

Rational design of a lysosome-targeted fluorescence probes for monitoring the generation of hydroxyl radicals in ferroptosis pathways

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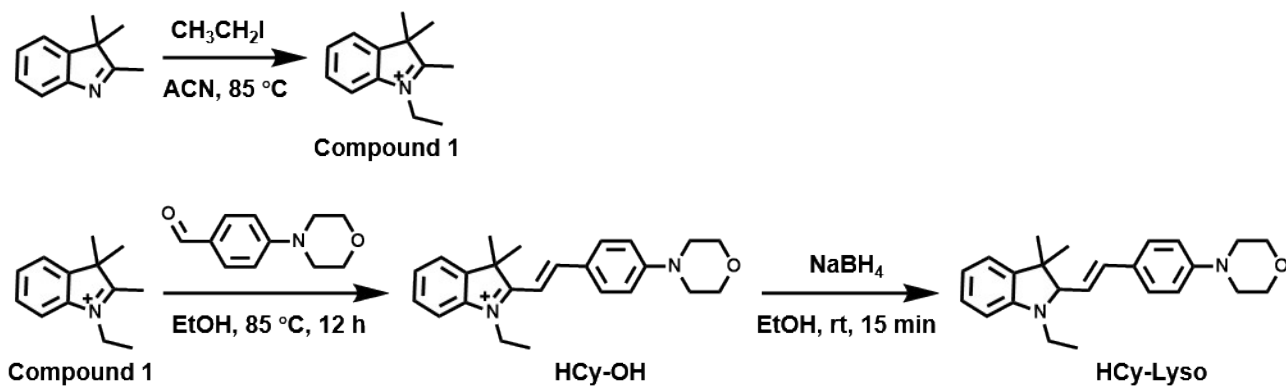
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Scheme S1. The synthesis route of HCy-Lyso.

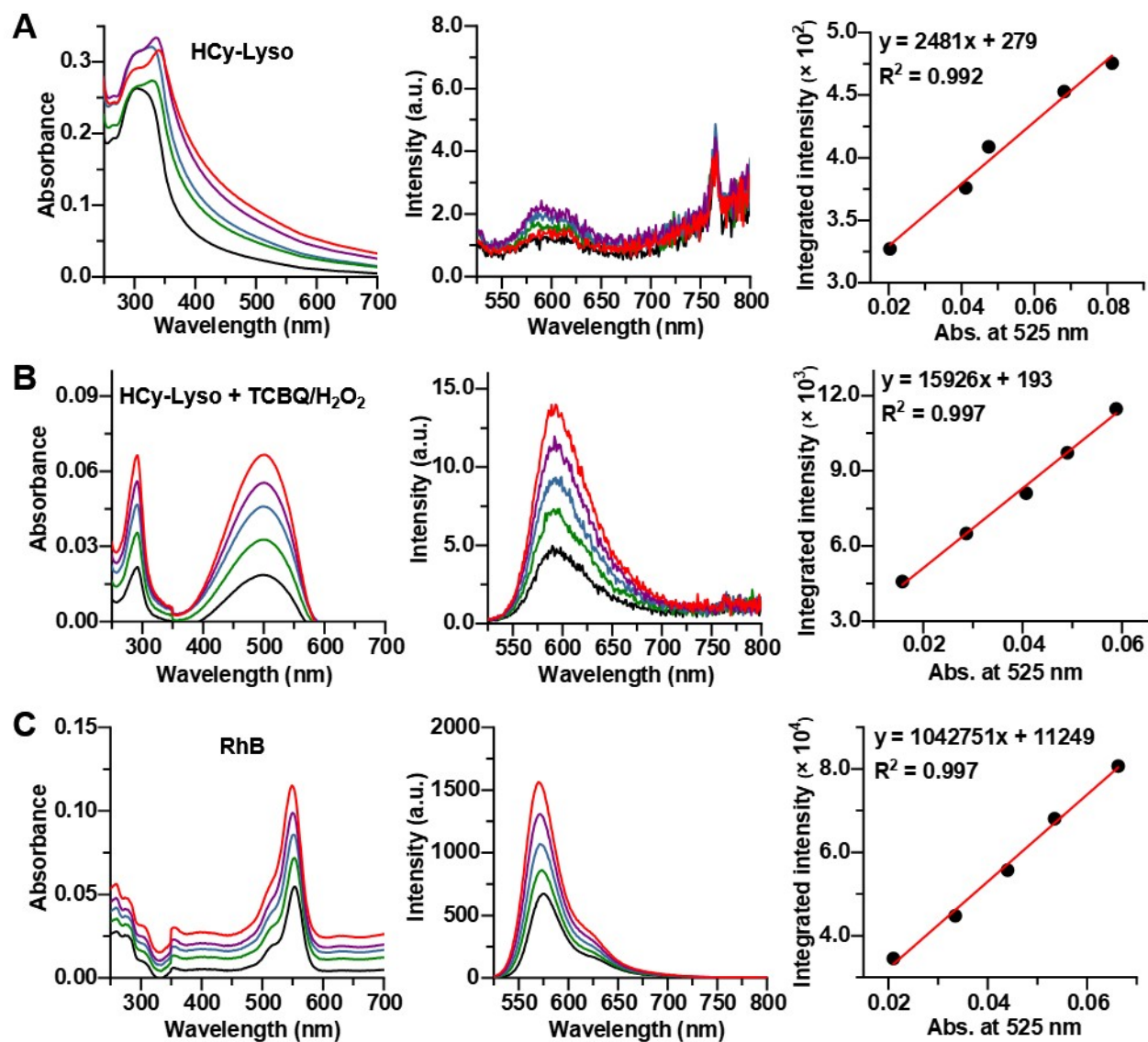


Fig. S1 Calculation the fluorescence quantum yield of HCy-Lyso, HCy-Lyso treating with TCBQ/H₂O₂ and RhB in pH 4.0 PBS solution and EtOH, respectively. Absorption spectra, fluorescence emission spectra, and plot of integrated fluorescence intensity (525-800 nm) versus absorbance at 510 nm of (a) HCy-Lyso, (b) HCy-Lyso treating with TCBQ/H₂O₂, and (c) RhB at various concentrations.

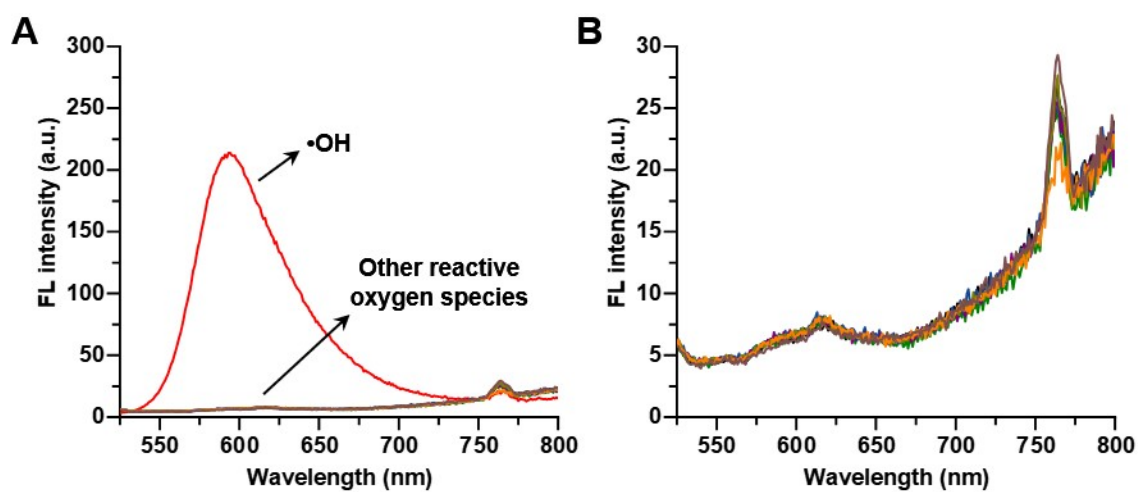


Fig. S2 (A) The fluorescence spectra of 10 μM HCy-Lyso toward various ROS in pH 4.0 phosphate buffer: control group; $\bullet\text{OH}$, 10 μM TCBQ + 10 μM H₂O₂; 100 μM OCl⁻; ¹O₂ (100 μM H₂O₂ + 500 μM OCl⁻); 100 μM NO; 100 μM H₂O₂; 100 μM ONOO⁻; 100 μM TBHP. (B) The corresponding other ROS from (A). $\lambda_{\text{ex/em}} = 510/592$ nm.

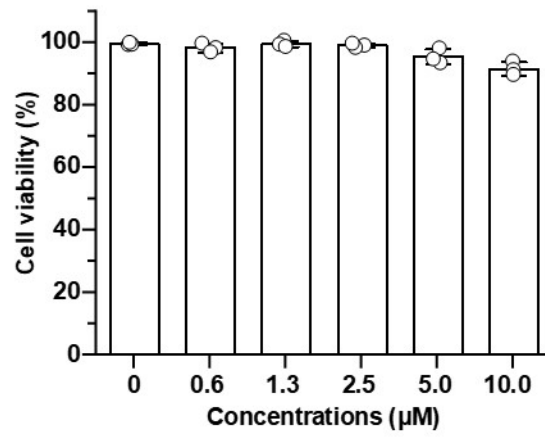


Fig. S3 Relative viability of 4T1 cells treated with various concentrations of HCy-Lyso (0-10 μM) for 12 h. Data are presented as the mean \pm SD (n = 3).

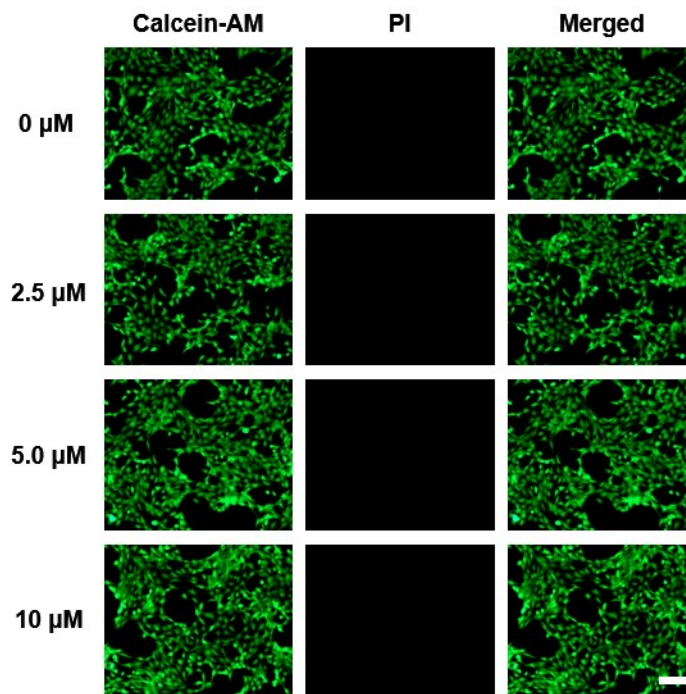


Fig. S4 Calcein-AM/PI co-staining of 4T1 cells after incubated with different concentrations of HCy-Lyso (0-10 μM) for 12 h in the dark. Scale bar: 100 μm .

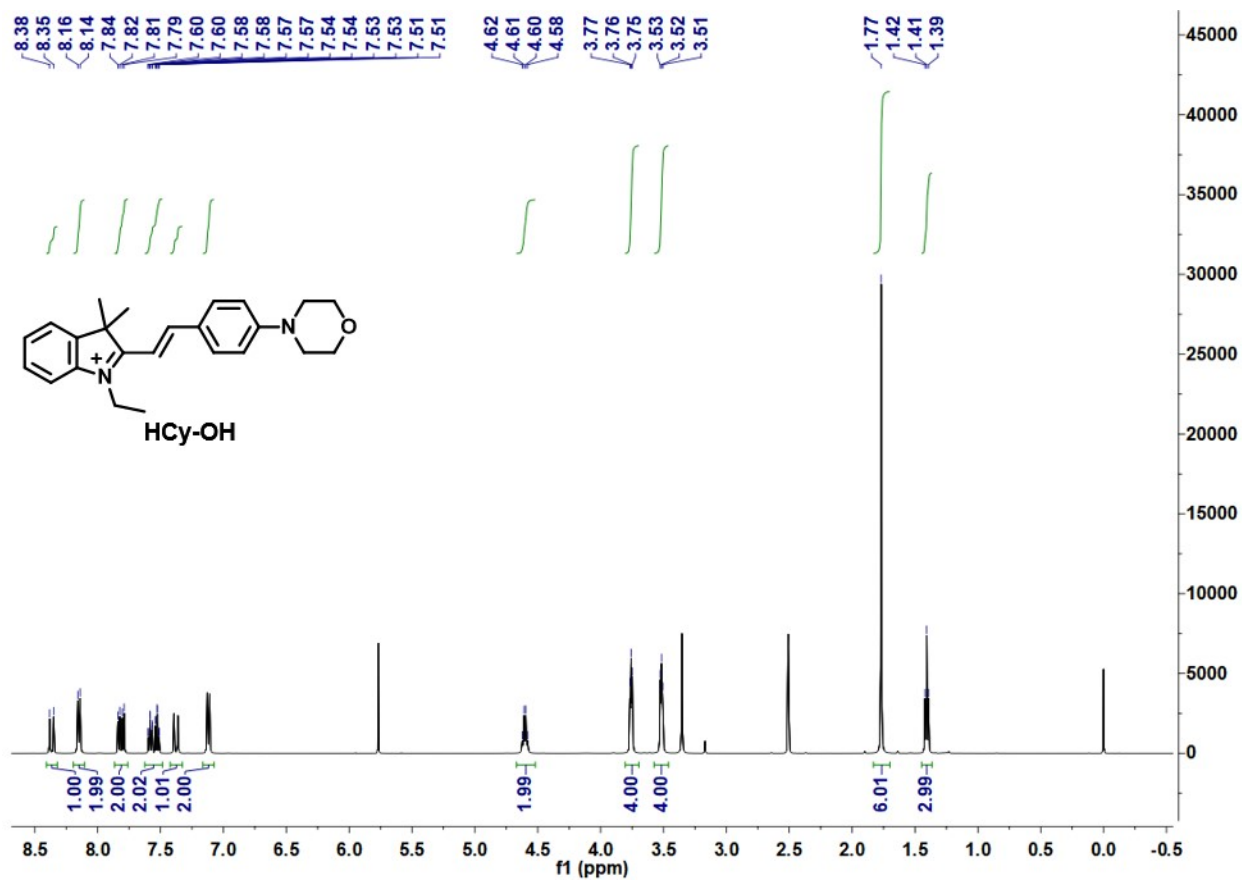


Fig. S5 ^1H NMR spectra of HCy-OH.

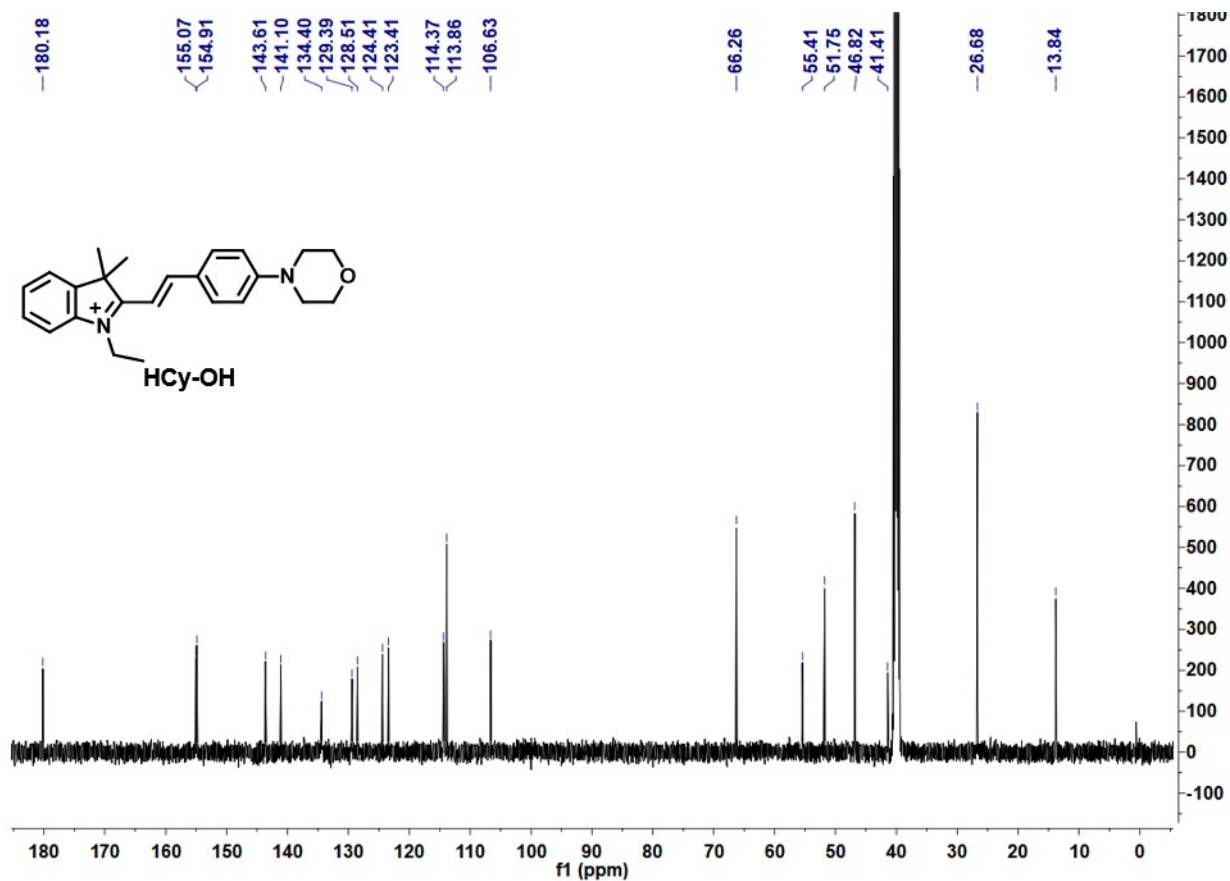


Fig. S6 ^{13}C NMR spectra of HCy-OH.

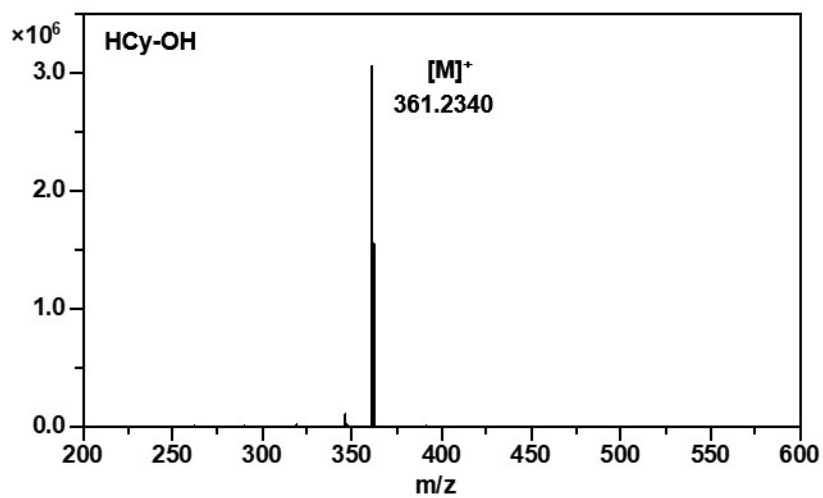


Fig. S7 HRMS spectra of HCy-OH.

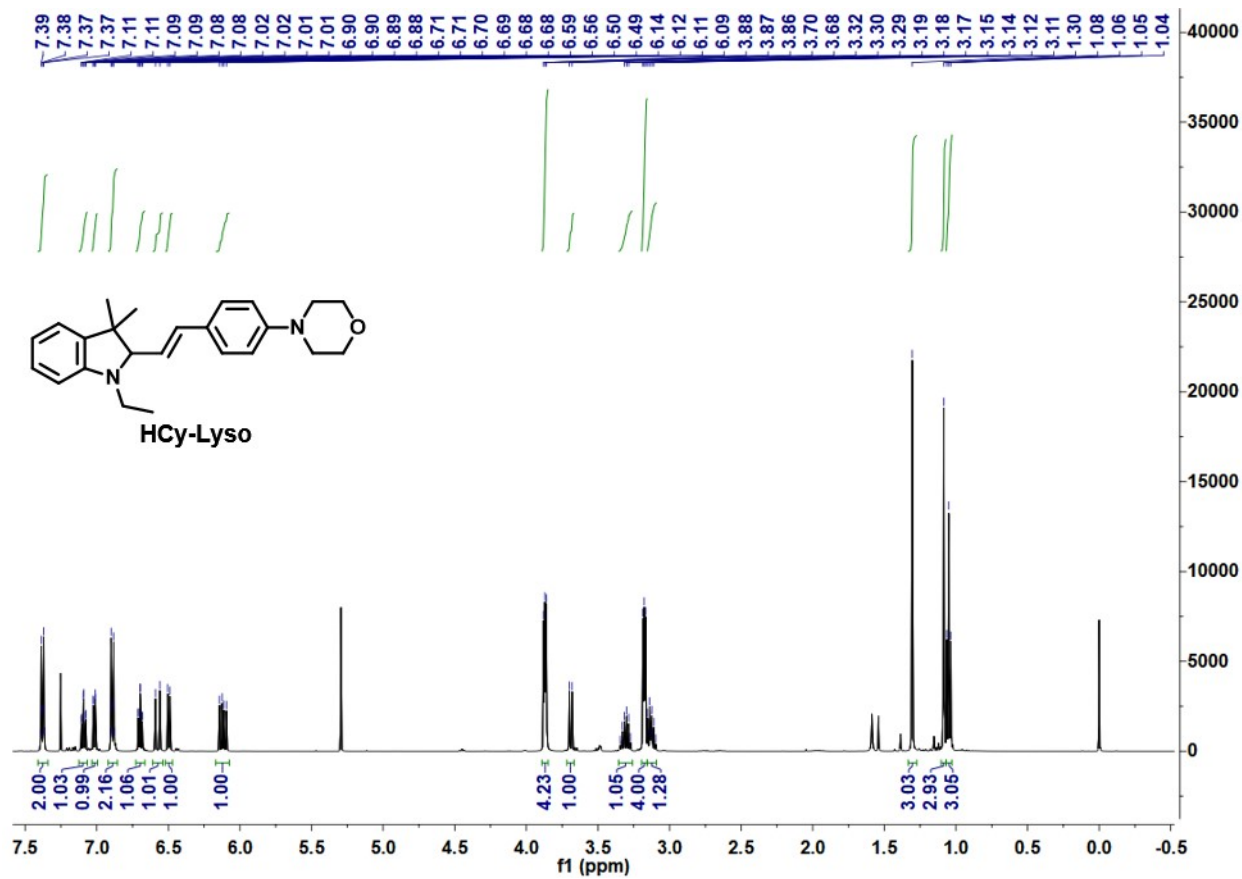


Fig. S8 ^1H NMR spectra of HCy-Lyso.

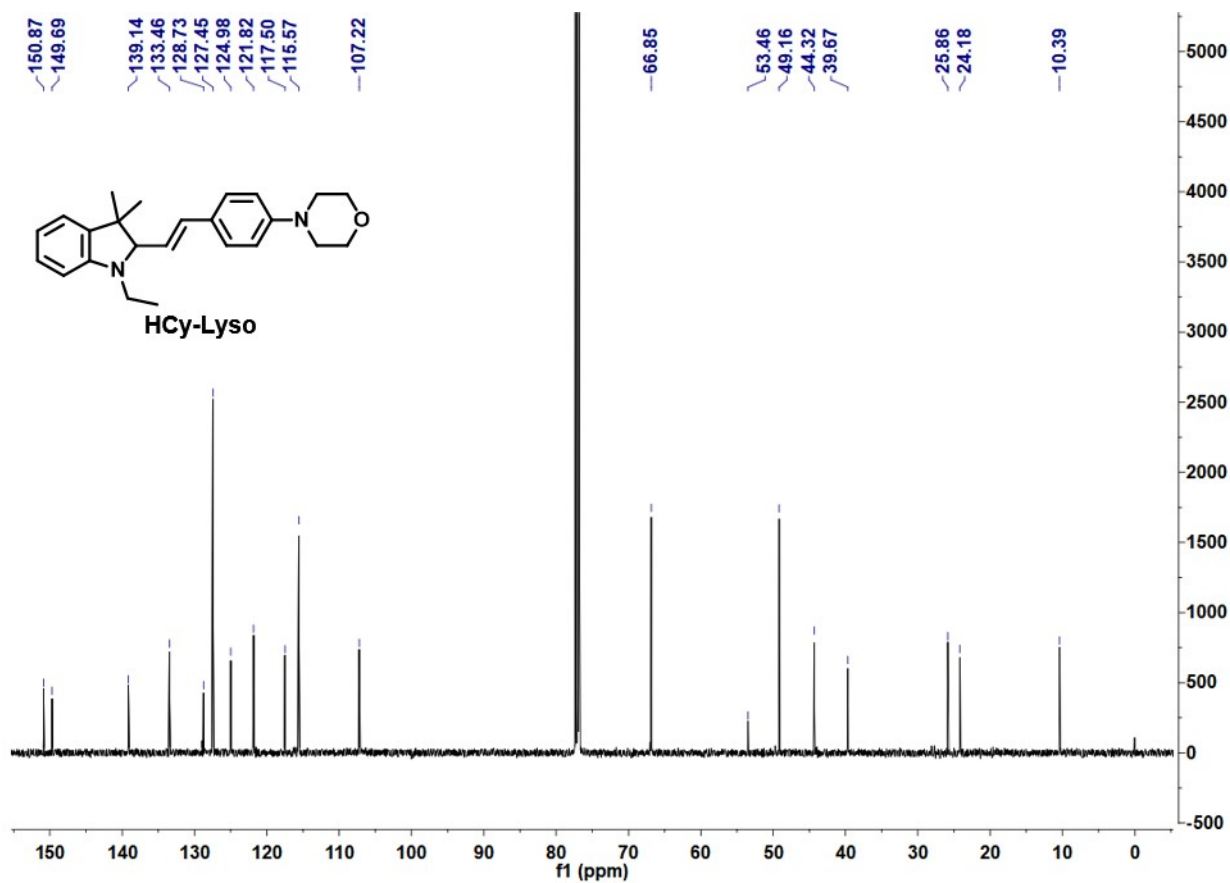


Fig. S9 ^{13}C NMR spectra of HCy-Lyso.

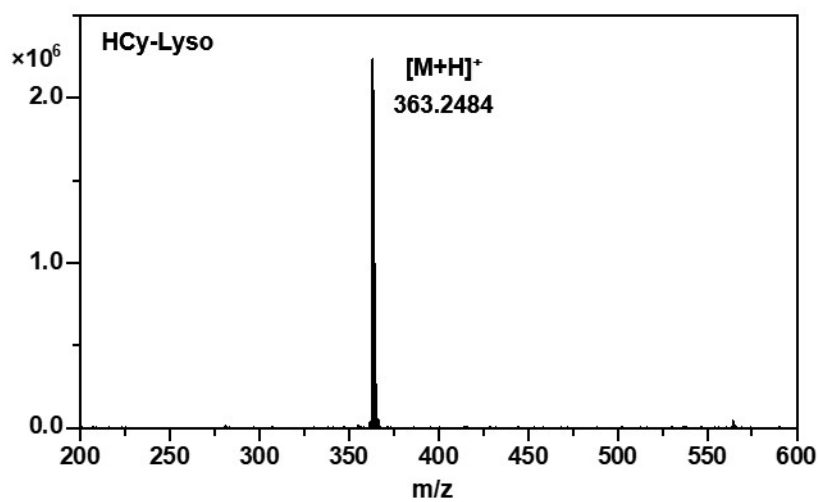


Fig. S10 HRMS spectra of HCy-Lyso.

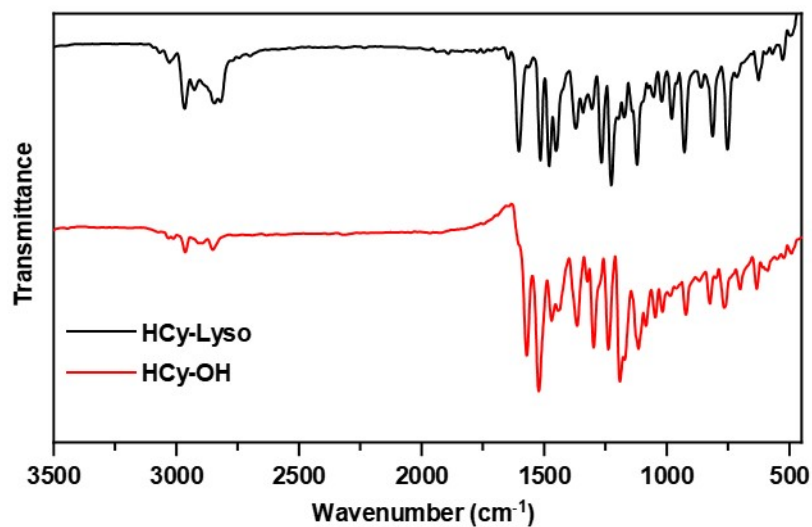


Fig. S11 FTIR spectra of HCy-Lyso and HCy-OH.

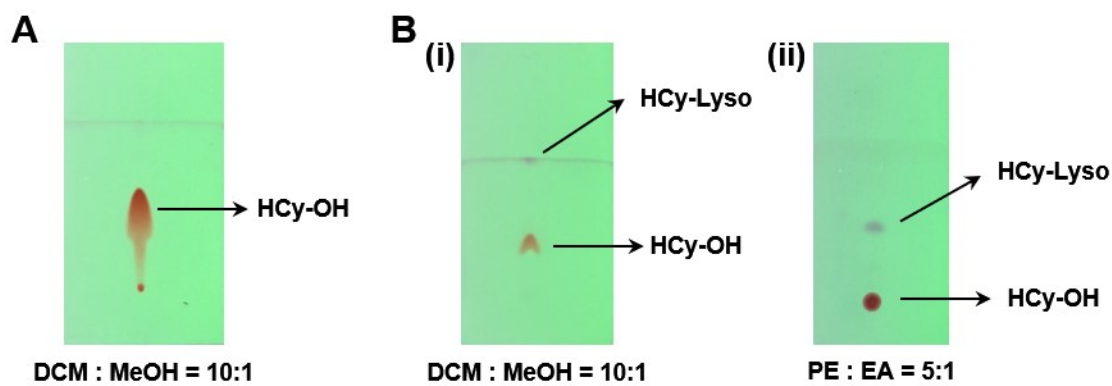


Fig. S12 (A) Thin layer chromatography plate of HCy-OH reaction solution, run with dichloromethane and methanol as eluent. (B) Thin layer chromatography of HCy-Lyso mixture with different eluent, (i) dichloromethane and methanol; (ii) petroleum ether and ethyl acetate.