Supporting Information

Title: Red blood cell membrane-coated upconversion nanoparticles for pretargeted multimodality imaging of triple-negative breast cancer

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Synthesizing the pristine $NaGdF_4$: Yb, Tm nanoparticles

$GdCl_3 \cdot 6H_2O$ (0.80 mmol), $YbCl_3 \cdot 6H_2O$ (0.18 mmol), and $TmCl_3 \cdot 6H_2O$ (0.02 mmol) will be mixed with 14 mL of OA and 16 mL of ODE in a 100 mL flask. The resultant mixture will be then heated to 150 ℃ under nitrogen protection to form a homogeneous solution. After the solution will be cooled to 50 ℃, 10 mL of a methanol solution containing NaOH (2.5 mmol)

and NH4F (4 mmol) will be slowly introduced, and the reaction system will be then kept under stirring at 50 ℃ for 30 min. Subsequently, methanol in the system will be removed by keeping the reaction system at 100 ℃ for 10 min under vacuum. Under atmospheric pressure, the finally formed reaction mixture will be heated to 300 ℃. The reactions will be allowed for 1 h under nitrogen protection and then terminated by cooling the reaction mixture to room temperature. The resultant nanoparticles will be precipitated by ethanol, collected by centrifugation, washed with ethanol several times, and finally redispersed in THF or cyclohexane for further experiments.

Physicochemical characterization of RBC membranes

The preparation of RBC-ghosts was monitored by using an inverted microscope (IX73, Olympus, Japan) with a colorful charge-coupled device (CCD; DP72, Olympus, Japan), which displayed an intact cellular shape but a changed intracellular component. The preparation of RBC membrane-derived vesicles was monitored by using a dynamic light scatter (DLS; Nano-Zen 3600, Malvern Instruments, UK) to measure the hydrodynamic diameters and zeta potentials of products after each step.

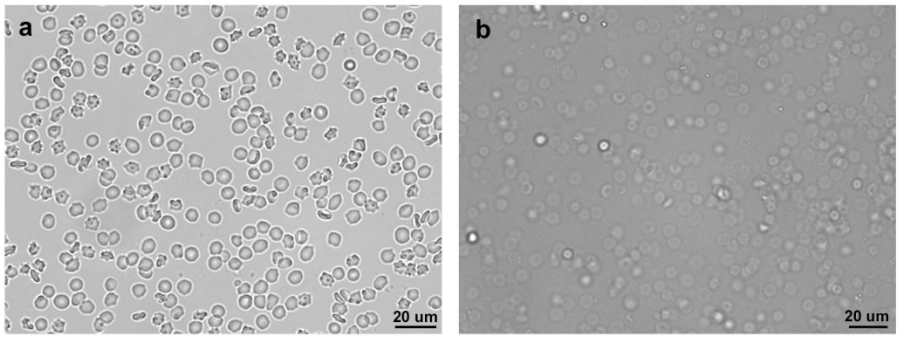

Figure S1. Bright field images of mice RBCs before (A) and after (B) the hypotonic treatment.

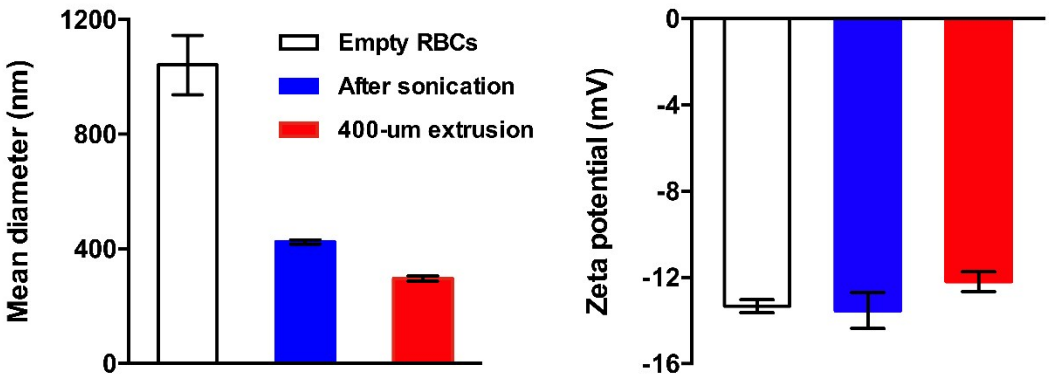

Figure S2. Average hydrodynamic diameter (left) and zeta potential (right) of the RBC membrane- derived vesicles following RBC-ghosts derivation (empty RBCs), sonication for 5 min, and extrusion through 400-nm polycarbonate porous membranes.

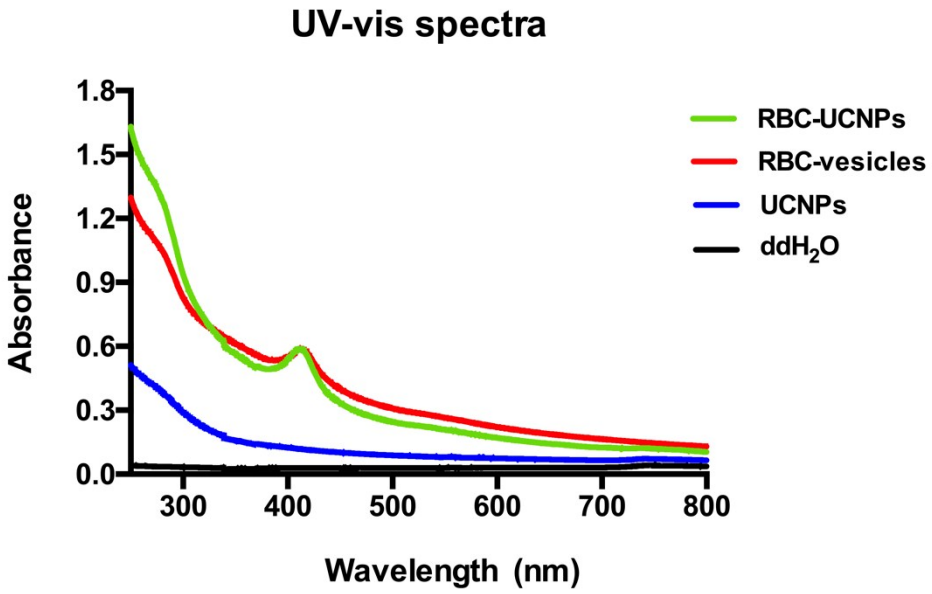

Figure S3. UV-vis spectra of RBC-vesicles, UCNPs and RBC-UCNPs.

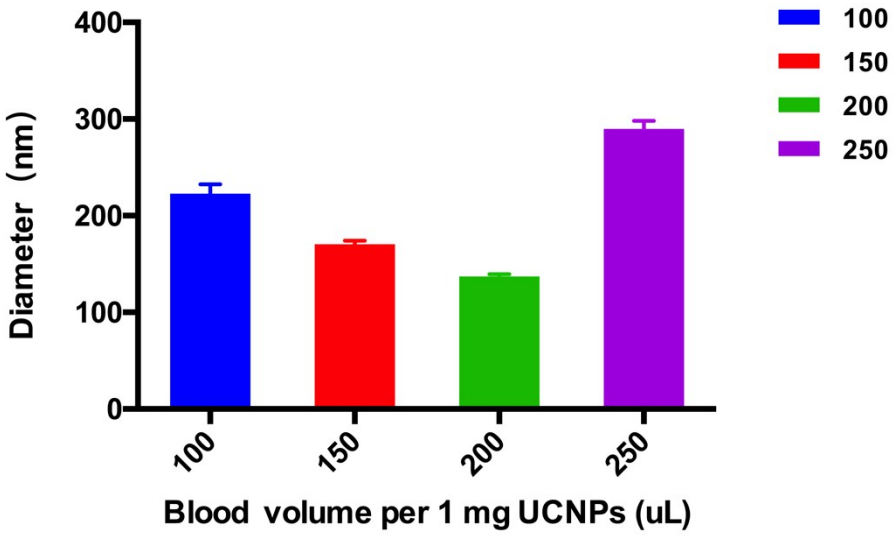

Figure S4. Hydrodynamic size of RBC-UCNPs at different RBC membrane to nanoparticle ratios right after synthesis in 1× PBS.

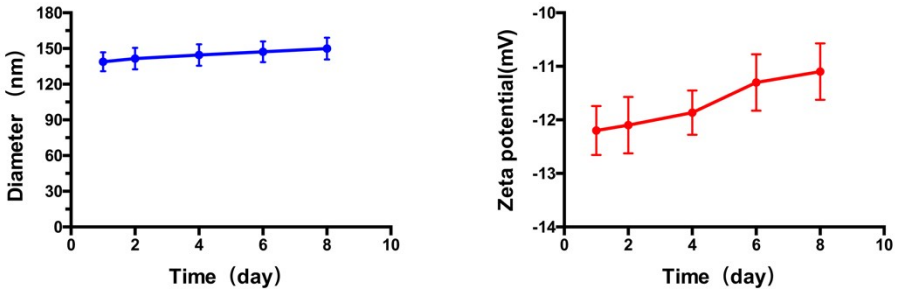

Figure S5. Diameter (left) and zeta potential (right) of RBC-UCNPs in 1× PBS over 7 days.

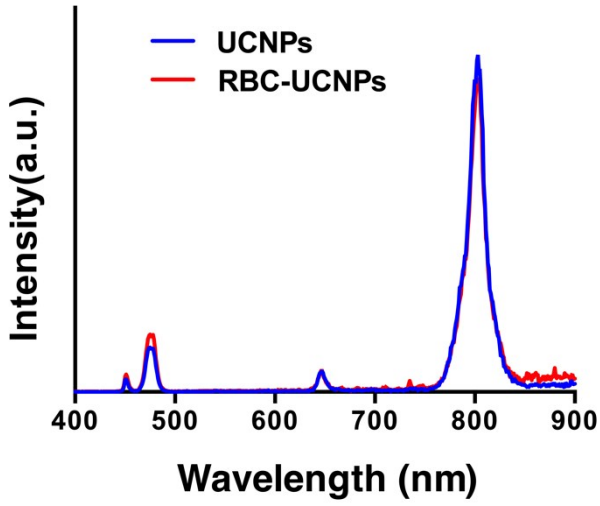

Figure S6. Upconversion fluorescence emission spectrum of UCNPs and RBC-UCNPs exposed to a 980-nm laser.

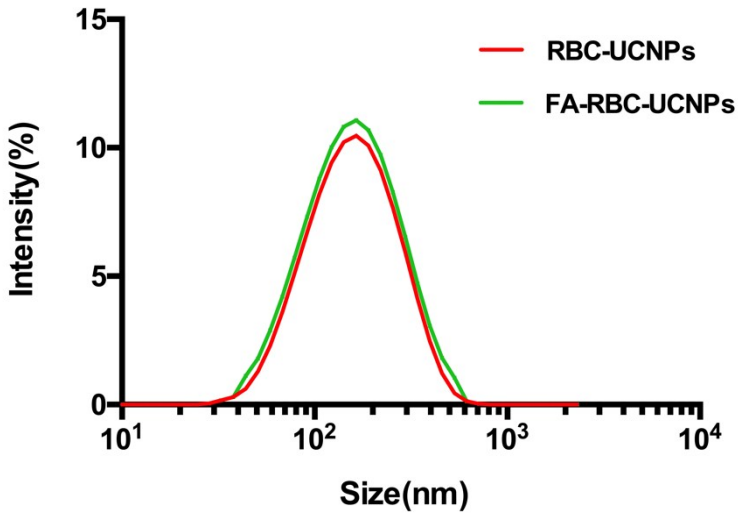

Figure S7. Size intensity curves of unmodified and modified RBC-UCNPs with DSPE-PEG-FA.

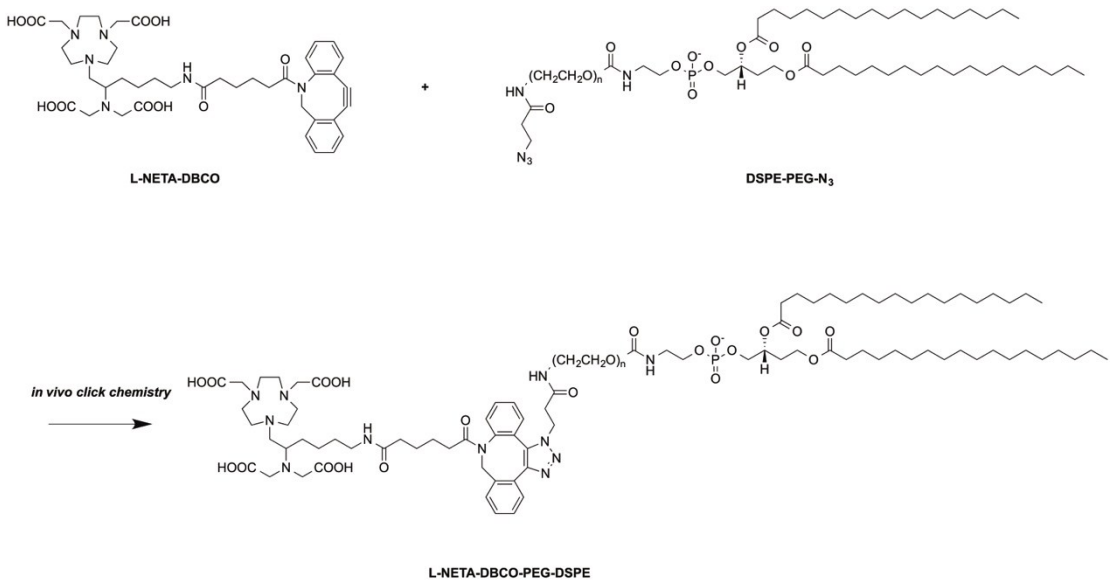

Figure S8. The scheme of in vivo strain-promoted azide-alkyne cycloaddition (SPAAC) reaction.

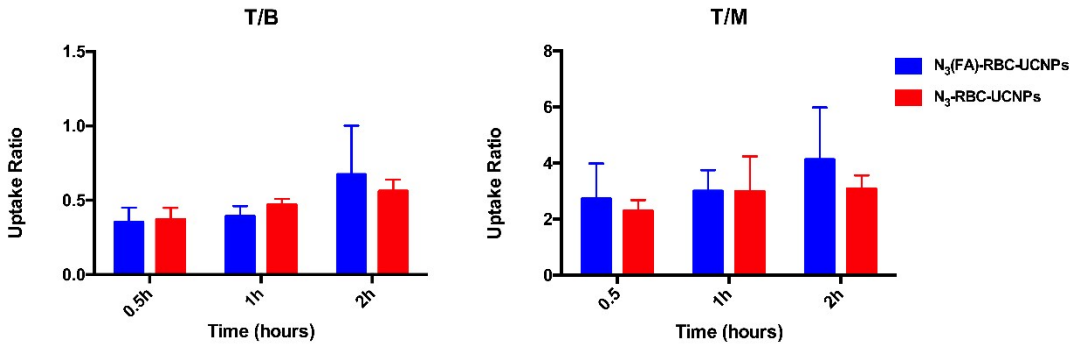

Figure S9. The uptake ratio of tumor to blood (T/B, left) and tumor to muscle (T/M, right) at different time points after injection of N_3(FA)-RBC-UCNPs and N_3-RBC-UCNPs.

	Unit	Control	UCNPs	RBC-vesicles	RBC-UCNPs
WBC	10^9/L	5.33 ± 1.87	5.40 ± 1.65	5.60 ± 1.21	5.13 ± 2.08
RBC	10^{12}/L	11.13 ±1.86	11.05 ±1.65	10.94 ± 1.30	10.91 ± 0.90
HGB	g/L	185.00 ± 33.50	173.20 ± 28.46	174.00 ± 21.32	179.50 ± 18.75
HCT	CV%	61.12 ± 10.08	57.96 ± 8.82	59.70 ± 6.92	60.77 ± 5.70
MCV	fL	55.00 ± 0.94	53.00 ± 2.13	54.63 ± 1.17	55.72 ± 1.70
MCH	pg	16.55 ± 0.39	15.58 ± 0.31	15.85 ± 0.46	16.37 ± 0.60
MCHC	g/L	301.33 ± 8.71	295.50 ± 9.59	290.83 ± 4.14	294.67 ± 6.65
PLT	10^9/L	927.40 ± 375.60	865.00 ± 284.64	997.40 ± 257.84	742.67 ± 100.89
PDW	CV%	16.75 ± 0.58	16.70 ± 0.41	16.55 ± 0.49	17.25 ± 0.56

Table S1. Complete blood panel analysis. Abbreviations as follows: WBC: white blood cell, RBC: red blood cell, HGB: hemoglobin, HCT: hematocrit, MCV: mean corpuscular volume, MCH: mean corpuscular hemoglobin, MCHC: mean corpuscular hemoglobin concentration, PLT: platelets, RDW: red blood cell distribution width.