

Supplementary Material

Table S1. Biometric features (length, width, height) and weight of *Anodonta cygnea* collected from aquaculture (T0) and exposed in three different rivers for short term and long term.

Exposure time	ID sample	Shell length (cm)	Shell width (cm)	Shell height (cm)	Wet soft tissue weight (g)	Wet gills weight (g)	Wet gastrointestinal tract weight (g)
T0	Ctr1	9.36	6.44	2.8	10.8	1.3	1.6
	Ctr2	8.12	5.73	2.5	8.5	1.4	1.5
	Ctr3	7.08	5.55	3	9.3	3.2	2.4
Short term 1 month	Ani1	9.37	6.53	3.3	22	5.7	8
	Ani2	9.9	7.33	3.5	27.8	11	4.2
	Mar 1	9.23	6.35	3.5	21.4	3.2	7.4
	Mar 2	8.69	6.98	3.5	17.9	2.2	5.7
	Sac1	9.57	6.94	3.5	45.6	11.2	13.3
	Sac2	10.13	7.66	4.5	48.1	7.1	13.7
Long term 3 months	Ani3	8.62	6.58	3.5	32.7	8.9	10.2
	Ani4	8.58	6.72	3.2	34.92	5.32	16
	Ani5	7.89	6.23	3	15.3	2.6	3.4
	Mar 3	9.29	6.75	3.5	22.6	9	3.4
	Mar 4	9.17	6.75	3.5	23.6	8.8	4.7
	Mar 5	9.1	6.6	3.5	22	8.5	3
	Sac3	8.61	6.63	3.5	16.6	3.3	1.5
	Sac4	11.14	7.76	4	39.6	12.6	13.5
	Sac5	10.55	7.68	4	38.5	9.4	11.9
Average		9.13	6.73	3.43	25.40	6.37	6.97
Dev. St.		0.96	0.61	0.46	12.10	3.68	4.92

Table S2. Physicochemical parameters of river water at the three sampling sites.

River	Sampling site	Temperature (°C)	Dissolved oxygen (mg/L)	pH	Conductivity (µs/cm)
Aniene	41°56'00.7"N, 12°32'41.3"E	23	6.39	7.79	1284
Marta	42°14'38.8"N, 11°42'41.4"E	25.1	7.5	7.89	720
Sacco	41°42'56.7"N, 13°05'08.7"E	25.8	6.85	7.68	588

Table S3. List of reference libraries employed for the analysis via MicroFTIR, software PARTICLE WIZARDS, Omnic™ Picta™.

Synthetic fibers by Microscope
 Plastic Fibers
 Polymer additives and plasticizers
 HR Polymer additive and plasticizers
 HR Hummel Polymer and plasticizers
 HR Nicolet Sampler Library
 HR Sprouse Polymer additives
 HR rubber compounding materials

Equations S1. Equations employed to obtain the total number of SMPs per gills or GITs and the weight of particles per specimen.

The total number of SMPs per set of gills or per gastrointestinal tract was then calculated according to equation 1 (from Corami et al., 2020a):

$$N_{tot} g^{-1} = \frac{(n*F)}{w} \quad (1)$$

where n = microplastics counted on every field, w = wet weight (expressed in grams) of gills or gastrointestinal tract analyzed, and F = count factor, calculated as follows (equation 2):

$$F = \frac{Filter\ area}{(Count\ field\ area*n\ count\ fields)} \quad (2)$$

Then, the weight of microplastics (μg) per organism is calculated according to equation 3, (from Corami et al., 2020a):

$$\mu g_{SMPs} g^{-1} = \frac{W_{SMPs}}{w} \quad (3)$$

where w_{SMPs} =total weight of small microplastics, μg , and w =wet weight (expressed in grams) of gills or hepatopancreas analyzed. The weight of each microplastic items is calculated according to its volume (V) and its density (ρ). The volume of each item is calculated according to the aspect ratio (elongation ratio). Volumes of items are then calculated according to their geometrical shape (i.e. sphere, ellipse, and cylinder).

Table S4. List of the polymers identified and quantified in the specimens of *A. cygnea* exposed in 3 different rivers for short term and long term, including T0. For each polymer the abbreviation and average density ($g\ cm^{-3}$) are reported.

Polymer	Abbreviation	Density
Acrylic Polymer	ACRYLIC POLYMER	1.19
Aramide	ARAMIDE	1.46
Brominated Polystyrene	BROMINATED PS	1.18
Epoxy resin mixture	EPOXY RESIN MIXTURE	1.2
Ethylene Methyl Acrylate Copolymer	EMA	0.96
Ethylene Vinyl Acetate	EVA	0.94
Modacrylic	MODACRYLIC	1.35
Nitrile-Butadiene Rubber	NBR	1
Polyamide 6	PA-6	1.14
Polyarylamide	PARA	1.51
Polyester	PES	1.38
Polyethylene acrylic acid copolymer-Zinc salt	PEEA-Zn	0.95
Polyolefin	PO	0.95
Polyphthalamide	PPA	1.11
Polypropylene	PP	0.905
Polytetrafluoroethylene	PTFE	2.2
Styrene-Butadiene Rubber	SBR	1.04
Synthetic urethane Rubber	SYNTHETIC URETHANE RUBBER	1.21

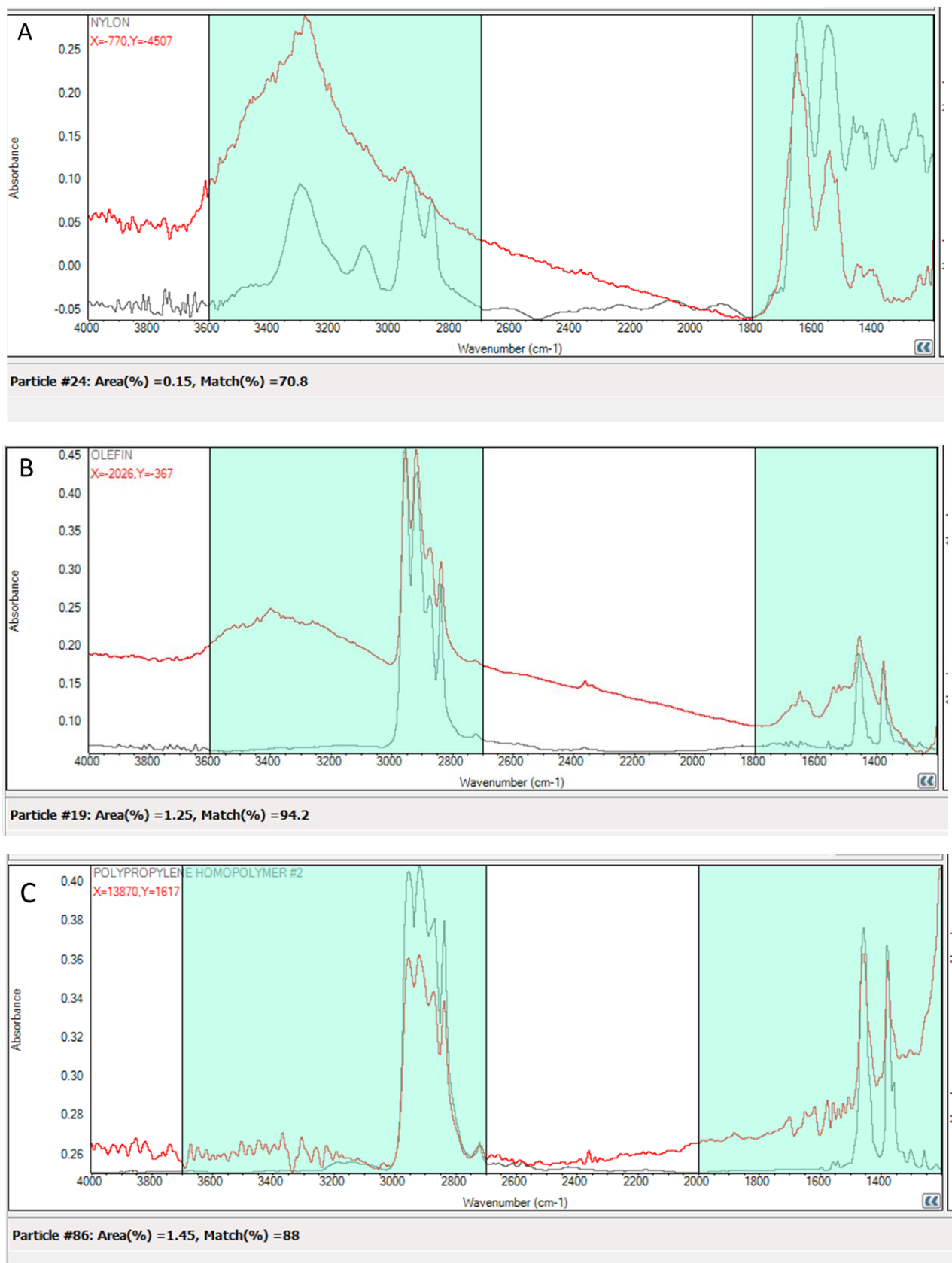


Figure S1. Some examples of the best FTIR spectra of polymers collected on the SMPs observed in gills and GITs of *Anodonta cygnea*. (A) Nylon or polyamide (PA), (B) polyolefin (PO) and (C) polyaryamide polypropilene (PP). The FTIR spectrum of the polymer collected is in red, the spectrum of the polymer present in the suite of reference libraries employed is in black. Spectral range of 4000–1200 cm⁻¹, 100-mm step size scanning (spatial resolution) at 100–100 mm aperture, and 32 co-added scans at the spectral resolution of 4 cm⁻¹. Optimal Match % of polymers was $\geq 65\%$.

Table S5. Microplastic particles size measured in μm as length (L, the longest dimension) and width (W, perpendicular to the length) found in gills and GITs.

		T0		Aniene ST		Aniene LT		Marta ST		Marta LT		Sacco ST		Sacco LT	
		L	W	L	W	L	W	L	W	L	W	L	W	L	W
Gills	Mean	62.5 4	25.1 3	48.4 3	24.5 5	53.2 7	26.5	45.0 5	25.1 9	59.5 5	32.4 8	43.2 3	21.1 3	56.7 5	30.9
	SD	5.2	0.68	1.22	0.49	2.15	0.95	2.89	1.53	5.59	3.62	0.44	0.2	4.4	2.8
	Dimensional range{	23.7	14	25.9	80.3	25.6	17.1	27.3	13.7	30.1	18.4	25.4	12.1	31.1	14.9
		500	66.6 2	200	80.3	116. 2	59.6	94.6	46.2	121. 2	72.7	107. 9	52.3	183. 7	122. 1
GITs	Mean	58.9 7	29.1 4	57.0 2	30.5	52.8 8	26.9 4	70.2 9	35.8 9	61.9 2	30.1 3	48	26.7 5	59.6	34.3
	SD	7.33	3.07	2.43	1.25	0.94	0.48	7.01	3.63	1.54	0.78	6.61	4.88	2.44	1.26
	Dimensional range{	26	17.6	26.5	17.5	26.8	13.5	27.5	13.7	28	13	5	5	27.5	16.6
		229. 8	97.9	114. 1	72	200. 7	127. 8	187. 3	93.9	221. 1	118. 7	105. 2	69.6	117	64.5

Table S6. List of the additives, plasticizers, and other micro-litter components identified and quantified in the specimens of *A. cygnea* exposed in 3 different rivers for short term and long term, including T0. For each additive, the abbreviation and average density (g cm^{-3}) are reported.

Additive	Abbreviation	Density
50% active glycerol monostearate in polyethylene carrier	GMS	0.97
Benzotriazole	BTA	1.36
Bromodichlorometahne	CHBrCl ₂	1.98
Butyl vinyl ether	NBVE	0.78
Calcium pelargonate	CALCIUM PELARGONATE	0.900
Calcium stearate	CS	1.08
Calcium sulfate dihydrate	CALCIUM SULFATE DIHYDRATE	2.22
Chloroalkyl phosphate ester	CPE	0.98
Cocoamidoproyl betaine	CAPB	1.05
Dodecenylsuccinic anhydride	DDSA	1.01
Hombitan TiO ₂	HOMBITAN TI02	0.6
Magnesium hydroxide	Mg(OH) ₂	2.34
Methyl alkyl imidazoline sodium salt	METHYL ALKYL IMIDAZOLINE SODIUM SALT	1.03
Methyl chloride	Methyl chloride	0.997
N-(2-ethoxyphenyl)-n-(2-ethylphenyl)-ethanediamide	NNE	1.2
Nichel-dibuthyldithiocarbamate	NDBC-O	1.3
Octadecanoic acid, calcium salt	OC SALT	0.941
Octenylsuccinic anhydride succinic	OCTENYLSUCCINIC ANHYDRIDE SUCCINIC	0.866
O-nitrobiphenyl	O-NITROBIPHENYL	1.44
Pentabromodiphenyl oxide	PENTABROMODIPHENYL OXIDE	2.27
Phosphate ester fatty acids blend	FAPE	1.15
Poly(N-methyl acrylamide)	PNMA	1.322
Polyether, quaternary ammonium salt mixture	POLYETHER, QUATERNARY AMMONIUM SALT MIXTURE	1.3
Polyvinylpyrrolidone	PVP	1.2
Propyl zithate	ZP-2-YMDT	3.5

Propylene carbonate	PC	1.2
Propylene glycol monostearate	PGMS	0.88
Rayon	RAYON	1.51
Reoflam dmmp	REOFLAM DMMP	1.145
Silk	SILK	1.3
Sodium polyacrylate, polyacrylic water	ACR	1.22
Stearamidopropyldimethyl- hydroxyethylammonium-dihydrogen	DIHYDROGEN PHOSPHATE	0.874
Triphenyl methane 4,4',4''- triisocyanate in ethyl acetate	FT172229	1.014
Varox 231 xl	VAROX	0.877
