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Supporting Information

Defect-rich Sonosensitizer Based on CeO_2 with Schottky Heterojunction for Boosting Sonodynamic/Chemodynamic Synergistic Therapy

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S1. Measurements and characterizations: All powder X-ray diffraction patterns were obtained on a Powder X-ray Diffractometer (D8 Advance, Bruker AXS, Germany). The average diameter of NPs fabricated was determined by Malvern Zetasizer Nano ZS90. Transmission electron microscopic (TEM) images were obtained from a TECNAI G2 high resolution transmission electron microscope, operating at 200 kV. The O_2 concentrations were measured by a portable dissolved oxygen meter (INESA Scientific Instrument Co., Ltd, Shanghai, China). Fluorescence spectrometer (F-7000, Hitachi) for detection of reactive oxygen species. UV-visible spectrum and UV-vis diffuse reflectance spectrum were recorded by UV-3600 spectrophotometer (SHIMADZU). The In-Vivo NIR-II fluorescence imaging system (Teledyne Princeton instruments, USA) was used for NIR-II fluorescence imaging *in vivo*. X-ray photoelectron spectroscopy (XPS) spectra were performed on an ESCALAB-MKII 250 photoelectron spectrometer (VG Co.).

S2. Detection of singlet oxygen generation *in vitro*: DPBF as a typical molecular probe was used to detect singlet oxygen ($^1O_2$) production. 60 μL DPBF (10 mM, dissolved in ethanol) was mixed with 2 mL CeO_2 solution at a concentration of 50 $\mu g \cdot mL^{-1}$ (ethanol: H_2O=6:4). Then, H_2O_2 (10 mM) is added to the solution. Subsequently, the mixture was exposed to US irradiation (1 MHz, 50% duty cycle, 1.5 W cm^{-2}) at fixed intervals in the dark. The change in DPBF concentration was recorded by the absorption intensity in UV-vis spectra. For comparison, $CeO_{2-x}S_x$ and $Pt/CeO_{2-x}S_x$ solution with the same concentration were respectively exposed to US irradiation (1 MHz, 50% duty cycle, 1.5 W cm^{-2}) for fixed intervals (0/2/4/6) to detect the changes in DPBF.

By using the trapping agent TEMP (2,2,6,6-Tetramethylpiperidine), the $^1O_2$ generation was also detected in the aqueous dispersion of $Pt/CeO_{2-x}S_x$ under US irradiation (1 MHz, 50% duty cycle, 1.5 W cm^{-2}) by an electron paramagnetic resonance spectrometer at ambient temperature. Note: The transducer with a diameter of 5 cm is used to generate US irradiation in the above experiments.

S3. Detection of hydroxyl radicals generation *in vitro*: 30 μL of MB (10 μg mL^{-1}), 100 μL of H_2O_2 (10 mM), and 1 mL of PBS buffer with $Pt/CeO_{2-x}S_x$ (50 μg mL^{-1}, pH=6.5) were mixed and placed at room temperature for 0/10/20/30/40/50 min, and the hydroxyl radical (·OH) induced degradation of MB was tracked by the change of

absorbance at 665 nm.

Alternatively, by using DMPO (5,5-dimethyl-1-pyrroline-N-oxide), the ·OH generation was also detected in Pt/$CeO_{2-x}S_x$ nanocomposites dispersed H_2O_2 solution (pH=6.5) by an electron paramagnetic resonance spectrometer at ambient temperature.

S4. Detection of ROS generation *in vitro*: 0.48 mg of DCFH-DA was dissolved in 1 mL of ethanol, mixed with 4 mL of NaOH (0.01 mM), and stirred for 30 min under light-avoidance conditions, and then mixed with 20 mL of PBS (pH=6.5) to make solution A. 250 μL of solution A was taken and mixed with 100 μL of H_2O_2 (10 mM) and 1 mL of Pt/$CeO_{2-x}S_x$ (50 μg mL^{-1}, pH=6.5) and then irradiated by US for 0/3/6/9 min (1 MHz, 50% duty cycle, 1.5 W cm^{-2}). A fluorescence spectrophotometer was used to excite at 488 nm, and the peak at 525 nm was recorded.

S5. *In vitro* detection of oxygen generation: The oxygen (O_2) produced was measured by a DO-958-BF portable dissolved oxygen meter (INESA Scientific Instrument Co., Ltd., Shanghai, China). Briefly, 20 μL of H_2O_2 (10 mM) was mixed with 20 mL of CeO_2, $CeO_{2-x}S_x$, and Pt/$CeO_{2-x}S_x$ aqueous solution (material concentration of 100 μg mL^{-1}) in a 25 ℃ environment. Dissolved oxygen values were recorded every 10 seconds.

S6. *In vitro* detection of GSH consumption: 0.198 g of DTNB was prepared into 50 mL of solution A (H_2O: DMSO=4:1) with 50 mM Na_2HPO_4. GSH solution (4 mL, 2.5 mM) was mixed with Pt/$CeO_{2-x}S_x$ solution (1 mL, 50 ppm) to form solution B. 1.8 mL of solution A was taken and mixed with 0.2 mL of solution B at 0/10/20/30/40/50 min. The GSH concentration was detected by the absorbance of the reaction products of GSH and DTNB.

S7. Cell culture: L929 fibroblast cells and 4T1 murine breast cancer cells were obtained from Fuheng Biotechnology and cultured in RPMI 1640 supplemented with 1% (v/v) penicillin/streptomycin and 10% (v/v) fetal bovine serum (FBS) at 37 ℃ under 5% CO_2.

S8. Cellular uptake behavior of Pt/$CeO_{2-x}S_x$ nanocomposites: 4T1 cells were planted in 96-well plates and allowed to adhere for 24 h. After that, RhB-labelled Pt/$CeO_{2-x}S_x$ (200 μg mL^{-1}) was added and co-incubated with cells for 0, 1, 2, and 4 h.

Afterward, the cells were washed with PBS for three times and stained by DAPI for 20 min before fluorescence images by inverted fluorescence microscope.

S9. Determination of ROS generation at the cellular level: To determine the production of intracellular ROS, 4T1 cells were seeded in 96-well plates and incubated for 24 h at 37 ℃ in a light-free environment. Subsequently, the cells were co-incubated with Pt/$CeO_{2-x}S_x$ (50 μg mL^{-1}) for an additional 12 h. Afterwards, the cells were exposed to 100 μM of H_2O_2 and incubated for 4 h. Following incubation, the cells were gently washed using PBS three times. Each well was then filled with DCFH-DA, and the mixture was left to incubate for 1 h. The cells were then exposed to ultrasound (1.0 MHz, 50% duty cycle, 1.5 W cm^{-2}, 2 min). Finally, the intracellular fluorescence intensity was measured to confirm the presence of ROS production.

S10. GSH depletion at the cellular level: Intracellular GSH consumption was tested by the micro-reduced glutathione assay kit. Briefly, cells were incubated with Pt/$CeO_{2-x}S_x$ for 24 h under different conditions. Then, the cells were treated with US irradiation (1.0 MHz, 50% duty cycle, 1.5 W cm^{-2}, 2 min). The amount of GSH was detected using the micro-reduced glutathione assay kit (Nanjing Jianjian Bioengineering Institute, A006-2-1) according to the manufacturer's instructions.

S11. Cytotoxicity of Pt/$CeO_{2-x}S_x$ at cellular level: The biocompatibility of Pt/$CeO_{2-x}S_x$ was evaluated using normal L929 murine fibroblast cells by CCK-8 assay. In brief, the cells were seeded in 96-well plates and grown in 5% CO_2 at 37 °C overnight. Then, different concentrations of Pt/$CeO_{2-x}S_x$ were added to the medium, and the cells were incubated in 5% CO_2 at 37 °C for another 24 h. At the end of incubation, 10 μL of CCK-8 solution was added to each well and the cells were incubated for another 1 h. Finally, the plate was examined using a microplate reader at the wavelength of 450 nm.

S12. *In vitro* SDT effects of Pt/$CeO_{2-x}S_x$: 4T1 cells were cultured in 96-well plates at 37 ℃ for 24 h in a light-free environment. Subsequently, the cells were co-incubated with different concentrations of Pt/$CeO_{2-x}S_x$ (20, 40, 80, 160, 320 μg mL^{-1}) for 24 h. Following this, the cells were incubated with 100 μM H_2O_2 for 4 h. Afterwards, the cells received US irradiation (1.0 MHz, 50% duty cycle, 1.5 W cm^{-2}) for 5 min. Following the treatment, 10 μL of CCK-8 was added to each well, and the cells were incubated for an additional hour. Finally, the microplate reader was utilized to examine

the plate at a wavelength of 450 nm.

S13. Live/dead assay: 4T1 cells were inoculated in 96-well plates, incubated at 37 ℃ for 24 h away from light, and then co-incubated with $Pt/CeO_{2-x}S_x$ (200 μg mL^{-1}) for 24 h. The cells were then further incubated with 100 μM H_2O_2 for 4 h. The cells were then treated with US irradiation (1.0 MHz, 50% duty cycle, 1.5 W cm^{-2}, 5 min). Gently wash the cells with PBS 3 times. Then a fresh medium containing Calcein-AM and PI was added. After staining for 20 min, the cells were washed with PBS and imaged by inversed fluorescent microscope.

S14. Tumor Model: The mice used in the animal experiments were female BALB/c mice aged 6-8 weeks. They were purchased from Changsheng Biotechnology. All of the animal experiments were approved by the Institutional Animal Care and Use Committee of Changchun Institute of Applied Chemistry (IACUC) (Grant no. 20220001).

S15. *In vivo* biocompatibility of $Pt/CeO_{2-x}S_x$ nanocomposites: Healthy female BALB/c mice were intravenously injected with $Pt/CeO_{2-x}S_x$ nanocomposites (5 mg kg^{-1}, 100 μL). The body weight was measured every 2 days to evaluate its *in vivo* biosafety. At the indicated time, the mice were euthanized, and their major organs, including the heart, liver, spleen, lung, and kidney, were harvested for H&E staining.

S16. *In vivo* biodistribution of $Pt/CeO_{2-x}S_x$: 1 mg of ICG was mixed with PBS solution of $Pt/CeO_{2-x}S_x$ (1 mg mL^{-1}) and stirred overnight. Excess ICG was removed by centrifugation, and then ICG-$Pt/CeO_{2-x}S_x$ nanoparticles (100 μL, 5 mg kg^{-1}) were administered intravenously to 4T1 tumor-bearing mice. Mice were dissected at 12, 24, 48, and 72 h after administration. NIR-II imaging was utilized to visualize tumor tissues and various major organs such as the heart, liver, spleen, lungs, and kidneys following their excision.

S17. Tumor suppression experiments *in vivo*: To establish a tumor model, 4T1 breast cancer cells suspended in PBS were subcutaneously injected into the right leg of female BALB/c mice. Once the tumor volume reached approximately 150 mm^3, the mice were randomly divided into five groups, each consisting of five mice: (i) control, (ii) US, (iii) $Pt/CeO_{2-x}S_x$, (iv) $CeO_{2-x}S_x$+US, and (v) $Pt/CeO_{2-x}S_x$+US. Subsequently, the mice were intravenously administered 100 μL of $CeO_{2-x}S_x$ or $Pt/CeO_{2-x}S_x$ NPs (5 mg kg^{-1}). In the

groups subjected to ultrasound irradiation, tumors were exposed to ultrasound after intravenous injection for 12, 24, 48, and 72 h (1.0 MHz, 50% duty cycle, 1.5 W cm^{-2}, 5 min). Throughout the treatment period, the size of the tumors and the body weight of the mice were monitored every two days.

S18. Hemolysis assay: BALB/c mice were used to harvest red blood cells in order to assess the *in vitro* hematotoxicity of $Pt/CeO_{2-x}S_x$. To get rid of the white blood cells, the red blood cells were first separated by centrifugation and then three times through PBS washing. Following this, different quantities of $Pt/CeO_{2-x}S_x$ were added to the red blood cells (19, 38, 75, 150, 300, and 600 μg mL^{-1}). After being left at room temperature for 6 h, the mixture was collected using centrifugation. The positive and negative groups were water and PBS, respectively. After gathering the supernatant, the absorbance at 540 nm was recorded. The following formula was used to get the hemolysis ratio: hemolysis rate (%) = (sample absorption - negative control absorption) / (positive control absorption - negative control absorption) * 100%.

S19. *In vivo* biocompatibility of $Pt/CeO_{2-x}S_x$ nanocomposites: Healthy female BALB/c mice were intravenously injected with $Pt/CeO_{2-x}S_x$ nanocomposites. The body weight was measured every 2 days to evaluate its *in vivo* biosafety. At the indicated time, the mice were euthanized and their blood samples were collected to perform blood panel analysis and blood biochemistry assay.

S20. Determination of O_2 generation at the cellular level: The intracellular O_2 generation was monitored using $[Ru(dpp)_3]Cl_2$ (RDPP) probe. 4T1 cells were first planted into 96-well plates for 24 h at 37 ℃ in the dark. Subsequently, the cells were incubated with RDPP (10 mg/L, 10μL) for 12 h. The cells were then incubated with $Pt/CeO_{2-x}S_x$ (40 μg mL^{-1}) and H_2O_2 (100 μM) for 4, 8, 12, and 24 h in a hypoxic chamber, respectively. Then, the cells were washed with PBS three times. Finally, the fluorescence of RDPP was observed through inversed fluorescent microscope.

S21. Statistical analysis: Significant differences between groups were determined using two-tailed tests, and P-values less than 0.05 were considered statistically significant (*$P < 0.05$, **$P < 0.01$, ***$P < 0.001$). All data were analyzed using Origin and Excel.

S22. XRD pattern of $Pt/CeO_{2-x}S_x$, $CeO_{2-x}S_x$, and CeO_2

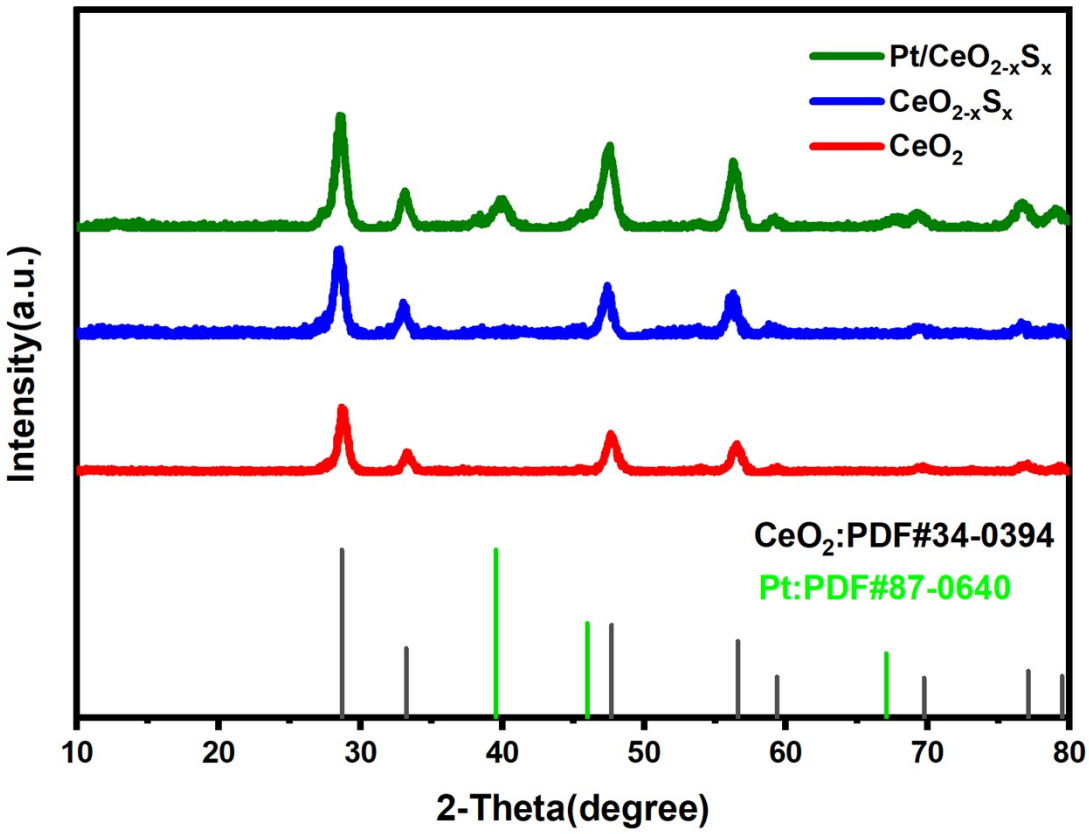

Figure S1. XRD pattern of $Pt/CeO_{2-x}S_x$, $CeO_{2-x}S_x$, and CeO_2.

S23. Zeta potential of $CeO_{2-x}S_x$, Pt, and $Pt/CeO_{2-x}S_x$.

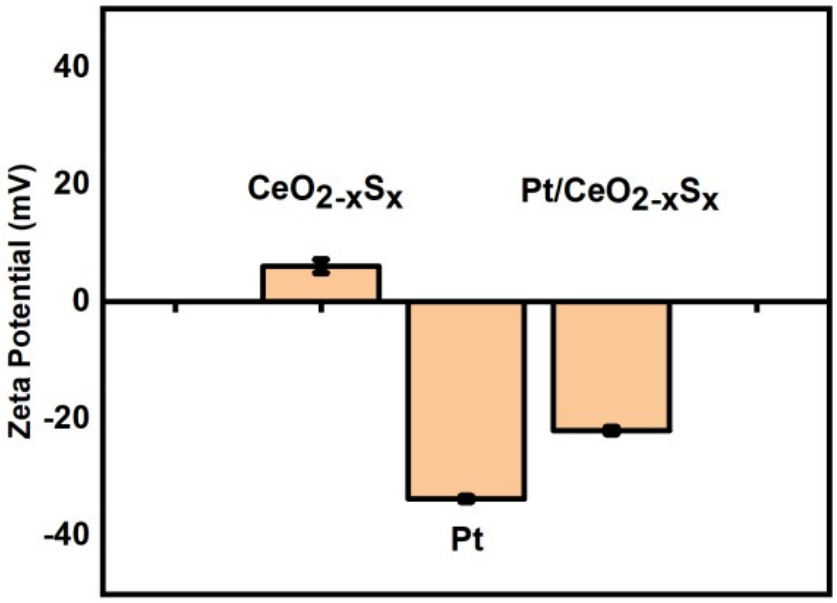

Figure S2. Zeta potential of $CeO_{2-x}S_x$, Pt, and $Pt/CeO_{2-x}S_x$.

S24. The size distribution profiles of CeO_2, $CeO_{2-x}S_x$, and $Pt/CeO_{2-x}S_x$ dispersed in H_2O measured by Dynamic light scattering (DLS) analysis

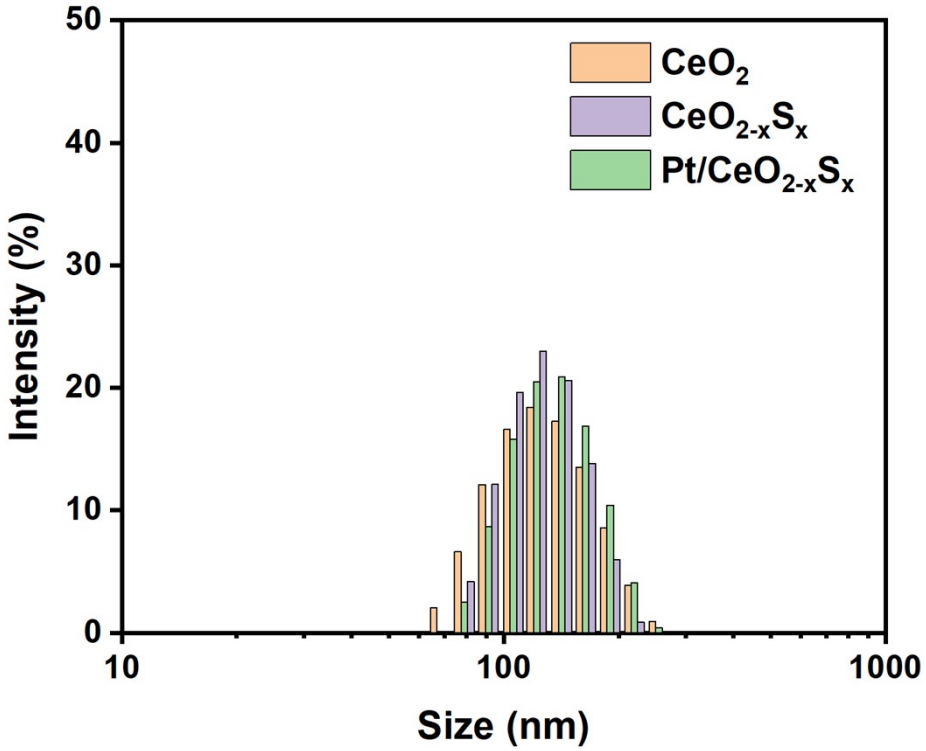

Figure S3. The size distribution of CeO_2, $CeO_{2-x}S_x$, and $Pt/CeO_{2-x}S_x$ dispersed in H_2O determined by dynamic light scattering (DLS).

S25. Dispersion stability of $Pt/CeO_{2-x}S_x$

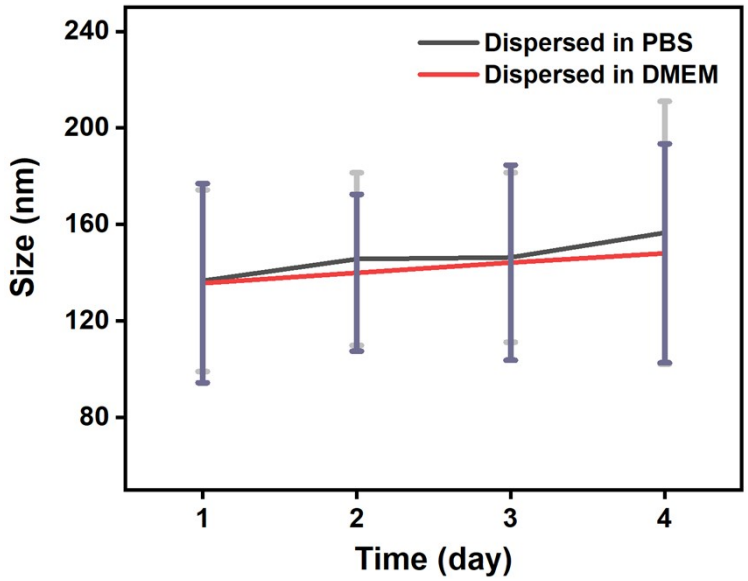

Figure S4. Changes in hydrodynamic size of $Pt/CeO_{2-x}S_x$ suspended in PBS buffer and DMEM, respectively.

S26. XPS spectrum of $Pt/CeO_{2-x}S_x$

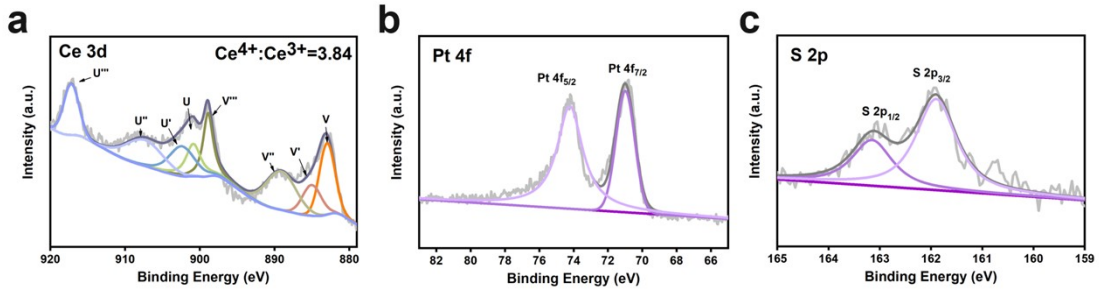

Figure S5. XPS spectrum of $Pt/CeO_{2-x}S_x$. (a) XPS spectrum of Ce 3d of $Pt/CeO_{2-x}S_x$. (b) XPS spectrum of Pt 4f of $Pt/CeO_{2-x}S_x$. (c) XPS spectrum of S 2p of $Pt/CeO_{2-x}S_x$.

S27. The depletion of glutathione by $Pt/CeO_{2-x}S_x$ under US using DTNB as a trapping agent

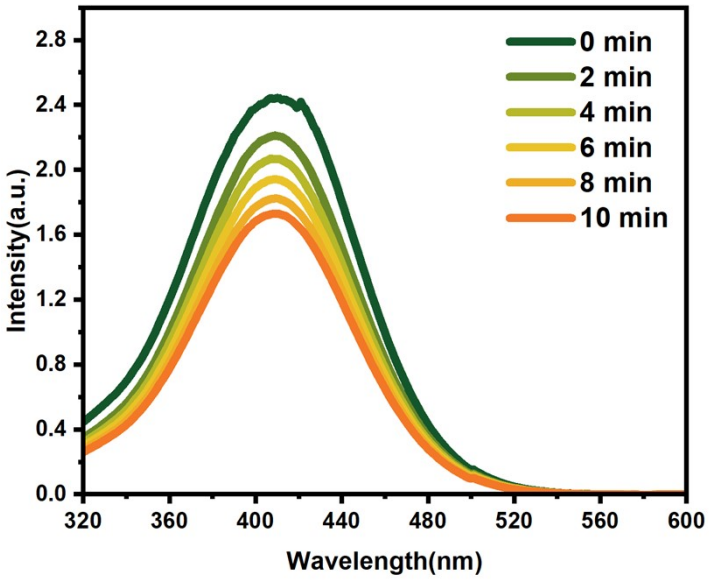

Figure S6. The depletion of glutathione by $Pt/CeO_{2-x}S_x$ under US using DTNB as a trapping agent (1.0 MHz, 50% duty cycle, 1.5 W cm^{-2}).

S28. *In vitro* cellular uptake of RhB-conjugated Pt/$CeO_{2-x}S_x$

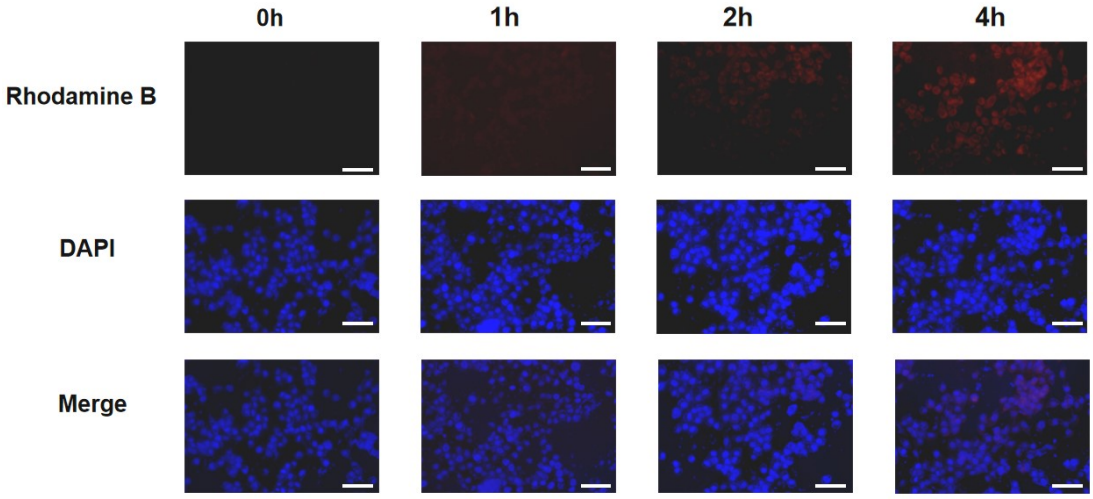

Figure S7. *In vitro* cellular uptake of RhB-conjugated Pt/$CeO_{2-x}S_x$, Scale bars = 50 μm.

S29. H&E staining images of control and therapy groups

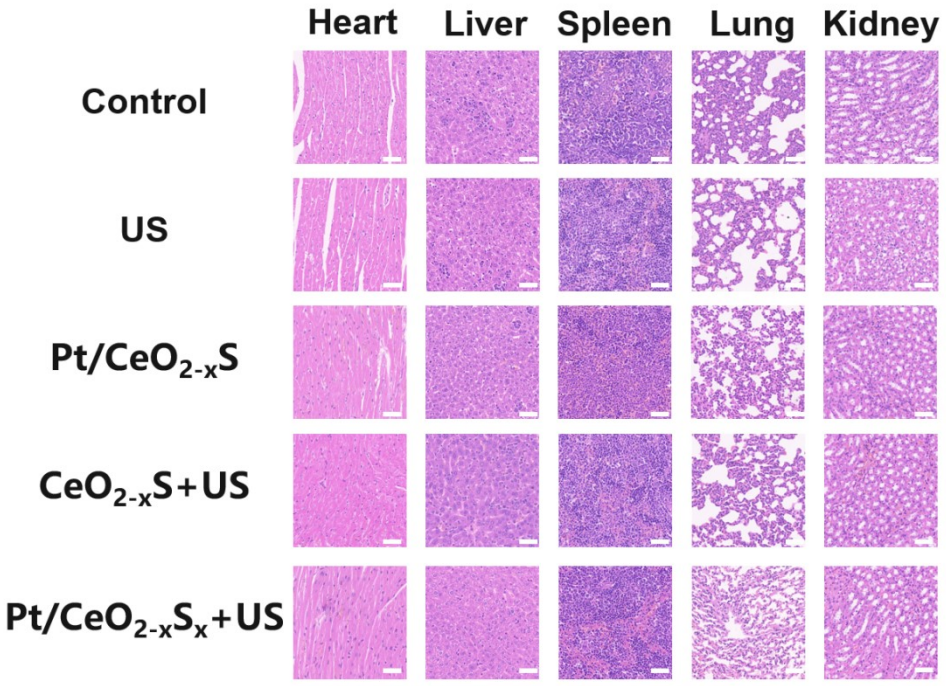

Figure S8. H&E staining images of control and therapy groups (Scale bars=50 μm).

S30. Blood routine examination

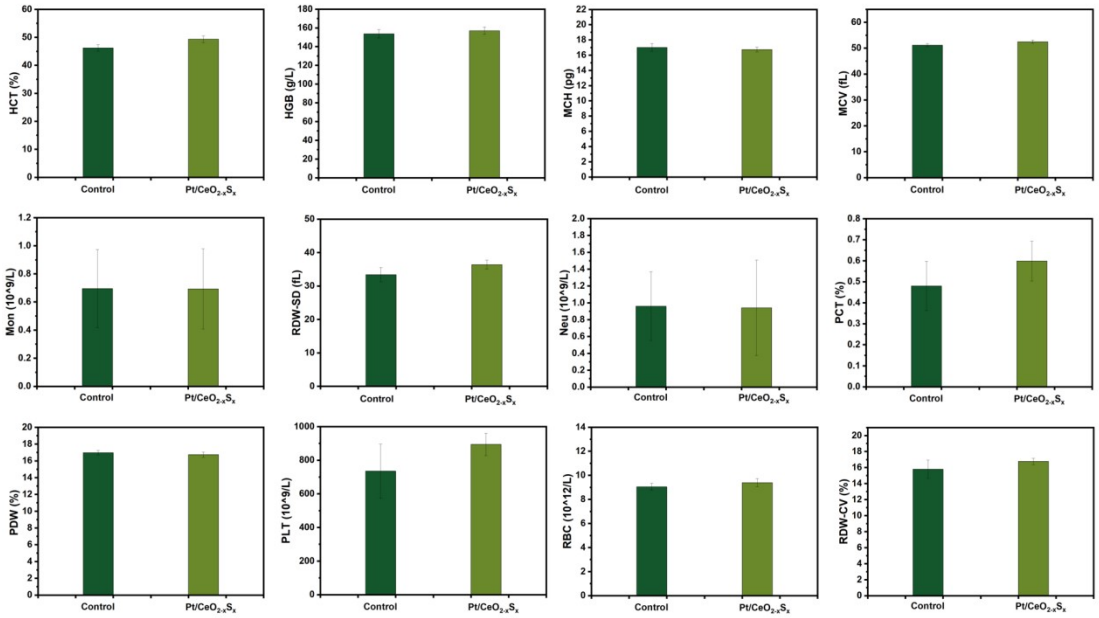

Figure S9. Blood routine indexes of healthy female BALB/c mice treated with $Pt/CeO_{2-x}S_x$ for 30 days. Data are represented as mean ± SD (n = 3).

S31. Blood biochemistry

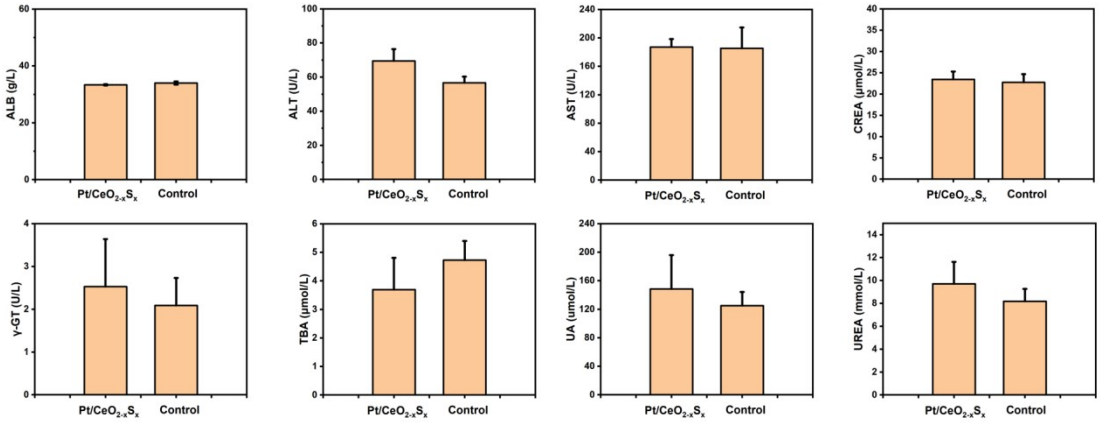

Figure S10. Blood biochemistry indexes of female BALB/c mice treated with $Pt/CeO_{2-x}S_x$ for 30 days. Data are represented as mean ± SD (n = 3).

S32. Digital photograph of hemolysis test and hemolysis rate of Pt/$CeO_{2-x}S_x$ nanocomposites

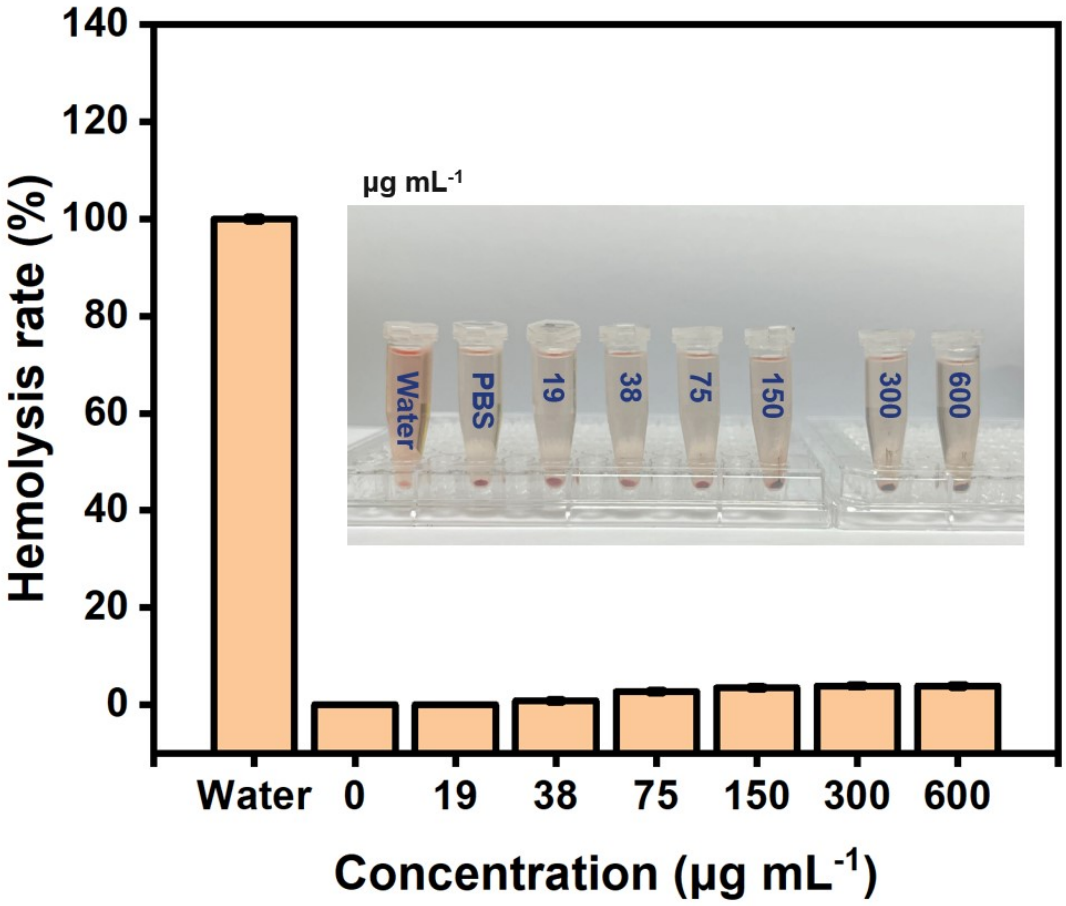

Figure S11. Digital photograph of hemolysis test and hemolysis rate of Pt/$CeO_{2-x}S_x$ nanocomposites.