

## Supplementary Information for

# A Self-assembling Polycationic Nanocarrier that Exhibits Exceptional Gene Transfection Efficiency

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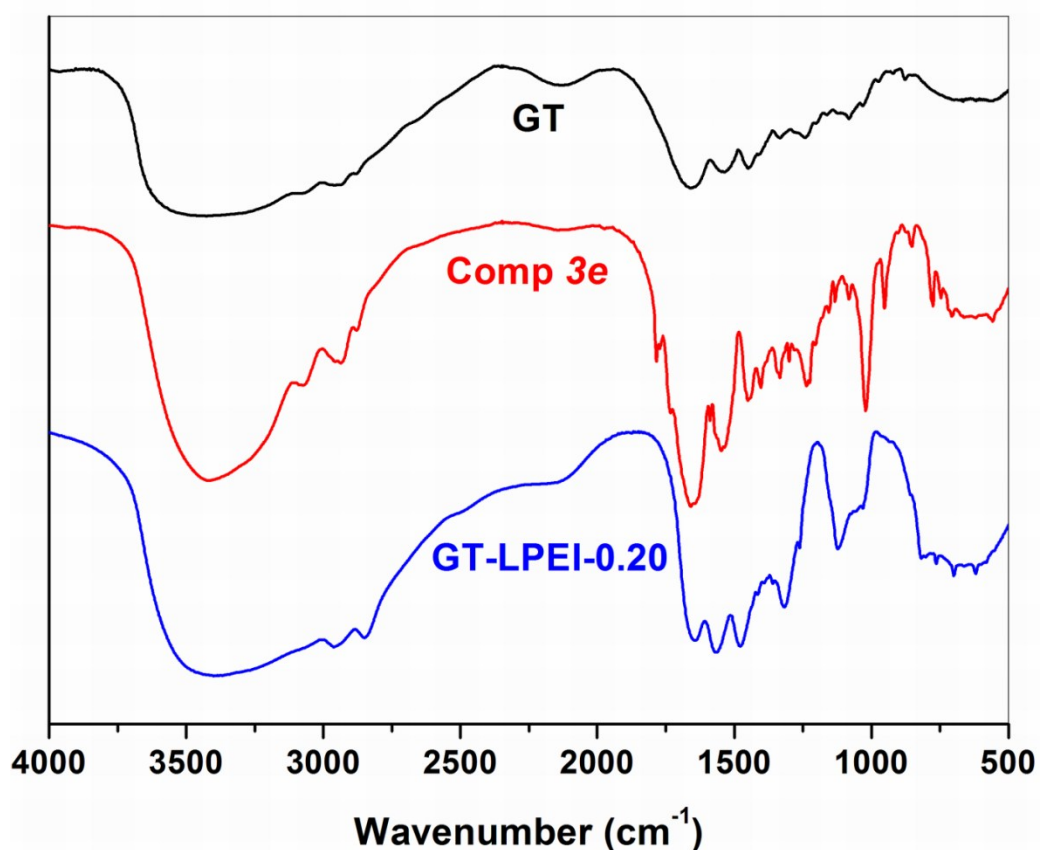
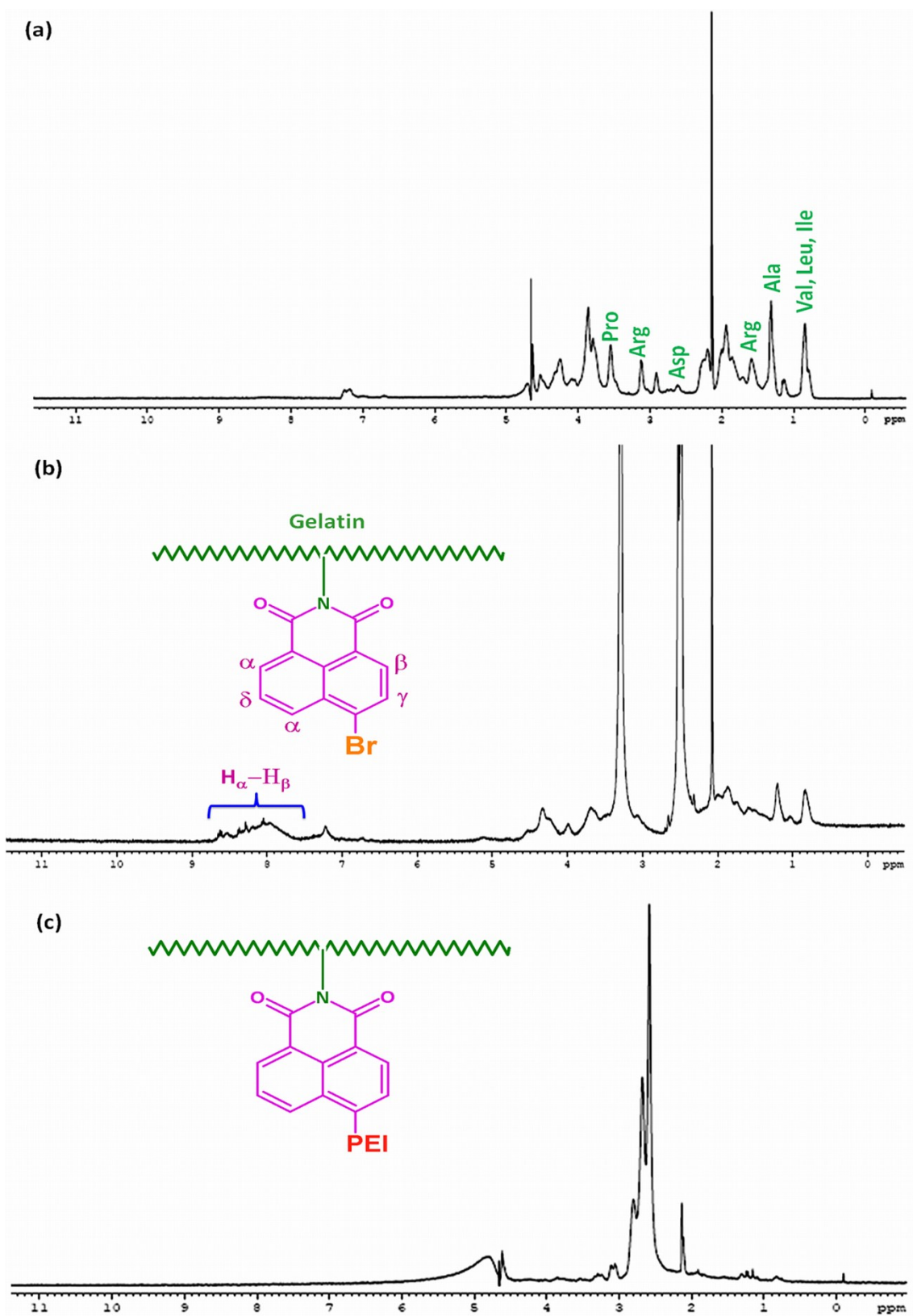
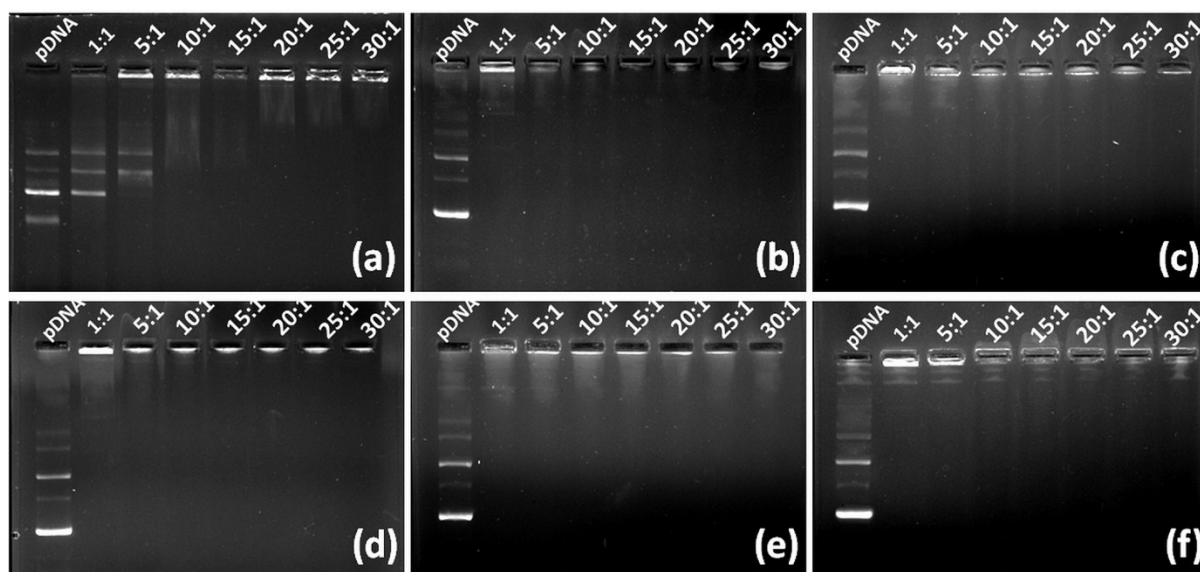


Figure S1. FTIR spectra of GT, Comp 3e and GT-LPEI-0.20.

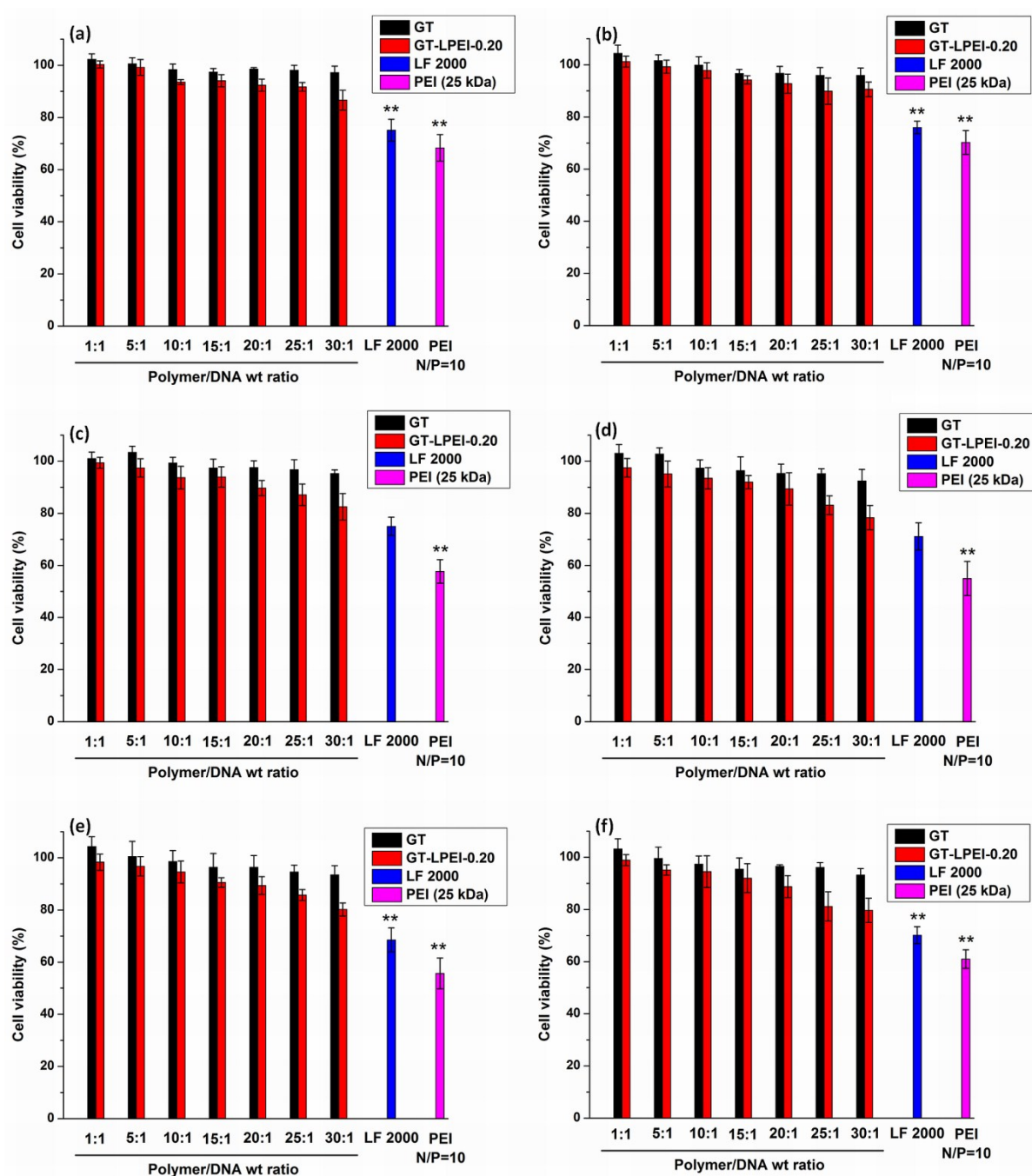




**Figure S3.** Image of the agarose gel after electrophoresis for (a) GT/pDNA, (b) GT-LPEI-0.01/pDNA, (c) GT-LPEI-0.06/pDNA, (d) GT-LPEI-0.12/pDNA, (e) GT-LPEI-0.18/pDNA and (f) GT-LPEI-0.20/pDNA complexes at different polymer: pDNA weight ratios of 1:1, 5:1, 10:1, 15:1, 20:1, 25:1 and 30:1.

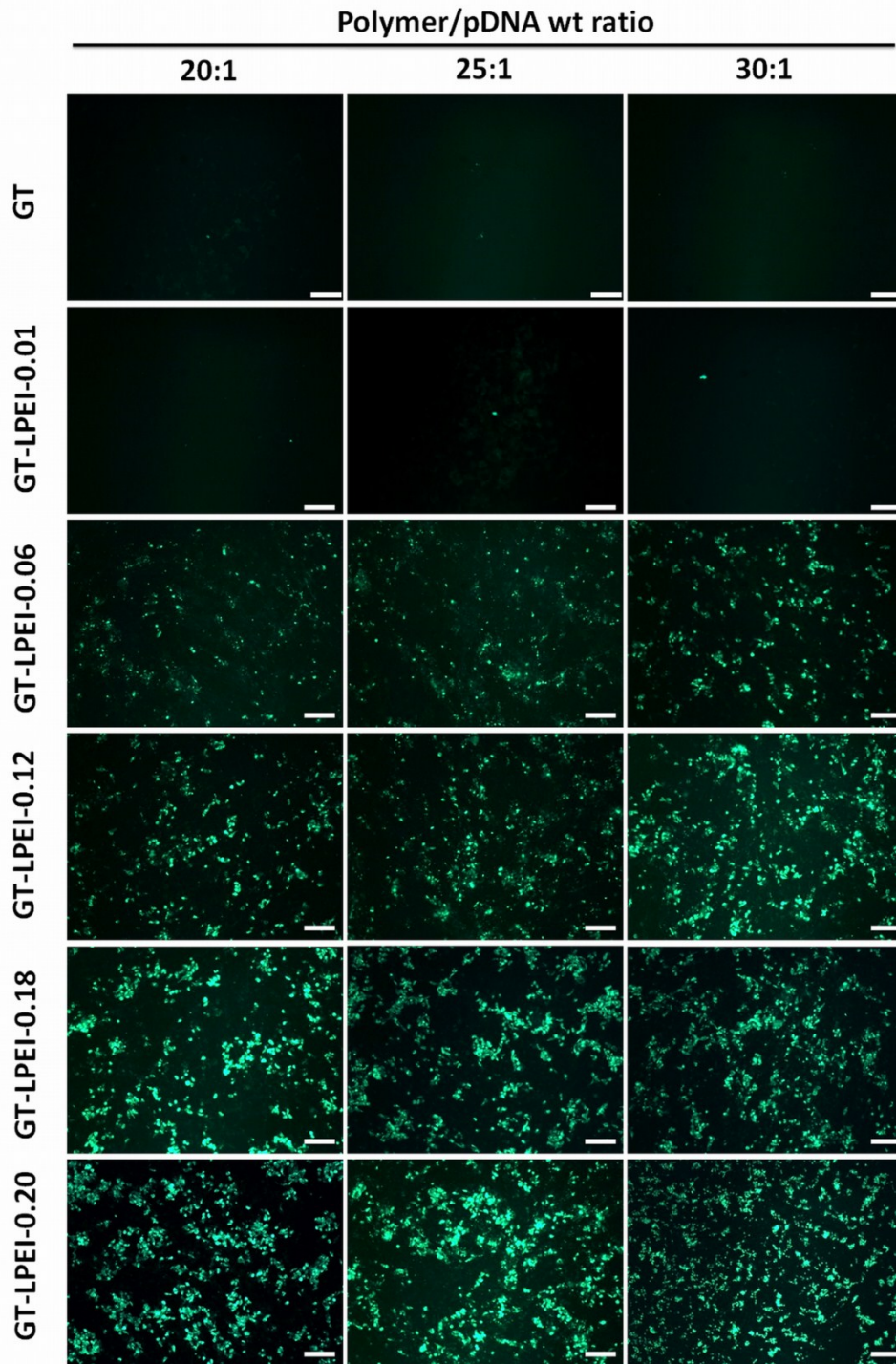


**Figure S5.** *In vitro* toxicity of GT and different GT-LPEIs at different concentrations in HeLa cells after 24 h. LPEI (2 kDa) and PEI (25 kDa) were used as positive controls and untreated cells were used as the negative control.

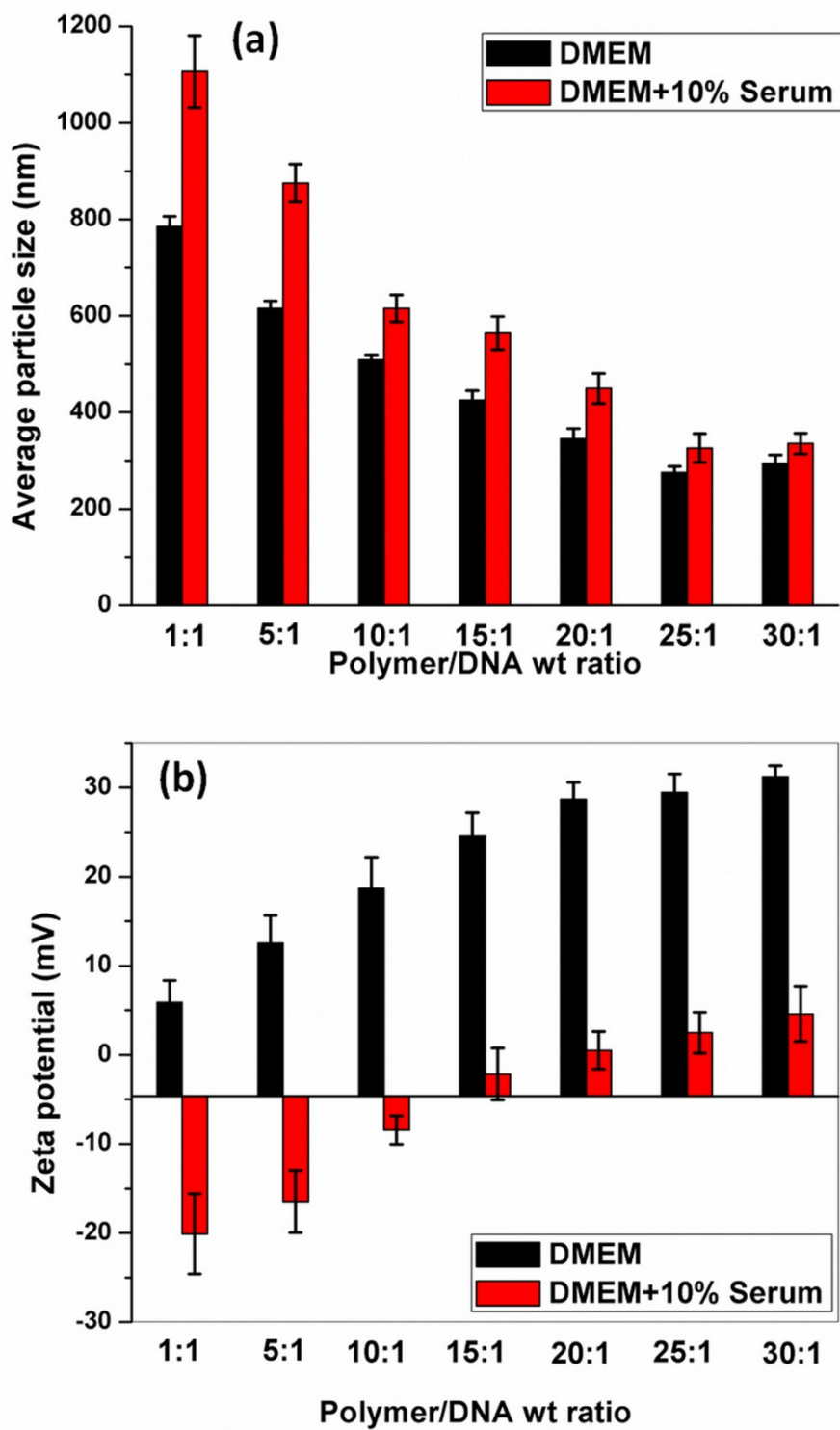


**Figure S6.** *In vitro* toxicity of GT/pDNA and different GT-LPEI/pDNA complexes at different weight ratios (1:1, 5:1, 10:1, 15:1, 20:1, 25:1 and 30:1) in various cells (a) HeLa, (b) A549, (c) SVEC, (d) MC3T3-E1, (e) RAW 264.7 and (f) hMSC. LF 2000/pDNA and PEI (25 kDa)/pDNA complexes were used as positive controls and untreated cells were used as the negative control.

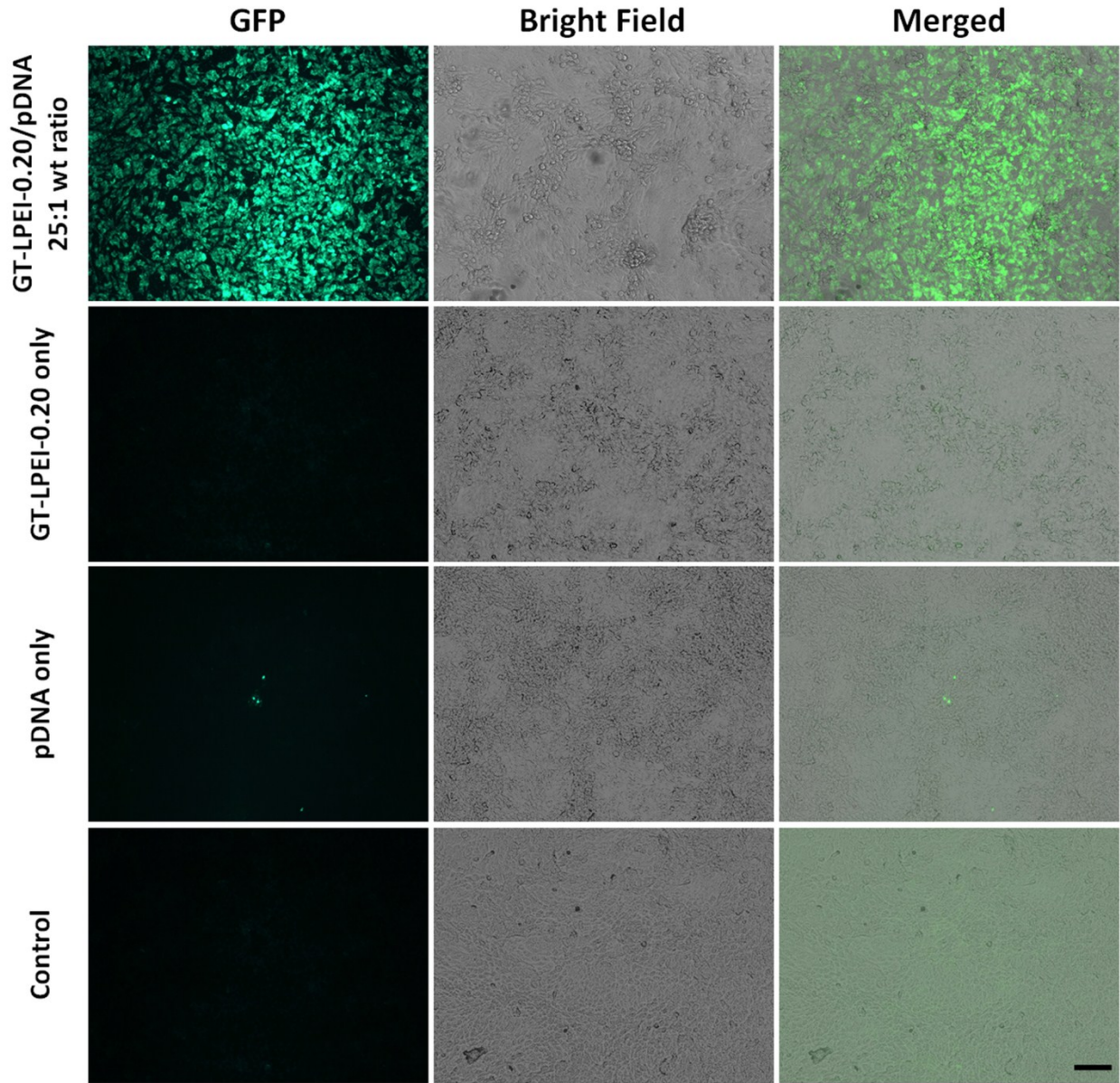




**Figure S7.** Representative fluorescence micrographs of HeLa cells by GT/pDNA and different GT-LPEI/pDNA complexes at weight ratios of 20:1, 25:1 and 30:1 containing 1  $\mu\text{g}$  of pDNA in each formulation. The scale bar is 10  $\mu\text{m}$ .

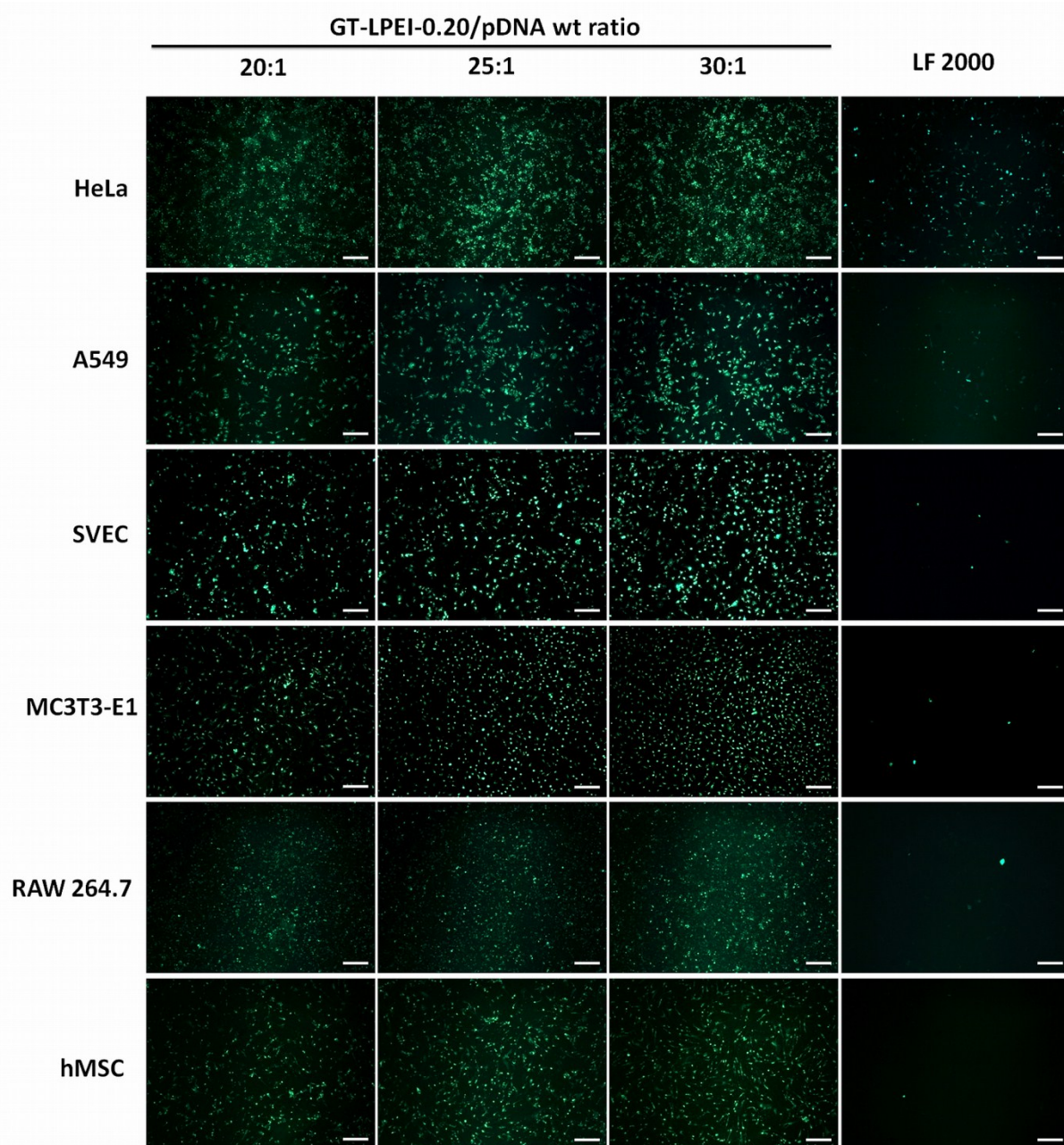


**Figure S8.** (a) Average particle size and (b) zeta potential of GT-LPEI-0.20/pDNA complexes at different weight ratios of 1:1, 5:1, 10:1, 15:1, 20:1, 25:1 and 30:1 in serum free and serum containing media determined by DLS.

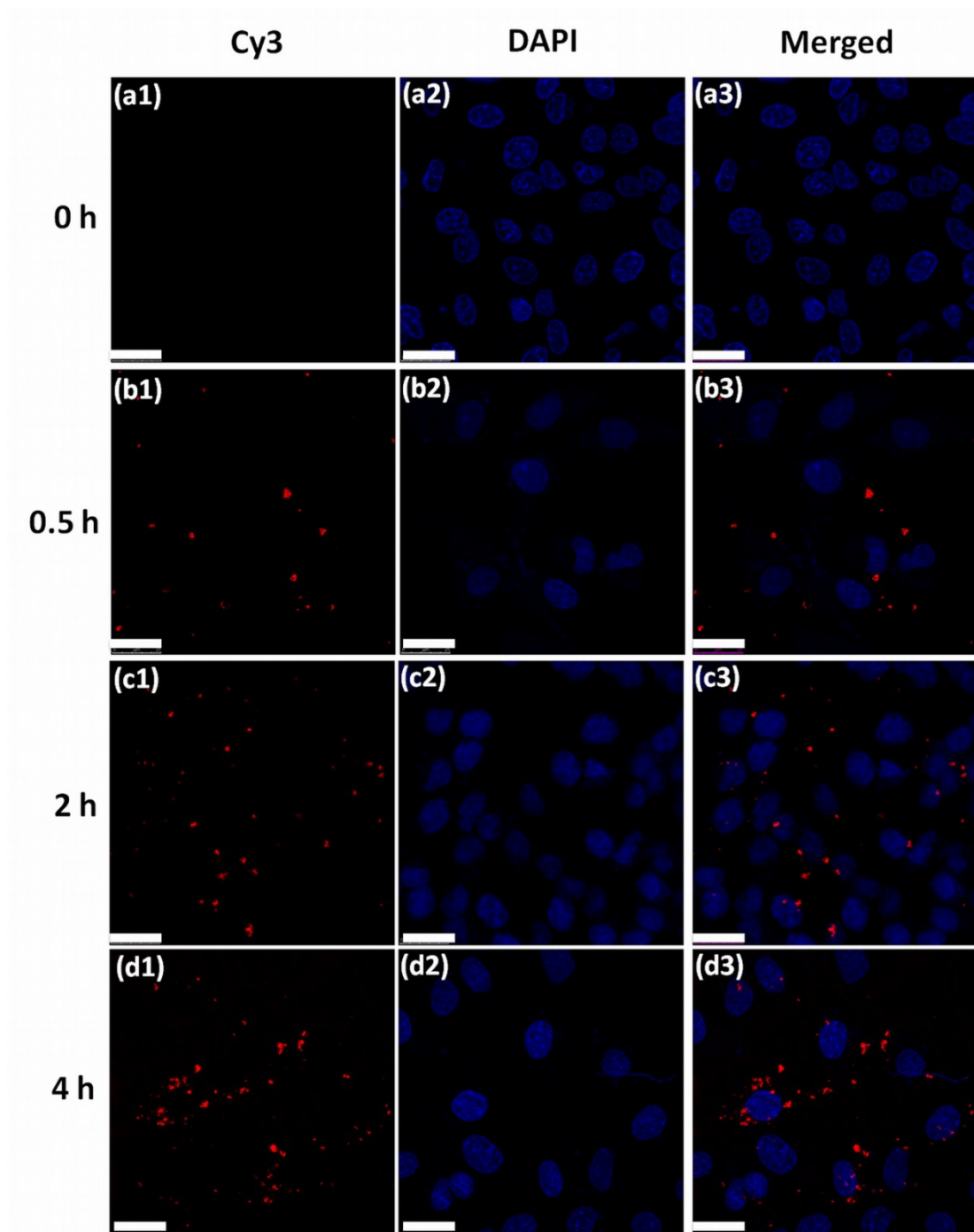


**Figure S9.** Representative fluorescence micrographs of transfected HeLa cells by GT-LPEI-0.20/pDNA complexes at weight ratio of 25:1 containing 1  $\mu$ g of pDNA. GT-LPEI-0.20 alone and pDNA alone were used as mock and negative controls, respectively. The scale bar is 1 mm.

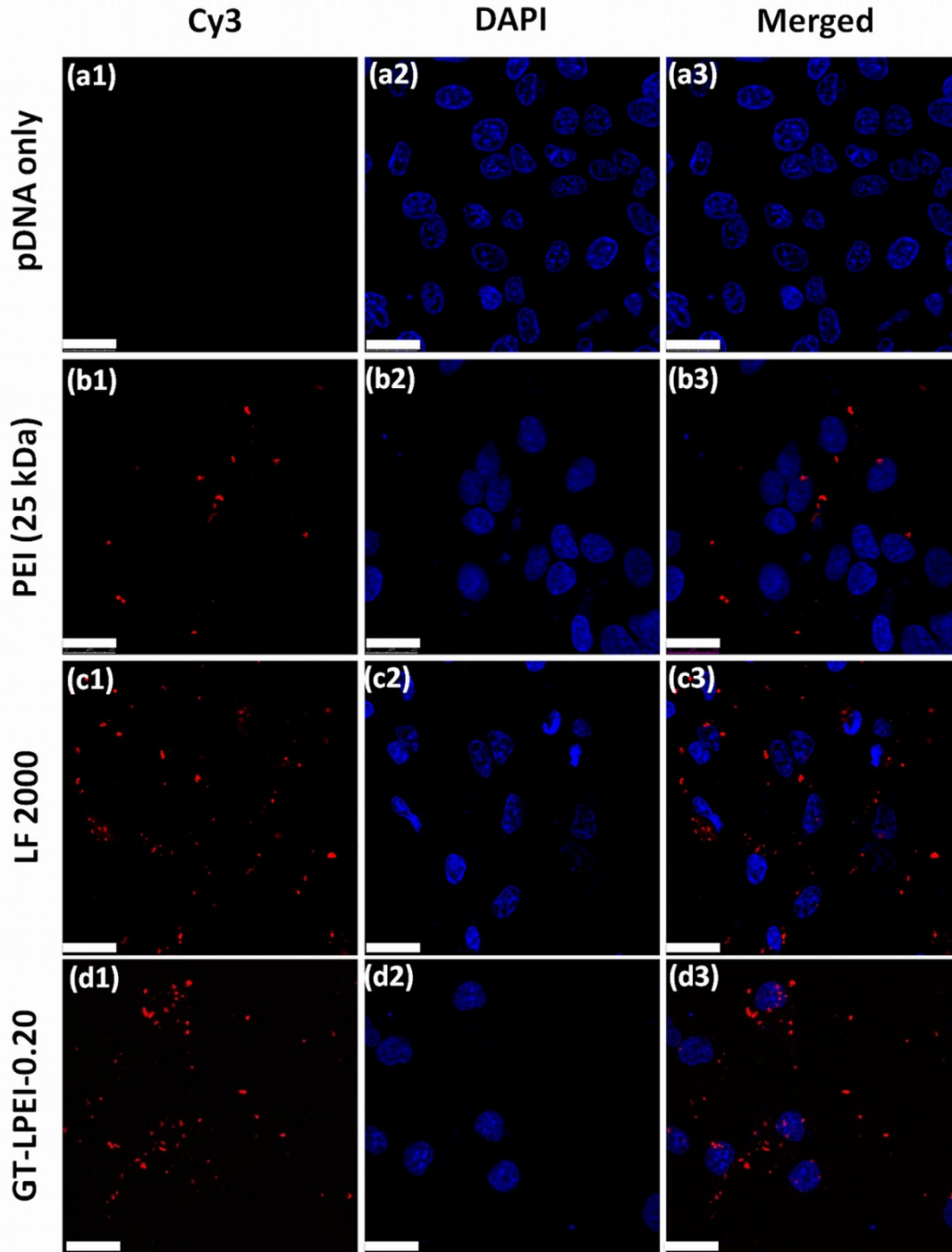




**Figure S10.** Representative fluorescence micrographs of different cells transfected by GT-LPEI-0.20/pDNA complexes at weight ratios of 20:1, 25:1 and 30:1 containing 1  $\mu$ g of pDNA in each formulation. LF 2000/pDNA complex was used as positive control. The scale bar is 5 mm.



**Figure S11.** CLSM images of intracellular distribution of GT-LPEI-0.20/ Cy3-labeled pDNA complex at weight ratio of 25:1 in HeLa cells at different time points including 0 h (a1-a3), 0.5 h (b1-b3), 2.0 h (c1-c3) and 4.0 h (d1-d3). For each panel, images from left to right show Cy3 labeled pDNA (red), stained cell nuclei (blue), and overlay of the two images. Scale bar is 25  $\mu$ m.



**Figure S12.** CLSM images of cellular uptake by HeLa cells after 4 h of transfection with Cy3-labeled pDNA alone (a1-a3), PEI/ Cy3-labeled pDNA complex at N/P ratio of 10 (b1-b3), LF 2000/ Cy3-labeled pDNA complex (c1-c3) and GT-LPEI-0.20/ Cy3-labeled pDNA complex at weight ratio of 25:1 (d1-d3) containing 1  $\mu$ g pDNA in each formulation. For each panel, images from left to right show Cy3 labelled pDNA (red), stained cell nuclei (blue), and overlay of the two images. Scale bar is 25  $\mu$ m.