Development of a Low Toxicity, Effective pDNA Vector Based on Non-covalent Assembly of Bioresponsive Amino-β-Cyclodextrin:Adamantane-Poly(vinyl alcohol)-Poly(ethylene glycol) Transfection Complexes

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Supporting information

Selective Complexation Analysis by ^{1}H NMR

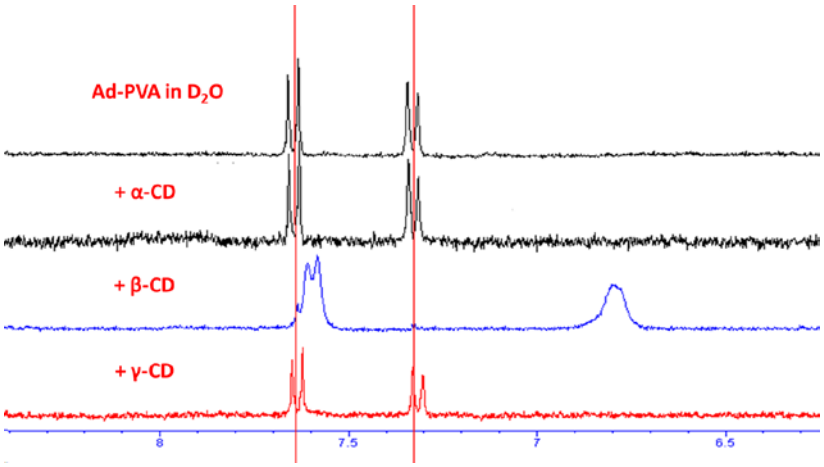

Figure 1s: 300 MHz ^{1}H NMR spectra of Ad-PVA with α-CD, β-CD and γ-CD in D_2O at 20 °C.

pDNA:Amino-β-CD$^+$:Ad-PVA-PEG Polyplex Characterization

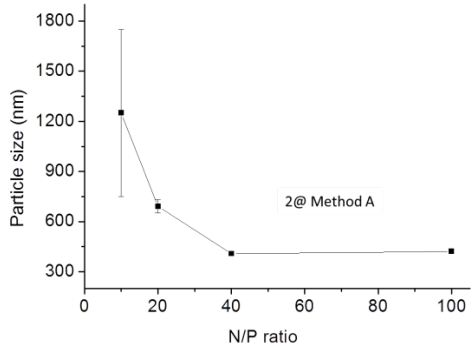

Figure 2s: Particle sizes of pDNA:2:Ad-PVA-PEG_{750} complexes formulated by Method A.

Formulation with Ad-PVA-PEG$_{750}$	Zeta Potentials (mV)				
	N:P = 5	N:P = 10	N:P = 15	N:P = 20	N:P = 30
1, Method B	-17 ± 1.3	-2.2 ± 0.1	1.3 ± 0.5	3.0 ± 0.8	3.1 ± 0.5
1 without Ad-PVA-PEG	-14.5 ± 0.5	-0.3 ± 0.2	2.6 ± 0.1	3.2 ± 0.6	2.8 ± 0.7
2, Method B	-2.3 ± 0.9	3.8 ± 1.1	4.1 ± 0.3	6.1 ± 0.2	6.3 ± 0.1
2 without Ad-PVA-PEG	-1.1 ± 0.1	4.1 ± 0.8	3.6 ± 0.3	7.2 ± 0.4	7.1 ± 0.2

Table 1s: Zeta potentials of formulations with Ad-PVA-PEG$_{750}$.

Amino-β-CD	Method of Formulation	Size		Zeta Potential
		Size (nm)	PDI	ζ (mV)
1	A	224.5	.39	-8.1 ± 0.1
	B	325.0	.40	-10.7 ± 0.7
2	A	192.6	.33	-6 ± 0.4
	B	280.8	.32	-6 ± 0.4
3	A	343.1	.36	-9.2 ± 0.5
	B	293.1	.30	-5.9 ± 0.6

Table 2s: Particle sizes and zeta potential measurements of pDNA:Amino-β-CD^{+}:Ad-PVA-PEG$_{2000}$ complexes at N:P = 20.

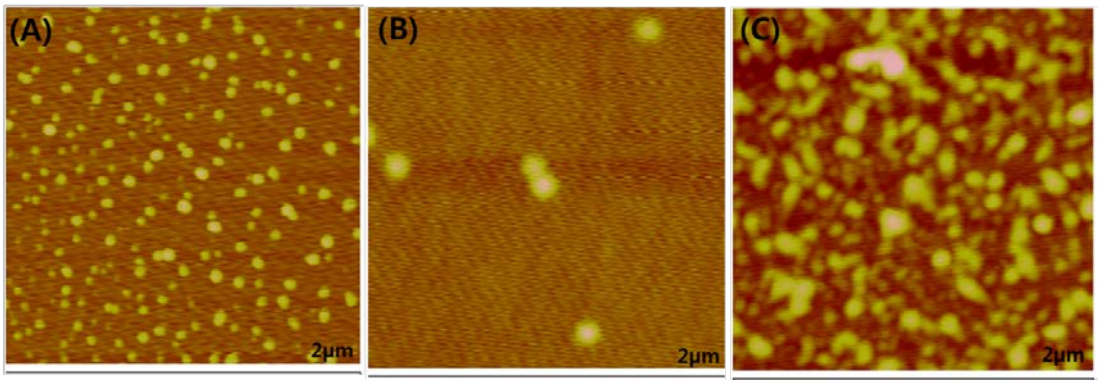

Figure 3s: AFM images of (A) Ad-PVA-PEG$_{2000}$; (B) 1:1 β-CD:Ad-PVA-PEG$_{2000}$; and pDNA:**3**:Ad-PVA-PEG$_{2000}$, prepared by Method B at (C) N:P = 2. The samples were prepared by adding a drop of solution to the mica surface and then slowly evaporating the sample at 25 °C overnight.

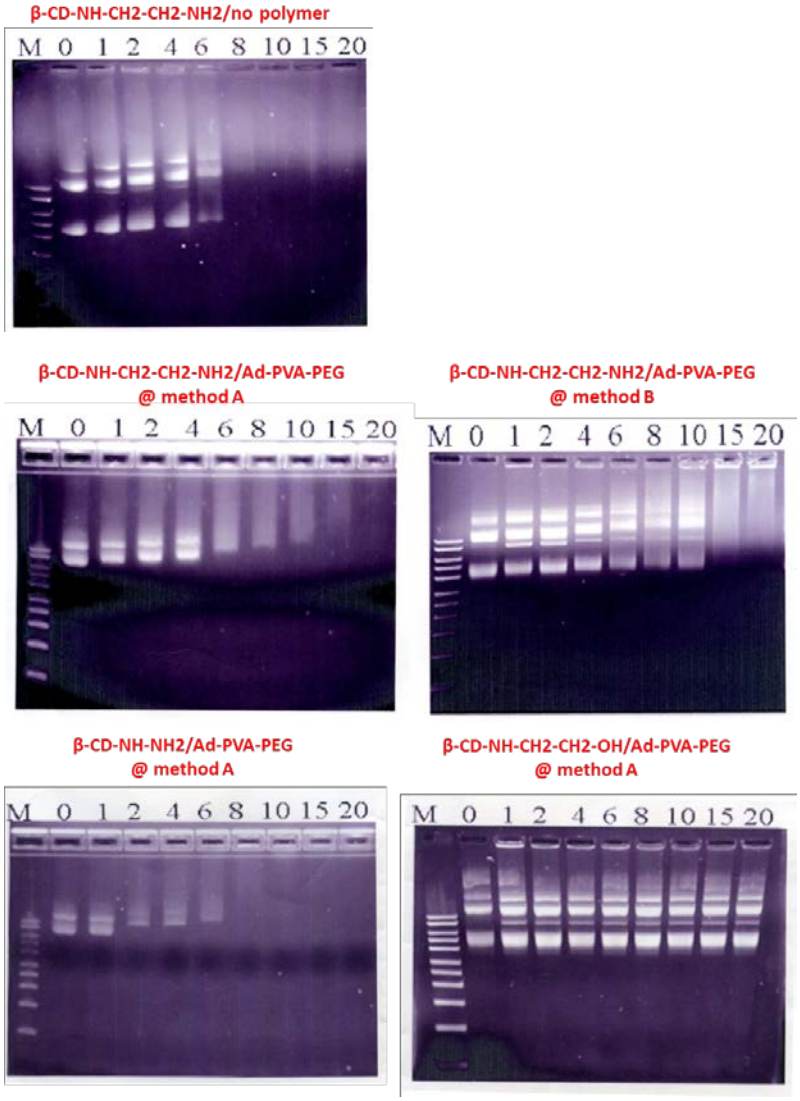

Figure 4s: Gel shift assay of amino-β-CD:Ad-PVA-PEG transfection complexes at various N:P ratios. Images showing pDNA condensation capabilities of **1**:no polymer, **1**:Ad-PVA-PEG @ Method A, **1**:Ad-PVA-PEG @ Method B, **3**:Ad-PVA-PEG @ Method A, and **2**:Ad-PVA-PEG @ Method A.

pDNA:Amino-β-CD^{+}:Ad-PVA-PEG Polyplex Transfection Performance

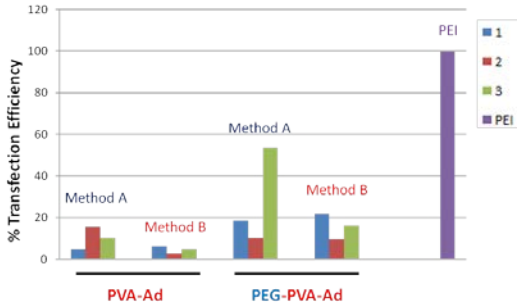

Figure 5s: In vitro gene transfection efficiency of the complexes of pDNA:amino-CD^{+}:Ad-PVA and Ad-PVA-PEG$_{750}$ relative to PEI (25K) at N:P = 20 in serum free media in HeLa cells.

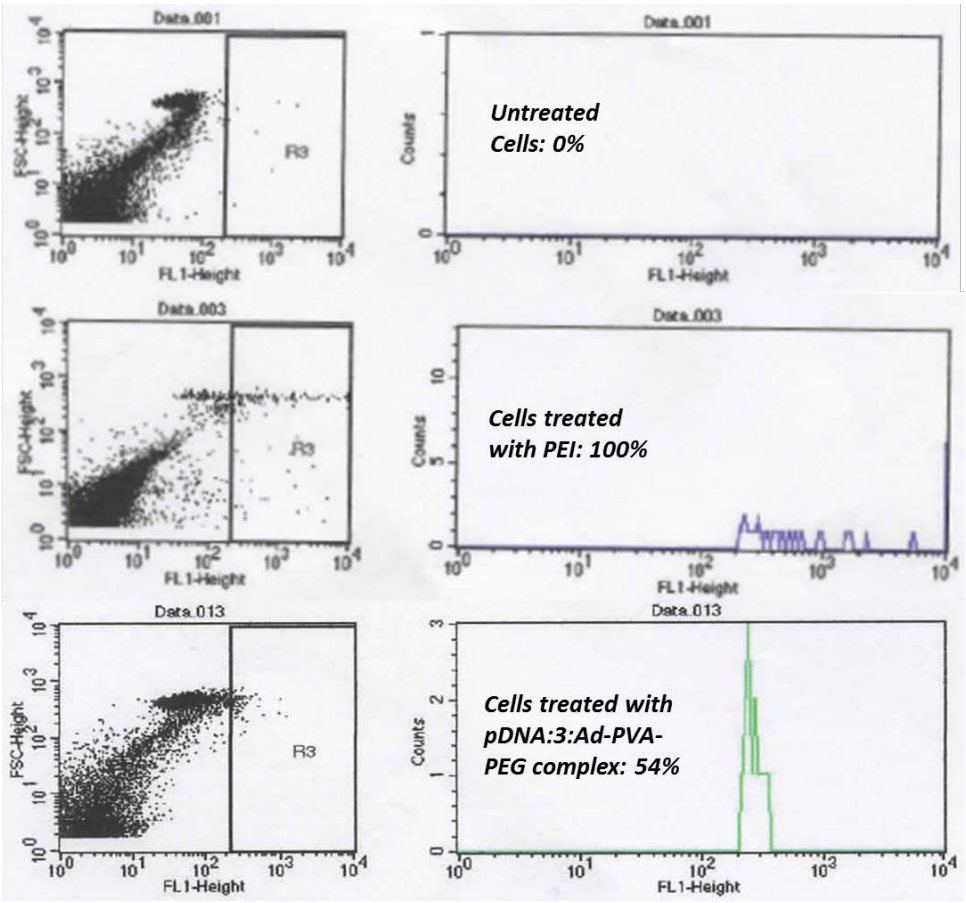

***Figure 6s*:** Flow cytometric analysis of plasmid DNA encoding mhGFP plasmid in HeLa cells. Comparison of complex of *p*DNA:**3**:Ad-PVA-PEG_{750} relative to pDNA:PEI (25kD) at N:P = 20 in serum free media using HeLa cells (2 μg/well pDNA).

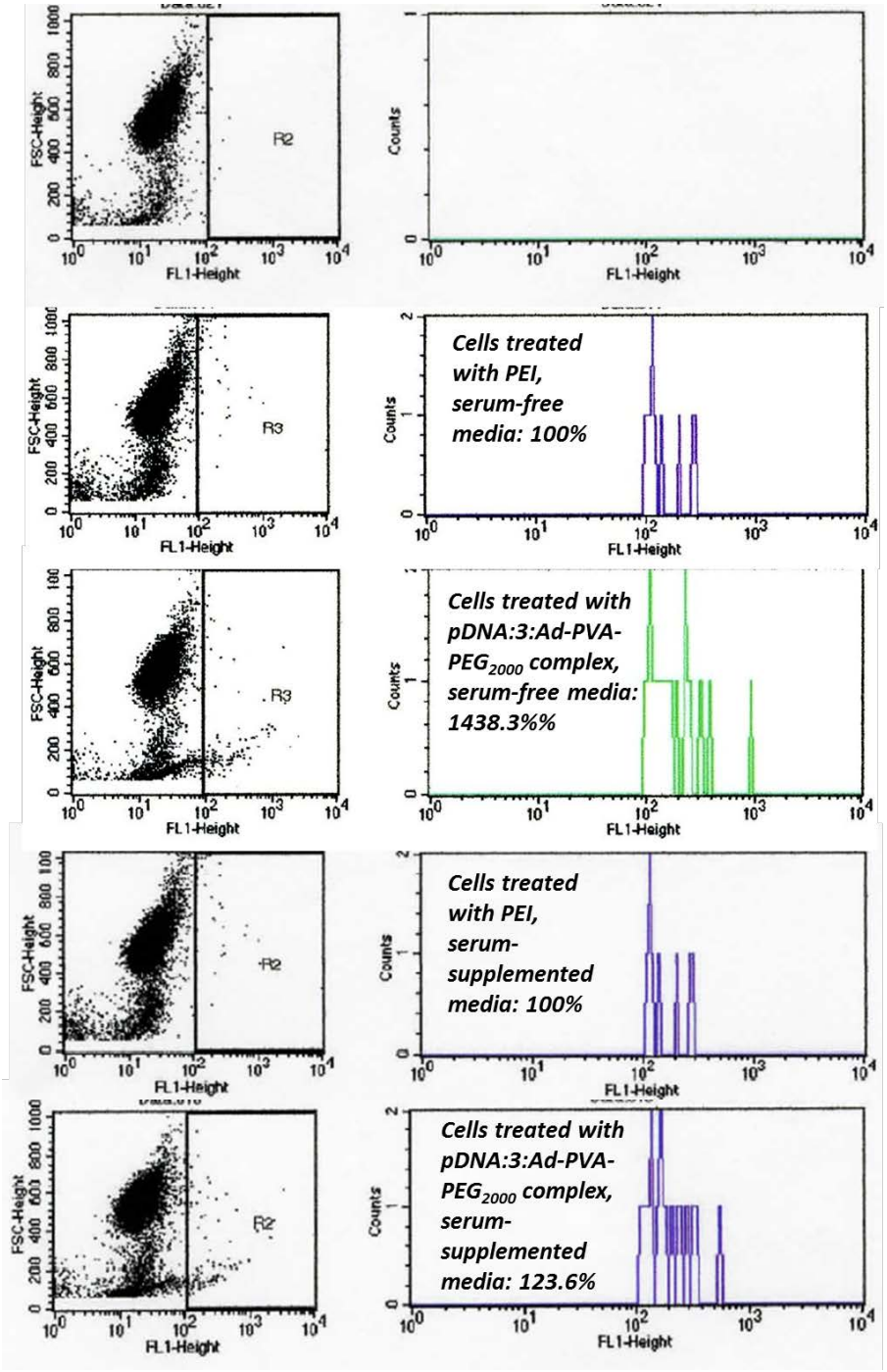

***Figure 7s*:** Flow cytometric analysis of plasmid DNA encoding mhGFP plasmid in HeLa cells. Comparison of complex of pDNA:**3**:Ad-PVA-PEG_{2000} relative to pDNA:PEI (25kD) at N:P = 20 in serum-free and serum-supplemented media using HeLa cells (2µg/well pDNA).