CleanAtlantic

Tackling Marine Litter in the Atlantic Area

WP 5: Monitoring and Data Management DELIVERABLE 5.5. - Evaluation of marine litter as transport facilitator for nuisance biota



WP	5 - MONITORING AND DATA MANAGEMENT
ACTION	5.5. EVALUATION OF MARINE LITTER AS TRANSPORT FACILITATOR FOR NUISANCE BIOTA
LAST UPDATED	30/04/2021
VERSION	1
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Evaluation of marine litter as transport facilitator for nuisance biota

INTRODUCTION

Most of the world's population lives within 100km of the ocean, and nearly 75% of all large cities are located on the coast (Brown et al., 2006). These coastal ecosystems are therefore of great importance because they are fundamental for human services on biological, social and economic grounds (Cardinale et al., 2012; Garcia Rodrigues et al., 2017). They sustain a variety of terrestrial and marine fauna and flora, and contribute to the economic sector through tourism, transportation, coastal development and resource extraction (Brown et al., 2006; Garcia Rodrigues et al., 2017). This human dependence on ocean and coastal ecosystems has resulted in their gradual modification, through direct and indirect **pressures** (Halpern et al., 2008).

Biological invasions by **non-indigenous species (NIS)** are one of the greatest environmental and economic threats and a leading cause of biodiversity loss worldwide (Diagne et al., 2021; Nentwig, 2007; Pimentel et al., 2000). In the marine system, biological invasions are largely concentrated in coastal areas, especially in port cities and tourist destinations, where the rate of detected invasions has significantly increased in the last two decades (Bailey et al., 2020; Ruiz et al., 2000). Moreover, coastal development resulted in disturbance agents, such as habitat modification and chemical pollutants that can increase invasion susceptibility (Canning-Clode et al., 2011; Ruiz et al., 2009). In the oceans many invasions have resulted from commercial shipping, stemming from the unintentional transfer of several species of animal and plant in **ballast water** and **hull fouling** (Ruiz et al., 2000). The majority of these transfers are from harbor to harbor, due to the nature of **marine traffic**, and thus typically involve estuarine species with striking euryhaline and eurythermal adaptations (MacIsaac et al., 2002; Ruiz et al., 2000). Once introduced, many NIS flourish on the vast amount of artificial hard substrate (e.g. floats, piers, docks, pontoons, buoys, or seawalls) growingly available in modern times in these environments (Bulleri and Chapman, 2010), due in part to their physiological and behavioral abilities to outcompete native species (Castro et al., 2021).

The accumulation of debris in the ocean also severely affects ocean and coastal ecosystems, as its ingestion and entanglement directly impacts marine life (Barnes et al., 2009). Furthermore, recent research indicates that **marine debris** is both a growing vector for the introduction of NIS, with transoceanic rafting already likely intensifying species invasions worldwide (Carlton et al., 2017), and a potential cause of marine diseases, whereby plastic waste in the oceans promotes microbial colonization and transmission of pathogens, as recently suggested for coral reef disease outbreaks (Lamb et al., 2018).

Islands ecosystems, in particular, are often biodiversity hotspots (Pouteau and Birnbaum, 2016) that not only are acutely sensitive to disturbances but also are experiencing rapid increases in tourism and other human pressures. In these coastal ecosystems, key human-driven stressors include **biological invasions**, marine debris, coastal development and climate change (Brierley and Kingsford, 2009; Halpern et al., 2008), all of which have diverse and significant consequences on biodiversity (Duffy et al., 2017; Fanin et al., 2018).



STUDY OBJECTIVES

In this context, this task examined the role of marine litter as a vector facilitator for dispersal of attached and mobile species in the Madeira Archipelago, Portugal. For this, we applied bioforensic techniques (biogeographic and systematic evidence) for assessing marine debris and quantifying epibiota on these debris. Using the Madeira islands as a model system, we specifically asked the following questions:

- i) Is marine litter showing up at Madeira waters?
- ii) Is marine litter a viable vector for NIS in Madeira?
- iii) What are the most represented biota colonizing marine litter?
- iv) What is the percentage of NIS colonizing marine litter?
- v) From where this litter (and NIS) are likely coming from?

METHODOLOGY

Study area

This study was conducted in the Madeira Archipelago, Portugal, which has been under increased pressure by human related activities over the last decades (e.g. demographics, tourism, coastal and urban development). The Madeira Archipelago is a volcanic islands system located in the NE Atlantic Ocean on the southwest of continental Europe and 700km off the Moroccan coast that comprises two inhabited islands: Madeira and Porto Santo. Madeira Island is the largest island with 144 km of coastline whereas Porto Santo Island is located about 42 km northeast of Madeira Island with about 33 km of coastline (Ramalhosa et al., 2019).

Opportunistic Sampling

During a 3-year period, we established a cooperation protocol with relevant stakeholders (e.g. fishers, SCUBA diving centers, and whale and bird watching companies) to evaluate if marine debris is showing up in Madeiran waters (NE Atlantic) and determine if it acts as an introduction vector of NIS. The surveys were opportunistic and depended on the operations and time availability of the stakeholders involved. Once detected an object in the sea or its proximity (beaches), stakeholders recorded its GPS position, photographed, collected and transferred it to the laboratory facilities of MARE-Madeira for further analysis. Each item was classified in terms of material and measured, and the biological hosts were sampled. All litter items were classified according to the OSPAR methodology for 100 m transects (OSPAR Commission, 2010). All macroalgae and macro-invertebrates (sessile and mobile) colonizing litter items were identified to the lowest possible taxonomic group and later assigned to four categories: native, NIS, cryptogenic (i.e. unknown origin) in accordance to literature and several current databases (AquaNIS, 2013; Fofonoff et al., 2020), or unresolved (based on an inability to identify to species level).

Object Origin Estimates

The origin of detected items was then estimated based upon three signatures: identification marks, biological, and physical oceanography. Specifically, object origin was estimated as follows:



A) Identification marks signature

This signature referred to any identification marks referring to insignia, manufacturer's marks or serial numbers found in each litter item.

B) Biological signature

The biological signature of each litter item referred to the biogeographic origin of the NIS found on each object. For each NIS present in each marine litter type, the probable native distribution range (origin) was assigned using the 18 large-scale IUCN marine bioregions as defined by Kelleher et al. (1995), and later modified by Hewitt & Campbell (2010) as follows: 1 – Antarctica; 2 – Arctic; 3 – Mediterranean including the Black and Azov Sea; 4 – North West Atlantic; 5 – North East Atlantic; 6 – Baltic; 7 – Wider Caribbean Sea; 8 – West Africa; 9 – South Atlantic; 10 – Central Indian Ocean; 11 – Arabian Seas; 12 – East Africa; 13 – East Asian Seas; 14a&b – South Pacific & Hawaii; 15 – North East Pacific; 16 – North West Pacific; 17 – Southeast Pacific; 18 – Australia and New Zealand. For assessing species potential native distribution, research articles were used (Appendix A, Table S1).

C) Physical oceanography signature

The Connectivity Modeling System (CMS v2.0) was used to track passive particle trajectories in the North Atlantic region. The CMS operates in offline mode, applying the 3D velocity fields (u, v, w) of the hydrodynamic velocity field to each particle using a Runge–Kutta 4th order advection scheme (Paris et al., 2013; van Sebille et al., 2012). In this study, CMS was coupled with the GLORYS12V1 reanalysis product, developed by Mercator-Océan in the framework of Copernicus Marine Environment Monitoring Service (CMEMS). This reanalysis product is based on the same ocean circulation models as the Nucleus for European Modeling of the Ocean (NEMO) framework. It is forced at the surface by the ERA-Interim wind reanalysis from the European Center for Medium-Range Weather Forecasts (ECMWF) and uses in-situ satellite data for data assimilation. With a ~9.25 km (1/12°) horizontal grid resolution and considering only the surface level (0.49 m), the study domain covers an area between 0 to 70°N in latitude and -80 to 20°W in longitude. A backward-intime simulation was conducted to determine the most probable origins of the floating macro litter reaching the Madeira Archipelago, carried for the period 2012-2017 with a daily interval. Each item corresponds to a set of passive particles, meaning that modelled particles do not consider variations in shape and density. Particles were released in 25 points, representative of the location (and time) where the items were collected (Appendix B, Table S2). A total of 50 particles were released at each release point, summing up to a total of 1250 particles. The trajectories were tracked up to a maximum of 2 years prior to their release date. In order to compare modelled results with in-situ transport pathways, the trajectories of surface drifting buoys that intercepted the Madeira Archipelago (delimited area in Fig. 8) were computed. These drifter trajectories are part of the Global Drifter Program (GDP) from the National Oceanic and Atmospheric Administration (NOAA). GDP drifters are drogued at 15 m depth so that they follow near-surface currents and have been deployed throughout the global ocean since 1988, maintaining an array of \sim 1,250 drifters throughout the years (Lumpkin and Pazos, 2007). From a total of 6536 drifters with recorded positions in the North Atlantic, only 129 intersected the delimited area around the Madeira Archipelago. To note is that due to a mechanical failure of the buoy-drogue connection, nearly 30 and 90% of GDP drifters lose their drogues in the first 3 and 18 months after deployment, respectively (Grodsky et al., 2011). Although this is estimated to increase the wind drag from 0.1 to





1% of the wind speed (Niiler et al., 1995), drogued and undrogued drifter trajectories were not differentiated in the analysis.

Data analysis

All data regarding category, material, litter type class and geographic position of found items were compiled together with information on the diversity of taxa detected. Taxa were labelled and grouped by major taxonomic groups (i.e. macroalgae, arthropoda, bryozoa, cnidaria, mollusca, annelida (polychaeta), sipuncula, chordata (tunicate), echinodermata, foraminifera, platyhelminthes, porifera and entoprocta). Based on identified species and distinct unidentified species, the total number of Operational Taxonomic Units (OTUs) was calculated for: i) each of the major taxonomic groups, and; ii) each of the four status categories (i.e. NIS, native, cryptogenic and unresolved). The total number of OTUs were then used to assess diversity richness per taxonomic and status.

A Bray-Curtis similarity matrix of major taxonomic groups' diversity richness (i.e. OTUs number per taxonomic group) was used to assess how they are differently associated with floating, beached and seafloor litter. Additionally, a Whittaker's Index of Association matrix (Clarke and Gorley, 2006; Zuur et al., 2007) was generated to assess the resemblance among major taxonomic groups, providing insight in the similarity of the OTU richness distribution of these groups.

Based on the presence-absence data of each OTU, the most frequently found taxa for floating, seafloor and beached litter was calculated to identify which OTUs were more common to hitch-hike on each of these litter types.

The proportion of NIS, native and cryptogenic OTUs occurring in floating, seafloor and beached litter was calculated by excluding all unresolved taxa, standardizing the total number of OTU and calculating the percentage of OTUs for each status.

All data analysis was performed in Primer V7 with PERMANOVA extension (Anderson et al., 2008; Clarke and Gorley, 2006).

RESULTS

During this 3-year study, a total of 71 litter items were registered in the archipelago of Madeira, from which 38 were found floating, 14 items were collected from the beach and 19 items were found at the seafloor (Fig. 1). Opportunistic sampling showed that floating litter was mostly detected on the south coast of Madeira Island as this is the area where the engaged stakeholders operate. In addition, seafloor items were sampled on the eastern part of Madeira Island and Porto Santo Island. Finally, our opportunistic sampling detected beached items at both the south and north coasts of Madeira Island (Fig. 1).



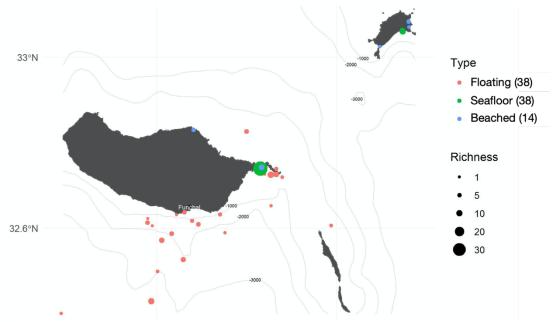


Figure 1 – Location of the 71 litter items from three categories (Floating, Beached, Seafloor) detected in Madeira waters during the 3-year period of the present study. Species Richness estimates per litter item is also shown.

Floating litter was mostly characterized by buoys and fishing gear of plastic nature. Items found at the seafloor (shallow waters < 20 meters depth) were mostly glass bottles, pieces of plastic containers and metal. Finally, beached litter was mostly characterized by packaging materials, ropes and plastic caps.

A total of 97 taxa were identified in the surveyed litter items (Fig.2, Appendix C, Table S3). Floating litter was mostly dominated by mollusks, arthropods and macroalgae (Fig. 2 and Fig. 3). Seafloor litter items were significantly dominated by bryozoans, followed by arthropods, macroalgae, mollusks and annelids (polychaeta) (Fig. 2 and Fig. 3). Finally, beached litter items were mostly colonized by bryozoans and arthropods (Fig. 2 and Fig. 3).

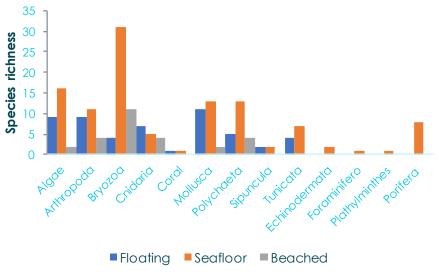


Figure 2 - Most representative taxonomic groups colonizing the three types of litter (Floating in blue, Beached in grey and Seafloor in orange) detected in Madeira waters during the present study.



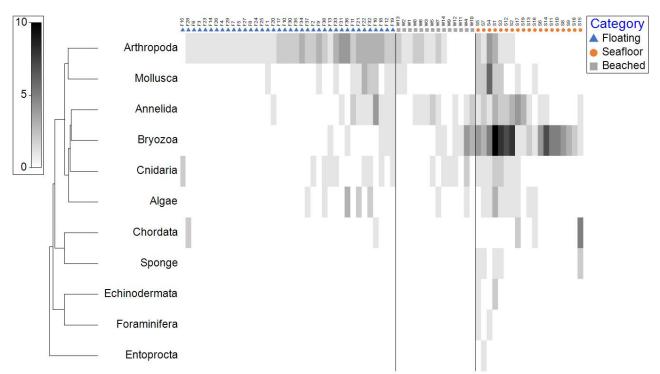


Figure 3 -Shadeplot showing differences in community composition across the three different types of litter (i.e. Floating, Beached, and Seafloor) found in Madeira during the present study.

In addition, a total of 16 non-indigenous species (NIS) and 26 cryptogenic species were identified in the 71 litter items surveyed during the present study across the 3 different litter types (Table 1). In particular, the floating litter was colonized by 6% of NIS and 65% of cryptogenic species, the seafloor litter by 17% of NIS and 14% of cryptogenic species, and the beached litter by 5% of NIS and 41% of cryptogenic species (Fig. 4). Among the most abundant species colonizing the different litter types, 3 cryptogenic species belong to the floating litter, 1 NIS to seafloor litter and 5 cryptogenic species to beached litter (Fig. 5).

Table 1 - Frequency (%) of non-indigenous species (NIS) found in different marine litter categories: Floating (F), Beached (B) and
Seafloor (S) on the south coast of Madeira Archipelago during this 3-year study. Species in bold indicate a new record for the Madeira
Archipelago.

	Frequ	iency (%	6)				
Таха	F B S			IUCN Bioregion	Literature		
Annelida (1)							
Amphinome rostrata	2.6	0.0	0.0	10. Central Indian Ocean	This study ; Wehe and Fiege, 2002		
Arthropoda (1)							
				14. S Pacific Ocean; 15. NE Pacific Ocean; 16.			
Balanus trigonus	5.3	0.0	21.1	NW Pacific Ocean; 17. SE Pacific Ocean	Carlton et al., 2011		
Bryozoa (5)							
Bugula neritina	0.0	0.0	5.3	Native region not known at this time			
Celleporaria inaudita	0.0	0.0	5.3	11. Arabian Seas; 13. E Asian Seas	McCann et al., 2019		
Parasmittina alba	0.0	0.0	15.8	9. S Atlantic Ocean	Ferrario et al., 2020 Carlton and Eldredge,		
Schizoporella errata	0.0	7.1	5.3	3. Mediterranean	2015		
					Pa		



Schizoporella unicornis	0.0	0.0	47.4	3. Mediterranean; 5. NE Atlantic	Hayward and Ryland, 1999
Chordata (3)					
				7. Wider Caribbean; 8. W Africa; 9. S Atlantic	
Botrylloides niger	0.0	0.0	5.3	Ocean	Sheets et al., 2016
Clavelina lepadiformis	0.0	0.0	5.3	3. Mediterranean; 5. NE Atlantic;	Canning-Clode et al., 2013
Distaplia corolla	0.0	0.0	15.8	7. Wider Caribbean	Canning-Clode et al., 2013
Cnidaria (1)					
Exaiptasia diaphana	2.6	0.0	0.0	3. Mediterranean	Canning-Clode et al., 2013
Mollusca (2)					
					Branch and Stefanni,
Mytilus galloprovincialis	2.6	7.1	0.0	3. Mediterranean; 5. NE Atlantic;	2004; Gestoso et al., 2015
				4. NW Atlantic; 7. Wider Caribbean; 9. S	
Pinctada imbricata	2.6	0.0	5.3	Atlantic Ocean	Holmes et al., 2015
Porifera 2)					
Mycale (Carmia)					
senegalensis	0.0	0.0	21.1	8. W Africa	Canning-Clode et al., 2013
Prosuberites longispinus	0.0	0.0	5.3	3. Mediterranean	Goodwin and Picton, 2011

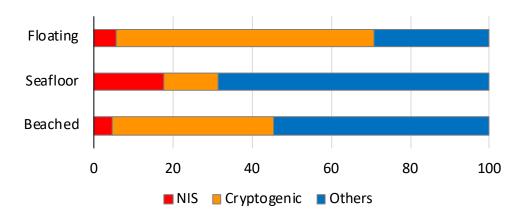


Figure 4 – Proportion (in %) of non-indigenous species (NIS), cryptogenic species (i.e. of unknown origin) and others (includes native species and unresolved) across the three different types of litter (i.e. Floating, Beached, and Seafloor) found in the Madeira Archipelago during the present study.



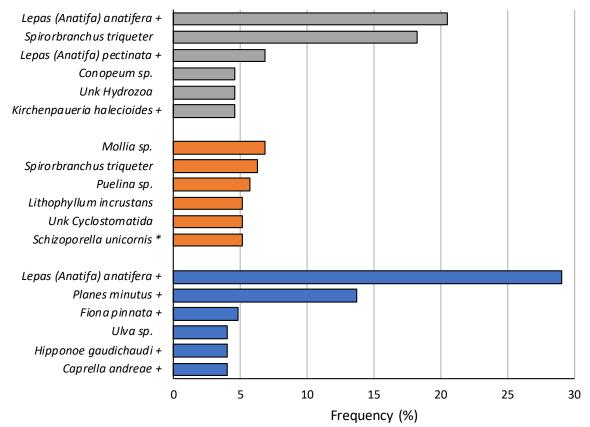


Figure 5 – Frequency of the most representative NIS (*) and cryptogenic (+) species (considering the number of objects where the species was found) across the three different types of litter (i.e. Floating, Beached, and Seafloor) found in the Madeira Archipelago during the present study.

Moreover, from all items of marine litter collected, approximately 10 floating objects had some identification marks that allowed us to backtrack its probable origin (i.e. country or vessel). Amongst these objects (See appendix D for some examples), there was a lifebuoy (#F1 object: NS-25, manufacture date 2004 Jun Nippon Sengu CO, LTD Tokyo, Japan) found in Madeira waters, with the name of the vessel Ardmore Seamaster (tanker). We were able to identify its call sign code, IMO number, Marshall Island flag, port of registry Majuro, and other additional information about the last port of call, country, destinations and, date/place of last dry dock and date of next dry dock due. Other objects showed a few marks allowing us to detect origin countries (e.g. Canada, Spain or USA). However, most litter items found had no identification marks.

Overall, most NIS collected in the present study were native to Mediterranean (22.2%), North West Atlantic and wider Caribbean (11.1% each), North West Pacific and West Africa (7.4% each), followed by other regions less represented (Fig. 6). Specifically, NIS colonizing floating litter were native for 16.7% to the Mediterranean and North West Pacific Ocean and 8.3% to other bioregions. Regarding seafloor litter, there was a higher signature of the Mediterranean (17.3%), South Atlantic Ocean and wider Caribbean (13.0%), North West Pacific, West Africa and North East Atlantic (8.7% each). Finally, for the beach litter, the Mediterranean and North East Atlantic were the only bioregions observed, with 66.7% and 33.3% of percentage of occurrence, respectively.



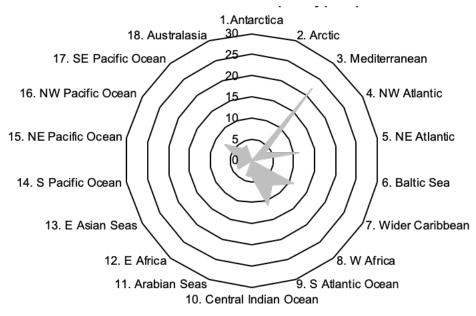


Figure 6 - Frequency of the origin of the non-indigenous species (NIS) colonizing floating litter in Madeira waters during the course of the present study. Potential native distributions were based on IUCN Bioregions (Kelleher et al., 1995) and several publications (Appendix S2). Note that some species have more than one native origin.

Results from the lagrangian simulation suggest the possible origins of the plastic items collected around Madeira islands, showing that virtual particles intercept the archipelago mostly from the North and from the West and tend to follow the large-scale circulation of North Atlantic Subtropical Gyre before reaching the archipelago (Figure 7). Some of these particles connect the archipelago to the American Continent within the simulation period (2 years), while others meander along the currents of the North Atlantic Ocean before reaching the archipelago.

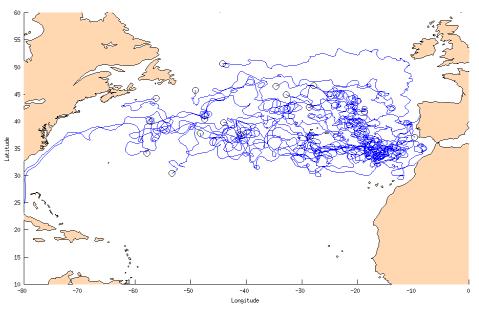


Figure 7 - Lagrangian particles trajectories tracked backward in time for up to 2 years before their release date. The black circles represent the particles' final position. A random particle was selected out of every 50 released per point for the sake of clarity.



In terms of in-situ drifter trajectories, GDP drifters (Figure 8) showed similar patterns as the modelled particles (Figure 7). From the 129 drifters that intercepted the delimited area around the Madeira Archipelago, the majority approached from the north and tended to follow the large-scale ocean circulation of the North Atlantic Subtropical Gyre, connecting the archipelago to the North American continent in 2 years minimum.

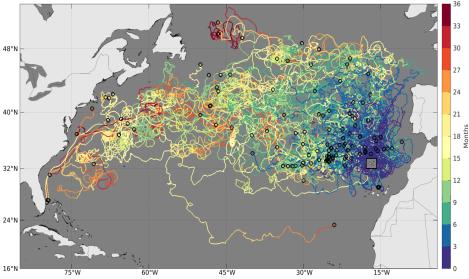


Figure 8 - Drifting buoys trajectories and travel time from the deployment (black circles) to the intersection of Madeira's delimited area.

FINAL CONSIDERATIONS

Biological invasions by non-indigenous species (NIS) are one of the greatest environmental and economic threats of global relevance, and, along with habitat destruction, a leading cause of biodiversity loss worldwide. In the ocean, biological invasions are largely concentrated in highly vulnerable coastal communities, where the rate of detected invasions has significantly increased during the last decades (Canning-Clode, 2015). In the Madeira Archipelago (NE Atlantic), the diversity and distribution of NIS has received some level of attention in recent years due to ongoing monitoring surveys, particularly in marinas on the southern coast of Madeira Island (Ramalhosa et al., 2019). While maritime traffic has received some level of attention (Castro et al., 2020), remaining uninvestigated in this Northeastern Atlantic theater is the expanding global role of marine litter in potentially mediating new invasions (Carlton et al., 2017). While ocean rafting is an ancient and natural phenomenon, modern-day marine debris generated by human activity consists largely of permanently floating materials (plastics, including fiberglass and polymers such as Styrofoam and PVC) which are non-biodegradable and thus serve as permanent rafts that are now potentially vastly extending the dispersal of marine species.

In this context, within the scope of the CleanAtlantic project and the established cooperation with local stakeholders (e.g. fishers, SCUBA diving centers and whale and bird watching companies), we were able to obtain valuable information on the litter items collected during a three year period



around the coasts of Madeira and Porto Santo islands. In particular, we collected and analyzed a total of 70 items (floating, on the seafloor or beached) that were hosting 97 different taxa.

The study has an opportunistic nature and is limited to Madeira, bestowing some limitations to extrapolations and to other regions. However, a total of 97 taxa were identified associated to the inspected items, corroborating that litter items can host and harbour numerous marine species. It is also notable that approximately 6% of the taxa colonizing floating litter and found in Madeira waters were NIS, showcasing how floating debris can act as rafts and facilitate the spread of species to non-native ranges. Interesingly, 17% of taxa found in seafloor litter were also NIS, suggesting that they may have also been colonized locally and raising questions on whether litter items serve as better suited substrate for NIS than natural substrates. Beached litter also had evidences of colonizing biota with 5% of NIS, which also corroborates the ability of either enabling NIS to arrive to shore or of being colonized by NIS locally. These numbers are likely underestimated given the significant number of recorded species classified as cryptogenic (i.e. of unknown origin), particularly in floating litter items (65%; Figure 4). This highlights the role of marine litter as a viable vector for the introduction species in Madeira waters.

The origin of detected items was estimated based upon: i) identification marks ii) biological signature referring to the biogeographic origin of the biota colonizing litter items; and iii) physical oceanography signature through lagrangian transport modeling to determine the most probable origin(s) of these litter items. With regards to identification marks, we detected some insignia, manufacturer's marks or serial numbers in only a small fraction (~14%) of litter items found in Madeira and Porto Santo. Although it is not expected that this method provides a robust response about the origin of litter items in the short run, it could provide solid results in long term monitoring. We therefore recommend future studies and actions to initiate an open access database on indentification marks found in litter items (floating, seafloor and beached).

In addition, most NIS collected in litter items during the course of this study were native to the Mediterranean, North West Atlantic, Caribbean, North West Pacific and West Africa. There seems to be a partial match between regions that are sources of NIS (Castro et al., 2020) and the predicted sources of litter items, substantiating the need to increase research on this field. Future research should focus, not only in standard protocols and dedicated monitoring but also the development of integrated approaches that enable to resolve pending questions, including: a) settlement and colonization patterns in the Madeira Archipelago on multiple litter types; b) quantities of litter items found with no associated biota; c) invasion risk associated to most common taxa found in litter items; d) ratio of viable transported biota (ie. live organisms that can viably establish themselves in the new region).

In respect to the lagrangian particles, their arrival to Madeira Archipelago is mostly determined by two ocean currents: the easternmost branch of the eastward-flowing Azores Current that drifts between 30°N and 40°N (Hernández-Guerra et al., 2001); and the southward-flowing Portugal Current that intercepts Madeira from the north (Pérez et al., 2001). These two currents interact with one another as they flow past the Madeira Archipelago and feed the southward-flowing Canary Current downstream (Klein and Siedler, 1989; Stramma, 1984; Zhou et al., 2000), making the



archipelago significantly exposed to the seasonal oscillations of the Azores, Portugal, and Canary Currents (Caldeira et al., 2002; Martins et al., 2002). At the far-field, lagrangian particles reach the North American continent through the North Atlantic Current (evidenced by the northernmost trajectories) and through the initial (western) branch of the Azores Current (evidenced by the southernmost trajectories; Figure 7). These two currents originate from the offshore extension of the Gulf Stream, which is marked by several bifurcations at ~55°W (Klein and Siedler, 1989). The fact that two particle trajectories follow the Gulf Stream up to its initial extension in less than two years (Figure 7) corroborates well with GDP drifter trajectories (Figure 8). Such results demonstrated that the archipelago is significantly vulnerable to North American sources, which together with Central America, account for 64–66% of the debris floating in the North Atlantic (Lebreton et al., 2012). The considerable amount of GDP drifter trajectories that intersect the Madeira Archipelago (129) and share the same general patterns illustrated by the modelled particles trajectories (25) is a good validation of modelled results. Most of the GDP drifter trajectories were deployed along the Azores Current, revealed by the narrow horizontal band of drifter deployments north of Madeira (black circles in Figure 8). Extending from ~10 to 24ºW (Pérez et al., 2001), the influence of the offshore branch of the Portugal Current is also revealed by the southward trajectories west of the Iberian coast.

During this 3-year study, our surveys were opportunistic and therefore depended on the operations and time availability of the stakeholders involved (i.e. fishers, SCUBA diving centers, whale watching and bird watching companies). Our results showed that floating litter was mostly detected on the south coast of Madeira Island as this is the area where the engaged stakeholders operate. Future efforts should continue and enhance engagement with stakeholder, but research should also explore dedicated sampling following standard protocols, both the south and north coasts of Madeira Island as well as Porto Santo Island.

In addition, this study indicates that Madeira is receiving litter items from different parts of the world, and some of these objects can be considered vehicles for a certain number of "hitch-hiking" species. However, certainly Madeira is also exporting litter, that in turn can be transporting local species to different regions. This issue was not subject of research in the current study, but should be addressed in future research.

Finally, the interplay between marine litter and NIS is receiving attention from the ICES Working Group on Introductions and Transfers of Marine Organisms (WGITMO). In this context, the first author of the current report, which is also a member of WGITMO, is coordinating a Term of Reference entitled "*Investigate the role of human-produced marine debris as a vector and facilitator for the introduction and spread of non-indigenous species (NIS)*". For the next 3 years, the WGITMO expert group will try to advance research and identify knowledge gaps on marine debris-NIS interactions as well as develop synergies with other working groups.



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APPENDIX A

Table S1 - References for	notential native	distributions for the	species used in the analysis
Table 31 - References for	polential native		species used in the analysis

Species	Reference for possible native ranges
Amphinome rostrata (Pallas, 1766)	(Wehe and Fiege, 2002)
Balanus trigonus Darwin, 1854	(Carlton et al., 2011)
Bugula neritina (Linnaeus, 1758)	Native region not known at this time
Celleporaria inaudita Tilbrook, Hayward & Gordon, 2001	(McCann et al., 2019)
Parasmittina alba Ramalho, Muricy & Taylor, 2011	(Ferrario et al., 2020)
Schizoporella errata (Waters, 1878)	(Carlton and Eldredge, 2015)
Schizoporella unicornis (Johnston in Wood, 1844)	(Hayward and Ryland, 1999)
Botrylloides niger Herdman, 1886	(Sheets et al., 2016)
Clavelina lepadiformis (Müller, 1776)	(Canning-Clode et al., 2013)
Distaplia corolla Monniot F., 1974	(Canning-Clode et al., 2013)
Exaiptasia diaphana (Rapp, 1829	(Canning-Clode et al., 2013)
Mytilus galloprovincialis Lamarck, 1819	(Branch and Nina Steffani, 2004; Gestoso et al., 2015)
Pinctada imbricata Röding, 1798	(Holmes et al., 2015)
Magallana gigas (Thunberg, 1793)	(Herbert et al., 2016)
Mycale (Carmia) senegalensis Lévi, 1952	(Canning-Clode et al., 2013)
Prosuberites longispinus Topsent, 1893	(Goodwin and Picton, 2011)



APPENDIX B

Table S2 – Floating macro-debris information provided by nautical companies. Each plastic item has been assigned an
identification, a collection date and the coordinates (longitude and latitude) corresponding to the location where it was captured.

Item	Longitude	Latitude
F1	-16.811	32.589
F2	-16.515	32.606
F3	-16.515	32.606
F4	-17.025	32.622
F5	-16.823	32.632
F6	-16.683	32.653
F7	-17.025	32.614
F8	-17.025	32.614
F9	-17.015	32.429
F10	-16.958	32.587
F11	-16.884	32.609
F12	-17.012	32.606
F13	-17.266	32.400
F14	-16.926	32.527
F15	-16.986	32.572
F16	-16.998	32.498
F17	-16.945	32.632
F18	-16.923	32.637
F19	-16.278	32.726
F20	-16.278	32.726
F21	-16.278	32.719
F22	-16.279	32.826
F23	-16.278	32.727
F24	-16.278	32.725
F25	-16.278	32.739



APPENDIX C

Table S3. List of frequency and percentage of recorded species found in different marine litter categories: Floating (F), Beached (B) and Seafloor (S) on the south coast of Madeira Archipelago during this 3-year study. Taxa were categorized as native (N), non-indigenous species (NIS) and cryptogenic (C) based on literature, or unresolved (U), based on an inability to identify to species level. Species in bold indicate a new record for the Madeira Archipelago.

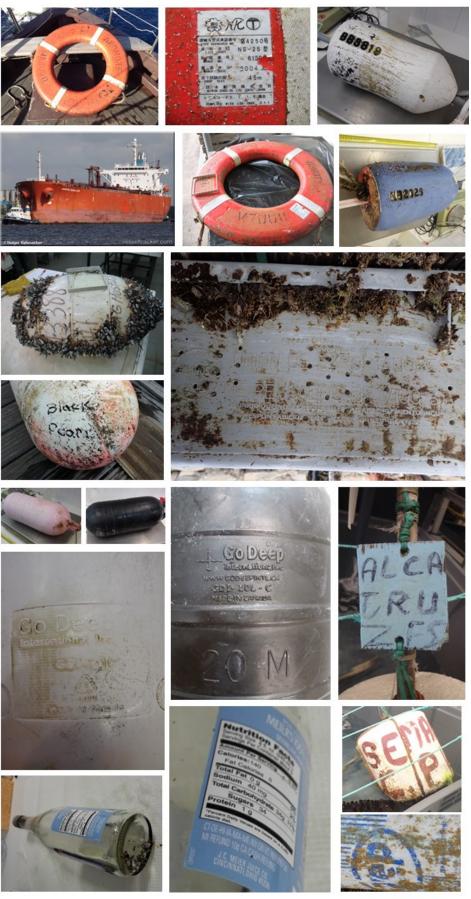
	Authority		Frequency			Per	centage	e (%)	_
Таха		Status	F	В	S	F	В	S	Literature Review
Algae (7)									
Anadyomene stellata	(Wulfen) C.Agardh, 1823	С	1	0	0	2.6	0.0	0.0	(Micael et al., 2014; Neto et al., 2001)
Dasycladus vermicularis	(Scopoli) Krasser, 1898	С	1	0	0	2.6	0.0	0.0	(Neto et al., 2001)
<i>Dictyota</i> sp.		U	0	0	1	0.0	0.0	5.3	
Lithophyllum incrustans	Philippi, 1837	Ν	0	1	9	0.0	7.1	47.4	(Canning-Clode et al., 2013; Levring, 1974; Ramalhosa et al., 2019)
Lobophora sp.		U	1	0	0	2.6	0.0	0.0	
Ulva sp.		U	5	1	1	13.2	7.1	5.3	
Unk Ceramiales		U	1	0	0	2.6	0.0	0.0	
Annelida (11)									
Amage adspersa	(Grube, 1863)	С	0	0	1	0.0	0.0	5.3	(Núnez and Talavera, 1995)
Amphinome rostrata	(Pallas, 1766)	NIS	1	0	0	2.6	0.0	0.0	This study
Hermodice carunculata	(Pallas, 1766)	С	0	0	1	0.0	0.0	5.3	(Núnez and Talavera, 1995)
Hipponoe gaudichaudi	Audouin & Milne Edwards, 1830	С	5	1	0	13.2	7.1	0.0	(David, 2017; Moore, 1903)
<i>Nereididae</i> sp.		U	2	0	0	5.3	0.0	0.0	
Phascolosoma sp.		U	3	0	1	7.9	0.0	5.3	
<i>Sabellidae</i> sp.		U	0	0	1	0.0	0.0	5.3	
Salmicina dysteri	(Huxley, 1855)	N	0	0	4	0.0	0.0	21.1	(Canning-Clode et al., 2013; Langerhans, 1879; Núnez and Talavera, 1995; Ramalhosa et al., 2019)
Serpula vermicularis	Linnaeus, 1767	Ν	0	0	4	0.0	0.0	21.1	(Canning-Clode et al., 2013; Núnez and Talavera, 1995)
<i>Spirobis</i> sp.		U	0	1	2	0.0	7.1	10.5	
	(Linnaeus, 1758)	Ν	3	8	11	7.9	57.1	57.9	(Gestoso et al., 2017; Ramalhosa et al., 2019)

Ammothella sp.	(Hodge, 1864)	U	2	0	1	5.3	0.0	5.3	
Ampithoe sp.		U	0	0	1	0.0	0.0	5.3	
Balanus trigonous	Darwin, 1854	NIS	2	0	4	5.3	0.0	21.1	(Canning-Clode et al., 2013; Chainho et al., 2015; Ramalhosa et al., 2019; Wirtz et al., 2006)
Caprella acanthifera	Leach, 1814	С	0	0	1	0.0	0.0	5.3	(Zeina et al., 2015)
Caprella andreae	Mayer, 1890	С	5	0	0	13.2	0.0	0.0	(Zeina et al., 2015)
<i>Caprella</i> sp.		U	2	0	1	5.3	0.0	5.3	
Eurynome spinosa	Hailstone, 1835	С	0	0	2	0.0	0.0	10.5	(Araújo et al., 2014)
		0	3						(1)(1) (1) (2) (2) (2) (2) (2) (2) (2) (2) (2) (2
Lepas (Anatifa) anatifera	Linnaeus, 1758	С	6	9	0	94.7	64.3	0.0	(Wirtz et al., 2006)
Lepas (Anatifa) pectinata	Spengler, 1793	С	0 1	3	0	0.0	21.4	0.0	(Wirtz et al., 2006)
Planes minutus	(Linnaeus, 1758)	С	7	0	0	44.7	0.0	0.0	(Dellinger et al., 1997)
Unk Amphipoda 1		U	4	1	1	10.5	7.1	5.3	
Unk Amphipoda 2		U	0	0	1	0.0	0.0	5.3	
Unk Hyalidae		U	1	0	0	2.6	0.0	0.0	
Unk Tanaidacea		U	0	0	1	0.0	0.0	5.3	
Bryozoa (26)									
Bryozoa (26) Bugula neritina	(Linnaeus, 1758)	NIS	0	0	1	0.0	0.0	5.3	(Norman, 1909; Ramalhosa et al., 2019, 2017) (Canning-Clode et al., 2013; Ramalhosa et al., 2019; Souto et al.,
	Tilbrook, Hayward & Gordon, 2001	NIS	0	0	1	0.0	0.0	5.3 5.3	(Norman, 1909; Ramalhosa et al., 2019, 2017) (Canning-Clode et al., 2013; Ramalhosa et al., 2019; Souto et al., 2018)
Bugula neritina	• • •								(Canning-Clode et al., 2013; Ramalhosa et al., 2019; Souto et al.,
Bugula neritina Celleporaria inaudita	Tilbrook, Hayward & Gordon, 2001 Souto, Reverter-Gil & Ostrovsky,	NIS	0	0	1	0.0	0.0	5.3	(Canning-Clode et al., 2013; Ramalhosa et al., 2019; Souto et al., 2018)
Bugula neritina Celleporaria inaudita Chorizopora sp.	Tilbrook, Hayward & Gordon, 2001 Souto, Reverter-Gil & Ostrovsky, 2014	NIS U	0	0 0	1 4	0.0 0.0	0.0 0.0	5.3 21.1	(Canning-Clode et al., 2013; Ramalhosa et al., 2019; Souto et al., 2018)
Bugula neritina Celleporaria inaudita Chorizopora sp. Conopeum sp.	Tilbrook, Hayward & Gordon, 2001 Souto, Reverter-Gil & Ostrovsky, 2014 (Linnaeus, 1767)	NIS U U	0 0 0	0 0 2	1 4 0	0.0 0.0 0.0	0.0 0.0 14.3	5.3 21.1 0.0	(Canning-Clode et al., 2013; Ramalhosa et al., 2019; Souto et al., 2018) (Souto et al., 2014)
Bugula neritina Celleporaria inaudita Chorizopora sp. Conopeum sp. Copidozoum tenuirostre	Tilbrook, Hayward & Gordon, 2001 Souto, Reverter-Gil & Ostrovsky, 2014 (Linnaeus, 1767) Hincks, 1880)	NIS U U C	0 0 0 0	0 0 2 1	1 4 0 2	0.0 0.0 0.0 0.0	0.0 0.0 14.3 7.1	5.3 21.1 0.0 10.5	(Canning-Clode et al., 2013; Ramalhosa et al., 2019; Souto et al., 2018) (Souto et al., 2014) (Hincks, 1880; Micael et al., 2019; Norman, 1909)
Bugula neritina Celleporaria inaudita Chorizopora sp. Conopeum sp. Copidozoum tenuirostre Corbulella sp.	Tilbrook, Hayward & Gordon, 2001 Souto, Reverter-Gil & Ostrovsky, 2014 (Linnaeus, 1767) Hincks, 1880)	NIS U U C U	0 0 0 0 0	0 0 2 1 0	1 4 0 2 6	0.0 0.0 0.0 0.0 0.0	0.0 0.0 14.3 7.1 0.0	5.3 21.1 0.0 10.5 31.6	(Canning-Clode et al., 2013; Ramalhosa et al., 2019; Souto et al., 2018) (Souto et al., 2014) (Hincks, 1880; Micael et al., 2019; Norman, 1909)
Bugula neritina Celleporaria inaudita Chorizopora sp. Conopeum sp. Copidozoum tenuirostre Corbulella sp. Crisia sp.	Tilbrook, Hayward & Gordon, 2001 Souto, Reverter-Gil & Ostrovsky, 2014 (Linnaeus, 1767) Hincks, 1880)	NIS U U C U U	0 0 0 0 0 0	0 2 1 0 1	1 4 0 2 6 0	0.0 0.0 0.0 0.0 0.0 0.0	0.0 0.0 14.3 7.1 0.0 7.1	5.3 21.1 0.0 10.5 31.6 0.0	(Canning-Clode et al., 2013; Ramalhosa et al., 2019; Souto et al., 2018) (Souto et al., 2014) (Hincks, 1880; Micael et al., 2019; Norman, 1909)
Bugula neritina Celleporaria inaudita Chorizopora sp. Conopeum sp. Copidozoum tenuirostre Corbulella sp. Crisia sp. Escharella sp.	Tilbrook, Hayward & Gordon, 2001 Souto, Reverter-Gil & Ostrovsky, 2014 (Linnaeus, 1767) Hincks, 1880) (Waters, 1898) (Moll, 1803) (Abildgaard, 1806)	NIS U U C U U U	0 0 0 0 0 0 0	0 2 1 0 1	1 4 0 2 6 0 0	0.0 0.0 0.0 0.0 0.0 0.0 0.0	0.0 0.0 14.3 7.1 0.0 7.1 7.1	5.3 21.1 0.0 10.5 31.6 0.0 0.0	(Canning-Clode et al., 2013; Ramalhosa et al., 2019; Souto et al., 2018) (Souto et al., 2014) (Hincks, 1880; Micael et al., 2019; Norman, 1909) (Norman, 1909)
Bugula neritina Celleporaria inaudita Chorizopora sp. Conopeum sp. Copidozoum tenuirostre Corbulella sp. Crisia sp. Escharella sp. Escharina vulgaris	Tilbrook, Hayward & Gordon, 2001 Souto, Reverter-Gil & Ostrovsky, 2014 (Linnaeus, 1767) Hincks, 1880) (Waters, 1898) (Moll, 1803)	NIS U C U U C	0 0 0 0 0 0 0 0	0 2 1 0 1 1 0	1 4 0 2 6 0 0 2	0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0	0.0 0.0 14.3 7.1 0.0 7.1 7.1 0.0	 5.3 21.1 0.0 10.5 31.6 0.0 0.0 10.5 	(Canning-Clode et al., 2013; Ramalhosa et al., 2019; Souto et al., 2018) (Souto et al., 2014) (Hincks, 1880; Micael et al., 2019; Norman, 1909) (Norman, 1909) (Busk, 1860, 1858; Norman, 1909; Waters, 1899)
Bugula neritina Celleporaria inaudita Chorizopora sp. Conopeum sp. Copidozoum tenuirostre Corbulella sp. Crisia sp. Escharella sp. Escharina vulgaris Escharoides sp.	Tilbrook, Hayward & Gordon, 2001 Souto, Reverter-Gil & Ostrovsky, 2014 (Linnaeus, 1767) Hincks, 1880) (Waters, 1898) (Moll, 1803) (Abildgaard, 1806) Souto, Kaufmann & Canning-Clode,	NIS U C U U U C U		0 2 1 0 1 1 0 0	1 4 0 2 6 0 0 2 1	0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0	0.0 0.0 14.3 7.1 0.0 7.1 7.1 0.0 0.0	 5.3 21.1 0.0 10.5 31.6 0.0 0.0 10.5 5.3 	(Canning-Clode et al., 2013; Ramalhosa et al., 2019; Souto et al., 2018) (Souto et al., 2014) (Hincks, 1880; Micael et al., 2019; Norman, 1909) (Norman, 1909) (Busk, 1860, 1858; Norman, 1909; Waters, 1899) (Norman, 1909; Waters, 1899)
Bugula neritina Celleporaria inaudita Chorizopora sp. Conopeum sp. Copidozoum tenuirostre Corbulella sp. Crisia sp. Escharella sp. Escharina vulgaris Escharoides sp. Favosipora purporea	Tilbrook, Hayward & Gordon, 2001 Souto, Reverter-Gil & Ostrovsky, 2014 (Linnaeus, 1767) Hincks, 1880) (Waters, 1898) (Moll, 1803) (Abildgaard, 1806) Souto, Kaufmann & Canning-Clode,	NIS U U U U U U N		0 2 1 0 1 1 0 0 1	1 4 0 2 6 0 0 2 1 2	0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0	0.0 0.0 14.3 7.1 0.0 7.1 7.1 0.0 0.0 7.1	 5.3 21.1 0.0 10.5 31.6 0.0 0.0 10.5 5.3 10.5 	(Canning-Clode et al., 2013; Ramalhosa et al., 2019; Souto et al., 2018) (Souto et al., 2014) (Hincks, 1880; Micael et al., 2019; Norman, 1909) (Norman, 1909) (Busk, 1860, 1858; Norman, 1909; Waters, 1899) (Norman, 1909; Waters, 1899)

<i>Membranipora</i> sp.	(Linnaeus, 1767)	U	1	0	0	2.6	0.0	0.0	(Norman, 1909)
Mollia sp.	(Waters, 1879)	U	0	1	12	0.0	7.1	63.2	
Parasmittina alba	Ramalho, Muricy & Taylor, 2011	NIS	0	0	3	0.0	0.0	15.8	(Ferrario et al., 2020; Ramalhosa et al., 2019; Souto et al., 2018)
Puelina sp.		U	0	1	10	0.0	7.1	52.6	
Reptadeonella violacea	(Johnston, 1847)	С	0	0	4	0.0	0.0	21.1	(Bianchi et al., 1998; Norman, 1909; Souto et al., 2015)
Schizomavella sp.		U	0	1	3	0.0	7.1	15.8	
Schizoporella errata	(Waters, 1878)	NIS	0	1	1	0.0	7.1	5.3	(Ramalhosa et al., 2019)
Schizoporella sp.		U	0	0	1	0.0	0.0	5.3	
Schizoporella unicornis	(Johnston in Wood, 1844)	NIS	0	0	9	0.0	0.0	47.4	(Busk, 1860; Souto et al., 2015; Waters, 1899)
Scruparia sp.	(Orbigny,1841)	U	0	1	0	0.0	7.1	0.0	
Scrupocaberea maderensis	(Busk, 1860)	Ν	0	0	1	0.0	0.0	5.3	(Busk, 1861, 1860; Norman, 1909; Souto et al., 2015)
Scrupocaberea maderensis	(Busk, 1860)	С	0	0	4	0.0	0.0	21.1	(Busk, 1861, 1860; Norman, 1909; Souto et al., 2015)
Unk Cyclostomatida		U	1	0	9	2.6	0.0	47.4	
Chordata (8)									
Botrylloides niger	Herdman, 1886	NIS	0	0	1	0.0	0.0	5.3	(Ramalhosa et al., 2021) (Canning-Clode et al., 2013; Ramalhosa et al., 2021, 2019; Wirtz,
Clavelina lepadiformis	(Müller, 1776)	NIS	0	0	1	0.0	0.0	5.3	1998)
Diplosoma listerianum	(Milne Edwards, 1841)	С	0	0	1	0.0	0.0	5.3	(Ramalhosa et al., 2021, 2019)
Distaplia corolla	Monniot F., 1974	NIS	0	0	3	0.0	0.0	15.8	(Canning-Clode et al., 2013; Ramalhosa et al., 2021, 2019)
Microcosmus sp.		U	1	0	1	2.6	0.0	5.3	
Seriola fasciata	(Plach 1702)	N	1	0	0	2.6	0.0	0.0	(Andaloro et al., 2005; Canning-Clode and Carlton, 2017; Wirtz et al., 2008)
Seriola jusciata	(Bloch, 1793) Valenciennes, 1833	C	1	0	0	2.6	0.0	0.0	(Wirtz et al., 2008)
Trididemnum cereum	(Giard, 1872)	c	0	0	1	0.0	0.0	5.3	(Canning-Clode et al., 2013; Ramalhosa et al., 2021, 2019)
Cnidaria (8)	(Glaid, 1872)	C	0	0	1	0.0	0.0	5.5	(Canning-Cloue et al., 2015, Kannaniosa et al., 2021, 2019)
Caryophyllia sp.		U	1	0	0	2.6	0.0	0.0	
Ectopleura larynx	(Ellis & Solander, 1786)	N	1	0	0	2.6	0.0	0.0	(Gestoso et al., 2017; Wirtz, 2007)
Exaiptasia diaphana	(Rapp, 1829)	NIS	1	0	0	2.6	0.0	0.0	(Canning-Clode et al., 2013; Ramalhosa et al., 2019; Wirtz, 2014)
Kirchenpaueria halecioides	(Alder, 1859)	C	2	2	3	5.3	14.3	15.8	(Ramalhosa et al., 2019; Wirtz, 2007)
Obelia dichotoma	(Linnaeus, 1758)	c	4	1	0	10.5	7.1	0.0	(Ramalhosa et al., 2019; Wirtz, 2007) (Ramalhosa et al., 2019; Wirtz, 2007)
Pennaria disticha	Goldfuss, 1820	c	4	0	1	0.0	0.0	5.3	(Wirtz, 2007)
Sertularella sp.	(Lamouroux, 1821)	U	0	1	4	0.0	7.1	21.1	(Vervoort, 2006)
sertuiurenu sp.	(Lamouloux, 1021)	U	U	т	4	0.0	/.1	21.1	

Unknown sp.		U	1	2	2	2.6	14.3	10.5	
Echinodermata (2)									
Arbacia lixula	(Linnaeus, 1758)	Ν	0	0	1	0.0	0.0	5.3	(Alves et al., 2001; Canning-Clode and Carlton, 2017; Gestoso et al., 2018)
<i>Ophiura</i> sp.		U	0	0	2	0.0	0.0	10.5	
Entoprocta (1)									
Barentsia discreta	(Busk, 1886)	С	0	0	1	0.0	0.0	5.3	(Ramalhosa et al., 2019)
Foraminifera (1)									
Miniacina cf. miniacea	(Pallas, 1766)	U	0	0	2	0.0	0.0	10.5	(George, 2014)
Mollusca (17)									
Bittium incile	R. B. Watson, 1897	Ν	0	0	2	0.0	0.0	10.5	(Segers et al., 2009a, 2009b)
Bittium latreillii	(Payraudeau, 1826)	Ν	1	0	3	2.6	0.0	15.8	(Segers et al., 2009a, 2009b)
Bittium sp.		U	0	0	1	0.0	0.0	5.3	
Calliostoma sp.		U	0	0	3	0.0	0.0	15.8	
Crisilla iunoniae	(Palazzi, 1988)	Ν	0	0	1	0.0	0.0	5.3	(Segers et al., 2009a, 2009b)
Doto fluctifraga	Ortea & Perez, 1982	С	0	1	0	0.0	7.1	0.0	
Fiona pinnata	(Eschsholtz, 1831)	С	6	0	0	15.8	0.0	0.0	(Segers et al., 2009a, 2009b)
Magallana gigas	(Thunberg, 1793)	С	1	0	1	2.6	0.0	5.3	This study
Mytilus galloprovincialis	Lamarck, 1819	NIS	1	1	0	2.6	7.1	0.0	(Segers et al., 2009a, 2009b)
Neopycnodonte cochlear	(Poli, 1795)	Ν	1	0	0	2.6	0.0	0.0	(Segers et al., 2009a, 2009b)
Odostomia sp.		U	0	0	1	0.0	0.0	5.3	
Palliolum incomparabile	(Risso, 1826) van Aartsen, Menkhorst &	Ν	0	0	1	0.0	0.0	5.3	(Segers et al., 2009a, 2009b)
Parvicardium vroomi	Gittenberger, 1984	Ν	0	0	1	0.0	0.0	5.3	(Segers et al., 2009a, 2009b)
Pinctada imbricata	Röding, 1798	NIS	1	0	1	2.6	0.0	5.3	(Segers et al., 2009a, 2009b)
Pinna rudis	Linnaeus, 1758	С	2	0	0	5.3	0.0	0.0	(Segers et al., 2009a, 2009b)
Stramonita haemastoma	(Linnaeus, 1767	С	1	0	0	2.6	0.0	0.0	(Segers et al., 2009a, 2009b)
Vermetidae sp.		U	0	0	1	0.0	0.0	5.3	
Porifera (3)									
Mycale (Carmia) senegalensis	Lévi, 1952	NIS	0	0	4	0.0	0.0	21.1	(Canning-Clode et al., 2013; Gestoso et al., 2017; Ramalhosa et al., 2019)
Prosuberites longispinus	Topsent, 1893	NIS	0	0	1	0.0	0.0	5.3	(Ramalhosa et al., 2019)

APPENDIX D – Examples of Identification Marks

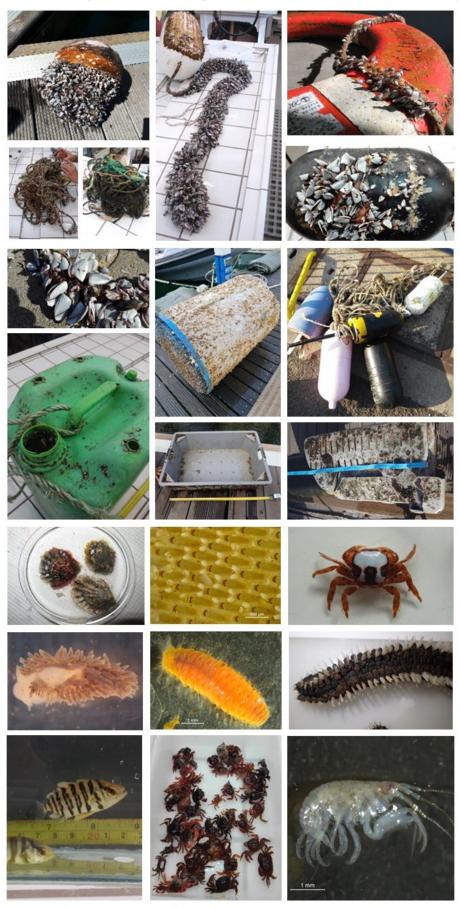


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Photos by Patrício Ramalhosa

APPENDIX E – Examples of Floating Marine Litter and associated species



Photos by Patrício Ramalhosa





APPENDIX F – Examples of Beached Marine Litter and associated species



Photos by Patrício Ramalhosa



APPENDIX G – Examples of Seafloor Marine Litter and associated species



Photos by Patrício Ramalhosa

