CleanAtlantic

Tackling Marine Litter in the Atlantic Area

Monitoring floating microlitter in offshore waters by manta-trawl (collaboration with iFADO project)

WP 5: Monitoring and data management WP 5.2: Monitoring the presence of marine litter in the marine environment



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Abstract

Floating marine litter is a recurrent pollutant in the Atlantic and worldwide oceans which has attracted increasing attention in the latest years. Meso- and microplastics derived from industrial pellets or through debris degradation have been also reported in surface and subsurface water samples from Atlantic coast and high-seas. The need of microplastic monitoring and assessment in EU countries and the Atlantic Area calls for the establishment of standardized sampling protocols and methods. In this work, the application of manta-trawl for micro-litter monitoring in coastal and offshore waters of the Galician coast (NW Spain) was assessed. The results obtained in this work suggest a moderate micro-litter density in both coastal and offshore stations. Also, manta-trawl was proved to be a feasible tool for surface and subsurface meso and micro-litter collection.

Introduction

Litter in the ocean has been reported since early 70s (Carpenter et al., 1972; Venrick et al., 1973). All along these years, the degradation and fragmentation of manufactured debris led tinny pieces of anthropogenic material (micro-litter) to "colonize" almost any spot of the planet (Colton et al., 1974; Ryan and Moloney, 1993; Thompson et al., 2004). The term Microplastics (MPs) refers to small plastic fragments, usually measuring below 5 mm in size, which have their sources either in minute plastic used in cosmetics, abrasives and plastic industry (primary MPs) or in macroplastic fractioning (secondary MPs) (Andrady, 2011; Barnes et al., 2009; Cole et al., 2011; Thompson et al., 2004).

Despite their small size, it has been reported an estimation of 14.9 to 51.2 trillion of plastic particles (93.3-236 thousand metric tons) globally, in the ocean (van Sebille et al., 2015) and they contribute to environ 80% of the total plastic litter in marine waters (Sharma and Chatterjee, 2017). Their micro and nano scale make these particles bioavailable for a wide range of organisms from those located at the basis of marine food webs to top predators (Avio et al., 2020; Farrell and Nelson, 2013; Frias et al., 2014; Nelms et al., 2018; Setälä et al., 2018, 2014; Zantis et al., 2021). Recent studies suggest not only a direct harm to organisms by synthetic polymer ingestion (such as intestine injuries, false satiation and poor nutrition) but also chemical harm due to their additives and capability to adsorb and concentrate a range of hazardous compounds (Costa et al., 2020; Hahladakis et al., 2018; Mato et al., 2001; Moore, 2008; Parra et al., 2021; Wang et al., 2020; Wright et al., 2013).

The evidences of MPs potential harm to marine environments and human food safety led to the need of their surveillance and monitoring in water compartments and biota to give a coordinated response and provide global solutions by governments and stakeholders. To this goal, standardization of sampling methods and sample treatment are required (Gago et al., 2016; Galgani et al., 2013).

Several approaches have been tested so far for floating micro-litter monitoring (Hidalgo-Ruz et al., 2012). Most common sampling strategies include direct water collection or water filtration through plankton nets (Barrows et al., 2017; Cole et al., 2014; Collignon et al., 2012; Covernton et al., 2019; Cózar et al., 2015; de



Lucia et al., 2014; Eriksen et al., 2018; Lusher et al., 2015; Maes et al., 2017). The use of manta trawls of 333 µm mesh-size has been recommended in certain studies and technical reports (Frias et al., 2019; Gago et al., 2016). Subsequent procedures comprise filtration through GF filters with previous digestion or/and density separation of organic material, if necessary (Cole et al., 2014; Collignon et al., 2012; Covernton et al., 2019). Finally, detection and identification of particles by binoculars and spectroscopic techniques are recommended (Hidalgo-Ruz et al., 2012; Song et al., 2015).

In the frame of the CleanAtlantic and in collaboration with iFADO project 10 samples from Galician offshore waters (NW Iberian Peninsula) were collected by means of a manta-trawl net for micro-litter categorization. The results will contribute to a better characterization of surface micro-litter and MPs distribution in the Atlantic area and to the identification of gaps and needs on sampling protocols and procedures for MPs detection and identification. These objectives are in the line with those envisaged in CleanAtlantic WP.5.2. *Reinforcement and support of harmonized monitoring of marine litter in the framework of the Marine Strategy Framework Directive (MSFD)* as well as with the goals of iFADO which, under the MSFD, aims at the use of traditional and innovative platforms for ocean observation and monitoring.

Methods

1. Sampling campaign

The oceanographic campaign iFADO20-RAD PROOF was performed by means of the research vessel Sarmiento de Gamboa in July, 2020. The sampling stations and coordinates are displayed in Table 1 and Figure 1. The samples were collected by means of a manta trawl net with 333 μ m mesh size and an aperture frame of 0.12 cm². The net was equipped with a flow-meter (Hydrobios, model 438 110). At each sampling point the net was positioned on the side of the boat and towed for 10 min at a speed of 3 knots. Once the net was retrieved onboard it was washed with clean water and the material deposited in the codend placed carefully in a glass jar.



Figure 1. Location of stations where samples were collected.

Table 1. Sampling stations with date and coordinates of transects where samples were collected.



Station		Time (GMT)	IN	ITIAL	FINAL		
	Date		LATITUDE	LONGITUDE	LATITUDE	LONGITUDE	
103	10-07-20	17:30	43º 0.27'	9º 20.24'	42º 59.78'	9º 20.20'	
133	12-07-20	7:40	43º 0.14'	14º 56.71'	42º 59.95'	14º 56.58'	
111	13-07-20	3:43	43º 0.21'	14º 3.97'	43º 0.01'	14º 4.09'	
108	14-07-20	10:45	43º 0.35'	12º 40.60'	43º 0.26'	12º 39.71'	
77	17-07-20	18:28	42.66º O'	9.36º 0'	42º 40.05'	9º 22.16'	
11	18-07-20	8:23	42º 59.89'	10º 0.77'	42º 59.93'	10º 0.95'	
5	19-07-20	4:17	43º 0.04'	9º 35.00'	42º 59.36'	9º 35.10'	
56	22-07-20	7:10	43º 45.88	8º 3.96'	43º 45.64'	8º 4.14'	
58	22-07-20	10:40	43º 57.23	8º 9.02'	43º 57.03'	8º 9.01'	
60	22-07-20	14:58	44º 11.77	8º 15.14'	44º 11.69'	8º 15.38'	

2. Sample processing

Samples were frozen in amber glass bottles at -20°C until their processing in the laboratory at the Spanish Oceanographic Institute (IEO) in Vigo. For micro-litter characterization, the glass-bottles containing approximately 150-200 mL of water were allowed to thaw overnight. When the load of organic material in the sample was low, the sample was directly filtered through a 47 mm-diameter glass fiber GF filter with 2 μ m pore size by means of a vacuum system. For some samples, the presence of high amounts of zooplankton hampered direct filtration. In this case, the sample was processed as follows. First, samples were treated with 10% KOH (ratio 1:3, sample:solution, v:v) and allowed to react for 3 days at 40°C. Subsequently, the samples were further digested with 15% H₂O₂ (1:1, v:v) and kept again at 40°C for 3 additional days (Frias et al., 2019). Since crustacean's carapaces were still profusely abundant in certain samples, those were avoided by density separation. For this purpose, the sample was mixed with a NaCl dilution (p=1.2 g L⁻¹) in a glass cylinder allowing for the sedimentation of organic remains during 24 hours. After this time, the upper layer was filtered as previously stated. Filters with micro-debris and MPs retained on them were placed in a clean glass petri-dish and examined under a Leica M165 C stereoscope equipped with a camera and LASX software.

Along all these procedures, quality control was carried out, which included working in an clean closed room, the avoidance of plastic ware, wearing cotton lab coats, as well as the performance of surface controls to check environmental contamination and procedural controls handled in parallel with the samples (Woodall et al., 2015).



For particles measures the software ImageJ was employed. The longitude and wide of fibers and filaments were recorded. For fragments, pellets and films the longest dimension, perimeter and surface area were also calculated using the same software.

All items resembling synthetic were photographed, pricked with a hot needle to check melting and a representative subsample was carefully picked with tweezers to be analyzed by RAMAN spectroscopy (Thermo). The obtained spectra were compared against data in the RAMAN polymer library by means of the Omnic Specta software furnished with the equipment. Approximately, 14 % of the samples were analyzed.

Results and discussion

1. Micro-litter abundance and density

The total number of items for all the sampling sites accounted 140. Over the 140 items, 74 were visually identified as fibers, while 66 were included in the fragment, pellet, filament and film categories, being the former the most numerous. Only one sample (sample 103) was free of synthetic items. This sample was also the one with the highest organic material, which could hampered the identification of small artificial particles. The sample with the highest number of particles was the sample 108 collected approximately 150 miles off the Galician coast (Figure 2). There was a degree of uncertainty about the composition of items identified as fibers, due to their resemblance with vegetal structures and crustacean appendixes. The spectra retrieved from these items by RAMAN spectrometry were also inconclusive. For this reason fibers were not considered in further analyses.



Figure 2. Number of items collected per station. For each sample counts with our without fiber are displayed.

For micro-litter density estimations, the number of items were expressed per m² and m³ referring to the distance towed and the volume filtered throughout the net, according to flow-meter recordings (Figure 3). Since data obtained with flow-meter could lead to errors due to the net behavior on the surface, the same



calculation was performed by taking distances into account, showing comparable results (data not show). The average of item density for the whole sample collection was 0.0089 ± 0.0091 item/m² or 0.0446 ± 0.0455 item/m³. As expected, the highest densities were obtained for station 108 (0.032 item/m² and 0.162 item/m³) followed by samples 133 and 77, which were located nearer the west coast of Galicia.

The average of micro-debris concentrations from the present study shows similar values to those previously reported in NW Iberian upwelling system (0.011-0.285 items/m²) (Gago et al., 2015) and Portuguese coasts (Frias et al., 2014) (0.03 particles/m³) and falls into the range of overall densities observed in the North Eastern Atlantic (Kanhai et al., 2017). However higher MPs densities were found in the Bay of Brest (0.24 \pm 0.35 item/m³), English Channel (0.27 particles/m³) (Cole et al., 2014) as well as North and Celtic seas (2.46 items/m³ and 0.14 fragments/m³ respectively) (Lusher et al., 2014; Maes et al., 2017). On the other hand, these differences may decrease if fibers were taken into consideration in our estimations.



Figure 3. Item density estimations for each sampling site expressed per unit of surface and volume of water filtered.

2. Micro-litter categorization

Only one filament and one film pieces could be categorically identified by RAMAN spectroscopy as polyethylene teraphtalate (PET) and Polyethylene (PE), in samples 133 and 5, respectively (Figure 4a,b and A, B). The percentage of match against the spectra in our RAMAN polymer library was 76.24% for PET and 95.15% for PE.

Unfortunately, for the remaining items, and despite their apparently synthetic origin by visual inspection, no reliable matches with this database were found (Figure 5). Fibers were identified in several samples, but



they were hard to distinguish from those of apparent natural origin and no peaks could be inferred from the spectra due to the absence of signal or samples showing fluorescence and noisy background. From a conservative approach, all fibers were excluded from the record. Additionally, several fragments resembled plastic paint scraps from vessels (Figure 5f). Due to the difficulties in identifying their actual origin, these scraps were included in the general category of fragments. By excluding fibers, which otherwise would be the main piece type in the totality of samples (52.86%), fragments accounted for 95.45% of the recorded particles.



Figure 4. Images of MPs identified in samples 133 and 5 (a and b) and corresponding spectra (A and B). Scale bars in the images indicate a) 500 µm and b) 250 µm. A) Spectrum of a) matching polyethylene teraphtalate (PET) with 76.24% of coincidence. B) Spectrum of b) matching Polyethylene (PE) with 95.15% similarity against the standard in the database.

In most studies, fibers are found to be the dominant MPs type in surface water samples from Atlantic European waters (Cole et al., 2014, 2011; Díez-Minguito et al., 2020; Lusher et al., 2014; Mendoza et al., 2020). However, several authors pointed fibers as the main cause of sample contamination during sampling procedures and handling, stressing the need of a tight quality control across all collection and treatment steps (Tamminga et al., 2018; Woodall et al., 2015). Furthermore, previous studies reported issues in identifying fibers by visual and spectroscopic techniques (Cózar et al., 2015; Frère et al., 2017; Song et al., 2015). For example, a high percentage of fibers found in water and biota are natural or made of Rayon, a semi synthetic material which could be misidentified as cellulosic (Lusher et al., 2014; Setälä et al., 2016). More specific libraries for RAMAN or F-TIR spectra identification would be needed to properly assign the nature of environmentally degraded fibers.





Figure 5. a) Blue fiber from 100% cotton lab coat (control filter); b-f) Example of items of unknown composition: b) Filament-like item of probably natural origin (e.g. crustacean chitin), c) Round item of unknown material, d) and e) Synthetic items behaving like plastics and f) Probable paint scrap. Scale bars in the images gauge a) 100 μ m; b) 100 μ m; c) 750 μ m; d) 250 μ m; e) 250 μ m; f) 2.5 mm.

After fibers, fragments are by far the second particle type most detected in the Atlantic (Frère et al., 2017; Frias et al., 2014; Maes et al., 2017). Paint scraps are also frequently found as marine debris in surface and subsurface water samples (Díez-Minguito et al., 2020; Frias et al., 2014; Setälä et al., 2016). Despite their occurrence could be, at some extent, due to the research vessel chipping, they may be mainly linked to locations with elevated presence of vessels. The Fisterra Traffic Separation Scheme, which is a shipping lane with important maritime traffic affluence, may have contributed to the large amount of paint sheet fragments in sample 108.

Regarding size, the majority of items measured less than 1 mm (62.12%), followed by pieces with size between 1 and 5 mm (34.85%) in their largest dimension (Figure 6). Although the PET filament found in sample 133 reached 10 cm in length, it is likely that it behaves as MPs since it was tightly curled up in a ball. The samples where the biggest fragments were found were 133 and 77 (Table 2). These values are slightly smaller than other studies were a 300 or 333 µm-mesh manta trawl was employed for sampling (Díez-Minguito et al., 2020; Maes et al., 2017; Sadri and Thompson, 2014). Nevertheless, these differences may be related to the exclusion of fibers in the analyses. Fibers retained in the manta trawl are usually slightly longer than 1 mm, increasing the 1-5 mm- size class percentage of the overall samples to 54.29%.





Figure 6. Number of items per class-size estimated for each sample.

Table 2. Average and standard deviation of main dimensions exhibited in fragments found in each sample. Length is considered as the largest dimension and wide the shortest one.

Sample	133	111	108	77	11	5	56	58	60
Length (mm)	6.17 ±20.53	0.73±0.11	1.43±1.04	1.29±1.09	1.18±1.24	2.54	2.73±3.81	2.43±1.76	2.72±1.76
Wide (mm)	0.19±0.52	0.51±0.01	0.42±0.33	0.37±0.73	0.11±0.16	0.93	0.31±0.49	0.59±0.17	0.16±0.30
Area (mm²)	20.24±32.22	0.10±0.10	4.50±17.65	24.25±36.94	6.16±10.92	1.20	1.00±0.65	0.38±0.26	0.50±0.60
Perimeter (mm)	2.12±2.82	1.96±0.31	2.59±1.39	3.44±3.98	1.11±0.77	6.09	3.94±1.35	2.65±1.09	2.97±1.42

As in previous reports from the Atlantic, when considering fibers in the analyses, the most common color was black and dark-blue (51.43%) (Cole et al., 2014; Díez-Minguito et al., 2020; Kanhai et al., 2017; Lusher et al., 2014). By excluding fibers, the green color (40.91%) was the one found mainly in fragments, followed by yellow (27.27%), red (21.21%) and blue (10.61%) (Figure 7).





Figure 7. Number of items sorted by colour estimated for each sample.

3. Feasibility of manta-trawl sampling

Although several sampling strategies have been suggested for MPs sampling, manta trawl has been recommended by several authors due to their capacity to sieve large volumes of water with relative low effort (Frias et al., 2019; Gago et al., 2016). The direct continuous collection of water by boat pumps or external ones has been suggested as an interesting alternative with the main advantages of sampling considerable volumes requiring little handling (especially when using boat pumps systems) (Kanhai et al., 2017; Lusher et al., 2015, 2014). They were also suggested to be more efficient than nets in retaining the smallest particles. On the other hand, in this study items lower than 1 mm were frequently found in most samples, since once the net is clogged particles measuring less than the mesh size are equally retained. In certain studies different nets and net sizes were explored finding similar size (< 1 mm) and particle retention (Eriksen et al., 2018). Discrete methods could be opportunistically used (*i.e.* collecting surface water with a bucket or Niskin bottles) having the obvious advantages of feasibility and simplicity as well as the retention of small items (Barrows et al., 2017; Covernton et al., 2019; Tamminga et al., 2018). However, the need of global standardised methods calls for a consensus of the scientific organisations responsible for marine environment monitoring.



Conclusions

In this study, the employment of a 333 µm-mesh manta trawl enabled the estimation of neustonic micro debris at several sites, offshore NW Spanish Atlantic waters. This methodology is suitable for sampling large water volumes and allows for the collection of micro particles, even tinier than the mesh-size. Fragments found in this work were abundant in both coastal and offshore stations and were mainly composed by particles with sizes smaller than 1 mm, and predominance of green colour. However, a great number of suspected anthropogenic items could not be conclusively identified. Strict control of environmental contamination and improved detection methods for synthetic and natural item discrimination are still needed.



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