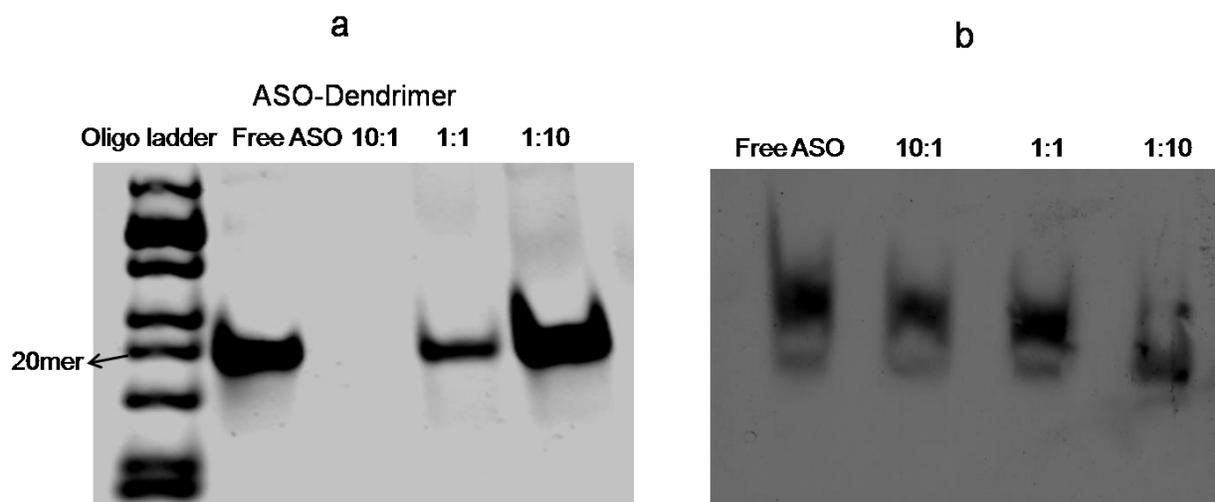
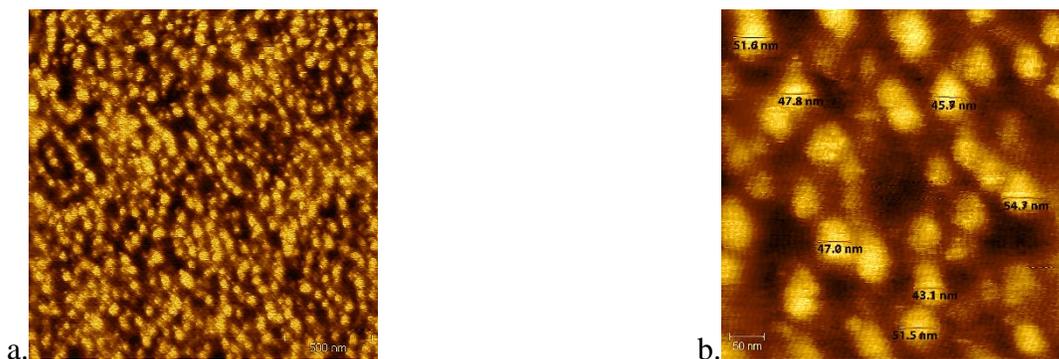


Topical Gene Silencing by iontophoretic delivery of antisense oligonucleotide-dendrimer nanocomplex: Proof of concept in a skin cancer mouse model

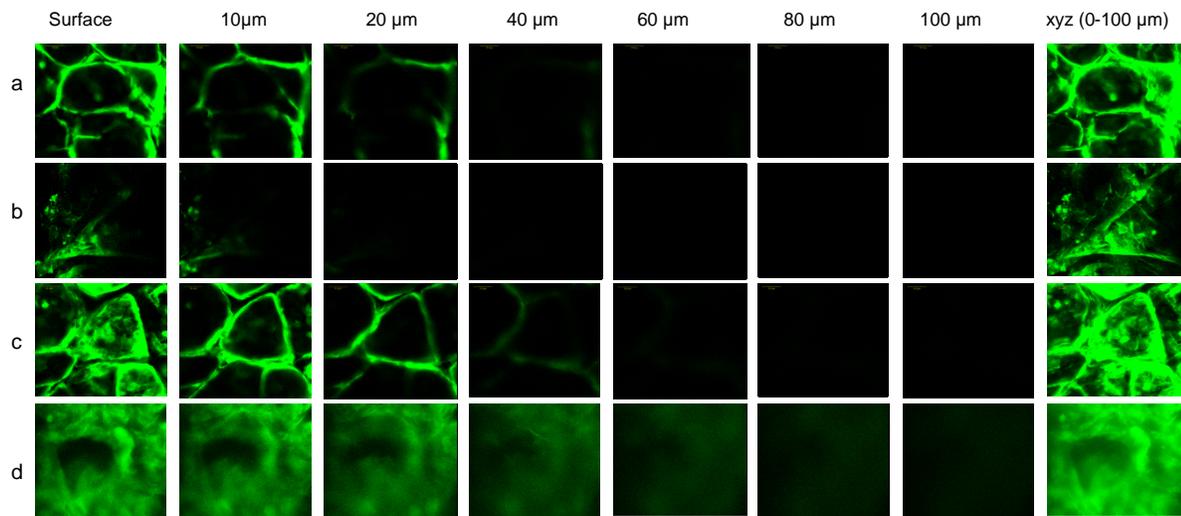
Venkata Vamsi K. Venuganti, Manju Saraswathy, Chandradhar Dwivedi, Radhey S. Kaushik,
Omathanu P. Perumal



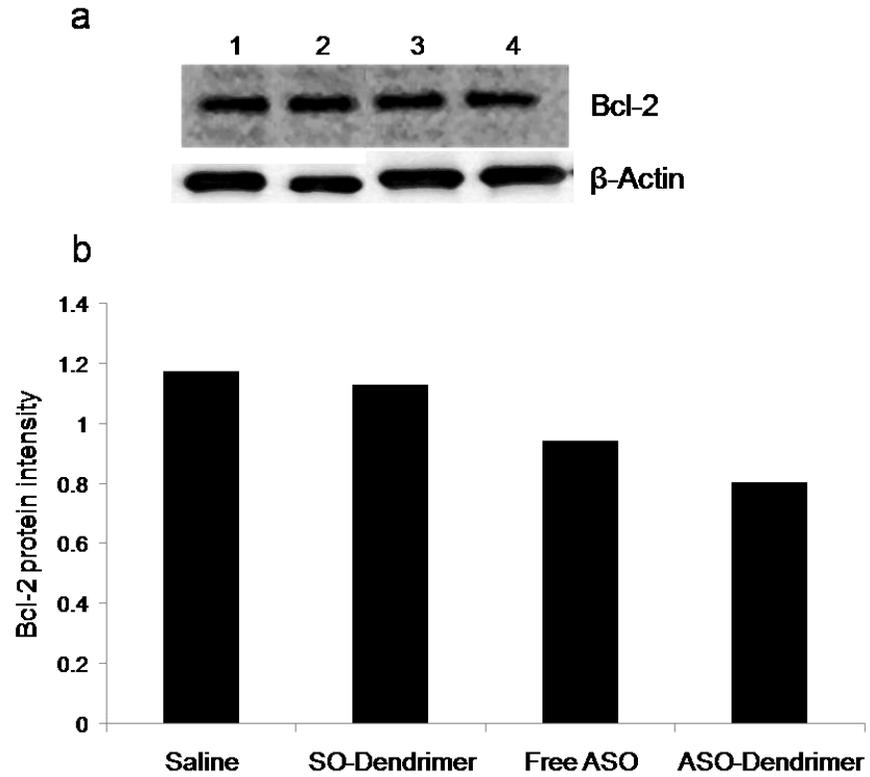
Supplementary Fig. S1. PAGE analysis of ASO-dendrimer complexes (a). Bands from ASO-dendrimer complexes after incubation with dextran sulfate for 4hrs (b). Complexes were formed with different N/P ratios and the gel was stained with SYBR gold.



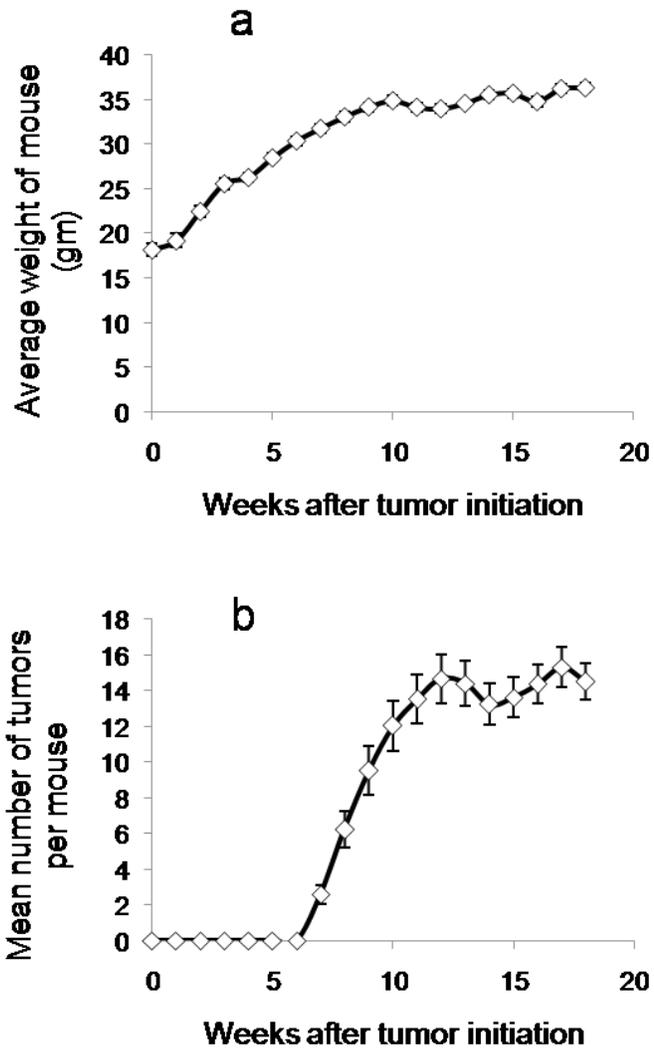
Supplementary Fig. S2. AFM images of Dendrimer-Bcl2 ASO complex (10:1). a) AFM image at lower magnification (scale is 500nm). b) AFM image at higher magnification (scale is 50nm). The dendrimer-ASO complex solution in deionized water was deposited on a mica surface and dried under a flow of nitrogen for the AFM study. The diameter of the complex is shown on the right hand panel.



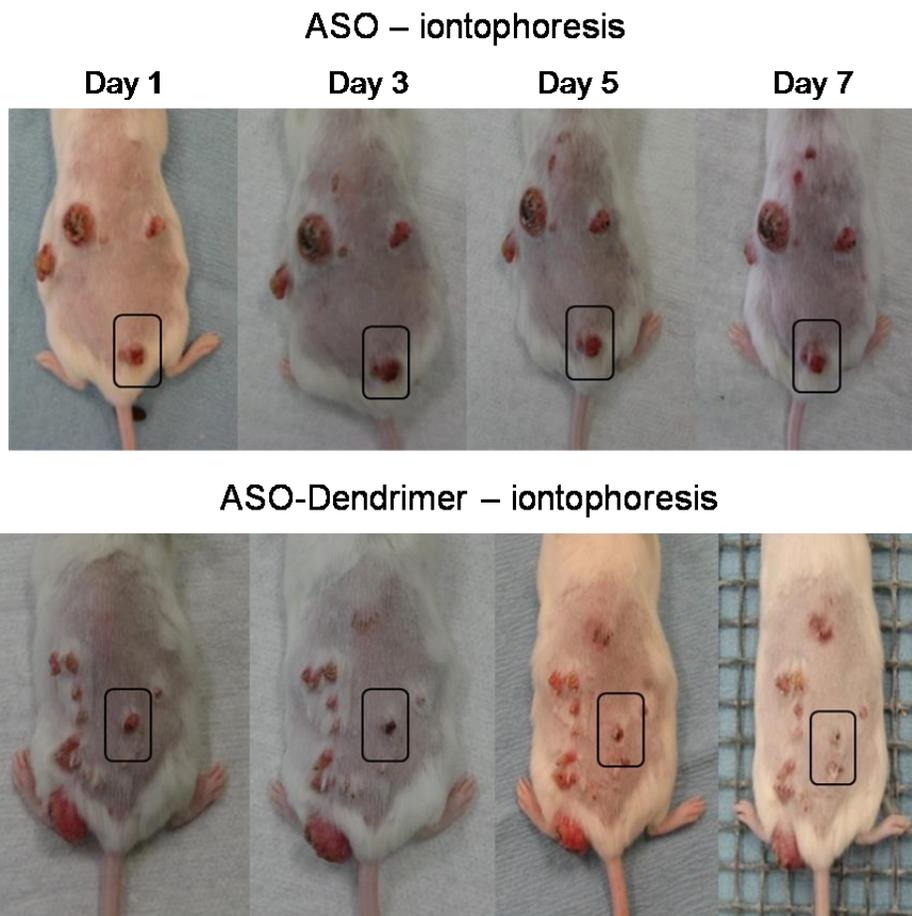
Supplementary Fig. S3. CLSM images of porcine skin in the xyz plane after 4hrs treatment. (a) Passive delivery of free ASO; (b) iontophoretic delivery of free ASO; (c) passive delivery of ASO-dendrimer complex; (d) iontophoretic delivery of ASO-dendrimer complex. A current density of $0.3\text{mA}/\text{cm}^2$ was applied for 4hrs. Optical sections are shown in xyz plane from the skinsurface to a depth of $100\ \mu\text{m}$.



Supplementary Fig. S4. Bcl-2 protein expression in A431 cells after treatment with PBS (1); SO-dendrimer complex (2); free ASO (3) and ASO-dendrimer complex (4). A) A431 cells were treated for 2 hrs with 1 μ M of ASO and a representative gel band is shown. b) The values represents the relative intensity of Bcl-2 band with respect to β -actin (control) band. Each value represent average of two experiments.



Supplementary Fig. S5. Animal weight (a) and tumor number (b) in mice as a function of time after tumor initiation. Values represent mean \pm SE (n = 30).



Supplementary Fig. S6. Photographs of mouse treated from days 1 to 7. A single tumor (indicated by rectangular area) was passively or iontophoretically (0.5 mA/cm^2 , 2hrs) treated with ASO-dendrimer complex on days 1, 3, 5 and 7. Images are representative of five experiments. A gradual reduction in tumor volume is seen in the iontophoretic delivery group.

Supplementary Table S1: Treatment groups for *in vivo* studies

Group	Treatment
Group I	DMBA+TPA+saline intradermal injection
Group II	DMBA+TPA+SO intradermal injection
Group III	DMBA+TPA+ASO intradermal injection
Group IV	DMBA+TPA+ASO-dendrimer complex solution intradermal injection
Group V	DMBA+TPA+ASO
Group VI	DMBA+TPA+ASO+iontophoresis
Group VII	DMBA+TPA+ASO-dendrimer complex
Group VIII	DMBA+TPA+ASO-dendrimer complex +iontophoresis

DMBA - 7,12-dimethyl benzanthracene, TPA - 12-O-tetradecanoylphorbol 13-acetate