



Micro(nano)plastics and plastic additives effects in marine annelids: A literature review

Adília Pires^{a,*}, Alessia Cuccaro^a, Montserrat Sole^b, Rosa Freitas^a

^a Centre for Environmental and Marine Studies (CESAM) & Department of Biology, University of Aveiro, 3810-193 Aveiro, Portugal

^b Instituto de Ciencias del Mar ICM-CSIC, E-08003, Barcelona, Spain

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ABSTRACT

Plastic debris are dispersed in the marine environment and are consequently available to many organisms of different trophic levels, including sediment-dwelling organisms such as polychaetae. Plastic degradation generates micro (MPs) and nanoplastics (NPs) and as well as releases bounded plastic additives, increasing the ecotoxicological risk for marine organisms. Therefore, this review summarizes current knowledge on the accumulation and effects of MPs and NPs and plastic additives in polychaetes, derived from laboratory and field evidences. Thirty-six papers (from January 2011 to September 2021) were selected and analysed: about 80% of the selected works were published since 2016, confirming the emerging role of this topic in environmental sciences.

The majority of the analysed manuscripts (68%) were carried out in the laboratory under controlled conditions. These studies showed that polychaetes accumulate and are responsive to this contaminant class, displaying behavioural, physiological, biochemical and immunological alterations. The polychaetes *Hediste diversicolor* and *Arenicola marina* were the most frequent used species to study MPs, NPs and plastic additive effects. The consideration of field studies revealed that MP accumulation was dependent on the plastic type present in the sediments and on the feeding strategy of the species.

Polychaetes are known to play an important role in coastal and estuarine food webs and exposure to MPs, NPs and plastic additives may impair their behavioural, physiological, biochemical and immunological responses. Thus, the estimated global increase of these contaminants in the marine environment could affect the health of these benthic organisms, with consequences at population and ecosystem levels.

1. Introduction

1.1. Micro and nanoplastics

Plastics debris, synthetic organic polymers manufactured from fossil fuels with many uses in everyday life, are currently recognized as among the most persistent contaminants in the environment (Gallo et al., 2018), and thereby an important environmental stressor, posing risks to aquatic life, ecosystems and potentially to human health (Oliveira et al., 2019). Synthetic polymers based on organic materials are cost-effective and easily modelled into different shapes, designed to have attractive properties such as resistance to microorganisms' action, among others (da Costa, 2018). In 2019, global plastic production reached 370 million tons (PlasticsEurope, 2021) as a result of a wide array of applications from packaging to cosmetics and biomedical applications, often consisting of single-use items that are easily discarded (Avio et al., 2017; da Costa, 2018; da Costa et al., 2016; Li et al., 2016).

Due to improper disposal, it becomes a serious problem since plastics frequently end up in aquatic environments, especially in the ocean, where they are not easily degradable, making them a class of emerging pollutants (Rios Mendoza et al., 2018).

There is a wide array of plastic polymers present in the marine environment and the most frequently found are: polyvinyl chloride (PVC), polyethylene (PE), polypropylene (PP), polyethylene terephthalate (PET) and polystyrene (PS) (Gallo et al., 2018). PVC is commonly used for window frames and pipes (PlasticsEurope, 2021); PE is used to make cosmetic bottles, plastic grocery bags and bullet-proof vests (PlasticsEurope, 2021); PP is widely used in everyday items such as utensils, housewares, toys, cars, plastic pallets, sportswear, and even in the lab and medical equipment (PlasticsEurope, 2021); PET is usually employed to make plastic bottles (Li et al., 2021); PS can be found in a variety of products from single-use packaging and plastic cups to eye-glasses frames, to building insulation items (PlasticsEurope, 2021).

* Corresponding author.

E-mail address: adilia@ua.pt (A. Pires).

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Once entering the ocean, plastics can migrate by currents, wind and tides. Due to colonization by microbial communities, adherence by phytoplankton and aggregation with organic debris, plastics may eventually sink and accumulate in sediments. The combined action of physical abrasion (e.g., wave action and sand crumbling), UV radiation and microbial activity result in degradation, embrittlement and plastic fragmentation (Liu et al., 2020), originating smaller particles called microplastics (MPs) (size between 1 and 5 mm) and eventually nanoplastics (NPs). The definition of NPs is not yet agreed, while some authors define NPs as plastic particles with sizes smaller than 1000 nm (Gigault et al., 2018), others as plastic materials having sizes smaller than 100 nm (as defined for other types of nanoparticles) (Lambert and Wagner, 2016; Ter Halle et al., 2017).

Different quantities of MPs in marine environments have already been found from diverse habitats, with concentrations detected in sediments surface varying from 1.5 to 23.4 particles/kg dw (MPs < 1 mm) in beach sediments along the French, Belgian and Dutch coastline (Van Cauwenbergh et al., 2015) to higher concentrations in the Norwegian Continental Shelf (39–3400 particles/kg dw MPs \geq 45 μ m) (Knutsen et al., 2020) and in the Nova Scotia's Eastern Shores, Canada (5000 items/kg dw, 0.063–5 mm) (Mathalon and Hill, 2014).

A wide range of physical and chemical effects of MPs and NPs on benthic organisms, such as changes on survivorship, behaviour, regeneration, oxidative stress and energy reserves have been documented (e.g., Gomiero et al., 2018; Green et al., 2016; Haegerbaeumer et al., 2019; Setälä et al., 2016; Silva et al., 2020a,b). Additionally, the ingestion of MPs, NPs and plastic additives by benthic organisms could result in the bioaccumulation and eventual biomagnification into the aquatic food web, altering its structure and functioning (Lin et al., 2020).

1.2. Plastic additives

Plastic products are made from the essential polymer mixed with additives, which are chemicals added to improve their performance, functionality and ageing properties. A large number of chemicals can be considered plastic additives, and their environmental threat, resulting from its leaching from the polymer and inadequate recycling processes, was extensively addressed by Hahladakis et al. (2018). Their initial regulation in the European Union (EU) No 10/2011 focused mainly on the potential to come into contact with food ((CEC, 2011) or children due to their inclusion in many products, such as toys. Among additives, functional additives such as flame retardants, biocides, stabilizers and plasticizers are considered as more relevant in this review on polychaete studies. Since most of these additives may be present at high concentrations in plastic products, and are not covalently bound to the polymers, during plastic deterioration they can be easily released into the environment (Gunaalan et al., 2020; Tyler et al., 2019). Several recent reviews on plastic additives, which also included marine species, have addressed the environmental problem of the toxicity induced by these compounds, usually associated to plastic products (Avio et al., 2017; Gunaalan et al., 2020; Hermabessiere et al., 2017; Tyler et al., 2019).

Flame retardants and, in particular, polybrominated diphenyl ethers (PBDEs) have been banned in the European Union since 2004, under the Existing Substances Regulation 793/93/EEC, because of their ubiquitous presence, toxicity, persistence and bioaccumulation properties (Gunaalan et al., 2020). Due to their lipophilic characteristics, they could be stored in sediment (Bollmann et al., 2012) and pose a threat to sediment-dwelling biota such as polychaetes. Among the different formulations, penta- and octa-BDEs are of greater environmental concern and have been replaced by others such as tetrabromo bisphenol A (TBBPA) and, more recently, by organophosphorus flame retardants (Gunaalan et al., 2020).

Biocides can also be included as additives since they confer antimicrobial properties to the plastic. Among them, triclosan (TCS) is one of

the most employed in industrial and household products. It is a suspected lipid and endocrine disruptor and therefore of environmental relevance (Gunaalan et al., 2020).

Stabilizers such as the surfactant and antioxidant by-product nonylphenol (NP) and its metabolite octylphenol (OP) are the final degradation product of non-ionic detergents such as alkylphenols (Andrady and Rajapakse, 2019). Due to NP-associated endocrine disrupting properties, these agents are under European Union's REACH regulation.

Bisphenol A (BPA), main monomer used in polycarbonate (PC) plastics (65% of volume used) and epoxy resins (30% of volume used), and phthalates constitute the largest group of additives or plasticizers (Andrady and Rajapakse, 2019). Their main environmental interest is related to their reported endocrine disrupting properties for which they are constantly under survey by REACH authorities and under The Waste Framework Directive, 2014/955/EU. Among phthalates, DEHP constitutes up to 80% of the PVC polymer. Dibutyl phthalate (DBP) is another commonly used plasticizer. From a quantitative perspective, plasticizers represent 10–70% in weight of the polymer, flame retardants 3–25%, stabilizers < 3% and biocides < 1% (Andrady and Rajapakse, 2019). Overall, plastic additives have been detected worldwide in estuarine and marine waters and sediments. The phthalate DEHP and flame retardants PBDEs were the most commonly reported plastic additives in the environment, being found at concentrations ranging from 0.52 to 5.3 ng/L (Xie et al., 2005) to 64,300 ng/L (Tan et al., 1995) for DEHP and from 0.002 to 0.082 ng/L (Wurl et al., 2006) to 4.2–19 ng/L for PBDE (Sánchez-Avila et al., 2012) in marine water. In sediments, DEHP were detected ranging from 170 to 3300 μ g/kg dry weight (dw), in North Sea (Klamer et al., 2005) and PBDEs were detected ranging from 0.2 to 1650 μ g/kg dw (Verslycke et al., 2005), being BDE-209 the most abundant PBDE quantified. In addition, BPA and NP are also frequently detected in seawater and sediments. NP concentrations in marine waters ranged from 0.09 to 1.4 ng/L (Xie et al., 2005) to 4100 ng/L (Petrovic et al., 2002) and from 0.9 μ g/kg dw (Jonkers and Laane, 2003) to more than 20,000 μ g/kg dw in the sediments (Kurihara et al., 2007). BPA, concentrations ranged from < 1 ng/L (Pojana et al., 2007) to 2.47 μ g/L (Basheer et al., 2004) in coastal waters and from 1 μ g/kg dw to 180 μ g/kg dw in marine sediments (Pojana et al., 2007).

1.3. Polychaetes

Polychaetes are the most abundant group of invertebrates in estuarine ecosystems (Rodrigues et al., 2011), being also key elements in the estuarine and coastal food webs, and the primary food source for many fish, crustaceans and birds, making them important vectors for the transfer of contaminants to higher trophic levels (Lewis and Watson, 2012; Serrano et al., 2003). Moreover, as polychaetes are in intimate contact with sediments, they are also particularly exposed to harmful materials both in the sediments and associated pore water (Banta and Andersen, 2003; Scaps, 2002). Polychaetes are considered good models in toxicity studies, being regarded as important sentinel species in contaminated sediment assessment (Dean, 2008).

The environmental impacts of MPs, NPs and plastic additives in aquatic systems have been the object of several studies throughout the last few years including also polychaetes (e.g., Avio et al., 2017; Brandts et al., 2018; Green et al., 2016; Haegerbaeumer et al., 2019; Oliveira et al., 2019; Revel et al., 2018; Soares et al., 2021). Several of these former studies reported their effects on polychaetes performance (e.g., Besseling et al., 2013; Browne et al., 2013; Silva et al., 2020a,b) in which oxidative stress and energy reserves are the most commonly investigated endpoints.

Some of these former studies also demonstrated a systematic impairment in behaviour. Behaviour is considered as a very relevant ecological endpoint, since its alteration at lower levels of biological organization may have an impact on animal's fitness and survival (Besseling et

al., 2013; Gebhardt and Forster, 2018; Green et al., 2016; Muller-karanassos et al., 2021; Silva et al., 2020b; Wright et al., 2013). It is already recognized that polychaetes play a key role in their habitat by providing protection, refuge and food to other species, including other marine invertebrates. Thus, harmful interference in their life cycle will probably have negative consequences to a broader range of species not directly associated with them.

2. Materials and methods

2.1. Literature search parameters

In the present work, the systematic literature review was conducted examining peer-reviewed scientific articles published between 2010 and 2021. The process of identifying relevant bibliographic data was performed using Scopus and Web of Science (WoS) search databases, and all studies found in online databases until September 2021 were included. Articles were retrieved using the following combination of terms as search inputs: Polychaeta AND plastic(s); Polychaeta AND microplastic(s); Polychaeta AND nanoplastic(s); Polychaeta AND plastic additive(s)/plasticizer (s).

2.2. Screening process

The title and abstract of each article found during the searches were evaluated according to the following criteria:

- i) if plastic particles and plastic additives were linked to biological effects on polychaetes (field and laboratory studies)
- ii) if microplastics, nanoplastics and plastic additives uptake was observed or quantified in polychaetes (field and laboratory studies)
- iii) studies with microplastics or nanoplastics or plastics additives and other contaminant types were also considered.

The selected papers were read, and appropriate information and data for this review were extracted. The obtained information is summarised in Tables 1 and 2. Selected papers were categorized according to: year of publication and laboratory experiments (Table 1) or field studies (Table 2). In the studies related to laboratory experiments, it was considered types, shapes and sizes of plastics and plastic additive classes, species used, concentrations adopted, exposure time and effects on polychaetes. In the field studies, it was highlighted the species, types, shapes and sizes of plastics and plastic additives, bioaccumulation, concentration in water and/or sediment and effects in polychaetes, if provided.

2.3. Data analysis and visualization by VOSviewer software

After the screening process, documents that met the former requirements were exported from databases into specific file formats (CSV for Scopus and Tab-delimited for WoS) for further interpretation and visualization by bibliometric analysis.

The bibliometric retrieved files contained bibliographical information (i.e., authors, title, year and source), authors' abstract and keywords, as well as counts of citations, allowing to determine research trends and topics of major interest by specific bibliometric software. In this review, VOSviewer (1.6.11) software (Van Eck and Waltman, 2010) was employed for data visualization by science mapping analysis.

Briefly, files were imported into VOSviewer and downloaded information was extracted. Based on the type of data (bibliographic or text ones), the software was able to create two different networks (co-occurrence links between keywords and terms, respectively) and then, generate the corresponding maps. The different types of links were cho-

sen to follow the evolution of the “plastic issue” over the selected time frame: the high-frequency keyword analysis revealed scientific research trends, while the co-term analysis highlighted the hot-topics investigated by researchers.

The corresponding maps were represented by two ways of visualization: I) the network visualization mapped link between terms, due to its immediate overview on relatedness intra- and inter-groups; II) the overlay visualization defined links between keywords, because of its additional information about research trends following the average publication year of articles in which a keyword occurred. In both maps, items were presented as circles, and their sizes depend on the weight attributed: i) *Occurrences weight* for keywords, which corresponds to the number of documents in which an item occurred; ii) *Total link strength weight* for terms, which indicates the total strength of the links of an item with others. Lines between items represented links: the closer two items were located to each other the stronger their relatedness.

3. Laboratory based studies on polychaetes

3.1. Micro and nanoplastic exposures

The negative impacts of MPs and NPs on polychaetes were evaluated by several authors, resulting in fourteen published manuscripts (Fig. 1). They included short and long-term laboratory assays on the impacts of PVC, PS, PP, Antifouling Paint Particles (APPs) and MPs mixtures in different polychaetes being *Arenicola marina* and *Hediste diversicolor* the most employed species. The concentrations tested varied from relevant to unrealistic (from 0 to 7.4% dw (Besseling et al., 2013) and 0–2000 part/kg on sediments (Gomiero et al., 2018); 5–1000 part/mL (Leung and Chan, 2018; Setälä et al., 2016) and from 0 to 50 mg/mL at water (Silva et al., 2020b). MPs were evaluated individually, as mixtures and also as mixtures of MPs with other contaminants (Table 1). The effects were mainly related to survivorship, growth and behaviour (feeding and burrowing activity).

3.1.1. Exposure to PVC

Four papers on the effects of PVC in polychaetes are addressed in this review (Table 1).

Green et al. (2016) evaluated the effects of PVC on the survivorship, biomass, behaviour and respiration rate of the lugworm *A. marina* after 31 days outdoor mesocosm experiment at an increasing range of concentrations (0.02, 0.2 and 2% of wet weight sediment). The obtained results demonstrated that, although survivorship and biomass were not affected, significantly fewer casts were produced by those exposed. Metabolic rates were increased under the presence of MPs, with significantly higher O₂ consumption at those at 2% of PVC, which was justified as a stress response (Green et al., 2016).

Similarly, Wright et al. (2013) exposed acutely (48 h) and chronically (4 weeks) the same species at laboratory mesocosms conditions to sediments containing clean and chemically-inert PVC (0–5% dw sediment). Worms acutely exposed significantly prolonged gut residence time at 5% of unplasticized polyvinyl chloride (UPVC), while those chronically exposed displayed significantly reduced feeding activity. The total available energy reserves in 1% and 5% UPVC exposed worms were significantly reduced with a maximal 50% depletion to 5% UPVC. The authors justified this response as a result of reduced feeding activity, longer gut residence times and inflammation. Chronic UPVC exposure also significantly increased the phagocytic activity of *A. marina* immune cells, indicative of a metabolically demanding inflammatory response.

Biological effects resulting from the exposure to PVC were also investigated in the polychaete *H. diversicolor* (Gomiero et al., 2018). Experiments lasted 10 and 28 days and sediments were spiked only with pristine PVC at 200 and 2000 particles/kg of sediment. Exposed polychaetes showed induction of immune responses and the suppression of

Table 1
Endpoints of the Micro, nanoplastics and plastic additives in polychaetes exposed under laboratory conditions.

Reference	Species	Contaminant characteristics	Matrix	time	Endpoints
Klosterhaus et al. (2011)	<i>Nereis virens</i>	PBDEs and PCBs (0–3000 ng/g dw)	Sediment	28 d	Bioaccumulation Survivorship Energy reserves (lipids) Biomass
Perron et al., 2012 Besseling et al. (2013)	<i>Nereis virens</i> <i>Arenicola marina</i>	Triclosan (80–800 mg/kg dw) PS Fragments 400–1300 µm (0.074–7.4% sed.dw) PS + PCBs (1.84 ± 0.22 µg/kg)	Sediment Sediment	28 d	Bioaccumulation Survivorship Feeding activity Egestion (faeces) Development (Biomass) PCBs bioaccumulation
Browne et al. (2013)	<i>Arenicola marina</i>	PVC (5%) microspheres 230 µm PVC + phenantrene (0.11 ± 0.01 µg/g) PVC + triclosan (57.30 ± 6.01 µg/g) PVC + PBDE-47 (9.49 ± 1.94 µg/g) PVC + nonylphenol (0.69 ± 0.01 µg/g) Nonylphenol (0.69 ± 0.01 µg/g)	Sediment	11 d	Survivorship Feeding activity Immunotoxicity Oxidative stress biomarkers
Wright et al. (2013)	<i>Arenicola marina</i>	PVC fragments 130 µm (0 and 5% d.w)	Sediment Sediment	2 d 28 d	Egestion (faeces) Energy reserves Feeding activity Immunotoxicity Bioaccumulation
Koelmans et al., 2014	<i>Arenicola marina</i>	Bisphenol A + nonylphenol	Biodynamic model based on previous studies		
Shin et al. (2014)	<i>Capitella sp. I</i>	BDE-47 (0,5 - 3 ppb)	Water	24 h 28 d	Larval settlement Growth
Zhao et al., 2014	<i>Perinereis aibuhitensis</i>	Bisphenol A (10, 50 and 100 µg/L)	Water	4, 7, 14 d	Expression of the Pa G gene
Van Cauwenberghé et al., 2015	<i>Arenicola marina</i>	PS microspheres 10, 30 µm (50 part/g) 90 µm (10 part/g)	Sediment	14 d	egestion (faeces) Bioaccumulation Energy reserves Respiration (energy consumption)
Díaz-Jaramillo et al., 2016	<i>Laeonereis acuta</i>	BDE-47 (0,17–410 ng/g dw)	Sediment	14 d	Bioaccumulation Oxidative stress biomarkers
Green et al. (2016)	<i>Arenicola marina</i>	PVC fragments 8,7–478 µm (0.02–2% ww) HDPE fragments 2,5–316 µm (0.02–2% ww) PLA fragments 1.4–107 µm (0.02–2% ww)	Sediment	31 d	Survivorship Biomass Behaviour Respiration (O2 consumption)
Setälä et al. (2016)	<i>Marenzelleria spp</i>	PS (fluorescent) microspheres 10 µm (5, 50 and 250 beads/mL)	Water	24 h	Ingestion
Lu et al. (2017)	<i>Galeolaria caespitosa</i>	Dibutyl phthalate (0.02–20 mg/L)	Water	15 min- 2.5 h	Embriotoxicity
Gebhardt and Forster (2018)	<i>Arenicola marina</i>	PS (20 g) + PA (10 g) fragments	Sediment surface	106–240 d	Sediments bioturbation
Gomiero et al. (2018)	<i>Hediste diversicolor</i>	1000, 500 µm PVC fragments 250 µm (200 and 2000 part/kg) PVC + B[a]P: 1 mg/L	Sediment	10 and 28 d	Accumulation Biomass Survivorship Immunotoxicity Oxidative stress biomarkers Genotoxicity B[a]P accumulation
Leung and Chan (2018)	<i>Perinereis aibuhitensis</i>	PS microspheres 8–12 µm and 32–38 µm (100 and 1000 part/ml)	Water	28 d	Survivorship Regeneration
Revel et al. (2018)	<i>Hediste diversicolor</i>	PE + PP Fragments 0.4–400 µm (10 and 100 µg/L and 10 and 50 mg/kg)	Water (10, 100 µg/L) Sediment (10, 50 mg/kg)	96 h 96 h and 10 d	Bioaccumulation Mucus production
Silva et al., 2020b	<i>Hediste diversicolor</i>	PS nanospheres 100 nm (0.005–50 mg/mL)	Water	28 d	Immunotoxicity Behaviour Neurotoxicity Oxidative stress biomarkers Energy reserves Metabolic activity

(continued on next page)

Table 1 (continued)

Reference	Species	Contaminant characteristics	Matrix	time	Endpoints
Silva et al., 2020a	<i>Hediste diversicolor</i>	PS nanospheres 100 nm (0.005–50 mg/mL)	Water	28 d	Regeneration
Missawi et al. (2021)	<i>Hediste diversicolor</i>	PE, PEVA, LDPE, HDPE, PP, PA particles from environment 5 mm–1 mm and 1 mm–300 µm (0, 10 and 100 mg/kg)	Sediment	1, 3, 7 and 14 d	Survivorship Growth Neurotoxicity Oxidative stress biomarkers
Muller-karanassos et al., 2021	<i>Hediste diversicolor</i>	APP particles (historic, modern biocidal APPs, non-biocidal silicone APPs) 0.0625–1 mm (0–18.8 g/L)	Sediment	5 and 18 d	LC50 and EC50 Feeding activity Biomass Behaviour Metallothionein-like protein

mitochondrial activity was directly correlated to concentration and exposure time. No consequences were seen in oxyradical production, lysosomal membrane stability and DNA damage. These authors also included the effects of PVC particles pre-incubated with benzo[a]pyrene (B[a]P) and observed higher impacts on those under co-exposure, indicating that MPs particles may enhance toxicity and play a role as vectors for organic pollutants.

Also, Browne et al. (2013) studied the effects in lugworms of sand containing 5% PVC microspheres previously presorbed with nonylphenol, phenanthrene, triclosan and PBDE-47. These authors claimed that MPs contributed to the transfer of these pollutants into the gut tissues of *A. marina*, with its associated biological effects. They also demonstrated that clean sand presorbed with these 4 chemicals transferred higher concentrations of pollutants into polychaete tissues than sand with 5% PVC microspheres alone. The uptake of nonylphenol from PVC or sand reduced the ability of coelomocytes to remove pathogen bacteria by more than 60%. The uptake of triclosan from PVC diminished the ability of worms to act as sediment engineers causing mortality (>55%), while PVC alone made worms more susceptible to oxidative stress (>30%).

One of these former studies revealed physiological and behavioural impairments, including lower egestion (faeces production), after acute exposures (0 and 5% dw) while chronic assays at the same concentrations decreased feeding activity in *A. marina* (Wright et al., 2013). This chronic effect may imply lower remobilization and oxygenation of the associated sediment (Wright et al., 2013). In other studies, the decrease of feeding activity also led to weight loss and reduction of total energy reserves, impacting polychaetes growth and health (Browne et al., 2013; Green et al., 2016). Moreover, chronic assays (0–5% and 200 to 2000 part/kg) demonstrated that exposure to PVC induced immune responses and enhanced metabolism in *A. marina* and *H. diversicolor* (Gomiero et al., 2018). Additionally, the studied papers showed that MPs may also act as vectors for organic pollutants.

3.1.2. Exposure to PS

Six papers studying the impact of PS in polychaetes, two with NPs and four with MPs of different sizes, were considered.

Oxidative stress, neurotoxicity, impacts on metabolism and burrowing activity were evaluated by Silva et al., 2020b after exposing *H. diversicolor* for 28 days to different concentrations of polystyrene NPs (PS NPs) (0, 0.005, 0.05, 0.5, 5 and 50 mg/L). At higher exposures, the activity of the enzyme superoxide dismutase (SOD) increased, while catalase (CAT) and glutathione S-transferases (GSTs) were inhibited, which lead to enhanced protein carbonylation although lipid peroxidation levels remained similar. Furthermore, an increase of the metabolic capacity of polychaetes, due to PS, was associated to the activation of the antioxidant defence system while their burrowing capacity was compromised. PS NPs tended to aggregate alongside time and concentration

(Silva et al., 2020b), yielding higher particle sizes that, may be less bioavailable (Brandts et al., 2018); which justified the lack of behavioural alterations at higher concentrations.

Silva et al., 2020a further demonstrated that after PS NPs exposure to 10-times lower each of the former concentrations (0.0, 0.0005, 0.005, 0.05, 0.5 and 5.0 mg/L) significantly decreased *H. diversicolor* regenerative capacity, with impacts already noticed at the lowest concentration.

Leung and Chan (2018) tested if particle size and concentration (100 and 1000 part/mL) affected the regeneration rate of *Perinereis aibuhitensis* exposed to PS for four weeks. Their results revealed that PS MPs increased mortality and reduced the posterior segment regeneration rate. The size-related impact confirmed that smaller beads were more detrimental than bigger ones. The authors justified these results by a preference to ingest smaller particles in deposit feeding polychaetes.

Besseling et al. (2013) investigated the effects of PS MPs on survival, feeding activity and body weight in *A. marina*. A positive relationship was observed between MPs concentration in the sediment alongside with the uptake of plastic particles and weight loss by the worms as result of a reduced feeding activity at the highest dose (7.4% dw). Besides the effects of PS on survival and body weight of *A. marina*, these authors further evaluated the transfer of 19 polychlorinated biphenyls (PCBs). A PS dose as low as 0.074% was responsible for increasing the bioaccumulation of PCBs while at higher PS doses (7.4%) PCBs bioaccumulation decreased (Besseling et al., 2013).

Van Cauwenberghe et al. (2015) exposed *A. marina* to a mixture of PS spheres of three different sizes (10, 30 and 90 µm) at a concentration of 110 MP/g of sediment for 14 days. Despite these high unrealistic concentrations, no significant adverse effects were seen on the organisms' overall energy budget and it was justified by the authors by the fact of being a short-term trial.

Setälä et al. (2016) conducted a mesocosm experiment designed to evaluate the ingestion of 10 µm PS beads (final concentration: 5, 50 and 250 beads/mL) in the polychaete *Marenzelleria* spp. In this study, the number of ingested beads by worms increased alongside concentration.

All these six former studies were carried out with bioturbator species (three with *H. diversicolor*, two with *A. marina* and one with *P. aibuhitensis*), and the majority of them (four) included parameters related to behaviour (burrowing, feeding activity and casts production) (Besseling et al., 2013; Leung and Chan, 2018; Setälä et al., 2016; ; Silva et al., 2020b; Van Cauwenberghe et al., 2015). These studies demonstrated that the effects of the exposure to PS MPs and NPs may depend on MPs size. In fact, it seems that small particles (100 nm and 8–12 µm in diameter) were responsible for higher impacts on polychaetes regenerative capacity, behaviour and oxidative status, which could constrain

Table 2

Microplastics and Plastic additives concentrations in sediment and water samples and polychaetes from marine environments. PA – polyamide, PAM - polyacrylamide, PAN - polyacrylonitrile, PBT - Polybutylene Terephthalate, PE – polyethylene, HDPE - high-density polyethylene, LDPE - low-density polyethylene, PET - polyethylene terephthalate, PEVA - polyethylene vinyl acetate, PP - polypropylene, PS - Polystyrene, PU – Polyurethane, PVC - polyvinyl chloride, S – sediment, P – Polychaetes, W - Water, NA – not analysed, ww – wet weight, dw – dry weight.

Reference	Species	Contaminant characteristics	Concentration in organism	Concentration in sediment (S)/water (W)	Methodology (S, W, P)	Location
Nurulnadia et al., 2014	<i>Paraprionospio</i> sp.	Nonylphenol	1460–4410 ng/g	11.3–36.5 ng/g ww (S)	LC-MS/MS	Japan
Van Cauwenberghe et al., 2015	<i>Arenicola marina</i>	Bisphenol A	22.5–39.6 ng/g	0.98–2.64 ng/g ww (S)	69% nitric acid (P) NaI(S) Microscope Raman spectrometer	French, Dutch Belgian
		Octylphenol	18.9–45.4 ng/g	2.72–5.36 ng/g ww (S)		
Gusmão et al. (2016)	<i>Saccocirrus</i> sp.	PE, HDPE and PS microparticles	1.2 ± 2.8 particles/g tissue	1.5 to 23.4 particles/kg dw (S)	Stereo microscope	Brazil, Italy Canary Islands Portugal,
		15–100 µm (tissue)	0.3 ± 0.6 part/g tissue (faeces)	0.4 ± 0.3 particles/L (W)		
Lourenço et al. (2017)	<i>Hediste diversicolor</i>	PAN; PET; Other plastics (PE, PP, nylon and PS)	2.7 ± 1.64 microfibers/ind <i>H. diversicolor</i>	3.1 ± 2.01 to 8.0 ± 2.41 microfiber/mL (S)	Enzyme neutrastase (P)S tereo microscope µ-FTIR	Mauritania
	<i>Diopatra neapolitana</i>	Microfibers	1.0 ± 0.82 microfibers/ind <i>D. neapolitana</i>			
	<i>Glycera alba</i>		3 microfibers/ind <i>G. alba</i>			
	<i>Nereis caudatus</i>		0.5 ± 0.97 microfibers/ind <i>N. caudatus</i>			
Bour et al. (2018)	<i>Hediste diversicolor</i>	PP, PE, polyester, polyacrylic, PBT, copolymer	40% contained at least one Mp	-	KOH (P), NaCl Stereomicroscope µ-FTIR	Norway
	<i>Sabella pavonina</i>	Fragments, flakes, fibres, <100 µm to >1 mm	-	-		
Cuvillier-Hot et al., 2018	<i>Hediste diversicolor</i>	PP, PE Fragments, flakes <100 µm to >1 mm	-	-	GC-MS	France
		Phthalates (DEHP >95%) and trace metals (Cd, Co, Ni, Cu, Pb, Zn, Mn, Fe)	-	3.47–23.28 DEHP µg/g 3.63–23.88 total µg/g		
Naidu et al. (2018)	<i>Sternaspis scutata</i> <i>Magelona cincta</i>	PS particles and fibres	-	-	-	India
Nel and Froneman, 2018	<i>Gunnarea gaimardi</i>	Synthetic polymers (not identified)	0.056 to 1.113 microplastic particles/g dw	-	NaCl, 100 g.L ⁻¹ Stereo microscope	South Africa
Md Amin et al., 2020	NA	Polyamide fibers (625 µm)	0.007 particle/ind	3.3 particles/L (W)	HNO ₃ (P). Light microscope observation	Malaysia
Knutsen et al. (2020)	<i>Oweniidae</i> family	PE-chlorinated, PVC, PAM, PE, PE:PP, PET Phenoxy resins and nylon (≥45 µm)	11 to 880 particles/g ww 390 to 1400 particles/g ww (tube)	39 to 3400 particles/kg dw (S)	NaOH (S and P) FTIR	Norway
Missawi et al. (2020)	<i>Hediste diversicolor</i>	PE, PEVA, LDPE, HDPE, PP, PA (3 µm–0.22 µm)	0.5–3.7 items/g	129 to 606 items/kg (S)	ZnCl ₂ solution (S). KOH (P) FTIR	Tunisia
Costa et al. (2021)	<i>Phragmatopoma caudata</i>	PE, PP, PET	blue MPs 81.29% (tissue)	-	KOH (P) Raman spectroscopy	Brazil
		Filaments and fragments shape	56.37% blue filaments (tube) 16.54% colorless filaments (tube) 13.87% blue fragment (tube)			
Hamzah et al. (2021)	<i>Namalycastis</i> sp.	PP, PA Filaments (99.79%) and fragments (0.21%)	20 to 46.79 Mps/ind	-	NaOH 10 M (P). µ-FTIR	Malaysia
Pequeno et al. (2021)	<i>Marphysa sanguinea</i>	PET (83%), PVC (17%) fibres and fragments	0.06 ± 0.25 fibers/ind	-	KOH 10%. Stereoscopic microscope FTIR	Portugal
		73–822 µm	0.33 ± 0.84 fragments/ind			
Vermeiren et al. (2021)	<i>Euzonus</i>	PE and PP (52.4%), PET (12.7%), PVC (11.1%),	0.2–1.0 fibres/mg	8.5–847.5 plastics/L (S)	Fenton's reagent (S) ZnCl ₂ (S) Nile Red dye (S,P)	Uruguay

(continued on next page)

Table 2 (continued)

Reference	Species	Contaminant characteristics	Concentration in organism	Concentration in sediment (S)/water (W)	Methodology (S, W, P)	Location
	(Thoracophelia)	Acryl (6.3%), PS (4.8%), PU (3.2%),	0.4–0.8 particles/mg		μFT-IR (S) Fenton's reagent + hydrogen 30% peroxide(P)	
	<i>furcifera</i>	Polysiloxane (1.6%) fibres and particles 484 ± 234 μm fibers, 5 ± 30 μm particles				

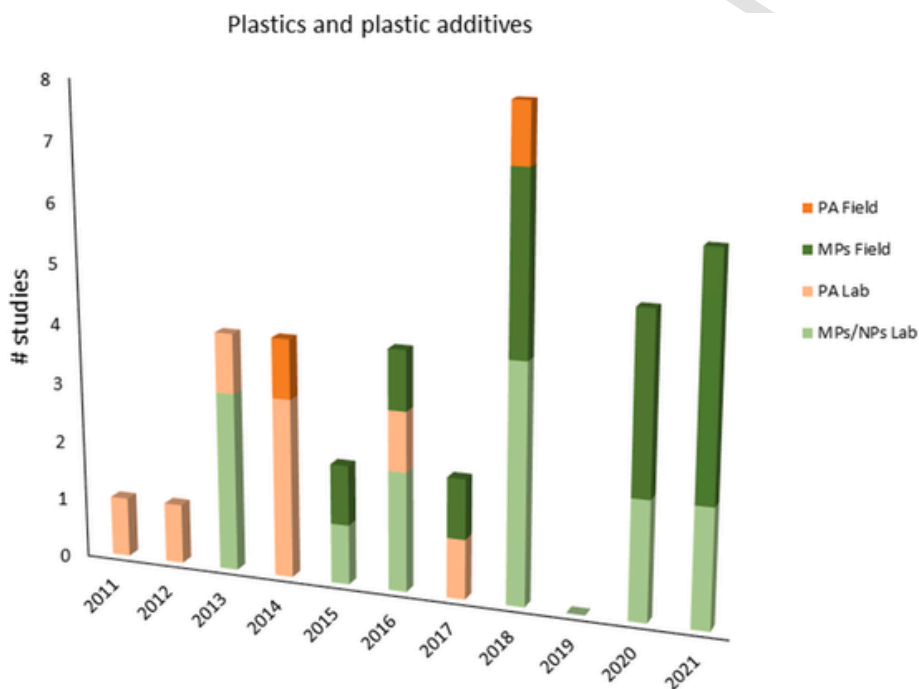


Fig. 1. Timeline of the number of laboratorial and field studies with Micro/Nanoplastics and Plastics additives on polychaetes from 2011 to September 2021. MPs – Microplastics; NPs, Nanoplastics; PA – Plastic additives; Lab – Laboratorial studies.

the normal growth, reproduction or defence capacity of polychaetes under adverse environmental conditions (Leung and Chan, 2018; Silva et al., 2020a). Exposure of large MPs (400–1300 μm) decreased feeding activity (7.4%) of *A. marina* at higher concentrations and caused weight loss, which may also impact polychaetes growth and health.

3.1.3. Exposure to high-density polyethylene (HDPE) and polylactic acid (PLA)

Green et al. (2016) also investigated the impacts of HDPE and PLA in *A. marina* and, as in their formerly described PCV study, 31 days of exposure to these MPs had no impact on worms' survivorship, biomass and behaviour, although respiration rate was affected by HDPE at higher MPs concentrations.

3.1.4. Exposure to antifouling paint particles (APPs)

The study carried out by Muller-karanassos et al. (2021) evaluated the acute (5 days) and chronic (18 days) toxicity of three types of APPs (modern, historic and silicone) on *H. diversicolor*, at environmentally relevant concentrations (0–18.8 g/L). Their results revealed that modern APPs had a higher impact than historic and silicone APPs, causing a decrease in their body weight, feeding activity, and burrowing behaviour. The authors justified their results by the fact that modern APPs contained high Cu concentrations (about 2.6 times) higher than historical ones, representing an additional threat already at environmentally relevant concentrations, while silicone APPs were the least toxic. Additional analyses in *H. diversicolor* tissues confirmed that APP ingestion was correlated with Cu accumulation (Muller-karanassos et al., 2021).

3.1.5. Exposure to mixtures of MPs

3.1.5.1. PS and PA. Gebhardt and Forster (2018) conducted a long-term mesocosm experiment (106–240 days) on the impact of MPs transport in sediments and accumulation of two sized MP particle types on *A. marina* bioturbation. No effects were due to incubation time, ingestion of contaminated sediment or interaction of these two factors on polychaetes' biomass. Nonetheless, the bioturbation activity of *A. marina* could contribute to the transport of MPs larger than 500 μm and favour their longer retention time in marine sediments.

3.1.5.2. PE and PP. Revel et al. (2018) evaluated the accumulation of MPs in *H. diversicolor* exposed to either sediment or waterborne of PE and PP mixture. For both types of MPs, two concentrations were selected, resembling medium and heavily polluted water (10 and 100 μg of MPs/L) and sediment compartments (10 and 50 mg of MPs/kg). In worms exposed through sediment for 96 h, the average number of PP particles ingested was lower than in waterborne exposed ones, due to the absence of secreted mucus on those sediment-exposed. MPs caused an impact on the immune system by reducing coelomocytes viability but they did not alter phagocytic, phenoloxdase and acid phosphatase activities in sediment exposed worms after 10 days.

3.1.5.3. PP, PE, HDPE, low-density polyethylene (LDPE), polyethylene vinyl acetate (PEVA) and polyamide (PA). Missawi et al. (2021) exposed via sediment the polychaete *H. diversicolor* to a mixture of PE, PEVA, LDPE, HDPE, PP and PA for 1, 3, 7 and 14 days. Results confirmed that worms accumulated them. Low doses and short-term exposures had no

impact on survival, growth and weight gain, but higher MPs concentration and longer exposure time, had an impact on survival and growth as well as affected neurotransmission and antioxidant pathways in a concentration- and time-dependent way.

The three above mentioned studies with MP mixtures were carried out with bioturbator species (two with *H. diversicolor* and one with *A. marina*). They revealed that effects on polychaetes increased alongside concentration and time although *H. diversicolor* seemed the most affected species.

3.2. Plastic additive exposures

Evaluation of plastic additives on polychaetes was assessed by few studies, comprising eight manuscripts from 2011 to 2017 (Fig. 1). Literature includes short and long-term laboratory assays, addressing mostly the impacts of flame retardants, biocides and plasticizers individually and/or as mixtures. Survivorship, growth and behaviour (feeding and burrowing activity) were the most selected endpoints (Browne et al., 2013; Klosterhaus et al., 2011; Shin et al., 2014).

3.2.1. Exposure to flame retardants

The bioaccumulation and toxicity associated to the flame retardants polybrominated diphenyl ethers (PBDEs), including the particular congener PCB 209, was considered in *Nereis virens* using contaminated sediment either from a field site or spiked under laboratory-controlled conditions with deca-BDE, penta-BDE and PCB 209 (Klosterhaus et al., 2011). The sediment exposure (from 0 to 3000 ng/g dw depending on the congener) lasted 28 days and bioaccumulation, survival, energy reserves (lipids) and biomass were the targeted endpoints. The results revealed that in both field-collected and laboratory-spiked sediments, worms selectively accumulated congeners in the penta-BDE mixture over BDE 209 and deca-BDEs, supporting the transfer role of polychaetes to species of higher trophic level. Survival ranged from 90% (control), 88% (sediment-spiked) to 83% in field specimens, with no impact on lipid reserves over time, or between control and field specimens. Growth was lower in sediment-spiked and field groups likely due to their lower ingestion rates (Klosterhaus et al., 2011).

The influence of a particular PBDE congener on larval settlement of *Capitella* sp. I, was tested under normoxia and hypoxia conditions for 24 h with spiked sediment at 0.5 and 3 ppb BDE-47 (Shin et al., 2014). Only the interactive effects of hypoxia and environmentally realistic concentrations of BDE-47 could compromise their settlement. The same congener, BDE-47, was targeted in adults of *Laeonereis acuta* exposed to contaminated sediment (0.17–410 ng/g dw) for 14 days and the effects on bioaccumulation and oxidative stress were investigated (Díaz-Jaramillo et al., 2016). Bioaccumulation trends revealed a biota sediment accumulation factor (BSAF) greater than 2 and GSTs activity increased over time and concentration although lipid peroxidation (by measuring the formation of thiobarbituric acid reactive substances) or total antioxidant capacity were not affected. The same study revealed that PBDE hydroxylation is one of the main biotransformation pathways of BDE-47 in *L. acuta*.

The same former PBDE congener and other plastic additives combined with the polymer PVC were considered in a study with the lugworm *A. marina*. Microspheres of 230 µm containing BDE-47 at 9.49 ± 1.94 µg/g dw and 5% PVC in spiked sediment for 11 days did not significantly affect lugworm survival, feeding activity or oxidative stress biomarkers but enhanced immunotoxicity (Browne et al., 2013). PVC microspheres, combined with the antimicrobial agent triclosan (TCS; 57.30 ± 6.01 µg/g dw), compromised survival and feeding activity after the 11 days. The addition of the polycyclic aromatic hydrocarbon, phenanthrene (Phe; 0.11 ± 0.01) to PVC microspheres, enhanced oxidative stress likely due to the presence of Phe. Similarly, the polymer (PVC) combined with the stabilizer nonylphenol (NP;

0.69 ± 0.01) only revealed immune compromised effects seen as reduced phagocytic activity (Browne et al., 2013).

Overall, polychaetes exposed to flame retardants accumulated them, causing biological effects such as reduced growth. Additionally, the combined exposure of these contaminants with other stressors, as hypoxia and other contaminants (as PVC), enhanced their impacts on the immune system.

3.2.2. Biocides

The bioaccumulation of TCS for 28 days under sediment-spiked (80–800 mg/kg dw) was also investigated in *N. virens* by Perron et al. (2012). The kinetics revealed TCS concentration increased to approximately 4.0 µg/g lipid after 14-days with a slight increase to 4.5 after 28 days, after 14-days depuration it decreased to 2.5. In terms of BSAF, it was 0.23 kg organic carbon/kg lipid and it decreased by half after the depuration period.

3.2.3. Plasticizers

The consequences of BPA waterborne exposure at 10, 50 and 100 µg/L in *Perinereis aibuhitensis* over time (4, 7 and 14 days) were assessed at molecular level (Zhao et al., 2014). The results revealed that BPA induced the gene expression of Pa Gα (G proteins alpha respond to an array of stimuli, including hormones, drugs and environmental factors), and this was positively related to concentration and length of exposure. Alterations on the Gα protein pathway might be associated to the endocrine-disrupting role of BPA in polychaetes.

The same monomer (BPA) combined with NP was tested in *A. marina*, using a biodynamic model based on former reported studies (Koelmans et al., 2014). The outcomes of this study revealed that plastic ingestion by the lugworms would yield NP and BPA concentrations well below the lower ends of global NP and BPA concentration, and therefore they would not represent a relevant exposure pathway.

Phthalates constitute an important group of plastic additives. However, to the best of our knowledge, dibutyl phthalate (DBP) is the only one tested at the embryonic stages of the serpulidae *Galeolaria caespitose*. Different water (from 0.02 to 20 mg/L) and time exposure (from 15 min to 2 h) were tested, revealing sperm dysfunction as well as embryogenesis impairment after 30 min (Lu et al., 2017). DBP was also found to suppress the activity of SOD in spermatozooids and, in association with this inhibition, DBP-treated cells experienced oxidative stress (as confirmed by the presence of lipid aldehydes, such as 4-hydroxynonenal).

In general, the analysed studies demonstrated that plasticizers caused toxicity to polychaetes, affecting their performance even at low concentrations. Additionally, the increase in exposure time enhanced the impacts on polychaetes. BPA seemed to be the most hazardous plasticizer, since it induced impacts at lower concentrations.

3.3. A comparison between MPs/NPs and plastic additive laboratorial studies

A total of twenty-two studies were performed under laboratory conditions with the aim to investigate the effects of MPs/NPs and plastic additives in polychaetes (Table 1, Fig. 1).

In the last years, since concern on the effects of plastic pollution in marine species has significantly increased, the number of studies reporting on their impacts has evolved accordingly. In most of the considered former studies, it was demonstrated that, in general, polychaetes are responsive to MPs/NPs and plastic additives, even at low concentrations, showing alterations in most of the endpoints analysed. Regarding the endpoints targeted in the considered works, parameters related to polychaetes behaviour (i.e., burrowing and feeding activity) and physiology (i.e., growth) were the most considered, followed by ones related with oxidative stress and immunotoxicity.

Instead, few studies focused on the effects of mixtures of MPs (Gebhardt and Forster, 2018; Missawi et al., 2021; Revel et al., 2018) as well as MPs and plastic additives (Besseling et al., 2013; Browne et al., 2013; Gomiero et al., 2018), and although they were made responsible for biological effects, no antagonistic or synergistic effects were clearly revealed. It was also demonstrated that MPs may vehiculate plastic additives (Browne et al., 2013).

4. Field based studies on polychaetes

4.1. Microplastic

Thirteen studies with MPs were found (Table 2, Fig. 1), and none with NPs, probably due to methodological constraints. Most of them only reported the presence of MPs and their accumulation *per* individual or their concentration in tissues *per* dw or wet weight (ww) (Table 2) (e.g., Bour et al., 2018; Costa et al., 2021; Gusmão et al., 2016; Knutsen et al., 2020; Pequeno et al., 2021; Van Cauwenberghe et al., 2015), with limited information on their biological effects. However, those on MP concentration in wild organisms (sediments or water) are essential as they provide valuable information for further laboratory experiments on biological and ecological effects at realistic exposure conditions.

Only three studies investigated the effects of MP accumulation in polychaetes' tissues (Gusmão et al., 2016; Missawi et al., 2020; Van Cauwenberghe et al., 2015). Van Cauwenberghe et al. (2015) combined laboratory and field research on MPs uptake by benthic species. First, these authors analysed MPs in *A. marina* collected along the French-Belgian-Dutch coastline, finding 1.2 ± 2.8 particles/g ww of tissue. To assess effects related to MPs, the authors exposed this species to sediment previously contaminated with PS microspheres (110 particles/mL). However, no significant adverse effects on their energetic metabolism were seen. Gusmão et al. (2016) observed that three species of annelid *Saccocirrus* spp., collected in the Atlantic Ocean and the Mediterranean sandy beaches, ingested microfibrils; however, only 1 microfibre *per* polychaete was detected, with no obvious detrimental consequences. The Missawi et al. (2020) study using *H. diversicolor*, collected from Tunisian coasts, revealed that the size of MPs accumulated in field-collected polychaetes, was correlated with oxidative stress biomarkers, namely lipid peroxidation and CAT activity. Additionally, a correlation between MP abundance in sediments and polychaete bioaccumulation capacity was underlined by the same authors, detecting MP accumulation of 0.5–3.7 particles/g in wild collected specimens.

The remaining ten field studies considered, only reported on MPs presence in biota, with a great variation in accumulation depending on species and study areas (from 0.007 to 46.79 MPs/ind or 0.056 to 1000 MP/g) (Table 2). Some of them pointed out that MP accumulation may vary according to MP type, size and concentration (Md Amin et al., 2020; Missawi et al., 2020; Pequeno et al., 2021) but could also depend on the organism's feeding strategy (Bour et al., 2018; Lourenço et al., 2017). Suggesting that non-selective deposit-feeding polychaetes usually are more likely to accumulate MPs. The most commonly accumulated MPs classes were PE, PP, PET and PS, matching the production of the plastic polymers worldwide. The study by Lourenço et al. (2017) found microfibrils in benthic polychaetes collected in the Tejo estuary (Portugal) and in Banc d'Arguin (France) (2.7 ± 1.64 microfibrils/ind *H. diversicolor*, 1.0 ± 0.82 microfibrils/ind *D. neapolitana*, 3 microfibrils/ind *G. alba*, 0.5 ± 0.97 microfibrils/ind *N. caudatus*, 0.6 ± 0.74 microfibrils/ind *S. squamata*). They observed higher microfibril concentrations in polychaetes than in bivalves from the same areas, reflecting microfibril intake according to foraging strategies: polychaetes are deposit-feeders and often ingest sediment, while bivalves are filter-feeders. Moreover, this study also found a positive correlation between the concentration of microfibrils in the sediment and the percentage of fine sediments in the Tejo estuary, indicating that the

microfibrils tend to accumulate in low hydrodynamic sites (comparably to fine sediment fractions) (Lourenço et al., 2017).

The observations of Bour et al. (2018) indicated that the occurrence of MPs in *H. diversicolor* was higher than in *Sabela pavonina* collected in Oslofjord (Oslo, Norway). The former species accumulating in average 2 MPs/ind of a broader range of MP (PE, polyacrylic, PP, PBT, copolymer) while *S. pavonina* accumulated only 1 MP/ind of the PP and PE types. As in the Lourenço et al. (2017) study, the feeding mode, with deposit-feeders (*H. diversicolor*) accumulating more than filter-feeders (*S. pavonina*), was the reason for the differences observed.

The study carried out by Naidu et al. (2018) provided preliminary evidence of the presence of PS particles and fibres in the gut content of the polychaetes *Sternaspis scutata* and *Magelona cincta* from coastal waters of India.

Md Amin et al. (2020) studied the presence of MPs in surface seawater and zooplankton from Terengganu coast in Malaysia (China Sea), showing how polychaetes selectively accumulated polyamide fibres (0.007 particles/ind). Hamzah et al. (2021) also investigated the ingestion of MPs by the deposit-feeder polychaete *Namalycastis* sp. collected in the estuarine area of the Setiu Wetlands (Malaysia). These authors observed that polychaetes accumulated from 20 to 46.79 MPs/ind, identifying filaments as the majority (>99%) of MP types.

Pequeno et al. (2021) reported that the polychaete *Marphysa sanguinea* collected at Sado estuary (Southwest of Portugal) accumulated MPs from 73 to 822 μm (average size $223 \pm 233 \mu\text{m}$), with an average concentration of 0.19 ± 0.40 MP/g (0.40 ± 0.88 MP/ind), showing a preference for MPs fragments (83% of the total detected MPs) in respect to fibres.

The polychaete *Euzonus (Thoracophelia) furcifera* from the beaches of Barra del Chuy and La Coronilla (eastern Uruguay) ingested between 0.2 and 1.0 fibres/mg and 0.4–0.8 particles/mg (Vermeiren et al., 2021). In this case, MPs accumulation did not reflect sediment exposure levels (1–100 plastics/118 mL of dw sediment). The sediment grain size of areas varied from 216 to 263 μm and the authors demonstrated that MPs abundance decreased exponentially with increasing grain size.

In addition, some studies, focusing on tubicolous polychaetes, reported that their tubes also accumulate MPs usually in higher concentrations than they occur in sediments or surrounding water (Nel and Froneman, 2018; Knutsen et al., 2020; Costa et al., 2021). Nel and Froneman (2018) detected the presence of MP particles in all tube structures of *Gunnarea gaimardi* (0.056–1.113 particles/g dw) collected along the coast of South Africa. Accordingly, polychaetes from Owenidae family worms, collected in the Norwegian Continental Shelf and the Barents Sea, accumulated in their tubes significantly higher MP levels in respect to concentrations found in the local sediments (0.039–3.4 particles/g dw) (Knutsen et al., 2020). MPs accumulation in soft tissues were also reported, with concentrations (11–880 particles/g ww, corresponding to 0.10–1.9 particles/ind) higher than those found in surrounding sediments but lower compared to their tube structures (Knutsen et al., 2020). The characterisation of the sediment grain size varied from silt, clay, fine and medium sand in the studied areas, but the authors did not find a correlation between sediment granulometry and MPs accumulation (Knutsen et al., 2020). Costa et al. (2021) also observed the presence of MPs filaments and fragments in the tissues, tubes and associated to colony tubes of *Phragmatopoma caudata*. The quantity of MPs found in the tubes was three times higher than those associated with the colony. Referring to MPs types, blue filaments were more abundant in their tubes as well as in their tissues, while blue fragments were associated to the colony.

Regarding field studies, in general the species that accumulated more MPs in their tissues were *Namalycastis* sp. (from 20 to 46.79 MP/ind) and refer mainly for PP and PA.

The MPs types most frequently detected in field sediments correspond to PE (8), PP (7) and PET (6) studies. Regarding those found in polychaetes, only four studies identified their nature, resulting PE the

most accumulated MPs. Overall they conclude that MPs accumulation depended on plastic sediment concentration and the species feeding strategy (Bour et al., 2018).

4.2. Plastic additive exposures

Under field conditions, six phthalates were detected in sediments, with DEHP being dominant (> 95% of the total). These plastic additives with the associated trace metals (Cd, Co, Ni, Cu, Pb, Zn, Mn, Fe) were considered as potential drivers of the reproductive and immune disorders observed in exposed *H. diversicolor* (Cuvillier-Hot et al., 2018). This study revealed a reduced survival upon infection with a local pathogen due to an over inflammatory state in lugworms from the polluted sites.

The polychaete *Paraprionospio* sp was the sentinel chosen to study bioaccumulation of plastic additives with endocrine disrupting properties such as NP, BPA and octylphenol (OP) from the Osaka Bay, Japan (Nurulnadia et al., 2014). Chemical analysis (in ng/g ww) revealed ranges of NP (11.3–36.5 in sediment and 1460–4410 in biota), BPA (0.98–2.64 in sediment and 22.5–39.6 in the organism) and OP (2.72–5.36 in soil and 18.9–45.4 in the worm).

4.3. A comparison between MPs and plastic additive field studies

A total of fifteen field studies were included considering MPs and plastic additives, namely thirteen with MPs and only two with plastic additives (Table 2, Fig. 1). Once more, this obvious gap could be related to the fact that MPs are easier to spot while the invisible threat of plastic additives is a more recent concern.

In field studies, although diverse species were used, *H. diversicolor* was the most considered in three of the thirteen studies with MPs and in one of the two with plastic additives. Those with plastic additives also included their biological effects, while almost all studies on MPs only reported their occurrence and bioaccumulation, except three, which considered the effects of MP accumulation (Gusmão et al., 2016; Missawi et al., 2020; Van Cauwenberghe et al., 2015).

5. Bibliometric-based research trends, current hotspots and future directions

Bibliometrics is a helpful meta-analytical tool to examine a scientific field by identifying the main research areas and/or current dynamics, suggesting future needed directions (Zhang et al., 2017). In the present review the combined use of bibliometrics and a systematic literature review allowed to explore interconnections between articles related to the topic “micro/nanoplastic and plastic additives on marine anellids”. From this perspective, a comprehensive overview on research efforts, developments and possible gaps was gathered (Figs. 2 and 3). The keyword analysis revealed scientific trends as well as the direction and evolution of this topic (Fig. 2), while the high-frequency term analysis summarised the current research status and the hot-topics most investigated (Fig. 3).

The co-keyword overlay map (Fig. 2) evidenced that this topic is still at a developing stage, with some already covered themes and complex thematic evolutionary paths identified. Although some pioneering studies date from 2012, this biometric analysis revealed that the vast majority of research has been conducted in the last five years, confirming the emerging role of this topic in environmental science. Since 2016, a particular focus on environmental relevance has been gained by I) detection of plastic debris in the environment including aquatic and terrestrial sediments and II) bioaccumulation in local organisms justified by their selective food uptake strategy. Moreover, since 2018, research interest has shifted towards the marine system, with a focus on MPs impact and ecotoxicological risks for invertebrates.

In this respect, analyses on current research status and hotspots (Fig. 3) pointed out different types of plastics in the network map: PP, PA and PS as plastics and PDBE, BDE, TCS, NP and BPA as plastic additives. Additionally, results underlined the two most frequent keystone species selected to evaluate the toxicity of MPs/NPs and/or their additives: the ragworm *H. diversicolor* and the lugworm *A. marina*. The most outstanding toxicity features, related to plastic particle type and size, on sentinel species were: I) alterations on biochemical (oxidative stress, lipid peroxidation and energy reserves) and immunological (coelomocytes viability) responses; II) alteration of body growth; III) impairment in organism's behaviour; IV) accumulation in soft tissues (especially in the

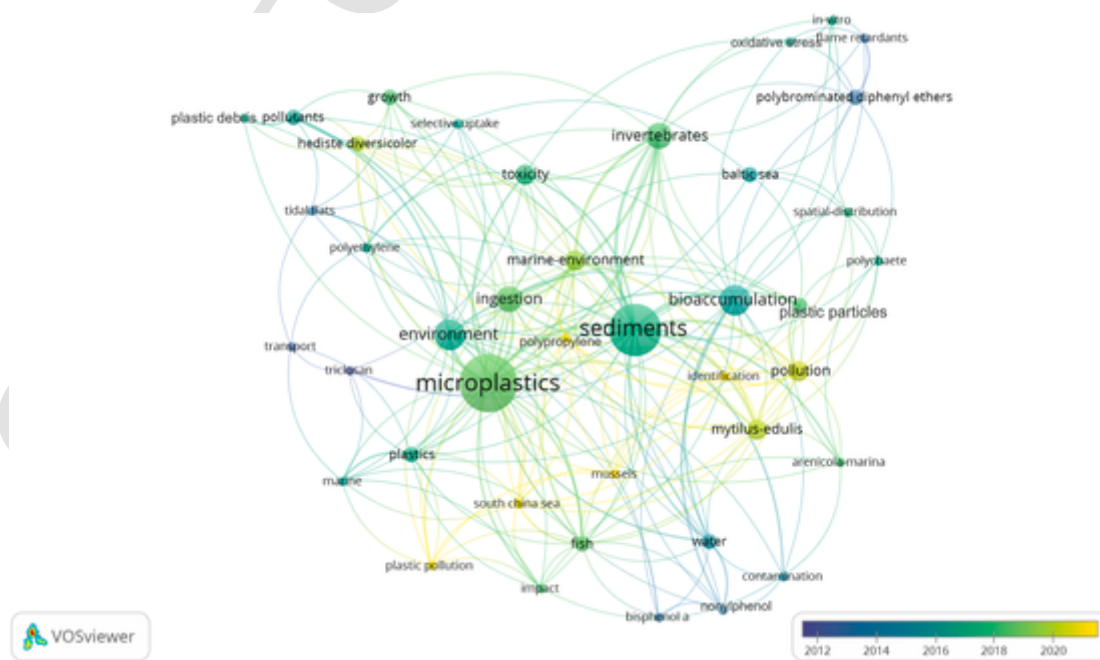


Fig. 2. Co-keyword VOSviewer overlay map of research trends and developments of the topic of MPs/NPs and plastic additives effects in marine annelids in the period of 2011–2021. The size of the circles reflects the frequency of keywords, while different colors indicate their average publication year according to the colour bar (bottom right). (For interpretation of the references to this figure legend, the reader is referred to the Web version of this article.)

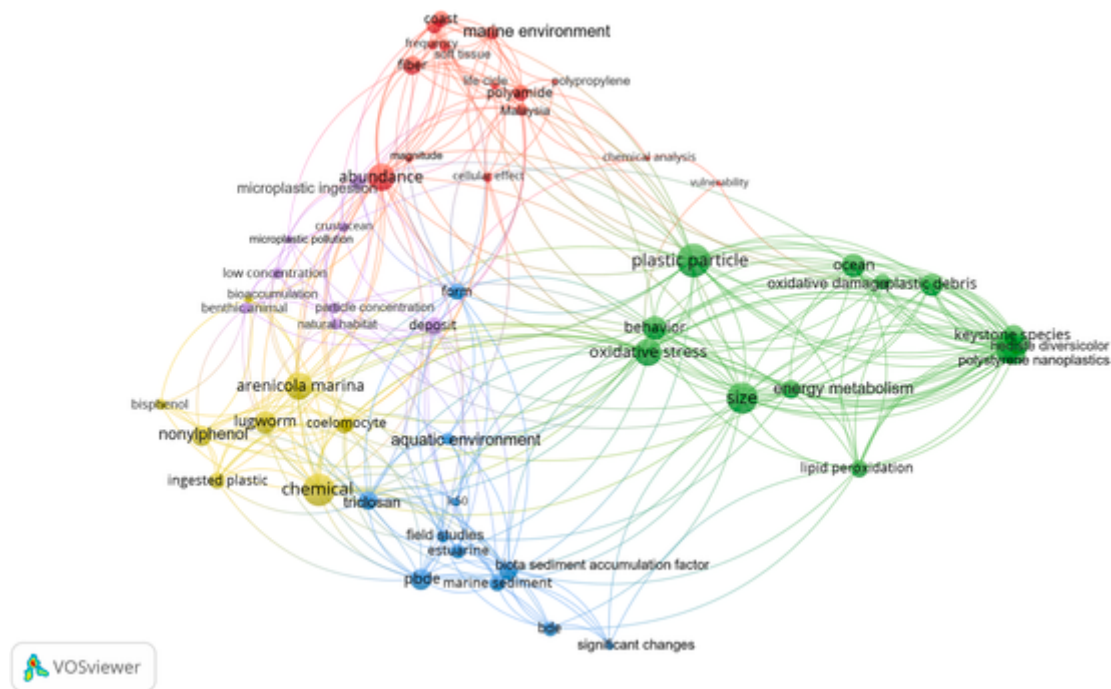


Fig. 3. Co-term VOSviewer network map of hot-topics investigated by researchers related to the topic of effects of MPs/NPs and plastic additives in marine annelids in the period of 2011–2021. The size of the circles reflects the frequency of terms, while different colors indicate their clustering division. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

fibre form). They were related to direct consequences of MPs/additives ingestion or their sediments' abundance.

In addition, the analysis pointed out the latest research interests together with the new study frontiers (Fig. 2), revealing two future emerging trends: firstly, the development of more selective and sensitive analytical methods and techniques for plastic detection in marine annelids; secondly, the investigation of the plastic impact on other marine organisms such as mussels, with special attention to *Mytilus* spp., and the ecotoxicological evaluation of PP as an emerging compound.

Considering the scarce ecotoxicological risk information on MPs/NPs and plastic additives in marine annelids, the present review recognized specific knowledge gaps to be addressed and future directions. Specifically, five prior gaps were identified: (1) develop appropriate and standardised analytical methods for extracting and detecting MPs/NPs as well as plastic additives; (2) optimise and implement sampling methodologies to better quantify plastic environmental levels and validate results from different geographical areas for reliable comparisons; (3) assess MPs/NPs exposure under more realistic laboratory-based conditions, using environmentally relevant concentrations and over time; (4) determine any possible Trojan Horse effects of plastics on polychaetes' biological responses and (5) use multiple marine taxa (in laboratory studies) or sentinel species (in field ones) to provide relevant environmental risk assessment data and understand MPs/NPs and plastic additives transfer throughout the marine food-chain.

6. Conclusions

This review considered laboratory and field studies conducted with polychaetes as model species that encompassed the accumulation and/or effects induced by MPs/NPs and plastics additives published between January 2011 and September 2021. These studies differed in terms of *via*, time and concentrations as well as targeted biomarkers. Thus, making difficult to realistically compare the impact of exposure to MPs/NPs and plastic additives and the magnitude of the responses. Nonetheless, they all evidenced that polychaetes accumulate and are usually very responsive to this contaminant class and, thus, could be ad-

equated sentinel species of estuarine/marine environments. It is also possible to infer that due to polychaetes' behaviour and feeding activity, they could play a key role in the dynamics of plastics and plastic additives in marine sediment. In particular, non-selective deposit-feeders seem to be the best candidates among polychaetes, since they are more likely to accumulate MPs. However, MPs type, size and concentration in sediments also play a role in MPs accumulation in polychaetes' tissues. From the analysed studies, PE, PP, PET and PS were the most common MPs classes accumulated by sediments, corresponding to the most manufactured plastic polymers. By far, PE was the most accumulated MPs in field situations.

In future, it would also be interesting to investigate how polychaetes' feeding and burrowing activity may influence the distribution of these contaminants in marine sediments and consequently their bioavailability for other benthic communities.

As far as field studies concerns, those analyzing the occurrence of these types of contaminants has risen in the last 5 years, probably due to the development of advanced MPs detection techniques, being undeniable that Polychaeta are particularly vulnerable to contaminated marine sediments. Given the ecological role of sediment dwelling marine polychaetes and the potential transfer from this group to higher trophic level biota of marine ecosystems, the consequences of sediment-associated MPs/NPs and plastic additives exposures and effects in this invertebrate group warrant further surveillance.

Uncited references

CEC, 2011, .

Declaration of competing interest

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

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