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Detection of low numbers of microplastics in North Sea fish using strict quality assurance criteria

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ABSTRACT

We investigated 400 individual fish of four North Sea species: Atlantic Herring, Sprat, Common Dab, and Whiting on ingestion of > 20 µm microplastic. Strict quality assurance criteria were followed in order to control contamination during the study. Two plastic particles were found in only 1 (a Sprat) out of 400 individuals (0.25%, with a 95% confidence interval of 0.09–1.1%). The particles were identified to consist of polymethylmethacrylate (PMMA) through FTIR spectroscopy. No contamination occurred during the study, showing the method applied to be suitable for microplastic ingestion studies in biota. We discuss the low particle count for North Sea fish with those in other studies and suggest a relation between reported particle count and degree of quality assurance applied. Microplastic ingestion by fish may be less common than thought initially, with low incidence shown in this study, and other studies adhering to strict quality assurance criteria.

1. Introduction

Plastic is one of the most used materials in the world nowadays (PlasticsEurope, 2015). As a consequence, plastic has been entering the marine environment in large quantities over the past decades, quantities that have been steadily increasing over the years (Copello and Quintana, 2003; Ribic et al., 2010; Wright et al., 2013). Nowadays, plastic is one of the most common and persistent pollutants in the oceans (Cole et al., 2011). Plastics constitute 60–80% of marine litter, reaching 90–95% in some areas (Moore, 2008). Plastic litter has been shown to have significant harmful effects on marine species both under laboratory conditions and in the field (Gall and Thompson, 2015; Derraik, 2002; Laist, 1987), and has been recognized as a global issue requiring immediate action (UNEP, 2014).

The occurrence of < 5 mm microplastic ingestion by marine organisms has been shown in a number of different studies (e.g., Boerger et al., 2010; Lusher et al., 2013; Van Cauwenberghe and Janssen, 2014). In laboratory experiments different planktonic organisms, polychaetes, bivalves, echinoderms, corals, and decapods have been shown to take up microplastic during feeding (Murray and Cowie, 2011; von Moos et al., 2012; Besseling et al., 2013; Cole et al., 2013; Hall et al., 2015; Setälä et al., 2016). These studies, however, often used unrealistically high concentrations of microplastics in their experiments (Phuong et al., 2016). Microplastic ingestion in fish and bivalves under field conditions has been observed in a range of studies as well

(Foekema et al., 2013; Van Cauwenberghe and Janssen, 2014; Desforges et al., 2015; Devriese et al., 2015; Cannon et al., 2016; Rummel et al., 2016; Nadal et al., 2016). The ingestion of microplastics raises several concerns; microplastic particles are thought to be able to evoke a biological response through both physical and chemical mechanisms.

First, physical impacts for small organisms like internal abrasions and blockages have been reported (Wright et al., 2013). Moreover, microplastic particles were shown to cause damage leading to cellular necrosis, inflammation, and lacerations of tissues in gastrointestinal tracts (Rochman et al., 2016).

Second, plastic's durability makes it a vector for the transport of non-indigenous species over longer distances than was possible before (Goldstein et al., 2014). Plastics may serve as alternative rafting substratum for the same organisms usually transported (Thiel and Haye, 2006), and many of the species found are known to be prolific and successful rafters (Goldstein et al., 2014). Colonization by invasive species is viewed as one of the greatest threats to global biodiversity (Barnes, 2002), which makes this rafting a reason for concern.

Third, persistent organic pollutants (POPs) can concentrate on the plastic particles (Endo and Koelmans, 2016) and it has been suggested this could pose a possible new route for POPs to enter the food chain (Teuten et al., 2009). However, it has not been convincingly shown that this actually happens under natural conditions (Koelmans, 2015; Koelmans et al., 2013, 2016; Bakir et al., 2016; Herzke et al., 2016).

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Contrarily, evidence for Northern Fulmars (*Fulmarus glacialis*) suggests a transfer of POPs from the lipids in the animal to the plastic, rather than the other way around (Herzke et al., 2016).

This leads to the fourth concern: human food-safety (Van Cauwenberghe and Janssen, 2014; Miranda and Carvalho-souza, 2016; Neves et al., 2015; Wright and Kelly, 2017). Laboratory studies have observed trophic transfer of microplastic particles between groups of zooplankton (Setälä et al., 2014), and mussels and crabs (Farrell and Nelson, 2013). Combined with the potential of plastic particles to translocate from the gut to the circulatory system in mussels (Browne et al., 2008), this has caused concerns with regard to human food safety. Measuring ingestion rates of microplastic in organisms can improve the understanding of the hazards to humans. Whether microplastic particles travel up the food chain and end up in humans, and if the microplastics or adhered POPs cause any adverse effect on human health, remains an open question.

Finally, despite the concern regarding accumulation of floating plastic debris in the open ocean, the extent and mechanisms of this accumulation are still largely unknown. A study by Cozar et al. (2014) reported a gap in the size distribution of floating plastic debris, and the global surface load of plastic was observed to be below what would be expected from production and input rates. These findings support the hypothesis of substantial losses of plastic from the ocean surface, and indicate the existence of four possible sinks: shore deposition, nano-fragmentation, biofouling causing submersion (Kooi et al., 2017), and ingestion (Cozar et al., 2014). The predominant size of plastic debris where global losses occur matches the most frequent plastic size ingested by fish in ingestion studies, marking the importance of ingestion studies as a valuable source of information for global inventory of plastic debris in the world's oceans.

Notwithstanding these concerns, studies examining the occurrence of microplastic in natural populations are relatively few. This field of research is fairly young, and a standardized method to detect and quantify microplastics has not been established yet. This has led to an array of studies performed with a variety of methods, resulting in largely incomparable data between studies. Often, studies found relatively large numbers of fibres (Lusher et al., 2013; Rummel et al., 2016; Nadal et al., 2016; Desforges et al., 2015; Devriese et al., 2015; Neves et al., 2015; Remy et al., 2015; Mathalon and Hill, 2014; Anastasopoulou et al., 2013; Foekema et al., 2013; Claessens et al., 2011; Davison and Asch, 2011), leading to the understanding that microfibrils are likely to be the most abundant shape of microplastic in the marine environment (Wright et al., 2013). However, the majority of these studies did not take into account the possibility of (airborne) contamination of microfibrils during the performance of the study, or did not take sufficient precautions to rule it out completely (Torre et al., 2016). Foekema et al. (2013) initially detected small fibres, but the abundance sharply decreased when working under clean air conditions. Additionally, studies performing polymer identification are rare; often visual identification is the only identification method applied. This underlines the need for a proper protocol for sampling, extraction, and identification of microplastics in biota, while mitigating airborne contamination (Wesch et al., 2016a,b; Vandermeersch et al., 2015).

Currently only the seabird Northern Fulmar (*Fulmarus glacialis*) is subjected to regular monitoring of plastic ingestion (Van Franeker et al., 2011). By expanding such monitoring with some pelagic, demersal, and benthic species a vertical assessment of the water column could be achieved. Fish can serve as a good indicator for the health of the foodweb, as they move freely through the water column and are able to cover fairly large distances, increasing their chance of encounters with microplastic particles. Additionally, in light of the human food safety concern, assessment of commercially interesting species is highly relevant.

The aim of this study was to show whether the occurrence of microplastic ingestion in North Sea fish is common, following strict quality assurance criteria in order to minimize and control

contamination during the study. The quality assurance applied in this study was stricter than that of recent studies, and involved a level of contamination mitigation that has not been seen in earlier studies, taking into account several different factors influencing contamination of samples. A secondary aim was to study if the frequency and quantity of microplastic found in the gastrointestinal tracts of the fish could be linked to species characteristics, such as feeding behaviour. To this end, a total of 400 individual fish of four different commercially valuable species from the North Sea was examined. The species covered were: *Clupea harengus* (Atlantic Herring), *Sprattus sprattus* (Sprat), *Limanda limanda* (Common Dab), and *Merlangius merlangus* (Whiting, or Merling). Common Dab is a species of right-eyed flounder, a demersal species. Both Atlantic Herring and Whiting are benthopelagic, Sprat is a pelagic-neritic species (FishBase). For all fish, size was recorded. Plastic was isolated from the digestive tract, and analysed with FTIR spectroscopy for polymer identification. Finally, our results are discussed with respect to other studies also in the light of quality assurance protocols applied.

2. Materials and methods

2.1. Sampling

The fish was caught in late January 2013, during the yearly International Bottom Trawl Survey (IBTS) performed by research institute IMARES in the Netherlands. A GOV (Grande Ouverture Verticale) gear was used. The cod-end was fitted with a mesh size of 20 mm so that the majority of young fish caught were retained. The area the fish for this study was caught, is the Southern Bight; an area making up the southern North Sea, bounded by the coasts of the Netherlands, Belgium, France and Great Britain (Fig. 1). The trawling was performed during daytime, between 15 min before sunrise until 15 min after sunset, and started from two locations (Fig. 1). Every trawl was performed with a speed of about 4 knots, and lasted 30 min. Samples of commercial species caught, were retained for this study. After the fish was caught it was sealed in a plastic bag, and frozen at -20°C , and kept frozen until examination. In order to maximise efficiency, with minimum loss of information, the following sample



Fig. 1. Sampling areas. The red dots indicate the starting point of the trawls. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

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