

Supplementary Materials: Non-Cationic RGD-Containing Protein Nanocarrier for Tumor-Targeted siRNA Delivery

Xiaolin Yu, Lu Xue, Jing Zhao, Shuhua Zhao, Daqing Wu and Hong Yan Liu

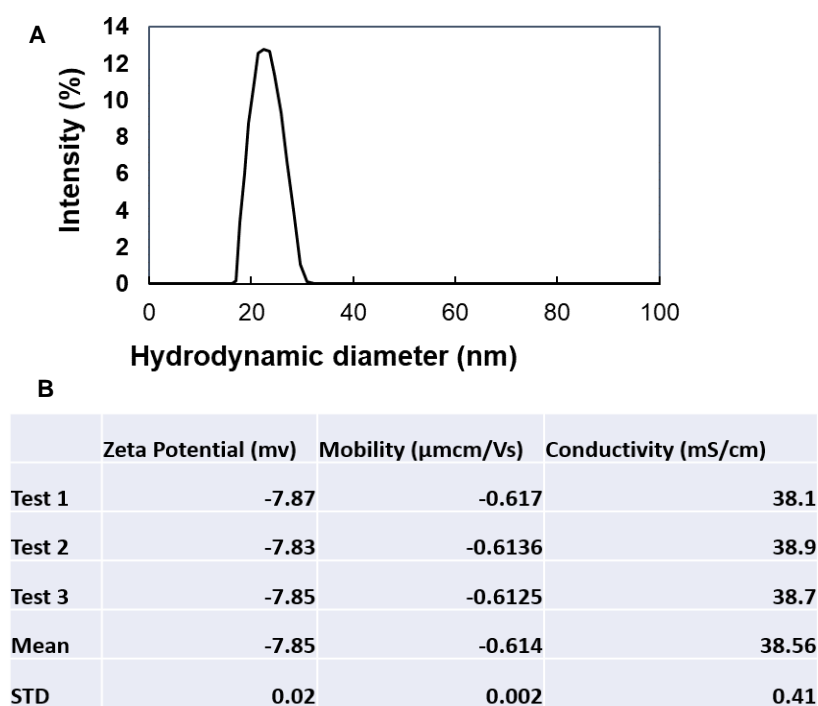


Figure S1. Detection of hydrodynamic diameter and zeta potential by Zetasizer. Monomer dual-RGD in PBS buffer were measured at 25°C. The hydro-dynamic diameter is about 23.5nm shown in (A) and zeta potential is about -7.85mv shown in (B).

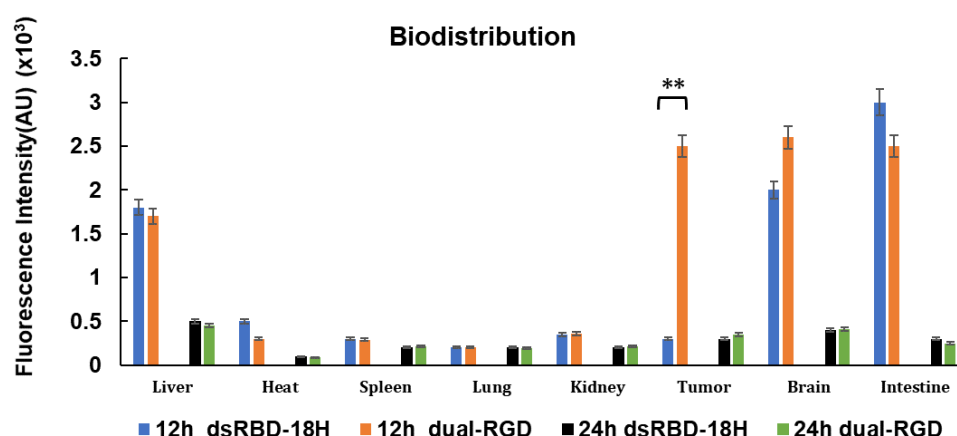


Figure S2. Ex vivo evaluation of biodistribution. (A) Athymic mice were injected with MDA-MB-231 cells (5×10^6) mixed with Matrigel (v/v 1:1) subcutaneously. After 4 weeks, tumor-bearing mice were tail-vein injected with 100 μl of Cy5-EGFR siRNA/dual-RGD complex (5 nmoles) or equal moles of Cy5-siRNA/dsRBD-18His complex. At time of 12h and 24h, major organs are removed and homogenized in buffer (10mM Tris pH7.4 and 0.5% Triton X-100) with mortar and pestle at a ratio of 100mg of tissue per ml buffer. 100 μl of tissue homogenate was loaded to a 96-well plate. The plate was measured by Tecan Infinite F200 Pro Microplate Reader. The results are the mean \pm SEM (N=3). **P<0.01.

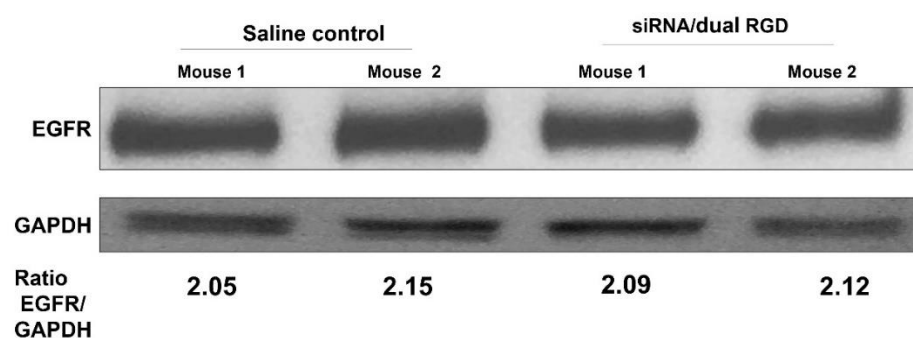


Figure S3. Evaluation of off-target gene silencing in brain. After treatment of saline or siRNA/dual RGD (5nmles) twice a week for 4 weeks. EGFR silencing in brain tissues was detected by Western blot. There is no significant difference of EGFR expression between saline treated or siRNA/dual RGD treated mouse brains. This result indicates dual-RGD did not induce siRNA endocytosis/gene knockdown in brain.