

Supplementary Information

Heteroaggregates of Polystyrene Nanospheres and Organic Matter: Preparation, Characterization and Evaluation of Their Toxicity to Algae in Environmentally Relevant Conditions

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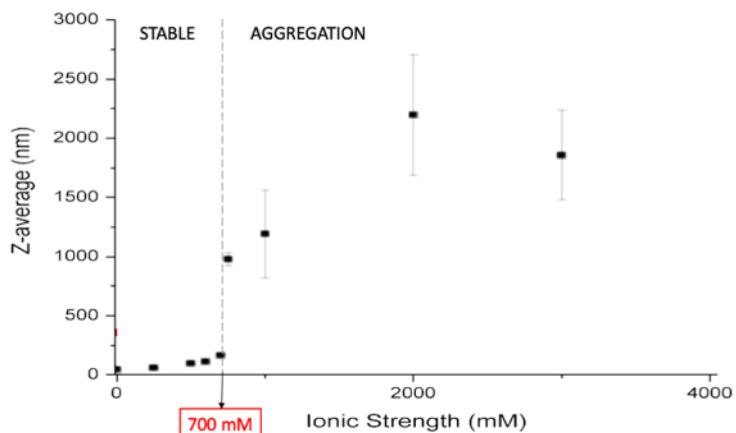


Figure S1: Size of the PS50 dispersion as a function of the ionic strength (with monovalent ions, KCl).

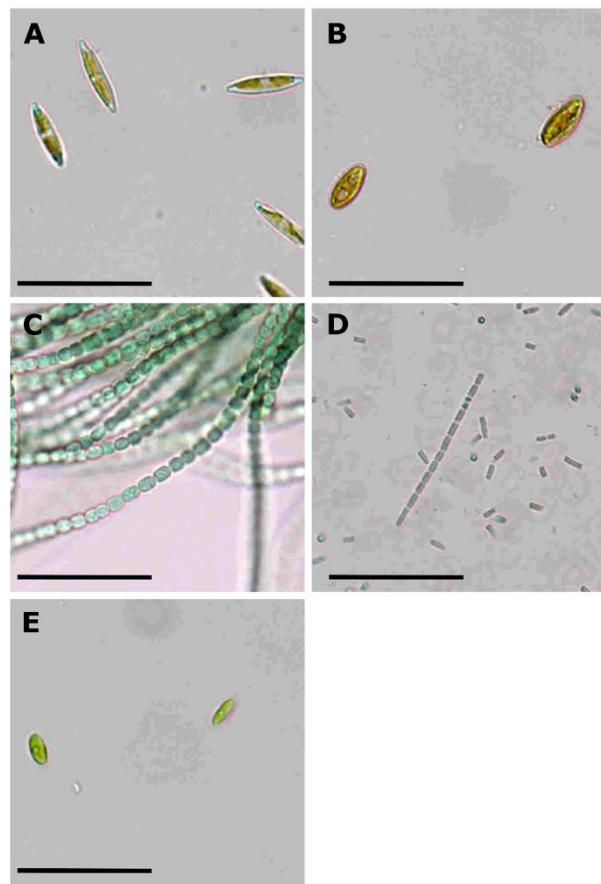


Figure S2. Micrographs of the 5 algal strains used in this study: *Nitzschia palea* (A), *Gomphonema parvulum* (B), *Nostoc* sp. (C), *Komvophoron* sp. (D) and *Scenedesmus obliquus* (E). Scale bar represents 10 μm .

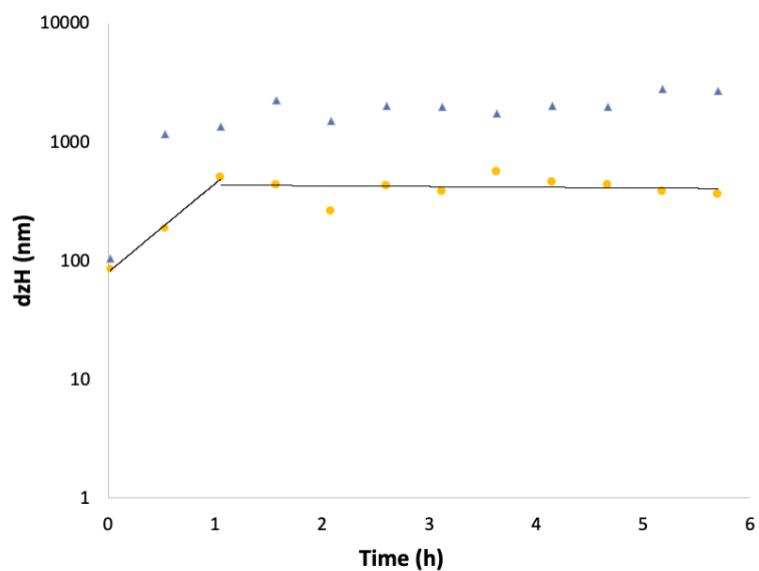


Figure S3: Kinetics of aggregation of PS50 (blue triangles) and PS50-HA (orange circles) in NaCl solution (700 mmol.L⁻¹, pH 7) : the first slope corresponding to a fast aggregation regime and the second one to a rearrangement phase.

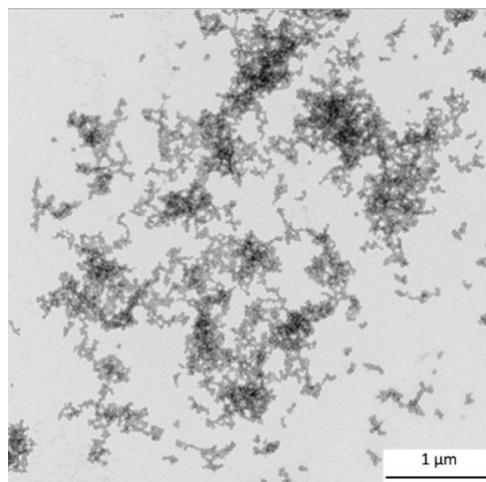


Figure S4: TEM observations of (a) PS-50 homoaggregation ($I = 700 \text{ mmol.L}^{-1}$).

Table S1: Characterization of biological media.

	T °C	pH	Conductivity mS/cm	Ionic strength (I) mmol.L ⁻¹
BGII	16.7	7.68	2.23	23.77
COMBO	16.1	7.88	0.338	5.98

Table S2: Characterization of the nanospheres by DLS.

Z-Average		PdI	Intensity Mean	Number Mean	Volume Mean	Derived Count Rate
	nm		nm	nm	nm	kcps
PS50	44.59	0.050	47.21	35.48	40.44	15691.2
	45.22	0.048	48.01	35.35	40.60	15828.6
	45.67	0.050	48.63	35.42	40.83	15635.8
347.0		0.019	364.1	320.5	379.8	16182.4
351.6	PS350	0.099	355.6	329.7	365.5	16583.1
347.0		0.158	346.2	322.5	354.9	16165.7
				0	44.59	0.05

Table S3: Zeta potential of particles in the exposure media after adjustment of the pH to 7 (10 mg/L). In brackets are reported the standard deviation values ($n = 4$).

Particles	Zeta potential	0	Zeta potential	45.67	0.05
	in COMBO exposition medium (mV)		In BG11 exposition medium (mV)		
PS350	-34.7 (\pm 1.3)		-48.7 (\pm 2.1)		
PS50	-24.3 (\pm 1.3)		-30.6 (\pm 1.9)		
PS50-HA	-25.2 (\pm 1.4)		-37.1 (\pm 3.4)		

Table S4: Chemical composition of COMBO algal exposure media.

Ingredients	Quantity (mg.L ⁻¹)
CaCl ₂ . 2H ₂ O (147.02)	36.76

MgSO₄. 7H₂O (246.48)	36.97
KH₂PO₄ (136.09)	8.71
NaNO₃ (84 .99)	85.01
NaHCO₃ (84.01)	12.60
Na₂O₂SiO₂. 2H₂O (218.16)	21.84
H₃BO₃ (61.83)	24.00
MnCl₂. 4H₂O (197.844)	0.18
CuSO₄. 5 H₂O (249.612)	0.001
ZnSO₄. 7 H₂O (287.54)	0.022
CoCl₂. 6 H₂O (237.839)	0.01
Na₂MoO₄. 2 H₂O (241.92)	0.022
NaVO₂ (121.93)	1.20
Na₂EDTA. 2H₂O * (328.25)	4.36
FeCl₃. 6H₂O (270.30)	1.50

Table S5: Chemical composition of BG11 algal exposure media.

Ingredients	Quantity (mg.L ⁻¹)
CaCl₂. 2H₂O (147.02)	36.0
MgSO₄. 7H₂O (246.48)	75.0
KH₂PO₄ (136.09)	28.6
NaNO₃ (84 .99)	1500.0
Na₂CO₃ 10H₂O (286.14)	54.0
H₃BO₃ (61.83)	29.0
C₆H₈O₇. H₂O (210.06)	6.0
MnCl₂. 4H₂O (197.844)	1.8
CuSO₄. 5 H₂O (249.612)	0.1
ZnSO₄. 7 H₂O (287.54)	0.2
Co(NO₃)₂. 6H₂O (290.933)	0.05
Na₂MoO₄. 2 H₂O (241.92)	0.4
Na₂EDTA. 2H₂O * (328.25)	1.0
Fe(III)(NH₄)₃ citrate	6.0