

Supplementary Information

A FRET-based ratiometric fluorescent probe to detect cysteine metabolism in mitochondria

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Cell Culture and Imaging

MCF-7 cells were grown in RPMI-1640 provided with 10% PBS, at 37 °C, under 5% CO₂. Probe **CP-K** (10 μM) was added to MCF-7 cells and incubated at 37 °C for 0.5 hour in 5%CO₂. Next, PBS buffer (pH 7.4) wash the cell for three times. Then, the cells were added to NaHSO₃ solution for another 30 min in 5% CO₂ and washed three times with PBS buffer. Colocalization experiment the MCF-7 cells were processed a mitochondria staining probe, Mito Tracker Green FM (500 nM) and probe **CP-K** (10 μM) for additional 0.5 h and washed with PBS buffer (pH 7.4). To monitor cysteine metabolism, MCF-7 cells were incubated with 10 mM of Cys for 0.5 h and then treated 10 μM probe for 4 h. The cells imaging were used by Leica TCS SP8 confocal microscope.

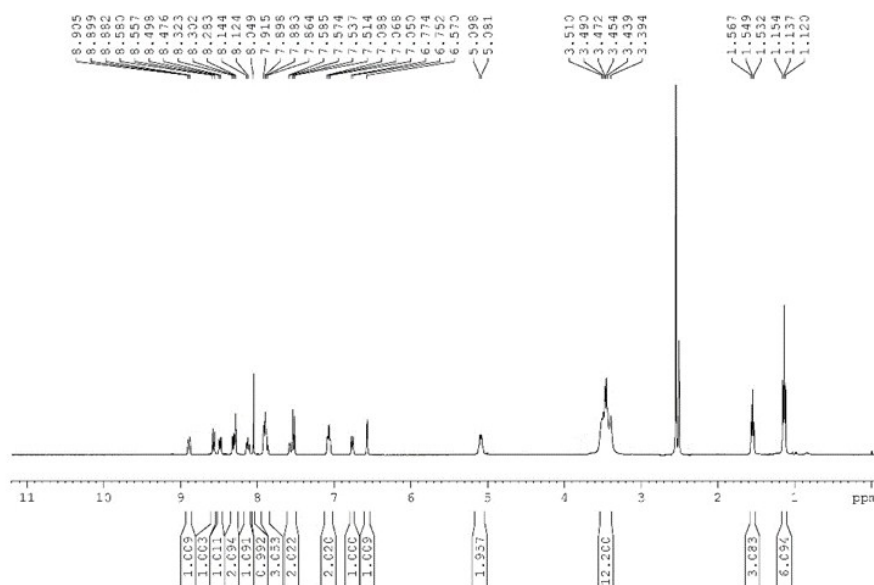


Figure S1. ¹H NMR spectrum of CP-K in DMSO-d₆.

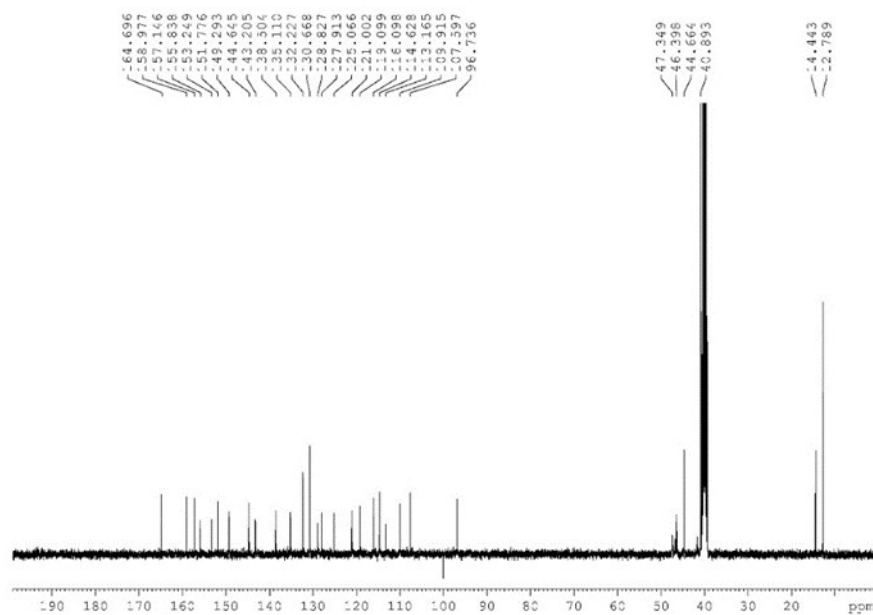


Figure S2. ^{13}C NMR spectrum of CP-K in DMSO-d_6 .

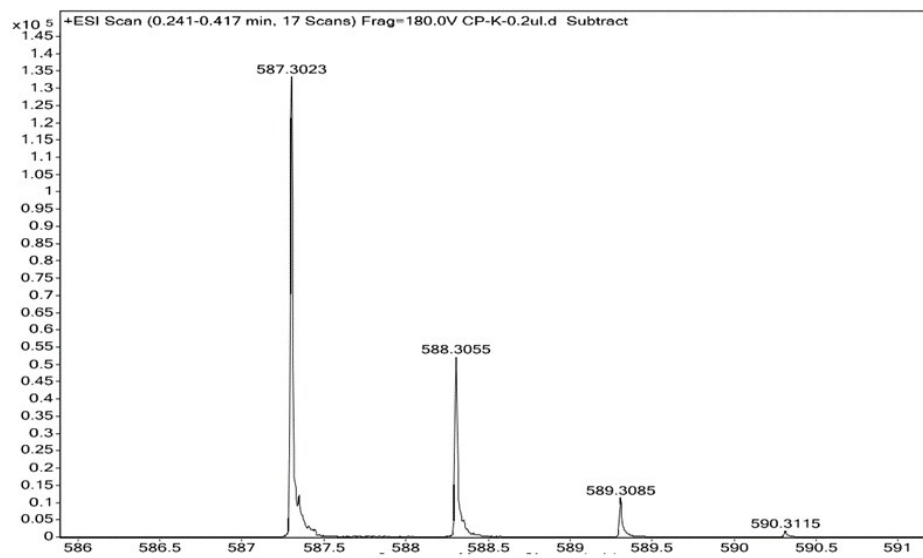


Figure S3. HR-MS spectra of CP-K.

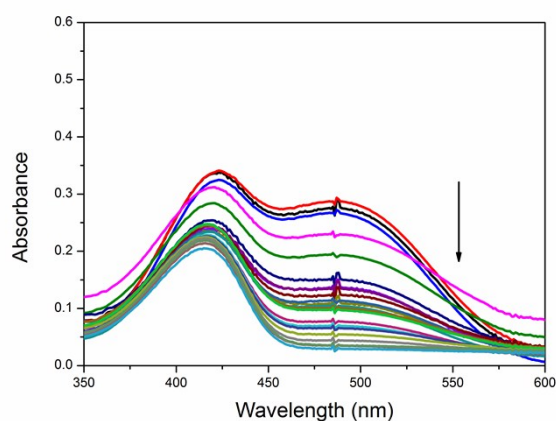


Figure S4. The UV-vis spectra of probe CP-K (10.0 μM) in the presence of NaHSO_3 (0–10 equiv.) in the PBS buffer (10 mM, pH=7.4, containing 50% ethanol).

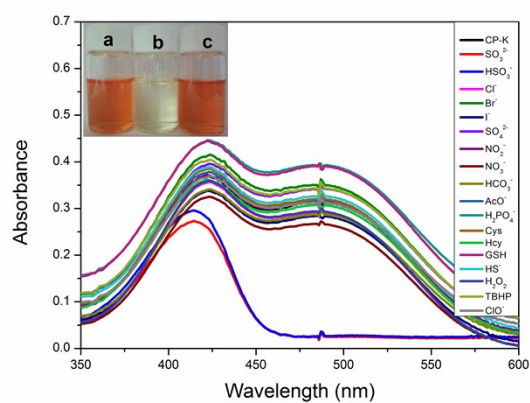


Figure S5. The UV-vis spectra of probe CP-K (10 μM) with NaHSO_3 (10 equiv.) and other various anions (10 equiv.) in the PBS buffer (10 mM, pH=7.4, containing 50% ethanol). (a: probe, b: probe+ NaHSO_3 , c: probe+Competing species)

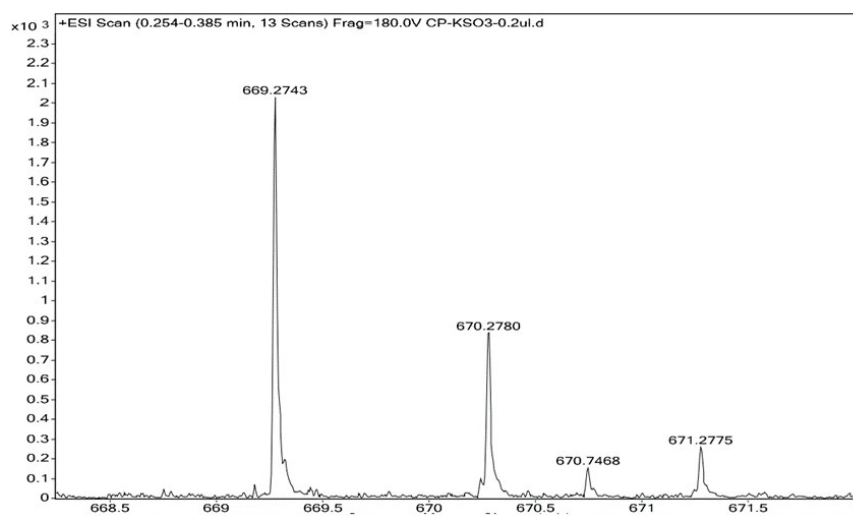


Figure S6. HR-MS spectra of **CP-K** in the presence of HSO-3 (10 equiv.).