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Baseline

A baseline assessment of beach macrolitter and microplastics along northeastern Atlantic shores

Nadja Velez[a,*], Gerardo I. Zardi[b], Roberto Lo Savio[c], Christopher D. McQuaid[b], Ugo Valbusa[c], Brahim Sabour[d], Katy R. Nicastro[a]

[a] CCMAR, CIMAR Associated Laboratory, University of Algarve, Campus de Gambelas, 8005-139, Faro, Portugal
[b] Department of Zoology and Entomology, Rhodes University, Grahamstown, 6140, South Africa
[c] Department of Physics, Università degli Studi di Genova, Genoa, 16146, Italy
[d] Department of Biology, Faculty of Sciences, Chouaib Doukkali University, P.O. Box 20, 24000, El Jadida, Morocco

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ABSTRACT

Marine litter is widely dispersed throughout coastal environments. Assessing the distribution and accumulation of such contaminants is crucial to understand their environmental impacts. This study presents a baseline for the monitoring of litter and microplastics in intertidal sediments along the Atlantic shores of southern Portugal and Morocco and identifies potential sources of contamination. Although variable, distribution and composition of both litter and microplastics did not follow a latitudinal pattern. Most of the litter had an undifferentiated source. Within the identifiable sources of litter, food packaging, fishing and tobacco were the most abundant, with variable contributions among sites. Over 97% of marine litter retrieved was plastic. Fragments and filaments were the most abundant categories of plastics at sites with the highest and lowest microplastic abundance respectively. Filaments were mainly made of Polypropylene (PP,50%) and Polyethylene terephthalate (PET,29%) while the predominant polymers for fragments were Polyethene (PE, 75%) and PP (25%).

The amount of litter discarded into or transported from land to the marine and coastal environment is an increasing threat to ecosystems and the services they provide to mankind worldwide (Haward, 2018; Barnes et al., 2009; Gall and Thompson, 2015; Newman et al., 2015; UNEP, 2005). Marine anthropogenic litter or debris is generally defined as "any persistent, manufactured or processed solid material discarded, disposed of or abandoned in the marine and coastal environment" (Galgani et al., 2010). It is mainly composed of items made of plastics, paper, metal, wood and textiles (Gall and Thompson, 2015; OSPAR Comission, 2007). In particular, since the mass production of plastics started in the 1950s, contamination of this long-lasting, versatile material has rapidly emerged as the largest component of marine debris and a serious environmental and economic threat which, together with other global key issues such as overfishing, raising temperatures and ocean acidification, seriously jeopardizes the biodiversity of marine ecosystems and the goods and services they provide (Claessens et al., 2013; Cole et al., 2011; Derraik, 2002; Gregory, 2009). Despite these environmental concerns, the demand for plastics is great. Globally, annual plastic production has increased dramatically from 1.5 million tonnes in the 1950s to approximately 322 million tonnes in 2015 (PlasticsEurope, 2016). Because of the lack of comprehensive waste management plans, the amount of plastic waste in the environment has accumulated at an uncontrollable rate and is expected to increase (Geyer et al., 2017).

Currently, most plastic in the marine environment is represented by small particles with an upper size limit of 5 mm, generally called microplastic (Claessens et al., 2013; Martins and Sobral, 2011; Wright et al., 2013). Primary microplastics are originally manufactured as industrial pellets used as precursors in the manufacture of larger plastic items or microbeads incorporated in a number of industrial (air-blasting media) and household (hand-cleaners and facial scrubbers) products (Cole et al., 2011). Secondary microplastics originate from the fragmentation that results from degrading processes such as photo-oxidation or mechanical degradation of larger items. Due to their relatively small size, microplastics becomes bioavailable to organisms throughout the food-web (e.g. Farrell and Nelson, 2013). There is ample evidence in a wide variety of taxa that microplastic ingestion can result in severe physical and physiological effects (e.g. reduced growth and reproduction, internal abrasions and blockages; Egbeocha et al., 2018). Additionally, microplastics may become heavily contaminated, thus acting as vectors of toxic chemicals originating from the plastic item itself or the surrounding environment through the food that affect marine

* Corresponding author.
E-mail address: nvrvelez@gmail.com (N. Velez).

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species and ecosystems and also carry a risk to human health (Gall and Thompson, 2015).

Baseline data on marine anthropogenic litter contamination are of crucial importance to evaluate changes through time and among regions and to define management and conservation strategies.

Surprisingly, marine anthropogenic litter contamination along Atlantic southern Europe and Northern African shores are still largely unexplored. Only a few studies have described plastic contamination along the Portuguese coastline with none for Atlantic Moroccan shores (e.g., Alshawafi et al., 2017). Most studies carried out in Portugal have focused on northern and central shores. Abundant plastic contamination has been reported at several intertidal beaches in the Lisbon region (Frias et al., 2013, 2011, 2010). It has been estimated that microplastics make up 72% of all marine plastic debris found on the west coast of Portugal (Martins and Sobral, 2011). Resin pellets found in ten beaches along the western Portuguese coastline represented 53% of the total marine debris (Antunes et al., 2013) and fishing nets are the predominant type of benthic debris in the canyon of Cabo de São Vicente (Oliveira et al., 2015). The only study carried out in southern Portugal focused on subtidal sediments, although in Browne et al., (2011), one of the 18 worldwide sampling sites was in Faro, southern Portugal), where 56% of samples contained microplastics (Frias et al., 2016).

Here, we provide a quantitative (number of items and total mass of litter) and qualitative (visual and spectroscopic assessment) baseline data on abundance, distribution and composition (including polymer type) of litter and microplastics in intertidal sediments along southern Portuguese and Atlantic Moroccan shores.

Marine anthropogenic litter assessment followed a sampling approach adapted from NOAA, using a standing-stock procedure (Lippiatt et al., 2013; Opfer et al., 2012), which fits less frequent and widely separated assessments better than accumulation stock surveys (Smith and Markic, 2013). At each sampling site (Table 1), a 200 m along shore section of the beach was selected. Within these 200 m, transects (n = 3) of 5 m width were randomly selected prior to sampling and running from the low to the high shore (Lippiatt et al., 2013).Transect lengths at the eight sites ranged from 21 to 27 m. In each transect, the surveyor followed the walking pattern described by Opfer et al. (2012). Each type of litter collected was sorted by material (Ceramic, Glass, Metal, Plastic, Paper, Other; Marine conservation Society, 2012) and source (Construction, Domestic, Fishing, Tobacco, Food Package, Hygiene/Medical, Undifferentiated). Litter size categories in Barnes et al. (2009) were used as guidelines: meso (between 0.5 mm and 2 cm), macro (from 2 cm to 10 cm) and mega (larger than 10 cm). Microplastics (smaller than 0.5 mm) were sampled separately.

For microplastic, at each site, a transect of 30 m was randomly selected on the high tide line mark. Within the selected section, 50 × 50 cm quadrats (n = 3) were randomly picked. Sand surface samples (2 cm) were placed into aluminum container, sealed and brought to the laboratory for further analyses. No plastic material was used while sampling.

Table 1
Codes, full names and coordinates of each sampling site. Population estimation of nearest village/city for each sampling site and year of the population census.

Code	Name	Coordinates	Population	Census
MA	Malhão	37°46′41.73″N 8°48′9.98″W	5031	2011
BO	Bordeira	37°11′54.15″N8°54′16.40″W	432	2011
SA	Sagres	37° 0′19.66″N 8°56′18.27″W	1909	2011
PA	Ponta D'Areia	37°10′10.84″N 7°24′31.48″W	19,156	2011
RA	Rabat	33°55′22.78″N 6°57′52.22″W	577,827	2014
SB	Sidi Bouzid	33°13′50.76″N 8°33′17.02″W	122,676	2014
MI	Mirleft	29°35′28.37″N 10° 2′16.73″W	7026	2004
TA	Tantan	28°29′52.94″N 11°20′9.35″W	73,209	2014

In the laboratory, samples were oven dried at 60 °C until constant weight was reached. Dried samples were weighed and then sieved through a 5 mm sieve to exclude macroplastics from the sample. The extraction of microplastics from the sediment followed the density separation principle (Hidalgo-Ruz et al., 2012). Less dense particles were suspended by adding NaCl saturated ultrapure water (purified by an Elix® equipment), mixed and left to settle. Supernatant was collected with a pipette and filtered through a qualitative filter (Whatman, 1820-047, 1.6 µm). Mixing and filtering was repeated two times (Claessens et al., 2013; Hidalgo-Ruz et al., 2012).

To prevent possible contamination, benches were cleaned and covered in paper and trays wrapped and covered with clean aluminum foil to store material in use. All solutions and glassware were kept covered at all times to avoid airborne contamination. Material was frequently cleaned with distilled water during the procedure and a laboratory coat (100% cotton) was used to prevent contamination by cloth fibers. To prevent cross sample filament contamination, metal sieves were cleaned often with a pure acetone bath (De Witte et al., 2014; Nel and Froneman, 2015). Three blank controls consisting of all microplastic extraction steps without sediment addition were carried out to assess potential contamination; an average of 1 filament (SD = 1.00) and 0.67 fragments (SD = 0.58) was present.

The qualitative filters were analyzed under a dissecting microscope inside closed glass petri dishes. Microplastic particles were counted and sorted into type category (filament, foam, pellet and fragment; (Hidalgo-Ruz et al., 2012; Marine & Environmental Research Institute). To obtain information on polymer composition and to validate microplastic identification, Raman spectroscopy analysis was performed (JASCO NRS-4100, *Laser Raman Spectrometer*) on a subsample (n = 15) of each category. A laser beam (532 or 785 nm) was focused on the sample surface using a 5 × or 20 × objective, resulting in a spot size of ~30 or ~5 µm, respectively; the laser power was in the 0.5–5.0 mW range depending on the specific sample, and it was kept low enough to prevent sample damage. Given the high spatial resolution of the Raman spectrometer, at least three spectra at three different points of the sample surface were acquired for each sample. In order to identify the polymer composition, the spectra were then compared with those of the most common polymers included in a home-made spectral database. When identification through Raman analysis was ambiguous or not possible, usually due to intense photoluminescence background, Fourier-Transform Infrared Spectroscopy (FTIR) was used as an additional technique (JASCO FT/IR-4700), performing both transmission and attenuated total reflectance (ATR) measurements.

To test for differences in macrolitter abundance among sites, two datasets were used. One with data grouped by material and one with data grouped by source. Similarly, to test for differences in microplastic abundance among sites, a dataset was created with data grouped by microplastic category.

For each dataset, a series of one way PERMANOVA was performed with site as a fixed factor and abundance as the dependent variable. These were run using Bray–Curtis dissimilarity matrices for square-root transformed multivariate measures. To search for significant sources of variation identified by the PERMANOVA, post hoc comparisons were performed using pair-wise tests and applying Bonferroni correction to reduce potential type I errors in multiple comparisons. The Monte Carlo P value was preferred over the permutation P-value when very few unique permutations were possible (Anderson, 2005). Permutation tests of multivariate dispersion (PERMDISP; Anderson, 2004) were used to check the homogeneity in the average dissimilarities of samples from the central location of their group. The SIMPER procedure (Clarke, 1993) was used to identify the percentage contribution (%) that each variable made to the between sites Bray-Curtis dissimilarities. The cut

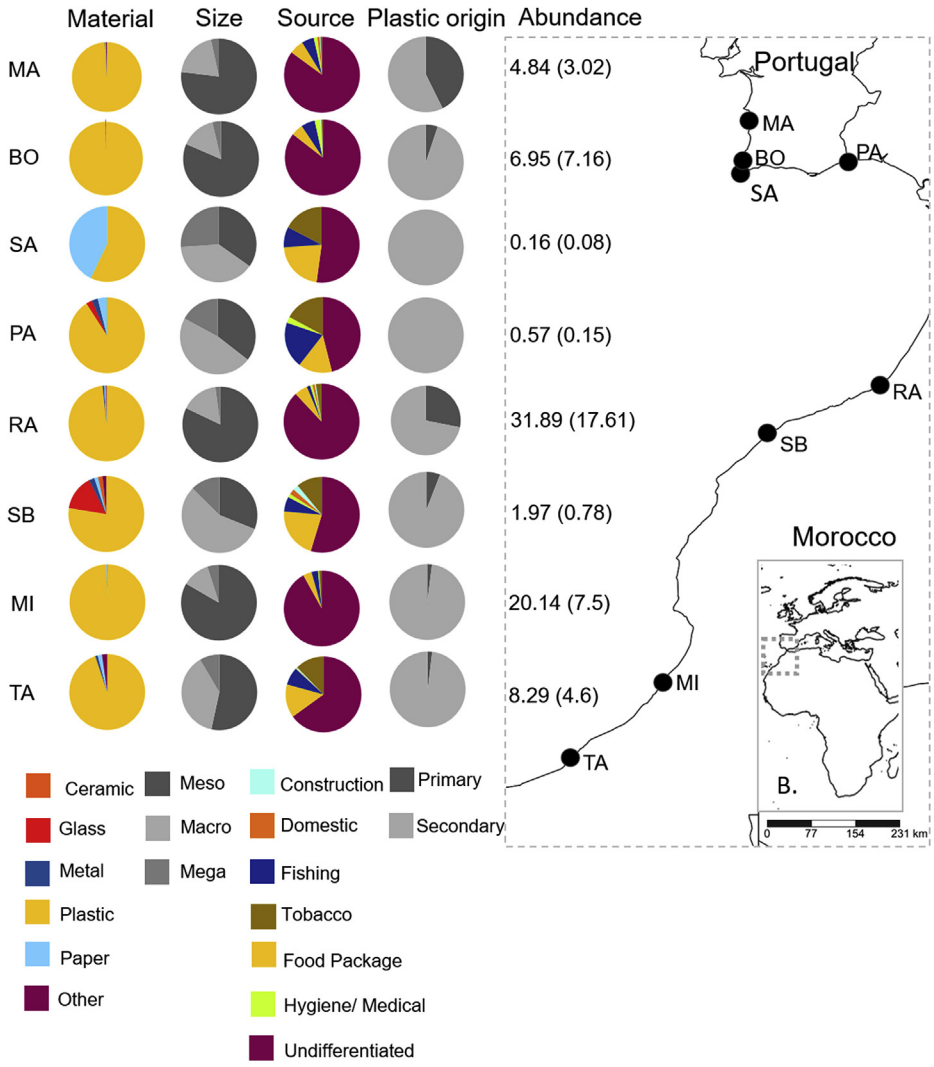

Fig. 1. Sampling site and mean debris abundance (± SD). Proportions of each material, size, source and plastic origin are described by colour coded pies for each site. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

off for low contributions was 50% of cumulative percentage of average dissimilarity between sites. To visualize the datasets spatially, a series of principal coordinate analysis (PCO) based on a Bray–Curtis similarity resemblance matrices were performed. All analyses were performed using PRIMER 6.1.15 & PERMANOVA+ 1.0.5 software (PRIMER-E Ltd, 2012).

A total of 10,023 macrolitter items were encountered across the standing-stock surveys with an average of 9.35 m^{-2}. Overall, our results show relatively higher concentrations of anthropogenic marine macrodebris and larger variability among sites (up to two orders of magnitude) compared to other studies conducted along European beaches. Generally, apart from a study reporting average litter densities in the range of 0.16–3.06 items m^{-2} in Scottish beach sediment, less than one item m^{-2} has been reported on other European beaches (e.g., Benton, 1995; Honorato-Zimmer et al., 2019; Munari et al., 2016; Velander and Mocogni, 1999). Litter was most abundant at RA, MI and TA, and least abundant at SA, PA and SB (Fig. 1). Plastic accounted for most of the litter on these shores, ranging from 57% to 100% of total litter. Similar ratios have been reported globally, where plastic is the main component of beach stranded litter (up to 95%; Galgani et al., 2015). Most litter items belonged to the smallest size categories (Meso or Macro). Most of the litter had an undifferentiated source. Within the identifiable sources of litter, food packaging, fishing and tobacco were the most abundant, with variable contributions among sites. This is in agreement with previous worldwide records that have food packaging as the most produced plastic item and plastic fishing gear debris as the most widespread. Tobacco debris is also commonly found on beaches; this has additional health and environmental implications as toxins adsorbed in cigarette stumps have been shown to promote bioaccumulation of compounds in invertebrates such as marine worms (Wright et al., 2015).

Site had a significant effect on debris abundance when grouped by material (PERMANOVA: $F_{7,16} = 11.16$; $p = 0.0001$). The two sites with lowest abundances of debris were significantly different from the two sites with the highest abundances of debris (pairwise comparison: SA, RA: $t(7) = 5.34$, $p(MC) < 0.0018$; SA, MI: $t(7) = 5.50$, $p(MC) < 0.0018$; PA, RA: $t(7) = 6.30$, $p(MC) < 0.0018$; PA, MI: $t(7) = 6.56$, $p(MC) < 0.0018$). Multivariate dispersion was homogeneous among levels (PERMDISP: $F_{7,16} = 2.01$; $p = 0.441$).

Site had a significant effect on debris abundance when grouped by source (PERMANOVA: $F_{7,16} = 9.4485$, $p = 0.0001$). Two pair-wise comparisons were significantly: PA vs. RA: ($t(7) = 7.59$, $p(MC) < 0.00179$) and PA vs. MI ($t(7) = 8.50$, $p(MC) < 0.00179$). Multivariate dispersion was homogeneous among levels (PERMDISP: $F_{7,16} = 1.44$; $p = 0.801$).

Principal coordinate analysis (PCO) did not show a geographical clustering pattern (Figs. 2 and 3). However, replicates within each site clustered together in all datasets (debris by material and debris by

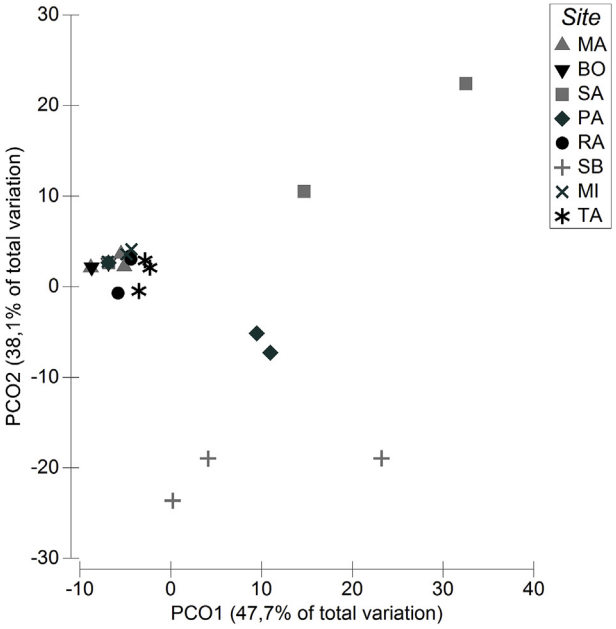

Fig. 2. Principal coordinate analysis (PCO) based on a Bray – Curtis similarity matrix for debris material composition at each site.

source). Overall, the sites that did not fall in the main cluster were those that had the lowest abundances of debris (see Supplementary materials, Table 1S and 2S).

When data were grouped by material, plastic was the main contributor to the dissimilarities among sites (SIMPER: 44%–91%) with the exception of the pair PA vs. SB where instead glass, which was more abundant at SB (34%) contributed the most, followed by plastic (29%), which was more abundant at PA.

The site with the highest abundance of plastic was RA while SA had the lowest (Table 1S). Glass was the highest in SB and was present in RA, TA and PA. Metal occurred at the highest abundance at RA and was present at all sites except for BO and SA. Paper was present at all sites with the exception of BO. The site with the highest abundance of paper was RA. Ceramic was only present at SB.

When data were grouped by sources, undifferentiated debris was the main contributor to the dissimilarities among sites (SIMPER: 28.87%–72.29%). Fishing (13.4%–26.4%), tobacco (11.35%–27.6%), medical/hygiene (13.4%–21.3%) and food packaging (12.9%) also had an important contribution to the dissimilarity between few pairs of sites (Table 2S). Sites with the lowest abundance of debris coincided with the lowest undifferentiated debris and this increased the contribution of other types of debris with recognizable sources.

Plastic items less than 5 mm in size class (microplastics) were present at all sites. In total, 2013 pieces of microplastic were recorded with an overall average of 335.5 particles m^{-2} (Fig. 4). Other studies reporting abundances per unit of surface (m^2) found very variable concentrations (Van Cauwenberghe et al., 2015 and references therein). For example, in South Korea (during the rainy season) an average of 27606 items m^{-2} was recorded (Jang et al., 2014). Along Portuguese beaches, highest microplastic concentrations have been observed in autumn, peaking in central sites with more than 3500 items m^{-2} (Antunes et al., 2018). Our results show that sites with the highest abundances of marine debris were also those with the highest quantities of microplastics (RA, BO, MI; see Supplementary materials, Table 3S). In contrast with previous studies, the most abundant types of microplastics were fragments, followed by filament (Claessens et al., 2011; Frias et al., 2016). It is estimated that one of the main sources of contamination by microplastic filaments is wastewater resulting from cloth washing machines. This is not surprising since evidence shows that each garment can produce > 1900 filaments per wash (Browne et al., 2011; Frias et al., 2016). Pellets were present at RA, BO and MI and absent at all other sites. Industrial pellets were the predominant type of pellets but few cosmetic-like pellets were present. All foam found was styrofoam, a type of material frequently used in the fishing industry.

Site had a significant effect on microplastic abundance (PERMANOVA: $F(7,16) = 8.019$; $p = 0.0001$). Two pairwise comparisons were significant: SA vs RA ($t(7) = 5.47$, $p(MC) < 0.00179$) and BO vs SA ($t(7) = 6.32$, $p(MC) < 0.00179$; Bonferroni: $\alpha = 0.00179$). Multivariate dispersion was homogeneous among levels (PERMDISP: $F7,16 = 3.02$; $p = 0.290$). The main contribution to the dissimilarities among sites was either fragment (SIMPER: 38.6%–63.8%) foam (30.5%-51.1) or filament (30.5%–51.1%). Pellet was only relevant for the dissimilarity between the pair BO and TA (25.13%). The dissimilarity between RA and BO was mostly explained by the category fragment.

Principal coordinate analysis (PCO) did not show a geographic pattern (Fig. 5). Replicates within each site clustered together. RA and BO grouped closely together and were slightly isolated from the remaining sites, which is consistent with the previously described SIMPER analysis.

Recent studies have discussed the need for specific analytical methods to avoid misidentification problems in the visual microscopic analysis of small microplastic (Remy et al., 2015; Rochman et al., 2016). Materials such as viscose, natural cellulose and lignin, can easily be mistaken for synthetic fibers when using only visual examination. There are a range of laboratory techniques that can increase analysis certainty such as pyrolysis-gas chromatography (GC) in combination

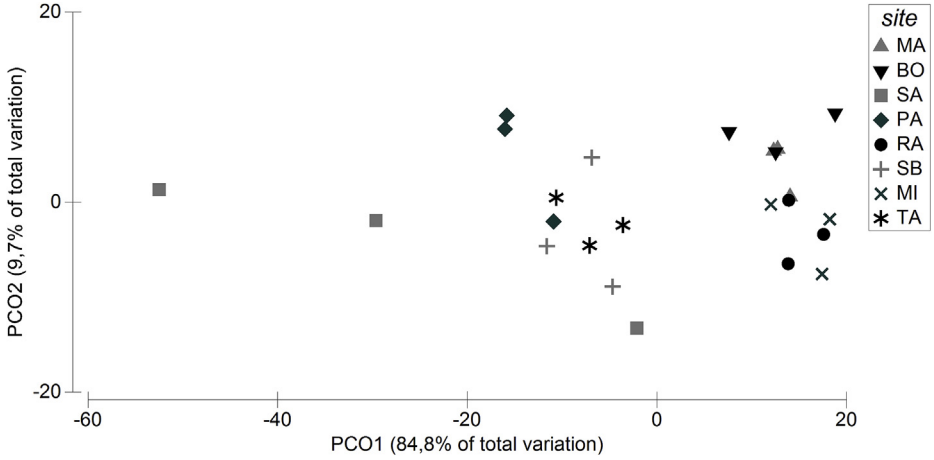

Fig. 3. Principal coordinate analysis (PCO) based on a Bray – Curtis similarity matrix for debris source at each site.

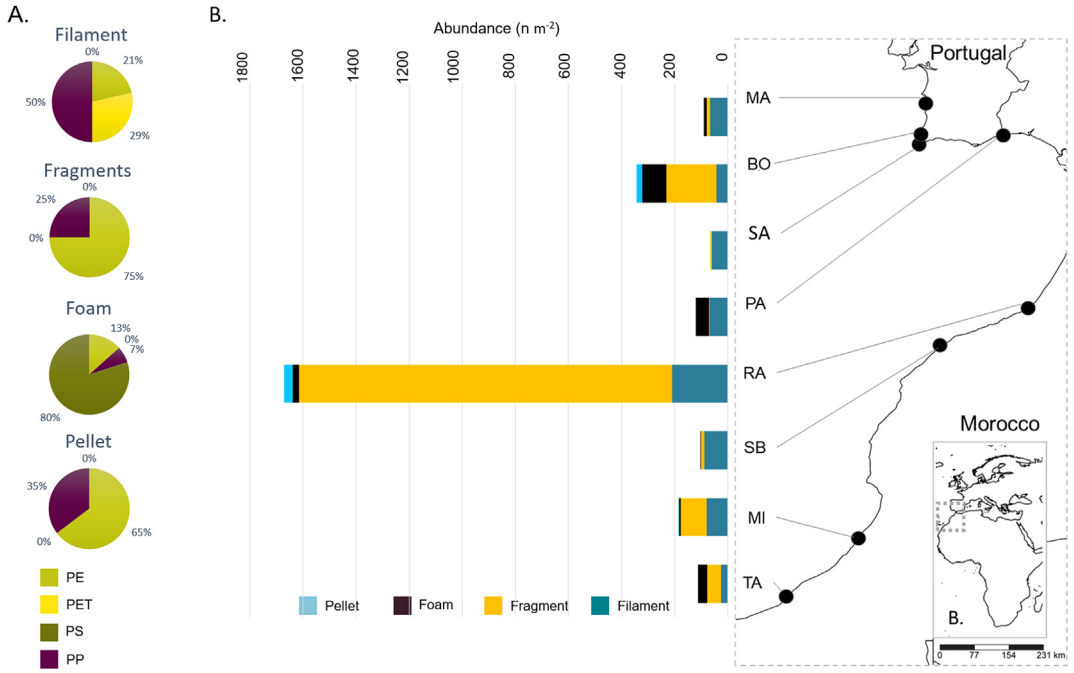

Fig. 4. Predominant polymers (A.) and categories (B.) of microplastic. A.- Proportion of each polymer per microplastic category described by color coded pies. B.- Mean abundance and proportion of each microplastic category at each site. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

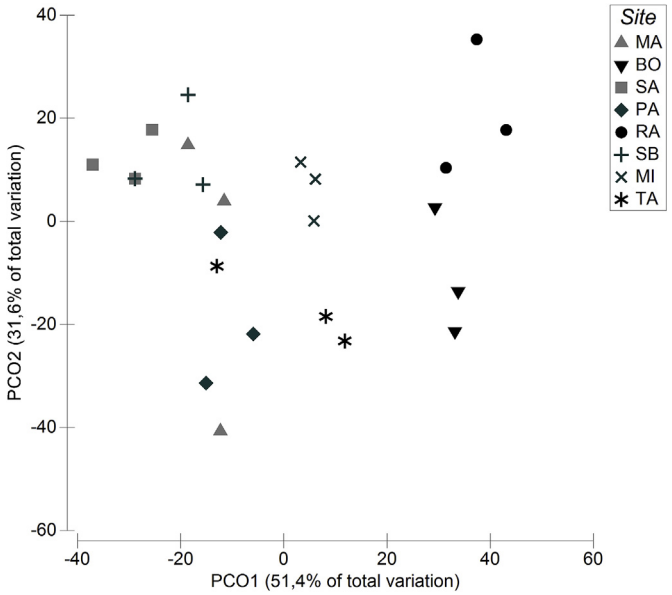

Fig. 5. Principal coordinate analysis (PCO) based on a Bray – Curtis similarity matrix for microplastic source at each site.

with mass spectrometry (MS), infrared (IR), or Fourier-transform infrared (FTIR; Collard et al., 2015; Remy et al., 2015; Wesch et al., 2016). The lack of use of such techniques may overestimate microplastic contamination and create a bias on the overall understanding of sources and impacts of this pollutant (Wesch et al., 2016). In addition, information on polymer type can further narrow down potential sources of plastics into the environment. In our study, combined Raman and FTIR analysis allowed the identification of the polymer type of 83% of microplastic subsamples, for the remaining 17% polymer type was unidentifiable. The predominant polymer differed among microplastic categories (Fig. 4). The predominant microplastic category, Filament was the most abundant category recorded and was mainly composed of Polypropylene (PP, 50%), Polyethylene terephthalate (PET, 29%) and Polyethylene (PE, 21%). This is in contrast with what was previously found subtidally on the south coast of Portugal (Frias et al., 2016), where 80% of microplastic filaments were rayon (a semisynthetic cellulose based polymer) and only the remaining 20% were PP. Fragment, which was also a very abundant category, was mostly composed of PE (75%) and PP (25%). Polystyrene (PS) was the predominant polymer for foam particles (80%) and industrial pellets were mostly made of PE (65%) and PP (35%).

This study provides a solid baseline data of litter and microplastic prevalence across a wide latitudinal range and over regions for which information is lacking. Morocco ranks as 18th in the list of countries that mismanage plastic waste and it is estimated to generate 0.05–0.18 MMT/year of plastic marine debris (Jambeck et al., 2015), a rank equivalent to what all the 23 coastal countries of EU would score together. This is mainly attributed to social factors such as the size of coastal populations, state of the economy, total waste generation and the percentage of plastic waste, and waste mismanagement (Jambeck et al., 2015). However, we show that, although spatially variable, the distribution and composition of debris and microplastics did not follow geographical or regional patterns neither was positively correlated to the number of inhabitants of the nearest populated area (Figs. 1 and 4; Table 1). These findings are in accordance with results from global surveys (e.g., Browne et al., 2011) and small spatial scale studies (Nel and Froneman, 2015).

In addition, we stress the importance of (1) implementing accepted, widely recognized protocols on beach surveying and replicates (Araújo et al., 2006; Ryan et al., 2009), (2) using multivariate analyses to provided critical information on large scale composition and distribution of debris (Underwood et al., 2017) and (3) categorizing plastic types according to polymer type; the application of Raman and FTIR spectroscopy is crucial to discriminate between plastic and natural items, and can provide a potential indication of plastic particle sources and impacts (Desforges et al., 2014; Nicastro et al., 2018).

CRediT authorship contribution statement

N. Velez: Investigation, Data curation, Writing - original draft. **G.I. Zardi:** Conceptualization, Investigation, Writing - review & editing, Funding acquisition, Supervision. **Roberto Lo Savio:** Investigation, Validation, Formal analysis. **Christopher D. McQuaid:** Investigation, Validation. **Ugo Valbusa:** Investigation, Validation. **B. Sabour:** Investigation, Validation. **K.R. Nicastro:** Conceptualization, Investigation, Formal analysis, Writing - review & editing, Funding acquisition, Supervision.

Declaration of competing interest

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

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.marpolbul.2019.110649.

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