Electronic Supplementary Material (ESI) for Journal of Materials Chemistry B.

Guanidine Modified Polyethyleneimine-g-Polyethylene Glycol Nanocarriers for Long Interfering RNA (liRNA) based Advanced Anticancer Therapy

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Supporting information

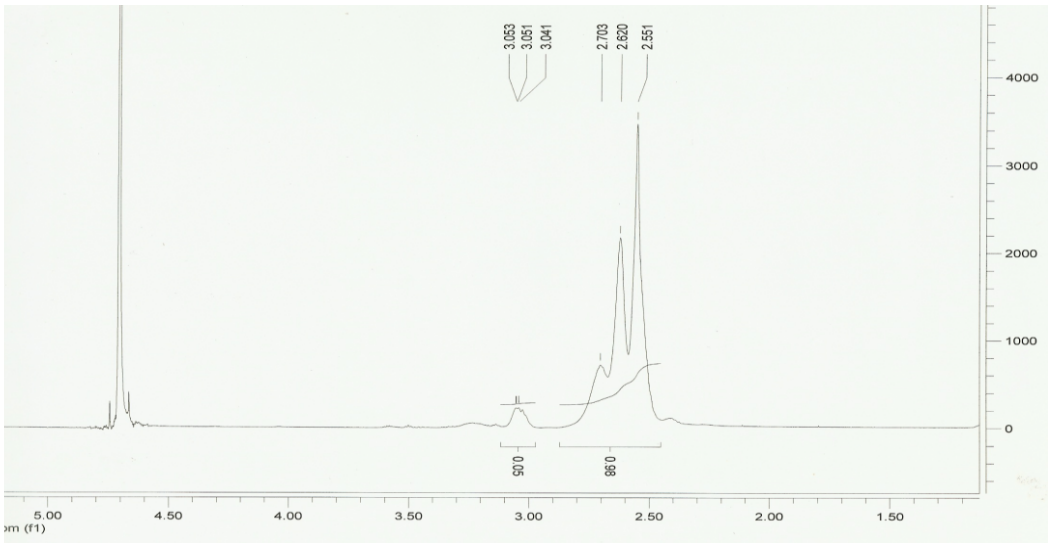
Table S1- Quantification of degree of guanidination in bPEI-g-PEG

Sample code	% of GU*
bPEI-g-PEG	-
GU bPEI-g-PEG 1	-
GU bPEI-g-PEG 2	6.7
GU bPEI-g-PEG 3	12.9
GU bPEI-g-PEG 4	16.2
GU bPEI-g-PEG 5	21.72

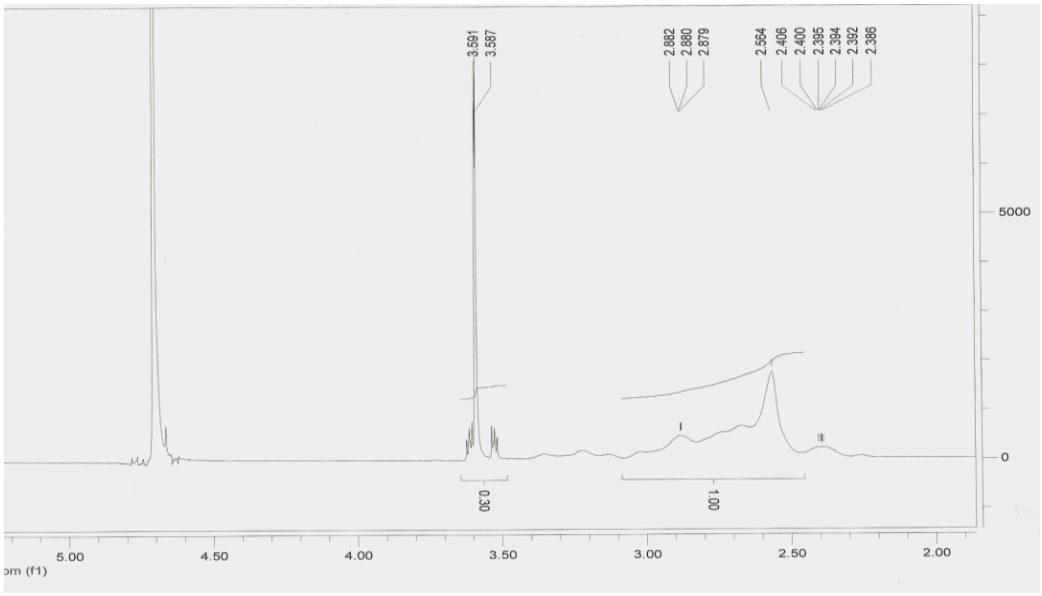
*GU content in the polymer was determined by Sakaguchi assay

Table S2 – IC 50 values for Dox in HeLa/U87MG cells

Cells	**IC-50 (µM) Control cells (Dox alone)**	**IC-50 (µM) siRNA treated cells (siRNA+Dox)**	**IC-50 (µM) liRNA treated cells (liRNA+Dox)**
U87MG	1.929	2.78	0.779
HeLa	1.107	0.902	0.203



A



B

Figure S1- H1-NMR spectra of (A) bPEI and (B) bPEI-g-PEG. 1H NMR spectra was obtained with Bruker Ascend™ 500 NMR using D_2O as solvent.

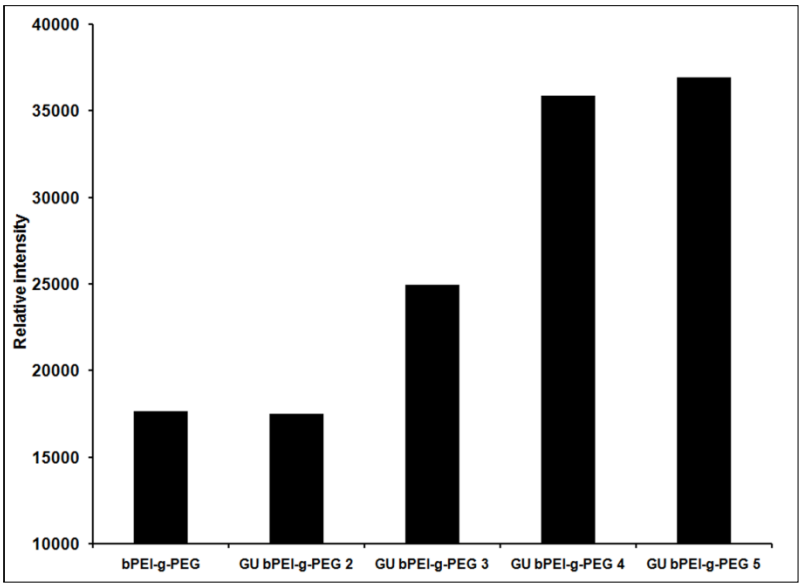


Figure S2- NC-3000 based quantification of liRNA uptake in U87MG cells using bPEI-g-PEG with varying degree of guanidination. Cells were treated with 100 nM liRNA complex (N/P 5) for 3h and intracellular Cy3 intensity was quantified by NC-3000 method

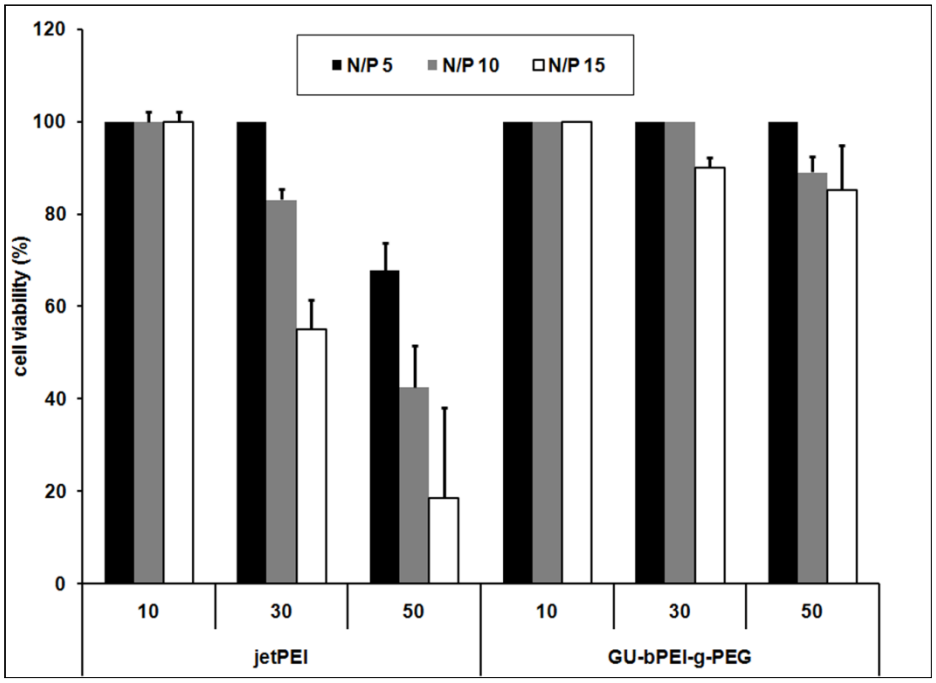


Figure S3- liRNA induced toxicity under transfection conditions. HeLa cells were treated with liRNA complex at different N/P ratios (5,10 and 15) and RNA concentration (10,30 and 50 nM). jetPEI and GU bPEI-g-PEG were used as the transfection reagents. Cell viability was assessed immediately after transfection by MTT assay.

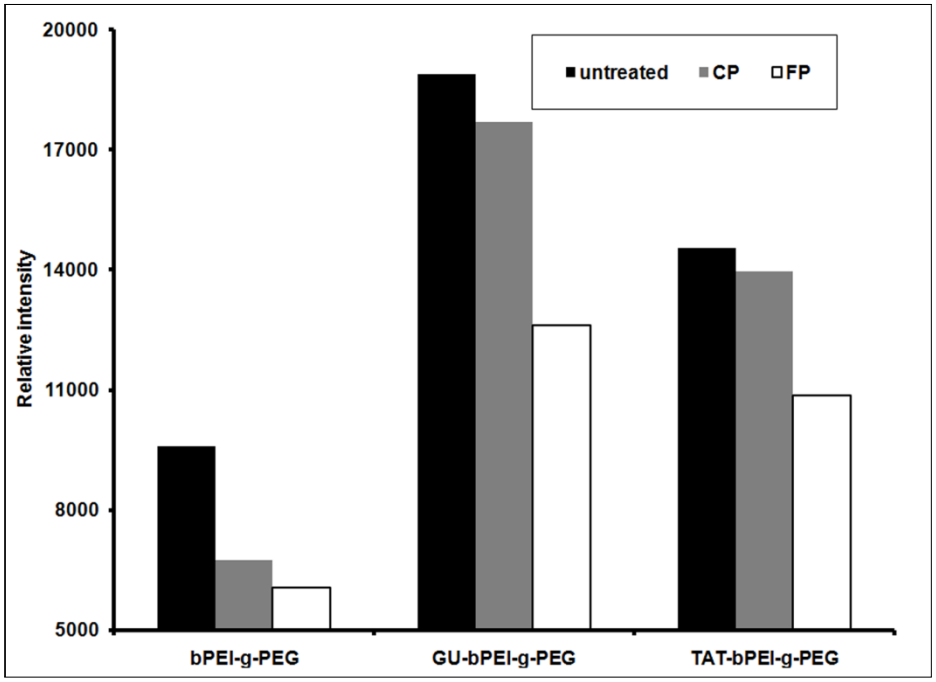


Figure S4- Effect of endocytic inhibitors on the cellular uptake liRNA complex. HeLa cells used for these studies were pretreated with chlorpromazine (CP, 2 μg/mL) to block clathrin mediated endocytosis and filipin III (FP, 5 μg/mL) to inhibit caveolae mediated endocytosis. Cells were then transfected with 50 nM liRNA complex with either bPEI-g-PEG, GU bPEI-g-PEG or TAT bPEI-g-PEG at N/P 5. Cellular uptake of liRNA was then compared between treated and untreated cells by NC-3000 method.