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Title	Hydrodynamic forcing and sand permeability influence the distribution of anthropogenic microparticles in beach sediment.
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Abstract

The distribution of anthropogenic microparticles (Mps), such as plastic and natural fibres used in textiles, in beach sediments was studied in a human-influenced pocket beach in Liguria (NW Mediterranean Sea). Information on environmental parameters such as rainfall, hydrodynamism and sediment texture was collected at the same time as the sediment samples. The Mps (416±202 Mps kg-1 on average) were mainly fibres (57-100%), while fragments and spheres showed irregular abundances linked to the draining action of waves on the beach. Uni- and multivariate statistical analyses highlighted that the different spatial and seasonal distribution of fibres primarily depended on the action of the waves that force seawater into the sand, rather than on sedimentation following depositional processes. Wave height and direction had a role in fibre distribution in the sand, as well as sediment permeability and sorting. The occurrence of short-term and spatially-localised hydrodynamic events such as rip currents were observed to influence the abundance of fibres, overlapping the seasonal sequences of beach accretion and erosion that is typical of the area and increasing fibre abundance by transporting those accumulated in the sediments of the submerged beach during winter.

Keywords	anthropogenic microparticles; beach sediment; hydrodynamism; sediment texture; NW Mediterranean.
Taxonomy	Beach Process, Litter, Pollution Impact on Marine Environment

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Submission Files Included in this PDF

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Genova, 24 September 2019

Dear Editor of Estuarine, Coastal and Shelf Science,

We submit the third revised version of the manuscript "Hydrodynamic forcing and sand permeability influence the distribution of anthropogenic microparticles in beach sediment" by C. Misic, A. Covazzi Harriague and M. Ferrari.

Thank you for your attention and do not hesitate to contact me at the address below for any further requirement.

Looking forward to hearing from you, I remain

Sincerely yours

Cristina Misic

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Response (in bold) to Reviewer 3

This is an interesting attempt to explain the distribution of microplastic particles in a wave-stirred beach. The English needs a final editing: for instance

-sea calm condition should be calm sea condition

- Hydrodynamism is not in the English online dictionary nor in Thesaurus. Try: 'hydrodynamic nature' etc.

The text has been subjected to a final editing by a native English speaker. The text of the first revision was processed by a certified English proofreading service before (the certificate was attached to the first revised manuscript). "Hydrodynamism" was changed with "hydrodynamic characteristics". "Sea calm condition" was changed with "calm-sea condition".

I would personally like to see more thinking and hopefully an additional interpretation about Figure 7. Indeed the January data are the odd points that do not fit in the general pattern. What happened in January (what additional processes occurred?)? With so much scatter in Figure 7 (if the January data are included), hardly any explanation is possible to explain Figure 7, even if statistics give you a 0.5 coefficient of fit - but visually that seems just a coincidence!) and the bottom half of the abstract and much of the highlights are not justified from looking at Figure 7. The data could possibly be explained if the January data were excluded provided other processes dominated in January and are explained or at least suggested? Can you do some statistics on that to better explain the hydrodynamics of the microplastic particles in different seasons?

In the text (lines 804-811 in Results section, lines 899-916 and 1082-1095 in Discussion section) and in Table 3 the peculiar features of January were reported and explained. The occurrence of exceptional wind events is, unfortunately, not rare on the Ligurian coast. The January information has the same value of the other months and, in our opinion, it has to be treated together with the other data in the statistical analysis, because it represents a scenario that can occur.

Figure 7 includes January data, in the A and B panels. The observations are indicated by grey circles, as reported in the legend. We choose the multivariate analysis because in the environment the trend of a single variable is often associated to a number of forcings and not only to one. Therefore, this is the best way to discover significant relationships between station and/or variables.

Classical univariate analysis (such as correlation) was performed and the significant results reported in lines 770-775. They are rather few, indicating that, actually, the microparticle distribution depends on more forcings acting at the same time or, eventually, on variables that were not recorded in the present study. To test whether the environmental variables we recorded were responsible of the station features in terms of microparticle content of the sediment, we performed the multivariate RDA. As reported in Material and Methods section, in this case the microparticle features of each observation are response variables, and the environmental features (such as permeability and waves) explanatory variables.

Fig. 7A is the plot related to fibres, Fig. 7B to fragments and spheres. In the first case the January observations are scattered as the July ones, while April showed a higher similarity between stations. In the second panel, instead, December station 1 was very different from the other December stations. It means that the beach sites may be alternatively different or similar depending on the microparticle type. We think that the point is that, notwithstanding the rather scattered observations, the entire multivariate analysis shows that permeability and wave direction significantly explain the microparticle distribution. It would be more surprising if all the observations were always grouped by month. And also alarming, because it would mean that the microparticle input to the beach, different for each month, is so high to mask totally the morphodynamic differences of the sites.

Actually, the Fig. 7 has a statistical value only if linked to Tables 4-7. We can delete the Fig. 7 if it can bring the reader to misunderstand what the text reported. However, given that the significant differences (or similarities) between stations have been already reported describing and discussing the single parameters, we added few sentences in the Discussion section, as requested by the Reviewer. We highlighted that also in the plots the stations grouped depending on the microparticle abundance and

that, especially for fibres, the relationship with permeability was not only statistically but also visually rather clear.

Highlights to:

“Hydrodynamic forcing and sand permeability influence the distribution of anthropogenic microparticles in beach sediment”

by C. Misic, A. Covazzi Harriague, M. Ferrari

- Anthropogenic microparticles (Mps) were studied on a NW Mediterranean beach
- Mps abundance (416±202 Mps kg^{-1}) and distribution were related to environmental characteristics
- Fibres were the main Mps morphology (>90%)
- Hydrodynamism (wave characteristics, rip current) and sediment permeability influenced fibre distribution
- Fragment accumulation depended on the efficiency of waves at removing stranded materials

Hydrodynamic forcing and sand permeability influence the distribution of anthropogenic microparticles in beach sediment.

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Abstract

The distribution of anthropogenic microparticles (Mps), such as plastic and natural fibres used in textiles, in beach sediments was studied in a human-influenced pocket beach in Liguria (NW Mediterranean Sea). Information on environmental parameters such as rainfall, hydrodynamic characteristics and sediment texture was collected at the same time as the sediment samples. The Mps (416±202 Mps kg^{-1} on average) were mainly fibres (57-100%), while fragments and spheres showed irregular abundances linked to the draining action of waves on the beach. Uni- and multivariate statistical analyses highlighted that the different spatial and seasonal distribution of fibres primarily depended on the action of the waves that force seawater into the sand, rather than on sedimentation following depositional processes. Wave height and direction had a role in fibre distribution in the sand, as well as sediment permeability and sorting. The occurrence of short-term and spatially-localised hydrodynamic events such as rip currents were observed to influence the abundance of fibres, overlapping the seasonal sequences of beach accretion and erosion that is typical of the area and increasing fibre abundance by transporting those accumulated in the sediments of the submerged beach during winter.

Key-words: Anthropogenic microparticles, Beach sediment, Hydrodynamic characteristics, Sediment texture, NW Mediterranean.

1. Introduction

The diffusion of microparticles (Mps) of anthropogenic origin in the marine environment is a highly debated topic, especially with respect to what composes microplastics (i.e., plastic particles ranging from 20 μm to 5 mm, Barnes et al., 2009). According to the scientific literature, primary and secondary microplastics can be distinguished as follows. The first type includes items produced to be small, such as microbeads derived for personal-care products (Bergmann et al., 2015), precursors or by-products of plastic production (Costa et al., 2010), sandblasting, and industrial-cleaning products (Browne, 2015). The second type includes small items (fragments, fibres, films, etc.) coming from the deterioration, weathering and photo-degradation of macroplastics that were used or abandoned in the environment (Andrady, 2011; Barnes et al., 2009; Claessens et al., 2011; Hidalgo-Ruz et al., 2012).

Natural materials used in textiles are another source of anthropogenic Mps. They can be confused with common microplastic fibres and hold similar environmental concerns as both have been found to adsorb chemical pollutants (Ladewig et al., 2015). Information related to the distribution at sea of these materials is nearly absent (Song et al., 2015), although they can reach the environment following the same pathways as synthetic fibres before biodegradation takes place. In fact, fibres of natural origin, such as cotton and wool, are biodegradable (Chen and Burns, 2006; Ladewig et al., 2015), and they may achieve a rather high importance; in the seawater surface they may account for up to 85% of the total fibre abundance (e.g., the coastal area of South Korea, Song et al., 2015). The principal input of anthropogenic-derived fibres is due to the machine-washing of natural and synthetic textiles that enter into wastewater, yet cannot be retained by sewage treatment plants (Desforges et al., 2014). Both natural and synthetic materials that adsorb dissolved pollutants may have deleterious effects (Ladewig et al., 2015), but they may also be a threat because of the chemicals that are used to improve performance and dye textiles (Chen and Burns, 2006). Recent scientific literature concerning microplastics reports their influence on both individual organisms and food webs (Rochman et al., 2013). The small size of Mps facilitates intake by organisms

(Lusher et al., 2013; Remy et al., 2015). Thus, planktonic and benthic organisms may ingest particles that accumulate in the water column and sediment (Deudero et al., 2014; VanCauwenberghe et al., 2015; Wright et al., 2013). The consequence of ingestion of anthropogenic-derived items could be the uptake of additives such as phthalates used to enhance plastic performance and/or the uptake of adsorbed persistent organic pollutants (Teuten et al., 2007). Other general negative impacts may be f mechanical in nature, such as suffocation or reduction of feeding activity (Lusher, 2015).

Mps have been found in all environmental components, from the water column to the sediment. Items denser than water may sink due to their physical and chemical features (Woodall et al., 2014), but biofilm accrual often reduces the buoyancy of materials lighter than water, making them sink as well (Andrady, 2011; Jorissen, 2014; Reisser et al., 2013; Zettler et al., 2013). Furthermore, wind and currents may carry the items far from their production or site of last-use (Suaria and Aliani, 2014), thus contaminating rather pristine environments (Alomar et al., 2016). Data related to plastics in the sediment have been published since the beginning of the 21st Century (Sanchez-Vidal et al., 2018; Thompson et al., 2004), but few have dealt with coastal sites (Alomar et al., 2016; Graham and Thompson, 2009; Palatinus et al., 2019). Little is known about seasonal variations and temporal trends of natural fibres and microplastics on beaches (Browne et al., 2011; Song et al., 2015; Stolte et al., 2015), although it has been estimated that microplastics contribute between 8 and 40% of the total weight of plastic in beach sediments on the Belgian coast (Van Cauwenberghe et al., 2015).

Despite the rising number of published studies over the last decades, many marine areas worldwide have not been extensively studied, including zones where a high abundance of anthropogenic Mps is documented such as the Mediterranean Sea (Fossi et al., 2017; Suaria et al., 2016). The Ligurian coast (NW Mediterranean Sea) hosts a large variety of human activities that are sources of Mps. A clear influence of these items on the marine ecosystem has not been highlighted yet, but potential threats of this kind of pollution should not be ignored, especially in densely populated areas such as the eastern Ligurian Riviera.

As observed for macrodebris (Browne et al., 2010), small-scale hydrodynamic characteristics of coastal areas may regulate the ability of Mps to enter the sediment. Therefore, local features such as coast exposure, sea-storm frequency and intensity, rip current formation, and presence of groynes and breakwaters may play a role in the accumulation of Mps in beach sediment. In order to investigate the influence of these physical and morphological features, we performed a seasonal sampling of sediment in a typical Ligurian pocket-beach, whose hydrodynamic and morphometric characteristics have been previously studied (Schiaffino et al., 2015). Environmental data were collected at the same time. Due to the extraction protocols used to isolate the anthropogenic Mps - employed for the microplastic extraction and described below - our data primarily related to plastic items. Nevertheless, the lack of confirmation by Fourier transform infrared spectroscopy (FT-IR) of the actual composition of these items and the possibility that some samples may contain fibres derived from natural material used in textiles, led us to call all our items Mps, even thou they are potentially only plastic.

In this study, we focused on sea wave features (i.e., wave direction and wave height) to determine if short-term and low-energy wave action on sampling days exerted an influence on Mps abundance and distribution in the sand, and whether this influence was different from the longer-term, high-energy wave action associated with sea storms. We also studied on the mean grain size, homogeneity, and permeability of the beach sediment, to determine if these sedimentary characteristics were related to the penetration of the Mps in the sediment.

2. Material and Methods

2.1 Study area and position of the stations

The Levanto beach is located on the eastern Ligurian coast (44°10.18' N, 9°36.73' E) (Fig. 1). The Levanto beach is a NNW-SSE oriented pocket beach delimited by two promontories and representing the seaward extension of the Ghiararo Creek catchment. The annual mean rainfall during the period 1932-2016 was 1050.4 mm as measured at the Levanto weather station. The coastline displays wave conditions typical of Liguria, with the most severe storms surging from the south (Ferrari et al., 2006). The most frequent and intense storms come from the southwest. The Levanto beach is a micro-tidal area with a maximum tidal excursion of about 30-40 cm (Schiaffino et al., 2015).

The Levanto beach extends for approximately 800 m and its width ranges from 35 to 50 m (Fig. 1). It is delimited, on its backside, by a continuous wall and touristic facilities. In its central section, this wall is interrupted to allow the Ghiararo Creek to flow to the sea. This is a very short, ephemeral water course and its water was not visible on the beach surface during the sampling period.

Some artificial structures protect the beach from wave action. In the central sector, two groynes delimit the beach portion that contained Stations 1, 2 and 3. This sector has experienced minor erosion in recent years (Schiaffino et al., 2015). In front of this part of the beach, approximately 65 m far from the shore, submerged and detached breakwater approximately 100-m in length was built to further protect the beach.

Station positions were located in order to determine whether the aforementioned local hydrodynamic characteristics and potential environmental sources have a role in the Mps content of the beach sand. Station 1 was placed at the creek outlet, whereas the Station 3 was placed on the opposite side where, during periods of bad sea conditions (when waves come from the southwest), a rip current is generally observed due to the interaction of the waves and the southernmost groyne. Station 2 was located in the middle of the offshore breakwater. Station 4 was placed south of the southernmost groyne, next to the outlet of a very small channel (less than 0.5 m wide) that presents a low but rather constant discharge. Station 5 was only sampled twice. It was placed on the southernmost part of the beach, in a site characterised by very different hydrodynamic conditions, due to the shelter of the existing external seawall and rocky peninsula.

Besides the structures built at sea and along the backside of the beach, and the intense use of the beach for bathing and sunbathing, other anthropogenic pressures are present in the area. In the northern part, the touristic harbour is highly frequented by small boats. A sewage treatment plant is located in the immediate outback, discharging north of the harbour. Beach replenishment in the central area (Stations 1-4) occurs nearly every year. In 2017, it began in May before the beginning of the touristic season, although the beach is frequented by tourists all year long. The sand used for replenishment is of fluvial origin and is washed and analysed before placement to avoid the presence of contaminants (http://www.comune.levanto.sp.it).

2.2 Sampling

The sampling took place during 2017by season, i.e., January (winter), April (spring), July (summer), and December (late autumn). Sation 5 was sampled only in July and December.

The sediment samples were collected at the watermark of the higher wave for each survey date following Claessens et al. (2011). Sediment was picked up with stainless-steel cores of 4 cm diameter and carefully cleaned with prefiltered (0.45 µmpore size) deionised water. The sediment was sampled from the surface to a depth of 5 cm (as recommended by the Joint Research Centre of the European Commission, 2013) and the samples were placed into clean glass containers, to be treated as described below. Four replicates were collected at each station, as in Thompson et al (2004), with an overall mean weight equal to 123±19 g for every sediment sample. The total sediment volume of 63 ml per replicate was similar to the 50 ml of sediment collected for each replicate by Browne et al. (2010) in a study of microplastic spatial patterns along the shoreline of the Tamar Estuary (UK).

Data concerning rainfall and waves were recorded by the Meteo-Hydrological Observatory of the Regione Liguria (OMIRL, http://www.arpal.org.it). Furthermore, sea conditions, consisting of wave height and direction, were recorded on the sampling day, for the latest sea storm, and for the week before sampling.

The wave attack from SW on the beach (mean wave direction observed in Levanto) was modelled using Xbeach. This is a two-dimensional model that simulates wave propagation and its mean flow (Roelvink et al., 2009). The simulated wave attacks are representative of calm and stormy conditions (i.e. Hs registering around 0.2 m for calm-sea conditions and Hs registering about 2.0 m for those of stormy sea conditions). The computational grid has been generated by merging bathymetric field data collected in 2015 and the data related to deeper bathymetry supplied by the Hydrographic Institute of the Italian Navy.

2.3 Sample treatment and analysis

2.3.1 Mps extraction and analysis

The equipment used for sample processing was washed with diluted HCl and rinsed with prefiltered (0.45 μm) deionised water; all working surfaces were cleaned with alcohol. A white lab coat was always worn while analyses were performed.

Sample analyses were based on the protocols described by Thompson et al. (2004), Hidalgo-Ruz et al. (2012), and the Joint Research Centre of the European Commission (2013) for microplastics, because no protocol was available in the literature for the extraction of natural fibres from sediment. However, the Joint Research Centre of the European Commission (2013) considered that the analytical approaches described are likely to capture other man-made particles.

Mps were extracted by density separation. A saturated solution of NaCl (density 1.2 g cm^{-3}, Thompson et al., 2004) and $ZnCl_2$ (density 1.6 g cm^{-3}, Imhof et al. 2012) was added to each sample. Each solution was prefiltered (0.45 μm) to remove any particles that may contaminate the sample. The NaCl solution was added to the sediment twice (200 ml + 200 ml), whereas the $ZnCl_2$ only once (200 ml), resulting in a three-step extraction. After each addition, the sediment was shaken for 5 minutes; after 10 minutes, the supernatant was transferred to a glass container. Next, 100 ml HCl (Desforges et al, 2014) was added to the sample in order to digest residual organic material. The concentration of HCl was 37%, therefore the final concentration was below 5%. This concentration allowed the samples to be cleaned and analysed. We tested the effect of this HCl concentration on textiles, and no change of colour or damage was observed, but a bleaching of the coloured fibres cannot be excluded *a priori*. The action of hydrogen peroxide (H_2O_2), whose use has already been suggested by other authors such as Stolte et al. (2015), was not enough to digest the vegetal debris sometimes found in samples.

After 24 h digestion, the samples were filtered through mixed cellulose-ester filters (47 mm, 0.45 µm pore size). This procedure was performed under vacuum filtration by using a glass filtering apparatus and rinsing carefully the containers with prefiltered (0.45 µm) deionised water.

The filters were placed in clean glass Petri dishes. Control samples, represented by filters subjected to the same conditions as the samples under investigation but without sediment, were made every time to evaluate laboratory background contamination expected from airborne particles. Each filter was carefully inspected under a dissecting stereo microscope (Leica EZ4) with 30x magnification. The Mps were identified according to morphological characteristics and physical properties (e.g., response to physical stress, whether they were bendable or soft, and colour) as reported in Hidalgo-Ruz et al. (2012). Furthermore, they were counted and categorized into three broad categories: fibres, fragments (including fragments and films), and spheres (i.e., pellets and all items that showed a spherical shape). The dimension of some representative fragments and spheres and the length of some typical fibres (e.g., the transparent ones) were determined with a Leica DFC290 stereomicroscope (magnification 90x) and associated Leica Application Suite (LAS) software to image acquisition and elaboration. Due to the very small size (identified fibres were substantially less than 2 mm in length, often approximately 0.1 mm long; the longest were too thin to be analysed properly), it was not possible to analyse them with FT-IR spectroscopy and distinguish between natural and synthetic materials.

Fibres were counted and divided by colour (transparent, black, blue, red, and violet), whereas fragments and spheres were only counted. After subtracting the control values, the values were expressed as Mps kg^{-1} dry weight sediment.

2.3.2 Sediment characteristics

The sediment of each replicate, after extraction, was washed using freshwater to remove inorganic salts, and then placed in a furnace (105°C) to dry for 24 h. Subsequently, the sediment texture was investigated. A stack of stainless-steel sieves was used, from 0.063 to 16 mm (i.e. from -4 phi to +4 phi) mesh size. The sediment in the stack was shaken for 5 minutes and the sediment retained in each sieve was weighed (Denver APX200 balance, d=0.1 mg) for the definition of the grain size.

Mean grain size, permeability, and sorting were calculated as follows:

Mean grain size (mm) = $(phi_{16}+phi_{50}+phi_{84})/3$

where phi_{16}, phi_{50}, and phi_{84} represent the dimension of the 16^{th}, 50^{th}, and 84^{th} percentile (Folk and Ward, 1957)

Permeability (cm s^{-1}) = $(phi_{90})^2 * c$

where phi_{90} represents the dimension of the 90^{th} percentile of retained sediment and c a coefficient equal to 0.011(Hazen, 1911); and

Sorting (phi) = $(phi_{84}-phi_{16})/4 + (phi_{95}-phi_{5})/6.6$

where phi_{95}, phi_{84}, phi_{16}, and phi_{5} represent the dimension of the 95^{th}, 84^{th}, 16^{th}, and 5^{th} percentile (Folk and Ward, 1957).

The sediment organic content was also investigated. Protein and carbohydrate contents were determined following the spectrophotometric methods of Hartree (1972) and Dubois et al. (1956) using albumin and glucose solutions to calibrate the Jasco V-530 spectrophotometer.

2.4 Statistical analyses

A two-tailed *t*-test was used to verify the differences between stations in the same sampling for the same variable, and a paired *t*-test was used to verify the differences among samplings over time. Moreover, a Spearman-rank correlation was used to test the significance of the relationships among variable trends (STATISTICA software). The redundancy analysis (RDA) (Zuur et al., 2007) was applied on normalised data to verify the influence of the environmental variables on the total abundance and colour of fibres and on the abundance of fragments and spheres using a Brodgar 2.5.6 package, 2011(Highland Statistics Ltd.). RDA is a form of constrained ordination that examines how much of the variation in one set of variables explains the variation in another set of variables. RDA is a direct gradient analysis technique which summarises linear relationships among components of response variables (in this case the features of Mps) that are explained by a set of explanatory variables (normalised mean grain size, permeability, carbohydrate and protein content, and protein/carbohydrate ratios). The sampling season and the wave direction on the sampling day and during the latest sea storm were nominal explanatory variables (ranked 0 or 1). Moreover, to test the order of importance of the explanatory variables an automated forward selection model was applied. First, the "marginal effects", i.e., the variance expressed by one explanatory variable only, were calculated. Then the "conditional effects" that showed the increase in the total sum of eigenvalues after including a new variable during the forward selection, were calculated. Finally, a permutation test was applied (number of permutations: 499) in order to test the null hypothesis, i.e., the explained variation is larger than a random contribution

3. Results

3.1 Mps morphology

In the sediment of the Levanto beach, a relevant dominance of fibres was observed. In most of the samples they represented more than 90% of the observed Mps and, when present, fragments were generally more abundant than spheres. The highest contribution of fragments and spheres to the total (41 ± 3%) characterised the January sampling at Stations 3 and 4 (Table 1). Except for these two stations (where fragments reached values up to 209±190 fragments kg^{-1}), fragments and spheres contributed 11 ± 3% during the January and April samplings (27±8 and 31±5 fragments kg^{-1}, 4 ± 3 and 1 ± 1 spheres kg^{-1}, respectively), whereas their contribution dropped to 1 ± 1% (ranging from 0 to 3%) in the July and December samplings. Fragment and sphere dimensions ranged between 0.05

and 0.5 mm. Fibres showed variable lengths and morphologies, as described in Section 3.2. No significant relationships were found between the abundance of fibres and the overall quantity of fragments and spheres at the same station and considering different seasons.

3.2 Fibre distribution and morphology

Fig. 2A reports the total abundance of fibres for each station on each sampling date. The same station registered rather different values on the four sampling dates. For instance, in the January sampling, Station 4 showed the lowest value recorded (124 ± 26 fibres kg^{-1}), which was significantly lower than the values registered at Stations 1 and 3 on the same sampling date. On the following sampling dates, the abundance of fibres increased, reaching a maximum value in December (563 ± 51 fibres kg^{-1}), when it was higher than the values registered at the other stations (significantly higher compared to Station 5, Fig. 2A). Station 1 showed the highest value in July (893 ± 109 fibres kg^{-1}), which was significantly higher than Stations 3 and 5 on the same sampling date (Fig. 2A). However, in the other samples the values were below 400 fibres kg^{-1} (significantly lower than Station 3 in January). Among the stations sampled four times (Stations 1 through 4), Station 3 reported the most stable values, ranging from 208 ± 98 fibres kg^{-1} in April to 499 ± 118 fibres kg^{-1} in December.

Among the fibres analysed, the transparent ones (Fig. 2A) were the most abundant in all of the samples (80 ± 11%). January samplings showed the lowest contribution (68 ± 12%, Stations 1-4) and July showed the highest (90 ± 7% at Stations 1-4 and 85 ± 11% at Station 5).

The coloured fibres (Fig. 2B) also showed variable values at the same sites. Nevertheless, the most abundant fibres were blue, contributing an average of 53 ± 18% for all the samplings, followed by black (20 ± 13%), violet (16 ± 16%), and red (11 ± 9%). The highest contribution of the coloured fibres to the total abundance was noticed in January (32 ± 12%); it was higher than the values registered in July (15±11%), December (16 ± 6%) and April (20 ± 10%).

Fibres were grouped into three classes depending on their length: small (from 0.1 to 0.5 mm), medium (from 0.5 to 2 mm), and large (longer than 2 mm). Small fibres (averaging 304 ± 188 fibres kg^{-1}) were significantly more abundant than medium (averaging 56 ± 39 fibres kg^{-1}) and large (averaging 25 ± 19 fibres kg^{-1}) fibres (t-tests, $p < 0.001$). Fig. 3 shows the contribution of each class grouped by colour (dark: blue + black, light: red + violet, and transparent). Temporal differences were observed for the small transparent and violet fibres. They were more abundant in samples taken in July and December, than in January and April (t-test, $p < 0.01$ and $p < 0.05$, respectively). The transparent fibres of medium and large length had a cylindrical section, whereas the small transparent fibres had flattened sections and were more curled than the large ones. Dark coloured fibres had a cylindrical section as well as the smallest ones. The edges were net-cut in the cylindrical fibres, whereas they were irregular in the flattened ones.

3.3 Sediment features

The sediment texture (Table 2) showed a rather high variability. Stations 1 and 2 showed the higher mean grain size (4.9 ± 2.1 mm and 3.4 ± 2.0 mm, respectively). Station 5 showed the finest grain size (0.4 ± 0.0 mm). The highest value was recorded in Station 1 in December, the lowest in Station 5 in July.

The sediment permeability (Table 2) was higher in January (3.17 ± 2.67 $*10^{-2}$cm s^{-1}) and July (2.67 ± 2.49 $*10^{-2}$ cm s^{-1}) than in April (0.47 ± 0.44 $*10^{-2}$ cm s^{-1}) and December (0.67 ± 0.67 $*10^{-2}$ cm s^{-1}). The highest mean value was recorded at Station 1 (3.33 ± 2. 70 $*10^{-2}$ cm s^{-1}), and the lowest at Station 5 (0.04 ± 0.02 $*10^{-2}$ cm s^{-1}).

The sediment sorting ranged between 0.73 ± 0.05 phi (moderately sorted) measured at Station 5 in July and 2.12 ± 0.48 phi (very poorly sorted) at Station 1 in December (Table 2). All the stations in January and April showed a poorly-sorted sediment, whereas higher variability and a dominance of moderately-sorted sediment was observed for July and December. Stations 1 through 4 showed similar mean values (from 1.29 ± 0.31 phi for station 3 to 1.36 ± 0.53 phi for station 1), and Station 5 (sampled only in July and December) showed the lowest value (0.78 ± 0.07 phi).

The sediment-permeability trends showed a significant and direct correlation with the abundance of fibres ($r = 0.58$, $n = 18$, $p < 0.01$), whereas the sorting resulted in significant and inverse correlation to the abundance of fibres ($r = -0.48$, $n = 18$, $p < 0.05$).

The organic matter content showed a general dominance of carbohydrates over proteins (Fig. 4). The highest values were recorded in April, (on average 89.5 ± 75.0 µg g^{-1} for carbohydrates and 19.9 ± 13.2 µg $g^{-1}$for proteins). They were higher than those documented in January (averaging 15.2 ± 10.3 and 5.6 ± 5.7 µg g^{-1}), July (on average 24.8 ± 16.2 µg $g^{-1}$and 16.7 ± 13.3 µg g^{-1}) and December (averaging 18.7 ± 6.8 µg g^{-1} and 11.9 ± 8.1 µg g^{-1}). The correlation between carbohydrates and proteins were significant ($r = 0.64$, $n = 18$, $p < 0.001$), but the organic matter did not show any correlation with the abundance of fibres, fragments or spheres.

3.4 Environmental features

The mean wave height, the significant wave height, and the wave direction were recorded for the day of sampling, for the latest sea storm, and for the week before the sampling (Table 3, Fig. 5A and 5B). During sampling, the wave height was generally low, except for July when the significant wave reached a value of 0.91 m. The lowest wave height was documented in January (lower than 10 cm). It was due to an exceptionally strong wind from 46°N (http://www.arpal.org.it) that completely flattened the sea on the shore, suppressing wave action. Except for January, the direction of the waves on the sampling days was from southwest.

Values of the mean wave height relate to the week before sampling, except the sea storm day. The values were similar to those on the respective sampling days (in April and July, December showed higher height) and the directions overlapped. Again, January showed different characteristics. The week before sampling, they were clearly divided into two very different conditions (Table 3).

Sea storms generally occurred 2 to 4 days before samplings. The wave direction was similar for January and July, while the sea storms of April and December came from the east (108°N) and southeast (122°N), respectively. Wave heights on storm days were higher than 2 m (January and December, Table 3) and showed lower values (1.50 ± 0.30 m) in July. The mean wave height in April had the lowest value for any storm day (less than 1 m).

The wave propagation models (Fig. 6) in both simulated conditions have not shown significant energy changes along the shoreline from Station 1 through Station 4. Only near station 5 was the wave height close to zero. Rip currents were observed. In stormy sea conditions (Fig. 6A) two rip currents were observed adjacent to the central groyne. Station 1 was located very close to the groyne and was not directly influenced by these water movements. A third rip current, of weak intensity, was observed near station 5. A strong rip current was present near station 3. The rip current placed south of Station 1 was also present, although very weak, in calm-sea conditions (Fig. 6B).

The year 2017 was rather dry, with a total rainfall slightly higher than 750 mm vs approximately 1050 mm for the period 1932-2016 (Levanto weather station). The cumulative rainfall registered over the 30 days before each sampling was 15 mm (0 - 10 mm per day) for January, 25 mm (0 - 10 mm per day) for April, 14 mm (0 - 9 mm per day) for July, and increased slightly in December for a total of 73 mm, ranging between 0 and 22 mm per day.

3.5 Multivariate statistical analysis

The RDA showed that the influence of the environmental features was different for fibres and for fragments and spheres. Fig. 7 reports the graphical outputs of the RDAs.

The RDA applied on fibres explained a rather high part of the variance (0.56, axis 1 explaining 29% of the variance and axis 2 13%, Table 4) and showed that sediment permeability and wave direction (during the day of sampling and during the latest sea storm) significantly influenced the distribution and abundance of fibres over the beach (Table 5).

The RDA applied on fragments and spheres explained 0.53 of the variance (axis 1 47%, axis 2 6%,Table 6), but showed that only the wave direction during the sampling was significantly related to the distribution of Mps on the shore (Table 7).

4. Discussion

The fragment and sphere abundance and the fibre abundance in the different samplings showed relevant differences. However, no significant relationships were found between these two groups, indicating that these Mps types may have mixed with the beach sediment following different processes (Browne et al., 2011; Stolte et al., 2015). Therefore, we analysed them separately.

In January, the fragment and sphere abundance was rather high, with the highest fragment abundance at Stations 3 and 4. Also macro-litter (such as packaging, ropes, funnels and highly degraded plastic items) was found at Stations 3 and 4. This macro-litter accumulation was not observed in the other samplings. The differences between the months are rather clear in the RDA plot (Fig. 7B), where the first axis explains 47% of the variance and the January sites have been separated from the observations made in the other months. The RDA highlighted that the abundance of fragments and spheres may be significantly influenced by the action of waves on the sampling day. Actually, wave direction and height on sampling days impeded accumulation. In fact, in January, the strong northeast wind event that started about two days before sampling led to negligible waves that did not have the force to drain stranded debris to sea, causing the relevant abundance of fragments and spheres found.

The higher abundance of fragments and spheres in April than in December, considering that the wave direction and height were similar, depended on the wave heights that occurred before the two samplings. In fact, we observed lower values for wave height in April for both the regular regime and in the sea-storm regime the week before each sampling (Table 3), while December showed higher wave heights. The draining of fragments and spheres by the waves in the days before the April sampling was less efficient than that of December. Part of these fragments and spheres may have derived from those observed in January. On the whole, the abundance of fragments and spheres was four times lower in April than in January, while no macro-litter was observed at Stations 3 and 4 in April. Different patterns in the distribution and characterisation of micro and macro-debris were previously observed by Browne et al. (2010), mainly related to wind action and the density of the materials.

The RDA highlighted that fibre distribution and abundance were, instead, influenced by the sediment permeability, the wave direction of the previous sea storms and the wave direction of the sampling day. The distribution of the observations in the RDA plot reported in Fig. 7A points to the significant relationship between the fibre abundance and permeability, already indicated by the univariate analysis. The July stations showing the highest fibre abundances are grouped on the left side of the first axis, where permeability presented the highest scores, while the samplings showing the lowest fibre abundances (April observations and station 4 in January) are, instead, on the right side of the axis, opposite to the permeability vector.

Huettel and Gust (1992), with their experiments in stirred chambers, have shown that penetration in the sediment of small items such as algal cells depended on sediment permeability and dynamic processes, such as water flushing. In our study, higher fibre abundance was found in the most permeable sediments. The penetration of Mps into the sediment was influenced by grain size, sorting and state of consolidation of sediments (Rusch and Huettel, 2000). In the Levanto beach, the inverse correlation between abundance of fibres and sorting values indicated that fibres accumulate mainly in homogeneous sediments. Station 5, although it was sampled only twice, showed the lowest permeability, finest grain size, and low abundance of fibres. The wave height of this area was generally low. The sheltered conditions allow for depositional processes of finer sediment grain sizes but does not for fibres. This observation and the previous statistical evidence supported the hypothesis that wave action led to a forced flushing of water into the sand, which may play a relevant role in the distribution of fibres inside the beach.

Waves were identified as an important parameter used in determining the fate of microparticles in the beach sediment. The waves were not uniform along the sampling sites because some Stations were protected by groynes and the submerged breakwater. In addition, wave direction at Levanto was primarily observed as coming from SW (Fig. 6). Therefore, we processed the hydrodynamic data in order to obtain a schematic diagram of the wave energy as well as direction along the shore in case of waves arriving from SW. The model results for low height of waves indicated the occurrence of a low-energy rip current in the area of station 1, possibly reducing the wave efficiency and thus also lowering the introduction in the sediment of the allochthonous particles floating into the water, as plastic ones. This current may have had a role in samples that were mainly taken in April and December, when the sampling-day wave on the shore had a mean height of 0.20±0.02 m and 0.30±0.05 m, respectively. In the April sampling, the fibre distribution for the area, which was influenced by the two groynes (stations 1 through 4) and partially protected by the submerged breakwater (stations 1 through 3) showed slightly lower values at Stations 1 and 2 than at Station 3, with an increase at Station 4. In December, the trend was similar; although the fibre abundance was higher. This difference may have different explanations, one of which could be related to previous hydrodynamic events such as the height of the storm-day waves that was lower for the month of April (0.90±0.27 m) than for that of December (2.12±0.15 m).

In July, the wave direction was registered as coming from the SW during the previous sea storm cycle and during the sampling, when we recorded the highest wave height on the shore (on average 0.85±0.07 m). Waves characterised at this height were known to interact with the submerged breakwater generating the transport of sediment on the beach and a strong rip current originating from the area of Stations 3 and 4. This was illustrated by the model output for high energy waves coming from SW and has also been reported in previous studies. Given the wave direction and the orientation of the groynes, Schiaffino et al. (2015) observed that wave heights approaching 1 m resulted in the selective erosion of the beach line in specific places, due to the temporary rip currents that moved sediment offshore. Therefore, in this case it was not a matter of a weak current affecting the particle penetration in the sediment, but sediment actually being physically removed. This has led to a lower abundance of fibres at Stations 3 and 4 than at the Stations 1 and 2. The rip current eroded some centimetres of sand, exposing the layers below. Jackson et al. (2014) showed that waves, together with tide movements, had a role in burying or re-suspending particles, such as invertebrate eggs, embedded in the sediment. The reworking of sediment reached 3 to 4 cm in the sediment, depending on the combined action of wave and tidal movement.

In our study, the rather high mean grain size of Stations 1 through 4 implied that fibre penetration in the sediment was not a matter of the top mm, but water flushing and particulate material transport occurred over several cm. This was possible, at least in Stations 1 through 4, where the mean grain size was higher than 2 mm, as observed previously for particulates in a neighbouring Ligurian pocket beach (the Baia Blu Beach, located 25 km eastward and having the same exposure and texture) (Misic and Covazzi Harriague, 2007). The ability of Mps such as microplastics to penetrate the beach sediment was previously observed by Carson et al. (2011), who found microplastics down to 25 cm in the sand, although 50% was observed in the topmost 5 cm. Therefore, the fibre contamination at Station 3 may be the result of previous infiltration processes and/or the transport in the deeper layers due to the waves, facilitated by the coarse grain size.

In the case of higher waves (reaching offshore heights of nearly 3 m), the influence of submerged breakwater was clear in the model output, where the area facing station 3 showed an increased wave energy compared to the other Stations. Station 5, due to its sheltered position, did not demonstrate these variation of wave energy and influence because the energy that was sustaining particle penetration in the sediment always remained low, and therefore it showed low fibre abundance.

Also, the January sampling was preceded by a sea storm from a southwest direction, but the following distribution of the fibres on the beach had a different pattern, with the highest value in Station 3 that, in July, showed the lowest fibre abundance. This difference was due to the peculiar wind-wave conditions after the storm event (as indicated by the significant influence of the wave direction on sampling days highlighted by the RDA), that is the presence of the aforementioned exceptional northeast wind, which completely changed the wave direction, prevented the formation of the rip current and, consequently, resulted in a different fibre distribution.

Most of the fibres were transparent in every season and small with curled shapes, especially in July and December. A small dimension (often below 0.3 mm) was also common also in coastal sediments of Croatia in the Adriatic Sea (Palatinus et al., 2019). The modification of the mean abundance and dimension of fibres registered after the first two samplings may have a seasonal explanation. In particular, the sharp increase recorded in July was associated with the recreational activities on the beach (Stolte et al., 2015) and in the neighbouring touristic harbour, both of which are generally known to increase the release of microplastics into seawater (Andrady, 2011; Desforges et al., 2014). Harbour sediments were also considered as reservoirs for pollutants due to the structure of the harbour itself in that it reduces water exchange with the area outside of the harbour (Claessens et al., 2013). An increase in vessel movements in the summer may cause resuspension of sediments and recirculation of settled particles, which may escape the harbour structure and be transported along the coast towards SE, as indicated by the model output for waves coming from SW. This phenomenon was observed in similar Ligurian touristic harbours, that in summer showed higher turbidity and concentrations of particulate matter, previously accumulated in the sediment (Misic and Covazzi Harriague, 2009).

The coloured fibres in the Levanto beach were mainly blue, as recorded by Stolte et al. (2015) for the Baltic Sea beaches. As these authors observed, it is possible that dark-coloured particles are more visible than light-coloured ones to the observer, thus causing an underestimation of the abundance of some category of coloured fibres.

The beach spring replenishment carried out in May, that covered the resident sand, was ineffective at increasing Mps abundance because the allochthonous sand was washed and analysed before loading onto the shore to avoid the introduction of pollutants. On the other hand, it is known that fibres are more abundant at coastal sites than offshore because they derive from the drainage of municipal wastewater (Alomar et al., 2016; Desforges et al., 2014). Machine washing of clothes is a major source of fibres (Browne et al., 2011) that are discharged to the sea via pipes. At Levanto, the pipe was placed north of the touristic port, in an area where the littoral drift carries the inorganic particulate materials, and therefore, also Mps, from west to east (Schiaffino et al., 2015), allowing the released fibres to reach the shore or to sink into the sediment in front of the beach. Thus, water movement able to move submerged sediment and carry it to the coast may represent an input of Mps to the beach, given that deep sediments (Pham et al., 2014; Sanchez-Vidal et al., 2018; Woodall et al., 2014) and coastal sediments (Alomar et al., 2016) are sinks for anthropogenic Mps. Beaches experience seasonal variations related to wave energy (Short, 1979). In the Ligurian Sea during the winter months (characterised by high wave energy), the backshore is affected by a diffused erosive sequence of sediment, whereas in the summer (low wave energy), it is subjected to a depositional sequence. It is possible that the July sampling recorded general accretion on the beach of sediment and fibres, whose transport was promoted by the morphodynamic evolution. The increase of smaller, curled, and frayed transparent fibres suggests that they may originate from the mechanical-abrasion process carried out on larger plastic items by waves in the submerged sediment during the aforementioned erosive sequence, a process that may develop for several months during the winter period.

Another hypothesis was that the fibres entered the beach carried by Ghiararo Creek water, whose mouth was next to Station 1. Its flowing water was not visible during samplings, but it may have had a feeble subsurface flow through the sand and gravel. In the Ligurian area the continental inputs are low and irregular, virtually absent during the summer. This was particularly true for 2017, which was characterised by a lower cumulative rainfall than the average value during the period 1932-2016. The January-July period was homogeneously dry, but the Mps abundance showed significant variations and reached rather high values, higher than those recorded in December after an increase of rainfall. These trends contrast with the observation that continental input may move large amounts of materials to the coastal area (Derraik, 2002).

A biological effect, i.e., the retention of Mps in organic materials suspended in seawater, was not likely, nor was an eventual chemical or mechanical binding of Mps with autochthonous organic material on the beach. In this study, we did not find any significant correlation between carbohydrate and protein content in sediments and the distribution of fibres or fragments. The sedimentary organic matter was related to natural biological processes, as the detected carbohydrates derived mainly from vegetal detritus which dominated on the protein content found in the sediment. In April, organic matter showed the highest values, primarily due to primary production processes in the seawater (Misic and Covazzi Harriague, 2008), whereas the Mps showed their lowest abundance.

5. Conclusions

As nearly all Ligurian beaches, the Levanto beach showed a strong anthropogenic influence, mainly due to urbanisation and touristic activities. The building of artificial structures, such as the groynes and the harbour, modified the natural sediment balance of the shore and introduced allochthonous materials such as the Mps we studied into the coastal system. Actually, the origin of the Mps was not determined, although for fibres, it is likely related to the input coming from the wastewater carried to the sea via a pipe located in the northern part of the study area. These fibres may accumulate in the submerged sediment all year along, and may be re-suspended and introduced to the beach sand during the summer via the natural accretion sequence characteristic of this season, with addition to vessel movements in the touristic harbour. The role of rainfall and of continental input was not observed, likely due to the very dry regime of the sampled year. The abundance and distribution of the fragments and spheres and of the fibres were mainly driven by hydrodynamic characteristics that in this area were modified by the anthropogenic structures built offshore and along the shore. This led to significant variations in the abundances of the Mps in the stations in the four samplings, despite the rather low extension of the beach. The Levanto beach is predominantly exposed to waves from the south. The southwest sector is the source of the most intensive and frequent storms, therefore it is possible that these conditions promote the preferential penetration of Mps in some sectors of the beach.

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References

Alomar, C., Estarellas, F., Deudero, S., 2016. Microplastics in the Mediterranean Sea: Deposition in coastal shallow sediments, spatial variation and preferential grain size. Mar. Environ. Res. 115, 1-10.

Andrady, A. L., 2011. Microplastics in the marine environment. Mar. Pollut. Bull. 62, 1596-1605.

Barnes, D.K.A., Galgani, F., Thompson, R.C., Barlaz, M., 2009. Accumulation and fragmentation of plastic debris in global environments. Philos. Trans. R. Soc. B 364, 1985–1998.

Bergmann, M., Gutow, L., Klages, M., 2015. Marine Anthropogenic Litter. Springer.

Browne, M.A., 2015. Sources and pathways of microplastic to habitats. In: Bergmann, M., Gutow, L., Klages, M. (Eds.), Marine anthropogenic litter. Springer, Berlin, 229-244. http://dx.doi.org/10.1007/978-3-319-16510-3.

Browne, M.A., Crump, P., Niven, S.J., Teuten, E., Tonkin, A., Galloway, T., Thompson, R., 2011. Accumulation of microplastic on shorelines worldwide: sources and sinks. Environ. Sci. Technol. 45, 9175–9178.

Browne, M.A., Galloway, T.S., Thompson, R., 2010. Spatial patterns of plastic debris along estuarine shorelines. Environ. Sci. Technol. 44, 3404–3409.

Carson, H.S., Colbert, S.L., Kaylor, M.J., McDermid, K.J., 2011. Small plastic debris changes water movement and heat transfer through beach sediments. Mar. Pollut. Bull. 62, 1708–1713.

Chen, H.-L, Burns, L.D., 2006. Environmental analysis of textile products. Cloth.Text. Res. J. 24, 248-261.

Claessens, M., Van Cauwenberghe, L., Vandegehuchte, M.B., Janssen, C.R., 2013. New techniques for the detection of microplastics in sediments and field collected organisms. Mar. Pollut. Bull. 70, 227–233.

Costa, M.F., Ivar do Sul, J.A., Silva-Cavalcanti, J.S., Araujo, M.C.B., Spengler, A., Tourinho, P.S., 2010. On the importance of size of plastic fragments and pellets on the strandline: a snapshot of a Brazilian beach. Environ. Monit. Assess. 168 (1-4), 299e304.

Derraik, J.G.B., 2002. The pollution of the marine environment by plastic debris: a review. Mar. Pollut. Bull. 44, 842-852.

Desforges, J.-P.W., Galbraith, M., Dangerfield, N., Ross, P.S., 2014. Widespread distribution of microplastics in subsurface seawater in the NE Pacifc Ocean. Mar. Pollut. Bull. 74, 94-99.

Deudero, S., Nadal, M.A., Estarellas, F., Alomar, C., 2014. Microplastic exposure in pelagic fishes and holothurians: a Mediterranean case study. In: 2nd International Ocean Research Conference. Barcelona 17-21 November 2014.

Dubois, M., Gilles, K.A., Hamilton, J.K., Rebers, P.A., Smith, F., 1956. Colorimetric method for determination of sugars and related substances. Anal. Chem. 39, 350-356.

Ferrari, M., Bolens, S., Bozzano, A.,Fierro, G., Gentile, R., 2006. The port of Genoa-Voltri (Liguria, Italy): a case of updrift erosion. Chem. Ecol. 22, 361–369.

Folk, R., Ward, W.C., 1957. Brazos River bar (Texas): a study in the significance of grain size parameters. J. Sediment. Res. 27, 3-26.

Fossi, M.C., Romeo, T., Baini, M., Panti, C., Marsili, L., Campani, T., Canese, S., Galgani, F., Druon, J.-N., Airoldi, S., Taddei, S., Fattorini, M., Brandini, C., Lapucci, C., (2017) Plastic debris occurrence, convergence areas and fin whales feeding ground in the Mediterranean Marine Protected Area Pelagos Sanctuary: a modeling approach. Front. Mar. Sci. 4, 167. doi: 10.3389/fmars.2017.00167

Graham, E.R., Thompson, J.T., 2009. Deposit- and suspension-feeding sea cucumbers (Echinodermata) ingest plastic fragments. J. Exp. Mar. Biol. Ecol. 368, 22-29. http://dx.doi.org/10.1016/j.jembe.2008.09.007.

Hazen, A., 1911. Discussion: dams on sand foundations. T. Am. Soc. Civ. Eng. 73, 199.

Hidalgo-Ruz, V., Gutow, L., Thompson, R.C., Thiel, M., 2012. Microplastics in the marine environment: a review of the methods used for identification and quantification. Environ. Sci. Technol. 46, 3060–3075.

Hartree, E.F., 1972. Determination of proteins: a modification of the Lowry method that gives a linear photometric response. Anal. Biochem. 48, 422–427.

Huettel, M., Gust, G., 1992. Impact of bioroughness on interfacial solute exchange in permeable sediments. Mar. Ecol. Prog. Ser. 89, 253–267.

Imhof, H. K., Schmid, J., Niessner, R., Ivleva, N. P., Laforsch, C., 2012. A novel, highly efficient method for the separation and quantification of plastic particles in sediments of aquatic environments. Limnol. . Oceanogr.: Methods 10, 524-537.

Jorissen, F.J., 2014. Colonization by the benthic foraminifer *Rosalina* (*Tretomphalus*) *concinna* of Mediterranean drifting plastics in CIESM 2014. In: Briand, F. (Ed.), Marine litter in the Mediterranean and Black Seas. CIESM Publisher, Monaco, p. 180. CIESM Workshop Monograph n. 46.

Joint Research Centre of the European Commission - MSFD technical subgroup on marine litter, 2013. Guidance on Monitoring of Marine Litter in European Seas. Luxembourg: Publications Office of the European Union,ISSN 1831-9424.54 doi:10.2788/99475

Ladewig, S.M., Bao, S., Chow, A.T., 2015. Natural Fibers: A Missing Link to Chemical Pollution Dispersion in Aquatic Environments. http://dx.doi.org/10.1021/acs.est.5b04754.

Lusher, A L., McHugh, M., Thompson, R.C., 2013. Occurrence of microplastics in the gastrointestinal tract of pelagic and demersal fish from the English channel. Mar. Pollut. Bull. 67, 94-99. http://dx.doi.org/10.1016/j.marpolbul.2012.11.028.

Lusher, A., 2015. Microplastics in the marine environment: distribution, interactions and effects. In: Bergmann, M., Gutow, L., Klages, M. (Eds.), Marine anthropogenic litter. Springer, Berlin, 245-307. http://dx.doi.org/10.1007/978-3-319-16510-3.

Misic, C., Covazzi Harriague, A., 2007. Enzymatic activity and organic substrates on a sandy beach of the Ligurian Sea (NW Mediterranean) influenced by anthropogenic pressure. Aquatic Microbial Ecology 47, 239-251.

Misic, C., Covazzi Harriague, A., 2009. Organic matter characterisation and turnover in the sediment and seawater of a tourist harbour. Mar. Environ. Res. 68, 227-235.

Misic, C., Covazzi Harriague, A., 2008. Organic matter recycling in a shallow coastal zone (NW Mediterranean): the influence of local and global climatic forcing and organic matter lability on hydrolytic enzyme activity. Cont. Shelf Res. 28, 2725–2735.

Palatinus, A., Kovač Viršek, M., Robič, U., Grego, M., Bajt, O., Šiljić, J., Suaria, G., Liubartseva, S., Coppini, G., Peterlink, M., 2019. Marine litter of the Croatian part of the middle Adriatic Sea: simultaneous assessment of floating and seabed macro e micro litter abundance and composition. Mar. Pollut. Bull. 139, 427-439.

Pham, C.K., Ramirez-Llodra, E., Alt, C.H.S., Amaro, T., Bergmann, M., Canals, M., Company, J.B., Davies, J., Duineveld, G., Galgani, F., Howell, K.L., Huvenne, V.A.I., Isidro, E., Jones, D.O.B., Lastras, G., Morato, T., Gomes-Pereira, J.N., Purser, A., Stewart, H., Tojeira, I., Tubau, X., Van Rooij, D., Tyler, P.A., 2014. Marine litter distribution and density in European seas, from the shelves to deep basins. PLoS One 9, e95839.

Reisser, J., Shaw, J., Wilcox, C., Hardesty, B.D., Proietti, M., Thums, M., Pattiaratchi, C., 2013. Marine plastic pollution in waters around Australia: characteristics, concentrations, and pathways. PLoS One 8, e80466. http://dx.doi.org/10.1371/journal.pone.0080466.

Remy, F., Collard, F., Gilbert, B., Compère, P., Eppe, G., Lepoint, G., 2015. When microplastic is not plastic: the ingestion of artificial cellulose fibers by macrofauna living in seagrass macrophytodetritus. Environ. Sci. Technol. 49, 11158–11166. http://dx.doi.org/10.1021/acs.est.5b02005.

Rochman, C.M., Hoh, E., Kurobe, T., Teh, S.J., 2013. Ingested plastic transfers hazardous chemicals to fish and induces hepatic stress. Sci. Rep. 3 (3263). http://dx.doi.org/10.1038/srep03263.

Roelvink, D., Reniers, A., Van Dongeren, A., de Vries, J. v. T., McCall, R., Lescinski, J., 2009. Modelling storm impacts on beaches, dunes and barrier islands. Coast. Eng. 56, 1133–1152.

Rush, A., Huettel, M., 2000. Advective particle transport into permeable sediments - evidence from experiments in an intertidal sandflat. Limnol. Oceanogr. 45, 525-533

Sanchez-Vidal., A., Thompson, R.C., Canals, M., de Haan, W.P., 2018. The imprint of microfibres in southern European deep seas. PlosONE 13 (11), e0207033.

Schiaffino, C. F., Dessy, C., Corradi, N., Fierro, G., Ferrari, M., 2015. Morphodynamics of a gravel beach protected by a detached low-crested breakwater. The case of Levanto (eastern Ligurian Sea, Italy). Ital. J. Eng. Geol. Environ. 1, 31-39.

Short, A.D., 1979. Wave power and beach stages: A global model. Proc. 16th Conf. Coastal Engineering, Hamburg, pp. 1145-1162.

Song, Y.K., Hong, S.H., Jang, M., Han, G.M., Rani, M., Lee, J., Shim, W.J., 2015. A comparison of microscopic and spectroscopic identification methods for analysis of microplastics in environmental samples. Mar. Pollut. Bull. 93, 202–209. http://dx.doi.org/10.1016/j.marpolbul.2015.01.015.

Stolte, A., Forster, S., Gerdts, G., Schubert, H., 2015. Microplastic concentrations in beach sediments along the German Baltic coast. Mar. Pollut. Bull. 99: 216–229

Suaria, G., Aliani, S., 2014. Floating debris in the Mediterranean sea. Mar. Pollut. Bull. 86, 494-504. http://dx.doi.org/10.1016/j.marpolbul.2014.06.025.

Suaria, G., Avio, C.G., Mineo, A., Lattin, G.L., Magaldi, M.G., Belmonte, G., Moore, C.J., Regoli, F., Aliani, S. 2016. The Mediterranean Plastic Soup: synthetic polymers in Mediterranean surface waters. Sci. Rep. 6, 37551. doi: 10.1038/srep37551

Teuten, E.L., Rowland, S.J., Galloway, T.S., Thompson, R.C., 2007. Potential for plastics to transport hydrophobic contaminants. Environ. Sci. Technol. 41, 7759–7764.

Thompson, R.C., Olsen, Y., Mitchell, R.P., Davis, A., Rowland, S.J., John, A.W.G., McGonigle, D, Russel., A.E., 2004. Lost at sea: where is all the plastic? Science 304 (5672), 838.

Van Cauwenberghe, L., Claessens, M., Vandegehuchte, M., Janssen, C.R., 2015. Microplastics are taken up by mussels (*Mytilus edulis*) and lugworms (*Arenicola marina*) living in natural habitats. Environ. Pollut. 199, 10-17.

Woodall, L.C., Sanchez-Vidal, A., Canals, M., Paterson, G.L.J., Coppock, R., Sleight, V., Calafat, A., Rogers, A.D., Narayanaswamy, B.E., Thompson, R.C., 2014. The deep sea is a major sink for microplastic debris. R. Soc. Open Sci. 1, 140317. http://dx.doi.org/10.1098/rsos.140317.

Wright, S.L., Thompson, R.C., Galloway, T.S., 2013. The physical impacts of microplastics on marine organisms: a review. Environ. Pollut. 178, 483-492.

Zettler, E.R., Mincer, T.J., Amaral-Zettler, L.A., 2013. Life in the “plastisphere”: microbial communities on plastic marine debris. Environ. Sci. Technol. 47, 7137-7146. http://dx.doi.org/10.1021/es401288x.

Zuur, A.F., E.N., Ieno, G.M., Smith, 2007. Analysing ecological data. Springer, New York.

Captions to figures

Fig. 1. Study area and location of Stations 1 -5. The dotted line denotes the beach width, delimited by a continuous wall interrupted at the mouth of Ghiararo Creek.

Fig. 2. Abundance ± sd (fibres kg^{-1}) of (A) total fibres and transparent fibres. Significant differences (t-test, $p<0.05$) for the total number of fibres among stations referred to the same sampling are indicated by the letters located on the histogram (the same letter means significant difference among stations); (B) coloured fibres.

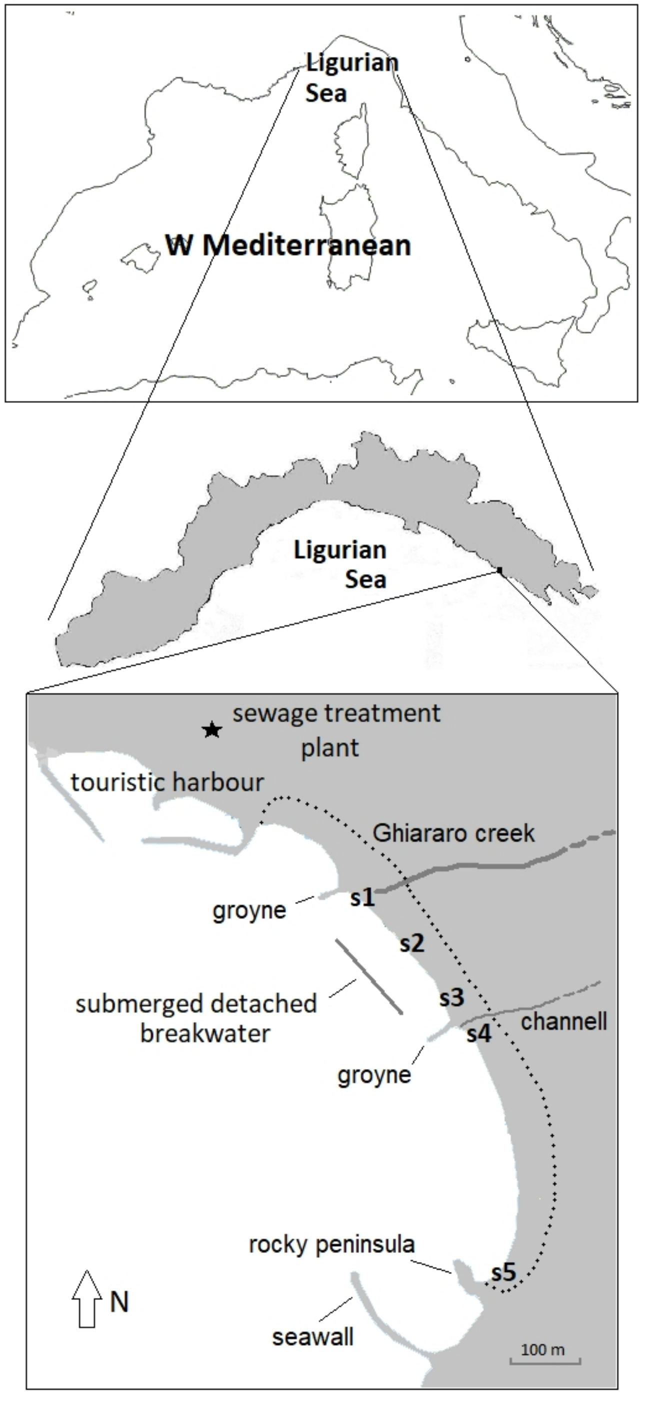
Fig. 3. Size-class distribution of the coloured and transparent fibres: small (0.1 - 0.5 mm), medium (0.5 - 2 mm), large (larger than 2 mm). Dark colour: black and blue fibres, light colour: red and violet fibres.

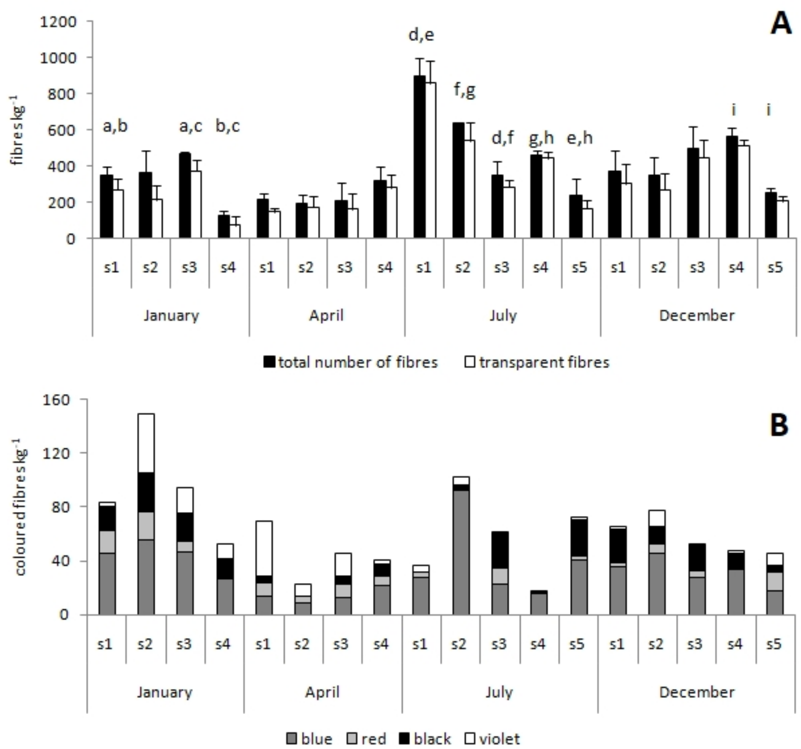
Fig. 4. Sediment content ± sd (µg g^{-1}) of carbohydrates and proteins.

Fig. 5. Characteristics of the waves on the sampling days (A) and for the latest sea storm (B). The vectors indicate the mean direction of waves, which were strongly related to wind direction.

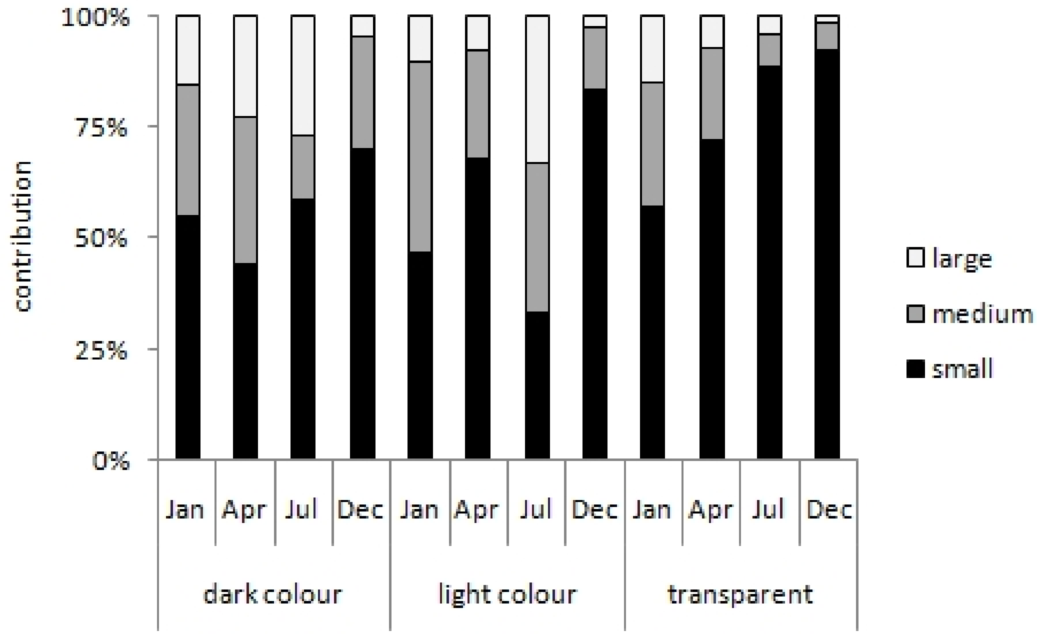
Fig. 6. Outputs of the Xbeach model for waves coming from SW. A: storm-like wave height, B: calm-sea conditions. Coloured legends report the wave height (m).

Fig. 7. Plot of the RDA on fibres (A) and on fragments and spheres (B). The markers report observations (coming from the analysis of the response variables) related to single stations (Stations 1-5) in the different samplings. The explanatory variables are represented by the vectors and the black squares (for the nominal variables).

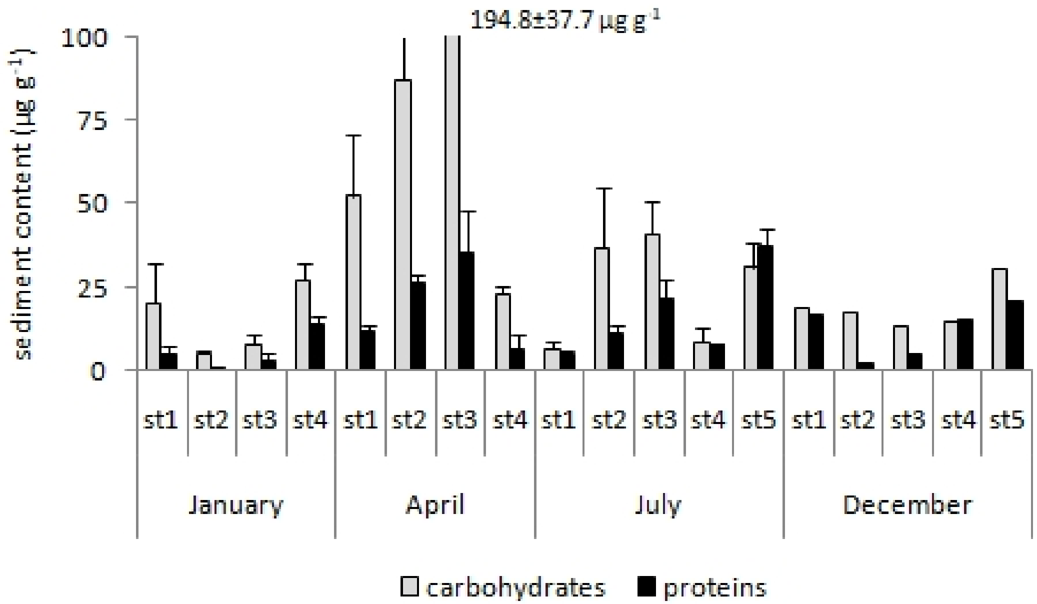

Ligurian Sea
W Mediterranean
Ligurian Sea
sewage treatment plant
touristic harbour
Ghiararo creek
groyne
s1
s2
submerged detached breakwater
s3
s4
channell
groyne
rocky peninsula
s5
N
seawall
100 m

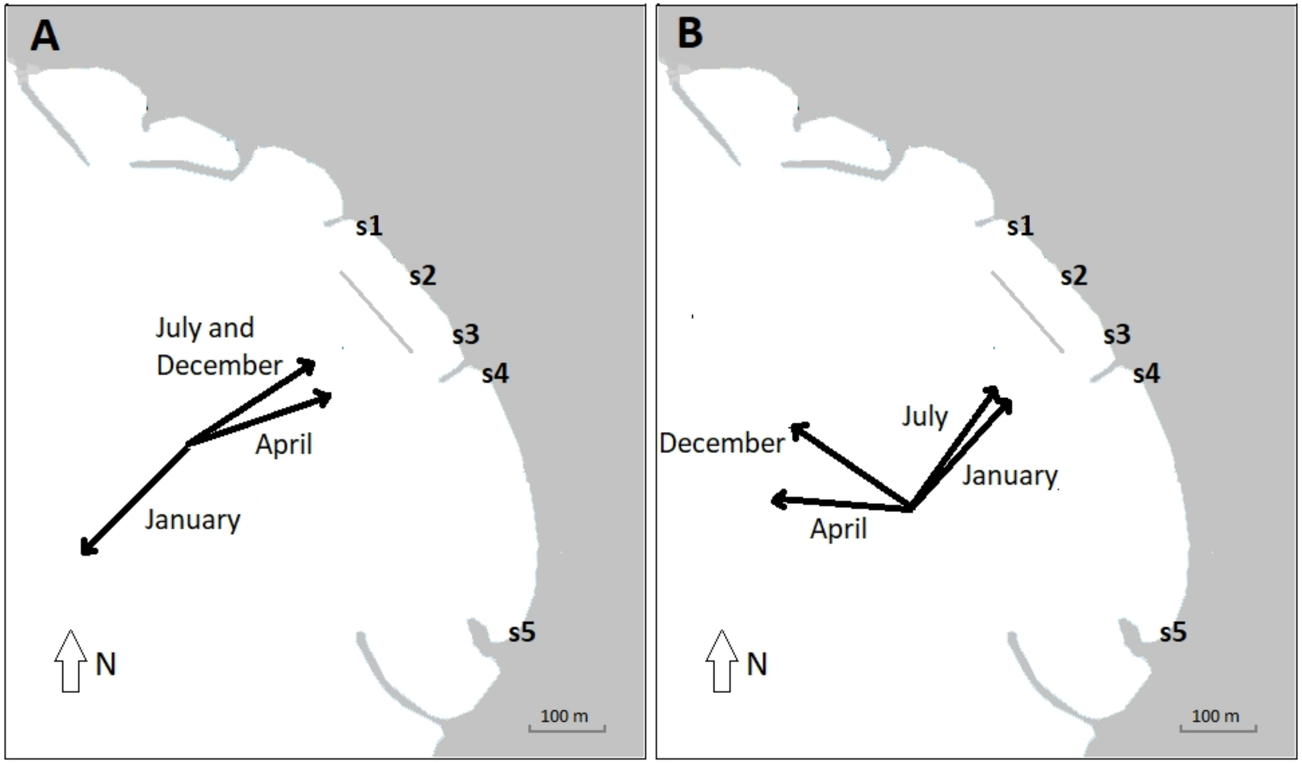


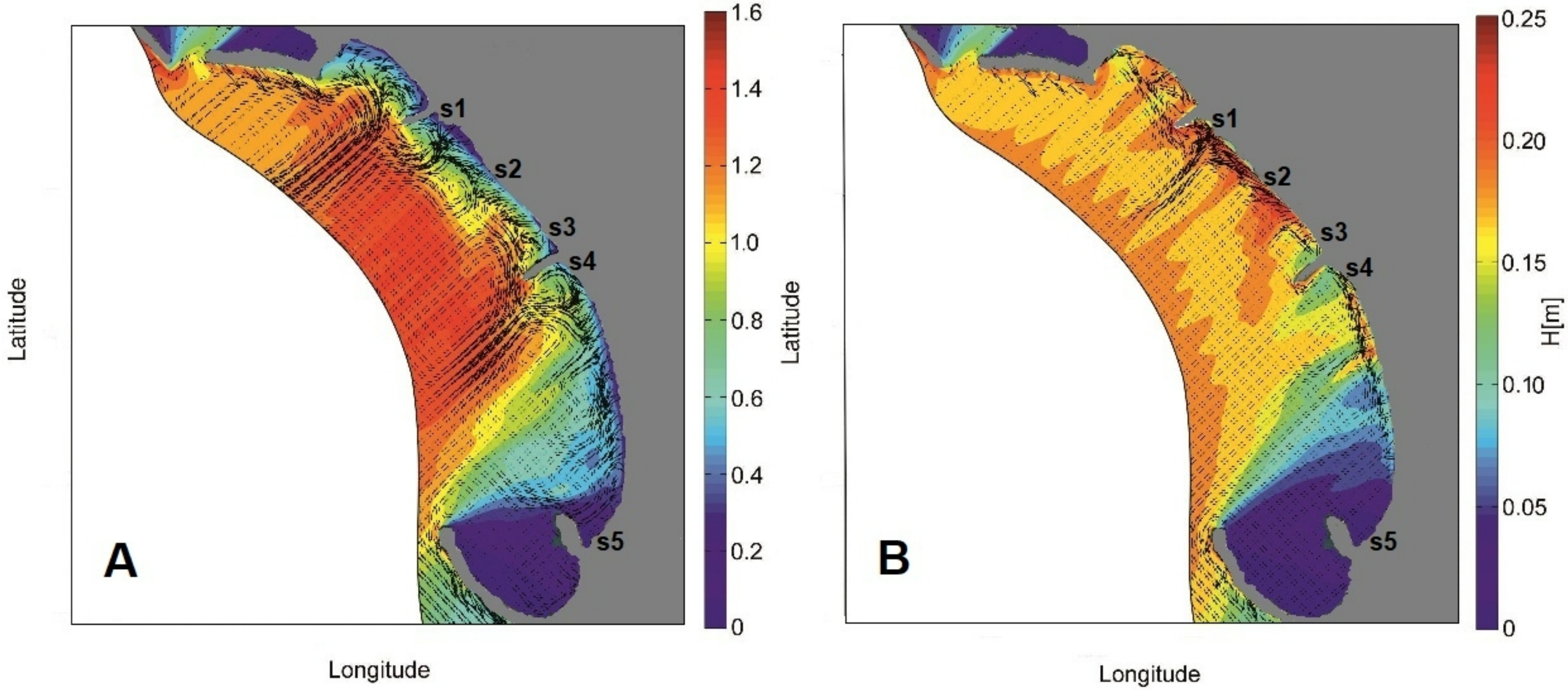
A
1200
1000
800
600
400
200
0
fibres kg-1
a,b
a,c
b,c
d,e
f,g
d,f
g,h
e,h
i
i
s1
s2
s3
s4
s5
January
April
July
December
total number of fibres
transparent fibres
B
160
120
80
40
coloured fibres kg-1
blue
red
black
violet

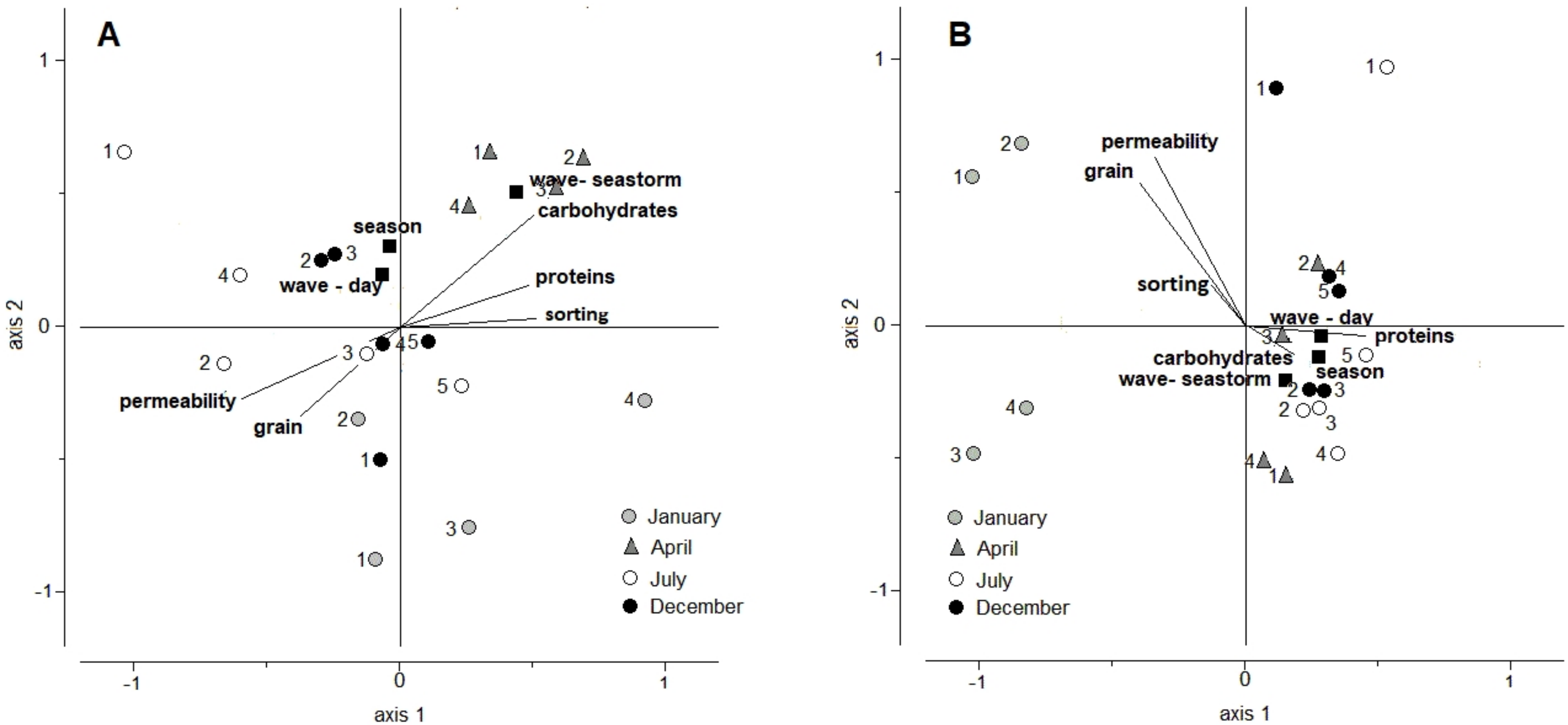


100%
75%
50%
25%
0%
contribution
Jan
Apr
Jul
Dec
Jan
Apr
Jul
Dec
Jan
Apr
Jul
Dec
dark colour
light colour
transparent
large
medium
small


194.8±37.7 µg g-1
sediment content (µg g-1)
100
75
50
25
0
st1 st2 st3 st4 st1 st2 st3 st4 st1 st2 st3 st4 st5 st1 st2 st3 st4 st5
January
April
July
December
carbohydrates
proteins


A
s1
s2
s3
s4
s5
July and
December
April
January
N
100 m
B
s1
s2
s3
s4
s5
July
December
January
April
N
100 m


s1
s2
s3
s4
s5
A
Latitude
Longitude
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0
B
Latitude
Longitude
0.25
0.20
0.15
0.10
0.05
0
H[m]



A
permeability
grain
sorting
proteins
carbohydrates
wave- seastorm
season
wave - day
January
April
July
December
axis 1
axis 2
B
permeability
grain
sorting
wave - day
proteins
carbohydrates
wave- seastorm
season
January
April
July
December
axis 1
axis 2

Table 1. Abundance ± sd of fragments or spheres per kg. Na: data not available. 0: complete absence in all replicates.

	station 1		station 2		station 3		station 4		station 5	
	fragments kg^{-1}	spheres kg^{-1}	fragments kg^{-1}	spheres kg^{-1}	fragments kg^{-1}	spheres kg^{-1}	fragments kg^{-1}	spheres kg^{-1}	fragments kg^{-1}	spheres kg^{-1}
January	21±18	8±11	32±43	2±4	344±144	2±4	75±98	3±4	na	na
April	27±19	0	30±22	2±5	38±23	0	30±20	0	na	na
July	13±16	0	0	0	0	0	0	0	4±5	0
December	0	0	12±11	0	0	2±4	2±4	0	2±4	0

Table 2. Sediment features (mean grain size, permeability, and sorting) for each date and station.

		mean grain size		permeability		sorting	
		mm	sd	$*10^{-2}$ cm s^{-1}	sd	phi	sd
17 Jan	s1	7.0	1.2	5.04	1.39	1.2	0.1
	s2	5.9	1.6	5.69	1.90	1.0	0.1
	s3	3.8	0.1	1.90	0.21	1.1	0.1
	s4	0.7	0.1	0.03	0.00	1.7	0.0
4 Apr	s1	2.1	0.4	0.52	0.06	1.2	0.2
	s2	2.1	0.5	0.17	0.02	2.0	0.3
	s3	1.5	0.1	0.12	0.01	1.6	0.1
	s4	3.6	0.2	1.08	0.19	1.2	0.1
26 Jul	s1	5.4	0.9	6.17	0.54	0.9	0.2
	s2	4.0	0.5	2.27	0.22	1.0	0.1
	s3	2.2	0.2	0.28	0.04	1.5	0.1
	s4	2.8	0.5	1.95	0.56	0.8	0.0
	s5	0.3	0.0	0.03	0.00	0.7	0.0
4 Dec	s1	5.2	2.6	1.58	2.51	2.1	0.5
	s2	1.5	0.1	0.16	0.02	1.3	0.1
	s3	0.9	0.0	0.16	0.04	1.0	0.1
	s4	1.5	0.2	0.79	0.40	0.8	0.1
	s5	0.5	0.1	0.06	0.01	0.8	0.1

Table 3. Hydrodynamic characteristics. The mean wave height (±sd), the significant wave height, and the direction of the waves were reported for the sampling day (only to the sampling hour), for the latest sea storm and for the other days of the week before the sampling. In January, the days of the week before the sampling are divided into two groups due to the exceptional wind event that completely changed the sea characteristics.

		mean wave height (m)	significant wave height (m)	direction (° from N)
January	sampling day	<0.1	<0.1	42-57
	storm day	2.46±0.33	2.98	216-260
	week before	1.06±0.41	2.04	144-260
	wind event	<0.1	<0.1	348-4
April	sampling day	0.20±0.02	0.24	234-294
	storm day	0.90±0.27	1.39	42-88
	week before	0.33±0.21	0.91	130-291
July	sampling day	0.85±0.07	0.91	234-240
	storm day	1.50±0.30	2.07	206-227
	week before	0.72±0.35	1.87	149-254
December	sampling day	0.30±0.05	0.40	207-247
	storm day	2.12±0.15	2.40	40-181
	week before	1.10±0.31	1.50	43-255

Table 4. RDA on abundance and colour of fibres.

axis	eigenvalue	eigenvalue as percentage of total variance	cumulative	eigenvalue as percentage of sum of all canonical eigenvalues	cumulative
1	0.285	28.5	28.5	51.3	51.3
2	0.132	13.2	41.2	23.7	74.9

Table 5. RDA on abundance and colour of fibres. The variables showing a statistically significant effect are reported in bold letters.

variable	marginal effects		conditional effects		
	eigenvalue using only one explanatory variable	eigenvalue as % (of sum all eigenvalues) using only one explanatory variable	increase total sum of eigenvalues after including new variable	F statistic	P-value
permeability	**0.16**	**28.11**	**0.16**	**2.963**	**0.038**
wave direction (day)	**0.11**	**19.45**	**0.14**	**3.075**	**0.014**
wave direction (latest seastorm)	**0.15**	**26.74**	**0.14**	**3.351**	**0.016**
mean grain size	0.11	19.18	0.03	0.810	0.504
proteins	0.09	16.87	0.03	0.718	0.614
season	0.05	9.79	0.02	0.515	0.756
sorting	0.07	13.44	0.02	0.346	0.860
carbohydrates	0.09	16.85	0.02	0.391	0.826

Table 6. RDA on abundance of fragments and spheres.

axis	eigenvalue	eigenvalue as percentage of total variance	cumulative	eigenvalue as percentage of sum of all canonical eigenvalues	cumulative
1	0.466	46.6	46.6	88.7	88.7
2	0.059	5.9	52.5	11.3	100.0

Table 7. RDA on abundance of fragments and spheres. The variables showing a statistically significant effect are reported in bold letters.

variable	marginal effects		conditional effects		
	eigenvalue using only one explanatory variable	eigenvalue as % (of sum all eigenvalues) using only one explanatory variable	increase total sum of eigenvalues after including new variable	F statistic	P-value
wave direction (day)	**0.44**	**82.84**	**0.44**	**12.333**	**0.002**
permeability	0.07	13.77	0.03	0.842	0.352
mean grain size	0.10	18.73	0.02	0.449	0.642
sorting	0.01	1.62	0.01	0.363	0.732
season	0.12	23.13	0.01	0.246	0.772
proteins	0.07	12.69	0.01	0.268	0.810
carbohydrates	0.01	2.11	0.01	0.118	0.814
wave direction (latest seastorm)	0.01	2.25	0.00	0.036	0.966

Declaration of interests

XThe 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.

☐The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:

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