Supporting information

Mitochondrial Directed Ratiometric Fluorescent Probe for Quantitively Detection of Sulfur Dioxide Derivatives

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Cytotoxicity assay

HeLa cells were cultured in culture media (DMEM) in an atmosphere of 5% CO₂ and 95% air at 37 °C. The cells were seeded into 96-well plates at a density of 5 × 10³ cells per well in culture media, then 0, 5, 10, 15, 20, and 25 μM MN (final concentration) were added, respectively. Next, the cells were incubated at 37 °C in an atmosphere of 5% CO₂ and 95% air for 24 h. Finally, 10 μL 3-(4,
5-dimethylthiazol-2-yl)-2, 5-diphenyltetrazolium bromide (MTT, 5 mg/mL) was added and the cells were cultured for another 4 h, respectively. When the purple precipitate is clearly visible under the microscope, add 100 μL DMSO to all wells, and swirl gently. Then measure the absorbance in each well, including the blanks, at 570 nm in a microtiter plate reader (Bio-Rad 680).

**Supplement Date**

Scheme. S1 The synthesis route of the probe MN

Fig. S1 Proposed reaction mechanism of MN with SO$_3^{2-}$/HSO$_3^−$

Fig. S2 The linear relationship between the fluorescence intensity ratio of the probe (10 μM) and the SO$_3^{2-}$/HSO$_3^−$ concentration (1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20 μM). Excitation at 405 nm.
Fig. S3 The fluorescence intensity of probe MN (10 µM) in the absence and presence of SO₃²⁻/HSO₃⁻ (10 µM) changes with the pH of PBS buffer solution (pH 7.4, PO₄³⁻ = 20 mM).

Excitation at 405 nm.

Fig. S4 Fluorescence responses of the probe (10 µM) toward various analytes: (100 µM): (1) Br⁻; (2) Cl⁻; (3) ClO⁻; (4) Hcy; (5) Cys; (6) GSH; (7) HPO₄²⁻; (8) SO₄²⁻; (9) NO₂⁻; (10) NO₃⁻; (11) ACO⁻; (12) HS⁻; (13) S²⁻; (14) SO₂O₃²⁻; (15) probe MN; (16) SO₃²⁻/HSO₃⁻ (20 µM) (except: GSH 1 mM, Hcy 1 mM, Cys 1 mM) in PBS buffer solution (pH 7.4, PO₄³⁻ = 20 mM).
Fig. S5 The photostability of the probe (10 μM) detected in PBS buffer, pH = 7.4, 20 mM at room temperature. Slit width: 5 nm/5 nm.

Fig. S6 (A) Absorption spectra and (B) fluorescence spectra of MN (10 μM) in the absence of SO$_3^{2-}$/HSO$_3^{-}$ (10 μM) at room temperature and 37°C, respectively. The measurements were performed in PBS (20 mM, pH = 7.4). Excitation wavelength = 405 nm. Slit width: 5 nm/5 nm.

Fig. S7 Cytotoxicity assays of MN in HeLa cells. Cells were treated with different concentrations of probe MN for 24 h. Data are expressed as the mean ± SD.

Fig. S8 $^1$H NMR spectrum of probe MN
Fig. S9 High resolution HRMS chart of probe MN treated without and with SO$_3^{2-}$/HSO$_3^-$