Supporting Information

In Vivo Integrity and Biological Fate of Chelator-Free Zirconium-89-Labeled Mesoporous Silica Nanoparticles

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Calculation of number of silanol groups per MSN (and $dSiO_2$) particle

To calculate the number of silanol groups per MSN (and $dSiO_2$) particle, we first calculated the number of nanoparticles per gram based on a previously reported method.[1, 2]

The volume of a MSN was first divided into two parts: the pore volume (V_p) and the solid silica volume (V_s). We then define the fraction of pore volume as $x = \frac{V_p}{V_p+V_s}$, where V_p was given by the BET data. For V_s, each gram of MSN will have $V_s = \frac{m_s}{\rho} = \frac{1\,g}{2.2\frac{g}{cm^3}} = 0.455\ cm^3$, where ρ is the density of silica (2.2 g/cm^3). The mass of each MSN was then defined as $m_{MSN} = \rho * V_{MSN} = \rho * \frac{4\pi r^3(1-x)}{3}$, where r is the radius of MSN determined from TEM images. Therefore, number of nanoparticles per gram of MSN could be calculated by using the following equation:

$$N_{MSN} = \frac{1}{m_{MSN}} = \frac{1}{\rho * \frac{4\pi r^3\left(1 - \frac{V_p}{V_p + V_s}\right)}{3}} = \frac{0.749\left(V_p + 0.455\right)}{\pi r^3}$$

For ~90 nm sized $dSiO_2$, the volume of each $dSiO_2$ is $V_{dSiO2} = \frac{4\pi r^3}{3} = \frac{4\pi(45\times10^{-7})^3}{3} = 3.8 \times 10^{-16} cm^3$. The number of $dSiO_2$ per gram can be calculated as $N_{dSiO2} = \frac{1}{V_{dSiO2}\times\rho} = \frac{1}{3.8\times10^{-16}\times2.2} = 1.2 \times 10^{15}/g$.

According to the Zhuravlev model, the concentration of silanol group is directly proportional to the specific surface area of amorphous silica particle, with the surface silanol group density (-Si-OH/nm^2) found to be in the range of 4 to 5 (also known as the Kiselev–Zhuravlev constant).[3] The number of silanol group per MSN particle can be estimated, which are listed in the following table.

Table S1. Number of silanol groups per MSN (and $dSiO_2$) particle.

	Radius (nm)	Surface area (m^2/g)	Pore volume (cm^3/g)	Number of particles per gram	Surface area per particle (nm^2)	Total silanol groups per particle
MSN	~75	581.5	1.31	1.0×10^{15}	5.8×10^{5}	$\sim2.6\times10^{6}$
$dSiO_2$	~45	40.8	0.06	1.2×10^{15}	3.4×10^{4}	$\sim1.5\times10^{5}$

Table S2. ^{89}Zr-labeling yields of MSN with varied concentrations

	2.0 mg/mL	**2×10^{-1} mg/mL**	**2×10^{-4} mg/mL**
15 min	69.0 %	43.0 %	6.3 %
30 min	74.2 %	49.9 %	8.4 %
60 min	80.5 %	60.3 %	7.7 %
120 min	82.5 %	72.2 %	10.8 %

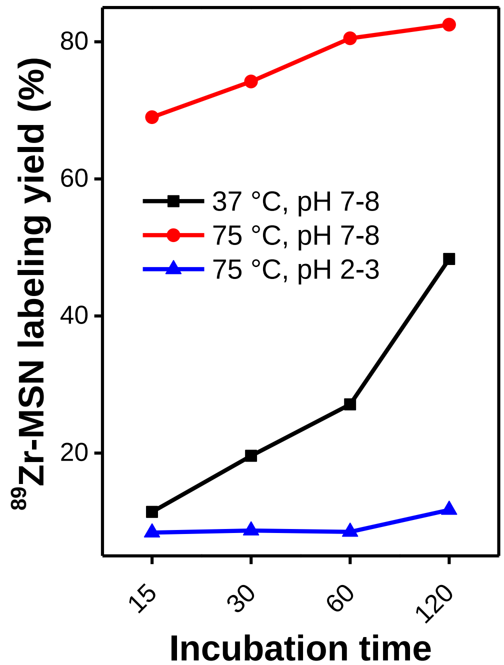

Figure S1. Labeling yields of ^{89}Zr to MSN (2 mg/mL) under different incubation temperatures or in solutions with different pH values.

Table S3. ^{89}Zr-labeling yields of MSN under varied incubation temperatures and in solutions with different pH values

	15 min	**30 min**	**60 min**	**120 min**
37 °C, pH 7-8	11.4 %	19.6 %	27.1 %	48.3 %
75 °C, pH 7-8	69.0 %	74.2 %	80.5 %	82.5 %
75 °C, pH 2-3	8.4 %	8.7 %	8.5 %	11.7 %

Table S4. Drug loading capacity of MSN(DOX) and ^{89}Zr-MSN(DOX).

Samples	Mass of samples	DOX used	Amount of free DOX	DOX in MSN	Loading efficiency	Loading capacity
MSN	500 μg	500 μg	84.9 μg	415.1 μg	83.0 %	830 μg/mg
^{89}Zr-MSN	500 μg	500 μg	103.5 μg	396.5 μg	79.3 %	793 μg/mg

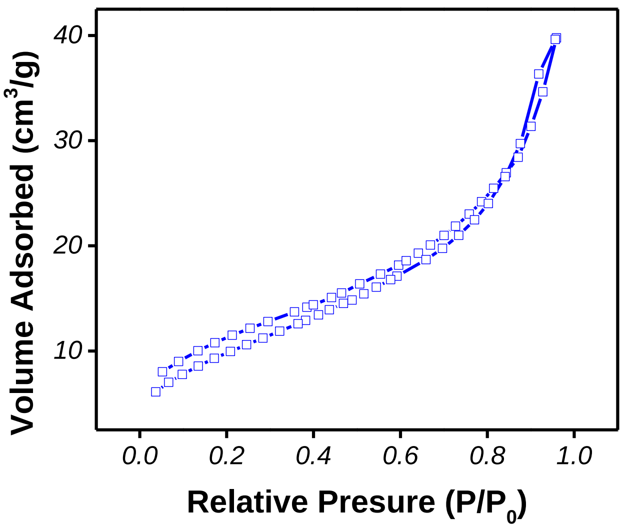

Figure S2. Nitrogen adsorption-desorption isotherms of ~90 nm sized $dSiO_2$. BET surface area was estimated to be 40.8 m^2/g.

Table S5. ^{89}Zr-labeling yields of $dSiO_2$ with varied concentrations

	2.0 mg/mL	**2×10^{-1} mg/mL**	**2×10^{-4} mg/mL**
15 min	30.6 %	9.4 %	7.4 %
30 min	38.7 %	18.5 %	5.3 %
60 min	57.5 %	33.7 %	6.9 %
120 min	76.6 %	38.1 %	10.1 %

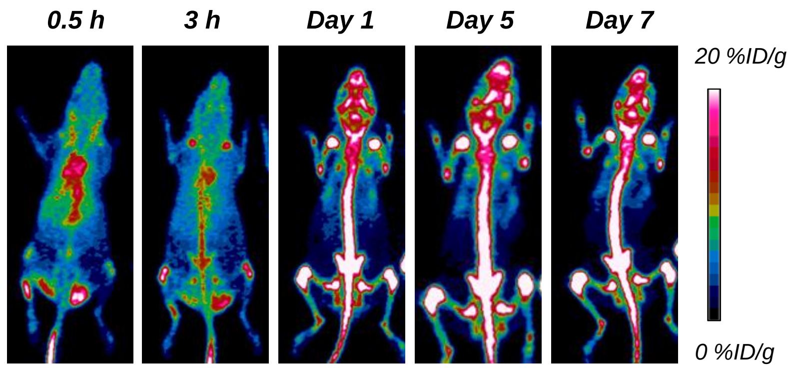

Figure S3. *In vivo* PET maximum intensity projection images of mice injected with free ^{89}Zr-oxalate.

Table S6. ROI quantification data of ^{89}Zr-dSiO_2 and ^{89}Zr-MSN uptake in bone at different time points post-injection.

Bone	**^{89}Zr-dSiO_2 (%ID/g, n=3)**		**^{89}Zr-MSN (%ID/g, n=3)**	
	Ave.	*Std.*	*Ave.*	*Std.*
0.5 h	0.2	0.1	0.2	0.1
3.0 h	0.6	0.5	0.2	0.1
Day 1	6.5	5.9	0.5	0.1
Day 3	11.1	7.5	0.7	0.1
Day 7	11.0	7.0	0.8	0.1
Day 14	11.8	6.6	1.1	0.2
Day 21	9.9	5.6	1.5	0.2

Table S7. ROI quantification data of ^{89}Zr-$dSiO_2$ and ^{89}Zr-MSN uptake in liver at different time points post-injection.

Liver	**^{89}Zr-$dSiO_2$ (%ID/g, n=3)**		**^{89}Zr-MSN (%ID/g, n=3)**	
	Ave.	*Std.*	*Ave.*	*Std.*
0.5 h	38.1	14.5	29.7	4.8
3.0 h	36.6	15.2	30.0	4.7
Day 1	25.2	2.7	32.8	4.8
Day 3	27.0	0.3	38.5	7.4
Day 7	27.3	2.6	39.5	4.5
Day 14	24.6	0.8	37.7	3.1
Day 21	21.1	1.4	34.2	3.3

Table S8. *Ex vivo* biodistribution data of ^{89}Zr-$dSiO_2$ and ^{89}Zr-MSN at Day 21 post-injection

Day 21	**^{89}Zr-$dSiO_2$ (%ID/g, n=3)**		**^{89}Zr-MSN (%ID/g, n=3)**	
	Ave.	*Std.*	*Ave.*	*Std.*
Blood	0.4	0.1	0.2	0.1
Skin	1.3	0.3	0.7	0.1
Muscle	0.4	0.1	0.2	0.1
Heart	0.4	0.1	0.2	0.1
Lung	0.8	0.3	0.7	0.4
Bone	6.3	3.4	1.2	0.4
Liver	23.2	0.3	39.3	5.5
Spleen	42.9	32.5	47.1	21.0
Kidney	1.2	0.3	1.0	0.1
Pancreas	0.4	0.2	0.3	0.2
Stomach	0.6	0.4	0.2	0.1
Intestine	0.3	0.2	0.1	0.0
Tail	1.9	0.5	0.9	0.4
Brain	0.2	0.1	0.32	0.20

References

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2. Lin, Y. S.; Haynes, C. L., Impacts of Mesoporous Silica Nanoparticle Size, Pore Ordering, and Pore Integrity on Hemolytic Activity. *J. Am. Chem. Soc.* 2010, 132, 4834-4842.

3. Zhuravlev, L. T., The Surface Chemistry of Amorphous Silica. Zhuravlev Model. *Colloids Surf. A* 2000, 173, 1-38.