Supporting Information

Using porous magnetic iron oxide nanomaterials as a facile photoporation nanoplatform for macromolecular delivery

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S1. Properties of P-MNPs



Figure S1. (A) Optical image of P-MNPs dispersed in H₂O. (B) Optical image of (A) attracted by a magnet. (C) Optical images showing the stability of P-MNPs dispersed in H₂O, PBS, DMEM and FBS+DMEM respectively.



Figure S2. The stability of P-MNPs in different buffers ranging from 0 h to 72 h determined by DLS. (A) Particle size. (B) Zeta potential.



Figure S3. SEM images of the P-MNPs deposited for the concentrations at (A) 100 μ g·mL⁻¹, (B) 200 μ g·mL⁻¹ and (C) 300 μ g·mL⁻¹ respectively. Scale bar: 2 μ m.

S2. Cell viability



Figure S4. Relative viability of HeLa cells cultured on the surface covered with P-MNPs 0 h and 72 h after laser irradiation under various conditions. I₀, I₁ and I₂ refer to 25 μ g of P-MNPs with no irradiation, 3.5 W·cm⁻² for 60 s and 5 W·cm⁻² for 45 s respectively; II₀, II₁, II₂ and II₃ refer to 50 μ g of P-MNPs with no irradiation, 2 W·cm⁻² for 60 s, 3.5 W·cm⁻² for 45 s and 5 W·cm⁻² for 30 s respectively; III₀, III₁, III₂ and III₃ refer to 75 μ g of P-MNPs with no irradiation, 2 W·cm⁻² for 45 s, 3.5 W·cm⁻² for 30 s respectively; III₀, III₁, III₂ and III₃ refer to 75 μ g of P-MNPs with no irradiation, 2 W·cm⁻² for 45 s, 3.5 W·cm⁻² for 30 s respectively.



Figure S5. Relative viability of HeLa cells 48 h after dextran delivery under various conditions. I₀, I₁ and I₂ refer to 25 μ g of P-MNPs with no irradiation, 3.5 W·cm⁻² for 60 s and 5 W·cm⁻² for 45 s respectively; II₀, II₁, II₂ and II₃ refer to 50 μ g of P-MNPs with no irradiation, 2 W·cm⁻² for 60 s, 3.5 W·cm⁻² for 45 s and 5 W·cm⁻² for 30 s respectively; III₀, III₁, III₂ and III₃ refer to 75 μ g of P-MNPs with no irradiation, 2 W·cm⁻² for 45 s and 5 W·cm⁻² for 30 s respectively; III₀, III₁, III₂ and III₃ refer to 75 μ g of P-MNPs with no irradiation, 2

S3. Transfection of pDNA



Figure S6. Transfection of GFP pDNA into HeLa cells mediated by PEI with or without 808 nm laser irradiation. (A) Typical fluorescence images. Cell nuclei were stained by DAPI (blue fluorescence). Scale bar: 100 μ m. (B) Percentage of GFP positive cells. (C) Green fluorescence intensity. (D) Relative viability of HeLa cells 48 h after gene transfection. Group Ctrl refer to gene transfection without treatment in cell plate; Groups P1, P2 and P3 refer to gene transfection mediated by PEI with N/P ratios of 20, 30 and 40, respectively. wo/Laser means gene transfection without laser irradiation; w/Laser means gene transfection under 808 nm laser irradiation at 5 W·cm⁻² for 30 s.



Figure S7. Typical bright field and fluorescence images of HeLa cells after the transfection of GFP pDNA. Ctrl group: cells after pDNA transfection without treatment in cell plate; M group: cells after pDNA transfection mediated by P-MNPs (50 μ g) under 808 nm laser irradiation at 5 W·cm⁻² for 30 s; M/P3 group: cells after pDNA transfection mediated by P-MNPs (50 μ g) under 808 nm laser irradiation at 5 W·cm⁻² for 30 s; M/P3 group: cells after state by P-MNPs (50 μ g) under 808 nm laser irradiation at 5 W·cm⁻² for 30 s with the assistance of PEI with N/P ratio of 40. Cell nuclei were stained by DAPI (blue fluorescence). Scale bar: 100 μ m.



Figure S8. Transfection efficiency of HeLa transfected with pRL-CMV under different conditions. The data are presented as the mean \pm SD (n = 6), 0.01 , ***<math>p < 0.001.



Figure S9. (A) Typical fluorescence microscopy images showing the expression of GFP in mEFs mediated by P-MNPs (50 µg) upon 808 nm laser irradiation at 5 W·cm⁻² for 30 s with the assistance of PEI with N/P ratio of 40 (Group M/P3). Ctrl group: cells after GFP pDNA transfection without treatment in cell plate. Cell nuclei were stained by DAPI (blue fluorescence). Scale bar: 100 µm. The corresponding green fluorescence intensities were summarized in (B). The data are presented as the mean \pm SD (n = 6).



Figure S10. Transfection efficacy of GFP pDNA into mEFs mediated by Lipo2000 and M/P3. (A) Percentage of GFP positive cells. (B) Green fluorescence intensity. Ctrl group: pDNA transfection without treatment in cell plate; Lipo2000 group: pDNA transfection mediated by lipo2000; M/P3 group: pDNA transfection mediated by P-MNPs (50 µg) under 808 nm laser irradiation at 5 W·cm⁻² for 30 s with the assistance of PEI with N/P ratio of 40. The data are presented as the mean \pm SD (n = 6), ***p < 0.001.



Figure S11. Typical fluorescence images of pDNA GFP transfected HeLa cells after trypsinization and 24 h after subculture. Ctrl group: cells after pDNA transfection without treatment in cell plate; M/P3 group: cells after pDNA transfection mediated by P-MNPs (50 μ g) under 808 nm laser irradiation at 5 W·cm⁻² for 30 s with the assistance of PEI with N/P ratio of 40; GNPL group: cells after pDNA transfection mediated by gold nanoparticle layer (GNPL) under 808 nm laser irradiation at 5 W·cm⁻² for 30 s. Scale bar: 100 μ m.

	Control	P-MNPs	GNPL
Cell viability ^a	100%	$98.5\pm4.3\%$	91.2± 5.7%
Delivery efficiency ^b	N.D.	$66.6\pm2.6\%$	$98.7\pm3.8\%$
Cell recovery efficiency ^c	100%	$97.0 \pm 1.3\%$	N.D.

 Table S1. Comparison of macromolecular delivery and cell recovery performance

 between P-MNPs and GNPL.

^a Relative viability of HeLa cells 48 h after GFP pDNA transfection, the viability of cells cultured on cell culture plate (Control) were taken as 100%.

^b The percentage of GFP positive cells after GFP pDNA transfection to HeLa.

^c The percentage of recovered cells when compared with control.