**Supporting information**

# Endosomolytic and Tumor-Penetrating Mesoporous Silica Nanoparticles for siRNA/miRNA Combination Cancer Therapy

*Yazhe Wang,[1, †, $] Ying Xie,[1, †, $] Kameron V. Kilchrist,[‡] Jing Li,[†] Craig L. Duvall,[‡] David Oupický[†*]*

[†] Center for Drug Delivery and Nanomedicine, Department of Pharmaceutical Sciences, University of Nebraska Medical Center, Omaha, Nebraska, United States

[‡] Department of Biomedical Engineering, Vanderbilt University, Nashville, Tennessee, United States

[$] Present address: Department of Biomedical Engineering, Yale University, New Haven, Connecticut, United States

[1] These authors contributed equally

*Corresponding author: david.oupicky@unmc.edu

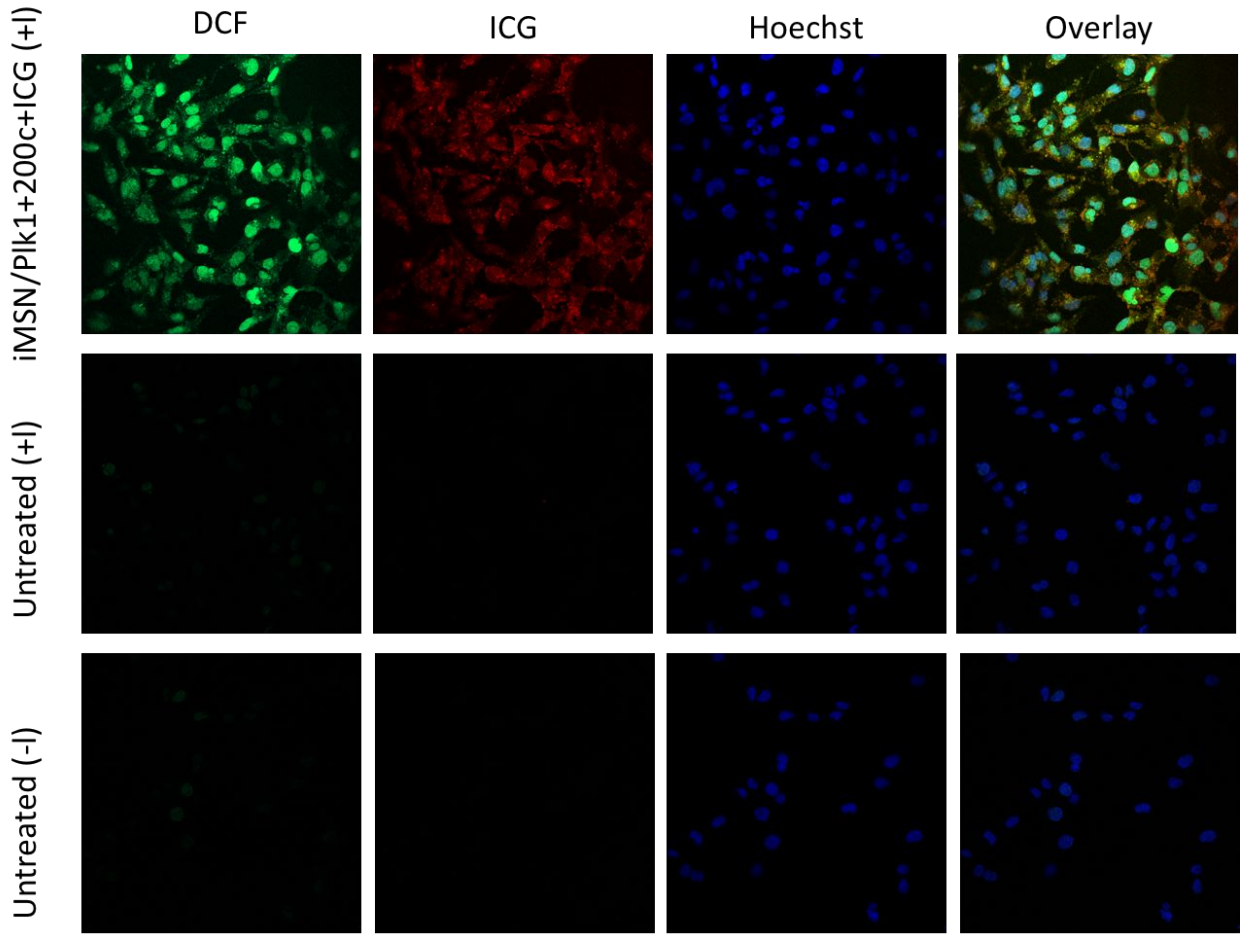


**Figure S1.** ROS generation following 4 h incubation of tumor cells with nanoparticle and laser irradiation. Confocal images of MDA-MB-231 cells stained by ROS detecting probe DCF (green). Cell nuclei were stained by Hoechst 33258.

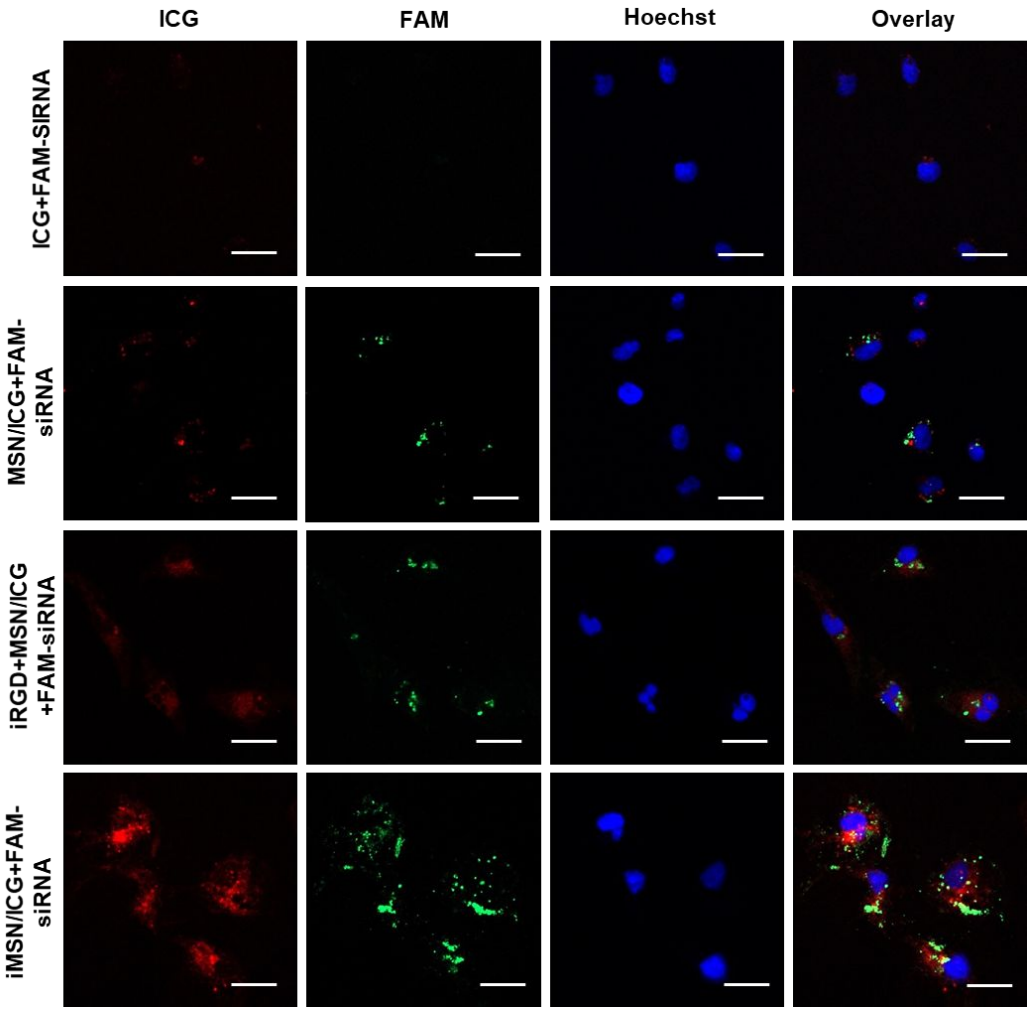


**Figure S2.** Cellular uptake in MDA-MB-231 cells by confocal microscopy. The cells were incubated with the nanoparticles or free ICG+FAM-siRNA for 4 h. Nuclei were stained with Hoechst dye. Red fluorescence of ICG, green fluorescence of FAM-siRNA. Scale bar, 20 μm.

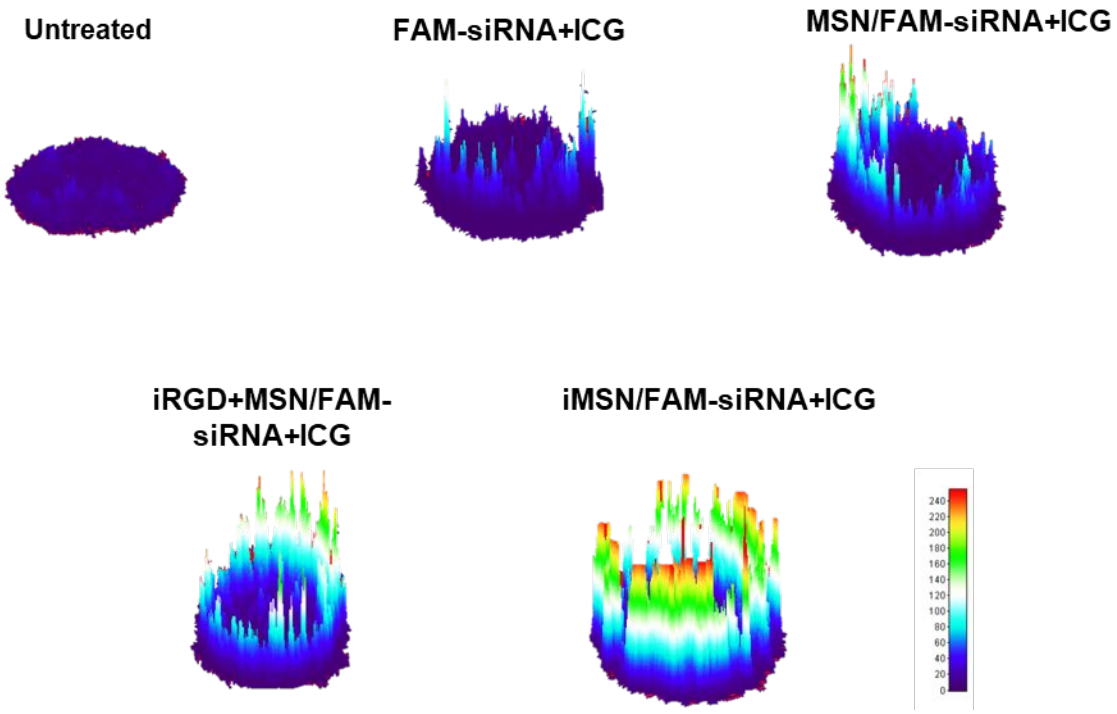


**Figure S3.** Surface plot images of MDA-MB-231 tumor spheroids treated with free FAM-siRNA+ICG, MSN/FAM-siRNA+ICG, iRGD+MSN/FAM-siRNA+ICG, iMSN/FAM-siRNA+ICG and negative control. Fluorescence signal was from FAM-siRNA channel.

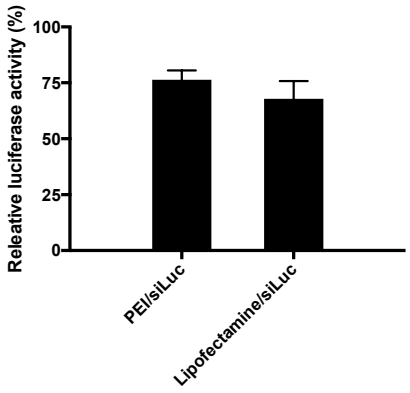


**Figure S4.** Luc silencing efficacy of siLuc loaded with PEI and Lipofectamine 3000 in MDA-MB-231.Luc cells. Data are shown as mean ± SD (n = 3).

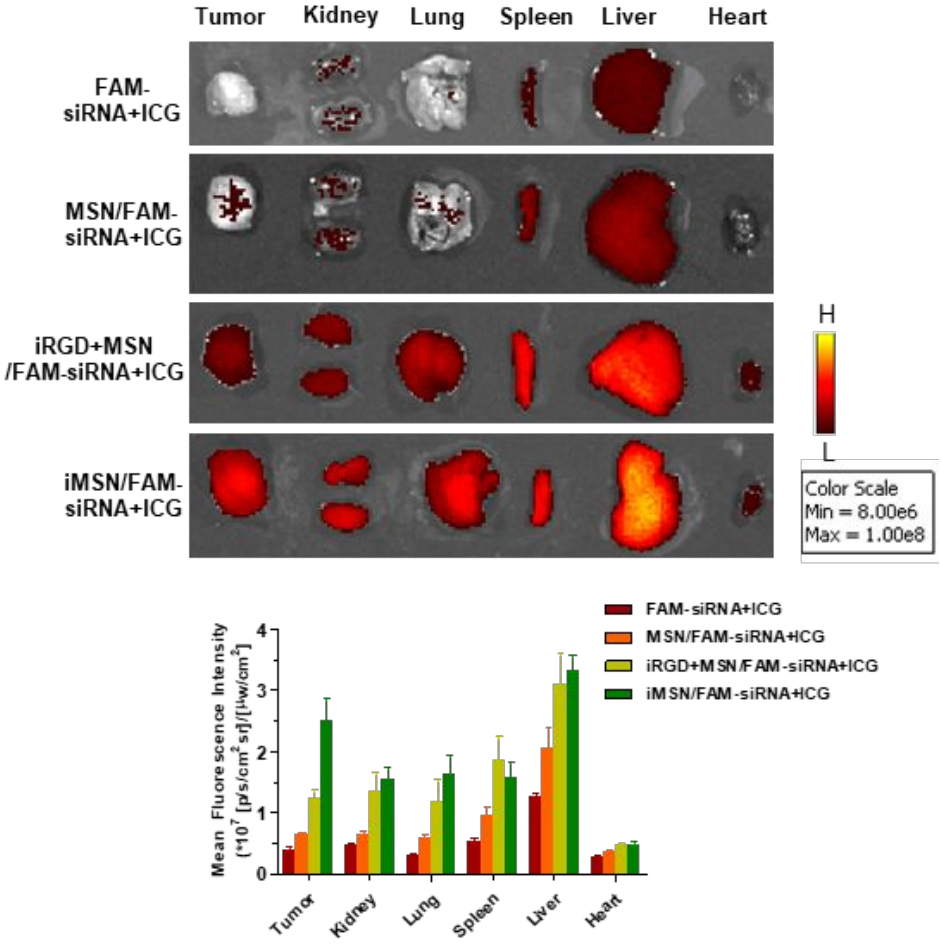


**Figure S5.** *Ex vivo* images of tumors and other tissues at 48 h after injection and semiquantification of nanoparticle biodistribution in mice tissues. Results are expressed as mean fluorescence intensity ± SD (n = 3).

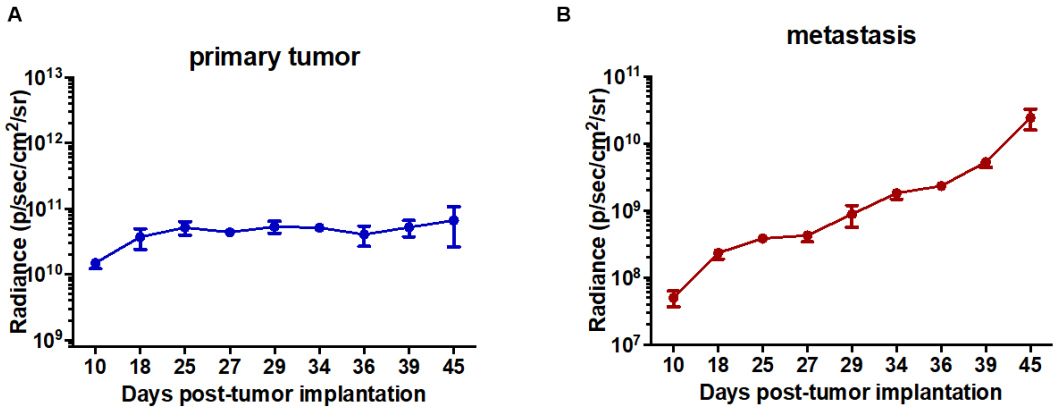


**Figure S6.** Quantitative analysis of bioluminescence signal from primary tumor (A) and metastasis (B) in untreated tumor-bearing mice post-tumor implantation. Results are expressed as mean fluorescence intensity ± SD (n = 5).

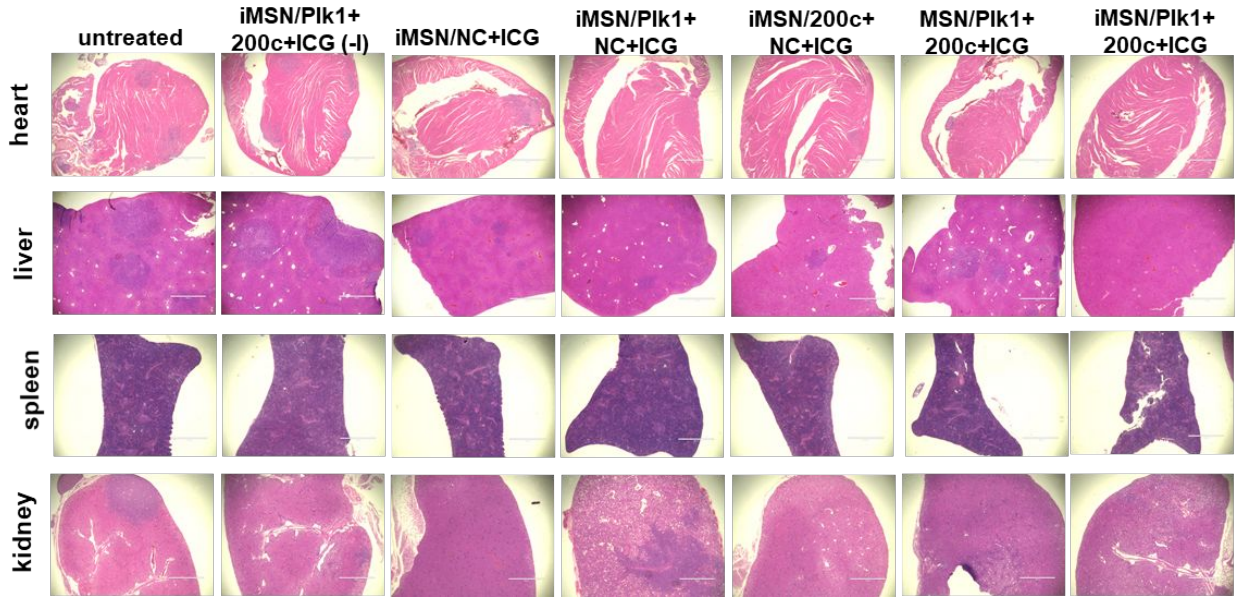


**Figure S7.** Histological observation of tissue sections from major organs of mice was performed after the treatment. The organ sections were stained with hematoxylin and eosin (H&E). Scale bar, 1000 μm (4x), 100 μm (40x).

Table S1. Blood biochemistry of tumor-bearing mice treated with iMSN/NC+ICG, iMSN/Plk1+200c+ICG (+/-light) or saline from the efficacy experiment.

| | Saline | NC+ICG | Plk1+200c+ICG (-l) | Plk1+200c+ICG (+l) |
|---|---|---|---|---|
| WBC ($10^9$/L) | 6.2±3.9 | 4.6±1.4 | 5.2±0.8 | 3.1±0.5 |
| LYM ($10^9$/L) | 0.7±0.6 | 0.6±0.3 | 0.5±0.2 | 0.9±0.6 |
| MON ($10^9$/L) | 0.4±0.2 | 0.4±0.1 | 0.4±0.1 | 0.2±0.2 |
| NEU ($10^9$/L) | 5.2±3.1 | 3.5±1.6 | 4.3±0.6 | 2.0±0.6 |
| RBC ($10^{12}$/L) | 8.9±0.5 | 8.9±0.3 | 9.2±0.7 | 8.1±0.3 |
| HGB (g/dL) | 15.2±0.6 | 15.0±0.7 | 15.3±1.1 | 14.0±0.8 |
| HCT (%) | 42.1±3.7 | 40.7±1.5 | 41.7±2.9 | 35.9±1.5 |
| MCV (fl) | 46.0±0.0 | 46.0±0.0 | 46.0±1.0 | 45.0±0.0 |
| MCH (pg) | 16.7±1.0 | 16.8±0.3 | 16.7±0.1 | 17.6±0.5 |
| MCHC (g/dL) | 37.3±1.0 | 36.8±0.5 | 36.8±0.7 | 39.1±0.9 |
| RDWc (%) | 18.6±0.2 | 18.5±0.5 | 18.1±0.3 | 19.1±0.6 |
| RDWs (fl) | 34.1±0.5 | 34.1±0.9 | 32.8±0.8 | 34.4±0.8 |
| PLT ($10^9$/L) | 656±77 | 594±52 | 614±78 | 705±172 |
| MPV (fl) | 6.0±0.3 | 6.2±0.2 | 6.2±0.1 | 6.0±0.1 |
| PCT (%) | 0.4±0.1 | 0.4±0.0 | 0.4±0.1 | 0.4±0.1 |
| PDWc (%) | 29.3±0.6 | 29.3±0.6 | 30.5±1.0 | 29.1±1.2 |
| PDWs (fl) | 6.1±0.3 | 6.1±0.3 | 6.6±0.5 | 6.0±0.5 |
| BUN (mg/dL) | 26.2±2.5 | 24.2±4.5 | 28.0±7.3 | 20.7±7.1 |
| CRE (mg/dL) | 2.2±0.2 | 2.2±0.6 | 1.9±0.1 | 1.7±0.3 |
| AST (U/L) | 29.0±1.2 | 27.5±6.6 | 26.3±4.7 | 26.1±2.5 |
| ALT (U/L) | 4.5±0.9 | 5.1±1.5 | 5.3±1.0 | 5.3±0.6 |