

**MODELING HEALTH RISKS OF
MICROPLASTICS: AN UNCERTAINTY ANALYSIS**

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Dedication

To my mother, Sheila, my father, Leigh and grandmother, Cecil, who instilled in me a love of nature, and who refused to give up on me when I was seemingly lost. You have my eternal gratitude for never leaving my side, for making me laugh, and for your faith in me.

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ABSTRACT

The global issue of ocean plastic pollution is undisputed and fast-growing. The preferential sorption of persistent organic pollutants to plastic debris is also proven. However, the relative contribution of microplastics to the total toxin load in consumers at all levels of the food web is unknown. Other potential pathways include dermal sorption and direct consumption of persistent bioaccumulative contaminants through food. An abundance of diffuse experimental data exists to test this pathway, but without standardization in this field of study, results are not directly transferable and translatable to examining toxin loading in higher organisms such as humans. As a result, microplastic effects on toxin loading in humans has heretofore been unknown or minimized, where it should be strongly considered in environmental policy decision making.

This study develops a stochastic model to test various scenarios of possible toxin loads to humans as a result of secondary microplastic consumption. The model incorporates a number of different investigations testing organic pollutant concentration on plastics and the rate of adsorption or desorption of these chemicals within the animal gut. Of the seven scenarios tested, two resulted in toxin loading to humans within an order of magnitude of allowable daily limits of these chemicals. The other five scenarios predicted no probable human toxin load within an order of magnitude of most organic pollutant Acceptable Daily Limits. Finally, the physiological and immunological effects of microplastic consumption are discussed from the currently available body of literature, and incorporated into recommendations for the control of ocean plastic pollution and of human microplastic exposure through seafood.

INTRODUCTION

The Emerging Ocean Plastics Problem

The issue of plastic accumulation in the ocean is vast and growing. Plastic debris kills about two million seabirds and 100,000 marine mammals each year (Castro, 2015). According to the Ocean Conservancy, plastic entering the ocean is projected to double from 2015 to 2025. A business-as-usual (no mitigation) projection predicts that by 2025, there will be a 1:3 ratio of plastic to finfish in the oceans by weight (Ocean Conservancy, 2015). According to a 2006 study by Allsopp et al., roughly 4.6% of all plastic produced ends up in the oceans, while Barnes et al. (2009) estimated this number at 10% of global plastic production. As of 2014, the ocean contains an estimated 5.35 trillion particles (~268,940 tons) of plastic (Eriksen et al., 2014). Of this global plastic debris, 92.4% of it is estimated to be microplastic, defined by this author as <5mm diameter plastic (Eriksen et al., 2014). The persistence time of plastics in the marine environment is uncertain, although one estimate concludes that plastics may lose only a small percentage of their total carbon content in a decade (Engler, 2012).

Roughly 80% of that plastic marine debris is land-derived, and the other 20% enters the ocean via sea routes and ship traffic (Allsopp et al., 2006 Ocean Conservancy, 2015). Despite the 2002 decision at The International Convention for the Prevention of Pollution from Ships (MARPOL) to ban all plastic dumping at sea, 6.5 million metric tons of plastic per year are estimated to enter the ocean by this route (Allsopp et al., 2006). The ICIS Supply and Demand database projects that the demand for plastics will increase from 250 million metric tons in 2015 to 380 million metric tons in 2025 due to user benefits, population growth, and economic growth.

As the mass production of plastics worldwide began relatively recently in the 1970s (SPI, 2015), issues surrounding excessive plastic pollution in the oceans are new and emerging, and pose unknown threats to human health. As plastics do not fully biodegrade -- instead breaking into progressively smaller pieces-- microplastic beads are of increasing concern in marine food chains. Microplastics (defined in this paper and others as pieces of plastic with diameter under 1 mm) are manufactured for use in personal care products such as skin exfoliates, toothpastes, and hand scrubbers, and are the feedstock of plastic products in general. Microplastic fibers are sloughed off fabrics containing plastic components during the laundering and wearing processes. Microplastics from this source can enter the environment after wastewater treatment, which fails to filter them. They also are generated in the breakdown of plastics in the natural environment.

Mitigation Issues

Policy, manufacturing, and mitigation decisions made over the next 10 years are critical for solving this problem. Total cost of mitigation is estimated at around \$5 billion/year by the Ocean Conservancy, which claims huge returns on investment in health, food supply chain, and tourism sectors, among others. Despite the enormity of the plastic pollution problem and the relatively low mitigation costs, no effective reduction strategy has been implemented.

Health Risks for Animals and Humans

Microplastics attract persistent organic pollutants (POPs) in the water, and are ingested by organisms as small as plankton and as large as baleen whales. The degree to which POPs are desorbed within the animal gut is still under investigation, but has been cause for concern within the scientific community. Consumption of larger-sized plastics has been documented as fatal in many ocean creatures, including sea turtles and pygmy sperm whales. Microplastics (of similar size and buoyancy as krill) have been sampled in both bivalves and fish taken for human consumption (Van Cauwenberghe and Janssen, 2014). However, the secondary effect of microplastics on humans as we consume animals that have ingested them is not well understood.

Ocean plastic pollution is seldom linked to human health, but a growing interest in this matter is fueling more investigation. Consumption of microplastics by organisms

harvested for seafood poses potential toxicological, immunological, and physiological risks to humans.

Literature Review & Background

Degrading plastic toxicity and its health effects is now a popular topic of research and debate, as evidenced since 2011 in the pages of the Journal of Marine Environmental Research. Contentious arguments exist over the relevance of microplastic to POP bioaccumulation. In these studies, bivalves are often used as monitors of aquatic pollution because they are widely distributed geographically, have a mostly-sessile lifecycle, have a relatively wide salinity tolerance range, exhibit stress resistance, show a high volume of water filtration, accumulate a high pollutant accumulation, and are easy to sample (Vandermeersch et al., 2015).

Despite varied risk analyses of microplastics as carriers of persistent organic pollutants (POPs), some researchers maintain that, because persistent organic pollutants preferentially sorb to microplastic beads, a bioaccumulation effect of POPs from microplastic consumption is certain. This is partially due to microplastic concentrating POPs from seawater. According to Mato and colleagues (2001), plastic debris sorbs Polychlorinated biphenols and degradant species of the pesticide DDT roughly 100 times better (on average) than natural organic matter in suspension within the water column, but sorption preferences (K_d) can be up to 1 million times greater than seawater (Mato et al., 2001).

Conversely, some recent research indicates that, due to the difference in dissociation constants between ocean and animal gut environments and the gradient of pollutant concentration within the animal gut, POP bioaccumulation does not necessarily occur (Koelmans et al., 2013). According to Koelmans et al.'s 2013 analysis, both closed laboratory bioassays as well as models predicting the behavior of open ocean systems indicate that ingestion of microplastics causes dilution of POPs – a cleaning effect – and in fact decreased toxin bioaccumulation. However, these changes are estimated to be minute (Koelmans et al., 2013). Sources that claim POP bioaccumulation is still a risk of microplastic ingestion far outnumber the studies to the contrary.

As Bouwmeester et al. (2015) maintain, the risks of microplastic ingestion don't stop at the parameters examined by Koelmans et al. Nanoplastics (<100 nm in size), the

probable next step in plastic degradation, are even more poorly studied than microplastics. With a greater surface area: volume ratio, they would expectedly move more easily between membranes (Bouwmeester et al., 2015). The risk of leeching plastic additives (also harmful chemicals), the physiological effect of consuming microplastics, and the POPs bound to the plastics all remain potential concerns (Bouwmeester et al., 2015). A number of factors of uncertainty, including the rate of sorbance and desorbance of chemicals from microplastics, are incorporated into this uncertainty analysis. Physiological and immunological effects such as translocation of micro- and nanoplastics to tissues outside of the gut are also addressed in this uncertainty analysis.

Study Goals

Accordingly, this paper examines key human toxicological variables in response to consuming mollusks that have been contaminated with plastic microbeads by using available data from literature as inputs to a probability model. Mollusks are both commonly consumed by humans and frequently used as test subjects in marine toxicology due to the volume of sea water they filter, their exposure levels as filter feeders, their ubiquity, their ease of collection, and other metrics. Incorporating physiological and immunological effects in other organisms, a new study on overall microplastic effect on mammals is suggested to elucidate the effects of secondary microplastic consumption on mammals.

To actively reduce this worldwide threat to ocean and human health, this study employs a threefold approach, intending to serve as a:

- a) Compilation of existing information on the toxicological and physiological effects of microplastic ingestion via seafood consumption;
- b) A stochastic and *in vivo* experimental framework for further risk assessment of human exposure to microplastics, and
- c) A call for policy changes that would mitigate human health and ocean health effects from plastic pollution.

METHODS

Here, the primary goal is to develop and assess a stochastic model accounting for much uncertainty in POP sorption to/desorption from microbeads within the animal gut. As there is a tremendous amount of variability in sorption/desorption behavior based on the contaminant and plastic type (Rochman, 2013c), ranges of values observed in the literature are used to account for known POP-plastic combinations. A multivariate sensitivity simulation takes into account a range of plastic types, pollutant concentrations (sorbed to and within the plastic), and possible chemical gradients within the animal gut (both human and bivalve). Variable ranges are based upon previous groups' findings (Bakir, 2014, and Rochman, 2013c, Koelmans et al., 2014). The following terms and equations are used in the model.

Terms and Equations

Abbreviations

MP: microplastic, size <1mm (for purpose of this experiment).

POP: persistent organic pollutant (includes PAHs, PBDEs dioxins, DDTs and PCBs). Many countries have outlawed these, but they are still in use in others, and are expected to persist for many years in the aquatic environment.

PBT: a persistent, bioaccumulative, and toxic substance. More inclusive than the classification of POP.

PAH: polycyclic aromatic hydrocarbon, a component in crude oil and byproduct of combustion. Classified as a POP.

PCB: polychlorinated biphenol, a transformer fluid and plasticizer, both an additive and a sorbent. Classified as a POP.

DDT: dichlorodiphenyltrichloroethane (the term 'DDTs' includes the degradants DDD and DDE as well as DDT). A pesticide and a POP.

Dioxins: polychlorinated dibenzo-p-dioxins. Industrial and incineration byproducts, carcinogenic. A POP.

PBDEs: polybrominated diphenyl ethers. Flame retardants, plastic additives, often added to seat cushions. Classified as a POP.

NP: nonylphenol

OP: octylphenol

BPA: bisphenol A

PE: polyethylene, a plastic

PP: polypropylene, a plastic

PS: polystyrene, a plastic

PVC: polyvinyl chloride, a plastic

(source: Engler, 2012)

Equations

$$C_t = C_{eq} (1 - e^{-kt})$$

Where:

C_t is the concentration at time t ,

C_{eq} is the predicted equilibrium concentration, and

k is the first-order rate constant (units $\frac{1}{day}$), where k_1 is often adsorption rate and k_2 is desorption rate. For the proposed model, both k_1 and k_2 are converted to a percentage of largest observed daily rate of desorption or adsorption, with adsorption represented by negative percentage values and desorption represented by positive percentage values.

$$K_d = \frac{[q_e]_{solid}}{[C_e]_{aqueous}}$$

Where K_d is the distribution coefficient (partition coefficient). High K_d means high affinity of a compound for plastic over water.

$[q_e]_{solid}$ is the amount of contaminant sorbed onto plastic at equilibrium

(in μg chemical /kg debris)

$[C_e]_{aqueous}$ is the contaminant concentration in the aqueous phase at equilibrium (in $\mu\text{g/L}$)

Likewise, rearranging this equation, we can solve for $[q_e]_{solid}$, the amount of contaminant associated with microplastic in equilibrium (in $\mu\text{g/kg}$)

$$[q_e]_{solid} = K_d \times [C_e]_{aqueous}$$

Building on this formula, we can solve for the mass of POP ingested via microbeads:

$$m_{POP} = [q_e]_{solid} \times m_{MP}$$

where m_{POP} is the mass of POP ingested via microbeads (in μg)

and m_{MP} is the mass of microplastic (in kg, for unit accounting in this equation)

Variations of these formulae were used to inform the model.

General Terms

K_{ow}: The octanol-water partition coefficient, or ratio of chemical concentration in the octanol phase to concentration of chemical in aqueous phase, at equilibrium.

R_fC: Reference concentration

ROS: Reactive Oxygen Species

ADI (also ADL or R_fD): Acceptable daily intake/Acceptable daily limit/Reference Dose

TDI: Tolerable daily intake

BMD: Benchmark dose

BMDL₁₀: Lower limit of BMD causing a 10% effect on test subjects

EC₂₀: The effective dose causing 20% response (subscript is variable)

NEL: No Effect Level

NOAEL: No observed adverse effect level

LED₁₀: Dose associated with 10% increase in tumor incidence, used to evaluate carcinogenicity

LD₅₀ : Dose of specific chemical that was lethal to half (50%) the animal test subjects used

EEQ: Oestradiol-equivalent concentrations: multiply concentrations of individual estrogen-like compounds by relative potency.

Software

The model itself is built in *Vensim PLE Plus* software. Monte Carlo analyses are multivariate sensitivity simulations in which sampling of constants over a range of values occurs automatically, per modeler specifications (Ventana Systems, 2016).

Literature Review

The modeling portion of this study is followed by a literature review on the observed immunological and physiological effects of plastic microbead consumption on mollusks and humans. A further study is designed and suggested due to the need for mammalian models to approximate human health impact.

In addition to modeling chemical fates, a toxicological viewpoint of POP consumption will also be briefly reviewed. Risk assessment of human consumption of common plastic-associated pollutants are reviewed based upon animal studies, case

reports, and *in silico* studies. Common POPs are reviewed and their toxic effects such as carcinogenicity, mutagenicity, neurotoxicity, and endocrine disruption are noted. Dose-response relationships are also reported for ingestion exposure routes of these chemicals. LD₅₀ levels (doses of specific POPs that were lethal to half the animal test subjects used) are noted for these chemicals, as well as lesser reactions to oral routes of exposure. Likely physiological responses of exposure in humans via microplastic-laden seafood is discussed for risk assessment.

Research Constraints

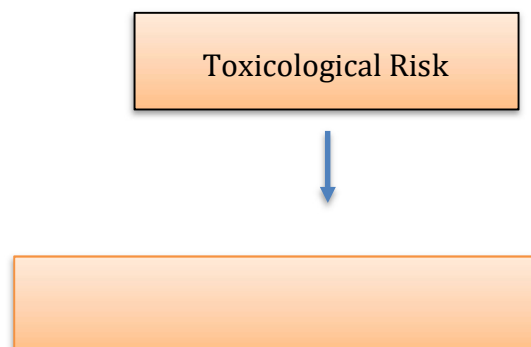
The modeling parameters in this study will be viewed by some as overly simplistic. However, this tradeoff must be weighed in light of the modeling goals:

- a) To gain generalized insight without losing the predictability that current models provide, and
- b) to scale the important factors involving toxin load up trophic levels.

With full acknowledgement that this approach is a broad sweep rather than precise data point delineation, the aim of this study is to illustrate likely scenarios of human toxin loading as a result of secondary microplastic consumption, based upon existing research. Note that synergistic effects of chemicals when consumed together have not been determined, and that mixes of organic chemicals vary at any given spatio-temporal point in the ocean (Rochman et al., 2013c, Hayes, 2015). Nevertheless, this model is a reasonable benchmark for human toxin load from secondary microplastic consumption, to the best of this author's knowledge. The model presented could act as a framework upon which to test future sensitivity analyses as more information comes to light.

Overall risk analysis subsequently takes into account not only toxin load, but also known immunological and physiological risk from testing. Below is a schematic of the risk assessment methodology implemented.

- model
- total human toxin load

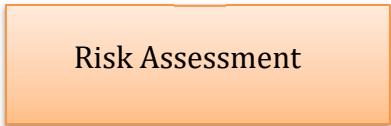


- literature review
- new experimentation

Immunological Risk



- literature review
- new experimentation



- relative risk, categorically
- risk in non-health sector

- cost of treatment/human lives (based on animal epidemiology studies)
- cost of mitigation (based on Ocean Conservancy estimates)
- time to mitigation

RESULTS

The fact that microplastic pellets preferentially sorb POPs is well-accepted in the scientific community—so much so that one global program measures ocean PCB content based solely upon their presence on microplastic beads found in seawater (pelletwatch.org scientific group). Passive sampling devices use plastics (PVC, PE, PS, POM) because of their ability to concentrate hydrophobic chemicals such as PCBs, PAHs, and PBDEs, and microplastics have been studied as bioremediation materials for soils and sediments due to their ability to concentrate organic chemicals (Koelmans, 2015). Interestingly, sorption to microplastics is expected to inhibit chemical degradation of sorbents by sequestering chemicals from microbes (among possible mechanisms), and thus may increase the persistence of these chemicals in the environment (Teuten et al., 2009).

Measuring the transfer of MP-associated pollutants and plastic additives has proven difficult for researchers. There is still a large amount of uncertainty surrounding the bioavailability of both sorbed and intrinsic organic pollutants. Van Cauwenberghe et al. (2014) are among many proponents that MPs present in food potentially threaten human health. However, because of the complexities of the toxicology associated with microplastics, evaluation of potential human health risks is deemed “not yet possible” by the scientific community (Koelmans et al., 2015, Engler et al., 2012, and others).

The seeming impossibility of measuring human toxin loads as a result of MPs should not deter policymakers from acting quickly on this matter. A probabilistic Monte Carlo model may shed more light upon possible contaminant loads in human consumers of ocean bivalves due to secondary microplastic consumption, based upon available data, without the delay of further experimentation (see **Appendix V**). This model, along with other risk assessment tools, may be useful to inform legislation concerning all persistent organic and endocrine disrupting/carcinogenic compounds associated with MPs. It can also be used to estimate possible transfer of a specific chemical, by simply changing the model parameters.

Model Premise

In 6 months of peer-reviewed scientific literature search from February–July 2016, no single model was found that accounted for the transfer of a stochastic mixture of chemicals associated with microplastics to the human body as a result of secondary ingestion of microplastics. The stochastic model presented in this paper provides an estimate of possible toxin loading in humans who consume MP-contaminated seafood, based on known desorption rates of POPs from MPs in the mammalian gut and bivalve gut. It is neither a chemical kinetic model nor a fate and transport model, but is instead meant solely for use in risk analysis of microplastic effects upon human health.

Confusion about the relevance of MPs as POP carriers compared to other sources (food, direct water desorption, etc.) has hindered policy development (Koelmans, 2015). While a number of modelers (Koelmans and Guin among them) have attempted to narrow down the POP uptake system to a set of first order partial differential equations, these have translated poorly to actual animal gut conditions and experimental results, despite the attempt to account for greater dissolution in gut surfactants, size of gut area,

and chemical gradient in the gut. There are factors at play that are not captured by these models and are in fact not yet understood, because toxin loading is physically as well as chemically facilitated by microplastics (Paul-Pont et al., 2016).

It has been suggested that a system of partial differential equations might yield more specific results than a Monte Carlo simulation. This would be true if data for each of these parameters was well-defined. Unfortunately, variables at play in a model of the gut desorption of POPs from microplastics involve a number of uncertain variables, which could change with any newly published experiment. In the absence of specific parameters but given a set of observed ranges, a Monte Carlo simulation allows for hypothesis testing and simulation, while providing a platform for more specific analyses (including systems of partial differential equations) in the future. These approaches are not mutually exclusive, but the Monte Carlo approach is more well-suited to the quality of data that exists today in this complex system of variables.

Another objection to this model regards the wide ranges of parameters it uses. It has been argued that such a wide range within a model could not predict useful outputs. While this may be true in general, Monte Carlo simulations with 2,000 iterations have yielded clear peaks in possible human toxin loads.

The dissolution and adsorption factors used in this model are based on actual experimental observations in the animals used or in simulated gut conditions. The *in silico* models by Koelmans et al. and Guin et al. translate well to experiments in lugworm but fail to predict toxin loads in higher animals (seabirds and fish). A dynamic variable model that accounts for a wide range of uncertainty and whose parameters could be easily changed to fit new data provides policymakers a new risk analysis tool with which to evaluate human toxin load as a result of secondary microplastic consumption. Though this model is, by comparison, very simplistic, it can be used to make the necessary speedy decisions on ocean plastics and global pollutants which otherwise would be stalled by science on toxin loading that is still in its infancy. Every new experiment may yield new surprises that change the game, but it would be inadvisable to fail to act on a severe global pollution issue due to uncertainty as to its human toxicological effect.

Previous Models: Explanation, Strengths and Deficiencies

Here, previous models created by Koelmans et al., Gouin et al., and Reijnders et al. will be discussed, as they provided the basis for the current experiment.

To begin, Koelmans et al., (2013 & 2015), Reijnders et al., (2011), and Gouin et al., (2011) assume that the flux of a chemical within the body is proportional to its concentration gradient, that is:

$$\frac{dc}{dt} = k \cdot \Delta C \quad (\text{Reijnders, 2011}).$$

While theoretically this is true, a number of experiments (Teuten et al., 2009, Avio et al., 2014, and others) have proven that concentrations of chemicals can accumulate in the animal body upstream of the chemical gradient. Clearly there are physiological factors at play in this toxicological model that are not fully understood, and thus are difficult if not impossible to model. This lack of understanding is one main driver for use of a Monte Carlo simulation rather than a Physiologically Based Kinetic (PBK) model, as suggested by Reijnders et al., or a variation on one of Koelmans et al.'s models. That said, a truly detailed PBK model including mammalian tissue/blood partition coefficients, tissue volume, cardiac flow, and kinetic constraints (including biotransformation reactions), could possibly yield better results for higher organisms than did Koelmans et al.'s model.

Koelmans et al., 2013 and 2015:

In Koelmans et al., 2015, a highly complex model was proposed which took into account a number of important parameters in absorption of POPs from microplastics within the animal gut.

The model begins with pollutant adsorption to plastic in seawater, in a situation with excess seawater and sediment in which the concentration of pollutant in the water (C_w) does not decrease with sorption to plastic, and there is a constant $C_{w,0}$:

$$C_{PL} = C_{w,0} \frac{k_1}{k_2} (1 - e^{-k_2 t})$$

Where C_{PL} is the concentration of pollutant on the plastic, and k_1 (in L/kg*day) and k_2 (in 1/day) are forward and reverse first order rate constants related to boundary layer thickness and chemical diffusivity (Koelmans, 2015).

The net absorption efficiency $a_{PL,t}$ for plastic is accounted for by the amount of contaminant removed in gut passage, per the equation:

$$a_{PL,t} = C_{PLR} / C_{PL,ING}$$

Where C_{PLR} is the concentration of chemical removed from plastic during gut passage, and

$C_{PL,ING}$ is the concentration of pollutant on the plastic upon ingestion.

Koelmans et al.'s model goes on to describe the bioaccumulation of hydrophobic chemicals from an environment containing plastic, taking into consideration three routes of exposure (dermal, food, and plastic), as well as elimination (egestion):

$$\frac{dC_{B,t}}{dt} = k_{derm} C_w + IR (S_{food} a_{food} C_{food} + S_{PL} C_{PLR,t}) - k_{loss} C_{B,t}$$

Where:

k_{derm} is the dermal first order rate constant governing gill uptake from water,

the middle term is the uptake from diet and exchange with plastic particles,

$S_{food} + S_{PL} = 1$, the mass fractions of food and plastic in ingested material,

IR (g/g*day) is the mass of food ingested per unit time and organism dry weight,

C_{food} is the chemical concentration in the food,

a_{food} is the absorption efficiency from the diet,

$a_{food} C_{food}$ is the contaminant concentration transferred from food,

$S_{PL} C_{PLR,t}$ is the transferred pollutant from the plastic during gut passage, and

$k_{loss} C_{B,t}$ is the loss of contaminant due to elimination and egestion.

This equation accounts for pollutant load from dermal contact (including gills), as well as food and microplastic consumption, taking into account the relativity of MPs as a pollutant source.

Finally, the concentration of transferred pollutant from plastic during gut passage is given by the following equation:

$$C_{PLR,t} = [(k_{1G}C_{PL} - k_{2G}C_{L,t}) / (k_{1G} + \frac{MPL}{ML} * k_{2G})] * [1 - e^{-(k_{1G} + (MPL/ML) * k_{2G}) * GRT}]$$

Where:

$C_{PLR,t}$ In $\mu\text{g}/\text{kg}$ is the transferred concentration of organic pollutant from plastic during gut passage.

k_{1G} and k_{2G} are forward and backward first order rate constants, respectively, in (1/days). These describe the transport between plastic and biota lipids inside the gut, and are used in the stochastic simulation in this paper.

$C_{L,t}$ is the concentration of lipid biota (fatty tissue) ($\mu\text{g}/\text{L}$)

C_{PL} is the concentration of ingested plastic ($\mu\text{g}/\text{kg}$)

MPL is the mass of plastic

ML is the mass of lipid in an organism (kg)

GRT is the gut residence time, in days

Theoretically, if the numerator of the above equation is positive, then transfer of pollutant from plastic to lipid occurs. On the other hand, if the numerator is negative, a “cleaning” effect occurs in which pollutants from within the organism adsorb to the MP from the gut. Koelmans et al. claim that the cleaning mechanism dominates in closed laboratory systems, and is expected to dominate in ocean environments also.

Koelmans et al. also argue that for plastic to be a significant POP source to animals, the plastic partition $M_{PL}K_{P,PL}$ must be sufficiently large compared to the terms $V_w + M_{SED}K_{P,SED}$ (for sediment component) and similarly compared to $M_i K_{Pn,i}$ for phytoplankton component. It also must compare in magnitude to POP transfer from dissolved organic content. These terms above are mass times partition coefficients with respect to various media (water, plastic, and sediment). V_w is the volume of water.

The same paper also introduces an equation to measure total pollutant body burden at steady state (equation not restated here), which takes into account all reasonable pathways of uptake and loss. In this way, Koelmans et al. account for pollutant partitioning between water, food matter, and plastic, as well as gut conditions including chemical gradient within the gut, gut size and gut surfactant. This model examines the importance of microplastic as an uptake/loss pathway.

Koelmans et al. conclude that small increases in body burden can occur at PE concentrations of 1-10% in sediment, but that this concentration is 100-1000 times higher than the highest plastic concentrations reported, 81 mg/kg, and even in this case effects are deemed too small to be considered relevant to a risk assessment (Koelmans et al., 2013). Koelmans et al. used a previously reported order of magnitude uncertainty value from bioaccumulation parameter studies in benthic invertebrates to disregard positive bioaccumulation data. These researchers predict a predominant cleaning effect in the marine environment, resulting in roughly 10% bioaccumulation decrease in marine animals (bioaccumulation percentages are not directly translatable to the proposed model).

The Koelmans et al. model was parameterized for lugworm consumption of PS MP, and the authors claim that it is easily translatable to other species and plastic types. However, for experiments on shearwater chicks (Teuten et al., 2009), mussels (Avio et al., 2015) and fish (Rochman et al., 2013a), among others, the model's prediction of overall decrease in bioaccumulation via cleaning mechanism simply does not translate to *in vivo* results, even when animals were dosed with pollutants from non-MP sources and thus had a lower gradient. Toxins were able to bioaccumulate counter-gradient via mechanisms that are not well understood.

To its credit, the Koelmans et al. model is thorough enough to be able to account for nanoplastics (<100 nm), and raises many important points in terms of parameters relevant to gut desorption/adsorption from/to microplastics. Their model predicts more interesting effects from nanoplastics with $k \gg 10/\text{day}$, though again bioaccumulation effects were predicted to be very small in magnitude as a result of nanoplastic ingestion.

Perhaps the largest drawback of Koelmans et al.'s model is its failure to account for a large range of POPs. PAHs and PBDEs break down in the natural environment but are preserved on MPs (Rochman et al., 2013a), which increases MPs' role as a relevant pathway (Koelmans, 2015). So, the cleaning effect/bioaccumulation suppression is less relevant for otherwise degradable POPs (Koelmans, 2015). Koelmans et al.'s model was created to model PCB bioaccumulation. (PCB is not degradable.)

Gouin et al., 2011:

Gouin et al. (2011) created a thermodynamic partitioning model that predicts the negligible contribution of PE MPs to body burden of organic pollutants. The model predicts a decreased body burden upon consumption of MPs for chemicals with $\log K_{ow}$ between 5.5 and 6.5, and claims higher POP sorption to non-lipid organic matter than to MPs for K_{ows} in this range. The predicted body burden decrease in this model as a result of MP consumption varied from 0 to 20% (not directly translatable to k-values for use in the proposed *Vensim* model). In this model, MPs are assumed to be 10% of the organism's diet.

Gouin et al.'s model does not include gut retention time or gastrointestinal fluid effects. The authors admit that increased prevalence of plastic would increase its relevance as a vector for POP transport. The fugacity within the gut depends, among other things, on the percentage of lipid and non-lipid organic matter (NLOM) content of the organism's food, while gut extraction efficiency also depends upon K_{ow} of the chemical. The authors claim that gut absorption efficiency decreases at K_{ow} values above 5.5, citing two other papers on biomagnification (Gouin et al., 2011).

Gouin et al. (2011) also found PE to be an insignificant sorbing material, sorbing less than 0.1% of chemical mass, in comparison to dissolved organic content (DOC) or phytoplankton. Yet, the cleaning effects predicted by Gouin et al. and Koelmans et al.'s *in silico* models is also claimed to be "too small to be relevant from a risk management perspective" (Koelmans, 2015).

Lessons from Gouin et al. and Koelmans et al.:

First-order forward and reverse rate constants are converted to positive (forward, desorption from plastic to gut) and negative (backward, sorption of POPs from gut to plastic) percentages of total possible ad/desorption in the Monte Carlo model.

First order rate constants are always multiplied by masses or concentrations. In the Monte Carlo model, k values are translated to percent desorption and percent adsorption (though not a direct translation, an approximation was needed for the simplistic single-step model).

k1: plastic to lipid transport first order rate constant, 1/day (in the Monte Carlo model, positive percentage values)

k2: lipid to plastic transport first order rate constant, 1/day (in the Monte Carlo model, negative percentage values)

For the first six scenarios in the Monte Carlo model, k values in the Monte Carlo model were calculated based upon a maximum desorption value of 100/day for nanoplastics, calculated by Koelmans et al. (2013). Scenario 7 takes k values as a percentage of a 10/day maximum, close to maxima found by Teuten et al. (2007) and Bakir et al. (2014a).

k1 plastic to lipid transport rates in the gut used in Koelmans et al.'s model range from 0.1 to 20 / day for 0.4-1.3mm particles, based on PCB diffusivity from PE (Koelmans et al., 2013). However, Koelmans et al. (2013) considered k1 values of 0.1 to 12/day for their modeling purposes, and these are the k1 values I have used in the Monte Carlo model in the invertebrate gut. In Teuten et al.'s experiments, k1 rates (from PE to seawater + surfactant) vary from 4-12/day, which is consistent, though they did not report reverse k2 values (Teuten et al., 2007). The maximum k1 values reported by Teuten et al., 2007 and Bakir et al., 2014a were 10-12/day. k1 rates would be even faster (>100/day) for 0.1-1µm micro-and nano-plastics (Koelmans et al., 2013).

k2 values (reverse rate constants) were calculated in Koelmans et al. as $k1/K_{PLIP}$, where K_{PLIP} is the ratio between lipid-water partition coefficient (K_{LIP}) and plastic water-coefficient (K_{PL}). k2 values used in Koelmans' experiment are not reported, either within the article text or in supporting information, nor are the K_{PLIP} values from which they would be calculated. They would be assumed higher than k1 values due to the gradient equation below, and the finding that the cleaning (adsorption) effect predominates within the animal gut:

Gradient equation: $k1C_{PL, ing.} - k2C_{L, ing.}$

That is, the forward first order desorption rate constant times the concentration of pollutant on plastic ingested, minus the adsorption rate constant times the concentration of biota lipid at the time of ingestion.

Koelmans et al.'s experiment uses k1 values between 1 and 10, with 10 being default, and shows linearly decreasing concentrations of POP in the gut with addition of more PE. That is, 1% plastic → 10% cleaning effect, 10% plastic → 25% cleaning effect

in open marine simulated conditions (Koelmans, 2013 supporting information). (Without plastic, dermal, and sediment sources contribute 100% of PCB load.) From this I theorize that the k_2 values used, all else held constant, must be greater than k_1 values, but their exact values would need to be sourced from the author. By fitting a line to the graph described above and taking its slope, I theorize that k_2 values are approximately a factor of 1.6 higher than k_1 values for the organism that the graph is adjusted to (presumably lugworm *A. marina*). I use higher k_2 values than k_1 values in one of the model runs for invertebrates only, as a nod to this experiment.

k_2 values used in the Monte Carlo experiment for invertebrates will be approximately $-1.6 * k_1$ values, yielding a range of 0.2 to 19.2 /day. These are incorporated into a single rate, such that k_2 values are negative percentages of total resorption (100%).

Gouin et al. did not publish values that are directly translatable to the proposed model, but rather reports results in body burden only. However, Gouin et al.'s and Koelmans et al.'s approaches and conclusions are similar, and Koelmans et al. use Guin et al.'s results in their model, thus the use of Koelmans et al.'s data also encompass Gouin's concept.

Koelmans et al. claim that most lab experiments use clean test subjects and contaminated MPs, a condition which favors chemical transfer, whereas in the field, the gradients would be lessened or reversed (Koelmans et al., 2015). To account for this critique and reduce error in the proposed study, I have cited data from experiments with more realistic toxin loads in test subjects. However, the only way to know for sure whether experimental gradients are truly realistic is to perform experiments upon resident animals, which is yet to be done (Rochman et al., 2013a).

Hypotheses

I propose that, by building a model which takes into account observations from a multitude of experiments (as well as many different POP-plastic pairs, and test animals), the consumption of a realistic amount of MPs will contribute to total body burden in humans, rather than decreasing it.

This hypothesis would be confirmed if the most probable model outputs estimating total human body burden of microplastic-associated organic pollutants were

nonzero and positive, as well as within an order of magnitude of established acceptable daily doses of most POPs.

The null hypothesis--that secondary consumption of microplastics does not affect human body burden of associated organic toxins --is supported if the most probable outcomes were near-zero, or negligible in comparison with established ADIs.

Finally, the cleaning effect hypothesis would be supported if the most probable model outputs were negative, and within an order of magnitude of ADIs, showing decrease in body burden.

Limitations and Assumptions

The proposed model cannot account for chemical mixtures/synergistic effects. To the best of this author's knowledge, the physiological effect of consuming a mix of POPs and other organic pollutants is not yet known.

This model does not account for specific POP-microplastic combinations, but rather incorporates available information on a multitude of POPs, additives, and microplastic types. In this way, it simulates a natural mixture of MPs and POPs. However, using available data on selected POPs and microbeads, it could be run as a specific model.

The proposed model does not account for toxicity of desorbing plastic polymers. For purposes of this experiment, plastics are considered biochemically inert (see discussion below for further details on plastic physiological effects). According to Araújo et al, 2002, residual monomer content of plastic ranges from 0.0001% to 4%, and monomers leach out of the plastic material. Some of these are toxic, with either carcinogenic or mutagenic properties (PVC, PS, PU). PVC, for example, has a carcinogenic monomer (Rochman et al., 2013a). However, as no daily allowable limits have been set for monomers, they are not analyzed in this experiment.

The proposed model does not have a time component, and thus does not allow for changes in residence time within the gut. (Though this variable could easily be added if need be.) Rather, all chemical transfer is presumed to take place within the course of a single day, per organism. Percent desorption or adsorption in this simulation is for total gut residence time of the plastic (in the case of the mussel, before it is consumed). In reality, residence time of MPs in a mollusk range from 12 (digestive tract) to 48

(hemolymph) days (Browne, 2008, unpublished data from Santana et al., 2016), while in a human the gut residence time of MPs is unknown.

Assumption 1: There are no sufficiently harmless metabolites of POPs produced in the timeframe of this model. Most POP metabolites found in literature review are also fat-soluble and have potential for bioaccumulation or endocrine effects. Metabolites are assumed to be toxic, though some metabolites of faster-degrading materials (additives) may be less toxic than their original forms.

This model assumes no metabolism of POPs or additives to inert or non-harmful forms (see last section for endpoints of the chemicals in question). In the short time frame of this model, it is assumed that faster-degrading additives are also not degraded to innate, non-toxic forms.

Assumption 2: POPs desorbed within the mollusk itself, as well as POPs desorbed from MPs post-human consumption, are bioavailable to the human system.

Assumption 3: Water and plastic have already equilibrated (which can take months), thus MPs contain concentrations of POPs similar to MPs measured in the ocean environment (Engler, 2012).

Assumption 4: The volume of microplastic sequestered from the surface (such that no chemical exchange of additives would occur) would be negligible due to the surface area: volume ratio of the particle. That is, all additives are assumed to desorb from the MP, regardless of their placement within the MP structure.

Assumption 5: Simple conversion of forward and reverse rate constants to percentages of observed maximum k values is a reasonable approximation for completeness of adsorption/desorption.

Sources of Contamination

Plastic toxicity comes from two main sources: additives and sorbed hydrophobic pollutants. Sorbed hydrophobic pollutants mostly consist of POPs and are chemicals of great concern globally. Their potential bioavailability to humans as a result of secondary MP consumption sparked the majority of MP studies being done today.

In addition to organic pollutants attracted to MPs and sorbed to their surface, plastics often contain toxic additives (NP, OP, BPA, and PBDE) that are intrinsic to the material, either mixed in or bound to the plastics' structure. These additives compose

about four percent of the plastic by weight (Brouwmeester et al., 2015). Their endpoints (endocrine disruption, carcinogenicity, and ADI values) are listed below. These can desorb from the plastic. As additives from the exterior diffuse out of the plastic, additives from the interior are pulled toward the surface of the plastic.

For purposes of this experiment, additives are pooled with overall human toxin load. Even though some are not classified as POPs and can be metabolized more readily, almost all have endocrine effects or carcinogenic effects on humans, and the overall toxin burden is thus pertinent to a discussion on control of plastic pollution. However, once more is known about a specific POP or additive, it may be more helpful to model it separately and specifically.

Parameters

The rate and completeness of sorption of POPs to MPs are dependent upon a number of factors including:

- Concentration of POP in seawater
- MP residence time in seawater
- Difference in concentration of POP inside the organism (human or mollusk, in this case), versus upon the microplastic
- Chemical affinity of plastic to a specific POP (Bakir et al., 2014a)
- Temperature (Bakir et al., 2014a)
- pH (Bakir et al., 2014a)
- Residence time of MP in animal gut
- Size of microplastic (Teuten et al., 2009)
- Porosity of microplastic (Teuten et al., 2009)
- Extent of weathering and/or biofouling of the MP

Likewise, the release of plastic additives out of plastics inside the animal gut are dependent upon many factors:

- Type of bond holding additive to plastic (covalently bonded additives are unlikely to desorb) (Teuten et al., 2009).
- Temperature (Teuten et al., 2009)
- pH (Teuten et al., 2009)

- Pore diameter of plastic in relation to size (molecular weight) of the additive (Teuten et al., 2009). Smaller additives move more readily.
- Co-migration
- Type of bond holding additive within plastic structure

(For more information on additives and their inclusion in this model, see Appendix 3.)

The mussel gut condition as described by Bakir et al., (15mM sodium taurocholate, 18 deg. C, pH= 7.5-8.4) was used to estimate first-order rate constant (k) values for desorption of POPs in the bivalve gut. Likewise, human gut condition as described in Bakir et al. (2014) (15mM sodium taurocholate, 38 deg. C, pH=4) was used to determine rate constants for POP desorption under those conditions.

Within the cold-blooded gut, desorption rates (k, per day) ranged from 0.27 +/- 0.1 to (PE -DEHP) to 3 +/- 0.87 (PE-Phe). (Notice there are no reverse rate constants reported in this experiment as none were observed, and experimental conditions would not allow for it). This range is used in Simulations 2 through 7, which eliminates in silico models (Koelmans et al. and Guin et al.) Within the warm-blooded gut, desorption rates (k, per day) ranged from 0.54 +/- 0.2 (PVC-DDT) to 12.10 +/- 2.09 (PE-Phe). These values are likewise used in Simulations 2 through 7.

Within Bakir et al.'s in vitro experiment, four POP-MP combinations were statistically lower than other combinations in terms of Kd (desorption constant) value (PFOA-PVC, PFOA-PE, Phe-PVC and DEHP-PVC), while three were significantly higher in terms of Kd value (PVC-DDT, PE-DEHP, PE-DDT, and PE-Phe). The latter are most concerning combinations in terms of transport and release of contaminants to warm-blooded animals in the marine environment (Bakir et al., 2014a). Note that the highest Kd value combination, PE-Phe, desorbs the most from plastic in both the cold-blooded gut and the warm-blooded gut conditions. In all cases, contaminants desorbed more readily in the warm-blooded gut than in the cold-blooded gut, and the surfactant enhancement over seawater is consistent (1.2 to 7 times enhancement in cold-blooded animals and 2.1 to 31.3 times enhancement over seawater in warm-blooded animals). No instances of the reverse reaction (cleaning effect) are reported, because simulated gut conditions in this experiment did not include a baseline toxin load (Bakir et al., 2014a). Thus, these rate constants may be accurate for a "clean" organism.

For the purpose of this model, only ranges of observed chemical concentrations of POPs on MPs collected from the ocean were used. This model represents a daily dose of MP from consumption of mussels. Desorption constants in the mussel portion are designed such that they represent sufficient residence time to reach equilibrium within the mussel gut as well as the human gut.

- Concentrations of sorbed POPs on MPs from Teuten et al., 2009 and Brouwmeester et al, 2015: (organic contaminant concentrations measured on marine plastics):
 - PCBs: 1ng/g to 200 ng/g
 - DDE: 0.1 ng/g to 60 ng/g
 - PAH: 10 ng/g to 1000 ng/g
 - PBDE: 0.4 ng/g to 57 ng/g
 - NP: 20 ng/g to 3,000 ng/g
 - OP: 0.4 ng/g to 8 ng/g
 - BPA: 5 ng/g to 300 ng/g
- Many BPA, NP, and PBDEs are from additives (Teuten et al., 2009).
- While not unequivocal, this study suggests that all contaminants were measured in the same sample (marked “North Pacific Central Gyre”. Therefore, it is reasonable to assume that any combination of these measurements are found in any one sample. Were these additive, a low of 11.5 ng POPs/g and a high of 1,310 ng POPs/g would be achieved. Likewise, (using NP, OP and BPA as additives), total additive loads are 25.4 ng additive/g and a high of 3,308 ng/g.
- MP dose information: A 300g average portion of mussels contains 300 plastic particles (about 1.5 µg of plastic) (Van Cauwenberghe et al., 2014).
- However, the actual MP load in humans is likely much higher due to exposure from other food sources such as honey and beer (Brouwmeester et al., 2015)
- Simulation 1: “Dirty Organisms”. This simulation uses k1 and k2 values (translated into positive percentages of total desorption, and negative percentage of total adsorption, respectively) from Koelmans et al. (2013) for mussels, and k1

values from Bakir et al. (2014) for humans. Model is run at 2,000 iterations for all simulations.

- Simulation 2: “Clean Organisms”. This simulation uses k_1 values from Bakir throughout.
- Simulation 3: “Observed Bioaccumulation”. Per Avio et al., 2014, tissue concentrations in bivalves can accumulate to as much as three times the concentration of the particles. Therefore, the high range of desorption rates of pollutants in the mussel is set to 100% in Simulation 3. Other values are the same as Simulation 2.
- Simulation 4: “Dirtier Plastics” uses the highest observed pollutant concentrations which are still realistic to support mussel growth. High end of PCB concentrations: marine PS microbeads in New England contained up to 5,000 ppb (5,000 $\mu\text{g/L}$ PCBs equivalent) (Engler, 2012). High additive concentrations: NP was measured in PP up to 16,000 $\mu\text{g/L}$ (Engler, 2012). High concentrations of DDTs: 64.4-87.7 ppb ($\mu\text{g/L}$) (Engler, 2012) in South Brazil. In the Pacific, DDTs are at higher concentrations of 22-7100ppb because DDT is still used to control mosquitos in this region (Engler, 2012). High PAH scenario: PAH concentrations of 39-1200 ppb on microplastics (up to 9,297 ppb near oil spills (Engler, 2012). This scenario uses fixed values for toxin loads: 21,485 $\mu\text{g/L}$ POPs and 16,000 $\mu\text{g/g}$ additives. (Note these are still an order of magnitude lower than highest reported POP values.) Simulation 4 uses k value ranges from Simulation 3.
- Simulation 5: “Welcome to 2050”. Daily dose of MPs consumed is increased by the same percentage by which total plastic pollution is expected to expand in the ocean: an additional 52% (Ocean Conservancy, 2015), or 2.25 μg plastic. Simulation 5 uses k values from Simulation 3, and normal toxin concentrations.
- Simulation 6: “How Big a Dose of Mussels (and their consumed MPs) Would it Take to Get a Non-Negligible Dose of Organic Pollutants in a Human?” Simulation 6 uses k values from Simulation 3.
- Simulation 7: “Recalibrating k .” Uses k values from Koelmans et al. for the mollusk portion, but instead of calibrating k values to a maximum of 100/day,

they are calibrated to a maximum of 10/day (a rate also referred to as ‘high’ and ‘fast’ by Bakir and Koelmans et al.).

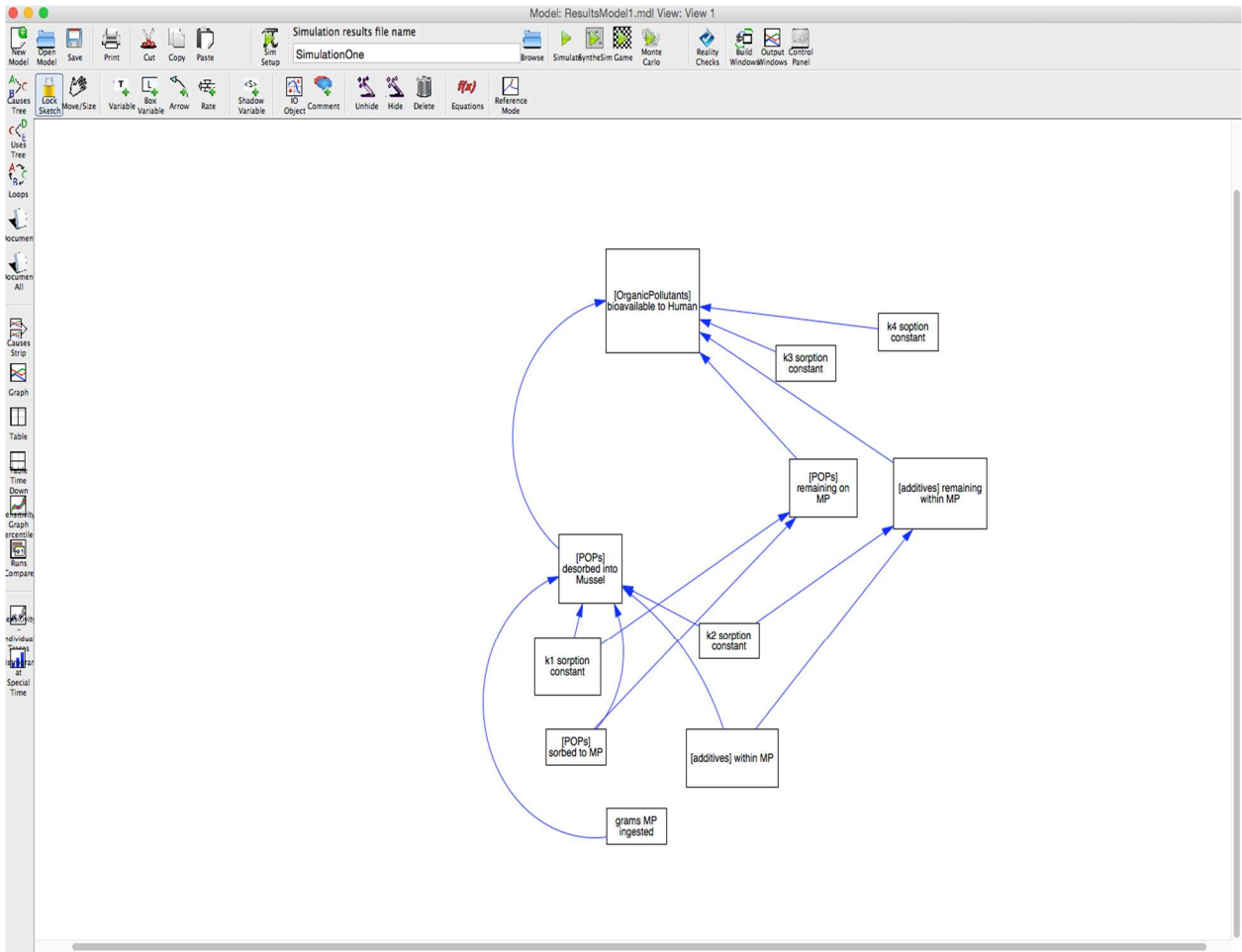


Figure 1: Generalized model, for visualization purposes, of secondary consumption.

Here, toxin loading pertains to human consuming blue mussels which have in turn consumed microplastics. This model was created using *Vensim PLE Plus* software.

Explanation of Variables:

In the secondary consumption model, POPs sorb or desorb in two stages: inside the mollusk gut and inside the human gut.

k1, k2, k3 and k4 are used to indicate percentage disassociation (or association) of the POP from (or to) the microplastic bead.

k1: percentage of POPs desorbed from MPs inside the mollusk gut.*

k2: percentage of additives desorbed from MPs inside the mollusk gut.*

k3: percentage of remaining POPs desorbed from MP inside the human gut.*

k4: percentage of remaining additives desorbed from MP inside the human gut.*

*All kn values can be negative, symbolizing resorption onto the MP bead.

Concentration of POP sorbed to MP and concentration of POP additives remaining within MP are varied in the Monte Carlo simulation, and have units ng/g microplastic. Amount of microplastic ingested is constant per scenario.

[POPs] desorbed into Mussels (that is, the blue mussel used in many microplastic experiments) is defined as [POPs sorbed to MP]*k1 + [additives within MP]*k2.

Likewise, “[POPs] bioavailable to Human” is defined as:
‘[POPs] desorbed into Mussel+ (k3* [POPs remaining on MP])+ (k4* [additives] remaining within MP).

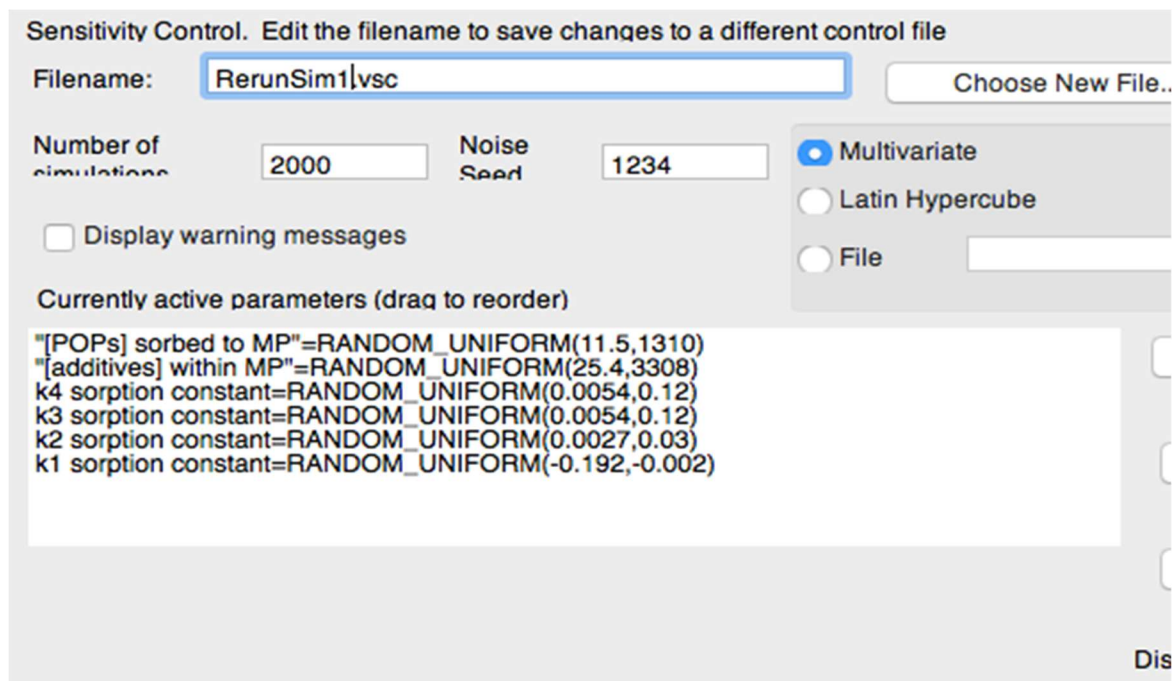


Figure 2: Ranges of variables used for the Monte Carlo simulation “Simulation 1”.

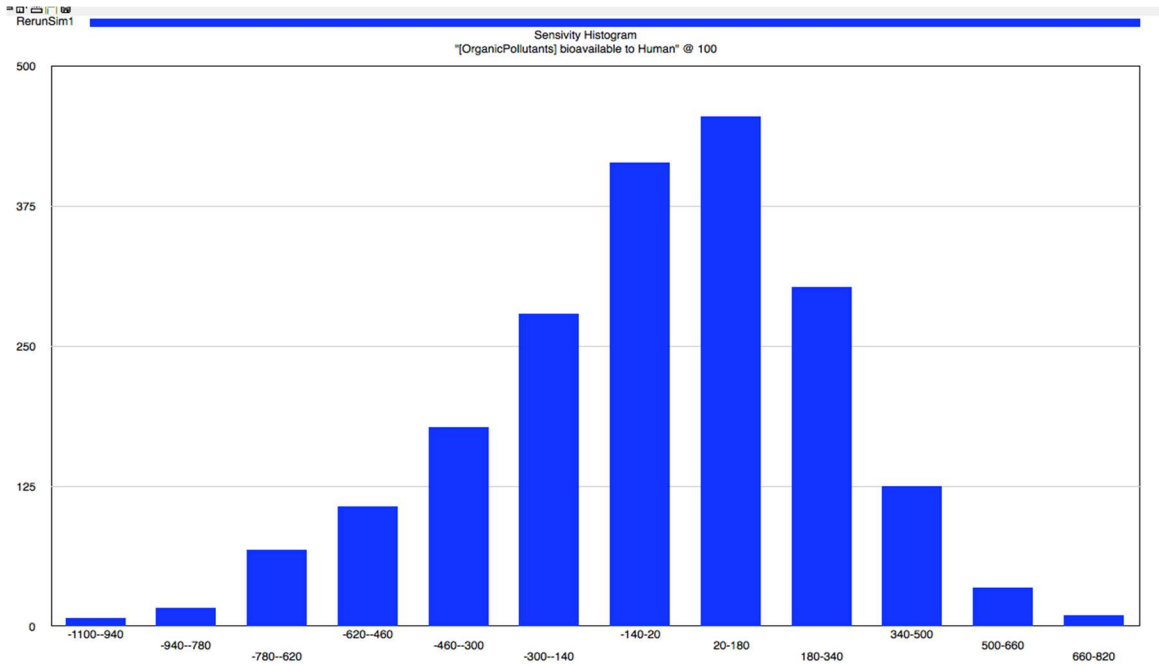


Figure 3: Results of Simulation 1, predicting probable toxin loads to the human body. Measurements (x-axis) are in nanograms (ng) of organic pollutant, while y-axis is number of runs. All distributions were set to ‘Random Uniform,’ in order to encompass all known values and not insert bias.

The most likely scenario in Simulation 1 is between 20 and 180 ng of human toxin load, supporting the null hypothesis.

Sensitivity Simulation Setup

Sensitivity Control. Edit the filename to save changes to a different control file

Filename:

Number of simulations: Noise Seed:

Display warning messages

Currently active parameters (drag to reorder)

```

"[POPs] sorbed to MP"=RANDOM_UNIFORM(11.5,1310)
k2 sorption constant=RANDOM_UNIFORM(0.0027,0.03)
k3 sorption constant=RANDOM_UNIFORM(0.0054,0.12)
k4 sorption constant=RANDOM_UNIFORM(0.0054,0.12)
"[additives] within MP"=RANDOM_UNIFORM(25.4,3308)
k1 sorption constant=RANDOM_UNIFORM(0.0027,0.03)

```

Multivariate
 Univariate
 Latin Hypercube
 Latin Grid
 File

Distribution

Parameter:

Model Value	Minimum Value	Maximum Value
--	<input type="text"/>	<input type="text"/>

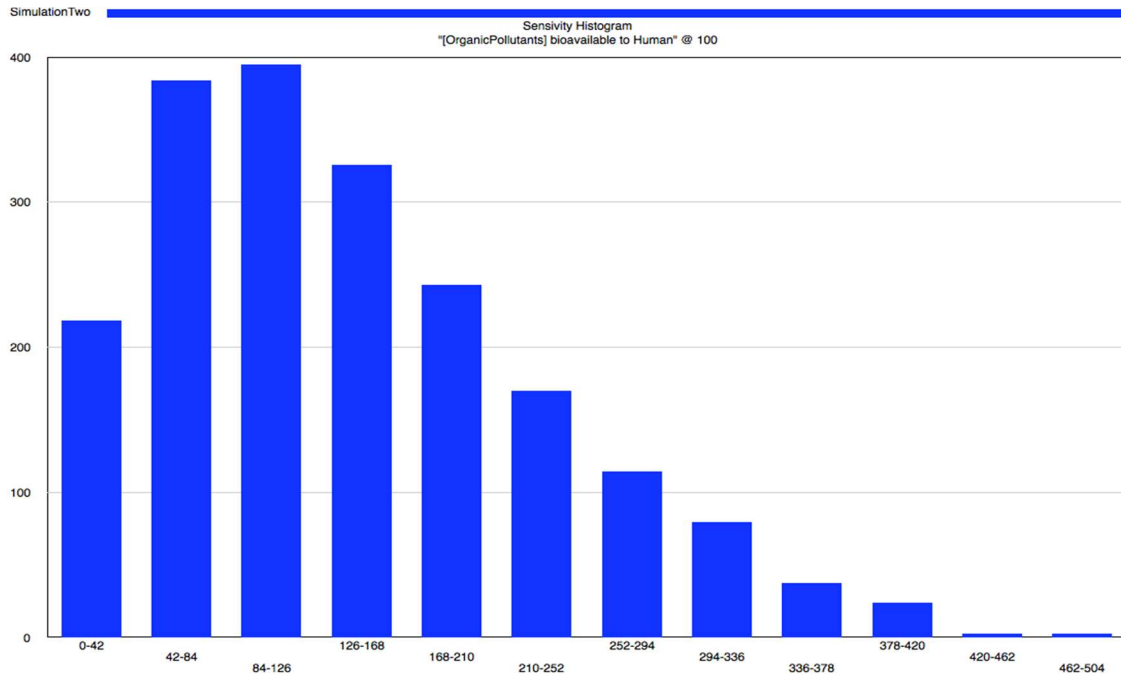


Figure 4: Parameters and Results of Simulation 2.

Simulation 2 supports the null hypothesis.

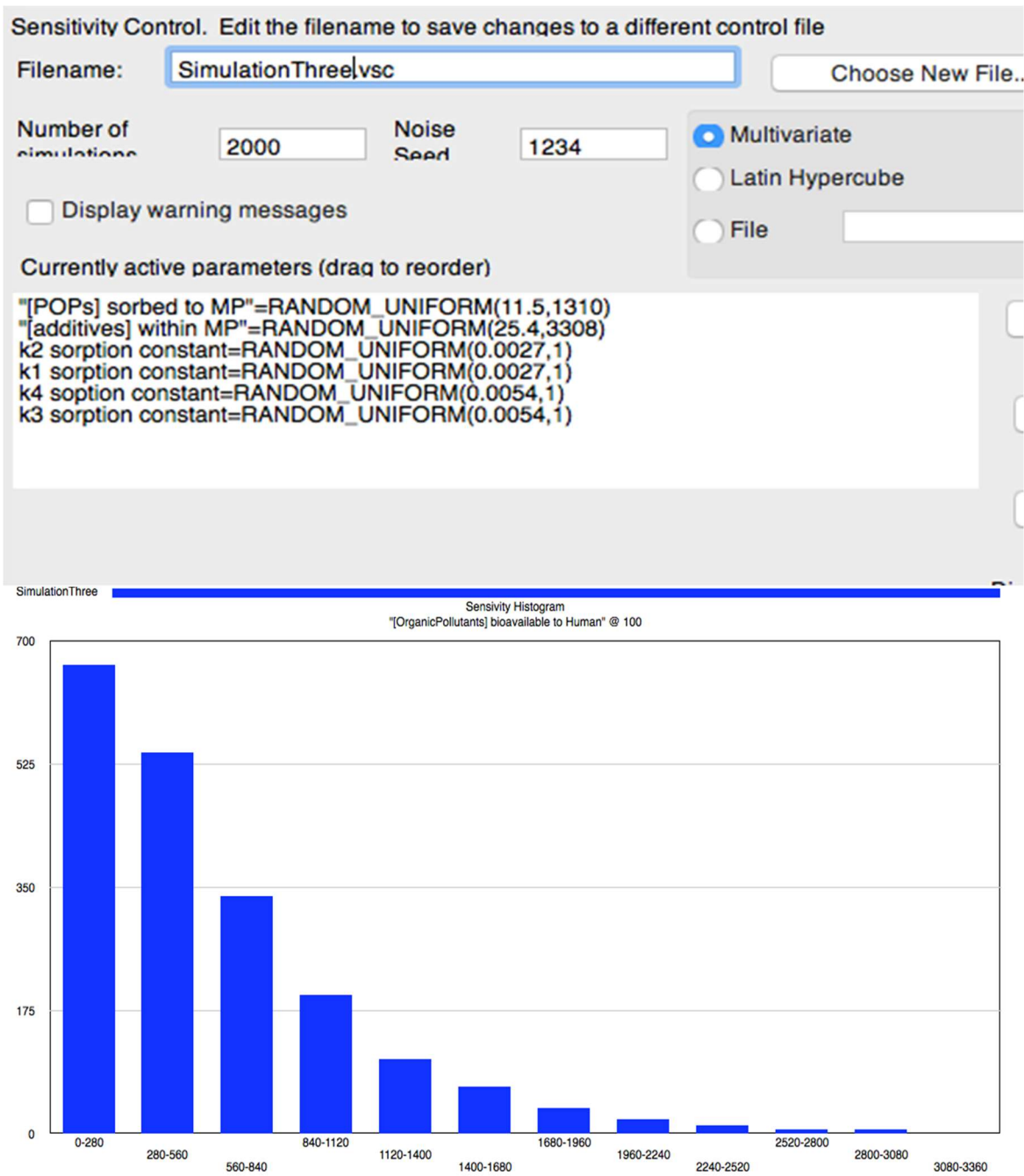


Figure 5: Parameters and Results of Simulation 3.

Simulation 3 supports the null hypothesis.

Sensitivity Simulation Setup

Sensitivity Control. Edit the filename to save changes to a different control file

Filename:

Number of simulations: Noise Seed:

Display warning messages

Multivariate
 Latin Hypercube
 File

Currently active parameters (drag to reorder)

```

k2 sorption constant=RANDOM_UNIFORM(0.0027,1)
k1 sorption constant=RANDOM_UNIFORM(0.0027,1)
k4 sorption constant=RANDOM_UNIFORM(0.0054,1)
k3 sorption constant=RANDOM_UNIFORM(0.0054,1)

```

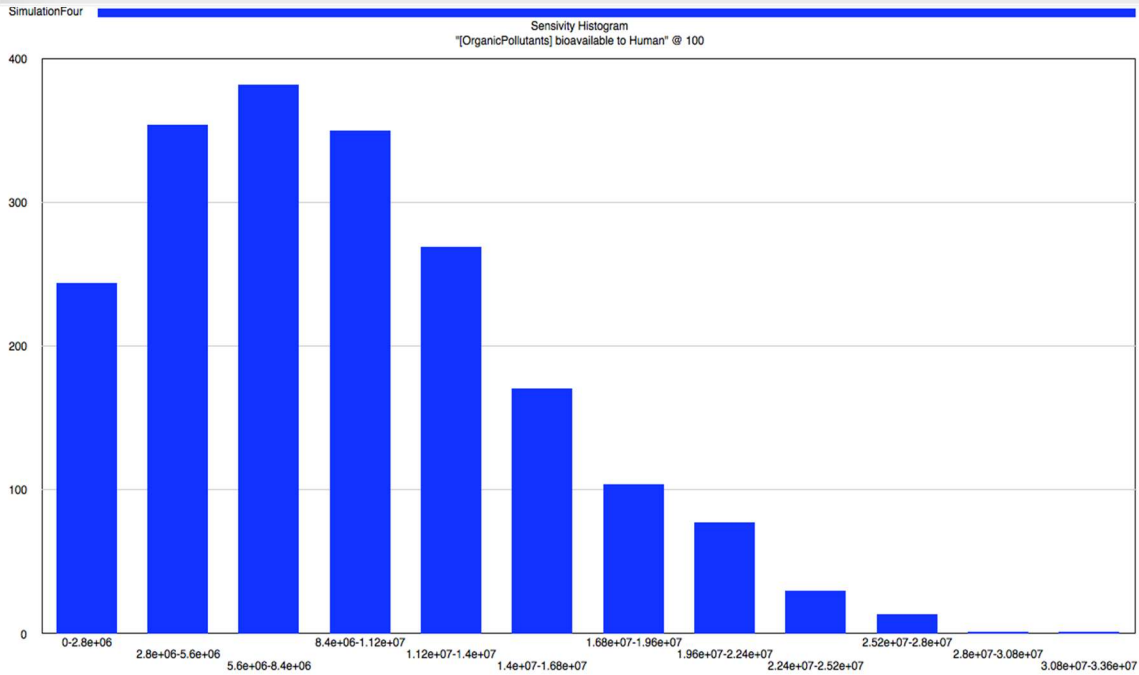


Figure 6: Parameters and Results of Simulation 4.

In Simulation 4, the most likely scenario puts total human toxin load at around 5,000,000 ng, or 5 mg, supporting the toxin loading hypothesis.

Sensitivity Control. Edit the filename to save changes to a different control file

Filename:

Number of simulations: Noise Seed:
 Multivariate
 Latin Hypercube
 File

Display warning messages

Currently active parameters (drag to reorder)

```

k2 sorption constant=RANDOM_UNIFORM(0.0027,1)
k1 sorption constant=RANDOM_UNIFORM(0.0027,1)
k4 soption constant=RANDOM_UNIFORM(0.0054,1)
k3 sorption constant=RANDOM_UNIFORM(0.0054,1)
"[POPs] sorbed to MP"=RANDOM_UNIFORM(11.5,1310)
"[additives] within MP"=RANDOM_UNIFORM(25.4,3308)

```

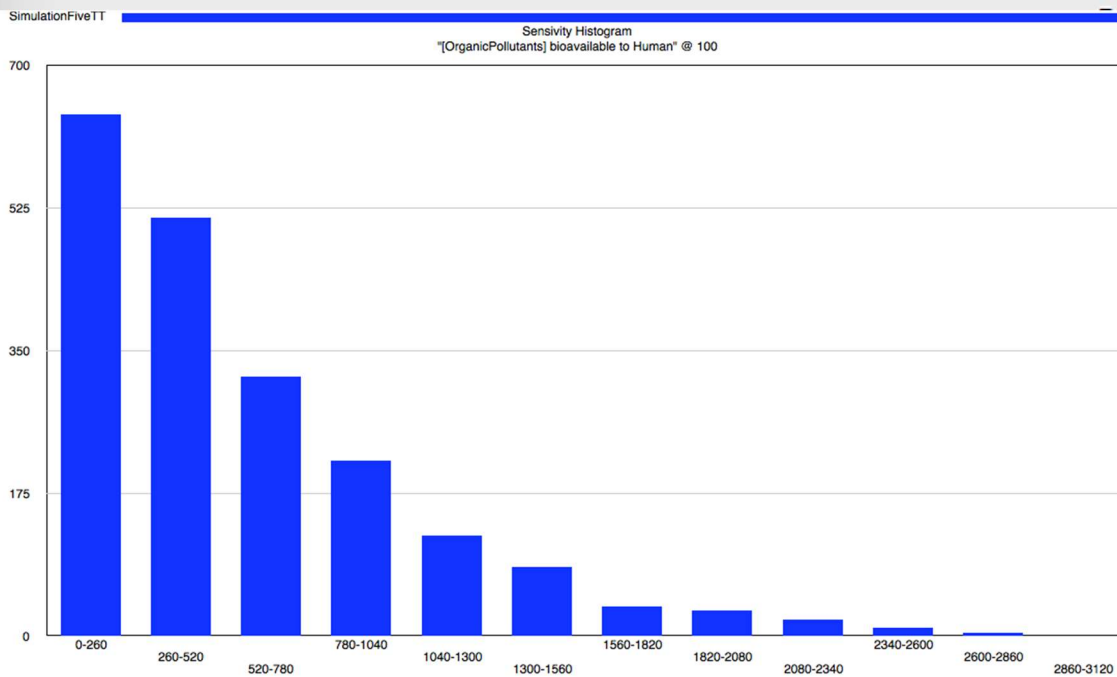


Figure 7: Parameters and Results of Simulation 5.

Most likely outcomes of Simulation 5 support the null hypothesis.

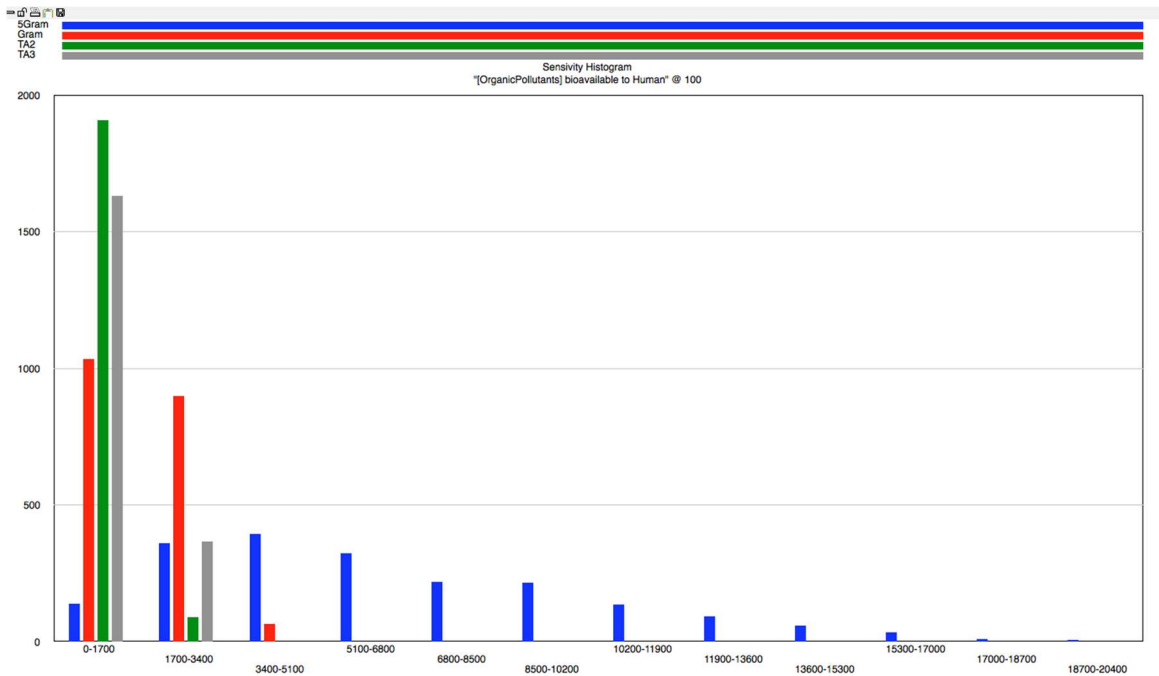


Figure 8: Results of Simulation 6.

Sensitivity graphs with 0.1g (“TA2”), 0.5g (“TA3”), 1g, and 5g plastic ingested. (For scale reference: Teuten et al. fed shearwater chicks a single 1g dose of PCB-laden MPs and saw bioaccumulation.) Only in the 5g simulation is a load in the μg range (most likely to be an effective dose) probable. The 5g ingestion scenario supports the toxin loading hypothesis.

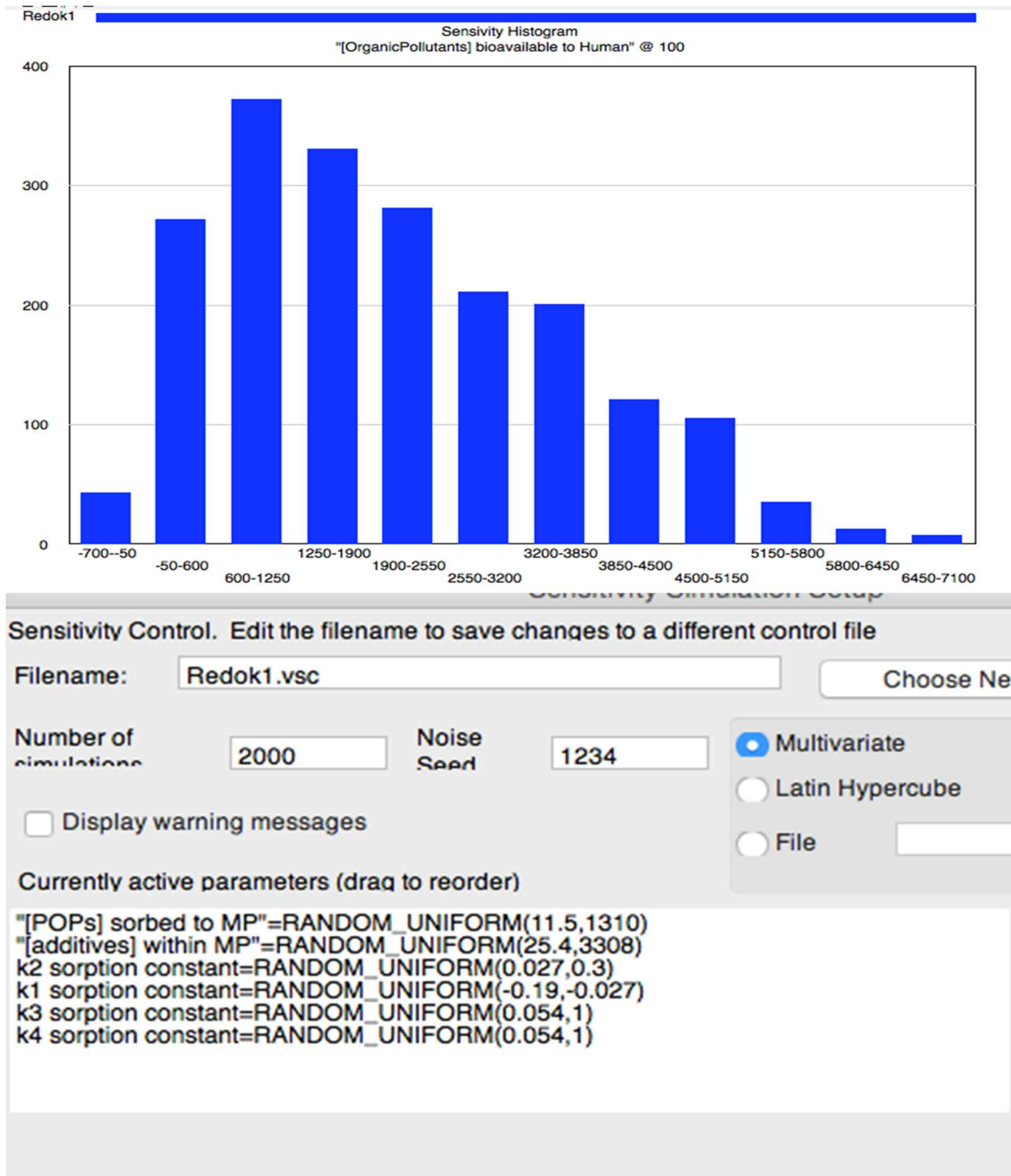


Figure 9: Parameters and Results of Simulation 7.

When k_1 values are calibrated to a maximum of 10/day rather than a maximum of 100/day, the results do not change much—human toxin loads are within the ng range, outside of one order of magnitude of ADL toxin loads for most toxins. This suggests that toxin loading inside the more facilitative human gut is more important than loading

within the mollusk gut for determining total human POP loads. Simulation 7 has the most likely human toxin load of 600-1250 ng, and supports the null hypothesis.

Relative Human Body Load, According to FDA

Chemical Effects and Dosage

Table 1
Toxicological guideline values established by EFSA and JECFA.

Element/species	Body	Year	Type	Value
Metals				
Mercury, inorganic	JECFA	2011a	PTWI	4 µg/kg bw/week
	EFSA	2012a	TWI	4 µg/kg bw/week
Methylmercury	JECFA	2003	PTWI	1.6 µg/kg bw/week
	EFSA	2012a	TWI	1.3 µg/kg bw/week
Lead	JECFA	2011b	PTWI	Withdrawn, no no-effect level identified
	EFSA	2010c	PTWI	Withdrawn, no no-effect level identified
Cadmium	JECFA	2011b	PTMI	25 µg/kg bw/month
	EFSA	2009c	TWI	2.5 µg/kg bw/week
Arsenic, inorganic	JECFA	2011a	BMDL _{0.5}	3 µg/kg bw/day (2–7 µg/kg bw/day)
	EFSA	2009a	BMDL ₀₁	0.3–8 µg/kg bw/day (lifetime risk)
Pharmaceuticals and personal care products				
Benzylpenicillin	JECFA	1990	ADI	< 30 µg/kg bw
Oxytetracycline	JECFA	2002	ADI	0–30 µg/kg bw
Emamectin	JECFA	2013	ADI	0–0.5 µg/kg bw
Derquantel	JECFA	2012	ADI	0–0.3 µg/kg bw
Flumequine	JECFA	2006a, 2006b	ADI	0–30 µg/kg bw
Carazolol	JECFA	1995	ADI	0–0.1 µg/kg bw
Dexamethasone	JECFA	2009	ADI	0–2 µg/kg bw
Tilmicosin	JECFA	1998	ADI	0–40 µg/kg bw
Triclabendazole	JECFA	1993	ADI	0–3 µg/kg bw
Tylosin	JECFA	2009	ADI	0–30 µg/kg bw
Avilamycin	JECFA	2009	ADI	0–2 µg/kg bw
Endocrine disruptors				
Bisphenol A	EFSA	2015	TDI	4 µg/kg bw
Estradiol-17β	JECFA	2000	ADI	0–0.05 µg/kg bw
Testosterone	JECFA	2000	ADI	0–2 µg/kg bw
Progesterone	JECFA	2000	ADI	0–30 µg/kg bw
Melengestrol acetate	JECFA	2001	ADI	0–0.03 µg/kg bw
PFOS	EFSA	2008a	TDI	150 ng/kg bw/day
PFOA	EFSA	2008a	TDI	1500 ng/kg/bw/day
Polycyclic aromatic hydrocarbons				
Benzo[a]pyrene	JECFA	2005	BMDL ₁₀	0.10–0.23 mg/kg bw/day
Benzo[a]pyrene	EFSA	2008c	BMDL ₁₀	0.07–0.20 mg/kg bw/day
Benzo[a]pyrene and chrysene (PAH2)	EFSA	2008c	BMDL ₁₀	0.17–0.45 mg/kg bw/day
Benzo[a]anthracene, benzo[a]pyrene, benzo[b]fluoranthene and chrysene (PAH4)	EFSA	2008c	BMDL ₁₀	0.34–0.93 mg/kg bw/day
benz[a]anthracene, benzo[b]fluoranthene, benzo[k]fluoranthene, benzo[ghi]perylene, benzo[a]pyrene, chrysene, dibenz[a,h]anthracene, and indeno[1,2,3-cd]pyrene (PAH8)	EFSA	2008c	BMDL ₁₀	0.49–1.35 mg/kg bw/day
Brominated flame retardants				
Pentabromodiphenyl ether	EFSA	2011a	LD ₅₀	2640–6200 mg/kg bw
Polybrominated biphenyls	EFSA	2010d	LD ₅₀	> 1000 mg/kg bw (single exposure) 64–150 mg/kg bw (after repeated exposure)
Hexabromocyclododecane	EFSA	2012b	NOEL	10 mg/kg bw (cited Ema et al. (2008))
Tetrabromobisphenol A	EFSA	2011b	BMDL ₁₀	16 mg/kg bw
1,2-Bis(2,4,6-tribromophenoxy)ethane	EFSA	2012c	LD ₅₀	> 10 g/kg for rats and dogs

ADI: acceptable daily intake; BMDL: benchmark dose lower limit of the 90% confidence interval; LD: lethal dose; NOEL: no-observed-effect level; PTMI: provisional tolerable monthly intake; PTWI: provisional tolerable weekly intake; TDI: tolerable daily intake; TWI: tolerable weekly intake.

Table 1: Image from Vandermeersch et al., 2015a: Acceptable Daily Intake Values set by EFSA and JECFA. This can be used as a reference for the overall human toxin load transferred to humans from microplastics.

POP	Global Historical Use/Source	Overview of U.S. Status
PCBs	Used for a variety of industrial processes and purposes, including in electrical transformers and capacitors, as heat exchange fluids, as paint additives, in carbonless copy paper, and in plastics. Also unintentionally produced during combustion.	Manufacture and new use prohibited in 1978 (TSCA). Regulated as a hazardous air pollutant (CAA). Priority toxic pollutant (CWA).
DDT	Insecticide used on agricultural crops, primarily cotton, and insects that carry diseases such as malaria and typhus.	Under FIFRA: No U.S. registrations; most uses canceled in <ul style="list-style-type: none"> • 1972; all uses by 1989. • Tolerances on food crops revoked in 1986. No U.S. production, import, or export. DDE (a metabolite of DDT) regulated as a hazardous air pollutant (CAA). Priority toxic pollutant (CWA).
dioxins and furans	Unintentionally produced during most forms of combustion, including burning of municipal and medical wastes, backyard burning of trash, and industrial processes. Also can be found as trace contaminants in certain herbicides, wood preservatives, and in PCB mixtures.	Regulated as hazardous air pollutants (CAA). Dioxin in the form of 2,3,7,8-TCDD is a priority toxic pollutant (CWA).

Table 2: Regulatory data for some POPs of interest to this study, from the EPA website, 2015.

Minimal risk levels proposed by ATSDR for the monitored substances

Name	Duration	MRL (mg/kg/day)	Factors of uncertainty	Endpoint
pp'DDT	Acute	0.0005	1000	Develop.
	Intermediate	0.0005	100	Hepat.
HCB	Acute	0.008	300	Develop.
	Intermediate	0.0001	90	Reprod.
	Chronic	0.00002	1000	Develop.
α -HCH	Chronic	0.008	100	Hepat.
β -HCH	Acute	0.2	100	Neur.
	Intermediate	0.0006	300	Hepat.
γ -HCH	Acute	0.01	100	Neur.
	Intermediate	0.00001	1000	Immun.
PCBs (Aroclor 1254)	Intermediate	0.00003	300	Neur.
	Chronic	0.00002	300	Immun.
Anthracene	Intermediate	10	100	Hepat.
Fluoranthene	Intermediate	0.4	300	Hepat.
Fluorene	Intermediate	0.4	300	Hepat.
Naphthalene	Acute	0.05	1000	Neur.
	Intermediate	0.02	300	Hepat.

Table 3: Minimal Risk Levels for various pollutants (POPs and PBTs) of interest to this study. Source: Binelli & Provini, 2003.

Important Pollutant Information: Endpoints

PVC: Has carcinogenic monomers (Rochman et al., 2013a).

Styrene: Neurological effects (ATSDR, 2010)

PE: Composes over 40% of MPs; toxic effects examined further in Discussion.

Contaminants of Emerging Concern: No maximum levels have been laid down in EU legislation. Always evaluated by persistence, bioactivity, and bioaccumulation (Vandermeersch et al., 2015a).

PBTs: Bioaccumulate in food webs, associated with fish population decline and endocrine disruption (Rochman et al., 2013a). Intake of fish and shellfish is a predictor of serum PBTs in pregnant women. Usually found at ng/g to μ g/g concentrations. (Teuten et al., 2009)

POPs:

DDTs: Carcinogenic. Linked to infertility, miscarriages. Developmental, nervous system, and hepatic effects.

Organochloride Pesticides: Target nuclear hormone signaling pathways.

(Vandermeersch et al., 2015a)

PBDEs: Neurological, thyroid, sex hormones, reproduction (rodents). Suspected thyroid disruptor in humans and wildlife (Teuten et al., 2009, Vandermeersch et al., 2015), (WHO/IPCS 1994). No legislation exists regarding PBDEs in seafood, while BDE-47 is often one of the most concentrated seafood pollutants (Vandermeersch et al., 2015a). In the 2010 Conference of the Parties of Stockholm convention, PBDEs were placed on a list of POPs for strict elimination (Priority POPs are under EU regulation #850/2004 (ECR)) (Vandermeersch et al., 2015).

PBDEs bioaccumulate and biomagnify, and can be transported long distances on microplastics (much like PCBs). Unfortunately, even with worldwide elimination of brominated flame retardants, they would persist in the environment for many years. Brominated flame retardants can be found mixed into plastics (e.g. polyurethane foams) or reacted to the plastic (e.g. epoxies or polystyrene). PBDE exposure is of particular concern to children with a developing brain.

PAHs: PAHs are easily biodegradable (Rochman et al., 2013a), however, their metabolites oxy-PAHs and alkylated PAHs are often more toxic than the parent compounds, inducing oxidative stress and endocrine disruption (Vandermeersch, 2015a). Endpoints: many are carcinogenic, and cause developmental toxicity. (Engler, 2012). They are often mutagenic as well (Vandermeersch, 2015a). PAHs have low metabolic activity in bivalves, and therefore accumulate within them (Binelli et al., 2003). In animal studies, chronic PAH ingestion had adverse effects to the cardiovascular, respiratory, gastrointestinal, immune, and central nervous system. PAHs are a possible human carcinogen (ATSDR, 1995). Bivalves do not metabolize PAHs readily, and therefore transport of these chemicals to humans via bivalves may have a profound effect upon human health, where it only had a negligible effect upon the bivalve (Binelli & Provini, 2003).

PCBs: Banned in many countries. EU tolerable weekly intake: 14 pg/kg body weight OR daily intake: 2pg/kg/day. (Extremely low). There is some concern for PCB toxicity at current level of PCB & plastic ocean pollution, and there is no room for an increase in

concentration (Engler, 2012). FDA maximum contamination of PCBs is 2µg/g w.w. for bivalves (Binelli, 2003).

Average exposure to non-dioxin like PCBs is 0.3-1.8µg/day for a 70 kg person, according to the European Food Safety (Brouwmeester et al., 2015).

The geometric mean of PCB body burden in humans is 103.6 ppb (µg/g).

According to a US national Health & Nutrition Examination Survey, diet is the dominant source of PCB body burden (Brouwmeester et al., 2015).

Additives

Alkylphenols: NP & OP have estrogenic properties. They are ubiquitous and endocrine disrupting. NP & OP are now included as priority pollutants in the European Water Framework Directive (2000) (Vandermeersch et al., 2015a). The concentration of concern for aquatic species is 0.7 ppb (0.7 µg/L) in water.

BPA: An endocrine disruptor, toxic to wildlife and humans (Van Cauweberge & Janssen, 2014). NOAEL for aquatic organisms is 8 ng/L water, after which malformation of female organs in freshwater snails occur. BPA is estrogenic in humans (Vandermeersch et al., 2015a), and thus could be associated with reproductive cancers and fertility problems.

In humans, BPA targets nuclear hormone signaling pathways. It is relatively hydrophilic, and degrades completely (mineralizes) only in aerobic conditions.

Organotin: Deterioration of human immune function and endocrine disruption (Teuten et al, 2009)

Phalates: Some phalates reduce testosterone (Teuten et al., 2009). Phalates target nuclear hormone signaling pathways. They are also endocrine disruptors. Phalates are toxic to wildlife and humans (Van Cauweberge & Janssen, 2014).

DISCUSSION

Conclusions from the Model

Of the seven scenarios run, only two supported the hypothesis of human toxin loading within an order of magnitude of most organic pollutant effective doses. Five scenarios supported the null hypothesis: that no toxin loading occurred within an order of magnitude of most effective organic toxins' ADI. While there were occurrences within the simulation of net cleaning effect, no scenario tested predicted this as a most likely outcome of secondary MP consumption.

The two simulations which resulted in net toxin load within an order of magnitude of ADIs were Simulation 4 and Simulation 6. Simulation 4 represents consumption of highly POP-loaded MPs, and uses toxin values on MPs from the areas of highest-measured POP levels on MPs. Simulation 6 represents consumption of a high dose of MP (5g), using k conditions of observed bioaccumulation rather than theoretical cleaning effect.

The *Vensim* simulation supports previously published rough estimates, which have claimed that human secondary consumption of microplastics does not result in a high toxin load, compared to ADIs of those toxins in humans (Brouwmeester et al., 2015). However, there are scenarios in this model which do support the human toxin loading hypothesis, and they may not be unrealistic in the near future. Human exposure to MPs is increasing, and MPs have been found in foods other than seafood, such as beer and honey (Brouwmeester et al., 2015 and sources cited therein). Furthermore, a continual increase of plastics (and thus microplastics) in the ocean is expected, following a business-as-usual scenario. The case of higher toxin loading is also not unrealistic. Unless a worldwide ban on certain organic compounds – including but not limited to POPs—is achieved, concentrations as high as those tested in Scenario 4 may become more common in areas without a POP ban. Since higher microplastic concentrations and higher POP concentrations are associated with more densely populated areas, these areas are likely to suffer most from seafood MP contamination (Van Cauwenberghe et al., 2015). Today, however, it is important to understand the combined effects of MPs, not only on toxin loading within organisms but on physiological and immunological responses.

Utility of the Model

The stochastic model presented above has a number of drawbacks, yet it maintains some utility. For example, a case can be made that the system in question is not inherently stochastic, and therefore may be better modeled by a system of stochastic partial differential equations, either as a function of time or in a steady state (Professor William Bossert, personal communication, 2016). A sensitivity analysis of this dynamical system within the same parameters, Prof. Bossert maintains, would be more convincing. A version of the same system as a set of stochastic partial differential equations in MATLAB may therefore be a next step to this modeling process.

However, to the best of this author's knowledge, estimates as to human toxin loading as a result of MP consumption have been only rough approximations in the past, and have not taken into account the range of variables—or uncertainties—which affect the buildup of toxins within the animal system. What has been presented here therefore may represent the first such system, providing an initial building block for more robust models of its kind once these biological systems are better understood.

Data from literature on animal toxin loading from MPs are still very limited, and a simulation is rarely indicative of the actual behaviors of an animal's body. Rather, natural systems are complicated and, as many modelers repeat: “no model is correct, but many are useful.” This is a systems model taking into account data for sorption of multiple POPs to multiple types of microplastics, although of course its parameters could easily be altered to model the effects of one pollutant-MP type pair.

Though ADI values for the organic pollutants in question are published, average human toxin load for these chemicals was not accounted for by this study by way of kinetic chemical gradient modeling. Though it has been suggested that a high toxin load within a human with respect to the toxin concentration on an MP may lead to a cleaning effect, this hypothesis has not been tested in mammals, nor has it been supported in consumers of seafood (e.g. birds) (Teuten et al., 2009). This model assumes that bioaccumulation is not only possible but probable in predators (including humans) based upon Teuten et al.'s experimentation in birds with a baseline toxin load. However, this is an untested theory in mammals, and therefore further experimentation is suggested below, along with experimental design.

Many of the conclusions formed by previous researchers differ in a predictable way that is correlated to experimental design. For example, Koelmans et al. (2013 and 2015) and Gouin et al. (2011) conclude that microplastics have an overall cleaning effect, and that no risk management is necessary surrounding transfer of POPs via MPs. Both used theoretical modeling to reach this conclusion, with limited *in vitro* studies in lower-trophic level organisms. Koelmans et al. used an *in vitro* study on lugworms to correlate their model. In contrast, while the cleaning effect has been confirmed in contaminated organisms with respect to completely clean, virgin plastic, most other studies on higher animals have had a higher instance of showing bioaccumulation effects from contaminated microplastics. This effect was also observed in organisms with a previous toxin load within their bodies (Teuten et al., 2009).

Synergistic effects, immunological perturbations, and physical harm caused by MPs POPs are not accounted for in the proposed model. Immunological and physiological effects are discussed further below.

Physiological & Immunological Effects of Microplastic Ingestion in Bivalves, Fish, and Higher Organisms.

Cellular Effects

Avio et al. (2014) measured cellular effects in terms of changes in immune responses, lysosomal activity, peroxisomal proliferation, antioxidant production, neurotoxic effects, genotoxicity, and changes in gene expression within test subjects *Mytilus galloprovincialis*. Similar indicators were used by Ferreira et al. (2016) on a marine fish. Avio et al. (2014) used a DNA microarray, among other tools, to test these cellular effects, and found that there is a risk of virgin and contaminated MPs having toxicological risk in terms of transcriptional and cellular responses. In fact, the immunological responses in this experiment were mostly a result of plastic ingestion itself, and not of the associated chemicals.

Significant enhancement of DNA strand breaks was observed in the hemocytes of mussels treated with virgin MPs (PS and PE). PS and PS-Pyrene treatments showed changes in several DNA-repair genes (Avio et al., 2014). It is not clear what role desorbing plastic monomers have in this response if any.

Numerous cellular oxidative markers were examined in *Mytilus* by Paul-Pont et al., (2016), who observed modulation of cellular oxidative balance as a result of micro-PS ingestion alone.

Canesi et al. (2015) observed cytotoxicity in hemocyte cells of *Mytilus galloprovincialis* at highest tested concentrations (50 µg/L) of polystyrene nanoparticles (50 nm). PS-NH₂ nanoplastics also induced apoptotic processes, which were evaluated by flow cytometry. Decreased phagocytotic activity, increase in lysozyme activity, and increase in extracellular (ROS) and NO (nitric oxide) production were also observed, with maximum effects at *lower* concentrations (Canesi et al., 2015). Paul-Pont et al. (2016) likewise found significant changes to cellular oxidative balance markers and increased hepatocyte mortality as a result of MP treatment alone in mussels, with greatest effects seen with a combination of fluoranthene-associated microplastic.

In fish, single-cell necrosis was significantly higher if fed marine plastic than in fish fed no plastic, under environmentally relevant conditions (Rochman et al., 2013a). Enhanced Reactive Oxygen Species (ROS) production was observed in response to MP ingestion (from which the researchers hypothesize DNA strand breaks result) (Rochman et al., 2013a). Acetylcholinesterase (AChE) levels were used to measure neurotoxic effects in mussels (Rochman et al., 2013a), and could be used as a neurotransmitter proxy in mammals in the proposed experiment below.

Immunological Effects

In marine invertebrates, the immune function suffers as a result of exposure to PS nanoplastics (Canesi et al., 2015). Mechanisms are similar to those in mammalian cells.

Granulocyte formation was used by von Moos et al. (2012) to measure inflammation as a result of MP ingestion by *Mytilus edulis*. Granulocytes are a collection of immune cells whose job is to wall off foreign tissue the body cannot eliminate. These are visible via histological observation. Lysosome formation and the stability of the lysosome were also measured. Granulocytomas formed after 6 hours and lysosomal membrane destabilization increased with increased exposure to PE particles 0-80µm in size (von Moos et al., 2012). Immuno-modulation in mussels due to uptake of PS beads was observed by Paul-Pont et al. (2016), as well as impairment of *Mytilus* metabolism.

Microplastic ingestion by fish *Oryzias latipes* was associated with severe depletion of glycogen stores, which was linked to the energy cost of detoxification (Rochman et al., 2013a).

In humans, it has been theorized that MP consumption would lead to gut inflammation, but that these effects would be limited only to the gut (Brouwmeester et al., 2015). However, translocation of MPs within the human system and inflammatory or cytotoxic responses as a result of MP consumption have not been disproven.

Physical Effects: Translocation

Browne et al. (2008) found that 3-9.6 μm -sized MPs accumulated in the digestive tissue and translocated to the hemolymph (circulatory fluid) of the blue mussel *Mytilus edulis*. Cilia movement in the stomach, intestine and digestive tubules partially determines MP endpoints and accumulation in the lysosomal compartments of *Mytilus*, while MP collection on the gills is aided by microvilli there, and by endocytosis (Avio et al., 2014). MPs persist in the digestive tract of *Mytilus* for 12 or more days, and hemolymph for 48 or more days after a single exposure (Browne et al., 2008, unpublished data reported in Santana et al., 2016).

MP and nanoplastic translocation has also been observed by a number of other researchers since. In Avio et al.'s (2015) experiment on *Mytilus spp.*, tissue localization of MPs to hemolymph, gills and most of all digestive tissues was observed. Digestive tissues showed an accumulation of pyrene. Avio et al. (2015) observed MPs translocated to the hepatic tissue of oceanic mullet.

In *medaka* fish, ingestion of microplastic was linked to hepatic stress, while fish exposed to both MPs and sorbed contaminants showed the greatest hepatic stress (Rochman et al., 2013a).

According to Hussain et al., the uptake of a large number of inert particulate matter--especially nanoparticles-- has been observed in the intestinal Peyer's patches in various animals including dogs and rats. The Peyer's patches (intestinal lymphatic tissue) seem to be indiscriminate as to the type and size of particles absorbed (Hussein et al., 2001).

Translocation across the mammalian gut has been proven in humans for particles 0.2 to 150 μm (Hussein et al., 2001), though not specifically for plastics. In dogs, PVC

particles of 5-110 μ m translocated to the liver (Brouwmeester et al., 2015 and Volkheimer et al., 1975).

Physical Effects: Detoxification Modulation

Paul-Pont et al. (2016) found that after marine mussel depuration following exposure to fluoroanthene and micro-PS, there was a higher concentration of fluoroanthene in mussels with micro-PS and FLU compared to mussels treated with only FLU alone (Paul-Pont et al., 2016). Their results suggest physiological mechanisms that affect detoxification in the marine mussel. Their primary hypothesis surrounding this phenomenon was that micro-PS caused the observed, associated down-regulation of P-glycoprotein involved in pollutant excretion. However, impairment of filtration activity and/or presence of remaining beads in the gut were not ruled out as causes for decreased detoxification (Paul-Pont et al., 2016).

Nanoplastics: Effects and Concerns

Nanoplastics are widely understood to have greater potential for translocation than larger particles. Their abundance in the marine environment has yet to be confirmed due to the difficulty in enumerating and visualizing them. However, Dr. Tracey Mincer and others at Woods Hole Oceanographic Institute are in the process of enumerating and tracking oceanic microplastics and their ecosystem effects.

Perhaps even more concerning is the possibility of nanoplastics crossing the blood brain barrier. Wick et al. (2010) demonstrated the uptake of 240nm fluorescent polystyrene particles by the human placenta *ex vivo*.

Intense research is underway on using nanoparticles (usually biodegradable) to transport drugs. Results of these studies and of those by ocean scientists like Dr. Mincer will shed light on the relative importance of nanoplastics to the human system and to the oceanic food web.

CONCLUSIONS & RECOMMENDATIONS

Plastics are an important material for global societies, allowing for tremendous advances in fields from medicine to food storage capacity. However, some of the most abundant uses of plastics have costs to global ecosystems and human health that outweigh the convenience of plastic as a material. In particular, the food industry's

widespread implementation of single-use plastic containers represents a tremendous opportunity for implementation of more suitable materials. The impact of microplastics from the breakdown of marine plastic litter has become noticeable in the human food chain, and is now both a cause for concern and a warning that a substitution of more intelligent materials choices is paramount to the health of our species and many others.

Ninety percent of human POP exposure is from food, and of this, about 90% of POP load from food is from animal products (Binelli & Provini, 2003, Safe, 1998, Schlummer et al., 1998, and Furst et al., 1990). Despite the small percentage of the human diet that seafood occupies, it is a major route of POP contamination to the human body (Binelli et al. and sources cited therein). Half of all plastics produced each year are polyolefins (PE and PP) used primarily for single-use packaging, which is designed to end its life in a landfill (Browne et al., 2010). However, these materials are light and volatile enough that they often end up in aquatic environments, and ultimately the ocean. According to the NGO Ecosurf, 32% of all plastic packaging ends its cycle in natural ecosystems, and 8 million tons of plastic are dumped in the ocean each year. Ecosurf's estimate of total load of ocean plastic is 150 million tons of plastic globally.

Because of the facilitated uptake of POPs due to MPs (Teuten et al., 2009), the physical and immunological perturbation that results from MP consumption (Rochman et al., 2013a), and the decrease in detoxification ability as a result of MP consumption (Paul-Pont et al., 2016), the secondary consumption of MPs from seafood is cause for concern. There is no contesting the ocean ecosystem damages that plastic pollution causes. However, connecting such an environmental disaster to human health risk is an important precursor to effective mitigation strategies.

Reducing Sources and Monitoring Sinks

Microplastics are now found in waters and on beaches worldwide. In 2011 Browne and colleagues sampled 18 sites on 6 continents, including the poles. Concentrations of MP were directly correlated to size of human population close to the test site. However, MPs were found at every site, no matter how remote (Browne et al., 2011). Sources of secondary MP were examined using FT-IR spectroscopy, and sewage from laundering polyester-fiber clothing was found to be a major source of MPs, with a single garment potentially creating over 1900 micro fibers per wash (Browne et al.,

2011). New and additional filtration systems for wastewater treatment plants may aid in the reduction of primary and secondary MP loading into aquatic ecosystems, reducing or eliminating fibers from clothes washing and pellets from facial scrubs (though distribution, size, and FT-IR analysis found that facial scrubs are a less important source of MPs than clothing).

Aquatic waste water treatment disposal sites—even ones which had been out of commission for decades—had significantly greater concentrations of MPs than did reference sites, with over 250% more MPs sampled (Browne et al, 2011). MP loading of this kind could also be reduced by regulating the use of MPs in beauty products, where they could be easily replaced by natural abrasives such as sugar, salt, and apricot seed. Clothing materials may be more difficult to replace and phase out, and the costs and benefits of doing so should be evaluated.

Song et al. (2014) found an abundance of alkyd polymer synthetic ship paint particles, in addition to regularly evaluated MPs, suggesting that reclassifying and regulating boat paints may be beneficial to the ocean environment. The same group found an abundance of MPs in the sea surface microlayer, when compared to sampling done at different depths of the water column (Song et al., 2014). Song et al. also found that as particle size decreased, the abundance of MPs increased, suggesting even higher concentrations of nanoplastics. It has also been suggested that the benthos acts as a final sink for MPs, particularly after biofouling of MPs (Wright et al., 2013 and others).

Better Materials

Currently, over 50% of all plastic produced is single-use disposable, while recovery rates in Europe (some of the best in the world) remain at 39% or less (Hopewell et al., 2009 and Gouin et al., 2011). Plastic Europe (2012) estimates that 10% of global plastic produced ends its life in the ocean. According to the NGO 5 Gyres, over a third of plastic debris collected in the oceans is disposable packaging. Microplastics represent more than 92% of plastic debris at sea (Andrady et al., 2011). Eliminating unnecessary uses of this persistent and harmful material is absolutely necessary. A number of safer materials have been proposed for single-use purposes, including within the food industry. For example, Engler (2012) proposes the following replacements in **Table 4**, with the reminder that “not all bioplastics are biodegradable, and not all biodegradable plastics are

bio-based.” For example, PLA, though technically not biodegradable, does degrade faster than PE and other non-bio based plastics in the marine environment, though as Rochman and colleagues point out, PLA still degrades only slightly in a single year in the ocean (Engler et al., 2012, Rochman et al., 2015). It may be possible to find additives that increase biodegradability of plastics--for example, one that enables microbial breakdown of PE (Engler, 2012).

polymer	may replace	potential uses
starch blends/modified starch	PET, PE, PP, PS	packaging, medical products, food service items
PHAs, including polyhydroxybutyrate and polyhydroxyvalerate	PP, PS, PVC, ABS, PC	food service items, durable goods, packing, coastal restoration
PLA	PET, PE, PP, PS	bottles, paper coating, produce containers, apparel

Table 4: Suggestions for improvement of plastic materials with respect to marine environmental health. Source: Engler et al., 2012.

Taking an approach such as the one discussed in McDonough and Braungart’s book Cradle to Cradle, single-use plastics can be replaced with high-quality 100% recyclable (not downcyclable) plastics. Programs could be implemented to financially incentivize recycling of these products. Low-residual value plastic is more likely to leak, presumably due to lack of incentive for retention. In the five countries with greatest ocean plastic pollution, no formal recycling exists, but waste-picking is prevalent. In addition to raising value, the more virgin and quality the plastic, the fewer toxic additives are necessary. At the point of degradation where addition of harmful additives would be necessary, a new use for the material could be found.

For many purposes, plastics may not be the best material. Rather, for foods with a short shelf life, a 100% biodegradable packaging option is feasible, so long as the packaging outlives the shelf life of the food by a small margin.

Products should be re-engineered with end of life in mind. That is, at the end of their lives, products should either enter the biological system or the technological system as fodder (McDonough & Braungart, 2002). Extending producer responsibility in every industry by holding them responsible for packaging would further encourage innovative solutions in materials science.

Reclassification of Microplastics as Harmful Pollutants: A Triage Strategy

In a 2013 Comment paper in the journal *Nature*, Rochman, Browne and others called for a reclassification of plastic waste as hazardous due to human health risk and ecosystem risk. According to this piece, the UN's Globally Harmonized System of Classification and Labeling of Chemicals includes the additives and monomers of over 50% of plastics in the 'hazardous' category. Rochman, Browne et al. found that 78% of the EPA's priority pollutants and 61% of the EU's priority pollutants for elimination are associated with microplastic debris, and cites studies in humans and mussels on MP translocation from ingestion and inhalation as further proof of health hazard. Citing the Montreal Protocol's success in virtually eliminating the use of CFCs, and the Stockholm Convention (2004) banning many POPs, these key researchers in the field of microplastic effects argue that the simple act of international reclassification can decimate groups of pollutants. Additionally, Rochman and colleagues issued a call for clearer and stricter definitions of 'plastic' and 'biodegradability' for the sake of labelling and global materials control (Rochman et al., 2015). By taking clear evidence of ecosystem destruction and human health impact associated with MPs into account, the business-as-usual projection of 33 billion tonnes of plastic produced by 2050 –up from 2012's 0.28 billion–can be curbed (Rochman, Browne et al., 2013b). Ultimately, Rochman, Browne, et al. call for a closed loop system of plastic reuse and recycling. However, these are far from the only options for materials reduction.

Cole et al. (2013), and Avio et al. (2014) have suggested ranking MPs and POPs by hazard rating to target systematic elimination (see **Table 5**). In this way, the danger of plastic type can be ranked and triaged for elimination or reduction. For example, Polyethylene (PE) is currently the most abundant plastic in the oceans worldwide (Gouin et al., 2011). PE also has a greater affinity for persistent organic pollutants (POPs) than does polystyrene (PS), another common microplastic (Koelmans et al., 2013). Weight of Evidence (WOE) evaluations in one experiment pairing different POPs with various MP types showed PE-Pyrene and PS-Pyrene ingestion to be major and severe on the hazard scale, while ingestion of just PE or PS was scaled at slight to moderate (Avio et al., 2014). This assessment included both bioaccumulation and the biomarkers tested (of which there were many).

	PP	PE	PET	PS	PVC	PU
Global production volume (tonnes/year) ^[1]	45	39 (LD) 32 (HD)	33	13 >4*	37	9
Sorption and desorption capacity for POPs** ^{[2] [3]}	Inter-mediate	High	Yes	Yes	Yes	Yes
Hazard rank for polymer 1=most hazardous 37=least hazardous ^[3]	37	36	36	35 34*	5-11	1-7

[1] Lithner 2011 [2] Rochman 2013b [3] Schönlau 2016 *Expandable PS ** PCBs, DDE, PAHs, and Ah-receptor response. PS, polystyrene. PVC, polyvinylchloride. PE, polyethylene. PP, polypropylene. HD, high density. LD, low density. PET, polyethylene terephthalate. PU, polyurethane.

Table 5: Relative abundance (Global Production Volume) of various MPs and their affinity for various POPs, along with a hazard ranking. Source: Cole et al., 2013 and sources cited therein.

More recently, a 2015 *Nature* editorial piece has called for a ban of MPs in the beauty industry. It points out that California is one of the first states to have enacted this ban recently, and that CA’s slow phase-out efforts are insufficient (*Nature*, 2015). Rochman et al also called for a microbead ban, citing single-use design and sheer volume of waste. Eight trillion microbeads per day are being released into aquatic habitats in the US alone from wastewater treatment plants (Rochman et al., 2015). Gouin et al. (2011) estimated the per capita usage of microbeads in the US at 2.4 mg/person/day. According to Gouin et al., 50% of primary microplastics produced in the US reach marine environments. Throughout the scientific community there has been a call for more apt materials applications, with over 70 NGOs in 30 countries as well as major brands like Target Corp and Johnson & Johnson calling for and pledging a plastic microbead ban (Rochman et al., 2015).

Ocean Plastic Mitigation Strategies

The plastics industry and demand for plastic consumer goods is expected to increase over the next 10 years, and by 2025 The Ocean Conservancy projects that the ocean could contain “one ton of plastic for every three tons of finfish,” as plastic entering the ocean may double from 2015 to 2025 (Ocean Conservancy, 2015). The ICIS Supply

and Demand Database projects that global demand for plastics will increase from 250 million metric tons in 2015 to 380 million metric tons in 2025 (ICIS, Ocean Conservancy, 2015). Plastic is extremely persistent in the marine environment—some plastic materials are recognizable in their original form 400 years after being discarded there (Ocean Conservancy, 2015). Due to the worldwide problem of marine plastic pollution and the difficulty of filtering MPs from plankton (Avio et al., 2015), the removal of plastics from the ocean is a problem of colossal scale. However, a few strategies have been suggested, including collecting ocean plastics with a boom using the natural circulation of the ocean gyre (The Ocean Cleanup, 2016). The Ocean Conservancy, however, has recommended that due to the enormity of the problem and its many logistical issues, the majority of ocean plastic effort should take place at its major source—the land (Ocean Conservancy, 2015).

The 2015 Ocean Conservancy report on ocean plastics points out that most plastics come from a small geographic area consisting of developing nations. More than half of the plastics sampled in this study came from China, Indonesia, the Philippines, Thailand, and Vietnam (Ocean Conservancy, 2015). These same countries also have the greatest projected growth rates for plastic waste production worldwide (Ocean Conservancy, 2015). The use of plastics in these countries has been correlated with GDP increase, greater global economic importance, and better quality of life, but waste-management has not kept up with the growth of plastic use, either in terms of scale of collection or waste retention. The authors of this report suggest using global funds to target this area of great importance (the five countries above) with respect to ocean plastics, and implementing a global ocean plastics reduction effort. Global cooperation and international supply chain management is necessary to achieve a worldwide ocean plastics and ocean pollutant reduction goal (The Ocean Conservancy, 2015).

The total cost of the mitigation strategies proposed by the Ocean Conservancy is estimated at \$5 billion USD/year. The organization claims high returns on investment, and proposes both public and private funding, though realistically private industry has the greatest ability to catalyze this effort due to the necessity of quick and immediate action. Of course, international political buy-in is essential.

Ocean cleanup solutions were not the focus of The Ocean Conservancy's 2015 call for action. As a result of a cost-benefit analysis, only preventative solutions were discussed in depth. However, I suggest that the following remediation approaches may prove cost-effective and should be seriously considered.

Wildlife-safe marine plastic collection systems could be built into offshore wind and oil rigs, or incorporated into routine coast guard and naval surveillance. A bounty on marine plastics would incentivize fishermen and other ocean-based businesses to collect ocean trash. There have been a number of businesses built solely upon ocean plastic removal and re-purposing, including NetPositiva skateboard company and others discussed below.

As suggested above, point-source solutions implemented at wastewater treatment plants could reduce marine input of MPs dramatically if incorporated at a large scale (Browne et al., 2011). Certainly, improvements in waste disposal and recycling, and municipal composting infrastructure on land, as well as incentives for recycling and use of compostable materials, may be extremely helpful in terms of ocean impacts. Less than 20% of plastic leakage comes from ocean-based sources (fisheries, etc), while over 80% is from land-based sources (Andrady et al., 2011). This means that plastics are not well managed after being discarded. Dumping waste from waste-transportation systems, and uncontained waste dumps should be penalized (Ocean Conservancy, 2015). More effective measures by which to transport plastic materials by ship should be implemented, and enforcement strategies for offshore dumping should be internationally discussed.

Improved waste to energy technology (gasification, pyrolysis, incineration with energy recovery) may aid in plastics reduction only if the amount of airborne dioxins and other carcinogenic chemicals emitted by these technologies can be reduced to safer levels.

Government organizations such as the European Marine Strategy Framework Directive, US EPA, NOAA, the UN Environment Programme, and the Intergovernmental Oceanographic Commission should be encouraged to take action in matters of marine plastic pollution collaboratively. In addition to government organizations, NGOs such as the Trash-Free Seas Alliance, Surfrider Foundation, and 5 Gyres may be helpful allies in

this effort. If, by international agreement, more PBTs can be phased out and ocean plastic pollution prevention programs adopted, then the removal of marine plastics may result in a net removal of ocean toxins associated with those plastics.

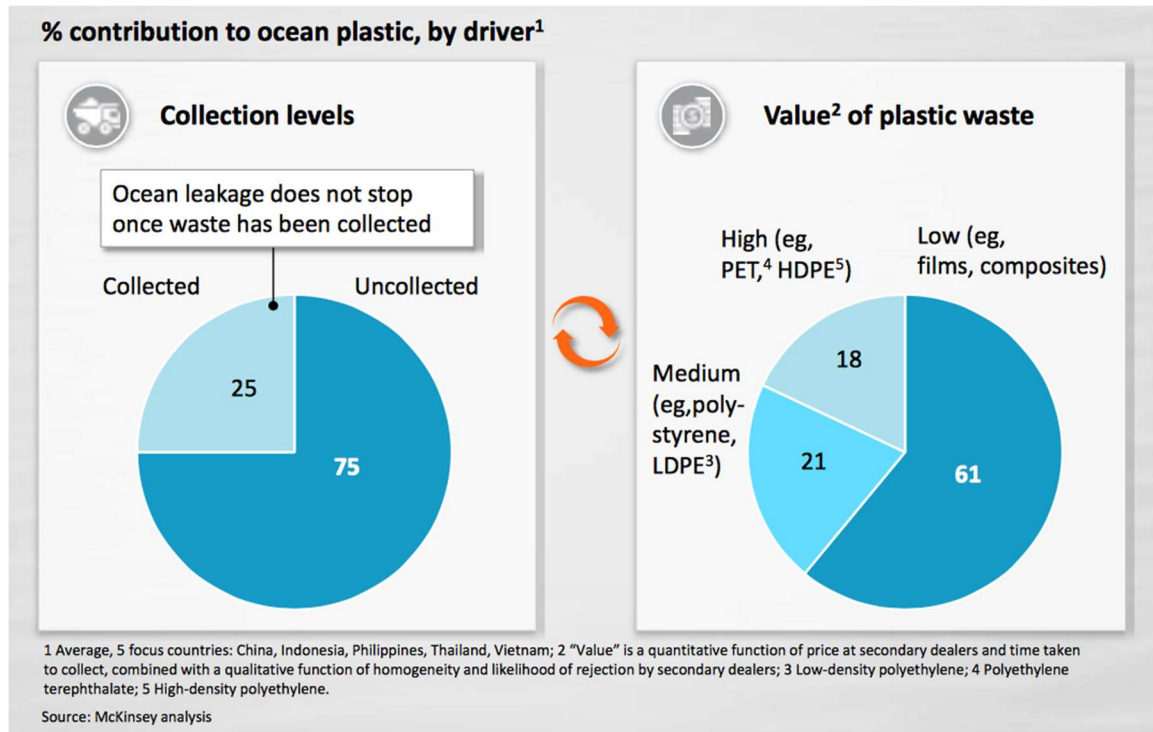


Figure 11: Prevalence of ocean plastics and collection levels in 5 focus countries China, Thailand, Vietnam, Indonesia, and the Philippines. Source: The Ocean Conservancy, 2015.

Ocean Plastic Cleanup as a Business Opportunity

The Ocean Cleanup is a relatively new company begun by Boyan Slat when he was only sixteen years old (Theoceancleanup.com). The company has developed a prototype boom for ocean plastics collection. A year-long prototype test of this product began in June of 2016, and will examine, among other things, the ability of the boom to weather rough seas and the potential for the boom to collect bycatch (although at only four feet of depth, the company does not expect bycatch to be an issue). The boom's materials are designed to be serviceable and it is fatigue tested to 100 years of service to prepare it for Pacific storms. Using technology from floating oil platforms, it is designed not to float away and become debris itself. The Ocean Cleanup expects full deployment of their product in the Great Pacific Gyre in 2020, and hopes to replace boat-and-net

models of plastic collection with networks of floating barriers (theoceancleanup.com). The company claims that this system could remove half the plastic in the Pacific Gyre in 10 years, though this claim is under scrutiny by other sources. The boom could also be deployed at river mouths to prevent plastic pollution from freshwater sources (theoceancleanup.com). Its efficacy at MP removal from the surface layer is not immediately apparent in the website, but the product seems to target macroplastic removal.

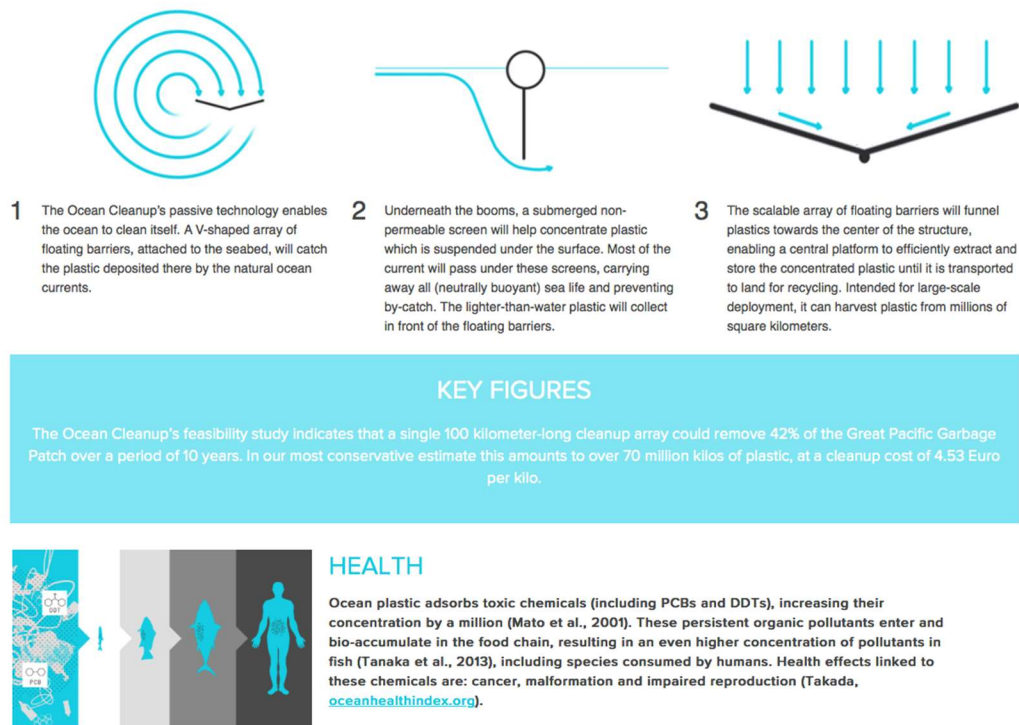


Figure 12: Infographic describing the boom technology created by The Ocean Cleanup company. Source: theoceancleanup.org

Regardless of efficacy of the boom technology proposed, The Ocean Cleanup company has successfully funded a project which makes a business opportunity out of a global environmental crisis. More efforts to this end would be most helpful to the microplastics problem, as cleanup of larger plastic debris would reduce production of secondary MPs. Ultimately, the ocean plastics problem will only worsen without solutions at the source of plastic pollution. There have been a number of land-based companies who have attempted to tackle the ocean plastics issue from its land-based

source. Terracycle is a market-based, independent company that turns waste plastic to profit by paying for empty branded packaging to use as inputs for its products. Covanta Energy uses derelict fishing gear as fodder for incineration and energy production. Finally, QinetiQ is a company that uses pyrolysis to turn plastic into fuel (Engler et al., 2012). Further innovation and green entrepreneurship is needed in the arena of marine plastic pollution prevention and mitigation.

Unfortunately for the green entrepreneur, it is extremely difficult to separate microplastics from benthic debris or particles in the water column based on charge or other chemical affinity, particularly in seawater, which is charged itself. Previous technology for microplastic removal in sand operates via electrostatic attraction (Ward, n.d.). An additional issue is posed by the fact that netting which catches microplastics also catches plankton, and separating biological material from plastics without loss of plankton would be challenging (Avio et al., 2015 and others). Successful filtration of nanoplastics from the ocean would be even less likely. The diffusivity of microplastics (only a few kg per square km in the Pacific Gyre) would be a hindrance to filtration-based removal (Engler, 2012). While further experimentation and innovation may prove helpful, the most cost-effective strategy for ocean pollutant reduction is likely source-focused rather than cleanup-focused (Ocean Conservancy, 2015).

LCA Strategies

Finally, the ocean health impacts of plastics should be incorporated into lifecycle assessment databases such as EcoInvent, particularly in terms of biodiversity and human health impacts. The number of threatened species found stranded and dead, for whom the cause of death is determined to be plastics has been evaluated in a number of reports compiled by The Ocean Conservancy (Ocean Conservancy, 2015 and Allsopp et al., 2006). (Survivorship can also be measured in terms of reduction in fecundity or reproduction. However, there are many confounding factors to survivorship.) From these numbers, an estimate can be made as to the number of threatened or endangered animals at risk of death per pound of plastic produced. Type of plastic may even be delineated. A biodiversity impact per pound of plastic produced should be introduced to account for the environmental cost of this material over biodegradable materials, and encourage changes in single-use packaging. Microplastics have been shown to have a direct impact upon

endangered species. For example, foraging habitat of filter-feeding fin whales overlap areas of greatest plastic density (Fozzi et al., 2016). Any human health impacts confirmed as a result of MP consumption can also be added to LCA databases.

Monetized Health Risk & Benefit of MP reduction in Aquatic Ecosystems

The scientific and food regulatory communities would benefit from a cost-benefit analysis of marine plastic cleanup. Such a study would help inform the cost of global seafood safety.

In the EU, mollusks are regulated solely on the basis of microbial properties—that is, microbes and microbe-associated toxins (Binelli & Provini, 2003). It may be useful to regulate bivalves also by POP concentration and to quantify MPs within a representative sample of a given shipment. Vandermeersh and colleagues have called for new standards for seafood contamination which include MP content (Vandermeersh et al., 2015). To rid bivalves of MP contaminants, a required depuration period (placement in clean water for a few days) may be partially effective (Santana et al., 2016).

In New England, for example, the stomach contents of ~60% of fish species sampled contained polystyrene. Health impacts to the fish and subsequently to the human consumer are not well understood. Similar findings were confirmed in mollusks.

Santana et al. (2016) found microplastics in 75% of mussels sampled from the populated Santos Bay, Brazil, with each sampling site containing contaminated mussels. This species, *Perna perna*, are a key foodsource in the area. Cultured mussels also use marine water, and are not exempt from marine pollution issues. Van Cauwenberge et al. (2014) found that mussels sampled had an average plastic load of 0.36 ± 0.07 particles per gram tissue (wet weight), while oysters sampled had on average 0.47 ± 0.16 particles per gram tissue (ww). Average human intake will vary by location and individual, but as mentioned earlier, Van Cauwenberghe et al. estimate that the annual dietary exposure of a European to MPs, via shellfish alone, is around 11,000 MPs/year. However, it is widely claimed that estimating human health risks of MP consumption is not yet possible. The study proposed above, along with a cost-benefit analysis of the assurance of food safety, would allow for better understanding on this matter.

Once human health impacts from MP consumption do become noticeable and are studied, human health impacts can be monetized using the following metrics:

- Earnings lost, days at work lost, & cost of care.
- Pain and suffering as a multiple of tangible metrics (money earned over a period of time, costs of health care, rehab, etc.,)
- Use of actuarial tables for the effects of immunomodulators on health(if this is the primary overall effect of MP consumption)

Shareholders

Engler et al. demonstrated that PBTs can be transported globally on MPs, and argued for international regulation (Engler et al., 2012). In fact, US and Japan have the highest concentrations of MP-associated PCBs on shoreline debris (Engler et al., 2012 and sources therein). Other researchers have speculated that certain pathogens can also be transported long distances via fouled MPs.

Issues of ocean plastic pollution pertain to human health and effect the food industry, policymakers, public health officials, citizens of nearshore areas, consumers, and companies concerned with seafood products. Action can be taken by anyone in these groups to facilitate food safety in relation to microplastics, and in turn implement actionable steps to reduce ocean plastic pollution. Increasing awareness of plastic pollution effects on human immune response, physiology, and toxin loading as a result of microplastic consumption may also draw attention to the overall effects of MPs on the food web. Microplastics and nanoplastics present a sufficiently concerning human health risk atop known ocean health risk, and warrant the attention of policymakers and manufacturers to mitigate global impacts.

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Appendix I: Bioaccumulation factors compiled by Koelmans et al. (2013)

Table S5: Summary of HOC desorption rate constant k_{IG} (d^{-1}) from plastic in the gut of marine biota, obtained from (a) bioaccumulation studies, (b) desorption studies, (c) first principles^a.

Value or range	Chemical/Plastic	Organism or condition	Plastic	Comment	Source
Laboratory bioaccumulation studies (modeled with SI Eqs. 1 and 2)					
4.1 – 9.8	Nonylphenol	Lugworm <i>A. marina</i>	PVC 230 μ m	Body wall - gut	This study, based on Browne et al 2013
0.76 – 1.6	Phenanthrene	Lugworm <i>A. marina</i>	PVC 230 μ m	Body wall - gut	This study, based on Browne et al 2013
2.5 → '> 50'	PBDE-47/PVC	Lugworm <i>A. marina</i>	PVC 230 μ m	Body wall - gut	This study, based on Browne et al 2013
0.073	PBDE-28, 47, 100, 99	Amphipod, <i>Allorchestes compressa</i>	PE 11-700 μ m	Chemicals at equilibrium only	This study, based on Chua et al, 2014
1.89	PCBs	Japanese Medaka. <i>Oryzias latipes</i>	PE < 500 μ m		This study, based on Rochman et al, 2013
1.16	PAH	Japanese Medaka. <i>Oryzias latipes</i>	PE < 500 μ m		This study, based on Rochman et al, 2013

<i>Desorption studies using artificial gut fluids</i>					
4.09	Phenanthrene	15.5 mM STC ^b 18°C	PE 200- 250 µm		Teuten et al, 2007
9.7	Phenanthrene	15.5 mM STC ^b 18°C	PP 200- 250 µm		Teuten et al, 2007
2.29	Phenanthrene	15.5 mM STC ^b 18°C	PVC 200- 250 µm		Teuten et al, 2007
2.3	Phenanthrene	15.5 mM STC ^b 18°C	PVC 130 µm		Teuten et al, 2007
3	Phenanthrene	15 mM STC ^b 18°C, pH=7.5- 8.4	PE		Bakir et al, 2014
1.68	DDT	15 mM STC ^b 18°C, pH=7.5- 8.4	PE		Bakir et al, 2014
0.27	DEHP	15 mM STC ^b 18°C, pH=7.5- 8.4	PE		Bakir et al, 2014
12.10	Phenanthrene	15 mM STC ^b 38°C, pH=4	PE		Bakir et al, 2014
7.2	DDT	15 mM STC ^b 38°C, pH=4	PE		Bakir et al, 2014
3.89	DEHP	15 mM STC ^b 38°C, pH=4	PE		Bakir et al, 2014

1.67	Phenanthrene	15 mM STC ^b 18°C, pH=7.5-8.4	PVC		Bakir et al, 2014
0.31	DDT	15 mM STC ^b 18°C, pH=7.5-8.4	PVC		Bakir et al, 2014
1.37	DEHP	15 mM STC ^b 18°C, pH=7.5-8.4	PVC		Bakir et al, 2014
4.8	Phenanthrene	15 mM STC ^a 38°C, pH=4	PVC		Bakir et al, 2014
0.54	DDT	15 mM STC ^b 38°C, pH=4	PVC		Bakir et al, 2014
4.86	DEHP	15 mM STC ^b 38°C, pH=4	PVC		Bakir et al, 2014
0.81	BDE209	Fish oil, 38°C	HDPE	See Table SI-4, calculated with SI Eqs. 3 to 9.	<i>This study</i>
0.085	BDE209	Stomach oil, 38°C	HDPE		
<i>Calculated from first principles^c</i>					
0.1	PCB	Polymer diffusion	1.3 mm PS	Besseling et al, 2013	Koelmans et al, 2013
2	PCB	Polymer diffusion	0.4 mm PS	Besseling et al, 2013	Koelmans et al, 2013
>100	PCB	Polymer diffusion	0.1 – 1 µm PS	Besseling et al, 2013	Koelmans et al, 2013
0.9 - 9.8	PCB (28, 52, 101, 153, 180)	Polymer diffusion	PE 0.5 mm	As used by Rochman 2013 (< 0.5 mm)	<i>This study</i>
1.67 - 53	PAH (BAA, CHRYS, PHEN, ANT, FLU)	Polymer diffusion	PE 0.5 mm	As used by Rochman 2013 (< 0.5 mm)	<i>This study</i>

^a The 10-90% inter quantile range $PR_{10-90\%}$ for all data is $0.3 - 9.8 \text{ d}^{-1}$ with a median of 2.1 d^{-1} . For the separate categories of studies, $PR_{10-90\%}$ overlaps with ranges of $0.6 - 17.8$; $0.3 - 8.0$; and $0.7 - 27.8$ for modeling studies, desorption studies and first principle calculations, respectively. The ranges are not affected by the uncertainty in the outlying values (indicated with '>').

^b Sodium taurocholate

^c First order rate constants based on $k_{1G} \approx 23D/r^2$ with D is the polymer diffusion coefficient and r is the (average) radius of the MP particle.

Koelmans, A.A., E. Besseling, A. Wegner, E.M. Fockema. (2013). Plastic as a carrier of POPs to aquatic organisms. A model analysis. *Environ. Sci. Technol.* 47. 7812–7820.

Appendix II: Summary of Results of Selected Relevant Experiments

Rochman et al., 2013a:

- Significant difference in PBDE totals after 2 months for fish fed a mixture of marine-equilibrated plastics and regular feed. Experiment takes into account actual marine concentrations of POP on MPs, some baseline contamination of fish food, and longer term exposure.
- “Polyethylene ingestion is a vector for the bioaccumulation of PBTs in fish, and that toxicity resulting from plastic ingestion is a consequence of both the sorbed contaminants and plastic material” (Rochman et al., 2013a).
- LDPE exposed to the marine environment had 129 ng/g PAHs, 17 ng/g PCB, and 1.9 ng/g PBDEs (Rochman et al., 2013). PAH and PCB concentrations were an order of magnitude higher than the amount found in a normal marine diet.
- PBTs sorbed to the plastic from seawater transferred from plastic to *medaka* (fish) upon ingestion, suggesting MPs may be a relevant pathway of exposure to these chemicals despite global contamination (Rochman et al., 2013a).

Avio et al., 2013

- MPs are definitively shown to transfer PAHs to blue mussel tissue and increase bioavailability there.
- There are toxicological effects of virgin PS and PE (mostly measured at cellular level)
- Pyrene was used because it is one of the more common PAHs found on plastic debris.
- Unlike some experiments that dose higher than environmental levels of contaminant, this experiment maintained environmentally sampled levels of pyrene. It used farm-raised, sea-bred *Mytilus spp.*
- <100µm microplastics were used, thus smaller MP size could have contributed to the bioaccumulation observed.
- Pollutant concentration was 200-260 ng/g of pyrene.
- 7 days’ exposure saw significant increase in pyrene in the gills, and even greater accumulation in the digestive glands. The glands had concentrations much greater

than the MPs, with tissues up to 470 ng/g, thirteen times greater bioaccumulation than the control. Digestive gland concentrations were up to three times greater than the concentration on the MPs.

- This experiment showed elevated desorption rates of POPs from MPs and an increase in bioconcentration processes within gut tissue.
- This suggests that there is more than just chemical gradient to consider in a risk assessment of tissue POP bioaccumulation.
- Greatest adsorption efficiency for pyrene to PE/PS was at moderate pollutant levels (5µg/L), with adsorption not differing much for each plastic in the case of pyrene.
- 5µg /L pollutant concentration is consistent with POP levels measured on the CA coast.
- Salinity of the water has relatively little effect on pollutant adsorption compared to chemical concentration, plastic density and residence time. Therefore, estuarine plastic can also pose an issue for marine biota.

Paul-Pont et al., 2016

- Fraction of fluoroanthene transferred from algae to microbeads of PS confirms a higher partition coefficient of micro-PS than other partition fractions measured (Log $K_p=6.6$)
- This didn't lead to significant change in FLU bioaccumulation in mussels. Micro-PS had a minor role in transferring FLU to tissues compared to other pathways (water, food).
- After depuration, MP exposed mussels still had more FLU in their tissue.
- Their detoxification mechanisms were down-regulated (presumably due to down-regulation of P-glycoprotein). Impaired filtration activity or presence of MPs still in gut could be other reasons for impaired detoxification.
- Micro-PS dosing alone led to higher hemocyte mortality, and changed cellular oxidative balance, increasing ROS production in hemocytes.
- This was a short-term study (14 Days)—longer term studies may show different bioaccumulation effects.

Teuten et al., 2009

- Teuten et al. performed experiments on shearwater chicks, feeding one group a single dose of 1g MP pellets with 15ng associated PCBs. One gram of MPs coincides with about a cubic centimeter of microplastics, a large daily dose. Chick body burden increased by 25-100% as a result of this single dose (containing about 100 ng PCBs total), even though both control birds and test birds were fed PCB burdened fish as well (0.298 to 0.706 ng/g w.w. fish). There was a statistically significant difference in MP-specific lower chlorinated congeners of PCBs, which would otherwise be degraded in the food chain if not bound to MPs.

Teuten et al., 2007

- 13% decrease in body burden of phenanthrene from lugworm upon addition of *clean* PE to sediment (Teuten et al., 2007).

Brouwmeester et al., 2014

- (Big-picture thought experiment) A 300g average portion of mussels contains 300 plastic particles (about 1.5 µg of plastic) (Van Cauwenberghe et al., 2014).
- Per capita yearly consumption of mollusks excluding cephalopods was measured at 2.4 kg/capita/year by the FAO in 2012. By this study, dietary exposure is found to be about 300 MPs per capita per year (about 0.8 MPs per day), three orders of magnitude lower than Van Cauwenberge's study for the same European population.
- Mussels and beer were similar in MP amounts: 0.3-2 per gram in mussels, 0.01-0.1 per gram in beer (Van Cauweberge & Jenssen, 2014), (Liebezeit et al., 2014), but fish and shellfish remain a likely important source of MP intake (Brouwmeester et al., 2014).
- "European Food Safety Authority estimated average exposure to non-dioxin like PCBs at 0.3-1.8µg/day for a 70 kg person" (Brouwmeester et al., 2015)
- Dietary intake of BPA for adolescents can be up to 1.449 µg/kg /day according to the EFSA. At this rate a 70kg adolescent would consume 27 µg per day, and the contribution of plastics from an estimated average mussel portion would contribute a negligible 0.25% of ADI (Brouwmeester et al., 2015).

- “Even if we assume that the PCBs are completely released from the plastics in the mussels, their consumption would change PCB exposure by less than 0.0001%” (Brouwmeester et al., 2015)
- “At time of human consumption, *M. edulis* contains on average 0.36 +/- 0.07 particles/gram wet weight” (Van Cauwenberghe et al., 2014). *C. gigas* had a plastic load of 0.47 +/- 0.16 particles/gram w.w. (Van Cauwenberghe et al., 2014).
- Average size of *M. edulis* grown for commercial purposes collected by Vandermeersch et al. (2015a) in France were 3.5 +/- 1.2 g each.
- Commercial mussels analyzed by Mathalon & Hill (2014) were found to have between 0.00 and 0.68 MPs/g w.w.

Appendix III: A Note on the inclusion of Plastic Additives

Additives

Plasticizers (and other additives) diffuse from the plastic surface. As the surface is depleted of additive, a chemical gradient is created which pulls additive from inside the plastic to the surface, where it can then leach out (Engler, 2012). This process is much slower for additives which are chemically bound to the plastic matrix. Many additives break down faster than POPs (except PBDE, which is a POP, and can be found either within the MP or sorbed to it). Approximately 4% of the weight of plastics is additives (NP, OP, BPA, and PBDE) (Brouwmeester et al., 2015), so there is plenty of chemical leaching potential from MPs which have a large surface area to volume ratio.

Phthalates (the most common plastic additive) are not persistent (they degrade), but leach steadily. They include DEHP, which is high MW, hydrophobic, and resists migration, as well as DMP, which has a low MW, easily migrates from resin, and is hydrophilic (Engler, 2012). Nonylphenol (NP) persists for months, does not biomagnify, but is toxic to mammals as an endocrine disruptor (Engler, 2012). Biomagnification of phenols is unlikely as they have a hydrophilic group and metabolize relatively easily (Teuten et al., 2009). NP concentrations on plastic are mostly due to the additive and not from sorbed chemicals in the environment, and therefore MPs may be a significant source of NP to aquatic species (Engler, 2012). The concentration of concern for aquatic species

is 0.7 ppb (0.7 µg/L) in water. Because of its high concentration in plastic and the fact that it does not biomagnify, the presence of NP in aquatic species may be assumed to be mostly due to plastic (Engler, 2012).

BPA has a moderate biodegradability, and degrades less in seawater. It does bioaccumulate in fish. The primary route of human exposure is through canned foods or fish ingestion, depending upon rates of ingestion. Human BPA limits are yet to be determined (Engler, 2012).

Phenols, NP, and BPA all have low concentration potentials. They do not concentrate in fish unless there is plastic in the fish diet (Engler, 2012). They thus serve not only as good proxies for plastic ingestion, but also may be of greater overall importance to toxin load in consumers due to lack of other sources for these chemicals.

For purposes of this experiment, additives are pooled with overall human toxin load. Even though some are not classified as POPs and can be metabolized more readily, almost all have endocrine effects or carcinogenic effects on humans, and overall toxin burden is thus pertinent to a discussion on control of plastic pollution. However, once more is known about a specific POP or additive, it may be more helpful to model it separately.

Appendix IV: Equations from *Vensim* model

"grams MP ingested"

Units: grams

"[additives] within MP"

Units: ng/g

Varied

"[POPs] sorbed to MP"

Units: ng/g

Varied

k1, k2, k3, and k4 sorption constants are dimensionless, percentages of total toxin load. They can be positive or negative.

"[POPs] desorbed into Mussel"= (k1 sorption constant*" [POPs] sorbed to MP"*grams MP ingested)+(k2 sorption constant*" [additives] within MP"*grams MP ingested)
Units: ng

"[POPs] remaining on MP"= "[POPs] sorbed to MP"-(k1 sorption constant*" [POPs] sorbed to MP")
Units: ng

"[additives] remaining within MP"= "[additives] within MP"-(k2 sorption constant*" [additives] within MP")
Units: ng

"[OrganicPollutants] bioavailable to Human"= "[POPs] desorbed into Mussel"+(k3 sorption constant*" [POPs] remaining on MP")+(k4 sorption constant*" [additives] remaining within MP")
Units: ng

Appendix V: Suggested Experimentation on Mammals

Closing a Knowledge Gap: An Experiment to Understand Effects of Microplastic consumption in Mammals

Modeling has suggested that MPs may not pose a large threat in terms of direct toxin loading in humans under current conditions, although some *in vivo* experimentation has suggested otherwise. Physiological and immunological effects on mammals still warrant further investigation.

I propose an experiment to directly measure the effects of secondary (or primary) MP consumption on mammals. This study would utilize a number of groups of piglets, fed a diet of clean bivalves with no POP contamination (control), POP-polluted bivalves (treatment 1), MP-only polluted bivalves (treatment 3) and POP-MP polluted bivalves

(treatment 4). Further possible treatments are described below. Pollutant levels could be varied but at least one set should be environmentally relevant. Pigs should also be fed a mixture of other foods as needed to maintain their health. Some treatment groups could be ethically sacrificed within 6 months to measure immediate effects of secondary MP consumption, while a second group set should be allowed to mature to measure long-term effects of MP consumption. Pigs were chosen for this experiment due to their physiological similarity to humans.

Protocol choice for such an experiment will be difficult due to lack of standardization in this new and growing field (Vandermeersch et al., 2015). In general, information on the effects of microplastics is reported in such a variety of different ways that assumptions have to be made by later researchers for their own experiments, leading to potential bias (Vandermeersch et al., 2015). Any new experimentation must take care to report results in as accessible a way as possible, communicating with others in the field.

As pointed out by Remy et al. (2015), it is important to characterize the particles removed from or visualized within an organism, lest a particle like cellulose (found within their test subjects) be mistaken for microplastic. To do so, they used Raman spectroscopy to identify fibers within one set of test subjects (small benthic crustaceans). Fibers were also identifiable via scanning electron microscopy (Remy et al., 2015).

Avio et al. (2015) found that frequently-used acid digestion techniques for MP enumeration within organisms actually destroy part of the MPs within the animal. They therefore tested and vetted a new protocol for extraction of MPs, which combines two former protocols. Each sample is added to a NaCl hypersaline solution, decanted, then twice filtered. The sample is then added to 15% hydrogen peroxide to dissolve flesh partially, and dried overnight at 50 deg. C. All solutions are pre-filtered and beakers cleaned with deionized (DI) water. The entire procedure is performed under a hood while wearing pure cotton lab coats to prevent contamination. FT-IR Raman Spectroscopy is then used to characterize plastic polymers (Avio et al., 2015). This is the method I propose using for MP enumeration in test subjects for this new experiment, as it was confirmed to be the most effective of the 6 protocols tested by Avio et al (2015).

For histological examination, various body parts can be removed from the organism (once slaughtered humanely), and flash-frozen in liquid nitrogen. Ten μm -wide cryostatic sections of these organs would then be stained and observed with polarized light microscopy (Avio et al., 2015).

This experiment should account for toxin loading via other means, and measure toxins in other (non-bivalve) pig feed, water and enclosure (Paul-Pont et al., 2016). Total pig toxin burden should be measured in control and test subjects and normalized. There is much debate in literature over the relative importance of the route of chemical exposure.

This experiment should use a representative mix of plastics and plastic sizes. To create MPs of different sizes for experimental use, pellet beads can simply be ground in a conical burr grinder as per Rochman et al., 2013a.

Such an experiment could test:

- Long-term vs. short-term exposure
- Primary vs. secondary ingestion
- The effect of secondary ingestion of MP-polluted bivalves in environmentally-relevant marine conditions by mammals similar to humans.
- Effect of nanoplastic, tagged with an easy-to-visualize material e.g. gold-PS or luminescent material.
- Virgin vs. contaminated plastics

Test criteria would include:

- Immunological, cytotoxic, and genotoxic markers: granulocyte formation, phagocytosis activity, lysosomal membrane stability, AChE activity, ROS production, DNA strand breaks, and any other mammal-specific markers commonly used.
- Histology of various tissues, including stomach, intestine, liver, and brain.
- Enumeration of plastics in tissue via digestion procedure described above.
- Toxin loading relative to MP toxin content and toxin content of other food and surroundings.

In analyzing the toxicology portion of this experiment, a Weight of Evidence approach should be used as well, as recommended by the European Directives. In this approach, each variable (including immune effects, bioavailability of pollutants, etc.) is given an index of hazard and then integrated in the WOE evaluation (Avio et al., 2014). The Hazard Quotient for Biomarkers formula published in Piva et al., 2011 may be of use.

An epidemiology-type experimental analysis should also be performed for long term nano- and microplastic exposure in pigs. The following describes the basic method for analyzing epidemiology results in paired group testing.

	Sick	Well
Exposed	a	b
Non-Exposed	c	d

Relative risk: $[a/(a+b)]/[c/(c+d)]$

Relative risk=1: no association

Relative risk >1: exposure harmful

Relative risk <1: exposure may be protective

Attributable risk: rate attributable to exposure. Amount of disease that could be eliminated by removing exposure = $(a-c)/[(a+b)-(c+d)]$.

Attributable risk is still not sufficient to prove causation. A cohort study is necessary for this type of analysis. There may be a multifactorial causation, but eliminating MPs (or nanoplastics) from the diet could eliminate attributable risk by removing exposure.

Population attributable risk is risk in the general population that could be eliminated, given by:

Incidence in general population – incidence in exposed population.

Because of its direct physiological relevance to humans, such an experiment may well inform further legislation on the uses of plastics, micro- and nanoplastics, and plastic-associated chemicals. It is necessary to understand not only the lower level trophic effects of microplastic ingestion, but its effects upon humans. Quick action is needed to transition to smarter materials, and limit the use of plastics for single-use purposes.

