradation and sinking process involves several physical activities such as being exposed to the sun, waves, and wind, through weathering and biofouling processes (Kaiser et al., 2017; ter Halle et al., 2016). These degradation mechanisms also apply to other microplastics types, including foam and film.

In general, marine bivalves globally consume a variety of plastic polymers (Figure 6). Polypropylene (PP) and polyester (PE) are two of the most frequently identified plastic polymer in papers that investigate the polymer types of microplastics ingested by marine bivalves. PP and PE are popular polymers used to create plastic products for customers due to its lightweight material and thermal resistance (thermoplastic) (Maddah, 2016; Plastics Europe, 2022). Several examples of PP and PE plastic products are plastic bags and packaging, synthetic fabric materials, household appliances, and disposable medical appliances. As the recycling rate of general plastic product is low, including in PP and PE, many PP and PE products are discarded and degraded in the environment, especially the sea. A meta-analysis on the distribution plastic polymer by Erni-Cassola et al., (2019) also confirms that PP and PE are amongst the common of plastic polymer types in the marine environment compartment, from the surface to the deep.

This review found that marine bivalves globally have been reported to ingest microplastics in various sizes, with the majority of microplastics being less than 250 µm in size (Table 2, Appendix 2). Different intake on various microplastics sizes by marine bivalves is possibly due to the different selection mechanisms of the bivalves. The shapes of microplastics particles and the size of their mouths are another consideration on microplastics intake by bivalves (Ward et al., 2019). As a sedentary organism, the size and number of microplastics ingested by marine bivalves can help give information on the level of microplastics contamination in its surrounding environment. A study showed that there was a decrease in the number of microplastics ingestion with the increase of distance to the wastewater treatment (Kazour & Amara, 2020).

3.3.3 Limitations around the investigations

Field investigation and monitoring are crucial in understanding the level of contamination of microplastics ingested by bivalves. However, studies used different standard of methods to analyze microplastics. To this day, the protocol uses a digestive method with a different solution and concentration. Some studies agree that a lack of standardization affects analysis results, such as underestimating or overestimating the result and failing to compare the findings with other research in the same region or globally (Bråte et al., 2018; Digka et al., 2018; Hermabessiere et al., 2019; Santana et al., 2016; Webb et al., 2019). A possible reason why a standard method is needed is because some solutions with certain concentration might not be efficient for digesting organic material. For example, Ding et al. (2018) found that high efficiency of digestion by using 10% KOH solutions are more efficient than 30% H_2O_2, while Reguera et al. (2019) found that 10% KOH is more efficient than 65% HNO_3. The current evaluation on the protocol is still testing the common solution being used (Phuong et al., 2018; Thiele et al., 2019). Nevertheless, researchers is still unable to determine which option to use.

Visual identification is also helpful for determining the morphometric of microplastics. Various microscopes are being used on the studies, yet the expertise of the researchers remains questionable, that implies that a professional researcher is required to get a valid identification (Ding et al., 2022; Li et al., 2019). However, this might be not practical at this moment, due to the present global contamination situation. One alternative to help improving the quality of identification is to set up online data base of microplastics which have been discovered around the world. This can be adopted from a marine biodiversity data base, such as matcher.org for manta ray or fishbase.se for fish species catalog.

Altough the majority of the samples were collected in the wild, the accurate information on the origin of the bivalve samples obtained from the market (e.g., de Witte et al., 2014; Mathalon & Hill, 2014; Phuong et al., 2018; van Cauwenberghe & Janssen, 2014) remained unclear. According to the global trend of the marine bivalve production, a large portion of bivalves is coming from

Asia, especially from China, and are being exported all over the world, including to the US, Japan, Hongkong, and Australia (Wijsman et al., 2018), and will be distributed to the fish market. This distribution is expected to cause bias in the sampling site origin for the specimen obtained from the market. Therefore, it is important to track the origin of samples from the market since the concentration of microplastics in the environment is varied from one region to another. As mentioned in the previous discussion, a relatively high ingestion rate occurs in the bivalves sampled near the area with intensive human activity. This kind of information is also related to the amount of microplastics consumed by humans.

Apart from the limitation on the sample origin from the market, two studies proposed an alternative through depuration process, to reduce the concentration of microplastics in bivalves before being consumed by humans (Birnstiel et al., 2019; van Cauwenberghe & Janssen, 2014). Moreover, Kazour and Amara (2020) used a cage technique after the depuration process to monitor the contamination level on the native bivalves. This technique can be a solution to minimize microplastics intake through bivalves and to protect the native bivalves in the wild. Therefore, further field-based experiments are needed to find other solutions on reducing microplastics in seafood.

4. Conclusion

In general, this review shows that marine bivalves globally have been contaminated by microplastic particles from various shapes, polymers, and sizes. This suggests that all species of marine bivalve, whether wild or cultivated, are vulnerable to microplastics contamination. Although some studies have reported that the microplastics intake is higher in maricultural or cultivated areas, the difference is not statistically significant. This review also highlights the promising role of marine bivalves as microplastics biomonitoring. However, it is important to set an international standard protocol for microplastics analysis and microplastics data set to achieve a comparable outcome. Furthermore, the suitable species, size, and sample size to use in the standard protocol must be determined to improve the result validity and comparability.

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