## **Supporting Information**

## Intrinsic Lysosomal Targeting Fluorescent Carbon Dots with ultrastability for Long-Term Lysosome Imaging

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Fig. S1. XRD spectrum of CDs.



Fig. S2. High-resolution C1s, N1s and O1s spectra of CDs.



**Fig. S3.** (A) Absorbance spectra of CDs and (B) plot of absorption intensity of 340 nm at different concentration of CDs.



Fig. S4. The fluorescence lifetime spectra of CDs.



Fig. S5. Fluorescence spectra of CDs at different pH conditions.



**Fig. S6.** Normalized Fluorescence intensity of CDs upon addition of 1 mM metal ion, anions, reactive oxygen species, amino acids, thiols and other analogues. (K<sup>+</sup>, Ca<sup>2+</sup>, Na<sup>+</sup>, Mg<sup>2+</sup>, Al<sup>3+</sup>, Zn<sup>2+</sup>, Fe<sup>3+</sup>, Cu<sup>2+</sup>, Mn<sup>2+</sup>, Cr<sup>3+</sup>, F<sup>-</sup>, Br<sup>-</sup>, I<sup>-</sup>, CO<sub>3</sub><sup>2-</sup>, NO<sup>2-</sup>, ClO<sup>-</sup>, O<sub>2</sub><sup>-</sup>, OH<sup>-</sup>, H<sub>2</sub>O<sub>2</sub>, Phe, Thr, Arg, Met, Pro, Gly, Lys, Leu, Trp, Ile, Ala, His, Glu, Asp, Val, Ser, Tyr, GSH, Hcy, Cys, UA, AA, NaHSO<sub>3</sub>) in PBS solution (0.01 M, pH 7.4) ( $\lambda$ ex = 405) nm).



Fig. S7. Cytotoxicity of CDs (60  $\mu$ g/mL) on HeLa cells.



**Fig. S8.** (A1) Co-localization images of CDs, (B1) MitoTracker<sup>TM</sup> Deep Red (MTDR) in HeLa cells. (C1) Panels A and B merged. (D1) Fluorescent intensity within the linear region of interest of CDs and MTDR; (A2) Co-localization images of CDs, (B2) ER-Tracker Red (ER-TD) in HeLa cells. (C2) Panels A and B merged. (D2) Fluorescent intensity within the linear region of interest of CDs and ER-TD. Scale bar:  $25 \,\mu m$ .



**Fig. S9.** (A) Co-localization images of CDs, (B) LTDR in HepG2 cells, HL-7702 cells, and 3T3 cells. (C) Panels A and B merged. (D) Fluorescent intensity within the linear region of interest of CDs and LTDR. Scale bar:  $25 \mu m$ .



Fig. S10. Fluorescent images of live cells cultured with CDs at different concentrations. Scale bar:  $25 \ \mu m$ .



Fig. S11. Fluorescent images of CDs stained in HeLa cells at different time. Scale bar:  $25 \ \mu m$ .



**Fig. S12.** (A) Confocal images of live cells cultured with 60  $\mu$ g/mL of CDs (for 48 h), (B) 50 nM LTDR (for 25 min), (C) merged images by panels A and B. (D) Fluorescent intensity within the linear region of interest of CDs and LTDR. Scale bar: 25  $\mu$ m.



**Fig. S13.** Fluorescent images (A) and fluorescence intensity (B) of HeLa cells stained with CDs stimulated by chloroquine (50  $\mu$ M) at different time (0, 5, 10, 15 and 20 min). Scale bar: 25  $\mu$ m.



**Fig. S14.** Fluorescent images of cells treated with CDs (A), LTDR (B) before and after the fixation treatment. (C) Merged images by panels of A and B. (D) Intensity profiles within the linear region of interest of CDs and LTDR. Scale bar:  $25 \mu m$ .



Scheme S1. The proposed synthetic mechanism of the CDs.

Citric acid was selected as the precursor because it contains three carboxylic groups and one hydroxyl group, which can not only serve as precursor but also serve as acid catalyst to catalyze the Friedel-Crafts reactions with *N*, *N*-dimethylaniline to generate the intermediate I. The intermediate can undergo further reaction with both citric acid and *N*, *N*-dimethylaniline. The resulting intermediate was expected to be polymerized and carbonized to form carbon nuclei. Through condensation, polymerization, carbonization and covalent attachment, the CDs were finally formed.