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# Marine litter colonization: Methodological challenges and recommendations

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Marine litter colonization by marine invertebrate species is a major global concern resulting in the dispersal of potentially invasive species has been widely reported. However, there are still several methodological challenges and uncertainties in this field of research. In this review, literature related to field studies on marine litter colonization was compiled and analyzed. A general overview of the current knowledge is presented. Major challenges and knowledge gaps were also identified, specifically concerning: 1) uncertainties in species identification, 2) lack of standardized sampling methodologies, 3) inconsistencies with the data reported, and 4) insufficient chemical-analytical approaches to understand this phenomenon. Aiming to serve as a guide for future studies, several recommendations are provided for each point, particularly considering the inaccessibility to advanced techniques and laboratories.

KEYWORDS

flotsam, colonization, invasive, dispersal, marine litter, rafting

# **1** Introduction

Marine litter is defined as all synthetic, or processed material, discarded, disposed or abandoned in the marine environment or beaches. One of the possible classifications of litter is based on material categories, such as plastics, metals, and glass, among others, and one of the main types of litter found in these environments is plastic (Ribeiro et al., 2021; Póvoa et al., 2022). Plastics are some of the most widely used materials in virtually all industries and commercially available products (Andrady and Neal, 2009; Hahladakis et al., 2018). Since the 1950s, plastic production has increased continuously due to its versatility, lightweight, resistance to corrosion, and low production costs (Andrady, 2017; Tuladhar and Yin, 2019; Torres and De-la-Torre, 2021). However, due to insufficient solid waste

management systems, infrastructure and reach, as well as the lack of environmental awareness (Prata et al., 2020; Mihai et al., 2021; Wichmann et al., 2022), plastic pollution has become one of the major issues threatening the world oceans (Walker, 2018; Chassignet et al., 2021; Haarr et al., 2022; De-la-Torre et al., 2022b). Upon entering marine and coastal environments, plastic litter interacts with biota in multiple ways (Costa et al., 2022). For instance, plastic ingestion and entanglement or entrapment are recognized as some of the most impactful plastic-biota interactions, which could compromise the survival of many top predators, such as marine birds, turtles, and mammals (Poeta et al., 2017; Battisti et al., 2019; Staffieri et al., 2019; Santillán et al., 2020; Ammendolia et al., 2022; Fukuoka et al., 2022; Provencher et al., 2022). A less considered plastic-biota interaction is the fouling of plastic litter surfaces, thus, acting as substrates for the development of invertebrate species or microbial communities (Barnes, 2002; Gong et al., 2019; Pinochet et al., 2020; Wright et al., 2021) (Figure 1). The most of plastic litter is affected by ocean surface currents due to the positive buoyancy of this object in seawater, possibly travelling for interoceanic distances (Maes and Blanke, 2015; Luna-Jorquera et al., 2019; Thiel et al., 2021). Other types of litter might reach the bottom sediments and being colonized by benthic organisms. However, less dense material can later detach or resurface. The transport of colonized litter for long distances is known as ocean rafting (Tutman et al., 2017). This phenomenon represents a threat to foreign ecosystems through the arrival of plastic rafts, which have been reported to host alien invasive species (AIS) (Rech et al., 2016; Rech et al., 2018b; Gracia and Rangel-Buitrago, 2020).

AIS is defined as exotic species that could potentially generate an impact on the ecosystem they invade (Koh et al., 2013). Upon settlement in a foreign environment, AIS may lead to the displacement of native species, particularly endemic ones, and are very difficult to eradicate (García-Gómez et al., 2021). They also represent a significant economic cost, as they compromise natural resources used to produce market goods and services (García-Gómez et al., 2021). While non-native species are able to travel long distances by natural means, like attaching to migratory biota (Thiel and Gutow, 2005), the influence of anthropic activities is undeniable. For instance, sessile species may be transported attached to ship hulls (Hänfling et al., 2011), transported in ship ballast water (Walker et al., 2019), and, as aforementioned, floating plastic litter may also act as a vector of AIS (Tutman et al., 2017; Rech et al., 2021). The latter has been subject to significant research in the last decade (Póvoa et al., 2021). However, multiple factors influencing the transportation of AIS through floating litter remain poorly understood, such as the main types of litter and polymers carrying attached biota, significant donors and recipients of colonized litter, and their overall contribution to the global issue of AIS dispersal (Rech et al., 2016).

Due to the relevancy and risk that marine litter represents for the dispersion of non-native and potentially invasive species, multiple authors have compiled and analyzed the literature. For instance, Kiessling et al. (2015) compiled studies on organisms that inhabit floating marine litter. The characteristics of the litter and biological traits of associated species were analyzed to understand their colonizing behavior. However, in the last six years, recent studies have allowed researchers to elucidate new findings related to marine



#### FIGURE 1

Examples of a plastic bottle (A) colonized by the bivalve Semimytilus algosus (B), anemones (Order: Actiniaria) (C) y bristle worms (Class: Polychaeta) (D), and a rubber boot (E) colonized by Balanus sp. (F). (Photographs by G.E.D.).

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litter colonization that were not previously understood, such as the influence of chemical-analytical aspects of synthetic substrates and the use of genetic identification of fouling species (e.g., molecular analyzes). More recently, García-Gómez et al. (2021) carried out a literature search aiming to compare the potential of plastic as a vector of non-native species compared to other sources of dispersal (e.g., ship hull biofouling and ballast waters). Also, their analysis of the composition of fouling communities on marine litter substrates, primarily plastic, indicated that these species generally have a short life cycle and larval development. Póvoa et al. (2021) conducted a literature review focused on the research questions proposed by Rech et al. (2016) and Gracia C. et al. (2018). Although the aforementioned studies correctly compiled information regarding marine litter colonization and carried out multiple analyzes, it is necessary to carry out an updated literature search to analyze the multiple challenges that remain in this field of research. Hence, in the present review, an updated overview of the current knowledge regarding the occurrence of marine litter colonized by marine macroinvertebrates is provided. We aim to identify and discuss the various factors surrounding this field of research and establish key recommendations to guide future studies, particularly concerning sampling, species identification, litter data reporting, and chemical characterization methodologies.

# 2 Literature search methodology

The literature search was conducted adjusting De-la-Torre et al.'s (2022a) criteria to the topic of the present study. On the  $10^{th}$  of August 2022, the Scopus (https://www.scopus.com/) database was consulted with the keywords "litter" OR "plastic" in conjunction with "fouling" OR "rafting" OR "invasive species" OR "non-native" OR "biological invasion" in conjunction with "marine" OR "ocean". The search was carried out within the title, abstract, and keywords of the documents in the database. Publication year intervals were set from 2000 to 2023. Additionally, three document types (editorials, book chapters, and reviews) and the subject areas (Computer Science, Social Sciences, Mathematics, Economics, and Business management) were excluded from the search. The search resulted in 274 documents, which were exported to the virtual platform Rayyan (https://www.rayyan.ai/), where the title and abstract of each document were revised in detail to determine whether to include or exclude studies according to a specific criteria. Studies were included if reports of colonizing organisms on marine litter (either marine or coastal zones), including those recovered from benthic areas, were presented. Only field studies (e.g., marine litter surveys) were included. Studies conducting involving litter colonization under controlled experimental conditions were excluded from the analysis.

# **3** Results and discussion

A total of 39 documents were selected. Relevant information (time, location, environmental matrix, number of species, type of litter, and sources) from each study was extracted to obtain a better understanding of the current state of the knowledge. Table 1 summarizes the main elements of each study. Additionally, a geographic map was constructed indicating the main locations of each of the consulted studies (Figure 2). As shown in Figure 2, most studies were conducted in South America, Oceania, and Europe, with a single study reported on the Antarctic Peninsula by Barnes and Fraser (2003), while East Asia and Africa have been poorly assessed, as well as the coasts surrounding the Indian Ocean, and the east coast of North America. It should be mentioned that the 2011 Tōhoku earthquake and tsunami sparked various studies conducted in the following years investigating marine litter arriving from Japan to North America (North Pacific Ocean) (Carlton et al., 2017; McCuller and Carlton, 2018; Póvoa et al., 2021).

The studies consulted addressed multiple research questions involving the occurrence of colonized litter in diverse areas, as well as the transportation of colonized litter (rafting). Most studies surveyed stranded litter, although the focus of the studies differed depending on specific sources of contamination. For instance, Rech et al. (2018b) focused on understanding the contribution of mariculture areas on the release of rafting AIS in Europe, while Rech et al. (2018c) investigated the arrival of colonized litter on beaches of the Rapa Nui Island, a remote island in the South Pacific Ocean. Furthermore, De-la-Torre et al. (2021) highlighted that the Peruvian coast may act as a source of colonized marine litter rather than the arrival of rafts containing AIS. McCuller and Carlton (2018) focused on a very specific event (e.g., the Great East Japan Earthquake and Tsunami of 2011) and its repercussion on transoceanic species dispersal on large-scale marine litter. On the other hand, Crocetta et al. (2020) investigated benthic marine litter composition and abundance of mega- and macrofauna by trawling at depths of 50 and 100 m. The number of studies focusing on benthic marine litter is reduced, while others combined approaches by investigating beaches, floating, and benthic marine litter (e.g., Subías-Baratau et al., 2022). Conducting multiple approaches allows researchers to understand important factors in the transport of floating marine litter, such as the role of biofouling on plastic sinking rates (Chen et al., 2019). While marine litter colonization remains less investigated than other dispersal pathways (García-Gómez et al., 2021), recent studies have provided multiple perspectives on the factors involved in biofouling colonization and dispersal. However, several challenges remain ahead.

As mentioned previously, 28 studies (71.8%) were conducted in coastal zones or beaches, while studies recovering fouled litter from the open ocean or sea are limited (11 studies, 28.2%) (Table 1). This is likely due to the ease and accessibility to coastal zones as also mentioned by Póvoa et al. (2021). Likewise, one study evaluated the state of colonization of submerged litter (Mantelatto et al., 2020), although submerged debris (generally dense materials) is unlikely to resurface and serve as AIS rafts. Nevertheless, submerged debris may be subject to abiotic influences (e.g., weathering conditions, water currents) and colonizing species than those experienced by floating debris.

Sampling methodologies have not been standardized to assess colonizing biota on beached litter. Seven studies (17.9%) chose to monitor the entire beach, from the tide line to the maximum limit of the beach (vegetation or trails) (e.g., Miralles et al., 2018; Rech et al., 2018a; Ibabe et al., 2020; De-la-Torre et al., 2021). This method (covering the entirety of the beach) allows recovering a greater amount of litter and obtaining a complete view of the abundance of

## TABLE 1 Characteristics of methodologies and results of the 39 studies evaluated.

Country	Specific zone	Environmental Matrix	Sampling year	Sampling type	Species list <sup>1</sup>	Taxonomic class <sup>2</sup>	Type of litter	Source	Polymer types	Ref.
Peru	Lima-Cañete	Beach	2019-2020	Whole beach survey	Balanus laevis Seminytilus algosus Prisogaster niger Alia unifasciata Perumytilus Purpuratus Hyalella sp. Tetrapygus niger Ocypode occidentalis Emerita analoga Chiton cumingsii Tegula atra	Bivalvia Gastropoda Thecostraca Ophiuroidea Malacostraca Polychaeta Echinoidea Polyplacophora	Clothing Bottles Food containers Plastic bags Sheetings Monofilament line Fishing net Other plastic Timber	Land- based (81.5%) Sea- based (18.5%)	Polyester/ PET (18.5%) Nylon/PA (11.1%) PP (25.9%) LDPE (22.2%) Latex (3.7%)	(De-la- Torre et al., 2021)
Spain	Gijon port	Beach	2017	Whole beach survey	Platynereis dumerilii Syllis gracilis Mytilus edulis Patella vulgata	Polychaeta Bivalvia Gastropoda	Plastic bag Bottles Fabric Buoy Fishing gear EPS Plastic debris	Land- based Sea- based	-	(Ibabe et al., 2020)
Pacific Ocean	North Pacific Subtropical Gyre	Ocean	2012	Floating debris picked up manually	Class: Hydrozoa Actiniidae Family: Actiniidae Family: Metridiidae Amphinome rostrata Chaetopterus sp. Parasabella sp. Hipponoe gaudichaudi Lepidonotus sp. Mytilus sp. Fiona pinnata Lottia pelta Lepas spp. Idotea metallica Pentidotea wosnesenskii Glebocarcinus amphioetus Planes major Planes major Planes major Planes major Planes major Planes major Spene sp. Pomacentridae	Hydrozoa Hexacorallia Polychaeta Bivalvia Gastropoda Thecostraca Malacostraca Gymnolaemata Actinopterygii	Buoy Toy ball Bottle cap Flat piece of plastic Bottle Boat bumper	Land- based Sea- based	-	(Gil and Pfaller, 2016)
Turkey	Sarayköy Beach	Beach	2016-2017	OSPAR transect protocol	Mytilus sp. Family: Balanidae Class: Gastropoda Phylum: Bryozoan	Bivalvia Thecostraca Gastropoda	Plastic bottle Foam Shoe	Land- based	-	(Aytan et al., 2019)
Brazil	Ilha Grande Bay	Beach	2018-2020	Along the strandline	Amphibalanus improvisus Amphibalanus reticulatus Amphibalanus sp. Newmanella radiata Lepas anatifera Ostrea puelchana Saccostrea cuculatta Hydroides elegans Hydroides sp. Family: Membraniporidae Tubastraea coccinea Tubastraea tagusensis	Thecostraca Bivalvia Polychaeta Gymnolaemata Anthozoa	Caps Shoes Buoy Shoes Rubbers Styrofoam Plastic fragments	Land- based Sea- based	~	(Póvoa et al., 2022)

Brazil	Ilha Grande Bay	Beach and deep sea	2012-2014 (Benthos) 2018-2020 (Beach)	Scuba diving Along the strandline	Tubastraea coccinea Tubastraea tagusensis	Hexacorallia	Buoy EPS Rope Electric cable Flip-flop (shoe) Wood Tire Glass bottle	Land- based (75%) Sea- based (25%)	-	(Mantelatto et al., 2020)
Spain	Catalan Sea (NW Mediterranean)	Beach and sea	2020-2021 (Trawling) 2020 (Beach)	Trawling Along the strandline	Spirobranchus triqueter Novocrania sp. Chorizopora brongniartii Scyliorhinus sp. Arbopercula tenella Aetea sica Annectocyma sp. Cryptosula pallasiana Fenestrulina malusii Phallusia mammillata Ophiothrix fragilis Barbatia barbata Aplousina sp. Copidozum tenuirostre Escharella variolosa Hagiosynodos latus Myriapora truncata Plagioecia sp. Reptadeonella violacea Schizoporella dunkeri Alcyonium palmatum Eunicella verrucosa Lepto grija sarmentosa Neopycnodonte cochlear	Polychaeta Craniata Gymnolaemata Chondrichthyes Stenolaemata Ascidiacea Ophiuroidea Bivalvia Stenolaemata Anthozoa Thecostraca	Not specified		PE (47%) PP (13.7%) PET (11.8%) Chlorinated poly-ethylene (CPE) (9.8%) PS (7.8%) PU (3.9%) PVC (3.9%) PA (2.0%)	(Subias- Baratau et al., 2022)
Argentina	Mar Chiquita, Buenos Aires	Beach	2017-2018	Debris collected from bins at beach	Amphibalanus improvisus Brachidontes rodriguezii Ostrea sp. Membranipora sp. Amphisbetia operculata Class: Polychaeta	Thecostraca Bivalvia Gymnolaemata Hydrozoa Polychaeta	Fishing line Buoy Fishing rope Cap Swim googles Strap Plastic bottle Sunglasses Food film Backpack strap Electric cable Aluminized paper Hose Tire Other plastics	Land- based (66.6%) Sea- based (33.3%)	-	(Rumbold et al., 2020)
Croatia	Mar Adriático	Sea	2014	Floating debris picked up manually	Planes minutus Liocarcinus navigator	Malacostraca	Tire Sandal Shoe	Land- based (100%	-	(Tutman et al., 2017)
Colombia	Atlantico department	Beach	NS	Transect protocol	Arbopercula tenella Arbopercula angulata 3 unidentified bryozoan Lepas anserifera Class: Polychaeta	Gymnolaemata Thecostraca Polychaeta	Wood Propagule Plastic debris Cap Plastic jar Plastic bottle Plastic ring Paint pot Glass bottle Other plastics	Land- based	-	(Gracia C. et al., 2018)

Colombia	Atlantico and Magdalena department	Beach	2018	-	Perna viridis	Bivalvia	Buoy	Sea- based	-	(Gracia and Rangel- Buitrago, 2020)
Italy and Portugal	Venice and Algarve	Beach	2016	Whole beach survey	Austrominius modestus Amphibalanus amphitrite Anomia epphipium Hesperibalanus fallax Magallana angulata Bugula neritina Bugula neritina Santa trigonus triqueter Lepas pectinata Lepas pectinata Lepas pectinata Lepas pectinata Buguloprovincialis Mytilus sp. Modiolula sp. Ostrea edulis Chthamalus montagui Perforatus Verruca stroemia Class: Ophiuroidea	Thecostraca Bivalvia Gymnolaemata Polychaeta Ophiuroidea	Buoy Mussel bag Fishing trap Ropes Plastic bottles Other plastics Processed timber	Land- based (36%) Sea- based (64%)		(Rech et al., 2018b)
Chile	Easter Island	Beach	2017	Along the strandline	Family: Serpulidae Planes major Halobates sericeus Lepas anatifera Chthamalidae sp. Jellyella eburnea Pocillopora sp. Class: Hydrozoa Other	Polychaeta Malacostraca Insecta Thecostraca Gymnolaemata Anthozoa Hydrozoa	Bottle caps Plastic bottles Crates/Baskets Fishing gear Rope Other plastic	Land- based Sea- based	-	(Rech et al., 2018c)
Australia	Victoria	Beach	2019	Not specified	Lepas pectinata	Thecostraca	Bottle	Land- based	-	(Cooke and Sumer, 2021)
Spain	Asturias	Beach	2016	Whole beach survey	Amphibalanus amphitrite Austrominius modestus Perforatus Chthamalus stellatus Neoacasta laevigata Lepas anatifera Lepas pectinata Pachygrapsus marmoratus Polybius henslowii Magallana gigas Mytilus edulis Mytilus edulis Mytilus edulis Mytilus torssulus Tritia reticulata Eumida bahusiensis Neodexiospira alveolata Paragorgia arborea	Thecostraca Malacostraca Bivalvia Gastropoda Polychaeta Anthozoa	Bottles Fishing gear	Land- based (~40%) Sea- based (~60%	-	(Miralles et al., 2018)

		1								
Italy	Ganzirri (Sicily)	Beach	2016-2019	Not specified	Megabalanus tulipiformis Pachylasma giganteum Octolasmis sp. Adna anglica Alcyonium coralloides Coencoyathus cylindricus Desmophyllum pertusum Desmophyllum dianthus Family: Caryophylliidae Madrepora oculata Order: Zoantharia Errina aspera Sertulareila sp. Family: Sertulariidae Class: Hydrozoa Pedicularia sicula Neopycnodonte cochlear Striarca lactea Family: Sertulariidae Class: Hydrozoa Pedicularia sicula Neopycnodonte cochlear Striarca lactea Family: Serpulidae Metavermilia multicristata Vermiliopsis sp Filogranula sp. Filogranula gracilis Filogranula gracilis Filogranus Semivermilia Semivermilia Semivermilia Serivermilia Senivermilia Senivermilia Senivermilia Senivermilia Sycon raphanus Order: Cheilostomatida Cellepora sp Haplopoma sp. Cellaria salicornoides Puellina cfr gatyae Puellina sp. Order:	Thecostraca Anthozoa Hydrozoa Gastropoda Bivalvia Polychaeta Calcarea Gymnolaemata Stenolaemata Stenolaemata	Fishing gear	Sea- based		(Battaglia et al., 2019)
USA	-	Beach	2012-2017	Not specified	<i>Cyclostomatida</i> 289 species	_	vessels, docks, buoys, totes (crates), wood, and many other objects, identified as JTMD	Land- based Sea- based	_	(Carlton et al., 2017)
Spain	Asturias	Beach	2016	Whole beach survey	Lepas anatifera Lepas anserifera Lepas pectinata Dosima fascicularis Austrominius modestus Chthamalus stellatus Chthamalus stellatus Chthamalus Balanidae sp Verruca stroemia Caprella andreae Mytilus edulis Mytilus edulis Mytilus ep Crassostrea gigas Ostrea stentina	Malacostraca Thecostraca Malacostraca Bivalvia Gastropoda Polychaeta Hydrozoa	Hard plastic Other plastic Foam Non-plastic	Land- based Sea- based	-	(Rech et al., 2018a)

		1							1	1
					Gibbula umbilicali Spirobranchus triqueter Spirobranchus taeniatus Serpula columbiana Neodexiospira sp. Spirobranchus sp. Bougainvillia muscus Obelia dichotoma Helix aspersa					
Sweden	Gothenburg	Beach	-	Transect protocol	63 species	Bivalvia Phylum: Bryozoan Polyplacophora Gastropoda Malacostraca Polychaeta Thecostraca Others	Glass Ceramic Wood Fabric Metal Plastic	-	-	(Garcia- Vazquez et al., 2018)
South Pacific Ocean	-	Ocean	2015-2017	Floating debris recovery using nets, trawls, and snorkeling	Campanulariidae sp. 1 Campanulariidae sp. 2 Obelia sp. Pocillopora sp. Fiona pinnata Hipponoe gaudichaudi Lepas sp. Lepas pectinata Order: Amphipoda Stenothoe gallensis Caprella andreae Planes marinus Idotea metallica Idotea sp. Jellyella eburnea Jellyella tuberculata Hirundichthys sp. Cheilopogon sp. Others	Hydrozoa Anthozoa Gastropoda Polychaeta Thecostraca Malacostraca Gymnolaemata	Plastic pieces Jerrycans and buckets Crates and baskets Buoys Others	Land- based (28.8%) Sea- based (41.2%)	PP (3.4%) EV (3.4%) PE (93.1%)	(Rech et al., 2021)
Norway	Svalbard	Beach	2017	Transect protocol	Electra spp. Eucratea loricata Lepas anatifera Semibalanus balanoides Class: Gastropoda Mytilus sp.	Gymnolaemata Thecostraca Gastropoda Bivalvia	Fishing box Barrel Containers	Land- based Sea- based	-	(Węsławski and Kotwicki, 2018)
Malaysia	Penang and Langkawi	Beach	-	Whole beach survey	Acanthodesia perambulata Acanthodesia irregulata Jellyella eburnea	Gymnolaemata	Plastic debris Glass bottle	-	-	(Taylor and Tan, 2015)
Chile	Coquimbo	Ocean	2001-2005	Floating debris picked up manually	102 species	Ascidiacea Polychaeta Malacostraca Thecostraca Others	Buoys	Sea- based	-	(Astudillo et al., 2009)
Antarctica	Adelaide Island	Beach	2003	Not specified	Laevilitorina antarctica Aimulosia antarctica Arachnopusia inchoata Ellisina antarctica Fenestrulina rugula Micropora Drevissima Others	Demospongiae Polychaeta Hydrozoa Gastropoda	Plastic band	Land- based	-	(Barnes and Fraser, 2003)

Uruguay	Rocha department	Beach	2014	Not specified	Pinctada imbricata	Bivalvia	Rope	Land- based	-	(Marques and Breves, 2014)
Brazil	Rio de Janeiro	Beach	2012	Not specified	Petaloconchus varians	Gastropoda	Marine debris	-	-	(Breves and Skinner, 2014)
The Netherlands	Texel	Beach	2009	Not specified	Favia fragum	Anthozoa	Metal cylinder	-	-	(Hoeksema et al., 2012)
Brazil	Sao Paulo Rio de Janeiro	Beach	_	Not specified	Tubastraea coccinea Tubastraea tagusensis	Anthozoa	Styrofoam	-	-	(Faria and Kitahara, 2020)
Iran	Coast of the Persian Gulf	Beach	2016-2018	Along the strandline	Amphibalanus amphirite Microeuraphia permitin Striatobalanus amaryllis Chthamalus barnesi Spirorbis sp. Hydroides sp. Saccostrea cucullata Isognomon legumen Class: Bivalvia Diodora funiculata Chiton sp. Parasmittina egyptica Microporella browni Antropora tincta Paracyathus stokesii Class: Ascidiacea	Bivalvia Gastropoda Thecostraca Polychaeta Gymnolaemata Anthozoa Ascidiacea	Plastic Wood Glass Metal cans	-	-	(Shabani et al., 2019)
North Pacific Ocean	-	Ocean	2009-2012	Floating debris picked up with a net	95 species	Polychaeta Malacostraca Pycnogonida Gymnolaemata Stenolaemata Perciformes Phylum: Chordata Heterotrichea Anthozoa Hydrozoa Ophiuroidea Polythalamea Bivalvia Gastropoda Rhabditophora Turbellaria Calcarea Demospongiae	Rigid fragment Rope clumps Flexible substrates Expanded foam	-	-	(Goldstein et al., 2014)
Stewart Island Falkland Island Bird Island	-	Beach	2001-2004	Not specified	Lepas australis	Thecostraca	Plastic debris Wood	-	-	(Barnes et al., 2004)
New Zealand	Western Coromandel Peninsula	Beach	2015-2016	Transect protocol	NS	Phylum: Porifera Phylum: Hydrozoa Phylum: Bryozoa Phylum: Arthropoda Phylum: Mollusca Phylum: Annelida	Plastic Ceramic/glass Metal Cloth Foam Rubber Wood	_	-	(Campbell et al., 2017)
Turkey	Mersin Bay	Ocean	_	Trawling	Bougainvillia muscus	Hydrozoa Polychaeta	Plastic debris	-	EPC (2%) PE (72%)	(Gündoğdu et al., 2017)

					Spirobranchus triqueter Hydroides sp. Serpula sp. Serpula sp. Serpula vermicularis Order: Sabellida Phylum: Bryozoa Neopycnodonte cochlear Musculus subpictus Anomia ephippium Corbula gibba Sephia officinalis Diodora sp. Lepas antifera Perforatus Galathea intermedia Ascidiella aspersa Phallusia mammillata Ciona intestinalis Styela sp.	Phylum: Bryozoa Bivalvia Cephalopoda Gastropoda Hexanauplia Malacostraca Ascidiacea			PET (3%) Poly-E (1%) PP (12%) NY-6 (1%) PVC (1%) PS (1%) SAC (1%) PET/PP (1%) Unidentified (7%)	
Israel	Shefayim	Beach	2014	Not specified	Cerithiopsis tenthrenois Crisilla semistriata Arca noae Striarca lactea Gregariella cf. ehrenbergi Musculus subpictus Musculus costulatus Lithophaga Modiolus cf. barbatus Arcuatula senhousia Brachidontes pharaonis Septifer cumingii Mytilaster cf. minimus Pinctada imbricata radiata Ostrea edulis Dendostrea cf. folium Malleus regula Chama pacifica Sphenia binghamii Cucurbitula cymbium Rocellaria dubia	Gastropoda Bivalvia	Buoy	Sea- based	-	(Ivkić et al., 2019)
Australia	-	Ocean	_	Trawling	Phylum: Bryozoa Lepas spp. Order: Isopoda Halobates sp. Phylum: Annelida	Phylum: Bryozoa Phylum: Annelida Insecta Malacostraca Thecostraca	Plastic fragments	-	PE (83%) PP (17%)	(Reisser et al., 2014)
Mediterranean Sea	Ligurian sea	Ocean	1997	Floating objects were collected	Arbacia lixula Bowerbankia gracilis Callopora lineata Clytia hemisphaerica Cymodocea nodosa Doto sp. Electra posidoniae Eudendrium sp. Fiona pinnata Gonothyrea Idotea metallica Laomedea	Echinoidea Gymnolaemata Hydrozoa Gastropoda Malacostraca Thecostraca Polychaeta	Plastic bag Hard plastic debris Styrofoam Bottles Wood Fishing gear	-	-	(Aliani and Molcard, 2003)

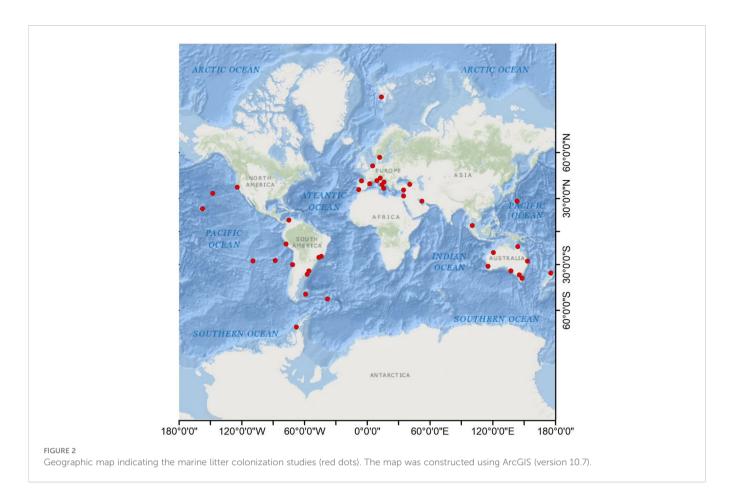
					angulata Lepas pectinata Membranipora membranacea Nereis falsa Obelia dichotoma Phitisica marina Spirobranchus polytrema					
Mediterranean Sea	Gulf of Pozzuoli	Ocean	2019	Trawling at different depths	-	Phylum: Bryozoa Phylum: Cnidaria Phylum: Porifera Phylum: Chordata Phylum: Arthropoda Phylum: Mollusca Phylum: Annelida	Cotton Glass Metal Nylon Paper Plastic Pottery Concrete Rubber Synthetic textile Wood	Land- based (83.6%) Sea- based (16.4%)	-	(Crocetta et al., 2020)
USA	-	Beach	2012-2017	Large-scale landing on coastlines (no specific methodology)	49 species of bryozoans	Stenolaemata Gymnolaemata	Totes, crates or containers Vessels Buoys, floats Other items Post and beam wood Trees/logs Pontoon sections Misawa docks	Land- based Sea- based	-	(McCuller and Carlton, 2018)
Morocco	Mediterranean coast	Beach	2022	Whole beach survey	Lepas pectinata Perforatus Lepas anatifera Phylum: Bryozoa Class: Thecostraca Spirobranchus triqueter Neopycnodonte cochlear Hydroides sp Ophiothrix fragilis Eunicella verrucosa Myriapora truncuta	Thecostraca Phylum: Bryozoa Polychaeta Bivalvia Ophiuroidea Octocorallia Gymnolaemata	Bottles <2 L Bottles >2 L Food containers Plastic bags Plastic buoys Ropes Plastic tube Clothing, shoes Bottles and jars Other wood	Land- based (93.3%) Sea- based (6.7%)	-	(Mghili et al., 2022)

ABS-indicates acrylonitrile butadiene styrene; PC-indicates polycarbonate; PE-indicates polyethylene; LDPE-indicates low-density polyethylene; MDPE-indicates medium-density polyethylene; HDPE-indicates high-density polyethylene; PET-indicates polyethylene terephthalate; PMMA-indicates poly (methyl methacrylate); PP-indicates polypropylene; PTFE-indicates polytetrafluoroethylene; PS-indicates polystyrene; PVC-indicates polyvinyl chloride; PU-indicates polyuethane. Species in bold were identified as non-indigenous species or AIS. <sup>1</sup>If organisms were not identified at species level, the lowest possible taxonomic classification was mentioned.

<sup>2</sup>If organisms were not identified at class level, the phylum was mentioned.

litter per beach. However, this method may incur in sampling bias when the sampling length or quadrants dimensions are different across sampling locations, in addition to requiring more effort, as determined by Pizarro-Ortega et al. (2022). To this end, the resolution of sampling designs should be standardized and similar across sampling locations. Conversely, other studies (n = 5; 12.8%)followed the tidal lines in search of colonized litter (Rech et al., 2018c; Mantelatto et al., 2020; Póvoa et al., 2022; Subías-Baratau et al., 2022). With this method, it is possible to quickly assess the litter that has been in contact with the tides that are concentrated in the tide line. Regardless, focusing on the strandline or whole beach area may be indicators of incoming or accumulated colonized litter, respectively. Thus, studies should consider this interpretation to determine the methodology that better aligns with their objectives. Other studies ran transect- and quadrat-based sampling methods (n = 5; 12.8%) (Taylor and Tan, 2015; Gracia C. et al., 2018; Aytan et al., 2019). On several occasions, the sampling methodology was not specified (n = 9; 23.1%). It could be assumed that colonized litters were collected opportunistically, although important data, such as colonized litter density (e.g., colonized litters per total number of litter or beach area), is not obtained. The heterogeneity in the methodologies used to search for colonized litter makes the comparison between studies difficult. For this reason, it is necessary to reach a consensus regarding the methodology used for this purpose, as well as the reported data, as also mentioned by Póvoa et al. (2021). For example, very few studies reported the percentage of colonized litter with respect to the total number of litter found (Rech et al., 2018c; Póvoa et al., 2022). Also, no previous study reported the number of colonized (and non-colonized) marine litter per unit area.

A significant number of species have been reported, belonging to 31 taxonomic classes, as displayed in Table 1. While some authors focused on one or two relevant species (Gracia and Rangel-Buitrago, 2020; Cooke and Sumer, 2021), others report hundreds of species found as a result of more extensive sampling methodologies



(Astudillo et al., 2009; Carlton et al., 2017). Some of the most frequent taxonomic classes among studies are Bivalvia (Phylum: Mollusca), Gastropoda (Phylum: Mollusca), Thecostraca (Phylum: Arthropoda), Malacostraca (Phylum: Arthropoda), Polychaeta (Phylum: Annelida), and Gymnolaemata (Phylum: Bryozoa). Most of the species in these classes are sessile or have a limited degree of mobility, while others, such as those of the class Malacostraca, have a higher degree of mobility. This suggests that, in terms of species transport, marine litter not only functions as a substrate for species proliferation but also as a potential hiding place for more mobile species. In a particular case, Subias-Baratau et al. (2022) reported the presence of egg capsules of a shark of the genus Scyliorhinus. This behavior would not only mean a possible dispersion of species, but also an impact on the survival of oviparous chondrichthyans. However, the dynamics between oviposition, dispersal and hatching of larger species require further investigation, as studies are limited (De-la-Torre et al., 2022c). It should be noted that biofilm-forming microbial communities that settle on the synthetic substrate in the first weeks in contact with seawater may influence macroinvertebrate assemblages. Diatoms and bacteria become attached to the surface of a substrate that is rich in protein, carbohydrates, and glycoproteins after being in contact with seawater; these organisms then secrete extracellular polymeric substances to become embedded (Müller et al., 2013). The proliferation of macroalgae and other organisms occurs later on (Kiessling et al., 2015). While this review focused on macroinvertebrates, the importance of the first biofilm-forming microbes to understanding complex macroinvertebrate assemblages on marine litter should be noted and further investigated.

Most of the studies reported native or cosmopolitan species (e.g., Lepas spp.) in their samples, or did not categorize them, while invasive species and their proportion with respect to the total species are reported in a few studies. For instance, in aquaculture areas of Italy and Portugal, Rech et al. (2018b) reported the presence of invasive species of the class Thecostraca [Amphibalanus amphitrite (Darwin, 1854), Austrominius modestus (Darwin, 1854), Balanus trigonus (Darwin, 1854) and Hesperibalanus fallax (Broch, 1927)], Polychaeta [Hydroides elegans (Haswell, 1883) and Hydroides sanctaecrucis (Krøyer, 1863)] and Bivalvia [Magallana angulata (Lamarck, 1819)]. Povoa et al. (2022) found several species of anthozoans (Class: Anthozoa) classified as invasive [Tubastraea coccinea (Lesson, 1830), Tubastraea sp., and Tubastraea tagusensis (Wells, 1982)] mainly in polystyrene-based materials (e.g., buoys and expanded polystyrene) on Brazilian beaches. Likewise, the bivalve Saccostrea cuculatta (Born, 1778), and polychaete H. elegans in expanded polystyrene and plastic fragments, respectively. Although several studies recognize invasive species by comparing international databases (e.g., Global invasive species database, http://www.issg.org/ database) and regional studies, considering the multiple species of various taxonomic levels, the list may be evolving. Regardless, it is almost impossible to determine the source of dispersal of non-native species that are already proliferating in foreign environments.

One of the main barriers in the study of invasive species is the identification of these at the species level as also mentioned by Póvoa et al. (2021). In multiple studies, organisms are identified at family, order, or class level (Gil and Pfaller, 2016; Carlton et al., 2017; De-la-Torre et al., 2021), which makes it difficult to determine potential

invaders. However, several studies opted for the genetic identification of some potentially invasive species through DNA barcoding (Miralles et al., 2018; Rech et al., 2018a; Rech et al., 2018b; Rech et al., 2018c; Ibabe et al., 2020; Rech et al., 2021). For example, Rech et al. (2018a) identified species classified as invasive Crassostrea gigas (Thunberg, 1793), Ostrea stentina (Payraudeau, 1826) (class: Bivalvia), A. modestus (class: Thecostraca), Serpula columbiana (Johnson, 1901), and Neodexiospira sp. (class: Polychaeta) through DNA barcoding. The main limitation of DNA barcoding is the availability of reference libraries for taxonomic identification of the genetic sequence (Hellberg et al., 2016). However, new studies are constantly contributing to filling the gaps in reference libraries (Leite et al., 2020). On the other hand, DNA barcoding is not always readily available to research groups and institutions worldwide, particularly in developing countries, such as Peru and Brazil. In this context, taxonomists or research groups must make efforts to consider taxonomic analyzes that help build barcoding libraries of invertebrates and make efforts to construct fruitful collaborations to gain access to genetic identification.

Types of colonized litter reported in these studies are generally diverse, including textiles, bottles, plastic containers and bags, processed wood, fishing gear (e.g., nets), and buoys, among others (e.g., Gracia C. et al., 2018; Mantelatto et al., 2020; Rumbold et al., 2020; De-la-Torre et al., 2021). However, the categorization of litter is often not standardized (Póvoa et al., 2021). For example, Rech et al. (2018a) classify most litter as "hard plastic", "other plastics", "foams", and "non-plastic", while Mantelatto et al. (2020) propose a more specific classification, including tires, plastic bottles, electric cables, Styrofoam, among others. Other studies report the predominance of only one or two types of colonized litter, mainly buoys and plastic bottles (e.g., Aytan et al., 2019; Cooke and Sumer, 2021), or they do not report it and concentrate on its chemical composition (e.g., Subías-Baratau et al., 2022). The divergence between the classifications used between studies generates complications when integrating the data reported in the literature to determine which substrates are widely preferred by organisms. For this reason, it is necessary to standardize the classification with remarkable specificity. Furthermore, substrate preference may also by influenced by litter type availability. In this sense, litter classification should also include a standard method for litter type (different substrate types) quantification.

The diversity of litter types found can be influenced by experimental design, sources of contamination, and sampling efforts (Rees and Pond, 1995; Velander and Mocogni, 1999). A temporary study carried out in a large coastal area contaminated by a variety of sources is likely to report a greater diversity of colonized litter than a shorter, more focused study. For instance, Carlton et al. (2017) carried out the most extensive study on the occurrence of colonized marine litter on the Pacific coast of the United States. In more than six years, 634 objects and litter from Japan of a great variety (wood, buoys, plastics, metals, ropes, boxes, and electronics) were reported. Secondly, Gracia and Rangel-Buitrago (2020) focused on reporting the occurrence of the invasive bivalve Perna viridis (Linnaeus, 1758) in buoys found on the Caribbean coast of Colombia, while Astudillo et al. (2009) compared the communities found in buoys in use and detached. In both cases, only buoys are reported; however, these areas remain susceptible to the arrival of other types of colonized litter.

The origin of litter colonized by epibionts can be tracked in specific scenarios. For instance, in Italy and Portugal, it has been reported that 64% of the colonized litter originated from aquaculture activities (Rech et al., 2018b). Aquaculture items are easily recognizable while considering that the study by Rech et al. (2018b) was carried out in those areas. However, associating the occurrence of colonized litter with specific anthropic activities may be a rather difficult task on most occasions. For instance, while plastic bottles are generally assumed to originate from land (e.g., incorrectly discarded by beachgoers), studies suggest that a significant number of PET bottles are dumped by ships off shore (Ryan et al., 2019; Ryan, 2020; Ryan et al., 2021), but a definitive number cannot be attributed to either source.

A factor that plays an important role in the colonization process, although it is generally underestimated, is the polymeric composition of the litter (normally plastics). Experimental studies have previously shown that some invertebrate species, such as bryozoans, have a preference for plastics with multiple polymer compositions (Li et al., 2016; Pinochet et al., 2020; Póvoa et al., 2022). A total of 13 different polymers have been identified in the literature, including blends. However, in field studies, there is still great uncertainty regarding the preference of observed organisms for different types of polymers. Only five studies were found that identified the polymeric composition of plastic litter. De-la-Torre et al. (2021) tried to relate the organisms found at the taxonomic class level with the polymers or materials that make up the substrates they inhabit. However, their results are not enough to be conclusive. Similarly, Rech et al. (2021) identified the polymer from the floating plastic debris found. However, the number of litters analyzed was not sufficient to evaluate this factor as a predictor variable. Subías-Baratau et al. (2022) carried out a more exhaustive identification, including a greater number of litters analyzed and reported a greater variety of polymers than previous studies. The three studies agree that the two predominant types of polymers are polyethylene (PE) and polypropylene (PP). This makes sense since these are the two types of polymers most produced and traded worldwide (PlasticsEurope, 2021). Other polymers found are polyethylene terephthalate (PET), which could be attributed to bottles and some textiles, polyamides (PA), which could originate in fishing nets, among others. One possible reason for the low use of polymeric identification techniques is the cost and scope of sophisticated equipment and analysis, such as Fourier Transform Infrared Radiation (FTIR) spectrometry, particularly in developing countries (Silva et al., 2018; Aragaw, 2021). However, more exhaustive monitoring of the polymers that transport marine organisms is necessary at a global level. Secondly, Cooke and Sumer (2021) reported the presence of Lepas pectinata (Spengler, 1793) in the neck of a plastic bottle. The particularity of this finding is that, after a contact angle analysis, the neck of the bottle presented higher hydrophilicity compared to the rest of the bottle. Although more in-depth studies are needed regarding the physicochemical characteristics of the colonized litter, the study by Cooke and Sumer (2021) preliminarily put into perspective the relevance of materials science aspects in this field of research.

The present review focused on the role of marine litter in the transportation of colonized floating marine litter (rafting), which could harbor invasive or potentially invasive species. Other anthropic sources of dispersal may include biofouling boat hulls and ballast water (Costello et al., 2022), as well as natural pathways. Natural dispersal pathways have been demonstrated on several occasions. For

instance, non-native cnidarian [e.g., Rhopilema nomadica (Galil, 1990)] and fishes [e.g., Lagocephalus sceleratus (Gmelin, 1789), and Pterois miles (Bennett, 1828)] have been found in the Mediterranean Sea as a result of drifting and swimming pathways (Deidun et al., 2011; Coro et al., 2018; Galanidi et al., 2018). Comparing the multiple anthropic and natural dispersal pathways is a complicated task as these may occur simultaneously, although more attention has been given to biofouling and ballast water pathways (García-Gómez et al., 2021). This is likely due to the increase and exacerbation of biological invasions associated with anthropic activities, as well as the capacity of transoceanic dispersal (e.g., international maritime activity) (Encarnação et al., 2021). Attributing specific pathways to already triggered biological invasions is extremely difficult if active monitoring of the various anthropic and natural vectors is lacking. On the other hand, ecological research must support determining if non-native organisms are harmful invasive species for specific ecosystems. Whether a certain dispersal pathway is more or less harmful than others may be debatable. However, constant monitoring is required to understand species dispersal dynamics and elaborate control and conservation programs, including natural pathways.

# **4** Recommendations

Based on the results of this review and identified limitations, several recommendations are proposed:

- In many cases, identifying organisms to the lowest taxonomical level by just applying morphological characteristics is insufficient, thus, it is recommended to carry out molecular analyzes of the predominant species that show traits of being an AIS. Considering the difficulty to access techniques, such as DNA barcoding, research groups are encouraged to seek collaborative projects with mutually beneficial objectives.
- In light of the lack of a standard marine litter classification, the item categorization according to Fleet et al. (2021) is recommended. Additionally, digital approaches are required to facilitate data collection in the field. Further, the item categories indicated by Fleet et al. (2021) should be taken as standardized litter categories master list, while maintaining some flexibility for local-level litter peculiarities. A clear example is the great number of laminated candy wrappers, which are specific to cities like Lima, Peru. On the other hand, specific events that induce unusual and unprecedented sources of marine litter should also be taken into account. For instance, during the COVID-19 pandemic, it would be relevant to include an additional category referring to face masks or personal protective equipment (such as gloves and face shields).
- The overall number or abundance of marine litter at each sampling site, regardless of their colonization status, should be taken as a reference frame. Thus, fouling litter surveys must report the percentage of fouled items with respect to the total number of litters at each sampling location.
- Comprehensive studies should include species preference analyses based on the chemical characteristics of a

substantial part of all the biofouled items. For instance, in the case of plastics, identifying the polymer type through FTIR or Raman spectroscopy is recommended, as well as the hydrophilicity of the material and surface microstructure.

# 5 Conclusions

The growing literature on floating marine litter have evidenced its indubitable ability to serve as a substrate for multiple organisms, possibly acting as a vehicle for species dispersal, including AIS. In the present review, an updated literature search was carried out focusing on field research reporting the occurrence of marine macro-biota colonizing or inhabiting marine litter. Results indicate that, from a geographic point of view, there is a lack of information on African and East Asian countries, as well as on the open ocean. Methodologies used to investigate colonized litter in coastal areas lack standardization, thus, making studies difficult to compare. Also, a consensus is needed regarding litter classification and unit expression, as well as combining litter classification with polymer identity. On the other hand, the difficult access to species identification through molecular analyzes and incomplete libraries are important barriers to precisely estimating the contribution of marine litter to AIS dispersal. Based on the main limitations identified in the present review, a list of recommendations was proposed.

# Author contributions

GD-l-T: Conceptualization, Methodology, Investigation, Project administration, Writing – original draft, Writing – review and editing. MA and VR: Conceptualization, Methodology Investigation, Writing – original. AP and TW: Writing – review and editing, Data curation, Validation. All authors contributed to the article and approved the submitted version.

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# Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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