A novel near-infrared ratiometric fluorescent probe for SO_2 detection with a large emission shift

Shaohui Han,[a] Xiuxiu Yue,[a] Jingpei Wang,[a] Yun Zhang,[a] Benhua Wang,[a,] * and Xiangzhi Song[a, b]

[a] College of Chemistry & Chemical Engineering, Central South University, Changsha, Hunan 410083, China

[b] Key Laboratory of Hunan Province for Water Environment and Agriculture Product Safety, Changsha, Hunan 410083, China

*Corresponding authors.

Fax: +86-731-88836954; E-mails: benhuawang@csu.edu.cn; song@rowland.harvard.edu

Experimental

Reagents, materials and apparatus

Unless otherwise stated, all reagents were purchased from commercial suppliers and used without further purification. Solvents were purified by standard methods prior to use. Silica gel thin layer chromatography (TLC) plates and silica gel (200-300 mesh) were purchased from Qingdao Ocean Ltd Co. All the reactions were monitored by TLC, and the intermediates were purified by a silica gel column. ^{1}HNMR and ^{13}C NMR were recorded on Bruker Avance 400 NMR and 500 NMR spectrometers. Twice-distilled water was used throughout all experiments. The fluorescence and UV-visible absorption spectra were measured on Hitachi F7000 spectrofluorometer and UV-2450 spectrophotometer, respectively. A Leici PHS-3C meter was used for pH measurements. Fluorescence imaging experiments were performed on Olympus FV1000 and Nikon ARsiMP confocal microscopes.

Spectral measurements

All spectral measurements were carried out in PBS buffer (10.0 mmol/L, pH = 7.4) containing 50% DMSO at 25 ℃. Fluorescence spectra were monitored with excitation wavelengths at 414 nm and 544 nm, respectively, and the slit widths were 10.0 nm/10.0 nm.

Detection limit

Detection limit was calculated according to the following equation:

$$\text{Detection limit} = 3\sigma/k$$

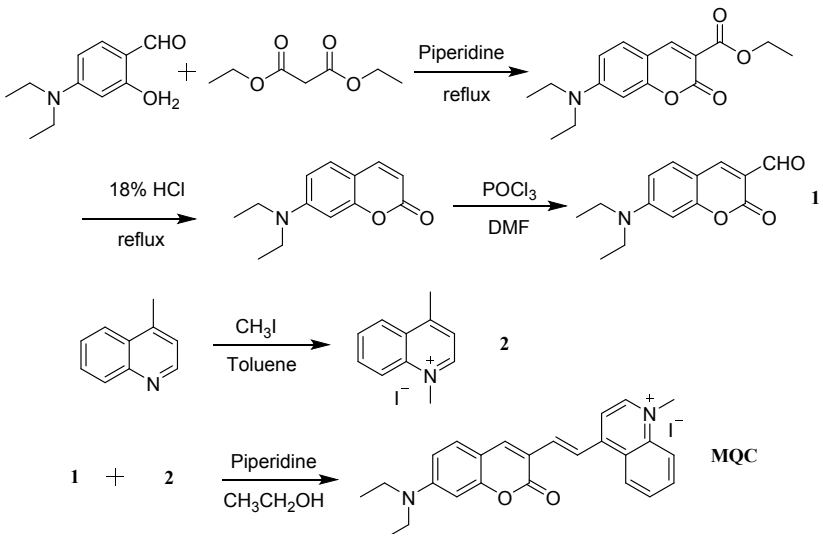
Where σ is the standard deviation of blank measurement, k is the slope between the $F_{470\ nm}/F_{715\ nm}$ versus SO_3^{2-} concentration.

Cell culture and fluorescence imaging experiments

Living MCF-7 cells were grown in Dulbecco's modified Eagle's medium (DMEM) supplemented with 10% fetal bovine serum and 1% penicillin under a humidified

atmosphere on glass-botton culture dishes and allowed to adhere for 24 h. Before use, cells were washed three times with PBS buffer. Cells were firstly incubated with **MQC** (10 μmol/L) for 20 min at 37 ℃, and then treated with Na_2SO_3 (0.3 mmol/L) for 20 min at 37 ℃. For the control experiment, MCF-7 cells were only incubated with **MQC** (10 μmol/L) for 20 min at 37 ℃. Cells were washed with PBS buffer before cell imaging experiments.

Synthesis



Scheme S1. The synthetic route of probe **MQC**.

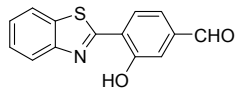
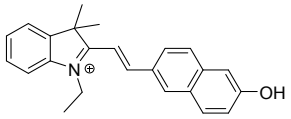
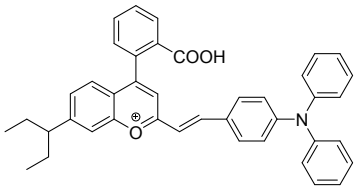
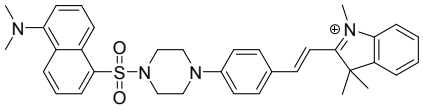
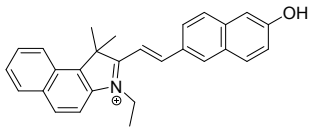
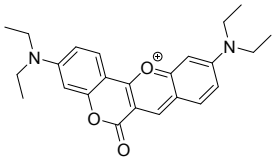
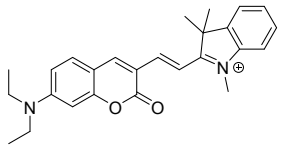
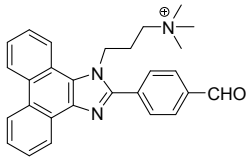
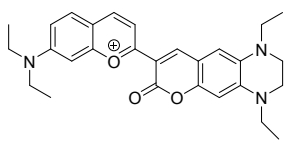
Compounds **1** and **2** were synthesized according to the reported procedures in the literature[1-2].

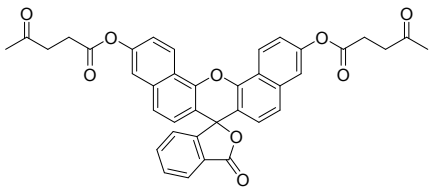
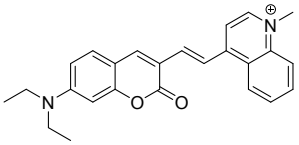
Synthesis of MQC

Compounds **1** (49.0 mg, 0.20 mmol/Lol) and **2** (68.4 mg, 0.24 mmol/Lol) and 5 μL piperidine were dissolved in 10 mL anhydrous ethanol. Then the mixture was stirred at 80 ℃ under nitrogen for 5 h. After ethanol was evaporated under a reduced pressure, the crude product was purified by silica gel flash chromatography with CH_2Cl_2 to CH_2Cl_2/MeOH=20:1 as eluent to afford compound **MQC** as a black powder (75.0 mg, 73.3%). ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.24 (d, J = 6.6 Hz, 1H), 8.68 (d, J = 8.5 Hz, 1H), 8.48 (s, 1H), 8.42 - 8.32 (m, 3H), 8.26 - 8.17 (m, 1H), 8.01 (dd, J = 17.3, 11.7 Hz, 2H), 7.51 (d, J = 9.0 Hz, 1H), 6.79 (dd, J = 9.0, 1.8 Hz, 1H),

6.56 (s, 1H), 4.50 (s, 3H), 3.49 (dd, J = 13.7, 6.7 Hz, 4H), 1.16 (t, J = 6.9 Hz, 6H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 160.46, 156.96, 152.80, 152.61, 148.15, 145.62, 139.16, 138.69, 135.30, 131.37, 129.78, 126.42, 126.07, 119.85, 118.83, 115.79, 114.36, 110.69, 109.03, 96.76, 56.49, 45.00, 44.92, 19.04, 12.91. MS (ESI) m/z calcd for $C_{25}H_{25}N_2O_2$ $[M]^+$: 385.1911, found: 385.1893.

Table. S1 Some fluorescent probes for SO_2.

Probes	Types	Emission wavelength /nm	Emission shift/nm	Response time	Medium	Ref.
	ratiometric	467, 563	96	seconds	PBS buffer (containing 10% CTAB)	3
	ratiometric	473, 573	100	5 min	PBS buffer (pH = 7)	4
	ratiometric	520, 740	220	80 min	PBS buffer (containing 10% DMSO)	5
	ratiometric	530, 582	52	2 min	PBS buffer (containing 30% DMF)	6
	ratiometric	467, 593	126	30 s	PBS buffer (pH = 7)	7
	ratiometric	450, 645	195	seconds	PBS buffer (containing 5% DMSO)	8
	ratiometric	475, 650	175	5 min	PBS buffer (containing 30% EtOH)	9
	turn on	388	0	30 s	PBS buffer (containing 2.5% CTAB)	10
	turn on	585	0	15 s	HEPES buffer	11

	turn on	695	0	20 min	PBS buffer (containing 50% DMSO)	[12]
	ratiometric	470, 715	245	5 min	PBS buffer (containing 50% DMSO)	this work

Characterization data

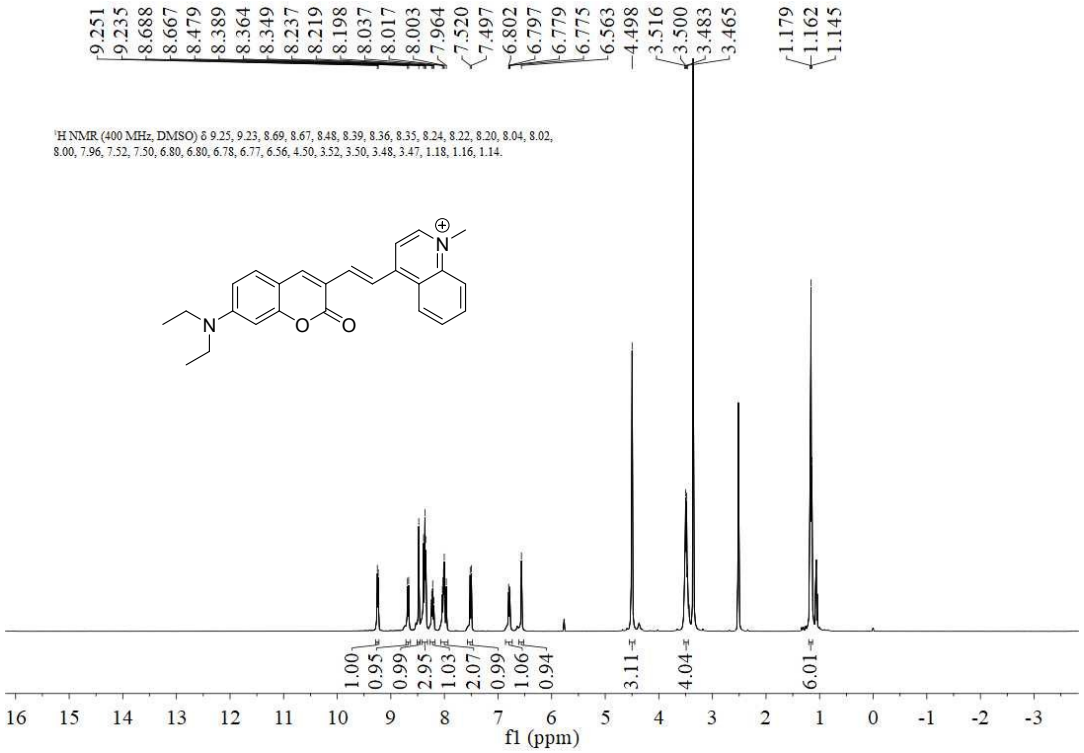


Fig. S1 ^{1}H NMR spectrum of probe **MQC** in DMSO-d_6.

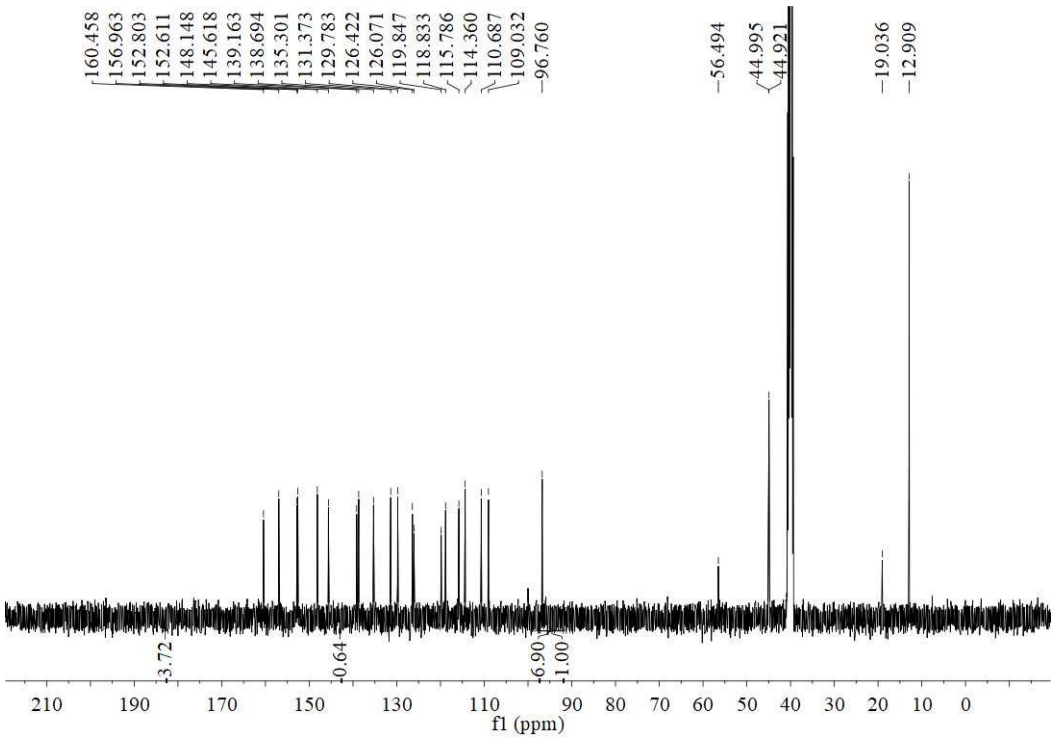


Fig. S2 ^{13}C NMR spectrum of probe **MQC** in DMSO-d_6.

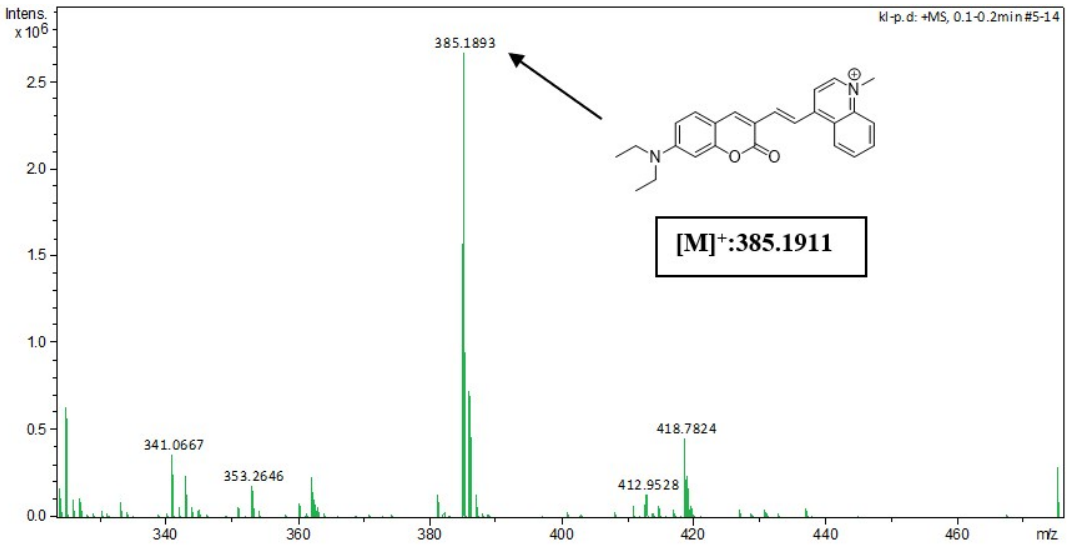


Fig. S3 HRMS spectrum of probe **MQC**.

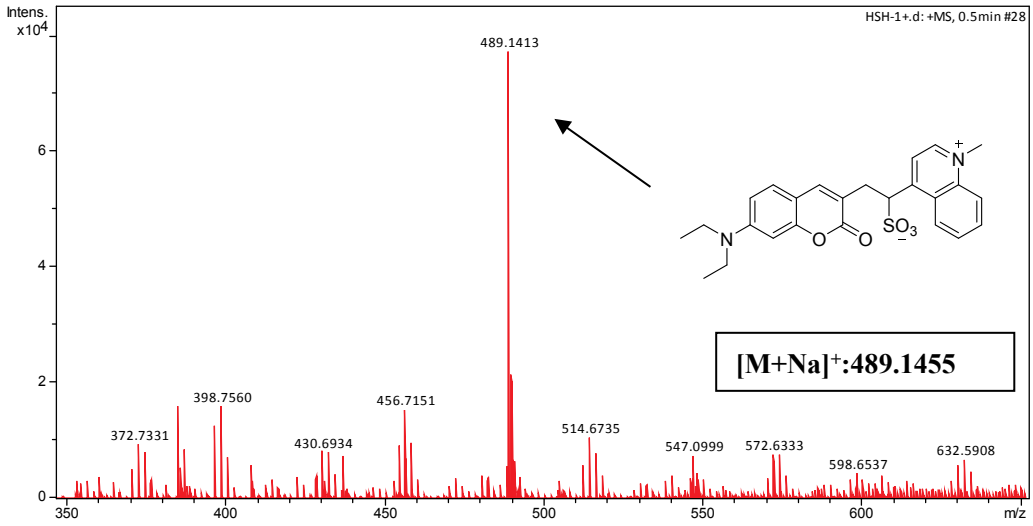


Fig. S4 HRMS spectrum of probe **MQC** in the presence of SO_3^{2-}.

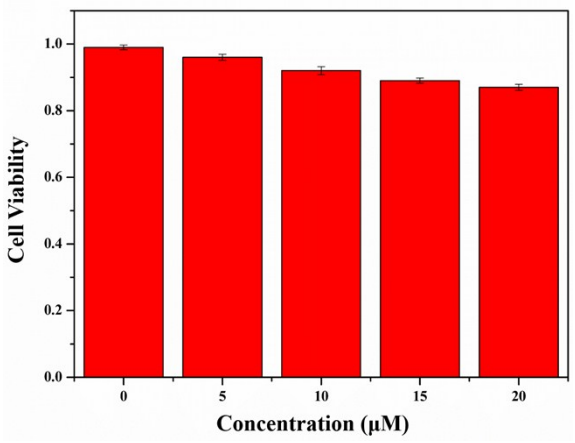


Fig. S5 MTT assay of MCF-7 cells incubated with different concentrations of probe **MQC**.

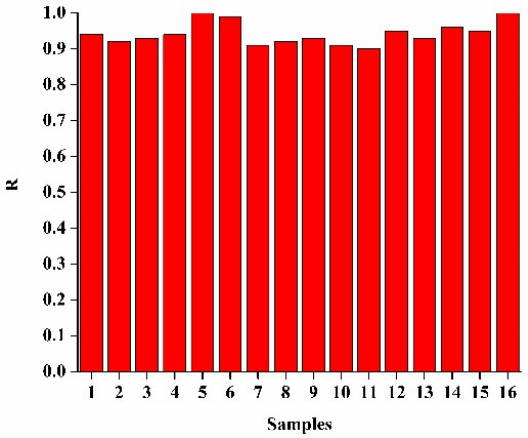


Fig. S6 The fluorescence intensity ratio $F_{470\ nm}/F_{715\ nm}$ of probe **MQC** (10 μM) with Na_2SO_3 (300 μM) in the co-existence of relevant species in PBS buffer (10.0 mM, pH = 7.4, containing 50% DMSO). Species: 1. Zn^{2+}; 2. Fe^{2+}; 3. F^-;4. Cl^-; 5. NO_3^-; 6. NO_2^-; 7. SO_4^{2-}; 8. Cys; 9. Hcy; 10. GSH; 11. BHP; 12. ROO^-; 13. H_2O_2; 14. NO; 15. S^{2-}; 16 SO_3^{2-}. The Y-axis R represents the ratio of $F_{470\ nm}/F_{715\ nm}$ of probe **MQC** and Na_2SO_3 with and without other species.

Reference

1. M. E. Aliaga, M. Gazitua, A. Rojas-Bolaños, M. Fuentes-Estrada, D. Durango and O. García-Beltrán, *Spectrochim. Acta, Part A,* 2020, **224**, 117372.
2. M. Xiang, H. Huang, X. Liu, Z. Tong, C. Zhang, F. Wang, R. Yu and J. Jiang, *Anal.Chem.,* 2019, **91**, 5489-5493.
3. X. Liu, Q. Yang, W. Chen, L. Mo, S. Chen, J. Kang and X. Song, *Org. Biomol. Chem.,* 2015, **13**, 8663-8668.
4. C. Gao, Y. Tian, R. Zhang, J. Jing and X. Zhang, *New J. Chem.,* 2019, **43**, 5255-5259.
5. K. Liu, Y. Chen, H. Sun, S. Wang and F. Kong, *J. Mater. Chem. B,* 2018, **6**, 7060-7065.
6. D. Li, Z. Wang, X. Cao, J. Cui, X. Wang, H. Cui, J. Miao and B. Zhao, *Chem. Comm.,* 2016, **52**, 2760-2763.
7. Y. Wang, Q. Meng, R. Zhang, H. Jia, X. Zhang and Z. Zhang, *Org. Biomol. Chem.,* 2017, **15**, 2734-2739.
8. Y. Ma, Y. Tang, Y. Zhao, S. Gao and W. Lin, *Anal.Chem.,* 2017, **89**, 9388-9393.
9. W. Xu, C. L. Teoh, J. Peng, D. Su, L. Yuan and Y. Chang, *Biomaterials,* 2015, **56**, 1-9.
10. H. Li, Q. Yao, J. Fan, C. Hu, F. Xu, J. Du, J. Wang and X. Peng, *Ind. Eng. Chem. Res.,* 2016, **55**, 1477-1483.
11. W. Chen, Q. Fang, D. Yang, H. Zhang, X. Song and J. Foley, *Anal. Chem.,* 2015, **87**, 609-616.
12. H. Zhang, S. Xue and G. Feng, *Sens. Actuators, B,* 2016, **231**, 752-758.