Supporting Information

Biodegradable Nano-Organosilica Gene Carrier for High-Efficiency Gene Transfection

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Experimental section

Part I. The ^{13}C solid state NMR spectra of o-SiNP and o-SiNP-E

The ^{13}C solid state NMR spectra of o-SiNP and o-SiNP-E were obtained on Bruker AVANCE AV III 400 WB, as shown in Figure S1. Three signals at 4.6, 22.7, and 41.2 ppm (Peak 1, 2, and 3 on the spectrum of o-SiNP) are assigned to the carbon atoms between S and Si in BTSPTS, and the signal at 11.7 ppm (Peak 4 on the spectrum of o-SiNP) is assigned to the carbon atoms between Si and Si in BTSE. The strong signals at around 72 ppm assigned to the carbon atoms in the unit of -CH_2-O-CH_2- emerge in the spectrum of o-SiNP-E, indicating the successful introduction of epoxy groups on o-SiNP.

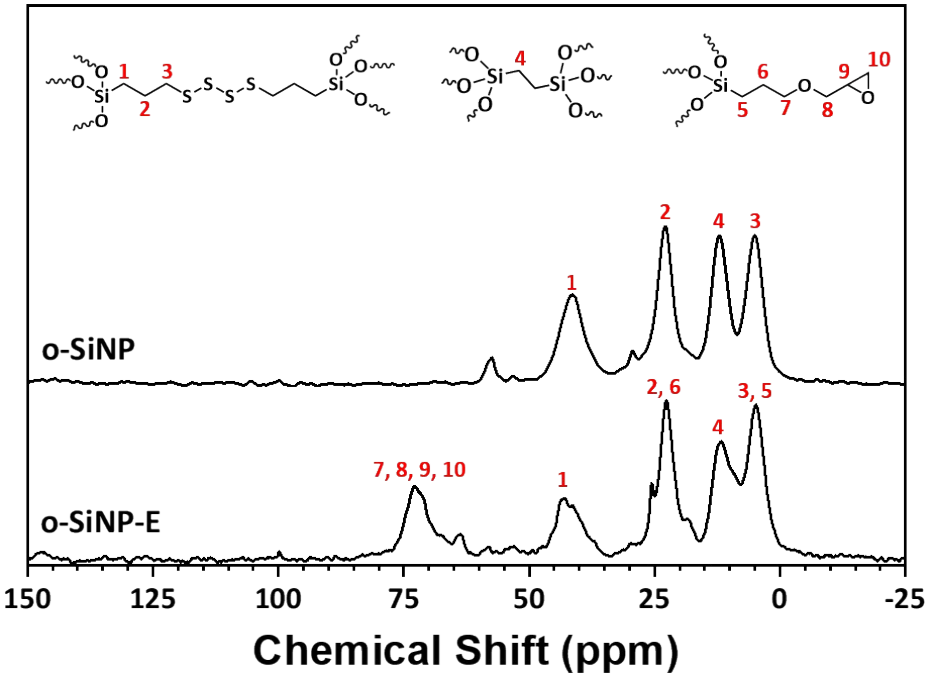

Figure S1. The ^{13}C solid state NMR spectra of o-SiNP and o-SiNP-E.

Part II. Synthesis and characterization of α-polylysine (PLL)

PLL was synthesized according to the previous work,[1-3] as shown in Figure S2.

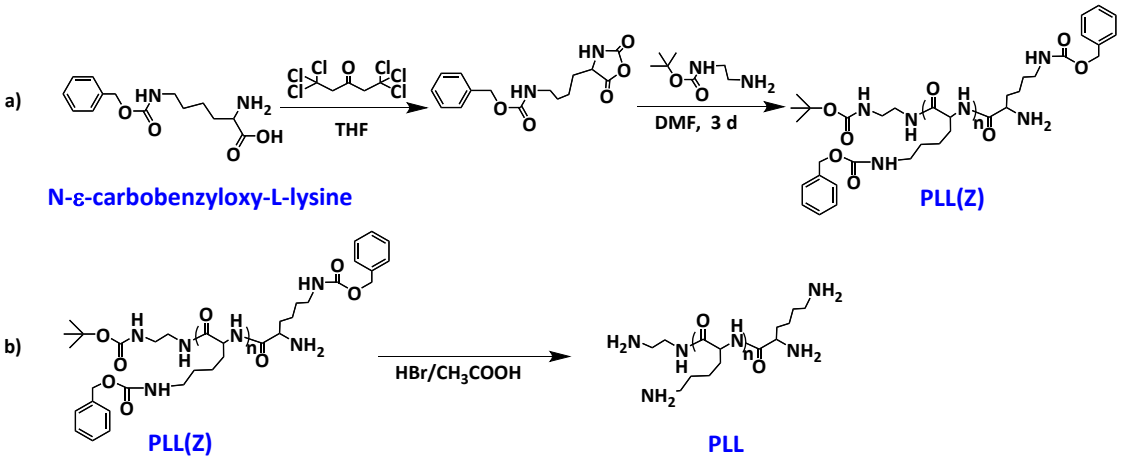

Figure S2. Synthesis of PLL

a) Synthesis of α-polylysine-carbobenzyloxy (PLL(Z))

N-ε-carbobenzyloxy-L-lysine (20.0 g) was added in 100 mL of THF. After the suspension was heated to 55 °C, 20 mL of THF containing 10 g of triphosgene was added dropwise during 30 min to obtain a clear solution. Then the solution was added into 500 mL of petroleum ether. The system was stored in the fridge overnight at -20 °C to let the crude product precipitate completely. The precipitation was collected by filtration, washed with diethyl ether for three times, and dried in a vacuum oven at 20 °C for 5 h to obtain the intermediate product N-ε-benzyloxycarbonyl-lysine-N-carboxyanhydride. The polymerization of N-ε-benzyloxycarbonyl-lysine-N-carboxyanhydride (2.5 g) was dissolved in 5 mL of DMF at 40 °C under nitrogen atmosphere with N-(tert-butyloxycarbonyl (Boc))-ethylenediamine (10 mg) as the initiator. The polymerization product, PLL(Z), was precipitated in diethyl ether, washed with ether twice, and dried in a vacuum oven at 20 °C for 5 h. Figure S3 shows the ^{1}H NMR spectrum of PLL(Z) (300 MHz, DMSO-d_6) measured by Bruker AVANCE AV300 nuclear magnetic resonance spectrometer. The chemical shifts (ppm) of PLL(Z): 7.72-8.21 (m, H), 7.40-7.09 (m, H), 4.98 (s, H), 3.05-2.84 (m, H), 1.78-1.16 (m, H), 1.08 (s, H).

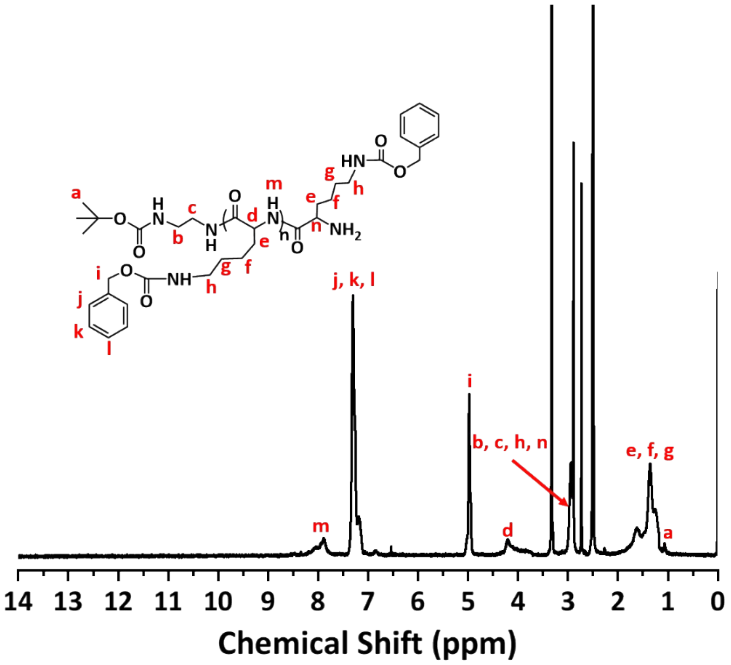

Figure S3. ^{1}H NMR spectrum of PLL(Z)

b) Deprotection of PLL(Z)

PLL(Z) (500 mg) was dissolved in 10 mL of acetic acid. The solution was stirred at room temperature for 3 h after 4 mL of hydrobromic acid in acetic acid (33%) was added. The system was dialyzed (molecular weight cut-off, 14000-7000 Da) in deionized water for 48 h. Finally, the synthesized PLL was collected by lyophilization. As shown in Figure S4, the chemical shifts (ppm) on the ^{1}H NMR spectrum of PLL (300 MHz, D_2O): 4.19 (s, H), 3.09-2.74 (m, H), 1.79-1.45 (m, H), 1.45-1.10 (m, H). The disappearance of the chemical shifts of 7.72-8.21, 4.98, and 1.08 ppm proved that carbobenzyloxy and Boc groups had been removed from PLL(Z).

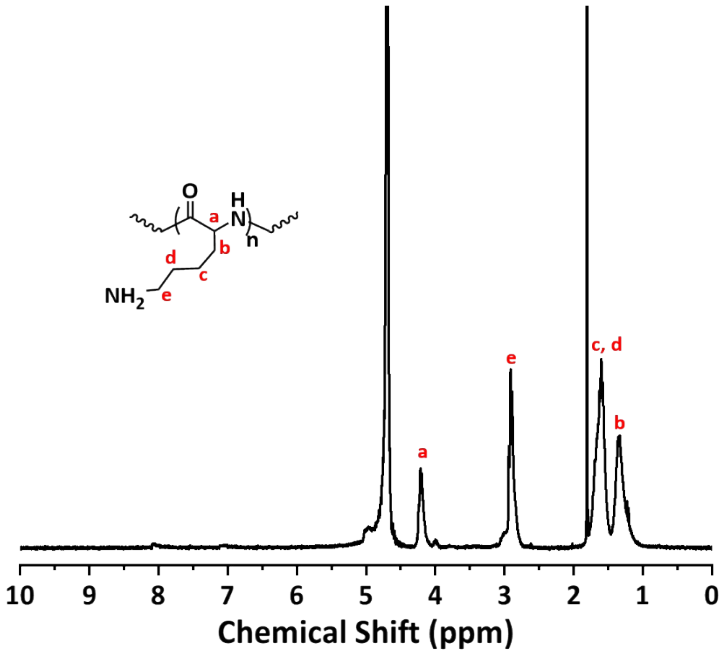

Figure S4. ^{1}H NMR spectrum of PLL

Part III. Synthesis of guanidinated-fluorinated α-polylysine (PLL-GF)

The synthesis route and the ^{1}H NMR spectrum of PLL-GF were shown in Figure S5 and S6, respectively. The chemical shifts (ppm) on the ^{1}H NMR spectrum (300 MHz, D_2O): 4.19 (s, H), 3.10 (s, H), 2.97-2.74 (m, H), 1.79-1.45 (m, H), 1.45-1.10 (m, H). The new chemical shift of 3.10 ppm proved the arginine had already been bonded on PLL.

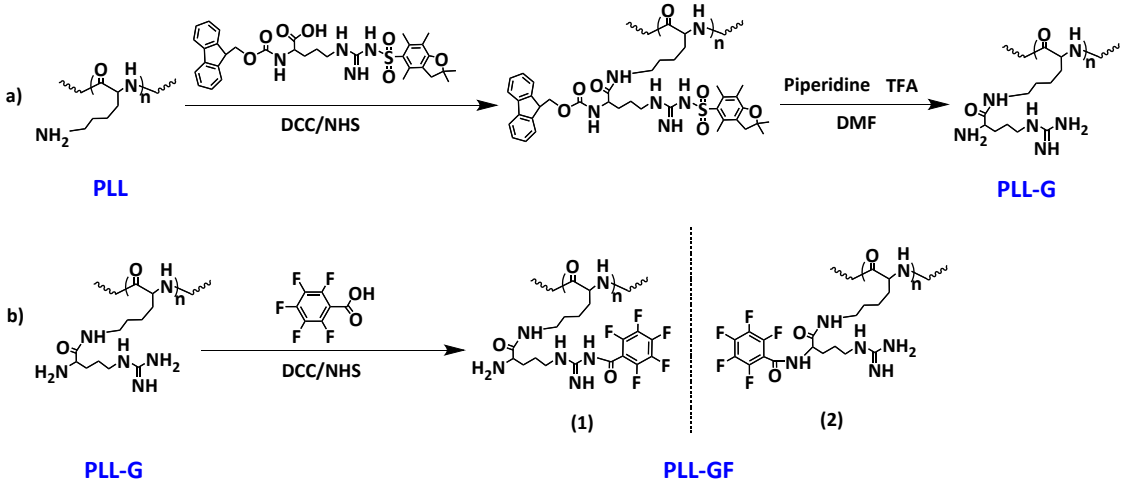

Figure S5. Synthesis of PLL-GF

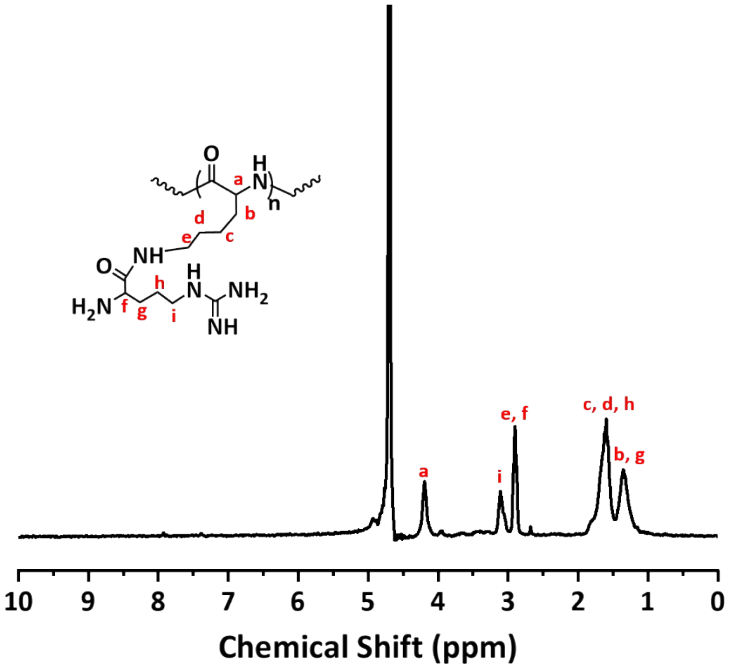

Figure S6. ^{1}H NMR spectrum of PLL-G

Part IV. Characterization

a) Fluorescamine assay of PLL-G and PLL-GF

The fluorescamine assay was measured according to the previous work.[4] Generally, 5 mg of polymer (PLL-G or PLL-GF) was dissolved in 1 mL of water, followed by the addition of 10 mL of an aqueous solution of fluorescamine (20 mg). The mixture was incubated at room temperature for 10 min. The fluorescence emission spectrum of the mixture excited by 290 nm laser was obtained by fluorescence spectrophotometer, as shown in Figure S7. The integral area of emission peak of PLL-GF was 81% of that of PLL-G, indicating that 19% of primary amine groups on PLL-G had reacted with pentafluorobenzoic acid.

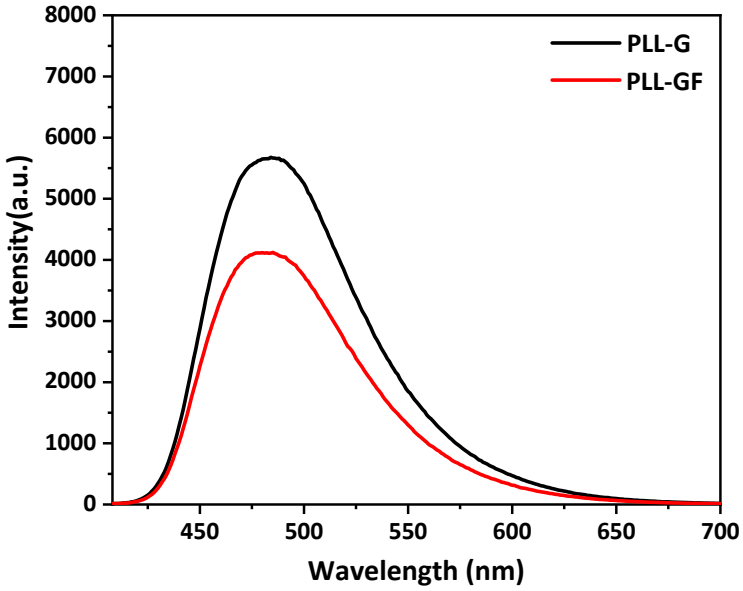

Figure S7. The fluorescence emission spectra of fluorescamine mixed with PLL-G and PLL-GF respectively

b) Element analysis of o-SiNP-GF, PLL-GF, and PEI

The mass proportions of N, C, and H elements in o-SiNP-GF, PLL-GF, and PEI were measured by element analyzer (VarioELIII, Elementar, Germany), as shown in Table S1.

Table S1. The mass proportions of N, C, and H elements in o-SiNP-GF, PLL-GF, and PEI

	N (%)	C (%)	H (%)
o-SiNP-GF	6.5	23.4	5.2
PLL-GF	18.2	47.8	9.1
PEI	31.8	54.5	13.7

c) Synthesis of organosilica nanoparticles based on TEOS and BTSPTS

TEA (0.80 g) and CTAB (0.25 g) were dissolved in 60 mL of water. After TEOS (0.36 mL) and BTSPTS (0.24 mL) were added in the aqueous solution, the mixture was stirred vigorously for 10 h at 80-85 °C. The solid product was collected by centrifugation and washed with ethanol for three times. Then, the product was extracted with 100 mL of ethanol containing 1 g of NH_4NO_3 for 24 h at 65 °C under reflux condition to remove CTAB. After that, the product was collected by centrifugation and washed with ethanol and water alternatively for three times, and dried in a vacuum oven at 50 °C for 16 h. The obtained nanoparticle was labeled as o-SiNP-T. The TEM image of o-SiNP-T (Figure S8) exhibits that the particles have a size more than 200 nm, and stick to each other.

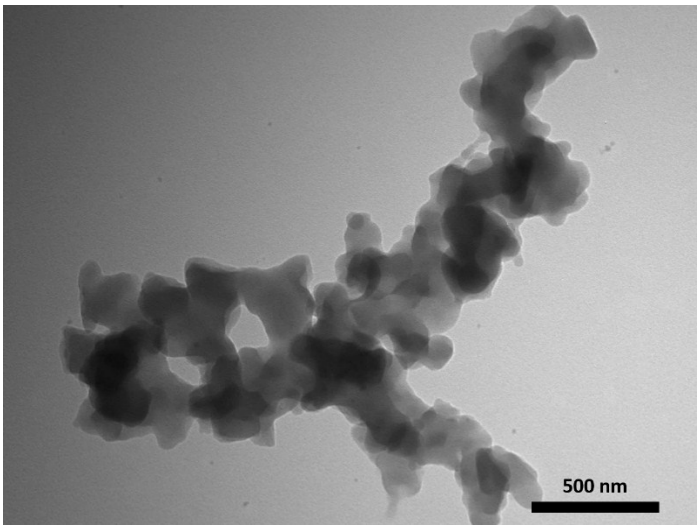

Figure S8. TEM image of o-SiNP-T

References

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