Supporting Information for

Upconversion Nanoparticle-Anthraquinone Sensors for Measuring pH

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Synthesis of β-NaYF4: Yb, Er Particles

The following describes the synthesis of 1 mmol of β -NaYF₄: 20% Yb, 2% Er particles, but the protocol can be scaled up to 20 mmol batches. The corresponding molar ratios of the lanthanide chloride hexahydrates were dissolved in 5 mL of methanol and transferred into a three-neck round bottom flask. Oleic acid (8 mL) and 1-octadecene (15 mL) were added under nitrogen flow. The mixture was heated to 160 °C then a vacuum was applied for 30 min, after which a clear solution formed. After the solution was cooled to room temperature, ammonium fluoride (4.0 mmol) and sodium hydroxide (2.5 mmol) were dissolved in methanol and added to the flask. The solution was maintained at 120 °C for 30 min then heated to 325 °C. A 980 nm cw laser module (200 mW, 130 W cm⁻²) was used to monitor the reaction progress. The reaction was maintained for an additional 8 minutes after upconversion luminescence could be observed by the naked eye. The reaction mixture was cooled to room temperature using a heating gun. The particles were precipitated by adding ethanol and collecting by centrifugation at 1,000 g for 5 min. The precipitate was washed twice with chloroform/ethanol (1:10 v/v) and three times with cyclohexane/ethanol (1:10 v/v) by repeated redispersion-precipitation-centrifugation cycles. The particles were dispersed in cyclohexane (10 mL), centrifuged at 1,000 g for 3 min to remove remaining aggregates, and the clear supernatant was collected.

Synthesis of α-NaYF₄

The synthesis procedure for α -NaYF₄ was identical to that for β -NaYF₄: Yb, Er (described above), except for the last heating step. Here, the mixture was heated to 240 °C for 30 min.

Synthesis of Core-Shell NaYF₄: Yb, Er@NaYF₄ Particles

The core material (β -NaYF₄: Yb, Er particles) and shell precursor (α -NaYF₄ particles) dispersed in cyclohexane were transferred into separate three-neck round bottom flasks under nitrogen flow. Oleic acid (5 mL) and 1-octadecene (5 mL) were added. Both flasks were heated to 100 °C, and a vacuum was applied for 30 min to remove the cyclohexane. Next, the hexagonal particles were heated to 325 °C and the cubic particles were kept at 100 °C under nitrogen flow. Small volumes (<3 mL) of the shell precursor were injected into the other flask every 10 min, making sure to prevent the temperature dropping below 300 °C. The suspension was cooled to room temperature using a heating gun 10 min after the last injection. The same redispersion-precipitation-centrifugation steps as described above were performed to precipitate and purify the core-shell nanoparticles.



Figure S1. Emission spectra of the same batch of thick-shell, PMA-coated UCNPs in water measured with two different spectrometers.



Figure S2. Emission spectra of two batches of thick-shell, PMA-coated UCNPs used in the pH titration experiments with the two UCNP-anthraquinone mixtures. There are slight differences in peak ratios and emission shoulders.



Figure S3. Size distribution of hexagonal-phase, oleate-capped NaYF₄: 20% Yb, 2% Er UCNPs calculated from transmission electron micrographs.



Figure S4. XRD pattern of core-shell UCNPs.



Figure S5. Absorption spectra of (a) CaR and (b) ARS in solution buffered with 0.2 M phosphate/citrate at different pHs.



Figure S6. Fluorescence emission spectra of (a) CaR and (b) ARS in solution buffered with 0.2 M phosphate/citrate at different pHs. At acidic pHs, the change in fluorescence of ARS could not be detected by the spectrofluorimeter.



Figure S7. Zeta potential measurement of UCNP coated with PMA (left) and PEI (right) after a two-step ligand exchange process.



Figure S8. DLS measurements of UCNPs coated with OA in cyclohexane and PMA in water.



Figure S9. pH response curve in 0.2 M phosphate/citrate buffer of (a) CaR solution at an absorbance of 533 nm and (b) ARS solution at an absorbance of 518 nm.



Figure S10. Cut and normalized decay data of the 540 nm emission of UCNP-CaR mixture (A-C) and UCNP-CaR conjugate (D-F) in phosphate/citrate buffer solution of different pH values. Each average lifetime was calculated from monoexponential fits of three decay curves. UCNP-ARS mixture and UCNP-ARS conjugate had similar trends in their decay data.



Figure S11. Absorption spectra of CaR, UCNP, UCNP-CaR mixture, and the sum value of the absorbance of UCNP and CaR (left). Absorption spectra of ARS, UCNP, UCNP-ARS mixture, and the sum value of the absorbance of UCNP and ARS (right).



Figure S12. Structures of the CaR (left) and ARS (right) anthraquinone derivatives used in this work. Drawings were created with ChemDoodle.



Figure S13. DLS measurements of UCNPs coated with PEI at different pHs.

				CaR on UCNPs (mg)	Fraction on UCNPs
	OD ₅₂₀	c _s (mM)	Start	0.357	
S1	0.183	0.028	Wash 1	0.328	0.92
S2	0.079	0.0073	Wash 2	0.320	0.90
S3	0.064	0.0044	Wash 3	0.315	0.88

Table S1. Fraction of excess starting CaR dye on UCNPs after wash steps

Table S2. Fraction of excess starting ARS dye on UCNPs after wash steps

				ARS on UCNPs (mg)	Fraction on UCNPs
	OD ₄₂₁	c _s (mM)	Start	0.342	
S1	0.063	0.019	Wash 1	0.322	0.94
S2	0.047	0.008	Wash 2	0.314	0.92
S3	0.048	0.008	Wash 3	0.305	0.89

S = supernatant

 c_s = concentration of supernatant

Calculating the Dye Loading on UCNPs

$$n_{dye} = c_{dye} V_{sample}$$

Where n = number of moles, c = concentration, and V = volume

$$N_{dye} = n_{dye} N_A$$

Where N = number of molecules and $N_A =$ Avogadro's number

$$V_{UCNP} = \frac{4}{3}\pi r_{UCNP}^3$$

Where r = radius

$$m_{UCNP} = V_{UCNP} \rho_{UCNP}$$

Where m = the mass of one particle and ρ = density

 $M_{UCNP} = V_{sample} c_{UCNP}$ Where M = total mass in the sample. C_{UCNP} is obtained from ICP measurements.

$$N_{UCNP} = M_{UCNP}/m_{UCNP}$$