Supporting Information for

## A Dual-Emission Fluorescence-Enhanced Probe for Imaging Copper(II) Ions in Lysosomes

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**Figure S1** The fluorescence spectral changes of control compounds **4** (10  $\mu$ M) upon addition of increasing concentrations of Cu<sup>2+</sup> (0-20 equiv) ( $\lambda_{ex} = 360$  nm) in PBS buffer, pH 4.7, containing 50 % CH<sub>3</sub>CN as a cosolvent.



**Figure S2** The fluorescence spectral changes of control compounds **5** (10  $\mu$ M) upon addition of increasing concentrations of Cu<sup>2+</sup> (0-20 equiv) ( $\lambda_{ex} = 360$  nm) in PBS buffer, pH 4.7, containing 50 % CH<sub>3</sub>CN as a cosolvent.



**Figure S3** The comparison of normalized fluorescence spectral of Lys-Cu, control compound **4** and **5** upon addition of concentrations of Cu<sup>2+</sup> (20 equiv) ( $\lambda_{ex} = 360$  nm) in PBS buffer, pH 4.7, containing 50 % CH<sub>3</sub>CN as a cosolvent.



**Figure S4** The emission intensity changes of **Lys-Cu** upon addition of  $Cu^{2+}$  (20 equiv) at different pH containing 50 % CH<sub>3</sub>CN as a cosolvent ( $\lambda_{ex} = 360$  nm). (Black lines represent the fluorescence spectra before added Cu<sup>2+</sup>; red lines represent the fluorescence spectra after added Cu<sup>2+</sup>).



**Figure S5** (A) The fluorescence intensity of probe Lys-Cu (10  $\mu$ M) at 440 nm in the presence of various analytes (30 equiv) in PBS buffer (pH 4.7, containing 50% CH<sub>3</sub>CN as a cosolvent). (B) The fluorescence intensity of probe Lys-Cu (10  $\mu$ M) at 440 nm in response to Cu<sup>2+</sup> in the presence of various metal species (30 equiv) in PBS buffer (pH 4.7, containing 50% CH<sub>3</sub>CN as a cosolvent).



**Figure S6** Visible color changes of the probe **Lys-Cu** solution (20  $\mu$ M) with different metal ions (20 equiv) in PBS buffer solution (pH 4.7, containing 50% CH<sub>3</sub>CN as a co-solvent): 1. K<sup>+</sup>; 2. Mg<sup>2+</sup>; 3. Na<sup>+</sup>; 4. Ni<sup>2+</sup>; 5. Pd<sup>2+</sup>; 6. Zn<sup>2+</sup>; 7. Ag<sup>+</sup>; 8. Ca<sup>2+</sup>; 9. Cu<sup>+</sup>; 10. Fe<sup>2+</sup>; 11. Co<sup>2+</sup>; 12. Cu<sup>2+</sup>.



**Figure S7** Cytotoxicity assays of **Lys-Cu** at different concentrations (a:  $0 \mu$ M; b:  $1 \mu$ M; c:  $5 \mu$ M; d:  $10 \mu$ M) for NIH3T3 cells



**Figure S8** Fluorescence images of SiHa cells stained with the probe Lys-Cu. a) overlay of blue and green channels ; b) Intensity profile of linear region of interest across the SiHa cell costained with LysoTracker Green and blue channel of Lys-Cu. c) Intensity scatter plot of blue and green channels.



Figure S9 <sup>1</sup>H-NMR (CDCl<sub>3</sub>) spectrum of compound 2.



Figure S10 <sup>13</sup>C-NMR (CDCl<sub>3</sub>) spectrum of compound 2.



Figure S11 HRMS (ESI) spectrum of compound 2.



Figure S12 <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>) spectrum of compound Lys-Cu.



Figure S13 <sup>13</sup>C-NMR (CDCl<sub>3</sub>) spectrum of compound Lys-Cu.



Figure S14 HRMS (ESI) spectrum of compound Lys-Cu.