Near-infrared light manipulation of non-visual opsin-expressing cells using upconversion nanoparticles

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Although the demand for UV light-induced photochemical reactions *in vivo* has been increasing, UV light has been limited due to its low tissue penetration from outside the organisms and high cytotoxicity. To address this, a technique is desired which uses upconversion nanoparticles (UCNPs), which when irradiated by near-infrared (NIR) light (with high tissue penetration from outside the organisms) convert it to UV light *in vivo*. In the field of optogenetics, visual opsins that respond to visible light have been studied. Recently, UV light-responsive non-visual opsin, OPN5[1] has much attraction, which is expressed in the deep brain and is involved in metabolic functions.[2]

In this study, we synthesized NIR→UV conversion-capable UCNPs and combined them with OPN5-expressing HEK293T cells. As UCNPs, $NaYF_4$:Yb,Tm@$NaLuF_4$ core@shell NPs with enhanced UV light emission were synthesized (**Fig. 1**).[3] For OPN5-expressing HEK293T cells, an OPN5 expression plasmid was synthesized in which the FLAG tag was expressed in the extracellular domain, and transfected into HEK293T cells. Then, UCNPs were bound to the cells using the FLAG tag. In this presentation, we will explain how we demonstrated that it is possible to manipulate OPN5 by binding UCNPs to OPN5-expressing HEK293T cells, and then generating UV light in the vicinity of OPN5 through NIR light irradiation, as evidenced by intracellular Ca^{2+} imaging.

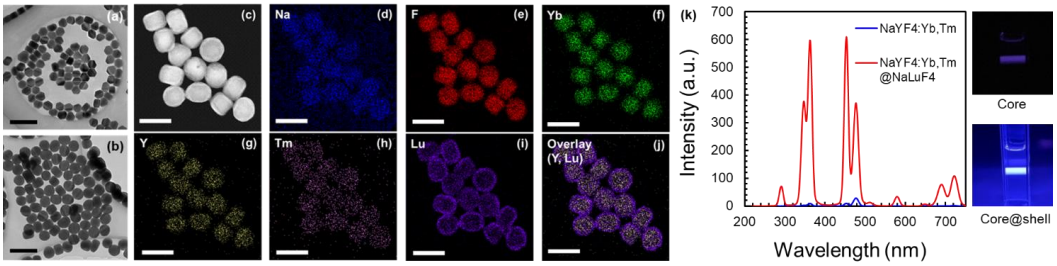

Fig. 1. TEM images of (a) $NaYF_4$:Yb,Tm core and (b) $NaYF_4$:Yb,Tm@$NaLuF_4$ core@shell UCNPs (scale bar = 200 nm). (c) HAADF-STEM image, and (d–j) EDS elemental maps of core@shell UCNPs (scale bar = 100 nm). (k) Emission spectra and photos of emission when irradiating NIR light (980 nm) to as-synthesized core (blue curve) and core@shell UCNPs (red curve) dispersed in hexane.

1) E. E. Tarttelin *et al*., *FEBS Lett*. **2003**, *554*, 410; 2) K. X. Zhang *et al*., *Nature* **2020**, *585*, 420; 3) N. T. Nguyen *et al*., *ACS Nano* **2023**, *17*, 382.