

## SUPPLEMENTARY INFORMATION

### Impact of magnetic nanoparticle surface coating on their long-term intracellular biodegradation in stem cells

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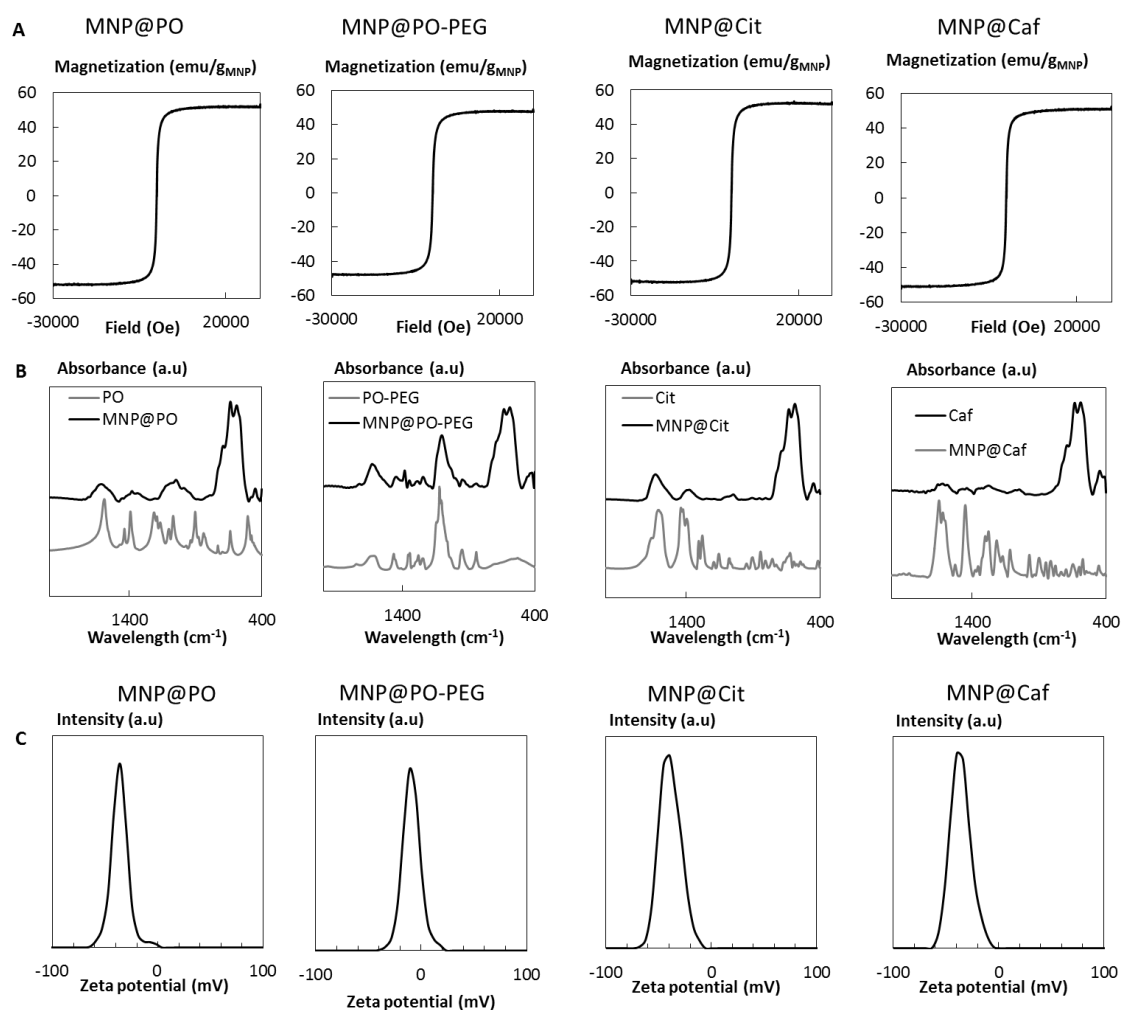
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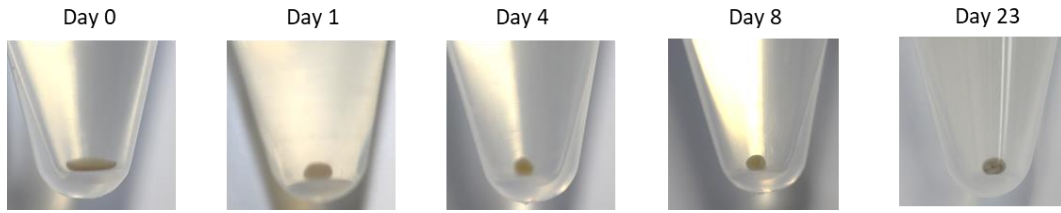
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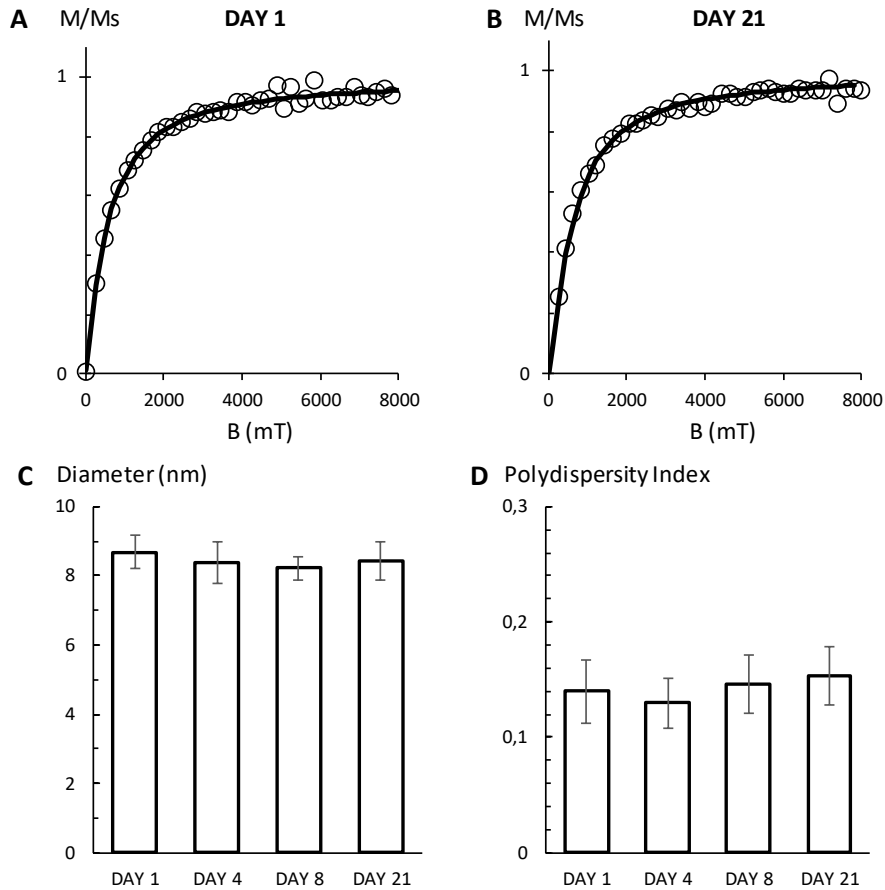
**Figure S1:** Physico-chemical characterisations of coated MNP. **A.** Magnetization curves at room temperature. **B.** FTIR spectra of each coated MNP and corresponding coating molecule. **C.** Zeta potential curves.

Ligand	Ligand/MNP	Chelating function/nm <sup>2</sup>
PO	670	2.6
PO-PEG	355	1.4
Cit	350	1.4
Caf	700	2.7

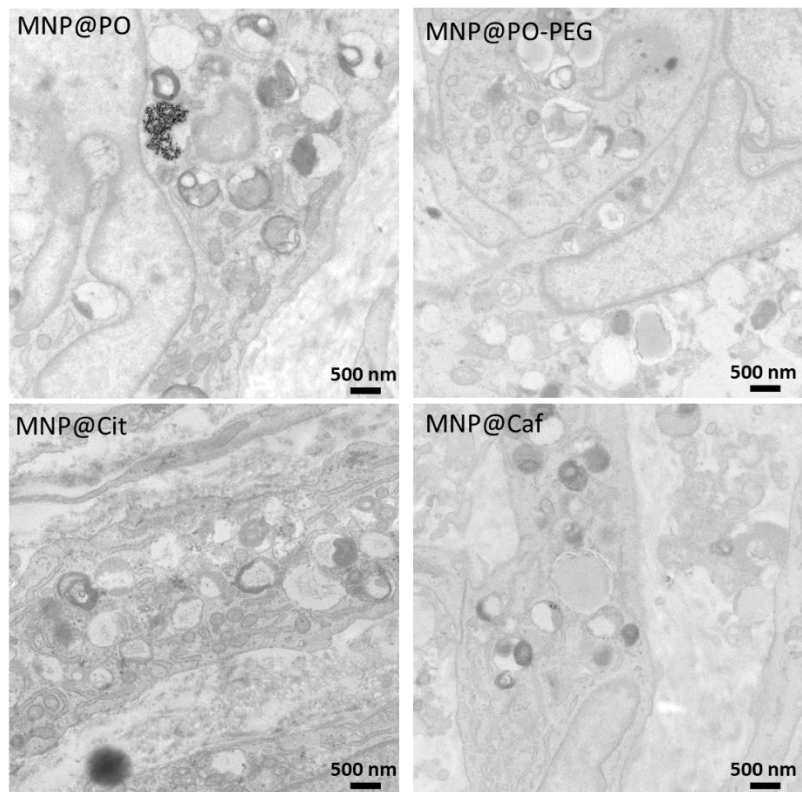
**Figure S2:** Quantification of ligand molecules at the surface of each nanoparticle. Available surface for the chelating functions (considering 3 chelating functions for Cit)



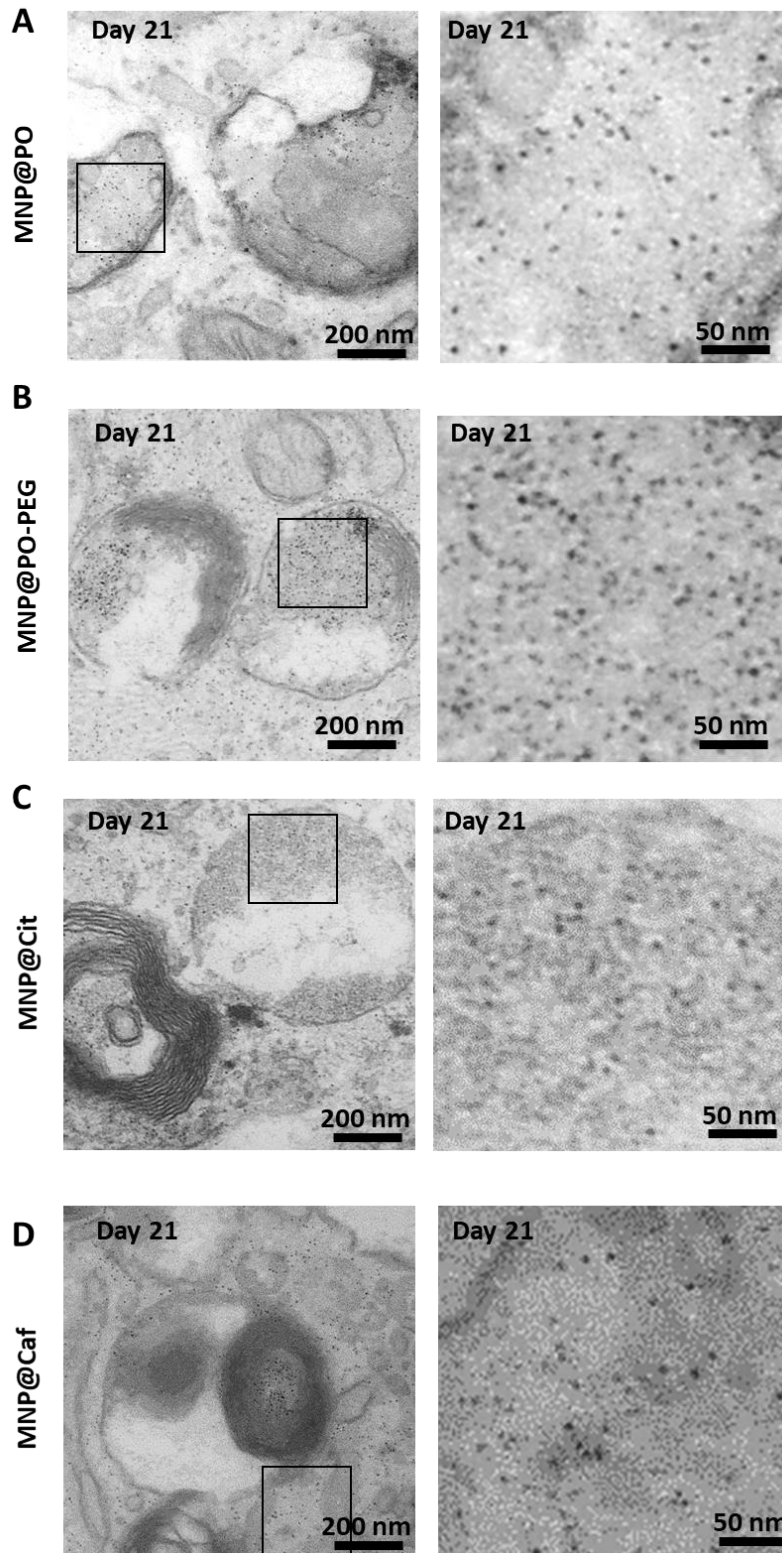
**Figure S3:** Camera images of the formation and maturation of stem-cells spheroid loaded with MNP after 0, 1, 4, 8 and 21 days.



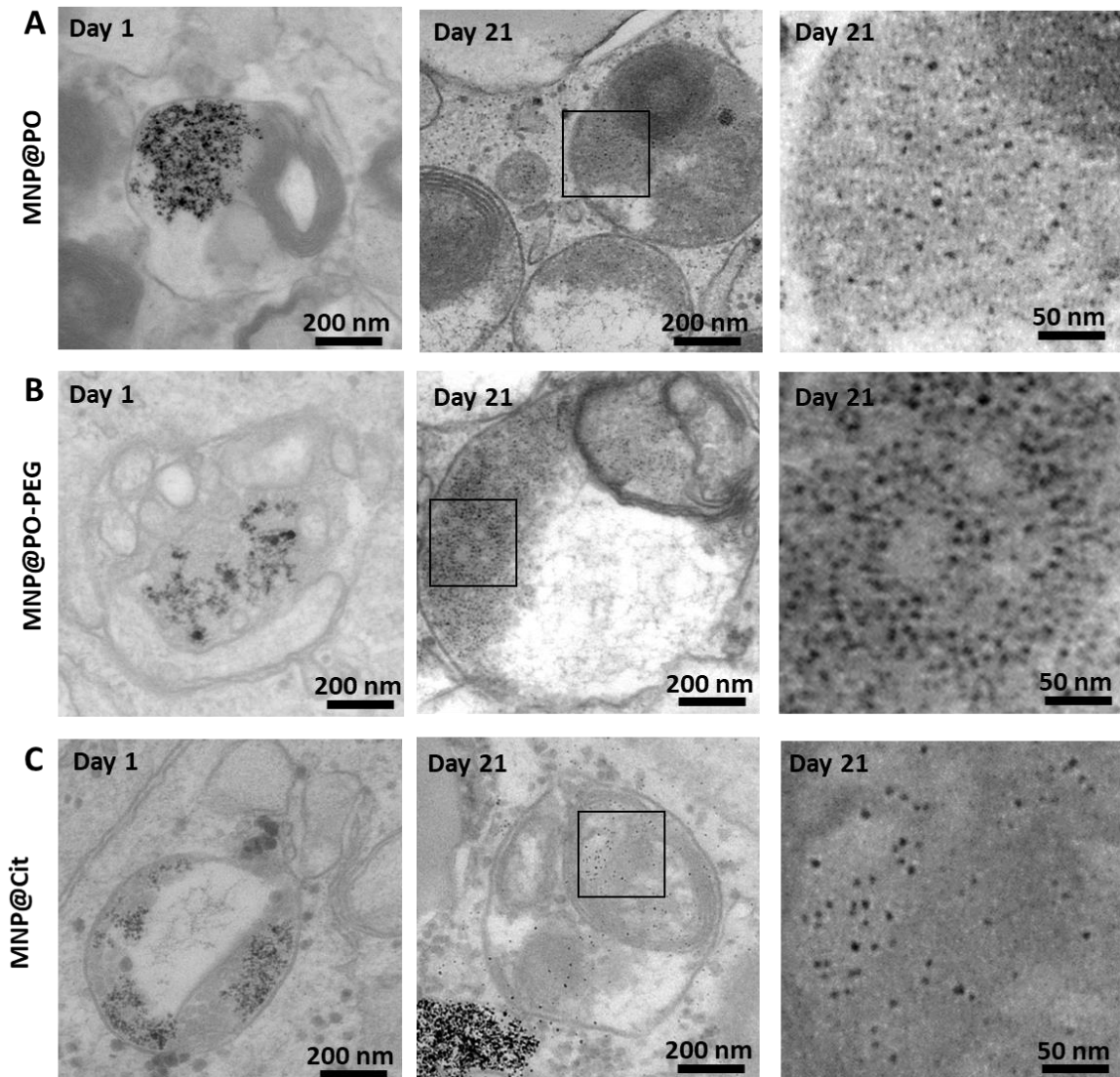
**Figure S4: A-B.** Typical magnetization curve at day 1 (**A**) and day 21 (**B**) of tissue maturation. The original data (empty circles) and the fit obtained with a Langevin formalism (black line) are plotted. To perform the analysis, the nanoparticle magnetic moment is expressed as Langevin law  $M(H) = m_s \phi (\coth \xi - 1/\xi)$ , with  $\xi = 10^{-4} M_s \pi d^3 B / 6kT$  the Langevin parameter,  $B$  being the applied magnetic field,  $k$  the Boltzmann constant,  $T$  the temperature and  $M_s$  the saturation magnetization of the magnetic material. Then, because the nanoparticles population is polydisperse in size, this Langevin expression must be weighted by a lognormal distribution of the nanoparticle diameter  $d$ :  $P(d) = \frac{1}{(\sqrt{2\pi}\sigma d)} \times \exp\left(-\frac{\ln^2\left(\frac{d}{d_{mag}}\right)}{2\sigma^2}\right)$ , with  $\sigma$  is the polydispersity index. **C-D.** Average  $d$  (**C**) and  $\sigma$  (**D**) values obtained for the different degradation times (days 1, 4, 8, and 21).



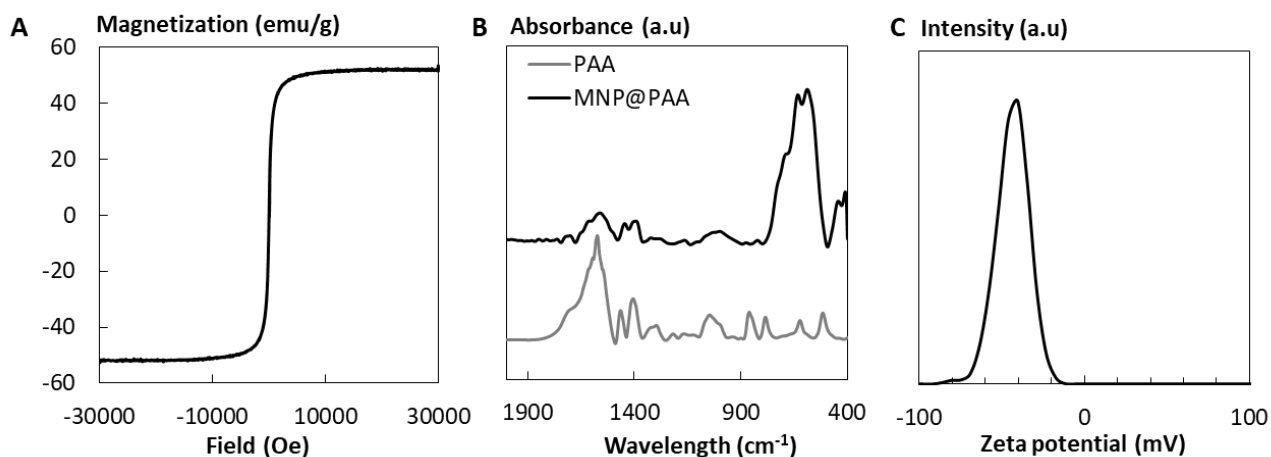
**Figure S5:** Large view TEM images of cells containing MNP 21 day after spheroid formation. MNP@PO, MNP@Cit and MNP@Caf were incubated in without FBS for 30 minutes and for 24 h for MNP@PO-PEG.



**Figure S6:** TEM images of endosomes 21 days after MNP internalization within cells for the different MNP incubated in the absence of FBS. For each coating, an endosome is represented 21 days after spheroid formation (left) with an image zoom in the black square (right) to clearly identify the ferritin spots. **A.** MNP@PO. **B.** MNP@PO-PEG. **C.** MNP@Cit. **D.** MNP@Caf



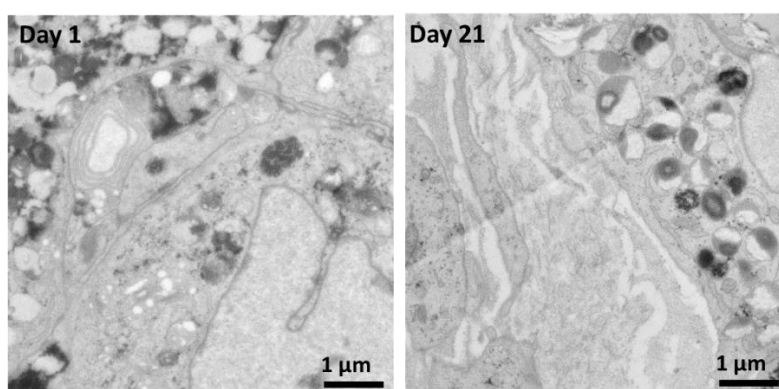
**Figure S7:** TEM images of endosomes 1 and 21 days after MNP internalization within cells for the different MNP incubated in the presence of FBS. For each coating, an endosome is represented 1 day after spheroid formation (left), then 21 days after spheroid formation (center) with an image zoom in the black square (right) to clearly identify the ferritin spots. **A.** MNP@PO. **B.** MNP@PO-PEG. **C.** MNP@Cit.



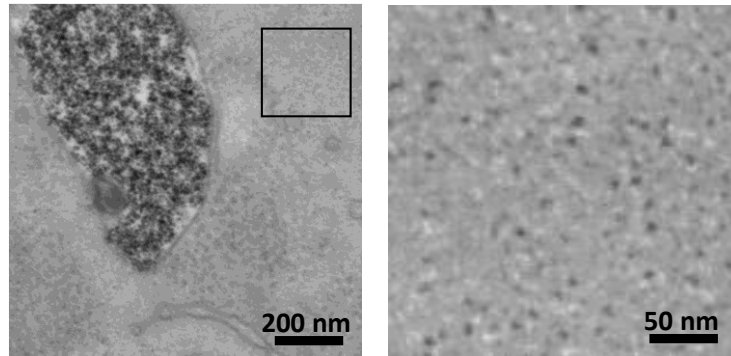
**Figure S8:** Physico-chemical characterisations of MNP@PAA. **A.** Magnetization curves at room temperature of MNP@PAA. **B.** FTIR spectra of PAA and MNP@PAA. **C.** Zeta potential curve of MNP@PAA.

Ligand	Ligands/MNP	COOH/nm <sup>2</sup>
PAA	17	0.9

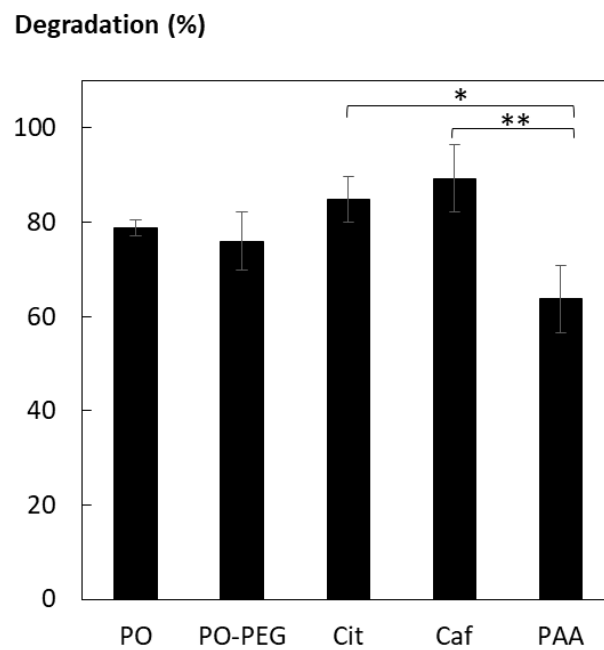
**Figure S9:** Quantification of PAA ligands and chelating (COOH) functions at the surface of each nanoparticle. Available surface for the chelating functions (considering 13 chelating functions per PAA Mw 1200 g.mol<sup>-1</sup>)



**Figure S10:** Large view TEM images of cells containing MNP@PAA 1 (left) and 21 (right) days after spheroid formation. MNP were incubated in without FBS for 30 minutes.



**Figure S11:** TEM images 21 days after MNP@PAA internalization within cells, showing still intact nanoparticles within endosomes, but with ferritin spots outside (right).



**Figure S12:** Degradation in lysosomal mimicking conditions (pH = 4.7, 20 mM citrate) for MNP@Caf, MNP@Cit, MNP@PAA, MNP@PO and MNP@PO-PEG. \*  $p < 0.05$  and \*\*  $p < 0.01$  between conditions (Student t.test) after 21 days.