Supporting Information for

Visualization of intercellular cargo transfer using upconverting nanoparticles

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Figure S1. Characterization of synthesized Er^{3+} -doped and Tm^{3+} -doped upconverting nanoparticles (UCNPs). (a-b) Transmission electron microscopy (TEM) images of the synthesized Er^{3+} -doped UCNPs (average diameter of 26 nm) and Tm^{3+} -doped UCNPs (average diameter of 27 nm). (c) Emission spectra of Er^{3+} -doped and Tm^{3+} -doped UCNPs along with the transmission spectra of the optical filters for the emission from Er^{3+} -doped UCNPs at ~650 nm (F1) and that from Tm^{3+} -doped UCNPs at ~800 nm (F2).



Figure S2. The characterizations of PEGylated Er³⁺-doped and PEGylated Tm³⁺-doped UCNPs (Er-UCNPs and Tm-UCNPs). (a-c) TEM image, dynamic light scattering (DLS) and zeta-potential of Er-UCNPs, respectively. The average diameter of Er-UCNPs from DLS was 40.2 nm, and zeta-potential value of Er-UCNPs was -10.9 mV. (d-f) TEM image, DLS and zeta-potential of Tm-UCNPs, respectively. The average diameter of Tm-UCNPs from DLS was 36.7 nm, and zeta-potential value of Tm-UCNPs was -12.2 mV.



Figure S3. Schematic of the real-time dual-color bioimaging setup. Er-UCNPs and Tm-UCNPs in HeLa cells were excited using a 980 nm CW laser (LC: live-cell incubator, S: sample, Obj: objective lens, L: lenses, DM: dichroic mirrors, F: optical filters, and M: mirror).



Er-UCNP Tm-UCNP

Figure S4. Images of Er-UCNPs and Tm-UCNPs in living HeLa cells. (a) Merged images of HeLa cells (bright-field) and photoluminescence (PL) images of Er-UCNPs and Tm-UCNPs after 6 h of incubation. (b) Merged image of HeLa cells and PL images of Er-UCNPs and Tm-UCNPs after 12 h of mixed incubation. Scale bars represent 10 µm.



Er-UCNP Tm-UCNP

Figure S5. Long-term observation results of living HeLa cells and internalized UCNPs with 20 min interval. Boundaries of HeLa cells containing Er-UCNPs and Tm-UCNPs are highlighted by dashed lines with the same color as the cells containing UCNPs. All HeLa cells migrate during the imaging. Scale bars represent 10 μm.



Figure S6. Single-particle tracking (SPT) analysis in a living HeLa cell. (a) Trajectory of Particle Q in Figure 3, which is shown in the video from 10 h 30 min to 10 h 40 min from the start of mixed incubation. Bottom image is the magnified image of the white box in (a). Red triangles and circles show the initial and final positions of each trajectory, respectively. Scale bar represents 10 μ m. Mean-square displacement (MSD) plots and exponent analysis of Trajectories (c) Q₁ and (d) Q₂.