

Supplementary Data

For

A novel naphthalene-based fluorescent probe for highly selective detection of cysteine with large Stokes shift and its application in bioimaging

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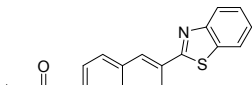
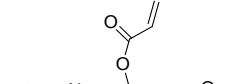
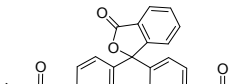
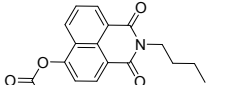
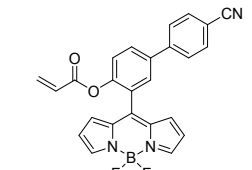
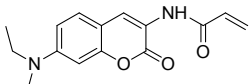
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| | Probe | Solvent (pH=7.4) | Dose of Cys (eq.) | Time (min) | Stokes shift (nm) | Analyte | Detection Limit (μM) | Reference |
|----------|---|--|--------------------------|-------------------|--------------------------|-------------------|---|------------------|
| 1 |  (Ex=365nm,Em=505nm) | DMSO-PBS (1/1,v/v) | 10 | 9 | 140 | Cys | 0.18 | This work |
| 2 |  (Ex=333nm,Em=450 nm) | DMSO-PBS (1/9,v/v) | 10 | 30 | 117 | Cys Hcy GSH | 0.80 | [1] |
| 3 |  (Ex=450nm,Em=520 nm) | EtOH-water (2/3, v/v) | 14.5 | 60 | 70 | Cys Hcy | 0.50 | [2] |
| 4 |  (Ex=452nm,Em=547 nm) | DMSO-PBS (containing 0.2% DMSO) | 20 | 7 | 95 | Cys Hcy | 1.8 | [3] |
| 5 |  (Ex=503nm,Em=525 nm) | CH ₃ CN-H ₂ O (1:1,v/v) | 50 | 150 | 22 | Cys | 0.037 | [4] |
| 6 |  (Ex=380nm,Em=490 nm) | H ₂ O-CH ₃ OH (1/9,v/v) | 30 | 180 | 110 | Cys Hcy GSH | 0.68 | [5] |

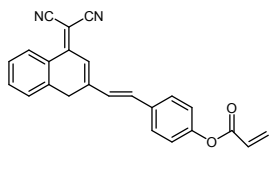
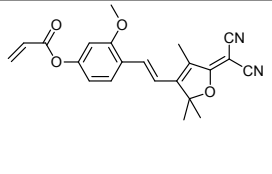
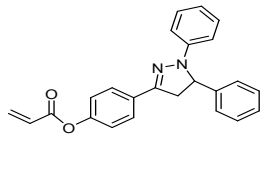
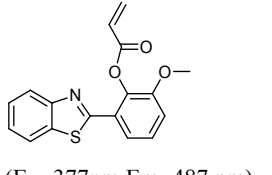
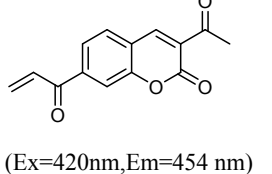
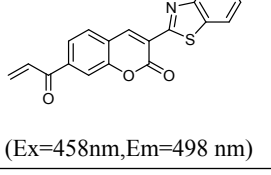
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|-----------|---|-------------------------------------|----|------------|-----|-------------------|---------|------|
| 7 |  (Ex=560nm,Em=676 nm) | DMSO-PBS (1/1,v/v) | 5 | 20 | 116 | Cys Hcy | 0.081 | [6] |
| 8 |  (Ex=585nm,Em=635 nm) | DMSO-PBS (1/1,v/v) | 10 | 30 | 50 | Cys Hcy | 0.3 | [7] |
| 9 |  (Ex=370nm,Em=474 nm) | EtOH-PBS (3/7, v/v) | 10 | no data | 104 | GSH | 0.082 | [8] |
| 10 |  (Ex=377nm,Em=487 nm) | EtOH-H ₂ O (2:8, v/v) | 1 | 40 | 110 | Cys Hcy | no data | [9] |
| 11 |  (Ex=420nm,Em=454 nm) | EtOH-HEPES (2:8, v/v) | 15 | 40 | 34 | Cys GSH | 0.657 | [10] |
| 12 |  (Ex=458nm,Em=498 nm) | DMSO-PBS (1/1,v/v) | 5 | 15 | 40 | Cys Hcy GSH | 0.02 | [11] |

Table S1. The comparison between probe **BTNA** and some similar probes

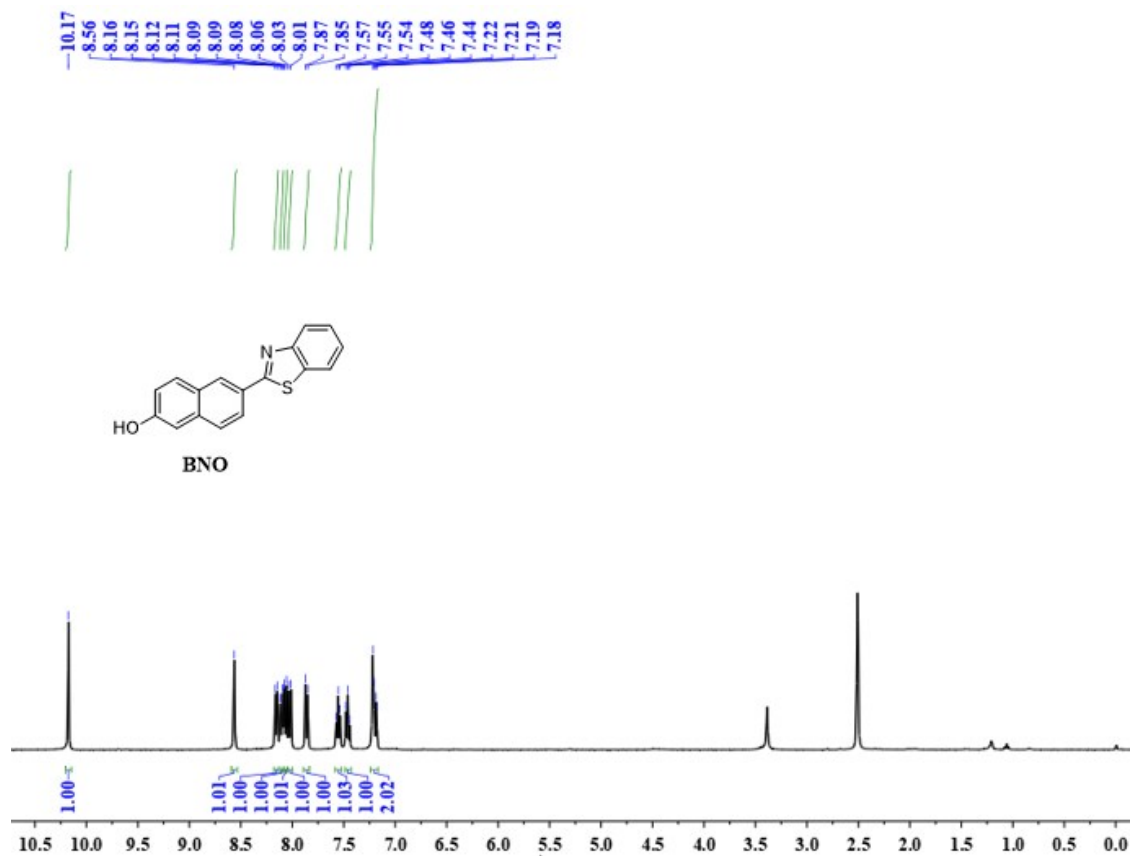


Figure S1. ¹H NMR (400 MHz) spectrum of compound **BNO** in DMSO-*d*₆.

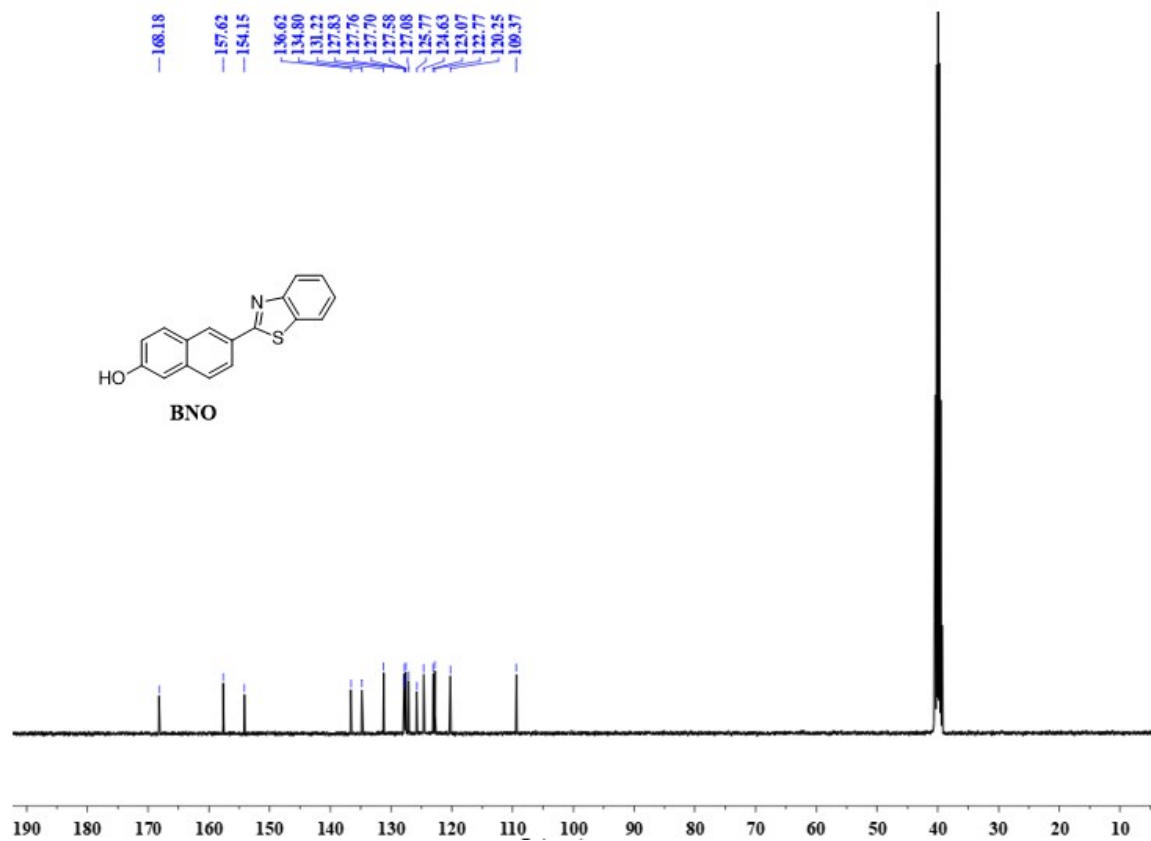


Figure S2. ¹³C NMR (100 MHz) spectrum of compound **BNO** in DMSO-*d*₆.

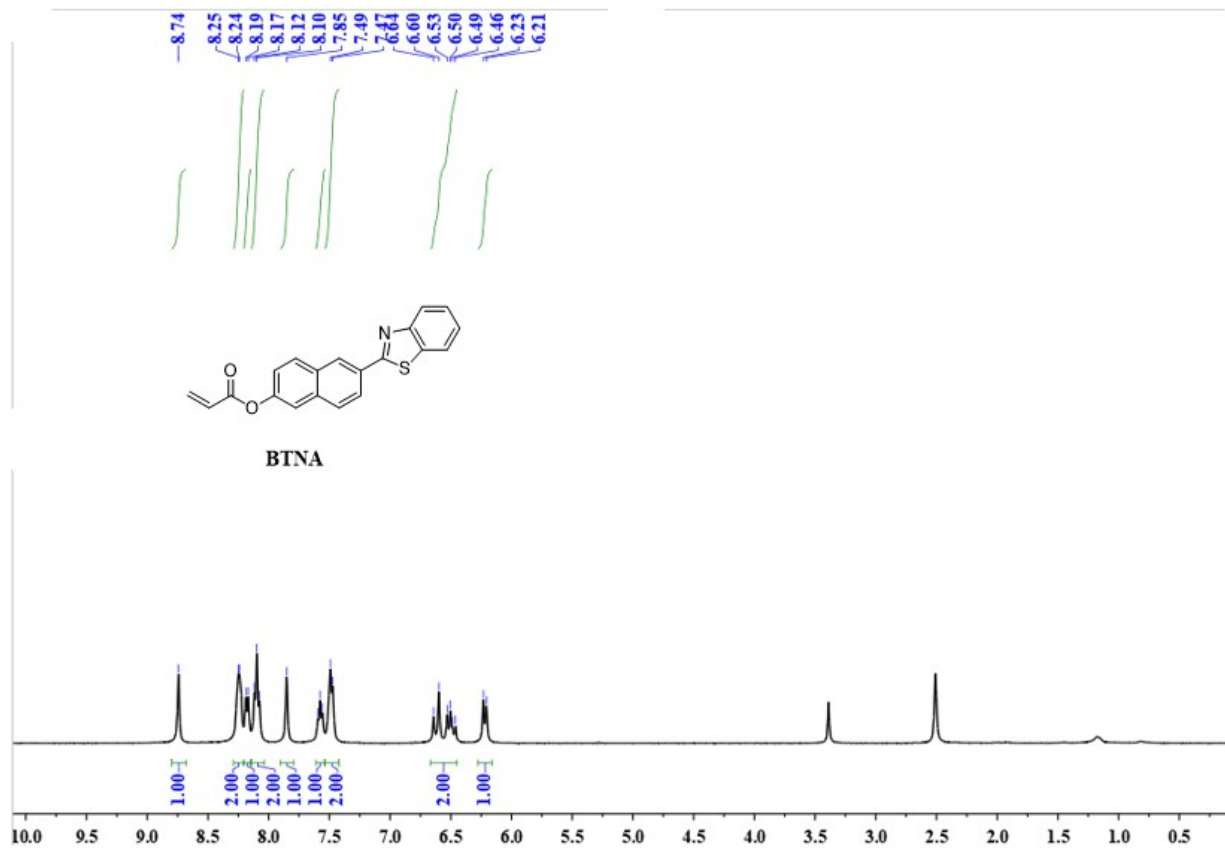


Figure S3. ¹H NMR (400 MHz) spectrum of probe BTNA in DMSO-*d*₆.

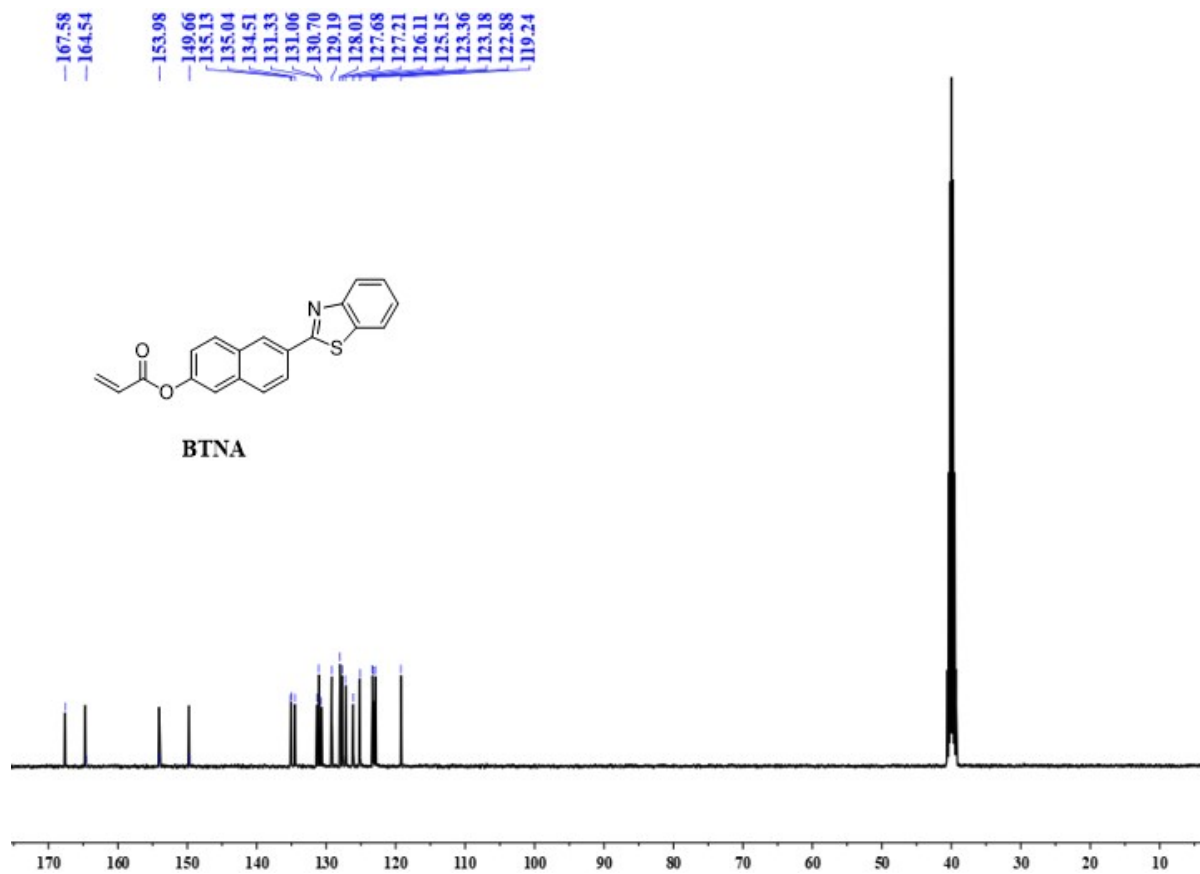


Figure S4. ¹³C NMR (100 MHz) spectrum of probe BTNA in DMSO-*d*₆.

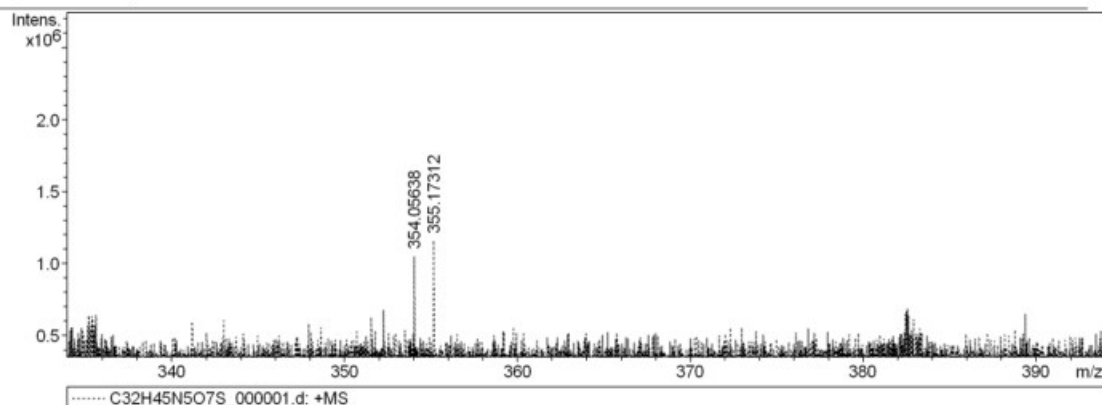
Mass Spectrum SmartFormula Report

Analysis Info

Analysis Name D:\Data\teachers\C32H45N5O7S_000001.d
Method 20180105_4M@Pos_lhl
Sample Name
Comment
Operator
Instrument solariX

Acquisition Parameter

| | | | | | |
|--------------------------|------------|-------------------|-----|-----------------------|--------------------------|
| Polarity | Positive | n/a | n/a | No. of Laser Shots | 200 |
| n/a | n/a | No. of Cell Fills | 1 | Laser Power | 20.0 lp |
| Broadband Low Mass | 101.1 m/z | n/a | n/a | n/a | n/a |
| Broadband High Mass | 1500.0 m/z | n/a | n/a | n/a | n/a |
| Acquisition Mode | Single MS | n/a | n/a | Calibration Date | Fri Jan 5 11:24:10 2018 |
| Pulse Program | basic | n/a | n/a | Data Acquisition Size | 4194304 |
| Source Accumulation | 0.050 sec | n/a | n/a | Apodization | Sine-Bell Multiplication |
| Ion Accumulation Time | 0.050 sec | n/a | n/a | | |
| Flight Time to Acq. Cell | 0.001 sec | | | | |



Meas. m/z # Formula Score m/z err [ppm] Mean err [ppm] mSigma rdb e⁻ Conf N-Rule

Figure S5. HRMS (ESI⁺) spectrum of probe BTNA.

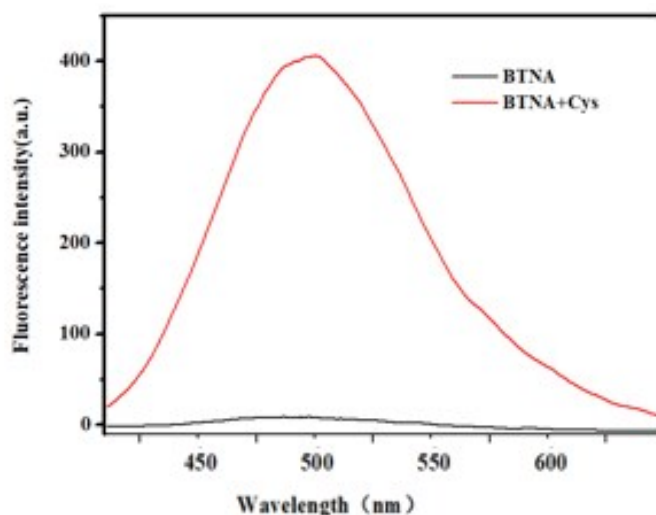


Figure S6. Fluorescence spectra of BTNA by itself (10 μ M, black line), and BTNA (10 μ M) in the presence of Cys (1 mM, red line). Ex=365 nm.

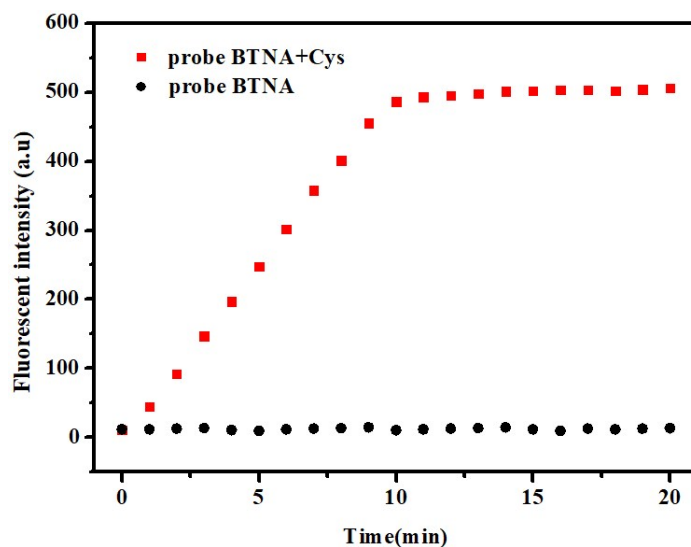


Figure S7. Time-dependent fluorescence intensity of **BTNA** (10 μ M) at 511 nm in the absence and presence of **Cys** (60 μ M) in PBS buffer (50% DMSO) solution. Ex=365 nm.



Figure S8. Emission color changes of probe **BTNA** with various analytes.

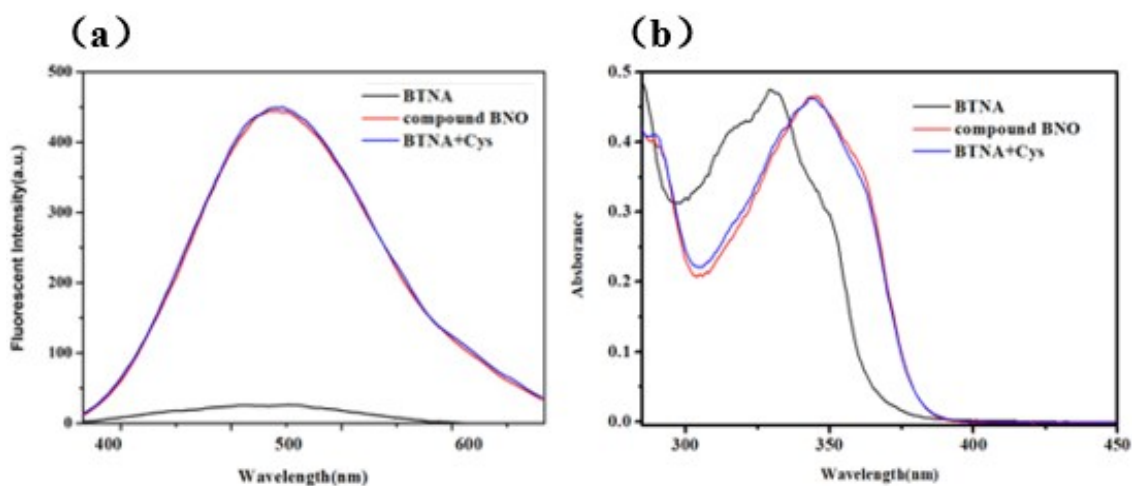
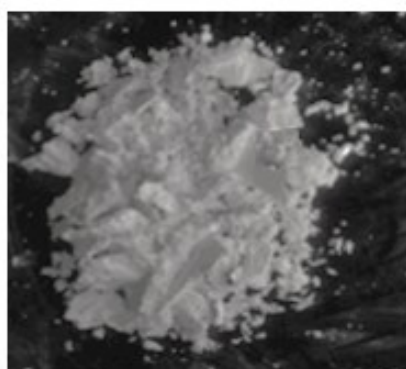
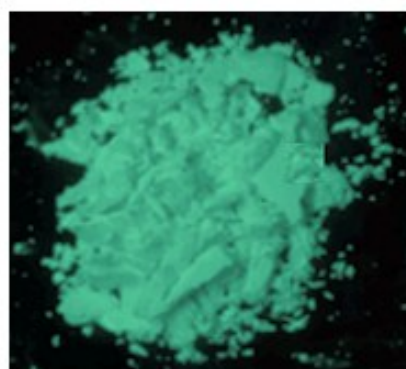


Figure S11. A comparison of (a) fluorescent spectra and (b) absorbance spectra of probe **BTNA** (10 μM), compound **BNO** (10 μM), and the mixture of probe **BTNA** (10 μM) + Cys (50 μM) after 15 min in PBS buffer (50% DMSO) solution.



BNO under room light



BNO under light of 365 nm

Figure S12. The color of solid compound **BNO** under room light and UV light.



Figure S13. Fluorescence changes in test paper (under a 365 nm UV lamp) with probe **BTNA** upon addition of some representative analytes (1. none, 2. Ala, 3. Arg, 4. Asn, 5. Asp, 6. Gln, 7. Glu, 8. His, 9. HIE, 10. Leu, 11. Lys, 12. Met, 13. Phe, 14. Pro, 15. Ser, 16. Trp, 17. Tyr, 18. GSH, 19. Hcy, 20. Cys. Except GSH were used 1mM, others were used 200 μM for the test).

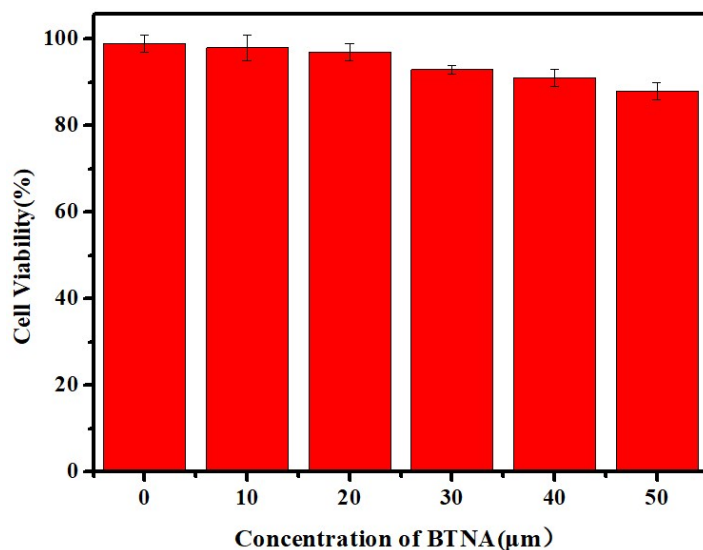


Figure S14. Cell viability to HeLa cells of probe **BTNA**.

References

- [1] Haiyan Zhang, Weiyong Feng, Guoqiang Feng, A simple and readily available fluorescent turn-on probe for cysteine detection and bioimaging in living cells, *Dyes and Pigments*. 139 (2017) 73-78.
- [2] Liguang Wang, Qi Zhou, Baocun Zhu, Lianguo Yan, Zhenmin Ma, Bin Du, Xiaoling Zhang, A colorimetric and fluorescent chemodosimeter for discriminative and simultaneous quantification of cysteine and homocysteine, *Dyes and Pigments*. 95 (2012) 275-279.
- [3] Jinmin Shi, Yujiao Wang, Xiaoliang Tang, Wei Liu, Huie Jiang, Wei Dou, Weisheng Liu. A colorimetric and fluorescent probe for thiols based on 1, 8-naphthalimide and its application for bioimaging, *Dyes and Pigments*. 100 (2014) 255—260.
- [4] Wenlong Fan, Ximing Huang, Xiaomin Shi, Zhuo Wang, Zhengliang Lu, Chunhua Fan, Qibing Bo. A simple fluorescent probe for sensing cysteine over homocysteine and glutathione based on PET, *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*. 173 (2017) 918–923.
- [5] Yuchen Liao, Paribthan, Lingfang Wei, Shupao Wu. A coumarin-based fluorescent probe for

- thiols and its application in cell imaging. *Sensors and Actuators B*.332 (2016) 732-737
- [6] De huan Yu, Qiong Zhang, Shuangshuang Ding and Guoqiang Feng, A colorimetric and near-infrared fluorescent probe for biothiols and its application in living cells, *RSC Adv.* 4 (2014) 46–61.
- [7] Ji Ting, Hou Jin Yang, Kun Li, Kang Yu, Xiao Qi Yu. A colorimetric and red emissive fluorescent probe for cysteine and its application in bioimaging, *Sensors and Actuators B Chem.* 214 (2017) 92-100.
- [8] Shengqing Wang, Qinghua Wu, Haoyang Wang, Junying Miao, Baoxiang Zhao. Novel pyrazoline-based fluorescent probe for detecting glutathione and its application in cells. *Biosensors and Bioelectronics.* 55 (2014) 386-390.
- [9] Xiaofeng Yang, Yixing Guo, Robert M, Strongin. Conjugate addition/cyclization sequence enables selective and simultaneous fluorescence detection of cysteine and homocysteine. *Angew Chem Int Ed Engl.* 50 (2011) 10690-10693.
- [10] Xi Dai, Qing Hua Wu, Peng-Chong Wang, Jie Tian, Yu Xu, Sheng Qing Wang, Jun Ying Miao, Baoxiang Zhao. A simple and effective coumarin-based fluorescent probe for cysteine, *Biosensors and Bioelectronics* 59 (2014) 35–39.
- [11] Qiong Zhang, Dehuan Yu, Shuangshuang Ding and Guoqiang Feng. A colorimetric and red emissive fluorescent probe for cysteine and its application in bioimaging. *50* (2014) 14002-14005.