Electronic Supplementary Material (ESI) for Environmental Science: Nano.

Supplementary Information

Nano and submicron fluorescent polystyrene particles internalization and translocation in seedlings of *Cichorium endivia* L.

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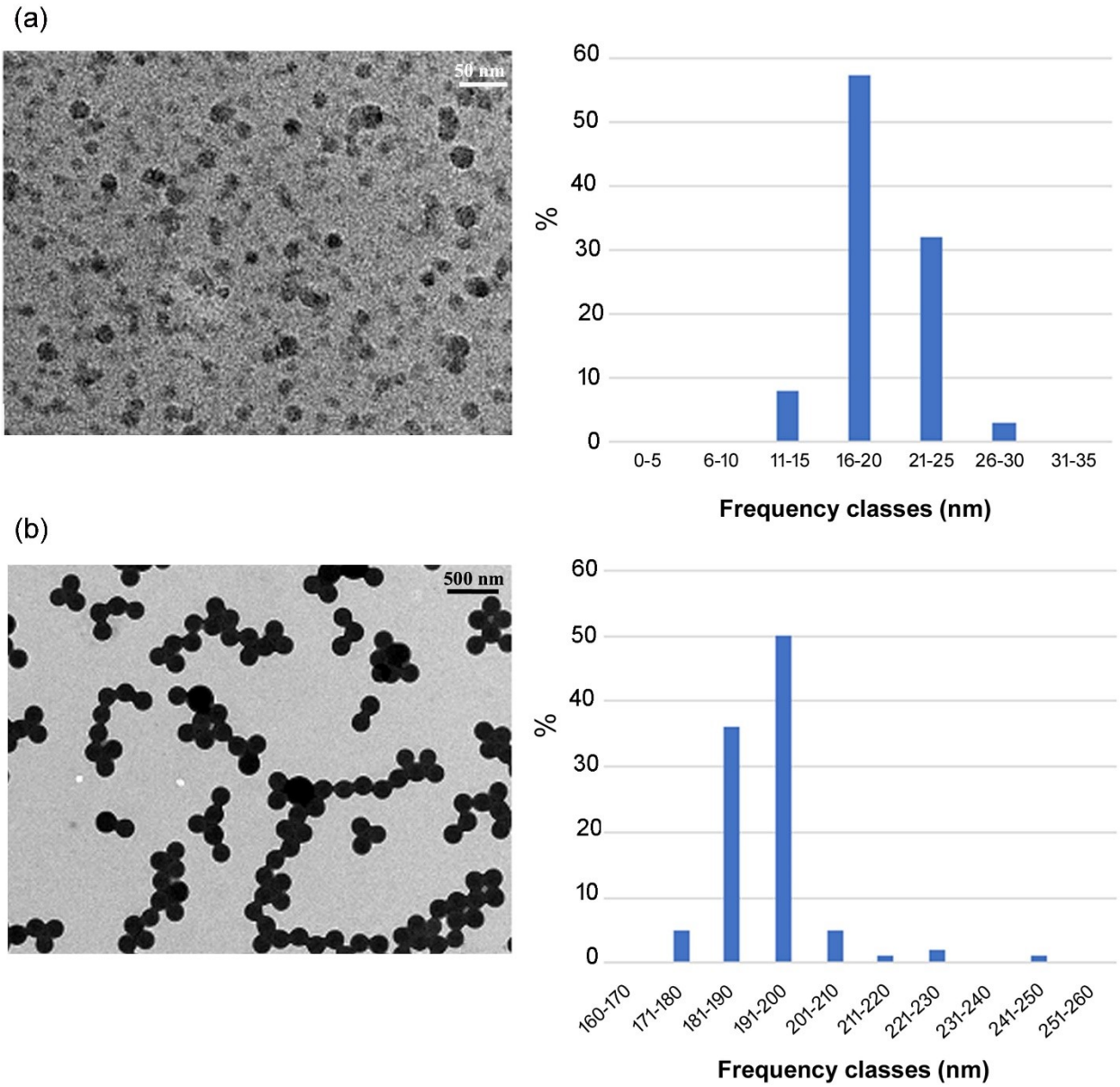

Fig S1 Frequency classes and TEM image of polystyrene particles. nPS (a); smPS (b).

Dynamic Light Scattering experiments have been performed with a Zetasizer Nano ZS90 instrument (Malvern, UK). The instrument uses a He/Ne laser (wavelength 633 nm) and the scattering angle is 173°. Hydrodynamic diameters are expressed as Z-average values. Particle size distribution curves were obtained by the instrument software using the non-negative least square fitting method. ζ-potential values were determined from the electrophoretic mobility measurements applying the Smoluchowski approximation (Table S1†).

Table S1. DSL and ζ potential measurements

Sample	Z-Average (nm)	peak ± standard deviation (nm)	PdI	ζ-potential (mV)
nPS (20 nm)	20±1	21±9	0.36±0.07	-40±3
smPS (200 nm)	167±3	183±55	0.08±0.02	-60±1

PdI is polydispersity index

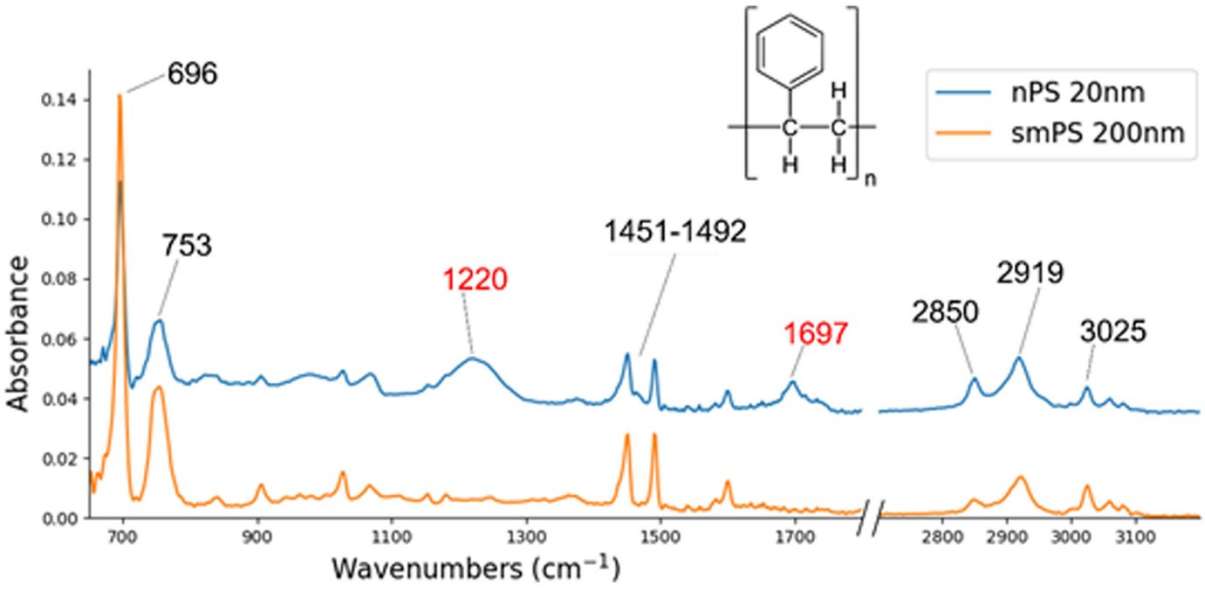

Fig S2 ATR-FTIR spectra of PS particles. Spectrum of nPS is uplifted from baseline for clarity of visualization. Distinctive peaks of PS are marked with corresponding wavenumbers. Peaks at 1220 and 1697 cm^{-1} (red) are not associated with PS IR fingerprint. Peak at 1220 cm^{-1} is assigned to surfactant and peak at 1697 cm^{-1} to fluorescent dye.

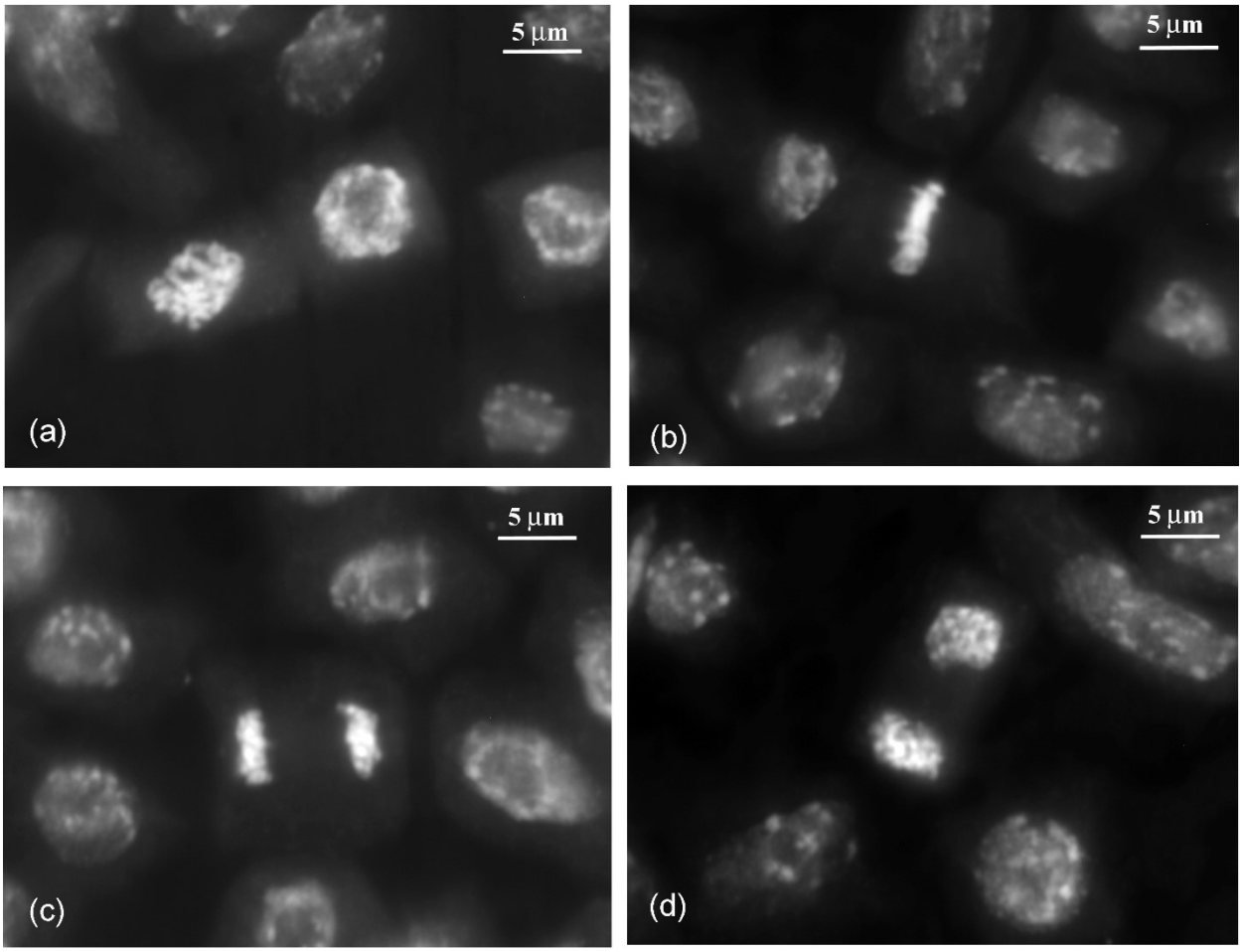

Fig S3 Cytological figures of 5 days control root meristems of *Cichorium endivia* detected at fluorescence microscope after Feulgen staining. Normal prophase (a), normal metaphase (b) normal anaphase (c) and normal telophase (d).

Cytogenetic analysis

Mitotic phases	Prophases %	Metaphases %	Abn Metaphases %	Ana/Telophases %	Abn Ana/Telophases %
Control	42.6 ±12.8b	25.6 ±9.3b	8.8 ±3.4a	23 ±5.1a	0 a
0.01 g L^{-1} nPS	27 ±7.4ab	14.8 ±3.6ab	25.4 ±7.3b	21.1 ±6.4a	11.8 ±3.2b
0.1 g L^{-1} nPS	22.6 ±7a	15 ±1.1ab	36.1 ±2.3b	17.8 ±5.1a	8.5 ±2.3b
1 g L^{-1} nPS	24 ±4.8ab	13.6 ±4.1ab	28 ±4.2b	20.6 ±4a	13.7 ±2.2b
0.01 g L^{-1} smPS	28.7 ±4.8ab	17.6 ±4.1ab	29.3 ±7.4b	17 ±1.5a	7.3 ±0.4b
0.1 g L^{-1} smPS	16.9 ±4.4a	10.4 ±6a	35.1 ±4.3b	24.6 ±2.9a	13 ±3.9b
1 g L^{-1} smPS	31.3 ±8.4ab	12.5 ±3.8a	27.7 ±2.7b	14.9 ±3.6a	13.6 ±3.3b

Table **S2**. Cytological analysis of *Cichorium endivia L.*. root meristems in control and after 5 days treatment with 0.01, 0.1, 1 g L^{-1} nPS and smPS particles. Mitotic phases were expressed as mean values ± standard deviation on 100 mitoses analysed. Within colums, values followed by different letters are statistically significant with Tukey test for $P \leq 0.05$

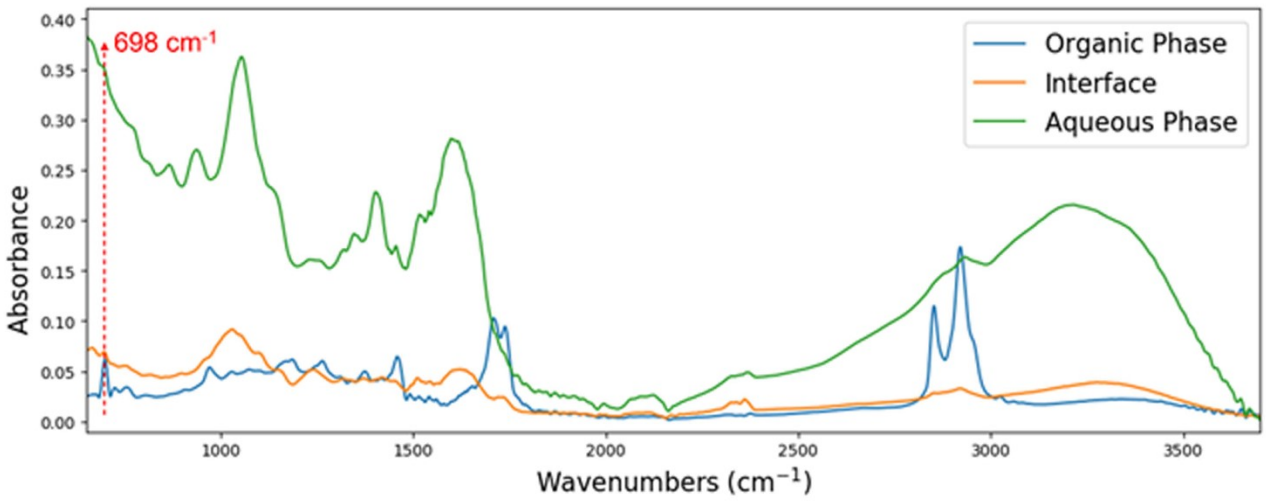

Fig. S4 ATR-FTIR spectra of extracts of nPS 1 g L^{-1} treated seedlings of *Cichorium endivia* of 14 days. Spectra of all resulting phases are shown: organic phase (blue); interface (orange); aqueous phase (green). PS is clearly detectable only in the organic phase (698 cm^{-1}).

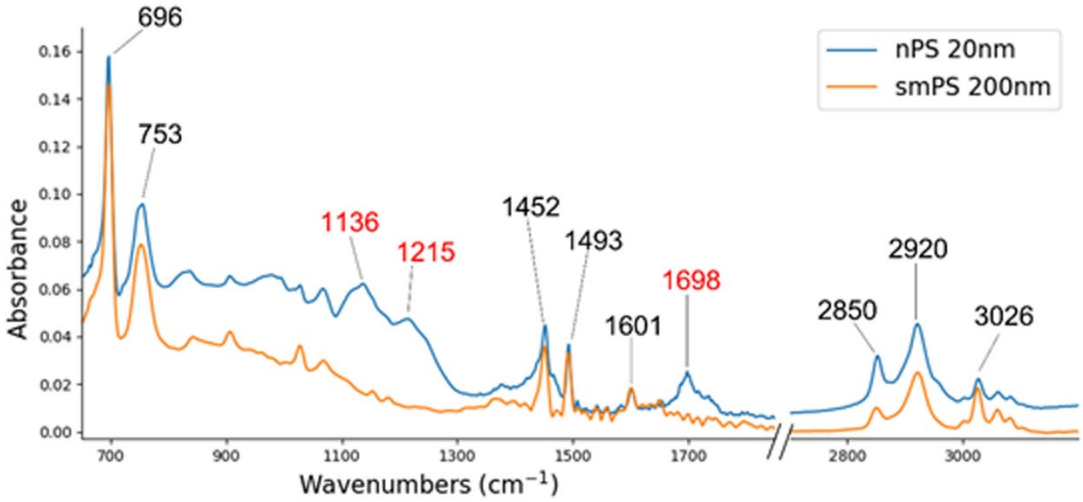

Fig. S5 ATR-FTIR spectra of nPS and smPS particles of stock solutions processed with the organic solvent extraction protocol. Spectra of polymer particles of both sizes show the infrared signature characteristic of PS. As for raw particles (Fig. 2), spectrum of nPS (blue) presents peaks around 1220 and 1700 cm^{-1}, due to surfactant and fluorescent dye.

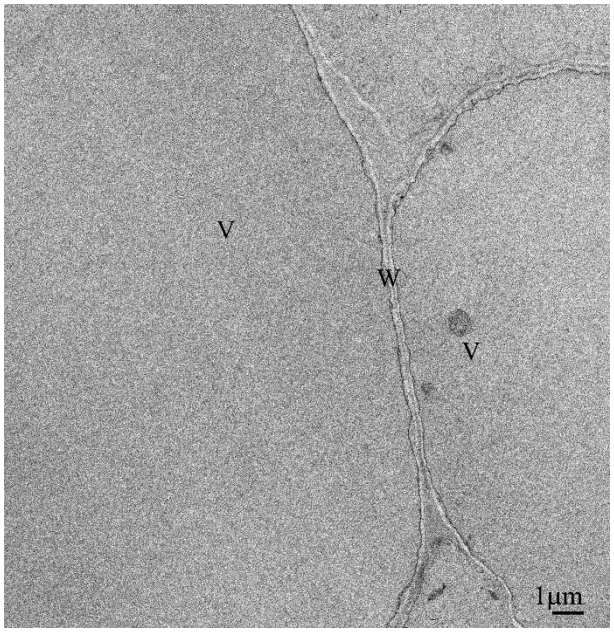

Fig. S6 TEM images of control root of *Cichorium endivia*, of 14 days. Cortical cells with large central vacuoles. V, vacuole; W, cell wall.