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Bioaccumulation of PCBs from microplastics in Norway lobster (*Nephrops norvegicus*): An experimental study

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HIGHLIGHTS

- Limited PCB bioaccumulation observed after ingestion of PCB-spiked polyethylene.
- Negligible PCB bioaccumulation observed after ingestion of PCB-spiked polystyrene.
- No effect of 3-week microplastic ingestion on nutritional state of *Nephrops*.
- No PCB depuration from *Nephrops* tissue to microplastics.

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ABSTRACT

Plastic debris acts as a sorbent phase for hydrophobic organic compounds like polychlorinated biphenyls (PCBs). Chemical partitioning models predict that the ingestion of microplastics with adsorbed chemicals in the field will tend not to result in significant net desorption of the chemical to the organism's tissues. This is expected due to the often limited differences in fugacity of the chemical between the indigestible plastic materials and the tissues, which are typically already exposed in the same environment to the same chemicals as the plastic. However laboratory trials validating these model predictions are scarce. In this study, PCB-loaded microplastics were offered to field-collected Norway lobsters (*Nephrops norvegicus)* during *in vivo* feeding laboratory experiments. Each ingestion experiment was repeated with and without loading a mixture of ten PCB congeners onto plastic microspheres (MS) made of polyethylene (PE) and polystyrene (PS) with diameters of either 500–600 μm or 6 μm. We observed that the presence of chemicals adsorbed to ingested microplastics did not lead to significant bioaccumulation of the chemicals in the exposed organisms. There was a limited uptake of PCBs in *Nephrops* tail tissue after ingestion of PCB-loaded PE MS, while almost no PCBs were detected in animals exposed to PS MS. In general, our results demonstrated that after 3 weeks of exposure the ingestion of plastic MS themselves did not affect the nutritional state of wild *Nephrops*.

1. Introduction

Do microplastics act as a vector for organic compounds into the marine ecosystem, and does that lead to an increased bioaccumulation of these contaminants in marine animals when the microplastics are ingested? We know that all organic matter in the marine environment has the potential to adsorb hydrophobic organic compounds from the environmental matrices it comes into contact with via a process known as partitioning (Schwarzenbach et al., 2017). Being organic materials, microplastics are no exception. Gauquie et al. (2015) identified more than 200 organic compounds or compound groups on plastic debris. Although banned since the 1980's, polychlorinated biphenyls (PCBs) are still present in the marine environment (e.g. Roose et al., 2005; OSPAR, 2010;

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OSPAR, 2012; De Witte et al., 2016) and omnipresent on plastic debris (e.g. Hirai et al., 2011; Karapanagioti et al., 2011; Antunes et al., 2013; Mizukawa et al., 2013). PCBs are able to partition to any type of plastic present in the marine environment. For example, for polyethylene (PE) the log plastic-water partition coefficients for PCB congeners are high, but comparable to the partition coefficients of other hydrophobic phases (e.g. log BCF or log K_{ow}) (e.g. Mackay, 1982; Shiu and Mackay, 1986; Hawker and Connell, 1988; Choi et al., 2013). For polystyrene (PS), the log PS-water partition coefficients for PCB congeners are lower compared to log K_{ow}. This can be illustrated for congener CB128 for example, with log $K_{PE\text{-}water}$ value of 6.5, log $K_{PS\text{-}water}$ value of 3.13 and K_{ow} value of 6.99 (O'Connor et al., 2016).

Because PCBs sorb well to plastic, plastic ingestion by marine organisms can provide an extra contaminant exposure route, in addition to direct inhalation, dermal adsorption and ingestion (e.g. Teuten et al., 2007, 2009). The transfer of trace metals, PCBs and other organic contaminants through the food chain by plastic ingestion has already been tested for different seabird species (e.g. Colabuono et al., 2010; Tanaka et al., 2013; Fife et al., 2015). These studies showed the importance of plastics as a source of persistent organic pollutants (POPs) and suggested the transfer of plastic derived chemicals from ingested plastic to the marine organisms ingesting them (Colabuono et al., 2010; Tanaka et al., 2013). For several marine species, controlled lab exposures with microplastics and chemical pollutants have been performed providing evidence that under certain conditions sorbed pollutants can desorb from ingested microplastic into aquatic organisms (e.g. Avio et al., 2015; Wardrop et al., 2016). On the other hand, model analyses revealed that microplastic ingestion is not likely to increase the risks of POPs such as PCBs to the aquatic ecosystem because the bioaccumulation of organic chemicals from plastic is probably overwhelmed by the uptake via water or food (Koelmans et al., 2013a,b; Koelmans et al., 2016).

In the current study, we assessed whether ingested PCB-spiked microplastics lead to an increase in PCB body residues in wild Norway lobsters *Nephrops norvegicus*. These animals were pre-exposed to low environmental levels of PCBs in the wild in the marine environment. For bioaccumulation to happen, PCBs need to passively diffuse from the plastic matrix into the biological tissue down a fugacity gradient until either an equilibrium is reached or until the plastic has been excreted. This can be described in thermodynamic terms as the PCB congener aiming to achieve equal fugacities ('escaping tendencies') in the plastic and biological phases (Mackay, 1979), meaning that the uptake of PCBs is expected if the fugacity of PCBs in plastic is higher than in *Nephrops* tissues. In line with modelling studies and the above mentioned partition coefficients, we hypothesized that very small amounts of PCBs would desorb from microplastics after ingestion. We used a set of seven omnipresent indicator PCB congeners and three non-indicator PCB congeners that are not commonly present in the marine environment. The seven indicator PCBs were selected by the OSPAR Commission for priority action as these PCB congeners cover the range of toxicological properties of the PCB group (PCB congeners 28, 52, 101, 118, 138, 153, 180) (OSPAR, 2010, 2012). The different background contamination of both PCB sets in the *Nephrops* individuals may influence the concentration gradient and flux of PCBs.

We investigated the effect of microplastic polymer type (PE and PS) and size of PS microplastics (6 μm and 500–600 μm) on the adsorption of the 10 PCB congeners to plastic microspheres (MS) and the desorption of these PCBs in the gut. Smaller particles have a greater surface area: volume ratio so we expected them to be able to adsorb more PCB per unit mass plastic than the large particles. For the PCBs in this experiment the values for the polystyrene-water partition coefficients ($K_{PS\text{-}water}$) are smaller than those for polyethylene-water ($K_{PE\text{-}water}$) (e.g. Pascall et al., 2005; Velzeboer et al., 2014; O'Connor et al., 2016). This means a relatively small amount of PCB can sorb to PS compared to PE of the same particle size.

We also investigated how PCB-spiked MS ingestion contributes to the bioaccumulation process and the $\sum$PCB body residue of *Nephrops*. Concerning the size, the bigger MS of 500–600 μm are expected to be excreted with faeces, as they are too big to be taken up by gut epithelial cells and small enough not to obstruct the gastro-intestinal tract of *Nephrops* (GESAMP, 2015). Finally, we investigated whether clean (i.e. non PCB-spiked) MS present in the gut negatively impacts the nutritional state of the exposed animals, and whether clean MS contribute to the depuration of PCBs already present in *Nephrops* tissue.

2. Materials and methods

2.1. Norway lobster Nephrops norvegicus as test organism

The Norway lobster *Nephrops norvegicus* is a commercial species for human consumption known to ingest significant amounts of microplastics (Murray and Cowie, 2011).

2.1.1. Nephrops sampling

Nephrops were caught at Cleaver bank ('Pitboeien') (*54°20.0072/005°10.0974)* by a commercial beam trawler (N.350) in November 2013 (Experiment A), June 2014 (Experiment B) and May 2014 (Experiment C) (Table 1). After 6 h trawling, *Nephrops* were kept in aerated tanks to ensure a high survival rate. In the laboratory, *Nephrops* were held in a large tank until the start of the experiment and fed pieces of sprat (*Sprattus sprattus)*, plaice (*Pleuronectes platessa*) and whiting (*Merlangius merlangus*) three times a week. As the ingestion rate ($g.g^{-1}$ body weight d^{-1}) increases with decreasing body size (Sardà and Valladares, 1990), the average carapace length of the test specimen was set between 9 and 13 cm (Table 2). Quarantine procedures were implemented to allow for a selection of healthy individuals, based on condition, lack of parasites or injuries, intermoulting (hard carapace) and carapace length.

Table 1
Overview of the laboratory treatments and composition of the feed for experiment A (500–600 μm polyethylene microspheres), experiment B (500–600 μm polystyrene MS) and experiment C (6 μm PS MS).

Exposure treatment	Feed	Experiment A 500–600 μm PE	Experiment B 500–600 μm PS	Experiment C 6 μm PS
T_0 control group	No feed, assessed before trial	x	x	x
Blank	Gelatin	x	x	x
Positive control	Gelatin+PCBs	x	x	x
Negative control	Gelatin+clean MS	x	x	x
Loaded microplastics	Gelatin+PCB-loaded MS	x	x	x
Positive control with microplastics	Gelatin+PCBs+clean MS	–	x	x