SUPPLEMENTARY INFORMATION

Oligoamine-tethered low generation polyamidoamine dendrimers as potential nucleic acids carriers

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Materials

Ethylenediamine-cored polyamidoamine (PAMAM) dendrimers generation 2, 3 and 4 (G2, G3 and G4) with molecular weights 3256, 6909 and 14215 Da, respectively, 2,4,6-trinitrobenzene sulfonic acid (TNBS), 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT), ethidium bromide and Dulbecco's Modified Eagle Medium (DMEM) were procured from Sigma-Aldrich Chemical Company (USA). Lipofectamine 2000 (cationic lipid-based highly efficient transfection reagent) and Superfect (heat-fractured form of G5-PAMAM dendrimer as competent transfection reagent) were purchased from Invitrogen (USA) and Qiagen (France), respectively. Transfection assays were carried out with the plasmid encoding enhanced green fluorescent protein (EGFP) (Clontech Inc., USA) under the cytomegalovirus (CMV) immediate early promoter. The plasmid was transformed into *E. coli* bacterial strain DH5α and extracted from the culture pellets using the Qiagen Maxi-prep Kit (France) as per manufacturer's instructions. Other analytical grade chemicals and reagents, used in the present study, were obtained locally.

Cell culture

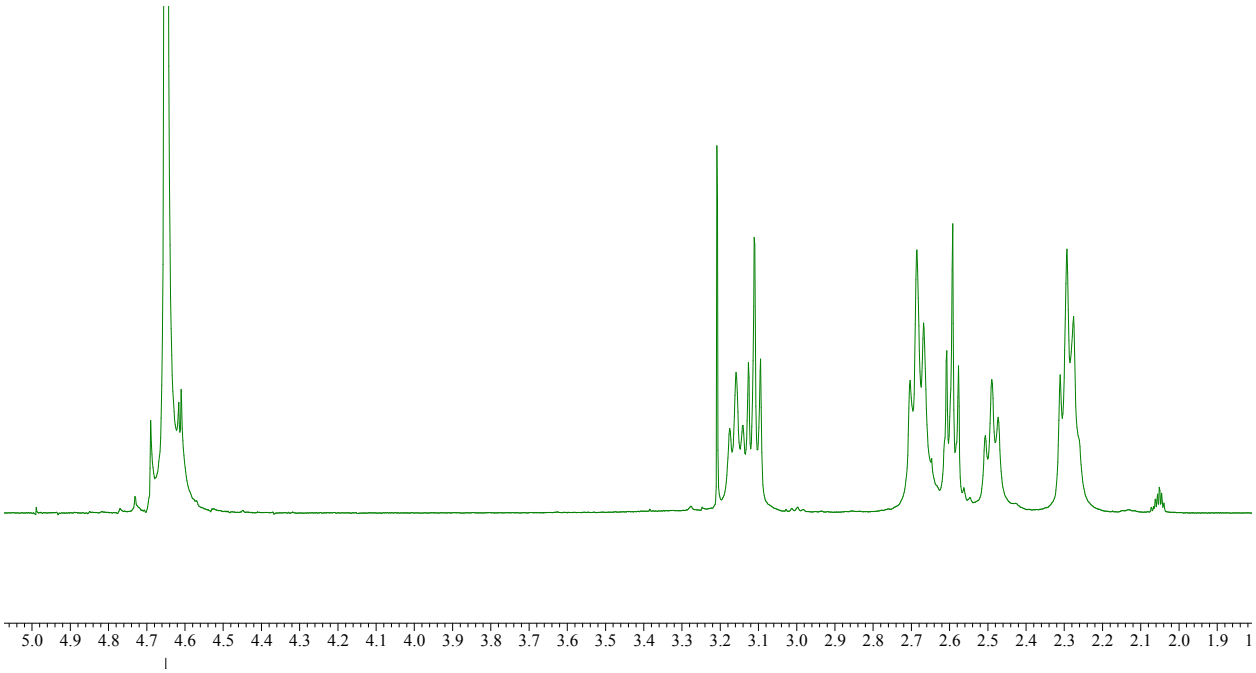
HeLa (Human cervical carcinoma cell line) and A549 (Human alveolar adenocarcinoma cell line) were obtained from National Centre for Cell Science, Pune, India and grown in DMEM containing 1% antibiotic cocktail of streptomycin and penicillin supplemented with 10% heat inactivated fetal bovine serum (FBS) (GIBCO-BRL-Life Technologies, UK) at 37 ^{o}C in a humidified 5% CO_2 incubator.

Particle size, morphology and zeta potential measurements

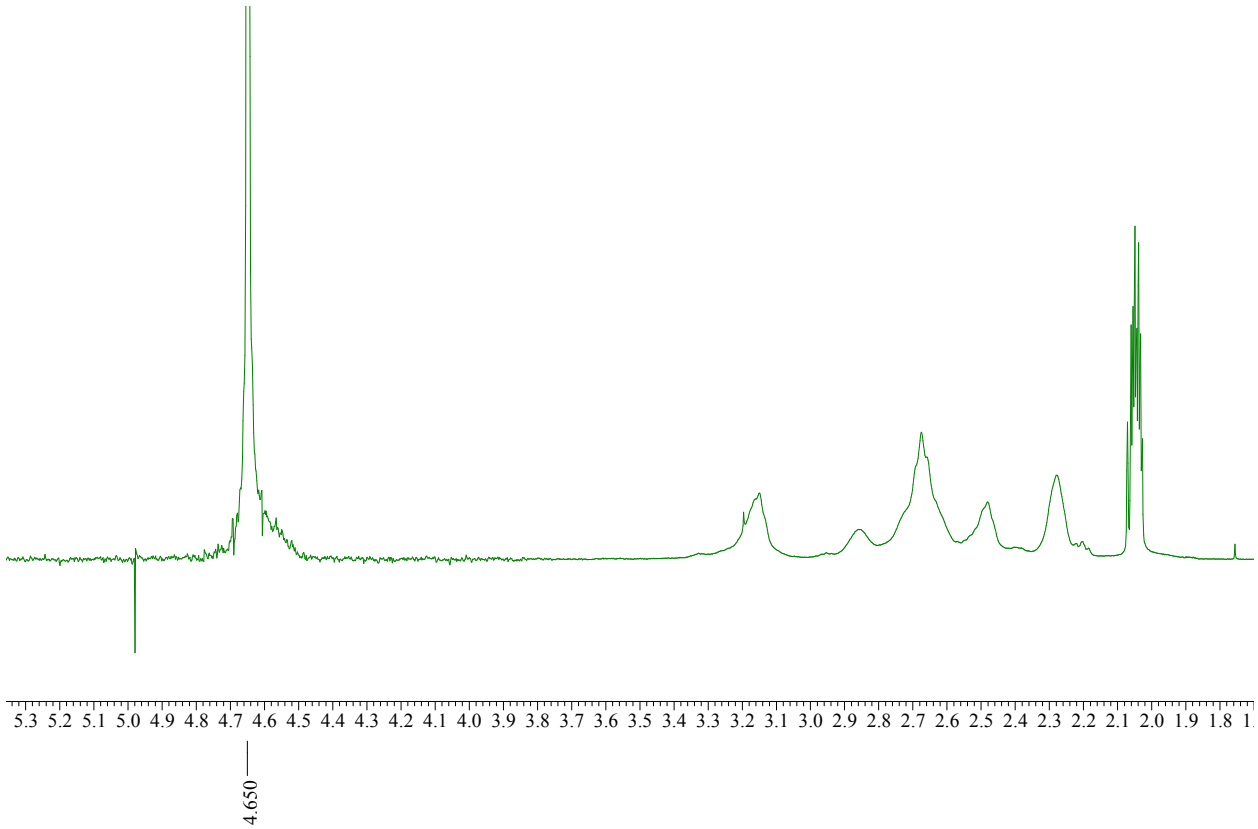
Size and zeta potential measurements of pDNA complexes of native and modified dendrimers were carried out by dynamic light scattering (DLS) using Zetasizer Nano-ZS (Malvern Instruments, UK). The measurements were performed in water and 10% FBS. The data analysis was performed in automatic mode and presented as the average value of 20 or 30 runs. Morphology and size of the complexes were also examined on high resolution transmission electron microscopy (HR-TEM) (Technai G2 30U-twin, Technai 200 kV ultratwin microscope, operating at 200 kV). The samples were prepared at their best working w/w ratio in water, deposited on carbon-coated copper grids and negatively stained with 1% uranyl acetate. The image was captured at an accelerating voltage of 200 kV.

^{1}H-NMR spectra of native and modified dendrimers

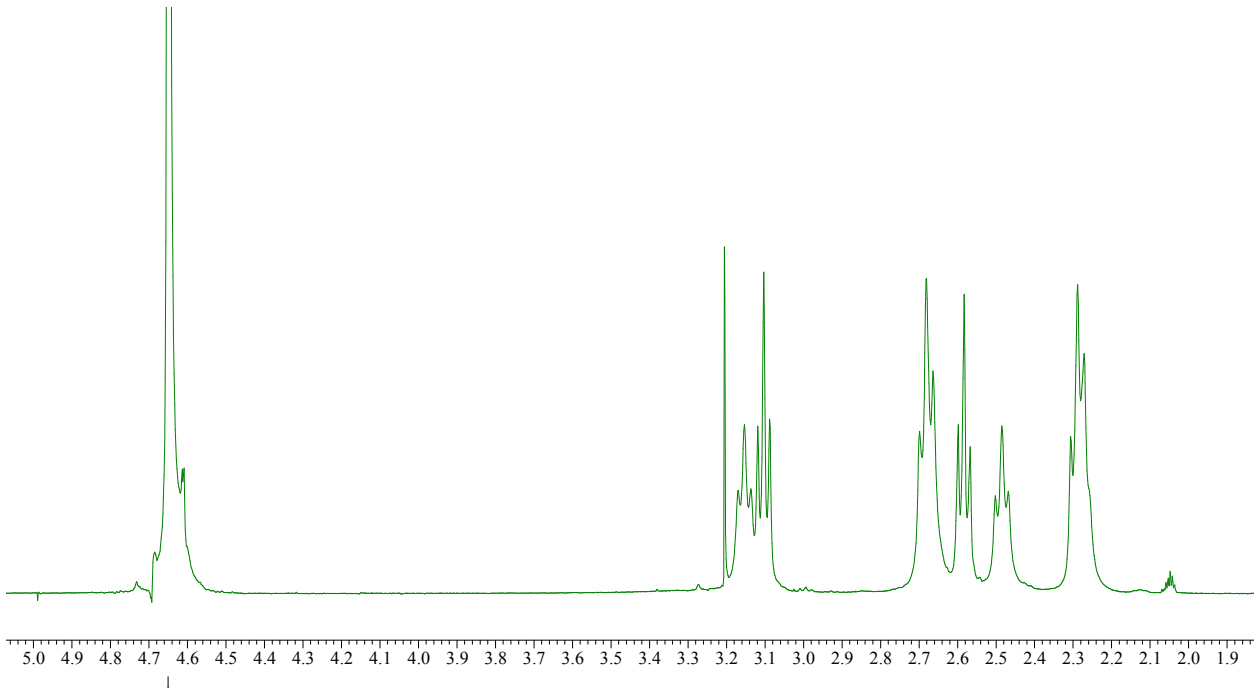
G2-PAMAM



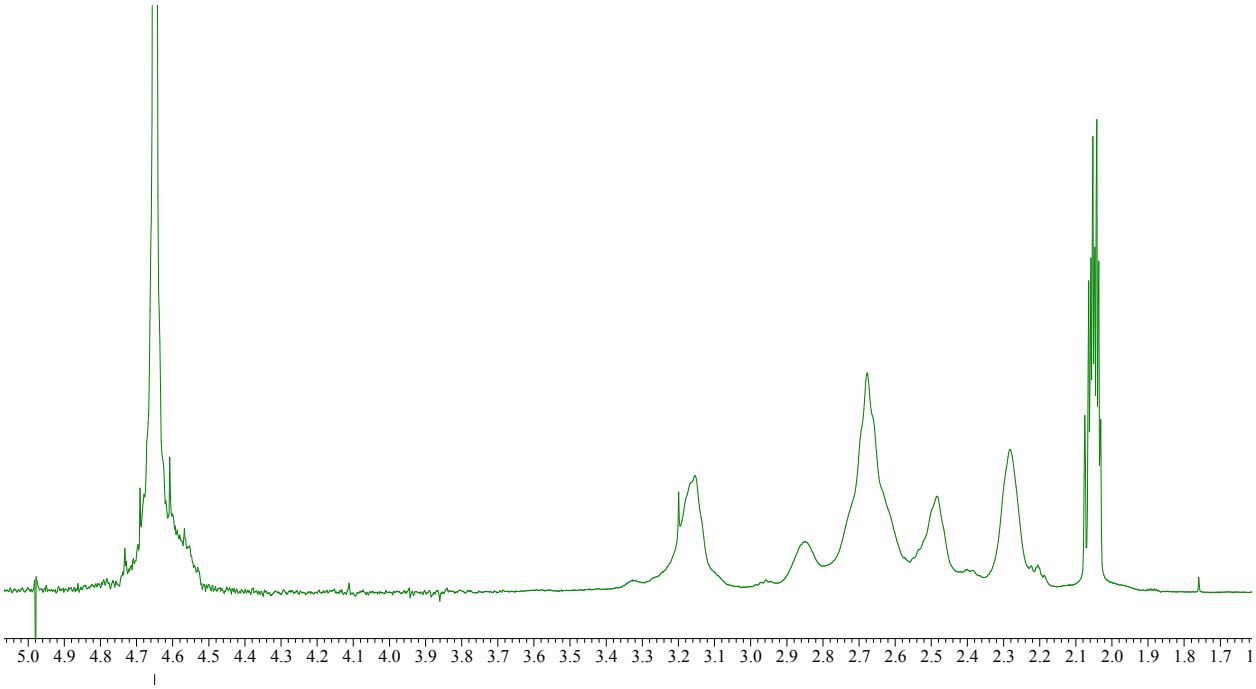
mG2-PAMAM



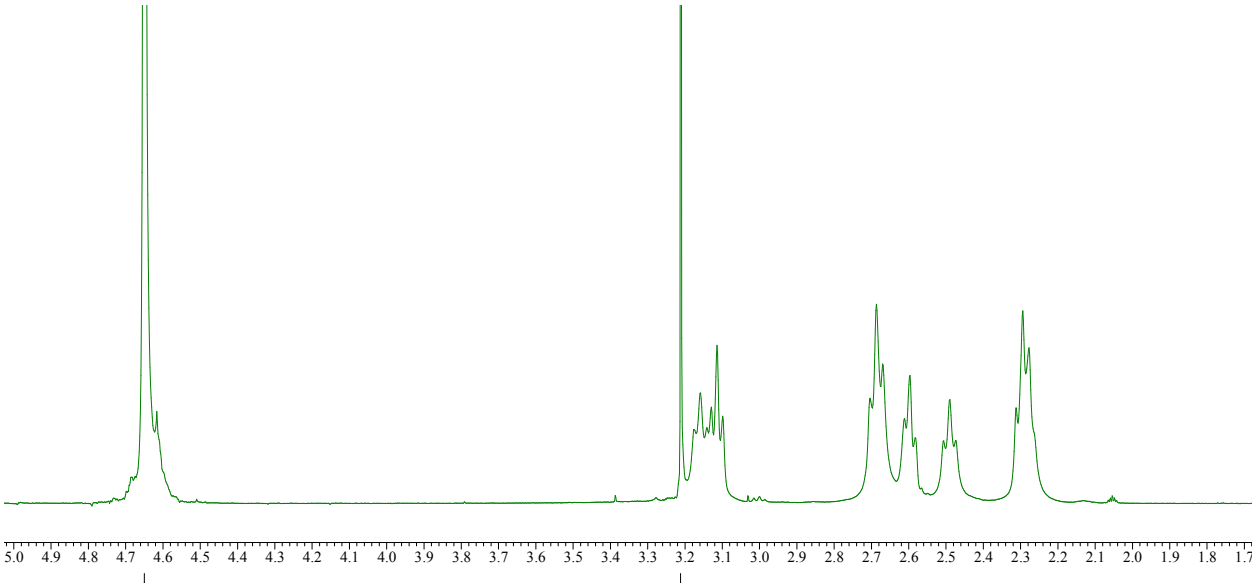
G3-PAMAM



mG3-PAMAM



G4-PAMAM



mG4-PAMAM

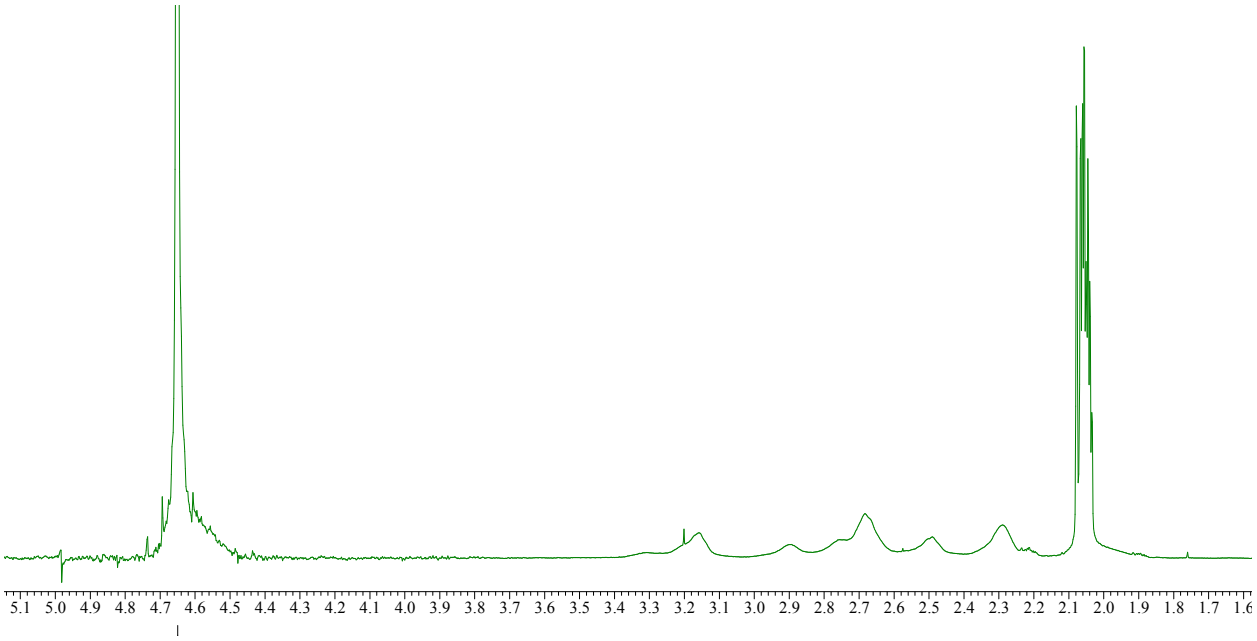


Figure S1. ^{1}H-NMR spectra of (i) G2 (ii) mG2 (iii) G3 (iv) mG3 (v) G4 and (vi) mG4 (in D_2O).

Table S1. Determination of dissociation constant (K_d) of binding from gel retardation assay

Dendrimer	G2	G3	G4	mG2	mG3	mG4
K_d (μM)	630	316	141	141	98	3

Table S2. IC50 values of native and modified PAMAM dendrimers/pDNA complexes in HeLa cells

S. No.	Dendrimers	IC50 value (μg/ml)
1	G2	123.1
2	G3	76.2
3	G4	74.1
4	mG2	122.4
5	mG3	79.8
6	mG4	72.2