Freshwater food webs amplify microplastic transfer to fish

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The following is included as supplementary information for this paper:
Supplementary Methods - Statistical rationale and additional details for the second meta-analysis
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Supplementary Methods − Statistical rationale and additional details for the second meta-analysis

Decile-stratified freshwater–marine contrasts conditioned on environmental exposure

Environmental MP concentrations in the global dataset span several orders of magnitude owing to heterogeneous sources, transport and measurement protocols. To compare freshwater and marine fish at comparable exposure levels and to reduce leverage from extremes, we divided observations into 10 equally sized groups (deciles) based on the mean-centred log10 environmental MP concentration. Deciles lacking records from both environments were omitted from contrasts.

Within each decile, freshwater–marine differences in fish MP burden (total and by compartment) were tested using Kruskal-Wallis tests in the two-group case, with Monte Carlo *P*-values (1,000 iterations) to stabilise inference under unequal sample sizes and non-normality. Effect sizes are reported as Hodges-Lehmann median differences (freshwater - marine) with 95% confidence intervals. This design explicitly conditions on exposure ("dose-dependent" contrasts) and minimises the influence of extreme values while preserving the rank structure of the data.

Consistency with parametric models

Non-parametric, exposure-conditioned contrasts were used alongside parametric models. Simple log–log regressions related fish MP burden to environmental concentration; between-environment differences were evaluated with tests of parallelism (slope contrasts) and, where appropriate, ANCOVA on intercepts (elevation), with bootstrap resampling. Across compartments where slopes were parallel (e.g., digestive tract and total), the decile-stratified results were concordant with the regression/ANCOVA findings: no slope differences between environments but significant elevation shifts (higher freshwater intercepts at comparable exposure). Where slopes were non-parallel (e.g., muscle, gills), inferences were based on the compartment-specific regression coefficients reported in the main tables.

Implementation notes

All concentration variables were log10-transformed and mean-centred prior to binning and modelling; ln(pBAF) was used for analyses of bioaccumulation ratios.

Decile cut-points were computed on the pooled dataset; contrasts were performed only within deciles containing both environments.

Model diagnostics and additional summaries (including FC1 loadings/variance explained) are reported in the Supplementary Tables referenced in the main text.

Supplementary Table 1 | Morphological and experimental traits of study taxa. Summary of zooplankton (*Simocephalus vetulus, Daphnia galeata, Tigriopus californicus, Acartia tonsa*) and fish (*Poecilia reticulata*) used in laboratory trials; taxonomic class, experimental environment, population source, mean body length, and dry weight at exposure. Morphometric values are means (± 1 SD), measured post-experiment. DW, dry weight.

Species	Class	Environment	Body length (mm)	Mean DW (mg)	Origin	Age (days)
S. vetulus	Branchiopoda	Freshwater	1.26 ± 0.08	0.23 ± 0.05	Lake Roś	6
D. galeata	Branchiopoda	Freshwater	1.34 ± 0.06	0.16 ± 0.03	Lake Roś	6
T. californicus	Copepoda	Marine	1.05 ± 0.06	0.14 ± 0.03	Commercial supplier	18
A. tonsa	Copepoda	Marine	0.76 ± 0.02	0.08 ± 0.01	Hatched from eggs Algova®	18
P. reticulata	Actinopterygii	Brackish	26.88 ± 3.90	107.6 ± 52.6	Commercial supplier	≈ 180

Supplementary Table 2 | Mean microplastic (MP) number and mass per millilitre of medium across treatments. Means (± 1 SD) for MP number and mass per mL of medium across experimental combinations varying by MP size (1 µm, 25 µm in diameter), zooplankton species (*Simocephalus vetulus*, *Daphnia galeata*, *Tigriopus californicus*, *Acartia tonsa*) and a control without zooplankton (C), and by medium type: MP_{zoo} (medium containing MPs previously ingested and excreted by zooplankton) and MP_{water} (MPs added directly to the fish medium). Columns report MP concentration (particles mL^{-1}) and MP mass (µg mL^{-1}). The same letters denote homogeneous groups from Holm-corrected *post hoc* comparisons; values sharing a letter within a column do not differ significantly ($\alpha = 0.05$).

MP size (µm)	Variant	MP concentration (particles mL^{-1})			MP mass (µg mL^{-1})		
		$mean_1$	SD_1	P_1	$mean_2$	SD_2	P_2
	C	4839108	543705	a	2.50	0.30	a
	S. vetulus × MP_{zoo}	5246647	1133227	a	2.88	0.65	a
	D. galeata × MP_{zoo}	5346036	887943	a	2.94	0.49	a
	T. californicus × MP_{zoo}	4975142	1017987	a	2.57	0.56	a
1	*A. tonsa* × MP_{zoo}	5374025	1230850	a	2.95	0.68	a
	S. vetulus × MP_{water}	5367685	1106826	a	2.95	0.61	a
	D. galeata × MP_{water}	5406698	1276960	a	2.97	0.70	a
	T. californicus × MP_{water}	4968507	881671	a	2.73	0.48	a
	A. tonsa × MP_{water}	4965647	848113	a	2.73	0.47	a
	C	298.85	50.99	b	2.48	0.44	a
	S. vetulus × MP_{zoo}	352.33	49.69	b	3.03	0.43	a
	D. galeata × MP_{zoo}	357.50	57.36	b	3.07	0.49	a
	T. californicus × MP_{zoo}	336.44	68.00	b	2.89	0.58	a
25	*A. tonsa* × MP_{zoo}	346.18	82.17	b	2.97	0.71	a
	S. vetulus × MP_{water}	299.83	100.34	b	2.58	0.86	a
	D. galeata × MP_{water}	320.40	80.95	b	2.75	0.70	a
	T. californicus × MP_{water}	307.22	93.75	b	2.64	0.81	a
	A. tonsa × MP_{water}	336.45	65.60	b	2.89	0.56	a

Supplementary Table 3 | Literature dataset for zooplankton foraging selectivity. Record-level inventory for the meta-analysis of microplastic–algae ingestion selectivity in zooplankton. Variables include environment (freshwater, marine), algal species, zooplankton species, algal concentration, algal ingestion rate, ingestion-to-concentration ratio, algal cell size, MP concentration, MP ingestion rate, MP size and shape, polymer type, and bibliographic reference. The curated dataset comprises 127 records from six studies, spanning three freshwater and six marine zooplankton species; inclusion criteria and search strategy are detailed in the *Methods*. Units are given per variable and brief data-quality notes are included where applicable. **Provided as an Excel file.**

Supplementary Table 4 | Global field dataset of fish microplastic burdens and co-located environmental concentrations Study- and sample-level metadata used in the field synthesis. Columns include environment (freshwater, marine), fish species, trophic level, feeding guild, MP concentrations in fish (per individual, per gram of muscle, digestive tract, gills), fish wet/dry mass, body length, wet muscle mass, environmental MP concentration, sampling depth, and reference/study identifiers. After screening (excluding saline lakes, estuaries and rivers), the dataset contains 883 records from 106 publications, covering 398 marine and 104 freshwater fish species; trophic levels and feeding guilds follow FishBase and SeaLifeBase. Environmental concentrations were taken either from the same study that quantified MPs in fish at a given site, or from another publication reporting environmental MPs for the same lake or the nearest marine location to the fish sampling site. See *Methods* for search terms, eligibility criteria, and transformation/normalisation steps. **Provided as an Excel file.**

Supplementary Table 5 | Effects of species identity and particle size on microplastic uptake and bioaccumulation in zooplankton. Analysis of deviance from generalized linear models (gamma distribution) testing the effects of zooplankton species, microplastic (MP) size and their interaction on four response variables: MP mass per individual, MP mass per unit body mass, and bioaccumulation factors (BAFs) calculated on a body-mass and body-volume basis. *Zooplankton body mass* was included as a covariate (italicized in the table). Statistically significant ($\alpha = 0.05$) effects are shown in bold. χ^2, chi-squared statistic; df, degrees of freedom; *P*, two-sided *P*-value.

Parameter	**Factor; interaction**	χ^2	**df**	***P***
MP mass per zooplankton individual	Species	**426.290**	**3**	**<0.001**
	MP size	**37.203**	**1**	**<0.001**
	Zooplankton body mass	*0.279*	*1*	*0.598*
	Species × MP size	**74.015**	**3**	**<0.001**
MP mass per unit zooplankton body mass	Species	**537.696**	**3**	**<0.001**
	MP size	**31.924**	**1**	**<0.001**
	Species × MP size	**70.250**	**3**	**<0.001**
BAF (body mass)	Species	**351.233**	**3**	**<0.001**
	MP size	**16.056**	**1**	**<0.001**
	Species × MP size	**49.262**	**3**	**<0.001**
BAF (body volume)	Species	**66.710**	**3**	**<0.001**
	MP size	**19.624**	**1**	**<0.001**
	Species × MP size	**52.392**	**3**	**<0.001**

Supplementary Table 6 | Pairwise contrasts reveal species- and size-specific patterns in zooplankton microplastic uptake and bioaccumulation. Estimated marginal-mean contrasts from generalized linear models (gamma distribution) comparing MP mass per individual, MP mass per unit body mass, and bioaccumulation factors (BAFs; body-mass and body-volume based) across treatments varying by zooplankton species (*Simocephalus vetulus*, *Daphnia galeata*, *Tigriopus californicus*, *Acartia tonsa*) and MP particle size (1 µm, 25 µm). Statistically significant (α = 0.05) differences are in bold. *E*, contrast estimate; SE, standard error; *z*-, *z*-statistic; *P*, Holm-adjusted two-sided *P*-value.

Parameter	Contrast	*E*	SE	*z*-	*P*
MP mass per zooplankton individual	*S. vetulus* × 1µm - *D. galeata* × 1µm	-0.079	0.208	-0.379	0.704
	S. vetulus × 1µm - *T. californicus* × 1µm	**2.897**	**0.298**	**9.726**	**<0.001**
	S. vetulus × 1µm - *A. tonsa* × 1µm	**3.817**	**0.287**	**13.298**	**<0.001**
	D. galeata × 1µm - *T. californicus* × 1µm	**2.976**	**0.257**	**11.583**	**<0.001**
	D. galeata × 1µm - *A. tonsa* × 1µm	**3.896**	**0.196**	**19.905**	**<0.001**
	T. californicus × 1µm - *A. tonsa* × 1µm	**0.920**	**0.268**	**3.431**	**<0.001**
	S. vetulus × 25µm - *D. galeata* × 25µm	**0.946**	**0.218**	**4.346**	**<0.001**
	S. vetulus × 25µm - *T. californicus* × 25µm	**3.359**	**0.314**	**10.687**	**<0.001**
	S. vetulus × 25µm - *A. tonsa* × 25µm	**3.011**	**0.308**	**9.789**	**<0.001**
	D. galeata × 25µm - *T. californicus* × 25µm	**2.413**	**0.259**	**9.317**	**<0.001**
	D. galeata × 25µm - *A. tonsa* × 25µm	**2.066**	**0.201**	**10.287**	**<0.001**
	T. californicus × 25µm - *A. tonsa* × 25µm	-0.348	0.265	-1.311	0.38
	S. vetulus × 1µm - *S. vetulus* × 25µm	**1.674**	**0.192**	**8.705**	**<0.001**
	D. galeata × 1µm - *D. galeata* × 25µm	**2.699**	**0.162**	**16.664**	**<0.001**
	T. californicus × 1µm - *T. californicus* × 25µm	**2.136**	**0.323**	**6.604**	**<0.001**
	A. tonsa × 1µm - *A. tonsa* × 25µm	**0.868**	**0.142**	**6.099**	**<0.001**
MP mass per unit zooplankton body mass	*S. vetulus* × 1µm - *D. galeata* × 1µm	-0.384	0.181	-2.123	0.055
	S. vetulus × 1µm - *T. californicus* × 1µm	**2.479**	**0.281**	**8.827**	**<0.001**
	S. vetulus × 1µm - *A. tonsa* × 1µm	**2.879**	**0.177**	**16.292**	**<0.001**
	D. galeata × 1µm - *T. californicus* × 1µm	**2.862**	**0.269**	**10.659**	**<0.001**
	D. galeata × 1µm - *A. tonsa* × 1µm	**3.262**	**0.156**	**20.849**	**<0.001**
	T. californicus × 1µm - *A. tonsa* × 1µm	0.400	0.266	1.504	0.133
	S. vetulus × 25µm - *D. galeata* × 25µm	**0.606**	**0.181**	**3.352**	**<0.001**
	S. vetulus × 25µm - *T. californicus* × 25µm	**2.866**	**0.281**	**10.205**	**<0.001**
	S. vetulus × 25µm - *A. tonsa* × 25µm	**2.028**	**0.177**	**11.477**	**<0.001**
	D. galeata × 25µm - *T. californicus* × 25µm	**2.260**	**0.269**	**8.416**	**<0.001**
	D. galeata × 25µm - *A. tonsa* × 25µm	**1.422**	**0.156**	**9.089**	**<0.001**
	T. californicus × 25µm - *A. tonsa* × 25µm	**-0.838**	**0.266**	**-3.151**	**0.006**
	S. vetulus × 1µm - *S. vetulus* × 25µm	**1.709**	**0.198**	**8.608**	**<0.001**
	D. galeata × 1µm - *D. galeata* × 25µm	**2.698**	**0.161**	**16.757**	**<0.001**
	T. californicus × 1µm - *T. californicus* × 25µm	**2.095**	**0.344**	**6.092**	**<0.001**
	A. tonsa × 1µm - *A. tonsa* × 25µm	**0.858**	**0.152**	**5.650**	**<0.001**
BAF (body mass)	*S. vetulus* × 1µm - *D. galeata* × 1µm	-0.293	0.189	-1.552	0.386
	S. vetulus × 1µm - *T. californicus* × 1µm	**2.381**	**0.300**	**7.939**	**<0.001**
	S. vetulus × 1µm - *A. tonsa* × 1µm	**2.865**	**0.212**	**13.507**	**<0.001**
	D. galeata × 1µm - *T. californicus* × 1µm	**2.674**	**0.283**	**9.461**	**<0.001**
	D. galeata × 1µm - *A. tonsa* × 1µm	**3.158**	**0.187**	**16.897**	**<0.001**
	T. californicus × 1µm - *A. tonsa* × 1µm	0.484	0.299	1.622	0.386
	S. vetulus × 25µm - *D. galeata* × 25µm	**0.506**	**0.189**	**2.679**	**0.045**
	S. vetulus × 25µm - *T. californicus* × 25µm	**2.768**	**0.300**	**9.230**	**<0.001**
	S. vetulus × 25µm - *A. tonsa* × 25µm	**1.825**	**0.212**	**8.606**	**<0.001**
	D. galeata × 25µm - *T. californicus* × 25µm	**2.262**	**0.283**	**8.003**	**<0.001**
	D. galeata × 25µm - *A. tonsa* × 25µm	**1.319**	**0.187**	**7.057**	**<0.001**
	T. californicus × 25µm - *A. tonsa* × 25µm	**-0.943**	**0.299**	**-3.157**	**0.011**
	S. vetulus × 1µm - *S. vetulus* × 25µm	**1.882**	**0.214**	**8.800**	**<0.001**
	D. galeata × 1µm - *D. galeata* × 25µm	**2.682**	**0.160**	**16.747**	**<0.001**
	T. californicus × 1µm - *T. californicus* × 25µm	**2.270**	**0.366**	**6.198**	**<0.001**
	A. tonsa × 1µm - *A. tonsa* × 25µm	**0.843**	**0.210**	**4.007**	**<0.001**
BAF (body volume)	*S. vetulus* × 1µm - *D. galeata* × 1µm	0.435	0.192	2.260	0.095
	S. vetulus × 1µm - *T. californicus* × 1µm	**1.106**	**0.288**	**3.844**	**<0.001**
	S. vetulus × 1µm - *A. tonsa* × 1µm	**1.524**	**0.201**	**7.580**	**<0.001**
	D. galeata × 1µm - *T. californicus* × 1µm	0.671	0.275	2.444	0.086
	D. galeata × 1µm - *A. tonsa* × 1µm	**1.089**	**0.182**	**5.986**	**<0.001**
	T. californicus × 1µm - *A. tonsa* × 1µm	0.418	0.281	1.489	0.409
	S. vetulus × 25µm - *D. galeata* × 25µm	**1.308**	**0.192**	**6.804**	**<0.001**
	S. vetulus × 25µm - *T. californicus* × 25µm	**1.581**	**0.288**	**5.495**	**<0.001**
	S. vetulus × 25µm - *A. tonsa* × 25µm	**0.561**	**0.201**	**2.792**	**0.037**
	D. galeata × 25µm - *T. californicus* × 25µm	0.272	0.275	0.992	0.409
	D. galeata × 25µm - *A. tonsa* × 25µm	**-0.747**	**0.182**	**-4.104**	**<0.001**
	T. californicus × 25µm - *A. tonsa* × 25µm	**-1.019**	**0.281**	**-3.629**	**<0.001**
	S. vetulus × 1µm - *S. vetulus* × 25µm	**1.810**	**0.210**	**8.602**	**<0.001**
	D. galeata × 1µm - *D. galeata* × 25µm	**2.683**	**0.172**	**15.579**	**<0.001**

T. californicus × 1µm - *T. californicus* × 25µm	**2.285**	**0.348**	**6.562**	**<0.001**
A. tonsa × 1µm - *A. tonsa* × 25µm	**0.847**	**0.191**	**4.430**	**<0.001**

Supplementary Table 7 | Drivers of MP accumulation in fish across zooplankton prey, particle sizes, and exposure types. Results of deviance analysis from generalized linear models (GLMs) with a gamma distribution, testing the effects of zooplankton species, MP particle size, MP exposure type, and their interactions on three response variables: (i) total MP mass per fish, (ii) MP mass ingested per zooplankton individual, and (iii) MP mass ingested per unit body mass of zooplankton prey. Fish body mass and gender were included as covariates (italicised). Statistically significant ($\alpha = 0.05$) effects are shown in bold. χ^2 = chi-squared statistic; df = degrees of freedom; P = P-value associated with χ^2.

Parameter	**Factor; Interaction**	χ^2	**df**	***P***
MP mass per fish	Species	**1226.905**	**4**	**<0.001**
	MP size	**271.375**	**1**	**<0.001**
	Exposure	3.122	1	0.066
	Fish body mass	*3.263*	*1*	*0.066*
	Gender	*1.047*	*1*	*0.355*
	Species × MP size	**132.384**	**4**	**<0.001**
	Species × Exposure	**22.282**	**3**	**<0.001**
	MP size × Exposure	**57.376**	**1**	**<0.001**
	Species × MP size × Exposure	**13.039**	**3**	**0.004**
MP mass ingested per zooplankton individual	Species	**457.310**	**3**	**<0.001**
	MP size	**349.330**	**1**	**<0.001**
	Fish body mass	*0.727*	*1*	*0.349*
	Gender	*0.019*	*1*	*0.839*
	Species × MP size	**27.089**	**3**	**<0.001**
MP mass ingested per unit body mass of zooplankton prey	Species	**291.128**	**3**	**<0.001**
	MP size	**377.220**	**1**	**<0.001**
	Fish body mass	*1.111*	*1*	*0.226*
	Gender	*0.278*	*1*	*0.552*
	Species × MP size	**25.348**	**3**	**<0.001**

Supplementary Table 8 | Pairwise contrasts showing how fish MP uptake varies with zooplankton prey, particle size and exposure route. Estimated marginal-mean contrasts from generalized linear models (gamma distribution) comparing: (i) MP mass in the fish digestive tract (digestive tract), (ii) MP mass ingested per zooplankton individual, and (iii) MP mass ingested per unit zooplankton body mass across treatment combinations. Treatments vary by zooplankton species (*Simocephalus vetulus*, *Daphnia galeata*, *Tigriopus californicus*, *Acartia tonsa*), MP size (1 µm, 25 µm) and exposure route (Tt, trophic transfer; DI$_{zoo}$, direct ingestion in the presence of zooplankton; DI$_{NO}$, direct ingestion in the absence of zooplankton). Statistically significant (α = 0.05) differences are in bold. E, contrast estimate; SE, standard error; *z*, *z*-statistic; *P*, two-sided *P*-value (Holm-adjusted, where applicable).

Parameter	Contrast	*E*	SE	*z*	*P*
MP mass per fish	DI$_{NO}$ × 1µm – *S. vetulus* × 1µm × Tt	**-6.444**	**0.289**	**-22.333**	**<0.001**
	DI$_{NO}$ × 1µm – *D. galeata* × 1µm × Tt	**-6.691**	**0.299**	**-22.409**	**<0.001**
	DI$_{NO}$ × 1µm – *T. californicus* × 1µm × Tt	**-3.254**	**0.314**	**-10.369**	**<0.001**
	DI$_{NO}$ × 1µm – *A. tonsa* × 1µm × Tt	**-3.021**	**0.300**	**-10.079**	**<0.001**
	DI$_{NO}$ × 1µm – *S. vetulus* × 1µm × DI$_{zoo}$	**-6.411**	**0.288**	**-22.278**	**<0.001**
	DI$_{NO}$ × 1µm – *D. galeata* × 1µm × DI$_{zoo}$	**-5.509**	**0.297**	**-18.537**	**<0.001**
	DI$_{NO}$ × 1µm – *T. californicus* × 1µm × DI$_{zoo}$	**-1.790**	**0.315**	**-5.689**	**<0.001**
	DI$_{NO}$ × 1µm – *A. tonsa* × 1µm × DI$_{zoo}$	**-3.166**	**0.300**	**-10.562**	**<0.001**
	S. vetulus × 1µm × Tt – *D. galeata* × 1µm × Tt	-0.247	0.269	-0.919	1.000
	S. vetulus × 1µm × Tt – *T. californicus* × 1µm × Tt	**3.190**	**0.296**	**10.779**	**<0.001**
	S. vetulus × 1µm × Tt – *A. tonsa* × 1µm × Tt	**3.423**	**0.280**	**12.210**	**<0.001**
	D. galeata × 1µm × Tt – *T. californicus* × 1µm × Tt	**3.437**	**0.307**	**11.214**	**<0.001**
	D. galeata × 1µm × Tt – *A. tonsa* × 1µm × Tt	**3.670**	**0.291**	**12.613**	**<0.001**
	T. californicus × 1µm × Tt – *A. tonsa* × 1µm × Tt	0.233	0.306	0.762	1.000
	S. vetulus × 1µm × DI$_{zoo}$ – *D. galeata* × 1µm × DI$_{zoo}$	0.903	0.269	3.358	0.120
	S. vetulus × 1µm × DI$_{zoo}$ – *T. californicus* × 1µm × DI$_{zoo}$	**4.621**	**0.297**	**15.548**	**<0.001**
	S. vetulus × 1µm × DI$_{zoo}$ – *A. tonsa* × 1µm × DI$_{zoo}$	**3.245**	**0.280**	**11.580**	**<0.001**
	D. galeata × 1µm × DI$_{zoo}$ – *T. californicus* × 1µm × DI$_{zoo}$	**3.719**	**0.307**	**12.131**	**<0.001**
	D. galeata × 1µm × DI$_{zoo}$ – *A. tonsa* × 1µm × DI$_{zoo}$	**2.343**	**0.290**	**8.067**	**<0.001**
	T. californicus × 1µm × DI$_{zoo}$ – *A. tonsa* × 1µm × DI$_{zoo}$	**-1.376**	**0.307**	**-4.477**	**<0.001**
	S. vetulus × 1µm × Tt – *S. vetulus* × 1µm × DI$_{zoo}$	0.033	0.261	0.125	1.000
	D. galeata × 1µm × Tt – *D. galeata* × 1µm × DI$_{zoo}$	**1.182**	**0.275**	**4.293**	**<0.001**
	T. californicus × 1µm × Tt – *T. californicus* × 1µm × DI$_{zoo}$	**1.464**	**0.319**	**4.588**	**<0.001**
	A. tonsa × 1µm × Tt – *A. tonsa* × 1µm × DI$_{zoo}$	-0.145	0.292	-0.497	1.000
	DI$_{NO}$ × 25µm – *S. vetulus* × 25µm × Tt	**-3.641**	**0.292**	**-12.488**	**<0.001**
	DI$_{NO}$ × 25µm – *D. galeata* × 25µm × Tt	**-1.902**	**0.293**	**-6.485**	**<0.001**
	DI$_{NO}$ × 25µm – *T. californicus* × 25µm × Tt	0.351	0.314	1.119	1.000
	DI$_{NO}$ × 25µm – *A. tonsa* × 25µm × Tt	0.535	0.301	1.778	1.000
	DI$_{NO}$ × 25µm – *S. vetulus* × 25µm × DI$_{zoo}$	**-4.242**	**0.286**	**-14.826**	**<0.001**
	DI$_{NO}$ × 25µm – *D. galeata* × 25µm × DI$_{zoo}$	**-3.272**	**0.295**	**-11.084**	**<0.001**
	DI$_{NO}$ × 25µm – *T. californicus* × 25µm × DI$_{zoo}$	0.124	0.313	0.395	1.000
	DI$_{NO}$ × 25µm – *A. tonsa* × 25µm × DI$_{zoo}$	-0.938	0.300	-3.125	0.272
	S. vetulus × 25µm × Tt – *D. galeata* × 25µm × Tt	**1.740**	**0.270**	**6.434**	**<0.001**
	S. vetulus × 25µm × Tt – *T. californicus* × 25µm × Tt	**3.992**	**0.294**	**13.561**	**<0.001**
	S. vetulus × 25µm × Tt – *A. tonsa* × 25µm × Tt	**4.176**	**0.285**	**14.665**	**<0.001**
	D. galeata × 25µm × Tt – *T. californicus* × 25µm × Tt	**2.253**	**0.298**	**7.562**	**<0.001**
	D. galeata × 25µm × Tt – *A. tonsa* × 25µm × Tt	**2.436**	**0.287**	**8.483**	**<0.001**
	T. californicus × 25µm × Tt – *A. tonsa* × 25µm × Tt	0.184	0.308	0.596	1.000
	S. vetulus × 25µm × DI$_{zoo}$ – *D. galeata* × 25µm × DI$_{zoo}$	0.969	0.270	3.590	0.051
	S. vetulus × 25µm × DI$_{zoo}$ – *T. californicus* × 25µm × DI$_{zoo}$	**4.365**	**0.293**	**14.880**	**<0.001**
	S. vetulus × 25µm × DI$_{zoo}$ – *A. tonsa* × 25µm × DI$_{zoo}$	**3.303**	**0.278**	**11.863**	**<0.001**
	D. galeata × 25µm × DI$_{zoo}$ – *T. californicus* × 25µm × DI$_{zoo}$	**3.396**	**0.303**	**11.194**	**<0.001**
	D. galeata × 25µm × DI$_{zoo}$ – *A. tonsa* × 25µm × DI$_{zoo}$	**2.334**	**0.290**	**8.048**	**<0.001**
	T. californicus × 25µm × DI$_{zoo}$ – *A. tonsa* × 25µm × DI$_{zoo}$	-1.062	0.306	-3.472	0.079

	S. vetulus × 25µm × Tt – *S. vetulus* × 25µm × DI_{zoo}	0.600	0.265	2.269	1.000
	D. galeata × 25µm × Tt – *D. galeata* × 25µm × DI_{zoo}	**1.371**	**0.276**	**4.972**	**<0.001**
	T. californicus × 25µm × Tt – *T. californicus* × 25µm × DI_{zoo}	0.227	0.320	0.709	1.000
	A. tonsa × 25µm × Tt – *A. tonsa* × 25µm × DI_{zoo}	**1.473**	**0.292**	**5.049**	**<0.001**
	DI_{NO} × 1µm – DI_{NO} × 25µm	-1.050	0.308	-3.412	0.098
	S. vetulus × 1µm × Tt – *S. vetulus* × 25µm × Tt	**1.753**	**0.261**	**6.703**	**<0.001**
	D. galeata × 1µm × Tt – *D. galeata* × 25µm × Tt	**3.739**	**0.278**	**13.462**	**<0.001**
	T. californicus × 1µm × Tt – *T. californicus* × 25µm × Tt	**2.555**	**0.320**	**7.986**	**<0.001**
	A. tonsa × 1µm × Tt – *A. tonsa* × 25µm × Tt	**2.506**	**0.292**	**8.569**	**<0.001**
	S. vetulus × 1µm × DI_{zoo} – *S. vetulus* × 25µm × DI_{zoo}	**1.120**	**0.262**	**4.276**	**<0.001**
	D. galeata × 1µm × DI_{zoo} – *D. galeata* × 25µm × DI_{zoo}	**1.186**	**0.276**	**4.301**	**<0.001**
	T. californicus × 1µm × DI_{zoo} – *T. californicus* × 25µm × DI_{zoo}	0.864	0.319	2.705	1.000
	A. tonsa × 1µm × DI_{zoo} – *A. tonsa* × 25µm × DI_{zoo}	**1.177**	**0.292**	**4.038**	**0.008**
MP mass ingested per zooplankton individual	*S. vetulus* × 1µm - *D. galeata* × 1µm	-0.213	0.269	-0.791	1.000
	S. vetulus × 1µm - *T. californicus* × 1µm	**3.290**	**0.324**	**10.152**	**<0.001**
	S. vetulus × 1µm - *A. tonsa* × 1µm	**3.510**	**0.266**	**13.201**	**<0.001**
	D. galeata × 1µm - *T. californicus* × 1µm	**3.503**	**0.341**	**10.260**	**<0.001**
	D. galeata × 1µm - *A. tonsa* × 1µm	**3.723**	**0.285**	**13.049**	**<0.001**
	T. californicus × 1µm - *A. tonsa* × 1µm	-0.220	0.312	0.705	1.000
	S. vetulus × 25µm - *D. galeata* × 25µm	**1.795**	**0.273**	**6.585**	**<0.001**
	S. vetulus × 25µm - *T. californicus* × 25µm	**4.140**	**0.321**	**12.899**	**<0.001**
	S. vetulus × 25µm - *A. tonsa* × 25µm	**4.252**	**0.278**	**15.283**	**<0.001**
	D. galeata - × 25µm *T. californicus* × 25µm	**2.346**	**0.321**	**7.311**	**<0.001**
	D. galeata × 25µm - *A. tonsa* × 25µm	**2.457**	**0.272**	**9.018**	**<0.001**
	T. californicus × 25µm - *A. tonsa* × 25µm	0.111	0.316	0.353	1.000
	S. vetulus × 1µm - *S. vetulus* × 25µm	**1.741**	**0.260**	**6.687**	**<0.001**
	D. galeata × 1µm - *D. galeata* × 25µm	**3.749**	**0.282**	**13.304**	**<0.001**
	T. californicus × 1µm - *T. californicus* × 25µm	**2.591**	**0.362**	**7.150**	**<0.001**
	A. tonsa × 1µm - *A. tonsa* × 25µm	**2.483**	**0.256**	**9.698**	**<0.001**
MP mass ingested per unit body mass of zooplankton prey	*S. vetulus* × 1µm - *D. galeata* × 1µm	-0.491	0.270	-1.818	0.276
	S. vetulus × 1µm - *T. californicus* × 1µm	**2.756**	**0.343**	**8.028**	**<0.001**
	S. vetulus × 1µm - *A. tonsa* × 1µm	**2.453**	**0.268**	**9.154**	**<0.001**
	D. galeata × 1µm - *T. californicus* × 1µm	**3.246**	**0.341**	**9.512**	**<0.001**
	D. galeata × 1µm - *A. tonsa* × 1µm	**2.943**	**0.264**	**11.136**	**<0.001**
	T. californicus × 1µm - *A. tonsa* × 1µm	-0.303	0.316	-0.958	0.495
	S. vetulus × 25µm - *D. galeata* × 25µm	**1.439**	**0.273**	**5.268**	**<0.001**
	S. vetulus × 25µm - *T. californicus* × 25µm	**3.676**	**0.341**	**10.786**	**<0.001**
	S. vetulus × 25µm - *A. tonsa* × 25µm	**3.231**	**0.279**	**11.576**	**<0.001**
	D. galeata - × 25µm *T. californicus* × 25µm	**2.237**	**0.322**	**6.951**	**<0.001**
	D. galeata × 25µm - *A. tonsa* × 25µm	**1.792**	**0.250**	**7.164**	**<0.001**
	T. californicus × 25µm - *A. tonsa* × 25µm	-0.445	0.320	-1.389	0.495
	S. vetulus × 1µm - *S. vetulus* × 25µm	**1.706**	**0.285**	**5.998**	**<0.001**
	D. galeata × 1µm - *D. galeata* × 25µm	**3.636**	**0.258**	**14.074**	**<0.001**
	T. californicus × 1µm - *T. californicus* × 25µm	**2.627**	**0.382**	**6.873**	**<0.001**
	A. tonsa × 1µm - *A. tonsa* × 25µm	**2.485**	**0.237**	**10.505**	**<0.001**

Supplementary Table 9 | Drivers of zooplankton foraging selectivity for microplastics across freshwater and marine environments. Multivariate random-effects meta-analysis (REML) **testing** how the relationship between microplastic ingestion and algal ingestion by zooplankton differs between freshwater and marine environments. Statistically significant effects ($\alpha = 0.05$) are shown in bold. *E*, estimate; SE, standard error; *Z*, *z*-statistic; *P*, *P*-value; 95% CI, 95% confidence interval.

Effect	**E**	**SE**	**Z**	***P***	**95% CI**	
					lower	**upper**
Intercept (Freshwater)	**1.049**	**0.063**	**16.65**	**<0.001**	**0.926**	**1.173**
Environment (Marine)	**–0.985**	**0.062**	**–15.80**	**<0.001**	**−1.107**	**−0.863**

Residual heterogeneity: $\tau^2 = 0.0013$; QE(3) = 4.86, $P = 0.182$. Test of moderators: QM(1) = 249.6, $P < 0.0001$.

Supplementary Table 10 | Freshwater–marine contrasts in fish MP burden within environmental deciles. Kruskal–Wallis χ^2 with Monte Carlo *P*-values (1,000 resamples) computed within deciles of mean-centred log10 environmental MP concentration. Reported χ^2 values are the sums across deciles (df = the number of deciles containing observations from both environments). Individual per-decile test results and Hodges–Lehmann between-environment differences (with CIs) are available in the repository. Deciles lacking records from both environments were omitted. Statistically significant (α = 0.05) effects are shown in bold.

Compartment	HL diff (F–M, log10)	95% CI (HL)	KWχ^2	df	*P* (MC)	Direction
Total	1.609	-1.986, 4.709	71.082	10	**<0.001**	Freshwater > Marine
Digestive tract	1.625	-5.686, 6.969	329.241	17	**<0.001**	Freshwater > Marine
Muscle	0.596	-5.869, 3.120	55.934	11	**<0.001**	Freshwater > Marine
Gills	0.876	-2.975, 4.259	90.727	12	**<0.001**	Freshwater > Marine

Note: Full per-decile medians and HL CIs are provided in the data repository; summary χ^2 and directions are reported here to prioritise clarity.

Supplementary Table 11 | Log–log regressions of fish microplastic burden on environmental concentration, by compartment and environment, with between-environment contrasts. For each compartment (total, digestive tract, gills, muscle) and environment (freshwater, marine), we report slope and intercept with 95% confidence intervals, model F and R^2 from simple regressions of log10 fish MP burden on log10 environmental MP concentration. Between-environment comparisons include slope contrasts (t-tests; test of parallelism) and ANCOVA intercept contrasts (elevation). Total and digestive tract **(Fig. 2A)** exhibited parallel slopes with higher freshwater elevation, whereas muscle and gills were non-parallel. All variables are log10-scaled. Statistically significant (α = 0.05) effects are shown in bold.

Compartment	**Slope difference (FWS vs MAR)**	***P***	**Intercept difference (ANCOVA)**	***P***	**Overall model *F***	***P***
Total	$t = -0.936$	0.350	$F = 99.145$	**<0.001**	$F = 166.270$	**<0.001**
Digestive tract	$t = -0.130$	0.897	$F = 22.873$	**<0.001**	$F = 17.673$	**<0.001**
Muscle	non-parallel (slopes differ)	—	—	—	—	—
Gills	non-parallel (slopes differ)	—	—	—	—	—

Notes: Exact slopes, intercepts and 95% CIs for muscle and gills are not reported here because the fitted regressions were non-parallel (“lines crossed”); full coefficients are provided in the data-repository export.

Supplementary Table 12 | Environment-independent freshwater–marine contrasts in fish microplastic burden by compartment. Box-plot summaries and Monte Carlo Kruskal–Wallis tests (1,000 resamples) corresponding to **Fig. 2B**. Freshwater burdens exceeded marine in total, digestive tract, and muscle (df = 1, $P < 0.05$), whereas gills showed no significant difference (df = 1, $P > 0.05$).

Compartment	**Direction**	**Significance**
Total	Freshwater > Marine	$\boldsymbol{P} < \mathbf{0.05}$ (Monte Carlo)
Digestive tract	Freshwater > Marine	$\boldsymbol{P} < \mathbf{0.05}$ (Monte Carlo)
Muscle	Freshwater > Marine	$\boldsymbol{P} < \mathbf{0.05}$ (Monte Carlo)
Gills	— (weakest contrast)	n.s. ($P > 0.05$, Monte Carlo)

Supplementary Table 13 | Particle-based bioaccumulation factors (pBAF): effects of environment and compartment. **(A)** Two-way permutational ANOVA on ln(pBAF) with factors Environment (freshwater vs marine) and Body part (total, digestive tract, gills, muscle); Type-III SS, df, MS, *F*, *P*. Permutation scheme: Freedman–Lane, 10,000 permutations. **(B)** Estimated marginal means (± SE, 95% CI) of ln(pBAF) by environment. **(C)** Compartment-wise freshwater–marine contrasts (Kruskal–Wallis χ^2 with 1,000-iteration Monte Carlo *P*-values) for total, digestive tract, gills and muscle. **(D)** *Post hoc* within-compartment comparisons; all pairwise n.s. except total vs muscle ($P = 0.003$). ANOVA uses the natural-log scale; contrast panels report the corresponding per-compartment tests.

(A) Two-way permutational ANOVA (ln pBAF)

Source	Type III SS	df	MS	*F*	*P*
Environment (FWS vs MAR)	1438.446	1	1438.446	81.228	**<0.001**
Body part (Total, digestive tract, Gills, Muscle)	104.965	3	34.988	1.976	0.116
Env × Part	166.702	3	55.567	3.138	**0.025**

(B) Estimated marginal means (ln pBAF)

Environment	Mean	SE	95% CI (Lower, Upper)
Freshwater	3.873	0.330	3.226, 4.520
Marine	7.325	0.195	6.942, 7.708

(C) Compartment-wise FWS–MAR contrasts (KW, Monte Carlo)

Compartment	χ^2	df	*P* (Monte Carlo)
Digestive tract	31.731	1	**<0.001**
Gills	23.855	1	**<0.001**
Total	4.771	1	**0.029**
Muscle	5.023	1	**0.025**

(D) *Post hoc* within-compartment comparisons

All pairwise comparisons n.s. except total vs muscle ($P = 0.003$).
Homogeneous subsets (Body part): {total} vs {digestive tract, gills, muscle}.

Supplementary Table 14 | Trophic-level (TL 2–5) regressions of fish microplastic (MP) burden by environment and compartment. **(A)** Marine. **(B)** Freshwater. **(C)** Pooled (marine + freshwater). For each compartment (digestive tract, gills, muscle, total) we report separate simple linear models of log10 MP burden vs trophic level for marine and freshwater, and a pooled model (marine + freshwater). Reported statistics: *R*, R^2, adjusted R^2, SE of estimate, slope *B* on the log10 scale ± SE with 95% CI, *t*, two-sided *P*, and model *F* (df1, df2). In marine fish, digestive tract, gills and total decline with TL, while muscle increases. In freshwater fish, all compartments decline with TL. In the pooled dataset, digestive tract, gills and total decline, muscle is non-significant. Note: negative *B* indicates decreasing MP burden with increasing TL on the log10 scale. Statistically significant ($\alpha = 0.05$) effects are shown in bold.

(A) Marine

Compartment	*R*	R^2	Adj R^2	SE	Slope *B* (log10)	SE(*B*)	*t*	*P*	Model *F*	df1, df2	*P*
Digestive tract	0.078	0.006	0.005	0.5807	−0.078	0.038	−2.050	**0.041**	4.204	1, 691	**0.041**
Gills	0.075	0.006	0.004	0.5809	−0.061	0.031	−1.982	**0.048**	3.927	1, 691	**0.048**
Muscle	0.098	0.010	0.008	0.5797	0.153	0.059	+2.601	**0.010**	6.763	1, 691	**0.010**
Total	0.078	0.006	0.005	0.5807	−0.150	0.073	−2.044	**0.041**	4.179	1, 691	**0.041**

(B) Freshwater

Compartment	*R*	R^2	Adj R^2	SE	Slope *B* (log10)	SE(*B*)	*t*	*P*	Model *F*	df1, df2	*P*
Digestive tract	0.175	0.031	0.025	0.650	−0.233	0.096	−2.434	**0.016**	5.923	1, 188	**0.016**
Gills	0.172	0.030	0.025	0.651	−0.106	0.044	−2.400	**0.017**	5.758	1, 188	**0.017**
Muscle	0.168	0.028	0.023	0.651	−0.047	0.020	−2.335	**0.021**	5.453	1, 188	**0.021**
Total	0.174	0.030	0.025	0.650	−0.418	0.172	−2.429	**0.016**	5.900	1, 188	**0.016**

(C) Pooled (marine + freshwater)

Compartment	*R*	R^2	Adj R^2	SE	Slope *B* (log10)	SE(*B*)	*t*	*P*	Model *F*	df1, df2	*P*
Digestive tract	0.343	0.118	0.117	0.601	−0.222	0.021	−10.83	**<0.001**	117.340	1, 881	**<0.001**
Gills	0.304	0.092	0.091	0.610	−0.184	0.019	−9.47	**<0.001**	89.721	1, 881	**<0.001**
Muscle	0.033	0.001	0.000	0.640	+0.018	0.019	+0.99	0.321	0.985	1, 881	0.321
Total	0.365	0.133	0.132	0.596	−0.245	0.021	−11.63	**<0.001**	135.305	1, 881	**<0.001**

Supplementary Table 15 | Feeding-guild contrasts (freshwater vs marine) in digestive tract MP burdens, with cross-compartment checks. **(A)** Digestive tract (primary panel in **Fig. 2D**): within-decile freshwater–marine contrasts by guild (herbivores, planktivores, carnivores) using Kruskal–Wallis χ^2 with 1,000-iteration Monte Carlo *P*-values; n shown per guild; interpretation indicated. Deciles are defined on mean-centred log10 environmental MP concentration; deciles lacking both environments are omitted. **(B)** Gills: complementary freshwater–marine checks by guild (all significant). **(C)** Muscle and total: complementary checks by guild (muscle generally non-significant; total significant only in carnivores). *Note:* exact n by guild for gills/muscle/total were not listed in the notes; "—" indicates counts available in the raw dataset. Statistically significant (α = 0.05) effects are shown in bold.

(A) Digestive tract (primary panel in Fig. 2D)

Guild	n	χ^2	df	*P* (Monte Carlo)	Interpretation
Carnivores	473	6.061	1	**0.014**	Freshwater > Marine
Herbivores	59	2.011	1	0.157	n.s.
Planktivores	129	0.817	1	0.366	n.s.

(B) Gills (complementary checks)

Guild	n	χ^2	df	*P* (Monte Carlo)	Interpretation
Carnivores	—	61.805	1	**<0.001**	Freshwater–marine contrast present
Herbivores	—	13.224	1	**<0.001**	Freshwater–marine contrast present
Planktivores	—	31.359	1	**<0.001**	Freshwater–marine contrast present

(C) Muscle & Total (complementary checks)

Guild	Compartment	χ^2	df	*P* (Monte Carlo)
Carnivores	Muscle	0.196	1	0.279
Carnivores	Total	11.278	1	**<0.001**
Herbivores	Muscle	1.102	1	0.294
Herbivores	Total	3.100	1	0.078
Planktivores	Muscle	0.535	1	0.453
Planktivores	Total	2.571	1	0.109

Notes: Exact n by guild for gills/muscle/total were not listed in the notes; "—" indicates count available in the raw dataset.

Supplementary Table 16 | Composite index of whole-body burden (FC1): two-factor bootstrap ANOVA. Bootstrap ANOVA (n = 1,000 resamples) with factors Environment (freshwater vs marine) and Environmental MP decile (10 levels) on FC1, the first component from dimensionality reduction across compartmental MP measures (≈55% variance explained). Reported: SS, df, MS, *F*, *p*, partial ç^{2}. FC1 increases with environmental decile and is higher in freshwater; environmental concentration explains ~56.3% of FC1 variance. Statistically significant (α = 0.05) effects are shown in bold.

Effect	**SS**	**df**	**MS**	***F***	***P***	**Partial ç^{2}**
Model	25.013	8	3.127	7.564	**0.001**	0.676
Environment (FWS vs MAR)	5.921	1	5.921	14.324	**<0.001**	0.331
Environmental MP Decile	15.453	7	2.208	5.341	**0.001**	0.563

Notes: FC1 derives from dimensionality reduction across compartmental MP measures and explains ~55% of total variance. Environmental MP concentration accounts for ~56.3% of FC1 variance across deciles.