

Degradation of Upconverting Nanoparticles in Simulated Fluids Evaluated by Ratiometric Luminescence

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Experimental Section

Materials: Gd(CH₃CO₂)₃·xH₂O (99.9%), Eu(CH₃CO₂)₃·xH₂O (99.9%), Yb(CH₃CO₂)₃·xH₂O (99.9%), Tm(CH₃CO₂)₃·xH₂O (99.9%), Tb(CH₃CO₂)₃·xH₂O (99.9%), NaOH (>98%), NH₄F (>98%), 1-octadecene (ODE, 90%), oleic acid (OA, 90%), serum, and cell culture medium were all purchased from Sigma Aldrich. S9 microsomal was obtained from Xinrun Biotechnology Co., Ltd. Wuxi, China. Ethanol (AR), cyclohexane (AR), hydrochloric acid (AR), nitric acid (MOS grade), and hydrogen peroxide (MOS grade) were purchased from Sinopharm Chemical Reagent Co., Ltd. Shanghai, China. All chemicals were directly used without further purification.

Synthesis of NaGdF₄:15%Eu core nanoparticles: NaGdF₄:15%Eu core nanoparticles were prepared using a classical thermal decomposition method with a modified procedure.¹ Briefly, 2 mL water solution of Gd(CH₃CO₂)₃ (0.34 mmol) and Eu(CH₃CO₂)₃ (0.06 mmol) were added into a 50 mL flask along with OA (6 mL) and ODE (4 mL). The mixture was then heated to 150 °C and retained for 1 hour to remove water until the solution became clear and transparent. After cooling down to 50 °C, 4.4 mL of a methanol solution containing NH₄F (0.05 g, 1.36 mmol) and NaOH (0.04 g, 1 mmol) was quickly added into the mixture and stirred for 30 min. Subsequently, the generated mixture was heated to 100 °C and then in vacuo for 20 min to evaporate the remaining methanol. The generated solution was heated to 300 °C under nitrogen and maintained for 1.5 h, and then cooled down to room temperature. The resulting nanoparticles were precipitated by the addition of ethanol, collected by centrifugation at 8000 rpm for 5 min. After washing three times with ethanol, the core nanoparticles were redispersed in 4 mL of cyclohexane.

Synthesis of NaGdF₄:15%Eu@NaGdF₄:49%Yb,1%Tm (Eu@Tm) core-shell nanoparticles: NaGdF₄:15%Eu@NaGdF₄:49%Yb,1%Tm core-shell nanoparticles were prepared using a hot injection method with a modified procedure.² The first process was the preparation of shell precursors. A 2 mL of aqueous solution containing Gd(CH₃CO₂)₃ (0.2 mmol), Yb(CH₃CO₂)₃ (0.196 mmol), and Tm(CH₃CO₂)₃ (0.004 mmol) was added into a 50 mL flask along with OA (4 mL) and ODE (6 mL). The mixture was then heated to 150 °C and retained for 1 hour to remove water. After cooling down to 50 °C, 4.4 mL of a methanol solution containing NH₄F (0.05 g, 1.36 mmol) and NaOH (0.04 g, 1 mmol) was quickly added into the mixture and stirred for 0.5 h. Next, the mixture was continuously heated to 80 °C and kept in this temperature for 40 min to remove

methanol, and then cooled down to room temperature naturally.

Second, the ordinary procedure for growing NaGdF₄:49%Yb,1%Tm shells. Core nanoparticles, OA (4 mL) and ODE (6 mL) were added to a 50 mL flask. The mixture was then heated to 80 °C and maintained for 40 min to remove cyclohexane. The resulting mixture was continuously heated to 100 °C and then in vacuo for 20 min to evaporate residual cyclohexane, air, and moisture. Subsequently, the generated mixture was heated to 310 °C under the nitrogen atmosphere, and the shell precursor solution was injected into the reaction system within 20 min with a syringe pump, and then cooled down to room temperature naturally under nitrogen atmosphere. The resulting nanoparticles were precipitated by the addition of ethanol, and then collected by centrifugation at 8000 rpm for 5 min. After washing three times with ethanol, the core nanoparticles were redispersed in 4 mL of cyclohexane.

Synthesis of NaGdF₄:15%Eu@NaGdF₄:49%Yb,1%Tm@NaGdF₄:15%Tb (Eu@Tm@Tb) core-shell-shell upconversion nanoparticles: The preparation process of core-shell-shell upconversion nanoparticles were identical with the core-shell upconversion nanoparticles mentioned above.

Synthesis ligand-free nanoparticles: Ligand-free nanoparticles were prepared with a modified procedure.³ 1 mL of Eu@Tm@Tb UCNPs was taken from 4 mL of the cyclohexane dispersion of core-shell-shell nanoparticles and precipitated by adding ethanol. Subsequently, 2 mL of ethanol was added and sonicated for 5 min to redisperse, 1 mL of a HCl solution (0.2 M) was added and sonicated for 10 min to remove the oleic acid ligand. After reaction, the nanoparticles were collected by 12000 rpm for 20 min, washed with ethanol and H₂O for two times and finally redispersed the nanoparticles in deionized water and saved for use.

Stability of Eu@Tm@Tb UCNPs in SBF, ALF, Serum, CCM, and S9: SBF, ALF and CCF were prepared following the reported literature.^{4,5}

100 µg/mL of Eu@Tm@Tb UCNPs was added into to a glass bottle containing 1 mL SBF. The resulting mixture was incubated for 28, 14, 7, 3, 1 d, and 0 d at 37 °C in a time-reversed order. Fluorescence spectra of all samples were measured on the last day. The resulting mixed solution was then centrifuged at 12,000 rpm for 30 min to collect the supernatant and precipitate, respectively.

The supernatant was digested with 5 mL of nitric acid (HNO₃, 70%) and 1 mL of hydrogen peroxide (H₂O₂, 30%) by a microwave digestion system (MARS, USA). After cooling down to 80 °C, the resulting mixture was heated at 200 °C to remove the remaining nitric acid. When water was evaporated, the resulting solution was diluted to 10 mL by 2% HNO₃ solution, then the metal contents of Gd, Eu, Yb and Tb in solution were measured by ICP-MS (ELAN DRC-e, PerkinElmer Co., Ltd., USA). In addition, the precipitate was redispersed in deionized water and characterized by TEM.

2.9. Characterization

Luminescence spectra were measured at room temperature with an FS5 in conjunction with 980 nm (cnilaser, MDLIII-980-2.0W, China) diode lasers. TEM images were taken using HT7700 field emission transmission electron microscope operated at an acceleration voltage of 120 kV. Energy dispersive X-ray (EDX) spectrum was recorded by an HT7700 field emission transmission electron microscope equipped with Oxford Instruments. High-resolution TEM (HRTEM) and elemental mapping were performed on an FEI Talos F200S transmission electron microscope operating at an acceleration voltage of 200 kV. Powder X-ray diffraction (XRD) analysis was

carried out on a Rigaku D/MAX-2200 attached with a rotating anode and a Cu K α radiation source ($\lambda = 0.15418$ nm). The size of our nanoparticles is the average diameter obtained by size statistical analysis of TEM images via a Nano Measurer software.

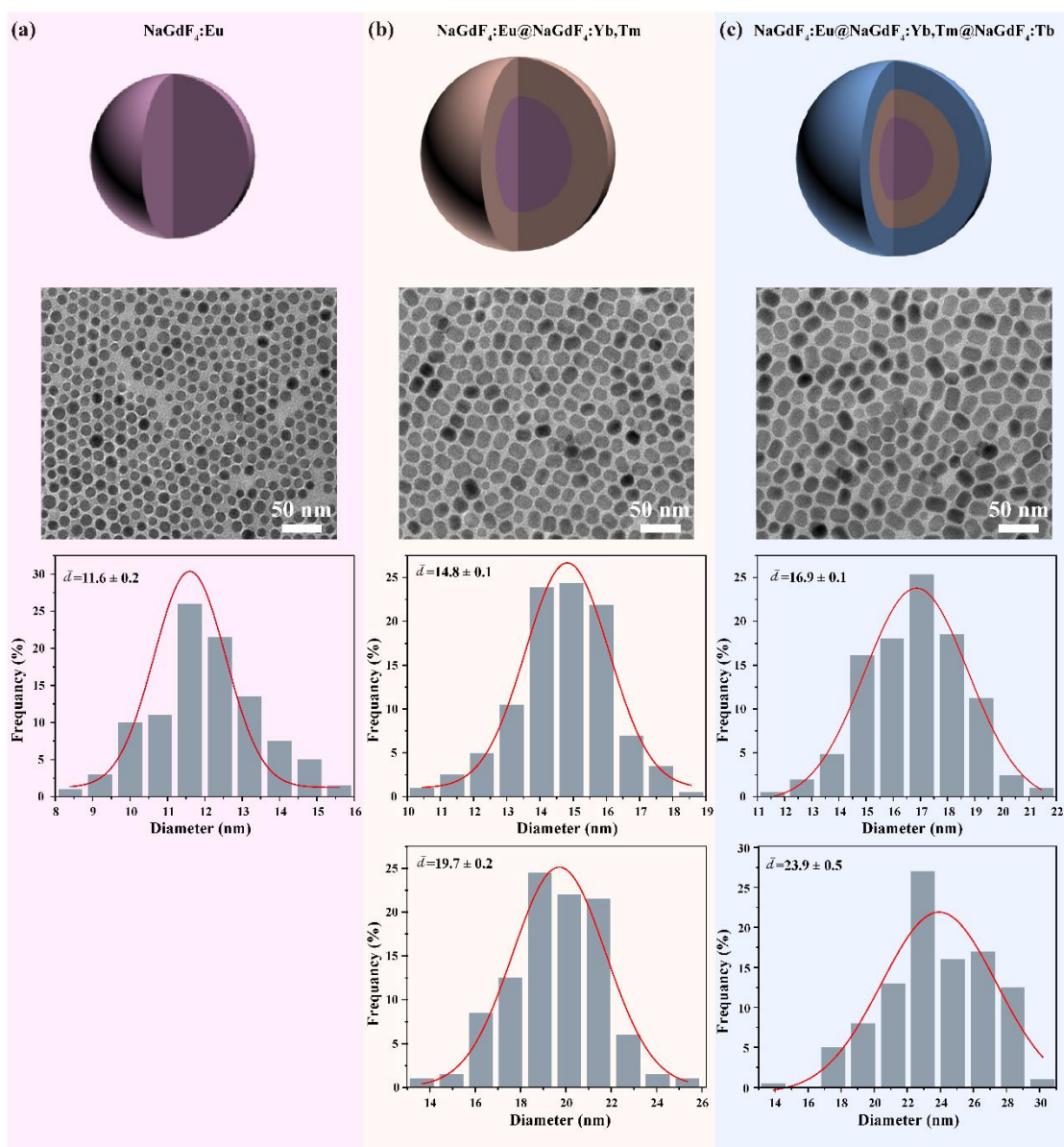


Fig. S1. Schematic illustration, TEM images and size distributions of core (a), core-shell (b), core-shell-shell (c), of the as-synthesized Eu@Tm@Tb nanoparticles. The size distribution is fitted by a Gaussian curve (full red line).

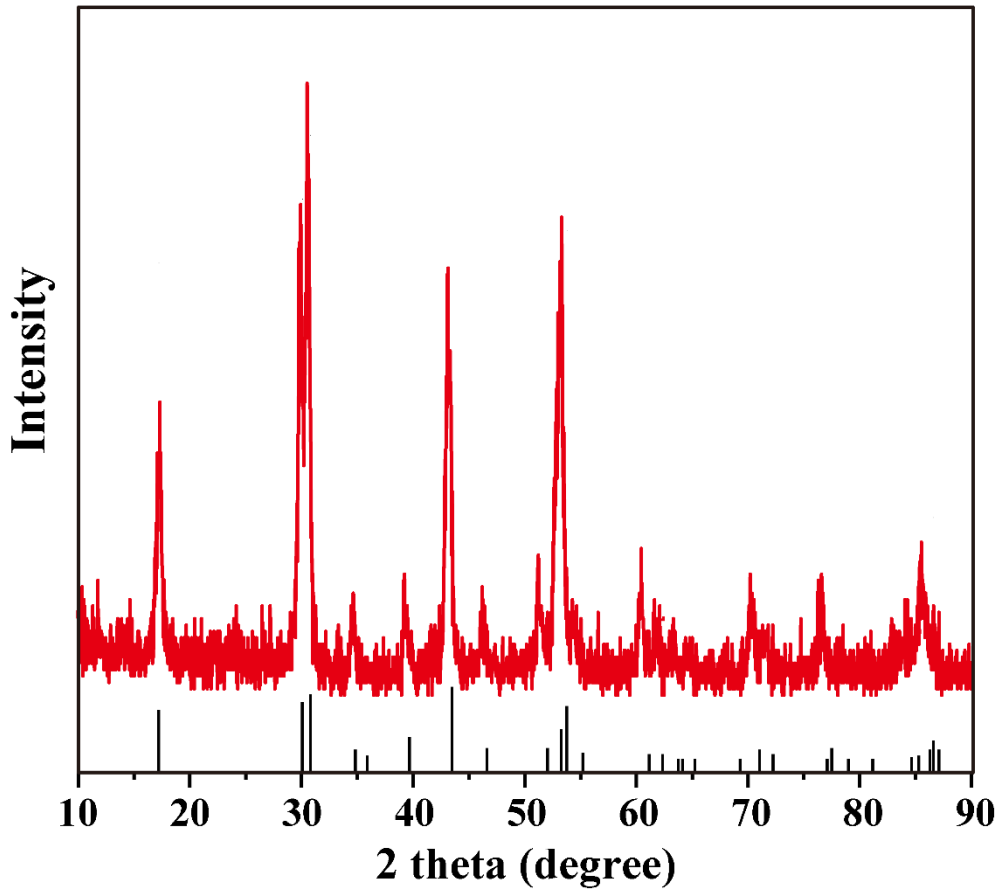


Fig. S2. XRD pattern of Eu@Tm@Tb nanoparticles.

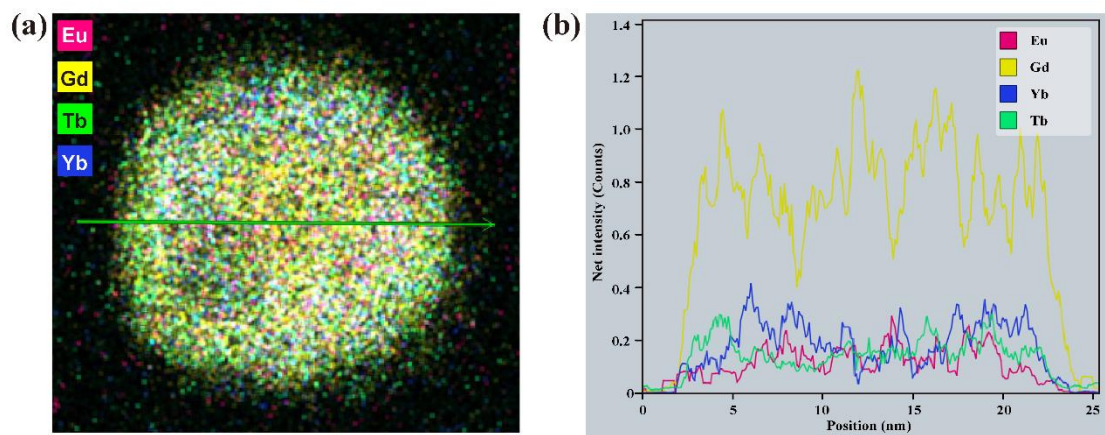


Fig. S3. (a) Elemental mapping of a single Eu@Tm@Tb nanoparticle. (b) Energy-dispersive X-ray mapping analysis of the heterogeneous core-shell-shell nanoparticle.

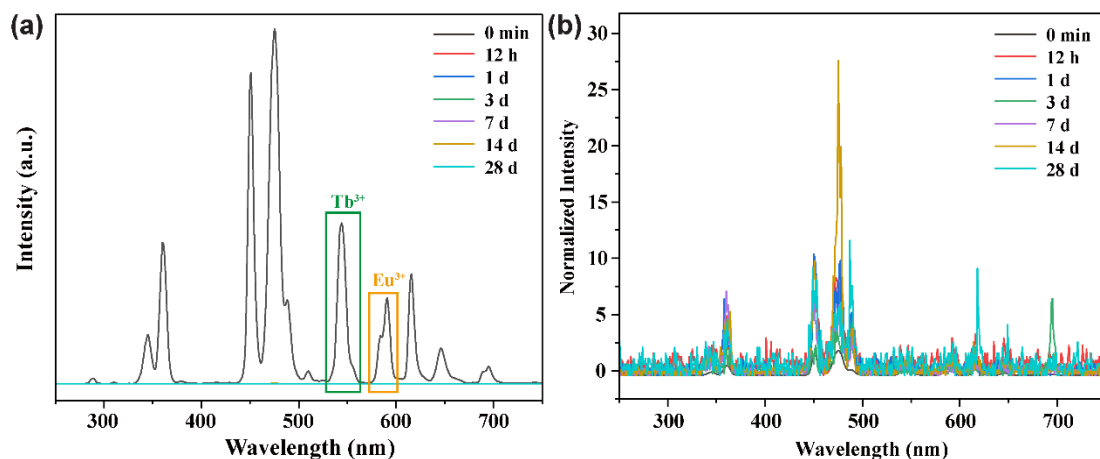


Fig. S4. The emission spectra of UCNPs incubated in PBS solution for different times (a) and the emission spectra normalized with Tb^{3+} characteristic peak under 980 nm excitation (b).

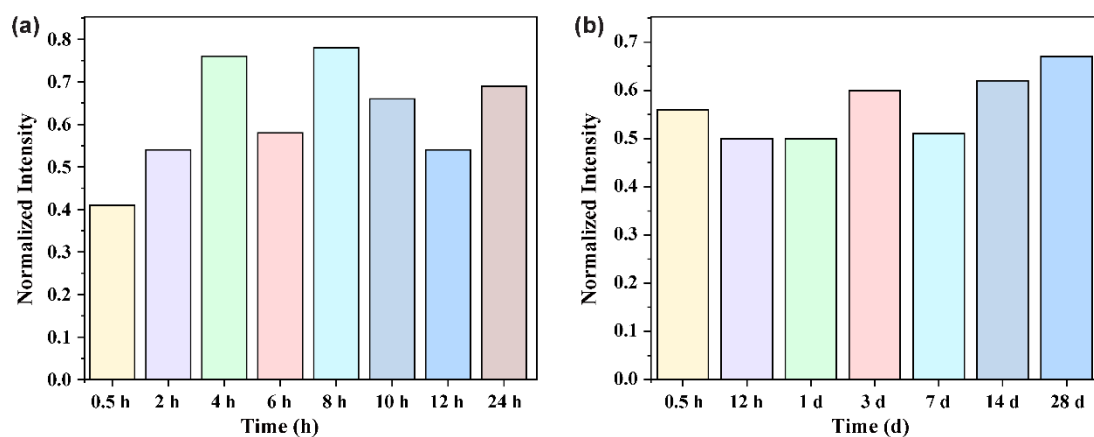


Fig. S5. Luminescence Eu^{590}/Tb^{545} ratio of the as-prepared nanoparticles incubated in in PBS (a) and pH=4.5 aqueous solution (b) for different times.

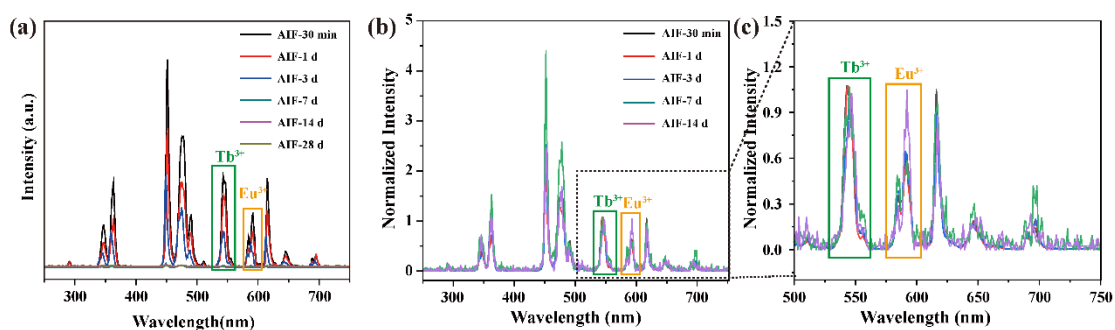


Fig. S6. The emission spectrum of UCNPs incubated in AIF media for different time (a) and the emission spectrum normalized with Tb^{3+} characteristic peak under 980 nm excitation (b-c).

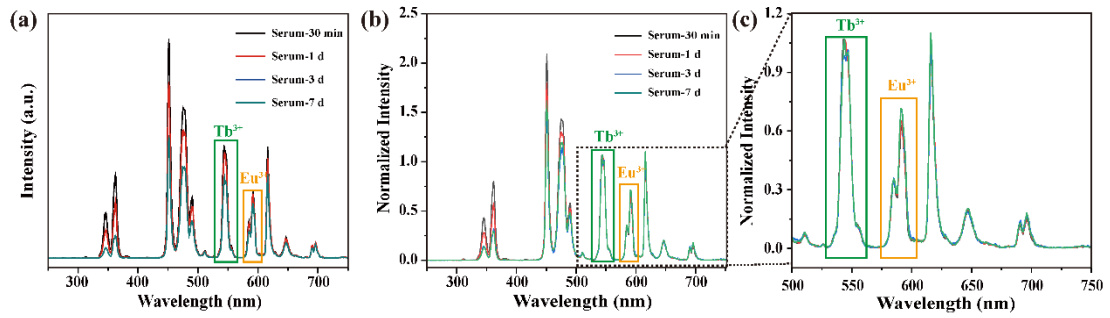


Fig. S7. The emission spectrum of UCNPs incubated in serum media for different times (a) and the emission spectrum normalized with Tb^{3+} characteristic peak under 980 nm excitation (b-c).

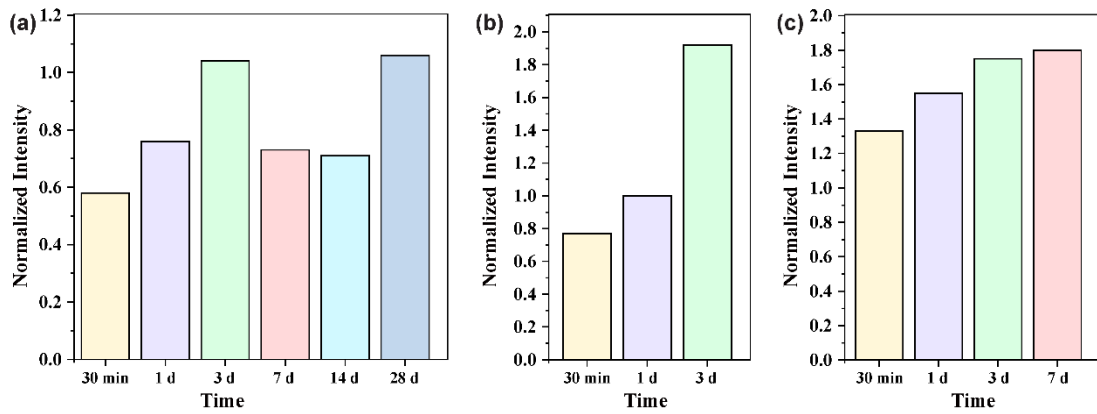


Fig. S8. Luminescence Eu_{590}/Tb_{545} ratio of the as-prepared nanoparticles incubated in SBF (a), S9 (b) and CCM (c) for different times.

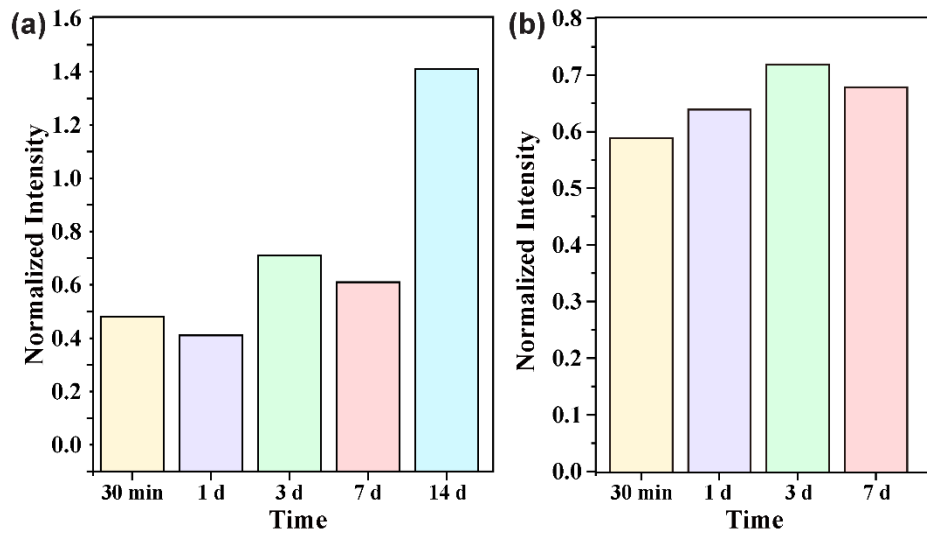


Fig. S9. Luminescence Eu_{590}/Tb_{545} ratio of the as-prepared nanoparticles incubated in AIF (a) and serum (b) for different times.

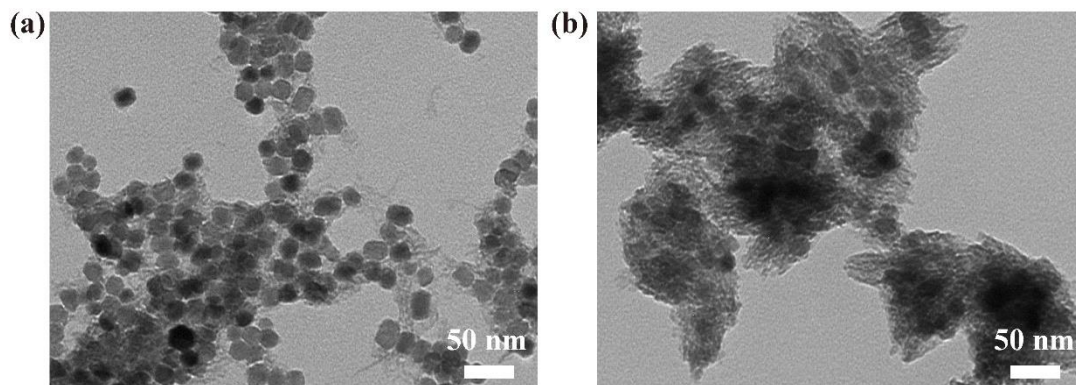


Fig. S10. TEM images of UCNPs incubated in serum media for 3 d (a) and 7 d (b).

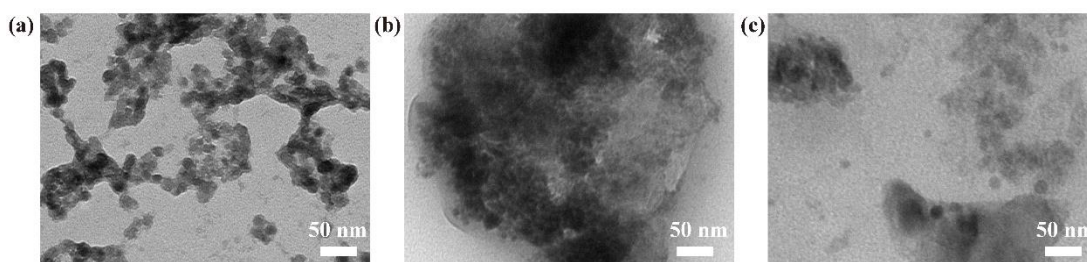


Fig. S11. TEM images of UCNPs incubated in AIF media at 7 d (a), 14d (b) and 28 d (c).

References

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