



**MICROPLASTIC CONCENTRATIONS IN THE ZANDVLEI
CATCHMENT AREA AND ESTUARY, CAPE TOWN**

by

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ABSTRACT

On a global scale, there is an increasing concern about the extent of plastic pollution in coastal areas. Global demand for plastic is increasing annually. South Africa has an extensive plastics manufacturing industry, but recycling is restricted and inadequate, with a large fraction of plastic waste entering the environment, which eventually degrades in smaller plastic particles, referred to as microplastic when smaller than 5 mm. Knowledge about the abundances and characteristics of microplastics is becoming ever increasingly important to assess the potential effects microplastics have on organisms and ecologically sensitive ecosystems. The aim of this study is to determine the abundance and polymer characteristics microplastics in the Zandvlei catchment and vlei area in Cape Town, South Africa from 2018 to 2021. Seasonal sampling was conducted to determine the characteristics (type, colour and size) of microplastics in the surface water and sediment. Microplastic polymer type was verified with a Fourier-transform infrared spectroscopy (FTIR). Microplastics were mainly transparent fibres smaller than 1000 μm size class. Concentrations were higher in sediment across the study 293 (\pm 37.1 SEM) with 18157 particles recorded across the whole study. This study provides baseline and quantitative information on microplastics in the catchment and Zandvlei Estuary. It will assess bioavailability of microplastics within the area and contribute to long term monitoring of the estuary and the surrounding areas. A risk assessment (Pollution Load Index, Polymer Risk Index and Pollution Risk Index) of microplastics sampled indicated that microplastics in the catchment compared to the vlei pose a greater threat to the environment. The results indicate that there is a need to monitor microplastics in riverine and estuarine ecosystems in South Africa.

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In everything give thanks; for this is God's will for you in Christ Jesus.

1 Thessalonians 5:18

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GLOSSARY

Acronyms	Definition / Explanation
ANOVA	Analysis of Variance
FTIR	Fourier Transform Infrared Spectroscopy
MMT/a	Million Metric Tonnes per Annum
BRICS	Brazil, Russia, India, China and South Africa
aMSL	aMSL Above Mean Sea Level
ABS	Acrylonitrile butadiene styrene
CPVC	Chlorinated polyvinyl chloride
EPDM	Ethylene propylene di-monomer
EPS	Expanded polystyrene
EVA	Ethylene vinyl acetate
HDPE	High density polyethylene
LDPE	Low density polyethylene
MF	Melamine-formaldehyde
PA	Polyamide, Nylon
PB	Polybutylene
PC	Polycarbonate
MP	Microplastic

CHAPTER 1: GENERAL INTRODUCTION

1.1 Statement of the research problem

Microplastics have become so ubiquitous that they can be found in almost every type of ecosystem globally (Vetrimurugan et al. 2020; Jong 2018), to even the most remote of locations and have been highlighted in a number of studies, including the Arctic (Mohamed and Obbard, 2014), mid-ocean convergence zones, such as the North Atlantic (Carpenter and Smith, 1972), Antarctica (Zarfl and Matthies, 2010) and South Pacific subtropical gyre (Eriksen et al. 2013).

There is increased concern over the exponential increase in plastic production and its prevalence in the environment globally which increased over the last decade (Duis and Coors, 2016). Global plastic demands had risen annually and stood at 245 MMT/a in 2011 (Andrady, 2011) and had further risen to 367 MMT/a in 2020, with a forecasted increase of 8.5% MMT/a (Howard et al. 2019). At the present rate, plastics production is estimated to double within the next 20 years (Lebreton and Andrady, 2019). The increase in the consumption of plastic drives an increase in the stocks of the material in the different sectors of the economy (Olatayo et al. 2021). This impressive success of plastics is unparalleled by any competing materials used in packaging or construction, the two major applications areas of plastics (Lebreton and Andrady, 2019).

Similarly, with South Africa following the same trend as the 11th largest plastic waste producer globally (Ryan, 2020). The South African economy is characterized by a fast-growing economy, growing middle class and high consumption of plastic resources (Vlad et al. 2014), which is consistent with emerging BRICS economies (O'Brien and Thondhlana, 2019). The state of waste management in South Africa is poor, with significant leakage of plastic debris to the environment, largely because of inadequate waste collection and disposal (Arabi and Nahman, 2020). This makes South Africa a key player in contributing to global plastic pollution with consumers in the country using approximately 8 billion plastic bags per year (Dikgang et al. 2012). The high production of plastic is associated with high consumption of resources and waste generation (Chen et al. 2020). Furthermore, given that South Africa has a relatively long coastline (of up to 2 500 km), its contribution and susceptibility (in socio economic and environmental terms) to plastic pollution of world seas and oceans cannot be underestimated (O'Brien and Thondhlana, 2019). With the increased flow of plastic into environmental systems resulting in a corresponding increase of its share in waste generated. The ever-increasing volume of plastic waste entering the land, river systems, oceans and food samples is reported to have a huge environmental impact on biodiversity, ecosystems and human health (Wright et al. 2013).

Micro and possibly also nano-scale plastic particles are common in the environment. In myriads of microplastic research papers, microplastics were discovered to differ in size, shape, chemical composition and spatial and temporal distribution (Wright et al. 2013). In the last 30 years, microplastics started to largely collect in surface water and sediments globally, with record concentrations reaching cubic meters (Wright et al. 2013). Plastics are versatile, lightweight, durable, transparent material, are ideal for a multitude of applications and are inexpensive to produce (Laist, 1987). At a global level, terrestrial sources are responsible for 80% of plastic debris and the remaining 20% originates from marine sources (Andrady, 2011). Microplastics currently have a worldwide distribution and have been identified at all levels of the natural environment (Solomon and Palanisami, 2016).

South African coastal ecosystems support a high degree of biodiversity and can be regarded as moderately pristine (Wepener and Degger, 2012). However, the rapid increase of coastal population densities, urbanization and industrialization are placing strain on the environment and its resources (Fisner et al. 2013). According to Taljaard et al. (2006), 30% of South Africa's population lives along the coastline, with the largest aggregation of people in Cape Town, Western Cape (Krige, 2019). With coastal areas being home to a large percentage of the South African population, many anthropogenic activities place stress on and releases excessive amounts of pollution into the coastal environment (Davenport et al. 2006). In highly populated coastlines Nel et al. (2017) is in agreement that plastic, specifically microplastics have been found. South Africa has an extensive plastics manufacturing industry, but recycling is restricted and inadequate, with a large fraction of unregulated waste entering the environment (Verster et al. 2017). The unregulated waste i.e., microplastics found in lotic (lakes and wetlands) environments have a longer residency time because of low circulation, however, in lentic (oceans) the microplastics would be dispersed easier and more widely apart (Soballe and Kimmel, 1987; Narvsten, 2009).

Actual plastic pollution studies only started to be conducted in South Africa in the 80's when Ryan (1988) conducted the first marine and coastal plastic study in South Africa. This looked at ingestion by seabirds and was followed up by (Ryan and Moloney, 1990) with the discovery of microplastics in South Africa in the form of polyethylene pellets. Following this there has been a number of marine and coastal studies have followed suite, with research efforts increasing from 2017.

Coastal systems like estuaries are among the most productive systems where freshwater from streams and rivers meets marine waters of coastal bays and mixing occurs. An estuary cannot

function efficiently without freshwater inflows from rivers and streams. Freshwater inflows are important to estuaries because they provide low-salinity nurseries and transportation of nutrients, sediment, and organic material which affects species movement and reproductive timing (Montagna et al. 2002). Sadly, not enough research has focused on microplastic contamination in freshwater and estuarine settings. The literature that is available highlights the importance of rivers and streams but also the fact that they can act as conduits for plastic into marine and coastal environments, estuaries (Naidoo et al. 2015), beach sediment (Ryan et al. 2009; De Villiers, 2018; Ryan et al. 2018) and wetlands (Reynolds and Ryan, 2018).

The respective study focuses on Zandvlei estuary and its catchment with permission granted from City of Cape Town, Conservation Services Unit. Zandvlei Estuary plays an integral role on the False Bay coast as it is the largest of eight and the only functioning estuary on the False Bay Coast. The estuary alone is a biodiversity rich area, supporting an abundance of fish species and acts as a nursery to juveniles and critically endangered species like the white steenbras (*Lithognathus lithognathus*). This haven allows the juveniles to mature and later exit to the ocean. The estuary is artificially opened and managed by City of Cape Town's Catchment Management Department. The boundaries of the vlei are a proclaimed bird sanctuary and acts as a habitat for indigenous bird species as well as migrants. This area provided much needed ecosystem services such as food security, carbon storage and nutrient filtering and sediment run-off from the catchment. Much of the original estuary and related services has been destroyed over the past four decades through the processes of farming, introduction of invasive species, dredging and urban development.

Although, Zandvlei estuary is ecologically significant in terms of its biodiversity, conservation importance and is ranked in the top 25% of ecologically important estuaries in South Africa, no prior research on microplastics has been conducted in the area. The only plastic related study conducted in Zandvlei estuary (Ryan and Perold, 2021) study which focused on the interchange of macroplastics/litter between the adjacent rivers and coastal waters. Botterelle et al. (2019) investigated the factors that affects the bioavailability of microplastics within a different estuarine environment, which included microplastic size, colour and type. Although this study was not conducted in Zandvlei estuary, the categorizations established will help to address key knowledge gaps with regards to the natural environment in the present study. Furthermore, it is imperative to understand the potential chemical effects microplastics pose on estuarine health, trophic level transfer, foodwebs, fauna and flora populations in the ecologically sensitive Zandvlei and the resulting economic effects this may have. While, conserving the health of the estuary is ecologically important, the area it also has significance to the non-scientific community, public,

residents and various stakeholders, as the area is used for sports, recreational fishing, religious activities and studies.

1.2 Research questions

- What are the concentrations of microplastics in Zandvlei sediment and water?
- How do these differ spatially between sampling sites i.e. mouth, vlei and catchment?
- How do these differ between mediums i.e. sediment and water?
- What types of microplastics are present in Zandvlei Estuary i.e., size, colour, and shape?

1.3 Aims and objectives of the study

The aim of this study is to determine the characteristics and potential effects of microplastics in the Zandvlei Catchment area in Cape Town, South Africa.

The main objectives of the study were:

- i) To determine spatio-temporal features of microplastics in surface water and sediment in the Zandvlei catchment.
- ii) To determine the general microplastic characteristics i.e (colour, shape and size) between catchment and vlei.
- iii) To determine the difference in abundance of microplastics between catchment and vlei.
- iv) To identify microplastic polymers in order to assess the impact of microplastics on the environment

1.4 Overview of the Dissertation

The aim of this research was to address some key knowledge gaps that existed regarding microplastic research in Zandvlei Estuary and Catchment.

Chapter 1 provides a general introduction, highlighting the importance of the respective study by giving insight into research gaps and evaluating the current situation.

Chapter 2 comprises of a literature review, general discussion on plastic, its history, its applications and its role in estuarine pollution, aims objectives and the essential concepts of the respective study.

Chapter 3 comprises of the study area and describes the materials and methods used.

Chapter 4 comprises of the results section describing the characteristics of microplastics at sites based spatial (catchment and estuary) and temporal (seasonal and annual) scales. The potential effects of microplastics are based on determining the hazard scores of polymers recorded.

Chapter 5 is a general discussion that provides a synopsis of the study.

Chapter 6 is recommendations, shortcomings, and conclusions.

CHAPTER 2: LITERATURE REVIEW

2.1 History of Plastic Production

The etymology of the term plastics traces back to its Greek origin *plastikos* which directly translates to fit for moulding (Hammer et al. 2012). The etymology is of keen interest because it has been noted by (Hosler et al. 1999) that ancient Mesoamericans have benefited from the use of polymers since approximately 1600 BC, when they moulded natural rubber into balls, figurines and bands. In the following years society has heavily relied on plastics and rubber, first experimenting with natural polymers, horn, waxes, natural rubber and resins, until the nineteenth century, when the development of modern thermoplastics began (Andrady & Neal, 2009). During the first 50 years of the twentieth century the development of plastics expanded with the manufacture of 15 new classes of polymers. The unique properties of plastics altered the way we live. These polymers have become a vital component of modern life since its mass manufacturing in the 1940s (Karami et al. 2017). Plastics have a distinctive set of characteristics which allowed it to be in high demand for everyday use. Plastics are usable at a broad temperature range, possessed low thermal conductivity, elevated strength to weight ratio, and was bio-inert, durable and most importantly inexpensive (Andrady & Neal, 2009). Furthermore, with the many uses of plastics and the fact that it is cost effective to produce, it had driven the worldwide consumption for plastics to reach exuberant amounts (Yu et al. 2020). There are hundreds, if not thousands of plastic materials, but the market is dominated by six groups of plastics: Polyethylene (PE, high and low density), Polypropylene (PP), Polyvinyl Chloride (PVC), Polystyrene (PS), Polyurethane (PUR) and Polyethylene Terephthalate (PET) (GESAMP, 2015). Consequently by the 1940's and 1950's, global production of plastics had risen steeply to approximately 1.5 million tonnes per annum (Barnes et al. 2009; Claessens et al. 2011; Naidoo et al. 2015). In fact, this was due to plastic manufacturers using coal to produce resins, polystyrenes and nylons (Brydson, 1966). Petroleum had become the main raw material for plastic production by the 1960's, which overtook the whole manufacturing process (Brydson, 1966).

By the 1950's, global plastic production was at 1.5 MMT/a and increased to 250 MMT/a in 2011 (Wright et al. 2013) and 300 MMT/a by 2014 (Frolich, 2014), with the latest global production information available at 368 MMT/a for 2019 (Tiseo, 2021). There is a general agreement in literature that the ever-increasing plastic production numbers are unsustainable. Studies by Rochman et al. (2013) and Henry et al. (2019) both hold the same view that if plastic production trends continue the cumulative total mass of plastics produced will increase into the billions.

Rochman et al. (2013) predicted by 2050 the MMT/a will increase to 33 billion tonnes and Henry et al. (2019) predicting 12 billion tonnes. These predicted totals are heavier than all the fish combined in the ocean (Auta et al. 2017). Overall, these studies highlight the need for adequate guidelines in global plastic production.

2.2 Characteristics of Plastics and Their Uses

Modern society relies so heavily on plastics that they have become an indispensable part of our daily life. According to Wabnitz and Nichols (2010) plastics are desirable to society because of its high durability, transparency, low mass, low costs, high insulation and high resilience to biological break down. These include clothing and footwear, together with products for use in food and public health applications and many others (Andrady & Neal, 2009). Additionally, plastic polymers are used in food and beverage products including bottles, lids/caps, bags, drinking straws (Ryan et al. 2014). Plastics are durable, having a long lifespan that prolongs their use. Because they are engineered to be durable and inexpensive, approximately 50% of plastic products, including utensils, plastic bags and packaging are designed to be disposable (Hopewell et al. 2009). Additionally, several plastic products are only used a single time before being discarded, which are termed throw away, disposable and single-use plastics (Rios et al. 2007). Plastic has also proven to be an essential part of the automotive and shipping industries, as it is used for protective packaging and weight reduction in transit which ultimately lowers fuel consumption (Alimba & Faggio, 2019). Plastic practically has an infinite number of possible applications (Boucher & Friot, 2017).

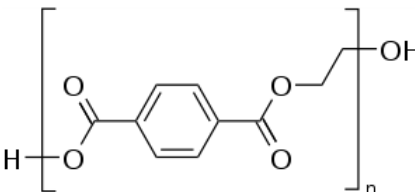
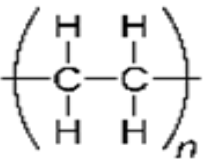
2.3 General Structure of Plastics

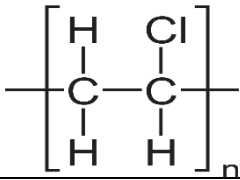
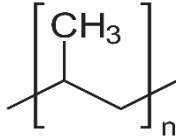
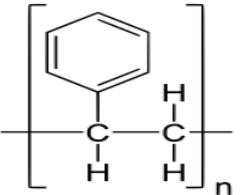
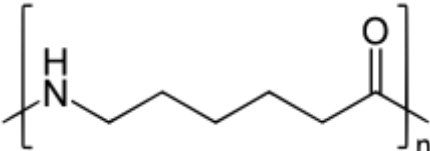
Plastics are defined as synthetic organic polymers made from the polymerization of monomers (Derraik, 2002; Cole et al. 2011). Plastics are derived from natural organic occurring organic materials examples of these include cellulose, coal, natural gas, salt and crude oil (Plastics Europe, 2021). The crude oil needs to be distilled in an oil refinery where the heavier crude oil is separated into groups with lighter components known as fractions. Durable as crude oil derived polymers and melt at high temperatures (Kade and Dalton 2021). Corn made plastics are made from sugar found in corn to make polylactide polymers. The polymers are not as Plastic polymers have a hydrocarbon structure (Table 2.1). They are made up of carbon and hydrogen atoms. They are well recognized for their malleability and durability, as well as light weight and chemically stable with good insulation and low thermal conductivity. Plastic polymers are made by polymerising monomers into macromolecular chains, known as polymerization. This occurs when the small molecules made up of the hydrocarbons called monomers, combine

and form chain like networks or structures of the same monomer. Plastic is not sold as a pure unadulterated substance but is instead mixed with various chemicals and other materials, which are collectively known as additives. These are added during the compounding stage and include substances such as stabilizers, plasticizers and dyes, which are intended to improve the lifespan, workability or appearance of the final item. In some cases, this can involve mixing different types of plastic together to form a polymer blend. Besides monomers, water, ethanol and benzyl alcohol are often needed for polymerization to occur, e.g. initiators, catalysts, and depending on the manufacturing process solvents may also be used (Lithner et al. 2011). Plastics can further be divided into two classifications thermosets and thermoplastics. Thermoset plastics are non-recyclable and non-heatable applications of thermosets include textile fibres (polyester) and polyurethane (electrical equipment). Thermoplastics are recyclable and can be re-heated applications include Polystyrene (toys), PVC (pipes), Polyethylene (bottles). Below are a few of the most common plastic polymers with their uses and functional groups.

Essentially additives are chemicals added to improve the processability and prolong the lifespan and achieve the desired physical and chemical properties in plastics eg PVC. Anti-ageing stabilizers are used to extend the lifespan of plastics and prohibit deterioration from UV light. An example elemental carbon or black dyes are added as black is a good UV absorber. Flame retardants slow down combustion. These additives are common in electrical products to avoid burning/ignition of plastic. Halogens such as bromines as well as phosphorus and nitrogen are common flame retardants.

Table 2.1: Common plastic polymers with their uses and functional groups

Polymer	Uses	Functional Group	Reference
Polyethylene terephthalate (PETE or PET)	Polyester Fibres Shatter proofing Water bottles Film		(Crichton et al. 2017)
Polyethylene (PE) High-density/Low-Density	Plastic bags Medical applications		(Mayoma et al. 2020)

Polyvinyl Chloride (PVC)	Plumbing Insulation Electrical wires		(Ghayebzadeh et al. 2021)
Polypropylene (PP)	Toys Carpets Packaging		(Andrady and Neal, 2009)
Polystyrene (PS)	Foam Applications		(Besseling et al. 2013)
Nylon (PA)	Clothing Car tires Rope/Thread		(Reddy et al. 2006)

Furthermore, plastics are also manufactured in synthetic polymer blends. These polymer blends are physical blends of two or more macromolecular substances. Polymer blends could belong to the same chemical family i.e low and ultra-high molecular weight Polyethylene (PE) or to different ones such as Polyethylene (PP) and Nylon (PA) blends. Certain polymer blends have become of keen interest as these materials have the ability to alter certain properties of an item. Of most interest is rubber toughening of tyres with butadiene rubber and styrene butadiene rubber polymer blend and the ability to make clothing more robust with blends such as polyester and nylon polymer fibre blends.

2.4 Microplastic Types and Sources

Microplastics are referred to as plastics smaller than 5mm in diameter (GESAMP, 2019) with the Zhang et al. (2021) stating that they can be separated into primary and secondary, based on their origin (Ajith et al. 2020). To date the literature separates plastic debris into size classes of either <5 mm (microplastics) or >5 mm (macroplastics) (Arthur et al. 2009).

2.4.1 Primary Microplastics

There has been a general consensus between the studies of (Boucher & Friot, 2017) and (Verster & Bouwman, 2020) on the theory that primary microplastics are plastics directly released into the environment in their manufactured form i.e. small particles, beads, spheres,

ellipses, rods, fibres, granules (Fendall & Sewell, 2009). In particular, polyethylene, polypropylene, and polystyrene microplastics have replaced natural scrubbers like pumice and oatmeal (Gregory, 1983), microplastics are now manufactured in industrial and commercial products such as scrubbing agents in toiletries (Andrady, 2011), cosmetics (Browne et al. 2011), beads in face wash (Van Cauwenberghe et al. 2015), toothpaste (Hammer et al. 2012) medical products (Horton & Dixon, 2018) and blast cleaning (Barboza et al. 2018). Granulated particles called microbeads are also classified as primary microplastics, with their incorporation in a number of industrial (air-blasting media) and household (hand-cleaners and facial scrubbers) products (Gregory, 1977). In addition primary microplastics can also originate from the abrasion of large plastic objects during manufacturing, use or maintenance such as the erosion of tires when driving (Van Wijnen et al. 2019) or the abrasion of synthetic textiles during washing (Alam et al. 2019), sewage discharge and spillage (Gregory and Thompson, 1978).

Primary microplastics are microplastics that are directly released into the environment in the form of small particulates. The main known primary microplastic sources are synthetic textiles, personal care products and plastic pellets.

In the primary form many plastics are in the pellet form. Pellets are either used in shipping or they are used to create other larger plastic items (Liebezeit & Dubaish, 2012). The pellets are then transported to plastic transformers that generate the large plastic products. During transport these pellets can also be spilled. Other plastic forms are microbeads (spheres) which are manufactured for face scrubbers and toothpaste. The nurdles are melted to form other products.

The production of a primary microplastic like nurdles begins when oil such as crude oil is extracted from the earth. The oil is then shipped to the processing factories where it undergoes refinement. The oil is then heated and is split into ethane and propene. The cracking process then begins where ethane and propane is broken down into their original state into smaller units. In this process molecular bonds are broken creating monomers by the process of polymerization. Polymerization causes ethane to become ethylene and propylene becomes polypropylene (Egessa et al. 2020). The stabilizers are then added. The plastic is then chopped into smaller pieces or nurdles.

2.4.2 Secondary Microplastics

In contrast to primary, secondary microplastics originate from the degradation of larger plastic items into smaller plastic fragments (Boucher and Friot, 2017), known as fragmentation (Napper and Thompson, 2019). Thompson (2006) stated that fragmentation is a result of physical, chemical, and biological processes. Breaking down of plastic debris makes pollution rates less

obvious since the particles are smaller, but it does not make the effects of it less harmful and instead poses new challenges in detecting microplastics.

2.4.3 Fragmentation of Secondary Microplastics

Microplastic fragmentation can further be divided into two categories abiotic and biotic degradation. Abiotic degradation refers to the physical or chemical changes of properties that occur for plastics due to abiotic factors such as light, temperature, air, water, and mechanical forces (Andrady, 2015). Microplastic degradation most commonly undergoes abiotic degradation as opposed to biotic (Zhang et al. 2021). Abiotic degradation can further be divided into:

2.4.4 Chemical Process

2.4.4.1 Photo degradation

Photo degradation is recognized as the most important abiotic process that initiates plastic degradation (Zhang et al. 2021). Photo degradation also known as oxidative photo degradation, is caused by the ultra violet component of solar radiation which involves a free radical mediated reaction that is induced by solar irradiation (Liu et al. 2019). The main ultra violet radiation which catalyzes photo oxidation are High energy ultraviolet (UV), irradiation UV-B (290e315 nm) and medium energy UV-A (315e400 nm) (Akdogan & Guven, 2019) . During the photo oxidation reaction the ultra violet light is absorbed by the plastic and induces the breaking of the plastic polymers' bonds. When the bonds of the plastic breaks it causes the plastic to lose colour, become brittle and break off from the weathered surface layer of the larger plastic object (Derraik, 2002). Iwalaye et al. (2020) reported that increased temperatures, due to climate change, may speed up photo degradation as increased elevated temperatures will also speed up the disintegration of macroplastics to microplastics due to photochemical processes activated by ultraviolet (UV) light and result in increased microplastic concentrations in the environment (Cao et al. 2017). Alternatively, (Harshvardhan and Jha, 2013) found that while heat, sunlight, and well aerated conditions are ideal for plastic fragmentation by chemical pathways, cold and anoxic conditions of aquatic environments and sediments can also cause fragmentation but at a much slower rate, which may even take centuries.

2.4.4.2 Thermal degradation

Thermal degradation occurs when plastics are exposed to high temperatures this results in the breakdown of plastics due to the energy input from the high temperatures (Zhang et al. 2021). The plastic will undergo a thermo oxidative reaction at these elevated temperatures, when the temperature is high enough, heat will be absorbed by the plastic polymer to overcome the energy barrier. The polymer chains break (chain scission) and react with one another (cross-link) to

change the properties of the polymer. This can cause the polymer to become brittle, lose colour and lower ductility.

2.4.5 Physical Process

Mechanical degradation refers to the breakdown of plastics due to the action of external forces (Zhang et al 2021). Mechanical degradation reduces the average molecular weight of the polymer by external forces such as surface scraping with sand and rocks under the influence of wave and wind action (Andrady, 2011). Pal et al. (2008) noted that freezing and thawing of plastics can also cause degradation to the polymer in areas of extreme low temperatures such as the arctic. The fracture strain of the polymer indicates how capable the plastic is able to resist the changes in shape without cracking. Lower fracture strain plastics are more prone to fragmentation and perforation under tensile forces.

2.4.6 Biotic degradation

Biotic degradation of plastics refers to the deterioration of plastics caused by organisms (Zhang et al. 2021) or biologically by biochemical processes (Danso et al. 2019).

2.4.6.1 Biological degradation

Plastics exposed in seawater can be colonized by microorganisms that form biofilm, which affects the light transmittance of plastics and causes biodegradation (Sudhakar et al. 2007; Eich et al. 2015; Horton and Dixon 2018). Biodegradation is the process by which organic substances are broken down by living organisms. Plastics are biodegraded aerobically in wild nature, anaerobically in sediments and landfills and partly aerobically and partly anaerobically in composts and soil. Carbon dioxide and water are produced during aerobic biodegradation and carbon dioxide, water and methane are produced during anaerobic biodegradation (Gu et al. 2000). Generally, the breakdown of large polymers to carbon dioxide (mineralization) requires several different organisms, with one breaking down the polymer into its constituent monomers, one able to use the monomers and excreting simpler waste compounds as by products and one able to use the excreted wastes.

2.4.6.2 Bacterial Degradation

Microorganisms are able to survive under various conditions and temperatures and can reach high biomasses. These microorganisms can make use of any surface area in the ocean to colonize. In seawater bacterial colonization on plastic material starts almost immediately. Within a few hours microorganisms are able to form micro assemblages and cover the surface area of

plastic also known as attachment. During this stage microbial assemblages catalyze metabolic reactions that leads to adsorption, desorption and fragmentation of the plastic (Harrison et al. 2011).

2.4.6.3 Microalgae Degradation

Building biofilms provides a platform for the settlement of other organisms such as microalgae (diatoms, flagellates, protists) and microscopic algae. Due to phylogenetic, functional and ecological variety. Biofilms are termed biofilming community (Rummel et al. 2017). Biofouling increases the density of the particle, and it may sink to the seafloor (Pauli et al. 2017). Reisser et al. (2014) stated that it is expected that biofouled materials could attract invertebrates capable of grazing on plastic.

Biodegradation is a process by which microbial organisms (bacteria and fungi) transfer or alter (through metabolic or enzymatic action) the structure of chemicals introduced into the environment. Microbial attachment on the surface and formation of biofilms depend not only on microorganisms but ability to build the surface structure/roughness and material of plastic. Consequently, increased temperature results in the increased rate of degradation.

2.4.7 Fibres

Fibres can be defined as objects resembling thread (Table 2). Fibres can be generated separated into naturally occurring such as silk, cotton, wool and jute or man-made which are derived from polymers. Furthermore, man-made fibres can be separated into natural, synthetic and regenerated fibres.

2.4.7.1 Natural Fibres

Material of natural origin examples include cellulose fibres, protein fibres and cellulose Ester fibres.

2.4.7.2 Synthetic Fibres

Synthetic fibres are derived from products of petroleum. Synthetic fibres are man-made through the process of polymerization examples include polyester, nylon and acrylic, polyetherane and polypropylene. The fossil fuels (gas, oil, coal) are polymerized at high temperatures to create a polymer solution. The solution is then pushed through spinnerets as the solution passes it cools and solidifies in the form of threads (Thompson et al. 2009).

2.4.7.3 Semi-synthetic Fibres

Cellulose are considered semi-synthetic because it undergoes chemical processing of the fibres and the addition of artificial dyes during the production of Rayon, which is an artificial textile. It then becomes a challenge to identify original cellulose fibres from plants such as cotton (Renny et al. 2015). Semi synthetic fibres also known as regenerated fibres are derived naturally occurring fibres through chemical processes, the fibre is broken down, reconstructed most commonly done with cellulose. Regenerated fibres are produced from cellulose polymers that occur in plants naturally. These include cotton, wood, hemp. Rayon and acetate fibres were historically the first fibres to be produced from cellulose, where the cellulose was taken and made into fibres.

2.4.7.4 Sources of fibres

Lastly, microplastic literature often lacks clarity on the exact sources of microfiber contamination, with different theories existing in the literature. Browne et al. (2011) stated that microfibers are the most commonly reported form of microplastics in literature but lacking the exact origin. According to Browne et al. (2011) an important source of microfiber contamination in the environment originates from washing laundry, with laboratory experiments demonstrating that a single garment can potentially produce N1900 fibres per cycle , and an average 6 kg load of acrylic fabric could release over 700, 000 fibres into the environment (Napper & Thompson, 2019). Cole et al. (2011) study concurs that microfibers are released during the washing of synthetic clothing, with textile mills also being a point-source release into the environment (Akdogan & Guven, 2019). Elevated microfiber levels have also been linked to domestic waste water and sewage-sludge disposal sites (Leslie et al. 2017).

Table 2.2: Description of common microplastic types

Type	Shape description	Potential source	Reference
Fibre	Thin and hair like	Synthetic fabrics Rope	(Lots et al. 2017)
Fragment	Irregular and chipped shape	Particles broken off from larger plastic	(Vilakati et al. 2020)
Film	Sheet like	Plastic bags, wrappers and foil packets	(Kalogerakis et al. 2017)
Foam	Foamed plastics	Polystyrene packaging	(Lozano et al. 2021)

Spheres	Round	Shipping and packaging	(Chubarenko et al. 2016)
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2.5 Plastic Sizes and Shape

Microplastics are of < 5 mm in size and have a unique set of characteristics. Different sources and methods of fragmentation cause them to occur in diverse shapes such as fibres, spheres, fragments, foam and films in environmental samples (Klein et al. 2015). GESAMP (2015) suggests that more rigorous definitions should be adopted for separating plastics into nano (< 100 nm), micro (1– 5 mm, < 1 mm, > 330 µm), meso (< 25 mm, 1 – 25 mm), macro (25 – 50 mm), and mega-size (>1 m) size classes in order to ensure uniformity in microplastic representation in literature. According to Zhao et al. (2015) microplastics are currently being classified into 4 groups dependent on their shape types: fibre, film, granule and pellet. Microplastics are also categorized into five groups: fragments (hard, jagged-edged particles), micropellets (hard, rounded particles), and fibres (fibrous or thin uniform plastic strands), films (thin, 2 dimensional plastic films) and foam (Styrofoam type material) as described by (Anderson et al. 2016).

2.6 Factors Affecting Microplastic Abundances in Estuaries

2.6.1 Transportation of microplastics into the environment

Estuaries have become densely populated and industrialized which presents additional microplastic contamination. Population clusters such as cities and towns have become increasingly important sources for microplastic hotspots. Social and economic activities and productions have impacts on the concentrations and distributions of microplastics. Additionally, urbanization intensifies this. Microplastics originate from land-based sources and enter the marine environment through direct and indirect pathways (Li et al. 2021). These include informal and illegal dumping and littering (Verster & Bouwman, 2020), estuaries (Naidoo et al. 2015), rivers (Atwood et al. 2019; Weideman et al. 2020b), sewage outlets (Carr et al. 2016; Edo et al. 2020) and storm water inputs (Weideman et al. 2020a). In addition, about 80% of marine microplastics are of land-based origins such as wastewater treatment plants, coastal landfills, and touristic activities (Andrady, 2011; Sadri and Thompson, 2014; Sousa et al., 2021), with the remaining 20% from sea-based activities such as fishing and shipping (Díez-Minguito et al. 2020).

Moreover, rivers and stormwater runoff transport microplastics from the terrestrial and freshwater

compartments into the ocean, with the estuary acting as transition zone (Preston-Whyte et al. 2021). The increased urbanization of estuaries makes them susceptible to plastic pollution due to the positive correlation between plastic generation and economic development (Jambeck et al. 2015). While estuaries act as sources of microplastic pathways into the ocean, the presence of these particles in the sediments suggests that they may act as potential sinks as well (Alves and Figueiredo, 2019; Baptista Neto et al. 2019; McEachern et al. 2019).

By location estuaries act as the interface between oceans and rivers. Microplastics that are suspended in water are very sensitive to hydrodynamics. Therefore, the tidal currents coming in and out of the estuary are most important for the transport of microplastics in suspension. The following estuarine hydrodynamics have profound implications for microplastics.

2.6.2 Movement and temporal variation of microplastics in estuaries

2.6.2.1 Daily Tides (Ebb and Flood)

Estuaries are characterized by their brackish waters that result from the mixing of salty seawater and riverine freshwater, with intense mixing occurring in the centre of the estuary. The water exchange in such environments is a factor of river discharge on the continental side (upper estuary) and tidal cycles on the marine side (lower estuary) (Dris, 2015). The estuary is affected by the daily tidal action of ebb and flood tides. The ebb tide is a seaward flow that flushes water out of the estuary creating a low water level. As opposed to ebb tide the flood tide is an upstream tide resulting in a high-water level (Malli et al. 2022). An estuary dominated by ebb flow is characterized by higher amounts of particles, sediments, and water being flushed out of the estuary rather than being carried into it (Gallagher et al., 2016) resulting in lower microplastic concentrations as opposed to flood dominated estuaries.

2.6.2.2 Monthly Tides (Spring and Neap)

Estuarine water levels vary monthly because of spring and neap tides. There is a pronounced difference between high and low and water levels during spring and neap tide. These tides follow the lunar cycle with spring tides occurring during new and full moons (higher water levels) and neap tides occurring during the first and third quarter moon (lower water levels). Elevated microplastic concentrations can be observed during spring tide because stronger currents of the spring tide are capable of mobilizing higher quantities of microplastics into the estuary (Malli et al. 2022).

2.6.2.3 Seasonal Variations (wet and dry seasons)

During the wet season a period of high precipitation causes riverine input, a high freshwater discharge entering the estuary. Conversely, the dry season is characterized by low river flows

due to a decrease in rainfall.

2.6.2.4 Windage

Windage is defined as the wind-induced motion of microplastics (Bermúdez et al. 2021). Low-density microplastics are more prone to be affected by windage. The wind factor affects the transport, suspension, and deposition of microplastics (Pazos et al. 2021). Winds are capable of importing or exporting low density microplastics to or from the estuary (Cheung et al, 2016; Naidoo et al.,2015; Vermeiren et al. 2016). Furthermore, microplastics become redistributed in the water column due to turbulent mixing caused by the wind (Kooi et al. 2017; Vermeiren et al. 2016). When the wind velocity increases the surface microplastics become submerged in the mixed layer (Kobayashi et al. 2021). As a result, the amount of microplastics decreases with depth (Kooi et al. 2017). This is because the effect of wind decreases in the deeper layers (Vermeiren et al. 2016). Additionally, in shallow estuaries, the wind-induced mixing reaches the sediments and causes the resuspension of microplastics (Naidoo et al. 2015; Vermeiren et al. 2016; Yonkos et al. 2014; Zhao et al. 2014).

2.6.3 Endpoint

An observation globally and locally are that estuaries act as microplastic hotspots and sinks (Avellán-Llaguno et al. 2021), studies have been conducted on different mediums, water (Weideman et al. 2020), biota (Naidoo et al. 2020) and sediment (Naidoo et al. 2015). Mud and sand flats retain sediments from inflowing rivers and streams (Marissa et al. 2020). Microplastics are retained in these areas. Factors facilitating the sinking and the residency of microplastics in estuaries include: sedimentation, aggregation, resuspension, and biofouling.

2.6.4 Sedimentation

Several authors reported elevated concentrations in surface sediments compared to the water column (Gray et al. 2018; Wu et al. 2020a). It is hypothesized that estuarine sediments may act as a sink for these particles (Luo et al. 2019; McEachern et al. 2019; Sanchez-Hernandez et al. 2021). Microplastics float in estuarine waters but because of the fluctuations and density differences the high density microplastics sink and become trapped within the sediments. Additionally, the shape of microplastics plays a role in their settling mechanisms where those with irregular shapes have more complex sedimentation schemes (Horton & Dixon, 2018). The surface sediments are where microplastics are first deposited (Xu et al. 2020), with many other studies reporting higher microplastic abundance in the upper layer of the sediments, compared to the deeper layers (Díaz-Jaramillo et al., 2021; Willis et al., 2017; Zhou et al. 2021). In short, sedimentation hinders the transport process of microplastics from the river to the ocean.

2.6.5 Aggregation

Aggregation of microplastics is caused by a physiochemical process that transports microplastic vertically and horizontally across water bodies. Aggregation can be further separated into heteroaggregation and homoaggregation. Homoaggregation can be described as particles of the same kind attracting each other and heteroaggregation particles of dissimilar sizes and makeup attracting each other (Yang et al. 2022). Homogenous aggregations are affected by factors of the particle size, aging degree and physicochemical properties of water (Dargo et al. 2020). Therefore, larger microplastics have greater stability and are less prone to aggregation. Heterogeneous aggregation is of more concern because microplastics aggregate with other solid constituents such as organisms, clay minerals and proteins. Under the same conditions, microplastics with smaller sizes are more prone to aggregation than those with larger sizes (Wang et al. 2021). Size is the main contributing factor to aggregation as larger plastic particles are less likely to aggregate together as opposed to smaller microplastics. Microplastics with small particle size can adsorb on the surface of large, suspended sediments and form heteroaggregates, which may cause a rapid settlement of the suspended microplastics in the water. Microplastics after entering the aquatic environment may sink depending on their buoyancy to the sediment (Wong et al. 2020). Waldschläger and Schüttrumpf (2019) found that microplastics with larger size rose rapidly, which also explained why more microplastics with smaller size were found in sediments.

2.6.6 Resuspension

Microplastic resuspension is caused by the disturbance of settled microplastics, often already settled in sediment and in the deeper layers of the water column. Furthermore, resuspension can occur during periods of turbulence and mixing driven by wind and resulting in microplastics being overturned in the water column (Eo et al. 2019). Resuspension can also be driven by human impact in an area of high fishing and urbanisation by boat activity overturning settled microplastics (Xia et al. 2021).

2.6.7 Biofouling

Microplastics are hydrophobic, attracting different ions, films and organic material, increasing the density of the material. In general, biofouling makes microplastics denser increasing their sinking velocity (Kooi et al. 2017; Pinheiro et al. 2021; Rogers et al. 2020). If the overall density of the microplastics surpasses that of the water it is suspended in it will sink. If the biofilm grows enough, it could increase the density of the MPs enough to eventually reach the estuarine sediments

(Zhang, 2017).

Lastly, microplastics transport is also affected by a set of characteristics: size, density and associated settling velocity. Neutrally-buoyant microplastics easily spread through the entire water column and they are more affected by tides. They are flushed from the estuary within few tidal cycles, while heavier microplastics tend to settle in the estuary. Neutrally-buoyant microplastics are concentrated in the surface layer with a reduced residency time as opposed to higher-density microplastics. Naidu et al. (2018) concurs that the high-density polymer sinks and accumulates in estuarine sediment. Additionally, an issue is of particular importance in the challenging context of estuaries, where the competition between density stratification and turbulent mixing can drastically affect the behaviour of water masses and suspended particles. Intense periods of mixing (i.e. ebb) followed by strong stratification periods (i.e. flood) have a great impact on the behaviour of suspended sediment (Defontaine et al., 2019). Similarly, microplastic distributions is expected to be strongly affected by the complex estuarine hydrodynamics, impacting the contamination of both inner estuary and connected coastal waters. Globally, estuaries have been identified as microplastic hotspots as a result of exposure to plastic contaminants (Wright et al. 2013). Densely populated areas adjacent to estuaries inadequately dispose of plastic waste, which enters the ocean through estuaries, yet published information on this area of research is severely lacking (do Sul and Costa, 2013). Estuaries allow tributaries to drain into the ocean when the tidal water (and litter in it) becomes diluted by riverine freshwater that flows seaward (Barletta and Dantas, 2016). The prevailing weather i.e. wind and rainfall which acts as an external force, had been proven to play an integral role in influencing the abundance and distribution patterns of microplastics in an aquatic environment (Eerkes-Medrano et al. 2015). Despite the long-term threat of microplastics on coastal ecosystems, sampling challenges have prevented the regular monitoring of microplastic abundance (Mohamed et al. 2014)

2.7 Global Microplastic Contamination

Currently microplastics are widespread across many different mediums and regions but with this growing concern research is still more ocean-centric (Cole et al. 2011). Microplastics pollution has received a lot of media attention and research, however despite growing attention, the actual amounts of plastics in environmental compartments (terrestrial, marine, freshwater, and atmospheric) and their ecological significance are still unclear. This is in part due to the recency of attention and lack of adequate sampling and analysis approaches, as well as the immensity and diversity of the oceans.

Majority of the studies globally focused on marine studies but in recent years studies have started shifting research efforts to deeper waters, sediments, freshwaters, soils, air, and biological systems. Furthermore, with the shift in focus on microplastic study areas, areas which

were presumed to be pristine, including Arctic sea ice (Peeken et al. 2018), the Antarctic (Waller et al, 2017), remote mountain ranges (Allen et al. 2019), and deep ocean (Jamieson et al. 2019) are now displaying microplastic contamination.

Microplastics is only a fraction of a larger emergent threat, which is litter. On a global scale and in most countries litter is not adequately disposed of. These larger plastics are disposed of in wastewater treatments landfills, incinerated, or recycled, although much is mismanaged and enters the natural environment.

Additionally, emergent and poor countries heavily rely on plastics and have produced the most waste (Figure 2.1) because of disposal in landfills, incinerated, or recycled, although much is mismanaged and enters the natural environment. Resulting in an upsurge in global plastic production and consumption. The rate of plastic production has recently surpassed that for carbon emissions (Figure 2.2).

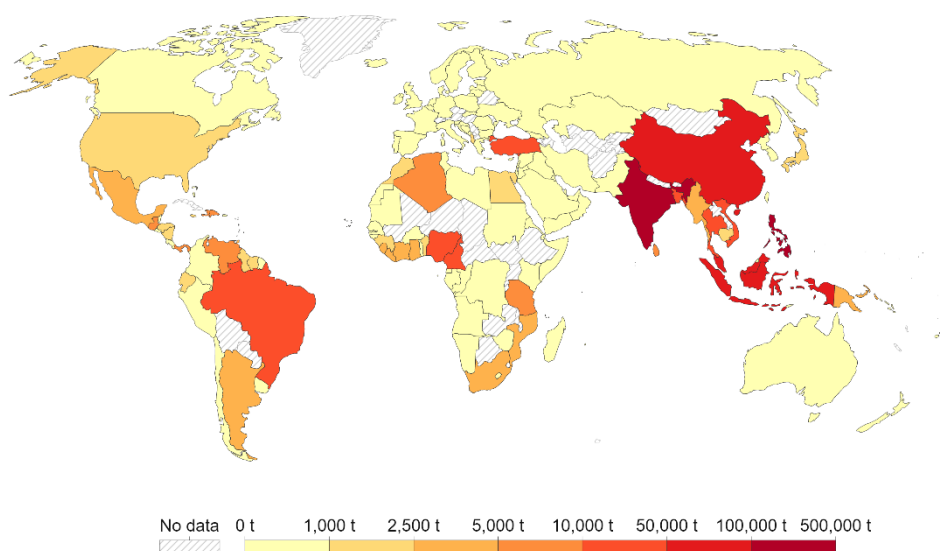


Figure 2.1: Global plastic production (Mejer et al. 2021)

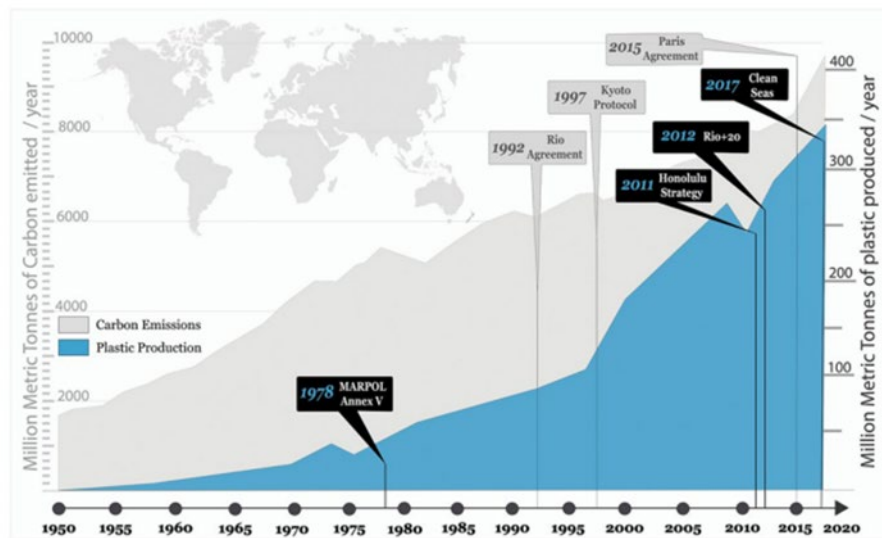


Figure 2.2: Global plastic production exceeds global carbon emission (Borelle et al. 2017).

2.7.1 South African Microplastic Contamination

South Africa is one of the largest plastic consumers and producers in Africa with a production of (Figure 1). This production is supported by the South African government as it seen as a priority sector to promote the country’s economic growth through DTI the Industrial Policy Action Plan (IPAP; the DTI 2016; Verster et al. 2017). According to Verster et al. (2017) 72% of plastic packaging is not recovered at all: 40% is landfilled and 32% leaks out of the collection system, where the packaging either is never collected and recycled or becomes part of illegal dumping. Taking these facts into account secondary microplastics are formed from fragmentation and degradation of these larger pieces of plastic if not adequately disposed of. Subsequently, these fragmented plastic particles end up in rivers, wastewater treatments, and other aquatic environments and are eventually transported to the ocean.

In recent years microplastic contamination has been on an exponential increase with numbers reaching into MMT/a with highest densities of microplastics occurring around major coastal metropolitan areas such as Durban and Cape Town (Nel and Froneman, 2015). Research gaps is imminent on less dense and presumed pristine areas. In light of this microplastic research has become an emerging research priority area in South Africa, with many of the focus of the studies being on the presence or absence of microplastics in aquatic environments, with key knowledge gaps still prevalent. Alternatively, this has changed in the last five years as more studies have emerged focussing on a diverse range of study areas including a bio diverse range

of organisms, variety of sediment types and depths, store-bought items and bivalves and surface and subsurface waters (see Appendix A for a tabulated review of microplastics research in South African waters, sediment, biota and retail stores).

2.7.2 Microplastic Sampling and Quantification Methodology

Microplastic research is globally and locally a generally new field of study with the GESAMP guidelines for sampling only being published in 2015, however many of the microplastic studies were conducted before these guidelines were set in place. Methods of sampling, extraction and records of microplastic abundances are relatively limited and unharmonized throughout the literature (Besley et al. 2017). The inconsistencies in sampling methodologies have led to inconsistencies in microplastic quantification which has become a real concern as it has become difficult to do inter-study comparisons (Van Cauwenberghe et al. 2015). The inconsistencies are not only in sampling but in the preparation of sample sets for microscopy and spectroscopy, these include differences in density separation, extraction techniques, and digestion as well as user bias.

Density separation methods aim to utilize density differences to separate different types of polymers from organic and inorganic material (Enders, 2016). There are some definitive differences in how density separation is conducted globally and locally (Table 3-8). The key differences are in the differences in the time that the samples are allowed to settle after agitation. The hypersaline mixture is mixed with the sample and agitated for (30 min to 2hrs) and allowed to settle (10 min to 24 hours) these vary amongst studies. The more buoyant microplastics will float and remain suspended. The supernatant is then filtered through filter paper or mesh. As per Hidalgo-Ruz et al. (2012) recommendations the extraction process should be repeated twice to have the best retention of microplastics however this is not the case in all studies.

Acid digestion is a reaction where strong mineral acids are used to break down biological material (Enders, 2016). Acid digestion usually takes place at room temperature over a period of 24 hours. This method is not always recommended as it causes damage to certain polymers with weaker structures and is not always the most effective in removing biological material. Acids vary across different studies, but common acids used includes: HNO₃, H₂SO₄, NaCl, KOH, KCl, CaCl₂, H₂O₂ and ZnCl₂ (Table 3-8).

Regardless of the type of sampling, density separation and digestion process conducted microscopic identification is an obligatory step to collect data. This method is subject to user bias and does not discern between plastic and other organic particles. There is a high probability

of missing microplastics in the < 1mm category and mistaking organic particles for microplastics due to similar colouring and shape. Furthermore, fragments, for example, have a higher chance of being disregarded due to their similarity in appearance to natural materials. However, Cole et al. (2014) paper suggests that microplastic counts may not always be the same due to presence of an 'operator selection biases towards fibres as they are not as easily discernible between actual plastic and biological material. Similarly Nel et al. (2017) and De Villiers (2019) found difficulty in visual identification of microplastics. Nel et al. (2017) found in her study that microplastics may be underrepresented in the results as microbeads were similar in size to actual sediment and it became difficult to distinguish between the two. De Villiers (2019) found that identification of microfibrils as plastic, based on visual inspection only, is prone to error and possible inclusion of natural fibres, it was also suggested that this may be particularly true for light-coloured fibres (white and yellow).

Daana et al. (2017) study explains that one of the issues that emerged was the prevalence of rayon fibres in the environment. Rayon, is artificial textile material composed of regenerated and purified cellulose derived from plant sources (Britanica, 2016; Daana et al. 2017) study confirms that 63% of the particles analysed by FTIR spectroscopy were rayon fibres. Other South African studies that found Rayon include: in beach sediments (Vetrimurugan et al. 2020) and juvenile fish species (Naidoo et al. 2020). The only way to surpass user bias is to make use of spectrometry to elucidate the polymers spectra.

Spectroscopy is mainly used to elucidate the elements and compounds of atoms and molecules. They are measured by examining the absorbed or emitted radiant energy by the sample or object. Spectroscopy is used to measure or identify the chemical characterization of microplastics when microscopic observation is not enough to identify the polymer. By using spectroscopy the user can get a better understanding of the composition of the polymer i.e parent materials and additives as well as the analysis of plastic associated toxic chemicals.

The two most common types of spectroscopy used to identify polymer types in microplastic research are Fourier-Transform Infra-Red (FTIR) and Raman Spectroscopy.

Raman spectroscopy is an analytical technique where scattered light is used to measure the vibrational energy modes of a sample. Raman spectroscopy is a molecular spectroscopic technique that utilizes the interaction of light with matter to gain insight into a material's make up or characteristics, like FTIR (Nandi, 2021). Raman spectroscopy is a vibrational spectroscopy technique based on the inelastic scattering of light that provides information upon the molecular vibrations of a system in the form of a vibrational spectrum. The Raman spectrum is akin to a fingerprint of chemical structure allowing identification of the components present in

the sample (Araujo et al. 2018).

Raman spectroscopy can be very laborious and time consuming as each individual particle needs to be cleaned from biological material and removed from the filter paper which runs the risk of particles being missed. Obtaining the instrumentation and accompanying parts can be quite expensive to replace. Exact focussing of the polymer for the laser is required but there is a substantial risk of the laser damaging the polymer. There is also a potential risk of not having the most accurate data as there is interference from the pigments.

Using FTIR, polymer composition can rapidly be identified by comparing the unique spectral signal of each polymer type to a library of known polymer spectral signals. FTIR Spectroscopy is obtained by an infrared spectrum of absorption or emission of a polymer and is widely used for analysing and determination of plastics and compositions. FTIR spectroscopy provides information on the specific chemical bonds and functional groups of each plastic polymer, which are easily identified with this method. The different bond compositions produce unique spectra that discriminate plastics from other organic and inorganic particles (Löder and Gerdts 2015). The FTIR comes with different components to accommodate different size classes of microplastics.

The challenges of using FTIR are that each microplastic particle has to be individually and manually selected this can be a tedious and time-consuming task because each polymer needs to be removed from the filter paper, have the biological material removed, the polymer must make contact with the metal plate and ATR. This runs a risk of particles being missed. Obtaining the instrumentation and accompanying parts can be quite expensive and to replace. Without the accompanying polymer identification library, it can be time consuming to scrutinize the spectra and one would need a good expertise in spectral interpretation.

Taking into account all the shortfalls of FTIR the respective study makes use of FTIR to validate the microscope counts and to represent the chemical composition of each polymer. The advantages of using FTIR is that there are no false positives, there is confirmation of all plastic like particles. There is a substantial reduction in false negatives. The FTIR equipment is non-destructive and doesn't compromise the integrity of the sample. The detection limit is for particles as small as 20 µm particles.

2.8 Microplastic Pollution Effects in Organisms and Aquatic Food Webs

According to Thompson et al. (2004) and Browne et al. (2008) microplastics have become an evolving problem in both marine and freshwater systems, with increased interest in understanding the effects on biodiversity. As a result, several studies carried out on biota have attempted to understand microplastics' toxicological effects (Nor and Obbard, 2014). Together the studies of (Browne et al. 2008), (Van Cauwenberghe et al. 2012) and (Lusher et al. 2013) highlight the fact that microplastics potentially pose a bigger threat than previously thought, as organisms often mistake microplastics for food. Through ingestion organisms at lower trophic levels pass on microplastics to those at higher levels through the process of bioaccumulation (Auta et al. 2017).

Wright et al. (2013) laboratory and field tests have demonstrated that ingesting and translocating microplastics adversely affect aquatic species. This view is also supported by studies on invertebrates (von Moos et al. 2012) and zooplankton (Cole et al. 2013). Whilst the following studies looked at fish (Lusher et al. 2013) and birds (Provencher et al. 2014). In addition Eerkes-Medrano et al. (2015) questions the usefulness of mainly researching marine systems when freshwater systems are more closely linked to terrestrial sources of microplastics.

According to Lusher et al. (2017) more than 220 different species were found to consume microplastics and of these, it was reported that the route of ingestion was as common as 80% in the sampled populations of invertebrate species. Cole et al. (2013) confirmed that microplastics had affected invertebrates through various methods, with ingestion being the most prominent in 7 marine phyla. Murray and Cowie (2011) maintained that ingested microplastics may result in serious physical and toxicological effects which includes obstruction of feeding appendages, aggregation, and blockage of the alimentary canal, limitation of food intake or translocation into the circulatory system. Therefore, according to the findings of (Karami et al. 2017) and (Rainieri et al. 2018) investigations of aquatic organism health should be investigated.

The academic community had extensively explored microplastics but (Karami et al. 2017) study managed to categorize the effects of microplastics into physical (as a result of colour, shape and dimension) and chemical (as a result of additives and contaminants), these features had allowed microplastics to become fatal. Nel and Froneman (2017) conducted research which looked at reef-building polychaete worms. The purpose of this study was to evaluate if the polychaete *Gunnarea gaimardi* (Quatrefages 1848) made use of microplastics to build its tube structure. Branch et al. (2010) supports (Nel and Froneman, 2017) findings that the polychaete used microplastics occurring in the water in conjunction with sand grains to build the tube structure. Comparative studies focusing on ingestion of microplastics by polychaete larvae (Setälä et al.

2016) and deposit feeders (Wright et al. 2013) demonstrated that adverse effects were more likely due to the physical effects as discussed by (Karami et al. 2017). Whereas other researchers have looked at the chemical component and have found that once ingested by the organisms, a percentage or fraction of the microplastics can be transferred to the liver resulting in lipid buildup (Lu et al. 2016) stunted growth (Au et al. 2015) immobilization (Rehse et al. 2016) and mortality (Jemec et al. 2016). Biomagnification of microplastics in food webs is thought to be a potential consequence of microplastic accumulation in natural environments (Galloway et al. 2017).

Plastic debris is known to enter food chains and anthropogenic microplastics had been described in various organisms ranging from those that form the foundation of the trophic food chain, zooplankton (Cole et al. 2013), fish (Lusher et al. 2017) to top predators (Nelms et al. 2018) and sea birds (Wright et al. 2013). Desforges et al. (2015) explains that by using model particles under laboratory conditions the movement of microplastics between trophic levels can be replicated in simple food chains. In contrast Au et al. (2015) could not fully understand the trophic transfer in natural ecosystems. However coastal food webs are thought to experience higher microplastic exposure stress, facilitating higher intakes into aquatic food webs.

Microplastics ultimately enters the food web through ingestion, entanglement, respiratory intake (inhalation) or adherence of microplastics (Watts et al. 2015). Nevertheless, ingestion of anthropogenic deposits of microplastics is believed to be the most prominent pathway of microplastic entry into biota and trophic chains (Ašmonaitė and Almroth, 2019).

2.9 Potential Impacts on Humans, Communities and Ecosystems

To date, most of the research examining potential consequences of microplastics for biota has focused on examining biological consequences at the organismal or suborganismal level (Ašmonaitė and Almroth, 2019). Due to the widespread occurrence and persistence of microplastics in the environment, they are said to physically alter biogeochemical cycling, change the dynamics of aquatic food webs, and negatively impact on large-scale ecosystem processes (Geyer et al. 2017). The introduction of microplastics into ecosystems could create a new niche that supports microbial growth and has the potential to affect carbon cycling in aquatic ecosystems (Ašmonaitė and Almroth, 2019). Ingestion of microplastics through seafood is an exposure route for humans (Smith et al. 2018) but there are indications the proximity of microplastics to humans and human microbiomes (i.e. in sewage effluent) could potentiate evolution of pathogenic species, or increase antibiotic resistance reservoirs in the environment (Ašmonaitė and Almroth, 2019). Smith et al. (2018) writes that microplastics can act as vectors

for harmful colonizing bacteria when they are ingested resulting in direct physiological effects i.e developmental, immunological, nutritional and toxicological. Comparatively, Wright and Kelly (2017) explain that although different studies have shown that microplastics can be found in food following ingestion, there had been no further data on what happens to microplastics in the body, with the effects still not well understood and still remaining a controversial topic.

CHAPTER 3: STUDY AREA AND MATERIALS AND METHODS

Name: ZANDVLEI ESTUARY NATURE RESERVE

Location: (340513.8S 182837.8E) Northwestern shore of False Bay

Hectares: 570 ha

Surface area: 1km²

Depth: 1-2 m

3.1 Study area

Estuarine systems like Zandvlei Estuary provides a wide range of ecologically important services, which includes acting as nurseries for juvenile fish species, feeding sites for resident and migratory bird species, and most importantly biological filters that break down waste and detoxify pollution (Jackson et al. 2011; van Niekerk and Turpie 2012; van Niekerk et al. 2013; Viskich et al. 2016). Estuaries are used for aesthetics, sports and cultural practices, however these ecologically sensitive areas face great adversity as they are among the most modified and threatened of all aquatic environments (Blaber et al. 2000; McQuaid 2013). There are only 300 functional estuaries along the south Africa coast and are divided into warm-temperate South and subtropical East coasts, with only 16 being on the cool temperate West Coast, west of Cape Point (van Niekerk et al. 2015). Zandvlei Estuary is a typical example of estuarine systems and the long-term modifications and disturbances estuaries are subjected to (Clark, 1998). Despite all the human manipulation Zandvlei Estuary still remains the only functioning estuary in the False Bay region (Heydorn and Grindley, 1982).

3.1.1 Spatial delineation of Zandvlei Estuary

One of the central and most dominant features of this estuary is the only seasonal estuary found in the far northwestern shore of False Bay (Hawly, 1999). The full area of Zandvlei estuary is bound by four high traffic roads, Prince George Drive East, Military Road North, Main Road west and Royal Road South which links up to Prince George Drive and runs above the estuary mouth. This link between the two roads follows the approximate high tide mark of 237 m (Zandvlei Trust, 2022). The adjacent Marina Da Gama canals constitute artificial estuarine habitat, but they are a connected and functioning part of the Zandvlei estuarine system (Hutchings et al 2016).

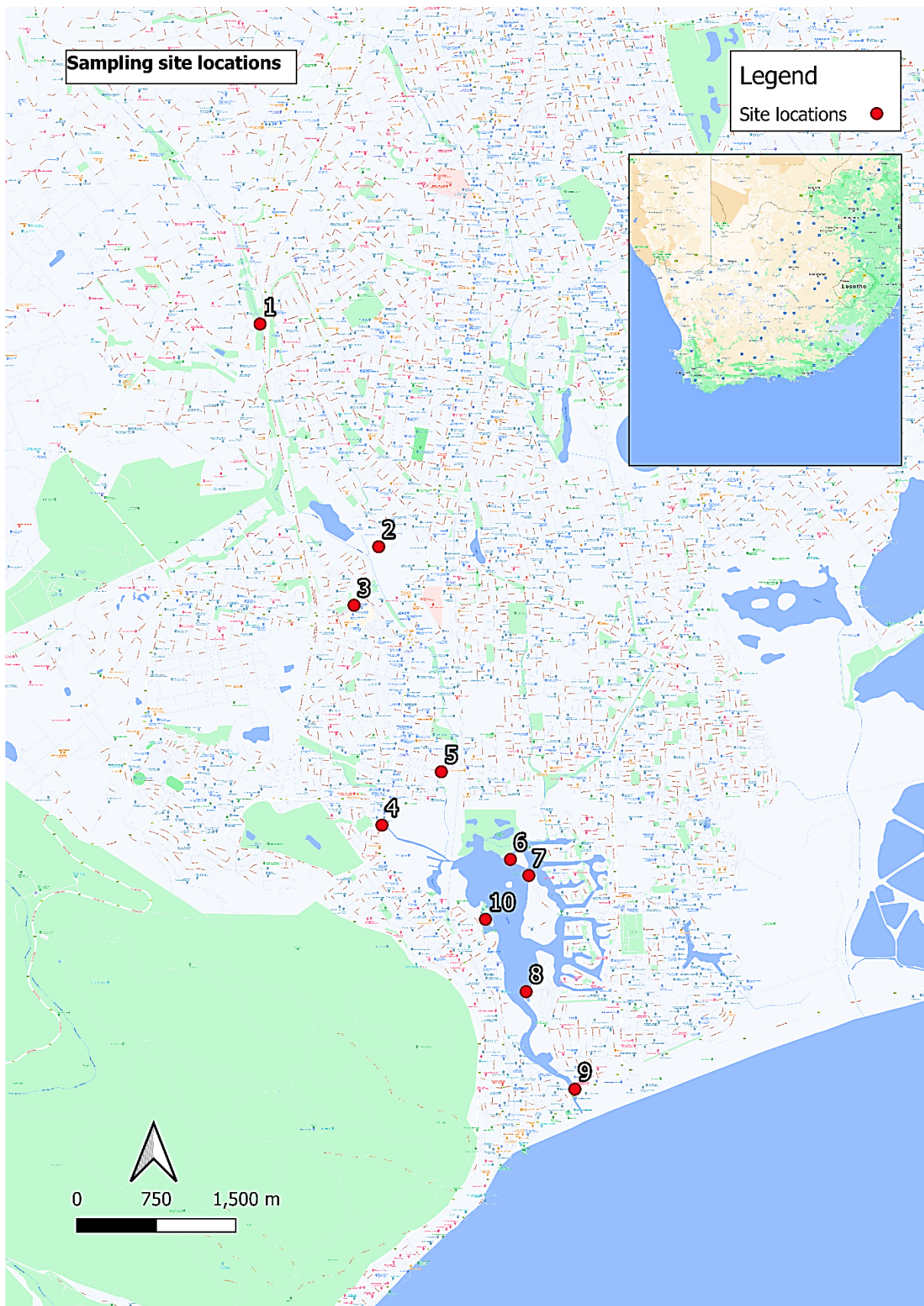


Figure 3.1: Spatial extent of the Zandvlei Estuary defined for the purposes of this study

3.1.2 Catchment and hydrology

Zandvlei estuary's catchment is approximately 92km² and bordered by Muizenberg Mountain, Silvermine Plateau and Constantiaberg to the West, and Wynberg Hill to the North (Hutchings et al. 2016). Due to the Mediterranean climate, the seasonal streams flow in winter after the rainfall and stop flowing in the drier summer months. The key streams which drain into the estuary are the Westlake Stream, Keysers River, Langvlei Canal and the lower end of the Diep River, known as the Sand River Canal (Quick & Harding, 1994). The Westlake and Keysers rivers converge and enter the north west of the estuary via a reed filled wetland while the Langvlei and Sand rivers which flow in constructed concrete canals converging and entering the estuary in the north east (Figure 3.1).

The current contribution of these aquifers to the hydrology of the system is now likely severely limited due to the extensive canalisation of some of the river courses in their lower reaches which prevents movement of water into the system through interflow or baseflow. This indicates that the majority of the water entering the system is coming from overland flow transported by the inflowing rivers. As a result of the altered canalization, weir construction and artificial mouth management the tidal influence is greatly influenced (Thornton et al. 1995). The mouth is opened artificially by the City of Cape Town's Catchment Management Department when a high spring tide is expected (CoCT, 2010).

3.1.3 Physical and chemical components

According to Hutchings et al. (2016) some earlier maps suggest that the mouth of the Zandvlei estuary was much wider than in the past and potentially a periodical shift of the mouth could have occurred. Subsequently, if no human modifications had occurred the estuary would have been open to tidal flushing most of the year maintaining water heights of between 0 m and 0.3 m above Mean Sea Level (aMSL). During late summer months when the river inflow was at its lowest, the mouth would have closed, resulting in a gradual increase in water level, potentially reaching as high as 2.5-3 m aMSL, flooding a much larger area than that which is inundated currently. The inflowing freshwater from the rivers would have then caused the salinity in the estuary to drop. At some point the mouth would be breached by the high-water level behind the natural sand beam which would result in flushing out most of the water, transporting a large amount of sediment along with it. The newly opened and widened mouth would then allow a tidal influence, once again increasing the salinity in the estuary. Additionally, according to Thornton et al. (1995) the combination of high wind velocities that run parallel to the estuary and the shallow depth allows for the vlei to be well mixed in most parts of the year.

3.1.4 Conservation importance of Zandvlei Estuary

Farming, urban development, invasive alien plants and dredging of the vlei have destroyed much of the natural vegetation around Zandvlei (CoCT, 2010). While the estuary has undergone many changes and degradation, it still acts a range of habitats for many species. IUCN red listed species are found within the Zandvlei estuary and surrounds. The estuary does provide some areas of mostly intact habitat, although large portions of the margins are completely converted and of little conservation value, however rehabilitation may be possible for some of these areas. The estuary is also listed as an important estuary in terms of providing a nursery habitat for estuarine dependent fish, amphibians and birds. The estuary has a relatively rich species of fish and avifauna which utilizes the estuary, with 40 fish species and 173 bird species being observed and of the 173 bird species six is listed on the red data species list (Gibbs et al. 2011; Hutchings et al. 2016). The only listed species on the IUCN Red List is the Endangered Western Leopard Toad that is known to breed in the upper estuary and surrounding wetland, where it completes its entire lifecycle in and around the estuary itself. In addition, Zandvlei Estuary can be described as the only estuary of real importance as a fish nursery along the False Bay coast (Quick & Harding, 1994). Lastly, the estuary performs a vital role in sustaining the fishing industry in False Bay as fish leave the estuary when they mature, and also provide for the piscivorous birds which frequent the area (Thornton, 1995).

3.2 Materials and Methods

3.2.1 Sample collection

Samples were collected seasonally between the years 2018 and 2021 (Table 3.1). Due to the COVID-19 pandemic sampling was not able to take place as planned and was restricted to certain seasons and sites.

The reaches of the whole system was categorized into the following:

Upper Reaches: site 1 of Catchment and 6-7 of Vlei;

Middle Reaches: sites 2-3 of Catchment and 8 and 10 of Vlei;

Lower Reaches: sites 4-5 of Catchment and 9 of Vlei.

Table 3.1: Sampling layout for study area, reaches and sites

Year	Season	Catch/Vlei	Sites	Reaches
2018	Autumn	Vlei	6-10	Upper Middle Lower

2019	Winter	Catchment	1-5	Upper Middle Lower
2020	Summer	Catchment and Vlei	1-10	Upper Middle Lower
2021	Autumn	Catchment and Vlei	1-10	Upper Middle Lower

Water collection

Surface waters were sampled from Zandvlei Estuary catchment and vlei area. Five replicates per site were captured with a 20 L bucket. Five replicates were collected at each site by pouring four separate 20 L buckets of water through a 250 µm sieve 5 times. The retained particles were rinsed with deionized water concentrated into a falcon tube. Buckets with 20 L of surface water from each site were taken back to the laboratory for processing. Falcon tubes were stored in a freezer at -20 °C until processing.

Sediment collection and granulometric analysis

Sediment samples were collected from the top 5 cm (oxygenated zone) using a spatula. Five 0.5 m x 0.5 m quadrats per site located a minimum of 5m apart in undisturbed areas as per recommendations of (Hidalgo-ruz et al. 2012). One kilogram of sediment was collected and taken back to the laboratory for processing. Sediment samples were decanted into aluminium containers and covered with additional aluminium foil. Each aluminium container had a sample of 500 g. These samples were split into A and B containers. All the A containers were used for grain size analysis and B containers for microplastic analysis.

3.2.2 Laboratory analyses: digestion and extraction

Water

Prior to processing, surface water samples were removed from the freezer and defrosted at room temperature (± 25 °C). The sample was manually agitated for 2 minutes and surface water samples vacuum-filtered (model Rotary Vane VP 145 1/3 HP) onto 20 µm nylon filters. The same was done for the 20 L bucket water. Each filter was placed into a new petri dish and allowed to dry before analysis. The samples were visually sorted under a dissecting microscope

(Olympus SC30). Microscopic particles were identified by possessing unnatural shape, type, coloration and size. This was recorded as microplastics per litre (MP/L) and converted accordingly.

Sediment

Samples were placed into a drying oven (model DHG 9070A) and dried at 60 °C for 48 hours until constant mass as per (Naidoo et al. 2015). Each of the A sediment samples underwent grain size analysis whereby the sample was placed in universal test sieves and sieved. Remains were weighed at the following intervals as per the Wentworth scale 500, 250, 125, 63 and the retriever ($63 > \mu\text{m}$).

An adjusted floatation technique was used to extract microplastics from B sediment samples (500g). Density separation was used to separate the sediment from the microplastics. A fully saturated Sodium Chloride (NaCl) solution was prepared by adding 360 g of commercially available iodated table salt to a beaker containing 1 L of distilled water and a magnetic stirrer bead. The beaker opening was covered with aluminium foil and placed on a magnetic stirrer. The hypersaline solution was mixed using a magnetic stirrer at a high speed at room temperature (25 °C) for ten minutes or until the solution became saturated. Commercially available table salt have been found to contain microplastics (Yang et al. 2015; Karami et al. 2017), therefore the NaCl solution was vacuum-filtered through a 20 μm nylon mesh before use. The supernatant was collected and 10% KOH (w:v) acid solution was added and placed in an oven for 48 h at 60 °C.

Each dried sediment sample was weighed to the nearest 0.001 g and thereafter mixed with 200 mL of saturated NaCl solution with a glass rod in a 250 mL glass beaker (both previously rinsed with deionized water) for 2 minutes. The salt-sediment mixture was left to settle for 15 minutes until the sediment has visibly settled. As per recommendations of (Yonkos et al. 2014) and (Besley et al. 2017). The extraction procedure was repeated three times per sediment sample in order to improve microplastic recovery rates. Microscopic particles were identified by possessing unnatural shape, type, colours and size. Concentrations were converted to MP per kg/meter and recorded as MPs/Kg dry weight (dw) MPs/m².

3.2.3 Microscopic identification of microplastics

For the most part visual identification of microplastics is considered to be one of the most rapid and technically simple methods, by categorizing plastics based on their morphological differences (Lusher et al. 2017). To aid in the identification of microplastics a dichotomous key was developed amongst senior students, by using recommendations of (Gerber, 2017) and

other previous studies for identification. Additionally, microplastic counts were validated by at least one other counter.

Table 4.2: Guideline for visual identification of microplastics under microscope

Visual	Present	Reference
<5 mm	Yes (Proceed to 2) No (No not microplastic)	(Fok & Cheung, 2015)
Organic material present	Yes (Not microplastic) No (Fibre like-proceed to 3) Other (proceed to 6)	(Weideman et al. 2020a)
Fibre is uniform throughout	Yes (Proceed to 4) No (Not microplastic)	(Halstead et al. 2018)
Is the fibre frayed?	Yes (Proceed to 5) No (Not microplastic)	(Gerber, 2017)
Homogenous in colour throughout length	Yes (Microplastic) No (Proceed to 6)	(Fotopoulou & Karapanagioti, 2012)
Evaluat using fine tweezers and hot needle test	Yes (Microplastic) No (Not microplastics)	(Silva et al. 2018)

Individual filter papers for water and sediment were visually examined under a dissecting microscope (Olympus SC30) equipped with a camera (DinoCapture Camera V1.5.39.C). Microplastics were identified based on the following characteristics: size, colour and type. Microplastic size classes categorized as: <1000, 1000-5000, >5000 μm . Microplastic colour categorized as: White, Black/Grey, Blue/Green, Red/Orange/Pink, Yellow/Brown, Other and Transparent. Microplastic type categorized as: Fibre, Fragment, Sphere, Film, Nurdle and Foam. Reverse Osmosis water was filtered through a 10 μm mesh and used for all solutions of samples processed.

3.2.3 Polymer Identification using spectrophotometry

Suspected plastic particles above 500 μm were removed from filter papers using fine tweezers, for validation using a Perkin Elmer Two FTIR-ATR Spectrometer. The microplastics were compressed against either the diamond or flathead at a force of at least 80 N. The spectra were recorded in the wavenumber ranging from 4000 to 450 cm^{-1} with a resolution set to 4 cm^{-1} and a data interval of 1 cm^{-1} and scans set to 10. Before each particle was analysed the ATR crystal was cleaned with 70% propanol and background scans done prior to each sample analysis. ST

Japan Polymer and Polymer Additives Library was used to verify polymer types and scans 70% and above were accepted. The respective polymers' spectral was plotted using Origin 9 software.

3.2.4 Risk assessment

Microplastics indices were applied in a similar manner as metals in order to provide comparative assessments of the potential effects of MPs (and APPs), with risk categories presented in Table 1. The MPs contamination factor (MPCF) assesses the concentrations of MPs ($C_{microplastic}$) compared to background concentrations.

$$MPCF_i = \left(\frac{C_{microplastic}}{C_{baseline}} \right)$$

where the $C_{baseline}$ value selected was the average microplastics sediment concentration for site 1 (control site) as there are no historic values for the region and this method is considered acceptable (Kabir et al. 2021). Microplastic pollution index (MPPLI) calculations were similar to that of metals.

$$MPPLI_{site} = \sqrt[2]{MPCFr \times MPCFi}$$

Where MPCFr and MPCFi were MPCFs for fragments and filaments, respectively. The chemical toxicity of polymers were analysed based on the method by Lithner et al. (2011), where hazard scores are assigned to polymer types to assess the risk of polymers.

$$H_i = \sum P_n \times S_n$$

where H_i is the calculated polymer risk index, P_n the ratio of a polymer type recorded at a site and S_n the polymer hazard score assigned by Lithner et al. (2011). The pollution risk index (PRI) is calculated as follows.

$$PRI_i = \sum H_i \times MPPLI_{site}$$

Where PRI_i indicates the ecological hazard of polymers when associated with the polymer risk index (H_i).

Table 4.3: Risk categories of indices for microplastic contamination in Zandvlei Estuary and Catchment, Cape Town

Risk Category:	Low (I)	Moderate (II)	High (III)	Very High (IV)	Dangerous (V)
Polymer Risk Index (H)	< 10	10 – 100	101 – 1000	1000–10,000	> 10,000
Pollution Risk Index (PRI)	< 150	150 – 300	300 – 600	600 – 1200	> 1200
Contamination Factor (CF)	< 1	1 – 3	3 – 6	> 6	
Pollution Load Index (PLI)	< 1	1–3	3–4	4–5	> 5

3.2.5 Quality Control

Method blanks and controls were set up in order to ascertain if there was any contamination during the sampling and laboratory processing. Clean petri dishes with 20µm filter paper was left exposed during filtration in order to determine if there was any airborne contamination. Blanks (10) for foil, falcon tubes and sediment bags were taken by rinsing these items with MilliQ ultra-pure water filtering it onto 20µm filter paper. The following protocols were observed in the laboratory as per (Lusher et al. 2014) suggestions: Cotton lab coats were worn, all containers used during processing were rinsed with distilled water, collecting samples in aluminium foil and using all metal equipment (scoops, tweezers), covering samples with aluminium foil, using glass materials previously washed with acid and MilliQ ultra-pure water, and opening samples only when strictly necessary (Prata et al. 2020). The jars used for digestion as well as the petri dishes were rinsed three times with MilliQ ultra-pure water and blank control samples for jars taken. As far as possible, no plastic items were used in the lab. MilliQ water was used to make up all solutions used i.e hypersaline solutions and acid solution NaOH. Petri dishes were always kept closed and only opened when being processed under the microscope to record microplastics.

3.2.6 Data analysis

Univariate statistics were conducted using IBM SPSS Statistics® (Version 28 for Microsoft® Windows® 10). Statistical tests for normality using the Kolmogorov-Smirnov test and equal variance was conducted using the ANOVA test. The data did not meet either of the assumptions to do parametric analyses. Even after transformations of the data. Non-parametric tests were subsequently done using the Kruskal-Wallis (KW) for multiple groups (sediment, water250 and water20) and the Mann-Whitney (MW) test between two groups (eg. catchment vs vlei). Non-parametric Spearman Rank correlations (Rs) were used to assess relationships between groups. Results are reported as means, variances as standard error of the mean (\pm SE) and significances set at $p < 0.05$.

CHAPTER 4: RESULTS

4.1 Quality Control measures

Microplastic (MP) quality control/assurance of samples were taken in field sampling by leaving glass jars open during the duration of sampling events to ascertain if any airborne contamination took place. These jars were rinsed with RO water and filtered onto filter paper. In the lab airborne contamination was controlled by placing empty wet petri dishes on workbenches for the duration of all lab work. These positive controls were checked at the start and end of each day and any contamination recorded. A total of 58 fibres were recorded for the duration of the study and the data adjusted accordingly. Blanks (negative controls) were included in all sample filtrations and no MPs contamination reported. Extraction efficiencies were done by filtering known quantities of filaments and fragments. Extraction efficiencies for fibres recorded recovery rates of 83-85% for MPs between 250 and 5000 μm in size and 91-94% for fragments.

4.2 Microplastic Abundances

Microplastics were sampled in the catchment area and vlei of the Zandvlei catchment system between 2018 and 2021. Samples were collected in water and sediment, seasonally (the COVID pandemic interrupted sampling) and at 10 sites in total. Microplastics were recorded in all estuarine surface water samples and estuarine sediment samples. A total of 485 samples were processed with a total of 18157 MPs counted. Based on respective units (per L for water and per Kg for sediment), a total of 56704 MPs were recorded. Water samples were sieved through 250 ($N = 170$) and 20 μm ($N = 220$) sieves and 190 sediment samples processed. No microplastics were found in blank samples.

MPs were highest in sediment samples with an average of 293 (± 37.1 SEM) MPs/Kg dw followed by water sieved through a 20 μm mesh (5.8 ± 0.5 MPs/L) and lowest in water sampled sieved through a 250 μm mesh (2.5 ± 0.4 MPs/L) (Figure 4.1a). Based on area, the MP concentrations had similar trends with highest average MP concentrations recorded in sediment (144 ± 15.2 MPs/m²), 4.9 ± 0.5 MPs/m² in water filtered through a 20 μm mesh and 2 ± 0.4 MPs/m² in 250 μm filters water (Figure 4.1b).

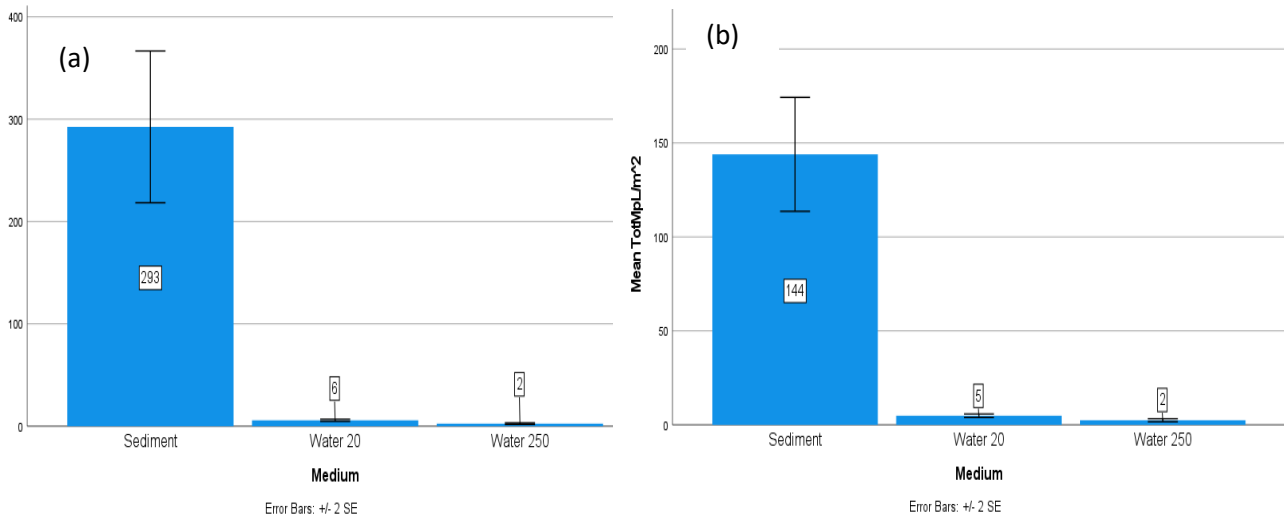


Figure 4.1: Microplastic concentrations in sediment, 20 µm and 250 µm filtered water based on (a) weight (MPs/Kg dry weight) and (b) area (MPs/m²)

Overall, most microplastics were found in sediment samples (39.7%) among all the sites, with surface water 20 µm (25.77%) and surface water 250 µm (35.05%) accounting for relatively less of the total microplastic abundance. Most microplastics were found in combined sediment and water samples from the Sandriver Canal (17.73%), with relatively fewer from the Keyzers River (17.92%) and Westlake River (9.07%) (Figure 4.2).

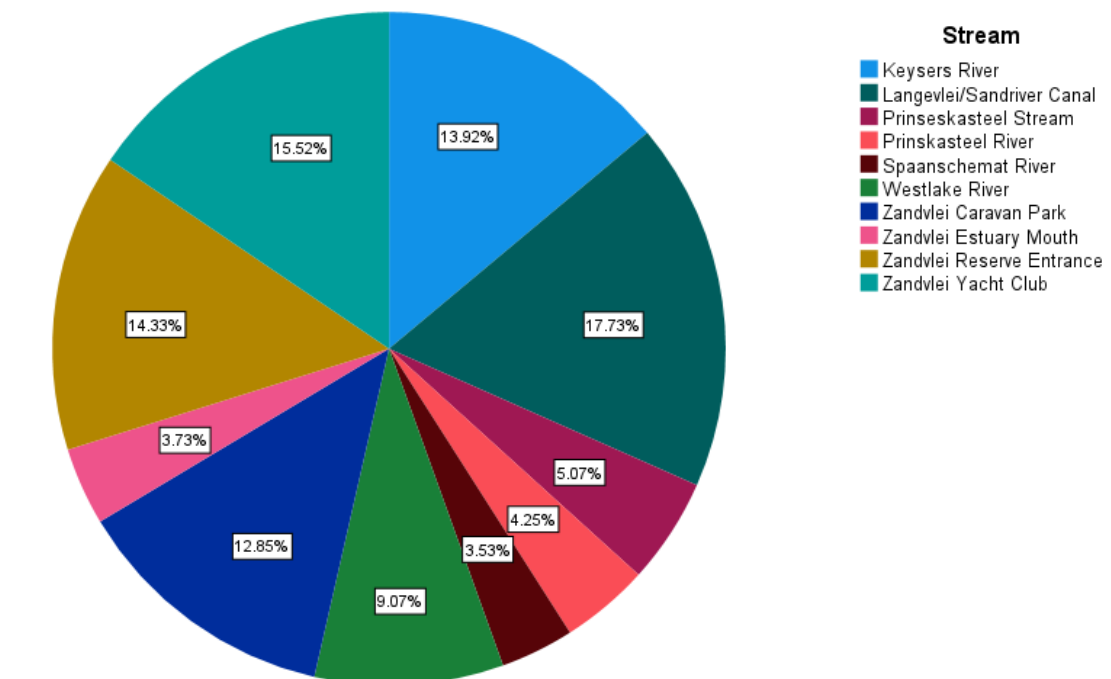


Figure 4.2: Microplastic composition based on input rivers, streams and sites

4.2.1. Characteristics of microplastics in areas, reaches and sites (spatial) in different media (water & sediment)

Microplastics were highest in the vlei across all mediums with an average of 395 particles in sediment MPs/Kg dw, followed by water sieved through a 250 µm mesh 4 particles MPs/L with water sieved through 20 µm mesh in both vlei and catchment 6 particles MPs/L (Figure 4.3a). Based on the area the results tend to show a similar trend where the highest MPs can be found in the vlei 152 particles, water sieved through 250 µm mesh 4 particles, except for water sieved through a 20 µm mesh where the vlei had more MP particles (Figure 4.3b).

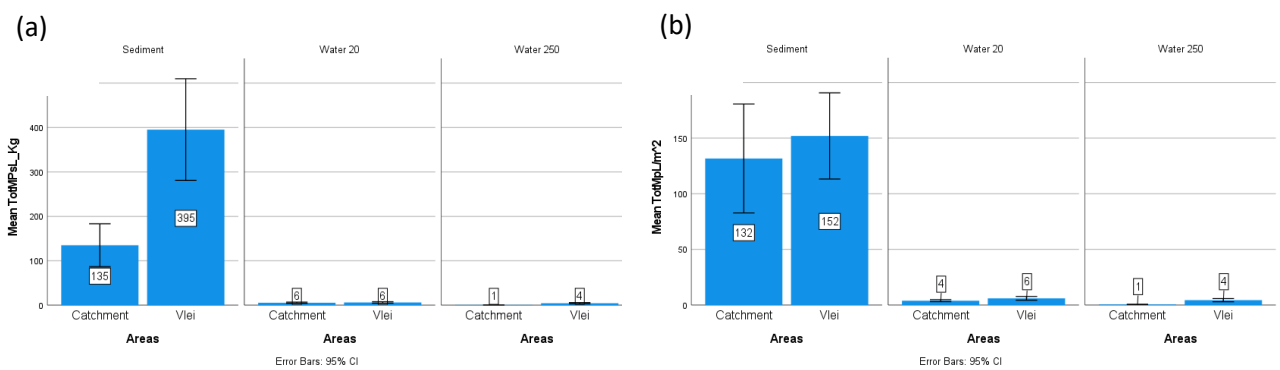


Figure 4.3: Microplastic concentrations in catchment and vlei sediment, 20 µm and 250 µm filtered water based on (a) weight (MPs/Kg dry weight) and (b) area (MPs/m²)

MPs were highest in the middle reaches sediment 164.5 MPs/Kg dry weight, followed by water sieved through 20 µm mesh 7.3 MPs/L in lower reaches and water sieved through 250 µm mesh 0.7 particles in the lower reaches (Figure 4.4a). Results based on the area do not follow the same trend as all microplastic abundances throughout the different mediums peak in different reaches (Figure 4.4b).

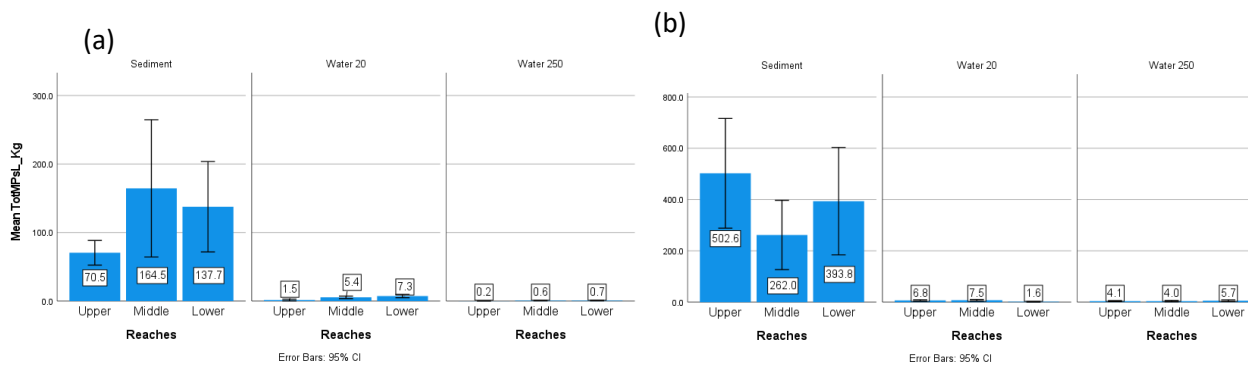


Figure 4.4: Microplastic concentrations based on reaches, sediment, 20 µm and 250 µm filtered water based on (a) weight (MPs/Kg dry weight) and (b) area (MPs/m²)

MPs were highest in sediment across all sites, followed by water sieved through 20 µm mesh

and then water sieved through 250 μm mesh (Figure 4.5a). MPs were highest in the upper reaches for sediment but this trend is dissimilar for both water sieved through 20 μm mesh and 250 μm mesh as MPs peak in the middle and lower reaches (Figure 4.5b).

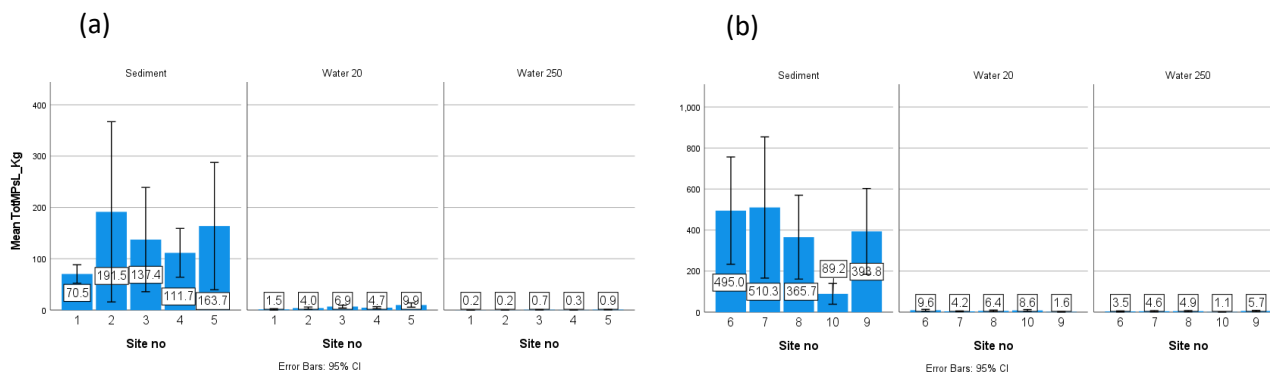


Figure 4.5: Microplastic concentrations across sites in sediment, 20 μm and 250 μm filtered water based on (a) weight (MPs/Kg dry weight) and (b) area (MPs/m²)

4.2.2 Temporal characteristics of microplastic abundances (season and year)

MPs were highest in 2018 (475 particles) and lowest in 2021 (28 particles) (Figure 4.6a). Total MPs are highest in autumn 183 particles MPs/Kg dw followed by winter (79 particles) and then summer (79 particles) Figure 4.6b). The same yearly trend follows for Mps per area but not for season, MPs were highest in winter followed by autumn and summer (Figure 4.6 c-d).

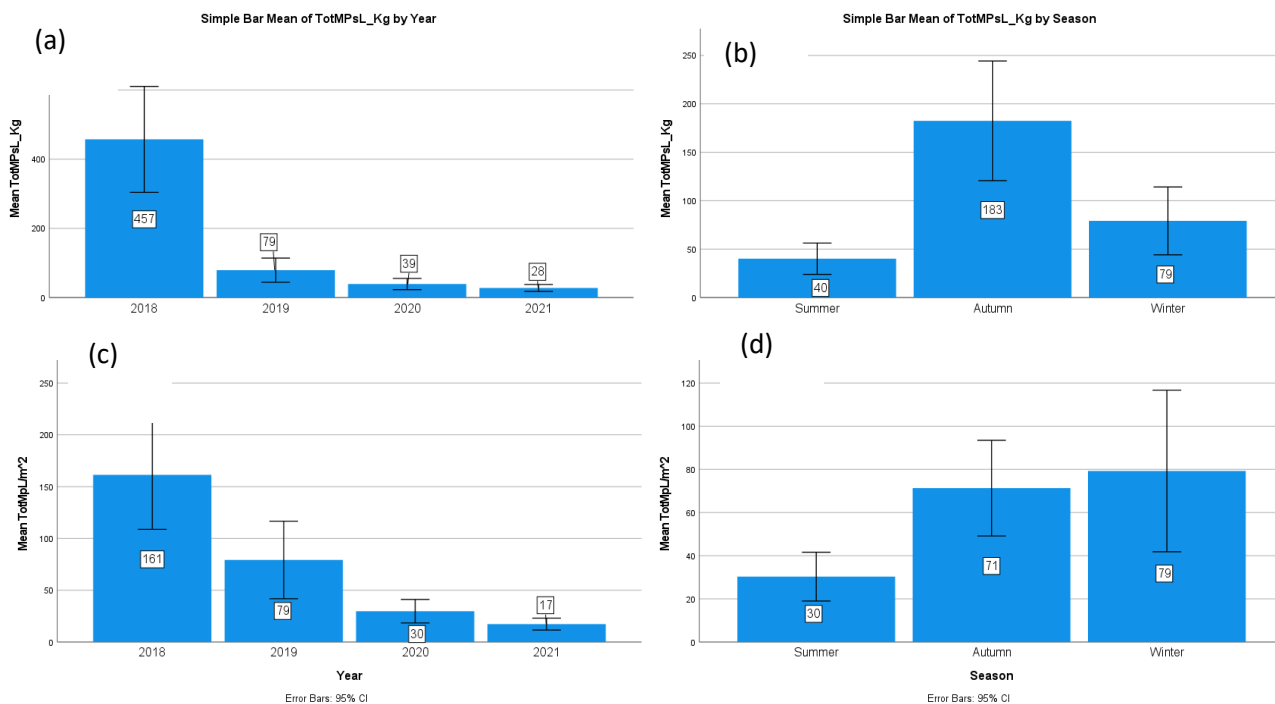


Figure 4.6: Yearly and seasonal microplastic concentrations based on weight (MPs/Kg dry weight) (a-b) and area (MPs/m²) (c-d)

MPs were highest in 2018 for sediment (962 particles MPs/Kg) and lowest in 2021 (79 particles) (Figure 4.7a). MPs were highest in 2019 for water sieved through 20 µm mesh (8.7 particles) and lowest in 2018. MPs were highest in 2018 (8.2 particles) and lowest in 2021 for water sieved through 250 µm mesh. The trend remains similar for area with the only deviation being MPs was lowest in 2019 for water sieved through 20 µm mesh (Figure 4.7 c).

MPs/Kg dry weight in sediment were highest in autumn (470 particles), more than twice the concentrations of summer and winter (Figure 4.7b). Water sieved through 20 µm mesh and 250 µm mesh do not follow the same trend as MPs concentrations are highest in winter in water sieved through 20 µm mesh and water sieved through 20 µm mesh. The trend remains similar for area in terms of sediment and water 250 µm expect for MPs is highest in winter and lowest in summer (Figure 4.7d).

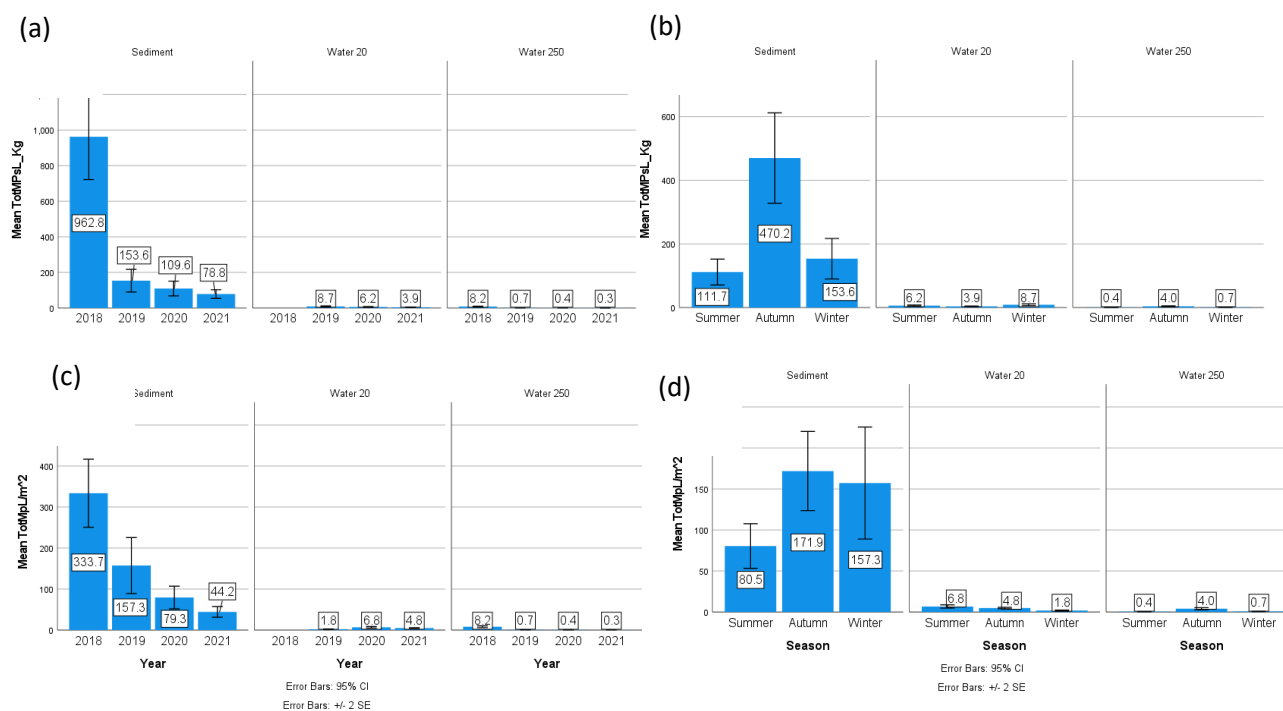


Figure 4.7: Yearly and seasonal microplastic concentrations in sediment, 20 µm and 250 µm filtered water based on (a-b) weight (MPs/Kg dry weight) and (c-d) area (MPs/m²)

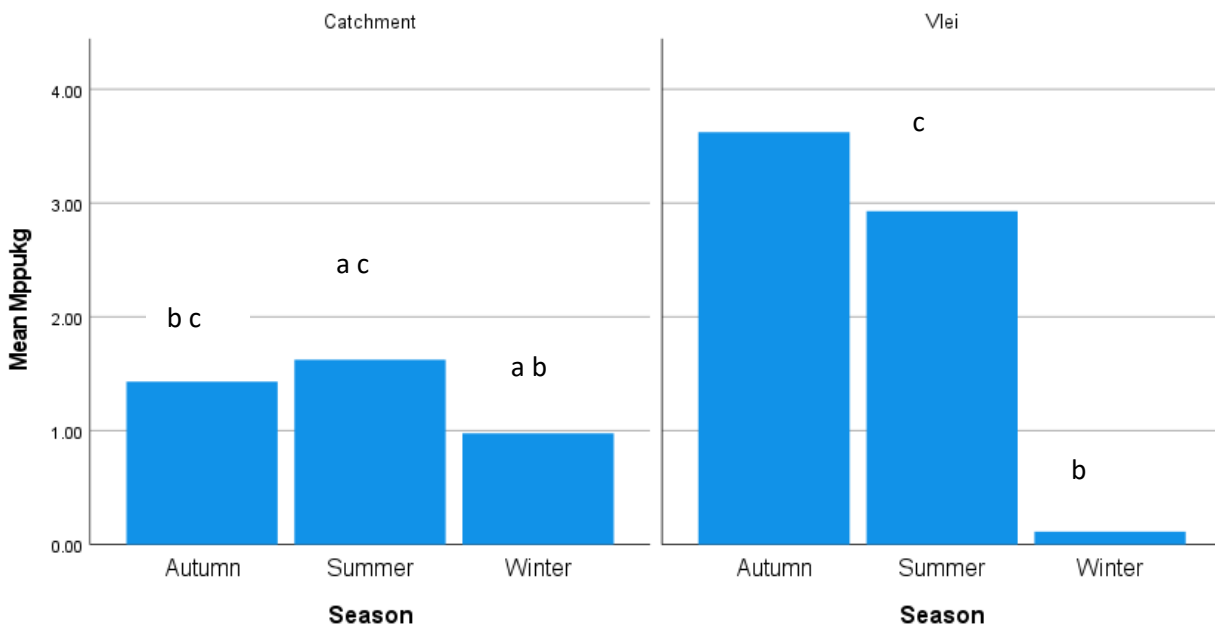


Figure 4.8: Statistical significances between catchment and vlei across seasons

Relationships between all seasons in the catchment are all statistically significant and relationships between summer and winter in the vlei are statistically significant at $p > 0.05$ (Figure 4.8).

4.3 Microplastic characteristics (type, colour and size)

4.3.1 Microplastic abundances based on type

Microplastic fibres were most abundant across all mediums (Figure 4.9), all regions (catchment and vlei) (Figure 4.10), all reaches (upper, middle and lower) (Figure 4.11), all years (2018 to 2019) (Figure 4.12) and all seasons (summer, autumn and winter) (Figure 4.13). For all categorical criteria above, fragments were generally the second-most abundant MP type recorded.

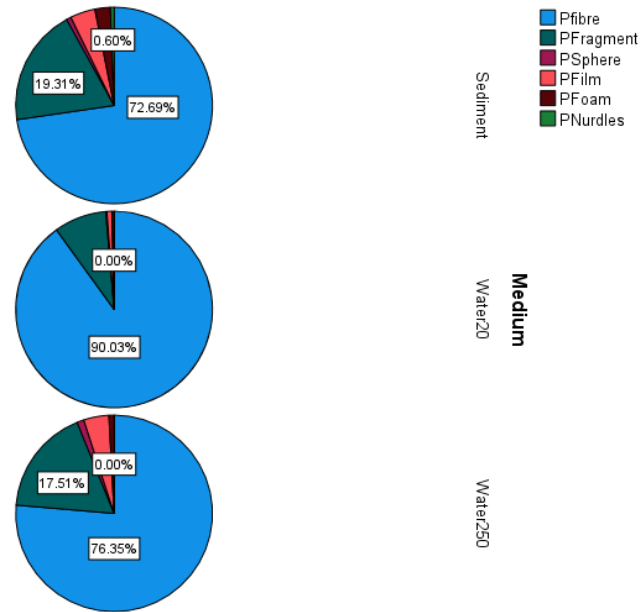


Figure 14.9: Pie charts depicting ratio of morphotype ratios across medium

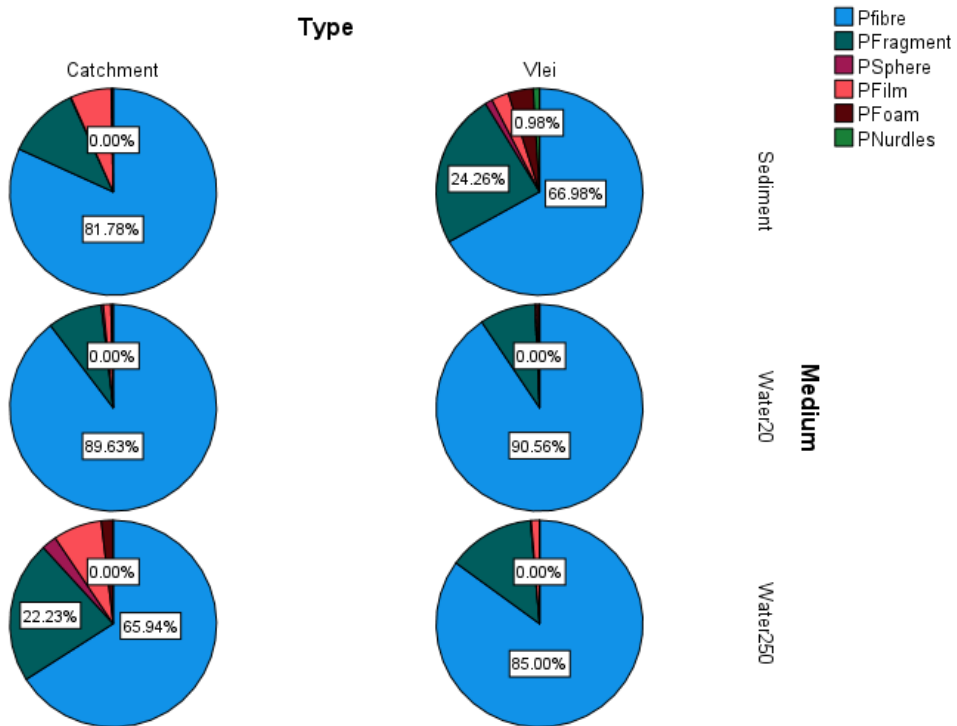


Figure 4.10: Pie charts depicting ratio of morphotypes across region and medium

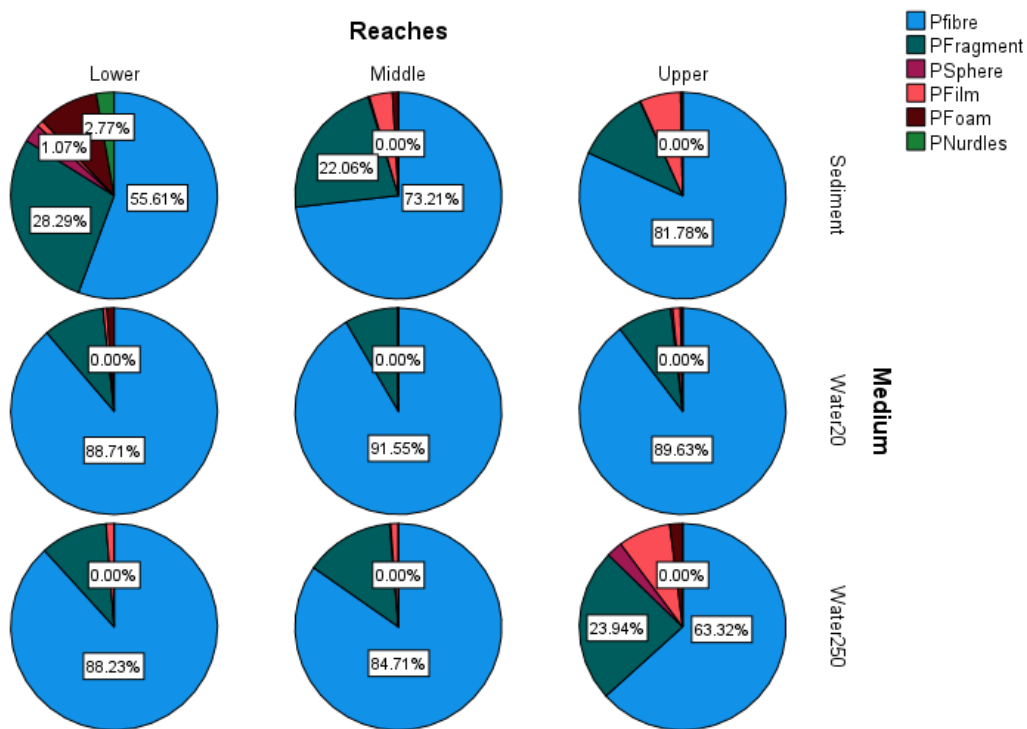


Figure 24.11: Pie charts depicting ratio of morphotypes across reaches and mediums

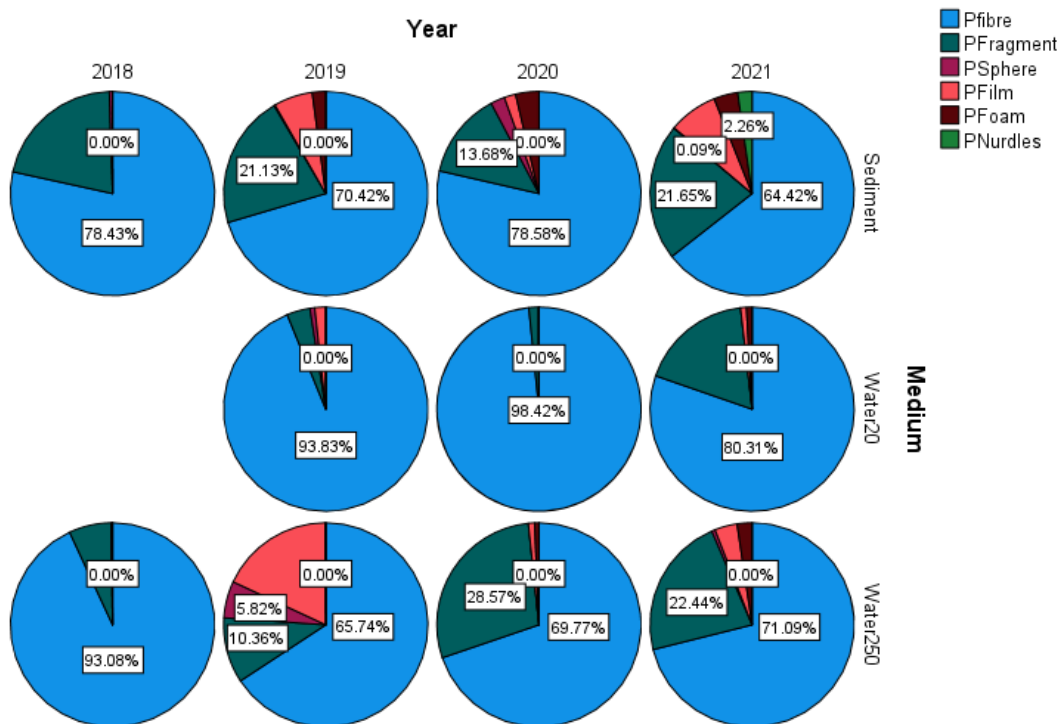


Figure 4.12: Pie charts depicting ratio of morphotypes across mediums and years

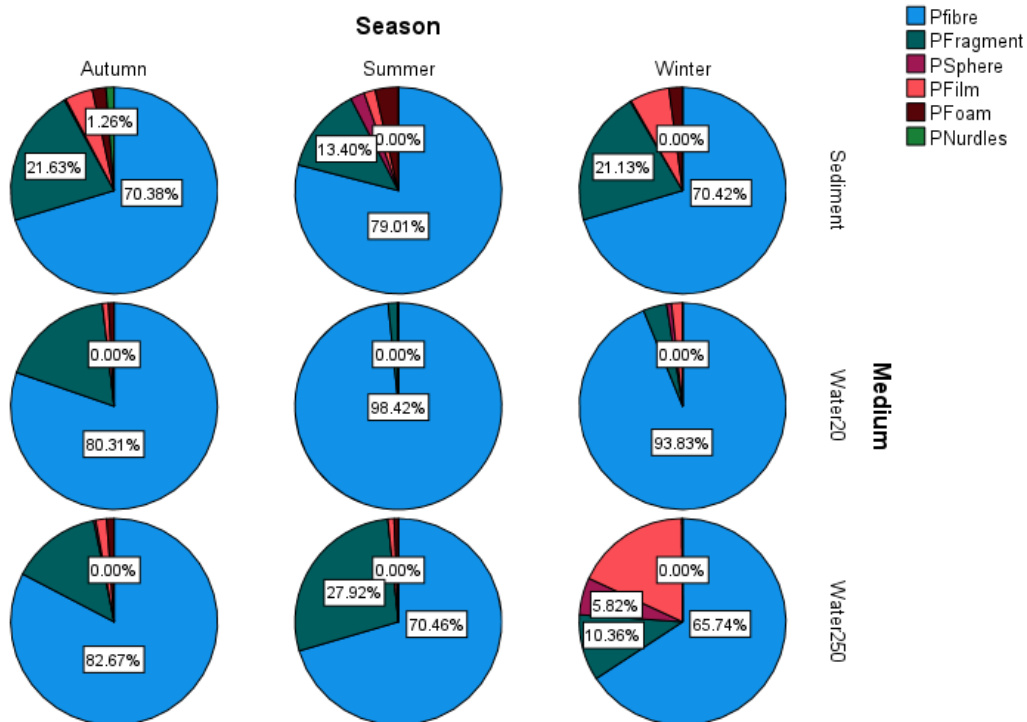


Figure 4.13: Pie charts depicting ratio of morphotypes across seasons and mediums

4.3.2 Microplastic abundances based on colour

Transparent MPs were generally most abundant across all mediums (sediment, water20 and water 250) (Figure 4.14), regions (Figure 4.15), reaches (Figure 4.16), years (Figure 4.17) and seasons (Figure 4.18). However there were exceptions where grey/black was more predominant in catchment water 250 μm (33.23%), upper reaches water 250 μm (35.69 %), water 250 μm 2020 (41.33%), sediment 2018 (33.61%), water 20 μm winter (31.59%) and water 250 μm winter (31.31%).

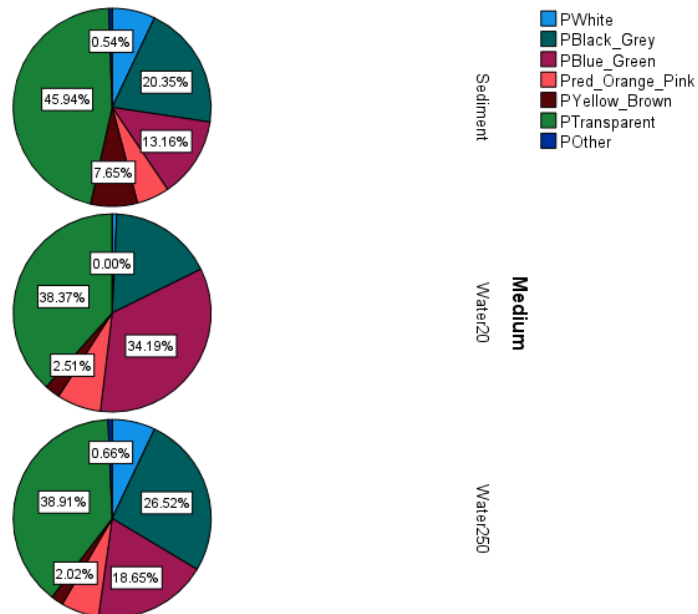


Figure 4.14: Pie chart ratio based on colour and medium

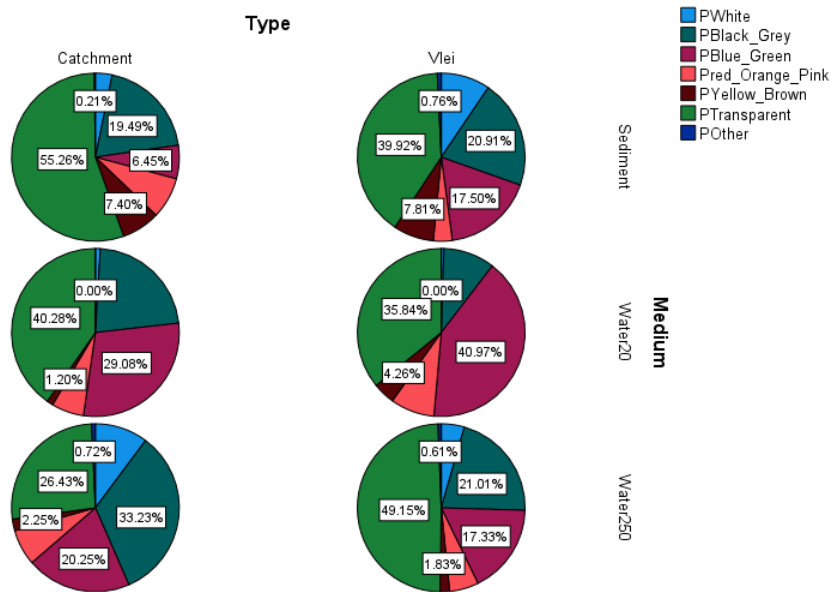


Figure 4.15: Pie charts ratio based on colour, medium and region

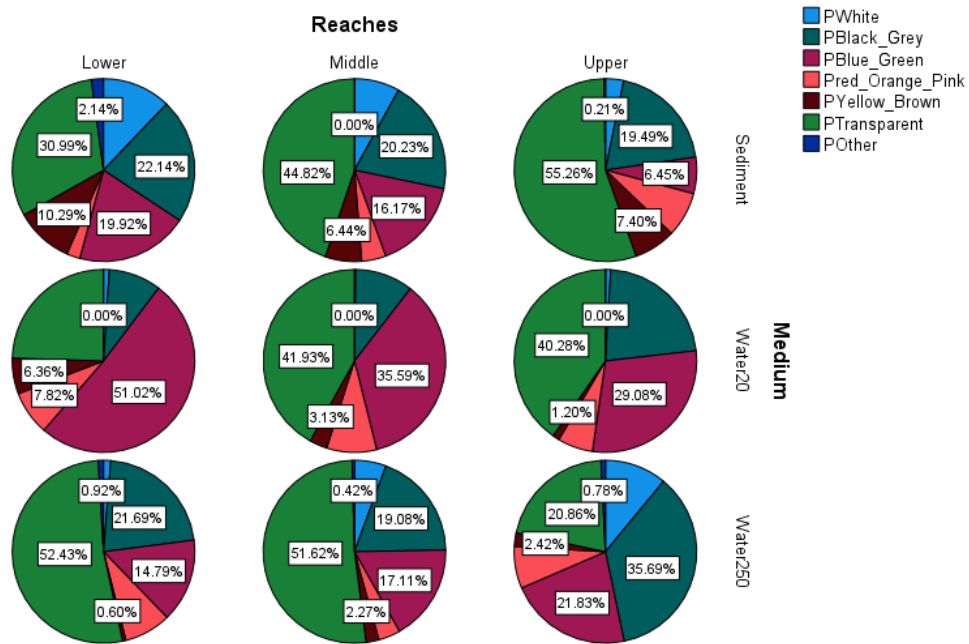


Figure 4.16: Pie charts ratio based on colour, medium and reaches

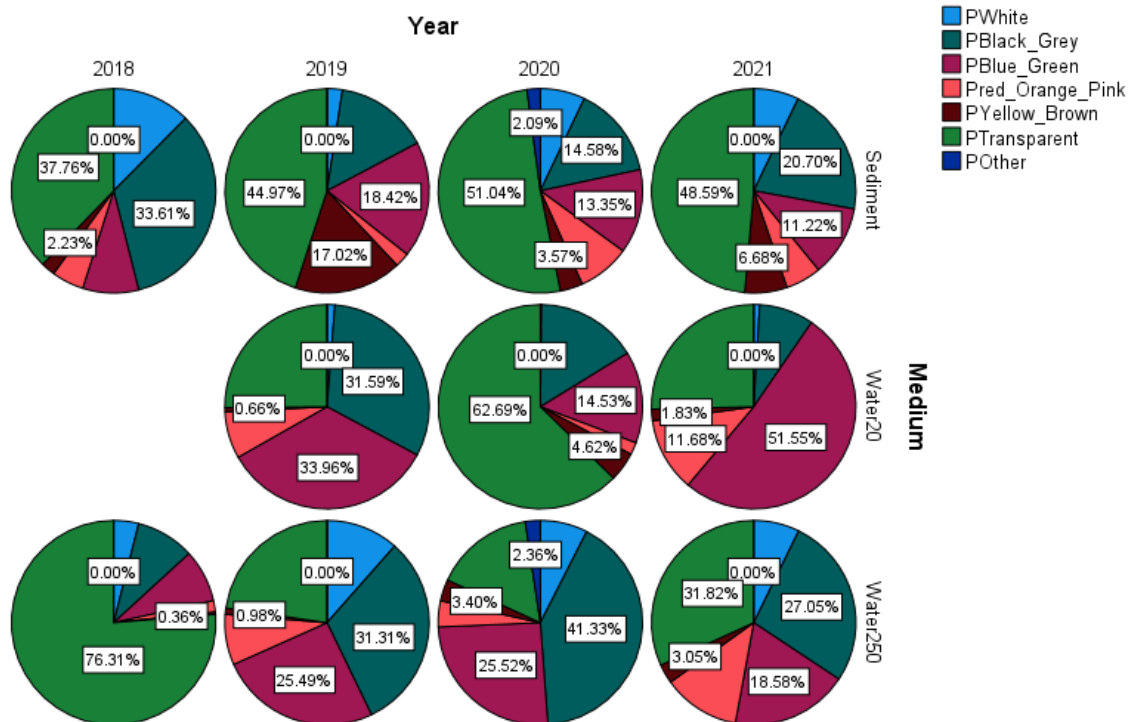


Figure 4.17: Pie charts ratio based on colour, medium and year

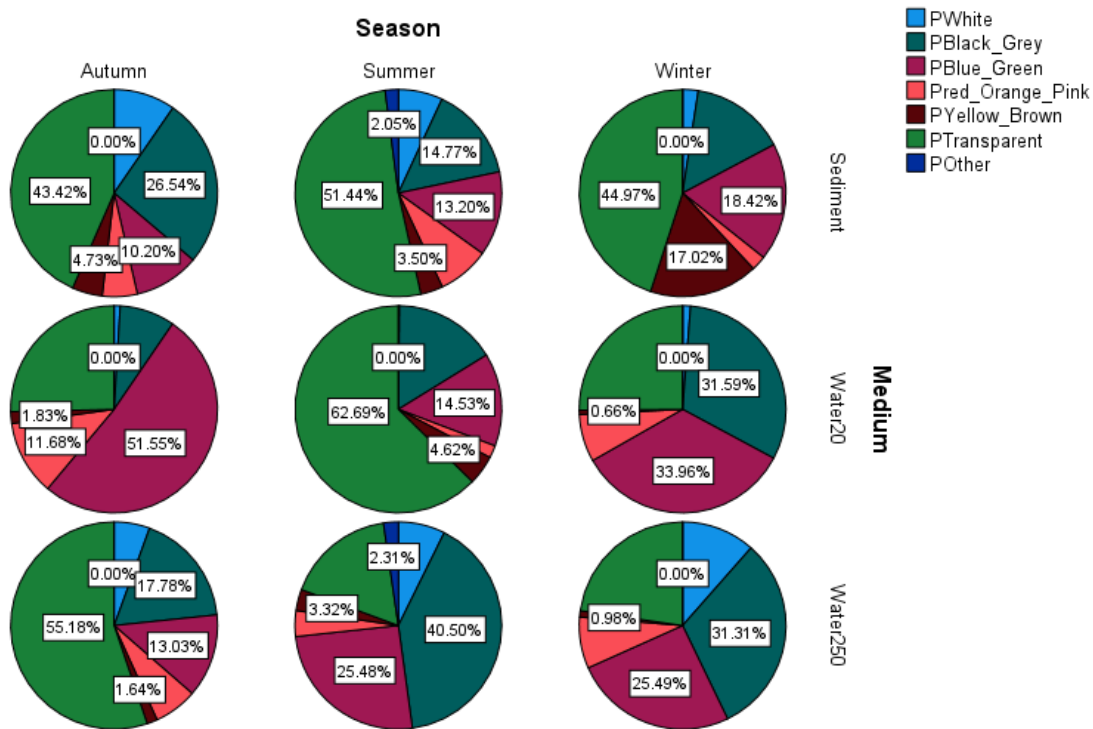


Figure 4.18: Pie charts ratio based on colour, medium and season

4.3.3 Microplastic abundances based on size

MPs were generally smaller than 1000 μm when analyzed based on different media (Figure 4.19), region (Figure 4.20), reaches (Figure 4.21), year (Figure 4.22) and seasons (Figure 4.23). The vlei and lower reaches of the study deviated from this trend, where MPs were larger than 5000 μm , during 2021.

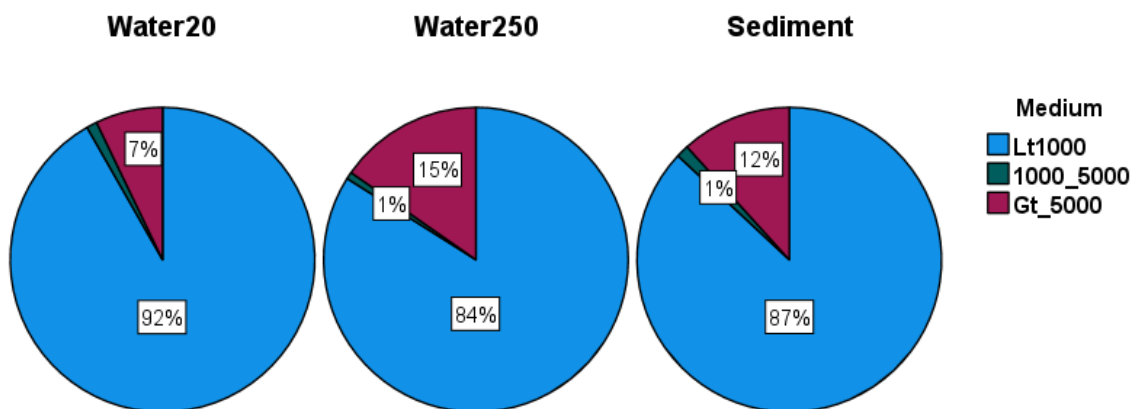


Figure 4.19: Pie charts ratio based on size and medium

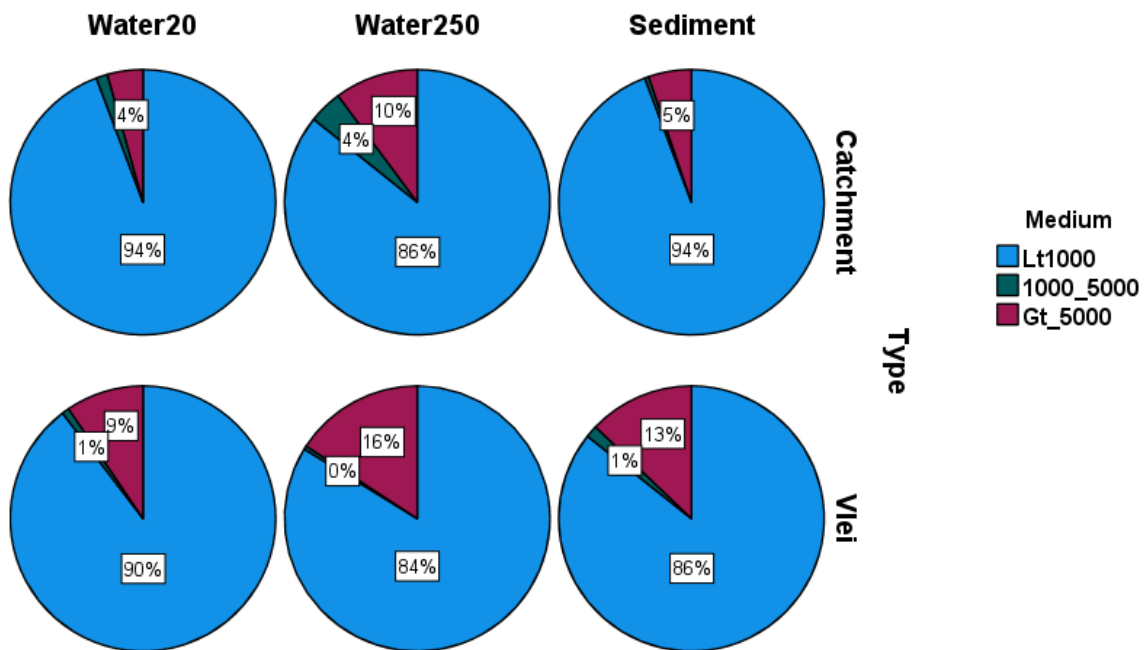


Figure 4.20: Pie charts ratio based on size, medium and region

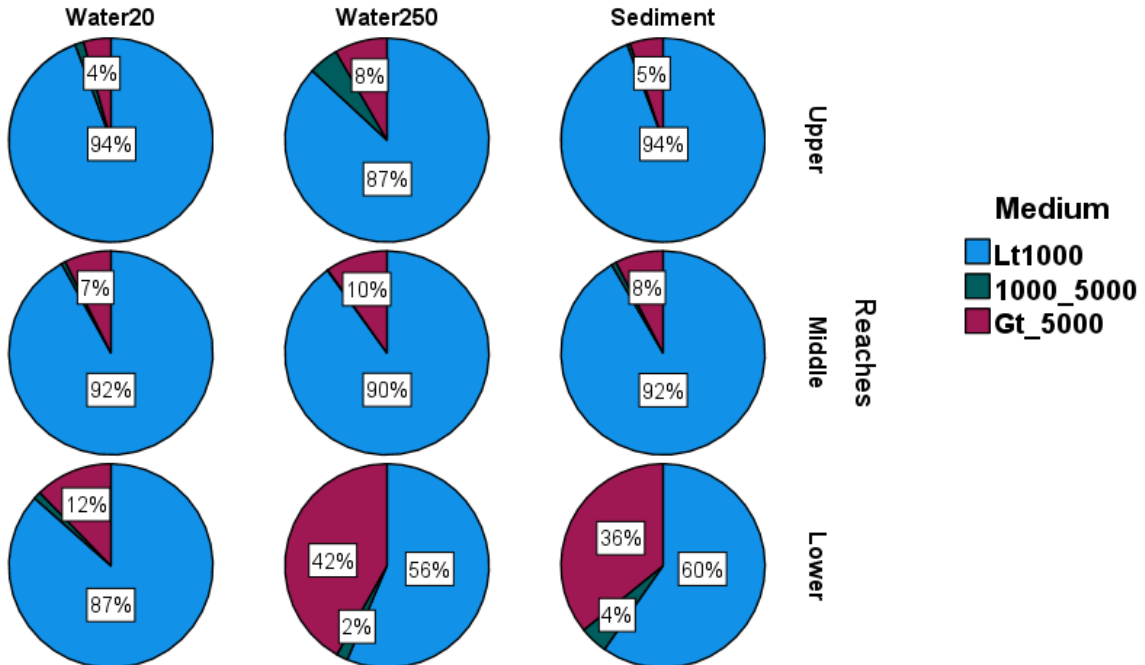


Figure 4.21: Pie charts ratio based on size, medium and reaches

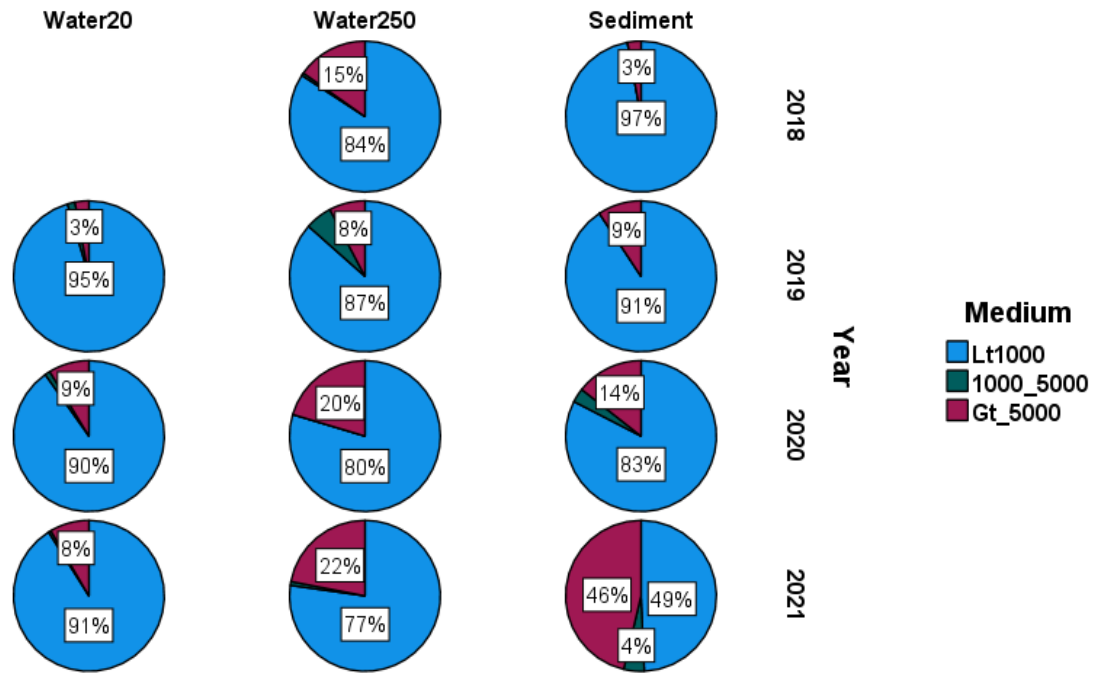


Figure 4.22: Pie charts ratio based on size, medium and year

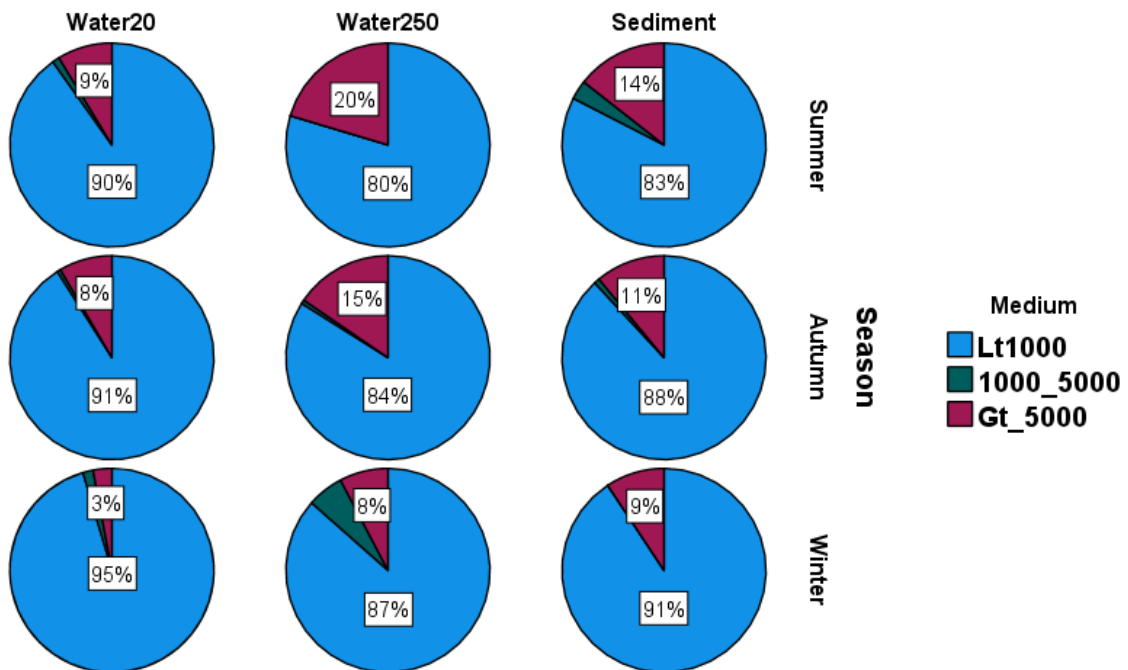


Figure 4.23: Pie charts ratio based on size, medium and season

4.4 Polymer characteristics

A wide array of polymers were recorded in the study (Figure 4.24). Polyethylene was the most

predominant synthetic polymer recorded in the study, as evident for the catchment and vlei (Figure 4.25a), mediums (Figure 4.25b) and seasons (Figure 4.26). Deviations of the trend was from Catchment Rivers, Prinseskasteel River (50% polyester and 50% styrene), Spaanschemat River (100% cotton), Westake River (50% polypropylene) and Zandvlei Yacht Club (50% Polypropylene). Polypropylene was dominant across all morphotypes except foam (57. 15%) (Figure 4.27a). Most of the synthetic polymers were either transparent or white (Figure 4.27b) with no distinct pattern based on polymer type and size of MPs (Figure 4.27c). Figure 4.28 indicates the spectral scans of representative MPs recorded in the study.

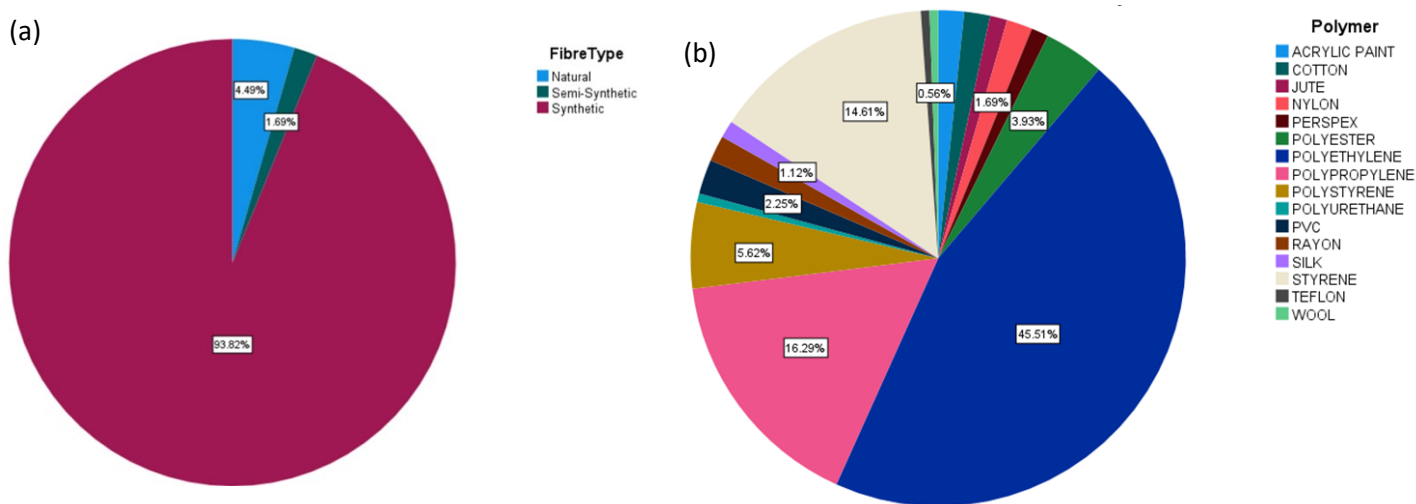
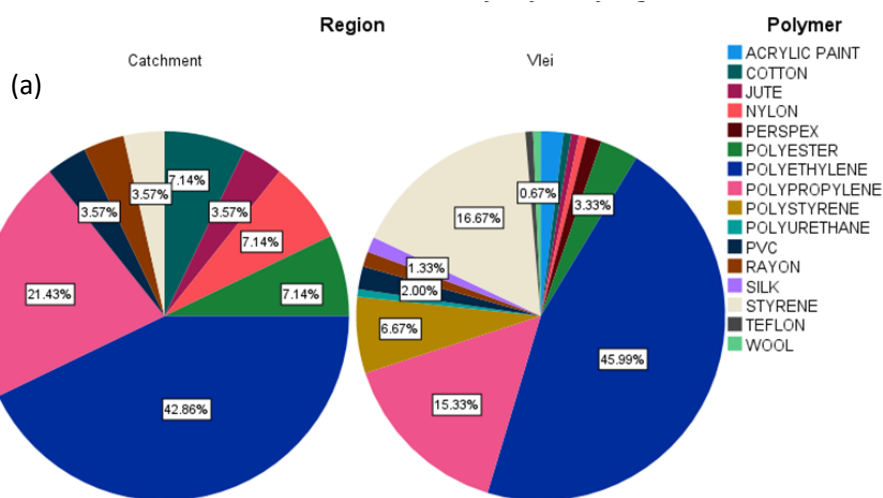


Figure 4.24: Pie chart depicting ratio of (a) synthetic to natural fibres and (b) different polymers across study



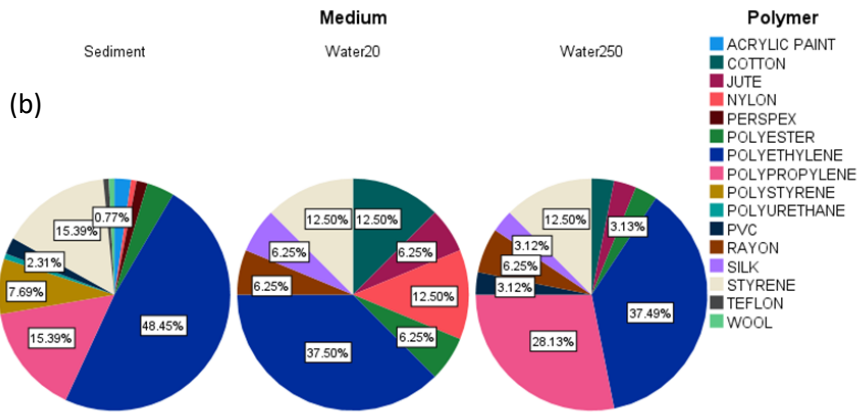


Figure 4.25: Pie chart depicting ratio of (a) different polymers between region (b) different polymers between mediums

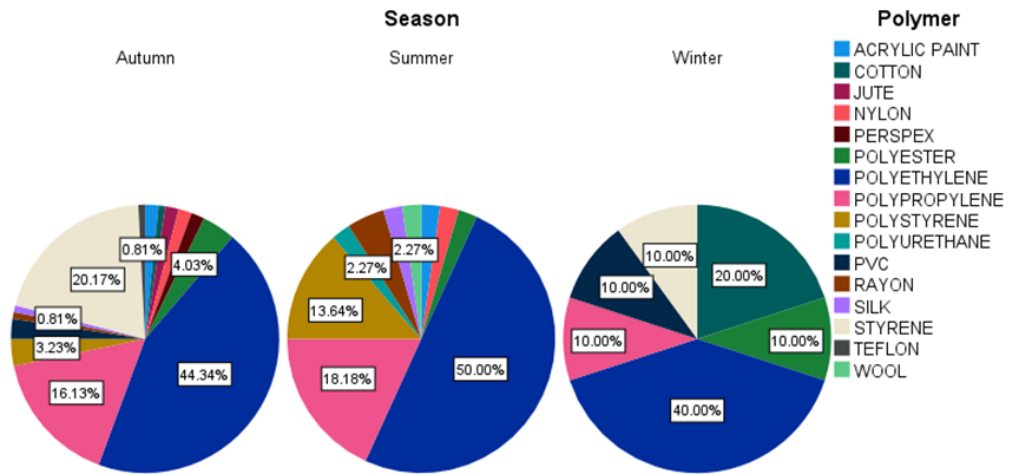


Figure 4.26: Pie chart depicting ratio of polymers across seasons

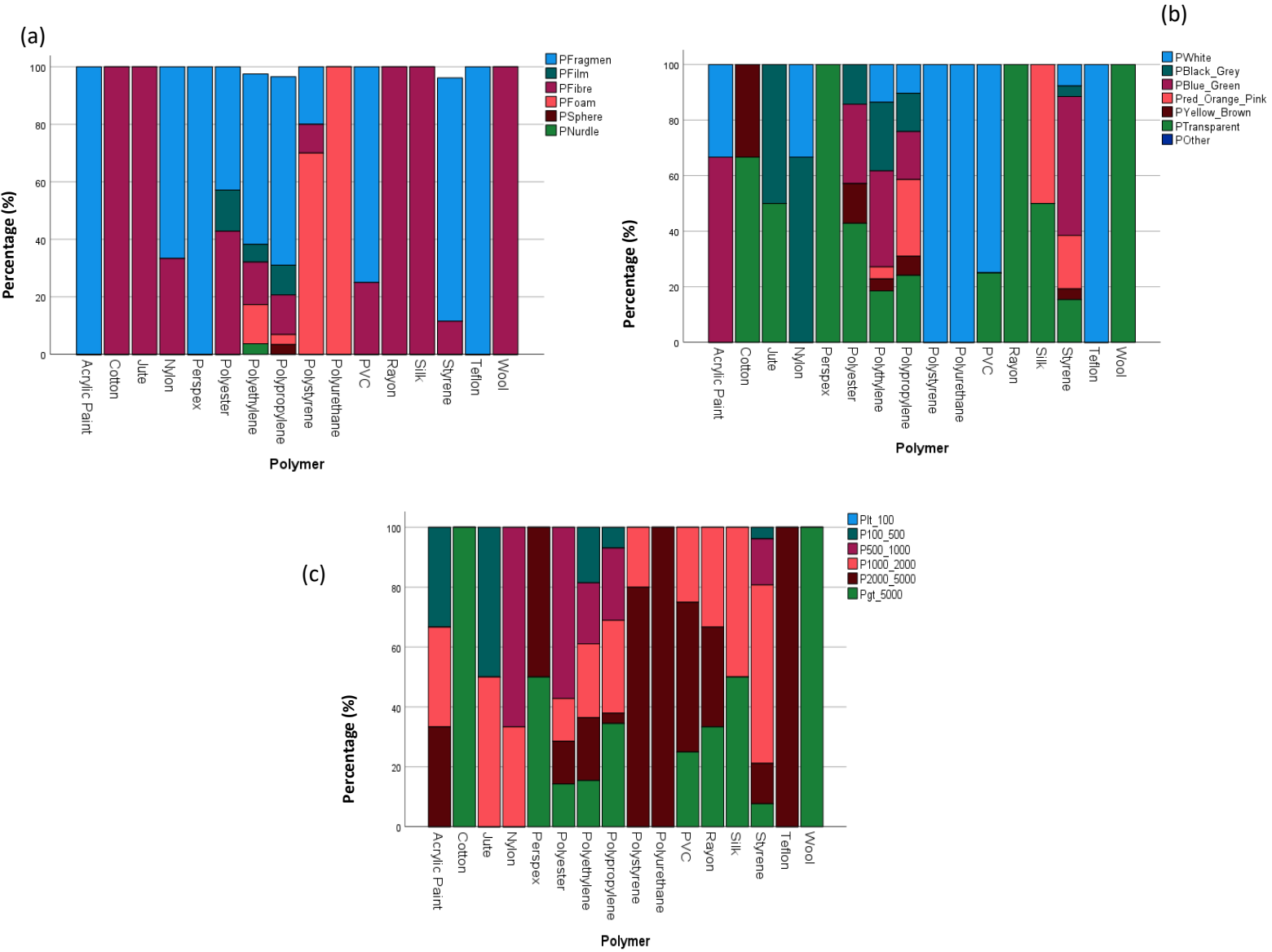


Figure 4.27: Polymer percentage (%) composition based on morphotype (a) Polymer percentage composition (%) based on colour (b) Polymer percentage composition (%) based on size (c)

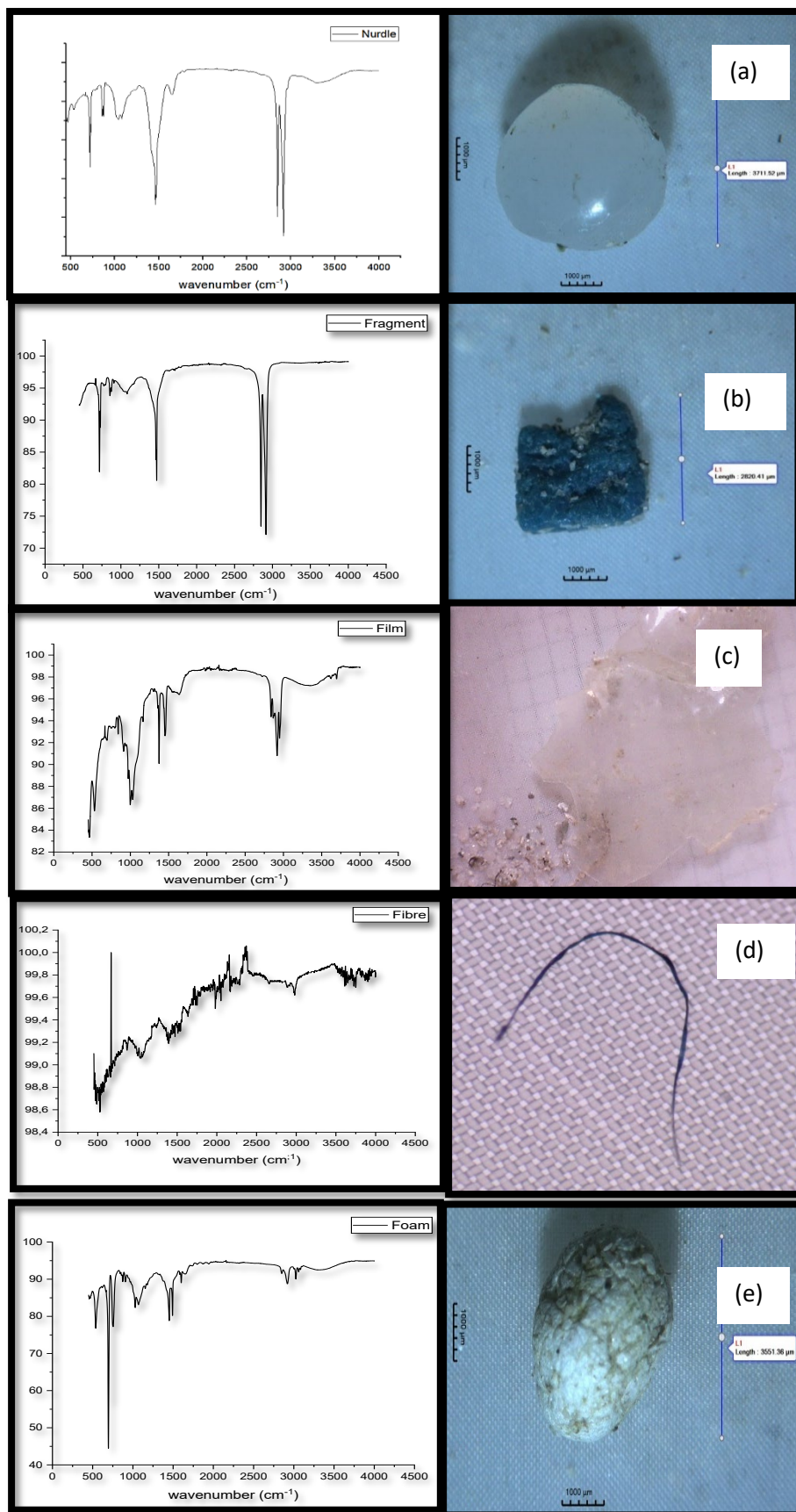


Figure 4.28: Infrared spectra of representative microplastic polymer samples in Zandvlei: (a) Polyethylene (b) Polyethylene Wax: High Density (c) Polypropylene (d) Polystyrene (e) Polyethylene: Olefin fibre

4.5 Grain size analysis

Grain size across the study was uniform with the grain size for sediment in catchment, vlei, upper middle and lower reaches consisting mainly of sediment in the 500 μm size class (Figure 4.29).

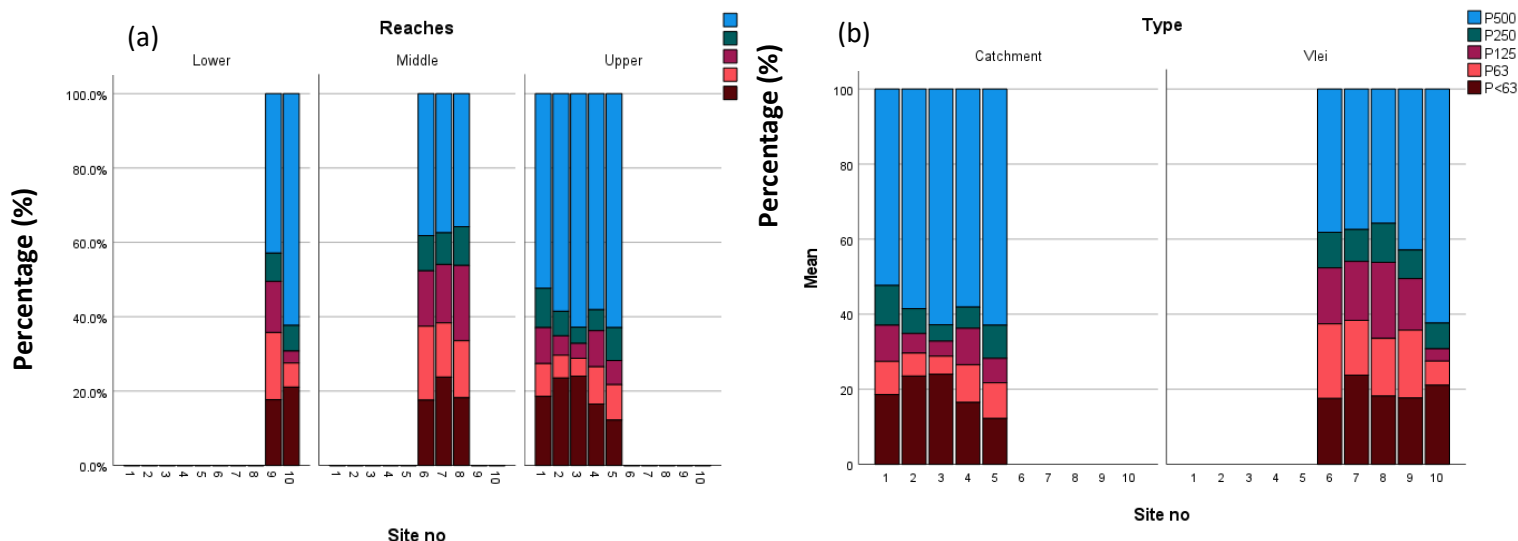


Figure 4.29: Grain size analysis percentage (%) based on reaches (a) Grain size analysis based on region (b)

4.6 Risk Assessment

The risk assessment displayed polymer risk index values between moderate and dangerous across the catchment and vlei across all mediums. Most risks were categorized as very high / dangerous for MPs in water 20 samples from the catchment. Moderate to low risk assessment scores were recorded for water 250 and sediment samples (Table 4.4).

Table 4.4: Risk assessment for media sampled in the catchment and vlei. Categories are colour-coded as low (green), moderate (blue), high (orange) and very high/dangerous (red)

Medium	Risk Assessment	Catchment	Vlei
Water20	MCFI	93.46 (V) dangerous	73.63 (V) dangerous
Water20	PLI	1969.61 (V) dangerous	0.12 (I) Low
Water20	H	2798.61 (IV) very high	1877.93 (IV) very high
Water20	PRI	5512194 (V) dangerous	203.75 (II) moderate
Water250	MCFI	1.09 (II) moderate	22.80 (V) dangerous
Water250	PLI	0.03 (I) low	1.11 (II) moderate
Water250	H	507.66 (III) high	614 (III) high
Water250	PRI	1.69 (I) low	675.62 (IV) very high

Sediment	MCFI	34.58 (V) dangerous	94.47 (V) dangerous
Sediment	PLI	0.45 (I) low	0.15 (I) low
Sediment	H	2.35 (I) low	419.44 (II) moderate
Sediment	PRI	1.07 (I) low	66.38 (I) low

CHAPTER 5: DISCUSSION

As plastic debris can be carried to the marine environment via rivers, transitional systems such as estuaries play a key role in the transportation of these particles from the land to the sea; and due to the dynamic nature of these ecosystems, microplastics can potentially remain in these habitats for extended periods of time and be ingested by several species. Pollution of coastal areas with plastic debris is a well-known problem, globally and in South Africa and is due to widespread improper disposal practices.

The respective study recorded 18,157 plastic particles collectively across the estuary and vleis these findings were not similarly aligned as expected with abundances discovered in other similar urban estuaries globally. The study of the urban Yangtze Estuary, China recorded 4,137.3 particles (Zhao et al. 2014) and Urban Tropical Goiana Estuary 14,724 particles (Barletta et al. 2020).

5.1 Medium

Microplastic abundances (MPs/Kg dry weight) and (MPs/m²) were most prevalent in sediment, specifically in vleis sediment. The increase of microplastic abundances closer to the mouth of the estuary as opposed to further away is evidence that estuaries are pathways of MPs to the marine environment. Similar patterns displayed in the respective study was displayed in a local KwaZulu Natal study conducted by Naidoo et al. (2015) (Table 3-8) where higher MP abundances were reported at polluted sites closest to the estuary mouth as opposed to further away. Elevated microplastic abundances closest to the mouth may be due to the influence of the artificially controlled estuary mouth which is opened during periods of low rainfall and closed with a sandbank during periods of high rainfall (CoCT, 2010).

Furthermore, estuaries act as sediment sinks for plastic particles, through the process of sedimentation as high density, larger size classes and fragmented microplastics are more likely to get trapped in sediments (Van Cauwenberghe et al. 2015) the higher density microplastics accumulate in sediment due to gravitational settling, while low density microplastics accumulate in sediment due to biological factors like biofouling (Li et al. 2019).

Estuaries are eutrophic environments with a high diversity of fouling organisms which facilitates algal and invertebrate attachment to the microplastic increasing the density causing them to sink into the sediment (Moore et al. 2001).

5.2 Season variations and region

The hydrodynamics of Zandvlei Estuary play an important role in the deposition and location of microplastics as it is situated in False Bay where there are numerous potential sources of litter and MPs (Pfaff et al. 2019). The hydrodynamics facilitate the notion that microplastics are more abundant closest to the mouth as False Bay is governed by South Easterly winds during the summer months (Dufois and Roualt, 2012). False Bay possesses a clockwise circulation rotation which is driven by the south-easterly winds and causes north-westerly current flow into Zandvlei Estuary (Wainman et al. 1987).

Tides also play a key role in the abundance and facilitation of microplastic movement from the ocean into the estuary through tidal flow. When the vlei mouth is open, flooding tides will be a northward flow into Zandvlei Estuary and during ebb tides there will be southward flow away from the estuary (Coleman et al 2021).

MPs (MPs/Kg dry weight) were highest during the rainy seasons (autumn and winter). According to Clarke et al. (1996) seasonal variations influence wind movement and circulation in False Bay. During winter the north-westerly winds are strongest, which could be a driver for circulation of microplastics into Zandvlei Estuary as well as increased rainfall during this time period. Key evidence of this is the presence of nurdles at site 9, closest to the estuary mouth, which can be inferred were introduced by wind driven movement into the vlei. The increased rainfall also allows MPs from the catchment to be flushed through the catchment system into the vlei area.

MP abundances (MPs/Kg dry weight) were highest in the middle reaches. This was also reported by Lima et al. (2014) study. In light of this, the hydrographic components of False Bay and the estuary itself play an integral role in the mixing and stratification in the middle reaches of the vlei. During the summer stable hydrographic conditions are observed in the upper and middle reaches of the estuary and these areas are a transition region between fresh water and marine water, generating turbulence and creating stratification in the water column of the mid reaches of the estuary restricting the transport of microplastics to different areas of the system (Barletta et al. 2020).

5.3 Particle Size Variation and Morphology

Most of the MPs were smaller than 1000 μm . The abundance of fibres across the whole study superseded all other morphotypes this remained constant seasonally, between reaches, between catchment and vlei and between mediums. According to Dalu et al. (2021) the smaller class microplastics may be from the fragmentation and degradation of larger microplastics in the system. Zandvlei estuary is adjacent to residential areas with the presence of the

Prinseskasteel, Prinskasteel, Sand, Spaanschemat and Westlaker Rivers draining into the populated vlei area.

The high concentrations of fibres may be related to the close proximity of highly populated domestic areas where there is inadequate litter removals and stormwater drainage (Kandasamy and Murugesan, 2011). Residential areas such as Marina da Gama are adjacent to the vlei area with the abrasion of synthetic textiles during washing of clothes and use of cosmetics are released with domestic effluent (Rahman et al. 2018). Zandvlei is also an area well-known for tourism and recreational fishing, with commercial fishing taking place in False Bay. Illegal dumping, inadequate disposal of waste such as cigarettes and broken fishing gear i.e. wrapping bands and fish nets are primary sources of the high fibre abundances (Wang et al. 2018). These high abundances may also be driven by the atmospheric and hydrographic processes in False Bay that may result in strong winds bringing in fibres from outside the study area.

5.4 Polymer Type

Synthetic polyethylene followed by polypropylene polymers were the most prevalent polymer types uniformly across all seasons, mediums and within the catchment and vlei. The same trend followed for the main rivers i.e. Westlake River and Keysers River and the mouth which is point source areas that feeds into the vlei area. These areas are highly urbanized with houses and large retail and grocery stores.

Polyethylene is most prevalent because it is a very versatile, inexpensive and has a wide range of applications. Polyethylene is largely used in the manufacturing of single use plastics, plastic bags, food packaging, bottles, caps, toys and containers. These are the main types of plastic litter in South Africa (Ryan and Moloney, 1990). This polymer is also not easily degraded by external forces and is more likely to aggregate amongst the same polymer particles. High density polyethylene is used for fishing and agricultural nets and mulch. On account of the drainage of the respective key rivers and agricultural region of Zeekoevlei into the system as well as input from False Bay through the mouth, this accounts for the high polyethylene abundances within the system.

5.6 Risk Assessment for catchment and vlei

The risk assessment of polymers analyzed from 2018 to 2021 recorded in Zandvlei catchment and vlei possesses environmental risk. Based on the results generally across the catchment and vlei pollution loads were in the low (I) to moderate (II) category with the pollution loads not being at high risk to the system. Alternatively, the polymer risk index across the catchment and vlei were in the very high to dangerous categories because additives are used in these synthetic

polymers which causes them to be ranked in a higher hazard score allowing for the polymer risk index to be elevated. These polymers are known to degrade and become bioavailable to not only organisms but humans as well. According to Lithner et al. (2011) high polymer risk index can allow for the following adverse effects: respiratory irritation, skin irritation, may cause cancer and severe skin burns and eye damage.

CHAPTER 6:

CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

The aim of this study was to determine the characteristics and potential effects of microplastics in the Zandvlei catchment area in Cape Town, South Africa. This study was novel because it was the first study that provided a report on microplastic contamination in Zandvlei Estuary and Catchment. This study assessed microplastic concentrations, characteristics, and ecological risks of the Zandvlei estuary and catchment. The microplastic concentrations recorded were comparatively higher than two other urban estuaries studies in South Africa and provided the first account of ecological risk assessment of microplastics in the Zandvlei Estuary. Microplastics were mainly fibres, transparent and smaller than 1000 μm . The main polymer type were polyethylene fibres and fragments. The high-risk values reported provides evidence to suggest that microplastics in Zandvlei Catchment and Vlei have the potential to negatively affect estuarine ecosystems. Hence the results of the research undertaken provide a strong rationale for microplastics to become part of riverine and estuarine monitoring programmes in the future.

6.2 Challenges and shortcomings

The most difficult challenges associated with the respective study was the lack of uniform and standardized microplastic sampling, quantification, acid digestion and density separation methodologies available in literature.

The methods used in the respective study (Chapter 3) had their own limitations because NaCl was used to extract microplastics from the catchment and vlei sediment. This may underestimate microplastics which are denser than the NaCl solution. Even though the NaCl solution was prefiltered before density separation there was still a chance that contaminant microplastics from the NaCl could introduce microplastics to the sample. However, this method was used because it was more cost effective, was simple to replicate and could be used across different kinds of microplastic studies to accommodate the postgraduate microplastics working group.

Furthermore, only surface water was sampled in the respective study (Chapter 3) this may underestimate the abundance of microplastics with higher densities that are not buoyant enough to stay suspended in surface water.

6.3 Recommendations

We need to find practical solutions to address the increase in microplastic contamination whilst highlighting the need for further study in ecologically sensitive areas.

Despite the need to urgently conduct more microplastic studies on ecologically sensitive areas it is imperative that sampling procedures and especially quantification methodologies are kept standard across studies in order to do inter-study comparisons on local, global and long-term monitoring scales.

There needs to be increased and strengthened capacity building between research institutions, universities, and NGO's as well as shared funding opportunities on key priority research areas.

More emphasis needs to be placed on the importance of ecological risk assessments in ecologically sensitive areas as well as estuarine areas as this component of research and literature is lacking on a local and global scale.

Promote cheaper and more cost-effective alternatives for microplastic surveys. The respective study's methodology for sampling, acid digestion and density separation can be used as a guide for a cost-effective approach to sampling and quantifying microplastic abundances in freshwater and estuarine studies.

Lastly, the findings of this research could potentially be used to initiate and promote microplastic pollution in estuarine health surveys and potentially introduce legislation to include more stringent plastic waste management policies in South Africa.

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APPENDIX

Appendix A1: Literature review of subsurface water microplastic studies in South Africa

Subsurface Waters											
Location	Microplastic Densities	Medium	Collection Method	Extraction Type	Pre-filter and extra extraction	Mesh Size	Morphotypes	Colour	Types of polymers	Polymer identification of particles	Study
Atlantic Ocean South West, South Africa	Mean range: 1.15 ± 1.45; 0-8.5	Subsurface Oceanic Waters	Klaus Union Sealex Centrifugal Pump (Bochum, Germany)	Water filtered through Buchner funnel and a vacuum flask	N/A	Whatman: 47 mm	Fibres Fragments	Pink Blue Other Transparent	Polyester Blends- (Nylon and Kevlar) Polyamide Polypropylene Acrylic Polyvinyl chloride Polystyrene Polyurethane Polyethylene terephthalate	Fourier transform infrared (FT-IR) spectroscopy on a Bruker Vertex 70 Infrared Spectrometer The instrument was equipped with a potassium bromide (KBr) beamsplitter and an internal	(Daan et al. 2017)

										mercury cadmium telluride (MCT) detector	
Indian, Atlantic, Southern Ocean	Indian: 1.20.3–3.0 Atlantic: .00.1–2.2 Southern ocean: 1.70.4–3.5	Surface and subsurface Water from cruises ACE, Antarctic Circumnavigation Expedition; IIOE2, Second International Indian Ocean Expedition.	Underway	N/A	N/A	N/A	N/A	N/A	Acrylic Polyester Polypropylene Nylon aramid	FTIR	(Suari et al. 2020)
Atlantic Ocean, South Africa	Average: 1.15±1.45 particles m ⁻³ .	Sub-surface waters	Klaus Union Sealex Centrifugal Pump (Bochum, Germany)	Buchner funnel and a vacuum flask	N/A	Whatman: 47 mm, pore size: 1.2 µm	Fibres Fragments	Blue Transparent Pink Purple Brown Red Green Grey Black Yellow white	Polyester Polyamide Polypropylene Acrylic Polyvinyl chloride Polystyrene Polyurethane Polyethylene terephthalate	(FT-IR) spectroscopy on a Bruker Vertex 70 Infrared Spectrometer	(Daan et al. 2017)

Appendix A2: Literature review of microplastic studies on sediment in South Africa

Sediment											
Location	Microp lastic Densiti es	Medium	Collectio n Method	Extraction Type	Pre-filter and additio nal extracti on	Mesh Size	Morphotypes	Colour	Types of polymers	Polymer identifica tion of particles	Study
Western Cape, South Africa	Numbe r of micropl astic 250 mL ⁻¹ in sedime nt 21-40	Beach sediment	Sample depth of 1cm	Density separation: Sodium Chloride	Pre-filter Sodium Chloride Extracti on repeat ed thrice	N/A	Fibre	N/A	Polyamide	FTIR	(Browne et al. 2011)
Five urban estuaries of KwaZulu-Natal South Africa	Mean: 745.4 ± 129.7 (±S.D.) particle s per 500 mL	Surface Water Estuarine Sediment Beach Sediment	500 ml water x 5 replicat es 10 cm depth with a corer With 50 mm	Density Separatio n: 140 g L ⁻¹ hypersatur ated sodium chloride (NaCl) solution Twice	Extracti on repeat ed twice	20 µm filters	Pellets Fragments Filament twine, Scrubbers Fibre	N/A	Polystyrene	(FT-IR) Perkin Elmer Spectrum 100 Series FT-IR spectromet er	(Naidoo et al. 2015)

			internal diameter	Water filtered through vacuum pump							
			Zooplankton net ((model 2035 MK4) 300 µm								
South Eastern Coastline South Africa	1215 ± 276.7, 927.4 ± 114.5 and 858.5 ± 241.0 particle s·m ⁻³ recorded in the water column 2636 ± 612.2, 2411 ± 297.7 and 3308 ± 1449 particle s·m ⁻² within	Beach sediment Surf-zone water	Top 5 cm of beach sediment Bulk water sample 45 cm deep	Density Separation: saturated saline solution WP-2 type net (80 µm) to filter water	Extraction repeated 5 times	65 µm mesh	Fibres Fragments Polystyrene-Spheres	Blue Black Red Yellow Green	Spectral not available	Spectral not available	(Nel & Froneman, 2015)

	the beach sediment										
Durban harbour, South Africa	750 microplastics per unit found in the surface layer (2.5–5 cm) 400 microplastics per unit found in the deeper layer (20–22.5 cm)	Sediment	Gravity corer of 11 cm i.d. and 50 cm length	Density Separation: 500 mL of 5.3 M NaI solution (density: 1.6 g/cm ³), Digestion: 150 mL of 30% H ₂ O ₂	N/A	315 mm mesh size nylon sieve	Fibre Fragment Film	Blue Black Brown White Green Yellow Red Grey Pink Cream	Polyethylene Polyester-Alkyd Polyamide-Nylon Polypropylene Polycaprolactone Polystyrene Polyethylene terephthalate Polyestradiol phosphate	FTIR (NICOLET iS5, Thermofisher Scientific) with Attenuated Total Reflectance (ATR)	(Matsuguma et al. 2017)
South African coastline	Bulk water (10L): 413.3 ±		Top 5 cm of beach sediment	Density Separation: hyper-saturated	N/A	63-µm mesh	Fibres Fragments Nurdles	Blue Black Red	Spectral not available	Spectral not available	(Nel et al. 2017)

	77.53 and 1200 ± 133.2 particle s·m ⁻³ Sediment microplastic loads ranged from 86.67 ± 48.68 to 754.7 ± 393 particle s·m ⁻²		Bulk water: 10 L buckets	solution (NaCl, 100 g/l) Water: vacuum pump							
West Coast to East Coast South Africa	Average microfiber content : 80 ± 102 F/dm ³ in Feb/ March 2017	Beach sediment	10 cm×5 cm mini-quadrant depth of 5 cm	Density separation: 500 cm ³ of a saturated NaCl solution NaCl filtered	Extraction repeated three times	N/A	Fibres	Blue Black	Spectral not available	Spectral not available	(De Villiers, 2018)

	87 ± 84 F/dm ³ in May/June 2016										
	0 to 797 F/dm ³ in 2017										
	4 to 772 F/dm ³ in 2016										
Bloukrans River system, Eastern Cape province of South Africa	6.3±4.3 (n=21; ±standard deviation) and 160.1±139.5 particles kg ⁻¹ (n=23), respectively for the summer and winter seasons mean of 0.37 ±	River sediment 2 kg and 5 cm top layer Deposit feeders (<i>Chironomid</i> larvae)	Hand-held nylon net (500 µm)	Digestion : nitric acid (55%) Density separation: hyper-saturated saline solution (100 g L ⁻¹)	N/A	Deposit feeders (<i>Chironomid</i> larvae): 2 µm Millipore membrane filter Sediment: 63 µm mesh	Morphotypes not represented in data	N/A	Spectral not available	Spectral not available	(Nel et al. 2018)

	0.44 and 1.12 ± 1.19 particles mg ⁻¹ ww for the summer and winter, respectively										
Cape Town Breede River Mossel Bay Port Elizabeth East London Durban South Africa	Industrial pellets were most abundant (55.1% of all mesodebris), followed by rigid plastic fragments (33.7%)	Beach surface area, sediment	Upper 50 mm of sand by sieving through a 2 mm mesh sieve (square frame, 0.5 m x 0.5 m, and 100mm deep)	Density Separation: using 20 L bucket of seawater	N/A	N/A	Morphotypes not represented in data	N/A	Spectral not available	Spectral not available	(Ryan et al. 2018)
South African coast line	0 to 567 fibres/d m ³	River sediment	5 m above the	Density separation:	No filter	Extraction repeated 3 times	Fibres	N/A	Spectral not available		(De Villiers, 2019)

			water's edge Samples were taken to a depth of 5 cm	saturated 1.2 g/cm ³ sodium chloride (NaCl) solution	paper/mesh					Spectral not available	
Braamfontein Spruit, Johannesburg South Africa	Water (mean of 705 particles m ⁻³) <i>Chironomus</i> sp. larvae (mean of 53.4 particles g ⁻¹ wet weight) Sediment (mean of 166.8 particles kg ⁻¹ dry weight)	Water <i>Chironomus</i> sp. larvae and sediment	100 L of water <i>Chironomus</i> sp. larvae were caught using a 1mm mesh size net 2 kg of top sediment in the river (≈ 10 cm depth)	Water Digestion: 10% KOH solution for the digestion <i>Chironomus</i> sp. larvae Digestion 1.9 mL of 10% KOH solution Density separation sediment: hypersaline NaCl solution (339 g l ⁻¹) until	N/A	N/A	Fibre Film Fragment Round Angular	Black Blue Green Other	Spectral not available	Spectral not available	(Dahms et al. 2020)

South Eastern Coast, South Africa	318 items 30g ⁻¹	Beach sediment	3kg sediment with stainless steel spatula	Digestion: 30 ml of 30% H ₂ O ₂ Density separation: Zinc Chloride (ZnCl ₂) ~1.5 g/cm ³	N/A	Merck Millipore, 25 mm diameter, diameter, 1.2 µm pore size)	Fibres Films Filaments Sheets Strands Clustered-Filaments	Black Blue White Pink Brown Red Green	Polypropylene High Density Polyethylene Low Density Polyethylene Rayon Nylon Polyester Polystyrene Polyethylene Terephthalate Polyacrylonitrile Polycarbonate	Scanning Electron Microscope (SEM) (IRAffinity-1 Shimadzu, Japan) FTIR	(Vetrimurugan et al. 2020)
Harbour port of Durban (Study A)	Sediment Total: 694 Mean: 99.14 ± 36.29 (±S.D) Min: 50 Max: 144.5 Water Total: 0.69 Mean: 0.099 ± 0.036 (±S.D) Min: 0.05	Surface Water Harbour Sediment	Particle pump (200 µm to >5 mm)	Density Separation: 1.2 g mL ⁻¹ solution of saturated sodium chloride (NaCl)	N/A	7 mm diameter Whatman GF/D filter with a 2.7 µm pore size	Morphotypes not represented in data	N/A	Water: Polypropylene (46%), followed by Polyethylene (38%) and Polyethylene terephthalate/ Polystyrene (8% each) Microplastic polymers: Polyethylene (47%), followed by Polyethylene terephthalate (16%) and Polystyrene (9%) Sediment: Polyethylene	Attenuated total reflection Fourier Transform infrared spectroscopy (ATR-FT-IR) with a Thermo Fisher Scientific Nicolet iS5 ATR-FTIR with OMNIC software (version 9.9.473)	(Preston-Whyte et al. 2021)

	Max: 0.145								(47%), followed by Cellophane (21%), Polypropylene (16%) and Polystyrene (5%) 11% of the particles were not successfully identified or were of biological origin		
Harbour port of Durban (Study B)	Sediment Total : 651.5 Mean: 93.07 ± 36.78 (±S.D) Min: 41.5 Max: 143.5 Water Total: 0.65 Mean: 0.093 ± 0.037 (±S.D) Min: 0.04	Surface Water Harbour Sediment	Particle Pump (200 µm - 5 mm)	Density Separation: 1.2 g mL ⁻¹ solution of saturated sodium chloride (NaCl)	N/A	7 mm diameter Whatman GF/D filter with a 2.7 µm pore size.	Morphotypes not represented in data	N/A	Water: Polypropylene (46%), followed by Polyethylene (38%) and Polyethylene terephthalate/ Polystyrene (8% each) Microplastic Polymers: Polyethylene (47%), followed by Polyethylene terephthalate (16%) and Polystyrene (9%) Sediment: Polyethylene	Attenuated total reflection Fourier Transform infrared spectroscopy (ATR-FT-IR) with a Thermo Fisher Scientific Nicolet iS5 ATR-FTIR with OMNIC software (version 9.9.473)	(Preston-whyte et al. 2021)

	Max:0.144								(47%), followed by Cellophane (21%), Polypropylene (16%) and Polystyrene (5%) 11% of the particles were not successfully identified or were of biological origin		
South African coastline	Bulk water (10L): 413.3 ± 77.53 and 1200 ± 133.2 particle s·m ⁻³ Sediment microplastic loads ranged from 86.67 ± 48.68 to 754.7 ±	Surface water Sediment	Top 5 cm of beach sediment Bulk water: 10 L buckets	Density Separation: hyper-saturated solution (NaCl, 100 g/l) Water: vacuum pump	N/A	63-µm mesh	Fibres Fragments Nurdles	Blue, Black Red	Spectral not available	Spectral not available	(Nel et al. 2017)

393	particle										
	$s \cdot m^{-2}$										

Appendix A3: Literature review on microplastic research in South Africa

Sediment											
Location	Microplastic Densities	Medium	Collection Method	Extraction Type	Prefilter and extra extraction	Mesh Size	Morphotypes	Colour	Types of polymers	Polymer identification of particles	Study
Review	Review	Review	Review	Review	Review	Review	Review	Review	Review	Review	(Verster and Bouwman, 2017)
Indian Ocean, South Africa	Review	Review	Review	Review	Review	Review	Review	Review	Review	Review	(Pattiaratchi et al. 2022)
Review	Review	Review	Review	Review	Review	Review	Review	Review	Review	Review	(Pereao et al., 2020)
Review	Review	Review	Review	Review	Review	Review	Review	Review	Review	Review	(Naidoo et al. 2020)
Review	Review	Review	Review	Review	Review	Review	Review	Review	Review	Review	(Verster and Bouwman, 2020A)

Review	Review	Review	Review	Review	Review	Review	Review	Review	Review	Review	(Nel et al. 2021)
Review	Review	Review	Review	Review	Review	Review	Review	Review	Review	Review	(Ryan et al. 2020)
Review	Review	Review	Review	Review	Review	Review	Review	Review	Review	Review	(Alimi et al. 2021)
Review	Review	Review	Review	Review	Review	Review	Review	Review	Review	Review	(Verster and Bouwman , 2020B)
Biofouling on Microplastics, South Africa	Experimental study on microplastic buoyancy	Review	Review	Review	Review	Review	Review	Review	Review	Review	(Fazey & Ryan, 2016)
Review	Sustainable policy framework for managing microplastic waste in Africa	Review	Review	Review	Review	Review	Review	Review	Review	Review	(Deme et al. 2022)
Review	Treatment options for marine plastic waste in	Review	Review	Review	Review	Review	Review	Review	Review	Review	(Williams-Wynn & Naidoo, 2020)

	South Africa											
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Appendix A4: Literature review on microplastics in organisms in South Africa

Organisms											
Location	Microplastic Densities	Medium	Collection Method	Extraction Type	Pre-filter and extra extraction	Mesh Size	Morphotypes	Colour	Types of polymers	Polymer identification of particles	Study
Urban Harbour KwaZulu-Natal, South Africa	3.8 particles per fish (SD 4.7)	Estuarine mullet <i>Mugil cephalus</i> (<i>Mugilidae</i>)	Castnet	Dissecting microscope	N/A	N/A	Fragments Polystyrene-Spheres Films Monofilament-line Twine	White Clear Opaque Black Red	Spectral not available	Spectral not available	(Naidoo et al. 2016)
KwaZulu-Natal, South Africa	No significant differences in the physiology (ingestion, respiration, photosynthetic)	Corals <i>Anomastrea irregularis</i> <i>Pocillopora verrucosa</i>	Twelve colonies of each species were carefully removed using a chisel and a hammer from intertidal rock pools	4% Formalin	N/A	100-µm nylon mesh filter 47 mm diameter reinforced glass	Fragments	N/A	Polypropylene	Fourier transform infrared spectrometer with an attenuated total reflectance	(Boodraj & Glassom, 2022)

	and growth rates) and tissue composition (<i>Symbiodiniaceae</i> , chlorophyll a and lipid contents) were found between corals fed the mix and those fed zooplankton only ($p>0.05$).					fbre filters with 5 μm pore size (GIC Scientific ©, South Africa)				attachme nt (Perkin Elmer Spectrum 100 Series Attenuate d Total Refectanc e spectrom eter)	
Bloukrans River system, Eastern Cape province of South Africa	6.3 \pm 4.3 (n=21; \pm standard deviation) and 160.1 \pm 139.5 particles kg^{-1} (n=23), respectively for the summer and winter seasons	River sediment 2kg and 5cm top layer Deposit feeders (<i>Chironomid</i> larvae)	Hand-held nylon net (500 μm)	Digestion: nitric acid (55%) Density separation: hyper-saturated saline solution (100 g L^{-1})	N/A	Deposit feeders (<i>Chironomid</i> larvae): 2 μm Millipore membrane filter Sediment: 63 μm mesh	Morphotypes not represented in data	N/A	Spectral not available	Spectral not available	(Nel et al. 2018)

mean of 0.37 ± 0.44 and 1.12 ± 1.19 particles mg ⁻¹ ww for the summer and winter, respectivel y											
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Appendix A5: Literature review on microplastics in store bought items in South Africa

Store Bought Items											
Location	Microplastic Densities	Medium	Collection Method	Extraction Type	Pre-filter and additional extraction	Mesh Size	Morphotypes	Colour	Types of polymers	Polymer identification of particles	Study
South Africa	maximum 37 particles per individual per annum	One package of sea salt (200–400 g)	Store bought	KOH digestion	Filtered twice	pore size (149 μ m)	Fragment Film	Turquoise Blue Yellow Green	Polyethylene terephthalate Polystyrene	Raman spectroscopy	(Karami et al. 2017)
South Africa	Average of: 0.04 MPs/g soft tissue and 3.8	Mussels (supermarket and wholesaler)	Store bought	10% KOH (w:v) acid solution to digest tissue	NaCl pre-filtered	20 μ m nylon mesh using	Filaments Fragments	White Transparent Yellow Red Pink	Cellulose Acetate Cotton Ethylene-vinyl Acetate	Perkin Elmer Two FTIR-ATR Spectrometer	(Sparks et al. 2021)

	MPs/mussel			Digestates filtered through a vacuum pump (model Rotary Vane VP 145 1/3 HP)				Blue Green Black Grey	High density polyethylene Latex Nylon Polyester-Cotton PVC Polyethylene terephthalate		
South Africa	(0–1.33 ± 0.32 particles/kg)	table salts	Store bought	Hydrogen peroxide (H ₂ O ₂)	N/A	Millipore cellulose nitrate membrane filter (having 0.3 µm pore size and 47 mm diameter)	Microfibres Fragments	N/A	Polyethylene Polypropylene Polyethylene terephthalate Polyethylene isophthalate Polyvinyl alcohol	FTIR	Fadare et al. 2021

Appendix A6: Literature review on microplastics in surface water in South Africa

Surface water											
Location	Microplastic Densities	Medium	Collection Method	Extraction Type	Pre-filter and extra extraction	Mesh Size	Morphotypes	Colour	Types of polymers	Polymer identification of particles	Study
South Eastern Coastline South Africa	1215 ± 276.7, 927.4 ± 114.5 and 858.5 ± 241.0 particles·m ⁻³ recorded in the water column 2636 ± 612.2, 2411 ± 297.7 and 3308 ± 1449 particles·m ⁻² within the beach sediment	Beach sediment Surf-zone water	Top 5 cm of beach sediment Bulk water sample 45 cm deep	Density Separation: saturated saline solution WP-2 type net (80 µm) to filter water	Extraction repeated 5 times	65 µm mesh	Fibres Fragments Polystyrene-Spheres	Blue Black Red Yellow Green	Spectral not available	Spectral not available	(Nel & Froneman, 2015)

Five urban estuaries of KwaZulu-Natal South Africa	Mean: 745.4 ± 129.7 (±S.D.) particles per 500 mL.	Surface water Estuarine Sediment Beach Sediment	500 ml water x 5 replicates 10 cm depth with a corer With 50 mm internal diameter Zooplankton net ((model 2035 MK4) 300 µm	Density Separation: 140 g L ⁻¹ hypertaturated sodium chloride solution Twice Water filtered through vacuum pump	Extraction repeated twice	20 µm filters	Pellets Fragments Filament twine, Scrubbers Fibre	N/A	Polystyrene	(FT-IR) Perkin Elmer Spectrum 100 Series FT-IR spectrometer	Naidoo et al (2015)
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Indian Ocean South East South Africa	N/A	Surface water	Modelling	N/A	N/A	N/A	Nurdles	N/A	N/A	N/A	(Schumann et al. 2019)
Orange Vaal river system South Africa	Bulk water: 0.21 ± 0.27 items·L ⁻¹ Net: 0.04 ± 0.16 items·m ⁻²	Top 30 cm of the water column	Neuston net 300 µm Bulk water samples (10L)	Density Separation for sediment removal from water samples: NaCl (prefiltered) Water samples filtered through	N/A	25 µm filter	Balls Beads Fibres Films Fragments Pellets Polystyrene-Spheres String Twine	N/A	Spectral not available	Spectral not available	(Weideman et al. 2020)

				h vacuum pump							
South African coastline	Bulk water (10L): 413.3 ± 77.53 and 1200 ± 133.2 particles·m ⁻³ Sediment microplastic loads ranged from 86.67 ± 48.68 to 754.7 ± 393 particles·m ⁻²	Surface water	Top 5 cm of beach sediment Bulk water: 10 L buckets	Density Separation: hyper-saturated solution (NaCl, 100 g/l) Water: vacuum pump	N/A	63-µm mesh	Fibres Fragments Nurdles	Blue Black Red	Spectral not available	Spectral not available	(Nel et al. 2017)
Agulhas Current, South Africa	of microplastic particles that enter the ocean from the five major coastal	Seasurface water	Modelling	Modelling	Modelling	Modelling	Modelling	Grey Teal	Low- and high-density Polyethylene Polyethylene Terephthalate	Modelling	(Collins & Hermes, 2019)

	urban-industrialized centers beach along the coastline of South Africa, a third is exported to the open ocean								Polyvinyl Chloride		
Harbour port of Durban	<p>Sediment Total: 694 Mean: 99.14 ± 36.29 (±S.D) Min: 50 Max: 144.5</p> <p>Water Total: 0.69 Mean: 0.099 ± 0.036 (±S.D) Min: 0.05 Max: 0.145</p>	Surface Water Harbour Sediment	Particle pump (200 µm to >5 mm)	Density Separation: 1.2 g mL ⁻¹ solution of saturated sodium chloride (NaCl)	N/A	7 mm diameter Whatman GF/D filter with a 2.7 µm pore size	Morphotypes not represented in data	N/A	<p>Water: Polypropylene (46%), followed by Polyethylene (38%) and Polyethylene terephthalate/Polystyrene (8% each)</p> <p>Microplastic polymers:</p>	Attenuated total reflection Fourier Transform infrared spectroscopy (ATR-FT-IR) with a Thermo Fisher Scientific Nicolet iS5 ATR-FTIR with OMNIC software (version 9.9.473) and	(Preston-Whyte et al. 2021)

									<p>Polyethylene (47%), followed by Polyethylene terephthalate (16%) and Polystyrene (9%)</p> <p>Sediment: Polyethylene (47%), followed by Cellophane (21%), Polypropylene (16%) and Polystyrene (5%)</p>		
Harbour port of Durban (Study B)	<p>Sediment Total : 651.5</p> <p>Mean: 93.07 ± 36.78 (±S.D)</p> <p>Min: 41.5</p>	Surface Water Harbour Sediment	Particle Pump (200 µm - 5 mm)	Density Separation: 1.2 g mL ⁻¹ solution of saturated	N/A	7 mm diameter Whatman GF/D filter with a 2.7 µm pore size.	Morphotypes not represented in data	N/A	<p>Water: Polypropylene (46%), followed by Polyethylene (38%) and</p>	Attenuated total reflection Fourier Transform infrared spectroscopy (ATR-FT-IR) with a	(Preston-whyte et al. 2021)

	<p>Max: 143.5</p> <p>Water Total: 0.65</p> <p>Mean: 0.093 ± 0.037 (±S.D)</p> <p>Min: 0.04</p> <p>Max:0.144</p>			sodium chloride (NaCl)					<p>Polyethylene terephthalate/ Polystyrene (8% each)</p> <p>Microplastic Polymers: Polyethylene (47%), followed by Polyethylene terephthalate (16%) and Polystyrene (9%)</p> <p>Sediment: Polyethylene (47%), followed by Cellophane (21%), Polypropylene (16%) and</p>	<p>Thermo Fisher Scientific Nicolet iS5 ATR-FTIR with OMNIC software (version 9.9.473)</p>	
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									Polystyrene (5%)			11% of the particles were not successfully identified or were of biological origin
Cape Province South Africa	3640 p. km ⁻²	Surface Coastal waters	Neuston trawls	N/A	N/A	N/A	N/A	N/A	Spectral not available	Spectral not available	(Ryan et al., 1988)	
Kwazulu-Natal Coastal shelf South Africa	Average: 4.01 ± 3.28 plastic particles/ 100 m ²	Surface Coastal waters	Stainless steel manta trawl (333 µm mesh)	Sieving 1000, 500 and 250 µm stacked sieves	N/A	N/A	Fragments Films Fibre Line Pellet Polystyrene sphere	White Clear Opaque Blue Black, Green Grey Red Yellow Pink	Spectral not available	Spectral not available	(Naidoo et al., 2019)	
Indian, Atlantic,	Indian: 1.20.3–3.0 Atlantic: .00.1–2.2 Southern ocean:	Surface and subsurface Water from cruises ACE, Antarctic Circumnavigation Expedition; IIOE2, Second	Underway	N/A	N/A	N/A	N/A	N/A	Acrylic Polyester Polypropylene Nylon aramid	FTIR	(Suaria et al. 2020)	

Southern Ocean	1.70.4–3.5	International Indian Ocean Expedition.									
Atlantic Ocean, Cape Town, South Africa	29 pyrolyzate compounds of marine water samples Out of 16 identified polymers in the study area, polythene (PE) was the dominant in six out of seven locations with 87.5% followed by polyethylene terephthalate (PET) and polyvinylc	Surface Coastal Waters	300 µm neuston net	Digestion: 20 mL of 30% hydrogen peroxide oxidation in the presence of 0.05M iron (II) sulphate (20 mL) Density separation: 5M NaCl to isolate microplastics through flotation	N/A	N/A	N/A	N/A	Polythene (PE) Polyethylene terephthalate (PET) Polyvinyl chloride (PVC) Polystyrene (PS), Polyamide 12 (PA-12) Polyacrylic acid (PAA) Ethyl vinyl acetate (EVA) copolymer	Tungsten scanning electron microscopy Thermal studies (Discovery TGA 5500) Fourier Transform Infrared (FTIR) Spectrum 100 PerkinElmer Analysis of plastic micro particles using pyrolysis GC-TOF-MS The LECO pyrolysis Gas Chromatography	(Vilakati et al. 2020)

<p>chloride (PVC) in five (71.4%) and four (57.1%) out of seven locations respectively. The other constituent polymers of microplastics identified through pyrolyzates were polystyrene (PS), polyamide 12 (PA-12) polyacrylic acid (PAA) and ethyl vinyl acetate (EVA)</p>										
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