## Different cell response induced by exposure to maghemite nanoparticles

Yurena Luengo,<sup>*a*,1</sup> Stefania Nardecchia,<sup>*a*,1</sup> María Puerto Morales,<sup>*a*</sup> and M. Concepción Serrano<sup>\*,*a*</sup>

## **Supporting information**

The crystalline structure of the samples was identified by X-ray powder diffraction performed in a Bruker D8 Advance powder diffractometer using Cu K $\alpha$  radiation with an energy-discriminator (Sol-X) detector. The patterns were collected between 10° and 70° in 20. The XRD spectra were indexed to an inverse spinel structure. The average crystallite size was calculated by Scherrer's equation using the half width of the (311) X-ray diffraction peak using the utilities of the automatic powder diffraction computer program (APD) from Phillips.

Particle size and shape were determined by transmission electron microscopy (TEM) using a 200 keV JEOL-2000 FXII microscope. TEM samples were prepared by placing one drop of a dilute suspension of NPs in water on a copper grid covered with a perforated carbon film and allowing the solvent to evaporate slowly at room temperature. The mean particle size and distribution were evaluated by measuring the largest internal dimension of at least 100 particles. Afterward, data were fitted to a log normal distribution obtaining the mean size ( $D_{TEM}$ ) and standard deviation ( $\sigma$ ) that were considered to be representative of the absolute error of the measurement.

Magnetic **characterization** of the samples was carried out in a vibrating sample magnetometer MagLabVSM Oxford Instrument with a maximum field of 50 kOe. Samples were measured in liquid state. To minimize interactions in the liquid samples, the dispersions were prepared at low concentrations (0.25 % wt). To avoid NP aggregation due to the VSM magnetic field, liquid samples were always measured in a

frozen state. 100  $\mu$ l of sample were placed in the sample-holder and immersed in liquid nitrogen before set it into the VSM. Temperature **was** always kept under 250 K. Hysteresis loops of the samples were measured at **room temperature** at 5 kOe min<sup>-1</sup>. The saturation magnetization was evaluated by extrapolating to infinite field the experimental data obtained in the field range where magnetization increases and it can be approximated to a 1/H law.



Figure 1-SI. Basic characterization of maghemite nanoparticles comprising: in the top, X-ray diffraction and magnetic behavior of non-coated NPs at room temperature; in the middle, TEM images showing particle size and shape before and after APS coating (left and right panels, respectively); at the bottom, size distribution of the core of non-coated NPs and aggregates of exemplary APS-coated NPs obtained from TEM images and fitted to a log normal function (left and right panels, respectively).



**Figure 2-SI. Representative** CLSM images of Saos-2 **osteoblast** control cultures at 1 and 7 days without exposure to any NPs. Cells were stained with the probes calcein and EthD-1 for monitoring cell viability over time. Scale bars represent **50**  $\mu$ m (top) and 250  $\mu$ m (bottom).