Marine microplastics spell big problems for future generations

Tamara S Galloway and Ceri Lewis

College of Life and Environmental Sciences

University of Exeter

Exeter EX 4 4QD, Devon UK

Tel + 44 1392 263436

Email t.s.galloway@exeter.ac.uk

There are certain human environmental perturbations that are so major, they are capable of destabilising the earth's normal function at a global scale (1). These so-called planetary boundary threats include climate change, ozone depletion and ocean acidification. Emerging as a novel addition to this list is the vast quantity of discarded plastic waste that is accumulating in the oceans on an unprecedented scale, where it breaks down to form microscopic and nanoscopic fragments, or microplastics. Microplastics (particles of a diameter < 1 mm, with no lower limit) derive from progressive fragmentation of larger plastic items, or may be manufactured to be of a small size; for use in personal care products, medicines and industry (2). They reach the seas through beach littering, road runoff, sewage and illegal dumping activities. Microplastics are ubiquitous in marine waters; from deep ocean sediments to polar icecaps, a result of the estimated 8 million tonnes of plastic that enters the oceans each year (3). Despite calls for plastic to be reclassified as hazardous (4), legislation to restrict marine debris accumulation is hindered by a lack of evidence that it causes ecological harm. In this issue of PNAS, Sussarellu and colleagues (5) provide an

important starting point for assembling this evidence: using an integrative approach, they show that ingestion of microplastics during gametogenesis impacts on feeding and reproduction in oysters, with negative impacts on adult fecundity and offspring quality, both key components of an organisms' individual fitness.

These results are important because they support an emerging paradigm that microplastics can reduce reproductive output and fitness in marine species by altering their food consumption and energy allocation. Marine plastic debris is a global threat because of its abundance, persistence and mobility across scales with subsequent widespread distribution and potential geophysical and biological impacts (1). Compelling images of large marine species such as birds and turtles entangled in plastic debris are widespread (6) and many hundreds of marine species have been recorded to ingest plastic debris, leading to physical injury and death. As plastic polymers degrade to form microplastics, their impacts become more subtle. Microplastics are a cause for concern because their size range overlaps with the preferred particle size ingested by animals at the base of the marine food web. Detritus, suspension and filter feeders can readily ingest them, leading to uptake and trophic transfer of the plastics themselves and any chemicals they contain or have absorbed from seawater. Many of these species are important to fisheries or perform vital ecosystem functions.

The impacts of plastic ingestion in laboratory studies include gut blockage and physical injury, oxidative stress, altered feeding behaviour (7, 8) and reduced energy allocation (9) with knock on effects for growth and reproduction (5). Transfer to tissues of plastics associated chemicals, many of which possess endocrine disrupting activity (10) adds to the potential toxicity of ingested particles through activation of signal transduction pathways relevant to hormone action.

Susarellu and colleagues (5) studied oysters, a keystone species of high ecological and economic performance. In shallow, coastal waters, oysters typically form reefs, filtering vast quantities of water and improving water quality and biodiversity. Adult oysters were exposed to microscopic polystyrene at environmentally relevant concentrations for 2 months during a critical point in the reproductive cycle when adults were growing their gametes. Exposed oysters had altered rates of feeding and absorption efficiency from food, reduced fecundity (number of eggs produced), oocyte quality and sperm swimming speed. Importantly, these impacts had clear carry over effects on offspring quality measured as reduced growth intheir larval progeny.

This reallocation of energy reserves from reproduction to maintenance, with resulting reductions in reproductive success, is a recurring theme emerging from chronic exposure studies with microplastics (6). Sediment dwelling worms exposed to sediments contaminated with polyvinylchloride (PVC) microparticles had increased gut transit times and reduced lipid accumulation (8). Similarly, planktonic copepods exposed to micropolystyrene for prolonged periods had reduced food consumption resulting in reduced reproductive output (9). They also showed a downward shift in their preference for algal prey, suggesting altered feeding behaviour post capture or post ingestion.

The cultured oysters showed a high capacity to ingest micropolystyrene with surprisingly high efficiency, clearing up to 70% of the 6 μm beads supplied to each tank each day (roughly 9.6 mg, or 100 beads per ml). Oysters in the wild are evidently capable of ingesting microplastics with similar efficiency. A recent study of oysters cultured in the northeast Atlantic being sold for human consumption found them to contain an average of 0.47 +/- 0.16 microplastics g^{-1} wet weight of tissue, with the most abundant particles and fibres in the size range 11-15 μm (29.6%) and 16-20 μm (33.3%) (11). Based on this, an average dietary portion of 6 oysters (100g) would contain around 50 plastic particles. Even higher

concentrations of microplastic fibres were reported in wild and farmed mussels (12), up to 178 fibres per farmed mussel, presumably due to the presence of ropes and aquaculture related paraphernalia.

In Susarella's study, there was no apparent translocation of the 2-6 μM diameter microplastics across the gut, although translocation of microplastics occurs in other bivalves (11, 13). In laboratory studies, early life stage oysters showed enhanced uptake of nano- compared with micro- polystyrene (14), which would tend to favour uptake across both gut barrier and cell membranes. However, detecting the uptake of nanoparticles in the wild remains beyond the limits of what is technically possible, despite recent advances (15).

Susarellu found that stress responses were activated in exposed oyster digestive tissues, with Dynamic Energy Budget models predicting diversion of energy allocation from reproductive output to structural growth and maintenance. There was reduced activity in the insulin pathway in gonadal tissues, suggesting that the typical mobilisation of resources that accompanies gametogenesis was not occurring. Oysters are broadcast spawners, which release their eggs and sperm into seawater, where external fertilisation occurs. Reduced sperm swimming speeds together with smaller, fewer eggs will reduce fertilisation success (16). Studies of other stressors, such as ocean acidification, show that carry over effects in oyster larvae can persist through to later life, reducing settlement success, population growth and productivity (17).

In Figure 1, we have incorporated these results, supported by previous findings, within a tentative Adverse Outcome Pathway (AOP) scheme. AOPs are extremely useful in deducing the key events linking an apical endpoint such as reduced reproductive output with a perturbation such as particle ingestion because they describe generalised motifs of biological response, or key events that are not necessarily specific to any one chemical or substance. For

example, applying the AOP concept to growth retardation in fish allowed (19) to distinguish the mode of action of cadmium, which reduced growth through increased metabolic demand, from that of pyrethroid pesticides and selective serotonin reuptake inhibitors, which reduced food intake through changes in behaviour and appetite (19). In relation to microplastics, the situation is further complicated by their potential to associate with chemical contaminants and the as-yet unknown extent to which these absorbed contaminants are transferred from the ingested particle into the organisms' tissues.

The wider implications of these finding relate to the similarity in mode of action between the microplastics themselves and the chemicals that are associated with them. Persistent, bioaccumulative and toxic organic contaminants that associate with microplastics in the ocean include polychlorinated biphenyls (PCBs), polyaromatic hydrocarbons (PAHs) and polybrominated diphenylethers (BDEs), all of which possess endocrine disrupting activity (10). This includes a subgroup, termed obesogens, that enhance weight change by shifting energy balance in favour of fat storage in adipocytes and altering basic metabolic rate (20). Obesogenic effects are not limited to vertebrates. The biocide tributyltin (TBT) is a high affinity ligand for the peroxisome proliferation activated receptor gamma (PPARγ) and its heterodimer partner retinoid receptor X (RXR), which regulate lipid metabolism in vertebrates. Waterfleas exposed to TBT showed disrupted lipid metabolism, with reduced transfer of triacylglycerols from adults to eggs, prompting their accumulation in the adults. Similar to Susarellu's microplastics exposed oysters, the life history responses of the progeny of females exposed to TBT showed reduced fitness, had lower survival and produced fewer eggs (21). Thus, a situation could well arise where significant potentiation of the mode of action of microplastics and the contaminants they are associated with could occur.

Strategies for buffering marine biodiversity against global threats such as climate change and ocean acidification include reducing additional stressors such as pollution and over fishing

(22). As plastic waste is one of the most prevalent of marine pollutants, reducing plastics input should be a high priority. Given the impossibility of removing all microplastics contamination from the oceans, the impetus is on all of us, governments, scientists and individuals to reduce our utterly ridiculous levels of plastics consumption and waste before we induce permanently alterations to our fragile marine ecosystem.

Figure legend

Figure 1, A tentative Adverse Outcome Pathway scheme for microplastics exposure of aquatic species showing potential pathways linking ingestion, uptake across membranes and chemical release with adverse outcomes of growth inhibition and reproductive decline.

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