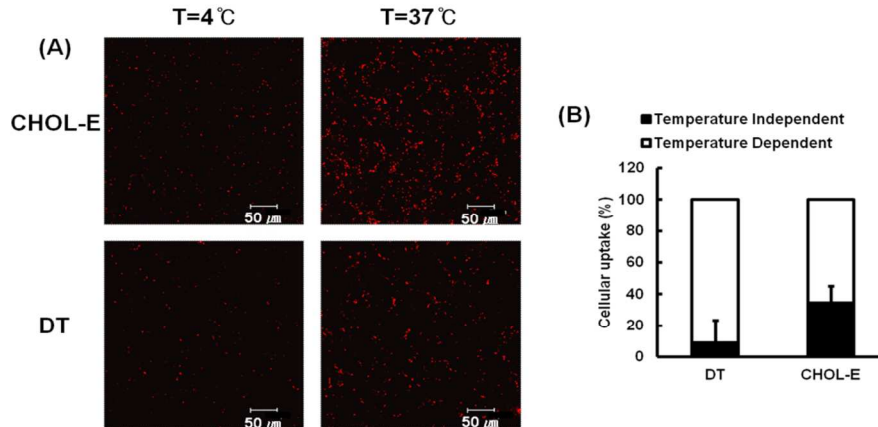
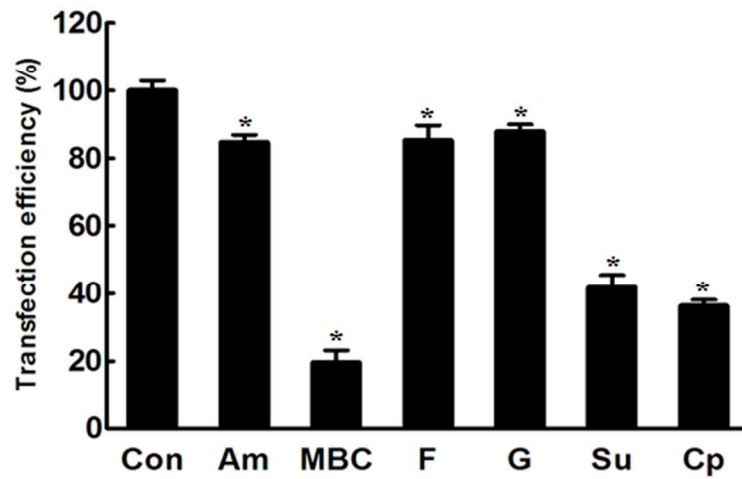


**Figure S 1.** Comparison of transfection efficiency with commercial cationic lipids. COS-7 cells were transfected with CHOL-E, LFA, DT lipoplexes in the absence (- or empty bar) or presence (+ or solid bar) of the serum. (A) GFP expression was observed via fluorescence microscopy 24 h after transfection. (B), Luciferase activity was determined 24 h after transfection. Data were expressed as relative light Unit (RLU)/ug total protein content. Values are given as means  $\pm$  SD of triplicates. (LFA: Lipofectamin, DT: DOTAP)



**Figure S 2.** Energy dependent and independent internalization of CHOL-E and DT lipoplex. COS-7 cells were transfected with Cy3-labeled DNA (red) using CHOL-E and DT lipoplexes in 4°C or 37°C for 2 h. (A) Pictures were taken via confocal microscopy. Scale bar; 50  $\mu$ m (B) Cy3-labeled CHOL-E and DT lipoplexes were analyzed by flow cytometry. Estimation of the percentage of temperature dependent (empty bar) and independent (solid bar) uptake for CHOL-E and DT lipoplex. Values are given as means  $\pm$  SD of triplicates. \*  $p < 0.05$  compared with DT 37°C or CHOL-E 37°C.



**Figure S 3.** Effect of endocytosis inhibitors on the transfection efficiency of CHOL-E lipoplexes in the presence of serum. COS-7 cells were pretreated with clathrin-, caveolae-mediated endocytosis and macropinocytosis inhibitor, then transfected with CHOL-E lipoplex in the presence of serum. Luciferase activity was determined 24 h after the transfection, and relative expression efficiency compared to control were displayed. (Am: amiloride, MBC: methyl- $\beta$ -cyclodextrin, F: filipin III, G: genistein, Su: sucrose, Cp: chlorpromazine, Con: without inhibitor) Values are given as means  $\pm$  SD of triplicates. \*  $p < 0.05$  compared with control.