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Trophic transfer of polyester microfibres across a multi-level marine food web

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ABSTRACT

Microplastics are multifarious contaminants and their transfer by marine organisms can impact health. Yet, despite considerable research, microplastic quantification across multiple trophic levels is incomplete. Here, ingestion, retention, depuration and transfer of environmentally relevant polyester (PEST) microplastics, with plasticising bis(2-ethylhexyl) phthalate (DEHP) as a co-contaminant, is reported for three reef species. Exposed copepods (*Parvocalanus crassirostris*), mysid shrimps (Mysida) and moon wrasse fish (*Thalassoma lunare*) ingested ($<21.33 \pm 15.94$ PEST individual^{-1}), retained (<48 h), and depurated ($<5.77 \pm 1.27$ PEST h^{-1}) PEST. Trophic transfer was observed as a 14.6-fold (copepods to shrimps) and 4.3-fold (shrimps to fish) increase of PEST. All organisms demonstrated PEST bioconcentration, PEST biomagnification only occurred in shrimp. DEHP positively influenced PEST intake by copepods, yet had no impact on PEST transfer. Demonstrated PEST transfer across this food chain, coupled with species-specific bioconcentration and biomagnification dynamics, enhances understanding of microplastics fate and informs ecological impact assessments.

1. Introduction

Estimates of global emissions of plastic into marine ecosystems are as high as 25 million metric tonnes (MMt) per year (Lau et al., 2020), with a predicted increase in annual emissions of up to 53 MMt by 2030 (Borrelle et al., 2020). Specifically, global production of synthetic fibres (e.g., polyester; PEST) has reached 113 MMt in 2021 (Textile Exchange, 2022), accounting for 14.5 % of total plastic production by mass (Geyer et al., 2017). Many of these fibres enter the marine environment via waste discharge, land run-off and improper waste disposal (Kershaw and Rochman, 2015). As a consequence, fibres are the most prevalent anthropogenic debris reported in pollution surveys (Gago et al., 2018; Suaria et al., 2020). Following their uncontrolled introduction into marine environments, mechanical abrasion and exposure to UV light can damage the integrity of plastic polymers (Zhu et al., 2019) and over time these processes promote fragmentation (Hahladakis et al., 2018) into microplastics (MPs; plastics between 1 μm and 5 mm (Arthur et al., 2008; Hartmann et al., 2019)). Due to their persistence in the marine environment and intake by marine organisms across all trophic levels, MP fibres are of major global concern (Lusher, 2015; Miller et al., 2020; Osman et al., 2023).

Virgin unadulterated plastic polymers are considered chemically inert; however, are often not fit for purpose in primary form. Hence, polymers are often enhanced with chemical additives (e.g., plasticisers, flame retardants, UV stabilisers, etc.) to impart specific properties such as flexibility and malleability, or to prolong the life of plastics (Hahladakis et al., 2018). A major group of chemical additives commonly used to increase the flexibility and elasticity of rigid plastics is the phthalic acid esters (or phthalates; commonly referred to as plasticisers) (Net et al., 2015). Phthalates are readily adsorbed to plastics, filling the free volume between polymer chains (i.e., noncovalently binding to plastics) (Rowdhwal and Chen, 2018). As such, the phthalates can migrate into surrounding aqueous matrices when immersed in the environment (Gulizia et al., 2023; Liang et al., 2008). Bis(2-ethylhexyl) phthalate (DEHP) is one of the most frequently used plasticisers worldwide (Rowdhwal and Chen, 2018). It is commonly used in the manufacturing of clothing and textile fibres (Aldegunde-Louzao et al., 2023), yet there are significant health concerns. DEHP acts as a

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carcinogen and, along with its degradation products, an endocrine disruptor (Kamrin, 2009). It is readily found contaminating the marine environment with concentrations of up to 64.3 μg L^{-1} (median of 1 μg L^{-1}) in seawater globally (Bergé et al., 2013). The bulk of this pollution arises from urban sewage effluents (up to 69 μg L^{-1}), emissions from metal, mineral and chemical production (up to 4.4 mg L^{-1}), and the 'end-of-life' phase of some plastics (e.g., abraded plastics, or plastics left in the environment) (Bergé et al., 2013; European Commission, 2011). MPs have been shown to act as a major vector for chemical contamination, and, as DEHP is lipophilic, MPs have the potential to promote its concentration, distribution and persistence within the marine environment (Amelia et al., 2021; Ye et al., 2020). This, along with its documented detrimental health impacts to animals and humans (Net et al., 2015; Rowdhwal and Chen, 2018), has elevated DEHP to a priority pollutant under the European Commission's Water Framework Directive (European Commission, 2011) and United States Environmental Protection Agency (USEPA, 2019).

Examining trophic models has proven critical to fully understand the downstream ecological fate of various contaminants in marine ecosystems (Briand et al., 2018). In coral reef ecosystems, zooplankton, small benthic invertebrates, and reef fish play crucial roles in energy transfer processes, i.e., as an important food source, and as intermediate prey species and predators, respectively (Holmes et al., 2012; Kramer et al., 2015; Oliveira et al., 2023; Verslycke et al., 2007). These organisms predominately feed within the water column or forage through benthic sediments for prey, both environments in which MPs are abundant (Botterell et al., 2019). In situ ingestion of MPs has been confirmed (Miller et al., 2023), and ecotoxicology studies investigating the effects of MPs on zooplankton (i.e., copepods) and benthic invertebrates (i.e., mysid shrimp) under laboratory conditions (Lee et al., 2021; Setala et al., 2014) indicate these organisms are likely to be negatively impacted despite the caveats of exposure variations (e.g., concentrations, sizes, shapes, colours; see Supplementary Literature Review for more details). The inclusion of higher order mesopredators (i.e., reef fish) in such studies is less frequent (Critchell and Hoogenboom, 2018; Santana et al., 2021), leaving a gap in our understanding of the mechanisms of MPs transfer to and their ecological fate in these higher trophic level organisms.

While inferred within the literature (Farrell and Nelson, 2013; Nelms et al., 2018), trophic transfer of MPs and associated chemical additives in marine ecosystems is seldom investigated in detail (Miller et al., 2020). Although controlled laboratory exposure studies have demonstrated MP ingestion and short-term retention can facilitate transfer across two trophic levels (Supplementary Literature Review and Supplementary Tables 1 and 2), few quantify the MPs ingested (Elizalde-Velázquez et al., 2020; Hasegawa and Nakaoka, 2021; Xu et al., 2022). Studies utilising MPs intentionally pre-adsorbed with chemical pollutants are further limited (Athey et al., 2020). Notwithstanding, the chemical signal of MPs can play an important role in MP ingestion and retention, in some instances promoting MP ingestion (Savoca et al., 2017) or acting as an attractant (Greenshields et al., 2021) for marine organisms. For example, larval fish preferentially ingest MPs contaminated with dichlorodiphenyltrichloroethane (DDT) compared to MPs alone (Athey et al., 2020). While plastic additive contamination within the tissues of marine organisms has been correlated to MP ingestion (Guo et al., 2020), evidence is lacking as to whether plastic additives, specifically phthalates, impact the quantity of MPs ingested, retained, depurated or transferred up trophic levels (Miller et al., 2020).

Bioconcentration (i.e., the increase of contaminants in an organism relative to the concentration in its surrounding environment (USEPA, 1997)) and biomagnification (i.e., the increase in contaminant levels in an organism relative to the concentration present in its prey (USEPA, 2008)) are fundamental concepts utilised in ecological risk assessments to evaluate the movement of pollutants within food webs (Beek et al., 2000; Chormare and Kumar, 2022; Feijtel et al., 1997). Initially developed to assess dissolved chemical contamination (Alexander, 1999), these concepts are only now being adapted to evaluate particulate MP contamination (Akhbarizadeh et al., 2019; Covernton et al., 2022; Rochman et al., 2019) – a shift still subject to conjecture within the scientific literature. Establishing the relevance of these parameters for MP risk assessments, including in complex ecosystems such as coral reefs, requires targeted investigation of realistic, species-interactive food webs.

This study offers a novel exploration of the ecological fate of the pervasive MP, PEST, including in the presence of its oft-associated phthalate additive DEHP, within a simple linear coral reef food chain. Specifically, this study quantified ingestion, retention and depuration rates and trophic transfer efficiency of PEST MPs and assessed the impact of DEHP (as the co-contaminant) on these endpoints. Three marine taxa, namely copepods (*Parvocalanus crassirostris*; trophic level 1), mysid shrimp (species of the Order: Mysida; trophic level 2) and moon wrasse fish (*Thalassoma lunare*; trophic level 3), were exposed individually to (a) PEST alone, (b) DEHP alone, (c) PEST intentionally pre-adsorbed with DEHP (PEST:DEHP), (d) PEST and DEHP added in tandem (PEST+DEHP), and (e) controls (i.e., no PEST or DEHP) to quantify ingestion, retention and depuration of PEST MPs. Mysid shrimp and fish were also exposed to prey burdened with each PEST treatment via a linear food chain arrangement to assess the potential for trophic transfer. Analyses made between species used both raw MP quantifications (i.e., per individual), as well as standardised MP ingestion, retention, depuration and transfer quantities (i.e., by weight) to allow for intercomparison of data. In addition, bioconcentration and biomagnification factors were calculated to better understand the accumulation dynamics of PEST. Critical quantitative insights and transferability of these findings to broader research contexts is discussed and recommendations are offered to guide future research efforts.

2. Methods

2.1. Permits and ethics

All experimental aquaria work was conducted at the Australian Institute of Marine Science's (AIMS) National Sea Simulator (SeaSim), located near Gurumbilbarra and Thul Garrie Waja, on Wulgurukaba and Bindal Country (Townsville, Queensland) in accordance with relevant institutional and national guidelines (Great Barrier Reef Marine Park Authority permit G12/35236.1 and James Cook University (JCU) Animal Ethics Committee Approval Number A2722). All sample sizes employed within this study were chosen to reduce the number of organisms required to be sacrificed, while still ensuring the experiments were statistically sound.

2.2. Animal culturing, collection, and husbandry

Water quality parameters in experimental tanks were set to match local mid-shelf coral reef conditions (e.g., Davies Reef, Great Barrier Reef [GBR]), including temperature (25.5 °C), pH (8.2), salinity (35.5 ppt), and dissolved oxygen (DO; 7.8 mg L^{-1}). These parameters were continuously monitored using the Supervisory Control and Data Acquisition (SCADA, Siemens WinCC) system within SeaSim and checked twice weekly using a portable water quality meter (HACH HQ11D; Supplementary Note 1). Light conditions were set to 12 h:12 h light:dark and achieved using SOL blue LED lights, which were incrementally increased to a final level of 80 % intensity of 100 Photosynthetic Active Radiation over 4 days (20 % per day) of acclimation.

Copepods (*P. crassirostris*) were cultured and reared under laboratory conditions at SeaSim for the purposes of these experiments (Supplementary Table 3). In brief, adult *P. crassirostris* were obtained from JCU's Marine and Aquaculture Research Facilities Unit (MARFU) and reared in two 200 L conical tanks at SeaSim. Copepods were fed a 200 mL algal mixture of *Tisochrysis lutea* (T-Iso) and *Chaetoceros muelleri* (1:1 ratio) daily and water changes were conducted twice weekly. In culture, the

sex ratio of *P. crassirostris* is highly skewed towards females (>85 %), the males do not feed after maturation (Alajmi and Zeng, 2015). To ensure the experiments simulated environmental conditions, no effort was made to remove the males.

Mysid shrimps (Order: Mysida) were cultured and reared using a stock population originating from local offshore mid-shelf reefs (e.g., Davies Reef) and maintained in large mesocosm coral reef tanks (1200 L) within SeaSim (Supplementary Table 3). For each experiment, mysid shrimps were transferred and maintained in a 100 L conical tank as a stock population. Mysid shrimp were fed a minimum of 25,000 copepods (*P. crassirostris*) daily to ensure cannibalism did not occur (Heindler et al., 2017); feed was either frozen (Experiment 1; Ocean Nutrition Frozen) or live (Experiment 2).

Moon wrasse fish (*T. lunare*, $n = 130$) were collected by Cairns Marine using hand and barrier nets at various mid-shelf reefs within the GBR World Heritage Area (GBRWHA) (Supplementary Fig. 1) and transported to SeaSim where they underwent acclimation in glass 50 L experimental tanks. Fish were kept individually in tanks, each equipped with an air supply (i.e., titanium rod) and a silica-glazed ceramic structure (13 × 11 cm; hollow) to provide refuge. Fish were fed a minimum of 1.2 mg per g fish weight of mysid shrimp daily, either frozen (Experiment 1; Ocean Nutrition Frozen) or live (Experiment 2). All fish were observed feeding within 24 h of arrival to the SeaSim, an indication of healthy and normal behaviour.

2.3. Experimental design

Fluorescent yellow PEST fibres were chosen as the model MP, as PEST fibres are the most common MPs found in the GBRWHA (Jensen et al., 2019; Miller et al., 2022; Santana, 2022). The fluorescent colour was chosen to allow easy discovery within the individual GITs of copepods and mysid shrimps without the need for chemical digestion. DEHP was chosen as the plasticising chemical additive, being defined as a priority waterway pollutant by the European Commission (European Commission, 2011).

First, copepods, mysid shrimp and fish were individually exposed to PEST and DEHP plasticiser treatments to determine ingestion and retention of PEST in the presence and absence of DEHP (Experiment 1). Second, the transfer of PEST from copepods to mysid shrimp to fish was assessed (Experiment 2). Treatments for both experiments included dosing experimental tanks with (a) PEST alone, (b) DEHP alone, (c) PEST pre-adsorbed with DEHP (PEST:DEHP), (d) PEST and DEHP added in tandem (PEST+DEHP), or (e) 1 μm filtered seawater (FSW) controls absent of PEST and DEHP. Following exposures, organisms were analysed for PEST contamination only. Experimental tank location in the experimental room, and treatment allocation were randomised using a customised random generator script in R (Version 4.04 with Rstudio, version 1.4.1106). For information regarding contamination control and exposure validation, see Supplementary Note 2 and Supplementary Table 4.

2.4. Treatment preparation

PEST microfibres were prepared using a fluorescent yellow thread purchased from a local craft store (Spotlight; Birch Polyester Thread Fluoro Yellow 1000 m; Supplementary Fig. 2). MP polymer type, shape, colour, and concentration were consistent for all exposures, yet differed in size. To expose organisms to MPs comparable to that of their natural prey (Experiment 1), PEST microfibres were prepared in sizes ranging from 10 ± 2 μm for copepods, 350 ± 28 μm for mysid shrimp, and 750 ± 14 μm for fish. For Experiment 2, only copepods were exposed to PEST, after which they were fed to mysid shrimp; therefore, only the smallest size range (i.e., 10 μm) was utilised. Larger microfibres (i.e., >300 μm) were cut from thread using a scalpel blade under microscope (Leica M80, 0.75–6.0× magnification) and length confirmed using a micrometre ruler. Smaller microfibres (i.e., 10 μm) were produced using a cryostat (Supplementary Fig. 3). A 10 μm PEST stock solution (1250 MPs mL^{-1}) was prepared (Supplementary Note 3) to facilitate dosing of copepods, while the 350 μm and 750 μm PEST microfibres were counted and dosed directly.

Concentrations of PEST and DEHP were kept as environmentally realistic as possible, while still ensuring detectable levels within a laboratory setting (Supplementary Note 4). PEST concentrations used in treatments were 10 microfibres per individual organism, one order of magnitude larger than in situ levels reported within copepods, shrimps, and fish (0.02 ± 0.01, 0.43 ± 0.87, and 1.75 ± 1.54 MPs individual^{-1}, respectively (Miller et al., 2023)). For Experiment 1, this equates to 5000 PEST L^{-1} (approx. 0.04 mg L^{-1}) for copepods, 120 PEST L^{-1} (approx. 0.003 mg L^{-1}) for mysid shrimp and 0.2 PEST L^{-1} (approx. 1.2×10^{-4} mg L^{-1}) for fish; for Experiment 2, this resulted in 26,000 PEST L^{-1} (approx. 0.21 mg L^{-1}). For both experiments, DEHP was dosed at a concentration of 1 mg L^{-1}, one order of magnitude higher than median levels found in marine environments globally but still within the reported levels (Bergé et al., 2013).

All treatments were prepared in 20 mL glass scintillation vials (Supplementary Note 3) and stored in the dark at 4 °C. Treatments included:

a) PEST only. For copepods, a 2.5 mL aliquot of the 10 μm PEST stock solution was diluted to obtain an exposure concentration of ~2500 microfibres (Experiment 1), and 13 mL to obtain ~13,000 microfibres (Experiment 2). For mysid shrimp and fish, larger microfibres (i.e., 350 μm; $n = 60$ for mysid shrimp, and 750 μm; $n = 10$ for fish) were handpicked under magnification (Leica M80, 0.75–6.0× magnification) using a long needle and placed into individual vials. All PEST vials were made up to a final volume of 15 mL with 1 μm FSW.
b) DEHP only. DEHP vials were prepared by adding DEHP (0.5 mg vial^{-1} for copepods and mysid shrimp, 50 mg vial^{-1} for fish) to 1 mL dimethyl sulfoxide (DMSO) for a final concentration of 1 mg L^{-1} DEHP in the experimental tank. DEHP is lipophilic, hence DMSO was used as a carrier solvent to ensure homogeneous distribution of DEHP during exposures. Vials were made up to a final volume of 15 mL with 1 μm FSW immediately prior to dosing into experimental tanks.
c) DEHP pre-adsorbed to PEST (PEST:DEHP). For the preparation of vials assigned to the 10 μm PEST:DEHP treatments, vials were similarly prepared for both experiments as described for PEST only treatments (refer above). Volumes were evaporated to dryness under a slow stream of nitrogen at 30 °C overnight. Larger PEST microfibres were handpicked (as per PEST only vials; refer above). Hexane (1 mL; Fisher Scientific, CAS Number 110–54-3) was added to each vial followed by the relevant amount of DEHP to give final concentration of 1 mg L^{-1} for each organism (0.5 mg vial^{-1} for copepods (experiment 1 and 2) and mysid shrimp, 50 mg vial^{-1} for fish). Vials were capped (aluminium foil lined) and shaken on an orbital shaker (125 rpm, Baxter Multi-Tube Vortexer, Baxter Diagnostics) for 1 h. Hexane was then left to fully evaporate overnight under a continuous nitrogen stream at 30 °C and vials were stored capped (Supplementary Note 5 and Supplementary Fig. 4). Immediately prior to dosing, 15 mL of 1 μm FSW was added to each vial and vigorously shaken.
d) PEST and DEHP added in tandem (PEST+DEHP). These treatments were dosed using two separate scintillation vials, one containing PEST and the other DEHP; all vials were prepared as described above.
e) Controls. For Experiment 1, each timepoint included two control replicates containing only FSW (15 mL of 1 μm FSW with no PEST or DEHP) and one solvent control comprising 1 mL dimethyl sulfoxide in 14 mL 1 μm FSW (DMSO; 18 controls total, 6 as solvent controls). For experiment 2, each treatment included five FSW control replicates and three DMSO solvent controls (8 controls total, 3 as solvent controls).

Treatments were dosed by emptying prepared vials into experimental tanks and rinsing vials (x3) with 1 μm FSW. Alongside exposures, a small amount of normal feed for each organism was added: 1250 (Experiment 1) or 7500 (Experiment 2) cells of T-Iso and *C. muelleri* mixture (1:1) for copepods, 30 frozen copepods for mysid shrimp, or 5 frozen mysid shrimps for the fish. As Experiment 2 involved feeding pre-exposed prey items to the next trophic level, i.e., copepods to mysid shrimp, and mysid shrimp to fish, additional algal feed was only provided to copepods. Feed was kept at a ratio of 50 % of the PEST exposure quantity to promote normal feeding during the experiment (Savoca et al., 2016) and emulate environmentally realistic conditions. There was no starvation period for any organism during experimentation.

2.5. Experimental procedures

2.5.1. Experiment 1: ingestion and retention

All three species were individually exposed to one of five treatments for 30 min (copepods and mysid shrimp) or 1 h (fish) and left to depurate for 24 h and 48 h, respectively. Copepods (n = 250 per timepoint replicate) and mysid shrimp (n = 6 per timepoint replicate) were sampled at timepoints 0, 1, 3, 6, 12 and 24 h while fish (n = 1 per timepoint replicate) were sampled at 0, 3, 6, 12, 24, and 48 h (fish) (Fig. 1). A total of 3 replicates were conducted for each treatment timepoint for each species.

2.5.2. Copepods

Adult copepods were aliquoted from the culture population and transferred into 600 mL experimental glass tanks 24 h prior to exposures (n = 250 copepods per tank; 22,500 total). Tanks were covered with aluminium foil and continuously aerated using titanium rods for the duration of the acclimation and experimental periods. The health status of copepods was considered normal if active swimming and movement was observed throughout the water column. Only healthy copepods were exposed to the treatments mentioned above. Following a 30 min exposure period, tank contents were filtered over a 26 μm stainless steel filter net and rinsed gently with FSW. The filter net was then backwashed into new FSW and left for a set depuration period (0, 1, 3, 6, 12 or 24 h). Subsequently, copepods were emptied over a 26 μm stainless steel filter net, rinsed with 1 μm FSW, and then backwashed into an aluminium foil weigh-boat. Copepods were collected using a glass pipette and immediately fixed in 70 % ethanol (EtOH) for PEST analysis.

2.5.3. Mysid shrimp

Prior to exposures, mysid shrimp were removed from their culture tank, transferred into 600 mL experimental tanks (n = 6 per tank; 540 total) and left to acclimate (>12 h). Following a 30 min exposure to one of the treatments, tank contents were emptied over a 547 μm stainless steel filter and rinsed with FSW. Mysid shrimps were then backwashed into clean FSW and left to depurate (0, 1, 3, 6, 12 or 24 h). Following the set depuration period, mysid shrimp were filtered, rinsed with FSW, sampled used forceps, and immediately fixed in 70 % EtOH for PEST analysis.

2.5.4. Fish

Fish were left to acclimate in their individual 50 L glass flow-through tanks (n = 1 fish per tank; 90 fish total) for a minimum of 7 days prior to experimentation. Tanks were maintained at a flow rate of 0.8 L min^{-1} under continuous aeration. Fish were exposed to one of the five

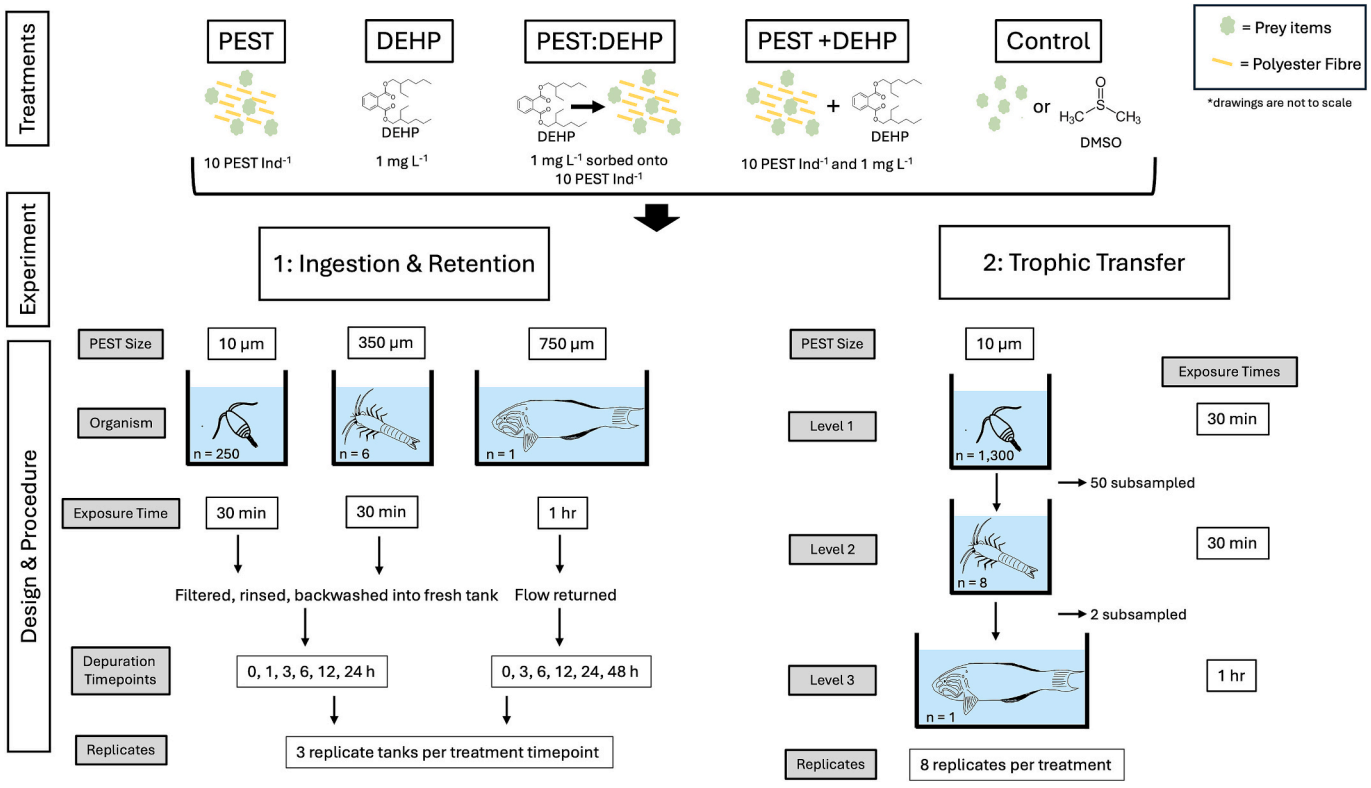

Fig. 1. Study designs for the two experiments conducted. Experiment 1: copepods (*Parvocalanus crassirostris*), mysid shrimp (Order: Mysida) and moon wrasse fish (*Thalassoma lunare*) exposed to a single dose of polyester (PEST) microfibres alone, bis(2-ethylhexyl) phthalate (DEHP) alone, PEST intentionally pre-adsorbed with DEHP (PEST:DEHP), PEST and DEHP added in tandem (PEST+DEHP), and 1 μm filtered seawater control (i.e., no PEST or DEHP) to determine PEST microfibre ingestion and retention rates. Experiment 2: the fate of PEST in PEST-burdened copepods was tracked through a simple linear food web consisting of mysid shrimp and moon wrasse. For Experiment 1, each timepoint included two control replicates containing only FSW (15 mL of 1 μm FSW with no PEST or DEHP) and one solvent control comprising 1 mL dimethyl sulfoxide in 14 mL 1 μm FSW (DMSO; 18 controls total, 6 as solvent controls). For experiment 2, each treatment included five FSW control replicates and three DMSO solvent controls (8 controls total, 3 as solvent controls).

treatments in triplicate with flow halted (i.e., static). Following the 1 h exposure period, tanks were siphoned to remove any debris accumulating at the bottom and flow was returned to the tanks. Fish were left for set depuration times (0, 3, 6, 12, 24, or 48 h) after which they were euthanised via ice slurry (ice and FSW mixture) and stored frozen (−20 °C) in aluminium foil until dissection.

2.5.5. Experiment 2: trophic transfer

Trophic transfer of PEST was assessed across a linear three-tier food web, copepods representing the primary consumer (level 1), mysid shrimp representing the secondary consumer (level 2), and fish representing the tertiary consumer (level 3) (Fig. 1). Copepods (n = 1300 tank^{-1}; n = 8 replicate tanks per treatment) were pre-exposed to treatments with food for 30 mins, then tank contents were emptied over a 26 μm stainless steel filter net and gently rinsed with FSW. The filter was partially submerged in clean 1 μm FSW (20 mL) and 50 copepods were subsampled using a glass pipette and fixed with 70 % EtOH to confirm PEST intake. The remaining copepods (n = 1250) were subsequently fed to mysid shrimp (n = 8 per tank; n = 8 replicate tanks per treatment). After 30 mins, mysid shrimp tank contents were emptied over a 547 μm stainless steel filter net and rinsed with FSW. Mysid shrimp (n = 2 mysid shrimp per tank) were subsampled using forceps and fixed in 70 % EtOH. The remaining mysid shrimp (n = 6) were fed to the fish (n = 6 mysid shrimp per fish; n = 8 replicate tanks per treatment). Fish were allowed 1 h to consume prey during which time prey consumption was recorded using GoPro video cameras (Model Hero5 and above). GoPro cameras were installed at each fish tank, outside of the water, at least 12 h prior to experimentation to ensure behavioural acclimation. The experiment was terminated by euthanising fish via ice slurry (ice and FSW). Fish were stored frozen (−20 °C) in aluminium foil until dissection.

2.5.6. PEST quantification

Copepods were emptied over 0.45 μm polytetrafluoroethylene (PTFE; Omnipore) filters and rinsed with 70 % EtOH to remove extraneous MPs. Copepods were then transferred to a pre-cleaned glass microscope slide and a cover slide applied. Individual mysid shrimp were positioned on a microscope slide with the dorsal side facing upwards using forceps and compressed with a cover slide. Frozen fish were thawed and dissected to extricate the gastrointestinal tract (GIT; stomach and intestines) and gills. Following an established protocol (Schlawinsky et al., 2022), fish GITs were digested using potassium hydroxide (10 % KOH) for 48 h at 40 °C. Digestate was filtered onto 0.45 μm PTFE filters (13 mm diameter) and rinsed liberally with Milli-Q water. Filters were transferred to a glass microscope slide using forceps and secured in place with a cover slide. Gills were laid flat and compressed between two glass microscope slides.

As the PEST was fluorescent, an inverted fluorescent microscope (Leica DMI6000B; DFC310FX camera attachment) was used to identify and quantify PEST microfibres within each organism. Copepods, mysid shrimp, fish gills and remnants of digested fish GITs were visualised under magnification and photographed under brightfield light (EMP TL-BF) using a blue light filter (I3). This filter was chosen to ensure PEST fibres were clearly visible while minimising any interference by autofluorescencing biological material. For Experiment 1, PEST microfibres were observed under magnification, counted, photographed and size measured using the freehand line tool in ImageJ (FIJI; version 1.53e). For Experiment 2, sample filters were photographed, and PEST counts performed using the ImageJ 'analyse particles' function. Each photo contained an embedded scale bar that was used to calibrate the ImageJ scale and transformed into an 8-bit image with a threshold set, ensuring all fluorescent microfibres were selected (Supplementary Fig. 5). Due to the tendency of 10 μm PEST microfibres to cluster within the GIT of mysid shrimp (Experiment 2), the area measurements obtained from ImageJ were corrected using the size of the PEST microfibres (i.e., 10 × 10 μm or 100 μm^2) to obtain a PEST microfibre count estimate.

2.6. Data analysis

Reporting MP individual^{-1} for contaminated organisms is standard practice in MP research as plastics are in essence heterogenous particulate matter varying in shape, size and polymer type (Andrady, 2011). Traditionally, body burden of a pollutant is reported as the weight of pollutant per gram weight of the analysed tissue (Thornton et al., 2002); however, quantifying the weight of MPs ingested is not routinely done, due primarily to difficulties associated with handling particulate matter less than 5 mm (Rivers et al., 2019). Therefore, to account for the potential difference in impacts particulate MPs may have on smaller compared to larger organisms, the literature has adapted the classical definition of body burden to be representative of the number of MPs per gram weight of organism (Jensen et al., 2019; Kroon et al., 2018; Santana et al., 2021). Here, PEST ingestion, retention and transfer are reported in both units (i.e., per gram weight of organism and per individual) for all three organisms, to allow appropriate conclusions to be made, as well as facilitate comparisons with previous studies (Cowger et al., 2020).

PEST ingestion and retention was determined individually for copepods, mysid shrimp, and fish as the number of PEST microfibres present in the GIT of each organism at a given timepoint. Ingestion is determined based on the number of PEST microfibres within an organism's GIT immediately following the exposure period (i.e., T_0). Retention is representative of the number of PEST microfibres in an organism's GIT throughout the depuration period. Depuration rates for each species treatment were calculated by subtracting the average number of PEST microfibres in an organism's GIT at a particular timepoint from the quantity of PEST microfibres at T_0 (i.e., initial ingestion), and dividing by the elapsed time since exposure ended to get an egestion rate per hour. Mean ingestion, retention, and depuration rates of PEST microfibres are presented as average ± standard deviation (S.D.) of replicates per treatment timepoint. To transform to gram per weight, average weights of copepods and shrimps previously collected from field studies (Miller et al., 2023), as well as total fish weights measured during experimentation, were utilised. Bioconcentration and biomagnification factors were calculated by adapting the formulas outlined in Miller et al. (2023). In brief, bioconcentration factors (BCF) were considered the quantity of PEST microfibres ingested (g per gram weight of organism) divided by the nominal exposure concentration (g L^{-1}); biomagnification factors (BMF) were considered the quantity of PEST microfibres ingested (g per gram weight of organism) divided by the quantity of PEST microfibres ingested by the previous trophic level (g per gram weight of organism). BMF values for copepods could not be determined given prey was not analysed for PEST. If BCF and BMF values are greater than 1, PEST is considered to be bioconcentrating and biomagnifying, respectively.

To assess differences in initial ingestion between treatments, three general linear models (GLMs) were run with Gaussian distributions for each species from Experiment 1 (same GLM formula for each, see Supplementary Table 5). To determine the differences of PEST ingestion between species and treatment from Experiment 2, a separate GLM was conducted using a negative binomial distribution due to the non-normality of the data (see Supplementary Table 6). Post-hoc analyses were done using the estimated marginal means (EMM) from the models for pairwise comparisons. To account for a possible tank effect, tank number was included as a nested variable within each model. All statistical analysis and graph construction was done using PEST individual^{-1} data in R (Version 4.0.4 with Rstudio, version 1.4.1106), with the 'emmeans' package for calculating EMMs, the 'ggfortify' and 'DHARMa' packages to check model assumptions, the 'pairs' package to conduct post-hoc pairwise comparisons and the 'ggplot2' package for graph creations.

3. Results

3.1. PEST ingestion and retention (Experiment 1)

All three taxa individually exposed to five treatments in short-term acute exposure laboratory experiments were confirmed to ingest PEST microfibres (Fig. 1). PEST microfibres were found within all individual mysid shrimp and fish exposed to PEST (regardless of treatment); however, only 93.3 %, 95.2 % and 97.4 % of copepods demonstrated PEST ingestion, depending on the treatment (i.e., PEST alone, PEST+DEHP, and PEST:DEHP, respectively). Considering all treatments, initial (i.e., T_0) ingestion of PEST microfibres varied for each taxon (Table 1a; Fig. 2), with copepods ingesting <1 % of offered PEST microfibres (average 0.48 ± 0.20 %), mysid shrimp consuming an average of 27.28 ± 18.63 %, and fish ingesting an average of 68.89 ± 19.00 % (Supplementary Table 7). For all species, levels of PEST contamination decreased over the defined experimental period, indicating PEST retention is temporary and that full PEST depuration is possible. No PEST microfibres were found in organisms exposed to control or DEHP only treatments. No changes in organism behaviour were observed between controls or any of the treatments.

Assessment of exposed copepods found T_0 ingestion ranged from 8.08 ± 1.71 to 17.47 ± 1.76 PEST individual^{-1} (1625.85 ± 343.38 to 3514.42 ± 353.70 PEST g^{-1} copepod) and was dependent on the PEST treatment (Fig. 2, Table 1a). For some copepods, there were signs of PEST depuration at T_1 and by T_3 PEST contamination had dropped to similar levels (2.00 ± 1.33 to 3.17 ± 1.96 PEST individual^{-1}) irrespective of treatment (Fig. 2). Mean copepod depuration rates were treatment-specific with ranges between 1.55 ± 0.77 PEST h^{-1} (PEST only) and 5.77 ± 1.27 PEST h^{-1} (PEST + DEHP) (Supplementary Table 5). The recovered PEST MPs were confirmed by microscopy to be of the same shape and similar size (10.2 ± 2.3 μm) as the original stock PEST microfibres. No PEST microfibres were present in copepods at T_6, indicating that retention in *P. crassirostris* is less than 6 h.

For mysid shrimp at T_0, ingestion across the three PEST treatments ranged between 9.11 ± 4.70 and 21.33 ± 15.94 PEST individual^{-1} (169.67 ± 87.57 and 397.27 ± 296.79 PEST g^{-1} mysid; Table 1a). Average retention times for PEST microfibres in mysid shrimp were at least 24 h; however, many individuals (n = 15, 55.6 %) demonstrated complete depuration by 12 h, with some clear of PEST as early as 3 h post-exposure (n = 6, 22.2 %). Mean depuration rates followed an opposite pattern to copepods, with depuration quicker for PEST only treatments (3.33 ± 2.83 PEST h^{-1}) compared to treatments with DEHP added either in tandem (1.12 ± 0.63 PEST h^{-1}) or pre-adsorbed to PEST (0.73 ± 0.51 PEST h^{-1}). Furthermore, the PEST microfibres recovered from mysid shrimp GITs were considerably smaller (57.6 ± 27.6 μm) than the original stock 350 μm PEST, indicating fragmentation of microfibres had occurred during ingestion.

At T_0, ingestion of PEST by fish across the three PEST treatments ranged from 5.67 ± 2.31 to 8.33 ± 1.53 PEST individual^{-1} (0.19 ± 0.10 to 0.35 ± 0.32 PEST g^{-1} fish; Table 1a). All fish demonstrated retention of PEST for at least 12 h, with most individuals retaining PEST up to 24 h (n = 7, 77.8 %), and some up to 48 h (n = 3, 33.3 %). Similar to the mysid shrimp, fish exposed to PEST alone had the quickest depuration rates (0.51 ± 0.31 PEST h^{-1}) compared to DEHP treatments (both 0.32 ± 0.15 PEST h^{-1}). The PEST recovered from the fish were of sizes similar to that of the original stock PEST (i.e., 731.7 ± 62.0 μm). No PEST was observed in the gills of exposed fish.

DEHP, either added simultaneously with PEST, or pre-adsorbed to PEST, influenced the ingestion of PEST microfibres and was species-specific. Copepods exposed to treatments containing DEHP ingested higher amounts of PEST (PEST+DEHP > PEST:DEHP > PEST; Fig. 2), with T_0 ingestion significantly different across the three PEST treatments (Experiment 1; GLM; $p < 0.05$; Supplementary Table 6). Regardless, and despite differences in PEST quantities ingested for the different PEST treatments, DEHP did not affect the final depuration time. All copepods were free of PEST by T_6. In contrast, for mysid shrimp, the quantity of PEST ingested at T_0 was significantly reduced when PEST microfibres were pre-adsorbed with DEHP (PEST:DEHP) compared to PEST alone (p = 0.046; Supplementary Table 6b), yet the simultaneous addition of DEHP (PEST+DEHP) had no influence on PEST ingestion. Mysid shrimp exposed to treatments containing DEHP were clear of PEST under 24 h (T_{24}), with percent PEST retained lower than those exposed to PEST alone (Supplementary Table 7). Similarly, initial PEST ingestion (T_0) by fish was lower when exposed to the two DEHP treatments (Fig. 2), albeit this was not significant (Supplementary Table 6). Across all treatments MPs were still present in some fish up to 48 h, although for those exposed to PEST+DEHP treatments, most individuals (66.7 %) had fully depurated by 12 h (Fig. 2).

3.2. Trophic transfer of PEST (Experiment 2)

Copepods exposed to the three PEST treatments were confirmed by fluorescence microscopy to have ingested these PEST microfibres (Figs. 3 and 4a). Similarly, PEST microfibres were observed in mysid shrimp fed PEST-burdened copepods (Figs. 3 and 4b). Video evidence revealed consumption of (assumed) PEST-burdened mysid shrimp prey - offered in a sequential linear trophic transfer design - by fish predators (Figs. 3 and 4c). No changes in fish behaviour were observed between experiments. PEST microfibres were found within all individual organisms that had consumed prey burdened with PEST, regardless of PEST treatment or trophic level. PEST was again absent from fish gills and no PEST was found in the control or DEHP alone exposed organisms. Based on PEST individual^{-1}, an increase in PEST contamination was observed from the primary trophic level to the tertiary level, with PEST contamination significantly different between the three species (all p-values <0.001; Supplementary Table 8; Figs. 3 and 4). The overall trend was a 14.6-fold increase of MPs from copepods to mysid shrimp and a 4.3-fold increase from mysid shrimp to fish (Table 1). From trophic level 1 to 3 (i.e., copepods through to fish), there was a 62.5-fold increase in PEST concentration. Interestingly, if using standardised PEST quantities (by weight; PEST gram^{-1} organism), a trend of decreasing PEST ingestion is observed as trophic level increases (Table 1b–c).

The influence of DEHP on ingestion of PEST-burdened prey followed the same trend as observed in Experiment 1, with highest ingestion occurring for PEST+DEHP treatments, albeit not significantly so (all p-values >0.05; Supplementary Table 6). DEHP did not significantly influence the quantity of PEST transferred from copepods to mysid shrimp or mysid shrimp to fish (all p-values >0.05; Supplementary Table 8).

3.3. Bioconcentration and biomagnification factors

Bioconcentration factors (BCF) and biomagnification factors (BMF) were calculated for the Experiment 1 and 2, respectively (Table 2). Copepods and mysid shrimp, regardless of treatment, resulted in a BCF > 1, indicating bioconcentration of PEST occurred. Fish exposed to PEST alone demonstrated bioconcentration of PEST, whereas fish exposed to PEST+DEHP or PEST:DEHP did not. Despite PEST microfibres bioconcentrating in copepods across all treatments in Experiment 1, PEST microfibres were only shown to biomagnify from copepods to mysid shrimp in PEST alone and PEST:DEHP treatments in Experiment 2. Fish showed no biomagnification of PEST microfibres (all BMFs <1) in Experiment 2 (Table 2). Bioconcentration factors (BCF) (a; Experiment 1), and biomagnification factors (BMF) (b; Experiment 2) for copepods, mysid shrimp and fish following exposure to PEST alone (PEST), PEST pre-adsorbed with DEHP (PEST:DEHP), and PEST added in tandem with DEHP (PEST+DEHP). Experiment 1 consisted of individual exposures, whereas Experiment 2 was comprised of a linear food chain of exposing the base level of copepods only, then feeding copepods to mysid shrimp and the mysid shrimp to the fish. BCFs are representative of the weight of PEST ingested (g gram^{-1} organism) divided by the weight of the PEST dose (g L^{-1}). BMFs are representative of the weight of PEST ingested by

Table 1
Ingestion (T_0) of polyester microfibres (PEST individual^{-1} and PEST gram^{-1} organism) for copepods, mysid shrimp, and fish (a) over time to assess ingestion and retention (Experiment 1) and (b + c) within a trophic transfer scenario (Experiment 2). Treatments include PEST alone (PEST), bis(2-ethyl hexyl) phthalate (DEHP) alone, PEST pre-adsorbed with DEHP (PEST:DEHP), and PEST added in tandem with DEHP (PEST+DEHP). Experiment 1 includes ingestion (T_0) and retention times, while Experiment 2 includes T_0 ingestion and percentage increases along trophic interactions. Ingestion and retention data are presented as mean ± standard deviations among replicates ($n = 3$ Experiment 1, $n = 8$ Experiment 2), and percent increases are presented as percent ± standard deviation (SD).

(a) Experiment 1: ingestion

Organism	Retention time (h)	**PEST individual^{-1}** *(PEST gram^{-1} organism)*		
		PEST	PEST:DEHP	PEST+DEHP
Copepod ($n = 750$)	0	**8.08 ± 1.71** *(1625.85 ± 343.38)*	**9.50 ± 4.63** *(1911.47 ± 932.43)*	**17.47 ± 1.76** *(3514.42 ± 353.70)*
	1	**5.65 ± 2.36** *(1136.82 ± 475.60)*	**6.96 ± 4.13** *(1401.00 ± 831.09)*	**10.80 ± 3.65** *(2173.04 ± 734.46)*
	3	**2.00 ± 1.33** *(402.41 ± 266.86)*	**3.17 ± 1.96** *(614.21 ± 394.10)*	**2.85 ± 2.59** *(573.44 ± 521.04)*
	6	**0.0**	**0.0**	**0.0**
	12	**0.0**	**0.0**	**0.0**
	24	**0.0**	**0.0**	**0.0**
Mysid shrimp ($n = 18$)	0	**21.33 ± 15.94** *(397.27 ± 296.79)*	**9.11 ± 4.70** *(169.67 ± 87.57)*	**18.67 ± 6.08** *(347.61 ± 113.27)*
	1	**13.56 ± 7.86** *(252.43 ± 146.37)*	**8.89 ± 5.93** *(165.53 ± 110.34)*	**18.44 ± 6.77** *(343.47 ± 125.99)*
	3	**8.22 ± 5.14** *(153.11 ± 95.76)*	**4.67 ± 6.63** *(86.90 ± 123.52)*	**14.44 ± 7.73** *(268.98 ± 143.98)*
	6	**7.56 ± 10.09** *(140.70 ± 187.87)*	**3.11 ± 4.91** *(57.94 ± 91.44)*	**7.56 ± 9.68** *(140.70 ± 180.33)*
	12	**3.78 ± 3.38** *(70.35 ± 63.00)*	**1.56 ± 3.43** *(28.97 ± 63.91)*	**2.44 ± 3.97** *(45.52 ± 73.97)*
	24	**3.33 ± 4.36** *(62.07 ± 81.77)*	**0.89 ± 2.03** *(16.55 ± 37.76)*	**0.67 ± 1.41** *(12.41 ± 26.34)*
Fish (n = 3)	0	**8.33 ± 1.53** *(0.35 ± 0.32)*	**6.67 ± 1.15** *(0.11 ± 0.03)*	**5.67 ± 2.31** *(0.19 ± 0.10)*
	3	**5.67 ± 0.58** *(0.33 ± 0.43)*	**5.67 ± 1.15** *(0.28 ± 0.34)*	**4.33 ± 0.58** *(0.07 ± 0.02)*
	6	**3.67 ± 1.15** *(0.14 ± 0.15)*	**3.67 ± 1.15** *(0.12 ± 0.06)*	**3.00 ± 1.00** *(0.07 ± 0.05)*
	12	**2.67 ± 1.15** *(0.12 ± 0.01)*	**1.33 ± 0.58** *(0.07 ± 0.06)*	**1.00 ± 1.00** *(0.04 ± 0.04)*
	24	**2.00 ± 1.00** *(0.07 ± 0.03)*	**1.67 ± 1.15** *(0.13 ± 0.18)*	**0.67 ± 0.58** *(0.03 ± 0.04)*
	48	**0.33 ± 0.58** *(0.05 ± 0.09)*	**0.33 ± 0.58** *(0.02 ± 0.04)*	**0.33 ± 0.58** *(0.01 ± 0.02)*

(b) Experiment 2: ingestion

Treatment	**PEST individual^{-1} ± SD** *(PEST gram^{-1} organism ± SD)*		
	Copepod	Mysid shrimp	Fish
PEST	**2.16 ± 0.86** *(86.40 ± 34.32)*	**31.63 ± 24.95** *(158.13 ± 124.76)*	**135.13 ± 62.41** *(3.90 ± 1.97)*
PEST:DEHP	**2.53 ± 2.34** *(101.10 ± 93.53)*	**28.63 ± 22.83** *(143.13 ± 114.17)*	**146.13 ± 61.65** *(4.19 ± 2.42)*
PEST+DEHP	**4.06 ± 4.75** *(162.40 ± 190.17)*	**30.13 ± 13.28** *(150.63 ± 66.41)*	**163.63 ± 99.15** *(6.38 ± 5.05)*

(c) Experiment 2: percent changes

Treatment	**% ± SD for PEST individual^{-1}** *(% ± SD for PEST gram^{-1} organism)*		
	Copepod → Mysid	Mysid Shrimp → Fish	Copepod → Fish
PEST	**1739.42 ± 1669.00** *(70.24 ± 154.47)*	**1115.25 ± 1503.16** *(−98.20 ± 2.09)*	**7759.77 ± 6193.75** *(−98.98 ± 0.66)*
PEST:DEHP	**2309.13 ± 3926.92** *(122.97 ± 363.41)*	**749.83 ± 758.25** *(−98.79 ± 0.99)*	**14,645.20 ± 21,623.00** *(−97.52 ± 4.46)*
PEST+DEHP	**1845.02 ± 2170.30** *(80.01 ± 200.86)*	**438.49 ± 252.48** *(−98.86 ± 0.94)*	**10,937.55 ± 13,588.20** *(−97.44 ± 3.65)*

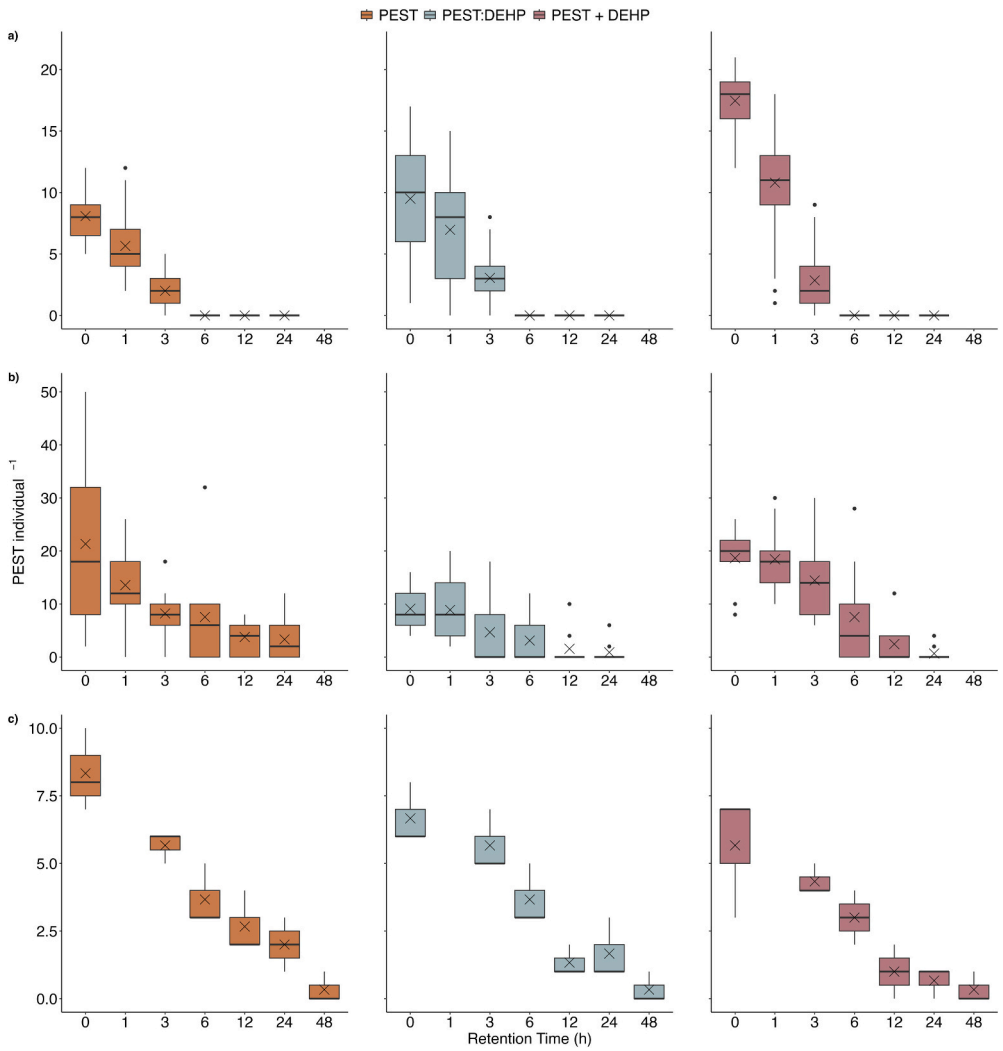

Fig. 2. Mean polyester (PEST) ingestion and retention by (a) copepods (*Parvocalanus crassirostris*, $n = 250$ per replicate), (b) mysid shrimp (Order: Mysida; $n = 6$ per replicate) and (c) moon wrasse fish (*Thalassoma lunare*; $n = 1$ per replicate), and the cumulative impact on PEST ingestion of the plasticiser bis(2-ethylhexyl) phthalate (DEHP). Treatments include a single dose of: PEST, PEST intentionally pre-adsorbed with DEHP (PEST:DEHP), and PEST dosed simultaneously with DEHP (PEST+DEHP). Treatments were dosed at a concentration of 10 PEST individual^{-1}; DEHP at a concentration of 1 mg L^{-1}. Data are presented as PEST Individual^{-1}. Each timepoint sampled three replicates per treatment; however, T_{48} was not measured for copepods or mysid shrimp and T_1 was not measured for fish. Middle line = median, X = mean, boxes = interquartile range (IQR), and whiskers = 1.5 times IQR.

the predator (g gram^{-1} predator) divided by the weight of PEST ingested by the prey (g gram^{-1} prey); BMF for copepods is not possible given prey was not analysed for PEST. If BCF and BMFs values are greater than 1, PEST is considered to be bioconcentrating or biomagnifying.

4. Discussion

Quantifying the ingestion, retention, depuration, and transferability of MPs is critical to elucidate ecological fate and potential impact of MPs in marine food webs. This study quantifies the ingestion, retention and depuration of environmentally relevant PEST microfibres by organisms linked intrinsically through a linear food web (i.e., copepods, mysid shrimp and fish), and demonstrates the trophic transfer of PEST MPs from the lowest to the highest trophic level. All three trophic levels readily ingested PEST microfibres when directly exposed. Ingestion levels for mysid shrimp and fish increased 1.3-fold and 25-fold higher, respectively, under trophic transfer scenarios and retention times increased substantially with higher trophic position. The direct ingestion, retention and depuration of PEST was significantly influenced by the presence of the plasticising phthalate DEHP, suggesting possible species specificity. In contrast, the addition of DEHP had no influence on the trophic transfer of PEST once ingested by the lowest trophic level. Once standardised by weight of the organism, trophic transfer trends reversed; albeit PEST microfibres were still found to bioconcentrate in copepods exposed to PEST+DEHP and biomagnify in mysid shrimp exposed to PEST-burden copepods. Despite this, given the high turnover rate of lower trophic level consumption within oceanic food webs (Barbier and Loreau, 2019), the potential for transfer of MPs, and associated chemical contaminants, to higher tropic levels is probable. While the concept of trophic transfer of MPs and chemical additives is underdeveloped and warrants further investigation, our findings suggest that the potential ecological implications could be significant.

4.1. Ingestion, retention and depuration rates

Ingestion of PEST by copepods here is lower than previously reported for MP consumption in other calanoid (i.e., *Calanus helgolandicus* (Cole et al., 2015; Procter et al., 2019)) and cyclopoid (i.e., *Mesocyclops isabellae* (Yadav et al., 2024)) copepods. Given MP ingestion is often correlated to exposure concentration (Domínguez-López et al., 2022; Santana et al., 2021), the disparity between this study and others could be attributed to the lower (and hence being more environmentally

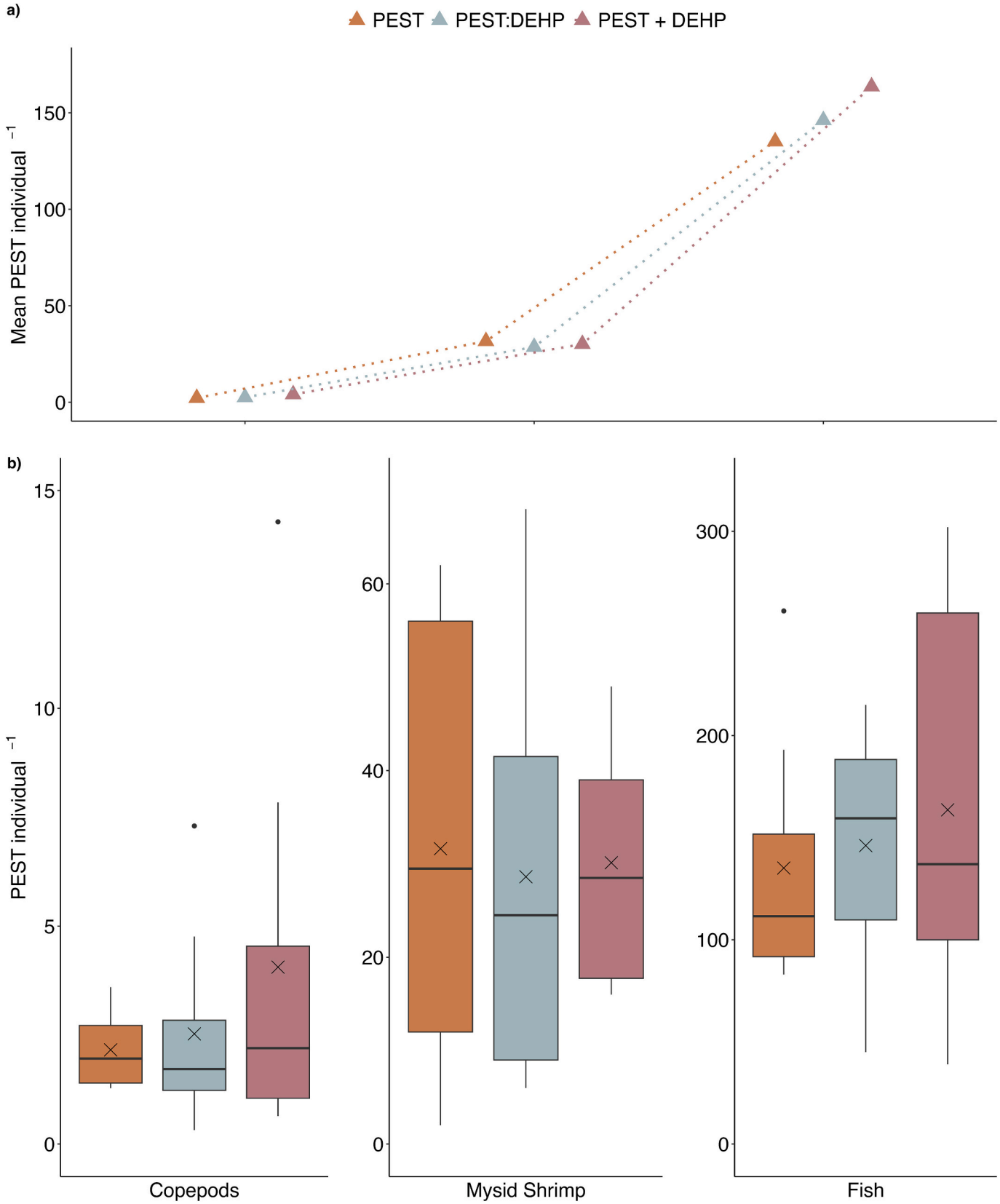

Fig. 3. Trophic transfer of polyester microfibres (10 μm PEST) and the cumulative influence of the plasticiser bis(2-ethylhexyl) phthalate (DEHP). Copepods were exposed to an acute dose of one of five treatments: PEST, 1 mg L^{-1} bis(2-ethyl hexyl) phthalate (DEHP), PEST pre-adsorbed with DEHP (PEST:DEHP), PEST dosed simultaneously with DEHP (PEST+DEHP) and controls (i.e., no PEST or DEHP). a) Mean PEST recovered per individual PEST-burdened copepod (*Parvocalanus crassirostris*), and per individual mysid shrimp (Order: Mysida) and moon wrasse fish (*Thalassoma lunare*) exposed to PEST via trophic transfer. No PEST were recovered from control or DEHP alone individuals and therefore these treatments are not visually shown. b) A statistical description of each dataset (middle line = median, X = mean, boxes = interquartile range (IQR), and whiskers = 1.5 times IQR) is presented.

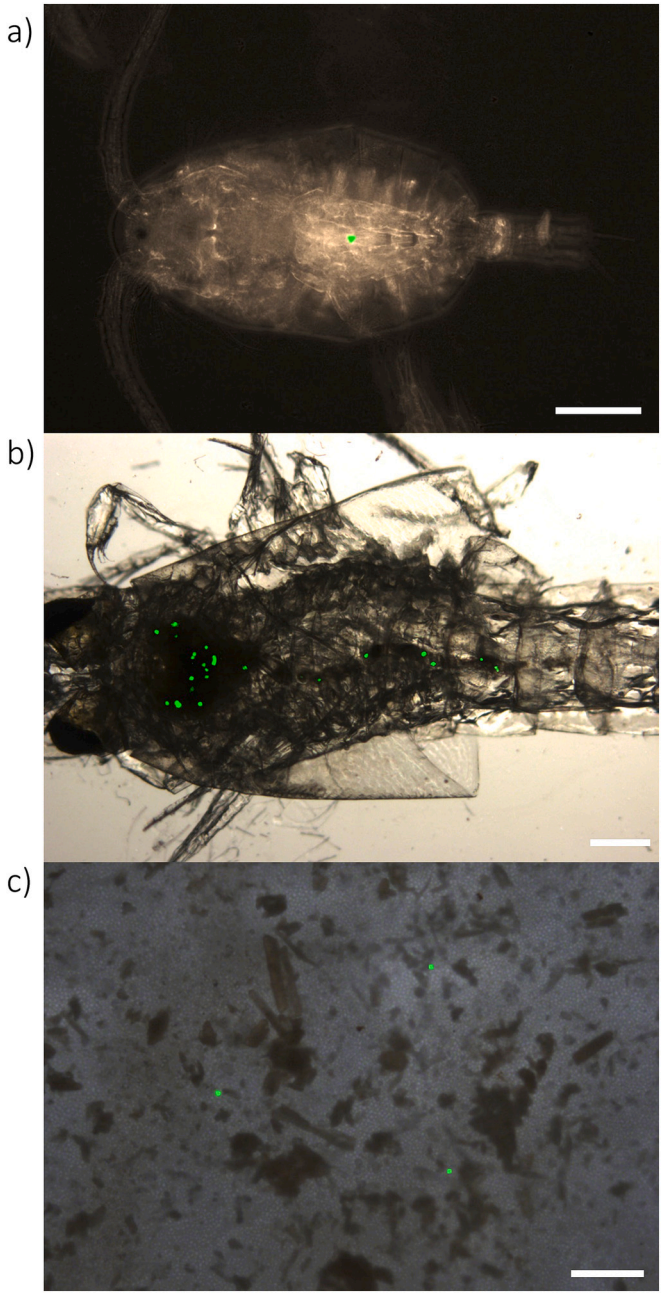

Fig. 4. Fluorescent polyester microfibres (10 μm PEST) identified in (a) copepod, *Parvocalanus crassirostris*, (b) mysid shrimp, Order: Mysida, and (c) in the gut contents of moon wrasse fish, *Thalassoma lunare*. PEST in mysid shrimp and fish are a direct result of trophic transfer from copepods. White scale bar is representative of 100 μm.

relevant) exposure quantities (i.e., 5 PEST mL^{-1}; Experiment 1) employed here. Alternatively, the MP polymer type (PEST versus polystyrene (PS) (Cole et al., 2015; Yadav et al., 2024) and nylon (Procter et al., 2019)) may have influenced the ingestion. However, when assessing trophic transfer (Experiment 2), PEST ingestion was lower despite a concomitant increase in copepod density, food and PEST concentration (i.e., 26 MPs mL^{-1}). This lower rate is potentially a result of increased competition between conspecifics. Yet, irrespective of the exposure concentration, the percent MP ingestion quantified here, and in previous studies (Cole et al., 2015; Procter et al., 2019), is extremely low (i.e., <1 %). The lower PEST ingestion rates observed for copepods when at higher density, coupled with the <6 h retention time and quick depuration established in Experiment 1, emphasises that if exposure to

Table 2
(a) BCFs (Experiment 1).

Organism	PEST dose (g L^{-1})	Treatment	Average ingested PEST		BCF
			Number $gram^{-1}$ organism	g $gram^{-1}$ organism	
Copepod	4×10^{-5}	PEST	1625.75	1.30×10^{-4}	3.25
		PEST+DEHP	3514.42	2.81×10^{-4}	7.02
		PEST:DEHP	1911.47	1.53×10^{-4}	3.82
Mysid shrimp	3×10^{-6}	PEST	397.27	1.11×10^{-4}	37.08
		PEST+DEHP	347.61	9.73×10^{-5}	32.44
		PEST:DEHP	169.67	4.75×10^{-5}	15.84
Fish	1.2×10^{-7}	PEST	0.35	2.08×10^{-7}	1.73
		PEST+DEHP	0.19	1.17×10^{-7}	0.97
		PEST:DEHP	0.11	6.68×10^{-8}	0.56

(b) BMFs (Experiment 2)

Organism	Treatment	Ingested PEST		BMF
		Number $gram^{-1}$ organism	g $gram^{-1}$ organism	
Copepod	PEST	434.61	3.48×10^{-5}	
	PEST+DEHP	816.9	6.54×10^{-5}	
	PEST:DEHP	508.55	4.07×10^{-5}	
Mysid shrimp	PEST	588.92	4.71×10^{-5}	1.36
	PEST+DEHP	560.99	4.49×10^{-5}	0.69
	PEST:DEHP	533.05	4.26×10^{-5}	1.05
Fish	PEST	3.9	3.12×10^{-7}	0.01
	PEST+DEHP	6.38	5.11×10^{-7}	0.01
	PEST:DEHP	4.19	3.25×10^{-7}	0.01

PEST is acute and intermittent (i.e., single exposure or non-continuous), complete clearance of fibrous MPs can be achieved in under 6 h for copepods. Adverse health outcomes have been reported for copepods exposed to MPs for >24 h (i.e., 10,000–80,000 polyethylene terephthalate fragments mL^{-1} (Heindler et al., 2017)) and may potentially be a consequence of continuous MP turn-over (i.e., intake, depuration, re-intake; over-burden scenario).

The observed positive influence of DEHP on copepod PEST ingestion and retention (PEST+DEHP > PEST:DEHP > PEST) is puzzling, especially given the toxic nature of DEHP (Seo et al., 2006); albeit copepod mortality rate was low for *P. crassirostris* adults even after exposure to 5120 μg L^{-1} DEHP (Heindler et al., 2017). The larger surface area-to-volume ratio of the smaller PEST particles (10 μm) to which copepods were exposed is expected to increase the (relative) amount of DEHP able to be adsorbed (PEST:DEHP) or which is available for sorption (PEST+DEHP) (Fred-Ahmadu et al., 2020). Expectations were that ingestion of PEST:DEHP and PEST+DEHP by copepods would have been lower compared to PEST alone, and ingestion by mysid shrimp and fish exposed to larger sized PEST:DEHP or PEST+DEHP microfibres would be less impacted. DMSO, used here as the carrier solvent (Brayton, 1986; Turner et al., 2012) for DEHP, may be a contributing factor. Copepods use chemosensory mechanisms to identify algal prey; for example, the presence of the algal gaseous metabolite dimethyl sulphide (DMS; the reduced form of DMSO and a known foraging attractant (Asher et al., 2017)) results in increased MP ingestion (Procter et al., 2019). The elevated ingestion of PEST+DEHP (i.e., DEHP introduced in DMSO) compared to PEST:DEHP (no DMSO) points to DMSO or its chemical reduction product DMS (Asher et al., 2015) acting not only as a copepod attractant but as a potential promoter of egestion. Thus, the use of DMSO in copepod ecotoxicity studies for the dispersal of lipophilic chemicals such as DEHP should be carefully considered.

The ingestion and retention of PEST microfibres by mysid shrimp here was lower than that observed for polyethylene (PE) beads in *Neomysis* sp. (Hasegawa and Nakaoka, 2021). It is difficult to establish whether this difference is a function of the higher concentrations (0.2 to 2 mg L^{-1} vs 0.003 mg L^{-1}) to which *Neomysis* sp. were exposed, or the smaller MP size (27–32 μm versus 350 μm), shape (bead versus fibre) or polymer type (PE versus PEST) (Bucci et al., 2020). Despite the larger

MP size used here, retention of PEST by mysid shrimp corroborates previous reported ranges determined for 1 and 10 μm PS beads travelling from the gastric mill to the anus of *Neomysis awatschensis* (Lee et al., 2021) (i.e., 35 min to 13.67 h post-exposure to PS). While this is the first report of detailed depuration rates for mysid shrimp, rates calculated here are higher than those reported for similar organisms (i.e., Antarctic krill *Euphausia superba*, 0.22 h^{-1} (Dawson et al., 2018)), potentially a consequence of their smaller size.

Being selective predators, mysid shrimp have the ability to actively reject food items they deem unpalatable (Viitasalo and Viitasalo, 2004). Somewhat perplexingly, the sorption of DEHP on PEST reduced the number of PEST microfibres ingested by mysid shrimp compared to exposure to PEST alone, yet the addition of DEHP and PEST in tandem had no influence on ingestion. The sorption of DEHP to PEST prior to exposure may act to concentrate DEHP on the surface of the PEST, causing active rejection or avoidance of ingestion in mysid shrimp, albeit the mechanisms behind this are not clear. Furthermore, DEHP exerted an opposing effect on mysid shrimp compared to copepods, evidenced by slower depuration rates relatively to treatments containing PEST microfibres alone. Additionally, the reduced ingestion of copepods burdened with PEST and DEHP indicates mysid shrimp may be capable of detecting the DEHP, or alternatively, that copepods emitted an unpalatable odour resulting from metabolised DEHP or DEHP-induced physiological stress. Yuan et al. (2022) established that intermediate trophic level organisms, such as the aquatic microcrustacean *Daphnia magna*, exhibit the highest sensitivity to phthalates, including DEHP, following 15-days exposure, in comparison to species at lower and higher trophic levels. The authors surmised that prolonged exposure to phthalates may disrupt aquatic food chain dynamics, potentially leading to an overabundance of primary producers such as algae. While this study investigated acute exposure (Mysid shrimp exposed to treatments or exposed copepod prey for 30 min, experiment 1 and 2, respectively), the findings not only support this disruption hypothesis but heighten concerns regarding the ecological impacts of MP-additive interactions on marine life.

Mysid shrimp were found to fragment larger 350 μm PEST microfibres upon ingestion (57.6 ± 27.6 μm), a process previously observed for other small crustaceans including *Neomysis* sp. (Hasegawa and Nakaoka, 2021) and Antarctic krill *E. superba* (Dawson et al., 2018). Small crustaceans have well-developed mandibles, as well as chitinous and thick barbed spines in their stomach (Dawson et al., 2018; Friesen et al., 1986) suited to grinding hard phytoplankton cell structures, and which promotes the mechanical breakdown of prey material for digestion. For Antarctic krill, the fragmentation of 31.5 μm MPs to <1 μm mimics this process (Dawson et al., 2018). Mysid shrimp have similar digestive mechanisms to krill, however, their larger size, their omnivorous diet, and preferential consumption of prey >10 μm in size (Friesen et al., 1986) may preclude fragmentation of MPs less than this size, with 57.6 ± 27.6 μm being the lower limit. The incorporation of MPs in copepod biomass may further reduce the efficiency of mysid shrimp mastication and limit the mechanical breakdown of <10 μm MPs, as fragmentation of 10 μm PE beads was similarly not reported following mysid shrimp exposure to pre-treated copepods and polychaete worms (Setala et al., 2014). If fragmentation of larger size MPs (i.e., >10 μm) by small crustaceans is common, the by-product may pose hazards to higher trophic level organisms (Hasegawa and Nakaoka, 2021) and could be a potential route for increased MP concentrations.

Ingestion of PEST microfibres, exposed contemporaneously with food, was confirmed for fish, with no PEST microfibres detected in the gills. Given the selective predatory nature of moon wrasse (Holmes and McCormick, 2006), the PEST intake is likely to have been incidental (i.e., collateral to the ingestion of food); however, selective MP intake is species-specific (Chan et al., 2019; Ryan et al., 2019) and the mechanism of intake of MPs for moon wrasse fish remains to be confirmed. The retention of PEST up to 48 h is longer than that reported for polypropylene (PP) fragments or PEST fibres in other reef fish (Santana et al., 2021), albeit *Pomacentrus amboinensis* are smaller in size and potentially have a faster gut passage time compared to *T. lunare* (Welden and Cowie, 2016). It is not known whether 48 h is considered prolonged retention, or whether MP retention beyond this time significantly impacts on fish health. Even so, impacts are likely given some MPs have been demonstrated to affect fish consumption, growth, reproduction, and survival (Foley et al., 2018).

Influences of DEHP on the ingestion and depuration of PEST microfibres in fish are comparable to those observed for the mysid shrimps. While not significantly so, PEST ingestion was lowest in the presence of DEHP with fish displaying signs of selectivity, i.e., active rejection of PEST microfibres. DEHP caused an analogous impact on egestion rates to mysid shrimp, with quicker depuration rates observed in the two treatments containing DEHP. However, this does not necessarily negate the potential adverse impacts of DEHP uptake (e.g., survival, fertility, swimming behaviour and growth) (Foley et al., 2018; Heindler et al., 2017; Lee et al., 2021). Although the impacts of retained PEST microfibres were not considered in this study, there is a need to investigate physiological and behavioural endpoints to further assess potential risk (Santana, 2022). The longer MPs are retained within the gut of an organism, the higher the likelihood that the associated phthalate additives will translocate into the body tissue, accumulate, metabolise and have further impact on animal health (Chua et al., 2014).

4.2. Trophic transfer of MPs to secondary and tertiary consumers

The trophic transfer of MPs through food webs, while increasingly studied, is seldom detailed (see Supplementary Literature Review and Supplementary Table 2). In situ research often speculates about trophic transfer (Dool and Bosker, 2022; Gamarra-Toledo et al., 2023; Justino et al., 2023; Welden et al., 2018) due to challenges in proving direct ingestion, including lack of exposure control, complex food webs, and the heterogeneous nature of MPs (e.g., varying shapes, sizes, colours), which complicates tracing the source of MPs in predators to ingested prey rather than (incidental) environmental exposure. In contrast, laboratory studies frequently use simplified exposure scenarios with high doses or uniform microplastics, limiting ecological relevance (Miller et al., 2020). Additionally, experimental studies rarely quantify MP transfer across more than two trophic levels, leaving gaps in our understanding of their fate in complex marine systems. Foundational knowledge in this area has been established by two experimental studies (Saikumar et al., 2024; Yadav et al., 2024). Saikumar et al. (2024) exposed *Artemia salina* to 1 μm fluorescent PS microspheres at a concentration of 10^6 MPs mL^{-1} for 24 and 48 h, and fed these to shrimp (*Litopenaeus vanamei*) and Nile tilapia (*Oreochromis niloticus)*, observing a decrease in MPs ingested as trophic levels increased. In contrast, Yadav et al. (2024) found exposing rotifers (*Brachionus plicatilis*) to <500 μm PP fragments at concentrations of up to 800 MPs L^{-1} for 20 min, followed by feeding them to copepods (*M. isabellae)* and subsequently seabass (*Lates calcarifer)*, resulted in an increase in MPs ingested as trophic level increased, matching observations in the current study. However, neither of these studies explored the impacts of a co-contaminant (e.g., DEHP or similar), as was done here. The shorter exposure times used here and employed by Yadav et al. (2024) likely reduced the opportunity of egestion of MPs prior to trophic transfer, potentially contributing to the difference in trophic accumulation patterns. These results highlight the importance of investigating trophic transfer under multiple scenarios, such as various MP sizes, shapes, concentrations, and exposure times for organisms of ecological importance.

Ingestion levels increased for mysid shrimp (level 2) and fish (level 3) by 1.3- to 25-fold, respectively, under trophic transfer scenarios (i.e., Experiment 2) in comparison to individual exposures (i.e., Experiment 1). Yu et al. (2024) reported a similar observation, where mysid shrimp, albeit the freshwater *Limnomysis benedeni,* consumed greater amounts

(ingested MPs were not quantified) of 4.8 μm PS microspheres when exposed via pre-burdened prey, comparatively to those exposed directly via the surrounding water. The selective feeding nature of both mysid shrimp (Viitasalo and Viitasalo, 2004) and moon wrasse fish (Holmes and McCormick, 2006) may also contribute to our findings, and indicate that these organisms could reject un-palatable food (e.g., MPs); however, further research is needed to substantiate these claims.

4.3. Bioconcentration and biomagnification

The classical concepts of bioconcentration and biomagnification, which are primarily applied to dissolved chemicals (Alexander, 1999), are now being considered to assess the ecological fate and risk of plastics (Covernton et al., 2022; Miller et al., 2023; Rochman et al., 2019). The body burden of a pollutant is traditionally reported as the weight of pollutant per gram weight of the analysed tissue (Thornton et al., 2002) and is an important component in (eco)toxicology assessments. Here, results reveal there is bioconcentration, or an increase of PEST in organisms compared to that of its surroundings (i.e., dosed concentration), for copepods and mysid shrimp exposed all treatments. This aligns with in situ observations of MPs bioconcentrating within copepods and benthic crustaceans (e.g., shrimps) compared to quantities in their surrounding environment (0.005 ± 0.004 MPs L^{-1}; (Miller et al., 2023)). Interestingly, PEST microfibres did not bioconcentrate for fish exposed to any treatments containing DEHP (PEST+DEHP or PEST:DEHP), despite in situ observations showing its potential (Miller et al., 2023).

Biomagnification (i.e., an increase in PEST within an organism compared to that of its prey) was only observed in mysid shrimp exposed to pre-burdened copepods exposed to PEST alone and PEST:DEHP treatments, returning BMFs of >1. The short transitional turn-over times (i.e., time between ending exposure and feeding to the next level) may explain why these trends are not also observed in mysid shrimp in situ (Miller et al., 2023). Alternatively, here, no fish exposed to the treatments generated a BMF > 1, corroborating in situ observations (Akhbarizadeh et al., 2019; Covernton et al., 2022; Miller et al., 2023). This may be a result of how BMF factors are calculated, and that the burden of relatively small-sized MPs (e.g., 10 μm PEST microfibres) coupled with the short exposure period (e.g., 1 h) and small number of prey consumed (e.g., 6 mysid shrimp) limits the chances of discerning biomagnification using the classical definition and formula. Furthermore, problems in data interpretation arise when transforming PEST ingestion from PEST per individual to PEST per gram weight of the organism analysed. For example, when looking simply at the number of PEST microfibres in each trophic level, there is an increase in PEST ingested observed across trophic levels. However, when standardised by body weight, magnification is no longer observed. Instead, the trend matches closer to that of global in situ data, with lower trophic levels containing higher levels of MPs and a decrease in contamination levels as trophic level increases. Global calculations of these endpoints were acquired using MPs $individual^{-1}$ (Miller et al., 2020) necessitated by the majority of MP reports employing these units and not reporting the body weight of organisms investigated, prohibiting traditional calculations and meaningful interstudy comparisons. Yet, it can be argued that as MPs are particulate matter, analysing ingestion reports in a similar way to dissolved chemical contamination (i.e., per gram organism) may not be appropriate for assessing ecological endpoints. To measure an endpoint that is directly comparing contamination in an organism to that in its environment (i.e., assessing bioconcentration), the reporting of all matrices in comparable units is paramount (Hartmann et al., 2019; Rochman et al., 2019). Hence, standardising by weight is necessary. However, to determine endpoints associated with an organism's prey (i.e., assessing biomagnification through trophic transfer), understanding the numeric quantity of MPs per individual will establish whether this contamination is retained and increases in quantity over time or trophic level. Other complicating factors, such as growth correction (where organismal growth, influenced by experimental conditions or life stage, skews elimination rate constants), bio-dilution (where high food intake relative to the contaminant concentration masks the bioaccumulation signal) and variable chemical concentration gradients (an inherent challenge of particulate contamination), can influence BCF and BMF estimations (Adolfsson-Erici et al., 2012; Gobas and Lee, 2019; Mackay et al., 2016). For MPs, sorption dynamics and particle aging further complicate assessments by modifying contaminant bioavailability and uptake (Pan et al., 2022). These complexities underscore the urgent need for a consensus for how classical bioaccumulation frameworks should be adapted to heterogeneous particulate matter (e.g., MPs). Further research is essential to disentangle these influences and improve MP risk assessments.

4.4. Conclusions and recommendations

Through detailed quantification, this research offers pivotal insights into the ecological fate of PEST microfibres in the presence and absence of a co-contaminant, DEHP, through a simple marine food chain. The pervasive ingestion, variable retention and consistent trophic transfer of PEST microfibres across copepods, mysid shrimp and moon wrasse fish underscore the widespread ecological exposure to these contaminants. The nuanced role of DEHP impacting PEST ingestion in copepods, without influencing subsequent transfer points to complex contaminant interactions, and provides the foundation for future experiments using copepods, mysid shrimp and fish. Notably, while bioconcentration was universally observed, biomagnification factors revealed a species-specific pattern and highlights the requirement for standardised MP units to allow these endpoints to be employed. These findings are essential for advancing the understanding of MP fate within a tropical marine ecosystem.

To build upon the foundational knowledge provided here, the following recommendations for future research are provided:

1. Investigate the impacts of MP and additive contaminants on more complex food webs to assess broader, ecosystem-level risks, and evaluate whether similar accumulation dynamics persist under more ecologically realistic conditions.
2. Assess the impacts of long-term and chronic exposure of MPs and chemical additives on physiological (e.g., reproduction, energy metabolism, growth), behavioural (e.g., feeding, predator-prey interactions) and toxicological (e.g., oxidative stress, endocrine disruption, mortality) endpoints.
3. Expand investigation to include the wide range of physicochemical characteristics that comprise MPs, including various shapes, sizes, colours and chemical additives, using both environmentally realistic and future-looking exposure concentrations.
4. Establish robust bioaccumulation frameworks suited to MP risk assessments that integrates complex factors like diverse MP characteristics, chemical co-contaminants, food web complexity, and environmental variability to better discern the real-world ecological impacts of MP contamination.

CRediT authorship contribution statement

Michaela E. Miller: Writing – review & editing, Writing – original draft, Visualization, Validation, Resources, Project administration, Methodology, Investigation, Funding acquisition, Formal analysis, Data curation, Conceptualization. **Cherie A. Motti:** Writing – review & editing, Supervision, Project administration. **Vilde K. Snekkevik:** Writing – review & editing, Methodology, Investigation. **Keegan Vickers:** Writing – review & editing, Methodology, Investigation. **Hannah Kennedy:** Writing – review & editing, Methodology, Investigation. **Lee**

Bastin: Investigation. **Mark Hamann:** Writing – review & editing, Supervision, Project administration. **Frederieke J. Kroon:** Writing – review & editing, Supervision, Resources, Funding acquisition, Conceptualization.

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Declaration of competing interest

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

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Appendix A. Supplementary data

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Data availability

Data will be made available on request.

References

Adolfsson-Erici, M., Åkerman, G., McLachlan, M.S., 2012. Measuring bioconcentration factors in fish using exposure to multiple chemicals and internal benchmarking to correct for growth dilution. Environ. Toxicol. Chem. 31, 1853–1860.
Akhbarizadeh, R., Moore, F., Keshavarzi, B., 2019. Investigating microplastics bioaccumuation and biomagnification in seafood from the Persian Gulf: a threat to human health? Food Addit. Contam. Part A Chem. Anal. Control Expo. Risk Assess. 36, 1696–1708.
Alajmi, F., Zeng, C., 2015. Evaluation of microalgal diets for the intensive cultivation of the tropical calanoid copepod, *Parvocalanus crassirostris*. Aquacult. Res. 46, 1025–1038.
Aldegunde-Louzao, N., López, P.P., Aira, M.L., Latorre, C.H., 2023. Seven-year-long screening of phthalate esters in clothing and textile products from a quality control laboratory. Text. Res. J. 93, 1670–1685.
Alexander, D.E., 1999. Bioaccumulation, bioconcentration, biomagnification. In: Environmental Geology. Springer Netherlands, Dordrecht, pp. 43–44.
Amelia, T.S.M., Khalik, W.M.A.W.M., Ong, M.C., Shao, Y.T., Pan, H.-J., Bhubalan, K., 2021. Marine microplastics as vectors of major ocean pollutants and its hazards to the marine ecosystem and humans. Prog Earth Planet Sci 8, 1–26.
Andrady, A.L., 2011. Microplastics in the marine environment. Mar. Pollut. Bull. 62, 1596–1605.
Arthur, C., Bamford, H., Baker, J., 2008. The Occurrence, Effects and Fate of Small Plastic Debris in the Oceans, pp. 9–11.
Asher, E.C., Dacey, J.W.H., Jarniková, T., Tortell, P.D., 2015. Measurement of DMS, DMSO, and DMSP in natural waters by automated sequential chemical analysis. Limnol. Oceanogr.: Methods 13, 451–462.
Asher, E.C., Dacey, J.W.H., Stukel, M., Long, M.C., Tortell, P.D., 2017. Processes driving seasonal variability in DMS, DMSP, and DMSO concentrations and turnover in coastal Antarctic waters. Limnol. Oceanogr. 62, 104–124.
Athey, S.N., Albotra, S.D., Gordon, C.A., Monteleone, B., Seaton, P., Andrady, A.L., Taylor, A.R., Brander, S.M., 2020. Trophic transfer of microplastics in an estuarine food chain and the effects of a sorbed legacy pollutant. Limnol. Oceanogr. Lett. 5, 154–162.
Barbier, M., Loreau, M., 2019. Pyramids and cascades: a synthesis of food chain functioning and stability. Ecol. Lett. 22, 405–419.
Beek, B., Böhling, S., Bruckmann, U., Franke, C., Jöhncke, U., Studinger, G., 2000. The assessment of bioaccumulation. In: Bioaccumulation–New Aspects and Developments. Springer, pp. 235–276.
Bergé, A., Cladière, M., Gasperi, J., Coursimault, A., Tassin, B., Moilleron, R., 2013. Meta-analysis of environmental contamination by phthalates. Environ. Sci. Pollut. Res. 20, 8057–8076.
Borrelle, S.B., Ringma, J., Law, K.L., Monnahan, C.C., Lebreton, L., McGivern, A., Murphy, E., Jambeck, J., Leonard, G.H., Hilleary, M.A., 2020. Predicted growth in plastic waste exceeds efforts to mitigate plastic pollution. Science 369, 1515–1518.
Botterell, Z.L.R., Beaumont, N., Dorrington, T., Steinke, M., Thompson, R.C., Lindeque, P. K., 2019. Bioavailability and effects of microplastics on marine zooplankton: a review. Environ. Pollut. 245, 98–110.
Brayton, C.F., 1986. Dimethyl sulfoxide (DMSO): a review. Cornell Vet. 76, 61–90.
Briand, M.J., Bustamante, P., Bonnet, X., Churlaud, C., Letourneur, Y., 2018. Tracking trace elements into complex coral reef trophic networks. Sci. Total Environ. 612, 1091–1104.
Bucci, K., Tulio, M., Rochman, C.M., 2020. What is known and unknown about the effects of plastic pollution: a meta-analysis and systematic review. Ecol. Appl. 30, e02044.
Chan, H.S.H., Dingle, C., Not, C., 2019. Evidence for non-selective ingestion of microplastic in demersal fish. Mar. Pollut. Bull. 149, 110523.
Chormare, R., Kumar, M.A., 2022. Environmental health and risk assessment metrics with special mention to biotransfer, bioaccumulation and biomagnification of environmental pollutants. Chemosphere 302, 134836. https://doi.org/10.1016/j.chemosphere.2022.134836.
Chua, E.M., Shimeta, J., Nugegoda, D., Morrison, P.D., Clarke, B.O., 2014. Assimilation of polybrominated diphenyl ethers from microplastics by the marine amphipod, *Allorchestes compressa*. Environ. Sci. Technol. 48, 8127–8134.
Cole, M., Lindeque, P., Fileman, E., Halsband, C., Galloway, T.S., 2015. The impact of polystyrene microplastics on feeding, function and fecundity in the marine copepod *Calanus helgolandicus*. Environ. Sci. Technol. 49, 1130–1137.
Commission, E, 2011. Technical Support for the Impact Assessment of the Review of Priority Substances under Directive 2000/60/EC. Limited, E.U. (Ed.).
Covernton, G.A., Cox, K.D., Fleming, W.L., Buirs, B.M., Davies, H.L., Juanes, F., Dudas, S. E., Dower, J.F., 2022. Large size (>100-μm) microplastics are not biomagnifying in coastal marine food webs of British Columbia, Canada. Ecol. Applic. 32, e2654.
Cowger, W., Booth, A.M., Hamilton, B.M., Thaysen, C., Primpke, S., Munno, K., Lusher, A.L., Dehaut, A., Vaz, V.P., Liboiron, M., 2020. Reporting guidelines to increase the reproducibility and comparability of research on microplastics. Appl. Spectrosc. 74, 1066–1077.
Critchell, K., Hoogenboom, M.O., 2018. Effects of microplastic exposure on the body condition and behaviour of planktivorous reef fish (*Acanthochromis polyacanthus*). PLoS One 13, 19.
Dawson, A.L., Kawaguchi, S., King, C.K., Townsend, K.A., King, R., Huston, W.M., Nash, S.M.B., 2018. Turning microplastics into nanoplastics through digestive fragmentation by Antarctic krill. Nat. Commun. 9, 8.
Domínguez-López, M., Bellas, J., Sánchez-Ruiloba, L., Planas, M., Hernández-Urcera, J., 2022. First evidence of ingestion and retention of microplastics in seahorses (*Hippocampus reidi*) using copepods (*Acartia tonsa*) as transfer vectors. Sci. Total Environ. 818, 151688.
Dool, T., Bosker, T., 2022. Predicted microplastic uptake through trophic transfer by the short-beaked common dolphin (*Delphinus delphis*) and common bottlenose dolphin (*Tursiops truncatus*) in the Northeast Atlantic Ocean and Mediterranean Sea. Mar. Pollut. Bull. 180, 113745.
Elizalde-Velázquez, A., Carcano, A.M., Crago, J., Green, M.J., Shah, S.A., Cañas-Carrell, J.E., 2020. Translocation, trophic transfer, accumulation and depuration of polystyrene microplastics in *Daphnia magna* and *Pimephales promelas*. Environ. Pollut. 259, 113937.
Exchange, T., 2022. Preferred Fiber And Materials Market Report 2022.
Farrell, P., Nelson, K., 2013. Trophic level transfer of microplastic: *Mytilus edulis* (L.) to *Carcinus maenas* (L.). Environ. Pollut. 177, 1–3.
Feijtel, T., Kloepper-Sams, P., Den Haan, K., Van Egmond, R., Comber, M., Heusel, R., Wierich, P., Ten Berge, W., Gard, A., De Wolf, W., 1997. Integration of bioaccumulation in an environmental risk assessment. Chemosphere 34, 2337–2350.
Foley, C.J., Feiner, Z.S., Malinich, T.D., Hook, T.O., 2018. A meta-analysis of the effects of exposure to microplastics on fish and aquatic invertebrates. Sci. Total Environ. 631-632, 550–559.
Fred-Ahmadu, O.H., Bhagwat, G., Oluyoye, I., Benson, N.U., Ayejuyo, O.O., Palanisami, T., 2020. Interaction of chemical contaminants with microplastics: principles and perspectives. Sci. Total Environ. 706, 135978.
Friesen, J.A., Mann, K.H., Willison, J.H.M., 1986. Gross anatomy and fine structure of the gut of the marine mysid shrimp *Mysis stenolepis* Smith. Can. J. Zool. 64, 431–441.

Gago, J., Carretero, O., Filgueiras, A.V., Viñas, L., 2018. Synthetic microfibers in the marine environment: a review on their occurrence in seawater and sediments. Mar. Pollut. Bull. 127, 365–376.
Gamarra-Toledo, V., Plaza, P.I., Peña, Y.A., Bermejo, P.A., López, J., Cano, G.L., Barreto, S., Cáceres-Medina, S., Lambertucci, S.A., 2023. High incidence of plastic debris in Andean condors from remote areas: evidence for marine-terrestrial trophic transfer. Environ. Pollut. 317, 120742.
Geyer, R., Jambeck, J.R., Law, K.L., 2017. Production, use, and fate of all plastics ever made. Sci. Adv. 3 (5).
Gobas, F.A., Lee, Y.S., 2019. Growth-correcting the bioconcentration factor and biomagnification factor in bioaccumulation assessments. Environ. Toxicol. Chem. 38, 2065–2072.
Greenshields, J., Schirrmacher, P., Hardege, J.D., 2021. Plastic additive oleamide elicits hyperactivity in hermit crabs. Mar. Pollut. Bull. 169, 112533.
Gulizia, A.M., Patel, K., Philippa, B., Motti, C.A., van Herwerden, L., Vamvounis, G., 2023. Understanding plasticiser leaching from polystyrene microplastics. Sci. Total Environ. 857, 159099.
Guo, J.-J., Huang, X.-P., Xiang, L., Wang, Y.-Z., Li, Y.-W., Li, H., Cai, Q.-Y., Mo, C.-H., Wong, M.-H., 2020. Source, migration and toxicology of microplastics in soil. Environ. Int. 137, 105263.
Hahladakis, J.N., Velis, C.A., Weber, R., Iacovidou, E., Purnell, P., 2018. An overview of chemical additives present in plastics: migration, release, fate and environmental impact during their use, disposal and recycling. J. Hazard. Mater. 344, 179–199.
Hartmann, N.B., Huffer, T., Thompson, R.C., Hassellov, M., Verschoor, A., Daugaard, A. E., Rist, S., Karlsson, T., Brennholt, N., Cole, M., Herrling, M.P., Hess, M.C., Ivleva, N. P., Lusher, A.L., Wagner, M., 2019. Are we speaking the same language? Recommendations for a definition and categorization framework for plastic debris. Environ. Sci. Technol. 53, 1039–1047.
Hasegawa, T., Nakaoka, M., 2021. Trophic transfer of microplastics from mysids to fish greatly exceeds direct ingestion from the water column. Environ. Pollut. 273, 116468.
Heindler, F.M., Alajmi, F., Huerlimann, R., Zeng, C., Newman, S.J., Vamvounis, G., van Herwerden, L., 2017. Toxic effects of polyethylene terephthalate microparticles and di(2-ethylhexyl)phthalate on the calanoid copepod, parvocalanus crassirostris. Ecotoxicol. Environ. Saf. 141, 298–305.
Holmes, T.H., McCormick, M.I., 2006. Location influences size-selective predation on newly settled reef fish. Mar. Ecol. Prog. Ser. 317, 203–209.
Holmes, T.H., Wilson, S.K., Vanderklift, M., Babcock, R., Fraser, M., 2012. The role of *Thalassoma lunare* as a predator of juvenile fish on a sub-tropical coral reef. Coral Reefs 31, 1113–1123.
Jensen, L.H., Motti, C.A., Garm, A.L., Tonin, H., Kroon, F.J., 2019. Sources, distribution and fate of microfibres on the Great Barrier Reef, Australia. Sci. Rep. 9, 1–15.
Justino, A.K., Ferreira, G.V., Fauvelle, V., Schmidt, N., Lenoble, V., Pelage, L., Martins, K., Travassos, P., Lucena-Frédou, F., 2023. From prey to predators: evidence of microplastic trophic transfer in tuna and large pelagic species in the southwestern tropical Atlantic. Environ. Pollut. 327, 121532.
Kamrin, M.A., 2009. Phthalate risks, phthalate regulation, and public health: a review. J. Toxic. Environ. Health, Part B 12, 157–174.
Kershaw, P., Rochman, C., 2015. Sources, fate and effects of microplastics in the marine environment: part 2 of a global assessment. Reports and studies-IMO/FAO/Unesco-IOC/WMO/IAEA/UN/UNEP joint Group of Experts on the scientific aspects of marine environmental protection (GESAMP) eng no. 93.
Kramer, M.J., Bellwood, O., Fulton, C.J., Bellwood, D.R., 2015. Refining the invertivore: diversity and specialisation in fish predation on coral reef crustaceans. Mar. Biol. 162, 1779–1786.
Kroon, F.J., Motti, C.E., Jensen, L.H., Berry, K.L.E., 2018. Classification of marine microdebris: a review and case study on fish from the Great Barrier Reef, Australia. Sci. Rep. 8, 1–15.
Lau, W.W.Y., Shiran, Y., Bailey, R.M., Cook, E., Stuchtey, M.R., Koskella, J., Velis, C.A., Godfrey, L., Boucher, J., Murphy, M.B., 2020. Evaluating scenarios toward zero plastic pollution. Science 369, 1455–1461.
Lee, D.-H., Lee, S., Rhee, J.-S., 2021. Consistent exposure to microplastics induces age-specific physiological and biochemical changes in a marine mysid. Mar. Pollut. Bull. 162, 111850.
Liang, D.-W., Zhang, T., Fang, H.H.P., He, J., 2008. Phthalates biodegradation in the environment. Appl. Microbiol. Biotechnol. 80, 183–198.
Lusher, A., 2015. Microplastics in the Marine Environment: Distribution, Interactions and Effects, Marine Anthropogenic Litter. Springer, Cham, pp. 245–307.
Mackay, D., Celsie, A.K., Arnot, J.A., Powell, D.E., 2016. Processes influencing chemical biomagnification and trophic magnification factors in aquatic ecosystems: implications for chemical hazard and risk assessment. Chemosphere 154, 99–108.
Miller, M.E., Hamann, M., Kroon, F.J., 2020. Bioaccumulation and biomagnification of microplastics in marine organisms: a review and meta-analysis of current data. PLoS One 15, e0240792.
Miller, M.E., Santana, M.F.M., Carsique, M., Motti, C.A., Hamann, M., Kroon, F.J., 2022. Temporal patterns of plastic contamination in surface waters at the SS Yongala shipwreck, Great Barrier Reef, Australia. Environ. Pollut. 307, 119545. https://doi.org/10.1016/j.envpol.2022.119545.
Miller, M.E., Motti, C.A., Hamann, M., Kroon, F.J., 2023. Assessment of microplastic bioconcentration, bioaccumulation and biomagnification in a simple coral reef food web. Sci. Total Environ. 858, 159615.
Nelms, S.E., Galloway, T.S., Godley, B.J., Jarvis, D.S., Lindeque, P.K., 2018. Investigating microplastic trophic transfer in marine top predators. Environ. Pollut. 238, 999–1007.
Net, S., Sempere, R., Delmont, A., Paluselli, A., Ouddane, B., 2015. Occurrence, fate, behavior and ecotoxicological state of phthalates in different environmental matrices. Environ. Sci. Technol. 49, 4019–4035.
Oliveira, A.F., Marques, S.C., Pereira, J.L., Azeiteiro, U.M., 2023. A review of the order mysida in marine ecosystems: what we know what is yet to be known. Mar. Environ. 188, 106019. https://doi.org/10.1016/j.marenvres.2023.106019.
Osman, A.I., Hosny, M., Eltaweil, A.S., Omar, S., Elgarahy, A.M., Farghali, M., Yap, P.-S., Wu, Y.-S., Nagandran, S., Batumalaie, K., 2023. Microplastic sources, formation, toxicity and remediation: a review. Environ. Chem. Lett. 21, 2129–2169.
Pan, K., Chen, C.C., Lin, L., Xu, H., Chen, F., Li, Y., Zhu, X., Ma, J., Lan, W., 2022. Adsorption of di (2-ethylhexyl) phthalate (DEHP) to microplastics in seawater: a comparison between pristine and aged particles. Bull. Environ. Contam. Toxicol. 109, 776–782.
Procter, J., Hopkins, F.E., Fileman, E.S., Lindeque, P.K., 2019. Smells good enough to eat: dimethyl sulfide (DMS) enhances copepod ingestion of microplastics. Mar. Pollut. Bull. 138, 1–6.
Rivers, M.L., Gwinnett, C., Woodall, L.C., 2019. Quantification is more than counting: actions required to accurately quantify and report isolated marine microplastics. Mar. Pollut. Bull. 139, 100–104.
Rochman, C.M., Brookson, C., Bikker, J., Djuric, N., Earn, A., Bucci, K., Athey, S., Huntington, A., McIlwraith, H., Munno, K., De Frond, H., Kolomijeca, A., Erdle, L., Grbic, J., Bayoumi, M., Borrelle, S.B., Wu, T.N., Santoro, S., Werbowski, L.M., Zhu, X., Giles, R.K., Hamilton, B.M., Thaysen, C., Kaura, A., Klasios, N., Ead, L., Kim, J., Sherlock, C., Ho, A., Hung, C., 2019. Rethinking microplastics as a diverse contaminant suite. Environ. Toxicol. Chem. 38, 703–711.
Rowdhwal, S.S.S., Chen, J., 2018. Toxic effects of di-2-ethylhexyl phthalate: an overview. In: BioMed Research International 2018.
Ryan, M.G., Watkins, L., Walter, M.T., 2019. Hudson River juvenile Blueback herring avoid ingesting microplastics. Mar. Pollut. Bull. 146, 935–939.
Saikumar, S., Mani, R., Ganesan, M., Dhinakarasamy, I., Palanisami, T., Gopal, D., 2024. Trophic transfer and their impact of microplastics on estuarine food chain model. J. Hazard. Mater. 464, 132927.
Santana, M.F.M., 2022. Presence, Abundance and Effects of Microplastics on the Great Barrier Reef. James Cook University, College of Science and Engineering.
Santana, M.F.M., Dawson, A.L., Motti, C.A., Van Herwerden, L., Lefevre, C., Kroon, F.J., 2021. Ingestion and depuration of microplastics by a planktivorous coral reef fish, *Pomacentrus amboinensis*. Front. Environ. Sci. 9, 641135.
Savoca, M.S., Wohlfeil, M.E., Ebeler, S.E., Nevitt, G.A., 2016. Marine plastic debris emits a keystone infochemical for olfactory foraging seabirds. Sci. Adv. 2 (8).
Savoca, M.S., Tyson, C.W., McGill, M., Slager, C.J., 2017. Odours from marine plastic debris induce food search behaviours in a forage fish. Proc. R. Soc. B Biol. Sci. 284, 20171000.
Schlawinsky, M., Santana, M.F.M., Motti, C.A., Martins, A.B., Thomas-Hall, P., Miller, M. E., Lefèvre, C., Kroon, F.J., 2022. Improved microplastic processing from complex biological samples using a customized vacuum filtration apparatus. Limnol. Oceanogr. Methods 20, 553–567.
Seo, J.S., Park, T.J., Lee, Y.M., Park, H.G., Yoon, Y.D., Lee, J.S., 2006. Small heat shock protein 20 gene (Hsp20) of the intertidal copepod *Tigriopus japonicus* as a possible biomarker for exposure to endocrine disruptors. Bull. Environ. Contam. Toxicol. 76.
Setala, O., Fleming-Lehtinen, V., Lehtiniemi, M., 2014. Ingestion and transfer of microplastics in the planktonic food web. Environ. Pollut. 185, 77–83.
Suaria, G., Achtypi, A., Perold, V., Lee, J.R., Pierucci, A., Bornman, T.G., Aliani, S., Ryan, P.G., 2020. Microfibers in oceanic surface waters: a global characterization. Sci. Adv. 6, eaay8493.
Thornton, J.W., McCally, M., Houlihan, J., 2002. Biomonitoring of industrial pollutants: health and policy implications of the chemical body burden. Public Health Rep. 117, 315.
Turner, C., Sawle, A., Fenske, M., Cossins, A., 2012. Implications of the solvent vehicles dimethylformamide and dimethylsulfoxide for establishing transcriptomic endpoints in the zebrafish embryo toxicity test. Environ. Toxicol. Chem. 31, 593–604.
USEPA, 1997. Terms of Environment: Glossary, Abbreviations, and Acronyms. Agency, U.S.E.P. (Ed.).
USEPA, 2008. White Paper on Methods for Assessing Ecological Risks of Pesticides with Persistent, Bioaccumulative and Toxic Characteristics. Office of Prevention, P.A.T.S., environmental fate and effects division (Ed.).
USEPA, 2019. Proposed Designation of Di-Ethylhexyl Phthalate (DEHP) (1,2-Benzene-Dicarboxylic Acid, 1,2-Bis(2-Ethylhexyl) Ester) (CASRN 117-81-7) as a High-Priority Substance for Risk Evaluation. Prevention, O.O.C.S.A.P. (Ed.).
Verslycke, T., Ghekiere, A., Raimondo, S., Janssen, C., 2007. Mysid crustaceans as standard models for the screening and testing of endocrine-disrupting chemicals. Ecotoxicology 16, 205–219.
Viitasalo, S., Viitasalo, M., 2004. Predation by the mysid shrimps Mysis mixta and M. relicta on benthic eggs of *Bosmina longispina maritima* (Cladocera) in the northern Baltic Sea. Mar. Ecol. Prog. Ser. 281, 155–163.
Welden, N.A.C., Cowie, P.R., 2016. Environment and gut morphology influence microplastic retention in langoustine, *Nephrops norvegicus*. Environ. Pollut. 214, 859–865.
Welden, N.A., Abylkhani, B., Howarth, L.M., 2018. The effects of trophic transfer and environmental factors on microplastic uptake by plaice, Pleuronectes plastessa, and spider crab, Maja squinado. Environ. Pollut. 239, 351–358.
Xu, X., Fang, J.K.-H., Wong, C.-Y., Cheung, S.-G., 2022. The significance of trophic transfer in the uptake of microplastics by carnivorous gastropod *Reishia clavigera*. Environ. Pollut. 298, 118862.
Yadav, D.K., Samantaray, B.P., Kumar, R., 2024. Effect of alternative natural diet on microplastic ingestion, functional responses and trophic transfer in a tri-trophic coastal pelagic food web. Sci. Total Environ. 950, 174999.

Ye, X., Wang, P., Wu, Y., Zhou, Y., Sheng, Y., Lao, K., 2020. Microplastic acts as a vector for contaminants: the release behavior of dibutyl phthalate from polyvinyl chloride pipe fragments in water phase. Environ. Sci. Pollut. Res. 27, 42082–42091.
Yu, Q., Nederstigt, T.A.P., Wang, Z., Wu, J., Bosker, T., Peijnenburg, W.J.G.M., Vijver, M. G., 2024. Accumulation kinetics of polystyrene nano- and microplastics in the waterflea *Daphnia magna* and trophic transfer to the mysid *Limnomysis benedeni*. Environ. Pollut. 363, 125029.
Yuan, L., Liu, J., Huang, Y., Shen, G., Pang, S., Wang, C., Li, Y., Mu, X., 2022. Integrated toxicity assessment of DEHP and DBP toward aquatic ecosystem based on multiple trophic model assays. Environ. Sci. Pollut. Res. 29, 87402–87412.
Zhu, F.X., Zhu, C.Y., Wang, C., Gu, C., 2019. Occurrence and ecological impacts of microplastics in soil systems: a review. Bull. Environ. Contam. Toxicol. 102, 741–749.