Supporting information

Enhanced Intracellular Ca^{2+} Nanogenerator for Tumor Specific Synergistic Therapy *via* Disruption of Mitochondrial Ca^{2+} Homeostasis and Photothermal Therapy

Lihua Xu [a,b,c,1], *Guihua Tong* [a,1], *Qiaoli Song* [a], *Chunyu Zhu* [a], *Hongling Zhang* [a,b,c], *Jinjin Shi* [a,b,c,*], *Zhenzhong Zhang* [a,b,c,*]

a School of Pharmaceutical Sciences, Zhengzhou University, Zhengzhou, People's Republic of China
b Key Laboratory of Targeting Therapy and Diagnosis for Critical Diseases, Henan Province, People's Republic of China
c Collaborative Innovation Center of New Drug Research and Safety Evaluation, Henan Province, Zhengzhou, China.

*Corresponding authors: Prof. Zhenzhong Zhang and Dr. Jinjin Shi. Email: zhangzhenzhong@zzu.edu.cn, shijinyxy@zzu.edu.cn.

Lihua Xu and Guihua Tong contributed equally to this work.

EXPERIMENTAL SECTION

Snythesis of HMCuS

Poly-vinylpyrrolidone (PVP-K30, 0.2400 g) was dissolved in deinonized water (25 mL) under magnetic stirring at room temperature. And then $CuCl_2.2H_2O$ (100 μL, 0.5 M) was added to the above solution to form homogeneous solution. Subsequently, NaOH (25 mL, 0.02 mM) and hydrazine hydrate (6.4 μL) were added into the mixture with stirring for another 5 min to form Cu_2O nanoparticles. And then Na_2S (200 μL, 32 mg/mL) was added to the reaction mixture, and stirred at 60℃ for 4 h. The resulting product was centrifugated at 12000 rpm for 5 min and washed twice with deionized water and ethanol, respectively. At last, the synthetic nanoparticles (HMCuS) were lyophilized for further use.

The effect of weight ratio of CaNG and CUR on CUR loading efficienvy

CaNG (16 mg) was divided into quarters, and dispersed in ethanol (4 mL). Each part was added into 2, 4, 8 and 16 mg CUR respectively, and stirred for 24 h at room temperature. The reaction mixtures were then centrifugated at 12000 rpm for 5 min to remove the free CUR. The precipitates (CUR loaded nanoparticles) were washed with anhydrous ethanol three times. The loading efficacy of CUR was detected by HPLC with following chromatographic conditions: a symmetry C18 (250 mm × 4.6 mm, 5 μm); mobile phase, acetonitrile and 0.4% glacial acetic acid with volume ratio of 44:56; column temperature, 30℃; flow rate, 1.0 mL/min; and injection volume, 10 μL.

Loading efficiency of CUR was calculated by the formula: $\text{Loading capacity}(\%) = \frac{M(CUR_{total}) - M(CUR_{free})}{M(CaNG)} \times 100\%$, where $M(CUR_{total})$ is the total CUR added into the CaNG

solution, M(CUR_{free}) is the unloaded CUR in the supernatant, and M(HMCuS@CaP) is the total adding amount of CaNG.

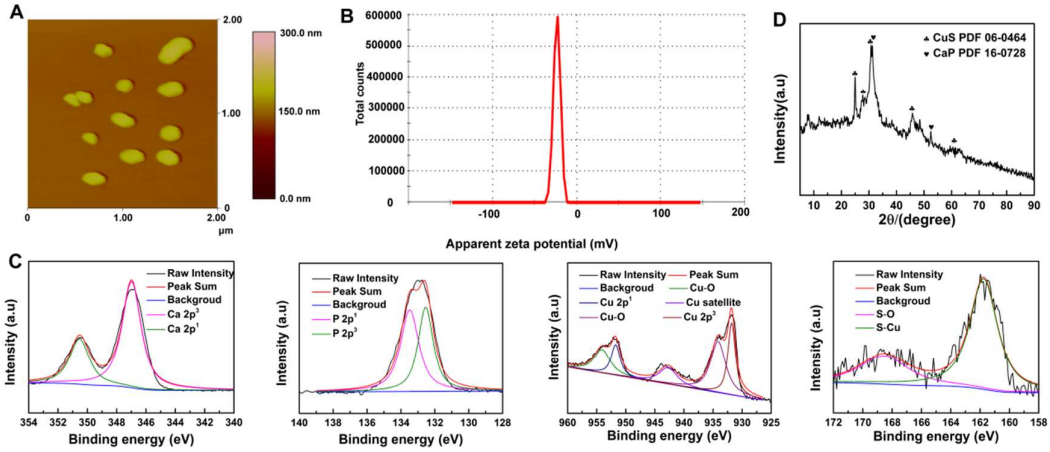

Figure S1. (A) AFM images of CaNG. (B) Zeta potential measurement of CaNG. (C) XPS analysis of various elements in CaNG. (D) XRD spectrum of CaNG.

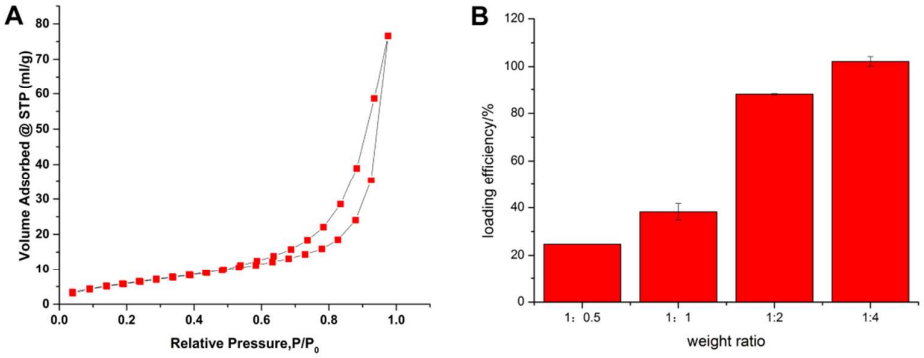

Figure S2. (A) Nitrogen adsorption-desorption isotherms plots of CaNG. (B) CUR loading at different weight ratio of CaNG and CUR.(n=3)

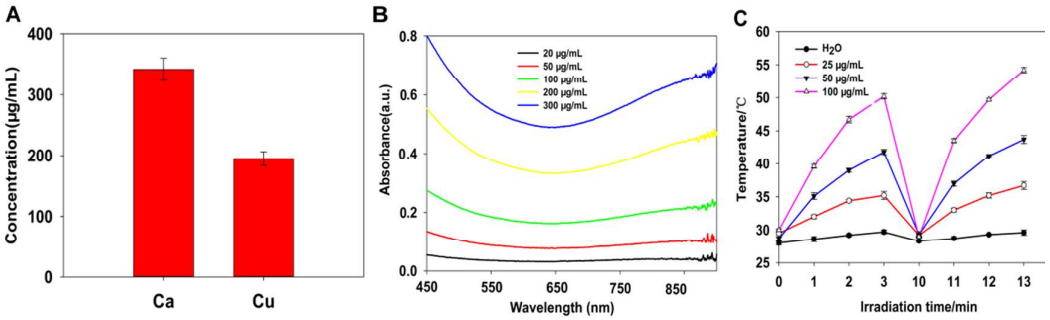

Figure S3. (A) The content of Cu and Ca of CaNG treated by nitrohydrochloric acid for 1 h. (n=3) (B) UV-vis absorption spectra of various concentrations of CaNG. (C) Photostability of CaNG with different concentrations under 808 nm laser irradiation twice. Laser was cut off at 3min and used again at 10 min. (n=3)

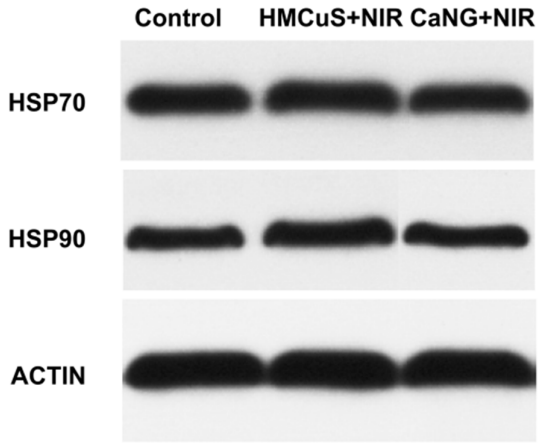

Figure S4 Western blot assays of HSP family regulation when cells were treated with HMCuS or CaNG with NIR at the same concentration of Cu^{2+}.

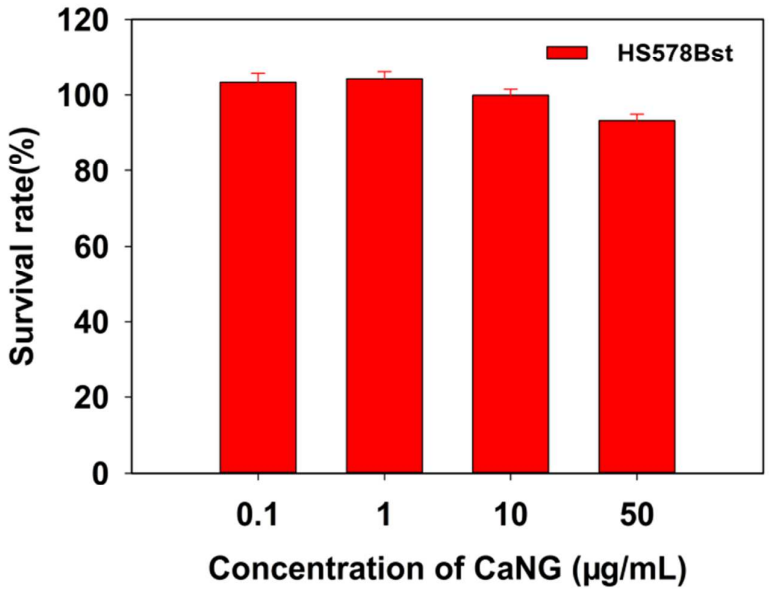

Figure S5. MCF-7 cell cytotoxicity after treated with different concentrations of CaNG for 24 h.

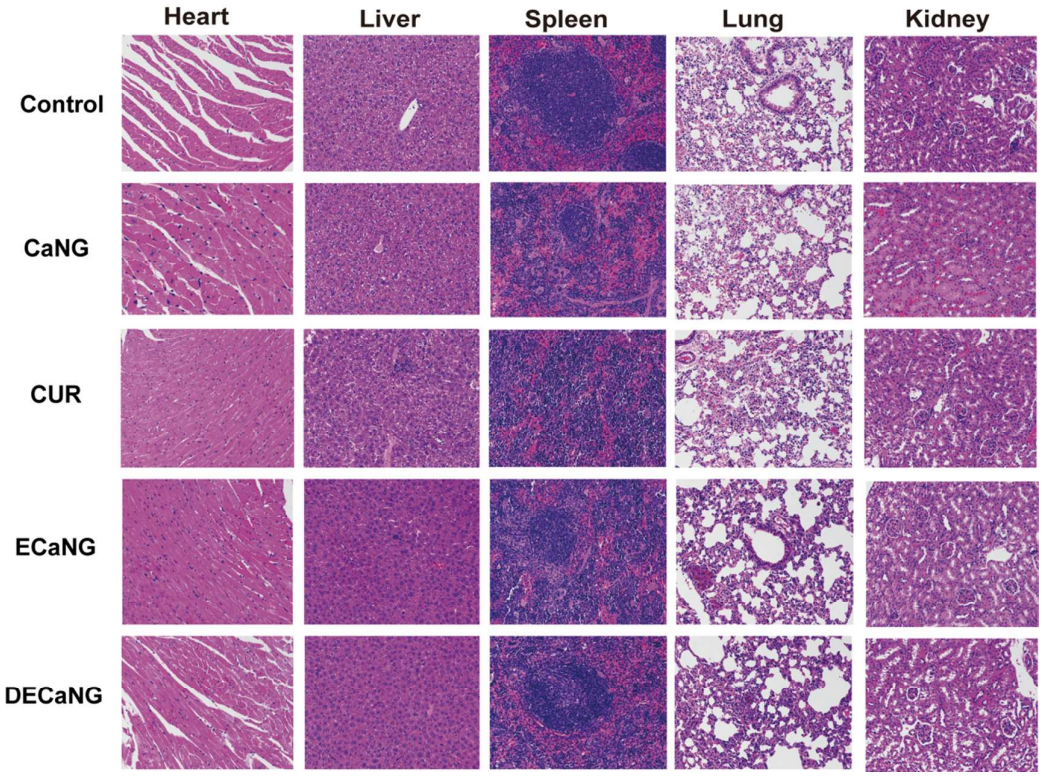

Figure S6. H&E staining of normal tissues harvested from tumor bearing mice treated with different formula (200×).