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Accumulation and distribution of microplastics in coastal sediments from the inner Oslofjord, Norway

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ABSTRACT

Microplastic presence in benthic marine systems is a widely discussed topic. The influence of the natural matrix on microplastic distribution within the sedimentary matrix is often overlooked. Marine sediments from the western inner Oslofjord, Norway, were investigated for temporal trends, with a particular focus on the relationship between sediment grain-sizes and microplastic distribution. Density separation, optical microscopy and chemical validation were used to categorize microplastics. Microplastic concentrations ranged from 0.02 to 1.71 MPs g^{-1} dry weight (dw). Fibres were the most common (76%), followed by fragments and films (18%, 6%). Common polymers were polyesters (50%), polypropylene (18%), polymethylmethacrylate (9%), rayon and viscose (5%) and elastane (4%). Microplastics appear to accumulate preferentially according to their morphology and polymer type in certain sediment grain-sizes. Microplastics inputs to the Oslofjord appear to derive from a wastewater treatment plant in the vicinity. Although, the redistribution of microplastics within the fjord needs further investigation.

1. Introduction

Plastic products have a fundamental role in everyday life, with many applications from packaging and textiles, to building, automotive, and within the medical field (PlasticsEurope, 2019). Versatility and low production costs have fuelled the global demand of plastic items, reaching an annual production of 368 million tons in 2018 to which Europe contributes with 57.9 million tons (16% of the global production; PlasticsEurope, 2020). The increasing production rates have been linked to the growing concern of plastics escaping waste handling and entering the environment (Borrelle et al., 2020). Plastics can be moved between terrestrial and aquatic reservoirs, with marine environments acting as a recipient of discarded plastic items, both from land and sea (Van Sebille et al., 2012; Martin et al., 2017; Law, 2017).

Plastic debris can be found in the marine environment in a wide variety of sizes, ranging from meters to micrometres (Barnes et al., 2009). Microplastics are described as a "*heterogeneous mixture of differently shaped materials referred to as fragments, fibres/filaments, beads/spheres, films/sheets and pellets*" (EFSA, 2016; Lusher et al., 2017a). In the current paper we defined them in the 1 μm to the 1000 μm size range as suggested by Hartmann et al. (2019). Microplastics can enter the environment as "primary" and "secondary" microplastics (Andrady, 2011; Cole et al., 2011; Hidalgo-Ruz et al., 2012). Whilst secondary microplastics result from fragmentation and weathering of larger plastic items, primary microplastics result in the direct release of small particles such as pellets, powders, and fibres (Sundt et al., 2014; Lebreton and Andrady, 2019). Primary microplastics, specifically designed to be small in their application, such as microbeads incorporated into personal care products that can be carried with wastewaters via sewers and released to aquatic systems (Napper and Thompson, 2016; De Falco et al., 2019). Most treatment schemes employ an initial screening of influent to eliminate macrodebris and settling to remove dense particles and grit. During this first step it has been estimated that the 78% of microplastics can be removed, whilst sequential steps can often remove the remaining 20% (Hale et al., 2020). Microplastics density, size and shape, highly influence retaining capacities of wastewater treatment plants (WWTPs). Similarly, the filters employed at WWTPs are not specifically designed to retain microplastic fibres which may escape the filtering process and can be released within effluent. Research has suggested that effluent may contribute to 35% of the world ocean's microplastic burden (Boucher

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and Friot, 2017; Salvador Cesa et al., 2017; Long et al., 2019).

The fate of microplastics in the marine environment is influenced by several complex biotic or abiotic processes (Van Sebille et al., 2020). Firstly, the physical characteristics of plastic polymers - including their density - play a fundamental role in the vertical distribution of microplastics in the water and in benthic habitats (Murray and Cowie, 2011). Whilst low-density microplastics tend to be buoyant, plastics with a density that exceeds that of seawater (>1.02 g cm^{-3}) will readily sink (Van Cauwenberghe et al., 2015). Additionally, concurrent processes such as additives leaching out of microplastics, biofouling and incorporation within marine aggregates can cause changes in density of floating microplastics, which may finally sink to the seabed (Còzar et al., 2014; Martin et al., 2017). Marine sediments have been identified as a possible final destination for microplastics in the marine environment (Woodall et al., 2014). Here bottom currents, sediment depth, sedimentation rate, biota redistribution, submarine physiography, distance from the shoreline and human activities (Shen et al., 2019; Harris, 2020; Canals et al., 2021), may further influence their distribution and accumulation.

As a general trend observed for coastal surface waters, microplastics in sediments appear to positively correlate with nearby population density (Van Cauwenberghe et al., 2015). In Norwegian coastal areas most of the population as well as the main industrial and tourism centres are concentrated close to fjords (Gomiero et al., 2019). In particular, fjords have the greatest sediment trapping efficiency of all coastal sedimentary environments (Smith et al., 2015). This is enhanced by the transition from freshwater to saltwater, where sediments suspended in the fresh river water mix with saltwater and give rise to the so-called estuarine turbidity maximum (Burchard et al., 2018). As an example, average numbers of microplastics identified in Norwegian fjords ranged from 190 to 77,000 particles kg^{-1} and exhibit a median of ~7000 particles kg^{-1} (Noren, 2007; Kazmiruk et al., 2018; Black et al., 2018; Haave et al., 2019; Singdahl-Larsen, 2019). This median value is an order of magnitude greater than that for shallow coastal environments or other estuarine environments (Harris, 2020). Although the contamination rate of fjords sediments by microplastics is substantial, the related research is patchy (Noren, 2007; Black et al., 2018; Kazmiruk et al., 2018; Haave et al., 2019; Olsen et al., 2020). Considering currently available published literature, half of the research on microplastics distribution in fjord sediments was conducted on the Norwegian coast near Bergen (Haave et al., 2019) and in the Bunnefjord, eastern Inner Oslofjord (Olsen et al., 2020). In the studies mentioned above, the distribution and the behaviour of microplastics considering sediment's characteristics is generally overlooked. There is the need to understand how microplastics distribute within the sediment column to have a general overview of the possible areas of accumulation and how the natural matrix could influence microplastic occurrence (Martin et al., 2021).

Therefore, the aims of this study were to: i) optimize the approach to sampling microplastics in the inner Oslofjord sediments, by quantifying the distribution of microplastics in sediments within and between sites; ii) understand sources and paths of microplastics and how these particles tend to accumulate in specific areas; and, iii) explain how microplastics abundance varies concerning age of sediments and grain-sizes.

2. Materials and methods

2.1. Study area

The Oslofjord is located on the south-eastern Norwegian coastline and it stretches 100 km northwards from the Inner Skagerrak, from which it is separated by a sill at about 120 m of water depth (Staalstrøm, 2005). The whole fjord system has a surface area of approximately 1644 km^2 (Staalstrøm and Ghaffari, 2015) and it is further divided into the inner and outer Oslofjord by the 12 km long Drøbak Sound, which develops with a shallow sill (19.5 m water depth) at its northern end (Staalstrøm et al., 2012). The inner Oslofjord, is a fjord system consisting of two main basins, the Vestfjord on the west side and the Bunnefjord on the east side, with a water depth exceeding 150 m in both basins and with an average depth of 49 m (Staalstrøm, 2005). The two inner basins are divided by the Nesodden Peninsula and by two sills at 55 m water depth extending from the northern tip of the Nesodden to the mainland (Lepland et al., 2010).

The seafloor is characterised by numerous small basins, shoals and ridges that extend in the NE-SW direction, following fault patterns (Solheim and Grøne, 1983; Lepland et al., 2010). In the inner Oslofjord area, the sediment accumulation rate (SAR) may vary considerably from zero in areas of bottom erosion, to 2.6 mm/year or higher (Lepland et al., 2010). The uppermost part of the seabed's sedimentary sequence is typically very loose and rich in organic matter. Anoxic conditions have developed at the bottom of several deep basins because of restricted deep-water renewals, and eutrophication resulting from a high supply of municipal waste, organic material, pollutants and nutrients (Lepland et al., 2010). Deep water is renewed by the denser water from the outer fjord/inner Skagerrak during the winter and early spring (October/November—April). The main driving force is northerly winds over the Oslofjord/inner Skagerrak area and the strength and duration of northerly winds determine the amount of renewed water (Gade, 1968). Furthermore, the fine grain-size and fluffy surface character of sediments in this depositional basin indicate weak bottom currents and limited bottom transport. The main currents near the bottom have a north-south direction and originate from flow-topography interaction (Staalstrøm and Ghaffari, 2015). In general, the water masses in the fjord are a mixture of Skagerrak water and locally formed waters affected by rivers, where the latter constitute the principal supply of water into the region (Baalsrud and Magnusson, 2002).

2.2. Sample collection

Surface sediments were sampled on the 7th November 2019 from three stations aboard the R/V Trygve Braarud (University of Oslo, UiO). They were collected along a transect in the deep-water channel that runs from the Vestfjordens Avløpsselskap wastewater treatment plant (VEAS WWTP) to the inner Oslofjord inlet in proximity of Håøya island (Fig. 1). The locations of sediment sampling were decided according to prevailing tidally driven water currents, suitable depth (~100 m water depth; Table 1), seabed topography and to avoid areas subject to trawling (Green et al., 2020). Due to safety distance from VEAS diffuser (discharge point) and suitable depth conditions, two stations were placed northeast of the diffuser (ST3, 0 km from the diffuser) and southeast of the diffuser (ST2, 0.4 km from the diffuser). The third station was located at Gråøyrenna (ST1, 11.2 km away from the diffuser).

Surface sediments were collected by a double Gemini corer (10 cm inner diameter) and were sampled using a steel slicer which was rinsed between samples with seawater, and wooden spatulas, discarded after each sample slice. The water-sediment interface was preserved in the cores. Both corer tubes were subsampled to provide duplicate samples from each depth at each sediment station. Sediments were sliced into 1 cm layers from 0 to 5 cm sediment column depth, resulting in a total of 10 samples for each station. Once sliced, samples were placed into cleaned glass jars, rinsed with Reverse Osmosis-water (RO-water) in the laboratory, previously to fieldwork, and were stored at +10 °C in a fridge until processing the following day. During fieldwork, 30 procedural blanks were collected. These consisted of sample jars which were kept open for the duration of the core slicing. They were closed and stored in the same way as the samples.

2.3. Sample preparation and analysis

Wet sediments stored in glass jars were oven dried at 40 °C. Dry sediment was subsampled in 10 g replicates and moved into 50 ml Falcon tubes. A procedural blank was included between 5 samples to test

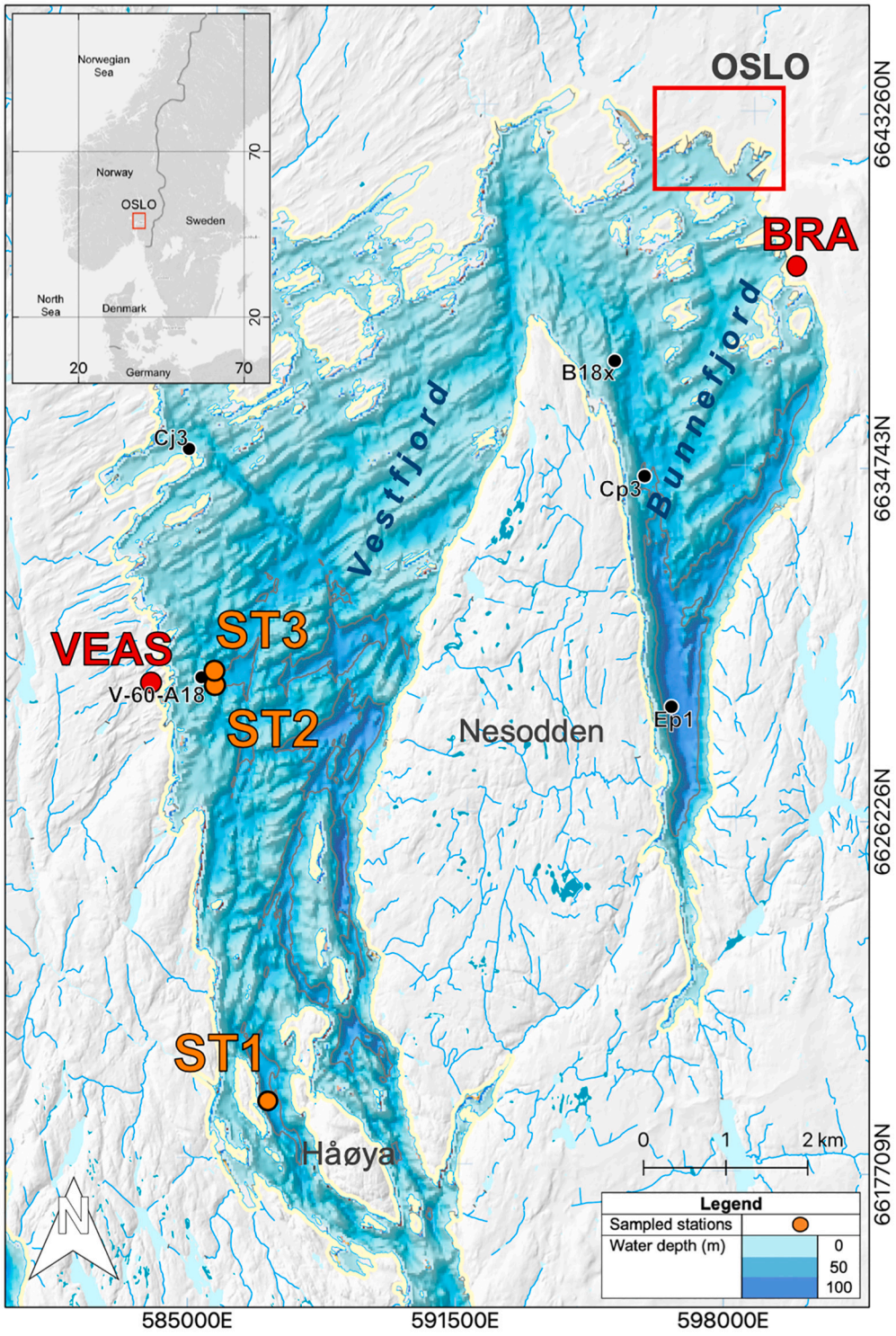

Fig. 1. Depth-coloured shaded-relief image of the Inner Oslofjord and sampled stations. Data are derived from the high-resolution bathymetric dataset collected by the Geological Survey of Norway (water depth 0–100 m). Red dots indicate the location of the two main WWTPs operating in the Inner Oslofjord, VEAS (Vestfjordens Avløpsselskap) and BRA (Bekkelaget). Black dots indicate the locations of the dated cores: Cj3, B18x, Cp3, Ep1 (Dolven et al., 2012) and V-60A18 (Decelles, 2019). (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Table 1
Coordinates (in UTM Zone 32 N) and depths of sampled stations in the Inner Oslofjord.

Stations	ST1	ST2	ST3
North	6,619,347.25	6,629,374.13	6,629,751.75
East	586,924.94	585,438.94	585,432.41
Depth (m)	113.5	98.5	100

background laboratory contamination. During this phase of the work, procedural blanks consisted of empty 50 ml Falcon tubes which were previously rinsed using RO-water following NIVA laboratory protocols.

To extract microplastics from sediments, a high-density Sodium Iodide (NaI) solution (1.8 g cm^{-3}) was applied because it is capable of isolating the majority of common polymer types from sediment matrices (Hurley et al., 2018). Two extractions were performed per sediment sample (and corresponding field and procedural blanks). Prefiltered-NaI solution was added to the Falcon tubes with the dry sediment. Similarly, the procedural blanks (empty Falcon tubes) were filled with the high-

density solution. The samples and blanks were shaken to mix for 30 s, after which they were left to stand for at least one day, until turbidity dissolved. Sediments and blanks were filtered from the solution with a vacuum filtering system (Büchner), samples were passed through a 47 mm Ø Whatmann GF glass fibre filter (pore size: 1.6 μm). When clay clusters occurred, they were crushed with a steel spoon between the first and the second extraction.

After sample preparation, a total of 170 glass fibre filters were analysed, 30 of which were field blanks and 10 were processing blanks. For the visual identification, a stereomicroscope (NIKON S'MZ745T') with a zoom magnification of 7.5× and a long working distance of 115 mm was used. A magnification up to 50× was used with a detection limit of ~30 μm. The microscope was equipped with a camera (Infinity 1, Lumenera) and pictures were processed with 'Infinity Analyze' software. During this step, potential microplastics were counted and sorted into fragments, films and fibres as suggested by Lusher et al. (2020). The longest (L) and shortest (S) axes of particles were measured, and the colour of particles was recorded.

Lastly, chemical validation of potential microplastics was performed using single-point measurements with Attenuated Total Reflectance – μFT-IR ('Perkin Elmer Spotlight 400 μFT-IR Microscope'). The ATR is a rapid surface analysis method that can be used for particles down to a diameter of 2 mm (Renner et al., 2016). To improve the quality of the spectra generated, particles were prepared for analysis using a diamond compression cell (DCC) accessory. Particles were carefully transferred from filter papers to the DCC with use of extra fine micro forceps. The DCC was used to compress particles to a thin, homogeneous thickness. The DCC was then loaded onto the μFT-IR microscope stage for analysis. Measurements were obtained in transmission mode at 4 cm^{-1} spectral resolution for the range 4000 to 600 cm^{-1}. Spectra were produced from a composite of 3 scans. Background measurements were taken before each batch of particle was analysed (Bråte et al., 2020). Once spectra were obtained, they were compared to library spectra. This included the commercially available libraries: PerkinElmer ATR Polymers library, STJapan Polymers ATR library; the BASEMAN library (Primpke et al., 2018); and, in-house curated libraries (including reference polymers, different textile materials, and potential sources of laboratory contamination). Library search matches were compared against the tested particle. The basic principle of library searching is to calculate a numeric score which ranges from 0 to 1 and describes the difference of two spectra. In the case of two identical spectra, the score is equal to 1, whilst if the two spectra differ from each other, the score decreases (Mecozzi et al., 2016). All spectra and matches were manually inspected to confirm the match. Matches between the polymer and reference spectra which exceeded 0.7 were automatically accepted, in accordance with current international procedures (e.g. Primpke et al., 2018; Knutsen et al., 2020; Olsen et al., 2020; Lusher et al., 2020). Microplastics with a match between 0.6 and 0.7 were subjected to further visual examination of spectra characteristics before being accepted or rejected (Lusher et al., 2015; Comnea-Stancu et al., 2017; Kim et al., 2018). This additional analysis was needed because values which fell >0.6 and <0.7 were often a consequence of bio-fouled particles or very thick fragments (i.e. with a thickness > 100 μm). A total of 45 (16% of the total confirmed) low-scoring spectra (>0.6) were included based on visual inspection of the spectra.

All the particles isolated from the samples and the blanks were analysed for chemical characterization, except those that were lost during processing (n = 20, 5.6%). This approach exceeds the recommendation for reporting under European Union's Marine Strategy Framework Directive (MSFD) (Gago et al., 2016). Lost particles were included in the final counts if their colour and form clearly fit within the classification of "Anthropogenic". This follows the protocol of Lusher et al. (2020) when using visual to define microplastics. Transparent, light yellow and black fibres were excluded from the data set to avoid any false positive as these are more likely to be of natural origin.

After chemical validation the number of microplastics per sample was corrected to remove non-plastic materials. Particles were removed if they produced spectra of natural particles, such as merino wool (n = 35), silk (n = 12), non-modified cellulose (e.g. identified as tissue paper) (n = 1). Rayon and viscose were considered microplastics because of the origin of those fibres could be linked with the release of acetate fibres from cigarette butts, sanitary products such as feminine tampons or garments (Wright et al., 2015). Field blanks did not present evidence of procedural contamination during sampling, whilst only two fibres were observed in the procedural blanks: these were identified as silk and merino wool. No sample correction was applied as these materials were not considered to be plastic.

2.4. Contamination control

Strict controls were followed during sample collection and processing in the laboratory to eliminate post-depositional contamination. Prior to fieldwork, all sample glass containers were pre-washed thoroughly with RO-water (filter Millipak® Express 20′ for 0.22 μm particulate) before use. During sample collection, nitrile gloves (NBR) and security coveralls were used, glass and metal equipment was used whenever possible.

Laboratory analyses were conducted in the "Mikroplast lab" at NIVA in Oslo, a dedicated laboratory for microplastics analyses. Processing was performed in a fume hood/laminar flow whilst, microscopy work was performed outside of the laminar flow bench. However, to minimize the contamination from the laboratory atmosphere, the samples on filter papers were enclosed in petri dishes whilst they were analyzed under the microscope. The upper lid was removed just only when a potential microplastics needed to be manipulated, or whilst taking pictures.

Reagents were vacuum filtered through 'Whatmann glass microfiber filters GF/A' (Ø = 47 mm, pore size: 1.6 μm) immediately prior to use. All equipment was cleaned with RO-water and the use of plastic laboratory equipment was kept to a minimum. In the laboratory gloves are mandatory as personal protective equipment (PPE; NIVA security protocols), NBR blue gloved were used. This polymer is resistant to oils and acids and due to its colour is easily recognizable during optical identification. Any particles identified to match NBR polymer and morphology combination were excluded from further analysis.

2.5. Sediments analyses and sediment accumulation rate

Grain-size analyses were performed at the Earth Science Department, University of Pisa. Sediments were investigated with the particle size analyser: the 'CAMSIZER x2 (RETSCH technology, particle characterization)'. The particle size analyzer is based on the principle of dynamic image analysis which provides precise particle size and shape information of sediment grains. Ultrabright LED stroboscopic light sources and two high-resolution cameras achieve a frame rate than 300 images per second which are evaluated in real time by the CAMSIZER x2 software. For the analysis of sediments, the X-Flow module was used. Thanks to this feature, liquid suspensions of sediments can be investigated. An ultrasonic bath and a strong centrifugal pump ensured efficient dispersion of particles contained in the suspension. With this analysis mode, particles within a dimensional range of 0.8 mm and 1000 μm can be detected (CAMSIZER x2, user's manual).

Prior to grain-size analysis, sediments were stored in 50 ml Falcon tubes. Demineralized water was added to each Falcon tube 48 h before grain-size analysis in order to dilute the sediment suspension. The solution was picked up with a 5 ml pipette and few drops were released in the dispersion bath of the particle size analyzer. From here, the solution traversed a close loop to the flow cell where the camera system captured particle images, for which an upper limit of 30,000 images per cycle was set.

Sediment dating was calculated from two previous works, to assess the degree of microplastic burial within the sediment column after deposition. In the work of Dolven et al. (2012), four cores were collected

and dated, three in the Bunnefjord (Ep1, Cp3, B18x) and one in the Vestfjord (Cj3; Fig. 1). The samples were collected in February and April 2009 and the sites were chosen considering the highest possible sedimentation rate and the least disturbed locations. The samples were sent to the Gamma Dating Centre in Denmark and were analysed for ^{210}Pb-, ^{226}Ra- and ^{137}Cs- activity via gamma spectrometry carried out on a Canberra Ultraslow-background Ge-detector. The other work considered is the Decelles' (2019) master thesis. The core V-60-A18 was sampled in May 2018 using a double Gemini corer (Fig. 1). The core was sent to the Environmental Radioactivity Research Centre at the University of Liverpool for the radiometric dating. The samples were analysed based upon direct gamma assay of ^{210}Pb, ^{226}Ra and ^{137}Cs radionuclides. From the data of these two works, the mean sediment accumulation rate (SAR) for the Vestfjord was calculated.

2.6. Multivariate analysis

Past4.03 software was used to perform Canonical Correspondence Analysis (CCA, Legendre and Legendre, 1998). This type of multivariate analysis was used to examine linear relationships between the dataset and environmental variables. The occurrence of different grain-sizes (clay, silt, sand) was considered as environmental variable and was compared to the occurrence of fibres, films, fragments and the type of polymers (polyesters, polypropylene, polymethylmethacrylates, etc.).

3. Results

3.1. Plastic particles distribution

After chemical confirmation, a total of 310 microplastics were found in all environmental samples. Considering the full sum of plastic particles within each core, the majority of microplastics were found at ST2 (62%), followed by ST3 (33%) and ST1 (5%). A total of 191 particles were identified at ST2 which represented a concentration of 1.04 microplastics per gram of sediment dry weight (MPs g^{-1} dw). A total of 102 particles were found at ST3, equating to 0.52 MP g^{-1} dw. The lowest concentration was found at ST1. Only 17 microplastics were found, equating to 0.08 MPs g^{-1} dw (Fig. 2).

Between sites, there appeared to be some differences between the number of microplastics at different depths within each core. For example, there was a difference in abundance of microplastics when the sediment layers (1 cm slices) were investigated at ST2.

The layer 4–5 cm appeared to be the most influenced by the presence of microplastics, 1.7 MPs g^{-1} dw, followed by the superficial layer (0–1 cm) with 1.12 MPs g^{-1} dw. The layers 1–2 cm and 2–3 cm showed a similar distribution with 0.94 and 0.91 MPs g^{-1} dw respectively. Whereas, section 3–4 cm (0.53 MPs g^{-1} dw) had the lowest concentration of microplastics. The distribution of microplastics at ST3 decreases toward the top of the core. The highest concentration was observed in 4–5 cm (0.80 MPs g^{-1} dw) whereas the lowest concentration was observed at 1–2 cm (0.34 MPs g^{-1} dw). The remaining layers 0–1 cm, 2–3 cm, 3–4 cm presented 0.46, 0.50 and 0.52 MPs g^{-1} dw respectively.

The sediment layers with the highest microplastic concentrations at ST1 were 4–5 cm and 3–4 cm, with 0.17 and 0.10 MPs g^{-1} dw each. The lowest microplastic concentration was found in layer 2–3 cm (0.02 MPs g^{-1} dw). This was followed by layer 0–1 cm (0.06 MPs g^{-1} dw) and layer 1–2 cm (0.07 MPs g^{-1} dw).

3.2. Microplastics dimension and morphology

The plastic particles found in Oslofjord's sediments had a wide range of dimensions. The smallest particle detected with the optical microscope had the longest axis dimension of 35 μm, whereas the longest was 8754 μm long. Considering the longest dimension of particles (L), two main dimensional classes were identified: microplastics (30 μm < L < 1000 μm) and mesoplastics (1000 μm < L < 10,000 μm). Microplastics

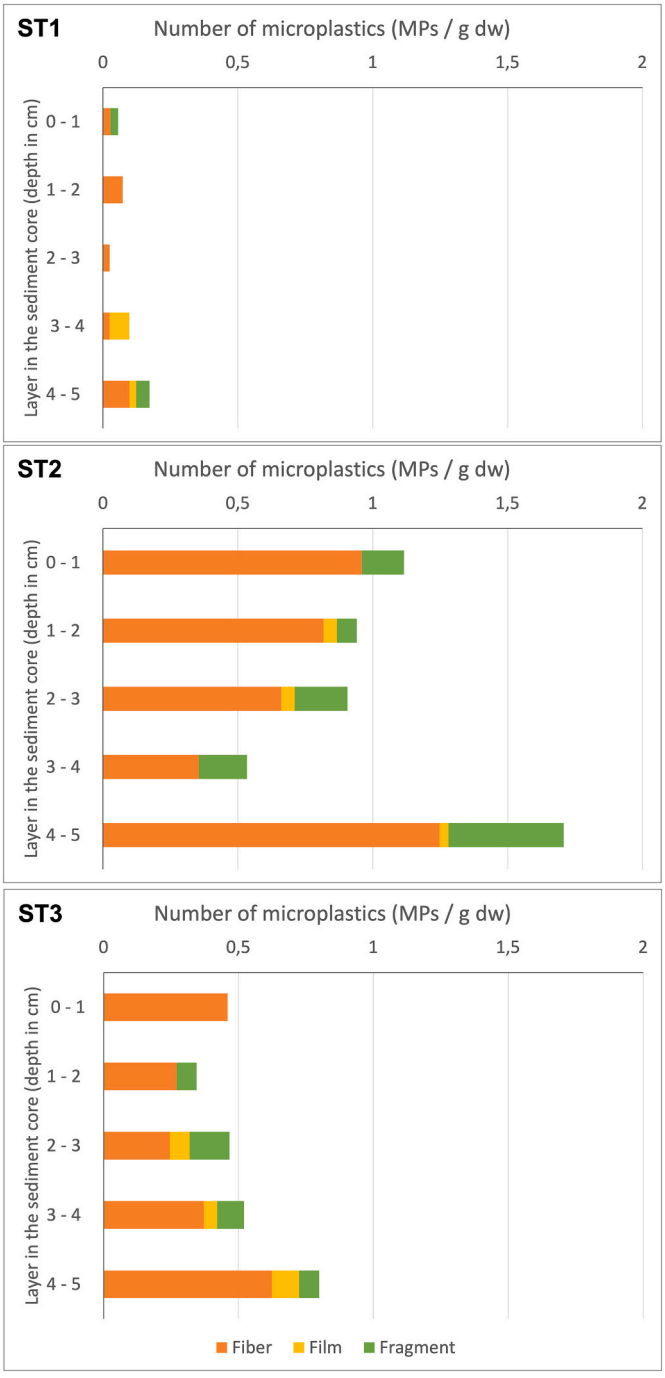

Fig. 2. Distribution of plastic particles in Oslofjord samples and layers of each core. Morphologies (fibres, films and fragments) plus microplastics abundances are normalized to MPs/g d.w.

were further divided in subclasses. The size classes of 30–200 μm, 200–400 μm and 400–600 μm showed a similar distribution, comprising of between 25% and 21% of the overall particle count. Abundances decreased in the subclasses 600–800 μm (16%) and 800–1000 μm (14%). The subclass of 1000–2000 μm contained the majority of the observed particles, with an abundance of the 54%. Largest particles were gathered in the 2000–3000 μm subclass and accounted for the 21%.

Fibres were the most common shape found, they accounted for 76% of all particles, followed by fragments (18%) and films (6%). Fibres were pervasive throughout cores, whilst the distribution of fragments and films were patchier.

Fibres accounted for the 59% of the particles at ST1, followed by

films (24%) and fragments (18%) Fibres were found in every layer and were most abundant in 1–2 cm (n = 3) and 4–5 cm (n = 4). Fragments were only present in the layers 0–1 cm (n = 1), and in 4–5 cm (n = 2). Films were found in deeper layers: 3–4 cm (n = 3) and 4–5 cm (n = 1).

Similarly, fibres accounted for 78% of the particles found in core ST2, whilst fragments and films accounted the remaining 19% and 3% respectively. Generally, fibres were abundant in every layer, although they decreased in layer 3–4 cm. Fibres and fragments were more abundant in layer 4–5 cm where 38 fibres and 13 fragments were counted. Films occurred in layers 1–2 (n = 2), 2–3 (n = 2) and 4–5 cm (n = 1).

Fibres were found in all layers at ST3, but were more abundant in layer 4–5 cm (n = 25). No other particle morphologies were observed in the first layer (0–1 cm) whereas in layer 1–2 cm three fragments were present in addition to fibres. The abundance of fragments decreased from layer 2–3 cm (n = 6) to 4–5 cm (n = 3), whereas the distribution of films accounted for one particle for each of these last layers.

Considering all stations and all intervals, the most frequent colours observed were transparent (37%), blue (21%), black (13%), green (10%), red and yellow (7% each). Few particles were pink (3%), orange (1%) and brown (<1%).

3.3. Chemical characterization

A total of 359 potential microplastics were initially counted, of this number a total 285 (79%) particles were chemically confirmed as microplastics. Due to sample loss, 21 particles were just visually confirmed and not excluded from plastics counts, 4 particles were confirmed as plastics but could not be assigned to a polymer (Table 2). Lost and undetermined particles accounted together for the 7%. In total across every station, the most abundant polymer was polyesters (PES) (50%, 154 particles) followed up by polypropylene (PP) (18%, 56 particles), acrylates and polymethylmethacrylates (A + PMMA) (9%, 26 particles). These three polymers represented more than 75% of all particles (Fig. 3).

3.4. Sediments analyses

Sediment grain-size distribution was similar between samples in the Oslofjord. Generally, cores were dominated by both silt (from very fine to very coarse) and sand (from very fine to medium) (Friedman and Sanders, 1978). A low portion of clay was detected (1–4%). The range between coarse silt and fine sand was the most common, with some exceptions. The silt/sand ratio was favourable for the sand grain-size in 9 layers of the 15 total layers analysed between stations (ST1, ST2, ST3). The end members (clay and coarse sand) were found only in few layers. Overall, ST1 and ST3 were characterised by relatively coarser sediments, whilst the abundances of finer sediments were slightly higher for ST2.

3.5. Age of sediments

An average SAR of 0.296 ± 0.02 cm/year was calculated for the Oslofjord with varying accumulation rates found throughout the fjord. The maximum of 0.42 and 0.36 cm/year were observed at station Ep1 and Cj3 respectively. The station Cp3 and V-60-A18 showed a similar pattern of approximately 0.20 cm/year (Fig. 4).

The age of sediments for the ST1 layers was calculated considering the 0.29 cm/year average accumulation rate of the Oslofjord. The year 2019 was set as the upper limit of the 0–1 cm layer, thus 2015 was identified as the lower limit. Starting from here, the lower limits of the core bottom layer (4–5 cm) were individuated at 2005 for the upper boundary and 2002 for the lowest one.

Considering the estimated SAR for station V-60-A18 (0.2 cm/year), the age of sediment layers was calculated for both ST2 and ST3 due to the proximity of the three stations. As for ST1, 2019 was considered as the upper boundary of layer 0–1 cm, then 2014 was identified as the lower limit. Lastly, 1994 was individuated as the lowest boundary of the core bottom layer (4–5 cm) with 1999 as the upper limit of this last layer.

Table 2
Quantities of microplastics confirmed across all sampling locations.

	ST1	ST2	ST3	Total
Potential MPs	22	202	135	359
Confirmed MPs	17	191	102	310
Lost	2	11	8	21

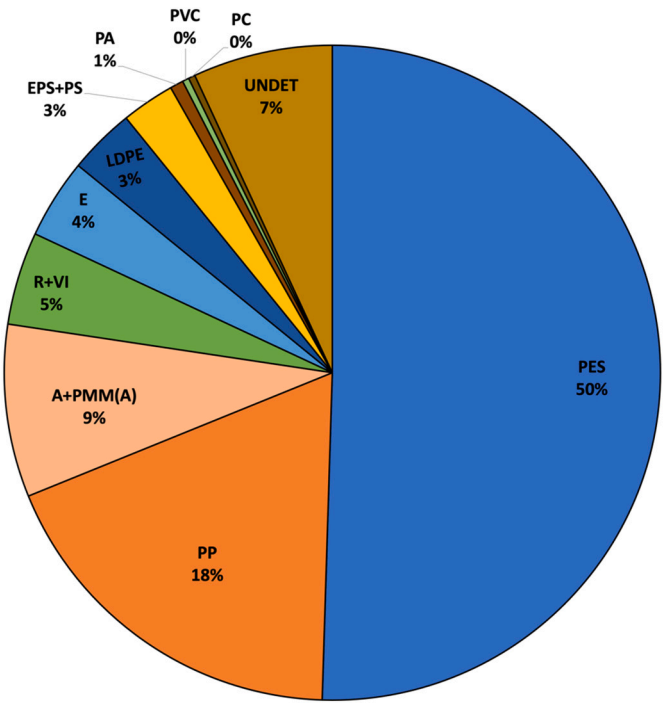

Fig. 3. Polymers detected in all Oslofjord samples (ST1, ST2, ST3). Abundances are expressed in percentages.

3.6. Multivariate analysis

From CCA was observed that plastic particles appeared to accumulate preferentially according to their morphology (Fig. 5). Fibres and fragments were more influenced by sandy sediments, whilst films accumulated mostly in silty sediments. Polymers too showed a preferential accumulation according to sediments. Such as PES, low-density polyester (LD-PE), expanded polystyrene and polystyrene (EPS + PS) and polyvinylchloride (PVC) which settled in finer grain-sizes (silt), like A + PMMA, elastane mix (E), rayon and viscose (R + VI), polyamide (PA) and polycarbonate (PC) but with a stronger correlation with clay. PP is the only polymer that occurred preferentially in sand.

4. Discussions

4.1. Visual identification quality

Of the 359 potential microplastics identified by visual microscope assessment, 285 particles (79%) were confirmed as microplastics using FT-IR analyses. Whilst 21 particles (6%) could not be chemically confirmed as they were lost prior to chemical inspection.

The protocol presented in Lusher et al. (2020) allowed the identification of potential microplastics under an optical microscope. It addresses the definition of particles' morphology, considering their optical

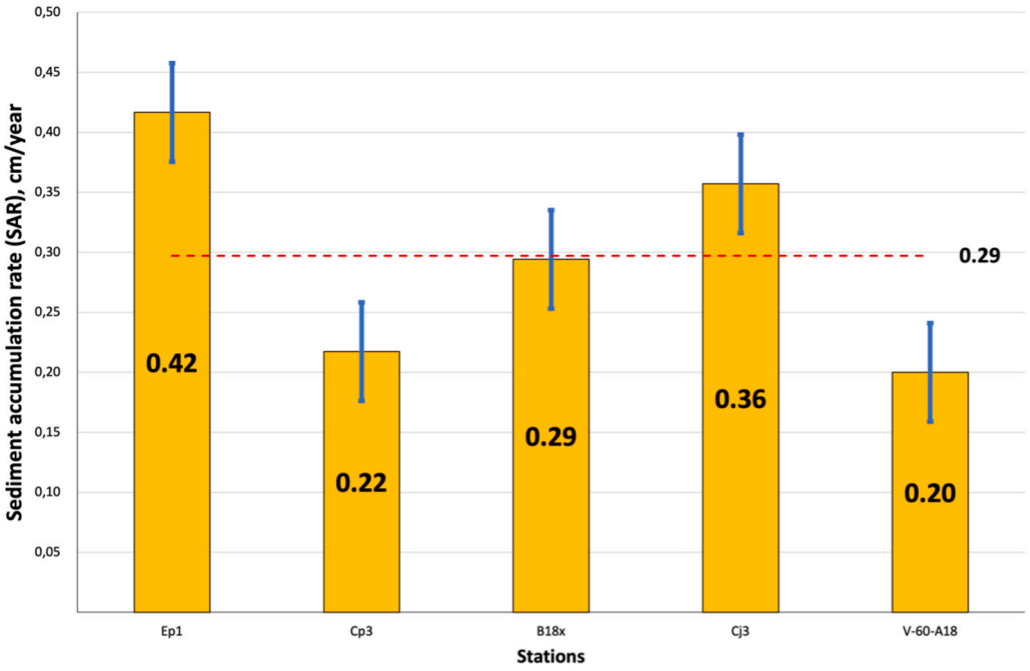

Fig. 4. Sediment accumulation rate (SAR) for the dated locations (EP1, Cp3, B18x, Cj3, V-60-A18) in the Inner Oslofjord. The red dotted line indicates the average accumulation rate calculated for the whole fjord. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

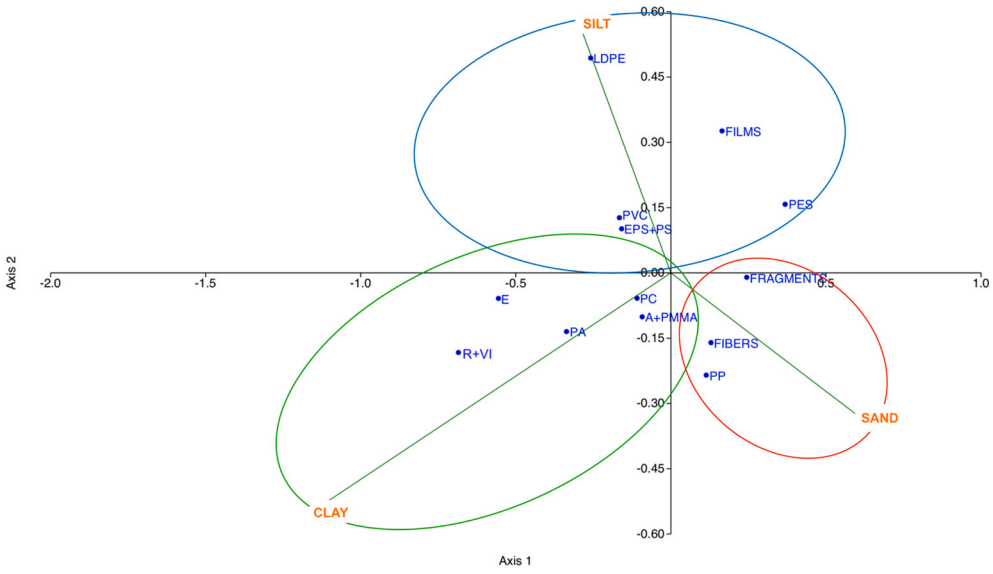

Fig. 5. CCA in which grain-sizes are included as environmental variables and are compared to plastic particles characteristics (morphology and polymeric composition). In the red circle are gathered those particles influenced by sandy sediments, in the blue circle particles influenced by silty sediments and in the green circle particles that show a better correlation with the clay grain-size. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

properties, whilst defining their physical behaviour manipulating them with forceps during the analysis. The protocol has been deemed efficient, especially considering fragments (59 potential, 50 chemically confirmed, 85% correct) and fibres (255 potential, 217 chemically confirmed, 85% correct). Bright and anthropogenic colours (red, blue, green, yellow, etc.) appeared to be reliable markers of a particles' polymeric origin, even considering very small particles – e.g. the smallest fragment found (35 μm) was identified thanks to its greenish colour (confirmed with FT-IR as PP). Transparent fragments and fibres were visually identified and were the two most common combinations (37%) among the other morphotypes characterised by different colours. However, no transparent fragment <250 μm were found. It is possible that smaller transparent fragments may have been overlook, and thus underestimated.

Some issues were encountered for films when applying visual identification: of 28 potential plastic films, 18 of these were confirmed with FT-IR analysis. Discarded films were all fragmented, yellowish in colour, easily foldable and, when folded, did not broke. These characteristics can be easily shared with both anthropogenic films and organic matter (e.g. algae). For these latter, FT-IR spectra were characteristic of material with a cellulose origin. The best match reference spectra were those of silk and wool, although it is not possible to differentiate these from natural cellulose, such as algae. This misinterpretation could be either related to inexperience of the user or operation error, meaning that sufficient training and or experience is required for microplastic identification in environmental samples. This supports conclusions by

several authors that visual identification alone (without the use of confirmation techniques) cannot be used for the identification of smaller microplastics (<1 mm) (Isobe et al., 2019; Lusher et al., 2020; AMAP, 2021). Lastly, FT-IR confirmation analyses allowed users to not overestimate microplastic abundances, as seen here with films.

4.2. Abundances

The abundances and the distribution of microplastics between stations in the Oslofjord varied widely even when considering stations in proximity (i.e., 0.37 km between ST2 and ST3). Microplastics were found at every station and within every core. The sampling site downstream and close to the VEAS WWTP diffuser (ST2) was the most polluted location identified in this study, where a total of 104 particles were identified in 100 g of sediment (0–5 cm, normalized values). This value was not reflected at other sites within the same quantity of sediment ($n_{ST3} = 52$, $n_{ST1} = 8$; normalized values). Interestingly, there was a difference in microplastic numbers between ST2 and ST3 even with the close proximity. The abundance of plastic particles at ST2 was doubled that of ST3 One of the principal sources of microplastics in this area is believed to be the VEAS WWTP system. A diffuser releases treated water into the Oslofjord via an outlet tunnel and five long distribution pipes in proximity to the stations downstream (ST2) and upstream (ST3). This outlet spreads the treated effluent from a depth of 20 m (https://www.veas.nu/produkter/vann). The distribution of outlet waters was assessed before building and the operation of the system during a three-day dye drop experiment (Bjerkeng et al., 1978). The flow from VEAS showed that the dye released in the Oslofjord moves generally toward the southeast, carried by tidally driven surface waters (Staalstrøm and Røed, 2016), with varying concentrations considering the distance from the discharge point. The finding of the dye drop experiment appeared to be consistent with the quantity of microplastics found at ST2 which is located more southerly than the VEAS diffuser. ST3 would be expected to have a higher occurrence of microplastics because it is located close to the diffuser. However, the results of this study showed a reverse distribution considering ST2 and ST3. Considering that the microplastic composition was mostly dominated by fibres and by polymers that are less dense than seawater, it could be expected that microplastics did not readily sink to the seafloor but tended to be buoyant in the water column for a period of time. The average SAR for these two sites was calculated to be of 0.2 cm/year but these estimates can be valid for a mineral grain, such as spherical quartz, which can have a density of 2.65 g cm^{-3} (Harris, 2020). The polymer with the highest density observed during analysis was polystyrene with a density of 1.05 g cm^{-3} in the non-expanded form. Thus, the SAR for microplastics is expected to be even lower than that for sediment particles, even when microplastics are weathered or biofouled.

Additionally, when considering the seafloor geomorphology of the area, it is more likely that microplastics accumulate in proximity of ST2 than of ST3. Indeed, whilst the first site is located in an almost flat area with a bathymetric high in its southernmost part, ST3 is located in proximity of a threshold where the basin gets deeper below the sampling station. It is predictable that the major burden of microplastics which pollutes that area will probably be found in the bottom part of this enclosed area.

The deepest site sampled in the Oslofjord was ST1 (113.5 ± 0.38 m) which also contained the lowest number of microplastics (n = 17, 0–5 cm). Interestingly, this site is influenced by the action of both superficial and bottom currents which cause major changes of water masses between the outer and the inner Oslofjord. Between September and November, the deep water entering the inner Oslofjord from the outer fjord, has a velocity of 0.25 m/s at a water depth between 111 m and 116 m with the strongest current in the north-south direction (Staalstrøm and Ghaffari, 2015). This current splits into two main northward branches, due to the presence of several islands in the Oslofjord inlet. Plus, the presence of the Drøbak Sound further reduces the strength of this bottom current. The current that flows above ST1, has a flux diffusivity rate of ~2 cm^2/s at a depth between 90 and 125 m (Staalstrøm et al., 2012), resulting in an area more influenced by bottom sediment transport. Thus, it is unlikely for high numbers of microplastics to accumulate in this area. Instead, surface currents show a maximum strength outward from the inner Oslofjord (Albretsen et al., 2004). Collignon et al. (2012) observed that strong currents could increase the mixing and the vertical redistribution of microplastics in the upper and mid layers of the water column. Thus, the strong overflowing current could even transport slowly sinking microplastics that will accumulate on the seafloor when increasing in density or when flow conditions allow settling. It is thus uncertain if the main supply of microplastics at ST1 comes from the outer Oslofjord via bottom currents and turbulent flows, or if even sub-superficial currents contribute to the transport and deposition of microplastics. As only one sample was analysed in this area, further research is required to better understand the pattern of distribution of microplastics and elucidate the hypothesis.

4.3. Vertical distribution and plastic particles characteristics

Following the estimation of the SAR, microplastics were calculated to accumulate in the Oslofjord sediments over the last 26 years (Table 3). Sediments at ST1 were 8 years younger than those of ST2 and ST3. Indeed, the bottom age for core ST1 was calculated to be 2002, whereas 1994 was calculated for both ST2 and ST3. The layer 4–5 cm of ST1 settled in correspondence of the transition between layer 3–4 cm and 2–3 cm of ST2 and ST3. From the analysis of microplastics abundance between cores, this correlation seems to be confirmed because the relative distribution of fibres, films and fragments follow the same distribution in these layers. A general trend can be observed between sites, especially considering ST1 and ST3 where the two cores show a general decreasing accumulation of microplastics from 2002 to 2014/2015. This is in agreement with the study performed in the eastern inner Oslofjord (Singdahl-Larsen, 2019). Additionally, the lowest quantity of microplastics deposited for both ST1 and ST3 was for the year 2009. This trend is reversed at ST2, as the quantity of microplastics increased from 2002 to 2014 suggesting a higher accumulation capability for microplastics in the ST2 area.

The present study shows that settling of microplastics is not sufficient to explain their distribution in benthic habitats. Benthic organisms were not investigated here although their influence on the distribution of microplastics cannot be ruled out. It is well known that bioturbation has a key role in the vertical and horizontal redistribution of microplastics in sediments (Graham and Thompson, 2009; Taylor et al., 2016; Courtene-Jones et al., 2017; Näkki et al., 2017; Bour et al., 2018). The intensity of bioturbation is dependent on species composition due to the difference on specific characteristics such as typical burrowing depth and habitat of certain species (Josefson et al., 2012). The small size of microplastics makes them available for interactions with marine biota in many other ways including feeding mode and thus ingestion/egestion at different trophic levels (Wright et al., 2013). Existing data indicate that microplastics are widely distributed in digestive tracts of a range of organisms living in marine benthic habitats (Gonçalves et al., 2019; Iannilli et al., 2019; Sfriso et al., 2020). As an example, in a recent study of Bråte et al. (2020) for the Nordic Council of Ministers, several Nordic environments were analysed. Many stations were sampled in the Baltic Sea, in Denmark, on the Norwegian Coast, in Faroe Islands, Iceland and Greenland. Between all these locations, it was demonstrated that the species inhabiting the inner Oslofjord are the most polluted ones between those analysed (*Limelicola balthica, Abra nitida* and *Thyasira* spp., *Mytilus* spp.). An average of 61 microplastics per individual was reported. In a similar study, Bour et al. (2018) highlighted that the ingestion rate and occurrence rate of microplastics in deposit-feeders (*Ennucula tenuis, Ophiura alida, Brissopsis lyfera, Hediste diversicolor*) was equal that of predators (*Hippoglossoides platessoides, Enchelyopus cimbrius, Trisopterus esmark*i), but higher than that for filter-feeding

Table 3
Age of sediments along the layers of ST1, ST2 and ST3 cores.

ST1 layers (cm)	year		ST2-ST3 layers (cm)	year	
0 - 1	2015	SAR (cm/year) = 0.297	0 - 1	2014	SAR (cm/year) = 0.2
1 - 2	2012		1 - 2	2009	
2 - 3	2009		2 - 3	2004	
3 - 4	2005		3 - 4	1999	
4 - 5	2002		4 - 5	1994	

species (*Amphiura filiformis, Sabella pavonina, Crangon allmanni*). Authors themselves suggested to improve the investigation of the distribution of microplastics in water and sediments of the Oslofjord to evaluate how biological or environmental factors could explain their results. Based on this information, the benthic fauna from the sampled locations in the Oslofjord may play a role in the redistribution of microplastics and future research should investigate benthic fauna in the same locations.

Considering the morphological and distributional characteristics of microplastics, it appeared that fibres were pervasive throughout the cores, whereas films and fragments tended to accumulate in deeper layers. Moreover, due to the proximity of inhabited areas, it was expected that beads and pellets would be found in sediment samples, although none were observed. It is possible that the efficiency of the WWTPs that operate in the Oslofjord may have contributed to this observation. Both VEAS (western inner Oslofjord) and Bekkelaget (BRA, eastern inner Oslofjord) WWTPs have tertiary treatment facilities meaning that waters undergo both chemical and biological treatment steps. For instance, differently from secondary treatments the biological step in these tertiary systems employs nitrogen removal from wastewaters, whilst in primary systems waters just passes through fine screens (~350 μm in Tomasjord WWTP, Tromsø). Additionally, the VEAS system has a separate treatment for excess stormwater which employ mechanical and chemical cleaning processes (Lusher et al., 2017b). Research has shown that sludge produced from these two of the WWTPs operating in the Oslofjord region (VEAS and Bekkelaget) were capable of retaining a total of 965,535,470 particles/m^3 per day, mostly composed by beads and fragments. The average particle size of fragments identified in sludge samples was 414 μm for Bekkelaget sludge and 312 μm for VEAS sludge.

Microplastics analysed in the present study from the Oslofjord had an average dimension of 256 μm. Thus, it is possible that smaller plastic fragments are not retained by WWTP filtering systems and can be easily released into the marine environment from diffusers (Browne et al., 2011; Carr et al., 2016). As an example, setting the lower microplastics dimensional limit at 400 μm, 81% of the microplastics identified in this study may not be retained in WWTPs and if we consider 300 μm as the lower limit for the longest microplastic's axis, 65% of microplastics will be released from both diffusers.

Plastic fibres constitute the largest burden of microplastics in Oslofjord sediments. Although larger debris is removed in WWTPs, filters are not specifically designed to retain them (Zubris and Richards, 2005). As stated above, the WWTPs that operate in the Oslofjord, especially Bekkelaget system, have a higher retaining capacity for beads (nV = 23, nB = 93) and fragments (nV = 10, nB = 80) than for fibres (nV = 11, nB = 35; Lusher et al., 2017b). Some experiments demonstrated that a single garment could produce >1900 fibres per wash (Browne et al., 2011), the direct consequence is that a large portion of microplastic fibres which were found in Oslofjord sediments may have derived WWTPs which showed low capacity to retain fibres.

Many polymer types were identified in the Oslofjord, with fibres mostly composed by PES (68%) and PP (33%), which was more common for fragments (36%) and films (33%). Generally, PP and PES are very versatile polymers, together they constitute the most demanded resin types globally. Both are used for the clothing production, but PP is also used for food packaging, hinged caps, microwave containers, pipes, etc. For PP, the 2018 annual demand accounted for the 19.3%. PES differentiates for the low- and high- to medium-density forms, it is generally employed for reusable bags, trays and containers, agricultural film, food packaging, etc. in its lighter composition (17.5% 2018 global demand). Whilst the heavier form is used to produce toys, milk bottles, shampoo bottles, pipes, houseware, etc. (12.2% 2018 global demand; PlasticsEurope, 2019). A major part of microplastics analysed in Oslofjord samples looked weathered (51%) and biofouled (28%), indicating a high residence time in seawater before they were able to reach the benthic domain. Lastly, strong correlations have been observed from CCA between polymers and sediment grain-size, such as the relationship between PES and fine sediments, PP with sand grain-sizes and clay with PMMA and E.

5. Conclusions

Microplastics have been found in surface sediments (0–5 cm) within every sample analysed in this study from the inner Oslofjord. Considering the sum of all the stations (ST1, ST2, ST3), the total concentration of microplastics was about 0.55 MP g^{-1} dw. The stations upstream (ST3) and downstream (ST2) of the VEAS WWTP diffuser were the most polluted stations, whilst the furthest station outward of the fjord (ST1) showed a low occurrence of microplastics, probably due to the high hydrodynamics of the location in which this sample was collected. Sites ST1 and ST3 showed that the concentration of microplastics decreased from 2002 and 2014/2015, indicating a reduction in release of microplastics or an improvement in WWTPs systems' retaining capacity. Reversely, at ST2 the amount of microplastics increased, likely indicating an area susceptible for microplastic accumulation.

Besides sinking of microplastics driven by increasing of density, it was supposed that the main transport agents in the locations analysed are shallow currents which redistribute microplastics between locations. For ST1, further analyses are needed to understand the main direction from which microplastics reach this area and thus if the main input is from the inner or the outer Oslofjord. From the correlation between microplastics characteristics and composition of sediments found in every sample analysed in this study, it was observed that there are some peculiarities in the deposition of microplastics. For example, fibres and fragments will preferentially accumulate in relatively coarse sediments such as sand from very fine to coarse, whilst films accumulate preferentially in fine sediments like silt. Acrylates, polyesters, elastane, rayon and viscose, polyamides and polystyrenes will accumulate preferentially in finer sediments such as clay and silt, whilst polypropylene was the only polymer that occurred in sandy sediments in our study.

CRediT authorship contribution statement

Laura Bronzo: Investigation, Data curation, Formal analysis, Writing – original draft. **Amy L. Lusher:** Methodology, Data curation, Funding acquisition, Writing – review & editing. **Merete Schøyen:**

Investigation. **Caterina Morigi:** Supervision, Writing – review & editing.

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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Appendix A. Supplementary data

Supplementary data associated with this article can be found in the online version, at https://doi.org/10.1016/j.marpolbul.2021.113076. These data include the Google map of the most important areas described in this article.

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