

Supporting Information

Dong-Shuai Shang^{a,†}, Ming-Zhu Shao^{a,†}, Sheng-Shuo Xiao^b, Yun-Bo Wei^b, Xin-Yu Zhou^a, Ya-Nan Jin^a, Li-Ya Xin^a, Yu Liu^a, Zhong-Long Zhou^a, Chang-Sheng Xu^c, Cheng-Shi Jiang^{a,*}, Ning Meng^{a,*}, Kai-Ming Wang^{a,*}

^a *School of Biological Science and Technology, University of Jinan, Jinan 250022, China*

^b *Laboratory of Immunology for Environment and Health, Shandong Analysis and Test Center, Qilu University of Technology (Shandong Academy of Sciences), Jinan 250022, China*

^c *Thoracic Surgery Department, Lung Cancer Center, The Third Affiliated Hospital of Shandong First Medical University, Jinan 250000, China*

1. The Synthesis of Compound 1

4-(Diethylamino)-2-hydroxybenzaldehyde (3.86 g, 20 mmol) was placed in a 150 mL round-bottom flask and dissolved in 50 mL of anhydrous ethanol. Ethyl acetoacetate (3.12 g, 24 mmol) was then added, followed by the dropwise addition of 5 drops of anhydrous piperidine. Under a nitrogen atmosphere, the mixture was stirred and refluxed at 80 °C for 16 hours. After cooling, the reaction mixture was filtered by suction to afford a golden-yellow solid product (3.96 g, 76.4% yield). ¹H NMR (600 MHz, DMSO) δ 8.47 (s, 1H), 7.64 (d, *J* = 9.0 Hz, 1H), 6.78 (dd, *J* = 9.0, 2.4 Hz, 1H), 6.56 (d, *J* = 2.4 Hz, 1H), 3.48 (q, *J* = 7.2 Hz, 4H), 2.49 (s, 3H), 1.13 (t, *J* = 7.2 Hz, 6H). ¹³C NMR (150 MHz, DMSO-*d*₆) δ 194.69, 160.34, 158.74, 153.42, 148.12, 132.90, 115.45, 110.59, 107.94, 96.25, 44.89, 30.60, 12.80. HR-MS *m/z*: calculated for C₁₅H₁₇NO₃⁺ [M+H]⁺: 260.1287, found: 260.1285.

2. Supplementary results and discussion

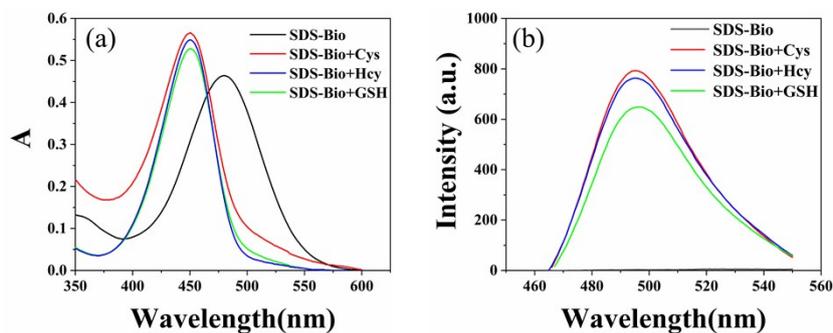


Fig. S1. The UV-Vis (a) and fluorescence spectra (b) of probe SDS-Bio in response to biothiols(Cys/Hcy/GSH)(300 μM).

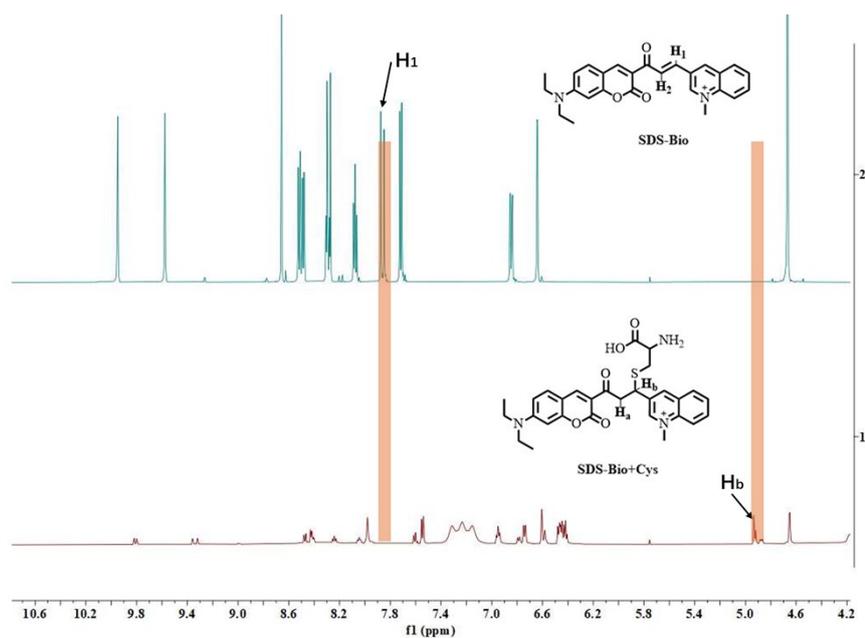


Fig. S2. Comparison of the ^1H NMR spectra of SDS-Bio in $\text{DMSO-}d_6$ with and without treatment using 10 equivalents of cysteine.

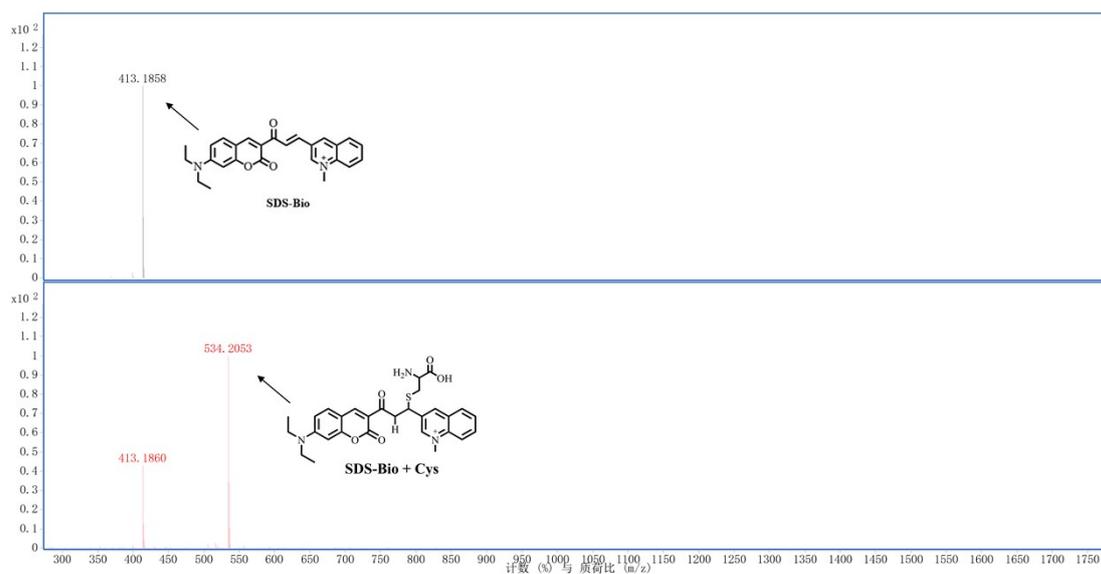


Fig. S3. HRMS spectra of SDS-Bio treated with and without 10 equivalents of cysteine.

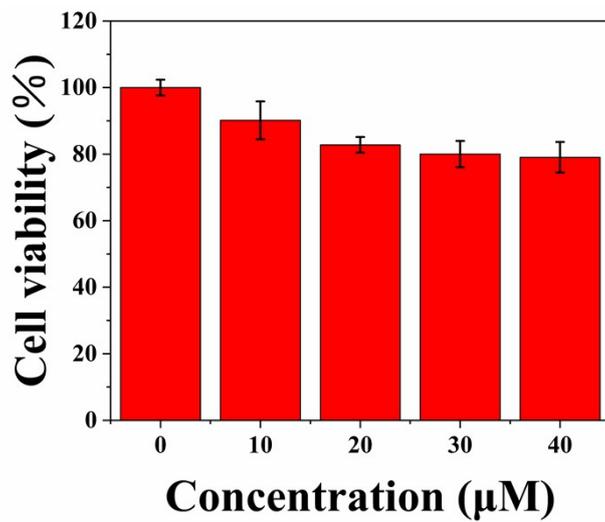


Fig. S4. Cell viability assay of probe SDS-Bio.

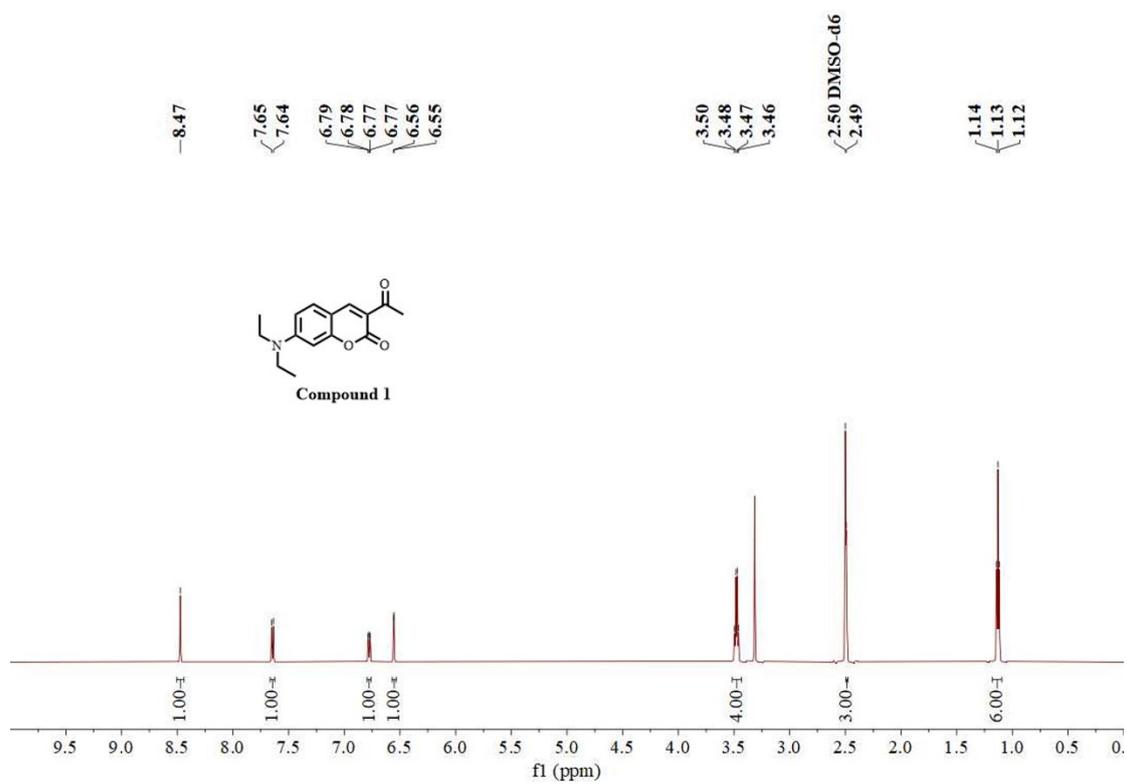
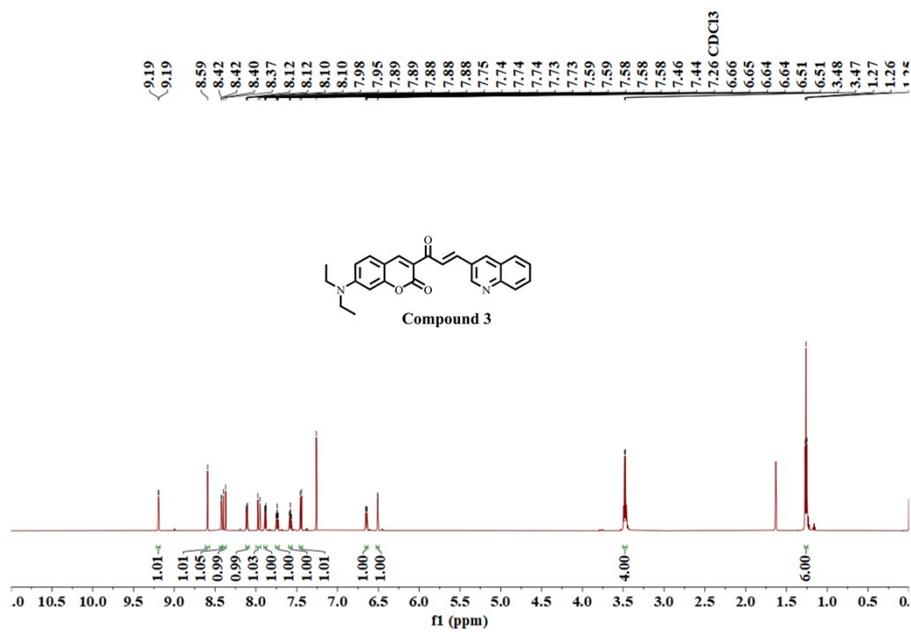
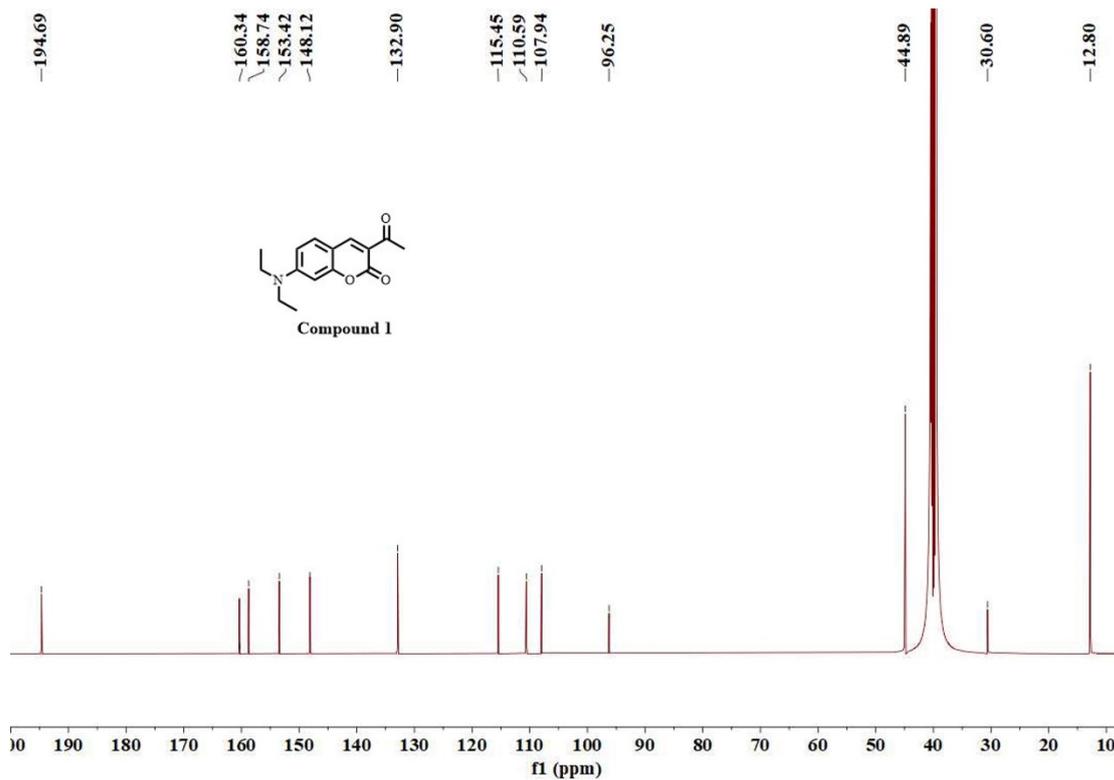


Fig. S5. ^1H NMR spectra of compound 1 in $\text{DMSO-}d_6$



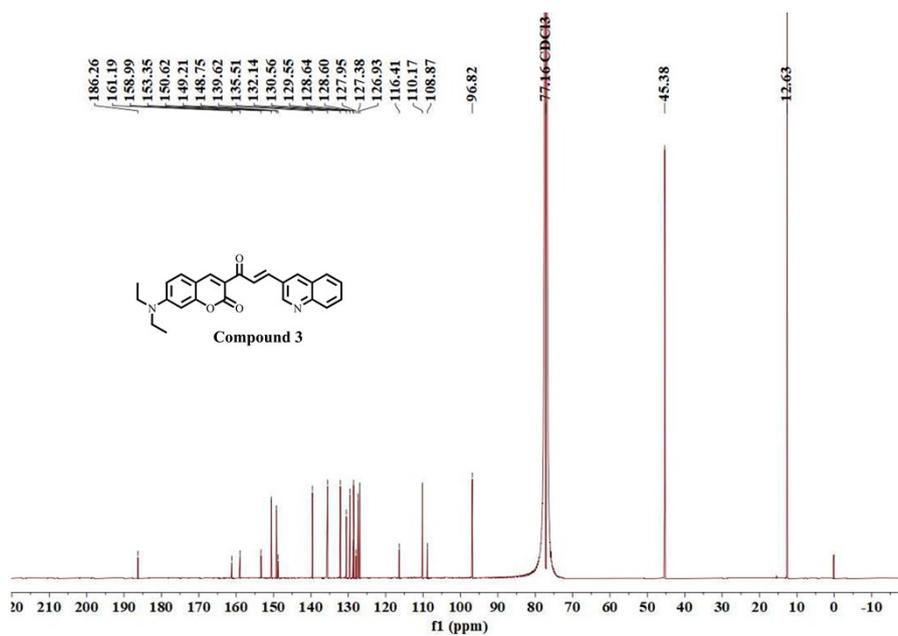


Fig. S8. ¹³C NMR spectrum of compound 3 in CDCl₃

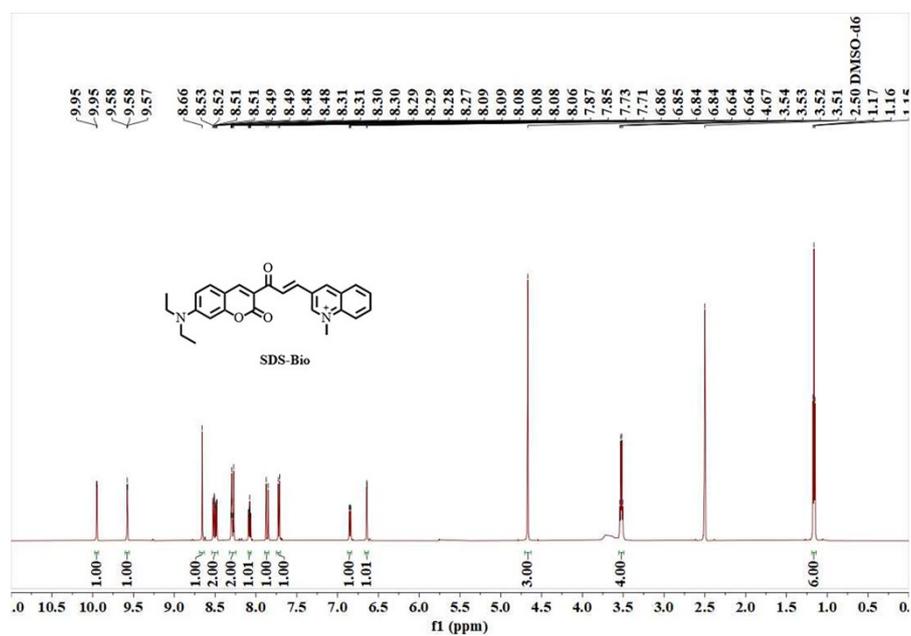


Fig. S9. ¹H NMR spectra of SDS-Bio in DMSO-d₆

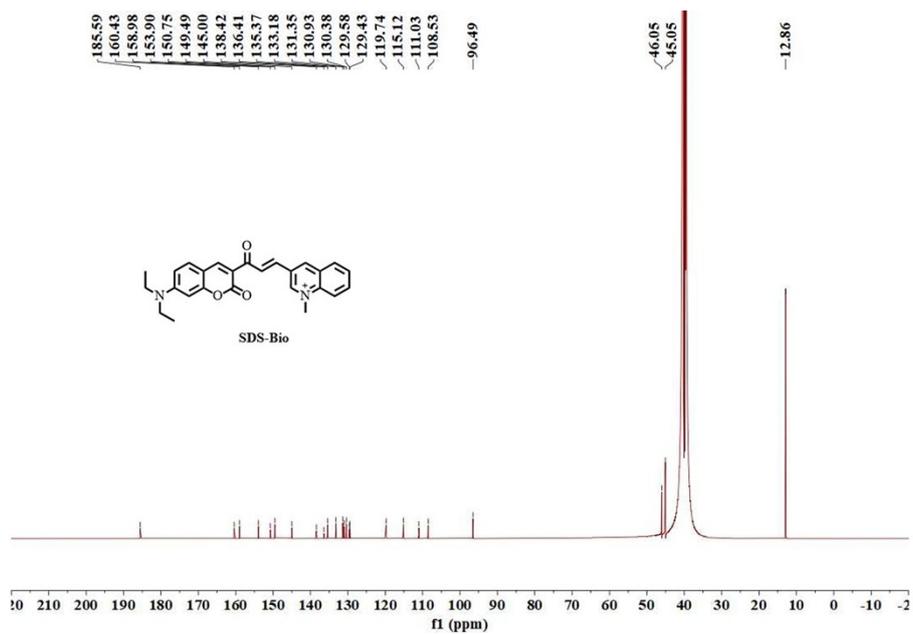


Fig. S10. ¹³C NMR spectrum of SDS-Bio in DMSO-*d*₆

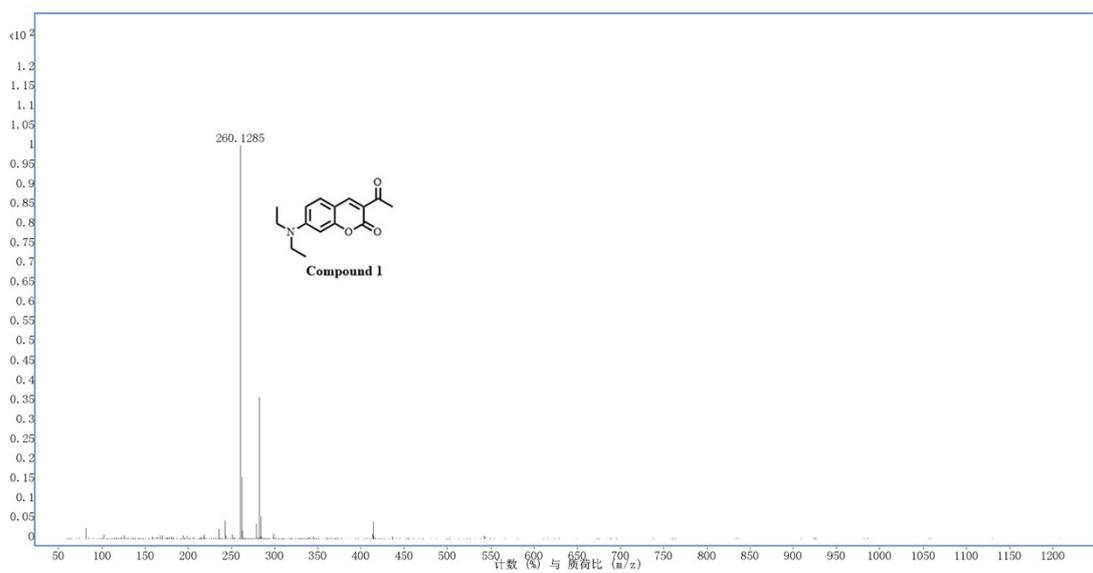


Fig. S11. HRMS spectrum of compound 1

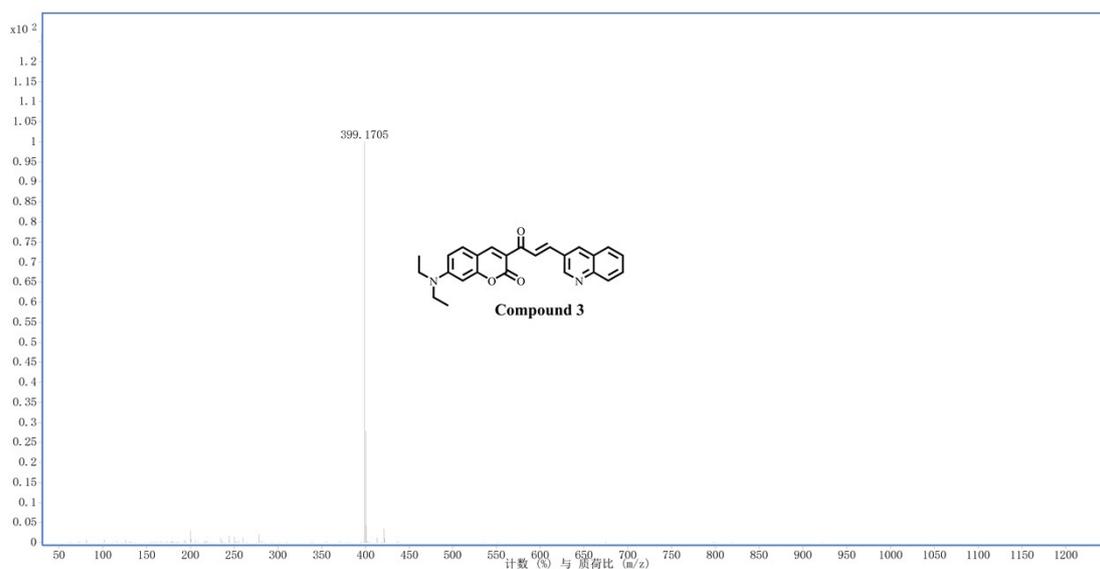


Fig. S12. HRMS spectrum of compound 3

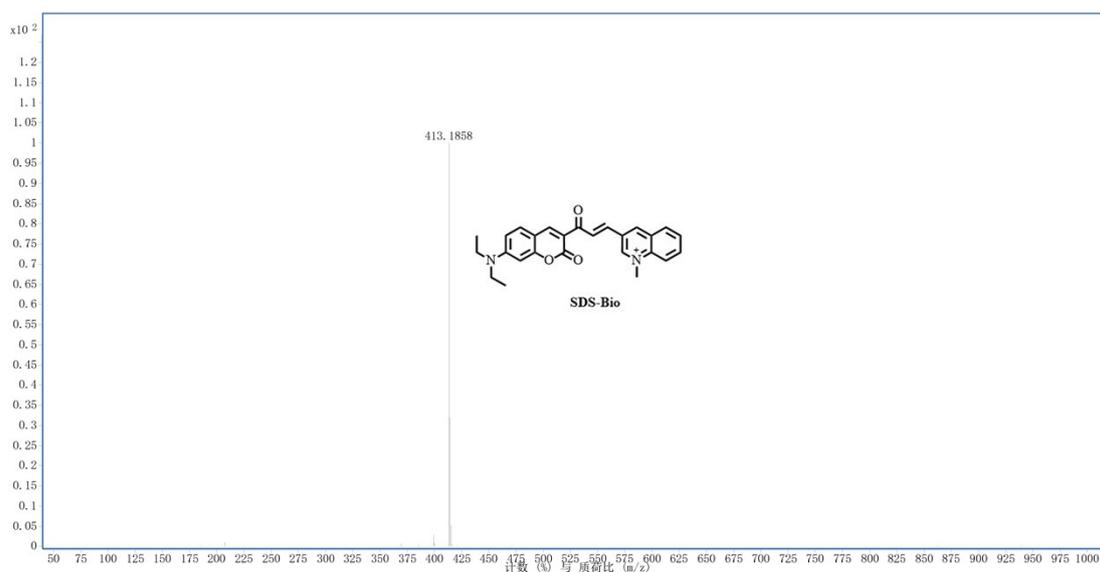


Fig. S13. HRMS spectrum of compound Probe

Table S1. This study compares SDS-Bio with other reported probes for detecting biothiols.

| Probe | Solution | $\lambda_{\text{ex}}/\lambda_{\text{em}}$ (nm) | The detection limit. (Cys/Hcy/GSH) | Response time |
|---------------------|-----------------------------------|--|---------------------------------------|---------------|
| MCY-NB ¹ | PBS (10 mM, 20% DMSO , pH=7.4) | 522 nm 566 nm | 330 nM/319 nM/174 nM | 20 min. |
| BD-MSN ² | PBS (10 mM, 20% DMSO , pH=7.4) | 550 nm/635 nm | 3.4 nM/3.3 nM /3.4 nM | 50 min |

| | | | | |
|------------------------|-------------------------------|----------------------|---|--|
| 3-STF ³ | PBS (10 mM, 20% DMSO, pH=7.4) | 390 nm 536/452 nm | 1.82 μM/0.46μM/0.67μM | / |
| XB-F ⁴ | PBS (pH = 7.4, 10 mM) | 539/700 nm | 119 nM/152 nM/ 121 nM | < 6 min |
| Cy-DNBS ⁵ | PBS (10 mM, 10% DMSO, pH=7.4) | 540 nm/604 nm | 1.34×10^{-7} M/ 2.29×10^{-7} M/ 1.38×10^{-7} M | 8 min |
| Probe 1 ⁶ | PBS (10 mM, 50% DMSO, pH=7.4) | 458 nM 498/567 nm | 149 nM /116 nM /133 nM | 15 min |
| TMN-Cl-S ⁷ | PBS (10 mM, 20% DMSO, pH=7.4) | 430 nm/578 nm | 3.41 nM/ 3.39 nM 3.38 nM | 30 min |
| BT-DNBS ⁸ | PBS (10 mM, 10% DMSO, pH=7.4) | 434 nm/630 nm | 49 nM/80 nM/83 nM | 1200 s |
| Acr-QX9A ⁹ | PBS (10 mM, 50% DMSO, pH=7.4) | 460 nm/532 nm | 75.2 nM/92.7 nM/370 nM | 25 min |
| LY-DCM-P ¹⁰ | PBS (10 mM, 40% DMSO, pH=7.4) | 520 nm/670 nm | 42.9 nM/75.3 nM/140.4 nM | Cys:10 min, Hcy:25 min, and GSH:50 min |
| This work | PBS (10 mM, 30% DMSO, pH=7.4) | 450 nm/495 nm | 48.5 nM/28.7 nM /49.3 nM | Cys:<30s, Hcy:200s, and GSH:600s |

References

1. M. Wei, X. Wang, R. Liu, Z. Yang, P. Li, Q. Yao, Y. Su, W. Qian, S. Shan, Y. Bao and L. Niu, *Spectrochim. Acta A*, 2025, **343**, 126618.
2. M. Ali, C. Zhou, Z. Gao, G. Fan, J. Ren, E. Wang and F. Wang, *Dyes Pigm.*, 2025, **235**, 112650.
3. H. Wei, F. Zhan, Y. Zheng and Y. Xu, *Spectrochim. Acta A.*, 2024, **305**, 123487.
4. M. Xie, T. Yang, M. Chen, L. Yu, T. Wang, H. Wei, J. Liu, Q. Jiang and P. Xing, *New J. Chem.*, 2025, **49**, 13958-13962.
5. Y. Zhu, H. Pan, Y. Song, C. Jing, J.-A. Gan and J. Zhang, *Dyes Pigm.*, 2021, **191**, 109376.
6. A. Ren, L. Qiao, K. Li, D. Zhu and Y. Zhang, *Anal. Bioanal. Chem.*, 2024, 416, 6223-6235.
7. Z. Wang, Z. Gao, Y. Gao, X. Pang and X. Xia, *Tetrahedron*, 2025, **179**, 134615.
8. S. Zhang, Y. Fujioka, Y. Ohba and K. Yamada, *RSC Advances*, 2025, **15**, 135-141.
9. Y. Zhang, B. Zhu, L. Sun, Y. Yan, Y. Sun, M. Zhang, L. Huang, H. Fei, C. Yang, Y. Chen, C. Hao, S. Ma, J. Zhu, Z. Wang, X. Han and M. Ji, *Microchem J.*, 2024, **199**, 110011.
10. T. Zhao, T. Zhang, Z. Tao, Z. Zhou, X. Xia, Z. Wu, F. Wang, J. Ren and E. Wang,

iScience, 2024, **27**, 111334.