Supporting Information

Maximizing gene delivery efficiencies of cationic helical polypeptides via balanced membrane penetration and cellular targeting

Nan Zheng,[§] Lichen Yin,[§] Ziyuan Song, Liang Ma, Haoyu Tang, Nathan P. Gabrielson, Hua Lu and Jianjun Cheng*

Department of Materials Science and Engineering,

University of Illinois at Urbana-Champaign, 1304 W. Green Street, Urbana, IL, 61801, USA

*Corresponding Author: jianjunc@illinois.edu; Phone: 217-244-3924; Fax: 217-333-2736



Fig. S1. ¹H NMR spectrum of PVBLG in CDCl₃/TFA-d (85:15, v/v).



Fig. S2. ¹H NMR spectrum of PABLG in CDCl₃/TFA-d (85:15, v/v).



Fig. S3. ¹H NMR spectrum of PVBLG-8 in TFA-*d*.



Fig. S4. ¹H NMR spectrum of PVBLG-7 in TFA-*d*.



Fig. S5. ¹H NMR spectrum of PVBLG-8-*r*-7 in TFA-*d*.



Fig. S6. GPC curve of PVBLG obtained from HMDS-initiated polymerization of VB_{-L} -Glu-NCA at the M/I ratio of 150/1. The MW and PDI of the resulting PVBLG were summarized in the figure.



Fig. S7. Residue molar ellipticity at 222 nm of PVBLG-8 and PVBLG-7 at different pH (A) and NaCl concentrations (B).



Fig. S8. CD spectra of PVBLG-8/PVBLG-7 mixtures and their ternary complexes with DNA in DI water at pH 7.



Fig. S9. EB exclusion assay showing the condensation of DNA in the binary complexes (A) and ternary complexes (B) PVBLG-8/DNA weight ratio fixed at 15/1 (n=3).



Fig. S10. CLSM images showing the cellular internalization of ternary complexes (PVBLG-8/PVBLG-7/DNA weight ratio of 15/7/1) in HeLa cells following incubation at 37 °C for 0.5, 1, 2, and 4 h (bar = $20 \mu m$).



Fig. S11. Uptake level of YOYO-1-DNA-containing ternary complexes (PVBLG-8/PVBLG-7/DNA weight ratio of 15/3/1, A) and binary complexes (PVBLG-8/DNA weight ratio of 15/1, B) in COS-7 and Raw 264.7 cells in the presence of free mannose at various concentrations (n=3). (C) Uptake level of ternary complexes (PVBLG-8/PVBLG-7/DNA weight ratio of 15/3/1) in the presence of various endocytosis inhibitors in COS-7 and Raw 264.7 cells (n = 3).



Fig. S12. *In vitro* transfection efficiency of ternary complexes (PVBLG-8/PVBLG-7/DNA weight ratio of 15/3/1, A) and binary complexes (PVBLG-8/DNA weight ratio of 15/1, B) in COS-7 and Raw 264.7 cells in the presence of free mannose at various concentrations (n=3).



Fig. S13. Cytotoxicity of PVBLG-8/PVBLG-7 mixtures at different weight ratios in HeLa, COS-7, and Raw 264.7 cells following 24-h treatment (n=3).