Supporting information for

Hydrophobic organic contaminants are not linked to microplastic uptake in Baltic Sea herring

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1.1 Data handling of FTIR spectra

The original FTIR spectra contained residual signals from atmospheric CO_2. These were subtracted using five spectra recorded without sample in which the regions without CO_2 signals were replaced by straight lines. Data outside the range of 3845 to 400 cm^{-1} were ignored. In order to treat the data in the same way as those in the reference data base (Primpke et al. 2018) the spectra were baseline corrected (64 baseline points, 10 iterations) with the OPUS 5.5 software. Their wavenumber values were then made compatible with those in the reference data base using one of its spectra as the principal file.

To reduce noise and enhance the spectrum quality without losing subtle spectral information, each spectrum passed through a baseline correction and denoising procedure using a second order derivative Savitzky-Golay filter to minimize baseline effects (Renner et al. 2019) as implemented in Unscrambler® X (version 10.5.1; Camo Software, 2018). Pre-processed spectra were then compared to the spectral libraries assembled by Primpke and co-workers (Primpke et al. 2018) using BioRad KnowItAll® Informatics System software. The Correlation algorithm implemented in the software was used to evaluate each query spectrum to the spectra of the databases. The most appropriate match (Hit Quality Index; HQI) was selected based on matching peak wavenumber positions and a minimum 70% correlation between unknown and matched spectra. In all spectra, we focused on the 3799 to 850 wavenumber (cm^{-1}) region, excluding the interval 2690 to 1900 wavenumber (cm^{-1}).

2.1 Model parameterization

2.1.1 Clearance rate (CR)

Clearance rates (CR) for Baltic herring were calculated based on intake rates of *Calanus finmarchicus* in the North Sea (Fig. 5 in Varpe and Fiksen 2010), which is the main prey for herring in this area. These copepods are also of similar size (2-3 mm prosome length) as the microplastic particles considered in this study (Pasternak et al. 2004). We used the reported values on *C. finmarchicus* consumption by herring expressed as (J copepod [J herring]$^{-1}$ day^{-1}) and ambient *C. finmarchicus* abundance to obtain the CR.

The average CR of the Baltic herring population examined in this study (L h^{-1}) was calculated assuming an energy content of 3.5 kJ and 10 kJ g wet weight^{-1} for *C. finmarchicus* and herring, respectively (Varpe et al. 2005). The average weight of the sampled Baltic herring (35 g) was used to derive the consumption rate on an individual basis using a first-order exponential decay function fitted to data on the CR and prey abundance for the North Sea herring feeding on *C. finmarchicus* (Figure S 2). The asymptote value (1.04×10^3 L ind.$^{-1}$ h^{-1}) was assumed to represent CR of the Baltic herring, because mesozooplankton abundance in the Baltic Sea normally supersede the maximum reported abundance for *C. finmarchicus* in the North Sea (Varpe and Fiksen 2010, Gorokhova et al. 2016).

2.1.2 Ambient MP concentrations in the Baltic Sea (CMP)

We used the average microplastic concentrations reported by Gewert et al. (2017) in the outer Stockholm archipelago (0.58 MP m^{-3}) estimated by surface manta trawls (335 µm mesh). These values were used, because the size range (median MP size and inter quartile range, IQR: fragment diameter = 1 mm (IQR 0.6-1.5 mm), fiber length = 1 mm (IQR 1-3 mm)) fits well the size of MP recovered from the fish guts. Also, the polymer materials have been rigorously identified by FT-IR in this selection of the field-collected MP, thus ensuring that the fragments collected were indeed microplastics.

2.1.3. Gut evacuation rates (GER)

We were not able to find data on gut evacuation rates for adult herring; therefore, a lower and an upper limit reported for two clupeid species of similar size and feeding ecology as the herring analyzed here (Collard et al. 2015) were chosen. The lower limit (0.05 h^{-1}) was adopted from the experimental and field data collected for adult South American pilchard (*Sardinops sagax*) (van der Lingen 1998). The upper limit (0.26 h^{-1}) was experimentally derived for adult European pilchard (*Sardina pilchardus*) (Costalago and Palomera 2014).

2.2 Monte Carlo simulation of MP burden in the Baltic herring

To estimate MP burden (MP ind^{-1}) dynamics at a given MP abundance from time 0 to the point when it is stabilized (48 h), we performed Monte Carlo simulation with 1000 permutations using STELLA® ver. 9.4.1 software (iSee systems, Inc. Lebanon, NH, U.S.A.), with the equations (Eqs. 1 to 3) integrated as shown in Figure S 3**.** Ambient MP concentrations (CMP) were allowed to vary randomly following a Poisson distribution as were the data presented in Gewert et al. (2017), whereas the CR values were normally distributed with a mean and SD of 1041 L h^{-1} and 27 h^{-1}, respectively, and GER values varied randomly between 0.05 and 0.26 h^{-1} without any assumption regarding the distribution (Table S 3). The final value of each run was used to represent an individual in the population.

SI Tables and Figures

Table S 1. FTIR classification results based on the comparison with a reference database (Primpke et al. 2018). A match to one or more equally plausible compounds was accepted at HQI ≥ 70%. Spectra with HQI-scores below this threshold were classified as unknown.

Sample	Sample type	Classification	Group
1	transparent fiber	fur	natural
2	transparent fiber	polyvinyl	MP
3	blue fiber	plant fiber, cellulose, hydroxypropyl methyl cellulose, polyvinyl alcohol	MP
4	transparent fiber	algae	natural
5	transparent fiber	fur	natural
6	transparent fiber	algae	natural
7	red fiber	fur	natural
8	black fiber	plant fiber	natural
9	black fiber	plant fiber, cellulose	natural
10	blue fiber	plant fiber, viscose	MP
11	blue fiber	viscose, polyvinyl	MP
12	transparent fiber	chitin	natural
13	blue fiber	unknown	unknown
14	blue fiber	unknown	unknown
15	blue fiber	plant fiber, cellulose, hydroxypropyl methyl cellulose, polyvinyl alcohol	MP
16	red fiber	plant fiber, cellulose, hydroxypropyl methyl cellulose, rubber	MP
17	black fiber	unknown	unknown
18	white fragment	acrylonitrile butadiene styrene, alkyd varnish	MP
19	blue fiber	unknown	unknown
20	black fiber	unknown	unknown
21	red fiber	rubber, ethylene propylene, poly 1-butene isotactic	MP
22	brown fragment	polyamide, fur	natural
23	black fiber	rubber, poly 1-butene isotactic	MP
24	black fiber	rubber, polyethylene	MP
25	blue fiber	fur	natural
26	red fiber	unknown	unknown

Table S 2. Descriptive statistics for the microplastics recovered from the gastrointestinal tract of Baltic Sea herring as well as gut fullness of the examined fish. For each basin, the number of differently colored fragments and fibers that were recovered from the fish are shown as well as MP frequency of occurrence (FO), median, range (min-max) and mean; moreover, the mean values were calculated for all fish and for the fish that contained putative MP in their GIT (i.e., excluding zero values). Gut fullness (GF) is presented as percent of fish with empty stomachs, median gut fullness and the corresponding inter-quartile range (IQR). Values in parentheses represent FTIR-validated MP data. Data are ordered north to south.

	Fibers				Fragments			MP total					GF		
	Black/Brown	Red	Blue	Clear	Black/Brown	Red	Green	FO (%)	Median	Mean all samples	Mean of presences	Range (min-max)	% fish with empty GIT	Median	IQR
Bothnian Bay	0-7	0-38	0-0	0-3	0-0	0-3	0-12	46.7 (40.0)	0	4.1 (3.5)	8.9 (3.9)	0-40 (0-15)	10	50	25-50
Bothnian Sea	0-8	0-18	0-0	0-5	0-7	0-0	0-2	30.0 (12.5)	0	1.3 (1.0)	4.2 (3.4)	0-18 (0-7)	7.5	25	25-50
Northern Baltic Proper	0-1	0-0	0-0	0-51	0-0	0-2	0-2	30.0 (20.0)	0	6.7 (6.6)	22.2 (12.8)	0-51 (0-20)	25	25	19-50
Western Gotland Basin	0-1	0-3	0-1	0-1	0-0	0-7	0-0	40.0 (25.0)	0	1.0 (0.9)	2.5 (1.4)	0-7 (0-3)	10	25	25-25
Bornholm Basin	0-1	0-13	0-0	0-2	0-0	0-0	0-0	20.0 (15.0)	0	0.9 (0.9)	4.5 (2.3)	0-13 (0-5)	0	50	25-56
Total	**0-8**	**0-38**	**0-1**	**0-51**	**0-7**	**0-7**	**0-12**	**33.8 (22.3)**	**0**	**2.7 (1.0)**	**7.8 (4.4)**	**0-51 (0-20)**	**9.2**	**25**	**25-50**

Table S 3. Variables and simulation settings used to model microplastic ingestion in Baltic Sea herring. Details regarding derivation of the values are provided in the Supporting Information 2.1.

Parameter	Unit	Average	Min	Max	S.D	Distribution	Species	Meaning	Reference
CMP	MP L^{-1}	5.8×10^{-4}				Poisson		MP concentration in the water column	Gewert et al. 2017
CR	L $ind.^{-1}$ h^{-1}	1.04×10^{3}			2.6×10^{2}	Normal	*Clupea harengus*	Clearance rate	Varpe & Fiksen 2010
GER^{1}	h^{-1}		5×10^{-2}	2.6×10^{-1}			*Sardinops sagax*, *Sardina pilchardus*	Gut evacuation rate	Van der Lingen 1998, Costalago & Palomera 2014
IR	MP h^{-1}							Number of MP ingested at time *t*	
MP	MP							Number of MP in fish stomach at time *t*	
Eg	MP h^{-1}							Number of MP egested at time *t*	

1. The lower value for GER is based on data for *Sardinops sagax* (Van der Lingen 1998) while the higher is derived from *Sardina pilchardus* (Costalago & Palomera 2014).

Table S 4. Descriptive statistics for the predicted (modelled) and observed distributions of the MP burden in the Baltic herring. The data are presented as either "Total", i.e., where individuals without MP in the GIT are included, or "Zeros excluded" that shows only the fish with positive MP burden.

	Total		Zeros excluded	
	Mod	Obs	Mod	Obs
n	1000	130	806	25
Mean	4.7	0.9	5.9	4.7
SD	4.7	3.0	4.5	5.6
Median	3.6	0.0	4.4	2.0
Min	0.0	0.0	1.3	1.0
Max	33.3	20.0	33.3	20.0
Range	33.3	20.0	32.0	19.0
Skew	1.8	4.4	2.0	1.5
Kurtosis	5.0	20.1	5.7	0.8
SE	0.1	0.3	0.2	0.1

Table S 5. Summary statistics and factor loadings for the variables used in the factor analysis. WS MP burden = weight specific MP burden. Factor loadings > 0.7 are considered statistically significant.

	Factor 1	**Factor 2**
WS MP burden	-0.135	0.578
BDE sum	**0.921**	0.243
HBCD	**0.997**	-0.042
DD sum	**0.953**	0.083
HCB	**0.941**	-0.099
PCB sum	0.562	**0.824**
SS	3.969	1.091
Proportion var	0.662	0.182
Cumulative var	0.662	0.843

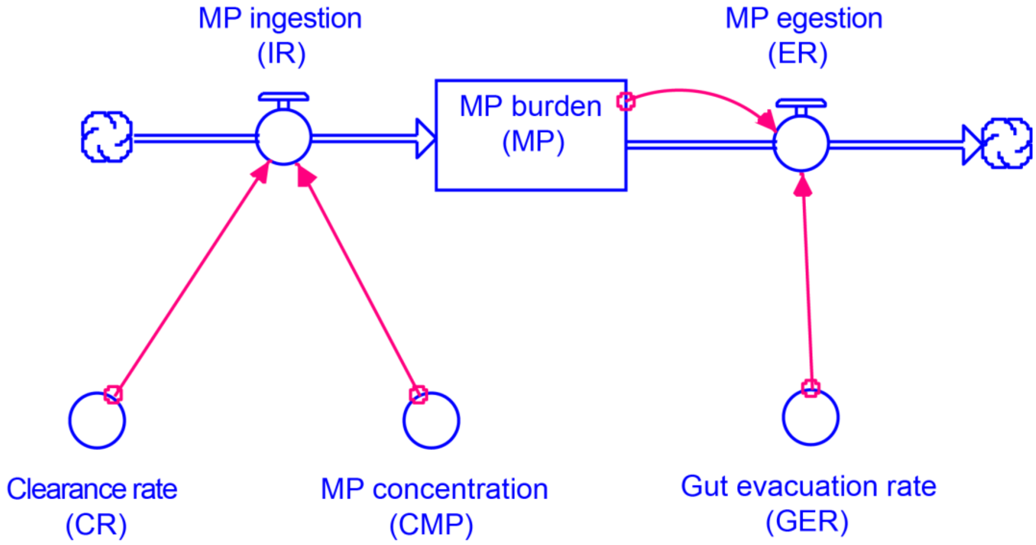

Figure S 1. Schematic representation of the model used to predict microplastic ingestion in Baltic herring.

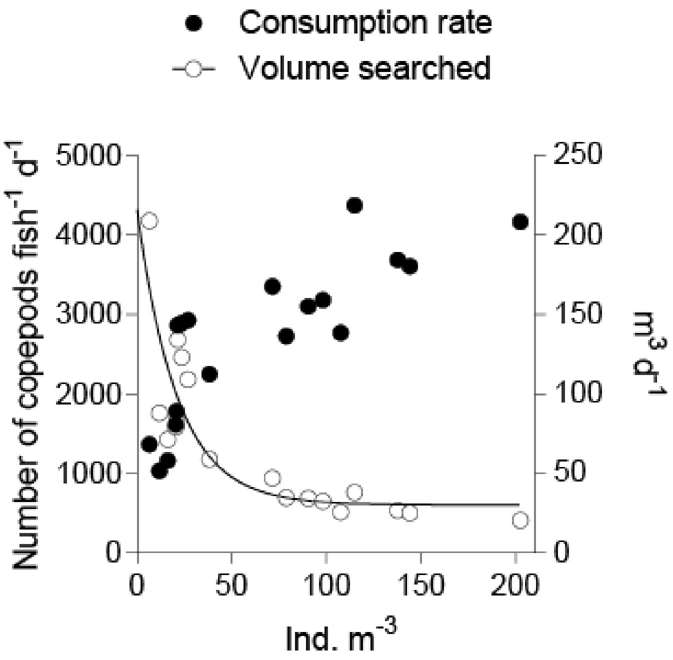

Figure S 2. Consumption rates (left axis) and clearance rates (right axis) as a function of *Calanus finmarchicus* abundance. The values are based on the data presented in Fig. 5, Varpe and Fiksen (2010) and adjusted for fish with average body weight of 35 g.

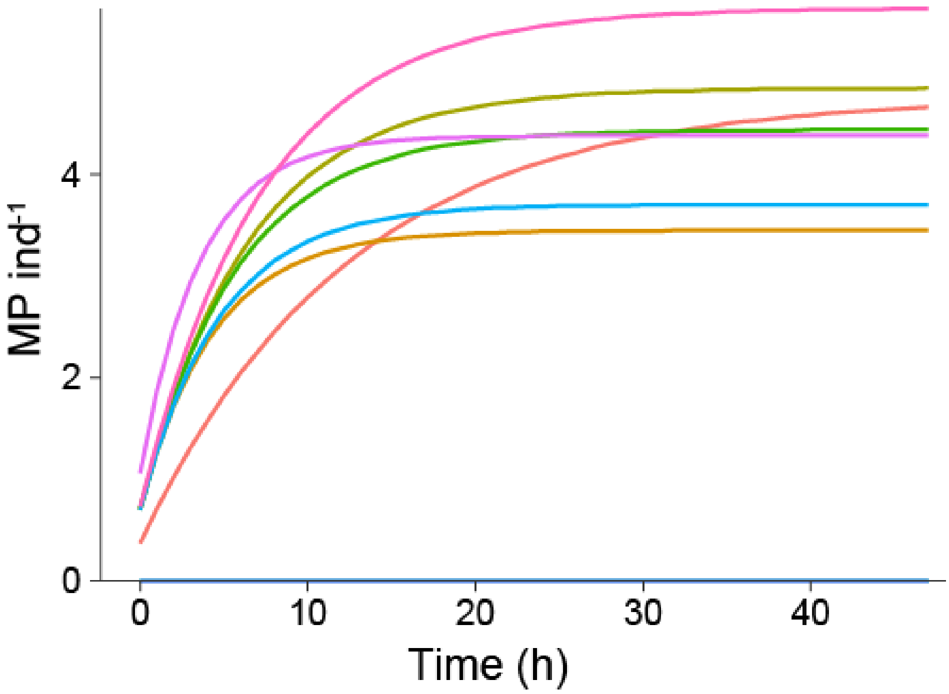

Figure S 3. Modeled MP burden (MP ind^{-1}) in the first ten simulation runs for 48 h. Observe that values are stabilized at the end of the simulation; these values are used to represent intrapopulation variability. Three out of ten individuals contain no MP.

References

Collard, F., B. Gilbert, G. Eppe, E. Parmentier, and K. Das. 2015. Detection of Anthropogenic Particles in Fish Stomachs: An Isolation Method Adapted to Identification by Raman Spectroscopy. Archives of Environmental Contamination and Toxicology 69:331–339.

Costalago, D., and I. Palomera. 2014. Feeding of European pilchard (Sardina pilchardus) in the northwestern Mediterranean: from late larvae to adults. Scientia Marina 78:41–54.

Gorokhova E, Lehtiniemi M, Postel L, Rubene G, Amid C, Lesutiene J, Uusitalo L, Strake S, Demereckiene 2016. Indicator properties of Baltic zooplankton for classification of environmental status within Marine Strategy Framework Directive. *PLoS ONE* 11(7): e0158326.doi:10.1371/journal.pone.0158326

van der Lingen, C. D. 1998. Gastric evacuation, feeding periodicity and daily ration of sardine *Sardinops sagax* in the southern Benguela upwelling ecosystem. South African Journal of Marine Science 19:305–316.

Ojaveer, H., A. Lankov, T. Raid, A. Põllumäe, and R. Klais. 2018. Selecting for three copepods—feeding of sprat and herring in the Baltic Sea. ICES Journal of Marine Science.

Pasternak, A., K. S. Tande, E. Arashkevich, and W. Melle. 2004. Reproductive patterns of Calanus finmarchicus at the Norwegian midshelf in 1997. Journal of Plankton Research 26:839–849.

Primpke, S., M. Wirth, C. Lorenz, and G. Gerdts. 2018. Reference database design for the automated analysis of microplastic samples based on Fourier transform infrared (FTIR) spectroscopy. Analytical and Bioanalytical Chemistry 410:5131–5141.

Renner, G., A. Nellessen, A. Schwiers, M. Wenzel, T. C. Schmidt, and J. Schram. 2019. Data preprocessing & evaluation used in the microplastics identification process: A critical review & practical guide. TrAC Trends in Analytical Chemistry 111:229–238.

Varpe, Ø., and Ø. Fiksen. 2010. Seasonal plankton–fish interactions: light regime, prey phenology, and herring foraging. Ecology 91:311–318.

Varpe, Ø., Ø. Fiksen, and A. Slotte. 2005. Meta-ecosystems and biological energy transport from ocean to coast: the ecological importance of herring migration. Oecologia 146:443.