Supporting Information

Stimuli-Responsive Biodegradable and Gadolinium-Based Poly[*N*-(2-hydroxypropyl)methacrylamide] Copolymers: Potential as Targeting and Safe Magnetic Resonance Imaging Probes

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1. Materials and Methods

The MWs and polydispersity (PDI) of the copolymer were tested via size-exclusion chromatography (SEC) on a Superose 6 HR10/30 column and on an ÄKTA/FPLC system (GE Healthcare). Sodium acetate buffer/acetonitrile (7:3, pH 6.5) was used as mobile phase with a corresponding flow rate of 0.4 mL/min. The copolymers were purified by SEC via a Superose 6 HR10/30 column, while the mobile phase was sodium acetate buffer/acetonitrile (7: 3, pH 6.2), and the flow rate was 2.5 mL/min, and the temperature was 4 ^{o}C. NMR spectroscopy data was obtained using a 400 MHz Bruker Advanced Spectrometer, and chemical shifts were reported in ppm on the δ scale.

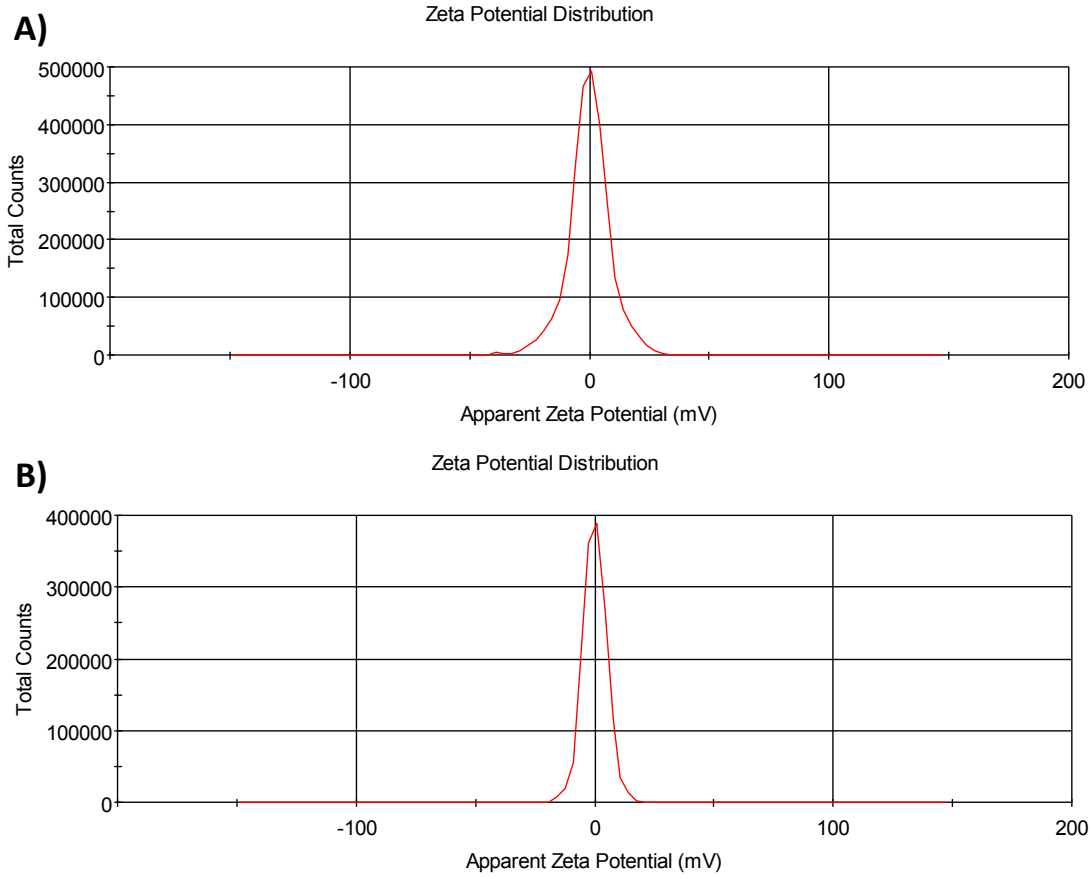

Figure S1. Zeta potential of pHPMA-DOTA-Gd (-0.13 mV) and pHPMA-DOTA-Gd-cRGD (0.04 mV) based nanoscale system.

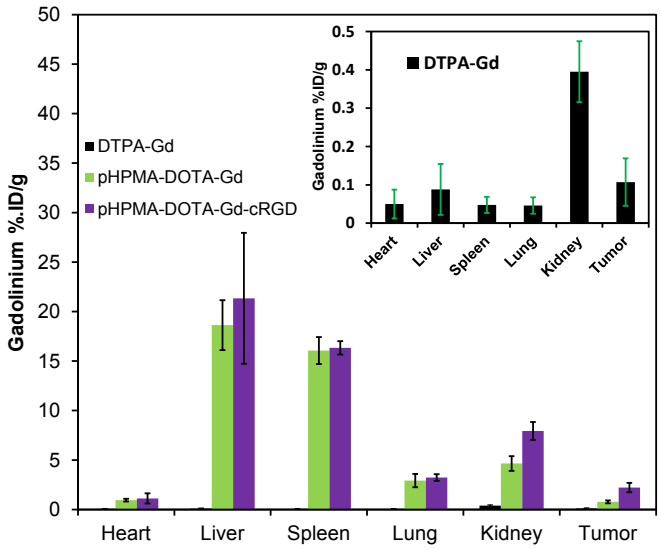

Figure S2. Quantitative analysis of gadolinium content of the different tissues and tumors from the nude mice with U87 tumors. The data was obtained at 24 h post-treatment with DTPA-Gd, pHPMA-DOTA-Gd and pHPMA-DOTA-Gd-cRGD [0.08 mmol Gd(III)/kg mice], respectively. The tumor size was about 3 mm in diameter. (*p<0.01 versus DTPA-Gd) (n=7/ group).

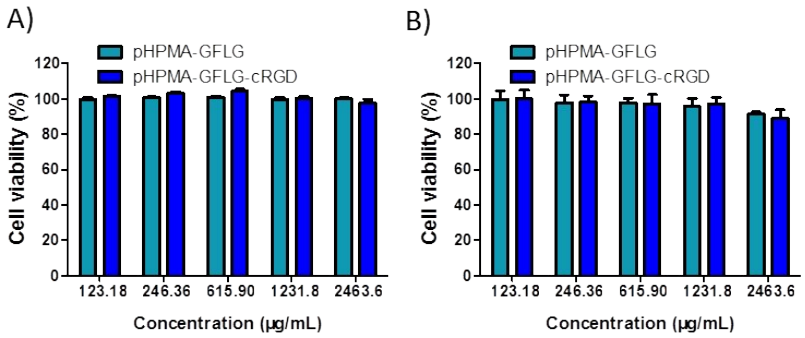

Figure S3. Cytotoxicity of two polymers without Gd(III) in L02 (A) and U87 (B) cell lines.

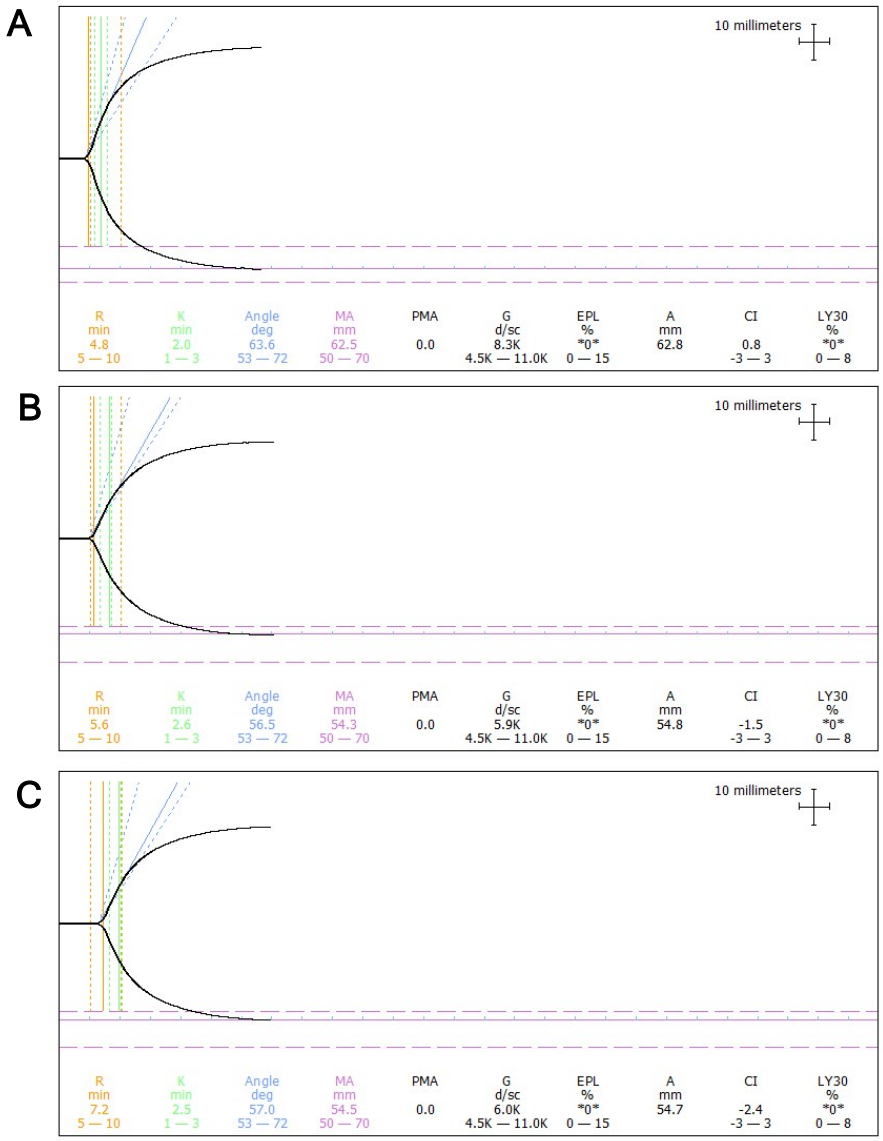

Figure S4. Representative thromboelastography (TEG) traces of whole blood clotting in the presence of PBS (A), pHPMA-DOTA-Gd (B) and pHPMA-DOTA-Gd-cRGD (C), respectively.

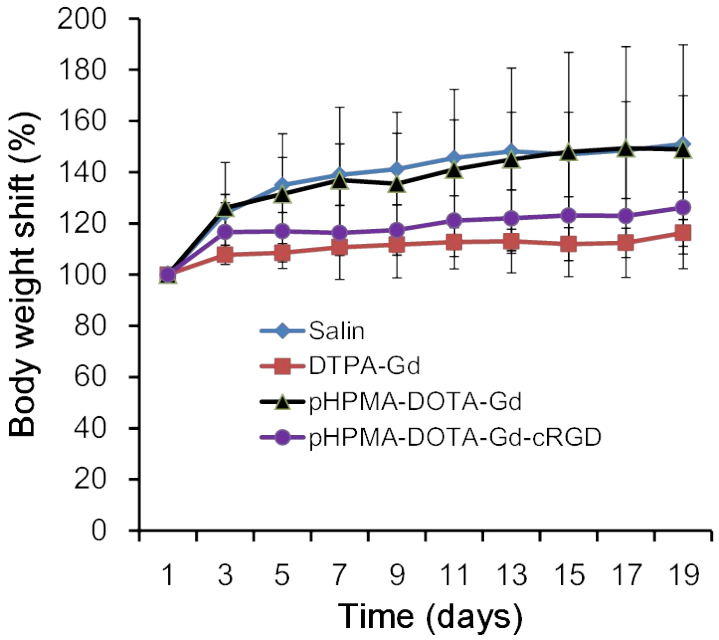

Figure S5. Normal animal body weight shifts post-injections of saline, clinical agent DTPA-Gd, pHPMA-DOTA-Gd and pHPMA-DOTA-Gd-cRGD conjugates up to day 19 (n=7, per group).

Table S1. The characterization of the pHPMA-DOTA-Gd and pHPMA-DOTA-Gd-cRGD products.

Comp.	MW (kDa)	PDI	Content of thiol group (mmol/g product)	Content of cRGDyK (weight%)	gadolinium content(weight%)
pHPMA-DOTA-Gd	85	1.18	0.66	-	6.5%
pHPMA-DOTA-Gd-cRGD	94	1.20	0.10	~22%	4.0%

Table S2. The MW and PDI of the degraded products after incubation of conjugates (3 mg/mL) with 2.4 μM cathepsin B in McIlvaine's buffer at 37 °C (pH 5.4).

Comp.	0 h	2 h	5 h	8 h	12 h
pHPMA-DOTA-Gd	85 kDa, PDI 1.18	78 kDa, PDI 2.94	49 kDa, PDI 1.41	43 kDa, PDI 1.03	43 kDa, PDI 1.02
pHPMA-DOTA-Gd-cRGD	94 kDa, PDI 1.20	76 kDa, PDI 2.64	54 kDa, PDI 1.45	43 kDa, PDI 1.15	44 kDa, PDI 1.12

Table S3. The MW and PDI of the degraded products after incubation of conjugates in PBS at 37 °C (pH 7.4).

Comp.	0 h	2 h	5 h	8 h	12 h
pHPMA-DOTA-Gd	85 kDa, PDI 1.16	85 kDa, PDI 1.17	84 kDa, PDI 1.19	84 kDa, PDI 1.19	83 kDa, PDI 1.16
pHPMA-DOTA-Gd-cRGD	94 kDa, PDI 1.20	94 kDa, PDI 1.22	93 kDa, PDI 1.22	93 kDa, PDI 1.21	93 kDa, PDI 1.22

PBS: phosphate buffered saline

Table S4. Clotting kinetics values of human whole blood mixed with the two conjugates.

	R (min)	*K* (min)	*α* (deg)	*MA* (mm)
Normal range	5−10	1−3	53−72	50−70
PBS control	4.8	2.0	63.5	62.5
pHPMA-DOTA-Gd	5.6	2.6	56.5	54.3
pHPMA-DOTA-Gd-cRGD	7.2	2.5	57.0	54.5