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## \*CORRESPONDENCE

Guido Pietroluongo  
✉ [guido.pietroluongo@unipd.it](mailto:guido.pietroluongo@unipd.it)

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# Assessment of marine litter interaction in cetaceans stranded along the Italian coastline and the Adriatic Sea

Guido Pietroluongo<sup>1,2\*</sup>, Cinzia Centelleghes<sup>1,3</sup>,  
Matteo Baini<sup>4</sup>, Cristiano Cocumelli<sup>5</sup>, Cristina Casalone<sup>6,7</sup>,  
Giorgia Corazzola<sup>1</sup>, Gabriella Di Francesco<sup>8</sup>, Fabio Di Nocera<sup>9</sup>,  
Ludovica Di Renzo<sup>8,10</sup>, Martina Đuras<sup>11</sup>, Maria Cristina Fossi<sup>4</sup>,  
Stefano Gavaudan<sup>12</sup>, Tilen Genov<sup>13</sup>, Federica Giorda<sup>6,7</sup>,  
Giuseppe Lucifora<sup>9</sup>, Ilaria Pascucci<sup>12</sup>, Antonio Petrella<sup>14</sup>,  
Antonio Pintore<sup>15</sup>, Roberto Puleio<sup>16</sup>, Silva Rubini<sup>17</sup>,  
Giuliana Terracciano<sup>18</sup>, Carla Grattarola<sup>6,7</sup>  
and Sandro Mazzariol<sup>1,3</sup>

<sup>1</sup>Department of Comparative Biomedicine and Food Science, University of Padova, Legnaro, PD, Italy,

<sup>2</sup>Consorzio Nazionale Interuniversitario per le Scienze del Mare (CoNISMa), Roma, Italy,

<sup>3</sup>Interuniversity Center for Cetacean Research (CIRCE), Genova, Italy, <sup>4</sup>Department of Physical Sciences, Earth and Environment, University of Siena, Siena, Italy, <sup>5</sup>Istituto Zooprofilattico Sperimentale del Lazio e della Toscana, Roma, Italy, <sup>6</sup>Istituto Zooprofilattico Sperimentale del Piemonte, Torino, Italy, <sup>7</sup>WOAH Collaborating Centre for the Health of Marine Mammals, Torino, Italy,

<sup>8</sup>Istituto Zooprofilattico Sperimentale dell'Abruzzo e del Molise "G. Caporale", Teramo, Italy, <sup>9</sup>Istituto Zooprofilattico Sperimentale del Mezzogiorno, Portici, Italy, <sup>10</sup>Centro Studi Cetacei, Pescara, Italy,

<sup>11</sup>Department of Anatomy, Histology and Embryology, Faculty of Veterinary Medicine, University of Zagreb, Zagreb, Croatia, <sup>12</sup>Istituto Zooprofilattico Sperimentale dell'Umbria e delle Marche "Togo Rosati", Perugia, Italy, <sup>13</sup>Morigenos–Slovenian Marine Mammal Society, Piran, Slovenia, <sup>14</sup>Istituto Zooprofilattico Sperimentale della Puglia e della Basilicata, Foggia, Italy, <sup>15</sup>Istituto Zooprofilattico Sperimentale della Sardegna, Sassari, Italy, <sup>16</sup>Istituto Zooprofilattico Sperimentale della Sicilia, Palermo, Italy, <sup>17</sup>Istituto Zooprofilattico Sperimentale della Lombardia e dell'Emilia Romagna "Bruno Ubertini", Ferrara, Italy, <sup>18</sup>Istituto Zooprofilattico Sperimentale del Lazio e della Toscana, Pisa, Italy

Marine litter represents a growing threat to marine biodiversity, particularly to cetaceans, yet its impacts on these sentinel species remain insufficiently quantified. This study provides the first comprehensive, transboundary assessment of litter ingestion in stranded cetaceans along the Italian coastline and across the wider Adriatic basin, including Croatia and Slovenia, between 2009 and 2023. Through harmonized *post-mortem* examinations, and focusing on the period of consistent data collection and analysis (2009–2023), this study documented plastic litter ingestion in 2.9% of necropsied cetaceans in Italy and 3.7% in the broader Adriatic subregion, with sperm whales (*Physeter macrocephalus*) showing the highest frequency (50% FO) and susceptibility. In 11 cases, ingestion was associated with health deterioration and mortality. The most commonly ingested items were plastic sheets and fragments. The Italian Adriatic subregion emerged as a hotspot for plastic interactions, reflecting regional hydrodynamics and anthropogenic pressures. Applying criteria from regional and international frameworks, the results showed that 60% of *P. macrocephalus* had ingested more than 1 kg of plastic, with 40% exhibiting harmful effects. These data provide baseline values that can serve as reference points for proposing thresholds to achieve Good Environmental Status under the

Marine Strategy Framework Directive. Despite these results supporting the use of sperm whales as suitable sentinel species for monitoring macroplastic pollution, the available data are limited to Italy and influenced by distributional patterns and unusual mortality events. A combined approach, where *T. truncatus* is monitored for its broad spatial representativeness and *P. macrocephalus* for its ecological susceptibility, may be a useful strategy to guide further research and inform management measures in the future. These findings underscore the need for standardized monitoring protocols, enhanced cross-border data sharing, and policy measures to mitigate plastic impacts. This work provides crucial baseline knowledge for conservation planning and reinforces the role of cetaceans as indicators of ecosystem health in the Mediterranean.

#### KEYWORDS

marine litter, plastic ingestion, cetaceans, post-mortem, Italy, Adriatic sea, marine strategy framework directive, conservation

## 1 Introduction

Marine litter is widely recognized as a serious threat to marine ecosystems (Stelfox et al., 2016). Both ingestion and entanglement are documented to cause injury and mortality in marine fauna, along with sub-lethal effects that compromise essential functions such as foraging, health, and reproductive output (Fossi et al., 2018a; Kühn and Van Franeker, 2020; Senko et al., 2020). More than 900 marine species, including fish, turtles, seabirds, and marine mammals, are known to be affected by marine litter (Stelfox et al., 2016; Thiel et al., 2018; Claro et al., 2019; Woods et al., 2019; Beneli et al., 2020; Kühn and Van Franeker, 2020; Høiberg et al., 2022; Perroca et al., 2022), and approximately 70% of marine mammal species have been observed interacting with anthropogenic debris through ingestion or entanglement (Fossi et al., 2018a; Kühn and Van Franeker, 2020). Over the past two decades, ingestion of marine debris by marine mammals has been widely reported (Fossi et al., 2018a), involving a variety of item types. However, relatively few studies have explored the physiological and pathological impacts of this ingestion on individual animals or populations. These impacts range from no apparent harm, such as from microplastics and associated contaminants, to gastrointestinal blockage, suffocation, starvation, and even death. Entanglement, on the other hand, can lead to severe injury, drowning, or strangulation (Laist, 1997; Duras et al., 2009; Baulch and Perry, 2014; Solomando et al., 2022; Zantis et al., 2021).

Cetaceans serve as key indicators of marine ecosystem health (Wells et al., 2004) and have been proposed as sentinel species for assessing the impact of marine litter at global and regional scales (Zantis et al., 2021). Data collected from stranded marine mammals offer critical insight into the types and frequency of marine litter interactions (Nelms et al., 2019; Fossi et al., 2018b, 2020). Beyond their ecological role, cetaceans also provide significant economic, cultural, and educational value, functioning as flagship species that

foster public engagement in marine conservation (Germanov et al., 2018).

The Mediterranean Sea, characterized by a semi-enclosed geography and intensive anthropogenic use, is particularly vulnerable to marine pollution, with significant consequences for marine biodiversity (Deudero and Alomar, 2015; Darmon et al., 2017; Fossi et al., 2017, 2020; Alomar et al., 2020). It is considered one of the most impacted marine regions globally, with seafloor surveys reporting up to 43.55 items of anthropogenic litter per 100 m<sup>2</sup>, in comparison to Northeast Atlantic (0 to 10.1 items/100 m<sup>2</sup>) (Consoli et al., 2018), and floating marine litter densities of approximately 0.00021 items per 100 m<sup>2</sup>, compared to other areas such as the Baltic or North Seas (0.000126 ± 0.000082 items/100 m<sup>2</sup>) (Pham et al., 2014). Within the Mediterranean, the Adriatic Sea has been identified as one of the areas most affected by benthic litter accumulation (Pasquini et al., 2016). Further studies have demonstrated that the Adriatic acts as a convergence zone for plastic debris due to its geomorphology and hydrodynamics (Liubartseva et al., 2016; Ruiz-Orejón et al., 2016; Vianello et al., 2018; Zambianchi et al., 2017). Major litter inputs originate from coastal activities, urban and industrial discharges, maritime transport, fisheries, and aquaculture, with transboundary transport facilitated by sea currents (Vlachogianni et al., 2018; Fortibuoni et al., 2019; Palatinus et al., 2019; Galli et al., 2023).

At the policy level, the European Commission, with the 2008/56/EC Marine Strategy Framework Directive (MSFD; 2008/56/EC), has designated sea turtles as an indicator taxon for monitoring the amount and composition of litter ingested by marine animals (Galgani et al., 2013, 2023; Matiddi et al., 2017; Darmon et al., 2017). An analogous framework guiding marine litter monitoring is the Regional Plan on Marine Litter Management (UNEP/MAP, 2012) for the protection of the Mediterranean Sea, following the Ecosystem Approach. At the international level, both the

International Whaling Commission (IWC) and the Agreement on the Conservation of Cetaceans of the Black Sea, Mediterranean Sea and Contiguous Atlantic Area (ACCOBAMS) have recognized marine litter as a significant conservation threat. Both organizations have promoted coordinated monitoring efforts and organized workshops to assess the impact of marine litter on cetaceans. Notably, the IWC's 2019 Marine Debris Workshop in La Garriga (Barcelona) reviewed existing evidence and recommended best practices for *post-mortem* debris assessment (International Whaling Commission, 2020). Similarly, ACCOBAMS prioritized marine litter in its 2017–2019 Work Program and launched pilot projects using standardized necropsy protocols for collecting data on litter interactions from stranded cetaceans. Within the ACCOBAMS area, stranding monitoring has proven effective in generating baseline data on this issue.

*Post-mortem* examinations, especially those analyzing the gastrointestinal tract, are vital for assessing both lethal and sub-lethal effects of marine litter, often in conjunction with demographic and spatial data (Bond and Lavers, 2013; Provencher et al., 2014; Duncan et al., 2017; Nelms et al., 2019). However, the assessment of marine litter ingestion in cetaceans poses significant methodological and logistical challenges due to the large size of the gastrointestinal tract, the decomposition state of carcasses, often remote nature of stranding locations, and limitations during necropsy procedures. Therefore, the presence of marine litter has historically been reported only as an incidental finding during dietary analyses and has mostly been limited to macroplastic items (De Stephanis et al., 2013; Alexiadou et al., 2019; Panti et al., 2019). This aspect has made it difficult to properly assess the impact. To address this significant knowledge gap, several authors have proposed specific, standardized necropsy protocols for the detection and quantification of both macro and microplastics in gastrointestinal samples to enhance data comparability and better assess the role of marine debris in cetacean mortality (Moore and van der Hoop, 2012; Fossi et al., 2018a, 2018b; Corazzola et al., 2021).

Although marine litter ingestion and entanglement have been documented in several cetacean species along the Adriatic coasts, particularly in *Tursiops truncatus* (common bottlenose dolphin), *Physeter macrocephalus* (sperm whale), and *Ziphius cavirostris* (Cuvier's beaked whale) (Gomerči et al., 2006; Mazzariol et al., 2011, 2018; Đuras et al., 2021), the overall magnitude and consequences of these interactions remain poorly understood. These species display markedly different ecological characteristics, spatial distributions in the Mediterranean, and life-history traits. *T. truncatus* is the most common coastal cetacean, widely distributed along continental shelves and nearshore waters exposed to intense human activity, and is listed as Vulnerable in the Mediterranean by the International Union for Conservation of Nature (IUCN); *P. macrocephalus* inhabits deep offshore waters of the Western and Central Mediterranean, forming a small, resident subpopulation classified as Endangered; *Z. cavirostris* occupies slope and canyon habitats across the basin and is also assessed as Vulnerable (ACCOBAMS, 2021). Their contrasting spatial distributions and diving capacities influence species-specific foraging behavior and,

consequently, the degree of exposure to marine litter and other anthropogenic stressors.

In the Mediterranean region, Italy, Slovenia, and Croatia have established national cetacean stranding networks to monitor and collect data on stranded individuals. While all three countries are committed to this objective, their networks differ in terms of geography, logistics, investigative capacity, facilities, and available expertise. As member states of ACCOBAMS, common protocols and resolutions, specifically Resolution 8.15 on cetacean stranding networks, are available for adoption to enhance coordination, apply standardized procedures, and ensure harmonized data collection and *post-mortem* examinations across the region (IJseldijk et al., 2019). Routine monitoring of strandings along the regional coastlines facilitates the collection of demographic and mortality data, including information on evidence of interactions with human activities, and supports surveillance of emerging and zoonotic diseases of concern. However, despite these shared frameworks, the actual implementation is not consistently applied, which can lead to discrepancies in data collection practices and interpretation of results.

The primary objective of this study is to provide a comprehensive overview of marine litter interactions, unrelated to fishery activities, in stranded cetaceans along the Italian coastline. A secondary objective is to extend this overview to the wider Adriatic basin, including available data from Slovenia and Croatia. Ultimately, the study aims to assess the impact of marine litter on cetacean health, providing recommendations within the context of the most relevant regional and international conservation frameworks.

## 2 Materials and methods

To achieve the objectives of this study, we reviewed the data provided by the three national stranding networks (Italy, Slovenia, Croatia), focusing particularly on interactions with marine litter. During *post-mortem* examinations, marine litter was recorded when found either in the gastrointestinal tract (esophagus, stomach chambers, intestines) or entangled around anatomical features such as the flippers, dorsal fin, fluke, larynx, or the whole body. When possible, the litter items were classified and categorized according to type and origin. Only items not related to fishery activities are included in this study, following the UNEP definition as any persistent material that has been manufactured or processed and ends up in the marine and coastal environment after being discarded, abandoned, or improperly disposed of (Cheshire et al., 2009). Fishery-related items were excluded because their assessment is the subject of a separate study (Pietroluongo et al., 2025) not included in the scope of the present study. Furthermore, according to the definition provided by the MSFD Technical Subgroup on Marine Litter (Galgani et al., 2013, 2023), the analysis focused exclusively on macro litter (items larger than 2.5 cm) and meso litter (items between 5 mm and 2.5 cm). No compositional analyses (e.g., FTIR or Raman spectroscopy) were performed to confirm the polymeric nature of the materials visually identified as plastics. The classification of use-type (USE) marine litter sources was based on

the categorization used for biota (birds and sea turtles) by the same MSFD Technical Subgroup (Table 1). Additionally, the stranded locations were categorized according to MSFD subregions in the Western Mediterranean Sea (WMED), the Central Mediterranean Sea (CMED), and the Adriatic Sea (ADRIA) (Table 1), as defined by Jensen et al. (2015).

The datasets from the three countries cover the period 1986 to 2023, although the duration and intensity of monitoring efforts varied. In Italy, the dataset spans from 1986 to 2023 (38 years) with 6393 stranding reports and 1238 undergoing necropsy. In Slovenia, data were collected between 2002 and 2023 (22 years), with 26 carcasses analyzed. For Croatia, data span from 1990 to 2023 (34 years), with 310 carcasses analyzed, incorporating updates and revisions to the previously published dataset (1990–2019) by Đuras et al. (2021). Given that systematic analyses of marine litter during *post-mortem* examinations began in Italy in 2009, and to ensure methodological consistency across the 3 countries, this study included only cetaceans stranded over the past 15 years (2009–2023), as detailed in Table 2.

When possible, the frequency of occurrence (%FO) of marine litter ingestion is reported per species, based on findings during necropsy, in relation to the total number of examined carcasses as follows:

$$\%FO = \left( \frac{Ni}{N} \right) \times 100$$

where  $N_i$  corresponds to the number of a species of cetaceans with evidence of marine litter ingestion, and  $N$  corresponds to the total number of necropsied cetaceans of the same species in the study period.

To further evaluate temporal and regional variability, the %FO was calculated for each species in each year within the 2009–2023 study period. Annual %FO values were then used to derive the mean and standard deviation (SD) across years, providing an estimate of uncertainty around ingestion frequency and allowing comparison of interannual variability between Italy and Croatia.

For Croatia, %FO values were calculated based on available data from 2016 and 2018.

This approach allows a quantitative assessment of temporal fluctuations and uncertainty within and between national datasets under the MSFD framework.

Due to the heterogeneity of *post-mortem* investigations, characterized by differing protocols, resources, and interpretive approaches regarding the impact of marine litter on animal health, the criteria proposed by Mazzariol et al. (2020) in Annex 5 of the International Whaling Commission (2020) were retrospectively applied to ensure standardization. These evidence-based diagnostic frameworks classify findings according to the severity of gastrointestinal involvement: from incidental presence of debris without lesions, to contributory cases showing partial repletion/obstruction and associated pathology, up to probable cause of death characterized by perforation, severe impaction/obstruction, with severe presence of lesions associated. In addition to documenting the presence and characteristics of foreign objects, all pathological *post-mortem* findings described in the original necropsy reports and referenced within the diagnostic framework were considered to assess the potential impact of marine debris on the health of the examined animals.

Finally, to establish a baseline data for species-specific threshold value (TV), the general MSFD criteria were followed: D10C3 (Ingestion: “The amount of litter and microlitter ingested by marine animals is at a level that does not affect the health of the species concerned”) and D10C4 (Health impact: “The number of individuals of each species that are adversely affected due to litter, such as through entanglement, other types of injury or mortality, or health effects”). Numeric cut-offs were based on the empirical distribution of debris mass in the Italian dataset. The ingestion baseline value was defined as the lowest debris mass consistently associated with pathological signs, thus distinguishing incidental ingestion from biologically harmful ingestion. The baseline value was expressed as the maximum proportion of individuals exceeding the ingestion and impact criteria within a rolling six-year

TABLE 1 Classification of marine litter per category and MSFD subregions (Galvani et al., 2013, 2023; Jensen et al., 2015).

| Classification           | Type                      | Code     | Description                                                                                                                                                                      |
|--------------------------|---------------------------|----------|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Marine litter categories | Industrial plastic        | IND PLA  | Industrial plastic granules, usually cylindrical but also sometimes oval, spherical, or cubical shapes, or suspected industrial items, used for the tiny spheres (glassy, milky) |
|                          | Use sheet                 | USE SHE  | Remains of sheet, e.g., from bag, cling-foil, agricultural sheets, rubbish bags, etc.                                                                                            |
|                          | Use thread                | USE THR  | Threadlike materials, e.g., pieces of nylon wire, net fragments, woven clothing; includes ‘balls’ of compacted such material                                                     |
|                          | Use foam                  | USE FOA  | All foamed plastics, e.g., polystyrene foam, foamed soft rubber (as in mattress filling)                                                                                         |
|                          | Use fragment              | USE FRAG | Fragments, broken pieces of thicker type plastics, can be a bit flexible, but not like sheet-like materials                                                                      |
|                          | Other use plastics        | USE POTH | Any other type of plastics, including elastics, dense rubber, cigarette filters, balloon pieces, and soft airgun bullets                                                         |
|                          | Litter other than plastic | OTHER    | All non-plastic rubbish and pollutants                                                                                                                                           |
| MSFD subregions          |                           | WMED     | Western Mediterranean Sea                                                                                                                                                        |
|                          |                           | CMED     | Central Mediterranean Sea                                                                                                                                                        |
|                          |                           | ADRIA    | Adriatic Sea                                                                                                                                                                     |

TABLE 2 Stranding reports and analyzed carcasses within Italy, Slovenia, and Croatia between 2009 and 2023, and the MSFD subregions.

| 2009–2023 (15 years) | MSFD subregion | N° stranding reports | N° analyzed carcasses |
|----------------------|----------------|----------------------|-----------------------|
| Italy                | WMED           | 1423                 | 555                   |
|                      | CMED           | 374                  | 155                   |
|                      | ADRIA          | 790                  | 287                   |
|                      | TOT            | 2587                 | 997                   |
| Slovenia             | ADRIA          | 20                   | 20                    |
| Croatia              | ADRIA          | 361                  | 153                   |

assessment window, as established by the MSFD during the review process cycle by the Member States.

3 Results

The geographic distribution of stranded cetacean with evidence of marine litter ingestion along the study area and corresponding MSFD subregions (WMED, CMED, ADRIA) are shown in Figure 1.

3.1 Italy

In Italy, 29 animals were reported to have ingested plastic items between 2009 and 2023 (Supplementary Table 1), out of 997 analyzed carcasses (2.9% FO, with a yearly rate of 1.9 cases per year) (Figure 2).

Overall, four odontocete species were found to have ingested plastic debris (Table 3). *P. macrocephalus* showed the highest frequency of occurrence (%FO = 50%), followed by *Z. cavirostris* (12.5%), *Stenella coeruleoalba* (striped dolphin) (2.3%), and *T. truncatus* (1.3%). The majority of the animals were adults (20 adults, 8 juveniles, 1 not determined-ND) and females (16 females, 12 males, 1 ND). When annual %FO values were examined across the 15-year study period (2009–2023), clear interannual fluctuations were observed. In Italian waters, the mean %FO ( $\pm$  SD) was  $5.2 \pm 3.7$  for *S. coeruleoalba*,  $1.4 \pm 1.7$  for *T. truncatus*,  $30.0 \pm 43.0$  for *P. macrocephalus*, and  $11.1 \pm 33.3$  for *Z. cavirostris* (Table 4). Temporal fluctuations were evident, with marked peaks in 2009, 2014, and 2019, years corresponding to unusual mortality events (UMEs) involving sperm whales. These results highlight the strong interannual variability and emphasize the importance of incorporating uncertainty estimates when evaluating ingestion rates across long-term datasets.

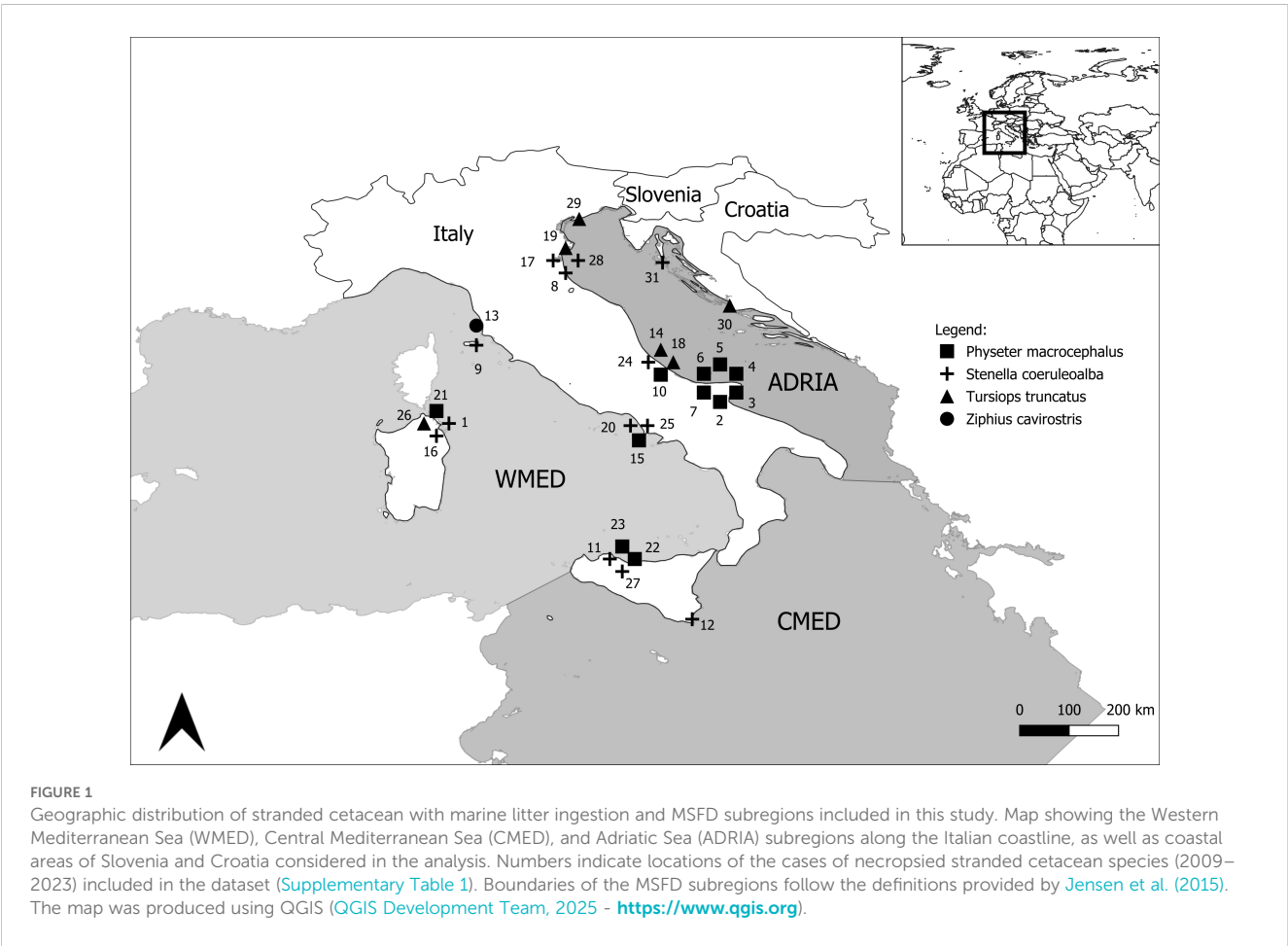




TABLE 3 Frequency of occurrence (%FO) and yearly rate of marine litter ingestion by species in Italy (2009–2023).

| Species | N° of analyzed carcasses (N) | N° of analyzed carcasses with evidence of marine litter ingestion (Ni) | %FO  | Yearly rate |
|---------|------------------------------|------------------------------------------------------------------------|------|-------------|
| Sc      | 521                          | 12                                                                     | 2.3  | 0.8         |
| Tt      | 385                          | 5                                                                      | 1.3  | 0.3         |
| Pm      | 22                           | 11                                                                     | 50.0 | 0.7         |
| Zc      | 8                            | 1                                                                      | 12.5 | 0.06        |

Species: Sc: *Stenella coeruleoalba*; Tt: *Tursiops truncatus*; Pm: *Physeter macrocephalus*; Zc: *Ziphius cavirostris*.

TABLE 4 Annual and mean ( $\pm$  SD) frequency of occurrence (%FO) of marine litter ingestion for cetacean species analyzed in Italy (2009–2022) and Croatia (2016–2018). Mean and standard deviation summarize interannual variability and uncertainty across the study period.

| Italy   |       |      |          |                        |      |      |      |      |      |          |                        |
|---------|-------|------|----------|------------------------|------|------|------|------|------|----------|------------------------|
| Species | 2009  | 2012 | 2014     | 2017                   | 2018 | 2019 | 2020 | 2021 | 2022 | Mean FO% | Standard deviation %FO |
| Sc      | 14.28 | 7.69 | 3.22     | 4.25                   | 4.76 | 4.16 | 3.22 | 2.27 | 3.33 | 5.24     | $\pm$ 3.72             |
| Tt      | 0     | 0    | 0        | 0                      | 3.70 | 2.04 | 4.16 | 0    | 2.38 | 1.36     | $\pm$ 1.73             |
| Pm      | 100   | 0    | 20       | 0                      | 100  | 50   | 0    | 0    | 0    | 30.00    | $\pm$ 43.01            |
| Zc      | 0     | 0    | 0        | 100                    | 0    | 0    | 0    | 0    | 0    | 11.11    | $\pm$ 33.33            |
| Croatia |       |      |          |                        |      |      |      |      |      |          |                        |
| Species | 2016  | 2018 | Mean FO% | Standard deviation %FO |      |      |      |      |      |          |                        |
| Sc      | 0     | 100  | 50.00    | $\pm$ 70.71            |      |      |      |      |      |          |                        |
| Tt      | 7.14  | 0    | 3.57     | $\pm$ 5.05             |      |      |      |      |      |          |                        |

According to the MSFD classification, 3 categories of marine litter were reported (Figures 3, 4), such as USE SHE (45.2%), consisting of remains of sheets from plastic bags, plastic cups, plastic tarp (from agricultural activities), plastic wrappings; USE FRAG (41.9%) of plastic cups and broken pieces from thick plastics; and USE THR (12.9%), consisting of plastic ropes and strings.

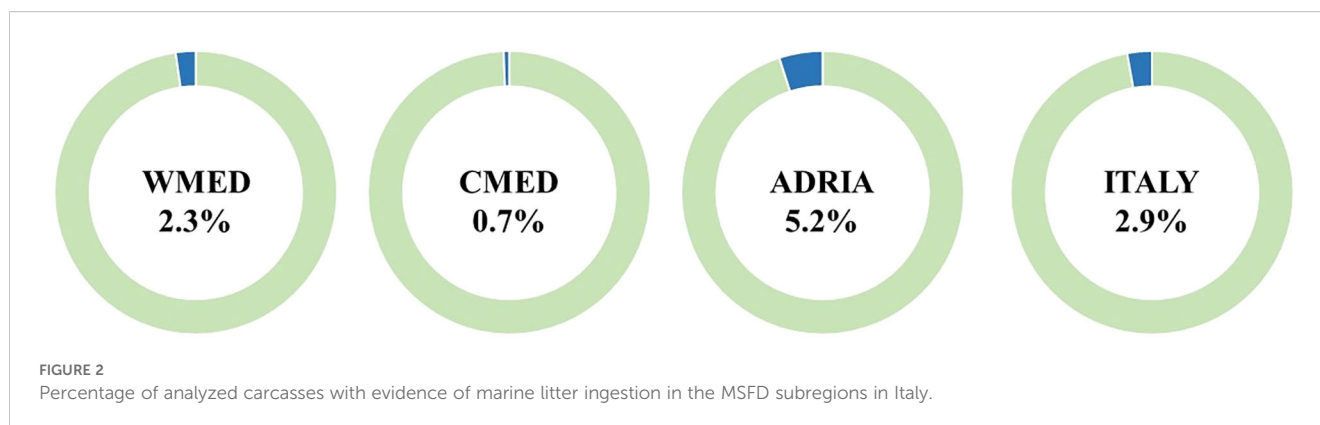
As shown in the Supplementary Table 1, all 3 categories of marine litter were equally documented in WMED and ADRIA, while in CMED only 1 case of USE THR was recorded (Figure 3).

Among the Italian MSFD subregions, ADRIA showed the highest percentage of plastic litter ingestion (5.2%) (Figure 4) and yearly rate (WMED: 0.9; CMED: 0.06; ADRIA: 1; total yearly rate: 1.9).

Among the species, *P. macrocephalus* showed the highest exposure to plastic ingestion (Supplementary Table 1). The most frequently reported marine litter category for *P. macrocephalus* was USE SHE, and the most affected MSFD subregions were ADRIA and WMED; in the cases of 2009, 2014, and 2019, the specimens were stranded during 3 different unusual mortality events (UME) (Mazzariol et al., 2011; 2018). For *S. coeruleoalba*, the most reported marine litter category was also USE SHE, with WMED as the main subregion. Finally, for *T. truncatus*, the predominant marine litter category was USE FRAG, with ADRIA as the most affected MSFD subregion. It is interesting to note the presence of a plastic label from Libya (case 27 in Supplementary Table 1) in an adult male *S. coeruleoalba* found stranded in the WMED, highlighting how both

cetaceans (Genov et al., 2022) and marine litter can actively or passively travel long distances.

Among all the cases with plastic litter ingestion, this finding was considered to have an impact on the animal's health status in 20.7% of cases (6/29) and may have contributed to their strandings (0.6% of the analyzed carcasses). Specifically, in an adult female *S. coeruleoalba* found stranded in the WMED subregion (Sardinia) in 2009 (case 1 in Supplementary Table 1), stomach occlusion caused by a piece of plastic bag (USE SHE) was determined to be fatal. Similarly, in another *S. coeruleoalba* found stranded in the ADRIA subregion (Veneto) in 2012 (case 8 in Supplementary Table 1), a piece of plastic bag (USE SHE) obstructed the opening between the first and second stomach chambers, and was associated with a heavy parasitic burden of *Anisakis* spp. and *Pholeter gastrophyllus*. Plastic litter was also identified as a key contributing factor to the poor health conditions observed in 4 sperm whales, accounting for 36.4% of the individuals of this species that had ingested plastic (cases 2, 3, 10, and 21 in Supplementary Table 1). In these cases, the weight and volume of the ingested plastic were deemed sufficient to predispose the animals to poor nutritional condition, often accompanied by gastric impaction and/or concurrent diseases. For the remaining individuals, logistical constraints or an advanced state of decomposition prevented a conclusive *post-mortem* assessment of the impact of marine litter on their health status.



Only the reports on *P. macrocephalus* described the plastic litter mass, ranging from 10 g to 29 kg (Supplementary Table 1), with a median of 1151 g. No individual with health impact had a debris load below 1 kg. Based on these observations, 1 kg was identified as the lower bound separating incidental ingestion from harmful ingestion, accounting for 22.7% of the analyzed carcasses (5/22) and 6.3% of the total stranding of sperm whales (5/80) in Italy. In the Adriatic Sea subregion (ADRIA), plastic litter ingestion above 1 kg (D10C3) occurred in 57.1% of analyzed carcasses (4/7), and 26.7% of the total stranding of sperm whales (4/15) in this subregion. Related health impact (D10C4) was reported in 42.9% (3/7) and 20% (3/15) of the total stranding in this subregion. For the Western Mediterranean Sea subregion, 1 kg (D10C3) occurred in 6.7% of analyzed carcasses (1/15), and 1.7% of the total stranding of sperm whales (4/58) in this subregion. Related health impact (D10C4) was reported with the same percentage. Based on these observations, 1 kg was identified as the lower bound separating incidental from harmful ingestion. These observed proportions were used to define the preliminary GES baseline percentages, rounded to the nearest ten percentage points (60% for D10C3 and 40% for D10C4) to avoid false precision given the small sample size. These represent empirical, provisional baseline values that will require confirmation and refinement as additional data become available.

These proportions were therefore used to propose the preliminary GES baseline values for the species:

- DC10C3: 60% of *P. macrocephalus* with more than 1 kg of ingested plastic in samples of at least 7 dead individuals.
- DC10C4: 40% of *P. macrocephalus* with more than 1 kg of harmful effect of ingested plastic in samples at least 7 dead individuals.

### 3.2 Adriatic Sea

All 3 Adriatic countries conducted analyses over the same 15-year period from 2009 to 2023. The review of Slovenian and Croatian cases revealed a different scenario, as shown in Table 5. While no marine litter items were documented in Slovenia among

20 analyzed carcasses, plastic litter ingestion was confirmed in Croatia in 1 adult female *T. truncatus* (case 30 in Supplementary Table 1) and 1 adult male *S. coeruleoalba* (case 31 in Supplementary Table 1), out of a total of 153 analyzed carcasses. In both cases, plastic litter was hypothesized to have impacted the health of the animals due to impaction and/or occlusion. In Croatian waters, % FO values were available for 2016 and 2018. The mean %FO ( $\pm$  SD) was  $50.0 \pm 70.7$  for *S. coeruleoalba* and  $3.6 \pm 5.0$  for *T. truncatus* (Table 4). Although based on a limited number of necropsies, these data confirm that marine litter ingestion occurs along the eastern Adriatic as well. Overall, including the aforementioned Italian cases (Table 3), the ADRIA subregion shows a 3.7%FO of plastic litter ingestion among all examined carcasses, with 6 animals showing health impacts associated with plastic ingestion (Supplementary Table 1), representing 35.3% of the individuals with plastic ingestion (6/17) and 1.3% of the total analyzed carcasses (6/460). Including the Croatian cases, the USE SHE category remained the most frequently represented MSFD marine litter type in this subregion.

## 4 Discussion

Between 2009 and 2023, 2.9% of stranded cetaceans along the Italian coastline were found to have ingested plastic, based on frequency of occurrence (%FO). The Italian Adriatic subregion (ADRIA) exhibited a slightly higher %FO of 5.2% (15/287), indicating a relatively greater impact compared to other Italian subregions, such as the Western Mediterranean (WMED), where the ingestion rate was lower (2.3%). A similar pattern is apparent when considering the broader Adriatic region, although comprehensive data for comparing such transboundary patterns in the Western Mediterranean remain limited.

Notably, all ingestion cases involved Odontoceti, supporting existing evidence that this taxonomic group is particularly susceptible to plastic ingestion due to its feeding behavior and ecological traits (Walker and Coe, 1989). However, this may also be, at least in part, due to the fact that only 1 species of Mysticeti regularly inhabits the Mediterranean Sea, out of a total of 10 regular species (ACCOBAMS, 2021). In Italy, two odontocete species were most frequently affected: *S. coeruleoalba* (n=12) and *P.*

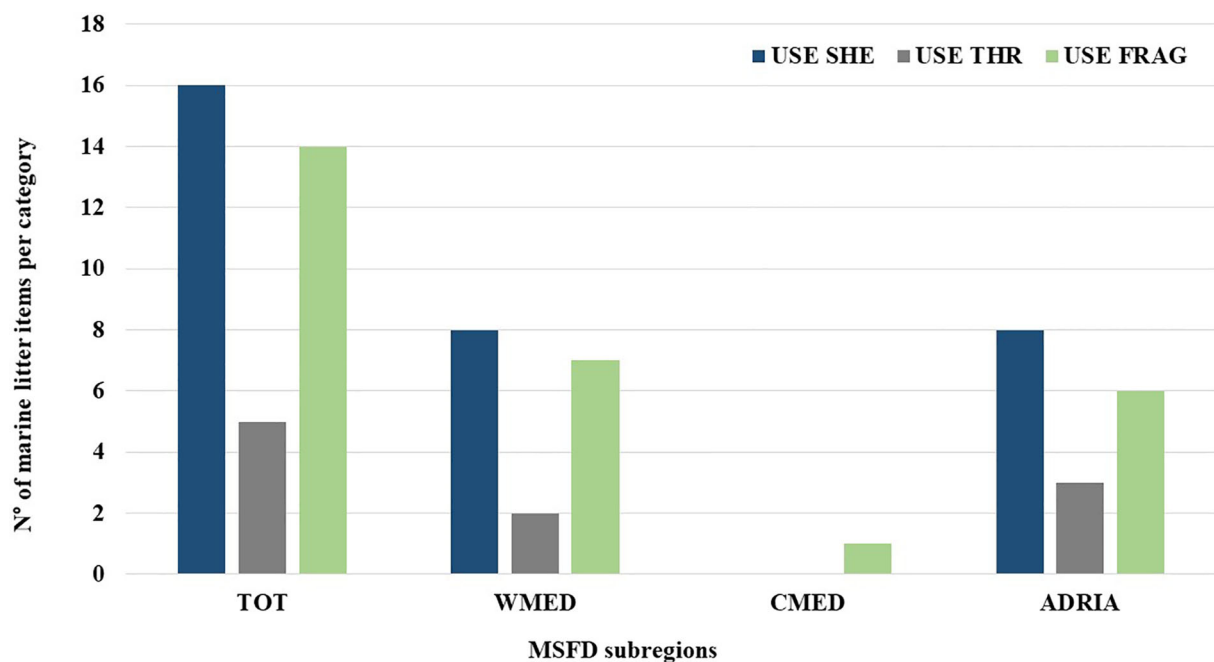


FIGURE 3  
Marine litter categories per MSFD subregions in Italy.

*macrocephalus* (n=11). However, their relative vulnerability differed markedly. *S. coeruleoalba* had a lower %FO of 2.3%, while *P. macrocephalus* showed a notably high %FO of 50%. This disparity likely reflects both biological and methodological factors. In particular, *S. coeruleoalba* may be exposed to floating debris in offshore pelagic zones, and the individuals stranded outside their typical distribution range in the central and northern Adriatic may reflect altered foraging behavior or compromised health, potentially increasing their risk of debris ingestion. Conversely, the deep-diving, suction-feeding behavior of sperm whales (Werth, 2004), along with their large forestomachs adapted for storing prey (Cozzi et al., 2017), increases their exposure to plastic debris, particularly on or near the seafloor. Furthermore, the size and number of ingested items in sperm whales often make them more easily detectable through macroscopic visual analysis. Merrill et al. (2024) further support this explanation, demonstrating that 100% of plastic debris possess acoustic target strengths equal to or greater than those of typical deep-sea prey, potentially increasing the risk of mistaken ingestion. Given these traits, *P. macrocephalus* has been

proposed as a strategic indicator species for monitoring marine litter ingestion in the region (Fossi et al., 2020).

Although *T. truncatus* is the most widely distributed species in the Adriatic, the number of plastic ingestion cases was equal to that of *S. coeruleoalba* (5 each). This may reflect the bottlenose dolphin's greater ability to distinguish prey from debris (Kellogg, 1962) and its adaptation to synanthropic habitat (Bonizzoni et al., 2021). In contrast, *S. coeruleoalba* stranded in central and northern Adriatic areas, outside their typical range (ACCOBAMS, 2021), may reflect altered foraging behavior due to compromised health. It should be noted, however, that the percentages reported may be partially biased, since during the first study period gastrointestinal tracts were not systematically examined for litter.

Plastic ingestion can severely impair cetacean digestive physiology, particularly in species with complex gastrointestinal systems such as deep-diving odontocetes like *P. macrocephalus*. The accumulation of indigestible materials in the forestomach (first stomach chamber) may reduce gastric filling capacity, create a false sense of satiety, suppress appetite, and lead to malnutrition (Werth,

TABLE 5 Summary of number and %FO of analyzed carcasses with evidence of plastic litter ingestion, yearly rate, and number of animals with health impairment in the Adriatic Sea subregion (ADRIA).

| 2009-2023 | N° of analyzed carcasses with evidence of marine litter ingestion | %FO of analyzed carcasses with evidence of marine litter ingestion | Yearly rate | N° of animals with health impairment |
|-----------|-------------------------------------------------------------------|--------------------------------------------------------------------|-------------|--------------------------------------|
| ITALY     | 15                                                                | 5.2 (15/287)                                                       | 1           | 5                                    |
| CROATIA   | 2                                                                 | 1.3 (2/153)                                                        | 0.1         | 2                                    |
| SLOVENIA  | 0                                                                 | 0 (0/20)                                                           | 0           | 0                                    |
| ADRIA     | 17                                                                | 3.7 (17/460)                                                       | 1.1         | 7                                    |



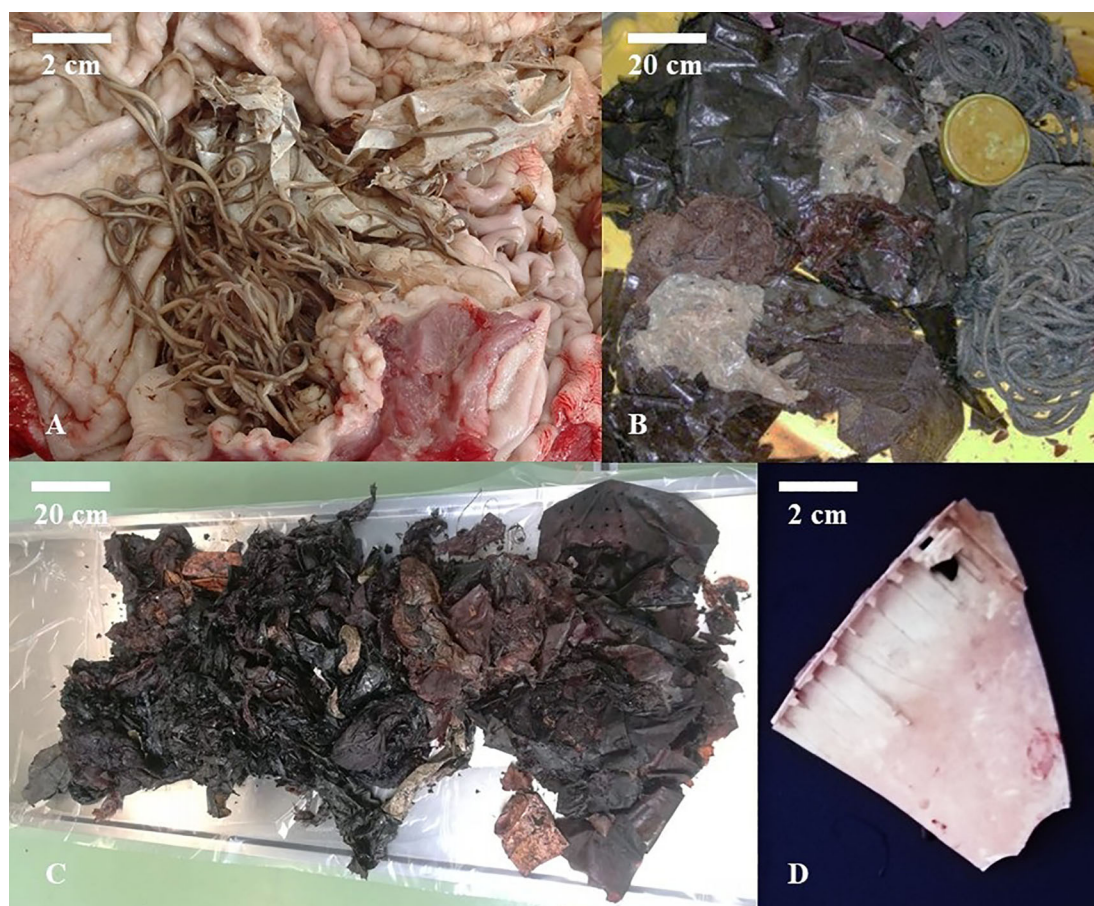


FIGURE 4

Plastic litter found in the stomach chambers of stranded cetacean carcasses. (A) A piece of a plastic bag (USE SHE) in the opening between the I and II stomach chambers of *S. coeruleoalba* causing occlusion (case 8 [Supplementary Table 1](#)); (B) Plastic bags, plastic tarp, and rope (USE SHE, USE THR) inside the I stomach chamber of *P. macrocephalus* (case 3 [Supplementary Table 1](#)); (C) 29 kg plastic mixed materials (USE SHE, USE THR, USE FRAG) in the I stomach chamber of *P. macrocephalus* (case 21 [Supplementary Table 1](#)); (D) Hard plastic fragment (USE FRAG) inside the I stomach chamber of *P. macrocephalus* (case 23 [Supplementary Table 1](#)).

2004; Cozzi et al., 2017). Plastic debris can also obstruct peristaltic movement between stomach compartments and mechanically interfere with gastric emptying (Baulch and Perry, 2014; Alexiadou et al., 2019). The present study documented cases reinforcing these concerns. In Italy, 93.1% of ingestion cases showed plastic localized in the forestomach, with only isolated findings in the mouth and duodenum. This anatomical compartment appears particularly vulnerable, as its structure facilitates the entrapment of flat, flexible items like bags, strings, and ropes, especially when combined with prey remains, which may accumulate and obstruct digestion (Cozzi et al., 2017). Health impacts were recorded in 31% of ingestion cases in Italy and 41.2% in the broader Adriatic region. For instance, occlusion of stomach chamber openings and impaction of the stomach chamber were recorded in 2 *S. coeruleoalba* and 1 *T. truncatus*, where the characteristics of plastic items may have prevented their passage into the glandular stomach, causing further entrapment and digestive impairment. Additional impacts were observed in another *S. coeruleoalba* and 7 *P. macrocephalus* in the Adriatic Sea, where plastic presence was associated with poor

nutritional status and concurrent diseases, such as Morbillivirus infections (Mazzariol et al., 2011; 2018). It remains unclear whether plastic ingestion acted as a predisposing factor or if it was a consequence of the animals' condition.

Geographical variability in plastic ingestion patterns was observed. Most cases were along the Italian coast ( $n=15$ ), compared to two in Croatian waters. The analysis of temporal and regional %FO patterns provided additional insight into interannual variability and uncertainty. Peaks in 2009, 2014, and 2019 coincided with UMEs involving *P. macrocephalus*. The comparatively higher %FO values in Croatian waters, especially for *S. coeruleoalba*, is based on limited years. This discrepancy may be influenced by coastline length and type, anthropogenic pressure, and transboundary processes such as marine currents that transport debris across regions (Baulch and Perry, 2014; Vianello et al., 2018; Fortibuoni et al., 2019; Palatinus et al., 2019). Interestingly, the relatively low ingestion rate reported contrasts with the high estimated plastic load entering the Adriatic (Arcangeli et al., 2018). These results underline the need for harmonized, long-

term monitoring of ingestion rates across the basin to ensure robust MSFD indicator evaluation. Additional factors such as carcass drift, oceanographic dynamics, and differences in stranding response systems may also contribute. Further research, including carcass drift modeling (Genov et al., 2016; Carlucci et al., 2020), is recommended to better understand spatial patterns of marine litter and cetacean exposure.

In other Mediterranean regions with long-term monitoring, such as the Balearic Islands, with a study by Solomando et al. (2022) between 2019 and 2022 (4 years), marine litter ingestion was observed in 1 *P. macrocephalus*, which had ingested three packaging straps, one plastic bag, and two plastic sheets as described also in this study. In Greek seas, a study between 1993 and 2014 (22 years) (Alexiadou et al., 2019) showed a %FO of 26.5% (7/34), with a yearly rate of 0.41. As for Italy and the Adriatic subregion, in the Eastern Mediterranean Sea subregion (EMED), the most affected species was *P. macrocephalus* with 60%FO, slightly higher compared to Italy and the Adriatic Sea, followed by *Z. cavirostris* (25%FO, 1/4), *Grampus griseus* (Risso's dolphin) (20% FO, 1/5), and *Phocoena phocoena* (harbor porpoise) (20%FO, 1/5). No cases were reported for *T. truncatus* or *S. coeruleoalba*. Only in 3 animals (8.8%) was the cause of death hypothesized to be related to the ingestion of marine litter, a lower percentage compared to Italy. Beyond the Mediterranean Sea, other long-term monitoring efforts showed a higher %FO compared to Italy (2.8%) and the Adriatic subregion (3.7%), such as 7.7% recorded in the Canary Islands (Puig-Lozano et al., 2018). Among species, *P. macrocephalus* showed greater susceptibility in Italy (50%FO) than in the Canary Islands (21.4%FO), whereas *S. coeruleoalba* and *T. truncatus* had higher %FO in the Canary Islands (3.3%FO and 2.5%FO, respectively) compared to Italy (2.3%FO and 1.3%FO). Severe health effects from ingested plastics (USE SHE) were more frequent in the Canary Islands (80.6% of ingestion cases, 6.2% of total analyzed carcasses) than in Italy (20.7%, 0.6%) and the Adriatic (35.3%, 1.3%). Another study in the Western South Atlantic found a %FO of 33.8 (52/154) across all investigated species, but a lower %FO of 3.3% (1/30) for *T. truncatus*, which was the only species in common with our study (Zimmer-Correa et al., 2024). In the study by Zimmer-Correa et al. (2024), FRAG was reported in 70.5% of cases, substantially higher compared to the present study. No cause of death related to the interaction with plastic litter was reported. Other studies reported isolated cases without long-term datasets (e.g., Walker and Coe, 1989; De Stephanis et al., 2013; Tonay et al., 2021; Çanakçı et al., 2023), underscoring the need for sustained monitoring and the underestimation of this threat in certain areas and species. Limitations shared by Italy, Slovenia, and Croatia include geographic and logistical constraints that hinder carcass retrieval and *post-mortem* analysis, as well as the application of the harmonized regional protocols for evaluating marine litter, especially microplastics. An additional aspect not included in the present study is that no compositional analyses (e.g., FTIR, Raman) were conducted to confirm the chemical nature of the marine litter identified as plastic. Moreover, the present study focused exclusively

on macro- and meso-litter, while microplastics were not investigated. Future studies should include compositional verification and microplastic quantification to provide a more comprehensive evaluation of plastic contamination in cetaceans. Advanced decomposition also impairs diagnostic capacity, restricting the reliability of necropsy data and the correlation of plastic presence with pathological features, as well as the health impact. A coordinated approach to data collection and standardization is needed to ensure consistent reporting, particularly regarding emerging threats. In this regard, beyond mechanical effects, plastics pose additional risks through chemical pollutants such as PCBs and PFAS (Genov et al., 2019; Sciancalepore et al., 2021) and biological pathways. Plastic surfaces can also adsorb hydrophobic contaminants such as PCBs and PAHs, which may be transferred to cetaceans upon ingestion, potentially disrupting endocrine and immune functions (Fossi et al., 2012; Alomar et al., 2020). Moreover, plastics can host opportunistic or pathogenic microbial communities, the so-called “plastisphere” (Bowley et al., 2021; Du et al., 2022). Although this study did not assess chemical or microbial contamination directly, these pathways represent probable sub-lethal effects, especially under chronic exposure conditions. The application of new tools, such as blow sampling via UAVs (Centelleghé et al., 2020), may support future health assessments in live populations in regard to microplastics (Dziobak et al., 2024). Given that the three countries share transboundary cetacean populations, particularly *T. truncatus*, enhanced cross-border data sharing is essential to obtain reliable information on stranded specimens, contextualized in light of known population estimates. While collaborative initiatives have already begun, such as workshops and joint projects under ACCOBAMS and the MSFD, data inconsistencies continue to hinder comprehensive, large-scale assessments.

A previous study in Italy by the same stranding network (Pietroluongo et al., 2025) reported gillnet ingestion in 29 of 171 cetacean carcasses (2009–2023; FO = 16.3%), a rate higher than plastic litter ingestion. Most cases involved *T. truncatus* (n = 22), followed by *S. coeruleoalba* (n = 4) and *P. macrocephalus* (n = 1). Gillnet ingestion was identified as the cause of death in 5 *T. truncatus*, representing 6.1% of fishery-related mortalities and 0.6% of all analyzed carcasses, similar to the impact from plastic litter ingestion. Fishing gear also caused laryngeal entanglement, often associated with ingestion, in 27 of 82 identified causes of death (32.9%) and 27 of 790 carcasses (3.4%); no such cases were linked to plastic litter. Comparison between the two studies indicates *T. truncatus* is more affected by fishery interactions than plastic litter, while *P. macrocephalus* shows the reverse trend.

In comparison to cetaceans, sea turtles examined in Italy between 2017 and 2021 showed significantly higher rates of marine litter ingestion. Matiddi et al. (2024) reported plastic ingestion in 63.4% of turtle carcasses in Italy (291/459) and 41.2% in the Adriatic subregion, versus 2.9% for all cetaceans in Italy and 5.2% in the Italian Adriatic Sea. Plastic ingestion was the cause of death in 4.12% of turtles (12 cases; 2.61% of all carcasses). Under MSFD Descriptor 10, Good Environmental Status (GES) requires

that marine litter cause no harm, applying the precautionary principle (Commission Decision (EU) 2017/848, Art. 4(1)(e)) and using threshold values low enough to prevent harmful effects (Werner et al., 2020). Matiddi et al. (2024) proposed a GES threshold of <33% of turtles with >0.05 g ingested plastic, in samples of  $\geq 50$  dead individuals per subregion. In the present study, *P. macrocephalus* was the only cetacean with comparable ingestion values and is identified as a suitable sentinel species, as previously proposed by Fossi et al. (2020). Following the MSFD and the results of the present study, the starting point to propose a GES threshold for this species over a rolling six-year period (minimum sample: 7 individuals) corresponds to a baseline value of <60% of examined carcasses with >1 kg ingested plastic and <40% showing health impacts directly attributable to plastic ingestion. Based on these criteria, GES is not met for *P. macrocephalus* in the ADRIA MSFD subregion, but it was for the WMED. However, these values should be considered as preliminary baseline data rather than definitive thresholds, given the limited geographical scope of the sampled individuals (Mediterranean only), which does not allow for generalized MSFD-related threshold setting as in studies based on larger and more diverse datasets (e.g., Matiddi et al. on *Caretta caretta*). Furthermore, data interpretation is limited to Italian data, and biased by the fact that this species is not distributed throughout the study region. In that respect, *T. truncatus* is the only species distributed throughout the study region and would therefore qualify as a better sentinel species in terms of its geographic distribution. However, as noted above, this species appears to be a rather poor proxy for the occurrence of marine debris. Finally, most data on *P. macrocephalus* in this study originated from UMEs, which may not necessarily be representative of this species in the region overall. Thresholds are intended for early detection of critical plastic exposure trends in top marine predators and harmonized GES assessments across species and subregions. Defined as a provisional Mediterranean benchmark, the baseline values of the present study need to be refined as further necropsy data become available, reinforcing the sperm whale's role as a sentinel of macroplastic pollution in deep pelagic ecosystems, particularly given the endangered status of the Mediterranean subpopulation on the IUCN Red List (Pirrotta et al., 2021).

## 4.1 Choosing the right sentinel species for marine litter monitoring

Identifying an appropriate sentinel species for monitoring plastic ingestion in Mediterranean context is challenging, as no single taxon fulfills all the required criteria. *T. truncatus*, although widespread and present across all MSFD subregions in the Mediterranean, as well as most subregions in other parts of Europe, appears relatively invulnerable to plastic ingestion, likely due to its adaptability and foraging strategies, which may underestimate the magnitude of the threat to cetaceans generally. *S. coeruleoalba*, while affected more frequently, abundant and also relatively widespread, also shows relatively low ingestion rates,

limiting its suitability as a proxy. In contrast, *P. macrocephalus* consistently exhibits high %FO values and associated health impacts, suggesting it is a particularly vulnerable species, which may be an informative indicator of plastic-related risks. However, its restricted distribution and the predominance of data derived from UMEs introduce biases that caution against relying on it as the sole sentinel. A combined approach, where *T. truncatus* is monitored for its broad spatial representativeness and *P. macrocephalus* for its ecological susceptibility, may therefore provide a more balanced strategy. Neither of these two species is a perfect sentinel candidate on its own, but combined they may be very informative in guiding further monitoring and management in the future. Moreover, specific species may be considered in specific regions, for example *T. truncatus* in the Adriatic Sea and *P. macrocephalus* in the Tyrrhenian Sea. This approach, and the combined or individual use of these species, may be re-evaluated in the future, as more data become available. Furthermore, other species should be investigated, particularly deep divers such as *Grampus griseus* and *Z. cavirostris*, both with similar behavioral and ecological traits and potential susceptibility to marine litter ingestion, despite their lower abundance and stranding rates. Such a multi-species framework would allow for both geographical coverage and sensitivity to marine litter ingestion, while maintaining flexibility to refine species selection as new evidence emerges from coordinated, long-term monitoring.

## 4.2 Final remarks

To effectively mitigate the impacts of marine litter, simulation models integrated with empirical data can help identify high-risk areas and forecast future trends (Darmon et al., 2017; Fossi et al., 2025). These models also help overcome the limitations of sporadic observations and subjective assessments (Li Veli et al., 2023). Furthermore, strengthening national stranding networks is essential to facilitate the standardized collection and sharing of samples for comprehensive marine litter analyses. This will allow for future comparisons of temporal trends and differences among geographic areas. In addition, specific behaviors propagated through social learning, observed within and across cetacean generations (Rendell and Whitehead, 2001; von Bubnoff, 2005), highlight the need to better understand behavioral drivers of marine litter interactions.

Overall, this study emphasizes the multifactorial threat posed by plastic pollution to cetacean health in the Mediterranean, with particular concern for deep-diving odontocetes such as *P. macrocephalus*. The combination of mechanical obstruction, physiological impairment, potential chemical and microbial exposure, and interaction with other stressors underlines the urgent need for mitigation strategies. Moreover, it reinforces the importance of integrating both lethal and sub-lethal effects of marine litter into ecological risk assessments and conservation planning. Ultimately, progress depends on continuing and expanding transboundary cooperation, harmonizing methodologies, and translating research



findings into actionable conservation policies, in line with the goals of ACCOBAMS and the MSFD.

## Data availability statement

The original contributions presented in the study are included in the article/[Supplementary Material](#). Further inquiries can be directed to the corresponding author.

## Ethics statement

Ethical approval was not required for the study in accordance with the local legislation and institutional requirements because this research consisted solely of post-mortem examinations and sampling of stranded cetacean carcasses. No live animals were handled or subjected to procedures.

## Author contributions

GP: Writing – review & editing, Writing – original draft, Investigation, Visualization, Resources, Methodology, Data curation, Funding acquisition, Supervision, Conceptualization, Project administration, Formal Analysis. CCe: Funding acquisition, Writing – review & editing, Resources, Formal Analysis, Project administration, Writing – original draft, Conceptualization, Data curation, Methodology, Supervision, Investigation. MB: Writing – review & editing, Investigation, Data curation, Methodology. CCo: Writing – review & editing, Investigation. CCa: Writing – review & editing, Investigation, Methodology, Data curation. GC: Formal Analysis, Data curation, Methodology, Investigation, Writing – review & editing. GD: Writing – review & editing, Investigation. FD: Writing – review & editing, Investigation. LD: Writing – review & editing, Investigation. MĐ: Data curation, Validation, Methodology, Writing – review & editing, Investigation, Formal Analysis. MF: Methodology, Data curation, Investigation, Writing – review & editing. SG: Writing – review & editing, Investigation. TG: Investigation, Writing – review & editing. FG: Data curation, Methodology, Investigation, Writing – review & editing. GL: Investigation, Writing – review & editing. IP: Investigation, Writing – review & editing. APe: Investigation, Writing – review & editing. APi: Investigation, Writing – review & editing. RP: Writing – review & editing, Investigation. SR: Investigation, Writing – review & editing. GT: Investigation, Writing – review & editing. CG: Writing – review & editing, Investigation, Methodology, Formal Analysis, Data curation. SM: Resources, Writing – original draft, Project administration, Validation, Writing – review & editing, Conceptualization, Supervision, Funding acquisition, Methodology, Investigation.

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## Conflict of interest

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## Supplementary material

The Supplementary Material for this article can be found online at: <https://www.frontiersin.org/articles/10.3389/fmars.2025.1713820/full#supplementary-material>

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