

A highly sensitive fluorescent probe for selective detection of cysteine/homocysteine from glutathione and its application in living cells and tissues

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Contents

Fig. S1 ¹ H-NMR of NIPY-OH	S3
Fig. S2 HRMS spectrum of reaction products of the probe NIPY-NBD with Cys.	S4
Fig. S3 HRMS spectrum of reaction products of the probe NIPY-NBD with Hcy.	S5
Fig. S4 HRMS spectrum of reaction products of the probe NIPY-NBD with GSH.	S6
Fig. S5 ¹ H-NMR of the probe NIPY-NBD	S8
Fig. S6 ¹³ C-NMR of the probe NIPY-NBD	S9
Fig. S7 HRMS spectrum of the probe NIPY-NBD	S10
Fig. S8 The response time fluorescence map of the probe NIPY-NBD	S11
Table. S1 Comparison of fluorescent probes for Cys/Hcy from GSH with NBD group.	S12

References.....S17

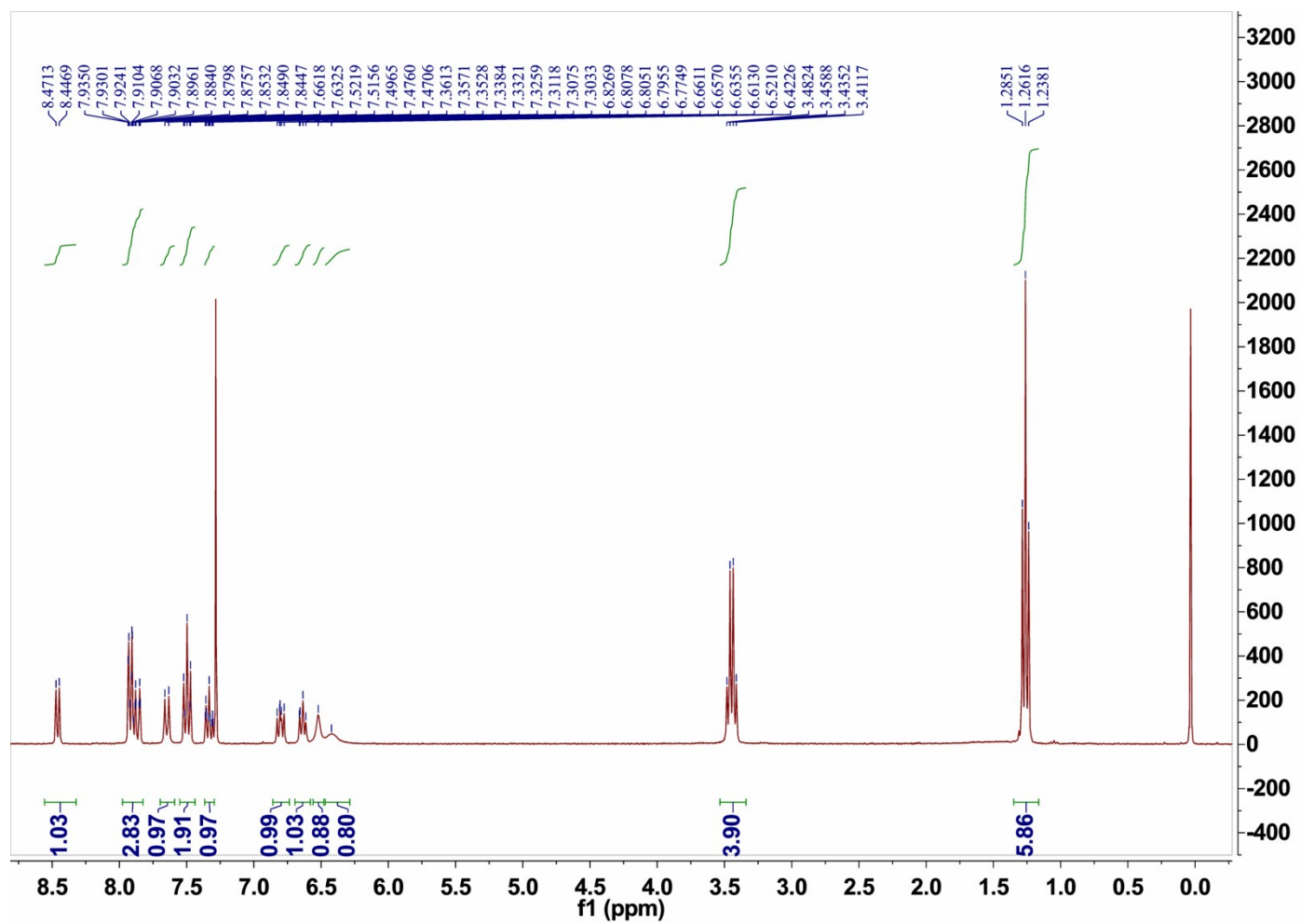


Fig. S1 ¹H-NMR of NIPY-OH

Sample Name		Position	p2f2	Instrument Name	Instrument 1
User Name	QTOF-PC/QTOF	Inj Vol	0.1	InjPosition	
Sample Type	Sample	IRM Calibration Status	Success	Data Filename	IP-CY-p.d
ACQ Method	20110418-MSonly-p.m	Comment		Acquired Time	5/17/2018 12:39:55 PM

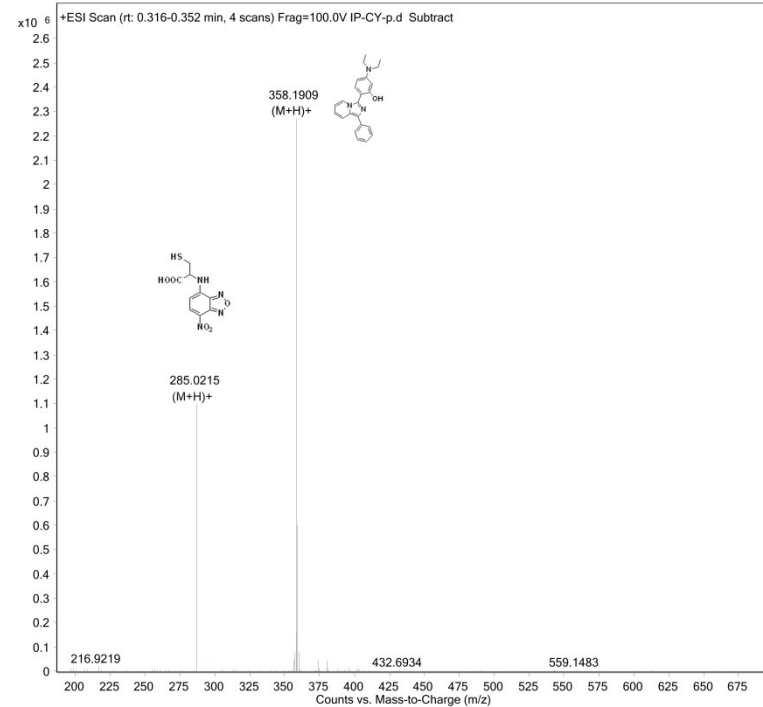


Fig. S2 HRMS spectrum of reaction products of the probe **NIPY-NBD** with Cys.

Sample Name		Position	P1f3	Instrument Name	Instrument 1	User Name	QTOF-PC\QTOF
Inj Vol	1	InjPosition		SampleType	Sample	IRM Calibration Status	Success
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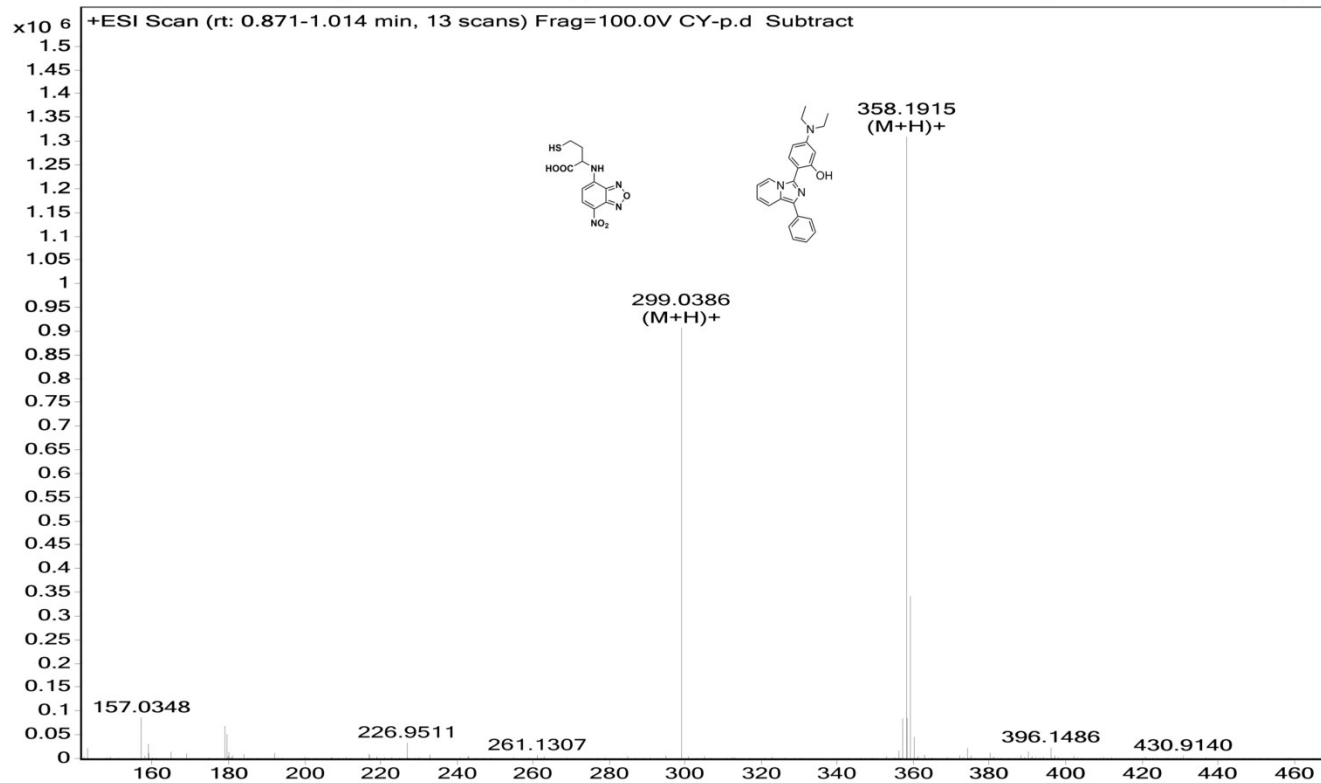


Fig. S3 HRMS spectrum of reaction products of the probe **NIPY-NBD** with Hey.

Sample Name		Position	p2f3	Instrument Name	Instrument 1
User Name	QTOF-PC\QTOF	Inj Vol	0.1	InjPosition	
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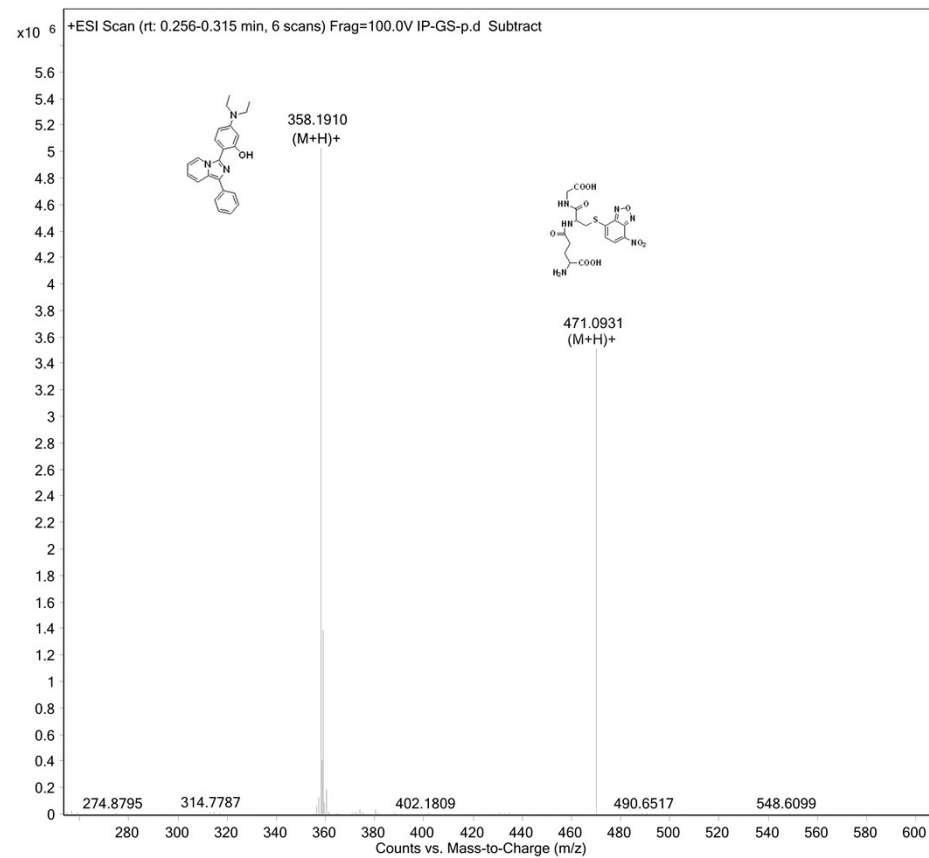
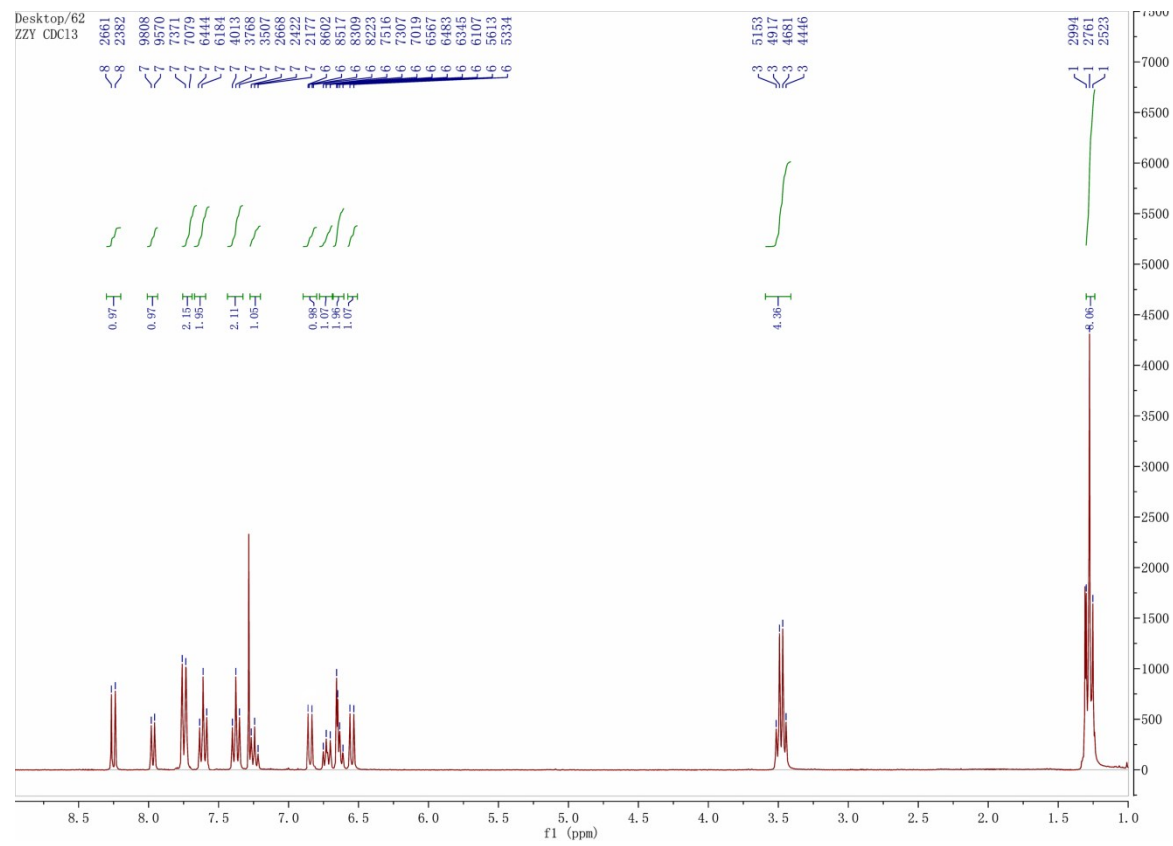


Fig. S4 HRMS spectrum of reaction products of the probe **NIPY-NBD** with GSH.



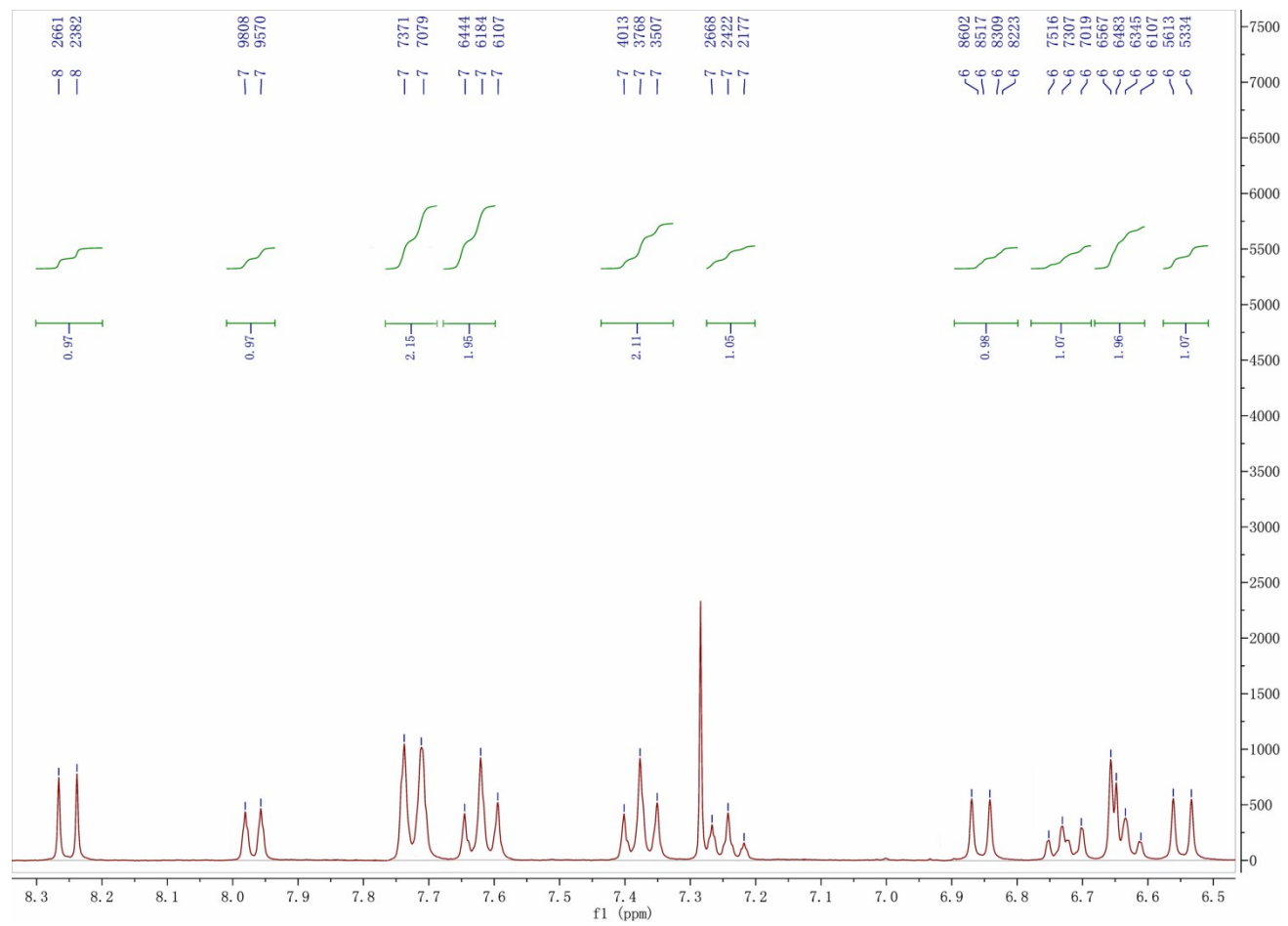


Fig. S5 ^1H -NMR of the probe NIPY-NBD.

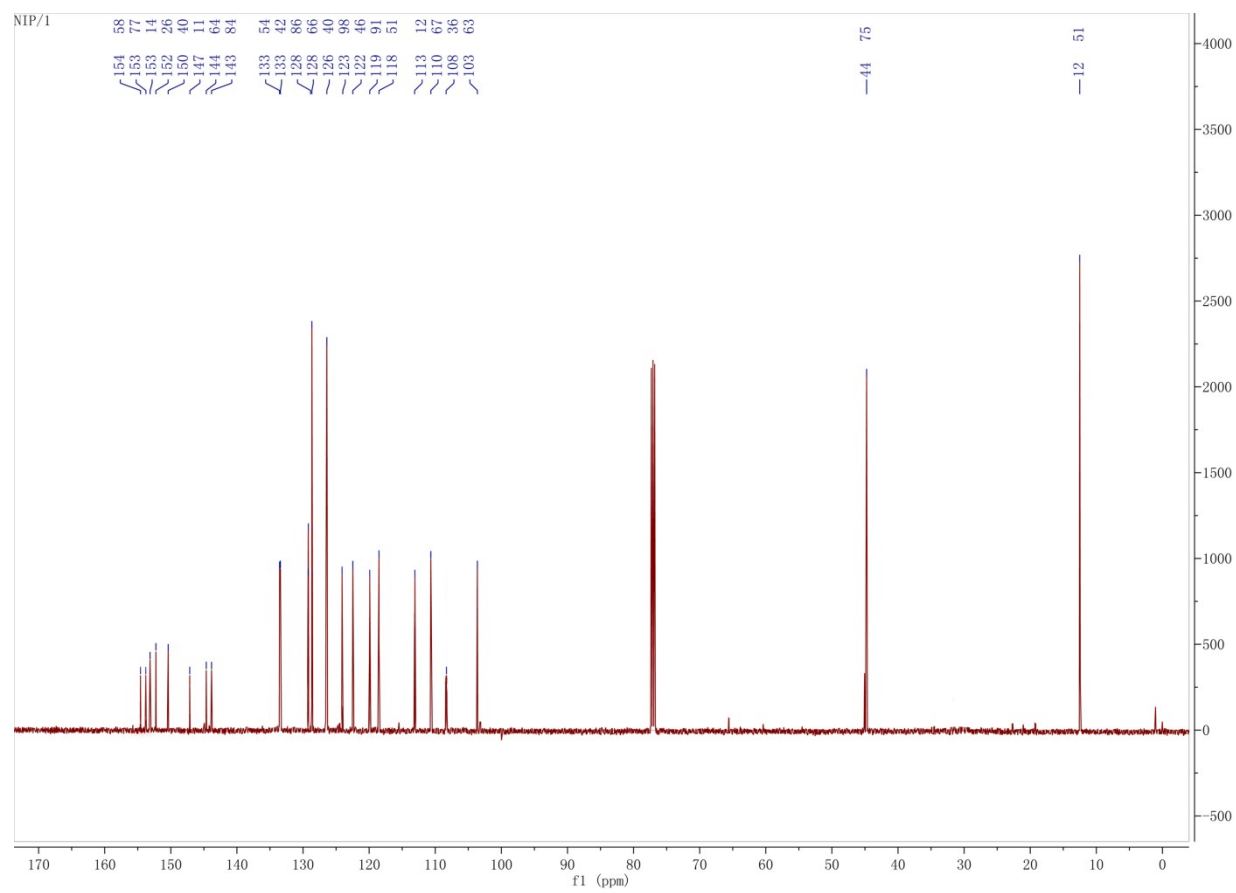


Fig. S6 ^{13}C -NMR of the probe **NIPY-NBD**.

Sample Name		Position	p2f1	Instrument Name	Instrument 1
User Name	QTOF-PCIQTOF	Inj Vol	0.1	InjPosition	
Sample Type	Sample	IRM Calibration Status	Success	Data Filename	IP-N-p.d
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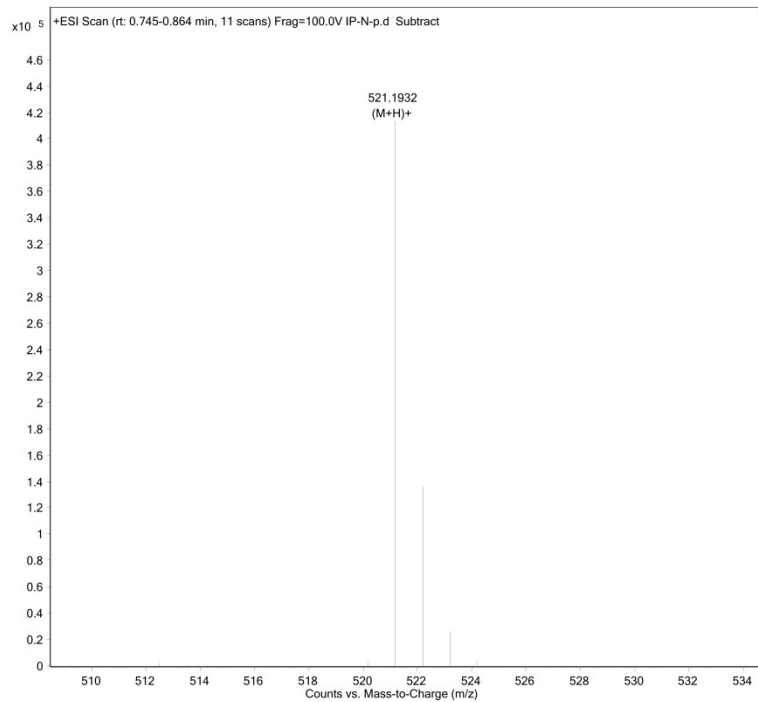


Fig. S7 HRMS spectrum of the probe **NIPY-NBD**.

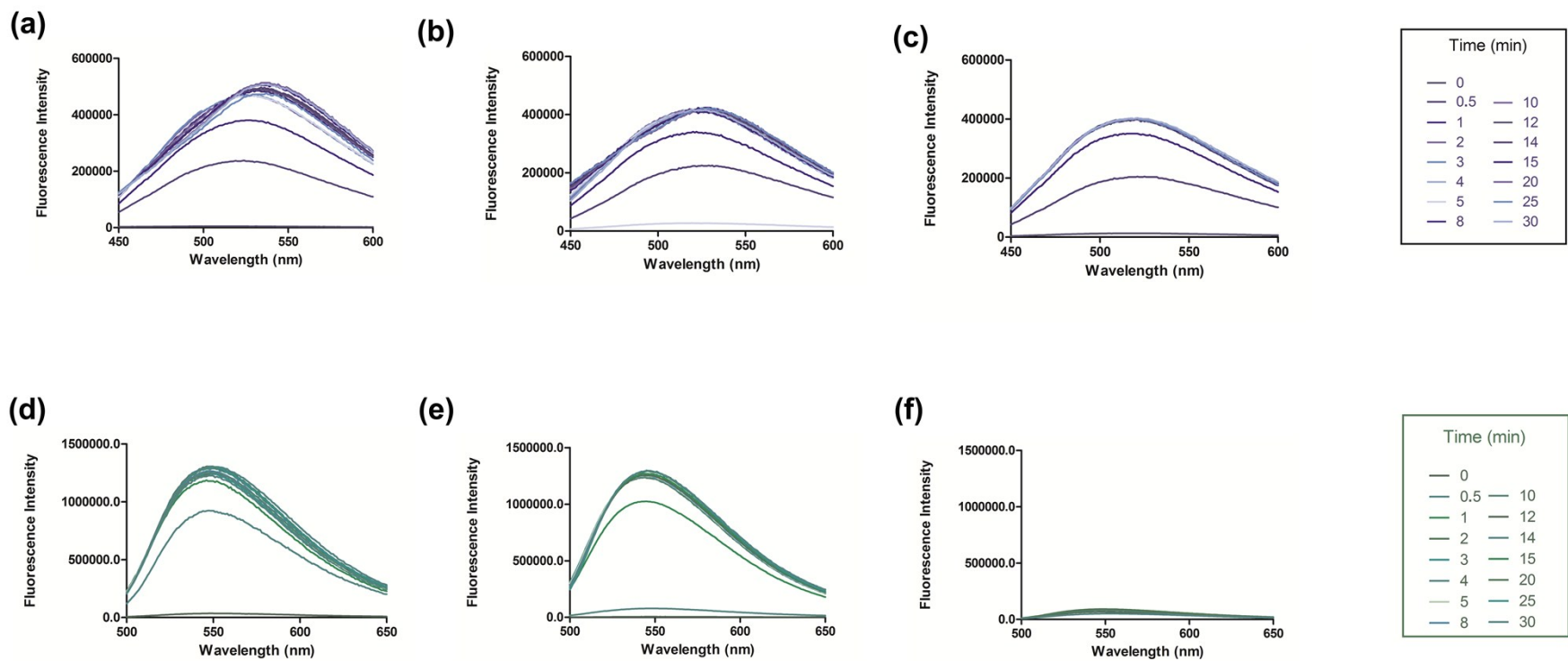
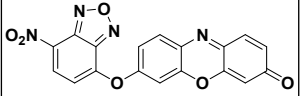
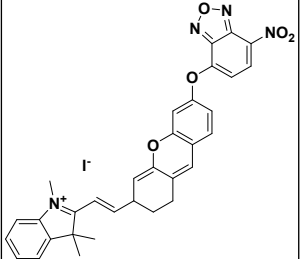
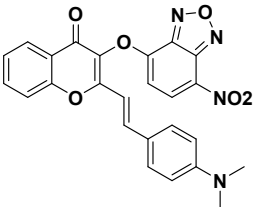
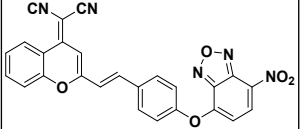
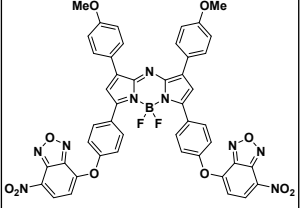
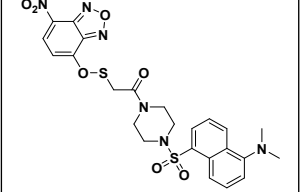


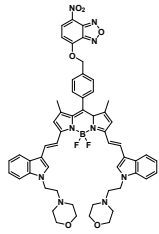
Fig. S8 The response time fluorescence map of the probe **NIPY-NBD** (10 μM) with time after adding 20 μM Cys/Hcy and GSH in PBS-DMSO buffer (v/v = 1/1). (a,d: adding Cys, b,e: adding Hcy, c,f: adding GSH, a-c: $\lambda_{\text{ex/em}} = 310 / 520 \text{ nm}$, d-f: $\lambda_{\text{ex/em}} = 470 / 550 \text{ nm}$).

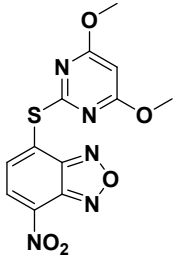
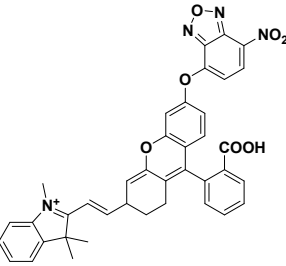
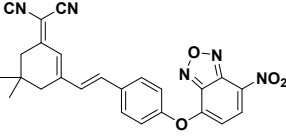
Table. S1 Comparison of fluorescent probes for Cys/Hcy from GSH with NBD group.

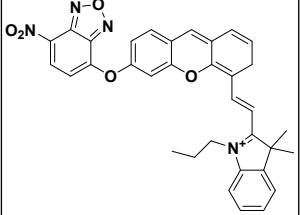
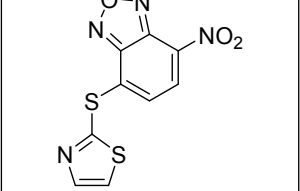
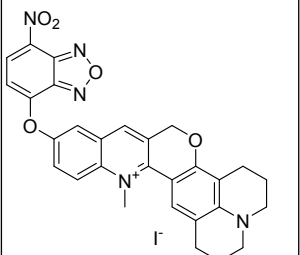
Probes	Fluorescence enhancement (at the concentration of Cys)	Response time/Limit of detection	Spectral separation Probe between two emissions/Stokes Shift	Reference
 <p><i>Chem. Commun</i>, 2015</p>	81 fold (I403/I519) at 320 μ M	10 min; For Cys and GSH - 130 and 70 nM, respectively	$\lambda_{\text{ex1}}=470$ nm ; $\lambda_{\text{em1}}=540$ nm $\lambda_{\text{ex2}}=470$ nm ; $\lambda_{\text{em2}}=585$ nm 70 & 115 nm	1
 <p><i>Biosens. Bioelectron</i> , 2016</p>	100 fold at 100 μ M	5 min /20 min; For Cys, Hcy and GSH - 27, 25 and 16 nM, respectively	$\lambda_{\text{ex1}}=470$ nm ; $\lambda_{\text{em1}}=550$ nm $\lambda_{\text{ex2}}=670$ nm ; $\lambda_{\text{em2}}=716$ nm 80 & 46 nm	2

 <p><i>Anal. Chem.</i> 2016</p>	70 fold at 30 μ M	<p>120 min</p> <p>For Cys, Hcy and GSH- 2100, 2700 and 6400 nM, respectively</p>	<p>$\lambda_{ex1}=650$ nm; $\lambda_{em1}=705$ nm;</p> <p>$\lambda_{ex2}=476$ nm; $\lambda_{em2}=545$ nm;</p> <p>55 & 79 nm</p>	3
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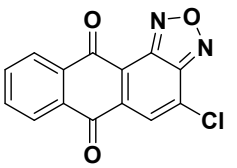
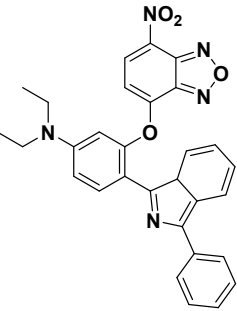
 <p><i>Sens. Actuators B-Chem</i>, 2017</p>	54 fold at 20 μ M	15 min; For Cys, Hcy and GSH - 21, 17 and 26 nM, respectively	$\lambda_{\text{ex1}}=475$ nm; $\lambda_{\text{em1}}=560$ nm $\lambda_{\text{ex2}}=560$ nm; $\lambda_{\text{em2}}=700$ nm 85 & 140 nm	4
 <p><i>Chem. Commun.</i> 2017</p>	30 fold at 10 mM	10 min; For Cys, Hcy and GSH - 80, 170 and 50 nM, respectively	$\lambda_{\text{ex1}}=470$ nm; $\lambda_{\text{em1}}=540$ nm; $\lambda_{\text{ex2}}=670$ nm; $\lambda_{\text{em2}}=730$ nm 70 & 60 nm	5
 <p><i>Tetrahedron</i>, 2017</p>	118 fold at 100 μ M	12 min; For Cys, Hcy – 35 and 26 nM	$\lambda_{\text{ex}}=470$ nm; $\lambda_{\text{em}}=550$ nm; 80 nm	6

 <p><i>J.Mater. Chem. B</i>, 2017</p>	<p>*</p>	<p>30 min; For Cys - 22 nM</p>	<p>$\lambda_{\text{ex1}}=466 \text{ nm}; \lambda_{\text{em1}}=540 \text{ nm};$ $\lambda_{\text{ex2}}=650 \text{ nm}; \lambda_{\text{em2}}=735 \text{ nm}$ 74 & 85 nm</p>	<p>7</p>
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 <p><i>Acta. A. Mol. Biomol. Spectrosc</i>, 2017</p>	78 fold at 10 μ M	10 min; For Cys and Hcy - 95 and 138 nM, respectively	λ_{ex} =419 nm; λ_{em} =547 nm; 128 nm	8
 <p><i>Anal. Chim. Acta</i>, 2017</p>	9.3 fold at 10 μ M	8 min; For Cys, Hcy and GSH- 21, 37 and 28 nM, respectively	λ_{ex1} =463 nm; λ_{em1} =544 nm; λ_{ex2} =650 nm; λ_{em2} =707 nm; 81 & 57 nm	9
 <p><i>Dyes Pigments</i>, 2017</p>	35 fold at 10 μ M	10 min For Cys, Hcy and GSH- 51, 16 and 34 nM, respectively	λ_{ex1} =440 nm; λ_{em1} =500 nm; λ_{ex2} =400 nm; λ_{em2} =443 nm; 60 & 43 nm	10

 <p><i>Sens. Actuators B-Chem</i>, 2017</p>	<p>50 fold at 30 μM</p>	<p>5 min For Cys and GSH- 68 and 81 nM</p>	<p>λ_{ex1}=480 nm; λ_{em1}=540 nm; λ_{ex2}=670 nm; λ_{em2}=702 nm; 60 & 32 nm</p>	<p>11</p>
 <p><i>Dyes Pigments</i>, 2018</p>	<p>103 fold at 100 μM</p>	<p>10 min; For Cys, Hcy - 176 and 124 nM</p>	<p>λ_{ex}=460 nm; λ_{em}=565 nm; 105 nm</p>	<p>12</p>
	<p>10 fold at 120 μM</p>	<p>15 min / 28 min; For Cys, Hcy and GSH –12, 13 and 6 nM, respectively</p>	<p>λ_{ex1}=502 nm; λ_{em1}=610 nm; λ_{ex2}=487 nm; λ_{em2}=547 nm 108 & 60 nm</p>	<p>13</p>

<i>Sens. Actuators B-Chem</i> , 2018				
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 <p><i>Sens. Actuators B Chem</i>, 2018</p>	<p>17 fold at 150 μM with only GSH</p>	<p>30 min; For GSH- 89 nM</p>	<p>λ_{ex}=465 nm; λ_{em}=558 nm; 93 nm</p>	<p>14</p>
	<p>100 fold (λ_{ex}/em = 310 nm /520 nm) & 210 fold (λ_{ex}/em = 340 nm /550 nm) at 20 μM</p>	<p>2.5 min; For Cys, Hcy and GSH- 21, 46 and 63 nM, respectively</p>	<p>λ_{ex1}=310 nm; λ_{em1}=520 nm; λ_{ex2}=470 nm; λ_{em2}=550 nm 210 & 80 nm</p>	<p>This work</p>

*The data was not mentioned.

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