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ÁREAS MARINHAS PROTEGIDAS SOB AMEAÇA DE MICROPLÁSTICOS: UMA ABORDAGEM MULTI-ESCALAR

BEATRIZ ZACHELLO NUNES

Tese apresentada ao Programa de Pós-Graduação em Oceanologia, como parte dos requisitos para a obtenção do Título de Doutor.

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Rio Grande, RS, Brasil

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por

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Rio Grande, RS, Brasil

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De acordo com a ONU, a tripla crise planetária inclui as mudanças climáticas e a poluição como principais propulsores da perda contemporânea de biodiversidade. Nessa perspectiva, os aportes de resíduos sólidos em ambientes naturais, geram impactos sobre diferentes níveis de organização. Entre os materiais de origem antropogênica, os produzidos a base de polímeros plásticos, desde macro à microplásticos (MPs), têm sido frequentemente identificados nos ambientes marinhos, representando uma ameaça significativa à biodiversidade. Em resposta as estas ameaças, a implementação de Áreas Marinhas Protegidas (AMPs) é parte de uma estratégia global para alcançar objetivos de conservação, tais como as Metas de Aichi e a Agenda 2030. Contudo, AMPs, especialmente em categorias de manejo mais restritivas (*no-take*), permanecem vulneráveis à vários estressores ambientais. Embora os MPs sejam reconhecidos como contaminantes de preocupação emergente onipresentes, de alta persistência, e prejudiciais à vida humana e marinha, sua ocorrência, distribuição e impactos foram escassamente estudos em AMPs. Nesse sentido, o presente estudo avaliou em escalas global e nacional a contaminação por MPs atingindo AMPs, testando a hipótese de que a contaminação no interior dessas áreas é similar àquela observada em áreas não protegidas. Enquanto a avaliação global empregou dados secundários da literatura, avaliando amostras de água sedimento e biota, as abordagens nacionais foram realizadas usando moluscos bivalves como sentinelas da contaminação por MPs, em AMPs selecionadas no Brasil e na Austrália. A avaliação global, analisando dados de contaminação em água do mar, com base em publicações realizadas entre os anos de 2017 e 2020, mostrou que 68 AMPs apresentaram contaminação por MPs. Similarmente, um total de 186 AMPs tiveram registros de contaminação com base em estudos que avaliaram a ocorrência de MPs em amostras de sedimento e biota. Em ambos os casos, as AMPs, pertencentes a diferentes categorias de gestão, mostraram níveis de contaminação indistintamente distribuídos quando comparados a áreas não protegidas, com concentrações mais elevadas em áreas de uso múltiplo ou não categorizadas pela IUCN. Adicionalmente, aproximadamente metade dos registros foram associados aos quartis de alta concentração. Os estudos de campo empregando sentinelas, em áreas de proteção integral do Brasil e da Austrália, mostraram níveis de moderados a baixos quando comparados a áreas sem proteção ou de uso múltiplo. Mais além, os tipos de polímeros encontrados foram consistentes com aqueles reportados para regiões costeiras e urbanizadas, indicando potenciais fontes múltiplas de contaminação para esta AMPs. Em ambos os países do Sul global, nenhuma correlação significativa foi encontrada entre as concentrações de MPs e níveis de urbanização, sugerindo que fatores como fontes locais e condições hidrodinâmicas podem estar influenciando a contaminação. Tais achados sugerem que as áreas estudadas oferecem algum nível de proteção contra a contaminação, embora mais esforços de manejo sejam necessários para atingir níveis de proteção compatíveis com os internacionalmente almejados. Embora a implementação de AMPs ao redor do mundo possam fornecer algum grau de proteção a essas áreas, os níveis de contaminação encontrados com base em dados secundários e nas análises *in situ*, indicam fontes difusas de contaminação. Assim, monitoramentos de longo prazo devem ser realizados avaliando simultaneamente o risco ecológico e tendências temporais. Tal abordagem é essencial, considerando o arcabouço regulatório que potencialmente derivará das decisões tomadas junto ao tratado global dos plásticos.

Palavras-Chave: microplástico, conservação, poluição, biodiversidade.

Abstract

According to the UN, the triple planetary crisis includes climate change and pollution as the main drivers of contemporary biodiversity loss. From this perspective, the input of solid waste into natural environments generates impacts on different levels of biological organization. Among materials of anthropogenic origin, plastic polymers, from macro to microplastics (MPs), have been frequently identified in marine environments, representing a significant threat to biodiversity. In response to these threats, the implementation of Marine Protected Areas (MPAs) is part of a global strategy to achieve conservation objectives, such as the Aichi Targets and the 2030 Agenda. However, MPAs, especially in more restrictive management categories (*no-take*), remain vulnerable to several environmental stressors. Although MPs are recognized as ubiquitous, highly persistent contaminants of emerging concern that are harmful to human and marine life, their occurrence, distribution, and impacts have been scarcely studied in MPAs. In this sense, the present study assessed microplastic contamination reaching MPAs on a global and national scales, testing the hypothesis that contamination within these areas is similar to that observed in non-protected areas. While the global assessment used secondary data from the literature evaluating water, sediment and biota samples, the national approaches were carried out using bivalve molluscs as sentinels of MP contamination, in selected strictly protected areas in Brazil and Australia. The global assessments analyzing contamination data in seawater showed, based on publications carried out between 2017 and 2020, that 68 MPAs presented contamination by MPs. Similarly, a total of 186 MPAs had contamination records based on studies that evaluated the occurrence of MPs in sediment and biota samples. In both cases, MPAs belonging to different management categories showed indistinctly distributed contamination levels when compared to unprotected areas, with higher concentrations in areas of multiple use or not categorized by the IUCN. Additionally, approximately half of the records were associated with the high concentration quartiles. Field studies using sentinels in strictly protected areas in Brazil and Australia showed moderate to low levels when compared to unprotected or multiple use areas. Furthermore, the types of polymers found were consistent with those reported for coastal and urbanized regions, indicating potential multiple sources of contamination for these MPAs. In both countries of the global South, no significant correlation was found between microplastic concentrations and levels of urbanization, suggesting that factors such as local sources and hydrodynamic conditions may be influencing contamination. These findings suggest that the studied areas offer some level of protection against contamination, although more management efforts are needed to achieve protection levels compatible with Aichi Target 11 and the 2030 Agenda. Although MPAs around the world can provide some degree of protection, the contamination levels found based on secondary data and in situ analyses are concerned for diffuse sources of contamination. Thus, long-term monitoring should be undertaken to simultaneously assess ecological risk and temporal trends. This approach is essential considering the regulatory framework that will potentially derive from decisions taken under the global plastics treaty.

Keywords: microplastic, conservation, pollution, biodiversity.

Contents

Capítulo I: Introdução Geral

De acordo com a Organização das Nações Unidas, a humanidade está diante de uma tripla crise global que interconecta as mudanças climáticas, a poluição e a perda sistêmica de biodiversidade (Almroth et al., 2022). No contexto da poluição, as zonas costeiras e marinhas têm sido alvo de lançamentos de substâncias químicas perigosas e resíduos em proporções nunca vistas na história. Essa situação é majoritariamente consequência do crescimento da população global, urbanização desordenada e mudanças no estilo de vida da sociedade, desde a revolução industrial (Hoornweg et al., 2013). No centro do debate global sobre a poluição marinha, estão os produtos a base de polímeros plásticos, que representam o principal tipo de resíduo sólido de origem antropogênica encontrado nos mares e oceanos (Andrades et al., 2020). Nesse contexto, a comunidade científica internacional e gestores públicos em vários países, têm concentrado esforços para entender, dimensionar e mitigar os impactos do consumo disseminado do plástico pelas sociedades contemporâneas (Thompson, 2022).

De acordo com Young et al. (2016), a poluição é uma das principais ameaças globais à biodiversidade oceânica, frequentemente causada pelo descarte deliberado ou acidental de resíduos ou substâncias químicas perigosas no ambiente (Tilman et al., 2001). Para fazer frente às ameaças recentes à biodiversidade global, algumas ferramentas de conservação, entre elas a criação de unidades de conservação, têm sido implementadas por organizações internacionais como a "*International Union for Conservation of Nature*" (IUCN) (Edgar et al. 2014). Unidades de Conservação (UC) são definidas como áreas legalmente delimitadas e protegidas, abrangendo várias formas de utilização de seu espaço e recursos e que tenham como objetivo a conservação da biodiversidade.

Expandir e implementar sistemas de áreas protegidas, visa atingir a décima primeira Meta de Aichi estabelecida durante a 10ª Convenção sobre Diversidade Biológica (COP-10) ocorrida em Nagoya (Japão). Portanto, nos últimos anos, a quantidade de UCs no mundo tem aumentado principalmente no que tange as Áreas Marinhas Protegidas (AMPs) (Araújo e Bernard, 2016; UNEP -

WCMC e IUCN - WCPA, 2018). O engajamento global para atingir as Metas de Aichi no prazo estabelecido, tem se mostrado crescente desde 2014. De fato, AMPs de grande escala têm sido criadas como estratégia para atingir as metas de conservação marinha (Ban et al. 2017). Até 2017, ocorreu um incremento da cobertura de áreas protegidas, principalmente em áreas sob jurisdições nacionais, atribuídos, principalmente, à criação de novas UCs em diversos países, incluindo a criação da maior AMP do mundo localizada no mar de Ross no Oceano Antártico (CCAMLR-XXXV 2016). A convenção de Aichi estabeleceu 20 metas de biodiversidade para serem alcançadas até 2020, incluindo a proteção de 17% das áreas terrestres e de águas continentais e 10% das áreas marinhas e costeiras. Considerando que estas metas não foram atingidas, após 2020, a Agenda 2030 para o Desenvolvimento Sustentável introduziu novos objetivos, mantendo a criação e expansão dos sistemas de áreas protegidas como uma prioridade essencial para a conservação global da biodiversidade (UNEP-WCMC, 2021).

Segundo as categorias de manejo estabelecidas pela IUCN, as AMPs podem ter diferentes usos e objetivos. Por exemplo, reservas biológicas, refúgios de vida silvestre, parques nacionais e monumentos naturais, são considerados AMPs de uso restrito (ou proteção integral), onde as atividades humanas não são permitidas ou são limitadas. Por outro lado, AMPs de uso sustentável permitem algum grau de interação humana incluindo, em alguns casos, a exploração sustentável de recursos naturais e a ocupação por comunidades tradicionais e são elas reservas extrativistas, áreas de proteção ambiental, áreas de relevante interesse ecológico e florestas nacionais (Dudley, N, 2008). Assim, as AMPs são um instrumento político eficiente para conservar a biodiversidade marinha e costeira e seus aspectos ambientais e socioculturais relevantes (Day et al., 2012).

Considerando os fatores responsáveis pela perda contemporânea de diversidade biológica (Young et al. 2016), as áreas protegidas contribuem para a conservação desses recursos e são instrumentos potenciais para a reversão de tendências negativas (Watson et al. 2014). Além disso, do ponto de vista ecológico, AMPs de larga escala apresentam maior importância devido a sua capacidade de alcançar ecossistemas inteiros, permitindo ligações dinâmicas entre eles e aumentando a resiliência a distúrbios (Ban et al. 2017). Entretanto, a estratégia de criação de grandes AMPs visando apenas atingir as metas globais estabelecidas, de forma a atender apenas a compromissos políticos em detrimento da conservação efetiva da biodiversidade, vem sendo recentemente debatido (Ban et al. 2017; Lewis et al. 2019). Diferentemente das áreas protegidas terrestres, as áreas marinhas protegidas estão sujeitas a ação das marés e das correntes oceânicas, que podem atuar diretamente na entrada de agentes externos (Day et al., 2012).

Entre as várias classes de contaminantes que atingem os oceanos globais, os MPs tornaram-se uma questão particularmente preocupante devido à sua ampla distribuição, persistência e potencial de dano à vida humana e marinha (Boucher and Friot, 2017). Essas partículas medem entre 1 µm e 5 mm (Frias and Nash, 2019), sendo internacionalmente reconhecidas como uma ameaça ambiental. Os principais tipos de MPs encontrados em mares e oceanos incluem fragmentos de resíduos plásticos descartados irregularmente ou que escaparam aos mecanismos de manejo, fibras sintéticas de roupas, resíduos de atividade pesqueira e microesferas de produtos cosméticos e de limpeza (Schmid et al., 2021). Por causa de sua alta persistência e mobilidade, os MPs tornaram-se globalmente onipresentes em ambientes urbanos e naturais (Bindoff et al., 2019). Nesse sentido, os MPs têm sido amplamente detectados em diferentes matrizes ambientais de habitats costeiros, como praias (Akkajit et al., 2021; Alvarez-Zeferino et al., 2020), manguezais (Celis-Hernández et al., 2021; Deng et al., 2020), estuários (Harris, 2020; Pagter et al., 2020; Villagran et al., 2020), plataforma continental (Carretero et al., 2021), águas superficiais (Silvestrova e Stepanova, 2021), coluna d'água (Defontaine et al., 2020), sedimentos (Cruz et al., 2019; Pagter et al., 2020) e até mesmo no fundo do oceânico (Zhang et al., 2020). Além disso, estudos sistemáticos recentes demonstraram que os MPs são uma ameaça à saúde das áreas marinhas protegidas (Nunes et al., 2023a, 2023b) podendo induzir efeitos deletérios na biota marinha (Pauna et al., 2019) com potencial para comprometer a biodiversidade.

Esforços globais para implementar AMPs têm sido realizados por meio de agendas e convenções internacionais, incluindo a recente discussão sobre um tratado juridicamente vinculante para reduzir os impactos ambientais do plástico (March et al., 2022). Apesar disso, mais da metade das áreas marinhas protegidas do mundo têm falhado em proteger a biodiversidade oceânica devido a vários estressores ambientais (Ohayon et al., 2021). A poluição, que tem sido amplamente negligenciada nos planos de gestão de áreas protegidas em todo o mundo, é uma questão particularmente desafiadora para as AMPs, uma vez que evitar que moléculas e resíduos prejudiciais atinjam essas zonas é uma impossibilidade óbvia (Campbell et al., 2016). Essas áreas são criadas e projetadas para conservar ambientes marinhos, entretanto, a entrada de MPs tem interferido, por exemplo, na reprodução e alimentação de espécies sensíveis (Di Renzo et al., 2021). Além disso, os MPs podem atuar como vetores de outras substâncias perigosas adsorvidas na superfície dos materiais, induzindo efeitos ainda mais deletérios nos organismos expostos (Gola et al., 2021). Portanto, o controle e monitoramento da contaminação é importante para garantir a conservação das AMPs, desempenhando um papel essencial nas avaliações de impactos e fontes, permitindo o desenvolvimento de medidas eficazes para minimizar os efeitos negativos.

As AMPs sob categorias de gestão mais restritivas (IUCN - categorias Ia, Ib, II e III) são especialmente vulneráveis a poluição, o que pode afetar a biodiversidade a ser protegida (Liao et al., 2021). Portanto, elas devem ser priorizadas por estudos que avaliem os potenciais impactos da poluição. Paralelamente, espécies de bivalves filtradores são globalmente aceitos como ferramentas adequadas para investigar a contaminação por MPs em sistemas aquáticos (Kazour and Amara, 2020). Nessa perspectiva, entender o quanto a implementação de AMPs tem sido efetiva na proteção quanto a contaminação por MPs, tanto em escala global como nacional, é essencial para subsidiar as discussões internacionais e os tomadores de decisões com informações cientificamente embasadas.

Capítulo II: Objetivo

Avaliar em escalas globais e nacionais a contaminação por microplásticos atingindo Áreas Marinhas Protegidas.

Objetivos Específicos

1. Avaliar globalmente a contaminação por microplásticos em água do mar de Áreas Marinhas Protegidas;

2. Avaliar globalmente a contaminação por microplásticos em sedimentos e organismos de Áreas Marinhas Protegidas;

3. Avaliar a contaminação por microplásticos em Áreas Marinhas de Proteção Integral no litoral brasileiro, utilizando moluscos bivalves como sentinelas;

4. Avaliar a contaminação por microplásticos, em Áreas Marinhas de Proteção Integral no litoral nordeste australiano utilizando moluscos bivalves como sentinelas.

Capítulo III: Hipótese

Áreas Marinhas Protegidas, em escalas globais e nacionais, estão sujeitas a contaminação por microplásticos de forma similar a áreas não protegidas.

Capítulo IV: Área de Estudo

As áreas de estudo abrangem áreas protegidas na Austrália e no Brasil. Na Austrália, a pesquisa foi realizada em Townsville, situado em Queensland, na região nordeste do país, em Cleveland Bay, voltada para o Mar de Coral. Townsville, estabelecida em 1864, é um importante centro portuário de comércio de containers e automóveis no norte da Austrália, com uma população estimada de 201.433 habitantes em 2023. A região inclui a Pallarenda Beach, situada próxima à cidade e parte do Parque de Conservação Terrestre e Águas Interiores de Cape Pallarenda (Categoria III da IUCN), conhecida por sua acessibilidade e oportunidades recreativas, como natação e pesca. A Ilha Magnetic, localizada a 8 km de Townsville, faz parte do Parque Marinho da Grande Barreira de Coral, enfrentando potenciais contaminações por MPs devido a atividades humanas, como dragagem de manutenção no Nelly Bay Marina, e processos naturais como o transporte de sedimentos durante tempestades e ciclones. A ilha tem uma parte protegida pelo Parque Nacional da Ilha Magnetic (Categoria II da IUCN – zona de no-take) e áreas menores protegidas por Parques de Conservação Terrestre (Categoria III da IUCN – zona de no-take). Todas as áreas costeiras de Townsville e da Ilha Magnetic são protegidas pelo Patrimônio Mundial da Grande Barreira de Coral e pelo Parque Marinho da Grande Barreira de Coral (Categoria Ia da IUCN – no-take) e uma zona de uso múltiplo (Categoria IV da IUCN).

No Brasil, o estudo incluiu dez AMPs de "no-take", onde foram coletadas amostras de bivalves para avaliar a contaminação por MPs. Essas AMPs foram escolhidas para análise comparativa com áreas não protegidas, visando estudar a proximidade das fontes de poluição e a eficácia das estratégias de manejo de cada região.

Capítulo V: Material e Métodos

Estudos Sistemáticos

Os estudos sistemáticos foram conduzidos por meio da análise de dados disponíveis na literatura científica, com o objetivo de identificar a presença de MPs em diferentes ambientes marinhos. A seleção dos estudos foi feita com base em critérios rigorosos, incluindo a qualidade das metodologias empregadas e a relevância geográfica dos locais analisados. As amostras de dados foram extraídas de publicações que detalhavam métodos de coleta, processamento e análise de MPs em organismos marinhos, como bivalves. Para a análise, foram incluídos dados de concentrações de microplásticos em áreas de conservação marinha, com foco na comparação entre regiões de proteção integral e áreas de uso sustentável. A busca por estudos relevantes foi realizada em bases de dados científicas reconhecidas, como Web of Science e Google Scholar, e os critérios de inclusão incluíram apenas estudos com amostras que passaram por um processo de análise laboratorial rigoroso, como microscopia de alta resolução e espectroscopia.

Estudo de Campo

O estudo de campo foi realizado em locais selecionados com o objetivo de investigar a presença de MPs em organismos marinhos em diferentes tipos de áreas de conservação e uso. As amostras foram coletadas manualmente de locais específicos, utilizando bivalves como organismos sentinelas para a detecção de MPs. As coletas foram conduzidas em áreas de proteção integral, onde atividades extrativas são proibidas para preservar a biodiversidade, e em zonas de uso sustentável, permitindo atividades reguladas. Após a coleta, os bivalves foram transportados em condições apropriadas para o laboratório, onde os tecidos moles foram separados das conchas e submetidos a um processo de digestão em solução de hidróxido de potássio (KOH) a 40°C por um período de 48 horas.

O processo de filtragem foi realizado com malhas de diferentes tamanhos para capturar partículas de MPs, usando água destilada filtrada para evitar contaminações externas. As amostras filtradas foram analisadas com microscopia de alta resolução para identificar a presença de MPs, e a identificação dos polímeros foi feita com a técnica de espectroscopia ATR-FTIR. O controle de qualidade foi garantido pela inclusão de amostras em branco, para assegurar a ausência de contaminação durante todas as etapas do processo de análise.

Capítulo VI: Artigos Científicos

Para a obtenção do título de Doutor pelo Programa de Pós-Graduação em Oceanografia Física, Química e Geológica, é requerido que o discente realize a submissão de pelo menos dois artigos científicos como primeiro autor em periódico com corpo indexado. Desse modo, os resultados da pesquisa desenvolvida durante o período de doutorado e a discussão dos resultados serão apresentados em forma de artigos neste Capítulo. O primeiro manuscrito, de autoria de Beatriz Zachello Nunes, Yuyue Huang, Victor Vasques Ribeiro, Siqi Wu, Henrik Holbech, Lucas Buruaem Moreira, Elvis Genbo Xu e Italo B. Castro, é intitulado "***Microplastic contamination in seawater across global marine protected areas boundaries***" e foi submetido para publicação no periódico "***Environmental Pollution***. O segundo manuscrito, de autoria de Beatriz Zachello Nunes, Lucas Buruaem Moreira, Elvis Genbo Xu e Ítalo Braga Castro é intitulado "***A global snapshot of microplastic contamination in sediments and biota of Marine Protected Areas***" e foi publicado no periódico ***Science of The Total Environment***. Os artigos a seguir estão submetidos ou em fase final de redação para submissão e são respectivamente "***Microplastic in no-take Marine Protected Areas from Brazil: bivalves as sentinels***" de autoria Beatriz Zachello Nunes, Victor Vasques Ribeiro, Clara Galacho Leal, Cherie Ann Mottib e Ítalo Braga Castro, e "***Microplastic in bivalves from Great Barrier Reef, an Australian World Heritage Site***" de autoria Beatriz Zachello Nunes, Marina F. M. Santana, Cherie A. Motti e Ítalo Braga Castro.

Microplastic contamination in seawater across global marine protected areas boundaries

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Microplastic contamination across global Marine Protected Areas boundaries

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Abstract

Despite the relatively rich literature on the omnipresence of MPs in marine environments, the current status and potential ecological impacts of MPs on global Marine Protected Areas (MPAs) are still unknown. Their ubiquitous occurrence, increasing volume, and ecotoxicological effects have made microplastic an emerging marine pollutant. Given the critical conservation roles of global MPAs that aim to protect vulnerable marine species, biodiversity, and resources, it is essential to have a comprehensive overview of the occurrence, abundance, distribution, and characteristics of MPs in MPAs including their buffer zones. In the present study, extensive data were collected and screened based on 1,549 peer-reviewed literature from 2017 to 2020, and a GIS-based approach was applied to improve the outcomes by considering boundary limits. MPs in seawater samples were verified within the boundaries of 52 MPAs; after including the buffer zones, 31% more (68 MPAs) were identified as contaminated by MPs. A large range of microplastic levels in MPAs was summarized based on water volume (0 to 809,000 items/m3) and surface water area (21.3 to 1,650,000,000 items/km2), which was likely due to discrepancy in methods among different studies. Fragments were the most frequently observed shapes, followed by fibers, films, and foam, while fiber was the most abundant shape. PE and PP were the most common and also most abundant polymer types identified in MPAs. Overall, 65.8% of available data reported that microplastic levels in MPAs were higher than 12,429 items/km2, indicating that global MPAs alone cannot protect against microplastic pollution. The limitations of current microplastic studies on MPAs and future directions were also discussed toward the post-2020 Global Biodiversity Framework (GDF) goals.

Keywords: Pollution; Seawater; Conservation; Polymer; Debris; GIS

Introduction

Marine Protected Areas (MPAs) provide protection, restoration, and understanding of the global marine heritage through the creation of a representative system of conservation units (Dudley, 2008). Such areas are created and designed in accordance with the World Conservation Strategy proposed by the International Union for Conservation of Nature and Natural Resources (IUCN, 1980). MPAs are zones geographically delimited under pre-defined management goals, which may include economic resource concerns, biodiversity conservation, and species protection. According to MPAs categories and objectives established by IUCN, there are permitted and non-permitted uses. On the other hand, nature reserves, wilderness areas, national parks, and natural monuments are considered no-take MPAs (where human activities are limited) while sustainable-use MPAs and some degrees of human interactions are allowed (Dudley, 2008).

MPAs are expected to exhibit pristine conditions with no or minimal degradation, and for those affected by anthropic activities, natural restoration can occur to ensure that the biodiversity is effectively protected from different stressors (Abessa et al., 2018), addressing thus the conservation goals established by the Convention on Biological Diversity (CBD). Along with other strategies, MPAs represent a global effort to mitigate biodiversity loss that is largely driven by contamination/pollution (Young et al., 2016). Despite this, threats to the integrity and objectives of these units have been reported in the literature. In this regard, previous studies demonstrated wide contamination by organic contaminants, for example, endocrine disrupting chemicals (EDCs) posing ecological risks to MPAs in Hong Kong (Xu et al., 2019, 2016, 2015), and polycyclic aromatic hydrocarbons (PAHs) (Nunes et al., 2021) and tributyltin (TBT) (Castro et al., 2021) affecting dozens of MPAs from Latin America. In addition, studies assessing levels and biological effects of hazardous residues within specific MPAs have demonstrated simultaneously environmental occurrence and potential impacts (Abessa et al., 2017; Araujo et al., 2013; Baztan et al., 2014; Cruz et al., 2019). However, pollution information is available for only 67 of 13,674 MPAs worldwide according to

Abessa et al., (2018). Therefore, it is urgent to systematize the available data, allowing overviews and management actions in a global context.

MPs are widely present in the contemporary world with fast-increasing records reported in different environmental matrices such as water, sediment, and biota (Jiang et al., 2022) Nguyen et al., 2019), as well as in drinking water, foods, and air that poses a hazard to human health (Zhang et al. 2020). Although the exact dimensions of MPs are still under debate, these small plastics, with size range between 0.1 μm and 5 mm, can originate from the fragmentation of larger plastic pieces (Sorasan et al., 2022), or can be directly released into the environment as primary MPs, such as microbeads used in personal care products (Xanthos and Walker, 2017) or pellets leaked during the transport of raw polymer materials (Loubet et al., 2022). Also, synthetic fiber released from textile utensils during washing, drying, and wearing has been reported as one of the most commonly detected types of microplastic in natural environments (Periyasamy and Tehrani-Bagha, 2022). In addition, tire MPs are identified as one of the most abundant types of MPs, which originate from tire wear particles, recycled tire crumb , and tire repair-polished debris (Luo et al., 2021). It is known that MPs, particularly fibers, can be transported for long-range distances to isolated protected areas by wind and rain (Brahney et al., 2020), providing direct evidence of MPA susceptibility to microplastic pollution. Recently, OECD warned that the microplastic leakage is projected to more than double, from 2019 to 2060 (> 5Mt; OECD 2022). Overall, the continuous and increasing inputs of MPs to the ocean, long-distance transport, and potential toxicity of MPs (Matthews et al., 2021) have led to global concerns even on remote MPAs (Dehaut et al., 2016).

Experimental and field studies have demonstrated the impacts induced by microplastic exposure on marine organisms. At molecular to organism levels, microplastic ingestion may lead to obstruction and failure of the digestive tracts as observed in Locations of Australia’s Great Barrier Reef (Hall et al., 2015), oxidative stress in fish exposed in the laboratory to food and seawater containing MP (Capó et al., 2021), structural alterations of gills and digestive glands of mussels contaminated with MP, collected in a harbor area (Vasanthi et al., 2021), decreases in energy reserves of worms in toxicity bioassay with sediments spiked with MP (Wright et al., 2013), alteration in predatory and reproductive behavior of fish exposed to MPs-enriched food (Rios-Fuster et al., 2021).Further, MPs are known vectors of hydrophobic contaminants and may also release toxic additives and pigments from their composition. Thus, microplastic alone or associated with other hazardous chemicals could be transferred along the food chain affecting fecundity, survival, and development of organisms at different trophic levels (Wang et al., 2021). Therefore, although the impacts of MPs at the ecosystem level are still unclear, MPs have been recognized as emerging contaminants causing a great threat to marine biodiversity (Khalid et al., 2021), particularly in MPAs of high ecological value.

Numerous studies described the occurrence and distribution of MPs in coastal and oceanic zones worldwide, including in MPAs. However, the data are fragmented as a result of the different regional scopes of the studies combined with sampling and analytical methodological discrepancies (Kutralam-Muniasamy et al., 2021), which makes it difficult to conduct systematic evaluations from a global perspective. Few reviews are available on MPA contamination by MPs, which gathered information from peer-reviewed literature in scientific databases (e.g., Scopus, Web of Science, Scielo and Google Scholar) by using specific keywords (e.g., marine protected areas, marine reserve, marine park, marine nature reserves, MPs) (Abessa et al., 2018; Kutralam-Muniasamy et al., 2021). By using this approach, however, the status of microplastic pollution might be underestimated when studies do not address MPA boundary limits in their sampling design or even in the study scope. One promising way to deal with such limitations is to leverage geographic data and information (GIS) across MPA boundary limits via vector analysis (Castro et al., 2021; Nunes et al., 2021).

Ambitious global efforts in marine conservation forums aim to reach 30% of the ocean coverage area protected by MPAs (MPA News, 2014) and the current status is merely 8% (WDPA, 2022), revealing a long way to meet this objective of MPA enforcement. In this regard, increasing and more accurate information on the contamination status of global MPA is necessary. Thus, the present study aimed to assess the state-of-the-art microplastic contamination in seawater across Marine Protected Areas boundaries. Given the exponential growth of microplastic monitoring data in recent years, extensive data were collected and screened based on 1,549 recent peer-reviewed publications from 2017 to 2020, and a GIS-based approach was applied to improve the outcomes by considering spatial limits. The new results and approach allowed us to simultaneously identify marine areas that may be critically threatened by MPs, reveal knowledge gaps, and provide new perspectives on the assessment and management of microplastic pollution.

Material and methods

Data collecting and screening

Peer-reviewed articles published between 2017 and 2020 were searched using the keywords "microplastic" and "seawater" in the Scopus database (http://www.scopus.com), resulting in 1,549 articles. Manual screenings were conducted, and articles were considered valid if providing the following attributes: Sampling area, country, sampling period, sampling method, and microplastic concentrations. When available, data on geographic coordinates, the month of sampling, mesh, sampling depth, polymer composition, and shape were also collected. Articles describing or modeling the environmental behavior of MPs in natural environments or investigating physiological and chemical properties of MPs in the laboratory were excluded. Microplastic abundance provided only total amounts were not used in quantitative analyses. A microplastic database was set up for MPAs based on the available attributes described above. Subsequently, data on geographic coordinates were re-checked and converted to the decimal degrees format for GIS analysis.

GIS approach and data analysis

The microplastic database was imported into QGis, open-source software for delimitating GIS data, and intersected with a shapefile containing MPAs boundaries (available in the WDPA database;

www.protectedplanet.com), using a vector analysis tool. The number of records inside each MPA polygon was inserted as a third layer for graphically visualizing the occurrence of MPs within MPAs. Then, an overview of the collected data was generated based on the abundance of MPs reported for the Continental Regions (Asia, Africa, North America, Latin America, Europe, Oceania, and international areas). MPA contaminated by microplastic was considered by analyzing the records of MPs in seawater within MPA boundaries as well as in their buffer zones consisting of a 0.01o perimeter. Abundance records obtained from the microplastic database were calculated into four quartiles and summarized for both items/km2 and items/m3. After, the abundance of MPs reported within and around MPAs was assessed and compared according to respective quartiles. The frequencies of MPs in terms of mesh size, shapes, and polymer compositions were also comprehensively analyzed. Furthermore, the status of microplastic contamination among different MPA categories was also assessed based on their management objectives (IUCN management categories; Dudley, 2008).

Results

Overview of global marine MPs

To have a global overview of the marine microplastic occurrence beyond MPA boundaries, one hundred and fifty-four valid articles were identified, totalizing 2,099 individual records of microplastic in marine and coastal waters, collected between 2017 to 2020 (Table S1). Over 80% of total abundance data (1,866) was reported as the number of MPs per volume (items/m3 or items/L). In addition, 231 records were reported by area (items/km2 or items/m2) and 2 records were expressed in mass (ng/L). The concentration of MPs per volume ranged from 0 to 809,000 items/m3; 21.3 to 1,650,000,000 items/km2 were observed per area (Figure 1).

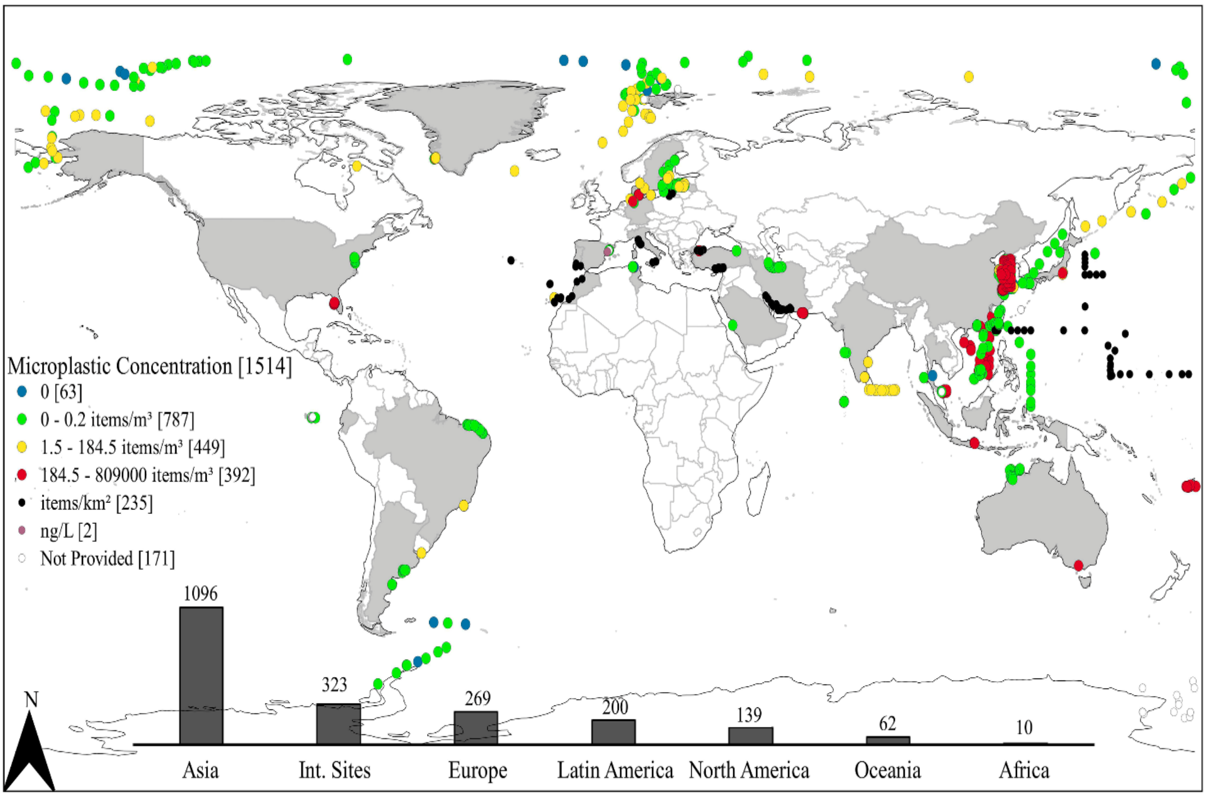

Figure 1. Global occurrence and concentration ranges of MPs in marine and coastal waters.

MPs were ubiquitously distributed along coastal and maritime zones of all continents including the Arctic and Antarctica, reaching 42 countries' territories and 12 international sites. China (24% of total records; 511 records), Turkey (6% of total records; 120 records), and India (5% of total records; 105 records) contributed the highest number of records in the database (Figure 1). 1,349 records (64.3%) reported the occurrence of MPs in surface waters (0 to 0.1 m), and 253 data (12.1%) were recorded between 0.2 and 0.5 m water depth. 494 records (23.5%) were obtained at water depth greater than 1 m, of which only 22 were sampled deeper than 1,000 meters. Approximately 28% of records (548) do not provide coordinates.

Notably, fifty-two different mesh sizes were used in different studies to collect MPs from seawater, ranging from 0.45 to 800 µm, making it impractical to evaluate and compare the size distribution of MPs in different regions and studies. Among these records, the most used meshes for sampling were 300 µm (13.5%), 330 µm (23.4%), and 335 µm (9.6%). Bulk samples (46 records) and records without mesh data (91) accounted for 2.2% and 4.3% of total records, respectively (Figure 2a).

Information on shapes and polymer compositions of MPs were provided by 1,708 (81,3%) and 1,216 (57.9%) records, respectively. A total of six main shape categories were reported, including fragments, fibers, foams, films, pellets, and others. Fragments (1,630 records) and fibers (1,505 records) were the most frequently reported (Figure 2b), while polyethylene (PE) and polypropylene (PP) were the most frequent polymers (Figure 2c). The fragments were the most abundant shape followed by fibers, foams, films, and pellets (Figure 2d). The most abundant polymer types reported included polyethylene (PE) followed by polypropylene (PP), polystyrene (PS), polyamide (PA), polyacrylic acid (PAA), and poly ethyl acrylate (PEA). It should also be noted that over 40% of microplastic records did not provide chemical information on polymer types (Figure 2e).

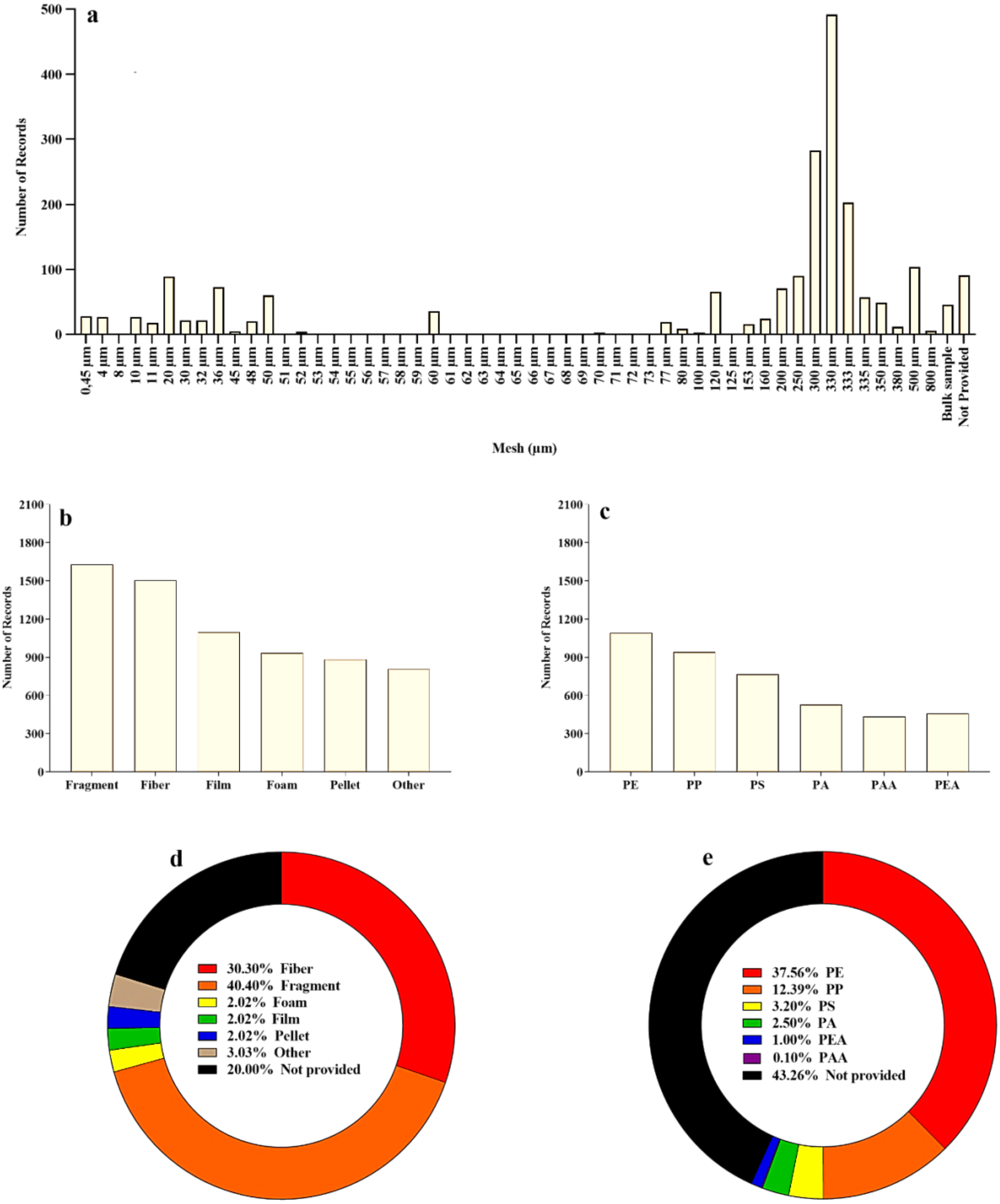

Figure 2. The total record numbers of mesh sizes (a), shapes (b), and polymer compositions (c) of microplastic in marine environments. Percentage of shapes (d) and polymer compositions (e) of MPs in marine environments.

MPs in Marine Protected Areas and buffer zones

A total of 471 microplastic records showed sampling sites within MPAs and their buffer zones (Table 1 and Figure 3). Among these studies, samples from 7 sites in South Georgia / South Sandwich Islands and Suratthani, Thailand, showed no microplastic contamination; 146 records did not provide data on microplastic abundance. A total of 375 records of microplastic occurrence were reported within the boundaries of 52 global MPAs; after including the buffer zones, 31% more (68 MPAs) were identified as contaminated by MPs (e.g. at least 1 particle found per unit in any site) (Figure 3). The 68 contaminated MPAs were distributed in Asia (9 MPAs), Europe (41 MPAs), South America (7 MPAs), North America (4 MPAs), and Oceania (7 MPAs), and no studies assessing MPs within MPAs in the African continent were found. Among MPAs assessed for MPs contamination but presenting zero values were found 1 in Asia, 11 in Europe and 2 in Antarctica.

Table 1. Marine Protected Areas affected by microplastic contamination under Management Categories (with and without buffer zone). BR= Biological Reserve; EPA= Environmental Protection Area; RS = Ramsar sites; NR= Category Not reported.

Marine Protected Areas	IUCN Category	Records in MPAs (all units)		Concentration Range			Country	References
		No Buffer	With Buffer (0.01°)	Mín - Máx (items/km²)	Mín - Máx (items/m³)	Mín - Máx (ng/L)		
Calderinhas	Ia	0	1	300			Portugal	(Herrera et al., 2020)
The Spit W.R.	Ia	0	1	253			Australia	(Su et al., 2020)
National parck of Archipiélago De Chinijo	II	2	2	68- 153			Portugal	(Herrera et al., 2020)
National Park of Kimberley	II	2	2		0.01 - 0.4		Australia	(Kroon et al., 2018)
EPAAkmensrags	IV	2	4		0.16 - 0.8		Latvia	(Aigars et al., 2021)
EPA Irbes saurums	IV	3	4		0.09 - 0.8		Latvia	(Aigars et al., 2021)
Massaciuccoli lake and marsh	IV	0	2	20936- 59,730			Italy	(Baini et al., 2018)
EPA Miankaleh	IV	1	1		0.5		Iran	(Manbohi et al., 2021)
Parco Naturale Di Migliarino, San Rossore E Massaciuccoli	IV	0	2	20936 – 59,730			Italy	(Baini et al., 2018)
Rigas Lica Rietumu Piekraste	IV	7	9		0.1 - 1.4		Latvia	(Aigars et al., 2021)
Santuario Per I Mammiferi Marini	IV	24	24	3157 – 347,040			Italy	(Baini et al., 2018)
Selga uz rietumiem no Tujas	IV	3	6		0.3 - 0.4		Latvia	(Aigars et al., 2021)
Vitrupe – Tuja	IV	1	2		0.3		Latvia	(Aigars et al., 2021)
Ærø Kommune	IV	0	2		1,010 – 1,270		Denmark	(Tamminga et al., 2018)
Mariana Trench	IV	2	2	1,584			Japan	(Pan et al., 2019)
Río Guadiana y Ribera de Chanza	IV	0	1	150,000,000			Spain	(Velez et al., 2019)

Terra Ceia Aquatic Preserve	IV	2	2		700 - 900		USA	(McEachern et al., 2019)
Pinellas County Aquatic Preserve	IV	3	4		570 - 2000		USA	(McEachern et al., 2019)
Cockroach Bay Aquatic Preserve	IV	0	1		1100		USA	(McEachern et al., 2019)
EPA Delta do Parnaiba	V	4	4		0.002 - 0.1		Brazil	(Garcia et al., 2020)
EPA Praia de Ponta Grossa	V	2	2		0.01		Brazil	(Garcia et al., 2020)
Delta de l'Ebre	V	1	3			1.08 - 137	Spain	(Schirinzi et al., 2019)
Monte da Guia	V	0	1	300			Portugal	(Herrera et al., 2020)
Sudoeste Alentejano e Costa Vicentina	V	3	3	70,000,000 – 380,000,000			Portugal	(Velez et al., 2019)
Florida Coastal Islands Sanctuaries	V	0	1		600		USA	(McEachern et al., 2019)
Amazon Estuary and its Mangroves	VI	4	4		0.002 - 0.1		Brazil	(Garcia et al., 2020)
Bahía Blanca, Bahía Falsa y Bahía Verde	VI	1	1		0.02		Argentina	(Ronda et al., 2019)
Canal Faial-Pico/Sector Faial	VI	2	2	143.8 - 300			Portugal	(Herrera et al., 2020)
Lalang-garram / Camden Sound	VI	6	6		0.01 - 0.4		Australia	(Kroon et al., 2018)
Lalang-garram / Horizontal Falls	VI	4	0		0.01 - 0.4		Australia	(Kroon et al., 2018)
North Kimberley	VI	8	0		0.01 - 0.4		Australia	(Kroon et al., 2018)
Marine Extrative reserve of Itaipu	VI	18	0		1.1 - 2.9		Brazil	(Castro et al., 2020)
Isla Graciosa Y De Los Islotes Del Norte De Lanzarote	VI	1	1	153.3			Portugal	(Herrera et al., 2020)
Archipiélágo de Colón (Galápagos)	BR	36	38		0.89		Ecuador	(Jones et al., 2021)
Biosphere Reserve of Miankaleh	BR	1	1		0.50		Iran	(Manbohi et al., 2021)

Penínsu Valdés	BR	3	3		0.01	Argentina	(Ríos et al., 2020)
Pelagos Sanctuary	BR	24	24	3,157 – 347,040		Italy	(Baini et al., 2018)
Dune costiere del Parco dell'Uccellina	RS	0	2	1,0104		Italy	(Baini et al., 2018)
Espacio marino del Delta de l'Ebre-Illes Columbretes	RS	8	10		0.4 - 0.8	Spain	(Expósito et al., 2021)
Miankaleh peninsula	RS	2	4		0.5	Iran	(Manbohi et al., 2021)
Tamarit-Punta De La Móra-Costes Del Tarragones	NR	1	2		0.8	Spain	(Expósito et al., 2021)
Área Marina de la Isleta	NR	0	1	894		Portugal	(Herrera et al., 2020)
Bahía del Confital	NR	1	1	894		Portugal	(Herrera et al., 2020)
Cetáceos da Madeira	NR	3	4	40 - 88		Portugal	(Herrera et al., 2020)
Costes del Tarragonès	NR	1	2		0.8	Spain	(Expósito et al., 2021)
Espacio marino de los Islotes de Lanzarote	NR	2	2	153		Spain	(Herrera et al., 2020)
Espacio marino del oriente y sur de Lanzarote-Fuerteventura	NR	2	2	153		Spain	(Herrera et al., 2020)
Faial-Pico Channel	NR	2	2	144 - 300		Portugal	(Herrera et al., 2020)
Hoburgs bank och Midsjöbankarna	NR	6	6		0.07 - 0.2	Sweeden	(Schönlau et al., 2020)
Monte da Guia - Ilha do Faial	NR	1	1	300		Portugal	(Herrera et al., 2020)
Palau National Marine Sanctuary	NR	4	4		0.035 - 0.04	Palau	(Liu et al., 2021)
Pineta Granducale dell'Uccellina	NR	0	2	10104		Italy	(Baini et al., 2018)
Selva Pisana	NR	0	4	59730		Italy	(Baini et al., 2018)
Skagens Gren og Skagerrak	NR	6	6		0.02 - 2.6	Denmark	(Schönlau et al., 2020)

Stora Middelgrund och Röde Bank	NR	9	9		0.01 - 14	Sweden	(Schönlau et al., 2020)
Port Phillip Bay (Western Shoreline) and Bellarine Peninsula	NR	1	1		2530	Australia	(Su et al., 2020)
Sydfynske Øhav	NR	18	26		200 – 1,270	Denmark	(Tamminga et al., 2018)
South Funen Archipelago	NR	18	26		200 – 1,270	Denmark	(Tamminga et al., 2018)
Vueti Navakavu	NR	3	4		3,200	Fiji	(Dehm et al., 2020)
Rukurukulevu/Cuvu/Sila/Tore/Naevuevu/Yadua	NR	3	4		1,000	Fiji	(Dehm et al., 2020)
Namada/Votua/Vatuolalai/Tagaqe	NR	4	4		1,000	Fiji	(Dehm et al., 2020)
Kiuva	NR	4	4		1,800	Fiji	(Dehm et al., 2020)
Biausevu/Navola/Vanua Komave-Komave/Namatakula	NR	0	4		1,000	Fiji	(Dehm et al., 2020)
Unterweser	NR	1	1		9,700	Germany	(Roscher et al., 2021)
Monti Peloritani, Dorsale Curcuraci, Antennamare e area marina dello stretto di Messina	NR	1	3		3	Italy	(Savoca et al., 2019)
Ria Formosa / Castro Marim	NR	0	1	150,000,000		Portugal	(Velez et al., 2019)
Costa Sudoeste	NR	6	6	70,000,000 – 380,000,000		Portugal	(Velez et al., 2019)
Göksu Delta	NR	0	1	120,660		Turkey	(Güven et al., 2017)

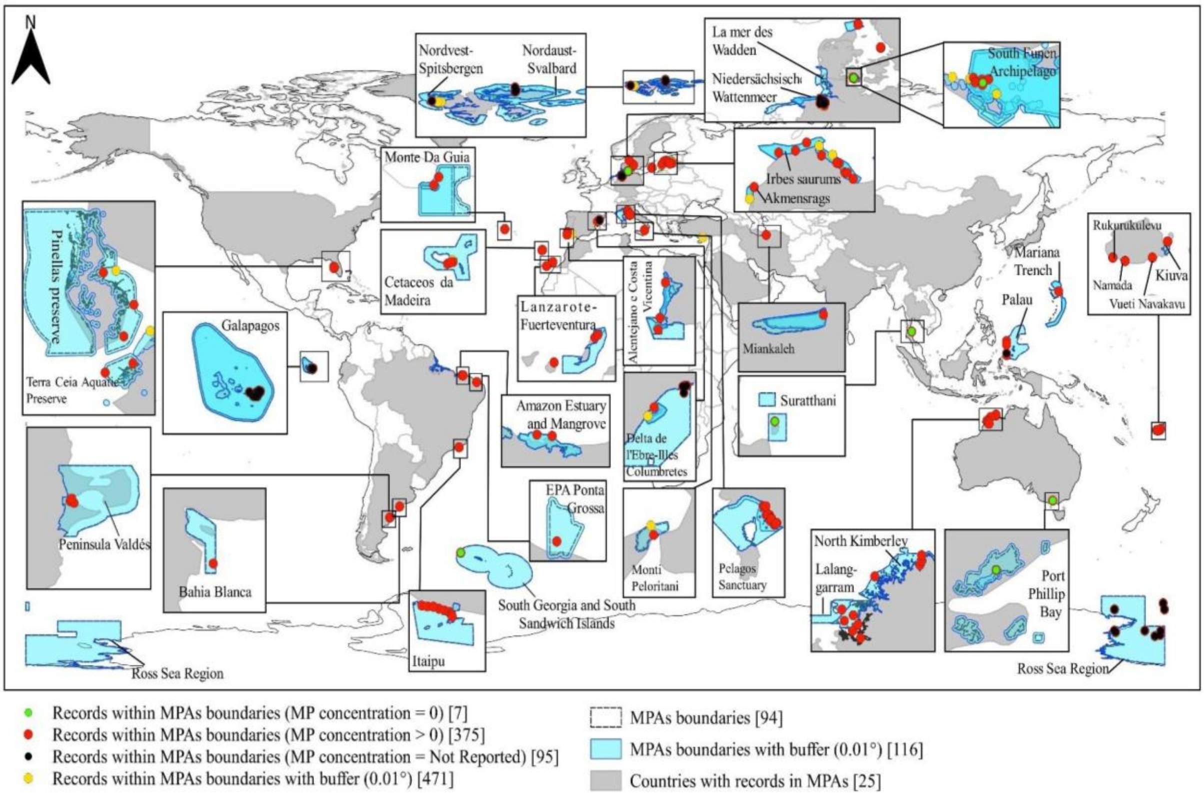

Figure 3. Global occurrence of MPs in Marine Protected Areas and buffer zones.

Based on 146 microplastic records expressed as items/km^2 in Marine Protected Areas and buffer zones, the quartile analysis resulted in four concentration ranges of microplastic, including Q1 (21.3 to 813 items/km^2), Q2 (813 to 12,429 items/km^2), Q3 (12,429 to 46,150 items/km^2) and Q4 (46,150 to 1,650,000,000 items/km^2) (Figure 4a). Among these records, 96 (65.8%) fell in the high concentration quartiles, i.e., Q3 and Q4. The number of microplastic-contaminated MPAs falling within the quartiles Q1, Q2, Q3, and Q4 were 10, 7, 5, and 11, respectively. Compared to area-based concentrations, more volume-based levels (321 records) indicated a higher number of microplastic-contaminated MPAs (48). In total, 83, 112, 14, 112 volume-based abundance records fell within Q1 (0.000042 to 0.2 items/m^3), Q2 (0.2 to 1.7 items/m^3) and Q3 (1.7 to 144.7 items/m^3), Q4 (144.7 to 809,000 items/m^3), respectively (Figure 4b). The microplastic levels per volume in 16, 15, 3, and 15 MPAs fell in the quartiles Q1, Q2, Q3, and Q4, respectively. Overall, 1/3 of microplastic-contaminated MPAs belong to the highest quartile (Q4) according to both area- and volume-based abundance data.

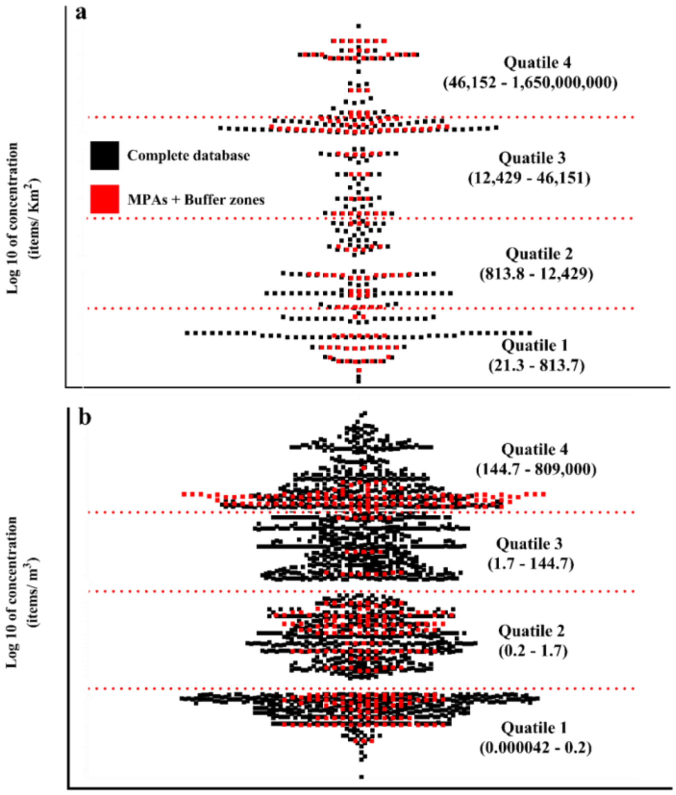

Figure 4. Quartiles of microplastic concentrations in items/m3 (a) and items/km2 (b). Black dots indicate the complete database of marine MPs; red dots indicate MPs in MPAs and buffer zones.

In terms of current sampling strategies, similar to the results of the global overview on marine microplastic, most microplastic samples in MPAs were taken from surface water (< 5 m in depth) and MP abundance in deeper zones of MPAs was largely unknown. Also, the mesh sizes of nets used in microplastic sampling varied significantly in different studies on different MPAs. Again, ~300 µm was the most frequent (44.3%) used mesh size out of a total of 11 different mesh sizes reported, and 4.7% of records contained no information on mesh size for sampling. 17% of microplastic records were obtained from bulk sampling (Figure 5a). Fragments were the most frequently observed shapes in MPAs, followed by fibers, films, foam, pellets, and others (Figure 5b), while fibers were the most abundant (Figure 5d). PE and PP were both the most common and abundant polymer types identified in MPAs (Figures 5c and 5e).

The status of microplastic contamination among MPAs under different IUCN categories was also compared to better inform MPA management (Table 1). In two nature reserves (Ia), 253 and 300 items/km2 of MPs were reported in the buffer zones around The Spit wildlife reserve (Australia) and Calderinhas strict natural reserve (Portugal), respectively. Two records (up to 153 items/km2) were found in the national parks of Chinijo Archipelago (Spain) and Kimberley (Australia). No records were available in MPAs of categories Ib (Wilderness Area) and III (Natural Monument or Feature). Among the multiple-use categories (VI, V, and VI), MPs were detected in 28 MPAs, including Habitat/Species Management Area (14 MPAs; 60 records), Protected Landscape/ Seascape (6 MPAs; 14 records), and Protected area with sustainable use of natural resources (8 MPAs; 14 records). MPAs with unassigned categories (NR) accounted for 139 microplastic records. Also, the occurrence of MPs was reported in protected areas with special diplomas, such as RAMSAR sites (3 MPAs; 16 records) and Biosphere Reserves (4 MPAs; 66 records)(Table 1). In general, fewer microplastic-contaminated MPAs belonged to no-take than multiple-use categories due to fewer records and relatively lower levels of MPs reported in MPA categories I and II. However, it was unknown whether categories I and II were more protected from MPs than the multiple-use ones because of the limited sample size per MPA category.

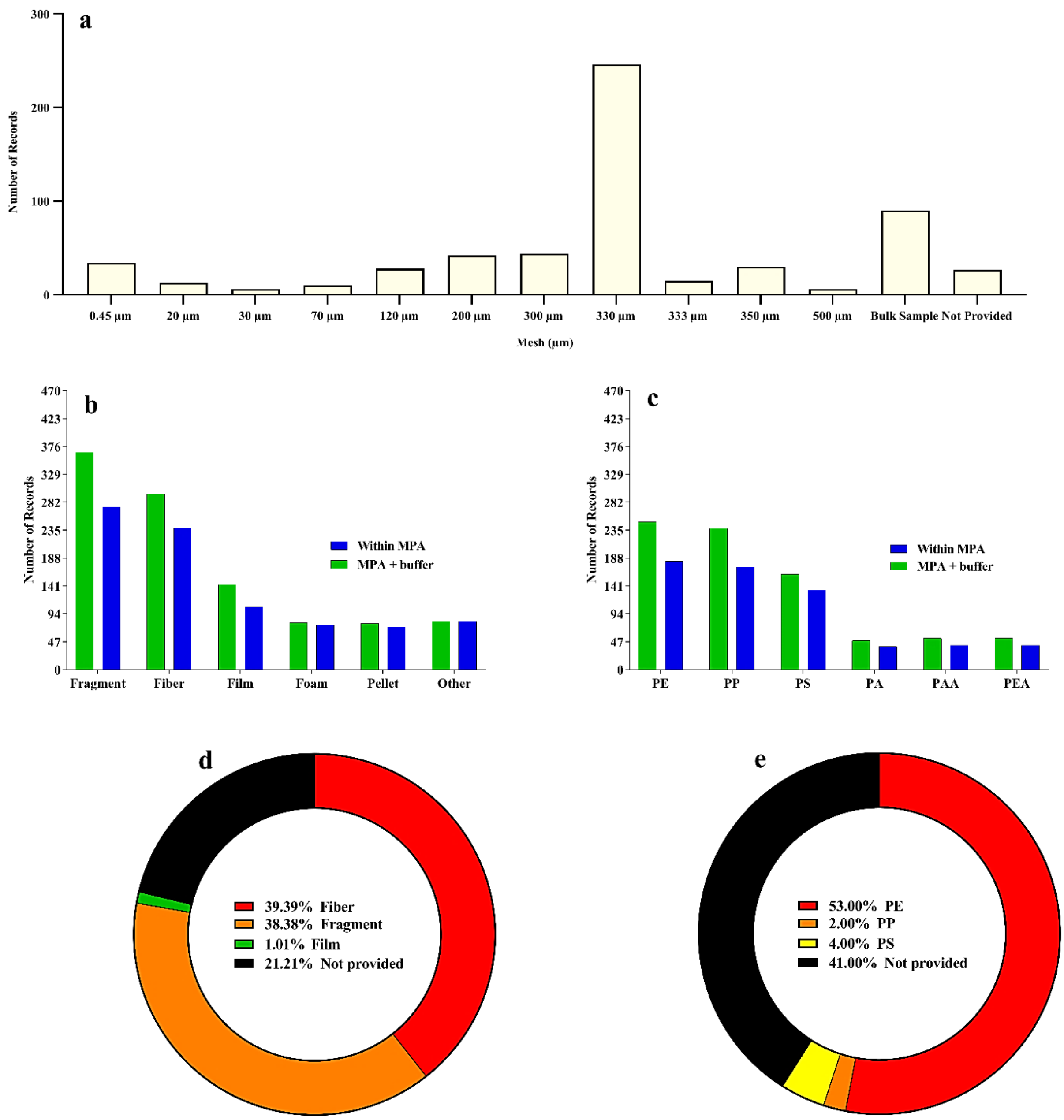

Figure 5. The total record numbers of mesh sizes (a), shapes (b), and polymer compositions (c) of microplastic in MPAs and buffer zones. Percentage of shapes (d) and polymer compositions (e) in MPAs and buffer zones.

Discussion

In the present review, the microplastic contamination across global MPA boundaries was evaluated by screening 1,549 published literature combined with a GIS-based approach considering their buffer zones. In fact, the concerns about microplastic contamination in MPAs have been raised. For example, Kutralam-Muniasamy et al. (2021) assessed the microplastic occurrence in environmental matrices (seawater, sediment, and biota) within MPAs and reported that 29 MPAs were contaminated by MPs (Kutralam-Muniasamy et al., 2021). Different from the study focusing on only areas within the MPA boundaries, we identified a larger number of microplastic-contaminated MPAs (52 within the boundaries and 68 when considering buffer zones) based on seawater data from 2017 to 2020. Such differences in results between our study and previous studies are probably due to the use of a more accurate search methodology here, allowing the inclusion of microplastic records that are not originally reported by the articles that belong to MPAs and their buffer zones (Castro et al., 2021; Nunes et al., 2021). The geospatial overlay using the shapefiles provided by the WDPA database (www.protectedplanet.com) allows for identifying records of MPs that fell into overlapping boundaries of MPAs, meanwhile highlighting the challenges of managing the microplastic pollution beyond the spatial boundaries of MPAs.

Overall, over 60% of available data reported that microplastic levels in MPAs are higher than 12,429 items/km2 (Figure 4a). Interestingly, the abundance values in MPAs are in general higher than those reported on non-protected marine areas, suggesting that MPAs are not effective to protect against microplastic pollution. Similar issues have also been demonstrated for other marine micropollutants (Abessa et al., 2018; Castro et al., 2021; Nunes et al., 2021). Hence, concerning the limitations of conservation roles of national and regional MPAs, globally integrated management and actions are necessary to mitigate and prevent marine microplastic pollution. Although not fully covered in the present review, it has been noted that different methodological approaches used from sampling and separating to identifying and quantifying MPs in different environmental samples result in high variation in microplastic abundance or concentration (Manbohi et al., 2021; Nguyen et al., 2019; Schönlau et al., 2020). Thus, the wide range in microplastic levels in MPAs, reported as both volume (0 to 809,000 items/m3) and area basis (21.3 to 1,650,000,000 items/km2), is first likely due to the use of a wide range of mesh sizes for sampling and collecting MPs (Figures 2a and 5a). Specifically, trawling nets are the most commonly used to collect MPs in surface waters, which can cover large surface water areas but lose smaller MPs that pass through the net. On the other hand, bulk samples and pumps are used to better collect smaller MPs, but the covered sampling area is limited, requiring large volumes and replicates to represent real environmental conditions. According to Lindeque et al. (2020), the use of a 100 μm net ensured a 2.5-fold and 10-fold increase in microplastic concentrations compared to 333 μm and 500 μm nets, respectively. Similarly, using a 160 μm net resulted in a 3-fold increase in microplastic levels than using a 500 μm mesh (Sun et al., 2018; Wang et al., 2019). We find that most MPA studies used mesh sizes of 300 to 335 µm (Figures 2a and 5a), which likely lead to significantly underestimated the levels of MPs in MPAs. Thus, it is suggested to employ the smaller mesh size (< 100 μm) supplemented by bulk pump sampling (McEachern et al., 2019). It is worth noting that the information on the smallest MPs (< 1 μm; i.e., nanoplastics) in MPAs is still missing due to the limitation of current methodologies (Cai et al., 2021). Besides the influence of sampling and analytical methods, the abundance of MPs in seawater is affected by a core set of factors, including oceanic currents actions, and hydrodynamic at local to global scale, seasonality, coastal geomorphology, the proximity of potential sources, and physico-chemical characteristics of plastics (Jeyasanta et al. 2020; Ibrahim et al. 2021).

Our results indicate that fibers and fragments are the most frequent shapes reported in MPAs, and PP and PE are the most prevalent polymer types (Figures 2b,c and 5b,c). One of the primary sources of environmental MPs is industrial, for instance, from the unprocessed virgin granules (pellets; 1 to 8 mm) that are released accidentally during transportation or operations(Galafassi et al., 2019). Primary MPs found in the ocean also include microbeads in personal care products and cosmetics such as toothpaste, shower gel, and face and body washes, as exfoliating materials substitutes (Rochman et al., 2015). Some products for skin peeling, for instance, plastic particles of 420 μm size, can reach natural water bodies by passing through the sewage treatment systems (Waldschläger et al., 2020).

Thus, increased care in transport and processing combined with efficient wastewater treatment technologies are needed to reduce the input of primary emissions (Ngo et al., 2019). Secondary MPs originate from the wethering, degradation, and fragmentation of larger plastic debris, e.g., single-use plastic bags, often generating plastic microfibers and fragments (Cole et al., 2011; Coyle et al., 2020; Abreu and Pedrotti, 2019; Doğan, 2021). Cloth washing also generates large number of microfibers from different synthetic polymers, such as nylon, PET, PP, PS (Gago et al., 2018; Waldschläger et al., 2020). Other sources include plastic waste in landfills (Cole et al., 2011; Coyle et al., 2020), maritime activities like fishing ropes and netting (Napper et al., 2022), and rubber MPs (Luo et al., 2021). The usage and environmental disposal of plastic face masks during the Covid-19 pandemic have been pointed out as an emergent source of microfibers to the environment (Fadare and Okoffo, 2020; Ribeiro et al., 2022; Xu and Ren, 2021). The continuous release of secondary MPs originating from complex sources poses a big threat to MPAs and how to control secondary microplastic in MPA is a difficult management task.

Most microplastic-affected MPAs are identified near the coast (Figure 3) in zones under the direct influence of urban activities or river mouths that are recognized as the major land sources of MPs in marine environments (Yuan et al., 2022). Also, the most contaminated MPAs belong to the multiple-use management categories with certain levels of human interventions allowed (Dudley, 2008). Alarmingly, some records have also been reported inside MPAs located in remote and sparsely populated areas such as Palau (Ferreira et al., 2020), Canary (Vega-Moreno et al., 2021), and Galápagos (Jones et al., 2021) islands, suggesting probable allochthonous sources such as ocean currents and airborne transport, once the influence of both means of transport on MP distribution are documented (Evangeliou et al., 2020). The occurrence of MPs in pristine areas now is evident around the world including in both aquatic and terrestrial environments beyond MPAs (Lim, 2021; Nguyen et al., 2019). Given the increasing ecotoxicological data on MPs in various organisms but limited knowledge of precise toxicity mechanisms (Matthews et al., 2021), the increasing presence of MPs in the MPAs, which are specially designed to protect and preserve unique and vulnerable species and biodiversity, is very worrying. Moreover, it is important to highlight the co-occurrence and joint impacts of other chemical contaminants in MPAs (Abessa et al., 2018; Castro et al., 2021; Nunes et al., 2021) and the vector effects of MPs MPs (Torres et al., 2021) in MPAs requires future studies.

We need to particularly acknowledge that the non-harmonized methods of sampling MPs, use of different abundance units, lacking vertical distribution data, and lacking location information make comparative assessment difficult among different studies and regions. Furthermore, only a small portion of the MPA studies provide quantitative data on the size, shape, and chemical composition of MPs, preventing a detailed evaluation of the occurrence profiles and potential sources of MPs in MPAs. Thus, it is urgent to establish and apply standardized approaches to collect and analyze MPs in various environmental matrices, particularly for multiple-regional large-scale comparative studies, aiming to achieve a meaningful global perspective on microplastic pollution across MPA boundaries.

Knowing the occurrence and levels of MPs in MPAs is the first and fundamental step for ecological risk assessment. To date, the impacts and risks of MPs and associated chemicals on MPAs at the ecosystem level are still unclear. MPs have been recognized as emerging contaminants threatening marine biodiversity (Khalid et al., 2021). The Aichi biodiversity targets were established in 2010 by the 10th Convention on Biological Diversity (CBD) with unachieved goals by 2020 (Ferreira et al., 2022). Later, a decision was issued by the CBD Parties to adopt a comprehensive and participatory process for the preparation of a post-2020 Global Biodiversity Framework (GDF). Such initiative is currently based on global-scale targets and indicators related to the main drivers of biodiversity loss including reducing pollution from all sources (CDB, 2018). In this regard, plastic debris density has been proposed by GBF as a putative indicator, although there is still no consensus on microplastic safety levels for marine environments. Still, coverage of effective protected areas and area-based conservation measures have been proposed desired among other indicators. Thus, future large-scale field studies using a harmonized approach to assess microplastic contamination and biota health across MPA boundaries may provide valuable information to support post-2020 GDF goals.

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A global snapshot of microplastic contamination in sediments and biota of marine protected areas

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A global snapshot of microplastic contamination in sediments and biota of Marine Protected Areas

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Abstract

Microplastics (MPs) become ubiquitous contaminants in Marine Protected Areas (MPA) that have been planned as a conservation strategy. The present study provides a comprehensive overview of the occurrence, abundance, and distribution of MPs potentially affecting MPA worldwide. Data on MP occurrence and levels in sediment and biota samples were collected from recent peer-reviewed literature and screened using a GIS-based approach overlapping MP records with MPA boundaries. MPs were found in 186 MPAs, with levels ranging from 0 to 9187.5 items/kg in sediment and up to 17,461.9 items/kg in organisms. Peaked MPs concentrations occurred within multiple-use areas, and no-take MPAs were also affected. About half of MP levels found within MPA fell into the higher concentration quartiles, suggesting potential impacts on these areas. In general, benthic species were likely more affected than pelagic ones due to the higher concentrations of MP reported in the tissues of benthic species. Alarmingly, MPs were found in tissues of two threatened species on the IUCN Red List. The findings denote urgent concerns about the effectiveness of the global system of protected areas and their proposed conservation goals.

Keywords: ocean; marine debris; conservation; environmental impacts; GIS

Introduction

Implementing Marine Protected Areas (MPAs) is a policy instrument to conserve marine and coastal biodiversity, its landscapes, and their relevant environmental and socio-cultural aspects (Day et al., 2012). The main targets of MPAs aim to attenuate anthropogenic threats including over-exploitation of natural resources, habitat loss, unregulated tourism, coastal urbanization, and impacts of chemical pollution (de Oliveira Júnior et al., 2021; Zupan et al., 2018). Considering that most marine biodiversity hotspots are still not protected so far, the 2030 Agenda for Sustainable Development of United Nations have proposed Sustainable Development Goals (SDGs) supporting MPAs objectives, such as SDG 14 (life below water), 13 (climate action), and 15 (life on land). In addition, the Aichi Biodiversity Targets issued by the 10th Convention on Biological Diversity, also support the management of MPAs to prevent pressures on vulnerable ecosystems, including those caused by marine pollution (United Nations, 2021). Despite these global efforts, over half of marine protected areas worldwide fail to protect ocean biodiversity due to several environmental stressors (Ohayon et al., 2021).

The release of hazardous substances and residues from anthropogenic sources has been listed as a major stressor to marine biodiversity (Landrigan et al., 2018). In this regard, the occurrence of contaminants of emerging concern (Chaves et al., 2020), antifouling biocides (Castro et al., 2021; Ribeiro-Brasil et al., 2021), polycyclic aromatic hydrocarbons (Nunes et al., 2021), toxic trace metals (Cruz et al., 2019), organ halogenated compounds including pesticides and polychlorinated biphenyls (Commendatore et al., 2015), and marine litter (Kroon et al., 2020) have been reported within MPAs. Moreover, deleterious biological effects related to the occurrence of these residues in MPAs have also been addressed (Abessa et al., 2018; Rodríguez Grimón et al., 2016; Rodríguez-Grimon et al., 2020).

Plastic debris is one of the main health threats to marine ecosystems and many previous studies have assessed their direct and indirect effects on biota (Pauna et al., 2019). Although still under debate, microplastic (MP) sizes classes were defined by Frias and Nash (2019) as particles between 1 µm and 5 mm. Such particles occur as pellets, fragments, films, and microfibers derived from the fragmentation of larger plastic or microbeads, pellets used as abrasives in personal care products, and raw materials used in the fabrication of plastic utensils (Schmid et al., 2020). Because of their high persistence and mobility, MP has become omnipresent on a global scale (Bindoff et al., 2019). MPs have been widely detected in coastal habitats and various environmental compartments, such as beaches (Akkajit et al., 2021; Alvarez-Zeferino et al., 2020), mangroves (Celis-Hernández et al., 2021; Deng et al., 2020), estuaries (Harris, 2020; Pagter et al., 2020; Villagran et al., 2020), continental shelf (Carretero et al., 2021), surface waters (Silvestrova and Stepanova, 2021), water column (Defontaine et al., 2020), sediments (Cruz et al., 2019; Pagter et al., 2020) and even in the deep ocean (Zhang et al., 2020).

Recent studies assessing MP intake by marine organisms raise global concerns about the potential ecological impacts of MP particles. Furthermore, inorganic and organic chemicals adsorbed by MP particles in the environment may induce additional damage compared to virgin MP particles (Khalid et al., 2021a). Since it is assumed that an effective MPA ensures the protection of biodiversity (Day et al., 2012), organisms inhabitant such areas are expected to be protected from the effects of MP as well. While the number of studies regarding the environmental occurrence of MP has increased over the last decade, few studies reported its impacts inside MPAs boundaries (Grillo and Mello, 2021; Kutralam-Muniasamy et al., 2021a; Lorenzi et al., 2021; Masiá et al., 2019; Ronda et al., 2019; Sevwandi Dharmadasa et al., 2021). Moreover, fewer studies investigated MPs in MPAs that are located around urban centers (Celis-Hernández et al., 2021; Ferreira et al., 2020; Ríos et al., 2020).

Kutralam-Muniasamy et al. (2021) reviewed 36 articles pointing out 35 MPAs affected by MPs contamination considering water, biota, and sediment samples. However, this review included articles by searching keywords related to MPAs but in fact, many assessments on MP contamination did not mention whether the sample grid was within of MPAs or not (Nunes et al. 2021; Castro et al. 2021). To overcome this, an approach using data collected from scientific literature overlapped with georeferenced MPA polygons can efficiently identify protected areas contaminated by MPs, producing a snapshot of MP contamination and status in global MPAs, useful for global MPA managers and stakeholders.

Sediments are known as the main sink of various chemical pollutants and MPs in the oceans. Aquatic organisms may accumulate MP from the diet, water, and sediments contaminated by MPs. In this regard, higher levels of environmental contaminants including MP are expected to be found in benthic environments in comparison to the pelagic ones, suggesting that benthic organisms are at higher risk of MP pollution. Thus, we hypothesize that sediments and biota samples can be useful and relevant targets in assessments of MP impacts on MPA. To test the hypothesis, based on data from peer-reviewed literature (2010 to 2021), the present study aims to assess the levels of MP contamination in sediments and biota of MPAs by using a GIS approach as mentioned above.

Material and methods

To assess MP contamination within global MPAs, a 7-step approach developed for polycyclic aromatic hydrocarbons (Nunes et al. 2021) and tributyltin (Castro et al. 2021) was used as follows: (1) study area delimitation, (2) bibliographic survey, (3) construction of attribute tables, (4) data insertion in a Geographic Information System (GIS), (5) overlapping with worldwide shapefiles of MPAs, (6) identification of affected MPAs, and (7) impacts assessment using quartiles.

Study area delimitation and bibliographic survey

Peer-reviewed articles published in 2020 and 2021 were searched using the keyword "Microplastic" combined with "Marine Sediment", "Marine Litter", "Marine Protected Area", "Occurrence", and "Pollution". Other keyword combinations used as an addition to fill the information gaps, were "Microplastic + [country]" or "Microplastic + [continent]". The keywords were searched in the Scopus (http://www.scopus.com) and Web of Science (https://www.webofknowledge.com/) databases. Papers describing the environmental partition of MP in environmental matrices, investigating experimental physiological responses, and reviews were not included. Articles providing attributes as described below (Table 1) were considered valid.

Table 1 Selected attributes considered to delimitate bibliographic survey

Attributes	Description
Geographic coordinates	Latitude and Longitude (decimal degrees)
Original site name	The name provided by the author to each sampling site
Country	
Sampling date	Year (whenever available, month or season)
Microplastic occurrence in sediment and/or organisms	General concentration of microplastic, concentrations by type of plastic
Taxonomic group	Class, Order, or Family
Species	Scientific name
Unit of measurement	e.g., items/kg, items/m^2, items/sample, items/site and items/organism
DOI link	
Site code	Unique assigned code for each site

Attribute tables

A dataset based on attributes (Table 1) consisted of a spreadsheet including the occurrence of MP on a site-by-site basis. Data provided graphically were extracted using WebPlotDigitizer, an open-source and free-use software. The geographical coordinates (latitude and longitude), sampling periods, and the sampling method of each site were obtained from the articles and included in a spreadsheet. Subsequently, they were individually checked and converted to the decimal degrees format, commonly used in geographic information systems (GIS). During this step, only papers providing georeferenced data on MP occurrence were considered.

Sampling methods varied widely among the consulted articles. Hence, different units were used to report MP concentrations. The most frequent data presentation was items/kg (44% of total – 1,201 records) for sediments and item/organism for biota (9% - 251 records). Whenever possible, concentrations based on the number of particles per sample mass were converted into items/kg, increasing its representation to 79% (2,164 records), for both sediment and biota samples. Data provided as numbers of particles by area and volume were used as occurrence registries and excluded from quantitative analysis.

Identification of MP-contaminated MPAs

To identify MPAs affected by MP, the database with MP occurrence was imported to QGis, a tool for delimitating GIS data. Then, data were intersected with a shapefile containing MPAs boundaries, available for free download in the World Database on Protected Areas (WDPA) (https://www.protectedplanet.net/en), using a vector analysis tool. The number of records inside each MPA polygon was inserted as a third layer allowing to visualize graphically the occurrence of MP within MPAs. The status of MP contamination among different MPA categories was also assessed based on their management objectives (IUCN management categories; Dudley, 2008).

Identifying MP-contaminated species

The IUCN categorize threatened species using a hierarchical structure assessing the risk of global extinction based on several parameters. In the red list (https://www.iucnredlist.org/), species Critically Endangered (CR) are under "extremely high risk", while Endangered (EN) and Vulnerable (VU) categories indicate respectively "very high risk" and "high risk" of extinction in the wild (IUCN, 2001). This database was used to identify the threatened species sampled within MPAs presenting MPs accumulated in their tissues. In addition, MP records of species reported as items/kg were assessed for ecological attributes of the domain (benthic or pelagic) and feeding (herbivores, detritivores, omnivores, and carnivores). For each species, information was retrieved from its respective article when available or consulted on biodiversity databases (e.g., https://www.marinespecies.org/ and https://www.fishbase.se).

Statistical Analyses

Descriptive statistics including the means and standard errors of microplastic concentrations were provided to describe the occurrence, abundance, and distribution of MPs in the different MPAs. Four quartiles of concentration data were calculated and analyzed based on abundance records in biota and sediment samples, respectively. The quartile assessments were conducted for MPs in global coastal zones (i.e., the complete dataset) and also for MPs reported within MPAs.

Results and Discussion

Considering that methodological parameters may influence the data of MP occurrence, the dataset was assessed for the adopted sampling methods, sample size, and mesh porosity used in the extractions. For sediments, 134 studies assessed MP occurrence by using four different sampling instruments. Scoops were used to collect surface sediments or sand beach in 33% of studies, while grabs (i.e., Ekman and Van Veen grab), box corer, and cylindric cores represented 30, 30, and 20% respectively (Figure 1a). Similarly, the sediment mass used in the analysis varied widely, and 69 studies did not provide this information. A total of 34 studies (24%) reported only transect areas providing information in items/m^2, while three reported only sample volumes. Zinc chloride, sodium chloride, and sodium iodide were often used as flotation solutions to extract MPs from sediment samples (Fang et al., 2021; Patti et al., 2020; Sunitha et al., 2021). In addition, studies assessing sediments with high organic matter amounts also included oxidative treatment generally using hydrogen peroxide (Abel et al., 2021). Thirty-five different mesh sizes were used ranging from 0.45 to 1,000 µm, which may affect recoveries for each method, thus influencing the size distribution of the reported MP frequency (Sánchez-Hernández et al., 2021). Overall, 19 out of 134 studies used 0.45 µm filter/mesh, 26 used 1.2 µm, and 73 remained between 1 and 1,000 µm. Further, 12 studies did not provide mesh information (Figure 1b).

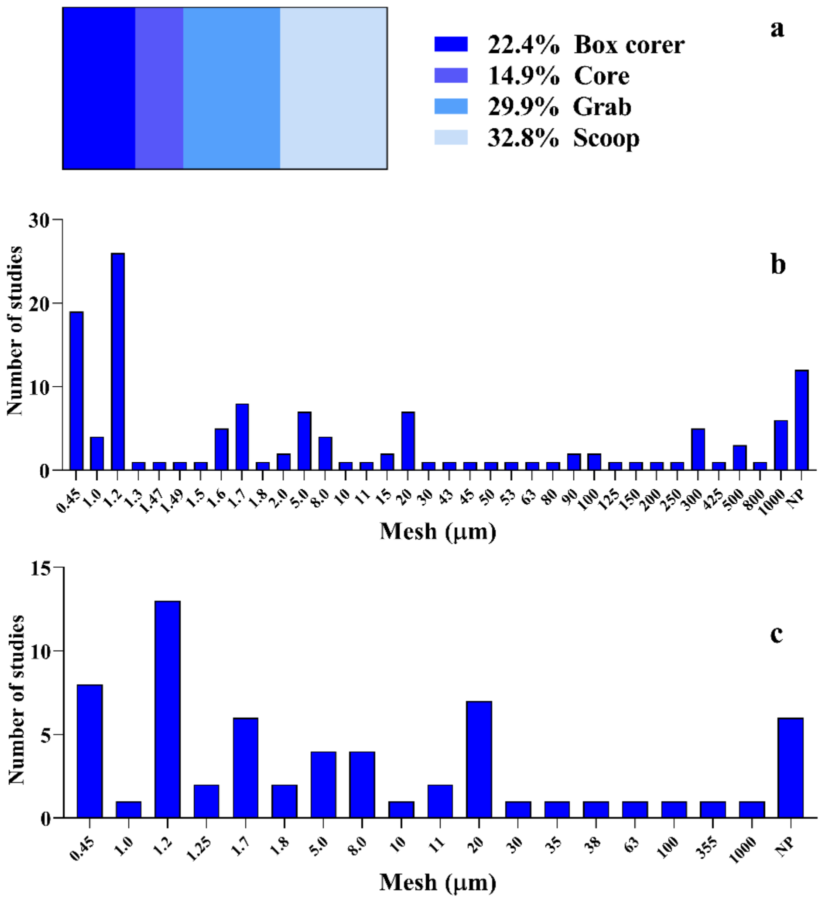

Figure 1: Instruments used to collect sediment samples (a) and distribution frequency of mesh sizes used in microplastic extractions in sediments (b) and biota samples (c).

As expected, wide variation in the sampling methods used to collect biota samples was seen across the 63 studies analyzed. Different approaches were adopted to capture pelagic and benthic organisms including bottom and pelagic trawls, longlines, and manual collection. In this regard, 33 studies (52%) did not detail the sampling method and almost all studies using fish analyzed gastrointestinal contents (Ferreira et al., 2020). Potassium hydroxide was the most used for tissue digestion (Wang et al., 2021), although enzymatic digestion had been used in some studies (Bagheri, 2020; Dahl et al., 2021; Expósito et al., 2021; Stockin et al., 2021). Considering filter porosity, the same range (0.45 to 1,000 µm) and pattern observed for sediment samples was seen for marine biota, with 1.2 µm being the most frequently used (21%) mesh size (Figure 1c). The lack of standardized methods is a recurring topic in MP research, mentioned since at least 2017 (Hanvey et al., 2017). Such methodological variability challenges the search for relevant patterns in studies aiming to synthesize information from the literature, as in the present study. However, data on MP contamination spatialized in MPAs exhibited no bias from method variability.

Overview of MPs in coastal zones

Considering all concentration units (items/kg, items/m^2, items/sample, items/site, and items/organism), the survey resulted in 177 valid MP articles. These results lead to 2,745 records of sediment and/or biota samples analyzed along the coastal zones of 59 countries and 2 International Sites (Figure 2). A total of 1,896 records were found exclusively for sediment and 766 for biota. A total of 83 records were simultaneously observed for both biota and sediments. Most records were reported in Asia (56% - 1529 records) (Table S1), with China accounting for 21% of the total (589 records), followed by India (10% of the total – 279 records), and Iran in the Middle East (7% of the total - 193 records). On the other hand, the north Brazil, the west coasts of the USA and Canada, north Australia, and coastal areas of Africa were not properly covered, due to a lack of research available within the selected time frame (see light gray countries in Figure 2). Based on records reported in sediment samples as items/kg (1609 records), the results ranged from 0 (n = 30) to 148,000 in Japan (Wang et al., 2021). The highest concentrations observed in sediments were sampled around Tokyo, strongly affected by the runoffs during heavy rains (Wang et al., 2021). For biota, MP occurrence ranged from zero (n = 26) to 29,500 items/kg for several taxonomic groups, being more representative in Mollusca (n = 244) and Actinopterygii fish (n = 202). The highest concentrations were found in oysters from Taiwan (29,500 items/kg), collected near famous tourist beaches (Liao et al., 2021). Despite the efforts to cover all countries, studies investigating MP occurrence in coastal areas of North America, Australia, Africa, and Russia were scarce in the time frame surveyed.

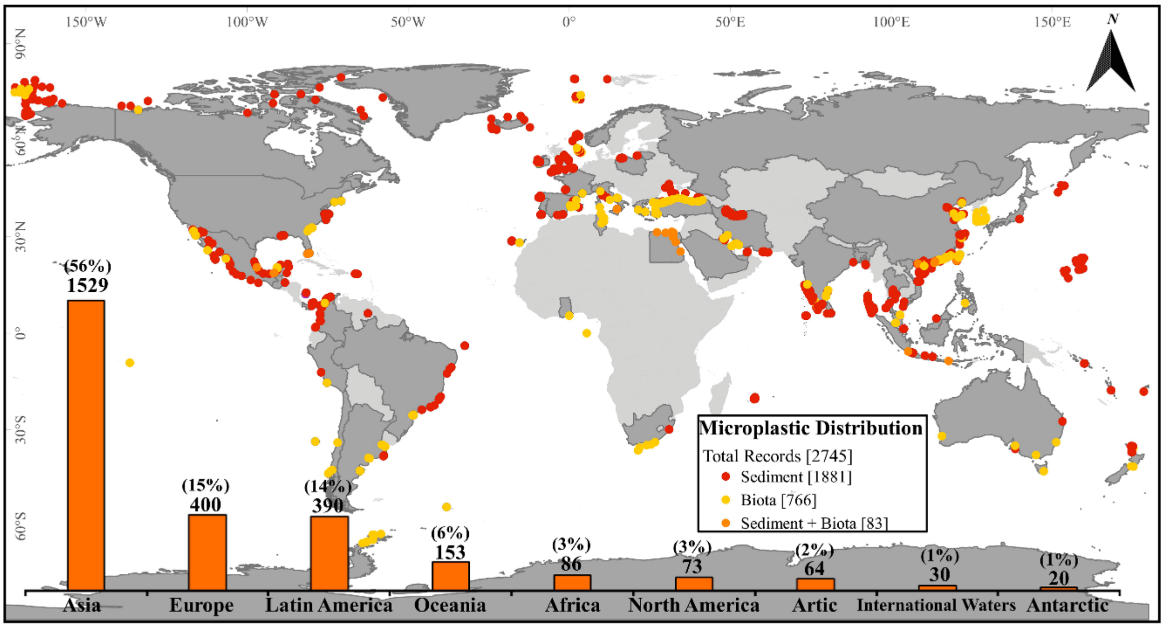

Figure 2: Spatial distribution of microplastic records in sediment and biota samples from coastal areas worldwide between 2006 and 2021.

MPs in MPAs

Based on the outcomes of step 5 (data overlapping with MPAs boundaries) MP occurrence was reported in 186 different MPAs considering data expressed as items/kg, items/m^2, items/sample, items/site, and items/organism. Such findings were obtained from 766 samples, including 557 records of sediments (Supplementary Material), 180 of biota, and 29 of both sediment and biota simultaneously. The affected MPAs were distributed among 31 countries, being more frequent in the USA (14%; n = 106 records), Iran (13%; n = 97), Mexico (10%; n = 76), and Colombia (10%; n = 76) (Figure 3).

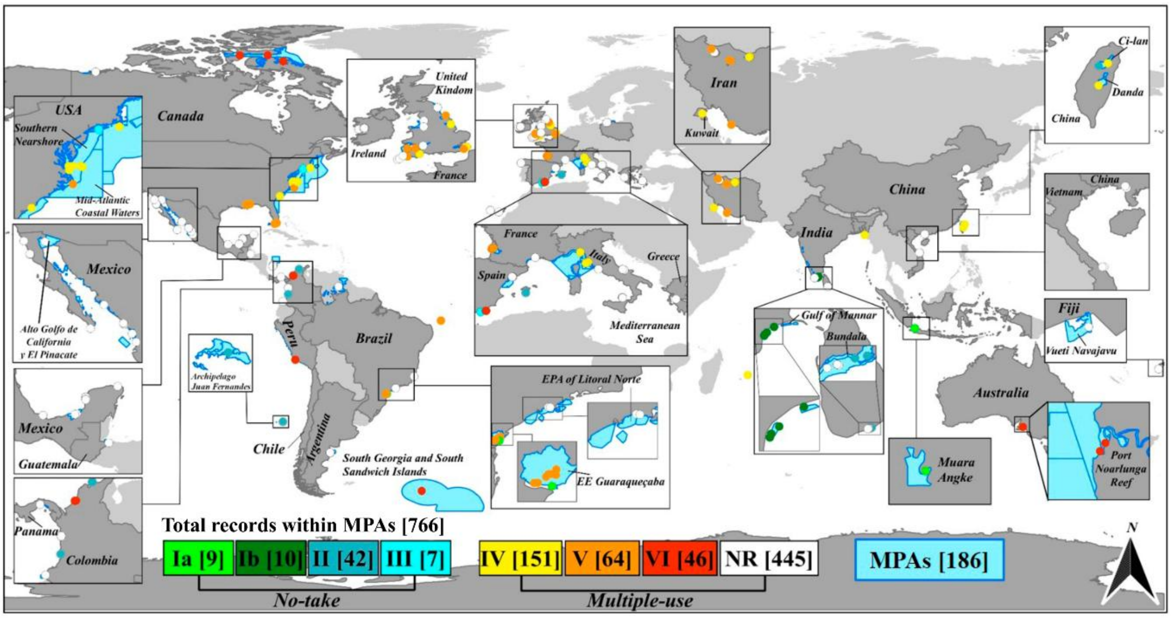

Figure 3: Microplastic records inside Marine Protected Areas boundaries by IUCN categories between 2010 and 2021.

Protected Areas are classified into different categories by IUCN according to their conservation goals and uses, thus clustered into two groups: no-take and multiple-use. No-take reserves are the most restrictive types of protected areas, which include the IUCN categories Ia, Ib II, and III. Multiple-use reserves (IUCN categories IV, V, and VI) aim to conserve specific features, such as sustainable use and knowledge from human interactions via traditional management practices (IUCN, 2013), with moderate effectiveness in maintaining, conserving and restoring species and habitats (Rodríguez-Rodríguez and Sinoga, 2022). Moreover, multiple-use MPAs are important to provide supporting ecosystem services on local and worldwide scales (Roberts et al., 2001). Based on the 766 records of MPs contamination inside MPAs, 66 occurred in no-take areas and 700 in multiple-use areas. No-take MPAs presented 57 records in sediments and 9 in biota, while multiple-use exhibit 500 records in sediments with 171 in biota, and 29 in both. The ranges of MP concentrations reported as items/kg and items/m^2 by management categories were summarized in Table 2.

Table 2 Ranges of microplastic concentrations (items/kg and items/m^2) reported between 2010 and 2021 for sediment and biota samples in MPAs belonging to different categories. NA= Not assigned, NR= Not reported.

IUCN Category		MPAs	Records	Items/kg		Items/m^2
				Sediment	**Biota**	**Sediment**
No-take	**Ia**	**3**	**7**	2 – 6	-	-
	Ib	**1**	**10**	33.3 – 133.2	-	76
	II	**11**	**42**	33.3 – 362	4849.1	2.8 - 791
	III	**4**	**7**	217		279
	Total	**18**	**66**	**2 – 2173**	**7 – 4849.1**	**2.8 - 791**
Multiple-use	**IV**	**31**	**147**	0 – 2224	42.1 – 17,461.9	0.21 - 70
	V	**23**	**64**	0 – 3819	5 – 18	0.26 - 38.7
	VI	**13**	**46**	1 – 3819	0 – 3526.5	12 - 12
	NA/NR	**101**	**443**	0.7 – 9187.5	0.2 – 2176.3	0.21 - 286
	Total	**168**	**700**	**0 – 9187.5**	**0 – 17,461.9**	**0.21 - 286**

MPs in no-take MPAs

Considering 34 records expressed as items/kg within no-take areas, 29 records were found for sediments, while five were found for biota. Sediment concentrations ranged from 2 to 2,173 items/kg while biota exhibited levels between 7 and 4,849 items/kg (Table 3). The affected no-take MPAs were Arrecife Barrera de Posidonia, Archipielago De Cabrera, Gulf of Mannar, Muara Angke, Guaraqueçaba, Shei-pa, Ilha do Mel, Sanctuary Playa El Verde Camacho, and Archipelago Juan Fernadez. These areas were distributed in seven countries: Taiwan, Chile, Brazil, Mexico, Spain, India, and Indonesia (Supplementary material). The highest values found in sediments (2,173 items/kg) were observed in the National Monument (NM) Arrecife Barrera de Posidonia (Spain), and the highest concentrations of MPs reported in no-take MPAs occurred in oysters collected in the NP of Shei-pa, located in the northwestern part of Taiwan Island (4,849 items/kg).

The IUCN management category Ia, known as Ecologic Stations (ES), is managed to ensure minimal disturbance limiting access, excluding settlement by people (IUCN, 2013). Indeed, the lowest levels of MP in sediments were in this category (2 to 6 ±1.5 items/kg) (Table 3). Regarding biota, as an example, MP levels were observed in *Crassostrea gasar* (5 to 18 items/kg) from Paranaguá Estuarine System (PES) (Brazil), which is classified in this category. However, PES has two important harbors, artisanal fishery, and aquaculture installed (Vieira et al., 2021). These areas are under several chemical impacts caused by residues of polycyclic aromatic hydrocarbons (Martins et al., 2015) and butyltin compounds (Castro et al., 2012). Thus, the area has been included among the most impacted estuarine complexes on the Brazilian coast, due to the presence of contamination sources, including MPs (Mengatto and Nagai, 2022).

In contrast, MPAs categorized as Ib are unmodified, or slightly modified areas, that can be used by indigenous and local communities (IUCN, 2013). Nine records ranged from 33 to 133 items/kg of MPs in sediments that occurred in Wilderness Areas of the Gulf of Mannar (India), related to sewage discharges (Patterson et al., 2020). Such areas contemplate four of the seven species of sea turtles found worldwide and are protected under Schedule I of the Indian Wildlife Protection Act (Sivakumar et al., 2018). Despite this, tourism activities are allowed in the buffer zone, even though the anthropic pressure is listed as a preexisting threat to local wildlife (Sivakumar et al., 2018). In this regard, solid waste, including MPs, can be considered a potential threat to conservation goals.

Table 3: Marine Protected Areas affected by Microplastic contamination considering IUCN management categories [IUNC-MC], sample type [Sediment (Sed) and Biota (Bio)], units and concentration ranges. Biosphere Reserve (BR), World Heritage Site (WHS), Environmental Protected Area (EPA)

Marine Protected Area	IUCN MC	Country	Sample Type	Unit	Sed range	Biota range	References
Aeolian Islands	NA/NR	Italy	Sed/Bio	itens/kg	100	100	Renzi et al. 2020
Alborz e Markazy	V	Iran	Sed	itens/kg	188-285		Ghayebzadeh et al. 2020
Al-Kuwaisat (Al-Jahra) Natural Reserve	IV	Kuwait	Sed	itens/site	3		Saeed et al. 2020
Alto Golfo de California y Delta del Río Colorado	VI	Mexico	Sed	itens/kg	9		Alvarez-Zeferino et al. 2020
Alto Golfo de California y El Pinacate	NA/NR	Mexico	Sed	itens/kg	9		Alvarez-Zeferino et al. 2020
Anzali Mordab (Talab) complex	NA/NR	Iran	Sed	itens/kg	113-1216		Rasta et al. 2020
Arcipelago delle Eolie/ Area Marina e Terrestre	NA/NR	Italy	Sed/Bio	itens/kg	99-100	100-116	Renzi et al. 2020
Arkoi; Leipsoi, Agathonisi Kai Vrachonisides	NA/NR	Greece	Biota	itens/kg		150-1957	Malcolm-Mckay et al. 2021
Arrecife Barrera de Posidonia	III	Spain	Sed	itens/kg	2173		Dahl et al. 2021
Arrecifes de Roquetas de Mar	NA/NR	Spain	Sed	itens/kg	2173		Dahl et al. 2021
Arxipèlag de Cabrera	NA/NR	Spain	Sed	itens/kg	68-362		Dahl et al. 2021
Back Bay	IV	USA	Sed	itens/kg	596		Dodson et al 2020
Bahía de Almería	NA/NR	Spain	Sed	itens/kg	2173		Dahl et al. 2021
Bahía de Panamá	NA/NR	Panama	Sed	itens/m^2	105		Delvalle de Borrero et al 2020
Bahía de San Quintín	NA/NR	Mexico	Bio	itens/kg		0.5-6	Lozano-Hernandez et al. 2021
Bahía del Confital	NA/NR	Spain	Bio	itens/sample		1.2-2	Rapp et al. 2021
Ban Dao Son Tra	NA/NR	Vietnam	Sed	itens/kg	9187		Tran Nguyen et al. 2020
Bancs Sableux de l'Espiguette	NA/NR	France	Bio	itens/kg		436	Tsangaris et al. 2020

Bassin d'Arcachon	V	France	Sed	itens/sample	22		Lefebvre et al. 2021
Bassin d'Arcachon et Cap Ferret	NA/NR	France	Sed	itens/sample	35-443		Lefebvre et al. 2021
Los Tuxtlas	VI	Mexico	Sed	itens/kg	13		Alvarez-Zeferino et al. 2020
Bristol Channel Approaches / Dynesfeydd Môr Hafren	NA/NR	England	Sed	itens/m^2	5		Green and Johnson 2020
Cabo de Gata Nijar Natura 2000	NA/NR	Spain	Sed	itens/kg	3819		Dahl et al. 2021
Cabo Manglares Bajo Mira y Frontera	NA/NR	Colombia	Sed	itens/m^2	9		Garces-Ordonez et al 2020
Cabo San Lucas	VI	Mexico	Sed	itens/kg	10		Alvarez-Zeferino et al. 2020
Canaima	II	Canada	Sed	itens/sample	16		Geoffroy et al. 2021
Cape Cod South	IV	USA	Bio	itens/sample		0-0.1	Lato et al. 2021
Cape Hatteras	V	USA	Sed	itens/kg	1351		Dodson et al 2020
Ci-lan	IV	Taiwan	Bio	itens/kg		11351- 17462	Liao et al. 2021
Hancock County Marsh Coastal Preserve	IV	USA	Sed	itens/kg	0-104		Weitzel et al. 2021
Jourdan River Coastal Preserve	IV	USA	Sed	itens/kg	0-16		Weitzel et al. 2021
Hancock County Marsh	V	USA	Sed	itens/kg	0-104		Weitzel et al. 2021
Jourdan River	V	USA	Sed	itens/kg	0-16		Weitzel et al. 2021
Comprensorio Tolfetano-Cerite-Manziate	NA/NR	Italy	Sed	itens/kg	117-280		Piazzolla et al. 2020
Coquet to St. Mary's	NA/NR	England	Sed	itens/m^2	35		Green and Johnson 2020
Cornwall	V	England	Sed	itens/m^2	39		Green and Johnson 2020
Costes del Tarragonès	NA/NR	Spain	Sed	itens/kg	0.7-32		Exposito et al. 2021
Côte languedocienne	NA/NR	France	Bio	itens/kg		436	Tsangaris et al. 2020

Danda	IV	Taiwan	Bio	itens/kg		13899	Liao et al. 2021
Ecologic Station da Ilha Do Mel	Ia	Brazil	Bio	itens/kg		7	Vieira et al. 2021
Ecologic Station de Guaraqueçaba	Ia	Brazil	Bio	itens/kg		9	Vieira et al. 2021
EPA Cabo de Gata-Nijar	NA/NR	Spain	Sed	itens/kg	3819		Dahl et al. 2021
EPA Cinque Terre	IV	Italy	Bio	itens/kg		828	Tsangaris et al. 2020
EPA de Flora y Fauna Laguna de Términos	NA/NR	Mexico	Sed/Bio	itens/kg	6.4-11	2754-3526	Celis-Hernandez et al. 2021
EPA de Flora y Fauna Yum Balam	NA/NR	Mexico	Sed	itens/kg	7		Alvarez-Zeferino et al. 2020
EPA Do Maciço Central	NA/NR	Brazil	Sed	itens/kg	8-32		Zamprogno et al. 2021
EPA Fernando De Noronha	V	Brazil	Sed	itens/kg	50-3703		Carvalho et al. 2021
EPA Guaraqueçaba	V	Brazil	Bio	itens/kg		5-18	Vieira et al. 2021
Epa Marinha Do Litoral Centro	NA/NR	Brazil	Sed	itens/sample	667		Tsukada et al. 2021
Epa Marinha Do Litoral Norte	NA/NR	Brazil	Sed	itens/sample	27		Tsukada et al. 2021
Espacio marino de las Rias Baixas de Galicia	NA/NR	Spain	Sed	itens/kg	13-69		Carretero et al. 2021
Espacio marino del Delta de l'Ebre-Illes Columbretes	NA/NR	Spain	Sed	itens/kg	0.7-41		Exposito et al. 2021
Estero de Punta Banda	NA/NR	Mexico	Bio	itens/kg		0,5-6	Lozano-Hernandez et al. 2021
Exmoor	V	United Kindom	Sed	itens/kg	2-174		Wilson et al. 2021
Exmouth	IV	England	Sed	itens/m^2	31		Green and Johnson 2020
Florida Keys	V	USA	Sed/Bio	itens/sample	1-6	0-7	Plee and C.M. Pomory 2020
Foce Biferno-Litorale di Campomarino	NA/NR	Italy	Bio	itens/kg		1.7-8	Di Renzo et al. 2021
Foce Trigno-Marina di Petacciato	NA/NR	Italy	Bio	itens/kg		9-10	Di Renzo et al. 2021
Fondali dell'isola di Salina	NA/NR	Italy	Sed/Bio	itens/kg	99.6-99.8	100-117	Renzi et al. 2020

Franja marina de Fuencaliente	NA/NR	Spain	Sed	itens/kg	3325-4279		Villanova-Solano et al. 2022
Galway Bay Complex	NA/NR	Ireland	Sed	itens/kg	5-1441		O Briain et al. 2020; Pagter et al. 2020
Golfo de Tribuga Cabo Corrientes	NA/NR	Colombia	Sed	itens/m^2	12-286		Garces-Ordonez et al 2020
Hai Van-Hon Son Tra	NA/NR	Vietnam	Sed	itens/kg	919		Tran Nguyen et al. 2020
Heritage Coast Lundy	V	England	Sed	itens/m^2	5		Green and Johnson 2020
Himchari	IV	Banglades h	Sed	itens/kg	311		Hossain et al. 2021
Humber Estuary	IV	England	Sed	itens/m^2	2		Green and Johnson 2020
Ilot Gabriel	IV	Mauritius	Sed	itens/m^2	4		Mattan-Moorgawa et al. 2021
Indian Key	V	USA	Sed/Bio	itens/sample	2	0	Plee and C.M. Pomory 2020
Inishmore Island SAC	NA/NR	Ireland	Sed	itens/kg	5		Pagter et al. 2020
Inner Galway Bay	NA/NR	Ireland	Sed	itens/kg	5-1441		O Briain et al. 2020; Pagter et al. 2020
Inner Galway Bay SPA	NA/NR	Ireland	Sed	itens/kg	51-1441		O Briain et al. 2020; Pagter et al. 2020
Integrated Management Regional District Manglar de la Bahia de Cispata y Sector Aledano del Delta Estuarino del Rio Sinu	VI	Colombia	Bio	itens/kg		0-181	Garces-Ordonez et al 2020
Integrated Management Regional District Manglar de la Bahia de Cispata y Sector Aledano del Delta Estuarino del Rio Sinu	VI	Colombia	Sed	itens/m^2	12		Garces-Ordonez et al 2020
International significance Natural Marine Area Santuario per i Mammiferi Marini	IV	Italy	Bio/Sed	itens/kg	8-73	828	Mistri et al 2020; Tsangaris et al. 2020
Islands and Protected Areas of the Gulf of California	NA/NR	Mexico	Sed	itens/kg	9,2-14,3		Alvarez-Zeferino et al. 2020
Jezioro Kopań	NA/NR	Poland	Sed	itens/kg	105		Urban-Malinga et al. 2020
Kent Downs	V	England	Sed	itens/m^2	5		Green and Johnson 2020
La Sierpe	II	Colombia	Sed	itens/m^2	152		Garces-Ordonez et al 2020

Lago di Guardialfiera-Foce fiume Biferno	NA/NR	Italy	Biota	itens/kg		2-8	Di Renzo et al. 2021
Laguna de Términos	VI	Mexico	Sed/Bio	itens/kg	6-11	2754-3526	Celis-Hernandez et al. 2021
Lignumvitae Key	IV	USA	Sed/Bio	itens/sample	2	0	Plee and C.M. Pomory 2020
Lisar	V	Iran	Sed	itens/kg	74		Manbohi et al. 2021
Liverpool Bay / Bae Lerpwl	NA/NR	England	Sed	itens/m^2	43		Green and Johnson 2020
Locally Managed Marine Area Vueti Navakavu	NA/NR	Fiji	Sed	itens/kg	100		Ferreira et al. 2020
Los Petenes	VI	Mexico	Bio	itens/kg		2-9	Borges-Ramirez et al. 2020
Manglares y humedales de la Laguna de Sontecomapan	NA/NR	Mexico	Sed	itens/kg	12.6		Alvarez-Zeferino et al. 2020
Marine Conservation Zone Lundy	NA/NR	England	Sed	itens/m^2	5		Green and Johnson 2020
Marine Park Encounter	II	Australia	Sed	itens/sample	5		Hayes et al. 2021
Marine Park Encounter	IV	Australia	Sed	itens/ sample	29		Hayes et al. 2021
Marine Protected Area (OSPAR) Exe Estuary	NA/NR	England	Sed	itens/m^2	31		Green and Johnson 2020
Marismas Nacionales	NA/NR	Mexico	Sed	itens/kg	10		Alvarez-Zeferino et al. 2020
Messolonghi lagoons	NA/NR	Greece	Bio	itens/kg		177	Tsangaris et al. 2020
Miankaleh Peninsula, Gorgan Bay and Lapoo-Zaghmarz Ab-bandan	NA/NR	Iran	Sed	itens/kg	78-741		Bagheri et al. 2020; Manbohi et al. 2021
Mid-Atlantic Coastal Waters Area	IV	USA	Sed	itens/kg	0-2224		Dodson et al 2020; Jones et al 2022
Mond	V	Iran	Bio	itens/sample		2664	Maghsodian et al 2021
Morecambe Bay	NA/NR	England	Sed	itens/m^2	5		Green and Johnson 2020
Mounts Bay	NA/NR	England	Sed	itens/m^2	1		Green and Johnson 2020
Muara Angke	Ia	Indonesia	Sed	itens/kg	2-6		Cordova et al. 2021

Nacional Park Archipielago De Cabrera	II	Spain	Sed	itens/kg	68-362		Dahl et al. 2021
National Key Deer	IV	USA	Sed/Bio	itens/sample	1-2	1-2	Plee and C.M. Pomory 2020
National Park Archipielago Juan Fernández	II	Chile	Bio	itens/kg		1881-2176	Perez-Venegas et al. 2020
National Park Bundala	II	Sri Lanka	Sed	itens/m^2	39-196		Sevwandi Dharmadasa et al. 2021
National Park Gulf of Mannar	Ib	India	Sed	itens/kg	33-133		Jeyasanta et al. 2020; Patterson et al. 2020
	Ib	India	Sed	itens/m^2	76		Jeyasanta et al. 2020; Patterson et al. 2020
National Park Gulf of Mannar	II	India	Sed	itens/kg	33-133		Jeyasanta et al. 2020; Patterson et al. 2020
	II	India	Sed	itens/m^2	76		Jeyasanta et al. 2020; Patterson et al. 2020
Natural Park Cabo de Gata-Níjar	V	Spain	Sed	itens/kg	3819		Dahl et al. 2021
Newquay and the Gannel	NA/NR	England	Sed	itens/m^2	5		Green and Johnson 2020
Nisides Voreion Dodekanison Kai Thalassia Periochi	NA/NR	Greece	Bio	itens/kg		150-1957	Malcolm-Mckay et al. 2021
North Exmoor	IV	UK	Sed	itens/kg	2-174		Wilson et al. 2021
North Northumberland	V	England	Sed	itens/m^2	35		Green and Johnson 2020
North York Moors	V	England	Sed	itens/m^2	3		Green and Johnson 2020
North Yorkshire & Cleveland	V	England	Sed	itens/m^2	3		Green and Johnson 2020
Northern Nearshore	IV	USA	Bio	itens/sample		0-0,1	Lato et al. 2021
Northumberland Marine	NA/NR	England	Sed	itens/m^2	36		Green and Johnson 2020
Northumbria Coast	NA/NR	England	Sed	itens/m^2	35		Green and Johnson 2020
Obszar Chronionego Krajobrazu	NA/NR	Poland	Sed	itens/kg	105		Urban-Malinga et al. 2020
Offshore	IV	USA	Sed	itens/kg	0-67		Jones et al 2022

Other Northeast	IV	USA	Bio	itens/sample		0-0,1	Lato et al. 2021
Pallemalala	IV	Sri Lanka	Sed	itens/m^2	51-70		Sevwandi Dharmadasa et al. 2021
Pascagoula River	V	USA	Sed	itens/kg	0-16		Weitzel et al. 2021
Pascagoula River Marsh	IV	USA	Sed	itens/kg	0-16		Weitzel et al. 2021
Penwith	V	England	Sed	itens/m^2	0.3		Green and Johnson 2020
Playa El Verde Camacho	II	Mexico	Sed	itens/kg	0.4-1.3		Rios-Mendoza et al. 2021
Playa Tortuguera el Verde Camacho	NA/NR	Mexico	Sed	itens/kg	0.4-74		Alvarez-Zeferino et al. 2020; Rios-Mendoza etal. 2021
Port Noarlunga Reef	VI	Australia	Sed	itens/sample	5-29		Hayes et al. 2021
Protected Areas Archipielago De Cabrera	NA/NR	Spain	Sed	itens/kg	68-362		Dahl et al. 2021
Przybrzezne Wody Baltyku	NA/NR	Poland	Sed	itens/kg	98-119		Urban-Malinga et al. 2020
Ramsar Site Bundala	Ramsar	Sri Lanka	Sed	itens/m^2	39-196		Sevwandi Dharmadasa et al. 2021
Ramsar Site Punta de Manabique	Ramsar	Guatemala	Sed	itens/m^2	279		Mazariegos-Ortiz et al. 2020
Reserva de Biosfera Seaflower	BR	Colombia	Sed	itens/m^2	30-152		Garces-Ordonez et al 2020; Portz et al. 2020
	BR	Colombia	Sed	itens/sample	0-120		Garces-Ordonez et al 2020; Portz et al. 2020
Reserva de la Biosfera Los Petenes	BR	Mexico	Bio	itens/kg		2-9	Borges-Ramirez et al. 2020
Reserva Marina de Cabo de Gata-Níjar	VI	Spain	Sed	itens/kg	3819		Dahl et al. 2021
Runnel Stone	NA/NR	England	Sed	itens/m^2	0.3		Green and Johnson 2020
Sandwich & Pegwell Bay	IV	England	Sed	itens/m^2	0.2		Green and Johnson 2020
Sandwich Bay	NA/NR	England	Sed	itens/m^2	0.2		Green and Johnson 2020
Sandwich Bay to Hacklinge Marshes	IV	England	Sed	itens/m^2	0.2		Green and Johnson 2020

Severn Estuary / Môr Hafren	NA/NR	England	Sed	itens/m^2	9		Green and Johnson 2020
Shankou Mangrove Nature Reserve	NA/NR	China	Sed	itens/kg	34-88		Zhou et al. 2020
Shanku Mangrove	NA/NR	China	Sed	itens/kg	88		Zhou et al. 2020
Shei-pa	II	Taiwan	Bio	itens/kg		4849	Liao et al. 2021
Shuangtai Estuary	NA/NR	China	Sed	itens/kg	98-266		Xu et al. 2020
Siahkeshim	V	Iran	Sed	itens/kg	163-470		Rasta et al. 2020
Sistema de Islas, Islotes y PuntasGuaneras	VI	Peru	Bio	itens/kg		711-1522	Perez-Venegas et al. 2020
Sistema Lagunar San Ignacio-Navachiste-Macapule	NA/NR	Mexico	Sed	itens/kg	5		Alvarez-Zeferino et al. 2020
Site of Community Importance Cabo de Gata-Níjar	NA/NR	Spain	Sed	itens/kg	3819		Dahl et al. 2021
Site of Special Scientific Interest Exe Estuary	IV	England	Sed	itens/m^2	31		Green and Johnson 2020
Sorkhanhol	IV	Iran	Sed	itens/kg	153-830		Rasta et al. 2020
Southaven	III	USA	Bio	itens/sample		0.3-3	Lato et al. 2021
Southeast U.S.	IV	USA	Bio	itens/kg		42	Battaglia et al. 2020
Southern Mid-Atlantic Waters	IV	USA	Sed	itens/kg	0-2224		Dodson et al 2020; Jones et al 2022
Southern Nearshore	IV	USA	Sed	itens/kg	0-2224		Dodson et al 2020; Jones et al 2022
Specially Protected Area Seaflower	NA/NR	Colombia	Sed	itens/m^2	30-152		Garces-Ordonez et al 2020; Portz et al. 2020
	NA/NR	Colombia	Sed	itens/sample	0-120		Garces-Ordonez et al 2020; Portz et al. 2020
Specially Protected Areas of Mediterranean Pelagos Sanctuary	NA/NR	Italy	Bio	itens/kg		828	Mistri et al 2020; Tsangaris et al. 2020
	NA/NR	Italy	Sed	itens/site	8-75		Mistri et al 2020; Tsangaris et al. 2020
Tallurutiup Imanga (Lancaster Sound) National Marine Conservation Area	VI	Canada	Sed	itens/kg	1-4		Adams et al. 2021

Tamarit-Punta de la Móra-Costes del Tarragonès	NA/NR	Spain	Sed	itens/kg	0.7-32		Exposito et al. 2021
Tarium Niryutait Marine Protected Area	NA/NR	Canada	Bio	itens/kg		18-147	Moore et al. 2020
Tayrona	II	Colombia	Sed	itens/m^2	3-791		Garces-Ordonez et al 2020
Thanet Coast & Sandwich Bay	NA/NR	England	Sed	itens/m^2	0.2		Green and Johnson 2020
Archipiélago Juan Fernández	BR	Chile	Bio	itens/kg		1881-2176	Perez-Venegas et al. 2020
Biosphere Reserve Los Tuxtlas	BR	Mexico	Sed	itens/kg	13		Alvarez-Zeferino et al. 2020
Biosphere Reserve Miankaleh	BR	Iran	Sed	itens/kg	78-741		Bagheri et al. 2020; Manbohi et al. 2021
Uramba Bahia Malaga	II	Colombia	Sed	itens/m^2	137		Garces-Ordonez et al 2020
Valdés	NA/NR	Argentina	Bio	itens/kg		0.2-3	Rios et al. 2020
Wildlife Refuge Miankaleh	IV	Iran	Sed	itens/kg	78-741		Bagheri et al. 2020; Manbohi et al. 2021
Wildlife Refuge Punta de Manabique	III	Guatemala	Sed	itens/m^2	279		Mazariegos-Ortiz et al. 2020
World Heritage Site Western Ghats	WHS	India	Sed/Bio	itens/kg	86	100-590	Cheng et al 2021
Yum Balam	VI	Mexico	Sed	itens/kg	7		Alvarez-Zeferino et al. 2020

National Parks (NP) represent category II providing ecological corridors at large-scale, strengthening thus biodiversity connectivity (IUCN, 2013). Seventeen records ranging from 33 to 362 items/kg were found in sediment samples from Archipielago de Cabrera (Spain), Gulf of Mannar (India), Archipelago Juan Fernadez (Chile), Sanctuary Playa El Verde Camacho (Mexico) and Shei-pa (Taiwan). The highest concentrations of MPs in sediments were reported in the Archipelago de Cabrera, formed by small islands in the Mediterranean Sea. Although the archipelago has been protected for over 30 years, a direct relationship with the increasing anthropogenic pressures on the coastal environment was demonstrated. In this MPA, marine litter composed mainly of shopping bags and plastic bottles was found on the seafloor and associated with tourism and recreational activities (Compa et al., 2022). As expected, high MP contamination was also found by Dahl et al. (2021), which used core samples to reveal increased sedimentary accumulation of MPs in the past 50 years. Thus, the lower restriction to visitation and recreation activities, often adopted in NPs, is probably leading to MP contamination rates slightly higher than MPAs included in Ia and Ib categories (Pongpattananurak, 2018).

MPAs designed to protect a specific natural monument are included in category III, also called Natural Monuments (NM) or Features. In this category, the NM Arrecife Barrera de Posidonia (Spain) presented the highest MPs concentration in sediment samples from no-take categories (2,173 items/kg) (Dahl et al., 2021). Since this MPA was implemented to ensure the safety of one of the few Posidonia reefs, the occurrence of MP in sediments at high levels can cause physiological disturbances on corals reefs, harming the conservation objectives of this important heritage (Day et al., 2012; Liao et al., 2021; Reichert et al., 2021). Also, the region presents a highly degraded environment caused by inputs of hazardous residues from agricultural and industrial sources located in the vicinity of the Almería region (Dahl et al., 2021). Posidonia is a highly complex marine ecosystem sheltering endemic species in the Mediterranean Sea, such as the grass benches of *Posidonia oceanic.* The area also shelters populations of *Pinna nobilis,* a mussel species classified as Critically Endangered according to IUCN criteria, which are in decline throughout the Mediterranean due to habitats suppression, and accidental or intentional capture (Fernández-Salas, 2012).

MPAs included in no-take categories have shown that conservation areas located in the vicinity of large centers with public visitation allowed, exhibited higher status of MP contamination. As pointed out by Liao et al. (2021), MPAs close to urban areas are under direct anthropogenic impacts, which contribute to contaminants inputs able to induce adverse effects on organisms. In these cases, conservation goals may also be impaired. These findings, already reported by Cordova et al. (2021), Perez-Venegas et al. (2020), and Vieira et al. (2021) highlight the urgent need to support proper waste management as well as educational campaigns for visitors, seeking to protect vulnerable coastal environments from MP impacts.

MPs in multiple-use MPAs

Multiple-use MPAs presented 700 records of MP occurrence considering sediments and biota samples. Most of those records were within Not Reported or Not Assigned Categories (n = 443; 63%) followed by categories IV (n = 147; 21%), V (n = 64; 9%), and VI (n = 46; 7%). According to the dataset (Table 3), surveys were performed in 169 multiple-use MPAs, and in 82 of them MP contamination levels were not found. Records of MP in sediments ranged from 0 to 9,187 items/kg (n = 325). Vietnam (9,187 items/kg) followed by the Canary Islands (4,278 items/kg) presented the highest levels of MP in sediments. In Da Nang Bay (Vietnam), the highest levels were attributed to river discharges carrying MP fibers released from domestic and industrial wastewater (Nguyen et al., 2020). On the other hand, in Puerto Naos beach (Canary Island), the contamination by fibers was associated with wastewater discharges combined with atmospheric deposition since this Spanish territory has a very limited presence of industries (Villanova-Solano et al., 2022).

Considering MPs records based on biota samples, several species belonging to different taxonomic groups were surveyed in multiple-use MPAs, such as Mollusca (61), Actinopterygii fish (48), Echinodermata (41), Plantae (18), Mammalia (13), Aves (7), Chelonia (6), Porifera (4) and Cnidaria (2). Bivalve species (oysters and mussels) were the most frequently used sentinels. The use of bivalve species has been proven to be an excellent tool for environmental monitoring, including MP assessments (Kazour and Amara, 2020). The use of such models has been increasing, mostly because they are widely distributed in coastal zones, abundant, easily collected, and important seafood items (Curpan et al., 2022). Actinopterygii fish, the second most accessed taxonomic group, have also been widely used as sentinels of MPs contamination. In these organisms, MPs residues were studied using both, inedible (Gastrointestinal tract - GIT) and edible tissues (muscle) (Daniel et al., 2020). The MPAs in categories IV, V, and VI often act as buffers or links among different reserves, conserving and restoring species and habitats. The sustainable use of the land is encouraged in these areas, which can also release hazardous materials and substances. The highest sediment concentrations of MPs were found in these areas (3,819 items/kg). In biota, the same scenario was found, with increased concentrations of MP observed in Mollusca, hitting 17,461 items/kg.

Even matching the standard definitions of protected areas, MPAs may be categorized by IUCN as Not Reported (NR) or Not Assigned (NA) due absence of specific information provided or by the choice of managers (UNEP-WCMC, 2016). Since the IUCN Management Categories system is not adopted by all countries, some specific designation types, such as World Heritage Sites, UNESCO Reserves, and Ramsar sites, are not reported to the WDPA (UNEP-WCMC, 2016). Therefore, the specific conservation goals or general objectives are attributed individually. A total of 62 NR and NA MPAs were affected by MPs contamination, accounting for 57% of all protected areas. From these totals, 25 were classified as NR (n = 112), 15 as Ramsar Sites (n = 114), 10 as NA (n = 19), 6 as UNESCO Biosphere Reserve (n = 34), 3 as World Heritage Site (n = 10), 2 as OSPAR (n = 17) and 1 as Natura 2000 (n = 1). The Natural Reserve Ban dao Son Tra (NR), in Vietnam, had the highest values in sediments (9,187 items/kg), while the Ramsar Site Laguna de Términos, in Mexico, exhibited the highest concentration in biota, 3,526 items/kg in sponges (*Haliclona implexiformis*). For this site, biodiversity is affected by pollution and the reduction of stocks because of fishing, agriculture, and livestock, combined with deforestation in the region of Laguna de Términos (Área de Protección de Flora y Fauna Laguna de Términos, 2004).

The results of this study revealed that MP contamination also affects various organisms in multiple-use MPAs (Celis-Hernández et al., 2021; Cheng et al., 2021; Plee and Pomory, 2020; Renzi et al., 2020). As for MPs intake by marine organisms, studies suggest that MPs are mostly ingested by fish (Lin et al., 2020). Recent studies on MP ingestion in oysters concluded that MPs could cause histopathological alterations in the gill and digestive gland, inducing inflammatory responses (Teng et al., 2020). Also, Bom et al. (2022) correlated negative effects on bivalve weights with MP concentrations in the environment. As a sublethal response, oxidative stress induction in mussels has also been reported by Provenza et al. (2022).

It was also notable the low number of records in no-take MPAs (n = 34) compared with multiple-use MPAs (n = 474), in addition to the same trend of contamination status, considering the highest MP concentrations reported in sediments from these areas (2,173 items/kg in no-take and 9,187.5 items/kg in multiple-use MPAs). Such divergence can be explained by the different flexibility in the restrictions adopted in each management strategy. In this case, regardless of the protection category, diffuse sources of MPs in their surrounding areas are related to cities wastewater discharges, atmospheric deposition, riverine inputs, run-off, fishing, tourism, agricultural and livestock(Fernández-Salas, 2012; Kutralam-Muniasamy et al., 2021b; Liao et al., 2021; Pongpattananurak, 2018; Sivakumar et al., 2018; Vieira et al., 2021).

Threatened species by microplastic contamination and Ecological attributes

Levels of MPs in tissues were found in 277 species, belonging to Actinopterygii fish (200), Mollusca (28), Decapoda (22), Echinodermata (7), Aves (5), Polychaeta (5), Mammalia (4), Cnidaria (3), Chelonia (1), Plantae (1) and Porifera (1). Two species (*Notarius bonillai* and *Caretta caretta*) contaminated by MPs are considered threatened according to the currently available IUCN Red List. The Cazon Sea Catfish (*N.bonillai*) is an endangered species endemic to marine and fresh waters on the coast of the Caribbean Sea of Colombia (Betancur-R. et al., 2007; Garcés-Ordóñez et al., 2020; Santana et al., 2019). MP contamination in tissues of *N. bonillai* was found in the Integrated Management Regional District Manglar de la Bahia de Cispata y Sector Aledano del Delta Estuarino del Rio Sinu in Colombia (Garcés-Ordóñez et al., 2020), with a concentration of 2.56 items/kg in the gastrointestinal tract. Studies had shown that MP exposure in fish can cause reduced feeding and interrupted digestion, histological alterations in the gastrointestinal tract, reduced gill functioning, neurotoxicity, reduced immunity, and impaired reproduction (Mallik et al., 2021; Tongo and Erhunmwunse, 2022; Yu et al., 2022). Apart from the physical impact, MPs can act as carriers of various toxic and bioaccumulative chemicals. It was found that hydrophobic contaminants can be transferred in the trophic chain much more effectively due to the ability to get adsorbed onto the plastic (Wardrop et al., 2016).

Sea turtles are considered fitting organisms for the investigation of the spatial exposure of MPs due to their migratory habits (Di Renzo et al., 2021). Di Renzo et al. (2021) studied the MP accumulation in 28 individuals of Loggerhead Turtle (*Caretta. caretta*) from

Italy. Six of them were sampled inside the MPAs, including Site of Community Importance Foce Biferno - Litorale di Campomarino (1), Site of Community Importance Foce Trigno - Marina di Petacciato (3), and Special Protection Area Lago di Guardialfiera (2). Gastrointestinal tracts of *C. caretta* exhibited MP concentrations ranging from 1.74 to 9.59 items/kg (n = 6). This species was classified as globally Vulnerable due to several threats leading to its population decrease, as a result of solid waste and litter exposure, effects of climate change, invasive species, and health disturbances at individual levels (e.g., diseases) (Casale and Tucker, 2017). Also, recent findings in Italy reported the occurrence of MP acting as a carrier of organic pollutants (Capriotti et al., 2021) that can be bioavailable following its ingestion. According to Gola et al. (2021), plastic ingestion may induce damage to the digestive tract, such as a reduction in stomach capacity and toxicity including alteration in swimming behavior, immunologic performance, growth rate, and ability to escape.

As for the ecological attributes, assessed only for records expresses as items/kg, 230 records were found for pelagic species while 439 records were found for benthic organisms. According to the feeding preferences of pelagic fauna, 7 records of MP contamination were found for herbivores, 74 for omnivores, and 149 for carnivores. For benthic fauna, 36 records were found for herbivores, 64 for detritivores, 274 for omnivores, and 65 for carnivores (Figure 4). In general, mean values of MP levels in tissues of benthic species were higher compared to pelagic organisms, with herbivores (627.4 ±71.87 items/kg) and omnivores (1007 ±193.2 items/kg) presenting elevated values (mean ±standard error). Such patterns may be related to the different traits of benthic organisms, once they interact directly with sediments, which are considered sinks of MP, even with its sinking mechanisms not thoroughly comprehended (Phuong et al., 2021; Zhang et al., 2022).

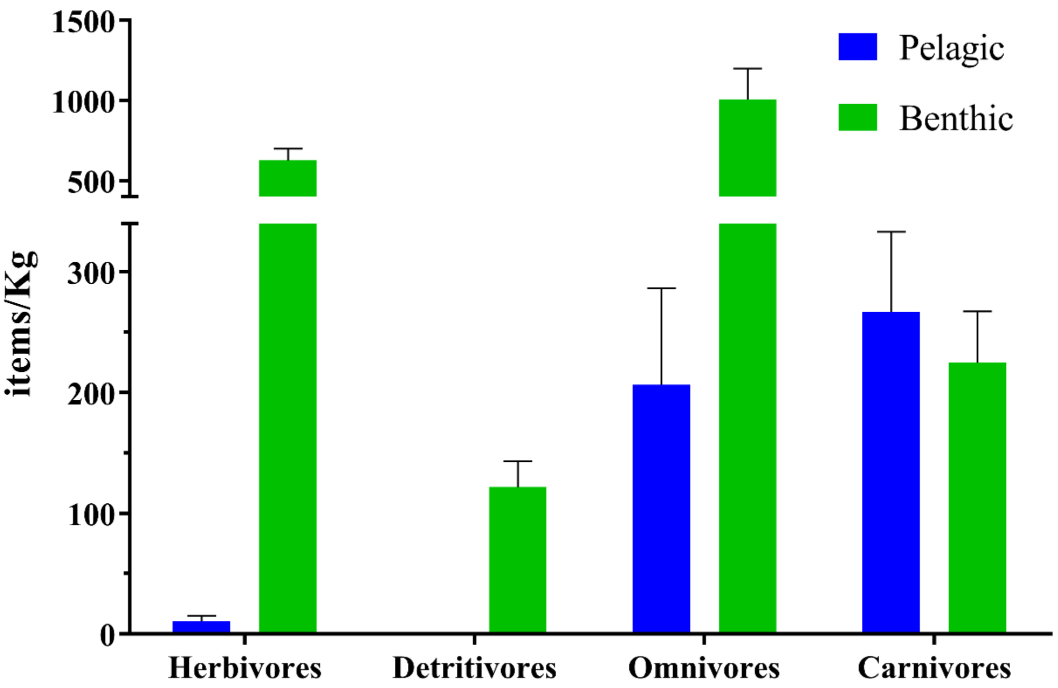

Figure 4 Concentrations of MPs detected in tissues of benthic and pelagic organisms from different feeding types. Values expressed as items/Kg (mean ±standard error)

The results of MP contamination across feeding preferences of pelagic organisms suggest a possible trophic transfer as one of the major pathways of MP ingestion of marine biota, from primary consumers (direct ingestion) to top predators (indirect ingestion of contaminated prey), especially for those species that feed on whole prey (Nelms et al., 2018). At a low-trophic level, under controled conditions, the mysid *Neomysis* spp ingested 2 to 3 more polyethylene beads from contaminated water, with elevated levels of fragments, indicating a fragmentation caused by the species (Hasegawa and Nakaoka, 2021). In the same study, the carnivore fish *Myoxocephalus brandti* ingested 3 to 11 times more polyethylene beads from contaminated mysids than from contaminated water, corroborating the mechanism of MP trophic transfer as a relevant issue. However, other authors recognized the evidence of biological ingestion of MP but highlight that they do not bioaccumulate and cannot biomagnify because of the trophic transfer, since most field observations report MP particles retained in the digestive system such as in the gastrointestinal tract (Gouin, 2020).

Quartile analysis

Considering the complete dataset, a total of 1,722 records expressed as items/kg were found for sediment and 588 for biota, with 350 and 159 reported within MPAs boundaries, respectively. The quartile analysis resulted in the four concentration intervals for sediments: Q1 (0.1 to 22 items/kg), Q2 (22 to 106.7 items/kg), Q3 (106.7 to 362.1 items/kg) and Q4 (362.1 to 145,435 items/Kg) (Figure 5a). Eight hundred and ninety records, 51.4% of the complete database, fell into the highest quartiles (Q3 and Q4) affecting 42 MPA categorized as no-take (Ia, Ib, II and III) and multiple-use (IV, V, VI, NA, and NR). Among these MPAs are the region of Muara Angke (Indonesia), National Parks of Gulf of Mannar (India), Archipelago de Cabrera (Spain), and Sanctuary Playa El Verde Camacho (Mexico). These MPAs are unique and highly vulnerable ecosystems and are classified as no-take categories. Although they play an important role in protecting coral reefs and mangrove areas, by acting as a sanctuary for faunal and floral diversity (from migratory sea birds to breeding centers for sea turtles) (Dahl et al., 2021; Jeyasanta et al., 2020; Rios-Mendoza et al., 2021), these areas are also close to metropolitan centers and port facilities which are potential contamination sources as already mentioned (Cordova et al., 2021; Patterson et al., 2020).
Similarity, quartiles were determined for biota as follows: Q1 (0.01 to 2.6 items/kg), Q2 (2.6 to 29.8 items/kg), Q3 (29.8 to 290.4 items/kg), and Q4 (290.4 to 29,500 items/kg) (Figure 5b). The same pattern was observed in organisms, with 50.4% of records of the entire dataset associated with Q3 and Q4 (29.8 to 29,500 items/kg). In Q4, the species presenting the highest MP levels was the oyster *Saccostrea sp*., collected in two Major Wildlife Habitats from China (Category IV). High MP concentrations (Q4) were also seen within the National Parks of Shei-pa and Archipelago Juan Fernadez, both belonging to no-take category II. Furthermore, organisms collected in 47 no-take and multiple-use MPAs presented measurable levels of MP contamination.

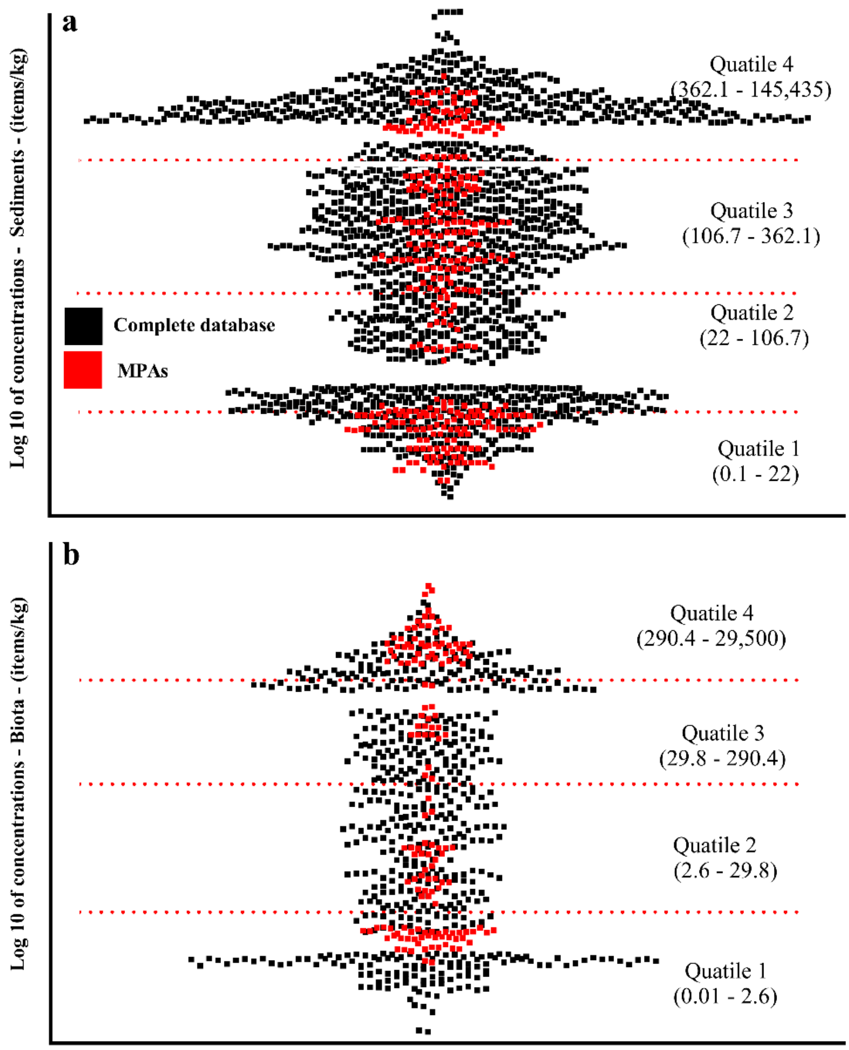

Figure 5: Quartiles of microplastic concentrations in items/kg for Sediment (a) and Biota (b) samples. Black dots indicate the complete database of MPs; red dots indicate microplastic records within MPAs.

Indeed, MPs have been found in different environmental matrices at low to high concentrations, even inside marine protected areas. This is mainly due to the high mobility of MPs that can be transported by the atmosphere and hydrodynamics reaching sediment layers and organisms of several feeding types (Kane et al., 2020). Around half of the available data showed high MP levels in MPAs (Figure 5). Although there are not yet established safe limits for MP exposures, several deleterious effects have been observed in organisms exposed to MPs (Missawi et al., 2020; Xu et al., 2020). Such effects have even been reported at population and community levels, causing ecosystem imbalances and threatening biodiversity (Khalid et al., 2021b). Moreover, considering that chemical contamination and consequent biological effects are already affecting MPAs worldwide (Abessa et al., 2018; Castro et al., 2021; Nunes et al., 2021), the global conservation goals based only on spatial features may not be effective. Therefore, international efforts focused on preventing and mitigating marine pollution by MPs are urgently needed (Borrelle et al, 2017), and the outcomes of this study can serve as a baseline for ecological risk assessment and management of this global environmental issue.

It should be fully acknowledged that further studies are needed to fill the current knowledge gaps of MP occurrence in global MPAs. As pointed out by Nunes et al. (2023), many previous MPA studies reporting the environmental occurrence of MPs do not provide information on the shape, size, and polymeric compositions of MPs. In the present review, we find that data on MP polymeric composition was provided only for 2 MPAs, while shapes of MPs were reported in only 4 MPAs. MPs with different characteristics could manifest different environmental fates and biological impacts. For example, denser MPs preferentially accumulate in sediment layers (Abel et al., 2021), while sizes, shapes, and surface charges of MPs also affect the exposure pathways and toxic effects of MPs on organisms (Borges-Ramírez et al.,2020; Ding et al., 2022; Matthews et al. 2021). In addition, the concentrations of MPs reported in different studies using non-harmonized methods for sampling, digestion, flotation, and separation may largely influence spatial comparison and conclusion (Nunes et al., 2023).

Conclusions

MP contamination was reported in 186 global MPAs belonging to no-take and multiple-use management categories in at least one environmental matrices (sediment and biota). Higher levels of MPs were found within multiple-use and not categorized MPA, which are less restrictive to anthropic interactions and located near urban centers. Mollusks were the most frequently used group to assess MP contamination, among affected organisms. Alarmingly, MP residues were found in tissues of two threatened species according to IUCN Red List, including an endangered fish (*Notarius bonillai*) and a vulnerable sea turtle (*Caretta caretta*). The assessment of MP contamination based on ecological attributes showed that MP levels in tissues of benthic species were higher than in pelagic organisms. Although there are still no guidelines for the safety levels of MP in different environmental matrices, the analysis based on quartiles showed that half of MP levels found within MPA fell in the high-concentration quartiles, rasing concerns on the effectiveness of the global system of MPAs and their proposed conservation goals.

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Microplastic in no-take Marine Protected Areas from Brazil: bivalves as sentinels

Microplastic in no-take Marine Protected Areas from Brazil: bivalves as sentinels

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Abstract

Microplastics (MPs) are pervasive environmental contaminants even in remote and pristine locations. Despite relatively rich literature revealing MPs to be omnipresent in marine environments, their occurrence in Marine Protected Areas (MPAs) is less documented, particularly in developing countries. Bivalves have been heralded as effective sentinels for quantitative and qualitative monitoring of MP contamination. Here, MP contamination was assessed in ten no-take MPAs in Brazil, using several bivalve species as sentinels. In addition, an extensive comparative analysis was conducted with the scientific literature to grade the level of contamination. MPs were found in bivalves from all studied areas with higher levels found in natural monuments managed within International Union for Conservation of Nature management category III, while those from strict nature reserves (category Ia) were less contaminated. Moreover, no-take MPAs had lower MP concentrations than multiple-use MPAs or unprotected areas when compared to the literature. Across all studied MPAs, the majority of particles associated with bivalve tissues were black, white or transparent cellulosic fragments <1000 µm in size. Although MPAs do afford some level of protection against MP contamination, yet further efforts are required to meet the 11th Aichi Target. The contamination levels found are considered moderate warranting long-term monitoring of MP occurrence in these vulnerable areas and the ecological consequences.

Keywords: polymer; contamination; conservation; monitoring; management

1. Introduction

Pollution is a primary driver of the triple planetary crisis, which also includes climate change and biodiversity loss (Almroth et al., 2022). It results from the intentional or unintentional release of substances and/or hazardous materials into natural environments (Tilman et al., 2001). These enter the oceans and seas mostly from continental sources (Cardoso and Caldeira, 2021) and are associated with industrial, port, domestic, mining, agricultural, or tourist activities (Angeli et al., 2021). Of the contaminants of emergent concern often found in aquatic systems, solid waste, particularly plastic, has been identified as a pervasive environmental stressor for marine and coastal zones (Haarr et al., 2022).

The environmental impacts of plastics are related to their widespread use, and are a direct result of inadequate disposal and the lack of effective management mechanisms (Dimassi et al., 2022; Islam et al., 2024). Poorly managed plastic waste can enter into the oceans and be exposed to physical friction, wave action, UV radiation and oxidation, and even interact with organisms potentially resulting in biofouling (Fazey and Ryan, 2016) and/or ingestion (Du et al., 2022). These processes, either singularly or in combination, can lead to fragmentation into smaller plastic particles and microplastics (MPs; those measuring between 1 μm and 5 mm), which then become increasingly bioavailable to a wider range of organisms (Botterell et al., 2019). There is now extensive literature reporting on the omnipresence of MPs in natural systems, with MPs detected even in remote and protected areas around the world (Nunes et al., 2023b, 2023a). The environmental distribution of MPs is strongly influenced by socioenvironmental factors, with urban rivers often serving as links between terrestrial and marine systems (Perumal and Muthuramalingam, 2022). These areas, characterized by high population density and frequent urban runoff and effluent discharge, expose local ecosystems and biota to significant MP contamination (Parker et al., 2022).

Indeed, river discharge is widely recognized as a primary source of MPs to coastal areas, and, under the influence of tides, currents, and other oceanographic forcings, can be further distributed to adjacent environmental compartments (Dimassi et al., 2022).

Microplastics can induce deleterious effects in marine species, for example, negatively impacting growth and reproduction rates, and overall survival (Ugwu et al., 2021). Moreover, long-term ecological consequences, such as changes in energy budgets and impairments in nutrient cycling have been attributed to MPs and can threaten ecosystem stability (Ma et al., 2020). In coastal environments, bivalve molluscs are particularly vulnerable to MPs exposure given their sessile nature and feeding habit, i.e., filtration of the water (Baroja et al., 2021). Significant MP concentrations can accumulate in their soft tissues, and some species of clams, mussels, and oysters have been identified as suitable sentinels of MP contamination (Bom and Sá, 2021a; Ribeiro et al., 2024a). In this regard, a study assessing global data on MPs occurrence in Marine Protected Areas (MPAs) demonstrated extensive contamination in filter-feeding bivalves (Nunes et al., 2023b). Such findings suggest that MPAs, which are legally designated zones seeking to conserve marine biodiversity (Edgar et al., 2014), may be failing to achieve established environmental goals due to the impacts of ever increasing pollution (Abessa et al., 2018; Castro et al., 2021; Nunes et al., 2024, 2023c).

The 11th Aichi Target established during the 10th Convention on Biological Diversity (COP-10) in Nagoya, Japan, has the aspiration goal of protecting ocean biodiversity from anthropogenic-driven stressors and, in response, many countries are now implementing and expanding their protected area management strategies (IPBES, 2019). Yet despite global efforts, over half of the global MPAs have failed to meet this target (Ohayon et al., 2021). The 11th Aichi Global Target has now been incorporated into the 2030 Agenda for Sustainable Development, with a stronger focus on conservation and sustainable use of the oceans, seas, and marine resources, addressing issues such as pollution, ocean acidification, and sustainable fishing (UNEP-WCMC, 2021). The primary objectives to achieve this are to reduce anthropogenic threats, including overexploitation of natural resources, habitat degradation, uncontrolled tourism, coastal development, and pollution (Davidson and Dulvy, 2017). Despite this intent and the significant progress made to address mismanaged waste (OECD, 2022) no systematic studies assessing MPs contamination in MPAs have so far been performed in Brazil, the fifth largest country in the world and producer of 7.1 million metric tons of plastic per year (Do Amparo et al., 2023; Statista, 2022; Zamora, A. M. et al., 2020). Compounded by limited waste management infrastructure, Brazil contributes 79 million metric tons of plastic waste annually and is ranked among the top plastic polluters globally, with a substantial amount of plastic leakage into natural environments (Pincelli et al., 2021). This ranking emphasizes the need to assess the presence and impact of microplastics in Brazilian MPAs, zones that are crucial for biodiversity conservation.

Marine Protected Areas (MPAs) are categorized by the International Union for Conservation of Nature (IUCN) to serve different purposes and objectives. Human activities are restricted or entirely prohibited in no-take MPAs, categorized as nature reserves (Ia), national parks (II), and natural monuments (III). The restriction changes gradually across the categories, in which Ia is the most restricted, followed by Ib, II and III where the restriction is more flexible, although all is considered no-take. In contrast, sustainable-use MPAs allow some level of human activities within their boundaries (Dudley, 2008). Consequently, no-take MPAs are effective tools for preserving marine and coastal biodiversity, along with their associated environmental aspects (Costello and Ballantine, 2015). In Brazil, there are currently 57 no-take and 122 multiple-use MPAs, representing approximately 26.8% of national marine zones (UNEP/WCMC, 2023), however, contamination and pollution effects are often neglected in the management plans of these zones (Moreira et al., 2021a) mainly because there is little information available (Abessa et al., 2017; Moreira et al., 2021b, 2017; Nunes et al., 2021). Therefore, there is an urgency to conduct systematic assessments of contamination within these MPAs to identify potential pollution sources and guide management strategies to enact effective mitigation and negate any adverse impacts. Given these issues, the present study uses filter-feeding bivalves species as sentinels to establish baseline MP contamination in ten no-take MPAs along the Brazilian coast and includes comparative analyses with existing literature to assess contamination levels to infer impacts across Brazil's entire marine estate.

2. Materials and Methods

2.1 Study Area and sampling

A total of ten no-take MPAs from different IUCN management categories (Ia, II and III) were selected along the Brazilian coast (Figure 1). The 2022 sampling campaigns were performed in the Abrolhos National Park (ANP), Alcatrazes Archipelago Wildlife Refuge (AAWR), Arvoredo Biological Reserve (ABR), Atol das Rocas Biological Reserve (ARBR), Carijós Ecological Station (CES), Fernando de Noronha National Park (FNNP), Frades River Wildlife Refuge (FRWR), Guarequeçaba Ecological Station (GES), Jericoacoara National Park (JNP) and Tamoios Ecological Station (TES) under the Brazilian environmental SISBIO license No. 64646-3. Whenever possible, three sites were sampled in each MPA seeking to archive local representativity (Figure 1). At each selected site, twenty to thirty adults of oysters (*Chama sinuosa*, *Crassostrea brasiliana*, *Ostrea equestris*) and mussels (*Perna perna*) were sampled by scuba diving or snorkeling (Table S1). Specimens were immediately stored in decontaminated (see Section 2.2.1) and refrigerated aluminum boxes to avoid cross-contamination until further laboratory analysis.

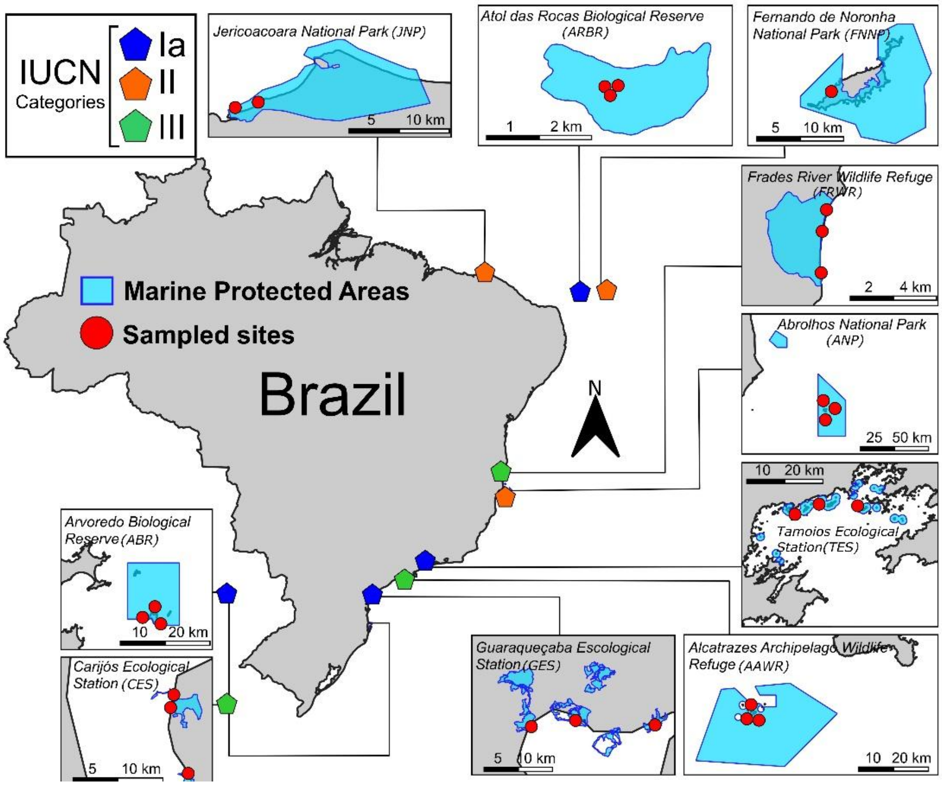

Figure 1. Map of Brazil showing the ten no-take marine protected areas (MPAs) and local sites from which filter-feeding bivalves were sampled to assess microplastic contamination.

2.2 Laboratory procedures

The collected organisms were identified to species and biometric parameters (shell length, width and height, and weight of wet tissues) were measured using a digital caliper and analytical balance. The complete list of species name, shell length (52.6 ±16.6 mm), coordinates, MPA site details, sample data and IUCN category is available in Table S1. After, integral tissues (muscle, soft organs, gill, mantle) of each specimen were transferred to decontaminated glass vials, pooled according to each MPA, and stored at −20°C then freeze-dried to facilitate further digestions. Dried tissue samples were homogenized, macerated, and separated into at least 5 pools of approximately 5 g. The chemical digestion was carried out according to Ribeiro et al. (2024a), using 10 ml of 10% potassium hydroxide (KOH) solution per gram of tissue, and vials were incubated at 40°C for 48–96 hours. Digested samples were vacuum filtered through 1.2 μm fiberglass membranes which were then transferred to pre-cleaned Petri dishes and dried at 40°C for 24 hours (Ding et al., 2021).

The MPs quantification was performed by visual inspection of filters under stereomicroscopy with a 40x magnification. Particle size was measured using AxioVision (Zeiss, Germany) software and classified according to the following MPs size categories: 0–1,000; 1,000–2,000; 2,000-3,000; 3,000–4,000; 4,000–5,000; >5,000 µm (Bom and Sá, 2021). Shapes (fibers, fragments, pellets, films) and colors of MPs were also catalogued (Jankauskas et al., 2024a; Naji et al., 2018). Subsequently, each particle was transferred to glass plates and individually analyzed to determine the polymer composition using Attenuated Total Reflectance Fourier transform infrared (ATR-FTIR) spectroscopy operating in ATR mode (PerkinElmer Spectrum 100; 1 mm ATR window, pressure gauge = 150, atmospheric [CO_2/H_2O] suppression and atmospheric vapor compensation, and background scans acquired hourly). The recorded spectra were compared with NICODOM IR libraries (ATR): Polymers and Additives, Coatings, Fibres, Dyes and Pigments, Petrochemicals; NICODOM Ltd., Czech Republic. Polymers were identified positively in the database when the match coefficient was higher than 70% (Jankauskas et al., 2024a; Kroon et al., 2018). Finally, particles presenting a polymeric match coefficient lower than 70% were not further considered.

2.2.1 Quality assurance and quality control

To avoid cross-contamination during laboratory procedures, where possible only glass or metal utensils were used. Moreover, all aluminum foils, metal and nonvolumetric utensils and glassware used were calcined (400^{o}C, 4 hrs) and then washed twice with filtered ultrapure water (1.2 µm). Water and digestion solutions were filtered (1.2 µm) and stored in pre-calcinated glass containers. Laboratory users wore 100% cotton lab coats (Enders et al., 2020). Digestion, filtration, and MPs separation steps were conducted under laminar flow, with exposed control filters positioned adjacent the workspace to monitor for potential airborne contamination (Prata et al., 2021). Finally, procedural blanks were regularly performed in-line with real sample batches. Concentrations below 1 particle.filter^{-1}.hour^{-1} for exposed filters and 1 particle.filter^{-1} for procedural blanks were considered acceptable (Bruzaca et al., 2022; Ribeiro et al., 2023; Thiele et al., 2019).

2.3 Comparison with microplastic concentrations reported in other Brazilian studies

MP concentrations found in the present study were compared with data reported in previously published studies. The search was performed using Scopus and Google Scholar databases and a combination of keywords ('microplastic', 'bivalve 'and 'Brazil') without filters used to screen information. A national database was constructed to collate data reporting on MP concentrations (particles.g^{-1}ww) in Brazilian bivalve tissues. Studies reporting concentrations in other units (particles.individual^{-1} or particles.g^{-1} dry weight) were not considered due to non-harmonization of the reported data. Associated data on location (latitude and longitude) and MP concentrations were tabulated (CSV files) and imported into QGis software, and overlaid with polygons of the MPAs (available in the WDPA database; www.protectedplanet.com) according to (Nunes et al., 2021). Such an approach allowed the identification of points within and outside the boundaries of the MPAs (Table S3).

2.4 Statistical analysis

Data were reported as mean ±stand deviations (SD). The spatial differences of MP concentrations (particles g^{-1} ww) accumulated in bivalves across MPAs and IUCN categories were analyzed separately by One-way ANOVA followed by Tukey multiple comparisons test. Qualitative data across MPAs were analyzed by Two-way ANOVA followed by Tukey multiple comparisons test, taking into account the class size variable (1.2–1,000; 1,000–2,000; 3,000–4,000; and 4,000–5,000 µm), shape (fibers, films, fragments or pellets) and colors. All statistical analyses were performed using Statistica® (version 13.0 Statsoft) with a significance level of 0.01.

3. Results and Discussions

3.1 Microplastic concentrations in bivalves in Brazilian marine protected areas

A total of 90 particles were identified in the tissues of five different oyster and mussel species at the ten sampled MPAs, representing an average MP particle concentration of 0.42 ±0.34 (0.17 – 2.00) particles.g^{-1} ww. The highest concentrations were observed at AAWR, where the average concentration was 0.90 ±0.59 (0.49 – 2.00) particles.g^{-1} ww (Fig. 2). Five MPAs were found to have average concentrations between 0.45 and 0.51 particles.g^{-1} ww (FNNP, TES, GES, FRWR and ABR), while three were between 0.25 and 0.28 particles.g^{-1} ww (ANP, JNP and CES) (Fig. 2). Finally, the lowest concentration was seen in ARBR (0.23 ±0.07 particles.g^{-1} ww). Only few significant differences were observed in MP concentrations between the ten MPAs (p = 0.04) (Fig. S1), these being between AAWR and ARBR (p = 0.03) and JNP (p = 0.02) (Fig. 2).

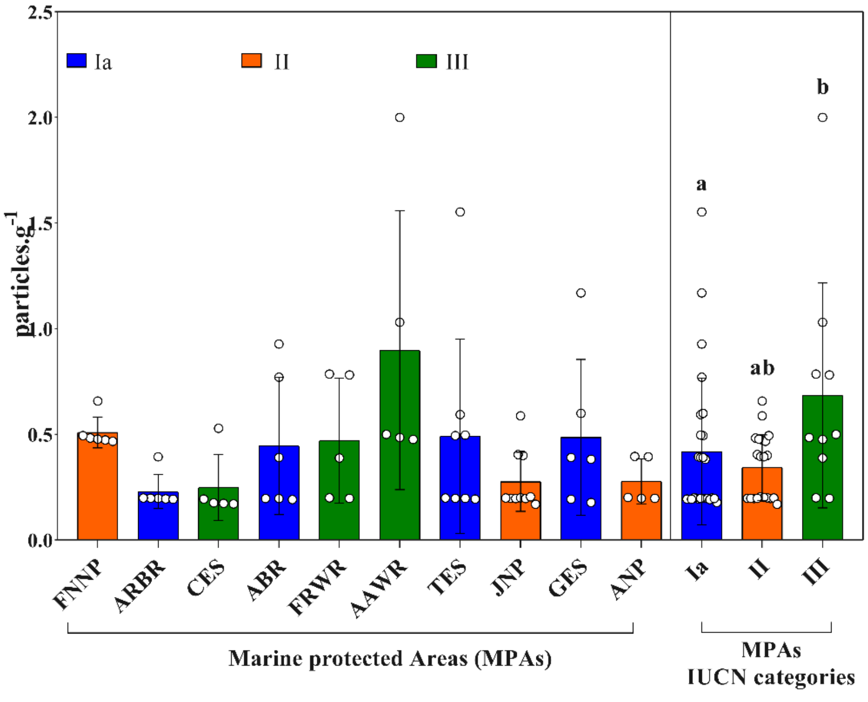

Figure 2. Particle concentrations (particles.g^{-1} ww) in bivalves from ten no-take marine protected areas (MPAs) along the coast of Brazil and within their IUCN categories Ia, II and III. Refer to Fig. 1 for locations and full names of each MPA.

Alcatrazes Archipelago Wildlife Refuge (AAWR) is considered one of the most pristine Brazilian marine regions, consisting of seven islands and islets, six submerged rocky reefs and three rocky parcels (Savio et al., 2021). This archipelago is recognized as a key area for the protection of threatened species, including several endemic birds and reptiles, with prohibition of fishing activities imposed since 1982 (Rolim et al., 2024). This region was designated a no-take MPA in 2016 and a management plan enacted one year later, authorizing recreational visitation through diving and nautical activities only as well as scientific research (Giglio et al., 2022; Rolim et al., 2024). Unfortunately, this MPA suffers from an invasive coral species (Savio et al., 2021), while the endemic species suffers from bleaching (Zanotti et al., 2024). Furthermore, linear alkylbenzenes (LABs), polycyclic aromatic hydrocarbons (PAHs) and organochlorine compounds have been found contaminating AAWR bivalves (Nunes et al., 2024). However, extensive assessments of pollution are scarce in this MPA, and none related to MPs or solid waste have so far been performed. This study reveals this archipelago to be the most contaminated compared to the other nine Brazilian MPAs investigated. Furthermore, contamination levels are as high as those recorded in other non-MPAs of Brazil (Ribeiro et al., 2024a).

The FNNP, comprised of a main island of 18 km^2 and 20 other small islands and islets, is a key area for biodiversity and harbors endangered species of the South Atlantic Ocean, including those of high environmental and economic importance (Grillo and Mello, 2021; UNESCO, 2020). Despite the distance to the coast (~360 km) as well as the low level of urbanization (~3,100 inhabitants) and the 2019 ban of several single-use plastics, plastic debris is still observed entering the FNNP, mostly likely via surface currents and winds (Ivar Do Sul et al., 2009). The FNNP is largely contaminated with cigarette filters made of acetate cellulose, and according to Grillo and Mello (2021) are likely related to high tourism rates. Indeed, MPs were found in the sediments of six beaches across the FNNP by Carvalho et al. (2021), with concentrations from 0.6 to 1,059.3 particles.m^{-2} and 325.7 to 469,774.6 particles m^{-3}. Moreover, MPs were also detected in the abundant deep-sea lanternfishes of the FNNP (Ferreira et al., 2023).

The Tamoios Ecological Station (TES) was established to protect, investigate, and monitor the marine ecosystem of Ilha Grande Bay (Trimble et al., 2014). Managed by the Chico Mendes Institute for Conservation of Biodiversity (ICMBio), TES strictly prohibits the use of marine resources, which has led to conflicts between fishers and MPA managers (Freitas et al., 2017). It comprises 29 islands, slabs, rocks, and acts as a buffer zone for a nuclear power plant in the southern portion of Angra dos Reis (ICMBio, 2006). The proximity of this type of activity and that of urban centers raises greater environmental concerns, and this concern is further supported by the detection of macro- and micro-marine litter in the region (Póvoa et al., 2022). Additionally, the currents within Ilha Grande Bay, where Tamoios is located, exhibit a permanent circulation pattern, which may increase the retention of contaminants within the bay (Rodrigues et al., 2022).

The Guaraqueçaba Ecological Station (GES), recognized as a RAMSAR site, extends across a significant portion of the Brazilian coastal and marine ecosystems, including estuaries, salt marshes, and tidal flats (MMA, 2023). These habitats are crucial for various marine species and sustains the livelihoods of traditional fishing communities (Barata et al., 2024). The detection of plastic polymer-based paint fragments in the sediments within the GES (Mengatto and Nagai, 2022) has been attributed to the proximity of human activities and raises significant environmental concerns for this vulnerable MPA (Mengatto and Nagai, 2022). The presence of high levels of MPs in sediments and in bivalves suggests a potential transfer from abiotic matrices into the biota. Thus, the plausible ingestion by marine species of lower trophic levels raises concerns about bioaccumulation and food web disruption in these MPAs (Kangas et al., 2023; Zhang et al., 2024).

Established in 2007, the Frades River Wildlife Refuge (FRWR) acts as an ecological corridor, running from the coastline to the river mouth, to protect the pockets of sand dune vegetation and inhabiting fauna (Guedes et al., 2017). The Refuge's primary objectives are to preserve ecologically significant and scenic natural ecosystems, facilitate scientific research, and support environmental education, recreation, and eco-tourism activities (MMA, 2007). Despite the ecological importance of the FRWR, the region remains understudied, with few baseline data available regarding pollution levels or anthropogenic impacts on its ecosystems. In this regard Nunes et al., (2024) recently reported low levels of LABs, PAHs and organochlorine compounds in oysters from FRWR. On the other hand, this is the first study assessing MPs contamination in the region.

Arvoredo Archipelago, known for its rocky coastline and limited human presence, sees a substantial rise in tourism and fishing during the summer and fall, leading to increased littering (Machado and Fillmann, 2010). Although only a small section of the main island permits regulated activities like scuba diving and artisanal fishing, pollutants from human activities along the coast can be transported into the Arvoredo Biological Marine Reserve (ABR) via local coastal hydrodynamic features, potentially threatening the integrity of this protected environment (Paquette et al., 2016).

The Abrolhos Archipelago, located 60 km off the coast, consists of five small islands that are part of the Abrolhos Marine National Park (ANP), which has been legally protected since 1983 (Creed and Amado Filho, 1999). This region covers a vast 45,000 km² area of the Eastern Brazilian shelf and is subject to significant anthropogenic pressures, including mining, dredging, oil extraction, and fishing, as well as the impacts of climate change (Zoffoli et al., 2022). Because it holds the major reef bank of south Atlantic and acts as the most important humpback whale nursery, it’s also target of tourist activities regarding these natural features (ICMBio, 2024). The presence of contaminants such as metals from chronic discharge of untreated sewage and waste have been detected in the ANP (Gama et al., 2022) and the present findings of microplastic presence, although in low concentration, confirms coastal activities are having an impact. Moreover, muddy plumes from the Doce River often extend into the Abrolhos archipelago following seasonal rainfalls (Coimbra et al. (2020) and the continental shelf acts to transport sediments (Kane et al., 2020), providing a mechanism for microplastic transport from land to sea. The introduction of these anthropogenic stressors via natural coastal processes highlights the urgent need for comprehensive research to understand their effects and better inform conservation planning and management strategies (Cardial et al., 2024).

Jericoacoara National Park (JNP), established in 2002, spans 8,416 hectares and is dedicated to safeguarding coastal ecosystems while balancing local tourism with natural resource preservation (ICMBio, 2002). This park, now the fourth most-visited in Brazil, attracts nearly one million tourists annually and features a diverse array of coastal landscapes, including sandy beaches, mobile dunes, mangroves, and rocky shores (Melo et al., 2022). However, the park faces significant threats from litter, particularly due to its extensive beach areas and popular tourist spots, which influences the distribution of various waste types (Brabo et al., 2022). Key factors contributing to litter accumulation include the proximity to estuaries and nearby villages (Crosti et al., 2018) like Jericoacoara city. Notably, specific types of waste, such as cigarette butts, styrofoam, plastics and rope were found in JNP beach sediments (Brabo et al., 2022). These findings highlight the need for targeted management strategies, including beach cleanups, strategic placement of trash cans, and awareness campaigns focused on reducing specific types of litter. Implementing these measures would help promote the sustainable use of the beaches and attain the ultimate goal of the conservation unit, to preserve the ecosystem.

Located in one of the largest tourist cities in southern Brazil, Carijós Ecological Station (CES) is home to several critically endangered, endangered, and vulnerable species according to the IUCN Red List of Threatened Species. Oysters sampled in CES face significant contamination from pesticides, PAHs and polychlorinated biphenyls (PCBs) due to the low sewage collection which consequently has led to increased contamination ratios and oxidative stress and lipid peroxidation in local oysters (Bastolla et al., 2024).

The Atol das Rocas Biological Reserve (ARBR), located off the coast of Rio Grande do Norte, Brazil, was established by Decree No. 83.549 on June 5, 1979. As the first marine protected area in Brazil, it encompasses over 36,000 hectares of ocean, including a submerged mountain within the Fernando de Noronha Chain. The reserve features the only atoll in the South Atlantic, characterized by its unique coral algal formations and geomorphological features reminiscent of both Atlantic and Pacific atolls. The atoll supports diverse marine life, including ornamental fish, sharks, crustaceans, mollusks, corals, and sea turtles, and provides critical nesting sites for migratory and endangered seabirds (ICMBio, 2007). The lack of studies regarding contamination status in ARBR leaves a significant knowledge gap. Oil spills are the most commonly investigated issue in the Reserve and pose a significant threat to the biodiversity (Zacharias et al., 2023). The detection of MPs in bivalves in the present study, and in the abundant deep-sea lanternfishes of the ARBR (Ferreira et al., 2023) indicates plastic pollution is an additional concern for the region.

Microplastic concentrations were higher in bivalves from management category III (natural monuments) MPAs (0.54 ±0.47 particles.g^{-1} ww) (Table S2), biased somewhat by the elevated levels in AAWR (0.89 ±0.59 particles.g^{-1} ww) which exhibited the highest concentration across all sites. Category II MPAs displayed the lowest average concentrations, 0.34 ±0.15 particles.g^{-1} ww, with FNNP recording the highest concentration within this category (0.65 particles.g^{-1} ww). Notable concentrations were also observed in category Ia strict nature reserves with an average of 0.42 ±0.33 particles.g^{-1} ww. The trend was III>Ia=II, with significant differences in MP concentrations observed only between Ia and III ($p = 0.03$), and mirrors the increasing level of protection from III to Ia. The variation observed within each category can be explained by the wide range of regional characteristics, from remote and uninhabited to highly urbanized and densely populated areas, as well as distinct geomorphological features and proximity to ports and river discharges, all of which influence MP input rates, transport potential, distribution, and thus MP bioavailability and levels ingested and retained by filter-feeding bivalves (Nunes et al., 2023b).

Seeking to compare findings from this study, a total of 496 records of MP concentrations in marine bivalve tissues collected across Brazil were identified in the scientific literature (Table S3). Outside MPAs, 250 records were found, reporting an average concentration of 4.82 particles.g^{-1} ww in bivalves (Fig. 3). These MP concentrations are considered high, and are in the country's coastal northeastern (Bruzaca et al., 2022), southeastern (Bom et al., 2022a; Costa et al., 2023; Neves et al., 2024; Ribeiro et al., 2023, 2024a), and southern (Jankauskas et al., 2024a; Saldaña-Serrano et al., 2022) regions. Unsurprisingly, bivalves from highly urbanized areas were found to have higher MP concentrations (Jankauskas et al., 2024a; Ribeiro et al., 2023, 2024a). Inside MPAs (246 records), an average concentration of 1.15 particles.g^{-1} ww was observed (Fig. 3). In comparison, bivalves obtained from the no-take MPAs showed low to moderate contamination levels (0.42 particles.g^{-1} ww) indicating some level of protection (Fig. 3). Similar or lower MP concentrations have been previously observed in bivalves from Amazonian estuaries (Pantoja et al., 2024; Rodrigues et al., 2024).

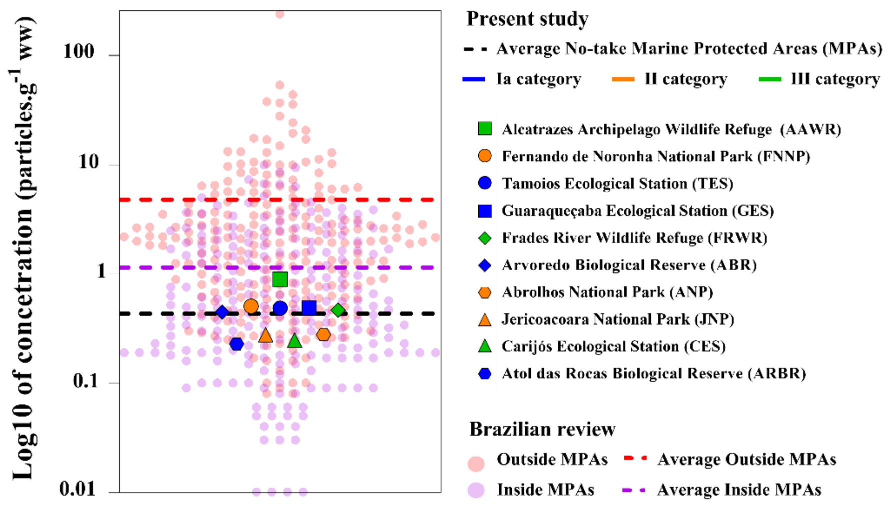

Figure 3. Average microplastic concentrations (particles.g^{-1} ww) in bivalves from Marine Protected Areas (MPAs) within IUCN categories Ia (blue), II (orange) and III (green) compared to literature reports (496) within (light purple) and outside (light red) MPAs.

3.3 Microplastic qualitative aspects

Polymeric composition analysis of all 90 particles found in bivalve tissues revealed only 20 particles (22.2%) had an acceptable library match (>70%) to known polymers. Ideally, all found particles should be fully identified, but several known technical difficulties are faced in this challenging analytical procedure. Despite the low number (representing 37% of the found items), further analysis was undertaken (Jankauskas et al., 2024a; Ribeiro et al., 2024a). Of the 20 identified particles, thirteen (60%) were confirmed to be cellulosic (CEL), three (15%) polyethylene (PE), and two polyester (PES) and rayon:polyester (RY:PES) (10% each) (Fig. 4).

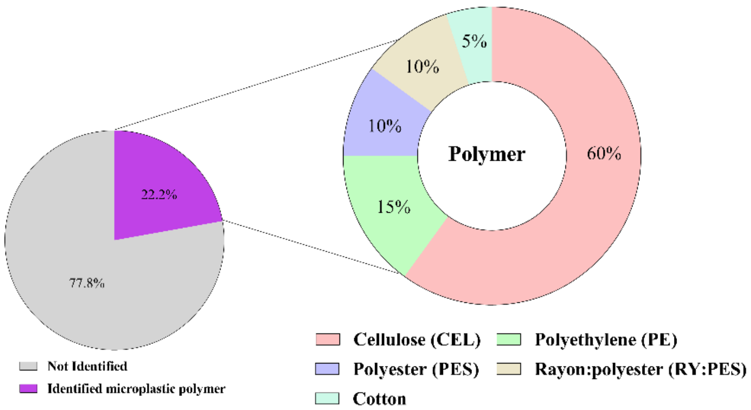

Figure 4. Polymer composition of microplastics found in bivalves from no-take Brazilian marine protected areas.

The substantial presence of CEL aligns with findings from other global research (Ergas et al., 2023; Hasenmueller et al., 2023; Liu et al., 2024), including for bivalves. Indeed, cellulosic MPs were prevalent in bivalves from Argentina (Truchet et al., 2021), Australia (Colombo et al., 2023), Brazil (Ribeiro et al., 2023), Canada (Coverton et al., 2019; Rowenczyk et al., 2022), South Africa (Sparks et al., 2021) and UK (Scott et al., 2019). MPs made from CEL (i.e., naturally-derived) or synthetically modified CEL (i.e., rayon and cellulose acetate) are used in many different industrial and clothing applications (Ding et al., 2022; Hartmann et al., 2019). Natural-derived CEL MPs (cotton), while themselves biodegradable, are often treated with harmful stabilizers and dyes that can be released into the environment (Surana et al., 2024) and induce toxicity in living organisms (Ribeiro et al., 2023). Moreover, CEL acetate is a component of cigarette filters (Belzagui et al., 2021; Joly and Coulis, 2018) and, with cigarette butt litter now considered one of the most prevalent litter types, represents an emerging global concern (Araújo et al., 2022; Conradi and Sánchez-Moyano, 2022; Green et al., 2023) given the impact on wildlife (da Silva et al., 2023; Lima et al., 2021)

Polyethylene is widely used in plastic bags, juice containers, and cling wrap due to it being simple to make and its chemical resistance, toughness, durability, moisture resistance, and low production cost (Deus et al., 2024; Ghatge et al., 2020). It is also the most prevalent type of plastic found in coastal environments. Polyester is a synthetic polymer widely used in textiles and various industrial applications and has emerged as a significant source of MPs in natural environments due to its high use, durability, and persistence (Gao et al., 2024). Rayon, although a CEL-based semi-synthetic polymer, is a key contributor to microfiber contamination in marine ecosystems, commonly released from treated textiles that also contain harmful substances such as PCBs (Herrero et al., 2022).

The size of MPs found in bivalve tissues ranged from 149.8 to 7,290.1 µm. The most abundant size class was 1.2–1,000 µm (75.6%), followed by 1,000–2,000 µm (17.8%), 2,000–3,000 µm (3.3%), >5,000 µm (2.2%) and 3,000–4,000 µm (1.1%) (Fig. 5). Indeed, the presence of smaller MPs in bivalves from Brazilian no-take MPAs is consistent with data from bivalves in unprotected areas in Brazil and global patterns (Bom et al., 2022b; Jankauskas et al., 2024b; Truchet et al., 2021). No significant differences in MP sizes were observed among MPAs ($p = 0.11$) or the IUCN categories ($p = 0.01$) (Fig. S2) suggesting that the feeding habit of bivalves across all locations was the same, (i.e., having a preference for smaller MPs), and that this size range was present in all environments examined and bioavailable to the bivalves.

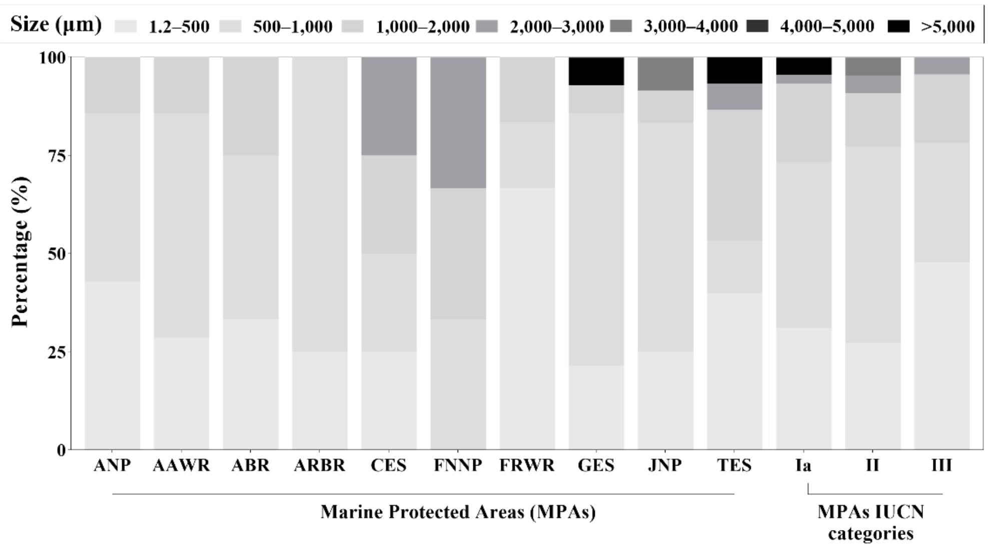

Figure 5. Percentages of microplastic size classes across sampled no-take Brazilian marine protected areas (MPAs) and management categories Ia, II and III. Refer to Fig. 1 for locations and full names of each MPA.

Three MP shapes were consistently found across all sites, i.e., fragments, fibers and films (Fig. 6); while pellets were not found. Fragments were the most prevalent (60.0%) across nearly all sites, particularly in AAWR. A total of 35.6% of MPs were fibers, found in all MPAs except JNP. Films represented only 4.4% of MPs, and were found at ABR, ARBR and FRWR only. As for size class, no significant differences ($p > 0.05$) in MP shape were observed across MPAs or management categories (Fig. S3).

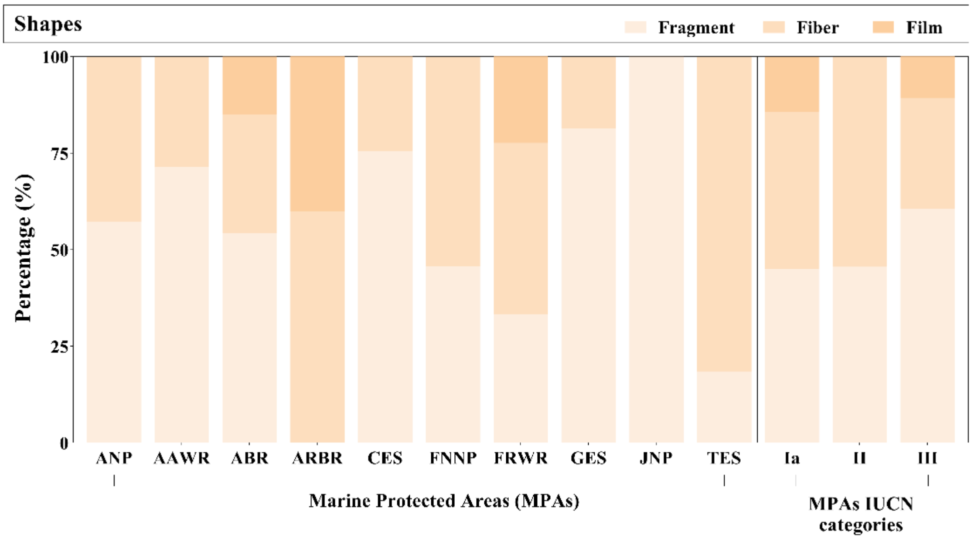

Figure 6. Percentages of microplastic shape across no-take Brazilian marine protected areas (MPAs) and management categories Ia, II and III. Refer to Fig. 1 for locations and full names of each MPA.

Fibers represent >90.0 % of global MP contamination in natural environments (Barrows et al., 2018; Do Amparo et al., 2023). This is reflected at a regional scale, with fibers commonly reported in Brazilian urbanized zones and environments outside MPAs (Jankauskas et al., 2024b; Ribeiro et al., 2023, 2024a). Fibers are also the predominant MP shape found in bivalves (Bom and Sá, 2021b) and ingestion of fibers is known to cause harmful effects, including stunted growth, tissue damage, compromised body condition, and increased mortality (Khanjani et al., 2023; Sharifinia et al., 2020; Sussarellu et al., 2016; Zhang et al., 2020). The lower load of fibrous MPs in Brazilian MPAs reported here is possibly because of the distances of the MPAs from the likely sources, i.e., raw sewage discharges, garbage dumping sites and slums (Ribeiro et al., 2023). While fibers are highly mobile in aquatic systems mostly due to their reduced density (Brahney et al., 2020; Liu et al., 2021), the high abundance of fragments in the present study may reflect their environment persistence. Fragments are increasingly found on shorelines and in oceans worldwide (Barnes et al., 2009; Baroja et al., 2021; Thompson et al., 2004) and pose physical hazards to marine organisms also acting as vectors for harmful chemicals (Koch and Calafat, 2009).Microplastic fragments, which originate from the degradation of larger plastic debris, are the second most persistent form of MPs in marine environments (Do Amparo et al., 2023; Peng et al., 2020). Their sharp edges can induce physical tissue and cellular damage, leading to heightened physiological impacts (Park et al., 2024). In this regard, experimental studies have demonstrated that fragmented PE and PS result in greater oxidative stress, immunotoxicity, and behavioral alterations in aquatic organisms, than spherical equivalents suggesting that the morphology of MPs is a more critical determinant of toxicity than their size (Choi et al., 2018; Na et al., 2021; Park et al., 2024; Qiao et al., 2019). Thus, based on higher abundance of fragments compared to fibers found in filter-feeding bivalves from Brazilian no-take MPAs, and with no spherical MPs found, a certain degree of risk is expected.

Despite little evidence that color is a factor in ingestion, assessment of MP color is needed to aid source detection (Jankauskas et al., 2024a). Here, eleven colors were observed for MPs recovered from bivalve tissues. The most prevalent colors found were black (31.1%), followed by white (15.6%), and brown (13.3%), the remaining 40% were composed of yellow (11.1%), orange (7.8%), transparent (7.8%), blue (3.3%), pink (3.3%), purple (3.3%), red (2.2%) and gray (1.1%) (Fig. 7). Considering individual MPAs and management categories, no statistical differences ($p > 0.05$) were seen among MP colors (Fig. S4). Therefore, no location-specific sources can be attributed to the MPs found in bivalve tissues, rather, the results reflect the ubiquitous presence of black, white and brown MPs in the environment. Systematic studies using different organisms have also identified these MPs colors (Santana, 2021).

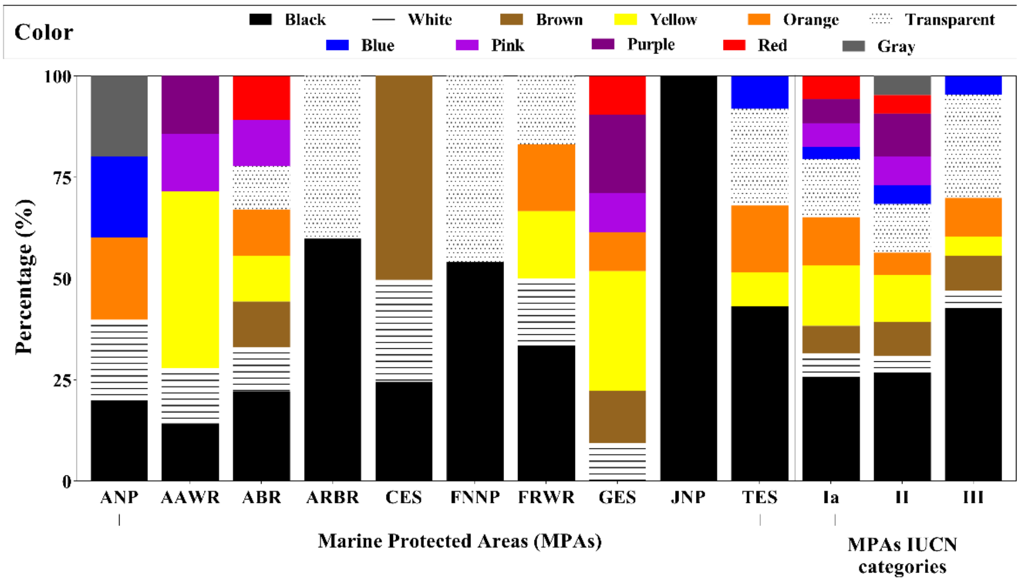

Figure 7. Percentages of microplastic colors across sampled no-take Brazilian marine protected areas (MPAs) and its IUCN categories Ia, II and III. Refer to Fig. 1 for locations and full names of each MPA.

Black MPs mainly enter the environment due to abrasion of tires on the road surfaces as regular wear and tear (Gurjar et al., 2022; Sewwandi et al., 2024), or from industrial and electronics waste. Moreover, black plastics are the least recycled plastic type due to intrinsic technical difficulties in waste sorting that relies on infrared technologies. This results in a significant proportion of black plastic waste being dumped rather than recycled or repurposed and therefore a higher percentage that can be fragmented into microplastics (Huang and Xu, 2022). There is also evidence showing that some marine species preferentially select black MPs over other colors due to visual similarity to their food, such as demersal fish (Atamanalp et al., 2021; Kılıç and Yücel, 2022). In contrast, the presence of colorless (i.e., white and transparent) particles in aquatic environments is often linked to sources like fishing lines and nets (Davidson and Dudas, 2016; Nithin et al., 2021) or the breakdown of larger transparent and white plastics, including bags, cups, and bottles used in packaging and clothing (Fu and Wang, 2019). Colorless particles have been found to be prevalent in bivalves from southern and southeastern Brazil (Ribeiro et al., 2023; Ríos et al., 2020), Canada (Wardlaw and Prosser, 2020), Africa (Wakkaf et al., 2020), Asian countries

(Cho et al., 2021; Sathish et al., 2020), Oceania (Jahan et al., 2019) and Europe (Marques et al., 2021). It is important to note that colorless particles might dominate in aquatic systems because they can also result from fading and bleaching of previously colored particles due to environmental exposure (Nithin et al., 2021). Colored MPs are commonly used in a multitude of applications, including objects that are a part of daily life, i.e., synthetic textiles, packaging, and clothing, (Gurjar et al., 2022; Wang et al., 2017) yet because of this source identification based on color alone is prone to inaccuracies (Teng et al., 2020).

4. Potential impacts and conservation remarks

Based on the current scientific understanding and an inherent lack of established safety thresholds, it is difficult to accurately assess potential risks related to MPs contamination associated with bivalve tissues within the MPAs. Studies have shown that MPs can accumulate in bivalves, leading to various physiological disruptions, including a decrease in filtration and respiration rates, oxidative stress, and reduced reproductive success (Bringer et al., 2021; Ribeiro et al., 2021). Despite the absence of standardized limits, the MPs levels observed here in no-take MPAs are alarming, especially considering the potential of synergistic or additive effects when combined with other environmental pollutants (Osman et al., 2023; Sun et al., 2020). This is further compounded by the fact that low to moderate levels of PAHs, LABs, and persistent organic pollutants (POPs) like dichlorodiphenyltrichloroethane (DDTs) and PCBs were also detected in these same bivalve samples as part of an integrated study (Nunes et al., 2024). Collectively, these findings corroborate the hypothesis that anthropogenic sources simultaneously release MPs and hazardous chemicals into the marine environment including regions considered to be vulnerable. Thus, despite uncertainties regarding toxicological effects, the levels of contamination observed in no-take MPAs of Brazil is of concern (Khanjani et al., 2023).

Pollution is a leading cause of biodiversity loss globally, and the widespread contamination by MPs in MPAs is a growing concern (Nunes et al., 2023b, 2023a). The impact of MPs extends beyond direct physiological harm to bivalves; it also affects entire ecosystems by disrupting food sources and nutrient cycles (Green, 2016; Sjollema et al., 2016). Given that MPs can also carry other harmful contaminants, such as toxic metals, hydrocarbons and persistent organic pollutants, their presence in MPAs raises the level of threat to marine life (Sana et al., 2020). Therefore, implementing regular monitoring programs to assess MP and chemical contamination is crucial to guiding management strategies and ensuring the effectiveness of conservation efforts in MPAs (Khanjani et al., 2023). Still, the impacts induced by MPs exposure may extend through the marine food web, posing potential risks to higher trophic levels (Ribeiro et al., 2024b). For instance, the ingestion of MPs by zooplankton, which are crucial food sources for bivalves, has been shown to impair bivalve health and reproduction, further affecting predator species that rely on these organisms (Jaikumar et al., 2019). Additionally, MPs can carry pathogenic bacteria and toxins, potentially leading to the bioaccumulation of harmful substances up the food chain (Frère et al., 2018; Sababadichetty et al., 2024). Microplastics, once ingested by bivalves have been shown to be transferred to predator species (Mercogliano et al., 2020; Revel et al., 2018). As MPs move through the trophic levels, they can impact the nutritional quality of prey and contribute to broader ecological disruptions (Danopoulos et al., 2020; Khanjani et al., 2023), ultimately affecting the biodiversity and, therefore, the intent of MPAs, that being conservation. Therefore, such results underscore the urgency of addressing MPs contamination, through continuous monitoring programs, and adopting strategies to safeguard marine ecosystems.

5. Conclusions

Filter-feeding bivalves from ten no-take MPAs along the Brazilian coast presented MPs contamination regardless of the IUCN management category. Considering all MPAs, the majority of particles associated with the bivalves were black, white or transparent fragments composed by cellulosic polymers with size less than <1000 µm. However, no statistical differences were found among these qualitative parameters when considering individual MPAs and management categories. Highest MPs concentrations were seen in the Alcatrazes Archipelago Wildlife Refuge (category III), and while strict nature reserves and national parks, assigned management categories Ia and II, respectively, had lower levels, these were found to have concentrations similar to each other. Overall, no-take MPAs were found to be less contaminated by MPs than multiple-use MPAs and unprotected areas. Such findings indicate MPAs do afford some level of protection against MP contamination, yet further efforts are required to meet the 11th Aichi Target. Considering the low to moderate MP contamination levels found here, long-term monitoring of MPs occurrence and ecological impacts is recommended across all these highly vulnerable MPAs.

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Microplastic in bivalves from Great Barrier Reef, an Australian World Heritage Site

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ABSTRACT

The global decline in ocean biodiversity is largely driven by pollution, with plastic debris emerging as a significant threat due to its transformation into MPs spread across marine ecosystems. Coastal areas, including protected zones, are not immune to microplastic contamination, as human activities and natural processes facilitate their distribution, even in Marine Protected Areas (MPAs). This study aims to evaluate MPs contamination in MPAs at the coastal areas of Townsville in northeastern Australia, using bivalve mollusks as sentinels. MPs (MP) were detected in bivalve tissues across all 10 sampled sites. Great Barrier Reef Marine Park (GBRMP, Category IV, multiple-use) showed the highest average MP concentration (0.12 ±0.23 particles.g^{-1}) among the MPAs studied. No relationships between human modification levels and MP concentration were observed, suggesting alternative contamination sources. Among MP items, 11 different polymers, including cellulose and polyester, were identified. MPs were primarily found in fibrous forms, constituting 83% of samples, while fragments accounted for 17%. The measured MPs ranged from 101 to 3655 μm, with a significant majority being smaller than 1000 μm, indicating their potential for environmental transport and ecological impact. The Port of Townsville and Ross River estuary are potential contamination sources of local MP inputs. Compared to global averages, MP concentrations in the present study were relatively low, indicating some levels of effectiveness of studied MPAs.

Keywords: Protected area; conservations, marine debris; contamination; plastic

1. Introduction

Threats to ocean biodiversity on global scale are largely attributed to pollution (Young et al., 2016), which is commonly related as intentional or unintentional discharge of waste and hazardous chemicals into natural environments. Plastic debris stands out as a primary residue threating the health of marine ecosystems, with numerous studies investigating its direct and indirect impacts on biota (Celis-Hernández et al., 2021; Wootton et al., 2022). The transformation of plastic debris, ranging from larger fragments to smaller particles, contributes significantly to the widespread formation of MPs in marine environments (Frias and Nash, 2019). In addition, primary microplastic, including pellets and microbeads, have also been widely found in coastal and marine zones around the world (Hernán et al., 2024; Isobe, 2016; Kushwaha et al., 2024; Wu et al., 2024; Zainuddin et al., 2022).

After release in coastal environments, MP can be transported reaching distant and isolated zones like protected coastal areas. Thus, MP distribution patterns vary across different shoreline regions according to drift currents, winds and inputs (Andrady, 2011; Tiwari et al., 2023). Due to their bioavailability in coastal areas, MPs can interact and even be ingested by organisms, belonging various trophic levels (Nunes et al., 2023b). In this regard, bivalves are essential filter-feeding organisms highly susceptible to MPs uptake due to their feeding habits, global distribution and sedentary lifestyle (Can Tunçelli and Erkan, 2024). Such animals are often exposed to MPs from the water column, leading to reported adverse effects on gills and other physiological systems (Z. Li et al., 2022; Ribeiro et al., 2023). Furthermore, the wide geographical distribution of most of bivalves species makes them ideal sentinels to monitor global levels of MPs in aquatic environments (Ding et al., 2021).

As MPs ubiquity and effects in marine environments are widely acknowledged (Z. Li et al., 2022), monitor their occurrence and potential sources are necessary steps in assessing potential risks to Marine Protected Areas (MPAs). While MPAs are known as conservation tools to preserve natural resources and biodiversity (Rice et al., 2012), they are not immune to microplastic pollution (Nunes et al., 2023b, 2023a). MPAs are the focus of global goals, such as the Aichi Targets and the 2030 Agenda, which aim to promote the sustainable use of terrestrial and marine life, along with the services these ecosystems provide, through the expansion of their coverage (UNEP-WCMC & IUCN, 2021). Besides that, MPAs are usually categorized by the International Union for Conservation of Nature (IUCN) based on their objectives. No-take MPAs, including nature reserves (Ia), national parks (II), and natural monuments (III), restrict or prohibit human activities. In contrast, sustainable-use MPAs permit certain human activities within their boundaries (Dudley, 2008). Despite protection measures, MPAs may experience similar levels of microplastic contamination as urban areas due to long-range atmospheric (Hee et al., 2023) and currents transport as well (Nunes et al., 2023a). Moreover, direct MPs releases have been reported within MPAs (Nunes et al., 2023b, 2023a). This challenges the assumption that MPAs are fully protected from pollution.

Microplastic pollution is especially concerning in Australia, where an advanced legislative framework on marine protected areas is into force (Yin and Techera, 2020). On the other hand, the combination of human activities and natural processes along the northeastern coast provides an opportunity to study the occurrence and distribution of MPs. Indeed, urban areas, such as Townsville in Northeast Queensland including regions near the Great Barrier Reef, are subject to significant human pressures, which may lead to microplastic contamination (Shand and Taylor, 2024). The city of Townsville includes islands, rivers, rock shores, sandy beaches, mangroves and the reef barriers, suffering with extensive human pressures such as tourism, urban development, port installations and fishing, all of which may contribute to plastics inputs into marine environments (Shand and Taylor, 2024). Also, the area is under frequent wave action and tropical cyclones, which can further influence the distribution and deposition of MPs along the coast (Hitchcock, 2020; Nott et al., 2009). Therefore, the interaction between intense human activities and these natural dynamic processes makes Townsville region particularly vulnerable to microplastic pollution, affecting both protected areas and sensitive ecosystems (Hitchcock, 2020; Shand and Taylor, 2024). Thus, the present study hypothesized that protected areas located in zones under high levels of human modification will be more affected by microplastic contamination. Therefore, this study aims to evaluate MPs contamination in MPAs at the coastal areas of Townsville in northeastern Australia, using bivalve mollusks as sentinels.

2. Material and Methods

2.1. Study area

Townsville, situated in Queensland in the northeastern part of Australia, at Cleveland Bay, faces towards the Coral Sea. It was established in 1864 and has served as a port ever since, being considered the largest hub for containers and automotive trade in Northern Australia (Townsville City Council, 2024). By 2023, Townsville had an estimated population of 201,433 residents and featured a wastewater treatment facility located in the mangrove area of the Ross River delta(QLD, 2024). Pallarenda Beach, near Townsville, are popular for their accessibility, facilities, and recreational opportunities such as swimming and fishing, being part of Cape Pallarenda Terrestrial and Inland Waters Conservation Park (IUCN Category III) (Australian Goverment, 2024; UNEP-WCMC, 2024a) (Figure 1).

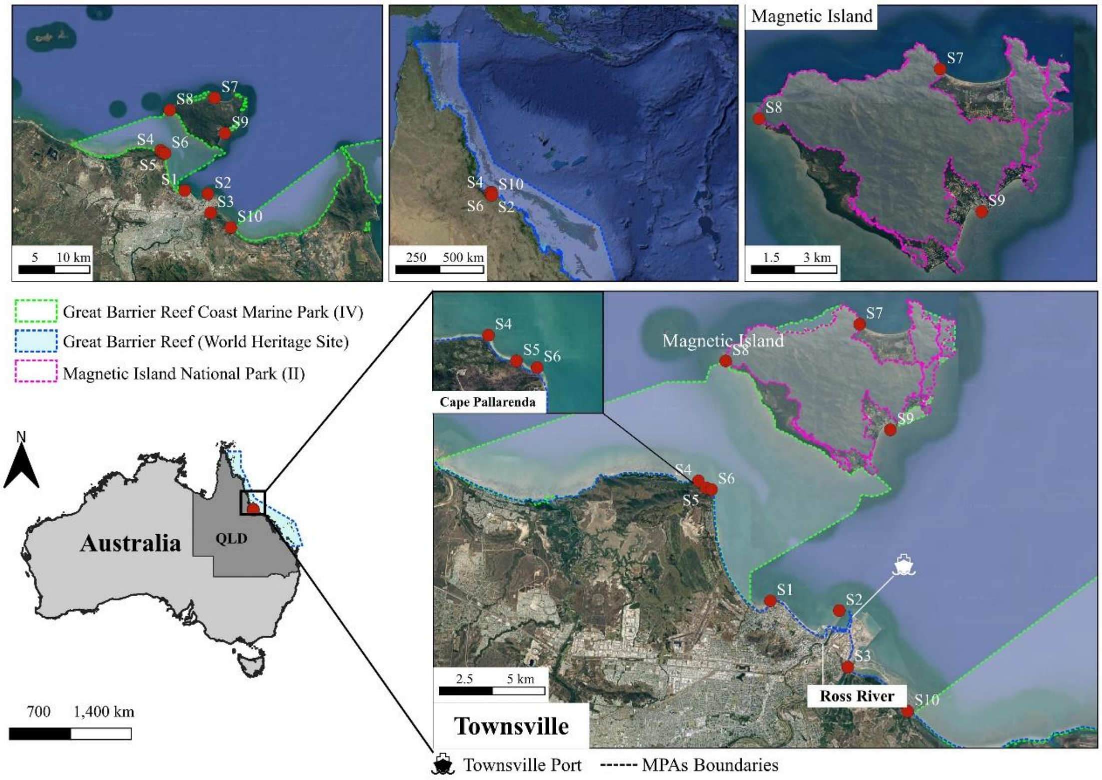

Figure 1. Map of Townsville and Magnetic Island showing the ten local sites from which filter-feeding bivalves were sampled.

Magnetic Island is located 8km offshore from Townsville within the Great Barrier Reef Marine Park, facing potential contamination from MPs due to human activities, such as maintenance dredging at Nelly Bay Marina, and natural processes like sediment transport during storms and cyclones (Lewis et al., 2012; Moscardo and Murphy, 2016). Its proximity to Townsville, combined with runoff from Ross River, increases the likelihood of microplastic inputs affecting the island's fringing reefs and delicate ecosystems. Most parts of the island are protected by a terrestrial National Park (IUCN Category II – no-take) and small areas by terrestrial Conservation Parks (IUCN Category III – no-take). Additionally, all coastal area in both Townsville and Magnetic Island is protected by the Great Barrier Reef World Heritage Site (Not Applicable to a IUCN Category), also the Great Barrier Reef Marine Park (IUCN Category Ia – no-take) and a Great Barrier Reef Marine Park mosaic (IUCN Category IV – multiple-use) (UNEP-WCMC, 2024b).

2.2. Field sampling

Between October and November 2023, oysters (*Saccostrea sp.* or *Crassostrea sp.*) were sampled in ten sampling sites under Fisheries permit no. 267475/P-MPP-100495386. In the sites S1 to S9 adult individuals of *Saccostrea sp.* were manually sampled, while in S10 the available species was *Crassostrea sp.* (Table 1). Sites S1 to S9 were sampled within the Great Barrier Reef World Heritage Site (GBRWH), which is designated as a no-take area. This classification ensures the highest level of protection, prohibiting all extractive activities, including fishing and collecting, to preserve the area's ecological integrity. Sites S7 and S8 were also located within Magnetic Island National Park (MINP), another no-take zone, aimed at conserving the island's unique biodiversity and mitigating human impact. Conversely, sites S4, S5, S6, and S10 were sampled within the Great Barrier Reef Marine Park (GBRMP), a multiple-use area under a mosaic zoning system, where a balance between conservation and sustainable use is promoted, allowing regulated human activities alongside protected zones (see Table S1).

Table 1: Sampled sites within Australian Marine Protected Areas (MPAs), designation according to IUCN management categories, sample size (n) and collected species.

Site	Site name	MPA Name	Designation (IUCN Category)	Classification	n	Specie
S1	Garabarra Lawn	Great Barrier Reef	World Heritage Site	No take	10	*Saccostrea* sp
S2	Rock Wall	Great Barrier Reef	World Heritage Site	No take	13	*Saccostrea* sp
S3	Southern Port Rd Bridge	Great Barrier Reef	World Heritage Site	No take	11	*Saccostrea* sp
S4	Cape Pallarenda	Great Barrier Reef Coast	Marine Park (IV)	Multiple use	17	*Saccostrea* sp
		Great Barrier Reef	World Heritage Site	No take		
S5	Shelly Beach	Great Barrier Reef Coast	Marine Park (IV)	Multiple use	7	*Saccostrea* sp
		Great Barrier Reef	World Heritage Site	No take		
S6	Middle Rock	Great Barrier Reef Coast	Marine Park (IV)	Multiple use	10	*Saccostrea* sp
		Great Barrier Reef	World Heritage Site	No take		
S7	Westward Horseshoe Bay	Magnetic Island	National Park (II)	No take	15	*Saccostrea* sp
		Great Barrier Reef	World Heritage Site	No take		

S8	West Point Rocks	Magnetic Island Great Barrier Reef	National Park (II) World Heritage Site	No take No take	19	*Saccostrea* sp
S9	Nelly Beach	Great Barrier Reef	World Heritage Site	No take	13	*Saccostrea* sp
S10	Cleveland Bay	Great Barrier Reef Coast	Marine Park (VI)	Multiple use	19	*Crassostrea* sp

2.3. Microplastic analysis

The microplastic analysis and quality control procedures followed Dawson et al. (2023) using individual organisms as replicates for each sampled site. The shell of each specimen was first measured using digital calipers, and the animal was weighed in analytical balance with and without the valves to obtain the condition index (CI= soft tissues weight / shells weight) according to Lawrence and Scott (1982). Subsequently, the integral tissues were removed, and samples were digested in a 10% potassium hydroxide (KOH) solution for 48 hours at 40°C in a temperature-controlled oven. After digestion, the mixture was vacuum filtered through mesh screens 547 and subsequently a 250um mesh as well. The filtration was carried out using pre-filtered distilled water to avoid potential cross contamination. After filtration, all samples were visually inspected for MPs identification using a Leica M205C microscope and analyzed using a PerkinElmer Spectrum 100 ATR-FTIR. Spectral data were collected between 650 and 4000 cm^{-1}, with 16 scans at a resolution of 4 cm^{-1}, followed by baseline correction. Polymer identification relied on NICODOM spectral libraries, with reliable matches considered for those above 70%. For each particle, detailed records of physical characteristics such as shapes (fiber, fragment and microbeads), colors (black, blue, brown, green, orange, pink, red, transparent, white and yellow) and size classes (<1.000; 1.000–2.000; 3.000–4.000; 4.000–5.000; >5.000 μm) were assessed according to (Ribeiro et al., 2024). Obtained concentrations were expressed as particles.g^{-1} of wet weight (w.w.).

To avoid background contamination from solutions, the laboratory environment and equipment, all liquids (water, H_2O and KOH) were filtered through a 1.2 μm polycarbonate filter prior to use and stored in pre-washed glass vessels. All containers, beakers and centrifuge tubes were thoroughly scrubbed with a bamboo brush, washed first in rainwater and then rinsed a minimum of 3 times with pre-filtered water. Only cotton clothing was worn, and air exposure was minimized by covering all samples, solutions and clean equipment with aluminum foil or lids. For every 3 samples, 1 blank was prepared simultaneously following the same procedure to check for potential contamination.

2.4. Human Modification Metric (HMc)

Urbanization levels were assessed using the Human Modification metric (HMc), derived from data provided by NASA's Socioeconomic Data and Applications Center (Kennedy et al., 2019). This analysis utilized the 'raster' (Hijmans et al., 2023) and 'rgdal' (Bivand, 2021) packages. The HMc measures the degree of human alteration within a 1 km radius of each location, producing continuous variables between 0 (least modified) and 1 (most modified). It is calculated by combining the spatial extent and intensity of five key factors: 1) human settlements (e.g., population density, built-up areas), 2) agriculture (e.g., farmland, livestock), 3) transportation networks (e.g., main roads, secondary roads, railways), 4) resource extraction and energy production (e.g., mining areas, oil wells, wind turbines), and 5) electrical infrastructure (e.g., high-voltage lines, nighttime lighting). As a result, HMc serves as a reliable measure of urbanization (Barboza et al., 2021; Kennedy et al., 2019).

2.5. Data analysis

A comparison of among CI values from different sampled sites was used to verify the homogeneity of obtained samples, while MPs concentrations were also compared among sites and areas. To assess data normality and variance homogeneity Shapiro-Wilk and

Levene tests were previously performed. Based on obtained results, non-parametric Kruskal-Wallis tests followed by Dunn multi-comparative analysis were selected for further analysis. The relation between human modification metric and microplastic concentrations was assessed using a Spearman correlation. All tests were performed using Statistica 7.0 and significance was set at $p < 0.05$.

3. Results and discussion

3.1 Biometric parameters

Across all sites, the shell lengths averaged 53.5 ±18.2 mm for *Saccostrea* sp and 50.4 ± 10.5 mm for *Crassostrea* sp (Table S3). The largest individuals were found at S8 (MI West Point Rocks), measuring an average of 73.4 ±13.6 mm, while the smallest came from S1 (Townsville Garabarra Lawn), with an average of 24.3 ±4.4 mm. A more reliable indicator of maturity and, by extension, indicating the capacity to accumulate contaminants (Voets et al., 2006), including MPs, is the Condition Index (CI). The CI ranged from 0.02 to 0.86 (average= 1.2 ±0.8) among the 134 oysters. The smallest values were seen in S3 in the city side of Southern Port Rd Bridge, while the highest was measured in S7, the Horseshoe Bay in Magnetic Island (see Table S3 and Fig. S1 in the supplementary material). The statistical analysis showed significant differences among some of the sites ($p < 0,0001$; see Fig. S2).

2. Microplastic concentrations in bivalves from marine protected areas

Results showed MP occurrence in bivalve tissues from all 10 sampled sites (Figure 2). However, MP was observed in 50.7% (68 organisms) of all 134 oysters dissected (Table S3) while the absence of MP was observed in 49.3% (66 organisms). Average concentrations ranged from 0.04 ±0.14 particles.g^{-1} at S3, located on the city side of Southern Port Rd Bridge, to 0.24 ±0.3 particles.g^{-1}ww at S6, Pallarenda Beach. The concentration levels followed the pattern: S6 > S10 > S1 > S8 > S4 >S 3> S5 > S7 > S9 > S2 (Table S1).

Considering the MPAs, Great Barrier Reef Marine Park (GBRMP, Category IV) had the higher average concentration of MP, 0.12 ±0.23 particles.g^{-1}, followed by the Magnetic Island National Park (MINP, Category II) and Great Barrier Reef World Heritage (GBRWH) 0.11 ±0.17 and 0.1 ±0.19 particles.g^{-1}, respectively. The Kruskal-Wallis test conducted to understand the difference among MPs concentrations considering sampled sites ($p = 0.05$) and studied MPAs ($p = 0.18$) showed no statistical variations.

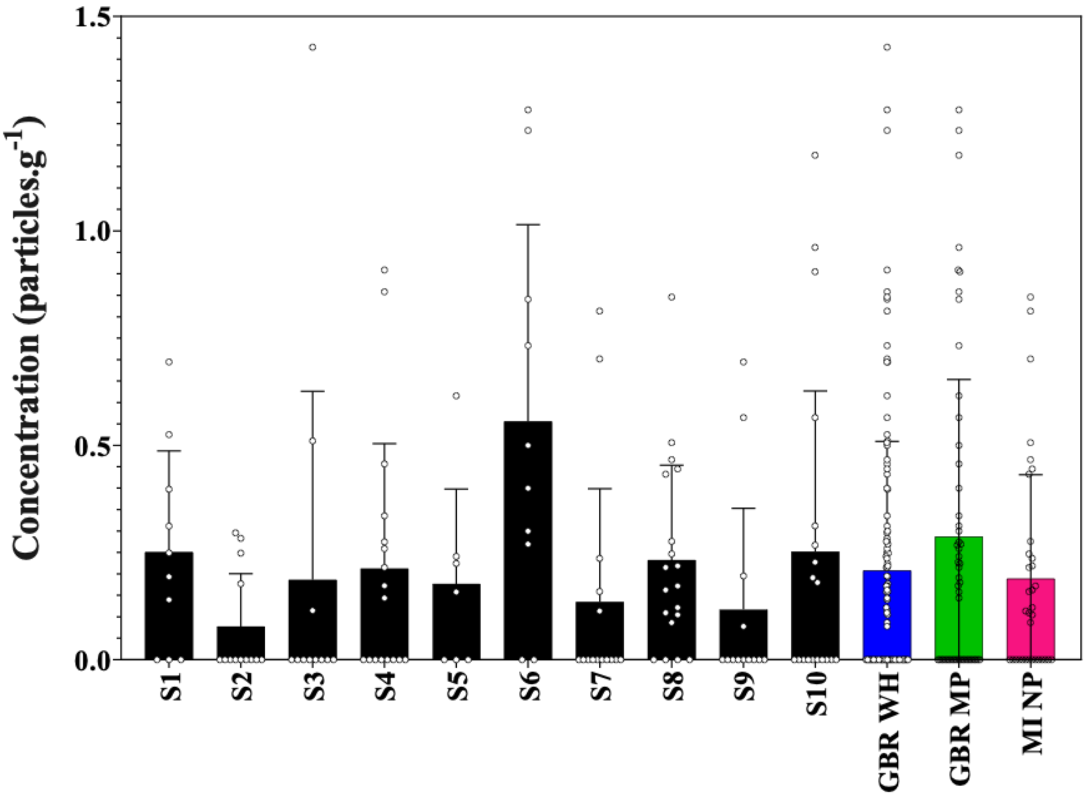

Figure 2. Microplastic concentrations (particles.g^{-1}) in bivalve tissues from ten sampling sites in marine protected areas (MPAs) along the northeast coast of Australia and within no-take and multiple-use areas.

3.1. The Human Modification metric (HMc) results

The Human Modification metric (HMc) values across the ten sampling sites revealed a range from 0.04 to 0.56, indicating varying degrees of urbanization and human impact. Garabarra Lawn (S1) recorded the highest HMc at 0.56, followed closely by Nelly Beach

(S9) at 0.55 and Rock Wall (S2) at 0.51, suggesting significant alterations due to human activities in these locations. Conversely, West Point Rocks (S8) exhibited the lowest HMc at 0.04, indicating it is the least disturbing site. Notably, despite its lower HMc, S8 had a relatively high average microplastic concentration of 0.09 particles.g^{-1}, exceeding that of more impacted sites such as Southern Port Rd Bridge (S3), which had a lower concentration of 0.039 particles.g^{-1} despite an HMc of 0.46. Interestingly, First Rock (S6) recorded the highest average microplastic concentration at 0.067 particles.g^{-1}, with a moderate HMc of 0.40. Spearman correlation analysis indicated a non-significant relation between microplastic concentrations ($p = 0.83$) and HMc, suggesting such variables are not monotonically related. These findings indicate the other sources non-related to urbanization levels are inducing microplastic contamination in the studies areas. Thus, factors like local pollution sources (Di et al., 2024) and water dynamics likely play a role (Kılıç et al., 2024).

One important contribution to coastal pollution is the Port of Townsville and harbor installations. Previous studies have shown high concentrations of toxic metals from dredged harbor sediment disposal, posing a unique challenge due to the harbor's location within the environmentally sensitive Great Barrier Reef region (Gibbs, 1993). The local hydrodynamics contributes to the dredged material being dispersed by wave-induced turbulence near the seabed, making this material highly mobile, moving away from the disposal site with the prevailing currents (Wolanski et al., 1992). Queensland coast is particularly vulnerable to tropical cyclones occurrence (Bruyère et al., 2022), floods (Department of Environment and Science, 2020) and waves caused by these type of climate event and can contribute up to 0.45m (35%) in the wave setup (Hetzel et al., 2024). Also, Ross River acts as a coastal contamination source once it carries chemicals such as per- and poly-fluoroalkyl substances, a persistent pollutant and part of a group of anthropogenic contaminants of global concern that can be toxic to wildlife (Department of Environment and Science, 2020; Wang et al., 2017). Still, atmospheric deposition of MPs in MPAs has been found to be similar to that in urban areas, suggesting that both long-range atmospheric transport (Hee et al., 2023) and ocean currents contributes significantly to microplastic influx even in protected areas (Nunes et al., 2023a).

3.2. Comparison with Global Distribution of MPs in Bivalves

A total of 331 records of MP concentrations in marine bivalve tissues collected globally were identified in the scientific literature (Table S2) reporting an average concentration of 1.98 ±3.4 particles.g^{-1}. Inside MPAs (64 records), an average concentration of 2.08 ±3.1 particles.g^{-1} has been observed. When comparing to both, the whole database and inside MPAs, the concentrations in bivalves in the present study showed a low levels of microplastic contamination (Figure 3). Wootton et al. (2022) observed a similar trend, where oysters from southern Australia had a lower microplastic abundance compared to global levels.

Considering the records inside MPAs, 9 were in no-take areas with 0.71 ±1.47 particles.g^{-1} average, while 55 in multiple-use areas with 2.3 ±3.24 particles.g^{-1} average. These findings highlight the need for more specific research to understand how local sources influence microplastic distribution and suggest that MPAs alone may not be sufficient to reduce contamination, underscoring the importance of additional measures to mitigate human impact on marine environments (Adams et al., 2023). Depending on the country social, environmental, cultural and legislative structure, the use of strict, multiple-use, and other protected areas varies, with multiple-use areas representing the largest portion of all protected areas (Adams et al., 2023). However, variability within MPAs highlights the influence of local conditions, such as currents or proximity to urban areas.

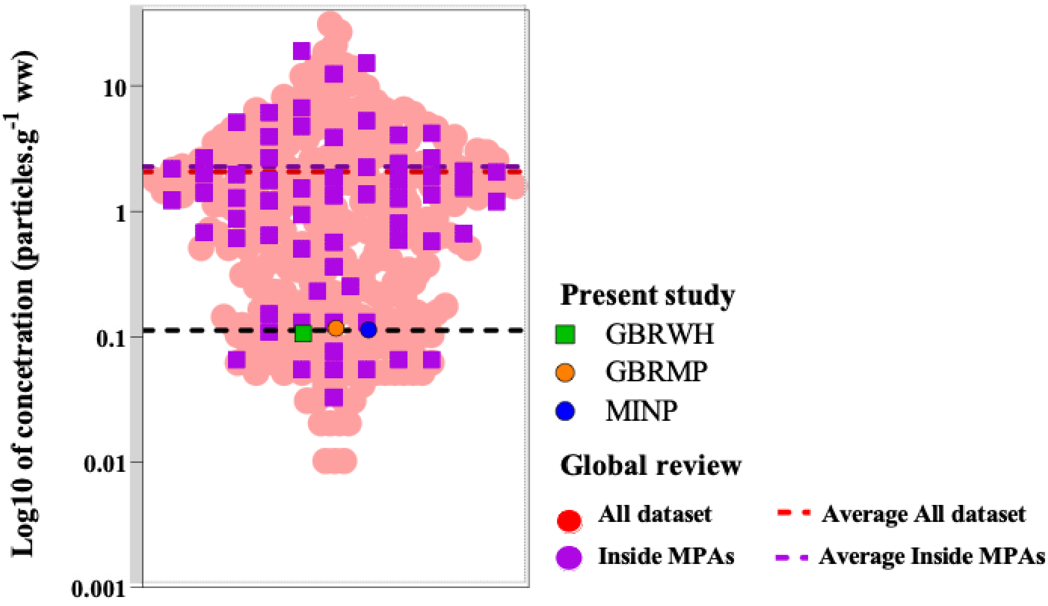

Figure 3. Average microplastic concentrations (Log10 of particles.g^{-1}) in bivalves from Marine Protected Areas (MPAs) within Magnetic Island National Park (blue), Great Barrier Reef Marine Park (orange) and Great Barrier Reef World Heritage Site (green) compared to literature reports (331) within (light purple) and outside (light red) MPAs. Refer to Fig. 1 regarding locations of each MPA.

MPs have been found in different mollusks species across several regions, including Asia, Europe, North America, and Latin America (Table S2). In the context of present study, the frequency of occurrence was relatively low at 50.7%, with a plastic load per tissue measured at 0.21 ±0.31 particles.g^{-1}, both of which are similar and below global averages, respectively. The highest average among continents was found in Europe (2.76 ±3.69), where almost 10% of the studies included in our global analysis originated. However, Asia, who usually represent the highest global values (Nunes et al., 2023a, 2023b), have an average of 2.12 ±3.99 particles.g^{-1} and represented 58% of the whole dataset. Despite this, overall, bivalves from Australia presented low levels of microplastic contamination when compared globally (0.88 ±2.03), likely due to the isolated location of sampling sites like Pallarenda Beach.

The filter-like feeding behavior and the potential to bioaccumulate in bivalves make them good sentinels of environmental contamination in marine and estuarine environments (Ding et al., 2021; Li et al., 2019). Oysters are known to be one of the best sentinel marine species for biological monitoring of water and sediment quality as they respond and reflect contamination in hotspots globally (Edge et al., 2014; Nunes et al., 2023a, 2023b; Ribeiro et al., 2023). Consequently, the microplastic presence and abundance in water and sediment are linked to contamination in oysters (Wootton et al., 2024).

3.3. Microplastic qualitative aspects

3.3.1.Microplastic shapes

Three MP shapes including fiber, fragment and microbeads were identified in the bivalve’s samples (Figure 4). The distribution of MP shapes differed among sites, while fibers represented 78%, fragments were 15% and microbeads achieved 8% of all analyzed samples. Similar pattern showing fiber prevalence have been globally found, representing more than 90.0% of MP contamination in natural environments (Barrows et al., 2018), as well as in bivalves (Bom and Sá, 2021). The higher load of fibrous MPs in the samples is often associated to the proximity of anthropogenic sources (Celis-Hernández et al., 2021; He et al., 2024; Wang et al., 2022). In fact, land-based sources, such as sewage (Wang et al., 2020), agricultural areas, and roadways (Alomar et al., 2021), atmospheric deposition (Hee et al., 2023; Zhang et al., 2020), and surface runoff (Wang et al., 2022) along with their related pathways, including wastewater treatment plants (WWTPs) and rivers, significantly contribute to the release of MPs (Wootton et al., 2024). Thus, the proximity to the Townsville Port may contribute to higher contamination by fibers. In this regard, recent findings have linked the significant presence of MP fibers in port areas to ballast water, which poses an relevant source (Allami et al., 2023; Su et al., 2024). In addition, dredging operations frequently carried out in port zones, may resuspend MPs and other contaminants historically accumulated in sedimentary layers, posing an environmental threats leading to MP bioaccumulation evidenced in organisms (Goswami et al., 2020). Moreover, the discharge from the Ross River can also act as a source of MPs, as coastal rivers are recognized as a primary pathway for microplastic inputs into the oceans (Le Breton et al., 2017). Additionally, delta regions as Rosso river estuary are particularly susceptible to becoming a pollution sink (Wang et al., 2023).

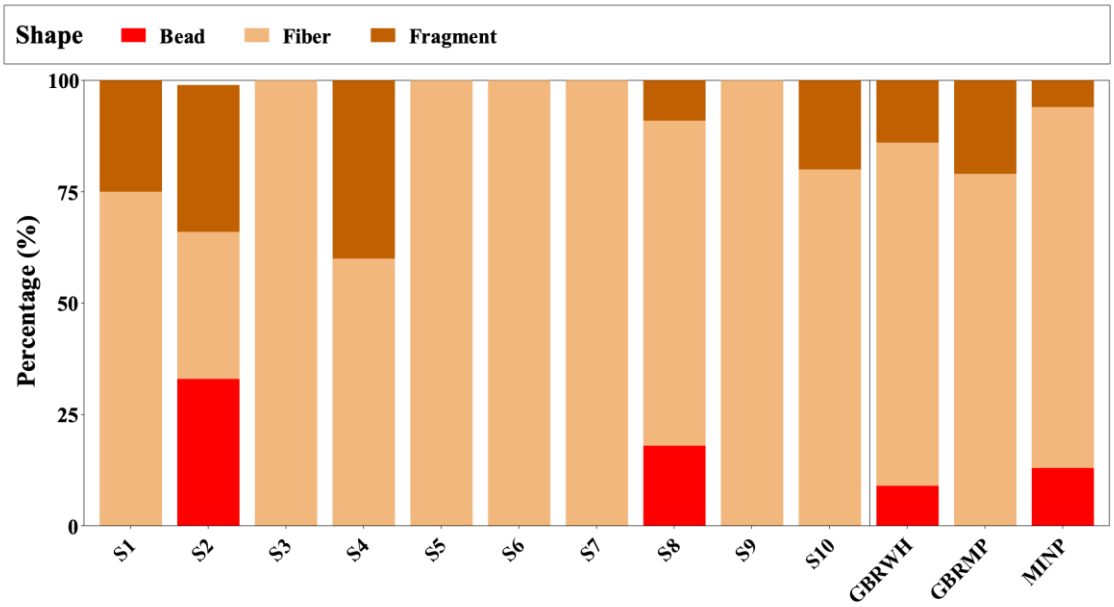

Figure 4. Percentages of microplastic shapes in bivalve tissues from ten sampling sites in marine protected areas (MPAs) along the northeast coast of Australia and within no-take and multiple-use areas.

Fragments, which originate from weathering of larger plastic debris represented 15% of the total MP, and are the second most predominant form of MPs in marine environments (Peng et al., 2020). Besides the physical damage to tissues and cells due to the sharp edges (Park et al., 2024), they also serve as carriers for harmful chemicals (Koch and Calafat, 2009) inducing deleterious effects such as spreading resistance genes and pathogenic bacteria (Arias-Andres et al., 2018). Sittl, MPs fragments may induce impacts on development and growth (Mason et al., 2022; Wen et al., 2024), decreased fertilization and abnormalities in larvae (Martínez-Gómez et al., 2017), oxidative and intestinal damages (Prokić et al., 2019), decreased metabolic rates (Welden and Cowie, 2016), neurotoxicity (Barboza et al., 2018), decreased energy allocation (Farrell and Nelson, 2013), decreased predatory functions (De Sá et al., 2015), altered behavioral responses (Barboza et al., 2018), and death (Kushwaha et al., 2024). Despite low fragment rates found in studied MPAs, these core set of potential effects may threaten the vulnerable biodiversity that is sought to be protected.

Microbeads represented the smallest amount of MP (8%) and are considered to be ubiquitous in practically all ecosystems (Zhou et al., 2023). The environmental impacts of microbeads from cosmetics were first highlighted by Zitko and Hanlon (1991), who raised concerns about pollution and ecosystem accumulation. The microbeads are used in personal care products (PCCPs) for various functions beyond exfoliation, such as abrasives and emulsifiers (Kozlowska et al., 2019; Leslie et al., 2022; Tian et al., 2022). The most common type of microbeads are polyethylene and polypropylene pellets, which appear more widespread in the marine environment (Zitko and Hanlon, 1991). At least in part, the low microbeads concentrations found may be related to bans implemented on its use in personal care products (Kukkola et al., 2024).

3.3.2.Microplastic Colors

Ten different colors were observed for MPs recovered from bivalve tissues (Figure 5). The most prevalent colors found were black (27.5%), followed by blue (20%) and white (12.5%), the remaining 40% were composed of orange (10%), transparent (7.5%), yellow (7.5%), red (5%), brown (5%), green (2.5%) and pink (2.5%).

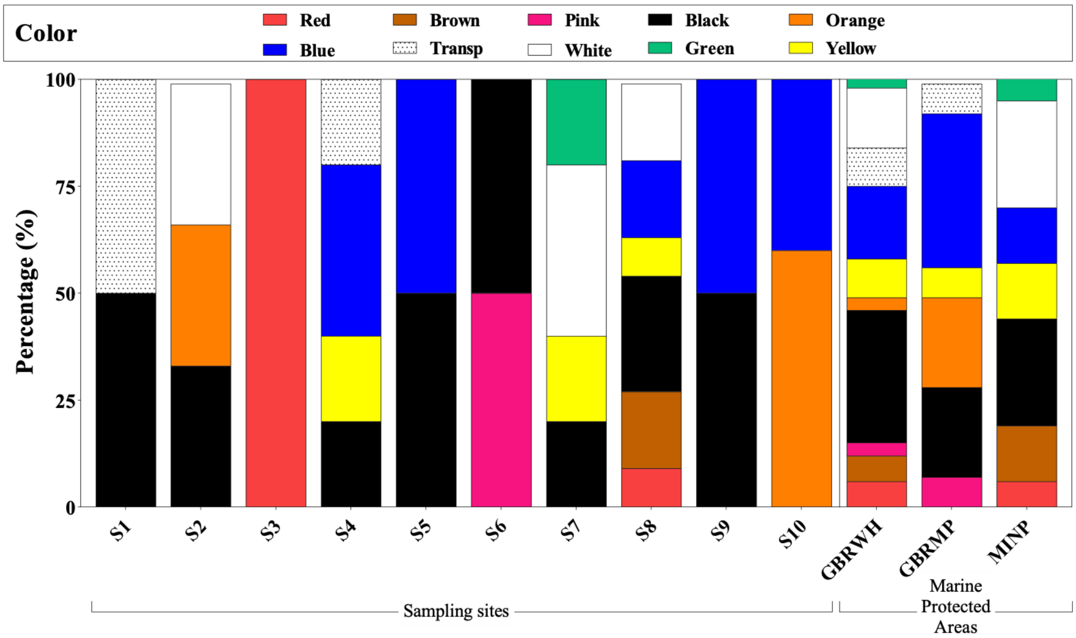

Figure 5. Percentages of microplastic colors in bivalve tissues from ten sampling sites in marine protected areas (MPAs) along the northeast coast of Australia and within no-take and multiple-use areas.

3.3.3.Microplastic size classes

The size of detected MPs in the bivalve samples ranged from 101 to 3655 µm. For the 10 sites, the majority of MPs were small (<1000 µm), with average size 928.6 ±616.1 µm. In the size class smaller than 1000 µm, the distribution rates were above 50% of the total MPs detected in each site (Figure 6), showing that smaller microplastic particles are prevalent and likely more susceptible to transport across environmental matrices (Horton et al., 2017), which may increase their bioavailability and potential ecological impact (Xu et al., 2020). In other studies, particles smaller than 1000 µm are prevalent in bivalve samples (Chinfak et al., 2024; Cho et al., 2021; Do et al., 2024; Le et al., 2024; Nikhil et al., 2024). However, at Australia's northern coast, the highest quantity of MPs found in water was in the 1000 to 2000 µm size range (Jensen et al., 2019) while in oysters of eastern coast was between 100 and 500 µm (Jahan et al., 2019).

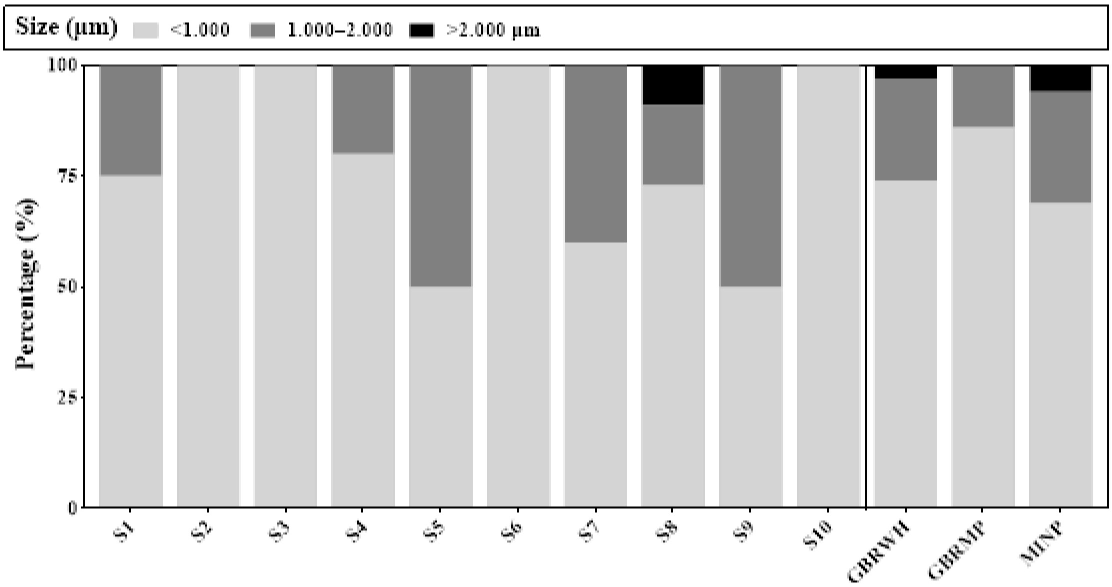

Figure 6. Size distribution of MPs in bivalve tissues from ten sampling sites in marine protected areas (MPAs) along the northeast coast of Australia and within no-take and multiple-use areas.

Our study also found that MPs between 1000 and 2000 μm were present in 6 out of 10 sampled sites, with a total occurrence rate of up to 20%, being more representative in S5 and S9. The size of MPs is an important factor influencing their bioavailability and accumulation in bivalve tissues, which can be explained by the ability of bivalves to select the particle size they ingest (Le et al., 2024). Additionally, as microplastic size decreases, they are mistaken for food particles, such as microalgae or diatoms, which increases the chance of accidental ingestion and, consequently, can accumulate in tissues (Joshy et al., 2022; H.-X. Li et al., 2022).

3.3.4.Polymers

Of all 215 items identified visually at the filters, 106 (49.3%) were analyzed in the FTIR. Spectra matches were higher than 0.7 for 88 particles (83%), while 18 (17%) particles were not confirmed due to matches below than 0.7. Thus, 11 different polymers were found and confirmed: cellulose (CEL), glycol fatty acid (GFA), metal fatty acid (MFA), nylon (NY), Natural fiber composites of nylon and polyurethane (NY-PU), polyacrylate (PAC), polyethylene glycol (PEG), polyester based (PES), naturally-derived (ND), polypropylene (PP) and polyurethane (PU) (Figure 7).

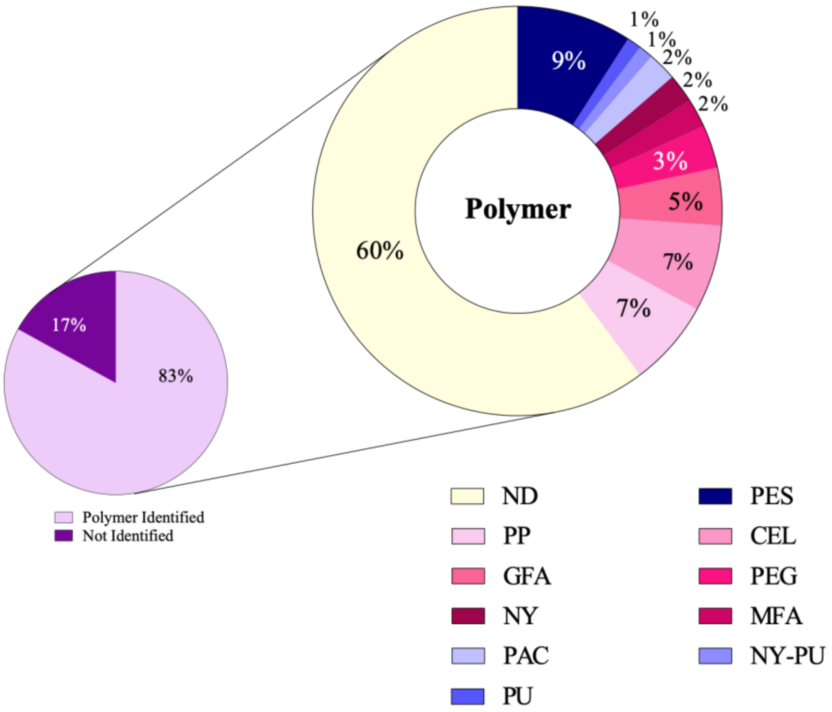

Figure 7. Polymer composition of MPs found in bivalve tissues from Australian marine protected areas. PU: polyurethane; NY: nylon; CEL: cellulose; PES: polyester; PAC: polyacrylate; PEG: polyethylene glycol; PP: polypropylene.

The microplastic polymers observed in the present study are ND > PES > CEL > PP > GFA > PEG > MFA > NY > PAC > PU > NY. The PES polymers comprise of alkyd-based polymer and polyester and represent 9% of microplastic polymers in the present study. An alkyd resin is a polyester modified with oils, incorporating fatty acids and several additional components (Onn et al., 2024). Currently, surface coatings based on alkyd resins as film formers continue to be one of the largest types of coatings used in the world (Abraham and Höfer, 2012). Polyester fibers, such as usually originate from clothes and are released in wastewater into bodies of water. The significant presence of PES in the environment (Awuor et al., 2024; Thao Le et al., 2024; Xu et al., 2024) was reflected in the current study. However, PES microfibers at current environmental concentrations seems to be a minimal threat to organisms (Thao Le et al., 2024).

The high amount of CEL over other polymers corresponds partially with findings from global studies (Ergas et al., 2023; Liu et al., 2024). Also, cellulosic MPs were found to be common in bivalves from south Australian mussels (Klein et al., 2022) and Australian prawns from several locations (Ogunola et al., 2022). Naturally derived cellulose and its synthetic variations, such as rayon and cellulose acetate, are utilized in diverse industrial and textile sectors (Ding et al., 2021; Hartmann et al., 2019). Despite their biodegradable characteristics, these materials are often treated with toxic stabilizers and dyes that may leach into the environment (Surana et al., 2024), potentially endangering living organisms. Furthermore, cigarette filters containing cellulose acetate are frequently littered (Belzagui et al., 2021; Joly and Coulis, 2018), representing a considerable threat to wildlife (Da Silva et al., 2023; Lima et al., 2021) and are increasingly acknowledged as a pressing global concern (Araújo et al., 2022; Conradi and Sánchez-Moyano, 2022; Green et al., 2023).

Polypropylene (PP) is one of the most commonly found MP in the environment (Shruti et al., 2021; Stang et al., 2022; Xun et al., 2024) and yet one of the most toxic polymer (Bobori et al., 2022; Xun et al., 2024). The ingestion of PP by freshwater organisms can cause restricted cellular function of the gills and hepatic cells by lipid peroxidation, DNA damage, protein ubiquitination, apoptosis, autophagy, changes in metabolite concentration, oxidative stress and liver damage (Bobori et al., 2022; Jeyavani et al., 2023; Xun et al., 2024). Regarding marine organisms, MP consumption may disrupt affected antioxidant biomarkers, ultimately causing oxidation of biomolecules and liver tissue injury (Jeyavani et al., 2022; Priyadharshini et al., 2024; Tian et al., 2024). In the present study, PP represented 7% of polymers analyzed, showing the global pattern on use of plastic for packaging and fishing materials (Bergmann et al., 2015; Jensen et al., 2019) and corroborating to the Australian trend as well (Jensen et al., 2019; Wootton et al., 2022).

Polyurethane (PU) is a synthetic polymer resistant to heat and difficult to decompose, due to disposal challenges (Choi et al., 2024). It is considered as one of the most hazardous polymers and appears in many forms, such as rigid and flexible foams, manufacturing of tires, gaskets, bumpers, fibers, plastic foam, synthetic leathers, jackets, adhesive, paints, sponges and cushions, rubber goods (Khan et al., 2017; Wang et al., 2024). In recent years, its extensive use in infrastructure construction is largely due to its ability to create a surface protective coatings in concrete (Dacuan et al., 2021). Although PU represents only 1% of the polymers identified in the present study, it is the sixth most used plastic globally (Kemona and Piotrowska, 2020). Moreover, studies have highlighted its susceptibility to fungal degradation in macro debris which can lead to surface cracking, erosion, pore formation or loss in tensile strength, contributing to multiplicate the microplastic pollution in the environment (Khan et al., 2017).

GFA, PAC, NY, NY-PU, MFA and PEG represent 16% of the polymers in the present study, but previous studies are more related to the effectiveness of those polymers (Mahmoud et al., 2017; Soleimani et al., 2023; Sparsø, 2014) and other topics (Zhao et al., 2024), than to their toxicity (Mejías et al., 2023; Zheng et al., 2022). For instance, PAC was assessed as a potential eco-friendly antifouling coating for aquaculture applications, but no toxicity tests were performed so far (Soleimani et al., 2023). Nylon is an important synthetic microfiber and are often detected in aquatic environments (Dharmaraj et al., 2021; Soleimani et al., 2023), however NY frequency was not representative in our study (2%).

4. Potential Impacts and Conservation Insights

MPs reaches coastal environments through several pathways and may include a wide range types, including different polymers, sizes, and chemical additives, making their management and mitigation highly complex (Peng et al., 2020; Zhang et al., 2020; Zhou et al., 2023). Stakeholders as government bodies, regulatory agencies, and the general public are globally concerned on the presence of plastic debris and MPs in coastal and marine ecosystems (Wootton et al., 2024). In response to such environmental challenges, intergovernmental negotiations for a legally binding instrument, named Plastic Treaty, are in progress (Gonçalves et al., 2024). These initiatives, together with sustainable development goals (SDG 14, for instance) and implementation of MPAs, seek to protect vital ecosystems while balancing human activities worldwide. For example, the Great Barrier Reef Marine Park (GBRMP), established in 1975 (McNeill, 1994), marked the beginning of Australia's efforts to mitigate threats to its marine environments while still allowing recreational use (Maestro et al., 2019). The presence of MPs and other pollutants impacts subsistence and cultural practices, such as oyster harvesting by Indigenous Australians. MPAs seek not only to protect the biodiversity itself, but also assure cultural and traditional practices that depend on the environment.

Indigenous Australians have been harvesting oysters along the Great Barrier Reef for around 9000 years, with a significant rise in exploitation noted approximately 3000 years ago, aligning with notable shifts in the archaeological record (Barker, 2004; Ulm et al., 2019). Across Australia, oysters historically served as a crucial resource, especially in coastal regions where natural intertidal beds initially provided abundance (Reid and Bone, 2020). Consuming contaminated bivalves poses health risks, as pollutants accumulate within marine food webs, affecting both Indigenous communities, who rely on these resources, and wider populations consuming shellfish (Dawson et al., 2023). Also, addressing the management of Australia's marine protected areas and recognizing Indigenous communities' rights are fundamental steps toward sustainable development (Isaac et al., 2024). Evidence shows that Aboriginal people successfully managed Australian landscapes for thousands of years, bringing a depth of knowledge valuable for modern conservation efforts (Reid and Bone, 2020). However, the complexity of MPs contamination—with their diverse sources, types, and impacts—poses significant challenges for MPAs (Nunes et al., 2023a, 2023b), especially in communities that rely on marine resources for their subsistence and cultural practices. This highlights the importance of MPAs in maintaining food security and cultural heritage between the increase occurrence and potential risks related to microplastic contamination. Therefore, this research can guide policymakers in establishing sustainable standards and regulations along Great Barrier Reef, and vulnerable Australian World Heritage Site.

5. Conclusions

The average concentrations of MPs differed between locations, with some areas exhibiting higher contamination levels. No significant correlation was found between microplastic levels and human modification (HMc), suggesting that factors such as local pollution sources and hydrodynamic conditions might be influencing contamination. The range of polymers detected in MPs indicates multiple sources of contamination, with cellulose from anthropogenic activities and polyester being the most common. When compared to other studies, the concentrations found in this research were consistent with those from coastal and urbanized regions globally, particularly concerning the types of MPs identified. However, the levels detected were lower than global averages, even in marine protected areas (MPAs). These results stress the importance of understanding how local conditions impact microplastic distribution within northern Australian MPAs. They also highlight that while MPAs can play a role in reducing contamination, additional measures are essential to minimize human impacts on marine ecosystems.

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Capítulo VII: Síntese da Discussão e Conclusões

Contemporaneamente os riscos ecológicos associados a contaminação por MPs, ainda não estão suficientemente claros a ponto de permitir o estabelecimento de limites ambientalmente seguros (Koelmans et al., 2023). Essas lacunas científicas limitam a formulação de diretrizes globais para mitigação e controle (Ohayon et al., 2021). A variabilidade nas abordagens metodológicas, como o uso de malhas de diferentes tamanhos e técnicas analíticas, dificulta a comparação e interpretação global dos dados (Kutralam-Muniasamy et al., 2021). Além disso, as características físicas e químicas dos MPs, como tamanho, forma e composição polimérica, aumentam a complexidade na avaliação de seus efeitos. Entretanto, há muitos estudos disponíveis na literatura científica especializada, indicando impactos em diferentes níveis de organização biológica (Cao et al., 2024). Portanto, o arcabouço de conhecimentos disponíveis permite inferir que a contaminação global por matérias a base de polímeros plásticos está entre os mais preocupantes desafios ambientais da modernidade.

Essa preocupação é reforçada pelos impactos já documentados em diferentes níveis biológicos, desde alterações moleculares até prejuízos celulares significativos. Em nível molecular e celular, os MPs podem causar estresse oxidativo, danos ao DNA e alterações em membranas celulares, frequentemente agravados por substâncias químicas adsorvidas (Khalid et al., 2021; Wang et al., 2021). Esses impactos escalam para níveis de organização ecologica mais altos, onde interferem na dinâmica de predação, competição e reprodução, resultando em mudanças estruturais nas comunidades ecológicas (Jaikumar et al., 2019; Ugwu et al., 2021). De fato, MPs foram reconhecidos como um dos contaminantes de preocupação emergentes que mais ameaçam a biodiversidade marinha (Khalid et al., 2021). Espécies como bivalves filtradores são particularmente vulneráveis, o que faz com que sejam amplamente utilizadas como sentinelas destes contaminantes (Kazour and Amara, 2020), principalmente devido ao fato de estarem diretamente em contato com matrizes ambientais onde os MPs sao encontrados em grandes quantidades (Li et al., 2019).

Correntes oceânicas e ventos contribuem para a ampla dispersão dos MPs, permitindo que esses contaminantes alcancem áreas remotas e protegidas (Brahney et al., 2020). Nesse contexto, avaliar a presença e os níveis de MPs em AMPs é crucial para subsidiar políticas públicas e estratégias mitigatórias. Entre 2017 e 2022, 254 AMPs em diferentes partes do mundo apresentaram registros de contaminação, em amostras de água do mar, sedimento e biota (Nunes et al., 2023a; 2023b). AMPs de proteção integral, mais restritivas quanto às atividades humanas, geralmente apresentam maior eficácia na contenção de MPs (Day et al., 2012; Dudley, 2008). Contudo, fatores externos, como correntes oceânicas, ventos e proximidade de fontes de poluição, comprometem sua efetividade (Brahney et al., 2020; Vieira et al., 2021). Em contraste, áreas de uso sustentável frequentemente exibem níveis mais elevados de contaminação devido à permissão de atividades como pesca e turismo (Kazour and Amara, 2020; Nunes et al., 2023b).

Estudos em dez áreas de proteção integral no Brasil mostraram contaminação em bivalves filtradores independentemente da categoria de manejo, com concentrações mais elevadas no Refúgio de Vida Selvagem do Arquipélago de Alcatrazes, enquanto Parques Nacionais e Reservas Naturais apresentaram níveis ligeiramente mais baixos e sem diferença estatística. Fatores locais e regionais também desempenham papel importante na distribuição de MPs, principalmente em estuários e regiões costeiras urbanizadas, que destacam-se pela alta contaminação decorrente de escoamento urbano e descargas de efluentes (Liao et al., 2021; Vieira et al., 2021). De forma semelhante, no nordeste da Austrália, os tipos de polímeros encontrados em AMPs sugerem múltiplas fontes de contaminação, compatíveis com regiões costeiras urbanizadas. Em ambos os países, não foi observada correlação significativa entre as concentrações de MPs e os níveis de urbanização, sugerindo que condições hidrodinâmicas e fontes locais podem ser determinantes. Embora esses achados reforcem o papel das áreas de proteção integral na mitigação da contaminação por MPs, é importante considerar que a estrutura e o funcionamento dessas áreas variam significativamente entre os países. Cada região adota sistemas de manejo que refletem suas particularidades socioambientais, culturais e legislativas, o que pode influenciar a vulnerabilidade e a efetividade das AMPs. Essa diversidade estrutural implica que os dados obtidos em áreas protegidas brasileiras e australianas devem ser interpretados com cautela e podem não ser diretamente extrapoláveis para outras regiões sem uma análise criteriosa das diferenças nas práticas de uso e proteção ambiental em escala global.

De forma geral, verificou-se que as áreas de proteção integral avaliadas em ambos os países apresentaram níveis de contaminação por MPs abaixo dos valores globais quando comparadas com áreas sem proteção ou de uso múltiplo. Esses resultados sugerem que, apesar das diferenças estruturais, as áreas protegidas estudadas oferecem algum nível de proteção contra a contaminação. Contudo, esforços adicionais de manejo são necessários para alcançar níveis de proteção compatíveis com a 11ª Meta de Aichi, que visa conservar áreas importantes para a biodiversidade de maneira eficaz e equitativa.

A interação entre MPs e outros estressores ambientais, como mudanças climáticas e acidificação dos oceanos, exacerba os impactos negativos na biodiversidade marinha e dificulta ainda mais a gestão de contaminantes emergentes. As mudanças climáticas influenciam diretamente os padrões de circulação oceânica e a distribuição de MPs, ampliando sua dispersão para áreas remotas, incluindo regiões polares e AMPs remotas (Brahney et al., 2020; Evangeliou et al., 2020). A acidificação dos oceanos, resultante do aumento da absorção de dióxido de carbono atmosférico pelas águas marinhas, representa uma ameaça crescente aos ecossistemas aquáticos, alterando processos biogeoquímicos e comprometendo a sobrevivência de diversas espécies (Qu et al., 2025; Zheng et al., 2023). A acidificação dos oceanos, por sua vez, pode modificar as propriedades químicas dos MPs e de contaminantes adsorvidos em sua superfície, potencializando efeitos tóxicos em organismos marinhos (Balbela et al., 2024; Wang and Chen, 2023). Essas condições também afetam organismos filtradores, como bivalves, que já estão sob pressão devido à bioacumulação de MPs. A exposição simultânea a MPs, temperaturas mais altas e águas acidificadas pode reduzir a taxa de crescimento, alterar o comportamento alimentar e comprometer processos reprodutivos em diversas espécies (Capo et al., 2021; Vasanthi et al., 2021). Além disso, as sinergias entre MPs e outros poluentes, como metais pesados e compostos orgânicos persistentes, representam um risco adicional (Liu et al., 2024). Os MPs podem atuar como vetores de transporte desses contaminantes, facilitando sua biodisponibilidade e aumentando os impactos em diferentes níveis tróficos (Khalid et al., 2021). Essas interações destacam a necessidade de uma abordagem holística para compreender e mitigar os impactos dos MPs em um contexto de múltiplos fatores de estresse. Estratégias de manejo integradas que considerem essas sinergias podem fornecer informações mais robustas para subsidiar políticas públicas globais, como o post-2020 Global Biodiversity Framework, e promover ações que minimizem os impactos cumulativos em ecossistemas marinhos.

Os resultados desta tese podem fornecer informações valiosas para subsidiar regulamentações no âmbito do Tratado Global dos Plásticos, especialmente no contexto dos países do Sul global. O tratado, que busca reduzir os impactos ambientais do plástico em escala global, exige uma base científica robusta para o estabelecimento de metas específicas e mecanismos de implementação. Estudos como os apresentados aqui, que destacam os níveis de contaminação por MPs em diferentes regiões e categorias de AMPs, são essenciais para embasar decisões políticas e identificar hotspots de poluição que requerem ações prioritárias (Nunes et al., 2023a, 2023b). Nos países do Sul global, onde a infraestrutura de gestão de resíduos ainda é limitada , a implementação de regulamentações como redução na produção e descarte correto, propostos pelo tratado, enfrenta desafios significativos (Kushwaha et al., 2024). No entanto, os dados apresentados nesta tese podem ajudar a direcionar esforços para áreas críticas, promovendo a alocação mais eficiente de recursos e o desenvolvimento de estratégias adaptadas às necessidades locais.

Os resultados obtidos reforçam a relevância da pesquisa cientifica para orientar tanto politicas publicas quanto o atingimento das metas do Tratado Global dos Plásticos, especialmente em países do Sul global, onde as ações de mitigação são mais urgentes e desafiadoras. Dessa forma, medidas como monitoramentos regulares e padronizados a longo prazo, mapeamento de fontes, fortalecimento da gestão integrada, zonas de amortecimento, fomento à pesquisa e educação ambiental, são algumas abordagens que permitem identificar tendencias de contaminação, auxiliam a alocação eficiente de recursos, descarte correto, compreender melhor os impactos de forma a desenvolver estratégias eficazes e que devem ser empreendidos nessas áreas altamente vulneráveis, avaliando simultaneamente o risco ecológico a partir dos níveis de base aqui apresentados. Tal abordagem será ainda mais relevante considerando o arcabouço regulatório que potencialmente derivará das decisões tomadas junto ao Tratado Global dos Plásticos.

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