



Microplastic ingestion in fish larvae in the western English Channel[☆]



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ABSTRACT

Microplastics have been documented in marine environments worldwide, where they pose a potential risk to biota. Environmental interactions between microplastics and lower trophic organisms are poorly understood. Coastal shelf seas are rich in productivity but also experience high levels of microplastic pollution. In these habitats, fish have an important ecological and economic role. In their early life stages, planktonic fish larvae are vulnerable to pollution, environmental stress and predation. Here we assess the occurrence of microplastic ingestion in wild fish larvae. Fish larvae and water samples were taken across three sites (10, 19 and 35 km from shore) in the western English Channel from April to June 2016. We identified 2.9% of fish larvae ($n = 347$) had ingested microplastics, of which 66% were blue fibres; ingested microfibrils closely resembled those identified within water samples. With distance from the coast, larval fish density increased significantly ($P < 0.05$), while waterborne microplastic concentrations ($P < 0.01$) and incidence of ingestion decreased. This study provides baseline ecological data illustrating the correlation between waterborne microplastics and the incidence of ingestion in fish larvae.

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1. Introduction

Microplastic (microscopic plastic, 0.1 μm –5 mm) debris has emerged as a persistent environmental pollutant, recognised within the scientific and political community as a ubiquitous contaminant of global concern (Thompson et al., 2004). The increasing abundance and widespread distribution of microplastics has led to concerns over the risks posed to the health of organisms and ecosystem processes (Clark et al., 2016). Since the emergence of mass-produced plastics in the 1930s (BPF, 2017), production has increased annually, currently reaching in excess of 322 million tonnes per year globally (Plastics, 2016). Its durability, low cost and widespread application has made plastic a popular manufacturing material worldwide (Cole et al., 2011). These same characteristics make it difficult to dispose of, and once in the environment could be considered a persistent and potentially hazardous pollutant (Rochman et al., 2013a). Marine plastic debris stems from poor waste management and accidental losses from fishing, industry, shipping and tourism among other sources (Jambeck et al., 2015).

Microplastic pollution originates from the photooxidative degradation and subsequent fragmentation of this larger debris (Jambeck et al., 2015), termed secondary microplastics, and the release of plastics manufactured to be of a microscopic size, such as exfoliates in cosmetics (Napper et al., 2015), termed primary microplastics. Microplastics in marine waters were first documented over forty years ago in the North Atlantic subtropical gyre (Carpenter et al., 1972). Microplastics have since been found in a diverse range of marine ecosystems, including deep ocean sediments (Van Cauwenberghe et al., 2013) and Arctic waters (Lusher et al., 2015). Recent estimates suggest over 5.25 trillion items of floating plastic litter are polluting the world's oceans, of which the vast majority are microscopic in size (Eriksen et al., 2014).

Microplastic pollution poses a threat to marine biota through ingestion or entanglement (Wright et al., 2013b). Continuous fragmentation and degradation of microplastics in the marine environment produces a wide range of particle sizes (Enders et al., 2015), which can be ingested by an equally wide range of marine organisms, including the Humboldt squid (Braid et al., 2012), blue mussel and Pacific oyster (Van Cauwenberghe and Janssen, 2014), gooseneck barnacle (Goldstein and Goodwin, 2013), Norway lobster (Murray and Cowie, 2011), brown shrimp (Devriese et al., 2015), zooplankton (Desforges et al., 2015), harbour seal (Rebolledo et al., 2013) and green turtle (Tourinho et al., 2010). The

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overlap between microplastics and marine biota is predicted to be most pronounced in shelf sea regions (Clark et al., 2016), owing to high levels of biological productivity and high microplastic concentrations stemming from the proximity to terrestrial sources of pollution (e.g. rivers, estuaries, sewage outfalls) (Browne et al., 2011; Desforges et al., 2014).

Zooplankton encompass a diverse group of planktonic animals, including the larval stages of vertebrates and invertebrates. Marine zooplankton predominantly inhabit surface waters when feeding, where microplastics are found in high abundance (Cozar et al., 2014), increasing the opportunity for them to ingest microplastics. Under laboratory conditions, zooplankton (e.g. copepods, urchin larvae, bivalve larvae, decapod larvae) have been observed to readily consume microplastics (Cole et al., 2013, 2015; Cole and Galloway, 2015; Cole et al., 2015; Nobre et al., 2015; Setala et al., 2014; Lee et al., 2013; Kaposi et al., 2014). Toxicity testing has highlighted the adverse physical (Wright et al., 2013a) and toxicological effects that microplastic exposure can have on marine biota (Ogonowski et al., 2016; Peda et al., 2016; Watts et al., 2016; Cole et al., 2015). Experiments using marine worms and zooplankton have demonstrated that microplastic ingestion can result in reduced feeding, increased mortality, decreased growth rates, decreased hatching success and reduced fecundity (Wright et al., 2013a; Cole et al., 2015). Marine zooplankton are a vital source of food for secondary consumers (e.g. fish, cetaceans), and, as such, may represent a route via which microplastics enter the food web, posing a risk to secondary producers, apex predators and potentially human health (Clark et al., 2016). Field observations detailing incidence of microplastic ingestion by organisms typically relate to larger organisms (e.g. squid, mussels, oysters, adult fish), owing to the constraints associated with collecting and processing samples (Lusher et al., 2017). Research by Desforges et al. (2014) on zooplankton communities in the North East Pacific has shown microplastic ingestion ratios of 1 in 17 copepods (*Neocalanus cristatus*), and 1 in 34 euphausiids (*Euphausia pacifica*), of which 50–68% were fibres. Microplastics have been further identified in zooplankton communities sampled from the South China Sea, with 70% of identified plastics being fibrous (Sun et al., 2016). Otherwise, very little is known about ingestion rates of microplastics in wild zooplankton and the type, source and distribution of plastic being ingested.

Fish stocks have considerable ecological and economic value. Global annual fisheries revenue fluctuates around USD 100 billion supporting about 12% of the world population, and providing 2.9 billion people with 20% of their animal protein (Lam et al., 2016). With over 30,000 species of fish worldwide, existing in all of the world's marine habitats, their abundance and diversity has significant ecological importance for the food chain, nutrient cycling and ecosystem services (Worm et al., 2006). Ichthyoplanktonic studies show that un-fished taxa account for the majority of fish larvae and contribute significantly to trophic food webs (Baran, 2002). Fish populations are vulnerable to a growing number of anthropogenic pressures, including overfishing, climate change and pollution, resulting in increased mortality and reduced fecundity. Incidence of microplastic consumption by adult fish has been widely reported for pelagic and demersal populations across the globe, including blue whiting (*Micromesistius poutassou*), red gurnard (*Aspitrigla cuculus*), john dory (*Zeus faber*) and dragonet (*Callionymus lyra*) (Lusher et al., 2013). However, there is currently no substantial published data regarding microplastic ingestion rates in fish larvae. Fish larvae play a pivotal role in marine food webs (Russell, 1976), and their health, development and survival is fundamental to the long-term sustainability of healthy fish populations. As such, data is urgently required to better assess the risks posed to fish larvae by microplastics *in natura*.

In this study we investigate the incidence of microplastic ingestion by fish larvae in the productive shelf-sea waters of the western English Channel, off the coast of Plymouth (UK). We look to test the hypotheses that: (1) microplastic concentrations increase with proximity to the coast; (2) fish larvae consume microplastic debris in their natural environment; and, (3) incidence of microplastic consumption is regulated by the abundance of larvae and the abundance of microplastics. Fish larvae and microplastics were collected via oblique tows, across three sites with varying distance from shore; microplastics were isolated using dissection and enzymatic digestion of samples.

2. Methodology

2.1. Field sampling

Field sampling was undertaken on board RV Plymouth Quest in the western English Channel off the coast of Plymouth (UK). Sampling was conducted at stations L4, L5 and E1 (10 km, 19 km and 35 km from shore respectively), which are routinely sampled as part of the Western Channel Observatory (WCO; www.westernchannelobservatory.org.uk). The sampling sites spanned distances of 10–35 km from the city of Plymouth (Fig. 1), accounting for habitats with a coastal (L4) and oceanic influence (E1); L5 was added as a reference site because it is a rocky reef known to be a favourable habitat for fish larvae. Eleven samples were collected between 11th April 2016 and 21st June 2016 across the three sites (L4, $n = 5$; L5, $n = 3$; E1, $n = 3$). For each trawl, tow distance and maximal sample depths were recorded using GPS and a Suunto vyper dive computer respectively; maximum depths reached were on average 50 m at L4 and L5, and 65 m at E1. Fish larvae were collected using a 500 μm metal-framed net (1 m^2 square aperture) towed for 20 min on an oblique tow. Following the trawl, larvae were passed through a 500 μm sieve and rinsed with filtered (0.22 μm) natural seawater. Subsequently, specimens were transferred into a 1 L Nalgene bottle and preserved in 4% formalin. Microplastics were sampled using a 100 μm WP2 net (47 cm diameter aperture), suspended below the net used for sampling the fish larvae. This concurrent sampling allowed for direct comparison of microplastics ingested by the fish larvae with 'prey-sized' microplastics in the surrounding water. Following sampling, the WP2 net was rinsed with filtered seawater and the sample poured through a 100 μm mesh; samples were immediately sealed and subsequently stored in a foil envelope in a $-80\text{ }^\circ\text{C}$ freezer prior to analysis. Control measures included collection of procedural blanks using filtered sea water, and sampling of boat paint for Fourier Transform Infrared Spectroscopy (FT-IR) analysis to ensure false positives were avoided in the plastics count.

2.2. Fish larvae

Fish larvae were isolated by screening the formalin preserved net samples through a 2000 μm sieve. Specimens were rinsed thoroughly, and the 2000 μm sieve placed in a tray of water to float the sample inside the sieve. Fish larvae >10 mm were handpicked and placed into a covered beaker containing ultrapure water. The total number of fish larvae per sample was recorded, and fish larvae density (individuals m^{-3}) calculated using the net dimensions, tow length and depth, and a net efficiency of 85% (Southward, 1970). All fish larvae larger than 9 mm were identified to species level.

2.3. Microplastic ingestion in marine fish larvae

Fish larvae were assessed under a dissection microscope (Wild Heerbrugg Switzerland M5-49361; 6x-50x magnification) with

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