# Supporting Information

# Magnetic hyperthermia-induced drug release from ureasil-PEO-Fe<sub>2</sub>O<sub>3</sub> nanocomposites

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# **EXPERIMENTAL SECTION**

## Synthesis of monolithic ureasil-PEO hybrids (U-PEO)

In the first step, the ureasil cross-linking agent was covalently bonded to both ends of the macromer by reacting the terminal aminopropyl groups of the end-functionalized PEO (O,O'-bis(2-aminopropyl)poly(ethylene oxide) (average molecular weight (Mw) 1900 g mol<sup>-1</sup>) with (3-isocyanatopropyl)-triethoxysilane in a molar ratio of 1:2. These commercially available reagents (Fluka, Aldrich) were stirred together in anhydrous ethanol (99.8%) under reflux for 6 h at 78 °C, resulting in a hybrid precursor solution with concentration of ~0.208 mol L<sup>-1</sup>. The hybrid precursor solution obtained was stored at room temperature in a desiccator to prevent any further hydrolysis reaction. The protocol used to synthesize the precursor was a modification of the original method.1 In the second step, silanol moieties were generated, followed by condensation reactions to form ureasil cross-linking nodes. The hydrolysis of -(SiOCH<sub>2</sub>CH<sub>3</sub>)<sub>3</sub> was initialized by adding 0.075 mL of aqueous HNO3 solution (0.5 mol L<sup>-1</sup>) to 0.5 mL of precursor solution, followed by the addition of 0.295 mL of acid water (pH ~1.5) in the case of the sample without  $\gamma$ -Fe<sub>2</sub>O<sub>3</sub>. For the nanocomposite synthesis, the acid solution was replaced by an equal volume of  $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> acid colloidal suspension (pH ~1.5) at different concentrations (3.2, 0.32, 0.032, and 0.0032 wt%), resulting in samples with 5, 0.5, 0.05, and 0.005 wt% of  $\gamma$ -Fe<sub>2</sub>O<sub>3</sub>, respectively. In the case of the samples loaded with SDCF, addition was made of 3 wt% (0.005 g) sodium diclofenac (2-[(2,6-dichlorophenyl)amino] benzeneacetic acid sodium salt) (SDCF). In the final step, cylindrical monolithic xerogels approximately 20 mm in diameter, 0.5 mm in height, and weighing ~0.2 g were obtained after drying at room temperature for 48 h.

# Synthesis of magnetic nanoparticles

The Fe<sub>3</sub>O<sub>4</sub> nanoparticles were synthesized by the co-precipitation of iron ions in an alkaline medium.<sup>2</sup> An ammonia solution (500 mL, 0.7 M) was added to an aqueous solution of ferric chloride (FeCl<sub>3</sub>, 40 mL, 1 M) and ferrous chloride (FeCl<sub>2</sub>, 10 mL, 2 M) in HCl (2 M). The precipitate was removed from the solution using a permanent magnet and the supernatant was discarded. The Fe<sub>3</sub>O<sub>4</sub> was washed with distilled water, acidified with HNO<sub>3</sub> to pH ~1.5, and the resulting colloidal solution was oxidized to maghemite ( $\gamma$ -Fe<sub>2</sub>O<sub>3</sub>) by the addition of Fe(NO<sub>3</sub>)<sub>3</sub> (20 mL, 1 M). The  $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> size polydispersity was reduced by size sorting based on destabilization of the dispersion by increasing the ionic strength. The  $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> NP obtained had an average diameter of around 11 nm.

# Characterization

# Small angle X-ray scattering (SAXS)

The nanostructure of the samples was analyzed by small angle X-ray scattering (SAXS) at the SWING beamline of the SOLEIL Synchrotron. The measurements were performed at energy of 7.7 keV, with a twodimensional CCD detector located at distances of 3.5 m or 1 m from the sample in order to achieve a wide q range.

#### Transmission electron microscopy (TEM)

TEM images of the  $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> MNPs were acquired with a JEOL 100CX instrument. A droplet of diluted nanoparticles suspended in water was deposited on a carbon-coated copper grid, and the excess was removed using filter paper. Cryo-TEM images of the PEO:Fe<sub>2</sub>O<sub>3</sub>(5) nanocomposite were obtained at ICMPE-CMTR, UMR 7182 THIAIS, using a Tecnai F20 instrument (FEI) operated at 200 kV. The samples were first prepared using ultramicrotomy in order to obtain thin slices with a thickness of 100 nm. The microstructures of the nanocomposites were then observed by means of bright field images recorded at different magnifications. Size analysis of the TEM and Cryo-TEM images employed ImageJ software.<sup>3</sup>

# DSC measurements and heat capacity values

Differential scanning calorimetry (DSC) measurements were performed using a TA Instruments Model Q100 calorimeter. Disk sections (~20 mg) were removed from the monolithic hybrid xerogels and placed into 40 µL aluminium cans. Each sample was heated from -90 °C to 180 °C, at 10 °C min<sup>-1</sup>, under high purity N<sub>2</sub> gas supplied at a flow rate of 10 mL min<sup>-1</sup>. The DSC curves obtained (Figure S4a) showed the endothermic peak corresponding to fusion of semi-crystalline PEO present in the hybrid matrix, enabling information to be obtained about the crystallinity degree (D<sub>c</sub>) and melting temperature (T<sub>f</sub>). The T<sub>f</sub> can be obtained from the minimum of the endothermic peak, while the D<sub>c</sub> (Equation 1) can be calculated using the ratio between the melting enthalpy of semi-crystalline PEO in the OIH sample and the melting enthalpy of 100% crystalline PEO1900 ( $\Delta$ H<sub>p</sub> = 188.9 J g<sup>-1</sup>).<sup>4</sup>

$$D_c = \frac{\Delta H_f}{\Delta H_p} \times 100 \tag{1}$$

The heat capacity (Cp) values of the U-PEO samples (Figure S4b) were calculated from the DSC data using Equation 2:

$$Cp = \frac{K \times \Delta y_{sample - BL}}{dT / dt \times m_{sample}}$$
(2)

where K is the calorimeter sensitivity, BL is the baseline, dT/dt is the heating rate, and  $m_s$  is the sample weight. The baseline (BL) was determined under similar DSC run conditions (dT/dt = 10 °C min<sup>-1</sup>, N<sub>2</sub> atmosphere), using an empty aluminium can. The calorimeter sensitivity (K) was determined from the DSC of sapphire, used as a standard sample.

## Magnetic hyperthermia measurements

Magnetic hyperthermia measurements were performed using a commercial system (DM100 Series, nanoScale Biomagnetics Corp.). The measurements of heating induced by an AC magnetic field were performed at frequencies of 420 kHz and 280 kHz, with maximum amplitude of the alternating magnetic field (AMF) of 14.9 kA m<sup>-1</sup>. 1 mL of aqueous colloid was used, and 0.2 g of U-PEO:Fe<sub>2</sub>O<sub>3</sub>(x) was used in the case of the solid samples. The samples were placed into holders suitable for liquid and solid materials. The sample holder was then positioned in the copper coil, and the temperature was measured with a fluoro-optic fibre thermometer. Heating of the coil was avoided by circulating water kept at a thermostatically controlled temperature of 4-5 °C. Equivalent masses of liquid and solid samples without  $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> were measured in order to confirm that no heating induced by the coil occurred in the sample holder in the absence of nanoparticles.

#### In vitro drug release

The release of SDCF was evaluated using ~0.2 g of the nanocomposite and 50 mL of phosphate buffer solution (pH 7.2). Five experiments were performed, using buffer temperatures of 0, 25, 37, and 50 °C, and one experiment started at 37 °C and with application of an AMF (14.9 kA m<sup>-1</sup>, 420 kHz). The amount of SDCF released was determined by UV-Vis absorbance at 276 nm, using an Agilent Technologies Cary 60 dual beam spectrophotometer. Aliquots were collected from the different systems at predetermined time intervals.

# RESULTS



**Fig. S1** Temperature increases of samples containing the indicated amouts of  $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> under applied external magnetic fields (14.9 kA m<sup>-1</sup>; 420 or 280 kHz frequency): a) and c)  $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> aqueous colloids; b) and d) U-PEO:Fe<sub>2</sub>O<sub>3</sub>:SDCF nanocomposites.



**Fig. S2** Linear temperature increases during the first 30 s of application of an external AMF of 14.9 kA m<sup>-1</sup> and frequency of 420 kHz, used for calculation of the SLP values (see Table S1) of a) the  $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> aqueous colloids, and b) the U-PEO:Fe<sub>2</sub>O<sub>3</sub>:SDCF nanocomposites.



**Fig. S3** Experimental conditions used for the assays of drug release induced by application of an AMF. Cumulative drug release (black), AMF magnetic field amplitude (blue), buffer solution temperature (red).



**Fig. S4** a) DSC curves for the U-PEO samples, showing the melting fusion temperatures ( $T_f$ ) and the crystallinity degrees ( $D_c$ ). b) Temperature dependence of the heat capacity ( $C_p$ ) of U-PEO:Fe<sub>2</sub>O<sub>3</sub>(5):SDCF.

	Samples	wt% γ-Fe <sub>2</sub> O <sub>3</sub>	$(dT/dt)_{max}$ (°C s <sup>-1</sup> )	<b>SLP</b> (W g <sup>-1</sup> )
		0.032	0.00845	110.38
	γ-Fe <sub>2</sub> O <sub>3</sub>	0.32	0.0809	105.67
		3.2	0.746	97.45
		0.5	0.0298	13.80
	0-r E0.re <sub>2</sub> 0 <sub>3</sub> (X).5DCr	5	0.277	12.80
0	01 000 0	1 00		

Table S1: Experimental data determined from hyperthermia measurements.

\* $C_{p \text{ (water, 25 °C)}}$ : 4.18 J  $g^{-1}$  °C<sup>-1</sup>;  $C_{p \text{ (U-PEO, 25 °C)}}$ : 2.12 J  $g^{-1}$  °C<sup>-1</sup>.

# References

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