Electronic Supplementary Information

Biocompatible Protamine Sulfate@Silicon Nanoparticles-Based Gene Nanocarriers Featuring Strong and Stable Fluorescence

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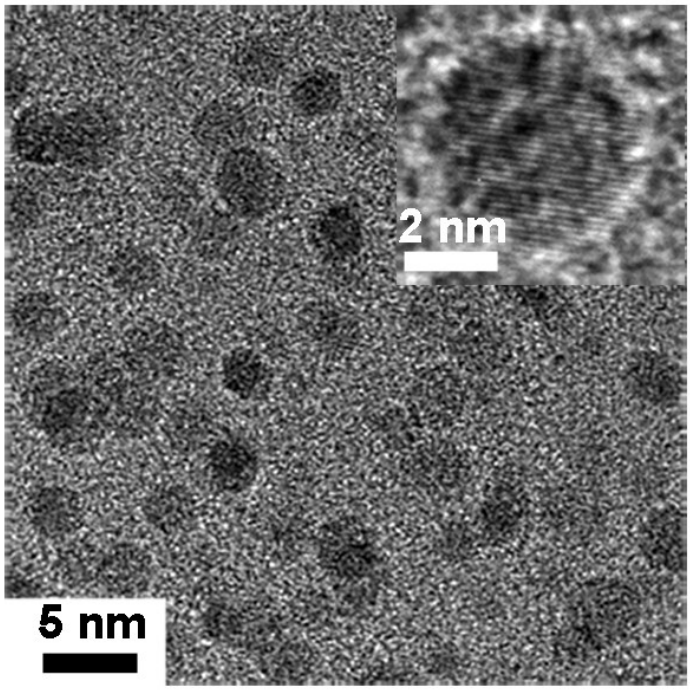

Fig. S1 TEM and HRTEM (inset) images of SiNPs.

As shown in Fig. S1, fluorescent SiNPs appear as spherical particles with good monodispersity. Moreover, the HRTEM image of a single NP clearly demonstrates excellent crystallinity of the resultant SiNPs (inset in TEM image).

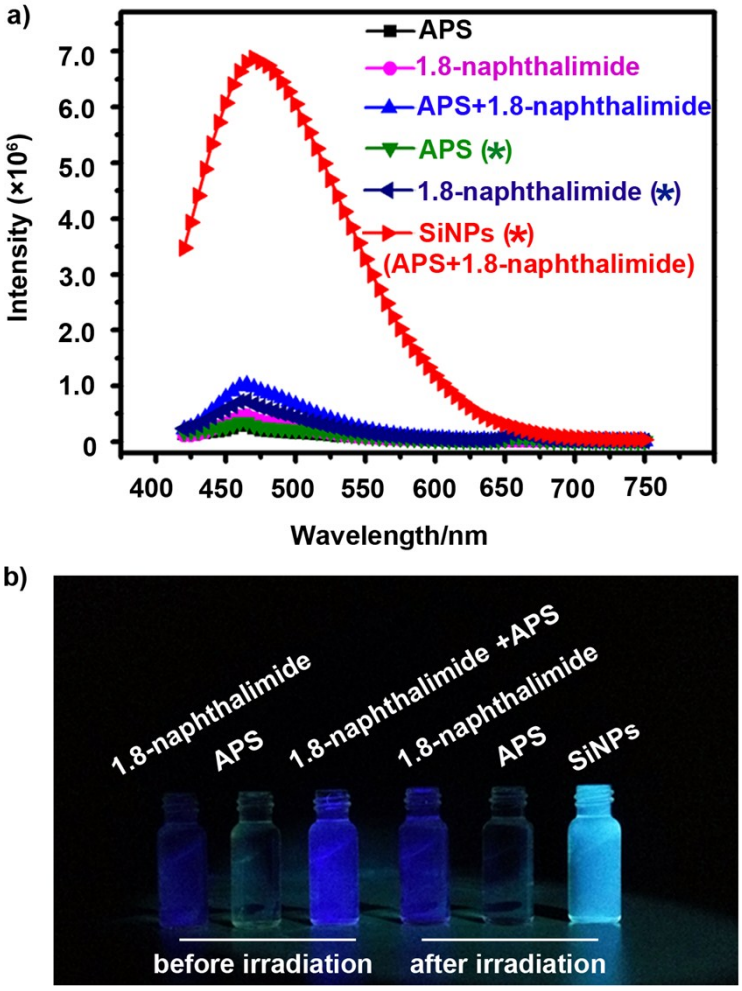

Fig. S2 (a) PL spectra of pure 1.8-naphthalimide, pure APS and reaction precursor (i.e., 1.8-naphthalimide + APS) before and after irradiation (*). (b) Photographs of corresponding aqueous solutions under UV irradiation (450 W xenon lamp of 365 nm).

As shown in Fig. S2a, feeble PL spectra are observed for pure 1.8-naphthalimide and pure APS before and after UV irradiation. In sharp contrast, for the reaction precursor solution (i.e., 1.8-naphthalimide + APS), distinct fluorescence is produced through UV-irradiation treatment. Furthermore, compared to feeble fluorescence of control groups (i.e., 1.8-naphthalimide, APS, 1.8-naphthalimide + APS), obvious fluorescence is readily observed for the aqueous solution of the resultant SiNPs under UV irradiation (Fig. S2b). These results demonstrate that the observed fluorescence is generated by the resultant NPs.

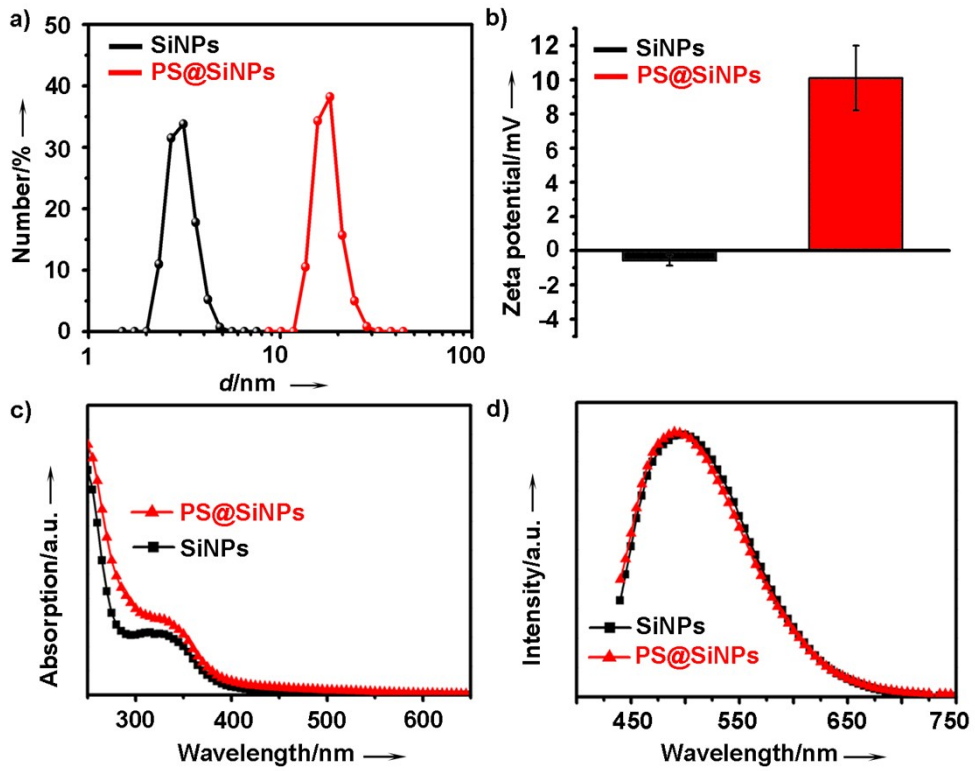

Fig. S3 Characterizations of SiNPs and the resultant PS@SiNPs. DLS (a), zeta potential (b), absorption and photoluminescence (UV-PL) spectra (c and d) of SiNPs (black line) and the resultant PS@SiNPs (red line).

As shown in Fig. 3a, the DLS diameter of SiNPs or PS@SiNPs equals to ~4 and ~20 nm, respectively. The SiNPs have a negligible negative charge of ~0.6 ± 0.3 mV; comparatively, PS@SiNPs-based gene carriers exhibit a positive charge of 10.1 ± 1.9 mV due to the positively charged PS (Fig. S3b). UV-PL spectra demonstrate that both of the SiNPs and PS@SiNPs-based gene carriers exhibit similar UV-PL spectral profiles (Fig. S3c and S3d).

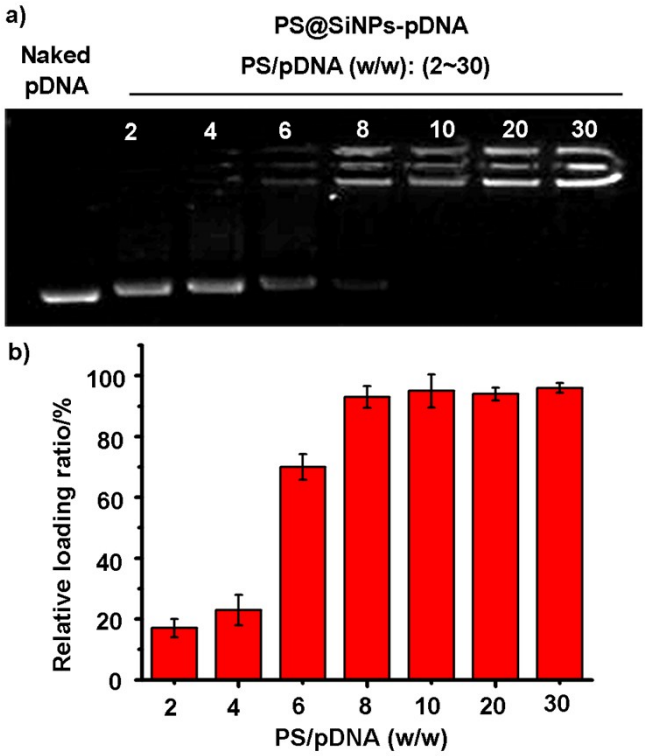

Fig. S4 DNA binding ability of PS@SiNPs. (a) Agarose gel electrophoresis patterns of PS@SiNPs-pDNA with different PS/pDNA ratios (i.e., 2-30). Naked pDNA is used as control. The amount of pDNA (0.2 μg) is constant in these samples. (b) Quantitative analysis of electrophoresis results (a) analyzed by Image J software.

DNA binding ability of the PS@SiNPs is tested by an established agarose gel electrophoresis assay. As shown in Fig. S4a, pDNA band is undetectable when the PS/pDNA ratio increases to 8. Quantitative data confirms that the more than 95% of pDNA binds on PS@SiNPs-based gene carriers when the ratios of PS/pDNA are over 8 (Fig. S4b).

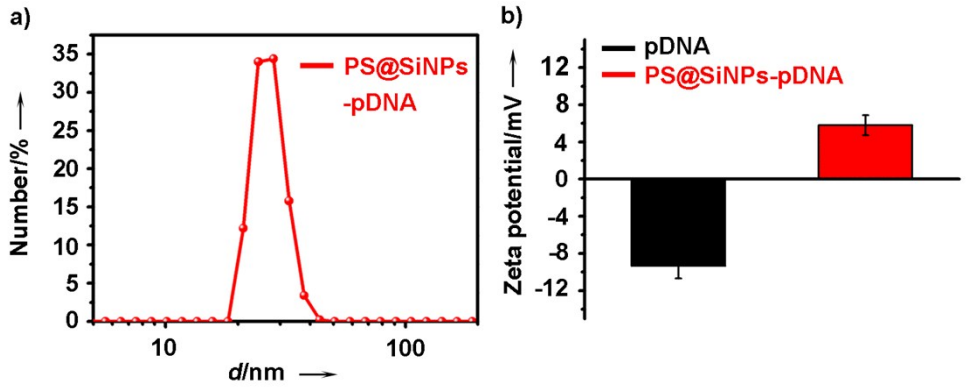

Fig. S5 Characterizations of the resultant PS@SiNPs-pDNA gene carriers with a PS/pDNA ratio of 40. DLS (a) and zeta potential (b) of naked pDNA and the resultant PS@SiNPs-pDNA.

As shown in Fig. S5a, the DLS diameter of the resultant PS@SiNPs-pDNA equals to ~28 nm. Naked pDNA has a negative charge of ~9.3 ± 1.3 mV; comparatively, PS@SiNPs-pDNA gene carriers exhibit a positive charge of 5.8 ± 1.1 mV (Fig. S5b).

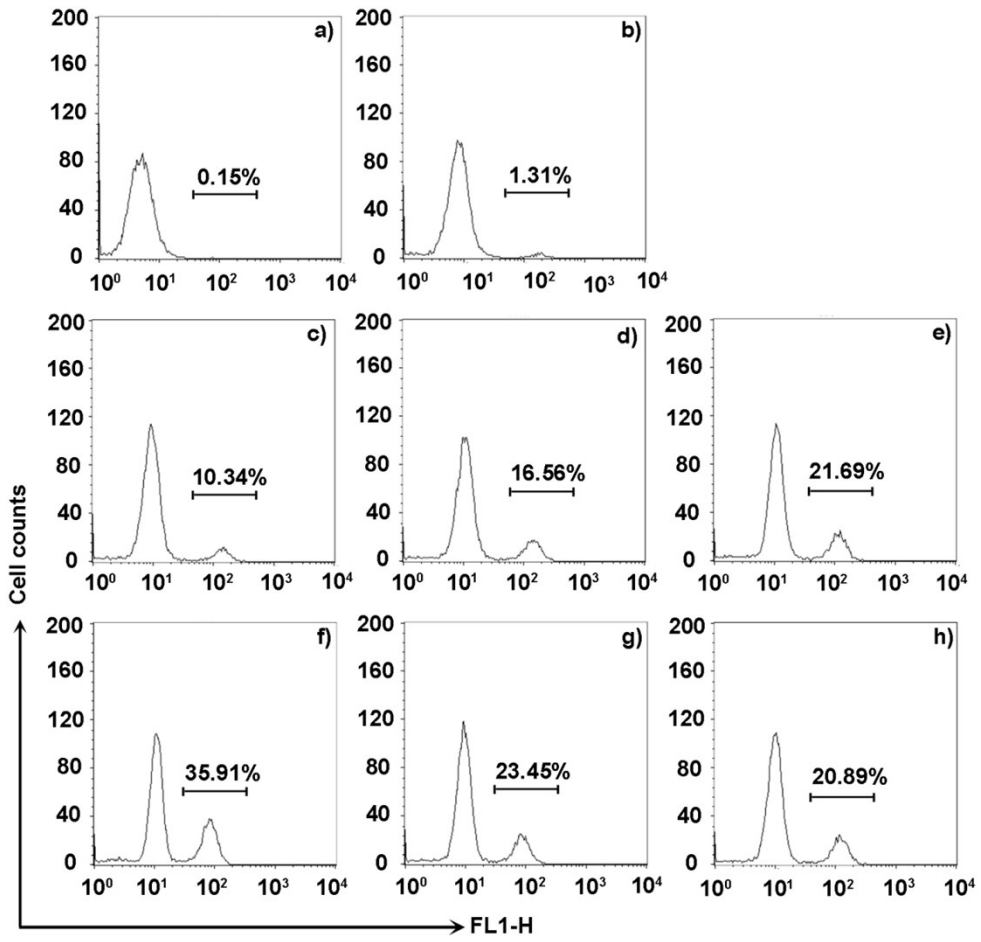

Fig. S6 Transfection efficiency measured by flow cytometry. Cells are non-treated (a), transfected with naked pDNA (b), and PS@SiNPs-pDNA with different PS/pDNA ratios (i.e., 10, 20, 30, 40, 50, and 60) (c-h). Transfection is performed at a dose of 1 μg/well of pDNA. The numbers present the transfection efficiency calculated based on the percentages of mCherry positive cells in total cells.

Flow cytometry is used to measure the transfection efficiencies of PS@SiNPs-pDNA with different PS/pDNA ratios. As shown in Fig. S6, for naked pDNA-treated cells, the percentage of mCherry positive cells in total cells is ~1.31%, indicating naked pDNA is difficult to transfect cells. In contrast, mCherry pDNA encapsulated in PS@SiNPs-pDNA gene carriers could be effectively translated into ARPE-19 cells. The maximal percentage of mCherry positive cells in total cells is 35.91 % at the PS/pDNA ratio of 40.

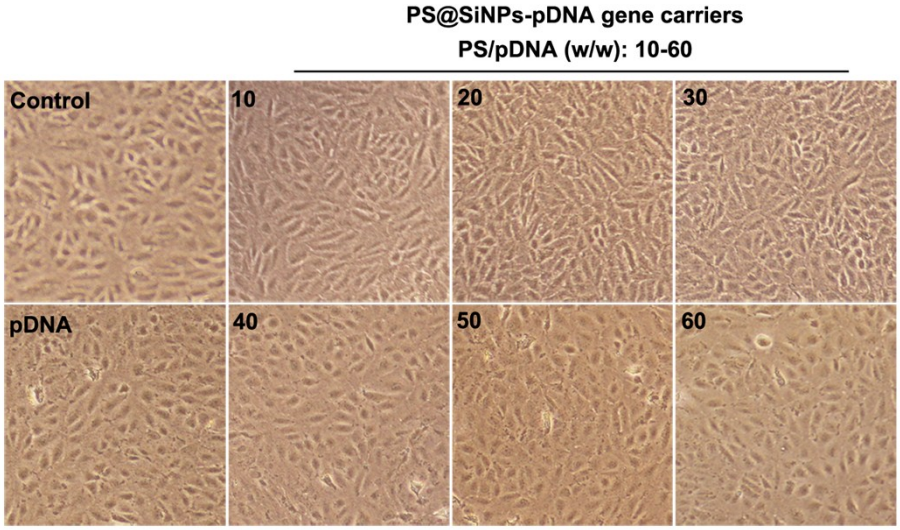

Fig. S7 Morphology of ARPE-19 cells treated by PS@SiNPs-pDNA gene carriers with different PS/pDNA ratios (i.e., 10-60) for 48 h.

As shown in Fig. S7, compared to non-treated cells, no obvious morphological changes are observed for the PS@SiNPs-pDNA gene carriers-treated ARPE-19 cells within 48-h incubation, indicating negligible cytotoxicity of the PS@SiNPs-pDNA gene carriers.

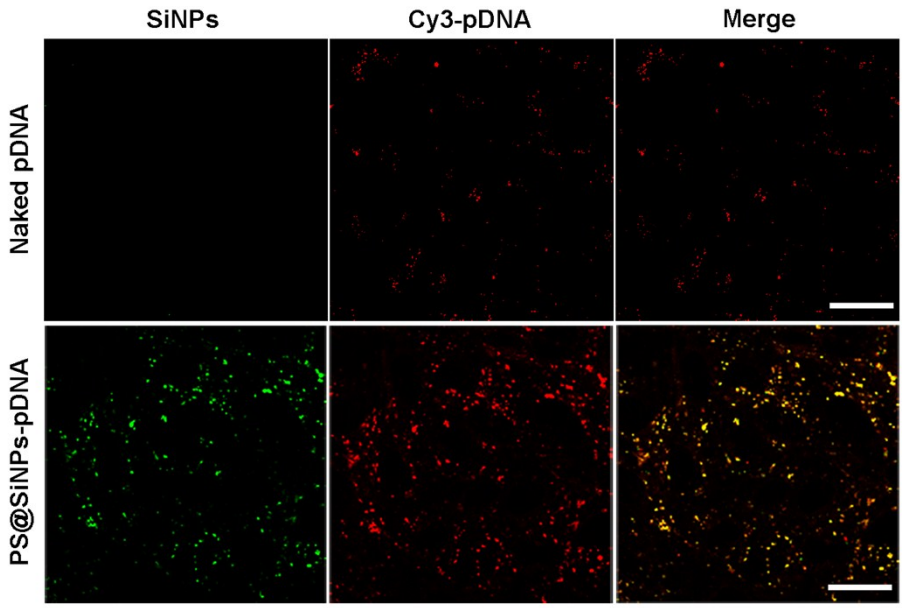

Fig. S8 Enhanced cellular internalization of pDNA mediated by PS@SiNPs-based gene carriers. Cells are incubated with PS@SiNPs-pDNA gene carriers with PS/pDNA ratio of 40 for 6 h. Fluorescence signals of SiNPs and Cy3-labeled pDNA are defined as green and red, respectively. Naked pDNA is used as control group.

As shown in Fig. S8, PS@SiNPs-pDNA gene carriers have better cellular internalization than naked pDNA. Typically, after treatment with naked pDNA, feeble fluorescence signals from Cy3-labeled pDNA are observed in cytoplasm, indicating naked pDNA can hardly enter into cells due to the negative charge and relative low stability. In sharp contrast, after 6-h incubation with PS@SiNPs-pDNA gene carriers, a mass of green (SiNPs) and red fluorescence signals (Cy3-labeled pDNA) with good colocalization are observed inside cells, which means pDNA formulated in PS@SiNPs-pDNA gene carriers could be effectively delivered into cells.

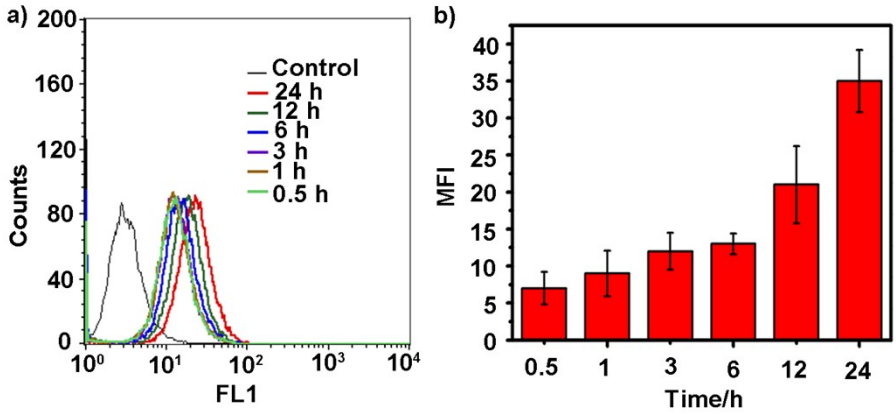

Fig. S9 Cellular uptake of PS@SiNPs-pDNA gene carriers in ARPE-19 cells. (a) Flow cytometry histogram and (b) corresponding mean fluorescent intensity (MFI).

Flow cytometry histogram presents time-dependent internalization of PS@SiNPs-pDNA gene carriers during 24 h incubation (Fig. S9a). Similarly, fluorescent intensities are gradually enhanced during 0.5-24 h incubation (Fig. S9b).