TALLINN UNIVERSITY OF TECHNOLOGY
SCHOOL OF ENGINEERING
Department of Civil Engineering and Architecture

ASSESSMENT OF MICROPLASTIC REMOVAL EFFICIENCY BY EXISTING WASTEWATER TREATMENT TECHNOLOGY IN ESTONIA: A CASE STUDY IN KEILA WWTP

MIKROPLASTI EEMALDAMISE TÕHUSUSE HINDAMINE OLEMASOLEVA REOVEEPUHASTUSTEHNOLOOGIA ABIL EESTIS- KEILA REOVEEPUHASTUSJAAMA JUHTUMIUURING

MASTER'S THESIS

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Tallinn 2024

AUTHOR'S DECLARATION

Hereby I declare that I have written this thesis independently.

No academic degree has been applied for based on this material. All works, major viewpoints, and data of the other authors used in this thesis have been referenced.

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THESIS TASK

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Thesis topic:

Assessment of microplastic removal efficiency by existing wastewater treatment technology in Estonia: A case study in Keila WWTP**.**

Thesis main objectives:

The objective of this study is to analyse liquid phase microplastics concentration, evaluate the removal efficiency of the subject WWTP, and propose process improvements.

Thesis tasks and schedule

No.	Task Description	Deadline
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2.	Research writing, data analysis and compilation	May 15, 2024
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List of Abbreviations and Terms

A2O: Anaerobic-Anoxic-Oxic

AS: After Screen

ASP: Activated Sludge Process

DAF: Dissolved Air Floatation

DF: Disc Filters

DW: Dry Weight

FT-IR: Fourier-Transform Infrared Spectroscopy

HDPE: High-density polyethylene

LW: Laundry Wash

MBR: Membrane Bioreactor

MPs: Microplastics

Mt: Million Tons

NOOA: National Oceanic and Atmospheric Administration

Nylon-6: Polycaprolactam

PA: Polyamide

PAN: Polyacrylonitrile

PB: Polybutene

PCPs: Personal Care Products

PDAP: Poly(diallyl phthalate)

PE: Polyethylene

PEST: Polyester

PET: Polyethylene Terephthalate

PP: Polypropylene

PS: Polystyrene

PTFE: Polytetrafluoroethylene

PUR: Polyurethane

PVC: Polyvinyl Chloride

Py-GC-MS: Pyrolysis-Gas Chromatography-Mass Spectrometry

QI: Quality Index

RE: Removal Efficiency

REACH: Registration, Evaluation, Authorisation and Restriction of Chemicals

RSF: Rapid Sand Filtration

SEM-EDX: Scanning electron microscope-energy-dispersive X-ray

SS: Suspended Solids

UWWTD 91/271/EEC: Urban Wastewater Treatment Directive

UV: Ultraviolet

WWTP: Wastewater Treatment Plant

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Table of Contents

List of Figures

List of Tables

Abstract

Wastewater treatment facilities are recognised as significant contributors of microplastics (MPs) to the environment. However, the specifics regarding the fate of MPs within these systems must be better understood. To explore this, a study was initiated at the wastewater treatment plant in Keila City, Harju, Estonia, focusing on microplastic distribution and removal in the raw influent, primary treatment (mechanical screening and grit/grease removal), and the effluent of the secondary treatment employing the Anaerobic-Anoxic-Oxic (A2O) process.

The MPs identified by visual inspection were fibres, fragments, sheets, film, and foam. The average concentration of MPs in the influent was 173.3 MPs/L, and the primary treatment removed approximately 75.4%. After the secondary treatment stage, the concentration of MPs in the final effluent significantly decreased to 1.31 MPs/L, demonstrating an overall removal efficiency of 99.2%. MP within the size range of 500 - 5000 μm were most prevalent in the influent and the effluent of the treatment plant.

The μ-FTIR analysis identified six polymer types: polyethylene terephthalate (PET), polypropylene (PP), polystyrene (PS), polyethylene (PE), polyvinyl chloride (PVC), and polyacrylonitrile (PAN), respectively. Fragments, sheets, foam, and films were composed of PVC, PP, and PE, while fibres were predominantly PET, PP, PS and PAN. The outcome of this study demonstrates that the Keila WWTP can significantly reduce MPs size greater than 300μm in raw wastewater. Despite the high removal efficiency exhibited by the treatment plant, a considerable number of MPs are released into the water body.

1. Introduction

The global marketplace for plastic and its associated products has seen significant growth since the 1950s, owing to low material cost, incredible versatility, and high quality. The use cases of plastic cuts across several sectors such as food packaging, construction, home and industry-grade appliances, medical instruments, and more. Global plastics production as of 2019 was about 460 million tons [1], 4 times less than steel production [1], and 33 times more than natural rubber production [2]. By 2060, global plastic production is projected to hit 1.2 billion tons under a business-as-usual scenario (Figure 1).

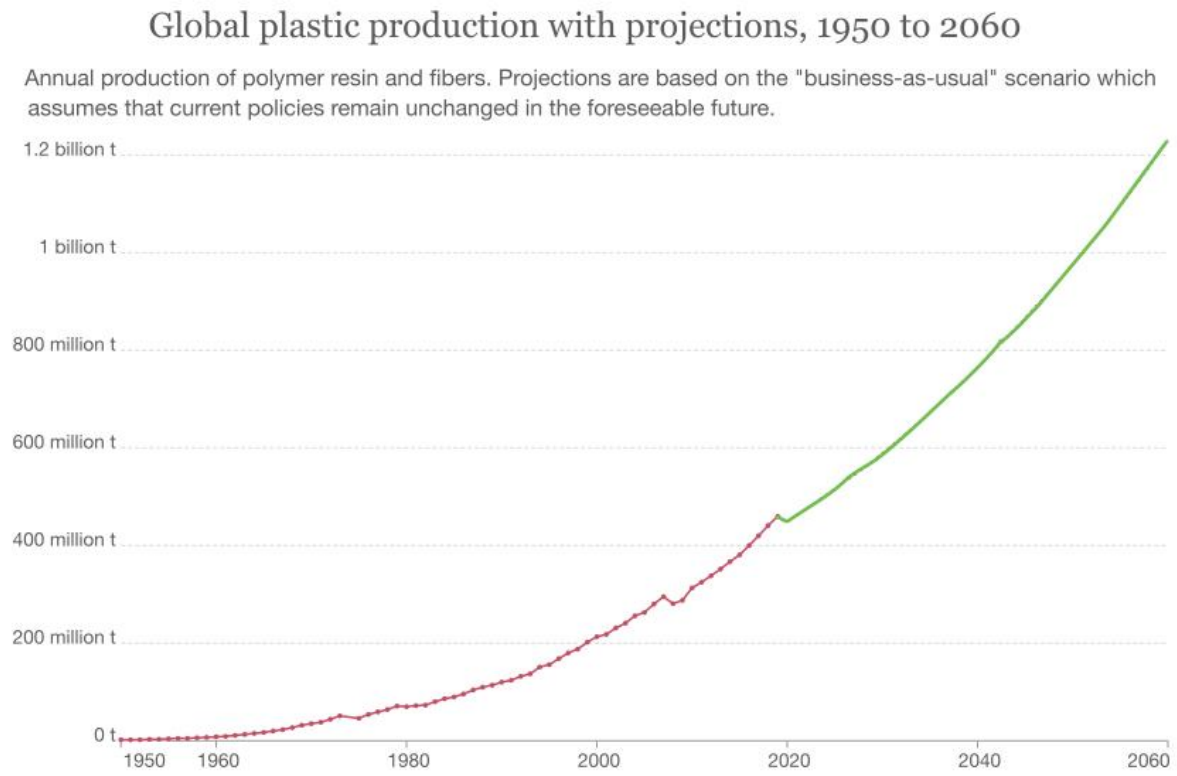


Figure 1. Global plastics production (1950 to 2019) and projection (2019 to 2060)[3]

The Organization for Economic Co-operation and Development (OECD) reports [4] that plastic waste generation increased to 353.3 million metric tons in 2019 (6.5 times more than global e-waste generation [5]), showing that waste generation is as much as the production metrics. About 9% of globally generated plastic waste goes to recycling, 50% is landfilled, 19% goes to incineration, and 22% evades collection systems, ending up in dumpsites [4]. In 2019, a total of 6.1 Mt of waste plastic entered aquatic habitats, with 1.7 Mt specifically entering the oceans

[6]. Recent estimates suggest that there are around 30 million metric tons of plastic garbage in seas and oceans, with an additional 109 million metric tons accumulating in rivers [7].
As plastic wastes persist in the environment, they undergo fragmentation due to the combined effects of photochemical and mechanical processes expedited by sunlight, wind, and waves, breaking down into smaller fragments (< 5mm) known as microplastics (MPs), now perceived as serious health and environmental risk, especially in marine ecosystems [8].

The study of microplastic pollution emanated in 1972, and since then, extensive research has revealed their presence in various compartments of the environment [9]. Microplastics have been found in oceans and rivers [10], [11], mangroves [12], and air [13], underscoring their widespread distribution. Apart from land and water-based plastic litter fragmenting to form MPs, wastewater treatment plants (WWTPs) have been identified as a significant sink and source of these micropollutants [14]. MPs reaching urban WWTPs through the sewage system originate mostly from the use of personal care products (PCPs) containing microbeads and laundry activities releasing synthetic fibre (ibid). The occurrence of this emerging micropollutant in WW (wastewater) means that there is the possibility that it may persist downstream of the treatment plants (rivers and oceans). Hence the emergence of several studies investigating the characteristics and removal efficiency of MPs in WWTPs [15], [16], [17].
In Estonia, there is limited research on microplastics in the WWTPs, therefore, addressing this research gap will enhance the comprehension of microplastic pollution's dynamics and the effectiveness of current wastewater treatment processes.

1.1 Goal of the study

The goal of this study is to:

1. analyse microplastics (MPs) concentrations in the liquid phase of the stages of the Keila WWTP.
2. assess the current stagewise and overall MPs removal efficiency by the existing wastewater treatment technology in Keila.
3. propose methods to increase the MPs removal efficiency in Keila WWTP

1.2 Justification of the Study

Estonia is predominantly sparsely populated, with a population of 1.35 million people served by numerous municipal wastewater treatment plants (WWTPs) (n = 664). Most of these WWTPs are small, each serving fewer than 300 population equivalents (PEs) and utilize similar

activated sludge process treatment technology. Keila WWTP, serving 2000 PEs, employs the same process technology as 80% of medium-sized WWTPs across Estonia. Recently, Keila WWTP has expressed interest in upgrading their treatment process units and is keen on studying how to address emerging contaminants such as microplastics (MPs).

1.2.1 Choice of Sampling Points

Raw wastewater entering the Keila WWTP goes through the mechanical screens and then the grit and grease removal chamber, both of which are the primary treatment stage. The effluent of the primary treatment flows into the secondary treatment phase comprising of a biological tank and a secondary clarifier. The choice of sampling at the influent, after screen and effluent is to ensure a comprehensive analysis of the efficiency of the primary and secondary treatment stages in reducing the concentration of microplastics in the liquid phase.

A clear understanding of the load and type of MPs entering into the Keila WWTP, the capture rate of the mechanical treatment (screening and grit/grease removal), and the efficiency of the biological treatment and clarifying process will provide us with the essential data to evaluate the impact of the treatment processes on MPs load reduction and adequate information to provide recommendations on technological upgrades or process optimization to improve removal efficiency.

2. Literature Review

2.1 Microplastics, Sources and Characteristics

According to the National Oceanic and Atmospheric Administration (NOOA), microplastics (MPs), a term first coined in a 2004 study of plastic pollution in UK waters [18], are defined as small synthetic plastic particles measuring ≤5mm in diameter [19]. By the nature of their original sizes, MPs are known to originate from primary and secondary sources [20]. Primary MPs emanate from engineered MPs (for example, microbeads in cosmetic products, medications, or detergents), and other processes where MPs are intentionally utilized as a component of commercial or industrial products [20], [21].

Secondary MPs result from the physical, chemical, or biological breakdown of synthetic fibres or larger polymers [22]. The process (Figure 2) begins with the initial degradation or fragmentation of macro-plastics (D >20 mm) facilitated by UV, heat, sea wave action, and abrasion to form meso-plastics (5 mm ≤ D ≤ 20 mm), and subsequently, microplastics (1 μm ≤ D ≤ 5 mm) and nanoplastics (NPs: 1 nm ≤ D ≤ 100 nm) [23].

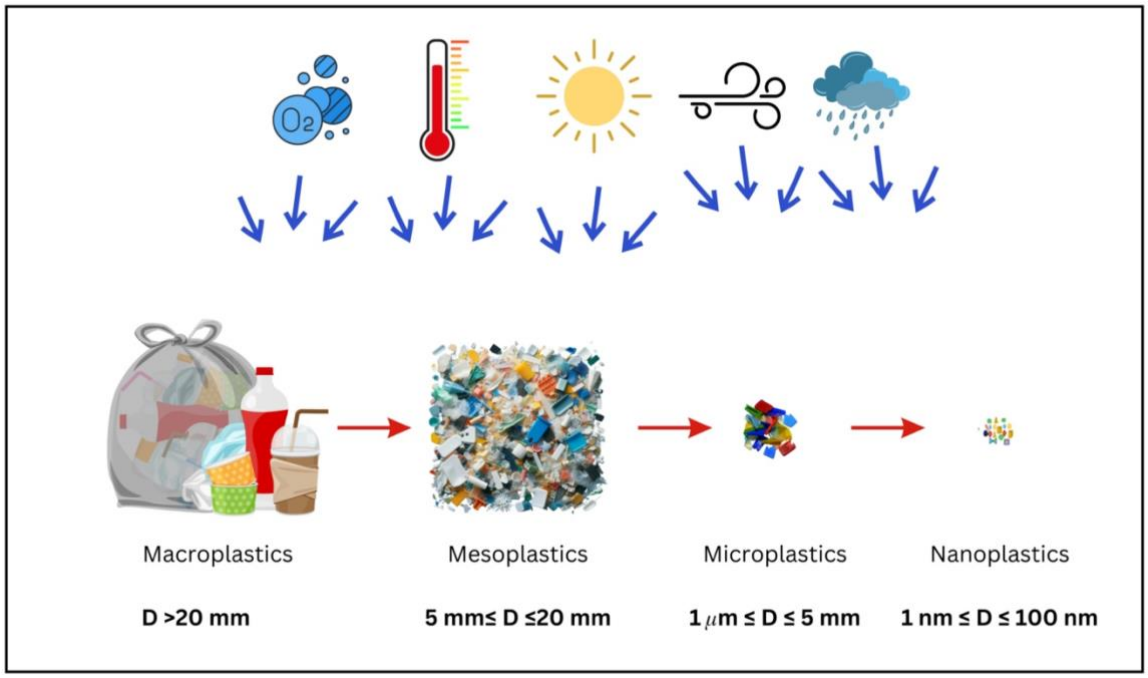


Figure 2 Fragmentation of plastic waste into smaller size fractions.

MPs are characterized by their small particle size, large surface area, and hydrophobicity [24]. While they are by themselves emerging contaminants, their surface area and hydrophobicity mean that they can adsorb other environmental pollutants, acting as micro-vectors for a cocktail of toxic substances and pathogens [25].

2.2 Microplastics Contamination in Different Environmental Compartments

The ubiquitous nature of MPs transcends ecological domains as their presence has been reported in not just the aquatic ecosystem, but also the terrestrial and atmospheric compartments of the environment (Figure 3) [26].

2.2.1 Microplastics in the Aquatic Environment

MPs may enter the aquatic environment from terrestrial sources such as stormwater overflow, leachates from landfills or runoff from polluted lands, tyre wear, WWTP effluents (containing MPs from PCPs or fibres from textile laundering), and atmospheric deposition [27]. In 2015, an estimation showed that there are between 15 to 51 trillion microplastic particles (about 93-236 thousand tons) floating in the world's oceans [28]. As of 2019, Eriksen et al. put this estimation at a mean of 171 trillion plastic particles (2.3 million tons) [29]. A Study that investigated the presence of MPs in various European waters, including smaller rivers and tributaries revealed a significant variation in microplastic concentrations, ranging from as low as 0.03 to as high as 187,000 particles per cubic meter [30]. In sediments, the concentration ranges from 18 to 72,400 particles per kilogram (ibid). Research focused on the Baltic Sea observed MP concentrations ranging from 0.07 - 3,300 particles per cubic meter. In sediments, the numbers vary from 0 - 10,179 particles per kilogram [31].

2.2.2 Microplastics in Soils and Sediments

MPs enter the soil via several routes including wastewater sludge recycling, landfill, and plastic mulching [32]. According to Nizzetto et al.[33], ca. 700,000 tonnes of MPs get into agricultural land in North America and Europe per year. Lofty et al. [34] suggested that European soils may represent the most significant repository of MPs. The United Kingdom, for instance, exhibits the highest level of contamination on agricultural land, with estimates ranging from 500 to 1000 particles per square meter annually. Spain, Portugal, and Germany are also significantly affected, in terms of microplastic accumulation. Adopting the data from the study and leveraging stats from the EC and Eurostat, lofty et al. calculated that the annual MP pollution in European soils could range from 31,000 to 42,000 tonnes for MPs sized between 1000–5000 μm, or between 8.6×10^{13} and 7.1×10^{14} particles for sizes 25–5000 μm (ibid).

2.2.3 MPs in the Atmosphere

Atmospheric deposition of MPs has been investigated by several studies, and the occurrence of these pollutants in the atmosphere indicates the potential for long-range airborne

transportation and human exposure [35]. Aside from disintegrated lightweight plastic litter from land sources ingressing the atmosphere due to wind action [36], other pathways of MPs into the atmosphere include waste incineration, industrial emissions, infrastructural degradation, sea sprays, particle resuspension, aerosolization of contaminated vegetation of soil and traffic-related particles (ibid).

The published data on the abundance of atmospheric MPs vary with location (urban or rural). Growing population density and increased outdoor and indoor activities are associated with the variation [37]. Dris et al. investigated MPs in urban and suburban sites and observed concentrations of 110 and 53 particles $m^{-2}d^{-1}$. Similarly, Liao et al. (2021) reported higher MPs at an urban transit station than in rural locations like farmland, wetlands, and mountains. Contrastingly, another study found higher MP concentrations in rural sites than in urban areas, with averages of 396 and 137 particles $m^{-2}d^{-1}$, respectively, due to a "comb-out effect," the ability of plants to filter particles from the air. The size range reported across studies also varies; Hamburg (60%: < 63 µm, 30%: 63-300 µm) [38], Portugal (indoor: 250 µm, outdoor: 299 µm) [39], and China (50%: < 500 µm) [40]

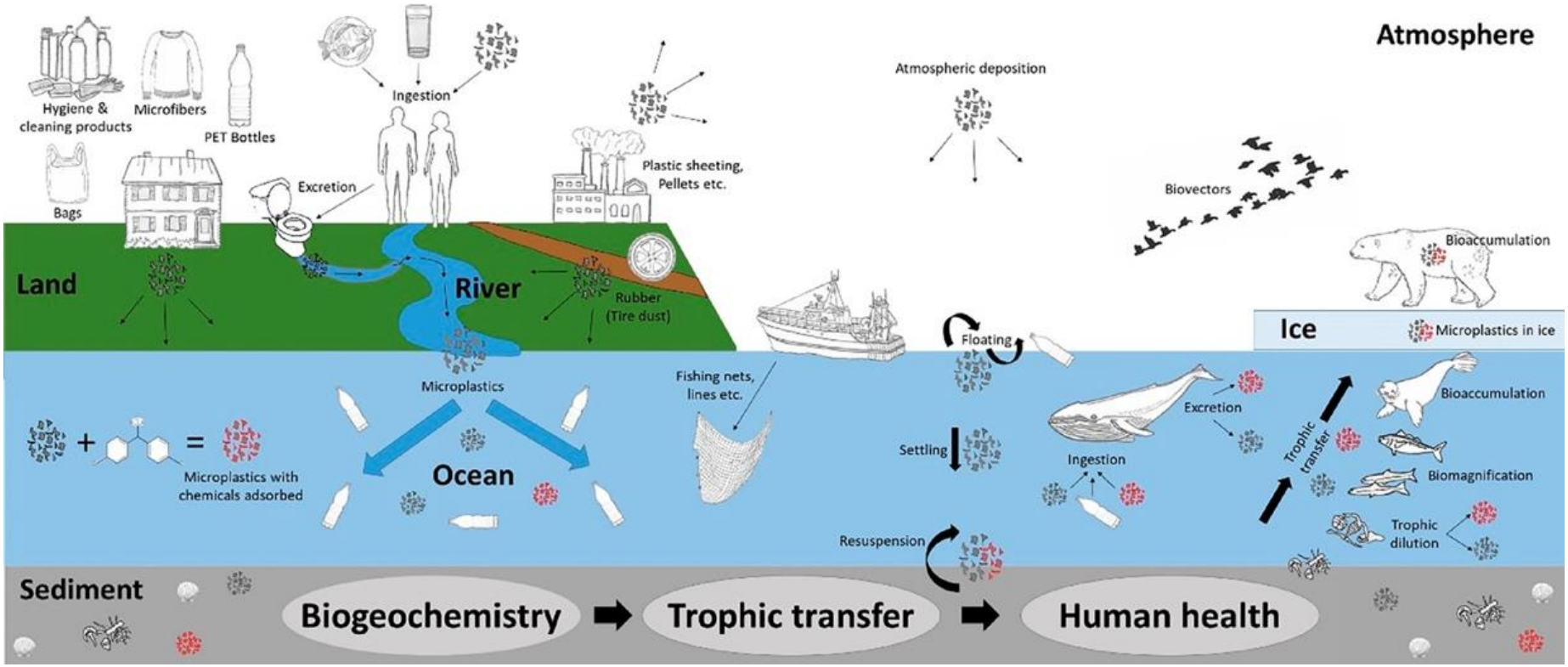


Figure 3 Microplastics distribution in different environmental compartments. The diagram represents the source and distribution routes of microplastics in the atmosphere, land, river, ocean and sediments.

2.3 Biological Impact of Microplastics

Ingestion and Physical Effects

One of the most direct impacts of microplastics on biological systems is through ingestion. Marine and freshwater organisms, ranging from plankton to larger fish and mammals, can mistake these tiny polymers for food due to their small size and often colorful appearance. This ingestion can lead to physical blockages in the digestive tracts of these animals, reduced energy intake, and even starvation, as the particles can accumulate and occupy space within the gastrointestinal system without providing any nutritional benefit [41].

Chemical Toxicity

Beyond the physical dangers posed by microplastics, they can also act as vectors for chemical harm. MPs, because of their large surface area and hydrophobicity are known to absorb and concentrate surrounding pollutants, including persistent organic pollutants (POPs), heavy metals, and other toxic substances from aquatic environments [42]. When ingested, microplastics can thus introduce these concentrated toxins into the bodies of aquatic organisms, leading to toxicological effects ranging from endocrine disruption to reproductive harm [43]. Furthermore, the additives and plasticizers inherently present in many plastics can leach into the organism, potentially causing further biochemical or physiological disturbances [44].

Trophic Transfer and Biodiversity Loss

The effects of microplastics can be magnified through trophic levels as predators consume prey that have ingested microplastics (Figure 3). This process, known as trophic transfer, can lead to bioaccumulation of MPs and associated pollutants in top predators, potentially impacting them negatively [45]. The widespread presence of microplastics can reduce the overall biodiversity of aquatic ecosystems by negatively affecting species survival and reproduction rates [27].

Implications for Human Health

The cycle of plastic pollution extends to human health risks through the consumption of contaminated food, inhalation, and skin exposure [46]. Although the extent of health impacts on humans is still under investigation, the potential for exposure to plastic-associated chemicals and pathogens adsorbed onto MPs raises valid public health concerns.

2.4 Microplastics as Emerging Micropollutants in Wastewater Treatment Plants

Plastics enter the environment through diffuse or point-source pollution, and WWTPs have been identified as a crucial pathway for MP emission [47]. WWTPs receive and treat raw wastewater from households and/or industrial sources, including organic or inorganic materials (grease, coarse debris, MPs, chemicals, and more), producing treated water and sludge.

Influent water into a WWTP is typically handled in a 2-stage treatment process, with an optional third (tertiary) treatment phase provided the effluent from the second stage needs to meet more stringent quality standards or is to be repurposed [48]. While WWTPs can function as a barrier to the incursion of pollutants into the aquatic environment, they can also become a significant entrance route for MPs. The pathways of MPs into WWTPs include [15][49]:

a. residential waste streams typically associated with the fibre emanating from laundry wastewater and the use of cosmetic products (i.e. shower gels, facial cleansers, toothpaste, etc.)
b. leachates from landfills
c. effluent wastewater from industrial activities
d. stormwater dry and wet deposition of MPs

MPs entering a WWTP will differ in characteristics (colour, shape, size, and polymer type) depending on the diversity of raw water into the plant, population density and lifestyle, seasonal changes, and economic conditions of the study area [17]. Based on morphology, the forms of MPs in raw water samples have been observed as fragments, fibers, foam, pellets, irregular and films [50] [51], with fiber and fragment observed as the predominant shapes [52], [53]. Another research showed a higher proportion of fragments than fibres, with 65% fragments (> 500 μm) and 21% fibers (250 - 500 μm) [54] observed. By polymer type, Polystyrene (PS), Polyvinyl Chloride (PVC), Polyethylene Terephthalate (PET), Polypropylene (PP), Polyurethane (PUR), and Polystyrene (PS) have been observed [55][56]. Some of the colour distributions of MPs include blue, black, red/orange, yellow, green, and white [57].

While the size of MPs in WWTPs (influent and effluent) is study-specific and cannot be generalized as they are subject to the origin of the plastic particle, level of fragmentation during transport to and/or in the plant and in-situ treatment technologies, some of the most common

distributions observed vary from small (1 micron to 1 millimetre) to large (1 to 5 mm) [17]. Likewise, MPs originating from PCPs are known to have predefined dimensions since they are purposely added [58]. Table 1 shows the shape, size, and polymer type distribution of MPs in different WWTPs as observed in some studies.

Besides the liquid effluent from WWTPs being a sink for MPs, sewage sludge has also been identified as a sink for these micropollutants [59]. Sewage sludge is generated in the primary treatment phase (primary sludge) and the secondary biological treatment stage (activated sludge). Zhang et al. [60] suggest that under 3% of influent MPs are discharged with liquid effluent, while over 90% are intercepted and sequestered within the sewage sludge matrix. Another study [61] shows that about 80% of MPs in raw water influent to a wastewater treatment plant are retained in the sludge.

As with the water line in WWTPs, the size, polymer type, shape, and abundance of MPs in sewage sludge have also been investigated. According to Zhou et al. (2020) [62] and Hu et al. (2019) [63], the size of MPs in sewage sludge varies from < 1mm to > 5mm, with fragments and fibers observed as the predominant shapes. Another study observed that over 85% of MPs in sewage sludge were smaller than 0.5 mm, of which 44.73% was between 0.1 to 0.2 mm in size, while 29.45% fell within the 0.2 to 0.5 mm range [64], [65]. Xiuna et al [66] detected four types of MP shapes in dewatered sewage sludge including fragment, fibre, film, and sphere. PVC, PE, PB and PTFE are some of the polymer types observed (ibid).

Like with the MPs in the liquid flow, the abundance of particles in the sewage sludge is influenced by varying environmental factors in the study area and treatment technologies [67]. Table 2 shows the characteristics of MPs in sewage sludge obtained from WWTPs in different countries.

Table 1 Polymer type, shape, abundance, and size distribution of MPs in the influent and effluent of different WWTPs

Location & Population Served	Size Distribution		Shape Distribution		Polymer Distribution	
	Influent	Effluent	Influent	Effluent	Influent	Effluent
Seyhan WWTP, Turkey (1,000,000) [68]	53.6% (1–5 mm) 23.0% (0.5–1 mm) 21.8% (0.1–0.5 mm) 1.7% (< 0.1 mm)	34.9% (1–5 mm), 34.9% (0.5–1 mm) 27.0% (0.1–0.5) 3.2% (< 0.1 mm)	54.8% fiber 26.8% fragment 18.4% film	44.4% fiber 30.2% film 25.4% fragment	50.8% PEST 29.2% PE 13.8% PP 6.2% others	43.80% PEST[1] 31.30% PE[2] 18.80% PP[3] 6.30% Nylon-6[4]
Yüreğir WWTP, Turkey (500,000) [68]	59.2% (1–5 mm), 24.6% (0.1–0.5 mm) 14.7% (0.5–1 mm) 1.4% (< 0.1 mm)	40.5% (1–5 mm) 27.0% (0.5–1 mm) 29.7% (0.1–0.5) 2.7% (< 0.1 mm)	87.7% fiber 10.0% fragment 2.4% film	86.5% fiber 10.8% fragment 2.7% film	61.9% PEST 23.8% PE 11.9% PP 2.4% others	68.80% PEST 18.80% PE 12.50% PP
Panyu District, Guangzhou, China (300,000) [69]	-	41.75% (<100 μm) 41.74% (100-550μm) 9.71% (550-1000 μm) 6.80% (>1000 μm)	-	43.69% fiber 38.83% fragment 17.48% pellet		40% PET[5] 17% PP 10% PDAP[6] 8% PE 8% HDPE[7] 6% Others
Liwan District Guangzhou, China (1,427,000) [69]	-	56.32% (<100 μm) 30.33% (100-550μm) 8.66% (550-1000 μm) 4.69% (>1000 μm)	-	57.40% fiber 33.5% fragment 9.03% pellet	-	35% PET 26% PP 12% PE 5% PDAP 4% PA[8] 4% PS[9] 10% Others

1. PEST: Polyester 2. PE: Polyethylene 3. Polypropylene 4. Nylon-6: Polycaprolactam 5. PET: Polyethylene terephthalate

6. PDAP: Poly (diallyl phthalate) 7. HDPE: High-density polyethylene 8. PA: Polyamide 9: PS: Polystyrene

Table 2 MPs abundance, polymer type, concentration, and size distribution in sewage sludge across select WWTPs in different regions.

Location & Capacity	MP Conc. in influent	Sludge Conc.	Polymer Type	Shape	MP range (μm)
Spain (8,000 m^3/day) [70]	16.1 MPs/L	24 MPs/g (DW)	36.0% PET 25.0% PS 20.0% PA 9.0% PVC	57.0% fragment 33.0% fiber	-
Korea (130,000 m^3/day) [71]	13.9 MPs/L	13.2 MPs/g	-	6.0 fibers/g 7.1 fragments/g	10.6 MP/g (106–300 μm) 2.5 MP/g (>300 μm)
China (300,000 m^3/day) [72]	16 MPs/L	2,920 MPs/kg	-	63.0% fiber 37.0% fragment	41.0% (0.08–0.55 mm) 51.0% (0.55–1.70 mm) 8.0% (1.70–5.00 mm)
France (80,000 m^3/day) [73]	244 MPs/L	16.1 MPs/g (DW)	25.0% PS 20.0% PET 18% PE 15.0% PP 10.0% PA and others	77.0% fiber and others	55.0% (200–500 μm) 20.0% (80–200 μm) 20.0% (>500 μm) 5.0% (20–80 μm)

2.5 Fate of Microplastics in Wastewater Treatment Plants

WWTPs conventionally operate in a sequence of phases: primary, secondary, and tertiary processes. These stages collectively function to treat the influent wastewater before the effluent is released into environmental water bodies or sludge is disposed of. Studies [74][75][76] have shown that the amount of MPs entering WWTPs can vary between 10^3 to 10^8 particles per cubic meter of water. Likewise, another research estimated that about 8 trillion microparticles are released daily into the aquatic environment through WWTP [77].

Therefore, an in-depth understanding of the movement of MPs within this treatment framework is crucial, as it enables a detailed assessment of the stage-specific removal efficiencies and fosters the optimization of treatment strategies to more effectively manage these emerging contaminants [78].

2.5.1 Microplastics in the Primary Treatment Stage

The primary treatment phase of a WWTP begins with the preliminary separation of large solids using bars and screens, followed by physicochemical treatments designed to sediment and precipitate suspended solids, reduce the biochemical oxygen demand of the organic matter, eliminate volatile contaminants, and remove substances like greases and oils.

During the primary treatment phase in WWTPs, the removal of microplastics (MPs) is significantly influenced by their physical properties, particularly density. Polymers that are denser than water such as polyvinyl chloride (PVC), polyethylene terephthalate (PET), and polystyrene (PS), are more likely to be eliminated by gravity through the

sedimentation/primary clarification process. Concurrently, MPs of lighter, and more buoyant polymers, can be effectively removed by skimming alongside grease, oils, and fats [79].

In a study conducted by Dris et al. [80], most MPs (45% fiber: 1000 to 5000 μm) from urban wastewater entering the primary treatment phase were removed by early-stage skimming and settling, attributable to their ability to adhere to suspended solids (SS). MP concentration of 260-320 particles/L was reduced to 50-120 particles/L - showing a 71% average extraction efficiency. Talvitie et al [81] also observed a removal efficiency of 97.76% for a grab sampling of wastewater containing 636.7 particles/L (D > 300 μm) in influent and 14.2 MPs/L (100-300 μm) in the effluent of the primary treatment. Removal efficiency lower than 70% is not uncommon for primary-stage treatment. Hyuk et al. observed a removal efficiency of 69.52% and 58.62% during screening and primary clarification [82].

2.5.2 Microplastics in the Secondary Treatment Stage

The secondary treatment phase of wastewater, also known as biological treatment, is designed to substantially remove organic matter and reduce suspended solids in wastewater with the aid of microorganisms. This phase can be carried out anaerobically, anoxically, or aerobically (activated sludge process, sequence batch reactor, trickling filters, membrane bioreactor, and moving bed biofilm reactor) [83]. In this phase, effluent MPs from primary treatment may attach to the unstable biomass due to biofouling and then settle out by density. The hydrophobic nature of MPs plays a significant role in accelerating their binding with biomass or sludge in biological reactors [82].

Removal efficiencies of MPs during the secondary treatment have been reported to vary widely. For the activated sludge process (ASP), MP removal efficiencies of >97% [75][84] [85] have commonly been shown. Secondary treatments such as anaerobic+anoxic+aerobic (A^2O) [86], bioreactor [76], and anoxic tank+clarifier [87] process have shown comparably high (>90% to >98%) removal rates. However secondary treatment using biofilters, ASP, and aeration mechanism with less efficient (<90%) MPs removal rates [80][80][88][89] are not uncommon and can be attributed to a variety of characteristics of the WWTP and the MPs inflow. The exact dynamics of how MPs interact with biomass and the effectiveness of MP removal with varying biological treatment mechanisms require further research.

2.5.3 Microplastics in the Tertiary Treatment Stage

After primary and secondary treatments, tertiary treatment systems target residual solids, pathogens, organics, nutrients, and emerging pollutants like MPs. Despite the reasonably high MP removal efficiency recorded by the primary and secondary stages, the effluent from these stages still contains significant microparticles, most especially fibres [90] that may be further eliminated by tertiary treatment technologies [91] such as dissolved air floatation (DAF), membrane bioreactor (MBR), rapid sand filtration (RSF), and disc filters (DF).

While WWTPs are not particularly designed to remove MPs, tertiary treatment technologies may significantly improve removal efficiency [92]. A study comparing the efficiency of DF (pore sizes 10 and 20 μm), RSF, DAF and MBR for MPs > 20 μm, shows a removal rate ≥95% for all technologies (MBR = 99.9%) except the DF with a pore size of 10 μm (40%) due to membrane fouling and accelerated backwashing [91].

The removal efficiency of MPs in WWTPs is influenced by particle size, morphology, and polymer type. Primary treatment effectively captures larger microplastics and fibres [14]. Secondary treatment significantly reduces smaller fragments through agglomeration and assimilation by activated sludge. Tertiary processes excel in removing specific forms, such as pellets, and are effective against microplastics of minimal particle sizes [93].

2.6 Microplastics Research in Estonian WWTPs

In Estonia, the study of MPs in WWTPs is still in its nascent stage, but some research is beginning to emerge, particularly spurred by the recent revision of the Urban Wastewater Treatment Directive (UWWTD 91/271/EEC) which mandates the inclusion of advanced treatment and monitoring of MPs in the sludge and liquid line of residential WWTPs.

Ayankunle et al. [94] conducted a recent study by estimating microplastic concentrations from laundry wash (LW) and personal care products (PCPs) in the wastewater influent and effluent of major Estonian cities, including Tallinn, Tartu, Narva, Pärnu, Rakvere, Viimsi, Viljandi, Kuressaare, Paide, Võru, Sillamäe, Valga, Keila, Põlva, Põltsamaa, Rapla, Haapsalu, Elva, and Haljala consisting about 760,000 inhabitants (57% of the Estonian population) in total. The study utilized a material flow analysis predictive model to measure the release of MPs from LW and PCPs in households connected to wastewater treatment plants (WWTPs) in the areas of interest. It is estimated that the annual MPs load per capita from PCPs ranges from 4.25 to 12 tons, while for LW, it is between 3.52 to 11.24 tons (ibid).

Furthermore, the estimated MP load relevant to wastewater varies significantly. In the influent stream of WWTPs, the load is estimated to be between 700 to 30,000 kg/year. In contrast, the effluent stream, which is the treated wastewater discharged from WWTPs, shows a considerably lower estimated microplastic load, ranging from 2 to 1500 kg/year (ibid). In alignment with Cai Y et al. [95], Ayankunle et al. [94] also discovered that microfibers (200 to 600 μm) were the most dominant MPs in the in-situ sampling results, taking up more than 75% of the total MP count. The outcome of the research though provides a great understanding of the Estonian environment, but it also underscores the need for a broader study into the identification and quantification of MPs and more efficient capture strategies.

Another systematic analysis of MPs in both the influent and effluent streams of Estonian WWTPs conducted by Ayankunle et al. unpublished [96], utilizing a continuous sampling filtration method coupled with Fourier-transform infrared spectroscopy (FTIR-spectroscopy) examines the efficiency of six WWTPs across Estonia. The findings of the research revealed the presence of MPs in all samples, with the highest concentrations detected in the influent to the screening phase of the primary treatment stage. The predominant types of polymers identified were polyester, polyethylene, and polypropylene.

While the cited studies establish fundamental baseline levels of MPs in the influent and effluent of Estonian WWTPs, this study goes further to investigate the abundance of MPs and estimate stagewise removal efficiencies.

2.7 Regulations for Mitigating Microplastics Pollution in Estonia

Estonia, as part of the European Union, aligns with EU regulations and strategies to combat microplastic pollution. The EU commission put out a proposal (first published in October 2022, and now amended) [97] for revising the existing outdated Urban Wastewater Treatment Directive (UWWTD 91/271) from 1991. This update is intended to make tertiary treatment mandatory for all urban WWTPs with p.e ≥100,000 (population equivalents) by the end of 2038, and for all residential WWTPs with 10,000-100,000 p.e. by the end of 2043 where higher concentrations of micropollutants (including microplastics) pose a risk to human health or the environment. The revision requires member states to monitor microplastics at the inlets and outlets of municipal WWTPs serving >10,000 people and in the sewage sludge of these WWTPs. The Commission also intends to establish a standardized methodology for measuring microplastics in municipal wastewater and sewage sludge to ensure the directive's uniform application.

3. Materials and Methods

3.1 Description of the Keila WWTP

The Keila City wastewater treatment plant (WWTP) (Figure 4) is in North-Western Estonia. Raw wastewater to the Keila plant originates from households, industries, and stormwater. The plant serves a population of 18,233, with an average flow rate of 1,746 m^3 per day and discharges its effluent into the biological pond prior reaching the flowing stream. The wastewater is pumped to the treatment process line from the primary wastewater pumping station within which also collects septic wastewater from household not connected to the sewer lines.

The primary treatment stage comprises a 6 mm lamella mechanical screen and a grease and grit separator unit converting the turbulent flow to a laminar flowrate prior reaching the biological process tank operating in anaerobic-anoxic-aerobic (A2O) phases. The facility adds aluminum Chloride ($AlCl_3$) to enhance mixed liquid suspended solid (MLSS) flocculation and coagulation processes. The MLSS flows into secondary clarifier for efficient liquid-solid phase separation, 20% of the MLSS is returned into the biological process tank to maintain the microorganism's population.

Three parallel sampling points (S1, S2, and S3) were selected: influent, after-screen (AS) and final effluent (Figure 5) the triplicate samples were collected – to evaluate the abundance of MPs and the stagewise removal efficiency of the treatment plant. The raw water samples were collected on July 10-11, 2023.

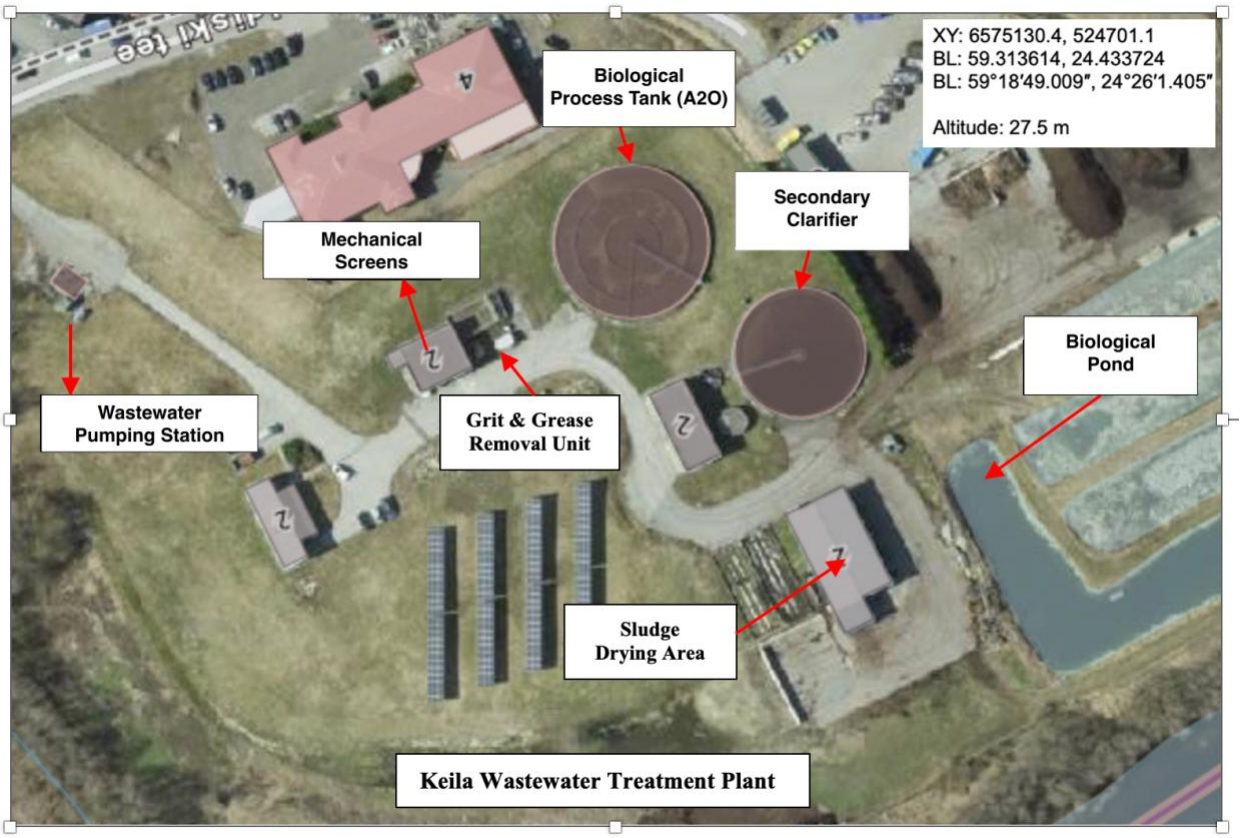


Figure 4 Location and description of the WWTP.

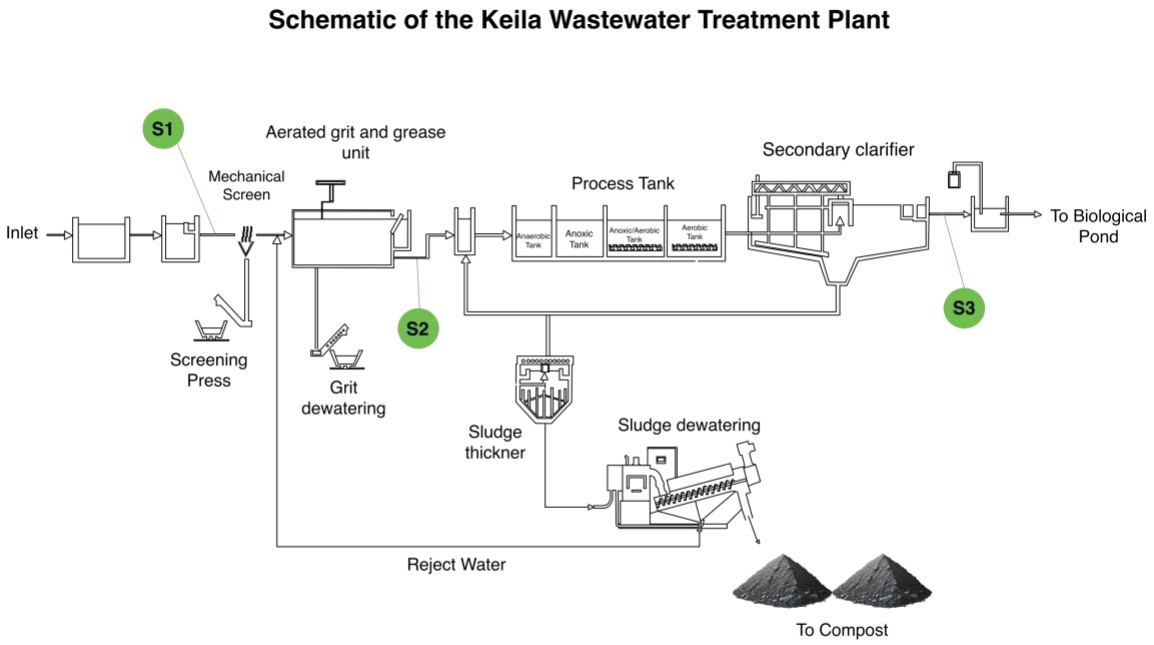


Figure 5 Schematic of the Keila WWTP, highlighting key unit operations and sampling points (S1, S2, and S3)

3.2 Sampling

24-hour composite samples were collected with automatic sampler to capture the varying pollutants concentrations in the wastewater. Wastewater samples from the raw influent (10 L), after primary treatment stage (48 L) and from the final effluent (1440 L) were filtered through a double-layered sieve system for immediate in-situ fractionation, upper layer sieve with 400 µm and the finest mesh with 100 µm pore diameter. The retentate materials on the mesh screens were rinsed with 50 ml of Milli-Q water into a glass beaker covered with aluminum foil paper, and transferred to the laboratory, stored at 4^{o} C until digestion. The sample was collected between 10.07.2023 to 11.07.2023 at Keila WWTP (Table 3).

Table 3 Description of sampling campaign at Keila WWTP.

Treatment Step	Raw Influent	After Mechanical Screen	Final Effluent
Sampling frequency	400 ml /45 mins	100 ml/30 mins	1000 ml/mins
Total Volume (L)	10 L	48 L	1440 L

3.3 Sample Digestion Protocol

In this study, Fenton's reagent protocol (H_2O_2 + Fe catalyst solution) as described by Mohammed et al. [98] is used to isolate MPs from organic-rich samples due to its high digestion efficiency, little polymer degradation, and rapid oxidation of organic matter [99], [100]. However, the protocol was optimized for this study, and the steps are:

a. Preparation of Fenton's Reagent

To prepare the Fenton's reagent, approximately 1g of $Fe_2(SO_4)_3$ was diluted in 50.945 ml of Milli-Q water to obtain 0.05M solution. The pH of the resulting solution was controlled by adding 0.1 ml of diluted sulphuric acid 0.5M H_2SO_4 to achieve a pH of 2.5 to 3.

b. Organic Digestion

The raw water samples were first homogenized using a mechanical laboratory shaker and three technical replicate subsamples 2 ml each was pipetted to a conical flask. 10 ml of Fenton's reagent was added and then shaken to ensure adequate mixing. 20 ml of Hydrogen Peroxide (H_2O_2) (35%) was added to the mix to initiate an exothermic reaction. After every 1 minute, 5 ml H_2O_2 is added up to 10 times. The reaction is allowed to cool for 20 minutes and 4 ml of concentrated H_2SO_4 is added to obtain a clear solution. To prevent the adhesion of MPs to the

walls of the flask, 5ml surfactant (1ml tween and 99 ml MilliQ) was added and rigorous shaking was avoided because of foaming.

c. Filtration

The digested samples were initially filtered through a metallic filter with a pore size of 10µm. The retentates is rinsed off the metal filter with Milli-Q water onto a cyclopore filter with a pore size of 10µm. The use of the metallic filter for the initial filtration was to prevent acid reaction with the cyclopore (nylon) filter. The filtered samples on cyclopore filter were transferred to petri dishes and dried in an oven (SANYOMOV-212F) for 15 minutes at 60 °C before visual identification.

3.4 Visual Identification of Microplastics

All filtered samples were visualized using a Nikon stereomicroscope SMZ1270 12.7x (0.63 - 8x magnification), with 4x magnification. The size, colour and shape of MPs were observed using the NIS Elements BR software®. All identified particles were counted, photographed, and suspected non-plastics material were picked with forceps, scratched to observe brittleness of the particles.

To effectively distinguish between MPs, some characteristic differences are provided in Table 4 [101], [102], [103]. According to Strady et al. [104] the visible internal structure of all MPs should not have repetitive identical sequences (organic structures), as with organic fibre, and they must exhibit a distinct and homogenous colour. The size range of interest for this study is particles >300 µm (0.3 mm) due to detection and analytical capabilities, and the need to assess the efficiency of the WWTP in removing larger particles otherwise considered easier to filter out.

Table 4 Microplastic type and characteristics

Microplastic Type	**Morphological Characteristics**
Fibers	Elongated, thread-like
Fragments	Irregular shapes, jagged edges
Films	Thin, flat, flexible
Foams	Lightweight, porous, sponge-like
Pellets	Small, spherical, or cylindrical, uniform, hard
Microbeads	Small, spherical, or irregular, smooth surface

3.5 Polymer Identification

In this study 60% of the visually identified MPs were chemically characterized with μFTIR-IR (PerkinElmer Spectrum Spotlight 400). The FTIR-analyses were performed by Dr. N. Buhhalko from the Department of Marine Systems (TalTech). The digested samples were analyzed in point mode, coupled with an MCT detector. The spectrometer scanned from 4000 to 420 cm^{-1}, covering the infrared range essential for identifying polymer-specific functional groups. From the observed particle spectra, those that matched a quality index (QI) > 85% when compared against a spectral library were accepted as polymers. The reference spectral library which includes the most frequently encountered types of plastic polymers as well as natural cellulose polymers was developed at the Department of Marine Systems at Tallinn University of Technology, alongside contributions from the Leibniz Institute for Polymer Research in Dresden and Aalborg University's Department of the Built Environment in Denmark.

3.6 Estimation of Removal Efficiency

The removal efficiency of MPs was calculated using the following equation [105]:

$$RE\ (\%) = \frac{Cinlet - Coutlet}{Cinlet}\ x\ 100 \text{ ..... (eqn 1)}$$

Where C_{inlet} is the concentration of MPs in the influent of the sampling stage and C_{outlet} is the concentration of MPs in the effluent of the subsequent stage. The expression is used to estimate stagewise removal efficiency and the overall MP removal efficiency of the WWTP.

3.7 Statistical Analysis

The data analysis in this study was performed using Microsoft Excel 2021. The mean and standard deviations of the data set were calculated, and an ANOVA single factor was conducted to examine the relationship between particles sizes observed in the different sampling points.

3.8 Quality Control

3.8.1 Contamination control

In this study, several measures to prevent contamination were implemented as detailed below:
1. To evaluate potential onsite or laboratory contamination, control and experimental samples were established. Cyclopore filter papers were placed inside glass containers located both within and near the sampling and testing areas.

2. All experimental procedures were conducted inside a fume hood, except for the filtration processes.
3. Laboratory coats made from 100% cotton were used throughout the experiments to prevent the introduction of synthetic fibres into the samples.
4. To protect against airborne contaminants, all samples-containing glassware, including the refrigerated samples were consistently covered with aluminum foil when not in active use, including times outside of handling or adding reagents.
5. All glassware and equipment were cleaned with MilliQ water to remove any residues.

4. Results and Discussion

4.1 Statistical Analysis of Results

The descriptive statistics of the size range observed during visual identification in Table 5.

Table 5 Descriptive statistics of the visually observed MP sizes across the three sampling points

Statistic	**Value**
Count	682
Mean	0.7 mm
Standard Deviation	0.42 mm
Minimum Size	0.3 mm
25th Percentile	0.42 mm
Median (50th Percentile)	0.56 mm
75th Percentile	0.8 mm
Maximum Size	5 mm

An ANOVA single factor was performed to test if there are statistically significant differences in the MP sizes at the influent, after screen and effluent observed during microscopic identification. The result of the ANOVA shows a p-value of 0.4116. Since the p-value is >0.05, there is no sufficient evidence to reject the null hypothesis. Hence, there are no statistically significant differences in the MP sizes across the sampling points.

4.2 Contamination Control

During sample collection at the treatment plant, and preparation and analysis at the laboratory, the deposition of air-bound particles might be inevitable. Due to the ubiquitous nature of MPs in the environment, they are transported by airflow and may contaminate samples intended for MP detection, posing a challenge to data consistency [106].

In order to estimate air contamination and ensure the consistency of data used in this study, petri dishes with cylopore filter were left open during sample collection, laboratory analysis, and microscopic inspection. Experiments were conducted in fume hoods to further mitigate air contamination as demonstrated by Bhat et al. 2024 (ibid). The result of the contamination showed that there was no contamination of the results, as the observed MPs were all below 300 µm in size and therefore were excluded from the analysis.

4.3 Recovery Controls

To validate the efficiency of the digestion protocol and evaluate the potential loss of MPs, a recovery experiment was conducted with cotton microfibers and cryo-milled polymer pellets PP (1 mm) and high-density polyethylene (HDPE) (0.5 mm) which were initially counted and subjected to the digestion and filtration protocol (Fenton's reagent). The recovery samples were observed under stereomicroscope to obtain the number count of MPs in other to estimate the recovery rate. The result showed that post digestion, 83% of PP (>300 µm), 76% of HDPE (>300 µm), and 91% of microfiber cotton were recovered.

4.4 Characterization of Microplastics by Shape

The morphology of MPs observed in wastewater samples in this study can be classified into five (5): fibre, fragment, film, foam, and sheet (Figure 6). Fibre exhibited significant length to width ratio, while fragments were opaque and with irregular shapes. Film and sheet have flat surface, whereas foams were sponge-like and porous.

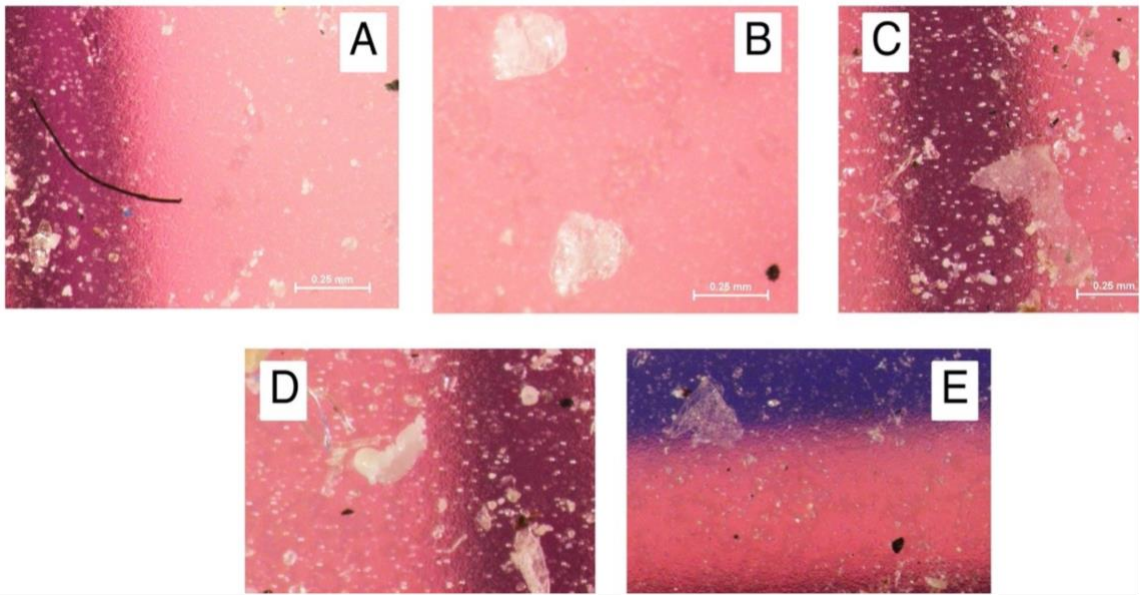


Figure 6 Types of microplastic particles found in this study (classified by shape) A. black fiber, B. white fragment, C. transparent film, D. white foam, E. transparent fragment.

The percentage distribution of MPs found in the influent, after screen, and effluent are shown in Figure 7. It can be seen that in all the sampling points, fibre was the dominant shape, followed by fragment, both constituting more than 98%. Previous studies report fibre and fragment as the dominant MPs found in wastewater samples [106] [107]. According to Cristaldi et al [108], fibers in the influent waters accounted for 79% of the total MPs. Yang et al [109] also reported a dominance of 85.9% for microfibers, which aligns with the results obtained in

this study. In the influent samples fibre accounted for 88.6 – 94.8% with an average of 91.5% of MPs while fragment, sheet and film constituted 7.06%, 0.9% and 0.50%. The percentage distribution of fibre in the after screen was 94.8%, while fragment, and foam made up the total with 4.9% and 0.4%, respectively.

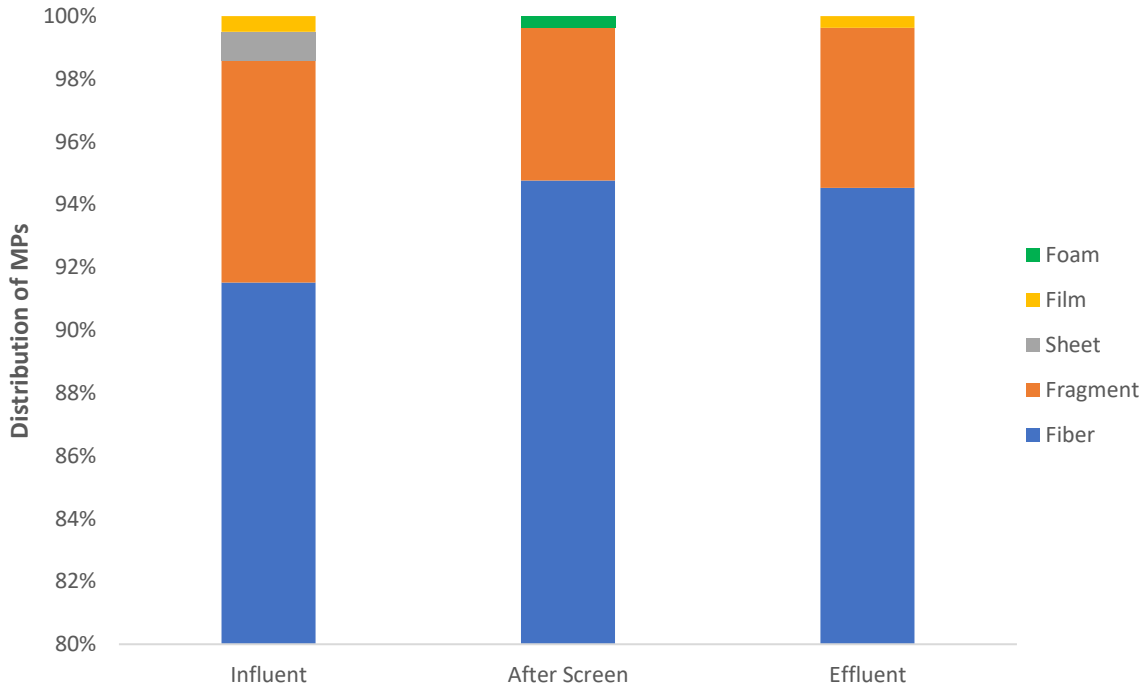


Figure 7 Microplastics categorization by shape in the influent, after screen and effluent samples.

In the effluent samples, the abundance of fibre ranged between 91.2 – 98.7%, with an average of 94.5%. Fragment and film constituted 5.1% and 0.4%. The dominance of fibre in the influent stream may be attributed to home laundering activities resulting in the release of large amounts of synthetic materials [86] [110].

4.5 Characterization of Microplastics by size and colour

The size distribution of MPs in the influent, after screen and effluent streams were analysed per the baseline for this study (>300 µm or 0.3 mm). Visually observed MPs ranged between 300 – 5000 µm (0.3 to 5 mm) - the categorization employed in this study is: 300-500 µm (0.3 to 0.5 mm) and >500 µm (0.5 mm). In the influent samples, MPs between 300-500 µm, represented 44.2% of the total MP load. Particles >0.5 mm, accounted for 55.8% of MPs in the influent (Figure 8), indicating that larger MPs (mostly fiber – Figure 8) are more abundant in

the influent than the smaller ones. An observation that aligns with the reported abundance in other studies where MPs > 500 µm were more dominant [111], [112] in collected samples.

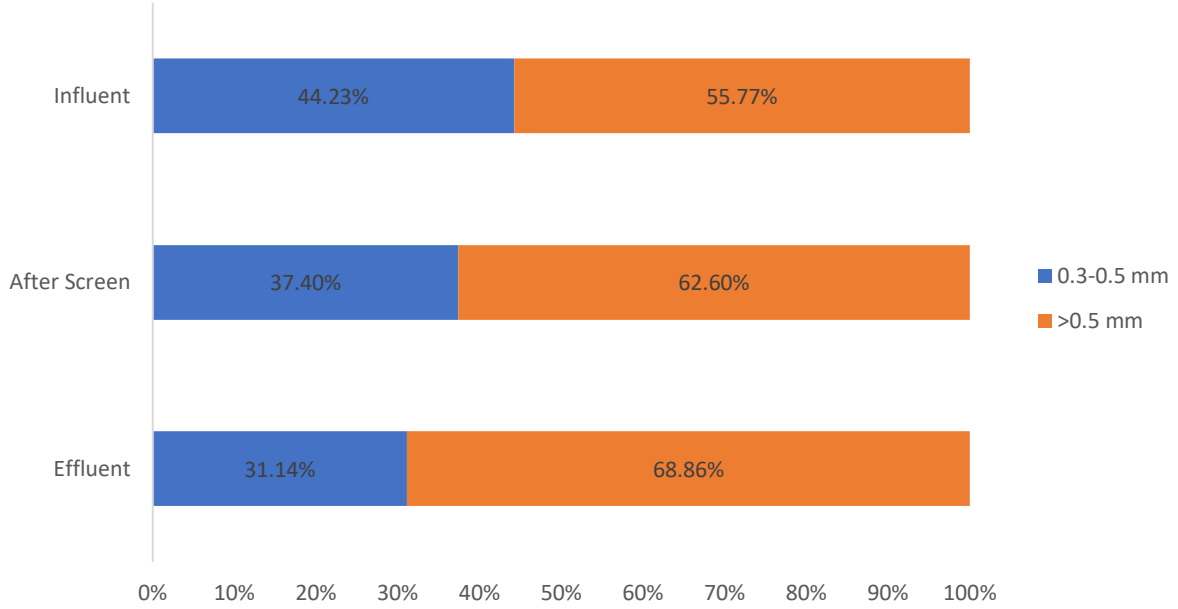


Figure 8 Microplastics categorization by size in the influent, after screen and effluent samples. In the after screen, MPs between 300 – 500 µm accounted for 37.4% while those >500 µm made up 62.6% of the total MPs. The abundance of MPs > 500 µm indicate that the grit and grease chamber removed particles between 300-500 µm more efficiently.

Table 6 Concentration of differently shaped microplastics (MPs/L) in two size categories (0.3-0.5 mm and >0.5 mm) in the influent, after screen and effluent samples.

	Influent		**After Screen**		**Effluent**	
Size Range (mm)	**0.3-0.5**	**>0.5**	**0.3-0.5**	**>0.5**	**0.3-0.5**	**>0.5**
Fiber	63.3	95	14.24	26.22	0.35	0.85
Fragment	10.8	1.67	1.56	0.52	0.052	0.046
Sheet	1.67	0.00	0.00	0.00	0.00	0.00
Film	0.83	0.00	0.00	0.00	0.006	0.00
Foam	0.00	0.00	0.17	0.00	0.00	0.00
Total (MPs/L)	**76.67**	**96.67**	**15.80**	**26.74**	**0.41**	**0.90**

In the effluent samples, the majority of MPs were larger than 500 μm (68.9%), while 31.1% were between 300 – 500 μm, an indication that the activated sludge system (A2O) also removed MPs between 300 – 500 μm more efficiently than those >500 μm.

The abundance of MPs > 500 μm in the after screen and effluent samples may be attributable to minimal fragmentation of plastic particles and low hydraulic retention time resulting in sediment overflow and particle resuspension. Petroody et al. [87] suggests that the size and shape of MPs may affect their retention in wastewater treatment processes. Table 6 shows that fibers were the most abundant MPs in the effluent of the primary treatment and final effluent, attributable to the tendency of fibers to pass longitudinally through treatment stages more easily than other forms [113].

The most common color distribution observed was white (31.43%), followed by black (20.08%), blue (16.47%), and brown (10.57%). Other colors (9.88%) observed in this study include orange, yellow, pink, red, grey, and green. The colour distribution in this study is consistent with the outcome of other studies where white/transparent and black MPs were the most abundant [114], [115].

4.6 Chemical Characterization of MPs

For the chemical characterization of MPs in this work, 60% of the visually identified MPs were subjected to FTIR analysis. The FTIR analysis identified six (6) polymers, which includes polyethylene terephthalate (PET), polypropylene (PP), polystyrene (PS), polyethylene (PE), polyvinylchloride (PVC), and Polyacrylonitrile (PAN). The spectra for each of the detected polymer is shown in Appendix I.

In the influent PET had the highest abundance (56.2%) followed by PP (26.6%) and PE (14.3%) (Figure 9). The least occurring polymers were PAN (1.9%) and PS (0.9%). Likewise, the most abundant polymer constituent in the after screen was PET (64.9%), followed by PP (23.4%). PE constituted 9.9%, while PAN and PS each made up 0.9%. The polymer type observed in the effluent were PET (74.5%), PE (15.5%), PP (7.3%), PAN (1.8%), and PVC (0.9%).

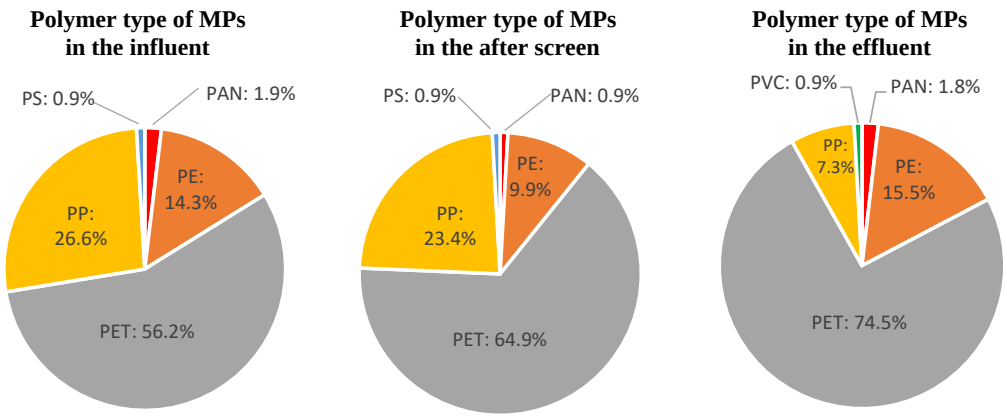


Figure 9 Chemical composition of MPs observed under μFTIR for the influent, after screen and effluent.

The proportion of polymers found by the FTIR analysis in this study follows the decreasing order PET > PP > PE > PAN > PS > PVC. When compared with existing literature [54], [86], the findings from this FTIR analysis align with the outcome of those that have discussed the prevalence and detection of polymers in WWTPs. PET, PE, PS and PP, in no particular order, were observed as the most occurring polymer types in the referenced literature. On the other hand, Hassan et al. [67] and Franco et al. reported high abundance for PVC in the effluent: 61% and for two WWTP, 15% and 40%, respectively. The differences in polymer prevalence found could be linked to the local population's plastic consumption and usage habits [116]. Also, the abundance of PET (fiber), also known as polyester fibre is indicative of the source of MPs – domestic laundering activities.

4.7 Concentration of MPs in Keila in Comparison with Other Studies

In the influent, MP concentration detected ranged from 155 – 197.5 MPs/L, with an average of 173.3 ± 21.8 MPs/L. Fiber, fragment, sheet, and film constituted 158.3 MPs/L, 12.5 MPs/L, 1.67 MPs/L and 0.83 MPs/L, respectively. Considering the daily influx of raw wastewater, which is approximately 1,746 cubic meters (1.746 million liters) in Keila WWTP, it can be estimated that the total number of microplastic particles that enter the plant each day is 302.6 $\times 10^6$ MPs/day.

In the after-screen samples, the concentration detected ranged between 40.1 – 47.9 MPs/L, with an average of 42.7 ± 3.7 MPs/L. Fibre was observed to be 40.5 MPs/L, while fragment and foam had average concentrations of 2.1 MPs/L and 0.2 MPs/L. The concentration of MPs in

the effluent samples ranged between 1.1 – 1.6 MPs/L, with an average value of 1.3 ± 0.3 MPs/L. Fiber, fragment, and film constituted 1.2 MPs/L, 0.1 MPs/L, and 0.01 MPs/L.

Considering the average concentration of MPs in the influent, after screen and effluent stage, the stage-wise and overall removal efficiency of the treatment plant can be estimated. The microplastics discharge from the plant to the environment can also be obtained. In the primary treatment phase of the treatment plant (from mechanical screen to grit and grease chamber), a removal efficiency of 75.4% was achieved (Table 7). This outcome is consistent with the findings across literature where primary treatment has been shown to remove between 70 to 98% of MPs [75], [81], [109].

The use of Aluminium based coagulant in the grit and grease removal chamber of the WWTP to enhance flocculation/coagulation may have contributed to the efficiency of the primary treatment stage, as seen in the study by Ma et al. [117] where the effects of Fe-based coagulant and Al-based coagulant on the removal efficiency of MPs in a portable water plant were compared. The result showed that the Al-based coagulant demonstrated significant effect on MP removal.

The secondary treatment phase which includes the biological tank (A2O) and secondary clarifier reduced microplastics concentration from 42.7 MPs/L to 1.3 MPs/L. According to studies, the secondary treatment makes up for 7 to 20% of microplastics retained in the WWTP [81], [118]. This study shows a 23.9% retention of MPs in the secondary treatment stage.

Table 7 Stagewise and overall MP removal efficiency and the estimated daily load and discharge to and from the WWTP.

Stages	**MP Conc. (MPs/L)**	**MP Conc. (MPs/d) x 10^6**	**MP Conc. (MPs/y) x 10^6**	**Removal Efficiency (%)**
Influent	173.3	302.6	110461.5	0
Primary Treatment	42.7	74.6	27218.7	75.4
Biological Treatment	1.31	2.3	834.9	23.9

Comparing the concentration of MPs in the influent to the final effluent, the overall removal efficiency of the WWTP in this study is 99.2% (Table 7), reasonably aligning with the finding of Edo et al (93.7%) [86] with the same biological treatment mechanism (A2O) Table 8. On the contrary, Do et al. [119] obtained a much lower efficiency (25.5%), for the HC WWTP with similar capacity and MP concentration in the influent. The study attributed the low

removal efficiency in the plant to the possible lack of treatment processes intended for MP capture. Taking the removal efficiency of the plant into account, the amount of MPs discharged to the environment can be estimated to be 2.3×10^6 MPs/day. Despite the noteworthy retention efficiency observed in this study, MPs are still released to the environment in the order of 10^6, highlighting the significance of WWTP as a pathway of microplastic pollution.

Table 8 MP removal efficiency observed across several WWTPs.

Plant Location	WWTP Capacity (m^3/day)	Size Range (μm)	Treatment Process	MPs Conc. (MPs/L)		Discharge/day	Removal Efficiency (%)	Ref.
				Influent	Effluent			
Madrid (Spain)	45,000	25-5,000	Primary, A2O	171	10.7	3×10^8	93.7	[86]
Wuhan, China	20,000	20-5,000	Secondary (Activated Sludge)	79.9	28.4	5.7×10^8	64.4	[53]
Sydney Australia (WWTP B)	17,000	25-500	Primary and secondary	1.44	0.48	8.16×10^6	66.7	[120]
HC WWTP	2,000	1.6-5,000	A2O	413	309	3.8 × 107	25.5	[119]
Keila WWTP	1,760	300-5,000	Primary, A2O	173.3	1.3	2.3×10^6	99.2	This study

5. Conclusions and Recommendations

5.1 Limitations of the Study

Like any research work, this study has its limitations which must be acknowledged.

1. Due to time constraints this study was confined to one WWTP, which potentially limits the generalizability of the findings to other plants with different technological setup and operational scale.
2. The size range of MPs studied were targeted (>300 μm), underestimating the occurrence of smaller MPs in the collected samples.
3. Since one-time 24-hr composite samples were used in this study, the results do not account for seasonal variation in the concentration of MPs which could influence the distribution of MPs at the sampling points.

Therefore, future work should consider expanding the scope of this study by assessing multiple WWTPs in Estonia, accounting for MPs < 300 μm, and considering seasonal dynamics to obtain a more comprehensive outlook of the microplastics distribution in Estonian WWTPs. Despite the limitations, the findings of this study establish the effectiveness of the current treatment technology at Keila WWTP and proposes operational enhancement to improve MPs capture efficiency.

5.2 Improvement of Microplastics Removal Efficiency

While the wastewater treatment technology at the Keila plant shows a high microplastics retention, the MPs discharged to the receiving water body is still very much significant, hence, the need to improve the plant's removal efficiency. Some of the recommendations for improvements include:

1. The efficiency of the primary treatment can be improved by using mechanical screen device less than 3mm diameter unlike the 6mm bars currently in use.
2. In addition, the hydraulic retention time through the primary treatment stage should be systematically increased and monitored to support efficient removal of MPs by density floatation in the oil and grit removal unit.

5.3 Conclusions

This study investigated the abundance and characteristics of MPs in an urban WWTP and estimated the stagewise and overall removal efficiency of the plant. The concentration of MPs detected in the influent, after screen and effluent of the Keila WWTP within the sampling period was 173 MPs/L, 42.7 MPs/L, and 1.3 MPs/L, respectively. Of microplastics shape, fibers were the most abundant, underscoring the significance of the discharge of synthetic fibre from household laundering activities. By size distribution, MPs > 500 μm particles were the most often occurring in all the treatment stages. The most frequently identified polymer type was PET. White-coloured MPs were predominant, followed by black.

The removal/retention efficiency of the plant was obtained as 75.4% for the primary treatment and a further 23.9% for biological treatment (A2O), resulting in an overall efficiency of 99.2%. Since MPs reduced from the influent to the effluent of the WWTP, it indicated that the treatment process was effective at removing them (retaining them in the sludge), despite not being designed for such application.

References

[1] "Global Steel Report 2019 Global Steel Report Introduction and Trends".

[2] "Production - SIPH." Accessed: Mar. 26, 2024. [Online]. Available: https://siph.groupesifca.com/en/natural-rubber/production

[3] "Global plastics production." Accessed: Jan. 18, 2024. [Online]. Available: https://ourworldindata.org/grapher/global-plastics-production

[4] "Plastic pollution is growing relentlessly as waste management and recycling fall short, says OECD." Accessed: Mar. 26, 2024. [Online]. Available: https://www.oecd.org/environment/plastic-pollution-is-growing-relentlessly-as-waste-management-and-recycling-fall-short.htm

[5] "Global e-waste generation 2010-2019 | Statista." Accessed: Mar. 26, 2024. [Online]. Available: https://www.statista.com/statistics/499891/projection-ewaste-generation-worldwide/#

[6] "Global Plastics Outlook," *Global Plastics Outlook*, Feb. 2022, doi: 10.1787/DE747AEF-EN.

[7] "Plastic pollution is growing relentlessly as waste management and recycling fall short, says OECD." Accessed: Jan. 20, 2024. [Online]. Available: https://www.oecd.org/environment/plastic-pollution-is-growing-relentlessly-as-waste-management-and-recycling-fall-short.htm#

[8] M. N. Issac and B. Kandasubramanian, "Effect of microplastics in water and aquatic systems," *Environmental Science and Pollution Research 2021 28:16*, vol. 28, no. 16, pp. 19544–19562, Mar. 2021, doi: 10.1007/S11356-021-13184-2.

[9] E. Curren, V. S. Kuwahara, T. Yoshida, and S. C. Y. Leong, "Marine microplastics in the ASEAN region: A review of the current state of knowledge," *Environmental Pollution*, vol. 288, p. 117776, Nov. 2021, doi: 10.1016/J.ENVPOL.2021.117776.

[10] M. Sighicelli *et al.*, "Microplastic pollution in the surface waters of Italian Subalpine Lakes," *Environmental Pollution*, vol. 236, pp. 645–651, May 2018, doi: 10.1016/J.ENVPOL.2018.02.008.

[11] J. Wagner, Z. M. Wang, S. Ghosal, C. Rochman, M. Gassel, and S. Wall, "Novel method for the extraction and identification of microplastics in ocean trawl and fish gut matrices," *Analytical Methods*, vol. 9, no. 9, pp. 1479–1490, Mar. 2017, doi: 10.1039/C6AY02396G.

[12] R. Li, L. Yu, M. Chai, H. Wu, and X. Zhu, “The distribution, characteristics and ecological risks of microplastics in the mangroves of Southern China,” *Science of The Total Environment*, vol. 708, p. 135025, Mar. 2020, doi: 10.1016/J.SCITOTENV.2019.135025.

[13] J. Gasperi *et al.*, “Microplastics in air: Are we breathing it in?,” *Curr Opin Environ Sci Health*, vol. 1, pp. 1–5, Feb. 2018, doi: 10.1016/J.COESH.2017.10.002.

[14] T. Reza, Z. H. Mohamad Riza, S. R. Sheikh Abdullah, H. Abu Hasan, N. ‘Izzati Ismail, and A. R. Othman, “Microplastic Removal in Wastewater Treatment Plants (WWTPs) by Natural Coagulation: A Literature Review,” *Toxics*, vol. 12, no. 1, Jan. 2024, doi: 10.3390/TOXICS12010012.

[15] I. Sheriff, M. S. Yusoff, and H. B. Halim, “Microplastics in wastewater treatment plants: A review of the occurrence, removal, impact on ecosystem, and abatement measures,” *Journal of Water Process Engineering*, vol. 54, p. 104039, Aug. 2023, doi: 10.1016/J.JWPE.2023.104039.

[16] N. Tang, X. Liu, and W. Xing, “Microplastics in wastewater treatment plants of Wuhan, Central China: Abundance, removal, and potential source in household wastewater,” *Science of the Total Environment*, vol. 745, p. 141026, 2020, doi: 10.1016/j.scitotenv.2020.141026.

[17] S. Acarer, “Microplastics in wastewater treatment plants: Sources, properties, removal efficiency, removal mechanisms, and interactions with pollutants,” *Water Science and Technology*, vol. 87, no. 3, pp. 685–710, Feb. 2023, doi: 10.2166/WST.2023.022.

[18] R. C. Thompson *et al.*, “Lost at Sea: Where Is All the Plastic?,” *Science (1979)*, vol. 304, no. 5672, p. 838, May 2004, doi: 10.1126/SCIENCE.1094559/SUPPL_FILE/THOMPSON.SOM.PDF.

[19] N. O. and A. A. US Department of Commerce, “What are microplastics?”.

[20] Z. Akdogan and B. Guven, “Microplastics in the environment: A critical review of current understanding and identification of future research needs,” *Environmental Pollution*, vol. 254, p. 113011, Nov. 2019, doi: 10.1016/J.ENVPOL.2019.113011.

[21] A. I. Osman *et al.*, “Microplastic sources, formation, toxicity and remediation: a review,” *Environ Chem Lett*, vol. 21, no. 4, p. 1, Aug. 2023, doi: 10.1007/S10311-023-01593-3.

[22] A. I. Osman *et al.*, “Microplastic sources, formation, toxicity and remediation: a review,” *Environ Chem Lett*, vol. 21, no. 4, p. 1, Aug. 2023, doi: 10.1007/S10311-023-01593-3.

[23] H. Sheikhi and R. Mirzaei, “Occurrence and abundance of macro, meso and microplastics along the coasts of the Persian Gulf (case study: Bushehr Province coast),” *Mar Pollut Bull*, vol. 194, p. 115261, Sep. 2023, doi: 10.1016/J.MARPOLBUL.2023.115261.

[24] H. Ma, L. Chao, H. Wan, and Q. Zhu, “Microplastic Pollution in Water Systems: Characteristics and Control Methods,” *Diversity 2024, Vol. 16, Page 70*, vol. 16, no. 1, p. 70, Jan. 2024, doi: 10.3390/D16010070.

[25] D. Saad and D. Saad, “Why Microplastics Are Exceptional Contaminants?,” Mar. 2023, doi: 10.5772/INTECHOPEN.109173.

[26] M. S. Bank and S. V. Hansson, “The Plastic Cycle: A Novel and Holistic Paradigm for the Anthropocene,” *Environ Sci Technol*, vol. 53, no. 13, pp. 7177–7179, Jul. 2019, doi: 10.1021/ACS.EST.9B02942.

[27] Z. Qaiser *et al.*, “Microplastics in wastewaters and their potential effects on aquatic and terrestrial biota,” *Case Studies in Chemical and Environmental Engineering*, vol. 8, p. 100536, Dec. 2023, doi: 10.1016/J.CSCEE.2023.100536.

[28] L. Peng, D. Fu, H. Qi, C. Q. Lan, H. Yu, and C. Ge, “Micro- and nano-plastics in marine environment: Source, distribution and threats — A review,” *Science of The Total Environment*, vol. 698, p. 134254, Jan. 2020, doi: 10.1016/J.SCITOTENV.2019.134254.

[29] M. Eriksen *et al.*, “A growing plastic smog, now estimated to be over 170 trillion plastic particles afloat in the world’s oceans—Urgent solutions required,” *PLoS One*, vol. 18, no. 3, p. e0281596, Mar. 2023, doi: 10.1371/JOURNAL.PONE.0281596.

[30] C. Scherer *et al.*, “Comparative assessment of microplastics in water and sediment of a large European river,” *Science of The Total Environment*, vol. 738, p. 139866, Oct. 2020, doi: 10.1016/J.SCITOTENV.2020.139866.

[31] I. Narloch, A. Gackowska, and G. Wejnerowska, “Microplastic in the Baltic Sea: A review of distribution processes, sources, analysis methods and regulatory policies,” *Environ Pollut*, vol. 315, Dec. 2022, doi: 10.1016/J.ENVPOL.2022.120453.

[32] M. Sajjad *et al.*, “Microplastics in the soil environment: A critical review,” *Environ Technol Innov*, vol. 27, p. 102408, Aug. 2022, doi: 10.1016/J.ETI.2022.102408.

[33] L. Nizzetto, M. Futter, and S. Langaas, “Are Agricultural Soils Dumps for Microplastics of Urban Origin?,” *Environ Sci Technol*, vol. 50, no. 20, pp. 10777–10779, Oct. 2016, doi: 10.1021/ACS.EST.6B04140/ASSET/IMAGES/LARGE/ES-2016-04140A_0001.JPEG.

[34] J. Lofty, V. Muhawenimana, C. A. M. E. Wilson, and P. Ouro, “Microplastics removal from a primary settler tank in a wastewater treatment plant and estimations of contamination onto European agricultural land via sewage sludge recycling,” *Environmental Pollution*, vol. 304, p. 119198, Jul. 2022, doi: 10.1016/J.ENVPOL.2022.119198.

[35] X. Yao, X. S. Luo, J. Fan, T. Zhang, H. Li, and Y. Wei, “Ecological and human health risks of atmospheric microplastics (MPs): a review,” *Environmental Science: Atmospheres*, vol. 2, no. 5, pp. 921–942, Sep. 2022, doi: 10.1039/D2EA00041E.

[36] P. Rocha-Santos, A. Luísa, P. Da Silva, F. Haque, and C. Fan, “Fate and Impacts of Microplastics in the Environment: Hydrosphere, Pedosphere, and Atmosphere,” *Environments 2023, Vol. 10, Page 70*, vol. 10, no. 5, p. 70, Apr. 2023, doi: 10.3390/ENVIRONMENTS10050070.

[37] M. Ahmad *et al.*, “Sources, analysis, and health implications of atmospheric microplastics,” *Emerg Contam*, vol. 9, no. 3, p. 100233, Sep. 2023, doi: 10.1016/J.EMCON.2023.100233.

[38] M. Klein and E. K. Fischer, “Microplastic abundance in atmospheric deposition within the Metropolitan area of Hamburg, Germany,” *Science of The Total Environment*, vol. 685, pp. 96–103, Oct. 2019, doi: 10.1016/J.SCITOTENV.2019.05.405.

[39] J. C. Prata, J. L. Castro, J. P. da Costa, A. C. Duarte, T. Rocha-Santos, and M. Cerqueira, “The importance of contamination control in airborne fibers and microplastic sampling: Experiences from indoor and outdoor air sampling in Aveiro, Portugal,” *Mar Pollut Bull*, vol. 159, p. 111522, Oct. 2020, doi: 10.1016/J.MARPOLBUL.2020.111522.

[40] K. Liu, X. Wang, N. Wei, Z. Song, and D. Li, “Accurate quantification and transport estimation of suspended atmospheric microplastics in megacities: Implications for human health,” *Environ Int*, vol. 132, p. 105127, Nov. 2019, doi: 10.1016/J.ENVINT.2019.105127.

[41] N. Ali *et al.*, “Insight into microplastics in the aquatic ecosystem: Properties, sources, threats and mitigation strategies,” *Science of The Total Environment*, vol. 913, p. 169489, Feb. 2024, doi: 10.1016/J.SCITOTENV.2023.169489.

[42] S. H. Joo, Y. Liang, M. Kim, J. Byun, and H. Choi, “Microplastics with adsorbed contaminants: Mechanisms and Treatment,” *Environmental Challenges*, vol. 3, p. 100042, Apr. 2021, doi: 10.1016/J.ENVC.2021.100042.

[43] K. Ziani *et al.*, “Microplastics: A Real Global Threat for Environment and Food Safety: A State of the Art Review,” *Nutrients*, vol. 15, no. 3, Feb. 2023, doi: 10.3390/NU15030617.

[44] V. Razaviarani, A. Saudagar, S. Gallage, S. Shrinath, and G. Arab, “Comprehensive investigation on microplastics from source to sink,” *Clean Technologies and Environmental Policy 2024*, pp. 1–28, Feb. 2024, doi: 10.1007/S10098-024-02738-W.

[45] S. Saikumar, R. Mani, M. Ganesan, I. Dhinakarasamy, T. Palanisami, and D. Gopal, “Trophic transfer and their impact of microplastics on estuarine food chain model,” *J Hazard Mater*, vol. 464, p. 132927, Feb. 2024, doi: 10.1016/J.JHAZMAT.2023.132927.

[46] S. Mariano, S. Tacconi, M. Fidaleo, M. Rossi, and L. Dini, “Micro and Nanoplastics Identification: Classic Methods and Innovative Detection Techniques,” *Frontiers in Toxicology*, vol. 3, p. 636640, Feb. 2021, doi: 10.3389/FTOX.2021.636640/BIBTEX.

[47] M. A. Browne *et al.*, “Accumulation of microplastic on shorelines woldwide: Sources and sinks,” *Environ Sci Technol*, vol. 45, no. 21, pp. 9175–9179, Nov. 2011, doi: 10.1021/ES201811S.

[48] “What is a WWTP? | Smart Water Magazine.” Accessed: Jan. 11, 2024. [Online]. Available: https://smartwatermagazine.com/q-a/what-a-wwtp

[49] X. Wu *et al.*, “Wastewater treatment plants act as essential sources of microplastic formation in aquatic environments: A critical review,” *Water Res*, vol. 221, p. 118825, Aug. 2022, doi: 10.1016/J.WATRES.2022.118825.

[50] P. J. Anderson, S. Warrack, V. Langen, J. K. Challis, M. L. Hanson, and M. D. Rennie, “Microplastic contamination in Lake Winnipeg, Canada *,” *Environmental Pollution*, vol. 225, pp. 223–231, 2017, doi: 10.1016/j.envpol.2017.02.072.

[51] A. L. Lusher, I. Lise, N. Bråte, K. Munno, R. R. Hurley, and N. A. Welden, “Is It or Isn’t It: The Importance of Visual Classification in Microplastic Characterization”, doi: 10.1177/0003702820930733.

[52] S. Abbasi, “Prevalence and physicochemical characteristics of microplastics in the sediment and water of Hashilan Wetland, a national heritage in NW Iran,” *Environ Technol Innov*, vol. 23, p. 101782, Aug. 2021, doi: 10.1016/J.ETI.2021.101782.

[53] X. Liu, W. Yuan, M. Di, Z. Li, and J. Wang, “Transfer and fate of microplastics during the conventional activated sludge process in one wastewater treatment plant of China,” *Chemical Engineering Journal*, vol. 362, pp. 176–182, Apr. 2019, doi: 10.1016/J.CEJ.2019.01.033.

[54] X. Lv, Q. Dong, Z. Zuo, Y. Liu, X. Huang, and W. M. Wu, “Microplastics in a municipal wastewater treatment plant: Fate, dynamic distribution, removal efficiencies, and control strategies,” *J Clean Prod*, vol. 225, pp. 579–586, Jul. 2019, doi: 10.1016/J.JCLEPRO.2019.03.321.

[55] S. Sarkar, H. Diab, and J. Thompson, “Microplastic Pollution: Chemical Characterization and Impact on Wildlife,” *Int J Environ Res Public Health*, vol. 20, no. 3, Feb. 2023, doi: 10.3390/IJERPH20031745.

[56] K. Ugwu, A. Herrera, and M. Gómez, “Microplastics in marine biota: A review,” *Mar Pollut Bull*, vol. 169, p. 112540, Aug. 2021, doi: 10.1016/J.MARPOLBUL.2021.112540.

[57] X. Ren *et al.*, “Abundance and characteristics of microplastic in sewage sludge: A case study of Yangling, Shaanxi province, China,” *Case Studies in Chemical and Environmental Engineering*, vol. 2, Sep. 2020, doi: 10.1016/J.CSCEE.2020.100050.

[58] J. H. Fernández *et al.*, “Identification and Quantification of Microplastics in Effluents of Wastewater Treatment Plant by Differential Scanning Calorimetry (DSC),” *Sustainability (Switzerland)*, vol. 14, no. 9, p. 4920, May 2022, doi: 10.3390/SU14094920/S1.

[59] A. A. Franco *et al.*, “Assessment and accumulation of microplastics in sewage sludge at wastewater treatment plants located in Cádiz, Spain,” *Environmental Pollution*, vol. 317, p. 120689, Jan. 2023, doi: 10.1016/J.ENVPOL.2022.120689.

[60] L. Zhang, Y. Xie, J. Liu, S. Zhong, Y. Qian, and P. Gao, “An Overlooked Entry Pathway of Microplastics into Agricultural Soils from Application of Sludge-Based Fertilizers,” *Environ Sci Technol*, vol. 54, no. 7, pp. 4248–4255, Apr. 2020, doi: 10.1021/ACS.EST.9B07905/ASSET/IMAGES/LARGE/ES9B07905_0006.JPEG.

[61] S. Ziajahromi, P. A. Neale, I. Telles Silveira, A. Chua, and F. D. L. Leusch, “An audit of microplastic abundance throughout three Australian wastewater treatment plants,” *Chemosphere*, vol. 263, p. 128294, Jan. 2021, doi: 10.1016/J.CHEMOSPHERE.2020.128294.

[62] G. Zhou *et al.*, “Distribution and characteristics of microplastics in urban waters of seven cities in the Tuojiang River basin, China,” *Environ Res*, vol. 189, p. 109893, Oct. 2020, doi: 10.1016/J.ENVRES.2020.109893.

[63] Y. Hu, M. Gong, J. Wang, and A. Bassi, “Current research trends on microplastic pollution from wastewater systems: a critical review,” *Reviews in Environmental*

Science and Bio/Technology 2019 18:2, vol. 18, no. 2, pp. 207–230, Apr. 2019, doi: 10.1007/S11157-019-09498-W.

[64] Z. Chen *et al.*, "Enhanced in situ biodegradation of microplastics in sewage sludge using hyperthermophilic composting technology," *J Hazard Mater*, vol. 384, p. 121271, Feb. 2020, doi: 10.1016/J.JHAZMAT.2019.121271.

[65] Z. Xu and X. Bai, "Microplastic Degradation in Sewage Sludge by Hydrothermal Carbonization: Efficiency and Mechanisms," *Chemosphere*, vol. 297, p. 134203, Jun. 2022, doi: 10.1016/J.CHEMOSPHERE.2022.134203.

[66] X. Ren *et al.*, "Abundance and characteristics of microplastic in sewage sludge: A case study of Yangling, Shaanxi province, China," *Case Studies in Chemical and Environmental Engineering*, vol. 2, p. 100050, Sep. 2020, doi: 10.1016/J.CSCEE.2020.100050.

[67] F. Hassan *et al.*, "Microplastic contamination in sewage sludge: Abundance, characteristics, and impacts on the environment and human health," *Environ Technol Innov*, vol. 31, p. 103176, Aug. 2023, doi: 10.1016/J.ETI.2023.103176.

[68] S. Gündoğdu, C. Çevik, E. Güzel, and S. Kilercioğlu, "Microplastics in municipal wastewater treatment plants in Turkey: a comparison of the influent and secondary effluent concentrations," *Environ Monit Assess*, vol. 190, no. 11, pp. 1–10, Nov. 2018, doi: 10.1007/S10661-018-7010-Y/TABLES/5.

[69] Y. Zou, C. Ye, and Y. Pan, "Abundance and characteristics of microplastics in municipal wastewater treatment plant effluent: a case study of Guangzhou, China," *Environmental Science and Pollution Research*, vol. 28, no. 9, pp. 11572–11585, Mar. 2021, doi: 10.1007/S11356-020-11431-6.

[70] A. Menéndez-Manjón *et al.*, "Long-Term Occurrence and Fate of Microplastics in WWTPs: A Case Study in Southwest Europe," *Applied Sciences (Switzerland)*, vol. 12, no. 4, p. 2133, Feb. 2022, doi: 10.3390/APP12042133/S1.

[71] H. Lee and Y. Kim, "Treatment characteristics of microplastics at biological sewage treatment facilities in Korea," *Mar Pollut Bull*, vol. 137, pp. 1–8, Dec. 2018, doi: 10.1016/J.MARPOLBUL.2018.09.050.

[72] P. J. Ren, M. Dou, C. Wang, G. Q. Li, and R. Jia, "Abundance and removal characteristics of microplastics at a wastewater treatment plant in Zhengzhou," *Environmental Science and Pollution Research*, vol. 27, no. 29, pp. 36295–36305, Oct. 2020, doi: 10.1007/S11356-020-09611-5/FIGURES/4.

[73] M. Kazour, S. Terki, K. Rabhi, S. Jemaa, G. Khalaf, and R. Amara, “Sources of microplastics pollution in the marine environment: Importance of wastewater treatment plant and coastal landfill,” *Mar Pollut Bull*, vol. 146, pp. 608–618, Sep. 2019, doi: 10.1016/J.MARPOLBUL.2019.06.066.

[74] S. A. Carr, J. Liu, and A. G. Tesoro, “Transport and fate of microplastic particles in wastewater treatment plants,” *Water Res*, vol. 91, pp. 174–182, Mar. 2016, doi: 10.1016/J.WATRES.2016.01.002.

[75] F. Murphy, C. Ewins, F. Carbonnier, and B. Quinn, “Wastewater Treatment Works (WwTW) as a Source of Microplastics in the Aquatic Environment,” *Environ Sci Technol*, vol. 50, no. 11, pp. 5800–5808, Jun. 2016, doi: 10.1021/ACS.EST.5B05416/ASSET/IMAGES/LARGE/ES-2015-05416E_0003.JPEG.

[76] H. Hidayaturrahman and T. G. Lee, “A study on characteristics of microplastic in wastewater of South Korea: Identification, quantification, and fate of microplastics during treatment process,” *Mar Pollut Bull*, vol. 146, pp. 696–702, Sep. 2019, doi: 10.1016/J.MARPOLBUL.2019.06.071.

[77] C. M. Rochman *et al.*, “Scientific Evidence Supports a Ban on Microbeads,” *Environ Sci Technol*, vol. 49, no. 18, pp. 10759–10761, Sep. 2015, doi: 10.1021/ACS.EST.5B03909/ASSET/IMAGES/LARGE/ES-2015-03909P_0001.JPEG.

[78] S. Raju *et al.*, “Transport and fate of microplastics in wastewater treatment plants: implications to environmental health,” *Rev Environ Sci Biotechnol*, vol. 17, no. 4, pp. 637–653, Dec. 2018, doi: 10.1007/S11157-018-9480-3/FIGURES/2.

[79] T. H. Nafea, A. J. Al-Maliki, and I. M. Al-Tameemi, “Sources, fate, effects, and analysis of microplastic in wastewater treatment plants: A review,” *Environmental Engineering Research*, vol. 29, no. 1, pp. 230040–0, Apr. 2024, doi: 10.4491/EER.2023.040.

[80] R. Dris *et al.*, “Microplastic contamination in an urban area: a case study in Greater Paris,” *Environmental Chemistry*, 2015, doi: 10.1071/EN14167ï.

[81] J. Talvitie, A. Mikola, O. Setälä, M. Heinonen, and A. Koistinen, “How well is microlitter purified from wastewater? – A detailed study on the stepwise removal of microlitter in a tertiary level wastewater treatment plant,” *Water Res*, vol. 109, pp. 164–172, Feb. 2017, doi: 10.1016/J.WATRES.2016.11.046.

[82] H. J. Kwon, H. Hidayaturrahman, S. G. Peera, and T. G. Lee, “Elimination of Microplastics at Different Stages in Wastewater Treatment Plants,” *Water (Switzerland)*, vol. 14, no. 15, p. 2404, Aug. 2022, doi: 10.3390/W14152404/S1.

[83] “Secondary treatment of Wastewater - Aerobic and Anaerobic Processes.” Accessed: Jan. 17, 2024. [Online]. Available: https://testbook.com/civil-engineering/secondary-treatment-of-wastewater#

[84] M. R. Michielssen, E. R. Michielssen, J. Ni, and M. B. Duhaime, “Fate of microplastics and other small anthropogenic litter (SAL) in wastewater treatment plants depends on unit processes employed,” *Environ Sci (Camb)*, vol. 2, no. 6, pp. 1064–1073, Nov. 2016, doi: 10.1039/C6EW00207B.

[85] K. Magnusson and F. Norén, “Screening of microplastic particles in and down-stream a wastewater treatment plant,” 2014, Accessed: Jan. 17, 2024. [Online]. Available: www.ivl.se

[86] C. Edo, M. González-Pleiter, F. Leganés, F. Fernández-Piñas, and R. Rosal, “Fate of microplastics in wastewater treatment plants and their environmental dispersion with effluent and sludge,” *Environmental Pollution*, vol. 259, Apr. 2020, doi: 10.1016/J.ENVPOL.2019.113837.

[87] S. S. Alavian Petroody, S. H. Hashemi, and C. A. M. van Gestel, “Factors affecting microplastic retention and emission by a wastewater treatment plant on the southern coast of Caspian Sea,” *Chemosphere*, vol. 261, Dec. 2020, doi: 10.1016/J.CHEMOSPHERE.2020.128179.

[88] J. Talvitie *et al.*, “Do wastewater treatment plants act as a potential point source of microplastics? Preliminary study in the coastal Gulf of Finland, Baltic Sea,” *Water Science and Technology*, vol. 72, no. 9, pp. 1495–1504, Nov. 2015, doi: 10.2166/WST.2015.360.

[89] N. Hongprasith, C. Kittimethawong, R. Lertluksanaporn, T. Eamchotchawalit, S. Kittipongvises, and J. Lohwacharin, “IR microspectroscopic identification of microplastics in municipal wastewater treatment plants,” *Environmental Science and Pollution Research*, vol. 27, no. 15, pp. 18557–18564, May 2020, doi: 10.1007/S11356-020-08265-7.

[90] F. Yuan, H. Zhao, H. Sun, J. Zhao, and Y. Sun, “Abundance, morphology, and removal efficiency of microplastics in two wastewater treatment plants in Nanjing, China,” *Environmental Science and Pollution Research*, vol. 28, no. 8, pp. 9327–9337, Feb. 2021, doi: 10.1007/S11356-020-11411-W/FIGURES/9.

[91] J. Talvitie, A. Mikola, A. Koistinen, and O. Setälä, “Solutions to microplastic pollution – Removal of microplastics from wastewater effluent with advanced wastewater treatment technologies,” *Water Res*, vol. 123, pp. 401–407, Oct. 2017, doi: 10.1016/J.WATRES.2017.07.005.

[92] S. Kittipongvises, A. Phetrak, N. Hongprasith, and J. Lohwacharin, “Unravelling capability of municipal wastewater treatment plant in Thailand for microplastics: Effects of seasonality on detection, fate and transport,” *J Environ Manage*, vol. 302, Jan. 2022, doi: 10.1016/J.JENVMAN.2021.113990.

[93] W. Liu *et al.*, “A review of the removal of microplastics in global wastewater treatment plants: Characteristics and mechanisms,” *Environ Int*, vol. 146, p. 106277, Jan. 2021, doi: 10.1016/J.ENVINT.2020.106277.

[94] A. Y. Ayankunle *et al.*, “Estimating Microplastics related to Laundry Wash and Personal Care Products released to Wastewater in Major Estonian Cities: a comparison of calculated and measured microplastics,” *J Environ Health Sci Eng*, vol. 21, no. 1, pp. 225–237, Jun. 2023, doi: 10.1007/S40201-023-00856-Z.

[95] Y. Cai, T. Yang, D. M. Mitrano, M. Heuberger, R. Hufenus, and B. Nowack, “Systematic Study of Microplastic Fiber Release from 12 Different Polyester Textiles during Washing,” *Environ Sci Technol*, vol. 54, no. 8, pp. 4847–4855, Apr. 2020, doi: 10.1021/ACS.EST.9B07395/ASSET/IMAGES/MEDIUM/ES9B07395_M001.GIF.

[96] A. Y. Ayankunle, N. Buhhalko, A. Lukjanova, K. Pachel, E. Lember, and M. Heinlaan, “Microplastics in Influents and Effluents of Estonian Wastewater Treatment Plants,” p. 55, Nov. 2023, doi: 10.3390/PROCEEDINGS2023092055.

[97] “REPORT on the proposal for a directive of the European Parliament and of the Council concerning urban wastewater treatment (recast) | A9-0276/2023 | European Parliament.” Accessed: May 17, 2024. [Online]. Available: https://www.europarl.europa.eu/doceo/document/A-9-2023-0276_EN.html

[98] M. S. M. Al-Azzawi *et al.*, “Validation of Sample Preparation Methods for Microplastic Analysis in Wastewater Matrices—Reproducibility and Standardization,” *Water 2020, Vol. 12, Page 2445*, vol. 12, no. 9, p. 2445, Aug. 2020, doi: 10.3390/W12092445.

[99] A. S. Tagg *et al.*, “Fenton’s reagent for the rapid and efficient isolation of microplastics from wastewater,” *Chemical Communications*, vol. 53, no. 2, pp. 372–375, 2017, doi: 10.1039/C6CC08798A.

[100] I. Schrank *et al.*, “Microplastic sample purification methods - Assessing detrimental effects of purification procedures on specific plastic types,” *Science of The Total Environment*, vol. 833, p. 154824, Aug. 2022, doi: 10.1016/J.SCITOTENV.2022.154824.

[101] “Microplastics Sampling and Processing Guidebook | Mississippi State University Extension Service.” Accessed: Mar. 11, 2024. [Online]. Available: http://extension.msstate.edu/publications/microplastics-sampling-and-processing-guidebook

[102] R. Singh, R. Kumar, and P. Sharma, “Microplastic in the subsurface system: Extraction and characterization from sediments of River Ganga near Patna, Bihar,” *Advances in Remediation Techniques for Polluted Soils and Groundwater*, pp. 191–217, Jan. 2021, doi: 10.1016/B978-0-12-823830-1.00013-4.

[103] C. M. Free, O. P. Jensen, S. A. Mason, M. Eriksen, N. J. Williamson, and B. Boldgiv, “High-levels of microplastic pollution in a large, remote, mountain lake,” *Mar Pollut Bull*, vol. 85, no. 1, pp. 156–163, Aug. 2014, doi: 10.1016/J.MARPOLBUL.2014.06.001.

[104] E. Strady *et al.*, “Baseline assessment of microplastic concentrations in marine and freshwater environments of a developing Southeast Asian country, Viet Nam,” *Mar Pollut Bull*, vol. 162, p. 111870, Jan. 2021, doi: 10.1016/J.MARPOLBUL.2020.111870.

[105] T. M. T. Le *et al.*, “Evaluation of microplastic removal efficiency of wastewater-treatment plants in a developing country, Vietnam,” *Environ Technol Innov*, vol. 29, p. 102994, Feb. 2023, doi: 10.1016/J.ETI.2022.102994.

[106] M. A. Bhat, E. O. Gaga, and K. Gedik, “How can contamination be prevented during laboratory analysis of atmospheric samples for microplastics?,” *Environ Monit Assess*, vol. 196, no. 2, Feb. 2024, doi: 10.1007/S10661-024-12345-3.

[107] X. Xu, Y. Jian, Y. Xue, Q. Hou, and L. P. Wang, “Microplastics in the wastewater treatment plants (WWTPs): Occurrence and removal,” *Chemosphere*, vol. 235, pp. 1089–1096, Nov. 2019, doi: 10.1016/J.CHEMOSPHERE.2019.06.197.

[108] A. Cristaldi *et al.*, “Efficiency of Wastewater Treatment Plants (WWTPs) for Microplastic Removal: A Systematic Review,” *Int J Environ Res Public Health*, vol. 17, no. 21, pp. 1–23, Nov. 2020, doi: 10.3390/IJERPH17218014.

[109] L. Yang, K. Li, S. Cui, Y. Kang, L. An, and K. Lei, “Removal of microplastics in municipal sewage from China’s largest water reclamation plant,” *Water Res*, vol. 155, pp. 175–181, May 2019, doi: 10.1016/J.WATRES.2019.02.046.

[110] E. Vassilenko *et al.*, “Domestic laundry and microfiber pollution: Exploring fiber shedding from consumer apparel textiles,” *PLoS One*, vol. 16, no. 7 July, Jul. 2021, doi: 10.1371/JOURNAL.PONE.0250346.

[111] M. Lares, M. C. Ncibi, M. Sillanpää, and M. Sillanpää, “Occurrence, identification and removal of microplastic particles and fibers in conventional activated sludge process and advanced MBR technology,” *Water Res*, vol. 133, pp. 236–246, Apr. 2018, doi: 10.1016/J.WATRES.2018.01.049.

[112] Y. Cao *et al.*, “Intra-day microplastic variations in wastewater: A case study of a sewage treatment plant in Hong Kong,” *Mar Pollut Bull*, vol. 160, p. 111535, Nov. 2020, doi: 10.1016/J.MARPOLBUL.2020.111535.

[113] J. Sun, X. Dai, Q. Wang, M. C. M. van Loosdrecht, and B. J. Ni, “Microplastics in wastewater treatment plants: Detection, occurrence and removal,” *Water Res*, vol. 152, pp. 21–37, Apr. 2019, doi: 10.1016/J.WATRES.2018.12.050.

[114] W. Shan *et al.*, “Distribution, characteristics and daily fluctuations of microplastics throughout wastewater treatment plants with mixed domestic—industrial influents in Wuxi City, China,” *Front Environ Sci Eng*, vol. 16, no. 1, pp. 1–9, Jan. 2022, doi: 10.1007/S11783-021-1440-4/METRICS.

[115] I. Uogintė, S. Pleskytė, J. Pauraitė, and G. Lujanienė, “Seasonal variation and complex analysis of microplastic distribution in different WWTP treatment stages in Lithuania,” *Environ Monit Assess*, vol. 194, no. 11, pp. 1–18, Nov. 2022, doi: 10.1007/S10661-022-10478-X/FIGURES/7.

[116] M. R. Sanz and J. R. B. Rodríguez, “Abundance and Characterization of Anthropogenic Microlitter in Effluent from Three Wastewater Treatment Plants in Gran Canaria (Canary Islands, Spain),” *Water (Switzerland)*, vol. 16, no. 1, p. 64, Jan. 2024, doi: 10.3390/W16010064/S1.

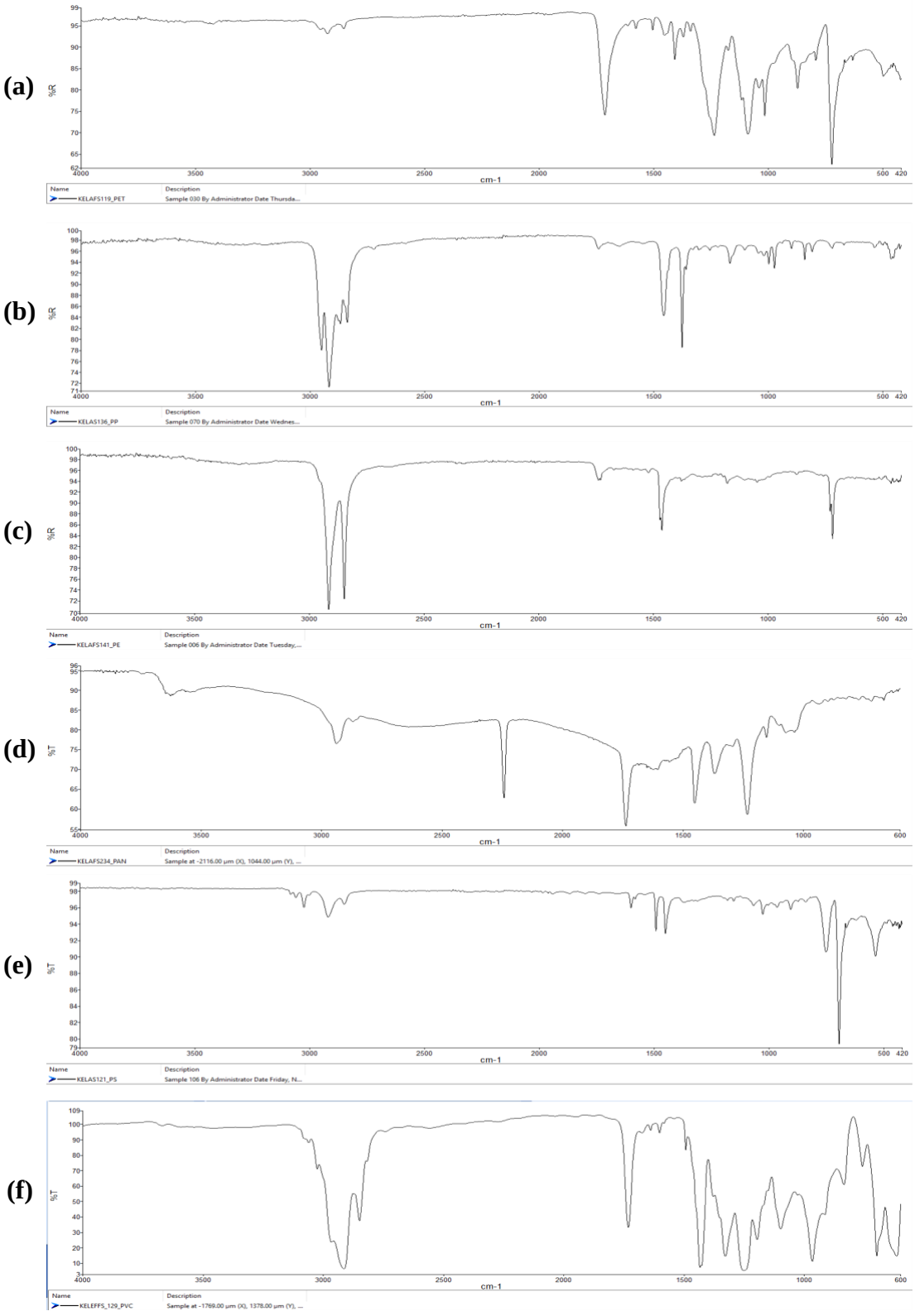
[117] B. Ma, W. Xue, C. Hu, H. Liu, J. Qu, and L. Li, “Characteristics of microplastic removal via coagulation and ultrafiltration during drinking water treatment,” *Chemical Engineering Journal*, vol. 359, pp. 159–167, Mar. 2019, doi: 10.1016/J.CEJ.2018.11.155.

[118] R. Z. Habib, T. Thiemann, and R. Al Kendi, “Microplastics and Wastewater Treatment Plants—A Review,” *J Water Resour Prot*, vol. 12, no. 01, pp. 1–35, Jan. 2020, doi: 10.4236/JWARP.2020.121001.

[119] M. Van Do, T. X. T. Le, N. D. Vu, and T. T. Dang, “Distribution and occurrence of microplastics in wastewater treatment plants,” *Environ Technol Innov*, vol. 26, p. 102286, May 2022, doi: 10.1016/J.ETI.2022.102286.

[120] S. Ziajahromi, P. A. Neale, L. Rintoul, and F. D. L. Leusch, “Wastewater treatment plants as a pathway for microplastics: Development of a new approach to sample wastewater-based microplastics,” *Water Res*, vol. 112, pp. 93–99, Apr. 2017, doi: 10.1016/J.WATRES.2017.01.042.

APPENDIX I



Spectra of PET (a), PP (b), PE (c), PAN (d), PS (e), and PVC (f) polymers observed from the FTIR analysis of visually identified MPs.