Supporting Information

Intelligent NIR-IIb responsive lanthanide@metal-organic framework core-shell nano-catalyst for combined deep-tumor therapy

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Chemicals and Materials

Rare earth YCl_3•$6H_2O$ (99.99%), $ErCl_3$•$6H_2O$ (99.99%), $LuCl_3$•$6H_2O$ (99.99%) were purchased from QingDa elaborate Chemical Reagent Co. Ltd (Shandong). Oleic acid (OA, 90%) and 1-Octadecene (90%, ODE), NH_4F and NaOH, CH_3OH (99.5%), anhydrous alcohol, polymers acrylic acid (PAA), indocyanine green molecules (ICG), N,N Dimethylformamide (DMF), 3-aminopropyl) triethoxysilane (APTES), Meso tetra(4 carboxyphenyl) porphine (TCPP), 1,4-Diphenyl-2,3-benzofuran (DPBF), nitrosonium tetrafluoroborate ($NOBF_4$), and all other reagents were supplied by Sigma-Aldrich. All chemical reagents were used without further purification.

Characterization

Transmission electron microscopy **(**TEM, FEI Tecnai F30) equipped with the energy dispersive X-ray spectroscopy (EDS, Oxford Xplore Instrument) system was used to test the morphology of the nanoparticles with an accelerating voltage of 300 kV. X-ray diffraction (XRD) was demonstrated on a Bruker D Advance system operating at 40 kV and 250 mA. The upconversion emission spectra were obtained with the Edinburgh FLS980 system at room temperature. The UV-vis absorbance spectra were tested by the spectrophotometer system (UV-1801, Beijing Beifen-Ruili Analytical Instruments Group Co., Ltd). Inductively coupled plasma mass spectrometry (ICP-MS) was performed on a PerkinElmer NexION 300X; iCAP6300. The thermal images were acquired with a HIKMICR K20 thermal camera.

Fabrication of $NaLuF_4$:30% Er core nanoparticles

The $NaLuF_4$:30% Er core nanoparticles were fabricated by using the high-temperature co-precipitation method [s1]. First, 0.7 mmol of $LuCl_3$, 0.3 mmol of $ErCl_3$, 30 mL of ODE and 12 mL of OA were mixed into a 100 ml three-neck flask, followed by constantly stirring at 160 ℃ for 1 h. The mixture solution was then cooled down to room temperature. Next, 10 mL of NaOH methanol solution (0.01 g/mL) and 20 mL of NH_4F methanol solution (0.0074 g/mL) were

immediately added into the solution and kept stirring for 1 h at room temperature. After that, the mixture solution was heated to 305 ℃ for 60 min and then cooled down to room temperature. All the procedures were demonstrated under N_2 protection. The resulting products were obtained through centrifugation at 5000 rpm and washed with ethanol for three times, the final product was redispersed in 5 mL of cyclohexane.

Synthesis of $NaLuF_4$:30%Er@$NaYF_4$ core-shell nanoparticles

Typically, 1 mmol of YCl_3, 30 mL of ODE and 12 mL of OA were added into a 100 mL three-neck flask. Then, the mixture solution was heated to 160 ℃ and maintained at 160 ℃ for 1 h. The mixture was then cooled down to room temperature, the NaOH methanol solution (0.01 g/mL), 20 mL of NH_4F methanol solution (0.0074 g/mL) and 5 mL of the prepared core nanoparticles were poured into the mixture solution and kept stirring for 1 h. Next, the solution was heated up to 305 ℃ and kept stirring for 60 min. All the procedures were demonstrated under N_2 protection. The final nanoparticles were obtained by centrifugation and washed with ethanol for three times, the final products were redispersed in cyclohexane.

Synthesis of PAA-modified $NaLuF_4$:30%Er@$NaYF_4$ core-shell nanoparticles

The PAA-modified $NaLuF_4$:30%Er@$NaYF_4$ core-shell nanoparticles were synthesized according to the previous work [s1]. 0.25 mmol of the $NaLuF_4$:30%Er@$NaYF_4$ core-shell nanoparticles in 5 mL of hexane was added into 5 mL of $NOBF_4$ DMF solution (0.01 M) and kept gently shaking for 1 h. The nanoparticles were obtained by centrifugation and washed with 5 mL of DMF and 10 mL of toluene. Next, the obtained nanoparticles were redispersed in 5 mL of DMF containing 200 mg PAA under vigorous stirring. The mixture solution was heated at 80 ℃ for 30 min. The product was precipitated by centrifugation and washed with water for three times. The obtained PAA-modified $NaLuF_4$:30%Er@$NaYF_4$ core-shell nanoparticles were dispersed in water.

Preparation of UCNPs@ZrMOF nanocomposite

The ZrMOF shell was coated on the UCNPs by using an in situ growth approach based on the

previous works [s2]. 30 mg of PAA-UCNPs and 30 mg $ZrOCl_2$ were added into 5 mL of DMF solution and kept stirring for 1 h. Then, 2 mL of TCPP DMF solution (5 mg/mL), 2 mL of benzoic acid DMF solution (140 mg/mL) and 3 mL DMF were introduced into the mixture solution and sonicated for 5 min. After that, the mixture solution was cultured at 90 ℃ for 5 h. Finally, the UCNPs@ZrMOF nanocomposite was obtained by centrifugation and redispersed in water.

Synthesis of UCNPs@ZrMOF@ICG nanocomposite

The ICG molecule was loaded into the UCNPs@ZrMOF nanocomposite according to our previous reports [s3]. For ICG loading, 5 mg of ICG was added into the 5 mL of UCNPs@ZrMOF water solution. After stirring for 12 h, the nanocomposite was centrifugated and washed with water.

Measurement of the förster resonance energy transfer (FRET) efficiency

The FRET efficiency was calculated according to the following formula:

$$E = 1 - \frac{I}{I_0}$$

Where I and I_0 represent the luminescence intensities of the donors in the presence of acceptors (ZrMOF), and in the absence of acceptors, respectively

ROS generation ability evaluation

The ROS production ability was tested by the DPBF. Typically, 50 μL of DPBF DMSO solution was added into 2 mL of UCNPs@ZrMOF@ICG solution (2 mg/mL). The mixture solution was irradiated by a 1532 nm laser with different power density. The character absorption peak at 417 nm was recorded by using the spectrophotometer system.

***In vitro* therapy test**

The HCT 116 cells were first cultured in the DEME solution containing fetal bovine serum (FBS) (10%) and penicillin-streptomycin (1%). For assessing the biocompatibility of the UCNPs@ZrMOF@ICG nanocomposite, the cells in 96-well microplate were cultivated with a

fresh culture medium containing various contents of UCNPs@ZrMOF@ICG nanocomposite (0, 100, 200, 400, 800, 1000 μg/mL). The cell survival rate was then measured by using the standard MTT assay.

For *in vitro* therapeutic efficiency test, the cells were divided into 6 groups: (1) PBS; (2) PBS + 1532/808 nm; (3) UCNPs@ZrMOF; (4) UCNPs@ZrMOF@ICG; (5) UCNPs@ZrMOF@ICG + 1532 nm; (6) UCNPs@ZrMOF@ICG + 1532/808 nm. After different treatments, the cell survival rate was also tested by using the standard MTT method. The living/dead cell staining test was performed to directly visualize the therapeutic effect. The tumor cells were divided into 6 groups with various treatments: (1) PBS; (2) PBS + 1532/808 nm; (3) UCNPs@ZrMOF; (4) UCNPs@ZrMOF@ICG; (5) UCNPs@ZrMOF@ICG + 1532 nm; (6) UCNPs@ZrMOF@ICG + 1532/808 nm. After another 12 h incubation, 500 μL of Calcein-AM/PI mixed liquor was added and cultured for 20 min. The living/dead stained results images were acquired with a fluorescence-inverted microscope.

Photothermal conversion efficiency evaluation

The UCNPs@ZrMOF@ICG nanocomposite (600 μg /mL) was treated with 808 nm laser for 6 min. A HIKMICR K20 thermal camera was used to record the temperature. The photothermal conversion efficiency can be determined by using the equation 1 [s4]:

$$\eta = \frac{hA(T_{max} - T_{surr}) - Q_{dis}}{I(1 - 10^{-A_{808}})} \quad (1)$$

In formula (1), h represents the heat transfer coefficient, A means the surface area of the cuvette, T_{max} and T_{surr} are the equilibrium and ambient temperatures, respectively. The term A_{808} pertains to the intensity of light absorption of UCNPs@ZrMOF@ICG nanocomposite at 808 nm, Q_{dis} means the heat loss of the quartz container. The hA was determined by the eq 2:

$$hA = \frac{mc_p}{\Gamma_s} \quad (2)$$

In this formula, A represents the sample time constant. m is the solution mass, and C_p represents the heat capacity. The Γ_s was measured based on eq 3:

$$\Gamma_s = \frac{t}{\ln \theta} \quad (3)$$

Where Γ_s was calculated based on linear time data to -ln θ.

Intracellular ROS generation ability test

The HCT 116 cells were cultured in a 12 well microplate. After incubation for 24 h, the cell culture medium was replaced by a fresh cell culture medium. The cells were divided into six groups with the following treatments: (1) PBS; (2) PBS + 1532/808 nm; (3) UCNPs@ZrMOF; (4) UCNPs@ZrMOF@ICG; (5) UCNPs@ZrMOF@ICG + 1532 nm; (6) UCNPs@ZrMOF@ICG + 1532/808 nm. After culturing for another 4 h, the HCT 116 cells were stained with 2',7'-dichlorodihydrofluorescein (DCFH-DA) and 4',6-diamidino-2-phenylindole (DAPI) for 30 min. The fluorescence images were obtained by a fluorescence inverted microscope.

Animal models

1×10^6 HCT 116 cells were subcutaneously injected into the BALB/C nude mice. The nude mice were cultured until the tumor had grown to 100 mm^3 for further use. All animal experiments have been approved by the institutional animal use guidelines of Changsha University of Science and Technology and Animal Ethics Committee of Hunan Province.

***In vivo* antitumor treatment evaluation**

The HCT 116 tumor bearing mice were split into 5 groups at random: (1) PBS; (2) PBS + 808/1532 nm; (3) UCNPs@ZrMOF@ICG; (4) UCNPs@ZrMOF@ICG + 1532 nm; (5)

UCNPs@ZrMOF@ICG + 1532/808 nm. The tumor bearing mice were injected with PBS (100 μL) for groups (1) and (2), UCNPs@ZrMOF@ICG (15 mg/Kg) for groups (3), (4) and (5), respectively. After 24 h of injection, the 1532 nm laser irradiation was demonstrated on groups (2), (4), and (5) for 10 min, and the 808 nm laser irradiation was performed on groups (2) and (5) for 10 min. The laser exposure treatment was demonstrated every 2 days. The injection treatment was performed every 4 days. The body weight and tumor volume of the tumor-bearing mice were recorded every 2 days. The tumor volumes were calculated according to the following formula: $V = (a \times b^2)/2$, a and b indicate the length and width of the tumor, respectively. The tumors were then dissected from the tumor-bearing mice after 14 days of treatments and were sliced up for HE and TUNEL staining.

In vivo pharmacokinetics test

The Kunming mice were first intravenously injected with the UCNPs@ZrMOF@ICG nanocomposite (100 μL, 3 mg/mL) for pharmacokinetics test. The blood samples were received from the mice at various time points post-injection. The concentrations of Lu^{3+} in the blood were measured by using ICP-MS analysis.

In vivo biocompatibility evaluation

The control group was not injected, while the normal BALB/C nude mice were intravenously injected with UCNPs@ZrMOF@ICG nanocomposite. After injection for 15 and 30 days, the treated groups and control group were dissected, the main organs (heart, liver, spleen, lung, and kidney) were obtained and stained with H&E for further analysis.

Statistical analysis

The test results were presented as mean ± standard deviation (S.D). We conducted a significance level analysis using a student's t-test.: $*p < 0.05$, $**p < 0.01$, $***p < 0.001$.

s1. Y. B. Li, M. Y. Jiang, Z. L. Xue, S. J. Zeng. *Theranostics* 2020, **10**, 6875.

s2. X. T. Zhao, Y. B. Li, L. M. Du, Z. M. Deng, M. Y. Jiang, S. J. Zeng, *Adv. Healthcare Mater.* 2021, **10**, 2101174.

s3. J. Q. Huang, Y. Huang, Z. L. Xue, S. J. Zeng, *Biomaterials* 2020, **262**, 120346.

s4. Y. B. Li, G. X. Bai, S. J. Zeng, J. H. Hao. *ACS. Appl. Mater. Interfaces* 2019, **11**, 4737.

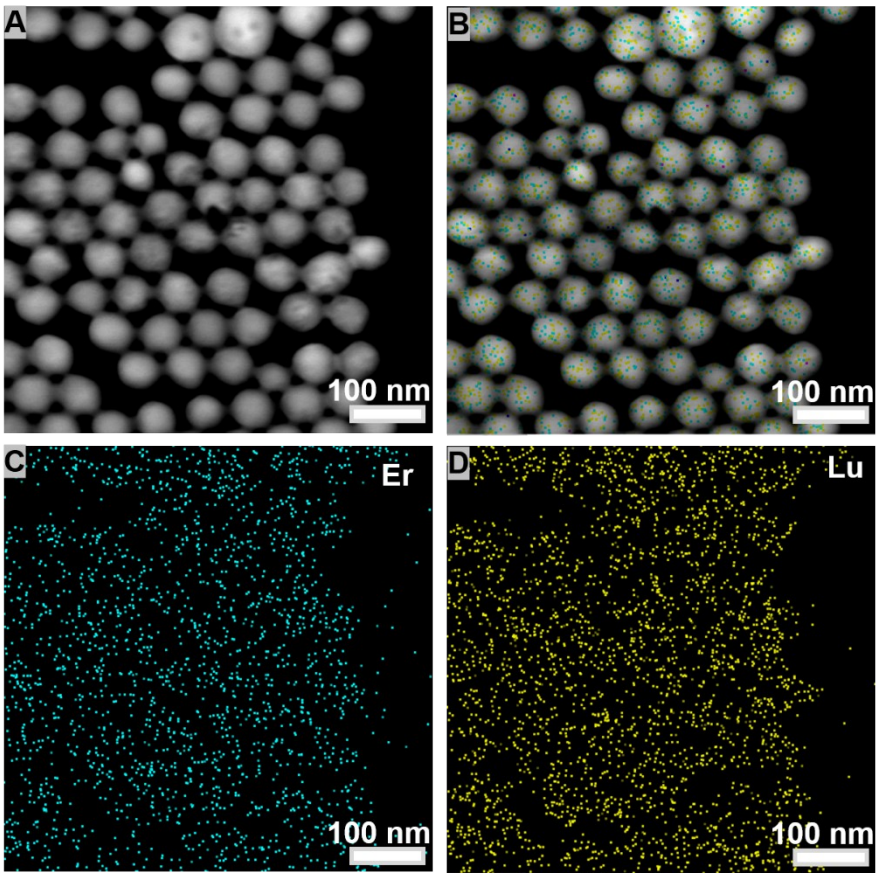

Figure S1. (A)-(D) Scanning TEM (STEM) pictures of $NaLuF_4$:30%Er core nanoparticles and the corresponding elemental mappings results.

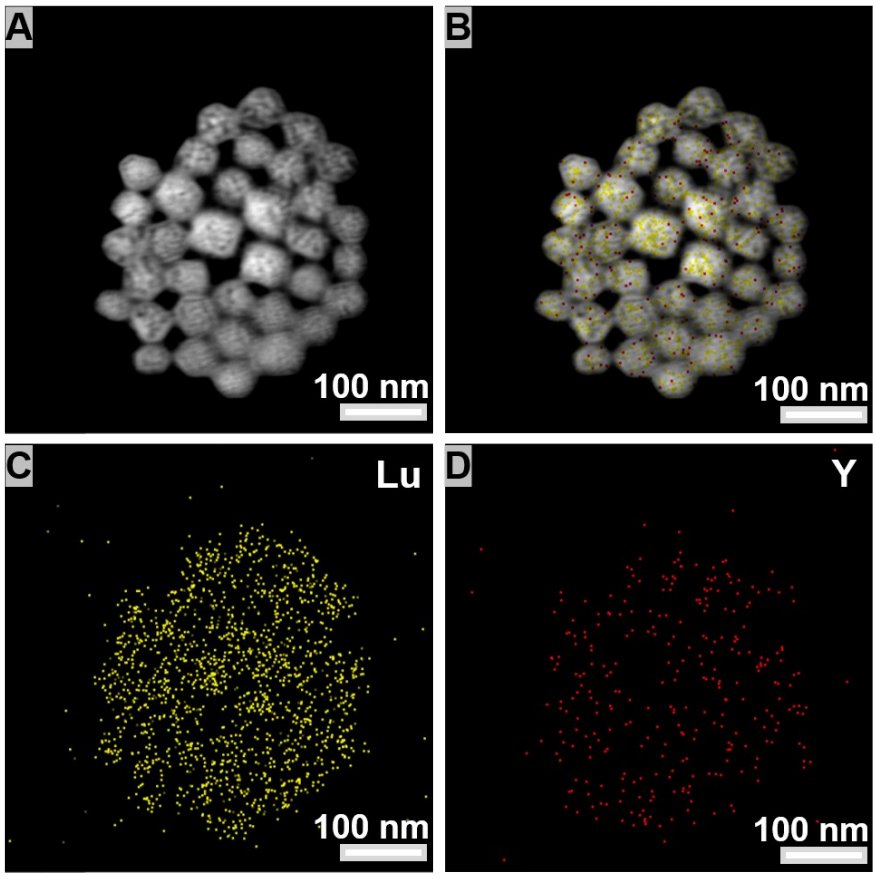

Figure S2. (A)-(D) STEM pictures of $NaLuF_4$:30%Er@$NaYF_4$ core-shell nanoparticles and the corresponding elemental mappings results.

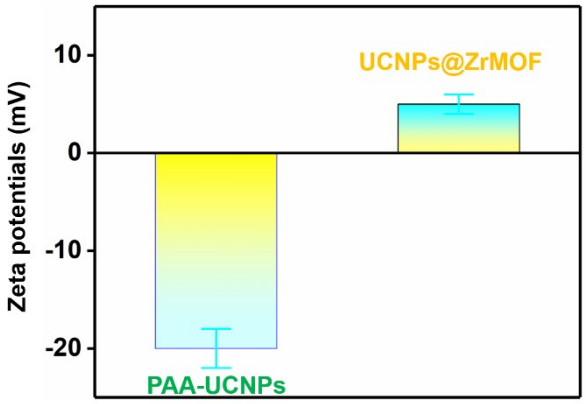

Figure S3. The zeta potential of the PAA-UCNPs and UCNPs@ZrMOF nanocomposite.

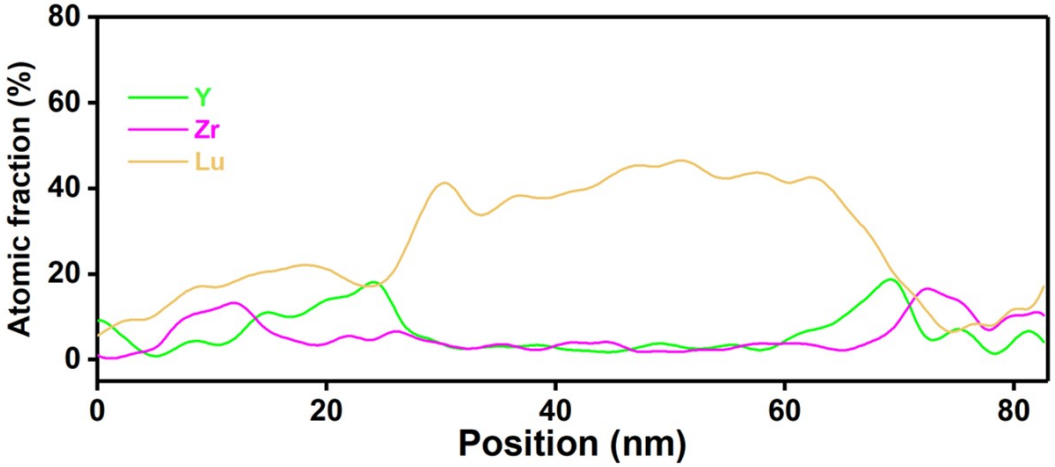

Figure S4. EDS line scan result of the as-prepared UCNPs@ZrMOF nanocomposite in Figure 1H.

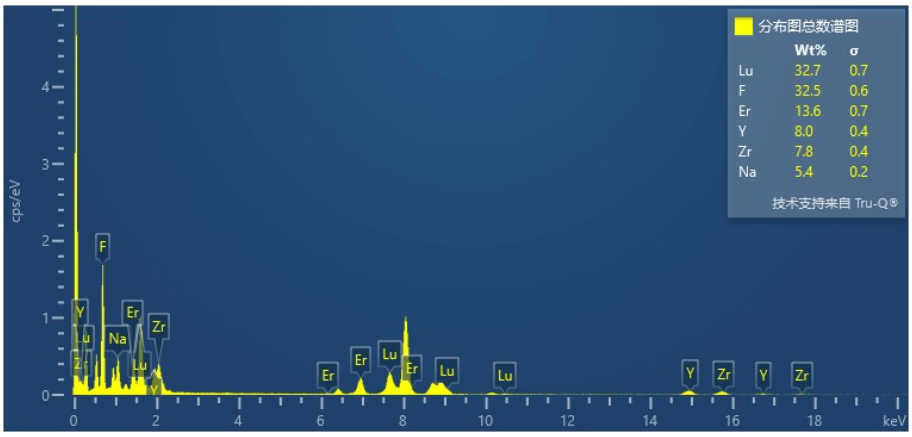

Figure S5. Energy dispersive X-ray spectrometer result of the as-prepared UCNPs@ZrMOF nanocomposite.

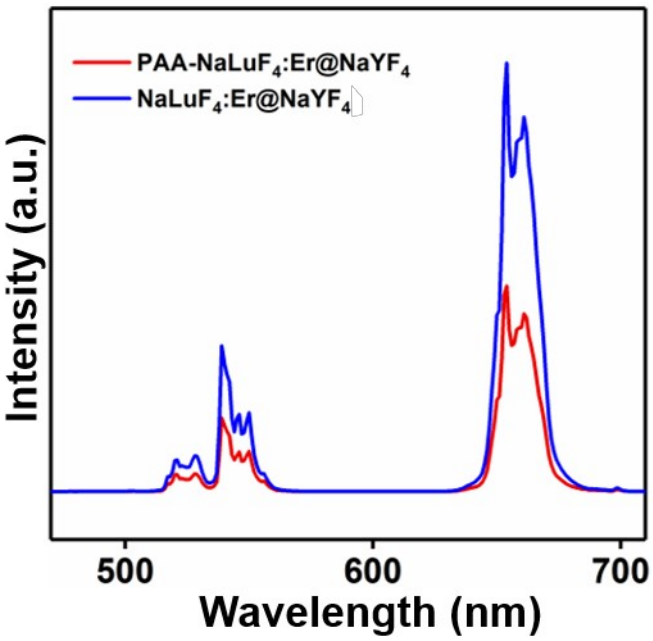

Figure S6. The upconversion emission spectra of the $NaLuF_4$:Er@$NaYF_4$ core-shell nanoparticles and PAA-$NaLuF_4$:Er@$NaYF_4$ nanoparticles.

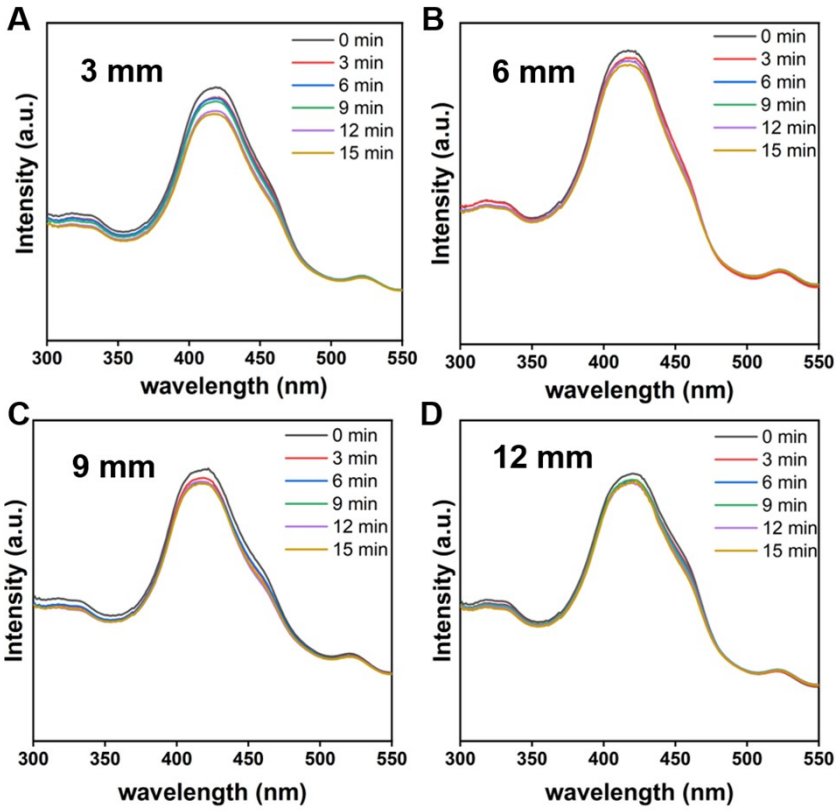

Figure S7. 1532 nm light triggered ROS generation of the UCNPs@ZrMOF nanocomposite covered with different thicknesses of pork tissues.

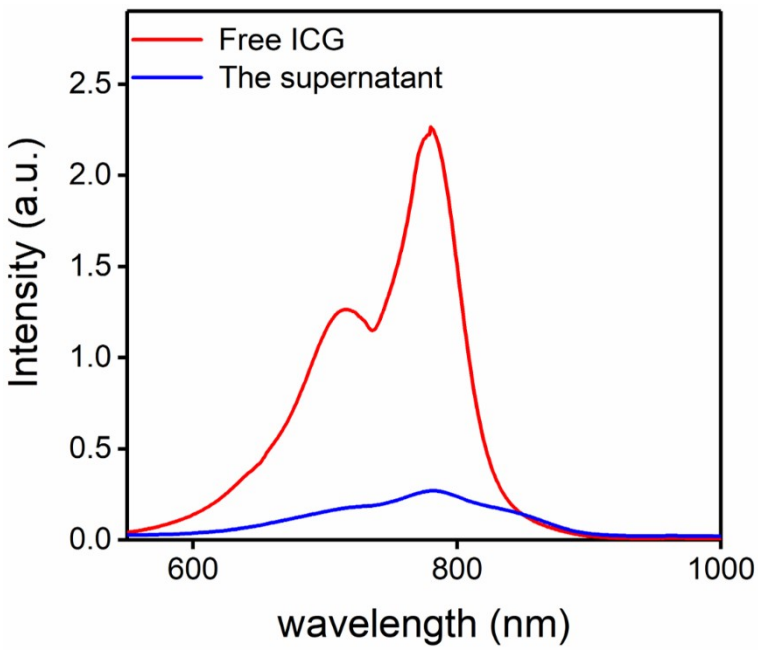

Figure S8. The absorbance spectra of the free ICG solution and the supernatant solution of the UCNPs@ZrMOF@ICG nanocomposite after centrifugation.

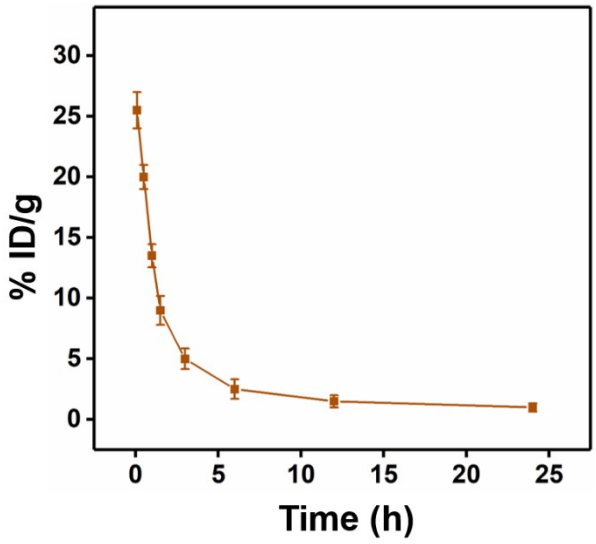

Figure S9. The blood circulation half-life time of the UCNPs@ZrMOF@ICG nanocomposite *in vivo*.

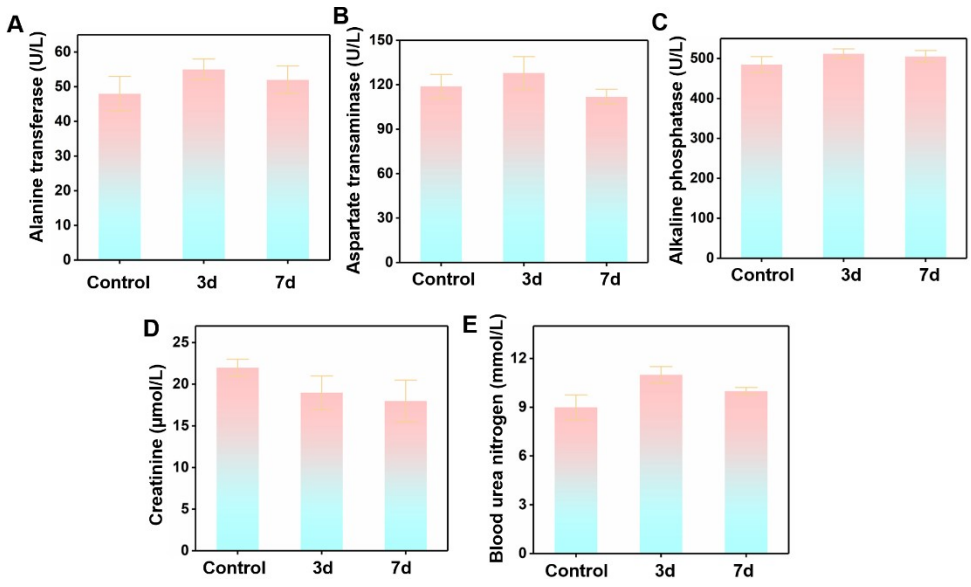

Figure S10. *In vivo* blood biochemistry tests of the mice after intravenous injection of UCNPs@ZrMOF@ICG nanocomposite and control mice with injection of PBS.

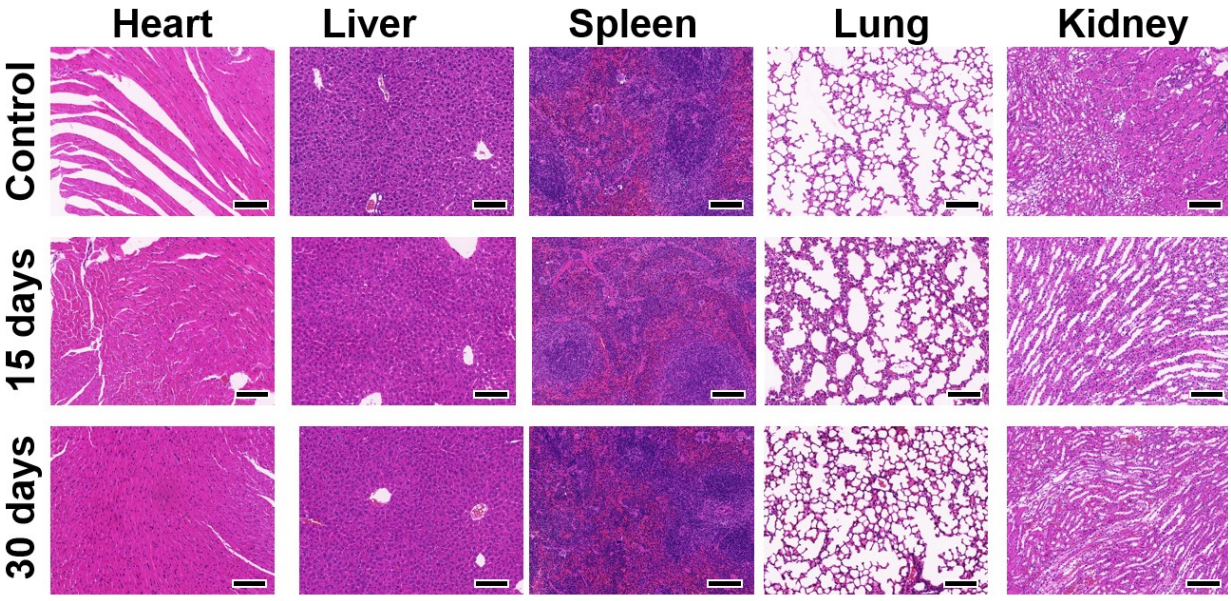

Figure S11. H&E stained heart, liver, spleen, lung and kidney obtained from the mice after injection of UCNPs@ZrMOF@ICG nanocomposite for 15 and 30 days and control groups without injection. All the scale bars are 100 μm.