

Supplementary information

What are the drivers of microplastic toxicity? Comparing the toxicity of plastic chemicals and particles to *Daphnia magna*

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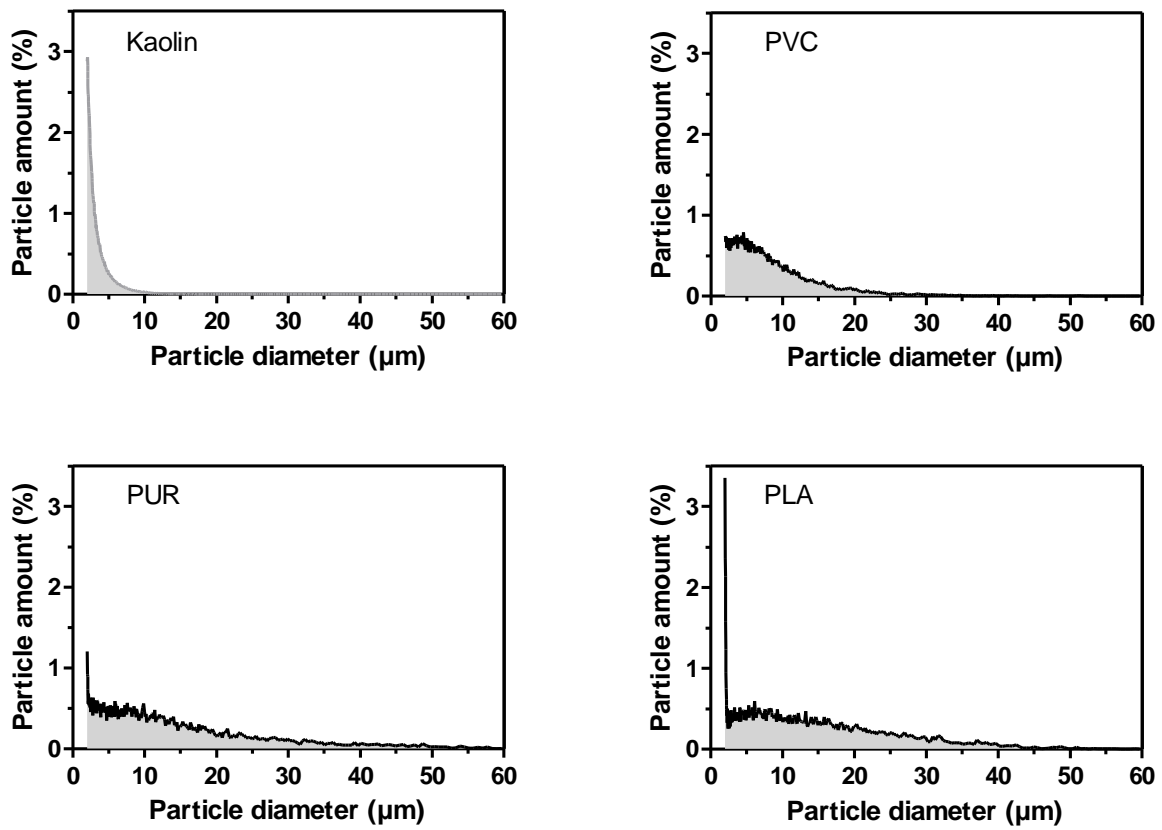


Figure S1: Relative particle size distributions for kaolin as well as PVC, PUR and PLA particle suspensions (100 mg/L in Elendt M4 medium) using a Beckman Coulter Multisizer 3.

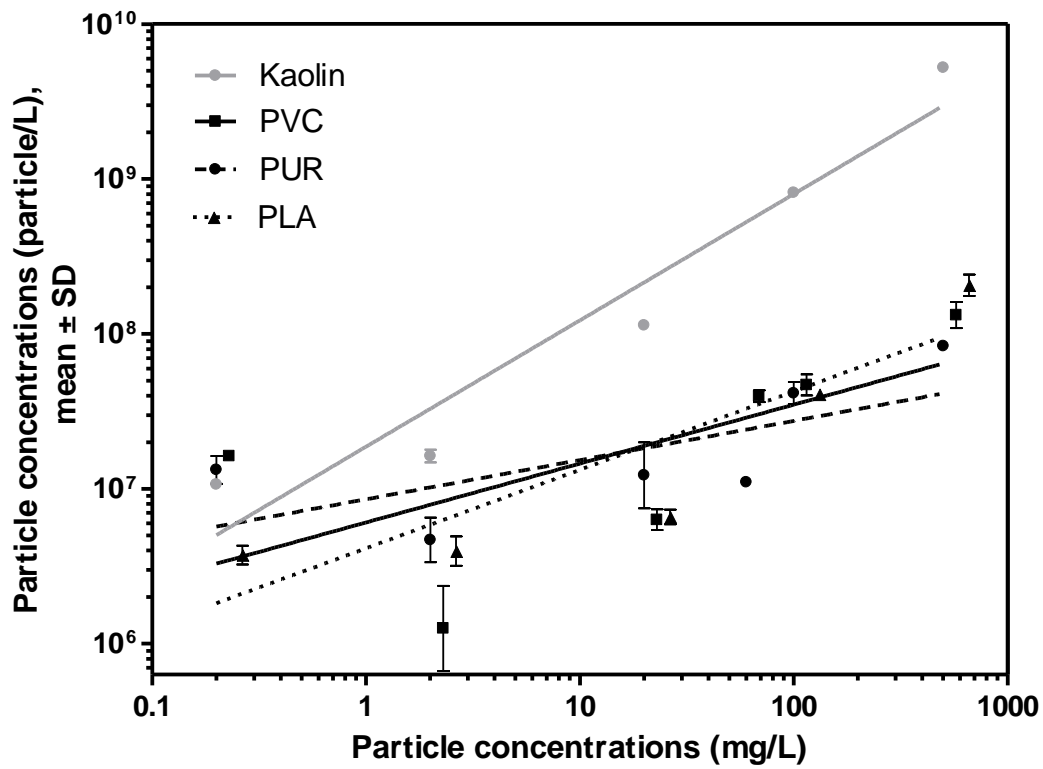


Figure S2: Calibration curve for nominal mass-based vs. measured numerical concentrations/L of microplastics and kaolin suspensions. SD: standard deviation.

Table S1: Nominal mass-based and mean measured numerical concentrations in the size range of 2.0 and 60 μm in kaolin, PVC, PUR and PLA suspensions used in the first chronic exposure experiment with *Daphnia magna*. Numerical concentrations were measured in vessels prepared identically to those used to expose daphnids but did neither contain algae nor animals. Numerical concentrations of exposures were corrected for the mean particle concentration in the Elendt M4 medium. Means are of three to six (0 mg/L) technical replicates \pm standard deviation (SD).

Treatment	Nominal mass concentrations (mg/L)	Mean measured numerical concentrations \pm SD (particle/L)
Elendt M4	0	$1.23 \times 10^6 \pm 5.97 \times 10^5$
Kaolin	10	$1.06 \times 10^8 \pm 1.52 \times 10^6$
	50	$4.70 \times 10^8 \pm 6.76 \times 10^6$
	100	$9.42 \times 10^8 \pm 2.44 \times 10^7$
	500	$4.75 \times 10^9 \pm 1.62 \times 10^8$
	PVC	10
PVC	50	$8.27 \times 10^6 \pm 8.03 \times 10^5$
	100	$8.12 \times 10^7 \pm 7.67 \times 10^6$
	500	$2.53 \times 10^8 \pm 3.18 \times 10^7$
	PUR	10
PUR	50	$2.31 \times 10^7 \pm 2.07 \times 10^6$
	100	$4.53 \times 10^7 \pm 2.27 \times 10^6$
	500	$1.13 \times 10^8 \pm 1.03 \times 10^7$
	PLA	10
PLA	50	$2.08 \times 10^7 \pm 1.10 \times 10^6$
	100	$4.46 \times 10^7 \pm 4.86 \times 10^6$
	500	$1.66 \times 10^8 \pm 1.78 \times 10^7$

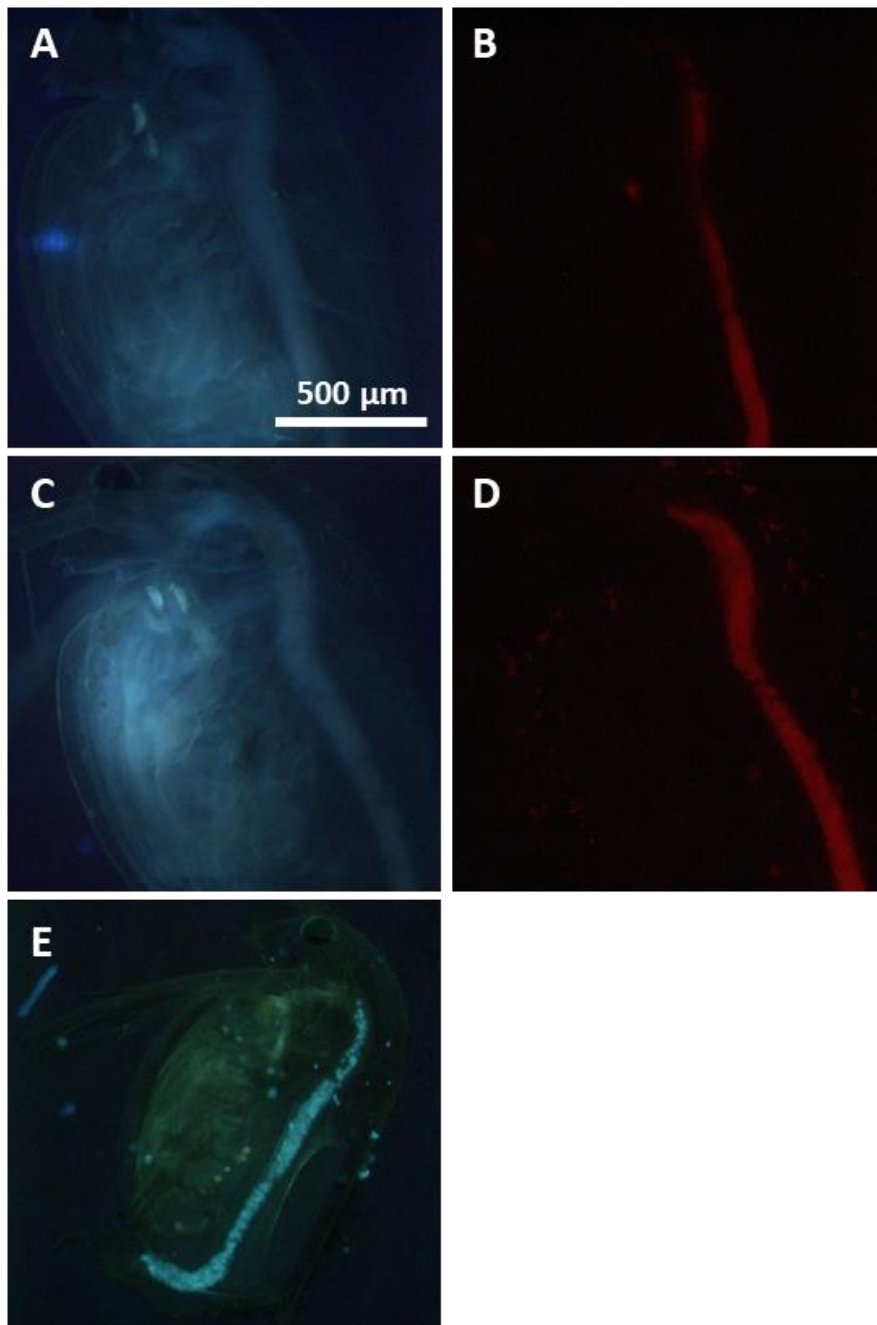


Figure S3: Fluorescence microscopy images of PVC (A + B), PLA (C + D) and PUR (E) microplastic ingestion by *Daphnia magna*. While PVC (B) and PLA (D) microplastics were stained with Nile red, PUR (E) was clearly visible without staining.

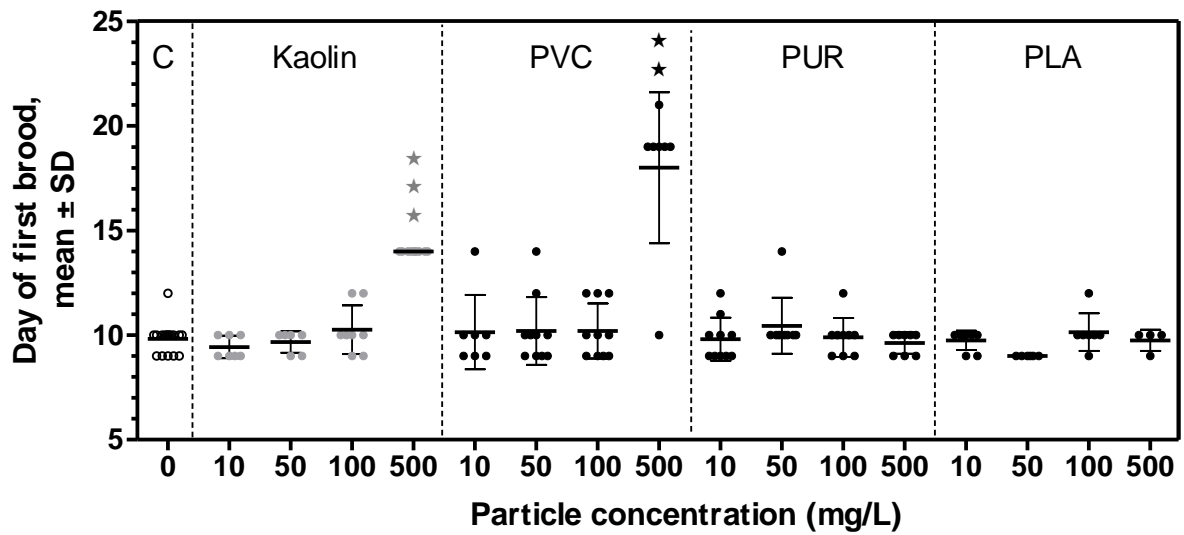


Figure S4: Timing of reproduction of *Daphnia magna* in 21 days exposure to 10, 50, 100 and 500 mg/L of PVC, PUR, PLA microplastics and kaolin. Data is presented as mean \pm standard deviation (SD). Asterisks indicate significant differences to the control (C): ** $p < 0.01$, *** $p < 0.001$ (Kruskal-Wallis with Dunn's multiple comparison post-test).

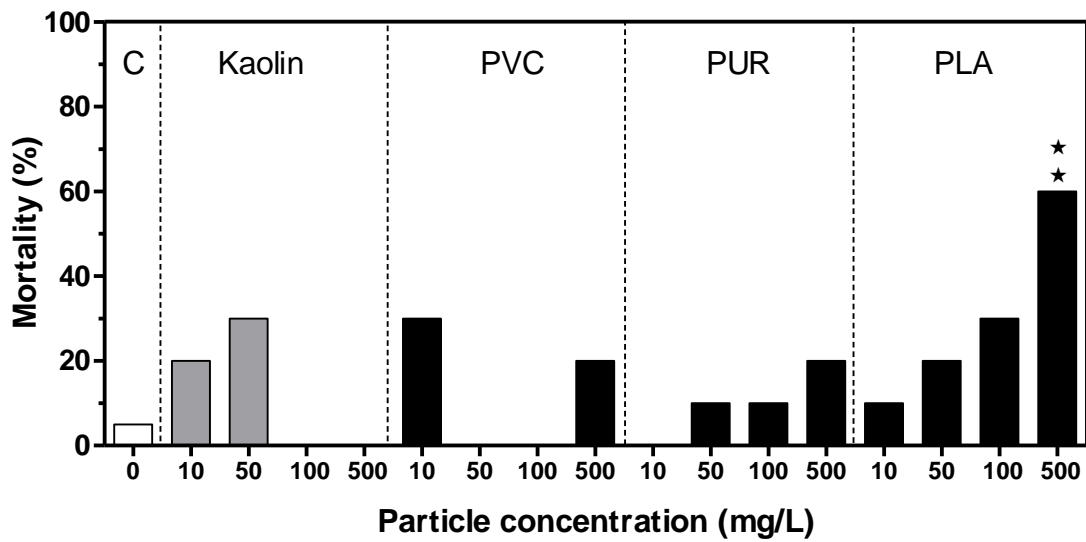


Figure S5: Mortality of *Daphnia magna* after exposure to 10, 50, 100 and 500 mg/L PVC, PUR, PLA microplastics and kaolin for 21 days. ** $p < 0.01$ (Fisher's exact test, comparison to the control (C)).

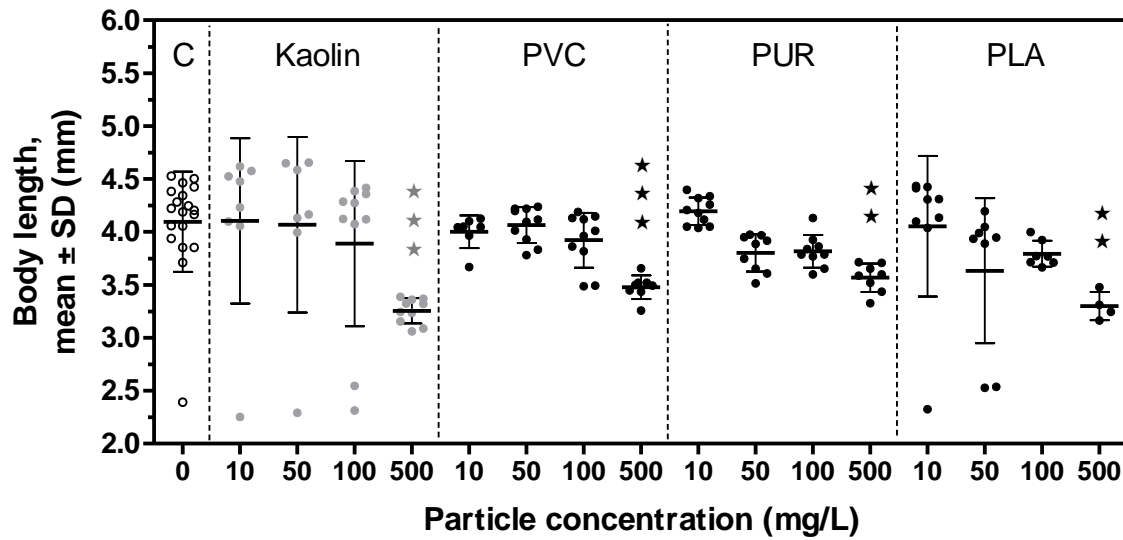


Figure S6: Size of adult *Daphnia magna* individuals after 21 days of exposure to kaolin as well as PVC, PUR and PLA microplastics. Data is presented as mean \pm standard deviation (SD). Asterisks indicate significant differences to the control (C): ** $p < 0.01$, *** $p < 0.001$ (Kruskal-Wallis with Dunn's multiple comparison post-test).

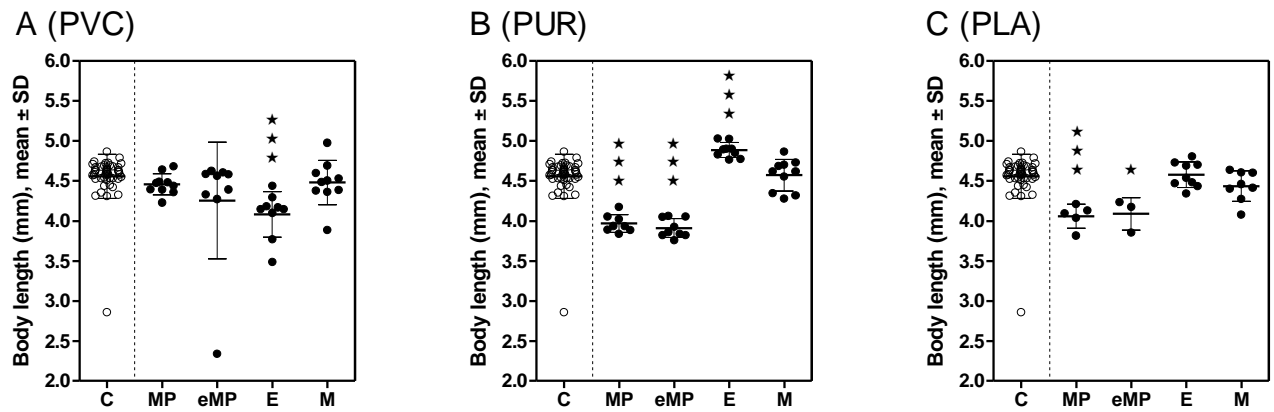


Figure S7: Size of adult *Daphnia magna* individuals after 21 days exposure to 45.5 mg/L PVC (A), 236 mg/L PUR (B) and 122 mg/L PLA (C) microplastics. Treatments include microplastics (MP), microplastics without extractable chemicals (eMP), the chemicals extracted (E) and migrating from microplastics (M). Data is presented as mean \pm standard deviation (SD). Asterisks indicate significant differences to pooled controls (C): \star $p < 0.05$, $\star\star\star$ $p < 0.001$ (Kruskal-Wallis with Dunn's multiple comparison post-test).

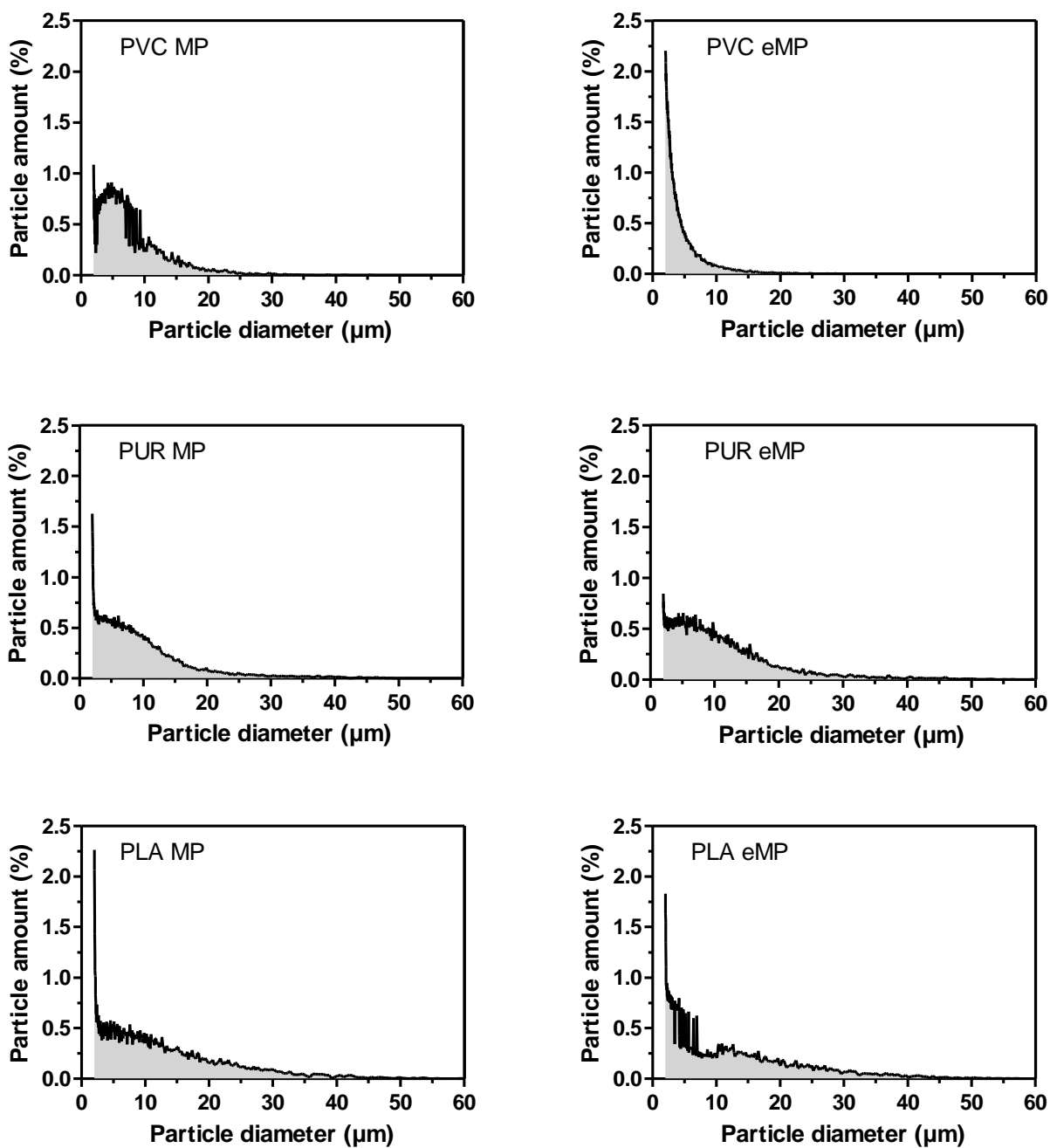


Figure S8: Relative particle size distributions of microplastics (MP) and microplastics without extractable chemicals (eMP). Size distributions were derived from exposure suspensions (PVC: 45.5 mg/L, PUR: 236 mg/L, PLA: 122 mg/L) in Elendt M4 medium using a Beckman Coulter Multisizer 3.

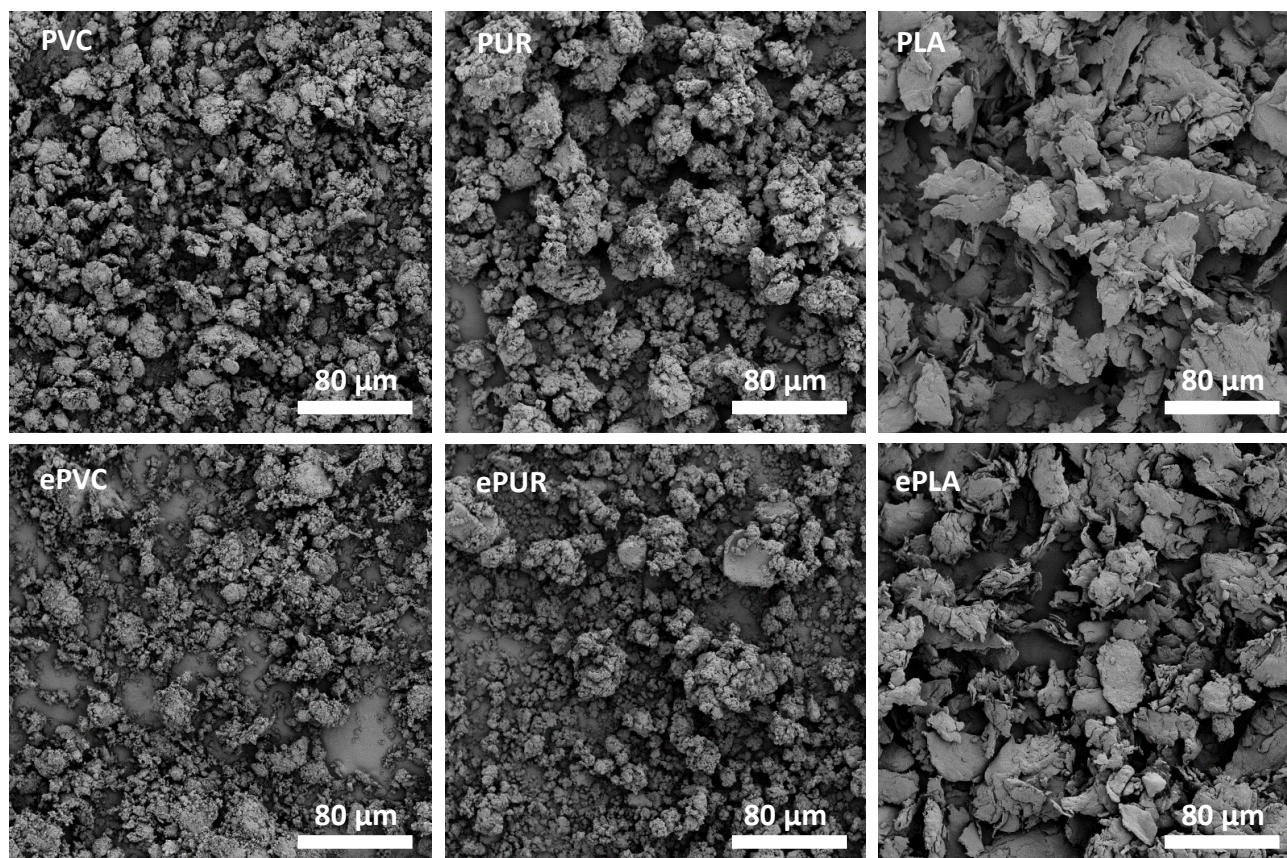


Figure S9: Scanning electron microscope (SEM) images of untreated PVC, PUR and PLA microplastics (MP, top) and extracted microplastics (e, bottom, 300× magnification).