

Figure S1. Effect of PEGylation of cationic liposomes on (A) the agglutination of cationic lipoplexes with erythrocytes, and (B and C) siRNA biodistribution after intravenous injection of cationic lipoplex. PEGylated cationic liposomes were prepared by inclusion of 1, 2, 3 and 5 mol% PEG₂₀₀₀-DSPE into the formulations of DOTAP/cholesterol liposomes (molar ratio of 1:1). (A) PEGylated cationic lipoplexes containing 2 μ g siRNA at a charge ratio (+:-) of 4:1 were added to erythrocyte suspensions, and agglutination was observed by phase contrast microscopy. Scale bar, 100 μ m. (B and C) PEGylated cationic lipoplexes were prepared by mixing PEGylated cationic liposomes with 20 μ g Cy5.5-siRNA at a charge ratio (+:-) of 4:1, and were administered intravenously to mice. (B) At 1 h post-injection, the mice were sacrificed, and Cy5.5 fluorescent imaging of the tissues was performed using a NightOWL LB981 NC100 system (Berthold Technologies, Bad Wildbad, Germany). The images were analyzed using the IndiGo2 software provided with the *in vivo* imaging system (Berthold Technologies). Fluorescence intensity is illustrated as a color-coded scale (red is maximum, purple is minimum). (C) The tissues following fluorescent imaging were frozen on dry ice and cut into 16- μ m-thick slices. The localization of Cy5.5-siRNA was examined using an Eclipse TS100-F microscope. Green signals indicated localization of Cy5.5-siRNA. Scale bar, 100 μ m.

