DETECTION OF MICROPLASTICS IN TAP WATER, KAMPAR PERAK

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DETECTION OF MICROPLASTICS IN TAP WATER, KAMPAR PERAK

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A project report submitted in partial fulfillment of the requirements for the award of Bachelor of Engineering (Hons) Environmental Engineering

> Faculty of Engineering and Green Technology Universiti Tunku Abdul Rahman

> > January 2022

DECLARATION

I hereby declare that this project report is based on my original work except for citations and quotations which have been duly acknowledged. I also declare that it has not been previously and concurrently submitted for any other degree or award at UTAR or other institutions

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Specially dedicated to my beloved family and friends

ACKNOWLEDGEMENTS

Throughout the whole project, I have received a great deal of kind assistance and endless support.

Firstly, I would first like to thank my supervisor, Prof. Dr. Sumathi a/p Sethupathi for her valuable advice, patience, and guidance for the successful completion of my project. Her knowledge, experience and expertise had brought my work to a higher level.

In addition, I would also like to express my gratitude to my beloved family, friends and seniors for their endless support and encouragement. I would like to acknowledge also the laboratory staff, Ms Amelia, Mr Yong, Puan Nisha for their assistance and technical support during the laboratory work.

Furthermore, I would like to appreciate Universiti Tunku Abdul Rahman (UTAR) for providing an education platform for me to conduct a research project.

Last but not least, I would like to acknowledge Lembaga Air Perak Water Treatment Plant and their friendly staffs, Mr Megan and Mr Saiful that providing kind assistance during my sample collection procedure.

DETECTION OF MICROPLASTICS IN TAP WATER, KAMPAR PERAK

ABSTRACT

Microplastic (MPs) have become a trendy environmental concern in recent decades because of their ubiquity in marine, freshwater, soil, and even human food chain and drinking water. However, the MPs research in Malaysia mainly focuses on environmental contexts and no research was reported on drinking water. This study focused on quantification and characterization of MPs in tap water, precisely in Kampar distinct of Perak State. Tap water samples were collected from three different location in Kampar within three consecutive days. The location included private institution (S1), household (S2) and discharge of water treatment plant (S3) for comparison of MPs occurrence in provider and end user. The analysis of MPs was performed using Nile Red (NR) stain visual sorting of MPs using fluorescent microscope. The experimental results reported that the concentration of MPs ranged from 169.7 \pm 46.5, 179.3 \pm 49.7 and 238.7 \pm 48.3 MPs/L in the source of S1, S2 and S3 respectively. MPs smaller than 10 µm was predominant in all samples. The average size was reported as 8.4 ± 4.9 , 2.7 ± 0.3 and $4.4 \pm 1.6 \mu m$ for S1, S2 and S3 respectively. Nano sized plastics (<1 µm) were also detected. Fragment shaped MPs was dominated in the water samples (55.3 - 60.0 %) followed by sphere (37.1 - 42.6 %) and fiber (1.0 %)- 3.4 %). Selected filtered samples were analyzed under ATR and SEM-EDX to determine the chemical composition, surface characteristic and elemental signature of MPs. Type of plastic determined were PET, PS/PA and PVC. Additional spectrum in fingerprint region indicated presence of additives in the MPs. This study indicated the existences of MPs in tap water and the unneglectable number of nano-plastics presences in tap water.

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LIST OF SYMBOLS / ABBREVIATIONS

%	Percentage
<	Less than
>	More than
±	Plus / Minus
-	То
Ø	Diameter
°C	Degree Celsius
cm ⁻¹	Per Centimetre
g/L	Gram per Litre
g/mL	Gram per Litre
kg	Kilogram
kg/capita/day	Kilogram per capita per day
L	Litre
М	Mole
mg/L	Milligram per Litre
mL	Millilitre
mm	Millimetre
MPs/L	Microplastics per Litre
nm	Nano Metre
particle/kg	Particles per Kilogram
particles.km ⁻²	Particles per Kilometre square
particles/g	Particles per Gram
particles/L	Particles per Litre
particles/m ³	Particle per Cubic Meter
µg/mL	Microgram per Millilitre

	٠	٠	٠
XV	1	1	1

μm	Micrometre
Δ1	Aluminium
C.	Carbon
C C-C	
C=0	Carbonyls
C-H-O	
C ₃ H ₆ O	n-Heyane
	Mathyl
	Alleyl Chlorida
	Mathylene group
	Chloring
C-N	Aryı
C-0	Ester, Etners, Alconols, Carboxylic Acid group
	Copper
Fe (II)	Iron with +2 oxidation state
H_2O_2	Hydrogen Peroxide
HCl	Hydrochloric Acid
HNO ₃	Nitric Acid
КОН	Potassium Hydroxide
Na	Sodium
Na	Sodium
NaCIO	Sodium hypochlorite
NaCl	Sodium Chloride
N-H	Nitrile
0	Oxygen
Si	Silicon
Sn	Stannum
Zn	Zinc
ZnCl ₂	Zinc Chloride
Zr	Zirconium
DM	Distancia Minara
DM	Dichroic Miltror

E	East
EDTA	Ethylenediaminetetraacetic Acid
EPDM	Ethylene Propylene Diene Monomer
EVA	Ethylene-Vinyl Acetate
HDPE	High Density Polyethylene
HW	Household water
LAP	Lembaga Air Perak
LDPE	Light Density Polyethylene
LW	Private institution water
MCE	Mixed Cellulose Esters
MOhm	MegaOhm
MPs	Microplastics
Ν	North
PA	Polyamide
PAHs	Polycyclic Aromatic Hydrocarbons
PCB	Polychlorinated Biphenyls
PE	Polyethylene
PE-PDM	Polyethylene – ethylene propylene diene monomer
PES	Polysulfone
PEST	Polyester
PET	Polyethylene Terephthalate
PI	Polyimide
POPs	Persistent Organic Pollutants
PP	Polypropylene
PPS	Polyphenylene Sulfide
PS	Polystyrene
PTFE	Polytetrafluoroethylene
PUR	Polyurethane
PVC	Polyvinyl Chloride
PVDC	Polyvinylidene Chloride
S1	Tap Water Source 1
S2	Tap Water Source 2
S3	Tap Water Source 3

SEBS	Styrene-ethylene-butylene-styrene					
SEM-EDX	Scanning Electron Microscopy (SEM) with Energy					
	Dispersive X-Ray Analysis (EDX)					
UV	Ultraviolet					
UV-B	Ultraviolet B					
WPO	Wet Peroxide Oxidation					
WW	Water Treatment Plant Water					
WWTPs	Wastewater Treatment Plant					

CHAPTER 1

INTRODUCTION

1.1 Background

The history of plastic can be traced back to 100 years ago. Plastic is developed from natural materials with intrinsic plastic properties. After years of evolution, wide range of synthetic and chemically modified plastic were found at all places. It is beneficial to society across all sectors as it is inexpensive, flexible, and high versatility. However, it is currently known as "the necessary evil". People can't live without it but don't want to live with it, particularly due to the plastic waste and its improper management (Jones, 2021). Figure 1.1 shows that there are about 350 million tons of plastics produced worldwide annually, and up to 40% is produced for packaging, in other word immediate discard. Additionally, it is estimated that about 8 million pieces of plastic reach to the oceans daily (Jambeck et al., 2015). Most of the plastics end up in the landfills and ultimately reach the environment in various sizes and shapes due to immature control of plastic waste management.



Figure 1.1: The global plastics production from 1950 to 2015 (in metric tons per year) (Jambeck et al., 2015)

The plastic debris which is visible and larger in size is known as megaplastic or macroplastic while the smaller plastic particle is classified as Microplastics (MPs). MPs is known as a solid microscopic and anthropogenic polymer particle which is water-insoluble and smaller than 5 μ m in diameter The official recognition of the size boundary of MPs was not launched, however the particles which are smaller than 1 μ m are labelled as nanoplastics instead of MPs (Bergmann et al., 2015).

Recent research has reported that MPs are found in the worldwide environment from the open surface water down to the deep sea and covers almost all the aquatic matrices (Bergmann et al., 2015). The MPs contamination has brought intense attention globally as the MPs is not only widespread over the aquatic and terrestrial environment but also entered the human sources food includes honey, seafood, table salt and tap water. People can minimise the consumption of some food intentionally to prevent exposure of MPs however, human cannot escape the daily intake of water (Zhang et al., 2020). The main challenge of this topic is the risks that MPs posed on human health are clueless and unknown (Koelmans et al., 2019). Additionally, the potential exposure route of MPs towards human through ingestion and inhalation is complex.

1.2 Effect of MPs to humans

The effects of MPs towards human health are still under investigation and the effect is expected to be dose dependent (Danopoulos et al., 2020). The possible risks associated with MPs exists in three forms which include the physical attributes (physically cause hazard), chemicals (broken monomers, additives, and adsorbed pollutants from the environment) and biofilms which the microorganisms can attach and colonized on the particles.

MPs with less than 130 μ m in diameter possess risks to translocate in human tissues and have potential to release its constituent monomers which may be the hazardous chemical additives added during the production. The hydrophobic properties of plastic enable them to adsorb chemical from the environment including PCB, PAH and heavy metals which are carcinogenic and toxic (Kosuth et al., 2018). The MPs may release those pollutants and heavy metal absorbed from the surroundings to human body once enter the body matrices (Cox et al., 2019). Furthermore, MPs have the potential to trigger toxic effect in human body by produce oxidative stress and cause tissue damage and chronic inflammation (Shen et al., 2020). The smaller the size of MPs can translocate into human body more effectively while the larger particles (> 2 μ m) will stay in the intestinal tract. It can penetrate to peripheral tissues and circulatory system through lymphatic system and cause systemic exposure (Shen et al., 2020).

1.3 Plastics in Malaysia

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In Malaysia, there are 0.5-1.9 kg of municipal solid waste (MSW) generated per capita per day and with 24 % of plastic comprised in the total composition (Aja and Al-Kayeim, 2014). By comparison with other Southeast Asia countries, the MSW generation is higher than Indonesia and Philippines with 0.22 kg and 0.4 kg/capita/day respectively (Chen et al., 2021). There are more than 1,300 plastic manufactures in Malaysia across all sectors and make Malaysia become one of the largest plastic production industries in worldwide (Amin et al., 2020). Figure 1.2 shows that Malaysia generates over 0.94 million tonnes of poor managed plastic waste per year (Jambeck et al., 2015). The mismanaged plastic waste, in other words not contained and disposal in dumps and open landfills, has high potential to enters the natural environment via wind, tidal and waterways transport, which eventually may enter human body in micro form by consumption of water (Ritchie and Roser, 2018). High income or developing countries who have effective waste management produced little of mismanaged plastic waste however in Asia, especially of the developing countries including Malaysia, having the high mismanaged plastic waste rates (Ritchie and Roser, 2018). Furthermore, Malaysia is rank 8th among the top ten countries who have huge contribution towards the plastic pollution crisis over the world (Ibrahim and Noordin, 2020). Nevertheless, the environmental investigation of plastic pollution especially MPs contamination is insufficient and almost blank data for MPs in drinking water (Amin et al., 2020).



Figure 1.2: Mismanaged plastic waste in Asia region (Jambeck et al., 2015)

1.4 MP in fresh water

The most possible and easiest way of MPs enters human body is through consumption of water. Moreover, the available research showed that the potential uptake of MPs consumption by human body is estimated to 39,00 to 52,000 particles varied from age and sex and it can reach up to 74,000 to 12,100 particles uptake annually if inhalation is considered (Cox et al., 2019). Therefore, the occurrence of MPs in tap water should not be overlooked. Another study also concluded that the average person may ingest more than 5,800 particles of MPs annually from tap water, beer and table salt with largest contribution from tap water (81%) (Kosuth et al., 2018). Research also proved that the concentration of MPs ranged from zero to more than thousands of particles per litre in drinking water globally (Novotna et al., 2019). The efficiency of wastewater treatment including coagulation, sedimentation and filtration directly affect the presence of MPs in tap water (Shen et al., 2020). Recent study showed that the MPs were detected in 81% of the tap water sample over 159 countries of globally sourced tap water including sample from developed and developing countries (Kosuth et al., 2018). However, the information regarding the concentration of MPs detected in drinking water or tap water from different countries are uneven and the data is limited. The studies that proved the MPs has contaminated global drinking water or tap water is limited. Additionally, the sampling method, sample pretreatment procedure, detection technique which is the microscopy used in the analysis are not uniform. The different method used among different individuals has the potential to bring false positive result (Shen et al., 2020). Therefore, these will be a great key challenges in this topic as the available data varies in quality and not comparable due to the method for collection and analysis of MPs is not standardized (Weber et al., 2019).

Recent research and investigation in Malaysia showed that MPs present in river (Zaki et al., 2021), surface seawater (Amin et al., 2020) and mangrove forest (Barasarathi et al., 2014). The research exploration regarding to the topic of MPs is still inadequate. Indeed, the studies and database documenting the MPs in tap water in Malaysia are blank.

1.5 Problem Statement

The presence of MPs has been proven in all aquatic environment including marine and freshwater and the research and database so far have focus mainly on marine environment (Danopoulos et al., 2020). As the MPs penetrated the food web, its contamination alerts people with the food safety issue. The occurrence of MPs in drinking water will cause unknown health effect towards human body and it is expected to reduce the drinking water quality. It will trigger an attention in Malaysia community as well as the social media, putting this issue on the agenda of public health agency in Malaysia because ensuring safe drinking water is significant political agenda in every country (Koelmans et al., 2019). Further research and investigation of MPs towards human body need to be carried out to prove the existence of MPs whether cause harm or neutral to human body. This is because MPs capable to pass through biological barriers, penetrates tissues and accumulate in vital organs, as well as disturb metabolism and behavior of human organisms (Mattsson et al., 2017). Current wastewater treatment is also needed to be assessed to make sure the optimum removal efficiency of MPs in drinking water.

To the best of our knowledge, there is blank data for MPs detection in tap water in Malaysia and the MPs concentration in tap water in end-user in the household distribution system only have been investigated by few studies (Kosuth et al., 2018; Mintenig et al., 2019; Tong et al., 2020). The piping system in household distribution system in Malaysia are mainly made by plastic and the pipe possess risk of increasing MPs in tap water (Tong et al., 2020). Hence, this study is to close the research gaps of the MPs contamination in tap water.

1.6 Aim

Thus, this study is aimed to detect the concentration of MPs and its characteristics in the tap water of end user household distribution system in Kampar, Malaysia.

1.7 Research Objectives

The objectives of this thesis are shown as follows:

- i. To determine the quantity, shape, and size of MPs in drinking water.
- ii. To identify the chemical functional group of the particles using ATR-FTIR.
- iii. To compare the findings of MP existences in three different water sources.

1.8 Scope of Study

The current study was to identify the concentration of MPs in tap water samples collected from 3 different sources in Kampar, Perak. The sample are collected in 5 sets at each sampling point at three continuous days during the similar timeslot with total of 45 samples collected by using glass bottle. All the samples stored at fridge at 4 °C

to prevent degradation and sunlight. The samples had undergone H_2O_2 digestion to eliminate all the possible organic matters before the analysis subsequently followed by Nile Red staining. After that, quantification and characterization of MPs were conducted by using fluorescence microscope with visual detection. Surface characteristic and its elemental signature of the MPs were determined by using SEM-EDX. Furthermore, ATR-FTIR was used to identify the polymeric information of MPs. The concentration of MPs obtained in this study was reported as MPs/L.

CHAPTER 2

LITERATURE REVIEW

2.1 Plastic

2.1.1 Brief history of plastic

Back to the middle of 19th century in which the first synthetic polymer was invented by John Wesley Hyatt which is the celluloid, to substitute for natural materials including ivory, tortoiseshell, horn and linen which obtained through massive slaughter of innocent wild animals (Science Matters, 2021). This revolutionary invention proved human could produce new materials, freed human from relying solely on natural resource. In early century of 19th, Leo Baekeland launched Bakelite, the first fully synthetic plastic which contained only synthetic molecule (Science Matters, 2021). Its' astonishing properties including malleability, versatility and affordability for mass production stunned the world, and here the "Plastic Age" began (Freinkel, 2011). The revolution of plastic is even expanded widely during the World War II. The synthetic silk nylon, discovered for production of medical and warfare applications, and the Plexiglas, replace glass for aircraft window (Science Matters, 2021). Other than, various type of plastic launched between two World Wars such as cellophane, polyvinyl chloride, polystyrene, and polyethylene. The plastic production surged even the wars ended, increased estimate twentyfold around 1950 - 1970, and continue to grow unstoppable until reaching around 350 million metric tons in 2017 (Chalmin, 2019). Plastic is the only industry experienced such growth in the world, successfully challenged the traditional materials and become the dominant material in most of the production over the world (Freinkel, 2011).



Figure 2.1: The global plastic production in million metric tons (Chalmin, 2019).

2.1.2 Type of plastic

Plastic, a word originated from Greek verb *plassein*, has the meaning of "to mold or shape" (Freinkel, 2011). It is extremely versatile to be applied in wide range of field and industry. After years of revolution and chemically modified, 7 type of common plastic exist and available in the market summarized in Table 2.1

Plastic type	Properties	Characteristic	Application	Reference
Polyethylene	Synthesized from	 Low density, transparent 	 containers, bottle 	(Sax, 2010)
Terephthalate	ethylene glycol and	High Strain resistance	 packaging 	(Sharon &
$(C_{10}H_8O_4)_n$	terephthalic acid	Chemical resistance		Sharon, 2012)
		Thermo-stability		(Lanaro et al.,
				2018)
Polyethylene	Obtained by catalytic	• Excellent chemical resistance	 Packaging 	(Ugbolue, 2017)
-(CH ₂ -CH ₂) _n -	polymerization of	 Near-zero moisture absorption 	 Films, laminates 	
	ethylene	Thermoplastic	 Household goods 	
			 Tubes and pipes 	

Table 2.1:

Table 2.1 The plastic type and its respective properties, characteristic and application with reference

Polyvinyl chloride	Obtained by	chain	•	High rigidity	•	Pipes	(Zohuri, 2012)
(PVC)	polymerization	of	•	Thermoplastic	•	Floor covering	(Doble and
-(CH ₂ -CHCl) _n -	vinyl chloride		•	Chemical resistance	•	Roofing sheets	Kumar, 2005)
					•	Cable insulation	
Polypropylene	Obtained by	chain	•	Excellent chemical and mechanical	•	Industrial application	(Jaffe and
-[CH ₂ -CH(CH ₃)] _n -	polymerization	of		properties	•	Reinforcing fiber in	Menczel, 2020)
	propylene		•	High softening point		concrete or soil	
					•	Sanitary product	
					•	Surgical sutures	
					•	Automotive fittings	
Polystyrene	Obtained by	chain	•	Low thermal conductivity	•	Fresh food packaging	(Begum et al.,
-[CH ₂ -	polymerization	of	•	Low density	•	Foamed packaging	2020)
$CH(C_6H_5)]_n-$	styrene (pro	oduced	•	Almost zero compressive strength	•	Toys	(Niaounakis,
	from ethylene	and	•	Gas transmissible			2019)
	benzene)						

2.2 MPs Characteristics

2.2.1 Trend of MP globally

The demand and reliant of plastic raised since commercial production started in the middle of 20th century (Hale et al., 2020). The variety in properties of plastic fulfilled global demand in every field and aspect. However, majority of the plastic discarded and abandoned on land. Riverine, atmospheric transport and precipitation, and currents redistributed the plastic and MPs over the world. The plastic and MPs sink and eventually reached the ocean sediment as the ultimate destination (Hale et al., 2020). It widely distributed and now existed in every corner on the Earth and marked as the major threat in environmental and ecology (Zhang et al., 2020).

MPs have been discovered on the sore, surface of the sea, seabed from the coast to the open sea, and it even reached Artic and Antarctic Oceans (Law and Thompson, 2014). Other than that, the MPs is even indicated in polar sea ice (Obbard et al., 2014). The first record on the existence of the tiny plastic particles in surface water and fish was around 1970s, and its increasing attention of their existence documented in the early 2000s after highlighted as "MPs" (Shim and Thomposon, 2015). Studies and research regarding the distribution, fate, effects of MPs increase exponentially over past decade globally (Kershaw, 2015). The majority research originated from Western Europe, UK and United States, while the contribution from developing countries was limited (Zhang et al., 2020).

2.2.2 Origin of MPs

The prevailing weather such as rain and wind act as external force in influencing and distribute the existing pattern of MPs in water or land (Zhao et al., 2015). The wind driven the turbulence in air which transport the plastic to farther place and the increase of rainfall raised the amount of plastic debris entering to the water body. Although majority of the plastic produced nowadays are wear resistance material, durable to environmental degradation and chemical wear, but once they are not properly treated and enter the environment, they will be worn down by the nature.

The most significant degradation can be divided into physical degradation by abrasive forces, high temperature, freezing and thawing, photodegradation which initiated by UV light, chemical degradation through hydrolysis and oxidation and the last, is the biodegradation by organisms such as bacteria, algae, or fungi (Klein et al., 2018). The substantial degradation weakens the plastic and become brittle and the chemical bond in the polymer backbone breaks lead to chain scission and depolymerisation (Bergmann et al., 2015). The plastic debris shredded into smaller pieces during the movement across different habitats carried by prevailing weather (Klein et al., 2018). Those tiny plastic particles can go through further degradation even these tiny fragments are invisible to naked eye. Mechanical and physical degradation is the most important factor of MPs formation in aquatic environment. Then, it proceeded with photooxidation and chemical oxidation. The UV-B radiation from the sun initiated the photo-oxidative degradation and progress if oxygen is available (Smith, 2018).

However, the rate of these process includes photodegradation and fragmentation under natural condition is still undiscovered (Koelmans et al., 2019). This is because of the vast variability in oxygen and temperature in the environment caused different rates of processes for different type of polymer (Bergmann et al., 2015). The MPs can reach to the complete mineralization stage when the environmental parameter and presence of microorganism with enzymes are in favourable condition, but the situation is very seldom occurred (Klein et al., 2018). Figure 2.2 illustrated the brief degradation pathway of plastic.



Figure 2.2: The degradation pathway of plastic (Klein et al., 2018).

Other than natural factor, the MPs originates from human production in which intentionally manufactured in small size for variety purpose such as microbeads and pellets found in cosmetic products, including scrubs and facial exfoliating cream, shampoos and make up product (Xu et al., 2020). Furthermore, the by-product released during the use of plastic product such as microfibers from textile and clothing industry, abrasion of synthetic rubber from tyres, wearing from synthetic polymer commercial goods, MPs from paints, as well as unintentional discharged of pelleted raw materials or wastewater into environment (Wang et al., 2019; Xu et al., 2020). These actions enable new micro sized plastic particles added to the environment.

2.2.3 Types of MPs

MPs can be categorized as primary and secondary MPs depending on the source. Primary MPs are anthropogenic manufactured in millimetric size which can be found in various household product including microbeads from personal care product, cleaning product and exfoliating creams (Europarl, 2020). Whilst secondary MPs are those formed by the photodegradation and fragmentation of larger debris of plastics into ever-smaller fragment or plastic product wear (Bergmann et al., 2015).
2.2.4 Characteristics of MPs

MPs' structure is high diversity and complexity in which their size, shape and density differ in various scale. Furthermore, when MPs exposed to natural environment, the chemical contaminants and formation of biofilm combined with their original polymers and chemical additives further increase the complexity (Kooi and Koelmans, 2019).

The various particle size of plastic shown in Table 2.2, which the plastic particles between 1 - 5 mm will be acknowledged as MPs (Noik and Tuah, 2015). The shape of MPs could be categorized into several main types such as fiber, fragment, beads, film, foam and sheet. However, the abundance of MPs in nature is hard to estimate due to spatial challenges and heterogeneity of the plastic (Noik and Tuah, 2015). The various type of MPs with its respective characteristics and origin illustrated in Table 2.3

Plastic group	Plastic size
Macroplastic	> 20 mm
Mesoplastic	5-20 mm
Microplastics	$1 \ \mu m - 5 \ mm$
Nanoplastic	< 1 µm

Table 2.2: Plastic group with their respective estimate size range

Plastic shape	Characteristic	Origin		
Fibre	Linear, and even in thickness	Shedding from textile and cloth		
Fragment	Flattened and angular	Fragmentation of plastic debris		
Beads	Granular and spherical	Manufactured in micro- sized, usually from cosmetic product		
Film	Thin and transparent	Fragment from soft plastic material such as plastic bag		
Foam	Compressible and sponge-like texture	Insulation or food packaging		
Sheet	Thick and not transparent	Hard fragment of thin polymer		

Table 2.3: The various type of MPs and their respective characteristic and origin(Bullard et al., 2021; Wu et al., 2017)

2.3 Effects of MPs

2.3.1 Environmental impact of MPs

The present is driven by the accumulation trace in the past, the future of the earth will bear the plastic mark caused by mankind (Davis, 2015). The limited recovery of discarded plastic led to the accumulation on the environment and the concern about the impact caused by the MPs in different compartment is raising. With the assistant of weather, MPs can move between different environment matrix including air, terrestrial, habitats, freshwater, and ocean, which including sediment (Rillig and Lehmann, 2020). Terrestrial system obtained less attention compared to aquatic system however the abundance of MPs in terrestrial matrix caused negative effect. The leaching of chemical additives and plasticizers from MPs can accumulate and eventually alter physical properties of the soil. Micro- and mesofauna such as mites, collembola and enchytraeids may ingest the MPs present in the soil and accumulate the contaminants in the terrestrial food web (Rillig, 2012).

2.3.2 Effects of plastic on marine life

The phenomenon of the entanglement and ingestion of macroplastic debris in marine organism are widely recognised, over 250 marine species are expected to be suffered by plastic ingestion (Wright et al., 2013). Macro filter feeders such as Franciscana dolphins and baleen whale tends to consume floating or submerged plastic in the sea column due to non-selective feeding habit, cause impairment to digestive system and blockage of intestinal tract (Guzzetti et al., 2018). Other than macroplastic, the invisible MPs possess serious threat to marine life as they are potentially bioavailable to wide range of marine life. Planktonic organism, deposit-and detritus feeders, and filter-and suspension feeders, and echinoderms are highly vulnerable to MPs (Guzzetti et al., 2018). This is because marine organisms lack of specific enzyme to digest the synthetic polymer and will considered as bioinert element once the MPs enters their body (Andrady, 2011).

The MPs accumulate within organism which result in internal abrasion and injuries, blockage of tract which induced physiological stress, alter feeding behaviour which retard growth and fertility (Ogonowski et al., 2018). The chemical additives and monomers added during manufacturing process to enhance the plastic's performance could contain heavy metal, hydrocarbons, and POPs which capable to cause toxicity and harm the marine biota (Wright et al., 2013). The toxicological effect includes distortion of metabolism and reproductive activity, weaken immune system, induce toxicity in cellular and cancerous tumour formation (Guzzetti et al., 2018). Other than that, research has proved that exposure to MPs enable marine fish to decrease of predatory activities which could cause reduction of catching prey and escape from predators (Ferreira et al., 2016). Furthermore, MPs contamination can affect coral as the unintentionally uptake of high concentration of MPs as food will damage the tissue within the coral gut which ultimately damage the coral's health (Hall et al., 2015). Moreover, MPs can be the vector of toxic contaminant due to its high surface area to volume ratio to accumulate hydrophobic POPs. Bioaccumulation and biomagnification could occur along the food chain when the contaminated MPs consumed by marine organism (Wright et al., 2013).

2.3.3 MPs and human health

While the debate of MPs on human health still under way, various studies regarding the ingestion of MPs on animal proved to cause deleterious consequences. The toxicity of MPs varies from physical properties, chemical properties (type of additives and polymer), exposure concentration and the microbial films (Braun et al., 2021). MPs ingestion has been proved in wide range of human-consumed seafood such as fish, bivalve, and crustacean (Barboza et al., 2018). Human ingestion of MPs contaminated marine organisms raise attention across the globe as the knowledge about the effects is still unknown and under investigation. Furthermore, research has shown that MPs present in various food product such as canned sardine (Karami et al., 2018), sea salt (Karami et al., 2017), table salt (Yang et al., 2015), honey, sugar (Liebezeit and Liebezeit, 2014), and beer (Kosuth et al., 2018). The toxic effect brought by the orally consumed MPs is still unknown, and due to the bioinert properties the effects are likely

to occur at extreme high dose and intake. No research or data related to human is available with only little research performed with mice (BfR, 2020).

Other than consumption, human may expose to MPs externally from various sources such as primary MPs in personal care product including hand wash, shower gel, toothpaste, and scrub (Sharma and Chatterjee, 2017). Research has concluded that prolonged usage of the product which presence of MPs result in MPs absorption in tissues and cause skin damage (BfR, 2020).

2.4 Methods to detect MPs

The analytical method and standard procedure are still in the beginning stage due to MPs study is new emerging topic in recent decades. For common practise, the analytical procedure for samples involves 3 steps which are sample collection, pre-treatment of sample and lastly the MPs identification and characterization (Sun et al., 2017).

2.4.1 Sample collection

The sample collection can be performed in several ways such as container collection, autosampler collection, separate pumping and filtration and surface filtration (Sun et al., 2019). Container collection or autosamplers allows only few litres per collection, which suitable for environment analysis of MPs. Separate pumping and filtration capable to provide sample volume up to hundred litres, which commonly used to collect MPs in effluent of wastewater treatment plants. The surface filtering assembled for skimming water surface at final discharge of wastewater treatment plant which can further increase the collection volume to thousands cubic meter (Sun et al., 2019). Trawling technology is the main method of water sampling and the most popular method is sampling by manta trawl (Lv et al., 2021). Towing of neuston nets or manta nets usually used for sampling of surface water while plankton nets with small mesh size are normally towed at low velocities flow (Prata et al., 2018). After collection, the samples are normally filtered to concentrate the MPs and the mesh size of filters has significant effect on the concentration of MPs (Magnusson and Noren, 2014). The mesh size of the filter ranged from 1 µm - 500 µm, and stack of sieve pans is performed for filtration in much research to increase filtration volume and differentiate size category of MPs (Sun et al., 2019).

2.4.2 Sample pre-treatment

Sample collected from the environment or WWTPs usually contained organic and inorganic matters, thus various type of method applicable to purify the sample and facilitate the characteristic and identification of MPs (Sun et al., 2019). Catalytic wet peroxidation (WPO) is one of the widely used methods to digest organic matter in seawater, freshwater, and sediment sample. The application of oxidizing agent includes H₂O₂, NaCIO and Fenton reagents, the plastic particles usually remain unaffected during the WPO process. Particularly, Fenton reagents able to breakdown organic material in short period without interfering the MPs (Tagg et al., 2017). Other than that, enzymatic degradation is another effective approach to digest organic matter however it is still only applied in small scale (Lv et al., 2021). Submerging of MPs

sample in mixture of enzyme include lipase, amylase, proteinase, chitinase and cellulase capable to effectively remove organic matter without disturb the MPs (Cole et al., 2014).

Alternative method includes alkaline treatment with sodium hydroxide and potassium hydroxide and acid digestion with nitric acid and hydrochloric acid. Appropriate concentration of acid and alkaline able to digest unwanted biological matters however both methods require more concern (Sun et al., 2019). This is due to strong oxidizing acid capable to damage and corrode the plastic in different degrees. Similarly, high concentration of alkaline could cause partially destruction, melding and yellowing of the MPs (Lv et al., 2021).

2.4.3 MPs identification

At present, the combination of physical and chemical analysis is the common method by using microscopy and spectroscopy to analyse MPs due to single method is not sufficient to provide representative and accurate data to identify MPs in various size, shape, and polymer type (Shim et al., 2016). However, the use of spectroscopy is limited by high cost, substantial time, and effort for processing sample, require of skilled personnel which limit the number of sample detection, particularly detection of weathered and contaminated MPs (Prata et al., 2019). Current method is expected to be improved or development of new method to decrease the identification time to detect the MPs in samples (Shim et al., 2016).

2.4.3.1 Visual Identification

Before the high popularity of MPs research, mostly large MPs was reported on beaches and only little in surface water. Sorting and identification were normally done simultaneously in a tray with forceps and observed with naked eyes. MPs with larger size, colourful MPs fragment and pre-production resin pellets can be observed easily for clear sample. However, there is high probability of missing counting of MPs in the sample which contain high level of interfering inorganic and organic particles which increase the difficulties to differentiate the interfering material with ambiguous MPs. Thus, this easy and fast method only applicable for non-professional volunteers (Shim et al., 2016).

2.4.3.2 Microscopy

Magnified figure by using microscopy provide surface texture and structural data in detailed which is important to identify ambiguous plastic particles. However, false identification usually occurred by using microscopy with over 20 % and over 70 % error especially for transparent plastic particle, then these results were further confirmed with spectroscopic analysis. Variation may occur between the observers which can lead to overestimation or underestimation, and it is very time-consuming (Prata et al., 2018). For instance, synthetic and natural fibres were hard to distinguish by only microscopy due to similar shape, and many coloured fragments were easily identified as synthetic resin from paints (Song et al., 2015). Scanning electron microscopy (SEM) is capable to provide extremely detailed and high-magnification figure of ambiguous plastic particles which can assist the discrimination of the real plastic particle from organic material. Furthermore, energy-disperse X-ray spectroscopy (EDS) can provide elemental information and composition of the particles to differentiate the carbon-dominant plastic from inorganic material. However, this SEM-EDS is expensive, require more effort and longer preparation time and examination for sample which limit the amount of sample handled.

To overcome the challenges of visual identification by microscopy, staining dye, which is the Nile Red and so far, it is the most promising staining method for MPs (Prata et al., 2018). Nile Red provide high recovery rates which reaching 96.6%, require only short incubation time and enable vibrational spectroscopy to skip the short cleaning step with bleach (Erni-Cassola et al., 2017). Biogenic materials including algae and seaweed will not stained by Nile Red and weathering of plastic will not disturb the effect of staining (Maes et al., 2017).

2.4.3.3 Fourier Transform Infrared (FTIR) spectroscopy

FTIR spectroscopy capable to provide data on specific chemical bonds of particles in which can distinguish plastic from organic and inorganic element due to different bond compositions will generate unique spectra, thus carbon-based polymer can be easily identified. By comparison, spectroscopy identification is more accurate than microscope, with minimum missing and miscount rates (Song et al., 2015). It generated an infrared spectrum resulting from the change in dipole moment (Prata et al., 2018). Identification of MPs by this method can reveal the chemical composition of polymer as well as its abundance, which can obtain clue to the possible origin, source, or pathway of the MPs to the study area. Other FTIR technique such as attenuated total reflection FTIR (ATR-FTIR) provide irregular MPs data, and micro-FTIR able to generate high-resolution map of sample size down to 20 μ m (Prata et al., 2018).

2.4.3.4 Raman Spectroscopy

The application of Raman spectroscopy enables to differentiate the polymer type and provide plausible abundance of MPs in the sample (Anger et al., 2018). It uses laser beam falling on a particle result in different frequencies of back-scattered light according to different molecular structure and atoms of to generate unique spectrum

for each specific polymer particle. Raman spectroscopy provides element composition of polymer which is similar to FTIR. Those spectrum responses from FTIR and Raman spectroscopy can ensure for complex MPs identification. Both spectroscopy method is non-destructible, which possible for further analysis and highly accurate (Prata et al., 2018). By comparison, Raman spectroscopy has smaller diameter of laser beam which enable the identification of MPs reach to as small as few μ m in size (Shim et al., 2016). It allows identification of MPs size less than 20 μ m, however weak signals may be produced due to extreme small size. It can be enhanced by increase of measurement time and fluorescence interference when weak signals generated (Prata et al., 2018). However, Raman spectroscopy has weakness which is sensitive to chemical additives and pigment in MPs which may contribute to the false identification of polymer type.

2.5 MP in the environment

As MPs is an emerging research topic in recent decades, majority of the study is the first-time data regarding the quantitative and occurrence of MPs. It is difficult and complicated to compare global data with the reported MPs abundance due to inconsistent of standard methodologies and reporting units for MPs analysis, and different sampling method will lead to result varied.

Several research regarding the MPs analysis in marine, river, tap water and bottled water has been revised to investigate different procedure performed and their respective founding. Table 2.4 - 2.12 shown the sampling method, sample pre-treatment method, identification and quantification method, MPs abundance, MPs morphology, type of MPs identified, MPs morphology in marine, river, tap and bottled water from different countries.

2.5.1 MPs in Marine

Marine, which is the ultimate destination and fate of MPs accumulation triggers most of the scientist's interest for MPs research related to marine system (Xu et al., 2020). The research of MPs in marine water was summarised in Table 2.4 and 2.5.

The exact amount of plastic entering to the ocean is uncountable, recent research estimate that there are at least 5 trillion plastic pieces on the ocean which comprised of 269,000 tonnes of floating plastic (Maes et al., 2017). The mega-plastic eventually degraded and become MPs. Other than primary and secondary source of MPs, the abrasion of tyres, road marking, and dust which escape the water treatment and brought to water body by city stormwater runoff and effluent and eventually reach to marine environment (Cheung and Fok, 2016). Besides, the fishing gear including fishing net and ropes is also one of the potential sources of MPs. Plastic mulching in agricultural activities, illegal dumping of waste, intentional or unintentional leakage from vessels and litter abandoned on beached due to recreational activities also directly increase the abundance of MPs in ocean (Simon-Sánchez et al., 2019).

In recent years, studies and research have been conducted on the occurrence and distribution of MPs in environment, and majority is focus on marine system. MPs has been detected in almost habitat around the world because it mainly distributed by prevailing environmental conditions such as weather and wind circulation (Kwon et al., 2020). Plastic usually will float on the surface of the sea due to less density than seawater, and the buoyancy and density of the plastic cause changes in their residence in sea because of weathering and biofouling, thus MPs is able to be discovered across surface, water column and sediment in the sea (Maes et al., 2017). The prevailing wind and tide cause formation of ocean current and mixing of surface water which responsible for the vertical distribution of MPs in water column of marine system (Murray and Cowie, 2011). The denser polymer such as PVC tend to settle and sink down, but they are still can be transported by underlying current (do Sul and Costa, 2014). The MPs concentration is high in near-shore seawater which directly reflect the relationship of terrestrial plastic pollution from anthropogenic activities and MPs contamination in marine (Kwon et al., 2020).

 Table 2.4: The case study summaries of sampling, sample pre-treatment method, identification/quantification method, MPs morphology

 and type of MPs identified in marine water

Sampling	Sampling	Sample Pre-	Identification/	MPs	Maximum MPs	Reference
Location	Method	treatment Method	quantification	Abundance	abundance density	
			Method	density		
Victoria Harbor,	Towing	30% hydrogen	stereomicroscope,	0.11 - 27	$0.22 - 36 \text{ particles/ } \text{m}^3$	(Tsang et al.,
Hong Kong	plankton net	peroxide	ATR-FTIR	particles/ m ³		2017)
Coast of Korea	Manta trawl	Hydrogen peroxide	FTIR	1.12 - 4.73	-	(Kwon et al.,
				particles/m ³		2020)
Bohai Sea, China	Manta net	Hydrogen peroxide	Micro-FTIR	0.33 particles/	1.0 - 1.5 particles/m ³	(Zhang et al.,
		and ferrous sulfate		m ³		2017)
Sweden	Manta trawl	-	Stereomicroscopy	0.04 - 2.74	50.4 particles/m ³	(Schönlau et al.,
	Pump			particles/m ³		2020)
North-East	Manta trawl	-	Spectroscopes	0.14	1.5 particles/m ³	(Maes et al.,
Atlantic, UK				particles/m ³		2017)
Persian Gulf, Iran	Neuston net	Wet sieved, Zinc	Stereomicroscope,	1.8 x 10^4	$4.6 \text{ x } 10^4 \text{ particles.km}^{-2}$	(Kor and
		salt-saturated	FTIR	particles.km ⁻²		Mehdinia, 2020)
		solution				
Tropical Eastern	Plankton net	30% hydrogen	AmScope	$0.15 \pm 0.05,$	-	(Alfaro-Núñez
Pacific and		peroxide	trinocular	0.03 ± 0.01		et al., 2021)
Galápagos			stereoscope	$\mu p/m^3$)		

Table 2.5:	The case study	y summaries of	sample volume,	MP morphology	, type of MP id	entified and %	of sample conta	aining MP in	marine
water									

Sampling	Sampling	Mesh size	MPs Morphology	Type of MPs identified	Reference
Location	sites	(µm)			
Victoria Harbor,	9	153	Pellet > fragment > fibre	PP, HDPE, LDPE,	(Tsang et al., 2017)
Hong Kong				PP+EPDM, styrene	
				acrylonitrile	
Coast of Korea	8	330	Fragment > sphere >	PEST, PP, PS	(Kwon et al., 2020)
			fibre > film		
Bohai Sea, China	11	330	Fragment > fibre > films	PE, PP, PS, PET	(Zhang et al., 2017)
Sweden	12	333	Fibre	PE, PP	(Schönlau et al., 2020)
North Fost		222	Encoments films nellet		(Mass et al. 2017)
Atlantic UK	-	333	rragment > mm > penet	-	(Maes et al., 2017)
Persian Gulf, Iran	15	300	Fibre > fragment > pellet >	PE, PP	(Kor and Mehdinia, 2020)
			film		
Tropical Eastern	40	150	Fibre	-	(Alfaro-Núñez et al., 2021)
Pacific and		500			
Galapagos					

2.5.2 MPs in River

The research of MPs discovered in river water was summarised in Table 2.6 and 2.7. River is the significant pathway for transportation of land-based plastic debris and MPs from terrestrial to the ocean (Eo et al., 2019). It contributed 1.1 to 2.4 million tonnes of plastic to ocean however it received far less attention compared to marine environment (Simon-Sánchez et al., 2019). Majority of the freshwater systems surrounded by dense population and lead to increasing of anthropogenic activities such as sewage discharge and illegal dumping of waste into the river thus raise the occurrence of MPs in the freshwater system (Rodrigues et al., 2018). The high concentration of MPs in freshwater such as river not only threaten river living organisms but also human due to direct and frequent contact with human compared to oceans (Jiang et al., 2019). This is because the freshwater in river provides drinking water and source of food which harbour millions of people lived along the riverine and the river habitat (Pan et al., 2020). The riverine discharge of the MPs to the ocean seasonally and annually are the important factor influencing the abundance of MPs in coastal seawater and shoreline (Eo et al., 2019). Weather and seasonal variation has great influence on the abundance of MPs in river as the wet deposition from atmosphere directly affect the hydrology of the river. The physical forces including water flow velocity and water level influences the concentration of MPs in river (Campanale et al., 2019).

Other than floating MPs concentrated on surface water, significant number of MPs also found in mid and bottom of the water column. This is due to the denser plastic tends to sink and will be transported though the bottom of water, and the sediment of the river is likely served as the reservoir for MPs deposition (Eo et al., 2019). Research also found that the abundance of MPs in the bottom water was higher than surface water in river in late rainy season (Lima et al., 2014). Thus, investigation on the abundance of MPs on surface water, including upstream and downstream as well as sediment is crucial to fully understand the fate and distribution of MPs in river, to develop effective strategies and policies to manage the MPs pollution and mitigate the negative effects on the environment and human.

Sampling	Sampling	Sample Pre-	Identification/	MPs	Maximum MPs	Reference
Location	Method	treatment Method	quantification Method	Abundance	abundance	Reference
Location	wiethou	ti catilient Methou	quantification Method	Abundance domaites	domaiter	
				density	density	
Ofanto river,	Plankton net	30% hydrogen	Microscope, spectrometry	0.9 - 1.3	13 - 18	(Campanale et
Sountern Italy		peroxide		particle/m ³	particle/m ³	al., 2019)
Nakdong River,	100 L water	Hydrogen peroxide	FTIR	293 - 376	4760 - 10002	(Eo et al., 2019)
South Korea	transfer to 20 µm	and Fe(II) solution		particle/m ³	particle/m ³	
	potable net					
Zhangjiang	Bulk sampling,	Wet peroxide	Microscope, micro-Raman	246	725 particle/m ³	(Pan et al.,
river, China	Manta net	oxidation, density	spectroscopic	particle/m ³		2020)
		separation				
River of Tiber	Flow sampler	Wet peroxide	Stereoscopic microscope,	483 - 967	1108 particle/m ³	(Jiang et al.,
Plateau, China		oxidation	Raman spectroscopy	particle/m ³		2019)
			~			
Antuã River,	Water pump	Wet peroxide	Stereomicroscope, FTIR	58 - 193	71 - 1265	(Rodrigues et
Portugal	with 0.055 mm	oxidation with		particle/m ³	particle/m ³	al., 2018)
	mesh net	addition of zinc				
		chloride				
Langat River,	Manual	Filtration	FTIR	4.39 ± 5.11	45.86 ± 24.76	(Chen et al.,
Selangor	collection	30% hydrogen		particles/ L	particles/L	2021)
Malaysia		peroxide			90.0 particles/L	
				1		

Table 2.6: The case study summaries of sampling, sample pre-treatment method, identification/quantification method, MPs morphologyand type of MPs identified in river system

Table 2.7: The case study summaries of sample volume, MP morphology, type of MP identified and % of sample containing MP in riverwater

Sampling Location	Mesh size (µm)	MPs Morphology	Type of MPs identified	Reference
Ofanto river, Sountern Italy	333	Fragment > flakes	PE	(Campanale et al., 2019)
Nakdong River, South Korea	300	-	PP, PES, PE, PS, alkyd, acrylic	(Eo et al., 2019)
Zhangjiang River, China	330	Fragment > fibre > pellet	PP, PE, PS, PES, PET	(Pan et al., 2020)
River of Tibet Plateau, China	-	Fibre > fragment > pellet	PET, PE, PP, PS, PA	(Jiang et al., 2019)
Antuã River, Portugal	-	Fibre > fragment> foams	PE, PP	(Rodrigues et al., 2018)
Langat River, Selangor Malaysia	-	Fragment> films > beads	PET, HDPE, LDPE, PP, EVA, PVDC, PS	(Chen et al., 2021)

2.6 MPs in drinking and tap water

MPs contamination in tap water grabbed great attention over the world due to its daily consumption, including cooking and washing of food materials (Pratesi et al., 2021). Drinking water is the essential element for survival which cannot be eliminated, and yet tap water is the dominant source of anthropogenic particles among other consumables as well as it is the simplest food matrix to analyse (Kosuth et al., 2018; Oßmann, 2021). Water for daily consumption usually obtained from various freshwater sources such as river, lakes and reservoirs which are subject to MPs exposure through different pathway (Eerkes-Medrano et al., 2019). The possible pathway of MPs reaching household tap water including the MPs-polluted water reservoirs, atmospheric deposition, and physical abrasion of piping in water supply and distribution system which made from plastic (Lam et al., 2020).

The identification of MPs in tap water is complicated due to the source of tap water obtained from different raw water sources such as ground water or surface water. Surface water has higher risks to MPs contamination due to exposure directly to the environment. The raw water is treated through several procedure include coagulation/flocculation, sedimentation, filtration/floatation, and disinfection (Oßmann, 2021). Almost all the large MPs (> 10 μ m) is effectively removed during the coagulant/flocculation process but only 80 % removal rate of small MPs (> 1 μ m) which may lead to remaining of small MPs exist in the water, or even increased in the treatment process (Li et al., 2020). This is because the residue of coagulant polyacrylamide (PAM) used in coagulation which made from polymer may remain in the treated water (Pivokonsky et al., 2018; Wang et al., 2020). Furthermore, the polymeric membranes for filtration process may cause leaching of MPs especially aged membranes (Oßmann, 2021). Although wastewater treatment plant capable to effectively remove majority of the MPs, however due to huge amount of MPs entered the WWTPs, the treated effluent may still contain significant amount of MPs and eventually reach to human by tap water (Eerkes-Medrano et al., 2019). The prolonged consumption of MPs potential to cause deleterious consequences to human health due to their bio-persistence properties which can accumulate and deposited in organs and tissue (Pratesi et al., 2021).

In 2018 the first research regarding the occurrence of MPs in tap water revealed that 81 % of the tap water sample obtained from 14 countries discovered the presence of MPs, with vast majority of fibres (Kosuth et al., 2018). According to recent research, there are various type of polymers have been identified in tap water including polyethylene, polypropylene, polyvinyl chloride, polystryrene, polyphenylene sulfite and polyethylene terephathalate (Tong et al., 2020). The polyethylene and polypropylene comprised the highest concentration of these particles. The common shapes of MPs found in tap water are usually fragment, fibre, film, pellets, and sphere (Tong et al., 2020; Koelmans et al., 2019). The research of MPs in tap water was summarised in Table 2.8 and 2.9.

On a global perspective, there is still insufficient database on MPs contamination in tap water. From 2018 to 2021, the research regarding MPs in drinking water only conducted in 24 countries and only minimal amount of research addressing MPs in the country. Germany has the highest research of MPs addressing drinking water followed by China (Kirstein et al., 2021). The Figure 2.3 illustrated the MPs research in drinking water conducted in global. The MPs concentration reported in tap water varied from 0.001 to 903 MPs/L due to different analytical procedure performed as displayed in Figure 2.4. MPs is not a single defined chemical substance but rather a substance consisted of special families of polymeric substances which is traditionally different from other micropollutants (Kirstein et al., 2021). MPs may not only contain different structures and properties but also have various amount of additives added for better performance. Therefore, the MPs analysis remain challenging, as the significant differences between reported MPs in various study are owing to real difference among the applied method or just simply differences between the quantification limit, sampling and preparation method, accuracy of techniques, laboratory procedure contamination and so on.

Other than that, bottled water is also found contaminated by MPs. According to Table 2.10 and 2.11, more than 90 % of bottled water tested positive with MPs across 250 samples (Mason et al., 2018). By comparison, the plastic particles exist in bottled water is roughly twice as compared to tap water and fragment was the majority plastic particle morphology in bottled water (Mason et al., 2018). Furthermore, as the polyethylene terephthalate and polypropylene were the dominant polymer found in the

bottled water research, it suggests that the MPs contamination may come from the industrial process of bottling of water and abrasion of plastic from caps and bottlenecks during the opening and closing of bottles (Winkler et al., 2019, Mason et al., 2018). If an individual consumes 2 - 3 L of beverages per day including water, or drinks derived from tap water such as tea or juice, the daily dose of anthropogenic particles may up to 16 particles, which contribute to 5,800 particles consumption annually. Other than water consumption, individuals may uptake MPs from other sources such as MPs contaminated seafood, beer, and sea salt (Kosuth et al., 2018).



Figure 2.3: The MPs research for drinking water (Kirstein et al., 2021)

 Table 2.8: The case study summaries of sampling, sample pre-treatment method, identification/quantification method, and MPs

 abundance in tap water

Sampling	Sample	Sampling Method	Sample Pre-treatment	Identification/	MPs	Reference
Location	Туре		Method	quantification Method	Abundance	
Germany	Tap water from ground water	filtered through 3 μm stainless steel cartridge filter	Filtered with diluted HCl and perform density separation using ZnCl ₂ sln	Micro-FTIR	0.0007 MPs/L	(Mintenig et al., 2019)
Developed countries	Tap water	Glass bottle	Wet peroxide oxidation, PTFE filtration	FTIR	1.9 - 225 MPs/L	(Mukotaka et al., 2021)
Mexico	Tap water	Glass bottle	Filtered through nitrocellulose filter paper	Micro-Raman spectroscopy, SEM- EDS	18 MPs/L	(Shruti et al., 2020)
Denmark	Tap water	Stainless steel filter	Acetic acid	Stereomicroscopy, Micro-FTIR	0.58 MPs/L	(Strand et al., 2018)
China	Tap water	HDPE bottle	PC membrane filter, Nile Red dye conc. HCl digestion	Raman spectroscopy, fluorescence microscope	440 MPs/L	(Tong et al., 2020)
China	Tap water	Glass bottle	vacuum filtration with nitrocellulose membrane	Stereomicroscope, FTIR	0.7 MPs/L	(Zhang et al., 2020)
Brasilia, Brazil	Tap water	Laboratory amber bottle	Add 1.37 g/mL settle overnight, supernatant added to Nile red dye and 10 µg/mL methanol	Fluorescence microscope	194 MPs/L 438 MPs/L	(Pratesi et al., 2021)

Sampling	Sample volume	MPs Morphology	% of sample	Type of MPs	Reference
Location			containing MPs	identified	
Germany	1200 – 2500 L	Fragment	42%	PEST, PVC, PA, PE and epoxy resin	(Mintenig et al., 2019)
Various developed countries	500 mL per sample	Fragment > fibres > spherical	100%	PS, SEBS, PP, PES	(Mukotaka et al., 2021)
Mexico	3 L x 3 per site	Fibres > fragment	100%	PEST, epoxy resin	(Shruti et al., 2020)
Denmark	50 L per sample	Fragment	24%	PP, PS, PET	(Strand et al., 2018)
China	2 L per sample	Fragment > fibres > spheres	95%	PE, PP, PPS, PS, PET	(Tong et al., 2020)
China	4.5 L x 3 per site	Fibres > fragment	100%	Rayon, PET, PE, PS, PEST, PI	(Zhang et al., 2020)
Brasilia, Brazil	500 mL per sample	-	100%	-	(Pratesi et al., 2021)

Table 2.9: The case study summaries of sample volume, MP morphology, type of MP identified and % of sample containing MP in tapwater

				Tap wate	er
Reference	Sample type	MPs/L	Quanti-/Qualification	~~~~	
Wang et al., 2020	DWTP	930	SEM; RM		
Pivokonsky et al., 2018	DWTP	338 ± 76 - 628 ± 28	SEM; µ-FTIR; RM	100 - 1000	
Tong et al., 2020	houshold	440 ± 275	Nile red; RM	100 - 1000	
Pivokonský et al., 2021	DWTP	151±4	SEM; µFTIR; RM		
Kosuth et al., 2018	houshold	0-61	Rose Bengal; spatula le:		
Kankanige & Babel, 2020	houshold	56 ± 14; 21 ± 7; 13 ± 5; 6 ± 3 (dif. Size)	Nile Red; ATR-FTR; RM		
Shruti et al., 2020	public drinking water fountains	18±7	SEM-EDX; RM	10 - 100	
Pivokonský et al., 2021	DWTP	14±1	SEM; µFTIR; RM		N N
Sarkar et al., 2021	DWTP	2.75 ± 0.92	Nile Red; ATR-FTR		S
Zhang et al., 2020	houshold	0.7±0.6	ATR-µFTIR		
Lam et al., 2020	houshold	2.181 ± 0.165	Rose Bengal		
Kirstein et al., 2021	DWTP; pumping station; hydrant	0.174 ± 0.405	Py-GCMS; µFTIR		
Dalmau-Soler et al., 2021	DWTP	0.08 ± 0.04	µFTIR	0 = 10	
Pittroff et al., 2021	DWTP, houshold	0,066 ± 0,076	RM		
Johnson et al., 2020	DWTP	0.00011	μFTIR		
Mintenig et al., 2019	DWTP	0.0007	µFTIR		
Almaiman et al., 2021	n.i.	<loq< td=""><td>μFTIR</td><td></td><td></td></loq<>	μFTIR		
Weber et al., 2021	houshold; transfer station	0	RM		
Gomiero et al. 2021	DWTP	n.a. (0.0061 - 0.0931 µg/L)	Py-GCMS		
Kirstein et al., 2021	DWTP; pumping station; hydrant	n.a. [0.0014 - 0.00543 µg/L]	Py-GCMS; µFTIR		

Figure 2.4: The summary of studies investigating MPs in tap water (Kirstein et al., 2021)

 Table 2.10: The case study summaries of sampling, sample pre-treatment method, identification/quantification method, MPs morphology

 and type of MPs identified in bottled water

Sampling Location	Sample Type	Sample Pre-treatment Method	Identification/ quantification Method	MPs Abundance	Reference
Thailand	Bottled water (spring and tap)	Vacuum filtration with cellulose nitrate filter and cellulose filter Nile Red dye	Microscope, fluorescence microscope (< 50 μm), ATR-FTIR, confocal Raman spectroscopy	140 MPs/L	(Kankanige and Babel, 2020)
9 countries	Bottled water (table and mineral)	Nile red dye Vacuum filtered through glass fibre filter	Optical microscope, FTIR	325 MPs/L	(Mason et al., 2018)
Bavarian	Bottled water (mineral)	EDTA solution digestion, SDS sln added to increase sample homogeneity	Micro-Raman spectroscopy	2649 MPs/L	(Oßman et al., 2018)
Germany	Bottled water (mineral)	Gold coated polycarbonate filter	Micro-Raman spectroscopy	14 MPs/L	(Schymanski et al., 2018)
Germany	Bottled water (mineral)	Filtration with cellulose nitrate filter	Raman micro-spectroscopy	10 – 19 MPs/L	(Wiesheu et al., 2016)
Iran	Bottled water (mineral)	Vacuum filtered with glass fibre filter	Stereomicroscope, FTIR	8.5 ± 10.2 particles/L	(Makhdoumi et al., 2021)

Table 2.11: The case study summaries of sample volume, MP morphology, type of MP identified and % of sample containing MP in bottledwater

Sampling Location	Sample volume	MPs Morphology	% of sample containing MPs	Type of MPs identified	Reference
Thailand	10 brands, total 43.23 L	Fibre > fragment	100%	PET, PE, PP, PA, PVC	(Kankanige and Babel, 2020)
9 countries	11 brands, 500-600 mL per bottle	Fragment > fibres > films	93%	PP, Nylon	(Mason et al., 2018)
Bavarian	0.5 – 1 L per bottle	-	-	PET, PP, PE, Olefin, Styrene Butadiene	(Oßman et al., 2018)
Germany	700 – 1500 mL per bottle	Fragment	100%	PET, PP, PE, PA	(Schymanski et al., 2018)
Germany	3 L	Fibres	-	PET	(Wiesheu et al., 2016)
Iran	500 mL	Fragment > fibre	81%	PET, PS, PP	(Makhdoumi et al., 2021)

2.7 MPs analysis case studies in Malaysia

The MPs analysis in different environment context in Malaysia is shown in Table 2.12. Studies related to MPs contamination in Malaysia is scant, majority from coastal zone from Terengganu and Sarawak. Furthermore, the river studies in East Malaysia are mainly focus on heavy metal and none focus on MPs contamination (Choong et al., 2021). In addition, almost no studies in freshwater and coastal from West Malaysia. Although the studies and research regarding the occurrence of MPs in Malaysia consist of river, marine, sediment and MPs ingestion by biological organisms, however, the research related to abundance and distribution in tap water is empty. This indicates the MPs in tap and drinking water is still gaining less attention.

The research regarding MPs should continue until capable to fully understand the fate, morphology, pathway, and possible effects to human health, expanding the dataset of MPs contamination in marine, freshwater, drinking water and other environmental context (Campanale et al., 2019). Although faces challenges and obstacles, a lot of scientists have attempted to assess the MPs in global occurrence and distribution through research and studies (Zhang et al., 2017). All the research would be useful and contributed to the MPs distribution database over the globe.

 Table 2.12: The case study summaries of sampling, sample pre-treatment method, identification/quantification method, MPs morphology

 and type of MPs identified in different environmental context in Malaysia

Sampling	Sampling	Sample Pre-	Identification/	MPs Abundance	MP morphology	Reference
Location & Type	Method / volume	treatment	quantification	(density)	and type of MP	
of sample		Method	Method		identified	
Mangrove forest, Selangor (sediment)	Quadrat sampling at different depth of soil	Conc. Saline solution mix with sediment and pour to metal sieve	Binocular microscope	65 - 117 particles per quadrat	Fragment, film, pellet, foam	(Barasarathi et al., 2014).
Skudai and Tebrau River, Johor (sediment)	Box corer	Density separation, wet peroxide oxidation	Microscope	200 ± 80 particle/kg (Skudai) 680 ± 140 particle/kg (Tebrau)	Film	(Sarijan et al., 2018)
Santubong and Trombol Beach, Kuching (sediment)	Sampling quadrat	wet digestion	Electron microscope and FTIR	0.0358 ± 0.062 (Santubong), 1.7343 ± 2.173 particles/g (Trombol)	-	(Noik et al., 2015)

Kuala Nerus and Kuantan port	5.7 L calibrated steel sampler	Filtration and density separation	ATR-FTIR	0.13 - 0.69 particles/L,	Fragment PES, PS, PA, PVC	(Khalik et al., 2018)
(marine water)		•		0.14 to 0.15	PP, PE	
				particles/L		
Sungai Dungun,	200 µm mesh size	Hydrogen	Stereoscope	22.8 - 300.8	Fibre > fragment	(Hwi et al.,
Terengganu		peroxide	FTIR	particles/m ³	PP,	2020)
(surface water)					polyacrylonitrile,	
					rayon	
Langat River,	Manual collection	Filtration	FTIR	4.39 - 45.86	Fragment, films,	(Chen et al.,
Selangor Malaysia		30% hydrogen		particles/ L	beads	2021)
		peroxide			PET, HDPE,	
					LDPE, PP, EVA,	
					PVDC, PS	
Terengganu	Norpac net (60	Vacuum filtration	Stereomicroscope	3.3 particles/L in	Fragment, fibre	(Amin et al.,
coastal water	μ m) for capture	for surface water	and FTIR for	surface water,	PA	2020)
(surface water and	zooplankton, 60 L		surface water,	0.003 - 0.14		
zooplankton)	surface water		Quantitative	particles per		
	collected		analysis by	individual of		
			counting cell for	zooplankton		
			zooplankton			

Klang	River	95 gastropods over	Digestion	with	Stereomicrosc	ope	0.50	- 1.75	Fibre	(Zaki	et	al.,
estuary,	Selangor	12 sampling points	HNO ₃ and	H_2O_2 ,	ATR-FTIR		particles	s/g	PE-PDM, PES	2021)		
(gastropod	d)		Floatation	with								
			conc.	NaCl								
			solution									
Gastrointe	estinal	Trawled and	Dissection:	GIT	Infrared	and	9.88		PE, PP, ABS, PS,	(Jaafar	et	al.,
tract and	gills of	purchase	and gill ren	noved	Raman		particles	/individuals	PET	2021)		
commerci	al marine		and digest	with	spectroscopy							
fish			potassium									
			hydroxide	and								
			KOH sln									

CHAPTER 3

METHODOLOGY

3.1 Sampling

3.1.1 Sampling Point

The tap water samples were collected from three different points in Kampar, during the period November to January 2022. The three sources including conventional water tap from private household which is the private institutions which is Universiti Tunku Abdul Rahman (labelled as LW), Westlake residence in Kampar (labelled as HW), and the discharge of LAP Sungai Kampar water treatment plant (labelled as WW) as shown in Figure 3.1. Their coordinate information displayed in Table 3.1. These sampling point provide microplastic abundance from the aspect of provider-end which is the treatment plant and the user-end which is the consumer for household and institution.



Figure 3.1: The sampling location selected for microplastic analysis.

Location	Coordinate	Location Name
S 1	4°20'19.0"N 101°08'37.3"E	Universiti Tunku Abdul Rahman
		Kampar Campus
S2	4°19'59.2"N 101°07'54.0"E	Kampar Westlake Residence
S 3	4°22'15.4"N 101°09'49.6"E	LAP Sungai Kampar Water Treatment Plant

Fable 3.1:	Coordinate	of Sampling	Locations
		· · · · ·	



Figure 3.2: The LAP Sungai Kampar Water Treatment Plant



Figure 3.3: The water sampling cabinet at LAP Sungai Kampar Water Treatment Plant

3.1.2 Sample Collection

Each sampling glass bottle was cleaned thoroughly prior to the sample collection by using detergent and rinsed using ultrapure water. The sampling glass bottles were dried at drying cabinet of 70 °C for at least 24 h to remove moisture. During the sampling process, the water tap was opened and left to run for around 1 min before the sample was taken to prevent contamination of the first portion of tap water, and the 1 L glass bottle is filled to the point of overflowing (Koelmans et al., 2019). The sample are collected in 5 sets at each sampling point at three continuous days during the similar timeslot. Before filling the tap water, the glass bottles are rinsed with the water tap thrice before sampling to avoid contamination and residue inside the bottles. At the LAP Sungai Kampar water treatment plant, there are 3 sampling points which is raw water influent from Sungai Kampar (marked as RW), water effluent before chlorination (marked as SW) and chlorinated clean water effluent (marked as DW) at the water sampling cabinet. Only the samples from DW were collected.



Figure 3.4: The glass sampling bottles with labelling.

3.1.3 Sample Storage

The sample are preserved in glass bottles and retrieved back to the laboratory for further analysis. All samples are stored in fridge at 4°C to avoid direct sunlight prior to analysis. Furthermore, the glass bottles are covered by aluminum foil and ensured close tight to minimize the exposure to the environment which include the airborne contaminants (Sarjian et al., 2018).

3.2 Sampling Extraction and Digestion

The glass bottles were slowly inverted 3 times to ensure even distribution of the liquid. 200 mL of water sample was poured into separate beakers of 500 mL. Organic digestion was conducted by using 20 mL of 30% of hydrogen peroxide (H_2O_2 , Grade AR) which with the ratio 1:10 to the volume of sample (Wang et al., 2020). The samples covered with aluminum foil were then heated to 75°C for 1 hour and left at room temperature for 24 hours to digest remaining organic matter. The samples were stirred by using magnetic stirrer to ensure homogeneous heating throughout the whole samples. After the digestion process, the sample was then proceeded to staining process.

3.3 Microplastic analysis with Nile Red Staining Method

3.3.1 Nile Red Stain

Nile Red dye (9-diethylamino-5-benzo[a]phenoxazinone) staining is effective to stain hydrophobic polymers which binds itself to polymer and give fluorescence effect in the hydrophobic environment. It is the simple staining method for the identification of the hidden microplastic especially transparent microplastic (Uhl et al., 2018).

3.3.1.1 Preparation of Nile Red standard solution

The Nile Red stock solution (0.05 g/L) was prepared in acetone because of low solubility of Nile Red grain (Sigma-Aldrich, Germany) in acetone. The Nile Red working solution with concentration of 10 mg/L was then prepared by diluting the Nile Red stock solution with n-hexane (C₆H₁₄, Grade AR) (Shim et al., 2016). Higher concentration of dye increases the fluorescence intensity but at the same time also increased the background interference. Thus, working solution with 10 mg/L (10 μ g/mL) provide satisfactory visibility, speed, and background effect (Maes et al., 2017). The Nile Red solution is transferred to amber glass bottle and kept inside the fridge at 4°C and covered with aluminum foil to prevent the sunlight exposure as shown in Figure 3.5.



Figure 3.5: The Nile Red working solution

3.3.1.2 Staining process

Approximately 2 drops of Nile Red standard solution added into the samples and left for 24 hours to ensure homogeneity of the solution. Fluorescence intensity of the dye increased swiftly with incubation time however it plateaued within 30 - 60 minutes and remained almost unchanged up to 66 hours (Maes et al., 2017). After that, the sample was passed through MCE membrane filter (mixed cellulose ester) (47 mm Ø with 0.45 μ m pore size) facilitated by vacuum filter apparatus. Quadrants were drawn on the with approximately 2 mm x 2 mm on each grid as shown in Figure 3.6. The vertical axis was labelled with number across the columns while horizontal axis was labelled with alphabets across the rows. The membrane filter placed in clean capped glass petri dish with aluminum weight boat and dried at room temperature for 24 h to remove moisture. The microplastic retained on the membrane filter was ready for the visual sorting.



Figure 3.6: The filter paper drawn with quadrant and numerical horizontal and alphabetical vertical label.

3.4 Microplastic Analysis

3.4.1 Microplastic Inspection by Fluorescence Microscope

After 24 hours, the dried membrane filter was cut in half and prepared in microscope glass slide as displayed in Figure 3.7. The microplastic particles filtered on the membrane were determined and photographed under the fluorescence microscope (Olympus BX51) with magnification range of 0.8x - 4x by using the Infinity 1-3C camera and Infinity Analyze and Capture application from Lumenera Software, which shown in Figure 3.8 to measure the size of all MPs particle found. The microplastic was examined under green (dichroic mirror: DM500; excitation filter: BP 450 - 480; barrier filter: BA 515) and red mode (dichroic mirror: DM 570; excitation filter: BP 510 - 550, barrier filter: BA 590). The stained microplastic were able to be observed fluorescing in both green and red color. The MPs were captured, counted, and classified into 6 size range: < 1 μ m, 1 < x < 5 μ m, 5 < x < 10 μ m, 10 < x < 20 μ m, 20 $< x < 100 \ \mu m$ and $> 100 \ \mu m$. The shape of the microplastic was classified into three morphotypes: (1) fragment (FM), (2) fiber (FB) and (3) sphere (SP), with fragment being irregular shaped, fiber being rod-shaped and sphere being round-shaped. The size of MPs was measured by its diameter length for fragment and sphere whilst fiber was measured by its length between 2 points.


Figure 3.7: The filter paper prepared in glass slide with label



Figure 3.8: The fluorescence microscope with camera

3.4.2 Scanning Electron Microscopy with Energy-Dispersive X-Ray Analysis (SEM-EDX)

SEM-EDX provides high-resolution images of the magnified MPs particles at the same time capable to provide elemental composition signatures which is the qualitative information related to the chemical composition of the investigated particles. It scanned the surface of the particle with electron beams which the contact between the beam and the surface generates secondary electrons and element-specific X-ray radiation. From each source, 1 sample were selected by optical microscopy for SEM-EDX analysis. The selected grid that contained MPs was cut and prepared in slide to prevent loss of sample due to extremely small size of particle as exhibited in Figure 3.9. Samples were magnified at 2,000 - 10,000x by using back scattered electron (BES) detector of the SEM.



Figure 3.9: The sample for SEM-EDX analysis



Figure 3.10: The SEM-EDX

3.4.3: Attenuated Total Reflectance (ATR) Analysis

For the functional group identification of the polymer, 5 specimens from each source which contain MPs were analyzed by ATR-FTIR spectrometer (PerkinElmer). The background data was obtained before every sample spectrum. The diamond crystal of ATR and the probe were cleaned with cotton pad by using isopropanol in one direction to avoid any contamination. The filter paper was inverted to allow the MPs touched the diamond crystal and subsequently screwed the probe with 79 – 80 force gauge to ensure good surface contact as suggested by Perkin Elmer (Jung et al., 2018). The spectra were observed over wavelengths ranging from 4000 cm⁻¹ to 400 cm⁻¹ by averaging 4 scans and 32 scans. The spectrum was analyzed by using Perkin Elmer Spectrum software. The data obtained was then proceeded for further interpretation and analysis.



Figure 3.11: The diamond crystal probe of ATR-FTIR



Figure 3.12: The ATR-FTIR

3.5 Quality Control

To avoid man-made and laboratory contamination, all the plastic appliance replaced with clean glassware (Lv et al., 2021). Only pure virgin cotton cloth and Nitrile-Butadiene gloves were worn during the laboratory analysis. The blanks sample is prepared by using 1 litre of ultrapure water filtered into a glass bottle (Tong et al., 2020). All glassware were thoroughly rinsed with ultrapure water (18.2 MOhm) to prevent sample contamination, and except for during the transfer of sample to filtration

process all the sample were always covered with aluminium foil (Stanton et al., 2019). The glassware was properly stored in clean cabinet and their sensitive surfaces and openings were covered with aluminium foil to avoid air contact until put into service (Uhl et al., 2018). In addition, all the glassware were acid washed by using 0.15 % of hydrochloric acid and rinse with a large bath of distilled water before experiment (WebPath, n.d.). The laboratory work was conducted on laminar flow working bench with regularly cleaned with 75 % ethanol which for instance, has been shown no effect to the characteristics of polymer (Courtene-Jones et al., 2017).

3.6 Statistical Analysis

The microplastic concentration in each sample was calculated by dividing the MPs counted with the total volume of water sample which reported as total MPs particles per litre (MPs/L). The microplastic abundance was reported as the mean \pm standard deviation. The data was tabulated in Microsoft Excel and One-way ANOVA test was conducted to recognise the differences in the microplastic abundance.

3.7 Schedule for Overall Research

The Flow chart and Gantt chart for overall study summarized in Figure 3.13 and 3.14 respectively. Water sampled from 3 different sources eventually digested by using hydrogen peroxide and stained with Nile Red dye. Then, all samples undergone fluorescence microscope analysis and selected sample were analysed by using SEM-EDX and ATR-FTIR.

According to Figure 3.12, the study began with project title analysis and preliminary report writing including introduction, literature review and methodology during the period of June to August 2021. After the first stage report submission and oral presentation conducted in September 2021, the preliminary laboratory test started on late October to November 2021. The preparation including equipment preparation and setup of laboratory work. After that, actual sample analysis conducted during the period of November to February 2022. Data compilation and interpretation were conducted along the sample analysis while discussion with supervisor was done whenever discussion was necessary. After completion of laboratory work, the report writing of remaining chapters was done during the March and April 2022 and completed in late of April. The project poster and presentation slide were prepared, and the final report was compiled and submitted at the early of May 2022.



Figure 3.11: The flowchart of overall research

YEAR				2021						2022		
MONTH	JUN	JUL	AUG	SEP	OCT	NOV	DEC	JAN	FEB	MAR	APR	MAY
TASK / ACTIVITY			•									
1.0 Preliminary Title Analysis												
1.1 FYP Title analysis												
1.2 Case studies Reviews												
2.0 Preliminary Report Writing				_								
2.1 Chapter 1: Introduction			_									
2.2 Chapter 2: Literature Review												
2.3 Chapter 3: Methodology				-								
3.0 FYP 1 Presentation												
3.1 Completion of FYP 1 report												
3.2 Progress Report												
Discussion with Supervisor												
4.0 Preliminary Lab Test							_					
4.1 Setup of laboratory work												
4.2 Sample & equipment preparation												
5.0 Actual Sample Analysis									_			
5.1 Water sampling										_		
5.2 Sample analysis												
6.0 Report Writing												
6.1 Chapter 4: Results and Discussion												
6.2 Chapter 5: Conclusion & recommendation												
7.0 FYP 2 Presentation												_
7.1 Completion of thesis writing												
7.2 Oral and poster Presentation												
7.3 Submission of Final Report												



CHAPTER 4

RESULTS AND DISCUSSIONS

4.1 Nile Red Staining effect of MPs under different wavelength

Before the actual analysis was performed, preliminary test was conducted to reduce possible laboratory error. The water sample was analyzed with both green and red fluorescence light with different excitation and emission wavelengths. The Figure 4.1 illustrated the comparison of microplastics detected under both green and red fluorescence light. Figure 4.1 (a), (b) and (c) shown the sphere, entangled fiber and fragment of MPs which were observable under both green and red fluorescence. Figure 4.1 (d), (e) and (f) exhibited the long fiber of MPs however the long fiber MPs was not observable in red fluorescence shown in Figure 4.1 (f).

The results showed that microplastics were visible under both lights but the some of the background staining under red light was bright and may cause difficulties in observing microplastic. This led to overestimation or underestimation of the abundance of microplastics in the water samples. For instance, the fiber type of MPs was barely detected or non-detectable in red as shown in Figure 4.1 (f). However, it is more visible under green fluorescence. Red fluorescence may have strong background interference. Furthermore, the strong red glare causes eye strain while observing the microplastic especially during a long-time analysis. In comparison, the microplastic detected under green light observable and provide clearer image which improve

readability to detect the microplastics. Thus, green light was selected for subsequent analysis.

Nile red dye is lipophilic fluorophore to adsorb onto the surface of lipophilic compound such as fats, cell membrane and plastic. In other words, it not only can be used to stain neutral lipids in biological components, but also can stain synthetic polymers such as plastic due to its origin of hydrocarbon derived from petroleum which is hydrophobic (Shim et al., 2016). The dye attached on the surface of the components generate sensitive bright which can be detected under fluorescence microscopy (Scircle and Cizdziel, 2019). Yellow and red fluorescence are generally applied in biological samples analysis, while the peak emission fluorescence ranged from red to green with rising hydrophobicity of targeted hydrophobic component (Diaz et al., 2008). The Nile Red fluorescence show peak effect at 576 nm in n-hexane solution (Elsey et al., 2007).





Figure 4.1: Comparison of Images between Green and Red Fluorescence (a) Sphere (b) Entangled Fiber (c) Fragment (d) Fiber (e) Fiber (f) Fiber of MPs observed under Fluorescence Microscope

4.2 MPs staining properties

All the MPs were effectively stained and identified under the staining condition and the concentration of the Nile Red working solution (10 mg/L) provide adequate bright green fluorescence for the MPs particles. According to Figure 4.2, the MPs appeared as barely detectable, dim green glow, and strong glow. This is due to different shape of MPs tended to be stained differently thus result in different fluorescence effect. Furthermore, the color of the plastics can influence the fluorescence effect due to it can affect the intensity of emission. The different weathering condition of the MPs also result in different surface roughness and stainable surface area for the Nile Red dye.

Nile Red favors the detection of highly hydrophobic components at short excitation wavelength between 450 – 500 nm (green fluorescence) compared to lipid with more neutrality, which performed better visual at higher excitation wavelength (red) with 515 – 560 nm. This is because different type of plastic with different density will affect the fluorescent effect as more hydrophobic plastic such as PE and PP will give strong fluorescent effect (Figure 4.2 (c)) than less hydrophobic such as PET, while they are not effectively stained by Nile Red as their fluorescent effect performed weak, barely detectable (Figure 4.2 (a)) or even absent in longer excitation wavelength (Figure 4.1 (f)). Nile Red protocol limits for less hydrophobic and higher density plastic such as PC, PUR, PET, PES, PA microfibers and PVC with density more than 1.2g/cm³ but effectively applied in polymer such as PE, PP, PS and nylon 6 with density less than 1.08 g/cm³ as they fluoresced strongly (Erni-Cassola et al., 2017; Shim et al., 2016).



Figure 4.2: Nile Red staining effect in MPs (a) Dim Green Glow (b) Barely Detectable Glow and (c) Strong Glow

4.3 The MPs Mean Abundances

The MPs were observed in all 45 tap water samples tested, indicating a 100 % MPs occurrence in the studied area. The Figure 4.3, 4.4 and 4.5 shown the concentration of MPs obtained in source 1 to 3 which the sources from private institution, household, and water treatment plant respectively whilst the Table 4.1 illustrated the mean concentration of MPs projected to 1 L sample volume (MPs/L), and its overall plot shown in Figure 4.6. In addition, the MPs in blank samples are negligible thus no correction were done for the background contamination. The ANOVA test presented in Appendix A shown that the *p* value of < 0.05 was determined. Thus, it had an obvious difference between the mean of the 3 groups of data, whereas there was no statistical difference between the concentration of MPs in source 1 and 2 which has similar concentration (p > 0.05) shown in Appendix B. In addition, both group of sources 1 and 3 and source 2 and 3 displayed significant differences in statistic with p < 0.05 as illustrated in Appendix C and D.

From the overall perspective, the range of MPs within all tap water samples were 85 to 335 MPs/L. It can be told that the MPs presented in water sample source 1 and 2 (receiver) are slightly less than the source 3 which obtained from the provider side. The concentration of MPs in source 1 and 2 showed similar result which is 169.7 \pm 47.5 MPs/L and 179.3 \pm 49.7 MPs/L respectively while the abundance of MPs in source 3 were 238.7 \pm 48.3 MPs/L.

The MPs from outflow of water treatment plant might loss in the gravity flow and fractured into smaller pieces during the water distribution transportation along the piping network system. Thus, the receiver of tap water from household or institution receive less MPs compared to the effluent of raw water treatment plant due to dispersed MPs along the transportation. The occurrence of MPs in treated tap water was based on the types of source water bodies and the anthropogenic activities nearby it, atmospheric deposition, and mechanical abrasion of plastic-lined piping system (Lam et al., 2020). High occurrence of MPs is expected to be found in open surface water bodies than groundwater due to less available to natural sand filtration. In this case, the tap water in Kampar area is sourced from the Sungai Kampar which is open surface and more prone to airborne contamination which is believed to represent a possible route of MPs to raw water (Lam et al., 2020). Furthermore, the presence of MPs in tap water may be due to the non 100 % removal of plastic particles during the water treatment system and the possibility of polymer particles in coagulant released to the effluent due to high shearing forces. The residue of the coagulant, PAM might remain in the treated water as well as the leaching of polymer in the polymeric membranes applied in filtration process due to ageing (Oßmann, 2021). The type of treatment process is likely to affect the occurrence of MPs in tap water as floatation seems to be efficient for MPs removal as plastic polymer due to its buoyancy and light characteristic (Di and Wang, 2018). However, the plastic removal efficiency is not the focus in this study, further and additional research were required to justify these unknown.

Therefore, the prevalence of MPs found in drinking water was significant and it's an unneglectable input of MPs to human body (Lam et al., 2020).

Water sample	Name of source	Mean Concentration of MPs
source		(MPs/L)
S 1	Private institution	169.7 ± 47.5
S 2	Household	179.3 ± 49.7
S 3	Water treatment plant	238.7 ± 48.3

Table 4.1: Mean concentration of MPs in 3 sources of water samples



Figure 4.3: The concentration of MPs in water sample from private institution (S1)



Figure 4.4: The concentration of MPs in water sample from household (S2)



Figure 4.5: The concentration of MPs in water sample from water treatment plant (S3)



Figure 4.6: The overall plot with 3 different water sampling source and its average concentration of MPs with error bar

4.3.1 The Size Distribution of MPs

In this study, there were six category of size classes divided to classify the size of MPs (< 1 μ m; 1 - 5 μ m; 5 - 10 μ m; 10 - 20 μ m; 20 - 100 μ m; > 100 μ m). The particle size distribution was displayed in Table 4.2.

The size of MPs ranged from 0.5 - 183.9 μ m, and no MPs larger than 200 μ m were detected in all 45 tap water samples. The percentage of different size of MPs illustrated in Figure 4.7. Generally, MPs in the five out of six-size category of < 1 μ m, 1 - 5 μ m, 5 - 10 μ m, 10 - 20 μ m and 20 - 100 μ m were determined in all tap water sample sources. MPs size of 1 –5 μ m was predominated in majority of the tap water sample which ranged from 49.4 – 71.1 %. The second dominant group was that of < 1 μ m which comprising approximately 12 – 25 % of MPs and followed by 5 – 10 μ m (roughly 7 – 22 % of total MPs). The occurrence of MPs decreased with increasing size category as MPs larger than 100 μ m were the least, which only comprising not more than 1% of total MPs found. Furthermore, only minimum of MPs ranged from 20 – 100 μ m and 10 – 20 μ m were observed with percentage from 1.5 – 7.0 % and 1.5 – 9.0 % respectively. This indicates the LAP Sungai Kampar water treatment plant able to remove almost all the large MPs.

From the overall perspective, MPs less than 10 μ m in size were major in tap water which comprised of 83.1 – 97.3% of total MPs. According to Table 4.3, the size of the MPs obtained for source 1 – 3 were 8.4 ± 4.9 μ m, 2.7 ± 0.3 μ m and 4.4 ± 1.6 μ m respectively. The smaller size of MPs obtained in source 2 may be due to the filtration system installed in the household which able to captured the large MPs thus only the remaining small MPs will escape from the filter membrane. The larger size of MPs was found in source 1 was due to more large fibre shaped of MPs were determined thus contributed to larger mean size. The increasing size of MPs for each source was displayed in Figure 4.8.

The nano plastic was captured in this study due to the membrane filter with pore size of 0.45 μ m were applied which able to retain the nano plastic on it. Although the microplastic less than 1 μ m was considered nano plastic which was not the main interest in this study, however the abundance of nano plastic comprised from 11.6 –

24.6% of the total microplastic found were not negligible and should not be ignored. This is because MPs smaller than 150 μ m might penetrate through the gut epithelium wall and translocate into human tissue which might be toxicological relevant and the MPs size less than 1.5 μ m may enter deeply into organ (Oßmann, 2021; EFSA Panel on Contaminants in the Food Chain (CONTAM), 2016). In addition, due to technical limitation such extreme small particle was difficult to be analysed for chemical composition and hardly to be observed during the microscopic analysis. Thus, it remained a challenge in the determination of nano-sized MPs in tap water. This founding even proved the importance of microplastic study and continual effort were required in this sector.

The extreme tiny size of MPs found in this study was expected to escape from the tiny pores of the filtration membrane during the final filtration process in the treatment plant thus contribute to significant number of MPs in tap water. However, the bigger size of fiber observed in this study suggested tap water might be contaminated by large MPs in the water storage facility and water piping.

Size Range	Size Distribution (MPs/L)						
-	Private institution	Household	Water treatment				
	(81)	(S2)	Plant (S3)				
> 100 µm	0.3 ± 0.2	0	0				
$20 < x \leq 100 \ \mu m$	2.4 ± 1.9	0.5 ± 0.1	1.2 ± 0.5				
$10 < x \le 20 \ \mu m$	3.1 ± 2.7	0.5 ± 0.3	1.5 ± 0.8				
$5 < x \le 10 \ \mu m$	7.5 ± 4.1	2.5 ± 1.0	3.7 ± 1.1				
$1 < x \le 5 \ \mu m$	16.5 ± 6.0	23.8 ± 5.2	34.3 ± 9.3				
< 1 µm	4.1 ± 3.8	10.9 ± 3.9	7.1 ± 1.7				

 Table 4.2: The Mean Concentration of MPs in 3 different sources according to

 the size range



Figure 4.7: Size Distribution of MPs in 3 different sources

Table 4.3:	The Mean	Size of	f each	water	sample	sources
	I ne nitean		. vavn		pre	Sour ces

Water Sample	Mean Size (µm)
<u>S1</u>	8.4 ± 4.9
S 2	2.7 ± 0.3
S 3	4.4 ± 1.6





(b)



Figure 4.8: The MPs size distribution in 3 sources of tap water sample (a) S1 (b) S2 and (c) S3

4.3.2 The Morphological Characteristics of MPs

The morphotype of fragment and sphere were observed in all 45 tap water samples, however fiber only observed in 53% of the total samples (24 samples out of 45). Among all three morphologies of MPs, fragment was the most dominant morphotype in this study which ranged from 55.3 - 60.0 %. Sphere comprised of substantial amount also which is the second abundant morphotype ranging from 37.1 - 42.6 %, whilst the fiber was the least ranging from 1.0 - 3.4 %. Table 4.4 illustrated the mean concentration of MPs shape in three different tap water sample sources and the overall result in percentage was further summarized in Figure 4.9. Interestingly, there were also several types of shapes determined in this study such as oval, fragment with different glow, long fragment, thin film, rod-liked or thread-liked fragment, rectangular and irregular shaped of MPs as illustrated in Figure 4.10 - 4.20 below.

Fragment type of MPs were believed to originate from breakdown or weathering of the bigger plastic debris into small one such as plastic bag, packaging, and mulching film. Sphere like MPs were expected to originate from cleaning products such as microbeads and exfoliants while fiber was believed to originate from discharge of laundry drain or airborne contamination (Chanpiwat and Damrongsiri, 2021).

Type of MPs shape	Shape Distribution (MPs/L)						
Type of MTS shape –	S1	S2	S 3				
Fragment	95.0 ± 31.2	102.0 ± 21.8	145.3 ± 44.2				
Sphere	68.7 ± 26.4	75.7 ± 5.8	86.0 ± 2.7				
Fiber	6.0 ± 7.0	1.7 ± 1.5	7.3 ± 4.2				

 Table 4.4: The shape distribution of MPs in three respective tap water sources



Figure 4.9: The percentage of MPs of different morphological characteristics for 3 different water sources



Figure 4.10: The sphere MPs with size (a) < 1 μ m and (b) > 1 μ m



Figure 4.11: The oval shaped MPs with size (a) $< 5 \mu m$ and (b) $> 5 \mu m$



Figure 4.12: The fragment of MPs with size (a) $< 5 \mu m$ and (b) $> 5 \mu m$



Figure 4.13: The fragment of MPs with blurry glow



Figure 4.14: The fragment of MPs with inconsistent glow



Figure 4.15: The long fragment of MPs with size (a) \leq 5 µm and (b) > 5 µm



Figure 4.16: The rectangular shaped fragment of MPs



Figure 4.17: The thin film fragment of MPs



Figure 4.18: The long MPs (a) rod-liked and (b) thread-liked



Figure 4.19: The irregular shaped of MPs with size (a) < 5 μ m, (b) 5 – 10 μ m and (c) > 20 μ m



Figure 4.20: The fiber MPs (a) entangled and (b) long fiber

4.4 SEM-EDX Identification

The Figure 4.21 illustrated the Scanning Electron Microscope (SEM) image of blank and 3 selected samples from respective sources in 10,000 magnifications displayed in Figure 4.22 – 4.24. The elemental composition was tabulated in Table 4.5 and their further details was shown in Appendix E. The Figure 4.21 exhibited a clear blank membrane filter with no particle stained above indicated no background contamination. In the EDX analysis, only Carbon (C) and Oxygen (O) presented in the blank sample.

The SEM image for 3 selected samples illustrated different surface characteristics. The MPs in Figure 4.22 showed the cracking and rough surface of the MPs with gouges and ragged edge while Figure 4.23 shown rough and uneven polymer surface and Figure 4.24 showed smooth and wrinkled surface. Those surface texture and characteristics were believed to be formed from by such as fragmentation of larger particle during the transportation, aging of polymer, and caused by environmental exposure such as UV degradation (Wang et al., 2017). Other than that, the mechanical and atmospheric wearing can cause grooves or gouges on the polymer surface, which was discovered in this study (Zbyszewski et al., 2014). Weathered MPs not only experienced changes in structure and shape, increased of roughness but also changes in chemical composition which also known as chemical weathering, such as development of carbonyl group or hydroxyl group due to increase of polarity (Tu et al., 2020). This might be due to highly exposure to UV or oxygen availability (Cai et al., 2017). Over the time, the surface of MPs with roughness and delamination available for microbial colonization or adsorption of hydrophilic pollutants which poses potential hazard on organism (Ding et al., 2019).

According to Figure 4.25, the strong chlorine peaks in the EDX spectrum and relatively uneven polymer texture were believed yielded by the PVC polymer as illustrated in Figure 4.25 (c) and corresponded to Figure 4.23. Specific composition of C indicated polymeric origin and clear ratio of C:O corresponding to PET as shown in Figure 4.25 (b) and (d) (Winkler et al., 2019). Plastic particle such as PP and PE also have strong carbon peak without any other distinct EDX peak. There was trace element identified in the MPs such as aluminium (Al), sodium (Na), silicon (Si) which can be found in the additives in the plastic manufacturing or elements attached onto the MPs surface (Miloloža, 2020). Furthermore, the EDX analysis also exhibited the evidence of adsorbed heavy metals such as Copper (Cu), Zirconium (Zr), Zinc (Zn) and stannum (Sn) on the MPs. However, these metals exhibited very low concentration mostly due to additives to provide unique features or to decrease the surface coefficient of friction on the polymer surface (Ranjan et al., 2021). Those metals added into plastic manufacturing provide various function such as UV stabilizer, colour pigment, stabiliser, catalysts, or antimicrobial material (Turner, 2016).

SEM coupled with EDX analysis able to identify carbon-dominated polymer from inorganic substances. In addition, EDX not only can identify the elemental signature but also can determine the foreign substances or potential hazardous heavy metals adsorbed onto the MPs surface. Thus, this elemental analysis shown all the samples tested to be polymeric material.

In short, SEM with EDX was the powerful instrument to obtain highmagnification image of MPs and details elementary information to distinguish MPs from non-plastic However, it had limitations such as expensive, time-consuming, and required competent technician thus it was not suitable for bulk sample analysis (Chen et al., 2020). In addition, SEM analysis could not identify discolouration of MPs and no differentiation of elemental details between additives of MPs and adsorbed substances on the surface of MPs (Silva et al., 2018)



Figure 4.21: The SEM image of blank membrane filter in x10,000 magnification



Figure 4.22: The SEM image of MPs in private institution (S1) in x10,000 magnification



Figure 4.23: The SEM image of MPs in household (S2) in x10,000 magnification



Figure 4.24: The SEM image of MPs in water treatment plant (S3) in x10,000 magnification


Figure 4.25: The EDX spectrum for (a) blank (b) S1 (c) S2 and (d) S3

Sample	Figure	Elements
(a) Blank (control)		C and O
(a) S1		C, O, Na, Al, Si, Cu, Zn, Zr and Sn
(b) S2		C, O, Na, Al, Cl, Cu and Zn
(c) S3		C, O, Si, Cu and Zn

Table 4.5: SEM-EDX Analysis of MPs in Blank and 3 selected samples

4.5 ATR analysis for Polymer Functional Group Verification

In this study, total of 15 samples were studied which 5 selected MPs sample from each tap water sources. The result obtained from the ATR-FTIR analysis was interpreted and compared of the sample spectra with several literatures and reference spectra of the polymer (Jung et al., 2018; Müller et al., 2009; Lobo and Bonilla, 2003). The Figure 4.26 shown the ATR-FTIR spectra for averaging 4 scans with no differences were observed. In comparison, the spectra with averaging 32 scans able to provide several differences which indicate effective and adequate scanning for the MPs. Thus, only the results with 32 scans will be considered. The Figure 4.27, 4.28 and 4.29 illustrated the ATR-FTIR spectra for MPs from S1, S2 and S3 respectively with 32 scans.

The virgin membrane filter was used as the control blank and the spectrum identified included: C=O stretch (1743 cm⁻¹), C=C stretch (1644 cm⁻¹), CH₂ bend (1374 cm⁻¹), NH bend, C-N stretch, or C-O stretch (1278 cm⁻¹), Aromatic CH bend or C-O stretch (1059 cm⁻¹), CH₂ rock or C-CH₃ stretch (835 cm⁻¹), CH₂ rock (748 cm⁻¹) and Aromatic CH out of plane bend (684 cm⁻¹). Thus, the similar spectrum of the sample would be eliminated from the results when compared to the blank spectrum. The additional spectra for each sample were summarised and tabulated in Table 4.6.

According to the Table 4.6, the S1 (b), S1 (e), S3 (c) and S3 (d) exhibit similar spectrum with broad N-H stretch (3373 cm⁻¹), border and shaper for functional group in the finger region which is aromatic C-H, C-O stretch, or alcohols (1032 cm⁻¹) and additional spectrum of aromatic ring out of plane bend (537 cm⁻¹). The spectrum corresponds to the reference spectrum of PS or PA (Nylon). The spectrum 472 cm⁻¹ was not taken into consideration due to limited library and information related.

Furthermore, the MPs sample of S1 (d), S2 (e) and S2 (f) illustrated similar spectrum with additional spectrum in the fingerprint region such as C-Cl stretch (612 cm⁻¹) and aromatic ring out of plane bend (540 cm⁻¹). The C-Cl stretch indicate the presence of chloride which is PVC in the MPs. Besides, the additional ambiguous spectra of 471 cm⁻¹ in the fingerprint region of might due to the additives or adhesive

added into the plastic during the manufacturing process (Chércoles Asensio et al., 2009).

According to the result obtained, there were only 7 samples out of the 15 samples successfully obtained the spectra while the rest remained undistinguished with the blank. This is because of the ATR-FTIR analysis required surface contact between the probe and the MPs. The pressure exerted on the MPs by the probe made of hard diamond crystal may damage the fragile or weathered MPs, as well as being pulled from the membrane filter by adhesion force or electrotactic force which cause undefined result (Shim et al., 2016). In addition, the interpretation of infrared spectrum especially the peaks of specific functional group were time-consuming and required practical experiences in order to obtain accurate interpretation (Lobo and Bonilla, 2003). The MPs obtained in this study with mean size ranged from 2.7 \pm 0.3 to 8.4 \pm 4.9 μ m require several trails and hard to achieve clear spectra for accurate interpretation due to extremely small size (Shim et al., 2016). In addition, the polymer may suffer from environmental degradation which may induce alteration in polymeric chain and functional group, as well as contact with impurities to affect its absorption bands (Barbes et al., 2014).

Sample	Absorption band (cm ⁻¹)	Assignment
Control	1743	C=O stretch
	1644	C=C stretch
	1374	CH ₂ bend
	1278	NH bend, C-N stretch, C-O stretch
	1059	Aromatic CH bend, C-O stretch
	835	CH ₂ rock, C-CH ₃ stretch
	748	CH ₂ rock
	684	Aromatic CH out of plane bend
S1 (b)	3370 (broad)	N-H stretch
	1032 (broad and shaper)	Aromatic C-H, C-O stretch, alcohols
	537	Aromatic ring out of plane bend
S1 (d)	612	C-Cl stretch
	540	Aromatic ring out of plane bend
S1 (e)	3373 (broad)	N-H stretch
	1032 (broad and shaper)	Aromatic C-H, C-O stretch, alcohols
	536	Aromatic ring out of plane bend
S2 (e)	623	C-Cl stretch
	543	Aromatic ring out of plane bend
S2 (f)	620	C-Cl stretch
	541	Aromatic ring out of plane bend
S3 (c)	3373 (broad)	N-H stretch
	1032 (broad and shaper)	Aromatic C-H, C-O stretch, alcohols
	536	Aromatic ring out of plane bend
S3 (d)	3377 (broad)	N-H stretch
	1033 (broad and shaper)	Aromatic C-H, C-O stretch, alcohols
	539	Aromatic ring out of plane bend

Table 4.6: List of absorption bands (cm⁻¹) for ATR-FTIR spectra with its respective functional group for blank and samples



Figure 4.26: The Comparison of ATR Spectra between (a) Control and MPs from (b) S1, (c) S2 and (d) S3 (4 scans)





Figure 4.27: The Comparison of ATR Spectra between (a) Control and MPs from S1 (32 scans)





Wavenumber (cm⁻¹)

Figure 4.28: The Comparison of ATR Spectra between (a) Control and MPs from S2 (32 scans)



Wavenumber (cm⁻¹)



Figure 4.29: The Comparison of ATR Spectra between (a) Control and MPs from S3 (32 scans)

CHAPTER 5

CONCLUSION AND RECOMMENDATION

5.1 Conclusion

In this study, the quantification and characterization of MPs in three different sources of tap water in Kampar distinct which included the private institution (S1), household (S2) and LAP water treatment plant (S3) was achieved. The average concentration of MPs in S1 and S2 which from the end user has no significant differences (p > 0.05) and their concentration ranged from 169.7 ± 47.5 and 179.3 ± 49.7 MPs/L respectively. However, the higher abundance of MPs in provider end (S3) was observed which ranged from 238.7 ± 48.3 MPs/L. This might due to the MPs diverted and fractured into smaller particle during the water distribution system along the piping network.

The size of MPs observed in the tap water ranged from $0.5 - 183.9 \,\mu\text{m}$ with no MPs bigger than 200 μm observed in all tap water samples. MPs size category of 1 $-5 \,\mu\text{m}$ was predominant in all the tap water samples which ranged from $49.4 - 71.1 \,\%$. Overall, majority of MPs which up to $83.1 - 97.3 \,\%$ were less than 10 μm in size. MPs larger than 100 μm was the least which consisted only not more than 1 % of total MPs. The MPs ranged from 20 - 100 μm also observed ranged from 1.5 - 7.0 %. Interestingly, the nanoplastic which smaller than 1 μm was observed in the tap water sample which comprised of $11.6 - 24.6 \,\%$ of total MPs found and should not be neglected as it possessed potential hazard to human body. Fragment was the dominant morphotype observed in the total MPs which up to 55.3 - 60.0 % followed by sphere which comprised 37.1 - 42.6 % whilst fiber was the least which only consisted of 1.0 - 3.4 % of total MPs. The fragmentation of MPs may lead to substantial amount of fragment shaped MPs in tap water sample while sphere was believed originate from microbeads of consumer product and fiber was originated from laundry drain or airborne contamination. There were also irregular, thin film, rod-liked and thread-liked shaped of MPs observed in this study.

SEM-EDX able to provide high magnification of MPs which displayed different surface characteristic such as rough, wrinkled, and smooth. The surface texture was believed due to the weathering of MPs due to environmental exposure. The type of plastic determined in EDX was PET and PVC. There was also possibility the PET could be PP or PE due to strong carbon peak. PVC was ensured due to strong chlorine peak observed. Heavy metals and trace element found in little concentration may be due to additives added for plastic features. Furthermore, in the ATR-FTIR analysis, 32 scans were able to provide efficient scanning compared to 4 scans. The spectrum corresponds to PS or PA, and PVC was also found due to C-Cl stretch. The PVC particle was observed in both EDX and ATR analysis and indicated it may originate from the wearing of piping network which made from PVC and added into the water during the transportation.

5.2 Recommendations

The below are the recommendations proposed for future study of MPs.

- 1. Adopt larger sample size across several sampling point for better determination of the concentration of MPs in the studied area.
- Sampling at different cities across the whole country of Malaysia is necessary to provide basic database of MPs of drinking water in Malaysia.

- 3. Apply Raman or micro-Raman spectroscopy for identification the type of MPs down to 1 μm in future study due to the limitation of ATR-FTIR analysis.
- 4. Filter all added chemical solutions and deionized water through syringe membrane before the laboratory procedure because the chemicals generally stored in plastic container.

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APPENDICES

Appendix A One Way ANOVA Test for 3 sources of tap water sample

Anova: Single Factor

SUMMARY

Groups	Count	Sum	Average	Variance
Column 1	15	2545	169.6667	2005.238
Column 2	15	2690	179.3333	2474.524
Column 3	15	3580	238.6667	2333.81

Source of						
Variation	SS	df	MS	F	P-value	F crit
Between						
Groups	41874.44	2	20937.22	9.218611	0.00048	3.219942
Within Groups	95390	42	2271.19			
Total	137264.4	44				

Appendix B One Way ANOVA Test for source 1 and 2 of tap water sample

Anova: Single Factor

SUMMARY

Groups	Count	Sum	Average	Variance
Column 1	15	2545	169.6667	2005.238
Column 2	15	2690	179.3333	2474.524

Source of						
Variation	SS	df	MS	F	P-value	F crit
Between						
Groups	700.8333	1	700.8333	0.312889	0.58036	4.195972
Within Groups	62716.67	28	2239.881			
Total	63417.5	29				

Appendix C One Way ANOVA Test for source 2 and 3 of tap water sample

Anova: Single Factor

SU	JN	4N	ΛA	١R	Y
\sim					-

Groups	Count	Sum	Average	Variance
Column 1	15	2690	179.3333	2474.524
Column 2	15	3580	238.6667	2333.81

Source of						
Variation	SS	df	MS	F	P-value	F crit
Between						
Groups	26403.33	1	26403.33	10.98232	0.002548	4.195972
Within Groups	67316.67	28	2404.167			
Total	93720	29				

Appendix D One Way ANOVA Test for source 1 and 3 of tap water sample

Anova: Single Factor

SU	JN	4N	ΛA	١R	Y
\sim					-

Groups	Count	Sum	Average	Variance
Column 1	15	2545	169.6667	2005.238
Column 2	15	3580	238.6667	2333.81

Source of						
Variation	SS	df	MS	F	P-value	F crit
Between						
Groups	35707.5	1	35707.5	16.45868	0.000361	4.195972
Within Groups	60746.67	28	2169.524			
Total	96454.17	29				

Sample	Element	App Conc.	Weight (%)	Atomic (%)
Blank	С	2.19	48.85	55.99
	0	1.56	51.15	44.01

Appendix E EDX Analysis for	Blank, sample from	S1, S2 and S3
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Sample	Element	App Conc.	Weight (%)	Atomic (%)
S1	С	2.00	47.16	55.13
	0	1.65	50.14	44.00
	Na	0.02	0.26	0.16
	Al	0.05	0.57	0.30
	Si	0.03	0.26	0.13
	Cu	0.04	0.44	0.10
	Zn	0.03	0.33	0.07
	Zr	0.05	0.55	0.08
	Sn	0.03	0.28	0.03

Sample	Element	App Conc.	Weight (%)	Atomic (%)
S2	С	2.95	48.57	56.05
	0	2.19	50.23	43.52
	Na	0.02	0.22	0.13
	Al	0.02	0.20	0.10
	Cl	0.02	0.14	0.05
	Cu	0.05	0.41	0.09
	Zn	0.03	0.24	0.05

Sample	Element	App Conc.	Weight (%)	Atomic (%)
S3	С	3.41	52.17	59.52
	0	1.94	46.99	40.25
	Si	0.03	0.20	0.10
	Cu	0.05	0.04	0.08
	Zn	0.04	0.05	0.06