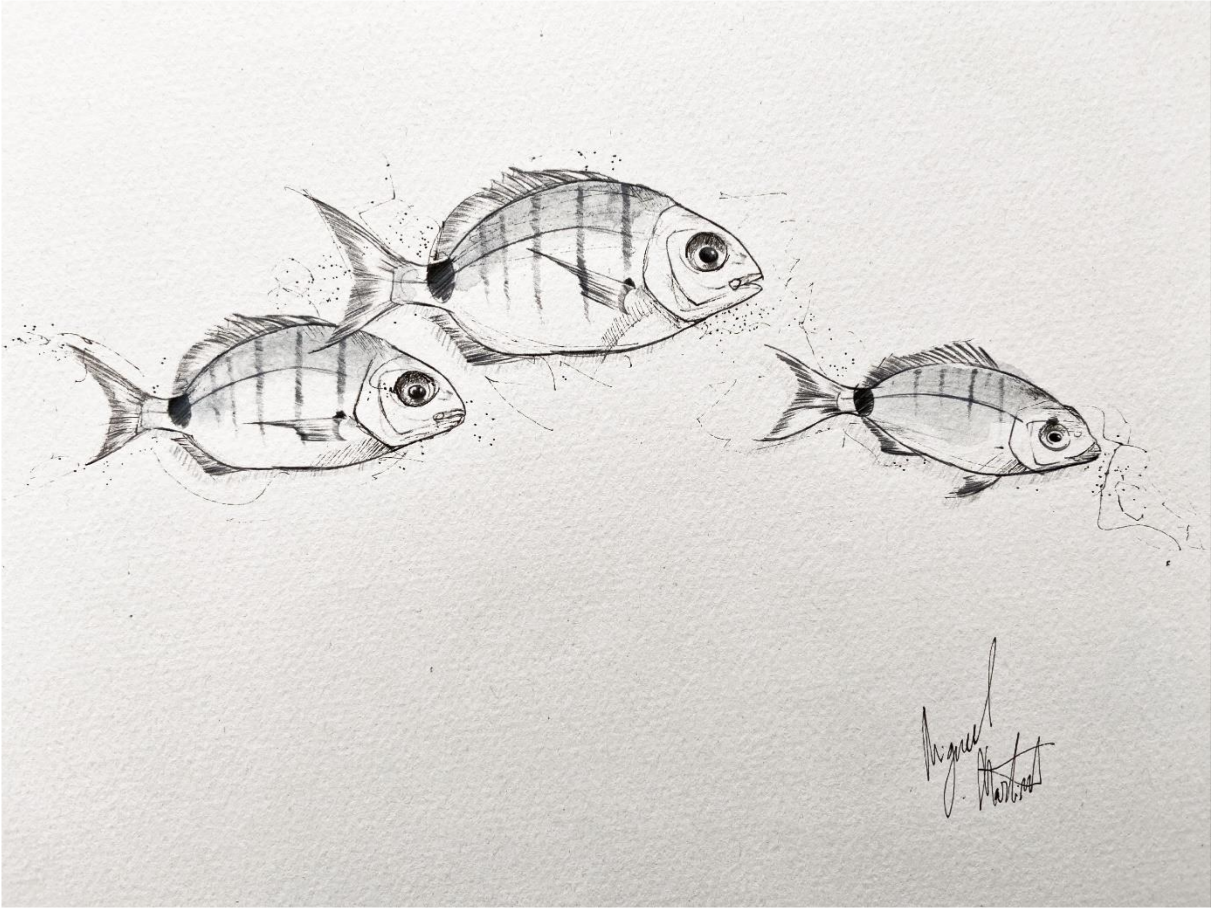

IMErSÃO

Impact of **M**icroplastic on **E**arly Life-History Stages of **S**eabream in the **A**tlantic **O**cean

Carolin Müller

© *Diplodus sargus* sketch, title page: Miguel Martins, Estúdio Onze, Faro, 2020.

Impact of Microplastic on Early Life-History Stages of Seabream in the Atlantic Ocean

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“’Plastic Man’ will come into a world of colour and bright shining surfaces… he is surrounded on every side by this tough, safe, clean material which human thought has created. ... It is a world free from moth and rust and full of colour, a world largely built up of synthetic materials made from the most universally distributed substances, ... a world in which man, like a magician, makes what he wants for almost every need, out of what is beneath him and around him, coal, water and air. ... We shall see growing up around us a new, brighter, cleaner and more beautiful world, ... the perfect expression of the new spirit of planned scientific control, the Plastics Age.”

Victor Emmanuel Yarsley & Edward Gordon Couzens (1941): Plastics

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Summary

In the light of global habitat and biodiversity loss, holistic research on the species- and life-stage-specific ecological, physiological, and behavioural responses to ambient conditions is required to evaluate the actual extent of interference, notably in nearshore environments which are exposed to anthropogenically driven changes while simultaneously offering essential ecosystem goods and services. One of these coastal ecosystem services is the provision of suitable habitats promoting the survival of ichthyofaunal early life-history stages, thereby enhancing the likelihood of recruitment to adult populations. Hence, in-depth understanding of the impact of environmental gradients and human-induced stressors on the growth and condition of larval and juvenile fish in coastal nurseries is of essential importance for the successful implementation of conservation measures as well as the sustainable exploitation of coastal fish stocks.

Plastic litter has been entering the marine realm ever since the mass production of synthetic polymers began in the 1950's, thereby potentially complementing and amplifying the effects of persistent stressors like ocean warming, acidification, overexploitation or habitat degradation. Despite the fact that first reports on plastic ingestion by marine fish date back to the 1970's and that considerable scientific and public concern has been raised recently over the potential detrimental effects of micro-sized (i.e., < 5 mm) plastic items, research on the explanatory variables for uptake as well as on the physiological effects of exposure and ingestion is still at an early stage, impeding a thorough evaluation of the actual implications for living marine resources.

The present thesis addresses the aforementioned knowledge gaps, by investigating the effects of microplastic (MP) exposure and uptake on growth, condition, and survival of early life-history stages of fish. It provides a critical analysis of the current state of the art on the extent of MP ingestion by marine fish, along with an examination of the effects and explanatory variables for MP uptake based on a literature review of 90 *in-situ* studies published between 1972 and 2019 (Chapter 1). This review simultaneously lays the groundwork for an integrated research design, combining an *in-situ* (Chapter 2) and an *in-vivo* (Chapter 3) study, to determine the impacts of and driving factors for MP ingestion by nursery-dependent, omnivorous juveniles of a commercially and recreationally important fish species, the white seabream *Diplodus sargus* (Linnaeus, 1758) in the Ria Formosa lagoon, Portugal.

Synthesising the key findings of the present thesis, it becomes apparent that ichthyofaunal early life-history stages of fish are underrepresented in scientific initiatives investigating the extent of marine MP pollution (Chapter 1), notwithstanding the vulnerability of larval and juvenile fish to environmental changes and anthropogenic disturbances, as well as their vital role in population dynamics. Moreover, the existing literature rarely contextualised the finding of MP in the gastrointestinal tract (GIT) of a fish with the ingestion and environmental availability of natural and artificial prey items. Collating the results for all taxa and life-history stages, around 1/3 of all individuals assessed were found with an average of 2.2 ± 4.02 MP items in their GIT (Chapter 1).

The *in-situ* and the *in-vivo* part of this research project verified the uptake of both synthetic fibres and particles by juvenile white seabream (Chapter 2 and 3). The majority of plastic-feeding *D. sargus* ingested ≤ 5 MP items; the low MP abundances in the GIT did not compromise the continuous uptake and digestion of natural prey. The ingestion of synthetic fibres was correlated to the uptake of plant material and detritus (Chapter 2), suggesting an accidental ingestion of fibrous plastic attached to or incorporated in prey of non-animal origin. Although minor MP uptake rates did not cause detrimental effects on condition, growth, and survival, feeding habits of certain individuals may lead to disproportionately high MP ingestion rates (Chapter 3), posing the potential risk of GIT blockage, reduced uptake of natural prey and consequently impaired body condition.

Evidence from the *in-situ* study (Chapter 2) emphasise that ontogenetic dietary shifts (e.g., from planktic to benthic prey) along with individual- and species-specific feeding habits may have a stronger explanatory power when assessing driving factors for MP ingestion than trophic guild alone. Despite its generalised, opportunistic feeding mode, *D. sargus* shows a specialisation in the prey uptake and food handling, enabling this fish species to distinguish inedible, hard-bodied items such as benthic gastropod shells, larger grains of sand or solid MP fragments from edible, soft-bodied prey already at an early developmental stage (Chapter 2 and 3).

In conclusion, the results presented herein challenge the existing hypotheses that omnivores in general and early life-history stages of fish growing up in coastal nurseries in particular are prone to elevated MP ingestion rates. Moreover, the notion of an inevitable deterioration of physiological condition upon uptake of synthetic polymers was refuted. The substantiated high inter-individual differences in trophic resource utilisation necessitate adequate sample sizes along with a contextualisation of GIT contents with environmental prey and MP availabilities. The outcomes further argue for the holistic integration of field and laboratory studies to determine the species- and life-stage specific driving factors for and potential detrimental effects of MP ingestion to enable a thorough evaluation of the ecological and economic risk arising for coastal fish stocks and fisheries.

Zusammenfassung

Angesichts des weltweiten Verlusts an Lebensraum und biologischer Vielfalt ist eine ganzheitliche Erforschung der arten- und lebensphasenspezifischen ökologischen, physiologischen und verhaltensbezogenen Reaktionen auf die Umgebungsbedingungen erforderlich, um das tatsächliche Ausmaß der Beeinträchtigung zu bewerten, insbesondere in küstennahen Ökosystemen, die in besonderem Maße anthropogenen Einflüssen ausgesetzt sind, und gleichzeitig die Versorgung mit wichtigen ökosystemaren Gütern und Dienstleistungen sichern. Eine dieser Leistungen des Küstenökosystems ist die Bereitstellung geeigneter Lebensräume, die das Überleben früher Entwicklungsstadien von Fischen fördern und damit die Rekrutierungswahrscheinlichkeit erwachsener Bestände erhöhen. Daher ist ein tiefgehendes Verständnis der Auswirkungen von Umweltgradienten sowie vom Menschen verursachten Stressfaktoren auf das Wachstum und die Kondition von Larven und Jungfischen in litoralen Kinderstuben von entscheidender Bedeutung für die erfolgreiche Umsetzung von Erhaltungsmaßnahmen und die nachhaltige Nutzung küstennaher Fischbestände.

Seit Beginn der Massenproduktion synthetischer Polymere in den 1950er Jahren gelangen große Mengen an Plastikmüll in die Weltmeere, wodurch die Auswirkungen anhaltender Stressfaktoren wie Ozeanerwärmung und -versauerung, Überfischung oder Lebensraumdegradierung potenziell verstärkt werden. Obwohl die ersten Berichte über die Aufnahme von Kunststoffen durch Meeresfische bereits in den 1970er Jahren veröffentlicht wurden, und heute sowohl in der Wissenschaft als auch in der Öffentlichkeit die potenziellen Gefahren von Mikroplastik (d.h. < 5mm) für Meerestiere große Aufmerksamkeit erfahren, befindet sich die Forschung zu den die Aufnahme bestimmenden Einflussfaktoren sowie zu den physiologischen Folgen noch in einem frühen Stadium, wodurch eine gründliche Bewertung der tatsächlichen Auswirkungen auf die lebenden Meeresressourcen erschwert wird.

Die vorliegende Arbeit befasst sich mit den zuvor aufgeführten Wissenslücken, indem sie die Auswirkungen der Exposition und Aufnahme von Mikroplastik (MP) auf das Wachstum, die Kondition und das Überleben früher Lebensstadien von Fischen untersucht. Sie bietet eine kritische Analyse des aktuellen Wissensstandes über das Ausmaß der MP-Aufnahme durch Meeresfische sowie eine Untersuchung der Auswirkungen und erklärenden Variablen für die MP-Aufnahme auf Grundlage einer 90 Studien umfassenden Literaturrecherche (Kapitel 1). Diese Literaturübersicht bildet gleichzeitig die Grundlage für ein integriertes Forschungsdesign, welches eine Feld- (Kapitel 2) und eine Laborstudie (Kapitel 3) kombiniert, um die Auswirkungen und treibenden Faktoren für die MP-Aufnahme durch omnivore

Jungfische einer kommerziell relevanten Fischart, der Geißbrasse *Diplodus sargus* (Linnaeus, 1758), welche in küstennahen Kinderstuben der Ria Formosa Lagune (Portugal) aufwächst, zu ermitteln.

Die Schlüsselresultate dieser Arbeit belegen, dass frühe Lebensstadien von Fischen in vorherig publizierten (1972 – 2019) wissenschaftlichen Untersuchungen zum Ausmaß der Meeresverschmutzung durch MP unterrepräsentiert sind (Kapitel 1), obwohl Larven und Jungfische gegenüber Umweltveränderungen und anthropogenen Störungen besonders anfällig sind und eine wichtige Rolle in der Populationsdynamik spielen. Darüber hinaus wurde in der vorhandenen Literatur der Nachweis von MP im Magen-Darm-Trakt (MDT) eines Fisches nur selten mit der Aufnahme und der Umweltverfügbarkeit natürlicher und künstlicher Beute in Zusammenhang gebracht. Fasst man die Ergebnisse für alle Taxa und Lebensstadien zusammen, so wurden bei etwa einem Drittel aller untersuchten Individuen durchschnittlich 2,2 ± 4,02 MP Partikel im MDT nachgewiesen (Kapitel 1).

Im Feld- und Laborteil dieses Forschungsprojekts wurde die Aufnahme von Kunststofffasern und -partikeln durch juvenile Meerbrassen nachgewiesen (Kapitel 2 und 3). Die Mehrheit der plastikfressenden *D. sargus* nahm ≤ 5 MP auf; die geringen MP-Mengen im MDT beeinträchtigten die kontinuierliche Aufnahme und Verdauung natürlicher Beute nicht. Die Korrelation in der Ingestion von synthetischen Fasern mit Pflanzenmaterial und Detritus (Kapitel 2) deutet auf eine unbeabsichtigte Aufnahme von Kunststofffasern hin, die an Nahrung nicht-tierischen Ursprungs angeheftet sein können. Obwohl geringe MP-Aufnahmeraten keine nachteiligen Auswirkungen auf Kondition, Wachstum und Überleben der Jungfische hatten, können die Nahrungspräferenzen einzelner Individuen zu unverhältnismäßig hohen MP-Aufnahmeraten führen (Kapitel 3), was das potenzielle Risiko einer Verstopfung des MDT, einer verringerten Aufnahme natürlicher Beutetiere und folglich einer Beeinträchtigung der Körperkondition birgt.

Die Erkenntnisse aus der Feldstudie (Kapitel 2) unterstreichen, dass ontogenetische Nahrungsumstellungen (z. B. von planktischer zu benthischer Beute) zusammen mit individuen- und artspezifischen Ernährungsgewohnheiten eine stärkere Aussagekraft bei der Bewertung der treibenden Faktoren für die MP-Aufnahme haben können als die trophische Ebene eines Organismus vermuten ließe. Trotz einer generalisierten, opportunistischen Ernährungsweise zeigt die Geißbrasse eine Spezialisierung bei der Beuteaufnahme und Nahrungsverarbeitung, die es dieser Fischart ermöglicht, bereits in einem frühen Entwicklungsstadium unverdaubare Festkörper wie benthische Schneckenschalen, größere

Sandkörner oder feste MP-Fragmente von essbarer, weicher Beute zu unterscheiden (Kapitel 2 und 3).

Zusammenfassend lässt sich sagen, dass die hier vorgestellten Ergebnisse die bestehenden Hypothesen in Frage stellen, wonach omnivore Fische im Allgemeinen und frühe Lebensstadien, die in küstennahen Kinderstuben aufwachsen im Besonderen, für eine erhöhte MP-Aufnahme anfällig sind. Darüber hinaus wurde die Vorstellung einer zwangsläufigen Verschlechterung des physiologischen Zustands bei Aufnahme synthetischer Polymere widerlegt. Die nachgewiesenen großen inter-individuellen Unterschiede in der Nutzung der trophischen Ressourcen erfordern eine angemessene Stichprobengröße sowie eine Kontextualisierung der MDT-Inhalte mit den in der Umwelt vorhandenen Beute natürlichen und künstlichen Ursprungs. Die Ergebnisse sprechen außerdem für eine ganzheitliche Integration von Feld- und Laborstudien, um die arten- und lebensphasenspezifischen treibenden Faktoren für die Aufnahme von MP und ihre potenziell schädlichen Auswirkungen zu bestimmen, um eine gründliche Bewertung der ökologischen und wirtschaftlichen Risiken für die Fischbestände und die Fischerei in Küstengebieten zu ermöglichen.

Resumo

Considerando a perda global de habitat e biodiversidade, uma investigação holística sobre as respostas ecológicas, fisiológicas e comportamentais específicas das espécies e estágio de vida às condições ambientais se faz necessária para avaliar a extensão real das interferências, especialmente em ambientes próximos à costa que estão expostos à alterações de origem antropogénica, oferecendo simultaneamente bens e serviços essenciais para o ecossistema. Um destes serviços é o fornecimento de habitats adequados de modo a promover a sobrevivência dos estágios iniciais da história de vida da ictiofauna, aumentando assim a probabilidade de recrutamento para populações adultas. Portanto, uma compreensão profunda do impacto dos gradientes ambientais e dos factores de estresse induzidos pelo homem no crescimento e condição das larvas e peixes juvenis em viveiros costeiros, é essencialmente importante para a o sucesso da implementação de medidas de conservação, bem como para a exploração sustentável dos recursos haliêuticos costeiros.

O lixo plástico vem sendo inserido nos ambientes marinhos desde que a produção em massa de polímeros sintéticos começou nos anos 50, complementando e amplificando potencialmente os efeitos de factores de stress persistentes como o aquecimento dos oceanos, a acidificação, a sobre-exploração ou a degradação do habitat. Apesar dos primeiros registos sobre a ingestão de plástico por peixes marinhos datarem dos anos 70 e de uma considerável preocupação científica e pública ter sido levantada recentemente sobre o potencial prejuízo dos artigos de plástico de micro dimensões (isto é, < 5 mm), a investigação sobre as variáveis explicativas para a absorção, bem como os efeitos fisiológicos da exposição e ingestão, ainda se encontra numa fase inicial, impedindo uma avaliação minuciosa das reais implicações para os organismos marinhos vivos.

A presente tese aborda as lacunas de conhecimento acima mencionadas, investigando os efeitos da exposição e absorção do microplástico (MP) no crescimento, condição e sobrevivência os estágios iniciais da história de vida dos peixes. Fornece uma análise crítica da situação actual da ingestão de MP por peixes marinhos, juntamente com um availiação dos efeitos e variáveis explicativas da absorção de MP com base numa revisão bibliográfica de 90 estudos in situ publicados entre 1972 e 2019 (Capítulo 1). Esta revisão estabelece simultaneamente as bases para um desenho de investigação integrado, combinando um estudo *in-situ* (Capítulo 2) e *in-vivo* (Capítulo 3), para determinar os impactos e os factores impulsionadores da ingestão de MP por juvenis omnívoros de uma espécie de peixe comercialmente relevante, o Sargo *Diplodus sargus* (Linnaeus, 1758), que cresce em viveiros próximos à lagoa da Ria Formosa (Portugal).

Sintetizando os principais resultados da presente tese, torna-se evidente que as fases iniciais da história de vida dos peixes estão sub-representadas em iniciativas científicas as quais investigam a dimensão da poluição marinha por MP (Capítulo 1), apesar da vulnerabilidade das fases larvais e juvenis às alterações ambientais e perturbações antropogénicas, bem como o seu papel vital na dinâmica populacional. Além disso, a literatura existente raramente contextualizou a ocorrência de MP no trato gastrointestinal (TGI) de um peixe com a ingestão e a disponibilidade ambiental de presas naturais e artificiais. Confrontando os resultados para todos os taxas e fases da história de vida, cerca de 1/3 de todos os indivíduos avaliados foram encontrados com uma média de 2,2 ± 4,02 MP de itens no seu TGI (Capítulo 1).

A parte in-situ e in-vivo deste projecto de investigação verificou a absorção tanto de fibras sintéticas como de partículas por juvenis de dourada branca (Capítulos 2 e 3). A maioria dos *D. sargus* que se alimentaram de plástico ingeriu ≤ 5 itens de MP; a baixa abundância de MP no TGI não comprometeu a contínua absorção e digestão das presas naturais. A ingestão de fibras sintéticas foi correlacionada com a absorção de material vegetal e detritos (Capítulo 2), sugerindo uma ingestão acidental de plástico fibroso ligado ou incorporado em presas de origem não animal. As menores taxas de absorção de MP não causaram efeitos prejudiciais sobre a condição, crescimento e sobrevivência, contudo, os hábitos alimentares de certos indivíduos podem levar a taxas de ingestão desproporcionadamente elevadas de MP (Capítulo 3), representando o risco potencial de bloqueio do TGI, redução da absorção de presas naturais e, consequentemente, deterioração da condição corporal.

As evidências do estudo in-situ (Capítulo 2) enfatizam que as mudanças alimentares ontogenéticas (por exemplo, desde presas planctônicas à bentónicas) juntamente com os hábitos alimentares individuais e específicos de cada espécie podem ter um poder explicativo mais forte quando se avaliam os factores impulsionadores da ingestão de MP do que a guilda trófica por si só. Apesar do seu modo de alimentação generalizado e oportunista, *D. sargus* mostra uma especialização na captação de presas e no manuseamento de alimentos, permitindo a esta espécie de peixe distinguir itens não comestíveis, e de composição dura, tais como conchas de gastrópodes bentónicos, grãos maiores de areia ou fragmentos sólidos de MP de presas comestíveis e de corpo mole já num estágio precoce de desenvolvimento (Capítulos 2 e 3).

Em conclusão, os resultados aqui apresentados desafiam as hipóteses existentes de que os omnívoros em geral e os estágios iniciais da história de vida dos peixes que crescem em viveiros costeiros, em particular, são propensos a taxas elevadas de ingestão de MP. Além disso, foi refutada a ideia de uma inevitável deterioração da condição fisiológica após a absorção de polímeros sintéticos. As elevadas diferenças fundamentadas entre indivíduos na utilização de recursos tróficos exigem tamanhos de amostra adequados juntamente com uma contextualização do conteúdo gastrointestinal, com disponibilidade de presas ambientais e MP. Os resultados defendem ainda a integração holística de estudos de campo e laboratorial para determinar os factores específicos das espécies e estágio de vida que motivam a ingestão de MP e seus potenciais efeitos prejudiciais para permitir uma avaliação exaustiva dos riscos ecológicos e económicos que surgem para as populações de peixes costeiros e a pesca.

1 Introduction

In this introduction, the early life-history stages (ELHS) of marine fish and the factors influencing recruitment are presented, with particular emphasis on the role of transitional zones, namely coastal lagoons and estuaries, as nurseries, and the potential effects of microplastic ingestion on ELHS. An overview of the study site, the Ria Formosa lagoon in the south of Portugal, an important nursery for many commercial species and source of recruits to coastal fisheries is given. The choice of the white seabream, *Diplodus sargus* (Linnaeus, 1758) as the model species for the study is justified based on its biology, ecology, and commercial as well as recreational importance. Finally, the research objectives and scientific questions addressed by the thesis are presented, along with the structure of the thesis.

1.1 Early life-history stages of fish

Advancing the understanding of the ecology and evolution of a species is inextricably linked to the holistic assessment of its life history, i.e., the age- or stage-specific patterns of survival, growth, maturation, and reproduction (Stearns 1992; Hutchings 2002; Braendle et al. 2011). Life history research approaches the interindividual and interspecific variability of quantitative, demographic fitness parameters (e.g., size at birth, number of offspring, age, and size at maturity) in response to environmental, ecological, physiological, and genetic conditions along with the constraining trade-offs between survival and reproduction (Roff 1992; Charnov 1993; Denney et al. 2002). As population growth is fundamentally associated with individual life-history traits, life-history research is thus considered essential for the management and conservation of flora (Aronne 2017) and fauna, ranging from terrestrial mammals (Wich et al. 2008) to aquatic invertebrates (Bauer 1991; Moore et al. 2013), as well as freshwater and marine fish (Heithaus and Laushman 1997; Foster and Vincent 2004; Young et al. 2006).

Fish, with more than 35,000 valid species described (Fricke et al. 2020; Froese and Pauly 2021), exhibit a level of biodiversity which is unparalleled among terrestrial vertebrates. Along with their taxonomic, ecological, and behavioural plasticity come developmental and morphological varieties which can be best understood when considering that upon their discovery, ELHS of several fish taxa were primarily assigned to their own genus (e.g., *Leptocephalus, Acronurus*) due to their dissimilarity with their adult forms (Fuiman 2002). Generally, the life-history of most fish taxa can be separated into five more or less morphologically and behaviourally distinct stages (Fig. 1) which may vary greatly in their ecological requirements: the egg (or embryo), larva, juvenile, adult and senescent (Hempel

1979; Patnaik et al. 1994; Fuiman 2002; Reznick et al. 2009; Miller and Kendall 2009). ELHS of fish (i.e., the embryo, larva, and juvenile stages) experience major developmental, morphological, and behavioural transitions which make them particularly sensitive to biotic and abiotic factors directly impacting their survival (Hjort 1914; Houde 1987; Cushing 1995; Chambers and Trippel 1997).

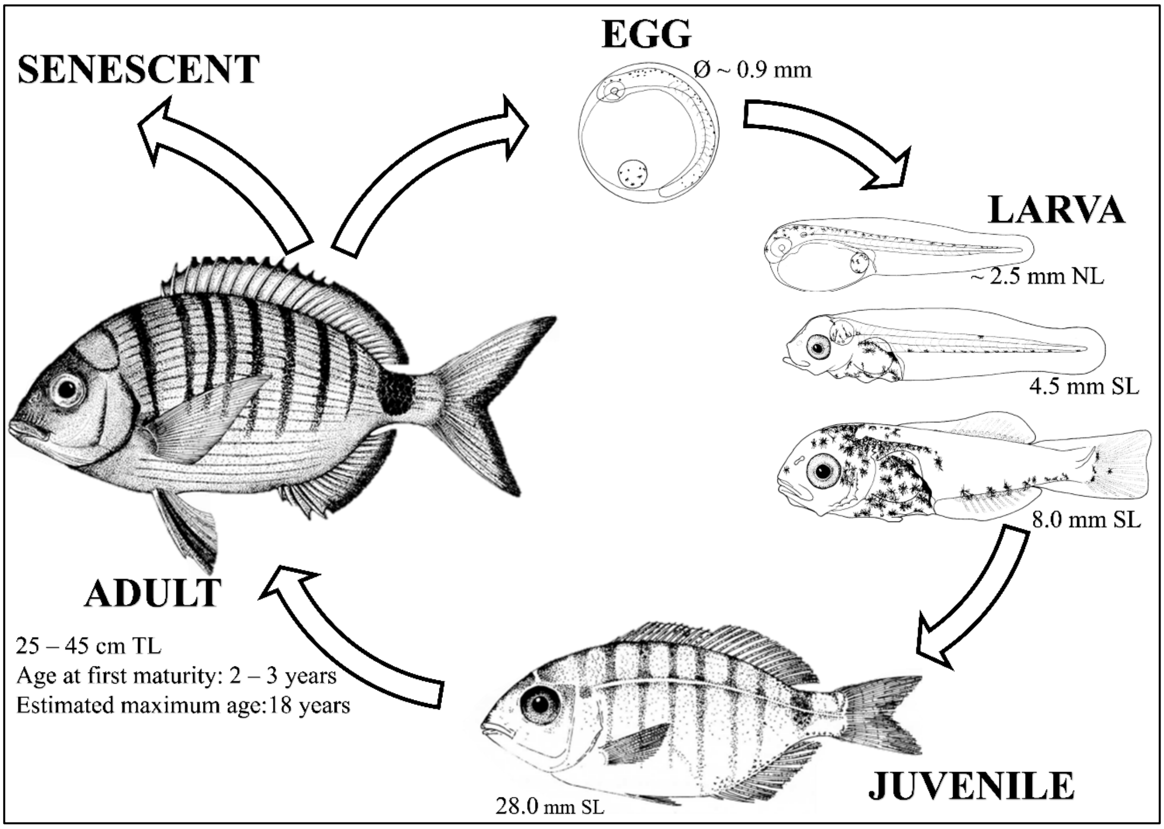

Fig. 1 Generalized life cycle of a fish, using *Diplodus sargus* as an example. Five life-history stages can be identified, the egg or embryonic stage, the larval stage (including several sub-stages), the juvenile stage, the adult stage, and the senescent stage. Abbreviations: NL = notochord length; SL = standard length; TL = total length. Illustrations taken from Brownell (1979); Bauchot (1987); Arias García and Moyano (1990). Data on growth and age: Bauchot (1987); Arias García and Moyano (1990); Abecasis et al. (2008); Mouine et al. (2011).

Besides high interspecific variability, key life-history traits may differ within and among populations of the same species, and spatial variation of such traits may be higher across small geographical scales than across large ones, resulting in fluctuations in the abundance of fish stocks (Hutchings 2002; Di Franco and Guidetti 2011). Early attempts to explain the variation in fish population size revolved around migration behaviour and fishing pressure as the primary driving factors (as retrospectively summarised by Houde 2008), however, this notion was revised by the discovery that reproductive success, precisely the variability of ELHS recruitment[1], has a stronger impact on year-class strength than the aforementioned parameters. Variability in mortality rates of the early larval stage was henceforth proposed to be the main determinant for interannual fluctuations in stock size.

Over the past century, a number of processes and theories governing recruitment have been proposed, that fall into two main categories: “mechanistic theories” and “synthesis theories” (Cole and McGlade 1998). The first mechanistic theory was that of Hjort (1914, 1926) who proposed the shift from endogenous to exogenous feeding (i.e., after adsorption of the yolk) to be the critical period in the early life-history of a fish during which the survival of the larva is dependent on the availability of sufficient densities of suitable prey, such as the copepod *Calanus finmarchicus*, for many gadoid species in the North Sea (Cushing 1984). Besides this trophodynamic impact on larval survival rates, Hjort (1914) suggested hydrodynamics may play a key role in recruitment variability as fish larvae may be passively dispersed by currents to unfavourable areas where growth, survival and consequently recruitment to the adult population are impeded. Hjort’s “Critical Period Hypothesis” along with his “Aberrant Drift Hypothesis” set the stage for more than 100 years of fisheries research which broadened the focus beyond first-feeding larvae and took into consideration multiple, interrelated factors (i.e., synthesis theories), to exert varying levels of impact on the different fish life-history stages (Houde 2008). Apart from the key tropho- and hydrodynamic processes (Fig. 2) that have since been proposed by various researchers (e.g., Cushing 1974, 1990; Lasker 1978; Sale 1978; Cury and Roy 1989; Govoni and Grimes 1992; Morote et al. 2010), correlations between recruit abundance (i.e., ELHS survival rates) and spawning stock characteristics (e.g., stock size / biomass; timing and location of spawning; age, size, and condition of spawning females) have also been recognised for some populations (Ricker 1954; Iles and Sinclair 1982; Cardinale et al. 2009; Hixon et al. 2014).

Additional abiotic and biotic factors such as temperature (e.g., Pepin 1991; Francis 1993, 1994; Stige et al. 2013), salinity (e.g., Alderice and Forrester 1971; Cushing 1995), dissolved oxygen (e.g., Ekau and Verheye 2005), predation (e.g., Bailey and Houde 1989; van der Veer et al. 1990), competition (e.g., Welker et al. 1994; Garvey and Stein 1998), as well as the availability of suitable habitats (Boehlert and Mundy 1988; Beck et al. 2001) are recognised as

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[1] The term ‘recruitment’ has different connotations (see Miller and Kendall 2009); in general, it can be defined as the number of fish of a cohort that are alive at a specific point in time during their lifetime, such as settlement in a nursery or when they become large enough to be caught by a commercial fishery.

essential factors shaping the survival probability of fish during their settlement and post-settlement phases (Fig. 2).

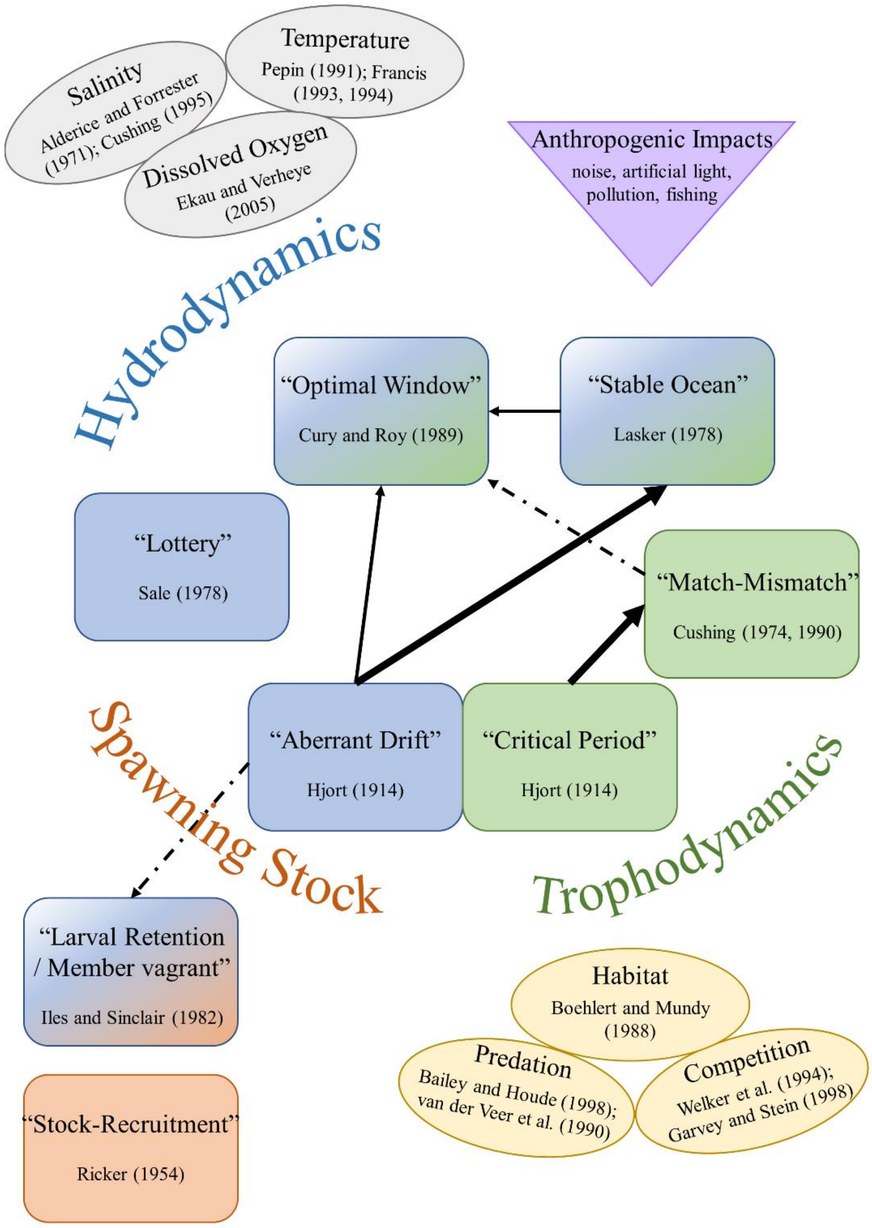

Fig. 2 Hypotheses on causes for recruitment variability along with biotic, abiotic and anthropogenic factors contributing to fluctuations in survival probability of early life-history stages of fish (including non-exhaustive list of exemplary references). Colour codes: blue = hydrodynamic processes; green = trophodynamic processes; orange = spawning stock characteristics; grey = abiotic factors; yellow = biotic factors; purple = anthropogenic factors. Solid arrows indicate direct extensions of Hjort's (1914) initial hypotheses, dashed arrows indicate indirect derivation. Thickness of arrows highlights strength of relationship between hypotheses. After Houde (2008) and Pritt et al. (2014).

Moreover, human-induced stressors such as noise (Simpson et al. 2016), artificial light at night (O'Connor et al. 2019), waterborne chemical and plastic pollutants (Naidoo and Glassom 2019; Besson et al. 2020; Pannetier et al. 2020), but also fishing pressure (Anderson et al. 2008; Perry et al. 2010), have been shown to either directly or indirectly affect offspring quality and the development, condition, and survival of fish ELHS; therefore, these human interferences also entail the potential to concomitantly alter recruitment success. It is nowadays widely acknowledged that mortality in ELHS is not caused by a single process but rather by an interaction of several processes across different developmental, spatial, and temporal scales (Fritz et al. 1990; Letcher et al. 1996).

Despite this long history of recruitment research and the existence of well-established theories, life-history research as well as fisheries science continuously aim at elucidating how the interaction of environmental (extrinsic) and organismal (intrinsic) factors shape condition and survival rates of fish ELHS across spatio-temporal gradients to holistically evaluate year-class fluctuations and stock size of individual fisheries resources (Houde 1987, 1997, 2001; Laurence and Howell 1981; Lehodey et al. 2006; Miller and Kendall 2009).

1.2 Ingress of ichthyofaunal early life-history stages to coastal environments

Many fish species from temperate zones occupy different marine habitats throughout their ontogeny (Sale 1980; Macpherson 1998; Fuiman 2002; Werner 2002). For coastal fish, several habitats are considered essential for ELHS (Fig. 3, after Boehlert and Mundy 1988). Spawning takes place **offshore**, thereby exposing planktic eggs and early larval stages to the pelagic environment and its prevalent physical processes (i.e., Ekman transport, [coastal] countercurrents, eddies) (Parrish et al. 1981; Miller et al. 1984). The timing and location of the adult reproductive activity have a significant impact on the effective transport of ELHS from offshore spawning grounds to inshore nursery areas (Whitehead et al. 1989; Begg and Marteinsdottir 2002; Vasconcelos et al. 2010). The hydrodynamics of the pelagic system, in combination with the vertical migration behaviour of the early larvae, the latter being influenced by abiotic and biotic factors (e.g., temperature, light, prey availability), govern the drift of the larval stages between off- and onshore regions as well as the nearshore accumulation along the coastline (Norcross and Shaw 1984; Kingsford and Choat 1986; Boehlert and Mundy 1988; Tiedemann and Brehmer 2017). The **nearshore zone** represents a transitory habitat for estuarine-dependent taxa: the photoperiod ("diel zeitgeber) stimulates circadian activity patterns (e.g., feeding periodicity, diel vertical migration) in late-stage, exogenously-feeding larvae (Werner 2002; Teodósio et al. 2016). The vertical migration is anticipated to allow for substantial horizontal movements via favourable currents towards the next transitory habitat, the **channel** or **mouth** of an estuary (Boehlert and Mundy 1988; Lough and Bolz 1989; Werner 2002).

Selective tidal stream transport (i.e., the larvae's circatidal behavioural response to synergistic effects of different environmental cues varying with the tidal flux) has been proposed to be the predominant mechanism for ingress and retention within estuaries for a variety of fish taxa (Boehlert and Mundy 1988; Miller 1988; Forward et al. 1998; Jager 1999; Silva et al. 2017). However, additional hypotheses, including wind forcing, residual bottom inflow and selective tidal stream transport (Weinstein et al. 1980; Joyeux 1999; Schultz et al. 2003; Hare et al. 2005; Schieler et al. 2014), as well as sensorial acuity and cue-oriented swimming of larval fish have also been proposed to shape successful ingress into estuaries (Teodósio et al. 2016; Baptista et al. 2019, 2020). Following the ingress into an **estuary**, the ELHS settle in suitable nursery (micro-)habitats, with species- and life-stage-specific preferences for certain abiotic and biotic conditions (e.g., substrate, vegetation, depth, turbulence) (Boehlert and Mundy 1988; Levin 1994; Ramos et al. 2006; Bastrikin et al. 2014; Teodósio et al. 2016). Both the effective ingress as well as the retention within an estuarine nursery ground (i.e., settlement) are essential components affecting the recruitment success and thus the connectivity to adult coastal stocks (Koutsikopoulos and Lacroix 1992; Able 2005; Jordan and Peterson 2012; Martinho et al. 2012).

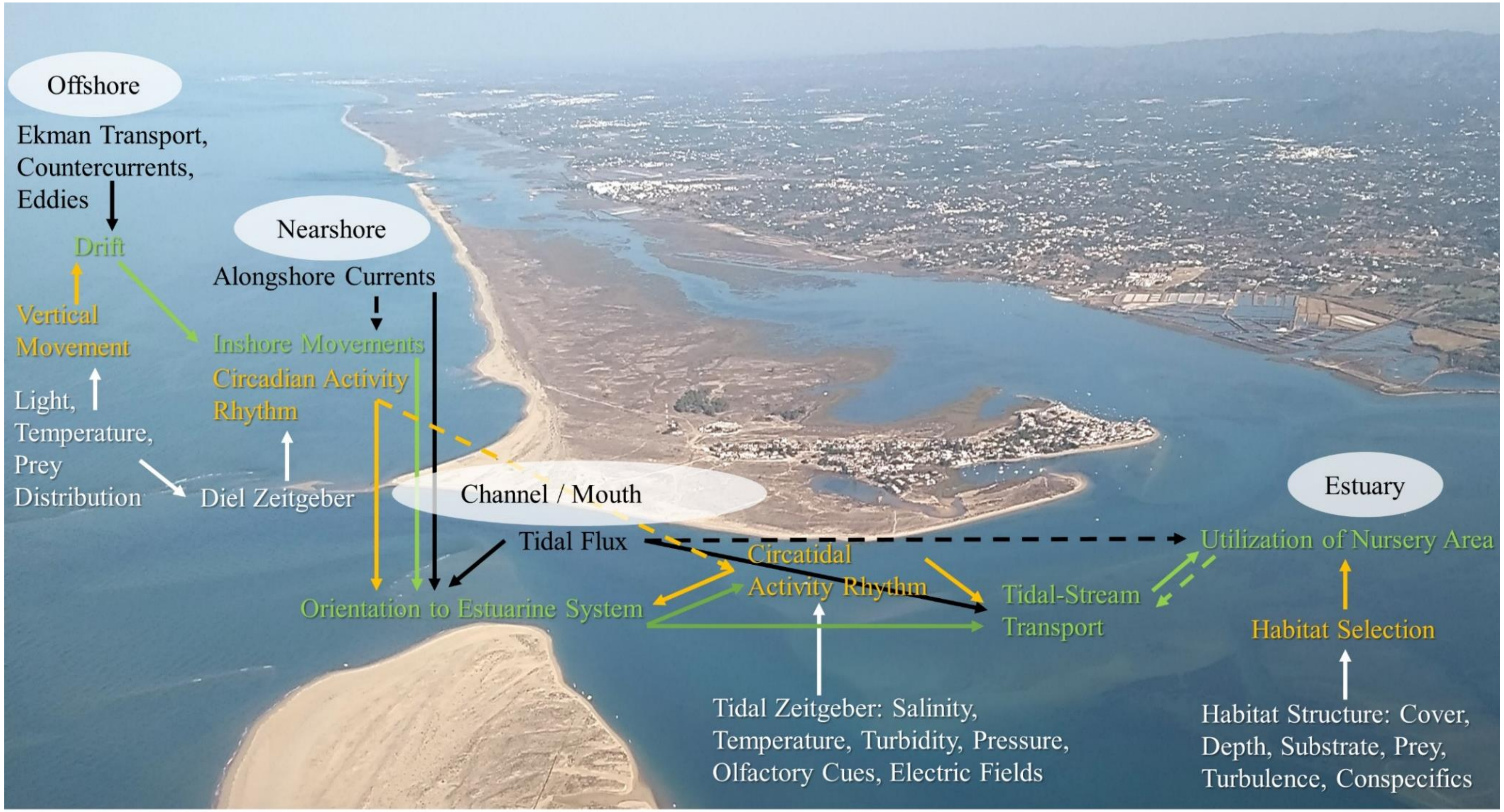

Fig. 3 Schematic diagram of the movement of ichthyofaunal early life-history stages across different habitats, from offshore spawning grounds, via nearshore environments, through channels to estuarine nursery grounds (after Boehlert and Mundy 1988). The physical processes as well as the abiotic and biotic stimuli acting on fish early life-history stages are represented in black and white respectively. The corresponding activity of the fish along with its behaviour are highlighted in yellow and green respectively.

1.3 Importance of transitional zones as nurseries

At the interface of terrestrial and marine realms, transitional environments, such as estuaries and lagoons, sustain highly productive aquatic ecosystems that provide valuable ecological, economic, and social/cultural goods and services (Nixon et al. 1986; Costanza et al. 1997; Worm et al. 2006; Beaumont et al. 2007; Atkins et al. 2011; Liquete et al. 2013). Lagoons and estuaries are composed of a diverse mosaic of (micro-) habitats which vary in their suitability and utilisation as habitats for various organisms and life-history stages (Addicott et al. 1987; Levin 1994; Isnard et al. 2015). Notably, specific estuarine and coastal habitat patches comprising a high structural complexity, such as mangrove forests, marshes, and seagrass meadows, have been acknowledged to play an essential role in the life cycle of fish, among them many taxa of commercial importance, which occupy these habitats as feeding, spawning or nursery grounds either permanently, transitionally (e.g., during specific life-history stages), or occasionally (Boesch and Turner 1984; Orth et al. 1984; Blaber 1997; Beck et al. 2001; Franco et al. 2008; Vasconcelos et al. 2010). On account of favourable physical conditions and advantageous hydrodynamics, as well as of the availability of biotopes, refuge, and trophic resources, these vegetated (micro-)habitats ensure a higher production of fish recruits per unit area when compared to other habitats frequented by juveniles of a given species and are therefore often referred to as nurseries or nursery grounds (Heck et al. 1997; Beck et al. 2001; Levin and Stunz 2005; Dahlgren et al. 2006; Nagelkerken 2009; Sheaves et al. 2015).

Despite offering particularly favourable conditions for ELHS of fish, nearshore habitats also feature a high environmental and spatial variability, and are frequently dominated by well-adapted, highly abundant species which show intra- and interspecific resource partitioning to counterbalance spatial and diet niche overlapping (Vinagre et al. 2005; Elliott and Quintino 2007; Pasquaud et al. 2010). While density-independent processes (e.g., abiotic environmental conditions) mainly shape recruitment variability at the margin of the distributional range, density-dependent processes (e.g., competition for food and refuge) prevail at distributional hotspots such as suitable (micro-)habitats, and recruitment success has been shown to be positively related to nursery surface area (Miller et al. 1991; Rijnsdorp et al. 1992). Thus, both habitat quality and quantity directly affect the nursery function of estuarine and coastal environments, which in return influence the condition and growth of ELHS of fish and consequently the recruitment success to adult populations offshore (van der Veer et al. 1990; Beverton and Iles 1992; Ciotti et al. 2014; Fonseca et al. 2015).

The significant contribution of estuarine nurseries towards the replenishment of nearshore fish populations has been widely acknowledged and can be regarded an ecosystem service in itself (Beck et al. 2001; Able 2005; Dahlgren et al. 2006; Liquete et al. 2016). Estuarine-dependent fish species constitute a major proportion of commercial catches in coastal areas (Houde and Rutherford 1993; Jordan and Peterson 2012; Martinho et al. 2012): members of the families Sciaenidae (e.g., Meagre), Moronidae (e.g., European bass), Clupeidae (e.g., sardines), Engraulidae (e.g., anchovies), or Sparidae (e.g., White seabream), for instance, are prime target species for subsistence, artisanal and commercial fisheries alike (Crawford et al. 1987; Rocklin et al. 2014; Soares et al. 2015; Chao et al. 2015; Leitão et al. 2016). As population dynamics and fish production of coastal species are reliant on the successful dispersal and migration of different life-history stages between various habitats, knowledge on the processes and conditions shaping this connectivity are of paramount importance for defining effective fisheries management and conservation measures (Gillanders et al. 2003; Anticamara et al. 2011; Martinho et al. 2012; Sheaves et al. 2015).

While coastal lagoons and estuaries offer a variety of favourable conditions for ELHS, the diverse mosaic of (micro-)habitats in combination with complex gradients of environmental conditions and anthropogenic interferences (i.e., plastic pollution) lead to a disparate spatio-temporal suitability of such areas as nurseries, with implications potentially arising particularly for ichthyofaunal ELHS critically dependent on certain habitats at a specific point in time (e.g., species / life-history stages showing a high site fidelity). Furthermore, temporal variation in the relative contribution of a particular nursery ground to coastal populations has been observed as well as the consistently higher connectivity of certain estuarine nurseries over others with minor output of recruits (Reis-Santos et al. 2013). Moreover, a particular area or habitat type (e.g., seagrass meadows) does not promote all fish taxa and life-history stages equally, i.e., certain micro-habitats within an estuary may be more favourable for the growth and condition of juveniles of a certain species while others support the juveniles of multiple species simultaneously, thereby contributing to interspecific variability in connectivity across spatial scales (Minello et al. 2003; Amorim et al. 2018; Schloesser and Fabrizio 2019; Guerreiro et al. 2021). Considering the relevance for future recruitment and population development, understanding and preserving the functioning of nursery (micro-)habitats is vital to sustain many and especially artisanal fisheries, yet particularly complex due to high spatial and temporal variability of transitional ecosystems in which nurseries are frequently situated, as well as due to the various anthropogenic interests, usages and pressures exerted on coastal and marine environments further diminishing habitat quality and quantity (McLusky and Elliott 2004; Orth et al. 2006; Elliott and Quintino 2007).

1.4 Plastic litter and its distribution in the marine environment

Among the multitude of co-occurring, anthropogenically driven impacts (e.g., habitat degradation, over-exploitation of resources, climate change) compromising the wealth of ecosystem services which marine and coastal environments provide, plastic pollution has become the focus of scientific and social attention: owing to its increasing production and durability, with plastic items nowadays representing the most abundant type of marine debris worldwide (GESAMP 2015, 2016; Cunningham and Sigwart 2019; Völker et al. 2019).

The mass-production and application of plastic commenced in the 1950s and has experienced undamped growth to this day, leading to an estimated annual production of more than 360 million tons (Lebreton and Andrady 2019; Plastics Europe 2020). Increasing plastic quantities in the marine realm have coincided with cumulative manufacturing and industrial application of synthetic polymers, lack of waste management regimes, along with increasing urbanisation (including land- and sea-based activities). However, the plastic fraction entering the oceans can only be guessed (Thompson 2004; Borrelle et al. 2017; Schmidt et al. 2017; Lebreton and Andrady 2019; Harris et al. 2021), with projections on the quantity of plastics floating at the surface of the world's ocean ranging from 7000 tons (Cózar et al. 2014) to 268,940 tons (Eriksen et al. 2014), whereas extrapolations on the amount of mismanaged plastic waste entering the global ocean in the year of 2010 alone revealed a range of 3.1 to 12.7 million tons (Jambeck et al. 2015; Lebreton and Andrady 2019).

Despite the uncertainty regarding estimates of the abundance of plastic in the oceans, it is nowadays undisputed that approximately 80% of marine plastic debris originates from land-based sources and thus spreads from terrestrial through coastal ecosystems to the global oceans (Browne et al. 2011; Jennerjahn and Mitchell 2013; Morritt et al. 2014; Auta et al. 2017; Hitchcock and Mitrovic 2019) In fact, coastal areas have been recognised as plastic hotspots and a significant relationship between plastic abundance and human population density has been established, mainly driven by the combination of contaminated river discharges, coastal urbanisation (e.g., ports, tourism, shipping, fisheries and aquaculture) and the ability of structurally complex habitats, such as seagrass meadows, to retain microplastic (MP) particles in the ecosystem (Cole et al. 2011; Frias et al. 2014; Lechner et al. 2014; Auta et al. 2017; Hitchcock and Mitrovic 2019; De los Santos 2021). Similar to the natural spatial and environmental variability of vital nursery habitats, the extent of plastic input may vary across small spatio-temporal scales with potential consequences for recruitment of fish populations in adjacent waters.

1.5 The ecological dimension of plastic pollution - microplastic uptake by fish

Since the earliest reports on encounters of marine organisms with anthropogenic waste items were published in the 1930's (Gudger and Hoffman 1931; Gudger 1938), interaction with marine litter has now been reported for more than 1,400 different species (Claro et al. 2019). Notably, the potential detrimental effects of plastic debris for biota are of growing concern due to the durability of this pollutant along with its global distribution and increasing abundances in coastal and marine environments as described above. The entanglement in and ingestion of macroplastic items by marine wildlife, especially by marine mammals (e.g., Fowler 1982; Kraus 1985; Alexiadou et al. 2019), elasmobranchs (e.g., Sazima et al. 2002; Parton et al. 2019), sea turtles (e.g., Nelms et al. 2016; Wilcox et al. 2018) and seabirds (e.g., Baltz and Morejohn 1976; Roman et al. 2016), is well documented and associated mortalities are typically regarded as an emotive issue triggering scientific research initiatives, environmental campaigns, and extensive media coverage (Laist 1997; Wright et al. 2013; Provencher et al. 2017; Claro et al. 2019; Völker et al. 2019). Although the effect of marine litter on charismatic marine megafauna attracts most of the media attention (Mazzoldi et al. 2019; Kühn and van Franeker 2020), fish are in fact among the most thoroughly studied group of organisms in relation to plastic uptake (de Sá et al. 2018; Azevedo-Santos et al. 2019).

Plastic ingestion by fish has been described from freshwater to coastal and marine environments worldwide, and particles of all size classes (macro- to nano-sized plastics) have been detected in the gastrointestinal tract (GIT) of both cartilaginous (Chondrichthyes) and bony (Osteichthyes) fish species (e.g., Boerger et al. 2010; Liboiron et al. 2016; Güven et al. 2017; Parton et al. 2020; Parker et al. 2021). In relation to the ingestion of plastic debris and associated adverse impacts, micro-sized particles have more recently come to the forefront of scientific and public attention (de Sá et al. 2018).

Despite the increasing number of scientific publications in recent years, the first estimates of the abundance of microplastic (MP) particles in surface waters of the Sargasso Sea and in New England coastal waters were published almost five decades ago, and already at that time researchers highlighted the potential implications of MP ingestion, particularly for smaller fish (Carpenter and Smith 1972; Carpenter et al. 1972; Colton et al. 1974; Kartar et al. 1976). Due to their size range, MP particles allow for interaction with and bioaccumulation in a wide variety of (ichthyo-)planktic organisms, and biomagnification along the food chain to fish taxa destined for human utilisation (i.e., in fisheries, aquaculture, ornamental trade or as bait or game fish) may be anticipated (Cole et al. 2013; Chagnon et al. 2018; Nelms et al. 2018; Provencher et al. 2019). However, a review of the literature shows terminological inconsistencies especially in the definition of the lower plastic size classes (Gregory and Andrady 2003), leading to a rather broad use of the term microplastics for particles ranging from a few µm up to < 10 mm (Arthur et al. 2009; Graham and Thompson 2009; Andrady 2011; Hanke et al. 2013). However, the most frequently applied definition of MP is particles of a size range less than 5 mm, as proposed by Arthur et al. (2009).

Albeit the long history of research on this topic, less than 500 different fish species have been investigated for MP ingestion with highly diverging outcomes concerning the proportion of MP-feeding individuals as well as the MP load per individual (Markic et al. 2020). In addition to the numeric differences across studies and taxa, significant uncertainties about the key drivers for MP uptake remain (Provencher et al. 2019). A multitude of different factors has been hypothesized to determine the susceptibility of fish taxa and life-history stages for MP uptake, such as trophic guild (Anastasopoulou et al. 2013; Vendel et al. 2017; Sun et al. 2019), preferred habitat (Güven et al. 2017; Markic et al. 2018), environmental parameters (Ferreira et al. 2016b; Welden et al. 2018), morphometrics, condition, gender or life-history stage (Boerger et al. 2010; Critchell and Hoogenboom 2018; Horton et al. 2018; Ory et al. 2018b), yet the majority of studies neither considered any of the above in their field work design or laboratory set-up nor verified the relevance of any parameter for MP ingestion probability beyond doubt. From both an ecological and economic point of view, however, addressing knowledge gaps related to the detrimental impacts of plastic ingestion on fish growth, condition and survival seems to be of even greater importance to infer the hazardous potential of this anthropogenic pollutant for one of the most essential marine living resources. Even though the outcomes of both field and laboratory studies indicate that MP exposure has the potential to detrimentally affect fish condition and survival, the environmental relevance especially of experimental set-ups has been questioned lately, further reinforcing the need for integrated, comprehensive study designs to address persistent knowledge gaps in this field (Phuong et al. 2016; Paul-Pont et al. 2018).

1.6 Implications of plastic pollution for fish stocks and fisheries

Although the gross of plastic waste input into the world's oceans originates from land-based sources (Lechner et al. 2014; Rochman 2018), the fisheries and aquaculture sectors need to be understood both as a considerable source of and sufferer from marine plastic (Lusher et al. 2017a; Boucher and Billard 2019). Evidently, fisheries catching, processing, and distributing sub-sectors have benefited from the industrial production of synthetic materials, including plastic-based paints and anti-fouling agents deployed in ship construction and maintenance, cheap and durable plastic fibres used for fishing gear, and the industrial plastic packaging for transportation and distribution of fish and fish products (Lusher et al. 2017a). As much as the manufacturing of plastic advanced the entire sector and continues to do so, it has also been contributing ever since to the issue of marine litter, with an estimated 18% of marine plastic debris being attributed to the fisheries sector (Andrady 2011). Lost or abandoned gears and materials from fisheries and aquaculture have been documented globally on coastlines and beaches (Merrell 1980; Slip and Burton 1991; Browne 2015; Unger and Harrison 2016), as well as from surface waters (Garcia-Garin et al. 2020; Ruiz et al. 2020) to the ocean floor / deep sea (Hess et al. 1999; Woddall et al. 2015). Despite the growing amount of evidence for marine plastic litter originating from fisheries and aquaculture, reliable accounts to evaluate the micro-sized waste input generated by these industries across spatio-temporal scales are lacking (Lusher et al. 2017a).

Fisheries and aquaculture are frequently identified as a chronic source of marine litter, yet these sectors are also subjected to the detrimental impacts of micro- and macro-sized plastic pollution which may reduce the efficiency and productivity of fisheries (and aquaculture) in both direct and indirect ways (Fig. 4).

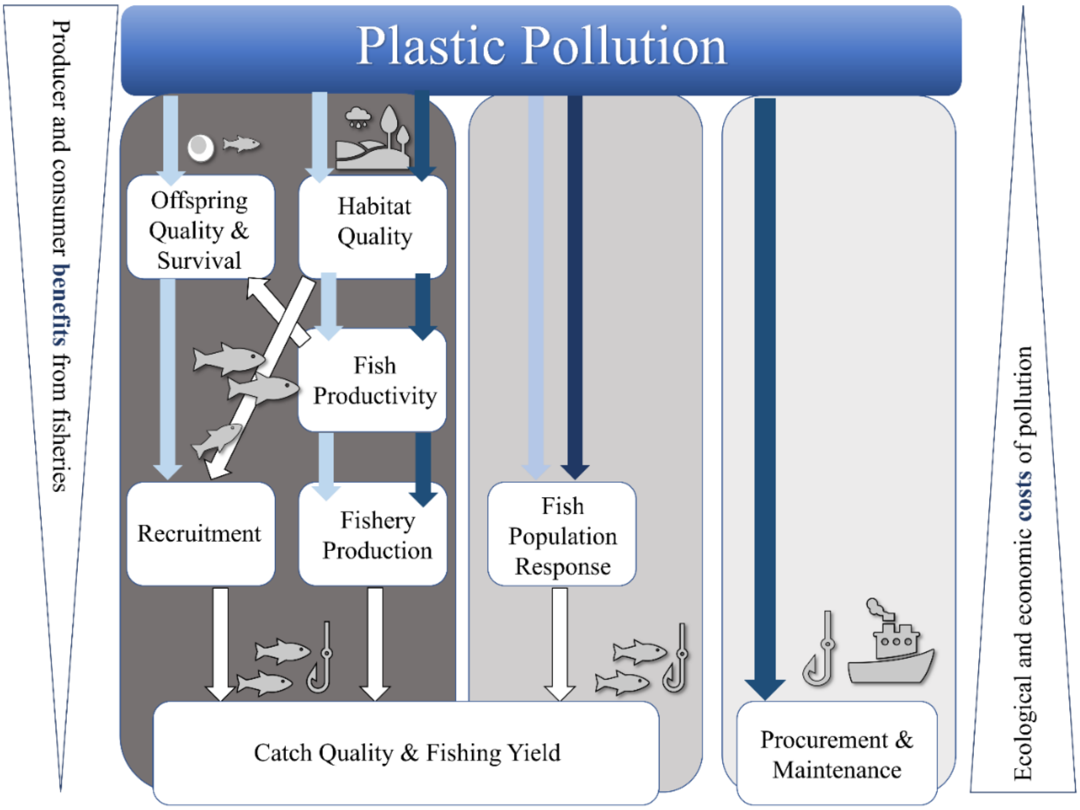

Fig. 4 Schematic diagram showing the effects of plastic pollution on the fisheries sector. With persistent and cumulative plastic pollution, the benefits for producers and costumers decrease, whereas the ecological and economic costs increase (indicated by the enclosing triangles). The effects of micro- and macroplastic pollution are indicated by light and dark blue arrows respectively; white arrows highlight internal links. The direct impact of plastic pollution on the sector is highlighted in light grey; the indirect impact on the sector via affected catch quality and fishing yield is highlighted in darker shades of grey. Modified after Islam and Tanaka (2004).

1.6.1 Impact of plastic pollution on the quality of nursery habitats and fish offspring

Plastic pollution has been shown to adversely affect estuarine and marine habitat quality (Fig 4, dark grey column) in different ways: macro-sized litter, such as derelict or lost fishing gear in vital nursery and fishing grounds, poses the direct risk of entanglement to marine biota (Laist 1997; Kühn et al. 2015). Furthermore, macro-plastic items may cause structural damage, reduced light and oxygen availability in seagrass meadows, mangroves and coral reefs known as essential nursery grounds (Uhrin and Schellinger 2011; Chapron et al. 2018; Luo et al. 2021; Menicagly et al. 2021). The deteriorating habitat quality is consequently expected to adversely impact fish productivity (Islam and Tanaka 2004), with direct and indirect consequences for ELHS, recruitment success and fishery production (Fig. 4, dark grey column) (Chambers 1992; Beck et al. 2001; Hughes et al. 2002; Brehmer et al. 2013).

Coastal vegetated habitats have been shown to act as a sink for micro-sized plastics thereby potentially enhancing the intentional, accidental, or secondary ingestion of MP items by marine biota (Cozzolino et al. 2020; Jones et al. 2020; De los Santos et al. 2021). Larval and juvenile fish may consequently be prone to the uptake of smaller-sized plastic fragments and fibres due to a combination of habitat preferences, feeding mode and disadvantageous size ratio (Lusher 2015; Critchell and Hoogenboom 2018; Gove et al. 2019). Moreover, the potential detrimental effects of exposure and ingestion could impair recruitment success, and eventually fishing yield (Fig. 4, dark grey column).

1.6.2 Impacts of plastic pollution on fish stocks and the fisheries sector

One of the essential concepts for the sustainable use of living marine resources is the ecosystem-based approach to (fisheries) management, that acknowledges both stakeholder perspectives and biotic, abiotic, as well as anthropogenic ecosystem components along with their interaction with each other rather than focusing on the management of a single species in isolation (Garcia et al. 2003). In this respect, the response of natural fish stocks to environmental and anthropogenic changes is mainly accounted for at the population level (Fig. 4, medium grey column), whereas monitoring the extent of response variation across individuals of the same population may be worth considering both for management and conservation purposes (Cooke and O’Connor 2010; Ward et al. 2016).

To implement sustainable exploitation of coastal and marine fish stocks, reliable figures on all sources of fishing mortality are needed (FAO 2011), including those associated to large plastic litter items such as abandoned, lost or otherwise discarded fishing gear (ALDFG), yet fisheries management rarely takes this source into consideration due to a lack of both adequate data and accurate estimation methods, even though this mortality factor may be significant for certain species and fisheries (Gilman et al. 2013). Recently, Lively and Good (2019) reviewed studies assessing the impact of ‘ghost fishing’ by experimentally sunk or derelict gear across the world. It was shown that the ecological and economic impacts of ALDFG depend on the type of gear, with substantial variation across spatio-temporal scales, while catch rates being highest immediately upon loss and decreasing over time and in relation to local conditions. To thoroughly estimate the contribution of ghost fishing to fishing mortality, key parameters such as average species-specific mortality rates per gear and unit time, the local amount of ALDFG as well as the total fishing area need to be taken into account (Matsuoka et al. 2005).

The fisheries sector experiences a direct decrease in profits due to the loss of gear itself, or due to the necessity to clean, repair or replace damaged equipment as well as by the disruptive impact of ghost fishing nets and other large marine plastic debris on gear and vessels (Fig. A, light grey column “Procurement & Maintenance”) (MacFayden et al. 2009; Mouat et al. 2010; Sheavly and Register 2007). Reliable local and global accounts on the implications of macro-sized plastic litter for the fisheries sector are scarce and difficult to obtain, in part because specific fishing methods / gear may vary in their vulnerability to economic losses (MacFadyen et al. 2009; Richardson et al. 2018; Gilman et al. 2021), and due to the challenges of calculating indirect costs stemming from reduced catch quality and fishing yield (Fig. 4, medium grey column) of commercially relevant fish stocks.

In relation to the potentially harmful effects of smaller sized plastic items, MP ingestion has been demonstrated in various commercially important fish taxa across different trophic levels and various habitats e.g., the Atlantic herring *Clupea harengus* (Collard et al. 2017b), the European anchovy *Engraulis encrasicolus* (Collard et al. 2017b), the Atlantic and Pacific cod *Gadus morhua* and *Gadus macrocephalus* (Sun et al. 2019; Liboiron et al. 2016) as well as the common two-banded seabream *Diplodus vulgaris* (Bessa et al. 2018b). Summarising the outcomes of the currently available studies, around 1/3 of all individuals assessed had micro-sized plastic items in the GIT, with an average MP-load of two pieces per plastic-feeding individual (Markic et al. 2020). Despite the aforementioned findings and the increasing scientific effort focussing on plastic uptake by marine fish taxa, the ecological and economic implications of micro-sized plastic litter for fish populations (Fig. 4, medium grey column) remain to be understood (De-la-Torre 2020). The variability of MP exposure and uptake across biological, temporal, and spatial scales as well as the still insufficient data situation (i.e., scientific information exists only for approximately 8% of the species actually targeted by the fisheries sector) impede a comprehensive evaluation of the actual threat of plastic debris to aquatic fish production and food security (Islam and Tanaka 2004; Galloway 2015; Markic et al. 2020; De-la-Torre 2020).

Despite the persistent knowledge gaps outlined above, plastic exposure and ingestion may lead to reduced fishing yield and loss of earnings related to fish as a marine living resource. Together with other (interrelated) pressures on the fisheries and aquaculture sectors (e.g., climate change, habitat degradation, overexploitation), plastic marine litter therefore encompasses the potential to change the availability and quality of certain fish stocks as well as other marine living resources (Lusher 2017a; Barange et al. 2014, 2018; Cunnigham et al.

2020). Consequently, more species- and life-history-stage specific surveys across small spatio-temporal scales may be beneficial to understand the potential of micro- and macroplastic litter to detrimentally affect population development of local fish stocks and thus to compromise the sustainable utilisation of fishery resources.

2 Scope of the Thesis

As ichthyofaunal communities have been successfully used as biological indicators to investigate the health of an ecosystem and monitor anthropogenically-induced changes to environmental quality (Whitfield 1996; Whitfield and Elliott 2002; Lloret and Planes 2003; Roset et al. 2007; Ribeiro et al. 2008; Breine et al. 2010; Whitfield and Harrison 2014), assessing the potential effects of plastic pollution on ELHS of fish in estuaries and coastal lagoons is of mutual benefit from the perspective of life-history research, fisheries biology, and conservation ecology alike.

Despite this acknowledged suitability as biological indicators and the growing body of scientific evidence on the ubiquity of marine plastic litter, detailed research on individual fish taxa in transitional ecosystems remains limited, even though fish are among the most intensively studied group of organisms in relation to plastic pollution over the past decades. MP uptake by fish and potential detrimental effects of ingestion have been commonly investigated via *in-situ* assessments or laboratory feeding trials. However, both approaches are limited and partially inconsistent in their explanatory power due to shortcomings in relation to different methodological approaches and experimental designs, inadequate sample sizes, underrepresentation of different life-history stages and taxa of commercial importance, as well as to the lack of contextualisation and condition assessments.

The overall goal of the present thesis is therefore to deepen the knowledge of the effects of MP on growth, development, and survival of ELHS of fish depending on nursery habitats in transitional ecosystems. By selecting an emblematic study site (the Ria Formosa lagoon, Southern Portugal) along with a generic fish taxon (family Sparidae), it is envisaged to provide a holistic evaluation of the potential extent and consequences of marine plastic pollution for recruitment and population dynamics of a commercially important coastal fish stock in relation to different habitat quality parameters. The thesis is based on a comprehensive literature review facilitating the adaptation of an integrated study design which involves an *in-situ* assessment in combination with a laboratory feeding experiment. To the best knowledge available, a synthesis of scientifically sound laboratory trials and contextualised field investigations of the same fish species has not yet been published, although the combination of both approaches is expected to improve the understanding of the implications of MP exposure and ingestion.

To achieve this objective, the following main research questions are addressed in three separate chapters:

1) **What are the driving factors for and the impacts of microplastic ingestion by fish?**

1st Chapter:

The first chapter of the thesis quantitatively and qualitatively reviews the outcomes of 90 *in-situ* MP ingestions studies, published between 1972 and 2019. It provides a synthesis of records for more than 480 different fish taxa that have been examined within the past decades and assesses the state-of-the-art regarding explanatory variables of MP uptake. This chapter reveals persistent knowledge gaps in relation to the inclusion of ELHS in field investigations, to the examination of *in-situ* ingestion effects, as well as to the contextualization of MP ingestion. It therefore recommends specific measures to promote a holistic consideration of the outcomes of future field studies. Furthermore, the existence of publication biases in the rapidly growing research area of MP ingestion by fish is analysed; results from the literature review and the bias analysis enhance a sound evaluation of the dimensions of MP uptake as well as consequent effects and form the basis for the empirical and experimental design of the two following chapters of this thesis.

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2) **What is the level of MP exposure and ingestion by omnivorous juvenile fish in a vital coastal lagoon nursery in relation to prey availability and environmental conditions?**

2nd Chapter:

The second chapter of the thesis provides an integrated, species-specific *in-situ* assessment of MP uptake and the potential detrimental effects of exposure and ingestion on the ELHS of a commercially important, omnivorous fish species in a transitional environment. According to the underlying hypothesis that the majority of MP enters the marine realm through coastal ecosystems, and vegetated nursery micro-habitats (i.e., seagrass meadows) act as a sink for MP, the potential vulnerability of juvenile white seabream *Diplodus sargus* was evaluated across a spatial gradient of anthropogenic interference. The analysis of the gastrointestinal tract of the juvenile fish was complemented with the assessment of zooplanktonic and benthic prey availability along with MP abundances in the respective habitats. Results obtained by this field study challenge the assumption of feeding mode and habitat use as driving factors for elevated MP uptake rates. Ingestion of synthetic particles and fibres (> 150 µm) was less prevalent than hypothesized for ELHS in a coastal lagoon and MP uptake was rather associated with inter-individual variability in trophic resource utilization than with the omnivorous feeding mode, morphometrics or micro-habitat use. MP exposure and ingestion of minor MP loads were not found to impair juvenile fish condition or uptake of natural prey items.

This chapter has been submitted to *Environmental Biology of Fishes* as:

Müller C, Erzini K, Dudeck T, Cruz J, Santos Corona L, Abrunhosa FE, Afonso CML, Mateus MAF, Orro C and Ekau W (subm). Growing up in a plastic ocean: variability of prey preferences, microplastic exposure and uptake by juvenile white seabream in a coastal lagoon nursery ground.

3) What are the physiological impacts of MP exposure and uptake for juvenile omnivorous fish?

3rd Chapter:

The main objective of the third chapter is to advance our understanding of potential physiological implications of MP ingestion by juvenile omnivorous fish, using a laboratory feeding experiment. In a novel set-up, applying polystyrene MP fragments (size range 500 – 1000 µm), the effects of MP exposure and uptake on survival, growth, and condition of juvenile white seabream *Diplodus sargus* (Linnaeus, 1758) were studied over a course of 3.5 weeks. Following the underlying premise of this thesis, the laboratory study took into consideration the application of either pristine or biofilm-coated, blue MP fragments, deployed in densities mimicking natural conditions while simultaneously feeding live prey items. This chapter shows that detrimental impacts of MP on growth and condition occurred only when particles are taken up in exceptionally high quantities (> 20 MP per fish). The uptake of only a few particles did not impair juvenile fish survival, growth, and condition over the course of the trial. Most fish presumably either did not ingest the MP particles offered at all – or efficiently egested the particles before the end of the experiment. Seabream were able to discriminate between edible and non-edible prey items; a preference for biofilm-coated over pristine MP was not verified.

To reliably assess the physiological effects of MP exposure and ingestion by ELHS of fish, the outcomes of this study reinforce laboratory studies as a suitable measure, however this chapter emphasizes the necessity to conduct MP feeding experiments mimicking natural conditions.

This chapter has been published as:

Müller C, Erzini K, Teodósio MA, Pousão-Ferreira P, Baptista V and Ekau W (2020) Assessing microplastic uptake and impact on omnivorous juvenile white seabream Diplodus sargus *(Linnaeus, 1758) under laboratory conditions. Marine Pollution Bulletin. DOI:10.1016/j.marpolbul.2020.111162*

The contribution of the PhD Candidate to each chapter of the present thesis is given in Table 1. Additionally, Chapter 2 and Chapter 3 contain an authorship contribution statement as requested by the respective journal.

Table 1 Contribution (in %) of PhD Candidate to conceptualisation, data acquisition and analysis, interpretation of results and preparation of manuscript, including visualisation of results (in figures and tables) for each of the three chapters presented in this thesis.

Task	**Chapter 1**	**Chapter 2**	**Chapter 3**
Concept and Design	100	90	85
Acquisition of Data	100	85	90
Data Analysis and Interpretation	100	95	90
Preparation of Figures and Tables	100	90	90
Drafting of Manuscript (incl. first draft and revision)	100	95	95

In a final discussion, the key results and findings of the three studies are integrated and evaluated in the context of the current state of knowledge concerning MP and ELHS. The applied methodologies along with the significance of the results are assessed in a broader, holistic context in order to identify future scientific objectives and make recommendations.

3 Study Area and Model Organism

3.1 Study area: Ria Formosa lagoon

The Ria Formosa lagoon is a triangularly-shaped, multi-inlet barrier island system, extending approximately 55 km in length and 6 km in width along the Southern Portuguese coast (36°58’N, 8°02’W to 37°03’N, 7°32’W, Fig. 5). Due to its high ecological and economic importance, the Ria Formosa lagoon has been nationally designated a Natural Park in 1987, is part of the international Natura 2000 European network for nature conservation as well as a classified Ramsar wetland and Special Bird Protection Area (Birds Directive 79/409/EEC; Habitats Directive 92/43/EEC; Decree-law 45/78, 1978; Decree-law 373/87, 1987).

Geography and hydrography

The mesotidal lagoon is characterised by a semi-diurnal tidal regime, with tidal amplitudes ranging between 3.5 m during spring tides and 0.5 m at neap tides, exposing an intertidal area of approximately 1/3 of the total lagoon area (Falcão and Vale 1990; Ribeiro et al. 2006). A seaward belt of sand-barrier islands protects the lagoon’s sand and mud flats, its wetlands and salt marshes as well as the salt and aquaculture ponds; six connecting inlets facilitate a daily tidal water exchange of 50 – 75 % between the complex system of highly branched subtidal creeks and channels and the Atlantic Ocean (Águas 1986). The water residence time ranges between 0.5 to > 7 days (mean 2.4 days) depending on the location (Saraiva et al. 2007; Mudge et al. 2008). While most of the area is less than 2 m deep at low tide, an average depth of approximately 6 m is recorded in the main channels of the lagoon (Newton and Mudge 2003). Though there is only one major river near Tavira in the eastern part of the lagoon, and river discharges are generally considered to be relatively small (many of the smaller rivers and streams dry out completely during summer), the system experiences direct inputs of urban wastewater and considerable occasional (i.e., during heavy rainfall episodes) or seasonal (i.e., during winter) land runoff (Bebianno 1995; Newton and Mudge 2003; Tett et al. 2003).

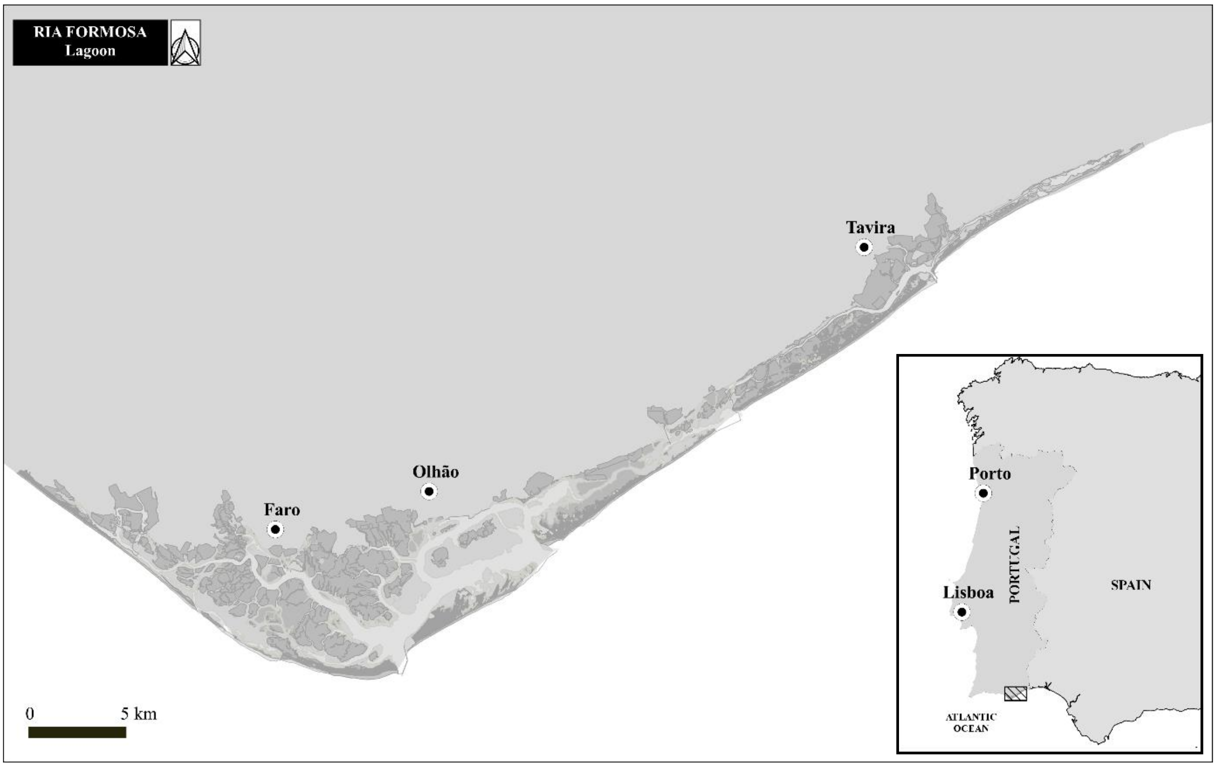

Fig. 5 Map of mainland Portugal with the location of the Algarve coast; enlarged view of the Ria Formosa lagoon.

Ecology and ichthyofauna

Located at the interface of land and sea, this transitional ecosystem is characterised by a unique set of hydrodynamic, biological, and ecological features shaping the diverse floral and faunal communities of the lagoon (Sprung 1994; Erzini et al. 2002; Gamito 2008). The Ria Formosa is constituted by a wide range of different habitats, which vary in depth, substrate type, vegetation and anthropogenic interference: the extensive intertidal area is composed of saltmarshes, sandy sediments and mudflats as well as macrophyte beds hosting three out of four European seagrass species, namely *Zostera marina* Linnaeus 1753, *Zostera noltei* Hornemann 1832 and *Cymodocea nodosa* (Ucria) Ascherson 1870 (Borum and Greve 2004). Seagrass meadows in the Ria Formosa have been identified as essential habitats, supporting a higher biodiversity and densities of both invertebrate and vertebrate taxa than adjacent bare-sediment sites (Gamito 2008; Ribeiro et al. 2012).

The Ria Formosa accommodates a diverse ichthyofauna which is known to comprise at least 112 different species which can be classified as either resident, migratory or occasional (Erzini et al. 2002). The fish assemblage structure in the lagoon shows a spatio-temporal pattern which is predominantly shaped by cyclic settlement (immigration) and recruitment (emigration) of coastal species that use the lagoon as a nursery. Highest diversity and abundance are recorded in summer and autumn which is mainly due to the presence of non-resident species spending their ELHS in the lagoon during the warmer seasons and migrating to coastal waters in late autumn or winter (Monteiro et al. 1987). Furthermore, peaks in abundance and diversity have been generally associated with shallow, less turbulent waters (with depths < 3 m during low tide) and a cover of marine plants over fine, muddy sediment bottoms towards the interior of the lagoon (Monteiro et al. 1990; Erzini et al. 2002). As significant populations of juvenile fish (including commercially relevant taxa) have been recorded in the vegetated habitats of the lagoon, the Ria Formosa is recognised as an essential nursery for the adjacent coastal fish populations and thus a suitable site for the purposes of this study (Monteiro et al. 1987, 1990; Monteiro 1989; Erzini et al. 2002; Ribeiro et al. 2006, 2012).

Socio-Economic importance and anthropogenic pressures

Owing to its high biological productivity and ecological importance, the Ria Formosa is of high socio-economic value for fisheries, aquaculture, salt extraction as well as the tourism sector (Newton et al. 2003; Tett et al. 2003; Instituto Nacional de Estatística 2019). Almost 90% of the national clam production is generated in the Ria Formosa (Serpa et al. 2005; DRPASul 2006). Furthermore, substantial revenues are generated by the fisheries of cephalopod molluscs (i.e., cuttlefish, octopus) and finfish (i.e., mullets and seabream), the cultivation of finfish (e.g., sea bass and seabream), and the harvesting of invertebrates for bait (Cabral et al. 2019; Instituto Nacional de Estatística 2019). The tourism sector can be considered the booster of economic growth and development in the Algarve over the past decades (Sousa et al. 2020), propelling the continuous urbanisation of the adjacent terrestrial and coastal areas. The entire region, including the Ria Formosa, experiences seasonal, tourism-related fluctuations in population densities with increasing numbers of visitors recorded particularly during the summer months (Nobre 2009; Martins 2010; Newton et al. 2014).

Despite its acknowledged provision of ecosystem services and partial protection status, this highly dynamic coastal environment has been experiencing a deterioration of the water quality directly associated to the progressive urbanisation and economic exploitation of the lagoon’s living and non-living resources (Bebianno 1995; Newon et al. 2003; Newton and Mudge 2005). During the past decades, extensive research effort has been dedicated to the assessment of habitat degradation and various types of pollution, e.g., metals, microbes, noise, endocrine disruptors, pesticides (Bebianno 1995, Newton and Mudge 2003; Cunha et al. 2013; Rocha et al. 2013; Cruzeiro et al. 2015; Soares et al. 2020), and the level of impairment has been shown to vary along a spatio-temporal gradient, with higher anthropogenic pressures being exerted on floral and faunal communities at interior parts of the lagoon during the summer months (Newton et al. 2003; Cravo et al. 2012). In addition to the compromised water quality, the habitat integrity of the lagoon has been adversely impacted by the combination of anthropogenic activities (e.g., clam harvesting, fishing, tourism) and climate change, resulting in a substantial decline in seagrass coverage which amounts to 75% loss of *Zostera noltii* beds (Guimarães et al. 2012; Cunha et al. 2013). Apart from habitat degradation and loss, detrimental effects of coastal wetland reclamation and fishing mortality have been identified as the prevailing pressures on the Ria Formosa and its biological communities (Vasconcelos et al. 2007; Sousa et al. 2020). Annual beach seine monitoring of juveniles since 2000 has shown a decline in both fish diversity and density over the past two decades (pers. comm. K. Erzini; unpublished data).

The continuous improvement of the domestic sewage treatment since the mid 1990's, the renewal of the wastewater treatment plants in Olhão Nascente (2005), Faro Noroeste (2009) and Faro-Olhão (2018) and an increase in environmental education and awareness for human-induced threats affecting biodiversity initiated a slow recovery of the lagoonal system since the early 2000's (Newton and Icely 2002; Barracosa et al. 2019; Turismo de Portugal 2021 ; Águas do Algarve 2021). Nevertheless, the efficiency of wastewater treatment plants in removing MP pollution as another anthropogenic pollutant is still debated (Pommepuy et al. 2004; Raju et al. 2018; Liu et al. 2021) and the extent to which the Ria Formosa and its biota face exposure to plastic litter has only recently become the focus of scientific studies and consequently needs further investigation (e.g., Cozzolino et al. 2020; Lopes et al. 2020; Oliveira et al. 2020; Velez et al. 2020).

3.2 Model organism: White seabream

Sparidae (English: seabream or porgies; deutsch: Meerbrassen) is a percoid fish family comprising 159 different species across 38 genera which are distributed widely in the temperate and tropical Eastern and Western Atlantic, the Mediterranean, as well as in the Indian and Pacific Ocean (Hanel and Tsigenopoulos 2011; Froese and Pauly 2021). Members of this chiefly marine, bottom-dwelling fish family show a high morphological and ecological diversity as well as reproductive plasticity (Buxton and Garratt 1990), enabling them to sympatrically occupy various ecological niches across a range of habitats (Sala and Ballesteros 1997; Hanel and Tsigenopoulus 2011). Seabream are of commercial importance in many coastal regions (Erzini et al. 2002; Costa and Cautadella 2007; Vitale et al. 2011), and frequently use nearshore habitats as nursery grounds (Ribeiro et al. 2006; Abecasis et al. 2009; Vinagre et al. 2010). Juvenile sparids are known to show a high site fidelity while temporally and spatially partitioning the available micro-habitats among congeners (Erzini et al. 2002; Ventura et al. 2015), therefore allowing population dynamics and feeding ecology to be studied across different spatial and temporal scales.

Considering their wide distributional range along with their high economic importance, especially for small-scale fisheries, which provide 90 % of employment in the marine fisheries sector (The World Bank 2012), studies on the potential susceptibility of seabreams towards plastic pollution may be considered relevant for the sustainability of this target fish family and thus the livelihood of coastal communities depending on them. Given that ELHS of fish are the bottleneck of population development, research on potential impacts on these life stages is essential to understand variation in recruitment success as well as implications for fisheries. ELHS of seabream are a suitable model organism for this study due to their preferred habitat choice at the gateway of plastic pollution to the global oceans, along with their largely omnivorous feeding mode, which hypothetically makes them prone to encountering and ingesting MP of different shapes and sizes (Mizraji et al. 2017). One seabream species of the genus *Diplodus* will be studied in detail in this thesis: the white seabream (*Diplodus sargus*).

Diplodus sargus (Linnaeus, 1758)

The white seabream (Fig. 6 and Fig. 7) shows a distributional range from the Eastern Atlantic Ocean, throughout the Mediterranean to the Southwestern Black Sea (Joubert and Hanekom 1980; Sala and Ballesteros 1997; Abecasis et al. 2009; Froese and Pauly 2021). It inhabits a variety of marine habitats, e.g., coastal rocky shores, seagrass meadows over sandy bottoms, and its preferred vertical distribution ranges from 0 to 50 m depth (Whitehead et al. 1986; Erzini et al. 1996). *D. sargus* is of high commercial importance, especially for small-scale coastal fisheries and recreational fisheries along the Southern Portuguese coast and the Mediterranean, and is one of the dominant species in inshore coastal fish communities throughout its range (Figueiredo et al. 2005; Veiga et al. 2010; Terlizzi et al. 2011; Leitão et al. 2016). Based on official statistics from the Algarve auctions from 2005 to 2017, an average of 61.7 t (s.e. 8.7 t) per year with a first sale value of 727,341 € (s.e. 90,040 €) of white seabream were landed by the Algarve commercial fleet (Direcção Geral de Recursos Naturais, Segurança e Serviços Marítimos [DGRM], pers. comm.). However, this is an underestimate of the total catches and economic value as the data do not include undeclared commercial landings or those of recreational anglers and spear fishers that may exceed those of the commercial fishery (Veiga et al. 2010). Due to its high market value, it has also become a popular aquaculture species during the past 20 years particularly in the Mediterranean (Sá et al. 2006b; Hanel and Tsigenopoulos 2011).

Fig. 6 Juvenile white seabream *Diplodus sargus*. © Michel Bariche (2004).

Fig. 7 Adult white seabream *Diplodus sargus* off the Canary Islands. © Daphnis de Pooter (2013).

The white seabream displays a high variability of sexual phenotypes. While the majority of individuals are protandrous hermaphrodites, digyny, where both sexes mature from a non-functional intersexual phase, with males being still able to convert into secondary females, has also been reported for this species (Micale and Perdichizzi 1994; Mann and Buxton 1998; Morato et al. 2003). At the age of two to three years (TL at first maturity: 21.0 ± 0.3 cm), *D. sargus* reaches sexual maturity (Divanach et al. 1982; Micale and Perdichizzi 1994; Morato et al. 2003; González-Wangüemert and Pérez-Ruzafa 2012; Mouine et al. 2012); off the Southern Portuguese coast, white seabream spawn from December to May (Morato et al. 2003). ELHS settle in shallow coastal areas where they stay for several months and show a high site fidelity before migrating out of their nurseries to join adult populations (Macpherson 1998; Ribeiro et al. 2012).

The generalist *D. sargus* is known to undergo several ontogenetic dietary shifts (Christensen 1978); it feeds on a variety of prey items, among them crustaceans, molluscs, hydroids, anthozoans, echinoderms, polychaetes, other fish species as well as seagrass and algae (Sala and Ballesteros 1997; Mariani et al. 2002; Figueireido et al. 2005; Costa and Cautadella 2007; Leitão et al. 2007). Despite the omnivorous feeding habits, commercial importance and high abundances across micro- and macro-spatial scales, the sensitivity of *D. sargus* to plastic pollution, specifically to MP ingestion, has been investigated in a single study (Shabaka et al. 2020), though numerous dietary studies have been published over the past decades. The multi-species assessment on MP ingestion from a Mediterranean harbour (Egypt), found high MP-loads in all *D. sargus* individuals (n = 40) under examination, and proposed the omnivorous feeding mode as a driving factor for elevated plastic ingestion rates (Shabaka et al. 2020).

Considering the high urbanisation of the Ria Formosa in general, and the disconcerting outcomes of recent studies on MP distribution and uptake in the lagoon in particular (Cozzolino et al. 2020; Oliveira et al. 2020), further research initiatives, like the present one, are needed to provide more details on the potential ecological and economic implications of plastic debris on commercially relevant fish taxa and particularly vulnerable early life-history stages inhabiting the vegetated habitats of this vital ecosystem.

4 Publications

Chapter 1

“‘Oh really, and I can prove it to you with a few simple figures.’

‘Bosh!’ Ned replied. 'You can make figures do anything you want!’”

Jules Verne (1872): 20,000 Leagues under the Sea

Not as bad as it seems? A literature review on the case of microplastic uptake in fish

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Abstract

Within the past decade, microplastic (MP) particles (<5 mm in size) became the focus of both scientific and public attention, on one hand due to their cumulative industrial application and global presence, on the other hand due to their size range allowing the interaction with organisms at the base of the marine food web. Along with the growing evidence of their ubiquitous distribution, the ingestion of MP fibres and fragments has been verified for a variety of marine biota, with fish species being among the group of organisms most intensively studied both in the laboratory and in the field. While the gross of scientific literature focuses on the quantification and chemical characterization of MP in the gastrointestinal tract of fish, in-depth investigations on the impacts or a contextualization of ingestion are rarely accomplished. Yet, the constant media-coverage and omnipresence of the topic present a (threat) scenario among civil society which might lack a solid scientific foundation. This review, therefore, analyses the scientific output of 90 field studies covering 487 different fish taxa with due regard to explanatory variables for MP ingestion. Additionally, it highlights persistent knowledge gaps in relation to the examination of *in-situ* ingestion effects and proposes measures how to approach them in future research initiatives. Moreover, the potential existence of a publication bias and a consequent distortion of the perception of the topic is evaluated.

Keywords: Marine litter, Anthropogenic particles, Ingestion, Marine fish, Publication bias

1 Introduction

1.1 The history of field studies on MP uptake in fish

Among the diverse anthropogenically caused environmental issues threatening coastal and oceanic ecosystems, plastic litter along with its potential detrimental effects on marine biota has been gaining particular attention and awareness both in the scientific community and civil society (Cunningham and Sigwart 2019; Völker et al. 2019). A concomitant of the growing research effort to describe the distribution and abundance of plastic pollution across the marine realm is the application of different size classifications in the scientific literature (Andrady 2015; Thompson 2015); a common, rigorous definition of the different size classes of plastic litter, as proposed by GESAMP (2015; Fig. 1), remains to be formally adopted by the scientific community.

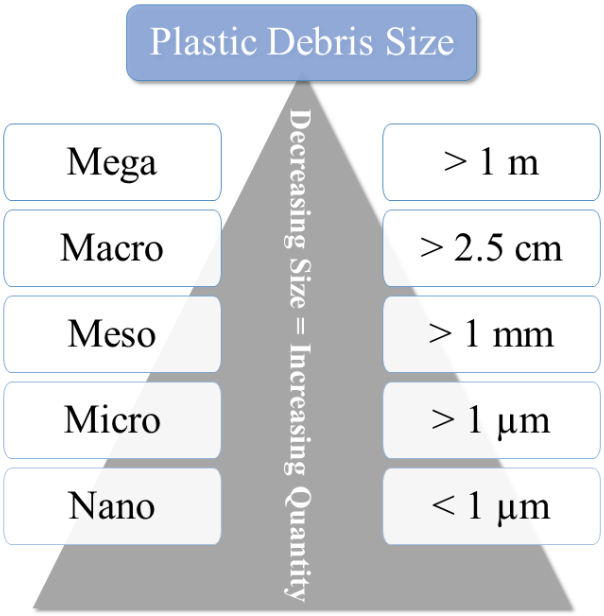


Fig. 1 Classification of plastic litter size ranges proposed by GESAMP (2015). With a decrease in size class, an increase in abundance (and, thus, bioavailability) is expected (Wright et al. 2013; Song et al. 2014).

With size being identified as one of the key factors determining the bioavailability of MP in the ocean (Wright et al. 2013), standardization of the size class definitions of MP is considered a top priority to holistically report and evaluate debris abundances in the marine realm and ingestion rates by different organisms on various levels of the trophic food web (Galgani et al. 2015). Irrespective of a lacking coherent categorization, especially micro-sized particles and fibres (commonly defined to range between 5 mm and 1 µm in, e.g., Arthur et al. 2009; GESAMP 2015, 2016) are of growing interest and concern: on the one hand, this is due to their increasing quantities in freshwater and marine environments as a result of cumulative industrial application of primary microplastics (Ryan 2015) along with the fragmentation of bigger-sized particles into secondary microplastics. On the other hand, due to their size range making them available for ingestion by a wide range of organisms at the base of the food web (Cole et al. 2013).

As early as in the 1970's, along with the onset of reports on MP in environmental samples (Buchanan 1971), early life-history stages of fish, as part of the marine zooplankton community, have been observed to take up MP. In a case study in the Northern Atlantic Ocean (Carpenter et al. 1972), the researchers collected fourteen different fish species by oblique plankton tows, of which eight were found to have ingested white, opaque polystyrene spherules. In investigations from the Severn Estuary (United Kingdom) by Kartar et al. (1973), the analysis of the stomach content of 0+ and 1+ year class flounder *Platichthys flesus* also revealed the presence of polystyrene particles in the guts. Upon publishing a progress report three years later, however, Kartar et al. (1976) stated that the same species of flounder (among other fish species investigated) collected over several years in the same area, was found to have varying levels of MP ingestion, including no uptake at all in some years. During the same time period, Colton et al. (1974) used a combined approach of large-scale field surveys and laboratory feeding experiments in an initial attempt to investigate the potential effects of MP uptake on larval and juvenile fish of different species. In more than half of the samples collected with a neuston net in ichthyoplankton surveys from Cape Cod to the Caribbean, plastic particles were found with varying abundances and plastic compositions. Yet, upon analysing the guts of over 500 larval and juvenile fish of 22 species collected in the respective areas, the authors did not find ingested plastic particles in any of the fish. Published in the same report, the outcomes of the feeding experiments with larval and juvenile fish of different species, which were offered polystyrene and acrylonitrile-butadiene-styrene suspension beads, likewise showed no MP uptake, as well as no MP-related mortality or detrimental effects (Colton et al. 1974). Following these early investigations, studies were published that highlighted the facts that the presence of MP in fish guts might be caused by postcapture ingestion (Lancraft and Robison 1980) and that not all fish species and age groups living in MP-contaminated environments are prone to ingest fragments or spherules to a high degree (Hoss and Settle 1990).

Since the start of investigations on MP ingestion by fish in the 1970’s, the number of research articles on this subject has been growing continuously (de Sá et al. 2018). Nowadays, MP ingestion has been verified for a multitude of different fish species from all around the world, including the North Pacific Central Gyre (e.g., Boerger et al. 2010; Choy and Drazen 2013), the North-Atlantic (e.g., Liboiron et al. 2016; Lusher et al. 2016) and South-Atlantic (e.g., Possatto et al. 2011; Pegado et al. 2018), the Mediterranean (e.g., Anastasopoulou et al. 2013; Güven et al. 2017), the North and Baltic Sea (e.g., Rummel et al. 2016; Beer et al. 2018) and even the polar regions (Kühn et al. 2018). Comparable to the early investigations from the 1970’s to the 1990’s, recent studies often tend to assess species assemblages rather than an individual taxon or life stage when examining the degree to which fish ingest MP (e.g., Davison and Asch 2011; Lusher et al. 2013; Jabeen et al. 2017; McGoran et al. 2018).

1.2 Reviews on MP ingestion by marine fish

To assemble and sort the growing number of reports from all over the world, several review articles have to date been published, covering MP ingestion by marine biota in general (Laist 1997) as well as by fish in particular. The first comprehensive list of plastic-ingesting marine fish species was collated almost twenty years after the early reports on this matter were published (Hoss and Settle 1990); this review included both scientific articles and anecdotal references of larval and juvenile stages of twelve fish species from nine different families which were reported to ingest MP in varying intensities. Reports on MP presence in the gastrointestinal tract (GIT) of adult fish were considered subsidiary results of dietary studies rather than specific investigations on MP uptake.

Within the past decade, publications of field-investigations on MP uptake by fish regularly incorporated topically limited literature reviews to put the obtained results into context: in a study on plastic ingestion by Atlantic cod, the authors reviewed 97 publications for plastic ingestion rates by different fish species, deliberately excluding species with an ingestion rate of 0% in their summary (Liboiron et al. 2016). In a baseline study on plastic ingestion by fish in the southern hemisphere, the researchers conducted a literature review of 15 studies with a special focus on methodologies used to establish a standardised sampling protocol for future data collections (Cannon et al. 2016). As part of their study on MP occurrence in commercial fish from a Portuguese estuary, a methodological review by Bessa et al. (2018b) took into account 22 different publications. In addition to the uptake rates among the number of different species investigated, the authors also compared the method of extraction and polymer verification. In a further MP ingestion study with Atlantic chub mackerel (*Scomber colias*), the researchers also conducted a brief literature review with the ISI Web of Science search, listed the respective outcomes of 31 studies in tabular form in the publication and put their obtained results in terms of MP uptake rate and predominant MP type and colour into perspective (Herrera et al. 2019).

The latest stand-alone review on field studies on plastic ingestion by marine fish systematically analysed 93 publications (Markic et al. 2020) and gave special attention to sample sizes and analytical methods. The review synthesized the information available on the number of marine fish species reported to ingest plastic and aimed to investigate patterns in plastic ingestion related to habitat, feeding strategy and geographical distribution. According to the authors, plastic ingestion was demonstrated for 65% of the fish species (n = 494) investigated up to January 2019, and the detection of plastic in the gastrointestinal tract (GIT) of a fish was stated to be highly dependent on adequate sampling sizes, as well as on the analytical methods applied.

Until today, however, methodological ambiguities remain regarding the detection and identification of MP and smaller sized particles (e.g., nanoplastics, Fig. 1) both in environmental samples and in marine biota (Käppler et al. 2016; Markic et al. 2020). The interpretation and comparison of the outcomes of different studies in terms of presence or absence of MP need to be done with caution, under consideration of the potential methodological bias resulting from different laboratory protocols (e.g., mesh sizes used for sampling and filtering, contamination control measures, inclusion or exclusion of fibres in MP reporting) and analytical approaches (e.g., naked-eye, infrared spectroscopy or mass spectrometry for identification and quantification).

1.3 Aim of this study

Beyond substantiating the mere presence of one or several MP particles in the GIT of marine fish, information on the actual ecological and physiological implications of MP ingestion remains scarce, despite the long history of MP-uptake investigations. Previously published literature reviews, both stand-alone articles as well as incorporated summaries in scientific field reports, primarily involved the collation of fish species, the respective frequency of MP-feeding and MP particles in the GIT, the representation of geographical locations and the appraisal of methodological approaches. To thoroughly evaluate the significance of MP ingestion by marine fish, however, these findings need to be contextualized (Fig. 2) and correlated with oceanographic properties (e.g., temperature, salinity, oxygen), environmental surveys (e.g., prey and MP abundance), GIT content analysis (e.g., abundance or volume ratio of natural prey items to MP particles) and an investigation of the potential impacts (e.g., assessment of the condition of the fish by various approaches).

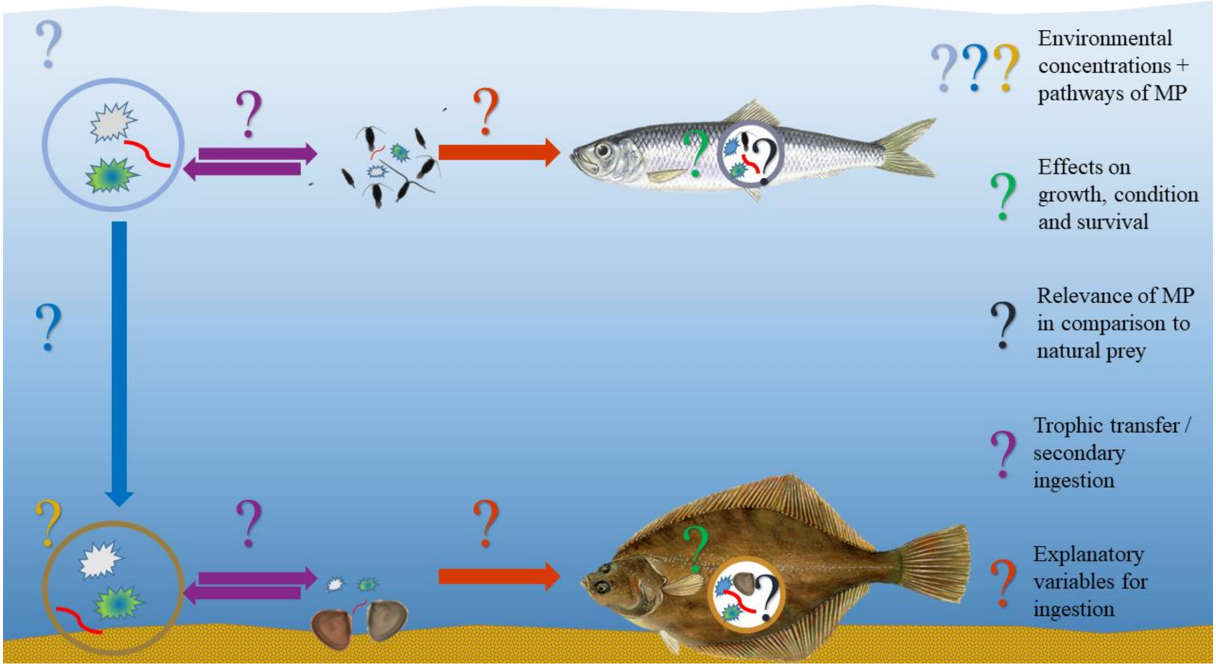


Fig. 2 Graphical overview displaying the challenges and open questions related to *in-situ* microplastic ingestion studies by showcasing two exemplary fish species, the pelagic *Clupea harengus* and the demersal *Platichthys flesus*, along with their potential MP exposure and ingestion, the latter being reported by several studies included in this review. Fish graphics ©Scandposters.com.

The constant increase in media coverage of the topic of marine (micro-)litter along with the increasing number of scientific studies reporting uptake and consequent detrimental effects for fish and a variety of marine biota, might lead to the assumption that ingestion is the natural corollary of MP presence in the ocean. Upon closer examination of the literature published within the past decades, however, it becomes obvious that there has been evidence proving the contrary: the scientific reports do not exclusively refer to fish species taking up MP. Among the MP-feeding species, there are frequently also those, which were found without any plastic at all in the GIT (Carpenter et al. 1972; Kartar et al. 1976; Steer et al. 2017; Vendel et al. 2017). Yet these ‘*negative*’ findings or even entire studies reporting a strikingly low or even zero-uptake along with minor or no effects observed both in the field and in laboratory set-ups appear to be rather unnoticed, underrepresented or even unpublished (Liboiron et al. 2018). This phenomenon, called (positive) publication bias is known across all scientific disciplines (Fanelli 2012; Mlinarić et al. 2017) and could potentially affect scientific and civil society's perception of the actual extent of MP ingestion by fish.

As fish are among the group of organisms potentially affected by MP ingestion which are most intensively investigated both in the field and in laboratory set-ups (de Sá et al. 2018), the aim of this study is threefold:

1. it will provide a brief quantitative and qualitative summary of the fish taxa examined in *in-situ* MP ingestion studies.
2. it will assess the current level of knowledge regarding driving factors of MP ingestion, along with the ecological and physiological implications of MP-uptake by fish to identify persistent knowledge gaps, future research projects should address.
3. it will investigate the potential existence of a publication bias in almost five decades of research on MP uptake to contribute to a sound evaluation of the extent and impact of MP-ingestion by marine fish.

2 Materials and Methods

2.1 Bibliographic research

A review of scientific papers on *in-situ* reports only was conducted to obtain a representative number of reports on fish and MP uptake along with the potential effects. The research was performed by using major scientific databases, including Google Scholar and ISI Web of Knowledge. Two key research terms were entered to identify relevant scientific papers: *plastic* and *fish*. The focus of the bibliographic research was laid on microplastic materials, i.e., anthropogenic particles and fibres of less than 5 mm in at least one dimension (Galgani et al. 2015). Studies focusing on this particular size range were considered relevant for this review; results reported on meso- or macroplastic even within the same studies were (if possible) deliberately excluded, the same holds true for studies in which a clear size attribution of the MP particles found was lacking. Studies on freshwater fish species were also not included.

Based on the ISI Web of Science data base inquiry ('*Web of Science Core Collection'*) with the keywords *plastic* and *fish* for TOPIC (TOPIC: (plastic fish); Timespan: All years.

Indexes: SCI-EXPANDED, SSCI, A&HCI, CPCI-S, CPCI-SSH, BKCI-S, BKCI-SSH, ESCI, CCR-EXPANDED, IC.), a total of 2608 studies were listed (status: 3rd July, 2019). Upon consulting the abstract of the listed publications, a total of 116 publications were preselected as potentially relevant. Combined with the results of a Google Scholar database inquiry and a meta-analysis of cited publications in the preselected literature, a total of 90 papers based on *in-situ* investigations was considered relevant for the objectives of this review.

2.2 Data analysis

To provide a broad perspective on *in-situ* MP uptake by fish, two databases were set up, focussing either on the fish taxa under investigation (Table 1, Annex 1) or on the publications and their respective characteristics under review (Table 1, Annex 2). The following main categories were assessed (if available) for each publication and analysed by descriptive means:

1) **Fish-related information** (family, species, life-stage, trophic guild, habitat, commercial importance for fisheries)
2) **Sampling-related information** (geographic region and location of the sampling, sampling time, environmental samples and parameters)
3) **Output-related information** (sample size, GIT analysis incl. prey ID or abundance, percentage of MP-feeding individuals, mean number of ingested MP per individual, impacts of ingestion)

The classification to either of the three life stages (adult = ad; sub-adult = sub-ad; larval/juvenile = juv) was based on the information given in the respective publication. If this information was lacking, the categories were derived from comparing the length measurements listed in the publication – if available - to the description accessed on FishBase (Froese and Pauly 2019). In case neither the life stage category in the actual publication nor the maturity record in FishBase were available for individual species, the term '*not applicable'* (n/a) was entered. A classification into trophic guilds, habitats and potential interest for commercial fisheries was also based on the data available on Fishbase (Froese and Pauly 2019). Furthermore, the number of citations was included; if accessible, the citation history as well (i.e., number of citations year^{-1}) based on the ISI Web of Knowledge or Scopus database, assessed on the 06th September 2019.

All statistical analyses included in this review, along with the visual representation of results, have been computed using a combination of Microsoft Office 365 Excel and RStudio, Version 1.2.1335. Level of significance was set to $p < 0.05$.

An in-depth review of the methodologies applied to detect and identify MP particles in fish GIT was not in the focus of this review as it has been comprehensively discussed in, e.g., Cannon et al. (2016), Bessa et al. (2018b) and Markic et al. (2020).

3 Results and Discussion

3.1 Bibliographic research

This study reviews a total of 90 papers based on *in-situ* investigations. Although the number and content of the research articles assessed were considered representative for a review on *in-situ* MP uptake by fish, this study does not intend to be an exhaustive collection of all reports published, especially given the growth of peer-reviewed publications over the past ten years. In previously published reviews, the number of reports assessed ranged between 8 and 97. To the best of our knowledge, the only other comparable stand-alone review published so far included 93 reports (Markic et al. 2020), thus, the number of publications included here was considered adequately representative. The results of the data compilation are given in Table 1 (Annex 1) and Table 1 (Annex 2).

While the earliest report on MP ingestion in fish dates back to 1972 (Carpenter et al. 1972), followed by studies from 1974 and 1976 (Colton et al. 1974; Kartar et al. 1976), the majority of studies reviewed here were published within the last four years (Fig. 3). Noteworthy is the gap between the years 1976 and 2010: a review on MP ingestion (Hoss and Settle 1990) as well as several dietary studies (e.g., Manooch and Hogarth 1983; Young et al. 1997; Joyce et al. 2002) were published in between those years, however, their content (i.e., the size spectrum of MP particles detected was not clearly defined) was not matching the criteria for this review and, therefore, not included in this bar chart. However, the general tendency of an increasing publication effort during the past five to ten years – as reflected by the selection of manuscripts in this review – was also attested by other reviews (Barboza and Gimenez 2015; Cunningham and Sigwart 2019; Markic et al. 2020).

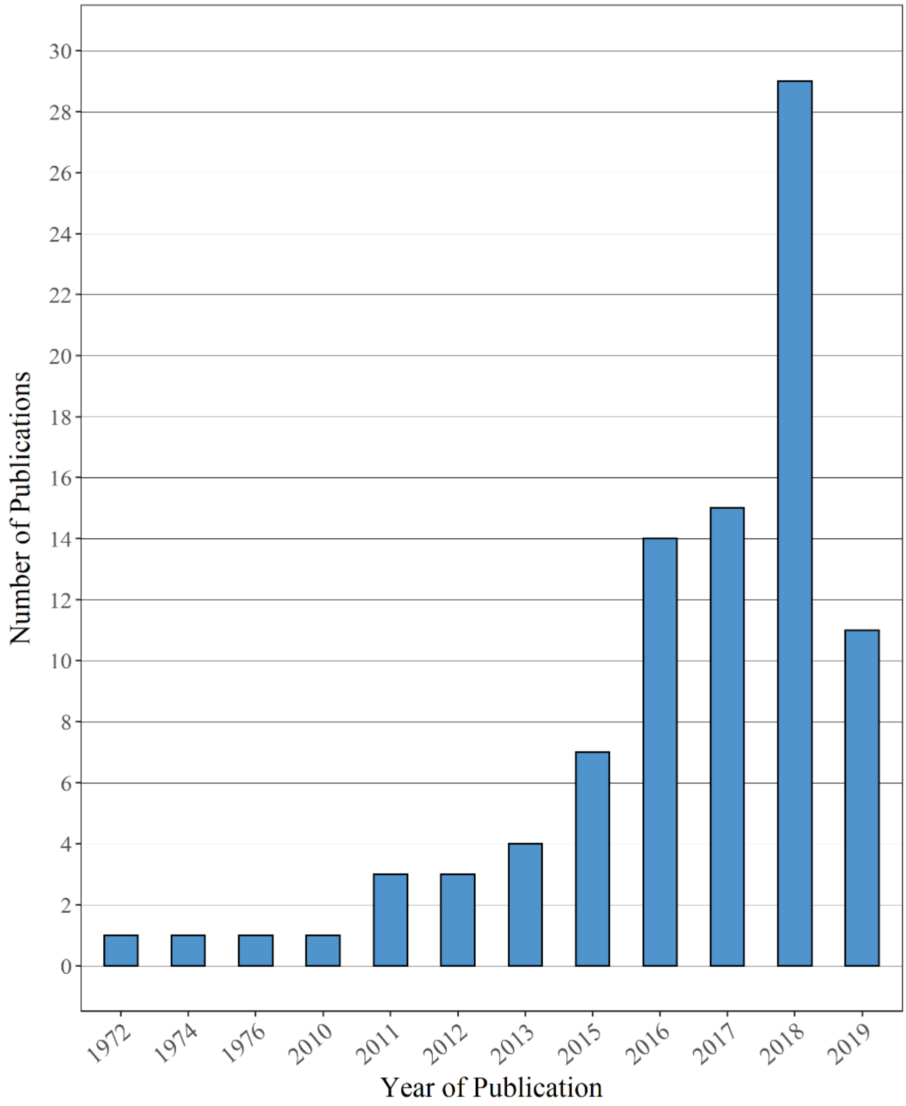


Fig. 3 Overview of the number of publications reviewed here, sorted by the respective publication year. Papers published until 3 July 2019 are considered in this review.

3.2 Quantitative and qualitative representation of fish taxa

3.2.1 Number of taxa per study and sample sizes

The MP uptake of a single species was assessed by 29 of the 90 studies reviewed here (e.g., Gassel et al. 2013; Naidoo et al. 2016; Smith 2018), the majority of field studies, thus, covering more than one species. Among those studies, 29 others dealt with up to five species, nine investigated six to ten species and nine others examined eleven to twenty different species. A total of 14 studies dealt with more than twenty different species and the highest number of species included in one report was 69 (Vendel et al. 2017). Several different studies investigated the same species (e.g., Kartar et al. 1976; Rummel et al. 2016; McGoran et al. 2017; Liboiron et al. 2019): upon examining those species, which were reported in more than one publication, the results in terms of MP uptake as well as in terms of mean number of particles per individual GIT vary greatly (Table 1). To provide a brief overview of those variations, the results for three frequently assessed species are exemplarily listed in Table 2, reporting on the minimum and maximum sample sizes given in one study, along with the corresponding minimum and maximum percentages of MP-feeding individuals as well as minimum and maximum number of particles detected in the GIT of these species.

Table 2 Exemplary overview on the MP-uptake results for three fish species, investigated in several different studies from different geographic areas (see references a – l). The outcomes of studies reporting on MP uptake of the same species sampled from various locations with clear differentiation between those are intentionally not summed here, e.g., Anastopoulou et al. (2018) or Kartar et al. (1976). References: [a]Güven et al. (2017); [b]McGoran et al. (2018); [c]Compa et al. (2018); [d]Kartar et al. (1976); [e]Liboiron et al. (2019); [f]Neves et al. (2015); [g]Rummel et al. (2016); [h]Renzi et al. (2019); [i]McGoran et al. (2017); [j]Foekema et al. (2013); [k]Anastasopoulou et al. (2018); [l]Bessa et al. (2018b).

	European pilchard (Sardina pilchardus)	**European flounder (*Platichthys flesus*)**	**Atlantic cod (*Gadus morhua*)**
Sample size minimum	7[a]	8[b]	1[b]
Sample size maximum	105[c]	530[d]	1010[e]
MP-feeding minimum %	15.24[c]; [0[f]]	5[d]; [0[b,d,g]]	1.4[g]; [0[b,g]]
MP-feeding maximum %	96[h]	75[i]	13[j]
No of MP / GIT minimum	0.09[k]; [0[f]]	0.18[l]; [0[b,d,g]]	1.1[j]; [0[b,g]]
No of MP / GIT maximum	4.63[h]	3.1[b]	1.12[e]
Geographic regions	Mediterranean, English Channel, Eastern North-Atlantic	Eastern North-Atlantic, English Channel, North & Baltic Sea	Western North-Atlantic, English Channel, Norwegian Sea, North & Baltic Sea

As can be seen from this exemplary list for three species only, the range in terms of sample sizes, frequency of MP-feeding, and MP particle load per individual fish seems to be quite substantial, without considering the variability in detection methodologies, particles under investigation, size ranges of both particles and fish as well as the life stages under investigation. Assessing all entries for individual taxa and life stages, collected across distinct geographic sampling locations, around one third of the fish were found with MP in their GIT (28.9 ± 33.91%), with an average particle load of around 2.2 ± 4.02 fibres and /or fragments among the

MP-feeding individuals. These results are largely consistent with the findings of another stand-alone review that found a slightly higher frequency of plastic-feeding fish 37.6 ± 0.6 %, with a comparable ingestion rate of 2.6 ± 0.2 pieces per individual (Markic et al. 2020).

Apart from this, the average sample size per species and study was calculated: in 28 of the studies included in this review, the average sample size per individual species was below thirty, in eight studies it was lower than ten (Table 1, Annex 1). Studies reporting on a single species tended to have higher sample sizes per taxon (e.g., Bråte et al. 2016; Ferreira et al. 2016a, 2018; Alomar et al. 2017) than studies that investigated species assemblages (e.g., Davison and Asch 2011; Rochman et al. 2015; Miranda and de Carvalho-Souza 2016; Pegado et al. 2018). According to the outcomes of the only other known stand-alone review of this dimension by Markic et al. (2020), the recommended sample size with a level of confidence of 95% and a 10% margin of error was $n \geq 91$. Applying this recommendation to the results included here, only 16 of 90 studies had an average sample size per species matching this criterion. Accepting a margin of error of 20%, the sample size recommended by Markic et al. (2020) was $n \geq 23$; 49 of the 90 studies reviewed here dealt with an average number of at least 23 individuals per species. With sample sizes below ten individuals per species in some studies, the significance of these results in terms of species-specific MP-uptake rates appears to be rather debatable (Liboiron et al. 2018; Markic et al. 2020). Moreover, comparing the results of different assessments to identify baselines of ingestion frequencies or patterns related to the uptake of MP per species is also not considered reasonable (Table 1), without even taking into account the variances in analyses, detection methodologies or sampling sites (Liboiron et al. 2019; Collicutt et al. 2019). Thus, the following results (Sect. 3.2.2) are considered to provide an overview rather than to quantitatively compare the results collated.

3.2.2 Representation of fish taxa and life stages

Representation of fish families

In total, the field studies assessed here were concerned with 142 different families, including both bony fish and cartilaginous fish taxa (Table 1, Annex 1). Members of the family Myctophidae ($n_{species} = 32$), Sciaenidae ($n_{species} = 26$), Carangidae ($n_{species} = 24$) were investigated most frequently, followed by Clupeidae ($n_{species} = 19$) and Sparidae ($n_{species} = 17$) (Fig. 4). Overall, 62 different families were represented by a single species only, e.g., Blenniidae, Chaetodontidae, Nemichthyidae, Pomacanthidae, and Scomberesocidae. Two taxa were investigated in 28 different families, and 16 more families were represented by three taxa each.

Representation of fish species and commercial importance

The present review collates information on a total of 487 different marine fish species, MP uptake was verified for around 67% of them (n = 327). The majority of species (80%; n = 393) was mentioned in only one study each. Among the remaining 95 species which were included in more than one publication, were several taxa of commercial importance (Table 1, Annex 1) such as the European pilchard *Sardina pilchardus*, the Flathead grey mullet *Mugil cephalus* and the European flounder *Platichthys flesus*. Of the 487 species included in this review, about 75% (n = 367) were considered commercially important for the fisheries sector based on the information accessed on FishBase (Froese and Pauly 2019). Data were not available for 86 species, with an additional 35 species rated as not relevant for the fisheries sector. According to FishBase (Froese and Pauly 2019), approximately 7,400 finfish species are used by humans (i.e., in fisheries and aquaculture, as bait or game fish, as well as in ornamental trade), 4657 of these freshwater and marine taxa are considered relevant for industrial and artisanal fisheries. Thus, this review collates information on 7.9% of the species of importance for the total fisheries sector. Irrespective of the individual outcomes of the MP-feeding studies on commercial fish taxa listed here, the overall significance of MP as a threat to the living marine resources is hardly assessable.

Representation of life stages

For the majority of the species listed (286 of 488), no clear classification of life stage was possible. This lack of data in this category may be explained by two major reasons: first of all, the missing data in the original publication – in terms of division to either of the categories or in terms of size dimension which would have been subsequently used here to derive the classification from the data accessible on FishBase (Froese and Pauly 2019) – or the lack of data on maturity size ranges for particular species in the online database itself. In a few cases, the results of different life stages were grouped in the publication and could not be assessed individually, which resulted in a combination of different life stages, e.g., juveniles + adults of *Allosa fallax* in Skóra et al. (2012) or in *Anchoa januaria* in Vendel et al. (2017); sub-adults + adults of *Engraulis encrasicolus* and *Sardina pilchardus* in Compa et al. (2018). For a total of 121 species, early life-history stages were considered with varying sample sizes across the different studies, ranging from one individual only, e.g., *Trisopterus minutus* in McGoran et al. (2018) to a maximum of 1090 individuals of *Platichthys flesus* in Kartar et al. (1976). For another 81 species, only adult individuals were investigated for MP ingestion.

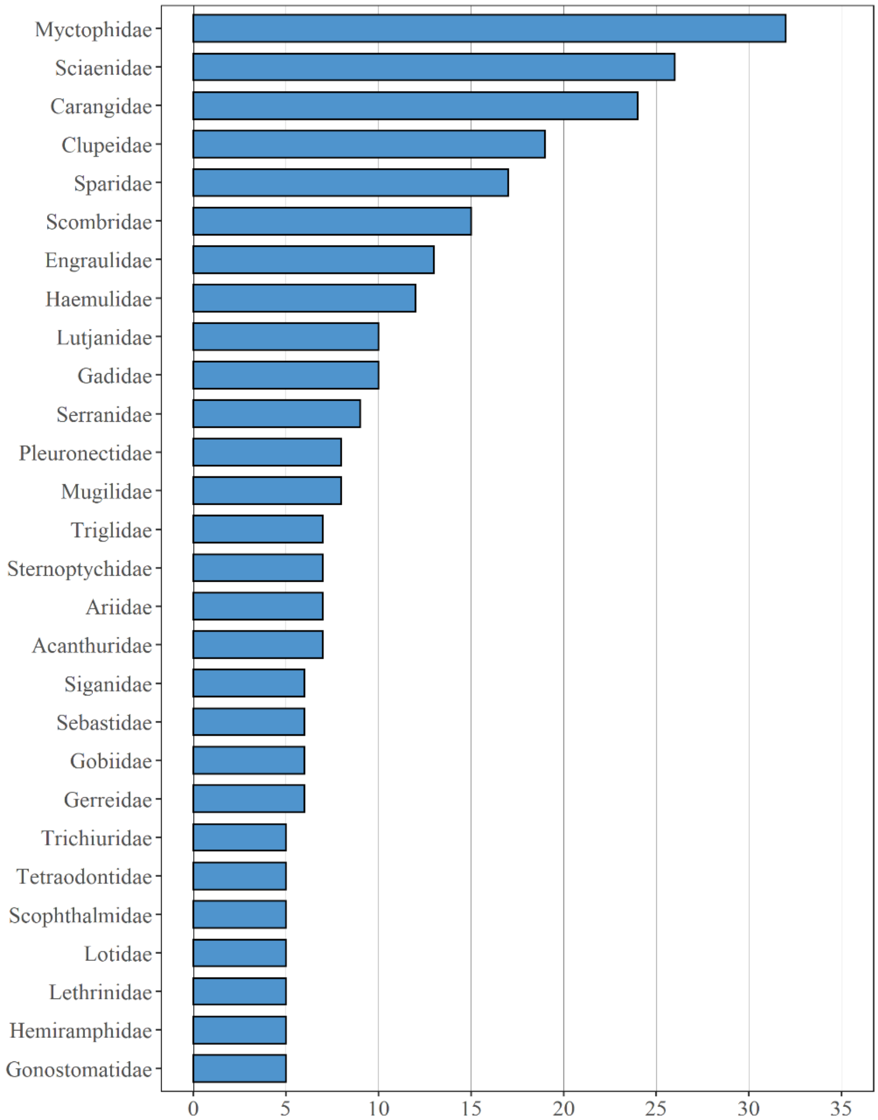


Fig. 4 Fish families which are represented by at least five different species mentioned in the 90 studies under review here.

3.3 Drivers of MP ingestion

Along with establishing growing evidence for MP-ingestion by a wide range of marine fish species, researchers seek to elucidate the driving factors of MP uptake. One of the most frequently investigated hypotheses correlates the size of the fish (e.g., standard length or total length) with the amount or size of MP particles taken up (e.g., Boerger et al. 2010; Foekema et al. 2013; Güven et al. 2017; McGoran et al. 2018), while two other prominent hypotheses aim to identify patterns related to the trophic guild / feeding mode of the fish (e.g., Anastasopoulou et al. 2013; Vendel et al. 2017; Sun et al. 2019) as well as the preferred habitat (e.g., Güven et al. 2017; Markic et al. 2018).

3.3.1 Body dimensions (size)

In fishery biology surveys and studies, the assessment of the size and weight of a fish has been routinely performed for decades (Smith 2002) – the same routine seems to be applied for studies on MP ingestion in fish. Frequently, measurements of standard length (SL) (e.g., Boerger et al. 2010; Dantas et al. 2012), total length (TL) (e.g., Anastasopoulou et al. 2013; Ory et al. 2018a) or fork length (FL) (e.g., Battaglia et al. 2016; Chagnon et al. 2018) were taken upon collecting or dissecting the fish. In other cases, the lengths of the fish were recorded, but no specification of the measurement was given in the publication (e.g., Neves et al. 2015; Peters et al. 2017) while in some investigations, no length was assessed at all (e.g., Davison and Asch 2011; Wesch et al. 2016a) or the data are not provided in the text even though the researchers reported measuring size and weight of the fish (Miranda and de Carvalho-Souza 2016; Su et al. 2019a). Due to these inconsistencies or entire absence of data on fish body dimensions, the results obtained are difficult to compare across all studies under review here. Notwithstanding, within the individual studies that collected these size-related data, the measurements were occasionally used to investigate a potential correlation between MP uptake (i.e., number of particles taken up, size of particles ingested) and body size of the fish. Comparable to the outcomes of impact-assessments, the results of these correlations turn out to be rather inconsistent. In their study on seven North Sea fish taxa, Foekema et al. (2013) were not able to relate the size of the MP particles ingested to the size of the MP-feeding fish beyond doubt. Likewise, the size of the ingested MP (fibres and non-fibres) by fish collected in Turkish territorial waters was not correlated to the size of the MP-feeding fish itself (Güven et al. 2017). Contrastingly, Sun et al. (2019) found a positive correlation between the length of MP particles detected in the GIT of fish from the Yellow Sea and the fish length.

The existence of a potential correlation between the number of MP particles detected in the GIT and the size of the fish was investigated in several of the studies reviewed here, again with partially divergent outcomes: in a study on 60 lemon damselfish *Pomacentrus moluccensis* from two locations of the Great Barrier Reef, Jensen et al. (2019) did not find a correlation between the number of MP ingested and the fish TL. Giani et al. (2019) were also not able to detect a correlation between the TL or weight of the examined 132 red mullet *Mullus barbatus barbatus* and 97 European hake *Merluccius merluccius* and MP ingestion – neither in terms of occurrence of MP ingestion by fish (in %) nor in terms of particles per individual. For some elasmobranch species, the observations made by Alomar and Deudero (2017) and Bernardini et al. (2018) suggest that early life-history stages (i.e., juvenile or immature individuals) show a higher probability to ingest MP particles than adults.

As opposed to the abovementioned results, Cheung et al. (2018) found a positive correlation between the abundance of MP and the FL of flathead grey mullet *Mugil cephalus*, suggesting larger individuals tend to take up MP particles to a higher extent. Similar observations were made by McGoran et al. (2018) in their comparative study of MP ingestion by fish from the Thames estuary and the Firth of Clyde: according to their results, larger fish (based on SL) ingested more MP particles than smaller ones. The same positive correlation between SL of the fish and the number of MP particles ingested was found for various fish species from the Amazon River estuary (Pegado et al. 2018) and the North Pacific Central Gyre (Boerger et al. 2010). For both Atlantic herring *Clupea harengus* and European sprat *Sprattus sprattus*, Beer et al. (2018), furthermore, substantiated the same correlation for fish TL and the number of MP particles ingested.

A different connection was investigated by Azad et al. (2018): in their study, the researchers aimed to identify a potential correlation between the mouth size of the fish and the frequency of occurrence of MP in the GIT as well as the size of the MP particles detected. No significant relationship could be attested between the mouth size and either of the MP characteristics. However, as the 24 different fish species under examination showed contrasting tendencies, both positive and negative correlations were found between mouth size and MP frequency of occurrence.

Reasons for the lack of a definite tendency on how body size and MP particle ingestion potentially correlate may be seen for one part in the – to some extent - insufficient data basis of the *in-situ* studies: comparisons of various taxa with (partially) small sample sizes (Vendel et al. 2017; Pegado et al. 2018), little variation in terms of size classes (Boerger et al. 2010), differing sampling sites (Alomar and Deudero 2017) and variation in the residence time of prey and/or MP in the GIT related to the feeding mode of the fish (Gassel et al. 2013; Ory et al. 2018a) may contribute to the vagueness of results. For the other part, secondary ingestion or trophic transfer along the food chain may play an important role, currently underestimated in the scientific literature (Peters et al. 2017). Furthermore, shifts in dietary preferences and changes of foraging strategy related to ontogenetic development, with younger individuals showing a tendency for more opportunistic feeding strategies (Balon 1986; Rønnestad et al. 2013; Critchell and Hoogenboom 2018) may confound the relationship between body size and MP ingestion. Lastly, the possibility of a non-existence of a correlation for all fish taxa alike should not be precluded. Repetitive analyses of the same species or populations from the same sampling site, sampled in adequate numbers over a sufficiently long timespan, may establish evidence for or against the existence of a correlation of body size and MP ingestion on a species-specific level.

3.3.2 Trophic guild and feeding strategies

Different feeding strategies in fish may involve various expenditures of time for satiation (i.e., feeding) and also gastric evacuation; to cover their energy demand, filter feeding, detrivorous or herbivorous taxa for instance spend more time collecting (numerically more but less nutritious) prey items than predatory, carnivorous taxa which rely only on few, substantial meals (Pandian and Vivekanandan 1985). These higher temporal costs of feeding and, thus, the potentially higher probability to encounter MP particles and fibres while feeding, however, may be counterbalanced by shorter digestion rates for herbivorous taxa than carnivorous ones (Fänge and Grove 1979). The trophic guild may, therefore, represent an important explanatory factor for MP presence in the GIT of a fish.

The fish taxa assessed in this review were divided into different feeding categories, following the data accessed on FishBase (Froese and Pauly 2019) on preferred prey items ingested; the entries can be assessed for each species in Table 1 (Annex 1). The different categories were defined as follows: herbivore (feeding on plant material exclusively), planktivore (including filter feeders, ingesting both zoo- and phytoplankton), carnivore (preying on other animals), and omnivore (taking up plant-based materials and other animals alike). According to the data available on FishBase, two species were rated exclusively piscivorous, (*Caranx papuensis* and *Scomberoides tol*). The majority of taxa was classified as carnivorous (n = 254), followed by planktivorous (n = 61), omnivorous (n = 26), and herbivorous (n = 20). Moreover, due to the lack of information available, a number of species (n = 125) could not be attributed to any of the abovementioned categories.

The proportion of MP-feeding individuals across the different trophic guilds was compared by taking into account those entries which included both the MP uptake given for the respective fish taxa in the publication as well as the trophic guild listed according to FishBase (Fig. 5) – this figure includes also the different entries made for the same taxa by various studies which explains the variation in total numbers given in the boxplot as compared to the listing above. The median proportion of MP-feeding individuals ranged between 11.25% (omnivorous taxa) and 20.0 % (planktivorous taxa). The proportion of MP feeding individuals, represented by the interquartile ranges, is less variable in the herbivorous guild compared to the three others. Moreover, except for two outliers, the overall range in the proportion of MP-feeding individuals in this guild is also smaller. However, this feeding guild was also represented by the lowest number of taxa. The Kruskal-Wallis rank sum test was applied as the data were not normally distributed (Shapiro-Wilk normality test, ***P*** < 0.01): the tendencies described above were not substantial and no significant difference in the proportion of MP feeding individuals across the different trophic guilds was verified (***P*** = 0.7316; df = 3). A comparison of the mean number of ingested MP particles per individual likewise revealed no significant difference between the MP uptake rate of the four feeding guilds (Kruskal-Wallis rank sum test, ***P*** = 0.4949; df = 3). In the only other stand-alone review of comparable dimensions, the authors also compared the representation of feeding guilds among MP-feeding and MP-non-feeding taxa – likewise, no correlation between MP ingestion and feeding mode was ascertained (Markic et al. 2020).

Taking into consideration the results collated at a study-specific level, without interference of different sampling efforts and detection methodologies, the potential existence of correlations between MP ingestion and feeding mode or trophic level was investigated in several publications under review here. Vendel et al. (2017) chose a comparable approach to the one applied here, by analysing the prey items identified in the full GITs of the fish, and subsequently identifying five different feeding guilds to which the fish were classified: generalist species, benthivorous, zoobenthivorous, algae eating or zooplanktivorous species. Although MP was found in all the feeding guilds, the highest MP ingestion was detected in zoobenthivorous species (*A. lineatus* and *D. auratus*). A different categorization was chosen by Markic et al. (2018), describing not only the preferred prey category but also partially incorporating the feeding mode: grazers, omnivores, planktivores, benthic predators, and pelagic predators. The study found a significant difference among the five trophic guilds with benthic predatory fish showing the lowest MP uptake rate and omnivorous fish showing the highest, even though the authors acknowledged that only two fish species were classified as omnivorous, potentially limiting the significance of results obtained.

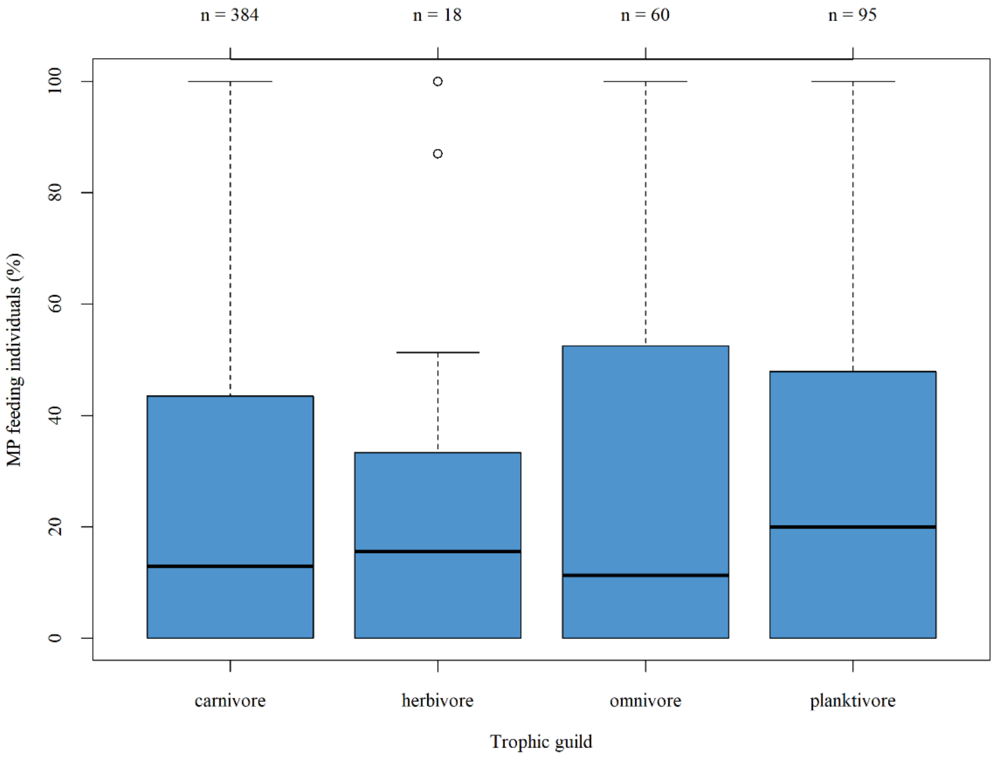


Fig. 5 Boxplot showing the proportion of MP-feeding individuals per trophic guild. Piscivorous taxa are included in the carnivorous guild, as only two taxa were classified as piscivorous according to FishBase.

Omnivorous fish species were also reported by Mizraji et al. (2017) to display higher MP ingestion rates in comparison to herbivorous or carnivorous species. These findings, however, are contradicted by the study of Jabeen et al. (2017), who detected the lowest MP uptake rate in omnivores, with higher rates detected in carnivores and planktivores. Following the categorization applied by Bour et al. (2018), the results indicate a significantly lower number of MP particles ingested by filter-feeders in comparison to predators, though the results concerning the frequency of MP-feeders among the different guilds did not differ significantly. As opposed to those tendencies, research by Güven et al. (2017) and Sun et al. (2019) as well as the review provided by Markic et al. (2020), did not establish significant evidence for a correlation between feeding mode / trophic guild and MP uptake rates.

The relationship between feeding mode and the type of MP taken up was assessed for instance by Anastasopoulou et al. (2013) who found that nektobenthic opportunistic feeders, represented by *G. melastomus*, ingested all identified plastic debris categories, whereas pelagic and bathypelagic feeders ingested either plastic bags and hard plastics, respectively. Another observation by Markic et al. (2018) highlighted the tendency of omnivorous and grazing fish species to ingest predominantly fibres, with ingestion of MP fragments more important in benthic and pelagic predators.

According to the majority of studies, there is no statistically significant correlation between the trophic level of a fish species and the quantity of MP particles taken up (Güven et al. 2017; Bour et al. 2018; Markic et al. 2018; Pegado et al. 2018; Sun et al. 2019). Only in a single study did results indicate that organisms at higher trophic levels also show a higher abundance of MP particles in the GIT, supporting the idea of trophic transfer of particles (Rios-Fuster et al. 2019). Evidence for trophic transfer of MP was, furthermore, found in 10 out of 57 fish examined by Markic et al. (2018), indicating an indirect uptake of MP from prey to predator, previously described on the basis of laboratory feeding experiments (Farrell and Nelson 2013; Setälä et al. 2014).

As the GIT content features only a current snapshot of the recent food intake, the rates at which marine fish taxa ingest synthetic particles and the extent to which they are exposed to potentially detrimental effects of this ingestion should be contemplated in the context of species- and life stage-specific GIT retention times (Beer et al. 2018; Markic et al. 2018) or in the special context of prey availability and environmental parameters (see Sect. 3.5.2 and 3.5.3).

3.3.3 Habitat

As essential feeding, spawning, and nursery habitats of fish overlap with input sources and fluxes of MP which, in return, facilitates biota-MP-interactions (Fig. 6), categorizing fish taxa according to their preferred habitat is another approach commonly used in the scientific literature to identify potential explanatory variables for MP ingestion. As the marine realm can be divided horizontally (e.g., inshore, neritic, oceanic) and vertically (e.g., benthic, epipelagic), it seems advisable to combine both to thoroughly investigate potential patterns in MP uptake related to the choice of habitats.

The only study in which a consistent classification to both zones was applied, found a tendency related to the vertical distribution of fish: benthopelagic fish showed the highest uptake rates in comparison to pelagic or demersal taxa (Markic et al. 2018). The study, however, did not detect any statistically significant horizontal patterns. In contrast to these results, a study from Turkish territorial waters, in which a vertical-horizontal combination of categories was applied that was based on the entries available on FishBase (Froese and Pauly 2019), detected slightly higher ingestion rates in fish species from the pelagic-neritic zone than in taxa from the benthopelagic, reef-associated, demersal, or pelagic-oceanic zones (Güven et al. 2017). Due to the overall lack of a consistent classification scheme in the literature reviewed here along with the variability in fieldwork approaches and laboratory methodologies applied for analysing MP uptake in fish, a quantitative evaluation and assessment of the results collated was not considered expedient. A similar conclusion was obtained in another stand-alone review on *in-situ* studies of MP ingestion in fish (Markic et al. 2020). Therefore, a brief overview of the reported outcomes and tendencies will be given in the following.

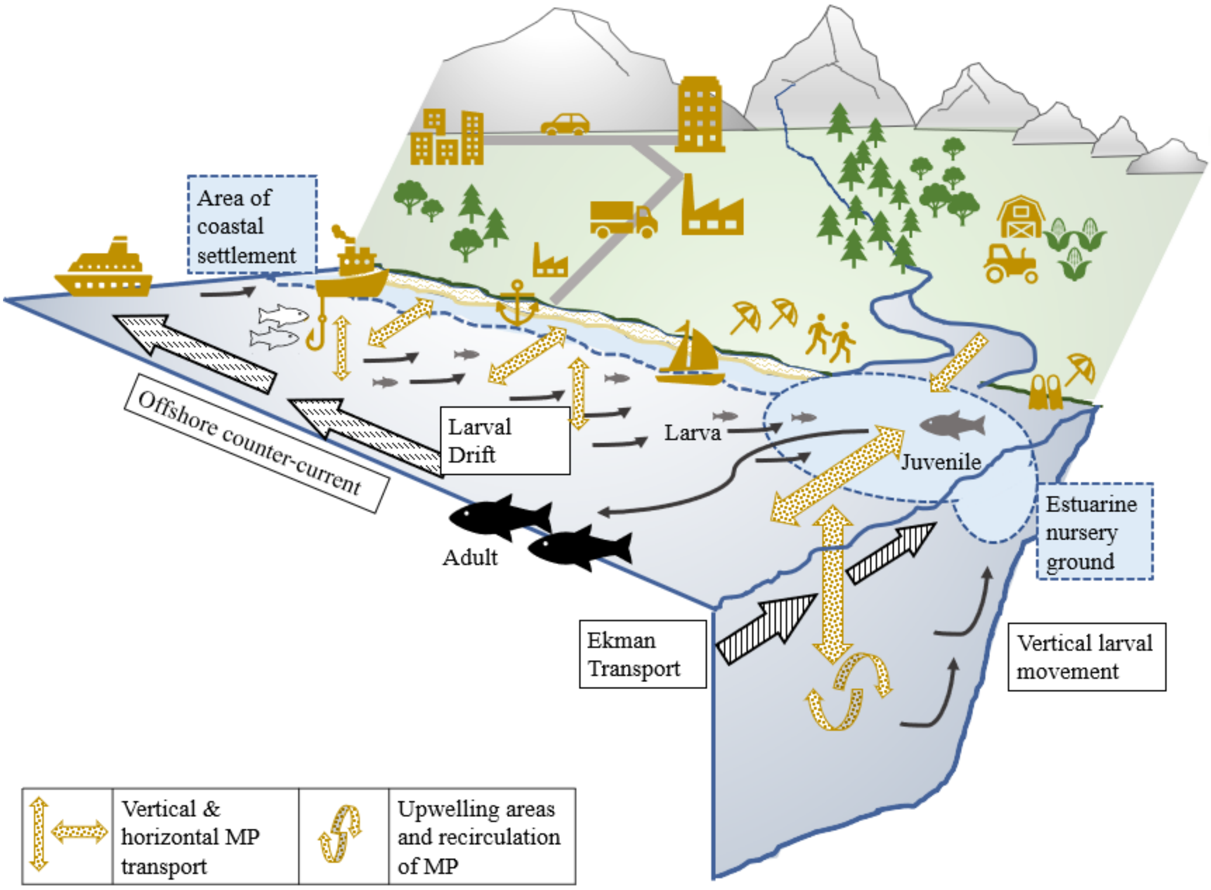
Horizontal Habitat Classification

As the majority of marine MP originates from land-based sources (GESAMP 2016), the horizontal distribution and pathways of plastic litter across neritic and oceanic habitats may represent a relevant explanatory variable of MP ingestion; however, only few studies applied a horizontal habitat classification to identify potential driving factors for MP ingestion. Murphy et al. (2017) found a tendency for coastal species to ingest significantly higher amounts of MP than offshore species. Sun et al. (2019) also reported lower numbers of MP per fish, if the fish was collected with greater distance to the coast and in the central Yellow Sea. In comparison to the amount of published MP research conducted in surface waters and accumulation zones (e.g., Boerger et al. 2010; Gassel et al. 2013; Ory et al. 2017), there is an apparent knowledge-gap concerning the exposure and uptake of MP across horizontal scales, especially in estuarine and coastal environments which are of special ecological, economic, and social importance (Martínez et al. 2007). Moreover, the connectivity between coastal and open-ocean areas in relation to fish abundances and MP uptake by various life stages along horizontal gradients (Fig. 6) should be prioritized by prospective research initiatives to enhance the assessment of potential vulnerabilities of specific fish taxa or vital life stages.

Vertical Habitat classification

The vertical distribution and pathways of plastic particles and fibres in the water column have been investigated by various studies (e.g., Kukulka et al. 2012; Enders et al. 2015; Cole et al. 2016; Choy et al. 2019). Similarly, published research included in this review more commonly categorized fish taxa according to their distribution across vertical marine zones. A broad allocation to either the benthic or pelagic realm was chosen by Neves et al. (2015): in their study of 26 commercial fish species sampled off the Portuguese coast, 63.5 % of the benthic fish species and 36.5% of the pelagic species ingested MP. A comparable categorization was chosen by Zhu et al. (2019b), who found significantly higher MP uptake in demersal fish than in pelagic fish from the Maowei Sea. In line with these observations, Murphy et al. (2017) reported significantly higher numbers of ingested MP in demersal fish species than in pelagic ones. In contrast, in a comparison of MP ingestion across three fish species, a significantly higher MP number per gram of GIT content (wet weight) was found for fish from the pelagic zone in comparison to demersal fish (Digka et al. 2018). In the Mondego estuary (Portugal) significantly lower ingestion rates were detected again for demersal fish species *D. labrax* and *P. flesus* in comparison to the benthopelagic *D. vulgaris* (Bessa et al. 2018b). No significant difference in MP uptake was verified, however, between five demersal and five pelagic fish species collected in the English Channel (Lusher et al. 2013).

Comparable to the attempt to link MP uptake to feeding modes, illustrated in Sect. 3.3.2, the abovementioned brief collection of outcomes from different studies of various geographic regions and fish taxa highlights the inconsistency of results as well the remaining ambiguities in establishing habitat selection as an explanatory variable for MP ingestion in fish. Conceivable reasons for the (currently) missing links between habitat selection and MP ingestion may be the lack of environmental samples collected along with the fish-samples for contextualisation (Sect. 3.4.3 and 3.4.3) as well as the rarity of investigations on early life-history stages (ELHS) of fish (Table 1, Annex 1), as part of the plankton community potentially representing a proxy for horizontal MP fluxes. Besides the classification of fish to specific feeding modes or preferred habitats, studies also investigated the relevance of other internal and external influencing factors of MP uptake and ingestion (Supplementary Materials).



	MP Input: Land-based	Exemplary Reference
🏢	Urbanized areas (e.g., waste water, municipal litter, landfills)	Wolff et al. 2019
🚚 🚗	Private and commercial traffic (e.g., tyre abrasion and road marking)	Kole et al. 2017
🏭	Industries (e.g., pellet spillage, industrial litter)	Derraik 2002
🚜	Agriculture (e.g., sewage sludge)	Corradini et al. 2019

	MP Input: Sea-based	Exemplary Reference
⚓	Harbours (e.g., paints, antifouling, abrasives)	GESAMP 2016
🎣	Fisheries and Aquaculture (e.g., fishing gear, mariculture structures)	Lusher et al. 2017
🚢 ⛵ 🚶 ⛱	Tourism (e.g., litter items, personal care products, transportation)	Arcadis and EUCC 2014 Galgani et al. 2015

Fig. 6 Schematic representation of the transportation and movement processes of marine fish larvae (spawned offshore) to inshore, estuarine nursery grounds (after Boehlert and Mundy 1988) in combination with the illustration of the spatial distribution of micro-sized plastic particles and fibres from land- and sea-based sources into different estuarine and marine compartments which overlap with vital fish habitats.

3.4 MP ingestion put into context

Scientific research efforts (especially based on laboratory experiments) have been substantiating a range of ecological, behavioural, and physiological implications for freshwater and marine fish taxa, though the effects of exposure and ingestion appear to vary across species and life stages, including even no measurable negative impact (Jovanović 2017; Foley et al. 2018). Given the broad research area of MP ingestion and MP-related effects, this review paper does not aim to reproduce the various impacts that have been verified to date. To give a brief summary, however, some of the detrimental implications of MP ingestion for fish, evidenced predominantly by laboratory studies, should be mentioned here: histopathological intestinal alterations (Pedà et al. 2016), translocation of particles (Avio et al. 2015) and consequent hepatic stress (Rochman et al. 2013), metabolic changes (Cedervall et al. 2012; Lu et al. 2016) and neurotoxic effects (Oliveira et al. 2013), reduction of predatory performance (de Sá et al. 2015), as well as increasing mortality of ELHS (Mazurais et al. 2015). By presenting the descriptive measures commonly applied in field studies, the aim of the present review is rather to focus on what *in-situ* studies on marine fish species have so far substantiated along with suggestions regarding the advancement and amendments of the state-of-the-art procedures.

3.4.1 Impact of ingestion

The detrimental effects of intentional, accidental, or secondary ingestion of plastic debris by marine biota are manifold and both, physical (Goldstein and Goodwin 2013; Lusher et al. 2013) and chemical hazards (Teuten et al. 2009; Rochman et al. 2013), have been shown to directly or indirectly impact growth, condition, and survival of different life stages of fish (Supplementary Materials). At present, the investigation of MP-uptake related impacts on fish are predominantly based on laboratory feeding experiments (Morgana et al. 2018), which in many cases lack consistency between natural conditions and experimental set-ups (Phuong et al. 2016; Lusher et al. 2017a). As the focus of this literature review is entirely on *in-situ* studies, it will report exclusively on the MP-related impacts investigated in those studies under consideration; the analysis of potential effects of MP-ingestion was, however, performed rather infrequently (in 11 of 90 studies). The majority of these studies investigated the impact of MP in the GIT on the condition of the fish based on morphometrics (Supplementary Materials): eight studies assessed the condition based on Fulton's condition factor K (Foekema et al. 2013; Rummel et al. 2016; Mizraji et al. 2017; Cardozo et al. 2018; Compa et al. 2018; Kazour et al.

2018; McGoran et al. 2018; Arias et al. 2019), two more studies (Ramos et al. 2012; Dantas et al. 2019) made use of the formula of Richardson's condition factor CF (Richardson et al. 2011). The outcomes of these investigations are equivocal; taking into consideration only the seven reports in which Fulton's K was calculated, one found a statistical significant difference between MP-feeding and non-feeding individuals (Cardozo et al. 2018). In their study on Atlantic bigeye (*Priacanthus arenatus*), Cardozo et al. (2018) observed that the ingestion of plastic fragments affected the condition of these fish with MP non-feeding individuals showing a significantly higher condition factor than the MP-feeding individuals in their study. The authors hypothesized that the presence of plastic particles in the GIT might ultimately lead to reduced body weight (and, thus, a lower condition factor) due to a decrease in feeding activity as a result of the false saturation feeling. Contrasting these findings, Mizraji et al. (2017) verified a significant negative relationship between the ingestion of MP and the condition of the omnivorous *Girella laevifrons*, which was interpreted by the authors as an indicator for a potential negative effect of MP uptake on the health status of an individual.

According to the results of a model by Compa et al. (2018) for *Sardina pilchardus* from the Mediterranean, fish with lower condition factors were more likely to ingest MP particles. However, in the same study, the model predictions for *Engraulis encrasicolus* showed the opposite: individuals with higher condition factors would rather take up MP than those with lower body condition. The authors state, however, that no significant relationship between the MP uptake rate and the body condition of a fish could be established by their model. In their study of seven different North Sea fish species, Foekema et al. (2013) were not able to verify any fundamental effect of MP ingestion on the condition of the MP-feeding fish, originating from five of the seven species under investigation. In an assessment of MP uptake by wild and caged juvenile European flounder *Platichthys flesus*, Kazour et al. (2018) also found no significant correlation between the condition and the number of MP particles taken up at different sampling sites in the eastern English Channel. In an Argentinian estuary, a similar observation was made by Arias et al. (2019) who did not detect statistically significant differences in the condition of individuals of the whitemouth croaker *Micropogonias furnieri* with plastic in their GIT in comparison to individuals without MP particles. Comparably, the condition factor of MP-feeding and MP non-feeding individuals did not differ significantly in the demersal bigeye sculpin *Triglops nybelini* and the pelagic polar cod *Boreogadus saida* collected off Northeast Greenland by Morgana et al. (2018). Rummel et al. (2016) also did not detect any effect of MP ingestion on the condition of different demersal and pelagic fish species collected in the North Sea and Baltic Sea.

The results obtained by calculating Richardson's condition factor appear to be equally inconclusive: according to Dantas et al. (2019), a higher condition factor for juvenile and adult fish without MP fragments in the GIT was found in their study. Following the interpretation of the authors, this result indicates that the ingestion of plastic could cause a reduced body condition. In contrast, Ramos et al. (2012) could not establish any evidence for a lower condition in MP-feeding members of the family Gerreidae in their study from the Goiana estuary.

While the assessment of condition factors is a commonly used tool to describe the physical state of individual fish or a fish population, potential differences in condition based on morphometric measurements should be evaluated with care. According to Froese (2006), the intraspecific variation in weight-length relationships may be significant, depending on the population itself, the season or varying environmental conditions between years. Depending on the group of organisms to compare, Froese (2006) suggests making use of different condition factors such as the Le Cren (1951) relative condition factor for comparing the relative condition of individuals within a sample. For studies of the relative condition across populations or taxa, relative weight in relation to mean weight was advised. One constraint in the evaluation based on condition factors, however, is their dependence on body length and weight of the fish, which on the one hand can be easily assessed in all routine sampling procedures making them available without additional handling effort, yet on the other hand, these morphometrics display different responses to varying environmental pressures (e.g., MP exposure or uptake) rather slowly (Markic et al. 2018). Unless the ingested MP particles accumulate over a long period of time in the individual, leading to a reduced food-uptake due to false saturation feeling and consequent reduced fitness and condition of the individual, the indicative meaning of condition factors based on body dimensions might bear only limited significance.

Another option to assess the potential impact of MP exposure and uptake is to analyse only specific organs such as the liver. Due to its detoxification function and the potential impact of contamination exposure to the size and weight dimensions of this organ, the hepatosomatic index (HSI) was calculated in one study (Arias et al. 2019). Even though their assessment of Fulton's condition factor did not establish evidence for a reduced body condition of MP-feeding individuals, the HSI was significantly higher in fish collected at one sampling site which also was positively correlated with a higher MP ingestion rate in individuals from this specific site.

An enzymatic approach to assess the effect of MP ingestion was chosen by Alomar et al. (2017): the researchers analysed the antioxidant and detoxification system of fish by measuring different enzymatic activities in samples of the liver of MP-feeding and non-feeding red mullet *Mullus surmuletus*. The authors did not find signs of oxidative stress or cellular damage in the fish liver of MP-feeding fish. Nevertheless, they assume an induction of the detoxification system of the liver based on their findings of a small increase in the activity of GST (glutathione *S*-transferase).

These partly contrasting results highlight the challenges inherent to *in-situ* studies and impact assessments based on snapshot samplings: to effectively evaluate the impact of MP ingestion on the condition of a fish – either assessed via morphometric indices or by investigations on molecular, cellular, or organ level – long-term studies under controlled laboratory conditions across all life stages and trophic guilds are necessary (Morgana et al. 2018). Another major obstacle in examining the effect of MP ingestion based on in-situ studies is the ascertainment of a baseline for comparison, which hypothetically would involve fish growing up in an MP-free environment with no possibility of ingesting MP particles at any given point in time (Markic et al. 2018). Studies comparing the condition of MP feeding individuals with MP non-feeding ones face the uncertainty of former MP-uptake (and consequent egestion) by the latter. Thus, the informative value of these comparisons may be partially limited due to potential effects originating from previous feeding incidences in the now detected MP non-feeding fish. Nevertheless, future research should be strongly encouraged to focus on impact assessments based on field investigations, by continuing to calculate adequate condition factors with reference to the data available. As substantiated by Ryan (2016), the physical impacts of MP ingestion could include injuries of the GIT (i.e., ruptures, abrasions, lesions), thus, the examination of potential damages of the GIT prior to content analysis should be done by default. Both measures are comparably cost-efficient in terms of labour as well as financial input and could potentially contribute towards an enhanced understanding of MP uptake effects, considering the current underrepresentation of such studies and the lack of conclusive results. Another more labour-intensive still financially reasonable approach may involve the reading of otolith microstructures as an indicator for fish condition. The measurements of growth increment widths could offer insights into environmental pressures experienced by the fish with a higher accuracy than body dimensions (Suthers et al.

1992), even though a definite identification of MP exposure or uptake as the causative environmental pressure may prove difficult. Beyond, it appears expedient to further advance the analysis of the ratio of RNA:DNA as a proxy for fish condition in *in-situ* investigations (Vasconcelos et al. 2009), as well as the examination of inflammatory responses or enzymatic activities in response to MP exposure and ingestion. In summary, it is worth mentioning that the proxies for fish condition frequently applied in ichthyology (i.e., somatic growth, otolith microstructure, and RNA:DNA ratio) are known to heterogeneously reflect recent growth history patterns (Peck et al. 2015), and that this varying degree of suitability in detecting responses to MP exposure and ingestion in fish should be envisioned prior to identifying research objectives.

3.4.2 GIT content: natural prey items and MP

Besides the exclusive verification of MP in the GIT of the fish, a limited number of studies included reports on natural prey items (e.g.. Gassel et al. 2013; Chagnon et al. 2018; Halstead et al. 2018), both qualitatively (i.e., identification of prey items) and quantitatively (e.g., volume or weight of natural prey items in comparison to MP particles). This lack of information in the majority of studies (79 of 90) may be predominantly due to the analytical method applied, which often includes the digestion of organic material as an initial step in MP-GIT-studies.

Accounting for the fact that the in-depth analysis of prey items in fish GIT may be time consuming and potentially susceptible to misidentification, the assessment of stomach fullness may be considered a feasible approach for many studies under time-constraints. Stomach fullness, i.e., the presence of food in the stomach or GIT, could represent a relevant factor in predicting the presence of ingested plastic. In their study on Atlantic cod *Gadus morhua*, Atlantic salmon *Salmo salar* and capelin *Mallotus villosus*, Liboiron et al. (2019) found varying levels of food presence (i.e., stomach fullness) across the three taxa under investigation which was linked to the probability of MP ingestion. A comparable observation was made by Alomar and Deudero (2017) who found a significantly positive correlation between the stomach fullness of *Galeus melastomus* and the amount of MP (particles g_{FW}^{-1}) in the stomach of these catsharks. In contrast to these findings, Wieczorek et al. (2018) did not detect a significant difference between the median stomach fullness of MP-feeding fish and MP-non-feeding fish in their study of seven fish species from North Atlantic waters, indicating no correlation between stomach fullness and MP ingestion.

Nevertheless, analysing the entire GIT content should be considered beneficial, as it could yield essential information on the importance of MP as targeted or accidentally ingested prey items. In dietary studies, the relative importance of a specific prey item is frequently inferred from the proportion of all GITs included in the study containing each prey item (Baker et al. 2014). This leads to the questions, concerning why MP as a 'prey item' is treated significantly different from natural prey items and how outcomes of MP ingestion studies should be interpreted and evaluated thoroughly without this essential information. By including the analysis of the entire GIT content, researchers will be able to deduce information on the feeding preferences of the fish population or species under investigation – insights that could be subsequently used to further investigate patterns of MP ingestion. This approach was chosen for instance in a study on six different fish species collected off the Texas Gulf coast (Peters et al. 2017): based on their assessment of prey items in the GIT of the fish, the authors were able to statistically correlate MP uptake with the ingestion of specific prey items in each fish species (e.g., shellfish, crabs, vegetation), enabling the researchers to identify patterns in MP ingestion related to foraging modes. Peters et al. (2017), thus, found a tendency for higher MP ingestion related to a generalist foraging mode and methods of prey capture in comparison to selective (invertebrate) foraging, as displayed by grunt (pigfish) *Orthopristis chrysoptera* in their study.

The assessment of the entire GIT content may imply other valuable insights into modes of MP ingestion in fish, such as trophic transfer and bioaccumulation which are currently rather neglected by the most of the scientific literature (Wright et al. 2013; Lusher et al. 2017b; Chagnon et al. 2018). Upon chemically or enzymatically digesting the entire GIT content for MP analysis, information about feeding preferences are lost, which could be relevant for investigating the probability of trophic transfer. Obviously, the size relation between the prey or predator and the MP particles and fibres represents an essential constraint, as the prey organisms need to be capable of ingesting MP particles and fibres >1 µm in size to fall into the category of MP. Only few studies included in this review investigated the potential existence of MP transfer from prey to predator organisms. In a study on different fish species (i.e., predators) and shrimp (i.e., potential prey), McGoran et al. (2018) found higher MP ingestion rates in pelagic fish and flatfish species (MP-feeding fish: 36%) than in the invertebrate taxon (MP-feeding shrimp: 6%). This observation was considered to indicate bioaccumulation of MP from prey to predator. In a study by Chagnon et al. (2018), MP were found in flying fish (*Cheilopogon rapanouiensis*) preyed upon by yellowfin tuna (*Thunnus albacares*), thus, the authors argued that MP may be transferred from prey to predator. Rios-Fuster et al. (2019)

found undigested *Sardina pilchardus* in the GIT of *Trachurus mediterraneus*, and upon analysing the GIT of the prey fish, anthropogenic particles were found, supporting the hypothesis of trophic transfer in fish. The extent to which the secondary ingestion of MP contributes towards bioaccumulation in fish remains to be critically evaluated on the basis of more in-depth studies on a variety of fish taxa, especially as many of the higher trophic species are consumed by humans. Particular consideration should be devoted, however, to the potential capability of fish taxa to egest small-sized particles so that MP may not necessarily accumulate inside the GIT over time (Foekema et al. 2013; Chagnon et al. 2018).

3.4.3 Environmental samples

Establishing a link between the content analysis of the fish GIT and the environment in which the fish were sampled is complex; the majority of *in-situ* studies (73/90) did not include water, plankton or benthos samples as a reference for MP abundance. In the studies which did so, however, the resemblance between the environmental MP and the ingested MP varied greatly. Concerning the abundance of MP in environmental samples and in fish GIT, Lusher et al. (2016) were not able to detect any correlation between the MP numbers in subsurface waters and the amount of MP ingested by fish in the same area. A similar observation was made by Collicutt et al. (2019) comparing the GIT MP concentrations in juvenile chinook salmon to environmental (water and sediment) MP concentrations. In contrast to this, Güven et al. (2017) detected a different pattern in fish from Turkish territorial waters: individuals with higher MP loads in their GIT originated from sampling sites with higher MP particles abundances, thus mirroring the local conditions in terms of MP pollution.

Concerning the colour and particle types (i.e., fragments or fibres) of MP detected in the environment and also in the fish GIT, the outcomes of the few studies that took environmental samples into account seem to be equally divergent. Ory et al. (2017) for example found a significantly different representation of the most common coloured MP particles ingested by *Decapterus muroadsi* in comparison to the representation of coloured MP detected most frequently in water samples. Specifically, the researchers verified a selective feeding for blue MP consistent with a higher prey selectivity for transparent-blue copepods in the diet of the fish, both more commonly ingested than expected based on their relative representation in the surrounding environment. Consistent with the observation of this selective feeding, the damselfish *Pomacentrus moluccensis* was found to ingest white or transparent fibres, potentially reflecting their natural prey items, with a higher frequency than this observed in the corresponding surface water samples collected in the habitat of the fish (Jensen et al. 2019).

In contrast, other studies clearly demonstrated the mirroring of the assemblage of MP sampled from surface waters or the water column and the MP assemblage found in the fish GIT, thus, indicating non-selective or random feeding. In mesopelagic fish species from the North-western Atlantic, for instance, no evidence for a colour-selective feeding was established, as the MP detected in the alimentary tract were similar to those sampled from surface waters (Wieczorek et al. 2018). Supporting this observation, fish larvae from the western English Channel ingested predominantly blue fibres, corresponding to the particle category most represented in the water column (Steer et al. 2017). In a time series analysis of MP in plankton samples and fish from the Baltic Sea, the GIT content of herring (*Clupea harengus*) and sprat (*Sprattus sprattus*) also mirrored the composition and abundance of MP in the water column, both in terms of MP types (i.e., fibres, fragments) and size classes of MP over the years (Beer et al. 2018). Juvenile chinook salmon *Oncorhynchus tshawytscha* were also reported to non-selectively ingest MP of various colours and types (i.e., fibres) with reference to water and sediment samples assessed from the respective sampling sites (Collicutt et al. 2019).

The inconsistent findings and apparent mismatches between ambient samples and fish GIT both in terms of MP abundance and MP categories and colours may be explained on the one hand by the mobile and often migrating behaviour of fish on horizontal and vertical scales (Lusher et al. 2016), and on the other by the spatial and temporal heterogeneous distribution of MP in the marine environment (Barnes et al. 2009; Goldstein et al. 2013; Antunes et al. 2018). Moreover, fish species showing specific feeding preferences with regards to sub-surface or benthic prey items may, further, distort the interpretation of results due to varying probabilities of MP encounter and ingestion across certain local, regional, or even seasonal scales (Lusher et al. 2016; Silva et al. 2018b). Thus, considerations of the ecology of the study organism as well as the hydrography of the sampling site are essential to holistically evaluate the correlation between MP in fish GIT and in the ambient environment. As laboratory experiments verified harmful effects of adsorbed toxins (Rochman et al. 2013), the collected MP from environmental samples should be considered for analysis of ambient toxins as well as harmful microorganisms adsorbed or for chemical additives inherent to the particles; the outcomes of such assessments may shed light on the potential implications of MP ingestion for fish.

3.4.4 Environmental parameters

Environmental parameters may have significant impacts on the physiological and ecological performance of fish. Related to the uptake of prey and, thus, the potential presence of MP particles in the GIT of a fish, the following abiotic factors may be considered relevant: oxygen availability determines, for instance, the swimming and feeding activity (Fry 1971), temperature is known to impact the feeding and the egestion rates (Pandian and Vivekanandan 1985; Fänge and Grove 1979; Beer et al. 2018), light intensity has been shown to also affect activity levels, swimming speed, and visual capacity relevant for near-field location and predation success (Downing and Litvak 2000; Trippel and Neil 2003), salinity may shape fish assemblage distribution and with that the probability of MP exposure (e.g., within estuaries; Blaber and Blaber 1980). Furthermore, current velocity or tidal currents could also influence not only the activity, distribution, and feeding behaviour of fish (Stoner 2004), but also the distribution of MP particles and fibres in the ambient waters (Kanhai et al. 2017; Welden and Lusher 2017; Zhang 2017). Concerning the distribution and abundance of MP, rainfall (i.e., seasonality) may be also considered a relevant factor especially in estuarine-based studies, as increased precipitation leads to higher MP abundance caused by increased river discharges (Lima et al. 2014) which in return also affects levels of salinity or temperature in the estuarine environment. All of the above highlight the importance of recording abiotic variables along with prey/MP availability in *in-situ* MP ingestion studies. However, only a minor part of the studies investigated here (4/90) explicitly reported on any environmental factors (i.e., Ferreira et al. 2016a, 2018; Vendel et al. 2017; Silva et al. 2018b).

In studies based on sampling campaigns during the dry and rainy season in the Goiana estuary, Brazil, a spatio-temporal pattern of filament ingestion was revealed for different life stages of *Cynoscion acoupa* (Ferreira et al. 2016a, 2018). According to the authors, the highest ingestion of filaments was recorded for adult individuals during the rainy season; the increased availability of filaments during this time of the year was hypothesized to be related to the peak in fishing activities acting as the origin of the filaments. From the same estuary, a spatio-temporal pattern of MP ingestion was also reported for different life stages of *Pomadasys ramosus* and *Haemulopsis corvinaeformis* (Silva et al. 2018b). All of the three studies recorded a variety of different environmental parameters, including salinity, dissolved oxygen, Secchi depth, and rainfall rates obtained from local meteorological stations. A significant impact of any of the assessed environmental parameters on the MP uptake of the fish species studied was, however, neither investigated nor substantiated in either of the studies. The same holds true for a comparative study performed during rainy and dry season along the salinity gradient in two other Brazilian estuaries subjected to varying levels of anthropogenic pressures (Vendel et al. 2017): the authors did not relate MP uptake to the seasonality, the salinity gradient assessed, or any other environmental factor. Notwithstanding the above, it may be expected, however, that a larger number of studies has information on abiotic data available for correlation analyses, especially those studies conducted on board research cruises as part of continuous monitoring campaigns (e.g., Bellas et al. 2016; Hermsen et al. 2017; Beer et al. 2018; Pellini et al. 2018b). As the metabolic demands and, thus, the feeding and egestion rates vary not only with life stage and species but also with prevalent abiotic factors, past, present, and future studies should (re-)evaluate the presence of prey or MP particles in the fish GIT with primary emphasis on temperature and oxygen, and secondary with regards to light intensity, salinity, current velocity, and seasonality.

3.5 Publication Biases

Within the previously mentioned broad category of publication biases, different sub-categories were suggested for further differentiation (Fig. 7), of which several of the delineated above may be recognized in MP research on fish on the basis of this review.

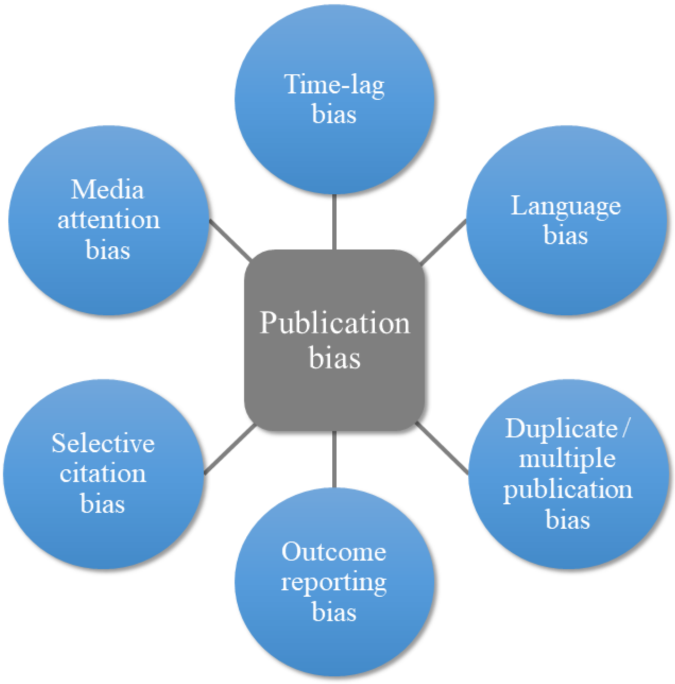


Fig. 7 Different types of publication bias (Ekmekci 2017).

3.5.1 Time-lag bias

According to Song et al. (2013), studies reporting significant or major results may be prone to more timely publishing than those stating insignificant outcomes. Taking into account the time-consuming procedures associated with alimentary tract analysis and MP particle identification (e.g., Fourier-transform infrared spectroscopy (FTIR) or Pyrolysis gas-chromatography mass-spectrometry (Py-GC/MS)) used in many studies, and consequently the small amount of samples that can be processed in a short period of time (Käppler et al. 2016, 2018; Silva et al. 2018a), a time lag between the sampling campaign and the consequent publication is inevitable, irrespective of the potential outcomes. In cases where only month and year were given, the last day of the respective month was chosen to calculate the time lag between sampling campaign and publication date online. Moreover, publications which indicated only a season (e.g., spring) as a reference for the sampling campaign were excluded for this analysis. As the entire submission and publication process (including the precise dates) was not uniformly trackable for all publications under review and due to the varying reporting schemes for MP feeding across fish taxa, the number of studies considered for the time lag analyses varies. Based on the accessible information for 67 of the 90 studies under review, 28.21 ± 18.92 months is the average time between the actual sampling campaign and the online publication of the study. Looking at the submission and publication time lag separately, the average time between the last month of sampling and the submission of the manuscript was 24.43 ± 19.34 months ($n = 58$), whereas after the submission, the review and publication process took on average 4.12 ± 2.26 months ($n = 57$). Studies reporting an uptake rate of <10% ($n = 16$) across all individuals investigated took 23.13 ± 12.27 months to be published online, whereas studies with 10 - 100% MP ingestion ($n = 48$) across individuals were published within 27.19 ± 15.51 months after the last month in the field. There was no correlation between the proportion of MP feeding individuals in a study and the total time lag between the last month of field work and the online publication (data not normally distributed, Spearman rank correlation coefficient $n = 64$, $R = -0.013$, $\boldsymbol{P} = 0.92$). As opposed to the hypothesis by Song et al. (2013), according to which studies with major results might be published faster, no significant correlation was found between the proportion of MP feeding individuals and the time lag between submission and publication (Spearman rank correlation coefficient $n = 55$, $R = -0.019$, $\boldsymbol{P} = 0.89$). Furthermore, no significant correlation was found between the number of fish taxa included and the total time lag between field work and publication (Spearman rank correlation coefficient $n = 67$, $R = -0.15$, $\boldsymbol{P} = 0.22$) nor between the number of individuals assessed and the total time lag (Spearman rank correlation coefficient n = 66, R = 0.062, ***P*** = 0.62). The existence of a time-lag bias may, thus, be considered incidental. Given the overall low number of insignificant, low, or negative reports in terms of MP-feeding currently published, the existence of this bias should be, nonetheless, re-evaluated in the future.

3.5.2 Language bias

The language in which a report is written may pose a significant obstacle to the thorough and holistic evaluation of the current state of knowledge in a specific research area (Ekmekci 2017). The literature reviewed here was exclusively written in English and published in peer-reviewed journals, in which the publication-language is English. Reasons for this may be seen in the database inquiry itself (Mongeon and Paul-Hus 2016), which was based on English keywords (*plastic* and *fish*), as well as on the fact, that most peer-reviewed, high impact journals in natural sciences are published in English (Meneghini and Packer 2007; Mongeon and Paul-Hus 2016). The limitation of a review to English-language literature only may lead to adding a certain tendency – or bias – to the outcome of the review. Beyond the actual language in which a report is published, the framing of titles, abstracts or results using certain phrases or terms may also contribute to a distorted perception of MP ingestion across fish taxa (Sect. 3.5.4). From the perspective of scientific ethics, the impact of a language bias in the field of biological/natural sciences may be valued differently in comparison with the other biases discussed hereinafter. Notwithstanding the above, researchers, editors, and reviewers should be equally encouraged to pay special attention to the compliance and usage of neutral language in scientific reports. Moreover, researchers may always consider the publication of results for a non-scientific audience in a transparent, easily understandable, and potentially local language.

Besides the actual publication language, the areas under investigation may also add to a certain language bias: considering the geographic areas under investigation by the studies reviewed here (Fig. 8), certain regions such as the Mediterranean, the Baltic and/or North Sea as well as the Western Atlantic seem to be represented to a greater extent than others, i.e., the Southern Ocean, the Persian Gulf or the Arctic Ocean, are all represented by a single study only (Abbasi et al. 2018; Cannon et al. 2016; Kühn et al. 2018). Other regions, such as the Red Sea or the Indian Ocean are not included at all. It cannot be excluded, that references for the abovementioned areas and beyond do exist and have simply not been assessed by this literature review and data base inquiry. Nonetheless, collating, mapping, and evaluating the knowledge on MP distribution and extent of exposure and ingestion may be substantially impaired by the heterogeneous distribution of geographical areas examined as well as by the existence of unsampled spots on the world ocean map. International collaboration between researchers and institutes should, therefore, promote investigations in areas currently underrepresented to contribute to a holistic overview on the global extent of MP distribution and the potential impact of ingestion.

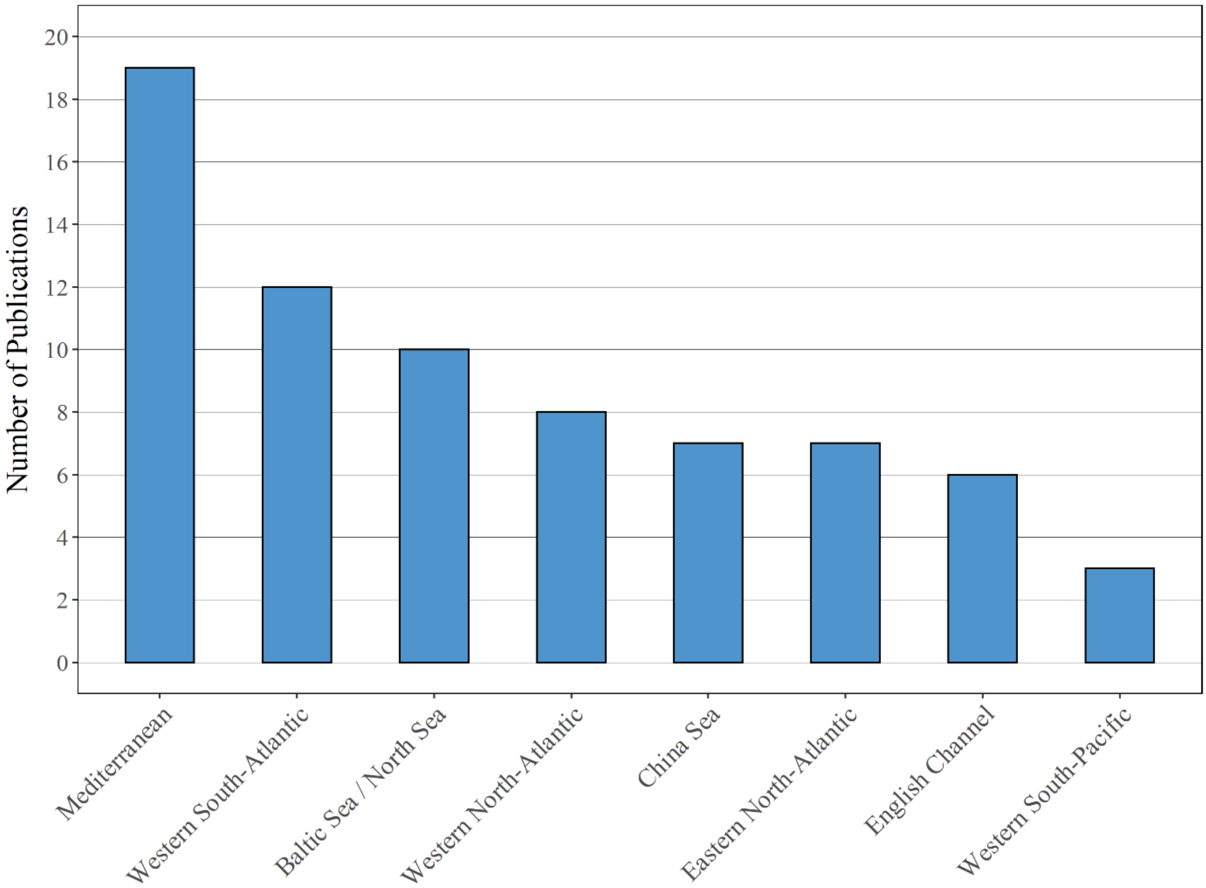


Fig. 8 Representation of geographic regions which were covered by at least three or more studies included in this review. Studies from the Baltic and the North Sea were combined for this graphical illustration.

3.5.3 Duplicate / multiple publication bias

Publishing the same research or data set twice, with substantial analogies in both manuscripts in relation to the underlying scientific theories, methodologies, outcomes, or conclusions, may lead to an erroneous perception of the significance of results both among the scientific community and potentially also among civil society (Ekmekci 2017). In many cases, the same researchers authored similar manuscripts, cross-referencing to both publications happens infrequently, and duplicate publication of results goes beyond abbreviated reports for the media or for a narrow audience, or communicating outcomes in different languages (Johnson 2006). Even though brief abstracts in conference proceedings are considered acceptable (COPE - Committee on Publication Ethics 2003), the admissibility of publishing the entire study in a conference proceeding may be questioned when later on published as a stand-alone study in another journal. Upon going through the results of the data base inquiry for this review, the following example stood out: Pellini et al. (2018a) published their study on MP ingestion by common soles in the Adriatic Sea within the *Proceedings of the International Conference on Microplastic Pollution in the Mediterranean Sea.* In Pellini et al. (2018b), MP in the gastrointestinal tract of common soles from the Adriatic Sea was published in the journal *Environmental Pollution*. Cross-checking both publications revealed a considerable duplication of the area under investigation, the number of individuals examined as well as the results obtained and discussed. Yet, these almost identical studies were listed separately by the means of the data base research, and a cross-reference to each other was not found. Comparably, Bessa et al. (2018a) published a brief version of their study on MP uptake in three commercially important fish species (namely *Dicentrarchus labrax*, *Diplodus vulgaris* and *Platichthys flesus*), likewise, within the *Proceedings of the International Conference on Microplastic Pollution in the Mediterranean Sea*, with the extended version reporting on the same species, sample sizes and results, however, is published separately as Bessa et al. (2018b) in the journal *Marine Pollution Bulletin*. In another case, papers by Ferreira et al. (2016a) and Ferreira et al. (2018) on acoupa weakfish appear to report on the same data set for a publication on dietary habits (incl. MP) and a more detailed study exclusively on MP ingestion. Even though the objectives of both publications are slightly different from each other, it seems that almost the identical data set of biological samples was used for two reports on MP ingestion by this fish species. These examples obtained by a single data base inquiry exemplify how the significance of MP ingestion by fish might be overemphasized through repetitive publications. Besides the mere increase in published literature on the topic, the waste of resources and ethical implications for the authors, editors, and journals involved, the repetitive release of fundamentally the same information may lead to the misperception of environmental pollution. This, in turn, could ultimately lead to the loss of scientific credibility, which might affect future research initiatives, funding availability, and policy making.

3.5.4 Outcome reporting bias

The publication of findings from scientific investigations with multiple outcomes is considered biased if ‘positive’ outcomes have a higher probability to be included in the report than ‘negative’ ones (Song et al. 2013) – this selective editing of results is also called ‘within-study publication bias”. The following examples were identified from the results of the literature review conducted here: in a study by Steer et al. (2017), the authors only mention those fish species explicitly for which MP uptake was found. In comparison to a total of 23 different species investigated, MP-feeding was reported for five species, which were given in the report by scientific names and abundances investigated. Information on the remaining 18 non-MP feeding species cannot be accessed on the basis of the actual publication. Upon personal request, however, Steer et al. (2017) provided original data on the investigated species for this review. Vendel et al. (2017) also reported on only a sub-set (n = 27) of the total number of species investigated (n = 69), explaining this selection by the inadequate sampling sizes for the other species (i.e., below 10 individuals per species) which could potentially impair statistical analysis. A record of the remaining 42 species with minor abundances is not included in the report. In a study by McGoran et al. (2018), the authors also do not mention the non-feeding species in the publication text. However, details on these species are accessible via the table included in the accompanying supplemental material. Taking not only recent studies into consideration, it becomes obvious that the existence of this outcome reporting bias dates back almost five decades: Carpenter et al. (1972) explicitly mentioned only those species with common names, scientific names and abundances that were found with MP spheres in their gastrointestinal tract.

Another instance of outcome reporting bias is the nonpublication of entire studies if a specific outcome could not be inferred from the data analysed, i.e., no MP fibres or particles present in the GIT of a fish species examined (Liboiron et al. 2018). A conceivable additional source of bias for MP ingestion studies might be the ‘relabelling’ of a MP-uptake analysis as a dietary analysis of a fish species, which was not found (but initially suspected) to ingest MP particles. The consequent lack of explicit specification of absence of MP in the GIT by focussing on reporting of natural prey items may further distort the perception of MP exposure and ingestion by fish.

3.5.5 Selective citation bias

According to Leimu and Koricheva (2005), several different factors may contribute to a selective citation of ecological papers: apart from the journal's impact factor, the number and consortium of researchers and institutions involved (including their nationalities), results supportive of a widely accepted hypothesis are cited more often. Following Ekmekci's (2017) reasoning, studies with statistically significant and 'positive' results are cited more frequently than others. Identifying this bias on the basis of the literature reviewed here is hardly possible due to the lack of reports on minor or none uptake of MP in fish (Liboiron et al. 2018). However, due to the high potential of the existence of an outcome reporting bias (see above), the existence of a consequent selective citation bias seems plausible. By assessing the citation history of all articles included in this review (conducted on the 6th September 2019), the potential existence of a selective citation bias (Fig. 9) was investigated.

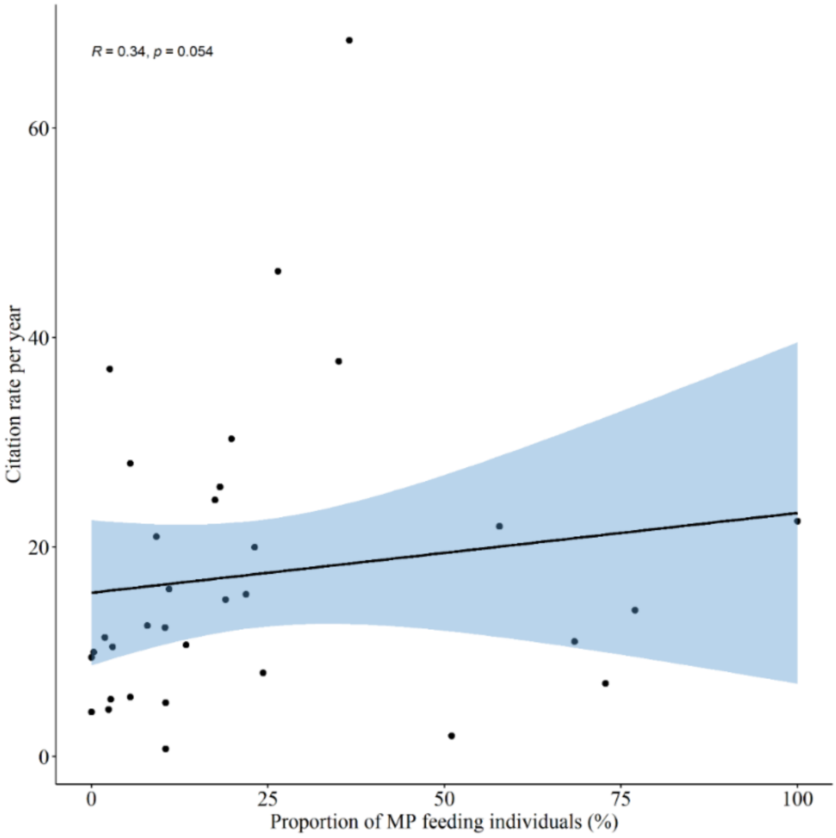


Fig. 9 Scatter plot showing the relationship between the citation rate per year and the proportion of MP feeding fish (%) for a subset of studies (n = 33) included in this review. Spearman's rank correlation coefficient (R) was computed, along with the corresponding significance level (***P***-value) of the correlation. Light-blue shaded area represents the 95% confidence interval.

The number of citations in the year 2019 were excluded, moreover, the respective first year in which a paper was published was not considered either. By this, a mean number of citations per year was computed. If the paper was published in 2017, citations for 2017 and 2019 were not taken into consideration as the calculation of a citation rate per year would have been then based on one year only (2018). Therefore, these references were also excluded. Due to the high number of papers published within the past two years, the citation per year was, thus, calculated for a subset of 33 manuscripts. Figure 9 shows the computed correlation based on Spearman rank correlation coefficient for not normally distributed data. The correlation coefficient between the citation rate per year and the proportion of MP-feeding individuals shows a marginal trend towards a significant positive relationship (R = 0.3379; ***P*** = 0.054), yet with the minority of data within the 95% confidence interval. Based on this analysis, the existence of a selective citation bias was not verified beyond doubt, although given the slight tendencies detected for a subset of papers included in this review, the potential occurrence of this bias may not be completely ruled out.

3.5.6 Media attention bias

Online and print media play an important role in distributing information on global environmental concerns such as marine (micro-)plastic pollution, impinging public awareness and political actions alike (SAPEA 2019). The increase in scientific and media coverage of the topic of MP litter is indubitable (Lusher 2015; Phuong et al. 2016; de Sá et al. 2018; Liboiron et al. 2018; Cunningham and Sigwart 2019). Irrespective of the fact that the actual implications of MP presence in the oceans have not been clarified beyond doubt, public perception of marine MP appears to be rather unequivocal. A recent study by Völker et al. (2019) confirmed the media framing of the topic by showing that the minority of scientific studies (24%) actually considers the risk of MP as evidenced beyond doubt, whereas the majority of media coverage (93%) seems to take the existence of detrimental effects of MP for granted. Derived from this, a media attention bias and consequent framing can be ascertained for both the topic of marine MP in general and for MP ingestion by marine biota in particular.

4 Conclusion and recommendations

4.1 Focus of future studies

In the 90 studies under review here, approximately one third of the individual fish assessed were found with MP particles or fibres in their GIT, with an average plastic load of around 2 particles. Yet, comparing the outcomes of different *in-situ* MP uptake investigations in marine and estuarine fish is challenging due to a lack of harmonized methodologies and standardized protocols as well as to the fact that only few species are examined in sufficient sample sizes. Furthermore, MP identification was frequently conducted exclusively by visual means. Individual ecological parameters (e.g., body dimensions, trophic level, feeding guild, preferred habitat) do not seem to explain the frequency of MP-feeding or the variation of MP ingestion rates beyond doubt. Thus, scientific efforts should go beyond the commonly applied categories to further investigate determining factors for MP ingestion, bearing in mind the following guidelines for *in-situ* studies on MP ingestion by fish (Fig. 10).

On the basis of the present review and the accompanying Tables 1 and 1OS, the following fundamentals for conducting *in-situ* assessments of MP uptake in marine and estuarine fish are suggested:

Selection of relevant fish taxa und life stages

Based on the outcomes of this review, a number of fish species and families can be identified relevant for future research initiatives, as either these taxa have not been studied yet or have been found to vary in terms of MP uptake across different studies and geographic regions. Moreover, ELHS (especially of commercially important fish taxa) should be targeted by prospective scientific efforts as they are hypothesized to show a high vulnerability to the ingestion and impact of MP. To significantly contribute towards an enhanced understanding of MP pollution as a threat for marine and estuarine fish, adequate sample sizes (i.e., >20 individuals per taxon) need to be taken and analysed – prioritisation should be given to single- or ‘few-species-‘studies with sufficient sample sizes over multi-species assessments with less than ten individuals per taxon.

Publication of field data

To ensure comparability of studies and a sound evaluation of the outcomes, essential details of the sampling-strategy and fieldwork need to be recorded and shared with the scientific community as well as civil society. Key information on the precise sampling date, time, location, and depth are just as relevant as specifications about the sampling gear applied (e.g., mesh sizes). Furthermore, the assessment of a potential methodological bias, e.g., stemming from consequent laboratory procedures, is greatly enhanced if details on the sampling and processing protocols are shared.

Investigation of the gastrointestinal tract

As demonstrated by this review, future research efforts should take an essential step towards the integrated evaluation of the hazardous potential of MP ingestion by analysing the entire content of the GIT and, thus, quantitatively and qualitatively compare the uptake of natural prey items to MP fragments and fibres. Thereby, also the importance of trophic transfer of MP along the food chain could be further elucidated.

Contextualization of MP ingestion

To identify potential MP uptake selectivity across fish taxa, feeding strategies, or life stages, environmental samples (e.g., water, plankton, or benthos collections) should be taken in the respective habitat or at the specific fish-sampling site and, consequently, compared to the outcomes of the GIT analysis. By integrating ambient samples into the assessment of MP uptake by fish, in-depth knowledge is gained on the distribution and abundance of MP in different compartments which may contribute also to an enhanced understanding of the vulnerability of individual taxa or life stages depending on specific habitats or prey items.

Correlation of MP exposure and ingestion with abiotic parameters

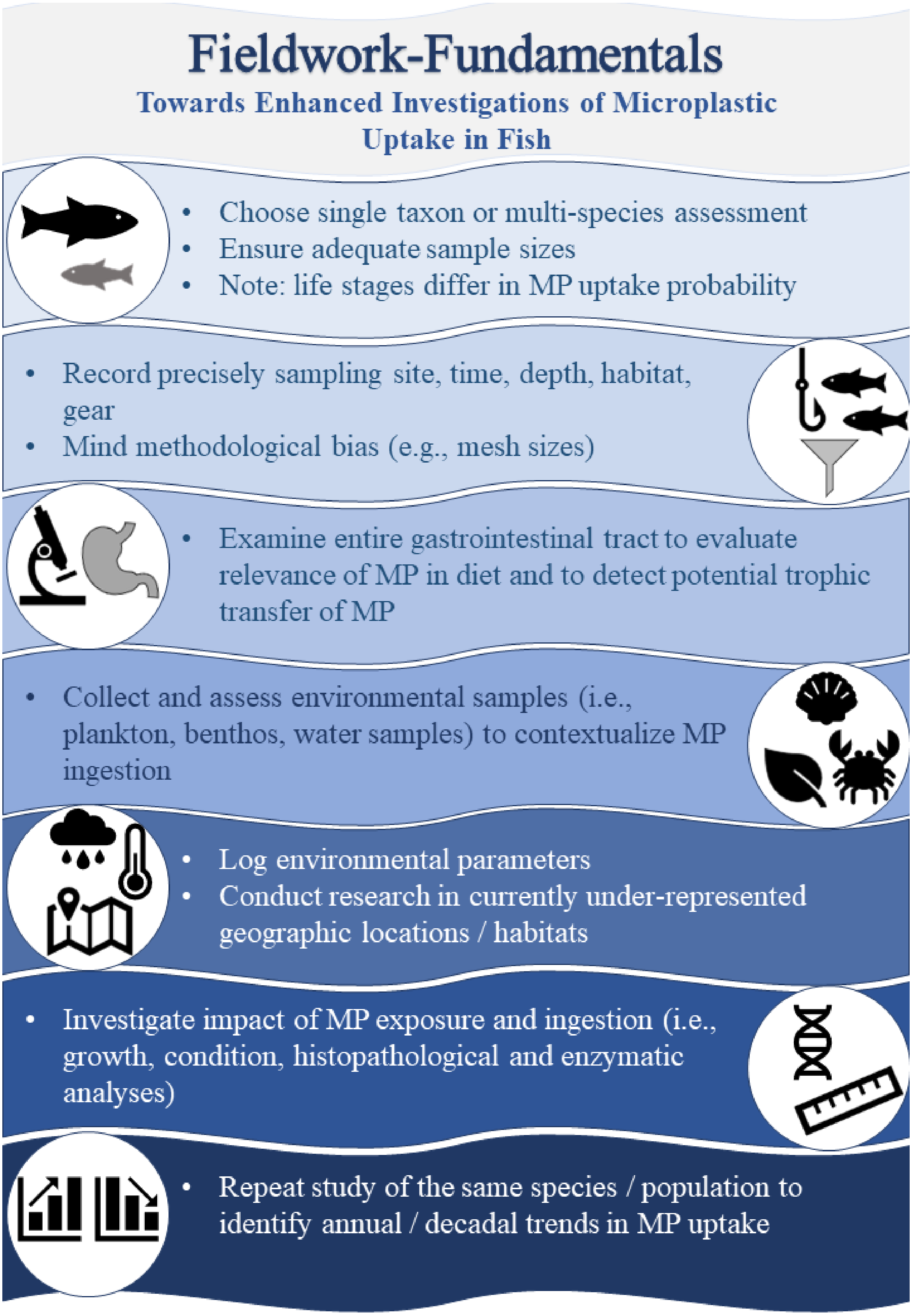
As environmental parameters and local conditions may significantly affect the physiological and ecological performance of a fish and consequently also the ingestion (as well as egestion) of MP particles and fibres, future investigations should aim at detecting links between the uptake of MP and abiotic environmental parameters such as temperature, oxygen, light intensity, or local weather conditions. To comprehensively analyse the horizontal and vertical pathways of MP distribution and the resulting implications for complex food-webs, more research is needed in currently under-represented geographic areas as well as in ecosystems which exhibit specific hydrographic and oceanographic properties (e.g., coastal habitats, marine upwelling ecosystems).

Investigation of MP-uptake related physiological and ecological effects

To further unravel the underlying drivers for and impacts of MP ingestion across marine and estuarine fish, the precise recording of morphological/anatomical features is considered essential (e.g., standard length, height, eviscerated weight, mouth gape). The assessment of fish condition and potential differences in growth between MP-feeding and MP-non-feeding individuals may be considered expedient in determining the impact of MP uptake on a species- or population-specific scale. Enzymatic, toxicological as well as histopathological approaches should be also considered by prospective research initiatives to holistically evaluate the physiological as well as ecological effects of MP ingestion across fish species and families.

Verification of spatio-temporal trends in MP exposure and uptake

An extension of short-term investigations of MP uptake towards long-term monitoring initiatives should be implemented by future scientific investigations. Therefore, repetitive sampling campaigns in the same area on key indicator species will assist in understanding the progression of marine plastic litter as a potential threat over the years to come and to evaluate the effectiveness of applied ecosystem conservation and litter reduction measures. In this respect, a *de novo* analysis of already existing samples collected by annual (or at least continuous) monitoring campaigns may be advisable with special focus on MP detection to clarify previous trends in MP abundances and distribution.



Fieldwork-Fundamentals

Towards Enhanced Investigations of Microplastic Uptake in Fish

- Choose single taxon or multi-species assessment
- Ensure adequate sample sizes
- Note: life stages differ in MP uptake probability

- Record precisely sampling site, time, depth, habitat, gear
- Mind methodological bias (e.g., mesh sizes)

- Examine entire gastrointestinal tract to evaluate relevance of MP in diet and to detect potential trophic transfer of MP

- Collect and assess environmental samples (i.e., plankton, benthos, water samples) to contextualize MP ingestion

- Log environmental parameters
- Conduct research in currently under-represented geographic locations / habitats

- Investigate impact of MP exposure and ingestion (i.e., growth, condition, histopathological and enzymatic analyses)

- Repeat study of the same species / population to identify annual / decadal trends in MP uptake

Fig. 10 Recommendations of essential details future *in-situ* studies on MP ingestion by fish to assure effective contributions towards an integrated assessment and holistic evaluation of the extent and impact of MP uptake.

4.2 Publications Biases

Irrespective of the reasons for why the non-feeding individuals/taxa are not included in the actual publication or for why studies reporting minor or zero uptake rates seem to be underrepresented, one or several biases may be attested for the reporting on MP uptake by fish, both in the past and in recent scientific investigations. With regards to the continuously growing research area, the incidence of these biases needs to be reduced to accurately inform both the scientific and public community about the extent and impact of MP ingestion on fish. Considering the high media coverage along with the easy comprehensibility and accessibility of the topic for a broad audience in comparison to other more complex environmental threats (e.g., ocean acidification), more attention should be devoted to critically examining and reporting about MP and its consequences to preserve the public's trust in scientific research. Scientists and editors should, thus, be encouraged to consider studies contradicting the common notion of an environmentally relevant topic for publication and may even contemplate a re-evaluation of scientific outcomes beyond the mere statistical significance. Moreover, both editors and authors should take steps towards preventing publication biases such as duplicate publication bias.

5 Acknowledgements

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Supplementary Materials

Other factors potentially influencing MP uptake

Microplastic ingestion appears to be common across different life stages and species in varying intensities, with body size, feeding modes, and preferred habitat individually not explaining the variation observed to a sufficient extent. Several studies suggested other factors that could potentially drive MP ingestion in marine fish such as the body shape, mouth/gape size, and swimming ability (Vendel et al. 2017) or behavioural traits, i.e., migrating behaviour (Romeo et al. 2015). Furthermore, assessing the following factors may be considered expedient in assessing relevant explanatory variables: life stage and associated shifts in diet of ELHS and adult fish (Silva et al. 2018b), sex (McGoran et al. 2018; Su et al. 2019b), and site of origin (i.e., distance to shore) rather than preferred habitat (Steer et al. 2017; Chan et al. 2019). As feeding modes, and, thus, preferred prey items may also vary among the same species, originating from different locations, species- and site-specific factors should also be taken into account (Silva et al. 2018b).

Impacts on Early Life-History Stages of Fish

During the period of switching from yolk-sac to exogenous feeding, the majority of marine fish larvae are considered to be planktivorous, while more diverse feeding patterns (including species-specific prey preferences) are observed during the juvenile life stage (Nunn et al. 2012). Deriving from this, early life-history stages (ELHS) may be particularly susceptible towards microplastic (MP) ingestion in relation to their feeding mode during the larval stage along with species-specific feeding habits at later ontogenetic phases. Knowledge about the feeding ecology of the respective taxa under investigation is, therefore, considered essential when evaluating the extent to which ELHS are vulnerable to MP uptake and potential detrimental effects.

As the impact of ingestion is likely to differ, depending on the size relation of the organism affected and the particles ingested (Lusher 2015), ELHS of fish may be considered particularly vulnerable to the impact of MP-sized particles (Mazurais et al. 2015). Digestive injuries and starvation may be among the conceivable consequences for ELHS ingesting large amounts of MP (Ferreira et al. 2016a). Studies assessing the impact on early life-history stages of fish should, thus, pay special attention to signs of malnutrition in MP-feeding fish. Regarding the potentially increased risk of mortality caused by MP uptake, Hoss and Settle (1990) came to the conclusion that the overall ingestion of inert MP by ELHS presumably did not contribute towards an increased mortality, yet nowadays, the extent to which MP ingestion adds to mortality in these vital life stages still remains to be understood. Even though these first in-situ reports of MP ingestion included ELHS of fish (Carpenter et al. 1972; Kartar et al. 1976; Hoss and Settle 1990), information on the impacts of ingestion remain insufficient, with field and laboratory studies commonly focusing on adult fish or giving no indication concerning the life stage investigated. To precisely appraise the vulnerability of ELHS to MP pollution, future studies should, therefore, pay attention to recording the actual size and/or life stage of the organisms under examination. Moreover, the immediate as well as the long-term effects of MP ingestion are yet to be assessed. Due to the special vulnerability of ELHS, priority consideration should be given to in-depth studies on larval and juvenile stages both in field and laboratory studies.

Condition indices based on fish morphometrics

Based on the assumption that heavier fish of a given length are in better shape or condition than equally-sized individuals of lighter weight, fishery ecologists frequently use external measures of fish size (e.g., standard length, height, body mass) and the thereof computed ratios as a proxy for individual or population fitness and well-being (Hayes and Shonkwiler 2001). The main advantages of morphometric-derived condition indices are the rapid and non-destructive collection of the required data along with its simplicity in roughly estimating the well-being of a fish. Apparent limitations may be caused by the various approaches to determine length (standard length, fork length, total length) or weight (wet weight or eviscerated wet weight), impeding comparisons across different studies and the apparent loss of information by reducing multi-dimensional relationships into a single factor (Cone 1989).

At the beginning of the 20th Century, Fulton proposed a condition factor *K,* which assumes isometric growth (i.e., growth with constant body proportions), based on the following formula:

$$K = \frac{weight}{length^3}$$

with wet weight in grams and length in centimetre (Fulton 1904).

Until today, Fulton's K is still one of the most frequently used indices (Nash et al. 2006). However, allometric growth is observed in most fish taxa (Le Cren 1951), which requires a species-specific adjustment of the slope of the length-weight relationship (LWR), as proposed by Ricker (1975) in the relative condition factor *K*', an extension of Fulton's *K*:

$$K' = \frac{weight}{length^b}$$

with wet weight in grams, length in centimeter and *b* as a species-specific constant.

To estimate the LWR of a fish (and, thus, the species-specific constant), the following linear regression of the log-transformed equation is used:

$$\log(Weight) = \log(a) + b\log(Length)$$

with the parameter a representing the intercept and the parameter *b* the slope of the relationship.

Derived from these two basic condition formulas, researchers have been applying various condition factors based on morphometrics, such as the Richardson condition factor used in two of the studies under review:

$$CF = 100\ \times\ \left(\frac{weight}{(length)^b}\right),$$

Other factors have been proposed which take into account not only the length and weight of a fish, but also its height (Jones et al. 1999). Within a given fish taxon, however, the relationship between body length and weight may vary considerably, depending on various factors such as the fish population itself (i.e., sex, maturation, or life stage), or on the annual variability of environmental conditions (Froese 2006). The usage of the relative condition factor after Le Cren (1951) may be, thus, advisable as it compensates for the variation in condition in relation to increasing body lengths:

$$K_{rel} = \frac{observed\ weight}{theoretically\ estimated\ weight}$$

Here, the observed wet weight (in grams) of an individual is compared to the calculated weight for that respective length in a given population (based on the calculation of the LWR).

A comprehensive overview on the different condition indices based on morphometrics, their strengths and potential limitations can be found, inter alia, in Bolger and Connolly (1989), Richter et al. (2000), and Froese (2006).

Chapter 2

"It is a curious situation that the sea, from which life first arose should now be threatened by the activities of one form of that life. But the sea, though changed in a sinister way, will continue to exist – the threat is rather to life itself."

Rachel Carson (1961): The Sea Around Us

Growing up in a plastic ocean: variability of prey preferences, microplastic exposure and uptake by juvenile white seabream in a coastal lagoon nursery ground

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Disclaimer: According to the guidelines and revision of the journal to which the manuscript was submitted, the content and structure of Chapter 2 may have changed in the process of publication of the present thesis.

Abstract

Marine plastic litter, originating from land-based sources, enters the marine environment by passing through coastal ecosystems such as lagoons and estuaries. As early life-history stages (ELHS) of many commercially important fish species rely on these transitional areas as nursery grounds, it is hypothesized that they encounter a spatial gradient of habitat quality and pollution from inner to outer parts of their vital environment. With sizes < 5mm, microplastic (MP) fibres and fragments entail a high bioavailability for ELHS of fish, potentially facilitating MP uptake at early developmental stages which may have implications for their survival and growth. This study provides a contextualisation baseline between feeding preferences and MP uptake by the white seabream *Diplodus sargus* (Linnaeus, 1758) in an estuarine nursery ground on the southern coast of Portugal. Juvenile fish showed a generalised, omnivorous feeding mode with slight differences in trophic resource utilization between individuals collected at distinct seagrass meadows in the lagoon. A total of 23.13 % of the fish were detected with MP in the gastrointestinal tract, the mean number of MP per MP-feeding individual was 1.64 ± 1.04, with plastic fibres occurring more frequently than particles. We verified no significant impact on juvenile fish condition and neither fish standard length nor wet weight were related to MP ingestion. The uptake of MP fibres was correlated with the ingestion of marine plants as well as with the overall fullness of the gastrointestinal tract, yet no correlation between MP ingestion and sampling site was established although results indicate the existence of a spatial gradient of plastic pollution. Knowledge of the underlying factors for MP ingestion will be greatly enhanced by considering environmental conditions along with species- and life-stage specific feeding modes and prey preferences which shape the uptake probability of artificial fibres and fragments.

Keywords: Sparidae, microplastic, ingestion, feeding mode, stomach content, estuarine nursery

1 Introduction

Coastal lagoons and estuaries are recognised as highly productive transitional ecosystems providing vital habitats and ecosystem services considered biologically, societally, and economically relevant (Costanza et al. 1997; Beck et al. 2001). Their importance as nursery grounds for many commercially valuable fish species arises from the combination of high structural complexity (comprised for instance of seagrass meadows) and particular favourable environmental conditions (Beck et al. 2001; Elliott and Hemingway 2002; Amara and Paul 2003; McLusky and Elliott 2004; Seitz et al. 2014). Irrespective of their acknowledged relevance, coastal ecosystems face severe exposure to habitat degradation (Kennish 2002; Lotze et al. 2006) and pollution as a result of the continuously increasing urbanization of on- and offshore regions (Browne et al. 2011) along with the riverine input of plastic particles from land-based sources (Lechner et al. 2014; Morritt et al. 2014). Within the past decade, microplastic (MP) fragments and fibres of < 5mm in size became the centre of scientific and public interest: arising from the cumulative industrial application of plastic materials and the lack of efficient waste-management (Jambeck et al. 2015; Ryan 2015), increasing quantities of MP have been documented in coastal areas around the world (Barnes et al. 2009; Cole et al. 2011; Kumar et al. 2021). Due to their size range, MP particles are available for ingestion (and potential trophic transfer) for a variety of organisms at the base of the marine food web, among them early life-history stages (ELHS) of fish (Cole et al. 2013; Gove et al. 2019). Growth, condition, and survival of ELHS is strongly shaped by gradients in abiotic and biotic conditions in vital nursery grounds, with direct consequences for recruitment success to adult fish stocks (Boehlert and Mundy 1988; van der Veer et al. 1990; Beverton and Iles 1992; Ciotti et al. 2014). Therefore, in-depth research on the potential uptake and consequent physiological impact of MP is needed to holistically assess the underlying factors contributing to recruitment variability in commercially important fish species. Although plastic ingestion in fish has been reported across more than 140 families (Azevedo-Santos et al. 2019), the majority of field studies neither assessed ELHS of commercially important fish taxa nor investigated prey selectivity by comparing diet composition with prey availability (Gamito et al. 2003; Selleslagh and Amara 2015; Markic et al. 2020; Müller 2021). Feeding strategy and prey preferences are known to undergo ontogenetic changes (Galarowicz et al. 2006; Sánchez-Hernández et al. 2019) and understanding of the feeding ecology of a life stage or species is considered of major importance to establish and improve sustainable management and conservation (Braga et al. 2012).

Our study therefore aims to contribute to the filling of existing knowledge gaps in relation to the potential uptake and effects of MP for growth and survival of ELHS of a commercially important fish species by complementing the analysis of the gastrointestinal tract of the fish with the assessment of zooplanktic and benthic prey availability. Juvenile white seabream, *Diplodus sargus* (Linnaeus, 1758), were chosen as model organisms for this field study. ELHS supposedly show a higher ingestion probability and sensitivity than adults (Critchell and Hoogenboom 2018; Salerno et al. 2021), and the omnivorous feeding mode of the white seabream has been hypothesized to be an influencing factor for elevated MP uptake rates (Mizraji et al. 2017; Garcia et al. 2020; Shabaka et al. 2020), yet this species has been shown to be able to discriminate between natural and artificial prey items in a feeding experiment (Müller et al. 2020), challenging the abovementioned notions. Based on the hypotheses that the gross of microplastic enters the marine environment through coastal ecosystems (Browne et al. 2011; Wright et al. 2013; Lechner et al. 2014; Auta et al. 2017) and seagrass habitats act as a sink for MP (Cozzolino et al. 2020; Jones et al. 2020; De los Santos et al. 2021), which are vital nursery grounds for a wide range of fish ELHS, including the white seabream, our research questions are:

i) Do juvenile *Diplodus sargus* encounter a gradient of habitat quality and MP pollution in their vital coastal lagoon nursery area?

ii) What are the driving factors for and the potential detrimental effects of MP ingestion for ELHS of an omnivorous fish species?

2 Materials and Methods

2.1 Study Area

The field study was conducted in the western part of the Ria Formosa lagoon, which stretches across 55 km along the southern Portuguese coast (Fig. 1) and has a maximal width of 6 km. Its widely ramified creeks and subtidal channels, which are on average less than 3 m deep, are connected to the Atlantic Ocean by six inlets which facilitate a continuous exchange of up to 75% of the water volume during each tidal cycle (Águas 1986). During spring tides, the maximum tidal amplitude can reach 3.5 m, resulting in an exposure of approximately 1/3 of the entire lagoon area (Ribeiro et al. 2006). Except for surface waters and salt pans, the salinity varies between 35.5 and 36.9 all year round (Falcão et al. 1992) and the water temperature ranges between 12°C in winter and up to 27°C in summer (Newton and Mudge 2003).

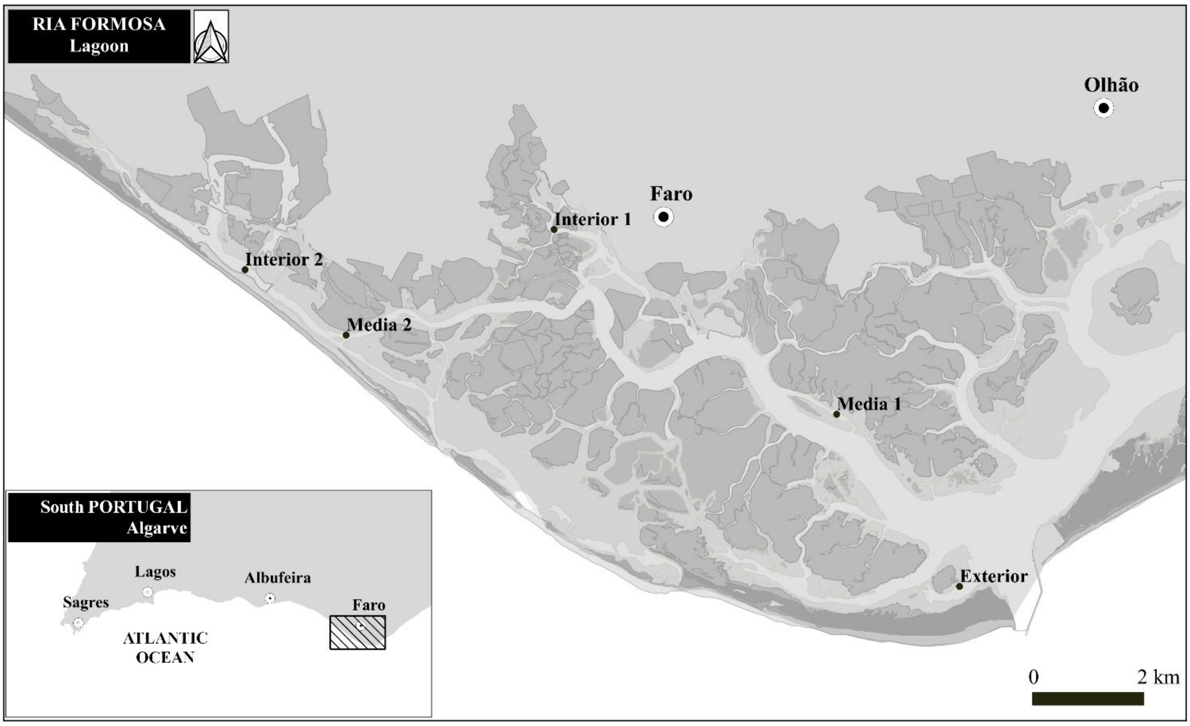

Fig. 1 Map of mainland Portugal, highlighting the location of the Ria Formosa lagoon at the Algarve coast. Enlarged map of the western part of the lagoon displays the five distinct sampling sites selected for this study and two inlets.

The lagoon serves as an important habitat for many occasional, migratory, and resident fish species, with the highest ichthyofaunal diversity and abundance recorded during summer in shallow, less turbulent waters over fine, muddy sediments vegetated by marine plants (Erzini et al. 2002). The Ria Formosa is recognised as an essential nursery for the adjacent coastal fish populations, fostering significant populations of juvenile fish, including commercially relevant members of the seabream family (Sparidae) (Monteiro et al. 1987, 1990; Monteiro 1989; Erzini et al. 2002; Ribeiro et al. 2006, 2012). Despite the acknowledged ecological and economic importance, the ecosystem is facing persistent anthropogenic pressures (e.g., habitat degradation, impaired water quality) across a spatio-temporal gradient, with higher detrimental effects being exerted on biological communities in the interior, urbanized parts of the lagoon during summer months (Newton et al. 2003; Cravo et al. 2012; Guimarães et al. 2012).

2.2 Field Sampling Design

To investigate potential spatial abiotic and biotic gradients as well as differences in MP abundance, five distinct seagrass-vegetated samplings sites were selected for this comparative study (Fig. 1): two sampling sites were chosen for their proximity to urbanized areas, namely the city centre of Faro and the Praia de Faro (thereinafter called ‘Interior 1’ and ‘Interior 2’), two additional sampling sites were selected in intermediate distance to the urbanized areas of Faro (‘Media 1’ and ‘Media 2’). One more sampling site was designated close to the inlet of the main channel to the Atlantic Ocean (‘Exterior’); as the only other western Ria Formosa inlet yielded no seagrass-vegetated sites to be sampled for comparison, only one station at the greatest distance to the city was considered appropriate. The distance between the most interior site (Interior 2) and the Exterior site is approximately 13 km. The field work design included three sampling campaigns in summer 2018: the first sampling was realised in the beginning of July (04. / 05.07.2018), the second one took place end of July (27.07.2018) and the final sampling was conducted mid-August (10. / 13.08.2018). All samplings took place during slack side (max. two hours prior and after low tide); at each site, different physico-chemical parameters (i.e., temperature, salinity, dissolved oxygen) were recorded using a YSI Professional Plus Multiparameter Instrument (YSI Pro2030), and sampling of juvenile seabream, zooplankton and benthos was conducted.

2.2.1 Field Sample Collection

Zooplankton & Macrozoobenthos

Zooplankton was collected using a conical net (200 µm mesh size, 0.13 m^2 mouth opening, equipped with a HydroBios flowmeter to precisely assess the sampled water volume) which was towed just below the sea surface over approximately 300 m behind the boat around the sampling site; GPS waypoints with a Garmin GPS device were taken to facilitate the recording of the actual distance. The samples were preserved in glass jars using a seawater-ethanol solution (70% ethanol) until further analysis. Macrozoobenthos was taken with a push net (1000 µm mesh size, rectangular opening of 20 cm height and 50 cm width), operated manually and pushed over a distance of 10 m through the seagrass meadow. The samples were stored deep-frozen in individual zip-lock bags until further analysis. Both zooplankton and macrozoobenthos samples were collected during the first and third sampling campaign.

Seabream

Juvenile seabream were collected during all three sampling campaigns using a beach seine (25 m width, 3.5 m height, 9 mm stretched mesh size) with a cod end type sac in the middle to facilitate handling of the catch (Erzini et al. 2002). The net was deployed from a 6.5 m boat and towed over a distance of 20 m along the shore by the boat from one side and by two to three people on the shore from the other side. After half of the towing distance, the boat headed to the shore, the net was hauled in and closed, allowing to sample an area of approximately 1087 m^2. The catch, which was retained in the cod end, was then emptied into a box and sorted for seabreams. Individuals that were gilled in the mesh of the seine net were also collected. Juvenile seabream were stored in labelled plastic bags inside a cooler and transported to the laboratory where the fish were preserved in a freezer at CCMAR, Faro, Portugal, until further processing and analyses.

2.3 Laboratory Processing

2.3.1 Contamination control

Potential contamination of the samples was reduced by applying the following procedural measures (Lusher et al. 2017b) in the laboratories of CCMAR in Faro, Portugal, and ZMT in Bremen, Germany: work benches and equipment (i.e., glassware, tweezers, needles and Bogorov chambers) were cleaned with ethanol prior to use, the latter being additionally checked under the microscope for contamination. Contamination with fibres was minimized by washing hands and forearms thoroughly as well as by wearing cotton laboratory coats and nitrile gloves during all analytical steps performed. Care was taken also by wearing cotton clothing underneath the laboratory coat and the colour of the clothing was recorded to trace back potential contamination. If in doubt about the origin, fibres found in the samples were excluded from the analysis.

To avoid misidentification of MP, the criteria proposed by Hidalgo-Ruz et al. (2012), Lusher et al. (2017b, 2020) were adopted to the visual sorting of particles and fibres in the fish gastro-intestinal tract (GIT) and zooplankton samples – if in doubt about the origin of a fragmented or fibrous object, it was excluded from the analysis, thereby applying a conservative visual classification and quantification protocol.

2.3.2 Zooplankton & Macrozoobenthos

Based on the principle of taxonomic sufficiency (TS) (Ellis 1985; Ferraro and Cole 1990), prey organisms were mainly classified to genus- or family-level; the abundance (individuals m^{-3}) was calculated for each zooplanktic and benthic prey taxon from the neuston and push net samples.

Zooplankton

Zooplankton samples were transported to the laboratory facilities of ZMT, Bremen, for further analyses. In the laboratory, samples were split into 1/4 – 1/128 fractions (depending on the amount of zooplanktic organisms collected) using a modified Motoda plankton sub-sampler. Zooplankton images with a resolution of 2400 dpi were taken from the respective subsamples with a ZooScan (ZooScan Model V4, Hydroptic Inc., France), following the procedures described by Gorsky et al. (2010). The scans obtained were analysed using the software ZooProcess on the ImageJ macro language (Gorsky et al. 2010), allowing automated processing and measuring of the scanned images. ZooProcess associates the images with the available metadata and divides the scanned images into multiple single images ideally depicting a single organism only. Images containing multiple or overlapping organisms and particles were manually edited using the software and subsequently processed again. For identification, all images were uploaded to the website EcoTaxa (http://ecotaxa.obs-vlfr.fr/prj/) where Random Forest Algorithm in combination with convolutional neural network feature extraction automatically classifies them; afterwards the preliminary classification was manually validated.

Macrozoobenthos

The entire zip-lock bag containing the sampling material was placed into a sorting tray and defrosted using tap water. Upon defrosting, one or several trays were filled with either the entire sample or several subsamples (depending on the amount of material collected) to facilitate handling. In an initial step, seagrass shoots, algae, and larger prey items (e.g., broken mollusc shells, larger crustaceans) were separated from the rest of the sample and excluded from the analysis. All remaining prey organisms, as well as artificial items, were manually sorted, identified, and counted.

2.3.3 Seabream

2.3.3.1 Fish Growth and Condition

In the laboratory of CCMAR, fish were thawed at room temperature before examination. Individuals of the species *Diplodus sargus* were identified by visually inspecting distinctive external characteristics (i.e., pigmentation and dentition), measured to the nearest mm (standard length = SL, total length = TL, height = H) and weighed before and after dissection (ww).

Body condition was evaluated for all individuals by calculating the Fulton's condition factor (Fulton 1904); as only a subset of fish was chosen for gastro-intestinal tract (GIT) analysis, Fulton's K was calculated using the total wet weight instead of the eviscerated wet weight as done in Müller et al. (2020) on the same species:

$$K = \left(\frac{wet\ weight}{standard\ length^3}\right) \times 100$$

2.3.3.2 Analysis of gastro-intestinal tract (GIT)

Upon dissection, the proportional fullness of the GIT of the fish was visually assessed and ranked in five classes, i.e., 0 (empty GIT), 1 (< 10% fullness), 2 (< 50% fullness), 3 (> 50% fullness), 4 (> 90% fullness), 5 (100% fullness). The wet weight of the entire GIT was measured to the nearest mg, afterwards, the entire GIT was preserved in 70 % ethanol and stored in Eppendorf tubes until further analysis in the laboratory facilities at ZMT in Bremen. Upon content analysis, the GIT was put in a Bogorov counting chamber (filled with 70% ethanol) and opened with fine scissors and tweezers under a stereo-microscope (Zeiss Stemi 2000-C). The content was visually inspected for both artificial (i.e., MP fibres and particles) and natural prey items, the latter were identified to higher taxonomic levels following the TS approach. Fibres detected in the GIT were carefully inspected for vegetal morphological features such as organic structures or segmentation to avoid misidentification with artificial fibres. Furthermore, only fibres were counted that were found attached to GIT content remains; free-floating fibres in the Bogorov counting chamber as well as fibres matching the clothing underneath the cotton lab-coat were also excluded. Though this conservative procedure could result in partial underestimation of plastic ingestion, the potential bias caused by airborne contamination is reduced to a minimum to ensure reliable results. The GIT analysis time was not standardised for this study due to the variability of GIT content and volume; to enhance comparability of results, a fixed 20 x magnification was chosen and maintained throughout all analysis.

Percentage frequency of occurrence (%FO) was chosen to be an appropriate, robust measure to analyse the GIT content of fish (Baker et al. 2014); it was calculated according to the following formula:

$$\%FO = \frac{number\ of\ GIT\ containing\ prey\ item\ i}{total\ number\ of\ GIT\ containing\ prey} \times 100$$

Moreover, prey-specific abundance (%P) was calculated for all countable faunal prey items:

$$\%P = \frac{abundance\ of\ prey\ item\ i}{total\ number\ of\ prey\ items\ in\ all\ GIT\ containing\ prey\ item\ i} \times 100$$

The analyses of the feeding strategy, prey importance, and inter- as well as intra-individual components of niche width, was based on the two-dimensional plot (Fig. 2) of prey-specific abundance %P (y-axis) and frequency of occurrence %FO (x-axis), following the approach suggested by Amundsen et al. (1996) as a modification of the Costello Method (Costello 1990). The distribution of points along the axes and diagonals of the diagram provide the following information: specialised or generalized feeding strategy (vertical axis), rare or dominant prey items (diagonal from lower left to upper right corner), high between-phenotype or within-phenotype contribution to the niche width (diagonal from upper left to lower right corner).

To contextualise MP uptake with environmental parameters, morphometrics, fish condition and other prey items, the data obtained for the MP-feeding individuals were analysed for potential correlation by computing the Spearman rank correlation coefficient (r_s) where values can vary between -1 (indicating a strong negative correlation of the variables compared), 0 (indicating no association between the variables compared), and +1 (indicating a strong positive correlation of the variables compared). Level of significance was set to $\boldsymbol{P} \leq 0.05$.

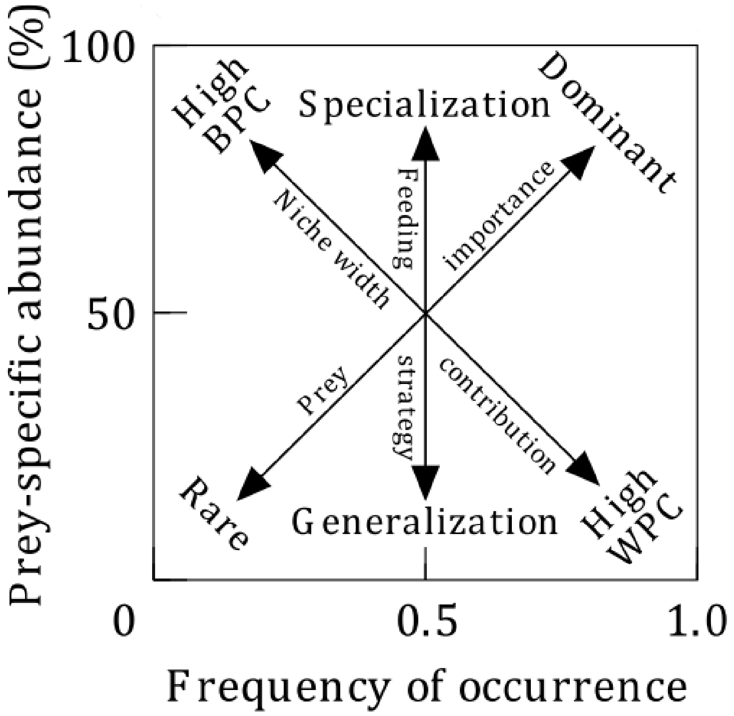

Fig. 2 Modified Costello plot (after Amundsen et al. 1996), in which the prey-specific abundance %P is plotted against the frequency of occurrence %FO. BPC = Between-phenotype component; WPC = Within-phenotype component.

2.4 Statistical Analyses

Data were tested for normality of distribution (Shapiro-Wilk test) and homogeneity of variance (Fligner-Killeen test for not-normally distributed data). In case of violation of normality, the Kruskal-Wallis test was used to investigate statistical differences between the groups - if significant differences were detected, Dunn's test (with Holm correction) was computed to perform a pairwise comparison between the groups to identify which groups differ.

Prey preferences across the different stations were examined with non-metric multidimensional scaling (nomads) ordinations, using Bray-Curtis similarity coefficient on presence-absence data (R-package: 'vegan' by Oksanen et al. 2020). Analysis of similarity (ANOSIM) was computed to examine the significance of prey item groups in the ordination pattern (Clarke 1993). A subsequent indicator species analysis (R-package: 'indicspecies' by De Caceres and Legendre 2009) was performed to identify prey items that were found more frequently in the GIT of fish from one station compared to another. The same analyses were run to detect potential differences in prey availability across the different sampling sites and campaigns. Level of significance was set to $\boldsymbol{P} \leq 0.05$.

Statistical analyses and data visualization were realised with Microsoft 365 and R (version 4.0.5) (R Core Team 2020).

3 Results

3.1 Station description and habitat quality

The five stations chosen for this study were all vegetated by seagrass and marine algae. However, their bottom characteristics differed slightly: the station "Exterior" was characterised by coarse, sandy sediments whereas, with increasing distance to the Atlantic Ocean inlet, the sediment became muddier and finer. Slight differences were also detected in the physico-chemical characteristics of the five sites (Table 1).

Table 1 Environmental parameters and microplastic concentrations recorded at five different stations in the Ria Formosa lagoon, Portugal, during three campaigns in the summer of 2018. Na = not assessed; plankton and macrozoobenthos samples were collected only during the first and third campaign.

Station	**Latitude / Longitude**	**Sampling Campaign**	**Temperature (°C)**	**Salinity (PSU)**	**Oxygen (mgl^{-1})**	**Microplastic concentrations (particle m^{-3})**
Exterior	36.97205 / - 7.877817	1 2 3 **Mean ± SD**	19.8 19.8 19.8 **19.8 ± 0**	35.73 36.53 36.47 **36.24 ± 0.46**	8.56 5.46 5.72 **6.58 ± 1.72**	0.0 na 7.01
Media 1	36.991633 / -7.899833	1 2 3 **Mean ± SD**	21.0 20.8 19.8 **20.5 ± 0.6**	35.92 36.42 35.66 **36.0 ± 0.39**	7.57 6.3 6.2 **6.69 ± 0.76**	5.63 na 4.68
Media 2	37.003297 / -7.976471	1 2 3 **Mean ± SD**	21.9 23.6 22.9 **22.8 ± 0.85**	36.76 37.74 37.18 **37.23 ± 0.49**	7.4 6.53 7.8 **7.24 ± 0.65**	1.33 na 12.97
Interior 1	37.014883 / -7.947183	1 2 3 **Mean ± SD**	23.1 23.7 23.4 **23.4 ± 0.3**	35.76 37.21 37.65 **36.87 ± 0.99**	6.87 4.08 5.92 **5.62 ± 1.42**	1.51 na 4.62
Interior 2	37.011292 / -7.999077	1 2 3 **Mean ± SD**	23.4 24.2 24 **23.9 ± 0.4**	37.34 38.11 37.78 **37.74 ± 0.39**	7.18 6.48 5.63 **6.43 ± 0.78**	1.08 na 18.54

Temperature ranged between 19.8 – 24.2 °C across the different sampling sites and campaigns, showing a spatio-temporal gradient from inside the lagoon to the Atlantic Ocean inlet over the different campaigns, with a minor peak during the second campaign (end of July 2018). Salinity ranged between 35.66 – 38.11 PSU, showing comparable spatio-temporal fluctuations to those for temperature. Oxygen was generally higher at all stations during the first campaign as compared to the following two samplings and ranged between 4.08 – 8.56 mgl^{-1}, with the highest value recorded at the station closest to the Atlantic Ocean, and the lowest oxygen concentration measured at station “Interior 1”.

Following a conservative visual identification using the images produced by the ZooScan, the planktic plastic concentrations varied between 0.0 – 18.54 particles m^{-3}; the lowest concentration was measured at station “Exterior” during the first campaign (beginning July 2018), whereas the highest concentrations were associated with stations “Interior 2” and “Media 2” during the third sampling (Mid-August 2018). Station “Interior 1” had overall lower MP concentrations during both samplings than “Media 1”. During the third campaign, MP concentrations were higher at station “Exterior” in comparison to both “Interior 1” and “Media 1”. Using the ZooScan images, the most commonly identified plastic types in plankton samples were fibres and threads (Fig. 3 A – D), frequently being larger in dimensions than what is usually considered microplastic.

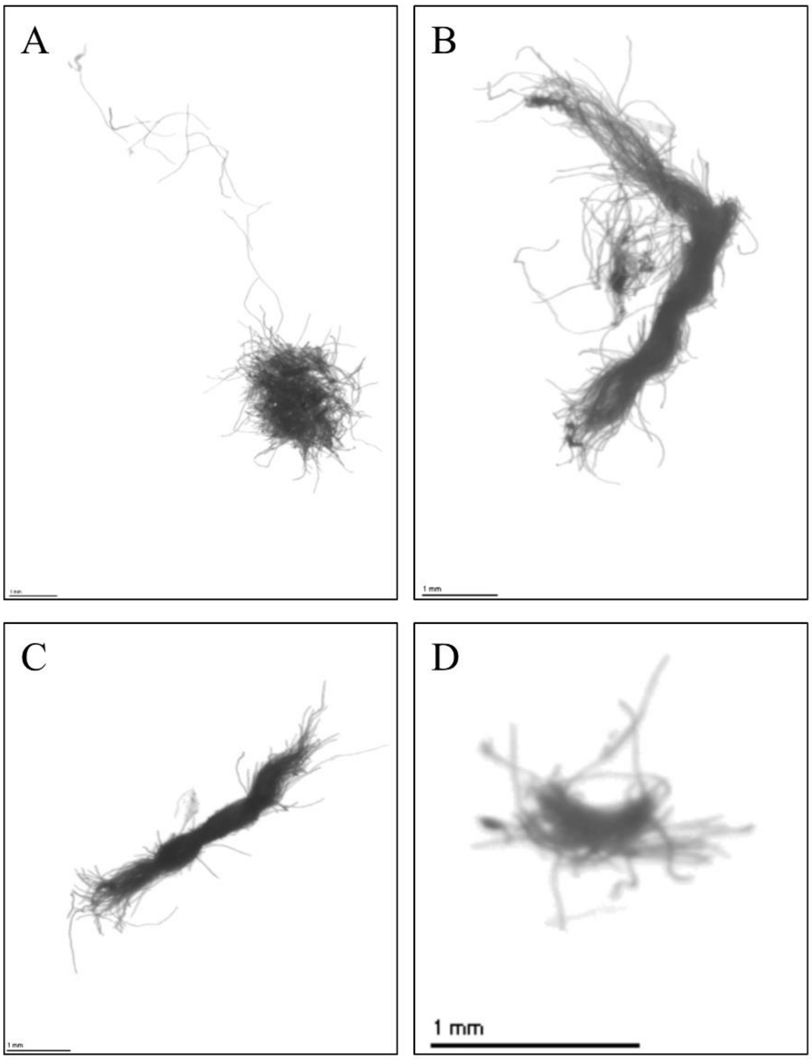

Fig. 3 A - D ZooScan images of artificial fibres and threads detected in plankton samples from the Ria Formosa lagoon, Portugal in summer 2018.

3.2 Fish Morphometrics

A total of 306 juvenile *D. sargus* were collected for this study in July and August 2018. A detailed overview of the abundances, key morphometric parameters and Fulton's condition factor of the juvenile fish collected at the different sites at three sampling campaigns is given in Figure 4 and in the supplementary materials, Table S1. The standard length (SL) ranged between 15 - 62 mm and total wet weight varied between 0.05 – 8.38 g. As only one juvenile *D. sargus* was collected at the station closest to the inlet to the Atlantic Ocean ("Exterior"), the analyses were performed including fish collected at the remaining four stations.

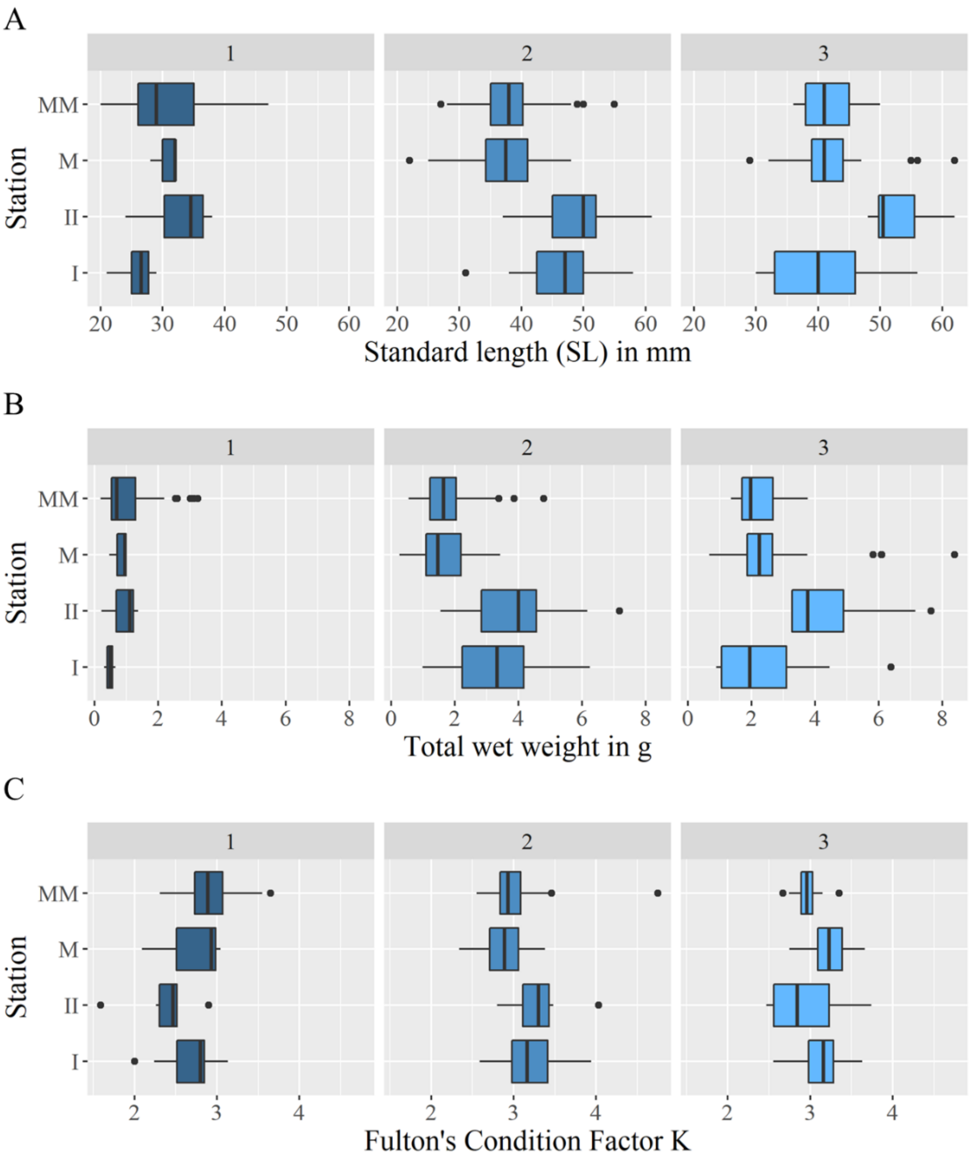

Fig. 4 A – C Comparative overview on key morphometrics, i.e., standard length (SL in mm), total wet weight (in g) and Fulton's Condition Factor K, recorded for 305 juvenile D. sargus at four distinct sampling sites (I = Interior 1; II = Interior 2; M = Media 1; MM = Media 2) over three sampling campaigns (1 – 3) in July and August 2018. Results for one juvenile fish, collected at the station "Exterior" are left out on purpose.

Regarding the morphometrics, juvenile *D. sargus* did not show any statistically significant differences in standard length or total wet weight during the first sampling campaign (Dunn's test, ***P*** > 0.05). In contrast, fish collected during the second campaign were significantly larger and heavier at both "Interior" stations in comparison to both "Media" stations (Dunn's test, ***P*** = 0.001 – 0.03). During the third campaign, no significant differences in standard length or total wet weight were detected. Comparably, Fulton's K showed no significant differences during the first campaign (Dunn's test, ***P*** = 0.34 – 1), however it was significantly higher during the second sampling in fish from "Interior 2" in comparison to fish collected at both "Media" stations (Dunn's test, ***P*** = 0.002 – 0.03). During the third campaign, seabream showed no statistically significant differences in Fulton's K across the different stations (Dunn's test, ***P*** = 0.3 – 1.0).

3.3 Dietary preferences of juvenile white seabream

3.3.1 Prey items

The GIT analysis of a subset of white seabream (n = 147) revealed a variety of different natural prey items, dominated by crustaceans, detritus, and marine flora (algae and seagrass), along with artificial items, namely MP fibres and particles (Table 2). Crustaceans were the most abundant prey item in terms of frequency of occurrence (94.56 %FO) and prey-specific abundance (90.45 %P). Within this prey taxon, noticeable differences between %FO and %P were recorded for copepods (70.75 %FO; 50.87 %N), Gnathiidae (49.66 %FO; 12.76 %P), as well as for decapod zoea stages (14.97 %FO; 85.7 %P). Comparable differences in the overall presence of a prey item in the diet and its relative abundance were also detected in other prey taxa, such as polychaetes, insects, and ascidian tadpoles (Table 2). While marine flora was present in more than 2/3 of the fish GIT, its %N was not computed as the amount of algae and plant material was numerically not quantifiable.

Table 2 Diet composition of juvenile *Diplodus sargus* (n = 147) collected in summer 2018 at five different sampling sites in the Ria Formosa lagoon, Portugal. * Not identified.

Prey Item			**Number of fish with prey$_i$**	**Frequency of occurrence % FO**	**Total abundance of prey$_i$**	**Prey-specific abundance (%P)**
Crustacea total			**139**	**94.56**	**6111**	**90.45**
	Copepoda total		**107**	**72.79**	**1968**	**48.72**
		Copepoda*	104	70.75	1783	50.87
		Harpacticoida	25	17.01	119	6.9
		Caligus spp.	9	6.12	66	29.46
	Amphipoda total		**20**	**13.61**	**55**	**9.05**
		Amphipoda*	17	11.56	39	19.8
		Gammaridae	5	3.40	5	1.23
		Caprellidae	4	2.72	11	26.83
	Isopoda total		**73**	**49.66**	**366**	**12.94**
		Isopoda*	1	0.68	1	3.57
		Gnathiidae	73	49.66	361	12.76
		Idoteidae	1	0.68	2	22.22
		Sphaeromatidae	1	0.68	2	4.55
	Tanaidacea	Tanaididae	**5**	**3.40**	**5**	**1.11**
	Decapoda	Zoea stages	**22**	**14.97**	**3452**	**85.7**
	Ostracoda		**34**	**23.13**	**63**	**2.35**
	Branchiopoda	Cladocera	**1**	**0.68**	**1**	**0.24**
	Maxillopoda	Cirripedia - cyprid larva	**50**	**34.01**	**186**	**10.49**
	Pantopoda		**1**	**0.68**	**1**	**0.83**
	Crustacea*		**11**	**7.48**	**14**	**1.2**
	Crustacea remains		**36**	**24.49**	**Nc**	**Nc**
Ascidiacea		tadpole	**15**	**10.20**	**90**	**8.91**
Actinopterygii			**5**	**3.40**	**5**	**6.49**
Polychaeta total			**23**	**15.65**	**19**	**11.31**
		Polychaeta*	11	7.48	14	11.48
		Polychaeta remains	13	8.84	Nc	Nc
		Polychaeta larva	1	0.68	2	22.22
		Serpulidae	1	0.68	1	2.70
		Glyceridae	1	0.68	2	7.14
Insecta total			**38**	**25.85**	**282**	**9.85**
		Insecta*	8	5.44	13	1.12
		Insecta remains	13	8.84	Nc	Nc
	Diptera total		**26**	**17.69**	**68**	**4.29**
		Chironomidae - larva	19	12.93	56	5.97
		Dolichopodidae - larva	7	4.76	8	3.65
		Meta-adult*	2	1.36	2	0.3
		Larva*	1	0.68	2	40.0

Prey Item			**Number of fish with prey$_i$**	**Frequency of occurrence % FO**	**Total abundance of prey$_i$**	**Prey-specific abundance (%P)**
	Hemiptera total		**11**	**7.48**	**201**	**8.16**
		Hemiptera*	4	2.72	13	1.75
		Auchenorrhyncha	11	7.48	188	7.63
Mollusca			**13**	**8.84**	**22**	**5.29**
		Mollusca remains	9	6.12	Nc	Nc
	Gastropoda					
		Planktic*	3	2.04	5	1.27
		Benthic*	2	1.36	2	9.52
	Bivalvia	Planktic*	1	0.68	1	5.58
Miscellaneous						
		Various*	28	19.05	185	15.14
		Sand grains	73	49.66	Nc	Nc
		Eggs*	5	3.40	Nc	Nc
		Detritus	125	85.03	Nc	Nc
		Alga 1*	109	70.07	Nc	Nc
		Alga 2*	77	52.38	Nc	Nc
		Seagrass*	20	13.61	Nc	Nc
		Fish scales	111	75.51	Nc	Nc
Plastic			**34**	**23.13**	**56**	**4.19**
		Fibre	30	20.41	47	7.31
		Particle	7	4.76	9	1.16

3.3.2 Feeding strategy and prey preferences

The feeding strategy and the importance of individual prey items in the diet of *D. sargus* are visualised in a modified Costello plot (Fig. 5) after Amundsen et al. (1996). As for feeding strategy (indicated by the solid line in Fig. 5), the majority of prey items ingested by *D. sargus* are represented in the lower part of the graph, hence suggesting a rather generalized feeding mode of the juvenile white seabream. Concerning the niche width contribution (dotted line), the location of prey items in the plot is an indicator for a population with a high between-phenotype component where the resource utilization varies between individuals. The location of the zoea stages in the upper left of the plot implies a specialization of certain individuals towards this prey taxon (high specific abundance, low occurrence), whereas copepods were ingested regularly by many individuals (intermediate specific abundance, high occurrence).

Most prey items (except for Copepoda) were ingested with minor frequencies and abundances (prey item points located in lower left corner of the plot, including MP fibres and particles) thus being of relatively low prey importance (dashed line). Though detritus and algae are not included in this representation due to their uncountable amount, they appeared with a high frequency of occurrence (52.38 – 85.03 %FO) as well as in high quantities, constituting an important share of the GIT fullness. Hence, these prey items would hypothetically be positioned towards the upper right part of the diagram. In summary, juvenile *D. sargus* are opportunistic feeders, taking up a variety of prey items with interindividual preferences for certain faunal taxa.

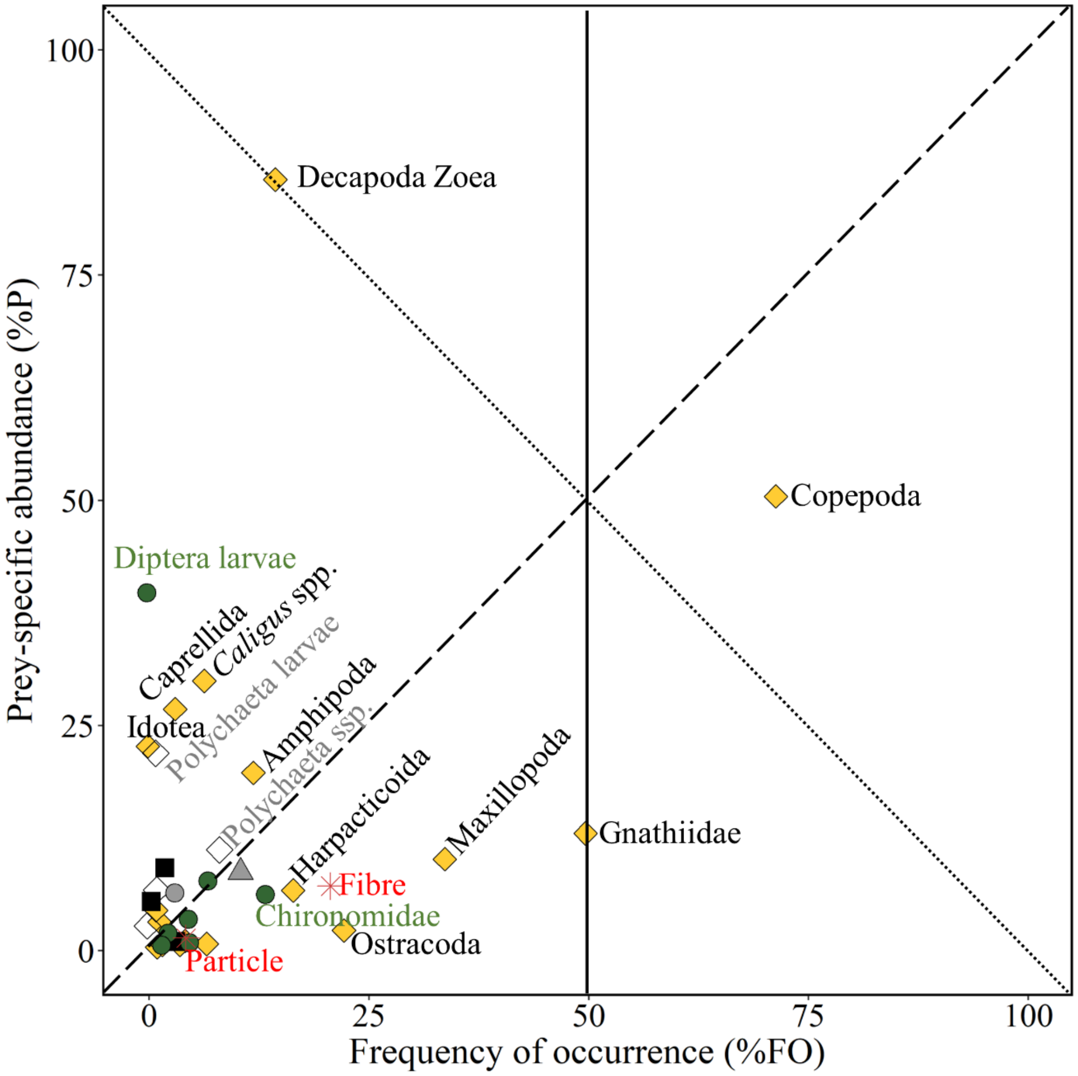

Fig. 5 Modified Costello plot, after Amundsen et al. (1996). Feeding strategy (solid line), niche width contribution (dotted line), and prey importance (dashed lined) of juvenile white seabream, D. sargus, are visualised based on the two-dimensional plot of prey-specific abundance (%P) and frequency of occurrence (%FO). Red asterisk = Plastic items; yellow rhombus = Crustacea; white rhombus = Polychaeta; green circle = Insecta; grey circle = Actinopterygii; grey triangle = Ascidiacea; black square = Mollusca.

To compare prey uptake across the five different stations, a non-metric multidimensional scaling (nMDS) was performed based on presence / absence data of identified prey items in the GIT of white seabream collected at five stations (Fig. 6). Although the ordination biplot shows major overlaps in prey item consumption across the different stations (stress = 0.184), weak yet significant differences in prey uptake were detected in relation to sampling site and campaign (ANOSIM R = 0.3806, ***P*** = 1 e-04).

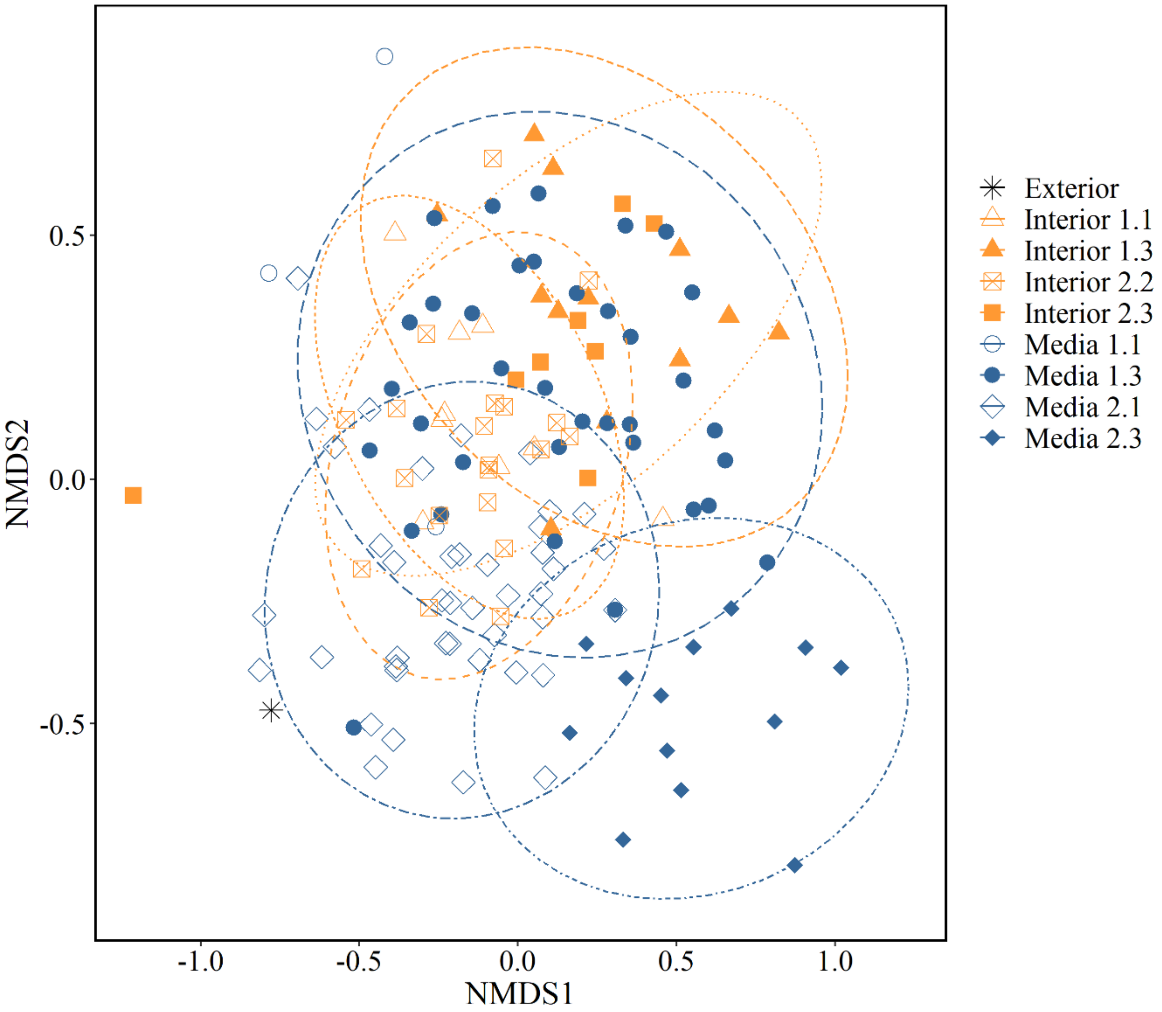

Fig. 6 Non-metric multidimensional scaling (nMDS) analysis ordination biplot based on Bray-Curtis coefficient of similarities between presence / absence data of prey items found in the GIT of juvenile white seabream D. sargus at five different sampling sites in the Ria Formosa lagoon (stress = 0.179, dimensions = 3, non-metric fit R^2 = 0.968, linear fit R^2 = 0.799; ellipses drawn based on 95% confidence interval where applicable). Station names, e.g., "Interior 1" are extended by the sampling campaign (i.e., 1, 2, 3).

According to the indicator species analysis, 8 out of 46 prey items were significantly associated with one or several groupings (*P* < 0.05, see supplemental materials for detailed results). During the first and second campaign, fish showed a preference for Gnathiidae (***P*** = 0.0478) across both Interior stations and at “Media 1”. For juvenile white seabream collected during the third campaign at the aforementioned stations, crustacean prey items were of significant importance (***P*** = 0.0037). Except for the first sampling in “Media 1” and the one individual collected at “Exterior”, algae were a key item across all sampling stations and campaigns in the diet of juvenile seabream (***P*** < 0.0016). At “Media 2”, *D. sargus* collected during the third campaign had a significant uptake tendency for Decapoda zoea stages (***P*** = 0.0001), and insect prey, particularly of the hemipteran order (***P*** = 0.0001 – 0.0279) which were of no importance at any other site or campaign, explaining the more distinct aggregation of this group in the nMDS plot (“Media 2.3”, blue rhombus, Fig. 6).

3.3.3 Microplastic uptake

A total of 34 fish were found with MP in their GIT (23.13 %FO). The proportion of MP-feeding individuals ranged between 13.64% and 31.03% across the four different stations located in the interior and middle sections of the Ria Formosa lagoon (Table 3), the only white seabream collected at the station “Exterior” had no MP in its GIT. In total, 56 MPs (4.19 %P) were found in the GIT of the juvenile white seabream across the four different stations, the majority of them being fibres (47/56), the remaining MPs were particles. The mean number of MP per MP-feeding fish was 1.64 ± 1.04. Most MP-feeding individuals had only one MP in their GIT (22 of 34 fish), two and three MP were found in five fish respectively, four and five MP were detected in only one fish each. A pairwise comparison across sampling sites and campaigns revealed no statistically significant differences in the uptake of MP (pairwise t-Test, adjustment: Benjamini and Hochberg, ***P*** = 0.46 – 0.93).

Of the MP detected in the GIT, the majority were of blue colour (19/56), followed by black (11/56) and red (11/56). A total of eight green / blue green MPs were found, additionally, four transparent fibres, two yellow particles as well as one purple fibre. Fibres were frequently found entangled within algae, detritus, and digested material (Fig. 7). However, the few, small particles detected were also incorporated into the GIT content. Both MP categories were carefully extracted to examine them in detail for verification of their artificial origin. Considering the overall small amount of ingested MP (Table 3), as well as their rather negligible size relation in comparison to natural prey items and digested materials (Fig. 7), the overall importance of MP in the diet of juvenile white seabream appears to be marginal. This is also confirmed by the fact that neither plastic fibres nor particles as artificial prey items were significant contributors to any observed trends in the analyses of feeding strategy and preferred prey uptake (Fig. 5 and 6).

Table 3 Overview of microplastic (MP) uptake by juvenile *D. sargus* across four different stations. The single white seabream, collected at station "Exterior" had no plastic in its GIT and is therefore not represented here. %FO = Frequency of occurrence; n_{MP}/n_{total} = number of MP-feeding individuals per total number of individuals investigated; the MP category (plus total numbers per category) detected in the GIT; the mean number of ingested particles plus standard deviation along with the range of MP ingested by MP-feeding individuals.

Station	%FO (n_{MP}/n_{total})	MP category (n_{total})	MP mean ± SD (min – max)
Media 1	28.21% (11/39)	Fibre (12), particle (3)	1.36 ± 0.81 (1-3)
Media 2	19.6% (11/56)	Fibre (15), particle (4)	1.73 ± 1.01 (1-4)
Interior 1	13.64% (3/22)	Fibre (3)	1 ± 0 (1)
Interior 2	31.03% (9/29)	Fibre (17), particle (2)	2.11 ± 1.36 (1-5)

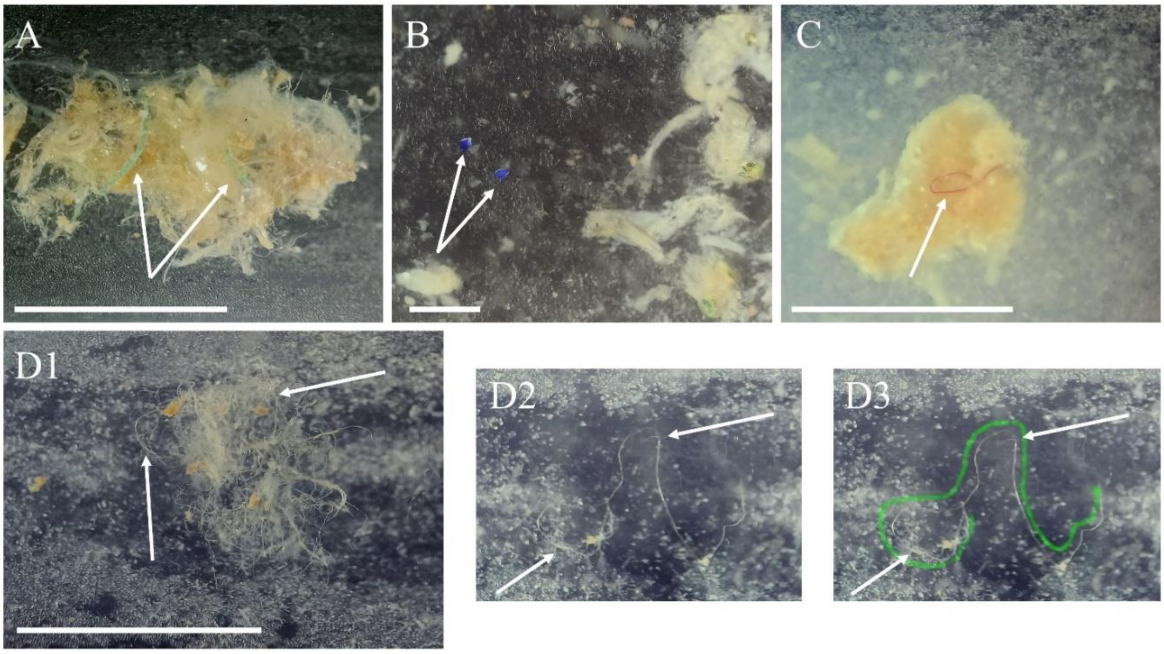

Fig. 7 Exemplary photographs showing different fibres and particles detected in the GIT of juvenile white seabream, Diplodus sargus. A: green fibre entangled within algae and detritus; B: two blue particles with zoea remains for size comparison; C: red fibre in digested material; D1 – D3: transparent fibre entangled within algae - separated from algae – separated and outlined in green for enhanced visibility. Scale bar = 500 µm.

3.4 Prey and MP uptake contextualisation

3.4.1 Prey availability across stations

Potential spatial and temporal differences in planktic and benthic prey availability (individuals m^{-3}) were assessed by computing an ANOSIM (based on Bray-Curtis-dissimilarity), followed by the indicator species analysis (as done in section 3.3.2). Several taxa (particularly benthic ones) were excluded from the analysis due to their inconsistent representation, namely benthic Mollusca and Polychaeta, but also planktic items such as Insecta and Actinopterygii. For the remaining groups, counts per taxon (individuals m^{-3}) from benthos and plankton sampling were grouped where applicable (e.g., Gammaridae, Caprellidae). The prey composition and abundance showed no significant differences (ANOSIM R = -0.14, $\boldsymbol{P} = 0.6995$), neither between the five sampling sites nor the two sampling campaigns (beginning of July, middle of August 2018). Moreover, according to the indicator species analysis, neither natural nor artificial prey items were significantly associated with any sampling site or campaign. Due to the mismatch between environmental prey availability and GIT content, computation of selectivity indices such as Chesson's α (Chesson 1978) could not be computed.

3.4.2 Contextualisation of MP uptake

MP uptake was contextualised with sampling data (i.e., sampling site, sampling campaign), environmental parameters (e.g., temperature, salinity, oxygen) as well as morphometrics and condition (e.g., SL, H, total wet weight, wet weight of GIT, GIT fullness, Fulton's K) (Fig. 8). The number of MP (including both fibres and particles) in the GIT was not correlated with station, sampling, or any abiotic parameter recorded. The number of fibres in the GIT was weakly positively correlated with GIT wet weight ($r_s = 0.2$; $\boldsymbol{P} \leq 0.05$) and moderately positively correlated with GIT fullness ($r_s = 0.42$; $\boldsymbol{P} \leq 0.05$), however there was no correlation verified between MP uptake and any other morphometric measurement or fish condition.

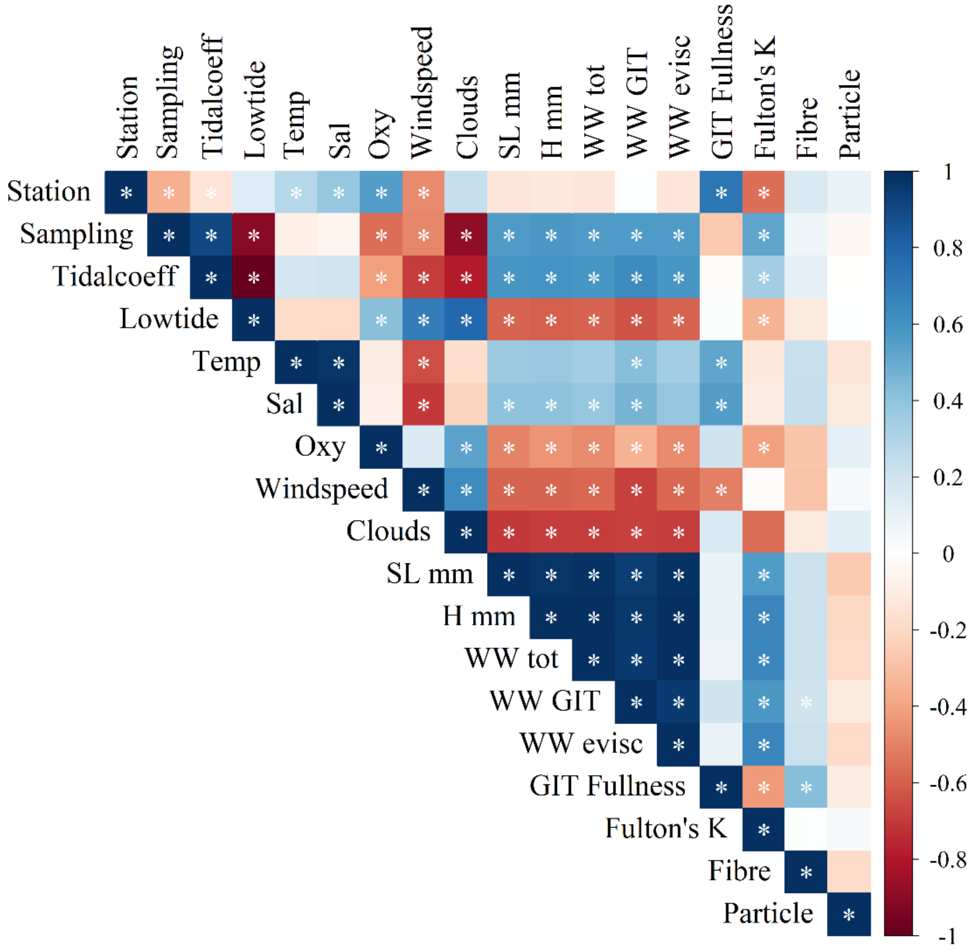

Fig. 8 Correlogram of MP uptake and sampling data, abiotic factors, morphometrics and fish condition, computation based on Spearman rank correlation coefficient for MP-feeding individuals of *D. sargus*. Negative correlation (r_s -1 to 0) is indicated by shades of red, while positive correlation (r_s 0 to +1) is given in shades of blue. Significant associations ($\boldsymbol{P} \leq 0.05$) are highlighted by *.

Contextualising the presence of fibres and particles in the GIT with other prey items (Fig. 9), revealed a strong, negative correlation between the two MP categories present in the GIT (r_s = -0.72; $\boldsymbol{P} \leq$ 0.001). The presence of fibres in the GIT was weakly to moderately positively correlated with algae (alga 1: r_s = 0.44; $\boldsymbol{P} \leq$ 0.01; alga 2: r_s = 0.34; $\boldsymbol{P} \leq$ 0.05). Furthermore, the uptake of fibres was weakly positively correlated with the uptake of detritus (r_s = 0.36; $\boldsymbol{P} \leq$ 0.05). Contrasting this, the presence of particles was moderately negatively correlated to the uptake of detritus (r_s = -0.61; $\boldsymbol{P} \leq$ 0.001) and alga 1 (r_s = -0.4; $\boldsymbol{P} \leq$ 0.05), as well as weakly negatively correlated to the uptake of crustaceans (r_s = -0.35; $\boldsymbol{P} \leq$ 0.05). Additionally, a weak positive correlation was detected between the presence of particles and ascidian tadpoles in the GIT ($r_s = 0.35$; $\boldsymbol{P} \leq 0.05$). No other association of MP and natural prey items was of statistical significance.

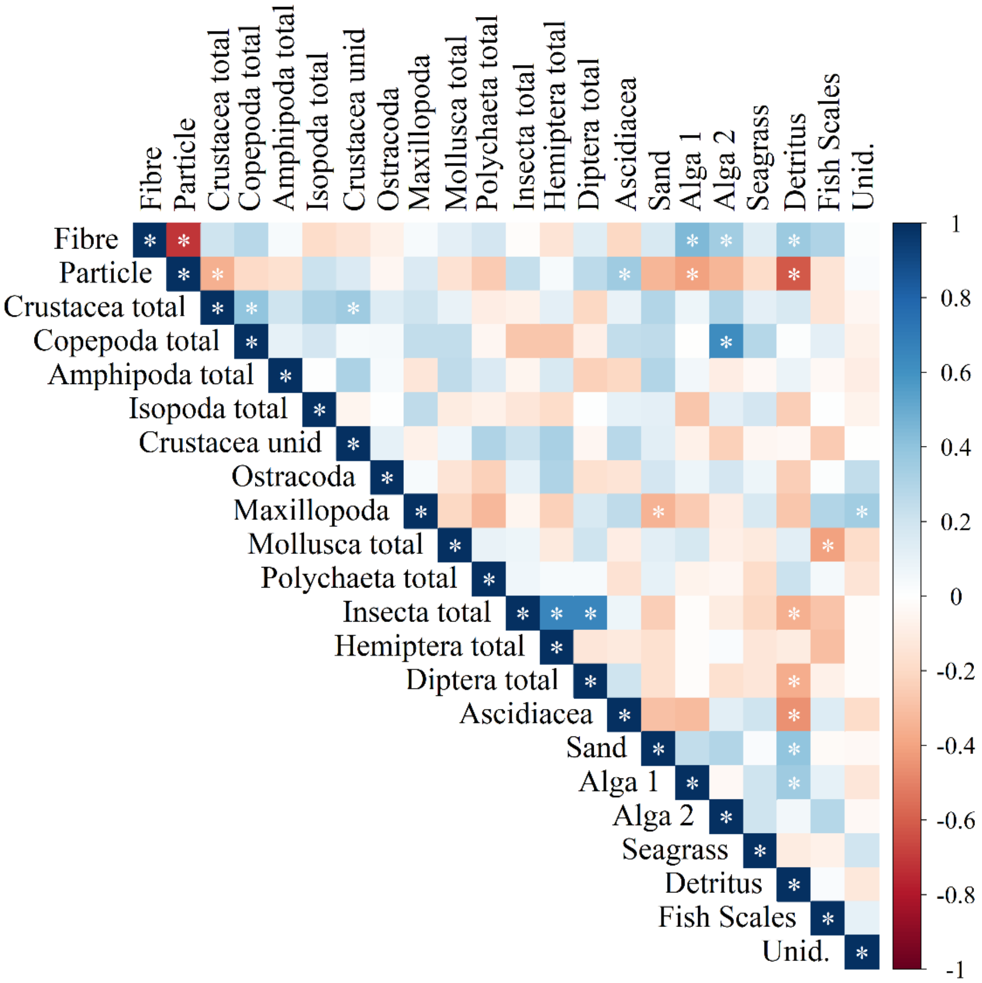

Fig. 9 Correlogram of MP uptake and other key prey items, computation based on Spearman rank correlation coefficient for MP-feeding individuals of *D. sargus*. Negative correlation (r_s -1 to 0) is indicated by shades of red, while positive correlation (r_s 0 to +1) is given in shades of blue. Significant associations ($\boldsymbol{P} \leq 0.05$) are highlighted by *.

4 Discussion

4.1 Quality control and MP identification limitations

Although the development of different digestion protocols (e.g., chemical digestion, enzymatic digestion) to extract MP from biological samples has been advanced over recent years, and the application of such methods is continuously increasing, visual identification and optical analyses are still an important, rapid and relatively cheap method to classify and quantify plastic (Lusher et al. 2017b, 2020). However, without subsequent polymer analysis, these methodologies also bear the risk of high observer biases and identification inaccuracies, especially in relation to smaller MP size spectra (Lenz et al. 2015; Hanvey et al. 2017; Angelini et al. 2019). In the present study, the abovementioned risks were partially overcome by applying strict quality control measures and a conservative MP identification and quantification approach. As the overall goal of this ecological investigation was to provide a contextualisation of MP uptake in a juvenile fish species from an estuarine nursery, the visual assessment of both natural and artificial prey items was necessary despite lowering the reliability of MP detection to particles and fibres > 150 µm (Fig. 7 B). Although the benefits of the ZooScan lie in the quick, semi-automated analysis of zooplankton samples (especially for large sample sets), as well as in the obtainment of meta-data for each scanned image and thus the comparability of data, its limitations in MP detection are entailed in the depiction of images in grey-scales only (Fig. 10), therefore impeding an MP identification based on colour or shades other than within the black-white-range. As can be seen from Figure 10 A – C, artificial prey items, such as fibres (Fig. 10 A) and particles (Fig. 10 B) closely resemble natural ones (i.e., algae and seagrass or gastropod shells) in shape and grey-scales. A digestion of scanned subsamples, a subsequent second image analysis on the ZooScan for verification along with a polymer identification, using for example FTIR, would therefore be advised for future studies aiming at precise quantification of MP concentrations in coastal ecosystems (Lins-Silva et al. 2021). As the present investigation applied a conservative MP identification approach, omitting fibres and particles of questionable origin, the results presented herein offer a baseline of MP concentrations from the respective region. In the macrozoobenthos samples, only a few hard plastic fragments were found, which were in a size dimension larger than any prey item ingestible (> 5 cm) by the target species of this study, thus these infrequent accounts were also omitted. The separation of seagrass and algae from the zoobenthic prey may have led to underestimating the environmental concentrations of MP fibres, which have been found incorporated into detritus and digested plant material in the fish GIT (Fig. 7), yet this MP category was otherwise well represented and identified in the plankton samples.

4.2 Spatio-temporal gradient of habitat quality and prey availability

Similar to the natural fluctuation of abiotic parameters shaping habitat quality and prey availability alike, MP input and distribution in nearshore ecosystems are expected to vary across small spatio-temporal scales, with potential implications for certain taxa and life stages (Ysebaert and Herman 2002; Telesh and Khlebovich 2010; Brito et al. 2012; Cruz et al. 2020).

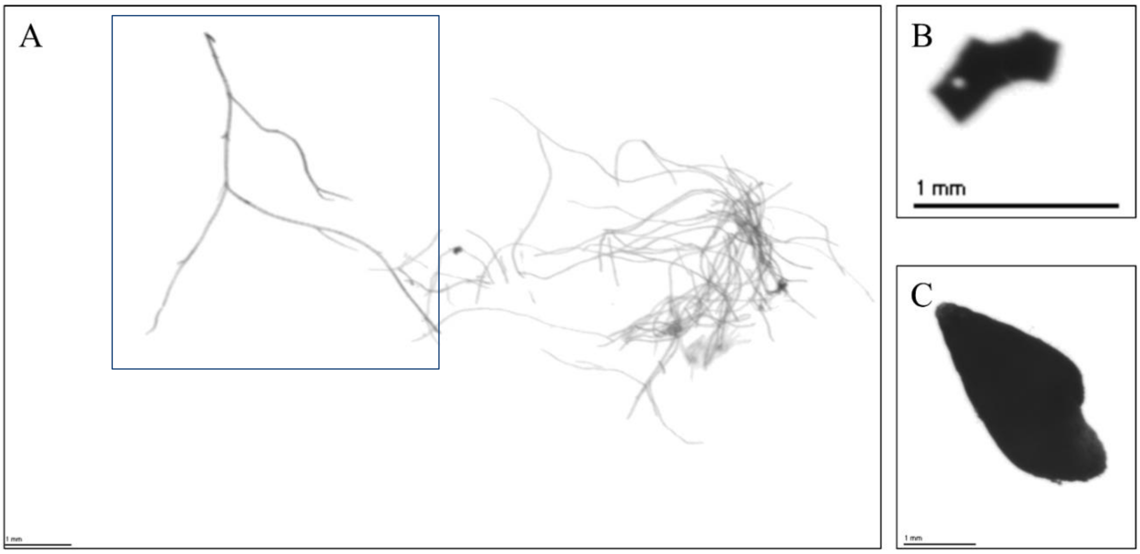

Fig. 10 A – C Natural prey in comparison to Microplastic and Macroplastic items: A) alga, highlighted in blue box to the left, and artificial fibres; B) microplastic item; C) Gastropod shell.

The present study, conducted in summer 2018 in the Ria Formosa lagoon, an important nursery ground on the Southern Portuguese coast, found small fluctuations of abiotic parameters (i.e., temperature, salinity, dissolved oxygen) from inside the lagoon to the inlet to the Atlantic Ocean, verifying the spatio-temporal trends previously described (Newton and Mudge 2003). As oxygen availability, salinity and temperature have been found to significantly impact fish behaviour (e.g., the swimming and feeding activity), ingestion and egestion rates as well as the overall spatial distribution in a given habitat (Fry 1971; Fänge and Grove 1979; Blaber and Blaber 1980; Pandian and Vivekanandan 1985; Beer et al. 2018), the MP uptake probability may also vary in relation to these environmental parameters. The present study did not find any correlation between MP uptake and station, sampling campaign, or abiotic parameter, presumably due to the lack of pronounced differences caused by the rather short time frame of the field study along with the high turnover rate of the water volume in the Ria Formosa during each tidal cycle. Previous investigations from estuarine environments were also not able to establish a significant correlation between MP uptake and environmental parameters even if a spatio-temporal pattern of MP ingestion was verified (e.g., Ferreira et al. 2016a, 2018; Vendel et al. 2017; Silva et al. 2018b). Nevertheless, the number of fibres in the GIT was moderately correlated with GIT fullness, which in return was positively correlated with station, temperature, and salinity, indicating an indirect spatial effect of these parameters on MP uptake which needs further investigation, for example by extending the sampling campaign over several seasons, or adding a denser grid of sampling stations from the interior of the lagoon to the exterior.

Based on the zooplankton and macrozoobenthos sampling, no spatial differences in the availability of prey items predominantly consumed by juvenile *D. sargus* were verified, though previous studies found planktic and benthic communities of the Ria Formosa lagoon to vary in relation to site, season and year, bottom characteristics, and vegetation, as well as to abiotic parameters such as tidal elevation or salinity (Sprung 1994; Gamito 2008; Barbosa 2010; Gamito et al. 2012; Cruz et al. 2020). The fact that only prey items actually detected in the fish GIT were considered for this spatial comparison over the course of six weeks (including three sampling campaigns) may have caused this deviation from earlier investigations. Certain taxa moreover had to be excluded from this analysis: though juvenile white seabream ingested insects of the hemipteran order to a significant extent at station “Media 2”, these items were basically non-existent in the zooplankton and macrozoobenthos samples, suggesting a certain selectivity of the gear in use. Additionally, benthic molluscs and polychaetes, which were highly abundant in the respective benthos samples and could potentially have driven differences in prey availability across the different stations, were hardly detected at all in the fish GIT.

Next to the detrimental effects of climate change scenarios, anthropogenic disturbances and pollutants entering the Ria Formosa (Bebianno 1995; Cortesão et al. 1986; Newton et al. 2003; Newton and Mudge 2005), plastic litter represents an additional stressor to this vital ecosystem and its biological communities which nevertheless has been rarely accounted for in the past (Velez et al. 2020; Cozzolino et al. 2020; Oliveira et al. 2020). Regardless the applied conservative approach in MP quantification, the results of this study suggest a tendency of higher MP concentrations at stations inside the lagoon, confirming a spatial trend which has been detected in the Ria Formosa lagoon by Velez et al. (2020) as well as in other estuaries worldwide (Lima et al. 2014; Hitchcock and Mitrovic 2019). The detection of a more pronounced, significant spatial trend in MP concentrations from inside the lagoon to the outer parts of the system may have been impeded by several factors other than sampling gear and MP identification methodologies, mainly: a) the present study sites were located towards the western part of the lagoon, where average water residence time is approximately one day (Nobre et al. 2005; Saraiva et al. 2007), resulting in a high dispersion capacity of this area, while systems dominated by a low wave / tidal regime have been found to bear an overall higher retention potential associated with a lower distribution of plastic items (Velez et al. 2020; Harris et al. 2021); b) due to the absence of ELHS of seabream in unvegetated areas, sampling over bare sediment was not conducted, thus the selected sampling sites for this study were all equally vegetated by seagrass, which is known to act as a sink for plastic particles and fibres in the Ria Formosa (Cozzolino et al. 2020); c) sampling was conducted over six weeks in summer with no or little precipitation recorded, however, MP concentrations have been found to vary with season / rainfall, suggesting higher MP loads and thus encounter probabilities of ELHS of fish and artificial particles earlier during the recruitment season (Lima et al. 2015; Balthazar-Silva et al. 2020).

4.3 Distribution of ELHS of seabream in the Ria Formosa

Members of the family Sparidae frequently use structurally complex, nearshore habitats as nursery grounds with juvenile sparids showing a high site-fidelity (Erzini et al. 2002; Ribeiro et al. 2006; Abecasis et al. 2009; Vinagre et al. 2010), thus the chosen field study design was considered appropriate to reflect potential differences in feeding preferences and MP uptake probabilities. The distribution of ELHS of white seabream in the Ria Formosa lagoon suggests a preference of this species for nursery sites inside the lagoon, which were generally characterised by higher temperatures and salinities along with lower concentrations of dissolved oxygen, and a muddier, finer sediment. The slightly higher values in standard length, wet weight and Fulton's K, recorded at both "Interior" stations at the end of July 2018, may be indicative for the variability in ingress rates into the Ria Formosa, with individuals of high body condition and thus high swimming capabilities advancing to nursery sites further inside the lagoon (Baptista et al. 2019, 2020). This observation needs further verification in order to clearly define the significance of different micro-habitats as nursery grounds inside the Ria Formosa and thus the potential installation of customised protection and conservation measures for the white seabream. The almost complete absence of ELHS of *D. sargus* at the station “Exterior”, the one closest to the Atlantic Ocean, may be regarded as an indicator for the inadequacy of this site for ELHS of seabream in the Ria Formosa, however currently unpublished results from this research project suggest that this station is frequently used by juveniles of the two-banded seabream, *Diplodus vulgaris,* which was also collected in high numbers at both “Interior” stations but only in minor numbers at the “Media” stations of this study. Juveniles of other abundant seabream species of the Ria Formosa, the black seabream, *Spondyliosoma cantharus,* the Senegal seabream, *Diplodus bellottii*, and the annular seabream, *Diplodus annularis*, again showed different distributional patterns over the different sites and sampling campaigns, with peaks in abundances even later in autumn (September / October 2018). This observation is in accordance with previous studies which found seabream species to temporally partition the available habitat and trophic resources inside their vital nurseries (Sánchez-Velasco and Norbis 1997; Erzini et al. 2002; Gamito et al. 2003; Ventura et al. 2015). Taking this niche partitioning behaviour into consideration, the MP encounter probabilities across distinct seagrass meadows and thus the potential detrimental effects for ELHS and recruitment success may vary between the different seabream species, posing a currently unexplored risk to this viable ecological and economical resource for local artisanal fisheries and aquaculture (Leitão et al. 2009; Bonanno and Orlando-Bonaca 2020).

4.4 Feeding ecology of white seabream and MP uptake contextualization

Ichthyofaunal communities have been proven to be useful biological indicators to monitor ecosystem health and environmental quality (Whitfield 1996; Whitfield and Elliott 2002; Ribeiro et al. 2008; Ramos et al. 2012), thus assessing the feeding ecology of ELHS of the omnivorous white seabream was considered an important first step towards a holistic understanding of the potential impacts of plastic pollution in an estuarine nursery ground. The assessment of the entire GIT content allows for the evaluation of inter-individual and species-specific prey preferences along with feeding habits potentially facilitating the disproportionate uptake of MP and therefore enables a sound estimation of the ecological threat this anthropogenic pollutant represents (Ory et al. 2017; Cardozo et al. 2018; Lopes et al. 2020). As management and conservation measures rely on a sound scientific basis, the GIT analysis performed herein was complemented by the assessment of environmental parameters and prey availability, a necessary data integration only infrequently accounted for in previous studies on MP uptake by fish (Gamito et al. 2003; Cardozo et al. 2018; McGregor and Strydom 2020; Müller 2021; Wootton et al. 2021).

The GIT content analysis revealed a generalized, omnivorous feeding mode of juvenile white seabream in the Ria Formosa: both floral and faunal prey items were ingested across the different sampling sites and campaigns, with slight differences in resource utilization detected between individuals of the different stations. Algae and seagrass were important prey items across all stations in July and August 2018, which is in line with previous studies describing the feeding preferences of white seabream (Sala and Ballesteros 1997; Figueiredo et al. 2005; Merciai et al. 2018). The importance of crustacean and insect prey organisms, which are less rapidly digested than smaller, softer items, may be slightly over-estimated (Windell and Bowen 1978; Buckland et al. 2017), however the relevance of these prey taxa in the diet of *D. sargus* has also been reported before (Rosecchi 1987; Sala and Ballesteros 1997; Osman and Mahmoud 2009). Noteworthy is the presence of two fish ectoparasites, namely Gnathiidae and *Caligus* spp., in the GIT of the juvenile fish, further establishing evidence for a facultative cleaning behaviour contributing to the trophic resource utilisation of *D. sargus* (Rosecchi 1987; Mariani 2001; Neto et al. 2019).

The majority of studies on MP uptake in fish examined adult individuals, thus ambiguities remain particularly regarding the driving factors for MP ingestion in ELHS along with the potential detrimental effects this pollutant exerts on vulnerable life stages in estuarine ecosystems (Browne et al. 2011; Steer et al. 2017; Vendel et al. 2017; Critchell and Hoogenboom 2018; Müller 2021). Although several *in-situ* studies on the feeding ecology of juvenile white seabream have been published over the past decades, MP uptake by this omnivorous species of commercial importance has been rarely investigated, thus intraspecific comparisons of MP uptake can be drawn only to a limited extent. Across the different stations considered for this study, the proportion of individuals with MP detected in their GIT ranged between 13.64 – 31.03 %FO, with a mean load of 1.64 ± 1.04 MP per MP-feeding individual. The proportion of MP-feeding fish *in-situ* is slightly higher than the MP-uptake verified by a feeding experiment conducted on juveniles of the same species: in the laboratory set-up, the ingestion rate of polystyrene particles (500–1000 µm) across the different treatment groups varied between 9.8 and 17.65 %FO, however higher mean MP loads per MP-feeding individual as well as pronounced inter-individual differences in MP-uptake were reported (Müller et al.

2020). A recent multi-species assessment conducted at the Egyptian coast found a MP-feeding proportion of 100 %FO (n = 40) along with high average loads of 3593 ± 3985 particles in juvenile *D. sargus* (Shabaka et al. 2020). Upon closer examination of the substantial variation in the reported %FO and MP-loads in juvenile *D. sargus,* the studies vary in several factors previously identified to impede the comparability of findings, namely the study/sampling characteristics (i.e., number and age/size of fish examined, sampling location), the applied quality control measures along with MP detection and quantification methodologies, as well as the MP size ranges and types considered (Collard et al. 2019; Markic et al. 2020; Müller 2021).

Although the role of feeding mode in MP uptake is still debated and no common consensus has been achieved (Mizraji et al. 2017; Markic et al. 2020), the present study found the ingestion of MP fibres to be correlated with feeding on marine plants, which may suggest a higher potential of herbivorous or omnivorous fish species to take up MP fibres along with their natural, soft-bodied prey (Peters et al. 2017; van der Hal et al. 2020). In benthic invertebrates, the combination of MP characteristics (i.e., size, shape) and the feeding habits of a species were reported to be more decisive in relation to MP ingestion than the trophic guild in itself, a finding which still requires further verification for ichthyofaunal taxa (Piarulli et al. 2020). The sensory perception of prey before and during intake along with their specialised food handling apparatus, notably the different types of teeth, enable members of the sparid family to utilise a broad food spectrum: all prey items are initially sucked in, those without a carapace directly reach the pharyngeal jaws while the buccal jaws hold back hard-bodied prey items for seizing, crushing, and rejecting via the mouth (Vandewalle et al. 1995). Due to this specialisation, ontogenetic shifts in dietary preferences, which have been described for the white seabream with larger individuals (> 150 mm SL) showing a tendency to ingest hard-bodied prey, such as gastropods and echinoderms (Figueiredo et al. 2005), may not necessarily affect the amount and type of plastic intake with varying age/size of the fish. The present study detected only a small number of hard-bodied natural and artificial prey items in the GIT of the juvenile white seabream, which have been found to discriminate polystyrene fragments from crustacean prey (Müller et al. 2020), further supporting the abovementioned specialisation in food intake. The observation of higher loads of MP fibres than MP particles in the GIT of the fish is furthermore in agreement with previous investigations reporting fibrous MP to be more abundant both in the marine realm (Wright et al. 2013; Rochman et al. 2015) and in fish GIT (Lusher et al. 2013; Bessa et al. 2018). Given the intense commercial fishing activity in the Ria Formosa lagoon and adjacent coastal waters, the most probable source of the fibres are fishing gears. Although the threads frequently detected in the plankton samples (Fig. 3 B + C) were beyond the ingestible size range for juvenile white seabream, their fragmentation products, i.e., smaller fibre bundles or individual fibres (Fig. 7 D1 – D3) were found in the GIT of the fish, and thus the spatial trend described for bigger sized plastics may be still considered ecologically relevant.

Considering the ratio of natural to artificial prey items detected in the GIT of juvenile white seabream from the Ria Formosa, the relevance of plastic in the diet seems to be rather marginal. This study could not verify accumulation of plastics in the GIT or detrimental effects of MP ingestion on this life stage and species. Smaller plastic items and associated chemicals have been found to be translocated to other organs and tissues, posing an ecotoxicological risk to the fish and potentially also for human health upon consumption of contaminated tissues (Rochman et al. 2014; Avio et al. 2015; Barboza et al. 2020). The arising implications, particularly for commercially relevant fish species, need further evaluation by assessing the amount of smaller-sized micro- and nanoplastics taken up accidentally along with the food, through trophic transfer or via drinking, making use of advanced identification and quantification methodologies such as FTIR spectroscopy (Setälä et al. 2014; Roch et al. 2020; Veerasingam et al. 2020).

5 Conclusion

Integrative studies, considering both the feeding biology of a species and the environmental availability of plastic pollution, have the potential to enhance our understanding on the extent to which fish deliberately or unintentionally ingest MP. Despite the continuously increasing number of studies on MP ingestion by fish, the extent of MP uptake by ELHS, being crucially dependent on nursery grounds in transitional ecosystems, needs further scientific exploration. Though juvenile white seabream show an omnivorous feeding habit and may encounter a gradient of habitat quality and elevated plastic encounter rates in their nurseries, their specialised mode of prey uptake may prevent them from ingesting high concentrations of hard-bodied MP particles, yet they may still be prone to taking up fibrous MP as well as smaller-sized micro- and nanoplastics along with their natural prey. Using ichthyofaunal communities as biological indicators for MP pollution in coastal ecosystems should be realised only under consideration of species- and life-stage-specific feeding modes and prey preferences as these factors may affect the uptake probability of different MP shapes, sizes, and colours.

6 CRediT authorship contribution statement:

Carolin Müller: Conceptualization, Methodology, Validation, Formal analysis, Investigation, Writing – Original Draft, Visualization, Funding acquisition. **Karim Erzini:** Supervision, Conceptualization, Methodology, Validation, Investigation, Resources, Writing – Review & Editing, Project Administration, Funding acquisition. **Tim Dudeck:** Formal analysis, Investigation, Writing – Review & Editing, Visualization. **Joana Cruz:** Validation, Investigation, Writing – Review & Editing. **Luana Santos Corona:** Investigation, Writing – Review & Editing. **Felipe Eloy Abrunhosa:** Investigation, Writing – Review & Editing. **Carlos Manuel Lourenço Afonso:** Validation, Investigation, Writing – Review & Editing. **Miguel Ângelo Franco Mateus:** Validation, Investigation, Writing – Review & Editing. **Cristina Orro:** Investigation, Writing – Review & Editing. **Pedro Monteiro:** Investigation, Writing – Review & Editing. **Werner Ekau:** Supervision, Conceptualization, Methodology, Validation, Resources, Writing – Review & Editing, Project Administration, Funding acquisition.

7 Declaration of interests

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

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Chapter 3

"It is a capital mistake to theorize before one has data. Insensibly one begins to twist facts to suit theories, instead of theories to suit facts." – Sherlock Holmes

Arthur Conan Doyle (1891): A Scandal in Bohemia

Assessing microplastic uptake and impact on omnivorous juvenile white seabream *Diplodus sargus* (Linnaeus, 1758) under laboratory conditions

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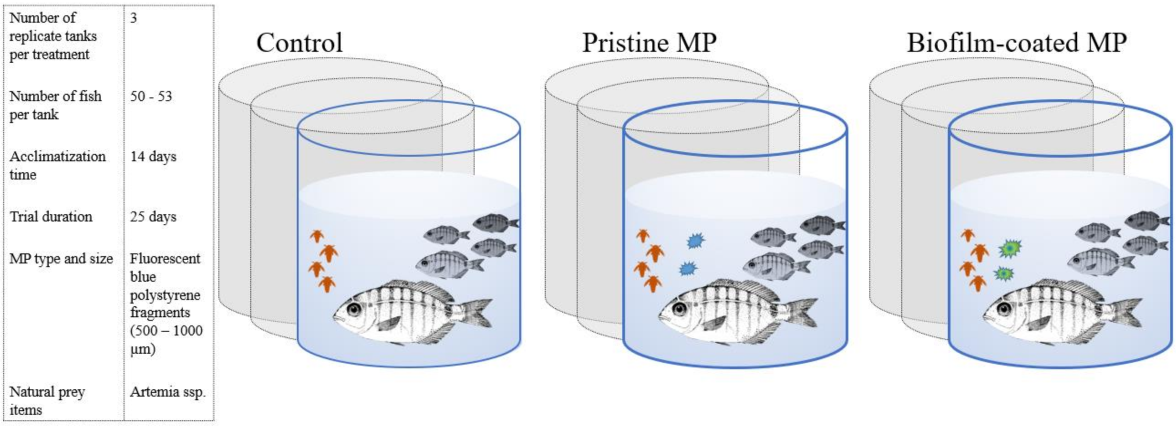
Abstract

Previous laboratory feeding experiments, representing the state-of-the-art methodology to investigate microplastic (MP) ingestion and its impact for fish, tend to disregard both the significance of applying realistic MP densities and the potential relevance of biofilm-coating for ingestion probability. This experiment assessed the uptake of either pristine or biofilm-coated MP particles and the physiological impacts for juvenile white seabream for MP concentrations consistent with those found in the field along with natural prey over a course of 3.5 weeks. Results indicate the ability of juvenile *D. sargus* to discriminate between edible and non-edible prey. A distinct preference for biofilm-coated over pristine particles could not be verified. No significant impact on growth and condition was found except for high levels of MP ingestion. The outcomes highlight the importance of performing MP feeding experiments mimicking natural conditions to reliably assess the impact of MP on early life-history stages of fish.

Keywords: Sparidae, early life stages of fish, polystyrene, RNA:DNA, otoliths, ingestion, stomach content



Number of replicate tanks per treatment	3
Number of fish per tank	50 - 53
Acclimatization time	14 days
Trial duration	25 days
MP type and size	Fluorescent blue polystyrene fragments (500 – 1000 µm)
Natural prey items	Artemia ssp.

Graphical Abstract Overview of the experimental design of the 3.5-week-long microplastic (MP) feeding trials with juvenile *Diplodus sargus*. All treatments are tested by three replicate tanks, each holding approx. 50 juveniles. Fish of the control group were fed with live prey exclusively, whereas fish of the two MP treatments were additionally offered polystyrene particles (either pristine or biofilm-coated). © Figure white seabream *Diplodus sargus*: Arias García and Drake Moyano (1990).

1 Introduction

As early as in the 1970's, along with the onset of reports on microplastic (MP) in sea water samples, early life-history stages (ELHS) of fish, as part of the marine zooplankton community, have been observed to take up MP in case studies in the Atlantic Ocean (Carpenter et al. 1972; Kartar et al. 1973, 1976). Besides reporting the presence or absence of MP in the gastrointestinal tract (GIT) of a fish from *in-situ* investigations, the potential impacts of ingestion are studied via laboratory feeding experiments (Colton et al. 1974; Morgana et al. 2018). Since the pioneering studies in the 1970s, the number of research projects studying MP-uptake based on field investigations (e.g., Boerger et al. 2010; Choy and Drazen 2013; Lusher et al. 2013; Bellas et al. 2016; Vendel et al. 2017) and feeding experiments in laboratories (Colton et al. 1974; Rochman et al. 2013; de Sá et al. 2015) has steadily increased. Collating the outcomes of these investigations, the present state of knowledge suggests that direct ingestion as targeted food or mistaken prey items is expected to be the major pathway of MP uptake in fish (Lusher 2015), though the levels of uptake across different age groups, populations and species seem to vary greatly (Carpenter et al. 1972; Kartar et al. 1976; Davison and Asch 2011; Dantas et al. 2012; Jabeen et al. 2017).

In almost five decades of investigations on MP ingestion by fish, the outcomes are equivocal and in-depth investigations on effects of the ingestion are still scarce, inconsistent and seldom cover juvenile life stages of commercially important fish species (Oliveira et al. 2013; Neves et al. 2015). Although it has been hypothesized that ELHS have a higher potential of MP uptake than adult fish (Critchell and Hoogenboom 2018), studies assessing the potential implications of MP exposure for ELHS are relatively scarce (Browne et al. 2011). Previous laboratory feeding experiments, representing the state-of-the-art methodology to investigate the potential impacts of MP ingestion, hardly reflected natural conditions in the experimental design, both in terms of polymer types and quantities applied (Phuong et al. 2016). Moreover, the role of colonization (i.e., biofilm-coating) in odour emission and thus inducement of MP uptake has rarely been considered in the set-up of the feeding experiments (Hoss and Settle 1990; Savoca et al. 2017), with application of exclusively pristine particles in most laboratory studies even though colonization of MP particles is initiated within hours in nature (Oberbeckmann et al. 2015).

Considering the lack of consistency in MP feeding experiments and the limited information on the actual impacts of MP uptake, with this study we aim to contribute to filling existing knowledge gaps in relation to the potential effects of MP ingestion on growth, condition and survival of ELHS of fish. Based on the hypothesis that juvenile fish of an omnivorous species show a potential for MP ingestion and that biofilm-coating impacts feeding behaviour, our primary research question is as follows: if applied in realistic densities, does MP exposure and uptake affect growth, condition, and survival of juvenile fish?

To answer this question and to assess the validity of feeding experiments as a measure to investigate MP effects on juvenile fish, juvenile white seabream *Diplodus sargus* (Linnaeus, 1758) were tested in a novel set-up, applying MP exposure densities close to environmental conditions, along with live prey over a course of 3.5 weeks.

2 Materials and Methods

Juvenile *D. sargus* were used as model organisms for this study. The species is classified as omnivorous (Joubert and Hanekom 1980) and distributed widely in Atlantic and Mediterranean waters (Joubert and Hanekom 1980; Figueiredo et al. 2005; Abecasis et al. 2009). It is a recreationally (Veiga et al. 2010) and commercially important fish species both for artisanal fisheries and aquaculture. Known to use near-shore habitats as nursery grounds and to show a high site fidelity (Erzini et al. 2002; Abecasis et al. 2009), it can be expected to have a high potential for MP ingestion (Mizraji et al. 2017).

2.1 Feeding experiments

The feeding experiments were conducted from April to May 2018 inside the facilities of the Ramalhete Marine Biology Fieldwork Station, of the Centre of Marine Sciences (CCMAR), Faro (Portugal). The experiments were performed in accordance to the National (Decreto-Lei 113/2013) and EU legislation (Directive 2010/63/EU) regulations on the protection of animals used for scientific purposes. A description of the laboratory set-up is given hereinafter; the supplementary material includes a flow diagram (Fig. S1) which provides an overview of the timeline of the preparations and the actual experimental design.

2.1.1 Microplastic Preparations

Microplastic (MP) fragments within a size range of 500 – 1000 µm were prepared from blue fluorescent polystyrene granulate with an original size of 2.5 x 2 mm, purchased from Magic Pyramid, Bruecher & Partner KG, Frechen, Germany (see supplementary material for details). One half of the prepared fragments was put into a fine mesh stocking and submerged into a tidal pond next to the Ramalhete Marine Biology Fieldwork Station for 21 days to induce biofilm formation; the second half was used in its pristine form.

2.1.2 Stock and Maintenance of Cultured Fish

A total of 476 juvenile *D. sargus* (78 days after hatching) were obtained from an aquaculture facility, the Estação Piloto de Piscicultura (EPPO) in Olhão, Portugal. For experimental use, 456 fish were transferred and allocated about equally across the nine 70 l experimental tanks inside the facilities of Ramalhete. The remaining 20 fish were transferred to another 70 l tank; these fish were initially kept as spare fish for potential replacement during the acclimatization period in case of high mortalities as a direct consequence of transportation or initial measurements. Ten of them were later sampled for RNA:DNA ratio analysis, the remaining ten individuals, however, were not used during the trial. Upon transfer, all fish were precautionarily treated with H_2O_2 to prevent potential infections due to injuries caused by the transport. During an acclimation period of 14 days, the photoperiod was adjusted to 12L/12D, the transition from inert to live food (cultured *Artemia salina*) was performed and measurements on standard length (SL), total length (TL), height (H) were taken for all fish by digitally photographing (Canon Powershot SX 220 HS) the fish on a wetted transparent film with graph paper underneath and measuring them afterwards using the software ImageJ.

2.1.3 Experimental Design

Calculation of MP density

The calculation of the number of particles in each tank i.e., the number of particles passing by the feeding radius of a juvenile fish over a 12-hour period, was based on several assumptions and approximations with regard to findings from the field (Fig. 1, additional information in supplementary materials). In brief, the volume of a cylinder was calculated (i.e., the volume of water passing by an individual fish, Fig. 1) to calculate the number of particles passing by within a 12-hour period of daylight. Based on the estimated number of particles m^{-3} obtained by conducting a pilot zooplankton survey in the Ria Formosa lagoon, 4241 particles m^{-3} were deployed in the respective experimental tanks. The number of particles actually deployed during the experiment was based on the derived weight of the particles rather than on counts of individual particles (Table S1, supplementary material).

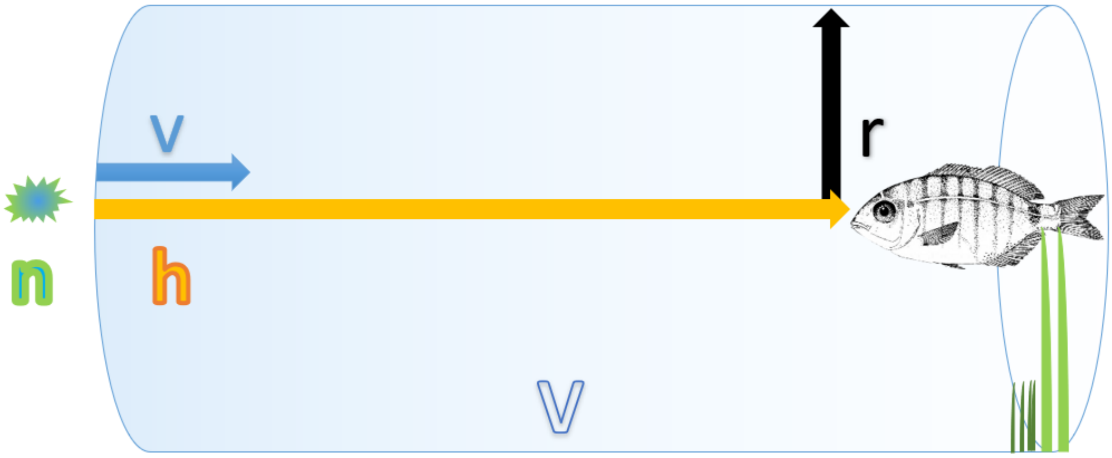


Fig. 1 Schematic visualization of the microplastic density estimation which was based on the following assumptions to calculate the number of particles (**n**) passing by an individual fish during 12 hours of daylight: juvenile fish show a high site fidelity which results in a feeding radius **r** = 0.25 m

(i.e., the size dimension of the experimental tank). In their seagrass habitats, they encounter a medium current velocity **v** = 0.5 m sec^{-1} (Silva et al. 2002; Newton and Mudge 2003). The water length **h** passing by an individual fish is **v** x 60 x 60 x 12 = 21,600 m. The volume V of water passing by an individual fish is $\pi \times r^2 \times h \approx 4241$ m^3. Given there is 1 microplastic particle m^{-3}, the number of particles passing by the feeding radius of a juvenile fish over 12 hours of daylight was estimated to be **n** = **4241**. Figure of juvenile *Diplodus sargus* after Arias García and Drake Moyano (1990).

Experimental Set-up

The experimental set-up consisted of nine conical 70 l fibreglass tanks, equipped with a continuous flow of filtered water from the Ria Formosa lagoon and aeration. To prevent potential contamination with plastic particles in the relevant size spectrum from the Ria Formosa lagoon water, a three-step filtration system was applied: the general filtration system of the facility is a sand filter which uses two filters with a total filtration area of 2.67 m^2. The thickness of the sand bed is 1.2 m and the sand has a particle size of 0.6 – 1.2 mm, the maximum filtration speed is 15 m^3 h^{-1} m^{-2}. The filters applied in the secondary filtration circuit were equipped with a mesh size of 150 µm. Additionally, tube filters installed inside each tank had a mesh size of 150 µm. Along with the procedural monitoring of the temperature and salinity of the ground tank in the station, temperature during the experiment was monitored by deploying probes in three of the experimental tanks (Fig. S2, supplementary material). An additional spare tank was used for the fish that were sampled for RNA:DNA analysis at the beginning of the trial.

The experimental design comprised three different treatment groups which were tested in three separate tanks each (Table 1, Fig. S1): a control group (abbreviation: 'ART') kept as a reference for fish growth and survival without plastic as an artificial prey item. A pristine MP group ('ART+NP') was tested for its potential MP uptake of pristine polystyrene particles along with natural prey items. The potential MP uptake of biofilm-coated particles was investigated (treatment group 'ART+OP') by offering particles which were submerged outdoors in a tidal pond prior to the experiment and kept inside with continuous water flow and aeration during the experiments.

All treatment groups were fed equal quantities of *Artemia spp.* two to five times a day (total amount: ~ 40000 *Artemia spp.* per tank per day) depending on the amount of natural prey per feeding. *Artemia salina,* was hatched and cultured inside the facilities of Ramalhete, fed with microalgae concentrate (*Nanochloropsis oculata*) to enrich their nutritional value. At a later stage of the experiment, *Artemia franciscana* (collected in the field) was given as an additional natural prey item to meet the feeding demands of the growing fish.

Table 1 Overview of the experimental design with three different treatment groups tested for growth and survival of juvenile *Diplodus sargus* under different feeding regimes in triplicate tanks.

Treatment	**Abbreviation**	**Feeding**	**Tanks**	**Total number of fish / treatments**
Control	ART	*Artemia spp.*	H1, H4, H10	153
Pristine MP	ART + **NP** (‘**n**ew **p**lastic’)	*Artemia spp.* + pristine MP particles	H2, H5, H11	151
Biofilm-coated MP	ART + **OP** (‘**o**ld **p**lastic’)	*Artemia spp.* + biofilm-coated MP particles	H3, H6, H12	152

The feeding trial started on 11.04.2018 by applying pristine and biofilm-coated MP to the respective experimental tanks along with the natural prey items fed to all experimental groups. Every second day, the tanks were cleaned from debris, faeces and MP particles (sedimented as well as free-floating ones) by removing approximately 75 % of the water in each tank and siphoning the bottom and walls of the tanks. By removing the MP, the distinction between pristine and biofilm-coated particles in the tanks was maintained, as after the cleaning procedure a new dose of either pristine or biofilm-coated MP particles was applied. The overflow filter in each tank was cleaned every day. This routine was maintained throughout the duration of the trial.

The feeding trial ended on the 05.05.2018. However, to assess the potential egestion time of MP particles by juvenile seabream, half of the fish were removed from the experimental tanks on the 03.05.2018 (i.e., while MP were still present in the tanks), sacrificed by an overdose of an anaesthetic (2-phenoxyethanol), measured (SL, TL, H), weighed (WW) and stored deep-frozen until further analysis. The remaining fish were then transferred into their respective cleaned tank where they stayed for another 48 hours in clean seawater without any new MP dose but fed with natural prey items. On the 05.05.2018, the second half of the fish were finally removed from the tanks and likewise sacrificed and processed.

2.2 Analyses

2.2.1 Mortality

All experimental tanks were routinely checked several times a day for the occurrence of mortality during the trial. Upon detection, dead fish were instantly removed from the tanks, measured (SL, H) and stored individually in Eppendorf tubes filled with ethanol (70%). The GIT content of these fish was analysed according to the procedure described below.

2.2.2 MP uptake by stomach content analysis

Defrosted fish from the feeding trial were dissected to examine stomach and intestinal tract for the presence of natural prey items and blue, fluorescent MP particles, using a UV torch for the latter to increase the detectability. The proportional stomach and intestinal tract fullness with natural prey was assessed and in case of microplastic presence, the location (stomach or intestine) and the number of particles were recorded.

2.2.3 Growth based on morphometrics and otolith analysis

Growth based on morphometrics

To assess potential differences in somatic growth, measurements of SL, TL, H and WW were taken in the beginning of the trial during the acclimatization stage (04.04.2018) and at the end of the experiment (03.05.2018 and 05.05.2018). Based on these measurements, the proportional increase in SL (in %) as well as the absolute growth rate (AGR, in mm day^{-1}) was computed.

Growth based on otolith analysis

As daily growth increment formation for *D. sargus* has been verified (Vigliola 1997), potential differences in growth over the experimental period were examined by measuring the daily growth increment widths for this time period. The sagittae of a subset of fish were removed, otolith measurements (anterior-posterior length, dorsal-ventral height, both in µm) were taken using a Keyence Microscope VHX-5000, and the weight of the otoliths was assessed (weight in mg) using a micro balance (Sartorius ME 36S). Based on the known date of hatching, the pre-experimental life stage was compared to the experimental life stage (maximum last 24 increments) of the fish by measuring the total radius of the otolith from the core to the tip of the rostrum using a digital microscope (Keyence VHX-5000). A second measurement was taken from the core to the daily growth ring which marked the transition from acclimatization to the onset of the laboratory trial, i.e., twenty-second or twenty-fourth-to-last ring respectively (Fig. S3).

2.2.4 Condition based on Fulton's condition factor *K* and RNA:DNA analysis

Condition based on morphometrics (Fulton's condition factor)

Based on the measurements of SL and biomass (i.e., wet weight, with and without alimentary tract), the condition of the fish was calculated, following the formula proposed by Fulton (1904):

$$K = \left(\frac{wet\ weight}{standard\ length^3}\right) \times 100$$

with eviscerated wet weight in g and standard length in cm.

Fulton's condition factor is commonly used as a proxy for the assessment and intra specific comparison of fish health or well-being (Froese 2006); according to the underlying concept of this length-based condition index, fish with a greater mass (i.e., weight) than their conspecifics of a given length are considered to be in better condition (i.e., higher Fulton's condition factor) and vice versa.

Condition based on RNA:DNA analysis

The main premises of the RNA:DNA (RD) ratio are that the concentration of DNA in the somatic cells of animals remains relatively constant, while the concentration of RNA is related to the protein synthesis rates and the recent growth (Bulow 1970). As the ratio of RNA:DNA has been verified to be an appropriate indicator for changes in feeding conditions and growth within short periods of only a few days for a variety of fish species (Buckley et al. 1999; Chícharo and Chícharo 2008), this method was considered suitable to complement the analysis of potential differences across treatments based on morphometrics. As a reference, muscle tissue from between the dorsal and caudal fin of ten fish was sampled at the beginning of the trial (Fig. S1). These fish were kept in an additional tank and fed identically to the ones of the control group. At the end of the trial, a subset of ten fish per tank was then randomly selected for RD analysis; muscle tissue was likewise removed from each selected individual, stored and preserved separately in RNA*later*™ Storage Solution. The processing of the total of 100 samples and the subsequent analysis were conducted at the laboratory facilities of CCMAR, following established protocols (Morais et al. 2015; Cruz et al. 2017; Teodósio et al. 2017;

Baptista et al. 2019). The laboratory procedures are described in detail in the supplementary materials.

2.3 Statistical analysis

Statistical analyses and data visualization were done by using the following software: Microsoft Excel, JMP PRO 14 and R (Version 3.6.0). Data were tested for normality of distribution (Shapiro-Wilk normality test) and homogeneity of variance (*F*-test). If normality of distribution and homogeneity of variance was violated, non-parametric statistical tests were used (Mann-Whitney U-test, Welch's Two-sample t-test). To investigate differences in mortality and growth across the different treatments, Student's t-test was used for normally distributed data with equal variances. The difference in somatic growth from the beginning to the end of the trial was assessed with the Paired t-test for normally distributed data. One-way ANOVA was used to investigate a potential impact of MP-exposure and ingestion on GIT fullness, growth (based on otolith analysis) and condition (based on RNA:DNA ratio). Tukey's test for multiple comparisons of means was applied for the post-hoc evaluation of the GIT fullness and Fulton's *K*. Regression analysis was performed to assess the correlation between SL and otolith length, WW and otolith weight. Level of significance was set to $\boldsymbol{P} \leq 0.05$.

3 Results

3.1 Mortality

A total of 38 fish across all treatments died during the trial: Table 2 gives an overview on the number of fish per tank and treatment at the beginning and at the end of the trial. Mortality was below 12% in all treatment groups, with no significant differences in mean mortality found between the control treatment and either of the two MP treatments (ART and ART+NP: *F*-test, $\boldsymbol{P} = 0.651$, Student's t-test, $\boldsymbol{P} = 0.8169$; ART and ART+OP: *F*-test, $\boldsymbol{P} = 0.014$, Welch's Two-sample t-test for unequal variances, $\boldsymbol{P} = 0.2868$). Furthermore, there was no statistically significant difference in the mortality between the two MP treatment groups (ART+NP and ART+OP: *F*-test, $\boldsymbol{P} = 0.004$; Welch's Two-sample t-test for unequal variances, $\boldsymbol{P} = 0.1976$). Of the 24 fish that died in either of the plastic treatments, only two had plastic in their alimentary tract, with each originating from a different treatment group (1 fish in H12 and 1 fish in H11).

The majority (34/38) of fish that died during the trial had a stomach and intestinal tract fullness of below 50%.

Table 2 Total number of fish (juvenile Diplodus sargus) at the beginning of the trial and alive until the end, total number of dead fish during trial and respective percentage of mortality during the trial. The different treatments are indicated by ART = control; ART+NP = pristine MP; ART+OP = biofilm-coated MP.

Tank	Treatment	Total number of fish at the start	Total number of fish alive at the end	Total Mortality during trial (%)
H1	ART	51	47	4 (7.84%)
H4	ART	50	45	5 (10.00%)
H10	ART	52	47	5 (9.62%)
Total	ART	153	139	14 (9.15%)
H2	ART+NP	49	46	3 (6.12%)
H5	ART+NP	51	45	6 (11.76%)
H11	ART+NP	51	45	6 (11.76%)
Total	ART+NP	151	136	15 (9.93%)
H3	ART+OP	51	46	5 (9.80%)
H6	ART+OP	50	47	3 (6.00%)
H12	ART+OP	51	50	1 (1.96%)
Total	ART+OP	152	143	9 (5.92%)

Given the low mortality rates of less than 12% across all tanks, including the control group triplicates, along with the rare incidence of MP in the GIT of the dead fish of the MP-treatment groups, no effect of the actual treatment on mortality of fish can be inferred.

3.2 Ingestion of MP particles and natural prey items

The uptake of natural prey items and MP particles was assessed by analysing the alimentary tract of all 456 juvenile *D. sargus* at the end of the trial.

Uptake of pristine and biofilm-coated MP particles

Results of the MP uptake analysis for fish of the two MP-treatment groups are shown in Table 3.

Table 3 Results of the alimentary tract analysis of the fish (juvenile *Diplodus sargus*) from the MP feeding trial, sorted by treatment: ART+NP = pristine MP treatment, ART+OP = biofilm-coated MP treatment. Fish from the control group are not included in this table.

Tank	**Treatment**	**Total number of fish at the start**	**Number of fish with plastic alive + dead**	**Percentage of plastic feeders**	**Min. number of MP / fish**	**Max. number of MP / fish**	**Mean number of MP / MP-feeding fish ± SD**
H2	ART+NP	49	6	12.24%	1	78	18.50 ± 30.18
H5	ART+NP	51	9	17.65%	1	53	8.00 ± 16.98
H11	ART+NP	51	6 + 1	13.73%	1	4	2.00 ± 1.41
Total	ART+NP	151	21 + 1	14.57%	1	78	8.95 ± 19.23
H3	ART+OP	51	5	9.80%	1	3	1.60 ± 0.89
H6	ART+OP	50	5	10.00%	1	20	7.80 ± 9.04
H12	ART+OP	51	5 + 1	11.76%	1	3	1.50 ± 0.84
Total	ART+OP	152	15 + 1	10.53%	1	20	3.50 ± 5.58

Within the two plastic treatments (pristine MP plus biofilm MP; n_{total}=303 fish), 38 fish were found with plastic in their alimentary tract, 36 of them alive until the end of the trial. One fish from both the pristine and the biofilm MP treatment that died during the trial was found to have ingested MP particles. Except for five individual fish, which were found with particle loads of more than ten pieces, the majority of fish (33 of 38 MP-feeding individuals) ingested on average 2 ± 1.8 polystyrene fragments.

The highest number of MP-feeding fish was recorded in the pristine MP treatment (ART+NP), with 22 of 151 fish containing MP in their GIT (Percentage of MP-feeding fish = 14.57%; range 12.2 – 17.7%). The mean number of pristine particles per MP-feeding fish across all triplicate tanks was 8.95 ± 19.23. In this treatment, the highest amount of ingested MP particles was also observed across all MP treatments (max. 78 particles in an individual fish; see also Figure 2 with 53 MP in another individual of the same treatment group).

In comparison, the total number of fish with biofilm-coated MP (ART+OP) in their GIT was 16 of 152 (Percentage of MP-feeding fish = 10.53%; range 9.8 – 11.76%). The percentage of plastic feeding fish was similar in all tanks and lower in comparison to the pristine MP treatment. The highest number of particles taken up by a single fish in the biofilm MP treatment was 20. The mean number of biofilm-coated particles per fish across all three tanks was 3.5 ±

5.58. There was no significant difference in the amount of ingested MP between both treatment groups (*F*-test, ***P*** ≤ 0.05, Welch's Two-sample t-test for unequal variances, ***P*** = 0.157).

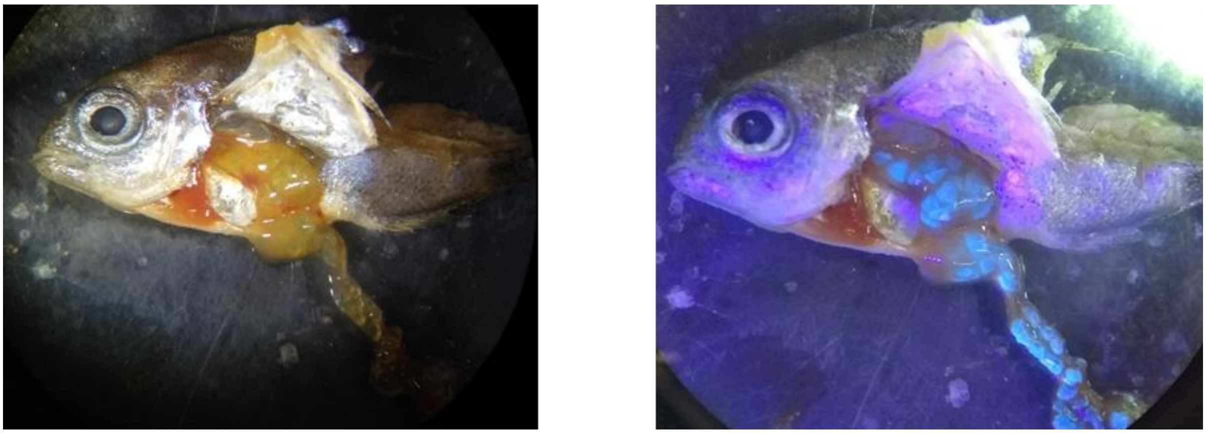


Fig. 2 Photos of a juvenile *Diplodus sargus*, with the intestinal tract lined up outside of the abdominal cavity. This fish was tested for MP uptake in the pristine MP treatment group and was found with 53 polystyrene particles in the GIT at the end of the trial (the picture on the right was taken under UV-light to make the MP particles inside the GIT visible). The same individual was furthermore included in the RNA:DNA analysis.

During the trial, fish in tanks of both MP treatments were frequently observed taking up MP particles and spitting them out again, but a statistical evaluation of this behaviour was not feasible as a constant video-monitoring was not installed. However, as proof of this behaviour, a short video sequence was recorded (see ES1, video sequence in Electronic Supplements).

Relationship between MP uptake and ingestion of natural prey items

Along with the total number of MP particles in the stomach and intestinal tract, the proportional GIT fullness with natural prey items (in %) was estimated (Fig. 3a). The two fish that died during the trial (red symbols in Fig. 3a) which were found with MP in their GIT, had ingested 1 biofilm-coated or 4 pristine MP particles and were found with either near empty (20%) or empty (0%) GITs respectively. Of the fish alive until the end of the trial, the majority (29 of 36) ingested ≤ 5 particles and the mean GIT fullness with natural prey items in these MP-feeding fish ranged from empty (0%) to full (100%). According to Figure 3a, the fish were able to ingest natural prey items along with a modest number of MP particles, unless exceptionally high quantities of synthetic particles were taken up (i.e., more than 15) in which case the GIT fullness ranged between 0 and 40 %. GIT fullness with natural prey items was also assessed for all other fish that were found with no MP in their GIT as well as for all fish of the control group (Fig. 3b).

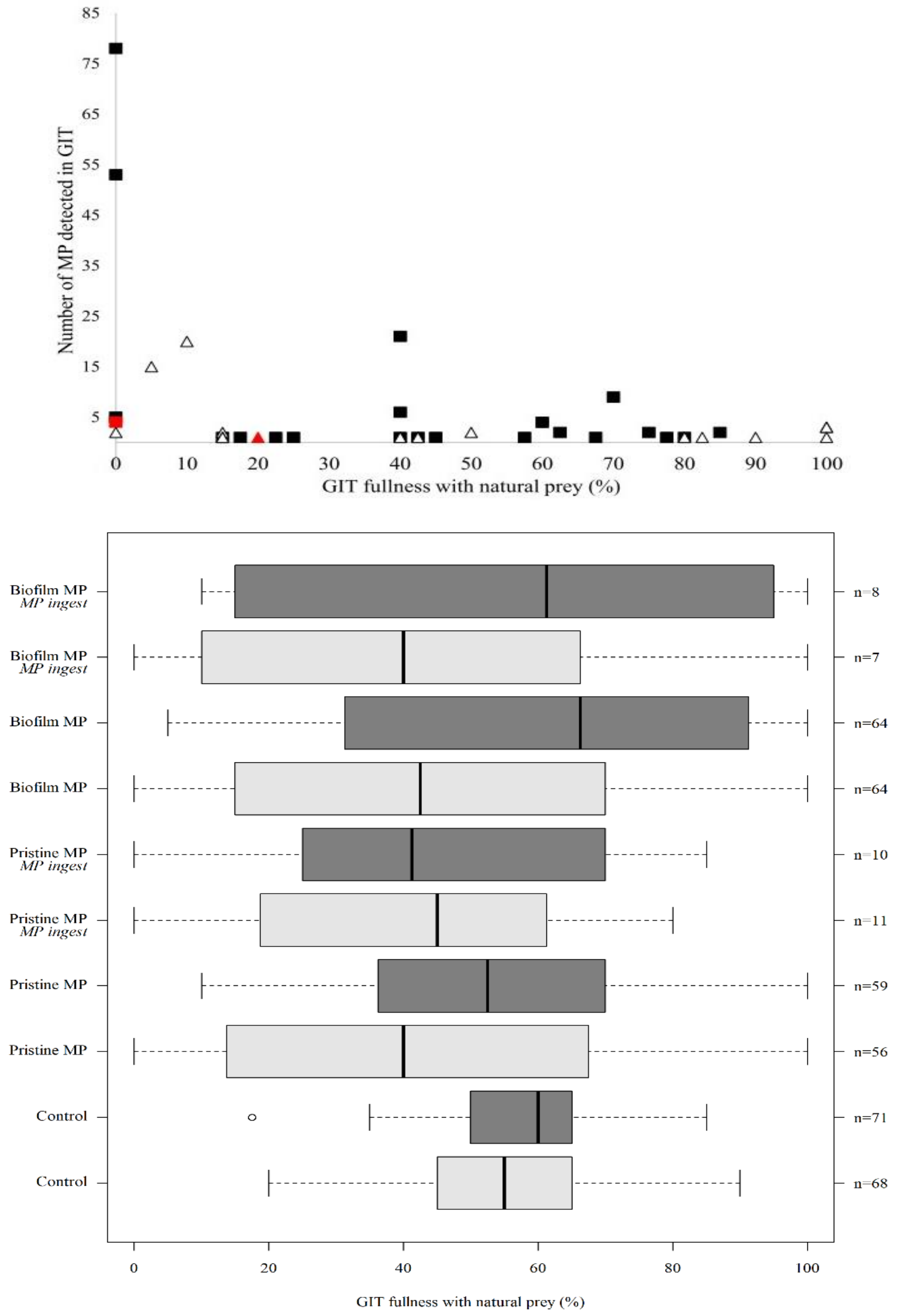


Fig. 3a Number of ingested MP particles (y-axis) in relation to the mean GIT fullness with natural prey items (x-axis). Results for fish (juvenile *Diplodus sargus*) of the pristine MP-treatment are given by ■, whereas Δ indicates results of the fish from the biofilm-coated MP treatment. Red symbols indicate the data of the two fish that died during the trial. **3b.** Boxplots showing the range of GIT fullness with natural prey items for all treatment groups; fish with MP in their GIT ("*MP ingest*") are displayed separately from fish of the same treatment that were found without MP at the end of the trial. The results of the GIT analysis for fish taken out of the trial on the second but last day of the trial (03.05.2018) are highlighted in light grey, results for fish taken out the final day of the trial (05.05.2018) are given in dark grey.

GIT fullness of fish that were taken out of the trial two days prior to the final end (i.e., under MP exposure) was compared to the GIT fullness of the remaining half of the fish that were kept until the termination of the experiment (i.e., in clean tanks without MP for 48 hours) (ANOVA: One-way analysis of means – not assuming equal variances: $\boldsymbol{P} = 0.010$, df = 9). In most cases, fish that were kept until the final day of the experiment had a higher mean GIT fullness compared to the fish that were removed from the tanks on the second but last day (Fig. 3b) – the only exception were MP feeding fish of the pristine plastic treatment, which showed a lower mean GIT fullness on the final day of the experiment. However, significant differences were detected only between MP-non-feeding individuals of the biofilm-coated MP treatment group (Tukey's test for multiple comparisons of means, $\boldsymbol{P} = 0.024$, see Table S2 for detailed comparisons). Fish of the control group showed a slightly higher GIT fullness with natural prey items than fish of the pristine MP treatment group (Fig. 3b). Moreover, the GIT fullness within control-group individuals was less variable compared to fish of both MP treatments, though the tendencies detected were not statistically significant (Table S2).

3.3 Growth

The growth of the fish during the trial was analysed using morphometric measurements, taken at the beginning and the end of the trial (i.e., SL, Table 4), along with the analysis of the otolith daily growth rings, comparing the pre-trial life stage to the phase of the experiment (i.e., the last 24 rings on the otolith).

Somatic growth

The SL of all fish was measured in the beginning and in the end of the trial; the data were tested for normality (Shapiro-Wilk test) and homogeneity of variances (F-test). As there were no statistically significant differences in SL between tanks of the same treatment (Welch's Two-Sample t-test for unequal variances, $\boldsymbol{P} = 0.268$ for comparison of tank H6 with H12; Student's t-test for all other tank comparisons, $\boldsymbol{P} = 0.059 – 0.954$), SL measurements were grouped per treatment in the following analysis. In all treatments, significant somatic growth from the beginning to the end of the experiments was recorded (Shapiro-Wilk normality test, $\boldsymbol{P} = 0.322 – 0.832$, Paired t-test of all comparisons, $\boldsymbol{P} \leq 0.05$). No significant difference in SL between the three treatments was found either at the beginning (F-test, $\boldsymbol{P} \geq 0.05$, Student's t-test, $\boldsymbol{P} = 0.519 – 0.988$) or at the end of the trial (F-test, $\boldsymbol{P} \geq 0.05$, Student's t-test, $\boldsymbol{P} = 0.750 – 0.941$). Moreover, by comparing the SL at the end of the trial for the individual tanks with each other, no significant difference was found in either of the comparisons (Welch's Two-sample t-test for unequal variances, ***P*** = 0.299 – 0.712; Two sample t-test, ***P*** = 0.076 – 0.990).

Table 4 Comparative overview on the mean standard length (SL) of the fish (juvenile Diplodus sargus) in mm (plus standard deviation), with measurements from 04.04.2018, prior to the actual trial phase, and from 03.05. + 05.05.2018, at the end of the trial. Results of the final measurement are grouped into one final measurement. The different treatments are indicated by ART = control; ART+NP = pristine MP; ART+OP = biofilm-coated MP. The proportional increase in length (Growth in %) is given along with the absolute growth rate (AGR in mm day^{-1}).

TANK	**Treatment**	**Mean SL (mm) ± SD**		**Growth (%)**	**AGR (mm day^{-1})**
		Start	**End**		
H1	ART	21.57 ± 3.29	25.33 ± 2.83	17.42	0.121
H4	ART	21.48 ± 2.79	24.56 ± 2.87	14.37	0.099
H10	ART	22.45 ± 2.77	25.59 ± 3.15	13.95	0.101
Total	ART	21.84 ± 2.98	25.17 ± 2.96	15.24	0.107
H2	ART+NP	21.99 ± 3.5	24.83 ± 4.0	12.92	0.091
H5	ART+NP	21.75 ± 2.88	25.05 ± 2.99	15.22	0.106
H11	ART+NP	21.78 ± 3.37	25.55 ± 2.96	17.29	0.121
Total	ART+NP	21.84 ± 3.24	25.14 ± 3.35	15.13	0.106
H3	ART+OP	21.60 ± 2.87	25.10 ± 3.07	16.17	0.112
H6	ART+OP	21.97 ± 3.23	25.12 ± 3.42	14.38	0.101
H12	ART+OP	22.60 ± 2.38	25.54 ± 2.44	13.02	0.094
Total	ART+OP	22.06 ± 2.86	25.26 ± 2.98	14.53	0.103

In terms of proportional growth, all tanks showed an increase in standard length between 12.9 – 17.4% (ART = 15.24%; ART + NP = 15.13%; ART + OP = 14.53%) over the course of the 31 days from the initial to the final measurement (Table 4). The AGR across tanks of the control group during the trial period was 0.107 mm day^{-1}, in the pristine treatment group 0.106 mm day^{-1} and in the biofilm-treatment group 0.103 mm day^{-1}.

Growth based on otolith radii

A total of 121 fish were selected for otolith analysis: the sagittal otoliths of all fish found with plastic in the alimentary tract alive until the end of the trial were removed (n = 36), and 85 fish without MP detected in their GIT were additionally randomly selected across all tanks for otolith analysis ($n_{ART} = 30$; $n_{ART+NP} = 38$; $n_{ART+OP} = 17$). As there were no significant differences between the left and the right otolith in terms of size dimensions (Paired t-test, ***P*** = 0.2609), only left otoliths ($n_{total} = 117$) were used for subsequent analyses of dimension and weight. The relationship between fish length and left otolith length, as well as eviscerated fish weight and otolith weight were described by linear regression models and a high correlation was indicated for both relationships investigated (left otolith length vs. fish SL: $y = 43.855x + 377.96$, $r = 0.909$; left otolith weight vs. fish eviscerated weight: $y = 1.1456x + 0.2583$, $r = 0.891$), thus otoliths were considered appropriate to evaluate growth.

To assess potential differences in growth related to the treatment, the pre-experimental life stage was compared to the experimental life stage of the fish. For a total of 50 MP-non-feeding fish, the core, the tip of the rostrum as well as the growth rings were clearly identifiable on the otoliths ($n_{\text{Control}} = 18$; $n_{\text{Pristine MP}} = 18$; $n_{\text{Biofilm MP}} = 14$). In the MP-feeding fish, a total of 31 otoliths were considered ($n_{\text{Pristine MP}} = 18$; $n_{\text{Biofilm MP}} = 13$). Based on the radii measured, the proportional growth during the experimental trial was calculated; Figure 4 shows the results for the different treatment groups, representing the results of the MP-non-feeding and MP-feeding individuals separately. The proportional growth during the experimental trial was highest in fish in the biofilm-coated MP treatment which were found with no particles in their GIT at the end of the trial, followed by fish of the control group. MP-non-feeding fish of the pristine MP treatment, along with MP-feeding fish of both plastic treatments showed comparable growth rates based on the otolith radii measurements. No statistically significant difference was verified across the different groups of fish (ANOVA: One-way analysis of means – not assuming equal variances: $\boldsymbol{P} = 0.067$, df = 4.0). The proportional growth calculated from the otolith radii thus reflected the growth rate calculated from the SL of the fish.

Additionally, the relative growth per day during the pre-experimental life stage as well as during the experimental life stage was calculated by measuring both otolith radii and dividing them by the respective number of days of each life stage. This was done to assess if fish with a potentially higher relative growth before the experiments also showed a higher growth during the trial and if hypothetical differences were related to the treatment groups. The results are displayed in Figure 5. The relative growth per day was significantly higher during the pre-experimental life stage in comparison to the life stage during the trial within all treatment groups tested (Shapiro-Wilk normality test, $\boldsymbol{P} = 0.194 – 0.709$, Paired t-test of all comparisons, $\boldsymbol{P} < 0.01$). Besides that, no significant differences in the relative growth between the different treatment groups were verified, neither before nor after the experiment (Tukey's test for multiple comparisons of means, $\boldsymbol{P} = 0.769 – 1.0$). Fish with MP in their GIT at the end of the trial did not show a distinct growth pattern (i.e., slower relative growth) in comparison to fish of the same treatment without MP in their GIT (Tukey' test for multiple comparisons of means, $\boldsymbol{P} = 0.971 – 1.0$).

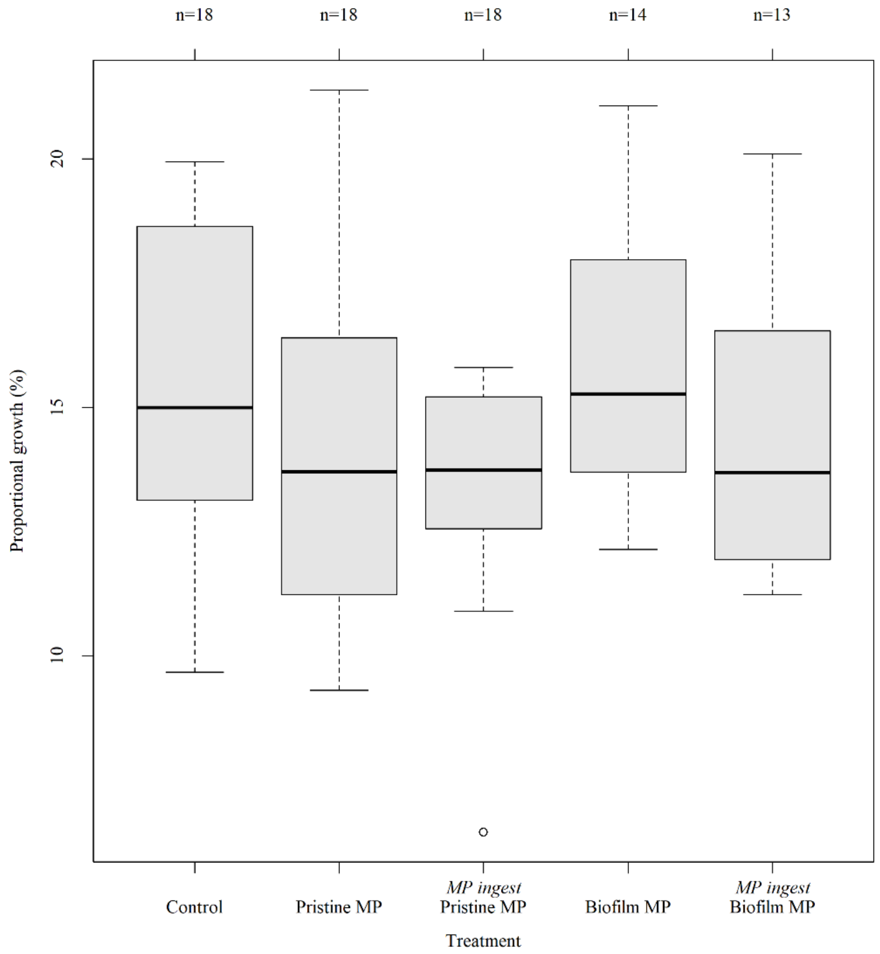


Fig. 4 Boxplots of the proportional growth calculated from otolith radii measured for a sub-set of individuals (juvenile *Diplodus sargus*). Individuals with polystyrene MP particles detected in their gastrointestinal tract at the end of the trial are represented separately ("*MP ingest*") from those of the same treatment without MP in their gastrointestinal tract.

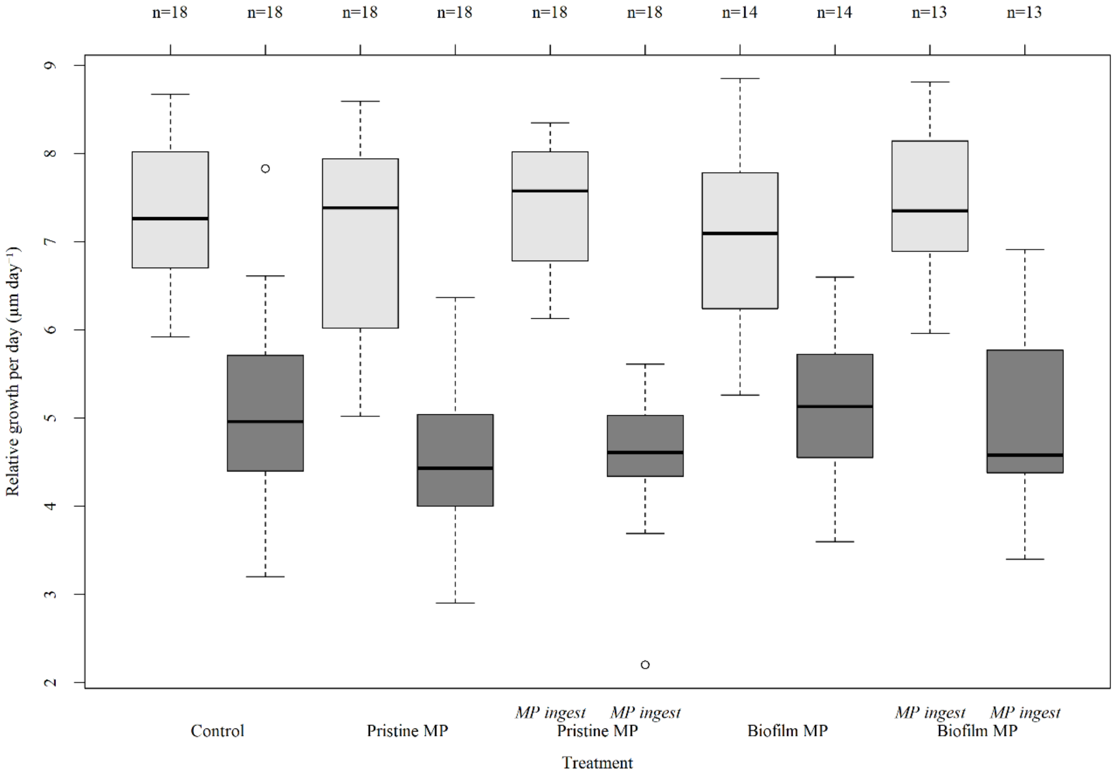


Fig. 5 Boxplots of the relative growth per day calculated from otolith radii measured for a sub-set of individuals. The results for the pre-experimental life stage is highlighted in light grey, results from the trial life stage in dark grey. Individuals with polystyrene MP particles detected in their gastrointestinal tract at the end of the trial are represented separately ("*MP ingest*") from those of the same treatment without MP in their gastrointestinal tract.

3.4 Fish condition

Condition based on morphometrics: Fulton's K

Juvenile fish condition was assessed using the condition factor *K* proposed by Fulton (1904). Figure 6 summarizes the results of the condition calculation of a subset of fish (used also in otolith analysis). Statistically significant differences in the condition were found between both the MP-feeding individuals of the pristine and biofilm-coated MP treatment compared to fish from the control group (Tukey's test for multiple comparisons of means, Control vs. Pristine MP-feeders, ***P*** = 0.039; Control vs. Biofilm MP-feeders, ***P*** < 0.001; see Table S3 for detailed comparisons). Moreover, the plastic-feeding fish of the biofilm-coated MP treatment also had a statistically significant lower condition compared to the MP-non-feeding fish of both MP treatments (Tukey's test for multiple comparisons of means, Biofilm MP-feeders vs. Biofilm MP, ***P*** = 0.029; Biofilm MP-feeders vs. Pristine MP, ***P*** = 0.026). No significant difference was found for fish of the control group compared to fish of the MP treatments which were found with no plastic in their GIT at the end of the trial (Tukey's test for multiple comparisons of means, ***P*** > 0.05, see Table S3 for details).

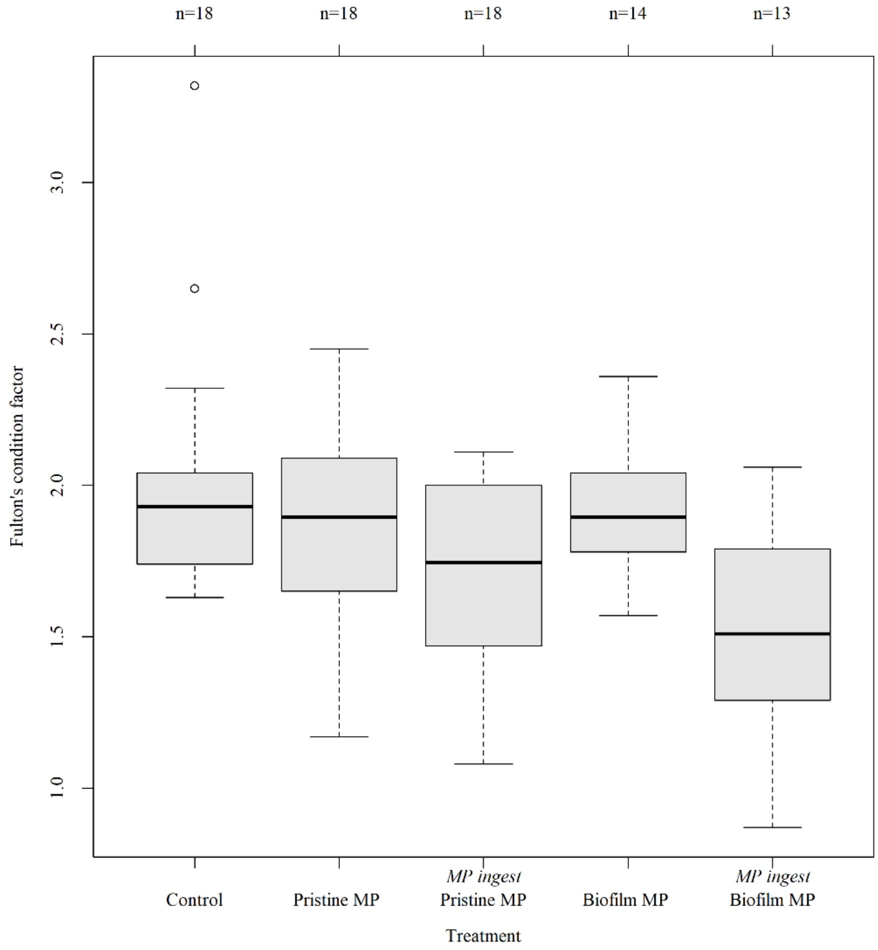


Fig. 6 Boxplot of the condition factor Fulton's K calculated for a subset of fish (juvenile *Diplodus sargus*) values per treatment. Individuals with polystyrene MP particles detected in their gastrointestinal tract at the end of the trial are represented separately ("*MP ingest*") from those of the same treatment without MP in their gastrointestinal tract.

Condition based on RNA:DNA ratio

The analysis of the standardized RNA:DNA (sRD) ratio between fish of the different treatment groups shows the widest inter-quartile range and furthermore the highest upper quartile and total value in sRD ratio for fish sampled at the beginning ('Start') in comparison to all other treatment groups (Fig. 7).

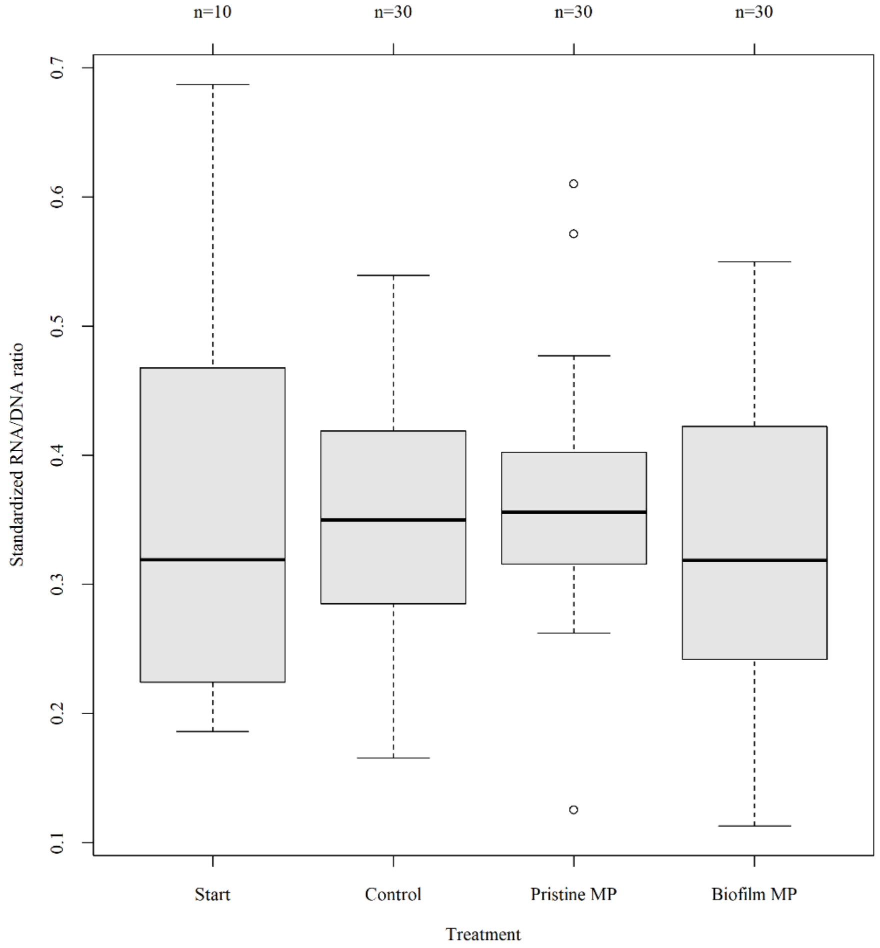


Fig. 7 Boxplot of the standardized RNA:DNA ratios of juvenile *Diplodus sargus* per treatment; 10 fish were sampled at the beginning ('Start') as a reference, 30 fish per treatment group (Control, pristine MP and biofilm-coated MP treatment) were randomly selected at the end of the feeding trial.

The median sRD ratio in the control and pristine MP treatment group are similar, however the interquartile range as well as the upper and lower whiskers of the control group are wider whereas the sRD ratio for fish of the pristine MP group varies less, suggesting a lower variation in condition based on sRD ratio for fish of the pristine MP treatment. Fish of the biofilm-coated

MP treatment show a slightly lower median sRD ratio along with the second widest inter-quartile range, indicating a higher variation in condition based on the sRD ratio for fish of the biofilm-coated MP treatment compared to fish of the pristine MP treatment. Despite the slight tendencies, no statistically significant differences were found, neither between the start of the trial compared to the respective measurements at the end nor between the three treatments compared with each other (ANOVA: One-way analysis of means – not assuming equal variances: ***P*** = 0.539, df = 3; Tukey's test for multiple comparisons of means, ***P*** = 0.505 – 0.997). The outlier with the lowest condition found in the pristine MP treatment was identified to be the fish which ingested 53 MP particles (Fig. 2).

4 Discussion

Methodology

With the onset of MP exposure experiments in the early 1970's by Colton et al. (1974), the number of studies assessing MP ingestion via laboratory set-ups has steadily increased with a peak in published laboratory studies after 2012 and fish being among the group of taxa most frequently studied in experimental set-ups (de Sá et al. 2018). Along with the growing number of feeding trials conducted, objections were raised in scientific publications to the consistency of these experimental designs with natural conditions, including remarks on the limited number of sample sizes and life stages tested over short trial durations without control treatments (Lusher et al. 2017a; Cunningham and Sigwart, 2019), the MP particle types, size ranges, polymers and concentrations deployed (Phuong et al. 2016; Mouneyrac et al. 2017; Ašmonaitė et al. 2018). Moreover, important gaps were revealed concerning the MP degradation state applied (i.e., pristine MP instead of biofilm-coated, weathered MP) and the incidence of secondary ingestion / trophic transfer as a mode of MP uptake considered in the set-up (Lehtiniemi et al. 2018; Paul-Pont et al. 2018). The present study took these critiques into consideration to improve the set-up: the number of fish tested in each treatment (i.e., ~ 50 per tank, thus ~ 150 per treatment) as well as the level of replication (triplicates) and the time scale were considered adequate to deliver meaningful results within the given experimental set-up (Thorarensen et al. 2015). The uptake of either pristine or biofilm-coated blue polystyrene particles was compared, a polymer type and colour that have been reported to be abundant in the marine environment (Boerger et al. 2010; Song et al. 2014; Erni-Cassola et al. 2019). Moreover, irregularly shaped particles of an ingestible size-range were deployed in environmentally realistic concentrations along with natural prey items. To the best of our knowledge, the current study is the first to assess MP uptake along with potential detrimental effects on growth and condition for an ELHS of a commercially important fish species under these environmentally realistic experimental conditions. Even though the otolith microstructure analysis showed a marked difference between the increment width of the daily growth rings prior to the experiments compared to those during the trial, the low mortality rates along with the significant somatic growth across all tanks and treatments are a valid argument to consider the husbandry conditions as well as the nutritional supply adequate. The reduced growth of juvenile *D. sargus* during the experimental trials we assume to be linked more to environmental factors such as lower temperature during the experiments (Fig. S2.; Pedro Pousão-Ferreira, pers. comm.) rather than to the exposure and uptake of MP.

Due to the distinctiveness of the experimental design, evaluating the results in terms of percentage of MP-feeding individuals or the number of ingested MP particles per individual with respect to other MP feeding studies on this very species or other taxa seems hardly expedient. Nevertheless, to provide an overview, experimental set-ups in MP feeding trials conducted with ELHS of marine fish species were collated in Table S5, complemented by a synoptic analysis of the different approaches (see supplementary materials).

Feeding behaviour

The overall proportion of MP-feeding fish in this experiment ranged from 9.8 to 17.7% across tanks and MP treatment groups. Comparing these results to more than 90 *in-situ* assessments summarized recently in two reviews (Markic et al. 2020; Müller, unpubl.), which found approximately one third of individual fish with MP particles and / or fibres in the GIT, the present MP-feeding frequency may be considered rather low, even though field studies regularly also take fibres into account which was not the case in the present study. Concerning the MP-uptake rate or particle load per MP-feeding individual, the juvenile white seabream showed a wide range of 1 to 78 MP particles ingested. The average particle load detected in MP-feeding fish sampled in the field (including both fragments and fibres) amounted to roughly two particles per individual (Markic et al. 2020; Müller, unpubl.). While *D. sargus* is known to undergo several ontogenetic dietary shifts (Christensen 1978) and displays an omnivorous feeding mode (Leitão et al. 2007), which in return is hypothesized to be related to a higher probability of MP uptake (Mizraji et al. 2017; Markic et al. 2018), the rather low proportion of MP-feeding individuals partially contradicts this notion. As the majority of MP-feeding fish ingested on average 2 ± 1.8 polystyrene fragments (excluding five fish with exceptionally high MP uptake), the hypothesis of trophic guild acting as the main driver for MP ingestion may be challenged (Güven et al. 2017; Sun et al. 2019; Markic et al. 2020). These findings suggest inter-individual or species-specific prey preferences and may thus represent a determining factor for the ingestion of synthetic particles which should be taken into account in future investigations.

With a hydrophobic surface, rapidly stimulating biofilm formation in the water column, plastic marine debris might emit odour which was shown to induce foraging behaviour in zooplankton (Vroom et al. 2017) and fish (Savoca et al. 2017). In the present study, no preference for biofilm-coated MP particles over pristine ones was verified; on the contrary, pristine MP were taken up by a higher number of fish in the respective treatment compared to the frequency of ingestion in the biofilm-coated treatment group. Moreover, fish of the pristine treatment ingested on average more and higher numbers of particles compared to fish of the biofilm-treatment. Irrespective of the lower frequency of uptake and lower ingested particle counts per individual in the biofilm-treatment, individuals of this group showed a slightly lower condition (Fulton's K, RD) and reduced growth compared to the control and pristine treatment even though food intake with natural prey items based on the assessment of GIT fullness at the end of the trial was not significantly different from the other fish.. This may lead to the conclusion that biofilm-coating on the one hand does not necessarily induce foraging behaviour and on the other hand that the constitution of this biofilm might represent a crucial parameter when evaluating the impact of MP ingestion on fish growth and condition. Thus future studies should be encouraged to closely examine the microbial species composition along with the concentration of persistent organic pollutants inherent to the biofilm-coating of MP fragments (Ašmonaitė et al. 2018). Independent of whether the particles were pristine or biofilm-coated, juvenile white seabream tested in the present study were observed to frequently expel MP fragments immediately upon ingestion – a comparable observation was made by Colton et al. (1974) and Ory et al. (2018b). As no statistical evaluation of this behaviour was possible in this study, the question arises if fish in the biofilm-treatment group had to invest more time and effort to find suitable (i.e., natural) food items due to confusion of biofilm-coated MP with organic prey, which in return could explain the slightly lower condition observed in this group.

The extent to which individual fish are able to discriminate between natural prey items and artificial particles or fibres as well as their ability to actively reject synthetic particles certainly needs more examination on the basis of scientifically sound experimental designs. In relation to the potential impact of biofilm-coating on growth and condition as well as on the risk of accumulation of MP in the GIT, the retention time of MP in the stomach and intestinal tract may be of particular relevance: results of the current study indicate that juvenile *D. sargus* were not able to egest MP particles within 48 hours. This preliminary assessment is in accordance with the outcomes of other studies that found the retention time of MP in the GIT to range between days to several weeks (Gassel et al. 2013; Ory et al. 2018b). The retention of MP in the GIT may therefore be considerably longer than the time needed to digest and egest natural food items (Ory et al. 2018b). Based on the detection of several polystyrene particles at the far end of the intestinal tract of the juvenile *D. sargus* in this study, the general ability to egest MP particles of a size range between 500 – 1000 µm is indicated. While GIT fullness with natural prey items was significantly higher only in MP-non-feeding fish of the biofilm-treatment that were kept for two days without MP exposure (in comparison to fish that were taken out the second but last day of the trial), this trend was present in all groups tested to some extent (Fig. 3b; Table S2); this could indicate that on the one hand the natural prey density was not consistently adjusted to the lower number of fish in the tanks and on the other hand the absence of MP may affect and enhance uptake of natural prey items. More research is needed therefore on the GIT clearance rate for different life stages and species under varying environmental conditions (Fänge and Grove 1979; Pandian and Vivekanandan 1985; Beer et al. 2018) in order to evaluate the accumulation potential of MP along with the impact of retained particles on the condition of ELHS of fish as well as on the effect of MP exposure on uptake efficiency and feeding behaviour.

MP impact on life performance of juveniles

Among the documented effects of MP exposure and ingestion relevant for comparison with this study are an increased mortality (Mazurais et al. 2015), the reduction of predatory performance and efficiency (de Sá et al. 2015) as well as a compromised feeding ability and potential blocking of the digestive tract in EHLS of fish (Cole et al. 2013; Foley et al. 2018). In the present study, an increased mortality caused by MP exposure and uptake was not detected. Moreover, substantial impacts of MP on juvenile fish condition were not verified: unless taken up in high quantities, the uptake of MP did not impair the uptake of natural food items; egestion of MP particles in the given size range appears to be feasible. Concomitant with the ability to ingest a sufficient amount of prey, the growth of fish of the control group compared to the individuals of the plastic treatments did not differ significantly over the course of the study, which was reflected by both the somatic growth (SL) as well as by the otolith radii measurements. An extension of trial periods however is strongly suggested for future studies in order to thoroughly assess the long-term impact of MP ingestion on growth (Foley et al. 2018). Individuals with MP detected in their GIT at the end of this trial however had a significantly lower condition (based on Fulton's K) compared to the fish of the control group. The lack of significant differences on the basis of sRD analysis may be related to the fact that among the fish randomly sampled before dissection (i.e., 30 per treatment), only a small proportion were also found with MP in their GIT (pristine MP-feeding individuals = 5; biofilm-coated MP-feeding individuals = 3). As verified also by Fulton's K, the mere exposure to MP polystyrene fragments of a size range between 500 – 1000 µm might therefore not cause an alteration in the condition of juvenile fish of that age and body size. An excessive uptake of plastic particles however may have an effect on sRD as could be inferred from the outlier of the pristine MP treatment, which was identified as the fish that ingested 53 polystyrene particles. As the impact of ingestion is likely to be dependent on the size relation between the organism affected and the particle encountered (Lusher 2015), smaller-sized particles may be taken up in higher quantities by ELHS of fish (Critchell and Hoogenboom 2018) and could thus adversely affect growth and condition. Taking into account that the classification of MP-feeding and MP-non-feeding individuals in this study was based solely on the presence or absence of plastic particles in the GIT at the end of the trial, the absence of clear tendencies with regards to detrimental effects on growth and condition could be explained. It cannot be excluded that a higher number of individual fish took up plastic during the course of this study and therefore potential differences in growth and condition in these individuals may confound the patterns observed. Continuous video-monitoring (as frequently done in bio-assays) and individual tagging of the fish may assist in eliminating this source of inaccuracy.

Contextualization

Relating the findings from this laboratory trial with the preliminary results of an *in-situ* study of the same species (within the same size range, caught in a comparable temperature regime in a lagoon in Southern Portugal), the presumption of a selective, omnivorous feeding habit along with inter-individual prey preferences and minor to no MP uptake in this species seems to prevail (Müller et al. unpubl.). Taxon-specific or inter-individual feeding preferences for certain prey items such as gastropods and bivalves - as verified for *D. sargus* (Osman and Mahmoud, 2009; Leitão et al. 2007) – contrast with the general perception of fish feeding preferentially on soft-bodied prey items (Jobling et al. 2001), and thus could represent a potential driver for varying levels of MP uptake across taxa or individuals. As fish taxa with distinct prey preferences may show diverging levels of MP uptake, depending on the polymer size (Critchell and Hoogenboom 2018), colour (Ory et al. 2017) and shape (Silva et al. 2018b), future laboratory set-ups should devote special attention to the feeding ecology and biology of the species under investigation. The outcomes of environmentally sound experimental set-ups will assist in properly assessing the implications of ingested MP detected in the GIT of wild-caught fish.

5 Conclusion

Laboratory feeding experiments have been shown to be a suitable approach to investigate uptake and potential effects of MP ingestion in ELHS of fish, although particular consideration should be given to designing experimental set-ups simulating natural conditions as much as possible. Even though a preferential uptake of biofilm-coated MP over pristine particles was not verified for juvenile white seabream, analysing the composition of the biofilm-coating on MP to assess the potential effect for forage inducement as well as the impact on fish condition and ingestion rates may be considered expedient. The exposure of a juvenile omnivorous fish to environmentally realistic concentrations of polystyrene MP (500 – 1000 µm), along with the ingestion of either pristine or biofilm-coated particles in minor numbers did not adversely affect survival, growth and condition in these fish over a course of 3.5 weeks. Future investigations should continue to assess MP uptake and gastric evacuation rates of different particle shapes (i.e., fibres, filamentous MP) and size spectra; moreover, potential impacts of MP ingestion on larval and juvenile stages of fish under varying environmental conditions need to be comprehensively examined. In-depth research is needed to thoroughly evaluate the findings of both laboratory and *in-situ* studies and to holistically assess the effect of this anthropogenically induced stressor to ELHS of fish, acting as a potential bottleneck for successful recruitment.

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7 CRediT authorship contribution statement

Carolin Müller: Conceptualization, Methodology, Formal analysis, Investigation, Writing – original draft, review & editing, Visualization, Funding acquisition

Karim Erzini: Conceptualization, Resources, Writing – review & editing, Supervision, Funding acquisition

Maria Alexandra Teodósio: Methodology – RNA:DNA, Resources, Formal analysis – RNA:DNA, writing – review & editing

Pedro Pousão-Ferreira: Resources – provision of animals, Writing – review & editing

Vânia Baptista: Investigation – RNA:DNA, Formal analysis – RNA:DNA, Writing – review & editing

Werner Ekau: Conceptualization, Formal analysis, Resources, Writing – review & editing, Supervision, Funding acquisition

Supplementary Materials

Materials & Methods

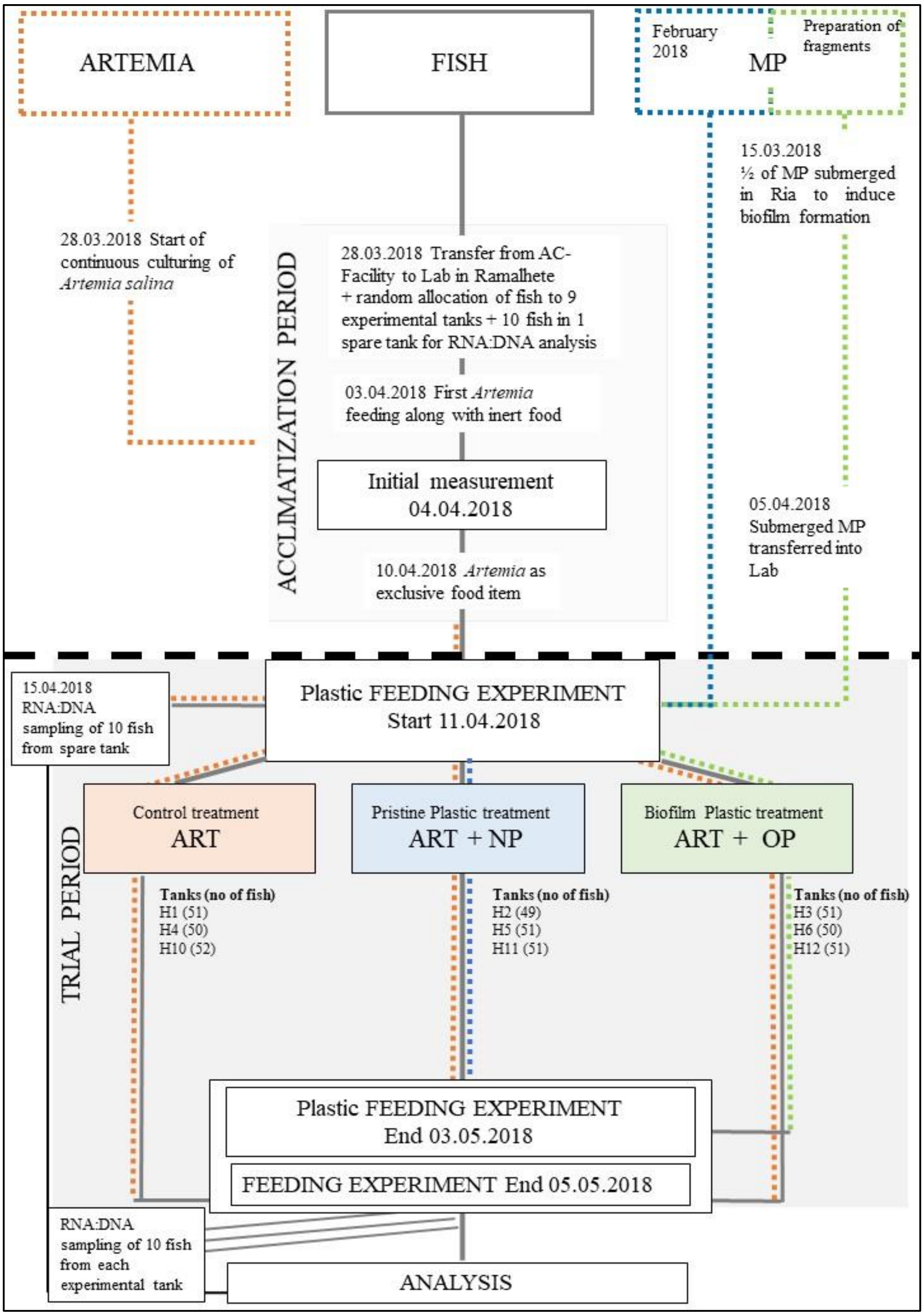


Fig. S1 Flow diagram of the preparations and experimental set-up of the MP feeding experiments conducted with juvenile white seabream. After an acclimatization period, fish were tested in three different treatments (each with three replicate tanks): the control treatment (‘ART’) which was fed with natural prey items only, the pristine MP treatment (‘ART+NP’) which received pristine polystyrene particles along with natural prey, and the biofilm-coated MP treatment (‘ART+OP’) which was offered natural prey and polystyrene particles that were submerged in a tidal pond three weeks prior to the experiments. The actual MP feeding trial lasted from the 11.04.2018 to the 05.05.2018.

Experimental Design

Preparation of MP particles

MP fragments within a size range of 500 – 1000 µm were prepared from blue, fluorescent polystyrene granulate with an original size of 2.5 x 2 mm, purchased from Magic Pyramid, Bruecher & Partner KG, Frechen, Germany. The granulate was ground using a 450 Watt hand blender in combination with an electric coffee grinder which yielded irregularly shaped particles which were subsequently sieved with metal sieves to obtain the desired size spectrum that was considered to match the prey size of the fish at that age / size. One half of the prepared fragments was put into a fine mesh stocking and submerged into a tidal pond next to the Ramalhete Marine Biology Fieldwork Station in order to induce biofilm formation; the second half was used in its pristine form. In order to facilitate the application of MP densities in coherence with MP densities found in the field (i.e., the Ria Formosa, southern Portugal), 10 x 500 dry particles were weighed to derive a mean weight per particle which was then used to calculate the total weight of dry particles to be deployed in the respective experimental tanks. The difference in weight between pristine dry and biofilm-coated wet particles was taken into account by weighing three samples with wet MP of the calculated target weight and counting the particles afterwards to assess the difference in particle numbers. By accounting for this difference, comparable numbers of dry and wet particles were applied to the respective treatment groups (Table S1).

Table S1 Applied polystyrene particle weights in g per tank and treatment. Based on the derived weight of 500 particles (ten individual weightings), the target weight of 1.26683 g pristine MP and 1.31282 g biofilm-coated MP were applied.

	Pristine MP Treatment			Biofilm MP Treatment		
Date	Tank H2	Tank H5	Tank H12	Tank H3	Tank H6	Tank H12
11.04.2018	1.26675	1.26665	1.26668	1.3126	1.3111	1.31261
13.04.2018	1.26631	1.26685	1.26628	1.31139	1.31162	1.31148
15.04.2018	1.2663	1.26613	1.26633	1.31186	1.3111	1.31131
17.04.2018	1.26662	1.26678	1.26662	1.31217	1.31231	1.31228
19.04.2018	1.26646	1.26634	1.26645	1.31282	1.31266	1.31275
21.04.2018	1.26636	1.26641	1.26637	1.31276	1.31282	1.31278
24.04.2018	1.2668	1.26677	1.26678	1.31281	1.31279	1.31282
26.04.2018	1.26669	1.26678	1.26676	1.31259	1.31268	1.31264
28.04.2018	1.26671	1.2667	1.2667	1.31278	1.31274	1.3128
30.04.2018	1.26677	1.2668	1.26677	1.3125	1.31253	1.31249
02.05.2018	1.26679	1.26672	1.26678	1.31268	1.31264	1.3127

Calculation of MP density

In order to establish experimental conditions resembling natural microplastic (MP) abundances, a pilot zooplankton and MP survey at three distinct sites in the Ria Formosa lagoon was conducted using a neuston net with a mesh size of 200 µm. Based on the visual assessment using a stereo microscope, a mean particle density of 1.1 particle m^{-3} was found.

The calculation of the number of particles in each tank i.e., the number of particles passing by the feeding radius of a juvenile fish over a 12-hour period, was then based on the following assumptions and approximations with regard to findings from the field:

- Juvenile white seabream show a high site fidelity
- Juvenile white seabream are particle feeders, selecting prey (i.e., zooplankton) from the water column
- There is a certain density of microplastic particles in the habitat $n = 1.1$ particles m^{-3}

- Juvenile fish feed within a radius of $r = 0.25$ m (i.e., in accordance with the size dimensions of the experimental tanks), facing into the mean current velocity $v = 0.5$ m s^{-1}
- Juvenile fish feed during daylight (i.e., 12 hours a day)

To specify the number of particles to be deployed in the experimental tanks, the volume of a cylinder was calculated (i.e., the volume of water passing by the individual fish) as

$$V = \pi \times r^2 \times h$$

where h is the length of the cylinder, which is determined by the flow velocity multiplied by time, and r is the radius of the cylinder, defined by the feeding radius of the fish (Fig. 2). The number of particles m^{-3} was rounded to 1, resulting in 4241 particles m^{-3} to be deployed in the respective experimental tanks.

Experimental Set-up

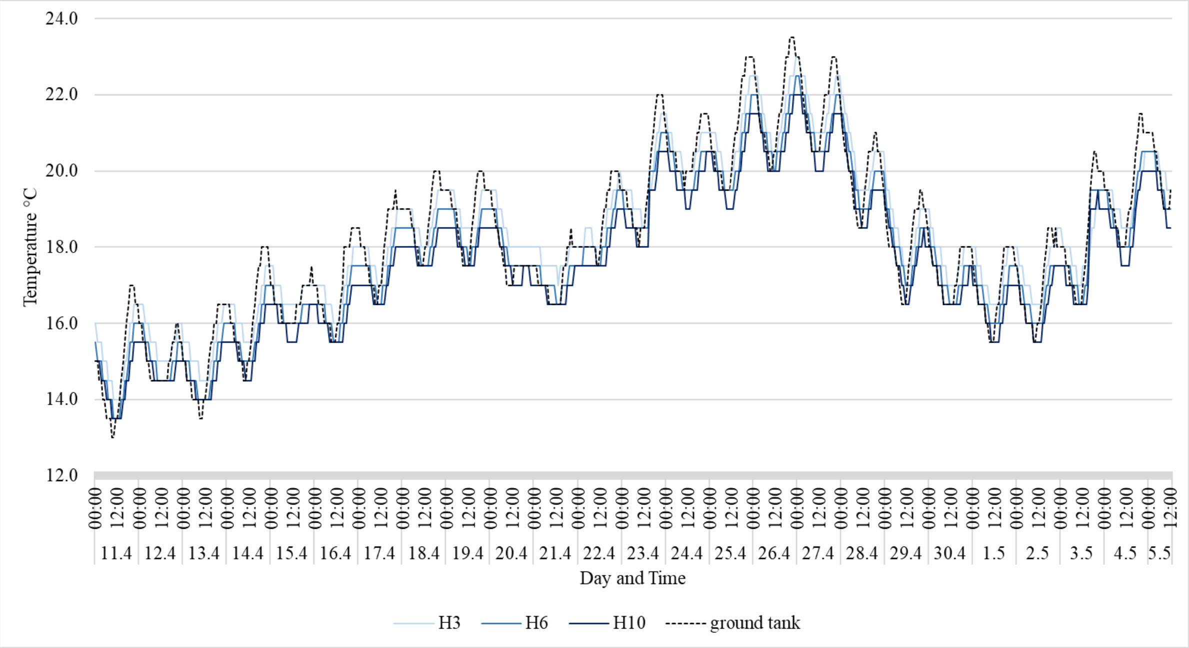


Fig. S2 Temperature profile recorded in three different experimental tanks (H3, H6, H10) and in the ground tank at the Marine Biology Fieldwork Station, Ramalhete, Faro (Portugal) during the feeding experiment in April – May 2018.

Analyses

Growth based on otolith analysis

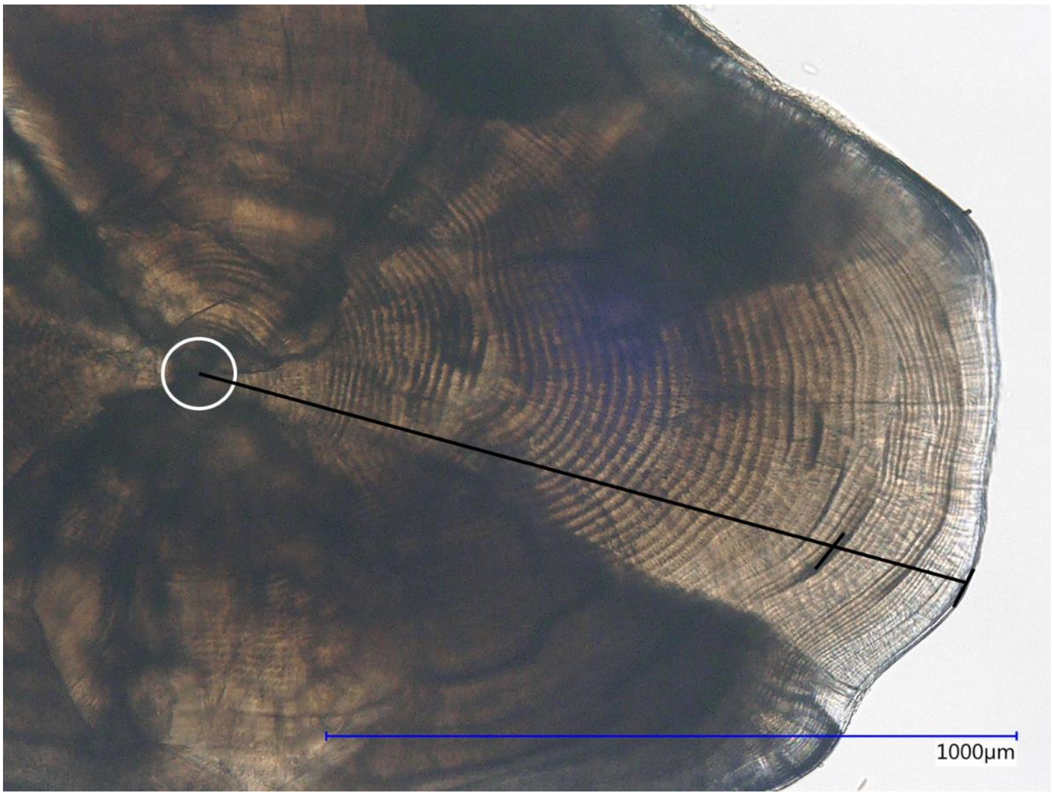


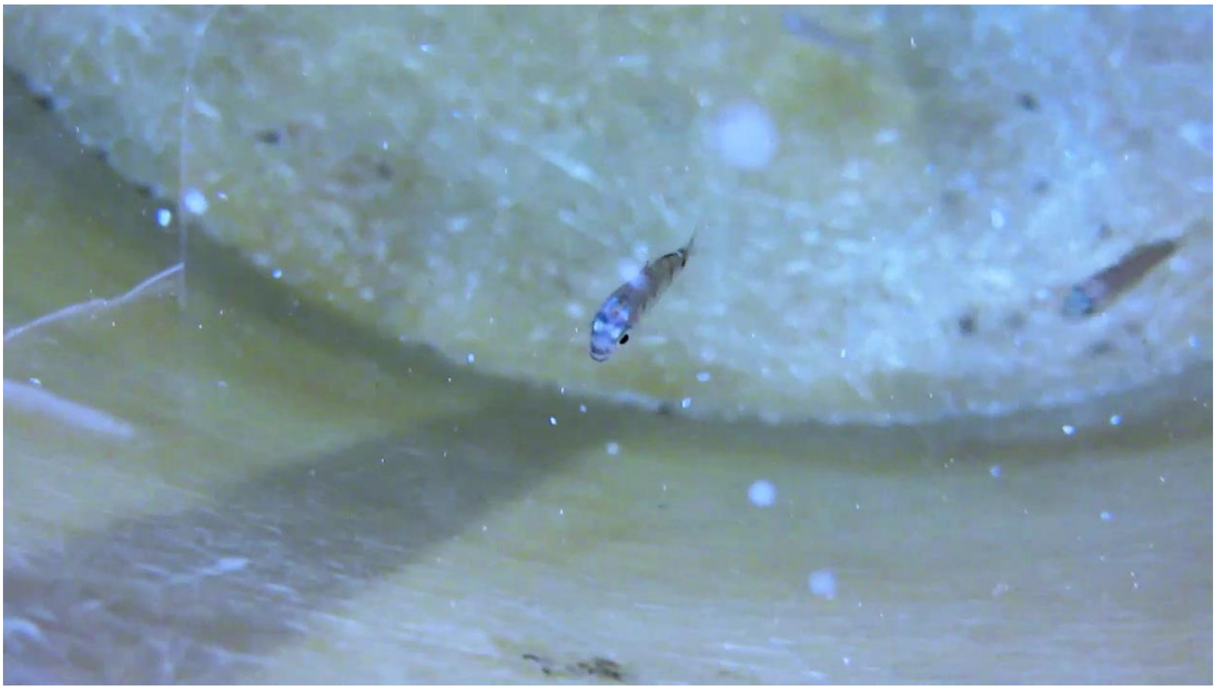
Fig. S3 Photo of a left otolith of a juvenile *Diplodus sargus*, highlighting exemplarily the respective measurements taken from the core of the otolith (encircled in white) to the first day of the experimental phase (black division marks) as well as to the outer margin. The photo does not display the actual measurements, as these were taken from the core to the tip of the rostrum for all fish.

Condition based on RNA:DNA analysis

The concentration of nucleic acids (RNA, DNA) was quantified for each juvenile fish using a microplate fluorescent assay (Caldarone et al. 2001; Wagner et al. 1998). The extraction of nucleic acids from juveniles involved the mechanical (sonication: 3 pulses at 50 A during 1 minute) and chemical homogenization of tissues (cold sarcosyl Tris-EDTA extraction buffer), the addition of Gel Red (GR) (a nucleic acid fluorochromedye), and subsequent fluorescence-photometric measurements. Fluorescence was measured on a microplate reader (Biotek Synergy HT model SIAFRTD – BioTek® Instruments, Inc., Vermont, USA) with an excitation wavelength of 365 nm and an emission wavelength of 590 nm. Following the first scan to determine total fluorescence of RNA and DNA, a ribonuclease A (Type-II A) solution was added to each well, and the concentration of DNA was calculated directly from the standard curve. The RNA fluorescence was calculated by subtracting the DNA fluorescence (second scan) from total fluorescence (first scan). Concentrations were determined by running standard curves of DNA-GR and RNA-GR every day with known concentrations of λ-phagus DNA (0.25 $\mu g\ \mu l^{-1}$) and 16S–23S E. coli RNA (4 $\mu g\ \mu l^{-1}$) (Roche). The average ratio of DNA and RNA slopes (average ± SD) was 3.56 ± 0.59. The RNA/DNA ratios were standardized (sRD) using DNA and RNA slope ratios and the reference slope ratio of 2.4 (Caldarone et al. 2006).

Results

Electronic supplement: video sequence plus description



Video 1 Juvenile white seabream *Diplodus sargus* in one of the experimental tanks of the microplastic treatment groups, taking up a blue polystyrene particle and rejecting it immediately afterwards.

Relationship between MP uptake and ingestion of natural prey items

Table S2 Results of the statistical evaluation (***P***-values of the Tukey's test for multiple comparisons of means) of the gastrointestinal tract (GIT) fullness of fish that were taken out of the trial two days prior to the final end of the trial (03.05.2018) and the remaining half of the fish which was used until the final day of the trial (05.05.2018). Analysis of variance (ANOVA: One-way analysis of means – not assuming equal variances: ***P*** = 0.01, df = 9) showed a significant difference between the two measurements, thus the detailed post-hoc analysis was run to detect potential differences in the relevant groupings. Relevant comparisons are listed for fish of the control group (ART), individuals of the pristine plastic treatment (ART+NP) and biofilm-coated plastic treatment (ART+OP) that were found with no MP particles in their GIT at the end of the trial, well as MP-feeding individuals of both plastic treatments (ART+NP *MP ingest;* ART+OP *MP ingest*). Significant differences (***P*** ≤ 0.05) are highlighted with '*'.

Second to last day (03.05.2018)	Final day (05.05.2018)	***P*** Tukey's test
ART	ART	0.998
ART+NP	ART+NP	0.381
ART+NP *MP ingest*	ART+NP *MP ingest*	0.999
ART+OP	ART+OP	0.024 *
ART+OP *MP ingest*	ART+OP *MP ingest*	0.984

Condition based on morphometrics: Fulton's K

Table S3 Results of the statistical evaluation (***P***-values of the Tukey's test for multiple comparisons of means) of the juvenile fish condition based on Fulton's condition factor *K* calculated for a subset of fish. Analysis of variance (ANOVA: One-way analysis of means – not assuming equal variances: ***P*** < 0.001, df = 4) was statistically significant, thus the detailed post-hoc analysis was run to detect potential differences in the groupings. Relevant comparisons are listed for fish of the control group (ART), individuals of the pristine plastic treatment (ART+NP) and biofilm-coated plastic treatment (ART+OP) that were found with no MP particles in their GIT at the end of the trial, well as MP-feeding individuals of both plastic treatments (ART+NP *MP ingest;* ART+OP *MP ingest*). Significant differences (***P*** ≤ 0.05) are highlighted with '*'.

	ART	ART+NP *MP ingest*	ART+NP	ART+OP *MP ingest*
ART+NP *MP ingest*	0.039*			
ART+NP	0.679	0.525		
ART+OP *MP ingest*	< 0.001*	0.522	0.029*	
ART+OP	0.846	0.451	0.999	0.026*

Discussion

Laboratory set-ups testing ELHS of marine fish for MP uptake

Considering only the MP densities, which are either given in particle numbers and show a range of 100 MP l^{-1} (de Sá et al. 2015), over 1000 MP l^{-1} (Hoss and Settle 1990) to 1 x 10^6 MP l^{-1} (Yin et al. 2018), or are given in weights, ranging for instance from 18.4 µg l^{-1} (Oliveira et al. 2013) to 0.216 mg l^{-1} (Luís et al. 2015), the key variable in MP feeding experiments is hardly comparable across trials (see tab. S4).

Besides the actual application of MP particle numbers per unit volume and independent of their condition (i.e., pristine or biofilm-coated), the objectives of laboratory MP ingestion studies also vary greatly and thereby the methodological approaches as well as the outcomes: in bioassays, performed to assess behavioural or physiological impacts of acute MP exposure (e.g., de Sá et al. 2015; Luís et al. 2015; Ferreira et al. 2016b; Barboza et al. 2018a), trial duration rarely exceeded 96 hours. Moreover, the actual MP uptake in terms of frequency of occurrence or MP particle counts per individual were not necessarily recorded in those bioassays. Only few recent studies were conducted over a duration of at least two weeks with juvenile marine fish species (i.e., Mazurais et al. 2015; Critchell and Hoogenboom 2018; Ašmonaitė et al. 2018; Naidoo and Glassom 2019). Frequently, inert food pellets were given along with MP particles which partially counteracts the natural feeding mode of the fish taxa and life stages under investigation and makes a comparison across laboratory studies or between laboratory and field studies questionable. In several experimental set-ups, fish were furthermore starved prior to the actual trial phase (e.g., de Sá et al. 2015; Ory et al. 2018b) or during the trial (e.g., Barboza et al. 2018a) which might also prevent the thorough evaluation of the uptake of MP.

In relation to the various objectives and laboratory set-ups applied in previous studies, the total number of individuals tested differs up to three orders of magnitude (see sample size in Table S4). The total number of individuals used in bioassays is usually lower than in other MP-related experimental designs. Furthermore, the number of fish tested simultaneously in the same experimental tank varies: in bioassays set-ups fish are frequently tested individually or in small groups of less than 10 individuals per tank (see e.g., de Sá et al. 2015; Luís et al. 2015; Critchell and Hoogenboom 2018). In several publications (e.g., Colton et al. 1974; Hoss and Settle 1990) no precise statement of the number of fish per tank is given though feeding behaviour may vary with group size. Summarizing all of the above, it becomes obvious that drawing comparisons across MP-feeding trials is hardly expedient and informative value may be limited.

Table S4 Overview on the experimental designs of microplastic feeding trials conducted with early life-history stages of marine fish taxa (incl. freshwater taxa known to occur in brackish habitats). No conversions were made on microplastic density or trial duration in order to highlight the variability in set-ups. The entry ‘n/a’ (not available) indicates the lack of information to a specific category. This table does not take into account the detailed objectives of individual studies, according to which the respective laboratory set-up was designed.

Species	**Sample size**	**No of fish per trial tank**	**Polymer type and size**	**Pristine / biofilm**	**Density**	**Prey type**	**Duration**	**Uptake (FO%)**	**MP / Ind**	**Objective**	**Reference**
Acanthochromis polyacanthus	112	4	PET particles, 1 – 2 mm	biofilm	0.021 – 0.86 mg l^{-1}	Food pellets	1 weeks	19.6	1 – 8	Growth, condition, behaviour	Critchell & Hoogenboom 2018
Acanthochromis polyacanthus	69	3 – 4	PET particles, 125 – 5000 µm	biofilm	0.025 – 0.1 mg l^{-1}	Food pellets	6 weeks	20 – 100	5 – 2102	Growth, condition, behaviour	Critchell & Hoogenboom 2018
Ambassis dussumieri	450	5 – 10	Mixture of PE, PVC, PE, PS, 250 – 1000 µm	pristine + biofilm	0.01 g per 20 l	Flake food	95 days	31 – 93	0.5 – 29.35	Growth, survival	Naidoo & Glassom 2019
Brevoortia tyrannus	n/a	n/a	PS microspheres, 100 – 500 µm	biofilm	200 - 1000 MP l^{-1}	none	10 minutes – 19 hours	0	n/a	MP uptake, impacts	Hoss & Settle, 1990
Dicentrarchus labrax	Initial Larval stock: 37,800; MP analysis: 1440	2100	PE microbeads, 10 – 45 µm	pristine	1.2 – 12 mg g^{-1} diet	Food pellets	37 days	35 – 68	1.4 – 3.3	MP uptake, growth, inflammatory response	Mazurais et al. 2015
Dicentrarchus labrax	81	1	Microspheres, 1 – 5 µm	pristine	0.26 – 0.69 mg l^{-1}	none	96 hours	n/a	n/a	Swimming performance, behaviour	Barboza et al. 2018a
Dicentrarchus labrax	162	n/a	PVC pellets,	pristine + ‘polluted’	Diets with 0.1 % (w/w) plastic	Food pellets	90 days	n/a	n/a	Intestinal alterations	Peda et al. 2016
Fundulus majalis	n/a	n/a	PS and acrylonitrile-butadiene-styrene spherules	n/a	43500 MP per 0.04 m^3	Chopped squid	8 weeks	0	0	uptake + potential effects	Colton et al. 1974
Gasterosteus aculeatus	n/a	n/a	PS and acrylonitrile-butadiene-styrene spherules	n/a	43500 MP per 0.04 m^3	Chopped squid	8 weeks	0	0	uptake + potential effects	Colton et al. 1974
Lagodon rhomboides	n/a	n/a	PS microspheres, 350 – 500 µm	biofilm	200 - 1000 MP l^{-1}	none	10 minutes – 19 hours	15	n/a	MP uptake, impacts	Hoss & Settle, 1990

Species	**Sample size**	**No of fish per trial tank**	**Polymer type and size**	**Pristine / biofilm**	**Density**	**Prey type**	**Duration**	**Uptake (FO%)**	**MP / Ind**	**Objective**	**Reference**
Leiostomus xanthurus	n/a	n/a	PS microspheres, 350 – 500 µm	biofilm	200 - 1000 MP l^{-1}	none	10 minutes – 19 hours	15	n/a (max. 45 MP)	MP uptake, impacts	Hoss & Settle, 1990
Leiostomus xanthurus	24	n/a	Plastic microspheres	n/a	1000 MP l^{-1}	Artemia spp.	10 days	25	n/a	Mortality	Hoss & Settle, 1990
Melanogrammus aeglefinus	n/a	n/a	PS and acrylonitrile-butadiene-styrene spherules	n/a	43500 MP per 0.04 m^3	fresh plankton	2 weeks	0	0	uptake + potential effects	Colton et al. 1974
Microgadus tomcod	n/a	n/a	PS and acrylonitrile-butadiene-styrene spherules	n/a	43500 MP per 0.04 m^3	Chopped squid	8 weeks	0	0	uptake + potential effects	Colton et al. 1974
Mugil cephalus	n/a	n/a	PS microspheres, 210 – 350 µm	biofilm	200 - 1000 MP l^{-1}	none	10 minutes – 19 hours	75	n/a (max. 45 MP)	MP uptake, impacts	Hoss & Settle, 1990
Mugil cephalus	20	n/a	Plastic microspheres	n/a	1000 MP l^{-1}	Artemia spp.	10 days	0	0	Mortality	Hoss & Settle, 1990
Oncorhynchus mykiss	48	1	polystyrene powder, 100 - 400 µm	pristine + ‘polluted’	n/a; "Estimated intake of MPs in plastic-containing diets varied from 500 − 700 to 2226 − 2411 particles/ fish/day"	handmade MP-feed-pellets	28 days	n/a	n/a	biological effects (incl. gene expression and enzymatic measurements)	Ašmonaitė et al. 2018
Paralichthys lethostigma	n/a	n/a	PS microspheres, 210 – 250 µm	biofilm	200 - 1000 MP l^{-1}	none	10 minutes – 19 hours	6	n/a	MP uptake, impacts	Hoss & Settle, 1990
Paralichthys spp	n/a	n/a	PS microspheres, 350 – 500 µm	biofilm	200 - 1000 MP l^{-1}	none	10 minutes – 19 hours	0	n/a	MP uptake, impacts	Hoss & Settle, 1990
Pomatoschistus microps	234	1	PE microspheres, 1 – 5 µm	pristine	0.216 mg l^{-1}	None	96 hours	n/a	n/a	Toxic effects in combination with chromium (VI) à predatory performance + biomarkers	Luís et al. 2015
Pomatoschistus microps	126	1	PE microspheres, 420 – 500 µm	pristine	15 or 30 MP per 300 ml	*Artemia franciscana* nauplii	3 minutes	n/a	n/a	MP uptake, predatory performance	de Sá et al. 2015

Species	**Sample size**	**No of fish per trial tank**	**Polymer type and size**	**Pristine / biofilm**	**Density**	**Prey type**	**Duration**	**Uptake (FO%)**	**MP / Ind**	**Objective**	**Reference**
Pomatoschistus microps	72	1	PE microspheres, 1 – 5 µm	pristine	18.4 or 184 µg l^{-1}	none	96 hours	n/a	n/a	Toxic effects in combination with pyrene à biomarkers	Oliveira et al. 2013
Pomatoschistus microps	108	1	Polyethylene spheres, 1 – 5 µm diameter	pristine	max. 0.182 ± 0.030 mg l^{-1}	none	96 hours	n/a	n/a	Combined effects with Au-Nanoparticles and temperature on predatory performance, biomarkers	Ferreira et al. 2016b
Pseudopleuronectes americanus	n/a	n/a	PS and acrylonitrile-butadiene-styrene spherules	n/a	43500 MP per 0.04 m^3	Chopped squid	8 weeks	0	0	uptake + potential effects	Colton et al. 1974
Pseudopleuronectes americanus	n/a	n/a	PS and acrylonitrile-butadiene-styrene spherules	n/a	43500 MP per 0.04 m^3	fresh plankton	2 weeks	0	0	uptake + potential effects	Colton et al. 1974
Sebastes schlegelii	100	10	polystyrene microspheres, diameter 15 µm	pristine	1 x 10^6 l^{-1}	commercial diet, 5% of body weight	14 days uptake, 7 days depuration	n/a	n/a	behaviour, energy reserve and nutritional quality	Yin et al. 2018
Seriolella violacea	33	2	nylon cable ties (1.2 mm length, 1.0 mm diameter), different colours	pristine	10 food pellets and 2 MP; max. 80 pellets and 8 MP	food pellets	max. 20 minutes	88	n/a	ingestion + egestion of MP	Ory et al. 2018b

5 Discussion

Towards a deeper understanding of microplastic uptake in fish

The growing evidence for microplastic (MP) ingestion by marine fish requires an increasing effort to identify both the explanatory variables for MP uptake and the adverse effects on fish condition, growth, and survival across life-history stages and taxa. The main goal of the present thesis was therefore to enhance the understanding of the potential detriments of MP pollution on fish by

i. Identifying the driving factors for and the impacts of MP ingestion by marine fish
ii. Determining the extent of MP exposure and uptake by juvenile fish in a vital coastal lagoon nursery in relation to prey availability and environmental conditions
iii. Investigating the physiological impacts of MP exposure and uptake for juvenile fish.

To achieve the aforementioned objectives, a thorough literature review (Chapter 1: Müller 2021), in combination with a well-grounded, comparative field (Chapter 2: Müller et al. subm.) and laboratory study (Chapter 3: Müller et al. 2020), using an omnivorous fish species, the white seabream *Diplodus sargus* (Linnaeus, 1758) as a model organism, was conducted.

Ninety *in-situ* studies, published between 1972 and 2019, were reviewed to provide a qualitative and quantitative overview on the fish taxa and life-history stages examined for MP uptake. Moreover, the current state-of-the-art was summarised regarding the driving factors for MP ingestion and the available information on the physiological implications verified by field reports which allowed highlighting their diverging outcomes and identify persistent knowledge gaps (Chapter 1: Müller 2021). Despite the fact that ingestion assessments for approximately 500 different marine fish taxa exist, information on ecological parameters (e.g., life-history stage, morphometrics, trophic level, dietary and habitat preferences) and environmental conditions (e.g., abiotic factors, ambient prey and MP abundance) to contextualise the frequency and impact of MP feeding, which is particularly relevant to evaluate MP pollution as a potential threat for fish as a marine living resource, remains scarce (Barboza et al. 2018b; De-la-Torre 2020; Provencher et al. 2020).

In the *in-situ* study of the present thesis, the abovementioned knowledge deficits were addressed by simultaneously assessing the contents of the gastrointestinal tract (GIT) of juvenile white seabream along with environmental parameters, MP abundance as well as prey availability across different sampling sites within a coastal lagoon nursery ground. To holistically examine the potential implications of MP exposure and uptake for the omnivorous ELHS, the findings with juvenile fish condition and feeding preferences were integrated (Chapter 2: Müller et al. subm.). This approach was chosen to address the complex challenge of establishing proofs of a MP ingestion-related impact in field studies which derives from the potentially additive, antagonistic and synergistic effects of different environmental stressors (e.g., temperature, salinity, oxygen availability) acting upon an organism, especially in transitional ecosystems, as well as by the difficulties in verifying both non-lethal and lethal impacts in field studies (Elliott and Quintino 2007; Fonte et al. 2016; McCormick et al. 2020).

Complementing the field study results, an *in-vivo* feeding experiment under controlled conditions was conducted to determine MP uptake rates and reliably allocate potential impacts on juvenile fish survival, growth, and condition (based on morphometrics and nucleic acid ratios) to this anthropogenic stressor in isolation (Chapter 3: Müller et al. 2020). Environmentally relevant MP concentrations of both pristine and weathered polystyrene fragments along with natural prey items were applied, thereby resolving the discrepancies of previously published trials: although laboratory experiments have been primarily used to investigate MP exposure and ingestion-related effects on fish survival, growth, and condition (Wang et al. 2020), the majority of set-ups were rather inconsistent with natural conditions in terms of MP concentrations, applied polymer types and MP shapes (Phuong et al. 2016; Lusher et al. 2017a; Burns and Boxall 2018; McGoran et al. 2018; Cunningham and Sigwart 2019).

In the following discussion, the results featured within the three Chapters of the present thesis will be amalgamated and related to the current state-of-the-art, following the rationale shown in the schematic diagram of Figure 1. In the first part of the discussion (section 5.1), the results obtained on the dimension of MP feeding and its physiological effects on fish populations will be recapitulated. Thereafter, the level of knowledge on the extent of MP uptake by early life-history stages (ELHS) of fish (section 5.2) will be summarised, and the findings connected with an evaluation of the potential detrimental effects of MP-feeding (sections 5.3 and 5.4), as well as with the identification of the driving factors for plastic ingestion (section 5.5). Moreover, the impact of MP pollution on the quality of nursery habitats and thus on recruitment variability will be assessed (section 5.6). This holistic reflection is expected to contribute towards an enhanced understanding of the level and impact of MP pollution on early life-history stages (ELHS) of fish growing up in coastal nursery grounds.

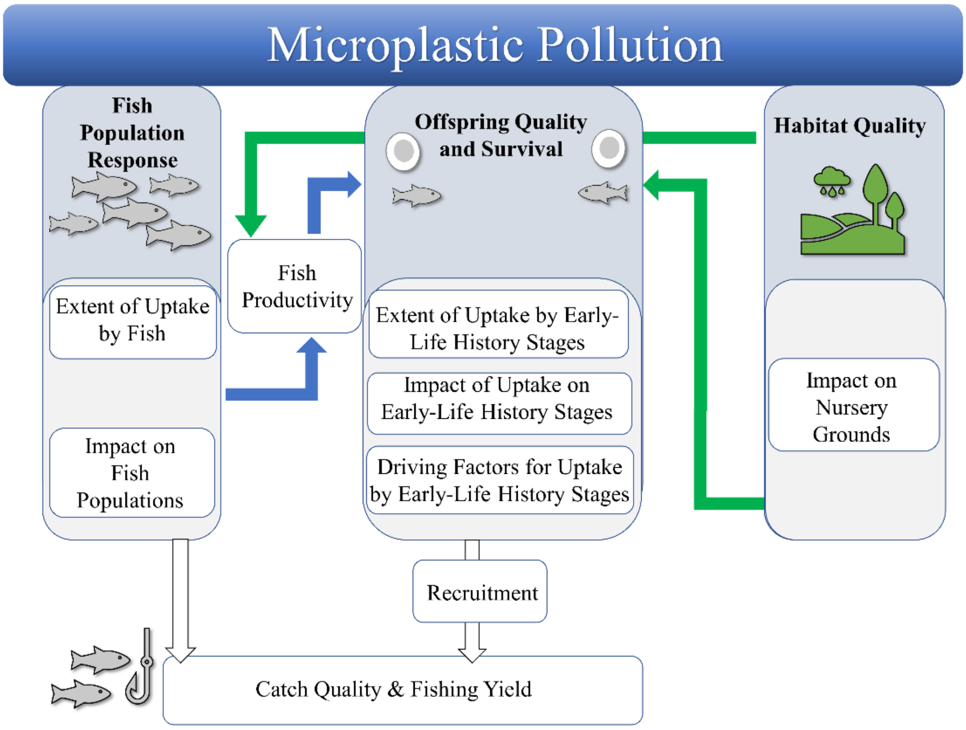


Fig. 1 Schematic diagram showing the rationale of the discussion reflecting on the influence of microplastic pollution on fish as a marine living resource. First of all, the implications of microplastic pollution on fish populations will be assessed (5.1), followed by an evaluation of the extent of MP ingestion (5.2) by early life-history stages of fish, the MP-uptake related impact on offspring survival (5.3 + 5.4), and the driving factors (5.5) for MP-feeding by larval and juvenile fish. Finally, the impact of microplastic pollution on the quality of nearshore nursery grounds will be appraised.

5.1 Extent of microplastic uptake by fish and its impact on fish populations

The rather insufficient proportion of marine and estuarine fish taxa investigated for MP uptake, along with the actually verified extent and impact of ingestion, do not seem to adequately support the increasing scientific and public attention for marine MP pollution and the prevailing concern about the associated detrimental effects for marine biota and ultimately food security (Toussaint et al. 2019; Völker et al. 2019; Cunningham et al. 2020; Provencher et al. 2020). Chapter 1 (Müller 2021) of this thesis showed that MP uptake has been verified for only 327 marine fish species (of approximately 500 fish species assessed). Not only does the present level of knowledge cover a mere 1.5% of all valid fish species described (Fricke et al. 2020; Froese and Pauly 2021), the extent to which a certain fish species ingests MP fibres, fragments or pellets shows furthermore substantial variation across geographical scales and sampling seasons, as well as between scientific studies and the methodologies applied therein (Chapter 1: Müller 2021; Chapter 2; Chapter 3: Müller et al. 2020). The majority of fish taxa considered by previous field studies are of commercial relevance (Chapter 1: Müller 2021), however, just 1/3 of all individuals assessed were found to actually contain MP in their GIT (Markic et al. 2020; Müller 2021). Moreover, the proportion of MP-feeding fish shows substantial variation across individuals of the same species (Chapter 1: Müller 2021), a finding which was also corroborated in the field and in the laboratory study presented herein (Chapter 2: Müller et al. subm.; Chapter 3: Müller et al. 2020), reflecting the intraspecific variation in physiological sensitivity exhibited by natural fish populations which shape their responses to anthropogenic stressors (Johnson et al. 2014; McKenzie et al. 2016). Though outnumbered by records of high MP ingestion rates, studies detecting no MP-feeding individuals or confirming low MP loads in fish GIT have been published in parallel to the earliest investigations (Colton et al. 1974; Liboiron et al. 2016, 2018, 2019; Ryan et al. 2019), indicating an unequal interaction of fish of different age groups and species with this anthropogenic pollutant. Phenomena such as MP post-capture ingestion / net feeding (Lancraft and Robinson 1980; Lusher et al. 2013) and post-sampling airborne contamination (Davison and Asch 2011; Foekema et al. 2013), along with the snap-shot-issue of GIT analyses (Baker et al. 2014) and species- and life-stage specific GIT retention times (Beer et al. 2018; Markic et al. 2018) add further uncertainties to the thorough evaluation of the actual extent of MP uptake by entire fish populations.

To achieve a holistic evaluation of the actual MP associated hazards for fish stocks, the associated physiological effects of ingestion may be considered more relevant than the actual MP load per fish. Yet, Chapter 1 (Müller 2021) revealed that impact assessments have been infrequently conducted in previous *in-situ* studies: only 11 out of 90 studies under review investigated the potential effects of MP ingestion. Fish morphometrics as a proxy for condition (e.g., Fulton's condition index K, Richardson's condition factor CF) have been most commonly calculated and evaluated either as a consequence of MP ingestion (e.g., Cardozo et al. 2018; Dantas et al. 2019) or as driving factor for it (Compa et al. 2018). The outcomes of both considerations, however, are to date equivocal and several studies failed to verify a statistically significant relation between body condition and MP presence in the GIT (e.g., Foekema et al. 2013; Rummel et al. 2016; Kazour et al. 2018; Arias et al. 2019).

Similarly, results from Chapter 2 (Müller et al. subm.) and Chapter 3 (Müller et al. 2020) indicate that juvenile fish condition (based on Fulton's K) is not significantly compromised provided that the individuals take up < 10 particles or fibres. It was shown that these MP loads did neither critically inhibit uptake of natural prey items nor significantly reduce growth, condition, and survival rates in juvenile white seabream. Depending on the size and shape of the ingested MP, an average load of two micro-sized plastics in around 1/3 of all individuals, as verified in Chapter 1 (Müller 2021), may have relatively minor effects on fish condition and thus population development for specific taxa. It is therefore not surprising that significant detrimental impacts on fish condition have not been verified beyond doubt in previous research initiatives.

Moreover, a drawback of morphometrics-based condition indices may be seen in the comparably slow response of fish growth and weight towards external pressures, potentially limiting these otherwise fast and easy approaches to assess fish condition (Markic et al. 2018; De Vries et al. 2020). Hence, molecular, toxicological as well as histopathological approaches are considered an expedient option to enhance the current understanding of the physiological as well as ecological impacts of MP uptake (Vasconcelos et al. 2009; Ryan 2016; Alomar et al. 2017; Arias et al. 2019). In this sense, a molecular condition index, the standardized ratio of RNA to DNA (sRD) in the fish tissue, reflects the changes in protein synthesis, growth rate, and nutritional condition in response to environmental conditions over shorter periods of time (Bulow 1970; Buckley et al. 1999; Chícharo and Chícharo 2008) and has therefore been applied to complement the morphometrics-based condition index assessed in the laboratory trial of the present thesis (Chapter 3: Müller et al. 2020). The sRD ratio of juvenile white seabream assessed in the *in-vivo* study supported the trends observed on the basis of Fulton's K: the uptake of exceptionally high quantities of solid MP fragments inhibits the intake of sufficient food, which translates into decreasing sRD ratios and thus lower body condition. However, the exposure to realistic MP concentrations along with the ingestion of low or minor numbers of MP, as observed in most MP-feeding individuals, does not detrimentally affect juvenile fish condition and survival over the course of several weeks.

Taking all of the above into consideration, the actual implications of MP availability for fish recruitment, population development and ultimately food security still remain to be clarified, yet media framing of the topic and consequent public perception seem to consider the detrimental effects of MP presence in the marine environment as a proven fact (Toussaint et al. 2019; Völker et al. 2019; Cunningham et al. 2020). Although the impact of a certain stressor or pollutant on an ecosystem-scale is of paramount scientific and public interest, the investigation of the immediate effects has to be realised on an individual and local scale, implying the need for an integration across scales (Cooke et al. 2014; Prinz and Korez 2020). To understand the impact of anthropogenic disturbances such as MP pollution at a population or ecosystem-level, it may not be expedient to consider a fish stock a homogenous group of individuals, as abiotic tolerances, trophic and habitat resource utilisation as well as behavioural adaptations may vary between individuals (Chapter 2: Müller et al. subm.), sexes, and life-history stages, thereby forming the basis for population dynamics and life-history variability (Chambers 1993; Bolnick et al. 2003, 2011; Roswell et al. 2013; Harding et al. 2019). Furthermore, plastic pollution is expected to affect organisms across different biological scales – from the cellular to the population level with subsequent implications arising for the entire ecosystem (Galloway et al. 2017). An in-depth understanding of the potential implications of MP pollution on all biological scales is needed to establish effective risk assessments and legitimately inform the general public and policy-makers alike. Nevertheless, studies at the individual level are considered to be an appropriate starting point as the individual's physiological and behavioural response is a relevant indicator of environmental stressors (Ward et al. 2016; Galloway et al. 2017; Nanninga et al. 2021). Despite the challenges inherent to biological upscaling, research initiatives such as local *in-situ* investigations or laboratory studies - both realised within the present thesis - have been proven applicable approaches to be translated into broader conservation and management-relevant context (Cooke et al. 2014).

5.2 Extent of plastic uptake by ELHS

Upon closer examination of the field studies which considered ELHS in adequate sample sizes (i.e., ≥ 23 individuals per taxon, as defined by Markic et al. 2020), the extent of MP-feeding shows substantial variation across taxa, trophic guilds, marine life-style as well as study sites, ranging from 0% in juvenile carnivorous Horse-eye jack *Caranx latus* (Vendel et al. 2017) to 96% (4.63 MP per fish) in juvenile planktivorous European pilchard *Sardina pilchardus* (Renzi et al. 2019). However, as most studies did not explicitly mention the life-history stage or size (e.g., standard length) of the individuals examined, a precise quantification of MP uptake by ELHS of fish and thus an assessment of the actual threat to recruitment success is not possible based on the current level of knowledge. Chapter 1 (Müller 2021) revealed that MP ingestion has been studied for ELHS of ~ 120 different taxa in the field, with uptake rates shown to differ substantially between fish populations of the same species collected across different geographic regions. Consequently, it seems advisable to initially evaluate the level of MP interference on local populations across small spatio-temporal scales as realised in Chapter 2 (Müller et al.

subm.) and carefully compare results between studies under consideration of different methodological approaches and ambient conditions (Markic et al. 2020).

In the case of the juvenile white seabream, the proportion of MP-feeding individuals varied with sampling site between 13.64 – 31.03% and the mean number of MP per MP-feeding individual was 1.64 ± 1.04, with a maximum of 5 MPs (> 150 µm) detected in a single individual (Chapter 2: Müller et al. subm.). In comparison, the proportion of MP-feeding individuals in the laboratory trial (Chapter 3: Müller et al. 2020) ranged between 9.8 – 17.65%, depending on MP treatment group. Although fewer individuals in the laboratory trial fed on MP, the mean MP load per MP-feeding individual was higher than in the field study (8.95 ± 19.23), as was the maximum MP uptake in a single individual which amounted to 78 polystyrene particles (500 – 1000 µm), which may be a result of the comparably deficient availability of different natural prey items in the laboratory in comparison to the diverse food supply in the Ria Formosa lagoon. The variability in plastic-feeding rates across studies is further validated when expanding the comparison of MP uptake by *D. sargus* beyond the two studies presented herein by another study from an urban harbour on the Mediterranean coast: Shabaka et al. (2020) found a feeding incidence of 100% in juvenile *D. sargus* (n = 40) with high average loads of 3593 ± 3985 MP per individual.

Reasons for the large intraspecific variation may lie within the different methods used for MP investigations. The methodological procedures to assess MP in environmental samples currently lack formally adopted, harmonised sampling protocols and gears, common methodologies for extraction and identification (which may vary in their suitability across different matrices), as well as reporting units for quantification (Hidalgo-Ruz et al. 2012; Lusher et al. 2017b; Hermsen et al. 2018; Koelmans et al. 2020). Moreover, the heterogeneity of MP particles in e.g., size, shape, polymer composition (and thus density) or colouration further impedes integrated assessments and comparisons across broader scales (Galgani et al. 2013; Lindeque et al. 2020). In principle, the visual sorting and quantification of MP either by naked eye or by stereo- / dissecting microscopy as applied in Chapter 2 (Müller et al. subm.) is a widely used methodology due to its easy and relatively fast application (Shim et al. 2017; Lusher et al. 2020). However, this method also entails the risk of misidentification, especially of smaller-sized MP, wherefore preceding sample treatment (e.g., digestion of organic materials) or subsequent verification by spectroscopic or thermo-analytical techniques are commonly employed (Lusher et al. 2020).

As the scope of the present thesis was to provide a contextualisation of MP availability and uptake by omnivorous juvenile fish, the digestion of the organic material both in environmental samples and in the fish GIT was not considered a viable option for the field study (Chapter 2: Müller et al. subm.). To minimize the problem of false-positive identification of MP particles, strict measures for contamination control along with coherent, conservative classification of micro-sized plastics were applied according to the guidelines provided by Lusher et al. (2013, 2017b, 2020). Therefore, the results obtained by the field study (Chapter 2: Müller et al. subm.) constitute an adequate representation of the MP ratios across different matrices without claiming to represent a 100% precise quantification of MP abundances in plankton, benthos, and biota samples. In contrast, the aforementioned study from Egypt (Shabaka et al. 2020) used optimized visual techniques in combination with diffraction scanning calorimetry for MP identification, thereby being able to define polymer characteristics and set the lower detection limit to < 25 µm (as compared to the minimum size > 150 µm applied in the present field study), which is one conceivable explanation for the differences in MP uptake verified by the different studies.

The meaningfulness of comparisons between different studies depends, on the one hand, on the methodologies applied therein to identify and quantify MP in fish GIT (Lusher et al. 2017b; Markic et al. 2020). On the other hand, local conditions, ontogenetic dietary shifts, along with site-, season- and individual-specific resource utilisation patterns may cause differences between MP ingestion rates (Chapter 2: Müller et al. subm.). Eastern Harbour (Egypt) was found to be heavily polluted by a variety of marine plastics (Shabaka et al. 2019) and white seabream (along with other fish taxa examined) showed higher MP loads than verified by the present study (Shabaka et al. 2020). Although the actual geo-hydrodynamic and ecological characteristics of the Egyptian harbour were not described in the publications, upon comparison of the satellite images of the two areas (Fig. 2) it can be anticipated that local conditions (e.g., input sources, hydrodynamics, tidal range, anthropogenic usage) differ considerably between the two study sites and may therefore cause variation in MP encounter and uptake rates as described in more detail in the sections 5.5.1 and 5.6.

Consequently, the variability in local conditions along with the currently available level of information do not allow for generalised statements on the vulnerability of certain life-history stages, stocks, or taxa towards MP ingestion. The existing perception of a ubiquitous interference of this anthropogenic pollutant with fish as a living resource needs to be reconsidered on the basis of local monitoring campaigns across small spatio-temporal scales (Galgani et al. 2013; Vandermeersch et al. 2015; Bonanno and Orlando-Bonaca 2018).

Fig. 2 Comparison of the two study sites on microplastic uptake by white seabream, *Diplodus sargus* (Shabaka et al. 2020; Chapter 2). Left satellite image: Eastern Harbour, Alexandria (Egypt). Right satellite image: Ria Formosa lagoon (Portugal). Scale bar = 1 km. Map data © Aerodata International Surveys, CNES / Airbus, IGP/DGRF, Landsat / Copernicus, Maxar Technologies (2021).

5.3 Impact of plastic uptake for ELHS - a matter of quantity, size, and shape

The bioavailability of primary and secondary MP debris may affect the physiological condition and survival rates of ELHS of fish, depending on the size relation between the organism affected and the particle encountered (Lusher 2015), the feeding habits and GIT anatomy (Dos Santos and Jobling 1991; Jabeen et al. 2017; McNeish et al. 2018), particle shape and size (Neves et al. 2015; Roch et al. 2021), the polymer composition and additive leaching (Lithner et al. 2011; Pannetier et al. 2020), weathering and associated biofilm-formation as well as absorbance of pathogens (Viršek et al. 2017; Michels et al. 2018) and persistent organic pollutants (POPs) (Gassel et al. 2013; Bakir et al. 2014).

Among the scientifically verified impacts of MP ingestion, the physical blocking or mechanical injuring of the digestive system can be considered the most immediate one, with implications arising particularly for ELHS due to their body: MP size ratio and enhanced vulnerability to environmental and anthropogenic stressors (Boehlert and Mundy 1988; Cole et al. 2013; Lusher 2015; Simpson et al. 2016; Foley et al. 2018). As has been demonstrated in Chapter 3 (Müller et al. 2020), accumulating MP in the GIT of a fish impairs efficient food intake and digestion, potentially leading to false satiation, decreasing growth rates and deteriorating physiological condition which may ultimately entail the risk to increase mortality rates and reduce recruitment success (Fig. 1) (Lusher et al. 2013; Wright et al. 2013; Mattsson et al. 2015; Jovanović 2017; Jabeen et al. 2018; McCormick et al. 2020).

According to Planes et al. (2009), up to 80% of *D. sargus* individuals may be expected to die within the first four months after settlement to their nursery grounds, highlighting the substantial mortality rates recorded for ELHS caused by a multitude of interrelated abiotic and biotic factors (Houde 1987, 1997; Cowan and Shaw 2002). In the case of the juvenile white seabream, however, MP exposure and uptake alone do not seem to have a significant repercussion on population dynamics, as the majority of fish both in the field and laboratory study presented herein did not contain any MP particles or fibres in their GIT (Chapter 2: Müller et al. subm.; Chapter 3: Müller et al. 2020). Although MP-feeding individuals had a significantly lower body condition (Fulton’s K) in comparison to fish of the control group which were fed exclusively with natural prey items, MP exposure and uptake during the 3.5 week-long laboratory experiment did not affect survival rates (Chapter 3: Müller et al. 2020). Furthermore, most MP-feeding fish both in the *in-situ* and *in-vivo* part of the study ingested ≤ 5 MP while GIT fullness with natural prey varied between 0 – 100% indicating the natural variability in food intake (Leitão et al. 2007). Fish from the field study (Chapter 2: Müller et al. subm.), which generally showed lower MP loads per MP-feeding individual than fish from the laboratory study, exhibited no signs of impaired body condition (Fulton’s K). Overall, only those individuals with exceptionally high MP uptake rates of > 10 particles had lower GIT fullness with natural prey as well as lower body condition (Fulton’s K, sRD) as compared to both the control group and MP-non-feeding individuals of their respective MP treatment group (Chapter 3: Müller et al. 2020).

It remains questionable if MP availability in itself and ingestion of minor quantities have the potential to induce significant fluctuations in recruitment success and stock development beyond natural variability (Fig. 1). Apart from the actual MP load, the combination of size and shape of the ingested plastics influences the GIT retention time as well as the accumulation potential, hence, the immediate and long-term ecotoxicological implications for ELHS will vary with MP size and shape (Dos Santos and Jobling 1991; Wright et al. 2013; Rebelein et al. 2021). The poorer body condition detected in fish from the laboratory trial may be directly related to the uptake of higher numbers of solid polystyrene fragments (500 – 1000 µm) in comparison to fish from the field study which predominantly ingested MP fibres instead of particles. While the length of an ingested fibre can be greater than the size of an ingestible particle, its potential to cause any of the abovementioned immediate harmful effects (e.g., GIT blockage, mechanical injuries) may be lower due to its elasticity and smoothness (Grigorakis et al. 2017; Hu et al. 2020). Furthermore, the translocation potential of MP fibres is expected to be minor as a consequence of its size and elongated dimension (Gouin et al. 2011; Su et al. 2019a; Garrido Gamarro et al. 2020). Nevertheless, laboratory experiments on MP fibre ingestion also revealed histological alterations in the GIT along with implications for metabolism and reproduction in certain fish species (Jabeen et al. 2018; Fackelmann and Sommer 2019; Qiao et al. 2019). A recent study on ELHS of Japanese medaka *Oryzias latipes* furthermore identified larvae to be more susceptible to alterations in gene expression and gut microbiota composition upon exposure to environmentally relevant polyethylene microfibre concentrations, with long-term implications potentially arising for digestive function and metabolism (DiBona et al. 2021). Smaller-sized plastics, moreover, have been shown to pass the intestinal mucosa, thereby entering the circulatory system of a fish, and being translocated to muscle tissues or other internal organs such as the liver (Avio et al. 2015; Collard et al. 2017a; 2018; Yin et al. 2018; Roch et al. 2020). Long-term effects such as inflammatory responses, disruption of the immune system and alteration of the metabolic profiles have furthermore been associated with MP ingestion (Mattsson et al. 2015; Greven et al. 2016; Lu et al. 2016).

On the basis of the present thesis, the physiological consequences of smaller micro- or nanoplastics cannot be evaluated. Although the study on white seabream from the Egyptian harbour (Shabaka et al. 2020) detected smaller-sized MP (≤ 25 µm and ≤ 500 µm) in the GIT, an impact assessment or contextualisation with natural prey items was not conducted, which limits the explanatory power of the respective results. In-depth assessments on different MP sizes and shape related impacts are advisable also for fish taxa, such as the white seabream, which generally seem to be able to discriminate between edible and non-edible prey items of a certain size spectrum (Chapter 3: Müller et al. 2020) and may nevertheless be susceptible to accidental and secondary uptake of micro- or nanoplastics (Barría et al. 2020; Gu et al. 2021). Moreover, the extent to which the bioavailability of MP of different size classes and shapes in combination with additive anthropogenic stressors such as habitat degradation, climate change, trophic resource alteration and shortages may eventually result in lower survival rates of different life-history stages (Fig. 1) requires further comprehensive experiments (Ferreira et al. 2016b; Kratina et al. 2019; Cunningham et al. 2020; Rebelein et al. 2021).

5.4 Impact of plastic uptake for ELHS – beyond size dimensions

Due to its physico-chemical properties, marine MP represents a suitable substratum for biofilm-formation (Harrison et al. 2011; Oberbeckman et al. 2015), the odour of which has been shown to induce foraging behaviour in zooplankton and fish (Savoca et al. 2017; Vroom et al. 2017). The large surface-area-to-volume ratio of the hydrophobic MP facilitates not only microbial colonisation but also the transport, accumulation, and bioavailability of contaminants such as persistent organic pollutants (e.g., polychlorinated biphenyls (PCBs), dichlorodiphenyltrichloroethane (DDT)) or trace metals (Mato et al. 2001; Holmes et al. 2012; Zhang et al. 2015). The combined effects of these adsorbed microorganisms, toxins, and metals together with the polymeric composition and chemical additives are of particular environmental concern as they entail the ability to, firstly, bioaccumulate in aquatic organisms once the MP particle is ingested, and, secondly, to biomagnify along the food chain (Teuten et al. 2007; Rochman et al. 2013; Hahladakis et al. 2018).

Despite this observation, the majority of laboratory exposure experiments used pristine plastic particles in their set-up (Phuong et al. 2016). In the present *in-vivo* study (Chapter 3: Müller et al. 2020), both pristine and weathered MP were applied in separate treatment groups: against initial expectation, the uptake of pristine MP was higher than the uptake of weathered MP. Although it was beyond the scope of the present thesis to chemically and microbiologically examine the MP detected in the field study as well as the biofilm-formation on the polystyrene particles deployed in the laboratory trial, it may be concluded that biofilm-coating does not necessarily induce MP uptake in ELHS of fish (Chapter 3: Müller et al. 2020), implying a potential repellent effect of certain biofilm components worth investigating in the future. Furthermore, fish from the biofilm MP treatment group showed a lower body condition (Fulton's K, sRD) and reduced growth in comparison to fish of the control group and the pristine MP treatment group, which supports the hypothesis of a detrimental effect of biofilm-coating on fish condition.

Based on the current state-of-the-art, microorganisms, chemical contaminants and heavy metals (individually or in combination with MP particle exposure) have the potential to impede crucial physiological processes and may lead to reduced larval abundance (Waring et al. 1996), inhibit hormone synthesis causing morphological, developmental and reproductive issues (Talsness et al. 2009), promote behavioural and metabolic changes (Kienle et al. 2008; Nematdoost Haghi and Banaee 2017; Barboza et al. 2018a), induce hepatic stress (Rochman et al. 2013), increase cases of deformity (Hu et al. 2009), and ultimately decrease growth and survival rates (Bervoets and Blust 2003). Future risks assessments should therefore be encouraged to analyse the chemical properties of MP along with the taxonomic composition of the microbiological colonisation to advance our comprehension of the ecotoxicological hazards of ingestion for ELHS and thus of recruitment variability (Fig. 1) (Koelmans et al. 2017; Lohmann 2017).

5.5 Driving factors and explanatory variables

In this part of the discussion, the validity of existing hypotheses on the factors influencing MP ingestion will be discussed in the light of the findings of this thesis.

5.5.1 Environmental availability and inter-individual variability in trophic resource utilisation

Juvenile fish are generally assumed to adopt an opportunistic / generalist feeding strategy, implying that prey uptake is simply a matter of prey availability (Sá et al. 2006a; Nunn et al. 2012; Selleslagh and Amara 2015) Accordingly, the omnipresence of MP both in environmental matrices and in fish GIT may lead to the conclusion that MP ingestion is directly associated to the actual concentrations in the surrounding environment (Wright et al. 2013). However, only few studies have actually contextualised the GIT content with the environmental availability of the respective natural prey items (e.g., Gamito et al. 2003; Pasquaud et al. 2010) – a similar lack of contextualisation can be identified in the current body of literature on MP ingestion by fish (Chapter 1: Müller 2021). As demonstrated by Chapter 1 (Müller 2021), although the correlation between bioavailability of MP and uptake rates in fish has only been investigated in a limited number of *in-situ* investigations, without a contextualisation of environmental MP abundances and MP in the fish, it remains largely unclear if differences in MP presence in the GIT reflect certain dietary preferences or differences in MP abundance.

Chapter 2 (Müller et al. subm.) and Chapter 3 (Müller et al. 2020) evidenced the high inter-individual variability in trophic resource utilization in omnivorous juvenile white seabream, a phenomenon potentially neglected by scientific studies in the past (Roswell et al. 2013; Nanninga et al. 2020). Although Chapter 2 (Müller et al. subm.) revealed highest MP-feeding proportions in omnivorous juvenile fish collected at sampling sites which also showed the highest MP concentration per m^3 during the summer sampling in 2018 in a temperate coastal nursery ground, MP uptake was not significantly correlated to sampling site or to sampling campaign. While some previous studies detected a correlation between sampling sites with high MP abundances and elevated ingestion incidences in fish (e.g., Güven et al. 2017), others did not observe the same pattern (e.g., Lusher et al. 2016; Collicutt et al. 2019; Bosshart et al. 2020). Likewise, fish GIT content did not evenly mirror MP categories detected in environmental samples of previous field investigations: the fish either showed a selective feeding for certain MP colours and shapes (e.g., Ory et al. 2017; Jensen et al. 2019) or randomly ingested any MP available in the respective habitat (e.g., Steer et al. 2017; Beer et al. 2018; Wieczorek et al. 2018; Collicutt et al. 2019). Comparable to the present findings from Chapter 2 (Müller et al. subm.), a ubiquitous trend between MP bioavailability and probability of ingestion has not been verified beyond doubt in previous research initiatives. Although it is advisable to assess environmental MP abundances to achieve a deeper understanding of the level of MP exposure and uptake, additional factors such as local habitat characteristics or intraspecific habits need to be considered as they may entail a stronger explanatory power.

5.5.2 Marine life-style

Originating from the ubiquitous distribution of MP in the marine realm overlapping with essential fish habitats (e.g., feeding, spawning or nursery grounds), the hypothesis of marine life-style (e.g., pelagic, benthic) as an explanatory variable for MP ingestion has been tested in several previous research initiatives (Neves et al. 2015; Güven et al. 2017; Markic et al. 2018; Zhu et al. 2019a). However, divergent outcomes were found (Chapter 1), with neither horizontal nor vertical fish distribution representing a driving factor for MP ingestion. Results from Chapter 2 showed that benthic juvenile seabream do not feed exclusively on benthic prey items but may utilize also planktic food resources, adding further evidence for the fact that marine life-style alone is not an adequate explanatory variable for elevated MP uptake rates (Avio et al. 2019; Markic et al. 2020). However, a common trait of the genus *Diplodus* is the ontogenetic migration pattern across different marine environments which are known to vary in their contamination with MP particles and fibres, hence, the probability and effect of MP encounter is expected to differ across pelagic larval and benthic juvenile life-history stages. Following the pelagic egg stage, planktonic pre-flexion larval seabream are passively dispersed by hydrographic processes into shallow littoral waters where they colonize as post-flexion larvae in the water column and settle as late-stage larvae by actively following environmental nursery cues before switching to a temporarily philopatric life-style during their juvenile stage (Harmelin-Vivien et al. 1995; Sabates and Olivar 1996; Vigliola et al. 1998; Baptista et al. 2019, 2020).

Ocean surface waters, specifically areas of convergence, have been verified to be important larval fish nurseries in which marine organisms at the base of the food web are aggregated, thereby providing favourable trophic conditions supporting the growth and development of ELHS – however, not only natural prey items are accumulated in these convergence zones, but also buoyant MP (Song et al. 2014; Gove et al. 2019). The extent to which pelagic fish larvae encounter and ingest prey-sized MP during their crucial period of first exogenous feeding along with the adverse effects related to MP uptake by larval fish need further scientific evaluation, particularly under consideration of the potentially disadvantageous body:MP size ratio (Lusher 2015) and the acknowledged relevance of larval fish survival rates for recruitment and population dynamics (Houde 1987, 1997, 2001; Lehodey et al. 2006; Miller and Kendall 2009).

5.5.3 Trophic level

In aquatic ecosystems, trophic level is often a function of morphometrics (e.g., body size) and ecological traits (e.g., feeding strategy) (Pauly et al. 1998; Keppeler et al. 2020). Thus, there is reason to believe in a relation between the trophic level of an organism and the MP ingestion probability due to the pollutant's size dimension and its ubiquitous bioavailability, particularly for smaller-sized, planktivorous biota. A recent review of MP uptake across different taxa and functional groups verified that organisms at the base of the marine food web, such as planktivorous bivalves, crustaceans, or small pelagic fish, representing lower trophic levels, are more prone to elevated MP uptake rates (Walkinshaw et al. 2020). Small pelagic fish, including sardines, sardinellas, anchovies, scads, mackerels, and sand smelts, play a vital ecological role in marine upwelling ecosystems and are of high value for global food security (Pikitch et al. 2012; FAO 2016). Due to their planktivorous feeding mode, which has been shown to be adaptable according to prey size and density (i.e., filter-feeding, particulate feeding), these species are, one the one hand, highly opportunistic feeders, and, on the other hand, potentially vulnerable to the elevated uptake of marine plastic litter (Blaxter and Hunter 1982; Fréon et al. 2005; van der Lingen et al. 2009; Collard et al. 2017b; Covernton et al. 2021).

Following this rationale, ELHS of fish typically occupy lower trophic positions than adult individuals thereby supposedly having a higher vulnerability towards MP uptake (Pepin and Dower 2007; Woodland and Secor 2011; Bernardini et al. 2018; Critchell and Hoogenboom 2018). Although trophic level may not be an explanatory variable for elevated MP uptake rates in fish per se, the underlying feeding strategy along with the body dimensions may be indicative for an increased vulnerability. Both aspects, however, need further scientific verification, by expanding the scope of research towards small pelagic fish in upwelling ecosystems as well as by combining research on different life-history stages of one species, preferably originating from the same geographic area.

Conversely, higher trophic levels may be prone to secondary ingestion via contaminated prey, though the transfer of MP in the food web is not fully understood and no consistent correlation between MP uptake and trophic level has been verified in the case of marine fish species although this taxonomic group is comparatively well studied (Nelms et al. 2018; Markic et al. 2020; Covernton et al. 2021). Holistic field studies such as the one presented in Chapter 2 (Müller et al. subm.) are expected to advance the current knowledge on the extent to which MP particles are intentionally and selectively fed on as mistaken food items and could shed light on the potential relationship between environmental availabilities and MP ingestion (Lusher 2015; Ory et al. 2017; Savoca et al. 2017).

In the case of the juvenile white seabream, the uptake of MP fibres was positively correlated with the uptake of marine vegetation (Chapter 2: Müller et al. subm.), hence an accidental ingestion of fibrous plastic adhering to seagrass and algae can be anticipated (Cozzolino et al. 2020). Moreover, several invertebrate prey items identified in Chapter 2 have been found contaminated with MP, such as Copepoda (Bai et al. 2021), Chironomidae larvae (Silva et al. 2019, 2021), or Bivalvia (Setälä et al. 2016). The relevance of accidental MP uptake along with natural food items or even passive uptake via drinking water containing MP needs further comprehensive evaluation, along with the scale of secondary ingestion of contaminated prey organisms, potentially contributing to high MP or nanoplastic loads in fish GIT with varying ecotoxicological potential (Peters et al. 2017; Chagnon et al. 2018; McNeish et al. 2018; Sun et al. 2019;Roch et al. 2020).

5.5.4 Morphometrics and condition

Comparable to the trophic guild and habitat preferences of a fish, morphometrics and condition have frequently been hypothesized to correlate with elevated MP uptake rates (Boerger et al. 2010; Foekema et al. 2013; Güven et al. 2017; Vendel et al. 2017; McGoran et al. 2018), yet no widespread, explicit trend has been detected (Chapter 1: Müller 2021). Though there is a tendency that higher MP feeding rates are associated to larger body dimensions, the significance of this relationship may be disputed partially on account of the underrepresentation of studies on smaller-sized individuals and ELHS of fish (Chapter 1: Müller 2021). Furthermore, no unambiguous trend has been verified in relation to body condition, and it remains largely debatable whether MP uptake is driven by lower or higher body condition or is causing a certain physiological status (Foekema et al. 2013; Mizraji et al. 2017; Compa et al. 2018; Cardozo et al. 2018).

The juvenile white seabream examined in the present thesis (Chapter 2; Chapter 3: Müller et al. 2020) showed a range in SL between 14 – 62 mm and the uptake of MP was recorded for the full size range of individuals. Moreover, morphometrics (i.e., SL, H, wet weight) and condition factor Fultons' K were not correlated to the uptake of MP in the field study (Chapter 2: Müller et al. subm.). In comparison, Shabaka et al. (2020) who found high MP ingestion rates in white seabream, reported the TL of the fish to vary between 65 – 175 mm. Upon initial assessment, the larger specimens showed higher MP uptake rates, tentatively endorsing the existing hypothesis of size being a driving factor for plastic feeding. However, the ontogenetic shift in diet and individual feeding preferences, along with the aforementioned methodological differences and the local conditions are expected to have a more significant effect on MP ingestion rates than the SL or TL of a fish (Giani et al. 2019; Jensen et al. 2019; Markic et al. 2020; Nanninga et al. 2021).

5.5.6 Ontogenetic dietary shifts and feeding mode

During the critical period when pelagic larvae develop into benthic juveniles, the fish undergo morphological (i.e., dentition, overall body shape) and ecological (i.e., feeding behaviour, swimming abilities) changes, which make them vulnerable to environmental disturbances and could result in variation in recruitment success (Garcia-Rubies and Macpherson 1995; McCormick et al. 2002; Hoey and McCormick 2004). In this respect, ELHS of fish have been considered more vulnerable to MP uptake (Critchell and Hoogenboom 2018) with omnivory being hypothesised to govern higher MP ingestion probabilities (Markic et al. 2020; Shabaka et al. 2020). Chapter 1 (Müller 2021) however revealed that MP uptake does not significantly correlate with feeding mode (i.e., carnivorous, herbivorous, omnivorous, planktivorous.

In support of the findings of Chapter 1, Chapter 2 (Müller et al. subm.) and Chapter 3 (Müller et al. 2020) showed a high inter-individual variability in the uptake of natural and artificial prey, which did not substantiate the prevalent perception of omnivory being an indicator for elevated MP uptake rates. Upon closer examination, the persistence of this hypothesis may generally be challenged as omnivorous species tend to utilize a broad dietary spectrum which leads to higher among-individual variation of both natural and potentially also artificial prey items (Svanbäck et al. 2015). Furthermore, results from Chapter 3 highlighted the ability of juvenile *D. sargus* to differentiate between edible, soft-bodied and inedible, hard-bodied prey items facilitated by their specialised food handling apparatus (Vandewalle et al. 1995). The presence of fibres in the GIT of *D. sargus* was positively correlated to the presence of detritus and algae, to which fibrous MP were incorporated or attached (Chapter 2: Müller et al. subm.), suggesting that taxa feeding on seagrass, algae, or detritus may be prone to accidental ingestion of MP fibres which have been shown to be filtered out of the water column by seagrass leaves or to rapidly aggregate with marine biogenic particles (Goss et al. 2018; Michels et al. 2018; Cozzolino et al. 2020; Seng et al. 2020). As has been demonstrated in Chapter 1 (Müller 2021), herbivorous and detritivorous taxa are currently underrepresented in comparison to other trophic guilds, therefore increased research effort should be devoted to MP ingestion by fish taxa feeding on marine vegetation (Chapter 1: Müller 2021).

Larval fish however may lack the ability to discriminate between natural and artificial food items, as the selection of prey is predominantly based on its ingestible size spectrum along with its accessibility (i.e., insufficient escape mechanism) (Rønnestad et al. 2013). In this respect, small synthetic particles and fibres may represent a convenient target for this early life-history stage, further arguing for the implementation of MP ingestion studies on larval fish both in the field and in the laboratory (Steer et al. 2017; Le Bihanic et al. 2020; Pannetier et al. 2020).

5.6 Impact of plastic pollution on the quality of nursery habitats

Nearshore nursery grounds and their floral and faunal communities face considerable loads of plastic debris, mainly due to the synergistic effects of coastal urbanisation and contaminated river discharges (Browne et al. 2011; Jennerjahn and Mitchell 2013; Hitchcock and Mitrovic 2019). In addition, structurally complex habitats, such as seagrass meadows have been shown to act as a sink for MP particles, thereby potentially amplifying the level of plastic exposure to organisms like ELHS of fish, critically dependent on these micro-habitats (Cozzolino et al.

2020; De los Santos 2021). Despite the elevated plastic input and accumulation potential in coastal zones, tide-dominated areas, like the Ria Formosa lagoon, may have high exchange of water volume with each tidal cycle along with small-scale variation in water residence times; the hydrodynamic properties may therefore affect MP dispersion capacity, ultimately counteracting the MP retention in vital nursery grounds (Águas 1986; Fabião et al. 2016; Velez et al. 2020; Harris et al. 2021). Moreover, both environmental conditions and the intensity of anthropogenic interferences (e.g., level of urbanisation, patterns in land- and coastal usage and resource exploitation, point-sources of pollution, location and efficiency of wastewater treatment plants) may vary along a spatio-temporal gradients coinciding with peaks in human activities during summer, with more pronounced effects detected at the inner parts of the system, a trend observed also in the field study of the present thesis (Newton et al. 2003, 2014; Nobre 2009; Martins 2010; Cravo et al. 2012; Vendel et al. 2017; Chapter 2: Müller et al. subm.). On this account, the interaction of (temporary) philopatric organisms like fish ELHS with MP pollution is also expected to vary within a single lagoon and requires further in-depth investigation across the entire lagoonal system.

Besides hydrodynamic and biological attributes, seasonality (i.e., rainfall, wind) may be considered factors influencing MP input and distribution in vital nursery grounds (Lima et al. 2014; Cheung et al. 2016). The fieldwork present herein (Chapter 2: Müller et al. subm.) was realised during summer months (July / August 2018) when anthropogenic usages are intense along the Algarve coast, while precipitation and thus river runoff is generally lower than during fall, winter, and spring (Newton et al. 2003; Newton and Mudge 2005). As the spawning period of *D. sargus* takes place from December to May off the southern Portuguese coast (Morato et al. 2003), potentially more vulnerable larval (and early juvenile) stages are hence hypothesized to encounter higher MP abundances caused by river runoff during spring than later juvenile stages during summer. Extending sampling campaigns over the course of the year is expected to elucidate the suspected variation in MP exposure across different ELHS and may provide deeper insights into the potential impact on the MP contamination of a coastal lagoon nursery and subsequent variation in recruitment success.

A recent study on personality traits of larval white seabream indicates that habitat preferences may be consistent throughout ontogeny (Baptista 2019). As shown by Chapter 2 (Müller et al. subm.), juvenile white seabream show a preference for nurseries located in the interior regions of the Ria Formosa, thereby partitioning the available habitat and trophic resources with other seabream species across the lagoon over the course of the year (Erzini et al. 2002; Ventura et al. 2015). On the one hand, it may be anticipated that anthropogenic stressors constantly exert higher impacts on particular life-history stages and taxa showing a high site-fidelity for certain micro-habitats, such as larval and juvenile white seabream at interior nurseries in the Ria Formosa. At the periphery of distribution, human-induced plastic pollution may therefore shape recruitment success just like density-independent processes such as abiotic conditions are considered to do (Miller et al. 1991; Rijnsdorp et al. 1992). On the other hand, estuarine species, and potentially also specific life-history stages, may be more resilient towards fluctuations in environmental and anthropogenic stressors (Elliott and Quintino 2007; Teichert et al. 2017).

In summary, the geological, hydrodynamic, geochemical, biological, and anthropogenic conditions of transitional environments influence the MP accumulation potential across spatial and temporal gradients which need to be taken into account in future research initiatives (Rynne et al. 2016; Iglesias et al. 2019). Due to local variability, an evaluation of the ecological implications of MP pollution on vital fish nurseries should carried out on the basis of in-depth studies, including reports on local abiotic and biotic conditions to facilitate comparisons of nursery grounds across geographic regions as suggested in Chapter 1 (Müller 2021).

6 Outlook and Future Perspectives

In this concluding section, future areas of research will be outlined, and recommendations provided on the basis of the results and discussion of the present thesis.

Despite the long history of MP research and the cumulative scientific effort to document MP presence in the oceans, Chapter 1 (Müller 2021) exposed several deficits in the understanding of the explanatory variables for MP uptake along with its detrimental effects on different fish taxa and life-history stages. In light of the results presented herein, ichthyofaunal communities are expected to vary in their vulnerability towards anthropogenic stressors and their ability to cope with plastic pollution, due to their spatio-temporal distribution patterns along with species- and life-history stage-specific feeding modes, as demonstrated for juvenile *D. sargus* in the Ria Formosa lagoon (Chapter 2: Müller et al. subm.; Chapter 3: Müller et al. 2020).

To implement effective monitoring campaigns investigating the impact of MP on biota, as prioritised by scientific initiatives and legislative frameworks alike (including the EU Marine Strategy Framework Directive MSFD 2008/56/EC), particular attention needs to be devoted to the selection of suitable indicator species (Galgani et al. 2014; Wesch et al. 2016b; Fossi et al. 2018; Bray et al. 2019). Besides the previously suggested factors determining the suitability of a species for monitoring campaigns (i.e., large scale distribution, gut length, commercial value, home range, level of plastic ingestion) (Bray et al. 2019), Chapter 1 (Müller 2021) provided recommendations for conducting efficient in-situ assessment of MP uptake in marine and estuarine fish (see "Fieldwork Fundamentals", Fig. 10 in Chapter 1 for details). Results from Chapter 2 (Müller et al. subm.) and Chapter 3 (Müller et al. 2020) complement the aforementioned suggestions by highlighting the importance of

i. Recording local biotic, abiotic as well as anthropogenic parameters to acknowledge their potential to shape the spatio-temporal pattern of MP distribution in relation to local and seasonal effects
ii. Investigating the probability of interaction of marine organisms with prey-sized plastic items by simultaneously assessing environmental availability of natural and artificial prey items along with the feeding biology of specific life-history stages of a species
iii. Considering ontogenetic as well as inter-individual differences in trophic resource utilization and prey handling by integrating sufficient sample sizes of different trophic levels, feeding modes and life-history stages (e.g., lower trophic levels

such as ichthyofaunal ELHS as well as small pelagic fish) in multi-species assessments

iv. Assessing the physiological resilience of certain taxa and life-history stages towards minor MP uptake rates on the basis of sound fieldwork and laboratory trial design
v. Retrospectively analysing MP distributional patterns and ingestion rates in key taxa to understand trends and pathways and ultimately enhance modelled predictions of accumulation zones and taxa at risk
vi. Focusing on the environmental implications of MP pollution in biota-rich, transitional waters such as coastal ecosystems or upwelling regions due to their ecological and economic importance

In the absence of any action reducing plastic pollution in the future, the amount of marine litter will triple by 2052 (in comparison to 2011), and fragmentation of macroplastic items to micro- and nano-sized particles will continue for decades even if the output of plastic waste could be eliminated immediately (Hohn et al. 2020). In light of the continuously increasing fish consumption across developed, developing and least-developed states over the past six decades, coinciding with increasing plastic production and usage, and the lack of proper waste management schemes, the ecological and economic implications especially for low and middle-income countries critically dependent on fish as a marine living resource need intensified scientific attention (Hoornweg et al. 2013; Beaumont et al. 2019; Edelson et al. 2021). While research on small spatial scales, like in the Ria Formosa lagoon (Chapter 2: Müller et a. subm.), is relevant to assess ichthyofaunal responses to anthropogenic stressors locally, studies across larger spatio-temporal scales are needed to holistically understand MP pathways and its ramifications for marine ecosystem functioning. Hitherto, MP research has rarely examined marine upwelling ecosystems, despite their ecological and economical importance that derives from the high primary productivity which supports high biomasses of fisheries resources (Kanhai et al. 2017).

6.1 JPI-Oceans MicroplastiX Project

To address the aforementioned knowledge gaps in relation to the spatio-temporal bioavailability of MP for fish ELHS in transitional ecosystems, the Working Group Fisheries Biology at ZMT / I, will henceforth participate in the JPI-Oceans MicroplastiX research project (https://www.microplastix.org). By using a unique sample collection from two essential eastern Atlantic upwelling areas, the Canary Current and the Benguela Upwelling System, results from this project are expected to provide insights into how MP abundance developed over several decades, which will allow for a more holistic evaluation of this anthropogenically caused stressor of marine ecosystems and the living resources therein.

The combination of different analytical procedures (image analysis with ZooScan, sample digestion, polymer characterisation and quantification using fluorescent staining as well as spectroscopic methods) will allow for a contextualisation of MP abundance in relation to planktic organisms (abundance, taxonomical composition) as prioritised by the present thesis (Chapter 1: Müller 2021; Chapter 2: Müller et al. subm.). As MP are differentially distributed in the water column (Cole et al. 2011; Figueireido et al. 2018), the chosen sample collection of neuston from the surface and plankton from the water column will greatly enhance our understanding on the interaction potential of marine organisms with plastic litter as well as on the effect of coastal upwelling on the distribution of MP in the world's oceans (**Task 1**). Moreover, the study of MP in samples stored in natural history collections or at research institutions like the present one will make an essential contribution towards an in-depth comprehension of temporal trends in MP abundance (**Task 2**), relevant also for modelling of future accumulation and dispersion patterns as envisaged also within the framework of the JPI-Oceans MicroplastiX project (Beer et al. 2018; Modica et al. 2020; Hou et al. 2021). Furthermore, by refining the visual (Nile Red staining and microscopic imaging) and spectroscopic methods for MP analysis, the outcomes of the MicroplastiX project will provide a precise characterisation and quantification of MP particles and fibres, allowing for an enhanced evaluation of the potential ecological detrimental effects caused by MP of different physical (i.e., size, shape) and chemical (i.e., polymer composition, additive incorporation) properties (Browne et al. 2011; Lenz et al. 2015; Shim et al. 2016; Lambert et al. 2017).

To comprehensively appraise both the ecological as well as the economic risk of marine (micro-) plastic pollution, persistent knowledge gaps in relation to the potential detrimental effects of this anthropogenic stressor for ichthyofaunal ELHS, especially in transitional ecosystems, need to be addressed just like the potential implications for commercially valuable fish taxa (Gregory 2009; Kanhai et al. 2017; Thushari and Senevirathna 2020; Chapter 1: Müller 2021). On these grounds, we will investigate the MP uptake by ELHS of some of the key species in upwelling food webs (**Task 3**), namely small pelagic Clupeiformes (i.e., sardines and anchovies), which are important economic resources for both human consumption and fishmeal and -oil production (Cury et al. 2000; Fréon et al. 2005; Alder et al. 2008; Pikitch et al. 2012), as well as advancing research on the larval stages of seabream, thereby carrying the work of the present thesis forward.

7 References

(* = qualitatively and quantitatively analysed for Chapter 1: Müller 2021)

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8 Annexes

Annex 1

Table 1 Data base on microplastic (MP) uptake by marine and estuarine fish taxa, established by reviewing 90 publications on field studies conducted between 1972 and 2019. Fish taxa entries are given in alphabetical order. Information on fish species (including the common and family name) are given, along with the trophic guild, habitat category, their potential commercial importance to fisheries as well as the IUCN threat status – entries were made based on the accessed information available on FishBase (Froese and Pauly 2019). Study-specific information on the respective sample size, the life stage under investigation, the uptake of MP (in terms of frequency of MP-feeding across all individuals sampled and particle load per MP-feeding individual) and the assessment of potential impacts of MP ingestion are listed. The location and geographic region of the sampling site for each study are given. Abbreviations are as follows: y = yes; n = no; n/a = not available or not assessed. Trophic Guild: P = planktivore; PI = piscivore; H = herbivore; C = carnivore; O = omnivore. IUCN status: DD = data deficient; LC = least concern; CR = critically endangered; EN = endangered; VU = vulnerable; NT = near threatened. Life stage: ad = adult; sub-ad = sub-adult; juv = juvenile (incl. larval stages if applicable); + = different life stages are mentioned but respective results are not listed separately within study (e.g., "juv + ad"). If no classification to a specific habitat category was available, the data entry followed the reported depth distribution of the species given on FishBase (Froese and Pauly 2019).

Species **Suffix** **Common name** **Family**	**Trophic guild**	**Habitat category**	**Commercial Fisheries Importance**	**IUCN threat status**	**Total sample size across studies**	**Reference**	**Life Stage**	**Sample size**	**MP Uptake per species and study**	**Percentage of MP feeding individuals (%)**	**MP particles per individual**	**Impact**	**Location**	**Geographic Region**
Abudefduf sexfasciatus (Lacepède, 1801) Scissortail sergeant Pomacentridae	P	reef-associated	y	LC	5	Baalkhuyur et al. 2018	n/a	5	y	20.00	1.00	n/a	Saudi Arabian coast	Red Sea
Acanthogobius ommaturus (Richardson, 1845) Asian freshwater goby Gobiidae	n/a	demersal	n/a	n/a	17	Su et al. 2019a	n/a	17	y	100.00	3.75	n/a	4 locations in Hangzhou Bay and Yangtze Estuary	East China Sea
Acanthopagrus australis (Günther, 1859) Yellowfin bream Sparidae	C	demersal	y	LC	24	Halstead et al. 2018	juv	24	y	25.00	0.60	n/a	Sydney Harbour, Australia	Southern West-Pacific
Acanthopagrus latus (Houttuyn, 1782) Yellowfin seabream Sparidae	n/a	demersal	y	DD	15	Zhu et al. 2019b	n/a	15	y	100.00	4.10	n/a	various	Maowei Sea
Acanthurus coeruleus Bloch & Schneider, 1801 Blue tang surgeonfish Acanthuridae	H	reef-associated	y	LC	1	Miranda and de Carvalho-Souza 2016	n/a	1	n	0.00	0.00	n/a	Salvador, Brazil	Western South-Atlantic
Acanthurus gahhm (Forsskål, 1775)	n/a	reef-associated	n	LC	10	Baalkhuyur et al. 2018	n/a	10	y	100.00	1.00	n/a	Saudi Arabian coast	Red Sea

Species Suffix Common name Family	Trophic guild	Habitat category	Commercial Fisheries Importance	IUCN threat status	Total sample size across studies	Reference	Life Stage	Sample size	MP Uptake per species and study	Percentage of MP feeding individuals (%)	MP particles per individual	Impact	Location	Geographic Region
Black surgeonfish Acanthuridae														
Acanthurus lineatus (Linnaeus, 1758) Lined surgeonfish Acanthuridae	H	reef-associated	y	LC	24	Markic et al. 2018	ad	24	y	16.70	1.50	n/a	Samoa	South Pacific
Acanthurus sohal (Forsskål, 1775) Sohal surgeonfish Acanthuridae	H	reef-associated	n	LC	3	Baalkhuyur et al. 2018	n/a	3	n	0.00	0.00	n/a	Saudi Arabian coast	Red Sea
Achirus lineatus (Linnaeus, 1758) Lined sole Achiridae	C	reef-associated	y	LC	6	Vendel et al. 2017	n/a	6	n	0.00	0.00	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Agonus cataphractus (Linnaeus, 1758) Hooknose Agonidae	C	demersal	n	LC	3	McGoran et al. 2018	n/a	3	n	0.00	0.00	n/a	Firth of Clyde	Firth of Clyde
Aldrichetta forsteri (Valenciennes, 1836) Yellow-eye mullet Mugilidae	O	demersal	y	LC	17	Cannon et al. 2016	n/a	17	n	0.00	0.00	n/a	Australia	Southern Pacific, Southern Ocean, Australia
Alepes apercna Grant, 1987 Smallmouth scad Carangidae	P	demersal	y	LC	3	Azad et al. 2018	n/a	3	y	67.00	2.00	n/a	Sathing Phra District	Gulf of Thailand
Alepes kleinii (Bloch, 1793) Razorbelly scad Carangidae	P	reef-associated	y	LC	4	Azad et al. 2018	ad	4	y	50.00	0.80	n/a	Sathing Phra District	Gulf of Thailand
Alepes melanoptera (Swainson, 1839) Blackfin scad Carangidae	P	inshore waters	y	LC	8	Azad et al. 2018	n/a	8	y	75.00	1.30	n/a	Sathing Phra District	Gulf of Thailand
Alepes vari (Cuvier, 1833) Herring scad Carangidae	C	0 - 10 m	y	LC	3	Azad et al. 2018	n/a	3	y	67.00	1.70	n/a	Sathing Phra District	Gulf of Thailand
Alepisaurus ferox Lowe, 1833	C	meso-pelagic / epipelagic/	n/a	LC	144	Choy and Drazen 2013	ad	144	y	30.00	2.70	n/a	n/a	Central North Pacific Ocean

Species **Suffix** **Common name** **Family**	**Trophic guild**	**Habitat category**	**Commercial Fisheries Importance**	**IUCN threat status**	**Total sample size across studies**	**Reference**	**Life Stage**	**Sample size**	**MP Uptake per species and study**	**Percentage of MP feeding individuals (%)**	**MP particles per individual**	**Impact**	**Location**	**Geographic Region**
Long snouted lancetfish Alepisauridae		bathy-pelagic												
Alosa fallax (Lacepède, 1803) Twaite shad Clupeidae	C	10 - 400 m	y	LC	206	Skóra et al. 2012	juv + ad	205	y	n/a	n/a	n/a	Gulf of Gdansk	Baltic Sea
						Neves et al. 2015	ad	1	y	100.00	1.00	n/a	off the coast of Portugal	Eastern North Atlantic
Ammodytes personatus Girard, 1856 Pacific sandlance Ammodytidae	n/a	demersal	y	n/a	50	Sun et al. 2019	n/a	50	y	40.00	1.40	n/a	Yellow Sea, 53 locations	East China Sea
Anchoa januaria (Steindachner, 1879) Rio anchovy Engraulidae	n/a	0 - 50 m	n	LC	109	Vendel et al. 2017	juv + ad	109	y	n/a	0.16	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Anchoa marinii Hildebrand, 1943 Marini's anchovy Engraulidae	P	0 - 20 m	n/a	LC	19	Vendel et al. 2017	n/a	19	y	n/a	0.05	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Anchovia clupeoides (Swainson, 1839) Zabaleta anchovy Engraulidae	P	bentho-pelagic	y	LC	8	Vendel et al. 2017	n/a	8	y	n/a	0.13	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Anchoviella commersonii Lacepède, 1803 Commerson's anchovy Engraulidae	P	0 - 50 m	y	LC	30	Sun et al. 2019	ad	30	y	33.00	1.20	n/a	Yellow Sea, 53 locations	East China Sea
Anguilla anguilla (Linnaeus, 1758) European eel Anguillidae	O	demersal	y	CR	3	McGoran et al. 2018	ad	2	y	50.00	2.00	n/a	Thames estuary,	English Channel
						Steer et al. 2017	juv	1	y	100.00	n/a	n/a	various	western English Channel
Anisotremus surinamensis (Bloch, 1791) Black margate Haemulidae	C	reef-associated	y	DD	1	Pegado et al. 2018	n/a	1	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Anisotremus virginicus (Linnaeus, 1758) Porkfish Haemulidae	C	reef-associated	y	LC	1	Pegado et al. 2018	n/a	1	n	0.00	0.00	n/a	Amazon River estuary, northern	Western South-Atlantic

Species Suffix Common name Family	Trophic guild	Habitat category	Commercial Fisheries Importance	IUCN threat status	Total sample size across studies	Reference	Life Stage	Sample size	MP Uptake per species and study	Percentage of MP feeding individuals (%)	MP particles per individual	Impact	Location	Geographic Region
													coast of Brazil	
Anodontostoma chacunda (Hamilton, 1822) Chacunda gizzard shad Clupeidae	C	0 - 50 m	y	LC	14	Azad et al. 2018	n/a	14	y	57.00	2.00	n/a	Sathing Phra District	Gulf of Thailand
Antigonia capros Lowe, 1843 Deepbody boarfish Caproidae	C	demersal	y	LC	3	Zhu et al. 2019a	n/a	3	y	100.00	n/a	n/a	various	South China Sea
Aphanopus carbo Lowe, 1843 Black scabbardfish Trichiuridae	C	bathy-pelagic	y	LC	5	Murphy et al. 2017	n/a	5	n	0.00	0.00	n/a	Scottish offshore waters	North Sea, Eastern North Atlantic
Apogon lineatus (Temminck & Schlegel, 184 Indian perch Apogonidae	n/a	demersal	n/a	n/a	20	Sun et al. 2019	juv + ad	20	y	35.00	1.10	n/a	Yellow Sea, 53 locations	East China Sea
Arctozenus risso (Bonaparte, 1840) Spotted barracudina Paralepididae	C	Bathy-pelagic / meso-pelagic / pseudo-oceanic	n	LC	14	Lusher et al. 2016	n/a	14	y	21.00	1.30	n/a	various	Eastern North-Atlantic
Argentina silus (Ascanius, 1775) Greater argentine Argentinidae	P	bathy-demersal	y	n/a	15	Murphy et al. 2017	n/a	15	y	6.70	0.10	n/a	Scottish offshore waters	North Sea, Eastern North Atlantic
Argyropelecus affinis Garman, 1899 Pacific hatchet fish Sternoptychidae	P	meso-pelagic / bathy-pelagic	n	LC	1	Davison and Asch 2011	ad	1	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Argyropelecus hemigymnus Cocco, 1829 Half-naked hatchet fish Sternoptychidae	P	meso-pelagic / bathy-pelagic	n	LC	3	Davison and Asch 2011	ad	3	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Argyrosomus regius (Asso, 1801) Meagre Sciaenidae	C	bentho-pelagic	y	LC	56	Güven et al. 2017	n/a	51	y	75.00	2.47	n/a	Turkish territorial waters	Mediterranean
						Neves et al. 2015	n/a	5	y	60.00	0.80	n/a	off the coast of Portugal	Eastern North-Atlantic

Species **Suffix** **Common name** **Family**	**Trophic guild**	**Habitat category**	**Commercial Fisheries Importance**	**IUCN threat status**	**Total sample size across studies**	**Reference**	**Life Stage**	**Sample size**	**MP Uptake per species and study**	**Percentage of MP feeding individuals (%)**	**MP particles per individual**	**Impact**	**Location**	**Geographic Region**
Arnoglossus imperialis (Rafinesque, 1810) Imperial scaldfish Bothidae	n/a	demersal	y	LC	8	Steer et al. 2017	juv	8	n	0.00	0.00	n/a	various	western English Channel
Astronesthes indopacificus Parin & Borodulina, 1997 Indo-Pacific snaggletooth Stomiidae	n/a	meso-pelagic	n/a	LC	7	Boerger et al. 2010	n/a	7	y	n/a	1.00	n/a	various	North Pacific Central Gyre
Atherinella brasiliensis (Quoy & Gaimard, 1825) Brazilian silversides Atherinopsidae	n/a	bentho-pelagic	y	LC	215	Vendel et al. 2017	juv + ad	215	y	n/a	0.05	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Atherinopsis californiensis Girard, 1854 Jack silverside Atherinopsidae	n/a	0 - 30 m	y	LC	7	Rochman et al. 2015	n/a	7	y	28.60	1.60	n/a	Half Moon Bay, California, USA	Eastern North-Pacific
Auchenionchus microcirrhis (Valenciennes, 1836) n/a Labrisomidae	C	demersal	n	LC	16	Mizraji et al. 2017	juv	16	y	100.00	10.00	condition K in g cm^{-3}	Las Cruces, Chile	Eastern South Pacific
Aurigequula fasciata *(Leiognathus fasciatus)* (Lacepède, 1803) Striped ponyfish Leiognathidae	C	demersal	y	LC	3	Azad et al. 2018	n/a	3	y	33.00	1.30	n/a	Sathing Phra District	Gulf of Thailand
Bagre bagre (Linnaeus, 1766) Coco sea catfish Ariidae	C	demersal	y	LC	7	Pegado et al. 2018	n/a	7	y	71.40	12.80	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Bagre marinus (Mitchill, 1815) Gafftopsail sea catfish Ariidae	C	demersal	y	LC	4	Pegado et al. 2018	n/a	4	y	100.00	7.80	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Bairdiella ronchus (Cuvier, 1830) Ground croaker Sciaenidae	C	demersal	y	LC	4	Pegado et al. 2018	n/a	4	n	0.00	0.00	n/a	Amazon River estuary, northern	Western South-Atlantic

Species **Suffix** **Common name** **Family**	**Trophic guild**	**Habitat category**	**Commercial Fisheries Importance**	**IUCN threat status**	**Total sample size across studies**	**Reference**	**Life Stage**	**Sample size**	**MP Uptake per species and study**	**Percentage of MP feeding individuals (%)**	**MP particles per individual**	**Impact**	**Location**	**Geographic Region**
													coast of Brazil	
Batrachoides surinamensis (Bloch & Schneider, 1801) Pacuma toadfish Batrachoididae	C	demersal	y	LC	1	Pegado et al. 2018	n/a	1	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Benthodesmus tenuis (Günther, 1877) Slender frostfish Trichiuridae	n/a	bentho-pelagic	n/a	LC	2	Zhu et al. 2019a	n/a	2	y	100.00	n/a	n/a	various	South China Sea
Benthosema glaciale (Reinhardt, 1837) Glacier lantern fish Myctophidae	P	pelagic-oceanic / meso-pelagic	n	LC	96	Lusher et al. 2016	n/a	27	y	22.00	1.50	n/a	various	Eastern North-Atlantic
						Wieczorek et al. 2018	ad	69	y	68.12	1.46	n/a	Galway (Ireland) to St. John's (Newfoundland)	Eastern North-Atlantic
Benthosema pterotum (Alcock, 1890) Skinnycheek lanternfish Myctophidae	P	bentho-pelagic / meso-pelagic	y	LC	10	Baalkhuyur et al. 2018	n/a	10	y	100.00	1.00	n/a	Saudi Arabian coast	Red Sea
Boleophtalmus pectinirostris (Linnaeus, 1758) Great blue spotted mudskipp Gobiidae	H	demersal	y	n/a	9	Su et al. 2019a	n/a	9	y	100.00	5.30	n/a	4 locations in Hangzhou Bay and Yangtze Estuary	East China Sea
Bolinichthys longipes (Brauer, 1906) Popeye lampfish Myctophidae	n/a	high-oceanic / meso-pelagic	n/a	n/a	3	Davison and Asch 2011	ad	3	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Boops boops (Linnaeus, 1758) Bogue Sparidae	O	demersal	y	LC	417	Nadal et al. 2016	juv + ad	337	y	57.80	4.98	n/a	Mallorca, Eivissa	Mediterranean
						Neves et al. 2015	ad	32	y	9.00	0.09	n/a	off the coast of Portugal	Eastern North Atlantic
						Rios-Fuster et al. 2019	juv + ad	48	y	27.10	0.50	n/a	Iberian Peninsula coast, Balearic Island	Mediterranean

Species **Suffix** **Common name** **Family**	**Trophic guild**	**Habitat category**	**Commercial Fisheries Importance**	**IUCN threat status**	**Total sample size across studies**	**Reference**	**Life Stage**	**Sample size**	**MP Uptake per species and study**	**Percentage of MP feeding individuals (%)**	**MP particles per individual**	**Impact**	**Location**	**Geographic Region**
Boreogadus saida (Lepechin, 1774) Polar cod Gadidae	C	demersal	y	n/a	157	Morgana et al. 2018	n/a	85	y	18.00	1.10	Fulton's K Condition factor	Northeast Greenland	Northern Atlantic
						Kühn et al. 2018	juv	72	y	2.80	1.00	n/a	ice-covered water in Eurasian Basin; open water near Svalbard	Central Arctic Ocean
Borostomias pacificus (Imai, 1941) n/a Stomiidae	n/a	bathy-pelagic / meso-pelagic	n/a	n/a	1	Zhu et al. 2019a	n/a	1	y	100.00	n/a	n/a	various	South China Sea
Brama brama (Bonnaterre, 1788) Atlantic pomfret Bramidae	C	oceanic, epipelagic	y	LC	12	Anastasopoulou et al. 2013	n/a	9	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
						Neves et al. 2015	n/a	3	y	33.00	0.67	n/a	off the coast of Portugal	Eastern North Atlantic
Branchiostegus japonicus (Houttuyn, 1782) Horsehead tilefish Malacanthidae	n/a	demersal	y	LC	18	Jabeen et al. 2017	juv	18	y	100.00	4.60	n/a	Yangtze estuary, East China Sea and South China Sea	China Sea
Brevoortia patronus Goode, 1878 Gulf menhaden Clupeidae	P	0 - 50 m	y	LC	9	Phillips and Bonner 2015	n/a	9	n	0.00	0.00	n/a	Laguna Madre, southern coast of Texas	Western North-Atlantic
Buglossidium luteum (Risso, 1810) Solenette Soleidae	C	demersal	y	LC	50	Lusher et al. 2013	n/a	50	y	26.00	1.25	n/a	10 km southwest of Plymouth	English Channel
Callionymus lyra Linnaeus, 1758 Dragonet Callionymidae	C	demersal	y	LC	101	Lusher et al. 2013	n/a	50	y	38.00	1.90	n/a	10 km southwest of Plymouth	English Channel
						Steer et al. 2017	juv	51	y	1.90	n/a	n/a	various	western English Channel
Callionymus planus Ochiai, 1955	n/a	demersal	n/a	n/a	18	Jabeen et al. 2017	n/a	18	y	100.00	4.80	n/a	Yangtze estuary, East	China Sea

Species **Suffix** **Common name** **Family**	**Trophic guild**	**Habitat category**	**Commercial Fisheries Importance**	**IUCN threat status**	**Total sample size across studies**	**Reference**	**Life Stage**	**Sample size**	**MP Uptake per species and study**	**Percentage of MP feeding individuals (%)**	**MP particles per individual**	**Impact**	**Location**	**Geographic Region**
Japanese darter dragonet Callionymidae													China Sea and South China Sea	
Callionymus reticulatus Valenciennes, 1837 Reticulated dragonet Callionymidae	n/a	demersal	n	n/a	1	Steer et al. 2017	n/a	1	n	0.00	0.00	n/a	various	western English Channel
Caranx crysos (Mitchill, 1815) Blue runner Carangidae	C	reef-associated	y	LC	7	Miranda and de Carvalho-Souza 2016	n/a	3	n	0.00	0.00	n/a	Salvador, Brazil	Western South-Atlantic
						Güven et al. 2017	n/a	1	y	100.00	5.00	n/a	Turkish territorial waters	Mediterranean
						Pegado et al. 2018	n/a	3	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Caranx hippos (Linnaeus, 1766) Crevalle jack Carangidae	C	reef-associated	y	LC	12	Phillips and Bonner 2015	n/a	9	n	0.00	0.00	n/a	Laguna Madre, southern coast of Texas	Western North-Atlantic
						Pegado et al. 2018	n/a	3	y	100.00	30.70		Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Caranx latus Agassiz, 1831 Horse-eye jack Carangidae	C	reef-associated	y	LC	55	Vendel et al. 2017	juv	55	n	0.00	0.00	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Caranx papuensis Alleyne & MacLeay, 1877 Brassy trevally Carangidae	PI	reef-associated	y	LC	32	Markic et al. 2018	n/a	32	y	43.80	2.40	n/a	Tahiti	South Pacific
Cathorops agassizii (Eigenmann &	n/a	bentho-pelagic	n/a	n/a	20	Possatto et al. 2011	juv	20	y	15.00	0.40	n/a	Goiana Estuary, Extractive Reserve	Western South-Atlantic

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Eigenmann, 1888) n/a Ariidae													Acaú-Goiana	
Cathorops spixii (Agassiz, 1829) Madamango sea catfish Ariidae	C	demersal	y	n/a	20	Possatto et al. 2011	juv	20	y	10.00	0.15	n/a	Goiana Estuary, Extractive Reserve Acaú-Goiana	Western South-Atlantic
Centroberyx lineatus (Cuvier, 1829) Swallow-tail Berycidae	n/a	bentho-pelagic	y	n/a	3	Zhu et al. 2019a	n/a	3	y	100.00	n/a	n/a	various	South China Sea
Centrophorus granulosus (Bloch & Schneider, 1801) Gulper shark Centrophoridae	C	bathy-demersal	y	DD	5	Anastasopoulou et al. 2013	n/a	5	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Cephalopholis argus Schneider, 1801 Peacock hind Serranidae	C	reef-associated	y	LC	4	Baalkhuyur et al. 2018	ad	4	n	0.00	0.00	n/a	Saudi Arabian coast	Red Sea
Cepola macrophthalma (Linnaeus, 1758) Red bandfish Cepolidae	P	demersal	y	LC	62	Lusher et al. 2013	n/a	62	y	32.30	2.30	n/a	10 km southwest of Plymouth	English Channel
Ceratoscopelus warmingii (Lütken, 1892) Warming's lantern fish Myctophidae	P	meso-pelagic / bathy-pelagic / oceanic	n/a	LC	8	Davison and Asch 2011	ad	8	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Cetengraulis mysticetus (Günther, 1867) Pacific anchoveta Engraulidae	P	0 - 32 m	y	LC	30	Ory et al. 2018a	ad	30	y	3.30	0.03	n/a	Colombia	South Eastern Pacific
Chaetodipterus faber (Broussonet, 1782) Atlantic spadefish Ephippidae	C	reef-associated	y	LC	108	Peters et al. 2017	n/a	103	y	46.60	2.96	n/a	Texas (TX) Gulf Coast, Galveston Bay to Freeport	Western North-Atlantic

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						Pegado et al. 2018	n/a	5	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Chaetodon austriacus Rüppell, 1836 Blacktail butterflyfish Chaetodontidae	n/a	reef-associated	n/a	LC	10	Baalkhuyur et al. 2018	n/a	10	y	100.00	1.00	n/a	Saudi Arabian coast	Red Sea
Cheilopogon pitcairnensis (Nichols & Breder, 1935) n/a Exocoetidae	C	0 - 20 m	y	n/a	21	Markic et al. 2018	n/a	21	y	9.50	1.00	n/a	Tahiti	South Pacific
Cheilopogon rapanoiiensis Parin, 1961 Easter island flyingfish Exocoetidae	C	0 - 20 m	n	LC	43	Chagnon et al. 2018	ad	43	y	16.30	1.50	n/a	Rapa Nui	South Pacific
Chelidonichthys cuculus *(Aspitrigla cuculus)* (Linnaeus, 1758) Red gurnard Triglidae	C	demersal	y	LC	72	McGoran et al. 2018	juv + ad, juv	5, 1	n, n	0, 0	0, 0	n/a	Thames estuary, Firth of Clydde	English Channel, Firth of Clyde
						Lusher et al. 2013	n/a	66	y	51.50	2.00	n/a	10 km southwest of Plymouth	English Channel
Chelidonichthys kumu (Cuvier, 1829) Bluefin gurnard Triglidae	n/a	demersal	y	LC	204	Markic et al. 2018	ad	27	y	3.70	2.00	n/a	Auckland	South Pacific
						Sun et al. 2019	juv	177	y	36.00	1.20	n/a	Yellow Sea, 53 locations	East China Sea
Chelidonichthys lucerna *(Trigla lucerna)* (Linnaeus, 1758) Tub gurnard Triglidae	C	demersal	y	LC	25	Anastasopoulou et al. 2018	ad	1	n	0.00	0.00	n/a	NE Ionian Sea	Mediterranean
						Güven et al. 2017	n/a	24	y	37.00	2.00	n/a	Turkish territorial waters	Mediterranean
Chelon auratus *(Liza aurata)* (Risso, 1810) Golden grey mullet Mugilidae	O	pelagic-neritic	y	LC	59	Anastasopoulou et al. 2018	ad	20	y	95.00	9.50	n/a	Slovenian Sea	Mediterranean
						Güven et al. 2017	n/a	39	y	44.00	7.47	n/a	Turkish territorial waters	Mediterranean

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Chlorophthalmus agassizi Bonaparte, 1840 Shortnose greeneye Chlorophthalmidae	C	bathy-demersal	y	LC	1	Zhu et al. 2019a	n/a	1	y	100.00	n/a	n/a	various	South China Sea
Chlorophthalmus albatrossis Jordan & Starks, 1904 n/a Chlorophthalmidae	C	bathy-demersal	y	n/a	4	Zhu et al. 2019a	n/a	4	y	100.00	n/a	n/a	various	South China Sea
Ciliata mustela (Linnaeus, 1758) Fivebeard rockling Lotidae	O	demersal	y	LC	125	Kartar et al. 1976	n/a	50, 5, 50, 8	y, n, y, n	2, 0, 10, 0	n/a, 0, n/a, 0	n/a, n/a, n/a, n/a	Oldbury	Eastern Nord-Atlantic
						Steer et al. 2017	juv	5, 7	n, n	0, 0	0, 0	n/a, n/a	various	western English Channel
Ciliata septentrionalis (Collett, 1875) Northern rockling Lotidae	C	demersal	y	LC	7	Steer et al. 2017	juv	7	n	0.00	0.00	n/a	various	western English Channel
Citharichthys sordidus (Girard, 1854) Pacific sanddab Paralichthyidae	n/a	demersal	y	n/a	5	Rochman et al. 2015	n/a	5	y	60.00	1.00	n/a	Half Moon Bay, California, USA	Eastern North-Pacific
Citharichthys spilopterus Günther, 1862 Bay whiff Paralichthyidae	P	demersal	y	LC	9	Vendel et al. 2017	n/a	9	n	0.00	0.00	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Citharus linguatula (Linnaeus, 1758) Spotted flounder Citharidae	C	demersal	y	LC	52	Anastasopoulou et al. 2018	ad	52	y	1.90	0.01	n/a	NE Ionian Sea	Mediterranean
Cleisthenes herzensteini (Schmidt, 1904) n/a Pleuronectidae	C	demersal	y	n/a	36	Sun et al. 2019	juv	36	y	33.00	1.30	n/a	Yellow Sea, 53 locations	East China Sea
Clupea harengus Linnaeus, 1758 Atlantic herring Clupeidae	P	bentho-pelagic	y	LC	1051	Collard et al. 2017b	ad	20	y	50.00	n/a	n/a	English Channel, Mediterranean, Eastern North-Atlantic	English Channel, Bay of Biscay, Northwestern Mediterranean
						Hermsen et al. 2017	ad	131	n	0.00	0.00	n/a	Southern Bight	North Sea

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						Carpenter et al. 1972	juv	2	y	100.00	1.00	n/a	Southern New England	Western North-Atlantic
						Foekema et al. 2013	n/a	566	y	1.40	2.00	condition based on morphometrics	various	North Sea
						Rummel et al. 2016	ad, ad	13, 20	n, n	0, 0	0, 0	Fulton's K condition factor	Tiefe Rinne, Helgoland; Herwig Cruise	North Sea, Baltic Sea
						Beer et al. 2018	juv + ad	299	y	21.10	1.15	n/a	Bornholm Basin	Baltic Sea
Coilia mystus (Linnaeus, 1758) Osbeck's grenadier anchovy Engraulidae	n/a	pelagic-neritic	y	EN	9	Su et al. 2019a	n/a	9	y	22.00	0.30	n/a	4 locations in Hangzhou Bay and Yangtze Estuary	East China Sea
Coilia nasus *(Coilia ectenes)* Temminck & Schlegel, 1846 Japanese grenadier anchovy Engraulidae	P	pelagic-neritic	y	EN	54	Su et al. 2019a	n/a	36	y	44.00	0.70	n/a	4 locations in Hangzhou Bay and Yangtze Estuary	East China Sea
						Jabeen et al. 2017	n/a	18	y	100.00	4.00	n/a	Yangtze estuary, East China Sea and South China Sea	China Sea
Collichthys lucidus (Richardson, 1844) n/a Sciaenidae	n/a	demersal	n/a	n/a	44	Jabeen et al. 2017	ad	18	y	100.00	6.20	n/a	Yangtze estuary, East China Sea and South China Sea	China Sea
						Su et al. 2019a	juv	26	y	46.00	1.20	n/a	4 locations in Hangzhou Bay and Yangtze Estuary	East China Sea
Cololabis saira (Brevoort, 1856) Pacific saury Scomberesocidae	P	pelagic-oceanic	n/a	n/a	53	Boerger et al. 2010	n/a	53	y	n/a	3.20	n/a	various	North Pacific Central Gyre

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Colomesus psittacus (Bloch & Schneider, 1801) Banded puffer Tetraodontidae	C	demersal	n/a	LC	2	Pegado et al. 2018	n/a	2	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Conger conger (Linnaeus, 1758) European conger Congridae	C	demersal	y	LC	44	Anastasopoulou et al. 2013	n/a	44	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Conger verreauxi Kaup, 1856 Conger Congridae	n/a	0 - 45 m	y	LC	25	Cannon et al. 2016	n/a	25	n	0.00	0.00	n/a	Australia	Southern Pacific, Southern Ocean, Australia
Conodon nobilis (Linnaeus, 1758) Banded grunt Haemulidae	C	demersal	y	LC	8	Pegado et al. 2018	n/a	8	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Coryphaena hippurus Linnaeus, 1758 Common dolphinfish Coryphaenidae	C	pelagic-neritic	y	LC	54	Choy and Drazen 2013	ad	42	y	2.00	n/a	n/a	n/a	Central North Pacific Ocean
						Phillips and Bonner 2015	n/a	2	y	n/a	n/a	n/a	Laguna Madre, southern coast of Texas	Western North-Atlantic
						Markic et al. 2018	ad	10	y	20.00	2.00	n/a	Tahiti	South Pacific
Coryphaenoides rupestris Gunnerus, 1765 Roundnose grenadier Macrouridae	C	bathy-pelagic / bentho-pelagic	y	CR	5	Murphy et al. 2017	n/a	5	n	0.00	0.00	n/a	Scottish offshore waters	North Sea, Eastern North Atlantic
Ctenochaetus striatus (Quoy & Gaimard, 1825) Striated surgeonfish Acanthuridae	P	reef-associated	y	LC	29	Markic et al. 2018	n/a	29	y	20.70	1.00	n/a	Samoa, Tahiti	South Pacific
Ctenogobius boleosoma (Jordan & Gilbert, 1882)	n/a	reef-associated	n/a	LC	16	Vendel et al. 2017	n/a	16	y	n/a	0.06	n/a	Paraiba estuary,	Western South-Atlantic

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Darter goby Gobiidae													Northeastern Brazil	
Ctenogobius giurinus (Rutter, 1897) n/a Gobiidae	C	demersal	y	LC	16	Zhu et al. 2019b	n/a	16	y	100.00	4.60	n/a	various	Maowei Sea
Ctenosciaena gracilicirrhus (Metzelaar, 1919) Barbel drum Sciaenidae	C	demersal	y	LC	11	Pegado et al. 2018	n/a	11	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Cyclothone acclinidens Garman, 1899 Benttooth bristlemouth Gonostomatidae	P	meso-pelagic / bathy-pelagic	n	LC	4	Davison and Asch 2011	ad	4	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Cyclothone atraria Gilbert, 1905 Deep-water bristlemouth Gonostomatidae	n/a	meso-pelagic / bathy-pelagic	n/a	n/a	3	Davison and Asch 2011	ad	3	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Cyclothone pallida Brauer, 1902 Tan bristlemouth Gonostomatidae	n/a	meso-pelagic / bathy-pelagic	n	LC	5	Davison and Asch 2011	ad	5	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Cyclothone pseudopallida Mukhacheva, 1964 Slender bristlemouth Gonostomatidae	n/a	meso-pelagic / bathy-pelagic	n	LC	4	Davison and Asch 2011	ad	4	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Cynoglossus abbreviatus (Gray, 1834) Three-lined tongue sole Cynoglossidae	C	demersal	y	n/a	33	Jabeen et al. 2017	n/a	18	y	100.00	6.90	n/a	Yangtze estuary, East China Sea and South China Sea	China Sea
						Abbasi et al. 2018	n/a	15	y	n/a	2.90	n/a	various	Persian Gulf
Cynoglossus robustus Günther, 1873 n/a Cynoglossidae	n/a	demersal	y	n/a	9	Su et al. 2019a	n/a	9	y	44.00	0.70	n/a	4 locations in Hangzhou Bay and Yangtze Estuary	East China Sea

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Cynoponticus savanna (Bancroft, 1831) Guayana pike-conger Muraenesocidae	C	demersal	y	LC	1	Pegado et al. 2018	n/a	1	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Cynoscion acoupa (Lacepède, 1801) Acoupa weakfish Sciaenidae	C	demersal	y	LC	1079	Ferreira et al. 2016a	juv, sub-ad, ad	469, 25, 33	y, y, y	64.4, 50, 100	1.8 - 2.5, 2 - 8, 5.4 - 25.9	n/a, n/a, n/a	Goiana Estuary, Extractive Reserve Acaú-Goiana	Western South-Atlantic
						Ferreira et al. 2018	juv + sub-ad + ad	552	y	51.00	3.03	n/a	Goiana Estuary, Extractive Reserve Acaú-Goiana	Western South-Atlantic
Cynoscion arenarius Ginsburg, 1930 Sand weakfish Sciaenidae	n/a	demersal	y	LC	139	Peters et al. 2017	n/a	139	y	43.20	1.83	n/a	Texas (TX) Gulf Coast, Galveston Bay to Freeport	Western North-Atlantic
Cynoscion jamaicensis (Vaillant & Bocourt, 1883) Jamaica weakfish Sciaenidae	C	demersal	y	LC	3	Pegado et al. 2018	n/a	3	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Cynoscion leiarchus (Cuvier, 1830) Smooth weakfish Sciaenidae	n/a	demersal	y	LC	2	Pegado et al. 2018	n/a	2	y	50.00	2.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Cynoscion microlepidotus (Cuvier, 1830) Smallscale weakfish Sciaenidae	n/a	demersal	y	LC	16	Pegado et al. 2018	n/a	16	y	18.70	1.30	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic

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Cynoscion nebulosus (Cuvier, 1830) Spotted weakfish Sciaenidae	C	demersal	y	LC	20	Phillips and Bonner 2015	n/a	20	y	n/a	n/a	n/a	Laguna Madre, southern coast of Texas	Western North-Atlantic
Cynoscion virescens (Cuvier, 1830) Green weakfish Sciaenidae	n/a	demersal	y	LC	7	Pegado et al. 2018	n/a	7	y	14.30	3.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Danaphos oculatus (Garman, 1899) Bottlelights Sternoptychidae	n/a	meso-pelagic	n/a	n/a	9	Davison and Asch 2011	ad	9	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Dascyllus trimaculatus (Rüppell, 1829) Threespot dascyllus Pomacentridae	O	reef-associated	y	n/a	2	Baalkhuyur et al. 2018	n/a	2	n	0.00	0.00	n/a	Saudi Arabian coast	Red Sea
Dasyatis zugei (Müller & Henle, 1841) Pale-edged stingray Dasyatidae	C	demersal	y	NT	3	Azad et al. 2018	ad	3	y	33.00	0.30	n/a	Sathing Phra District	Gulf of Thailand
Decapterus macrosoma Bleeker, 1851 Shortfin scad Carangidae	P	reef-associated	y	LC	42	Rochman et al. 2015	n/a	17	y	29.40	2.50	n/a	Makassar, Sulawesi, Indonesia;	Makassar Strait
						Markic et al. 2018	ad	25	y	28.00	1.10	n/a	Tahiti	South Pacific
Decapterus muroadsi (Temminck & Schlegel, 184 Amberstripe scad Carangidae	P	pelagic / oceanic	y	LC	123	Sun et al. 2019	n/a	78	y	31.00	1.30	n/a	Yellow Sea, 53 locations	East China Sea
						Ory et al. 2017	n/a	20	y	80.00	2.50	n/a	Rapa Nui, Easter Island, South Pacific subtropical gyre	South Pacific Subtropical Gyre
						Markic et al. 2018	n/a	25	y	64.00	2.40	n/a	Rapa Nui	South Pacific
Dendrophysa russelii (Cuvier, 1829) Goatee croaker Sciaenidae	n/a	demersal	y	n/a	3	Azad et al. 2018	n/a	3	y	33.00	0.30	n/a	Sathing Phra District	Gulf of Thailand

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Dentex dentex (Linnaeus, 1758) Common dentex Sparidae	C	bentho-pelagic	y	VU	1	Güven et al. 2017	n/a	1	n	0.00	0.00	n/a	Turkish territorial waters	Mediterranean
Dentex gibbosus (Rafinesque, 1810) Pink dentex Sparidae	C	bentho-pelagic	y	LC	14	Güven et al. 2017	n/a	14	y	29.00	1.00	n/a	Turkish territorial waters	Mediterranean
Dentex macrophthalmus (Bloch, 1791) Large-eye dentex Sparidae	C	bentho-pelagic	y	LC	1	Neves et al. 2015	n/a	1	y	100.00	1.00	n/a	off the coast of Portugal	Eastern North Atlantic
Diaphus anderseni Tåning, 1932 Andersen's lantern fish Myctophidae	n/a	meso-pelagic / oceanic	n/a	LC	13	Davison and Asch 2011	ad	13	y	15.40	n/a	n/a	various	North Pacific Subtropical Gyre
Diaphus fulgens (Brauer, 1904) n/a Myctophidae	n/a	meso-pelagic	n/a	n/a	7	Davison and Asch 2011	ad	7	y	14.30	n/a	n/a	various	North Pacific Subtropical Gyre
Diaphus metopoclampus (Cocco, 1829) Spothead lantern fish Myctophidae	n/a	oceanic / meso-pelagic	n/a	LC	296	Romeo et al. 2016	ad	296	y	0.30	n/a	n/a	Strait of Sicily	Central Mediterranean Sea
Diaphus phillipsi Fowler, 1934 Bolin's lantern fish Myctophidae	n/a	oceanic / meso-pelagic	n/a	n/a	1	Davison and Asch 2011	ad	1	y	100.00	n/a	n/a	various	North Pacific Subtropical Gyre
Diaphus rafinesquii (Cocco, 1838) White-spotted lantern fish Myctophidae	n/a	high-oceanic / bathy-pelagic	n/a	LC	34	Wieczorek et al. 2018	ad	34	y	70.59	1.15	n/a	Galway (Ireland) to St. John's (Newfoundland)	Eastern North-Atlantic
Diaphus watasei Jordan & Starks, 1904 Watases lanternfish Myctophidae	n/a	bentho-pelagic / meso-pelagic	y	n/a	4	Zhu et al. 2019a	juv + ad	4	y	100.00	n/a	n/a	various	South China Sea
Diapterus auratus Ranzani, 1842 Irish mojarra Gerreidae	C	demersal	y	LC	29	Vendel et al. 2017	n/a	29	y	n/a	0.97	n/a	Mamanguape estuary, Northeastern Brazil	Western South-Atlantic

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Diapterus rhombeus (Cuvier, 1829) Caitipa mojarra Gerreidae	C	demersal	y	LC	42	Vendel et al. 2017	juv, juv	13, 18	n, y	0, n/a	0, 0.06	n/a, n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
						Ramos et al. 2012	juv, sub-ad	4, 7	n, y	0, 14.3	0, 0.28	condition K (Richardson et al. 2011)	Goiana Estuary, Extractive Reserve Acaú-Goiana	Western South-Atlantic
Dicentrarchus labrax (Linnaeus, 1758) European seabass Moronidae	C	demersal	y	LC	40	Bessa et al. 2018b	juv	40	y	23.00	0.30	n/a	Portugal, Mondego estuary	Eastern North-Atlantic
Diogenichthys atlanticus (Tåning, 1928) Longfin lanternfish Myctophidae	n/a	meso-pelagic / epipelagic / oceanic	n/a	LC	5	Davison and Asch 2011	ad	5	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Diplodus annularis (Linnaeus, 1758) Annular seabream Sparidae	C	bentho-pelagic	y	LC	48	Güven et al. 2017	n/a	48	y	69.00	2.85	n/a	Turkish territorial waters	Mediterranean
Diplodus vulgaris (Geoffroy Saint-Hilaire, 181 Common two-banded seabre Sparidae	C	bentho-pelagic	y	LC	40	Bessa et al. 2018b	juv	40	y	73.00	3.14	n/a	Portugal, Mondego estuary	Eastern North-Atlantic
Dissostichus mawsoni Norman, 1937 Antarctic toothfish Nototheniidae	n/a	0 - 2200 m	y	n/a	10	Cannon et al. 2016	ad	10	y	10.00	2.00	n/a	Australia	Southern Pacific, Southern Ocean, Australia
Dorosoma cepedianum (Lesueur, 1818) American gizzard shad Clupeidae	H	0 - 33 m	y	LC	16	Phillips and Bonner 2015	n/a	16	y	n/a	n/a	n/a	Laguna Madre, southern coast of Texas	Western North-Atlantic
Dorosoma petenense (Günther, 1867) Threadfin shad Clupeidae	O	0 - 15 m	y	LC	5	Phillips and Bonner 2015	n/a	5	y	n/a	n/a	n/a	Laguna Madre, southern coast of Texas	Western North-Atlantic

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Drepane longimana (Bloch & Schneider, 1801) Concertina fish Drepaneidae	C	reef-associated	y	n/a	3	Azad et al. 2018	n/a	3	y	33.00	0.30	n/a	Sathing Phra District	Gulf of Thailand
Electrona risso (Cocco, 1829) Electric lantern fish Myctophidae	n/a	oceanic/bathy-pelagic/epipelagic / meso-pelagic	n/a	LC	82	Romeo et al. 2016	juv	82	y	6.10	n/a	n/a	Sicilian coast	Central Mediterranean Sea
Ellochelon vaigiensis (Quoy & Gaimard, 1825) Squaretail mullet Mugilidae	P	reef-associated	y	LC	33	Markic et al. 2018	n/a	33	y	48.50	4.30	n/a	Tahiti	South Pacific
Enchelyopus cimbrius (Linnaeus, 1766) Fourbeard rockling Lotidae	C	demersal	y	LC	17	Bour et al. 2018	n/a	17	y	5.90		n/a	Oslofjord	Skagerrak
Engraulis australis (White, 1790) Australian anchovy Engaulidae	P	pelagic-neritic	y	LC	10	Cannon et al. 2016	ad	10	n	0.00	0.00	n/a	Australia	Southern Pacific, Southern Ocean, Australia
Engraulis encrasicolus (Linnaeus, 1758) European anchovy Engraulidae	P	pelagic-neritic	y	LC	245	Collard et al. 2017b	ad	20	y	40.00	n/a	n/a	English Channel, Bay of Biscay, Northwestern Mediterranean	English Channel, Mediterranean, Eastern North-Atlantic
						Compa et al. 2018	sub-ad + ad	105	y	14.28	0.18	Fulton's K condition factor	western Spanish Mediterranean, Gulf of Lions to the Alboran Sea	Mediterranean
						Steer et al. 2017	juv	1	n	0.00	0.00	n/a	various	western English Channel
						Rios-Fuster et al. 2019	ad	39	y	2.56	0.03	n/a	Iberian Peninsula	Mediterranean

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													coast, Balearic Island	
						Renzi et al. 2019	ad	80	y	91.00	1.25	n/a	central Adriatic Sea	Mediterranean
Engraulis japonicus Temminck & Schlegel, 1846 Japanese anchovy Engraulidae	P	0 - 400 m	y	LC	344	Tanaka and Takada 2016	ad	64	y	77.00	2.30	n/a	Tokyo Bay	Western North-Pacific
						Sun et al. 2019	juv + ad	280	y	33.00	1.20	n/a	Yellow Sea, 53 locations	East China Sea
Engraulis mordax Girard, 1854 Californian anchovy Engraulidae	P	0 - 310 m	y	LC	10	Rochman et al. 2015	n/a	10	y	30.00	0.30	n/a	Half Moon Bay, California, USA	Eastern North-Pacific
Engraulis ringens Jenyns, 1842 Anchoveta Engraulidae	P	pelagic-neritic	y	LC	76	Ory et al. 2018a	ad	76	y	1.30	0.10	n/a	Chile, Peru	South Eastern Pacific
Epigonus telescopus (Risso, 1810) Black cardinal fish Epigonidae	C	bathy-demersal	y	LC	4	Anastasopoulou et al. 2013	n/a	4	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Epinephalus merra Bloch, 1793 Honeycomb grouper Serranidae	C	reef-associated	y	LC	20	Kumar et al. 2018	ad	20	y	n/a	n/a	n/a	Tamil Nadu, South East Coast of India	Indian Ocean
Epinephelus areolatus (Forsskål, 1775) Areolate grouper Serranidae	C	reef-associated	y	LC	5	Baalkhuyur et al. 2018	ad	5	y	20.00	1.00	n/a	Saudi Arabian coast	Red Sea
Epinephelus chlorostigma (Valenciennes, 1828) Brownspotted grouper Serranidae	C	reef-associated	y	LC	3	Baalkhuyur et al. 2018	ad	3	y	33.33	1.00	n/a	Saudi Arabian coast	Red Sea
Epinephelus epistictus (Temminck & Schlegel, 184 Dotted grouper Serranidae	n/a	demersal	y	LC	5	Baalkhuyur et al. 2018	n/a	5	y	20.00	1.00	n/a	Saudi Arabian coast	Red Sea
Epinephelus itajara (Lichtenstein, 1822) Atlantic goliath grouper Serranidae	C	reef-associated	y	VU	2	Pegado et al. 2018	n/a	2	n	0.00	0.00	n/a	Amazon River estuary, northern	Western South-Atlantic

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													coast of Brazil	
Epinephelus radiatus (Day, 1868) Oblique-banded grouper Serranidae	n/a	demersal	y	LC	7	Baalkhuyur et al. 2018	n/a	7	y	14.29	1.00	n/a	Saudi Arabian coast	Red Sea
Erisphex pottii (Steindachner, 1896) n/a Aploactinidae	n/a	demersal	n/a	LC	120	Sun et al. 2019	n/a	120	y	28.00	1.20	n/a	Yellow Sea, 53 locations	East China Sea
Etmopterus spinax (Linnaeus, 1758) Velvet belly Etmopteridae	C	bathy-demersal	y	LC	16	Anastasopoulou et al. 2013	n/a	16	y	6.30	n/a	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Eubleekeria splendens (Leiognathus splendens) (Cuvier, 1829) Splendid ponyfish Leiognathidae	C	demersal	y	LC	10	Azad et al. 2018	juv + ad	10	y	60.00	1.00	n/a	Sathing Phra District	Gulf of Thailand
Eucinostomus argenteus Baird & Girard, 1855 Silver mojarra Gerreidae	n/a	reef-associated	y	LC	52	Vendel et al. 2017	n/a	52	n	0.00	0.00	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Eucinostomus melanopterus (Bleeker, 1863) Flagfin mojarra Gerreidae	O	demersal	y	LC	162	Vendel et al. 2017	n/a, n/a	2, 19	n, n	0, 0	0, 0	n/a, n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
						Ramos et al. 2012	juv, sub-ad, ad	41, 56, 44	y, y, y	4,9, 8.9, 13.6	0.05, 0.07, 0.08	condition K (Richardson et al. 2011)	Goiana Estuary, Extractive Reserve Acaú-Goiana	Western South-Atlantic
Eugerres brasilianus (Cuvier, 1830) Brazilian mojarra Gerreidae	n/a	demersal	y	LC	304	Ramos et al. 2012	juv, sub-ad, ad	141, 72, 27	y, y, y	12.8, 16.7, 33.4	0.16, 0.18, 0.11	condition K (Richardson et al. 2011)	Goiana Estuary, Extractive Reserve Acaú-Goiana	Western South-Atlantic

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						Vendel et al. 2017	juv, juv	17, 47	n, y	0, n/a	0, 0.06	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Eupleurogrammus muticus (Gray, 183) Smallhead hairtail Trichiuridae	C	bentho-pelagic	y	n/a	15	Sun et al. 2019	n/a	15	y	33.00	1.00	n/a	Yellow Sea, 53 locations	East China Sea
Eutrigla gurnardus (Linnaeus, 1758) Grey gurnard Triglidae	C	demersal	y	n/a	180	Foekema et al. 2013	n/a	171	y	< 1%	n/a	condition based on morphometrics	various	North Sea
						Steer et al. 2017	juv	9	n	0.00	0.00	n/a	various	western English Channel
Gadus macrocephalus Tilesius, 1810 Pacific cod Gadidae	C	demersal	y	n/a	40	Sun et al. 2019	juv	40	y	40.00	1.10	n/a	Yellow Sea, 53 locations	East China Sea
Gadus morhua (Linnaeus, 1758) Atlantic cod Gadidae	O	bentho-pelagic	y	VU	1681	Liboiron et al. 2016	n/a	205	y	2.40	n/a	n/a	Eastern Coast of Newfoundland	Western North-Atlantic
						McGoran et al. 2018	ad, juv	1, 2	n, n	0, 0	0, 0	n/a, n/a	Thames estuary,	English Channel
						Bråte et al. 2016	n/a	302	y	3.00	n/a	n/a	Oslo, Bergen, Sorfjorden, Karihavet, Lofoten, Varangerfjorden, Norway	Norwegian Sea, Skagerrak
						Rummel et al. 2016	juv, ad	7, 74	n, y	0, 1.4	0, n/a	n/a, n/a	North Sea, Baltic Sea	
						Foekema et al. 2013	n/a	80	y	13.00	1.10	condition based on morphometrics	various	North Sea
						Liboiron et al. 2019	n/a	1010	y	1.68	1.12	n/a	Newfoundland	Western North-Atlantic

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Galeus melastomus Rafinesque, 1810 Blackmouth catshark Pentanchidae	C	demersal	y	LC	866	Anastasopoulou et al. 2013	n/a	741	y	3.20	n/a	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
						Alomar and Deudero 2017	juv	125	y	16.80	0.34	n/a	Mallorca	Mediterranean
Gambusia affinis (Baird & Girard, 1853) Mosquitofish Poeciliidae	P	bentho-pelagic	y	LC	5	Phillips and Bonner 2015	n/a	5	y	n/a	n/a	n/a	Laguna Madre, southern coast of Texas	Western North-Atlantic
Gempylus serpens Cuvier, 1829 Snake mackerel Gempylidae	C	oceanic	y	LC	104	Choy and Drazen 2013	ad	104	y	< 1%	n/a	n/a	n/a	Central North Pacific Ocean
Genidens genidens (Cuvier, 1829) Guri sea catfish Ariidae	n/a	demersal	y	LC	26	Dantas et al. 2019	juv	26	y	26.90	0.48	condition K (Richardson et al. 2011)	Southern Brazil, Santo Antonio dos Anjos + - Imarui Lagoons	Western South-Atlantic
Genyatremus luteus (Bloch, 1790) Torroto grunt Haemulidae	C	demersal	y	n/a	8	Pegado et al. 2018	n/a	8	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Gerres subfasciatus Cuvier, 1830 Common silver belly Gerreidae	n/a	demersal	n/a	LC	24	Halstead et al. 2018	n/a	24	y	21.00	0.10	n/a	Sydney Harbour, Australia	Southern West-Pacific
Girella laevifrons (Tschudi, 1846) n/a Kyphosidae	n/a	pelagic-neritic	n/a	n/a	16	Mizraji et al. 2017	juv	16	y	n/a	61.00	condition K in g cm-3	Las Cruces, Chile	Eastern South Pacific
Girella tricuspidata (Quoy & Gaimard, 1824) Parore Kyphosidae	n/a	bentho-pelagic	y	n/a	20	Markic et al. 2018	ad	20	y	70.00	5.90	n/a	Auckland	South Pacific

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Glyptocephalus cynoglossus (Linnaeus, 1758) Witch flounder Pleuronectidae	O	demersal	y	n/a	23	McGoran et al. 2018	juv	23	y	9.00	1.00	n/a	Firth of Clyde	Firth of Clyde
Gnathodentex aureolineatus (Lacepède, 1802) Striped large-eye bream Lethrinidae	C	reef-associated	y	LC	29	Markic et al. 2018	n/a	29	y	6.90	1.00	n/a	Tahiti	South Pacific
Gonostoma denudatum Rafinesque, 1810 n/a Gonostomatidae	n/a	bathy-pelagic	n	LC	5	Wieczorek et al. 2018	ad	5	y	100.00	2.20	n/a	Galway (Ireland) to St. John's (Newfoundland)	Eastern North-Atlantic
Graus nigra Philippi, 1887 n/a Kyphosidae	C	demersal	y		8	Mizraji et al. 2017	juv	8	y	n/a	10.00	condition K in g cm-3	Las Cruces, Chile	Eastern South Pacific
Gymnocranius grandoculis (Valenciennes, 1830) Blue-lined large-eye bream Lethrinidae	C	reef-associated	y	LC	10	Baalkhuyur et al. 2018	n/a	10	y	20.00	2.00	n/a	Saudi Arabian coast	Red Sea
Gymnoscopelus nicholsi (Gilbert, 1911) Nichol's lanternfish Myctophidae	P	meso-pelagic / bathy-pelagic	y	n/a	39	Cannon et al. 2016	n/a	39	n	0.00	0.00	n/a	Australia	Southern Pacific, Southern Ocean, Australia
Gymnothorax ocellatus Agassiz, 1831 Ocellated moray Muraenidae	C	reef-associated	y	LC	1	Pegado et al. 2018	n/a	1	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Haemulon plumierii (Lacepède, 1801) White grunt Haemulidae	C	reef-associated	y	LC	13	Pegado et al. 2018	n/a	13	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Haemulon steindachneri (Jordan & Gilbert, 1882) Chere-chere grunt Haemulidae	C	reef-associated	y	LC	5	Pegado et al. 2018	n/a	5	n	0.00	0.00	n/a	Amazon River estuary, northern	Western South-Atlantic

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													coast of Brazil	
Haemulopsis corvinaeformis (Steindachner, 1868) Roughneck grunt Haemulidae	C	demersal	y	LC	44	Silva et al. 2018b	sub-ad + ad	44	y	n/a	n/a	n/a	Goiana Estuary, upper, middle and lower	Western South-Atlantic
Haletta semifasciata (Valenciennes, 1840) Blue weed whiting Odacidae	C	demersal	y	LC	2	Cannon et al. 2016	n/a	2	n	0.00	0.00	n/a	Australia	Southern Pacific, Southern Ocean, Australia
Harpadon nehereus (Hamilton, 1822) Bombay-duck Synodontidae	C	bentho-pelagic	y	n/a	36	Jabeen et al. 2017	ad	18	y	100.00	3.80	n/a	Yangtze estuary, East China Sea and South China Sea	China Sea
						Su et al. 2019a	n/a	18	y	83.00	2.50	n/a	4 locations in Hangzhou Bay and Yangtze Estuary	East China Sea
Helcogrammoides chilensis (Cancino, 1960) n/a Tripterygiidae	P	demersal	n/a		3	Mizraji et al. 2017	juv	3	y	n/a	10.00	condition K in g cm-3	Las Cruces, Chile	Eastern South Pacific
Helicolenus dactylopterus (Delaroche, 1809) Blackbelly rosefish Sebastidae	C	bathy-demersal	y	LC	381	Neves et al. 2015	juv	1	n	0.00	0.00	n/a	off the coast of Portugal	Eastern North Atlantic
						Anastasopoulou et al. 2013	n/a	380	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Hemibarbus maculatus Bleeker, 1871 Spotted steed Cyprinidae	C	bentho-pelagic	y	n/a	9	Su et al. 2019a	n/a	9	y	44.00	0.90	n/a	4 locations in Hangzhou Bay and Yangtze Estuary	East China Sea
Heteropriacanthus cruentatus (Lacepède, 1801) Glasseye Priacanthidae	C	reef-associated	y	LC	10	Markic et al. 2018	n/a	10	y	30.00	1.00	n/a	Rapa Nui	South Pacific

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Hexagrammos otakii Jordan & Starks, 1895 Fat greenling Hexagrammidae	n/a	demersal	y	n/a	40	Sun et al. 2019	n/a	40	y	33.00	1.20	n/a	Yellow Sea, 53 locations	East China Sea
Hippoglossoides platessoides (Fabricius, 1780) American plaice Pleuronectidae	C	demersal	y	n/a	138	Bour et al. 2018	n/a	20	y	55.00	n/a	n/a	Oslofjord	Skagerrak
						McGoran et al. 2018	juv	104	y	23.00	2.67	n/a	Firth of Clyde	Firth of Clyde
						Murphy et al. 2017	n/a	14	n	0.00	0.00	n/a	Scottish offshore waters	North Sea, Eastern North Atlantic
Hygophum benoiti (Cocco, 1838) Benoit's lanternfish Myctophidae	n/a	bathy-pelagic / high-oceanic	n/a	LC	73	Romeo et al. 2016	n/a	73	y	6.80	n/a	n/a	Sicilian coast	Central Mediterranean Sea
Hygophum proximum Becker, 1965 Firefly lanternfish Myctophidae	n/a	meso-pelagic / oceanic	n/a	LC	2	Davison and Asch 2011	ad	2	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Hygophum reinhardtii (Lütken, 1892) Reinhardt's lantern fish Myctophidae	n/a	meso-pelagic	n/a	LC	49	Boerger et al. 2010	n/a	47	y	n/a	1.30	n/a	various	North Pacific Central Gyre
						Davison and Asch 2011	ad	2	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Hypanus americanus (Hildebrand & Schroeder, 1928) Southern stingray Dasyatidae	C	reef-associated	y	DD	3	Miranda and de Carvalho-Souza 2016	n/a	3	n	0.00	0.00	n/a	Salvador, Brazil	Western South-Atlantic
Hyperlophus vittatus (Castelnau, 1875) Sandy sprat Clupeidae	n/a	pelagic-neritic	y	LC	10	Cannon et al. 2016	n/a	10	n	0.00	0.00	n/a	Australia	Southern Pacific, Southern Ocean, Australia
Hyporhamphus ihi Phillipps, 1932 n/a Hemiramphidae	P	pelagic-neritic	y	n/a	24	Markic et al. 2018	n/a	24	n	0.00	0.00	n/a	Auckland	South Pacific
Hyporhamphus intermedius (Cantor, 1842) Asian pencil halfbeak Hemiramphidae	P	pelagic-neritic	y	n/a	18	Jabeen et al. 2017	ad	18	y	100.00	3.70	n/a	Yangtze estuary, East China Sea	China Sea

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													and South China Sea	
Hyporhamphus roberti roberti (Valenciennes, 1847) Slender halfbeak Hemiramphidae	n/a	n/a	y	LC	31	Vendel et al. 2017	n/a	31	y	n/a	0.03	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Hyporhamphus unifasciatus (Ranzani, 1841) Common halfbeak Hemiramphidae	O	reef-associated	y	LC	161	Vendel et al. 2017	juv	161	y	n/a	0.14	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Hyporhamphus melanochir (Valenciennes, 1847) Southern garfish Hemiramphidae	H	0 - 20 m	y	n/a	66	Cannon et al. 2016	ad	66	n	0.00	0.00	n/a	Australia	Southern Pacific, Southern Ocean, Australia
Idiacanthus antrostomus Gilbert, 1890 Pacific blackdragon Stomiidae	C	meso-pelagic	n/a	n/a	4	Davison and Asch 2011	ad	4	y	25.00	n/a	n/a	various	North Pacific Subtropical Gyre
Johnius borneensis (Bleeker, 1851) Sharpnose hammer croaker Sciaenidae	C	bentho-pelagic	y	n/a	3	Azad et al. 2018	juv	3	y	67.00	1.00	n/a	Sathing Phra District	Gulf of Thailand
Johnius carouna (Cuvier, 1830) Caroun croaker Sciaenidae	n/a	bentho-pelagic	y	n/a	20	Azad et al. 2018	n/a	20	y	85.00	3.80	n/a	Sathing Phra District	Gulf of Thailand
Katsuwonus pelamis (Linnaeus, 1758) Skipjack tuna Scombridae	C	pelagic-oceanic	y	LC	65	Cannon et al. 2016	ad	1	n	0.00	0.00	n/a	Australia	Southern Pacific, Southern Ocean, Australia
						Rochman et al. 2015	n/a	9	n	0.00	0.00	n/a	Makassar, Sulawesi, Indonesia;	Makassar Strait
						Choy and Drazen 2013	ad	29	n	0.00	0.00	n/a	n/a	Central North Pacific Ocean
						Markic et al. 2018	ad	26	y	23.10	1.50	n/a	Samoa	South Pacific
Konosirus punctatus (Temminck & Schlegel, 184	n/a	pelagic-neritic	y	LC	3	Zhu et al. 2019b	n/a	3	y	100.00	2.00	n/a	various	Maowei Sea

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Dotted gizzard shad Clupeidae														
Kyphosus sandwicensis (Sauvage, 1880) Pacific chub Kyphosidae	H	reef-associated	n/a	n/a	39	Markic et al. 2018	n/a	39	y	51.30	4.00	n/a	Rapa Nui	South Pacific
Labrus mixtus Linnaeus, 1758 Cuckoo wrasse Labridae	C	reef-associated	y	LC	1	Steer et al. 2017	juv	1	n	0.00	0.00	n/a	various	western English Channel
Lagocephalus laevigatus (Linnaeus, 1766) Smooth puffer Tetraodontidae	C	pelagic	y	LC	1	Miranda and de Carvalho-Souza 2016	n/a	1	n	0.00	0.00	n/a	Salvador, Brazil	Western South-Atlantic
Lagocephalus spadiceus (Richardson, 1845) Half-smooth golden pufferfis Tetraodontidae	n/a	demersal	n	LC	1	Güven et al. 2017	n/a	1	n	0.00	0.00	n/a	Turkish territorial waters	Mediterranean
Lagodon rhomboides (Linnaeus, 1766) Pinfish Sparidae	C	demersal	y	LC	497	Peters et al. 2017	ad	449	y	46.50	2.07	n/a	Texas (TX) Gulf Coast, Galveston Bay to Freeport	Western North-Atlantic
						Phillips and Bonner 2015	n/a	48	y	n/a	n/a	n/a	Laguna Madre, southern coast of Texas	Western North-Atlantic
Lampadena urophaos Paxton, 1963 Sunbeam lampfish Myctophidae	n/a	epipelagic / meso-pelagic	n/a	n/a	1	Davison and Asch 2011	ad	1	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Lampanyctus crocodilus (Risso, 1810) Jewel lanternfish Myctophidae	P	bathy-pelagic / high-oceanic	n	LC	2	Lusher et al. 2016	n/a	2	n	0.00	0.00	n/a	various	Eastern North-Atlantic
Lampanyctus macdonaldi (Goode & Bean, 1896) Rakery beaconlamp Myctophidae	C	oceanic / meso-pelagic / bathy-pelagic	n/a	LC	16	Wieczorek et al. 2018	ad	16	y	75.00	1.75	n/a	Galway (Ireland) to St. John's (Newfoundland)	Eastern North-Atlantic

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Lampris sp. big eye n/a n/a Lampridae	n/a	pelagic	n/a	n/a	115	Choy and Drazen 2013	ad	115	y	43.00	2.30	n/a	n/a	Central North Pacific Ocean
Lampris sp. small eye n/a n/a Lampridae	n/a	pelagic	n/a	n/a	24	Choy and Drazen 2013	ad	24	y	58.00	5.80	n/a	n/a	Central North Pacific Ocean
Larimichthys crocea (Richardson, 1846) Large yellow croaker Sciaenidae	C	bentho-pelagic	y	n/a	18	Jabeen et al. 2017	ad	18	y	100.00	4.60	n/a	Yangtze estuary, East China Sea and South China Sea	China Sea
Larimichthys polyactis (Bleeker, 1877) Yellow croaker Sciaenidae	n/a	bentho-pelagic	y	n/a	30	Sun et al. 2019	juv	30	y	37.00	2.60	n/a	Yellow Sea, 53 locations	East China Sea
Lateolabrax japonicus (Cuvier, 1828) Japanese seabass Lateolabracidae	P	reef-associated	y	n/a	18	Jabeen et al. 2017	juv	18	y	100.00	2.10	n/a	Yangtze estuary, East China Sea and South China Sea	China Sea
Leiognathus berbis (Valenciennes, 1835) Berber ponyfish Leiognathidae	C	demersal	y	n/a	8	Azad et al. 2018	n/a	8	y	50.00	0.90	n/a	Sathing Phra District	Gulf of Thailand
Lepidocybium flavobrunneum (Smith, 1843) Escolar Gempylidae	C	bentho-pelagic	y	LC	45	Choy and Drazen 2013	ad	45	n	0.00	0.00	n/a	n/a	Central North Pacific Ocean
Lepidopus caudatus (Euphrasen, 1788) Silver scabbardfish Trichiuridae	C	bentho-pelagic	y	DD	1	Anastasopoulou et al. 2013	n/a	1	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Lepidorhombus boscii (Risso, 1810) Four-spot megrim Scophthalmidae	n/a	demersal	y	n/a	2	Neves et al. 2015	n/a	2	y	50.00	n/a	n/a	off the coast of Portugal	Eastern North Atlantic
Lepidorhombus whiffiagonis (Walbaum, 1792)	C	bathy-demersal	y	n/a	16	Murphy et al. 2017	n/a	10	y	10.00	0.10	n/a	Scottish offshore waters	North Sea, Eastern North Atlantic

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Megrim Scophthalmidae						Neves et al. 2015	juv	2	n	0.00	0.00	n/a	off the coast of Portugal	Eastern North Atlantic
						Steer et al. 2017	juv	4	n	0.00	0.00	n/a	various	western English Channel
Lepidotrigla guentheri Hilgendorf, 1879 n/a Triglidae	n/a	demersal	n/a	n/a	4	Zhu et al. 2019a	n/a	4	y	100.00	n/a	n/a	various	South China Sea
Lethrinus amboinensis Bleeker, 1854 Ambon emperor Lethrinidae	C	reef-associated	y	LC	26	Markic et al. 2018	n/a	26	y	23.10	1.70	n/a	Samoa	South Pacific
Lethrinus microdon Valenciennes, 1830 Smalltooth emperor Lethrinidae	C	reef-associated	y	LC	10	Baalkhuyur et al. 2018	juv + ad	10	y	20.00	2.00	n/a	Saudi Arabian coast	Red Sea
Lethrinus obsoletus (Forsskål, 1775) Orange-striped emperor Lethrinidae	C	reef-associated	y	LC	30	Markic et al. 2018	ad	30	y	13.30	1.30	n/a	Samoa	South Pacific
Limanda limanda (Linnaeus, 1758) Common dab Pleuronectidae	C	demersal	y	LC	545	Hermsen et al. 2017	ad	100	n	0.00	0.00	n/a	Southern Bight	North Sea
						McGoran et al. 2018	ad, juv + ad	1, 307	y, y	100, 50	1, 4.5	n/a, n/a	Thames estuary, Firth of Clyde	English Channel, Firth of Clyde
						Murphy et al. 2017	n/a	19	y	47.00	1.30	n/a	coastal waters of Scotland to the east (Firth of Forth) and west (Clyde Estuary and Firth of Clyde)	North Sea, Eastern North Atlantic
						Rummel et al. 2016	ad, ad	74, 15	y, n	5.4, 0	n/a, 0	n/a, n/a	various	North Sea, Baltic Sea
						Steer et al. 2017	juv	29	n	0.00	0.00	n/a	various	western English Channel

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Liparis liparis (Linnaeus, 1766) Striped seasnail Liparidae	C	demersal	n/a	LC	50	Kartar et al. 1976	n/a	50	y	6.50	n/a	n/a	Oldbury, Hinkley	Eastern Nord-Atlantic
Liparis tanakae (Gilbert & Burke, 1912) Tanaka's snailfish Liparidae	n/a	demersal	n/a	n/a	245	Sun et al. 2019	n/a	245	y	32.00	1.10	n/a	Yellow Sea, 53 locations	East China Sea
Lipocheilus carnolabrum (Chan, 1970) Tang's snapper Lutjanidae	C	demersal	y	LC	7	Baalkhuyur et al. 2018	n/a	7	y	28.57	2.00	n/a	Saudi Arabian coast	Red Sea
Lithognathus mormyrus (Linnaeus, 1758) Sand steenbras Sparidae	C	demersal	y	LC	46	Güven et al. 2017	n/a	46	y	35.00	1.88	n/a	Turkish territorial waters	Mediterranean
Lobianchia gemellarii (Cocco, 1838) Cocco's lantern fish Myctophidae	n/a	high-oceanic / meso-pelagic	n/a	LC	3	Davison and Asch 2011	ad	3	y	33.30	n/a	n/a	various	North Pacific Subtropical Gyre
Lophius litulon (Jordan, 1902) Yellow goosefish Lophiidae	C	bathy-demersal	y	n/a	20	Sun et al. 2019	juv	20	y	25.00	1.00	n/a	Yellow Sea, 53 locations	East China Sea
Lophius piscatorius Linnaeus, 1758 Angler Lophiidae	C	bathy-demersal	y	LC	2	Neves et al. 2015	n/a	2	y	50.00	0.50	n/a	off the coast of Portugal	Eastern North Atlantic
Loweina interrupta (Tåning, 1928) n/a Myctophidae	n/a	high-oceanic / meso-pelagic	n/a	LC	27	Boerger et al. 2010	n/a	27	y	n/a	1.00	n/a	various	North Pacific Central Gyre
Lutjanus analis (Cuvier, 1828) Mutton snapper Lutjanidae	C	reef-associated	y	NT	5	Miranda and de Carvalho-Souza 2016	n/a	2	n	0.00	0.00	n/a	Salvador, Brazil	Western South-Atlantic
						Pegado et al. 2018	n/a	3	y	33.30	1.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic

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Lutjanus campechanus (Poey, 1860) Northern red snapper Lutjanidae	C	reef-associated	y	VU	2	Phillips and Bonner 2015	n/a	2	y	n/a	n/a	n/a	Laguna Madre, southern coast of Texas	Western North-Atlantic
Lutjanus gibbus (Forsskål, 1775) Humpback red snapper Lutjanidae	C	reef-associated, bentho-pelagic	y	LC	34	Rochman et al. 2015	n/a	5	n	0.00	0.00	n/a	Makassar, Sulawesi, Indonesia;	Makassar Strait
						Markic et al. 2018	ad	29	y	20.70	1.70	n/a	Samoa	South Pacific
Lutjanus griseus (Linnaeus, 1758) Grey snapper Lutjanidae	C	reef-associated	y	LC	5	Phillips and Bonner 2015	n/a	5	y	n/a	n/a	n/a	Laguna Madre, southern coast of Texas	Western North-Atlantic
Lutjanus jocu (Bloch & Schneider, 1801) Dog snapper Lutjanidae	C	reef-associated	y	DD	5	Miranda and de Carvalho-Souza 2016	n/a	5	n	0.00	0.00	n/a	Salvador, Brazil	Western South-Atlantic
Lutjanus kasmira (Forsskål, 1775) Common bluestripe snapper Lutjanidae	O	reef-associated	y	LC	12	Baalkhuyur et al. 2018	ad	12	y	16.67	2.00	n/a	Saudi Arabian coast	Red Sea
Lutjanus synagris (Linnaeus, 1758) Lane snapper Lutjanidae	C	reef-associated	y	NT	2	Pegado et al. 2018	n/a	2	y	50.00	1.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Lycengraulis grossidens (Spix & Agassiz, 1829) Atlantic sabretooth anchovy Engraulidae	C	pelagic-neritic	y	LC	50	Vendel et al. 2017	n/a	50	y	n/a	0.20	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Macrodon ancylodon (Bloch & Schneider, 1801) King weakfish Sciaenidae	C	demersal	y	LC	13	Pegado et al. 2018	n/a	13	y	7.70	2.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Malakichthys griseus Döderlein, 1883	n/a	pelagic-oceanic	n/a	n/a	1	Zhu et al. 2019a	n/a	1	y	100.00	n/a	n/a	various	South China Sea

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n/a Acropomatidae														
Mallotus villosus (Müller, 1776) Capelin Osmeridae	C	pelagic-oceanic	y	n/a	350	Liboiron et al. 2019	n/a	350	n	0.00	0.00	n/a	Newfoundland	Western North-Atlantic
Maurolicus mucronatus Klunzinger, 1871 n/a Sternoptychidae	n/a	bathy-pelagic	n/a	n/a	10	Baalkhuyur et al. 2018	n/a	10	y	100.00	1.00	n/a	Saudi Arabian coast	Red Sea
Maurolicus muelleri (Gmelin, 1789) Silvery lightfish Sternoptychidae	P	bathy-pelagic	y	n/a	282	Lusher et al. 2016	n/a	282	y	2.80	1.10	n/a	various	Eastern North-Atlantic
Megalaspis cordyla (Linnaeus, 1758) Torpedo scad Carangidae	C	oceanic	y	LC	29	Azad et al. 2018	juv	29	y	79.00	1.60	n/a	Sathing Phra District	Gulf of Thailand
Melanogrammus aeglefinus (Linnaeus, 1758) Haddock Gadidae	C	demersal	y	VU	103	McGoran et al. 2018	juv	6	y	100.00	5.83	n/a	Firth of Clyde	Firth of Clyde
						Foekema et al. 2013	n/a	97	y	6.20	1.00	condition based on morphometrics	various	North Sea
Menidia menidia (Linnaeus, 1766) Atlantic silverside Atherinopsidae	C	pelagic-neritic	y	LC	9	Carpenter et al. 1972	juv	9	y	33.00	n/a	n/a	Southern New England	Western North-Atlantic
Menticirrhus americanus (Linnaeus, 1758) Southern kingcroaker Sciaenidae	n/a	demersal	y	LC	151	Peters et al. 2017	juv + ad	150	y	35.30	1.62	n/a	Texas (TX) Gulf Coast, Galveston Bay to Freeport	Western North-Atlantic
						Pegado et al. 2018	n/a	1	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Merlangius merlangus (Linnaeus, 1758)	C	bentho-pelagic	y	LC	341	Foekema et al. 2013	n/a	105	y	5.70	1.30	condition based on	various	North Sea

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Whiting Gadidae												morphometrics		
						Hermsen et al. 2017	ad	28	n	0.00	0.00	n/a	Southern Bight	North Sea
						Lusher et al. 2013	n/a	50	y	32.00	1.70	n/a	10 km southwest of Plymouth	English Channel
						McGoran et al. 2018	juv + ad	29	y	10.00	4.67	n/a	Thames estuary	English Channel
						Steer et al. 2017	juv	129	y	3.90	n/a	n/a	various	western English Channel
Merluccius bilinearis (Mitchill, 1814) Silver hake Merlucciidae	C	demersal	y	NT	134	Liboiron et al. 2018	ad	134	n	0.00	0.00	n/a	South Coast of Newfoundland, Canada (Burgeo Bank, St. Pierre Bank, Southern Grand Bank)	Western North-Atlantic
Merluccius merluccius (Linnaeus, 1758) European hake Merlucciidae	C	demersal	y	LC	157	Anastasopoulou et al. 2013	n/a	36	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
						Giani et al. 2019	ad	97	y	26.80	1.38	n/a	around Italy, 3 different sites	Mediterranean
						Neves et al. 2015	juv, n/a	7, 5	y, y	29, 20	0.29, 0.4	n/a, n/a	off the coast of Portugal	Eastern North Atlantic
						Bellas et al. 2016	ad	12	y	16.70	1.20	n/a	Coast of Spain	East-Atlantic and Mediterranean
Meuschenia scaber (Forster, 1801) Velvet leatherjacket Monacanthidae	C	demersal	y	n/a	19	Markic et al. 2018	n/a	19	y	36.80	2.00	n/a	Auckland	South Pacific
Microchirus variegatus (Donovan, 1808)	C	demersal	y	LC	67	Lusher et al. 2013	n/a	51	y	23.50	1.60	n/a	10 km southwest of Plymouth	English Channel

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Thickback sole Soleidae						Steer et al. 2017	juv	16	y	12.50	n/a	n/a	various	western English Channel
Micromesistius poutassou (Risso, 1827) Blue whiting Gadidae	C	bathy-pelagic	y	n/a	50	Lusher et al. 2013	n/a	27	y	51.90	2.10	n/a	10 km southwest of Plymouth	English Channel
						Murphy et al. 2017	n/a	20	n	0.00	0.00	n/a	Scottish offshore waters	North Sea, Eastern North Atlantic
						Anastasopoulou et al. 2013	n/a	3	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Micropogonias furnieri (Desmarest, 1823) Whitemouth croaker Sciaenidae	C	demersal	y	LC	26	Pegado et al. 2018	n/a	6	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
						Arias et al. 2019	juv	20	y	100.00	12.10	Condition factor CF=(TWx100/TL^3) and Hepatosomatic index HSI=(LWx100)/TW with liver weight	Bahia Blanca Estuary, Argentina	Western South-Atlantic
Micropogonias undulatus (Linnaeus, 1766) Atlantic croaker Sciaenidae	C	demersal	y	LC	384	Peters et al. 2017	juv + ad	383	y	45.20	1.93	n/a	Texas (TX) Gulf Coast, Galveston Bay to Freeport	Western North-Atlantic
						Phillips and Bonner 2015	n/a	1	n	0.00	0.00	n/a	Laguna Madre, southern coast of Texas	Western North-Atlantic
Microstomus kitt (Walbaum, 1792)	C	demersal	y	n/a	10	Steer et al. 2017	juv	10	n	0.00	0.00	n/a	various	western English Channel

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Lemon sole Pleuronectidae														
Molva macrophthalma (Rafinesque, 1810) Spanish ling Lotidae	n/a	demersal	n	LC	13	Anastasopoulou et al. 2013	n/a	13	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Molva molva (Linnaeus, 1758) Ling Lotidae	C	demersal	y	n/a	5	Murphy et al. 2017	n/a	5	n	0.00	0.00	n/a	Scottish offshore waters	North Sea, Eastern North Atlantic
Mora moro (Risso, 1810) Common mora Moridae	O	bathy-pelagic	y	LC	19	Anastasopoulou et al. 2013	n/a	19	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Morone saxatilis (Walbaum, 1792) Striped bass Moronidae	C	demersal	y	LC	7	Rochman et al. 2015	n/a	7	y	28.60	0.90	n/a	Half Moon Bay, California, USA	Eastern North-Pacific
Mugil cephalus Linnaeus, 1758 Flathead grey mullet Mugilidae	O	bentho-pelagic	y	LC	237	Phillips and Bonner 2015	n/a	9	n	0.00	0.00	n/a	Laguna Madre, southern coast of Texas	Western North-Atlantic
						Jabeen et al. 2017	juv	18	y	100.00	3.70	n/a	Yangtze estuary, East China Sea and South China Sea	China Sea
						Cannon et al. 2016	juv	43	n	0.00	0.00	n/a	Australia	Southern Pacific, Southern Ocean, Australia
						Naidoo et al. 2016	juv + sub-ad	70	y	72.80	3.80	n/a	Bayhead Mangroves of Durban Harbour	Indian Ocean
						Markic et al. 2018	n/a	22	y	13.60	2.00	n/a	Auckland	South Pacific

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						Cheung et al. 2018	ad	30	y	60.00	4.30	n/a	Sam Mun Tsai, eastern coast of Hong Kong	South China Sea
						Halstead et al. 2018	juv	45	y	64.00	2.50	n/a	Sydney Harbour, Australia	Southern West-Pacific
Mugil curema Valenciennes, 1836 White mullet Mugilidae	P	reef-associated	y	LC	100	Vendel et al. 2017	juv	100	y	n/a	0.01	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Mugil hospes Jordan & Culver, 1895 Hospe mullet Mugilidae	n/a	demersal	y	LC	28	Vendel et al. 2017	juv	28	n	0.00	0.00	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Mullus barbatus barbatus Linnaeus, 1758 Red mullet Mullidae	C	demersal	y	LC	665	Digka et al. 2018	juv + ad	25	y	32.00	n/a	n/a	Northern Ionian Sea	Mediterranean
						Anastasopoulou et al. 2018	ad, ad, ad, ad	48, 50, 50, 25	n, y, y, y	0, 14, 8, 32	0, 0.18, 0.08, 0.5	n/a, n/a, n/a, n/a	North + South Adriatic Sea, NE Ionian Sea	Mediterranean
						Bellas et al. 2016	ad	128	y	18.80	1.75	n/a	Coast of Spain	East-Atlantic and Mediterranean
						Giani et al. 2019	ad	132	y	19.70	1.08	n/a	around Italy, 3 different sites	Mediterranean
						Güven et al. 2017	n/a	207	y	66.00	2.12	n/a	Turkish territorial waters	Mediterranean
Mullus surmuletus Linnaeus, 1758 Surmullet Mullidae	C	demersal	y	LC	510	Alomar et al. 2017	ad	417	y	27.30	0.42	oxidative stress, cellular damage, GST activity	Mallorca	Mediterranean
						Anastasopoulou et al. 2018	ad, ad	8, 30	n, y	0, 70	0, 1.8	n/a, n/a	North Adriatic Sea, Croatian Sea	Mediterranean
						Neves et al. 2015	ad, n/a	1, 3	y, y	100, 100	2, 1.66	n/a, n/a	off the coast of Portugal	Eastern North Atlantic

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						Güven et al. 2017	n/a	51	y	65.00	1.82	n/a	Turkish territorial waters	Mediterranean
Muraenesox cinereus (Forsskål, 1775) Daggertooth pike conger Muraenesocidae	C	demersal	y	n/a	18	Jabeen et al. 2017	n/a	18	y	100.00	2.40	n/a	Yangtze estuary, East China Sea and South China Sea	China Sea
Mustelus canis (Mitchill, 1815) Dusky smooth-hound Triakidae	C	demersal	y	NT	2	Pegado et al. 2018	n/a	2	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Mustelus higmani Springer & Lowe, 1963 Smalleye smooth-hound Triakidae	C	demersal	y	LC	3	Pegado et al. 2018	n/a	3	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Mycteroperca sp. Gill, 1862 n/a Serranidae	n/a	n/a	n/a	n/a	1	Miranda and de Carvalho-Souza 2016	n/a	1	n	0.00	0.00	n/a	Salvador, Brazil	Western South-Atlantic
Myctophum aurolaternatum Garman, 1899 Golden lanternfish Myctophidae	n/a	high-oceanic / meso-pelagic	n/a	n/a	462	Boerger et al. 2010	n/a	462	y	n/a	6.00	n/a	various	North Pacific Central Gyre
Myctophum nitidulum Garman, 1899 Pearly lanternfish Myctophidae	n/a	high-oceanic / meso-pelagic	n/a	LC	25	Davison and Asch 2011	ad	25	y	16.00	n/a	n/a	various	North Pacific Subtropical Gyre
Myctophum punctatum Rafinesque, 1810 Spotted lanternfish Myctophidae	P	bathy-pelagic / meso-pelagic / high-oceanic	n/a	LC	160	Lusher et al. 2016	n/a	3	n	0.00	0.00	n/a	various	Eastern North-Atlantic
						Wieczorek et al. 2018	ad	86	y	74.42	2.28	n/a	Galway (Ireland) to St. John's (Newfoundland)	Eastern North-Atlantic
						Romeo et al. 2016	juv + ad	71	y	4.20	n/a	n/a	Sicilian coast	Central Mediterranean Sea

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Myoxocephalus aenaeus (Mitchill, 1814) Grubby Cottidae	C	demersal	n/a	n/a	47	Carpenter et al. 1972	juv	47	y	4.20	n/a	n/a	Southern New England	Western North-Atlantic
Myoxocephalus scorpius (Linnaeus, 1758) Shorthorn sculpin Cottidae	C	demersal	n	n/a	5	McGoran et al. 2018	juv + ad	5	n	0.00	0.00	n/a	Firth of Clyde	Firth of Clyde
Nannobrachium fernae (Wisner, 1971) n/a Myctophidae	n/a	bathy-pelagic	n/a	n/a	1	Davison and Asch 2011	ad	1	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Nannobrachium hawaiiensis Zahuranec, 2000 n/a Myctophidae	n/a	meso-pelagic	n/a	n/a	17	Davison and Asch 2011	ad	17	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Nannobrachium regale (Gilbert, 1892) Pinpoint lampfish Myctophidae	P	epipelagic / meso-pelagic	n/a	n/a	1	Davison and Asch 2011	ad	1	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Narcine brasiliensis (Olfers, 1831) Brazilian electric ray Narcinidae	C	reef-associated	y	DD	6	Pegado et al. 2018	n/a	6	y	16.70	3.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Naso lituratus (Forster, 1801) Orangespine unicornfish Acanthuridae	H	reef-associated	y	LC	28	Markic et al. 2018	n/a	28	y	14.30	1.80	n/a	Samoa	South Pacific
Naso unicornis (Forsskål, 1775) Bluespine unicornfish Acanthuridae	H	reef-associated	y	LC	32	Markic et al. 2018	n/a	30	y	16.70	1.40	n/a	Samoa	South Pacific
						Baalkhuyur et al. 2018	n/a	2	n	0.00	0.00	n/a	Saudi Arabian coast	Red Sea
Nemadactylus macropterus (Forster, 1801) Tarakihi Cheilodactylidae	C	demersal	y	n/a	23	Markic et al. 2018	ad	23	y	8.70	3.50	n/a	Auckland	South Pacific
Nemichthys scolopaceus Richardson, 1848	C	bathy-pelagic /	n	LC	1	Lusher et al. 2016	n/a	1	y	100.00	1.00	n/a	various	Eastern North-Atlantic

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Slender snipe eel Nemichthyidae		mesopelagic												
Nemipterus randalli Russell, 1986 Randall's threadfin bream Nemipteridae	n/a	demersal	y	n/a	135	Güven et al. 2017	n/a	135	y	55.00	2.24	n/a	Turkish territorial waters	Mediterranean
Neoniphon sammara (Forsskål, 1775) Sammara squirrelfish Holocentridae	C	reef-associated	y	LC	5	Baalkhuyur et al. 2018	ad	5	y	20.00	1.00	n/a	Saudi Arabian coast	Red Sea
Neoscopelus microchir Matsubara, 1943 Shortfin neoscopelid Neoscopelidae	n/a	bathypelagic	n	n/a	1	Zhu et al. 2019a	juv	1	y	100.00	n/a	n/a	various	South China Sea
Nettastoma melanurum Rafinesque, 1810 Blackfin sorcerer Nettastomatidae	C	bathypelagic	n	LC	1	Anastasopoulou et al. 2013	n/a	1	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Notarius grandicassis (Valenciennes, 1840) Thomas sea catfish Ariidae	n/a	demersal	y	LC	4	Pegado et al. 2018	n/a	4	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Notemigonus crysoleucas (Mitchill, 1814) Golden shiner Cyprinidae	O	demersal	y	LC	7	Phillips and Bonner 2015	n/a	7	n	0.00	0.00	n/a	Laguna Madre, southern coast of Texas	Western North-Atlantic
Notolabrus tetricus (Richardson, 1840) Blue-throated wrasse Labridae	n/a	reef-associated	n/a	LC	7	Cannon et al. 2016	n/a	7	n	0.00	0.00	n/a	Australia	Southern Pacific, Southern Ocean, Australia
Notolychnus valdiviae (Brauer, 1904) Topside lampfish Myctophidae	P	high-oceanic / mesopelagic	n/a	LC	3	Davison and Asch 2011	ad	3	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Notoscopelus kroyeri (Malm, 1861)	C	pelagic-oceanic	n/a	n/a	417	Lusher et al. 2016	n/a	417	y	14.60	1.10	n/a	various	Eastern North-Atlantic

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Lancet fish Myctophidae														
Notoscopelus resplendens (Richardson, 1845) Patchwork lampfish Myctophidae	n/a	high-oceanic / meso-pelagic	n/a	LC	3	Davison and Asch 2011	ad	3	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Odontesthes regia (Humboldt, 1821) Chilean silverside Atherinopsidae	C	pelagic-neritic	n/a	LC	9	Ory et al. 2018a	ad	9	y	11.10	0.10	n/a	Chile	South Eastern Pacific
Oligoplites saurus (Bloch & Schneider, 1801) Leatherjacket Carangidae	C	reef-associated	y	LC	16	Vendel et al. 2017	n/a	16	n	0.00	0.00	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Oncorhynchus tshawytscha (Walbaum, 1792) Chinook salmon Salmonidae	C	bentho-pelagic	y	n/a	78	Rochman et al. 2015	n/a	4	y	25.00	0.25	n/a	Half Moon Bay, California, USA	Eastern North-Pacific
						Collicutt et al. 2019	juv	74	y	59.00	1.15	n/a	Vancouver Island, Strait of Georgia, 4 locations	Eastern North-Pacific
Ophichthus cylindroideus (Ranzani, 1839) Dusky snake eel Ophichthidae	n/a	demersal	n/a	LC	1	Pegado et al. 2018	n/a	1	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Ophichthus ophis (Linnaeus, 1758) Spotted snake eel Ophichthidae	C	demersal	y	LC	1	Pegado et al. 2018	n/a	1	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Ophiodon elongatus Girard, 1854 Lingcod Hexagrammidae	C	demersal	y	n/a	11	Rochman et al. 2015	n/a	11	y	9.10	0.10	n/a	Half Moon Bay, California, USA	Eastern North-Pacific
Opisthonema libertate (Günther, 1867) Pacific thread herring Clupeidae	P	pelagic-neritic	y	LC	40	Ory et al. 2018a	n/a	40	n	0.00	0.00	n/a	Colombia, Ecuador	South Eastern Pacific

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Opisthonema oglinum (Lesueur, 1818) Atlantic thread herring Clupeidae	P	reef-associated	y	LC	26	Vendel et al. 2017	juv + ad	26	y	n/a	0.12	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Opisthopterus tardoore (Cuvier, 1829) Tardoore Pristigasteridae	C	pelagic-neritic	y	LC	3	Azad et al. 2018	n/a	3	y	33.00	2.00	n/a	Sathing Phra District	Gulf of Thailand
Oreochromis aureus (Steindachner, 1864) Blue tilapia Cichlidae	P	bentho-pelagic	y	n/a	4	Phillips and Bonner 2015	n/a	4	y	n/a	n/a	n/a	Laguna Madre, southern coast of Texas	Western North-Atlantic
Oreochromis niloticus (Linnaeus, 1758) Nile tilapia Cichlidae	H	bentho-pelagic	y	LC	5	Rochman et al. 2015	n/a	5	n	0.00	0.00	n/a	Makassar, Sulawesi, Indonesia;	Makassar Strait
Oreochromis sp. Günther, 1889 n/a Cichlidae	n/a	n/a	n/a	n/a	3	Zhu et al. 2019b	n/a	3	y	100.00	8.50	n/a	various	Maowei Sea
Orthopristis chrysoptera (Linnaeus, 1766) Pigfish Haemulidae	C	demersal	y	LC	157	Peters et al. 2017	n/a	157	y	26.80	2.00	n/a	Texas (TX) Gulf Coast, Galveston Bay to Freeport	Western North-Atlantic
Orthopristis ruber (Cuvier, 1830) Corocoro grunt Haemulidae	C	demersal	y	LC	2	Pegado et al. 2018	n/a	2	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Osmerus eperlanus (Linnaeus, 1758) European smelt Osmeridae	C	pelagic-neritic	y	LC	10	McGoran et al. 2017	ad	10	y	20.00	0.20	n/a	Thames estuary	English Channel
Oxyeleotris marmorata (Bleeker, 1852) Marble goby Eleotridae	C	demersal	y	LC	18	Jabeen et al. 2017	n/a	18	y	100.00	4.20	n/a	Yangtze estuary, East China Sea and South China Sea	China Sea

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Pagellus acarne (Risso, 1827) Axillary seabream Sparidae	O	bentho-pelagic	y	LC	53	Neves et al. 2015	ad	1	y	100.00	1.00	n/a	off the coast of Portugal	Eastern North Atlantic
						Güven et al. 2017	n/a	52	y	67.00	2.46	n/a	Turkish territorial waters	Mediterranean
Pagellus bogaraveo (Brünnich, 1768) Blackspot seabream Sparidae	O	bentho-pelagic	y	NT	84	Anastasopoulou et al. 2013	n/a	60	y	1.70	n/a	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
						Savoca et al. 2019	ad	24	y	12.50	n/a	n/a	Thyrrhenian Sea	Mediterranean
Pagellus erythrinus (Linnaeus, 1758) Common pandora Sparidae	O	bentho-pelagic	y	LC	217	Anastasopoulou et al. 2018	ad, ad, ad, ad	30, 50, 30, 19	y, y, y, y	3.3, 2, 50, 42.1	0.03, 0.02, 1, 0.8	n/a, n/a, n/a, n/a	North Adriatic Sea, NE Ionian Sea, Croatian Sea	Mediterranean
						Güven et al. 2017	n/a	54	y	52.00	1.21	n/a	Turkish territorial waters	Mediterranean
						Savoca et al. 2019	ad	15	y	6.70	n/a	n/a	Thyrrhenian Sea	Mediterranean
						Digka et al. 2018	juv + ad	19	y	42.10	n/a	n/a	Northern Ionian Sea	Mediterranean
Pagrus auratus (Forster, 1801) Silver seabream Sparidae	C	reef-associated	y	LC	22	Markic et al. 2018	ad	22	y	4.50	1.00	n/a	Auckland	South Pacific
Pagrus pagrus (Linnaeus, 1758) Red porgy Sparidae	C	bentho-pelagic	y	LC	9	Güven et al. 2017	n/a	9	y	78.00	1.86	n/a	Turkish territorial waters	Mediterranean
Pampus argenteus *(Pampus cinereus)* (Euphrasen, 1788) Silver pomfret Stromateidae	P	bentho-pelagic	y	n/a	37	Sun et al. 2019	juv	10	y	20.00	1.00	n/a	Yellow Sea, 53 locations	East China Sea
						Jabeen et al. 2017	juv + ad	18	y	100.00	3.00	n/a	Yangtze estuary, East China Sea and South China Sea	China Sea
						Su et al. 2019a	n/a	9	y	56.00	1.10	n/a	4 locations in Hangzhou Bay and	East China Sea

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													Yangtze Estuary	
Paralichthys brasiliensis (Ranzani, 1842) Brazilian flounder Paralichtthyidae	n/a	demersal	y	n/a	1	Miranda and de Carvalho-Souza 2016	n/a	1	n	0.00	0.00	n/a	Salvador, Brazil	Western South-Atlantic
Paralichthys lethostigma Jordan & Gilbert, 1884 Southern flounder Paralichthyidae	C	demersal	y	NT	8	Phillips and Bonner 2015	n/a	8	y	n/a	n/a	n/a	Laguna Madre, southern coast of Texas	Western North-Atlantic
Paralonchurus brasiliensis (Steindachner, 1875) Banded croaker Sciaenidae	C	demersal	y	LC	6	Pegado et al. 2018	n/a	6	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Parascolopsis eriomma (Jordan & Richardson, 1909) Rosy dwarf monocle bream Nemipteridae	C	demersal	y	n/a	5	Baalkhuyur et al. 2018	n/a	5	y	60.00	5.00	n/a	Saudi Arabian coast	Red Sea
Pelates quadrilineatus (Bloch, 1790) Fourlined terapon Terapontidae	C	reef-associated	y	n/a	135	Güven et al. 2017	n/a	135	y	65.00	2.27	n/a	Turkish territorial waters	Mediterranean
Peprilus paru (Linnaeus, 1758) American harvestfish Stromateidae	C	bentho-pelagic	y	LC	2	Pegado et al. 2018	n/a	2	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Perca fluviatilis Linnaeus, 1758 European perch Percidae	O	demersal	y	LC	6	Zhu et al. 2019b	n/a	6	y	100.00	5.30	n/a	various	Maowei Sea
Pholis fangi (Wang & Wang, 1935) n/a Pholidae	n/a	demersal	n/a	n/a	79	Sun et al. 2019	n/a	79	y	38.00	1.30	n/a	Yellow Sea, 53 locations	East China Sea
Pholis gunnellus (Linnaeus, 1758)	C	demersal	n	n/a	1	McGoran et al. 2018	n/a	1	n	0.00	0.00	n/a	Firth of Clyde	Firth of Clyde

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Rock gunnel Pholidae														
Photopectoralis bindus (Valenciennes, 1835) Orangefin ponyfish Leiognathidae	n/a	demersal	y	n/a	18	Jabeen et al. 2017	ad	18	y	100.00	4.10	n/a	Yangtze estuary, East China Sea and South China Sea	China Sea
Phrynorhombus norvegicus (Günther, 1862) Norwegian topknot Scophthalmidae	C	bentho-pelagic	y	LC	3	McGoran et al. 2018	n/a	1	n	0.00	0.00	n/a	Firth of Clyde	Firth of Clyde
						Steer et al. 2017	juv	2	n	0.00	0.00	n/a	various	western English Channel
Phycis blennoides (Brünnich, 1768) Greater forkbeard Phycidae	C	bentho-pelagic	y	n/a	46	Anastasopoulou et al. 2013	n/a	46	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Planiliza affinis *(Chelon affinis)* (Günther, 1861) Eastern keelback mullet Mugilidae	n/a	n/a	n/a	n/a	4	Zhu et al. 2019b	n/a	4	y	100.00	3.80	n/a	various	Maowei Sea
Planiliza haematocheila *(Liza haematocheila)* (Temminck & Schlegel, 184 So-iuy mullet Mugilidae	O	pelagic-neritic	y	n/a	35	Su et al. 2019a	n/a	17	y	53.00	1.25	n/a	4 locations in Hangzhou Bay and Yangtze Estuary	East China Sea
						Jabeen et al. 2017	juv	18	y	100.00	3.30	n/a	Yangtze estuary, East China Sea and South China Sea	China Sea
Platichthys flesus (Linnaeus, 1758) European flounder Pleuronectidae	C	demersal	y	LC	1427	Kartar et al. 1976	juv, juv, juv, juv, juv	54, 530, 100, 206, 200	y, y, y, y, n	5.5, 20.7, 10.1, 5, 0	n/a, n/a, n/a, n/a, 0	n/a, n/a, n/a, n/a, n/a,	Oldbury	Eastern Nord-Atlantic
						Kazour et al. 2018	juv	22	y	58.00	2.04	Fulton's K condition factor	Canche and Seine Estuary, French coast	English Channel
						Bessa et al. 2018b	juv	40	y	13.00	0.18	n/a	Portugal, Mondego estuary	Eastern North-Atlantic

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						McGoran et al. 2017	ad	66	y	75.00	0.85	n/a	Thames estuary	English Channel
						McGoran et al. 2018	juv + ad, ad	118, 8	y, n	35, 0	3.1, 0	n/a, n/a	Thames estuary, Clyde of Firth	English Channel, Clyde of Firth
						Rummel et al. 2016	ad, ad	16, 20	n, y	0, 10	0, n/a	n/a, n/a	Tiefe Rinne, Helgoland; Herwig Cruise	North Sea, Baltic Sea
						Murphy et al. 2017	n/a	47	y	51.00	0.80		coastal waters of Scotland	North Sea, Eastern North Atlantic
Platycephalus sp. n/a n/a Platycephalidae	n/a	n/a	n/a	n/a	3	Zhu et al. 2019b	n/a	3	y	100.00	10.00	n/a	various	Maowei Sea
Platycephalus bassensis Cuvier, 1829 Southern sand flathead Platycephalidae	C	demersal	y	n/a	10	Cannon et al. 2016	ad	10	n	0.00	0.00	n/a	Australia	Southern Pacific, Southern Ocean, Australia
Platycephalus indicus (Linnaeus, 1758) Bartail flathead Platycephalidae	n/a	reef-associated	y	DD	12	Abbasi et al. 2018	juv	12	y	n/a	2.30	n/a	various	Persian Gulf
Platycephalus laevigatus Cuvier, 1829 Rock flathead Platycephalidae	n/a	demersal	n/a	n/a	7	Cannon et al. 2016	ad	7	n	0.00	0.00	n/a	Australia	Southern Pacific, Southern Ocean, Australia
Plectorhinchus gaterinus (Forsskål, 1775) Blackspotted rubberlip Haemulidae	n/a	reef-associated	y	n/a	6	Baalkhuyur et al. 2018	n/a	6	y	33.33	2.00	n/a	Saudi Arabian coast	Red Sea
Pleuronectes platessa (Linnaeus, 1758) European plaice Pleuronectidae	C	demersal	y	LC	163	McGoran et al. 2018	juv	99	y	31.00	2.52	n/a	Firth of Clyde	Firth of Clyde
						Steer et al. 2017	juv	2	n	0.00	0.00	n/a	various	western English Channel

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						Murphy et al. 2017	juv	62	y	45.00	0.90	n/a	coastal waters of Scotland to the east (Firth of Forth) and west (Clyde Estuary and Firth of Clyde)	North Sea, Eastern North Atlantic
Poecilia vivipara Bloch & Schneider, 1801 n/a Poeciliidae	P	bentho-pelagic	n	n/a	38	Vendel et al. 2017	n/a	38	y	n/a	0.08	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Pogonias cromis (Linnaeus, 1766) Black drum Sciaenidae	C	demersal	y	LC	1	Phillips and Bonner 2015	n/a	1	n	0.00	0.00	n/a	Laguna Madre, southern coast of Texas	Western North-Atlantic
Pollachius pollachius (Linnaeus, 1758) Pollack Gadidae	n/a	bentho-pelagic	y	LC	14	Murphy et al. 2017	n/a	5	n	0.00	0.00	n/a	Scottish offshore waters	North Sea, Eastern North Atlantic
						Steer et al. 2017	juv	9	n	0.00	0.00	n/a	various	western English Channel
Pollachius virens (Linnaeus, 1758) Saithe Gadidae	P	demersal	y	n/a	1	Carpenter et al. 1972	juv	1	y	100.00	1.00	n/a	Southern New England	Western North-Atlantic
Polydactylus oligodon (Günther, 1860) Littlescale threadfin Polynemidae	n/a	demersal	y	LC	1	Pegado et al. 2018	n/a	1	y	100.00	3.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Polydactylus virginicus (Linnaeus, 1758) Barbu Polynemidae	O	demersal	y	LC	27	Vendel et al. 2017	n/a	14	n	0.00	0.00	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
						Pegado et al. 2018	n/a	13	n	0.00	0.00	n/a	Amazon River estuary,	Western South-Atlantic

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													northern coast of Brazil	
Polymetme elongata (Matsubara, 1938) n/a Phosichthyidae	n/a	bentho-pelagic	n/a	n/a	1	Zhu et al. 2019a	n/a	1	y	100.00	n/a	n/a	various	South China Sea
Polyprion americanus (Bloch & Schneider, 1801) Wreckfish Polyprionidae	C	demersal	y	CR	15	Neves et al. 2015	n/a	1	n	0.00	0.00	n/a	off the coast of Portugal	Eastern North Atlantic
						Anastasopoulou et al. 2013	n/a	14	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Pomacentrus moluccensis Bleeker, 1853 Lemon damsel Pomacentridae	O	reef-associated	n/a	n/a	60	Jensen et al. 2019	n/a	60	y	95.00	n/a	n/a	Great Barrier Reef World Heritage Area	Western South-Pacific
Pomadasys incisus (Bowdich, 1825) Bastard grunt Haemulidae	C	demersal	y	LC	29	Güven et al. 2017	n/a	29	y	55.00	1.44	n/a	Turkish territorial waters	Mediterranean
Pomadasys ramosus (Poey, 1860) n/a Haemulidae	n/a	pelagic-neritic	n/a	n/a	125	Silva et al. 2018b	juv + sub-ad + ad	125	y	n/a	n/a	n/a	Goiana Estuary, upper, middle and lower	Western South-Atlantic
Pomatomus saltatrix (Linnaeus, 1766) Bluefish Pomatomidae	C	pelagic-oceanic	y	VU	2	Pegado et al. 2018	n/a	2	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Pomatoschistus minutus (Pallas, 1770) Sand goby Gobiidae	C	demersal	n	LC	50	Kartar et al. 1976	n/a	50	y	5.00	n/a	n/a	Oldbury	Eastern Nord-Atlantic
Priacanthus arenatus Cuvier, 1829 Atlantic bigeye Priacanthidae	C	reef-associated	y	LC	122	Cardozo et al. 2018	ad	122	y	49.17	6.00	Fulton's K condition factor	Southern Brazil near Garopaba,	Western South-Atlantic

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													Santa Catarina	
Prionace glauca (Linnaeus, 1758) Blue shark Carcharhinidae	O	epipelagic / oceanic	y	NT	43	Bernardini et al. 2018	juv	43	y	34.88	range from 1 to 30 litter particles	n/a	offshore area in front of the coast of Sanremo, Imperia and Nice	Mediterranean
Prionotus evolans (Linnaeus, 1766) Striped searobin Triglidae	C	reef-associated	y	LC	1	Carpenter et al. 1972	n/a	1	y	100.00	1.00	n/a	Southern New England	Western North-Atlantic
Pristipomoides multidens (Day, 1871) Goldbanded jobfish Lutjanidae	C	demersal	y	LC	10	Baalkhuyur et al. 2018	juv	10	y	20.00	2.00	n/a	Saudi Arabian coast	Red Sea
Pristipomoides typus Bleeker, 1852 Sharptooth jobfish Lutjanidae	C	demersal	y	LC	5	Baalkhuyur et al. 2018	juv + ad	5	n	0.00	0.00	n/a	Saudi Arabian coast	Red Sea
Psenopsis anomala (Temminck & Schlegel, 184 Pacific rudderfish Centrolophidae	n/a	bentho-pelagic	y	LC	28	Jabeen et al. 2017	ad	18	y	100.00	1.10	n/a	Yangtze estuary, East China Sea and South China Sea	China Sea
						Sun et al. 2019	juv	10	y	20.00	1.00	n/a	Yellow Sea, 53 locations	East China Sea
Pseudopleuronectes americanus (Walbaum, 1792) Winter flounder Pleuronectidae	C	demersal	y	n/a	95	Carpenter et al. 1972	juv	95	y	2.10	n/a	n/a	Southern New England	Western North-Atlantic
Pteroplatytrygon violacea (Bonaparte, 1832) Pelagic stingray Dasyatidae	C	pelagic	n/a	LC	2	Anastasopoulou et al. 2013	n/a	2	y	50.00	n/a	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Pygoplites diacanthus (Boddaert, 1772) Regal angelfish Pomacanthidae	C	reef-associated	y	LC	5	Baalkhuyur et al. 2018	n/a	5	n	0.00	0.00	n/a	Saudi Arabian coast	Red Sea

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Rachycentron canadum (Linnaeus, 1766) Cobia Rachycentridae	C	reef-associated	y	LC	1	Pegado et al. 2018	n/a	1	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Raja asterias Delaroche, 1809 Mediterranean starry ray Rajidae	C	demersal	y	NT	7	Neves et al. 2015	n/a	7	y	43.00	0.57	n/a	off the coast of Portugal	Eastern North Atlantic
Raja clavata (Linnaeus, 1758) Thornback ray Rajidae	C	demersal	y	NT	9	McGoran et al. 2018	juv + ad	7	y	14.00	1.00	n/a	Thames estuary,	English Channel
						Anastasopoulou et al. 2013	n/a	2	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Raja oxyrinchus Linnaeus, 1758 Longnosed skate Rajidae	C	bathy-demersal	y	NT	10	Anastasopoulou et al. 2013	n/a	10	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Rastrelliger brachysoma (Bleeker, 1851) Short mackerel Scombridae	P	pelagic-neritic	y	DD	3	Azad et al. 2018	ad	3	y	33.00	1.00	n/a	Sathing Phra District	Gulf of Thailand
Rastrelliger kanagurta (Cuvier, 1816) Indian mackerel Scombridae	P	pelagic-neritic	y	DD	29	Rochman et al. 2015	n/a	9	y	55.60	1.00	n/a	Makassar, Sulawesi, Indonesia;	Makassar Strait
						Kumar et al. 2018	ad	20	y	n/a	n/a	n/a	Tamil Nadu, South East Coast of India	Indian Ocean
Rexea solandri (Cuvier, 1832) Silver gemfish Gempylidae	C	bentho-pelagic	y	n/a	3	Zhu et al. 2019a	juv	3	y	100.00	n/a	n/a	various	South China Sea
Rhinoptera bonasus (Mitchill, 1815) Cownose ray Myliobatidae	C	bentho-pelagic	y	NT	1	Pegado et al. 2018	n/a	1	n	0.00	0.00	n/a	Amazon River estuary, northern	Western South-Atlantic

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													coast of Brazil	
Rhinosardinia bahiensis (Steindachner, 1879) Bahia sprat Clupeidae	n/a	pelagic	y	LC	139	Vendel et al. 2017	n/a	139	y	n/a	0.14	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Rhizoprionodon lalandii (Müller & Henle, 1839) Brazilian sharpnose shark Carcharhinidae	C	demersal	y	DD	6	Miranda and de Carvalho-Souza 2016	n/a	6	y	33.00	n/a	n/a	Salvador, Brazil	Western South-Atlantic
Roccus americanus (Gmelin, 1789) White perch Moronidae	n/a	demersal	y	LC	12	Carpenter et al. 1972	n/a	12	y	33.00	n/a	n/a	Southern New England	Western North-Atlantic
Salmo salar Linnaeus, 1758 Atlantic salmon Salmonidae	C	bentho-pelagic	y	LC	69	Liboiron et al. 2019	n/a	69	n	0.00	0.00	n/a	Newfoundland	Western North-Atlantic
Salmo trutta Linnaeus, 1758 Sea trout Salmonidae	C	pelagic-neritic	y	LC	62	Karlsson et al. 2017	ad	62	y	68.00	n/a	n/a	west coast Stenungsan, Sweden	Skagerrak
Sardina pilchardus (Walbaum, 1792) European pilchard Clupeidae	P	pelagic-neritic	y	LC	485	Anastasopoulou et al. 2018	ad, ad, ad, ad, ad	33, 48, 58, 30, 36	y, y, n, y, y	9.1, 50, 0, 36.6, 38.9	0.09, 1.12, 0, 0.9, 0.8,	n/a	North + South Adriatic Sea, NE Ionian Sea, Croatioan Sea	Mediterranean
						Collard et al. 2017b	sub-ad + ad	20	y	45.00	n/a	n/a	English Channel, Bay of Biscay, Northwestern Mediterranean	English Channel, Mediterranean, Eastern North-Atlantic
						Compa et al. 2018	ad	105	y	15.24	0.21	modelled Fulton's K	western Spanish Mediterranean, Gulf of	Mediterranean

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													Lions to the Alboran Sea	
						Neves et al. 2015	ad	12	n	0.00	0.00	n/a	off the coast of Portugal	Eastern North Atlantic
						Güven et al. 2017	n/a	7	y	57.00	3.75	n/a	Turkish territorial waters	Mediterranean
						Digka et al. 2018	juv	36	y	47.20	n/a	n/a	Northern Ionian Sea	Mediterranean
						Rios-Fuster et al. 2019	juv + ad	20	y	20.00	0.36	n/a	Iberian Peninsula coast, Balearic Island	Mediterranean
						Renzi et al. 2019	juv	80	y	96.00	4.63	n/a	central Adriatic Sea	Mediterranean
Sardinella albella (Valenciennes, 1847) White sardinella Clupeidae	P	reef-associated	y	LC	14	Azad et al. 2018	ad	14	y	86.00	2.30	n/a	Sathing Phra District	Gulf of Thailand
Sardinella gibbosa (Bleeker, 1849) Goldstripe sardinella Clupeidae	O	pelagic-neritic	y	LC	3	Azad et al. 2018	ad	3	y	33.00	0.30	n/a	Sathing Phra District	Gulf of Thailand
Sardinella jussieu (Lacepède, 1803) Mauritian sardinella Clupeidae	n/a	pelagic-neritic	y	DD	8	Azad et al. 2018	n/a	8	y	75.00	1.30	n/a	Sathing Phra District	Gulf of Thailand
Sardinops sagax (Jenyns, 1842) South American pilchard Clupeidae	P	pelagic-neritic	y	LC	7	Ory et al. 2018a	ad	7	n	0.00	0.00	n/a	Chile	South Eastern Pacific
Sargocentron spiniferum (Forsskål, 1775) Sabre squirrelfish Holocentridae	C	reef-associated	y	LC	5	Baalkhuyur et al. 2018	n/a	5	n	0.00	0.00	n/a	Saudi Arabian coast	Red Sea
Saurida tumbil (Bloch, 1795) Greater lizardfish Synodontidae	C	reef-associated	y	LC	4	Abbasi et al. 2018	juv	4	y	n/a	2.80	n/a	various	Persian Gulf

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Saurida undosquamis (Richardson, 1848) Brushtooth lizardfish Synodontidae	C	reef-associated	y	LC	99	Güven et al. 2017	n/a	99	y	55.00	2.20	n/a	Turkish territorial waters	Mediterranean
Scartichthys viridis (Valenciennes, 1836) n/a Blenniidae	H	demersal	n/a	LC	19	Mizraji et al. 2017	juv	19	y	n/a	14.00	condition K in g cm-3	Las Cruces, Chile	Eastern South Pacific
Scarus niger Forsskål, 1775 Dusky parrotfish Scaridae	H	reef-associated	y	LC	30	Markic et al. 2018	n/a	30	y	23.30	1.10	n/a	Samoa	South Pacific
Scarus oviceps Valenciennes, 1840 Dark capped parrotfish Scaridae	H	reef-associated	y	LC	45	Markic et al. 2018	n/a	45	y	11.10	2.80	n/a	Samoa	South Pacific
Scarus psittacus Forsskål, 1775 Common parrotfish Scaridae	H	reef-associated	y	LC	30	Markic et al. 2018	ad	30	y	16.70	1.00	n/a	Tahiti	South Pacific
Scatophagus argus (Linnaeus, 1766) Spotted scat Scatophagidae	O	reef-associated	y	LC	3	Zhu et al. 2019b	n/a	3	y	100.00	14.00	n/a	various	Maowei Sea
Schedophilus ovalis (Cuvier, 1833) Imperial blackfish Centrolophidae	C	bentho-pelagic	y	n/a	3	Anastasopoulou et al. 2013	n/a	3	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Schedophilus velaini (Sauvage, 1879) Violet warehou Centrolophidae	C	bathy-pelagic	n/a	LC	14	Markic et al. 2018	n/a	14	y	57.10	2.50	n/a	Rapa Nui	South Pacific
Sciades herzbergii (Bloch, 1794) Pemecou sea catfish Ariidae	C	demersal	y	LC	119	Vendel et al. 2017	juv	51, 6	y, n	n/a, 0	0.08, 0	n/a, n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
						Possatto et al. 2011	juv, sub-ad, ad	22, 20, 20	y, y, y	32, 10, 10	0.6, 0.1, 0.1	n/a, n/a, n/a	Goiana Estuary, Extractive Reserve	Western South-Atlantic

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													Acaú-Goiana	
Sciaena umbra Linnaeus, 1758 Brown meagre Sciaenidae	C	demersal	y	NT	1	Güven et al. 2017	n/a	1	y	100.00	3.00	n/a	Turkish territorial waters	Mediterranean
Sciaenops ocellatus (Linnaeus, 1766) Red drum Sciaenidae	C	demersal	y	LC	28	Phillips and Bonner 2015	n/a	28	y	n/a	n/a	n/a	Laguna Madre, southern coast of Texas	Western North-Atlantic
Scomber australasicus Cuvier, 1832 Blue mackerel Scombridae	P	pelagic-neritic	y	LC	4	Cannon et al. 2016	n/a	4	n	0.00	0.00	n/a	Australia	Southern Pacific, Southern Ocean, Australia
Scomber colias Gmelin, 1789 Atlantic chub mackerel Scombridae	n/a	pelagic-neritic	n/a	LC	120	Herrera et al. 2019	juv + ad	120	y	78.30	2.77	n/a	Gran Canaria + Lanzarote	Northern East-Atlantic
Scomber japonicus Houttuyn, 1782 Chub mackerel Scombridae	C	pelagic-neritic	y	LC	120	Su et al. 2019a	n/a	9	y	56.00	0.80	n/a	4 locations in Hangzhou Bay and Yangtze Estuary	East China Sea
						Anastasopoulou et al. 2018	ad	37	y	43.20	1.24	n/a	South Adriatic Sea	Mediterranean
						Neves et al. 2015	n/a	35	y	31.00	0.57	n/a	off the coast of Portugal	Eastern North Atlantic
						Rochman et al. 2015	n/a	1	n	0.00	0.00	n/a	Half Moon Bay, California, USA	Eastern North-Pacific
						Güven et al. 2017	n/a	7	y	71.00	9.40	n/a	Turkish territorial waters	Mediterranean
						Ory et al. 2018a	juv	31	y	3.30	0.03	n/a	Peru	South Eastern Pacific
Scomber scombrus Linnaeus, 1758	P	pelagic-neritic	y	LC	179	Nelms et al. 2018	juv	31	y	32.00	0.58	n/a	Celtic Sea/English Channel/We	Celtic Sea/English Channel/West

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Atlantic mackerel Scombridae													sternApproaches	ernApproaches
						Foekema et al. 2013	n/a	84	y	< 1	n/a	no impact on condition based on morphometrics	various	North Sea
						Neves et al. 2015	juv + ad	13	y	31.00	0.46	n/a	off the coast of Portugal	Eastern North Atlantic
						Rummel et al. 2016	ad, ad	38, 13	y, y	13.2, 30.8	n/a, n/a	n/a, n/a	various	North Sea, Baltic Sea
Scomberoides tala (Cuvier, 1832) Barred queenfish Carangidae	C	reef-associated	y	LC	3	Azad et al. 2018	n/a	3	y	67.00	0.70	n/a	Sathing Phra District	Gulf of Thailand
Scomberoides tol (Cuvier, 1832) Needlescaled queenfish Carangidae	PI	reef-associated	y	LC	3	Azad et al. 2018	juv	3	y	67.00	2.20	n/a	Sathing Phra District	Gulf of Thailand
Scomberomorus brasiliensis Collette, Russo & Zavala-Camin, 1978 Serra Spanish mackerel Scombridae	C	reef-associated	y	LC	1	Pegado et al. 2018	n/a	1	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Scomberomorus cavalla (Cuvier, 1829) King mackerel Scombridae	C	reef-associated	y	LC	9	Miranda and de Carvalho-Souza 2016	n/a	8	y	62.50	2.00	n/a	Salvador, Brazil	Western South-Atlantic
						Phillips and Bonner 2015	n/a	1	n	0.00	0.00	n/a	Laguna Madre, southern coast of Texas	Western North-Atlantic
Scomberomorus commerson (Lacepède, 1800) Narrow-barred Spanish mackerel Scombridae	C	pelagic-neritic	y	NT	4	Azad et al. 2018	juv	4	y	100.00	4.30	n/a	Sathing Phra District	Gulf of Thailand
Scomberomorus guttatus (Bloch & Schneider, 1801) Indo-Pacific king mackerel Scombridae	C	pelagic-neritic	y	DD	5	Azad et al. 2018	juv	5	y	60.00	0.60	n/a	Sathing Phra District	Gulf of Thailand

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Scophthalmus rhombus (Linnaeus, 1758) Brill Scophthalmidae	C	demersal	y	n/a	5	Steer et al. 2017	juv	5	n	0.00	0.00	n/a	various	western English Channel
Scorpaena elongata Cadenat, 1943 Slender rockfish Scorpaenidae	C	demersal	y	LC	1	Anastasopoulou et al. 2013	n/a	1	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Scorpaena jacksoniensis Steindachner, 1866 n/a Scorpaenidae	n/a	reef-associated	n/a	n/a	19	Cannon et al. 2016	n/a	19	n	0.00	0.00	n/a	Australia	Southern Pacific, Southern Ocean, Australia
Scyliorhinus canicula (Linnaeus, 1758) Lesser spotted dogfish Scyliorhinidae	C	demersal	y	LC	122	Anastasopoulou et al. 2013	n/a	1	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
						Neves et al. 2015	juv + ad, n/a	17, 3	y, y	12, 67	0.12, 0.67	n/a	off the coast of Portugal	Eastern North Atlantic
						Bellas et al. 2016	ad	72	y	15.30	1.20	n/a	Coast of Spain	East-Atlantic and Mediterranean
						Smith 2018	ad	20	y	5.00	1.00	n/a	n/a	North Sea
						McGoran et al. 2018	juv + ad, ad	7, 1	y, n	28, 0	1.5, 0	n/a, n/a		English Channel, Firth of Clyde
Sebastes caurinus Richardson, 1844 Copper rockfish Sebastidae	C	demersal	y	n/a	1	Rochman et al. 2015	n/a	1	n	0.00	0.00	n/a	Half Moon Bay, California, USA	Eastern North-Pacific
Sebastes flavidus (Ayres, 1862) Yellowtail rockfish Sebastidae	C	reef-associated	y	n/a	3	Rochman et al. 2015	n/a	3	y	33.30	0.30	n/a	Half Moon Bay, California, USA	Eastern North-Pacific
Sebastes miniatus (Jordan & Gilbert, 1880) Vermilion rockfish Sebastidae	n/a	reef-associated	y	n/a	3	Rochman et al. 2015	n/a	3	n	0.00	0.00	n/a	Half Moon Bay, California, USA	Eastern North-Pacific
Sebastes mystinus (Jordan & Gilbert, 1881)	C	reef-associated	y	n/a	10	Rochman et al. 2015	n/a	10	y	20.00	0.20	n/a	Half Moon Bay,	Eastern North-Pacific

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Blue rockfish Sebastidae													California, USA	
Sebastiscus marmoratus (Cuvier, 1829) False kelpfish Sebastidae	n/a	demersal	y	n/a	18	Jabeen et al. 2017	ad	18	y	100.00	4.20	n/a	Yangtze estuary, East China Sea and South China Sea	China Sea
Selar boops (Cuvier, 1833) Oxeye scad Carangidae	C	reef-associated	y	LC	7	Rochman et al. 2015	n/a	7	n	0.00	0.00	n/a	Makassar, Sulawesi, Indonesia	Makassar Strait
Selene setapinnis (Mitchill, 1815) Atlantic moonfish Carangidae	C	bentho-pelagic	y	LC	1	Pegado et al. 2018	n/a	1	n	0.00	0.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Selene vomer (Linnaeus, 1758) Lookdown Carangidae	C	demersal	y	LC	2	Pegado et al. 2018	n/a	2	y	50.00	2.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Seriola lalandi Valenciennes, 1833 Yellowtail amberjack Carangidae	C	bentho-pelagic	y	LC	34	Gassel et al. 2013	juv	19	y	10.50	1.00	n/a	over 1800 km from the coast of California.	North Pacific Subtropical Congervence Zone
						Markic et al. 2018	n/a	15	y	20.00	1.00	n/a	Auckland	South Pacific
Serranus cabrilla (Linnaeus, 1758) Comber Serranidae	C	demersal	y	LC	6	Güven et al. 2017	n/a	6	y	67.00	2.25	n/a	Turkish territorial waters	Mediterranean
Serrivomer beanii Gill & Ryder, 1883 Stout sawpalate Serrivomeridae	C	epibenthic-pelagic / bathy-pelagic	n	LC	14	Wieczorek et al. 2018	ad	14	y	92.86	2.36	n/a	Galway (Ireland) to St. John's (Newfoundland)	Eastern North-Atlantic
Setipinna taty (Valenciennes, 1848) Scaly hairfin anchovy Engraulidae	n/a	0-50	y	LC	20	Sun et al. 2019	n/a	20	y	30.00	1.20	n/a	Yellow Sea, 53 locations	East China Sea

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Siganus argenteus (Quoy & Gaimard, 1825) Streamlined spinefoot Siganidae	H	reef-associated	y	LC	2	Rochman et al. 2015	n/a	2	y	50.00	0.50	n/a	Makassar, Sulawesi, Indonesia;	Makassar Strait
Siganus canaliculatus (Park, 1797) White-spotted spinefoot Siganidae	H	reef-associated	y	LC	3	Rochman et al. 2015	n/a	3	y	33.30	0.30	n/a	Makassar, Sulawesi, Indonesia;	Makassar Strait
Siganus fuscescens (Houttuyn, 1782) Mottled spinefoot Siganidae	H	reef-associated	y	LC	2	Rochman et al. 2015	n/a	2	n	0.00	0.00	n/a	Makassar, Sulawesi, Indonesia;	Makassar Strait
Siganus luridus (Rüppell, 1829) Dusky spinefoot Siganidae	H	reef-associated	y	LC	n/a	Güven et al. 2017	n/a	15	y	87.00	3.62	n/a	Turkish territorial waters	Mediterranean
						van der Hal et al. 2018	n/a	n/a	y	n/a	n/a	n/a	Israeli coast	Mediterranean
Siganus punctatus (Schneider & Forster, 1801) Goldspotted spinefoot Siganidae	H	reef-associated	y	LC	29	Markic et al. 2018	juv	29	y	13.80	1.80	n/a	Samoa	South Pacific
Siganus rivulatus Forsskål & Niebuhr, 1775 Marbled spinefoot Siganidae	H	reef-associated	y	LC	n/a	van der Hal et al. 2018	n/a	n/a	y	n/a	n/a	n/a	Israeli coast	Mediterranean
Sillaginodes punctatus (Cuvier, 1829) King George whiting Sillaginidae	C	demersal	y	n/a	8	Cannon et al. 2016	juv	8	n	0.00	0.00	n/a	Australia	Southern Pacific, Southern Ocean, Australia
Sillago flindersi McKay, 1985 Eastern school whiting Sillaginidae	P	demersal	y	n/a	10	Cannon et al. 2016	ad	10	n	0.00	0.00	n/a	Australia	Southern Pacific, Southern Ocean, Australia
Sillago sihama (Forsskål, 1775) Silver sillago Sillaginidae	C	reef-associated	y	LC	39	Jabeen et al. 2017	ad	18	y	100.00	2.80	n/a	Yangtze estuary, East China Sea and South China Sea	China Sea
						Abbasi et al. 2018	juv + ad	17	y	n/a	1.50	n/a	various	Persian Gulf

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						Zhu et al. 2019b	n/a	4	y	100.00	1.70	n/a	various	Maowei Sea
Solea solea (Linnaeus, 1758) Common sole Soleidae	C	demersal	y	DD	664	Neves et al. 2015	n/a	1	n	0.00	0.00	n/a	off the coast of Portugal	Eastern North Atlantic
						Anastasopoulou et al. 2018	ad, ad, ad, ad	36, 50, 2, 20	n, y, n, y	0, 4, 0, 65	0, 0.04, 0, 1.9	n/a, n/a, n/a, n/a	North + South Adriatic Sea, NE Ionian Sea, Slovenian Sea	Mediterranean
						McGoran et al. 2018	juv + ad	18	y	17.00	1.00	n/a	Thames estuary	English Channel
						Steer et al. 2017	juv	4	n	0.00	0.00	n/a	various	western English Channel
						Pellini et al. 2018b	n/a	533	y	95.00	1.70	n/a	Adriatic Sea	Mediterranean
Sparus aurata Linnaeus, 1758 Gilthead seabream Sparidae	C	demersal	y	LC	130	Anastasopoulou et al. 2018	ad	20	y	100.00	7.30	n/a	Slovenian Sea	Mediterranean
						Güven et al. 2017	n/a	110	y	44.00	2.00	n/a	Turkish territorial waters	Mediterranean
Sparus macrocephalus (Bleeker, 1854) Blackhead seabream Sparidae	C	demersal	y	LC	3	Zhu et al. 2019b	n/a	3	y	100.00	2.00	n/a	various	Maowei Sea
Sphoeroides greeleyi Gilbert, 1900 Green puffer Tetraodontidae	C	reef-associated	n/a	LC	28	Vendel et al. 2017	juv + ad	28	n	0.00	0.00	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Sphoeroides testudineus (Linnaeus, 1758) Checkered puffer Tetraodontidae	C	reef-associated	n	LC	33	Vendel et al. 2017	juv + ad	33	n	0.00	0.00	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Sphyraena forsteri Cuvier, 1829 Bigeye barracuda Sphyraenidae	C	reef-associated	y	n/a	12	Markic et al. 2018	n/a	12	y	16.70	1.50	n/a	Samoa	South Pacific

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Sphyraena guachancho Cuvier, 1829 Guachanche barracuda Sphyraaenidae	C	0 - 100 m	y	LC	1	Miranda and de Carvalho-Souza 2016	n/a	1	n	0.00	0.00	n/a	Salvador, Brazil	Western South-Atlantic
Sphyrna tiburo (Linnaeus, 1758) Bonnethead Sphyrnidae	C	reef-associated	y	LC	2	Pegado et al. 2018	n/a	2	y	100.00	9.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Spratelloides gracilis (Temminck & Schlegel, 184 Silver-stripe round herring Clupeidae	n/a	0 - 40 m	y	LC	10	Rochman et al. 2015	n/a	10	y	40.00	1.10	n/a	Makassar, Sulawesi, Indonesia;	Makassar Strait
Sprattus sprattus (Linnaeus, 1758) European sprat Clupeidae	P	pelagic-neritic	y	LC	662	Hermsen et al. 2017	ad	141	y	0.71	2.00	n/a	Southern Bight	North Sea
						Steer et al. 2017	juv	6	n	0.00	0.00	n/a	various	western English Channel
						Beer et al. 2018	ad	515	y	18.80	1.15	n/a	Bornholm Basin	Baltic Sea
Squalus acanthias Linnaeus, 1758 Picked dogfish Squalidae	C	bentho-pelagic	y	LC	10	Anastasopoulou et al. 2013	n/a	10	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Squalus blainville (Risso, 1827) Longnose spurdog Squalidae	C	demersal	y	DD	75	Anastasopoulou et al. 2013	n/a	75	y	1.30	n/a	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Stellifer brasiliensis (Schultz, 1945) n/a Sciaenidae	n/a	demersal	n/a	n/a	330	Dantas et al. 2012	juv + sub-ad + ad	330	y	6.90	0.33	n/a	Goiana Estuary, Extractive Reserve Acaú-Goiana	Western South-Atlantic
Sternoptyx diaphana Hermann, 1781 Diaphanous hatchet fish Sternoptychidae	P	meso-pelagic / bathy-pelagic	n	LC	4	Davison and Asch 2011	ad	4	y	25.00	n/a	n/a	various	North Pacific Subtropical Gyre

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Sternoptyx pseudobscura Baird, 1971 Highlight hatchetfish Sternoptychidae	P	meso-pelagic / bathy-pelagic	n	LC	6	Davison and Asch 2011	ad	6	y	16.70	n/a	n/a	various	North Pacific Subtropical Gyre
Stomias boa boa (Risso, 1810) Boa dragonfish Stomiidae	C	bathy-pelagic	n/a	LC	14	Wieczorek et al. 2018	ad	9	y	66.67	1.33	n/a	Galway (Ireland) to St. John's (Newfoundland)	Eastern North-Atlantic
						Lusher et al. 2016	n/a	5	y	40.00	2.00	n/a	various	Eastern North-Atlantic
Strangomera bentincki (Norman, 1936) Araucanian herring Clupeidae	P	0 - 70 m	y	LC	10	Ory et al. 2018a	ad	10	n	0.00	0.00	n/a	Chile	South Eastern Pacific
Sudis hyalina Rafinesque, 1810 n/a Paralepididae	n/a	bathy-pelagic	n	DD	5	Anastasopoulou et al. 2013	n/a	5	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
Symbolophorus californiensis (Eigenmann & Eigenmann, 1889) Bigfin lanternfish Myctophidae	P	epipelagic / meso-pelagic	n/a	n/a	74	Boerger et al. 2010	n/a	74	y	n/a	7.20	n/a	various	North Pacific Central Gyre
Symphurus tessellatus (Quoy & Gaimard, 1824) n/a Cynoglossidae	n/a	demersal	n/a	LC	22	Vendel et al. 2017	n/a	22	n	0.00	0.00	n/a	Paraiba estuary, Northeastern Brazil	Western South-Atlantic
Synagrops japonicus (Döderlein, 1883) Blackmouth splitfin Acropomatidae	n/a	bentho-pelagic	y	LC	7	Zhu et al. 2019a	n/a	7	y	100.00	n/a	n/a	various	South China Sea
Taaningichthys bathyphilus (Tåning, 1928) Deepwater lanternfish Myctophidae	n/a	high-oceanic / bathy-pelagic	n/a	LC	3	Davison and Asch 2011	ad	3	n	0.00	0.00	n/a	various	North Pacific Subtropical Gyre
Taurulus bubalis (Euphrasen, 1786) Longspined bullhead Cottidae	C	demersal	n	LC	3	McGoran et al. 2018	n/a	3	y	67.00	2.00	n/a	Firth of Clyde	Firth of Clyde

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Terapon jarbua (Forsskål, 1775) Jarbua terapon Terapontidae	O	demersal	y	LC	18	Jabeen et al. 2017	ad	18	y	100.00	3.00	n/a	Yangtze estuary, East China Sea and South China Sea	China Sea
Terapon theraps Cuvier, 1829 Largescaled terapon Terapontidae	n/a	reef-associated	y	LC	8	Zhu et al. 2019b	n/a	3	y	100.00	5.70	n/a	various	Maowei Sea
						Azad et al. 2018	n/a	5	y	40.00	0.80	n/a	Sathing Phra District	Gulf of Thailand
Thalassoma rueppellii (Klunzinger, 1871) Klunzinger's wrasse Labridae	n/a	reef-associated	n/a	LC	12	Baalkhuyur et al. 2018	n/a	12	y	8.33	1.00	n/a	Saudi Arabian coast	Red Sea
Thamnaconus septentrionalis (Günther, 1874) n/a Monacanthidae	n/a	demersal	n/a	n/a	27	Jabeen et al. 2017	n/a	18	y	100.00	7.20	n/a	Yangtze estuary, East China Sea and South China Sea	China Sea
						Su et al. 2019a	n/a	9	y	33.00	0.70	n/a	4 locations in Hangzhou Bay and Yangtze Estuary	East China Sea
Thunnus alalunga (Bonnaterre, 1788) Albacore Scombridae	C	epipelagic / meso-pelagic / oceanic	y	NT	33	Rochman et al. 2015	n/a	2	n	0.00	0.00	n/a	Half Moon Bay, California, USA	Eastern North-Pacific
						Romeo et al. 2015	juv + ad	31	y	12.90	n/a	n/a	Eolian Islands, Strait of Messina	Mediterranean
Thunnus albacares (Bonnaterre, 1788) Yellowfin tuna Scombridae	C	oceanic / pelagic	y	NT	144	Markic et al. 2018	juv, juv, juv	33, 25, 10	y, y	15.2, 24, 70	1.4, 1.8, 3.1	n/a, n/a	South Pacific	Tahiti
						Chagnon et al. 2018	juv	50	n	0.00	0.00	n/a	Rapa Nui	South Pacific
						Choy and Drazen 2013	ad	26	n	0.00	0.00		n/a	Central North Pacific Ocean
Thunnus obesus (Lowe, 1839) Bigeye tuna Scombridae	C	0 - 1500 m	y	VU	35	Choy and Drazen 2013	ad	35	y	9.00	n/a	n/a	n/a	Central North Pacific Ocean

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Thunnus thynnus (Linnaeus, 1758) Atlantic bluefin tuna Scombridae	C	oceanic	y	EN	36	Romeo et al. 2015	ad	36	y	32.40	n/a	n/a	Eolian Islands, Strait of Messina	Mediterranean
Thyrsites atun (Euphrasen, 1791) Snoek Gempylidae	C	bentho-pelagic	y	n/a	30	Cannon et al. 2016	ad	2	n	0.00	0.00	n/a	Southern Pacific, Southern Ocean, Australia	Australia
						Markic et al. 2018	n/a	28	y	28.60	1.90	n/a	Rapa Nui	South Pacific
Torpedo torpedo (Linnaeus, 1758) Common torpedo Torpedinidae	C	demersal	n	DD	1	Neves et al. 2015	n/a	1	n	0.00	0.00	n/a	off the coast of Portugal	Eastern North Atlantic
Trachinotus ovatus (Linnaeus, 1758) Pompano Carangidae	C	50 - 200 m	y	LC	115	Battaglia et al. 2016	n/a	115	y	24.30	n/a	n/a	Strait of Messina	Mediterranean
Trachurus declivis (Jenyns, 1841) Greenback horse mackerel Carangidae	C	bentho-pelagic	y	LC	23	Cannon et al. 2016	ad	23	n	0.00	0.00	n/a	Australia	Southern Pacific, Southern Ocean, Australia
Trachurus mediterraneus (Steindachner, 1868) Mediterranean horse macker Carangidae	C	pelagic-oceanic	y	LC	187	Anastasopoulou et al. 2018	ad	2	n	0.00	0.00	n/a	NE Ionian Sea	Mediterranean
						Güven et al. 2017	n/a	98	y	67.00	1.58	n/a	Turkish territorial waters	Mediterranean
						Rios-Fuster et al. 2019	juv + ad	87	y	43.00	1.13	n/a	Iberian Peninsula coast, Balearic Island	Mediterranean
Trachurus novaezelandiae Richardson, 1843 Yellowtail horse mackerel Carangidae	n/a	22 - 500 m	y	LC	31	Markic et al. 2018	ad	31	y	3.20	1.00	n/a	Auckland	South Pacific
Trachurus picturatus (Bowdich, 1825)	C	bentho-pelagic	y	LC	38	Anastasopoulou et al. 2018	ad	9	n	0.00	0.00	n/a	NE Ionian Sea	Mediterranean

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Blue jack mackerel Carangidae						Neves et al. 2015	juv + ad	29	y	3.00	0.03	n/a	off the coast of Portugal	Eastern North Atlantic
Trachurus trachurus (Linnaeus, 1758) Atlantic horse mackerel Carangidae	C	pelagic-neritic	y	VU	255	Anastasopoulou et al. 2018	ad	50	y	24.00	0.52	n/a	South Adriatic Sea	Mediterranean
						Foekema et al. 2013	n/a	100	y	1.00	1.00	condition based on morphometrics	various	North Sea
						Lusher et al. 2013	n/a	56	y	28.60	1.50	n/a	10 km southwest of Plymouth	English Channel
						Murphy et al. 2017	n/a	5	n	0.00	0.00	n/a	Scottish offshore waters	North Sea, Eastern North Atlantic
						Neves et al. 2015	juv + ad	44	y	7.00	0.07	n/a	off the coast of Portugal	Eastern North Atlantic
Trichiurus lepturus Linnaeus, 1758 Largehead hairtail Trichiuridae	C	bentho-pelagic	y	LC	6	Neves et al. 2015	n/a	1	n	0.00	0.00	n/a	off the coast of Portugal	Eastern North Atlantic
						Pegado et al. 2018	n/a	5	y	20.00	2.00	n/a	Amazon River estuary, northern coast of Brazil	Western South-Atlantic
Tridentiger barbatus (Günther, 1861) Shokihaze goby Gobiidae	n/a	demersal	n/a	n/a	8	Su et al. 2019a	n/a	8	y	100.00	4.50	n/a	4 locations in Hangzhou Bay and Yangtze Estuary	East China Sea
Trigla lyra Linnaeus, 1758 Piper gurnard Triglidae	C	bathy-demersal	y	LC	31	Neves et al. 2015	n/a	31	y	19.00	0.26	n/a	off the coast of Portugal	Eastern North Atlantic
Triglops nybelini Jensen, 1944 Bigeye sculpin Cottidae	C	demersal	n	n/a	71	Morgana et al. 2018	n/a	71	y	34.00	1.00	Fulton's K Condition factor	Northeast Greenland	Northern Atlantic
Trisopterus esmarki (Nilsson, 1855) Norway pout Gadidae	C	bentho-pelagic	y	LC	20	Bour et al. 2018	n/a	20	y	25.00	n/a	n/a	Oslofjord	Skagerrak

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Trisopterus luscus (Linnaeus, 1758) Pouting Gadidae	C	bentho-pelagic	y	n/a	12	Neves et al. 2015	ad	1	n	0.00	0.00	n/a	off the coast of Portugal	Eastern North Atlantic
						Steer et al. 2017	juv	4	n	0.00	0.00	n/a	various	western English Channel
						McGoran et al. 2018	juv + ad	7	y	29.00	1.00	n/a	Thames estuary,	English Channel
Trisopterus minutus (Linnaeus, 1758) Poor cod Trisopteruidae	C	bentho-pelagic	y	n/a	93	McGoran et al. 2018	juv	1	n	0.00	0.00	n/a	Firth of Clyde	Firth of Clyde
						Lusher et al. 2013	n/a	50	y	40.00	2.00	n/a	10 km southwest of Plymouth	English Channel
						Steer et al. 2017	juv	42	y	2.30	n/a	n/a	various	western English Channel
Umbrina cirrosa (Linnaeus, 1758) Shi drum Sciaenidae	C	demersal	y	n/a	1	Güven et al. 2017	n/a	1	n	0.00	0.00	n/a	Turkish territorial waters	Mediterranean
Upeneus moluccensis (Bleeker, 1855) Goldband goatfish Mullidae	n/a	reef-associated	y	LC	18	Güven et al. 2017	n/a	18	y	44.00	1.75	n/a	Turkish territorial waters	Mediterranean
Upeneus pori Ben-Tuvia & Golani, 1989 Por's goatfish Mullidae	C	demersal	n/a	n/a	78	Güven et al. 2017	n/a	78	y	41.00	1.69	n/a	Turkish territorial waters	Mediterranean
Vinciguerria mabahiss Johnson & Feltes, 1984 n/a Phosichthyidae	n/a	bathy-pelagic	n/a	n/a	10	Baalkhuyur et al. 2018	n/a	10	n	0.00	0.00	n/a	Saudi Arabian coast	Red Sea
Xenodermichthys copei (Gill, 1884) Bluntsnout smooth-head Alepocephalidae	C	bathy-pelagic	n	LC	5	Lusher et al. 2016	n/a	5	y	60.00	2.00	n/a	various	Eastern North-Atlantic
Xiphias gladius Linnaeus, 1758 Swordfish Xiphiidae	C	oceanic	y	EN	88	Anastasopoulou et al. 2013	n/a	1	n	0.00	0.00	n/a	Cephalonia Island, Greece, Eastern Ionian Sea	Mediterranean
						Choy and Drazen 2013	ad	31	y	3.00	n/a	n/a	n/a	Central North Pacific Ocean

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						Romeo et al. 2015	juv + ad	56	y	12.50	n/a	n/a	Eolian Islands, Strait of Messina	Mediterranean
Zeugopterus punctatus (Bloch, 1787) Topknot Scophthalmidae	C	demersal	n	n/a	1	Steer et al. 2017	juv	1	n	0.00	0.00	n/a	various	western English Channel
Zeus faber Linnaeus, 1758 John dory Zeidae	C	bentho-pelagic	y	DD	43	Lusher et al. 2013	ad	42	y	47.60	1.00	n/a	10 km southwest of Plymouth	English Channel
						Neves et al. 2015	n/a	1	y	100.00	2.65	n/a	off the coast of Portugal	Eastern North Atlantic
Zoarces elongatus (Enchelyopus elongatus) Kner, 1868 n/a Zoarcidae	n/a	demersal	n/a	n/a	20	Sun et al. 2019	n/a	20	y	65.00	1.20	n/a	Yellow Sea, 53 locations	East China Sea
Zoarces viviparus (Linnaeus, 1758) Eelpout Zoarcidae	C	demersal	y		200	Wesch et al. 2016a	n/a	200	n	0.00	0.00	n/a	German Wadden Sea National Park, Bodden National Park	North Sea, Baltic Sea
22 different species (not mentioned individually)	n/a	n/a	n/a	n/a	> 500	Colton et al. 1974	juv	> 500	n	0.00	0.00	n/a	Cape Cod to Caribbean	Western North-Atlantic

Annex 2

Table 1 Data base on the 90 in-situ studies, conducted between 1972 and 2019, which were considered for the review on microplastic (MP) uptake by marine and estuarine fish taxa. References are listed in alphabetical order. Information on the location and geographic area of the respective studies are given, furthermore the average MP uptake across all individuals examined per study (in %), the total sample size of fish as well as the number of fish taxa investigated (additionally the calculated average number of individuals per taxa). The collection of environmental samples (e.g., plankton or benthos samples for reference), environmental parameters (e.g., salinity and temperature) is listed, furthermore analysis of the entire content of the gastrointestinal tract of the fish and a potential impact assessment. The last year of field study and the year of publication is given, along with the total number of citation (assessed on SCOPUS on 06th September 2019).
Citation history is not displayed, but available online via https://www.frontiersin.org/articles/10.3389/fmars.2021.672768/full#supplementary-material.
Abbreviations are as follows: y = yes; n = no; n/a = not available or not assessed; (y) = partially done or assessed. GIT = gastrointestinal tract. Environ = environmental.

Reference	**Location**	**Geographical Region**	**Average MP uptake in study (%)**	**Sample Size**	**Number of fish taxa**	**Average number of individuals per taxa**	**Environ. Samples**	**Environ. Parameters**	**GIT prey analysis**	**Impact**	**Field Study Year**	**Publication Year**	**Total no. of citations (SCOPUS)**
Abbasi et al. 2018	various	Persian Gulf	n/a	48	4	12.00	n	n	n	n/a	2015	2018	26
Alomar et al. 2017	Mallorca	Mediterranean	27.3	417	1	417.00	n	n	n	y	2015	2017	38
Alomar and Deudero 2017	Mallorca	Mediterranean	16.8	125	1	125.00	n	n	n	n/a	n/a	2017	34
Anastasopoulou et al. 2013	various	Mediterranean	1.9	1502	26	57.77	n	n	n	n/a	2010	2013	80
Anastasopoulou et al. 2018	various	Mediterranean	24	844	13	64.92	n	n	n	n/a	2015	2018	9
Arias et al. 2019	Bahia Blanca Estuary, Argentina	Western South-Atlantic	100	20	1	20.00	n	n	n	y	2018	2019	1
Azad et al. 2018	Sathing Phra District	Gulf of Thailand	66.7	165	24	6.88	n	n	n	n/a	2017	2018	0
Baalkhuyur et al. 2018	Saudi Arabian coast	Red Sea	14.6	178	26	6.85	n	n	n	n/a	2017	2018	10
Battaglia et al. 2016	Strait of Messina	Mediterranean	24.3	115	1	115.00	n	n	y	n/a	2012	2015	22
Beer et al. 2018	Bornholm Basin	Baltic Sea	20	814	2	407.00	y	n	n	n/a	2015	2017	22
Bellas et al. 2016	Coast of Spain	East-Atlantic and Mediterranean	17.5	212	3	70.67	n	n	n	n/a	2014	2016	87
Bernardini et al. 2018	offshore area in front of the coast of Sanremo, Imperia and Nice	Mediterranean	25.26	95	1	95.00	n	n	n	n/a	2015	2018	6
Bessa et al. 2018b	Portugal, Mondego estuary	Eastern North-Atlantic	38	120	3	40.00	n	n	n	n/a	2014	2018	31
Boerger et al. 2010	various	North Pacific Central Gyre	35	670	6	111.67	y	n	(y)	n/a	2008	2010	357
Bour et al. 2018	Oslofjord	Skagerrak	31.6	57	3	19.00	n	n	n	n/a	2017	2018	6

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Bråte et al. 2016	Oslo, Bergen, Sorfjorden, Karihavet, Lofoten, Varangerfjorden, Norway	Norwegian Sea, Skagerrak	3	302	1	302.00	n	n	n	n/a	n/a	2016	37
Cannon et al. 2016	Australia	Southern Ocean	0.3	342	21	16.29	n	n	n	n/a	2015	2016	34
Cardozo et al. 2018	Southern Brazil near Garopaba, Santa Catarina	Western South-Atlantic	49.17	122	1	122.00	n	n	y	y	2016	2018	5
Carpenter et al. 1972	Southern New England	Western North-Atlantic	5.5	270	14	19.29	y	n	n	n/a	1972	1972	307
Chagnon et al. 2018	Rapa Nui	South Pacific	8.6	93	2	46.50	n	n	y	n/a	2016	2018	6
Cheung et al. 2018	Sam Mun Tsai, eastern coast of Hong Kong	South China Sea	53.3	60	1	60.00	n	n	n	n/a	2017	2018	9
Choy and Drazen 2013	n/a	Central North Pacific Ocean	19	595	10	59.50	n	n	(y)	n/a	2012	2013	102
Collard et al. 2017b	English Channel, Bay of Biscay, Northwestern Mediterranean	English Channel, Mediterranean, Eastern North-Atlantic	45	60	3	20.00	n	n	n	n/a	2013	2017	22
Collicutt et al. 2019	Vancouver Island	Northern East-Pacific	59	74	1	74.00	y	n	n	n/a	2015	2018	4
Colton et al. 1974	Cape Cod to Caribbean	Western North-Atlantic	0	> 500	22	n/a	y	n	n	n/a	1972	1974	197
Compa et al. 2018	western Spanish Mediterranean, Gulf of Lions to the Alboran Sea	Mediterranean	14.8	210	2	105.00	n	n	n	y	2015	2018	24
Dantas et al. 2012	Goiana Estuary, Extractive Reserve Acaú-Goiana	Western South-Atlantic	7.9	569	2	284.50	n	n	n	n/a	2008	2011	90
Dantas et al. 2019	Southern Brazil, Santo Antonio dos Anjos + -Imarui Lagoons	Western South-Atlantic	14.1	92	1	92.00	n	n	y	y	2017	2019	0
Davison and Asch 2011	various	North Pacific Subtropical Gyre	9.2	141	27	5.22	n	n	n	n/a	2009	2011	170
Digka et al. 2018	Northern Ionian Sea	Mediterranean	41.3	80	3	26.67	n	n	n	n/a	2015	2018	23
Ferreira et al. 2016a	Goiana Estuary, Extractive	Western South-Atlantic	64.2	530	1	530.00	(y)	y	y	n/a	2008	2016	n/a

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	Reserve Acaú-Goiana												
Ferreira et al. 2018	Goiana Estuary, Extractive Reserve Acaú-Goiana	Western South-Atlantic	51	552	1	552.00	n	y	n	n/a	2008	2016	13
Foekema et al. 2013	various	North Sea	2.6	1203	7	171.86	n	n	n	y	2011	2013	259
Gassel et al. 2013	over 1800 km from the coast of California	North Pacific Subtropical Congervence Zone	10.5	19	1	19.00	y	n	y	n/a	2009	2013	41
Giani et al. 2019	various around Italy	Mediterranean	23.3	229	2	114.50	n	n	n	n/a	n/a	2019	4
Güven et al. 2017	Turkish territorial waters	Mediterranean	57.7	1337	28	47.75	y	n	n	n/a	2015	2017	83
Halstead et al. 2018	Sydney Harbour, Australia	Southern West-Pacific	43	93	3	31.00	n	n	y	n/a	2017	2018	20
Hermsen et al. 2017	Southern Bight	North Sea	0.25	400	4	100.00	n	n	n	n/a	2013	2017	40
Herrera et al. 2019	Gran Canaria + Lanzarote	Northern East-Atlantic	78.3	120	1	120.00	n	n	n	n/a	n/a	2019	5
Jabeen et al. 2017	Yangtze estuary, East China Sea and South China Sea	China Sea	100	378	21	18.00	n	n	n	n/a	2015	2016	97
Jensen et al. 2019	Great Barrier Reef World Heritage Area	Western South-Pacific	95	60	1	60.00	y	n	n	n/a	2016	2019	0
Karlsson et al. 2017	west coast Stenungsan, Sweden	Skagerrak	68	62	1	62.00	n	n	n	n/a	2014	2017	69
Kartar et al. 1976	various	Eastern North-Atlantic	10.5	1623	4	405.75	y	n	n	n/a	1975	1976	34
Kazour et al. 2018	Canche and Seine Estuary, French coast	English Channel	70.1	86	1	86.00	n	n	n	y	2017	2018	0
Kühn et al. 2018	ice-covered water in Eurasian Basin; open water near Svalbard	Central Arctic Ocean	2.8	72	1	72.00	n	n	n	n/a	2015	2018	10
Kumar et al. 2018	Tamil Nadu, South East Coast of India	Indian Ocean	30	40	2	20.00	n	n	n	n/a	n/a	2018	3
Liboiron et al. 2016	Eastern Coast of Newfoundland	Western North-Atlantic	2.4	205	1	205.00	n	n	n	n/a	2015	2016	18

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Liboiron et al. 2018	South Coast of Newfoundland, Canada (Burgeo Bank, St. Pierre Bank, Southern Grand Bank)	Western North-Atlantic	0	134	1	134.00	n	n	y	n/a	2016	2018	2
Liboiron et al. 2019	Newfoundland	Western North-Atlantic	1.2	1429	3	476.33	n	n	(y)	n/a	2016	2019	0
Lusher et al. 2013	10 km southwest of Plymouth	English Channel	36.5	504	10	50.40	n	n	n	n/a	2011	2012	464
Lusher et al. 2016	various	Eastern North-Atlantic	11	761	10	76.10	y	n	n	n/a	2014	2015	51
Markic et al. 2018	various	Western South-Pacific	24.3	932	34	27.41	n	n	(y)	n/a	2016	2018	3
McGoran et al. 2017	Thames estuary	English Channel	68.4	76	2	38.00	n	n	y	n/a	2014	2016	36
McGoran et al. 2018	Thames estuary, Firth of Clyde	English Channel, Firth of Clyde	33.2	760	20	38.00	n	n	n	n/a	2016	2018	1
Miranda and de Carvalho-Souza 2016	Salvador, Brazil	Western South-Atlantic	21.9	32	11	2.91	n	n	(y)	n/a	2011	2016	46
Mizraji et al. 2017	Las Cruces, Chile	Eastern South-Pacific	100	62	5	12.40	n	n	n	y	2015	2017	36
Morgana et al. 2018	Northeast Greenland	Western North-Atlantic	25	156	2	78.00	y	n	n	y	2015	2018	8
Murphy et al. 2017	various	Eastern North-Atlantic, North Sea	29.7	212	12	17.67	n	n	n	n/a	2014	2017	29
Nadal et al. 2016	Mallorca, Eivissa	Mediterranean	57.8	337	1	337.00	n	n	y	n/a	2014	2016	71
Naidoo et al. 2016	Bayhead Mangroves of Durban Harbour; South Africa, KwaZulu-Natal	Indian Ocean	72.8	70	1	70.00	n	n	(y)	n/a	2014	2016	21
Nelms et al. 2018	Celtic Sea/English Channel/Western Approaches	English Channel	32	31	1	31.00	n	n	n	n/a	n/a	2018	63
Neves et al. 2015	off the coast of Portugal	Eastern North-Atlantic	19.8	263	26	10.12	n	n	n	n/a	2013	2015	145
Ory et al. 2017	Rapa Nui, Easter Island, South Pacific subtropical gyre	South Pacific Subtropical Gyre	80	20	1	20.00	y	n	y	n/a	2015	2017	55
Ory et al. 2018a	various	Eastern South-Pacific	2.1	292	7	41.71	n	n	n	n/a	2016	2017	19

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Pegado et al. 2018	Amazon River estuary, northern coast of Brazil	Western South-Atlantic	13.7	189	46	4.11	n	n	n	n/a	2016	2018	10
Pellini et al. 2018b	Adriatic Sea	Mediterranean	95	533	1	533.00	n	n	n	n/a	2015	2017	35
Peters et al. 2017	Texas (TX) Gulf Coast, Galveston Bay to Freeport	Western North-Atlantic	42.4	1381	6	230.17	n	n	y	n/a	2015	2017	17
Phillips and Bonner 2015	Laguna Madre, southern coast of Texas	Western North-Atlantic	10.4	116	8	14.50	n	n	n	n/a	2013	2015	57
Possatto et al. 2011	Goiana Estuary, Extractive Reserve Acaú-Goiana	Western South-Atlantic	23.1	182	3	60.67	n	n	n	n/a	2008	2011	173
Ramos et al. 2012	Goiana Estuary, Extractive Reserve Acaú-Goiana	Western South-Atlantic	13.4	425	3	141.67	n	n	n	y	2008	2012	79
Renzi et al. 2019	central Adriatic Sea	Mediterranean	93.7	160	2	80.00	n	n	n	n/a	2014	2019	1
Rios-Fuster et al. 2019	Iberian Peninsula coast, Balearic Island	Mediterranean	28.4	197	4	49.25	n	n	(y)	n/a	2015	2019	1
Rochman et al. 2015	Makassar, Sulawesi, Indonesia; Half Moon Bay, California, USA	Eastern North-Pacific	26.4	140	23	6.09	n	n	n	n/a	2014	2015	222
Romeo et al. 2015	Eolian Islands, Strait of Messina	Mediterranean	18.2	121	3	40.33	n	n	n	n/a	2013	2015	142
Romeo et al. 2016	Sicilian coast	Mediterranean	2.7	522	4	130.50	n	n	n	n/a	2014	2016	15
Rummel et al. 2016	various	Baltic Sea + North Sea	5.5	290	5	58.00	n	n	n	y	2013	2015	125
Savoca et al. 2019	Thyrrhenian Sea	Mediterranean	9.1	39	2	19.50	y	n	n	n/a	2017	2019	15
Silva et al. 2018b	Goiana Estuary, upper, middle and lower	Western South-Atlantic	n/a	169	2	84.50	n	y	y	n/a	2013	2018	6
Skóra et al. 2012	Gulf of Gdansk	Baltic Sea	n/a	205	1	205.00	n	n	y	n/a	2002	2012	6
Smith 2018	n/a	North Sea	5	20	1	20.00	n	n	y	n/a	2018	2018	7
Steer et al. 2017	various	English Channel	2.9	347	23	15.09	y	n	n	n/a	2016	2017	64
Su et al. 2019a	4 locations in Hangzhou Bay and Yangtze Estuary	East China Sea	57.3	185	13	14.23	n	n	n	n/a	2017	2018	8

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Sun et al. 2019	Yellow Sea, 53 locations	East China Sea	34	1320	19	69.47	n	n	n	n/a	2016	2019	2
Tanaka and Takada 2016	Tokyo Bay	Western North-Pacific	77	64	1	64.00	n	n	n	n/a	2015	2016	78
van der Hal et al. 2018	Israeli coast	Mediterranean	92	88	2	44.00	n	n	n	n/a	2016	2018	0
Vendel et al. 2017	Paraiba estuary, Mamangua estuary, Northeastern Brazil	Western South-Atlantic	9	2233	69	32.36	n	y	(y)	n/a	2014	2017	49
Wesch et al. 2016a	German Wadden Sea National Park, Bodden National Park	Baltic Sea + North Sea	0	250	1	250.00	n	n	n	n/a	2014	2016	30
Wieczorek et al. 2018	Galway Ireland to St. John's Newfoundland	Eastern North-Atlantic	73.4	233	7	33.29	y	n	n	n/a	2015	2018	12
Zhu et al. 2019a	various	South China Sea	100	35	13	2.69	n	n	n	n/a	2017	2019	3
Zhu et al. 2019b	various	Maowei Sea	100	66	12	5.50	(y)	n	n	n/a	2017	2018	7

9 Acknowledgements

Towards the end of this dissertation (and my PhD journey), I would like to take the opportunity to express my gratitude to the group of encouraging, inspiring, and caring individuals whose support made me accomplish more than I would have been able to do alone, both in professional and private life. I promise to try my best in giving everyone the well-deserved appreciation, thus be prepared to read through some more pages; my apologies in advance to anyone whom I may have missed in this section – rest assured I acknowledge your support nonetheless!

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Werner, I thank you dearly for your leap of faith when affiliating me to your exquisite, small team without truly knowing me as a student (or person), for supporting me during this project and helping me advance my scientific skills, for sharing sound advice (and ironic comments) while leaving me enough freedom to work independently and shape my PhD project. I highly appreciate the chances (and challenges) you have been giving me to freely conduct and communicate my research, and to take major responsibilities beyond the scope of my own project. Special thanks also for giving me the once (or twice…) in a lifetime experience of cruising the Atlantic Ocean – I will never forget the marvellous sights of the Canary Current and the Benguela Upwelling System!

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I consider myself lucky that my professional and private network are quite often not clearly separable from each other, and many colleagues have become friends over the years, yet I would like to take the opportunity to explicitly express my sincere gratitude to the wonderful people beyond my pleasant working environment: my friends and family.

To all my friends who, over the last couple of years, endured the overly occupied version of me and accepted my absence, both physically and mentally, offered words of encouragement, constant support, and profound friendship: thank you dearly! Notably, my warmest thanks go out to Eva, for being the best friend one could hope for; to Sebi for being in the same boat and making that rough cruise a tolerable one; to Irina for cheerfully running with me towards that bright light at the end of each tunnel; to Fanny for being impartial and open-minded to let a noisy biologist into your world; to Alena, my last bastion of self-irony among the grand-aided paragons.

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من مشتاقانه منتظر آینده با هم و سفرهای مشترکمان هستم

Last but not least: *Mama und Papa, ich werde Euch niemals genug danken können für Euer liebevolles Vertrauen in mich und Euren unbegrenzten Glauben an meine Lebensträume. Dank Eurer bedingungslosen Unterstützung konnte ich immer genau das ausprobieren, was ich wollte und dem folgen, was mich von Kindesbeinen an faszinierte - ob nah oder fern, Ihr wart und seid stets an meiner Seite! Herzlichen Dank!*

10 Versicherung an Eides Statt

Ich, Carolin Müller,

versichere an Eides Statt durch meine Unterschrift, dass ich die vorstehende Arbeit mit dem Titel *Impact of Microplastic on Early Life-History Stages of Seabream in the Atlantic Ocean* selbständig und ohne fremde Hilfe angefertigt und alle Stellen, die ich wörtlich dem Sinne nach aus Veröffentlichungen entnommen habe, als solche kenntlich gemacht habe, mich auch keiner anderen als der angegebenen Literatur oder sonstiger Hilfsmittel bedient habe.

Ich versichere an Eides Statt, dass die zu Prüfungszwecken beigelegte elektronische Version der Dissertation identisch ist mit der abgegebenen gedruckten Version.

Ich versichere an Eides Statt, dass ich die vorgenannten Angaben nach bestem Wissen und Gewissen gemacht habe und dass die Angaben der Wahrheit entsprechen und ich nichts verschwiegen habe.

Die Strafbarkeit einer falschen eidesstattlichen Versicherung ist mir bekannt, namentlich die Strafandrohung gemäß § 156 StGB bis zu drei Jahren Freiheitsstrafe oder Geldstrafe bei vorsätzlicher Begehung der Tat bzw. gemäß § 161 Abs. 1 StGB bis zu einem Jahr Freiheitsstrafe oder Geldstrafe bei fahrlässiger Begehung.

Bad Zwischenahn, 17.11.2021

“The sea, once it casts its spell, holds one in its net of wonder forever.”

Jacques-Yves Cousteau