Supporting Information

A Highly Specific BODIPY-Based Probe Localized in Mitochondria for HClO Imaging

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1. MS of MitoClO upon addition of NaClO

Fig. S1. MS of MitoClO upon addition of NaClO.

2. Mitochondria colocalization experiments of MCF7 cells

Fig. S2. Confocal images of MitoClO plus a mitochondrially specific dye in MCF-7 cell.(a) Image of MCF-7 cells after treating with LPS (1 μg/mL) for 12 h and PMA (1 μg/mL) for 15 min and subsequent treatment of probe MitoClO (2 μM) for 15 min, λ<sub>ex</sub> = 488 nm; (b) Fluorescence image of MCF-7 cells stained with MitoTracker Deep Red FM, λ<sub>ex</sub> = 635 nm; (c) Bright-field of MCF-7 cell; (d) Merged image of (a), (b) and (c).
3. Living RAW264.7 cell imaging experiment

RAW264.7 cells (mouse macrophages cell line) were purchased from the Committee on Type Culture Collection of Chinese Academy of Sciences. Murine RAW264.7 macrophage cells were maintained following protocols provided by the American Type Culture Collection. Cells were seeded at a density of $1 \times 10^6$ cells mL$^{-1}$ for confocal imaging in RPMI 1640 Medium supplemented with 15% fetal bovine serum (FBS), NaHCO$_3$ (2 g/L), and 1% antibiotics (penicillin /streptomycin, 100 U/ml). Cultures were maintained at 37 °C under a humidified atmosphere containing 5% CO$_2$. The cells were subcultured by scraping and seeding on 20/12 mm Petri dishes according to the instructions from the manufacturer.

![Bright-field Fluorescence Overlay](image)

**Fig. S3.** Confocal fluorescence images of macrophages. RAW264.7 cells incubated with probe MitoClO (2 μM) for 15 min (top); image of cells after treatment with probe MitoClO (2 μM) for 15 min and subsequent treatment of the cells with 100 μM NaClO for 15 min (bottom). (a), (d) Bright-field images of the macrophages cells in samples; (b), (e) green emission (515-555 nm); (c) overlay image of (a) and (b); (f) overlay image of (d) and (e). $\lambda_{ex} = 488$ nm. Scale bar = 10 μm.
**Fig. S4.** Confocal images of MitoClO plus a mitochondrially specific dye in macrophages. (a) Bright-field of macrophages; (b) Fluorescence image of macrophages stained with MitoClO for 15 min and subsequent treatment of the cells with 100 μM NaClO for 15 min, λ<sub>ex</sub> = 488 nm; (c) Fluorescence image of macrophages stained with MitoTracker Deep Red FM, λ<sub>ex</sub> = 635 nm; (d) Merged image of (a), (b) and (c); Scale bar = 10 μm. (e) Colocalization coefficient of MitoClO and MitoTracker Deep Red FM is 0.95.

**Fig. S5.** Confocal fluorescence images of macrophages. RAW264.7 cells incubated with probe MitoClO (2 μM) for 15 min (top); image of cells after treating the cells with LPS (1 μg/mL) for 12 h and PMA (1 μg/mL) for 15 min and subsequent treatment of probe MitoClO (2 μM) for 15 min (bottom). (a), (d) Bright-field images of the macrophages in samples; (b), (e) green emission (515-555 nm); (c) overlay image of (a) and (b); (f) overlay image of (d) and (e). λ<sub>ex</sub> = 488 nm. Scale bar = 20 μm.
4. NMR and MS data for compound 1, 2, 3, 4 and MitoClO

Fig. S6. $^1$H NMR of intermediate 1

Fig. S7. MS of intermediate 1
Fig. S8. $^1$H NMR of intermediate 2

Fig. S9. $^{13}$C NMR of intermediate 2
Fig. S10. MS of intermediate 2

Fig. S11. $^1$H NMR of intermediate 3
**Fig. S12.** $^{13}$C NMR of intermediate 3

**Fig. S13.** MS of intermediate 3
**Fig. S14.** $^1$H NMR of intermediate 4

**Fig. S15.** $^{13}$C NMR of intermediate 4
Fig. S16. MS of intermediate 4

Fig. S17. $^1$H NMR of MitoClO
**Fig. S18.** $^{13}$C NMR of MitoClO

**Fig. S19.** MS of MitoClO