## **Supporting Information**

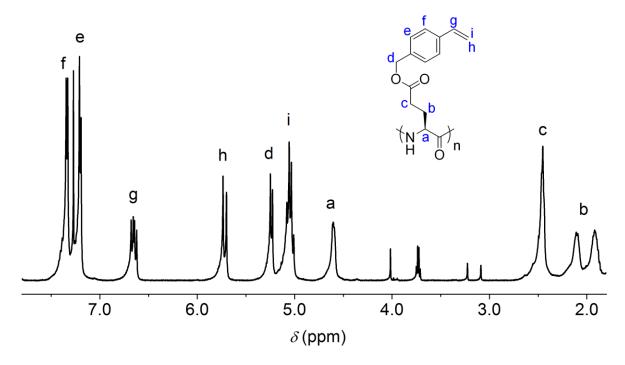
## Maximizing gene delivery efficiencies of cationic helical polypeptides via balanced membrane penetration and cellular targeting

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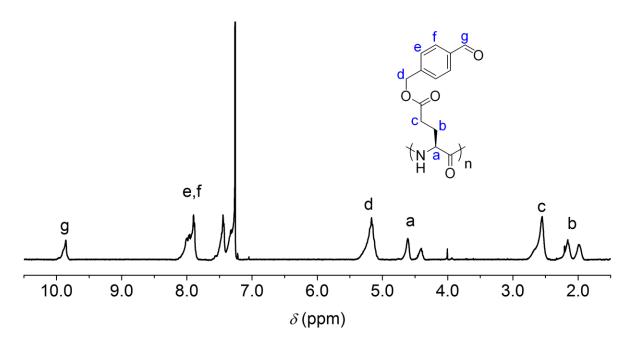
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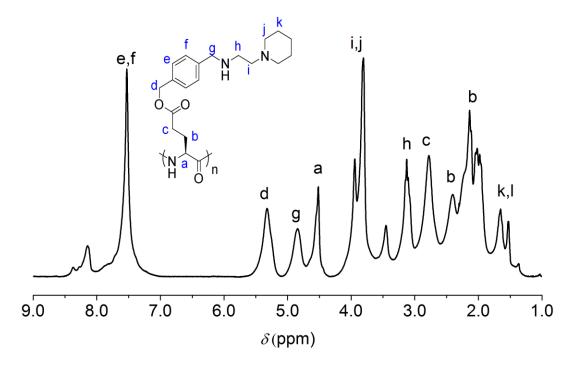
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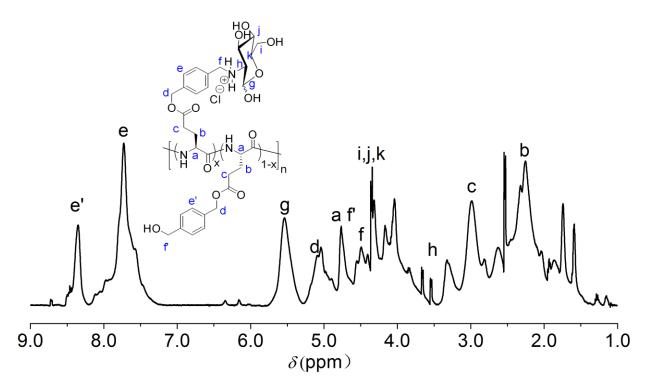
**Fig. S1.** <sup>1</sup>H NMR spectrum of PVBLG in CDCl<sub>3</sub>/TFA-d (85:15, v/v).



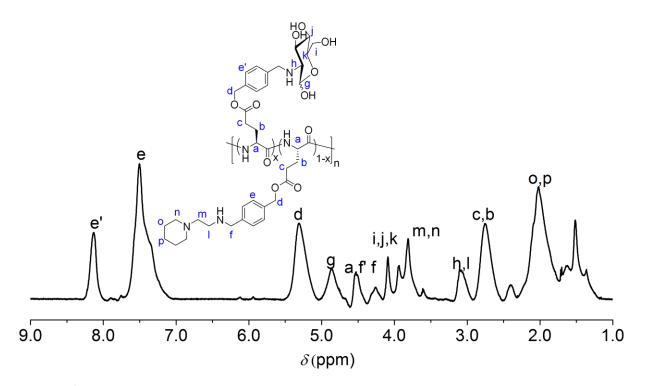
**Fig. S2.** <sup>1</sup>H NMR spectrum of PABLG in CDCl<sub>3</sub>/TFA-d (85:15, v/v).



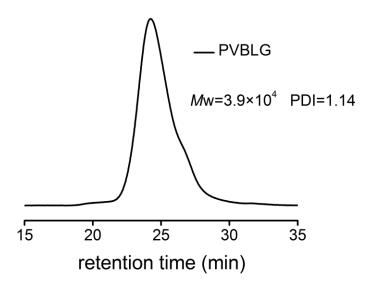
**Fig. S3.** <sup>1</sup>H NMR spectrum of PVBLG-8 in TFA-*d*.



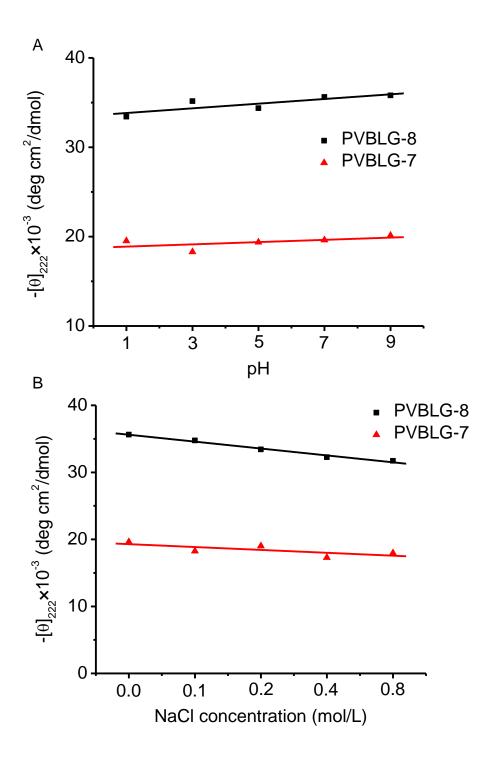
**Fig. S4.** <sup>1</sup>H NMR spectrum of PVBLG-7 in TFA-*d*.



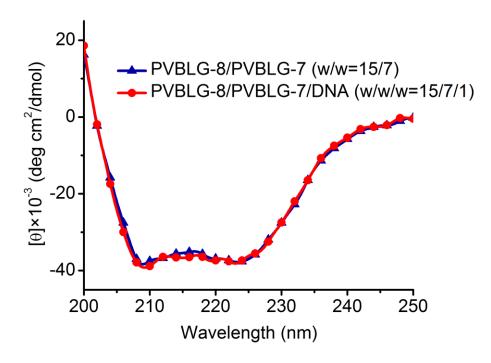
**Fig. S5.** <sup>1</sup>H NMR spectrum of PVBLG-8-*r*-7 in TFA-*d*.



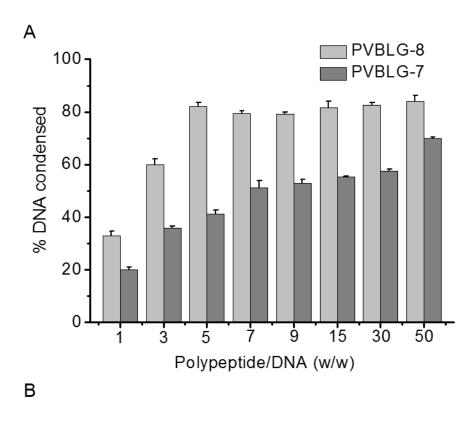
**Fig. S6.** GPC curve of PVBLG obtained from HMDS-initiated polymerization of VB-<sub>L</sub>-Glu-NCA at the M/I ratio of 150/1. The MW and PDI of the resulting PVBLG were summarized in the figure.

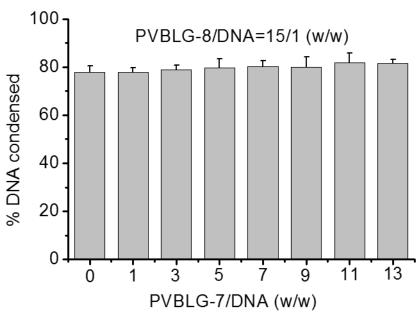


**Fig. S7.** Residue molar ellipticity at 222 nm of PVBLG-8 and PVBLG-7 at different pH (A) and NaCl concentrations (B).

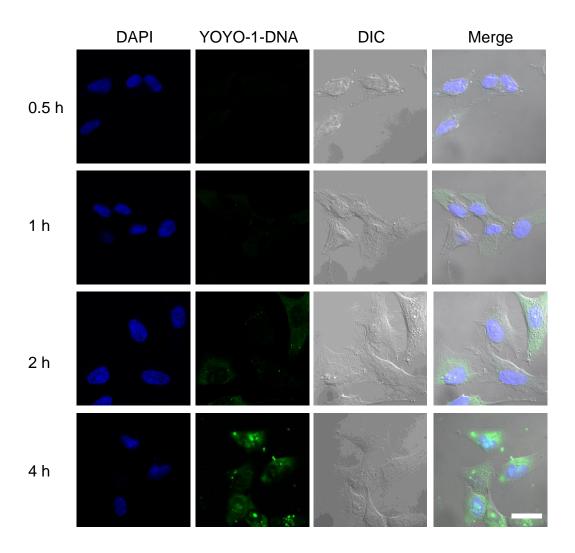


**Fig. S8.** CD spectra of PVBLG-8/PVBLG-7 mixtures and their ternary complexes with DNA in DI water at pH 7.

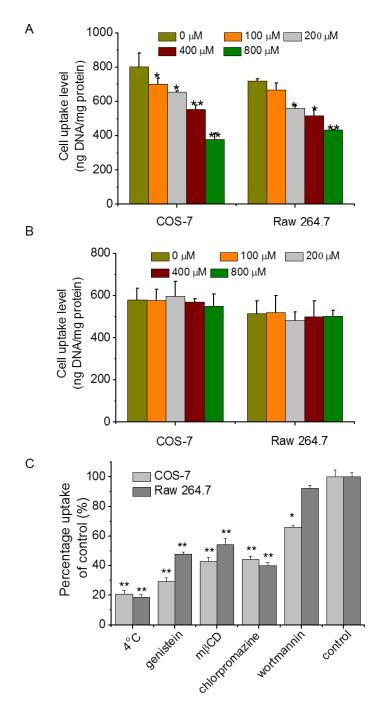




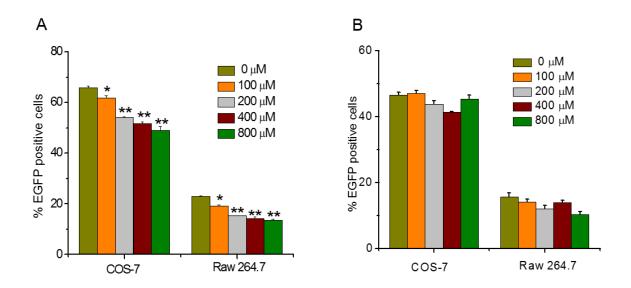
**Fig. S9.** EB exclusion assay showing the condensation of DNA in the binary complexes (A) and ternary complexes (B) PVBLG-8/DNA weight ratio fixed at 15/1 (n=3).



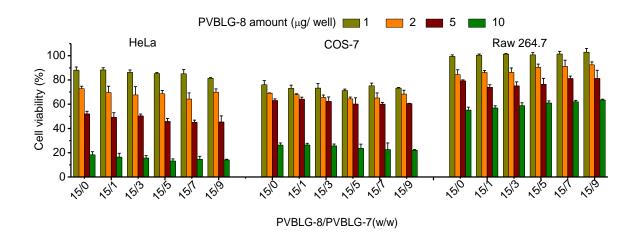
**Fig. S10.** CLSM images showing the cellular internalization of ternary complexes (PVBLG-8/PVBLG-7/DNA weight ratio of 15/7/1) in HeLa cells following incubation at 37 °C for 0.5, 1, 2, and 4 h (bar =  $20 \mu m$ ).



**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).