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Plastic litter transfer from sediments towards marine trophic webs: A case study on holothurians

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ABSTRACT

This study estimates for the very first time plastic litter levels in sea cucumbers (Echinodermata, Holothuroidea) sampled in situ and their intakes from sediments in three different rocky bottom habitats (slides, cliff, banks) settled in Salina Island (Aeolian Archipelago). Macroplastic were never recorded while meso- and microplastics were identified in all sediment (81–438 items/kg d.w.) and animal samples (1.8–22 items/ind.). Plastic intakes by sea cucumbers resulted frequently associated to the size range included within 100–2000 μm. Over than 70% of ingested plastic litter is represented by the size fraction > 500 μm. Sediment/animals ratios % are included 2.7 ± 2.0% in studied habitats with a selective intake of fragments occurring in slides. Furthermore, results support the occurrence of selective ingestion of plastic litter by holothurians in natural environments underlining the role of these species in microplastic transfer from abiotic towards biotic compartments of the marine trophic web.

1. Introduction

Litter transfer from the environment towards marine trophic webs represent an important task that should be better described and clarified by the literature to achieve Horizon 2020 targets concerning the Marine Strategy Framework Directive principal purposes (2008/56/EC). Plastic litter represent the larger part of the marine litter recovered along Mediterranean coasts (over than 50% in Baleari Islands, Martinez-Ribes et al., 2007). According to the literature (Galgani et al., 2013) maximum size of plastic marine litter determine the classification as macroplastic (> 25,000 μm); mesoplastic (5001–25,000 μm), and microplastic (MPs; 63–5000 μm).

Recent studies evidence that plastic litter affects different marine species (Avio et al., 2015; Collard et al., 2017; Dehaut et al., 2016; Fossi et al., 2016) and that it penetrates the marine trophic web (Ivar Do Sul and Costa, 2014; Setala et al., 2014). Cole et al. (2011) in a recent review, reported many different researches documenting microplastic ingestion by marine species such as seabirds, crustaceans and fish. Plastic ingestion could affect the feeding habits, reproductive success, and breathing of many marine organisms (Cole et al., 2014), which ingest plastic litter, such as cetaceans (Tonay et al., 2007), and large pelagic fish species (i.e. *Xiphias gladius*, *Thunnus thynnus* and *Thunnus alalunga*) (Romeo et al., 2015). Also, small pelagic planktivorous fishes, such as sardines, could actively ingest microplastic directly from the water column (Avio et al., 2015) nevertheless, in a laboratory study, only 19% of tested sardines predate floating microplastics and the principal route of ingestion is related to the feeding. On the contrary a large number of tested sardine and anchovy evidenced microplastics in stomach contents almost during the whole year with a clear difference related to their colours (Renzi et al., unpublished data). According to the literature, pelagic species ingested more particles than benthic ones, which, on the contrary, ingested more fibres (Neves et al., 2015). Recently, the presence of MPs in 45% of biota from the Adriatic Sea (Dehaut et al., 2016), and in particular in 95% of the benthic flatfish *Solea solea* (Pellini et al., 2018) were recorded supporting the hypothesis that benthic species could be highly impacted by litter stored in sediments.

Species belonging to the lower trophic level, that are indiscriminate feeders, are not able to differentiate between plastics and prey ingesting high levels of plastic. In vitro studies reported by Cole et al. (2011) evidenced the occurrence of microplastic ingestion by gastropods (7–70 μm), echinoderm larvae (10–20 μm), trochophore larvae (3–10 μm), scallop (16–18 μm), amphipods (20–2000 μm), and mussel (2–16 μm). Also sea cucumbers in vitro ingested microplastic in a wide dimensional range (Graham and Thompson, 2009).

Some authors reported that many marine animals are able to remove ingested materials that could cause harm (Thompson, 2006; Andrady, 2011), including microplastics (Graham and Thompson,

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2009). Nevertheless, ingestion could represent a possible risk as well as a recent research performed in mussels recorded translocation of polystyrene microspheres in mussels' haemolymph up to 48 days after the exposure (Browne et al., 2008). Another research evidenced that in blue mussel, the exposure to microplastic is able to induce immune response and granuloma formation in the digestive glands (Köhler, 2010).

In spite of recent studies, relationship between microplastic content in organisms and environments still remains unclear (Qu et al., 2018). In particular, relationships among plastic litter settled in sediments and benthic species are not jet clarified by the literature.

This research was performed to evaluate plastic litter transfer from sediments towards benthic species. Sea cucumber (Holothurians) was selected as benthic species of specific interest in this study representing a first step from the detritus net towards the trophic web in shallow marine ecosystems. This taxonomic group was selected as it is abundant in different marine habitats, it is represented by sedentary deposit feeding or suspension-feeding species that are indiscriminate feeders, it evidenced ingestion of plastics (Graham and Thompson, 2009) and it is reported to be a key benthic taxonomic group in marine ecosystems (Purcell et al., 2016). Furthermore, it is highly exploited by fishery for human food purposes allowing evaluations related to human exposure levels (Purcell et al., 2016).

2. Materials and methods

2.1. Study area features

The entire Aeolian Islands are inhabited even if islands are subjected to different degree levels of human pressure. This research focuses on Salina Island as it was designated for the institution of a Marine Protected Area (MPA) for the Aeolian Archipelago according to the Italian Law 979/82. At this moment Salina Island is not jet insert in the MPA Italian list.

Salina Island (Aeolian Archipelago) was the study area as this research represents an advancement of a previous published study developed in the Aeolian Archipelago islands by Fastelli et al. (2016). Authors focused on levels of plastic litter in sediments from different Islands of the Aeolian Archipelago (Alicudi, Filicudi, Lipari, Panarea, Stromboli, Vulcano, and Salina); and evaluated levels according to size, shape, and colour differences to define levels in sediments from Islands subjected to different direct local human pressure (Lipari; Panarea ≫ Alicudi; Filicudi; Salina). Fastelli et al. (2016) did not evidenced significant difference concerning plastic litter levels in sediments due to known local pollution sources. The lower MPs levels were found in sediments from Stromboli and Salina Islands while Lipari showed the highest level.

We selected Salina Island as study area on the basis of low pollution levels observed by Fastelli et al. (2016) and the future institution of MPA. According to the geomorphological structure of bottoms closed to Salina Island (Bosman et al., 2013), three different rocky bottom habitats (slides, cliff, banks) were sampled as reported in Fig. 1.

2.2. Principal features of selected species

Sea cucumber (Holothurians) was selected as benthic species of specific interest in this study due to different reasons. First of all holothurians are abundant in selected sampling sites and in all of the selected rocky bottom habitats (sampled species *H. tubulosa*). These animals are wide representative of marine benthic species (Purcell et al., 2016) and evidence a benthic deposit-feeding or suspension-feeding behaviour that is easy to be compared to MPs in sediments. Furthermore, holothurians are key benthic taxonomic group in marine ecosystems to preserve ecosystem integrity (Purcell et al., 2016). A recent in vitro study evidences a specific preference for plastic feeding (Graham and Thompson, 2009). Last but not least, holothurians are linked with the marine trophic web through predation by stars; crustaceans, gastropods, fishes (Francour, 1997; Dance et al., 2003) and they are highly exploited by humans for food purposes (Purcell et al., 2016).

On the basis of the ecological roles of these species in marine ecosystems, the Italian Ministry of Agriculture and Forestry, implemented regulatory measures centred on the Ecosystem-Based Fisheries Management to safeguard sea cucumbers from overexploited. At this date, holothurians are protected species in Italy and target fishes or bycatch are not allowed starting from 21/02/2018 till 31/12/2019 for conservation purposes and for the defence of national resource from overexploitation by fishing (Italian Ministry of Agriculture and Forestry, Law n. 156; 27/02/2018).

2.3. Logic model applied to samplings

Three different rocky bottom habitats (slides, cliff, banks) settled in Salina Island were selected for the evaluation of plastic litter features in sediments. Contextually, sea cucumbers were collected in statistical replicates in each sampling sites closed to sediment collection points to determine plastic levels in animals and their intakes in natural conditions and to perform comparisons to sediment levels and to different rocky bottom habitats considered. The sampling strategy included fixed and random factors a priori selected. Considered factors were: Rocky bottom habitats (three levels, fixed), sampling sites (R1–R2; two levels, fixed), sampling replicates (five levels, random). Overall, 30 samples were collected ($n = 30$) per each considered matrix.

2.4. Sampling activities

Starting from June to September 2017, samplings were performed from different rocky bottom habitats as reported in Fig. 1. In particular were sampled: i) Franata Tre Pietre, and Franata Punta Vallespina, Slides habitat (big blocks/boulders); ii) Secca del Capo, and Secca di Pollara, Banks habitats (open sea conditions); iii) Parete dei Gamberi, and Parete Scoglio Piramide, Cliffs habitats (vertical substrata).

Undisturbed surface sediments (5 cm depth) from Salina Island were collected at −30 m of depth by scientific scuba divers using wide mouth glass jars in statistical replicates following the same sampling strategy and methods of previous research performed in the same area (Fastelli et al., 2016) to favour comparisons even if, in this study, only Salina Island was considered. Every replicates were taken approximately within a radius of 1 m and collected samples were stored frozen at −15 °C until analyses (Galgani et al., 2013; adapted).

H. tubulosa species were collected contextually to sediment sampling in significant replicates per each sampling site and stored separately in glass jars submerged in ethylene to preserve animals until laboratory dissection and analyses. Body size (max length) of animals was standardized during collection to fix animal age and to perform comparison among sampling sites.

2.5. Laboratory analyses

2.5.1. Sediment pretreatment & extraction

Sediment samples were analysed following the extraction and classification method reported by Fastelli et al. (2016) for the Aeolian Archipelago Islands to a better comparison of collected data with data reported previously by the literature. In brief, sediments were dried at 40 °C and sieved throughout 4000–63 μm standard test sieves to determine grain-size. Fractions retrained on 4 mm, 2 mm, and 1 mm test sieves were collected and directly analysed by stereomicroscopy performing the manual sorting of plastic litter. Sediments retained by the 63 μm sieve were extracted by the exposure of saturated NaCl solution following the extraction method reported by the literature (Blašković et al., 2017). The extraction of this sediment fraction was necessary to reduce interferences during microscope analyses.