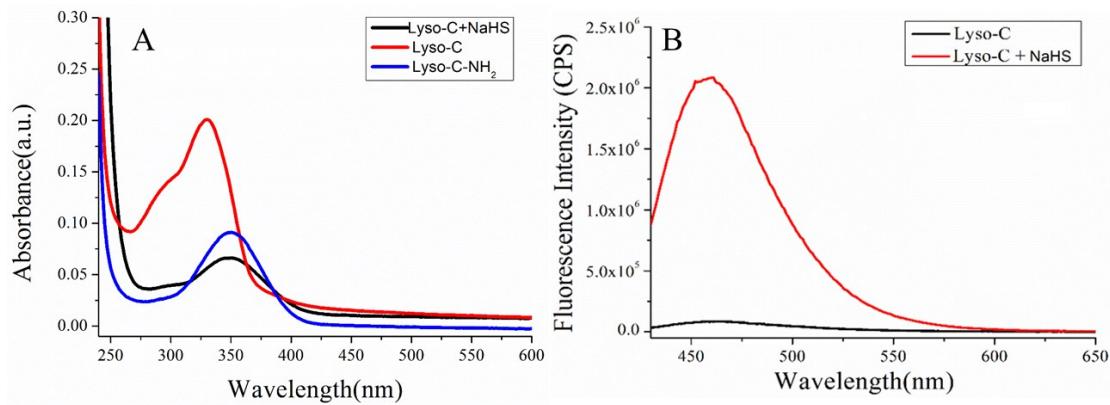


## Electronic Supplementary Information (ESI) for

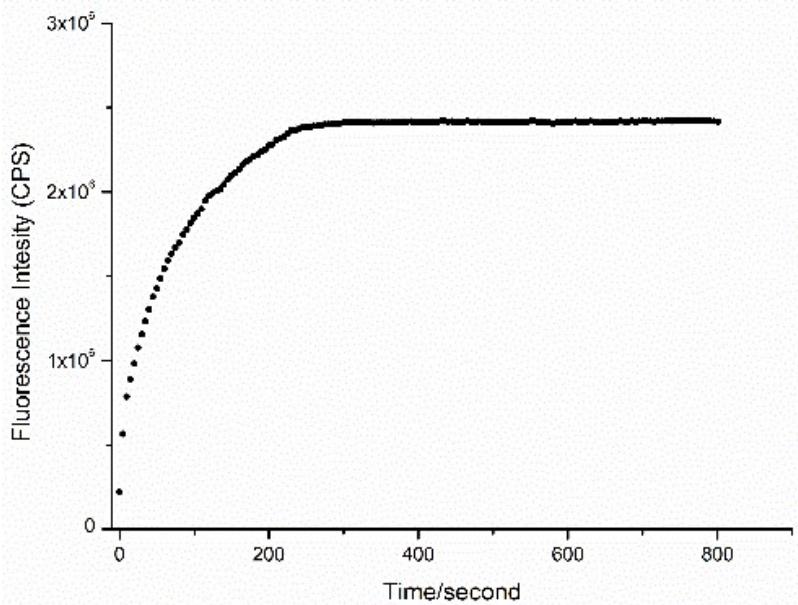
### **An azidocoumarin-based fluorescent probe for imaging lysosomal hydrogen sulfide in living cells**

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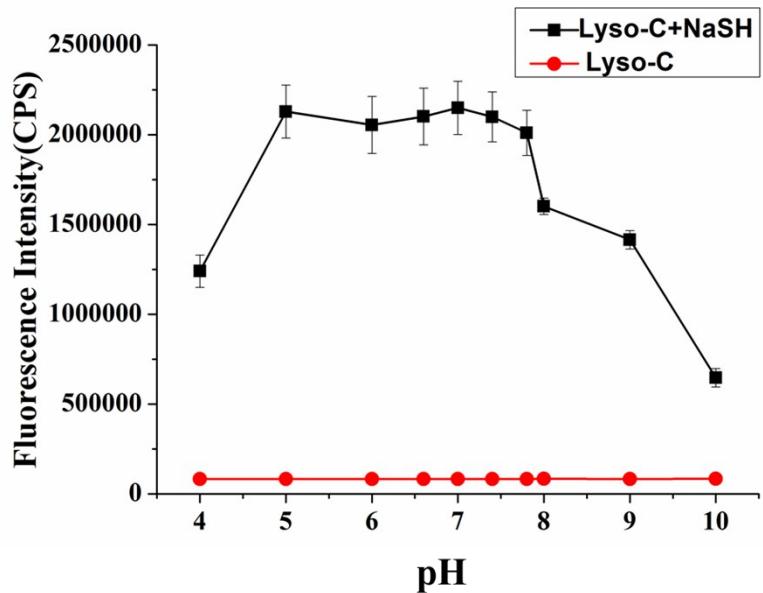
State Key Laboratory of Chemo/Biosensing & Chemometrics, Institute of Chemical Biology & Nanomedicine, College of Chemistry & Chemical Engineering, Hunan University, Changsha 410082, China



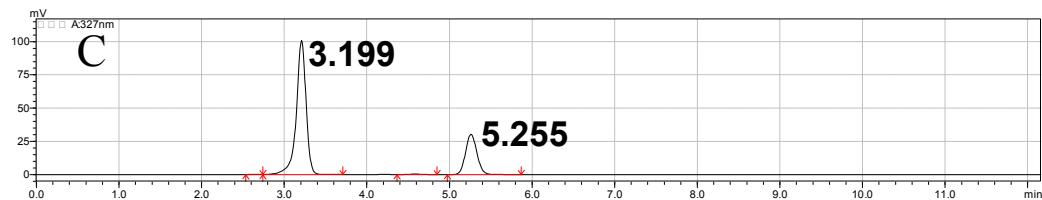
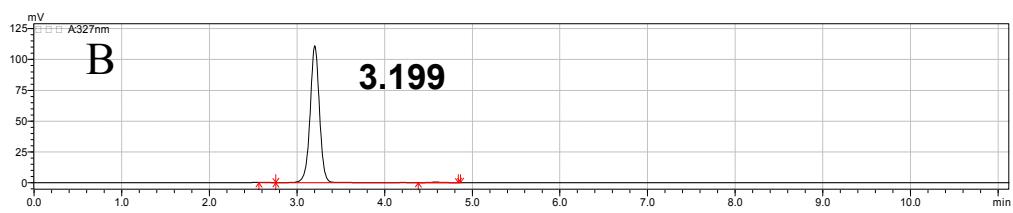
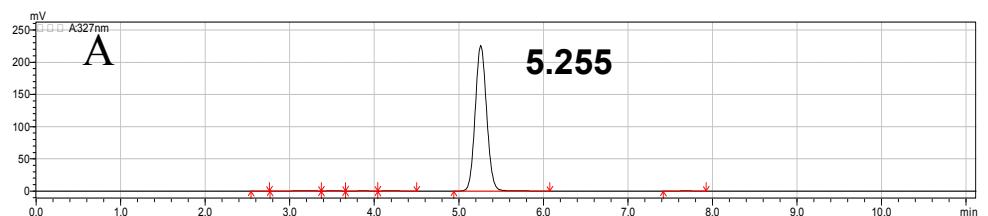
**Fig. S1** A) UV-vis response of probe Lyso-C toward H<sub>2</sub>S in aqueous solution. Red line: Lyso-C (10 μM); black line: the mixture of Lyso-C (10 μM) with NaHS (50 μM); blue line: reference sample of Lyso-C-NH<sub>2</sub> (10 μM); B) Fluorescence response of probe Lyso-C toward H<sub>2</sub>S in aqueous solution. Black line: Lyso-C (10 μM); red line: the mixture of Lyso-C (10 μM) with NaHS (50 μM).



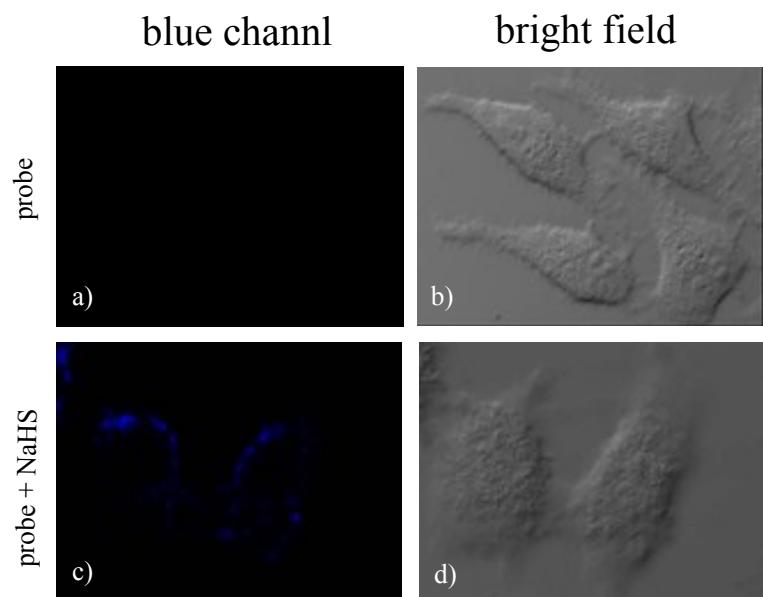
**Fig. S2** Time-dependent fluorescence intensity change of probe Lyso-C (10  $\mu\text{M}$ ) at 458 nm in response to NaHS (30  $\mu\text{M}$ ),  $\lambda_{\text{ex}} = 355 \text{ nm}$ .



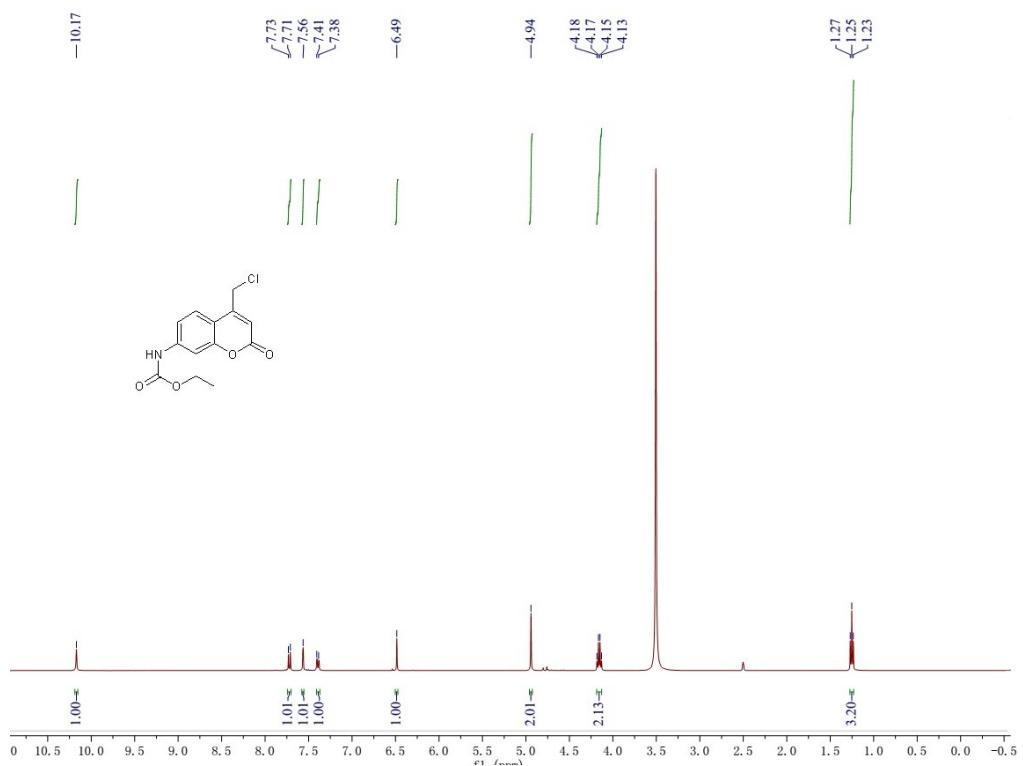
**Fig. S3** Effects of pH on the response of probe Lyso-C (10  $\mu\text{M}$ ) toward NaHS (30  $\mu\text{M}$ ),  $\lambda_{\text{ex}} = 355 \text{ nm}$ ,  $\lambda_{\text{em}} = 458 \text{ nm}$ .



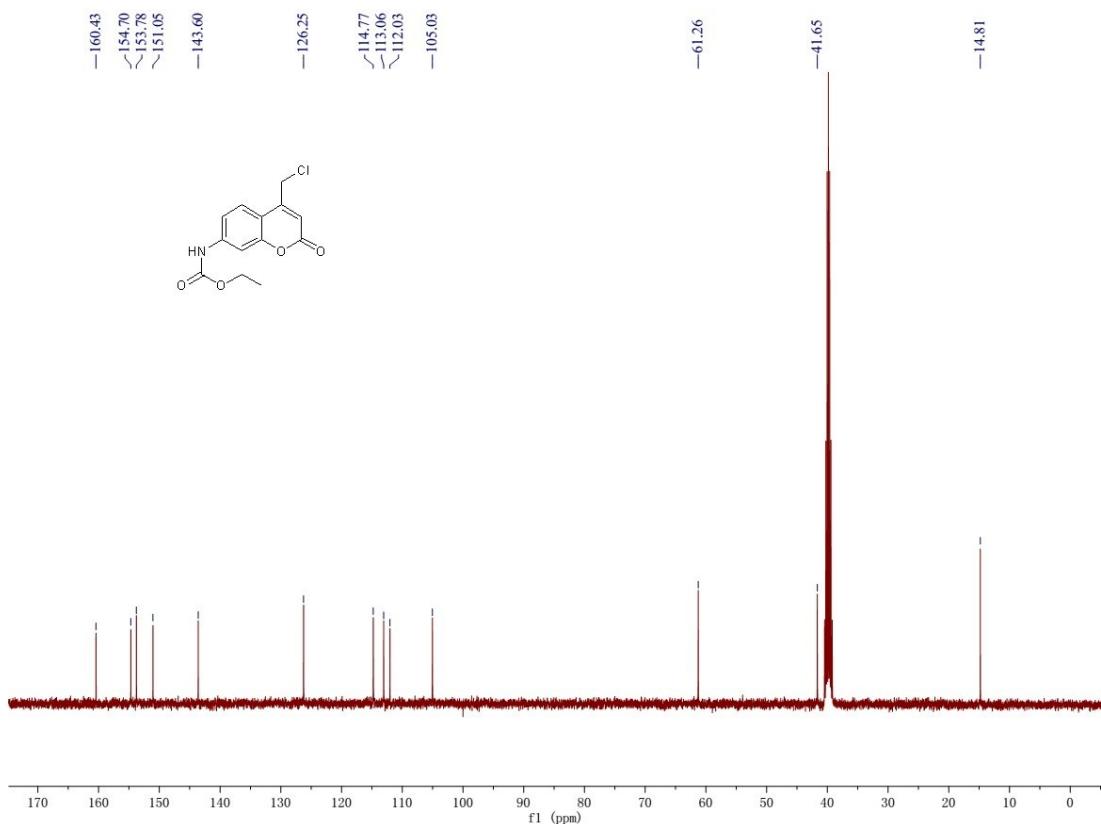
**Fig. S4** HPLC profiles: A) 100  $\mu\text{M}$  Lyso-C; B) 100  $\mu\text{M}$  Lyso-C-NH<sub>2</sub>; C) the reaction mixture of 100  $\mu\text{M}$  Lyso-C with 50  $\mu\text{M}$  NaHS. Detection: UV-vis (327 nm) detector. Flow rate: 1.0 mL/min. Temperature: 30 °C. Injection volume: 10  $\mu\text{L}$ . Mobile phase: MeOH/Water = 80/20 (v/v).



**Fig. S5** Fluorescence imaging of H<sub>2</sub>S in HepG-2 cells: (a) HepG-2 cells incubated with probe Lyso-C (10  $\mu$ M) for 30 min; (b) HepG-2 cells incubated with probe Lyso-C (10  $\mu$ M) for 30 min followed by incubation with NaHS (50  $\mu$ M) for another 30 min; (d) Bright-field image of HepG-2 cells in (c).

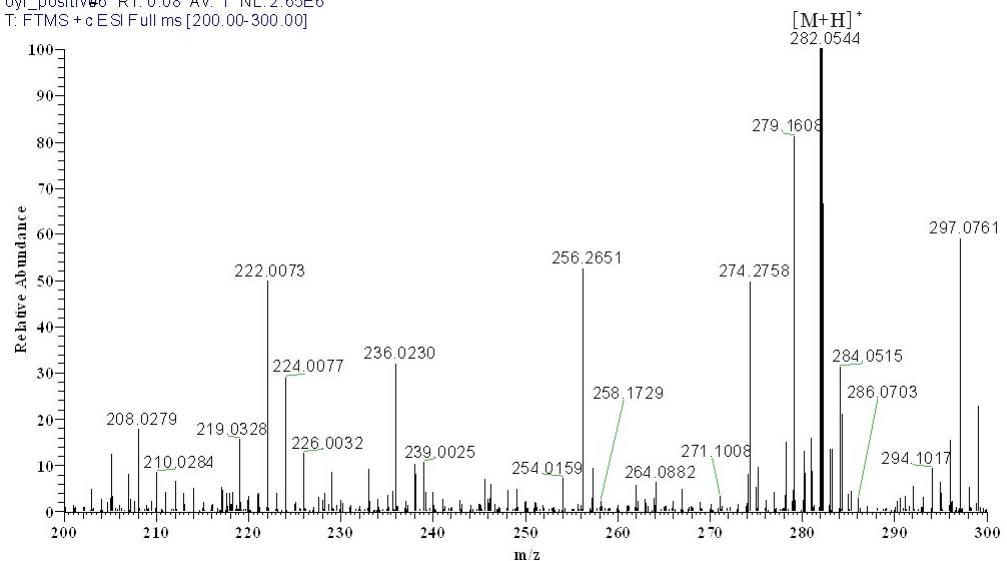


**Fig. S6**  $^1\text{H}$  NMR spectrum of compound **1** in  $\text{DMSO-d}_6$

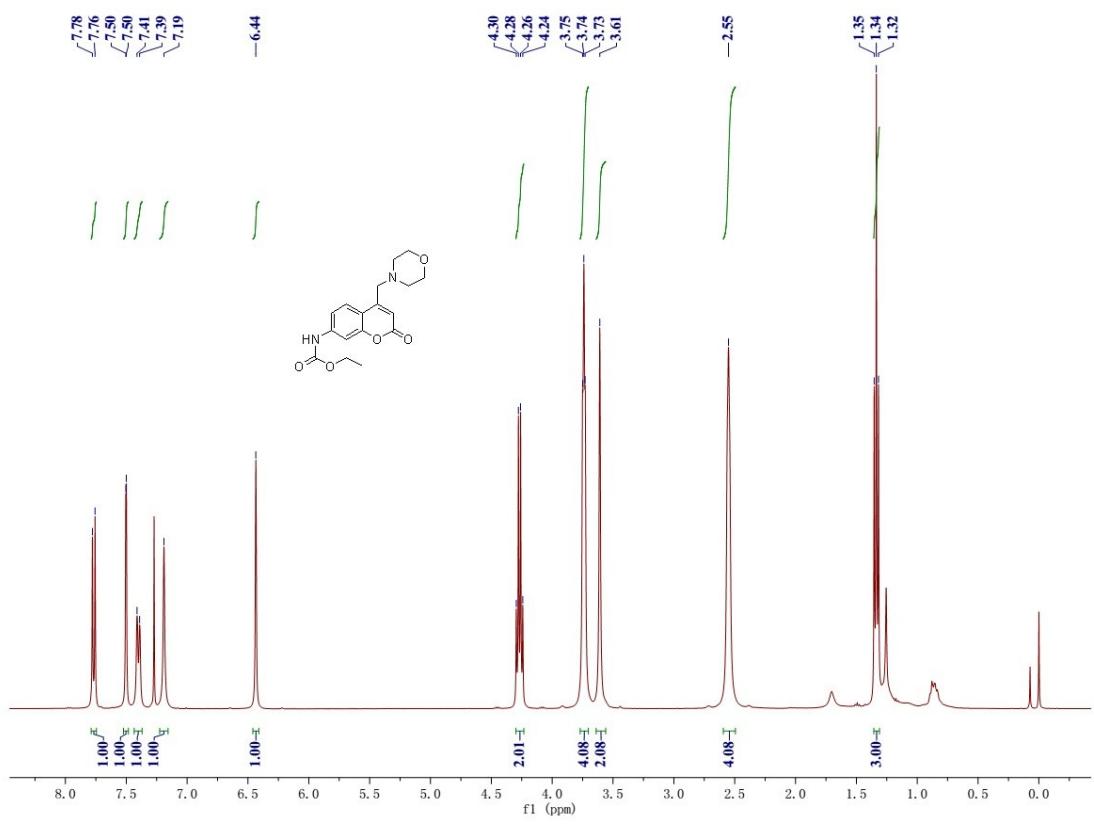


**Fig. S7**  $^{13}\text{C}$  NMR spectrum of compound **1** in  $\text{DMSO-d}_6$

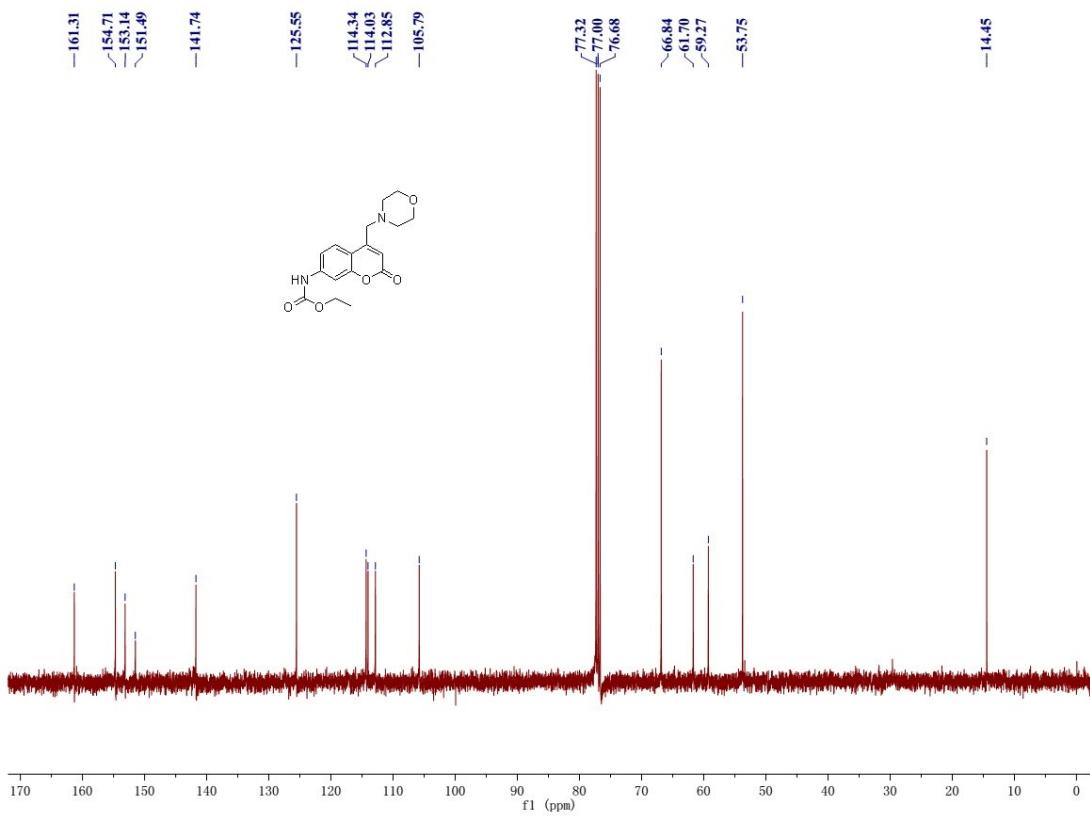
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T: FTMS + c ESI Full ms [200.00-300.00]



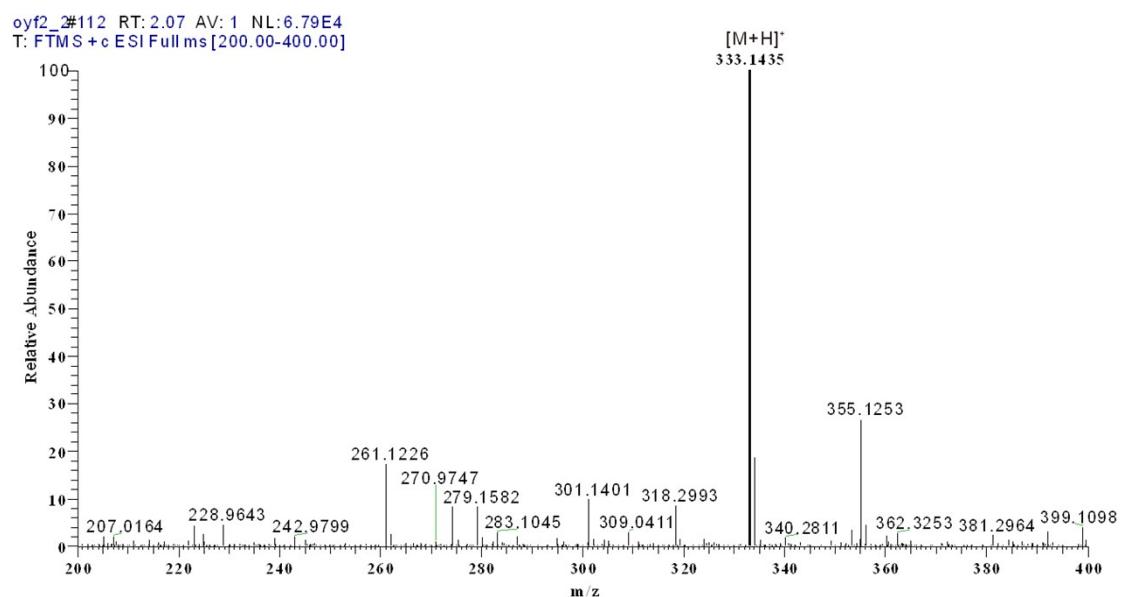
**Fig. S8** MS spectrum of compound 1



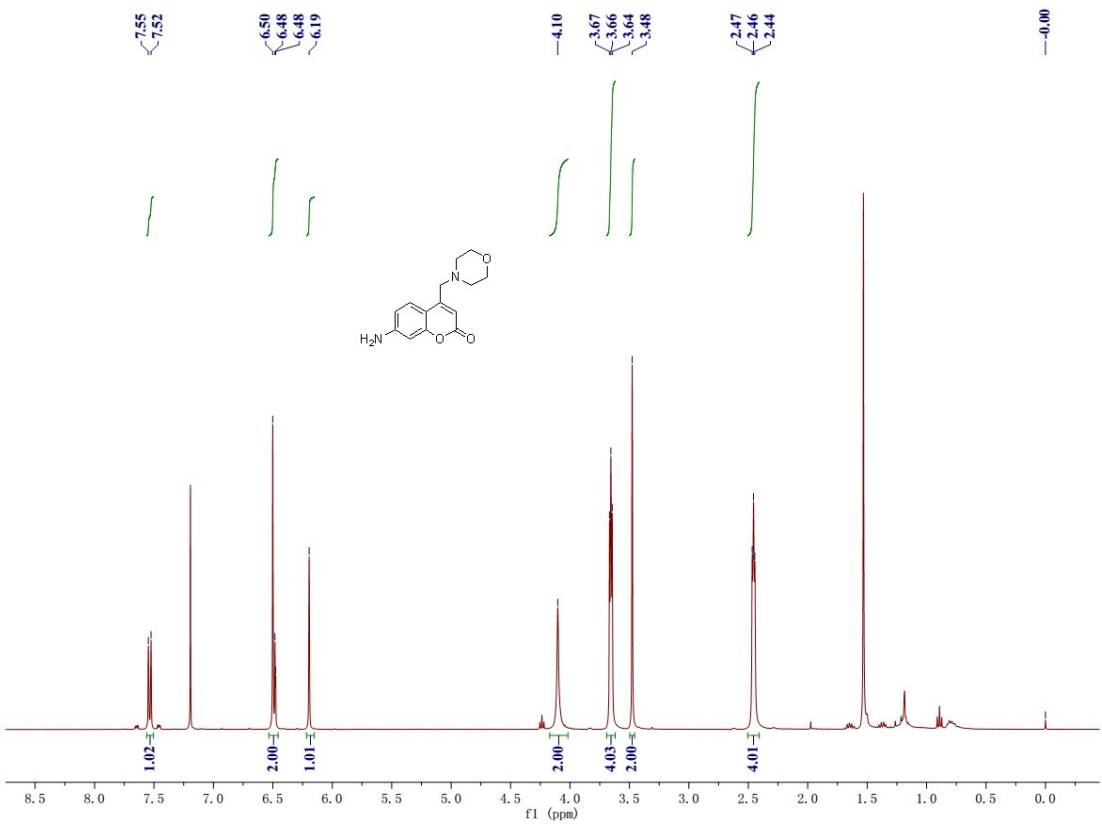
**Fig. S9**  $^1\text{H}$  NMR spectrum of compound **2** in  $\text{CDCl}_3$



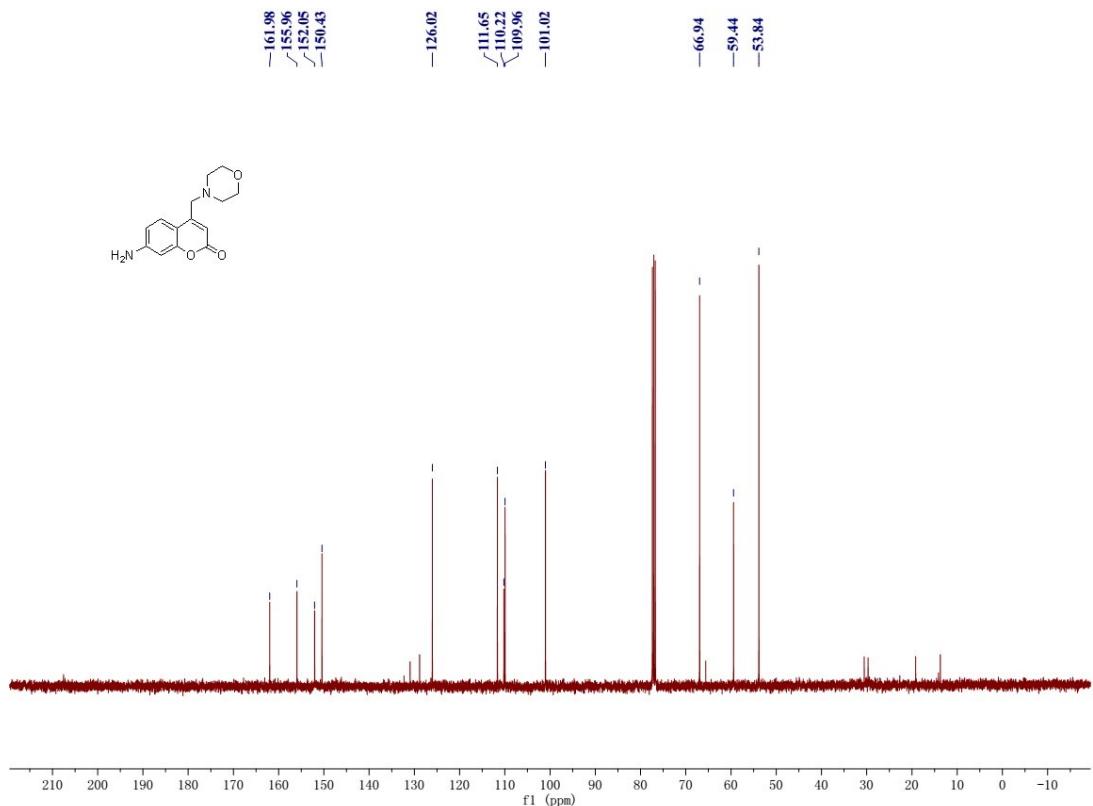
**Fig. S10**  $^{13}\text{C}$  NMR spectrum of compound 2 in  $\text{CDCl}_3$



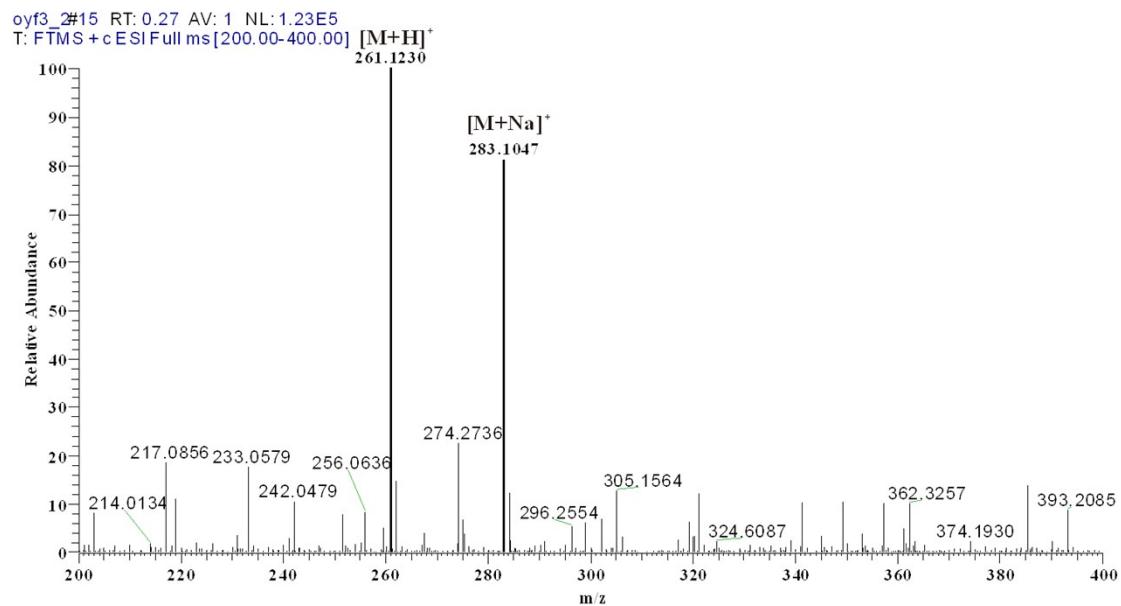
**Fig. S11** MS spectrum of compound 2



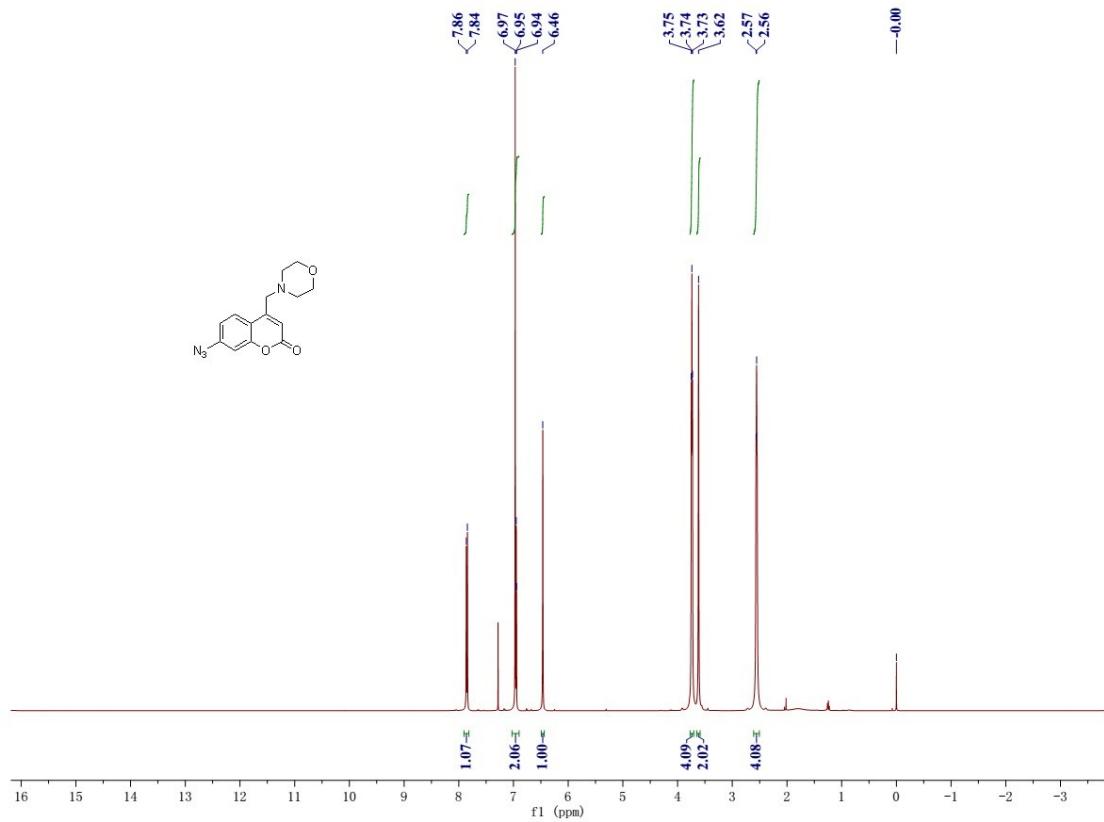
**Fig. S12**  $^1\text{H}$  NMR spectrum of Lyso-C-NH<sub>2</sub> in  $\text{CDCl}_3$



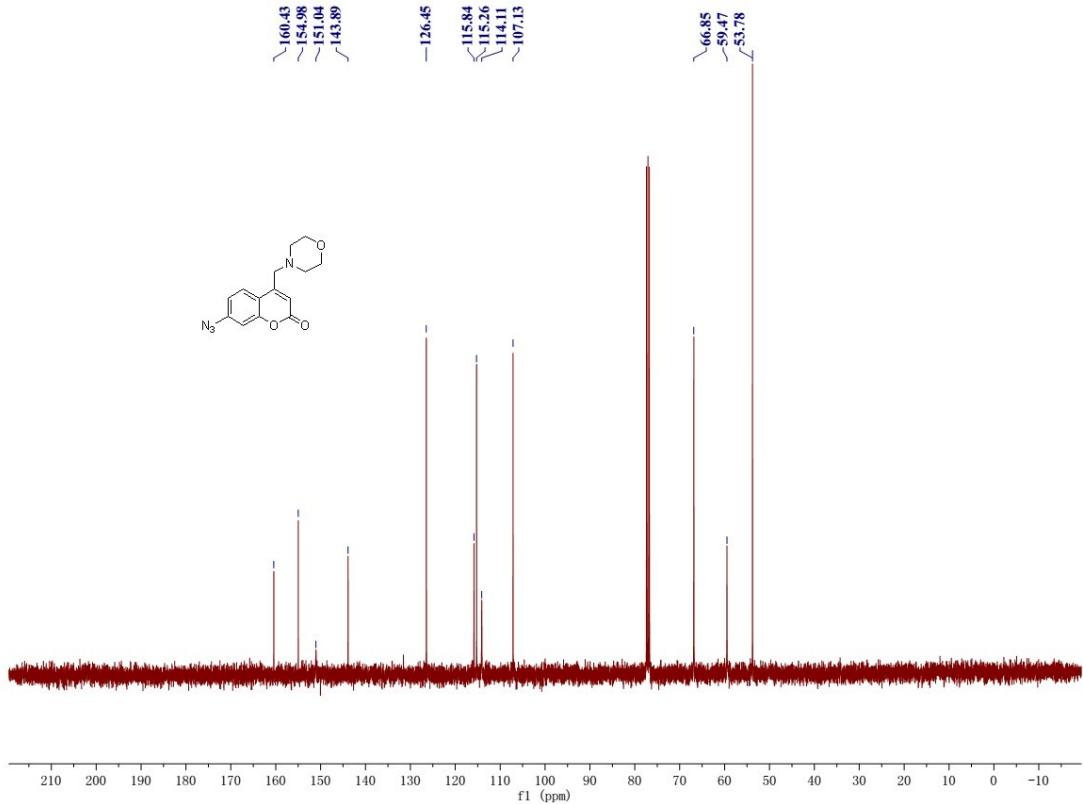
**Fig. S13**  $^{13}\text{C}$  NMR spectrum of Lyso-C-NH<sub>2</sub> in CDCl<sub>3</sub>



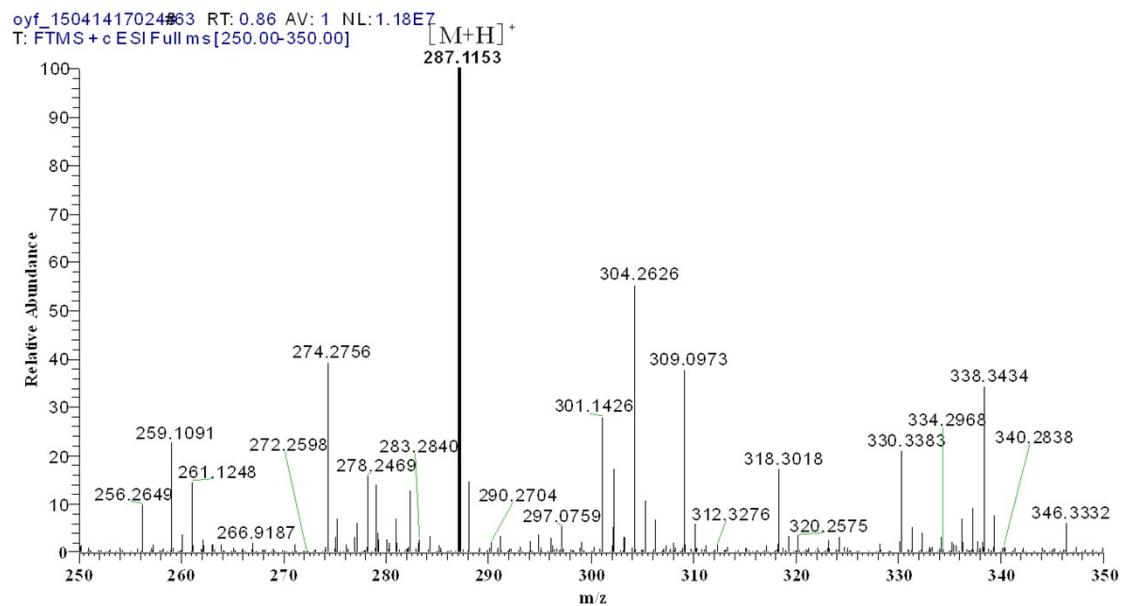
**Fig. S14** MS spectrum of Lyso-C-NH<sub>2</sub> in MeOH



**Fig. S15** <sup>1</sup>H NMR spectrum of Lyso-C in  $\text{CDCl}_3$



**Fig. S16**  $^{13}\text{C}$  NMR spectrum of Lyso-C in  $\text{CDCl}_3$



**Fig. S17** MS spectrum of Lyso-C in MeOH

**Table S1.** Comparison of probe Lyso-C with other reported lysosome-targeted H<sub>2</sub>S-responsive fluorescence probes.

Linear range	Limit of detection	Response time	Literature
<b>0 - 100 μM</b>	0.48 μM	20 min	Liu et al., <i>Org. Lett.</i> , 2013, <b>15</b> , 2310-2313
<b>Not mentioned</b>	Not mentioned	30 min	Qiao et al, <i>RSC Adv.</i> , 2014, <b>4</b> , 25790-25794.
<b>0-10 μM</b>	0.5 μM	60 min	Yang et al, <i>Anal. Chem.</i> , 2014, <b>86</b> , 7508-7515.
<b>25–2500 μM</b>	0.70 μM	Not mentioned	Feng et al, <i>Talanta</i> 2017, <b>167</b> , 134-142.
<b>0-300 μM</b>	$7.9 \times 10^{-7}$ M	30 min	Zou et al, <i>Chem. Commun.</i> , 2014, <b>50</b> , 13833.
<b>Not mentioned</b>	0.43 μM	Not mentioned	Zhang et al, <i>Terahedron</i> 2015, <b>71</b> , 8572-8576.
<b>Not mentioned</b>	3.2 μM	5 min	Liu et al, <i>Chem. Commun.</i> , 2016, <b>52</b> , 7016-7019.
<b>0-20 μM</b>	37 nM	5 min	This work