

Supporting Information

A new indoloquinoline-based fluorescence probe for imaging of hydrazine in vitro and in vivo

Caiju Zhang^{a, 1}, Shi Tang^{a, b, 1}, Jianyong Wu^a, wenxing Lv^c, Junjian Li^{b*}, Chu Tang^{d, c*}

a. Department of Radiology, Hainan Affiliated Hospital of Hainan Medical University (Hainan General Hospital), Haikou 570311, P. R. China

b. Key Laboratory of Tropical Biological Resources of Ministry of Education, School of Pharmaceutical Sciences, Hainan University, Haikou 570228, P. R. China

c. Hangzhou Institute of Advanced Technology Chinese Academy of Sciences, Hangzhou, 310024, P. R. China

d. Engineering Research Center of Molecular and Neuro Imaging, Ministry of Education, School of Life Science and Technology, Xidian University, Xi'an, Shaanxi 710126, China

***Corresponding author:**

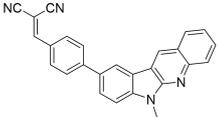
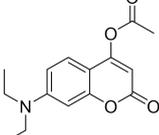
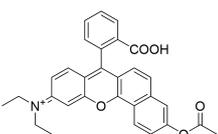
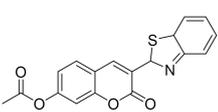
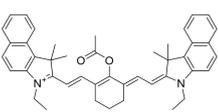
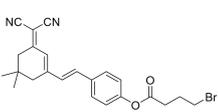
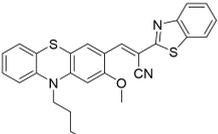
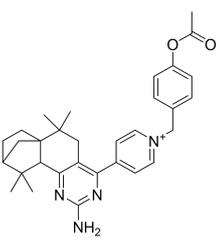
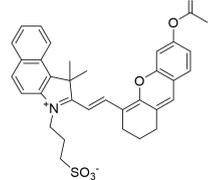
Junjian Li, E-mail: lijunjian@hainanu.edu.cn

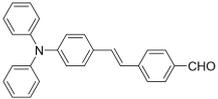
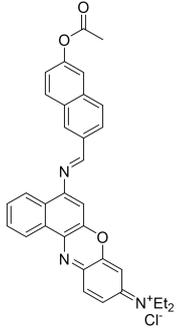
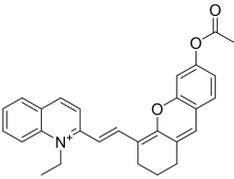
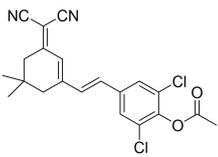
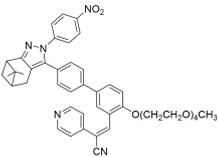
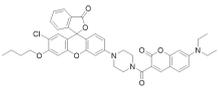
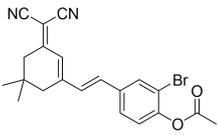
Chu Tang, E-mail: ctang@xidian.edu.cn

Table of Content

	Content	Page
1	Table S1	S3
2	Figure S1	S5
3	Figure S2	S5
4	Figure S3	S6
5	Figure S4	S6
6	Figure S5	S7
7	Figure S6	S7
8	Figure S7	S8
9	Figure S8	S8
10	NMR of Compounds	S8
11	References	S10

Table S1. Comparison of probe **1** with some hydrazine-detecting fluorescent probes

Name	Structure	Absorption/ emission (nm)	Type	LOD (nM)	Time (min)	Application	Ref.
1		410/615	Turn-off	680	< 2	MCF-7 tumor	The present word
6		382/470	Ratio	31000	7	HeLa cells	1
Rho-N₂H₄		575/645	Turn-on	365	40	4T1 cells, normal mice	2
Cou-1-N₂H₄		376/496	Turn-on	11.9	3	HeLa cells	3
Cy-OAc		730/825	Ratio	~15	~60	SMMC-772 1 cells, HepG2 cells kidney injury mice	4
DDPB		400/495	Turn-on	86.3	20	HeLa cells	5
PBT		440/690	Ratio	0.11	40	HepG2 cells	6
PBQ-AB		373/595	Ratio	48	< 1	HeLa cells	7
CyOE		606/705	Turn-on	82	15	HepG2 cells	8

TPA-CHO		~400/612		240	20	HeLa cells	9
BPN		660/725	Turn-on	0.45	30	MDA-MB-231 cells	10
QLN-Hy		588/692	Turn-on	250	12	MCF-7 cells, normal mice	11
DCPBCl₂-Hz		388/490	Ratio	300	30	HepG2 cells	12
TOPIB-PAN		401/520	Turn-off	74.5	24	HeLa cells	13
RC-<i>n</i>-Bu		413/470	Ratio	98	20	HeLa cells	14
HB		390/675	Turn-on	31	12	HeLa cells, normal mice	15

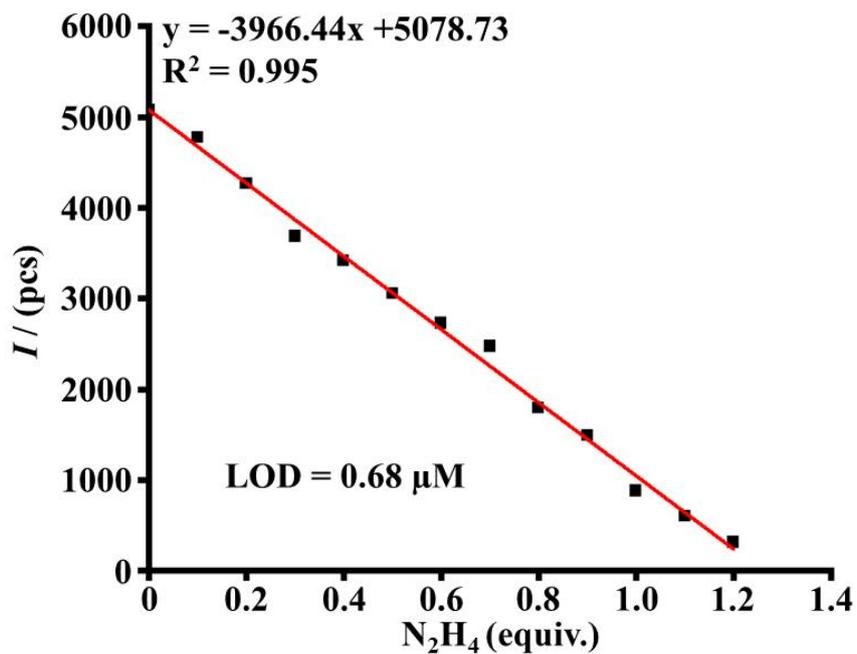


Figure S1. A linear relationship between fluorescence intensity at 615 nm and the equivalent of N₂H₄ for probe 1 (incubation time = 2 min at 25 °C).

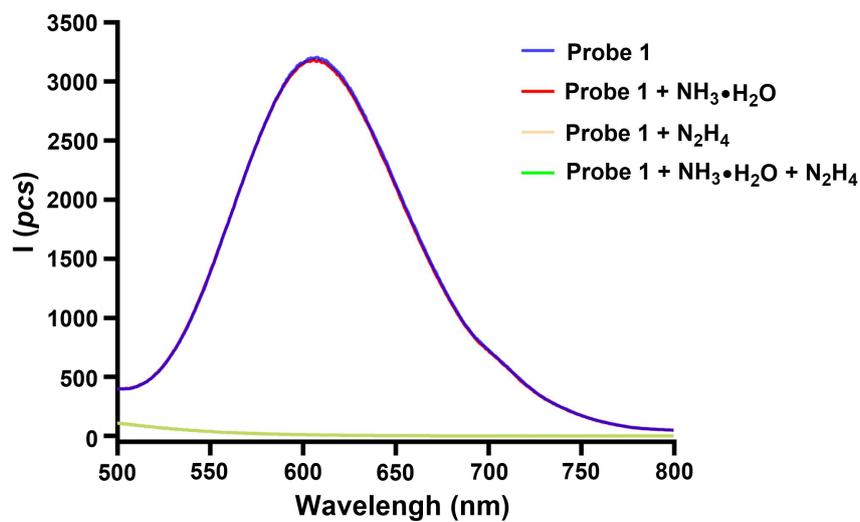


Figure S2. The fluorescence spectra of probe 1 (10 μM) after treatment with NH₃·H₂O (10 equiv.) or NH₃·H₂O + N₂H₄ (1.5 equiv.).

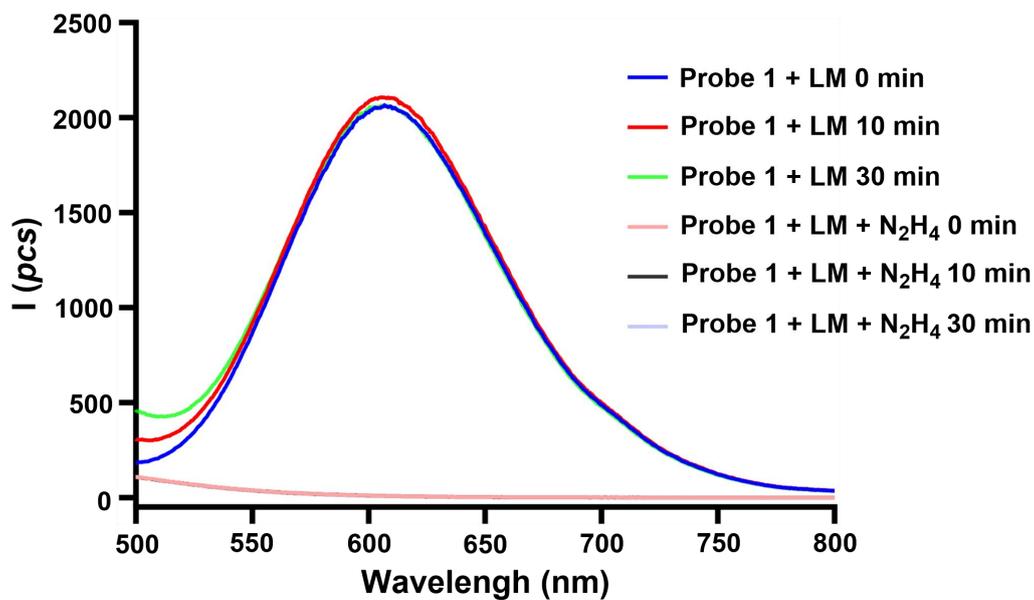


Figure S3. The fluorescence spectra of probe 1 (10 μ M) after treatment with rat liver microsomes (LM) (10 equiv.) or LM + N_2H_4 (1.5 equiv.) at 0, 10 and 30 min.

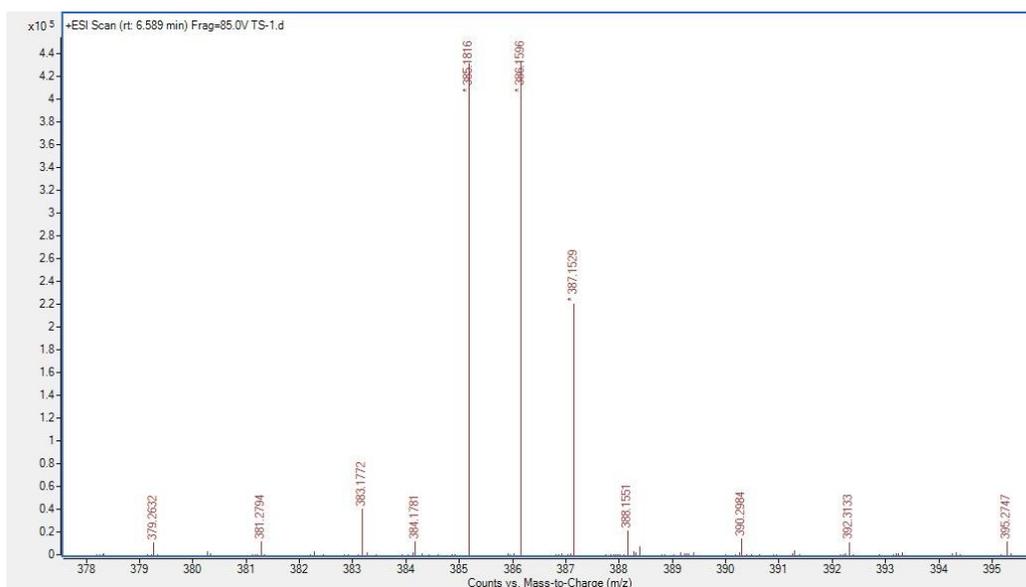


Figure S4. The HRMS spectra of probe 1

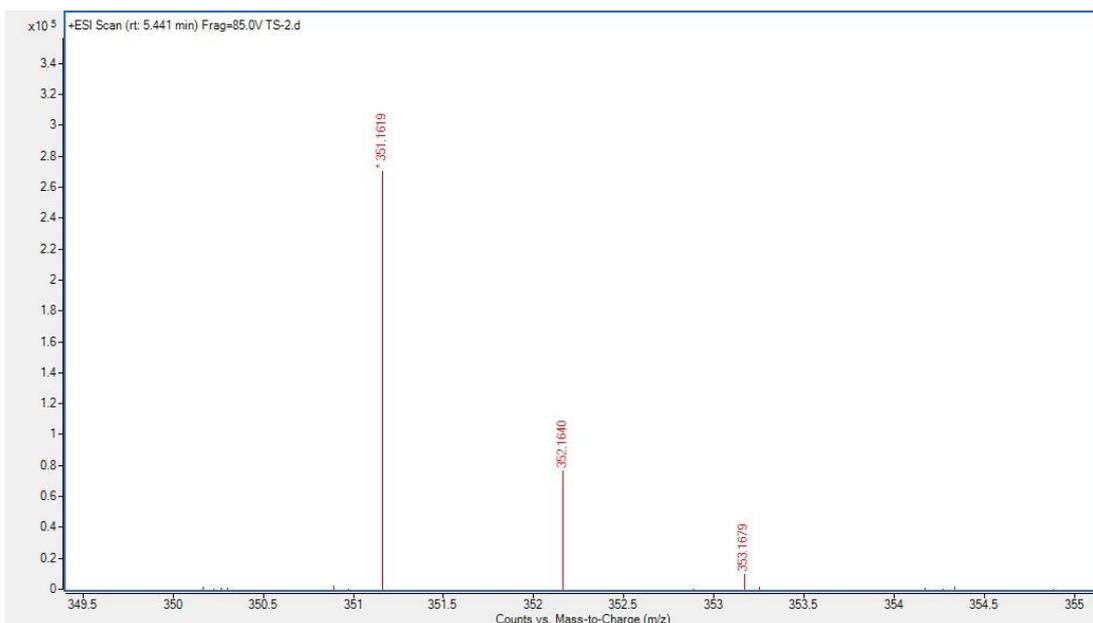


Fig. S5 The HRMS spectra of probe **1** after reacting with N₂H₄

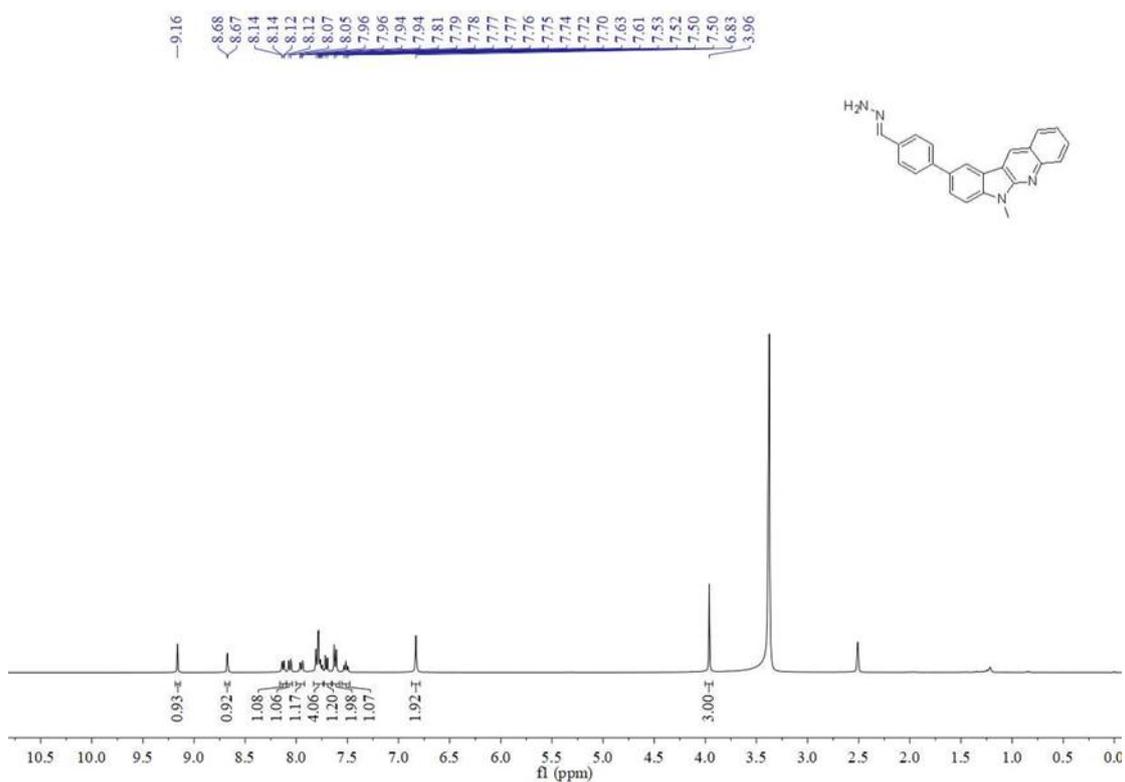


Fig. S6 The ¹H NMR spectra of probe **1** after reacting with N₂H₄

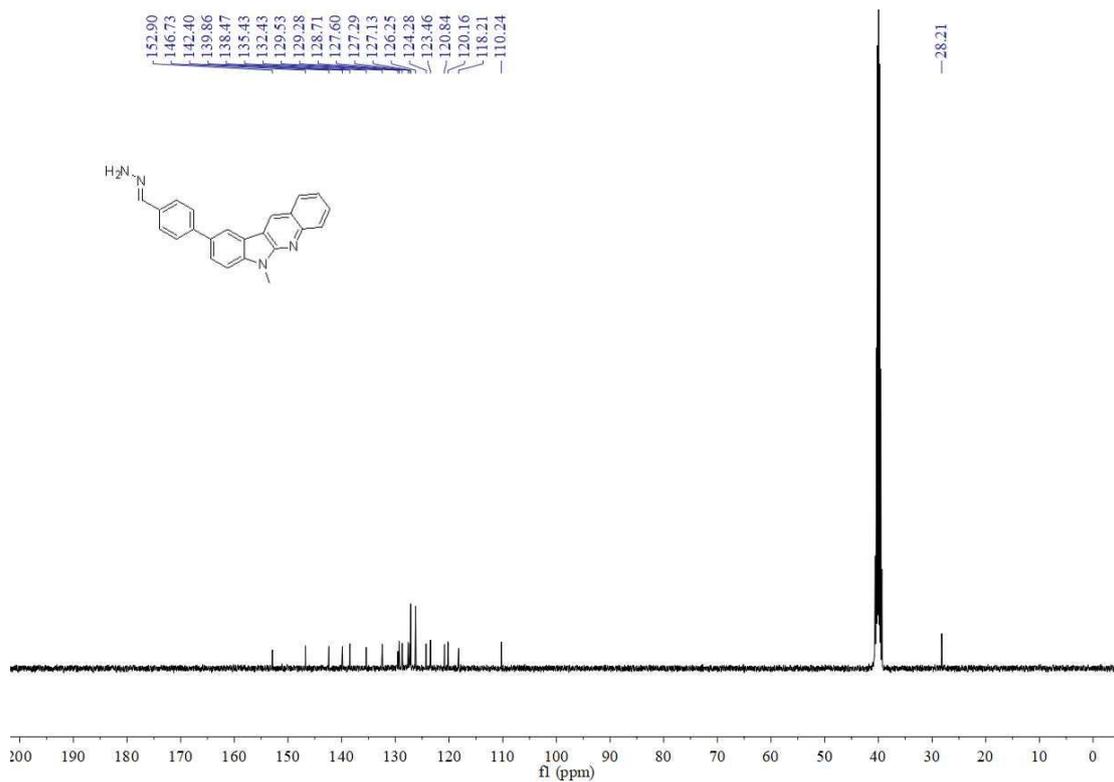


Fig. S7 The ¹³C NMR spectra of probe 1 after reacting with N₂H₄

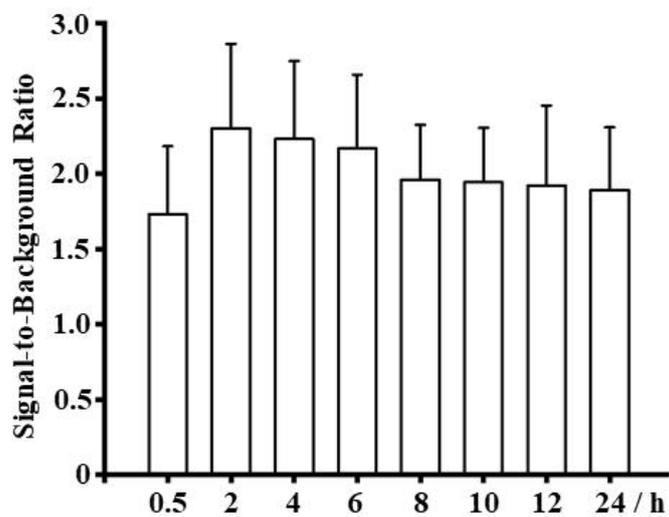
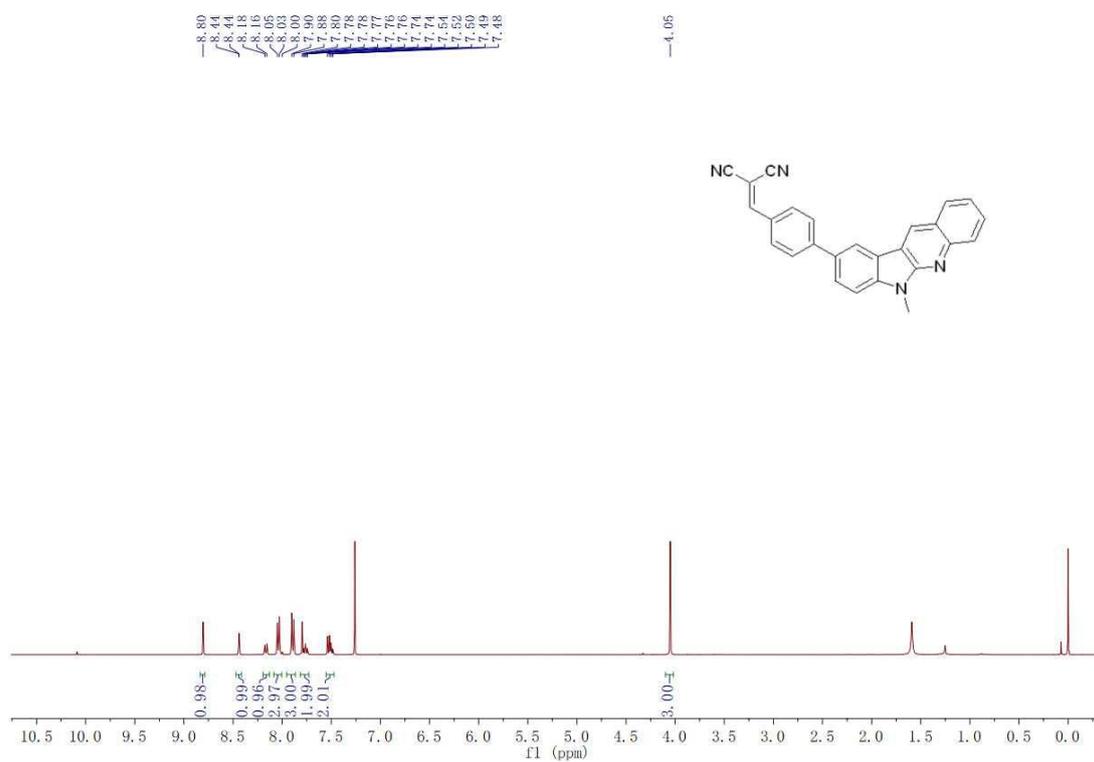
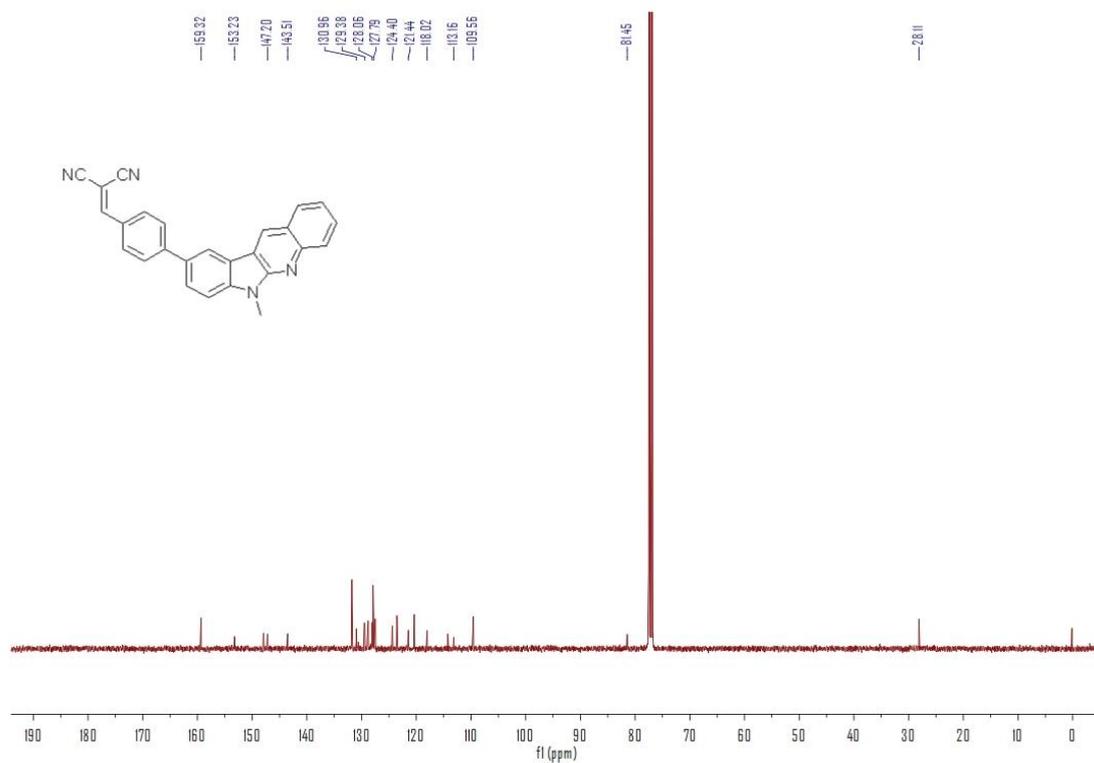


Fig. S8 Regions of interest (ROI) analysis of signal-to-background ratios.

The ^1H NMR spectra of probe 1



The ^{13}C NMR spectra of probe 1



References

- [1] X. R. Shi, F. J. Huo, J. B. Chao and C. X. Yin, A ratiometric fluorescent probe for hydrazine based on novel cyclization mechanism and its application in living cells, *Sensor. Actuat. B: Chem.*, 2018, **260**, 609-616
- [2] X. Q. Kong, B. L. Dong, C. Wang, N. Zhang, W. H. Song and W. Y. Lin, A novel mitochondria-targeted fluorescent probe for imaging hydrazine in living cells, tissues and animals, *J. Photoch. Photobiolo. A.*, 2018, **356**, 321-328
- [3] S. H. Guo, Z. Q. Guo, C. Y. Wang, Y. j. Shen and W. H. Zhu, An ultrasensitive fluorescent probe for hydrazine detection and its application in water samples and living cells, *Tetrahedron* 2019, **75**, 2642-2646
- [4] Y. Q. Song, G. Chen, X. Y. Han, J. M. You and F. B. Yu, A highly sensitive near-infrared ratiometric fluorescent probe for imaging of mitochondrial hydrazine in cells and in mice models, *Sensor. Actuat. B: Chem.*, 2019, **286**, 69-76.
- [5] M. Q. Zhu, Y. M. Xu, L. F. Sang, Z. Y. Zhao, L. J. Wang, X. Q. Wu, F. G. Fan, Y. Wang, H. Li, An ICT-based fluorescent probe with a large Stokes shift for measuring hydrazine in biological and water samples, *Environ. Pollut.*, 2020, **256**, 113427
- [6] T. G. Zhang, L. L. Zhu and W. Y. Lin, A near infrared ratiometric fluorescent probe with aggregation induced emission (AIE) characteristics for hydrazine detection in vitro and in vivo, *Dyes Pigm.*, 2021, **188**, 109177
- [7] Z. L. Wang, Y. Zhang, Z. Y. Meng, M. X. Li, C. L. Zhang, L. J. Yang, Y. Q. Yang, X. Xu and S. F. Wang, Development of a ratiometric fluorescent probe with large Stokes shift and emission wavelength shift for real-time tracking of hydrazine and its multiple applications in environmental analysis and biological imaging, *J. Hazard. Mater.*, 2022, **422**, 126891.
- [8] Y. Wang, X. L. Xue, Q. Zhang, K. P. Wang, S. j. Chen, L. S. Tang and Z. Q. Hu, A hemicyanine-based near-infrared fluorescent probe for vapor-phase hydrazine detection and bioimaging in a complete aqueous media, *Spectrochim. Acta A Mol. Biomol. Spectrosc.*, 2022, **279**, 121406.
- [9] Y. Y. Han, Y. Huang, Q. W. Lin, L. Y. Tang, G. Y. Yang, H. T. Xin, S. F. Zhao, R. F. Guan, K. N. Wang and D. X. Cao, Bifunctional fluorescent probe for the recognition of hydrazine and

- bisulfite in lipid droplets, *Sensor. Actuat. B: Chem.*, 2023, 393, 134181.
- [10] A. Maiti, D. Banik, S. Halder, S. K. Manna, A. Karak, K. Janab, and A. K. Mahapatra, Near-infrared fluorescent turn-on probe for hydrazine detection: environmental samples and live cell imaging, *Org. Biomol. Chem.*, 2023, **21**, 6046-6056
- [11] B. L. Cao, H. Li, Z. J. Wu, J. Yin, W. Z. Wang and C. L. Li, A near-infrared fluorescent off-on probe with large Stokes shift for sensing hydrazine in vitro and in vivo, *Tetrahedron* 2023, **147**, 133665.
- [12] L. J. Luo, J. Y. Cheng, S. Chen, P. S. Zhang, S. Chen, Z. L. Tang, R. J. Zeng, M. T. Xu and Y. Q. Hao, A near-infrared ratiometric fluorescent probe for hydrazine and its application for gaseous sensing and cell imaging, *Spectrochim. Acta A Mol. Biomol. Spectrosc.*, 2023, **296**, 122692.
- [13] S. Zhang, J. X. Tian, S. Gong, Z. Y. Meng, Y. Y. Liang, Y. Gu, M. R. Wang, Z. L. Wang and S. F. Wang, A novel biphenyl tetrahydroindazole-type fluorescent probe for the detection of hydrazine and its applications in food detection, environmental analysis and biological imaging, *Spectrochim. Acta A Mol. Biomol. Spectrosc.*, 2026, **347**, 126966.
- [14] S. Q. Tang, H. D. Wu and Y. F. Han, A FRET-based ratiometric fluorescent probe for selective hydrazine detection, live imaging, and portable sensing, *Spectrochim. Acta A Mol. Biomol. Spectrosc.*, 2026, **351**, 127479.
- [15] H. C. Xia, Y. H. Zhang, L. M. Lv, D. Han, H. K. Yang, H. J. Yan, J. L. Lv, J. T. Lin and Y. Y. Kong, Dicyanoisophorone-based near-infrared fluorescent probe with a large Stokes shift for the detection and bioimaging of hydrazine, *J. Hazard. Mater.*, 2026, **502**, 141061.