Supporting Information

Water soluble glucose-appended quinoline-benzothiazole conjugate as a selective and sensitive receptor for Cu^{+} ion in aqueous media and intracellular bio-imaging in live cells

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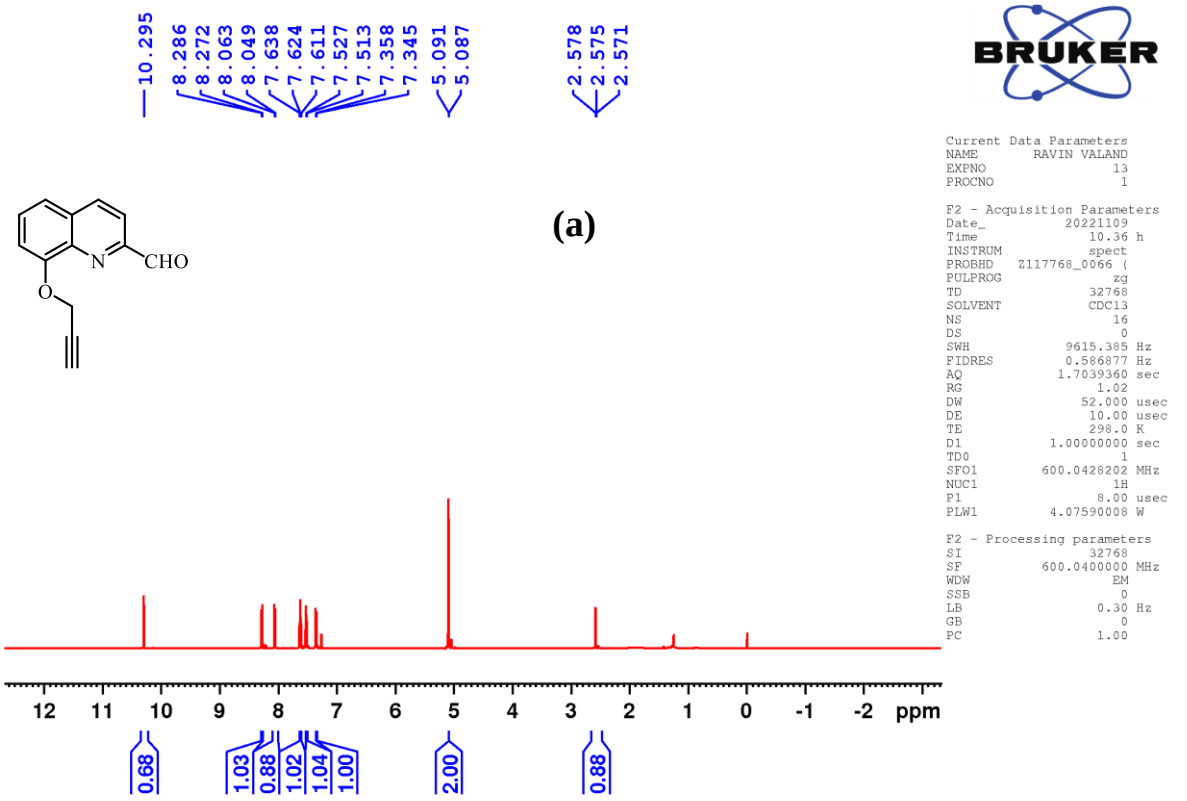
S 01. Synthesis and characterization of compound P_2

The **P_2** has been synthesized by a producer in the reported literature[1]. In a dry 100 mL round bottom flask, 8-hydroxyquinoline 2-carboxaldehyde (2 g, 11 mmol) and potassium carbonate (2.4 g, 17.3 mmol) were dissolved in *N*, *N*-dimethylformamide (DMF) and stirred for 5 minutes. Subsequently, a solution of propargyl bromide (1.3 mL, 17.3 mmol) was added dropwise to the reaction mixture. The resulting mixture was stirred at room temperature overnight. Upon completion of the reaction, the mixture was separated using ethyl acetate and water. The organic layer was extracted with ethyl acetate, dried over anhydrous sodium sulfate (Na_2SO_4), and concentrated under a vacuum. The crude product was then purified by column chromatography to yield a brown precipitate (1.94 g, Yield= 80 %) **^{1}H NMR ($CDCl_3$, 600 MHz):** δ 10.29 (s, 1H), 8.27 (d, *J*=8.46 Hz, 1H), 8.05 (d, *J*=8.45 Hz, 1H), 7.51 (d, *J*=8.15 Hz, 1H), 7.35 (d, *J*=7.74 Hz, 1H), 5.08 (d, *J*=2.31 Hz, 2H), 2.57 (t, *J*=2.30 Hz, 1H) **^{13}C NMR ($CDCl_3$, 150 MHz):** δ 193.99, 154.12, 151.96, 140.43, 137.69, 131.73, 129.78, 121.01, 118.28, 111.39, 78.17, 76.95, 57.23



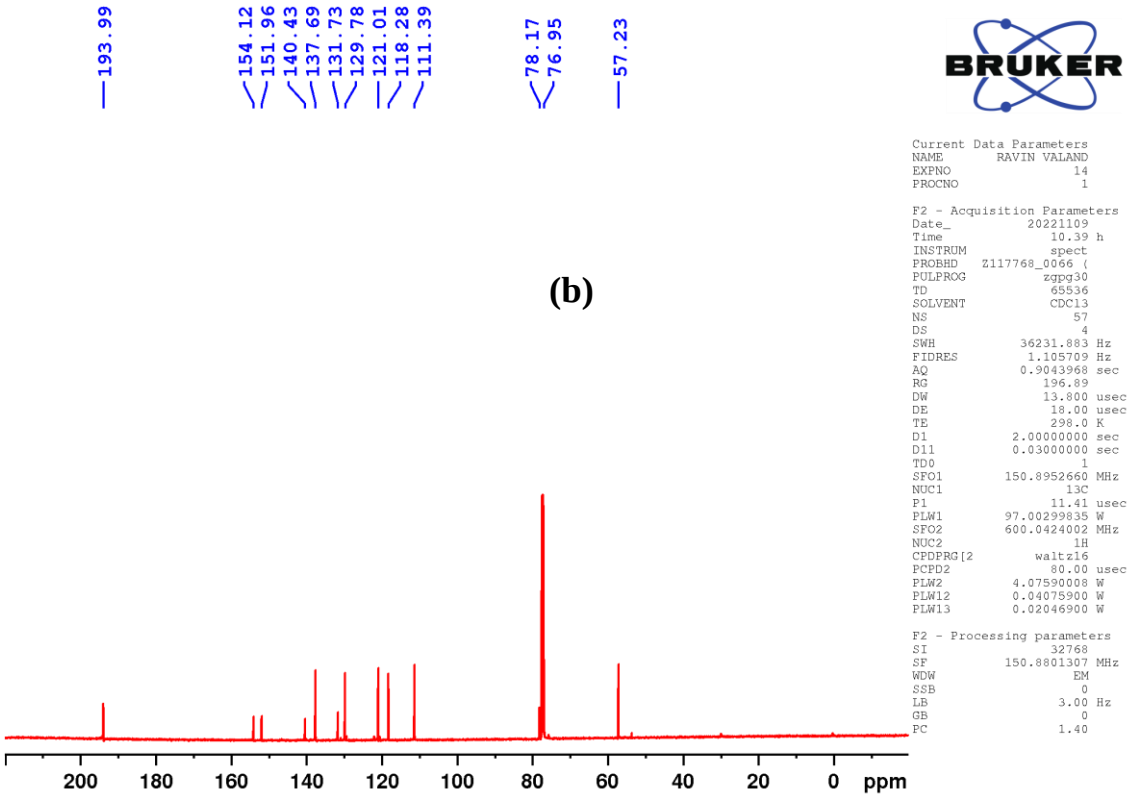
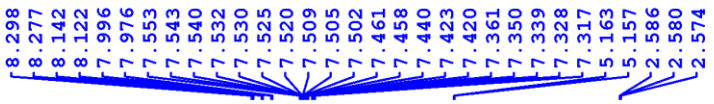
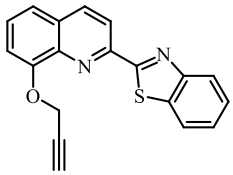


Fig. S1. Spectrum of compound **P₂** in $CDCl_3$ (a) ^{1}H NMR (b) ^{13}C NMR

S2. Synthesis and characterization of compound P₃

P₂ (1 g, 4.7 mmol) and 2-aminobenzene-1-thiol (608 µL, 5.6 mmol) were dissolved in DMSO and stirred at 120 ℃ for 2 hours. The reaction mixture cooled to room temperature and poured in water. The crude product was extracted with ethyl acetate and washed with water. The Organic layer was dried over anhydrous sodium sulfate and concentrate under vacuum. The crude product was then purified by silica gel column chromatography using a 4:6 mixture of Ethyl acetate and petroleum ether. The pure fraction was collected and evaporated to yield the product (1.12 g, Yield= 75 %) **^{1}H NMR ($CDCl_3$, 400 MHz):** δ 8.50 (d, *J*=8.57 Hz, 1H), 8.28 (d, *J*= 7.59 Hz, 1H), 8.13 (d, *J*=8.16 Hz, 1H), 7.98 (d, *J*=7.86 Hz, 1H), 7.54-7.50 (m, 1H), 7.45-7.42 (m, 1H), 7.33 (m, 1H) **^{13}C NMR ($CDCl_3$, 100 MHz):** δ 170.31, 154.73, 153.72, 150.66, 140.71, 137.46, 136.91, 130.67, 128.06, 126.59, 126.18, 124.09, 122.38, 121.57, 119.10, 113.51, 78.93, 76.49, 58.22 **ESI-MS:** calcd. for $C_{19}H_{12}N_2OSH^+[M+H]^+$ 316.0670, found 317.0748





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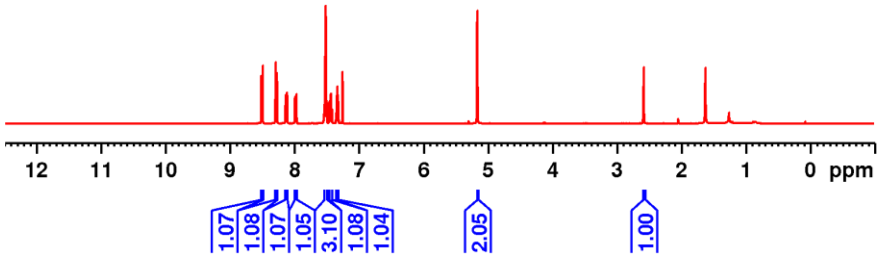


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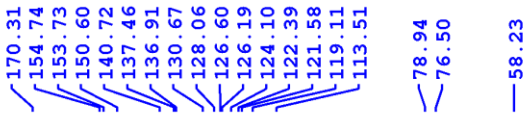

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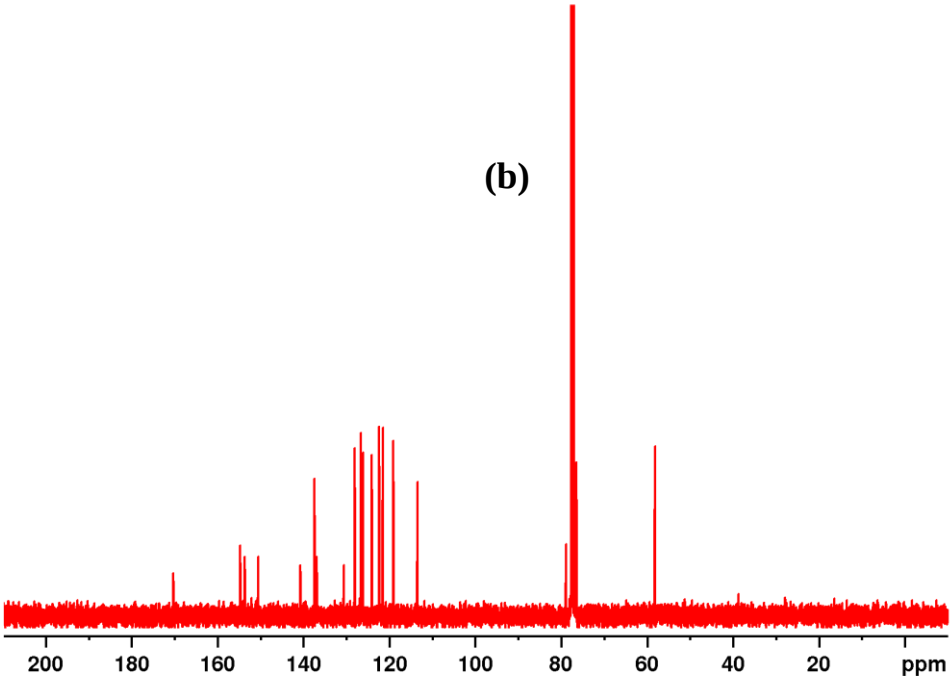


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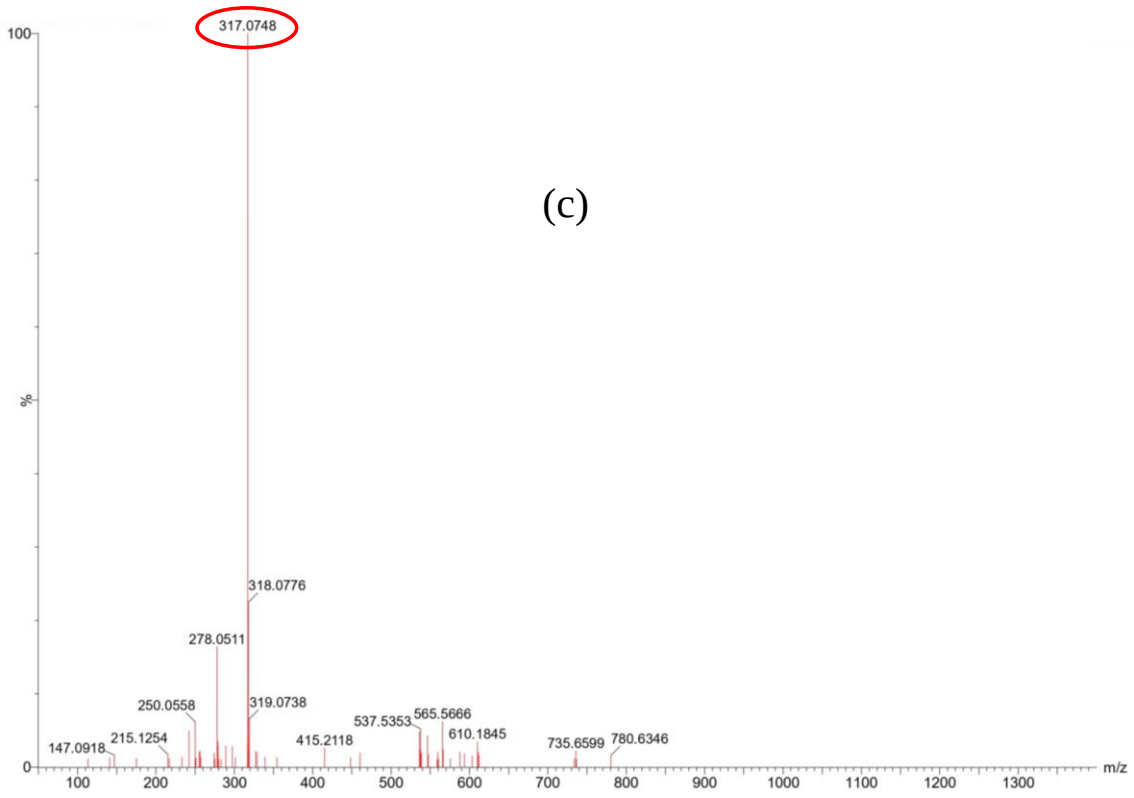


Fig. S2. **(a)** ^{1}H NMR ($CDCl_3$), **(b)** ^{13}C NMR ($CDCl_3$) and **(c)** ESI-MS spectrum of compound **P_3**

S3. Synthesis and Characterization of Compound P_5

The **P_5** has been synthesized by a producer in the reported literature[2]. The final yield was 61% resulting in off-white crystal. **^{1}H NMR ($CDCl_3$, 600 MHz):** δ 5.15 (t, *J*=9.50 Hz, 1 H), 5.04 (t, *J*=9.70 Hz, 1H), 4.89 (t, *J*=9.20 Hz, 1H), 4.60 (d, *J*=8.90 Hz, 1H), 4.21 (dd, *J*=12.40 Hz, *J*=4.45 Hz, 1H), 4.10 (d, *J*=12.40 Hz, 1H), 3.74 (dd, *J*=9.96 Hz, 1H), 2.03 (s, 3H), 2.01 (s, 3H), 1.96 (s, 3H), 1.94 (s, 3H) **^{13}C NMR ($CDCl_3$, 150 MHz):** δ 170.82, 170.33, 169.54, 169.43, 88.11, 74.24, 72.82, 70.86, 68.12, 61.89, 20.92, 20.78, 20.76

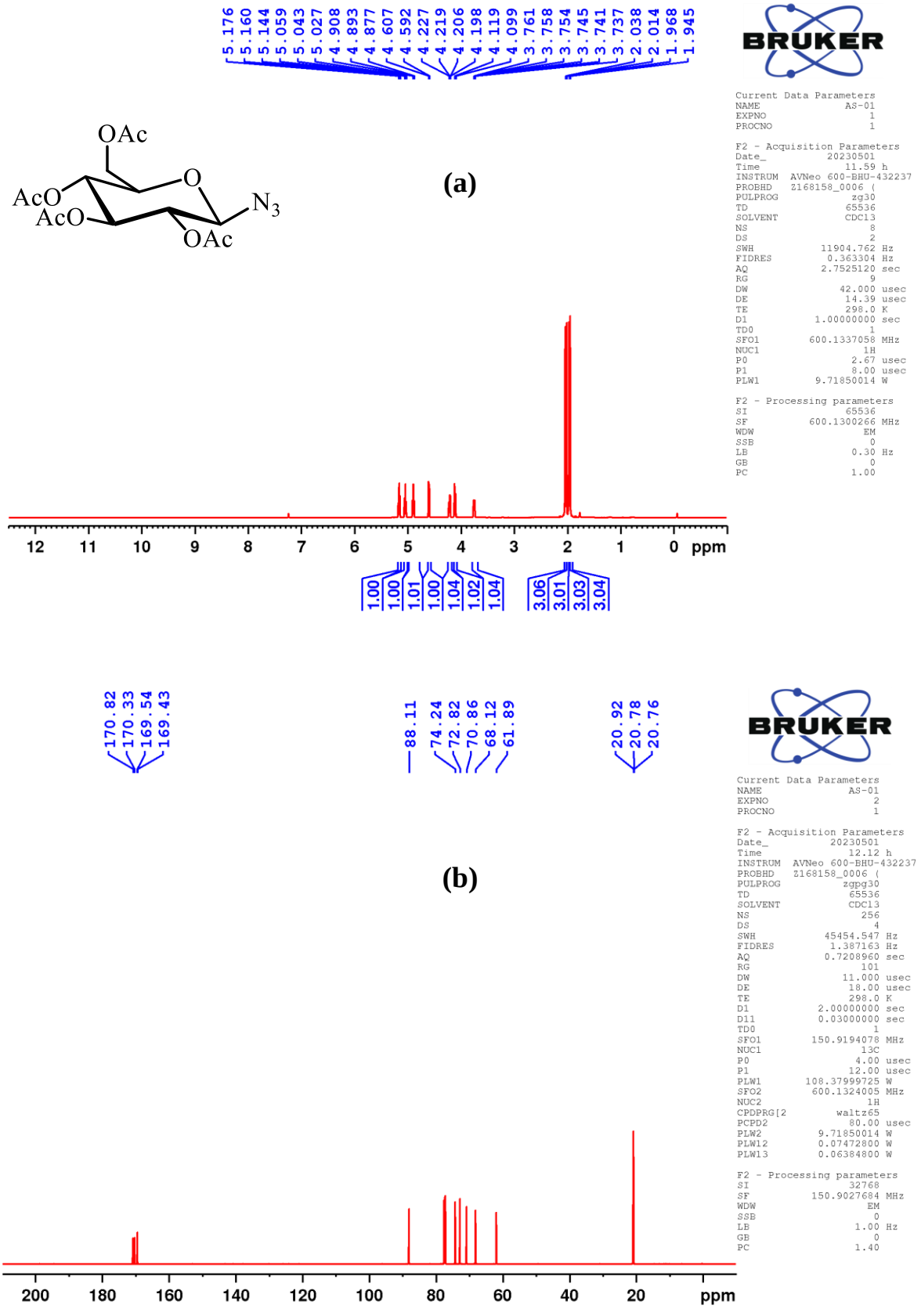
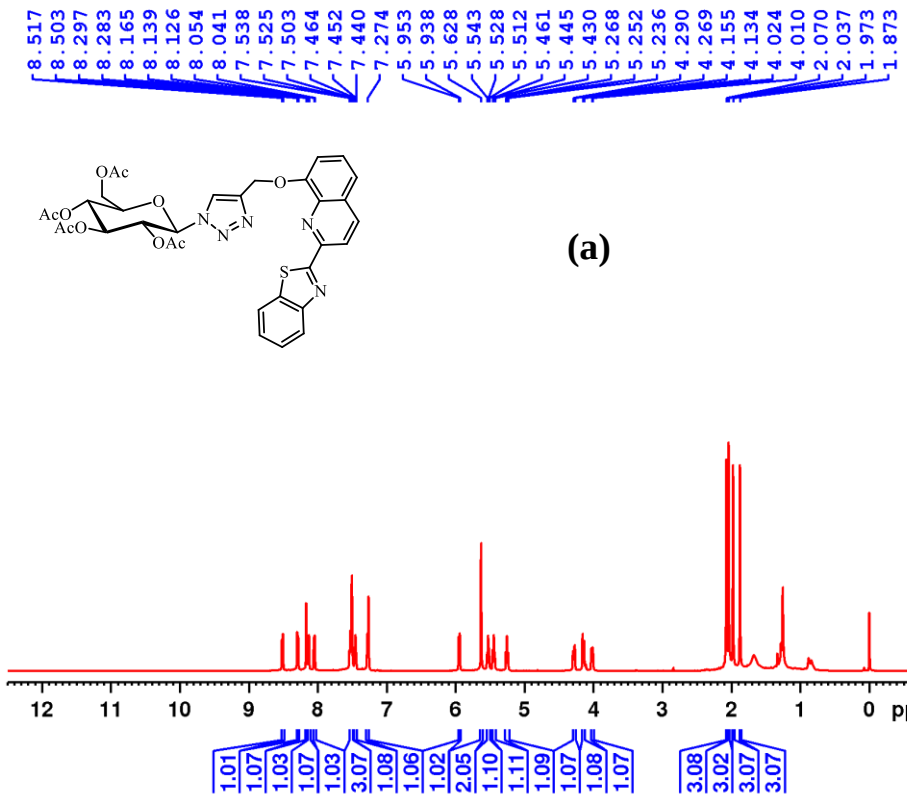


Fig. S3. Spectrum of compound **P5** in $CDCl_3$ **(a)** ^{1}H NMR and **(b)** ^{13}C NMR

S4. Characterization of compound P_6

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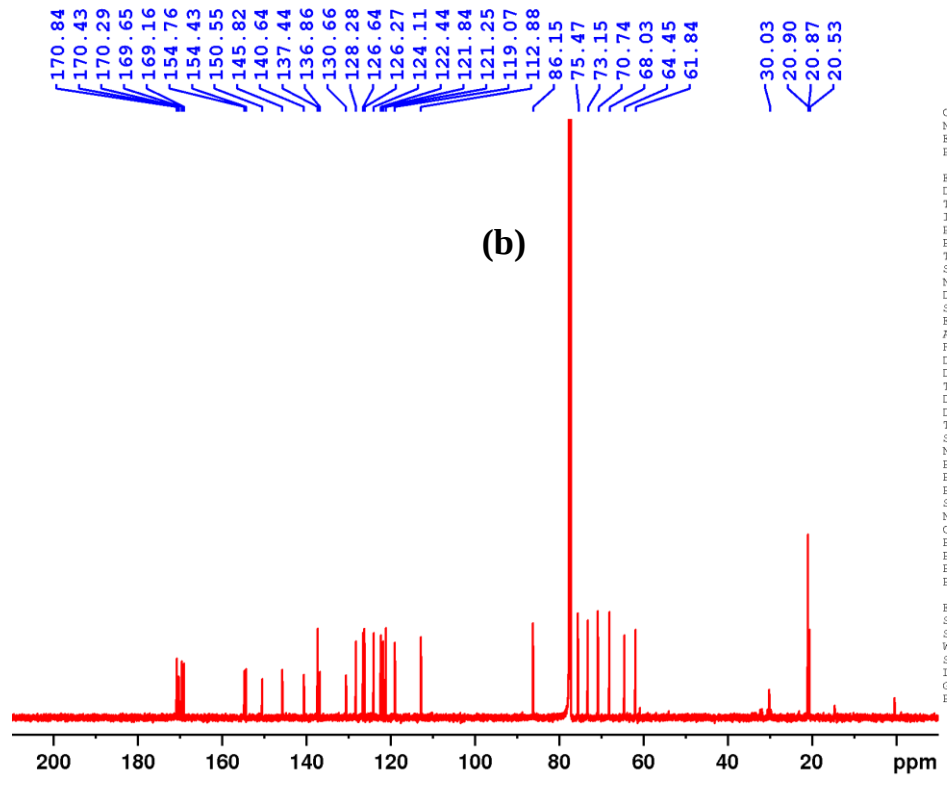


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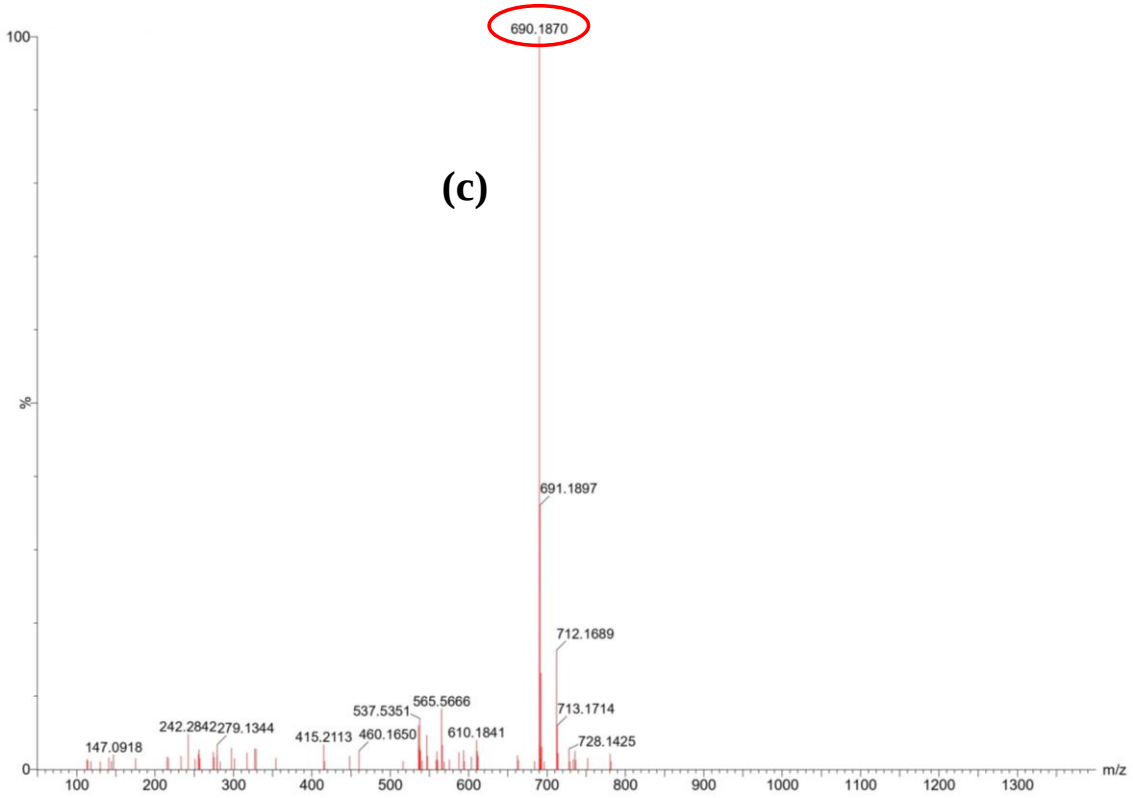
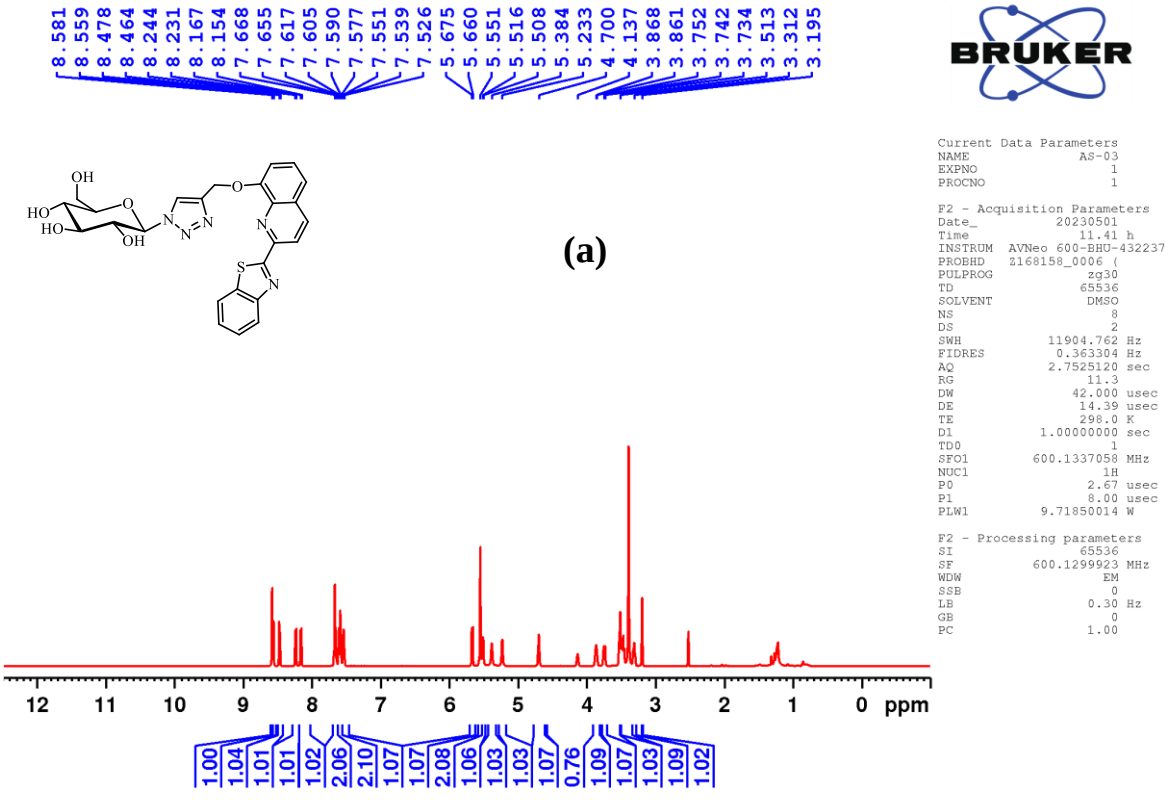


Fig. S4: **(a)** ^{1}H NMR ($CDCl_3$), **(b)** ^{13}C NMR ($CDCl_3$) and **(c)** ESI-MS spectrum of compound **P$_6$**

S5. Characterization of compound L



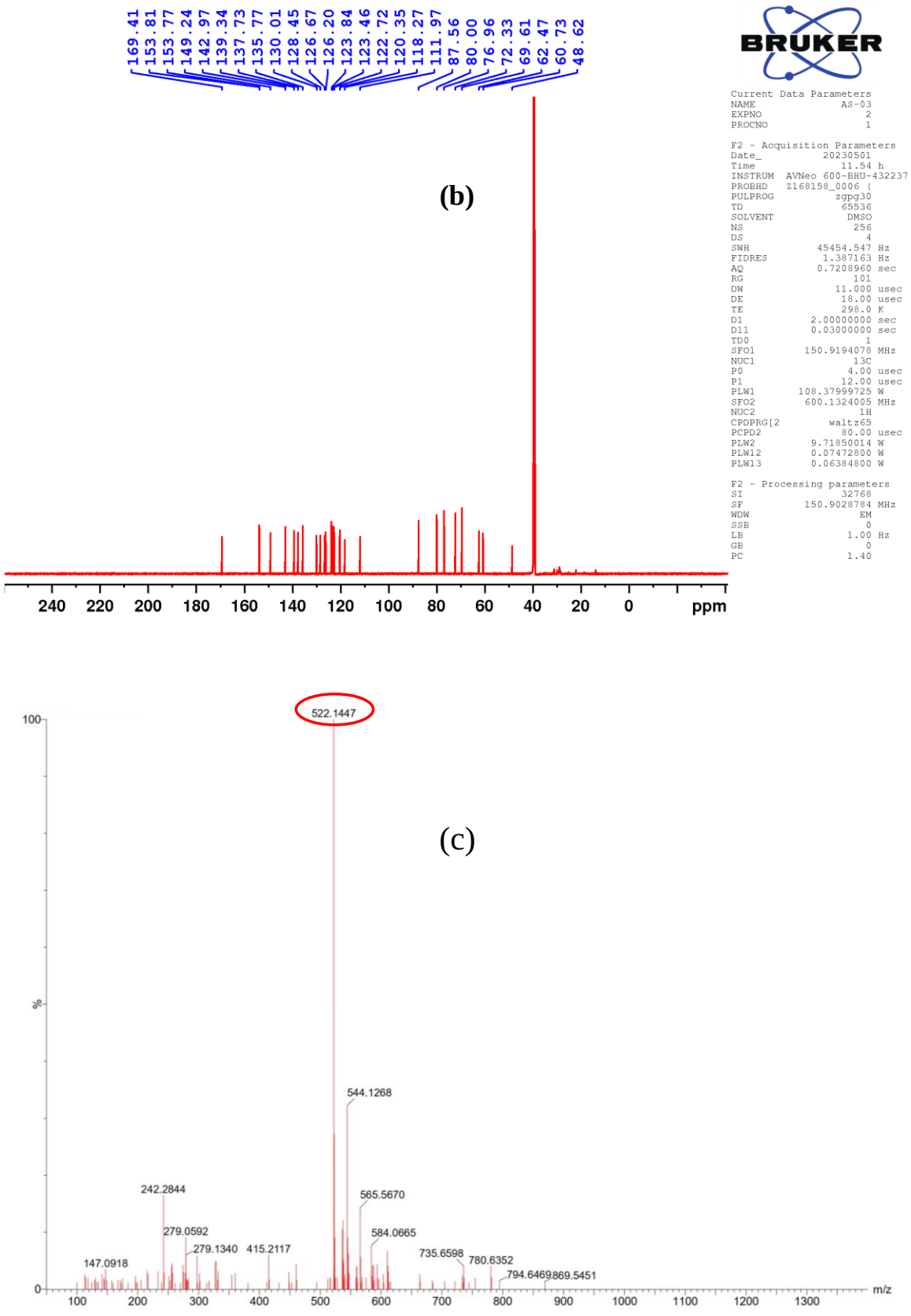


Fig. S5. (a) ^{1}H NMR (DMSO-d_6), **(b)** ^{13}C NMR (DMS)-d_6) and **(c)** ESI-MS spectrum of compound **L**

S6. Solubility of probe L in water

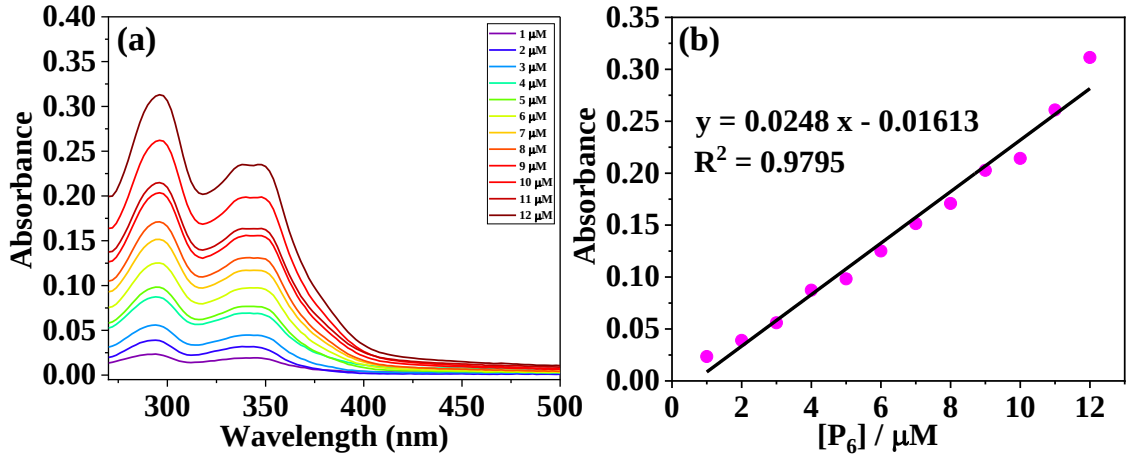


Fig. S6 (a) UV-Vis spectra of **L** at different concentrations ranging from 1-12 μM in water (b) A plot of absorbance (294 nm) vs. different concentrations of **L** in water.

S7. Effect of absorbance and Fluorescence on probe L using different metal ions

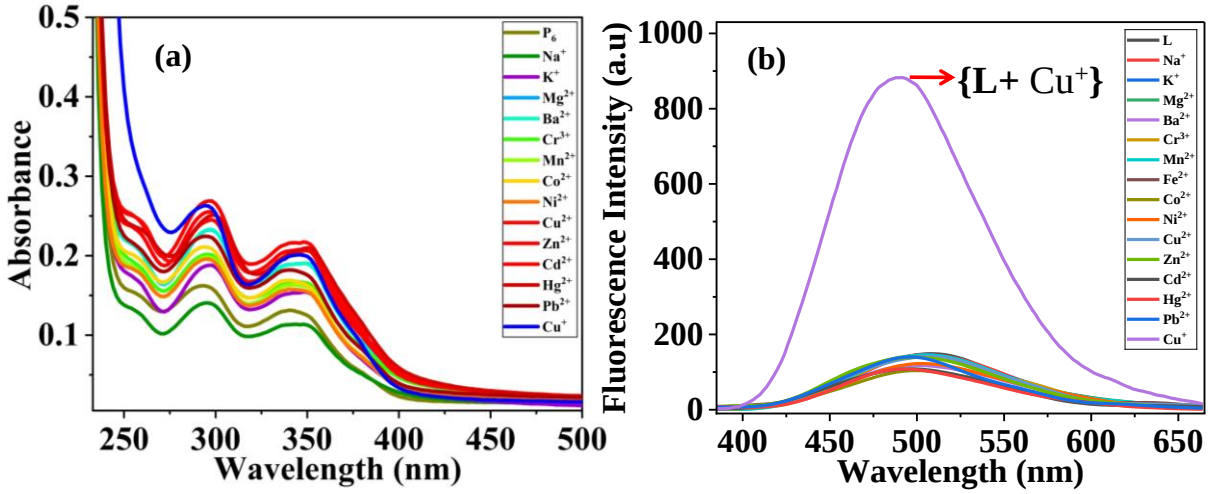


Fig. S7 (a) Absorbance of **L** (10 μM) with Cu^{+} (3 equivalent) and other metal ions (10 equivalent) in water **(b)** fluorescence spectral titration of **L** (10 μM) with Cu^{+} (3 equivalents) and other different metals (10 equivalents) (λ_{ex}=294 nm) in water.

S8 Fluorescence spectra of L with different metal ions

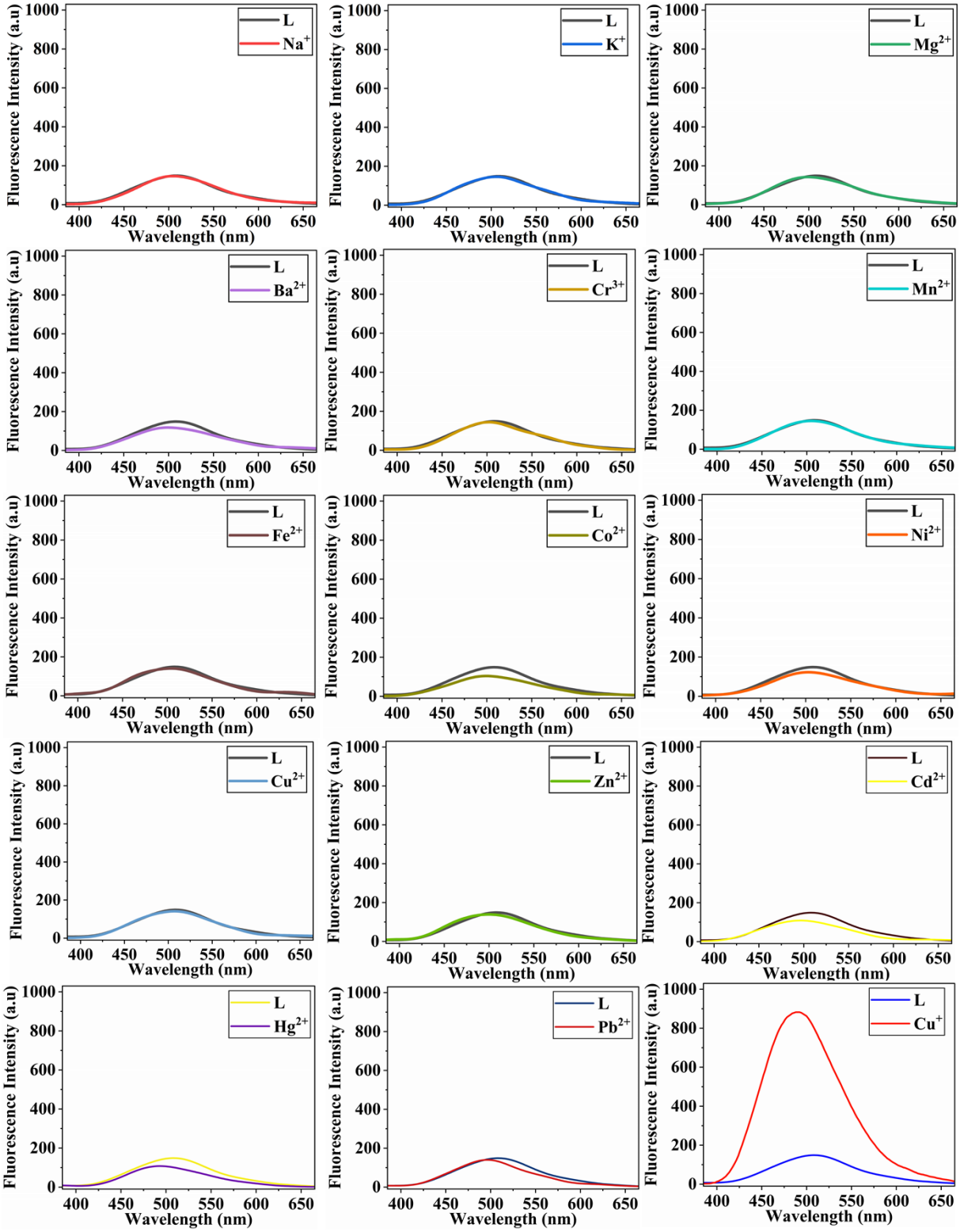


Fig. S8. Fluorescence spectra of **L** (10 μM) titrated with various metal ions (10 equivalents) in water.

S9. Association constant obtained from the fluorescence titration of {L+Cu^+} complex

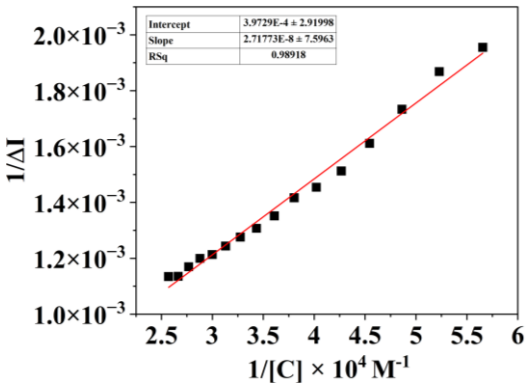


Fig. S9. Binding constant (K_a) of Ligand **L** with Cu^+ concentration in water

S10. Determination of Limit of Detection (LOD) of Cu^+ by L

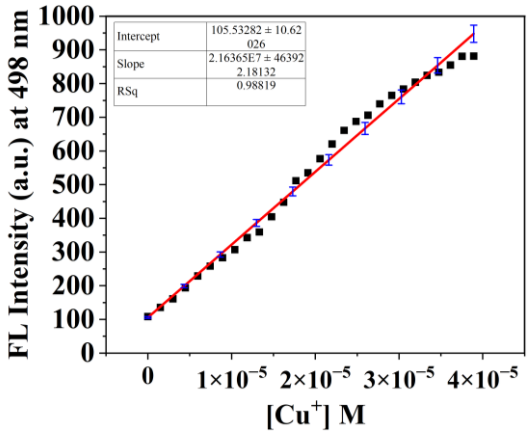


Fig. S 10. Linear fluorescence relationship of Ligand (**L**) with Cu^+ (3-90 µM) in water at λ_{em}=498 nm

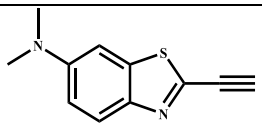
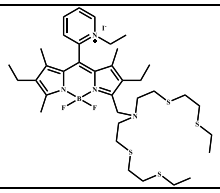
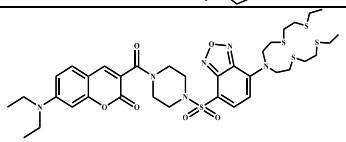
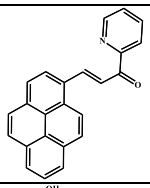
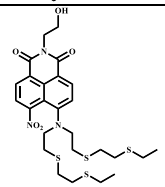
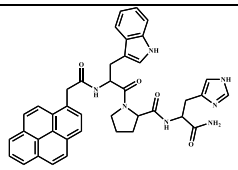
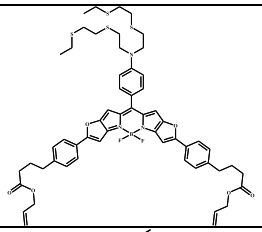
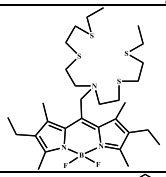
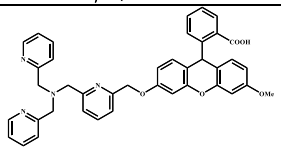
S11. Fluorescence quantum yield of L and {L+Cu^+} complex

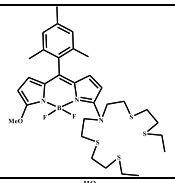
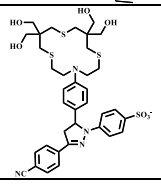
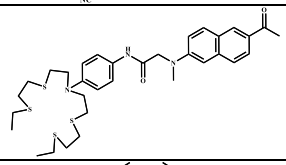
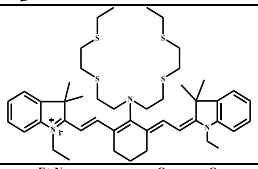
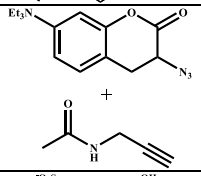
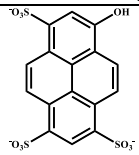
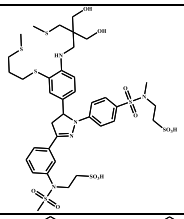
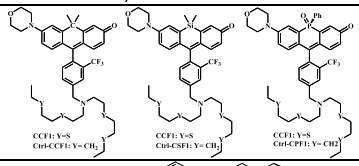
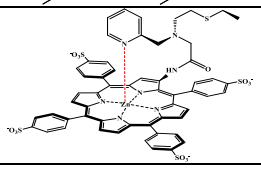
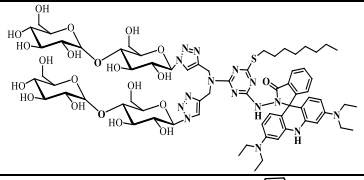
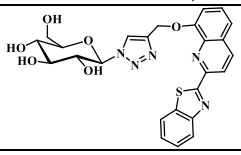
The quantum yield (Φ) was determined using literature reported method by the following equation.[3]

$$\Phi Y_{sample} = \Phi Y_{Reference} \left(\frac{\text{sample gradient}}{\text{reference gradient}} \right) \left(\frac{\text{sample refrective index}}{\text{reference refrective index}} \right)^2$$

where the subscripts R and S denote the reference standard (fluorescein in 0.1 M NaOH) and the synthesized sample **L**, respectively. Φ represents the quantum yield, F is the integrated fluorescence emission, A is the absorbance at the excitation wavelength, and n is the refractive index of the solvent. The quantum yield of **L** was calculated using fluorescein as the standard, which has a reported quantum yield of 0.79 in 0.1 M NaOH.

S12. Table 1: Comparisons between **L** and the molecular probes reported in the literature.

Sr. No.	Fluorescent molecular systems	Ion	Limit of detection (LOD)	Solvent system	Ref.
1.		Cu^{+}	N.R	PBS buffer (1 mM, pH 7.2)	[31] *Bioorg. Med. Chem. Lett*., 2012, **22**, 1747–1749.
2.		Cu^{+}	N.R	HEPES buffer (10 mM, pH 7.2)	[32] *Chem. Commun*., 2014, **50**, 9835–9838.
3.		Cu^{+}	N.R	Tris-HCl buffer/DMF 4:6 v/v pH=7.2	[33] *Sci. China: Chem*., 2019, **62**, 465–474.
4.		Cu^{+}	1.2×10^{-9} M	SDS (8 mM) micelles (EDTA 2 μM)	[34] *J. Mater. Chem. B*, 2023, **11**, 4111-4120
5.		Cu^{+}	N.R	HEPES buffer solution (with 50% of acetonitrile, pH 7.2)	[35] *Chem. Commun.*, 2013, **49**, 5565–5567.
6.		Cu^{+}	N.R	(10 mM HEPES, pH 7.4) containing 1% DMF	[36] *Sensors and Actuators B*, 2018, **256**, 393–401.
7.		Cu^{+}	N.R	Thiourea buffer in methanol	[37] *Angew. Chem., Int. Ed*., 2021, **133**, 23332–23337.
8.		Cu^{+}	N.R*	HEPES buffer (20 mM, pH 7)	[38] *J. Am. Chem. Soc.* 2006, **128**, 10–11
9.		Cu^{+}	N.R	HEPES buffer (50 mM, pH 7.2)	[39] *J. Am. Chem. Soc*. 2010, **132**, 5938–5939

10.		Cu^{+}	N.R	HEPES buffer (20 mM, pH 7.0)	[40] *J. Am. Chem. Soc*., 2010, **132**, 1194–1195.
11.		Cu^{+}	N.R	MOPS/K+, (10 mM, pH 7.2	[41] *J. Am. Chem. Soc.* 2011, **133**,15906–15909
12.		Cu^{+}	N.R	HEPES buffer (20 mM, pH 7.0)	[42] *Chem. Commun*., 2011, **47**, 7146–7148.
13.		Cu^{+}	4.1×10^{-13} M	PBS Buffer (25 mM, pH 7.0)	[43] *Chem. Commun*., 2012, **48**, 6247–6249.
14.		Cu^{+}	2.0×10^{-7} M	HEPES Buffer (0.10 M, pH 7.4)	[44] *Tetrahedron Lett*., 2012, **53**, 4473–4475.
15.		Cu^{+}	1.0×10^{-6} M	Water	[45] *Photochem. Photobiol. Sci*., 2014, **13**, 1427–1433.
16.		Cu^{+}	7.5×10^{-12} M	pH 6 buffer (10 mM MES, 100 nM MCL-1)	[46] *Chem. Sci.*, 2016, **7**, 1468-1473
17.		Cu^{+}	N.R	Thiourea-buffered	[47] *ACS Chem. Biol.* 2018, **13**, 1844–1852
18.		Cu^{+}	8.2×10^{-9} M	PBS (20 mM, pH 7.0)	[48] *ACS Sens*., 2019, **4**, 856–864.
19.		Cu^{+}	2.0×10^{-8} M	PBS Buffer (10 mM, pH 7.0)	[49] *ACS Sens.* 2024, 9, 1419–1427
20.		Cu^{+}	1.48×10^{-8} M	Water	Present work
***N.R. represents LOD not reported**					

S13. Reversibility experiments of L with Cu^{+}

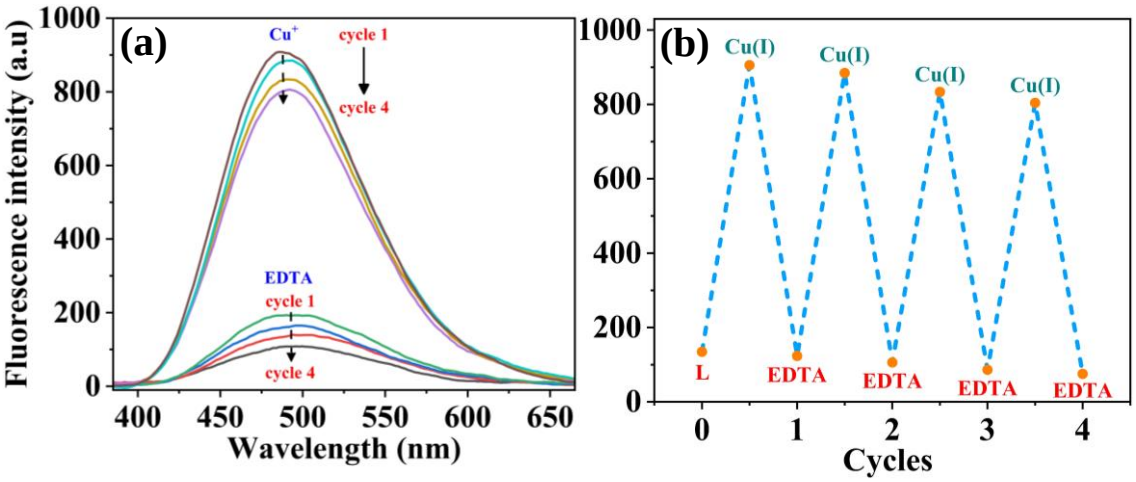


Fig. S13. (a) Fluorescence switching plot of **L** (n=4) after repeated additions of Cu^{+} followed by EDTA showing the reversibility of **L** (b) Reversible fluorescence up to four cycles of **L** with Cu^{+} (n=4).

S14. Optimized structure of L and {L+Cu^{+}}

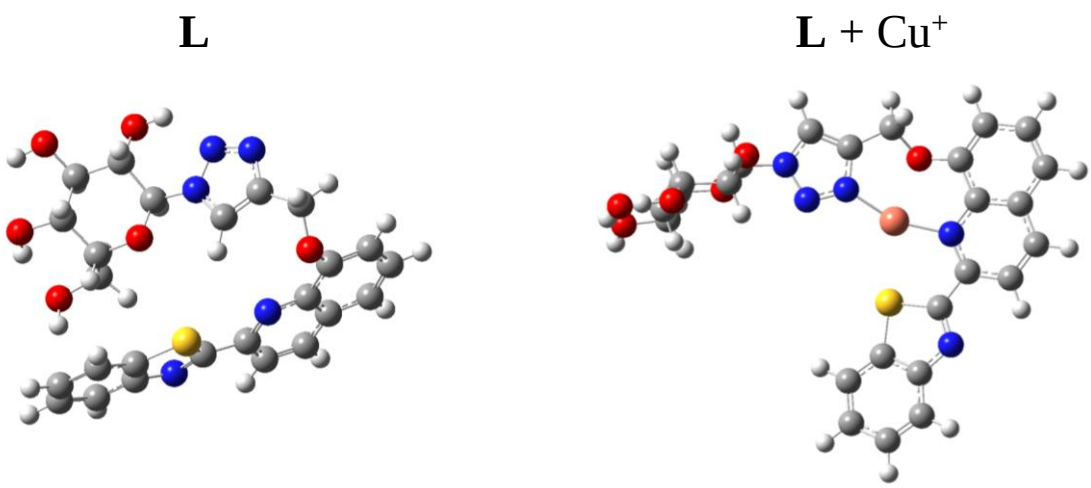


Fig. S14. Theoretical optimized structure of **L** and **{L+Cu^{+}}**

Table S 01. Cartesian coordinates of optimized L

Z	Coordinates			Z	Coordinates		
	X	Y	X		X	Y	Z
6	-1.98537	3.353833	-0.261477	1	3.372344	2.236552	2.968243
6	-1.00661	2.171346	-0.364928	8	3.73345	0.667185	1.76665
6	-2.70633	0.505563	-0.077015	6	4.331172	0.235729	0.620983
6	-3.75362	1.609585	0.079694	6	3.671765	-0.782799	-0.135165
6	-3.36584	2.875512	-0.684708	6	5.552688	0.706802	0.208751
1	-0.83092	1.969984	-1.430591	6	4.298129	-1.272246	-1.314017
1	-2.03484	3.675495	0.788766	6	6.164744	0.20606	-0.959069
1	-2.91865	-0.2876	0.646316	1	6.056358	1.465518	0.797125
1	-3.79111	1.881099	1.141289	6	1.836555	-2.121246	-0.462175
1	-3.32876	2.661	-1.765652	6	3.597205	-2.249605	-2.06335
8	-1.43384	1.015054	0.313662	6	5.557025	-0.764079	-1.708778
8	-1.58106	4.387103	-1.116895	1	7.130978	0.599474	-1.256147
1	-0.71984	4.695838	-0.789909	6	2.366046	-2.671617	-1.655199
8	-4.30977	3.881532	-0.418257	1	4.049353	-2.645151	-2.967818
1	-5.17187	3.449971	-0.508742	1	6.023559	-1.148198	-2.609986
8	-5.05787	1.203634	-0.284537	1	1.784782	-3.400822	-2.205018
1	-4.9773	0.53444	-0.985887	7	2.459984	-1.23089	0.274103
6	-2.65533	-0.143205	-1.465555	6	-1.5634	-2.823092	1.264316
1	-2.5479	0.598873	-2.259758	6	-1.5666	-3.284189	-0.066098
1	-1.79277	-0.816466	-1.516747	6	-2.73043	-3.841811	-0.609199
8	-3.87109	-0.829472	-1.740666	6	-3.86153	-3.930207	0.184654
1	-3.88814	-1.640065	-1.210982	6	-3.84564	-3.475188	1.512856
7	0.280002	2.490575	0.249584	6	-2.70229	-2.922509	2.064536
6	1.13087	1.650662	0.87155	6	0.494617	-2.516951	-0.003332
6	2.213338	2.439866	1.171478	1	-2.72142	-4.196327	-1.633951
1	0.926045	0.608197	1.037411	1	-4.7693	-4.366286	-0.218778
7	0.802078	3.724581	0.164501	1	-4.74296	-3.557646	2.11633
7	1.973661	3.697162	0.723331	1	-2.69655	-2.569294	3.089759
6	3.467963	2.062741	1.893646	7	-0.38025	-3.102033	-0.752359
1	4.298713	2.67443	1.529172	16	-0.00823	-2.137049	1.638587

Table S 02. Cartesian coordinates of optimized {L+Cu^{+}}

Z	Coordinates			Z	Coordinates		
	X	Y	X		X	Y	Z
6	5.562101	-0.02528	-1.00067	1	-2.9532	-5.07694	-0.76039
6	5.995367	0.53164	0.361971	6	-1.90957	3.72889	-0.27104
6	5.645415	-0.46792	1.450803	6	-3.14142	3.971464	0.36523
1	6.189418	-0.89828	-1.23314	6	-3.48848	5.273292	0.740823
1	6.237744	-1.37967	1.268324	6	-2.60227	6.296967	0.471793
1	5.445535	1.467594	0.551425	6	-1.37781	6.042246	-0.16445

8	4.181188	-0.40246	-0.97981	6	-1.0156	4.760623	-0.54255
7	2.448804	-1.65578	-0.1371	6	-3.34578	1.791197	0.1287
6	1.884187	-2.86218	-0.37644	1	-4.43817	5.450675	1.231709
6	0.538986	-2.62192	-0.35295	1	-2.85293	7.313189	0.754403
1	2.466057	-3.75625	-0.53041	1	-0.70085	6.865542	-0.36456
7	1.541046	-0.71116	0.025751	1	-0.06841	4.570474	-1.03385
7	0.381121	-1.28938	-0.10299	16	-1.76652	2.024776	-0.62308
6	-0.65361	-3.52194	-0.48799	7	-3.9263	2.85369	0.567433
1	-0.46561	-4.31152	-1.22116	6	5.668794	0.995651	-2.12208
1	-0.89935	-3.98918	0.472147	1	5.39336	0.52339	-3.07235
8	-1.73786	-2.69349	-0.90615	1	4.95608	1.806235	-1.9206
6	-3.01218	-2.96696	-0.48057	8	7.006563	1.464782	-2.13758
6	-3.79576	-1.83664	-0.11099	1	7.104668	2.141468	-2.8162
6	-5.15417	-2.03193	0.224956	8	7.385465	0.718394	0.446237
6	-4.89965	-4.41089	-0.08417	1	7.654589	1.248697	-0.32041
6	-3.9713	0.464808	0.183237	8	5.895575	0.031964	2.741255
6	-5.91662	-0.88036	0.544079	1	6.818478	0.320539	2.761981
6	-5.68977	-3.34104	0.238288	8	3.857349	-1.85995	2.304259
1	-5.30859	-5.41459	-0.07403	1	4.268287	-1.62269	3.147748
6	-5.34208	0.356025	0.506192	6	3.869172	-1.34436	0.000353
1	-6.96362	-0.99516	0.80551	1	4.397133	-2.29517	-0.15749
1	-6.73117	-3.4801	0.506806	6	4.174901	-0.83197	1.401562
1	-5.8892	1.263882	0.723357	1	3.572544	0.069076	1.580997
7	-3.21576	-0.59286	-0.08251	29	-1.34828	-0.60671	-0.17057
6	-3.55239	-4.22419	-0.46138				

S15. Effect of pH on the fluorescence emission of L and {L+Cu$^+$}

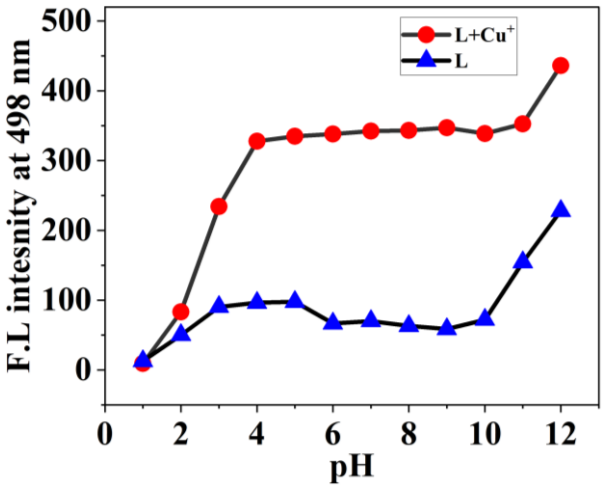


Fig. S15. Fluorescence response of (a) Free **L** (10 µM), (b) in the presence of Cu^+ (1 equiv.), and (c) fluorescence emission at 498 *vs.* pH range from 1-12.

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3. J. Hu and C.-y. Zhang, Quantum Yield Limits for the Detection of Single-Molecule Fluorescence Enhancement by a Gold Nanorod, Anal. Chem., 2013,85, 2000–2004.