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Oceanographic and anthropogenic variables driving marine litter distribution in Mediterranean protected areas: Extensive field data supported by forecasting modelling

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model-guided methodological

approach to simultaneously monitor

floating macrolitter and microplastics • Largest data collection on marine litter distribution in the NW Mediterranean

Macrolitter and microplastic items accumulated in the same hotspot areas.
Secondary-origin plastic items were the

• Anthropogenic factors deeply influence litter inputs into the Pelagos Sanctuary.

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HIGHLIGHTS

New

Sea

G R A P H I C A L A B S T R A C T



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most abundant.

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ABSTRACT

Marine litter concentration in the Mediterranean Sea is strongly influenced both by anthropogenic pressures and hydrodynamic factors that locally characterise the basin. Within the Plastic Busters MPAs (Marine Protected Areas) Interreg Mediterranean Project, a comprehensive assessment of floating macro- and microlitter in the Pelagos Sanctuary and the Tuscan Archipelago National Park was performed. An innovative multilevel

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Marine litter sources Pelagos Sanctuary Oceanographic factors Spatial risk assessment experimental design has been planned ad-hoc according to a litter provisional distribution model, harmonising and implementing the current sampling methodologies. The simultaneous presence of floating macro- and microlitter items and the potential influences of environmental and anthropogenic factors affecting litter distribution have been evaluated to identify hotspot accumulation areas representing a major hazard for marine species. A total of 273 monitoring transects of floating macrolitter and 141 manta trawl samples were collected in the study areas to evaluate the abundance and composition of marine litter. High mean concentrations of floating macrolitter (399 items/km²) and microplastics (259,490 items/km²) have been found in the facing waters of the Gulf of La Spezia and Tuscan Archipelago National Park as well in the Genova canyon and Janua seamount area. Accordingly, strong litter inputs were identified to originate from the mainland and accumulate in coastal waters within 10–15 nautical miles. Harbours and riverine outfalls contribute significantly to plastic pollution representing the main sources of contamination as well as areas with warmer waters and weak oceanographic features that could facilitate its accumulation. The results achieved may indicate a potentially threatening trend of litter accumulation that may pose a serious risk to the Pelagos Sanctuary biodiversity and provide further indications for dealing with plastic pollution in protected areas, facilitating future management recommendations and mitigation actions in these fragile marines and coastal environments.

1. Introduction

Nowadays, the irreversibility and global ubiquity of marine litter pollution, and plastic in particular, make this material a potential threat to planetary boundaries (Nash et al., 2017; Rockström et al., 2009; Villarrubia-Gómez et al., 2018). Information on litter quantities and trends are widely reported across the ocean basins (Cózar et al., 2014; Suaria et al., 2016), although comprehensive approaches evaluating and linking their presence and distribution with the ecological impacts (including human health and socio-economic impacts) are still poorly adopted. The lack of standardized and harmonized measurements and reporting, as well as the uncertainty concerning definition and baseline values, sources, transport and accumulation, and effects of litter and plastic, still represent existing knowledge gaps to address. These challenges need consideration in the establishment of future harmonized monitoring programmes at the global level and related strategies, both in terms of scientific approaches and feasibility, providing a valuable basis for the development of effective protection and mitigation measures to be taken forward.

In the Mediterranean Sea, the management and monitoring of marine litter fall within the framework of two main regional drivers: the Regional Plan on Marine Litter Management in the Mediterranean (UN Environmental Programme/Mediterranean Action Plan), and the Marine Strategy Framework Directive (MSFD; 2008/56/EC, Descriptor 10) for the European marine waters. In this context, the Interreg Mediterranean Project Plastic Busters MPAs (PB MPAs) was conceived to consolidate the fight against marine litter in specific Mediterranean protected areas (MPAs) through a harmonized multidisciplinary approach. A new simultaneous sampling model-guided strategy was developed and tested to create a standardized protocol providing comparable and reliable data on the abundance of litter in pelagic and coastal surface waters as well as on beaches, highlighting potential hotspot and coldspot areas of litter accumulation in one of the heavily polluted basins at the global scale, the Mediterranean Sea. Concurrently, the spatial distribution of marine mammals and others species (seabirds, sea turtles, rays and fishes) were assessed as well as several endangered and bioindicator species were collected to figure out the threat posed to the organisms and the potential related physical impacts and biological effects.

This study focuses on the Pelagos Sanctuary, the most extended protected area of the Mediterranean Sea and the Tuscan Archipelago National Park, located in its eastern part. The Pelagos Sanctuary for Mediterranean Marine Mammals, hereafter Pelagos Sanctuary, is a Special Protection Area of Mediterranean Importance (SPAMI) established in 1999, to protect marine mammals and their habitat and to assess the actual and potential threats to cetacean populations (e.g. intense shipping traffic, fishing, whale-watching activities, chemical pollution, coastal development, military exercises, seismic prospecting, and global climate change) (Coomber et al., 2016; Grossi et al., 2021;

Mackelworth, 2016; Notarbartolo di Sciara et al., 2008; Notarbartolo di Sciara and Birkun Jr., 2010; Panigada et al., 2017). Recently, it has been recognized as an area particularly affected by high concentrations of microplastics (MPs) and plastic additives, which may constitute an additional threat to the endangered species inhabiting this basin (baleen whales, sea turtles, filter-feeding sharks) (Baini et al., 2017; Fossi et al., 2012, 2014, 2016, 2017, 2018a; Germanov et al., 2018; Panti et al., 2015) and to the overall biodiversity of the Mediterranean Sea (Compa et al., 2019; Deudero and Alomar, 2015; Galgani et al., 2015; Romeo et al., 2015). The Tuscan Archipelago National Park is the largest marine park in the Mediterranean as well as being classified as a biosphere reserve (Angeletti et al., 2010). It consists of 7 main islands, Gorgona, Capraia, Elba, Pianosa, Montecristo, Giglio and Giannutri managed according to different levels of protection: protection zone 1 or integral reserve referring to adjacent strips of water up to 1 km offshore and protection zone 2 (general protection) (Fratini et al., 2013; Renzi et al., 2010). The Tuscan Archipelago represents a crucial ecosystem for the central Tyrrhenian Sea due to its geographical position, geomorphological structure and high biological value given by the presence of several fish nursery areas (Renzi et al., 2010; Sbrana et al., 2016; Serena et al., 1998). Nevertheless, the intense maritime traffic, the pressure of tourist activities and the presence of several local pollution sources (e.g., maritime and commercial ports, Arno and Serchio riverine inputs, agricultural land and industrial activities) (Renzi et al., 2010) make this area highly anthropized and prone to the accumulation of floating plastic. This is confirmed by the temporary formation of a well-known retention area near the island of Capraia, where floating litter may accumulate (Fossi et al., 2017; Suaria et al., 2016). According to the different oceanographical features, habitats and extent of the selected study areas, the aims of this study were: i) to review the litter pollution status of MPAs, identifying the current knowledge gaps; ii) to define and test a new simultaneous multilevel experimental design within ad hoc sampling campaigns guided by litter distribution provisional model; iii) to provide a comprehensive assessment of the quantities and composition of floating marine litter in pelagic and coastal MPAs, highlighting hot and coldspot areas of litter accumulation; iv) to evaluate the potential influences of environmental and anthropogenic factors affecting the litter inputs and accumulation; v) to develop a map highlighting areas at higher risk of exposure for the marine organisms.

2. Materials and methods

2.1. Marine litter distribution provisional model

Sampling campaigns for floating litter were planned a priori based on a lagrangian model, developed by the LaMMA consortium, simulating the dispersion of floating passive litter particles (Fossi et al., 2017). The purpose of this model was to validate the predicted distribution and concentration of marine litter with on-field data to verify the strength and usefulness of litter prediction and as a result to identify potential hotspots (accumulation areas), coldspots (dispersion areas), and convergence litter areas in the Pelagos Sanctuary and Tuscan Archipelago National Park. Dedicated bulletins were edited to directly guide the monitoring efforts in the study area according to litter pollution estimates by the Lagrangian model adopted (Fossi et al., 2017) and habitats and feeding grounds of investigated species. To compare marine litter abundances with simulation results, the linear correlation coefficient (Pearson correlation test) between the model data and the observed data was calculated, taking into consideration the estimations done at the same positions and times.

2.2. Sampling strategy

The ad hoc sampling activities for simultaneous monitoring of floating macrolitter and microplastic are shown in Fig. 1A and B. In the Pelagos Sanctuary SPAMI, monitoring transects were performed starting from one nautical mile (nm) offshore and repeated every 10 nm in pelagic areas (Fig. 1A). Additional macrolitter transects were conducted before the simultaneous sampling described above. In the Tuscan Archipelago National Park, a star-shaped experimental design was adopted on the coastal waters off the 7 main islands to assess potential differences in marine litter distribution as a function of different levels of protection (monitoring zones inside and outside the protected areas) and distance from the coast (Fig. 1B). Simultaneous transects were started one nautical mile offshore and repeated at 3 nm, while the macrolitter object monitoring was carried out throughout the circumnavigation of each island (Fig. 1B). Simultaneously at the floating litter evaluation, in both areas several bioindicator species (e.g. invertebrates, fishes and cetaceans) of plastic ingestion were collected or sampled to assess the potential physical and chemical impacts of plastics and their additive compounds (i.e., phthalic acid esters) (data not shown).

2.3. Marine litter monitoring activities

The sea surface floating litter (macrolitter and microplastic) sampling campaign in the Pelagos Sanctuary was carried out both in pelagic and neritic areas focusing on the northern sector (including the coasts of Liguria and Tuscany, Italy) and the central-western sector (including the coasts of Italy and France) of the Ligurian Sea from May to June 2019 and the northeast and northwest coasts of the island of Corsica (Fig. 1) in September 2019. Sampling effort was carried out taking into account the litter distribution provisional model outputs, different depths and slopes of the study area, focusing on the submarine canyons of Genova, Imperia and Saint-Florent, considered special feeding areas for cetaceans and fin whales in particular (Moulins et al., 2007, 2008; Würtz, 2012), as well as near potential sources of marine litter pollution, such as the port of Livorno and Marina di Pisa (along the Tuscan coast) and La Spezia, Genova and Loano (along the Ligurian coast), as well as the river discharges of Arno, Serchio and Magra. The sampling campaign was conducted onboard the 54-ft sailing catamaran "Headwind" property of CIMA Research Foundation. A total of 1568 nautical miles were navigated, with 168 floating macrolitter monitoring transects and 84 manta trawls conducted to assess the floating microplastic.

The Tuscan Archipelago National Park sampling campaign was conducted in July 2019 aboard the oceanographic ISPRA research vessel ASTREA, focussing on the coastal waters off the 7 main islands of the archipelago. A total of 585 nautical miles were covered, with 105 transects monitoring the floating macrolitter and 57 manta trawls assessing the floating microplastic.

2.4. Sea surface floating macrolitter: monitoring and characterization

The distribution, abundance and composition of floating macrolitter (>2.5 cm) were assessed using the fixed-width strip transect method as recommended in the monitoring guidelines developed by the EU MSFD Technical Subgroup on Marine Litter (Hanke et al., 2013) and the MEDSEALITTER project (Arcangeli et al., 2020). This method allows counting the number of objects detected within a fixed-width strip, which should be representative of the visibility conditions during the survey and depends mainly on the speed of the vessel and the height of the observer above sea level. All observations were made with the naked eve from the bow of the ship (3 m above sea level) at a mean speed of 4 knots for 30 min. Due to the characteristics of our observation set-up, a relatively narrow strip of 7 m was monitored, following the recommendation of Galgani et al. (2013). Each item was characterized according to the main list of litter categories (Galgani et al., 2013), which revised the original OSPAR and UNEP categories (Cheshire et al., 2009) and indicated the type (Artificial Polymer Materials, Rubber, Cloth/ Textile, Paper/Cardboard; Processed/Worked Wood and Metal), size classes (B. 2.5-5 cm, C. 5-10 cm, D.10-20 cm, E. 20-30 cm, F. 30-50 cm, G. > 50 cm) and colours (W. White; T. Transparent; B. Black; C. Cyan/ Blue; R. Red; G. Green; Y. Yellow; O. Other) of the floating objects. Finally, counts of scattered objects were converted to density values (Di) by dividing the total number of objects sighted by the effective area sampled in each transect:



Fig. 1. Monitoring activities carried out during the Pelagos Sanctuary (A) and Tuscan Archipelago National Park (B). Macrolitter (green) and microplastic transects (orange) were performed simultaneously starting one nautical mile from the coast and repeated every 3 and 10 nautical miles in the Pelagos Sanctuary and Tuscan Archipelago National Park, respectively.

$$Di = \frac{n}{L^*W}$$

Where *n* is the number of items seen on the transect, *L* is the length of the transect, *and W* (7 m) is the fixed width of the strip observed and expressed as items/km².

2.5. Sea surface floating microplastic: sampling and characterization

Floating microplastic samples were collected using a manta trawl (330 μ m mesh size, 16 \times 60 cm mouth opening) (simultaneously to the floating macrolitter survey) towed at 2–3 knots on the water surface for 30 min, held to the side of the boat to avoid the turbulence caused by the wake of the vessel. At the end of each transect, the net was thoroughly rinsed from the outside to ensure that both neuston and microplastics were washed into the end of the net. Samples were filtered through a 300 μ m metal sieve and stored in a 70 % ethanol solution for synthetic particle analysis. To avoid contamination throughout the sampling activities, all the materials used for sample collection, including the nets, were carefully cleaned and rinsed before each tow.

In the laboratory, the floating microplastic samples were filtered through a sieve (mesh size: 300 µm) and observed under an NBS stereo zoom microscope (Mod. NBS-STMDLX -T) equipped with an LED light and a micro metered eyepiece. The microplastics were manually isolated in a glass Petri dish and allowed to dry overnight at room temperature. Each Petri dish was then photographed and analysed for particle size measurement (expressed in mm) using ImageJ software (Fiji Distribution). Natural buoyant materials such as plant leaves, wood and bird feather were manually removed from the samples and rinsed with microfiltered waters (0.45 µm) to collect all the plastic particles sticking to them. Samples visually characterized both by heavy plastic particles and organic matter content were re-suspended with a NaCl solution (1.2 g/cm³) and let settled down overnight. Moreover, the dubious chemical origin of smaller natural particles such as chitin residues, was promptly tested through the hot needle technique (Bellas et al., 2016). The isolated particles were characterized according to different size classes into small microplastic (SMPs) (0.3-1 mm), large microplastic (LMPs) (1-5 mm), mesoparticles (5-25 mm), and macroparticles (>25 mm), shape (pellet, fragment, film, filament, microbead, and foam), and colour (black, blue, white/transparent, white/opaque, red, green, and others) and weighed using an OHAUS Explorer precision balance (± 0.1 mg) (Fossi et al., 2017). Glassware was used in the laboratory procedures, and special care was taken to prevent airborne contamination by performing sample analysis in a clean airflow cabinet and using two glass Petri dishes placed on either side of the stereomicroscope as blank controls. Despite the use of contamination control procedures, fibres and paint were not included in the total count of the particles due to the risk of external contamination during sampling.

The data obtained, expressed as concentration items/km² and mg/ m^2 were normalised, if necessary, by applying the correction factor proposed by Kukulka et al. (2012). This factor, widely accepted in the scientific literature (Baini et al., 2018; Faure et al., 2015; Fossi et al., 2017; Kooi et al., 2016) takes into account the unfavourable meteorological and maritime conditions (wave >0.50 m and wind speed >4 m/s) that may affect the accumulation of floating microplastic in surface waters due to the wind mixing effect, leading to an underestimation of their concentrations, and proposes an appropriate value to correct the final concentrations of the samples. Finally, the polymeric composition of 10 % of the isolated microplastics was evaluated and selected proportionally according to the relative abundance in the different sizes, shapes and colour classes for each sample. Using Fourier infrared spectroscopy (FTIR), each particle was scanned 16 times using an Agilent Cary 630 spectrophotometer. To identify the polymers, the spectrum obtained was processed using Agilent Micro Lab FTIR software and compared to a database of reference spectra. Only results that showed >80 % overlap were accepted (Baini et al., 2018).

2.6. Environmental and anthropogenic influences on marine litter distribution

The whole dataset related to floating macrolitter and microplastic was examined considering the habitat types (bathyal, canyon, seamount, slope and continental shelf) and the main environmental (SST: sea surface temperature; SSH: sea surface height; MLD: depth of mixed layer and current velocity) and anthropogenic factors (vessel traffic, distance from ports, distance from the coast and distance from estuaries) that may influence its distribution. Habitat types were identified using GEBCO bathymetric data (GEBCO, 2022); canyons were identified following Tepsich et al., 2014. Oceanographic data were taken daily from the Copernicus Marine Environmental Service, associating each floating macrolitter and microplastic sample with the corresponding daily value of the environmental variables considered. Vessel traffic and port data were downloaded from the European Marine Observation and Data Network (EMODnet, n.d., www.emodnet.eu). The vessel traffic data have a monthly resolution and include data from different vessel types (tankers, cargo vessels, fishing vessels, passenger vessels, sailing vessels and recreational vessels). Vessel densities are reported in hours/km²/month; each floating waste concentration sample was linked to the corresponding monthly traffic data. Discharge location data were obtained in QGIS using river data downloaded from the ISPRA website (http://www.sinanet.isprambiente.it/it/sia-ispra/ download-mais/reticolo-idrografico/view, n.d) for Italy and the French government (https://www.data.gouv.fr/fr/datasets/cours-deau-metropole-2017-bd-carthage, n.d) for France. In Italy, rivers were classified into two different groups (torrents and streams) according to their flow rate and classified as minor and major discharges, respectively. In France, rivers were divided into two classes according to their length: rivers longer than 25 km belong to class 1 (major discharges), and those longer than 10 km belong to class 2 (minor discharges).

2.7. GIS (geographical information systems) and statistical analysis

Floating marine litter concentration data were imported and processed using the Quantum GIS platform (version 3.10.1 A Coruña), and Rstudio (version 1.1.4.1106) to perform spatial and statistical analysis respectively. Descriptive statistics and normality tests (Shapiro-Wilk normality test and Anderson-Darling test) were performed to examine the entire floating litter datasets to determine whether parametric or non-parametric statistical analyses were appropriate.

Mann-Whitney-Wilcoxon for pairwise comparisons and Kolmogorov-Smirnov tests were used to compare differences in floating litter mean concentrations (items/km²) and characteristics of the items (size, shape and macrolitter categories) between the Pelagos Sanctuary and the Tuscan Archipelago National Park areas and the zoning protection among islands. The Kruskal-Wallis test for multiple comparisons and post hoc test analysis was conducted to compare differences in the distribution of floating litter among different habitats.

The analysis of environmental and anthropogenic factors that can influence the distribution of floating litter was performed in two steps, following the method of Kanhai et al. (2017). A Spearman's rank correlation test was performed between the factors considered and the scattering litter concentration. Then, generalised additive models (GAMs) were used to evaluate the influence of each variable on the distribution of floating litter. The response variable was always litter abundance (macrolitter or MPs), while the initial explanatory variables were potential pollution sources. The variables were considered separately so that a GAM was created for each variable for each type of floating litter (macrolitter or MPs). For the variables characterized by the presence of outliers, two models were created: one in which all values were included, and the other in which the outliers were excluded. Outliers have been identified by examining box-plots built separately for each variables, thus identifying points located outside the 1.5 times the interquartile range above the upper quartile and bellow the lower

quartile. Among the two GAMs built, the final model was then selected applying REML (Rpakage mgcv). This procedure was chosen to evaluate the influence of extreme situations that might not be representative of the general situation in the study area. To better understand the relationships represented by the GAM plots, a null line was used to define the positive effect of the predictors on litter accumulation, in a process called GAMvelope (Torres et al., 2008; Correia et al., 2015). The GAMvelope allowed the highlighting of areas favourable to litter accumulation in the Pelagos Conservation Area. A significance level (p < 0.05) was considered for all analyses.

2.8. Litter distribution risk map

Average oceanographic conditions in terms of SST, SSH and current velocity were determined using monthly maps corresponding to the period of the sampling campaigns (June–September 2019). A 5 km grid was overlaid to the Pelagos Sanctuary area, assigning a value of 1 to each cell characterized by environmental and anthropogenic variables that positively affect the litter distribution, as resulted from the GAMvelope model. A comprehensive hazard map was then generated, based on the distribution of floating macrolitter, with hazard indices ranging from 1 to 8 (considering the maximum number of variables influencing the litter distribution and accumulation) and indicating the areas at higher risk of exposure for the marine organisms.

3. Results and discussions

3.1. Marine litter distribution in marine protected areas: State of the art

Litter abundance and distribution in the Mediterranean Sea were reviewed with a particular focus on the sampling efforts carried out in the MPAs, their pollution status and the potential gaps to be covered to fully addressed the impact and effect of marine litter.

Although there are several studies on floating litter in the Mediterranean Sea covering different areas (Fig. 2), slightly >30 % have focused on marine protected areas (Supplementary material Tables S1 and S2). The Langrangian modelling analysis of plastic fluxes on six selected coasts of marine protected areas in the Mediterranean proposed by Liubartseva et al. (2019) represents one of the first attempts to predict marine litter distribution and potential impacts in MPAs. It showed that the input of litter was relatively low (0.4–3.6 kg (km/day) compared to the average flux of 6.2 \pm 0.8 kg (km/day) calculated for the Mediterranean Sea in 2013-2017 (Liubartseva et al., 2019), assessing the synergistic role of anthropogenic factors generating plastic and hydrodynamic transport influencing its distribution within MPAs. A different approach was proposed by Fossi et al. (2017) in the Pelagos Sanctuary SPAMI, where simulated and in situ MP concentrations were evaluated to verify the accuracy and strength of the predictive plastic distribution model and to highlight the potential risk associated with ingestion by fin whales in this important ecological MPA. This area is one of the most investigated in the Mediterranean Sea (Supplementary material Tables S1 and S2), 70 % of the studies conducted in MPAs are carried out here, with an average abundance of floating objects and MPs of 0.73 ± 82.3 items/km² and $85,122 \pm 35,726$ items/km² (0.30 ± 0.23 items/m³), respectively. This MPA also seems to be affected by the temporary formation of the marine litter convergence area between the islands of Corsica and Capraia, where a high concentration of plastic has been reported (Baini et al., 2018; Fossi et al., 2017; Suaria et al., 2016). In the western part of the Mediterranean, the presence and distribution of marine litter in other MPAs have been studied in the Menorca Channel (Ruiz-Orejón et al., 2019), the Cabrera National Park (Fagiano et al., 2022), Calanque National Park (Schmidt et al., 2021), Ischia and Ventotene Marine Protected Area (de Lucia et al., 2018) and Torre Flavia wetland (Battisti et al., 2019; Cesarini et al., 2021). Exceptionally high concentrations of floating macrolitter were detected in surface waters of the MPAs of Gozo and Malta (Ionian Sea and Central Mediterranean sub-



Fig. 2. Number of studies evaluating floating marine litter (macrolitter and microplastic) carried out in the Mediterranean Sea from 1980 to 2022, according to the Marine Strategy Framework Directive (MSFD) subregions.

regions). This study highlights the potential influence of seasonal variation and distance from the coast on the distribution and accumulation of litter, showing the highest levels during the winter season (2392 \pm 7477 items/km²) and in coastal areas (6371 \pm 11,968 items/km²) (Curmi and Axiak, 2021). Despite the data reported by previous studies carried out in the MPAs, no information is still available regarding the potential influence of oceanographic and anthropogenic variables affecting the litter accumulation and stranding as well as the potential sources of pollution insisting in this basin. In this context, the present study reports relevant data that shed light on the main inputs of litter contamination and factors driving litter dispersion, proposing for the first time a model-guided sampling approach.

3.2. Floating litter: abundances and composition

The experimental designs performed ad hoc in the selected areas showed valuable efficacy in the collection of data providing reliable information on the abundance, distribution and composition of macrolitter and microplastic. An overall correlation index (Pearson correlation test; R = 0.83, p < 0.05), between on-field marine litter abundances and simulation results computed by the Lagrangian model, was highlighted confirming the effectiveness of the adopted sampling strategy and validating the modelling indirect approach to correctly predict the concentration of floating litter.

3.2.1. Floating macrolitter

A total of 273 transects were conducted to monitor the presence of floating macrolitter throughout the study area. A total of 2169 items ranging from 0 to 3974 items/km² were sighted, with an average concentration of 399.01 \pm 485.84 items/km² (Fig. 3A). This value is onetwo orders of magnitude higher than the threshold level proposed by UNEP/MAP (2020) (5 objects/km²) and the average concentration calculated considering the published data on the assessment of litter in the western part of the Mediterranean (Supplementary material Table S1) (29.7 \pm 46.8 items/km²). As far as we know, this value represents the highest concentration of floating macrolitter recorded so far in the study area and could indicate a potential worsening of the macrolitter status in an important ecological area as the Pelagos Sanctuary (Arcangeli et al., 2018; Campanale et al., 2019; Di-Méglio and Campana, 2017; Fossi et al., 2017; Suaria and Aliani, 2014). As for the other Mediterranean subregions, few studies reported similar litter concentrations in the surface waters of MPAs located respectively in the Adriatic Sea (Palatinus et al., 2019) and near the islands of Malta and Gozo (Curmi and Axiak, 2021). Nevertheless, most published papers have been conducted with oceanographic vessels sailing at >6 knots and from an observing height of 6 to 25 m. Variability in observation conditions can affect the detection of small macrolitter objects (Class B. 2.5–5 cm), as previously acknowledged (Galgani et al., 2013; Zeri et al., 2018). Only recently studies have started to report the minimum size class detected (Compa et al., 2019; Di-Méglio and Campana, 2017; Fossi et al., 2017; Vlachogianni et al., 2018, 2020; Zeri et al., 2018), and relative information on the size characterization of sighted items (Zeri et al., 2018). Against this background, the application of harmonized monitoring protocols at the Mediterranean level, as proposed and implemented in the PB MPAs project, will improve the accuracy, and comparability of reported marine litter densities and effective identification of hot spot areas.

Litter and in particular artificial polymer materials items (99 % of the total) were observed in 90 % (245/273 transects) of the transects conducted. These results are consistent with previous studies published throughout the Mediterranean Sea (Campanale et al., 2019; Compa et al., 2019; Fossi et al., 2017; Tata et al., 2020). The majority of the sighted objects (80 %) had a size of <20 cm and a light-coloured characterization (>80 %), with size class B (2.5–5 cm; 58 %) being the most common. The account of this dimensional range as the most frequently sighted is consistent with other studies conducted aboard small vessels

at low speed, which allowed a homogeneous detection of all floating objects encountered in the sampled striped waters (Palatinus et al., 2019; Vlachogianni et al., 2018; Zeri et al., 2018).

Analysis of the most common objects revealed that >70 % of all objects floating on the sea surface were represented by 10 categories of litter (Supplementary material Table S3). Objects of secondary origin belonging to the categories G67 (Sheets and industrial packaging) and G79 (Plastic pieces 2.5 cm><50 cm) were most frequently sighted. Their presence could be an indication of the degradation processes and fragmentation that affect the litter objects once dispersed in the marine environment, allowing the formation of smaller items.

A statistical difference in the distribution of samples between the two monitored areas (W = 5413.5, p-value = 9.70e-10) was found, confirming a lower concentration of floating macrolitter in the Pelagos Sanctuary (280.36 \pm 423.88 items/km²) than in the surface waters of the Tuscan Archipelago National Park (617.76 \pm 599.15 items/km²). A statistical difference in mean concentration was also observed between the two areas considering different size classes (B. 2.5–5 cm, C. 5–10 cm, D. 10-20 cm, E. 20-30 cm, F. 30-50 cm, G. > 50 cm). Only for class E (20-30 cm), no difference in average concentration was observed, while the concentration for all other classes was higher in the Tuscan Archipelago National Park than in the Pelagos Sanctuary (Supplementary material Fig. 1). The highest abundances in surface waters of the islands of the Tuscany region, both in terms of the number of items and size classes, may be due to more recent inputs of pollution from land, as this area was particularly affected by tourist and recreational activities during the summer period of the sampling campaigns. Moreover, the stability of hydrodynamic features that characterise the Tuscan Archipelago during the summer season could favour the floating of larger objects in coastal waters once they are dispersed in the marine environment, delaying their potential accumulation in pelagic areas.

Considering the different types of litter, the categories with the highest average concentration in the Pelagos Sanctuary were G67 (Sheets and industrial packaging) and G79 (Plastic pieces 2.5 cm><50 cm), for which an average of >100 items/km² was recorded (Supplementary material Table S3). These categories were resulted the most abundant also in the Tuscan Archipelago National Park, reaching >300 items/km² and 150 items/km², respectively. The categories "G58: Fish boxes", "G94: Tablecloth", "G145: Other textiles" and "G149: Paper packaging" were only sighted in the Pelagos Sanctuary; while "G3: Buoys", "G74: foam packaging", "G135: Clothing", "G142: Rope, string, and nets" and "G160: Pallets" were only present in the Tuscan Archipelago.

Among the categories sampled in both study areas, "G6: Bottles", "G18: Crates and containers/baskets", "G45: Mussel nets/Oyster nets", "G48: Synthetic rope", "G67: Sheets, industrial packaging, plastic sheeting", "G79: Plastic pieces 2.5 cm> <50 cm" and "G124: Other plastic/polystyrene items (identifiable)" were found to have a higher statistically significant concentration in the Tuscan Archipelago National Park than in the Pelagos Sanctuary area (Supplementary material Table S3).

Considering the differences between the islands of the Tuscan Archipelago National Park, the highest concentration of macrolitter was found in the southern sector of the archipelago near the islands of Giglio and Giannutri (792.90 \pm 610.13 items/km²) and the northern sector between the islands of Gorgona and Capraia (726.42 \pm 735.20 items/km²) (Fig. 4). These patterns of accumulation may be influenced by the inputs of litter originating directly from the coast due to the short distance of these islands and the proximity of the Tevere river identified as a plastic pollution source by de Lucia et al. (2018) in the southern sector. The influence of rivers on plastic distribution in this area was pointed out also by Galgani et al. (2019), evidencing how during the summer period, the northern part of the Tyrrhenian Sea was particularly affected by plastics riverine inputs originating from the Ombrone and Tevere rivers and spatially distributed by the superficial currents insisting on this area. Oceanographical features, and in particular the currents



Fig. 3. Floating macrolitter (A) and microplastics (B) spatial distribution in the whole study area considered. The concentrations of litter objects sighted were expressed in items/km², and the floating macrolitter threshold proposed by UNEP/MAP (2020) reported.



Fig. 4. Total mean concentrations of floating macrolitter found in the facing waters of Tuscan Archipelago National Park from the northernmost to southernmost island (A) and inside (green) and outside (orange) the protection zones (B). The areas considered as "Inside and Outside Protection Zone" groups are also displayed (C). * indicates statistically significant difference (p < 0.05).

prevailing in the northern part of the Tuscan Archipelago National Park might be considered the main factors determining plastic distribution in this area. Other studies published have highlighted the transient formation of a convergence area between the islands of Capraia, Corsica and Gorgona, which influences the distribution and concentration of

litter (Fossi et al., 2017; Suaria et al., 2016).

Statistical differences among islands in terms of floating litter concentration (chi-squared = 14.401, df = 6, *p*-value = 2.5e-2) were found. In particular, Giannutri and Montecristo were statistically different from the other areas and were the islands with the highest (1040.35 \pm 648.34

items/km²) and lowest (264.93 \pm 210.92 items/km²) concentration values, respectively (p = 1e-3, p adjusted = 2e-2) (Fig. 4A). This difference could be explained both by the distance from the Tuscan coast and by the potential pollution sources that may affect these islands. Montecristo is the farthest island of the Tuscan Archipelago and it is located about 22 nm far from the Tuscan coast. Moreover, the waters of the island are fully protected up to a distance of 1 nm and tourist access is regulated and limited to 1000 visitors per year. For these reasons, the presence and accumulation of litter could be limited, as confirmed by our data. The second-highest concentration was found in the waters facing the island of Pianosa (748.32 \pm 522.32 items/km²) (Fig. 4A). The island is in the central sector of the Tuscan Archipelago and seems to be affected by intensive transport, accumulation and beaching ashore of litter items. So far, these data represent the first assessment of litter occurrence in this area, where surface currents seem to play a crucial role in marine litter accumulation.

As already highlighted for the whole study area, size class B was the most abundant in the different islands studied, especially in Giannutri Island with a concentration of 579.25 \pm 402.43 items/km² (p = 1e-3 - p adjusted = 2e-2) (Supplementary material Table S4). Litter with larger dimensions (>10 cm) was found statistically significant in the surface waters off the islands of Pianosa (classes D and E p = 2e-3 p adjusted = 4e-2) and again Giannutri (class D) (p adjusted = 2.3e-2) (Supplementary material Table S4). Considering the samples carried out inside or outside the protected areas in the Tuscan Archipelago National Park, the concentration of floating litter showed statistically significant lower values inside the marine protected area than outside (p = 6.2e-3) (Fig. 4B). Categories G67 (Sheets, industrial packaging, plastic sheeting), and G79 (Plastic pieces 2.5 cm > < 50 cm) were the most frequently found objects on the different islands. Their presence was assessed in higher concentrations in the samples collected outside the protected areas in the Tuscan Archipelago National Park. However, statistical differences were only found for category G67 (679.08 \pm $332.06 \text{ objects/km}^2$) (p-value = 7e-4).

3.2.2. Floating microplastic

A total of 141 manta trawl samples were simultaneously collected during the macrolitter monitoring to assess the concentration of smaller particles (<25 mm) at the sea surface. A total of 56,084 particles were isolated belonging 90 % (n. 50,985) and 10 % (n. 5099) to microplastics and mesoplastics, respectively. No rubber particles were found, so the following results refer to MPs only. An average concentration of 259,490 \pm 586,477 items/km² was found throughout the whole study area, ranging from 16,647 to 4,933,909 items/km² (Fig. 3B). This value was in agreement with the mean value of floating MPs abundances in the western Mediterranean subregion, which was calculated considering the studies available in the literature and set at 216,399 \pm 284,360 items/ km². Although no threshold values for MPs in the Mediterranean Sea have yet been proposed, the concentration found in this study and by Caldwell et al. (2019) (233,927 \pm 810,357 items/km²) in the Pelagos Sanctuary appear to be increasing compared to those reported during the sampling campaign carried out in 2018 (Caldwell et al., 2019) and by previous studies (Baini et al., 2018; Collignon et al., 2012, 2014; Fossi et al., 2017; Pedrotti et al., 2016; Tesán Onrubia et al., 2021). This threatening trend of particle accumulation may pose a threat to organisms living in this protected area throughout the marine trophic chain, as also highlighted by Fossi et al. (2017). The average concentration observed here (1.62 \pm 3.67 items/m³), expressed as particles per m³ to allow a proper comparison with other studies (Supplementary material Table S2), resulted higher than all those reported in the literature except for the values found by Fagiano et al. (2022) in the Cabrera National Park (3.52 \pm 8.81 items/m³) considered an area of high plastic waste density (Ruiz-Orejón et al., 2018).

MPs characterization analysis revealed that large MPs were the most abundant size class (76 %) ranging between 1 mm to 2.5 mm and accounting for 42 % in total. Fragments (86 %) and films (10 %) are the most represented shapes with 96 % of the isolated particles. These results are consistent with other studies conducted in the Mediterranean Sea (Baini et al., 2018; Compa et al., 2020; Suaria et al., 2016) and in other oceans (Cózar et al., 2014; Eriksen et al., 2013) and confirm that secondary microplastics are the most widespread in the marine environment. Colours can also influence the uptake of plastic particles. Particularly brightly coloured items, which were represented in this study at a concentration of >70 %, could increase the likelihood of ingestion as they resemble prey (Martí et al., 2020; Wright et al., 2013).

Polymer composition analysis showed that polyolefin thermoplastics, represented by polyethylene (PE) and polypropylene (PP) (95 % in total), were the most abundant. Their presence in the marine environment is widely recognized in all ocean basins (Baini et al., 2018; Enders et al., 2015; Pedrotti et al., 2016; Suaria et al., 2016), reflecting the increasing production and use of these plastic polymers. They are mainly used in packaging and disposable products and their production in Europe represents about 50 % of the total plastic demand (PlasticsEurope, 2020). Moreover, as PE and PP positively buoyant polymers $(0.90-0.99 \text{ g/cm}^3; 0.85-0.92 \text{ g/cm}^3)$ are sensitive to degradation in the marine environment and have a longer residence time at the sea surface, they tend to accumulate at the sea surface as confirmed by the plastictype here found, mainly fragments and films, made of these materials. The average weight density and concentration values of MPs were statistically lower in the Pelagos Sanctuary (0.068 \pm 0.162 mg/m² and 226,075 \pm 650,984 items/km²) than in the surface waters of the Tuscan Archipelago National Park (0.152 \pm 0.261 mg/m² and 355,281 \pm 616,782 items/km²) (weight density W = 1524, *p*-value = 1.8e-5; concentration W = 1524, p-value =3.2e-3).

This result confirms what was observed for the distribution and concentration of floating macrolitter objects and strengthens the hypothesis that the presence of larger objects (categories G67 and G79) may influence the formation of MPs as a result of degradation and fragmentation processes. According to that, a correlation between the spatial concentrations of floating macrolitter (273 monitoring transects) and microplastics (141 manta trawl samples) collected in the whole study area was investigated to reveal a statistically common distribution pattern. A significant strong direct correlation (p-value <2.2e-16, r = 0.63) (Fig. 5) was found confirming the effectiveness of the experimental plan performed and highlighting the importance of the simultaneous floating litter sampling to better address the presence and distribution of plastic pollution in the marine protected areas highlighting the presence of hotspot areas and finally providing also preliminary information on the potential impacts on marine organisms.

The shape analysis of the isolated particles revealed a significant concentration of fragments ($305,065 \pm 522,863$ items/km²) (p-value = 9.2e-03) and films ($37,479 \pm 101,232$ items/km²) (p-value = 4.1e-05) higher in the surface waters of the Tuscan Archipelago National Park than in the Pelagos Sanctuary (Fig. 6A).

According to the classification of size classes, the difference in mean concentration between the two areas was statistically significant when considering only larger particles, which resulted in a higher concentration in the Tuscan Archipelago National Park (287,744 \pm 497,983 items/km²) than in the Pelagos Sanctuary (163,084 \pm 466,917 items/ km²) (Fig. 6B). This accumulation pattern was confirmed by the study of Pedrotti et al. (2016), analysing the size distributions of plastic particles at different distances from land and showed an increase in plastic abundance from large to small items moving from coastal to pelagic areas. Moreover, this result is consistent also with the general size distribution found by Cózar et al. (2014) for ocean surface waters. According to Pedrotti et al. (2016), the highest presence of large MPs in the nearshore areas could be due to the combination of efficient removals of small fragments from the surface due to their potential stratification along the water column, sinking due to the biofouling processes and the interactions with marine organisms such as invertebrates species. In addition, the gradual fragmentation processes due to physical and chemical degradation of plastic particles moving towards the pelagic



Fig. 5. Correlation scatterplots among floating macrolitter and MPs concentration evaluated in the SPAMI Pelagos Sanctuary (cyan dots) and Tuscan Archipelago National Park (blue dots). Statistical significance for p-value <0.001.



Fig. 6. Floating MPs different distribution among the two study areas considered (Pelagos Sanctuary: blue boxplots; Tuscan Archipelago National Park: green boxplots) according to shape, size classes and total average concentration. The black line shows the reference value for mean MPs concentration in the Northwestern Mediterranean Sea, while the dashed line represents the reference value for the standard deviation of MPs concentration in the Northwestern Mediterranean Sea.* indicates difference statistically significative (p < 0.05).

areas may favourite the formation of smaller particles and their accumulation in offshore waters.

However, no differences among the islands of the Tuscan Archipelago National Park were found (Supplementary material Table S5).

The highest concentrations for both number and weight density of particles were found in the northern part of the Tuscan Archipelago facing the island of Gorgona (Supplementary material Table S5). This area was previously described as the most affected by the presence of floating macroparticles, indicating the formation of a temporary accumulation zone previously described in the literature (Fossi et al., 2017;

Suaria et al., 2016). High particle abundances were also detected around the islands of Elba and Pianosa in the central part of the archipelago, respectively, where MPs seem to accumulate. While the first area is under strong anthropogenic pressure, especially during the summer months, the distribution and accumulation of particles in the facing waters of Pianosa island again seem to be closely related to the surface currents that characterise the waters there. Differently from what was highlighted for macrolitter objects, the islands in the southern sector appear to be more vulnerable to recent inputs of plastic pollution from the coast. This is also confirmed by the greater extent of the sighted

objects, which may be displaced to more pelagic areas of the Tyrrhenian Sea where fragmentation processes occur, as indicated by the litter dispersion model (Northern Tyrrhenian Gyre, described in Fossi et al., 2017). No differences in MPs distribution were found between the different levels of protection regulating the monitored islands.

3.3. Marine litter distribution: influence of marine habitats

The potential distribution of floating macrolitter and MPs was assessed considering the different marine habitats according to the topographic features within the Pelagos Sanctuary and the Tuscan Archipelago National Park. The monitored areas were characterized by different habitat types in the bathyal, canyon, seamount, slope and continental shelf (Supplementary material Table S6).

A statistical difference between habitats (Kruskal-Wallis chi-squared = 39.79, df = 4, *p*-value = 4.8e-08) was detected only for floating macrolitter, underlining how the continental shelf (573 \pm 572 items/km²) and seamount (205 \pm 245 items/km²) areas were separated from all other habitats (Fig. 8B). No difference was found for the distribution of MPs (chi-square = 8.91, df = 4, p-value = 6.3e-2) in the study areas (Fig. 8A).

The highest concentration of floating macrolitter was found in the correspondence of the continental shelf $(573 \pm 572 \text{ items/km}^2)$. This area, which is the natural extent of the mainland, from the coastline to a depth of 200 m, is the most sensitive habitat for the accumulation of floating litter that enters the marine environment via land-based sources. Previously described as an area characterized by low litter seafloor density (Galgani et al., 1996; Pham et al., 2014) could be considered a transition zone of buoyant litter towards pelagic habitats such as submarine canyons, where marine litter has been shown to sink and accumulate (Galgani et al., 1996; Gerigny et al., 2019).

3.4. Marine litter distribution: influence of oceanographic and anthropogenic factors

The distribution of the floating macrolitter and MPs throughout the monitored study area was examined considering the main oceanographic (SST: sea surface temperature; SSH: sea surface height; MLD: mixed layer depth and current velocity) and anthropogenic factors (vessel traffic, distance from ports, distance from the coast and distance from estuaries) that may have influenced their spatial distribution during the sampling campaigns. Correlation analyses show a statistically significant relationship between many of the variables considered (76 %) and concentrations of floating macrolitter. In particular, SST, SSH, fishing vessel density and sailing vessel density showed a weak positive correlation (0 < rho < 0.3) with the amount of litter. Bathymetry showed a stronger significant positive correlation (0.3 < rho < 0.5), while a weak negative correlation (-0.3 > rho > 0) was found between floating macrolitter concentration and mixed layer depth (MLD), current velocity, tanker density, cargo vessel density, distance from nearest major outfalls, and distance from the nearest minor outfall. The correlation of floating macrolitter abundance respectively with distance from the coast and distance from the nearest port was also negative and stronger (-0.5 < rho < -0.3). The descriptive statistical values of each environmental and anthropogenic variable considered and the corresponding correlation values and scatter plots with floating macro pollution concentration were summarised in the Supplementary material (Table S7 and Fig. 2). MPs concentration was significantly related to 47 % of the variables studied. The statistically significant results show a weak positive correlation (0 < rho < 0.3) of microplastic density with sea surface temperature, sea surface height, bathymetry and sailing vessel density. Weak negative correlations (-0.3 < rho < 0) were found for currents, distance from the coast, distance from the nearest port and cargo ship density (Supplementary material Table S8 and Fig. 3A). Generalised additive models (GAM) were applied to further determine the influence of each variable on litter abundance (Supplementary



Fig. 8. Concentrations of floating macrolitter (A) and MPs (B) for different habitats within the study areas. Black lines represent the reference value of mean floating macrolitter/MPs concentration in the Western Mediterranean sub-region and the dashed line represents the mean concentration overall of Pelagos Sanctuary from the present study. * indicates statistically significant difference (p < 0.05).

material Tables S9 and S10). In addition, to better highlight the relationships represented by GAM, a zero line was used to define a positive effect of the predictors on litter accumulation. This was done in a procedure called GAMvelope, described by Torres et al. (2008) (Fig. 9A–C and D–E), and allowed the identification of areas affected by the presence of litter in the Pelagos Sanctuary.

Among the oceanographic variables, SST higher than 297.7 K (24.55 °C), SSH higher than -0.38 m, and currents slower than 0.101 m/s have a positive effect on the accumulation of floating litter (Fig. 9A-C). These results suggest that areas with warmer waters and weak oceanographic features such as lower wave height, slower currents, and no upwelling areas may favour macrolitter accumulation. The influence of certain physical and chemical parameters of oceanic waters on the distribution of litter and sampling activities was clearly outlined by Van Sebille et al. (2020). The so-called "vertical mixing effect" of plastic particles, first described by Kukulka et al. (2012) and also emphasised by Enders et al. (2015) and Reisser et al. (2015), is closely related to wave height and direct wind force, which could facilitate the stratification of plastic particles along the water column according to their physical properties (Kooi et al., 2016). A significant increase in litter has also been observed during daily ocean warming, leading to an accumulation of particles at the warmer sea surface (Kukulka et al., 2016).

Considering the anthropogenic factors, a statistical correlation was found between the amount of floating macrolitter and the distance from the coastline closer than 11 km, the distance from the nearest port closer than 25 km, and the distance from the river mouth between 8 and 37 km (Fig. 9D and E). These results confirm the findings of the spatial analysis of litter (Fig. 3A and B) and the distribution of floating plastics in the Mediterranean Sea modelled by Liubartseva et al. (2019), according to which >75 % of the litter scattered in the sea is located in the 50 km of nearshore waters. These areas can potentially be affected by large amounts of litter originating from nearby land-based sources and coastal maritime activities associated with densely populated areas, as well as inputs from rivers (Jambeck et al., 2015; Lebreton et al., 2017). In the Pelagos Sanctuary, the harbour of Livorno (one of the largest Italian

ports with 30 million tonnes of cargo and 2 million tourists), the Arno (240 km long and crossing several cities, agricultural areas and industrial zones) and Serchio rivers, and the intensive aquaculture and fishing activities near La Spezia could be the main sources of waste and plastic pollution (Cincinelli et al., 2001; Cortecci et al., 2002; Giovacchini et al., 2018; Merlino et al., 2020). Other minor litter inputs could be derived from the port of Genova, which is described to play an important role in litter distribution in the coastal areas of the northern part of the Pelagos Sanctuary, as well as the influence of the Magra river in the transport and accumulation of anthropogenic particles, especially during the summer season (Galgani et al., 2019). Its contribution appears particularly evident in the Tuscan Archipelago National Park due to the mediated transport of plastic by currents towards the southern sector of the SPAMI monitored (Galgani et al., 2019). This area may also be characterized by litter originating from the Tevere and Ombrone rivers, despite their influences that seem heavily affect the Pelagos Sanctuary, especially during the winter season (Galgani et al., 2019).

Sea surface temperature (SST), bathymetry and distance to the nearest port were shown to significantly influence MP distribution (Supplementary material Table S8). However, given the lower explained variance and the paucity of significant variables, no further analysis of MPs were conducted. Moreover, considering the existing predictive models for their distribution (Fossi et al., 2017, 2018a, 2018b; Liubartseva et al., 2019; Mansui et al., 2015; Politikos et al., 2020), the GAMvelope approach was not considered more effective and was applied only to floating macrolitter at the sea surface. Nevertheless, due to the strong correlation found between the spatial concentrations of floating macrolitter can also provide a reliable indication for the accumulation of smaller particles.

Overall, the study area was characterized by a high input of litter coming from the mainland (e.g., harbours and river inputs) and accumulating in coastal waters within about 10–15 nautical miles. The slope area off western Liguria, the continental shelf in the eastern part and the surrounding areas in the Tuscan Archipelago National Park and northeastern Corsica was shown to be particularly characterized by plastic



Fig. 9. GAMs plot of significative oceanographic (A: SST; B: SSH and C: current velocity) and anthropogenic variables (D: distance to the coast; E: distance to the port; and F: distance to river outfall) influencing the floating macrolitter accumulation. The degrees of freedom for non-linear fits are in parenthesis on the y-axis. Tick marks above the x-axis indicate the distribution of observations (with and without sightings). The shaded areas represent the 95 % confidence intervals of the spline functions.



Fig. 10. Floating macrolitter spatial hazard map created considering the oceanographic and anthropogenic factors statistically influencing litter distribution. A hazard score, ranging from 1 to 8, was assigned highlighting areas with different impacts.

litter accumulation (Fig. 10). A moderate risk was present in the canyons of western Liguria and western Corsica, while the least accumulation of plastic was found in the offshore waters over the bathyal plane (Fig. 10). The critical areas highlighted by this spatial risk analysis based on the collected field data show the ecological impact of plastics on the biodiversity inhabiting the Pelagos Sanctuary, especially in the continental shelf ecosystems. The oceanographic variables and anthropogenic activities and the related plastic accumulation in these areas pose a risk to marine species which are exposed to a plethora of anthropogenic stress.

4. Conclusions

The high heterogeneity of marine litter evidence in the available literature stresses the need to create and adopt shared monitoring protocols among the scientific community to collect comparable and consistent data. The harmonized multilevel protocol adopted by this study represents a clear and innovative effort towards a comprehensive assessment of litter impact including transport and accumulation pathways, pollution sources, and potential ecotoxicological effects on marine organisms. Data collected strengthen the effectiveness of the provisional model, as a reliable indirect tool to estimate the litter pollution status of ecologically valuable environments (i.e. SPAMI and National Park and pelagic and coastal protected areas) highlighting areas more at risk for marine organisms. The role of different anthropogenic variables as litteroriginating driving factors has been pointed out, confirming the evaluation of the pollution sources as one of the urgent existing gaps to be addressed and defined. The strong correlation found between the distribution of floating macrolitter objects and microplastics highlighted the significance and effectiveness of the simultaneous floating litter sampling design to better address the presence and distribution of plastic pollution in the marine environment. The multi-tier approach allowed to identify main litter sources: strong litter inputs were identified to originate from the mainland, with significant contribution of ports and estuaries as well as areas with warmer waters and weak oceanographic features (e.g., continental shelf) could facilitate plastic accumulation. Coastal waters, within 10-15 nautical miles, seem to represent litter retention zones, which in turn causes concerns about the underlying risk for marine biodiversity, especially considering the key ecological role of the protected areas of the Pelagos Sanctuary and the Tuscan Archipelago. Overall, the relevant information achieved in this study could serve as an affordable basis for implementing effective marine litter prevention, reduction and disposal policies in MPAs and facilitate future management recommendations and use of the marine and coastal environments of these protected areas.

CRediT authorship contribution statement

Matteo Galli: Formal analysis, Investigation, Writing – original draft, Writing – review & editing, Visualization. Matteo Baini: Conceptualization, Methodology, Formal analysis, Investigation, Writing – original draft, Writing – review & editing. Cristina Panti: Conceptualization, Methodology, Investigation, Resources, Writing – original draft, Writing – review & editing. Dario Giani: Investigation. Ilaria Caliani: Investigation, Writing – review & editing. Tommaso Campani: Investigation. Massimiliano Rosso: Investigation, Visualization. **Paola Tepsich:** Formal analysis, Investigation, Writing – review & editing, Visualization. **Vanessa Levati:** Formal analysis. **Federica Laface:** Investigation. **Teresa Romeo:** Project administration, Funding acquisition. **Gianfranco Scotti:** Investigation. **Francois Galgani:** Investigation, Supervision. **Maria Cristina Fossi:** Conceptualization, Methodology, Validation, Investigation, Resources, Writing – original draft, Writing – review & editing, Visualization, Supervision, Project administration, Funding acquisition.

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.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

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