DISTRIBUTION OF MICROPLASTICS IN TROPICAL ESTUARINE MANGROVE AND THE LEACHING OF HEAVY METALS UNDER THE INFLUENCE OF TEMPERATURE AND pH

TAN EVONNE

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by

TAN EVONNE

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LIST OF SYMBOLS

%	Percentage
°C	Degree Celsius
µ	micro
Al	Aluminium
As	Arsenic
Br	Bromine
C	Carbon
Ca	Calcium
Cd	Cadmium
Cr	Chromium
Cu	Copper
H	Hydrogen
Hg	Mercury
L	litre
M	Molarity
N	Normality
Ni	Nickel
O	Oxygen
Pb	Lead
Pd	Palladium
S	Sulphur
Zn	Zinc

LIST OF ABBREVIATIONS

$(CH_3)_2Hg$	Dimethlymercury
µ-FTIR	Micro-Fourier Transform Infrared Spectroscopy
µg	microgram
µL	microlitre
µm	micrometre
ADPC	Ammonium Pyrrolidindithiocarbamate
BP	Balik Pulau
$C_{10}H_{16}$	Adamantane
$C_{22}H_{46}$	Docosane
$C_{22}H_{46}O$	1-Docosanol
C_2H_4	Ethylene
$C_5H_6O_3$	Glutaric Anhydride
$CaCl_2$	Calcium Chloride
CH_3COOH	Acetic Acid
CH_3Hg	Methylmercury
$CHCl_3$	Chloroform
CIR	Coumarone-Indene Resin
cm	centimetre
cm^3	cubic centimetre
CO_2	Carbon Dioxide
DDDC	Diethylammonium Diethyldithiocarbamate
EDCs	Endocrine-disrupting chemicals
EU	European Union
FESEM-EDX	Field Emission Scanning Electron Microscope-Energy Dispersive X-Ray

$FeSO_4$	Iron (II) Sulphate
FIZ	Free Industrial Zone
FTIR-ATR	Fourier Transform Infrared Spectroscopy-Attenuated Total Reflection
g	gram
GPS	Global Positioning System
H_2O_2	Hydrogen Peroxide
HCl	Hydrochloric Acid
HDPE	High Density Polyethylene
HMs	Heavy Metals
HNO_3	Nitric Acid
IBA	Important Bird Area
ICP-MS	Inductively Coupled Plasma-Mass Spectrometry
IPCC	Intergovermental Panel on Climate Change
kg	kilogram
km	kilometre
KM	Kuala Muda
KOH	Potassium Hydroxide
LDPE	Low Density Polyethylene
LLDPE	Linear Low Density Polyethylene
m	Metre
mg	microgram
mL	microlitre
mm	micrometre
MPs	Microplastics
MSO	Mesityl Oxide
NaOH	Sodium Hydroxide

NCBI	National Centre for Biotechnology Information
NH_4Ac	Ammonium Acetate
NH_4OH	Ammonium Hydroxide
NPs	Nanoplastics
OSPW	Oil Sand Process-Affected Water
pcs	pieces
PE	Polyethylene
PEO	Polyethylene Oxide
PET	Polyethylene Terephthalate
PN	Penaga
PP	Polypropylene
PPE	Personal Protective Equipment
ppm	part per million
PS	Polystyrene
PSRP	Penang South Reclamation Project
PVC	Polyvinyl Chloride
ROS	Reactive Oxidative Species
ROS	Reactive Oxidative Species
Sb_2O_3	Antimony Oxide
SE	Standard Error
SP	Seberang Perai
SUPs	Single Used Plastics
TAT-KM	Teluk Air Tawar-Kuala Muda
UNEP	United Nations Environmental Programme
UV	Ultraviolet
UV	Ultraviolet
WWTPs	Waste Water Treatment Plants

$ZnCl_2$	Zinc Chloride
ZnO	Zinc Oxide

TABURAN MICROPLASTIK DI BAKAU MUARA TROPIKA DAN LARUT LESAP LOGAM BERAT BAWAH PENGARUH SUHU DAN pH

ABSTRAK

Kajian ini melaporkan taburan mikroplastik di kawasan bakau Seberang Perai, Kuala Muda, Penaga dan Balik Pulau, Pulau Pinang. Sebanyak 8775 keping mikroplastik telah dikumpulkan dari semua lokasi, dengan taburan di permukaan air pesisir pantai, dasar pesisir pantai dan sedimen kawasan muara yang berada di antara 201±21.214 - 1407±124.265 keping/L, 255±22.368 - 350±25.892 keping/kg dan 430±7.234 - 4000±29.174 keping/kg. Kelimpahan mikroplastik telah menunjukkan pengurangan daripada Seberang Perai (5757 keping) > Penaga (1589 keping) > Kuala Muda (901 keping) > Balik Pulau (528 keping). Morfologi mikroplastik dominan kajian ini adalah fragmen (53%). Sampel mikroplastik ditemui berwarna-warni dengan bentuk dan permukaan yang tidak sekata disebabkan oleh pengaruh cuaca. Polimer yang paling banyak ditemukan adalah polietilena (30%), polistirena (20%) dan poliacetal (20%). Sebatian kimia seperti dimethylmercury berpotensi memudaratkan organisma dan alam sekitar dikesan melalui micro-Fourier Transform Infrared Spectroscopy (μ-FTIR). Kajian ini menunjukkan dasar status pencemaran mikroplastik di kawasan muara Pulau Pinang dan tahap pencemaran adalah dipengaruhi oleh aktiviti manusia. Kajian ini menunjukkan jenis dan morfologi polimer yang dijumpai adalah berdasarkan aktiviti manusia di kawasan terbabit. Kajian mikroplastik jangka panjang adalah digalakkan untuk memudahkan pengurusan pencemaran dan pemuliharaan kawasan bakau pada masa depan. Kajian ini juga mengkaji larut lesap logam berat: aluminium (Al), zink (Zn), kromium (Cr), tembaga (Cu), plumbum (Pb) dan arsenik (As) dibawah pengaruh suhu dan pH dari

pelet polietilena ke dalam air laut. Kebaharuan kajian ini adalah untuk memahami sama ada peningkatan suhu dan penurunan pH air laut akan meningkatkan ketoksikan mikroplastik di bawah cuaca tropika. Kebanyakan logam berat dalam air mempunyai kepekatan tertinggi pada jam ke-24 dan mengalami pengurangan pada jam ke-120 sehingga ke-240, kecuali As yang kebanyakkannya tidak dapat dikesan dalam kajian ini. Al pada pH 7.3±0.05 menunjukkan larut lesap yang konsisten sepanjang kajian, ini menunjukkan keelektronegatifannya boleh mempengaruhi migrasi ke dalam air. Semua logam mencapai kepekatan tertinggi pada pH 7.5±0.05 menunjukkan ia adalah tahap optimum yang berkemungkinan menggalakkan aktiviti biologi dan kemerosotan pelet. Kedua-dua parameter mempunyai pengaruh terhadap tahap degradasi polimer. Tahap penuaan dan pemecahan permukaan pelet adalah lebih serius dalam kondisi yang lebih panas dan air yang lebih berasid. Analisis statistik menunjukkan bahawa suhu dan pH tidak mempunyai pengaruh terhadap larut lesap logam berat dari pelet ($p>0.05$). Walaubagaimanapun, kedua-dua parameter adalah penting dalam menyebabkan degradasi polimer dan seterusnya mendorong proses larut lesap bahan kimia ke dalam air. Kajian sifat logam berkaitan dengan biologi harus dipertimbangkan untuk kajian masa depan bagi menambahbaik pemahaman terhadap ketoksikan mikroplastik di dalam laut.

DISTRIBUTION OF MICROPLASTICS IN TROPICAL ESTUARINE MANGROVE AND THE LEACHING OF HEAVY METALS UNDER THE INFLUENCE OF TEMPERATURE AND pH

ABSTRACT

This study reports the microplastic distribution in Seberang Perai, Kuala Muda, Penaga and Balik Pulau estuaries of Penang. A total of 8775 microplastic pieces were collected, with abundance in coastal surface water, coastal bottom and estuarine sediment ranging between 201±21.214 - 1407±124.265 pcs/L, 255±22.368 - 350±25.892 pcs/kg and 430±7.234 - 4000±29.174 pcs/kg respectively. Overall microplastic abundance decreases from Seberang Perai (5757 pcs) > Penaga (1589 pcs) > Kuala Muda (901 pcs) > Balik Pulau (528 pcs). Fragment was the dominant morphology (53%). Severe surficial weathering was observed on colorful and irregular microplastic samples. Polyethylene (PE) (30%), polystyrene (PS) (20%) and polyacetal (20%) were the most common polymer type. Micro-Fourier Transform Infrared Spectroscopy (μ-FTIR) detected chemical compounds such as dimethylmercury that can be potentially harmful to organisms and the environment. This baseline study presents the fundamental microplastic pollution status of the estuarine environment in Penang, concluding that human activities are significant toward microplastic input. The dominance of polymer morphology and type in study sites is closely related to the nature of human activities. We encourage long-term microplastic studies for pollution management and mangrove conservation in the future. This study also examines the influence of temperature and pH on heavy metals: aluminum (Al), zinc (Zn), chromium (Cr), copper (Cu), lead (Pb) and arsenic (As) leaching from transparent PE pellets into seawater. The novelty of this study is

to understand whether rising water temperature and decreasing pH of the ocean would enhance microplastic toxicity under tropical weather. Most HMs concentration in water peaked by 24 hours and decreased by 120 hours until 240 hours, except As (mostly non-detectable). Al at pH 7.3±0.05 shows consistent leaching throughout the study, suggesting its electronegativity may influence its migration into water. HMs concentration peaked at pH 7.5±0.05 for all metals, indicating possible optimal levels for biological activities and promoting degradation of PE pellets. Both parameters have an influence on polymer degradation with pellets in warmer, acidic waters experiencing a greater extent of surface aging and breakdown. Statistical analysis revealed that temperature and pH do not influence HMs leaching from PE pellets ($p>0.05$). Although temperature and pH may not have a direct influence on metal leaching from PE pellets, they are crucial in inducing polymer degradation and paving the way towards chemical leaching. Consideration of metal properties and microbial studies should be included in future studies to improve our understanding of microplastic toxicity in the ocean.

CHAPTER 1

INTRODUCTION

1.1 Overview of Microplastics Pollution

In today's world, plastic can be found everywhere in the ocean because of improper waste management on land that brings them into nearby water bodies (Hammer et al., 2012). Plastic pollution is borderless as debris can be washed on and offshore to and from different countries, travelling throughout different water bodies, and even carrying to remote locations such as the poles by water and wind action (Klien et al., 2018). Plastics are widely used because they are cheap, light-weight, highly durable that brings huge convenience in everyday life (Zhou et al., 2022). High consumption of non-reusable single-used plastic (SUPs) is the major contributor to plastic pollution because of their minimal lifespan, with 40% of plastic production coming from them (Chen et al., 2021a). Plastic pollution has become more severe since the COVID-19 pandemic with high consumption of single-used PPE and mask globally. Face mask has been estimated to contribute at least 40000 kg of plastics into the ocean monthly (10 million pieces of 4 g face mask) (Ricciardi et al., 2021). Marine organisms ingesting plastic debris are often reported to suffer from physical entanglement, drowning, blockage of air and food pathway, ultimately leading to death (GESAMP, 2015).

Exposure of larger plastic to sunlight, mechanical, biological and chemical degrading mechanism increases the formation of microplastics (MPs) (GESAMP, 2015). MPs are small, fragmented polymer with size ranging from 1 μm to 5 mm (GESAMP, 2015). There are two types of MPs based on their origin: primary and secondary MPs. Primary MPs are commercially produced standard shaped plastics while secondary MPs are irregular pieces unintentionally formed from the

degradation of large plastic debris into small fragments (Zhang, 2017). Meanwhile, there are five morphological classifications of MPs as suggested by GESAMP (2019): foam, film, fragment, fibre and pellet. Increasing presence of MPs in the aquatic environment is expected to rise concurrently with the input of plastic waste because a single piece of large plastic may produce up to a thousand pieces of MPs (Gimiliani et al., 2021). High abundance of MPs recovered in water bodies near to land implies that terrestrial input is significant to the environment (Luo et al., 2018). Compared to larger plastics which can be collected through clean-up activities or deploying vessels, MPs are nearly impossible to recover once it enters into the aquatic environment because of its small size (Klien et al., 2018). The density of MPs influences their occupancy in different compartment of a water body, in turn affecting their distribution vertically and horizontally (Zhang, 2017). Low density MPs tends to float on surface waters or stay suspended in water columns while high density MPs often settles to the bottom of the ocean floor (Kumar et al., 2021). When MPs floats on surface water, they can travel longer distance by following surface current while particles that settle in the bottom sediment stays relatively static unless particle buoyancy changes which then allows them to resuspend into water column or float on water surfaces (GESAMP, 2015). The discovery of MPs in aquatic organisms across different trophic levels is a huge concern to human health as they are not biodegradable and may accumulate in the body if human accidentally consume contaminated sea-based food and beverages (Ricciardi et al., 2021). Nevertheless, research on the effects of MPs towards human health remains unclear, unlike the understanding towards aquatic animals and plants (Campanale et al., 2020).

The proximity of estuaries that sits between the land and ocean makes it highly vulnerable to pollution contamination, including MPs (Govender et al., 2020). Estuaries can accumulate MPs from direct terrestrial input, riverine flow and back washing of coastal current that carries them into the estuaries (Ricciardi et al., 2021). Riverine flow is considered as one of the most important MPs sources carried from land (Li et al., 2021). In fact, the hydrodynamics of estuaries encourage accumulation of larger and heavier MPs which makes them a potential sink for MPs (Defontaine et al., 2020). For example, the presence of finer sediment grains and muddy condition of estuaries can accumulate more MPs as they have strong cohesive forces (Furukawa & Wolanski, 1996, Lamichhane et al., 2021). Moreover, mangrove-dominated estuaries with numerous protruding roots that holds sediment and reduce coastal erosion would help build up MPs abundance as their roots can help filter out MPs from river input before entering nearby coastal waters (Govender et al., 2020). The authors also found out that the abundance of pneumatophores in estuaries influences the macro- and microplastic abundance as the roots can effectively trap larger plastic pieces and retain them for a longer period of them to cause fragmentation, forming MPs. Mangrove-dominated estuaries are important nurseries and habitats for numerous terrestrial and estuarine organisms (Bujang & Zakaria, 2020). Due to their high productivity and uniqueness of fresh-saltwater mixture, they also support a wide variety of fish species and serve as fishing grounds for fishery communities (Omar & Misman, 2020). Thus, MPs invasion into estuarine environment is harmful to both animals and humans as accidental ingestion may affect their development and health (Talbot et al., 2020).

Flora and fauna living in mangrove forest in Malaysia is highly susceptible to metal and plastic pollution because they receive significant amount of terrestrial and

oceanic waste that contains MPs and heavy metals (HMs) (Tan et al., 2021). Mangrove forests around the country have been extensively converted for urbanization and aquaculture activities (Hashim & Shahruzzaman, 2017). In Peninsular Malaysia, the mangrove ecosystem located along the west coast is subjected to pollution especially oil spill due to the congested traffic along Straits of Malacca (Rusli et al., 2019). Accidental oil leaking and spill from vessels will unavoidably contaminate surrounding water and sediment with hydrocarbons and HMs (Rashidi & Maimun, 2013; Zhang et al., 2019a). Mangrove forests in Penang also have been largely converted for development since 1972 (Foong et al., 2020). As of 2017, approximately 1050 hectares of mangrove forests remain in the state with only 35% of the area is protected under law (Foong et al., 2020). Natural services that mangrove ecosystem provided cannot be monetised, thus they are often overlooked for conservation purposes (Zakaria & Sharma, 2020). In fact, keeping up to update information on precise numbers and tracing of mangrove area and deforestation status in the country is deemed a challenging task (Zakaria & Sharma, 2020).

As our current environment is experiencing climate change like global warning and ocean acidification, it is crucial to study their consequences towards the environment and plastic pollution. Temperature and pH are important abiotic factors in polymer degradation (Wang et al., 2019). Polymer undergoes thermal degradation when temperature is sufficiently high to reduce intermolecular polymer bond which can ultimately change its structure (Izdebska, 2016). If biodegradation is involved, temperature is crucial in facilitating biological and enzymatic reaction where rate can accelerate by double with an increase of 10°C (Ball et al., 2011; Yao et al., 2022). Lei et al. (2020) added that acidic solution can act as oxidizing agent that initiate

chemical reaction and result in the breaking down of carbon bonds in the polymer chain. When polymer is placed in acidic medium, the presence of high concentration of free protons (H^+) will induce hydrolysis where chemical reaction occurs between polymer bond and H^+ (Baiti et al., 2015). A study also confirms that water pH influences weathering and aging of MPs where they found more profound aging and weathering on the MPs exposed to acidic water (pH 5) compared to pH 7 (Miranda et al., 2021). When polymer bond weakens and breaks, additives migration into ambient environment occurs which may be harmful to organisms and deteriorates water quality (Izdebska, 2016; Campanale et al., 2020). In the real world, both degrading agents existing as global warming and ocean acidification are also expected to become more severe as more carbon dioxide are anthropogenically entering the environment (Masson-Delmotte et al., 2021).

There is a lack of work and understanding on the status of mangrove forests in the country, let alone its resilience towards climate change (George, 2019). For example, there are two mangrove species in Malaysia classified by IUCN Red List as critically endangered, *Brugueira hainesii* and *Sonneratia griffithii*. *Brugueira* sp.. The former species was reported by Ono et al. (2016) in three ungazetted mangrove forests (Merbok, Klang and Kukup) while there is a lack of knowledge on the later species according to MyBIS (2022). This indicates that more studies need to be carried out on the mangrove forests around the country to help conserve mangrove biodiversity before losing them.

1.2 Statement of Problem

Our study is driven by the emerging reports of MPs in different compartments of estuarine environment in the country. All study sites have been

reported to provide ecological benefits and services to respective communities and organisms. Estuarine environment in Penang harbours significant mangrove trees species and marine birds that requires conservation and protection (MNS, 2015; Foong et al., 2016). There is high reliance of local communities towards the estuarine environment as the fishery activities are important to provide source of income and supply seafood to nearby states. Therefore, four study sites located around Penang with ecological and community importance was selected namely Seberang Perai, Teluk Air Tawar-Kuala Muda (TAT-KM) and Balik Pulau. Specifically, TAT-KM was recently considered to be proposed as a Ramsar Site for conservation purposes (Aziz, 2022). Yet, its pollution status has not been evaluated and remains unknown. A lack of supporting data and understanding on the pollution status of these study sites may hamper conservation and preservation effort in the near future.

The vulnerability of estuarine mangroves towards pollution is also one of the concerns of our study. These areas are facing threats from conversion for human settlement, industrial, aquaculture, agriculture activities, coastal reclamation and unsustainable tourism that would release pollutants into the water (Foong et al., 2016). The estuaries are experiencing busy shipping traffic and fishing boat activities, particularly in Seberang Perai that is near to the Perai Free Industrial Zone (FIZ). On top of that, the mangroves are located near highly populated settlements and industrial sites along Sungai Perai (Yii et al., 2020). Meanwhile in mangroves estuaries of Kuala Muda, gears used for fishing industry may release synthetic polymers such as fragments and fibres if stay submerged in water for a long time. Wright et al. (2021) outlined that fishing gears related MPs reflects the intensity of fishing activities in an area. These activities may contribute to the increase of MPs

abundance in the water and threaten public health as fish catch from this area are supplied to local markets.

As global warming and ocean acidification is expected to become much more critical in the future (Masson-Delmotte et al., 2021), our concern is the ecotoxicity of polymer degradation in warmer and acidic waters. The focus point is whether the process of heavy metals (HMs) leaching from polymers will be accelerated or decelerated under the influence of elevated temperature and reduced acidity. Although there are studies done to understand polymer degradation and MP toxicity under various conditions, nevertheless they are conducted under extreme conditions that are somewhat irrelevant to current environment conditions (Koelsman et al., 2022; Li & Tang, 2022). In fact, plastic degradation and toxicity under the influence of tropical weather remains relatively unexplored. Till today, it remains challenging to draw a definite conclusion as MPs are subjected to numerous degrading factors in the marine environment (Zhang et al., 2019b). Furthermore, the abundance of additives present in plastics is problematic as they are known to be a source of xenobiotics themselves (Verla et al., 2019). When polymers degrade, additives may leach into the waters (Zhang et al., 2019b). The presence of HMs and its leaching into the environment remains a threat to organisms as it does not metabolise and may gradually accumulates in the body (Munier & Bendell, 2018).

In addition, research on the influence of climate change towards mangrove trees globally remains below on par (Walden, 2019). Malaysia is lacking in understanding and research to conserve mangrove with current and future climate change despite having one of the largest mangrove coverages in the world (ADB, 2014; George, 2019). To our best knowledge, current studies conducted in Malaysia

on climate change impact associated with mangrove forests is limited to only sea level rise (Sarkar et al., 2014; Ehsan et al., 2019).

1.3 Statement of Purposes

Our study aims to provide a basic understanding on the abundance of MPs present in mangrove estuaries around Penang and assess the pollution extent. We are interested to see whether the influence of anthropogenic activities convinces with the severity of MP pollution of each study site. Our study also aims to provide meaningful data that can be used for pollution management and control. We aim to narrow the knowledge gap on MP pollution in Malaysia that will encourage mitigation efforts in the near future.

Our study also aims to understand the influence of temperature and pH towards HMs leaching from polymer under tropical weathers. Our interest is to observe properties change of polymer while enacting the current marine environment condition. In a bigger picture, we want to find out the possible effect of global warming and ocean acidification towards the toxicity of MPs. Although this study is only 10 days, it has been validated by European Union where chemical migration from plastic within this time frame can cause health problems (European Union, 2016). Our study believes that short-term study is the preliminary step to establish theoretical which can prompt similar environmentally relevant long-term study in the future. Therefore, our study sees the necessity of exploring the impact of ocean acidification and warming water temperatures towards chemical leaching from MPs.

1.4 Significance

This study is significant to the environment as quantifying MP abundance in the mangrove estuaries would provide an insight on the pollution status of the study

sites. This will help us to understand the influence of nearby anthropogenic activities towards the severity of MP pollution. More data will be available to suggest conservation and preservation effort to protect the ecosystem and maintain its environmental quality. This study is important for future pollution studies on MPs to contribute in data comparison.

It is also important to grasp the influence of tropical weathering towards polymer degradation and toxicity to improve our knowledge towards MPs exposure in the environment. In our study, the understanding of the toxicology of MPs while focusing specific environmental degrading factor will provide a deeper understanding the way how polymers will be affected. Additionally, we consider the extensive production and usage of polyethylene (PE) in the market and hence incorporating this type polymer in this study will help us to understand how majority MPs in the ocean degrades.

1.5 Scope of Study

The scope of study for MP study involved sample collection from coastal surface waters, coastal bottom sediment and estuarine sediment from four estuarine mangrove in Penang. All samples are used to assess and compare its abundance between each study sites, followed by physical characterization and chemical analysis of representative MPs samples. Samples were classified based on five morphologies as suggested by GESAMP (2019).

The study of HMs leaching from polyethylene pellets was limited at 10 days and includes six metal species in water which is aluminium (Al), zinc (Zn), lead (Pb), copper (Cu), chromium (Cr) and arsenic (As). The pellets are added into filtered seawater and exposure towards to outdoor environmental condition are maximised.

Measurements and adjustments of temperature and pH were carried out daily and water samples were collected on the 0, 24, 120 and 240 hours for analysis. Physical characterization of pellets was also carried out upon sample collection on 240 hours.

1.6 Unique Features

The first part of our study will be the first work to assess MP pollution in tropical mangrove estuaries of Penang, where no work has been done to date. The study sites were previously reported to have high diversity of mangrove tree species, compared to well-conserved Matang Mangrove Forest and mangrove park in Klang, despite located around highly urbanized area and ungazetted (Stiepani et al., 2021). In the second part of the study, realistic environment condition under tropical weather is applied to study MP toxicity in terms of HMs leaching. This study highlights the condition of tropical weather and its influence towards to polymer degradation and toxicity which have yet to be explored. This study also features two current environmental problems: global warming and ocean acidification where we will study its influencing effect towards HMs leaching from the polymer.

1.7 Objectives

Therefore, the objectives of this study are:

a) To determine the abundance of microplastics in coastal surface water, coastal bottom sediment and estuarine sediment of tropical estuarine mangrove in Penang.

b) To assess the physical changes and chemical composition of microplastics collected from coastal surface water, coastal bottom sediment and estuarine sediment of tropical estuarine mangrove in Penang.

c) To determine the influence of temperature and pH towards leaching of heavy metals (Al, Zn, Pb, Cu, Cr and As) from polyethylene microplastic.

CHAPTER 2

LITERATURE REVIEW

2.1 Microplastics (MPs)

Plastics are synthetic polymers comprising of inorganic (carbon, hydrogen, oxygen, nitrogen) and organic compounds (fossil fuels), formed from long chains of covalently bonded monomers (Pathak & Navneet, 2017). They have wide tolerable range towards temperature and pressure, inert, cheap and easily altered which makes them ideal to manufacture daily use items such as grocery bags, bottles and pails (Chia et al., 2020). About 50% global plastics production are single-used plastics (SUPs) while only 25% of them are manufactured long termed used (Kedzierski et al., 2020).

Microplastics (MPs) are small, fragmented plastic pieces with size ranging usually between 1µm-5mm (Fang et al., 2018). In natural environment, large plastic pieces undergo a series of physical weathering and chemical degradation processes to form MPs (Isaac & Kandasubramanian, 2021). Being hydrophobic and persistent, MPs will accumulate in the environment thus increasing its bioavailability towards organisms (Zantis et al., 2021). MPs is so abundant in the marine environment where it contributes at least 90% of plastic debris recovered from the Great Pacific Garbage Patch (Chen et al., 2019). MPs can accumulate even quicker in sediments and water bodies if hydrodynamics are slow or static (Padervand et al., 2020). At least 15 million pieces of MPs has entered the ocean since 2014, with expectation that concentration will increase tenth-fold by 2025 (Axworthy & Padilla-Gamino, 2019). MPs can be categorized into two types, primary and secondary MPs, as shown in Table 2.1 (Malankowska et al., 2021).

Table 2.1 Two main categories of microplastics (MPs).

Primary MPs	Secondary MPs
They are purposely made into small particles and notably used in personal care products such as facial scrubs and toothpaste, commercial plastic pellets.	Small plastic fragments from degradation of large piece of plastic influences by surrounding biological, chemical and physical factors.
In Europe, personal care products are the most significant source of primary microplastics.	Plastics can be degradation though abiotic and biotic processes in the ocean.

2.1.1 Degradation

Polymer degradation begins with physical weathering (wear and tear) of outer layer of the plastic and slowly extend into inner matrix where chemical bond weakens, breakdown and eventually mineralized by microorganism for food (Webb et al., 2013; Yousif & Haddad, 2013; Montesinos, 2018). Throughout degradation processes, different functional groups such as carbonyl, hydroxyl, carboxyl or ketone can be formed when the amorphous region of the polymer matrix is being attacked (Venkatachalam et al., 2012). The present of these functional groups will further promote polymer degradation because of its instability nature that tends to decompose under UV exposure (Montesinos, 2018).

Plastics rarely degrades 100% because small portion will be consumed by organisms and converted into natural products such as biomass (Shah et al., 2008). Each polymers have different susceptibility and sensitivity towards degradation mechanisms such as light, heat or chemical (Kulkarni & Dasari, 2018). For instance, polyethylene (PE) and polypropylene (PP) are the least sensitive polymer towards oxidative degradation (Jansen, 2015). Polymer degradation can occur abiotically and

biotically, where the latter comes after the former (Chamas et al., 2020). However, in terms of reaction rate, biotic degradation has been proven in laboratory conditions to occur faster than abiotic degradation (Min et al., 2020).

In the marine environment, the entire degradation process of plastic is relatively slow where at least 50 years is needed to fully degrade and incorporate into the environment (Webb et al., 2013). This is because factors that drives degradation process fluctuates and varies in different part of the ocean around the world (i.e., photodegradation is affected by sunlight intensity while oxidative degradation is influenced by the presence and concentration of oxygen in surrounding environment) (Webb et al., 2013). Although temperature is involved in degradation process, the presence and intensity of oxygen and sunlight is the most important factor to instigate abiotic degradation because the temperature of ocean surface or water column is not sufficient enough (Gewert et al., 2015). Thus, plastics occurring in bottom sediment would degrades extremely slow due to lower temperature and less sunlight radiation; those floating at water surface may degrade rapidly under sufficient oxygen and UV radiation (Andrary et al., 2018; Ronkay et al., 2021).

2.1.2 Source

MPs can enter the marine environment through three pathways: terrestrial, oceanic and atmospheric input (Wang et al., 2020). Terrestrial source of MPs may come from urbanization areas with intense human activities, industrial effluents, waste water treatment plant (WWTPs), household sewage, accidental runoffs from landfills due to improper waste management, careless littering, usage of personal care and medical products with microbeads (Du et al., 2020). Oceanic source may include paints and coating chipped from vessels, maritime instruments, sanitary waste released from cruises and vessels (Wang et al., 2020). Fishing gears and nets used in

aquaculture activities that regularly stay suspended in water surface or columns may also degrade to release microfibres (Kane & Clare, 2019). Seaports wastewater with plastic scrubbers that are used to remove rust from vessels may also contributes to MPs pollution into waterbodies (Beaman et al., 2016). Atmospheric input involving long-range transport of MPs into marine environment is an emerging source and concern as MPs has been found in deep sea sediment and invertebrates living in remote regions of the world such as Arctic and sub-Arctic (Wang et al., 2020).

2.1.3 Distribution

The distribution and transport of MPs in the marine realm is poorly understood due to complex ocean processes, coexistence of multiple degrading agents and the reaction-specific of polymers towards degradation (Lusher, 2015). Coastal waters and estuaries are reported to be most vulnerable towards MPs pollution due to its close proximity to land and river that directly receives terrestrial and river input respectively (Du et al., 2020). Rivers carrying MPs resembling the density and shape of sediments from land to the ocean relies on its water flow into nearby larger water bodies (Beaman et al., 2016; Kane & Clare, 2019; Padervand et al., 2020).

Microfibres have been reported to be the dominating MPs morphology in the marine environment because it was estimated that 1900 pieces of microfibres can be released from a single wash of one piece of clothing and current filtering technology in WWTPs does not have the ability to completely remove them off waste stream before being released into the environment (Browne, 2015; Beaman et al., 2016; Kane & Clare, 2019). MPs can be found suspended in water column or sink to sediment surfaces depending on their relative density to seawater (Anderson et al., 2016). MPs with lighter density tends to travel a greater distance compared to heavier ones because its easiness to be carried further by water and wind (Beaman et al., 2016).

Padervand et al. (2020) added that deliberated round shape MPs usually stay afloat on water surfaces while those naturally broken with irregular shapes and sizes usually stay suspended in water columns or sink to the bottom sediment.

There has also been emerging theory that deep sea regions and sediments such as submarine canyons and trenches contains more MPs that shallow coastal water despite receiving direct terrestrial input because as MPs sink passively to bottom of the ocean, it can be further carried into deeper parts of the ocean by deep water currents and accumulate because hydrodynamic are relatively slower than surface currents (Kane & Clare, 2019).

2.1.4 Problems

MPs concentration in marine environment has increases steadily over the years because of uncontrollable input of plastic debris that degrades into smaller pieces, hindering efforts to collected physically (Chen et al., 2021a). According to Deng et al. (2021), plastic debris contributes to about 80-85% of marine waste and United Nations Environmental Program (UNEP) has classified them as emerging environmental problem. Although plastic can tolerate a wide range of temperature, they are however defenseless towards degradation (Liang et al., 2021). The tendency for organisms to consume MPs varies on the physical attraction of particles (shape, size and color) and its availability in surrounding environment (Anderson et al., 2016).

Plastic waste is a huge environmental concern to the marine environment because of the threats and damage they possess (Secretariat of the CBD, 2016). Organism survival can be highly threatened because they can be easily ingested and move internally in the body (Chen et al., 2019). MPs is assumed to exert three different toxicological effects to marine organism: a) physical stress: blockage in important pathway, egestion (additional energy required), pseudofeces b) chemical

leaching from plastic c) pollutant carrier (Wright et al., 2013). To make matter worst, MPs or the formation of nanoplastics (NPs) are practically impossible to recover from the environment and can easily invade different compartments of the environment, especially the food chain (Secretariat of the CBD, 2016). Plastic persistency is an additional concern because they can act as a vector that absorbs and transport surrounding pollutants at the same time releasing toxic chemicals (Secretariat of the CBD, 2016). Till today, the duration for complete breakdown of plastic waste in the environment remains unresolved and it is speculated hundred to thousands of years is required (Wayman & Niemann, 2021).

Although recycling has been aggressively promoted as compared to reduce and reuse practice, this method is however economically not viable as it requires state-of-art technology and limits the usage of recycled plastic (Kedzierski et al., 2020). As mixing of different polymer type has been the most preferable practice in recycling that reduces processing cost and facilities, unavoidable degradation and mixture of additives from virgin plastic waste reduces its quality, market value and functionality (Azeez, 2019). In addition, plastic waste recovered from marine environment is an impractical recycling source because it is an arduous process from collection to processing compared to terrestrial source and it might be highly porous due to biofouling and long immersion in saltwater (Ronkay et al., 2021). With the emergence of COVID-19 pandemic in the 21st century, the fear of contamination and health risk at a global scale has dramatically increase the usage of single used medical devices and resources such as face mask and gloves (Schyns & Shaver, 2021). These further increases environmental pressure to extract more natural resources for production as they cannot be reused nor recycled (Schyns & Shaver, 2021). Whilst recycling is costly and merely supplying 10% of global plastic demand, it is nevertheless

important to explore for more constructive solution because less raw materials will be required by the market which implies lesser GHGs emission into the atmosphere (d'Ambrieres, 2019).

2.1.5 Plastic and Microplastics (MPs) Pollution in Malaysia

Plastic pollution came into the limelight of Malaysia when it was ranked 8th plastic contributor globally into the ocean (Ibrahim & Noordin, 2020). Various societal level and stakeholders contribute to the gravity of this environmental problem. Internationally, China's ban to stop importing contaminated plastic waste from 2018 caused redirection of developed nations towards Southeast Asian countries as a new dumpsite, including Malaysia (Wong & Jasmin, 2019). The lack of proper recycling capacities, facilities and strict enforcement resulted in overwhelming flow of plastic waste into the country (Ibrahim & Noordin, 2020).

Divided opinions at governmental level also contributes to the gravity of plastic pollution in Malaysia. Different ministries have different prioritization on the country's economy and environment (Ananthalakshmi & Chow, 2018). As a developing country, the recycling industry may potentially transform Malaysia's economic growth but the people's health will be jeopardised without a clean environment (Tan et al., 2021).

Due to its convenience and affordability, plastic products are the ultimate choice in daily use especially food delivery and packaging (Kumar et al., 2020). Emerging plastic capitalism from consumer to market creates chain reaction from thriving plastic production, unregulated usage to accumulating waste domestically (Tan et al., 2021). At an individual level, materialism culture coupled with poor environmental awareness among Malaysian also contributes to the severity of plastic pollution with accelerating plastic consumption (MESTECC, 2018).

Fortunately, the country is currently taking proactive measures. Malaysia's Roadmap Towards Zero Single-Used Plastics launched in 2018 (Tan et al., 2021). Large retailer stores and restaurants are discouraged to provide plastic bag and a charge will be implemented upon request (Hashim et al., 2019). Plastic straw bans are also implemented in several states (MESTECC, 2018). Nevertheless, the campaign is not looking effective as the public are still willingly to pay for the bags and seen as an indispensable part of the society (Tan et al., 2021).

2.1.6 Microplastics (MPs) Studies in Malaysia

Table 2.2 summarizes microplastic (MP) research of different marine compartments (sediment and water) in Malaysia relevant to this study. All MP research demonstrates stepwise analysis such as quantitative method using different types of microscopes and qualitative method involving budget friendly and/or high-end expensive instruments. From sampling method, we observed that most study uses quadrant for sediment collection. Meanwhile, water samples were collected using a variety of sampler which depends on individual researchers. Water sampler were observed to be commonly preferred by most studies as it may reduce biased sampling in terms of MP size (Cutroneo et al., 2020). This is because nets with specific mesh sizes that can selectively filter out smaller MPs (Cutroneo et al., 2020). However, this may be occasionally beneficial for study that focuses on study specific MPs size range (Razeghi et al., 2021). Sometimes, net towing at sites with high suspended particulate matter contamination may hinder sample collection as they may accumulate in the net (Razeghi et al., 2021). Net sampling is more effective in covering larger study area which can provide better picture on the contamination level of a specific region (Razeghi et al., 2021). Ultimately, the choice of sampling method depends on the

nature and objective of a study whether to cover large area or to study in detail the type of contamination presents in the water body.

Airborne contamination may occur when samples are left open throughout the studies (Lusher et al., 2018). Wet blank filter paper has been suggested to place alongside samples during analysis to enhance result accuracy (Lusher et al., 2018).

In sediment samples, high amount of organic matter may be present which requires removal before density separation. As such, suitable selection of reagent for digestion is necessary to effectively remove it while preserving samples as much as possible (Lusher et al., 2018). Based on Table 2.2, wet peroxide oxidation using hydrogen peroxide (H_2O_2) is commonly used. This method was reported to optimally digest organic matter while not damaging samples, compare to strong acidic or alkali solution such as HCl, HNO_3 and KOH that may even dissolve MPs (Lusher et al., 2018).

According to Ali et al. (2021), the density of polymers ranges between 0.8-1.4 g/cm^3. Density separation is a crucial analytical step to extract MPs from sample. By mixing high-density solution with samples, MPs will float up to solution surface and collect on filter paper via filtering (Cutroneo et al., 2021). Sodium chloride (NaCl) with a density of 1.6 g/cm^3 is the most commonly choice because of its affordability and environmentally friendly (Lusher et al., 2018). Although higher density solution is also used such as zinc chloride ($ZnCl_2$) or calcium chloride ($CaCl_2$) they are more expensive and possess more health hazard (Lusher et al., 2018). As polymer density range is gradually increasing, double extraction has been recommended when using lower density solution to reduce under-extraction (Lusher et al., 2018).

Quantitative analysis in MP research includes determining the abundance of MPs collected in a study. Based on Table 2.2, microscopes attached with digital

camera is primarily used to calculate abundance, measure size and taking photos of the samples. Despite being cost-effective, overestimation or underestimation of MP count may occur due to human error (Lusher et al., 2018). Thus, observer is required to be well-equipped on MP morphology and classification to reduce error as much as possible during analysis (Lusher et al., 2018). Hot needle test was also used in several studies mentioned in Table 2.2. This is an inexpensive and rapid method to differentiate plastic and non-plastic from samples (Cutroneo et al., 2020). But they are destructive and highly rely on melting temperature of tested pieces to show verify their plasticity; hence they are not well-preferred in studies (Cutroneo et al., 2020).

Qualitative analysis in MP research includes the studying of physical characteristics and chemical composition of the samples. Field Emission Scanning Electron Microscope (FESEM) is an expensive method commonly used to observe physical characteristics of MPs (e.g., surficial texture) (Mariano et al., 2021). Although it is destructive and requires knowledgeable individuals to operate the instrument such as pre-analysis sample coating, it is useful in obtaining high resolution images of sample surface to study the wear-and-tear condition (Mariano et al., 2021; Huang et al., 2023). FESEM equipped with EDX is useful to detect inorganic compounds in the polymer samples which can contribute in chemical composition study (Girão, 2020).

Fourier Transform Infrared Spectrometry (FTIR) is a non-destructive and highly accurate instrument where it uses fingerprint method to detect polymer compounds by comparing generated spectrum against library spectrum (Mariano et al., 2021). It is the most preferred choice in plastic identification as observed from Table 2.2 as it can detect MPs as small as 10 μm (Cutroneo et al., 2020). But FTIR can be expensive, occasionally time-consuming when there is large amount of sample

on a filter paper upon collection (Mariano et al., 2021). Pyrolysis GC-MS has been recommended as a much more promising method to analyse chemical composition of very small samples (< 10µm) (Ribeiro et al., 2020). Unlike FTIR, this method is destructive where samples are decomposed under high temperature and end-product are analysed using mass spectrometry (Huang et al., 2023). However, identification may become tricky because different polymer may have the same end-product as it does not have a standard reference (or library) for comparison (Huang et al., 2023). Ibrahim et al. (2021) is the only study to use this method based on Table 2.2.

Another limitation observed throughout all studies is the absence of standardized units in reporting the abundance, which is also globally acknowledge (Hamid et al., 2018; Tibbettes et al., 2018). This would hinder future data comparison for MP studies, where the pollution status around the world cannot be comprehend inclusively (Hamid et al., 2018). Whilst self-conversion of units has been suggested and used to overcome this shortcoming, it is however deemed inaccurate as conversion may under and/or overestimate initial reported abundance (Hamid et al., 2018).

There is no ultimate guideline or methodology for MPs research. The choice of sampler and analytical method depends greatly on the suitability of studied compartment, instrument availability and cost effectiveness. Each stepwise analysis is important to help compensate each other's limitation. Careful time management and work designing is crucial to maximise analysis outcome and achieve desirable of the study.

Table 2.2 summarises microplastic research of different marine compartments (sediment and water) in Malaysia relevant to this study.

Table 2.2 Comparison of microplastic studies in Malaysia based on location, study medium, sampling and analysis method and expressing unit.

Author(s) & Year	Location	Studied Compartment	Sampling Method/Instrument	Digestion/Extraction Method	Unit expressed
Barasarathi et al. (2014)	Sementa Mangrove, Selangor	Sediment	Quadrant	• Saline solution (unspecified)	Number of particles
Estim & Sudirman (2017)	Sebatik Island, Sabah	Sediment	Quadrant	• Sodium Chloride (NaCl) • Stereomicroscope	Number of particles/mL
Sarijan et al. (2018)	• Skudai river, Johor • Tebrau river, Johor	Sediment	Box corer	• Hydrogen Peroxide • Sodium Hydroxide Stereomicroscope	Number of particles/kg (dry weight)
Bitlus et al. (2020)	Sri Tujuh Beach, Kelantan	Sediment	Ekman grab	• NaCl • Microscope • Fourier Transform Infrared Spectrometry-Attenuated Total Reflection (FTIR-ATR)	Number of particles
Hamid et al. (2020)	Carey Island, Selangor	Sediment	Quadrant	• NaCl + Tween-80 • Optical microscope	Number of particles/kg (dry sediment)
Lim et al. (2020)	• Pulau Betong, Penang • Pasir Panjang, Penang • Teluk Aling, Penang	Sediment	Quadrant	• Calcium Chloride (CaCl2) • Stereomicroscope	Number of particles/ m^2

Table 2.2 Continue.

Author(s) & Year	Location	Studied Compartment	Sampling Method/Instrument	Digestion/Extraction Method	Unit expressed
Amin & Azman (2022)	Sungai Laloh, Johor	Sediment	Ekman dredge sampler	• NaCl • Hydrogen Peroxide (H_2O_2) • Stereomicroscope • FTIR-ATR	Percentage abundance (%)
Fauziah et al. (2015)	• Port Dickson, Negeri Sembilan • Kuala Terengganu, Terengganu • Kota Kinabalu, Sabah	Beach sand	Quadrant	• Seawater + Freshwater • Dry + wet sorting	Number of items/ m^2
Noik & Tuah (2015)	• Santubong, Sarawak • Trombol, Sarawak	Beach sand	Quadrant	• Hydrochloric Acid (HCl) + Nitric Acid (HNO_3) • NaCl • Compound stereoscope • Compound microscope • FTIR	Weight (g)
Lee et al. (2020)	• Kuala Perlis, Perlis • Kurung Tengar, Perlis • Pantai Miami, • Pantai Teluk Bahang, Penang • Pantai Pasir Panjang, Penang	Beach sand	Quadrant	• Optical Microscope • Hot Needle Test	Number of particles/ m^3