High-efficiency and water-quenching-resistant Tb$^{3+}$-based nanoparticles for single-particle imaging

Abstract: The structure of the host lattice has a substantial influence on the optical properties of lanthanide-doped luminescent materials. Hexagonal-phase ($\beta$-phase) NaREF$_4$ (RE = rare earth) is the most commonly used crystal structure for lanthanide-doped upconversion nanoparticles (UCNPs) owing to its high upconversion (UC) efficiency. In this work, we report, for the first time, that more efficient cooperative sensitization upconversion (CSU) can be achieved in cubic-phase ($\alpha$-phase) NaREF$_4$ UCNPs instead of their $\beta$-phase counterparts. With the passivation of an inert shell, the UC emission intensity of $\alpha$-NaYbF$_4$:Tb$_{40}\%$@CaF$_2$ is 10.5 times higher than that of $\beta$-NaYbF$_4$:Tb$_{40}\%$@NaYF$_4$. We propose that the high-symmetry crystal structure of the $\alpha$ phase facilitates the formations of [Yb–Yb] dimers and [Yb–Yb–Tb] clusters, which are particularly beneficial for CSU. Moreover, we prove that such Tb$^{3+}$-based UCNPs are almost impervious to water quenching because of the large energy gap ($\sim$15,000 cm$^{-1}$) that existed in Tb$^{3+}$ between its lowest emitting level ($^5D_4$) and next low-lying level ($^7F_0$). Finally, their potential application for single-nanoparticle imaging has also been demonstrated. As expected, the $\alpha$-core-shell UCNPs measured at the single-nanoparticle level are estimated to be 9-fold brighter than their $\beta$-core-shell counterparts. Importantly, the $\alpha$-NaYbF$_4$:Tb$_{40}\%$@CaF$_2$ UCNPs offer exciting opportunities for realizing single-nanoparticle imaging at ultralow irradiance (30 W/cm$^2$).

Keywords: cooperative sensitization upconversion; cubic phase; high-symmetry crystal structure; single-nanoparticle imaging; water-quenching-resistant.

1 Introduction

The host material is of great importance for lanthanide-doped upconversion nanoparticles (UCNPs), which play a crucial role in determining their upconversion (UC) efficiency, emission colors, and potential applications [1–3]. A desirable host material has the ability to host the sensitizer and activator ions within a proper distance, thus enabling the generation of intense UC emission. NaREF$_4$ (RE = rare earth) is one of the most popular host materials for lanthanide-doped UCNPs owing to its low phonon energy (<400 cm$^{-1}$) [4]. There are two types of crystal structure for NaREF$_4$, cubic ($\alpha$) and hexagonal ($\beta$) phases. The structure of $\alpha$-phase NaREF$_4$ is isomorphic with CaF$_2$, which contains one type of high-symmetry cation site randomly occupied...
by Na\(^+\) and Re\(^{3+}\) ions [5, 6]. In contrast, the structure of β-phase NaREF\(_4\) consists of two types of relatively low-symmetry cation sites selectively occupied by Na\(^+\) and Re\(^{3+}\) ions [7–9]. Of particular interest to researchers is the β-phase structure, normally with Yb\(^{3+}\) as sensitizer and Er\(^{3+}\), Tm\(^{3+}\), or Ho\(^{3+}\) as the activator, because of their excellent UC performance. In general, the UC efficiency of their β-phase structure exhibits about one order of magnitude higher than that of their α-phase counterpart [7, 10]. However, whether β-phase NaREF\(_4\) is beneficial for all the lanthanide-doped UCNPs is still unclear.

Tb\(^{3+}\)-based luminescent materials are extremely useful for the application of time-resolved spectroscopic technique because Tb\(^{3+}\) has much longer luminescent lifetime (up to milliseconds) compared with those of Er\(^{3+}\)/Tm\(^{3+}\)/Ho\(^{3+}\) [11–13]. It is known that Yb/Tb co-doped nanoparticles are able to generate UC emission relying to a process called cooperative sensitization upconversion (CSU), in which two adjacent Yb\(^{3+}\) ions simultaneously transfer the excitation energy to the nearest Tb\(^{3+}\) ion [14–17]. Compared with energy transfer upconversion (ETU, \(\eta = 10^{-3}\))，namely the dominant UC process in Yb/Er, Yb/Tm, and Yb/Ho co-doped systems, the process of CSU (\(\eta = 10^{-6}\)) shows much lower efficiency [3]. Hence, very few studies have been devoted to Yb/Tb co-doped UCNPs. Owing to the long luminescent lifetime of Tb\(^{3+}\)-based UC NPs, they may provide valuable applications in time-gated luminescence imaging, biosensing, and optical encoding [12, 18–20]. Therefore, it is of great significance to improve the efficiency of CSU in the Yb/Tb co-doped system.

Herein, we demonstrate that high-efficiency CSU can be achieved in α-phase instead of β-phase NaREF\(_4\). Both α- and β-NaYbF\(_4\):Tb\(^{40}\%\) core-only UCNPs, and their derivative core–shell UCNPs with highly comparable particle sizes, were synthesized and systematically investigated. The resulting α-phase UCNPs, no matter core-only or core–shell structure, is much brighter than their β-phase counterparts. From the view of crystal structure, α-phase NaREF\(_4\) is featured with higher-symmetry cation sites, thus benefiting the formations of [Yb–Yb] dimers and [Yb–Yb–Yb] clusters in the lattice. The existence of such dimers and clusters is able to dramatically boost the efficiency of CSU. In addition, we also found that Tb\(^{3+}\) is free from luminescence quenching caused by water because of the large energy gap between its 5D\(_4\) and 7F\(_0\) states, making Tb\(^{3+}\)-based UCNPs promising for biological applications. As lanthanide-doped UCNPs have been recently considered as ideal tools for single-nanoparticle imaging, we further investigated the as-prepared α- and β-phase core–shell UCNPs for such application. As a result, the α-core-shell UCNPs are bright enough for single UCNP observation, and their brightness is approximately an order of magnitude higher than that of β-core-shell counterparts.

### 2 Results and discussion

To make α- and β-phase UCNPs exactly comparable, the first is to ensure that both of them are comprised of the same RE ions and doping concentrations. The desirable doping concentration for Yb/Tb co-doped UCNPs is considered to be 60 mol% Yb\(^{3+}\) and 40 mol% Tb\(^{3+}\) [14, 21]. So firstly, both α- and β-phase NaYbF\(_4\):Tb\(^{40}\%\) were designed as the core-only UCNPs for comparison. As the UC efficiency is strongly particle-size dependent [22, 23], another important prerequisite for a rational comparison is that their particle sizes have to be approximately equal to each other. We successfully established a homostructured epitaxial growth strategy that allows us to obtain size-tunable α-NaYbF\(_4\):Tb\(^{40}\%\) UCNPs, with an initial size of ∼8.8 nm (Figure S1). Given that the as-prepared β-NaYbF\(_4\):Tb\(^{40}\%\) UCNPs have an average size of 13.2 ± 0.6 nm (Figures 1b and S2b), the α-NaYbF\(_4\):Tb\(^{40}\%\) UCNPs with an average size of 13.0 ± 1.8 nm (Figures 1a and S2a) were synthesized through the aforementioned homostructured epitaxial growth strategy. The XRD patterns of the as-synthesized α- and β-phase core-only UCNPs are presented in Figure 1e, which are in good agreement with the standard patterns of JCPDS 27-1426 and 27-1427, respectively, confirming they are pure α- and β-phase structures. Subsequently, rationally designed α- and β-phase core–shell UCNPs of 18.2 ± 2.2 nm α-NaYbF\(_4\):Tb\(^{40}\%\)@CaF\(_2\) (Figures 1c and S2c) and 20.4 ± 0.9 nm β-NaYbF\(_4\):Tb\(^{40}\%\)@NaYF\(_4\) (Figures 1d and S2d) were synthesized separately by using the above α- and β-phase core-only UCNPs. As α-NaREF\(_4\) and CaF\(_2\) are isostructural materials, CaF\(_2\) is easy to grow on α-NaREF\(_4\) seeds [5]. The formation of an inert CaF\(_2\) shell can be clearly seen from Figure 1c, and the shell thickness on the α-core is calculated to be ∼2.5 nm. On the other hand, the thickness of the inert β-NaYF\(_4\) shell on the β-core is determined to be ∼3.5 nm. The corresponding XRD patterns of the current two core–shell UCNPs are also shown in Figure 1e, indicating the successful preparation of our rationally designed core–shell structured UCNPs.

The UC emission spectra of the resulting α- and β-phase core–shell, and their core–shell samples, were then studied and the results are presented in Figure 2a. Based on the mechanism of CSU (Figure 2b), the UC emissions located at 486, 542, 587, and 620 nm are attributed to the transitions of 5D\(_4\) → 7F\(_6\), 5D\(_4\) → 7F\(_5\), 5D\(_4\) → 7F\(_4\), and 5D\(_4\) → 7F\(_3\) in Tb\(^{3+}\) ions, respectively. In many cases, the UC efficiency
of α-phase NaREF₄ is considered to be at least an order of magnitude lower than that of its β-phase counterpart, and one of the good examples is the Yb/Er co-doped UCNPs (Figure S3). But our study suggests that this is not applicable to the Yb/Tb co-doped system. First, we found that the α-core UCNPs are 3.1 times brighter than the β-core UCNPs (Figure 2a and c). Moreover, the decay time of α-NaYbF₄:Tb40% (1.9 ms) at 542 nm emission is also longer than that of β-NaYbF₄:Tb40% (1.7 ms), implicating a much faster decay rate in β phase (Figure 2d) [24, 25]. So the α-phase structure is more conducive to the accumulation of population at the ⁵D₄ emitting level. As the sizes of α- and β-phase core-only UCNPs are highly comparable (~13 nm), it can be concluded that the α phase enables a higher CSU efficiency than the β phase. Second, we also compared the brightness of their corresponding core–shell UCNPs. In the case of the α phase, with the passivation of an inert CaF₂ shell, an enhancement of 36.7 times in UC emission intensity was observed in comparison with the α-core alone. In addition, the decay time at 542 nm emission increases from 1.9 to 2.7 ms (Figure 2d). In the case of the β phase, after coating an inert β-NaYF₄ shell, 10.9-fold enhancement was realized compared with the β-core alone, accompanied by a prolongation of decay time from 1.7 to 2.5 ms (Figure 2d). More importantly, the UC emission intensity of α-NaYbF₄:Tb40%@CaF₂ is 10.5 times stronger than that of β-NaYbF₄:Tb40%@NaYF₄. The aforementioned results confirm that much more efficient CSU can be achieved in the α phase rather than in the β phase, especially in core–shell structure.

The reason why the α phase enabling more efficient CSU, is highly associated with its crystal lattice arrangements. The structure of α-NaREF₄ is isomorphic with the well-known face-centered CaF₂ fluorite type (space group Fm̅3m), in which the sites of Ca²⁺ ions are randomly occupied by 1/2 Na⁺ and 1/2 RE³⁺, and the O₆h symmetry of the cationic site is retained (Figure 2e) [8]. Historically, the crystal structure of β-NaREF₄ was debatable, and three models, namely P6₃, P6̅2m, and P6₃/m space groups were once proposed to rationalize the atomic arrangement in β-NaREF₄ [26, 27]. P6₃/m space group is currently a more widely adopted structure, in which there are two relatively low-symmetry cation sites, and the RE³⁺ ions possess a crystallographic site symmetry of C₃v (Figure 2f) [8]. Hence, the α-phase structure offers a much higher-symmetry environment for RE³⁺ ions. According to literature, clusters of RE³⁺ ions, such as [RE–RE] dimers are more likely to form in α-NaREF₄ owing to its higher-symmetry crystal structure [28–30]. But in most instances, the existence of [RE–RE] dimers in the host lattice is considered detrimental to UC, because it intensifies the cross-relaxation (CR) effect between activators and enormously weakens the UC brightness [31, 32]. Take the Yb/Er co-doped UCNPs as an example, as the high-symmetry α-phase NaREF₄ favors the formation of [Er–Er] dimers in the crystal lattice, the CR between two neighboring Er³⁺ ions are more likely to occur. The evidence for the enhanced CR effect between two Er³⁺ ions in α phase can be found in Figure S3e, where the red emission becomes dominant because of the CR mechanism depicted in Figure S4 [33]. On the contrary, the low-symmetry β-NaREF₄ can effectively avoid the formation of [Er–Er] dimers, thus significantly minimizing the adverse CR between activators. As can be seen from Figure S3e, the intense green emission in β-NaYF₄:Tb20%, Er2% indicates the reduced CR effect between Er³⁺ ions. A similar situation also prevails in Yb/Tm and Yb/Ho co-doped UCNPs. So β-NaREF₄ usually affords higher UC efficiency and is more preferable for most of the lanthanide-doped UCNPs. This, however, is not the case in Yb/Tb co-doped system. Different from the Yb/Er co-doped system, which is dominated by the process of ETU, the process of CSU in the Yb/Tb co-doped system requires two adjacent Yb³⁺ ions to simultaneously transfer their excitation energy to the nearest Tb³⁺. As aforementioned, the high-symmetry
The structure of $\alpha$-NaYbF$_4$:Tb not only facilitates the formation of [Yb–Yb] dimers but also benefits the formation of [Yb–Yb–Tb] clusters in the crystal lattice. Distinct from the $\beta$ phase (Figure 2h), the widely existed [Yb–Yb] dimers and [Yb–Yb–Tb] clusters in the $\alpha$ phase significantly boost the efficiency of CSU (Figure 2g). This proposed theory also well explains why when passivated with the shell of similar thicknesses, 36.7-fold enhancement was gained in $\alpha$-core-shell UCNPs, whereas merely 10.9-fold improvement was observed in $\beta$-core-shell UCNPs. Understandably, without shell protection, the excitation energy immediately migrates to the surface defects through [Yb–Yb] dimers [34]. Therefore, the existence of [Yb–Yb] dimers makes $\alpha$-core more vulnerable to surface quenching. But with the presence of an inert shell, the energy migration via [Yb–Yb] dimers to the surface defects is severely suppressed. In this case, [Yb–Yb] dimers ensure more excitation energy contributing to CSU instead of being dissipated by surface defects. As a result, the enhancement of UC emission in $\alpha$-core-shell UCNPs is much higher than that in $\beta$-core-shell UCNPs. It is also worth mentioning that the CR between Tb$^{3+}$ ions hardly quenches the emission originated from the $^5D_{4}$ emitting level [21]. Albeit the formation of [Yb–Tb] dimers in the $\alpha$ phase, it will not influence the UC emission of Tb$^{3+}$.

Water-induced quenching of UC emission widely exists in lanthanide-doped UCNPs. It is mainly caused by the overtone vibrational transitions of H$_2$O molecules, leading to serious depopulation of those emitting states in lanthanide ions such as Yb$^{3+}$, Er$^{3+}$, Tm$^{3+}$, and Ho$^{3+}$ [35, 36]. First, it is found that an inert shell of 4 nm thickness is able to suppress the quenching of Yb$^{3+}$ caused by water to some extent (Figure S6). Unfortunately, even with the protection of the shell (Figure S5b), the UC emission of Er$^{3+}$ is hugely quenched by water (Figure 3a). The energies required for the $^2H_{11/2}$, $^4S_{3/2} \rightarrow ^4I_{9/2}$ and $^4F_{9/2} \rightarrow ^4I_{9/2}$ transitions in Er$^{3+}$ match well with those of O–H stretch ($\sim$3500 cm$^{-1}$) in water molecules, thus resulting in the Er$^{3+}$ visible-emitting states ($^2H_{11/2}$, $^4S_{3/2}$, and $^4F_{9/2}$) highly deactivated (Figure 3b) [37, 38]. Likewise, the $^2F_{5/2} \rightarrow ^2F_{7/2}$ radiative transition in Yb$^{3+}$ is also quenched by water molecules [36]. Such strong nonradiative energy transfer from Yb$^{3+}$ and Er$^{3+}$ ions to H$_2$O molecules renders a marked decline of the lifetimes of both Yb$^{3+}$ and Er$^{3+}$ in water. In Yb/Er co-doped system, the apparent lifetime of 540 nm emission in Er$^{3+}$ is largely affected by the decay rate of the excitation level of Yb$^{3+}$, because the intrinsic lifetime of the $^2F_{5/2} \rightarrow ^2F_{7/2}$ transition in Yb$^{3+}$ is much longer than the intrinsic lifetime of the $^3H_{11/2}$, $^4S_{3/2} \rightarrow ^3I_{15/2}$ transition in Er$^{3+}$ [39]. As a consequence, a drastic shortening in the lifetime of 540 nm emission is found in water compared with that in cyclohexane (from 367.3 to 90.1 ms, inset of Figure 3a). Besides, the energies required for the $^4I_{13/2} \rightarrow ^4I_{15/2}$ and $^4I_{11/2} \rightarrow ^4I_{15/2}$ transitions match with those of the second and third overtone

Figure 2: (a) Corresponding UC emission spectra of the four samples in Figure 1 under 980 nm laser excitation at a power density of 3.6 W/cm$^2$. (b) Schematic energy level diagram showing the proposed CSU process in Yb/Tb co-doped UCNPs. (c) Normalized UC emission intensity of (a). Inset: their corresponding luminescence photographs. (d) Corresponding luminescence decay curves of Tb$^{3+}$ emission at 542 nm. (e–f) Crystal structures of (e) $\alpha$-phase and (f) $\beta$-phase NaREF$_4$. (g–h) Simplified energy level diagrams depicting the CSU process in (g) $\alpha$-phase NaREF$_4$ involving [Yb–Yb] dimer and [Yb–Yb–Tb] cluster and (h) $\beta$-phase NaREF$_4$. 
transitions of water molecules (Figure 3b), which will further intensify the non-radiative energy transfer from Er$^{3+}$ to H$_2$O and decrease its UC emission. Consequently, an obvious weakening of UC emissions in Er$^{3+}$ at 521, 540, and 660 nm are observed in Figure 3a. Significant water-induced quenching was also found in Ho$^{3+}$-based core–shell UCNPs (Figures 3c and S5d), the mechanism of which is analogous to that of Er$^{3+}$ (Figure 3d). However, Yb/Tb co-doped core–shell UCNPs are barely affected by water quenching, as evidenced by the experimental results presented in Figure 3e. This is because of the large energy gap ($\sim$15,000 cm$^{-1}$) between the lowest emitting level (5D$_4$) and its next low-lying level of Tb$^{3+}$, which is much larger than the energies required for overtone vibrational transitions of water molecules (Figure 3f). Hence, the nonradiative energy transfer from Tb$^{3+}$ to H$_2$O molecules could not occur. On the other hand, the CaF$_2$ shell is capable of hindering the diffusion of Yb$^{3+}$ to the shell layer, thus minimizing the nonradiative energy transfer from Yb$^{3+}$ to H$_2$O molecules [40]. The unaltered lifetime of Tb$^{3+}$ when measured in cyclohexane and water (inset of Figure 3e) strongly supports the above assumptions. It should be noted that the optical absorption coefficient of water at 980 nm is 0.485 cm$^{-1}$, suggesting transmission of 61.6% after propagating a path length of 1 cm in water [35, 41, 42]. Therefore, a slight decline in the UC emission intensity of Tb$^{3+}$ is probably resulted from the attenuation of 980 nm excitation light by water (Figure 3e). It is known that up to 70% of the human body is composed of water, so the property of being impervious to water quenching makes Tb$^{3+}$-based UCNPs very attractive for biomedical applications, such as bioimaging and biosensing.

Lanthanide-doped UCNPs are promising single-molecule biological probes, and the application of single-nanoparticle imaging based on the above $\alpha$- and $\beta$-phase core–shell UCNPs was also explored. Briefly, a lab-made microscopy with an scCMOS camera was used to collect the single-nanoparticle microscopic images (Figure 4a). The nanoparticle solutions of $\alpha$-NaYbF$_4$:Tb$_{40\%}$@CaF$_2$ (Figure S7a) and $\beta$-NaYbF$_4$:Tb$_{40\%}$@NaYF$_4$ (Figure S7b) were highly diluted and then were spin-coated uniformly on the coverslips respectively, and imaged under the 980 nm laser excitation with exactly the same test parameters. As shown in Figure 4b, the single $\alpha$-NaYbF$_4$:Tb$_{40\%}$@CaF$_2$ was clearly imaged with high brightness and satisfactory signal-to-noise ratio, even at an ultralow-power density of 30 W/cm$^2$. The corresponding line profile analysis with Gaussian fitting indicates a high resolution of 355 nm (Figure 4d). In comparison, the single $\beta$-NaYbF$_4$:Tb$_{40\%}$@NaYF$_4$ exhibits much weaker brightness and undesirable signal-to-noise ratio (Figure 4c). The corresponding line profile analysis with Gaussian fitting reveals a resolution of 350 nm.

Figure 3: UC emission spectra of (a) Er-based $\beta$-NaYF$_4$:Yb$_{20\%}$, Er$_{2\%}$@NaYF$_4$, (c) Ho-based $\beta$-NaYF$_4$:Yb$_{20\%}$, Ho$_{2\%}$@NaYF$_4$, and (e) Tb-based $\alpha$-NaYbF$_4$:Tb$_{40\%}$@CaF$_2$ core–shell UCNPs under 980 nm laser excitation measured in cyclohexane and water, respectively. Their corresponding TEM images are shown in Figures S5b, S5d, and S5f. Inset: their corresponding luminescence photographs and decay curves. (b, d) Proposed mechanism of water-induced UC luminescence quenching in (b) Er-based and (d) Ho-based core–shell UCNP (Figures 3c and S5d), the mechanism of which is analogous to that of Er$^{3+}$ (Figure 3d).
The single α-core-shell UCNP is approximately 9 times brighter than the single β-core-shell UCNP, which are generally consistent with the results of UC emission spectra measurements (Figure 2a and c). The above results suggest that the α-NaYbF₄: Tb₄₀%@CaF₂ UCNPs turn out to be a better candidate for single-nanoparticle imaging than their β-phase counterparts.

3 Conclusions

In summary, we have successfully synthesized α- and β-phase NaYbF₄: Tb₄₀% and their derivative core–shell UCNPs with comparable size and morphology. Our results have proved that more efficient CSU can be realized in the α phase rather than the traditionally believed β phase. The key factor lies in the high-symmetry cation sites of the α phase, which facilitates the formations of [Yb–Yb] dimers and [Yb–Yb–Tb] clusters. In addition, we have also demonstrated that Tb³⁺-based UCNPs are free from water quenching, and thus are attractive for biological applications. Finally, the single-nanoparticle imaging using the as-prepared α- and β-phase core–shell UCNPs have been investigated and quantified, confirming once again the high-efficiency CSU in α-phase structure.

4 Experimental section

Experimental details are given in the Supplementary Material.

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References


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