Supporting Information

Polyethyleneimine-Grafted Hyperbranched Conjugated

Polyelectrolytes: Synthesis and Imaging of Gene Delivery

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1. ¹H NMR spectra for HCP, HCPE, HCPEPEG, HCPEPEI-1, HCPEPEI-2 and details of calculation for N contents

The integration at 3.17 ppm has a ratio of 0.06 to that at 2.03 ppm, indicating 76% of the alkyne groups have been reacted. The number of newly formed benzene rings is 7 and degree of polymerization is 15. The M_n is estimated to be ~16000. After quanternization, the M_n is ~19000. After conjugation to 8 PEG chains, the M_n is ~23000. The integration ratios for peaks at 2.5 to 3.1 ppm (N⁺(CH₃)₃ and PEI protons) to that at 3.65 ppm (PEG protons) are 3.0 and 5.7 for HCPEPEI-1 and HCPEPEI-2, respectively. The 8 PEG chains gave totally ~320 protons for both HCPEPEI-1 and HCPEPEI-2. After deducting the protons from N⁺(CH₃)₃, there are ~420 and ~1284 PEI protons for HCPEPEI-1 and HCPEPEI-2, respectively. PEI600 has 14 NCH₂CH₂ repeating units, and PEI1800 has 42 NCH₂CH₂ repeating units. The number of protons for PEI ($M_n = 600$) is 56 and that for PEI ($M_n = 1800$) is 168. There are 420/56 = 7.5 mol of PEI600 and 1284/168 = 7.6 mol of PEI1800 conjugated to each HCPEPEG. The molecular weights for HCPEPEI-1 and HCPEPEI-2 are estimated to be

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~31800 and ~41400. The N contents are ~5%, ~13% for HCPEPEI-1 and HCPEPEI-2, respectively.

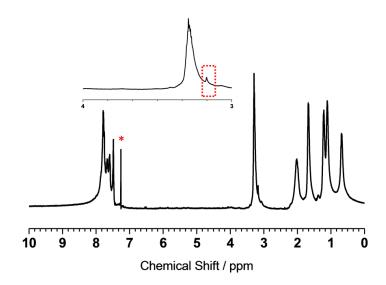


Figure S1. ¹H NMR spectrum of HCP in CDCl₃. * indicates solvent peak.

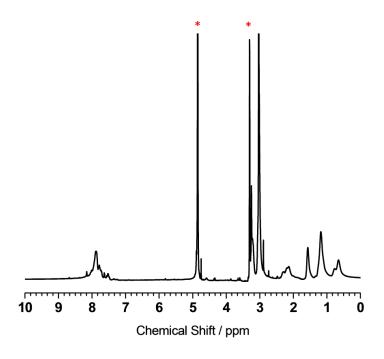


Figure S2. ¹H NMR spectrum of HCPE in MeOD. * indicates solvent peak and water peak.

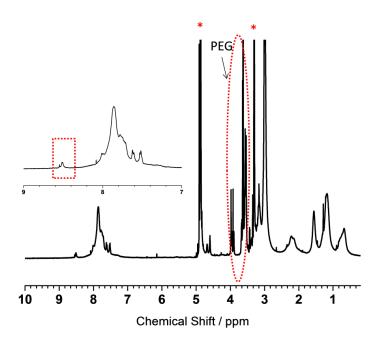


Figure S3. ¹H NMR spectrum of HCPEPEG in MeOD. * indicates solvent peak and water peak.

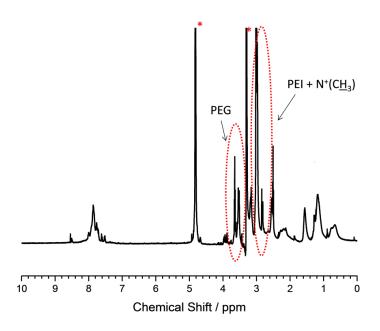


Figure S4. ¹H NMR spectrum of HCPEPEI-1 in MeOD. * indicates solvent peak and water peak.

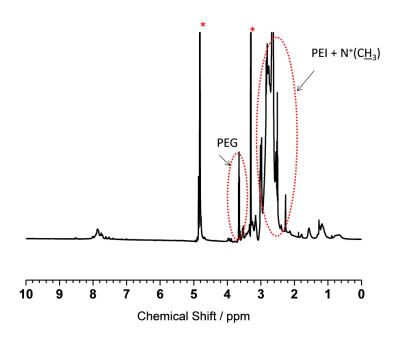


Figure S5. ¹H NMR spectrum of HCPEPEI-2 in MeOD. * indicates solvent peak and water peak.

2. The DLS results of HCPEPEI-1 and HCPEPEI-2 in H_2O

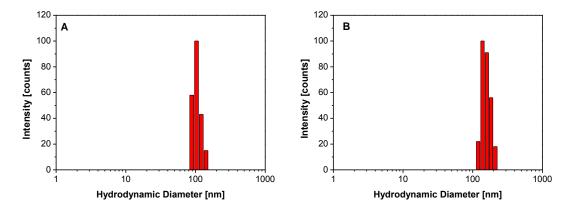


Figure S6. DLS spectra of HCPEPEI-1 (A) and HCPEPEI-2 (B) in H₂O.

3. Cytotoxicity study

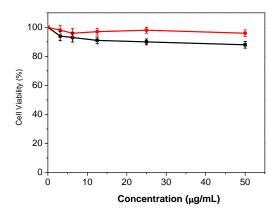


Figure S7. COS-7 cells viability of HCPE (black) and PEG (red) at different concentrations for 24 h.

4. Characterization of polymer/DNA nanoparticles

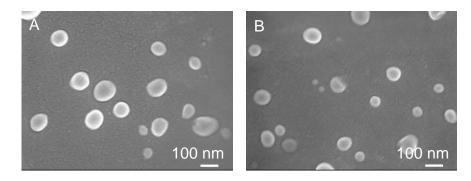


Figure S8. SEM images of HCPEPEI-1/DNA (A) and HCPEPEI-2/DNA (B) nanoparticles at N/P ratio of 30.

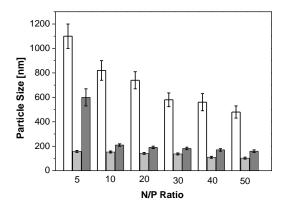


Figure S9. Particle sizes PEI600/DNA (white), PEI1800/DNA (light gray) and PEI25k/DNA (gray) nanoparticles as a function of the N/P ratio.

5. In vitro gene transfection study

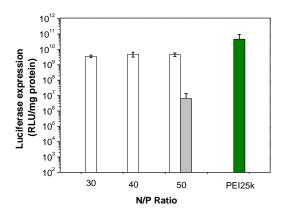


Figure S10. In vitro gene transfection efficiency of PEI600/DNA (gray) and PEI1800/DNA complex (white) in COS-7 cells in the absence of serum at 24 h post-transfection. The green column shows the transfection efficiency of PEI25k/DNA complex at an optimal N/P ratio of 30.

6. Photostability study

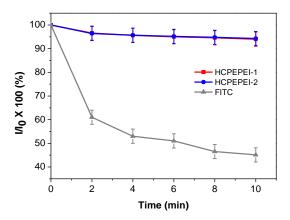


Figure S11. Photostability comparison among HCPEPEI-1, HCPEEPI-2 and FITC upon continuous laser excitation at 405 nm for 0 to 10 min. I_0 is the initial fluorescence intensity; I is the fluorescence intensity of the sample at various time points.