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A Bayesian analysis of the factors determining microplastics ingestion in fishes

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ABSTRACT

Microplastic particles (MPs) occur widely in aquatic ecosystems and are ingested by a wide range of organisms. While trophic transfer of MPs is known to occur, researchers do not yet fully understand the fate of MPs in food webs. We explored the factors influencing reported ingestion of MPs in marine and freshwater fishes by conducting a literature review of 123 studies published between January 2011 and June 2020. We used Bayesian generalized linear mixed models to determine whether MP ingestion by fishes varies by Food and Agricultural Organization fishing area, trophic level, body size, taxa, and study methodology. After accounting for methodology, strong regional differences were not present, although ingested MP concentrations were slightly different among some FAO areas. According to the reviewed studies, MP concentrations in fish digestive tracts did not increase with either trophic level or body size, suggesting that biomagnification of MPs did not occur, although larger fish were more likely to contain MPs. Researchers reported higher concentrations of MPs in clupeids compared with other commonly studied taxonomic families, which could be due to their planktivorous feeding strategy. Methodology played an influential role in predicting reported concentrations, highlighting the need to harmonize methods among studies.

1. Introduction

Microplastic particles (MPs) have been found worldwide in the digestive tracts of marine and freshwater animals, although the implications for ecosystem-level processes remain largely unknown (Akdogan and Guven, 2019; Hale et al., 2020). Researchers commonly define MPs as being between 1 and 5000 (and sometimes 1–1000) μm along their longest dimension. MPs are further categorized by source as either primary, meaning that they were manufactured to size for a particular function, or secondary, meaning that they were produced via fragmentation of larger plastic objects, including textiles. MPs can be further divided based on color and shape - generally fragments, foams, films, and lines or fibers (GESAMP, 2019). MP fibers are sometimes also referred to as microfibres, although this term may also include semi-synthetic (*e.g.* viscose) fibers, as well as anthropogenic natural fibers like cotton, wool, and silk (Singh et al., 2020; Barrows et al., 2018). In this study we only consider synthetic microfibres as MPs. MPs are complex contaminants, and can vary greatly depending on their size, shape, polymeric composition (*e.g.* polypropylene, high density polyethylene, low density polyethylene, polyvinyl chloride, polyurethane), chemical additives (*e.g.* flame retardants, plasticizers, UV stabilizers), and contaminants that have been sorbed from the environment, including heavy metals, pesticides, pharmaceuticals, and persistent organic pollutants (Rochman et al., 2019).

Once ingested, microplastics have the potential to cause harm to organisms via several pathways. A recent meta-analysis by Jacob et al. (2020) found that in fish, primary virgin MPs (fresh from the manufacturer) affected 32% of endpoints that had been studied, including significant effects on behavioral, sensory, and neuromuscular functions, metabolism, the alimentary and excretory system, the microbiome, and the immune system. The effects of MPs are highly variable across taxa, although smaller organisms such as larval fish and zooplankton may be more susceptible (Foley et al., 2018). In addition, the presence and extent of the effects of MPs on animals are highly dependent on the

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duration of exposure, concentration, shape, size, and polymer type of the particles, with only around 17% of experimental studies testing environmentally relevant concentrations, and often using particles smaller than those that are accurately reported by current analytical techniques (Bucci et al., 2020).

A key aspect of understanding the potential ecotoxicological effects of MPs is the process by which MPs move through food webs (Khalid et al., 2021). Determining whether a contaminant can bioaccumulate and biomagnify is vital to understanding which organisms are most affected by it and to what extent (Beek et al., 2000). In our paper, we define bioaccumulation as when the intake rate of MPs is greater than the excretion rate, which would lead to a net increase in MPs in the body of an individual through time (Arnot and Gobas, 2006). Biomagnification would occur if sufficient bioaccumulation is occurring, leading to increasing MP concentrations for higher trophic level animals (Katrine et al., 2011). If accumulation and magnification are both occurring, then animals at higher trophic levels will be at the greatest risk of suffering any negative consequence of exposure to MPs. If, however, insufficient accumulation is occurring to cause magnification (trophic dilution) then the organisms most at risk will be those with the highest encounter rates and lowest rates of depuration, for example low trophic level, benthic organisms (Lagesson et al., 2016). Recent work indicates that, in general, biomagnification of MPs does not occur, suggesting that accumulation is therefore not substantial for the size range of MPs that are commonly documented in biota, and that lower trophic level animals are at greatest exposure risk (Walkinshaw et al., 2020; Gouin, 2020). However, a consensus has not yet been reached.

Marine and freshwater fishes occupy a range of trophic levels, making them ideal organisms for studying the trophic dynamics of MPs. There are several direct and indirect ways in which microplastics can enter the bodies of fish, and the degree to which each of these pathways occurs may vary according to the feeding ecology of a given species. For example, fish may selectively ingest MPs when they match the characteristics of preferred food items (Ory et al., 2017), indirectly via trophic transfer from their prey (Santana et al., 2017; Welden et al., 2018), or accidentally during foraging, respiration, or through drinking water (Roch et al., 2020).

To evaluate the potential for biomagnification of MPs in marine and freshwater fishes, we conducted a literature review of studies that investigated the ingestion of MPs by marine and freshwater fishes from 123 studies published between January 2011 and June 2020. We used Bayesian generalized linear mixed models (GLMMs) to explore how geographic, methodological, and ecological factors (including trophic level and body size) influenced reported MP concentrations and occurrence rates in fish digestive tracts. This work will further advance the study of MPs and their ecotoxicology by determining what factors predict global ingestion of microplastics by fishes.

2. Material and methods

2.1. Data collection

We used the search terms “fish”, and “microplastic”, “plastic”, or “litter” to identify English-language peer-reviewed studies (via Web of Science™, the world’s leading scientific citation search platform) that measured the number of MPs in the digestive tract of marine and freshwater fishes. We also informally identified candidate studies by monitoring Google Scholar alerts for the terms “microplastic” and “microplastics” from January 2016 through June 2020. Publications were included in our analysis if average MP concentrations (defined as particles less than 5 mm along their longest dimension) in the digestive tracts of a particular fish species were reported, could be calculated, or if occurrence rates (proportion of fish containing MPs) were reported. We excluded studies if they did not explicitly report numbers in terms of MPs and instead examined a larger range of particle sizes. When studies further divided a species into groups (*e.g.*, by site, age, year) and reported separate MP ingestion numbers and/or rates, we calculated totals for the species using the overall sample size and pooling across individuals. Studies or data points were also excluded for individuals that were raised in aquaculture facilities, when the species-specific sample size was not reported, when the entire gastrointestinal tract was not isolated and analyzed for MPs, or when different digestion methodologies were used within a study for different samples of the same species.

This process resulted in a total of 123 useable studies (see Supplementary Materials for a full list of references). We also recorded data from each study on the sample size, lowest detectable particle size, whether polymer ID was performed (*i.e.*, FTIR or Raman spectroscopy, or pyrolysis), whether fibers were excluded and whether blanks were used to control for background contamination. Researchers sometimes exclude fibers when they are uncertain of their ability to detect them and/or distinguish them from background contamination. This practice is becoming less common, as the field advances, but still has potential implications for the interpretation of older studies. Data on the trophic level and habitat (*e.g.* bathydemersal, pelagic, demersal) for each fish species were added using the online resource FishBase (Froese and Pauly, 2019). We also recorded the Food and Agriculture Organization (FAO) major fishing area from which the samples in each study were collected. FAO major fishing areas were used since the designations include open ocean areas and inland waterbodies, as many of the studies that we utilized were not limited to coastal areas. We defined the lowest detectable particle size for a study as the pore or mesh size of the smallest filter or sieve used when preparing the samples for analysis, if the authors used a digestion step to prepare their samples for filtration. When samples were not pre-digested, we set the lowest detectable particle size at 500 μm. While it is difficult to know for certain what the lowest detectable particle size would be when a fish digestive tract is only analyzed via dissection and thus contains a lot of biological material, 500 μm seems reasonable for some mathematical reasons. Of 35 studies that did not digest and filter samples, 13 reported the smallest size of particle detected. Most of these numbers (8 of 13) were <200 μm, and the lowest size was 20 μm. Thus, if the probability of detecting a particle, p, is linearly and logistically related to the size of the particles, s:

$$\log\left(\frac{p}{1-p}\right) = a + b(s)$$

Assuming a 50% probability of detecting a 200-μm particle and 5% probability for a 20-μm particle, there would be a 75% probability of detecting a 500-μm particle. This is obviously a rough estimate but provides evidence that 500 μm is an appropriate estimate for the size cutoff for a high probability of noticing a potential MP particle in a complex sample.

2.2. Data analysis

Bayesian modeling was carried out using JAGS (Plummer, 2003), implemented in R v4.0.0 (R Core Team, 2020) using the R2jags package (Su and Yajima, 2020). Generalized linear mixed models (GLMMs) are powerful tools for modeling heterogeneous data as shrinkage due to random effects prevent overfitting for data groupings where the sample size is low (*e.g.*, for certain FAO regions). We fit five GLMMs to the data. Multiple candidate models were explored for each of the five models, including zero-inflated negative binomial, zero-inflated and Poisson, Poisson, and log-normal models for Models 1, 3, and 5, and beta-binomial models for Models 2, and 4. Models were assessed using the DHARMa package (Hartig, 2020), and the final models were chosen because their simulated scaled residuals plots did not suggest misspecification. We ran three Markov Chain Monte-Carlo (MCMC) chains for each model. When fitting models, the number of MCMC iterations was increased until $\widehat{R}$ values, a standard convergence metric, for each estimated parameter reached 1.01 or lower. For models 2 and 4 we added a study-level random effect to deal with heterogeneity in the simulated residuals. For all models, when a variable is indicated to be standardized, this was done by subtracting the mean and then dividing by the standard deviation for that variable from that dataset. A full description of the structure used for each model can be found in Text A.1.

2.2.1. *Model 1 – Effect of trophic level on MP concentration in digestive tracts*

We used this model to determine the effect of numerical trophic level, as estimated by FishBase, on the number of MPs contained in fish digestive tracts while controlling for FAO area, sample size, environment, and several methodological elements. The model was run using all the literature-extracted data where the average number of MPs contained in the digestive tracts of individuals from a species of fish in a study were either directly reported or could be calculated. This resulted in 735 data points from 106 studies spanning 550 species. The model originally included the environment occupied (*e.g.*, bathydemersal, pelagic, demersal) by the fish species according to Fishbase, but the addition of this variable did not improve model fit and the posteriors were not different among the groups, so the variable was excluded from this analysis (and all other models). We ran the model for 75,000 iterations with a burn-in of 5000 and at thinning factor of 25.

2.2.2. *Model 2 – Effect of trophic level on MP occurrence rate*

The second model investigated the effect of trophic level on the occurrence rate of MPs in fish digestive tracts while controlling for FAO area and methodological differences. The model was run using all literature-extracted data where the sample size and number of individuals containing MPs for a species of fish in a study were reported. This resulted in 642 data points from 108 studies and 478 species. We ran the model for 7000 iterations with a burn-in of 1000 and a thinning factor of 4.

2.2.3. *Model 3 – Effect of body size on MP concentration in digestive tracts*

The third model was used to determine the effect of body size on the number of MPs contained in fish digestive tracts while controlling for FAO area, lowest detectable particle size, and whether or not fibers were excluded (as these predictors all had some effect in Model 1). The model was run using all literature-extracted data where the average number of MPs contained in the digestive tracts of individuals from a species of fish in a study were either directly reported or could be calculated, as well as whether the average total length for the individuals of a species was reported. This resulted in 395 data points from 62 studies for 327 species. We ran the model for 50,000 iterations with a burn-in of 5000 and a thinning factor of 10.

2.2.4. *Model 4 – Effect of body size on MP occurrence rate*

The fourth model was used to determine the effect of body size on the occurrence rate of MPs in fish digestive tracts while controlling for FAO area, lowest detectable particle size, and whether fibers were excluded. The model was run using all literature-extracted data where the sample size and number of individuals containing MP for a given species of fish in a study were reported, and if the average total length for the individuals of a species was reported. This condition resulted in 258 data points from 48 studies spanning 215 species. We ran the model for 100,000 iterations with a burn-in of 2000 and a thinning factor of 50.

2.2.5. *Model 5 – Effect of family on MP concentration in digestive tracts*

The fifth and final model was used to determine the effect of taxonomic family on the number of MPs contained within fish digestive tracts while controlling for lowest detectable particle size, body size, and whether fibers were excluded. We derived the data for this model from the data for Model 3 by selecting only data for which there were at least 10 points from a taxonomic family. This resulted in 180 data points from 46 studies for 133 species from 12 families. We ran the model for 70,000 iterations with a burn-in of 5000 and a thinning factor of 40.

3. Results

Average MP concentrations in fish digestive tracts ranged from 0 to 40 particles individual^{-1}, with a global mean ± SD of 2.11 ± 3.81 (standard error of the mean of 0.14) and a median of 1.00 particle individual^{-1}. Reported mean concentrations were slightly higher in freshwater (2.84 ± 4.18 particles individual^{-1}) than the marine environment (2.03 ± 3.76 particles individual^{-1}), although overlap between the FAO area parameter posteriors did not suggest a meaningful difference between freshwater and marine environments. The highest reported concentrations were for the eastern central Atlantic and the western central Pacific, and the lowest for the Antarctic Indian Ocean and the northwestern Atlantic (Fig. 1). There was no strong or consistent relationship between trophic level and MP concentration in fish digestive tracts GLMM (Model 1), with the trophic level by FAO area random slope parameter posteriors showing substantial overlap with both zero and each other (Fig. A.1). Simulating from this model by holding all other parameters constant (*i.e.*, other than the random slope and intercepts) demonstrated that the only substantial differences according to trophic level were a negative correlative relationship for South American freshwater, and for southwest Pacific marine fishes (Fig. 2). The FAO areas with lower reported MP concentrations tended to have elevated lowest detectable particle sizes (Fig. 1). Extrapolating from our model suggests that skipping a digestion step and/or only analyzing the material left on a >100 μm sieve would result in the underestimation of MP concentrations by up to a few particles compared with digesting and filtering through a <100 μm filter (Fig. 3). Excluding fibers from a study also lowered the number of MPs reported in fish digestive tracts (Fig. 3). The use of either a polymer ID method or blanks did not have a substantial effect on reported MP concentrations (Fig. A.1).

Occurrence rates (the proportion of individuals within a species in each study having MPs in their digestive tracts) were higher in freshwater environments (0.56 ± 0.30) than in marine environments (0.31 ± 0.34). The highest mean occurrence rates in fresh water were reported in North America and Asia. The highest occurrence rates in the marine environment were in the western central Pacific, the northwest Pacific, and the Mediterranean and Black Sea (Fig. 4). The lowest reported marine occurrence rates occurred in the northwest Atlantic, the Antarctic Indian Ocean and the southwest Pacific. However, after accounting for the effects of trophic level, lowest detectable particle size, whether fibers were excluded, and study identity, the trophic level occurrence rate binomial GLMM (Model 2) predicted that the Mediterranean and Black Sea and southeast Pacific had the highest occurrence rates and the southwest Pacific the lowest occurrence rates for the marine environment. The former areas came out as higher following modeling as other FAO areas had many more zero MP occurrence reports and wider spreads in occurrence rates across trophic levels (Fig. A.2). There were very few (<10) data points for African fresh water, the eastern central and northwest Atlantic, the Antarctic Indian Ocean, and the northeast Pacific. There was no consistent relationship between trophic level and MP occurrence rate among FAO areas, although some patterns emerged within particular areas, which were diagnosed from the model when the posterior for the trophic level by FAO area slope minimally overlapped with zero (Fig. A.2). A relatively strong decrease in occurrence rate with trophic level – from over 50% to less than 25% ingestion – was observed for the northwest and southwest Pacific, as well as for North American fresh water. The model predicted an increase in MP concentration with trophic level for the eastern and western central Atlantic and for the Mediterranean and Black Sea (Fig. 5). Occurrence rate was negatively correlated with lowest detectable particle size. In contrast to Model 1, the exclusion of fibers was associated with higher reported MP occurrence rates. There was a high degree of variation by study according to the random effect standard

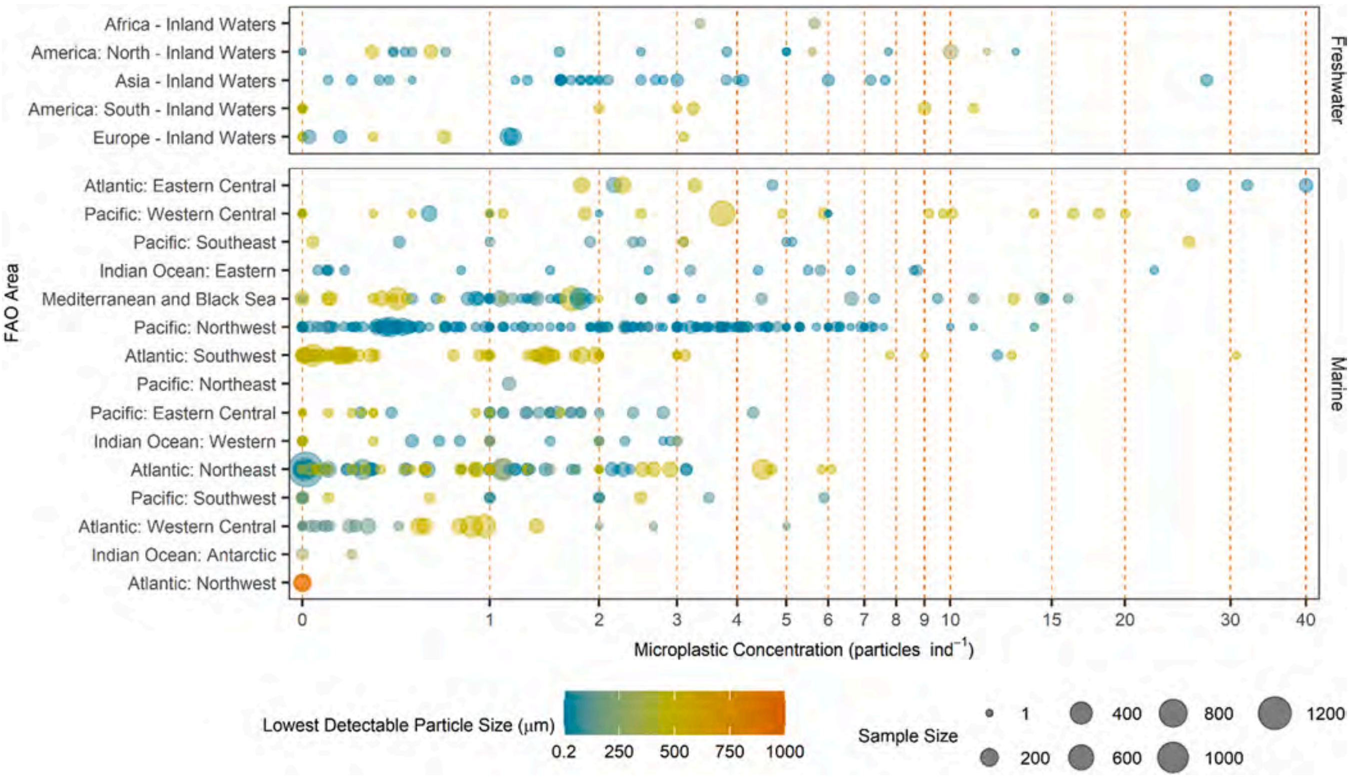

Fig. 1. Average microplastic concentrations (particles individual^{-1}) in the digestive tracts of fish from 104 studies and 552 species, plotted on a log scale within Food and Agricultural Organization major fishing area from highest to lowest microplastics concentration (top to bottom) by overall mean. Symbol sizes are proportional to sample size (number of fish from a species) used by each study. The points are colored according to the lowest detectable particle size (μm), from blue to orange with increasing size. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

deviation parameter, with a mean posterior estimate of 2.10 log-odds, or 0.89 in probability units (after logistic transformation).

According to the body size MP concentration GLMM (Model 3), the lowest detectable particle size had a strong negative correlation with MP concentration in fish digestive tracts, like Model 1 (Fig. A.3). The posteriors for the random slope by FAO area and total length generally overlapped heavily with zero, other than a positive correlation between total length and MP concentration for the northwest Pacific and negative correlations for the southwest Atlantic, the Mediterranean and the Black Sea, the eastern central Atlantic, and North American fresh water (Fig. A.3). As shown in Fig. 6, simulating from the model reveals a high uncertainty in the relationship between body size and MP concentration in fish digestive tracts, especially for larger body sizes (>50 cm) for which there were few data points.

According to the body size occurrence rate model (Model 4), MP occurrence rate was consistently and positively correlated with body size, with practically no difference in slopes among FAO areas (Fig. A.4). Simulating from the model suggested that although there is variation in occurrence rates by FAO area, in general, it might be expected that occurrence rate would rise from <50% to >50% moving from a body size of 1–200 cm (Fig. 7). However, uncertainty is also higher for larger body sizes, for which there were fewer data points.

The taxonomic family MP concentration GLMM (Model 5) revealed some overlap in the random intercept posteriors for family (Fig. A.5). Controlling for other variables and simulating from the model suggested that higher gut MP concentrations (or at least a wider spread in concentrations) might be expected for clupeids as compared to sparids, sciaenids, mugilids, gobiids, engraulidis, cyprinids, acanthurids, scombrids, gerreids, and carangids (Fig. 8). In the raw data, clupeids (*e.g.*, herring, shads, sardines, menhadens; mostly planktonic feeders) had on average nearly twice as many particles in their digestive tracts than most other families, with mean ± standard error gut MP concentrations of 6.15 ± 0.78 particles individual^{-1} compared with 3.61 ± 0.56 for sparids (porgies; benthic feeding carnivores) and <3 particles individual^{-1} for other families. Carangids (jacks and pompanos; large, fast-swimming, marine predators) and gerreids (mojarras; small, benthic feeders) were the lowest at 0.91 ± 0.07 and 0.71 ± 0.09 particles individual^{-1} respectively.

4. Discussion

4.1. Regional differences

The two models with the most data (Models 1 and 2) suggest that gut MP concentrations and the proportion of individuals of a given species that have ingested MPs do not necessarily show the same geographical patterns. According to the trophic-level MP gut concentration model (Model 1), concentrations of MPs in fish guts were higher in African and North American freshwater areas, the eastern central Atlantic, eastern Indian Ocean, southeast Pacific, and western central Pacific than in the western central Atlantic and southwest Pacific. Freshwater fish had both more MPs in their digestive tracts and were more likely (on average) to have any MPs in their digestive tracts. Chen et al. (2020) found that freshwater fish MP concentrations were highest in Asia, a trend not replicated in our analysis. The trophic-level occurrence rate model suggests that fish in the southeast Pacific more commonly contained MPs, which were less common in the northeast and northwest Atlantic, the eastern and western Indian Ocean, the eastern central Pacific, the southwest Pacific, and European fresh water. Taken together, it can be inferred that fish ingested MPs more frequently and had more MPs in their digestive tracts (on average) in the southeast Pacific (the South American west coast), but less frequently and with lower MP concentrations in the western central Atlantic (the Gulf of Mexico and the Caribbean Sea) and the southwest Pacific (Pacific islands and southeast

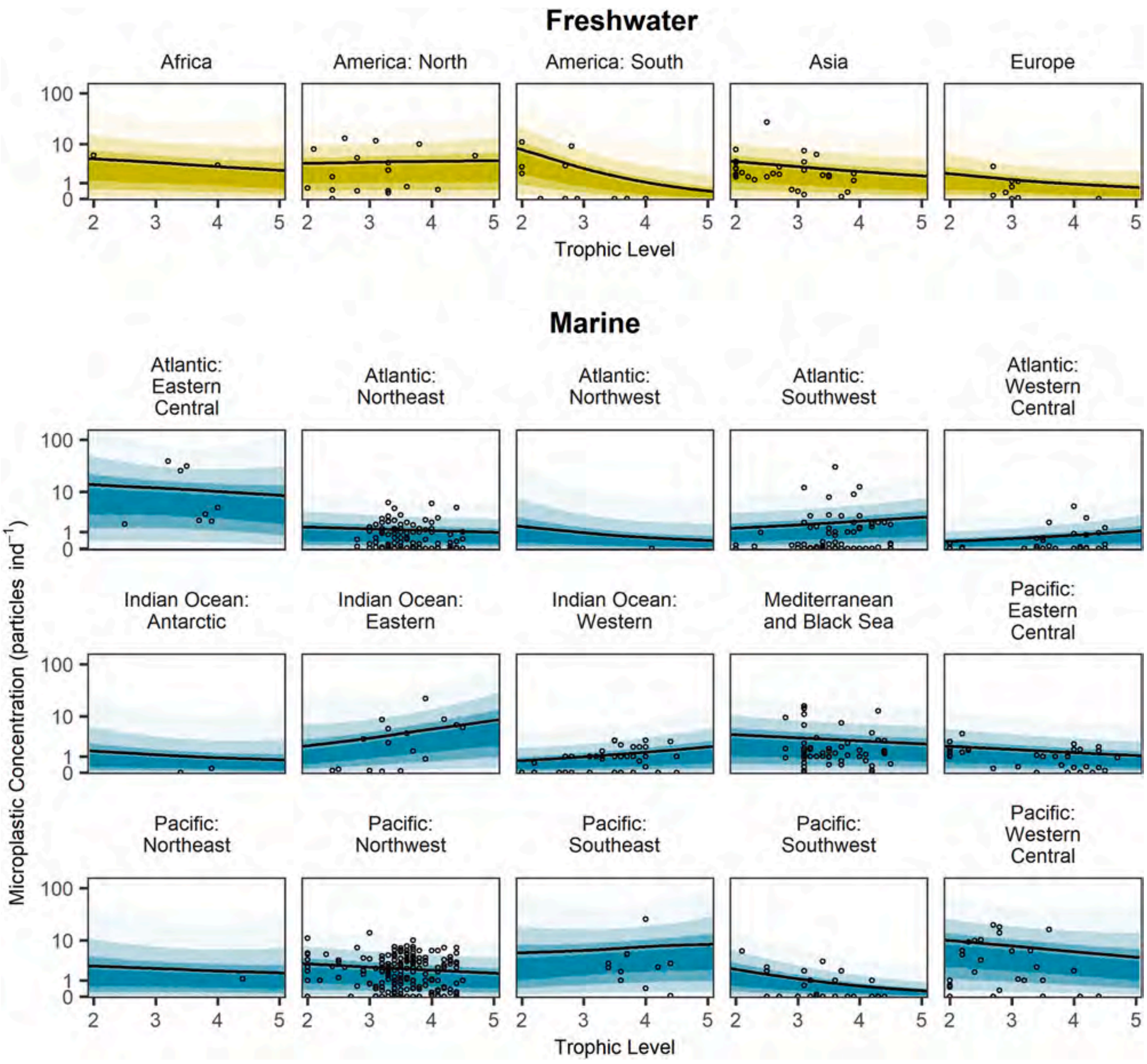

Fig. 2. Simulations from Model 1 holding all variables other than Food and Agricultural Organization region and trophic level constant. Plots show microplastics gut concentration in particles individual^{-1} on a log scale on the y-axis, and trophic level on the x-axis. We used MCMC chain samples to generate posterior predictive distributions over 7000 new observations, holding lowest detectable particle size at 100 µm, assuming that fibers were not excluded, blanks were used, and polymer identification was used. We generated sample sizes from a Poisson distribution with lambda (the mean sample size in the original data) of 38.7 and used trophic level values ranging from 1.9 to 5.9 (evenly spaced). Points represent the data used to build the model, solid lines are medians of the posterior for the mean, and the increasingly dark ribbons represent the 25%, 50%, 75%, and 95% highest posterior density intervals for predicted data.

coast of Australia). These results contrast with some other studies which commonly report that MP concentrations in the ocean – and thus ingestion risk for organisms – are highest in the northwest Pacific and in the Mediterranean Sea (Xu et al., 2020). In our analysis, these regions emerged close to the average in terms of both digestive tract MP concentrations and MP occurrence rate, potentially due to our ability to control for other factors such as trophic level and analytical methods. As mentioned above, however, this result may also be related to the feeding strategies of the most abundant fish species in each area.

It should be noted that many key geographical areas were data-poor (*i.e.*, the Arctic, the northwest Atlantic, the eastern central Atlantic, the southeast Atlantic, the Antarctic Atlantic, Indian, and Pacific, the northeast Pacific, and fresh water of Europe, Africa, and Oceania.). Our models are robust to the issue of low sample size in these areas, as FAO area was specified as a random effect in all models and uncertainty propagated through to predictions. Nevertheless this highlights the need for increased study of MPs in fish in the Arctic and Southern Ocean due to the high prevalence of microplastics and non-plastic microfibres in the surface waters of these areas (Barrows et al., 2018). This is especially salient for the Arctic, where many indigenous peoples rely heavily on subsistence hunting and fishing and have thus historically been highly exposed to pollutants (Van Oostdam et al., 2005). As the ocean warms, these regions will also become more accessible to commercial fisheries. There is a further need for more data on the ingestion of MPs by freshwater fish (Rochman, 2018). In Models 1 and 2, only 10.1% and 8.5% of the data points, respectively, were for freshwater fish.

4.2. *Effect of trophic level, body size, and taxonomy*

There was no effect of trophic level on either MP gut concentrations or MP occurrence rates beyond a change in a few particles per individual for different FAO areas. This result agrees with other recent reviews

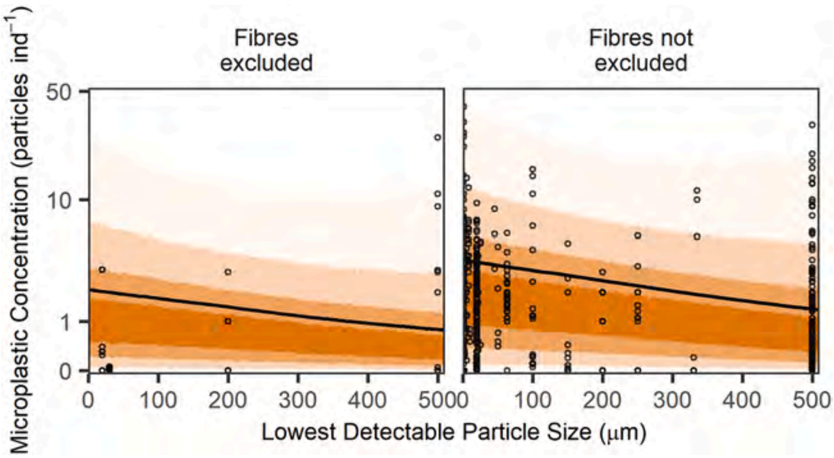

Fig. 3. Simulations from Model 1 holding all variables constant other than lowest detectable particle size and whether fibers were excluded. Plots show microplastics gut concentration in particles individual^{-1} on a log scale on the y-axis, and lowest detectable particle size in μm on the x-axis. We used Markov chain Monte Carlo samples to generate posterior predictive distributions over 7000 new observations, with lowest detectable particle size values ranging from 0.5 to 520 μm (evenly spaced), and assuming that blanks and polymer identification were used, and that the fish came from the northwest Pacific (*i.e.*, where the largest number of samples were present in the original data). We generated sample sizes from a Poisson distribution with lambda 38.7 (the mean sample size in the original data). The points represent the data used to build the model, the solid lines the median of the posterior for the mean, and the increasingly dark ribbons the 25%, 50%, 75%, and 95% highest posterior density intervals for predicted data.

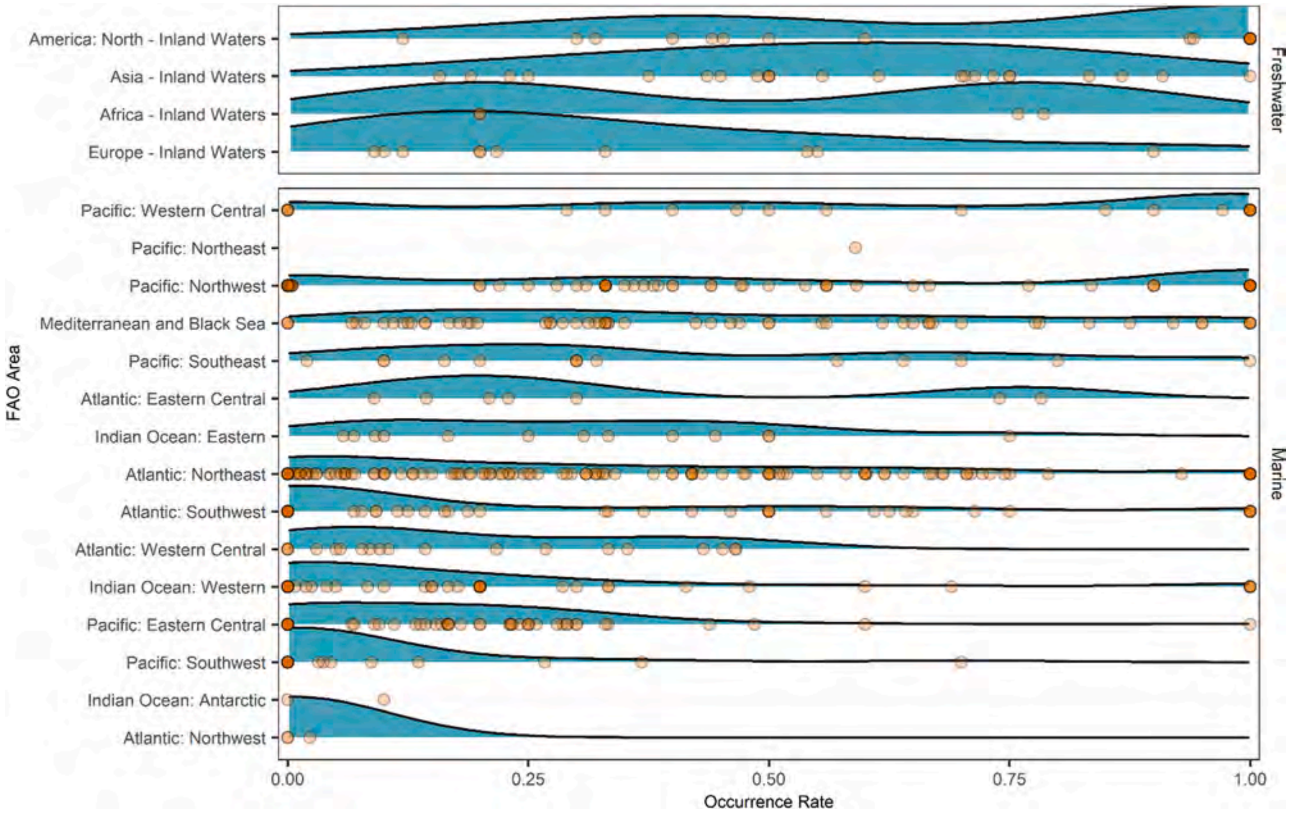

Fig. 4. Occurrence rate (proportion of individuals from a given species in a study having microplastics in their digestive tracts) by Food and Agricultural Organization major fishing area, divided into freshwater and marine environments. The ridges are kernel density estimates, and the points show the raw data. Each data point is consistently shaded, so darker points are multiple overlaid points.

suggesting that MP ingestion does not increase with trophic level (Walkinshaw et al., 2020; Gouin, 2020; Miller et al., 2020). Walkinshaw et al. concluded that lower trophic level marine species are at greater risk of MP ingestion than higher trophic level species (Walkinshaw et al., 2020). Our analysis did not confirm this finding for fish, as there was neither a strong increase nor decrease in digestive tract MP concentration with trophic level. However, occurrence rates increased with increasing trophic level in the eastern and western central Atlantic, and the Mediterranean and Black Sea, and decreased in North American fresh water, the western Indian Ocean, the northwest and southwest Pacific, and the western central Pacific. The largest trends were for the northwest Pacific and southwest Pacific, where the model predicted that occurrence rate would decline from >50% to close to zero going up from a trophic level of two to five.

Many studies have demonstrated trophic transfer of MPs in a variety of species and there has been much speculation about the entry and accumulation of MPs in food webs and the potential for biomagnification. One of the earliest laboratory studies to demonstrate trophic transfer of MPs between predator and prey was by Murray and Cowie in 2011 and demonstrated that the Norway lobster, *Nephrops norvegicus*, ingested and accumulated strands from polypropylene rope that had been incorporated into fish provided as a food source (Murray and Cowie, 2011). However, similar to our meta-analysis of published data for fish digestive tracts, most relevant field studies have found no relationship between trophic level and MP concentrations in digestive tracts, gills, or muscle of various fish and invertebrate species (Welden

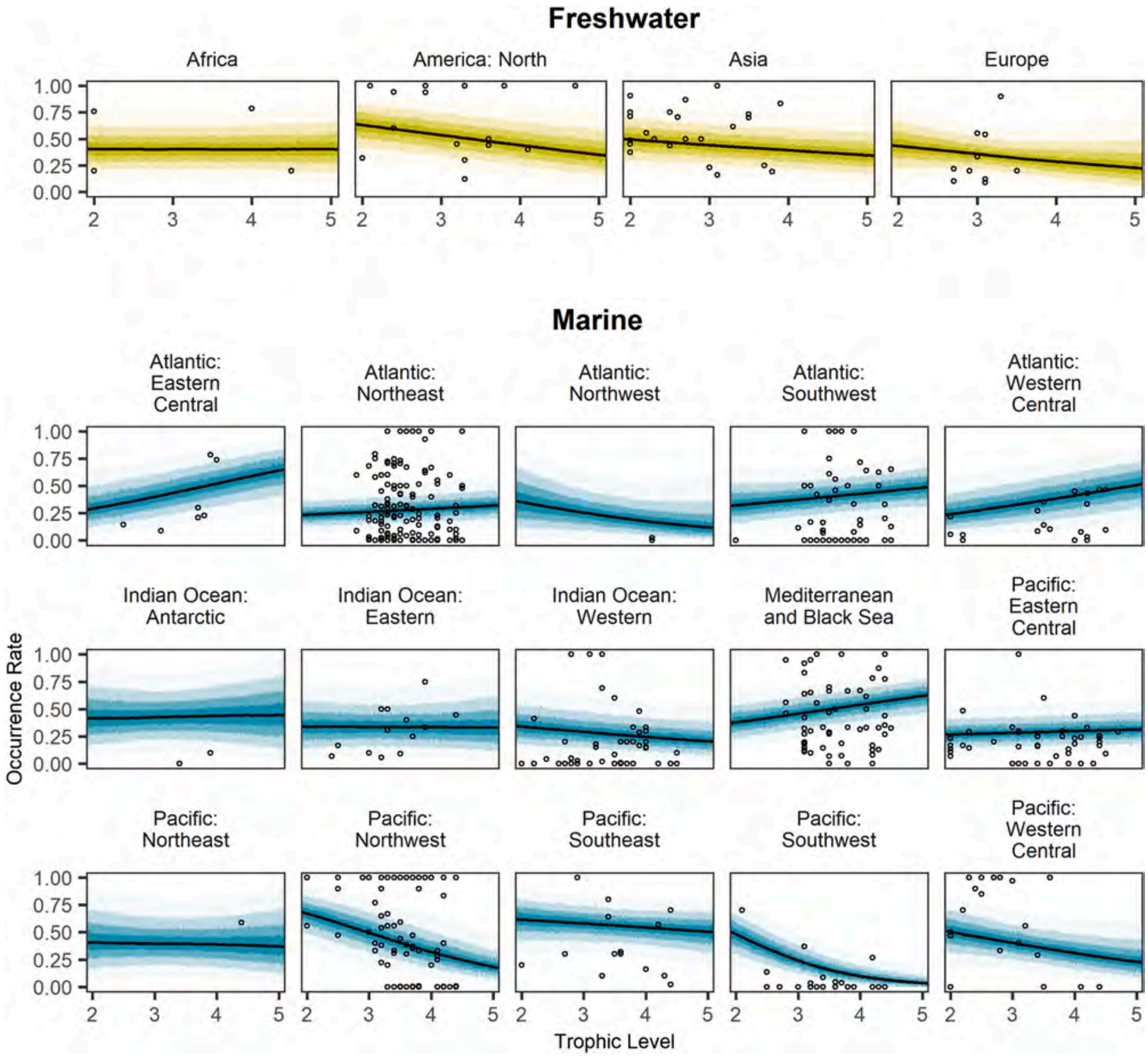

Fig. 5. Simulations from Model 2 holding all variables other than Food and Agricultural Organization region and trophic level constant. Plots show occurrence rate (proportion of individuals of a species from a study having any microplastics in their digestive tracts) plotted on the y-axis, and trophic level on the x-axis. We used Markov chain Monte Carlo samples to generate posterior predictive distributions over 10,000 new observations, holding lowest detectable particle size at 100 μm, assuming that fibers were not excluded, and that the effect of study was ignored (*i.e.,* averaging over that random effect). We generated sample sizes from a Poisson distribution with lambda 38.7 (the mean sample size in the original data) and used trophic level values ranging from 1.9 to 5.9 (evenly spaced). Points represent the data used to build the model, solid lines are the medians of the posterior for the mean, and the increasingly dark ribbons the 25%, 50%, 75%, and 95% highest posterior density intervals for predicted data.

et al., 2018; Güven et al., 2017; Bour et al., 2018; Akhbarizadeh et al., 2019; Filgueiras et al., 2020). One exception is a study from Zhang et al., which found a significant, positive correlation between MP concentrations in gastrointestinal tracts and gills and trophic level across 11 fish and eight crustacean species collect from the East China Sea (Zhang et al., 2019). In another study, Garcia et al. used stable isotope techniques to demonstrate that digestive tract concentrations of 700–5000-μm MPs did not increase with trophic position for freshwater fish and did for macroinvertebrates, but concluded that biomagnification was unlikely (Garcia et al., 2021). Laboratory studies further suggest that even under high MP exposure levels, trophic dilution is the predominant outcome (Sun et al., 2017; Kim et al., 2018; Elizalde-Velázquez et al., 2020).

We detected only a minor effect of body size on overall mean MP gut concentrations, with negative or neutral relationships between average total length and MP concentration. This result occurred everywhere except the northwest Pacific, although for this area there was still a large overlap in highest posterior density interval between the smallest and largest fish in the simulation from 1 to 200 cm. There was, however, a consistent, weak positive correlation between body size and MP occurrence rate for all FAO areas. Taken together, the results from these two models suggest that larger fish are more likely to ingest MPs, but not necessarily to retain more MPs in their digestive tracts. Larger fish need to eat more food than smaller fish due to metabolic scaling (Clarke and Johnston, 1999) and, on average, tend to eat larger prey items, but also a wider range of prey sizes (Scharf et al., 2000). It therefore seems likely that reported MP occurrence rates are higher in larger fish compared with smaller fish due to trophic transfer from prey driving higher probability of ingestion. Indeed, the trophic transfer of MPs from prey to predators has been documented in both the laboratory and the field

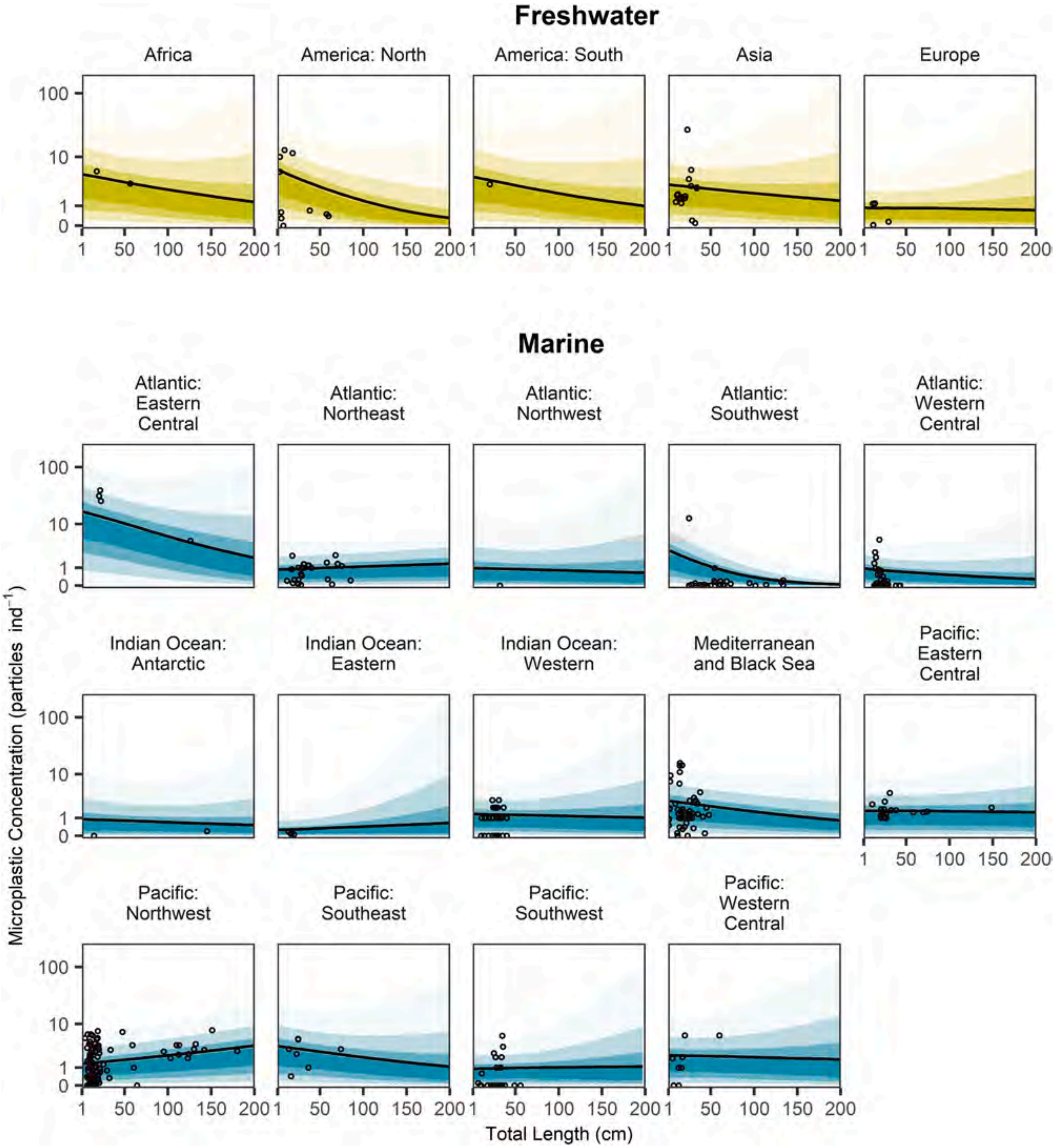

Fig. 6. Simulations from Model 3 holding all variables other than Food and Agricultural Organization region and body size (total length in cm) constant. Plots show microplastics gut concentration in particles individual^{-1} on the y-axis on a log-scale, and total length on the x-axis. We used Markov chain Monte Carlo samples to generate posterior predictive distributions over 7000 new observations, holding lowest detectable particle size at 100 μm, and assuming that fibers were not excluded. We generated sample sizes from a Poisson distribution with lambda 38.7 (the mean sample size in the original data) and used total length values ranging from 1 to 200 cm (evenly spaced). Points represent the data used to build the model, solid lines are the medians of the posterior for the mean, and the increasingly dark ribbons the 25%, 50%, 75%, and 95% highest posterior density intervals for predicted data.

(Welden et al., 2018; Nelms et al., 2018; Setälä et al., 2014). However, these particles are unlikely to remain in the digestive tract long enough to appear as higher MP concentrations in larger fish. As demonstrated by Santana et al. (2017), MPs did not persist in puffer fish (*Sphoeroides greeleyi*) digestive tracts, livers, gonads, or blood longer than 10 days following consumption via trophic transfer from prey. It is also possible that larger fish are more likely to accidentally ingest MPs via drinking water and respiration (Roch et al., 2020).

The trophic-level analysis and the size-based analyses did not produce similar results for several possible reasons. Primarily, body sizes, as reported by the various studies, did not correlate well with FishBase trophic level estimates (Fig. 9). This may be due to differences in ontogeny – which can have a large effect on the feeding behavior of fish, and thus their potential exposure to microplastics (Ferreira et al., 2019) – and the relationship between body size and trophic level which, although generally positive, is not consistent across different species

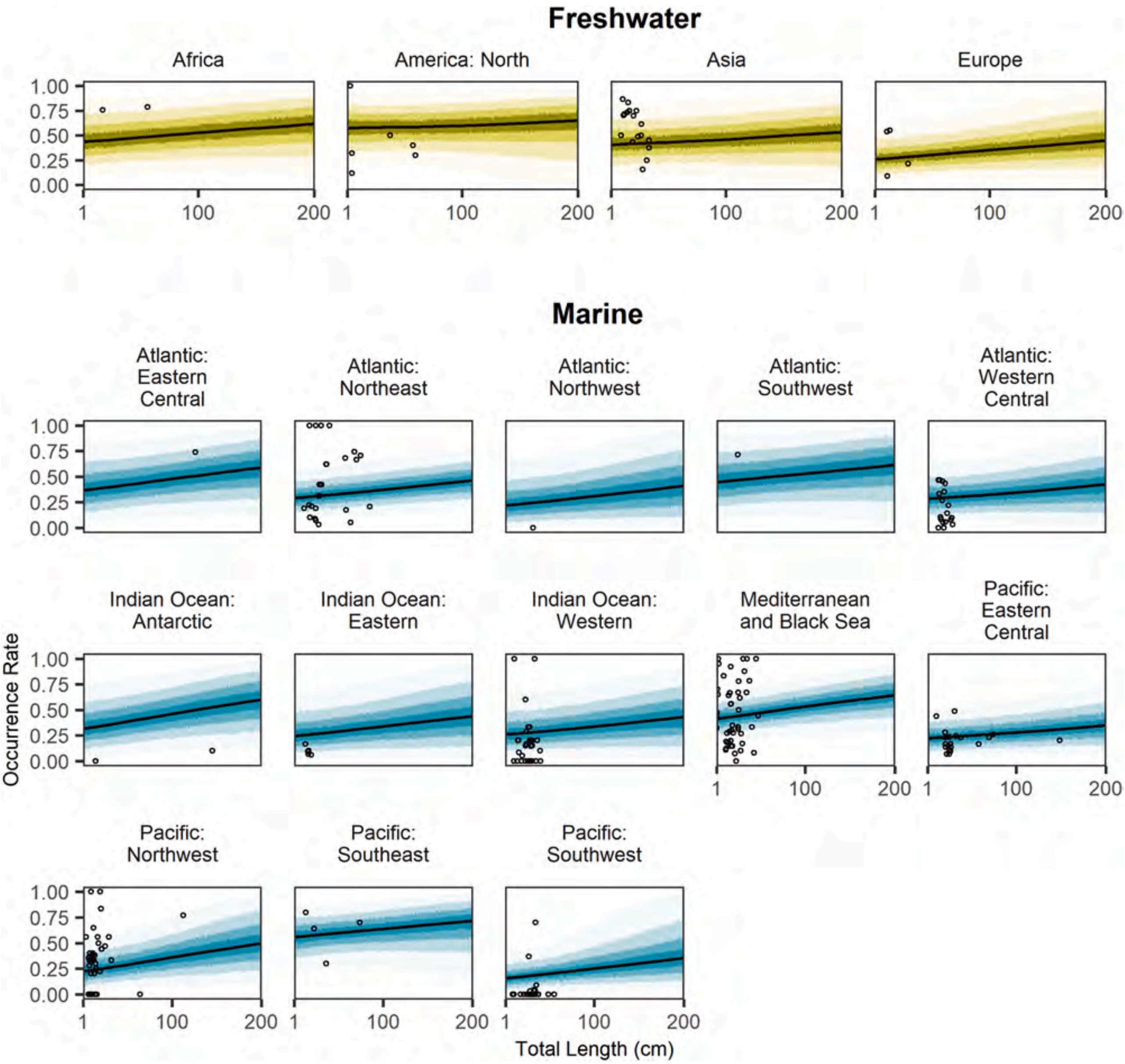

Fig. 7. Simulations from Model 4, holding all variables other than Food and Agricultural Organization region and body size (total length in cm) constant. Plots show microplastics occurrence rate (proportion of individuals of a species from a study having any microplastics in their digestive tracts) on the y-axis, and total length on the x-axis. We used Markov chain Monte Carlo samples to generate posterior predictive distributions over 10,000 new observations, holding lowest detectable particle size at 100 μm, and assuming that fibers were not excluded, and that the effect of study was ignored (averaging over that random effect). We generated sample sizes from a Poisson distribution with lambda 38.7 (the mean sample size in the original data) and used total length values ranging from 1 to 200 cm (evenly spaced). The points on the plot represent the original data used to build the model, the solid lines the median of the posterior for the mean, and the increasingly dark ribbons the 25%, 50%, 75%, and 95% highest posterior density intervals for predicted data.

(Olson et al., 2020). Two interpretations are possible. Either FishBase estimates of trophic level are largely unreliable for this type of analysis due to the aforementioned factors, or body size is simply more predictive of MP occurrence rates due to greater ingestion of food in general by larger fish, rather than any relationship between trophic level and MP ingestion. This could be explored further by relating metabolic scaling and allometry to MP consumption in fish in a laboratory setting with a variety of prey sizes available that have also been exposed to MPs in their diet.

While the size-based analysis suggests that larger fish are more likely to have MPs in their digestive tracts, our taxonomic analysis suggests that small, planktonic feeders are the most likely to have more MPs in their digestive tracts. Clupeids had approximately twice as many (or more) MPs in their digestive tracts compared to other fish families, including several reports of >10 particles individual^{-1}. Clupeids are generally mid-water feeders that either selectively ingest phytoplankton and/or zooplankton, or semi-selectively filter particles from the water column using their gill rakers, and are often able to switch between the two feeding strategies (James, 1988). According to Drenner et al. (1986), the filter-feeding clupeid *Dorosoma cepedianum* displayed increasing filtration efficiency with larger particle size, with the highest efficiency for polystyrene microspheres >40 μm in diameter. This suggests that filter-feeding clupeids would ingest high quantities of the MP size ranges considered in this analysis when they are available in the water column. However, the clupeids with higher MP concentrations in our datasets were all classified as selective plankton feeders or macrofaunal hunters, according to FishBase (Fig. 10). When feeding selectively, it is possible that clupeids might ingest high concentrations of

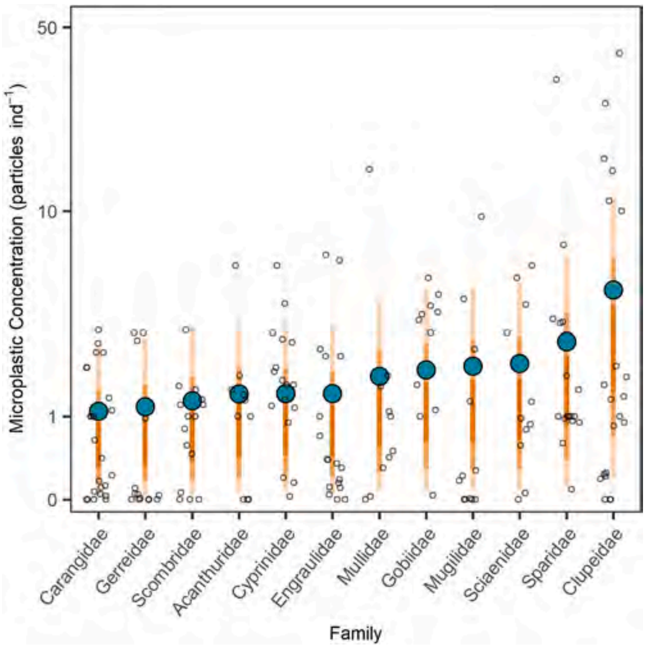

Fig. 8. Simulations from Model 5 holding all variables other than taxonomic family constant. Plots show gut microplastics concentration in particles individual^{-1} on a log scale on the y-axis, and family on the x-axis, arranged left to right by increasing average microplastics concentration. We used Markov chain Monte Carlo samples to generate posterior predictive distributions over each of the 12 families, holding lowest detectable particle size at 100 μm, total length at the mean total length in the data (24.38 cm), and assuming that fibers were not excluded. We generated sample sizes from a Poisson distribution with lambda 38.7 (the mean sample size in the original data). The smaller points in the plot represent the original data used to build the model, the larger points the median of the posterior for the mean, and the increasingly light line segments the 25%, 50%, 75%, and 95% highest posterior density intervals for predicted data.

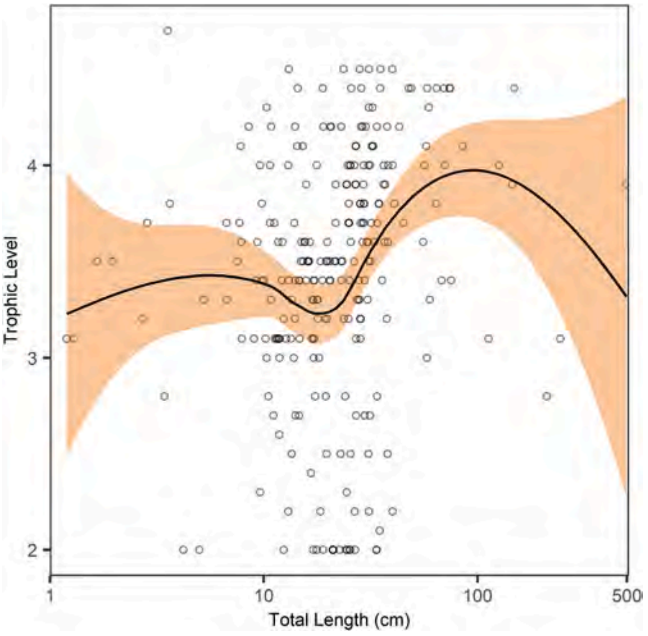

Fig. 9. Relationship between trophic level (from FishBase) and total length in cm, plotted on a log-scale. The line and ribbon are a LOESS (locally estimated scatterplot smoothing) fit and 95% confidence interval.

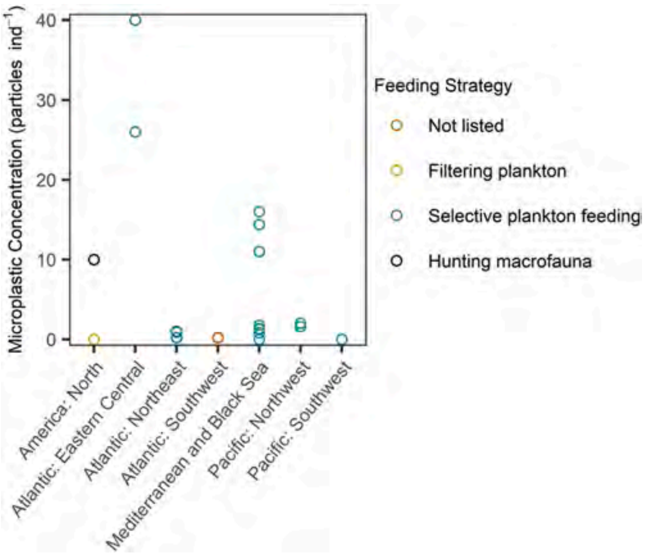

Fig. 10. Clupeid microplastic gut concentrations reported by analyzed studies, separated by Food and Agricultural Organization major fishing area where the fish were collected and feeding strategy, as reported by FishBase.

MPs due to the comparable size and similar appearance between MPs and their preferred prey items (Ory et al., 2017). MPs have been detected in 80% of livers of the European anchovy, *Engraulis encrasicolus*, supporting the idea that small forage fish such as clupeids might be at the greatest risk of ingesting higher concentrations of microplastics (Collard et al., 2017). However, low MP occurrence rates (2%) for Pacific Herring (*Clupea pallasii*) in the northeast Pacific suggest that all clupeids are not necessarily at greater risk of MP ingestion (Hipfner et al., 2018).

4.3. Methodological differences

As noted by several existing studies and reviews, methodology and reporting style play key roles in the scientific quality of MP ingestion studies (Dehaut et al., 2019; Hermsen et al., 2018, 2017; Provencher et al., 2020). Some of the most important suggestions for quality control in microplastics studies include the use of procedural blanks, chemical verification of potential MP particles, and extraction techniques - including whether or not digestion and filtration are carried out, and through what mesh size. In our analysis, we also tested the effect of excluding fibers. Our modeling results suggest the two most important methodological predictors of reported MP concentrations are (i) whether fibers were counted and (ii) the lowest detectable particle size.

We did not directly analyze differences produced by digestion and density separation methods, as there are a variety of combinations of chemicals, temperatures, and durations used by researchers during these processes. We used mesh size of the filter as both a direct measure of the smallest particle size that researchers could retain in their analyzed samples, as well as how thoroughly they had cleaned their samples. In general, more thorough digestion and separation procedures allow for smaller mesh sizes and should allow for easier visualization and detection of MPs. As shown in Fig. 3, the exclusion of fibers and the use of a large-meshed sieve or filter (or no digestion and filtration at all) is likely to result in reported concentrations of <1 particle individual^{-1} for fish digestive tracts. In contrast, counting fibers and processing/filtering samples down to <100 μm would result in estimates in the 1–10 particles individual^{-1} range.

It is important to note that current methods are not particularly good at detecting MPs < 100 μm, so our predictions are limited to more traditional methods, such as visual inspection of filters. Future methods that involve automated counting and spectroscopy of filters, as well as pyrolysis-GCMS methods, have the potential to result in exponentially higher MP counts due to the presence of smaller particles, which are not captured in this analysis (Roch et al., 2019). This may be particularly important given that at sizes < 100 μm MPs can translocate to (and potentially accumulate in) liver and other tissues (Collard et al., 2017; Abbasi et al., 2018; Akhbarizadeh et al., 2018; Avio et al., 2015; Crooks et al., 2019) where they may cause an oxidative stress response and inflammation, among other adverse outcomes (Ding et al., 2018; Lu et al., 2016). Although we did not detect biomagnification of MPs in fish digestive tracts, it is possible that these smaller particles might biomagnify in other tissues.

5. Conclusions

Our work suggests that larger MPs do not biomagnify in marine and freshwater fishes. Although accumulation could still be occurring – *i.e.*, a standing stock of MPs might exist in the bodies of fish if they are not being excreted fast enough – in some regions and taxa, we cannot make a definitive conclusion on this. It appears more likely that higher exposure to MPs *via* direct and indirect ingestion and/or slower rates of depuration can cause larger and higher trophic level fish to contain more MPs in their digestive tracts in certain regions. However, the overall increase in contamination is not large – and certainly nowhere near the exponential increase that would be expected if true biomagnification were occurring. Small planktivorous fish, such as clupeids, may be at the highest risk of ingesting MPs as the particles may resemble their preferred planktonic food. As the ingested volume and surface area of MPs will be higher relative to their body size compared with larger fish, Clupeids may also be at greater risk of health effects from both the physical and chemical effects of MPs. As a caveat, we only effectively consider larger microplastics ($>$100 μm) in this study, whereas small microplastics (1–100 μm) may still have the potential to bioaccumulate and biomagnify and thus pose a much greater ecological risk. In addition, methodological differences in sample collection and processing can influence reported gut concentrations of MPs (as well as occurrence rates). This highlights the need for researchers to carry out purification of their samples via digestion, and density separation where necessary, if their results are to be useful for quantifying the extent and ecological risks of MPs. Finally, our analysis reveals data gaps for MP ingestion by fish from certain geographical regions, especially the Arctic and Southern Oceans.

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CRediT authorship contribution statement

Garth A. Covernton: Conceptualization, Methodology, Software, Formal analysis, Investigation, Data curation, Writing - original draft, Visualization, Funding acquisition. **Hailey L. Davies:** Investigation, Writing - review & editing. **Kieran D. Cox:** Writing - review & editing, Funding acquisition. **Rana El-Sabaawi:** Writing - review & editing. **Francis Juanes:** Resources, Writing - review & editing, Supervision, Funding acquisition. **Sarah E. Dudas:** Writing - review & editing, Supervision, Funding acquisition. **John F. Dower:** Resources, Writing - review & editing, Supervision.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

All data and code used in this publication is available in the following repository: https://doi.org/10.5281/zenodo.4477765.

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.jhazmat.2021.125405.

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