Supporting Information

Critical Features for Mesoporous Silica Nanoparticles Encapsulated into Erythrocytes

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Figure S1

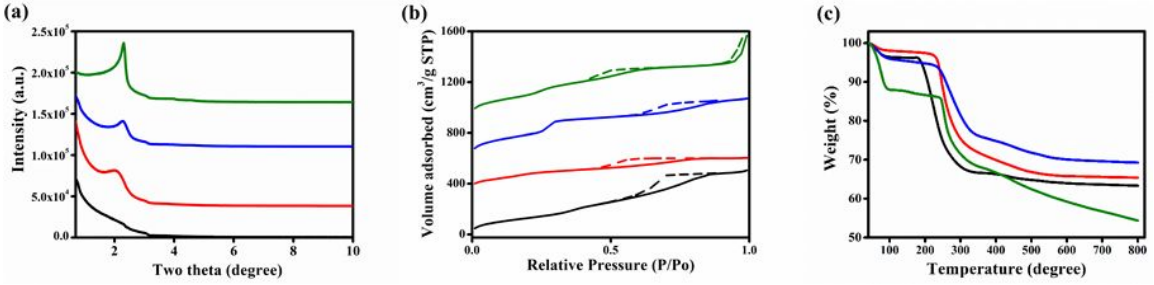

Figure S1. (a) X-ray diffraction (XRD) patterns, (b) Linear plot of the nitrogen sorption isotherms and (c) Thermogravimetric analysis (TGA) of RMSN-10 (black), RMSN-PEG-25 (red), RMSN-PEG-50 (blue) and RMSN-PEG-200 (green).

Table S1 Total surface area, interplanar spacing, pore size and weight loss of RMSN-PEG with various sizes.

Samples	d_{100} (nm)	S_{BET} (m^2/g)	D_{BJH} (nm)	TGA results for 150-800℃ (wt%)
Bare-RMSN-10	4.87	610.49	2.72	32.8%
RMSN-PEG-25	5.27	470.25	1.62	32.3%
RMSN-PEG-50	4.83	887.02	2.07	26.0%
RMSN-PEG-200	4.83	793.73	1.93	33.0%

d_{100}: interplanar spacing calculating from Bragg formulation. S_{BET}: surface area calculated from data using BET equation. D_{BJH}: pore diameter assigned from the maximum on the BJH pore size distribution. wt%: normalized weight loss from TGA analysis. Bare means particles without PEGylation.

Figure S2

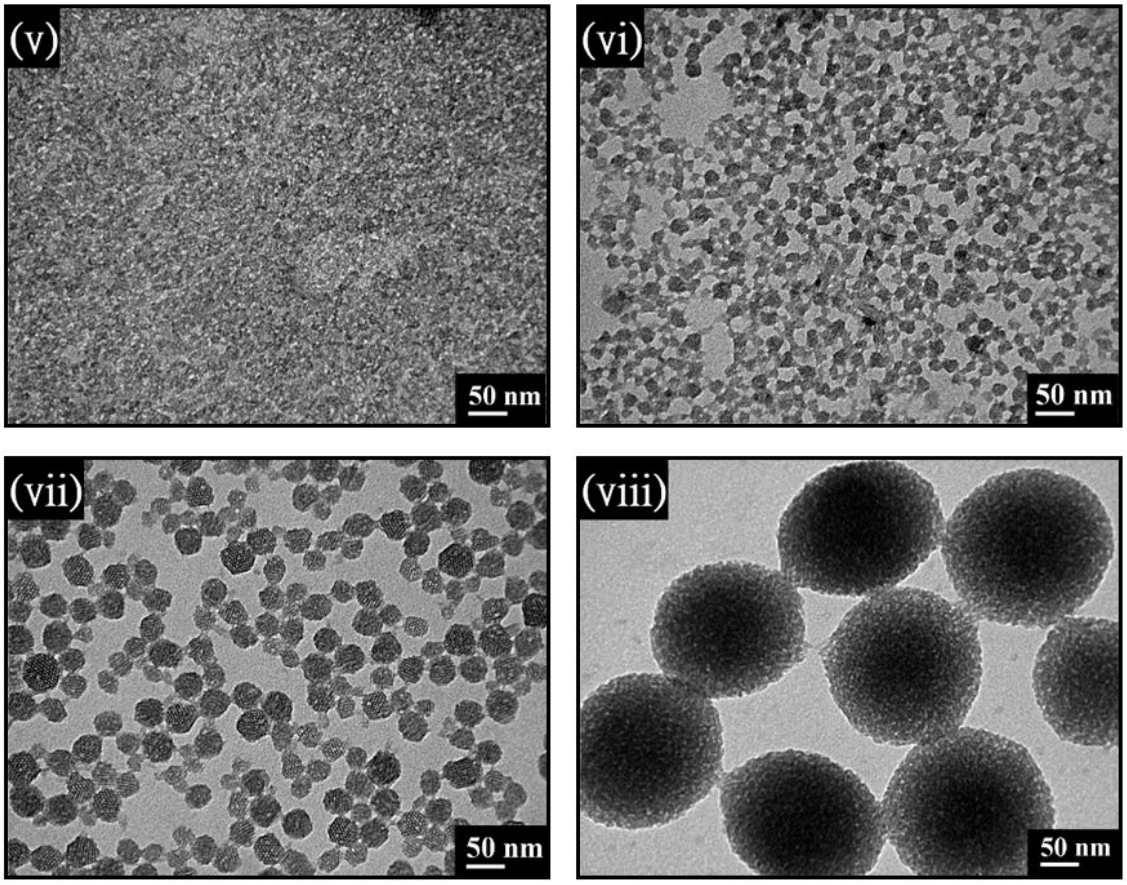

Figure S2. TEM imaging of MSN-PEG with various diameters: (v) MSN-PEG-10, (vi) MSN-PEG-25, (vii) MSN-PEG-50 and (viii) MSN-PEG-200.

Table S2 Hydrodynamic size distribution and zeta potential of MSN-PEG, RMSN-PEG and surface-modified MSN with various sizes in different solvent (including of DI water, PBS and HEPES buffer).

Samples	D_h / PdI in DI water (nm)	D_h / PdI in PBS (nm)	D_h / PdI in HEPES buffer (nm)	ζ in HEPES buffer (mV)
Bare-MSN-10	N/A	N/A	-	-7.1 ± 0.8
MSN-PEG-10	17.3 / 0.52	20.9 / 0.38	-	-1.6 ± 0.7
MSN-PEG-25	29.0 / 0.21	32.5 / 0.23	-	-1.0 ± 2.1
MSN-PEG-50	50.1 / 0.22	48.7 / 0.06	-	-1.4 ± 0.3
MSN-PEG-200	205.0 / 0.04	215.8 / 0.01	-	-2.5 ± 0.2
Bare-RMSN-10	N/A	N/A	N/A	-12.1 ± 2.0
RMSN-PEG-10	27.2 / 0.35	18.5 / 0.29	26.4 / 0.54 *	-1.6 ± 1.2
RMSN-PEG-25	34.4 / 0.23	32.7 / 0.15	38.8 / 0.20	-1.3 ± 0.5
RMSN-PEG-50	58.8 / 0.13	56.3 / 0.09	49.6 / 0.08	-1.3 ± 0.3
RMSN-PEG-200	192.5 / 0.03	206.4 / 0.03	215.2 / 0.06	-2.7 ± 0.02
RMSN-PEG-TA-10	38.4 / 0.66	23.8 / 0.42	54.3 / 0.41 *	0.4 ± 0.07
RMSN-PEG-PEI-10	31.2 / 0.31	27.2 / 0.30	47.9 / 0.20 *	0.7 ± 0.46

D_h = Z-average, harmonic intensity averaged particle diameter, PdI = polydispersity index. ζ= Zeta potential. * means multi-peak distribution. Bare means particles without PEGylation. R represents RITC dye reagent. TA = TA-silane, PEI = PEI-silane. N/A = not applicable.

Figure S3

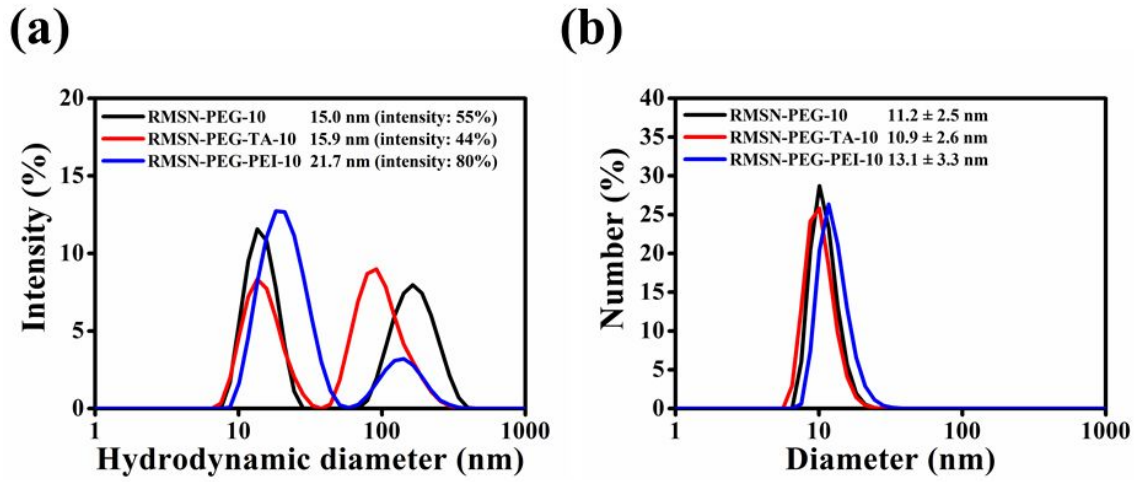

Figure S3. (a) Hydrodynamic diameter intensity distributions and (b) number distribution of RMSN-PEG-10 (black), RMSN-PEG-TA-10 (red) and RMSN-PEG-PEI-10 (blue) in HEPES buffer.

Figure S4

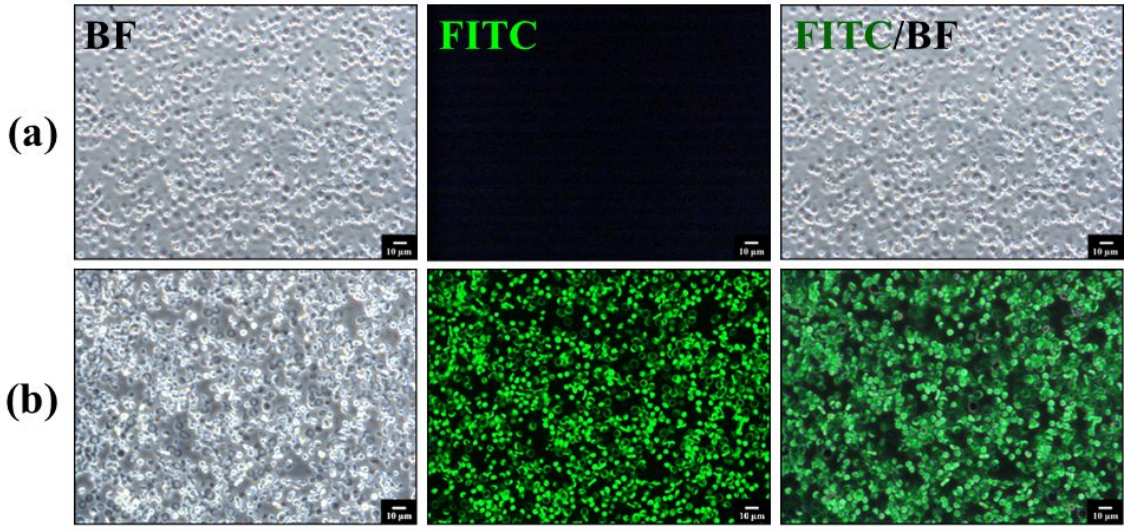

Figure S4. Optical and fluorescent imaging of (a) native RBCs and (b) the dextran@RBCs. BF: bright field, FITC: detecting fluorescent RBCs and FITC/BF: merge BF and FITC imaging. All of the images were taken at 40× original magnification.

Figure S5

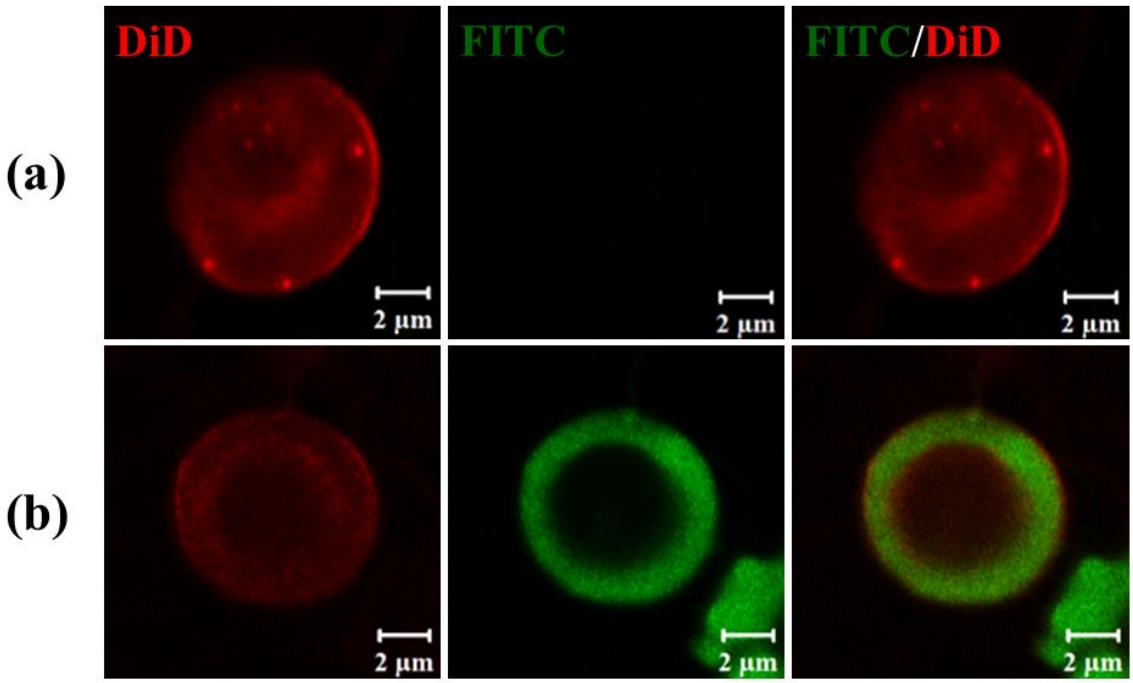

Figure S5. Confocal imaging of (a) native RBCs and (b) dextran@RBCs with DiD dye reagent. DiD: fluroscent membrane of RBCs, FITC: detecting FITC dye reagent and FITC/DiD: merge FITC and DiD imaging.

Figure S6

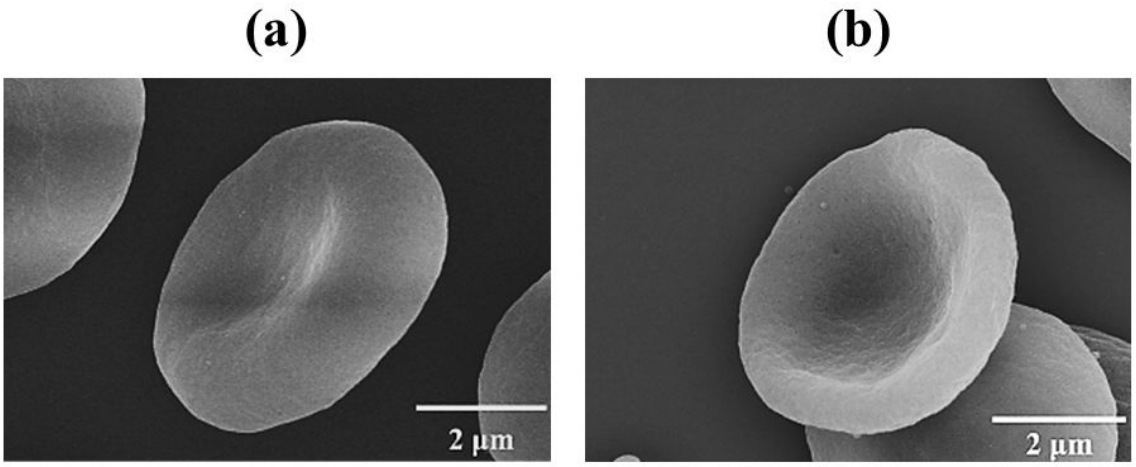

Figure S6. Scanning electron microscopy (SEM) imaging of (a) native RBCs and (b) the dextran@RBCs.

Figure S7

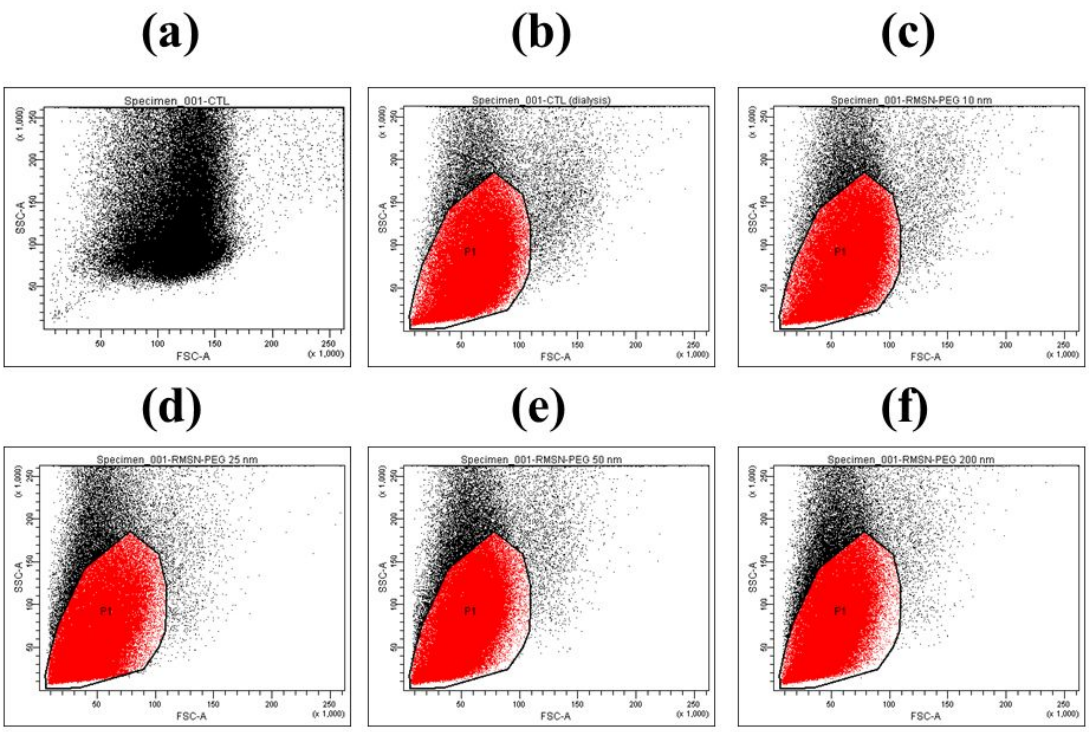

Figure S7. The FSC (X-axis) and SSC (Y-axis) parameters on flow cytometry determine the relative size and internal complexity of RBCs, which were processed via hypotonic dialysis based method. (a) Native RBCs, (b) dialyzed RBCs and (c-f) the engineered RMSN-RBCs. P1 meant the red area acted as control of the flow cytometry analysis for the engineered RMSN-RBCs shown in Fig 7a.

Table S3 Cell integrity evaluation of the engineered RMSN-RBCs measured with an automated hemocytometer.

Samples	MCV (fl)	MCH (pg)	MCHC (g/dl)
Native RBCs	101.5 ± 0.6	28.7 ± 1.8	28.2 ± 1.8
Dialyzed RBCs	53.8 ± 4.5	10.7 ± 0.9	20.0 ± 3.2
RMSN-RBC-10	53.0 ± 3.0	10.9 ± 0.9	20.5 ± 1.0
RMSN-RBC-25	53.1 ± 1.0	11.5 ± 0.9	21.6 ± 2.1
RMSN-RBC-50	51.9 ± 1.4	9.6 ± 0.2	18.5 ± 0.9
RMSN-RBC-200	43.7 ± 0.6	10.9 ± 0.2	24.9 ± 0.2

The results are means of three experiments ± standard deviation. MCV, MCH and MCHC were measured with an automated hemocytometer. MCH: Mean hemoglobin concentration; MCHC: Mean corpuscular hemoglobin concentration; MCV: Mean corpuscular volume; RBC: Red blood cell; Native RBCs: Not dialyzed red blood cells; Dialyzed RBCs: Red blood cell via hypotonic dialysis based method; RMSN-RBC-(10 to 200): the engineered RMSN-RBCs with various size of RMSN.

Table S4 Si content determined by ICP-MS analysis. The Delta Si values correspond to the values obtained by subtracting the Si content of native RBCs from the Si content of the engineered RMSN-RBCs (10^8 cells).

Samples	Si (μg/mL)	Delta Si (μg/mL)	SiO_2 (pg/cell)
Native RBCs	8.39	0	0
RMSN-RBC-10	12.40	4.01	0.085
RMSN-RBC-25	10.47	2.08	0.044
RMSN-RBC-50	9.93	1.54	0.032
RMSN-RBC-200	N/A	N/A	N/A

The results are determined by ICP-MS analysis. RBC: Red blood cell; Native RBCs: Not dialyzed red blood cells; RMSN-RBC-(10 to 200): the engineered RMSN-RBCs with various size of RMSNs. N/A = not applicable.

Figure S8

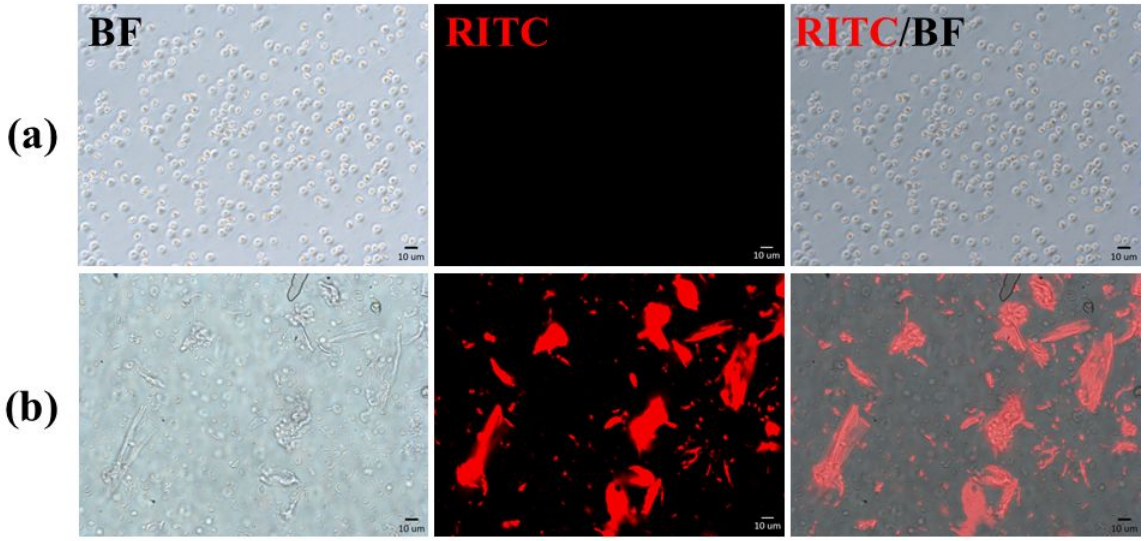

Figure S8. Optical and fluorescent imaging of the engineered RMSN-RBCs (mouse RBC), which is (a) native mRBCs and (b) RMSN-mRBC-50. From left to right of imaging channel, BF: bright field, RITC: detecting the RBCs containing RITC dye-containing particles and RITC/BF: merge BF and RITC imaging. All of the images were taken at 40× original magnification.