Electronic Supporting Information for:

Tuning the composition of biocompatible Gd nanohydrogels to achieve hypersensitive dual T_1/T_2 MRI contrast agents

Dr. M. Callewaert et al.

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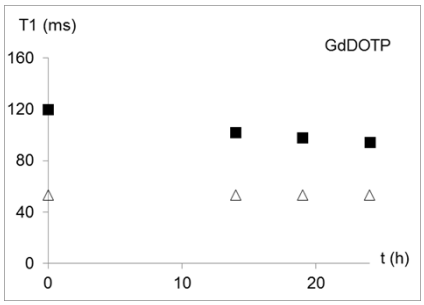
a)



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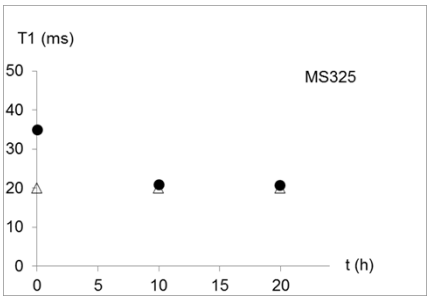
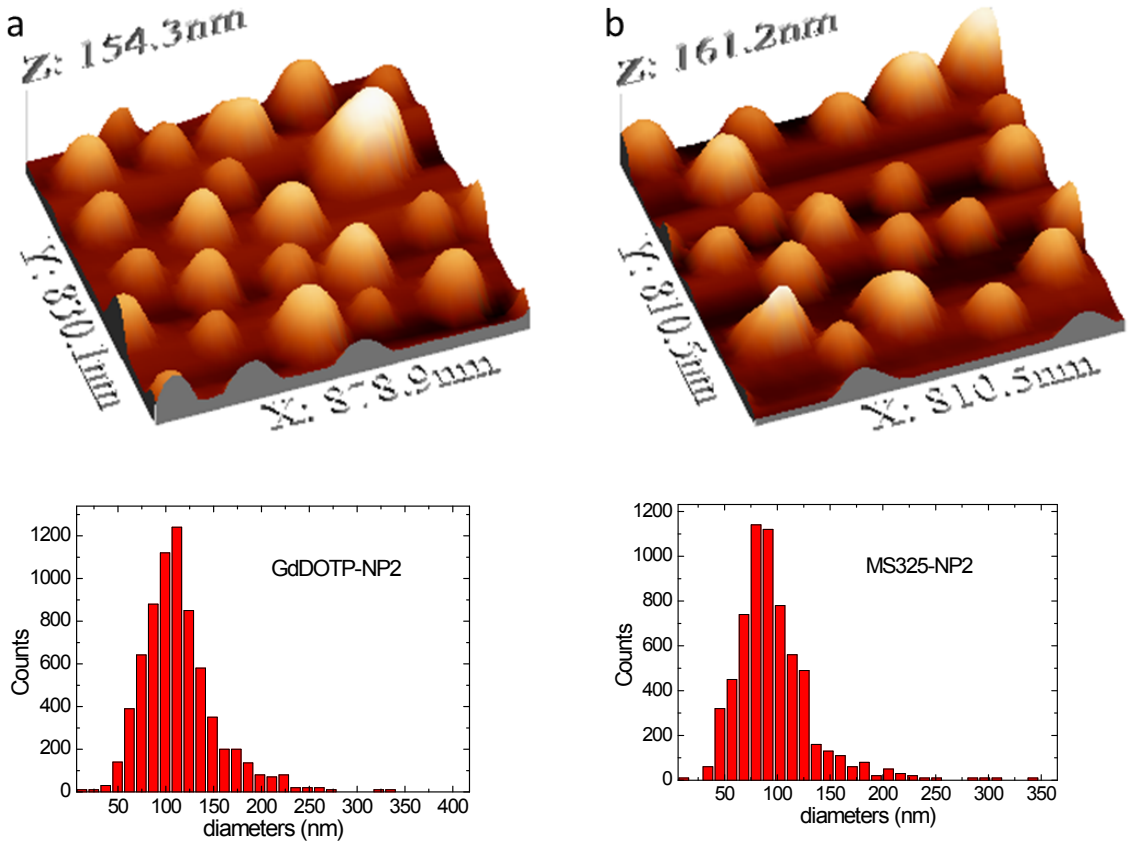
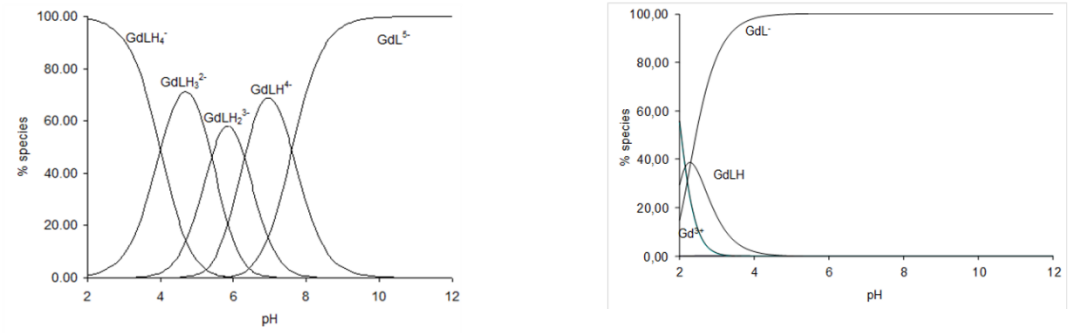


ESI-1. Evolution of T_1 relaxation rates for a- GdDOTP (■: 1.5×10^{-3}M) and b- MS325 (●: 4×10^{-3}M) in citric acid solution (10% v/v). T_1 relaxation rates of $GdCl_3$ (❒) in citric acid solution are given for comparison (concentrations identical to the ones of GdDOTP or MS325 respectively).



ESI-2. Size distribution of a- GdDOTP⊂NP2 and b- MS325⊂NP2 from AFM images

a) b)

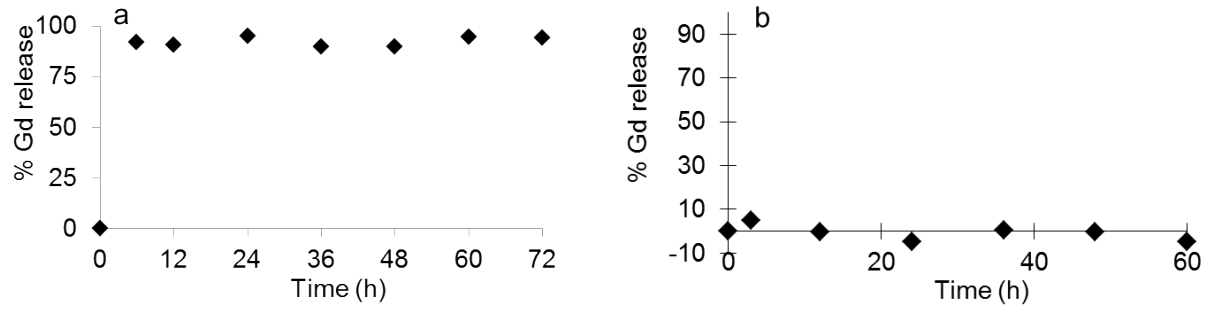


ESI-3. Speciation diagrams of a- GdDOTP[a] , b- GdDOTA[b] as a function of pH

References:

a. A. D. Sherry, J. Ren, J. Huskens, E. Brucher, E. Toth, C. F. C. G. Geraldes, M. M. C. A. Castro and W. P. Cacheris, *Inorg. Chem.*, 1996, **35**, 4604.

b. E. T. Clarke and A. E. Martell, *Inorg. Chim. Acta,* 1999, **190**, 37.



ESI-4. Release profiles at 37 °C of MS325⊂NP2 in a- phosphate buffer, b- simulated plasma

ESI-5. Relaxivity calculations

The reported relaxivities corresponded to the contribution of Gd centers buried inside the nanoparticles. To obtain these values:

- T_1 (and T_2) relaxation times of GdNPs were measured.

- $1/T_{1para}$ (and $1/T_{2para}$) values were calculated and the diamagnetic contribution was subtracted to these values (the diamagnetic contribution being obtained by measurements T_1 and T_2 relaxation times of unloaded NP). This gives R_1^{para} (and R_2^{para}) relaxation rates for the nanosuspensions. The values obtained at 20 MHz and 37°C for nanosuspensions given in Table 2 are the following:

	$[Gd]_{NPs}$ mM	$[Gd]_{free}$ mM	R_1^{para}	R_2^{para}
GdDOTP⊂NP1	0.397	0.022	16.57	19.01
GdDOTP⊂NP2	0.14	0.07	14.01	15.64
MS325⊂NP2	1.745	1.274	99.93	114.95

For each nanosuspension, the amount of Gd entrapped in the nanoparticles and the amount that remains free in the nanosuspension (or at least weakly bound to the nanoparticles) were systematically determined (details given in the experimental section – determination of the Gd loading by ICP OES). Therefore, to obtain r_1 values associated to GdNPs, the contribution of free chelates present in the nanosuspension must be removed according to:

r_1 (GdCA⊂NP) = $(R_1^{para}-([Gd]_{free}*r_1\ (GdCA_{\ free}))/[Gd]_{NPs}$ ($mM^{-1}\ s^{-1}$) that gives:

r_1 (GdDOTP⊂NP1) = (16.57-(0.022*4.2))/0.397 = 41.5 $mM^{-1}\ s^{-1}$

r_1 (GdDOTP⊂NP2) = (14.01-(0.07*4.2))/0.14 = 97.97 ~ 98 $mM^{-1}\ s^{-1}$

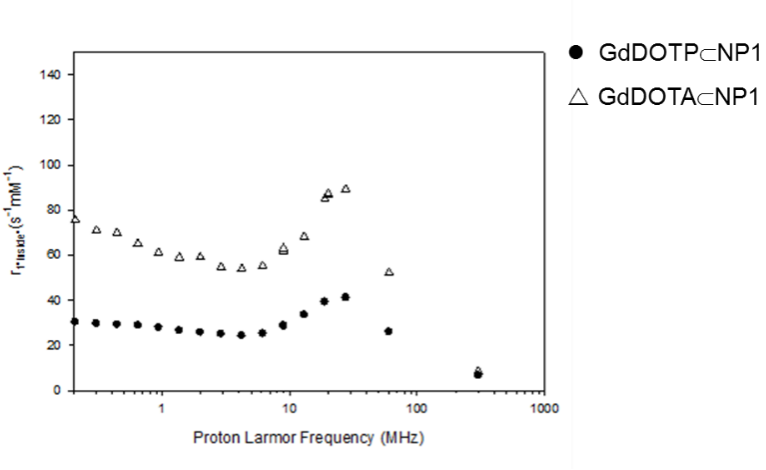
r_1 (MS325⊂NP2) = (99.93-(1.274*5.9))/1.745 = 52.9 $mM^{-1}\ s^{-1}$

with r_1 (GdDOTP) = 4.2 $mM^{-1}\ s^{-1}$ and r_1 (MS325) = 5.9 $mM^{-1}\ s^{-1}$

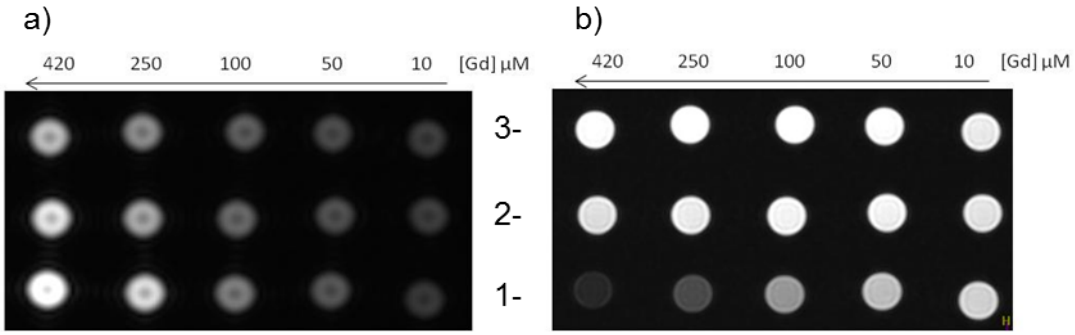
Similar calculations were done for r_2 values in Table 2. Similar treatments were applied to the NMRD profiles given in Figure 3.

For each nanosuspension the contribution of free chelates to the overall relaxivity (20MHz, 37°C) is given in the following table:

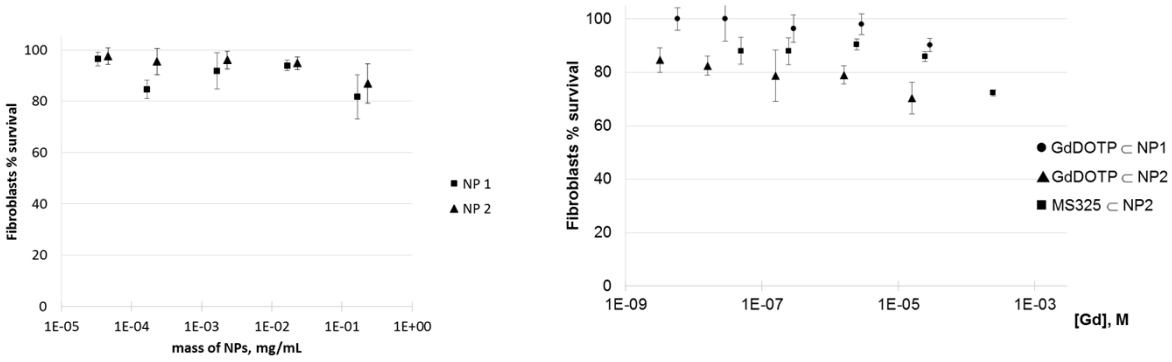
	$[Gd]_{free}$ mM	R_1^{para}	R_1^{para} (Gd_{free})/ R_1^{para}
GdDOTP⊂NP1	0.022	16.57	(0.022*4.2)/16.57 = 0.56%
GdDOTP⊂NP2	0.07	14.01	(0.07*4.2)/14.01 = 2.1%
MS325⊂NP2	1.274	99.93	(1.274*5.9)/99.93 = 7.5%



ESI-6. Comparison of GdDOTP⊂NP1 and GdDOTA⊂NP1 NMRD relaxivity profiles



ESI-7. a- T_1-weighted images of 1) GdDOTP⊂NPs – protocol 1, 2) $Na_5GdDOTP$ and 3) DOTAREM® as controls. b- T_2-weighted images for the same solutions as in a).
Samples imaged at 3 T, 37°C, and standard spin echo (SE) sequence.



ESI-8. Percentage of primary fibroblast cells survival a- in presence of unloaded NPs and b- in presence of Gd⊂NPs, as a function of increasing concentrations. Results obtained after 48 h of incubation.