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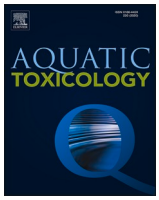

Microplastics in Estonian wastewater treatment plants: First evaluation of baseline concentrations and stage-wise removal efficiency

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ABSTRACT

Wastewater treatment plants (WWTPs) are important pathways of microplastics (MPs) into the environment. To date, the extent of MPs contamination from Estonian WWTPs, located at the Baltic Sea, is not known. To establish MPs baseline levels in the Estonian wastewater treatment system and evaluate MPs removal efficiency, six WWTPs were selected for evaluation. From each plant, 24 h composite samples were collected from raw influent, after primary treatment, and from secondary effluent using an automated sampler with a three-layered sieve system. Upon Fenton-H_2O_2 digestion of organic matter, $\geq$ 300 μm MPs were quantified by microscope and categorized by size, shape and color. At least 50 % of microscopically identified MPs were analyzed by μFTIR, identifying at least 78 % of these as artificial polymers.

The results showed that MPs concentrations in the WWTPs' influents were 205 – 520 MPs/L of which 36 – 94 % was removed during mechanical treatment. As a result of the overall MPs removal efficiency of 99.6 – 99.8 % compared to the influent, 0.5 – 1.4 MPs/L was quantified in the final effluent of the WWTPs. Fibers, fragments and films were recorded in the influent whereas the effluents were dominated by fibers. Fragments and films were mainly composed of polypropylene (PP) and -ethylene, while fibers had more diverse polymeric compositions incl. PP, polyethylene terephthalate and polyacrylonitrile. Despite high MPs removal rates, in total, the six studied WWTP discharge about 9,7E+07 MPs/day in the environment. The obtained results are significant for future regulatory and research endeavors.

1. Introduction

The widespread and increasing use of plastic products has led to an exponential increase and accumulation of plastics in the environment (Geyer et al., 2017), which poses a substantial threat to global ecosystems (Zhou et al., 2020). Research shows that wastewater treatment plants (WWTPs) are a significant pathway of the emerging contaminant, microplastics (MPs; $\leq$ 5 mm) into waterbodies (Arthur et al., 2009; Jagadeesh and Sundaram, 2021). The concern over MPs pollution is reflected in the recently adopted revision of the outdated Urban Wastewater Treatment Directive (UWWTD 91/271) which mandates that WWTPs in the EU start monitoring MPs, especially in the effluent discharges. This requirement will be phased in, starting with WWTPs serving >1E+05 population equivalents (p.e.) by 2035 and extending to those serving 1E+04 - 1E+05 p.e. by 2045 where elevated concentrations of micropollutants incl MPs pose a risk to the human health or the environment (EU Directive, 2024).

The efficiency of MPs removal is largely dependent on the wastewater treatment processes and type of technology deployed (Wu et al., 2021; Kurt et al., 2022; Nandakumar et al., 2022). In conventional WWTPs, MPs are mechanically removed with the skimming of fat and oil, while high density MPs are prone to co-sedimentation with other solids in the primary clarification tanks, leading to MPs removal efficiency of 78 – 98 % during the primary treatment processes (Carr et al., 2016; Murphy et al., 2016; Liu et al., 2019). Since MPs are hardly degradable (Amanna et al., 2023), they sink during the biological treatment and are consequently removed with the waste sludge, where the removal efficiency (7 – 20 %) at this stage depends on e.g. sludge age, sludge volume index, operator's competency and the choice of flocculant for sludge dewatering (Korgmaa et al., 2020; Frehland et al.,

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2020; Kurt et al., 2022). Tertiary treatment has shown to have limited removal efficiency for MPs but compliments the treatment process (Talvitie et al., 2017; Simon et al., 2019). Not only the WWTPs' specificity but MPs' properties such as size and shape are also affecting their removal efficiency in WWTPs. Larger MPs (> 500 μm) are easily removed and separated with the other solids, while smaller MPs particles (≤ 500 μm) and microfibers elude the treatment processes and dominate the MPs load of the WWTPs effluent discharge (Lares et al., 2018; Simon et al., 2019; Vardar et al., 2021). Understanding characteristics of MPs, including their transformation during the treatment process, was crucial for their effective removal in conventional WWTPs.

Despite the high (> 98 %) overall removal efficiency in WWTPs, MPs concentrations in effluents discharged into surface waters have been shown to reach 4.6E+08 MPs/day (Ziajahromi et al., 2021). The Baltic Sea is a vulnerable waterbody that has been shown to be significantly affected by the plastic pollution with MPs concentrations reaching 3.3E+03 MP/m^3 in the water and 1.1E+03 MPs/m^2 in sediments (Narloch et al., 2022). A 5-year monitoring study in the Eastern Baltic found a strong correlation between higher MP loads and sampling closeness to WWTP discharge areas, indicating that WWTPs could not efficiently retain the MPs and prevent their release into the waterbodies (Mishra et al., 2022). However, for Estonia, situated at the Gulf of Finland in the Eastern Baltic, MPs discharge from WWTPs has been unknown until now.

This study aims to quantify MPs ≥ 300 μm discharge from six WWTPs in Estonia, bordered by the Baltic Sea. For that, MPs loads in the influent, after primary screen and in the effluent of the WWTPs were quantified and the MPs' removal efficiency calculated. The new data not only assures early preparedness to comply with the revised wastewater directive but fills the gap in the plastic pollution mapping.

2. Material and methods

2.1. Microplastics sample collection

Six wastewater treatment plants (WWTPs) were chosen for the study and sampled June-August 2023. The selected WWTPs operate based on the plug flow suspended activated sludge principle with subsequent anaerobic, anoxic and aerobic zones, incorporating multiple treatment steps for mechanical and biological treatment (Table 1, Fig. 1). Additional information on the wastewater (WW) treatment process operation is available in Supplementary Information (SI). 24 h composite samples were collected from three sampling points in the WW treatment process (influent S1, after primary treatment S2 and effluent S3) (Fig. 1) using ISCO 3700 automatic sampler equipped with custom-made three-layered metal sieves (pore size: 5000 μm, 400 μm and 100 μm; diameter: 2 mm). A 16 m Teflon tube was connected to the suction port at the top of the sampler and another 2 m hose connected to the bottom for immediate discharge of the filtrate and redirected to the flowing effluent stream. 10 L, 48 L and 1440 L of water was simultaneously sampled from S1, S2 and S3, respectively (Fig. 1) following the sampling volume strategy by Hermsen et al. (2018), Koelmans et al. (2019). The retentate on the 400 μm and 100 μm metal sieves was carefully rinsed, including the edges from the backside of the sieve to one side of the sieve with sufficient volume of ultrapure (Type 1) Milli-Q water (Merck Millipore, Germany) in a plastic 500 ml squeeze bottle until metal sieves were visually clean. Finally, both sieves were scrapped with spatula into 300 ml glass beaker. To prevent excess dilution, the retentate and Milli-Q were kept at the level of 150 ml for the influent and after primary treatment sample while the final effluent with lesser organic matter was kept at 50 ml (Becucci et al., 2022), covered with aluminum foil, stored at 4 °C, and transported to the laboratory.

2.2. Sample treatment for microplastics analysis

All the samples were subjected to oxidation of the organics, following the procedure by Al-Azzawi et al. (2020). First, samples were vortexed (Vortex-Genie-2) for 1 min and three parallel sub-samples (2 mL each) were added in conical flasks using a plastic pipette. Fenton reagent (20 mL of 30 % H_2O_2 and 10 mL of Fe_2SO_4 x $7H_2O$ at 1 g/L) was introduced to each 2 ml sub-sample. An additional 5 mL of 30 % H_2O_2 was added every minute for ten minutes. The mixture was then allowed to cool for 20 min, after which 4 mL of concentrated H_2SO_4 (95–97 %, Merck) was added to dissolve iron precipitates and 5 mL of 1 % Tween (Tween 20 for synthesis (polyoxyethylensorbitan monolaurate, Merck) was added to prevent MP adhesion. The supernatant was passed through a 10 μm metal filter using vacuum-filtration and rinsed off the corrosive chemical with Milli-Q water and finally rinsed onto polycarbonate filters (Cytiva, Cyclopore™; 10 μm, diameter 47 mm) after which the sample retained on the filter was placed in a lidded plastic Petri dish pre-cleaned with acetone and air-dried before analysis. Digested samples passed Fenton - H_2O_2 treatment to break down organic matter. In the text, we used the term "undigested" only to refer to organic matter that was still distinguishable in the otherwise digested sample. The effect of sample treatment on polymer characteristics was not evaluated since no spectral damage on seven polymer types (PS, PE, PET, PP, PVC, PA, PLA) was detected in the original paper (Al-Azzawi et al., 2020).

2.3. Microplastics identification and characterization

2.3.1. Optical microscopy

The samples on Cyclopore membrane filters in Petri dishes were placed on colored paper divided into columns and four distinct quadrants (Figure S3) and examined in Nikon stereomicroscope SMZ1270. Abundance, size, color, and shape of the MPs in each sample was documented using NIS Elements BR software®. The filter was systematically moved vertically from the middle to the top and horizontally from left to right within each column, starting from quadrant 1 through 4 (Figure S3). During visualization, all the identified particles were counted, photographed, and recorded (Table S9). To account for potential counting errors, pristine plastic particles were randomly selected, and the number of MPs on membrane Cyclopore filters was counted by

Table 1
Characteristics of the studied wastewater treatment plants A, B1, B2, B3, B4, and C.

Features	A	B1	B2	B3	B4	C
Population Equivalents	454 000	80 000	38 284	33 499	18 233	4 400
Flow rate (m^3/day)	120 000	12 500	16 300	13 000	1 746	700
Mechanical Treatment Process	Belt Screen SGR Primary Clarifier	Step screen SGR Primary Clarifier	Mono Screen SGR	Step Screen SGR Sand filter	Lamella screen SGR	Screw screen SGR
Biological Treatment Process	A/A/O	A/A/O	A/A/O	A/A/O	A/A/O	A/O
Rainfall Status During sampling	Yes	No	No	No	No	No

SGR - Sand Grit Removal.

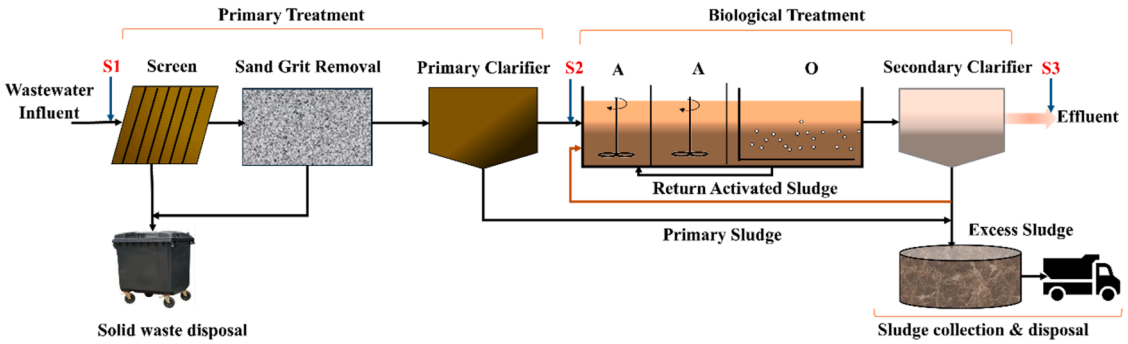

Fig. 1. Description of the wastewater treatment process indicating the microplastic sampling points. Influent sampling point (S1), after-primary treatment sampling point (S2) and effluent sampling point (S3). A - Anaerobic, A - Anoxic, O - Aerobic.

three ($n = 3$) experimenters. The results showed negligible differences in counting MPs ≥ 300 μm. MPs were classified into four shape categories: fragments, fibers, films, and foams. Under the stereomicroscope, fragments appeared as irregularly shaped particles with hardness and luster, films as thin, flat, flexible shapes, and foams as lightweight, porous, sponge-like structures. MP fibers are thread-like long chains with a consistent length, were classified by helicity and cross-sectional shape (e.g., round, oval, flat) and fiber ends (e.g., clear cut, tapered, frayed) (Lares et al., 2018).

2.3.2. μFTIR-IR spectroscopy

In this study, 50 - 78 % of the visually identified plastic particulates were chemically characterized using micro-Fourier Transform Infrared Spectroscopy (μFTIR-IR) with a PerkinElmer Spectrum Spotlight 400 (Table 2). The digested samples (prepared for analysis as described in chapter 2.2) were analyzed in point mode with an MCT detector. MP particles were physically picked up with tweezers and transfered by hand to the compression cell (Specac, DC-3). During the process, some particles were lost, while others were too small to be transferred onto the compression cell. The spectrometer scanned a range from 4000 to 420 cm^{-1}, encompassing the infrared spectrum necessary for identifying polymer-specific functional groups. Particle spectra with a quality index (QI) > 85 % when compared to a reference spectral library, were classified as polymers. Only particles, which were visually identified to clearly consist of organic or other non-plastic compounds, such as algae, sand and glass, were excluded. Some MPs (identified by microscopy) were not subjected to μFTIR analysis due to technical reasons. After analysis with SpectrumIMAGE software, each spectrum was reviewed and verified by an expert. Only spectra with correctly matched peaks were accepted as valid matches

2.4. Contamination control

Throughout the experiment, maximum attention was given to contamination risks and use of plastic-based materials was limited. For onsite wastewater sample collection, 10 L of tap water was filtered from the tap through the metal sieves device. When 10 L of filtrate was reached, tap was shut and sufficient volume of Milli-Q water was used to rinse the surface of the sieve, including the edges from the backside of the sieve to one side and carefully directed into a glass bottle was kept at 50 ml use as “Blank metal sieve control”. The metal sieves were covered with aluminum foil and placed inside enclosed automatic sampler. At the end of each sampling campaign, the automatic sampler was washed with detergent in warm water and rinsed with 10 L of tap water. Non-shedding raincoat was worn during sampling. Retentates were rinsed with appropriate amount of Milli-Q water into a glass beaker, covered with aluminum foil. During the sample treatment in the laboratory, nitrile gloves and 100 % cotton clothing were worn. Filters and Petri dishes were pre-examined by microscope for visible particles. Plastic Petri dishes were used to store the filters, but they had been rinsed with

Table 2
Share of μFTIR-IR studied and confirmed microplastics (MPs ≥ 300 μm) in microscopy-identified MPs across the different sampling points (S1, S2, S3) and wastewater treatment plants (WWTPs).

Sampling Point	WWTP	Microscope Identified MPs Total number of MPs in 3 technical parallels	μFTIR-IR Studied MPs % of total Microscope Identified MPs	Artificial Polymers* (%)	Organics (%)
Influent (S1)	A	145	73.8	85.0	15.0
	B1	131	61.8	90.1	9.9
	B2	82	52.4	97.7	2.3
	B3	141	63.1	88.8	11.2
	B4	208	68.8	86.7	13.3
	C	127	67.7	86.0	14.0
After Primary Treatment (S2)	A	277	78.3	82.0	18.0
	B1	39	66.7	88.5	11.5
	B2	252	53.6	96.3	3.7
	B3	252	66.7	88.1	11.9
	B4	246	78.5	78.8	21.2
	C	82	53.7	97.7	2.3
Effluent (S3)	A	94	68.1	87.5	12.5
	B1	243	70.0	85.9	14.1
	B2	114	50.0	100.0	0.0
	B3	111	60.4	91.0	9.0
	B4	228	69.3	84.8	15.2
	C	141	49.6	98.6	1.4

* particle spectra with a quality index > 85 % (compared to a reference spectral library), were classified as artificial polymers.

a) ethanol, b) checked for contamination. Contamination (size and shape of contaminant particles) was evaluated in different control samples: (a) airborne contamination during onsite sampling ("Air onsite"); scraping sample from sieve, (n = 1/sampling, total n = 6); (b) blank sample of metal sieve ("Blank MS"); contamination was checked after pre-cleaning the filter system, (n = 1/sampling, total n = 6); (c) airborne contamination during sample preparation and microscoping ("Air Lab"); a virgin Cyclopore filter was kept open next to the workstation in the lab (n = 1/sampling, total n = 6); (d) microplastics in used labware/reagents contamination ("Milli-Q"); sample was processed with 2 ml of MilliQ water, (n = 3/study); (e) contamination on Cyclopore filters ("Virgin filter"); filters were checked under stereomicroscope (40x) for contamination, (n = 1/day, total n = 6).

2.5. Data analysis

The results were analyzed in MS Office Excel (Version 2408) and DATAtab (DATAtab Team, 2024). Datasets were tested for normality of distribution (Kolmogorov-Smirnov test, $p > 0.05$) and for differences in MPs concentration between sampling points and WWTPs, one-way analysis of variances (ANOVA) was performed followed by Bonferroni Post-hoc-Tests. Spearman correlation analysis was performed to examine the relationship between MPs abundance in the influent stream and the population equivalents.

The MPs removal efficiency (RE) for each unit was calculated based on Eq. (1)

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

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 formula was used to estimate both the removal efficiency at each investigated treatment stage and the overall microplastics removal efficiency of the whole plant. Detail calculations steps for analyzed subsample were included in supplementary information.

3. Results and discussion

3.1. Microplastics recovery and contamination

To evaluate the efficiency and impact of sample treatment and account for potential microplastics (MPs) loss, a recovery experiment was conducted using pristine cryo-milled MPs, polypropylene (PP) and high-density polyethylene (HDPE) fragments and cotton fibers (Table S2). For each parallel (n = 3), 80 HDPE fragments, 60 PP fragments and 60 cotton fibers (all ≥ 300 μm being the lower studied particle size limit) were counted and rinsed with 2 ml MQ water into glass beaker. A piece of tissue paper was added to simulate the content of cellulose fibers, and the sample was subjected to the sample treatment protocol. The average recovery rate (n = 3) for cotton microfibers was 91.7 ± 1.7 % and for fragments, 85.6 ± 3.4 % and 77.5 ± 3.8 % for PP and HDPE, respectively, which showed satisfactory quality like (PS = 85.9 ± 3.54 %, 100 μm; PE = 81.5 ± 4.67 %, 200 μm) (Luo et al., 2023) and (PP = 97 %, 520 μm) (Salmi et al., 2021) to proceed with the study. Details are available in SI (Table S2).

The number of recorded particulate contaminants (≥ 300 μm) ranged from 0 to 6 particles/filter (Fig. 2), which was comparable to reported zero contaminant (Luo et al., 2023) and in the range (3.5 – 8 MPs) of blank control samples (Conley et al., 2019). In the current study, fibers were the prevalent type of contaminants across all the controls, with the highest counts observed in the blank control for the metal sieve ("Blank MS") used for onsite wastewater sample fractionation. This was followed by airborne contamination in the laboratory ("Air Lab"). No contamination was detected in MilliQ water and on virgin Cyclopore filters. To further reduce contamination, metal sieves were cleaned with air vacuum dryer to remove particles trapped in the sieve joints. In the laboratory, workspace was cleaned thoroughly at least three times prior to the experiment, and movement in the work zones was minimized.

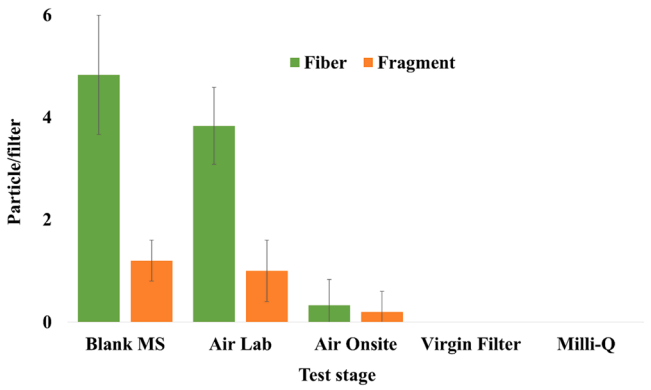

Fig. 2. Particulate contaminants (MPs ≥ 300 μm) at different steps of the study: Blank of metal sieve '**Blank MS'** (n = 1/sampling, total n = 6); Air contamination during sample manipulation in the laboratory **'Air Lab'** (n = 1/sampling, total n = 6); Air contamination during onsite sampling '**Air Onsite**' (n = 1/sampling, total n = 6); Contamination on Cyclopore polycarbonate 10 μm virgin filters '**Virgin filter'**(n = 1/day, total n = 6); lab wear/reagent '**Milli-Q**' (n = 3/study). For more detailed contamination control description, see chapter 2.4. The data are presented as AVG ± SD.

3.2. Concentration of microplastics across treatment stages

Building on previous MPs research (Talvitie et al., 2016; Lares et al., 2018), we aimed at high representativeness in our MPs sampling approach. First, continuous filtration of composite samples was collected from three critical sampling points (influent, after primary treatment, effluent) along the water line over a 24 h period to account for hourly fluctuations in MPs loads (Becucci et al., 2022). Second, we utilized larger sample volumes (10 - 1440 L) and deployed onsite size fractionation. Each of the three samples (S1, S2, S3) collected was analyzed in three technical sub-samples (parallels). All the digested sub-samples were initially examined under a microscope with the exclusion of seemingly undigested organic materials and subsequently subjected to μFTIR analysis. Specifically, 50 % - 78 % of the total visually identified MPs were further analyzed showing a high-level (78 - 100 %) of FTIR-identified artificial polymers amongst microscopically identified MPs (Table 2). Therefore, the reported particle concentrations of MPs in the WWTPs were based on microscopically identified MPs. Particle concentrations of MPs in the WWTPs based on artificial polymers excluding organics (Figure S5, Table S7).

Nine different polymer types were identified for the μFTIR-studied particles, including polyethylene terephthalate (PET), polypropylene (PP), polystyrene (PS), polyethylene (PE), polyvinyl chloride (PVC), polyacrylonitrile (PAN), polyamide (PA), acrylonitrile butadiene styrene (ABS), and polyvinyl acetate (PVA) (Fig. 3, Table S8). The most common polymer types were PET, PE, and PP, which were observed at all the sampling points. PET was the most abundant polymer across all WWTPs and sampling stages, with percentages ranging from 40 % to 88 %. The final effluent of most WWTPs showed an increased percentage of PET, especially in WWTP C, where it reached 85 % in the final effluent. However, in the effluent, MPs (≥ 300 μm) concentrations were just 0.5 - 1.4 MPs/L. PE was prominent in the influent of WWTP A (28 %) but decreased significantly in subsequent stages. The percentage of PE in the final effluent varied among WWTPs, with the lowest percentage observed in WWTP C (4 %) and higher values in WWTP A (46 %). Polymers PVA, PA, and ABS were detected in very small amounts or were absent across all WWTPs and stages. ABS, for instance, was only detected in WWTP A, contributing <2 % in any stage. PP showed significant variation between influent and effluent stages. For example, in

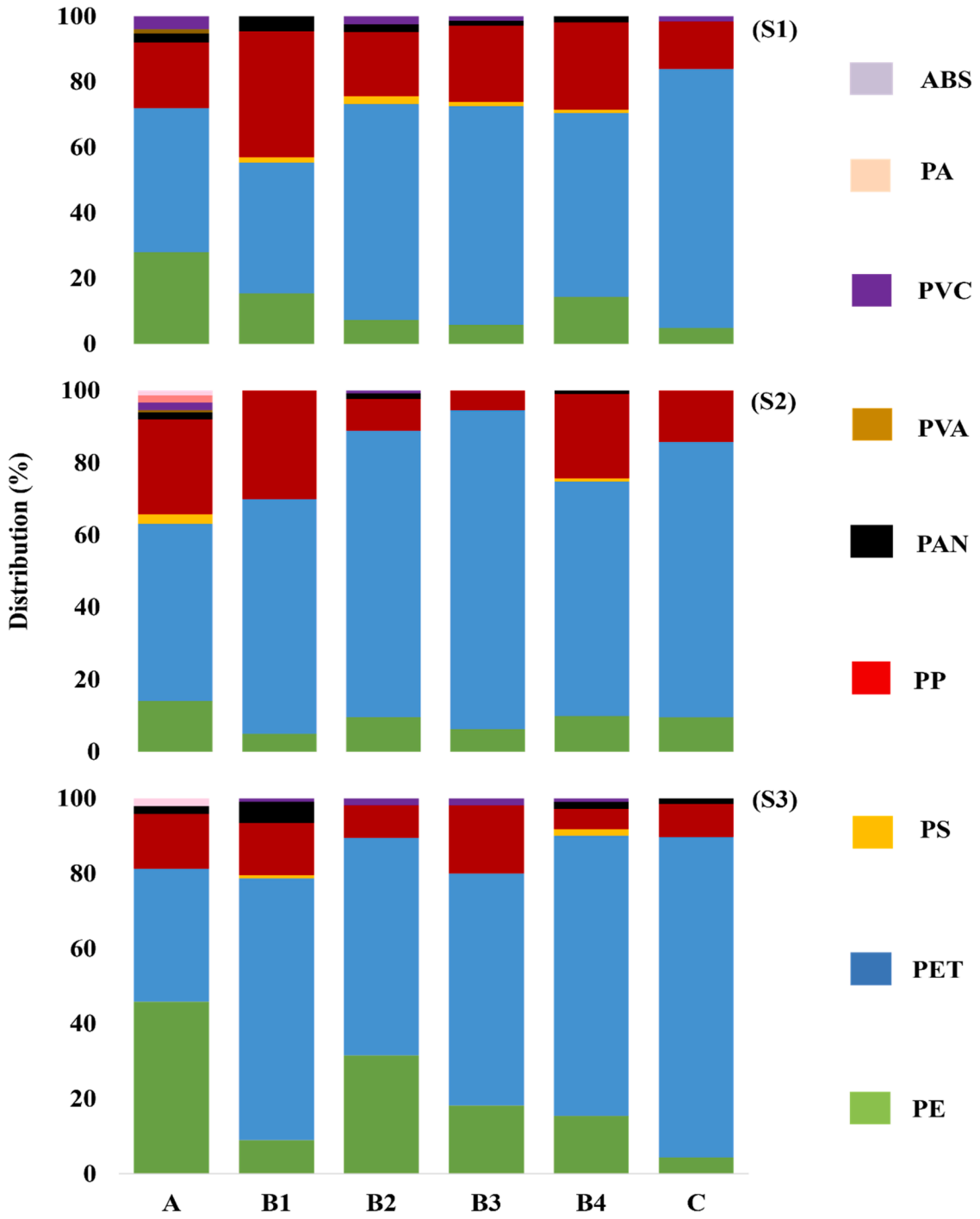

Fig. 3. Artificial polymer types of the μFTIR-analyzed microplastics (MPs ≥ 300 μm) in three sampling points (Influent (S1), After primary treatment (S2), Effluent (S3)) in six Estonian wastewater treatment plants. Average distribution data are presented ($n = 3$).

WWTP A, its proportion increased from 20 % in the influent to 26 % after primary treatment but decreased to 15 % in the final effluent. In the final effluent of the six WWTPs, the abundance of polymer types decreased in the order: PET > PE > PP > PAN > PS > PVC > ABS > PVA > PA.

Differential removal efficiency was observed for various polymers. For instance, the proportion of PET generally increased after primary treatment, possibly due to better retention of other polymer types like PE and PP. WWTP A showed a relatively balanced distribution among different polymer types in the influent compared to other WWTPs. However, WWTP C showed a high proportion of PET in all stages, peaking in the final effluent at 85 %, indicating less efficient removal of this polymer type. The polymer types of PET, PE, PP were also the most abundant in other WWTPs, as reported in Liu et al. (2021) originating from e.g. food packaging, plastic bags, textiles and rubber particles (Kurniawan et al., 2021) The findings of this study emphasize the significance of PET, PE and PP polymers in wastewater streams and their persistence through treatment processes.

The amount of microplastics in WWTP can be influenced by various factors, including the composition of raw wastewater, local lifestyles, population size, and differences in sampling techniques (e.g., grab vs. composite sampling, sampling duration, and sieve mesh size).

Additionally, extraction methods, such as digestion processes and density separation using salt solutions, as well as identification techniques and sampling locations, play a significant role. For instance, the number of MPs detected in samples depend on the pore size of the filters used during analysis. The smaller the filter size used, the greater the number of particles detected (Sun et al., 2019).

Microplastics concentration in the influent (S1) of the six WWTPs ranged from 205 ± 12 to 520 ± 66 MPs/L, with the respective values of 103 ± 9 to 263 ± 34 for FTIR-confirmed MPs (Figure S5; Table S7) with the highest concentrations observed in the influent of B4 and the lowest in the influent of B2 (Fig. 4). There was significant difference ($F = 46.41$, $p < 0.001$) among the concentration of MPs across the sampling points across the studied WWTPs. The average MPs/L in the influent in our study was of the same order of magnitude as reported in the neighboring country Finland (390 - 900 MPs/L; > 20 μm) (Talvitie et al., 2017) and similar to values in an urban WWTP located in Cadiz (Spain) with a concentration of (275 - 586 MP/L; >100 μm) (Franco et al., 2020) with varying 24 h composite sample volumes from the influent. Compared to the influent, MPs concentration after the primary treatment (S2) had reduced to 20.3 ± 8 to 144 ± 37 MPs/L with the lowest in WWTP B1 operating with four primary clarifiers and the highest in WWTP A operating eight primary clarifiers although the higher concentration could be attributed to low hydraulic retention time and impact of rainfall during sampling days. One of the primary factors influencing MPs removal in WWTPs were the initial MPs loads entering the system and the retention time of MPs within the operational units (Kurt et al., 2022).

As expected, MPs concentration in the final effluent was further reduced to 0.5 ± 0 to 1.4 ± 0 MPs/L across the six WWTPs (Fig. 4, Table 3). The highest concentration was recorded in WWTP B1 operating two secondary clarifiers, while the lowest was observed in WWTP A operating twelve secondary clarifiers. In comparison with other studies, MPs concentration in the effluent (1.05 MPs/L; > 250 μm) was similar to Lares et al. (2018).

However, despite the low concentration of MPs detected in the final effluent of the WWTPs and high removal rate (99.6 – 99.8 %), a substantial quantity is still being discharged into waterbodies daily (Table 3). For instance, WWTP A, serving a population of over 100,000, releases approximately 6.53E+04 MPs/day into surface waters. Based on this trend, the annual emission of MPs from WWTP A alone would amount to 23.8E+06 MPs. This accounts for 69 % of the total MPs from the investigated WWTPs in Estonia discharged into waterbodies connected to the Baltic Sea. In addition, factors like high coastal population, high industrialization rate of the region and slow water exchange make the Baltic Sea a very vulnerable ecosystem to (MPs) pollution. According to Narloch et al. (2022), knowledge of MPs input into the Baltic Sea is insufficient and in addition to the primary wastewater input pathway, rainwater and riverine emissions need attention as well.

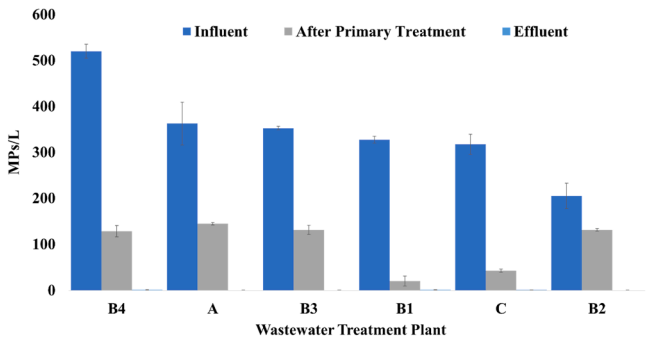

Fig. 4. Microplastics (MPs ≥ 300 μm) concentration (MPs/L) in wastewater systems: in the influent, after primary treatment and in the effluent (sampling locations on Fig. 1). Data are presented as AVG ± SD ($n = 3$).

Table 3
Microplastics (MPs ≥ 300 μm) concentration in the influent, after primary treatment and effluent of wastewater treatment plants, nominal discharge volumes and overall removal efficiency. Microplastics concentration data are presented as AVG ± SD ($n = 3$).

WWTP	Influent (MPs/L)	After PT (MPs/L)	Effluent (MPs/L)	MPs/day (10^3)	MPs/year (10^6)	Removal Efficiency (%)
A	363 ± 46	144 ± 37	0.5 ± 0	65.3	23.8	99.8
B1	328 ± 140	20.3 ± 8	1.4 ± 0	17.6	6.4	99.6
B2	205 ± 12	131 ± 30	0.7 ± 0	10.8	0.4	99.7
B3	353 ± 23	131 ± 32	0.6 ± 0	8.4	3.1	99.8
B4	520 ± 66	128 ± 11	1.3 ± 0	2.3	0.8	99.7
C	318 ± 83	42.7 ± 9	0.8 ± 0	0.6	0.2	99.7

WWTP – wastewater treatment plant; PT – primary treatment.

3.3. Removal of microplastics by size, shape and color

3.3.1. Size

Studied MPs were sub-divided into small, medium and large: 300–499 μm, 500–999 μm, and 1000–5000 μm, respectively (Fig. 5, Table S4). In the influent, WWTPs A, B1, B4, and C were characterized by higher prevalence of small MPs, accounting for over 40 % of the total. In contrast, WWTPs B2 and B3 exhibited a more balanced distribution, with large MPs making up 16 - 32 % of the total. Compared to the influent, small MPs showed a slight decrease across most WWTPs, indicating partial removal during treatment, while medium MPs generally exhibit an increase, likely due to higher retention or re-entry via reject water line within the WWTP system. However, large MPs consistently decreased (12 % – 18 %) across all WWTPs, reflecting effective retention and removal of this size fraction. However, in the final effluent of WWTP A, following biological treatment with activated sludge, 84 % of the MPs discharged into waterbody were small (60 %) and medium. This result highlights WWTP effluent as a major pathway for MPs to enter the open sea. These findings are comparable to earlier studies in Estonian surface waters (Mishra et al., 2022), where 75 % of detected MPs were within the 330–999 μm size range.

The shift towards smaller MPs can be attributed to the finer pore size (100 μm) of the metal sieve used during sampling, consistent with findings from other studies (Sutton et al., 2016). Notably, smaller pore sizes result in higher MP capture efficiency (Becucci et al., 2022). Our results are comparable with other studies that found abundant size fractions to be < 355 μm (Franco et al., 2020) and ranged between 45 – 500 μm (Dronjak et al., 2023). Overall, the results suggest that existing WWTP processes cannot effectively remove MP particles lesser than 1000 μm. However, bigger MPs can break down into smaller size fractions in different treatment units (Cheng et al., 2021) as observed in studied WWTP. Alarmingly, smaller particles (< 500 μm) are easily ingested by plankton and fish (Qiao et al., 2019) and pose a risk for the aquatic environment.

3.3.2. Shape

There was a significant difference ($F = 5.66; p < 0.05$) in the removal rates and shapes ($F = 63.97: p < 0.01$) of different MPs across various stages in all Table S5). Fibers dominated the MPs profile across all WWTPs, followed by fragments, which showed variable trends depending on the treatment plant. Films and foams were consistently the least represented shape in line with (Franco et al., 2020; Uogintė et al., 2022; Bastakoti et al., 2024). As shown in Fig. 5, fibers constituted over 90 % of MPs in both influent and effluent stages of studied WWTPs except for WWTP A with 79 % and 38 % in the influent and effluent

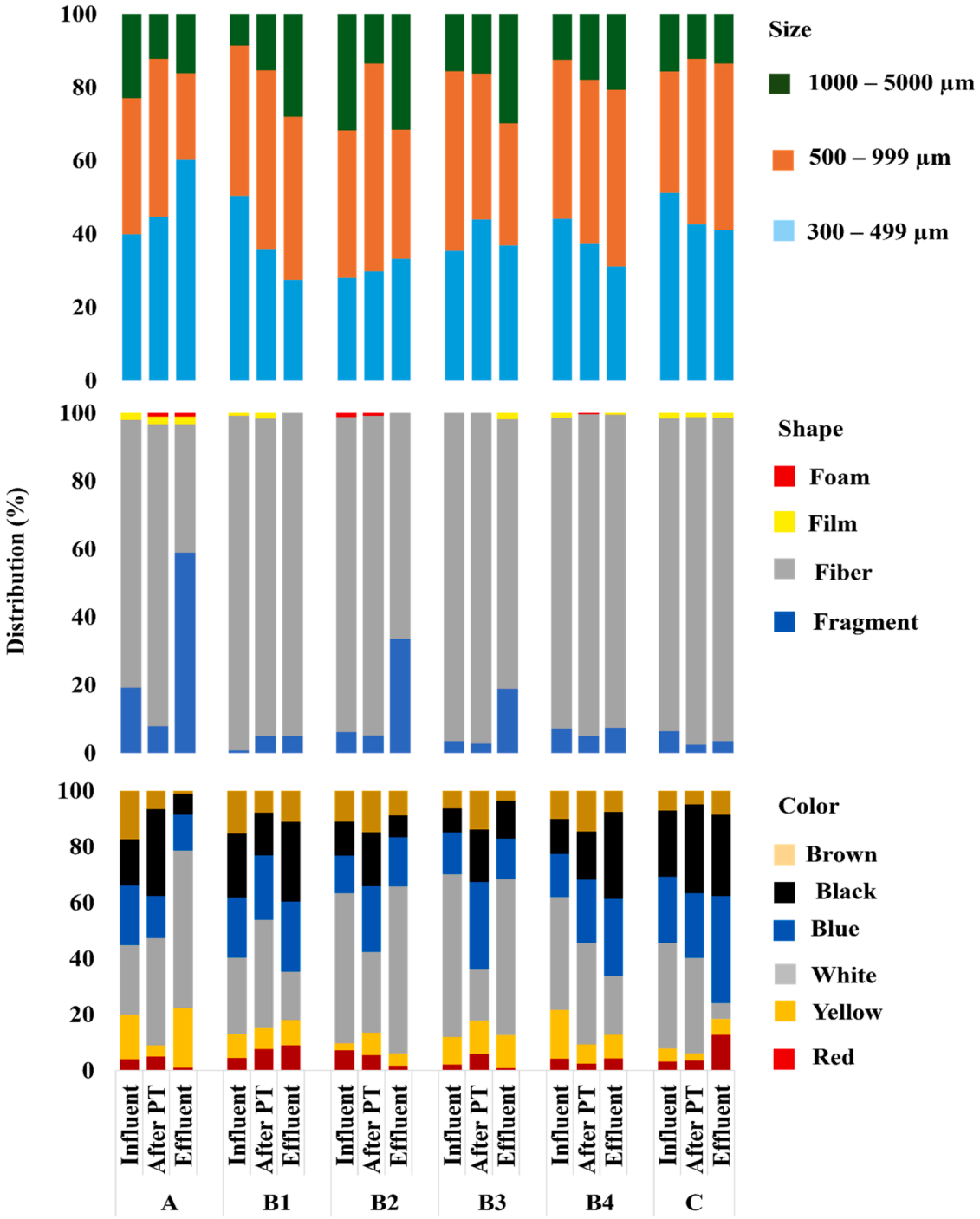

Fig. 5. Distribution of microplastics (MPs ≥ 300 µm) by size, shape and color, in six Estonian wastewater treatment plants across three sampling points. Average distribution data are presented ($n = 3$). After PT - after primary treatment.

stages respectively, highlighting their persistence through treatment processes. There was a major increase in fragment MPs shape in the effluent of WWTP A and B2, where their share increased from 19 % and 6 % in the influent to 59 % and 34 % in the effluent, respectively. This suggests either fragmentation of other MP shapes during treatment or low hydraulic retention efficiency for this MPs shape according to Poerio et al. (2019). Conversely, in other WWTPs (e.g., B1 and B4), the proportion of fragments remained consistently low across all stages, potentially indicating effective retention. Films constituted a minor proportion of the MPs, with no significant changes observed during treatment. Films were nearly absent in most WWTPs (e.g., B2, B3, and B4) and are only marginally present in WWTP A (2 % in both influent and effluent). Foams were rare, with negligible presence across all stages in all WWTPs. For example, WWTP A showed a slight increase from 0 % in the influent to 1 % in the effluent, while other WWTPs (e.g., B3, B4, and C) had no foam shape MPs at any stage. The results obtained in this study are consistent with literature data that reported fibers, fragments, and films the most widely detected MPs in wastewater, and their abundances in the influent within 48 – 66 %, 15 – 42 %, 10 – 19 % and after primary treatment 58 – 90 %, 9 – 16 %, 1 – 26 % respectively (Dronjak et al., 2023). The removal efficiency of MPs by shapes across the treatment stages in Table 4 complements the previously observed

Table 4
Removal efficiency of differently shaped microplastics (MPs ≥ 300 μm) after primary and secondary (biological) treatment. Removal efficiency was based on average values of MPs/L.

	After Primary Treatment				After Secondary Treatment			
	Fragment (%)	Fiber (%)	Film (%)	Foam (%)	Fragment (%)	Fiber (%)	Film (%)	Foam (%)
A	84	55	58	NA	97	100	100	100
B1	60	94	87	NA	93	93	100	NA
B2	46	35	NA	58	97	100	NA	100
B3	71	62	NA	NA	97	100	NA	NA
B4	83	74	100	NA	98	99	NA	100
C	95	86	90	NA	97	98	98	NA

Removal efficiency 0–50 % (red); 51–70 % (yellow); 71–100 % (green); NA- Not Applicable.

trends in percentage distribution and shows the effectiveness of the WWTPs in reducing MPs. During primary treatment, relatively higher percentage (71 – 95 %) of fragments were removed except for WWTP B2 and B1 which achieved moderate removal 46 % and 60 % respectively. Fibers were difficult to remove, half of the investigated WWTPs achieved moderate (35 – 62 %) and higher (74 – 94 %) removal rates. Film and foams were rarely detected and efficiently (> 87 %) removed but WWTP A, B2 achieves 58 % removal during primary treatment. In general, the biological treatment stage demonstrated relatively higher removal efficiency (93 – 100 %) for all MPs shapes in all the studied WWTPs. This higher efficiency was attributed to the ability of the activated sludge flocs (typically negatively charged) to entrap MPs, allowing them to be separated with the excess sludge after sedimentation in the secondary clarifier (Parrella et al., 2025).

3.3.3. Color

Recording of the colors of the MPs enables further insight into the sources of MPs before and after treatment in the WWTPs, as well as to assess potential contamination during sample preparation (Hartmann et al., 2019). However, it should be noted that the Fenton reaction can affect the color of MPs due to the generation of hydroxyl radicals, which can oxidize the surface of MPs, altering their chemical structure and potentially causing discoloration in the plastics (Yang et al., 2022). We categorized the particles into six distinct color types: white, blue, black, brown, yellow, and red (Fig. 5, Table S6). In all the influent samples, white MPs were predominant, ranging from 25 % (WWTP A) to 58 % (WWTP B3) and largely persistent in the effluents of WWTPs A, B2 and B3 up to 60 %. In contrast, WWTP C showed significant reductions of white-colored MPs from 38 % in influent to 6 % in effluent. Brown and red-colored MPs were generally present in lower proportions across the investigated WWTPs. Fragments, films, and foams were predominantly white, whereas fibers were mostly blue, black, brown, yellow, and red. Interestingly, these findings, especially for WWTP A, highly correlated with results from surface water samples MPs color distribution black (30 %), white (23 %), and blue (22 %) in Estonia (Mishra et al., 2022). Similarly, these results were comparable with recent study in Lithuania, where Uogintė et al. (2022) reported color distribution in wastewater samples (11 – 47 %) white, (24 – 73 %) black, (7 – 16 %) transparent while brown, yellow and blue present in relatively low proportion: 2 – 9 %, 2 – 7 % and 1 – 9 % respectively. The color of MPs was a critical factor for their ingestion by sea turtles and other marine species (Noh et al., 2024).

3.4. Factors influencing microplastics removal in wastewater treatment plants

The concentration of MPs in raw wastewater was influenced by several factors, including population density, the degree of urbanization, the level of industrial activity within the service area, stormwater surface run-off in case of combined sewage systems (Ma et al., 2024). The design and operational efficiency of treatment facilities also impact MP emissions in final effluents (Korgmaa, 2020; Hao and Shen, 2021). For instance, membrane bioreactors (MBR) can achieve 99.9 % MPs removal rates from raw influent, but are only economically feasible for larger WWTPs (Lares et al., 2018) whereas dissolved air flotation (DAF) in the primary treatment stage has also proven effective in removing MPs by reducing their densities, allowing them to be separated from the process and preventing MPs accumulation in the sewage sludge (Swart et al., 2022).

The concentration of MPs entering the selected Estonian WWTPs were of the same order of magnitude as those observed in other WWTPs in the Baltic Sea region (Table S3). In this study, variations in catchment areas resulted in a weak positive correlation between the abundance of MPs in wastewater influent and population density. WWTPs B1 to C, which mainly serve residential households connected to a centralized sewer system and some private households with septic tanks whose sewage was transported by truck to the WWTP, exhibited varying concentrations of MPs, predominantly composed of microfibers (91 – 98 %), which was in line with earlier study in Estonia (Ayankunle et al., 2023). Microfiber pollution largely originates from synthetic textiles. During the first wash cycle, the concentration of fibers released from textiles ranges from $1.0E{+}10^5$ to $6.3E{+}10^6$ kg^{-1} (Kärkkäinen and Sillanpää, 2020). Installation of microfiber filters in the effluent pipes of washing machines is a potentially cost-effective “point-source” method to prevent fibers from entering the sewer systems (Erdle et al., 2021).

In contrast, WWTP A, which serves an urban area where all the households were connected to the central sewer system and equipped with several sub-pumping stations with mechanical macro-screens, exhibited a different MPs profile. A substantial proportion of MPs in their influent and effluent were identified as fragments, accounting for 19 % and 59 %, respectively. Increased concentration of fragments in the effluent compared to influent could be attributed to MPs re-entering the process via reject water stream (Salmi et al., 2021), highlighting the need for filtration systems to enhance MP removal and prevent accumulation in sewage sludge, which was a major sink for MPs and a significant source of MPs emissions in case of agricultural sludge valorization (Hu et al., 2024). Similarly, WWTPs B2 and B3 had a higher fraction of fragments in the effluent compared to the influent, suggesting possible in-plant MPs pollution. MPs may originate from facility maintenance activities, such as using plastic-based cleaning tools like scrubbing brushes for washing secondary clarifier overflow weirs or from synthetic polymers used in the mechanical sludge dewatering process. Additionally, recirculation through return activated sludge and reject water lines may further contribute to MPs persistence (Salmi et al., 2021). Installing filtration devices in key treatment process streams could help reduce internal MP contamination.

Conventional WWTPs were not designed to effectively remove MPs and other emerging pollutants, making it challenging to efficiently retain the MPs just with the conventional widespread technical solutions. Therefore, in the case of point-source emissions, controlling MPs

pollution at the source may be a more effective approach. Nevertheless, for diffuse sources of MPs pollution, such as surface run-off from streets and roads, it was challenging to propose economically viable technical solutions, unless the run-off was collected in a stormwater overflow tank. Additionally, preventing MPs accumulation in the waste excess sludge generated at WWTPs will limit secondary contamination when the sludge was composted and subsequently valorized as a fertilizer in agricultural fields (Zhang and Chen, 2020).

While identifying and quantifying MPs in wastewater treatment systems is not new in the field of plastic pollution research, this study provides the first baseline data on MPs in WWTPs in Estonia, a country bordering the highly vulnerable and polluted Baltic Sea. These baseline values and the analysis of MPs removal efficiency were crucial for understanding the local context and planning effective mitigation strategies. The findings of this study contribute to the regional knowledge base, supporting the selection of appropriate technologies to reduce MP emissions from WWTPs. Furthermore, they prepare local authorities to comply with upcoming requirements under the revised Urban Wastewater Treatment Directive (UWWTD 91/271), which includes specific provisions for micropollutants, including MPs. This localized data fills a significant gap and provides actionable insights for environmental policy and management in Estonia.

4. Conclusions

For the first time, concentration and characteristics of microplastics (MPs) in six wastewater treatment plants (WWTPs) in Estonia was systematically investigated and compared. MPs $\geq$ 300 μm) concentration in the WWTPs' influents ranged from 205 to 520 MPs/L, comparable to other data from the region. MPs fragments and films were primarily composed of PP and PE, whereas fibers, that dominated the final effluent, had more diverse compositions, mainly PET, PP, and PAN. The overall WWTP removal efficiency of MPs was 99.6 – 99.8 % with notable variations across the individual treatment stages. In the current study, WWTPs with longer hydraulic retention time in the primary treatment stages were more successful in retaining the MPs while the majority of MPs were accumulated in the activated sludge during the biological treatment phase. Despite the high removal rates, approximately 9,7E+07 MPs/day were collectively discharged from these six WWTPs into waterbodies which are confluent into the Baltic Sea, highlighting their significance as a pathway of MPs into the environment. Further, as an important share of the MPs in the WWTPs' influent was accumulated in the sludge, the practice of sludge valorizing in landscaping and agriculture should be reconsidered due to the potential risk of environmental contamination. This study established the MPs baseline levels in Estonian WWTPs, which is crucial for future regulatory and research efforts but moreover for the development of prevention and control measures to reduce MPs emissions from the wastewater treatment utilities.

CRediT authorship contribution statement

Ayankoya Yemi Ayankunle: Writing – review & editing, Writing – original draft, Visualization, Validation, Supervision, Software, Resources, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Natalja Buhhalko:** Validation, Methodology, Investigation, Data curation. **Karin Pachel:** Supervision, Resources, Project administration, Funding acquisition. **Erki Lember:** Writing – review & editing, Visualization, Supervision, Software, Resources, Project administration, Investigation, Data curation. **Asya Drenkova-Tuhtan:** Writing – review & editing, Investigation. **Margit Heinlaan:** Writing – review & editing, Writing – original draft, Visualization, Validation, Supervision, Software, Resources, Project administration, Methodology, Investigation, Funding acquisition, Formal analysis, Data curation, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.aquatox.2025.107305.

Data availability

Data will be made available on request.

References

Al-Azzawi, M.S.M., Kefer, S., Weißer, J., Reichel, J., Schwaller, C., Glas, K., Knoop, O., Drewes, J.E., 2020. Validation of sample preparation methods for microplastic analysis in wastewater matrices-reproducibility and standardization. Water 12 (9). https://doi.org/10.3390/w12092445.

Amanna, R., Samavi, M., Rakshit, S.K., 2023. Biological degradation of microplastics and nanoplastics in water and wastewater. Curr. Dev. Biotechnol. Bioeng. 293–314. https://doi.org/10.1016/B978-0-323-99908-3.00002-6.

Arthur, C., Baker, J., Bamford, H., 2009. Proceedings of the International Research Workshop on the Occurrence, Effects, and Fate of microplastic marine debris. Group 530. https://doi.org/10.1007/978-3-319-16510-3. *January*.

Ayankunle, A.Y., Buhhalko, N., Pachel, K., Lember, E., Kõrgmaa, V., Mishra, A., Lind, K., 2023. 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. https://doi.org/10.1007/s40201-023-00856-z.

Bastakoti, S., Adhikari, A., Thaiba, B.M., Neupane, B.B., Gautam, B.R., Dangi, M.B., Giri, B., 2024. Characterization and removal of microplastics in the Guheshwori Wastewater Treatment Plant, Nepal. Sci. Total Environ. 935, 173324. https://doi.org/10.1016/J.SCITOTENV.2024.173324.

Becucci, M., Mancini, M., Campo, R., Paris, E., 2022. Microplastics in the Florence wastewater treatment plant studied by a continuous sampling method and Raman spectroscopy: a preliminary investigation. Sci. Total Environ. 808. https://doi.org/10.1016/j.scitotenv.2021.152025.

Carr, S.A., Liu, J., Tesoro, A.G., 2016. Transport and fate of microplastic particles in wastewater treatment plants. Water. Res. 91, 174–182. https://doi.org/10.1016/j.watres.2016.01.002.

Cheng, Y.L., Kim, J.-G., Kim, H.-B., Choi, J.H., Fai Tsang, Y., Baek, K., 2021. Occurrence and removal of microplastics in wastewater treatment plants and drinking water purification facilities: a review. Chem. Eng. J. 410, 128381. https://doi.org/10.1016/j.cej.2020.128381.

Conley, K., Clum, A., Deepe, J., Lane, H., Beckingham, B., 2019. Wastewater treatment plants as a source of microplastics to an urban estuary: removal efficiencies and loading per capita over one year. Water. Res. X 3, 100030. https://doi.org/10.1016/j.wroa.2019.100030.

DATAtab Team. (2024). *Online statistics calculator*. https://datatab.net/statistics-calculator/correlation.

Dronjak, L., Exposito, N., Sierra, J., Schuhmacher, M., Florencio, K., Corzo, B., Rovira, J., 2023. Tracing the fate of microplastic in wastewater treatment plant: a multi-stage analysis of treatment units and sludge. Environ. Pollut. 333, 122072. https://doi.org/10.1016/j.envpol.2023.122072.

Erdle, L.M., Nouri Parto, D., Sweetnam, D., Rochman, C.M., 2021. Washing machine filters reduce microfiber emissions: evidence from a community-scale pilot in Parry Sound, Ontario. Front. Mar. Sci. 8. https://doi.org/10.3389/fmars.2021.777865.

Directive, E.U., 2024. Directive Of The European Parliament and of the Council Concerning Urban Wastewater Treatment (Recast) (Text with EEA Relevance).

Franco, A.A., Arellano, J.M., Albendín, G., Rodríguez-Barroso, R., Zahedi, S., Quiroga, J. M., Coello, M.D., 2020. Mapping microplastics in Cadiz (Spain): occurrence of microplastics in municipal and industrial wastewaters. J. Water Process. Eng. 38 (June), 101596. https://doi.org/10.1016/j.jwpe.2020.101596.

Frehland, S., Kaegi, R., Hufenus, R., Mitrano, D.M., 2020. Long-term assessment of nanoplastic particle and microplastic fiber flux through a pilot wastewater treatment

plant using metal-doped plastics. Water Res. 182. https://doi.org/10.1016/j.watres.2020.115860.
Geyer, R., Jambeck, J.R., Law, K.L., 2017. Production, use, and fate of all plastics ever made. Sci. Adv. 3 (7), 25–29. https://doi.org/10.1126/sciadv.1700782.
Hao, F., Shen, M., 2021. Investigation of operational parameters for retaining properties of micro-plastics for typical aerobic wastewater treatment unit. Chem. Eng. J. 423, 130254. https://doi.org/10.1016/J.CEJ.2021.130254.
Hartmann, N.B., Hüffer, T., Thompson, R.C., Hassellöv, M., Verschoor, A., Daugaard, A. E., Rist, S., Karlsson, T., Brennholt, N., Cole, M., Herrling, M.P., Hess, M.C., Ivleva, N. P., Lusher, A.L., Wagner, M., 2019. Are we speaking the same language? Recommendations for a definition and categorization framework for plastic debris. Environ. Sci. Technol. 53 (3), 1039–1047. https://doi.org/10.1021/acs.est.8b05297.
Hermsen, E., Mintenig, S.M., Besseling, E., & Koelmans, A.A. (2018). Quality criteria for the analysis of microplastic in biota samples: a critical review. In *Environmental Science and Technology* (Vol. 52, Issue 18, pp. 10230–10240). Am. Chem. Soc. https://doi.org/10.1021/acs.est.8b01611.
Hu, Z., Sun, Y., Zhou, J., Sun, W., Shah, K.J., 2024. Microplastics in wastewater plants: a review of sources, characteristics, distribution and removal technologies. J. Contam. Hydrol. 267, 104448. https://doi.org/10.1016/J.JCONHYD.2024.104448.
Jagadeesh, N., Sundaram, B., 2021. A review of microplastics in wastewater, their persistence, interaction, and fate. *Journal of Environmental Chemical Engineering* (Vol. 9, Issue 6). Elsevier Ltd. https://doi.org/10.1016/j.jece.2021.106846.
Kärkkäinen, N., Sillanpää, M., 2020. Quantification of Different Microplastic Fibres Discharged from Textiles in Machine Wash and Tumble Drying. https://doi.org/10.1007/s11356-020-11988-2/Published.
Koelmans, A.A., Mohamed Nor, N.H., Hermsen, E., Kooi, M., Mintenig, S.M., De France, J., 2019. Microplastics in freshwaters and drinking water: critical review and assessment of data quality. Water Res. 155, 410–422. https://doi.org/10.1016/J.WATRES.2019.02.054.
Kurniawan, S.B., Said, N.S.M., Imron, M.F., Abdullah, S.R.S., 2021. Microplastic pollution in the environment: insights into emerging sources and potential threats. Environ. Technol. Innov. 23, 101790. https://doi.org/10.1016/j.eti.2021.101790.
Kurt, Z., Özdemir, I., James, R., A.M., 2022. Effectiveness of microplastics removal in wastewater treatment plants: a critical analysis of wastewater treatment processes. J. Environ. Chem. Eng. 10 (3). https://doi.org/10.1016/j.jece.2022.107831.
Lares, M., Ncibi, M.C., Sillanpää, M., Sillanpää, M., 2018. Occurrence, identification and removal of microplastic particles and fibers in conventional activated sludge process and advanced MBR technology. Water Res. 133, 236–246. https://doi.org/10.1016/j.watres.2018.01.049.
Liu, W., Zhang, J., Liu, H., Guo, X., Zhang, X., Yao, X., Cao, Z., Zhang, T., 2021. A review of the removal of microplastics in global wastewater treatment plants: characteristics and mechanisms. Environ. Int. 146, 106277. https://doi.org/10.1016/j.envint.2020.106277.
Liu, X., Yuan, W., Di, M., Li, Z., Wang, J., 2019. Transfer and fate of microplastics during the conventional activated sludge process in one wastewater treatment plant of China. Chem. Eng. J. 362, 176–182. https://doi.org/10.1016/j.cej.2019.01.033.
Luo, Y., Xie, H., Xu, H., Zhou, C., Wang, P., Liu, Z., Yang, Y., Huang, J., Wang, C., Zhao, X., 2023. Wastewater treatment plant serves as a potentially controllable source of microplastic: association of microplastic removal and operational parameters and water quality data. J. Hazard. Mater. 441, 129974. https://doi.org/10.1016/j.jhazmat.2022.129974.
Ma, M., Huo, M., Coulon, F., Ali, M., Tang, Z., Liu, X., Ying, Z., Wang, B., Song, X., 2024. Understanding microplastic presence in different wastewater treatment processes: removal efficiency and source identification. Sci. Total Environ. 929, 172680. https://doi.org/10.1016/J.SCITOTENV.2024.172680.
Mishra, A., Buhhalko, N., Lind, K., Lips, I., Liblik, T., Väli, G., Lips, U., 2022. Spatiotemporal variability of microplastics in the Eastern Baltic Sea. Front. Mar. Sci. 9. https://doi.org/10.3389/fmars.2022.875984.
Murphy, F., Ewins, C., Carbonnier, F., Quinn, B., 2016. Wastewater treatment works (WwTW) as a source of microplastics in the aquatic environment. Environ. Sci. Technol. 50 (11), 5800–5808. https://doi.org/10.1021/acs.est.5b05416.
Nandakumar, V.K., Palani, S.G., Varma, M.R.R., 2022. Interactions between microplastics and unit processes of wastewater treatment plants: a critical review. *Water Science and Technology* (Vol. 85, Issue 1, pp. 496–514). IWA Publishing. https://doi.org/10.2166/wst.2021.502.
Narloch, I., Gackowska, A., Wejnerowska, G., 2022. Microplastic in the Baltic Sea: a review of distribution processes, sources, analysis methods and regulatory policies. Environ. Pollut. 315, 120453. https://doi.org/10.1016/J.ENVPOL.2022.120453.
Noh, H.J., Moon, Y., Shim, W.J., Cho, E.V., Hong, S.H., 2024. Experimental study on color and texture as cues for plastic debris ingestion by captive sea turtles. Mar. Pollut. Bull. 200, 116055. https://doi.org/10.1016/J.MARPOLBUL.2024.116055.
Parrella, F., Brizzolara, S., Holzner, M., Mitrano, D.M., 2025. Microplastics settling in turbid water: impacts of sediments-induced flow patterns on particle deposition rates. Environ. Sci. Technol. https://doi.org/10.1021/acs.est.4c10551.
Poerio, T., Piacentini, E., Mazzei, R., 2019. Membrane processes for microplastic removal. Molecules 24 (22). https://doi.org/10.3390/molecules24224148.
Qiao, R., Deng, Y., Zhang, S., Wolosker, M.B., Zhu, Q., Ren, H., Zhang, Y., 2019. Accumulation of different shapes of microplastics initiates intestinal injury and gut microbiota dysbiosis in the gut of zebrafish. Chemosphere 236, 124334. https://doi.org/10.1016/J.CHEMOSPHERE.2019.07.065.
Salmi, P., Ryymin, K., Karjalainen, A.K., Mikola, A., Uurasjärvi, E., Talvitie, J., 2021. Particle balance and return loops for microplastics in a tertiary-level wastewater treatment plant. Water Sci. Technol. 84 (1), 89–100. https://doi.org/10.2166/wst.2021.209.
Simon, M., Vianello, A., Vollertsen, J., 2019. Removal of >10 µm microplastic particles from treated wastewater by a disc filter. Water 11 (9). https://doi.org/10.3390/w11091935.
Sun, J., Dai, X., Wang, Q., van Loosdrecht, M.C.M., Ni, B.J., 2019. Microplastics in wastewater treatment plants: detection, occurrence and removal. *Water Research* (Vol. 152, pp. 21–37). Elsevier Ltd. https://doi.org/10.1016/j.watres.2018.12.050.
Sutton, R., Mason, S.A., Stanek, S.K., Willis-Norton, E., Wren, I.F., Box, C., 2016. Microplastic contamination in the San Francisco Bay, California, USA. Mar. Pollut. Bull. 109 (1), 230–235. https://doi.org/10.1016/j.marpolbul.2016.05.077.
Swart, B., Pihlajamäki, A., John Chew, Y.M., Wenk, J., 2022. Microbubble-microplastic interactions in batch air flotation. Chem. Eng. J. 449, 137866. https://doi.org/10.1016/J.CEJ.2022.137866.
Talvitie, J., Mikola, A., Koistinen, A., Setälä, O., 2017. Solutions to microplastic pollution – removal of microplastics from wastewater effluent with advanced wastewater treatment technologies. Water. Res. 123, 401–407. https://doi.org/10.1016/j.watres.2017.07.005.
Talvitie, J., Mikola, A., Setälä, O., Heinonen, M., Koistinen, A., 2016. 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. 109, 164–172. https://doi.org/10.1016/j.watres.2016.11.046.
Uogintė, I., Pleskytė, S., Pauraitė, J., Lujanienė, G., 2022. Seasonal variation and complex analysis of microplastic distribution in different WWTP treatment stages in Lithuania. Environ. Monit. Assess. 194 (11), 829. https://doi.org/10.1007/s10661-022-10478-x.
Korgmaa, Vallo, Laht, Maillis, Rebane, Riin, Lember, Erki, Pachel, Karin, Kriipsalu, Mait, Tenno, Taavo, Iital, Arvo, 2020. Removal of Hazardous Substances in Municipal Wastewater Treatment Plants. https://doi.org/10.2166/wst.2020.264. TalTech.
Vardar, S., Onay, T.T., Demirel, B., Kideys, A.E., 2021. Evaluation of microplastics removal efficiency at a wastewater treatment plant discharging to the Sea of Marmara. Environ. Pollut. 289, 117862. https://doi.org/10.1016/J.ENVPOL.2021.117862.
Wu, M., Tang, W., Wu, S., Liu, H., Yang, C., 2021. Fate and effects of microplastics in wastewater treatment processes. Sci. Total Environ. 757, 143902. https://doi.org/10.1016/j.scitotenv.2020.143902.
Yang, Y., Chen, J., Chen, Z., Yu, Z., Xue, J., Luan, T., Chen, S., Zhou, S., 2022. Mechanisms of polystyrene microplastic degradation by the microbially driven Fenton reaction. Water Res. 223, 118979. https://doi.org/10.1016/J.WATRES.2022.118979.
Zhang, Z., Chen, Y., 2020. Effects of microplastics on wastewater and sewage sludge treatment and their removal: a review. Chem. Eng. J. 382, 122955. https://doi.org/10.1016/j.cej.2019.122955.
Zhou, Y., Wang, J., Zou, M., Jia, Z., Zhou, S., Li, Y., 2020. Microplastics in soils: a review of methods, occurrence, fate, transport, ecological and environmental risks. Sci. Total Environ. 748, 141368. https://doi.org/10.1016/j.scitotenv.2020.141368.
Ziajahromi, S., Neale, P.A., Telles Silveira, I., Chua, A., Leusch, F.D.L., 2021. An audit of microplastic abundance throughout three Australian wastewater treatment plants. Chemosphere 263, 128294. https://doi.org/10.1016/j.chemosphere.2020.128294.