Universitat Autònoma de Barcelona

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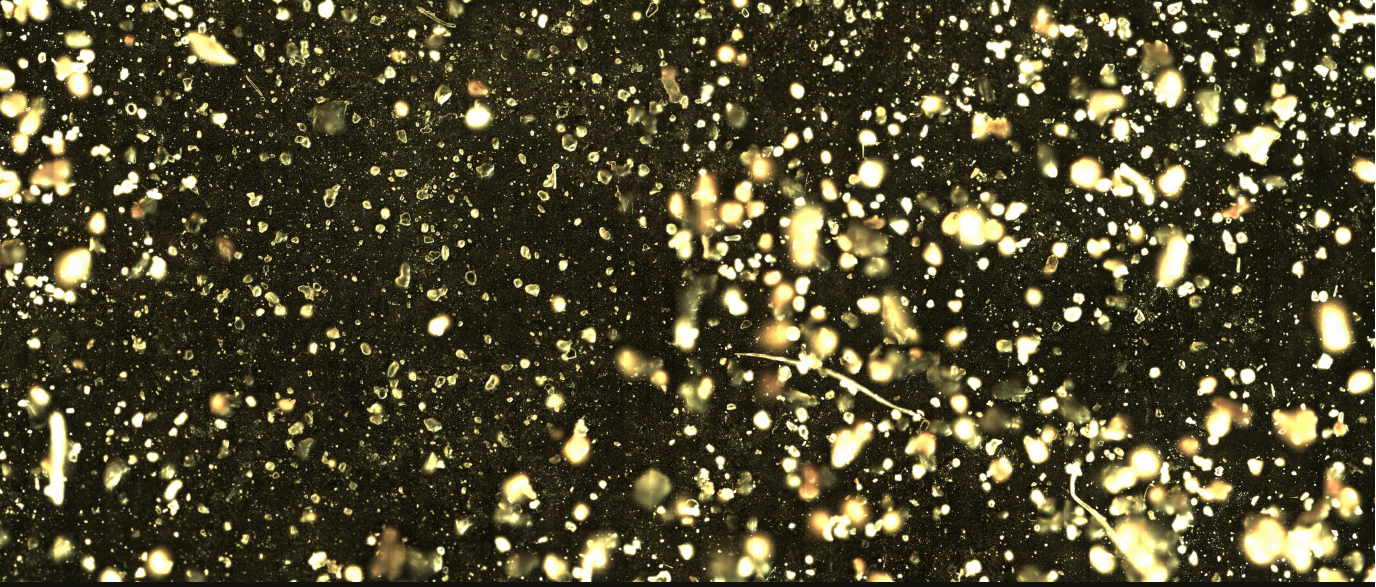
Laura Simon Sánchez



Doctoral Thesis 2022

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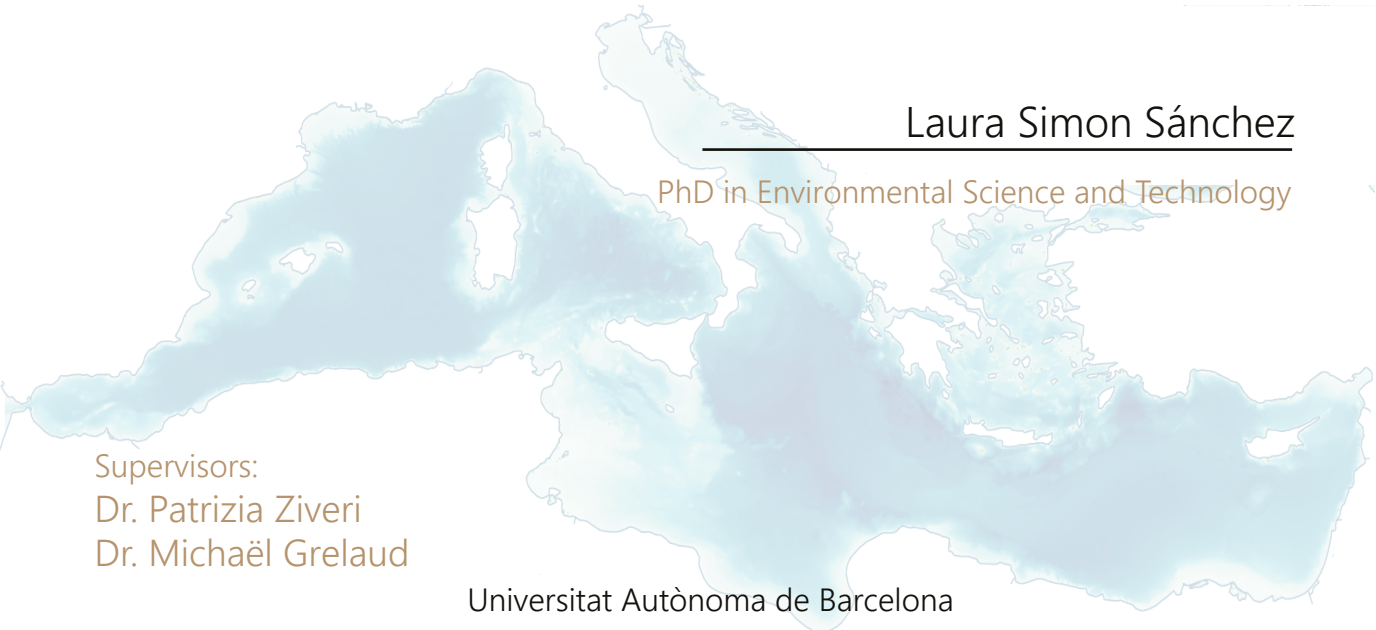
PhD in Environmental Science and Technology



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“Far and away, the greatest threat to the ocean, and thus to ourselves, is ignorance. But we can do something about that.”

Sylvia Earle

Abstract

In less than a century, plastic materials have become an indispensable part of our societies. Their diversity, versatility, and durability promoted their success. Besides, the reasonable low price of their manufacturing rapidly led to an exponential growth in their global production. This tendency is analogous to the pace at which plastics are discarded. Contrary to their manufacturing process, recycling these materials requires high economic and technological investments, resulting in prioritizing the use of new resources over the circular economy approach. All in all, their rapid production, in combination with the poor policies and low investments to cope with plastic waste, has resulted in these pollutants accumulating in the natural environment. Plastic pollution represents one more of the many anthropogenic stressors already threatening our planet. Concern over plastics, specifically microplastics, is severe when entering, accumulating, and interacting in the marine environment. These pollutants represent an increasing risk, from damaging at the individual cellular level to altering the ecosystem's function. The research efforts focus on providing evidence on microplastics' presence, dispersion, and fate across different environmental compartments to understand ultimately the exposure and hazard levels these pollutants pose. Further efforts transmitting the scientific findings to society about how rapidly and evidently plastics are changing our oceans have captured the general society's attention, encouraging and assisting in developing policies based on real-time data that lead toward marine protection. The main objective of this thesis is to contribute to this mission.

Considering that policies are based on current scientific findings, the robustness of the results and conclusions is crucial. The microplastic research community is dealing with a lack of standardization on sampling, sample preparation, and identification methodologies which jeopardizes the accurate comprehension of the microplastic pollution issue at global or regional scales. This thesis contributes to the interpretation of the microplastic research in the abiotic

compartments of the Mediterranean Sea, one of the greatest accumulations areas of (micro)plastic debris. A detailed literature review and interpretation of the methods and occurrence of microplastics are discussed and compared to the state-of-the-art methods and global knowledge of microplastic research. The results show how different methodologies shape our current understanding of these pollutants' occurrence, dispersion, and fate in the Mediterranean Sea. The study also highlights the future perspectives of microplastic research in the basin while claiming the need for an inter-basin collaboration to strengthen the efforts to address this demanding challenge.

Besides, this thesis aims to provide insights into the dispersion and fate of microplastics in transitional environments, the interface between land and marine environments. These areas play a crucial role in understanding the microplastic fluxes entering the marine environment, and potentially, they act as depositional areas preventing these pollutants from reaching the marine realm. In particular, three chapters of this thesis investigate the microplastic occurrence and properties along different Mediterranean beaches and in the Ebro Delta (NW Mediterranean Sea). The Mediterranean beaches, every year, attract millions of tourists promoting and sustaining the development of the local economies; however, the environmental and social impacts are not trivial. The study evaluates the seasonal variation of micro-litter as an effect of tourism on a large geographical and time scale. In seven Mediterranean islands, twenty-one beaches subjected to different anthropogenic pressure were selected and sampled four times during the high and low touristic seasons. The results showed a seasonal variation as an effect on the increase of visitors to the beaches, but the predominance of fibers emphasized the role of atmospheric deposition as a source of anthropogenic litter. The study also shows the challenges of monitoring micro-litter at high resolution on a large geographical scale, providing recommendations for further efforts. The studies in the Ebro Delta targeted i) the estuary to investigate the dispersion of the microplastic across the different environmental compartments and ii) the prodelta of the river

to assess the accumulation of microplastics over time in this depositional environment. The first study's findings showed the Ebro River's role as a source of microplastics to the Mediterranean Sea. Still, relevant concentrations of microplastics found in the benthic sediments indicated their potential as accumulation areas. The second study elaborated on these conclusions, aiming to assess the rate at which microplastics are sequestered on the seafloor, and their fate once buried within the sediments. To address these questions, the presence of microplastics was investigated in a sediment core combining palaeoceanographic approaches and microplastic analytical methods. The presence of microplastics in the sediments of the Ebro prodelta has increased over time, showing the sequestered mass of small microplastic (< 1mm) a similar exponential trend to global plastic production. Besides, the results indicated that microplastic pollutants remain preserved in the sediments, with no further degradation. Overall, the results reported in this dissertation provide further evidence of the increasing accumulation of microplastics in the marine environment, emphasizing the benthic sediments' role as a major reservoir of these pollutants.

Resumen

En menos de un siglo, los materiales plásticos se han convertido en una parte esencial de nuestras vidas. El éxito de estos materiales reside en su diversidad, versatilidad y durabilidad, pero también en el relativo bajo coste de su producción. Esto ha causado un crecimiento exponencial en la producción global de estos materiales. Consecuentemente, al mismo ritmo que estos materiales se producen, se descartan. Pero contrariamente a los procesos de producción, los de reciclaje y revalorización de los residuos plásticos requieren altas inversiones, tanto económicas como tecnológicas. Lo que ha derivado en la priorización del uso de nuevos recursos en lugar de seguir un modelo de economía circular. La combinación de su incesante producción con limitadas políticas de gestión de revalorización y reciclaje de los residuos, y las bajas inversiones para hacer frente a los residuos plásticos ha resultado en que estos residuos se están acumulando en el medio natural. La contaminación por plásticos se suma como uno más de los numerosos impactos antropogénicos a los que nuestro planeta está sometido. El problema de la contaminación por plásticos, específicamente microplásticos, es severo porque estos contaminantes están llegando al medio marino, donde se acumulan, interaccionando con diversos y numerosos organismos. Estos contaminantes representan un riesgo emergente, afectando desde individuos a nivel celular hasta alterar el funcionamiento de ecosistemas. Actualmente, los esfuerzos de investigación se centran en proporcionar las concentraciones en las que se encuentran estos contaminantes en diferentes matrices ambientales, pero también en su dispersión y destino ambiental, para en ultima estancia entender los niveles de exposición y amenaza ambiental que los microplásticos representan. Además, la comunidad científica está centrada en transmitir los hallazgos científicos a la sociedad, demostrando lo rápido y evidentemente que estos contaminantes se están acumulando y afectando a nuestros océanos. El impacto de estos esfuerzos ha derivado en una creciente preocupación ambiental, lo que está alentando y facilitando el desarrollo de políticas basadas

en *"datos a tiempo real"* que promueven la protección del medio marino contra este tipo de contaminantes. En este contexto se centra el principal objetivo de esta tesis, contribuir con datos empíricos a la misión de proteger nuestros océanos de la contaminación por microplásticos.

Teniendo en cuenta que el desarrollo de estas políticas se basa en los hallazgos científicos actuales, la solidez de los resultados y las conclusiones es crucial. El inconveniente, es que el campo de investigación de contaminación por microplásticos es incipiente, por lo que la comunidad científica se enfrenta a la falta de estandarización en las metodologías de muestreo, preparación de muestras e identificación, lo que limita la comprensión precisa del problema a escala mundial o regional. Esta tesis contribuye a la interpretación de la investigación sobre microplásticos en los compartimentos abióticos del mar Mediterráneo, una de las mayores áreas de acumulación de residuos (micro)plásticos. Esta contribución se llevó a cabo mediante una revisión detallada de la literatura, proporcionando una visión general de las metodologías empleadas en esta región y los niveles de concentración de microplásticos reportados en el Mar Mediterráneo. Los resultados muestran cómo las diferentes metodologías están afectando a la comprensión actual de la ocurrencia, dispersión y destino ambiental de estos contaminantes en la cuenca del Mar Mediterráneo. El estudio también destaca las futuras perspectivas de la investigación de microplásticos en esta región. Al mismo tiempo, el estudio reafirma la necesidad de una colaboración internacional, entre los países del Mediterráneo, para fortalecer los esfuerzos científicos y hacer frente al exigente desafío que representa la contaminación plástica en esta región.

Además, esta tesis tiene como objetivo proporcionar información sobre la dispersión y el destino ambiental de los microplásticos en zonas de transición, que representan la interfaz entre el medio terrestre y marino. Estas áreas representan un papel crucial en la comprensión de los flujos de microplásticos que llegan al mar, pero también estas áreas representan potencialmente zonas de acumulación que capturan estos contaminantes evitando que lleguen al

medio marino. En concreto, tres capítulos de esta tesis se centran en la investigación de la presencia y propiedades de los microplásticos a lo largo de diferentes playas del Mediterráneo y en el Delta del Ebro, Noroeste del Mar Mediterráneo. Las playas del Mediterráneo, cada año, atraen a millones de turistas promoviendo y sustentando el desarrollo de las economías locales; sin embargo, los impactos ambientales y sociales del turismo no son triviales. Nuestro estudio evaluó la variación estacional en la presencia de micro-residuos, plásticos y fibras de origen antropogénico, como consecuencia del turismo. El estudio se llevó a cabo cubriendo una gran escala geográfica y temporal. En siete islas del Mediterráneo, se seleccionaron veintiuna playas sometidas a diferentes grados de presión antropogénica. Las playas se muestrearon cuatro veces, durante las diferentes temporadas turísticas. Los resultados muestran una variación estacional que sigue el aumento de visitantes a las playas durante la temporada alta. Sin embargo, el predominio de las fibras, sobre otro tipo de partículas, resalta el papel de la deposición atmosférica como fuente de basura antropogénica. El estudio también muestra los desafíos de monitorear los niveles microplásticos con una alta resolución cubriendo una gran escala geográfica y temporal, proponiendo recomendaciones para ayudar en futuros esfuerzos de monitoreo en la presencia de microplásticos. Los estudios en el delta del Ebro se centran en i) el estuario donde se investiga la dispersión de los microplásticos en diferentes matrices ambientales y ii) el prodelta del río, con el fin de evaluar la acumulación de microplásticos en las últimas décadas. Los hallazgos del primer estudio muestran el papel del río Ebro como fuente de microplásticos al mar Mediterráneo. Aunque las altas concentraciones de microplásticos encontradas en los sedimentos bénticos indican el potencial del estuario como áreas de acumulación para estos contaminantes. El segundo estudio ahonda en estas conclusiones, con el objetivo de evaluar la tasa a la que los microplásticos se secuestran en el fondo marino y su destino ambiental una vez enterrados en los sedimentos. Para abordar estas cuestiones, se investigó la presencia de microplásticos en un testigo de sedimento marino, combinando métodos del campo de la

paleoceanografía y los métodos analíticos de vanguardia del campo de los microplásticos. Los resultados muestran que la presencia de microplásticos en los sedimentos del prodelta del Ebro ha aumentado con el tiempo, mostrando que la masa secuestrada de pequeños microplásticos (<1mm) sigue una tendencia exponencial similar a la tasa de producción global de materiales plásticos. Además, los resultados indican que estos contaminantes se preservan en los sedimentos, sin señales de mayor degradación con el paso del tiempo. En general, los resultados de esta tesis manifiestan la evidencia de la creciente acumulación de microplásticos en el ambiente marino, enfatizando el papel de los sedimentos bentónicos como un importante reservorio de estos contaminantes.

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Aims and thesis structure

The microplastic (MP) research field is commonly stated in its infant stage. A daunting number of studies have emerged in the last decades to shed light on their occurrence, dispersion, and potential threats (Everaert et al., 2020; Ivleva et al., 2017; Van Sebille et al., 2020), yet major knowledge gaps about the microplastic fluxes, dispersion, and fate in the marine environment remain. The rapid growth of scientific publications in the field comes with a drawback: studies are developed without a standardized framework regarding the methodologies for sampling, characterizing, and quantifying these pollutants. While this has hampered the intercomparison of studies, it also allowed to develop different techniques, advancing technical and scientific knowledge in the field. During the timeframe this thesis was conducted (2018-2022), it sought to evolve with the field and the state-of-the-art methods.

The specific objectives of this thesis were:

- To understand the dynamics of microplastic pollution in the transitional system, between land and marine environments for the accumulation and transfer of microplastics.
- To assess the seasonal pattern of microplastic variability under different degrees of anthropogenic pressure.
- To characterize the microplastic properties and identify potential sources and mechanisms driving the microplastic fate across different environmental compartments.

The study focus of this thesis is the Mediterranean Sea. This basin is recognized as one of the greatest accumulation areas for plastic pollution due to the combination of its hydrodynamic conditions and high anthropogenic pressure.

This Ph.D. dissertation is a compilation of scientific articles, with a general introduction to the research topic, a synthesis chapter compiling the research outputs and the future perspectives of microplastic pollution. The research

articles represent four manuscripts; two of them were published (Chapter 2, 4), one was submitted (Chapter 5), and one is in preparation (Chapter 3).

i. **Chapter 1-** Introduction

This chapter provides the reader with a general background on defining microplastic pollution, the sources and fate of these global pollutants in the marine environment, and the relevance of quantifying their presence across different environmental compartments. This is followed by a brief description of the Mediterranean Sea as a hotspot for microplastic pollutants.

ii. **Chapter 2-** Are research methods shaping our understanding of microplastic pollution? A literature review on the seawater and sediment bodies of the Mediterranean Sea.

In this chapter, the scientific literature reporting the microplastic presence in the abiotic compartments of the Mediterranean Sea, published until December 2020, was reviewed. The differences and similarities in the methods used to define and characterize the microplastic pollution levels in the basin were evaluated and critically discussed with implications for the microplastic research carried out on a global scale. The outcomes of this work were published in Environmental Pollution in October 2021.

iii. **Chapter 3**- Large geographical survey of micro-litter in Mediterranean beaches following a touristic gradient

In this chapter, large geographical and temporal distribution of micro-litter under an anthropogenic gradient has been targeted. The occurrence of micro-litter was evaluated in beaches located in Mediterranean islands subjected to three different degrees of anthropogenic pressure under a seasonal variability.

iv. **Chapter 4-** River Deltas as hotspots of microplastic accumulation: The case study of the Ebro River (NW Mediterranean)

In this chapter, one of the largest river delta systems in the western Mediterranean Sea, the Ebro River Delta, has been studied to define its role as a source and sink of microplastics to the Mediterranean Sea by quantifying and

characterizing microplastic particles across different environmental compartments. This work was published in Science of the Total Environment in June 2019.

v. **Chapter 5-** Sediment cores revealing the plastic age? – Microplastic preservation in coastal sedimentary records

This chapter combines different analytical approaches to reconstruct the microplastic accumulation in the sedimentary records of the Ebro prodelta. Besides, microplastic fate was investigated by assessing the variability in the size distribution and the weathering status of the polyolefins, polyethylene, and polypropylene. This study has been recently submitted for publication in Environmental Science and Technology.

vi. **Chapter 6**- Conclusions

This chapter synthesizes the main results and conclusions of this thesis. Besides, the prospects of microplastic pollution are outlined.

CHAPTER 1

Introduction

1. Plastic materials- *from bright inventions to ubiquitous pollutants*

Plastics were a promising alternative to scarce natural materials (e.g., ivory). Since the serendipitous creation of Bakelite, the first fully synthetic plastic polymer, by Leo Hendrick Baekeland in 1907, numerous and diverse synthetic plastics have emerged. During the Second World War, the versatility and durability of plastics did not pass unnoticed, which provided the momentum to advance their development (Crawford and Quinn, 2017). Rapidly, their success led to an exponential growth in plastic production, turning these materials into an integral part of our daily lives, from covering conventional commodities to leading medical and technological advances. Unfortunately, the design and end-of-life plastic treatments have not evolved at the same pace that plastics are produced and discarded. About one-third of the plastic is made to meet single-use standards and disposed of within a year (Koelmans et al., 2014). As of 2015, 8300 million tons of plastic had been manufactured, but only 24.1% of this plastic mass was still in use (Geyer et al., 2017). The remaining 6300 million tons were already discarded under the end-of-life scenarios of recycling (9.0%), incineration (12.0%), and accumulation in landfills or the natural environment (79%; Fig. 1; Geyer et al., 2017). Concern over the mismanaged plastic waste entering the aquatic environments is severe since the long-term environmental

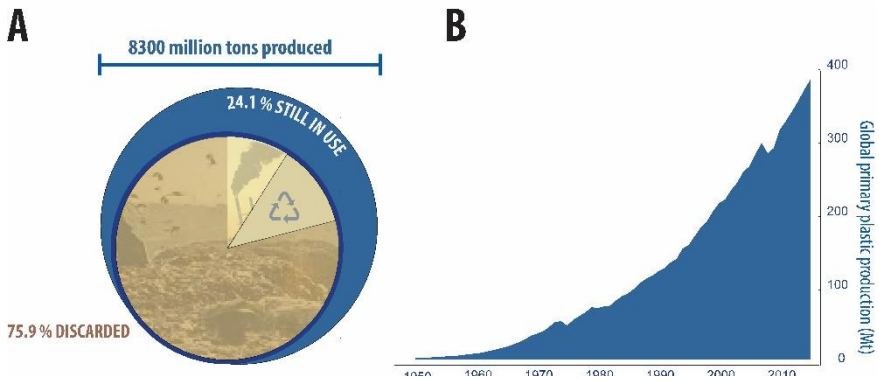

Fig. 1. 1. A. Fate of plastic produced as of 2015. **B.** Global plastic production 1950-2015. Adapted from Geyer et al., 2017. Landfill picture retrieved from National Geographic, author Randy Olson.

effects of plastic pollutants, specifically on the micro-and nano-size scale, remain largely unknown (SAPEA, 2019).

2. Microplastics – *defining microplastic pollutants*

The presence of synthetic fibers and small floating plastics was reported in the coastal waters of Northumberland, North Atlantic (Buchanan, 1971), and the Sargasso Sea (Carpenter and Smith, 1972), already back in 1970. The term 'micro-plastic' appeared for the first time in the scientific literature in 1990, when Ryan & Moloney recorded the temporal increase in the presence of micro-and macro-plastics stranded on the beaches of South Africa. However, these reports gained minimal attention from the scientific community. The social and scientific awareness of plastics accumulating in the oceans grew later with the discovery of the North Pacific Garbage Patch by Charles Moore, reporting relevant concentrations of neustonic plastic (Moore et al., 2001). Furthermore, the publication of Thompson et al. (2004) is generally considered as the trigger of the microplastic research field. In their work, Thompson and colleagues recovered the term 'microplastic' to describe the occurrence of microscopic plastics in marine sediments and the increasing abundance of these pollutants in archives of neustonic samples (1960-1990) collected in the North Atlantic (Thompson et al., 2004). Nevertheless, the term microplastic was not defined until 2008, when international environmental scientists were gathered at the research workshop "*on the Occurrence, Effects, and Fate of Microplastic Marine Debris*" organized by the NOAA. The participants agreed on the definition of microplastics as *"small plastic particles smaller than 5 mm"* (Arthur et al., 2009). The experts already foresaw the limitations of the definition but considering the early stage of the research field, they considered appropriate a broader definition that would redefine with the advancement of science and technology instead of restricting the scientific freedom (Arthur et al., 2009). Indeed, a decade later, the research community pursued a definition that encompasses the complex, multifaceted properties of microplastic pollutants (Frias and Nash, 2018; Hartmann et al., 2019). Still, experts differed

on the maximum bound size. Frias and Nash. (2018) proposed an inclusive size definition, from 1-5000 µm, contributing to the ongoing research efforts to monitor and compare microplastic pollution levels. Whereas Hartman et al. (2019) set the size limits following a pragmatic approach based on the International System of Units (SI) nomenclature, the prefix micro- refers to the scale 1-1000 µm. The relevance of finding consensus on a common definition goes beyond the scientific debate, as a standardized and relevant definition of microplastic is crucial for regulatory bodies to tackle the challenges of these pollutants not only in the natural environment but also in our food and indoor spaces (Brennholt et al., 2018; Hartmann et al., 2019). In this dissertation, I followed the definition proposed by Frias and Nash (2018), "*Microplastics are any synthetic solid particle or polymeric matrix, with regular or irregular shape and with size ranging from 1 µm to 5mm, of either primary or secondary manufacturing origin, which are insoluble in water"*. In this context, the upper limit was set at 5 mm, whereas the methodological approaches defined the lower limit of the particles characterized.

Microplastics are multifaceted pollutants whose physico-chemical properties and characteristics fall within a broad spectrum that can be categorized (Hartmann et al., 2019; Rochman et al., 2019).

2.1. Chemical composition

The primary and fundamental classification relies on the chemical identification of the particles to corroborate their synthetic or polymer matrix composition. Then, the categories are as vast as the diversity of plastic materials. Each plastic material has specific properties attributed to the arrangements of the atoms in their complex long chemical structures. Breaking down the complexity, plastics are composed of single molecules, monomers, that bond together to form large chains, known as polymers. The process of binding monomers is called polymerization. The polymerization may be: i) chain-growth, the monomers are added to each other, retaining all the atoms, or ii) step-growth, the functional group of the monomer reacts with the functional

group of another monomer, forming a molecule in the process (Crawford, chapter 4). If identical monomers are connected during the polymerization, the resulting polymer is classified as a homopolymer, whereas bonding more than one type of monomer produces heteropolymers, generally known as copolymers. Some typical homopolymers are polythene (PE), polypropylene (PP), polyvinyl chloride (PVC), while some typical copolymers are acrylonitrile-butadiene-styrene (ABS), ethylene-vinyl acetate (EVA), and styrene-butadiene rubber (SBR). Besides, the architecture of the polymers can have a linear, branched, or cross-linked structure (Edmondson and Gilbert, 2017). Thermoplastics, the most conventional and commercial plastics available, have a linear or branched structure that allows the materials to flow above certain temperatures but still recover the solid state at ambient temperature. On the contrary, thermoset plastics, with a cross-linked structure, are chemically and irrevocably changed at high temperatures, preventing them from melting down to be re-shaped.

Often referred to as commodity plastics, are petroleum-based plastics with a wide range of applications. Their success, durability, and versatility lead to the rapid production of these plastics. Comparably, they are the most common type reported as marine plastic debris. Under this category are Polyethylene terephthalate (PET), Polyethylene (PE-low density and PE-high density), PVC, PP, Polystyrene (PS), Polycarbonate (PC), Poly(methyl methacrylate) (PMA), Polytetrafluoroethylene (PTFE), Acrylonitrile butadiene styrene (ABS), Polyamide (nylon) and Polychloroprene (neoprene). Noteworthy, the advancements in biodegradable plastics, those synthesized from renewable feedstocks, have brought new polymers to the market, the most common and widely used are polycaprolactone (PCL), Polylactic acid (PLA), and Polyhydroxybutyrate (PHB) (Crawford and Quinn, 2017).

Finally, different chemical additives improve the plastic materials' properties during their production. There are several classes of additives: retardants, plasticizers, antioxidants, dyes, fillers, and stabilizers. While they do not define

the polymer composition, in some cases, additives represent a considerable weight of the material (i.e., PVC) (Hartmann et al., 2019; Rochman et al., 2019). The additives concentration in environmental microplastics is generally unknown, and this represents a challenge when elucidating the fate and impacts of the particles in the marine environment.

2.2. Origin

Microplastics are categorized as primary and secondary (Cole et al., 2011). Primary microplastics are manufactured and used in size range of 1- 5000 µm. Under this category, among others, are included virgin pellets, the raw material to produce plastic items; microbeads for the use of personal care products; and plastic beads used as abrasive media (e.g., cleaning ships' hull). Secondary microplastics are the result of the breakdown of larger plastic items. This process can occur in the natural environment where plastic items are exposed to physical, chemical, and biological processes that may lead to their degradation and fragmentation. However, these microplastics can also be originated from the use and tear of plastic materials, e.g., clothing, fibers are shed during laundering (Browne et al., 2011; De Falco et al., 2019; Napper and Thompson, 2016), as well as wearing them (De Falco et al., 2020); tire wear particles produced because of the abrasion of the tires and brakes against the road (Kole et al., 2017). As far as potential toxicological effects are concerned, categorizing microplastics based on their origin makes little difference; however, it plays a role for legislation and regulatory bodies to allocate responsibilities (Hartmann et al., 2019).

2.3. Morphology- shape, and structure

Microplastics are often classified according to their shape. The common categories considered are fibers, fragments, films, foams, beads, and pellets (Hidalgo-Ruz et al., 2012). These categories are often combined, and terms are used interchangeably. For example, the categories "pellet" and "bead," while the first term relates to the feedstock of plastic materials, the second describes the scrubbers from personal care and industrial products (Helm, 2017). This

issue can be addressed by describing the geometrical structure of the particles. Similarly, the category "fragment" encompasses vast geometric structures that, in some cases, are further described as rounded, angular, rough edges (Hidalgo-Ruz et al., 2012) but remain ambiguous. Because of this, the term "fragment" was suggested to be replaced by "irregular particles" (Hartmann et al., 2019). While the shape of the particles provides an overview of the type of particles encountered in the media, uttermost importance is the relevance of their physical behavior in the natural environment and the potential ecotoxicological effects of specific microplastic shapes (Hartmann et al., 2019; Rochman et al., 2019). Besides, the microplastic shape can indicate the potential source of the particle. Cózar et al. (2015) distinguished fibers in fishing threads (those originating from fishing activities) and textile fibers to discriminate potential contamination during sampling and sample processing. Nevertheless, categorizing the microplastics' shape is time-consuming and subject to the analyst's impression. Moreover, the size of the particles and/or the analytical methods may limit the possibility of visually allocating them to a shape category. Then, it may be resourceful to use the aspect ratio of the particles, as described in Vianello et al. (2019), who applied µFT-IR imaging spectroscopy to determine microplastics from air samples and discern between fibers and fragments by calculating the ratio between the length and width of the particles.

2.4. Color

Categorizing the color of microplastics is a common practice in the studies of microplastic pollution. Color is the property of the objects to emit or reflect light. In the marine environment, the exposure to light wildly varies. The vivid colors on the surface vanish with depth as the light decreases, as well the perception of color is different across species, even individuals (Allen, 2009). These concepts are essential when identifying potential predator-plastic prey relations under environmental conditions. Besides, the color of plastics changes under photo-oxidative agents. Weathering leads to discoloration and a shift in tonality to yellowish colors. Turner and Holmes (2011) reported that the

yellowish and darkening tones of plastic pellets stranded at the beaches of Malta, Mediterranean Sea, correlated with an increase in the carbonyl index, hence photooxidation. The lack of robust protocols and the limitations associated with determining the color category are driven by i) observer's subjectivity and ii) the methodological conditions (i.e., light exposure during microplastic characterization) and were recently addressed by Martí et al. (2020). In their work, Martí and colleagues proposed a semi-automated method using a reference color palette to categorize the color of plastics and emphasized the potential of color as a qualitative proxy to describe the age of plastic in the marine environment.

The relevance of providing data regarding the microplastic particles, chemical composition and morphology, is because the microplastic's properties influence to a certain degree their physical behavior and fate within the marine environment. Besides, there should be considered that the physical properties of microplastics might change under environmental mechanisms, which further challenge our current understanding of the occurrence, dispersion, and consequence of these pollutants accumulating in our oceans.

3. Relevance of microplastic pollution research

Plastic pollution is a global and intradisciplinary issue (Villarrubia-Gómez et al., 2018). The pace plastic materials are produced and discarded surpasses our current capabilities to evaluate not only the risks when these materials end up accumulating in the environment but also the potential adverse effects of manufacturing them in the first instance (Persson et al., 2022). Irretrievably, the unceasing plastic consumption of the last decades showed an overwhelming and unforeseen wide range of social, economic, and environmental impacts. Hence, the scientific efforts are critical to point toward a sustainable design, production, use, and disposal of plastics. Moreover, there is the responsibility to understand how plastics already in the natural environment affect ecosystems, from global processes to cellular levels.

Oceans are largely the final destination of mismanaged plastic waste. In particular, plastic pollution in the marine environment has drawn significant scientific attention. Plastics entering the marine environment break down into smaller fractions, microplastics and nanoplastics, whose both potential ecotoxicological effects and knowledge gaps increase as their size decreases (Covernton et al., 2019; Mitrano et al., 2021). Even if sustainable management of plastics is achieved, microplastics will be a persistent problem as their presence progressively increases over time (Barnes et al., 2009). There is a pressing need to understand their occurrence, pathways, behavior, and fate to ultimately address the hazard and exposure levels.

4. Sources, transport, and the fate of microplastics in the marine environment

4.1. Sources

Most of the plastic debris entering the marine environment is land-based sourced (Derraik, 2002; Li et al., 2016). The main pathway for plastic waste to reach the oceans are direct emissions in coastal areas (Jambeck et al., 2015) and rivers (Lebreton et al., 2017a; Meijer et al., 2021; Schmidt et al., 2017). As of 2010, Jambeck et al. (2015) estimated the direct influx between 4.8 million and 12.7 million metric tons of plastics to the global ocean, accounting for inputs within the first 50 km of the coast. The river plastic outflow were estimated to range between 0.8 million and 2.7 million metric tons per year (Meijer et al., 2021). Additionally, extreme weather events (i.e., hurricanes, tsunamis, floodings) may contribute to transporting plastic items to the coastal marine environment and the open ocean pelagic systems. These plastic inputs are considered a primary source of microplastics as their weathering and fragmentation lead to the production of secondary microplastics. Another relevant pathway of microplastics to the aquatic environment, and subsequently to the ocean, are allocated to wastewater treatment plant (WWTPs) effluents (Horton et al., 2017b; Talvitie et al., 2015). Recent studies showed the efficiency of secondary and tertiary plants in removing microplastics but

emphasized the large number of microplastics still emitted (Gatidou et al., 2019). Moreover, the unequal technological development and limitations of wastewater treatment facilities around the globe should be considered. Other sources can be allocated to the run-off dragging microplastics from terrestrial to aquatic environments (e.g., tire wear particles) (Goßmann et al., 2021), as well the atmospheric fall-out can contribute to the entrance of microplastics into the marine environment (Allen et al., 2020; Dris et al., 2016).

In contrast, 20% of marine plastic debris finds its origin in ocean-based sources, where fishing activities represent the major contribution (Li et al., 2016). Other sources are related to the illegal dumping of plastic waste or the loss of plastic products during transportation due to spillage or sinking of the vessels.

4.2. Transport and fate of microplastics in the marine environment

Microplastic fate can be described based on physical, chemical, and biological behavior (Crawford and Quinn, 2017; Wang et al., 2016). These behaviors are generally interconnected, and the synergetic effect of the microplastic characteristics and different environmental processes drive their fate in the marine environment. Remarkably, this dissertation's outputs contribute to understanding the physical behavior of microplastics in transitional environments (Chapter 3,4,5) and the chemical behavior, precisely the degradation status of microplastics buried in the sediment compartment (Chapter 5).

4.2.1. Physical behaviour

Migration (Micro)plastics entering the ocean can be effectively dispersed from the littoral zone to the open ocean. The convergence zones of the oceans, so-called subtropical gyres, are recognized for the **accumulation** of (micro)plastics, but particles do not remain indefinitely trapped in these areas. The oceanographic processes that control the transport of floating plastics on the ocean's surface are described in detail in Van Sebille et al. (2020).

Beaching, as early stated, most of the (micro)plastic inputs to the marine environment occur in the coastal areas. The transport model of positive buoyant plastics described by Onink et al. (2021), showed that on the global average about 48% of the beached plastics are sourced locally, and about 25% of the plastic inputs are trapped within the first 50 km of coast. Similarly, the model described by Kaandorp et al. (2020) addressing the plastic budget of the Mediterranean Sea, emphasized the relevance of coastal areas as depositional environment for plastic pollutants. Their results showed that the coastline retains between 49-63% of the estimated plastic inputs (Kaandorp et al., 2020). While these models addressed a wide spectrum of plastic sizes, the validation of the data was contrasted against environmental studies reporting the occurrence of microplastics (e.g.: Collignon et al., 2012; Gündoğdu, 2017; Güven et al., 2017; Zeri et al., 2018). Once in the beaches, the direct exposure of plastics to UV-light accelerates the chemical degradation (*Section 4.2.2*), inducing the physical **fragmentation** of plastic particles into smaller particles.

Export of microplastics from the sea surface to the seafloor can be induced by diverse sinking processes that transport microplastic pollutants along the water column. However, there is still a major knowledge gap on the removal mechanisms of microplastics from the surface to the bottom of the ocean (Egger et al., 2020; Van Sebille et al., 2020), as there are few *in situ* measurements addressing the occurrence of microplastics in the water column of the ocean (Choy et al., 2019; Pabortsava and Lampitt, 2020).

Sedimentation, some microplastics are expected to sink in the marine environment because of the commonsense approach that microplastics made of polymers with negative or neutral buoyancy in seawater do not float. However, as previously stated, microplastics are complex pollutants. Their ratio volume size, presence of additives, and weathering status may also affect their sinking potential. Indeed, microplastics with higher density than seawater have been found in the open surface ocean (Erni-Cassola et al., 2019). Similarly, buoyant polymers such as polyethylene and polypropylene are often reported in benthic

environments (Erni-Cassola et al., 2019; Int-Veen et al., 2021). The latter finding is likely a consequence of the interaction of microplastics with biota. Microplastics may be i) ingested-egested in fecal pellets, ii) trapped within organic and inorganic matter aggregates (e.g., marine snow), iii) colonized by microorganisms which cause an increase in the particle density, accelerating the export from the surface to the benthic environment, and also microplastics can be iv) transported within the vertical migration of marine species (de Haan et al., 2019; Van Sebille et al., 2020). Noteworthy, not all the microplastic reaching the marine sediments are **sequestered** in this compartment. Microplastics can be **remobilized** as bottom currents are responsible for erosion, transport, and deposition of sediments and microplastics in the deep sea (Kane et al., 2020; Kane and Clare, 2019). Similarly, sediments in submarine canyons are exposed to ephemeral gravity currents that contribute to the microplastic resuspension and transport in the benthic environment (Pohl et al., 2020).

4.2.2. Chemical behavior

Degradation is a chemical change induced by environmental factors that reduce the polymer's average weight affecting its properties (Andrady, 2011; Wang et al., 2016). Extensive degradation leads to the embrittlement of the plastic, triggering the **fragmentation** process. There are different environmental-specific processes that might cause the chemical change in the polymer. Biodegradation is induced by biological organisms, such as fungi, bacteria, or insects, which can deteriorate plastic materials (Crawford and Quinn, 2017) In the marine environment, the primary mechanisms, generally combined, driving the degradation of plastics are photooxidation, light-induced, and thermoxidative degradation caused by oxidation under moderate temperature (Andrady, 2011).

Adsorption, while plastics are inert materials, the additives added during the manufacturing process might drive the toxicity of these pollutants. Microplastics can adsorb and concentrate persistent organic pollutants (POPs)

and act as vectors to transport these chemical pollutants (Teuten et al., 2009). Consequently, the pollutants contaminating the microplastics can be desorbed. The **desorption** of the pollutants adhering to microplastic surface after ingestion may lead to their diffusion across different biological tissues (Crawford and Quinn, 2017).

4.2.3. Biological behavior

The biological behavior of microplastics is the interaction of these pollutants with marine life.

Biofouling is the development of a biofilm of microorganisms that may evolve into colonies of invertebrates, depending on the particle size (Andrady, 2011). This process might lead to the introduction of alien species, e.g., the presence of bacteria from WWTPs into the marine environment (Pedrotti et al., 2021).

The suspension of microplastics in the water or accumulation on the sediment compartment exposes a wide range of marine organisms to their **ingestion**. Moreover, the size range of marine microplastics overlaps with the prey's size of copepods (Rist et al., 2020) at the base of the marine food web. Experimental studies showed a high rate of **egestion** of these pollutants (Klein et al., 2021; Ward et al., 2019; Woods et al., 2018), although environmental studies reporting the occurrence of microplastics in marine biota reported relatively high abundances of anthropogenic particles, specifically fibers (Carreras-Colom et al., 2020). Moreover a recent study showed that the microplastic ingestion-egestion process from the freshwater amphipod *Gammarus duebeni* leads to their **fragmentation** into smaller particles (Mateos-Cárdenas et al., 2020). The retention of microplastics within the digestive tract is cause of concern due to the potential **translocation** into other tissues, the release of absorbed pollutants, and the biomagnification effects through the **trophic transfer** as a consequence of predation.

5. Microplastics in the Mediterranean Sea

The Mediterranean Sea is a semi-enclosed marginal sea, landlocked by the southern region of Europe, Western Asia, and Northern Africa. The basin lies between latitudes 30°- 45°N and longitudes 5°W- 36°E. The average depth of the Mediterranean basin is 1500 m, with a maximum depth of 5267 m in the Calypso Deep in the Ionian Sea. The only natural connection to the ocean is the narrow Strait of Gibraltar that opens on the Alboran Sea to the southwest and connects the Mediterranean Sea to the Atlantic Ocean. The Strait of Sicily (37.2°N 11.20°E) with its two sills subdivide the sea into the Eastern and Western basins. The general circulation of the Mediterranean Sea has been previously described (Béthoux, 1979; Hopkins, 1985; Pinardi et al., 2004; Pinardi and Masetti, 2000; Rohling et al., 2009). Briefly, it is characterized by anti-estuarine thermohaline circulation. The surface inflow of the Atlantic waters moves eastward, increasing its temperature and salinity. The evaporation exceeds precipitation resulting in eastern Mediterranean seawaters characterized by saltier and heavier waters that sink in the Levantine Basin to form the Levantine intermediate waters when colder winter temperature further increases the surface water density. From there, the thermohaline circulation extends to different regions with the formation of the Western Mediterranean Sea deep waters and the outflow at the Strait of Gibraltar. Besides the inflow of Atlantic waters, the water budget of the Mediterranean is affected by river discharge and precipitation (Struglia et al., 2004).

The Mediterranean region hosts about 6.7% of the global population, where nearly 150 million live by the coast, and more than 240 million are concentrated in the surrounding river basins (European Environment Agency, 2014; UNEP, 2020). Besides, the region receives millions of visitors every year, which gives it the title of one of the top world-leading touristic destinations (UNWTO, 2018). The basin also concentrates heavy industrial and marine activities, concentrating about 15-30% of the global shipping traffic (Piante and Ody, 2015; UNED-MAP, 2012).

For centuries, the Mediterranean Sea has been subject to high anthropogenic pressure (Coll et al., 2010; Lotze et al., 2011). The high population growth and intense use of resources, accelerated since the Industrial Revolution, resulted in changing environmental conditions boosted, among others, by climate change, habitat loss, pollution, and eutrophication (Coll et al., 2010; Karkanorachaki et al., 2018). The synergetic effect of these threats poses a risk to Mediterranean biodiversity (Coll et al., 2012). Despite its relatively small global surface representation (0.82%), this basin hosts 4-18% of the global biodiversity (Bianchi and Morri, 2000). The ubiquitous and increasing presence of microplastics has captured the research attention to understand the effects of this anthropogenic threat on Mediterranean biodiversity (Deudero and Alomar, 2015; Llorca et al., 2020 and references therein.). Emblematic species of the Mediterranean Sea, like the basking shark (*Cetorhinus maximus*) and the fin whale (*Balaenoptera physalus*), were proposed as sentinels to represent the microplastic levels in the pelagic environment (Fossi et al., 2014). The deep-sea shrimp (*Aristeus antennatus*) and Norwegian lobster (*Nephrops norvegicus*) were proposed as bioindicators for the benthic environment (Carreras-Colom et al., 2020; Cau et al., 2019). Additionally, microplastic ingestion was reported in relevant commercial species for human consumption: European sardine (*Sardina pilchardus*), anchovy (*Engraulis encrasicolus*), red mullet (*Mullus barbatus*), bogue (*Boops boops*), mussel (*Mytilus galloprovincialis*), spiny oyster (*Spondylus spinosus*) (Digka et al., 2018; Garcia-Garin et al., 2019; Kazour et al., 2019; Pennino et al., 2020; Renzi et al., 2019; Rodríguez-Romeu et al., 2020). The ecotoxicology effects of microplastic ingestion under natural environmental conditions remain unclear (Anbumani and Kakkar, 2018; Hamm and Lenz, 2021). Thus, it is crucial to systematically quantify and characterize the microplastic presence in the abiotic and biotic systems to comprehend their distribution, dispersion, and accumulation across the different environmental compartments.

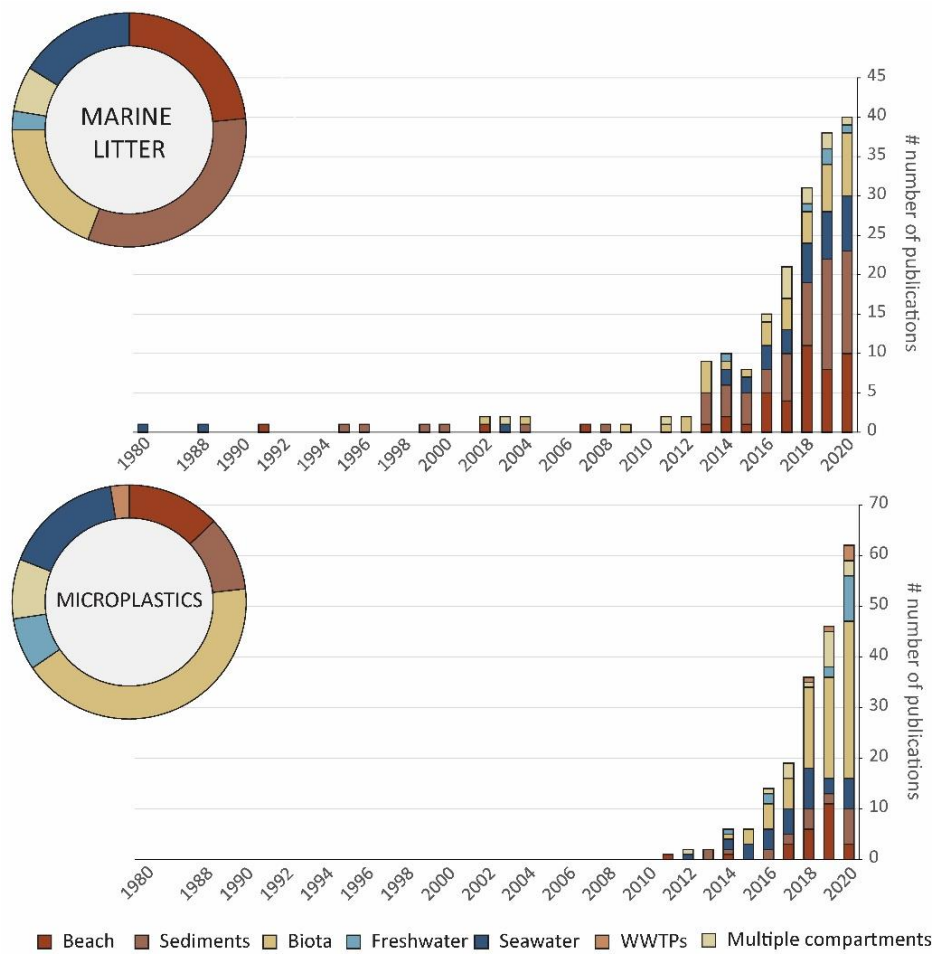

Fig. 1. 2. Comparison on the number of scientific publications of macro-litter and microplastics in the Mediterranean Sea. Data from SCOPUS and Web of Science using the search strings: A. ("marine debris" OR "Marine litter" OR "macro-litter" OR "plastic debris") AND "Mediterranean"; and B. ("microplastics" OR "microplastic pollution") AND "Mediterranean".

Indeed, numerous and growing research efforts have been conducted in the Mediterranean Sea to address (micro)plastic pollution from organism to environmental compartments (Fig 1.2.). Overall, the studies point to the Mediterranean Sea as one of the greatest accumulation areas of floating microplastic debris (Cózar et al., 2015; Lebreton et al., 2012; Suaria et al., 2016), surpassing levels that already pose an environmental risk (Everaert et al., 2020). However, major research gaps and challenges still remain, partly due to methodological limitations.

In this dissertation, Chapter 2 revises the scientific efforts to characterize the microplastic pollution in the abiotic compartments of the Mediterranean Sea. It highlights the limitations and significant research challenges to be addressed in the future to provide comprehensive knowledge on the occurrence, fate, and consequent impacts of microplastic pollutants in the Mediterranean basin. While the Chapter 3, 4 and 5 contribute, with empirical data, to the assessment of microplastic accumulation and dispersion levels, providing further understanding in their dynamics and fate across different environmental compartments.

CHAPTER 2

Are research methods shaping our understanding of microplastic pollution?

A literature review on the seawater and sediment bodies of the Mediterranean Sea

Abstract

The lack of standardization on the definition and methods in microplastic (MP) research has limited the overall interpretation and intercomparison of published data. This has presented different solutions to assess the presence of these pollutants in the natural environment, bringing the science forward. Microplastics have been reported worldwide across different biological levels and environmental compartments. In the Mediterranean Sea, numerous research efforts have been dedicated to defining the MP pollution levels. The reported MP concentrations are comparable to those found in the convergence zone of ocean gyres, pointing to this basin as one of the world's greatest plastic accumulation areas. However, to what extent are the data produced limited by the methods? Here, we present the results of a systematic review of MP research methods and occurrence targeting the seawater and sediment bodies of the Mediterranean Sea. Based on this dataset, we 1) assess the discrepancies and similarities in the methods, 2) analyze how these differences affect the reported concentrations, and 3) identify the limitations of the data produced for the Mediterranean Sea. Moreover, we reaffirm the pressing need of developing a common reporting terminology, and call for international collaboration between Mediterranean countries, especially with North African countries, to provide a complete picture of the MP pollution status in this basin.

Keywords: Microplastic, Mediterranean Sea, methods, seawater, marine sediments, beach

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1. Introduction

The presence of small plastic particles, microplastics (MPs), in the marine environment has been documented since the 1970s (Buchanan, 1971; Carpenter and Smith, 1972; Morris, 1980; Ryan and Moloney, 1990; Shiber, 1979). However, scientific and public concern have increased over the last 20 years once the accumulation and environmental effects of plastics in the environment were evident (Barnes et al., 2009; Derraik, 2002; Thompson et al., 2004). Since then, growing efforts are being devoted to characterize the presence and risk that these pollutants might pose to the natural environment and human health (Dick Vethaak and Legler, 2021; SAPEA, 2019). Consequently, the MP research field is constantly evolving, and different definitions and methodologies are being applied. Microplastics are generally defined as plastic particles smaller than 5 mm (Hidalgo-Ruz et al., 2012; Moore, 2008). This definition, restricted to the size criterion, overlooks one of the most valuable characters of plastic materials: their diversity. Recent proposals to standardize the MP definition consider their physiochemical properties, shape, size, and origin (Frias and Nash, 2018; Hartmann et al., 2019), although authors even differ on the size criterion (1 to <5000 µm - Frias and Nash, 2018; 1 to <1000 µm - Hartmann et al., 2019). Despite the lack of standardization, the MP research community is aware of the pressing need of working towards the harmonization of the data, reporting results using a common terminology (Hartmann et al., 2019; Provencher et al., 2020; Rochman et al., 2019) that would allow the large-scale interpretation and comparison of current knowledge.

In the Mediterranean Sea, numerous research efforts have been dedicated to define the plastic pollution issue. The reported MP concentration, modeled (Eriksen et al., 2014; Lebreton et al., 2012; Van Sebille et al., 2015) and empirically gathered (Cózar et al., 2015; Suaria et al., 2016), are comparable to those found in the convergence zone of the subtropical gyres, indicating that this basin is one of the world's greatest plastic accumulation areas (Cózar et al.,

2015; Lebreton et al., 2012). The high levels of MP pollution and its accumulation are likely driven by a combination of high anthropogenic pressure and the hydrodynamic conditions of the Mediterranean Sea (Morris, 1980). The Mediterranean region is inhabited by 480 million people, where one-third is concentrated along the coast, and about one-half is living in the surrounding hydrological basins (European Environment Agency, 2014). The basin supports intensive fishing, shipping, and industrial activities, and it is one of the world's top tourist destinations (UNWTO, 2018). Furthermore, the anti-estuarine circulation of the Mediterranean Sea contributes to this phenomenon by its limited outflow (Rohling et al., 2009). The Strait of Gibraltar is characterized by the inflow of Atlantic surface waters of relatively lower salinity, while the outflow of more saline and denser waters is restricted to deeper depth. Thus, floating plastic debris entering into, or generated within the Mediterranean Sea, will be trapped within the basin with few possibilities of escape (Aliani et al., 2003; Lebreton et al., 2012; Morris, 1980).

The high spatio-temporal variability of the Mediterranean Sea circulation prevents permanent accumulation areas of floating MP debris (Mansui et al., 2015). Most of the plastics are assumed to float at sea because of their positive buoyancy, as their average specific density (ρ= 0.9- 1.0 g cm^{-3}) is lower than seawater (ρ= 1.027 g cm^{-3}); except for few denser polymers (i.e., PET, ρ= 1.38 g cm^{-3}; or PVC, ρ= 1.39 g cm^{-3}) which are expected to sink once entering into the aquatic environment. Ocean turbulence induces the vertical transport of positively buoyant MPs along the water column (Kooi et al., 2016; Kukulka et al., 2012), yet despite this, the seafloor is considered the long-term sink of MP pollution in the marine environment (Woodall et al., 2014). The export of MPs from the sea surface to the deep sea can be facilitated by the formation of biofouling on the particle's surface, the sequestration of these pollutants into organic and inorganic aggregates, or transportation through the vertical migration of numerous organisms (Van Sebille et al., 2020 and references therein). In a recent study, Kaandorp et al. (2020) estimated the floating plastic

budget of the Mediterranean Sea: their results showed that 37-51% of the plastic input to the basin tend to settle on the seafloor and that 49-63% of these inputs end up beaching. Hence, the role of beaches cannot be neglected, as these transitional environments between terrestrial and marine systems act simultaneously as pathways and dynamic storage of MP pollutants.

Previously, authors have provided a general overview of the abundance of MPs in Mediterranean rivers (Guerranti et al., 2020), surface waters (Cincinelli et al., 2019), sediments (Martellini et al., 2018), and their interaction with biota (Llorca et al., 2020). However, none of the studies have discussed in depth the discrepancies in the methodologies and how these might be defining the current understanding of MP pollution in the Mediterranean basin. In this context, our approach for this paper is to: 1) Review and compile the available literature of MP occurrence in the seawater and sediment bodies of the Mediterranean Sea and 2) Describe the methods used for sampling, extracting, and identifying MPs, including the measures to prevent airborne contamination and quality control procedures. The main aims are to summarize how the differences and similarities in the methods influence the observed abundances of MP in the Mediterranean Sea, to outline where the research efforts are focused, and to identify the pressing research needs that will enhance our understanding of the fate of MP within this basin.

2. Methods

A systematic literature review was conducted to integrate the MP pollution data and the associated methods in the seawater and the sediment compartments of the Mediterranean Sea. Two scientific databases (Web of Science, www.webofknowledge.com; SCOPUS, www.scopus.com) were consulted, integrating logical operators, through the specific string search: ("microplastics" OR "microplastic pollution") AND "Mediterranean". The search was limited to English peer-reviewed articles published before January 2021. The first article selection was performed after a screening of both title and abstract. Only the articles investigating the concentration of MPs in

sediment and seawater bodies were considered for full-text review. This set of articles was then classified according to the environmental compartment they investigated: sea surface water, seawater column, marine sediments, and/or beach. For the articles in which more than one category was investigated, each compartment was included in the dataset as an independent study. The meeting criteria for inclusion after the full-text revision was that the study provided primary results on the concentration of MPs in at least one of the four targeted compartments of the Mediterranean Sea (Fig. 2.1).

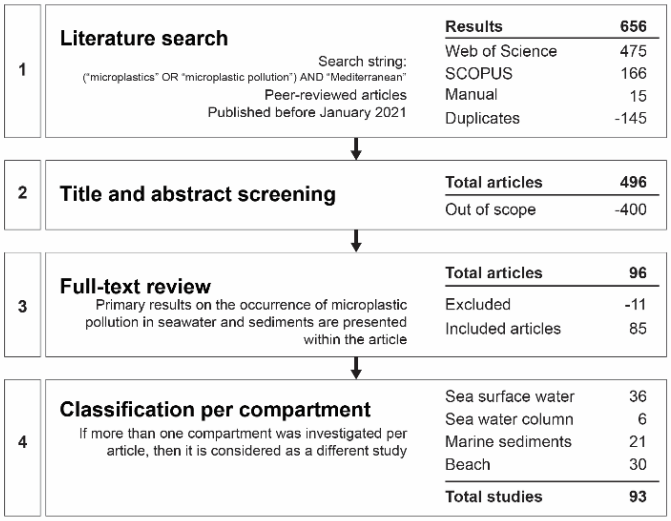

Fig. 2. 1. Flow diagram illustrating the selection process and systematic review of the published literature.

From each included study, the following information was extracted: the environmental compartment targeted, the description of the methods for MP sampling [e.g., date, instrument and its characteristics, flowmeter, wind correction, number of samples, sample preservation], extraction [e.g., sample's volume, purification treatment, flotation treatment parameters- brine solution, mixing and settling time, repetition], identification [visual, spectroscopy methods,% of particles analyzed,% of particles confirmed as MPs], quality control and contamination protocols, the registered MP occurrence per study [e.g., geographical area, mean abundance, if available, min. and max. abundance] and per sampling station [coordinates and abundance]. When data

were missing in the publication, the Online Portal for Marine Litter: LITTERBASE (https://litterbase.awi.de/) was used to complete the dataset to acquire the geographical coordinates and abundance per station, or the corresponding authors were contacted for further information.

We used QGIS Desktop 3.12 'București' (QGIS Development Team, 2020) to map the extracted information. Sampling stations were classified as "coastal" if they located within 12 nm from the coast, and classified as "open-sea" for those exceeding 12 nm from the coast (UNCLOS, 1982). The nearest distance between sampling stations and the coast was calculated using the NNJOIN plugin (Tveite, 2019). For data visualization, figures were produced in R-3.5.3 (RStudio Team., 2020), using ggplot (Wickham, 2016), ggalluvial (Brunson, 2020) packages, and post-edited in Adobe Illustrator CC 2021 (Adobe Inc. 2021).

3. Results

The present literature review draws data from 85 articles obtained from our initial search that identified a total of 641 articles. From this first set of articles, 545 were discarded because they were duplicates or out of the scope of this review. After the full-text review, another 11 articles were discarded as they were methodological or review articles, or did not provide primary results specifically on MPs. Within the suitable articles, eight presented data from more than one compartment, extending our final dataset to 93 independent studies (Table S1), from which 36 investigated the occurrence in sea surface water, 6 within the water column, 21 in marine sediments, and 30 in beaches.

3.1. Microplastic data distribution in the Mediterranean Sea

Although MP observations are available for the entire basin (Fig. 2.2), there is a clear bias towards the Western Mediterranean Sea, with fewer studies (25.8%) investigating MP occurrence in the Eastern Mediterranean Sea. Altogether, and depending on the precision of the sampling description in the reviewed literature, there were 3077 samples in the Mediterranean Sea used to assess the status of MP pollution in its abiotic compartments (Fig. 2.3).

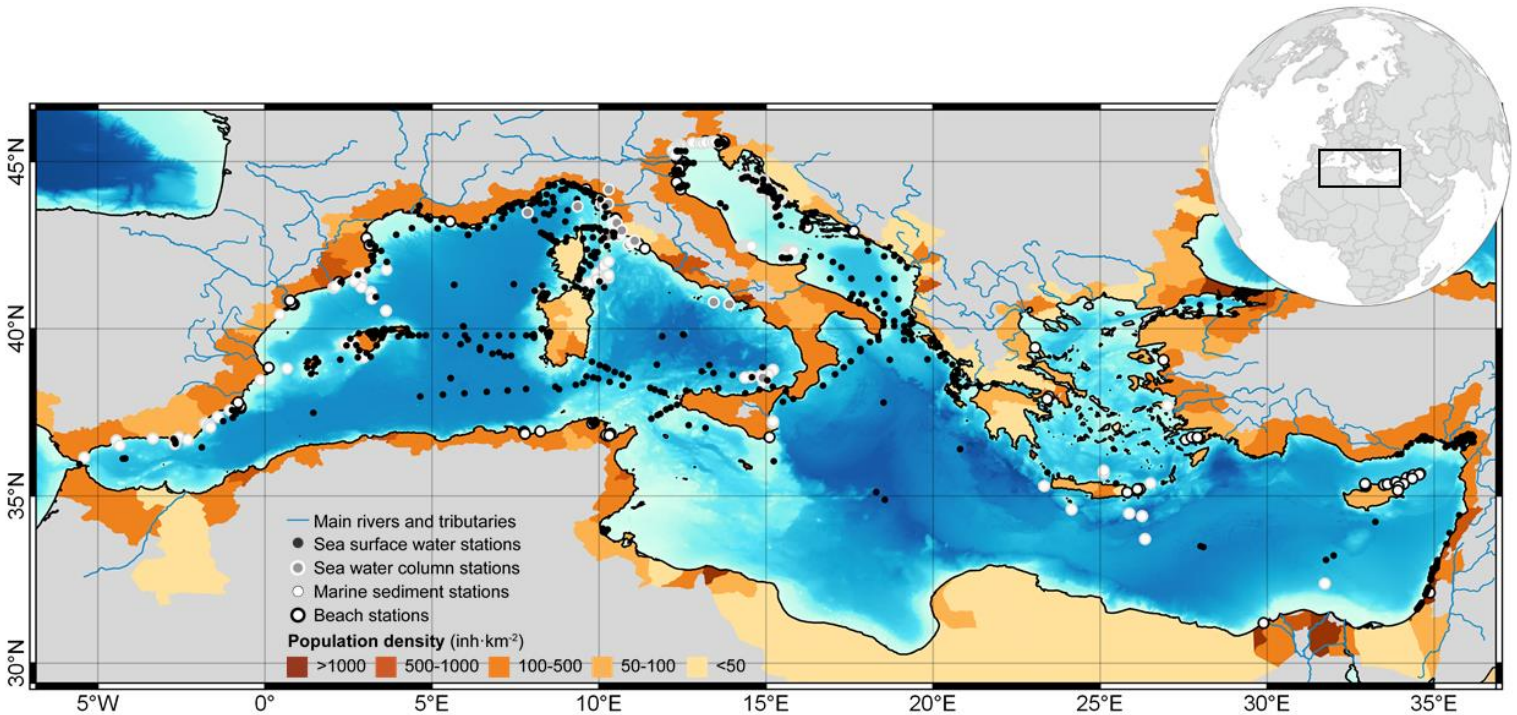

Fig. 2. 2. General map of the Mediterranean Sea. The location of sampling stations is shown in black dots for sea surface water stations, grey dots for water column stations, thin white circles for marine sediment stations, and thick black and white circles for beach stations. The population density of coastal areas is represented using an orange gradient. The population density data were obtained from several sources: Eurostat and National Statistical offices using the database City population [http://www.citypopulation.de]. NUTS3 regions and Mediterranean rivers were reprinted from Natural Earth free vector and raster map data [www.naturalearthdata.com]. The background bathymetric map was retrieved from the GEBCO 2019 grid [https://www.gebco.net/data_and_products/gridded_bathymetry_data]. Globe earth projection map showing the location of the Mediterranean Sea was created using the World Borders Dataset retrieved from thematic mapping [http://thematicmapping.org].

Most of the sampling efforts focused on the presence of these pollutants on the sea surface. Thirty-six studies collected a total of 1200 samples (40.0%). While less attention was devoted to defining the abundance of MPs within the water column, six studies collected a total of 76 samples (2.5%) restricted to the upper 100 m of the epipelagic layer. A similar picture was observed in the sediment compartment: sample scarcity is observed as the sampling sites deepen. Sediments from beaches and the neritic zone have been more extensively studied with 30 and 16 studies, collecting respectively 1302 (42.3%) and 452 (14.7%) samples. In contrast, only four studies reported MP concentration from 47 (1.5%) deep-sea sediment samples.

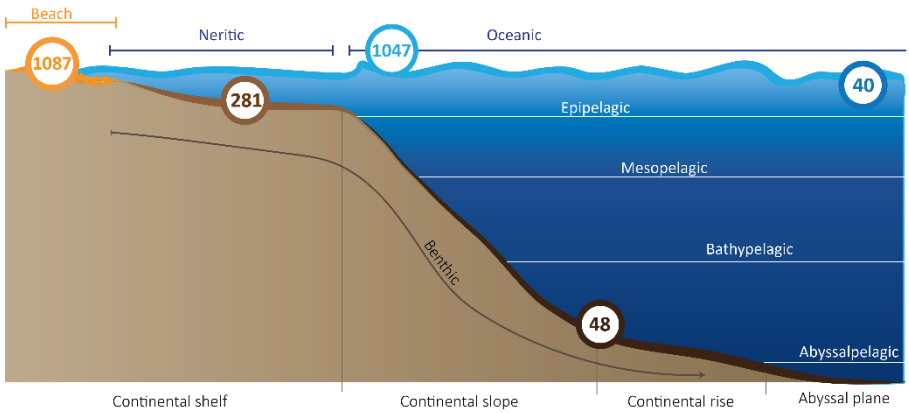

Fig. 2. 3. Distribution of sampling efforts depending on the zonation of the ocean.

3.2. Seawater

In the seawater compartment (surface water and water column: Fig. 2. 3), most of the studies collected coastal samples (77.4%), and higher efforts have been devoted to reporting the MP abundance in sea surface water (94.0%) rather than their dispersion within the water column (6.0%). Most of the samples were collected using a net (92.8%) and were volume reduced (i.e., concentrated in the cod-end of the net). Trawling speed depended on the sampling devices, currents, and weather conditions, but generally, it was adjusted between 1 and 4 knots. Reported trawling times ranged from 15 minutes up to 1 hour. Depending on the use of a flowmeter (55.4%) or not (44.6%), the absolute abundances were expressed respectively as MPs/unit of volume or MPs/unit of

surface. In the latter, the trawling distance was considered, and multiplied by the width opening of the net, under the assumption of constant seawater flux and ship speed. The authors generally reported concentration of MPs per unit of surface area (71.6%), rather than MPs per unit of volume (32.4%). Additionally, the mass of plastic debris was measured in 43.2% of the samples, and the concentrations are expressed as g MPs/unit of volume or g MPs/unit of surface (Table S2). For water column sampling, vertical or oblique tows were performed to report integrated values of MP abundance. Different mesh sizes were used to collect the seawater samples: 52 µm (0.3%), 200 µm (27.4%), 300 µm (3.6%), ≈333 µm (335, 333 and 330 µm; 62.8%), 500 µm (2.5%) and 780 µm (3.5%). Once the net is on-board, it is carefully rinsed from the outside to concentrate the sample into the cod-end. The cod-end content is emptied over a steel sieve with the same or smaller mesh size than the sampling net to avoid the loss of particles. The sample is then resuspended and transferred into the storage container. The authors reported different preservation methods. Frequently, samples were resuspended in pre-filtered seawater or deionized water and fixed with 4% formalin.

Once in the laboratory, the prevalent protocol for MP extraction consisted of manual separation of the putative MP particles under the stereomicroscope (67.0%). The identification of MPs primarily relied just on visual identification (49.3%). Further identification on the polymer composition was conducted by different spectroscopy methods (Attenuated total reflectance (ATR) - Fourier transform infrared (FTIR; 47.3%), Raman (2.0%), and Near-infrared (NIR; 1.3%)), analyzing a subsample that represented 1.2 to 100% of the putative MP particles selected under the stereomicroscope (Fig. 2. 4). When mentioned, the value of putative MP particles confirmed as MP particles after spectroscopy analyses ranged from 58.0% to 95.6%.

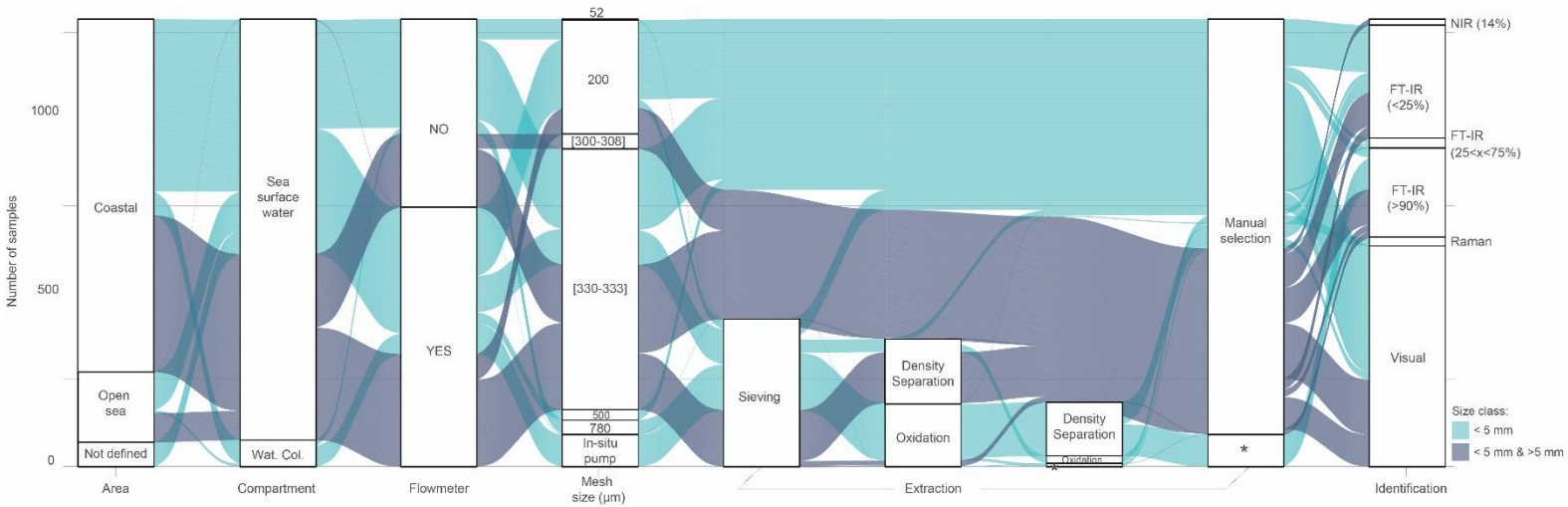

Fig. 2. 4. General overview of the sampling areas and methods for sampling, extraction processing, and characterization of microplastics in the seawater compartment of the Mediterranean Sea. The figure draws data from the 42 studies included in this review. The abbreviation DS refers to density separation, the asterisk (*) refers to the filtration process, and in the identification column, percentages are indicating the number of particles analyzed within the studies depending on the method used. The different size of plastic debris investigated by the authors is indicated in light blue (<5 mm) and dark blue (<5 mm and >5 mm).

Beyond the methodological approaches, high variability was reported for the MP pollution levels in the sea surface waters (Fig. 2. 5), the minimum concentration (200 µm mesh size, median $6.25 \cdot 10^3$ items km^{-2}; Q_1-Q_3, 0- $1.94 \cdot 10^4$ items km^{-2}) was reported in the Bay of Calvi, Corsica, NW Mediterranean Sea (Collignon et al., 2014). In contrast, the highest values were reported in the Levantine Basin, in the coastal waters of Lebanon (52 µm mesh size, median $2.24 \cdot 10^6$ items km^{-2}; Q_1-Q_3, - $1.85 \cdot 10^6$- $3.20 \cdot 10^6$ items km^{-2}; Kazour et al., 2019), Israel (333 µm mesh size, mean $1.52 \cdot 10^6$ items km^{-2}; van der Hal et al., 2017), and Turkey (333 µm mesh size, median $1.15 \cdot 10^6$ items km^{-2}; Q_1-Q_3, - $2.94 \cdot 10^5$- $5.12 \cdot 10^6$ items km^{-2}; Gündoğdu et al., 2018). In the compiled dataset (Table S2), we estimated that the MPs median concentration in the surface water of the Mediterranean Sea is $8.48 \cdot 10^4$ items km^{-2} (Q_1-Q_3, $2.89 \cdot 10^4$- $2.68 \cdot 10^5$ items km^{-2}) and the median weight is $1.0 \cdot 10^2$ g km^{-2} (Q_1-Q_3, $2.79 \cdot 10^1$- $3.63 \cdot 10^2$ g km^{-2}).

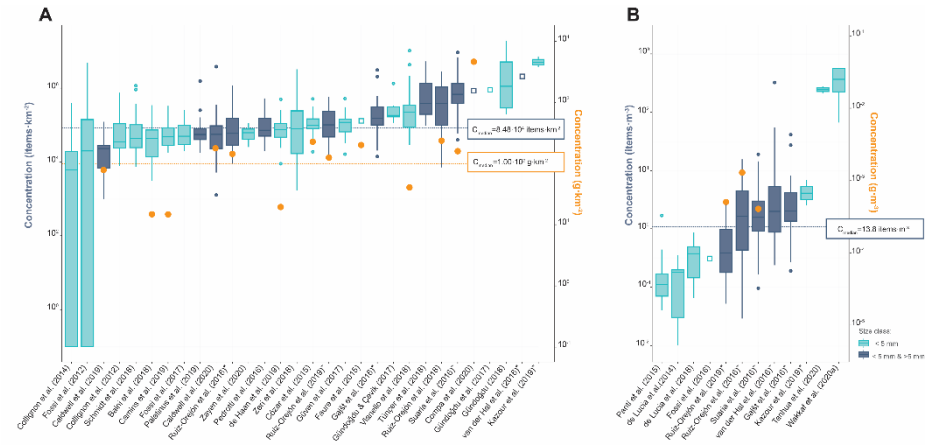

Fig. 2. 5. Overview of the concentration of plastic debris reported in the sea surface water studies (n = 36). (A) Studies reporting the concentration in items km^{-2} and g km^{-2}. (B) Studies reporting concentration in items m^{-3} and g m^{-3}. The studies marked with an asterisk (*) were plotted for both panels. The different sizes of plastic debris investigated by the authors are indicated in light blue (<5 mm) and dark blue (<5 mm and >5 mm). Note that the Y-axes are on a logarithmic scale. The squares represent the average concentration reported within the studies when data per station were not available.

3.3. Marine sediments

In the Mediterranean Sea, most of the MP observations were recorded within the coastal areas (94.4%), with the remaining 5.6% in the open sea. For shallow

and coastal sediments (<30 m), scientific scuba divers (26.7%) collected surface sediments manually (77.4%) or used hand cores (22.6%). In contrast, when the sampling was conducted from a research vessel (73.3%), Van Veen grab (76.5%), boxcorer (6.8%), multicorer (7.4%), or a mixed combination of these different devices (9.3%) were used. From these bulk sediment samples, a large volume of sediment is available, however, researchers collected a subsample of the first top centimeters (1-5 cm) and preserved it by freezing (-20º C) or cooling (4º C). It is worth to note that authors generally do not report to homogenize the sample before to further proceed with the MP extraction.

Microplastic extraction is characterized by multi-step procedures including sieving, purification, density separation, and filtration or concentration of the sample for further identification of MP particles (Hidalgo-Ruz et al., 2012). Some of these steps were applied in different combinations or individually by the authors (Fig.2. 6). Density separation was the prevalent procedure and used 77.4% of the samples. However, different settings for separation by density need to be considered: the type and volume of the brine solution, the mass of the processed sample, the mixing and settling times, and if the extraction is consecutively repeated to enhance the recovery rate. The most common brine solution was saturated sodium chloride (NaCl, $\rho \approx 1.16\text{-}1.20$ g cm^{-3}; 88.6%), followed by distilled water (H_2O, $\rho \approx 0.99$ g cm^{-3}; 6.2%;), zinc chloride ($ZnCl_2$, $\rho \approx 1.6\text{-}1.9$ g cm^{-3}; 4.7%) and sodium iodide (NaI, $\rho \approx 1.8$ g cm^{-3}; 0.5%). The mass of the sample treated varied from 10 g up to 1000 g for seabed sediment. The volume of the brine solution added to the sediment varies from 200 mL to 1000 mL. The mixing and settling time widely varied among the studies, from 30 seconds to 3 hours, and from 2 minutes to overnight periods, respectively. The extraction was repeated up to three times in 43.8% of cases. In general, the supernatant solution containing the MPs was collected and vacuum-filtered onto a filter (93.8%). In the studies where the 1-5 mm fraction was targeted (22.6%), MP particles were manually selected from the sediment.

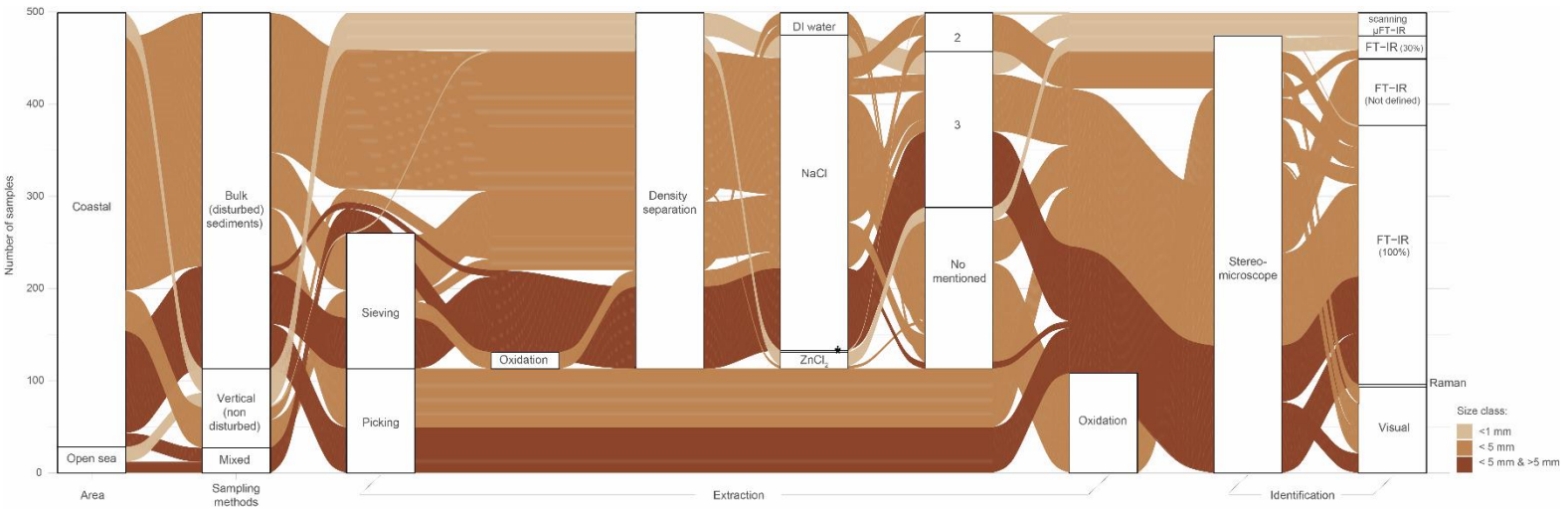

Fig. 2. 6. General overview of the sampling areas and methods for sampling, extraction processing, and characterization of microplastics in the marine sediments compartment of the Mediterranean Sea. The figure draws data from the 21 studies included in this review. From left to right, the sixth column represents the solution used for the flotation treatment, the asterisk (*) refers to NaI and DI water to deionized water, the seventh column represents the consecutive number of extractions performed during the density separation step, and in the tenth column, percentages are indicating the number of particles analyzed within the studies depending on the method used. The different size of plastic debris investigated by the authors is indicated in light brown (S-MPP; < 1 mm), brown (<5 mm), and dark brown (<5 mm and >5 mm).

In 24.4% of the samples, identification solely relied on visual identification; in 27.3% of the samples, a subsample was selected for spectroscopic analyses, and in 43.3% of the samples, all the putative MP particles were chemically characterized. Only Vianello et al. (2013) performed the identification of MPs via chemical mapping (5.0%) on a subsample of the filter.

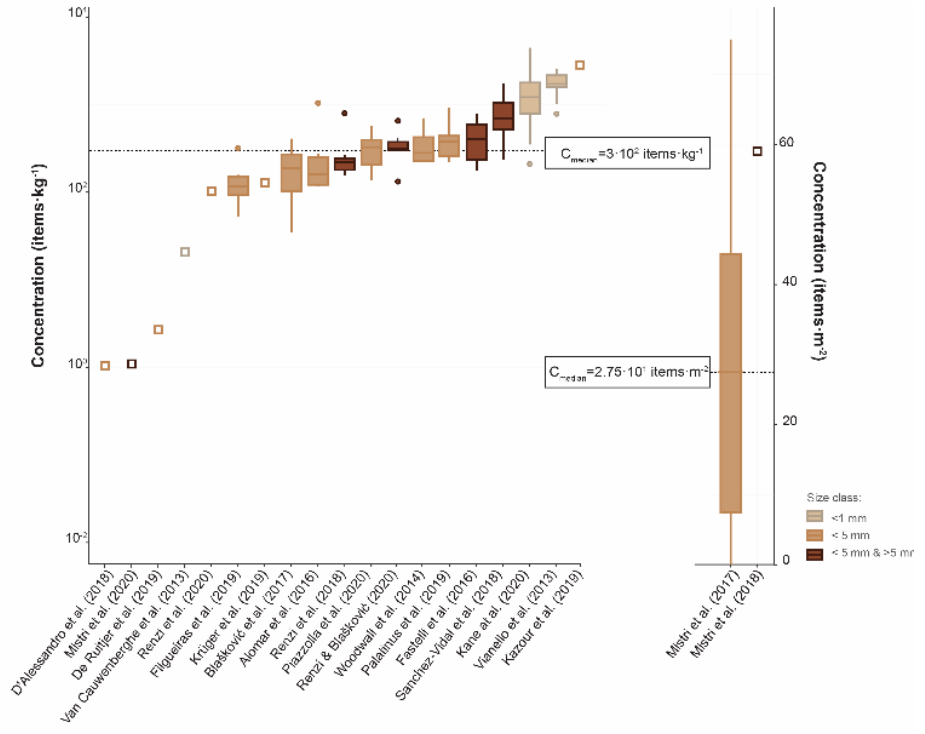

Fig. 2. 7. Concentration of plastic debris reported in the marine sediment studies (n = 21). On the left side of the figure, studies reporting the concentration in items kg−1, and on the right side, studies reporting concentration in items m−2. The different sizes of plastic debris investigated by the authors are indicated in light brown (S-MPP; < 1 mm), brown (<5 mm), and dark brown (<5 mm and >5 mm). Note that the left Y-axis is on a logarithmic scale. The squares represent the average concentration reported within the studies when data per station were not available.

The plastic debris concentration in the Mediterranean marine sediments presented high variability (Fig. 2.7), the median concentration reported is $3 \cdot 10^2$ items kg^{-1} (Q_1-Q_3, $1.49 \cdot 10^2$- $7.70 \cdot 10^2$ items· kg^{-1}). The lowest MP concentration (mean ± SD; 1.66 ± 1.77 items kg^{-1}) was reported in the Augusta Harbour, Central Mediterranean Sea (D'Alessandro et al., 2018). The highest concentration (mean ± SD; $2.43 \cdot 10^3 \pm 2 \cdot 10^3$ items kg^{-1}) was reported in the

coastal sediments of Lebanon (Kazour et al., 2019). The highest concentrations were found in the studies where the occurrence of S-MPP was investigated (Lagoon of Venice, Italy, median $1.48 \cdot 10^3$ items$\cdot$ kg^{-1}; Q_1-Q_3, $1.35 \cdot 10^3$-$1.86 \cdot 10^3$ items$\cdot$ kg^{-1} - Vianello et al., 2013; North Tyrrhenian Sea, median $1.04 \cdot 10^3$ items$\cdot$ kg^{-1}; Q_1-Q_3, $6.90 \cdot 10^2$- $1.54 \cdot 10^3$ items$\cdot$ kg^{-1} - Kane et al., 2020).

3.3. Beaches

The relatively easy access required for beach sampling has allowed researchers to widely collect sediment samples in this compartment and characterize the MP abundance along the Mediterranean coastline. However, there is considerable variability in the sampling strategies. The number of beaches sampled ranges from one to 23 in a single study that relied on citizen science (Lots et al., 2017). One study covered the entire extent of the beach, while others targeted accumulation zones or pooled samples from different tidal areas. However, most studies focus on the intertidal zone (37.0%) and the high tide line (29.6%). In the studies where the aim was to report the presence of plastic pellets (10.0%) or large microplastic particles (L-MPP; >1 mm, 6.7%;), selective sampling (3.3%), and volume-reduced sampling (13.3%) were conducted. The rest (83.3%) collected bulk sediment samples for further MP extraction in the laboratory. Samples were collected using a quadrat (56.6%), ranging from 0.04 m^2 to 1 m^2. From the surface down to a maximum of 15 cm in-depth, the top centimeters were scraped using a metal spoon, spatula, or shovel. In contrast, six studies out of the 30 collected the samples using a corer (16.6%) or boxcorer (3.3%).

Sieving was used as a pre-treatment of the sample to reduce the total volume, eliminating the particles larger than the MP size fraction (> 5 mm) in 15 studies. The majority of the studies (76.6%) used density separation to extract MPs from the sediments. The mass of sediment processed ranged from 50 g to 1000 g. Again, researchers prioritized the use of NaCl (78.3%), followed by $ZnCl_2$ (13.3%), while two studies (8.7%) combined the use of different brine solutions in the consecutive extractions to assure the recovery of denser polymers (Misic

et al., 2019; Piperagkas et al., 2019). Besides those two studies, consecutive extractions, from 2 up to 5, were performed to increase the recovery rate of particles (30.4%). The MPs particles were manually picked from the supernatant in 5 studies (21.7%), whereas in 18 studies (78.3%), the supernatant was concentrated onto a filter for further identification under the stereomicroscope. Additionally, two out of the 30 studies included a digestion step in their protocol, applied at different stages of the MP extraction process. Misic et al. (2019) performed acid digestion, adding 100 mL of HCl to the supernatant collected at the density separation step. Chouchene et al. (2019) applied alkaline digestion to the sample concentrated onto the filter, which was immersed into 50 mL of a KOH (20%) solution. When no density separation was performed (23.3%), the samples were sieved, and particles were visually selected (20%), or in one study (3.3%), a solvent extraction was performed to determine the content of polymeric and polymer-derived materials by spectroscopic techniques (Pyrolysis-gas chromatographic-mass spectroscopy (Py-GC/MS) - Ceccarini et al., 2018).

The identification of MPs relied on visual characterization in 9 studies (30.0%), differential scanning calorimetry (DSC) in one study (3.3% - Shabaka et al., 2019), and spectroscopy techniques were applied in 19 studies (63.3%). In the latter, 18 studies (60%) used ATR-FTIR on a subsample of particles; when mentioned, it ranges from a few particles to 100%, being confirmed as MPs between 11.3% and 100% of the particles. Lots et al., (2017) analyzed a subsample of particles (221 units) using the Raman spectrometry technique. Here, only 4.5% of the particles were confirmed as MPs, while the rest of the particles did not provide discernible peaks (42%), were categorized as dyes (18%), or did not provide a reliable match with the library used (36%).

In the compiled dataset, the median concentration of MPs along Mediterranean beaches was 58.6 items kg^{-1} (Q_1-Q_3, 11.6- $2.49 \cdot 10^2$ items$\cdot$ kg^{-1}) (Fig. 2.8), one order of magnitude lower than the median concentration found in marine sediments ($3 \cdot 10^2$ items kg^{-1}). The lowest abundance (particle size: <5 mm;

median $7.5 \cdot 10^{-1}$ items· kg^{-1}; Q1-Q3, $6.3 \cdot 10^{-1} – 8.8 \cdot 10^{-1}$ items kg^{-1}) was reported in Slovenian beaches (Korez et al., 2019). In this study, the authors reported a low rate of identification success, only 11.6% of the putative MPs were confirmed as MPs. The highest abundance (L-MPP; median $9.75 \cdot 10^{2}$ items· kg^{-1}; Q_1-Q_3, $6.16 \cdot 10^{2}$ – $1.55 \cdot 10^{3}$ items· kg^{-1}) was reported in the Datça Peninsula, North of Turkey (Yabanlı et al., 2019).

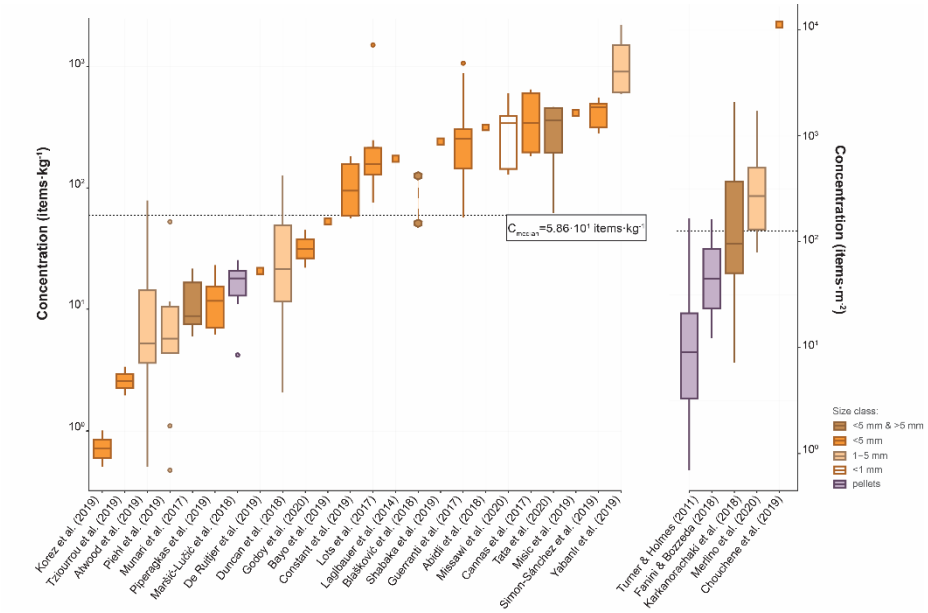

Fig. 2. 8. Concentration of plastic debris reported in the beach studies (n = 29). On the left side of the figure, studies reporting the concentration in items kg^{-1}, and on the right side, studies reporting concentration in items m^{-2}. The different sizes and types of plastic debris investigated by the authors are indicated in purple for pellets, white (S-MPP; <1 mm), light brown (L-MPP; 1–5 mm), orange (<5 mm), and dark brown (<5 mm and >5 mm). Note that the Y- axes are on a logarithmic scale. The squares and hexagons represent, respectively the average concentration and the full range of value (min. and max.) reported within the studies, when data per station were not available.

3.5. Quality control

To prevent airborne and cross-contamination of samples, especially if S-MPP and fibers are investigated, preventative measures are required, including the prioritization of using cotton clothes while sampling, the use of plastic-free and pre-cleaned storage containers, and pre-filtered solutions to resuspend the samples. The use of blanks and procedural blanks is still not standardized in MP studies. Generally, each study attempted to comply with at least some

appropriate measures to minimize contamination. For example, researchers wore cotton lab coats at the laboratory, used glass materials or stainless-steel materials, covered the samples to avoid airborne contamination, and all tools and surfaces were rinsed/cleaned (HCl (1M), ethanol or MilliQ water) before use.

For the studies on the seawater compartment, 38.1% reported the exclusion of fibers from their results due to the high risk of background contamination with this type of particle. To minimize this issue, 23.8% of the studies conducted the analysis below a laminar flow, 26.2% run blank controls (i.e., Petri dish or blank filters are exposed to laboratory conditions), and 7.1% reported to run procedural blank (i.e., MilliQ aliquot is treated as an environmental sample) to assess the cross-contamination while processing the samples.

In the marine sediment studies, 15.8% of studies performed the analysis inside of a specialized clean laboratory designed for microplastic analysis, which minimizes airborne contamination through pre-filtration of the air in the room. Blank controls were run in 42.9% of the studies, and 28.6% of the studies ran procedural blanks along with the environmental samples, to assess the potential contamination from laboratory background. Additionally, despite the use of procedural blanks, one study reported excluding fibers (<500 μm) in order to avoid potential overestimation.

In beach studies, 36.7% did not mention following any specific measure to prevent contamination. These measures were not relevant in 18.5% of studies due to the MP size investigated (L-MPP and pellets). Blank controls and procedural blanks were run in 20.0% and 23.3% of the studies, respectively. Three studies reported running spiked samples (virgin polymers) to assess the recovery rate of their protocols.

4. Discussion

In the Mediterranean Sea, numerous research efforts are focused on reporting MP occurrence in different abiotic compartments. Synthesis and integration of

these data are limited by the lack of standardization of the methods and reporting units. Notwithstanding the researchers' different approaches, similarities are found in the sampling, extraction, and identification protocols. Here, we discuss how these different approaches, rather than solely the prevention for intercomparison among studies, shape our current understanding of the Mediterranean Sea's MP pollution levels. After an extensive literature review, we have identified pressing research needs to be addressed within this basin.

4.1. The effect of sampling methods

4.1.1. Surface waters

In the Mediterranean Sea, most of the sea surface samples were volume-reduced using a net. This approach, preferably using a mesh size of 333 µm, was recommended by the Marine Strategy Framework Directive (MSFD) for the monitoring of MPs (Gago et al., 2016). The main advantage is that large volumes of water are sampled in a relatively short time. However, the mesh size of the net determines the size range and diversity of the particles. In a study of the Seine River (Paris, France) surface waters, Dris et al. (2015) found that MP concentrations measured with a mesh size of 80 µm were 30-fold greater than those collected with a 330 µm manta trawl, as a larger mesh size allows small size-class fibers (100–500 µm) to pass through it more easily. Kang et al. (2015) reported that floating MP (<2 mm) concentrations were two orders of magnitude higher using a 50 µm hand net when compared to a 330 µm mesh size net in the Southern Sea of Korea. In a comparison experiment on the efficiency of sampling devices along the North Atlantic coastal waters, Lindeque et al. (2020) demonstrated that MP concentrations collected with a 100 µm mesh size net were up to 2.5 and 10-fold greater than with a 333 µm and a 500 µm mesh size nets, respectively. Despite the large-scale-temporal variability of the compiled Mediterranean dataset (715 datapoints), we observed that lower MP abundances in the surface water were reported for larger mesh sizes (Fig. 2.9). The smallest mesh size (52 µm) was used by

Kazour et al. (2019) to collect samples from the coast of Lebanon (n=3). The predominance of waterborne MP samples collected with ≈333 µm (68.2%) and 200 µm (27.4%) mesh size nets indicate that smaller MPs (<333 µm and <200 µm) have been systematically underestimated in the surface waters of the Mediterranean basin. This is particularly relevant when considering that there is a negative relation between MP size and their abundance in the natural environment, with an increase in the number of particles accompanied by a decrease in size (Isobe et al., 2017; Pabortsava and Lampitt, 2020). Similarly and importantly, the potential environmental risk is negatively correlated with the size of the particles (Covernton et al., 2019; Ma et al., 2019).

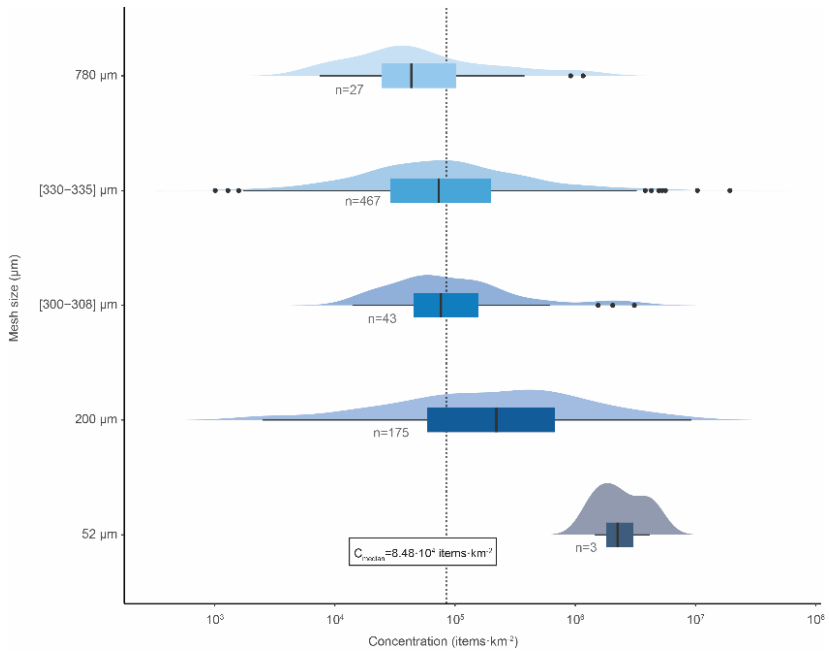

Fig. 2. 9. Violin graph showing plastic particle concentration (items km^{-2}) reported for 715 Mediterranean seawater locations, logarithmically transformed and plotted in relation to the mesh size of the net used to collect the seawater sample.

Other sampling approaches are required to account for the smaller fraction of floating MPs and potentially nanoplastics (NPs). Barrows et al. (2017) suggested implementing combined sampling, which involves collecting volume-reduced and bulk samples to ensure the characterization of a wide size spectrum of MPs (1-5000 µm). The pattern observed is that greater MP

concentrations were reported in bulk water samples (Barrows et al., 2017; Covernton et al., 2019; Green et al., 2018). Additionally, this approach allows for the assessment of fibers that generally are overlooked or discarded in the net sampling approach (Suaria et al., 2020). However, the volume and number of replicates required to provide a relevant statistical evaluation remain unclear. Ryan et al. (2020) reported that sampling a larger volume of water may increase the reproducibility of the measurement, but at the cost of underestimating the environmental concentration.

In addition to sampling devices, other factors may influence the accuracy of the measurements and possibly explain the differences between studies, such as using a flowmeter or considering the sea state. During sampling, the use of a flowmeter is critical, as it measures the water effectively passing through the net. Suaria et al. (2016) reported that the sampled area was on average two times higher when computed from GPS data compared to the same area when estimated using the flowmeter, making the GPS method a less reliable approach. During sampling, the weather and environmental conditions affect the mixing layer and, hence, MP's vertical distribution along the first meters of the water column (Collignon et al., 2012; Kooi et al., 2016; Kukulka et al., 2012). Manta trawls and neuston nets cover, respectively, the first 15-25 and 50 cm of the water column. Thus, accounting just for the surface tow concentration may underestimate MPs' total load in the surface waters, especially during high wind speed conditions (Kukulka et al., 2012).

4.1.2. Water column

Despite the numerous investigations on seawater, to date, subsurface data are still scarce. Depth integration models (Kooi et al., 2016; Kukulka et al., 2012) and multi-level trawls in the North Atlantic (150 µm mesh size, Reisser et al., 2015; and 330 µm mesh size, Kooi et al., 2016) indicated that MP concentration exponentially decreases within the first meters of the water column, yet reporting smaller sizes in deeper waters (Kooi et al., 2016; Reisser et al., 2015). In the Mediterranean Sea, few studies (n=6), always restricted to the upper 100

m of the photic zone, have investigated the MP occurrence within the water column. De Lucia et al. (2018) found higher MP concentration in coastal surface waters (0.32 ± 0.24 items·m^{-3}) of minor Italian islands compared to the abundance within the first 20 meters of the water column (0.18 ± 0.10 items·m^{-3}), although no significant variability was found. Along the Tuscany coastline, Baini et al. (2018) reported similar average concentrations between the water column (down to a maximum of 100 m; 0.16 ± 0.47 items·m^{-3}) and the floating MPs concentration at the surface waters (0.27 ± 0.33 items·m^{-3}), with the predominance of particles <1 mm. In the Gulf of Lion, Lefebvre et al. (2019) performed vertical tows from the bottom (max. depth of 100 m) to the surface, and solely fibers were found with an average abundance of 0.23 ± 0.20 items·m^{-3}. In the lagoon of Bizerte, Tunisia, seawater samples were collected using a submersible pump (300 μm mesh size; Wakkaf et al., 2020a, 2020b). The authors reported relevant high concentrations in the sea surface waters (453 ± 335 items·m^{-3}) and benthic waters (400 ± 200 items·m^{-3}). Outside the Mediterranean Sea, when similar sampling approaches were conducted (i.e., in-situ pumps) and deeper layers were investigated, the MP concentrations reported varied between 1-4 orders of magnitude higher (Choy et al., 2019; Pabortsava and Lampitt, 2020). At the Bay of Monterrey (California, USA), Choy et al. (2019) filtered a large volume of seawater (1007 to 2378 m^3) in depths ranging from 5 to 1000 m, and they found the highest MP (>100-5000 μm) concentration in the mesopelagic zone (200-600 m; 15 items·m^{-3}). In a latitudinal transect in the Atlantic Ocean, Pabortsava and Lampitt (2020) reported much higher concentrations of MPs (PE and PS, 32–651 μm; 1114 ± 542 items·m^{-3}) in the mesopelagic zone. These studies point to the water column as a major reservoir of MPs and highlight the importance of understanding the ecological and physical processes responsible for the export of these pollutants from the surface to deep-sea sediments. Specifically, the presence of smaller particles in the mesopelagic layer (Pabortsava and Lampitt, 2020) suggests the progressive degradation of MPs in the seawater environment, highlighting the existing incongruity of the oceanic plastic budget

(Eriksen et al., 2014; Pabortsava and Lampitt, 2020; Thompson et al., 2004; Van Sebille et al., 2015).

4.1.3. Marine sediments

The most common sampling approach was collecting bulk sediment samples, with a significant constraint related to the extraction and purification protocols (See section 4.2). Thus, the use of different sampling devices does not imply such substantial variability as it does for the water compartment. The reviewed studies (n=21) investigated the MPs presence in surface sediments, except for the study from De Ruijter et al. (2019), which investigated the vertical distribution of MPs in intertidal sediments cores. Despite the collection of sediment cores, palaeo-oceanographic approaches investigating historical accumulation patterns in sediment archives (Bancone et al., 2020) are absent from the Mediterranean literature. Future studies would need to address the role of geophysical processes (i.e., sediment and organic accumulation rate) and anthropogenic pressures in the area (i.e., bottom trawling) on the fate of MPs once they reach the seafloor. These aspects need to be taken into account to gain knowledge of MPs' sequestration within marine sediments, their dilution and accumulation patterns, and potential resuspension.

4.1.4. Beaches

The sampling strategy to assess the occurrence of MPs in the Mediterranean beaches encompasses a wide range of methodologies/approaches, depending on the main goal of a given study. For example, reduced volume samples were employed in studies where pellets or specifically L-MPP (>1-5 mm) were targeted (Atwood et al., 2019; Grelaud and Ziveri, 2020; Maršić-Lučić et al., 2018; Merlino et al., 2020; Piehl et al., 2019; Turner and Holmes, 2011a). However, the collection of bulk sediment samples was the most common (83.3%). Like in marine sediments, the whole MP size spectrum can be potentially characterized in this sample type, but the limitations are defined by the laboratory procedures (See section 4.2). The main differences in sampling

protocols' relied on the number of replicates, volume of sand collected, depth of sampling, beach area sampled, and the ratio of the sampled surface area to the beach's surface. Different protocols have been published for providing guidance on the monitoring of microlitter, targeting MPs particularly. The MSFD technical report recommended monitoring the presence of MPs above the strandline, collecting a minimum of five samples targeting the top 5 cm in a stratified random manner to cover the entire beach or a specific area (MSFD Technical Support group on marine litter, 2013). In an attempt to standardize the sampling protocol, Besley et al. (2017) investigated the sample size, sampling depth, and sampling location required to achieve statistical representativeness. Their proposed protocol developed a formula to calculate the number of samples required per 100 m transect and recommended collecting at least 100 g of sand targeting the top 5 cm. In contrast, the findings of Karkanorachaki et al. (2018) highlight the importance of collecting subsurface samples (down to 10 cm) to truly characterize the concentration of MPs in beaches. Their results showed that the concentration of MPs fragments and pellets was up to one order of magnitude higher in subsurface samples (339.8 ± 104.4 items·m^{-2}) than in surface samples (35.3 ± 11.5 items·m^{-2}). Future studies monitoring MP pollution on beaches need to strategically design their sampling to consider the morphological characteristics of the beaches. Generally, beaches are dynamic systems with changing conditions due to environmental and anthropogenic causes. Among these factors, for the Mediterranean Sea, studies should consider the low-tide amplitude, hydrodynamic forces, beach morphology, wind exposure, seasonal visitor pressure on the beach, proximity to MP sources, cleaning events, erosion after storm events, and potential maintenance works (i.e., restocking of sand).

4.2. The effect of extraction protocols

In seawater samples taken from the Mediterranean Sea, the extraction of MPs predominantly relies on physical separation processes – sieving, flotation, and manual selection under the stereomicroscope (Fig. 2.3). Although most of the

samples were collected within the continental margins, which are recognized as high productivity areas, pretreatment for organic matter (i.e., phyto- and zooplankton) oxidation protocols were rarely reported in the reviewed studies. Purification processes (i.e., enzymatic, digestion, oxidation) may reduce the processing time, enhance MPs' recovery, and simplify the subsequent step of identification (Cole et al., 2011; Löder et al., 2017). However, the protocol selection should be cautious, as temperature and different reagents may damage MPs (Enders et al., 2017; Munno et al., 2018). Following the manual MPs selection approach, the extraction efficiency highly depends on the user's experience, the particles' size, microscope's magnification, and the sample's complexity (e.g., rich biota-samples; Löder and Gerdts, 2015). Manual selection of particles is time-consuming and inevitably introduces a bias towards selecting larger and color particles because these are easier to recognize and isolate (Song et al., 2015). The predominance in the use of nets to sample the seawater compartment sets the lower size cut of the collected MPs, generally above 200-300 μm. The manual selection of the particles introduces a wide range of variables that undoubtedly affect the MPs detection limits and compromises the accuracy of the isolation. Such variability should be addressed by systematically implementing quality assurance practices (i.e., spiked samples, interlaboratory comparison exercises between Mediterranean laboratories) to validate the extraction's protocol and define the extraction rate and detection limits.

The MP extraction from marine sediments is challenging. For example, the organic content of the sediments, although less than surface water samples, may retain the MPs, complicating their separation. To address that, samples are subjected to a purification process to facilitate and enhance the recovery of MPs (R. R. Hurley et al., 2018; Löder et al., 2017; Masura et al., 2015). In the Mediterranean Sea, most of the samples were collected in the coastal margins. These areas represent sites of significant importance for many biogeochemical processes, including organic carbon burial and remineralization (Muller-Karger

et al., 2005). Thus, higher organic content is expected in these types of samples. In the reviewed studies, the predominant extraction protocol consisted in sieving and separation by density (Fig. 2.5), with few studies (n=2) performing oxidation treatment of the samples (De Ruijter et al., 2019; Krüger et al., 2019).

The wide variety of existing polymers differ on their specific density, which is generally used for the particles' physical separation. Due to the higher density of sediments (quartz, ρ= 2.65 g cm^{-3}) than the plastic materials, Thompson et al. (2004) proposed a flotation approach to extract the MPs from the sample matrix, using NaCl as a brine solution. Researchers used NaCl predominantly for the density separation in the Mediterranean studies because it is the most common brine solution in MP studies (Hidalgo-Ruz et al., 2012), it is environmentally friendly, and it is cost-effective. The limitation of using this brine solution is that plastics materials with a higher density (i.e., PET, ρ= 1.38 g cm^{-3}; or PVC, ρ= 1.39 g cm^{-3}) might be underestimated. Different brine solutions with a higher density, as NaI ($\rho\approx$1.8 g cm^{-3}), sodium polytungstate (SPT, $3Na_2WO_4 \cdot 9WO_3 \cdot H_2O$, p≈ 1.4 g cm^{-3}), zinc bromide ($ZnBr_2$, $\rho\approx$ 1.71 g cm^{-3}), sodium bromide (NaBr, $\rho\approx$ 1.37-1.40 g cm^{-3}), calcium chloride ($CaCl_2$, $\rho\approx$1.3–1.5) have been proposed as an alternative to enhance the recovery of denser polymers from sediment samples. While there is no standard recommendation, the selection of the appropriate brine solution should consider the polymers' density targeted in the study, hazard, and economic variables (Frias et al., 2018).

Besides the brine solution, other setting parameters may affect the rate of MP extraction from sediment samples. However, little attention was devoted to investigate these parameters' effect on the extraction efficiency (i.e., the mass of the sample processed, the ratio of the sediment/brine solution, mixing and settling times, and the number of consecutive extractions needed). Besley et al. (2017) addressed this question on beach sediment samples, suggesting that for 50 g dry weight of sediment samples, 200 mL of brine solution should be added to the sample, the mix stirred for two minutes and then left to settle for at least

6 hours. This process needs to be repeated three times, reaching a recovery rate of 83.0%. Similarly, Simon-Sánchez et al. (2019) reported a recovery rate of 79.4% after three out of five consecutive extractions from sandy samples using NaCl.

The lack of validation on the extraction protocols prevents a quantitive assessment of the effects of these diverse approaches on the reported MP concentration. In the current status of the research field, it is difficult to recommend the correct extraction protocol, as this should be chosen depending on the study's objective, the environmental matrix investigated, and the economic and time limitations. Under this context, the inter-comparison of results between studies will remain jeopardized by the researchers' numerous research approaches to characterize MP pollution. However, the implementation of good practices and different quality assurance measures, which can be easily fulfilled, can help to provide more accurate comparisons (e.g., providing a clear and concise description of the protocol, acknowledging the size limitation of the methods, analyzing spiked samples to characterize the recovery rate of the protocol, control and procedural blanks while sampling and analyzing the samples, presenting the results separately per type of MP pollutant and size fraction).

4.3. Identification

In the Mediterranean Sea, the identification of the particles relied on visual characterization in 49.3% of seawater samples, 27.3% of marine sediment samples, and 30.0% of beach studies. There are general recommendations to visually identify putative MP particles (Hidalgo-Ruz et al., 2012; MERI, 2012)or fibers (Stanton et al., 2019). This method is time-efficient but highly depends on the user and the microscope's magnification (Löder and Gerdts, 2015). Visual characterization may overlook smaller and transparent particles, significantly impacting the underestimation of the MP concentration (Song et al., 2015). Regarding size, the recommended limit for a correct identification is

1 mm for the naked-eye, 500-100 µm using optical microscopy (highly dependent of the magnification; Primpke et al., 2020 and references therein).

Infrared (IR) and Raman are the most common techniques for the chemical characterization of the particles. These analytical methods are recommended by the MSFD (MSFD Technical Support group on marine litter, 2013) and the Joint Group of Experts on the Scientific Aspects of Marine Environmental Protection (GESAMP, 2019). These spectroscopic techniques are typically non-destructive, provide particle number (units) and morphological characteristics of particles larger than 10 µm and 1 µm in size for FTIR and Raman spectroscopy, respectively (Löder and Gerdts, 2015). The analysis is relatively simple, as once the spectrum is retrieved, the polymer type can be determined by comparing it against a reference library. However, the purification and extraction methods are critical to isolate MPs from the environmental matrix (Löder et al., 2017). In the Mediterranean Sea, the general approach was that after visual sorting of the putative plastic particles, a subsample (1.2 - 100% of the particles) was selected for characterizing its chemical composition. In general, these subsamples were chosen randomly, with scarce reference to the statistical weight of the subsample to validate or correct the reported concentrations. Similarly, the rate of successful identification was often not reported in the studies, and when this was clearly stated, it ranged from 5% to 100%. The most common technique within the Mediterranean studies was the characterization of particles by ATR-FTIR. In this approach, the particle is pressed against a crystal and subjected to the IR laser beam to record its IR spectrum. This approach requires hand-picking the particles and manually analyzing them, which is time-consuming, and limits the minimum particle size to that can be manually isolated (≈300 µm; Primpke et al., 2020). As mentioned above, in the seawater compartment of the Mediterranean Sea, the knowledge is predominantly restricted to plastic particles >200-300 µm due to the sampling mesh size. In a few studies (n=4), the Raman spectroscopy technique was used to characterize the MP particles.

Lots et al. (2017) reported a low success for the identification of particles (4.5%). The authors indicated that spectra had low quality due to fluorescence that may result from biological material on the surface of the particles. In the same study, the authors reported the interference of additives and dyes that masked the spectrum, preventing polymer recognition.

Exclusively, Vianello et al. (2013) applied reflectance µFTIR based on chemical imaging of the filters. Under this approach, as the analysis is automatized, there is no need for a visual pre-sorting of the particles, minimizing the human bias. However, the analysis is time consuming, and requires a large initial investment for the acquisition of the instrument (Primpke et al., 2020a). Additionally, the authors reported difficulties in the analysis caused by the use of Glass Fiber filters to concentrate the sample. To minimize this issue, researchers reported concentrating the samples onto an IR transparent or reflectance surface (i.e., suitable membrane; Löder et al., 2015) or window (Simon et al., 2018) to record the spectrum. In the study of Vianello et al. (2013), the predominant size was in the range of 30-500 µm, and the smallest particle size reported was 15 µm.

Pyrolysis-Gas Chromatography-Mass Spectrometry (Py-GC-MS) was used to characterize MPs (<2 mm) in Marina di Vecchiano (Tuscany, Italy) (Ceccarini et al., 2018). To our knowledge, this is the only study in the Mediterranean Sea that applied Py-GC-MS. This technique is a destructive method, which provides accurate data on mass concentration. However, it does not determine the number of particles (units) or their morphological characteristics by itself, which makes the comparison between studies in the region difficult.

4.4. Microplastic pollution in the Mediterranean Sea

While the hydrodynamic conditions and wind-driven processes influence the redistribution of MP debris (Collignon et al., 2012; de Haan et al., 2019; Fossi et al., 2017; Suaria et al., 2016), even far from their sources (Ruiz-Orejón et al., 2018), higher MP presence is related to areas under high anthropogenic

pressure and proximity to (micro)plastics land-based sources (Pedrotti et al., 2016). Coastline morphology may also affect the dispersion of these pollutants as the rugosity of the coast may facilitate the MP retention in nearshore areas (Compa et al., 2020), while smooth and large bays may induce the exports of MPs to off-shore waters (Brennan et al., 2018). Most of the sea surface samples from the Mediterranean Sea were collected in coastal areas (78.2%). Few studies specifically investigated the relationship between MP occurrence and the proximity to the coast. In the Balearic Islands of Mallorca (Spain, NW Mediterranean), Compa et al. (2020) found high heterogeneity of the MP levels ($8.58 \pm 4.08 \cdot 10^6$ items km^{-2}) within the first km of coast, although MP concentration significantly decreased with increasing distance to the coast (<1km). The compiled dataset in Pedrotti et al. (2016) found higher MP concentration ($1.58 \cdot 10^5 \pm 1.57 \cdot 10^5$ itemss$\cdot km^{-2}$) in the first km adjacent to the coast, with a decrease in waters between 1 to 10 km from the coast ($8.0 \cdot 10^4 \pm 3.80 \cdot 10^4$, items km^{-2}), and again reaching high values in waters further away from the coast (>10 km; $1.76 \cdot 10^5 \pm 2.16 \cdot 10^5$ items km^{-2}). In the coastal waters of Tuscany, Baini et al. (2018) found a correlation between MP abundance, which significantly increased with distance from land. In the dataset compiled for this literature review, we observed that 61.5% of the sampling stations were located within the first 10 km of the coast (Fig. 2.10). No relation was observed between plastic debris concentration (items km^{-2}) and distance to the coast (km), and this observation is drawn using data from 636 Mediterranean locations (Fig. 2. 10). These findings agree with previous modelling studies (Liubartseva et al., 2018; Mansui et al., 2015) on the effective plastic dilution trend along the Mediterranean Sea's surface waters, facilitated by the basin's high surface dynamics, that prevent the formation of permanent accumulation areas (Mansui et al., 2015).

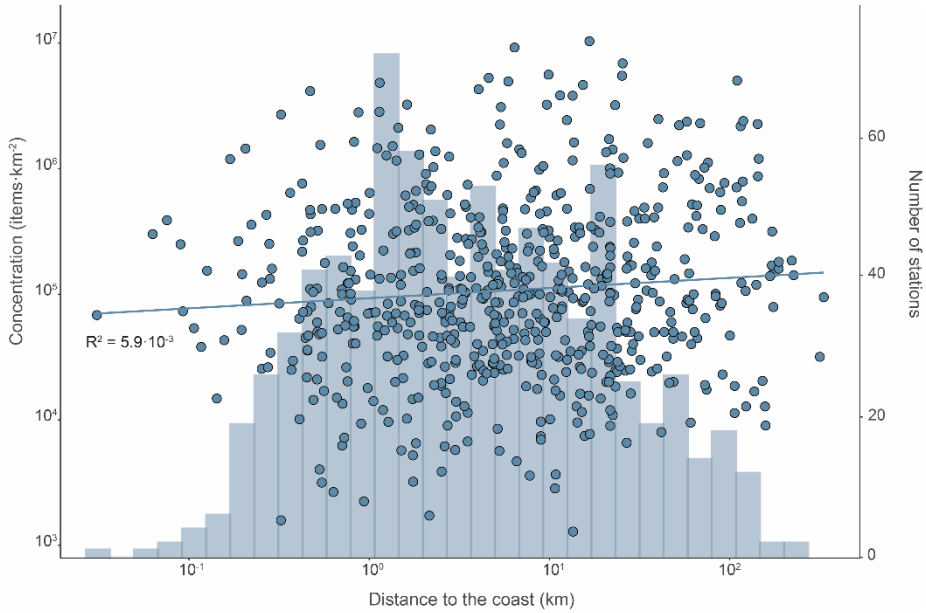

Fig. 2. 10. Plastic particle concentration (items km^{-2}) reported for 636 Mediterranean Sea surface water data points, logarithmically transformed (left Y-axis, blue dots) and plotted in relation to the distance (km) logarithmically transformed. The barplot indicates the number of sampling stations (#unit, right Y-axis) in relation to the distance to the coast.

The geographical distribution of surface water stations (Fig. 2) indicates a gap of knowledge on the MP pollution levels in the southern coast of the basin and in off-shore waters, specifically in the Central Mediterranean and the Levantine basin. Equally important as the spatial distribution of the sampling efforts is the temporal trend. Few studies investigated the seasonal variability of floating plastic debris in the Mediterranean Sea (van der Hal et al., 2017; Baini et al., 2018; Compa et al., 2020). Seasonal patterns, at local and regional scales, on the temporary retention of floating plastic debris were predicted by Mansui et al. (2020), who described three larger retention areas located east of the Balearic Islands, in the central Tyrrhenian Sea, and along the Tunisian and Lebanese coast during summer and autumn. Seasonal and interannual variability is critical to understand the physical transport and accumulation patterns of floating MPs. The detection of main circulation features (i.e., permanent and transient cyclonic and anticyclonic gyres) during sampling efforts may also contribute to our understanding of the accumulation and

dissipative trends of these pollutants (Brach et al., 2018) in the surface waters of the Mediterranean Sea.

We observed a decrease in the sampling efforts in the marine sediment compartment with depth. Few studies investigated MP occurrence in deep-sea sediment, while most of the efforts focused on the continental margins. In the Mediterranean Sea, continental shelf areas play an essential role, representing up to 20% of the basin's surface (Pinardi et al., 2004). These areas are characterized by highly dynamic biogeochemical processes, the presence of critical endemic ecosystems, and tremendous biodiversity (Coll et al., 2010; Muller-Karger et al., 2005). Thus, it can be expected that plastic pollutants in these areas may pose a higher environmental risk. The nearness to populated areas represents a continuous input of plastic debris to the marine environment. From these inputs, high sinking plastic flux (1 kg km^{-2}·day) occurs close to the coast (Kaandorp et al., 2020) in the Mediterranean Sea. Biogenic habitats likely sequester those particles in coastal ecosystems (de Smit et al., 2021). For example, seagrass meadows *(Posidonia oceanica)* trap plastic and MP debris and contribute to their removal by aggregating these pollutants within vegetal fibers that are washed up back to the coast (Sanchez-Vidal et al., 2021).

In the Mediterranean Sea, submarine canyons play an essential role in the exchanges between the continental shelf and the deep sea. These geomorphic features of the continental margins are subjected to ephemeral gravity currents, responsible for transporting terrestrial sediments, organic carbon, and recently also MPs to the deep-sea floor (Fernandez-Arcaya et al., 2017; Pohl et al., 2020). High loads of plastic litter were reported in the submarine canyons of the Mediterranean Sea, which are transported down-slope and expected to accumulate at depth (Ramirez-Llodra et al., 2013; Tubau et al., 2015). However, once on the deep-sea floor, bottom currents lead to the erosion, transport, and deposition of sediment (Stow et al., 2019). In the Tyrrhenian Sea, Kane et al. (2020) showed the strong influence of near-bed thermohaline currents on the fate of MPs. Therefore, focusing research efforts to coastal areas

may bias our understanding of MPs occurrence in the Mediterranean sediments. Future studies need to understand the MP presence within a bathymetric gradient, considering the Mediterranean physiographic settings, vertical settling fluxes, and deep-sea currents that may lead to the final accumulation of MPs in this environment. In the Mediterranean Sea, the deep sea represents the 80% of the basin, MP impacts to this environment may occur imperceptibly, as the remoteness of the deep sea still limits our knowledge on the biodiversity inhabiting it (Danovaro et al., 2010).

5. Perspectives

Detailed assessment of MP pollution is challenging, and while the lack of standardized methods persists in the field, researchers should be responsible for providing standardized results, and to highlight the limitations of their study (Gago et al., 2016; Hartmann et al., 2019; Provencher et al., 2020). Under this context, to advance the knowledge of MPs pollution in the abiotic compartments of the Mediterranean Sea and to understand the environmental risk that these pollutants pose to its biodiversity, major research questions/challenges need to be addressed.

- Combining efforts. Here, we have discussed the limitations of characterizing the whole spectrum of MPs while acknowledging the vast amount of work developed in the past years. Ideally, studies should target the whole MP spectrum, however temporal and economic constraints are evident. Thus, MP pollution research in the Mediterranean region will be highly benefited if the data produced – sampling parameters (i.e., coordinates, sea state, wind, depth, etc.), abundances, particle's size, polymer, shape and color distribution – will be made available in open data repositories.
- Quantifying MP pollution. The numerous research efforts conducted in the Mediterranean Sea provide an excellent opportunity for future monitoring of the temporal trend on the abundance of these pollutants within the basin. Future studies need to consider within their objectives

to apply similar approaches for the comparison of data already produced for the Mediterranean Sea. Notwithstanding, implementing good quality practices and quality control protocols will further validate the accuracy of the future knowledge on MP occurrence in the basin.

- Spatial distribution on marine MP data. Future sampling efforts need to consider sea surface features, and seasonal and interannual variability within the Mediterranean general circulation. There is also an urgent need to define the MP pollution levels in the North African coast and the Tyrrhenian, Aegean, and Levantine basins.
- Assessing MPs sinking/export in the water column. Very little is still known about the dynamics of MPs from the sea surface to the seafloor. The role of the water column is indubitable. Holistic approaches should be considered to gain comprehensive knowledge on the physical and biological processes distributing these pollutants across different environmental compartments. Understanding the MP abundance in the epipelagic and mesopelagic layers provides baseline knowledge to the MP exposure that fish stocks inhabiting these layers of the ocean are facing. This is particularly relevant, as crucial commercial fishing species for human consumption thrive in these layers.
- Assessing the accumulation of MPs. The deep-sea sediments are considered a major reservoir of MP pollution in the marine cycle. In the absence of studies linking geophysical processes with MP occurrence in sediments, the data is restricted to presenting only a snapshot of the potential exposure. However, to further understand the risk of these pollutants buried in the deep ocean, we need to gain knowledge of their patterns (accumulation, dilution, resuspension) and the hazard they might represent to the Mediterranean benthic communities.
- Caring about the macro-sized fraction. The effect of plastic pollutants is detrimental in all its size distribution, from macro to nano. The role

of macroplastics as a source of MPs cannot be overlooked, as well as the socio-economic impacts that this size fraction represents.

Our growing knowledge in the last decade shows the ubiquitous presence and high MP pollution levels in the Mediterranean Sea. The floating MP concentrations point to this basin as one of the most plastic polluted regions (Cózar et al., 2015; Suaria et al., 2016), already surpassing levels that pose an environmental risk (Everaert et al., 2020). As long as business continues as usual, without a significant reduction on single-use plastics and relevant investment in minimizing waste mismanagement, we can expect plastic pollution levels to increase (Lebreton and Andrady, 2019). Even if the methods constrain our understanding of the MP pollution issue in the Mediterranean basin, there is an urgent need to dedicate research efforts to produce quality, open, and comparable data on the occurrence of these pollutants, promoting broad basin-scale international collaboration. Only through quality science, well-informed and engaged politics, stakeholders, and society, effective measures, actions and regulations can be implemented to tackle the challenge of plastic pollution.

CHAPTER 3

Large geographical survey of micro-litter in Mediterranean beaches following a touristic gradient

1. Introduction

The Earth system is subjected to multiple anthropogenic and natural forcings driving rapid global environmental changes. Marine litter represents one of the many challenges that our oceans are facing (Derraik, 2002), and plastic is the most common type of marine anthropogenic debris (Morales-Caselles et al., 2021). Significant inputs of these pollutants reach annually the marine environment through river discharges (Lebreton et al., 2017a; Meijer et al., 2021; Schmidt et al., 2017). Other sources are fishing activities, transportation spillage, and direct inputs from coastal systems as a consequence of waste mismanagement and littering (Derraik, 2002). Still, providing estimates on the fluxes of plastic entering the ocean, where they increasingly accumulate and persist, remains challenging (Van Sebille et al., 2015). Transitional environments such as estuaries and beaches, provide the opportunity to evaluate the marine litter and plastic debris dynamics at the interface between land and marine systems including their potential accumulation (Onink et al., 2021; Simon-Sánchez et al., 2019).

The Mediterranean beaches are one of the world's leading tourist destinations. Tourism is the foundation of many Mediterranean economies (European Environment Agency, 2014). However, its social and environmental consequences are not trivial (Gössling, 2002). These impacts are exacerbated in islands, which must deal with a very large seasonal increase in population with a sea burdened infrastructure during a concentrated time: the high touristic season. As of 2017, the island of Rhodes, Greece, received about 2.6 million visitors multiplying by twenty-two the number of inhabitants on the island (Eurostats, 2022; Grelaud and Ziveri, 2020). Similarly, the island of Mallorca, Spain, received 13.9 million visitors, whereas the local population is nearly one million (Eurostats, 2022; Grelaud and Ziveri, 2020). This tendency is analogous across Mediterranean islands and is reflected in the marine litter generated on their beaches, whose pollution levels mimic the seasonal influx of visitors during the summer season (Grelaud and Ziveri, 2020). Moreover, the exposure

of these sea-locked regions to the open ocean increases the potential of stranding plastics across their coastline (Onink et al., 2021). Indeed, the microplastic (MP) abundance reported in beach islands typically surpasses continental beaches (Rey et al., 2021). Monitoring programs to systematically evaluate the occurrence of micro-litter and its characteristics are essential to 1) identify potential sources, 2) provide a temporal evaluation of the environmental health status of the coastal systems, and 3) to deliver empirical data to improve numerical models assessing the fluxes and dispersion of these pollutants in the marine environment. Ultimately this knowledge pursues the need to elaborate on effective solutions and targeted policies that lead to the development of sustainable tourism and the protection of the marine environment.

The scientific community has extensively reported the abundance of marine micro-litter, specifically MPs, in the Mediterranean Sea (Llorca et al., 2020; Martellini et al., 2018). Yet, few studies provided insights into MP occurrence at basin spatial and temporal scales (Baini et al., 2018; Compa et al., 2020; Cózar et al., 2015; Guerranti et al., 2017; van der Hal et al., 2017). Furthermore, the intercomparison and comprehension of MP pollution in the Mediterranean basin are jeopardized by the lack of standardization of the methods (Simon-Sánchez et al., 2021). In this context, this study aims to provide a systematic identification of the micro-litter, specifically MPs and anthropogenic fibers, occurrence in Mediterranean beaches by investigating the seasonal variation of anthropogenic micro-litter and its characteristics on a large geographical scale with the specific objectives of i) quantifying the abundance of micro-litter along Mediterranean beaches subjected to three different levels of anthropogenic pressure, ii) evaluating how the high and low touristic season influence their occurrence and characteristics, and iii) assessing the local spatial dispersion of the micro-litter in beaches by investigating the occurrence in intertidal and middle beach sediments.

2. Methods

2.1. Sampling

Seven Mediterranean islands were surveyed to assess the seasonal variation in the abundance of micro-litter as an effect of tourism during 2017 (Fig. 3.1). Three beaches with different degrees of anthropogenic pressure were selected and classified on every island. Three beach categories were considered: touristic, local, and remote, depending on the number of visitors, touristic infrastructure, and accessibility to the beach (Grelaud and Ziveri, 2020). Five samples were collected during four survey campaigns covering the low (from October to April) and high touristic seasons (from May to September) at every beach, two at the high-water mark and three in the mid-beach area. A quadrat of 20 x 20 cm was placed at every sampling point, and the upper 2-3 cm were collected in a clean, labeled glass jar (1L). Three hundred ninety samples were collected and sent to the Institute of Environmental Science and Technology – Autonomous University of Barcelona (ICTA-UAB) for sample processing and identification. Details of the sampling locations are provided in the Supplementary material Table S1.

2.2. Sample preparation

In the laboratory, 285 samples were prepared for micro-litter quantification and characterization. The sediments were homogenized. An aliquot of 100-150 g (wet weight) was dried at 50 °C until a constant dry weight (DW) was reached. The mass of sediment analyzed ranged from 71.2 - 131.7 g DW. Dried sediments were placed in a 2 L beaker, suspended in 250 mL of NaCl solution (≈ 1.2 g cm^{-3}), and stirred for 20 mins at 200 rpm. The supernatant was piped-out and placed in a beaker or separatory funnel. The density separation step was repeated, and the supernatant was collected and combined with the one from the first separation. After settling and discarding the non-floating solids, the combined supernatants were filtered through a glass-fiber filter (GF/F, ⌀47 mm, 0.7 μm pore size, Whatman).

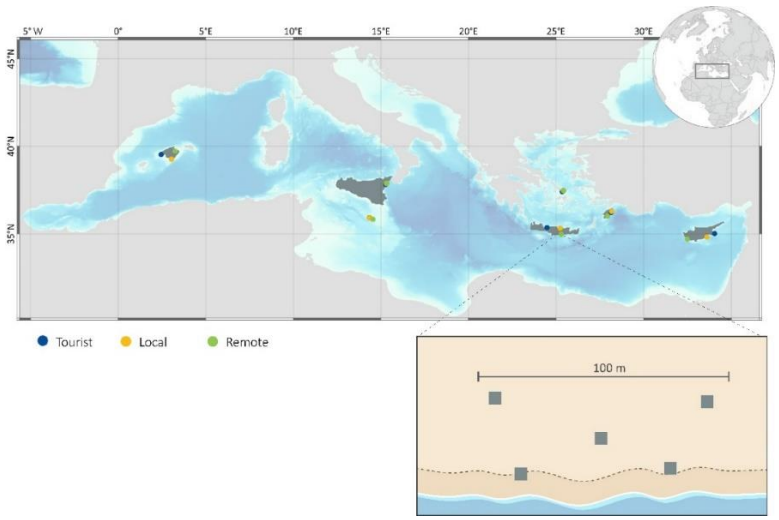

Fig. 3. 1. A. The location of the Mediterranean beaches monitored for micro-litter occurrence classified according to the type of beach: Touristic (blue), Local (yellow), and Remote (green). B. Scheme showing the sampling strategy, two samples were collected at the high-water mark and three in the middle part of the beach.

2.3. Recovery rate

A subset of 42 samples was treated to assess the recovery rate of the extraction protocol. In this subset, the density separation step was repeated five consecutive times. The supernatant of every extraction was filtered independently to assess the relative recovery after five extractions.

2.4. Identification and characterization of micro-litter

The filters were processed under a stereomicroscope (Leica Z16 APO, magnification 7.1×–115×). The entire filter surface was observed from the top left to the bottom right to avoid double-counting. Putative anthropogenic particles were photographed, counted, and classified by shape. The shape categories considered were fiber, fragment, film, foam, pellet, microbead, and aggregation of fibers. Remarkably, under this characterization, the category fiber includes all fibers of anthropogenic origin, synthetic and non-synthetic. A subset of 983 particles collected primarily during the first sampling campaign across all the islands and types of beaches was measured using ImageJ software (Schneider et al., 2012).

Moreover, a limited subsample of 26 particles (9 fragments and 17 fibers) randomly selected was chemically characterized using FPA-μFTIR-Imaging spectroscopy. Measurements were collected using an Agilent 620 FTIR microscope equipped with a 128 × 128 pixel FPA coupled with a Cary 670 FTIR spectrometer (Agilent Technologies, Santa Clara, CA, USA). Particles were hand-picked with high-precision tweezers from the GF/F and placed onto a Zinc Selenide (ZnSe) window or an anodisc. Optical images of the particles were determined with a 15× objective. The IR map of the particles was collected in transmission mode in the range of 3750 to 850 cm^{-1} (ZnSe window) and 3600 to 1200 cm^{-1} (anodisc), using a 15× IR Cassegrain objective-condenser system with a spectral resolution of 8 cm^{-1}, 30 co-added scans for the sample, and 120 for the background.

The resulting hyperspectral images were treated with the software Resolution Pro (Agilent Technologies, Santa Clara, CA, USA). Based on expert criteria, the best quality spectrum of each particle was exported. For chemical identification, the spectra were converted from %Transmittance (%T) to Absorbance (Abs) and compared to several reference libraries in OMNIC 8.3 software (Thermo Fisher Scientific, Madison, WI, USA).

2.5. Contamination

Sample preparation was conducted in a dedicated clean laboratory for MPs analysis at ICTA-UAB to prevent airborne contamination. The laboratory consists of an $8m^2$ hard wall laminar flow chamber ventilated through 4 laminar flow HEPA filters (H14). Besides, orange coveralls were used to identify potential fiber contamination (Fig. S3. 1).

During sample preparation, the reagents, specifically the brine solution NaCl, were filtered through a glass fiber filter (0.7 μm, Whatman) prior to being used. All the materials were rinsed three times with MilliQ® and immediately covered with aluminum foil before being used. Nine procedural blanks were run parallel to the environmental samples to assess airborne contamination.

2.6. Data analysis

The potential airborne contamination recorded in the procedural blanks during sample preparation is reported, and the efficiency of the methodology is presented. However, no correction factors were applied to the concentrations described here.

To calculate the concentrations, the counts of micro-litter yielded after two density separations were divided by the dry weight of sediment processed per sample and standardized to items· kg^{-1} DW. Data distribution followed a no normal distribution (Shapiro-Wilk, $W=0.6240$, $p < 0.01$). To test the differences in Mediterranean beaches' micro-litter abundance, non-parametric tests, Kruskal-Wallis, and a Mann-Whitney U test were considered for the following variables: type of beach (touristic, local and remote), touristic seasons (high and low), and area of the beach sampled (high-water mark and middle beach), respectively. For significant factors, a post hoc analysis Mann-Whitney Wilcoxon test was performed to determine specific differences between the type of beaches. Data analysis and visualization were conducted in R-4.1.1 (RStudio Team, 2020), using ggplot (Wickham, 2016) and cowplot (Wilke, 2020) packages. The level of statistical significance was set at $p < 0.05$.

3. Results

3.1. Particles' recovery and procedural blanks

The assessment of the recovery rate in the subset of 42 samples revealed a significant negative linear correlation between the percentage of particles recovered and the number of consecutive extractions (Pearson's, $r = -0.56$, $p< 0.01$). Fig. 3. 2 shows the relative abundances (%) of particles recovered after five extractions. After the first extraction, the median recovery was 33.3% (Q_1-Q_3: 25.0 – 40.6%). The relative recovery of the following extraction was 22.9% (Q_1-Q_3: 19.6 – 29.4%), 16.4% (Q_1-Q_3: 9.1 – 23.1%), 11.8% (Q_1-Q_3: 8.3 – 18.4%) and 10.0% (Q_1-Q_3: 5.9 – 20.0%).

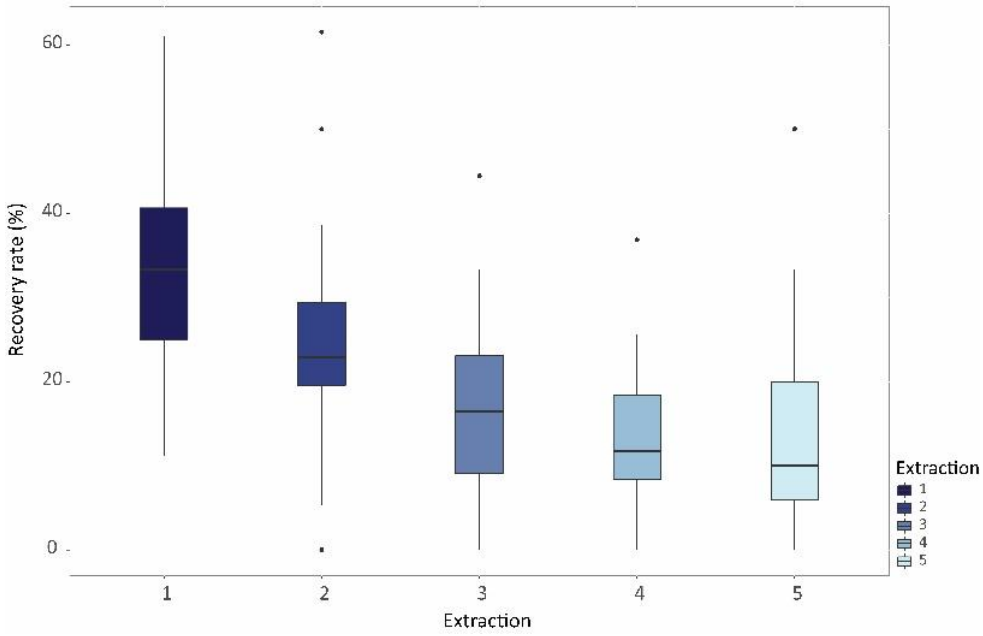

Fig. 3. 2. Boxplot comparison of the recovery rate of micro-litter and the number of consecutive density separations extractions to recover micro-litter. In the graph, the line stands for the median value, the lower and upper lines represent the Q_1-Q_3 percentiles, respectively, whiskers expand to extreme data points and outliers are plotted individually as points.

In the nine procedural blanks, the number of particles detected ranged from three to eight, with an average of four particles per blank. The predominant type of particles found were fibers (80.1%), followed by fragments (15.4%) and films (3.9%).

3.2. Abundance of micro-litter

A total of 5858 particles of micro-litter of suspected anthropogenic origin were retrieved and identified from the 285 samples analyzed. Overall, the median abundance of micro-litter registered in the dataset was 98 particles kg^{-1} DW (Q1-Q3: 47 – 224 particles kg^{-1} DW). The lowest abundance (9 particles kg^{-1} DW) was reported in a middle beach sample from the local beach Es Caragol in Mallorca, Spain, during the low touristic season, and the highest (2027 particles kg^{-1} DW) in a high-water mark sample from the touristic beach of Tsambika in Rhodes, Greece, during the low touristic season.

3.2.1. Micro-litter occurrence and composition per type of beach

To compute the median abundance per beach, the concentration of micro-litter recorded in every sample collected at each beach across all sampling campaigns was considered (Fig. 3.3).

In the touristic beaches, the median concentration was 215 items kg^{-1} (Q_1-Q_3: 94.2 - 529 items kg^{-1}). The minimum concentration of 108 items kg^{-1} (Q_1-Q_3: 94.2 - 529 items kg^{-1}) was registered in Platis Gyalos, Mykonos, Greece, and the maximum 598 items kg^{-1} (Q_1-Q_3: 385 - 1042 items kg^{-1}) in Sun Rise beach, Cyprus. In the local and remote beaches, the median concentration was one order of magnitude lower than on tourist beaches, 80 items kg^{-1} (Q_1-Q_3: 47.8 - 179 items kg^{-1}) and 68 items kg^{-1} (Q_1-Q_3: 30 – 114 items kg^{-1}), respectively. Significant differences were found in the occurrence of micro-litter depending on the type of beach (Kruskal-Wallis, X^2= 55.42, df = 2, $p < 0.01$). The post hoc test revealed that the concentration between the three types of beaches significantly differed, the touristic against the local and remote ($p < 0.01$), and the local from the remote ($p < 0.05$). Among the local beaches, the lower concentration 70 items kg^{-1} (Q_1-Q_3: 36 – 138 items kg^{-1}) was found in Es Caragol, Mallorca, Spain, and the highest in Arina, Crete, Greece, with 68 items kg^{-1} (Q_1-Q_3: 30 – 114 items kg^{-1}). On the remote beaches, the concentration ranged between 38 items kg^{-1} (Q_1-Q_3: 28 – 56 items kg^{-1}) in Tsoutsuras, Crete, Greece, and 114 items kg^{-1} (Q_1-Q_3: 56 – 219 items kg^{-1}) in Marsaxlokk, Malta.

Regarding the particles' composition per type of beach, from the 5858 particles recovered, 67.7% were found in the touristic beach samples, 18.3% in the local beaches, and 14.0% in the remote beaches. The composition of the micro-litter is displayed in Table 1. Overall, fibers were predominant, followed by fragments, films, foams, and others (microbeads, pellets, and aggregation of fibers).

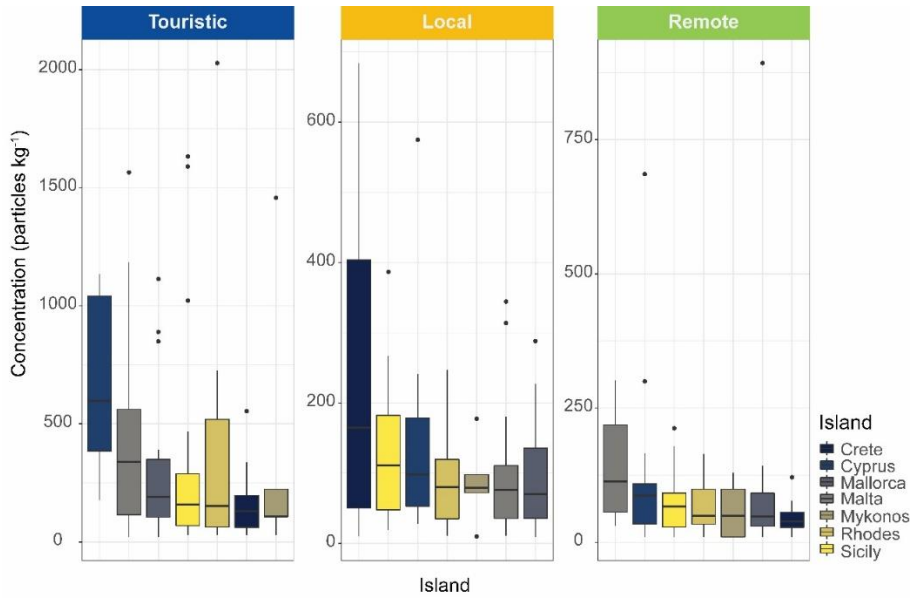

Fig. 3. 3. Boxplot of micro-litter item concentration classified according to the type of beach (touristic, local, and remote) and the Mediterranean island where the samples were collected (Crete, Cyprus, Mallorca, Malta, Mykonos, Rhodes, and Sicily). For each beach, the data are presented in descending order for the median. Indeed, the line stands for the median value, the lower and upper lines represent the Q_1-Q_3 percentiles, respectively, whiskers expand to extreme data points and outliers are plotted individually as points.

3.2.2. Micro-litter occurrence and composition per touristic season

Overall, significant differences were found between the abundance of micro-litter during the low and high touristic seasons (Mann-Whitney U, W = 12899, $p < 0.01$, Fig 3.4.A). A median micro-litter abundance of 74 items kg^{-1} (Q_1-Q_3: 38 – 172 items kg^{-1}) and 127 items kg^{-1} (Q_1-Q_3: 64– 337 items kg^{-1}) was quantified, during the low and high touristic seasons, respectively. There was an increase of 72% (69 - 95 %) of micro-litter occurrence during the high season compared to the median abundance registered during the low season. This increase widely varied depending on the type of beach (Fig. 3.4. B), as the median abundance of micro-litter increased by 95.0% on the touristic beaches, 94.2 % on the local beaches, and 33.3 % on the remote beaches.

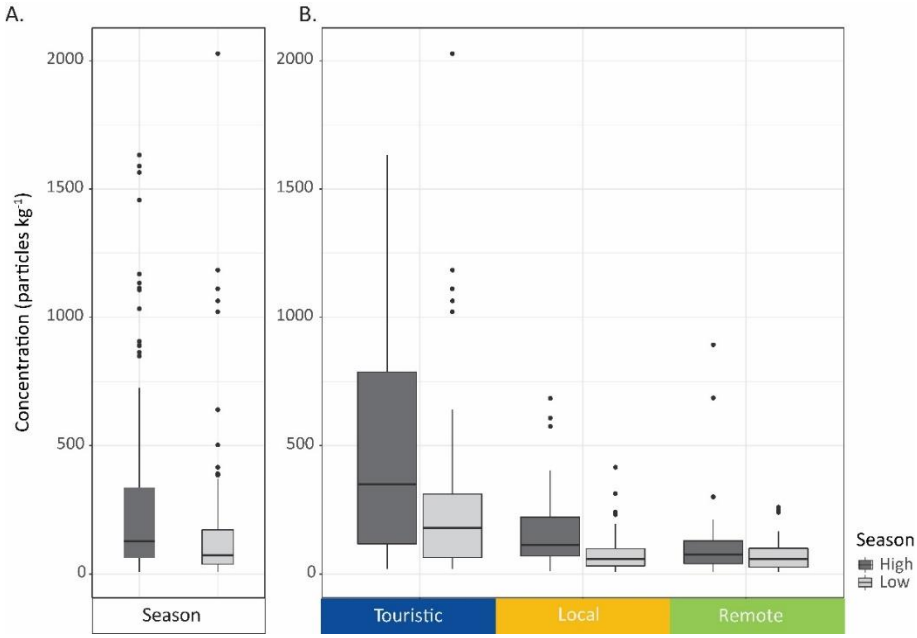

Fig. 3. 4. Boxplot of the overall micro-litter item concentration classified: A. according to the touristic season (high and low) and B. according to the type of beach (touristic, local, and remote) and touristic season (high and low). In the graph, the line stands for the median value, the lower and upper lines represent the Q_1-Q_3 percentiles, respectively, whiskers expand to extreme data points and outliers are plotted individually as points.

Table 3 1. Type of micro-litter items recovered in the Mediterranean beaches classified by the site and time of collection (beach and touristic season).

Beach	Season	Type of micro-litter (%)						
		Aggregation fibers	Fibers	Film	Foam	Fragment	Microbead	Pellet
Local	High	0.2	72.0	2.7	3.2	21.0	0.7	0.0
	Low	0.1	57.2	3.8	4.3	33.4	0.6	0.5
	Total	*0.6*	*60.5*	*3.6*	*0.5*	*34.1*	*0.6*	*0.0*
Remote	High	0.0	70.2	2.8	6.2	19.1	1.5	0.3
	Low	0.3	59.4	2.5	0.8	36.7	0.2	0.0
	Total	*0.2*	*48.8*	*4.4*	*3.0*	*42.7*	*0.0*	*0.6*
Touristic	High	0.1	64.0	2.8	0.7	32.2	0.1	0.0
	Low	0.3	60.1	2.9	1.4	34.8	0.3	0.1
	Total	*0.4*	*56.9*	*2.4*	*0.9*	*39.2*	*0.2*	*0.0*
Total		*0.3*	*60.1*	*2.9*	*1.4*	*34.8*	*0.3*	*0.1*

In general, the composition of the micro-litter is comparable during the different touristic seasons and types of beaches, with fibers as a predominant shape (Table 3.1). Remarkably, there is an increase in fragment occurrence in the remote (+17.6%) and local beaches (+12.4%) during the low season.

3.2.3. Micro-litter occurrence and composition per area of the beach

Concerning the area of the beach sampled, the samples collected in the middle part of the beach had a higher median abundance (110 items kg^{-1}; Q_1-Q_3: 50 – 285 items kg^{-1}) of micro-litter than those collected at the high-water mark (80 items kg^{-1}; Q_1-Q_3: 39 – 156 items kg^{-1}) as significant differences were found (Mann-Whitney U, W= 8182, $p < 0.05$; Fig 3. 5. A).

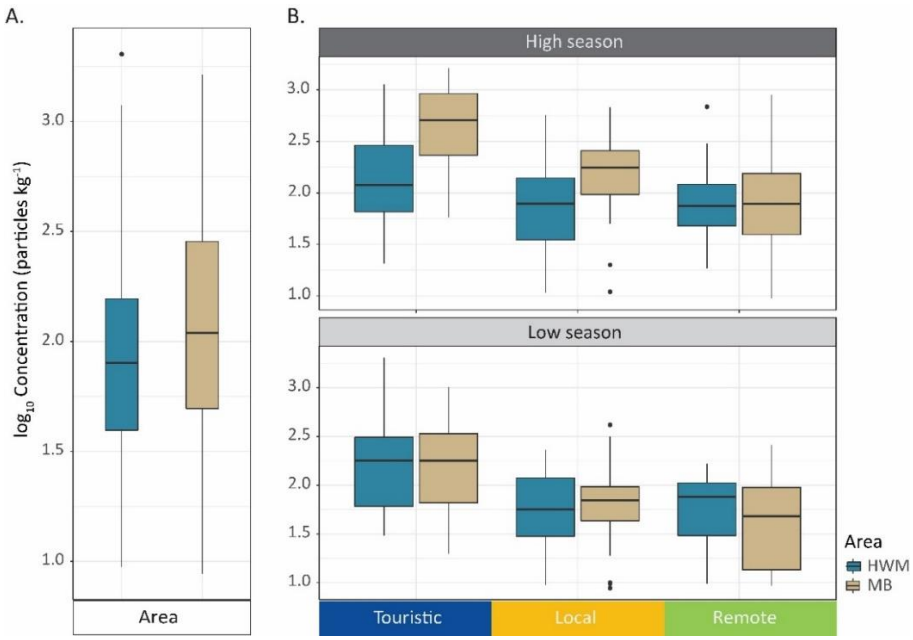

Fig. 3. 5 . Boxplot of micro-litter items classified: A. according to sampling area of the beach (MB: middle beach and HWM: high-water mark) and B. according to the touristic season (high and low), type of beach (touristic, local and remote) and touristic season (high and low). In the graph, the line stands for the median value, the lower and upper lines represent the Q_1-Q_3 percentiles, respectively, whiskers expand to extreme points, outliers are plotted individually as points, and y-axis is in logarithmic scale.

This pattern differs across different touristic seasons (Table 3.2, Fig 3.5. B). During the high-touristic season, the abundance of micro-litter in the beaches'

middle area was higher, representing up to fivefold and twofold, respectively, of the abundances recorded in the high-water mark in the touristic and local beaches. On the remote beaches, the concentration recorded at the high-water mark and the middle part of the beach was similar (Table 3.2.). In contrast, during the low touristic season, similar concentrations were found in the middle beach samples and at the high-water mark, except at remote beaches where the concentration at the high-water mark (76 items kg^{-1}; Q_1-Q_3: 30 – 105 items kg^{-1}) was slightly higher than at the middle area of the beach (48.2 items kg^{-1}; Q_1-Q_3: 14 – 105 items kg^{-1}).

Table 3 2. Summary of the abundance recorded during the different touristic seasons, beach type, and sampling area on the beach. MB stands for middle beach area and HWM for the high-water mark.

Tourism season	Beach type	Sampling area	Median	Q1	Q3	Min.	Max.
High	Touristic	HWM	120	66	291	21	1133
		MB	506	233	925	57	1632
	Local	HWM	79	35	141	11	575
		MB	176	96	257	11	684
	Remote	HWM	75	48	121	19	686
		MB	78	39	154	9	893
Low	Touristic	HWM	179	62	310	31	2027
		MB	178	66	339	20	1022
	Local	HWM	56	30	120	9	231
		MB	70	43	97	9	415
	Remote	HWM	76	30	105	10	166
		MB	48	14	95	9	260

The composition of the micro-litter varied depending on the type of beach, touristic season, and beach sampling area. Overall, the presence of fragments was higher at the high-water mark (38.1%) than at the middle beach (33.3%). In contrast, fibers were more abundant in the middle beach area (62.2%) than at the high-water mark (55.7%). During the high touristic season, the presence of fragments was higher at the high-water mark (touristic:44.6%, local:52.4%, and remote: 54.2%) than in the middle beach area (touristic: 37.9%, local: 29.1% and remote: 34.8%). This tendency was reversed during the low touristic

season in the local and remote beaches, where the presence of fragments was higher in the middle beach area (local:23% and remote 24%).

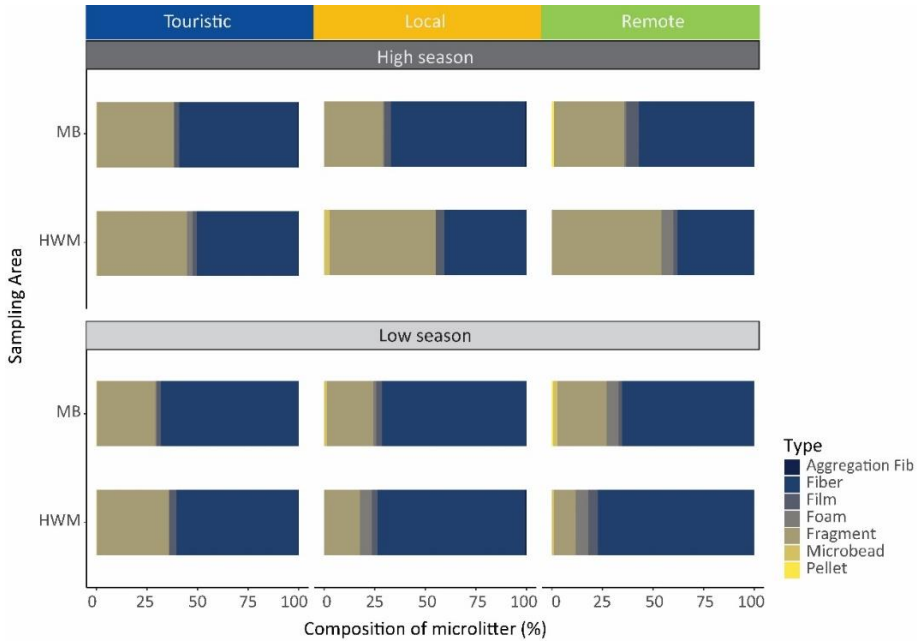

Fig. 3. 6. Comparison of the relative abundances of micro-litter types (aggregation of fibers, fiber, film, foam, fragment, microbead, and pellet) depending on the touristic season (high and low), type of beach (touristic, local and remote), and sampling location on the beach (MB: middle beach and HWM: high-water mark).

3.3. Size distribution and chemical composition

The size distribution of the particles followed a non-normal distribution (Shapiro-Wilk, W=0.7591, $p < 0.01$). The median size was 351 µm (Q_1-Q_3: 150 - 745 µm). 81.7 % of the particles were in the size fraction < 1mm, 13.4 % in the 1-2 mm, 3.6% in the 2-3 mm, and 1.1% were larger than 3 mm.

From the limited subsample analyzed for chemical characterization using Imaging µFT-IR, the results showed a predominance of cellulosic fibers (53.8 %), plastic materials represented 26.8% of the particles, with the presence of polyurethane (PU; 11.5%), polyester (PEsT; 7.7%), polyethylene (PE; 3.8%), polypropylene (PP; 3,8%) and nylon (3.8%). The rest of the spectra showed three visual misidentifications of anthropogenic micro-litter, as two of the fragments were made of calcium carbonate ($CaCO_3$; 7.7%), and another was

identified as an organic compound (a polysaccharide structure; 3.8%). The last spectrum could not be identified (3.8%) because of its low quality due to oversaturation.

3.4. Limitations

The protocol described for extracting and characterizing micro-litter, MPs, and anthropogenic fibers, was adapted from previous publications (Hidalgo-Ruz et al., 2012; Simon-Sánchez et al., 2019; Thompson et al., 2004). Some modifications were applied, and we investigated the recovery efficiency during the density separation by performing five consecutive extractions on a subset of samples. The results showed that after two consecutive extractions, the cumulative recovery rate ranged from 30.0 to 83.3%, with a median of 52.8% (Fig 3. 2).

In previous studies investigating the occurrence of MPs along Mediterranean beaches that applied similar methodologies, the number of consecutive density separation extractions ranged from one to five. However, little explanation of the variables considered to set the number of extractions was provided (Simon-Sánchez et al., 2021). In contrast, the standardized method for processing sand beach samples proposed in Besley et al. (2017) recommended performing the extraction three times to obtain a recovery rate of approximately 80% of the particles yielded after five extractions. Their results correlate well with our findings, which reported a recovery rate ranging between 47.1 and 76.5% after two extractions (Besley et al., 2017). Remarkably, these recovery rates were relative to five consecutive extractions. In Simon-Sánchez et al. (2019), the total recovery was investigated using a spiked sample with virgin fibers of different polymers. The results showed that after five extractions, the total recovery was 77.5 % (Simon-Sánchez et al., 2019). The loss of particles during the recovery was likely influenced by the polymers selected (polyester, acrylic, polyethylene, and nylon) for preparing the spiked sample. Despite the particular aspect ratio of fibers that facilitate their recovery, nylon and polyester still have non-buoyant behavior in NaCl solutions. Several studies have investigated the

efficiency of different protocols for recovering MPs in sand samples (Besley et al., 2017; Quinn et al., 2017). Applying different brine solutions (e.g., ZnCl, NaBr, NaI, $ZnBr_2$) enhances the recovery rate after the first extraction, besides retrieving a broader number of polymers due to their higher density in comparison to NaCl (Quinn et al., 2017). However, these solutions are more expensive and less environmentally friendly than NaCl (Frias et al., 2018).

This study relied on the visual characterization of the particles, with a restricted subsample analyzed for chemical characterization. This approach has been reported to fail on the detection of smaller MPs (Primpke et al., 2020a), particularly fragments and transparent particles (Song et al., 2015). At the same time, and due to false positives, this technique leads to overestimation, particularly with fibers, where synthetic and non-synthetic particles are difficult to distinguish even if visual criteria are considered (Dris et al., 2016). Noteworthy, we concentrated the samples on glass-fiber filters (GF/F). This substrate limited the automatic characterization of the particles using vibrational spectroscopic methods due to its self-absorption in specific spectral ranges and the unevenness of the filter surface. Hence, selected particles needed to be hand-picked and isolated from the filters. The size of the particles (61.5 % measuring <500 µm) and predominance of fibers (60.1%) hampered the applicability of benchtop ATR-FTIR measurements as the limited contact area between the particle and the crystal did not allow to obtain a proper optical contact and, therefore, collect spectra with reasonable quality for chemical identification. Alternatively, considering the properties of the particles, Raman micro-spectroscopy (µRaman) was a potentially powerful method to characterize them. However, samples were neither pre-oxidized nor submitted to any purification process (e.g., enzymatic treatment). Hence, preliminary tests with µRaman showed widespread fluorescence (organic residues on the surface of the particles). Moreover, the laser's choice, and related penetration into the particle's material, led to the identification of the dyes rather than the core chemical composition of the particles in most of the tested particles

(Supplementary material-Fig S3. 2). Similar outputs were reported in Lots et al. (2017), where out of 221 measurements, only 4.5% provided relevant results. Finally, the option was to analyze the particles by isolating and depositing them onto an IR-friendly substrate (Löder et al., 2015) and use an FTIR coupled to a microscope. By the opportunistic access to the instrument, the limited subset of particles characterized with no statistical representativeness showed the predominance of non-synthetic fibers.

Overall, these findings implied the underestimation of micro-litter abundances along the Mediterranean beaches due to method's constraints. Nevertheless, the strength and robustness of the presented data should also be highlighted. The samples covered a large geographical and temporal scale, and the extraction and identification of the particles followed a systematic protocol that allowed to capture the seasonal variability of the occurrence of micro-litter under gradients of beach touristic pressure. Micro-litter, especially MPs, is a global issue; the implementation of monitoring programs to assess the abundance of these pollutants in coastal areas lies on a broad spectrum of socio-economic conditions. Inevitably, there is the need to compromise considering time and economic constraints. Thus, a rigorous interpretation of the results and conclusion should always report the method's limitations to provide a better understanding of the data reported, especially in the MP research field, where policy is rapidly developing based on the real-time data produced by the growing scientific community (Provencher et al., 2020).

4. Discussion

4.1. Micro-litter occurrence in the Mediterranean beaches

High variability in micro-litter occurrence, especially MPs, stranded on Mediterranean beaches has been reported over the last two decades (Alshawafi et al., 2017; Korez et al., 2019; Laglbauer et al., 2014; Turner and Holmes, 2011b). However, the lack of standardization in identifying and characterizing these pollutants has limited the intercomparison of results, preventing an in-depth understanding of distribution patterns at the basin scale (Simon-Sánchez

et al., 2021). This study provides insights on the seasonal variation abundance of micro-litter in the beaches of the Mediterranean Sea subjected to different levels of touristic pressure.

Micro-litter was found at all the Mediterranean beaches investigated, with an overall abundance of (mean ± s.d.): 201 ± 168 items kg^{-1}; min.-max.:48-678 items kg^{-1}. These results are within one order of magnitude difference, below or higher, compared to previous studies reported for Mediterranean beaches (Fig. 3.8). Considering the methods used to determine the abundance of the micro-litter, the study of Lots et al. (2017) provides a relevant comparison. They found an average abundance of MPs (291 ± 62 items kg^{-1}) in Mediterranean beaches with remarkably higher concentrations in the Eastern basin (387 ± 100 items kg^{-1}). In this study, higher and lower abundances were reported in the beach of Sun Rise (677 ± 344 items kg^{-1}; Cyprus) and Tsoutsouras (48.2 ± 35.6 items kg^{-1}; Crete), both located in the Eastern part of the Mediterranean Sea. In our study, the abundance variability is likely influenced by the beach's type, characterized by different concentrations of visitors. This monitoring survey of Mediterranean beaches showed that the presence of micro-litter was significantly higher during the high touristic season, with a decrease in occurrence following the anthropogenic pressure gradient (higher abundances in touristic beaches, followed by local and remote ones). Overall, these observations support the evidence that higher anthropogenic pressure is correlated to a higher abundance of micro-litter on beaches (Ivar do Sul and Costa, 2007; Yu et al., 2016).

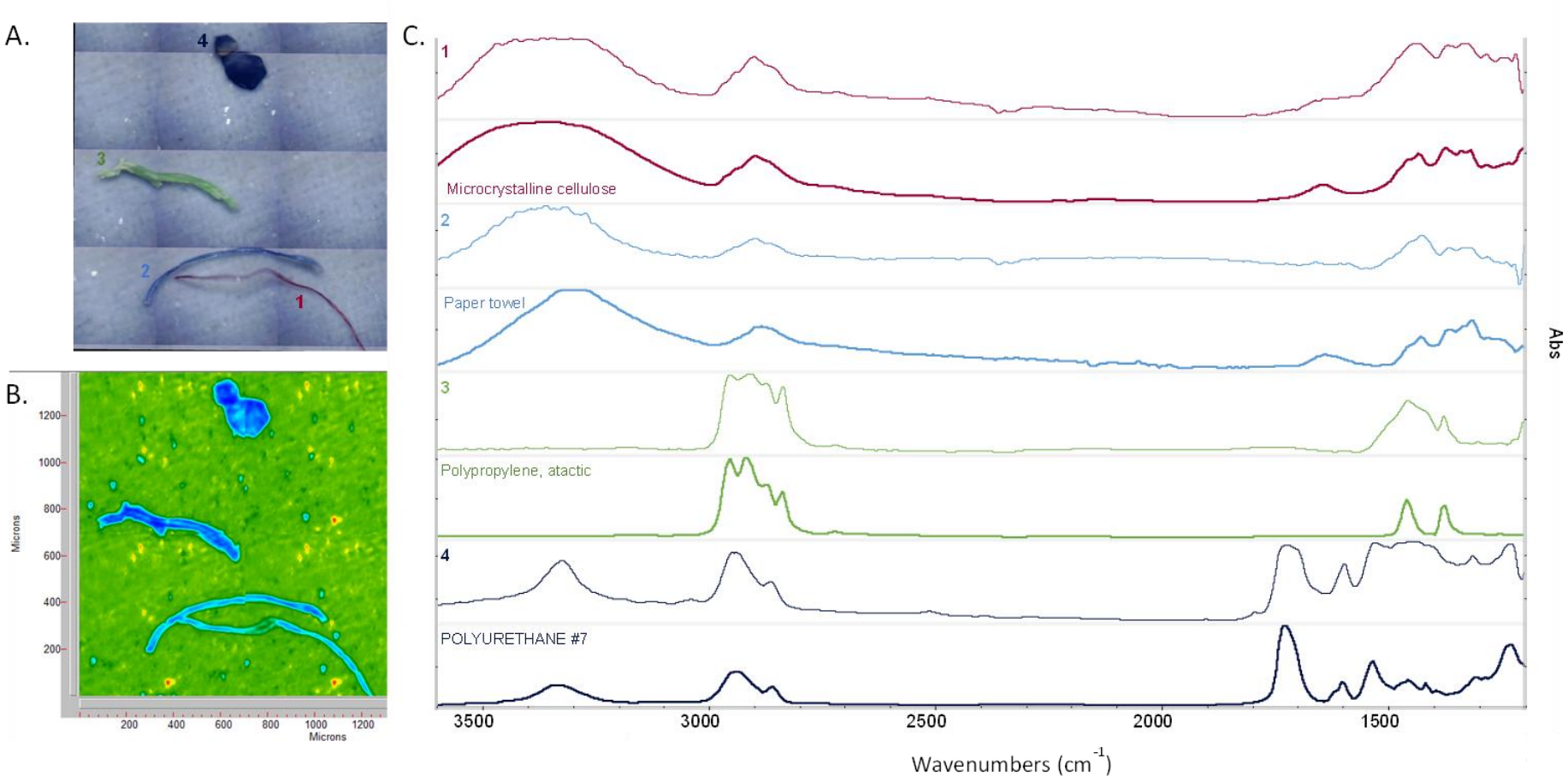

Fig. 3. 7. A. Visual image of four isolated particles for chemical analysis. B. Hyperspectral IR image. C. Absorbance spectra of the analyzed particles, and just below, the corresponding reference spectra to validate the chemical identification. Spectra from top to bottom: 1. Cellulose spectrum corresponding to the red fiber in the visual image, 2 Cellulose spectrum corresponding to the blue fiber, 3. Polypropylene spectrum corresponding to the green fragment and 4. Polyurethane spectrum corresponding to the dark blue fragment.

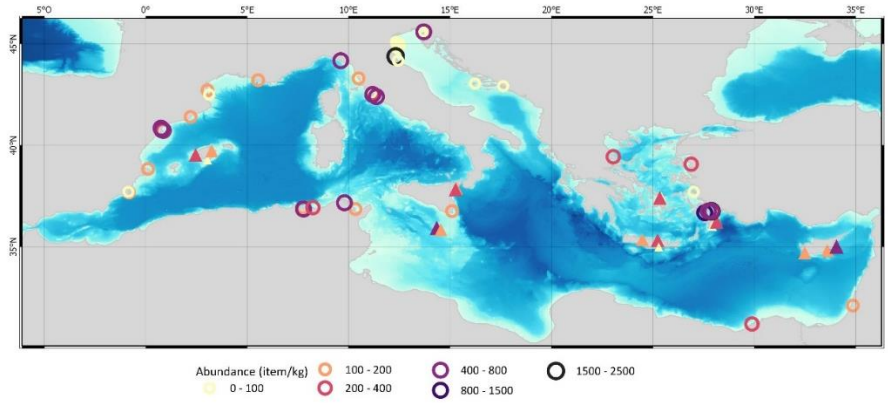

Fig. 3. 8. A map showing the micro-litter abundances across Mediterranean beaches [Δ: data from this study; ○: data retrieved from the literature listed in Supplementary table S3.2]

The sampling design, targeting the intertidal sediments (high-water mark) and shoreline area (middle-beach), allowed us to deduce information about the spatial distribution of micro-litter on the beach, and potential sources and fate, in terms of accumulation or dispersion. In general, in previous studies, most samples to evaluate the presence of micro-litter on beaches were collected at the high water mark (Hidalgo-Ruz et al., 2012; Simon-Sánchez et al., 2021). The sample collection at the intertidal sediments is related to the accumulation of micro-litter due to wave and wind influences. The sampling of these areas is crucial on beaches, particularly located on islands, where the deposition of sea-based marine litter is the dominant, governing the accumulation of these pollutants on their coastline (i.e: Canary Islands, Atlantic Ocean; Baztan et al., 2014; Hernández-Sánchez et al., 2021; and Hawaii, Pacific Ocean: Rey et al., 2021). The Mediterranean Sea is characterized by a micro-tidal regime, where the differences between the high-water mark during the low and high tide is below two meters, on average, about 0.2 - 0.3m. Overall, the abundance of micro-litter was higher in the middle beach (+37%) than at the high-water mark, particularly higher in the touristic (+321%) and local (+176%) beaches during the high touristic season (Fig 3.5). This observation, in combination with the micro-litter abundance per type of beaches, the low precipitation regime of the Mediterranean summer, and the limited efficiency of beach cleaning to collect

micro-litter suggest that small fragments and anthropogenic fibers are likely originated locally following an anthropogenic gradient and accumulate in the beaches during the high touristic season. These findings imply that touristic beaches are primary land-based sources of micro-litter to the Mediterranean Sea. The recorded decrease of micro-litter abundances after the high touristic season suggests that it does not permanently remain on the beach, pointing to the role of environmental processes inducing micro-litter removal (turn-over) in the Mediterranean beaches. Likely the seasonal decrease of micro-litter in the middle beach is a consequence of different weather conditions with precipitation events, where the run-off and wind facilitated the transport of pollutants to the seawater compartment.

Differences were found in the shape of the micro-litter particles across the different seasons and types of beaches. Anthropogenic fibers were predominant across sampling seasons and types of beaches. Their prevalent presence has been reported in different environmental compartments (Carreras-Colom et al., 2020; Dris et al., 2016; Frias et al., 2016; Stanton et al., 2019; Vianello et al., 2019), with an increasing number of publications reporting the dominance of non-synthetic fibers in the marine environment (Sanchez-Vidal et al., 2018a; Suaria et al., 2020). Despite the limited data on the chemical characterization of the particles presented in this study, we also found that most fibers were cellulose-based. One of the main threats of these pollutants is the general belief that their biodegradable composition prevents their persistence in the natural environment. During manufacturing processes, non-synthetic fibers are subjected to similar procedures as synthetic fibers, for example, the addition of artificial dyes and other compounds used to enhance their performance (Remy et al., 2015). Nevertheless, it is unknown at which rate these additives slow down biodegradation in the natural environment. The seasonal increase of fibers during the high touristic season (+9.8%), especially in the middle part of the beach (+14.1%), suggests that the source was likely local atmospheric deposition caused by increased anthropogenic pressure in the area. De Falco et

al. (2020) showed the significant emission of fibers into the air by the daily use of garments. While atmospheric transport might convey the deposition of these fibers far and to remote places (Allen et al., 2019; Suaria et al., 2020), it is also logical to believe in the potential of local deposition. Besides, the increase in visitors to the beaches is linked to bathing practices. The visitors bring along towels; the abrasive action against the sand may shred fibers explaining the predominance of fibers on the Mediterranean beaches. Concerning the increase of fragment presence in local (+12.4%) and remote (+17.6%) beaches during the low season, the local authorities' cleaning efforts to maintain the quality status of the beaches need to be considered. On local and remote beaches, the period between cleaning activities increases after the high touristic season (Grelaud and Ziveri, 2020). Consequently, this variability may lead to the generation and accumulation of a higher number of small plastic fragments during the high touristic season due to the UV exposure of larger plastic items and embrittlement of the particles (Andrady, 2011; Grelaud and Ziveri, 2020), which lately are likely impossible to remove because of their small size.

4.3. Monitoring the presence of micro-litter on beaches

This study provides an extensive geographical and temporal scale monitoring of the occurrence of micro-litter, MPs, and anthropogenic fibers, in Mediterranean beaches following a touristic gradient. The sampling design covering islands from the west to the east of the Mediterranean Sea, and the systematic extraction and identification of particles allowed to capture the seasonal trend on the occurrence of micro-litter at the basin scale. Different efforts to evaluate the presence of micro-litter (MPs in particular) in large geographical areas or temporal scales were reported in the literature. Lots et al. (2017) presented the occurrence of MPs on European beaches from 13 countries while showing citizen science's potential to contribute to the sampling effort. In the United States (US), 37 coastal locations in National Parks were sampled for MP quantification from 2015 to 2016 (Whitmire and Bloem, 2017). These previous studies provided a broad snapshot of MP occurrence on large geographical scales, reporting high variability in MPs accumulated on the

beaches, from remote areas (i.e., Alaska, Iceland) to large urban centers (i.e., Barcelona, Spain). The complexity of factors (i.e., nearness to sources, special geographical features) that potentially determined the presence of these pollutants both at a local and regional level hampered the interpretation of the results (Whitmire and Bloem, 2017). Nevertheless, both studies followed a similar methodology to the one reported here. The abundances of MPs along the National Parks of the US were in the same order of magnitude (min.-max. average; this study: 48-677 items kg^{-1}, US beaches: 21-225 items kg^{-1}; Whitmire and Bloem, 2017), whereas higher values were previously reported in European beaches: (72 – 1512 items kg^{-1}; Lots et al., 2017). Noteworthy, the mean concentrations of this study pooled samples collected across different seasons, while in Lots et al. (2017), the sampling date is unknown, which may explain the difference. Similarly to our findings, in both studies, fibers were the predominant shape of particles (>97%), which supports the evidence of the ubiquity of this anthropogenic particles in the marine environment (Suaria et al., 2020).

At the temporal scale, periodical monitoring efforts have focused on the sea-surface waters rather than beaches (Compa et al., 2020; Li et al., 2021; Thompson et al., 2004; van der Hal et al., 2017). Interestingly, from the studies investigating the variability of the occurrence of MPs at a temporal scale, different environmental processes were identified to affect the presence of these pollutants strongly. For example, MPs on the Mediterranean Turkey coast increased by 14-fold times after flooding events in the coastal surface waters (Gündoğdu et al., 2018). Along the same lines, flooding events were found to significantly decrease the presence of MPs in riverbed sediments by repeating the sampling under changing weather conditions (R. Hurley et al., 2018). Barrows et al. (2018) investigated the seasonal variation of MPs at a watershed scale using citizen science for two years, reporting the potential dilution of MP pollution close to source points during a high flow regime. Paradinas et al. (2021) also emphasized the potential of citizen science to monitor MPs;

notably, they investigated the seasonal variation of MPs across different environmental compartments (intertidal sediments, biota, and coastal water) on the North and West coast of Scotland. In their findings, the challenges of monitoring the presence of MPs at a temporal scale in coastal areas are highlighted, suggesting that year-long monitoring may not be representative of capturing the seasonal trends. In the beach compartment, Misic et al. (2019) carried out a high-resolution study on the Levanto beach (Liguria, Italy) following a seasonal sampling to reveal hydrodynamic events (rip currents, waves) and sand's permeability are governing factors influencing the spatial abundance of fibers. Hence, the relevance of a robust sampling design should be considered when aiming to capture the influence of anthropogenic stressors and environmental processes driving the accumulation, dispersion, and fate of micro-litter in the marine environment. All in all, the present study is the first to provide the abundance of micro-litter covering a large geographical scale and seasonal variation, besides showing the influence of tourism in the generation of micro-litter.

5. Conclusions

This study presents a unique dataset covering a large geographical and temporal scale, the findings indicated micro-litter, microplastics and anthropogenic fibers, are generated by coastal tourism. We found a strong seasonal pattern on the occurrence of micro-litter in the Mediterranean beaches. The presence of these micro-litter increased during the high touristic season and at the touristic beaches. Followed by a decrease in the occurrence during the low-touristic season, suggesting a removal mechanism preventing the permanent accumulation of micro-litter particles at the surface of the Mediterranean beaches. Likely, the land-based microliter originated during the high touristic season is transfer to the coastal waters of the Mediterranean Sea, which influence the turn-over dynamic on the occurrence of micro-litter across different touristic season. Moreover, the predominance of fibers rather than fragments, which may originate from the fragmentation of littering, emphasized

the role of human presence as an unavoidable source of micro-debris. Several economies of the region strongly rely on tourism, attracted annually by the landscapes, history, culture, endemic ecosystems, and biodiversity of the Mediterranean Sea. Hence, there is an urgent need to implement policies and management plans to develop sustainable coastal tourism while prioritizing the protection and conservation of the marine environment.

CHAPTER 4

River Deltas as hotspots of microplastic accumulation:

the case study of the Ebro River (NW Mediterranean)

Abstract

Microplastics (MPs) are considered pollutants that are ubiquitously distributed in aquatic environments. One of the key hotspot areas to understand fluxes of MPs entering into the oceans are transitional systems, between fresh and marine waters, where river estuaries in particular play an important role. In this study we analyzed MPs occurrence in the Ebro River Delta, Northeastern Iberian Peninsula, one of the largest wetland areas in the NW Mediterranean Basin. Microplastic profile, abundance, distribution, and characteristics were screened across different environmental matrices. MPs were collected in sandy beaches on the northern edge of the delta, in estuarine benthic sediments, and in surface waters of the Ebro River, with a mean abundance of 422 ± 119 MPs·kg^{-1} DW, 2052 ± 746 MPs kg^{-1} DW and 3.5 ± 1.4 MPs m^{-3}, respectively. Fibers were found to be the largest class (70 ± 22%) of the three different environmental matrices investigated. We estimated that the Ebro surface water represents an input of 2.14 × 109 MPs yr^{-1} to the Mediterranean Sea. The main contribution of this study is a new insight on the distribution of MPs across different environmental matrices in river estuaries, where estuarine benthic sediments were identified as a potential important sink for MPs.

Keywords: Microplastics, Esturine sediments, surface waters, beach

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1. Introduction

The first evidence of micro-sized plastic particles in the marine environment goes back to the 1970s, when huge amounts of synthetic fibers (Buchanan, 1971) and plastic particles (Carpenter and Smith, 1972) were detected offshore the north-eastern coasts of England and in the North Atlantic Ocean, respectively. The term "microplastic" (MPs) was introduced in 2004 (Thompson et al., 2004), and despite the fact that there is still no standardized definition of the size range of MPs, it is commonly accepted that MPs are those plastic particles smaller than 5 mm (Arthur et al., 2009; Gray et al., 2018; Hidalgo-Ruz et al., 2012; R. Hurley et al., 2018; Peng et al., 2017; Rodrigues et al., 2018; Vermeiren et al., 2016; Yan et al., 2019). Since then, MPs pollution has raised specific concern due to their worldwide widespread. Their presence has been reported in different aquatic environments such as fresh water systems, including lakes (Eriksen et al., 2013; Faure et al., 2013; Free et al., 2014), rivers (Castañeda et al., 2014; Moore et al., 2011; Wagner et al., 2014; Williams and Simmons, 1996) or estuaries (Sadri and Thompson, 2014; Wessel et al., 2016); as well as in marine environment such as shorelines (Browne et al., 2011; Ivar Do Sul and Costa, 2014), surface waters (Cole et al., 2011; Collignon et al., 2012; Cozar et al., 2014; Eriksson et al., 2013; Zhao et al., 2014) and even pristine deep marine sediments (Bergmann et al., 2017; Van Cauwenberghe et al., 2013). However, despite the abundance and occurrence of MPs in the aquatic environment, the fate (Ivar Do Sul and Costa, 2014) and impacts on living organisms (de Sá et al., 2018) remain complex and poorly understood. For example, one of the greatest concerns about MPs is their potential toxicity that can adversely affect freshwater and marine organisms, (e.g.: de Sá et al., 2018; Eerkes-Medrano et al., 2015) the food web and, by extension, human health (GESAMP, 2015). MPs under natural conditions can adsorb persistent organic pollutants (POPs) and act as carrier for these toxic compounds (Tsang et al., 2017; Wang et al., 2016). Besides, the MPs toxicity is related to the presence of inherent toxic monomers and additives used during

the plastic manufacturing process to improve specific properties (Oehlmann et al., 2009.; Teuten et al., 2009; Tsang et al., 2017).

Microplastics are separated into two classes: Primary and Secondary. Primary MPs are those manufactured and used at MP size range and can include, among other, raw material used to create plastic products (such as pellets; EPA, 1992), small particles used for cosmetics purposes (Zitko and Hanlon, 1991) (i.e.: microbeads/spheres), beads used for industrial abrasive blasting media (Sundt et al., 2014), or fibers used for textiles purposes (Browne et al., 2011) (Brown et al., 2011). Secondary MPs originate from the degradation of larger plastic items when exposed to different physical, chemical and biological processes in the natural environment which break down to form smaller fragments (Barnes et al., 2009; Wang et al., 2016). Secondary MPs will probably play an important role in the near future as plastic items already present in the natural environment may remain there for decades and their fragmentation will produce huge amounts of MPs, even if plastic pollution is stopped right away (Barnes et al., 2009; Eerkes-Medrano et al., 2015).

The sources of MPs into aquatic environments are numerous (e.g.: Duis and Coors., 2016; Windsor et al., 2019). On land, municipal run-off and wastewater treatment plants are considered as a critical entrance, mainly for microbeads and synthetic fibers to the aquatic environment (Horton et al., 2017a; Murphy et al., 2016). Atmospheric inputs could also represent a non-negligible source of MPs (mainly synthetic fibers, Dris et al., 2016, 2015) along with agriculture activity (Nizzetto et al., 2016) through the spreading of sewage sludge as fertilizer or plastic mulching (Steinmetz et al., 2016). The main inputs of plastic to the marine environment come from industrialized and densely populated coastal areas (Andrady, 2011; Derraik, 2002; Jambeck et al., 2015). In addition, plastic can enter into the marine environment through insufficient plastic waste management, illegal dumping, unintentional or accidental releases from vessels, and litter left on beaches (Crawford and Quinn, 2017b; Duis and Coors, 2016). The studies of MP pollution are mainly focused on marine systems (Avio

et al., 2017; Cole et al., 2011; Ivar do Sul and Costa, 2007; UNEP, 2011). Until now, rivers have received less attention despite the fact that they contribute 1.1-2.4 million tons of plastic to the marine environment annually (Lebreton et al., 2017b) and are considered the main pathway for plastic litter to reach oceans. Few ongoing research activities have recently focused their attention on plastic pollution in freshwater ecosystems, seeking to close the knowledge gap about the occurrence, sources and fate of MPs in rivers (Besseling et al., 2017; Lagarde et al., 2016; Leslie et al., 2017; Wagner et al., 2014; Wang et al., 2017; Zhang et al., 2016). Further to this, emerging studies highlight the need to understand the distribution of MP fluxes between the MPs retained within river catchments and MPs exported to the open oceans (Windsor et al., 2019; Xiong et al., 2019). In particular, estuaries, transitional areas between freshwater and marine systems, are recognized as important accumulation areas for several anthropogenic pollutants (Foufoula-Georgiou et al., 2011; Vörösmarty et al., 2009), making them critical zones to understand the accumulation and export fluxes of MPs to the oceans.

This study represents the first evaluation of MPs pollution in the Ebro River Delta, one of the largest wetland areas in the western Mediterranean region, a sea characterized by high accumulation of plastic debris (Cózar et al., 2015; de Haan et al., 2019). Thus, understanding the role of estuarine systems, such as Ebro River, as source and sink for MP pollutants will aid in the struggle of estimating the number of MPs reaching the world's oceans using rivers as corridors. The aims of this study were to investigate the occurrence of MPs pollutants in the Ebro Delta in NE Iberian Peninsula in order: 1) to determine the fluxes of MPs across different deltaic environmental matrices, 2) to determine the characteristics of the particles (type, size and colour) and 3) to explore the potential sources of MPs in the area. We assess MP abundance in three different environmental matrices: river surface water, sandy beaches, and estuarine benthic sediments ("benthic sediments" from here onwards). We hypothesized that estuarine systems would act as accumulation areas for MPs

pollutants, and MPs abundances were expected to be higher in the riverine benthic sediments.

2. Material and methods

2.1. Study site

The Ebro Delta is located in Northwestern Mediterranean, in Catalonia, Spain (Fig. 4. 1). It drains one of the main contributors of the Mediterranean Sea, the Ebro River, with a catchment area of 85,569 km^2 and annual mean discharge of 464 $m^3 \cdot s^{-1}$ (Gobierno de España, 2015). The estuarine section is one of the main major delta systems present in the Mediterranean basin. The total subaerial area is of about 320 km^2 and its coastlines length of approximately 50 km. The coastal zone is microtidal (<0.3 m) and characterized by sandy beaches and dunes accumulations (Palanques and Guillén, 1998). According to the classification of Hansen and Rattray (1966), the Ebro River shape is a Type 4-salt wedge estuary (Ibañez et al., 1997), which extends along the last 32 km of the river, with an average width of 250 m and an average depth of 7 m (Nebra et al., 2016). The climatology of the Ebro Delta is characterized by low thermal oscillation, high humidity and prevailing NW winds. The annual mean temperature is 17˚C and the annual mean precipitation of 548 mm, even though there is a high interannual variability (Gobierno de España, 2015).

The population density in the Ebro Delta is 78.1 inhab·km^2 (IDESCAT, 2018), which is relatively low considering that combined, the world's deltas host an average population density about ten time higher than the world average (Ericson et al., 2006; Foufoula-Georgiou et al., 2011). Nevertheless, the Ebro Delta is facing the challenge of finding balance between the anthropogenic activities and the conservation of the ecosystem. The ecological richness and vulnerability of the area lead to the declaration of specific zones of the Ebro Delta (77 km^2) as Natural Parks by the Catalonia Regional government in 1983. It is a Special Birds Protection Area (SPA) and is considered one of the Wetlands of International Importance under the Ramsar Convention. The main

human transformations in the area are related to the economic activities: the agricultural use covers more than half of the surface of the delta and is mainly devoted to rice crops followed by vegetables and fruit trees production (Fatorić and Chelleri, 2012). Fishing is another notable activity in the area: the harbour of Sant Carles de la Ràpita, one of the largest fishing ports in Catalonia, concentrates the commercial fishing, while fish farming production is present through 13 facilities in the area (Agència Catalana de l'Aigua). The main infrastructures in the Ebro Delta are the extensive irrigation and drainage channels and 6 wastewater treatment plants (WWTP) discharging directly or indirectly into the Ebro river (Fig. 4. 1, Agència Catalana de l'Aigua).

Finally, an important feature of the lower part of the Ebro River is the reduction in the flow of sediment and water as a result of river management during the 20th century, based on the construction of dams. Specifically, the dams of the Mequinenza and Ribaroja are considered responsible for strongly regulating the river flow dynamics in the lower Ebro River (Nebra et al., 2016).

2.2. Sampling

The microplastic contamination of the Ebro River Delta system was assessed in three different environmental matrices: in sandy beaches on the northern edge of the delta, in benthic sediments and in surface waters of the river. The sampling surveys were conducted in winter 2017 (see Table S4.1 for more details). Ten stations were selected to investigate the concentration and characteristic of MPs (Fig. 4.1). In order to evaluate the riverine influence of the presence of MPs on the seashore, four sites were selected along the northern beaches of the delta (S1-S4- Fig. 4.1). Sampling stations S1, S2, S3 and S4 were located at 0.6, 2.3, 3.4 and 6.9 km from the mouth of the river. A fifth station (S5) was selected out of the geographical boundaries of the delta system, 23 km to the north of Ebro River's mouth. Three riverbed sampling stations (GS1-GS3) were selected along the last 17 km of the Ebro River, from the river mouth (GS1) to the first towns: Sant Jaume d'Enveja and Deltebre (GS3). The surface waters were sampled at two sites: WS1 at Deltebre, the most urbanized area in

the delta, and WS2 at the nautical club of Riumar, about 3 km upstream of the river mouth.

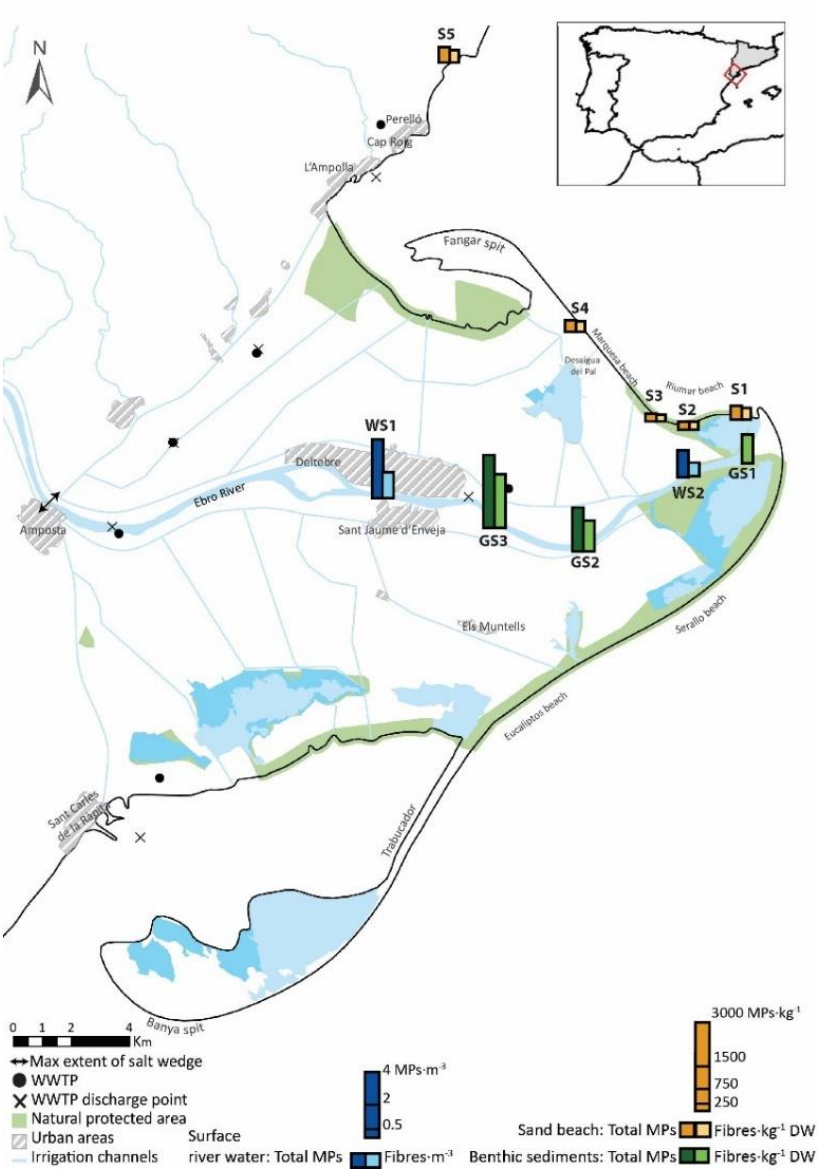


Fig. 4. 1. Geographical location of the Ebro Delta indicating the sampling stations and microplastics concentration (sand and sediments: MPs·kg^{-1} DW; surface water: MPs m−3) in the different environmental matrices. From the 13 sampling location: five beach sand station (S1-S5; ochre), three river bed sediments stations (GS1-GS3, green) and two river surface water stations (WS1-WS2; blue).

For the beach samples about 1 L of sand was collected at the upper limit of the last high watermark, using a 20x20 cm quadrant and scraping the first 2.5 cm of sand with a stainless-steel spoon. Riverbed samples were collected close to the sediment deposition margin of the river with all stations depth ranging from 4 - 7 m (Nebra et al., 2016), see Table 4. 1. Samples were collected using a van Veen grab sampler (0.046 m^2) deployed from a small boat. Once the grab sampler was loaded on board, from the top window of the sampler, the first 10 cm of surface benthic sediments were collected. Surface water sampling was performed with a 15 cm diameter round neuston net of 5 µm mesh. The net was deployed at the river surface against the water current flow during thirty minutes at the first station (WS1), where two replicates were collected, and during one hour at the second station (WS2) (Fig. 4. 1). To transfer the sample and to collect those particles that might remain on the mesh of the net, after each sampling, the net was rinsed, and the material collected was concentrated with water from Elix purification system. All samples were stored in unused, clean labelled glass containers and transferred to the laboratory where they were further processed. River flow data were retrieved from Confederación Hidrográfica del Ebro (CHE) (station A027- Tortosa).

Table 4.1. Benthic sediments site descriptions: distance to themouth of the Ebro, sediment fractions, total organic matter (TOM), depth, salinity and velocity. The parameters (annual mean±s.d; n= 4) with an asterisk (*) were extracted from Nebra et al. (2016).

ID	Lat N	Long E	Gravel (%) >2mm	Sand (%) 2 mm>x>63µm	Mud (%) <63µm	TOC* (%)	Depth* (m)	Mouth distance (km)	Salinity* (g l^{-1})	Velocity* (m s^{-1})
GS1	40.71	0.86	2.4	28.43	71.57	4.8 ± 1.5	6.00 ± 0.5	11	32.0 ± 11.7	0.16 ± 0.14
GS2	40.69	0.79	0.44	64.92	35.08	3.6 ± 1.3	7.25 ± 0.5	7.7	35.3 ± 2.0	0.06 ± 0.01
GS3	40.70	0.76	0.1	39.75	60.25	4.9 ± 0.5	6.25 ± 0.5	1.5	34.8 ± 2.7	0.07 ± 0.11

2.3. Microplastic extraction

The most common method to extract MPs from sediment samples is based on density separation using the saturated NaCl solution (Quinn et al., 2017). This solution, which has a density of 1.2 g cm^{-3}, might fail to separate polymers with

higher densities such as polyvinyl chloride (PVC, ρ=1.38 g cm^{-3}) or polyethylene terephthalate (PET, ρ=1.39 g cm^{-3}). However, it has been shown from previous studies that denser polymers can be separated with this brine solution, what is particularly true for fibers (Quinn et al., 2017; Sanchez-Vidal et al., 2018b). In the present study, the extraction of MPs from the sand beach samples followed a slightly modified methodology developed by Thompson et al. (2004). For a given sample, about 150 g (wet weight) of sediments and 250 mL of concentrated NaCl solution were added into a pre-rinsed 2 L glass beaker. The mixture was stirred for 20 min at 200 rpm. The solution was left to settle for two hours. The supernatant solution was piped out into a clean glass beaker and vacuum filtered using a glass-fiber filter (GF/F; 47 mm ø, 0.7 µm pore size). The walls of the funnel attached to the vacuum system were rinsed twice with ultrapure water (MilliQ®) in order to remove those particles that might remain attached. The glass-fiber filters were stored in Petri dishes and dried at 40 ^{o}C overnight. The extraction was performed during five consecutive times to test the efficiency of the methodology used. After every extraction, the beaker containing the sample was rinsed with concentrated NaCl solution, again to avoid the lost and underestimation of some particles that might remain attached to the beaker walls. Finally, the residual solids were washed with MilliQ®, drained and dried at 40 ^{o}C to record the dry weight.

For benthic sediments, grain size distribution was determined by wet sieving for gravel (ø>2 mm), sand (ø>63µm) and mud (ø<63µm). MP extraction analysis consisted in repeated density separations using Wet Peroxide Oxidation (WPO) to remove the presence of organic matter. For each sample, three different replicates of approximately 10-20 g of sediments were added to three separated clean glass beakers with 200 mL of concentrated NaCl solution and 20 mL of 30% hydrogen peroxide (H_2O_2). The mix was heated at 50 ^{o}C, stirred for 20 min at 200 rpm and left to settle first for an hour at 50 ^{o}C and then one additional hour at laboratory temperature. The foam from the reaction was transferred to a clean glass beaker, mixed with 50 mL of concentrated NaCl

solution and 20 mL of H_2O_2 (30%). After the heating, mixing and settling, the supernatant was collected and vacuum filtered (GF/F; 47 mm ø, 0.7 µm). In the meantime, 20 mL of H_2O_2 (30%) was added to the rest of the WPO solution and sediments. The mixture was again processed using the same aforementioned steps of heating, mixing and settling. The supernatant was then collected and transferred to a clean glass beaker. This mixture was again oxidized, heated, mixed and left to settle. Finally, the supernatant was collected and vacuum filtered (GF/F; 47 mm ø, 0.7 µm). For each replicate, the two filters were stored in Petri dishes and dried at 40 °C overnight. All the settled solids were collected, washed with MilliQ®, dried and weighed to record the dry weight.

To validate our extraction methodology, microplastics (n=40-80) were added to a benthic sediment sample (12.3 g DW) and to a sand sample (99.2 g DW). Acrylic, nylon, polyester and polyethylene fibers were added into the samples. Respectively, the MPs were gathered from cutting a line of acrylic pink wool, a transparent fishing line, a 100% polyester orange garment and a black backpack's strip. The spiked samples followed the same extraction treatment as the rest of the samples.

Surface water river samples were directly vacuum filtered (GF/F; 47 mm ø, 0.7 µm) in the laboratory. In order to remove particles that might remain attached to the walls of the glass container, they were rinsed twice with a squirt bottle containing MilliQ®, and also filtered as part of the sample. The glass-fiber filters were dried at 40°C overnight and stored in Petri dishes.

2.4. Microplastics identification and characterization

The filters were processed under a stereomicroscope (Leica Z16 APO, magnification 7.1x – 115x). The entire filter surface was examined from the top left to the bottom right to avoid double-counting. For accurate MPs identification, the criteria indicated by Hidalgo-Ruz et al. (2012) were followed, such as ensuring that there was no presence of organic or cellular structures, and, that there was a consistent thickness of the fiber across its length with the consideration that some colored fibers presented split ends (Marine

and Environmental Research Institute, n.d.). Similarly, for the criteria of homogeneous color throughout the entire particle, some exceptions were considered such as bleaching, embrittlement, biofouling and plastic design which can affect this characteristic of the particles (Lusher et al., 2017). Thus, those particles identified as MPs were quantified, photographed and classified under type, size and color categories. The categories of MPs types considered were: microbead, fragment, foam, fiber, film/foil and fiber aggregations. For size, the maximum particle length was measured using the software ImageJ (Schneider et al., 2012) and grouped under eight size classes (<50; 50-100; 100-200; 200-500; 500-1000; 1000-2000; 2000-3000; >3000 µm) (adapted from Song et al., 2015). Finally, particles were sorted under color properties considering four categories: transparency (transparent and translucent), black, white and color (light blue, dark blue, red, yellow, purple, grey, etc.).

In this study, a subsample of 25 particles were randomly selected for polymer identification by micro Fourier Transform Infrared Spectroscopy (µFT-IR; Micro FTIR Agilent Cary 610, optical bench Agilent Cary 680) operating in Attenuated Total Reflectance (ATR) mode. MP particles were directly measured on the glass-fiber filters. Analysis were performed with a variable size aperture, depending on the type and size of the particle, in the range of 600-4000 cm^{-1}, with 128 scans at a resolution of 2 cm^{-1}. Identification was based on the comparison of each FT-IR absorbance spectrum against the presence of specific absorption frequencies in accordance with the literature (Cincinelli et al., 2017; Jung et al., 2018).

2.5. Quality assurance

All analyses were performed inside a laminar flow cabinet in order to reduce contamination from airborne particles. Researchers wore cotton lab coats. Glass materials or stainless-steel materials were used when possible and all materials were rinsed twice with MilliQ® before being covered or used or stored. Finally, concerning the possible contamination, blanks (GF/F; 47 mm ø, 0.7 µm) were placed inside the laminar flow and then analyzed on the microscope to identify

and quantify potential background contamination. Airborne MPs such as fibers and fragments were observed in the procedural blanks (n=15) with an average of 0.5 ± 0.8 MPs·filter (ranging from 0-3 MPs·filter). Due to the relative low concentration of particles detected in the procedural blanks, no correction factor was used for the final concentrations of MPs reported in this study. Similarly, for the results reported in this study, no correction was applied following the recovery rate test nor was applied after the FT-IR analysis.

2.6. Data analysis

Microplastic concentrations were standardized and reported as particles·kg^{-1} D.W. for sandy and benthic sediments samples and as particles·m^{-3} for the river surface water samples. Overall, MPs abundances were reported as mean ± standard deviation. Statistical analyses were conducted using the statistical software RStudio (Version 1.1.453) with a significance level set at $\alpha \leq 0.05$. Non-parametric tests were chosen due to the small sample size per environmental matrix and the restrictive replication between samples. Differences in MPs abundance among stations within each environmental matrix were investigated using Kruskall-Wallis. Similarly, the presence of distributional differences between MPs size and environmental matrix were tested using Kruskall-Wallis test. Where significant differences were found, a post-hoc Dunn's test was used to reveal significant differences between groups.

3. Results

3.1. Microplastic extraction efficiency from sand samples

The spiked samples tested to assess the recovery rate of the extraction methods showed a recovery rate of 77.5% after five density separations in the sand samples, and a recovery rate of 70% in the benthic sediments. The lowest recovery rate was obtained for the transparent nylon fibers (13 out of 20 fibers) in sand beach samples and orange polyester fibers (5 out of 10 fibers) in the benthic sediments.

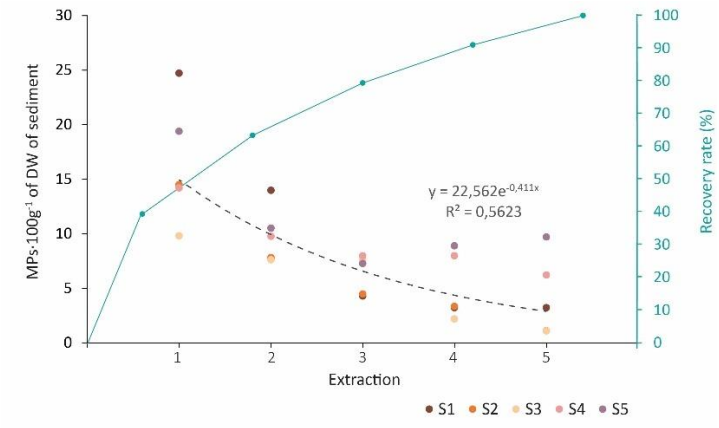


Fig. 4. 2. Number of microplastics recovered from sand beach samples, from a total of five NaCl consecutive extraction.

As mentioned, five sequential density separations were performed to increase the recovery rate of MPs from each of the sand beach samples. Results showed a negative exponential trend on the recovery of particles (Fig. 4. 2). The validation of the method proved that not all the particles were recovered after the fifth density separation. The recovery rate in the first extraction represented only an average of 39 ± 8% compared to the total number obtained after five separations. From the consecutives extractions, the recovery rate increased to 63.4% in the second extraction, to 79.4% in the third and 91.0% in the fourth extraction. These results correlate well with previous studies showing that 93.3% were recovered after fourth density separations (Besley et al., 2017).

3.2. Microplastics identification using µFT-IR

From the 25 particles investigated for chemical composition using µFT-IR, a total of 76% were identified as MPs. From the interpretation of the spectra, three particles could not be identified because the IR signal of the particle was not distinguishable from the background; three particles were identified as non-synthetic particles (cotton, vegetal fiber and coal) and from 19 particles the polymer type was identified. The polymer distribution was dominant by polyamide (24%), followed by polyethylene (16%), poly(methyl methacrylate) (acrylic, 12%), polyester (12%), polypropylene (8%) and polyacrylate (4%).

3.3. Microplastic concentrations in the Ebro River Delta

MPs were found in all analyzed samples. In the three environments studied, MPs were dominated by fibers, followed by plastic fragments and films. These three types of MPs categories were found at almost all sampling locations, except for the S2 sand beach sample where only fibers were detected (Fig. 4. 3).

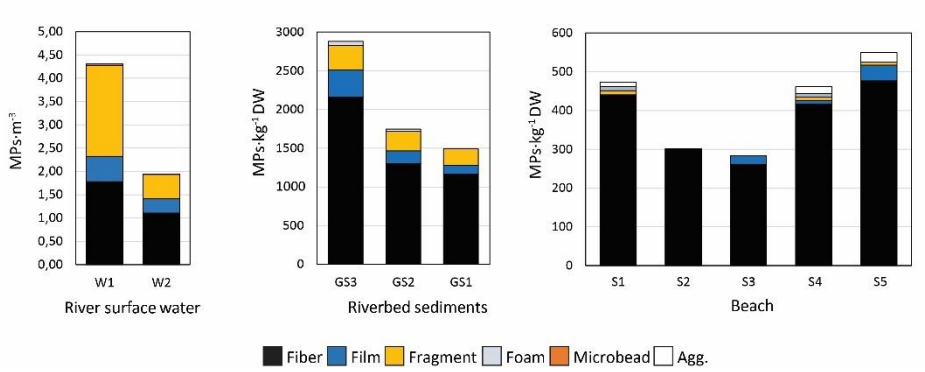


Fig. 4. 3. Concentration of particles at each sampling station: a) River surface water (MPs m^{-3}), b) River bed sediments (MPs·kg^{-1} DW) and c) sand beach (MPs·kg^{-1} DW)

The mean MPs concentration in sandy beaches samples were 422 ± 119 MPs·kg^{-1} of dry weight (DW) sediment (n=5) and 2052 ± 746 MPs·kg^{-1} of DW sediment in the riverbed (n=3). It is interesting to note that MPs concentrations were 4-fold higher in benthic sediments than in sandy beaches. Finally, the average concentration of MPs in surface waters of the Ebro River was 3.5 ± 1.4 MPs m^{-3} (n=3).

Of the 5 sand samples collected from the beach, a total of 220 particles were extracted and analyzed. Fibers (89.5%) were the most common type of particles observed. Concentrations ranged from 283 MPs·kg^{-1} DW sediment in S3 to 557 MPs·kg^{-1} DW sediment in S5, there was not a significant difference in the MPs abundance between the stations (X_2 = 4, df = 4, p = 0.406). The highest concentration was found at the station located out of the delta system (S5), while within the delta there is a spatial variability with higher concentration of MPs in S1 (494 MPs·kg^{-1} DW sediment), closer to the mouth of the river and

in S4 (461 MPs·kg^{-1} DW sediment), closer to the Desaigua del Pal (Fig. 4. 1). Reported concentrations are in the same order of magnitude to the concentration reported in other Mediterranean areas such as the littoral area of the Ombrone River, located in the Maremma Regional Park, Italy (166-318 MPs·kg^{-1}; winter; Guerranti et al., 2017), but lower in comparison to those concentrations reported close to the mouth of Albegna River, Italy (882-1069 MPs·kg^{-1} sediment; winter; Guerranti et al., 2017).

In benthic sediments samples, the highest concentration was found at the most distant station from the mouth of the river (GS3= 2899 ± 718 particles·kg^{-1} DW), located close to the urbanized areas of Sant Jaume d'Enveja and Deltebre (Fig. 4.1). The MPs concentration decreases downstream closer to the mouth of the river (GS2= 1766 ± 821 particles·kg^{-1} DW; GS1= 1491 ± 272 particles·kg^{-1} DW, but no significant difference was found between stations (X_2= 3.8222, df= 2, p= 0.1479).

The distribution of particles type in benthic sediments was led by high abundance of fibers (75.1%), followed by fragments (12.5%), film (10.5%) and the others categories (microbead, foam, and aggregation of fibers; 1.9%).

In river surface water samples, 634 particles were recovered. Fibers were the most predominant abundant type of MPs category (46.1%), but the proportion of fragments (39.4%) and films (13.5%) was relatively more abundant in water samples compared to the others two environmental matrices tested. The MPs abundance did not vary significantly between stations (X_2= 2, df= 2, p= 0.3679). The highest concentration was assessed at the upstream station (WS1: 4.3 ± 0.3 MPs m^{-3}), in the urbanized areas, and lower at the downstream station (WS2: 1.95 MPs m^{-3}), at the nautical club of Riumar.

3.4. Size and colour of microplastics

The size distribution of the particles varied significantly between the three environmental matrices (X_2= 137.83, df= 2, p< 2.2E-16*)*. Pairwise analysis identified significantly larger particles in the benthic sediments, compared with

particles from both sandy beaches (p= 0.0005) and river surface waters (p< 0.0001). When MPs are organized by size, the distribution of MPs is unimodal for all samples (Fig. 4. 4). Most MPs were small microplastic particles sizing less than 1000 µm, the most frequent being particles ranging from 200 µm to 500 µm (29 ± 3%). Similar size distribution was reported, specifically for fibers, in the study conducted by Dris et al. (2016) in Paris. However, this size distribution is in contrast to studies using micro spectroscopy techniques for the identification of the particles (Simon et al., 2018; A. Vianello et al., 2013) that reported higher concentration of MPs as the size of the particles decreases.

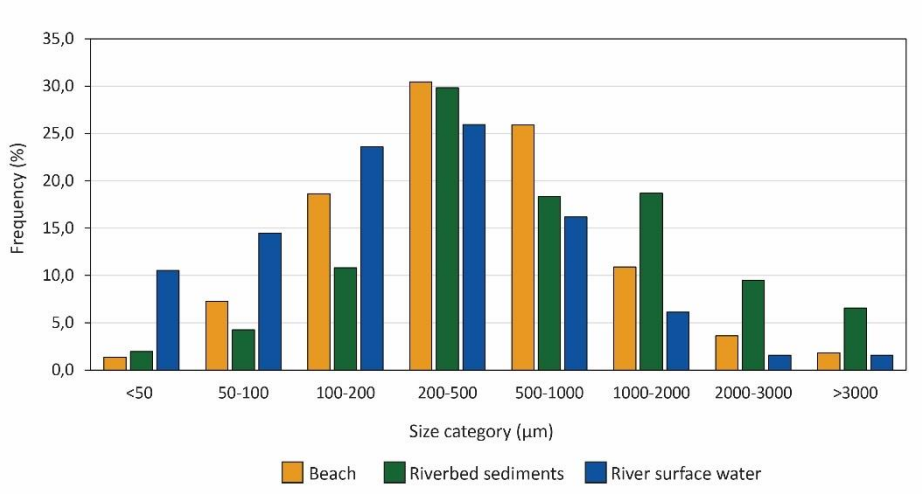


Fig. 4. 4. A comparison of the frequency of size class in the different environments sampled: sandy beaches (ochre), river bed sediments (green) and surface water river (blue).

The color distribution of MPs was investigated for the three different environments (Fig. 4. 5). The colored particles were the most abundant in the three environments, followed by black particles, with the exception of the river surface water samples that showed a higher concentration of white MPs (most of them being fragments; see Figure S4.1).

Fibers were the most common particles found in the three environments sampled. The size and color distribution of the fibers differed slightly between environmental matrices. Most of fibers detected were smaller than 1000 µm

with the highest frequency in the 200-500 µm class, but it is worth mentioning that the highest concentrations of large fibers (>1000 µm) were found in the benthic sediments. Here, these large fibers (33.6% of all MPs in the benthic sediments) were predominantly colored (54.0%) and transparent (35.3%). In comparison, small fibers (<1000 µm, 41.8%) were predominantly colored (51.2%) and black (33.8%) in the benthic sediments as well as in the surface water samples (respectively 41.6% and 36.1%).

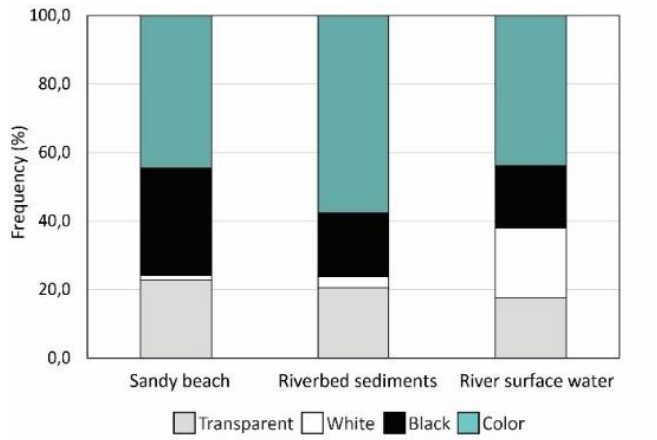


Fig. 4. 5. A comparison of the frequency of color class, Transparent, black, white and color, in the different environmental matrices sampled: sand beach, river bed sediments and river surface water.

The second most abundant type of particles were fragments (Fig. 4. 3). They showed a higher frequency in size of 200-500 µm and were in general, the most abundant particles of less than 500 µm. Those fragments characterizing sand beach samples were colored (42.9%) and transparent (57.1%), while in benthic sediments, the colored category largely prevails (81.6%), and in surface waters the white (50.6%) and colored (38.2%) categories dominate. Films were the third most common type of particles found in the three environmental matrices sampled. Their presence was higher in surface water samples. Those films identified were mainly colored, followed by the transparent and black categories. The presence of microbeads was observed in river surface waters

(n=4), and in benthic sediments (n=1). This type of particles was not found in sand beach samples.

4. Discussion

4.1. Microplastic occurrence in the Ebro Estuary

Estuaries are critical systems for the entrance of MPs to the open ocean. Emerging studies have reported the occurrence of MP in worldwide estuaries across different environmental matrices. MP pollution has been reported in the surface water of the Pearl Estuary (8902 particles·m^{-3}; Yan et al., 2019), in the sea surface microlayer of two South Carolina estuaries, USA (Charleston Harbor: 660 particles·m^{-3} and Winyah Bay: 3080 particles·m^{-3}; Gray et al., 2018) and in two estuarine systems of Southern U.K. (Hamble Estuary: 5380 particles·$m^{-3}$and Beaulieu estuary: 1050 particles·m^{-3};Anderson et al., 2018). These values are three orders of magnitude higher than the MP abundance reported for the Ebro Estuary (3.5 ± 1.4 particles·m^{-3}). In contrast, the concentration reported in the benthic sediments of Changjiang Estuary, China (121 ± 9 particles· kg^{-1} DW; Peng et al., 2017) is one order of magnitude lower than the concentration found in the river sediments of the Ebro Estuary (2052 ± 746 MPs·kg^{-1} DW). Albeit, the comparison of studies is difficult due to the different sampling, extraction/purification, identification methods and concentration units chosen by the researchers to characterize the occurrence of MPs in the natural environment. This first assessment of the MP occurrence in the Ebro Estuary suggests that MP pollution could be considered intermediate-low compared to the values reported in other estuaries. Nevertheless, few limitations of this study should be considered. For the extraction of MPs we used a NaCl solution, which might fail to recovered denser polymers such as PVC or PET. Our recovery test showed an efficiency of 77.5% and 70% for the extraction of MPs from sandy samples and benthic sediment samples, respectively. This indicates that the total concentration of MPs might have been underestimated in these two environmental matrices of the Ebro Estuary. Similarly, particles were identified visually using a stereomicroscope. In the

study conducted by Song et al. (2015), the identification of the particles by FT-IR spectroscopy reported higher MP abundances in comparison to visual identification. It is due to stereomicroscope techniques might neglect smaller and/or transparent particles, especially MPs fragments (Song et al., 2015). Additionally, the visual identification technique does not allow one to distinguish between the synthetic and non-synthetic (natural or artificial) fibers, even if criteria for visual identification are considered (Dris et al., 2015). This limitation of the technique has been reported to overestimate MPs fibers abundance (J.P.G.L. Frias et al., 2016; Hidalgo-Ruz et al., 2012; Song et al., 2015; Ziajahromi et al., 2017).In this study, a subsample of particles was selected for chemical characterization by μFT-IR spectroscopy to validate the visual characterization. The results of the analysis showed that a total of 76% of the particles were correctly identified as MPs.

4.2. Export and accumulation of microplastics in the Ebro river system

The presence of MPs throughout the Ebro Delta, in benthic sediments, river surface waters and sandy beaches corroborates the ubiquitous presence of MPs in aquatic environments. Our results contribute to the understanding of the MPs distribution in river estuaries across the different environmental matrices. MPs concentrations found in the benthic sediments (2052 ± 746 particles·kg^{-1} DW) were higher than those observed in sandy beaches (422 ± 119 particles·kg^{-1} DW). This first observation were reported in other studies that also found high MPs concentration in riverine benthic sediments (Horton et al., 2017; Hurley et al., 2018; Xiong et al., 2019) supporting the hypothesis that estuarine benthic sediments might be an important accumulation pool for MP pollutants. Several factors might induce the sinking of MPs in freshwater environments: 1) the density of the water is lower than in the marine environment, 2) the properties of MPs (density, surface-volume ratio), and 3) the variability of the flow speed due to the geomorphology of the river and human activities (discharges, accumulation, drainage or transfers of water). In the case of the Ebro Delta system, it is important to consider the fact that the Ebro River estuary is highly

stratified with two different water layers (Ibañez et al., 1997): a freshwater layer going from the surface to 1-3 m of water depth and a salt wedge right below. The salt wedge can extend up to ±30 km upstream from the river mouth when discharge is lower than ≈100 $m^3 \cdot s^{-1}$ and up to ±6 km when river discharges over ≈400 $m^3 \cdot s^{-1}$ (Ibañez et al., 1997), suggesting that the three benthic sediments samples were probably collected under the influence of the salt wedge. There are few studies that focus on the influence of salt wedge on the distribution of MPs in estuaries. Vermeiren et al. (2016) pointed out the lack of data regarding the abundance of plastic debris ,-both micro- or macroplastics, accumulating within deltas in relation to the river catchment concentrations. Lima et al. (2015) also showed that the highest densities of MPs within the Goiana estuary (NE Brazil) were found at the bottom waters of the lower estuary during the late rainy season, when the highest river flow induces a flushing of MPs toward the sea. Another study focusing on macroplastics in sediments from Rio de la Plata estuary (Argentina), showed that their concentrations were higher within the salt wedge (Acha et al., 2003). This evidence suggests that the salt wedge could be a critical boundary within an estuary system where increased sinking of MPs is recorded (Vermeiren et al., 2016). In the Ebro estuary, the velocity profiles clearly show that within the salt wedge, the velocity of the flow is low (Table 4. 1, Ibaňez et al., 1997; Nebra et al., 2016) suggesting that once trapped within the salt wedge, MPs will settle and accumulate on the riverbed, simulating sediment deposition dynamics. We found three to six time higher concentrations of MPs in benthic sediments than on sandy beaches. While the fate of MPs in the aquatic and sedimentary systems still remains poorly understood, there are two recognized processes of MPs transportation, which are suspended load and wash load. By contrast, the process of MPs accumulation in benthic sediments of estuaries could be driven by a lower buoyancy that facilitates the entrance of MPs into the salt wedge, where with low flow velocity and longer hydraulic retention time, the sediment flocculation might facilitate the settling and accumulation of MPs on benthic sediments. In contrast to the potential of riverine sediments as storage for MPs, Hurley et al.,

(2018) reported significant reduction in the abundance of microplastics after severe flooding events, with approximately about the 70% of MP pollution accumulated in the river sediments cleansed from the catchments after a high flood event (R. Hurley et al., 2018). Hence, further data on the accumulation time of these pollutants is needed to understand the dynamic on the fluxes of MPs to the oceans as well as to evaluate the level of exposure of benthic organisms to MPs pollutants.

The level of microplastic pollution in the surface waters of Ebro river (3.5 ± 1.4 particles m^{-3}) was below than other values reported in other studies, such as the Seine river (3-108 particles·m^{-3}; 80 µm mesh size) (Dris et al., 2015) or the Pearl River of China (379-7924 particles·m^{-3}; 20 µm mesh size) (Lin et al., 2018). The comparison of MPs abundance between areas is difficult and subject to uncertainty due to different sampling and analysis approaches. To some extent, the higher abundance of MPs could be expected in rivers located closer to urban areas under high anthropogenic pressure in contrast to the Ebro estuary, a high naturistic area with relatively low anthropogenic and demographic pressure (75.8 inhab·km^{-2}; IDESCAT, 2018). However, some additional factors such as the management of the Ebro River basin itself might influence the occurrence of MPs in the Ebro Delta area. Previous studies showed that the concentrations of MPs in surface water are usually higher upstream of a dam than downstream (Zhang et al., 2015). As was previously mentioned, the Ebro River contains one of the biggest dams of the Iberian Peninsula that affects the natural balance of water and sediments (Palanques and Guillén, 1998; Zografos, 2017). Thus, the dams of Mequinenza and Ribaroja when retaining water and also the underneath sediments can be a potential accumulation pools for MPs. However, further studies are needed to assess the role of dams in the fate of MPs, as these infrastructures might act as barriers retaining MPs and preventing them to reach the marine environment.

The distribution of MPs along the beaches of the northern delta suggests that once discharged by the river, the MPs are partially accumulated on the delta

beach. If only stations S1 to S4 are considered (i.e., within the delta system), the highest concentrations were observed at S1 located near the mouth of the river (S1) and next to the Desaigua del Pal (S4) (Fig.4. 1 and 4.3). Whilst for the station S2 and S3, the concentrations decrease as one moves away from the river mouth. On the other hand, the highest concentration of MPs was measured at S5, a beach outside of the geographical boundaries of the Ebro Delta (Fig.4.3). Although this concentration was slightly higher than at the river mouth (+11%) or next to the Desaigua del Pal (+17%), it remains in the same order of magnitude. The proximity of urban areas such as Cap Roig and Perellò might influence the highest MPs concentration observed in S5. However, the interaction of multiple factors might contribute to the heterogeneous variability of the MPs abundance between the stations, such as prevailing winds, currents, proximity to a point-source of pollution, diffuse pollution, urbanization and socioeconomic activities (Gray et al., 2018).

The variability of MP distribution along the Ebro Delta clearly shows a decreasing gradient from the upstream stations (WS1 and GS3) next to the town of Deltebre toward the river mouth (WS2 and GS1, Fig. 4. 1 and 4.3).A similar pattern was observed in the surface waters of the Douro river estuary, Portugal (Rodrigues et al., 2019): higher MP occurrence was found in the middle part of the estuary close to urban areas and lower MP concentrations in the lower part of the estuary. This gradient suggests that the flux of MPs reaching the open sea is lower than the inputs to the river basin due to the retention factor of the catchment. Nevertheless, the MP concentration recorded at WS2 (1.95 particles m^{-3}) could represent about 2.14 x 10^9 MPs·yr^{-1} reaching the Mediterranean Sea within the top layer (15 cm) of the Ebro River, considering the annual flow of the Ebro River 464 $m^3 \cdot s^{-1}$ (stream gauge A027-Tortosa, CHE). However, the estimation of the net flux of MPs to the Mediterranean Sea from the Ebro River requires further understanding on the fluxes of accumulation and deposition of MPs in the littoral area of the Ebro Delta as well as studies on the vertical distribution of MPs in the water column in the estuarine area. Therefore, there

is a need to present complete MP concentration profiles in the different compartments of estuaries, in order to provide more accurate estimation of the MP fluxes from rivers to the marine environment.

4.3. Tracking potential sources of MPs in the Ebro Delta

Here, we have discussed the potential sources of MPs considering the features and anthropogenic activities in the lower part of the Ebro River. The most common particles found in the collected samples were fibers (70 ± 22%), as shown in previous studies that reported the presence of large amount of fibers in aquatic ecosystems (Claessens et al., 2011; Peng et al., 2017; Thompson et al., 2004). The presence of this type of MP in aquatic environments is mainly attributed to wastewater effluents (Magnusson and Norén, 2014; Talvitie et al., 2015), as consequence of clothes washing. Browne et al. (2011) concluded that up to 1900 fibers can be released washing a single garment of clothing. In the studied area, there are two main WWTP that discharge directly into the Ebro River, and possibly 2 others indirectly (Fig. 4.1). The two WWTP that discharge directly into the river are located in the towns of Amposta and Deltebre, located 27 km and 14 km from the mouth of the Ebro River, respectively (Fig. 4.1). Both use a secondary treatment (biological degradation), which is not able to remove all the textile fibers from domestic and commercial laundry (Ziajahromi et al., 2017). In particular, Browne et al. (2011) found that the predominant type of fibers were polyester, acrylic and polyamide at the effluents of WWTP. These polymers were also identified in the Ebro Delta. Albeit it is difficult to track the origin of the fibers found in the three matrices, the main source of MPs pollution in the Ebro estuary could be related to the discharge of wastewater. Nevertheless, other sources cannot be disregarded as potential sources of fibers. Fishing and aquaculture activities, which have a relevant role in the study area, are other potential sources of MPs. Both use materials made of synthetic fibers and their degradation or direct disposal in the aquatic environment might lead to their degradation to the size of MP (Andrady, 2011). Finally, there might be contribution from the

atmospheric compartment, which has not been thoroughly investigated. Fibers can be transported by wind and deposited in terrestrial and aquatic environments (Dris et al., 2016), especially in the vicinity of populated areas.

In contrast to fibers, the presence of secondary MPs (fragments, film, foams) was observed in the lower portion of the Ebro Delta (37%). The origin of these types of MPs lies in the breakdown of larger plastics. One input of this type of MPs to the watercourse is related to the potential of rainfall run-off transporting plastic pieces already exposed to environmental factors (Horton et al., 2017a). Recent studies showed that MP pollution in soils is as severe as in aquatic environments (He et al., 2018). Particularly in the Ebro Delta, agriculture practices might enhance the release of MPs accumulated in the soil matrix to the aquatic environment. Agriculture is the main economic activity in the study area where more than 65% of its surface is devoted to intensive rice production (Genua-Olmedo et al., 2016). Briefly, the rice crop production in the area involved practices such as ploughing, flooding, sowing, re-flooding, and draining the fields before the harvest. The practice of ploughing might help to release those MPs trapped in soils, while the practices of flooding and draining will act similar to rainfall run-off, dragging MPs to the aquatic environment. Thus, cultivation practices associated with the production of flooded crop should not be neglected as a potential source of MPs in the aquatic environment.

5. Conclusion

The present study contributed to the scientific understanding of the MP fluxes accumulating in estuaries and the MP fluxes flowing into the open ocean. The results showed the widespread distribution of these pollutants across the different environmental matrices investigated in the Ebro Delta. The MPs concentration reported suggest that rivers (1) act as direct corridors for MPs to reach the marine systems, (2) once MPs are in the sea, the currents and tidal dynamics are responsible for partially depositing some of these particles in the littoral area, and (3) estuarine sediments are important sink areas for MP pollutants. Particularly in salt wedge estuaries, hydrological and sediment

transport dynamics might be responsible in facilitating the deposition of MPs onto bottom sediments. Considering that most of the world population is concentrated along rivers, estuaries and coastal areas, with the current plastic production, consumption and waste management system, the concentration of plastics and MPs in the aquatic ecosystems is expected to increase. More extensive and regular monitoring of MPs is required across river catchments, especially estuaries, to fully illustrate the partitioning of MPs across different environmental matrices and therefore assessing the level of hazard and exposure of these pollutants to aquatic organisms and, by extension, to human health.

CHAPTER 5

Sediment cores revealing the plastic age?

Microplastic preservation in coastal sedimentary records

Abstract

The seafloor is the major sink for microplastic pollutants. However, there is a lack of robust data on the historical evolution of microplastic pollution in the sediment compartment, particularly the sequestration and burial rate of small microplastics (<10-1000 μm). By combining a palaeoceanographic approach and state-of-the-art analytical methods for microplastic identification, we present the first high-resolution reconstruction of microplastic pollution from an undisturbed sediment core collected in the NW Mediterranean Sea. Furthermore, we investigate the fate of microplastics once buried in the sediments by evaluating the changes in the size distribution of the microplastics and the weathering status of the polyolefins, polyethylene, and polypropylene. Our results indicated that the microplastic mass sequestered in the sediment compartment mimics the global plastic production from 1965 to 2016. We observed an increase in the weathering status of the polyolefins as the size decreased. However, the variability in the size and weathering status of the microplastics along the sedimentary record indicated that these pollutants, once incorporated in the sediments, remain preserved with no further degradation under no remobilization events.

Chapter based on: Simon-Sánchez, L., Grelaud, M., Lorenz, C., Garcia-Orellana, J., Vianello, A.,Liu, F., Volltersen, J., Ziveri, P., (*Submitted*). Sediment cores revealing the plastic age? Microplastic preservation in coastal sedimentary records

1. Introduction

The Anthropocene epoch frames the geological time when intensified anthropogenic activities induced mounting changes in Earth-system processes (Crutzen, 2002). This time has been characterized by manufacturing new materials that are indispensable in our societies. Among these materials, plastics stand out. Not only does the global plastic mass outstrip the living animal biomass on Earth (Elhacham et al., 2020), but the rate of production and disposal of these materials already exceeds the planetary boundary (Persson et al., 2022). Moreover, their durability favors their plastic preservation as potential long-lasting distinctive markers in sedimentary records (Zalasiewicz et al., 2016). The increasing presence of plastics in our oceans raises concerns about the harm they represent to the ecosystems' functioning, from alterations to the marine carbon cycle (Smeaton, 2021) to individual ecotoxicological damage (Hamm and Lenz, 2021). Yet, elucidating the dispersion and accumulation of microplastics (MPs; <5 mm) and the elusive nanoplastics (NPs; <1µm), remain challenging as fluxes, fate, and residence time, are still poorly understood, partly due to the constraints of available analytical methods (Primpke et al., 2020a; Zarfl, 2019). The growing body of data on the presence of MPs in the marine realm points to the seabed as a significant sink for these pollutants (Woodall et al., 2014). The mechanisms for reaching this deep environment are primarily related to the MPs density, as buoyant MPs are expected to float at sea. However, MPs can sink in the water column (Pabortsava and Lampitt, 2020) and be transported by deeper currents (Kane and Clare, 2019). Furthermore, physico-chemical changes caused by fragmentation, weathering, and biological interaction, such as ingestion-egestion, aggregation with organic and inorganic matter, and biofilm formation, might facilitate the MP export to the ocean floor (Van Sebille et al., 2020 and references therein). Once on the seafloor, the fate, depositional trends, and environmental degradation of MPs, especially of the smaller fraction (<300 µm), remain unraveled (Martin et al., 2021).

Microplastics buried in sediments it can interact with benthic biota (Carreras-Colom et al., 2020; Cau et al., 2019). These pollutants potentially can also be used as a chronological tracer of sedimentary records (Bancone et al., 2020). However, its preservation and degradation in sediments and therefore its effectiveness as a temporal tracer has not been explored in detail. Depositional environments, such as river prodeltas, offer high-resolution stratigraphy over the last decades, making it suitable to track the evolution of MP pollution. Moreover, rivers play an essential role as a source of MP to the Mediterranean Sea and holds a relevant storage capacity for these pollutants (Simon-Sánchez et al., 2019). In this study, we investigate MP accumulation over time and the fate of these pollutants once buried in sediments. We combined different analytical approaches to reconstruct the accumulation of small MPs (11-1000 µm), including geochronology methods and state-of-the-art FPA-µFTIR-Imaging (Focal Plane Array-Fourier Transform Infrared-Imaging-Micro-Spectroscopy). The radiometric analysis (^{210}Pb,^{137}Cs) provides robust chronologies for the Plastic Age (i.e., post-1950s) with years to decades resolution (Bancone et al., 2020), while FPA-µFTIR-Imaging spectroscopy allows reliable detection of particles down to 11 µm in size, avoiding analyst's bias (Löder et al., 2015). Besides, we explore the degradation status of these pollutants once buried in the continental shelf by quantifying the weathered status of the polyolefins, polyethylene (PE), and polypropylene (PP). The investigated polymers represented, as of 2015, 50.3% of the global plastics waste generation (Geyer et al., 2017), and despite their positive buoyancy, PE and PP are the most abundant polymers reported in sedimentary environments (Abel et al., 2021; Int-Veen et al., 2021). We discuss our findings in the context of previous microplastic observations, the variability of MP properties over time, and the environmental factors leading to the MP sequestration and preservation in the seabed.

2. Methods

2.1. Core material

The sediment core ST17_MUC2 (40.7726º N, 1.1643º E; 104 m water depth, K/C Denmark Multi Corer) was recovered during the MERS_BI cruise in November 2019 onboard the R/V Sarmiento de Gamboa, in the Balearic Sea (NW Mediterranean; Fig. 5. 1. A). The selected sedimentary record (37 cm in length) was sliced onboard every centimeter. The samples were transferred to tared light-density polyethylene (LDPE) zip lock bags and stored at 4 °C. In the laboratory, the sediments were homogenized and dried at 50 °C until a constant dry weight (DW) was reached.

2.2. Age model for ST17_MUC2

Dried sediment samples were analyzed to determine ^{210}Pb specific activities by α-spectrometry through the analysis of its granddaughter ^{210}Po at the Grup de Recerca en Radioactivitat Ambiental de Barcelona (GRAB) at the Univesitat Autònoma de Barcelona – following the method described in Sanchez-Cabeza et al. (1998). After adding ^{209}Po as an internal tracer, 200–300 mg of sediment aliquots were totally digested in acid media using an analytical microwave oven, and Po isotopes plated on silver discs in HCl 1N at 70 °C while stirring for eight hours. Alpha emissions of ^{210}Po and ^{209}Po were measured using Passivated Implanted Planar Silicon detectors (PIPS; CANBERRA, Mod. PD-450.18 A.M.).

Age model derived from ^{210}Pb was obtained using the Constant Supply: Constant Flux (CS:CF) model (Krishnaswamy et al., 1971). The mean mass accumulation rate (MAR; g cm^{-2} yr^{-1}) was obtained by performing a non-linear least-square fit between the excess ^{210}Pb (Bq kg^{-1}) and the cumulative dry mass (g cm^{-2}) of the sediment core. The excess ^{210}Pb (^{210}Pb$_{xs}$) was determined by subtracting the constant ^{210}Pb supported, which was estimated as the average ^{210}Pb concentration of the deeper sediment layers analyzed, wherein ^{210}Pb activities reached constant values. To calculate the sedimentation rate (SR; cm yr^{-1}), the MAR was divided by the dry bulk density (DBD) of the section, where

the DBD of the section is obtained by dividing the DW mass (g) of the slice by its volume (cm^3).

The activities of ^{137}Cs were measured.using a HPGe γ-spectroscopy detector (CANBERRA, Mod. GCW3523 ad Mod.SAGe Well). Aliquots of dry sediments (3-5 g) were placed into PE counting vials using calibrated geometries and counted for around 186,000 seconds.

2.3. Microplastic analysis

The upper 10 cm of the sediment core at one cm resolution were analyzed for MP content following the protocol described in Liu et al. (2019).The sediment mass processed for MP analysis varied between slices, ranging between 22.8 and 59.5 g (Table S1). Briefly, the sample matrix was removed using a multi-step sample treatment including pre-oxidation, density separation, buffered multi-enzymatic treatment, and catalyzed oxidation. The resulting isolated particles were transferred into a glass 10 mL headspace vial with HPLC grade 50% ethanol, and the solvent was evaporated using an evaporator (TurboVap® LV, Biotage). Finally, a fixed volume (3 mL of HPLC grade 50% Ethanol) was used to re-mobilize the particles. After homogenization (vortex), multiple aliquots of the suspension were deposited with a capillary glass pipette (micro-classic, Brand GmbH, Germany) onto an area of 78.5 mm^2 (⌀10 mm) of a zinc selenide window (ZnSe – ⌀13 mm, 2 mm thickness, Crystran LTD, UK) held by a compression cell (Pike Technologies, USA). The deposited sample was dried overnight at 55 °C prior to analysis. To minimize data extrapolation, the total volume of each sample was deposited onto multiple windows (four to five per sample) and analyzed.

Sample analysis was carried out using FPA-μFTIR-Imaging spectroscopy. Measurements were performed using an Agilent 620 FTIR microscope equipped with a 128 × 128 pixel MCT - FPA detector (Mercury Cadmium Telluride – Focal Plane Array) coupled with a Cary 670 FTIR spectrometer (Agilent Technologies, Santa Clara, CA, USA). Optical images of the ZnSe window were determined with a 15× objective. The IR map was collected in

transmission mode in the range of 3750 to 850 cm^{-1}, using a 15× IR Cassegrain objective-condenser system with a spectral resolution of 8 cm^{-1}, 30 co-added scans for the sample, and 120 for the background. This setup allowed measuring the whole area of the ZnSe window (⌀10mm, 78.5 mm^2) with a pixel resolution of 5.5 × 5.5 µm.

The resulting hyperspectral images were analyzed for systematic automated MP identification with the software siMPle (Primpke et al., 2020b). After converting the recorded spectra from %Transmittance (%T) to Absorbance (Abs) and performing baseline correction (Primpke et al., 2018), the software chemically identifies the particles on the sample by comparing every pixel of the IR map with a custom-built library containing 441 spectra of inorganic and organic materials. The resulting scores are then used to provide a material-based map of the sample, quantitative data on particle abundance, and detailed physico-chemical information for each particle (polymer composition, two-dimensional size, estimated volume, and mass).

2.4. Contamination

Sample preparation was conducted inside a laminar flow bench (Telstar AV-100), and cotton lab coats were worn. All the reagents (e.g., H_2O_2, $ZnCl_2$, NaOH, $FeSO_4$, and enzymatic buffers) were filtered through a glass fiber filter (0.7 µm, Whatman) prior to use. Only glassware or stainless-steel materials were used whenever possible, except for the density separation step where silicon tubes and polytetrafluoroethylene (PTFE) stopcocks were unavoidable. All the materials were carefully rinsed three times with MilliQ® and immediately covered before use. Besides these measures, one analytical blank was run alongside each set of samples to assess contamination.

A section of the core below the Plastic Age (pre-1950s, 34-35 cm) was selected and analyzed along with the samples to account for potential contamination during sampling. We assumed that the MP recovered in this section represents the potential contamination during the coring and storing, since the existence

of plastic materials in these sections would correspond to the 19th century, according to the age model.

2.5. Data Analysis

For each sample, the results of each ZnSe window were combined and corrected for contamination. The contamination recorded for the sampling and in the analytical blanks were subtracted from the samples, considering the size class and polymer composition. The total amount of MPs (kg^{-1} DW and $\mu g\ kg^{-1}$ DW), MP fluxes (items $m^{-2}\ yr^{-1}$ and $\mu g\ m^{-2}\ yr^{-1}$), and polymer diversity were then calculated. The shape of particles was classified as fragments or fibers, according to Vianello et al. (2019), while size classes were adopted from Lorenz et al. (2019). Microplastic burial rates ($m^{-2}\ yr^{-1}$) were calculated by multiplying the MP concentration by the MAR of the core and standardized to m^2. Polymer diversity across the sediment core was assessed using the Shannon-Wiener diversity index (H') and Pielou's evenness index (J'). Data normality of the dataset was tested using the Shapiro Wilk test. Non-parametric tests (Kruskal-Wallis, followed by a post hoc Wilcoxon-Mann-Whitney) were applied to reveal differences. Data analysis and figures were produced in QGIS Desktop 3.12 'București' (QGIS Development Team, 2020) and R-4.1.1 (RStudio Team, 2020), using ggplot (Wickham, 2016), and cowplot (Wilke, 2020) packages. The level of statistical significance was set at $p < 0.05$.

2.6. Carbonyl index

The degradation status of polyolefins (PE, PP) was investigated using the carbonyl index (CI), which allows identifying the chemical changes in several polymeric materials by targeting the specific absorption band of the carbonyl species produced mainly during thermo- and photo-oxidation (Andrady, 2011; Ter Halle et al., 2017b). The CI was computed adopting the specified area under band (SUAB) method described in Almond et al. (2020), applying the equation:

$$CI = \frac{integrated\ area\ under\ band\ 1850{-}1650\ cm^{-1}}{integrated\ area\ under\ band\ 1500{-}1420\ cm^{-1}} \qquad (Eq.1)$$

Spectra of the particles were recorded in transmission mode through the entire particle's thickness, including the spectral contribution from the particle's inner core. All the spectra belonging to polyethylene and polypropylene particles were automatically exported from each sample dataset using a custom-design feature in siMPle. The exported spectra were loaded into SpectraGryph 1.2.15 software (Menges, 2016); the integrated areas under the selected bands were calculated using the peak analysis tool and used to compute the CI measurements.

3. Results and discussion

3.1. Sediment archive

The $^{210}Pb_{xs}$ profile of the sediment core showed an almost constant and continuos sediment accumulation rate (Fig. S5. 1). The application of the CS-CF dating model resulted in a SR of 0.12 ± 0.01 cm yr^{-1} The age model determined the beginning of the Plastic Age (50's) around the 10-11 cm (1951 ± 3 yr). The artificial radionuclide ^{137}Cs investigated to validate the geochronology did not provide discernible activities. This limitation can be related to the limited sediment mass analyzed, the decay of^{137}Cs, the low concentration in sediments, and its potential mobility (Arias-Ortiz et al., 2018). Nevertheless, in its absence, historical events can corroborate the ^{210}Pb geochronology. The Ebro River discharge records showed two marked trends after 1951, and after the maximum recorded in 1959 (CHE Ebro) was followed by a significant decrease in the annual contribution within the following years. Remarkably, the Ribaroja and Mequinenza dams located around 100 km upstream, whose constructions dated from 1958 to 1966, heavily impacted the Ebro River's water and sediment flow balance (Zografos, 2017). The anomaly of $^{210}Pb_{xs}$ observed in the general exponential trend between 8.5 and 10.5 cm corresponds to the years 1965 ± 2 and 1951 ± 3, coinciding with the years of construction of the dams, a fact that adds confidence to the chronological model obtained.

3.2. Microplastic concentration

Microplastics were successfully extracted and analyzed in nine out of the 11 samples, except for two of them (4-5 cm and 9-10 cm) that were lost during sample preparation. Microplastics were found in all the investigated sections of the sediment core. Fig. 5. 1 shows the MP concentration in the sediment core recovered in the Ebro prodelta. A total of 902 MPs ($1.39 \cdot 10^5$ ng) were recovered, considering the blank corrections (Table S5.1). The total MP abundances ranged from 706 to 6939 items kg^{-1} DW. The mass concentration ranged from $5.00 \cdot 10^{-2}$- 1.43 mg kg^{-1} DW. The highest and lowest concentrations were found at the surface of the core (0-1 cm) and in section 7-8cm, respectively. The highest mass concentration was recorded in the 1-2 cm of the core, whereas the lowest was in the 8-9 cm.

To our knowledge, there are no previous observations in the literature of MP abundance in sedimentary records combining palaeoecological approaches (i.e., radiometric analysis, varve counting) with measurements of imaging μFTIR for MP identification. Hence, this cutting-edge study provides one of the first high-resolution records of MP accumulation in coastal environments. However, this method was applied to characterize the MP presence in marine sediment samples, where the upper 5 cm of the seabed were retrieved using sediment corers (Abel et al., 2021; Bergmann et al., 2017) or a van-Veen grab sampler (Lorenz et al., 2019). We calculated the MP abundance for the top four cm (3122 items kg^{-1}) of our sediment core for the comparison. In the remoteness of the Kamchatka trench, northwest Pacific Ocean, MP concentrations were one to two orders of magnitude lower (14-209 items kg^{-1}; Abel et al., 2021) than our findings. Similarly, lower concentrations (3-1189 items kg^{-1}) were reported in the southern part of the North Sea (Lorenz et al., 2019). In contrast, Bergman et al. 2017 reported higher MP concentrations (42–6595 items kg^{-1}) at the Fram Strait, west of Svalbard. In the Mediterranean Sea, the closer analytical methods can be attributed to Vianello et al. (2013), who found relatively lower values (672-2175 items kg^{-1}) in the sediments of the Lagoon of Venice, Italy. These observations agree with our previous results (Simon-Sánchez et al., 2019) that

despite the Ebro River being a critical system for understanding the MP fluxes entering the Mediterranean Sea, and being under the influence of the Gulf of Lion current, the MP pollution levels in this system are intermediate to-low.

3.3. Microplastic sequestration and burial rate

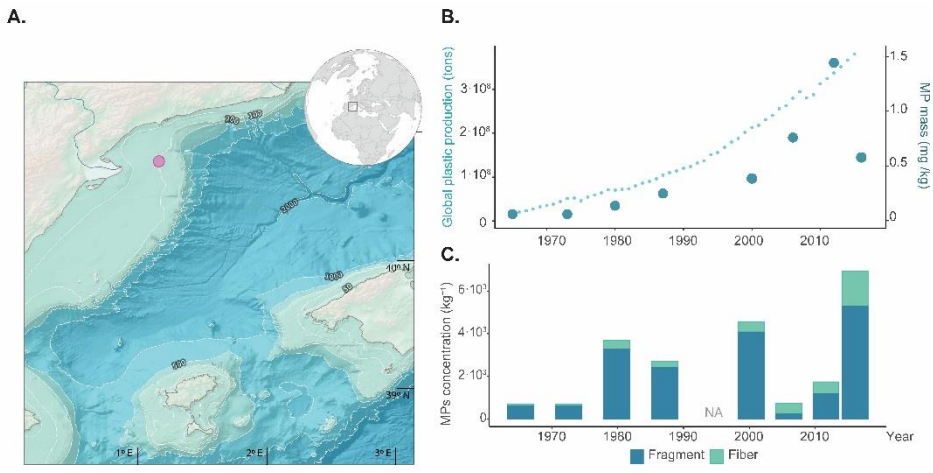


Fig. 5. 1. Geographical location of the sampling station. B. Abundance of mass-microplastic registered in the sediment core (y-axis right), against the global plastic production (y-axis left) from 1965 to 2016. C. Abundance of microplastic (number) classified as fragments and fibers, from 1965 to 2016.

The accumulated MP inventory since 1965 ± 2 was $1.44 \cdot 10^6$ items m^{-3} ($2.23 \cdot 10^{-1}$ g m^{-3}). MP burial rate ranged from 865 m^{-2} yr^{-1} in 1973 ± 2 to 8507 m^{-2} yr^{-1} in 2016 ± 1. The MP burial rate has increased 973% since 1965, with an average standardized rate of 18% per year. The mass of MP sequestered in the sediments ranged from 0.61 mg m^{-2} yr^{-1} in 1965 ± 2 to 1.76 mg m^{-2} yr^{-1} in 2012 ± 1. Kaandorp et al. (2020) calculated the sinking flux of plastics, considering all size classes, in the Mediterranean Sea over 2006 - 2016. They estimated that the sinking plastic fluxes from Algerian to the Spanish coast ranged from 0.1-1.0 g km^{-2} day^{-1}. Our results showed that in 2016 ± 1, the sinking mass of small MPs almost doubles their higher estimates value (1.89 g km^{-2} day^{-1}). There should be noted the relevance of the study area, river deltas are vulnerable systems subjected to upstream anthropogenic stressors and recognized

accumulation areas for several pollutants (Foufoula-Georgiou et al., 2011; Simon-Sánchez et al., 2019).

3.4. Are microplastic reporting units relevant?

In contrast to previous studies (Martin et al., 2021 and references therein), no significant correlation was found between the abundance of MPs (kg^{-1}) and the sediment depth (Pearson's, r= -0.52, p= 0.19; Fig. 5. 1. B-C). The MP fluxes reaching and accumulating in the sediments were reported to directly correlate with global plastic production, growing population (Brandon et al., 2019), and landscape changes by using plastic materials (e.g., greenhouse; Dahl et al., 2021). We acknowledge that as the global plastic mass and plastic waste production exponentially increase, it is unsurprisingly to find a negative correlation between MPs abundance and sediment depth in the core. Nonetheless, to compare and assess the plastic sinking fluxes and sequestration in the sediments regarding the global mass plastic produced, the unit MP mass concentration is more appropriate than the number of particles. The estimated MP mass sequestered in our sediment core over time shows a significant trend with sediment depth (Pearson's, r= -0.79, p< 0.05), as well a similar exponential trend was observed between the MP mass sequestered in the sedimentary records of Ebro prodelta and the global mass plastic production (Fig 5. 1.B).

Reconstructing the MP export (number of particles) to the benthic environment based on plastic production and waste generation requires the assumption that MP deposition is spatially homogeneous, constant, and increasing exponentially over time. In general, MP studies reported patchiness in the spatial occurrence of these pollutants across different environmental compartments (Korez et al., 2019; van der Hal et al., 2017; Vianello et al., 2018). When comparing the MP abundances at the sea surface and the sediments lying beneath, the concentrations and polymer composition significantly differed (Lorenz et al., 2019). The assumption of constant deposition of MP oversimplifies the complex and diverse compounds that MPs are. MPs compromise a wide size range (1-5000 µm), morphologies, specific

densities, and multifaceted chemical compositions (Frias and Nash, 2018; Hartmann et al., 2019) that undoubtedly affect their behavior and fate under natural environmental conditions (Rochman et al., 2019). Despite an incontestable increasing trend, the MP sequestration (number of particles) shows intra-variability over the last 54 years (Fig 5.1. C). This variability is likely driven by the synergistic combination of the heterogeneous distribution of microplastics along the surface waters of the Mediterranean Sea (Simon-Sánchez et al., 2021) and the different mechanisms, which are still poorly understood, leading the MP export from the surface to the benthic environment (e.g.: marine snow, biofilm formation, aggregates, ingestion-egestion, vertical migration of species, deep-ocean currents, ocean turbulence). Furthermore, these processes can affect the MP physico-chemical properties (de Haan et al., 2019; Mateos-Cárdenas et al., 2020; Tu et al., 2020). In our sediment core, the median MP size (58.7 μm, Q_1-Q_3: 41-91 μm) agrees with small MPs' prevalent dominance in the sediment compartment (Martin et al., 2020). Overall, this size fraction was reported to be scarce at the sea surface (Egger et al., 2020), highlighting the need for MP knowledge along the water column to identify processes leading the vertical and understand if these mechanisms are polymer- and size-selective by which the MP composition sequestered in the sediments may be shaped.

3.5. Microplastic characterization

The majority of MP found in the sediment core were fragments (Fig.5. 1.C), which accounted for 83.3% of the total particles. In contrast, Dong et al. (2020) exclusively found fibers along the sediment records recovered in the Donghu urban lake, China. Similarly, fibers were the predominant MPs morphology in the sedimentary records investigated in the Santa Barbara Basin (77.0%; Brandon et al., 2019) and in the Rockall Trough, North Atlantic Ocean (89.0%; Courtene-Jones et al., 2020). Noteworthy, the methods described in these studies implied visual presorting and isolation of the potential plastic particles for spectroscopy analysis. This visual approach is prone to analyst bias as fibers are easier to recognize than smaller MP fragments (Song et al., 2015).

Moreover, fibers are one of the primary sources of contamination in MP analysis (Torre et al., 2016). Thus, applying robust and quantitative protocols is essential to prevent contamination during sampling and sample preparation. Far more critical when asynchronous particles may jeopardize the interpretation of the MP depositional record.

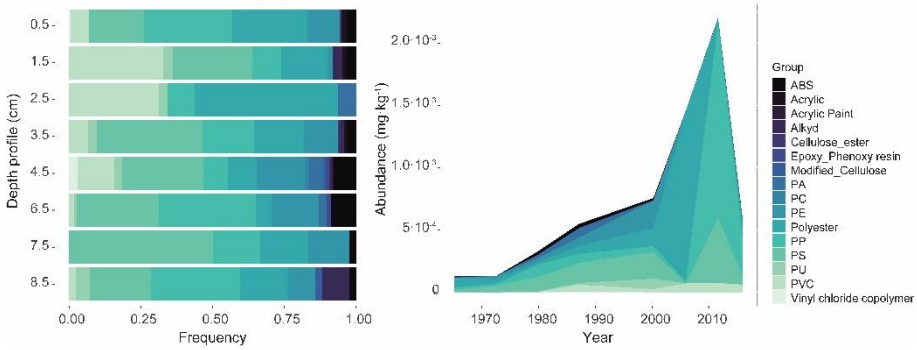


Fig. 5. 2. A. Microplastic polymer composition in the sediment profile. B. MP mass in the sediment core according to polymer composition from 1965 to 2016.

A total of 16 different synthetic polymers were identified in the sediment core (Fig. 5. 2). The most abundant were polystyrene (PS; 28.4%), followed by PP (22.4%), polyester (PET;15.9%), PE (12.6%), polyvinyl chloride (PVC; 8.5%), acrylonitrile butadiene styrene (ABS; 5.9%); polyurethane (PU; 1.9%), polyamide (PA; 1.7%), alkyds (1.1%) and others (2.5%). The synthetic polymers between sections ranged between 5 and 14 (Fig. 5.2. A). The Shannon-Wiener (H') values ranged from 1.0 in section 7-8 cm to 2.1 in section 5-6 cm, corresponding to 1973 ± 2 and 1987 ± 1, respectively. No significant trend was found between the polymer diversity and sediment depth (Pearson's, r = -0.04, p = 0.912). The synthetic polymer diversity evenness (J') was equal (J’ = 0.8) in all the sections investigated, except for section 7-8 cm (J’ = 0.6). Noteworthy, the MPs mass sequestered in the sediments sorted by polymer (Fig. 5.2. B.) showed a similar exponential growth by polymer group until early 2000. After 2006 ± 1, the major mass contribution is driven by the sequestration of PP (43.6%), PET (35.8%), and PS (14.0%).

3.6. Carbonyl index

FTIR spectroscopy is one of the most common techniques for MP characterization (Primpke et al., 2020). The potential of this technique was broadened to assess the aging of the polymers under natural environmental conditions (Brandon et al., 2016; ter Halle et al., 2017) and accelerated weathering in the laboratory (Kim et al., 2022). The measurements are generally gathered with attenuated total reflection (ATR)-FTIR recording the changes occurring at a single point of the particle's surface, and being the analysis limited to particles >300 μm in size. Noteworthy, the degradation of the particle may not occur evenly across the MP. In this study, spectra of small MPs (11-1000 μm) were collected in transmission. Under this setup, the limitation of the measurement is defined by the thickness of the particle as the IR light passes through the sample, recording the particle's inner core as well as its surface. However, by using hyperspectral images, where several spectra are measured per particle (Fig. 5.3), it is possible to characterize the weathering status across the MPs whole dimension in contrast to ATR-FTIR measurement. Furthermore, the reported method shows the potential of transmission measurements for computing the CI of small MPs in a standardized and automatic manner.

A total of 245 polyolefins particles were used to compute the CI, 83 PE, and 172 PP particles. These particles yielded a total of 11337 and 21661 spectra, respectively. Due to non-normal distribution, the median value was computed to describe the CI per particle. The overall median CI for PE was 1.05 (Q_1-Q_3: 0.90-1.25) and for PP 0.48 (Q_1-Q_3: 0.23-0.84). The SUAB method produces significantly higher CI values than previous methods, which statistically rejects the results' intercomparison (Almond et al., 2021). No other study has previously used this method to investigate MPs' weathering under natural environmental conditions. Potrykus et al. (2021) applied it to characterize the chemical modifications on a PP plastic sample after five years of degradation in a landfill. The authors reported a CI ranging from 0.37 to 1.29. However, it

should be noted that plastic degradation in seawater occurs severely slower than when exposed to sunlight in dry conditions (Andrady, 2011).

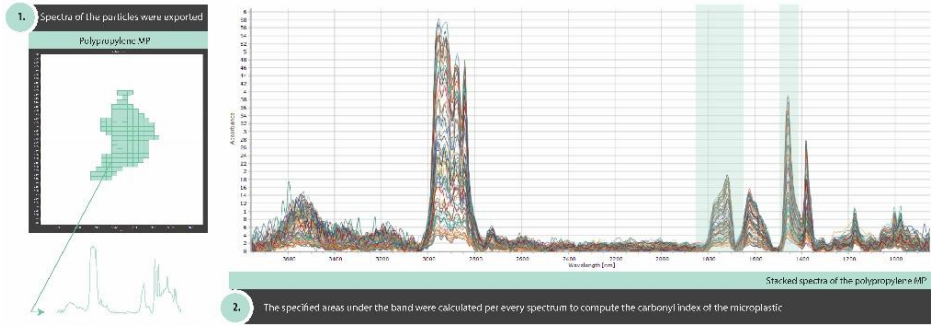


Fig. 5. 3. Illustrative example on the calculation of the carbonyl index for a polypropylene particle. From left to right, image on the particle's visualization, every pixel represents a collected spectrum. Spectra were exported from the siMPle software and treated in SpectraGryph 1.2.15 where the area below of the band was computed.

Along with the depth profile (Fig. 5.4. A), no significant differences were found for PP (Kruskal-Wallis, chi-squared = 11.379, df = 6, $p = 0.077$). In contrast, significant differences were found for the CI of PE (Kruskal-Wallis, chi-squared = 15.732, df = 6, $p < 0.05$), between section 5-6 cm and 7-8 cm ($p < 0.05$). Several uncertainties limit the explanation of the CI variability over time. First, although the MP oxidation may occur in benthic environments under aerobic conditions, the oxygen-rich products that allow CI estimation mainly occur under light-exposure conditions. Furthermore, this process is severely retarded in seawater and further impaired by biofilm formation (Andrady, 2011). Secondly, polyolefins' proneness to photooxidation is defined by the original chemical composition that might highly vary depending on the additives (plasticizers, retardants, antioxidants, stabilizers) used during their manufacturing. In this context, the interpretation of CI results relies on the uncertainties of the MP initial chemical composition, the distance to the sources, and the exposure time to natural conditions before its sequestration in the sediment.

A linear regression using the CI of PE and PP, and the major dimension of these polyolefins (logarithmically transformed) indicated a tendency that smaller

MPs were more oxidized (Fig. 5. 4. C). However, no significant correlation was found (PE: R^2=0.003 and PP: R^2=0.160). Overall, this observation agrees with the affirmation that MP degradation leads to particles' embrittlement, which favors the fragmentation of these pollutants (Andrady, 2011). However, our results indicate that this process occurs previously to sequestration into the sedimentary compartment, where MPs are accumulating with no signal of further physical degradation.

3.7. Is MP degradation an active process in the sedimentary record?

In our sediment core, the MP size, measured as the particle's major dimension, presented a non-normal distribution (Shapiro-Wilk normality test, W = 0.52413, $p < 0.001$), ranging from 13 to 983 μm. Fig 5. 4. A. displays the MP size along with the sediment core. Significant differences on the MP size were found between the sections investigated (Kruskal-Wallis, chi-squared = 76.54, df = 7, $p < 0.001$). The subsequent post hoc Wilcoxon-Mann-Whitney test revealed that the MP size in section 2-3 cm differs from the rest of the core (all $p < 0.001$), and section 6-7 cm differs from the top part of the core (section 0-1 cm to 5-6 cm, all $p< 0.05$). Overall, the variability in MP size with no significant differences between the oldest and most recent MPs sequestered in the sediment column suggests that MPs have not been subjected to physical degradation, which may indicate MP fragmentation after burial. Besides, the general decrease in MP occurrence with sediment depth and constant sediment accumulation in steady-state conditions showed by the ^{210}Pb profile rules out vertical remobilization of the MPs along the investigated sediment core. These observations support the MPs' feasibility as a long-lasting chronostratigraphic marker, as previously suggested in the literature (Bancone et al., 2020; Ivar do Sul and Labrenz, 2021). Limitations should be considered. The presence of MPs in sediment records can be used to corroborate geochronologies when the archive shows an undisturbed nature. Besides, the reliable characterization of MPs requires targeted analytical methods and prevention of cross-contamination that otherwise might mislead the interpretation of the historical records.

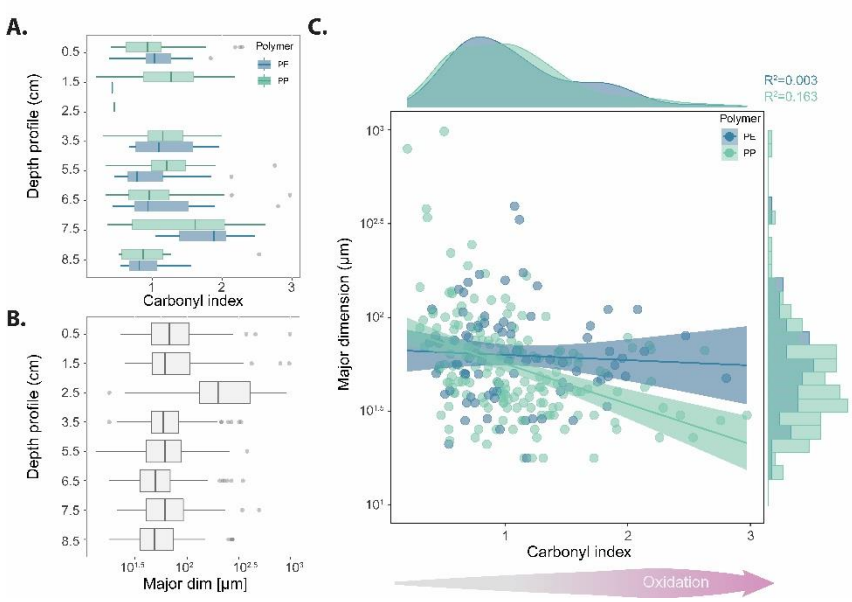


Fig. 5. 4. Summary of the MP weathering status and size variability in the sedimentary record A. Boxplot of the carbonyl index in the sediment profile classified as polyethylene (dark blue) and polypropylene (light blue). B. Boxplot of the size variability in the sediment core. C. Increase of the oxidation status of the polyolefins microplastics, measured as carbonyl index, as the particle size decreases.

4.Implications

In considering the limitations of this study, we acknowledge that coastal benthic environments are complex and highly dynamic, with exposure to disturbing natural and anthropogenic events that can alter the MP accumulation in these systems. The main objectives of this study were to investigate the sequestration and long-term fate of small MPs (11-1000 µm) buried in marine sediments. To comply with these research questions, it was indispensable to select an undisturbed sediment core with a high sedimentation rate from a relatively high MP polluted area to provide high-resolution data on the fate of buried MPs at a sub-decadal scale. The application of the state-of-the-art FPA-µFTIR-Imaging method for the MP characterization and CI computation, described in this study, provided robust results that indicated that i) the MP mass sequestered in marine sediments increased exponentially from 1965 to 2019 and, ii) MPs properties do not vary over time, suggesting preservation of these pollutants within the sedimentary records. These findings corroborated the importance of

benthic environments as major reservoir of microplastic pollutants. The rate MP mass is sequestered in undisturbed sedimentary records is the enduring testimony on the inefficient plastic waste management practices of our societies.

CHAPTER 6

Synthesis

6.1. Synthesis

The ubiquitous presence of MP pollutants across all the spheres of our planet indicates the global dimension of this environmental problem. The scientific community has responded in consequence, bringing together diverse research fields and providing a multidisciplinary approach to face the challenges of MP pollution. While significant efforts have been dedicated to characterizing MP pollution in the marine environment, major research questions still need to be addressed to contextualize these pollutants' exposure and hazard levels in our oceans. This knowledge is crucial to ultimately lead to the development of policies targeting the protection of the marine environment. In particular, this thesis directly contributes to the understanding of MP fluxes, dispersion, and fate in the Mediterranean Sea. This was done 1) to provide empirical data on the levels of MP pollution in the basin and 2) to understand the dual role of transitional systems as sources and retention areas of MPs in the marine environment

The extent of MP pollution in the Mediterranean Sea was evaluated by a systematic review presented in Chapter 2 and by gathering primary data on the occurrence of MPs across different Mediterranean beaches (Chapter 3) and the Ebro Delta system in the NW Mediterranean Sea (Chapter 4 and 5). Different analytical methods were presented in this thesis. In chapters 3 and 4, the presence of MPs and anthropogenic fibers were investigated following a common approach relying on density separation and visual characterization. These techniques provided baseline information on MP pollution levels while considering time and economic constraints. Indeed, as per research reported in Chapter 2, these approaches are predominant in the Mediterranean basin. While these methods may suppose limitations to addressing scientific questions reliably, they are efficient and cost-effective approaches that should be considered for potential monitoring protocols that the administrations may implement to conduct large geographical and temporal surveys. In contrast, the cutting-edge methods for extracting and characterizing MP through FPA-μFT-

IR imaging were employed in Chapter 5 to investigate the fate of small MPs (< 1mm) once buried in the marine sediments.

The systematic review of the methods and occurrence of MP pollution in the abiotic compartments of the Mediterranean Sea showed how different techniques are shaping our current knowledge of MP pollution in this basin. A closer comparison of the size distribution (Feret's diameter) of the particles (<1mm) investigated in this thesis showed differences due to the different analytical techniques, and morphological types of the particles (Fig 6.1). Similar sampling and sample preparation protocol were followed in Chapter 3 and 4, and the results indicated that the overall size of the micro-litter in the Ebro Delta beaches (median: 324 µm; Q_1-Q_3, 156-599 µm) were larger than those recovered across the Mediterranean island beaches (median: 268 µm; Q_1-Q_3, 125-498 µm). Similarly, it can be concluded that the MPs transported in the surface water of the Ebro River delta to the Mediterranean Sea were smaller (median: 184 µm; Q_1-Q_3, 94-378 µm) than those accumulating in the Ebro estuary sediments (median: 323 µm; Q_1-Q_3, 191-542 µm). In contrast, the size distribution revealed through FPA-µFTIR imaging for the MPs recovered from the sedimentary archive collected in the Ebro prodelta showed that the median of the particles was 40 µm (Q_1-Q_3, 29-58 µm) in the sedimentary compartment. However, Fig. 6. 1. demonstrates that the visual characterization failed to detect and identify smaller particles, as reported in previous studies (Primpke et al., 2020; Song et al., 2015). Thus, highlighting the employed methodological constraints is fundamental for the accuracy of the study conclusions to prevent misleading results. As concluded in Chapter 2, the MP research can address the current limitations hindering the overall interpretation of MP pollution by providing open, quality controlled, and comparable data to tackle the challenge of MP pollution.

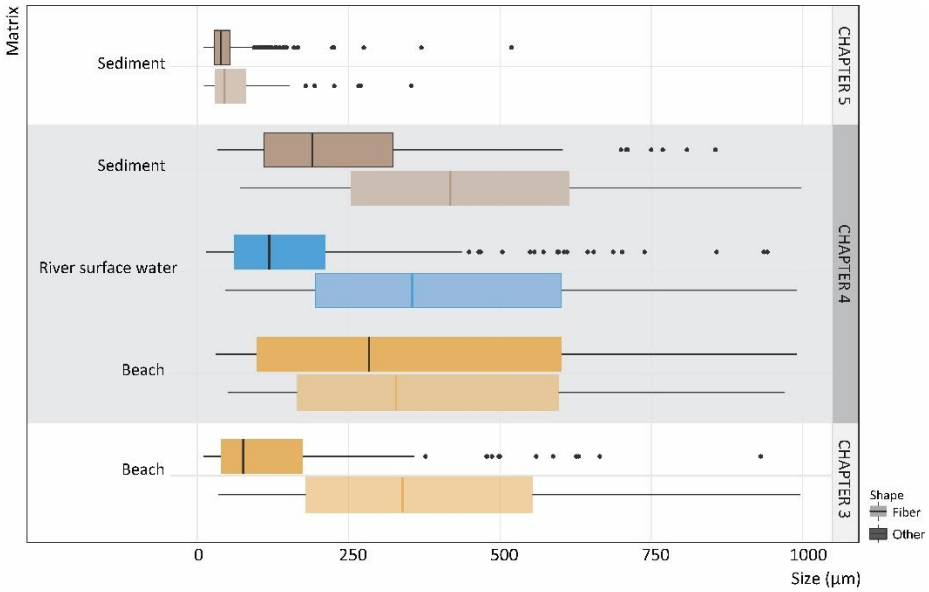


Fig. 6. 1. Boxplot showing the size distribution (Feret's diameter) of micro-debris, MPs (fragment, film, foam), and anthropogenic fibers, focusing on the size fraction <1mm. The y-axis indicates the sampling matrix, and the x-axis the size range (0-1 mm). The colors indicate the environmental matrix: yellow for sandy beaches, blue for surface waters, and brown for benthic sediments.

Besides the relevance of the methods, the findings reported in this thesis shed light on the dynamics of MPs in transitional environments of crucial importance for the Mediterranean Sea. The beaches of this basin attract millions of visitors every year, promoting the development of local and regional economies. Nevertheless, tourism benefits come with environmental and social costs. In Chapter 3, the clear seasonal distribution of the micro-litter in the Mediterranean beaches, following the touristic fluxes, showed that the recreational use of these transitional environments is the main driver controlling the accumulation of these pollutants. The predominance of fibers revealed the intrinsic and unavoidable relation between anthropogenic presence and micro-litter occurrence. Remarkably, the results showed a seasonal pattern of micro-litter abundance in the Mediterranean beaches suggesting clearance or removal mechanisms from the beaches to the Mediterranean coastal waters. Finally, this study showed the challenges and recommendations for future large-scale monitoring surveys.

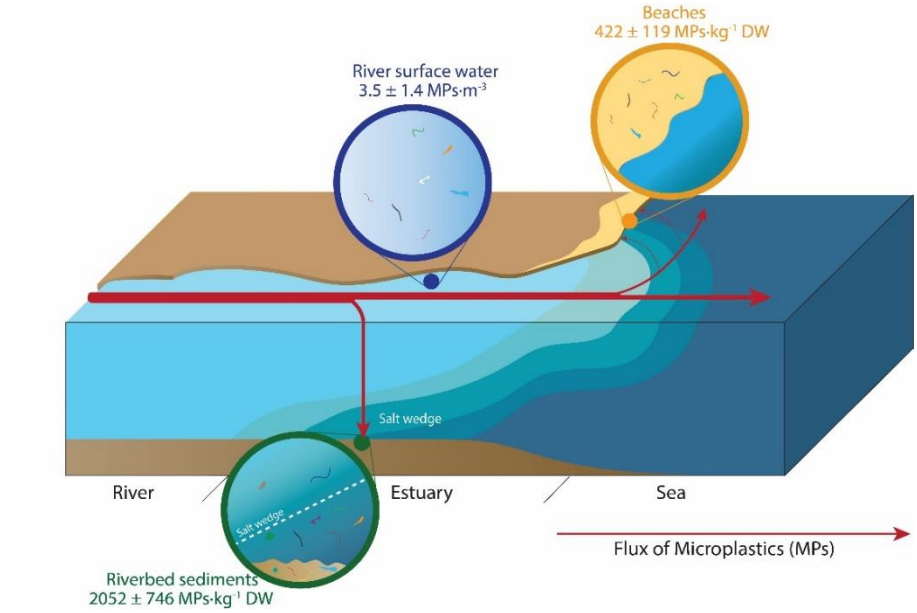


Fig. 6. 2. Graphical abstract summarizing the main findings of Chapter 4 on the distribution of microplastics in the Ebro Delta system**.**

The Ebro Delta system is one of the most extensive wetlands in the Mediterranean Sea, the ecological richness and vulnerability of the area led to declare the area a Natural Park, Special bird protection area, and Wetland of International Importance. Nevertheless, the presence of MP was detected across different environmental compartments of the Delta system (river surface water, beaches, and benthic sediments, Fig 6.2). The relatively higher MPs concentration found settling in the riverbed sedimentary compartment compared to the beaches indicated the potential of estuarine sediment as a relevant accumulation zone for these pollutants, preventing their entrance to the marine environment. However, the occurrence of MPs in the river surface water showed the continuous contribution of the riverine discharge as a source of MPs to the Mediterranean Sea. This MPs influx to the open sea is partly attenuated, remaining trapped in coastal areas due to the currents and tidal dynamics, which ultimately may drive the deposition of these pollutants in littoral areas. At the same time, part of these MPs' inputs is likely effectively dispersed and exported to the open sea, where their fate remains to be explored.

The seafloor is considered the major MP reservoir being the final sink of these pollutants (Martin et al., 2022; Woodall et al., 2014). This potential fate scenario was explored by combining palaeoceanographic approaches with the state-of-the-art methods of MP research. Here, the MP (mass) accumulation rate was found to mimic the exponential growth of global plastic production. Once buried in the sediments, the results suggested an absence of degradation of the MPs leading to their preservation (Fig 6.3).

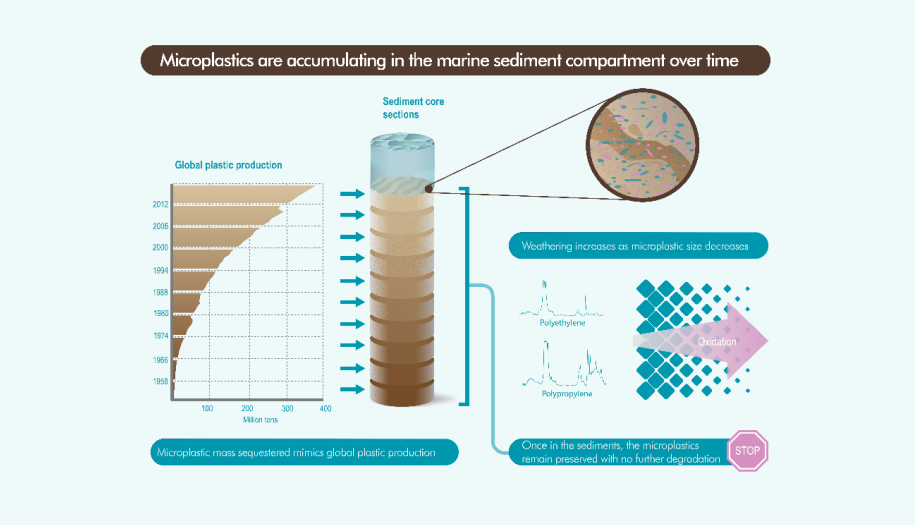


Fig. 6. 3. Graphical abstract summarizing the main conclusions of Chapter 5, the fate of microplastics in undisturbed sediment records.

The global implication of these findings emphasizes the role of river delta systems as a source of MPs and as important accumulation areas for MPs. These systems reflect the downstream influence of the anthropogenic pressure along the watershed, besides, concentrating high population level due to the deltas' high productivity and ecological richness. Under the current 'business as usual' scenario, where plastic materials remain indispensable to our daily lives and plastic waste management remains to be improved, the plastic pollution levels in the marine environment will likely increase. Therefore, transitional systems (estuaries and coastal zones) represent crucial areas to monitor the influx of

MPs to the open sea. This information is critical to model the risk of exposure and hazard levels in the open ocean.

The Mediterranean coastal areas are hotspots for biodiversity (Coll et al., 2012), being also home to several relevant commercial species. Considering that the MP pollution level in this semi-enclosed sea is already representing an ecotoxicological risk (Everaert et al., 2020), there is an urgent need to implement effective policies and regulations to reduce single-use plastics and mitigate plastic waste mismanagement to protect and preserve the unique ecosystems and biodiversity of the Mediterranean Sea.

6.2. Future perspectives

Temporal scale of microplastics pollution

There is daunting evidence of the pervasive presence of MPs in the marine environment. The scientific community has already provided numerous resources (protocols for sampling, sample processing, and identification) to implement effective monitoring efforts that are essential to evaluate the trends in exposure risk levels in the marine environment. Still, one of the major challenges ahead is to understand the evolution and fate of MPs after reaching the marine system. This task can be addressed by providing long time series on their occurrence to fill knowledge gaps on their dynamics. Moreover, these data can be contrasted against already available data series on several oceanographic variables (e.g., primary production, COPEPOD-global plankton database) to evaluate their role in the MP dispersion.

From a snapshot to a dynamic system – targeting the water column

Only few studies have focused on the three-dimensional ocean dispersion of MPs (horizontal and vertical displacement) with most of the sampling targeting the sea surface and the seafloor, despite the largest portion of the ocean being comprised between the surface and the sea sediments. Unavoidable, the occurrence of MPs on the sea surface represents a snapshot of the levels of these pollutants under the specific sampling conditions, whereas the concentration in

the sediments remains as the record of the final sink. Hence, we need to gain an understanding of the processes linking both compartments. Sediment traps can be employed to estimate seasonal and annual MP export rate, besides filtering large seawater volumes at specific depths will contribute to estimating MP standing stocks. In combination with robust analytical methods providing information on the polymer composition and size distribution of the particles, these sampling strategies can shed light on the sinking, residence time, and mechanisms governing the transport of MPs from the sea surface to the bottom of the oceans.

Accumulation and resuspension of microplastics

This thesis proved that MPs accumulate in the sediment compartment over time using an undisturbed high-resolution sediment core. The alternative of investigating MP occurrence in bioturbated and/or mixed sediment archives would contribute to quantify the MP bioavailability and their residence time in benthic biota, and the resuspension of these pollutants. Similarly, these data would be crucial to the ongoing discussion of using MPs as independent tracers to (co)validate chronologies of sediment archives.

Smaller particles, still big challenges

Complementary sampling strategies and analytical methods need to be considered to investigate the fate of plastics, from macro- to nano-scale. (Micro)plastics are fragmenting in the marine environment shredding smaller MPs and nanoplastics. Alternative approaches (i.e., bulk samples, pumps) to the conventional seawater sampling methods (neuston, and manta net) are required to target these particles. Equally, alternative analytical techniques (i.e., Atomic Force Microscopy, Field-Flow-Fractionation, Thermal Desorption - Proton Transfer Reaction - Mass Spectrometry) to vibrational spectroscopy (FTIR and Raman) are needed to overcome the detection limit of nanoplastics. Since the first observations in the early 70s, the MP research field has since then reached a certain maturity. As a next step, the scientific community is now developing these techniques to address the major challenge of sampling and

identifying environmental nanoplastics (Materić et al., 2022, 2020; Ter Halle et al., 2017; Weckhuysen et al., 2021).

References

Abel, S.M., Primpke, S., Int-Veen, I., Brandt, A., Gerdts, G., 2021. Systematic identification of microplastics in abyssal and hadal sediments of the Kuril Kamchatka trench. Environ. Pollut. 269, 116095. https://doi.org/10.1016/j.envpol.2020.116095

Acha, E.M., Mianzan, H.W., Iribarne, O., Gagliardini, D.A., Lasta, C., Daleo, P., 2003. The role of the Río de la Plata bottom salinity front in accumulating debris. Mar. Pollut. Bull. 46, 197–202. https://doi.org/10.1016/S0025-326X(02)00356-9

Adobe Inc., 2021. Adobe Illustrator, https://adobe.com/products/illustrator

Aliani, S., Griffa, A., Molcard, A., 2003. Floating debris in the Ligurian Sea, north-western Mediterranean. Mar. Pollut. Bull. 46, 1142–1149. https://doi.org/10.1016/S0025-326X(03)00192-9

Allen, K., 2009. Inter-species variation in colour perception. Philos. Stud. 142, 197–220. https://doi.org/10.1007/s11098-007-9183-z

Allen, S., Allen, D., Moss, K., Le Roux, G., Phoenix, V.R., Sonke, J.E., 2020. Examination of the ocean as a source for atmospheric microplastics. PLoS One 15, e0232746. https://doi.org/10.1371/journal.pone.0232746

Allen, S., Allen, D., Phoenix, V.R., Le Roux, G., Durántez Jiménez, P., Simonneau, A., Binet, S., Galop, D., 2019. Atmospheric transport and deposition of microplastics in a remote mountain catchment. Nat. Geosci. 2019 125 12, 339–344. https://doi.org/10.1038/s41561-019-0335-5

Almond, J., Sugumaar, P., Wenzel, M.N., Hill, G., Wallis, C., 2020. Determination of the carbonyl index of polyethylene and polypropylene using specified area under band methodology with ATR-FTIR spectroscopy. E-Polymers 20, 369–381. https://doi.org/10.1515/EPOLY-2020-0041/MACHINEREADABLECITATION/RIS

Alshawafi, A., Analla, M., Alwashali, E., Aksissou, M., 2017. Assessment of marine debris on the coastal wetland of Martil in the North-East of Morocco. Mar. Pollut. Bull. 117, 302–310. https://doi.org/10.1016/j.marpolbul.2017.01.079

Anbumani, S., Kakkar, P., 2018. Ecotoxicological effects of microplastics on biota: a review. Environ. Sci. Pollut. Res. 2018 2515 25, 14373–14396. https://doi.org/10.1007/S11356-018-1999-X

Anderson, Z.T., Cundy, A.B., Croudace, I.W., Warwick, P.E., Celis-Hernandez, O., Stead, J.L., 2018. A rapid method for assessing the accumulation of microplastics in the sea surface microlayer (SML) of

estuarine systems. Sci. Rep. 8, 9428. https://doi.org/10.1038/s41598-018-27612-w

Andrady, A.L., 2011. Microplastics in the marine environment. Mar. Pollut. Bull. 62, 1596–1605. https://doi.org/10.1016/j.marpolbul.2011.05.030

Anthony Browne, M., Crump, P., Niven, S.J., Teuten, E., Tonkin, A., Galloway, T., Thompson, R., 2011. Accumulation of Microplastic on Shorelines Woldwide: Sources and Sinks. Environ. Sci. Technol 45, 9175–9179. https://doi.org/10.1021/es201811s

Arias-Ortiz, A., Masqué, P., Garcia-Orellana, J., Serrano, O., Mazarrasa, I., Marbá, N., Lovelock, C.E., Lavery, P.S., Duarte, C.M., 2018. Reviews and syntheses: 210Pb-derived sediment and carbon accumulation rates in vegetated coastal ecosystems - Setting the record straight. Biogeosciences 15, 6791–6818. https://doi.org/10.5194/BG-15-6791-2018

Arthur, C., Baker, J., Bamford, H., 2009. Proceedings of the International Research Workshop on the Occurrence, Effects and Fate of Microplastic Marine Debris. NOAA Tech. Memo. NOS-OR&R-30 530. https://doi.org/10.1186/1744-9081-10-32

Atwood, E.C., Falcieri, F.M., Piehl, S., Bochow, M., Matthies, M., Franke, J., Carniel, S., Sclavo, M., Laforsch, C., Siegert, F., 2019. Coastal accumulation of microplastic particles emitted from the Po River, Northern Italy: Comparing remote sensing and hydrodynamic modelling with in situ sample collections. Mar. Pollut. Bull. 138, 561–574. https://doi.org/10.1016/j.marpolbul.2018.11.045

Avio, C.G., Gorbi, S., Regoli, F., 2017. Plastics and microplastics in the oceans: From emerging pollutants to emerged threat. Mar. Environ. Res. 128, 2–11. https://doi.org/10.1016/j.marenvres.2016.05.012

Baini, M., Fossi, M.C., Galli, M., Caliani, I., Campani, T., Finoia, M.G., Panti, C., 2018. Abundance and characterization of microplastics in the coastal waters of Tuscany (Italy): The application of the MSFD monitoring protocol in the Mediterranean Sea. Mar. Pollut. Bull. 133, 543–552. https://doi.org/10.1016/j.marpolbul.2018.06.016

Bancone, C.E.P., Turner, S.D., Ivar do Sul, J.A., Rose, N.L., 2020. The Paleoecology of Microplastic Contamination. Front. Environ. Sci. https://doi.org/10.3389/fenvs.2020.574008

Barnes, D.K.A., Galgani, F., Thompson, R.C., Barlaz, M., 2009. Accumulation and fragmentation of plastic debris in global environments. Philos. Trans. R. Soc. B Biol. Sci. 364, 1985–1998. https://doi.org/10.1098/rstb.2008.0205

Barrows, A.P.W., Neumann, C.A., Berger, M.L., Shaw, S.D., 2017. Grab: Vs. neuston tow net: A microplastic sampling performance comparison and

possible advances in the field. Anal. Methods 9, 1446–1453. https://doi.org/10.1039/c6ay02387h

Baztan, J., Carrasco, A., Chouinard, O., Cleaud, M., Gabaldon, J.E., Huck, T., Jaffrès, L., Jorgensen, B., Miguelez, A., Paillard, C., Vanderlinden, J.P., 2014. Protected areas in the Atlantic facing the hazards of micro-plastic pollution: First diagnosis of three islands in the Canary Current. Mar. Pollut. Bull. 80, 302–311. https://doi.org/10.1016/J.MARPOLBUL.2013.12.052

Bergmann, M., Wirzberger, V., Krumpen, T., Lorenz, C., Primpke, S., Tekman, M.B., Gerdts, G., 2017. High Quantities of Microplastic in Arctic Deep-Sea Sediments from the HAUSGARTEN Observatory. Environ. Sci. Technol. 51, 11000–11010. https://doi.org/10.1021/acs.est.7b03331

Besley, A., Vijver, M.G., Behrens, P., Bosker, T., 2017. A standardized method for sampling and extraction methods for quantifying microplastics in beach sand. Mar. Pollut. Bull. 114, 77–83. https://doi.org/10.1016/J.MARPOLBUL.2016.08.055

Besseling, E., Quik, J.T.K., Sun, M., Koelmans, A.A., 2017. Fate of nano- and microplastic in freshwater systems: A modeling study. Environ. Pollut. 220, 540–548. https://doi.org/10.1016/j.envpol.2016.10.001

Béthoux, J.P., 1979. Budgets of the Mediterranean Sea-Their dependance on the local climate and on the characteristics of the Atlantic waters. Oceanol. acta 2, 157–163.

Brach, L., Deixonne, P., Bernard, M.F., Durand, E., Desjean, M.C., Perez, E., van Sebille, E., ter Halle, A., 2018. Anticyclonic eddies increase accumulation of microplastic in the North Atlantic subtropical gyre. Mar. Pollut. Bull. 126, 191–196. https://doi.org/10.1016/j.marpolbul.2017.10.077

Brandon, J., Goldstein, M., Ohman, M.D., 2016. Long-term aging and degradation of microplastic particles: Comparing in situ oceanic and experimental weathering patterns. https://doi.org/10.1016/j.marpolbul.2016.06.048

Brandon, J.A., Jones, W., Ohman, M.D., 2019. Multidecadal increase in plastic particles in coastal ocean sediments. Sci. Adv 5.

Brennan, E., Wilcox, C., Hardesty, B.D., 2018. Connecting flux, deposition and resuspension in coastal debris surveys. Sci. Total Environ. 644, 1019–1026. https://doi.org/10.1016/j.scitotenv.2018.06.352

Brennholt, N., Heß, M., Reifferscheid, G., 2018. Freshwater microplastics: Challenges for regulation and management. Handb. Environ. Chem. 58, 239–272. https://doi.org/10.1007/978-3-319-61615-5_12

Browne, M.A., Crump, P., Niven, S.J., Teuten, E., Tonkin, A., Galloway, T., Thompson, R., 2011. Accumulation of Microplastic on Shorelines Woldwide: Sources and Sinks. Environ. Sci. Technol. 45, 9175–9179. https://doi.org/10.1021/es201811s

Browne, M.A., Galloway, T., Thompson, R., Velzeboer, I., Kwadijk, C.J.A.F., Koelmans, A.A., Using, F., Etoh, C.H.O., Rietzler, B., Bechtold, T., Schneider, N.S., Dusablon, L. V, Spano, L.A., Hopfenberg, H.B., Votta, F., 2014. Forscher finden erstmals Mikroplastik in Stuhlproben von Menschen. Environ. Sci. Technol. 48, 4869–4876. https://doi.org/10.1002/ieam.5630030412

Buchanan, J.B., 1971. Pollution by synthetic fibres. Mar. Pollut. Bull. 2, 23. https://doi.org/10.1016/0025-326X(71)90136-6

Brunson, J.C., 2020. "ggalluvial: Alluvial Plots in 'ggplot2'." R package version 0.12.3, http://corybrunson.github.io/ggalluvial/

Carpenter, E.J., Smith, K.L., 1972. Plastics on the Sargasso sea surface. Science (80-.). 175, 1240–1241. https://doi.org/10.1126/science.175.4027.1240

Carreras-Colom, E., Constenla, M., Soler-Membrives, A., Cartes, J.E., Baeza, M., Carrass On, M., 2020. A closer look at anthropogenic fiber ingestion in Aristeus antennatus in the NW Mediterranean Sea: Differences among years and locations and impact on health condition *. https://doi.org/10.1016/j.envpol.2020.114567

Castañeda, R.A., Avlijas, S., Simard, M.A., Ricciardi, A., Smith, R., 2014. Microplastic pollution in St. Lawrence River sediments. Can. J. Fish. Aquat. Sci. 71, 1767–1771. https://doi.org/10.1139/cjfas-2014-0281

Cau, A., Avio, C.G., Dessì, C., Follesa, M.C., Moccia, D., Regoli, F., Pusceddu, A., 2019. Microplastics in the crustaceans Nephrops norvegicus and Aristeus antennatus: Flagship species for deep-sea environments? Environ. Pollut. 255, 113107. https://doi.org/10.1016/j.envpol.2019.113107

Ceccarini, A., Corti, A., Erba, F., Modugno, F., Nasa, L., Bianchi, S., Castelvetro, V., 2018. The Hidden Microplastics: New Insights and Figures from the Thorough Separation and Characterization of Microplastics and of Their Degradation Byproducts in Coastal Sediments. https://doi.org/10.1021/acs.est.8b01487

Chouchene, K., da Costa, J.P., Wali, A., Girão, A. V., Hentati, O., Duarte, A.C., Rocha-Santos, T., Ksibi, M., 2019. Microplastic pollution in the sediments of Sidi Mansour Harbor in Southeast Tunisia. Mar. Pollut. Bull. 146, 92–99. https://doi.org/10.1016/j.marpolbul.2019.06.004

Choy, C.A., Robison, B.H., Gagne, T.O., Erwin, B., Firl, E., Halden, R.U., Hamilton, J.A., Katija, K., Lisin, S.E., Rolsky, C., S. Van Houtan, K.,

2019. The vertical distribution and biological transport of marine microplastics across the epipelagic and mesopelagic water column. Sci. Rep. 9, 1–9. https://doi.org/10.1038/s41598-019-44117-2

Cincinelli, A., Martellini, T., Guerranti, C., Scopetani, C., Chelazzi, D., Giarrizzo, T., 2019. A potpourri of microplastics in the sea surface and water column of the Mediterranean Sea. TrAC - Trends Anal. Chem. https://doi.org/10.1016/j.trac.2018.10.026

Cincinelli, A., Scopetani, C., Chelazzi, D., Lombardini, E., Martellini, T., Katsoyiannis, A., Fossi, M.C., Corsolini, S., 2017. Microplastic in the surface waters of the Ross Sea (Antarctica): Occurrence, distribution and characterization by FTIR. Chemosphere 175, 391–400. https://doi.org/10.1016/J.CHEMOSPHERE.2017.02.024

Claessens, M., Meester, S. De, Landuyt, L. Van, Clerck, K. De, Janssen, C.R., 2011. Occurrence and distribution of microplastics in marine sediments along the Belgian coast. Mar. Pollut. Bull. 62, 2199–2204. https://doi.org/10.1016/J.MARPOLBUL.2011.06.030

Cole, M., Lindeque, P., Halsband, C., Galloway, T.S., 2011. Microplastics as contaminants in the marine environment: A review. Mar. Pollut. Bull. 62, 2588–2597. https://doi.org/10.1016/j.marpolbul.2011.09.025

Coll, M., Piroddi, C., Albouy, C., Ben Rais Lasram, F., Cheung, W.W.L., Christensen, V., Karpouzi, V.S., Guilhaumon, F., Mouillot, D., Paleczny, M., Palomares, M.L., Steenbeek, J., Trujillo, P., Watson, R., Pauly, D., 2012. The Mediterranean Sea under siege: Spatial overlap between marine biodiversity, cumulative threats and marine reserves. Glob. Ecol. Biogeogr. 21, 465–480. https://doi.org/10.1111/j.1466-8238.2011.00697.x

Coll, M., Piroddi, C., Steenbeek, J., Kaschner, K., Ben Rais Lasram, F., Aguzzi, J., Ballesteros, E., Bianchi, C.N., Corbera, J., Dailianis, T., Danovaro, R., Estrada, M., Froglia, C., Galil, B.S., Gasol, J.M., Gertwagen, R., Gil, J., Guilhaumon, F., Kesner-Reyes, K., Kitsos, M.-S., Koukouras, A., Lampadariou, N., Laxamana, E., López-Fé de la Cuadra, C.M., Lotze, H.K., Martin, D., Mouillot, D., Oro, D., Raicevich, S., Rius-Barile, J., Saiz-Salinas, J.I., San Vicente, C., Somot, S., Templado, J., Turon, X., Vafidis, D., Villanueva, R., Voultsiadou, E., 2010. The Biodiversity of the Mediterranean Sea: Estimates, Patterns, and Threats. PLoS One 5, e11842. https://doi.org/10.1371/journal.pone.0011842

Collignon, A., Hecq, J.H., Galgani, F., Collard, F., Goffart, A., 2014. Annual variation in neustonic micro- and meso-plastic particles and zooplankton in the Bay of Calvi (Mediterranean-Corsica). Mar. Pollut. Bull. 79, 293–298. https://doi.org/10.1016/j.marpolbul.2013.11.023

Collignon, A., Hecq, J.H., Glagani, F., Voisin, P., Collard, F., Goffart, A., 2012.

Neustonic microplastic and zooplankton in the North Western Mediterranean Sea. Mar. Pollut. Bull. 64, 861–864. https://doi.org/10.1016/j.marpolbul.2012.01.011

Compa, M., Alomar, C., Mourre, B., March, D., Tintoré, J., Deudero, S., 2020. Nearshore spatio-temporal sea surface trawls of plastic debris in the Balearic Islands. Mar. Environ. Res. 158. https://doi.org/10.1016/j.marenvres.2020.104945

Courtene-Jones, W., Quinn, B., Ewins, C., Gary, S.F., Narayanaswamy, B.E., 2020. Microplastic accumulation in deep-sea sediments from the Rockall Trough. Mar. Pollut. Bull. 154, 111092. https://doi.org/10.1016/j.marpolbul.2020.111092

Covernton, G.A., Pearce, C.M., Gurney-Smith, H.J., Chastain, S.G., Ross, P.S., Dower, J.F., Dudas, S.E., 2019. Size and shape matter: A preliminary analysis of microplastic sampling technique in seawater studies with implications for ecological risk assessment. Sci. Total Environ. 667, 124–132. https://doi.org/10.1016/j.scitotenv.2019.02.346

Cozar, A., Echevarria, F., Gonzalez-Gordillo, J.I., Irigoien, X., Ubeda, B., Hernandez-Leon, S., Palma, A.T., Navarro, S., Garcia-de-Lomas, J., Ruiz, A., Fernandez-de-Puelles, M.L., Duarte, C.M., 2014. Plastic debris in the open ocean. Proc. Natl. Acad. Sci. 111, 10239–10244. https://doi.org/10.1073/pnas.1314705111

Cózar, A., Sanz-Martín, M., Martí, E., González-Gordillo, J.I., Ubeda, B., Gálvez, J.Á., Irigoien, X., Duarte, C.M., 2015. Plastic Accumulation in the Mediterranean Sea. PLoS One 10, e0121762. https://doi.org/10.1371/journal.pone.0121762

Crawford, C.B., Quinn, B., 2017a. Microplastic pollutants. Elsevier.

Crawford, C.B., Quinn, B., 2017b. Plastic production, waste and legislation, in: Microplastic Pollutants. Elsevier, pp. 39–56. https://doi.org/10.1016/B978-0-12-809406-8.00003-7

Crutzen, P.J., 2002. Geology of mankind. Nat. 2002 4156867 415, 23–23. https://doi.org/10.1038/415023a

D'Alessandro, M., Esposito, V., Porporato, E.M.D., Berto, D., Renzi, M., Giacobbe, S., Scotti, G., Consoli, P., Valastro, G., Andaloro, F., Romeo, T., 2018. Relationships between plastic litter and chemical pollutants on benthic biodiversity *. https://doi.org/10.1016/j.envpol.2018.08.002

Dahl, M., Bergman, S., Björk, M., Diaz-Almela, E., Granberg, M., Gullström, M., Leiva-Dueñas, C., Magnusson, K., Marco-Méndez, C., Piñeiro-Juncal, N., Mateo, M.Á., 2021. A temporal record of microplastic pollution in Mediterranean seagrass soils. Environ. Pollut. 273, 116451. https://doi.org/10.1016/J.ENVPOL.2021.116451

Danovaro, R., Company, J.B., Corinaldesi, C., D'Onghia, G., Galil, B., Gambi, C., Gooday, A.J., Lampadariou, N., Luna, G.M., Morigi, C., Olu, K., Polymenakou, P., Ramirez-Llodra, E., Sabbatini, A., Sardá, F., Sibuet, M., Tselepides, A., 2010. Deep-sea biodiversity in the Mediterranean Sea: The known, the unknown, and the unknowable. PLoS One 5, e11832. https://doi.org/10.1371/journal.pone.0011832

De Falco, F., Cocca, M., Avella, M., Thompson, R.C., 2020. Microfiber Release to Water, Via Laundering, and to Air, via Everyday Use: A Comparison between Polyester Clothing with Differing Textile Parameters. Cite This Environ. Sci. Technol 54, 3296. https://doi.org/10.1021/acs.est.9b06892

De Falco, F., Di Pace, E., Cocca, M., Avella, M., 2019. The contribution of washing processes of synthetic clothes to microplastic pollution. Sci. Reports 2019 91 9, 1–11. https://doi.org/10.1038/s41598-019-43023-x

de Haan, W.P., Sanchez-Vidal, A., Canals, M., 2019. Floating microplastics and aggregate formation in the Western Mediterranean Sea. Mar. Pollut. Bull. 140, 523–535. https://doi.org/10.1016/j.marpolbul.2019.01.053

de Lucia, G.A., Vianello, A., Camedda, A., Vani, D., Tomassetti, P., Coppa, S., Palazzo, L., Amici, M., Romanelli, G., Zampetti, G., Cicero, A.M., Carpentieri, S., Di Vito, S., Matiddi, M., 2018. Sea water contamination in the Vicinity of the Italian minor islands caused by microplastic pollution. Water (Switzerland) 10, 1108. https://doi.org/10.3390/w10081108

De Ruijter, V.N., Milou, A., Costa, V., 2019. Assessment of microplastics distribution and stratification in the shallow marine sediments of Samos island, Eastern Mediterranean sea, Greece. Mediterr. Mar. Sci. 20, 736–744. https://doi.org/10.12681/mms.19131

de Sá, L.C., Oliveira, M., Ribeiro, F., Rocha, T.L., Futter, M.N., 2018. Studies of the effects of microplastics on aquatic organisms: What do we know and where should we focus our efforts in the future? Sci. Total Environ. 645, 1029–1039. https://doi.org/10.1016/j.scitotenv.2018.07.207

de Smit, J.C., Anton, A., Martin, C., Rossbach, S., Bouma, T.J., Duarte, C.M., 2021. Habitat-forming species trap microplastics into coastal sediment sinks. Sci. Total Environ. 772, 145520. https://doi.org/10.1016/j.scitotenv.2021.145520

Derraik, J.G.., 2002. The pollution of the marine environment by plastic debris: a review. Mar. Pollut. Bull. 44, 842–852. https://doi.org/10.1016/S0025-326X(02)00220-5

Deudero, S., Alomar, C., 2015. Mediterranean marine biodiversity under threat: Reviewing influence of marine litter on species.

https://doi.org/10.1016/j.marpolbul.2015.07.012

Dick Vethaak, A., Legler, J., 2021. Microplastics and human health: Knowledge gaps should be addressed to ascertain the health risks of microplastics. Science (80-.). https://doi.org/10.1126/science.abe5041

Digka, N., Tsangaris, C., Torre, M., Anastasopoulou, A., Zeri, C., 2018. Microplastics in mussels and fish from the Northern Ionian Sea. Mar. Pollut. Bull. 135, 30–40. https://doi.org/10.1016/j.marpolbul.2018.06.063

Dris, R., Gasperi, J., Rocher, V., Saad, M., Renault, N., Tassin, B., 2015. Microplastic contamination in an urban area: A case study in Greater Paris. Environ. Chem. 12, 592–599. https://doi.org/10.1071/EN14167

Dris, R., Gasperi, J., Saad, M., Mirande, C., Tassin, B., 2016. Synthetic fibers in atmospheric fallout: A source of microplastics in the environment? Mar. Pollut. Bull. 104, 290–293. https://doi.org/10.1016/j.marpolbul.2016.01.006

Duis, K., Coors, A., 2016. Microplastics in the aquatic and terrestrial environment: sources (with a specific focus on personal care products), fate and effects. Environ. Sci. Eur. 28, 1–25. https://doi.org/10.1186/s12302-015-0069-y

Edmondson, S., Gilbert, M., 2017. The Chemical Nature of Plastics Polymerization. Brydson's Plast. Mater. Eighth Ed. 19–37. https://doi.org/10.1016/B978-0-323-35824-8.00002-5

Eerkes-Medrano, D., Thompson, R.C., Aldridge, D.C., 2015. Microplastics in freshwater systems: A review of the emerging threats, identification of knowledge gaps and prioritisation of research needs. Water Res. 75, 63–82. https://doi.org/10.1016/j.watres.2015.02.012

Egger, M., Nijhof, R., Quiros, L., Leone, G., Royer, S.J., McWhirter, A.C., Kantakov, G.A., Radchenko, V.I., Pakhomov, E.A., Hunt, B.P.V., Lebreton, L., 2020. A spatially variable scarcity of floating microplastics in the eastern North Pacific Ocean. Environ. Res. Lett. 15. https://doi.org/10.1088/1748-9326/ABBB4F

Elhacham, E., Ben-Uri, L., Grozovski, J., Bar-On, Y.M., Milo, R., 2020. Global human-made mass exceeds all living biomass. Nature 588. https://doi.org/10.1038/s41586-020-3010-5

Enders, K., Lenz, R., Beer, S., Stedmon, C.A., 2017. Extraction of microplastic from biota: Recommended acidic digestion destroys common plastic polymers. ICES J. Mar. Sci. 74, 326–331. https://doi.org/10.1093/icesjms/fsw173

EPA, 1992. Plastic Pellets in the Aquatic Environment: Sources and Recommendations. United States Environ. Prot. Agency 56.

https://doi.org/EPA 842-S-93-001

Ericson, J.P., Vörösmarty, C.J., Dingman, S.L., Ward, L.G., Meybeck, M., 2006. Effective sea-level rise and deltas: Causes of change and human dimension implications. Glob. Planet. Change 50, 63–82. https://doi.org/10.1016/j.gloplacha.2005.07.004

Eriksen, M., Lebreton, L.C.M., Carson, H.S., Thiel, M., Moore, C.J., Borerro, J.C., Galgani, F., Ryan, P.G., Reisser, J., 2014. Plastic Pollution in the World's Oceans: More than 5 Trillion Plastic Pieces Weighing over 250,000 Tons Afloat at Sea. PLoS One 9, e111913. https://doi.org/10.1371/journal.pone.0111913

Eriksen, M., Mason, S., Wilson, S., Box, C., Zellers, A., Edwards, W., Farley, H., Amato, S., 2013. Microplastic pollution in the surface waters of the Laurentian Great Lakes. Mar. Pollut. Bull. 77, 177–182. https://doi.org/10.1016/j.marpolbul.2013.10.007

Eriksson, C., Burton, H., Fitch, S., Schulz, M., Van Den Hoff, J., 2013. Daily accumulation rates of marine debris on sub-Antarctic island beaches. Mar. Pollut. Bull. 66, 199–208. https://doi.org/10.1016/j.marpolbul.2012.08.026

Erni-Cassola, G., Zadjelovic, V., Gibson, M.I., Christie-Oleza, J.A., 2019. Distribution of plastic polymer types in the marine environment; A meta-analysis. J. Hazard. Mater. 369, 691–698. https://doi.org/10.1016/j.jhazmat.2019.02.067

European Environment Agency, 2014. Horizon 2020 Mediterranean report. Toward shared environmental information systems.

Everaert, G., De Rijcke, M., Lonneville, B., Janssen, C.R., Backhaus, T., Mees, J., van Sebille, E., Koelmans, A.A., Catarino, A.I., Vandegehuchte, M.B., 2020. Risks of floating microplastic in the global ocean. Environ. Pollut. 267. https://doi.org/10.1016/j.envpol.2020.115499

Fatorić, S., Chelleri, L., 2012. Vulnerability to the effects of climate change and adaptation: The case of the Spanish Ebro Delta. Ocean Coast. Manag. 60, 1–10. https://doi.org/10.1016/j.ocecoaman.2011.12.015

Faure, F., Corbaz, M., Baecher, H., Neuhaus, V., de Alencastro, L., 2013. Pollution due to Plastics and Microplastics in Lake Geneva. 6th Int. Conf. water Resour. Environ. Res.

Fernandez-Arcaya, U., Ramirez-Llodra, E., Aguzzi, J., Allcock, A.L., Davies, J.S., Dissanayake, A., Harris, P., Howell, K., Huvenne, V.A.I., Macmillan-Lawler, M., Martín, J., Menot, L., Nizinski, M., Puig, P., Rowden, A.A., Sanchez, F., Van den Beld, I.M.J., 2017. Ecological role of submarine canyonsand need for canyon conservation: A review. Front. Mar. Sci. https://doi.org/10.3389/fmars.2017.00005

Fossi, M.C., Coppola, D., Baini, M., Giannetti, M., Guerranti, C., Marsili, L., Panti, C., de Sabata, E., Clò, S., 2014. Large filter feeding marine organisms as indicators of microplastic in the pelagic environment: The case studies of the Mediterranean basking shark (Cetorhinus maximus) and fin whale (Balaenoptera physalus). Mar. Environ. Res. 100, 17–24. https://doi.org/10.1016/j.marenvres.2014.02.002

Fossi, M.C., Romeo, T., Baini, M., Panti, C., Marsili, L., Campan, T., Canese, S., Galgani, F., Druon, J.N., Airoldi, S., Taddei, S., Fattorini, M., Brandini, C., Lapucci, C., 2017. Plastic debris occurrence, convergence areas and fin whales feeding ground in the Mediterranean marine protected area Pelagos Sanctuary: A modeling approach. Front. Mar. Sci. 4, 1–15. https://doi.org/10.3389/fmars.2017.00167

Foufoula-Georgiou, E., Syvitski, J., Paola, C., Hoanh, C.T., Tuong, P., Vörösmarty, C., Kremer, H., Brondizio, E., Saito, Y., Twilley, R., 2011. International Year of Deltas 2013: A proposal. Eos, Trans. Am. Geophys. Union 92, 340–341. https://doi.org/10.1029/2011EO400006

Free, C.M., Jensen, O.P., Mason, S.A., Eriksen, M., Williamson, N.J., Boldgiv, B., 2014. High-levels of microplastic pollution in a large, remote, mountain lake. Mar. Pollut. Bull. 85, 156–163. https://doi.org/10.1016/j.marpolbul.2014.06.001

Frias et al., 2018. Standardised protocol for monitoring microplastics in sediments. JPI-oceans BASEMAN Project.

Frias, J. P.G.L., Gago, J., Otero, V., Sobral, P., 2016. Microplastics in coastal sediments from Southern Portuguese shelf waters. Mar. Environ. Res. 114, 24–30. https://doi.org/10.1016/j.marenvres.2015.12.006

Frias, J.P.G.L., Gago, J., Otero, V., Sobral, P., 2016. Microplastics in coastal sediments from Southern Portuguese shelf waters. Mar. Environ. Res. 114, 24–30. https://doi.org/10.1016/J.MARENVRES.2015.12.006

Frias, J.P.G.L., Nash, R., 2018. Microplastics: Finding a consensus on the definition. https://doi.org/10.1016/j.marpolbul.2018.11.022

Gago, J., Galgani, F., Maes, T., Thompson, R.C., 2016. Microplastics in Seawater: Recommendations from the Marine Strategy Framework Directive Implementation Process. Front. Mar. Sci. 3, 219. https://doi.org/10.3389/fmars.2016.00219

Garcia-Garin, O., Vighi, M., Aguilar, A., Tsangaris, C., Digka, N., Kaberi, H., Borrell, A., 2019. Boops boops as a bioindicator of microplastic pollution along the Spanish Catalan coast. Mar. Pollut. Bull. 149, 110648. https://doi.org/10.1016/j.marpolbul.2019.110648

Gatidou, G., Arvaniti, O.S., Stasinakis, A.S., 2019. Review on the occurrence and fate of microplastics in Sewage Treatment Plants. J. Hazard. Mater.

367, 504–512. https://doi.org/10.1016/j.jhazmat.2018.12.081

Genua-Olmedo, A., Alcaraz, C., Caiola, N., Ibáñez, C., 2016. Sea level rise impacts on rice production: The Ebro Delta as an example. Sci. Total Environ. 571, 1200–1210. https://doi.org/10.1016/J.SCITOTENV.2016.07.136

GESAMP, 2015. Sources, fate and effects of microplastics in the marine environment: a global assessment. Reports Stud. 90, 96. https://doi.org/10.13140/RG.2.1.3803.7925

Geyer, R., Jambeck, J.R., Law, K.L., 2017. Production, use, and fate of all plastics ever made. Sci. Adv. 3. https://doi.org/10.1126/SCIADV.1700782/SUPPL_FILE/1700782_SM.PDF

Gobierno de España, 2015. PLAN HIDROLÓGICO de la parte española de la DEMARCACIÓN HIDROGRÁFICA DEL EBRO 2015-2021 MEMORIA.

Gössling, S., 2002. Global environmental consequences of tourism. Glob. Environ. Chang. 12, 283–302. https://doi.org/10.1016/S0959-3780(02)00044-4

Goßmann, I., Halbach, M., Scholz-Böttcher, B.M., 2021. Car and truck tire wear particles in complex environmental samples – A quantitative comparison with "traditional" microplastic polymer mass loads. Sci. Total Environ. 773, 145667. https://doi.org/10.1016/J.SCITOTENV.2021.145667

Gray, A.D., Wertz, H., Leads, R.R., Weinstein, J.E., 2018. Microplastic in two South Carolina Estuaries: Occurrence, distribution, and composition. Mar. Pollut. Bull. 128, 223–233. https://doi.org/10.1016/J.MARPOLBUL.2018.01.030

Green, D.S., Kregting, L., Boots, B., Blockley, D.J., Brickle, P., da Costa, M., Crowley, Q., 2018. A comparison of sampling methods for seawater microplastics and a first report of the microplastic litter in coastal waters of Ascension and Falkland Islands. Mar. Pollut. Bull. 137, 695–701. https://doi.org/10.1016/j.marpolbul.2018.11.004

Grelaud, M., Ziveri, P., 2020. The generation of marine litter in Mediterranean island beaches as an effect of tourism and its mitigation. Sci. Rep. 10, 1–11. https://doi.org/10.1038/s41598-020-77225-5

Guerranti, C., Cannas, S., Scopetani, C., Fastelli, P., Cincinelli, A., Renzi, M., 2017. Plastic litter in aquatic environments of Maremma Regional Park (Tyrrhenian Sea, Italy): Contribution by the Ombrone river and levels in marine sediments. Mar. Pollut. Bull. 117, 366–370. https://doi.org/10.1016/j.marpolbul.2017.02.021

Guerranti, C., Perra, G., Martellini, T., Giari, L., Cincinelli, A., 2020. Knowledge about Microplastic in Mediterranean Tributary River Ecosystems: Lack of Data and Research Needs on Such a Crucial Marine Pollution Source. J. Mar. Sci. Eng. 8, 216. https://doi.org/10.3390/jmse8030216

Gündoğdu, S., 2017. High level of micro-plastic pollution in the Iskenderun Bay NE Levantine coast of Turkey. Ege J. Fish. Aquat. Sci. 34, 401–408. https://doi.org/10.12714/egejfas.2017.34.4.06

Gündoğdu, S., Çevik, C., Ayat, B., Aydoğan, B., Karaca, S., 2018. How microplastics quantities increase with flood events? An example from Mersin Bay NE Levantine coast of Turkey. Environ. Pollut. 239, 342–350. https://doi.org/10.1016/j.envpol.2018.04.042

Güven, O., Gökdağ, K., Jovanović, B., Kıdeyş, A.E., 2017. Microplastic litter composition of the Turkish territorial waters of the Mediterranean Sea, and its occurrence in the gastrointestinal tract of fish. Environ. Pollut. 223, 286–294. https://doi.org/10.1016/j.envpol.2017.01.025

Hamm, T., Lenz, M., 2021. Negative impacts of realistic doses of spherical and irregular microplastics emerged late during a 42 weeks-long exposure experiment with blue mussels. Sci. Total Environ. 778. https://doi.org/10.1016/J.SCITOTENV.2021.146088

Hartmann, N.B., Hu, T., Thompson, R.C., Hassello, M., Verschoor, A., Daugaard, A.E., Rist, S., Karlsson, T., Brennholt, N., Cole, M., Herrling, M.P., Hess, M.C., Ivleva, N.P., Lusher, A.L., Wagner, M., 2019. Are We Speaking the Same Language? Recommendations for a Definition and Categorization Framework for Plastic Debris. https://doi.org/10.1021/acs.est.8b05297

Helm, P.A., 2017. Improving microplastics source apportionment: a role for microplastic morphology and taxonomy? Anal. Methods 9, 1328–1331. https://doi.org/10.1039/C7AY90016C

Hernández-Sánchez, C., González-Sálamo, J., Díaz-Peña, F.J., Fraile-Nuez, E., Hernández-Borges, J., 2021. Arenas Blancas (El Hierro island), a new hotspot of plastic debris in the Canary Islands (Spain). Mar. Pollut. Bull. 169, 112548. https://doi.org/10.1016/J.MARPOLBUL.2021.112548

Hidalgo-Ruz, V., Gutow, L., Thompson, R.C., Thiel, M., 2012. Microplastics in the marine environment: A review of the methods used for identification and quantification. Environ. Sci. Technol. 46, 3060–3075. https://doi.org/10.1021/es2031505

Horton, A.A., Svendsen, C., Williams, R.J., Spurgeon, D.J., Lahive, E., 2017a. Large microplastic particles in sediments of tributaries of the River Thames, UK – Abundance, sources and methods for effective

quantification. Mar. Pollut. Bull. 114, 218–226. https://doi.org/10.1016/j.marpolbul.2016.09.004

Horton, A.A., Walton, A., Spurgeon, D.J., Lahive, E., Svendsen, C., 2017b. Microplastics in freshwater and terrestrial environments: Evaluating the current understanding to identify the knowledge gaps and future research priorities. Sci. Total Environ. 586, 127–141. https://doi.org/10.1016/J.SCITOTENV.2017.01.190

Hurley, R., Woodward, J., Rothwell, J.J., 2018. Microplastic contamination of river beds significantly reduced by catchment-wide flooding. Nat. Geosci. https://doi.org/10.1038/s41561-018-0080-1

Hurley, R.R., Lusher, A.L., Olsen, M., Nizzetto, L., 2018. Validation of a Method for Extracting Microplastics from Complex, Organic-Rich, Environmental Matrices. Environ. Sci. Technol. 52, 7409–7417. https://doi.org/10.1021/acs.est.8b01517

Ibañez, C., Pont, D., Prat, N., 1997. Characterization of the Ebre and Rhone estuaries: A basis for defining and classifying salt-wedge estuaries. Limnol. Oceanogr. 42, 89–101. https://doi.org/10.4319/lo.1997.42.1.0089

Int-Veen, I., Nogueira, P., Isigkeit, J., Hanel, R., Kammann, U., 2021. Positively buoyant but sinking: Polymer identification and composition of marine litter at the seafloor of the North Sea and Baltic Sea. Mar. Pollut. Bull. 172, 112876. https://doi.org/10.1016/J.MARPOLBUL.2021.112876

Ivar do Sul, J.A., Costa, M.F., 2007. Marine debris review for Latin America and the Wider Caribbean Region: From the 1970s until now, and where do we go from here? Mar. Pollut. Bull. 54, 1087–1104. https://doi.org/10.1016/j.marpolbul.2007.05.004

Ivar Do Sul, J.A., Costa, M.F., 2014. The present and future of microplastic pollution in the marine environment. Environ. Pollut. 185, 352–364. https://doi.org/10.1016/j.envpol.2013.10.036

Ivar do Sul, J.A., Labrenz, M., 2021. Microplastics into the Anthropocene. Handb. Microplastics Environ. 1–16. https://doi.org/10.1007/978-3-030-10618-8_25-2

Ivleva, N.P., Wiesheu, A.C., Niessner, R., 2017. Microplastic in Aquatic Ecosystems. Angew. Chemie Int. Ed. 56, 1720–1739. https://doi.org/10.1002/ANIE.201606957

Jambeck, J.R., Geyer, R., Wilcox, C., Siegler, T.R., Perryman, M., Andrady, A., Narayan, R., Law, K.L., 2015. Plastic waste inputs from land into the ocean. Science (80-.). 347, 768–771. https://doi.org/10.1126/science.1260352

Jung, M.R., David Horgen, F., Orski, S. V, Rodriguez, V., Beers, K.L., Balazs, G.H., Jones, T.T., Work, T.M., Brignac, K.C., Royer, S.-J., David Hyrenbach, K., Jensen, B.A., Lynch, J.M., 2018. Validation of ATR FT-IR to identify polymers of plastic marine debris, including those ingested by marine organisms. https://doi.org/10.1016/j.marpolbul.2017.12.061

Kaandorp, M.L.A., Dijkstra, H.A., Van Sebille, E., 2020. Closing the Mediterranean Marine Floating Plastic Mass Budget: Inverse Modeling of Sources and Sinks. Environ. Sci. Technol. 54, 11980–11989. https://doi.org/10.1021/acs.est.0c01984

Kane, I.A., Clare, M.A., 2019. Dispersion, accumulation, and the ultimate fate of microplastics in deep-marine environments: A review and future directions. Front. Earth Sci. https://doi.org/10.3389/feart.2019.00080

Kane, I.A., Clare, M.A., Miramontes, E., Wogelius, R., Rothwell, J.J., Garreau, P., Pohl, F., 2020. Seafloor microplastic hotspots controlled by deep-sea circulation. Science (80-.). eaba5899. https://doi.org/10.1126/science.aba5899

Kazour, M., Jemaa, S., Issa, C., Khalaf, G., Amara, R., 2019. Microplastics pollution along the Lebanese coast (Eastern Mediterranean Basin): Occurrence in surface water, sediments and biota samples. Sci. Total Environ. 696. https://doi.org/10.1016/j.scitotenv.2019.133933

Kim, S., Sin, A., Nam, H., Park, Y., Lee, H., Han, C., 2022. Advanced oxidation processes for microplastics degradation: A recent trend. Chem. Eng. J. Adv. 9, 100213. https://doi.org/10.1016/J.CEJA.2021.100213

Klein, K., Heß, S., Nungeß, S., Schulte-Oehlmann, U., Oehlmann, J., 2021. Particle shape does not affect ingestion and egestion of microplastics by the freshwater shrimp Neocaridina palmata. Environ. Sci. Pollut. Res. 28, 62246–62254. https://doi.org/10.1007/S11356-021-15068-X/FIGURES/2

Koelmans, A.A., Gouin, T., Thompson, R., Wallace, N., Arthur, C., 2014. Plastics in the marine environment. Environ. Toxicol. Chem. 33, 5–10. https://doi.org/10.1002/ETC.2426

Kole, P.J., Löhr, A.J., Van Belleghem, F.G.A.J., Ragas, A.M.J., 2017. Wear and tear of tyres: A stealthy source of microplastics in the environment. Int. J. Environ. Res. Public Health. https://doi.org/10.3390/ijerph14101265

Kooi, M., Reisser, J., Slat, B., Ferrari, F.F., Schmid, M.S., Cunsolo, S., Brambini, R., Noble, K., Sirks, L.A., Linders, T.E.W., Schoeneich-Argent, R.I., Koelmans, A.A., 2016. The effect of particle properties on the depth profile of buoyant plastics in the ocean. Sci. Rep. 6, 1–10. https://doi.org/10.1038/srep33882

Korez, Š., Gutow, L., Saborowski, R., 2019. Microplastics at the strandlines of Slovenian beaches. Mar. Pollut. Bull. 145, 334–342. https://doi.org/10.1016/j.marpolbul.2019.05.054

Krüger, L., Casado-Coy, N., Valle, C., Ramos, M., S Anchez-Jerez, P., Gago, J., Carretero, O., Beltran-Sanahuja, A., Sanz-Lazaro, C., 2019. Plastic debris accumulation in the seabed derived from coastal fish farming *. https://doi.org/10.1016/j.envpol.2019.113336

Kukulka, T., Proskurowski, G., Morét-Ferguson, S., Meyer, D.W., Law, K.L., 2012. The effect of wind mixing on the vertical distribution of buoyant plastic debris. Geophys. Res. Lett. 39, n/a-n/a. https://doi.org/10.1029/2012GL051116

Lagarde, F., Olivier, O., Zanella, M., Daniel, P., Hiard, S., Caruso, A., 2016. Microplastic interactions with freshwater microalgae: Hetero-aggregation and changes in plastic density appear strongly dependent on polymer type. Environ. Pollut. 215, 331–339. https://doi.org/10.1016/j.envpol.2016.05.006

Laglbauer, B.J.L., Franco-Santos, R.M., Andreu-Cazenave, M., Brunelli, L., Papadatou, M., Palatinus, A., Grego, M., Deprez, T., 2014. Macrodebris and microplastics from beaches in Slovenia. Mar. Pollut. Bull. 89, 356–366. https://doi.org/10.1016/J.MARPOLBUL.2014.09.036

Lebreton, L., Andrady, A., 2019. Future scenarios of global plastic waste generation and disposal. Palgrave Commun. 5, 1–11. https://doi.org/10.1057/s41599-018-0212-7

Lebreton, L.C.M., Greer, S.D., Borrero, J.C., 2012. Numerical modelling of floating debris in the world's oceans. Mar. Pollut. Bull. 64, 653–661. https://doi.org/10.1016/j.marpolbul.2011.10.027

Lebreton, L.C.M., Van Der Zwet, J., Damsteeg, J.W., Slat, B., Andrady, A., Reisser, J., 2017a. River plastic emissions to the world's oceans. Nat. Commun. 8, 1–10. https://doi.org/10.1038/ncomms15611

Lefebvre, C., Saraux, C., Heitz, O., Nowaczyk, A., Bonnet, D., 2019. Microplastics FTIR characterisation and distribution in the water column and digestive tracts of small pelagic fish in the Gulf of Lions. Mar. Pollut. Bull. 142, 510–519. https://doi.org/10.1016/j.marpolbul.2019.03.025

Leslie, H.A., Brandsma, S.H., van Velzen, M.J.M., Vethaak, A.D., 2017. Microplastics en route: Field measurements in the Dutch river delta and Amsterdam canals, wastewater treatment plants, North Sea sediments and biota. Environ. Int. 101, 133–142. https://doi.org/10.1016/J.ENVINT.2017.01.018

Li, S., Wang, Y., Liu, L., Lai, H., Zeng, X., Chen, J., Liu, C., Luo, Q., 2021. Temporal and Spatial Distribution of Microplastics in a Coastal Region

of the Pearl River Estuary, China. Water 2021, Vol. 13, Page 1618 13, 1618. https://doi.org/10.3390/W13121618

Li, W.C., Tse, H.F., Fok, L., 2016. Plastic waste in the marine environment: A review of sources, occurrence and effects. Sci. Total Environ. 566–567, 333–349. https://doi.org/10.1016/J.SCITOTENV.2016.05.084

Lima, A.R.A., Barletta, M., Costa, M.F., 2015. Seasonal distribution and interactions between plankton and microplastics in a tropical estuary. Estuar. Coast. Shelf Sci. 165, 213–225. https://doi.org/10.1016/J.ECSS.2015.05.018

Lin, L., Zuo, L.-Z., Peng, J.-P., Cai, L.-Q., Fok, L., Yan, Y., Li, H.-X., Xu, X.-R., 2018. Occurrence and distribution of microplastics in an urban river: A case study in the Pearl River along Guangzhou City, China. Sci. Total Environ. 644, 375–381. https://doi.org/10.1016/J.SCITOTENV.2018.06.327

Lindeque, P.K., Cole, M., Coppock, R.L., Lewis, C.N., Miller, R.Z., Watts, A.J.R., Wilson-McNeal, A., Wright, S.L., Galloway, T.S., 2020. Are we underestimating microplastic abundance in the marine environment? A comparison of microplastic capture with nets of different mesh-size. Environ. Pollut. 265, 114721. https://doi.org/10.1016/j.envpol.2020.114721

Liu, F., Olesen, K.B., Borregaard, A.R., Vollertsen, J., 2019. Microplastics in urban and highway stormwater retention ponds. Sci. Total Environ. 671, 992–1000. https://doi.org/10.1016/j.scitotenv.2019.03.416

Liubartseva, S., Coppini, G., Lecci, R., Clementi, E., 2018. Tracking plastics in the Mediterranean: 2D Lagrangian model. Mar. Pollut. Bull. 129, 151–162. https://doi.org/10.1016/j.marpolbul.2018.02.019

Llorca, M., Álvarez-Muñoz, D., Ábalos, M., Rodríguez-Mozaz, S., Santos, L.H.M.L.M., León, V.M., Campillo, J.A., Martínez-Gómez, C., Abad, E., Farré, M., 2020. Microplastics in Mediterranean coastal area: toxicity and impact for the environment and human health. Trends Environ. Anal. Chem. https://doi.org/10.1016/j.teac.2020.e00090

Löder, M.G.J., Gerdts, G., 2015. Methodology used for the detection and identification of microplastics—a critical appraisal, in: Marine Anthropogenic Litter. Springer International Publishing, Cham, pp. 201–227. https://doi.org/10.1007/978-3-319-16510-3_8

Löder, M.G.J., Imhof, H.K., Ladehoff, M., Löschel, L.A., Lorenz, C., Mintenig, S., Piehl, S., Primpke, S., Schrank, I., Laforsch, C., Gerdts, G., 2017. Enzymatic Purification of Microplastics in Environmental Samples. Environ. Sci. Technol. 51, 14283–14292. https://doi.org/10.1021/acs.est.7b03055

Löder, M.G.J., Kuczera, M., Mintenig, S., Lorenz, C., Gerdts, G., 2015. Focal plane array detector-based micro-Fourier-transform infrared imaging for the analysis of microplastics in environmental samples. Environ. Chem. 12, 563–581. https://doi.org/10.1071/EN14205

Lorenz, C., Roscher, L., Meyer, M.S., Hildebrandt, L., Prume, J., Löder, M.G.J., Primpke, S., Gerdts, G., 2019. Spatial distribution of microplastics in sediments and surface waters of the southern North Sea. Environ. Pollut. 252, 1719–1729. https://doi.org/10.1016/j.envpol.2019.06.093

Lots, F.A.E., Behrens, P., Vijver, M.G., Horton, A.A., Bosker, T., 2017. A large-scale investigation of microplastic contamination: Abundance and characteristics of microplastics in European beach sediment. Mar. Pollut. Bull. 123, 219–226. https://doi.org/10.1016/j.marpolbul.2017.08.057

Lotze, H.K., Coll, M., Dunne, J.A., 2011. Historical Changes in Marine Resources, Food-web Structure and Ecosystem Functioning in the Adriatic Sea, Mediterranean. Ecosystems 14, 198–222. https://doi.org/10.1007/S10021-010-9404-8/FIGURES/8

Lusher, A., Hollman, P., Mendoza-Hill, J., 2017. Microplastics in fisheries and aquaculture. FAO Fish. Aquac. Tech. Pap. eng no. 615.

Ma, J., Zhao, J., Zhu, Z., Li, L., Yu, F., 2019. Effect of microplastic size on the adsorption behavior and mechanism of triclosan on polyvinyl chloride. Environ. Pollut. 254, 113104. https://doi.org/10.1016/j.envpol.2019.113104

Magnusson, K., Norén, F., 2014. Screening of microplastic particles in and down-stream a wastewater treatment plant. IVL Swedish Environ. Res. Inst. C 55, 22. https://doi.org/naturvardsverket-2226

Mansui, J., Molcard, A., Ourmières, Y., 2015. Modelling the transport and accumulation of floating marine debris in the Mediterranean basin. Mar. Pollut. Bull. 91, 249–257. https://doi.org/10.1016/j.marpolbul.2014.11.037

Maršić-Lučić, J., Lušić, J., Tutman, P., Bojanić Varezić, D., Šiljić, J., Pribudić, J., 2018. Levels of trace metals on microplastic particles in beach sediments of the island of Vis, Adriatic Sea, Croatia. Mar. Pollut. Bull. 137, 231–236. https://doi.org/10.1016/j.marpolbul.2018.10.027

Martellini, T., Guerranti, C., Scopetani, C., Ugolini, A., Chelazzi, D., Cincinelli, A., 2018. A snapshot of microplastics in the coastal areas of the Mediterranean Sea. TrAC - Trends Anal. Chem. https://doi.org/10.1016/j.trac.2018.09.028

Martí, E., Martin, C., Galli, M., Echevarría, F., Duarte, C.M., Andrés, A., Cózar, C., 2020. The Colors of the Ocean Plastics. Cite This Environ. Sci.

Technol 54, 6601. https://doi.org/10.1021/acs.est.9b06400

Martin, C., Baalkhuyur, F., Valluzzi, L., Saderne, V., Cusack, M., Almahasheer, H., Krishnakumar, P.K., Rabaoui, L., Qurban, M.A., Arias-Ortiz, A., Masqué, P., Duarte, C.M., 2020. Exponential increase of plastic burial in mangrove sediments as a major plastic sink. Sci. Adv. 6. https://doi.org/10.1126/SCIADV.AAZ5593

Martin, C., Young, C.A., Valluzzi, L., Duarte, C.M., 2022. Ocean sediments as the global sink for marine micro- and mesoplastics. Limnol. Oceanogr. Lett. https://doi.org/10.1002/LOL2.10257

Martin, J., Lusher, A.L., Nixon, F.C., 2021. A review of the use of microplastics in reconstructing dated sedimentary archives. Sci. Total Environ. 150818. https://doi.org/10.1016/J.SCITOTENV.2021.150818

Masura, J., Baker, J., Foster, G., Arthur, C., 2015. Laboratory Methods for the Analysis of Microplastics in the Marine Environment. NOAA Mar. Debris Progr. Natl. 1–39.

Mateos-Cárdenas, A., O'Halloran, J., van Pelt, F.N.A.M., Jansen, M.A.K., 2020. Rapid fragmentation of microplastics by the freshwater amphipod Gammarus duebeni (Lillj.). Sci. Reports 2020 101 10, 1–12. https://doi.org/10.1038/s41598-020-69635-2

Materić, D., Kasper-Giebl, A., Kau, D., Anten, M., Greilinger, M., Ludewig, E., Van Sebille, E., Röckmann, T., Holzinger, R., 2020. Micro-and Nanoplastics in Alpine Snow: A New Method for Chemical Identification and (Semi)Quantification in the Nanogram Range. Environ. Sci. Technol. 54, 2353–2359. https://doi.org/10.1021/ACS.EST.9B07540/SUPPL_FILE/ES9B07540_SI_005.ZIP

Materić, D., Kjær, H.A., Vallelonga, P., Tison, J.L., Röckmann, T., Holzinger, R., 2022. Nanoplastics measurements in Northern and Southern polar ice. Environ. Res. 208, 112741. https://doi.org/10.1016/J.ENVRES.2022.112741

Meijer, L.J.J., van Emmerik, T., van der Ent, R., Schmidt, C., Lebreton, L., 2021. More than 1000 rivers account for 80% of global riverine plastic emissions into the ocean. Sci. Adv. 7. https://doi.org/10.1126/SCIADV.AAZ5803/SUPPL_FILE/AAZ5803_SM.PDF

MERI, 2012. Marine & Environmental Research Institute - Guide to Microplastic Identification (Dissecting Microscope) 14.

Merlino, S., Locritani, M., Bernardi, G., Como, C., Legnaioli, S., Palleschi, V., Abbate, M., 2020. Spatial and Temporal Distribution of Chemically Characterized Microplastics within the Protected Area of Pelagos

Sanctuary (NW Mediterranean Sea): Focus on Natural and Urban Beaches. Water 2020, Vol. 12, Page 3389 12, 3389. https://doi.org/10.3390/W12123389

Misic, C., Covazzi Harriague, A., Ferrari, M., 2019. Hydrodynamic forcing and sand permeability influence the distribution of anthropogenic microparticles in beach sediment. Estuar. Coast. Shelf Sci. 230. https://doi.org/10.1016/j.ecss.2019.106429

Mitrano, D.M., Wick, P., Nowack, B., 2021. Placing nanoplastics in the context of global plastic pollution. Nat. Nanotechnol. https://doi.org/10.1038/s41565-021-00888-2

Moore, C.., Lattin, G.L., Zellers, A.F., 2011. Quantity and type of plastic debris flowing from two urban rivers to costal waters and beaches of Southern California, Journal of Integrated Coastal Zone Management.

Moore, C.J., 2008. Synthetic polymers in the marine environment: A rapidly increasing, long-term threat. Environ. Res. 108, 131–139. https://doi.org/10.1016/j.envres.2008.07.025

Moore, C.J., Moore, S.L., Leecaster, M.K., Weisberg, S.B., 2001. A Comparison of Plastic and Plankton in the North Pacific Central Gyre. Mar. Pollut. Bull. 42, 1297–1300. https://doi.org/10.1016/S0025-326X(01)00114-X

Morales-Caselles, C., Viejo, J., Martí, E., González-Fernández, D., Pragnell-Raasch, H., González-Gordillo, J.I., Montero, E., Arroyo, G.M., Hanke, G., Salvo, V.S., Basurko, O.C., Mallos, N., Lebreton, L., Echevarría, F., van Emmerik, T., Duarte, C.M., Gálvez, J.A., van Sebille, E., Galgani, F., García, C.M., Ross, P.S., Bartual, A., Ioakeimidis, C., Markalain, G., Isobe, A., Cózar, A., 2021. An inshore–offshore sorting system revealed from global classification of ocean litter. Nat. Sustain. 2021 46 4, 484–493. https://doi.org/10.1038/s41893-021-00720-8

Morris, R.J., 1980. Floating plastic debris in the Mediterranean. Mar. Pollut. Bull. 11, 125. https://doi.org/10.1016/0025-326X(80)90073-9

Morritt, D., Stefanoudis, P. V, Pearce, D., Crimmen, O.A., Clark, P.F., 2014. Plastic in the Thames: A river runs through it. https://doi.org/10.1016/j.marpolbul.2013.10.035

MSFD Technical Support group on marine litter, 2013. Guidance on Monitoring Marine Litter. https://doi.org/10.2788/99475

Muller-Karger, F.E., Thunell, R.C., Walsh, J., 2005. The importance of continental margins in the global carbon cycle. https://doi.org/10.1029/2004GL021346

Munno, K., Helm, P.A., Jackson, D.A., Rochman, C., Sims, A., 2018. Impacts

of temperature and selected chemical digestion methods on microplastic particles. Environ. Toxicol. Chem. 37, 91–98. https://doi.org/10.1002/etc.3935

Murphy, F., Ewins, C., Carbonnier, F., Quinn, B., 2016. Wastewater Treatment Works (WwTW) as a Source of Microplastics in the Aquatic Environment. Environ. Sci. Technol. 50, 5800–5808. https://doi.org/10.1021/acs.est.5b05416

Napper, I.E., Thompson, R.C., 2016. Release of synthetic microplastic plastic fibres from domestic washing machines: Effects of fabric type and washing conditions. Mar. Pollut. Bull. 112, 39–45. https://doi.org/10.1016/j.marpolbul.2016.09.025

Nebra, A., Alcaraz, C., Caiola, N., Muñoz-Camarillo, G., Ibáñez, C., 2016. Benthic macrofaunal dynamics and environmental stress across a salt wedge Mediterranean estuary. Mar. Environ. Res. 117, 21–31. https://doi.org/10.1016/j.marenvres.2016.03.009

Nizzetto, L., Futter, M., Langaas, S., 2016. Are Agricultural Soils Dumps for Microplastics of Urban Origin? Environ. Sci. Technol. 50, 10777–10779. https://doi.org/10.1021/acs.est.6b04140

Oehlmann, J., Schulte-Oehlmann, U., Kloas, W., Jagnytsch, O., Lutz, I., Kusk, K.O., Wollenberger, L., Santos, E.M., Paull, G.C., VanLook, K.J.W., Tyler, C.R., 2009. A critical analysis of the biological impacts of plasticizers on wildlife. Philos. Trans. R. Soc. B Biol. Sci. https://doi.org/10.1098/rstb.2008.0242

Onink, V., Jongedijk, C.E., Hoffman, M.J., van Sebille, E., Laufkötter, C., 2021. Global simulations of marine plastic transport show plastic trapping in coastal zones. Environ. Res. Lett. 16, 064053. https://doi.org/10.1088/1748-9326/ABECBD

Pabortsava, K., Lampitt, R.S., 2020. High concentrations of plastic hidden beneath the surface of the Atlantic Ocean. Nat. Commun. 11, 1–11. https://doi.org/10.1038/s41467-020-17932-9

Palanques, A., Guillén, J., 1998. Coastal changes in the Ebro delta: Natural and human factors. J. Coast. Conserv. 4, 17–26. https://doi.org/10.1007/BF02806485

Paradinas, L.M., James, N.A., Quinn, B., Dale, A., Narayanaswamy, B.E., 2021. A New Collection Tool-Kit to Sample Microplastics From the Marine Environment (Sediment, Seawater, and Biota) Using Citizen Science. Front. Mar. Sci. 8, 635. https://doi.org/10.3389/FMARS.2021.657709/BIBTEX

Pedrotti, M.L., Petit, S., Elineau, A., Bruzaud, S., Crebassa, J.-C., Dumontet, B., Martí, E., Gorsky, G., Cózar, A., 2016. Changes in the Floating Plastic

Pollution of the Mediterranean Sea in Relation to the Distance to Land. PLoS One 11, e0161581. https://doi.org/10.1371/journal.pone.0161581

Pedrotti, M.L., Petit, S., Eyheraguibel, B., Kerros, M.E., Elineau, A., Ghiglione, J.F., Loret, J.F., Rostan, A., Gorsky, G., 2021. Pollution by anthropogenic microfibers in North-West Mediterranean Sea and efficiency of microfiber removal by a wastewater treatment plant. Sci. Total Environ. 758, 144195. https://doi.org/10.1016/j.scitotenv.2020.144195

Peng, G., Zhu, B., Yang, D., Su, L., Shi, H., Li, D., 2017. Microplastics in sediments of the Changjiang Estuary, China. Environ. Pollut. 225, 283–290. https://doi.org/10.1016/j.envpol.2016.12.064

Pennino, M.G., Bachiller, E., Lloret-Lloret, E., Albo-Puigserver, M., Esteban, A., Jadaud, A., Bellido, J.M., Coll, M., 2020. Ingestion of microplastics and occurrence of parasite association in Mediterranean anchovy and sardine. Mar. Pollut. Bull. 158, 111399. https://doi.org/10.1016/j.marpolbul.2020.111399

Persson, L., Carney Almroth, B.M., Collins, C.D., Cornell, S., de Wit, C.A., Diamond, M.L., Fantke, P., Hassellöv, M., MacLeod, M., Ryberg, M.W., Søgaard Jørgensen, P., Villarrubia-Gómez, P., Wang, Z., Hauschild, M.Z., 2022. Outside the Safe Operating Space of the Planetary Boundary for Novel Entities. Environ. Sci. Technol. 56, 1510–1521. https://doi.org/10.1021/ACS.EST.1C04158/SUPPL_FILE/ES1C04158_SI_001.PDF

Piante, C., Ody, D., 2015. Blue Growth in the Mediterranean Sea: the Challenge of Good Environmental Status. MedTrends Project, Wwf.

Piehl, S., Mitterwallner, V., Atwood, E.C., Bochow, M., Laforsch, C., 2019. Abundance and distribution of large microplastics (1-5 mm) within beach sediments at the Po River Delta, northeast Italy. https://doi.org/10.1016/j.marpolbul.2019.110515

Pinardi, N., Arneri, E., Crise, A., Ravaioli, M., Zavatarelli, M., 2004. The physical, sedimentary and ecological structure and variability of shelf areas in the Mediterranean sea (27), in: Robinson, A.R., Brink, K. (Eds.), The Sea, The Global Costal Ocean. pp. 330–1243.

Pinardi, N., Masetti, E., 2000. Variability of the large scale general circulation of the Mediterranean Sea from observations and modelling: A review. Palaeogeogr. Palaeoclimatol. Palaeoecol. 158, 153–173. https://doi.org/10.1016/S0031-0182(00)00048-1

Piperagkas, O., Papageorgiou, N., Karakassis, I., 2019. Qualitative and quantitative assessment of microplastics in three sandy Mediterranean beaches, including different methodological approaches. https://doi.org/10.1016/j.ecss.2019.02.016

Pohl, F., Eggenhuisen, J.T., Kane, I.A., Clare, M.A., 2020. Transport and Burial of Microplastics in Deep-Marine Sediments by Turbidity Currents. Environ. Sci. Technol. 54, 4180–4189. https://doi.org/10.1021/acs.est.9b07527

Potrykus, M., Redko, V., Głowacka, K., Piotrowicz-Cieślak, A., Szarlej, P., Janik, H., Wolska, L., 2021. Polypropylene structure alterations after 5 years of natural degradation in a waste landfill. Sci. Total Environ. 758. https://doi.org/10.1016/j.scitotenv.2020.143649

Primpke, S., Christiansen, S.H., Cowger, W., De Frond, H., Deshpande, A., Fischer, M., Holland, E.B., Meyns, M., O'Donnell, B.A., Ossmann, B.E., Pittroff, M., Sarau, G., Scholz-Böttcher, B.M., Wiggin, K.J., 2020a. Critical Assessment of Analytical Methods for the Harmonized and Cost-Efficient Analysis of Microplastics. Appl. Spectrosc. 74, 1012–1047. https://doi.org/10.1177/0003702820921465

Primpke, S., Cross, R.K., Mintenig, S.M., Simon, M., Vianello, A., Gerdts, G., Vollertsen, J., 2020b. Toward the Systematic Identification of Microplastics in the Environment: Evaluation of a New Independent Software Tool (siMPle) for Spectroscopic Analysis. Appl. Spectrosc. 74, 1127–1138. https://doi.org/10.1177/0003702820917760

Provencher, J.F., Covernton, G.A., Moore, R.C., Horn, D.A., Conkle, J.L., Lusher, A.L., 2020. Proceed with caution: The need to raise the publication bar for microplastics research. Sci. Total Environ. https://doi.org/10.1016/j.scitotenv.2020.141426

QGIS Development Team, 2020. QGIS Geographic Information System. Open Source Geospatial Foundation Project. http://qgis.osgeo.org

Quinn, B., Murphy, F., Ewins, C., 2017. Validation of density separation for the rapid recovery of microplastics from sediment. Anal. Methods 9, 1491–1498. https://doi.org/10.1039/C6AY02542K

Ramirez-Llodra, E., De Mol, B., Company, J.B., Coll, M., Sardà, F., 2013. Effects of natural and anthropogenic processes in the distribution of marine litter in the deep Mediterranean Sea. Prog. Oceanogr. 118, 273–287. https://doi.org/10.1016/j.pocean.2013.07.027

Reisser, J., Slat, B., Noble, K., Du Plessis, K., Epp, M., Proietti, M., De Sonneville, J., Becker, T., Pattiaratchi, C., 2015. The vertical distribution of buoyant plastics at sea: An observational study in the North Atlantic Gyre. Biogeosciences 12, 1249–1256. https://doi.org/10.5194/bg-12-1249-2015

Remy, F., Collard, F., Gilbert, B., Compère, P., Eppe, G., Lepoint, G., 2015. When Microplastic Is Not Plastic: The Ingestion of Artificial Cellulose Fibers by Macrofauna Living in Seagrass Macrophytodetritus. Environ.

Sci. Technol. 49, 11158–11166. https://doi.org/10.1021/acs.est.5b02005

Renzi, M., Specchiulli, A., Blašković, A., Manzo, C., Mancinelli, G., Cilenti, L., 2019. Marine litter in stomach content of small pelagic fishes from the Adriatic Sea: sardines (Sardina pilchardus) and anchovies (Engraulis encrasicolus). Environ. Sci. Pollut. Res. 26, 2771–2781. https://doi.org/10.1007/s11356-018-3762-8

Rey, S.F., Franklin, J., Rey, S.J., 2021. Microplastic pollution on island beaches, Oahu, Hawai`i. PLoS One 16, e0247224. https://doi.org/10.1371/JOURNAL.PONE.0247224

Rist, S., Vianello, A., Winding, M.H.S., Nielsen, T.G., Almeda, R., Torres, R.R., Vollertsen, J., 2020. Quantification of plankton-sized microplastics in a productive coastal Arctic marine ecosystem. Environ. Pollut. 266, 115248. https://doi.org/10.1016/J.ENVPOL.2020.115248

Rochman, C.M., Brookson, C., Bikker, J., Djuric, N., Earn, A., Bucci, K., Athey, S., Huntington, A., McIlwraith, H., Munno, K., De Frond, H., Kolomijeca, A., Erdle, L., Grbic, J., Bayoumi, M., Borrelle, S.B., Wu, T., Santoro, S., Werbowski, L.M., Zhu, X., Giles, R.K., Hamilton, B.M., Thaysen, C., Kaura, A., Klasios, N., Ead, L., Kim, J., Sherlock, C., Ho, A., Hung, C., 2019. Rethinking microplastics as a diverse contaminant suite. Environ. Toxicol. Chem. 38, 703–711. https://doi.org/10.1002/etc.4371

Rodrigues, M.O., Gonçalves, A.M.M., Gonçalves, F.J.M., Nogueira, H., Marques, J.C., Abrantes, N., 2018. Effectiveness of a methodology of microplastics isolation for environmental monitoring in freshwater systems. Ecol. Indic. 89, 488–495. https://doi.org/10.1016/j.ecolind.2018.02.038

Rodrigues, S.M., Almeida, C.M.R., Silva, D., Cunha, J., Antunes, C., Freitas, V., Ramos, S., 2019. Microplastic contamination in an urban estuary: Abundance and distribution of microplastics and fish larvae in the Douro estuary. Sci. Total Environ. 659, 1071–1081. https://doi.org/10.1016/J.SCITOTENV.2018.12.273

Rodríguez-Romeu, O., Constenla, M., Carrassón, M., Campoy-Quiles, M., Soler-Membrives, A., 2020. Are anthropogenic fibres a real problem for red mullets (Mullus barbatus) from the NW Mediterranean? Sci. Total Environ. 733, 139336. https://doi.org/10.1016/j.scitotenv.2020.139336

Rohling, E., Abu-Zied, R., Rohling, E., Abu-Zied, R., 2009. The Marine Environment: Present and Past, in: Woodward, J. (Ed.), The Physical Geography of the Mediterranean. Oxford University Press, Oxford, pp. 33–67. https://doi.org/10.1093/oso/9780199268030.003.0012

RStudio Team, 2020. RStudio: Integrated Development for R. RStudio, PBC,

Boston, MA URL http://www.rstudio.com/

Ruiz-Orejón, L.F., Sardá, R., Ramis-Pujol, J., 2018. Now, you see me: High concentrations of floating plastic debris in the coastal waters of the Balearic Islands (Spain). Mar. Pollut. Bull. 133, 636–646. https://doi.org/10.1016/j.marpolbul.2018.06.010

Ryan, P.G., Moloney, C.L., 1990. Plastic and other artefacts on South African beaches: temporal trends in abundance and composition. S. Afr. J. Sci. 86, 450–452.

Sadri, S.S., Thompson, R.C., 2014. On the quantity and composition of floating plastic debris entering and leaving the Tamar Estuary, Southwest England. Mar. Pollut. Bull. 81, 55–60. https://doi.org/10.1016/J.MARPOLBUL.2014.02.020

Sanchez-Cabeza, J.A., Masqué, P., Ani-Ragolta, I., 1998. 210Pb and210Po analysis in sediments and soils by microwave acid digestion. J. Radioanal. Nucl. Chem. 227, 19–22. https://doi.org/10.1007/BF02386425

Sanchez-Vidal, A., Canals, M., de Haan, W.P., Romero, J., Veny, M., 2021. Seagrasses provide a novel ecosystem service by trapping marine plastics. Sci. Rep. 11, 254. https://doi.org/10.1038/s41598-020-79370-3

Sanchez-Vidal, A., Thompson, R.C., Canals, M., De Haan, W.P., 2018. The imprint of microfibres in southern European deep seas. https://doi.org/10.1371/journal.pone.0207033

SAPEA, S.A. for P. by E.A., 2019. A Scientific Perspective on Microplastics in Nature and Society., Evidence Review Report. https://doi.org/10.26356/microplastics

Schmidt, C., Krauth, T., Wagner, S., 2017. Export of Plastic Debris by Rivers into the Sea. Environ. Sci. Technol. 51, 12246–12253. https://doi.org/10.1021/ACS.EST.7B02368/SUPPL_FILE/ES7B02368_SI_002.ZIP

Schneider, C.A., Rasband, W.S., Eliceiri, K.W., 2012. NIH Image to ImageJ: 25 years of image analysis. Nat. Methods 9, 671–675. https://doi.org/10.1038/nmeth.2089

Shabaka, S.H., Ghobashy, M., Marey, R.S., 2019. Identification of marine microplastics in Eastern Harbor, Mediterranean Coast of Egypt, using differential scanning calorimetry. Mar. Pollut. Bull. 142, 494–503. https://doi.org/10.1016/j.marpolbul.2019.03.062

Shiber, J.G., 1979. Plastic pellets on the coast of Lebanon. Mar. Pollut. Bull. 10, 28–30. https://doi.org/10.1016/0025-326X(79)90321-7

Simon-Sánchez, L., Grelaud, M., Franci, M., Ziveri, P., 2021. Are research

methods shaping our understanding of microplastic pollution? A literature review on the seawater and sediment bodies of the Mediterranean Sea. Environ. Pollut. 292, 118275. https://doi.org/10.1016/j.envpol.2021.118275

Simon-Sánchez, L., Grelaud, M., Garcia-Orellana, J., Ziveri, P., 2019. River Deltas as hotspots of microplastic accumulation: The case study of the Ebro River (NW Mediterranean). Sci. Total Environ. 687, 1186–1196. https://doi.org/10.1016/J.SCITOTENV.2019.06.168

Simon, M., van Alst, N., Vollertsen, J., 2018. Quantification of microplastic mass and removal rates at wastewater treatment plants applying Focal Plane Array (FPA)-based Fourier Transform Infrared (FT-IR) imaging. Water Res. 142, 1–9. https://doi.org/10.1016/J.WATRES.2018.05.019

Smeaton, C., 2021. Augmentation of global marine sedimentary carbon storage in the age of plastic. Limnol. Oceanogr. Lett. 6, 113–118. https://doi.org/10.1002/LOL2.10187

Song, Y.K., Hong, S.H., Jang, M., Han, G.M., Rani, M., Lee, J., Shim, W.J., 2015. A comparison of microscopic and spectroscopic identification methods for analysis of microplastics in environmental samples. Mar. Pollut. Bull. 93, 202–209. https://doi.org/10.1016/j.marpolbul.2015.01.015

Stanton, T., Johnson, M., Nathanail, P., MacNaughtan, W., Gomes, R.L., 2019. Freshwater and airborne textile fibre populations are dominated by 'natural', not microplastic, fibres. Sci. Total Environ. 666, 377–389. https://doi.org/10.1016/j.scitotenv.2019.02.278

Steinmetz, Z., Wollmann, C., Schaefer, M., Buchmann, C., David, J., Tröger, J., Muñoz, K., Frör, O., Schaumann, G.E., 2016. Plastic mulching in agriculture. Trading short-term agronomic benefits for long-term soil degradation? Sci. Total Environ. 550, 690–705. https://doi.org/10.1016/j.scitotenv.2016.01.153

Stow, D., Smillie, Z., Esentia, I., 2019. Deep-sea bottom currents: Their nature and distribution, in: Encyclopedia of Ocean Sciences. Elsevier, pp. 90–96. https://doi.org/10.1016/B978-0-12-409548-9.10878-4

Struglia, M.V., Mariotti, A., Filograsso, A., 2004. River Discharge into the Mediterranean Sea: Climatology and Aspects of the Observed Variability. J. Clim. 17, 4740–4751. https://doi.org/10.1175/JCLI-3225.1

Suaria, G., Achtypi, A., Perold, V., Lee, J.R., Pierucci, A., Bornman, T.G., Aliani, S., Ryan, P.G., 2020. Microfibers in oceanic surface waters: A global characterization. Sci. Adv. 6, 8493–8498. https://doi.org/10.1126/sciadv.aay8493

Suaria, G., Avio, C.G., Mineo, A., Lattin, G.L., Magaldi, M.G., Belmonte, G.,

Moore, C.J., Regoli, F., Aliani, S., 2016. The Mediterranean Plastic Soup: Synthetic polymers in Mediterranean surface waters. Sci. Rep. 6, 1–10. https://doi.org/10.1038/srep37551

Sundt, P., Schultze, P.-E., Syversen, F., 2014. Sources of microplastic-pollution to the marine environment Project report. (Miljødirektoratet), Nor. Environ. Agency 86. https://doi.org/M-321|2015

Talvitie, J., Heinonen, M., Pääkkönen, J.P., Vahtera, E., Mikola, A., Setälä, O., Vahala, R., 2015. Do wastewater treatment plants act as a potential point source of microplastics? Preliminary study in the coastal Gulf of Finland, Baltic Sea. Water Sci. Technol. 72, 1495–1504. https://doi.org/10.2166/wst.2015.360

Ter Halle, A., Jeanneau, L., Martignac, M., Jardé, E., Pedrono, B., Brach, L., Gigault, J., 2017a. Nanoplastic in the North Atlantic Subtropical Gyre. Environ. Sci. Technol. 51, 13689–13697. https://doi.org/10.1021/ACS.EST.7B03667/SUPPL_FILE/ES7B03667_SI_001.PDF

Ter Halle, A., Ladirat, L., Martignac, M., Mingotaud, A.F., Boyron, O., Perez, E., 2017b. To what extent are microplastics from the open ocean weathered? *. https://doi.org/10.1016/j.envpol.2017.04.051

Teuten, Emma L., Saquing, J.M., Knappe, D.R.U., Barlaz, M.A., Jonsson, S., Björn, A., Rowland, S.J., Thompson, R.C., Galloway, T.S., Yamashita, R., Ochi, D., Watanuki, Y., Moore, C., Viet, P.H., Tana, T.S., Prudente, M., Boonyatumanond, R., Zakaria, M.P., Akkhavong, K., Ogata, Y., Hirai, H., Iwasa, S., Mizukawa, K., Hagino, Y., Imamura, A., Saha, M., Takada, H., 2009. Transport and release of chemicals from plastics to the environment and to wildlife. Philos. Trans. R. Soc. B Biol. Sci. 364, 2027–2045. https://doi.org/10.1098/rstb.2008.0284

Thompson, R.C., Olsen, Y., Mitchell, R.P., Davis, A., Rowland, S.J., John, A.W.G., McGonigle, D., Russell, A.E., 2004. Supporting online material to: Lost at sea: where is all the plastic? Science 304, 838. https://doi.org/10.1126/science.1094559

Torre, M., Digka, N., Anastasopoulou, A., Tsangaris, C., Mytilineou, C., 2016. Anthropogenic microfibres pollution in marine biota. A new and simple methodology to minimize airborne contamination. Mar. Pollut. Bull. 113, 55–61. https://doi.org/10.1016/J.MARPOLBUL.2016.07.050

Tsang, Y.Y., Mak, C.W., Liebich, C., Lam, S.W., Sze, E.T.P., Chan, K.M., 2017. Microplastic pollution in the marine waters and sediments of Hong Kong. Mar. Pollut. Bull. 115, 20–28. https://doi.org/10.1016/j.marpolbul.2016.11.003

Tu, C., Chen, T., Zhou, Q., Liu, Y., Wei, J., Waniek, J.J., Luo, Y., 2020. Biofilm

formation and its influences on the properties of microplastics as affected by exposure time and depth in the seawater. Sci. Total Environ. 734, 139237. https://doi.org/10.1016/j.scitotenv.2020.139237

Tubau, X., Canals, M., Lastras, G., Rayo, X., Rivera, J., Amblas, D., 2015. Marine litter on the floor of deep submarine canyons of the Northwestern Mediterranean Sea: The role of hydrodynamic processes. Prog. Oceanogr. 134, 379–403. https://doi.org/10.1016/j.pocean.2015.03.013

Turner, A., Holmes, L., 2011. Occurrence, distribution and characteristics of beached plastic production pellets on the island of Malta (central Mediterranean). Mar. Pollut. Bull. 62, 377–381. https://doi.org/10.1016/j.marpolbul.2010.09.027

UNED-MAP, 2012. State of the Marine and Coastal Mediterranean. UNEP/MAP – Barcelona Conv. 96.

UNEP, 2020. State of the Environment and Development in the Mediterranean: Summary for Decision Makers.

UNEP, 2011. Plastic Debris in the Ocean 20–33. https://doi.org/10.1021/es400931b.8

UNWTO, 2018. UNWTO Tourism Highlights: 2018 Edition, UNWTO Tourism Highlights: 2018 Edition. https://doi.org/10.18111/9789284419876

Van Cauwenberghe, L., Vanreusel, A., Mees, J., Janssen, C.R., 2013. Microplastic pollution in deep-sea sediments. Environ. Pollut. 182, 495–499. https://doi.org/10.1016/J.ENVPOL.2013.08.013

van der Hal, N., Ariel, A., Angel, D.L., 2017. Exceptionally high abundances of microplastics in the oligotrophic Israeli Mediterranean coastal waters. Mar. Pollut. Bull. 116, 151–155. https://doi.org/10.1016/j.marpolbul.2016.12.052

Van Sebille, E., Aliani, S., Law, K.L., Maximenko, N., Alsina, J.M., Bagaev, A., Bergmann, M., Chapron, B., Chubarenko, I., Cózar, A., Delandmeter, P., Egger, M., Fox-Kemper, B., Garaba, S.P., Goddijn-Murphy, L., Hardesty, B.D., Hoffman, M.J., Isobe, A., Jongedijk, C.E., Kaandorp, M.L.A., Khatmullina, L., Koelmans, A.A., Kukulka, T., Laufkötter, C., Lebreton, L., Lobelle, D., Maes, C., Martinez-Vicente, V., Morales Maqueda, M.A., Poulain-Zarcos, M., Rodríguez, E., Ryan, P.G., Shanks, A.L., Shim, W.J., Suaria, G., Thiel, M., Van Den Bremer, T.S., Wichmann, D., 2020. The physical oceanography of the transport of floating marine debris. Environ. Res. Lett. https://doi.org/10.1088/1748-9326/ab6d7d

Van Sebille, E., Wilcox, C., Lebreton, L., Maximenko, N., Hardesty, B.D., Van Franeker, J.A., Eriksen, M., Siegel, D., Galgani, F., Law, K.L., 2015. A

global inventory of small floating plastic debris. Environ. Res. Lett. 10, 124006. https://doi.org/10.1088/1748-9326/10/12/124006

Vermeiren, P., Muñoz, C.C., Ikejima, K., 2016. Sources and sinks of plastic debris in estuaries: A conceptual model integrating biological, physical and chemical distribution mechanisms. Mar. Pollut. Bull. 113, 7–16. https://doi.org/10.1016/J.MARPOLBUL.2016.10.002

Vianello, A, Boldrin, A., Guerriero, P., Moschino, V., Rella, R., Sturaro, A., Da Ros, L., 2013. Microplastic particles in sediments of Lagoon of Venice, Italy: First observations on occurrence, spatial patterns and identification. https://doi.org/10.1016/j.ecss.2013.03.022

Vianello, A., Da Ros, L., Boldrin, A., Marceta, T., Moschino, V., 2018. First evaluation of floating microplastics in the Northwestern Adriatic Sea. Environ. Sci. Pollut. Res. 25, 28546–28561. https://doi.org/10.1007/s11356-018-2812-6

Vianello, A., Jensen, R.L., Liu, L., Vollertsen, J., 2019. Simulating human exposure to indoor airborne microplastics using a Breathing Thermal Manikin. Sci. Reports 2019 91 9, 1–11. https://doi.org/10.1038/s41598-019-45054-w

Villarrubia-Gómez, P., Cornell, S.E., Fabres, J., 2018. Marine plastic pollution as a planetary boundary threat – The drifting piece in the sustainability puzzle. Mar. Policy 96, 213–220. https://doi.org/10.1016/J.MARPOL.2017.11.035

Vörösmarty, C.J., Syvitski, J., Day, J., de Sherbinin, A., Giosan, L., Paola, C., 2009. Battling to Save the World's River Deltas. Bull. At. Sci. 65, 31–43. https://doi.org/10.2968/065002005

Wagner, M., Scherer, C., Alvarez-Muñoz, D., Brennholt, N., Bourrain, X., Buchinger, S., Fries, E., Grosbois, C., Klasmeier, J., Marti, T., Rodriguez-Mozaz, S., Urbatzka, R., Vethaak, A.D., Winther-Nielsen, M., Reifferscheid, G., 2014. Microplastics in freshwater ecosystems: what we know and what we need to know. Environ. Sci. Eur. 26, 1–9. https://doi.org/10.1186/s12302-014-0012-7

Wakkaf, T., El Zrelli, R., Kedzierski, M., Balti, R., Shaiek, M., Mansour, L., Tlig-Zouari, S., Bruzaud, S., Rabaoui, L., 2020a. Characterization of microplastics in the surface waters of an urban lagoon (Bizerte lagoon, Southern Mediterranean Sea): Composition, density, distribution, and influence of environmental factors. Mar. Pollut. Bull. 160, 111625. https://doi.org/10.1016/J.MARPOLBUL.2020.111625

Wakkaf, T., El Zrelli, R., Kedzierski, M., Balti, R., Shaiek, M., Mansour, L., Tlig-Zouari, S., Bruzaud, S., Rabaoui, L., 2020b. Microplastics in edible mussels from a southern Mediterranean lagoon: Preliminary results on

seawater-mussel transfer and implications for environmental protection and seafood safety. Mar. Pollut. Bull. 158, 111355. https://doi.org/10.1016/J.MARPOLBUL.2020.111355

Wang, J., Peng, J., Tan, Z., Gao, Y., Zhan, Z., Chen, Q., Cai, L., 2017. Microplastics in the surface sediments from the Beijiang River littoral zone: Composition, abundance, surface textures and interaction with heavy metals. Chemosphere 171, 248–258. https://doi.org/10.1016/j.chemosphere.2016.12.074

Wang, J., Tan, Z., Peng, J., Qiu, Q., Li, M., 2016. The behaviors of microplastics in the marine environment. Mar. Environ. Res. 113, 7–17. https://doi.org/10.1016/J.MARENVRES.2015.10.014

Ward, J.E., Rosa, M., Shumway, S.E., 2019. Capture, ingestion, and egestion of microplastics by suspension-feeding bivalves: A 40-year history. Anthr. Coasts 2, 39–49. https://doi.org/10.1139/ANC-2018-0027/ASSET/IMAGES/LARGE/ANC-2018-0027F2.JPEG

Weckhuysen, B., Meirer, F., Oord, R., Zettler, E., Van Sebille, E., 2021. Nanoscale Infrared Spectroscopy Reveals Nanoplastics at 5000 m Depth in the South Atlantic Ocean. https://doi.org/10.21203/RS.3.RS-955379/V1

Wessel, C.C., Lockridge, G.R., Battiste, D., Cebrian, J., 2016. Abundance and characteristics of microplastics in beach sediments: Insights into microplastic accumulation in northern Gulf of Mexico estuaries. Mar. Pollut. Bull. 109, 178–183. https://doi.org/10.1016/j.marpolbul.2016.06.002

Whitmire, L.S., Bloem, S.J. Van, 2017. Quantification of Microplastics on National Park Beaches.NOAA Marine Debris Program;2017.

Wickham, H., 2016. ggplot2: Elegant Graphics for Data Analysis. Springer-Verlag New York. ISBN 978-3-319-24277-4, https://ggplot2.tidyverse.org

Wilke, C., 2020. cowplot: Streamlined Plot Theme and Plot Annotations for 'ggplot2'. https://wilkelab.org/cowplot/

Williams, A.T., Simmons, S.L., 1996. The degradation of plastic litter in rivers: Implications for beaches. J. Coast. Conserv. 2, 63–72. https://doi.org/10.1007/BF02743038

Windsor, F.M., Durance, I., Horton, A.A., Thompson, R.C., Tyler, C.R., Ormerod, S.J., 2019. A catchment-scale perspective of plastic pollution. Glob. Chang. Biol. 25, 1207–1221. https://doi.org/10.1111/gcb.14572

Woodall, L.C., Sanchez-Vidal, A., Canals, M., Paterson, G.L.J., Coppock, R., Sleight, V., Calafat, A., Rogers, A.D., Narayanaswamy, B.E., Thompson,

R.C., 2014. The deep sea is a major sink for microplastic debris. R. Soc. Open Sci. 1. https://doi.org/10.1098/rsos.140317

Woods, M.N., Stack, M.E., Fields, D.M., Shaw, S.D., Matrai, P.A., 2018. Microplastic fiber uptake, ingestion, and egestion rates in the blue mussel (Mytilus edulis). Mar. Pollut. Bull. 137, 638–645. https://doi.org/10.1016/J.MARPOLBUL.2018.10.061

Xiong, X., Wu, C., Elser, J.J., Mei, Z., Hao, Y., 2019. Occurrence and fate of microplastic debris in middle and lower reaches of the Yangtze River – From inland to the sea. Sci. Total Environ. 659, 66–73. https://doi.org/10.1016/J.SCITOTENV.2018.12.313

Yabanlı, M., Yozukmaz, A., Şener, İ., Ölmez, Ö.T., 2019. Microplastic pollution at the intersection of the Aegean and Mediterranean Seas: A study of the Datça Peninsula (Turkey). Mar. Pollut. Bull. 145, 47–55. https://doi.org/10.1016/j.marpolbul.2019.05.003

Yan, M., Nie, H., Xu, K., He, Y., Hu, Y., Huang, Y., Wang, J., 2019. Microplastic abundance, distribution and composition in the Pearl River along Guangzhou city and Pearl River estuary, China. Chemosphere 217, 879–886. https://doi.org/10.1016/J.CHEMOSPHERE.2018.11.093

Yu, X., Peng, J., Wang, J., Wang, K., Bao, S., 2016. Occurrence of microplastics in the beach sand of the Chinese inner sea: the Bohai Sea. Environ. Pollut. 214, 722–730. https://doi.org/10.1016/J.ENVPOL.2016.04.080

Zarfl, C., 2019. Promising techniques and open challenges for microplastic identification and quantification in environmental matrices. Anal. Bioanal. Chem. 411, 3743–3756. https://doi.org/10.1007/S00216-019-01763-9/FIGURES/2

Zeri, C., Adamopoulou, A., Bojanić Varezić, D., Fortibuoni, T., Kovač Viršek, M., Kržan, A., Mandic, M., Mazziotti, C., Palatinus, A., Peterlin, M., Prvan, M., Ronchi, F., Siljic, J., Tutman, P., Vlachogianni, T., 2018. Floating plastics in Adriatic waters (Mediterranean Sea): From the macro- to the micro-scale. Mar. Pollut. Bull. 136, 341–350. https://doi.org/10.1016/j.marpolbul.2018.09.016

Zhang, K., Gong, W., Lv, J., Xiong, X., Wu, C., 2015. Accumulation of floating microplastics behind the Three Gorges Dam. Environ. Pollut. 204, 117–123. https://doi.org/10.1016/J.ENVPOL.2015.04.023

Zhang, K., Su, J., Xiong, X., Wu, X., Wu, C., Liu, J., 2016. Microplastic pollution of lakeshore sediments from remote lakes in Tibet plateau, China. Environ. Pollut. 219, 450–455. https://doi.org/10.1016/j.envpol.2016.05.048

Zhao, S., Zhu, L., Wang, T., Li, D., 2014. Suspended microplastics in the

surface water of the Yangtze Estuary System, China: First observations on occurrence, distribution. Mar. Pollut. Bull. 86, 562–568. https://doi.org/10.1016/j.marpolbul.2014.06.032

Ziajahromi, S., Neale, P.A., Rintoul, L., Leusch, F.D.L., 2017. Wastewater treatment plants as a pathway for microplastics: Development of a new approach to sample wastewater-based microplastics. Water Res. 112, 93–99. https://doi.org/10.1016/j.watres.2017.01.042

Zitko, V., Hanlon, M., 1991. Another source of pollution by plastics: Skin cleaners with plastic scrubbers. Mar. Pollut. Bull. 22, 41–42. https://doi.org/10.1016/0025-326X(91)90444-W

Zografos, C., 2017. Flows of sediment, flows of insecurity: Climate change adaptation and the social contract in the Ebro Delta, Catalonia. Geoforum 80, 49–60. https://doi.org/10.1016/j.geoforum.2017.01.004

Supplementary material - Chapter 2

Table S2.1. References included in the literature review.

Table S2. 2. Literature data on plastics abundance in Mediterranean surface waters.

References

Table S2.1. References included in the literature review

The data obtained from these references – compartment, location, methods and occurrence- are the baseline of the current literature review. The data is visualized in the figures of the main manuscript.

Compartment	Reference	DOI
Beach	(Fanini and Bozzeda, 2018)	10.1016/j.ecolind.2018.02.027
Beach	(Abidli et al., 2018)	10.1016/j.ecss.2018.03.006
Beach	(Piperagkas et al., 2019)	10.1016/j.ecss.2019.02.016
Beach	(Misic et al., 2019)	10.1016/j.ecss.2019.106429
Beach	(Bayo et al., 2019)	10.1016/j.envpol.2019.06.024
Beach	(Turner and Holmes, 2011)	10.1016/j.marpolbul.2010.09.027
Beach	(Laglbauer et al., 2014)	10.1016/j.marpolbul.2014.09.036
Beach	(Guerranti et al., 2017)	10.1016/j.marpolbul.2017.02.021
Beach	(Cannas et al., 2017)	10.1016/j.marpolbul.2017.04.008
Beach	(Lots et al., 2017)	10.1016/j.marpolbul.2017.08.057
Beach	(Karkanorachaki et al., 2018)	10.1016/j.marpolbul.2018.06.011
Beach	(Blašković et al., 2018)	10.1016/j.marpolbul.2018.07.021
Beach	(Duncan et al., 2018)	10.1016/j.marpolbul.2018.09.019
Beach	(Maršić-Lučić et al., 2018)	10.1016/j.marpolbul.2018.10.027
Beach	(Atwood et al., 2019)	10.1016/j.marpolbul.2018.11.045
Beach	(Constant et al., 2019)	10.1016/j.marpolbul.2019.03.032
Beach	(Shabaka et al., 2019)	10.1016/j.marpolbul.2019.03.062
Beach	(Yabanlı et al., 2019)	10.1016/j.marpolbul.2019.05.003
Beach	(Korez et al., 2019)	10.1016/j.marpolbul.2019.05.054
Beach	(Chouchene et al., 2019)	10.1016/j.marpolbul.2019.06.004
Beach	(Piehl et al., 2019)	10.1016/j.marpolbul.2019.110515
Beach	(Tziourrou et al., 2019)	10.1016/j.marpolbul.2019.110531
Beach	(Simon-Sánchez et al., 2019)	10.1016/j.scitotenv.2019.06.168
Beach	(Tata et al., 2020)	10.1016/j.scitotenv.2020.136664
Beach	(Munari et al., 2017)	10.1016/j.wasman.2017.05.020
Beach	(Ceccarini et al., 2018)	10.1021/acs.est.8b01487
Beach	(De Ruijter et al., 2019)	10.12681/mms.19131
Beach	(Godoy et al., 2020)	10.1016/j.scitotenv.2020.142023
Beach	(Merlino et al., 2020)	10.3390/w12123389
Beach	(Missawi et al., 2020)	10.1016/j.envpol.2020.114634
Sea surface water	(Faure et al., 2015)	10.1007/s11356-015-4453-3
Sea surface water	(Vianello et al., 2018)	10.1007/s11356-018-2812-6

Sea surface water	(Fossi et al., 2016)	10.1016/j.envpol.2015.11.022
Sea surface water	(Gündoğdu et al., 2018)	10.1016/j.envpol.2018.04.042
Sea surface water	(Ruiz-Orejón et al., 2019)	10.1016/j.envpol.2019.06.063
Sea surface water	(de Lucia et al., 2014)	10.1016/j.marenvres.2014.03.017
Sea surface water	(Ruiz-Orejón et al., 2016)	10.1016/j.marenvres.2016.08.001
Sea surface water	(Compa et al., 2020)	10.1016/j.marenvres.2020.104945
Sea surface water	(Collignon et al., 2012)	10.1016/j.marpolbul.2012.01.011
Sea surface water	(Fossi et al., 2012)	10.1016/j.marpolbul.2012.08.013
Sea surface water	(Collignon et al., 2014)	10.1016/j.marpolbul.2013.11.023
Sea surface water	(Gajšt et al., 2016)	10.1016/j.marpolbul.2016.10.031
Sea surface water	(van der Hal et al., 2017)	10.1016/j.marpolbul.2016.12.052
Sea surface water	(Gündoğdu and Çevik, 2017)	10.1016/j.marpolbul.2017.03.002
Sea surface water	(Ruiz-Orejón et al., 2018)	10.1016/j.marpolbul.2018.06.010
Sea surface water	(Baini et al., 2018)	10.1016/j.marpolbul.2018.06.016
Sea surface water	(Tunçer et al., 2018)	10.1016/j.marpolbul.2018.06.054
Sea surface water	(Zeri et al., 2018)	10.1016/j.marpolbul.2018.09.016
Sea surface water	(Palatinus et al., 2019)	10.1016/j.marpolbul.2018.12.038
Sea surface water	(de Haan et al., 2019)	10.1016/j.marpolbul.2019.01.053
Sea surface water	(Caldwell et al., 2019)	10.1016/j.marpolbul.2019.110572
Sea surface water	(Schmidt et al., 2018)	10.1016/j.pocean.2017.11.010
Sea surface water	(Kazour et al., 2019)	10.1016/j.scitotenv.2019.133933
Sea surface water	(Camins et al., 2020)	10.1016/j.scitotenv.2019.136178
Sea surface water	(Suaria et al., 2016)	10.1038/srep37551
Sea surface water	(Panti et al., 2015)	10.1071/EN14234
Sea surface water	(Gündoğdu, 2017)	10.12714/egejfas.2017.34.4.06
Sea surface water	(Cózar et al., 2015)	10.1371/journal.pone.0121762
Sea surface water	(Pedrotti et al., 2016)	10.1371/journal.pone.0161581
Sea surface water	(Fossi et al., 2017)	10.3389/fmars.2017.00167
Sea surface water	(de Lucia et al., 2018)	10.3390/w10081108
Sea surface water	(Güven et al., 2017)	10.1016/j.envpol.2017.01.025
Sea surface water	(Caldwell et al., 2020)	10.1016/j.marpolbul.2020.111515
Sea surface water	(Tanhua et al., 2020)	10.1371/journal.pone.0243203
Sea surface water	(Zayen et al., 2020)	10.1016/j.ecss.2020.106832
Sea surface water	(Wakkaf et al., 2020a)	10.1016/j.marpolbul.2020.111625
Marine sediments	(Filgueiras et al., 2019)	10.1007/s11356-019-05341-5
Marine sediments	(Mistri et al., 2018)	10.1007/s12210-018-0736-1
Marine sediments	(Vianello et al., 2013)	10.1016/j.ecss.2013.03.022

Marine sediments	(D'Alessandro et al., 2018)	10.1016/j.envpol.2018.08.002
Marine sediments	(Krüger et al., 2019)	10.1016/j.envpol.2019.113336
Marine sediments	(Alomar et al., 2016)	10.1016/j.marenvres.2016.01.005
Marine sediments	(Fastelli et al., 2016)	10.1016/j.marpolbul.2016.08.054
Marine sediments	(Blašković et al., 2017)	10.1016/j.marpolbul.2016.09.018
Marine sediments	(Munari et al., 2017)	10.1016/j.marpolbul.2017.07.063
Marine sediments	(Renzi et al., 2018)	10.1016/j.marpolbul.2018.03.019
Marine sediments	(Renzi and Blašković, 2020)	10.1016/j.marpolbul.2020.110994
Marine sediments	(Palatinus et al., 2019)	10.1016/j.marpolbul.2018.12.038
Marine sediments	(Mistri et al., 2020)	10.1016/j.marpolbul.2020.111016
Marine sediments	(Kazour et al., 2019)	10.1016/j.scitotenv.2019.133933
Marine sediments	(Woodall et al., 2014)	10.1098/rsos.140317
Marine sediments	(Kane et al., 2020)	10.1126/science.aba5899
Marine sediments	(De Ruijter et al., 2019)	10.12681/mms.19131
Marine sediments	(Sanchez-Vidal et al., 2018)	10.1371/journal.pone.0207033
Marine sediments	(Van Cauwenberghe et al., 2013)	10.1016/j.envpol.2013.08.013
Marine sediments	(Piazzolla et al., 2020)	10.1016/j.ecss.2020.106819
Marine sediments	(Renzi et al., 2020)	10.1016/j.marpolbul.2020.110918
Sea water column	(Fossi et al., 2012)	10.1016/j.marpolbul.2012.08.013
Sea water column	(Baini et al., 2018)	10.1016/j.marpolbul.2018.06.016
Sea water column	(Lefebvre et al., 2019)	10.1016/j.marpolbul.2019.03.025
Sea water column	(de Lucia et al., 2018)	10.3390/w10081108
Sea water column	(Wakkaf et al., 2020b)	10.1016/j.marpolbul.2020.111355
Sea water column	(Güven et al., 2017)	10.1016/j.envpol.2017.01.025

Reference	Mesh Size	Size fraction	items·km^{-2}					g·km^{-2}					items·m^{-3}				
			median	Q1	Q3	mean	sd	median	Q1	Q3	mean	sd	median	Q1	Q3	mean	sd
Collignon et al. (2014)	200 µm	< 5 mm	6250	0	19375	33729	82154	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Fossi et al. (2012)	200 µm	< 5 mm; > 5mm	20000	0	14250	31087	10027	NA	NA	NA	NA	NA	0.04	0.0	0.2	0.62	2.01
Caldwell et al. (2019)	330 µm	< 5 mm	23132	6885	28428	28376	28917	77.4	24.1	365.3	268.6	421.2	NA	NA	NA	NA	NA
Schmidt et al. (2018)	780 µm	< 5 mm	43245	2460	10214	14575	27532	NA	NA	NA	61.9	178.0	NA	NA	NA	NA	NA
Baini et al. (2018)	330 µm	< 5 mm	43282	5	8	1	6	14.5	7.6	44.0	41.1	68.6	NA	NA	NA	NA	NA
Camins et al. (2019)	330 µm	< 5 mm	48850	1380	74041	69161	83243	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Fossi et al. (2017)	330 µm	< 5 mm; > 5mm	50000	2887	86600	94517	11870	14.5	6.8	16.5	19.9	23.8	NA	NA	NA	NA	NA
Palatinus et al. (2019)	308 µm	< 5 mm; > 5mm	55764	3000	10000	85238	74540	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Caldwell et al. (2020)	330 µm	< 5 mm; > 5mm	56999	4073	78350	12752	29472	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Ruiz-Orejón et al. (2016)	333 µm	< 5 mm	59395	2465	95405	25586	84122	175.7	47.2	286.7	394.2	760.9	NA	NA	NA	NA	NA
Zayen et al. (2020)	333 µm	< 5 mm; > 5mm	62151	2879	77997	15212	21100	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Pedrotti et al. (2016)	335 µm	< 5 mm; > 5mm	72178	4157	63739	14741	28748	141.0	52.4	536.5	579.3	1313.8	NA	NA	NA	NA	NA
De Haan et al. (2019)	335 µm	< 5 mm; > 5mm	76000	5034	77997	15395	11946	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
				3	9	4	2	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
				4700	11000	10266	10521		13.0	29.0				NA	NA		
				0	0	7	3	19.00	0	0	26.52	22.26	NA	NA	NA	NA	NA

Ruiz-Orejón et al. (2019)	333 µm	< 5 mm	10148 5	4792 1	23661 7	16101 6	15319 4	129.0 4	42.7 2	239. 74	1105. 19	3657. 00	0.38	0.1 8	0.9 7	0.66	0.63
Güven et al. (2017)	333 µm	< 5 mm	11848 0	6565 4	14316 5	14041 8	12067 1	NA	NA	NA	NA	NA	NA	N A	N A	NA	NA
Faure et al. (2015)	333 µm	< 5 mm; > 5mm	NA	NA	NA	12962 8	NA	NA	NA	NA	216	NA	NA	N A	N A	NA	NA
Gajšt et al. (2016)	300 µm	< 5 mm	15026 5	1006 64	31319 3	47176 7	82815 4	NA	NA	NA	NA	NA	2.01	1.3 5	4.1 8	6.29	11.0 5
Gündoğdu & Çevik. (2017)	333 µm	< 5 mm	18273 7	1680 00	31315 8	37073 7	43236 5	NA	NA	NA	NA	NA	NA	N A	N A	NA	NA
Vianello et al. (2018)	330 µm	< 5 mm; > 5mm	22722 2	8732 7	36771 0	12008 61	26830 14	65.0	20.0	152. 5	1503. 8	5476. 0	NA	N A	N A	NA	NA
Tunçer et al. (2018)	333 µm	< 5 mm; > 5mm	38997 7	1957 76	14430 15	11535 90	15074 35	NA	NA	NA	NA	NA	NA	N A	N A	NA	NA
Ruiz-Orejón et al. (2018)	333 µm	< 5 mm; > 5mm	39356 9	9996 8	10698 46	72299 8	82875 7	231.6 5	74.5 3	629. 36	1165. 72	2335. 84	1.65	0.4 3	4.4 9	3.28	4.05
Suaria et al. (2016)	200 µm	< 5 mm; > 5mm	68000 0	3850 00	13325 00	12521 62	16176 51	134.8 4	41.5 6	399. 83	671.9 1	1544. 16	1.69	0.9 6	3.3 2	3.13	4.05
Compa et al. (2020)	335 µm	< 5 mm	NA	NA	NA	85802 9	NA	NA	NA	NA	4520. 0	NA	NA	N A	N A	NA	NA
Gündoğdu et al. (2018)	333 µm	< 5 mm	11541 73	2941 44	51216 55	41194 53	65198 66	NA	NA	NA	NA	NA	NA	N A	N A	NA	NA

Table S2.2. Literature data on plastic abundance in Mediterranean surface waters

Reference	Mesh Size	Size fraction	items·km-2					g·km-2					items·m-3				
			median	Q1	Q3	mean	sd	median	Q1	Q3	mean	sd	median	Q1	Q3	mean	sd
Collignon et al. (2012)	333 µm	< 5 mm	38200	23950	108500	106894	169814	NA	NA	NA	202	NA	NA	NA	NA	NA	NA
Cózar et al. (2015)	200 µm	< 5 mm	99642	80726	147362	126593	94208	224.1	75.1	540.0	531.5	765.1	NA	NA	NA	NA	NA
de Lucia et al. (2018)	333 µm	< 5 mm	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	0.38	0.15	0.49	0.39	0.26
de Lucia et al. (2014)	500 µm	< 5 mm	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Fossi et al. (2016)	200 µm	< 5 mm	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	0.31	1.17
Gündoğdu (2017)	333 µm	< 5 mm	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Kazour et al. (2019)	52 µm	< 5 mm	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	4.10	3.30	5.55	4.53	2.28
Panti et al. (2015)	200 µm	< 5 mm	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	0.07	0.05	0.14	0.17	0.32
Tanhua et al. (2020)	Inlet	< 5 mm; > 5mm	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	246.00	229.00	263.00	246.00	48.08

van der Hal et al. (2017)	333 µm	< 5 mm	NA	NA	NA	1518340	NA	NA	NA	NA	NA	NA	1.99	0.88	5.36	7.68	31.66
Wakkaf et al. (2020a)	Pump	< 5 mm	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	366.70	224.98	566.70	452.99	335.24
Zeri et al. (2018)	330 µm	< 5 mm	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA

References

Abidli, S., Antunes, J.C., Ferreira, J.L., Lahbib, Y., Sobral, P., Trigui El Menif, N., 2018. Microplastics in sediments from the littoral zone of the north Tunisian coast (Mediterranean Sea). Estuar. Coast. Shelf Sci. 205, 1–9. https://doi.org/10.1016/j.ecss.2018.03.006

Alomar, C., Estarellas, F., Deudero, S., 2016. Microplastics in the Mediterranean Sea: Deposition in coastal shallow sediments, spatial variation and preferential grain size. Mar. Environ. Res. 115, 1–10. https://doi.org/10.1016/j.marenvres.2016.01.005

Atwood, E.C., Falcieri, F.M., Piehl, S., Bochow, M., Matthies, M., Franke, J., Carniel, S., Sclavo, M., Laforsch, C., Siegert, F., 2019. Coastal accumulation of microplastic particles emitted from the Po River, Northern Italy: Comparing remote sensing and hydrodynamic modelling with in situ sample collections. Mar. Pollut. Bull. 138, 561–574. https://doi.org/10.1016/j.marpolbul.2018.11.045

Baini, M., Fossi, M.C., Galli, M., Caliani, I., Campani, T., Finoia, M.G., Panti, C., 2018. Abundance and characterization of microplastics in the coastal waters of Tuscany (Italy): The application of the MSFD monitoring protocol in the Mediterranean Sea. Mar. Pollut. Bull. 133, 543–552. https://doi.org/10.1016/j.marpolbul.2018.06.016

Bayo, J., Rojo, D., Olmos, S., 2019. Abundance, morphology and chemical composition of microplastics in sand and sediments from a protected coastal area: The Mar Menor lagoon (SE Spain). Environ. Pollut. 252, 1357–1366. https://doi.org/10.1016/j.envpol.2019.06.024

Blašković, A., Fastelli, P., Čižmek, H., Guerranti, C., Renzi, M., 2017. Plastic litter in sediments from the Croatian marine protected area of the natural park of TelašČica bay (Adriatic Sea). Mar. Pollut. Bull. 114, 583–586. https://doi.org/10.1016/j.marpolbul.2016.09.018

Blašković, A., Guerranti, C., Fastelli, P., Anselmi, S., Renzi, M., 2018. Plastic levels in sediments closed to Cecina river estuary (Tuscany, Italy). https://doi.org/10.1016/j.marpolbul.2018.07.021

Caldwell, J., Muff, L.F., Pham, C.K., Petri-Fink, A., Rothen-Rutishauser, B., Lehner, R., 2020. Spatial and temporal analysis of meso- and microplastic pollution in the Ligurian and Tyrrhenian Seas. Mar. Pollut. Bull. 159, 111515. https://doi.org/10.1016/J.MARPOLBUL.2020.111515

Caldwell, J., Petri-Fink, A., Rothen-Rutishauser, B., Lehner, R., 2019. Assessing meso- and microplastic pollution in the Ligurian and Tyrrhenian Seas. Mar. Pollut. Bull. 149, 110572. https://doi.org/10.1016/j.marpolbul.2019.110572

Camins, E., de Haan, W.P., Salvo, V.S., Canals, M., Raffard, A., Sanchez-Vidal, A., 2020. Paddle surfing for science on microplastic pollution. Sci. Total Environ. 709, 1–10. https://doi.org/10.1016/j.scitotenv.2019.136178

Cannas, S., Fastelli, P., Guerranti, C., Renzi, M., 2017. Plastic litter in sediments from the coasts of south Tuscany (Tyrrhenian Sea). https://doi.org/10.1016/j.marpolbul.2017.04.008

Ceccarini, A., Corti, A., Erba, F., Modugno, F., Nasa, L., Bianchi, S., Castelvetro, V., 2018. The Hidden Microplastics: New Insights and Figures from the Thorough Separation and Characterization of Microplastics and of Their Degradation Byproducts in Coastal Sediments. https://doi.org/10.1021/acs.est.8b01487

Chouchene, K., da Costa, J.P., Wali, A., Girão, A. V., Hentati, O., Duarte, A.C., Rocha-Santos, T., Ksibi, M., 2019. Microplastic pollution in the sediments of Sidi Mansour Harbor in Southeast Tunisia. Mar. Pollut. Bull. 146, 92–99. https://doi.org/10.1016/j.marpolbul.2019.06.004

Collignon, A., Hecq, J.H., Galgani, F., Collard, F., Goffart, A., 2014. Annual variation in neustonic micro- and meso-plastic particles and zooplankton in the Bay of Calvi (Mediterranean-Corsica). Mar. Pollut. Bull. 79, 293–298. https://doi.org/10.1016/j.marpolbul.2013.11.023

Collignon, A., Hecq, J.H., Glagani, F., Voisin, P., Collard, F., Goffart, A., 2012. Neustonic microplastic and zooplankton in the North Western Mediterranean Sea. Mar. Pollut. Bull. 64, 861–864. https://doi.org/10.1016/j.marpolbul.2012.01.011

Compa, M., Alomar, C., Mourre, B., March, D., Tintoré, J., Deudero, S., 2020. Nearshore spatio-temporal sea surface trawls of plastic debris in the Balearic Islands. Mar. Environ. Res. 158. https://doi.org/10.1016/j.marenvres.2020.104945

Constant, M., Kerhervé, P., Mino-Vercellio-Verollet, M., Dumontier, M., Sànchez Vidal, A., Canals, M., Heussner, S., 2019. Beached microplastics in the Northwestern Mediterranean Sea. Mar. Pollut. Bull. 142, 263–273. https://doi.org/10.1016/j.marpolbul.2019.03.032

Cózar, A., Sanz-Martín, M., Martí, E., González-Gordillo, J.I., Ubeda, B., Gálvez, J.Á., Irigoien, X., Duarte, C.M., 2015. Plastic Accumulation in the Mediterranean Sea. PLoS One 10, e0121762. https://doi.org/10.1371/journal.pone.0121762

D'Alessandro, M., Esposito, V., Porporato, E.M.D., Berto, D., Renzi, M., Giacobbe, S., Scotti, G., Consoli, P., Valastro, G., Andaloro, F., Romeo, T., 2018. Relationships between plastic litter and chemical pollutants on benthic biodiversity *. https://doi.org/10.1016/j.envpol.2018.08.002

de Haan, W.P., Sanchez-Vidal, A., Canals, M., 2019. Floating microplastics and aggregate formation in the Western Mediterranean Sea. Mar. Pollut. Bull. 140, 523–535. https://doi.org/10.1016/j.marpolbul.2019.01.053

de Lucia, G.A., Caliani, I., Marra, S., Camedda, A., Coppa, S., Alcaro, L., Campani, T., Giannetti, M., Coppola, D., Cicero, A.M., Panti, C., Baini, M., Guerranti, C., Marsili, L., Massaro, G., Fossi, M.C., Matiddi, M., 2014. Amount and distribution of neustonic micro-plastic off the western Sardinian coast (Central-Western Mediterranean Sea). Mar. Environ. Res. 100, 10–16. https://doi.org/10.1016/j.marenvres.2014.03.017

de Lucia, G.A., Vianello, A., Camedda, A., Vani, D., Tomassetti, P., Coppa, S., Palazzo, L., Amici, M., Romanelli, G., Zampetti, G., Cicero, A.M., Carpentieri, S., Di Vito, S., Matiddi, M., 2018. Sea water contamination in the Vicinity of the Italian minor islands caused by microplastic pollution. Water (Switzerland) 10, 1108. https://doi.org/10.3390/w10081108

De Ruijter, V.N., Milou, A., Costa, V., 2019. Assessment of microplastics distribution and stratification in the shallow marine sediments of Samos island, Eastern Mediterranean sea, Greece. Mediterr. Mar. Sci. 20, 736–744. https://doi.org/10.12681/mms.19131

Duncan, E.M., Arrowsmith, J., Bain, C., Broderick, A.C., Lee, J., Metcalfe, K., Pikesley, S.K., Snape, R.T.E., Van Sebille, E., Godley, B.J., 2018. The true depth of the Mediterranean plastic problem: Extreme microplastic pollution on marine turtle nesting beaches in Cyprus. https://doi.org/10.1016/j.marpolbul.2018.09.019

Fanini, L., Bozzeda, F., 2018. Dynamics of plastic resin pellets deposition on a microtidal sandy beach: Informative variables and potential integration into sandy beach studies. Ecol. Indic. 89, 309–316. https://doi.org/10.1016/j.ecolind.2018.02.027

Fastelli, P., Blašković, A., Bernardi, G., Romeo, T., Čižmek, H., Andaloro, F., Russo, G.F., Guerranti, C., Renzi, M., 2016. Plastic litter in sediments from a marine area likely to become protected (Aeolian Archipelago's islands, Tyrrhenian sea). https://doi.org/10.1016/j.marpolbul.2016.08.054

Faure, F., Saini, C., Potter, G., Galgani, F., de Alencastro, L.F., Hagmann, P., 2015. An evaluation of surface micro- and mesoplastic pollution in pelagic ecosystems of the Western Mediterranean Sea. Environ. Sci. Pollut. Res. 22, 12190–12197. https://doi.org/10.1007/s11356-015-4453-3

Filgueiras, A.V., Gago, J., Campillo, J.A., León, V.M., 2019. Microplastic distribution in surface sediments along the Spanish Mediterranean continental shelf. Environ. Sci. Pollut. Res. 26, 21264–21273. https://doi.org/10.1007/s11356-019-05341-5

Fossi, M.C., Marsili, L., Baini, M., Giannetti, M., Coppola, D., Guerranti, C., Caliani, I., Minutoli, R., Lauriano, G., Finoia, M.G., Rubegni, F., Panigada, S., Bérubé, M., Urbán Ramírez, J., Panti, C., 2016. Fin whales and microplastics: The Mediterranean Sea and the Sea of Cortez scenarios. Environ. Pollut. 209, 68–78. https://doi.org/10.1016/j.envpol.2015.11.022

Fossi, M.C., Panti, C., Guerranti, C., Coppola, D., Giannetti, M., Marsili, L., Minutoli, R., 2012. Are baleen whales exposed to the threat of microplastics? A case study of the Mediterranean fin whale (Balaenoptera physalus). Mar. Pollut. Bull. 64, 2374–2379. https://doi.org/10.1016/j.marpolbul.2012.08.013

Fossi, M.C., Romeo, T., Baini, M., Panti, C., Marsili, L., Campan, T., Canese, S., Galgani, F., Druon, J.N., Airoldi, S., Taddei, S., Fattorini, M., Brandini, C., Lapucci, C., 2017. Plastic debris occurrence, convergence areas and fin whales feeding ground in the Mediterranean marine protected area Pelagos Sanctuary: A modeling approach. Front. Mar. Sci. 4, 1–15. https://doi.org/10.3389/fmars.2017.00167

Gajšt, T., Bizjak, T., Palatinus, A., Liubartseva, S., Kržan, A., 2016. Sea surface microplastics in Slovenian part of the Northern Adriatic. Mar. Pollut. Bull. 113, 392–399. https://doi.org/10.1016/j.marpolbul.2016.10.031

Godoy, V., Prata, J.C., Blázquez, G., Almendros, A.I., Duarte, A.C., Rocha-Santos, T., Calero, M., Martín-Lara, M.Á., 2020. Effects of distance to the sea and geomorphological characteristics on the quantity and distribution of microplastics in beach sediments of Granada (Spain). Sci. Total Environ. 746, 142023. https://doi.org/10.1016/J.SCITOTENV.2020.142023

Guerranti, C., Cannas, S., Scopetani, C., Fastelli, P., Cincinelli, A., Renzi, M., 2017. Plastic litter in aquatic environments of Maremma Regional Park (Tyrrhenian Sea, Italy): Contribution by the Ombrone river and levels in marine sediments. Mar. Pollut. Bull. 117, 366–370. https://doi.org/10.1016/j.marpolbul.2017.02.021

Gündoğdu, S., 2017. High level of micro-plastic pollution in the Iskenderun Bay NE Levantine coast of Turkey. Ege J. Fish. Aquat. Sci. 34, 401–408. https://doi.org/10.12714/egejfas.2017.34.4.06

Gündoğdu, S., Çevik, C., 2017. Micro- and mesoplastics in Northeast Levantine coast of Turkey: The preliminary results from surface samples. Mar. Pollut. Bull. 118, 341–347. https://doi.org/10.1016/j.marpolbul.2017.03.002

Gündoğdu, S., Çevik, C., Ayat, B., Aydoğan, B., Karaca, S., 2018. How microplastics quantities increase with flood events? An example from Mersin Bay NE Levantine coast of Turkey. Environ. Pollut. 239, 342–

350. https://doi.org/10.1016/j.envpol.2018.04.042

Güven, O., Gökdağ, K., Jovanović, B., Kıdeyş, A.E., 2017. Microplastic litter composition of the Turkish territorial waters of the Mediterranean Sea, and its occurrence in the gastrointestinal tract of fish. Environ. Pollut. 223, 286–294. https://doi.org/10.1016/j.envpol.2017.01.025

Kane, I.A., Clare, M.A., Miramontes, E., Wogelius, R., Rothwell, J.J., Garreau, P., Pohl, F., 2020. Seafloor microplastic hotspots controlled by deep-sea circulation. Science (80-.). eaba5899. https://doi.org/10.1126/science.aba5899

Karkanorachaki, K., Kiparissis, S., Kalogerakis, G.C., Yiantzi, E., Psillakis, E., Kalogerakis, N., 2018. Plastic pellets, meso- and microplastics on the coastline of Northern Crete: Distribution and organic pollution. Mar. Pollut. Bull. 133, 578–589. https://doi.org/10.1016/j.marpolbul.2018.06.011

Kazour, M., Jemaa, S., Issa, C., Khalaf, G., Amara, R., 2019. Microplastics pollution along the Lebanese coast (Eastern Mediterranean Basin): Occurrence in surface water, sediments and biota samples. Sci. Total Environ. 696. https://doi.org/10.1016/j.scitotenv.2019.133933

Korez, Š., Gutow, L., Saborowski, R., 2019. Microplastics at the strandlines of Slovenian beaches. Mar. Pollut. Bull. 145, 334–342. https://doi.org/10.1016/j.marpolbul.2019.05.054

Krüger, L., Casado-Coy, N., Valle, C., Ramos, M., S Anchez-Jerez, P., Gago, J., Carretero, O., Beltran-Sanahuja, A., Sanz-Lazaro, C., 2019. Plastic debris accumulation in the seabed derived from coastal fish farming *. https://doi.org/10.1016/j.envpol.2019.113336

Laglbauer, B.J.L., Franco-Santos, R.M., Andreu-Cazenave, M., Brunelli, L., Papadatou, M., Palatinus, A., Grego, M., Deprez, T., 2014. Macrodebris and microplastics from beaches in Slovenia. Mar. Pollut. Bull. 89, 356–366. https://doi.org/10.1016/j.marpolbul.2014.09.036

Lefebvre, C., Saraux, C., Heitz, O., Nowaczyk, A., Bonnet, D., 2019. Microplastics FTIR characterisation and distribution in the water column and digestive tracts of small pelagic fish in the Gulf of Lions. Mar. Pollut. Bull. 142, 510–519. https://doi.org/10.1016/j.marpolbul.2019.03.025

Lots, F.A.E., Behrens, P., Vijver, M.G., Horton, A.A., Bosker, T., 2017. A large-scale investigation of microplastic contamination: Abundance and characteristics of microplastics in European beach sediment. Mar. Pollut. Bull. 123, 219–226. https://doi.org/10.1016/j.marpolbul.2017.08.057

Maršić-Lučić, J., Lušić, J., Tutman, P., Bojanić Varezić, D., Šiljić, J., Pribudić, J., 2018. Levels of trace metals on microplastic particles in beach sediments of the island of Vis, Adriatic Sea, Croatia. Mar. Pollut. Bull.

137, 231–236. https://doi.org/10.1016/j.marpolbul.2018.10.027

Merlino, S., Locritani, M., Bernardi, G., Como, C., Legnaioli, S., Palleschi, V., Abbate, M., 2020. Spatial and Temporal Distribution of Chemically Characterized Microplastics within the Protected Area of Pelagos Sanctuary (NW Mediterranean Sea): Focus on Natural and Urban Beaches. Water 2020, Vol. 12, Page 3389 12, 3389. https://doi.org/10.3390/W12123389

Misic, C., Covazzi Harriague, A., Ferrari, M., 2019. Hydrodynamic forcing and sand permeability influence the distribution of anthropogenic microparticles in beach sediment. Estuar. Coast. Shelf Sci. 230. https://doi.org/10.1016/j.ecss.2019.106429

Missawi, O., Bousserrhine, N., Belbekhouche, S., Zitouni, N., Alphonse, V., Boughattas, I., Banni, M., 2020. Abundance and distribution of small microplastics (≤ 3 μm) in sediments and seaworms from the Southern Mediterranean coasts and characterisation of their potential harmful effects. Environ. Pollut. 263, 114634. https://doi.org/10.1016/J.ENVPOL.2020.114634

Mistri, M., Infantini, V., Scoponi, M., Granata, T., Moruzzi, L., Massara, F., De Donati, M., Munari, C., 2018. Microplastics in marine sediments in the area of Pianosa Island (Central Adriatic Sea). Rend. Lincei 29, 805–809. https://doi.org/10.1007/s12210-018-0736-1

Mistri, M., Scoponi, M., Granata, T., Moruzzi, L., Massara, F., Munari, C., 2020. Types, occurrence and distribution of microplastics in sediments from the northern Tyrrhenian Sea. Mar. Pollut. Bull. 153. https://doi.org/10.1016/j.marpolbul.2020.111016

Munari, C., Scoponi, M., Mistri, M., 2017. Plastic debris in the Mediterranean Sea: Types, occurrence and distribution along Adriatic shorelines. Waste Manag. 67, 385–391. https://doi.org/10.1016/j.wasman.2017.05.020

Palatinus, A., Kovač Viršek, M., Robič, U., Grego, M., Bajt, O., Šiljić, J., Suaria, G., Liubartseva, S., Coppini, G., Peterlin, M., 2019. Marine litter in the Croatian part of the middle Adriatic Sea: Simultaneous assessment of floating and seabed macro and micro litter abundance and composition. Mar. Pollut. Bull. 139, 427–439. https://doi.org/10.1016/j.marpolbul.2018.12.038

Panti, C., Giannetti, M., Baini, M., Rubegni, F., Minutoli, R., Fossi, M.C., 2015. Occurrence, relative abundance and spatial distribution of microplastics and zooplankton NW of Sardinia in the Pelagos Sanctuary Protected Area, Mediterranean Sea. Environ. Chem. 12, 618–626. https://doi.org/10.1071/EN14234

Pedrotti, M.L., Petit, S., Elineau, A., Bruzaud, S., Crebassa, J.-C., Dumontet,

B., Martí, E., Gorsky, G., Cózar, A., 2016. Changes in the Floating Plastic Pollution of the Mediterranean Sea in Relation to the Distance to Land. PLoS One 11, e0161581. https://doi.org/10.1371/journal.pone.0161581

Piazzolla, D., Cafaro, V., de Lucia, G.A., Mancini, E., Scanu, S., Bonamano, S., Piermattei, V., Vianello, A., Della Ventura, G., Marcelli, M., 2020. Microlitter pollution in coastal sediments of the northern Tyrrhenian Sea, Italy: microplastics and fly-ash occurrence and distribution. Estuar. Coast. Shelf Sci. 241, 106819. https://doi.org/10.1016/J.ECSS.2020.106819

Piehl, S., Mitterwallner, V., Atwood, E.C., Bochow, M., Laforsch, C., 2019. Abundance and distribution of large microplastics (1-5 mm) within beach sediments at the Po River Delta, northeast Italy. https://doi.org/10.1016/j.marpolbul.2019.110515

Piperagkas, O., Papageorgiou, N., Karakassis, I., 2019. Qualitative and quantitative assessment of microplastics in three sandy Mediterranean beaches, including different methodological approaches. https://doi.org/10.1016/j.ecss.2019.02.016

Renzi, M., Blašković, A., 2020. Chemical fingerprint of plastic litter in sediments and holothurians from Croatia: Assessment & relation to different environmental factors. https://doi.org/10.1016/j.marpolbul.2020.110994

Renzi, M., Blašković, A., Broccoli, A., Bernardi, G., Grazioli, E., Russo, G., 2020. Chemical composition of microplastic in sediments and protected detritivores from different marine habitats (Salina Island). Mar. Pollut. Bull. 152, 110918. https://doi.org/10.1016/J.MARPOLBUL.2020.110918

Renzi, M., Blašković, A., Fastelli, P., Marcelli, M., Guerranti, C., Cannas, S., Barone, L., Massara, F., 2018. Is the microplastic selective according to the habitat? Records in amphioxus sands, Mäerl bed habitats and Cymodocea nodosa habitats. https://doi.org/10.1016/j.marpolbul.2018.03.019

Ruiz-Orejón, L.F., Mourre, B., Sardá, R., Tintoré, J., Ramis-Pujol, J., 2019. Quarterly variability of floating plastic debris in the marine protected area of the Menorca Channel (Spain). Environ. Pollut. 252, 1742–1754. https://doi.org/10.1016/j.envpol.2019.06.063

Ruiz-Orejón, L.F., Sardá, R., Ramis-Pujol, J., 2018. Now, you see me: High concentrations of floating plastic debris in the coastal waters of the Balearic Islands (Spain). Mar. Pollut. Bull. 133, 636–646. https://doi.org/10.1016/j.marpolbul.2018.06.010

Ruiz-Orejón, L.F., Sardá, R., Ramis-Pujol, J., 2016. Floating plastic debris in the Central and Western Mediterranean Sea. Mar. Environ. Res. 120, 136–

144. https://doi.org/10.1016/j.marenvres.2016.08.001

Sanchez-Vidal, A., Thompson, R.C., Canals, M., De Haan, W.P., 2018. The imprint of microfibres in southern European deep seas. https://doi.org/10.1371/journal.pone.0207033

Schmidt, N., Thibault, D., Galgani, F., Paluselli, A., Sempéré, R., 2018. Occurrence of microplastics in surface waters of the Gulf of Lion (NW Mediterranean Sea). Prog. Oceanogr. 163, 214–220. https://doi.org/10.1016/j.pocean.2017.11.010

Shabaka, S.H., Ghobashy, M., Marey, R.S., 2019. Identification of marine microplastics in Eastern Harbor, Mediterranean Coast of Egypt, using differential scanning calorimetry. Mar. Pollut. Bull. 142, 494–503. https://doi.org/10.1016/j.marpolbul.2019.03.062

Simon-Sánchez, L., Grelaud, M., Garcia-Orellana, J., Ziveri, P., 2019. River Deltas as hotspots of microplastic accumulation: The case study of the Ebro River (NW Mediterranean). Sci. Total Environ. 687, 1186–1196. https://doi.org/10.1016/J.SCITOTENV.2019.06.168

Suaria, G., Avio, C.G., Mineo, A., Lattin, G.L., Magaldi, M.G., Belmonte, G., Moore, C.J., Regoli, F., Aliani, S., 2016. The Mediterranean Plastic Soup: Synthetic polymers in Mediterranean surface waters. Sci. Rep. 6. https://doi.org/10.1038/srep37551

Tanhua, T., Gutekunst, S.B., Biastoch, A., 2020. A near-synoptic survey of ocean microplastic concentration along an around-the-world sailing race. PLoS One 15, e0243203. https://doi.org/10.1371/JOURNAL.PONE.0243203

Tata, T., Belabed, B.E., Bououdina, M., Bellucci, S., 2020. Occurrence and characterization of surface sediment microplastics and litter from North African coasts of Mediterranean Sea: Preliminary research and first evidence. Sci. Total Environ. 713. https://doi.org/10.1016/j.scitotenv.2020.136664

Tunçer, S., Artüz, O.B., Demirkol, M., Artüz, M.L., 2018. First report of occurrence, distribution, and composition of microplastics in surface waters of the Sea of Marmara, Turkey. Mar. Pollut. Bull. 135, 283–289. https://doi.org/10.1016/j.marpolbul.2018.06.054

Turner, A., Holmes, L., 2011. Occurrence, distribution and characteristics of beached plastic production pellets on the island of Malta (central Mediterranean). Mar. Pollut. Bull. 62, 377–381. https://doi.org/10.1016/j.marpolbul.2010.09.027

Tziourrou, P., Megalovasilis, P., Tsounia, M., Karapanagioti, H.K., 2019. Characteristics of microplastics on two beaches affected by different land uses in Salamina Island in Saronikos Gulf, east Mediterranean. Mar.

Pollut. Bull. 149. https://doi.org/10.1016/j.marpolbul.2019.110531

Van Cauwenberghe, L., Vanreusel, A., Mees, J., Janssen, C.R., 2013. Microplastic pollution in deep-sea sediments. Environ. Pollut. 182, 495–499. https://doi.org/10.1016/J.ENVPOL.2013.08.013

van der Hal, N., Ariel, A., Angel, D.L., 2017. Exceptionally high abundances of microplastics in the oligotrophic Israeli Mediterranean coastal waters. Mar. Pollut. Bull. 116, 151–155. https://doi.org/10.1016/j.marpolbul.2016.12.052

Vianello, A., Boldrin, A., Guerriero, P., Moschino, V., Rella, R., Sturaro, A., Da Ros, L., 2013. Microplastic particles in sediments of Lagoon of Venice, Italy: First observations on occurrence, spatial patterns and identification. Estuar. Coast. Shelf Sci. 130, 54–61. https://doi.org/10.1016/j.ecss.2013.03.022

Vianello, A., Da Ros, L., Boldrin, A., Marceta, T., Moschino, V., 2018. First evaluation of floating microplastics in the Northwestern Adriatic Sea. Environ. Sci. Pollut. Res. 25, 28546–28561. https://doi.org/10.1007/s11356-018-2812-6

Wakkaf, T., El Zrelli, R., Kedzierski, M., Balti, R., Shaiek, M., Mansour, L., Tlig-Zouari, S., Bruzaud, S., Rabaoui, L., 2020a. Characterization of microplastics in the surface waters of an urban lagoon (Bizerte lagoon, Southern Mediterranean Sea): Composition, density, distribution, and influence of environmental factors. Mar. Pollut. Bull. 160, 111625. https://doi.org/10.1016/J.MARPOLBUL.2020.111625

Wakkaf, T., El Zrelli, R., Kedzierski, M., Balti, R., Shaiek, M., Mansour, L., Tlig-Zouari, S., Bruzaud, S., Rabaoui, L., 2020b. Microplastics in edible mussels from a southern Mediterranean lagoon: Preliminary results on seawater-mussel transfer and implications for environmental protection and seafood safety. Mar. Pollut. Bull. 158, 111355. https://doi.org/10.1016/J.MARPOLBUL.2020.111355

Woodall, L.C., Sanchez-Vidal, A., Canals, M., Paterson, G.L.J., Coppock, R., Sleight, V., Calafat, A., Rogers, A.D., Narayanaswamy, B.E., Thompson, R.C., 2014. The deep sea is a major sink for microplastic debris. R. Soc. Open Sci. 1. https://doi.org/10.1098/rsos.140317

Yabanlı, M., Yozukmaz, A., Şener, İ., Ölmez, Ö.T., 2019. Microplastic pollution at the intersection of the Aegean and Mediterranean Seas: A study of the Datça Peninsula (Turkey). Mar. Pollut. Bull. 145, 47–55. https://doi.org/10.1016/j.marpolbul.2019.05.003

Zayen, A., Sayadi, S., Chevalier, C., Boukthir, M., Ben Ismail, S., Tedetti, M., 2020. Microplastics in surface waters of the Gulf of Gabes, southern Mediterranean Sea: Distribution, composition and influence of

hydrodynamics. Estuar. Coast. Shelf Sci. 242, 106832. https://doi.org/10.1016/j.ecss.2020.106832

Zeri, C., Adamopoulou, A., Bojanić Varezić, D., Fortibuoni, T., Kovač Viršek, M., Kržan, A., Mandic, M., Mazziotti, C., Palatinus, A., Peterlin, M., Prvan, M., Ronchi, F., Siljic, J., Tutman, P., Vlachogianni, T., 2018. Floating plastics in Adriatic waters (Mediterranean Sea): From the macro- to the micro-scale. Mar. Pollut. Bull. 136, 341–350. https://doi.org/10.1016/j.marpolbul.2018.09.016

Supplementary material - Chapter 3

Table S3. 1. Summary of the beaches monitored for micro-litter occurrence classified by type of beach considered.

Fig S3. 1. Clean laboratory for microplastic analysis at Institute of Environmental Sciences and Technology- Autonomous University of Barcelona, and example of the coverall wore during sample preparation.

Fig S3. 2. Spectra of a red fragment showing fluorescence. The spectra was gathered with a Renishaw InVia instrument with a Leica microscope with 50× magnification len and a 785 nm diode laser, in the range of 2000 to 500 cm^{-1}, using a laser power of 5%, 10 s exposure time and four accumulations.

Table S3.2. Microplastic concentrations reported in beaches of the Mediterranean Sea. Data used to produce the map in Figure 3.8.

Table S3. 1. Summary of the beaches monitored for micro-litter occurrence classified by type of beach considered.

Island	Type of beach	Name of the beach	Latitude (ºE)	Longitude(ºN)
Crete	Touristic	Rethymno	35.3644	24.4822
	Local	Arina	35.33029	25.23613
	Remote	Tsoutsouras	34.98632	25.29227
Cyprus	Touristic	Sunrise	35.015908	34.054436
	Local	Faros	34.820764	33.604883
	Remote	Timi	34.709831	32.488094
Mallorca	Touristic	Torà	39.534889	2.457083
	Local	Es Caragol	39.276795	3.044037
	Remote	Sa Canova	39.729399	3.250852
Malta	Touristic	Golden Bay	35.934233	14.344583
	Local	Gnejna Bay	35.920217	14.343133
	Remote	Marsaxlokk	35.839017	14.548483
Mykonos	Touristic	Platis Gyalos	37.41411	25.34287
	Local	Fokos	37.48112	25.4101
	Remote	Merchia	37.47179	25.42839
Rhodes	Touristic	Tsambika	36.22914	28.14826
	Local	Afandou	36.28656	28.17663
	Remote	Gennadi	36.02011	27.92852
Sicily	Touristic	Naxos	37.825872	15.270125
	Local	Letojanni	37.884744	15.313469
	Remote	Fondaco Parrino	37.906536	15.337061

Fig. S3. 1. Clean laboratory for microplastic analysis at Institute of Environmental Sciences and Technology- Autonomous University of Barcelona, and example of the coverall wore during sample preparation.

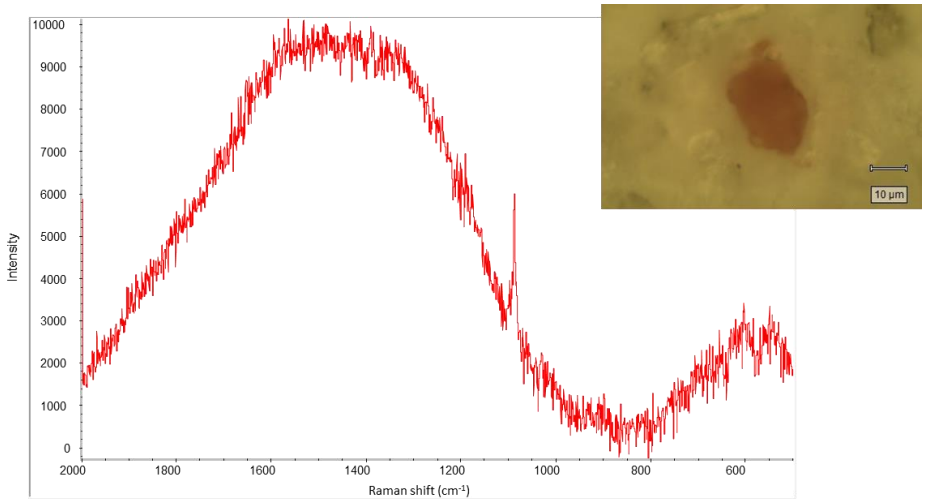


Fig. S3. 2. Spectra of a red fragment showing fluorescence. The spectra was gathered with a Renishaw InVia instrument with a Leica microscope with 50× magnification len and a 785 nm diode laser, in the range of 2000 to 500 cm^{-1}, using a laser power of 5%, 10 s exposure time and four accumulations.

Table S3.2. Microplastic concentrations reported in beaches of the Mediterranean Sea. Data used to produce the map in Figure 3.8.

Reference	Country	Station	Sampling year	LAT (N)	LONG (E)	MP	Units
Abidli et al. 2018	Tunisia	Carthage	2017	36.863	10.341	14 1	MP/kg
Abidli et al. 2018	Tunisia	Menzel Bourguiba	2017	37.172	9.795	46 1	MP/kg
Atwood et al. 2019	Italy	Caleri	2016	45.091	12.332	79	MP/kg
Atwood et al. 2019	Italy	Levante	2016	45.055	12.383	59	MP/kg
Atwood et al. 2019	Italy	Boccasette	2016	45.018	12.438	4	MP/kg
Atwood et al. 2019	Italy	Pila North 1	2016	44.990	12.514	2	MP/kg
Atwood et al. 2019	Italy	Pila North 2	2016	44.978	12.536	4	MP/kg
Atwood et al. 2019	Italy	Pila South	2016	44.953	12.536	8	MP/kg
Atwood et al. 2019	Italy	Allagamento	2016	44.873	12.482	1	MP/kg
Atwood et al. 2019	Italy	Barricata	2016	44.853	12.471	14	MP/kg
Atwood et al. 2019	Italy	Goro	2016	44.791	12.397	5	MP/kg
Bayo et al. 2019	Spain	Los Urrutias	NA	37.679	-0.827	16 6	MP/kg
Bayo et al. 2019	Spain	Marina del Carmolí Litoral Norte	NA	37.713	-0.857	8	MP/kg
Blaškovic et al. 2018	Italy	Free access beach-S2	2017	43.292	10.499	72	MP/kg
Blaškovic et al. 2018	Italy	Artificial Dam	2017	43.300	10.490	19 1	MP/kg
Cannas et al. 2017	Italy	Talamone	NA	42.557	11.136	62	MP/kg
Cannas et al. 2017	Italy	Osa	NA	42.550	11.169	28 6	MP/kg
Cannas et al. 2017	Italy	Albegna	NA	42.502	11.192	45 3	MP/kg
Cannas et al. 2017	Italy	Capalbio	NA	42.397	11.371	46 6	MP/kg
Constant et al. 2019	France	La Crouste	2016	42.717	3.040	18 2	MP/kg
Constant et al. 2019	France	Fourat Beach	2016	42.500	3.129	60	MP/kg
Constant et al. 2019	France	La Crouste	2016	42.717	3.040	15 0	MP/kg
Constant et al. 2019	France	Fourat Beach	2016	42.500	3.129	56	MP/kg
Korez et al. 2019	Slovenia	Bele skale	2017	45.534	13.628	0	MP/kg
Korez et al. 2019	Slovenia	Simonov zaliv	2017	45.532	13.644	0	MP/kg
Korez et al. 2019	Slovenia	Izola	2017	45.542	13.660	0	MP/kg
Korez et al. 2019	Slovenia	Ankaran	2017	45.570	13.743	0	MP/kg
Korez et al. 2019	Slovenia	Debeli rti?	2017	45.588	13.708	0	MP/kg
Korez et al. 2019	Slovenia	Se?a	2017	45.500	13.588	82	g

Korez et al. 2019	Slovenia	Portorož		2017	45.513	13.594	0	MP/kg
Korez et al. 2019	Slovenia	Koper		2017	45.547	13.708	0	MP/kg
Korez et al. 2019	Slovenia	Ankaran		2017	45.570	13.743	0	MP/kg
Laglbauer et al. 2014	Slovenia	Debeli Rti?		2012	45.590	13.704	44	MP/kg
Lots et al. 2017	Italy	Sicily	NA		36.760	15.100	16	MP/kg
Lots et al. 2017	Italy	Lido di Dante	NA		44.380	12.320	15	MP/kg
Lots et al. 2017	Italy	San Mauro	NA		44.170	12.440	12	MP/kg
Lots et al. 2017	Spain	Denia	NA		38.840	0.110	84	MP/kg
Lots et al. 2017	Spain	Barcelona	NA		41.400	2.210	15	MP/kg
Lots et al. 2017	France	Cassis	NA		43.210	5.540	14	MP/kg
Lots et al. 2017	Greece	Pilion	NA		39.440	23.050	12	MP/kg
Lots et al. 2017	Turkey	Dikili	NA		39.070	26.890	23	MP/kg
Lots et al. 2017	Israel	Tel Aviv	NA		32.110	34.860	24	MP/kg
Lots et al. 2017	Bosnia	Bosnia	NA		42.920	17.620	8	MP/kg
Misic et al. 2019	Italy	Levanto		2017	44.170	9.612	76	MP/kg
Piperagkas et al. 2019	Creete, Greece	Pacheia Ammos beach		2015	35.110	25.807	41	MP/kg
Piperagkas et al. 2019	Creete, Greece	Analoukas beach		2015	35.212	26.186	6	MP/kg
Piperagkas et al. 2019	Creete, Greece	Sitia Bay		2015	35.205	26.109	10	MP/kg
Piperagkas et al. 2019	Creete, Greece	Pacheia Ammos beach		2016	35.110	25.807	16	MP/kg
Piperagkas et al. 2019	Creete, Greece	Analoukas beach		2016	35.212	26.186	23	MP/kg
Piperagkas et al. 2019	Creete, Greece	Sitia Bay		2016	35.205	26.109	13	MP/kg
Shabaka et al. 2019	Egypt	Alexandria City		2017	31.203	29.888	6	MP/kg
Simon-Sánchez et al. 2019	Spain	Riumar		2017	40.736	0.863	24	MP/kg
Simon-Sánchez et al. 2019	Spain	Riumar		2018	40.731	0.842	49	MP/kg
Simon-Sánchez et al. 2019	Spain	Marquesa		2016	40.734	0.829	31	MP/kg
Simon-Sánchez et al. 2019	Spain	Marquesa		2017	40.762	0.798	28	MP/kg
Simon-Sánchez et al. 2019	Spain	Cala Moros		2017	40.845	0.751	46	MP/kg
Tata et al. 2020	Algeria	Chapuis Beach		2018	36.919	7.754	55	MP/kg
Tata et al. 2020	Algeria	Joannonville Beach		2018	36.871	7.769	18	MP/kg
Tata et al. 2020	Algeria	Sidi Salem Beach		2018	36.862	7.782	64	MP/kg
Tata et al. 2020	Algeria	Cap Rosa Beach		2018	36.935	8.242	58	MP/kg

Yabanli et al. 2019	Turkey	Aktur Beach	2018	36.755	27.889	22 18	MP/kg
Yabanli et al. 2019	Turkey	Ovabükü Beach	2018	36.681	27.558	13 28	MP/kg
Yabanli et al. 2019	Turkey	Surf Camping	2018	36.763	27.734	62 3	MP/kg
Yabanli et al. 2019	Turkey	Kurucabük Beach	2018	36.756	27.908	59 3	MP/kg
De Ruitjer et al. 2019	Samos Island	Psili Ammos	2017	37.708	27.018	21	MP/kg
Piehl et al. 2019	Italy	Lev-BSH	2016	43.040	16.231	12	MP/kg
This study	Rhodes	Afandou	2017	36.287	28.177	92	items/kg
This study	Crete	Arina	2017	35.330	25.236	22 7	items/kg
This study	Mallorca	Es Caragol	2017	39.277	3.044	96	items/kg
This study	Cyprus	Faros	2017	34.821	33.605	13 3	items/kg
This study	Mykonos	Fokos	2017	37.481	25.410	87	items/kg
This study	Sicily	Fondaco Parrino	2017	37.907	15.337	71	items/kg
This study	Rhodes	Gennadi	2017	36.020	27.929	66	items/kg
This study	Malta	Gnejna Bay	2017	35.920	14.343	10 4	items/kg
This study	Malta	Golden Bay	2017	35.934	14.345	47 2	items/kg
This study	Sicily	Letojanni	2017	37.885	15.313	13 2	items/kg
This study	Malta	Marsaxlokk	2017	35.839	14.548	13 4	items/kg
This study	Mykonos	Merchia	2017	37.472	25.428	59	items/kg
This study	Sicily	Naxos	2017	37.826	15.270	34 5	items/kg
This study	Mykonos	Platis gyalos	2017	37.414	25.343	38 5	items/kg
This study	Crete	Rethymno	2017	35.364	24.482	15 6	items/kg
This study	Mallorca	Sa Canova	2017	39.729	3.251	11 0	items/kg
This study	Cyprus	Sunrise	2017	35.016	34.054	67 7	items/kg
This study	Cyprus	Timi	2017	34.710	32.488	11 6	items/kg
This study	Mallorca	Torà	2017	39.535	2.457	33 0	items/kg
This study	Rhodes	Tsambika	2017	36.229	28.148	39 9	items/kg
This study	Crete	Tsoutsouras	2017	34.986	25.292	48	items/kg

Supplementary material - Chapter 4

Table S4.1. Date and geographical data of sampling

Fig S4. 1. Distribution of size and colour of the overall of fibres, fragments and films found in the different environments sampled at the Ebro Delta: river surface water, river bed sediments and sandy beaches.

Table S4. 1. Date and geographical data of sampling

Date	Environmental matrix	Sample	Coordinates		Sampling approach
			Latitude	Longitude	
24/02/2017	Surface water	WS1a	40,713764 N	0,716859 E	Neuston net: 15cm of diameter and 5µm mesh size
24/02/2017	Surface water	WS1b	40,713764 N	0,716859 E	
24/02/2017	Surface water	WS2	40,716047 N	0,841240 E	
24/02/2017	Beach	S1	40,735570 N	0,863472 E	Last high-water mark: quadrat of 20x20 cm and scraping the first 2.5 cm
24/02/2017	Beach	S2	40,730734 N	0,841889 E	
24/02/2017	Beach	S3	40,733619 N	0,828831 E	
24/02/2017	Beach	S4	40,761865 N	0,797965 E	
24/02/2017	Beach	S5	40,845452 N	0,751270 E	
10/03/2017	River bed sediments	GS1	40,719603 N	0,862288 E	Van veen grab sampler
10/03/2017	River bed sediments	GS2	40,694963 N	0,796825 E	
10/03/2017	River bed sediments	GS3	40,703477 N	0,761786 E	

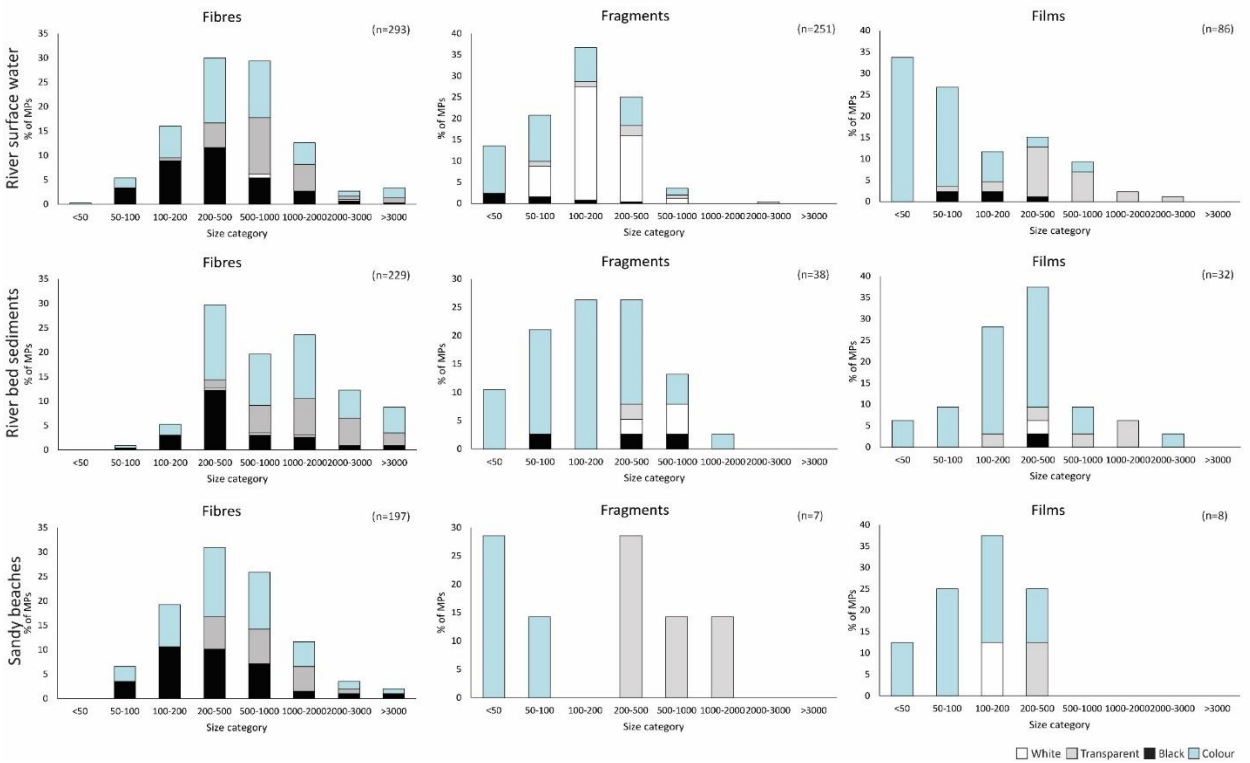


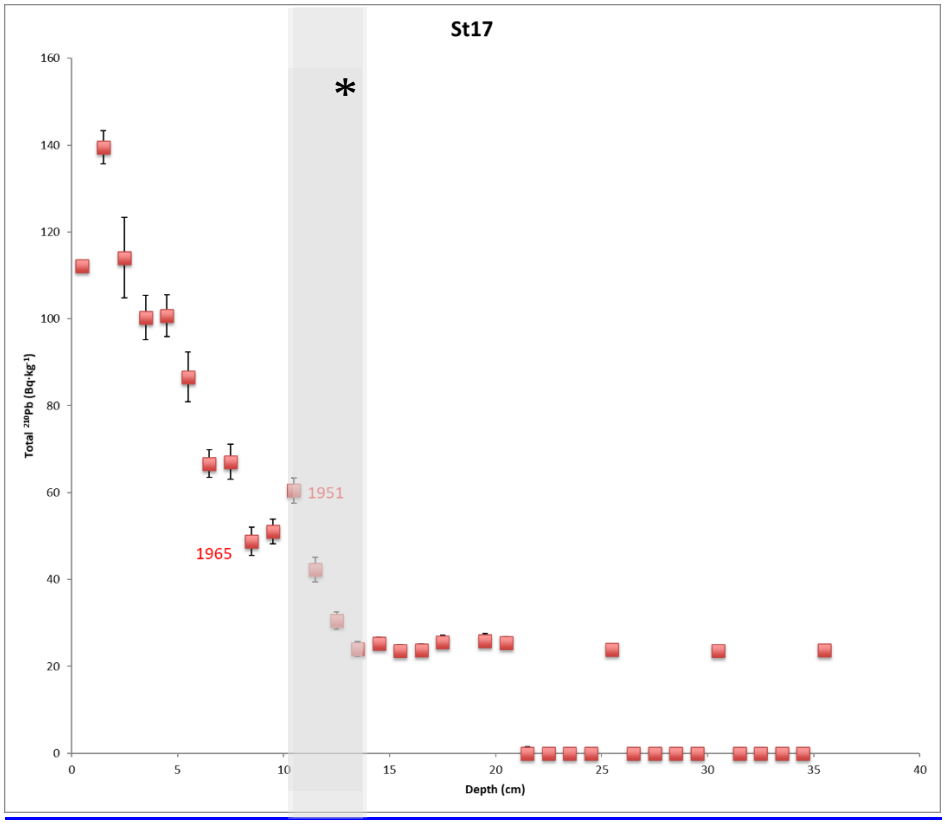
Fig. S4. 1. Distribution of size and colour of the overall of fibres, fragments and films found in the different environments sampled at the Ebro Delta: river surface water, river bed sediments and sandy beaches.

Supplementary material - Chapter 5

Fig. S5.1 $^{210}Pb_{xs}$ specific activity profile of the sediment core MERS_BI_ST17_MUC2. In grey the anomaly on the exponential trend characterizing the sediment accumulation in steady-state conditions. There, it can be observed a change in the sedimentation rate, most likely due to the construction of the Mequinenza and Ribaroja dams.

Fig S5.2. Ebro river discharge records from 1951 to 2016 recorded at the stream gauge of Tortosa. In red the decrease in the annual flow of the Ebro River. In grey the period of the dams' construction (1958-1966).

Table S5. 1- Microplastic data- Note that the absolute number of the blanks do not always correspond to the number of particles subtracted because the blank correction was applied per polymer and size class.



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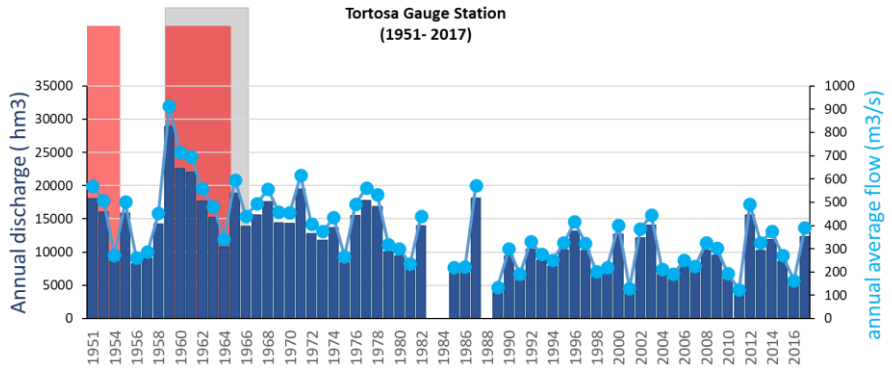

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Table S5. 1- Microplastic data- Note that the absolute number of the blanks do not always correspond to the number of particles subtracted because the blank correction was applied per polymer and size class.

Sample	Dry mass (g)	Blank	MPs No Corrected (n)	MPs corrected (n)	MP Mass No corrected (ng)	MP Mass corrected (ng)
PB02	NA	NA	13	13	2741.98737	2741.98737
PB01	NA	NA	6	6	1231.76903	1231.76903
3435cm		PB02	7	4	5283.22834	5104.23059
01cm	22.768	PB02+3435cm	174	158	13963.2174	12872.0385
12cm	35.03	PB02+3435cm	74	61	116256.838	50164.7075
23cm	44.31	PB01+3435cm	37	32	377312.365	33209.2504
34cm	45.849	PB02+3435cm	226	209	26620.1472	17223.6729
56cm	52.346	PB02+3435cm	154	141	13038.9305	12502.0483
67cm	58.565	PB01+3435cm	225	217	10944.117	7413.83108
78cm	59.508	PB02+3435cm	51	42	4404.10945	2978.99285
89cm	58.925	PB01+3435cm	46	42	2991.06673	2938.55209