Support information

CD44v6 Monoclonal Antibody-Conjugated Gold Nanostars for Targeted Photoacoustic Imaging and Plasmonic Photothermal Therapy of Gastric Cancer Stem-like Cells

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X-ray and CT imaging

GNSs have potential as X-ray contrast agents. Folic acid-conjugated and silica-modified gold nanorods had been applied for X-ray/CT imaging by Peng Huang et al, due to their strong X-ray attenuation. In this study, X-ray imaging of GNS *in vitro* was used to evaluate the ability of GNS-PEG-CD44v6 as contrast enhancement agents (Fig. S 2A and B). It is detected that there is a linear relationship ($R^2 = 0.9889$) between the intensity and concentration (X-ray imaging).

The Hounsfield Units (HU) of GNSs was also evaluated by CT. The scan was measured at 50kVp. Coronal, axial and sagittalia view of the nanoprobe in the concentration range of 0.0625-1 mg/mL were shown in Fig. S3A, B. With the concentration increased, the CT signal intensity raised continuously, as shown in Fig. S3C. The above results demonstrated that GNS may be applied as a

X-ray/CT imaging contrast agent.

To observed the feasibility of detecting GCSC *in vivo* by X-ray/CT imaging, 150 μl GNS-PEG-CD44v6 (0.867 mg Au/ml, test group) were intravenously injected to subcutaneous GC xenografted mice models. GNS-PEG injected group was set as control. The X-ray/CT images of mice were recorded on Kodak In-vivo Imaging System FX Pro I instrument and Micro-CT IVIS Quantum FX System respectively at 24 hour after injection. As shown in Figure S2 and S3, slightly signal enhancement of the tumor could be detected in the test proup, displaying positive-contrast than other soft tissues, which is attributed to the strong X-ray attenuation induced by gold element. However, there was no obvious change or signal enhancement perceived in the control group.(as hadn't been detaied)

Based on the above results, it is demonstrated that GNS-PEG-CD44v6 could be used to detect the distribution of GCSCs *in vivo* by X-ray/CT imaging and to predict prognosis and development of gastric cancer. X-ray/CT imaging were applied to provide a reference potential for GCSCs and GC targeted imaging.

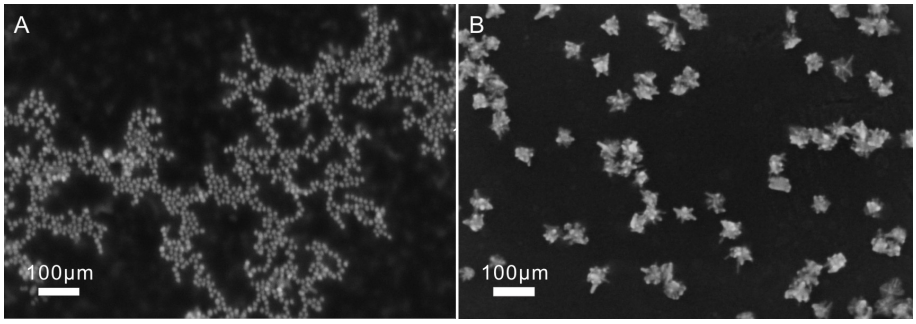

Fig. S1. SEM image of (A) citrate gold seeds and (B) GNSs in inlense mold.

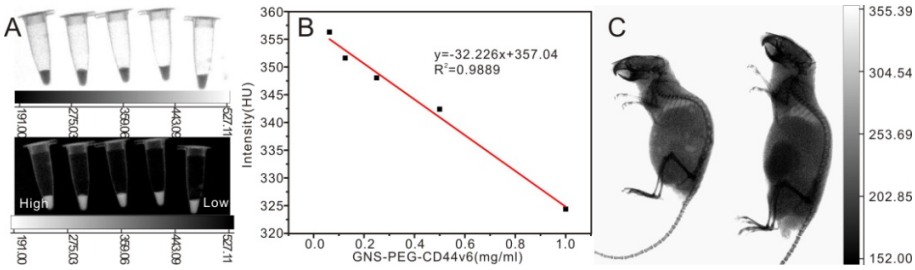

Fig. S2. (A) X-ray imaging of GNS-PEG-CD44v6 *in vitro*; (B) Dependence of HU signals on the concentration of nanoparticle; (C) X-ray imaging of tumor bearing mice injected with GNS-PEG-CD44v6 (right) and GNS-PEG (left) at 24 h.

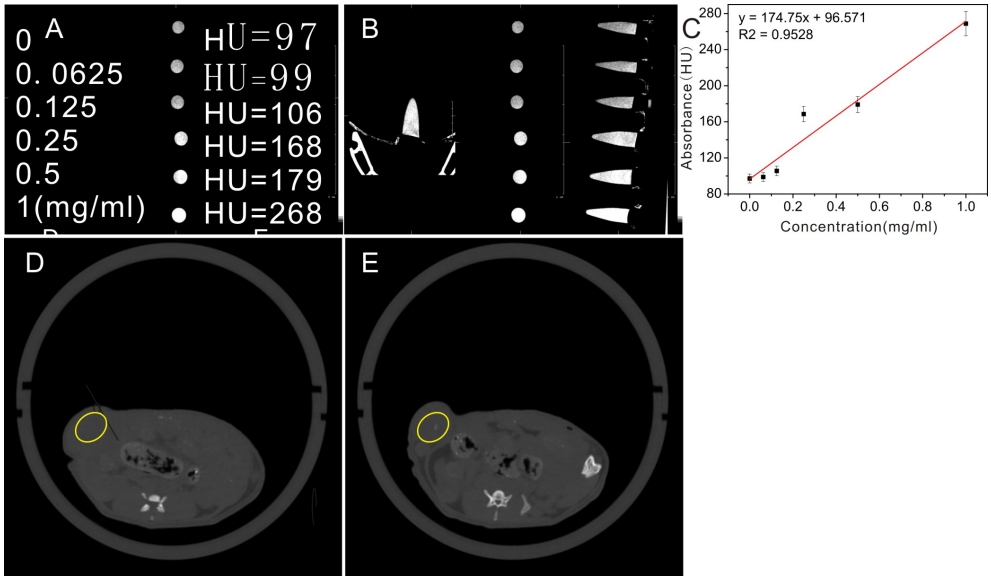

Fig. S3. CT images of GNS-PEG-CD44v6 of different concentration (A) and perspective (coronal, axial and sagittalia view) (B); Dependence of CT attenuation (HU) on the concentration of GNS-PEG-CD44v6; CT images of tumor bearing mice injected with GNS-PEG and GNS-PEG-CD44v6 (D and E) respectively at 24 h.

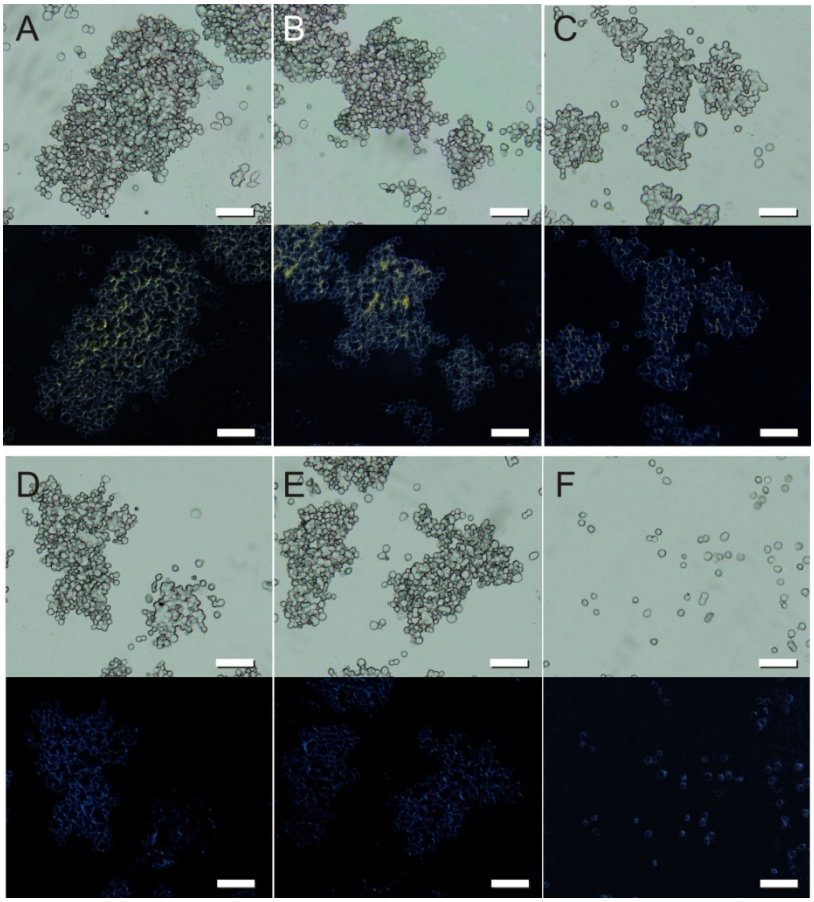

Fig. S4. DIC (differential interference contrast) in bright field and dark field scattering images of GCSCs (CD44+ sphere cells) and CD44- cells. CD44+ sphere cells incubated with GNS-PEG-CD44v6 for 2 h (A), 24 h (B) at 37 ^{o}C; (C) CD44+ sphere cells incubated with GNS-PEG-CD44v6 for 24 h at 4 ^{o}C; (D) pre-incubation of anti-CD44v6Ab before CD44+ sphere cells treated with GNS-PEG-CD44v6 for 24 h at 37 ^{o}C; (E) CD44+ sphere cells incubated with GNS-PEG for 24 h at 37 ^{o}C; (F) CD44- cells incubated with GNS-PEG-CD44v6 for 24 h at 37 ^{o}C; All scale bars are 50 μm.

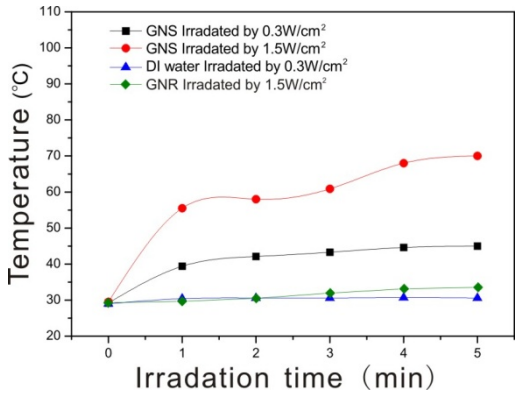

Fig. S5. Temperature profile of gold nanostars, and gold nanorods and DI water (upon laser irradiation (790 nm, 0.3, 1.5W/cm^2) for 5 min.

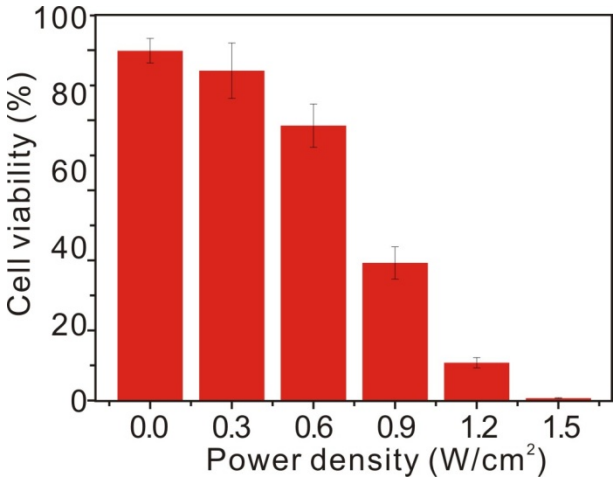

Fig. S6. Cell viability assay of the cells treated with the GNS-PEG-CD44v6 which were exposed to NIR laser irradiation with different power density (0.3, 0.6, 0.9, 1.2, 1.5 W/cm^2).

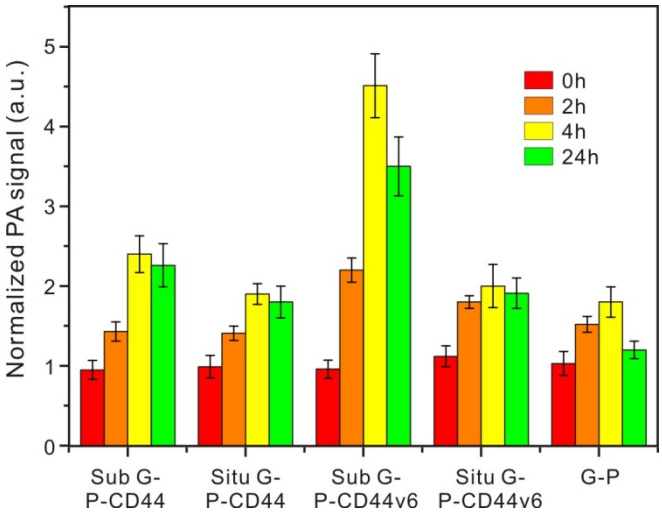

Fig. S7. The quantification of PA signal (n = 4). Error bars represent standard deviation.