*Supporting information for*

Selective Catecholamine Detection in Living Cells by a Copper-based Oxidative Reactivity

Ka Yan Tong,[a,†] Jia Zhao,[b,†] Chun-Wai Tse,[a] Pui-Ki Wan,[a] Jianhui Rong,[b] and Ho Yu Au-Yeung[a,*]

[a] State Key Laboratory for Synthetic Chemistry and Department of Chemistry, The University of Hong Kong, Pokfulam Road, Hong Kong, P. R. China. [b]School of Chinese Medicine, The University of Hong Kong, 10 Sassoon Road, Pokfulam, Hong Kong, P. R. China.

hoyuay@hku.hk
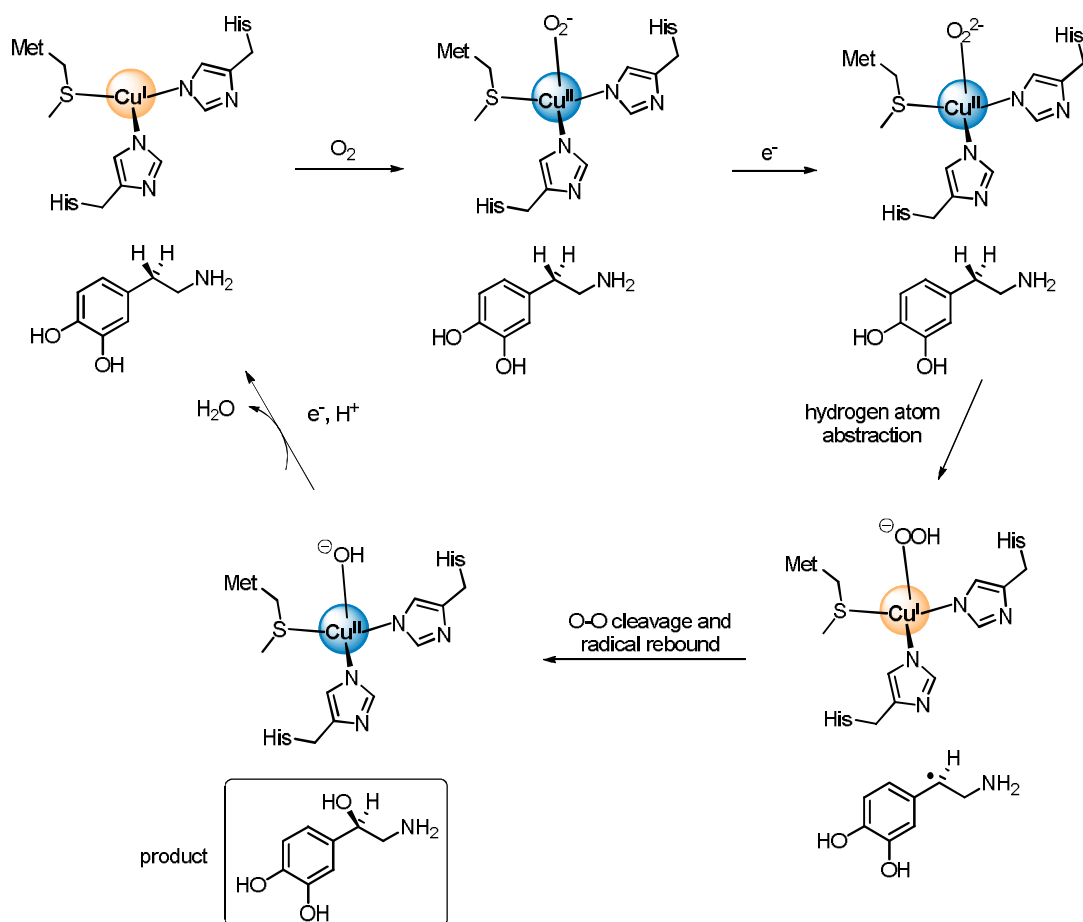
Scheme S1. Proposed mechanism of dopamine β-hydroxylase. A key Cu(II)-peroxo, formed from dioxygenation of the Cu(I) and electron transfer, is proposed to abstract a hydrogen atom from the substrate and result in a radical species for hydroxylation.[1] A similar mechanism maybe involved in the oxidative cleavage of CAP that the ether CH_2 group could be oxidized to a hemiacetal or an ester which upon hydrolysis will release the coumarin fluorophore in its phenolate form to give a fluorescent enhancement.

Synthesis

General. All solvents were of reagent grade. All commercially available chemicals were used as received. 6-(Chloromethyl)-*N*,*N*-bis(2-pyridinylmethyl)-2-pyridinemethanamine,[2] 3-(2-benzothiazolyl)-7-hydroxycoumarin,[3] 6-(hydroxymethyl)-2-pyridinecarboxaldehyde,[4] *O,O'*-di(tert-butyl-dimethylsilyl)dopamine (**TBS-DA**),[5] hydrogen peroxide probe **PC1**,[6] superoxide probe **SOP-orange**,[7] the general ROS probe H_2DCF,[8] 3-methoxytyramine[9] and *N*-methyltyramine[10] were synthesized according to literature procedures. LCMS analyses were carried out using a Waters-Alliance e2695 system coupled to a 2489 UV/Vis detector and an ACQUITY QDa MS detector. ^{1}H and ^{13}C{^{1}H} NMR spectra were obtained from a Brucker DPX 300, Brucker DPX 400, or Brucker DPX 500 spectrometers. Signals were internally referenced to solvent residues. HRMS data was obtained from a Waters Micromass Q-ToF Premier quadrupole time-of-flight tandem mass spectrometer. UV-Vis spectra were recorded by a Hitachi UH5300 UV-Vis spectrophotometer.
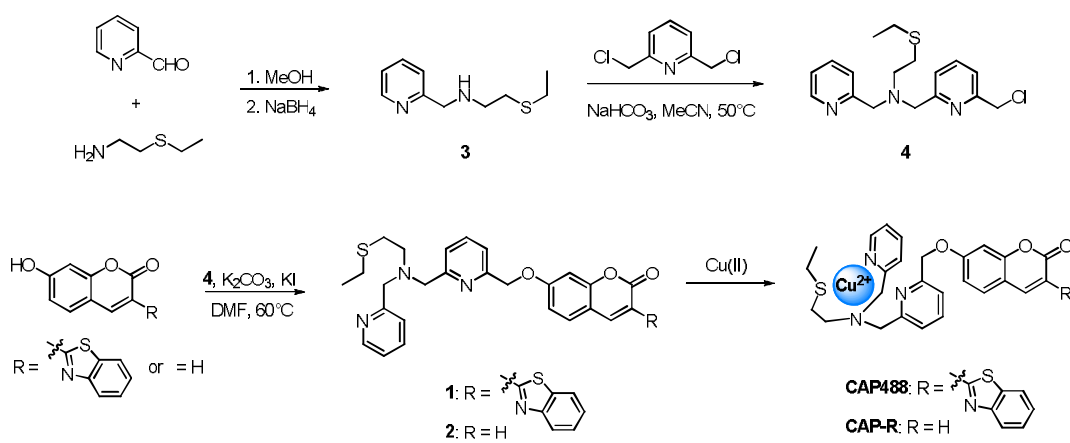
Scheme S2. Synthesis of **CAP488 and CAP-R**.

Synthesis of 3. A mixture of 2-pyridinecarboxaldehyde (0.48 mL, 5 mmol) and 2-(ethylthio)ethylamine (0.56 mL, 5 mmol) in 20 mL MeOH was stirred at room temperature for 2 hours. The mixture was cooled in an ice bath and $NaBH_4$ (0.38 g, 10 mmol) was added in small portions. The mixture was warmed to room temperature and stirred for an additional hour. Solvents were removed and the residue was re-dissolved in CH_2Cl_2 (20 mL) and washed with 0.1 M NaOH (20 mL) and brine. The organic layer was collected and dried over $MgSO_4$. Solvents were removed by a rotary evaporator to give **3** which was used in the next step without further purification. Yield = 1.08 g, quant. ^{1}H NMR (400 MHz, CDCl$_3$, 298 K) δ (ppm): 8.10 (d, *J* = 4.3 Hz, 1H), 7.20 (t, *J* = 7.7 Hz, 1H), 6.91 (d, *J* = 7.7 Hz, 1H), 6.75–6.69 (m, 1H), 3.49 (s, 2H), 2.64 (s, 1H), 2.41 (t, *J*

= 6.5 Hz, 2H), 2.28 (t, J = 6.7 Hz, 2H), 2.08 (q, J = 7.4 Hz, 2H), 0.79 (t, J = 7.4 Hz, 3H). ^{13}C{^{1}H} NMR (101 MHz, CDCl$_3$, 298 K) δ (ppm): 158.8, 148.3, 135.7, 121.4, 121.1, 53.8, 47.3, 30.9, 24.8, 14.0. ESI-MS (+ve): calcd. for C$_{10}$H$_{17}$N$_2$S [M+H]$^+$ m/z 197.1, found 197.1.

Synthesis of 4. A mixture of **3** (0.42 g, 2.12 mmol), 2,6-dichloromethylpyridine (1.49 g, 8.47 mmol) and NaHCO$_3$ (0.18 g, 2.12 mmol) in 60 mL MeCN was heated at 50 °C for overnight. Insoluble materials were removed by filtration. The filtrate was concentrated and purified by a silica column (4:1 hexane/ethyl acetate → 100% ethyl acetate). The product was obtained as a yellow oil. Yield = 0.41 g, 58%. ^{1}H NMR (400 MHz, CDCl$_3$, 298 K) δ (ppm): 8.46 (d, J = 4.8 Hz, 1H), 7.67–7.59 (m, 2H), 7.51 (t, J = 8.0 Hz, 2H), 7.28 (d, J = 7.6 Hz, 1H), 7.12–7.08 (m, 1H), 4.59 (s, 2H), 3.81 (s, 4H), 2.78–2.74 (t, 2H), 2.68–2.63 (t, 2H), 2.40 (q, J = 7.4 Hz, 2H), 1.14 (t, J = 7.4 Hz, 3H). ^{13}C{^{1}H} NMR (75 MHz, CDCl$_3$, 298 K) δ (ppm): 159.3, 159.1, 155.6, 148.8, 137.4, 136.4, 122.9, 122.2, 122.0, 121.0, 60.1, 59.8, 53.8, 46.6, 29.0, 25.9, 14.7. ESI-MS (+ve): calcd. for C$_{17}$H$_{23}$N$_3$S^{35}Cl [M+H]$^+$ m/z 336.1, found 336.2.

Synthesis of 1. A mixture of **4** (0.32 g, 0.94 mmol), 3-(2-benzothiazolyl)-7-hydroxycoumarin (0.28 g, 0.94 mmol), K$_2$CO$_3$ (0.65 g, 4.70 mmol) and KI (0.16 g, 0.94 mmol) in 50 mL DMF was heated at 60 °C for overnight. Insoluble materials were removed by filtration, and the filtrate was purified by a silica column (1:1 hexane/ethyl acetate → 100% ethyl acetate → 50:1 ethyl acetate/MeOH) to yield the ligand-fluorophore conjugate **1** as a yellow solid. Yield = 0.22 g, 39%. ^{1}H NMR (400 MHz, CDCl$_3$, 298 K) δ (ppm): 9.00 (s, 1H), 8.52 (dd, J = 4.9, 0.8 Hz, 1H), 8.05 (d, J = 8.1 Hz, 1H), 7.96 (d, J = 7.7 Hz, 1H), 7.72 (t, J = 7.7 Hz, 1H), 7.67 (td, J = 7.7, 1.8 Hz, 1H), 7.62 (d, J = 8.7 Hz, 1H), 7.58 (d, J = 7.8 Hz, 2H), 7.53–7.49 (m, 1H), 7.42–7.38 (m, 1H), 7.35 (d, J = 7.6 Hz, 1H), 7.18–7.14 (m, 1H), 7.04 (dd, J = 8.7, 2.4 Hz, 1H), 6.98 (d, J = 2.3 Hz, 1H), 5.26 (s, 2H), 3.90 (s, 2H), 3.88 (s, 2H), 2.85–2.80 (t, 2H), 2.75–2.72 (t, 2H), 2.46 (q, J = 7.4 Hz, 2H), 1.20 (t, J = 7.4 Hz, 3H). ^{13}C{^{1}H} NMR (101 MHz, CDCl$_3$, 298 K) δ (ppm):163.1, 160.5, 160.3, 159.8, 159.6, 156.0, 154.9, 152.6, 149.2, 141.7, 137.6, 136.7, 136.7, 130.8, 126.5, 125.3, 123.1, 122.8, 122.5, 122.3, 121.9, 119.9, 117.2, 114.4, 113.2, 102.06, 71.5, 60.4, 60.3, 54.1, 29.4, 26.2, 15.0. ESI-MS (+ve): calcd. for C$_{33}$H$_{31}$N$_4$O$_3$S$_2$ [M+H]$^+$ m/z 595.2, found 595.1.

Synthesis of 2. A mixture of **4** (0.12 g, 0.37 mmol), 7-hydroxycoumarin (0.06 g, 0.37 mmol), K$_2$CO$_3$ (0.26 g, 1.85 mmol) and KI (0.06 g, 0.37 mmol) in 50 mL DMF was heated at 60 °C for overnight. Insoluble materials were removed by filtration, and the filtrate was purified by a silica column (2:1 hexane/ethyl acetate → 100% ethyl acetate → 9:1 ethyl acetate/MeOH) to yield the ligand-fluorophore conjugate **2** as a brown oil. Yield = 0.14 g, 80%. ^{1}H NMR (500 MHz, CDCl$_3$, 298 K) δ (ppm): 8.52 (d, J = 4.2 Hz, 1H), 7.70 (t, J = 7.7 Hz, 1H), 7.66 (td, J = 7.7, 1.8 Hz, 1H), 7.62 (d, J = 9.5 Hz, 1H), 7.56 (t, J = 7.3 Hz, 2H), 7.37 (d, J = 8.6 Hz, 1H), 7.34 (d, J = 7.6 Hz, 1H), 7.17–7.13 (m, 1H),

6.93 (dd, J = 8.6, 2.4 Hz, 1H), 6.88 (d, J = 2.4 Hz, 1H), 6.24 (d, J = 9.5 Hz, 1H), 5.21 (s, 2H), 3.89 (s, 2H), 3.88 (s, 2H), 2.82 (t, J = 7.2 Hz, 2H), 2.71 (t, J = 8.2 Hz, 2H), 2.45 (q, J = 7.4 Hz, 2H), 1.18 (t, J = 7.4 Hz, 3H). $^{13}C\{^1H\}$ NMR (126 MHz, CDCl$_3$, 298K) δ (ppm): 161.7, 161.2, 159.6, 159.4, 155.9, 155.2, 149.1, 143.4, 137.5, 136.6, 129.0, 123.1, 122.3, 122.2, 119.8, 113.5, 113.0, 102.4, 76.9, 71.3, 60.3, 60.2, 54.0, 29.3, 26.2, 14.9. ESI-MS (+ve): calcd. for C$_{26}$H$_{28}$N$_3$O$_3$S [M+H]$^+$ m/z 462.2, found 462.2.

Synthesis of CAP488. A solution of CuCl$_2$·2H$_2$O (20.6 mg, 0.12 mmol) in 200 μL MeOH was added to a solution of **1** (72 mg, 0.12 mmol) in 1 mL CH$_2$Cl$_2$. Diethyl ether vapor was slowly diffused into the solution and a green precipitate was obtained. The green precipitate was collected and washed with diethyl ether. Yield = 57 mg, 65%. HRMS (ESI, +ve): calcd. for C$_{33}$H$_{30}$N$_4$O$_3$S$_2$Cl^{63}Cu [M+Cl]$^+$ m/z 692.0744, found 692.0698; calcd. for C$_{33}$H$_{30}$N$_4$O$_3$S$_2$Cl^{65}Cu [M+Cl]$^+$ m/z 694.0726, found 694.0624. UV-Vis (0.05 mM, MeOH) λ$_{max,}$ (ε): 261 nm (22,000 M^{-1} cm^{-1}), 380 nm (37,000 M^{-1} cm^{-1}), 682 nm (200 M^{-1} cm^{-1}).

Synthesis of CAP-R. A solution of CuCl$_2$·2H$_2$O (11 mg, 0.07 mmol) in 200 μL MeOH was added to a solution of **2** (31 mg, 0.07 mmol) in 800 μL MeOH. Diethyl ether vapour was slowly diffused into the solution and green crystals were obtained. The green crystals were collected and washed with diethyl ether. Yield = 39 mg, 84%. HRMS (ESI, +ve): calcd. for C$_{26}$H$_{27}$N$_3$O$_3$S^{63}CuCl [M+Cl]$^+$ m/z 559.0757, found 559.0782; calcd. for C$_{26}$H$_{27}$N$_3$O$_3$S^{65}CuCl [M+Cl]$^+$ m/z 561.0739, found 561.0781. UV-Vis (0.05 mM, MeOH) λ$_{max}$ (ε): 269 nm (900 M^{-1} cm^{-1}), 320 nm (15,000 M^{-1} cm^{-1}), 670 nm (200 M^{-1} cm^{-1}).
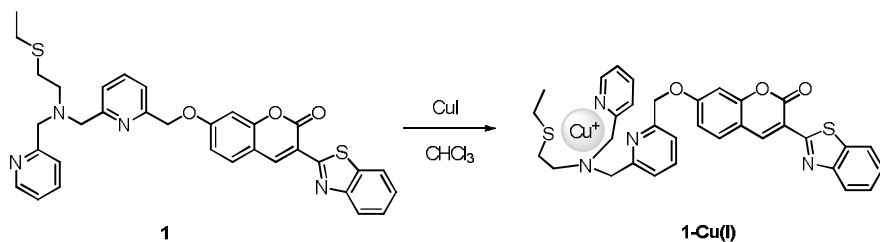
Scheme S3. Synthesis of **1-Cu(I)**.

Synthesis of 1-Cu(I). In a glove box, **1** (12 mg, 0.02 mmol) in 0.5 mL CHCl$_3$ was added to solid CuI (3.8 mg, 0.02 mmol). The solution was stirred for 1 hour and passed through a syringe filter. Hexane (1 mL) was then added to the solution after which a yellow solid precipitated from the solution. The yellow solid was washed with diethyl ether and dried under vacuum. Yield = 11 mg, 83%. ^{1}H NMR (500 MHz, CDCl$_3$, 298 K) δ (ppm): 8.98 (s, 1H), 8.77 (d, J = 3.9 Hz, 1H), 8.05 (d, J = 8.1 Hz, 1H), 7.95

(d, J = 7.9 Hz, 1H), 7.78–7.70 (m, 2H), 7.64 (d, J = 8.6 Hz, 1H), 7.50 (t, J = 7.2 Hz, 2H), 7.42–7.31 (m, 3H), 7.24 (s, 1H), 7.16 (d, J = 7.8 Hz, 1H), 6.96 (s, 1H), 5.75 (s, 2H), 4.10 (s, 2H), 3.95 (s, 2H), 3.02 (t, J = 7.0 Hz, 2H), 2.98 (t, J = 7.0 Hz, 2H), 2.61 (q, J = 7.1 Hz, 2H), 1.26 (t, J = 7.2 Hz, 3H). ^{13}C{^{1}H} NMR (126 MHz, CDCl$_3$, 298 K) δ (ppm): 162.6, 160.4, 160.2, 156.0, 155.8, 152.6, 152.6, 149.8, 141.7, 138.1, 137.6, 136.7, 131.0, 126.5, 125.2, 124.2, 123.9, 122.8, 121.8, 121.5, 120.2, 117.3, 114.0, 113.4, 102.7, 71.7, 59.9, 59.4, 54.8, 29.8, 28.2, 14.7. ESI-MS (+ve): calcd. for C$_{33}$H$_{30}$N$_4$O$_3$S$_2$^{63}Cu [M]$^+$ m/z 657.1050, found 657.1045; calcd for C$_{33}$H$_{30}$N$_4$O$_3$S$_2$^{65}Cu [M]$^+$ m/z 659.1038, found 659.1030.
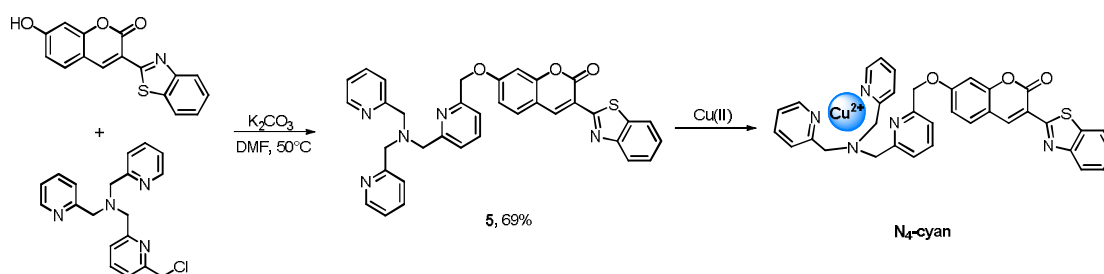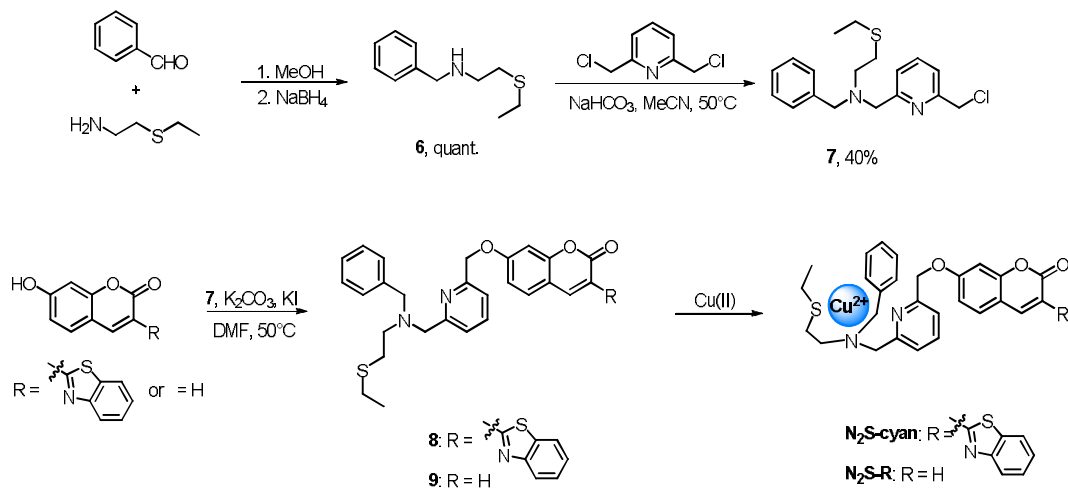

Scheme S4. Synthesis of **N$_4$-cyan**.

Synthesis of 5. A mixture of 6-(chloromethyl)-*N*,*N*-bis(2-pyridinylmethyl)-2-pyridinemethanamine (0.11 g, 0.33 mmol), 3-(2-benzothiazolyl)-7-hydroxycoumarin (0.10 g, 0.33 mmol) and K$_2$CO$_3$ (0.27 g, 1.95 mmol) in 50 mL DMF was heated at 50 °C for overnight. Insoluble materials were removed by filtration, and the filtrate was purified by a silica column (1:2 hexane/ethyl acetate → 100% ethyl acetate → 25:1 ethyl acetate/MeOH) to yield **5** as a yellow solid. Yield = 0.13 g, 69%. ^{1}H NMR (500 MHz, CDCl$_3$, 298 K) δ (ppm): 8.96 (s, 1H), 8.55 (d, J = 4.6 Hz, 2H), 8.03 (d, J = 8.2 Hz, 1H), 7.93 (d, J = 7.9 Hz, 1H), 7.71–7.66 (m, 3H), 7.59 (d, J = 8.6 Hz, 3H), 7.54 (d, J = 7.7 Hz, 1H), 7.49 (t, J = 7.6 Hz, 1H), 7.39–7.34 (m, 2H), 7.19–7.16 (m, 2H), 7.02 (d, J = 8.7 Hz, 1H), 6.95 (s, 1H), 5.26 (s, 2H), 4.05 (s, 6H). ^{13}C{^{1}H} NMR (126 MHz, CDCl$_3$, 298 K) δ (ppm): 163.0, 160.4, 160.2, 158.1, 157.9, 155.9, 155.2, 152.5, 149.0, 141.6, 141.5, 137.7, 137.1, 136.7, 130.7, 126.5, 125.2, 123.5, 122.6, 121.8, 120.2, 117.1, 114.3, 113.1, 102.0, 101.9, 71.4, 59.8. ESI-MS (+ve): calcd. for C$_{35}$H$_{28}$N$_5$O$_3$S [M+H]$^+$ m/z 598.2, found 598.2.

Synthesis of N$_4$-cyan. A solution of CuCl$_2$·2H$_2$O (3 mg, 0.02 mmol) in 200 µL MeOH was added to a solution of **10** (10 mg, 0.02 mmol) in 400 µL CH$_2$Cl$_2$ and mixed well. Vapour of diethyl ether was slowly diffused into the solution and a green precipitate was collected and washed with diethyl ether. Yield = 10 mg, 84%. ESI-MS (+ve): calcd. for C$_{35}$H$_{27}$N$_5$O$_3$SCl^{63}Cu [M+Cl]$^+$ m/z 695.1, found 695.2. UV-Vis (0.05 mM, MeOH) λ$_{max,}$ (ε): 260 nm (26,000 M^{-1} cm^{-1}), 379 nm (35,000 M^{-1} cm^{-1}), 679

nm (400 M^{-1} cm^{-1}).

Scheme S5. Synthesis of **N₂S-cyan** and **N₂S-R**.

Synthesis of 6. A mixture of benzaldehyde (0.46 mL, 4.5 mmol) and 2-(ethylthio)ethylamine (0.50 mL, 4.5 mmol) in 20 mL MeOH was stirred at room temperature for 2 hours. The mixture was cooled in an ice bath and $NaBH_4$ (0.34 g, 9 mmol) was added in small portions. The mixture was warmed to room temperature and stirred for an additional hour. Solvents were removed and the residue was re-dissolved in CH_2Cl_2 (20 mL) and washed with sat. K_2CO_3 (20 mL) and brine. The organic layer was collected and dried over $MgSO_4$. Solvents were removed by a rotary evaporator to give **6** which was used in the next step without further purification. Yield = 0.57 g, quant. ^{1}H NMR (500 MHz, $CDCl_3$, 298 K) δ (ppm): 7.31 (d, J = 6.4 Hz, 4H), 7.26–7.23 (m, 1H), 3.81 (s, 2H), 2.82 (t, J = 6.5 Hz, 2H), 2.71 (t, J = 6.4 Hz, 2H), 2.50 (q, J = 7.4 Hz, 2H), 1.23 (t, J = 7.4 Hz, 3H). ^{13}C{^{1}H} NMR (126 MHz, $CDCl_3$, 298 K) δ (ppm): 140.1, 128.5, 128.2, 127.1, 53.6, 47.9, 32.0, 25.8, 14.9. ESI-MS (+ve): calcd. for $C_{11}H_{18}NS$ [M+H]$^+$ m/z 196.1, found 196.2.

Synthesis of 7. A mixture of **6** (0.13 g, 0.67 mmol), 2,6-dichloromethylpyridine (0.47 g, 2.67 mmol) and $NaHCO_3$ (0.06 g, 0.67 mmol) in 50 mL MeCN was heated at 50 °C for overnight. Insoluble materials were removed by filtration. The filtrate was concentrated and purified by a basic alumina column (4:1 hexane/ethyl acetate). The product was obtained as a yellow oil. Yield = 0.09 g, 40%. ^{1}H NMR (500 MHz, $CDCl_3$, 298 K) δ (ppm): 7.69 (t, J = 8.9 Hz, 1H), 7.56 (d, J = 7.7 Hz, 1H), 7.38 (d, J = 7.4 Hz, 2H), 7.31 (t, J = 7.8 Hz, 3H), 7.23 (t, J = 7.0 Hz, 1H), 4.63 (s, 2H), 3.79 (s, 2H), 3.68 (s, 2H), 2.74 (t, J = 7.1 Hz, 2H), 2.68 (t, J = 7.1 Hz, 2H), 2.43 (q, J = 7.4 Hz, 2H), 1.18 (t, J = 7.4 Hz, 3H). ^{13}C{^{1}H} NMR (126 MHz, $CDCl_3$, 298 K) δ (ppm): ^{13}C NMR (126 MHz, $CDCl_3$) δ160.2, 155.7, 139.2, 137.5,

128.9, 128.4, 127.2, 122.3, 121.1, 60.0, 58.7, 53.7, 46.9, 29.4, 26.1, 14.9. ESI-MS (+ve): calcd. for $C_{18}H_{24}N_2S^{35}Cl$ [M+H]$^+$ m/z 335.1, found 335.0.

Synthesis of 8. A mixture of **7** (0.09 g, 0.27 mmol), 3-(2-benzothiazolyl)-7-hydroxycoumarin (0.08 g, 0.27 mmol), K_2CO_3 (0.19 g, 1.34 mmol) and KI (0.04 g, 0.27 mmol) in 30 mL DMF was heated at 50 °C for overnight. Insoluble materials were removed by filtration, and the filtrate was purified by a silica column (4:1 hexane/ethyl acetate) to yield **8** as a yellow solid. Yield = 0.04 g, 23%. ^{1}H NMR (500 MHz, CDCl$_3$, 298 K) δ (ppm): 9.00 (s, 1H), 8.06 (d, J = 8.1 Hz, 1H), 7.96 (d, J = 7.9 Hz, 1H), 7.72 (t, J = 7.7 Hz, 1H), 7.63–7.59 (m, 2H), 7.52 (t, J = 7.2 Hz, 1H), 7.42–7.38 (m, 3H), 7.37–7.29 (m, 4H), 7.04 (dd, J = 8.7, 2.3 Hz, 1H), 6.99 (d, J = 2.2 Hz, 1H), 5.26 (s, 2H), 3.83 (s, 2H), 3.70 (s, 2H), 2.76 (d, J = 7.6 Hz, 2H), 2.69 (d, J = 7.3 Hz, 2H), 2.44 (q, J = 7.4 Hz, 2H), 1.19 (t, J = 7.4 Hz, 3H). ^{13}C{^{1}H} NMR (126 MHz, CDCl$_3$, 298K) δ (ppm): 163.1, 160.5, 160.3, 156.0, 154.8, 152.6, 141.7, 139.1, 137.6, 136.8, 130.8, 129.0, 128.5, 127.3, 126.5, 125.3, 122.8, 122.4, 121.9, 119.9, 117.2, 114.4, 113.3, 102.1, 99.0, 71.6, 60.2, 58.8, 53.6, 29.8, 26.2, 14.9. ESI-MS (+ve): calcd. for $C_{27}H_{29}N_2O_3S$ [M+H]$^+$ m/z 461.6, found 461.4.

Synthesis of 9. A mixture of **7** (0.08 g, 0.25 mmol), 7-hydroxycoumarin (0.04 g, 0.26 mmol), K_2CO_3 (0.17 g, 1.25 mmol) and KI (0.04 g, 0.25 mmol) in 50 mL DMF was heated at 50 °C for overnight. Insoluble materials were removed by filtration, and the filtrate was purified by a basic alumina column (2:1 hexane/ethyl acetate) to yield the ligand-fluorophore conjugate **9** as a yellow oil. Yield = 0.11 g, 93%. ^{1}H NMR (500 MHz, CDCl$_3$, 298 K) δ (ppm): ^{1}H NMR (500 MHz, CDCl$_3$, 298K) δ (ppm): 7.70 (t, J = 7.7 Hz, 1H), 7.59 (dd, J = 18.5, 8.6 Hz, 2H), 7.41 – 7.28 (m, 6H), 7.23 (t, J = 7.3 Hz, 1H), 6.94 – 6.88 (m, 2H), 6.28 – 6.20 (m, 1H), 5.21 (s, 2H), 3.81 (s, 2H), 3.68 (s, 2H), 2.75 (t, J = 7.4 Hz, 2H), 2.68 (t, J = 7.4 Hz, 2H), 2.43 (q, J = 7.4 Hz, 2H), 1.17 (t, J = 7.4 Hz, 3H). ^{13}C{^{1}H} NMR (126 MHz, CDCl$_3$, 298K) δ (ppm): 161.7, 161.2, 160.2, 155.9, 155.0, 143.4, 139.1, 137.4, 128.9, 128.9, 128.4, 127.2, 122.2, 119.7, 113.4, 113.0, 113.0, 102.3, 71.3, 60.0, 58.7, 29.4, 26.1, 14.9. ESI-MS (+ve): calcd. for $C_{27}H_{29}N_2O_3S$ [M+H]$^+$ m/z 461.2, found 461.4.

Synthesis of N$_2$S-cyan. A solution of CuCl$_2$·2H$_2$O (2 mg, 0.01 mmol) in 200 µL MeOH was added to a solution of **8** (9 mg, 0.01 mmol) in 200 µL CH$_2$Cl$_2$ and mixed well. Diethyl ether vapour was slowly diffused into the solution and a green precipitate was collected and washed with diethyl ether. Yield = 8 mg, 75%. ESI-MS (+ve): calcd. for $C_{34}H_{31}N_3O_3S_2Cl^{63}Cu$ [M+Cl]$^+$ m/z 691.1, found 691.0. UV-Vis (0.05 mM, MeOH) $\lambda_{max,}$ (ε): 261 nm (14,000 M^{-1} cm^{-1}), 378 nm (30,000 M^{-1} cm^{-1}), 693 nm (500 M^{-1} cm^{-1}).

Synthesis of N$_2$S-R. A solution of CuCl$_2$·2H$_2$O (4 mg, 0.02 mmol) in 200 µL MeOH was added to a

solution of **9** (10 mg, 0.02 mmol) in 200 µL MeOH and mixed well. Diethyl ether vapour was slowly diffused into the solution and a green crystal was collected. Yield = 11 mg, 82%. ESI-MS (+ve): calcd. for $C_{27}H_{28}N_2O_3SCl^{63}Cu$ [M+Cl]$^+$ m/z 558.1, found 558.0. UV-Vis (0.05 mM, MeOH) $\lambda_{max,}$ (ε): 275 nm (7,000 M^{-1} cm^{-1}), 320 nm (12,000 M^{-1} cm^{-1}), 717 nm (200 M^{-1} cm^{-1}).
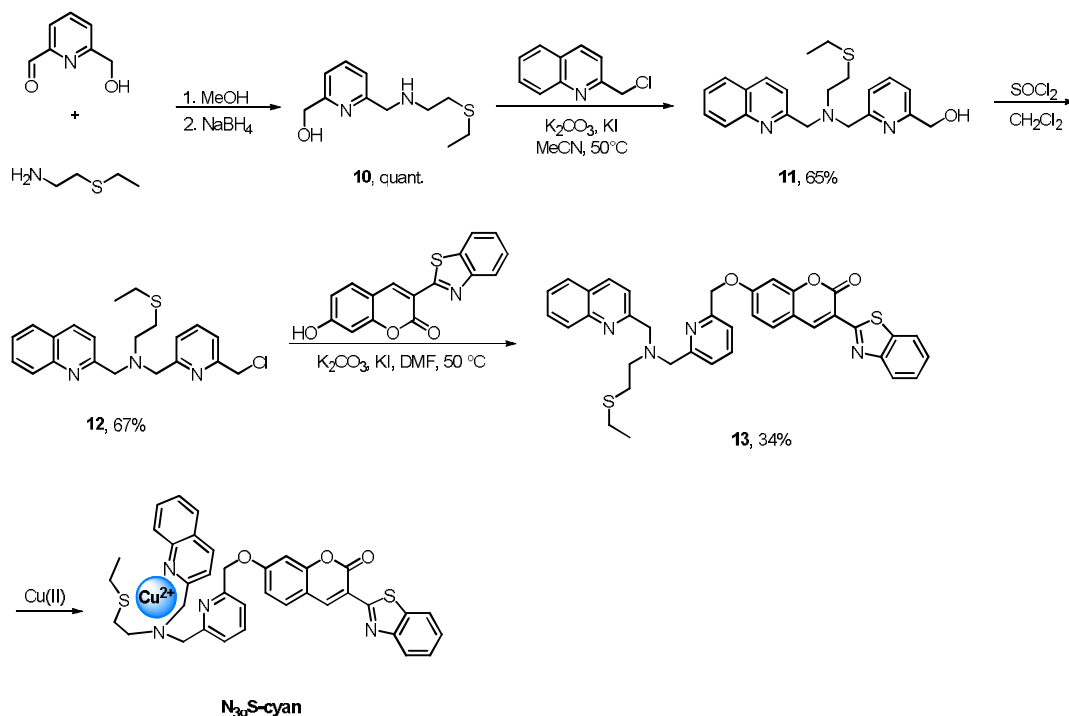
Scheme S6. Synthesis of **N$_{3q}$S-cyan**.

Synthesis of 10. A mixture of 6-(hydroxymethyl)-2-pyridinecarboxaldehyde (0.24 g, 1.76 mmol) and 2-(ethylthio)ethylamine (0.20 mL, 1.76 mmol) in 10 mL MeOH was stirred at room temperature for 4 hours. The mixture was cooled in an ice bath and NaBH$_4$ (0.13 g, 3.51 mmol) was added in small portions. The mixture was warmed to room temperature and stirred for an additional hour. Solvents were removed and the residue was re-dissolved in CH$_2$Cl$_2$ (20 mL) and washed with sat. NaHCO$_3$ (20 mL) and brine. The organic layer was collected and dried over MgSO$_4$. Solvents were removed by a rotary evaporator to give **9** which was used in the next step without further purification. Yield = 0.44 g, quant. ^{1}H NMR (400 MHz, CDCl$_3$, 298 K) δ (ppm): 7.62 (t, J = 7.7 Hz, 1H), 7.18 (d, J = 7.6 Hz, 1H), 7.11 (d, J = 7.7 Hz, 1H), 4.70 (s, 2H), 3.91 (s, 2H), 2.83 (t, J = 6.4 Hz, 2H), 2.70 (d, J = 6.4 Hz, 2H), 2.52 (q, J = 7.5 Hz, 2H), 1.23 (t, J = 7.4 Hz, 3H). ^{13}C{^{1}H} NMR (126 MHz, CDCl$_3$, 298 K) δ (ppm): 159.7, 158.1, 137.1, 120.5, 118.7, 64.2, 54.2, 47.9, 31.5, 25.6, 14.7. ESI-MS (+ve): calcd. for $C_{11}H_{19}N_2OS$ [M+H]$^+$ m/z 227.1, found 227.2.

Synthesis of 11. A mixture of **10** (0.19 g, 0.85 mmol), 2-(chloromethyl)quinoline hydrochloride (0.20 g, 0.93 mmol), K_2CO_3 (0.59 g, 4.24 mmol) and KI (0.14 g, 0.85 mmol) in 50 mL MeCN was heated at 50 °C for overnight. Insoluble materials were filtered off, and the filtrate was purified by a silica column (4:1 hexane/ethyl acetate → 100% ethyl acetate → 9:1 ethyl acetate/MeOH) to yield **11** as a yellow oil. Yield = 0.20 g, 65%. ^{1}H NMR (400 MHz, CDCl$_3$, 298 K) δ (ppm): 8.10 (d, J = 8.4 Hz, 1H), 8.02 (d, J = 8.2 Hz, 1H), 7.77–7.72 (m, 2H), 7.67–7.59 (m, 2H), 7.46 (dd, J = 17.9, 7.4 Hz, 2H), 7.08 (d, J = 7.7 Hz, 1H), 4.69 (s, 2H), 3.99 (s, 2H), 3.86 (s, 2H), 2.82 (t, J = 7.4 Hz, 2H), 2.70 (t, J = 7.4 Hz, 2H), 2.42 (q, J = 7.4 Hz, 2H), 1.15 (t, J = 7.4 Hz, 3H). ^{13}C{^{1}H} NMR (101 MHz, CDCl$_3$, 298K) δ (ppm): 160.4, 158.4, 158.2, 147.5, 137.1, 136.5, 129.5, 128.9, 127.6, 127.4, 126.2, 121.6, 121.1, 118.8, 64.0, 61.0, 59.9, 54.1, 29.4, 26.1, 14.8. ESI-MS (+ve): calcd. for $C_{21}H_{26}N_3OS$ [M+H]$^+$ m/z 368.2, found 368.3.

Synthesis of 12. To a solution of **11** (0.12 g, 0.55 mmol) in 10 ml CH$_2$Cl$_2$ was added 2 mL SOCl$_2$ and 2 drops of DMF. The mixture was stirred at room temperature for 2 hours and then neutralized with sat. NaHCO$_3$. The organic layer was washed with deionized water and brine. The organic phase was collected and dried over MgSO$_4$. Solvent was removed by a rotary evaporator to give **12** which was used in next step without further purification. Yield = 85 mg, 67%. ^{1}H NMR (500 MHz, CDCl$_3$, 298 K) δ (ppm): 8.10 (d, J = 8.5 Hz, 1H), 8.02 (d, J = 8.5 Hz, 1H), 7.78–7.72 (m, 2H), 7.69–7.64 (m, 2H), 7.54 (d, J = 7.7 Hz, 1H), 7.48 (t, J = 7.5 Hz, 1H), 7.30 (d, J = 7.6 Hz, 1H), 4.60 (s, 2H), 4.01 (s, 2H), 3.88 (s, 2H), 2.84 (t, J = 7.4 Hz, 2H), 2.71 (t, J = 7.4 Hz, 2H), 2.41 (q, J = 7.4 Hz, 2H), 1.15 (t, J = 7.4 Hz, 3H). ^{13}C{^{1}H} NMR (126 MHz, CDCl$_3$, 298K) δ (ppm): 160.3, 159.5, 155.8, 147.6, 137.5, 136.5, 129.5, 129.0, 127.6, 127.5, 126.2, 122.4, 121.1, 61.1, 60.2, 54.1, 46.8, 29.3, 26.1, 14.9. ESI-MS (+ve): calcd. for $C_{21}H_{25}N_3S^{35}Cl$ [M+H]$^+$ m/z 386.2, found 386.4.

Synthesis of 13. A mixture of **12** (85 mg, 0.22 mmol), 3-(2-benzothiazolyl)-7-hydroxycoumarin (65 mg, 0.22 mmol), K$_2$CO$_3$ (0.15 g, 1.10 mmol) and KI (37 mg, 0.22 mmol) in 30 mL DMF was heated at 50 °C for overnight. Insoluble materials were removed by filtration, and the filtrate was purified by a silica column (4:1 hexane/ethyl acetate → 3:1 hexane/ethyl acetate → 100% ethyl acetate) to yield **13** as a yellow solid. Yield = 49 mg, 34%. ^{1}H NMR (500 MHz, CDCl$_3$, 298 K) δ (ppm): 8.94 (s, 1H), 8.12 (d, J = 8.5 Hz, 1H), 8.04 (d, J = 8.3 Hz, 2H), 7.93 (d, J = 7.9 Hz, 1H), 7.77 (d, J = 7.9 Hz, 1H), 7.76–7.70 (m, 2H), 7.68 (t, J = 7.1 Hz, 1H), 7.60–7.57 (m, 2H), 7.51–7.47 (m, 2H), 7.38 (t, J = 7.5 Hz, 1H), 7.33 (d, J = 7.6 Hz, 1H), 7.00 (dd, J = 8.7, 2.3 Hz, 1H), 6.95 (d, J = 2.2 Hz, 1H), 5.23 (s, 2H), 4.04 (s, 2H), 3.94 (s, 2H), 2.87 (t, J = 7.3 Hz, 2H), 2.74 (t, J = 7.3 Hz, 2H), 2.44 (q, J = 7.4 Hz, 2H), 1.17 (t, J = 7.4 Hz, 3H). ^{13}C{^{1}H} NMR (126 MHz, CDCl$_3$, 298K) δ (ppm): 163.1, 160.4, 160.3, 160.2, 159.7, 155.9, 154.9, 152.6, 147.6, 141.6, 137.5, 136.7, 136.5, 130.7, 129.5, 129.0, 127.6, 127.5, 126.5, 126.3, 125.2, 122.8, 122.6, 121.8, 121.1, 119.9, 117.1, 114.3, 113.1, 102.0, 71.5, 61.1, 60.3, 54.2, 29.4, 26.2, 14.9. ESI-MS (+ve): calcd. for $C_{37}H_{33}N_4O_3S_2$ [M+H]$^+$ m/z 645.2, found 645.3.

Synthesis of N₃qS -cyan. A solution of CuCl$_2$·2H$_2$O (1.7 mg, 0.01 mmol) in 200 μL MeOH was added to a solution of **13** (4 mg, 0.01 mmol) in 200 μL CH$_2$Cl$_2$ and mixed well. Diethyl ether vapour was slowly diffused into the solution and a green precipitate was collected and washed with diethyl ether. Yield = 3 mg, 76%. ESI-MS (+ve): calcd. for C$_{37}$H$_{32}$N$_4$O$_3$S$_2$Cl^{63}Cu [M+Cl]$^+$ m/z 742.1, found 742.2. UV-Vis (0.05 mM, MeOH) λ$_{max,}$ (ε): 214 nm (50,000 M^{-1} cm^{-1}), 262 nm (18,000 M^{-1} cm^{-1}), 378 nm (33,000 M^{-1} cm^{-1}), 682 nm (400 M^{-1} cm^{-1}).
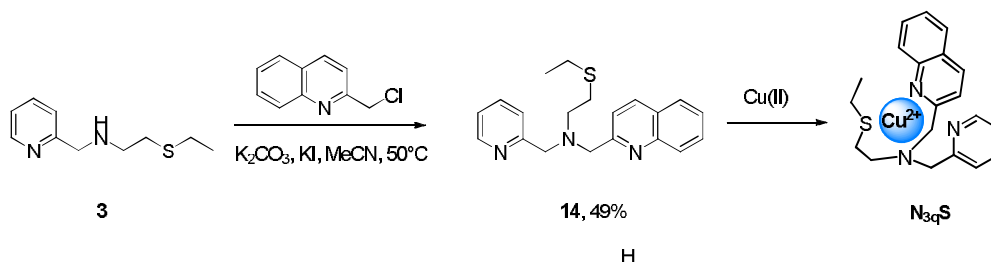
Scheme S7. Synthesis of **N₃qS**.

Synthesis of 14. A mixture of **3** (0.09 g, 0.46 mmol), 2-(chloromethyl)quinoline hydrochloride (0.13 g, 0.60 mmol), K$_2$CO$_3$ (0.32 g, 2.29 mmol) and KI (0.08 g, 0.46 mmol) in 30 mL MeCN was heated at 50 °C for overnight. Insoluble materials were removed by filtration, and the filtrate was purified by a silica column (2:1 hexane/ethyl acetate → 1:1 hexane/ethyl acetate) to yield **14** as a yellow oil. Yield = 76 mg, 49%. ^{1}H NMR (400 MHz, CDCl$_3$, 298 K) δ (ppm): 8.50 (d, J = 5.6 Hz, 1H), 8.12 (d, J = 8.5 Hz, 1H), 8.03 (d, J = 8.5 Hz, 1H), 7.81 – 7.72 (m, 2H), 7.66 (qd, J = 8.1, 1.6 Hz, 2H), 7.58 (d, J = 7.8 Hz, 1H), 7.51 – 7.45 (m, 1H), 7.16 – 7.09 (m, 1H), 4.01 (s, 2H), 3.89 (s, 2H), 2.84 (t, J = 7.6 Hz, 2H), 2.71 (t, J = 7.6 Hz, 2H), 2.41 (q, J = 7.4 Hz, 2H), 1.15 (t, J = 7.4 Hz, 3H). ^{13}C{^{1}H} NMR (101 MHz, CDCl$_3$, 298K) δ (ppm): 160.4, 159.5, 149.1, 147.6, 136.5, 129.5, 129.1, 127.6, 127.6, 127.6, 126.2, 123.2, 122.1, 121.2, 61.2, 60.5, 54.1, 29.3, 26.1, 14.9. ESI-MS (+ve): calcd. for C$_{20}$H$_{24}$N$_3$S [M+H]$^+$ m/z 338.1, found 338.1.

Synthesis of N₃qS. A solution of **14** (9 mg, 0.03 mmol) in 300 μL MeCN was added to solid CuCl$_2$·2H$_2$O (4.5 mg, 0.03 mmol) and stirred for 30 mins. Diethyl ether vapour was slowly diffused into the solution and green crystals were collected. Yield = 8 mg, 64%. ESI-MS (+ve): calcd. for C$_{20}$H$_{23}$N$_3$SCl^{63}Cu [M+Cl]$^+$ m/z 435.1, found 435.0. UV-Vis (0.05 mM, MeOH) λ$_{max,}$ (ε): 264 nm (5,000 M^{-1} cm^{-1}), 304 nm (5,000 M^{-1} cm^{-1}), 670 nm (60 M^{-1} cm^{-1}).
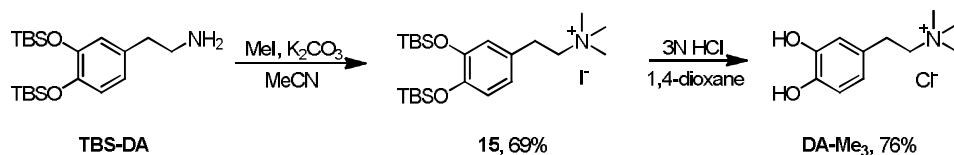
Scheme S8. Synthesis of **DA-Me₃**.

Synthesis of 15. A mixture of *O,O'*-di(tert-butyl-dimethylsilyl)dopamine (**TBS-DA**) (96 mg, 0.25 mmol), MeI (0.08 ml, 1.25 mmol) and K_2CO_3 (0.17 g, 1.25 mmol) in 30 mL MeCN was heat to reflux for overnight. Insoluble materials were removed by filtration. Solvent was removed by a rotary evaporator. The residue was recrystallized in CH_2Cl_2/hexane to yield **15** as a white solid. Yield = 95 mg, 69%. ^{1}H NMR (500 MHz, $CDCl_3$, 298 K) δ (ppm): 6.80 (dd, *J* = 8.2, 2.2 Hz, 1H), 6.74 (d, *J* = 8.2 Hz, 1H), 6.67 (d, *J* = 2.2 Hz, 1H), 3.72 (t, *J* = 8.3 Hz, 2H), 3.44 (s, 9H), 2.98 (t, *J* = 8.3 Hz, 2H), 0.93 (s, 9H), 0.91 (s, 9H), 0.15 (s, 6H), 0.13 (s, 6H). ^{13}C{^{1}H} NMR (126 MHz, $CDCl_3$, 298K) δ (ppm): 147.1, 146.5, 127.4, 122.1, 121.8, 121.6, 67.8, 54.1, 29.2, 26.0, 25.9, 18.4, 18.4, -3.9, -4.0.

Synthesis of DA-Me₃. To a solution of **13** (95 mg, 0.17 mmol) in 5 ml 1,4-dioxane was added 20 mL of 3N HCl (in dioxane). The mixture was stirred at room temperature for 1 hour. Solvent was removed by a rotary evaporator and the residue was recrystallized in MeOH to yield **DA-Me₃** as a white solid. Yield = 30 mg, 76%. ^{1}H NMR (400 MHz, DMSO-d_6, 298 K) δ (ppm): 8.95 (s, 1H), 8.90 (s, 1H), 6.72 – 6.66 (m, 2H), 6.53 (d, *J* = 9.7 Hz, 1H), 3.44 (t, *J* = 8.6 Hz, 2H), 3.10 (s, 9H), 2.85 (t, *J* = 8.5 Hz, 2H). ^{13}C{^{1}H} NMR (101 MHz, DMSO-d_6, 298K) δ (ppm): 145.4, 144.2, 126.7, 119.6, 116.6, 115.9, 66.2, 52.2, 27.9. ESI-MS (+ve): calcd. for $C_{11}H_{18}NO_2^+$ [M][+] *m/z* 196.1, found 196.2.

Characterization of CAP488 oxidation product. To a 25 mM solution of **CAP488** in 1:8 DMSO/PBS was added 10 eq. dopamine and the reaction mixture was stirred at room temperature for 1 hour. The solution was diluted with aqueous EDTA and extracted with CH_2Cl_2 (3 x 20 mL). The organic layers were combined and washed with aq. EDTA solution for several times and dried over anhydrous $MgSO_4$. Solvent was removed by rotary evaporator. The corresponding sulfoxide was identified by LCMS to be the major oxidation product, which was isolated from the mixture by a silica column (50:1 ethyl acetate/MeOH). ^{1}H NMR (300 MHz, $CDCl_3$, 298 K) δ (ppm): 9.00 (s, 1H), 8.56 (d, *J* = 3.9 Hz, 1H), 8.06 (d, *J* = 8.2 Hz, 1H), 7.96 (d, *J* = 7.9 Hz, 1H), 7.81–7.66 (m, 3H), 7.63–7.58 (m, 2H), 7.54–7.49 (m, 1H), 7.43–7.36 (m, 2H), 7.25–7.18 (m, 1H), 7.04 (dd, *J* = 8.6, 2.4 Hz, 1H), 6.99 (t, *J* = 3.0 Hz, 1H), 5.28 (s, 2H), 4.10 (s, 4H), 3.02 (s, 2H), 2.79 (t, *J* = 7.8 Hz, 2H), 2.47 (q, *J* = 7.4 Hz, 2H), 1.20 (t, *J* = 7.4 Hz, 3H). ^{13}C{^{1}H} NMR (101 MHz, $CDCl_3$, 298 K) δ (ppm): δ 163.1, 163.0, 160.4, 160.2, 155.9, 155.2, 152.1, 149.1, 141.6, 137.9, 137.1, 136.7, 130.7, 126.5, 125.2, 123.5, 123.3, 122.9, 122.8, 121.8, 120.4, 117.2, 114.4, 113.2, 102.0, 71.4, 59.8, 59.6, 54.0, 28.8, 26.2, 14.9.

ESI-MS (+ve): calcd. for $C_{33}H_{31}N_4O_4S_2$ [M+H]$^+$ *m/z* 611.2, found 611.2.

Fluorescence Spectroscopy

Fluorescence spectra were recorded on an Edinburgh Instruments FS5 Spectrophotometer equipped with a 150 W CW Ozone-free xenon arc lamp and a Photomultiplier R928P detection unit with spectral coverage of 200–870 nm. Samples for emission measurement were contained in a quartz cuvette with a path length of 1 cm and 1.5 mL cell volume. Millipore water was used to prepare all aqueous solutions. Dopamine hydrochloride, adrenaline hydrochloride and noradrenaline hydrochloride were purchased from Sigma-Aldrich and were dissolved in deionized water at the required concentrations before use. Human plasma samples were donated by Hong Kong Red Cross Blood Transfusion Service and the experiments involving the plasma were approved by the Human Research Ethics Committee, HKU (EA1801001).

Samples were prepared by diluting 4 µL of the 2.5 mM **CAP488** or **CAP-R** stock in 1:1 DMSO/H_2O with 986 µL of 10 mM PBS (pH 7.4) buffer, followed by addition of 10 µL of 1 mM of the catecholamine. Final concentrations of **CAP488** or **CAP-R** and the catecholamine are 10 µM. The solutions were mixed well in the quartz cuvette with a plastic disposable pipette after addition of each reagent and at 10 min intervals prior to collecting the emission spectra. For selectivity studies, 10 µM of the competing agents (except for amino acids at 100 µM and GSH at 2 mM) were used. The mixture was allowed to react for 1 hour before the emission spectra were collected. For measurements in the presence of reactive species, 200 µM of H_2O_2, ^tBuOOH, ClO^-, $\cdot OH$, $^1O_2$, NO, $ONOO^-$ and H_2S, either from commercial source or generated according to literature procedures,[11-13] was used. For emission measurement in the presence of catalase or superoxide dismutase (SOD), 100 µL of catalase (1000 U/mL) or SOD (1000 U/mL) was added to the **CAP488** solution in buffer to give a final concentration of 100 U/mL catalase or SOD. For fluorescence measurement under anaerobic conditions, the PBS buffer for preparing all the solutions was purged with argon for 20 min before use. A 2.5 mM stock solution of the Cu(I) analogue of **CAP488** in DMSO was prepared in a 1-mL microcentrifuge tube under anaerobic condition. The Cu(I) complex was characterized by ESI-MS. For dose-dependent response and determination of linear dynamic range, fluorescent intensity at emission maximum was measured after 30-min reactions. A 10 µM or 50 µM **CAP488** was used for measurements in PBS or 5% plasma in PBS respectively. Limit of detection was calculated using the 3σ method.[14] For **CAP488**, excitation was at 450 nm and emission spectra were collected from 465–650 nm. For **CAP-R**, excitation was at 325 nm and emission spectra were collected from 340–650 nm. All measurements were repeated in triplicate.

For the preparation of HeLa cell lysate, HeLa cells were seeded in a 100 mm culture dish with a seeding density of 2 x 10^6 cells, and were allowed to grow in DMEM/F12 medium supplemented

with 10% (*v/v*) fetal bovine serum, 1% (*v/v*) penicillin (100 units mL^{-1}), 1% *(v/v)* streptomycin (100 jigs mL^{-1}) at 37 °C with 5% CO_2. After reaching confluency, the medium was removed and the cells were trypsinized by trypsin-0.02% EDTA (0.05%). Cells were collected by centrifugation at 1100 rpm for 5 min, while solution was replaced with PBS with protease inhibitor cocktail (Roche). Cell samples were then subjected to freeze-thaw cycle twice to obtain lysates after centrifugation by 13,100 rpm at 4 °C for 20 min. Protein concentration was determined by Bio-Rad Protein Assay solution (protein concentration = 600 μg mL^{-1})

(a)

(b)
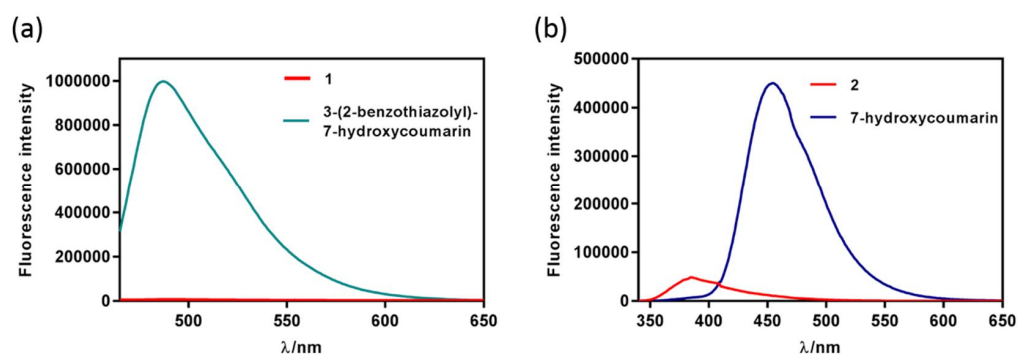
Figure S1. Emission spectra of 10 μM (a) ligand-fluorophore conjugate **1** and 3-(2-benzothiazolyl)-7-hydroxycoumarin; (b) ligand-fluorophore conjugate **2** and 7-hydroxycoumarin.
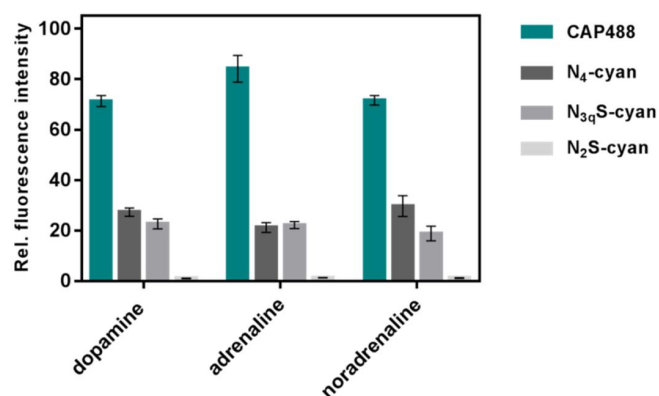
Figure S2. Fluorescence responses of 10 μM **CAP488**, **N$_4$-cyan** and **N$_2$S-cyan** towards 1 eq. of dopamine, adrenaline and noradrenaline after 1-hour of reaction. Relative fluorescence intensity was measured by the ratio of emission intensity at 488 nm before and after a 1-hour reaction. Error bars are ±SD (n = 3).
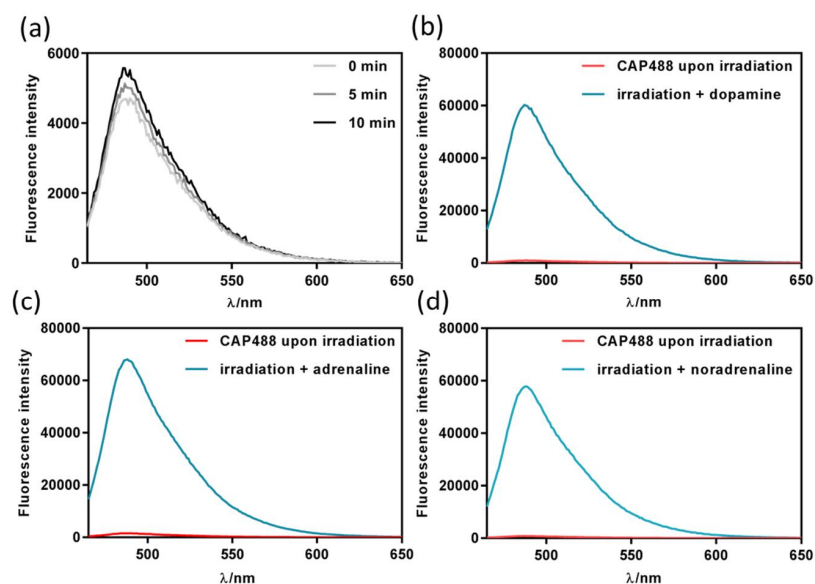
**Figure S3.** (a) Emission spectra of 10 µM **CAP488** in PBS after irradiation at 365 nm for 5 min and 10 min using a 6 W UV lamp. Emission spectra of 10 µM **CAP488** after the 10-min irradiation, followed by an 1-hour reaction with 1 eq. of (b) dopamine, (c) adrenaline and (d) noradrenaline.
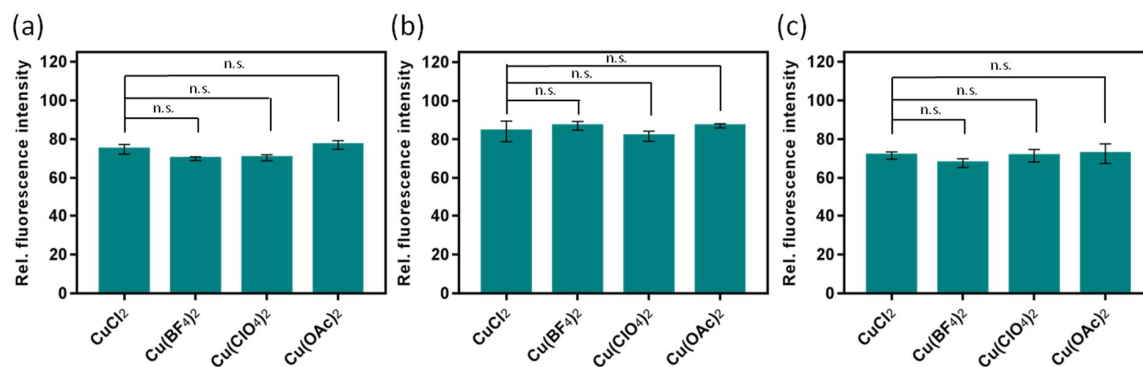
Figure S4. Fluorescence response of 10 µM **CAP488** prepared with different copper(II) salts towards 1 eq. of (a) dopamine, (b) adrenaline and (c) noradrenaline after 1-hour of reaction. Relative fluorescence intensity was measured by the ratio of emission intensity at 488 nm before and after a 1-hour reaction. No significant difference in the fluorescence response was observed for probes prepared from different copper(II) salts. Error bars are ±SD (n = 3). Statistical analysis was performed with t-test at 1% level of significance.
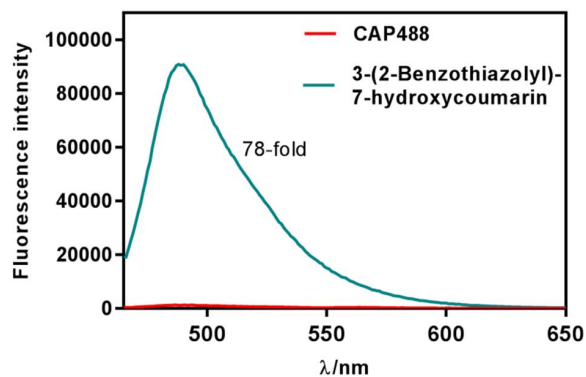
Figure S5. Comparison of emission intensity of 10 μM **CAP488** and 0.5 μM 3-(2-benzothiazolyl)-7-hydroxycoumarin in PBS. A bond cleavage of 5% yield of 10 μM **CAP488** would result in 0.5 μM 3-(2-benzothiazolyl)-7-hydroxycoumarin, corresponding to an emission intensity ratio of 78-fold.
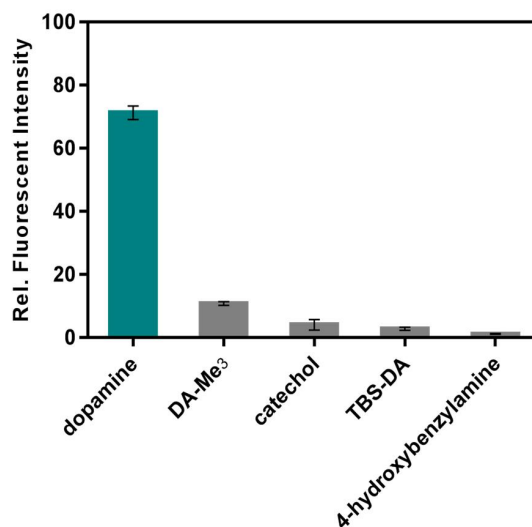
Figure S6. Fluorescence response of 10 μM **CAP488** towards 1 eq. dopamine, **DA-Me$_3$**, catechol, **TBS-DA** and 4-hydroxybenylamine. Relative fluorescence intensity was measured by the ratio of emission intensity at 488 nm before and after a 1-hour reaction. Error bars are ±SD (n = 3).
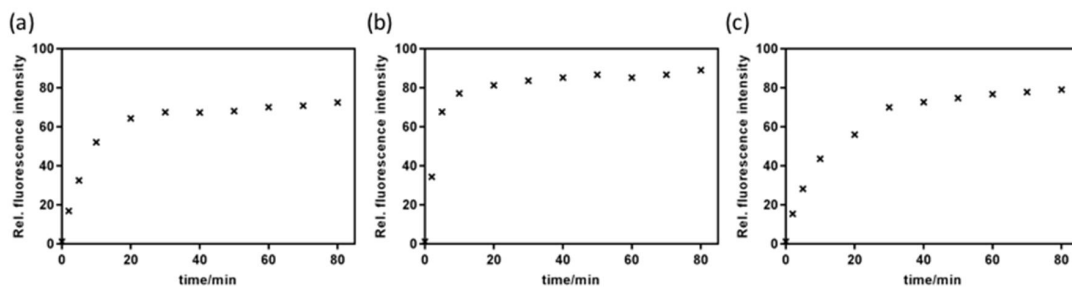
Figure S7. Time-dependent fluorescence response of 10 µM **CAP488** towards 1 eq. (a) dopamine, (b) adrenaline and (c) noradrenaline.
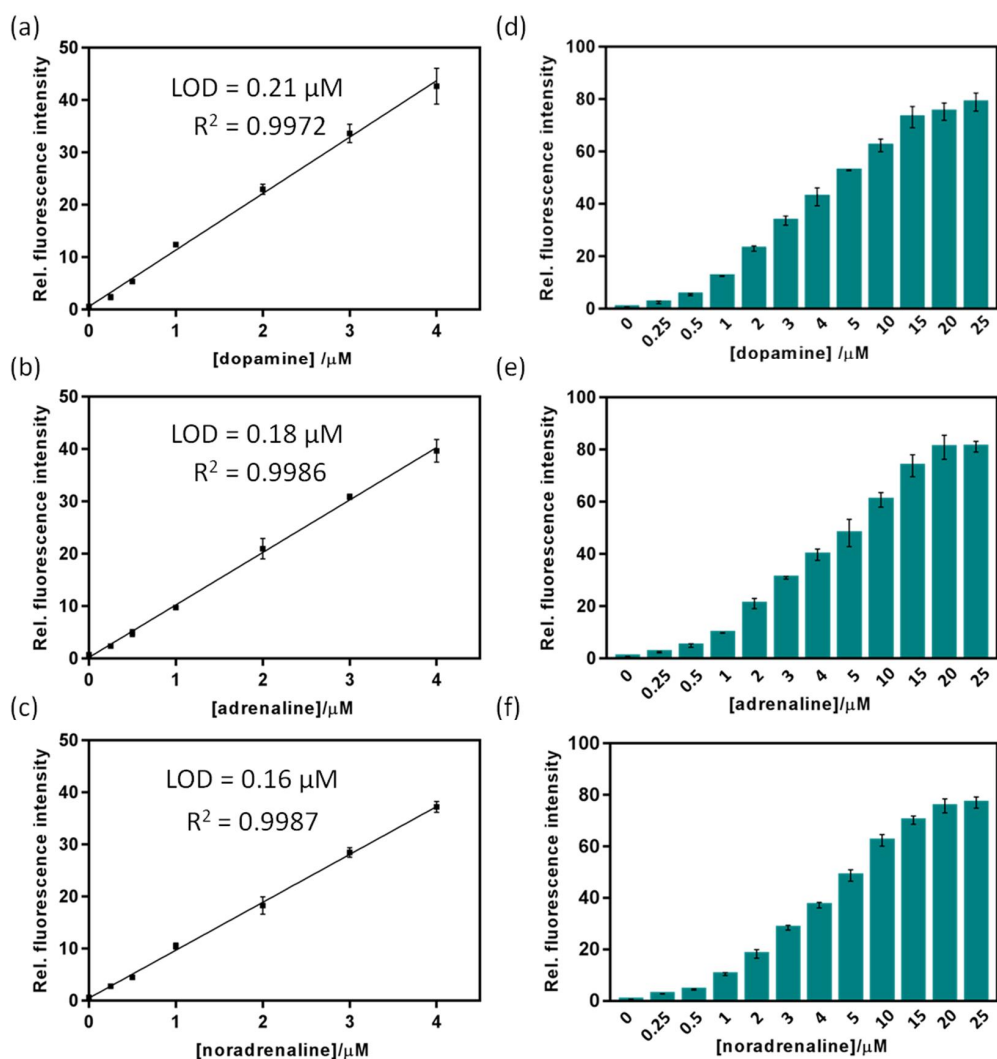
Figure S8. Linear dynamic range (a–c) and dose-dependent response (e–f) of 10 µM **CAP488** in 10 mM PBS (pH 7.4) towards (a and d) dopamine, (b and e) adrenaline and (c and f) noradrenaline. Relative fluorescence intensities were measured at 488 nm after reaction for 30 mins. Error bars are ±SD (n = 3).
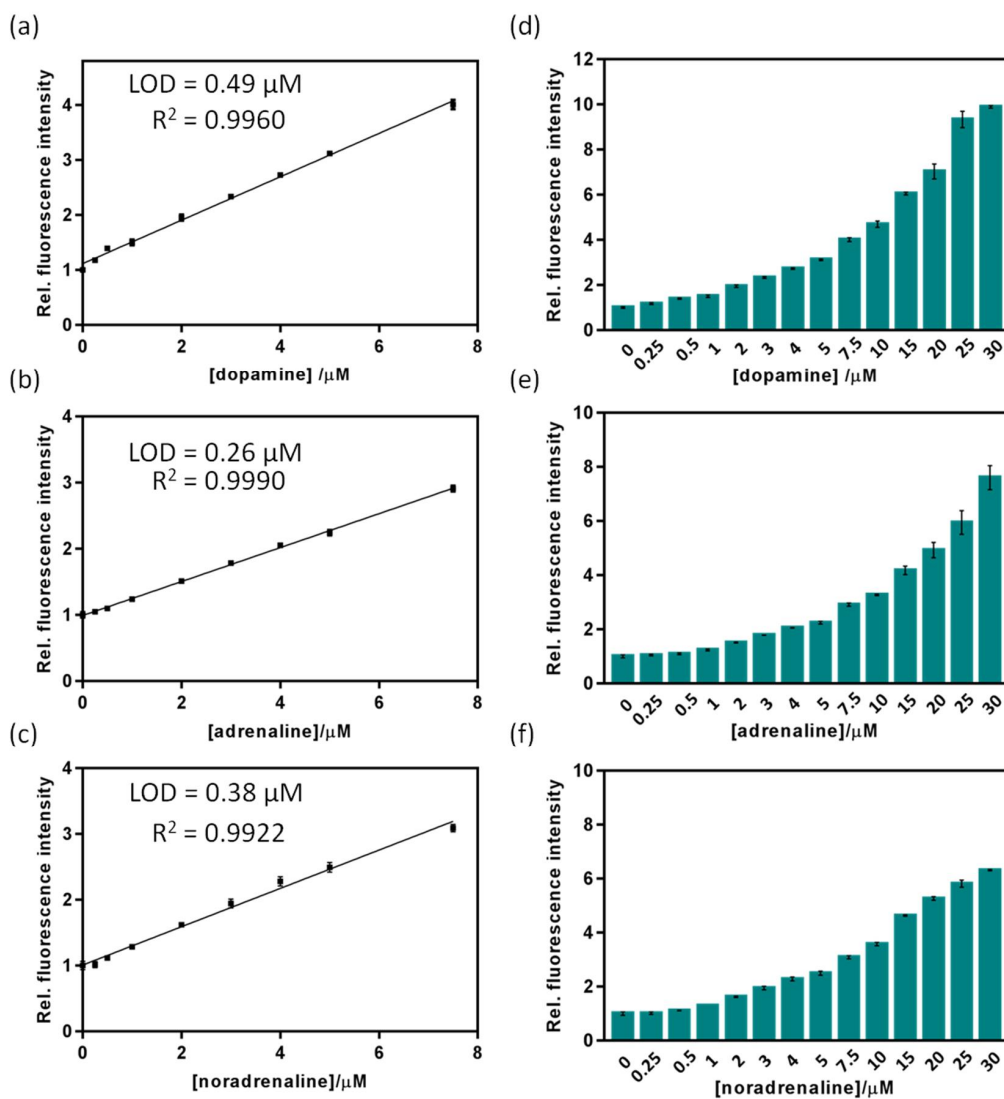
Figure S9. Linear dynamic range (a–c) and dose-dependent response (e–f) of 50 μM **CAP488** in 5% human plasma in 10 mM PBS (pH 7.4) towards (a and d) dopamine, (b and e) adrenaline and (c and f) noradrenaline. Relative fluorescence intensities were measured at 488 nm after reaction for 30 mins. Error bars are ±SD (n = 3).
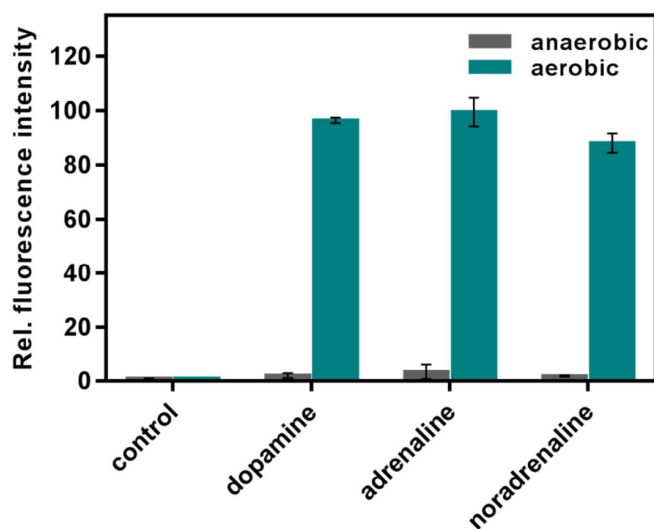
Figure S10. Fluorescence response of 10 µM **CAP488** towards 20 eq. of dopamine, adrenaline and noradrenaline under anaerobic and aerobic conditions. Relative fluorescence intensity was measured by the ratio of emission intensity at 488 nm before and after a 1-hour reaction. Error bars are ±SD (n = 3).
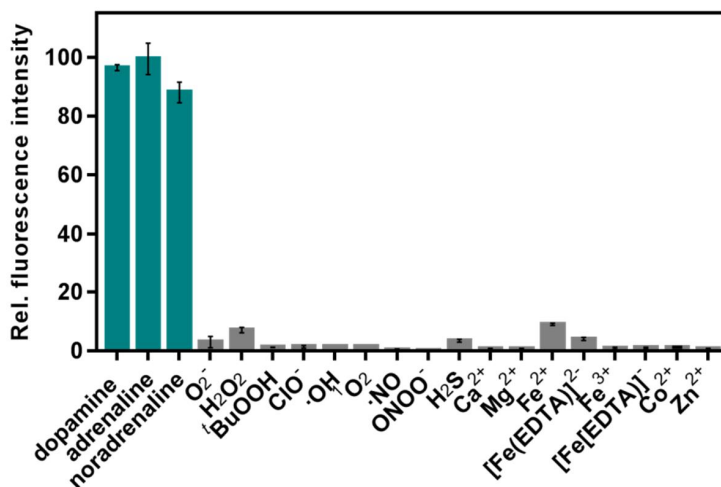
Figure S11. Fluorescence response of 10 µM **CAP488** towards 20 eq. of the catecholamines, L-DOPA, reactive oxygen, nitrogen, sulfur species, and metal ions. Relative fluorescence intensity was measured by the ratio of emission intensity at 488 nm before and after a 30-min reaction. Error bars are ±SD (n = 3).
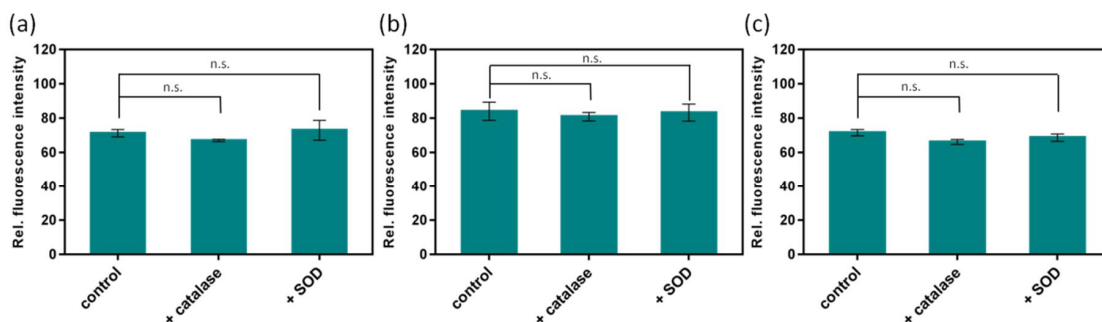
Figure S12. Fluorescence response of 10 µM **CAP488** towards 1 eq. of (a) dopamine, (b) adrenaline and (c) noradrenaline in the presence of catalase (100 U/mL) or SOD (100 U/mL). Relative fluorescence intensity was measured by the ratio of emission intensity at 488 nm before and after a 1-hour reaction. No significant effect of the enzymes was observed. Error bars are ±SD (n = 3). Statistical analysis was performed with t-test at 1% level of significance.
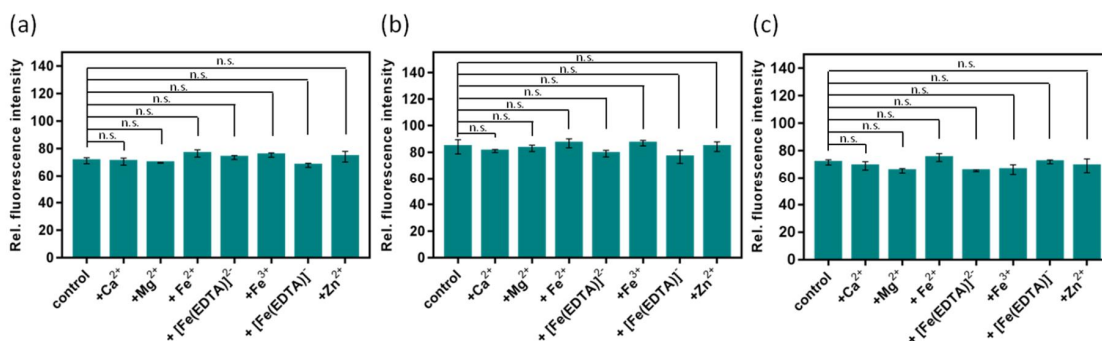
Figure S13. Fluorescence response of 10 µM **CAP488** towards 1 eq. (a) dopamine, (b) adrenaline and (c) noradrenaline in the presence of 20 eq. metal ions. Relative fluorescence intensity was measured by the ratio of emission intensity at 488 nm before and after a 1-hour reaction. No significant effect of the metal ions was observed. Error bars are ±SD (n = 3). Statistical analysis was performed with t-test at 1% level of significance.
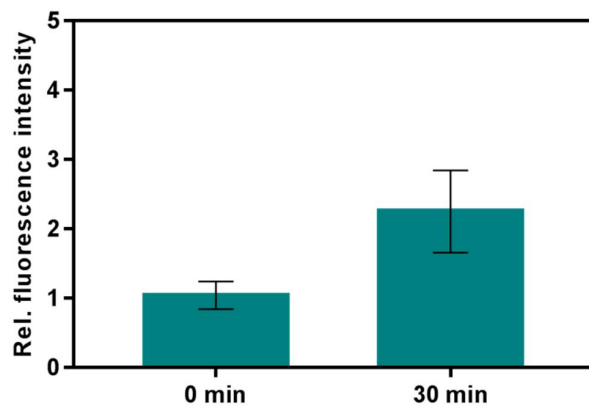
Figure S14. Fluorescence response of 10 µM of **1**-Cu(I) towards dioxygen. Error bars are ±SD (n = 3).
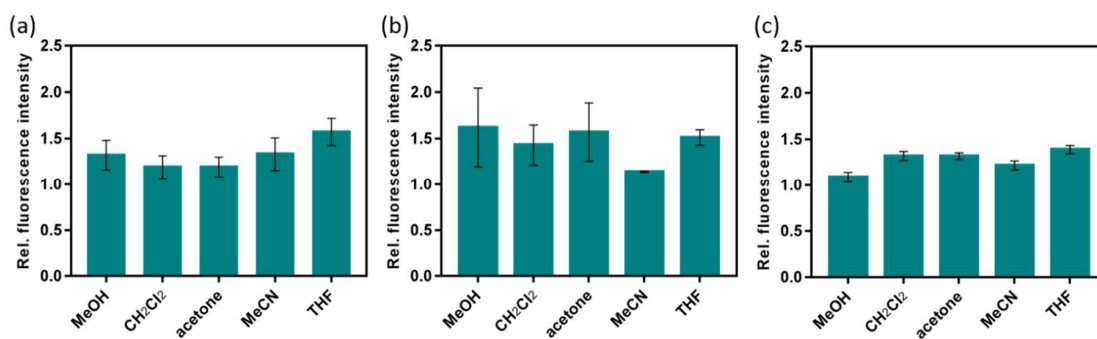
Figure S15. Fluorescence response of 10 µM of **CAP488** towards 1 eq. (a) dopamine, (b) adrenaline and (c) noradrenaline in organic solvents. Relative fluorescence intensity was measured by the ratio of emission intensity at 488 nm before and after a 1-hour reaction. Error bars are ±SD (n = 3).
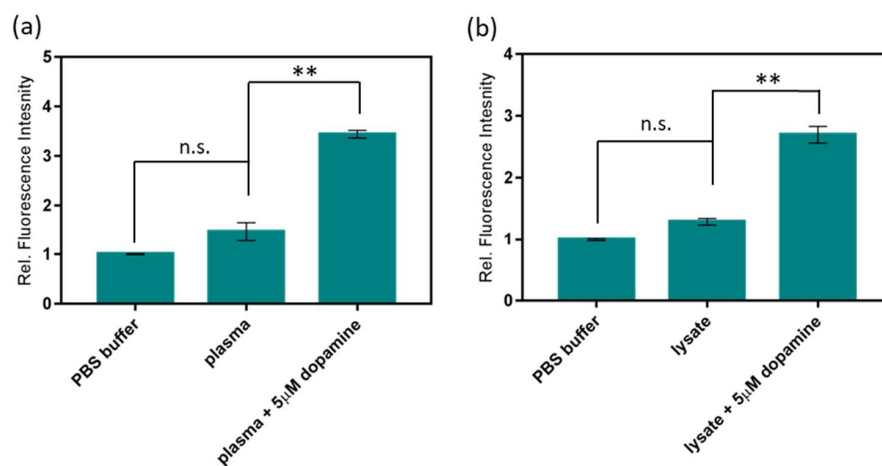
Figure S16. Dose-dependent response of 10 µM **CAP488** in (a) 5% plasma in 10 mM PBS (pH 7.4) and (b) HeLa cell lysate towards dopamine. Relative fluorescence intensities were measured at 488 nm after reaction for 1 h. Error bars are ±SD (n = 3). Statistical analysis was performed with t-test at 1% level of significance. **: $p < 0.01$.

LCMS and ESI-MS Analyses

LCMS analyses were carried out using a Waters-Alliance e2695 system coupled to a 2489 UV/Vis detector and an ACQUITY QDa MS detector. Samples were prepared by diluting a mixture of 8 µL of 2.5 mM **CAP488** and 4 µL of 100 mM catecholamine with in deionized water to a final volume of 200 µL. A 100 µM **PC1**, **SOP-orange** or H_2DCF were introduced for studying the involvement of free ROS. The mixtures were allowed to react for 1 hour and then injected onto a SunFire C18 column (3.5 µm, 3.0 × 150 mm) and eluted at 25 °C and 0.6 mL/min using a gradient of CH_3CN/water (with 0.5% formic acid) from 5% to 100%. Absorption was monitored at 254 nm or 350 nm. Yield of C–O bond cleavage was determined by the peak area of 3-(2-benzothiazolyl)-7-hydroxycoumarin in the corresponding chromatogram against a calibration curve constructed from stock solutions of 3-(2-benzothiazolyl)-7-hydroxycoumarin at 0.1–15 µM. Oxidation products were also characterized by a flow injection analysis of the reaction mixture after treatment with 4 µL of 5 mM EDTA (Fluka) using CH_3CN (with 0.5% formic acid) as the eluent.

HR-ESI-MS analyses were performed on a Waters Micromass Q-Tof Premier quadrupole time-of-flight tandem mass spectrometer. A mixture of 2.5 mM **CAP488** and 4 eq. of the catecholamine was diluted by *ca.* 50 times with CH_3CN and introduced into the ESI source using a syringe pump at a flow rate of 5 µL min^{-1}. Mass resolution was fixed at about 8000 (full width at half-height). For accurate mass measurements, the mass accuracy was calibrated to < 10 ppm using sodium formate as an external calibrant. Structural assignments in LCMS and HR-ESI-MS were based on the *m/z*.
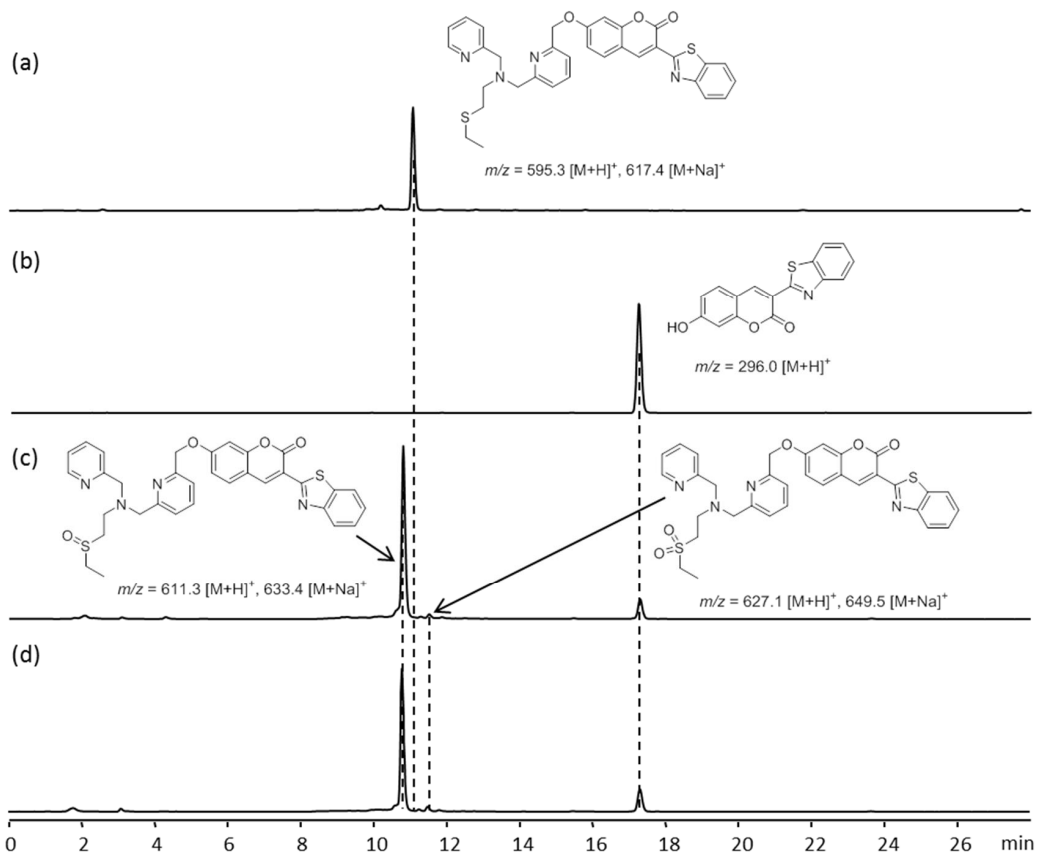
Figure S17. LCMS analysis of (a) **1**; (b) 3-(2-benzothiazolyl)-7-hydroxycoumarin; and reaction mixture of 100 µM of **CAP488** with 2 mM of (c) dopamine and (d) noradrenaline for 1 hour. UV absorbance was monitored at 350 nm. Yield of released 3-(2-benzothiazolyl)-7-hydroxycoumarin from the reaction of the probe with dopamine and noradrenaline are 4.4% and 4.8% respectively.
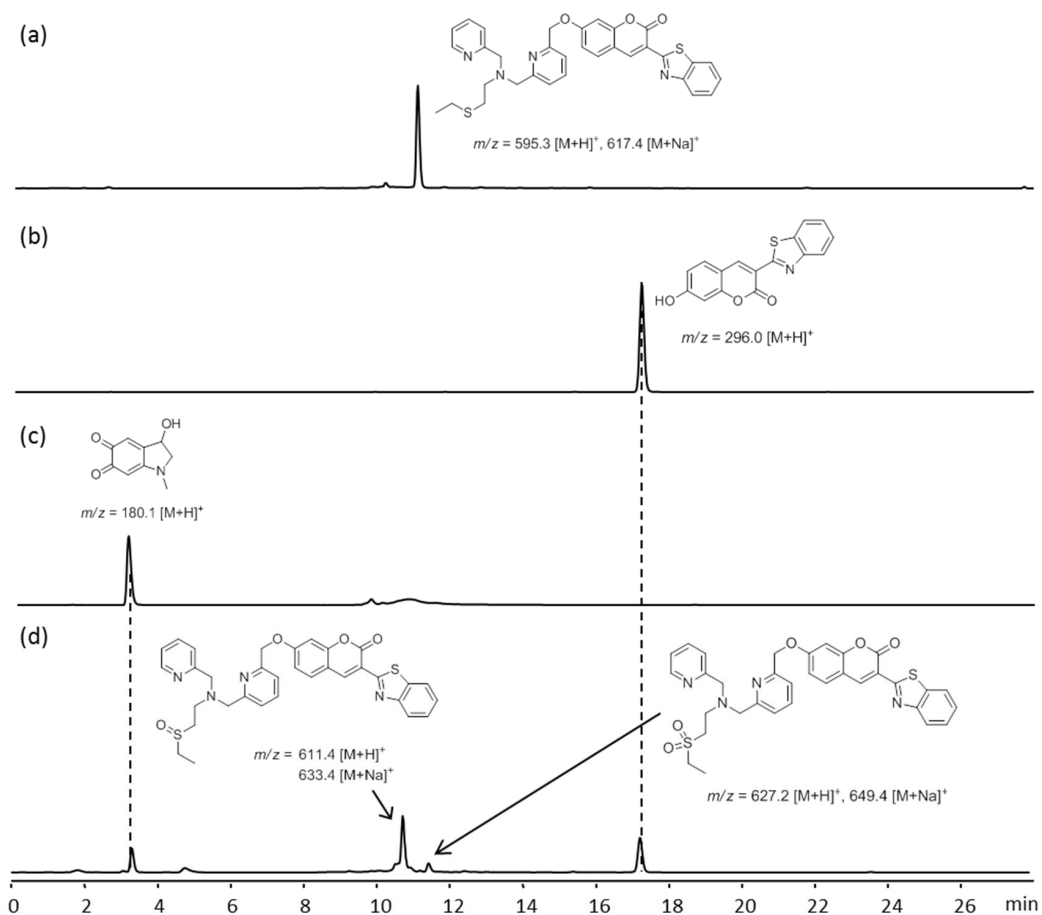
Figure S18. LCMS analysis of (a) **1**; (b) 3-(2-benzothiazolyl)-7-hydroxycoumarin; (c) adrenochrome; (c) reaction mixture of 100 µM **CAP488** with 2 mM of adrenaline for 1 hour. UV absorbance was monitored at 350 nm. Yield of released 3-(2-benzothiazolyl)-7-hydroxycoumarin is 5.2%.
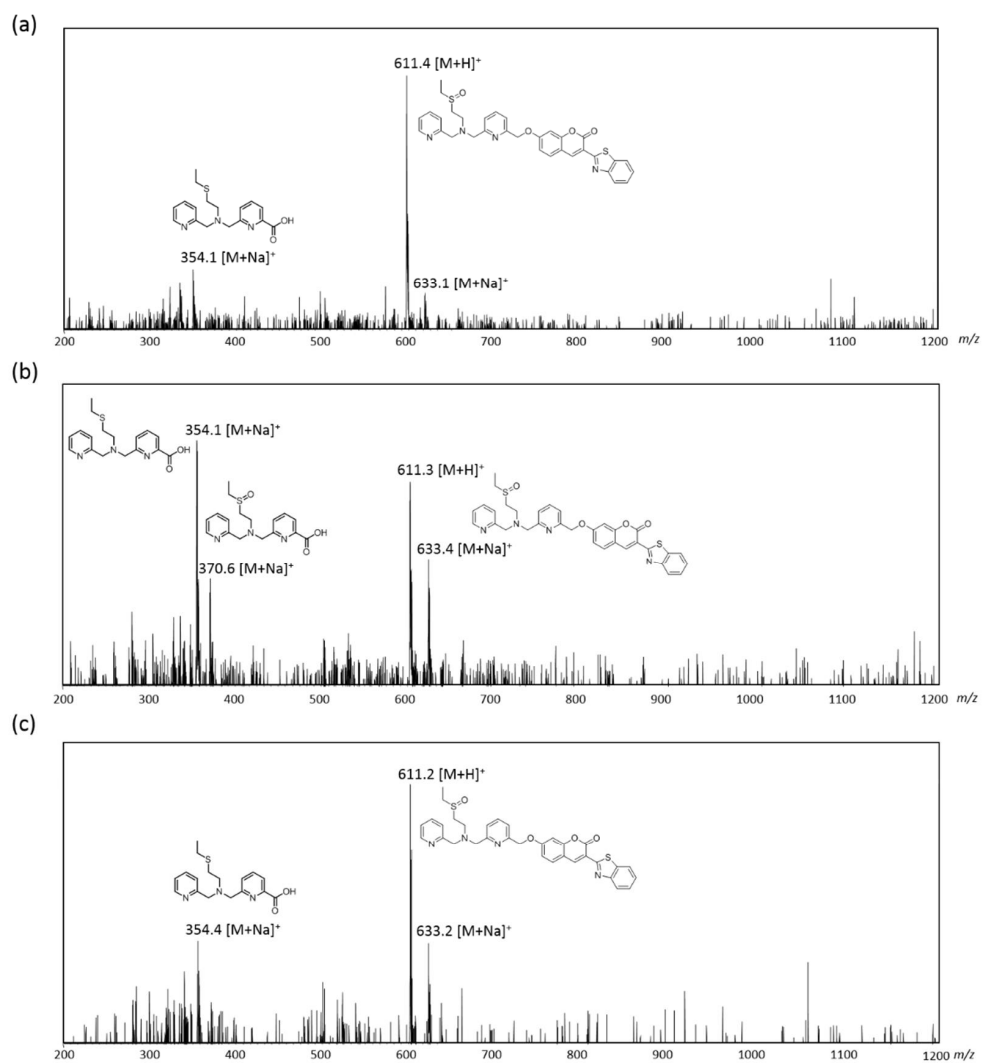
Figure S19. ESI-MS (+ve) analysis of reaction mixture of **CAP488** with (a) dopamine; (b) adrenaline and (c) noradrenaline.
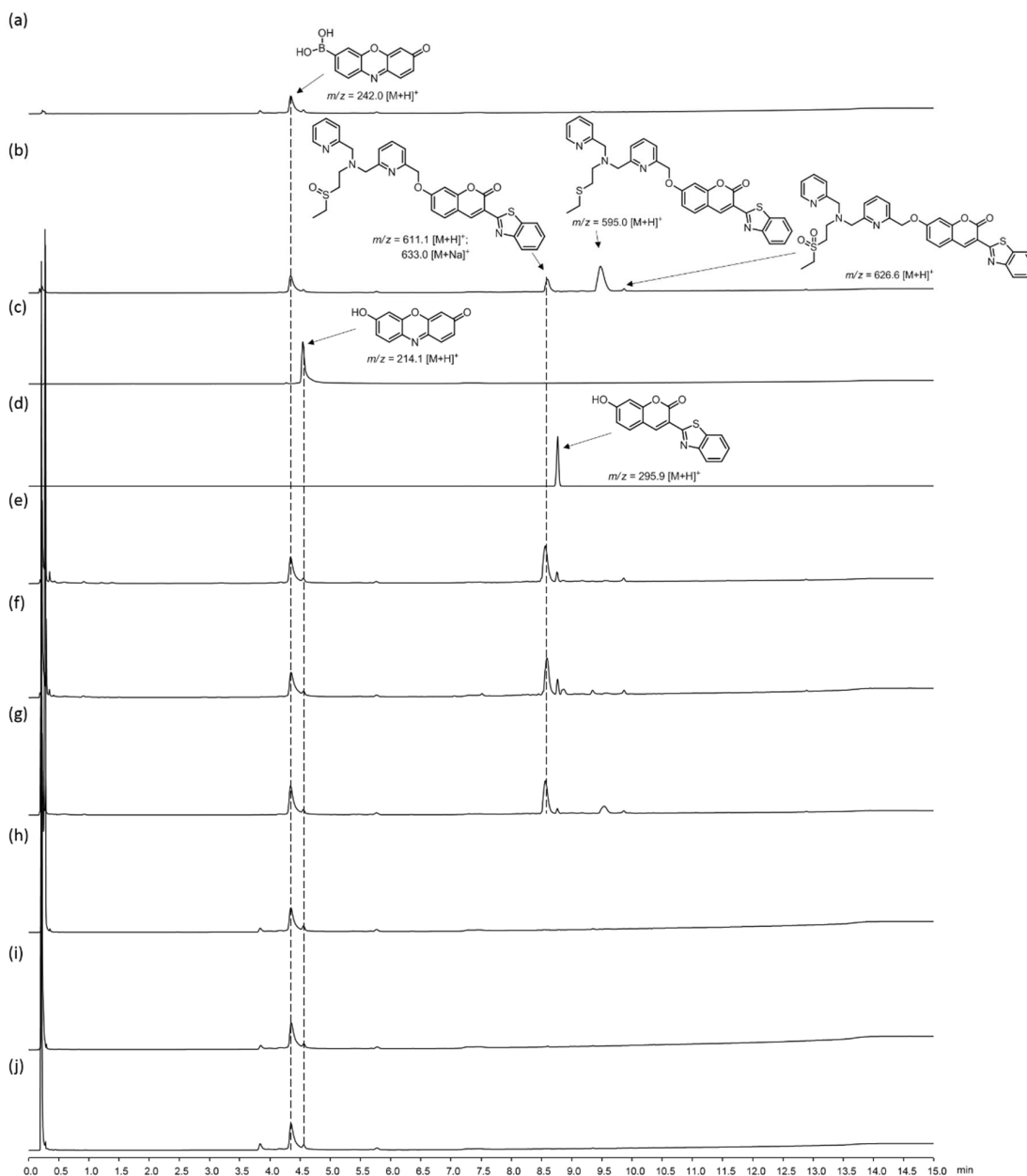
Figure S20. LCMS analysis of (a) **PC1**; (b) mixture of 100 µM **PC1** and 100 µM **CAP488**; (c) resorufin; (d) 3-(2-benzothiazolyl)-7-hydroxycoumarin; (e to g) reaction mixture of 100 µM **PC1**, 100 µM **CAP488** with 2 mM of (e) dopamine, (f) adrenaline, and (g) noradrenaline; (h to j) reaction mixture of 100 µM **PC1** with 2 mM of (h) dopamine, (i) adrenaline, and (j) noradrenaline for 1 hour. UV absorbance was monitored at 254 nm.
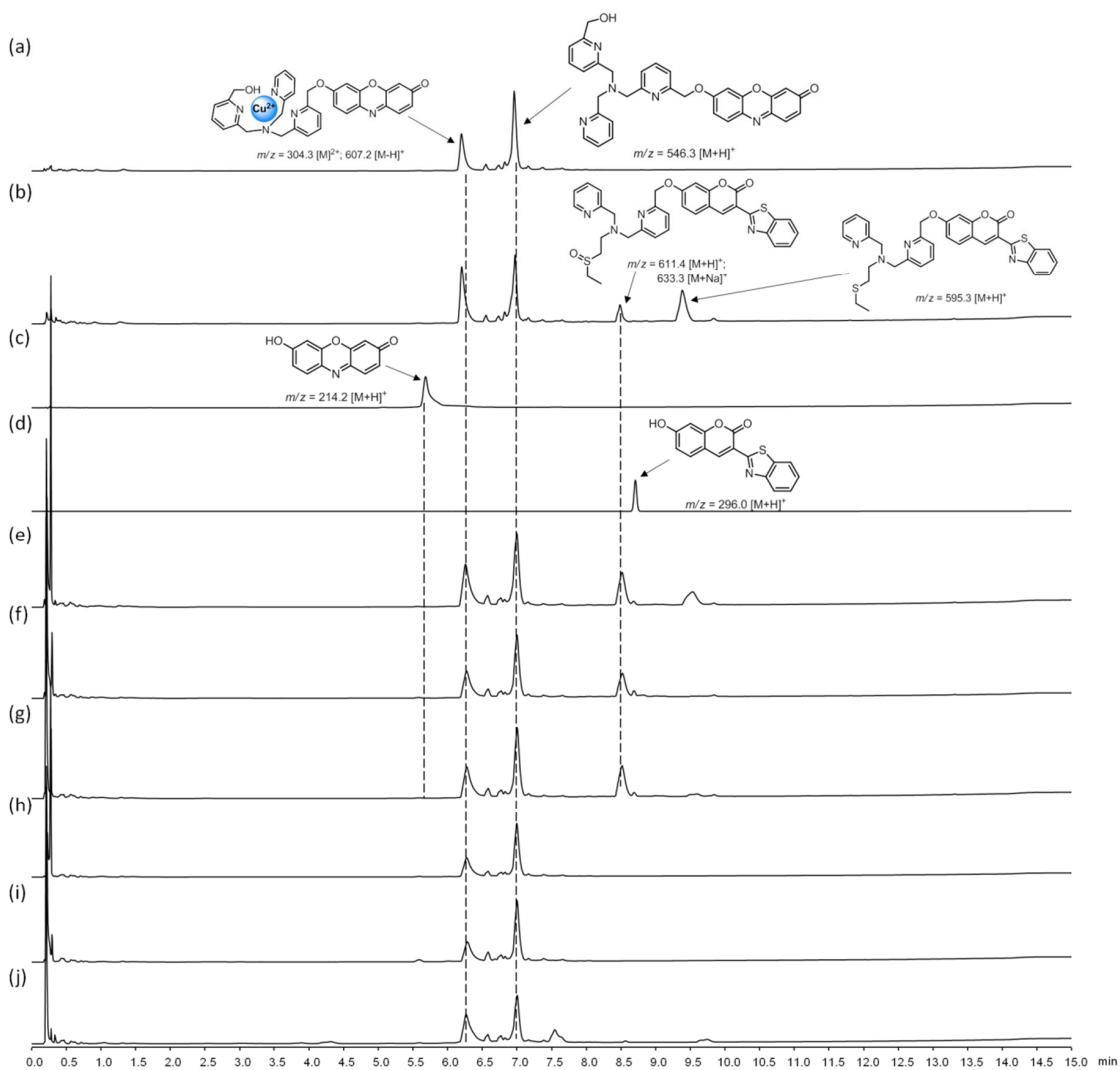
Figure S21. LCMS analysis of (a) **SOP-orange**; (b) mixture of 100 µM **SOP-orange** and 100 µM **CAP488**; (c) resorufin; (d) 3-(2-Benzothiazolyl)-7-hydroxycoumarin; reaction mixture of 100 µM **SOP-orange**, 100 µM **CAP488** with 2 mM of (e) dopamine, (f) adrenaline, and (g) noradrenaline; reaction mixture of 100 µM **SOP-orange** with 2 mM of (h) dopamine, (i) adrenaline, and (j) noradrenaline for 1 hour. UV absorbance was monitored at 254 nm.
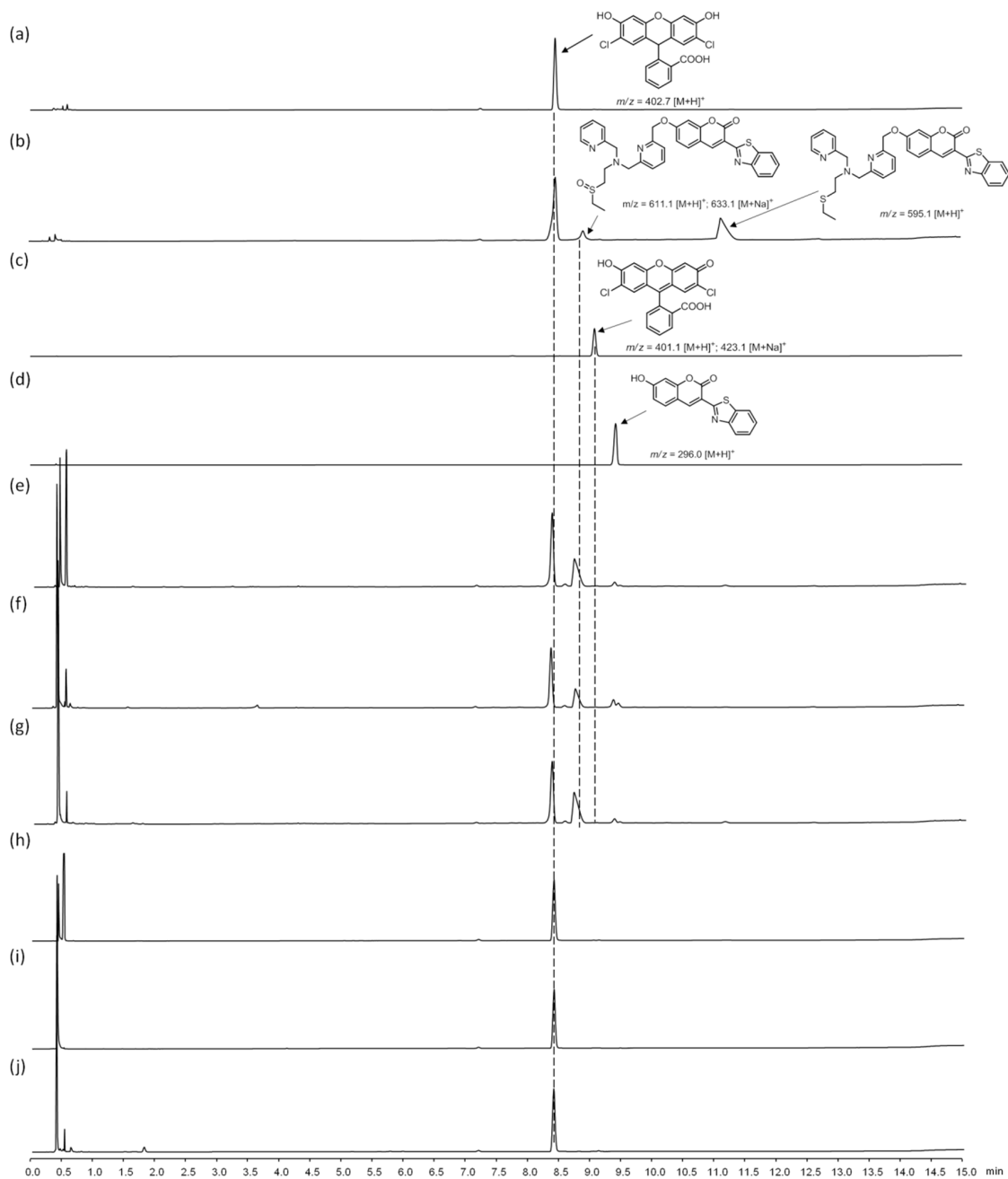
Figure S22. LCMS analysis of (a) H₂DCF; (b) a mixture of 100 μM H₂DCF and 100 μM **CAP488**; (c) dichlorofluorescein; (d) 3-(2-benzothiazolyl)-7-hydroxycoumarin; (e to g) reaction mixture of 100 μM H₂DCF, 100 μM **CAP488** with 2 mM of (e) dopamine, (f) adrenaline, and (g) noradrenaline; (h to j) reaction mixture of 100 μM H₂DCF with 2 mM of (h) dopamine, (i) adrenaline, and (j) noradrenaline for 1 hour. UV absorbance was monitored at 254 nm.
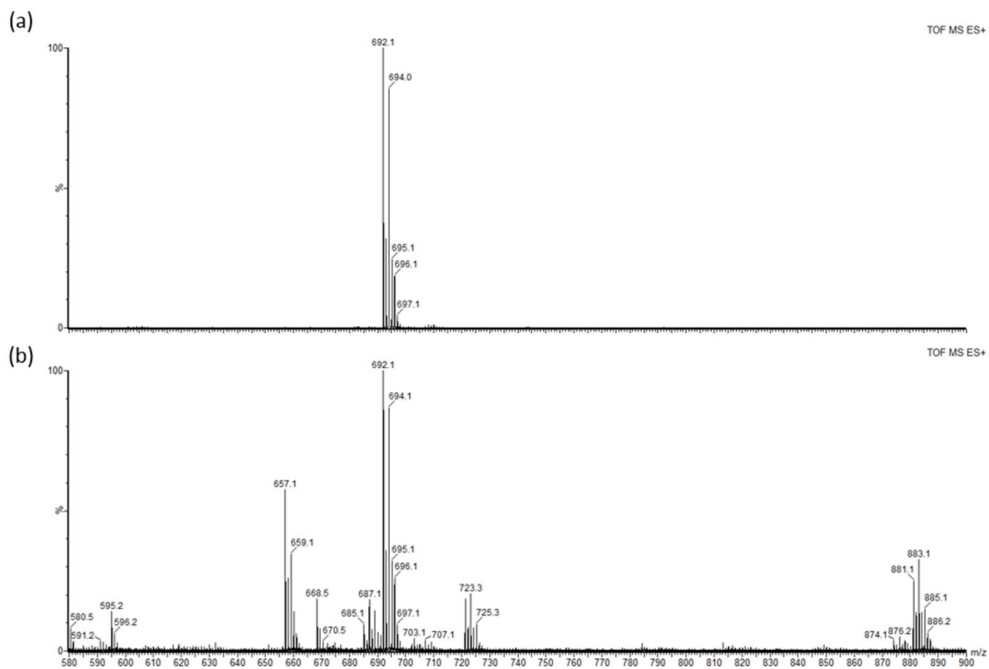
Figure S23. High resolution ESI-MS analysis of **CAP488** and dopamine. Full spectrum of (a) **CAP488**; (b) reaction mixture of **CAP488** and dopamine.
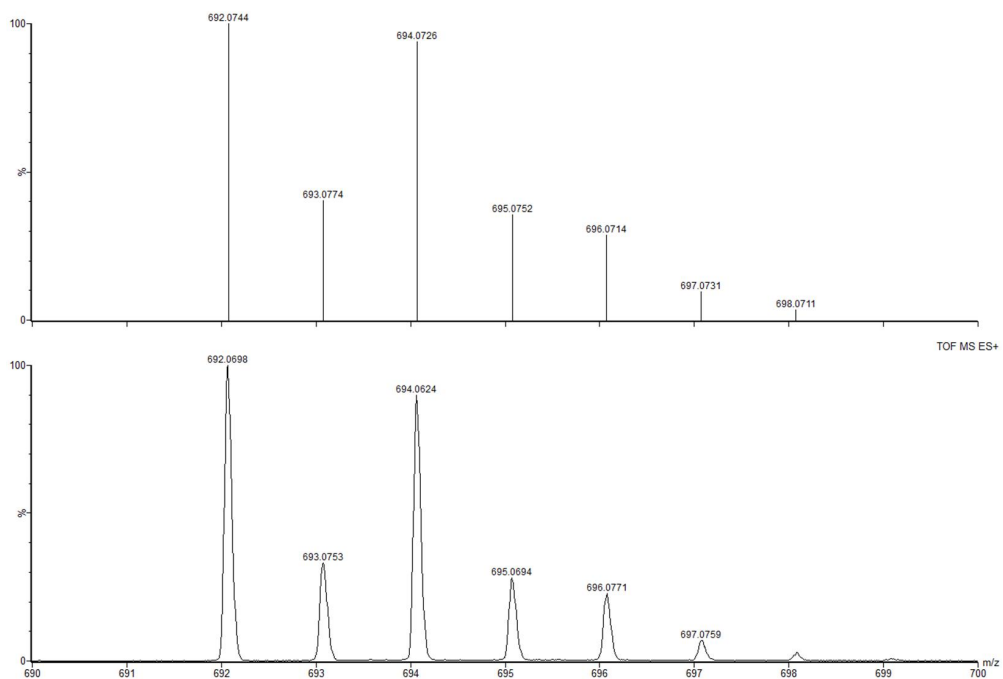
Figure S24. Isotopic distribution pattern matching of the *m/z* = 692.0698 ion attributed to **CAP488**. Mass accuracy: 7 ppm.
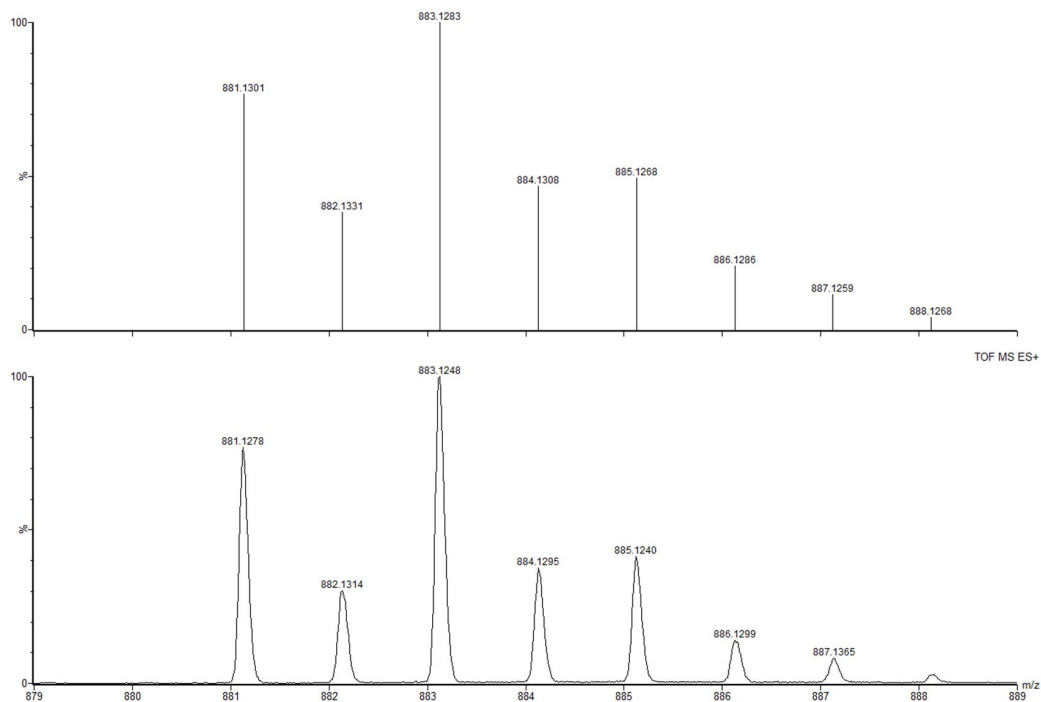
Figure S25. Isotopic distribution pattern matching of the *m/z* = 881.1278 ion observed in the reaction mixture of **CAP488** and dopamine. Mass accuracy: 3 ppm.
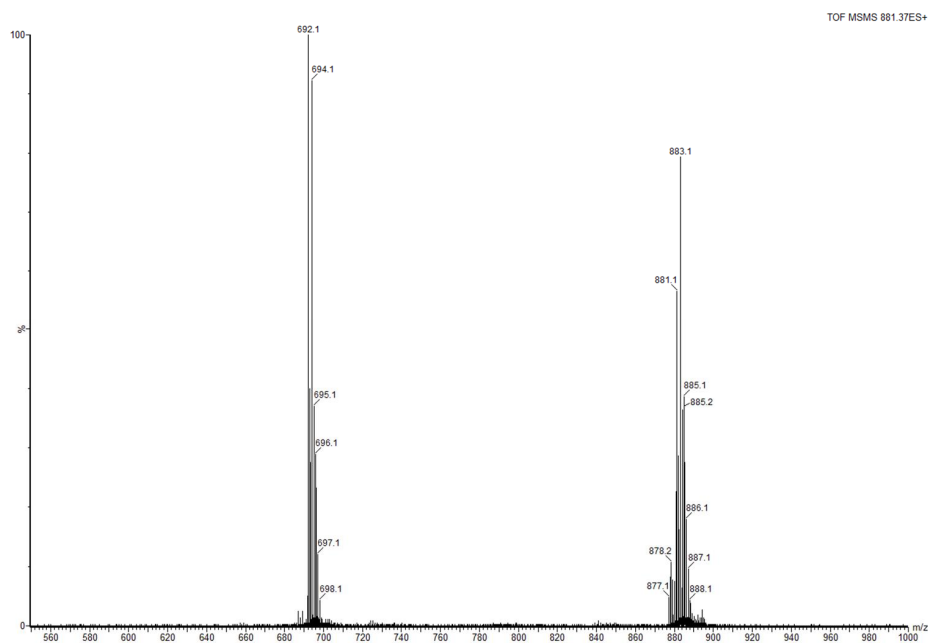

Figure S26. Collision-induced dissociation of the *m/z* = 883.1 ion observed in the reaction mixture of **CAP488** and dopamine.
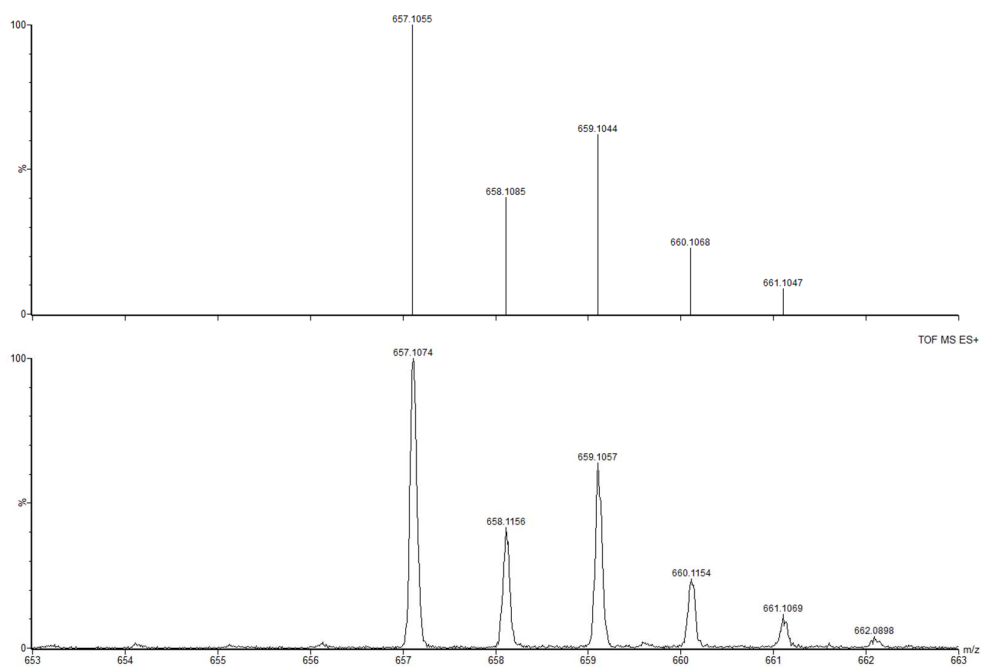
Figure S27. Isotopic distribution pattern matching of the *m/z* = 657.1074 ion observed in the reaction mixture of **CAP488** and dopamine. Mass accuracy: 3 ppm.
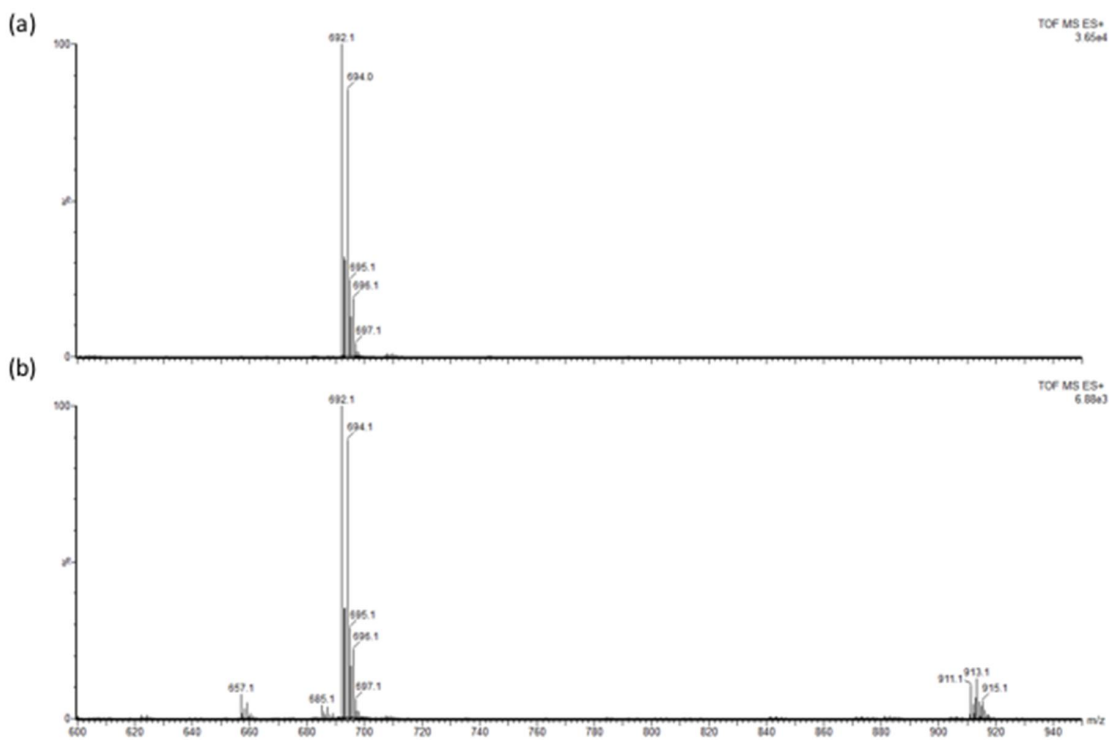
Figure S28. High resolution ESI-MS analysis of **CAP488** and adrenaline. Full spectrum of (a) **CAP488**; (b) reaction mixture of **CAP488** and adrenaline.
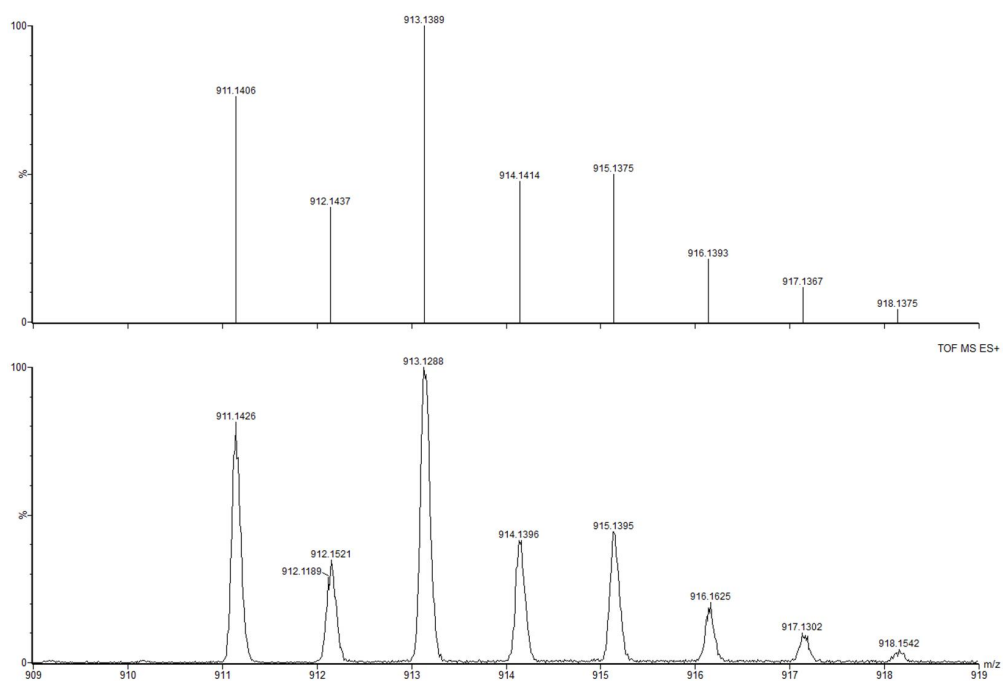
Figure S29. Isotopic distribution pattern matching of the *m/z* = 911.1426 ion observed in the reaction mixture of **CAP488** and adrenaline. Mass accuracy: 2 ppm.
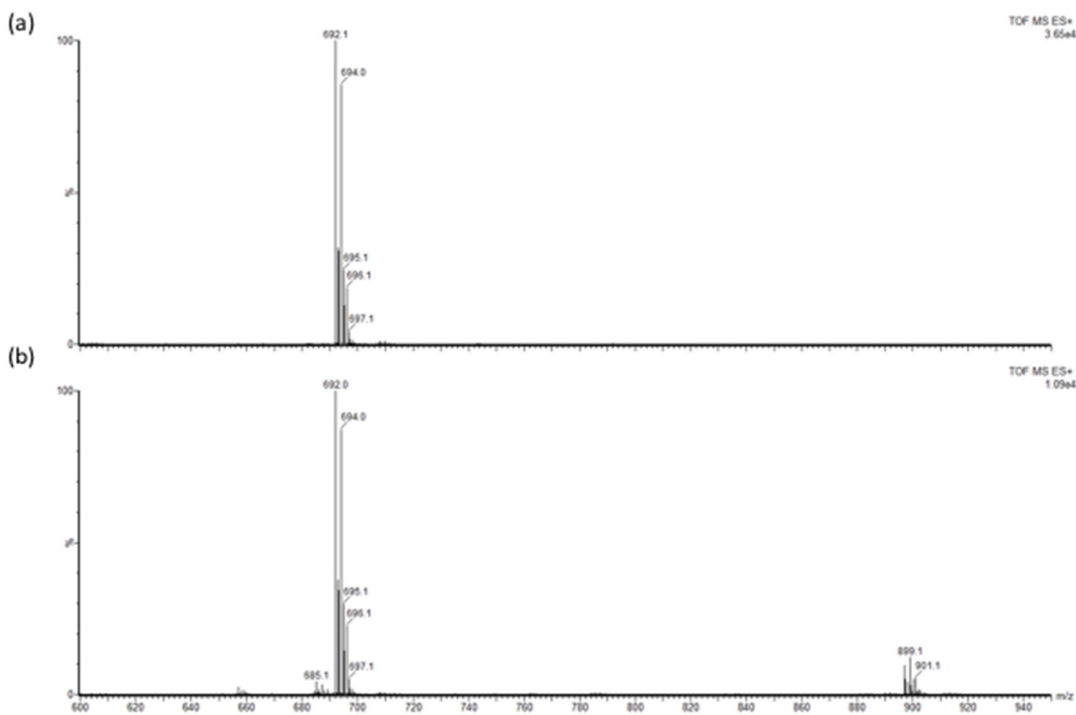
Figure S30. High resolution ESI-MS analysis of **CAP488** and noradrenaline. Full spectrum of (a) **CAP488**; (b) reaction mixture of **CAP488** and noradrenaline.
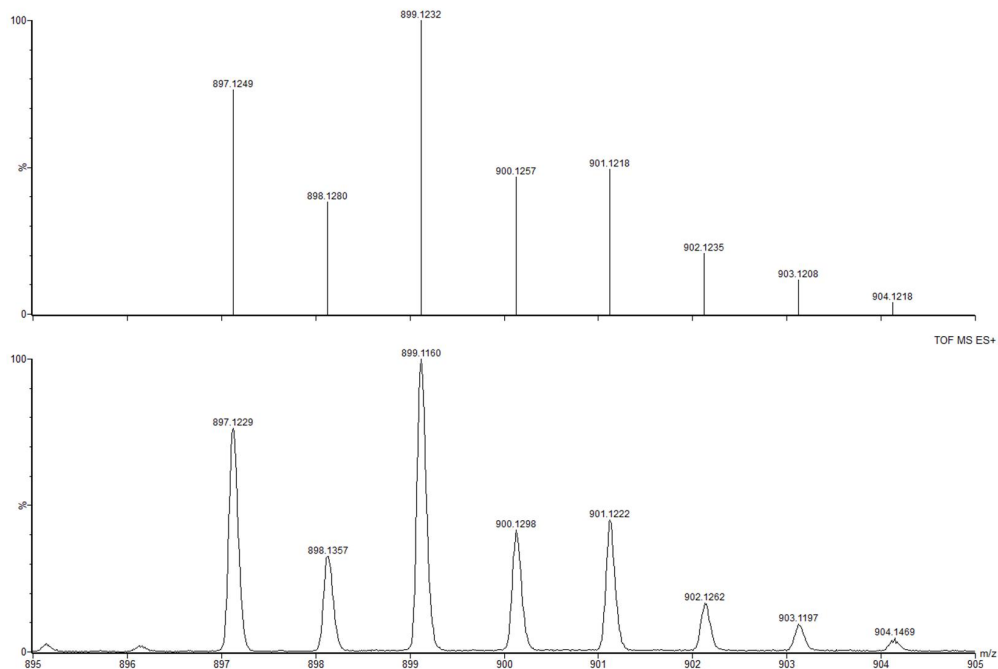
Figure S31. Isotopic distribution pattern matching of the *m/z* = 897.1229 ion observed in the reaction mixture of **CAP488** and noradrenaline. Mass accuracy: 2 ppm.
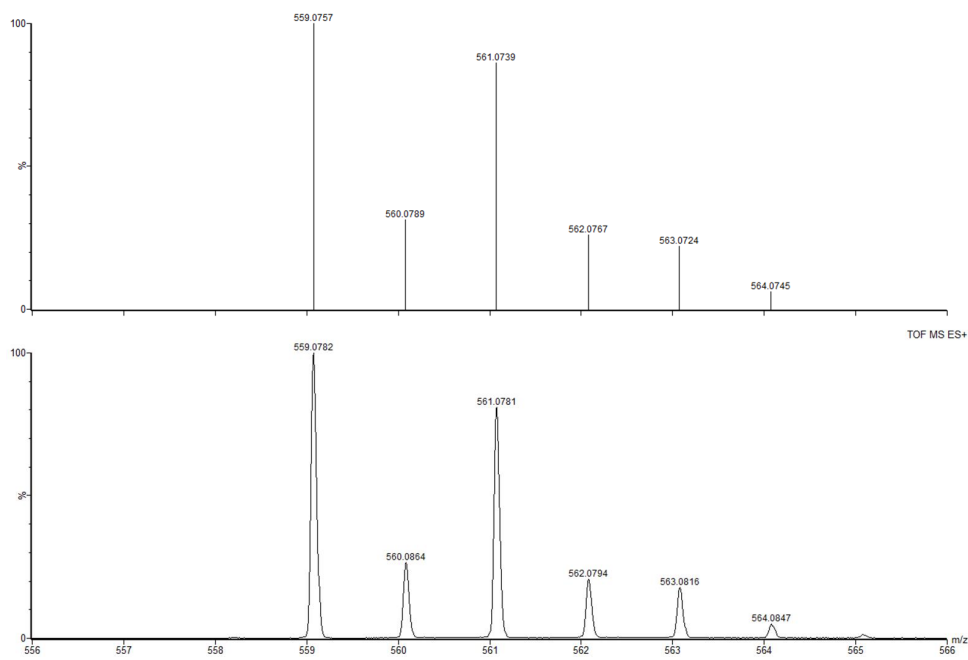
Figure S32. Isotopic distribution pattern matching of the *m/z* = 559.0782 ion attributed to **CAP-R**. Mass accuracy: 4 ppm.
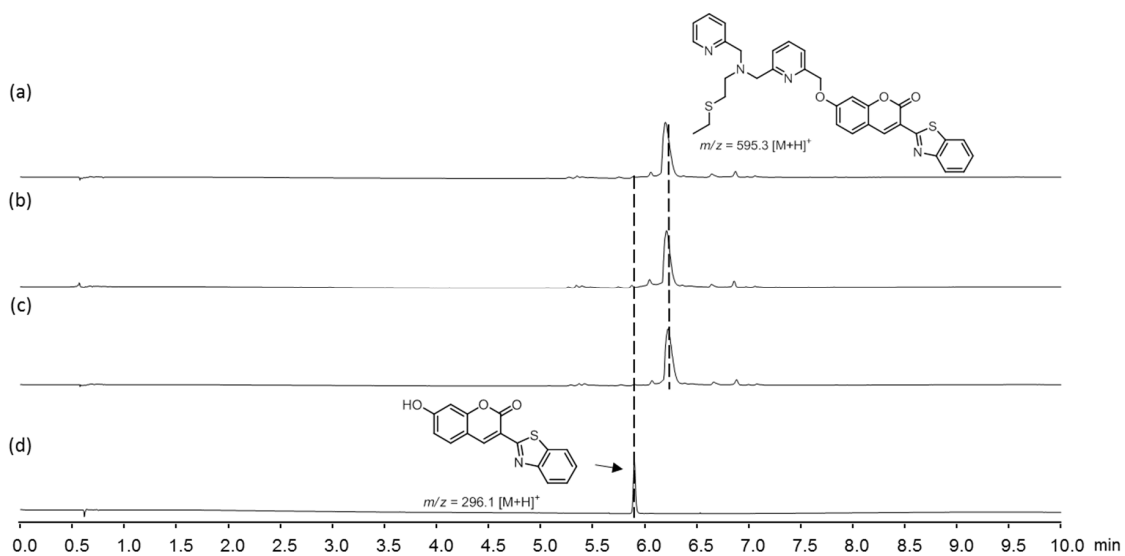
Figure S33. LCMS analysis of reaction mixture of 100 μM **CAP488** with 2 mM of (a) dopamine; (b) adrenaline and (c) noradrenaline in MeOH for 1 hour; (d) 3-(2-benzothiazolyl)-7-hydroxycoumarin. UV absorbance was monitored at 350 nm.

Cyclic Voltammetry

Cyclic voltammetry was conducted on a Princeton Applied Research PMC-1000 Potentiostat. The working electrode was glassy carbon; the reference electrode was SCE; the counter electrode was a platinum wire. Scan rate: 100 mV/s. All potentials are reported versus SCE (+0.241 V vs. NHE).

(a)

(b)
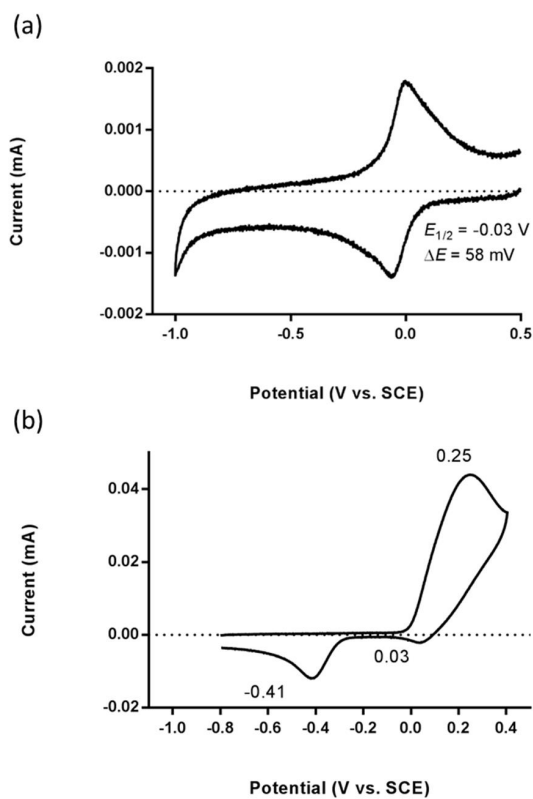
Figure S34. Cyclic voltammograms of (a) **CAP488** and (b) dopamine hydrochloride in 1:2 v/v DMSO/PBS (10 mM, pH 7.4) buffer. The signal of **CAP488** is relatively weak because of the limited solubility of **CAP488** in 1:2 v/v DMSO/PBS (< 1 mM). The irreversible dopamine oxidation observed in consistent with that reported in the literature.[15]

Kinetic Studies

In kinetic studies, the rate constants were obtained by a pseudo-first order measurement **CAP488** using 500 eq. of the catecholamines. Emission at 488 nm was monitored at 1-min time intervals and plotted against time. Pseudo-first order rate constants were obtained from the slope of the plot of $\ln(\{I\text{-}I_{max}\}/\{I_0\text{-}I_{max}\})$ against time. Reaction orders were studied using the initial rate method. For the determination of reaction order with respect to the catecholamines, emission intensities at 488 nm were recorded at a time interval of 1 second. Initial rates were determined from the initial slope of the plots against time.
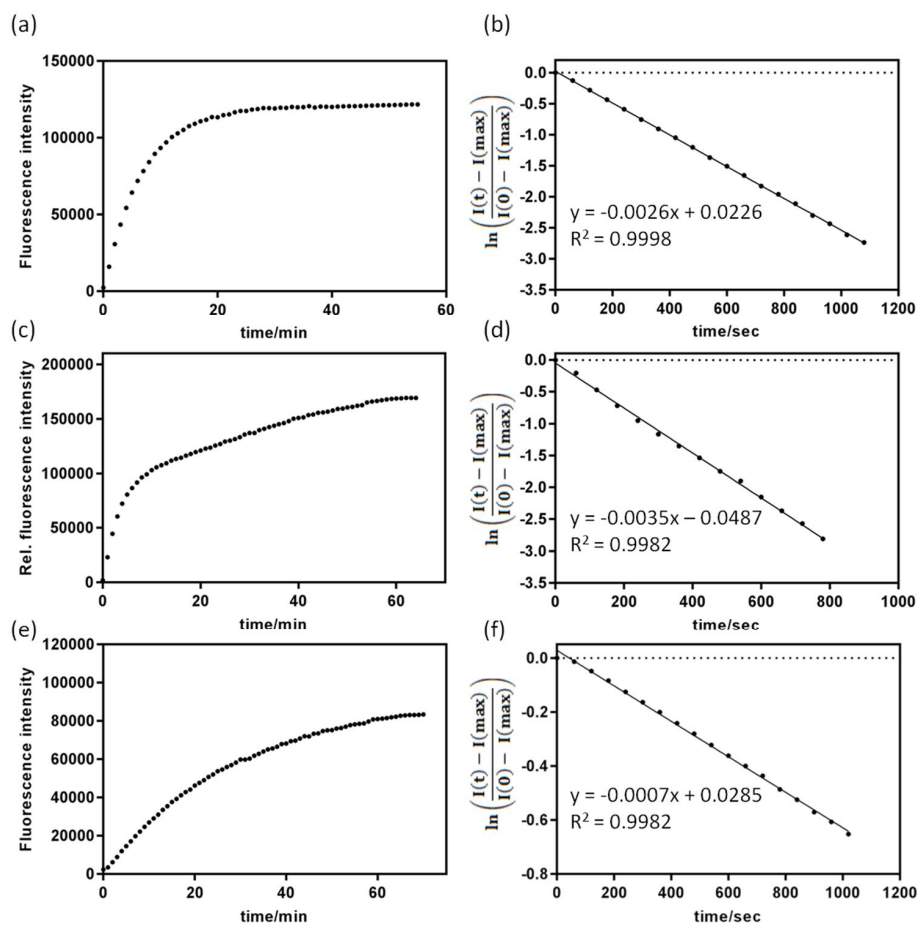
Figure S35. Kinetic profiles (a, c and e) and the pseudo-first order plots (b, d and f) of 10 μM **CAP488** towards (a and b) dopamine ($k_{obs} = 2.6 \times 10^{-3}$ s^{-1}); (c and d) adrenaline ($k_{obs} = 3.5 \times 10^{-3}$ s^{-1}); and (e and f) noradrenaline ($k_{obs} = 0.7 \times 10^{-3}$ s^{-1}). Fluorescent intensity was measured at 488 nm.

Table S1.

[**CAP488**] (µM)	[dopamine] (µM)	[adrenaline] (µM)	[noradrenaline] (µM)	initial rate (s^{-1})
5	2.5	–	–	218
5	5	–	–	340
5	10	–	–	533
5	–	2.5	–	266
5	–	5	–	447
5	–	10	–	690
5	–	–	2.5	214
5	–	–	5	332
5	–	–	10	484
7.5	10	–	–	738
10	10	–	–	1007
7.5	–	10	–	1020
10	–	10	–	1466
7.5	–	–	10	702
10	–	–	10	927

Table S2.

	reaction order
Dopamine	0.64 ± 0.01
Adrenaline	0.69 ± 0.06
Noradrenaline	0.59 ± 0.04
CAP488	0.99 ± 0.13

Cell Culture and Fluorescence Imaging of Live PC12 Cells

Rat pheochromocytoma PC12 cell line was purchased from American Type Culture Collection (Manassas, VA, USA) and the cell culture was maintained in Dulbecco's Modified Eagle Medium (DMEM) supplemented with 10 % horse serum and 5 % fetal bovine serum (Gibco) as previously described.[16-18] To induce neuronal differentiation, PC12 cells were seeded onto collagen-I-coated six-well plates at a density of 2×10^5/mL and treated with 20 ng/mL nerve growth factor (NGF) (Sigma Aldrich, St Louis, MO, USA) in the differentiation medium (DMEM with 1 % horse serum and 1 % fetal bovine serum) for 72 hours. For inhibition of the NGF receptor TrkA, 50 nM of K252a (Abcam, Cambridge, MA, USA) was added to the culture medium together with the NGF and the cells were stimulated for 72 hours. For studying dopamine level in a Parkinson's disease model, PC12 cells were differentiated with 20 ng/mL NGF for 72 hours and further treated with 4 mM of MPP^+ (Sigma Aldrich, St Louis, MO, USA) for 24 hours. Before imaging experiments, the cells were washed twice with PBS and then incubated with 20 μM of **CAP488** and 5 μg/mL Hoechst 33342 (Sigma Aldrich, St Louis, MO, USA) in PBS at 37 °C for 30 min. The cells were washed with PBS to remove excess probe in the culture medium. Fluorescent cell imaging was performed with a Carl Zeiss Axio Observer 3 fluorescence microscope with a 20x objective lens (Carl Zeiss, Jena, Germany) and filter-based monochromator was used for image acquisition. **CAP488** was excited at 495 nm and emission collected at 517 nm and Hoechst 33342 was excited at 359 nm and emission collected at 463 nm. The images were analyzed with Image J and the averaged fluorescent intensity of the entire field was measured. Cell viability was evaluated by a standard colorimetric MTT reduction assay. PC12 cells were seeded onto collagen I-coated 96-well plates at the density of 1.5×10^5/mL and treated with **CAP488** at concentrations ranging from 0 to 50 μM for 2 hours. The cells were then washed with PBS to remove excess probe and incubated with MTT solution (0.5 mg/mL) in PBS for 4 hours. The formation of purple formazan was quantified by measuring the absorbance at 540 nm using a Bio-Rad microplate reader (Hercules, CA, USA).

Preparation of cell lysate sample for HPLC analysis was based on previous literature.[19] Briefly, PC12 cells were seeded onto collagen I-coated 100 mm dishes at the density of 2.4×10^5/mL and treated with or without 20 ng/mL nerve growth factor (NGF) in the differentiation medium (DMEM with 1% horse serum and 1% fetal bovine serum) for 72 hours. At the end of the treatment, the cells were harvested and mixed with an extraction buffer consisting of 50 % acetonitrile, 40% of 0.1 M HCl and 10% of 27 mM EDTA in water. The cells were sonicated for 20 min in an ice-water bath. The cells were centrifuged at 6,500g for 15 min at 4°C. The supernatant was prepared for the analysis. Chromatographic analysis was performed on an Agilent HPLC system with a HPLC C18 column (ZORBAX Eclipse XDB-C18, 4.6v x 150 mm, 80 Å, 5 μM). The mobile phase consists of solvent A:

0.1% formic acid in water and solvent B: 0.1% formic acid in methanol. Chromatographic separation was achieved using isocratic elution of 99% A for 10 min, at a flow rate of 0.5 mL/min. Detection was performed with AB SCIEX 4000 QTRAP® system, operating in positive electrospray ionization (ESI) and multiple reaction monitoring (MRM) mode. The MRM transitions monitored were as follows: m/z 152.0 → 107.5 for noradrenaline; m/z 184.0 → 166.2 for adrenaline and m/z 154.0 → 137.2 for dopamine. The ion source parameters were as follows: ion-spray voltage of 4500V, temperature of 400°C, medium collision gas, curtain gas of 40 Psig, ion source gas 1 and 2 of 50 Psig. The compound dependent parameters were set as de-clustering potential of 25 V, entrance potentials of 10 V, collision cell exit potential of 10 V, and collision energy of 25 V for noradrenaline while 15 V for both adrenaline and dopamine.
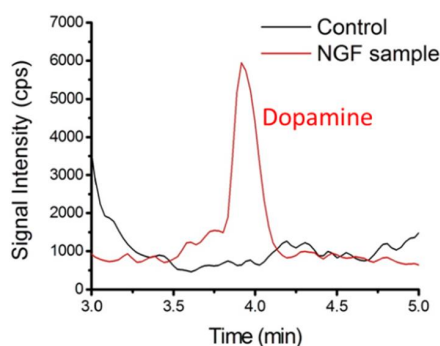
Figure S36. Overlaid chromatograms for MRM transition (dopamine: 154.0 →137.2) in vehicle control and NGF-treated samples.
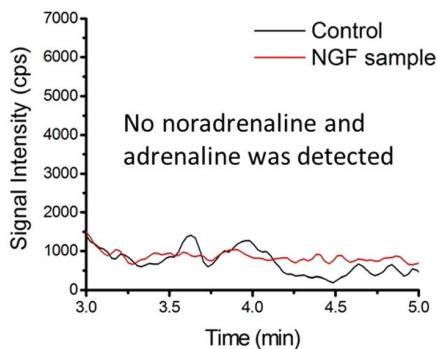
Figure S37. Overlaid chromatograms for MRM transition (Noradrenaline: 152.0→107.5; adrenaline: 184.0 →166.2) in vehicle control and NGF-treated samples.
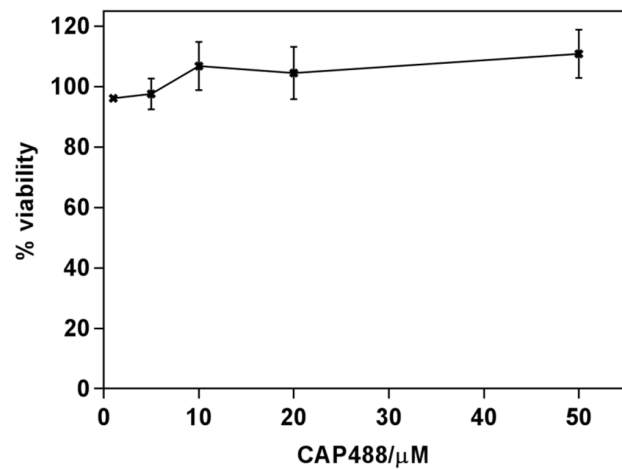
Figure S38. Percentage viability of PC12 cells at different **CAP488** concentrations by MTT assay. Error bars are ±SD (n = 3).

NMR
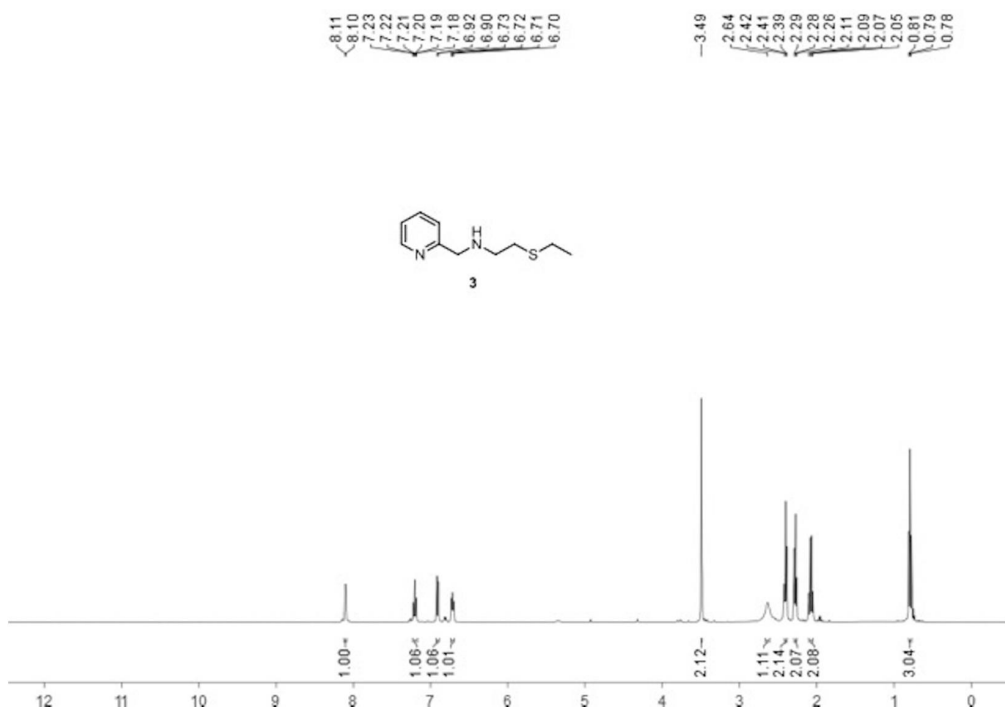
Figure S39. ^{1}H NMR (400 MHz, CDCl₃, 298K) of **3.**
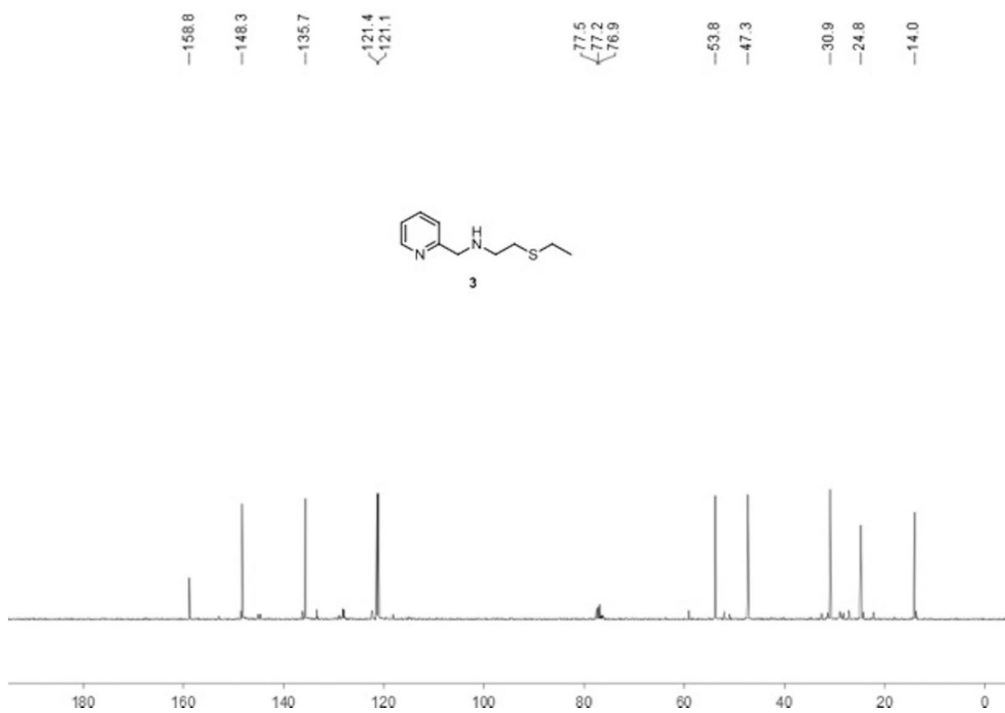
Figure S40. ^{13}C{^{1}H} NMR (101 MHz, CDCl₃, 298K) of **3**.
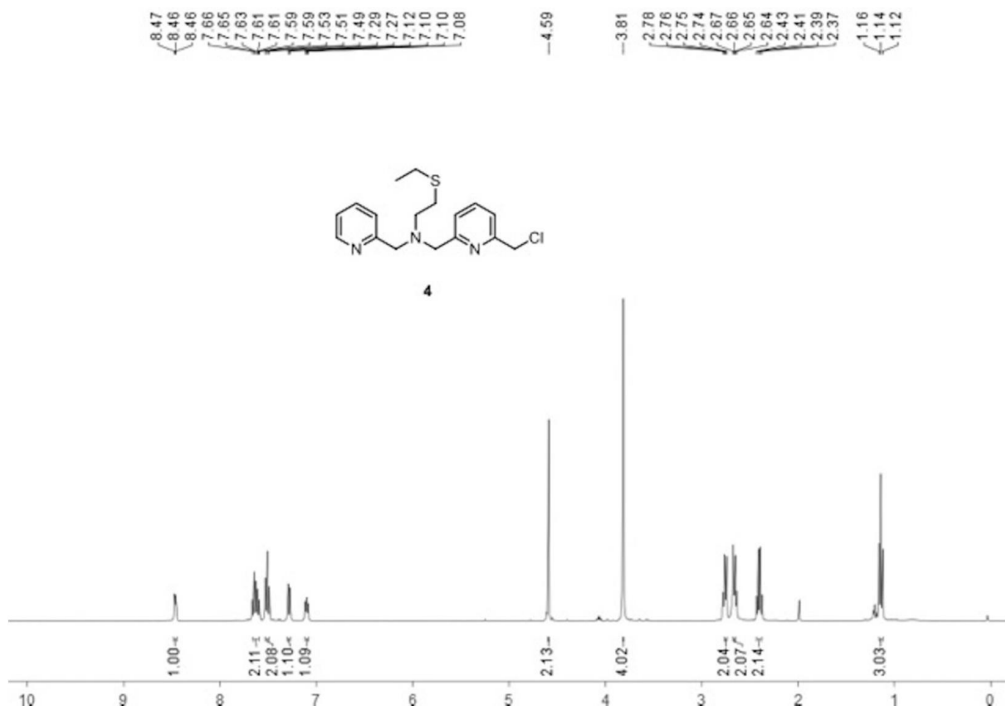
Figure S41. ^{1}H NMR (400 MHz, CDCl$_3$, 298K) of **4**.
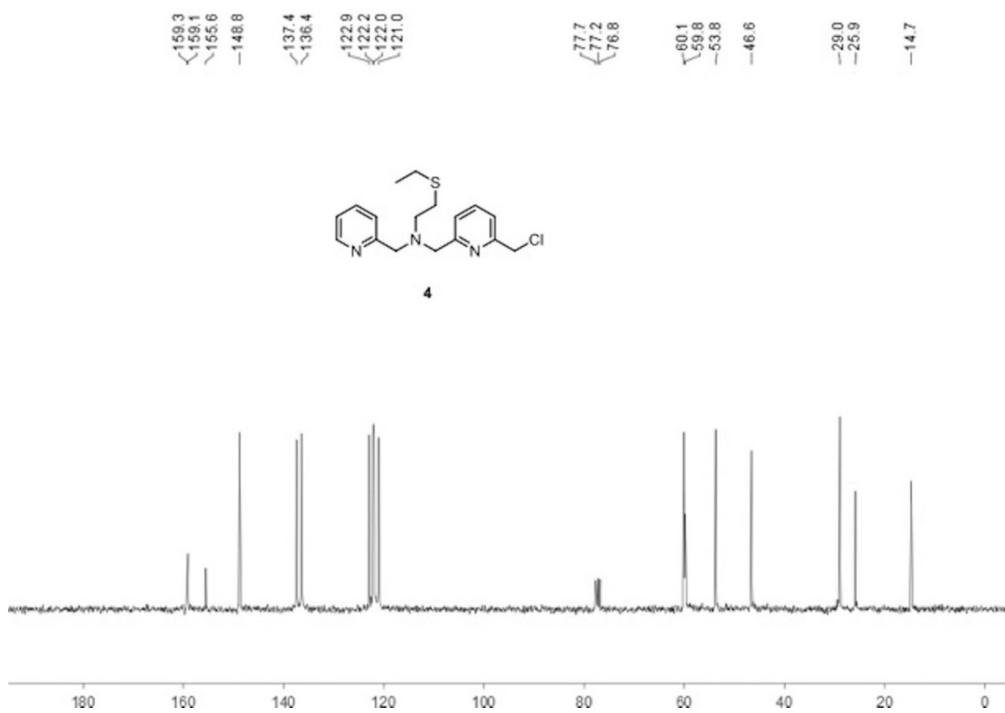
Figure S42. ^{13}C{^{1}H} NMR (75 MHz, CDCl$_3$, 298K) of **4**.
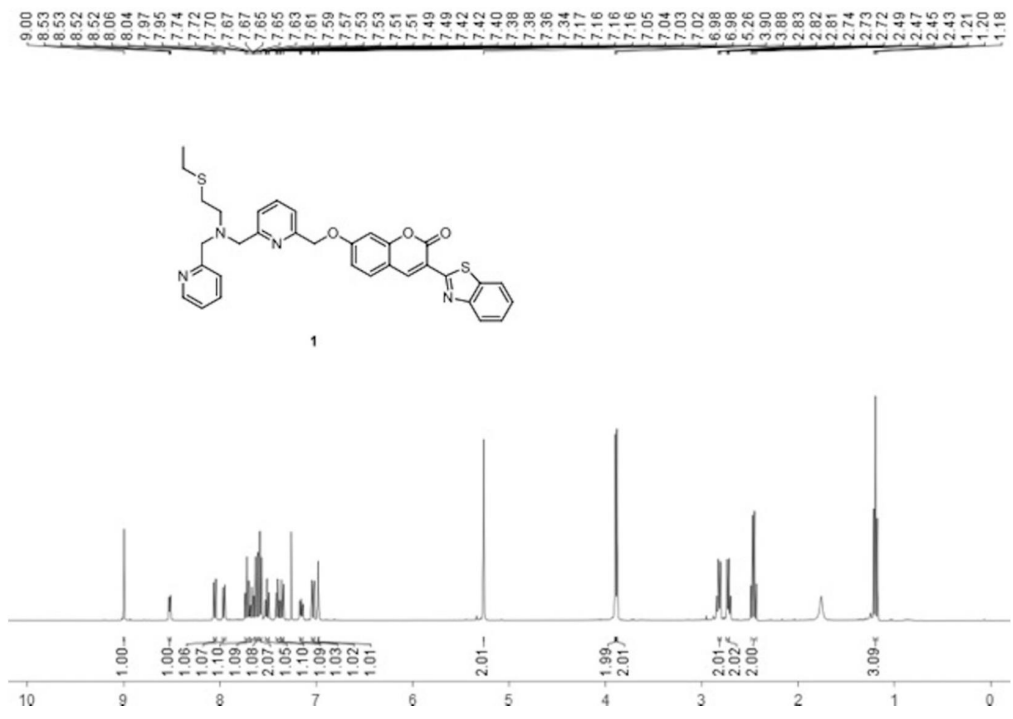
Figure S43. ^{1}H NMR (400 MHz, CDCl$_3$, 298K) of **1**.
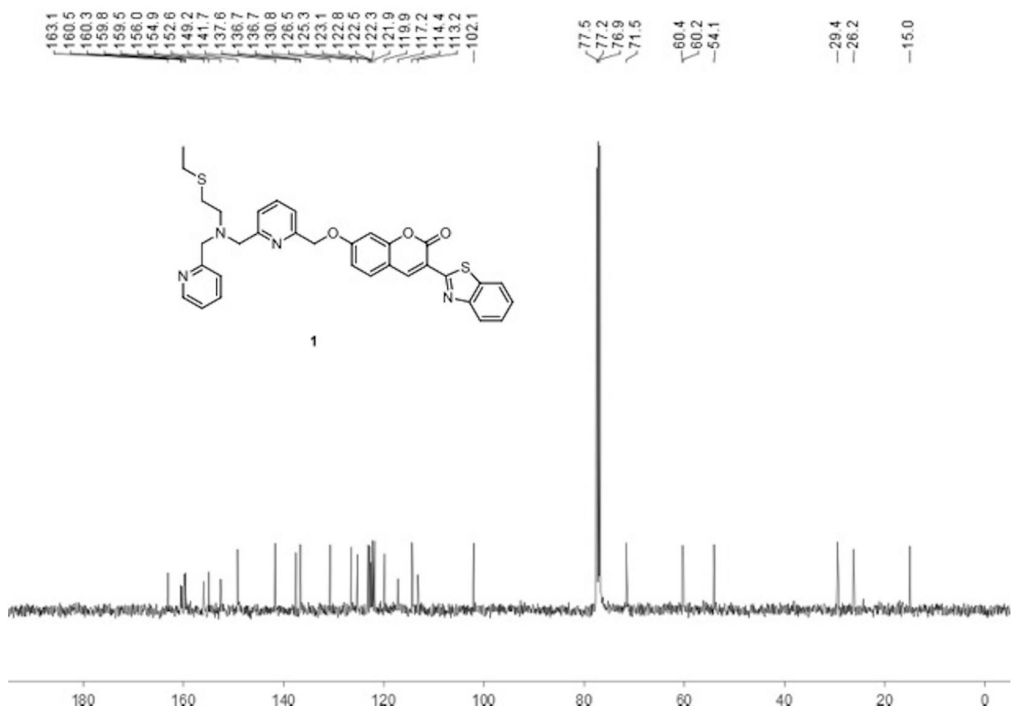
Figure S44. ^{13}C{^{1}H} NMR (101 MHz, CDCl$_3$, 298K) of **1**.
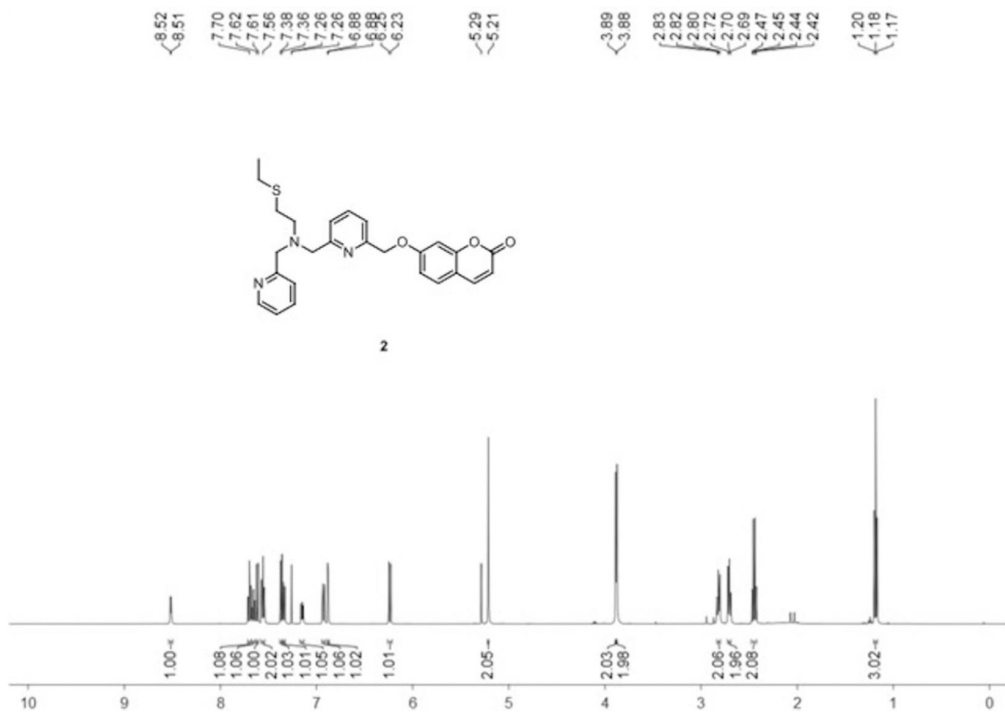
Figure S45. ^{1}H NMR (500 MHz, CDCl$_3$, 298K) of **2**.
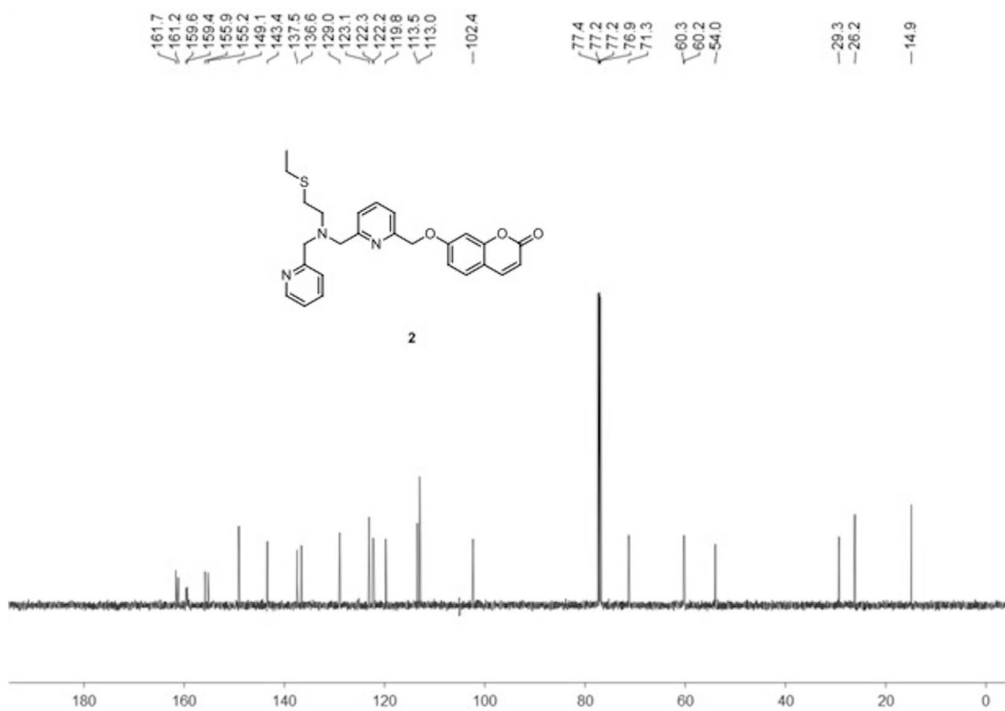
Figure S46. ^{13}C{^{1}H} NMR (126 MHz, CDCl$_3$, 298K) of **2**.

(a)

CAP488

(b)
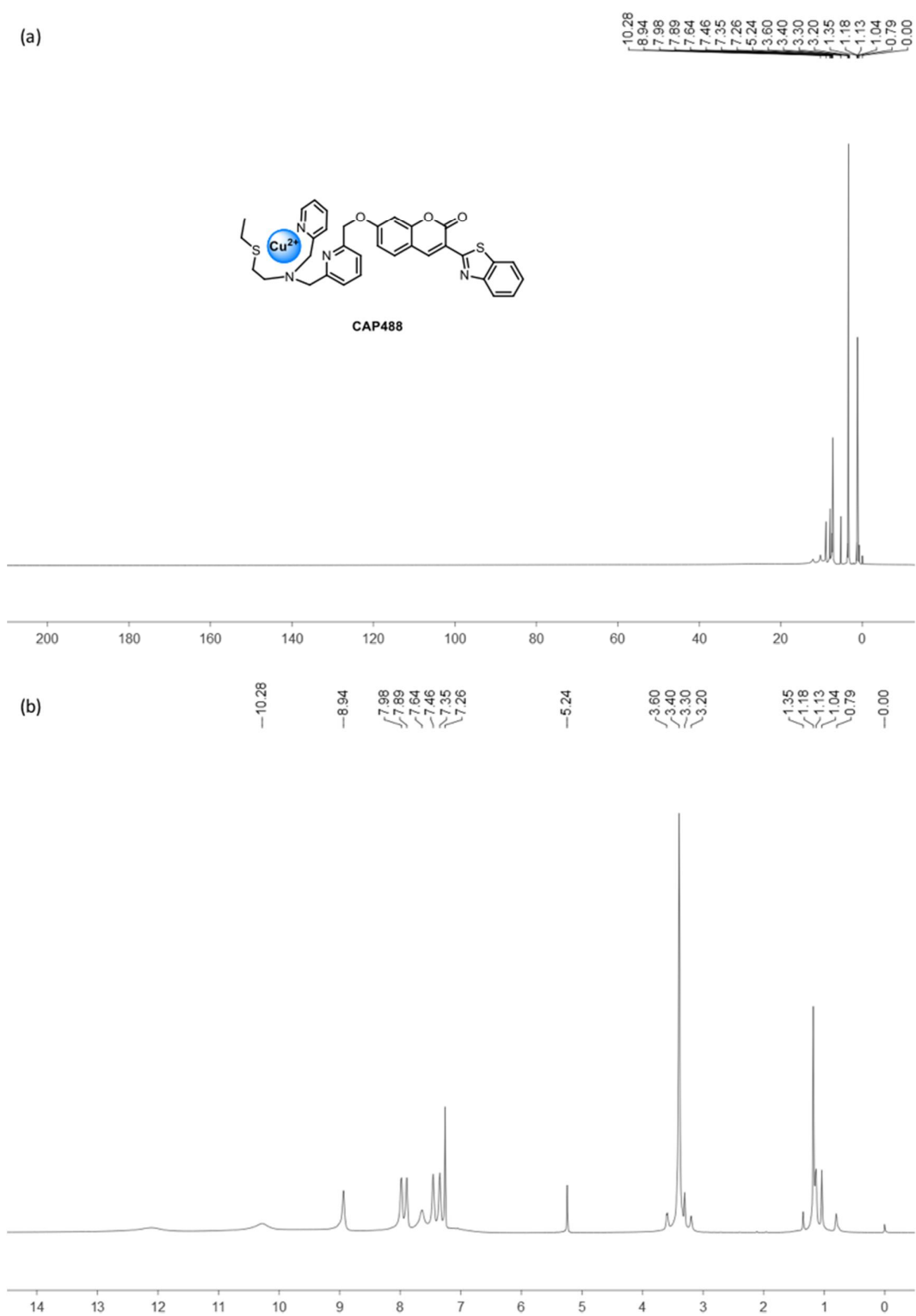
Figure S47. ^{1}H NMR (500 MHz, 9:1 CDCl$_3$/MeOD, 298K) of **CAP488** recorded at (a) -12 to 210 ppm; (b) 0 to 14 ppm.

(a)

11.97
9.89
7.87
7.54
7.11
6.22
4.86
3.48
3.31
1.17

CAP-R

200 180 160 140 120 100 80 60 40 20 0

(b)

—11.97
—9.89
7.87
7.54
7.11
—6.22
—4.86
3.48
3.31
—1.17

14 13 12 11 10 9 8 7 6 5 4 3 2 1 0
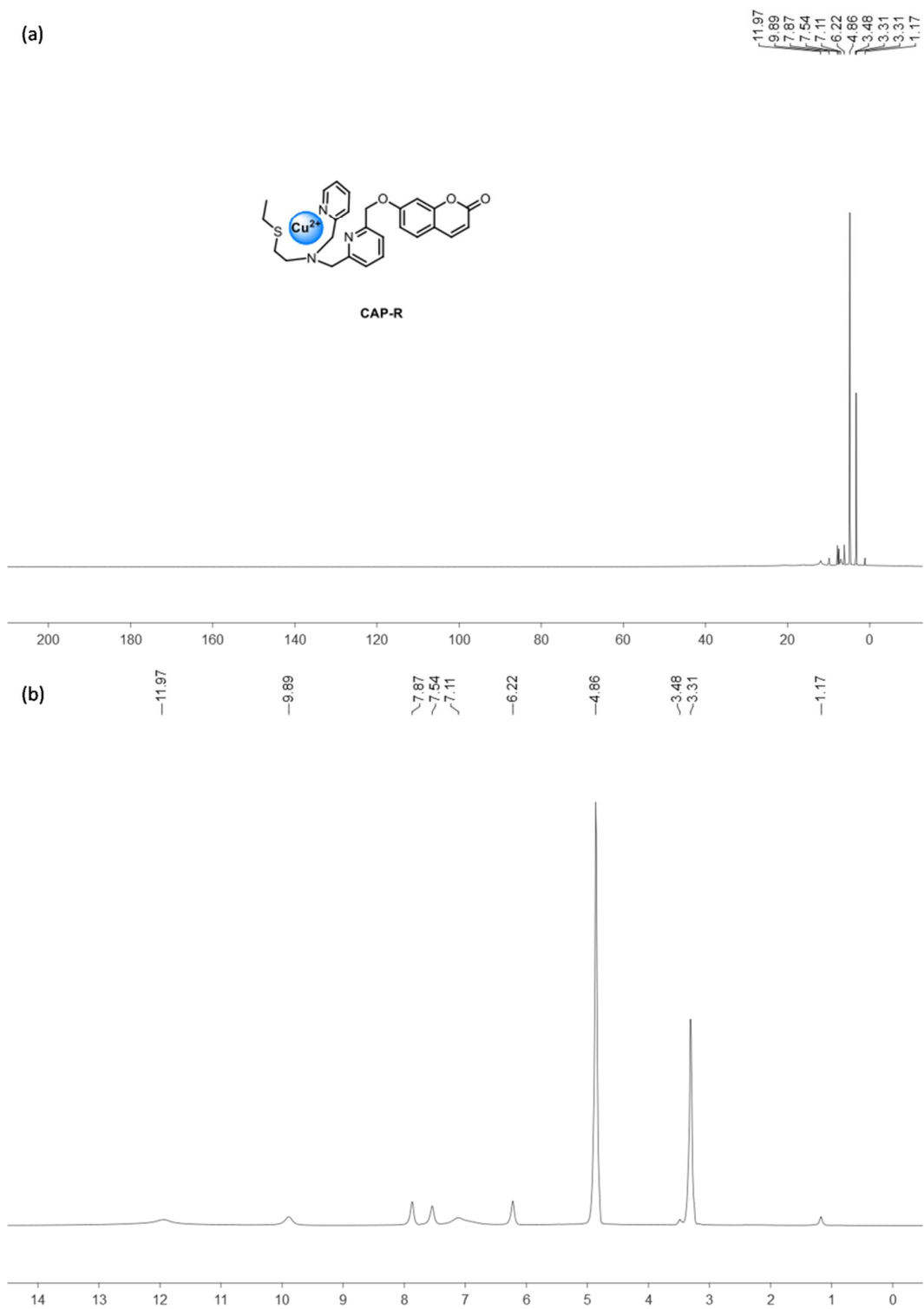
Figure S48. ^{1}H NMR (500 MHz, MeOD, 298K) of **CAP-R** recorded at (a) -12 to 210 ppm; (b) 0 to 14 ppm.
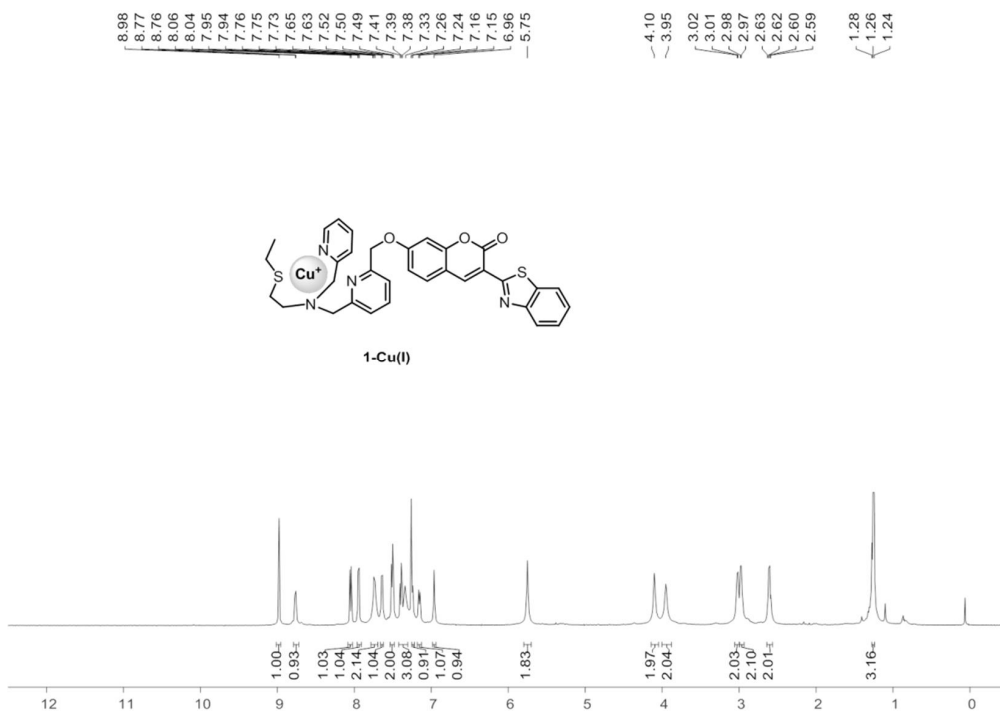
Figure S49. ^{1}H NMR (500 MHz, CDCl$_3$, 298K) of **1-Cu(I)**.
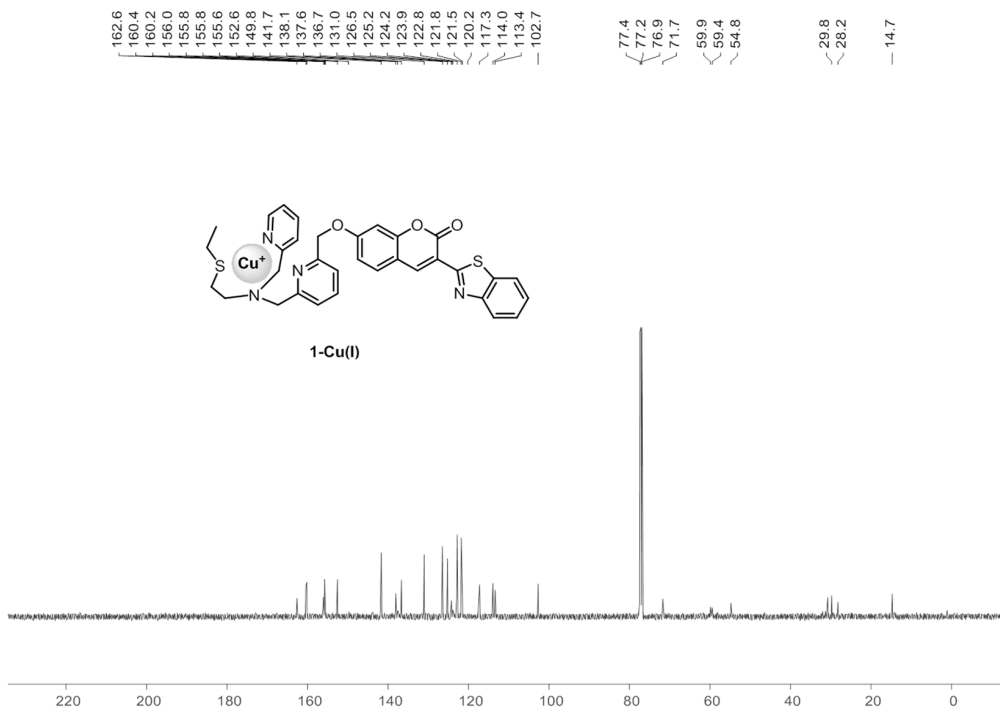
Figure S50. ^{13}C{^{1}H} NMR (126 MHz, CDCl$_3$, 298K) of **1-Cu(I)**.
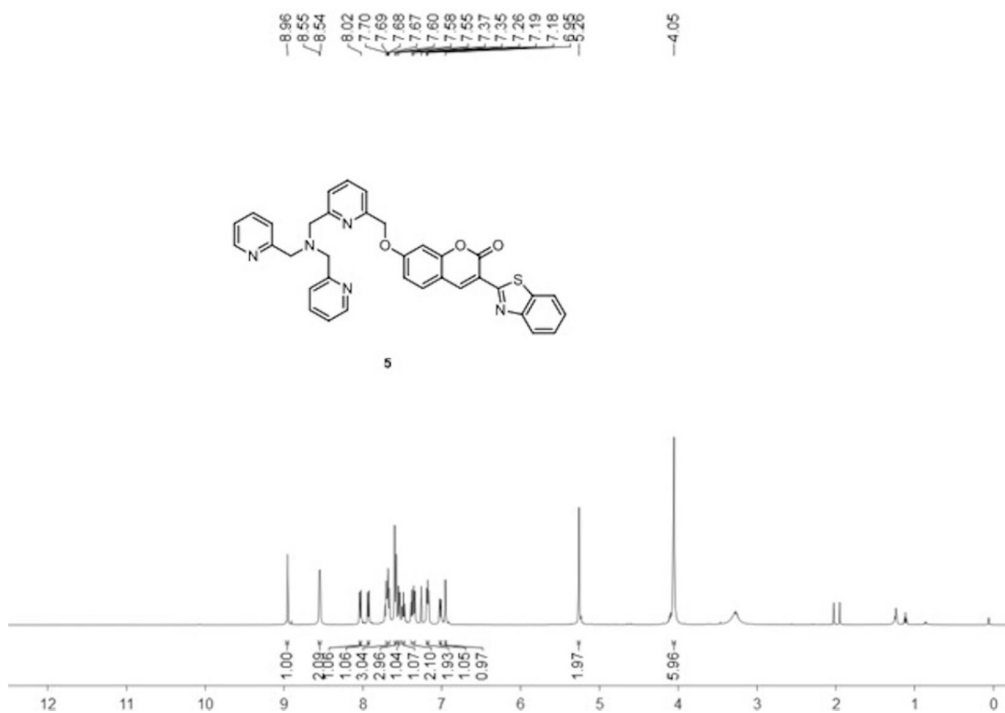
Figure S51. ^{1}H NMR (500 MHz, CDCl$_3$, 298K) of **5**.
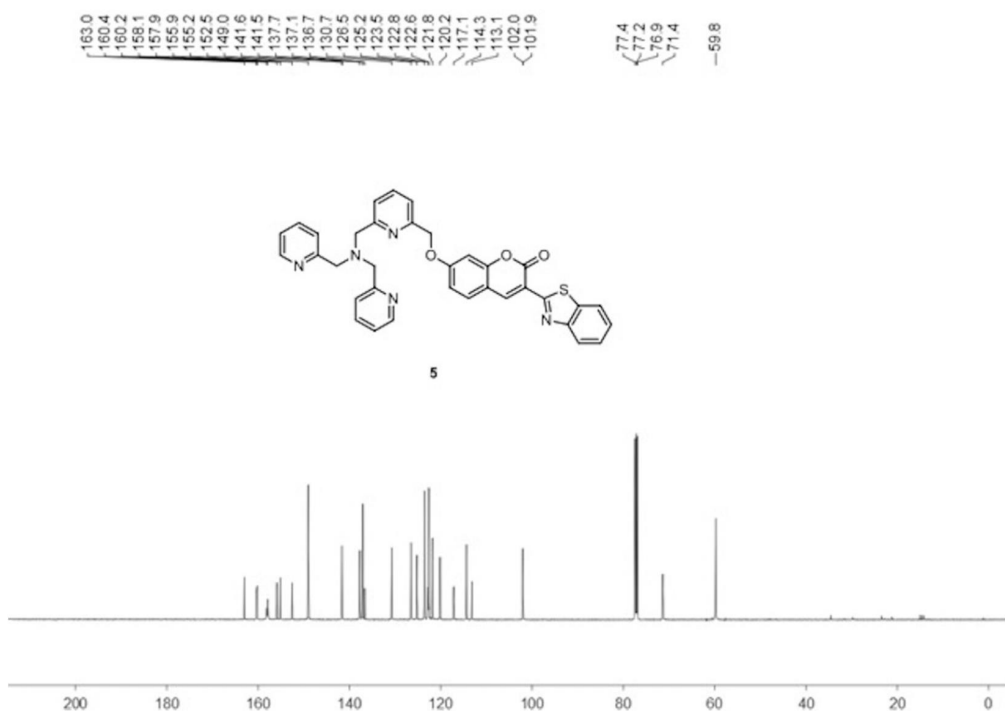
Figure S52. ^{13}C{^{1}H} NMR (126 MHz, CDCl$_3$, 298K) of **5**.

(a)

N₄-cyan

(b)
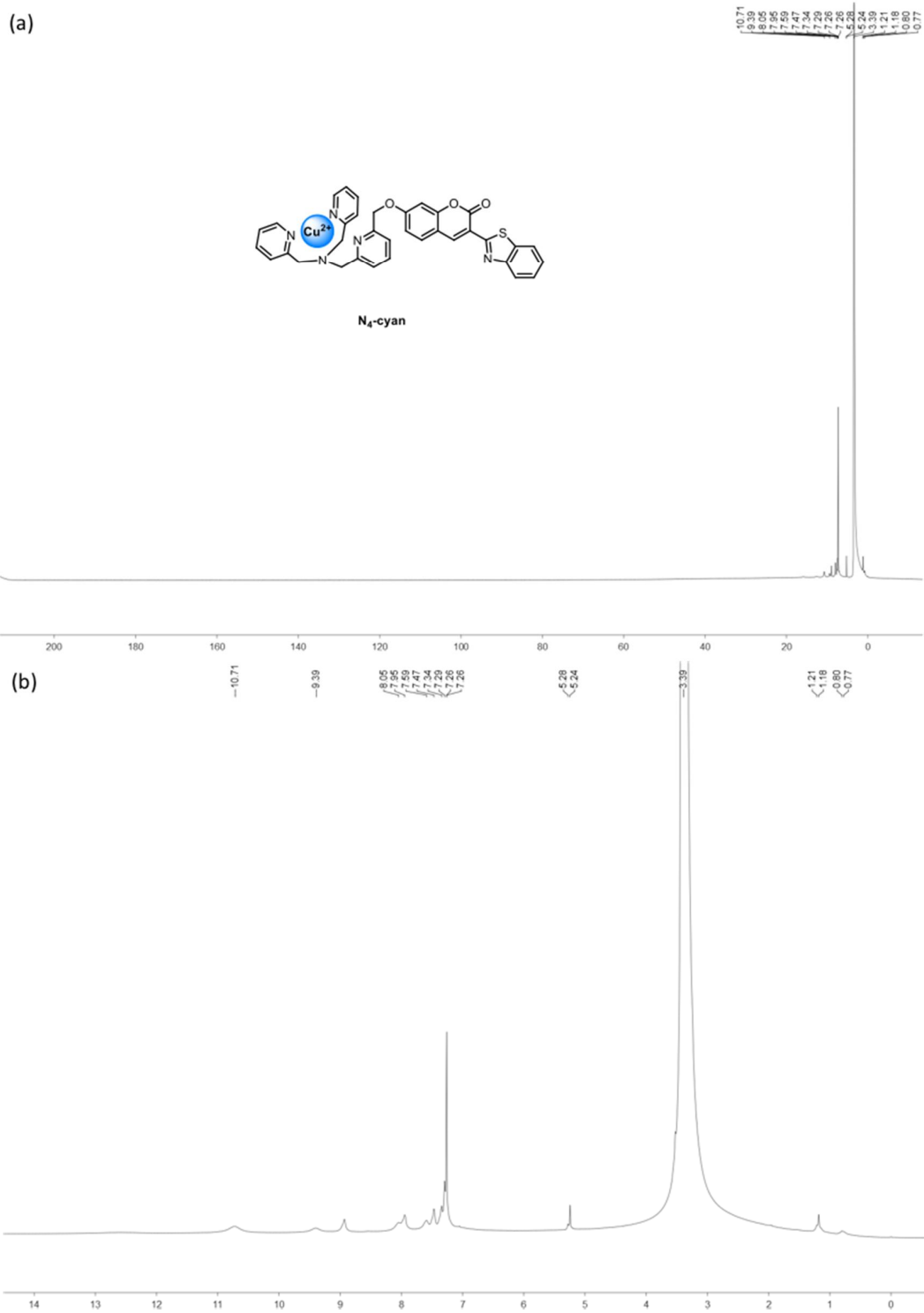
Figure S53. ¹H NMR (500 MHz, 9:1 CDCl₃/MeOD, 298K) of **N₄-cyan** recorded at (a) -12 to 210 ppm; (b) 0 to 14 ppm.
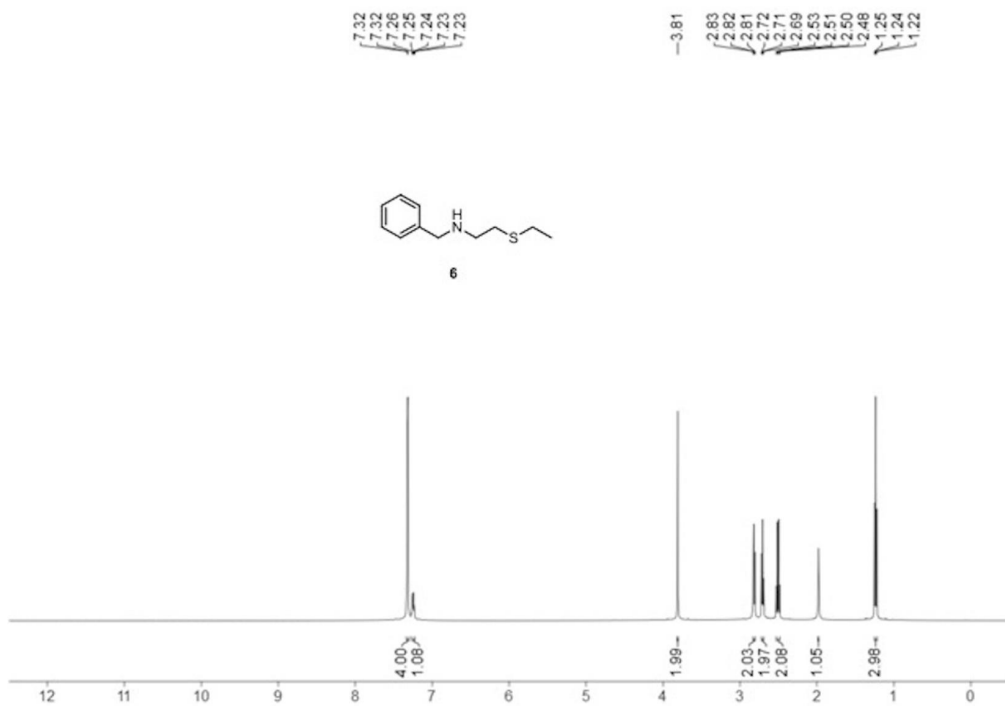
Figure S54. ^{1}H NMR (500 MHz, CDCl$_3$, 298K) of **6**.
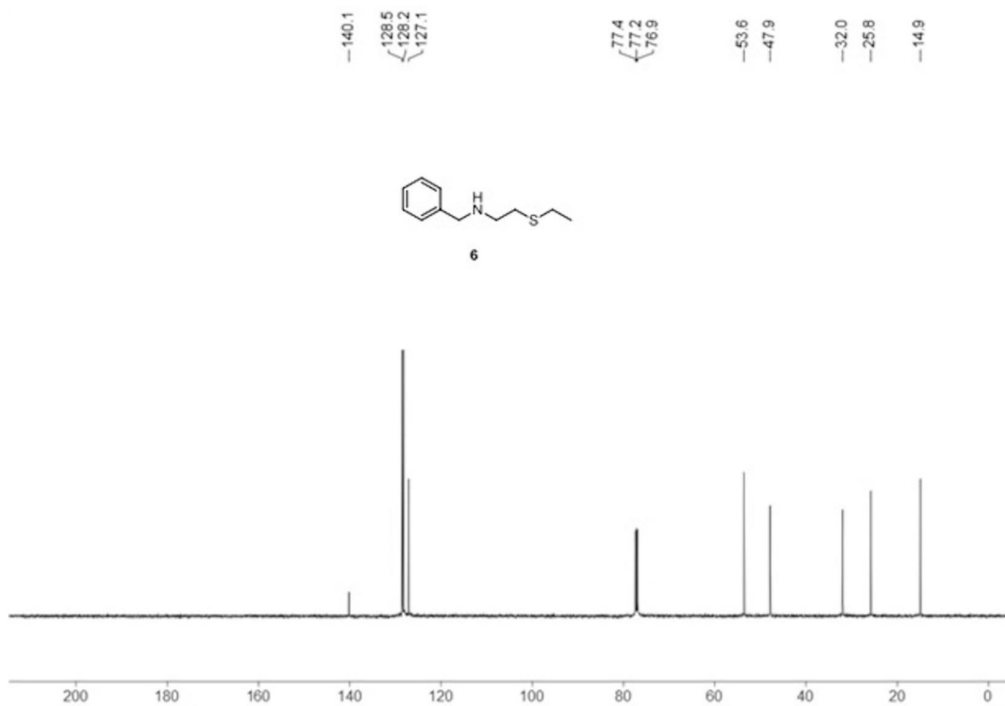
Figure S55. ^{13}C{^{1}H} NMR (126 MHz, CDCl$_3$, 298K) of **6**.

Figure S56. ^{1}H NMR (500 MHz, CDCl$_3$, 298K) of **7**.
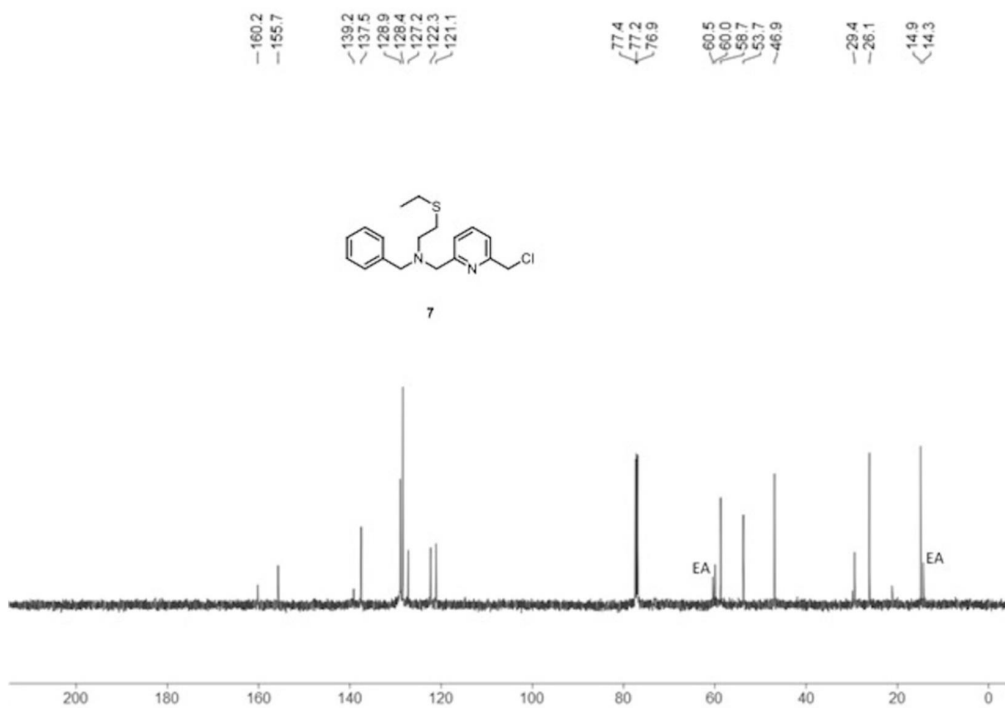
Figure S57. ^{13}C{^{1}H} NMR (126 MHz, CDCl$_3$, 298K) of **7**.
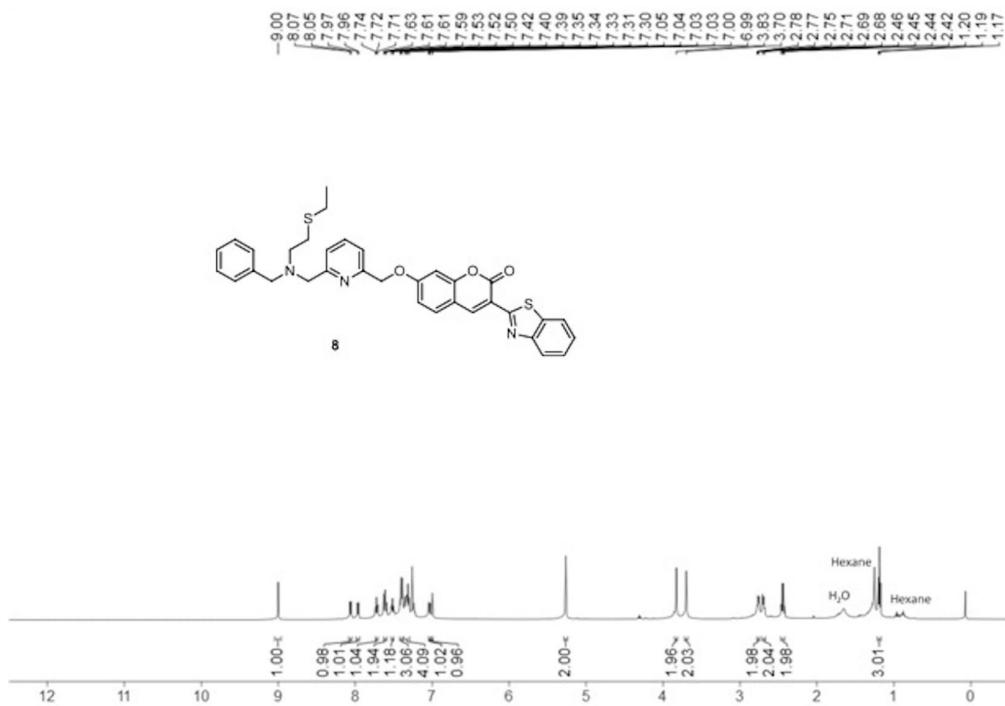
Figure S58. ^{1}H NMR (500 MHz, CDCl$_3$, 298K) of **8**.
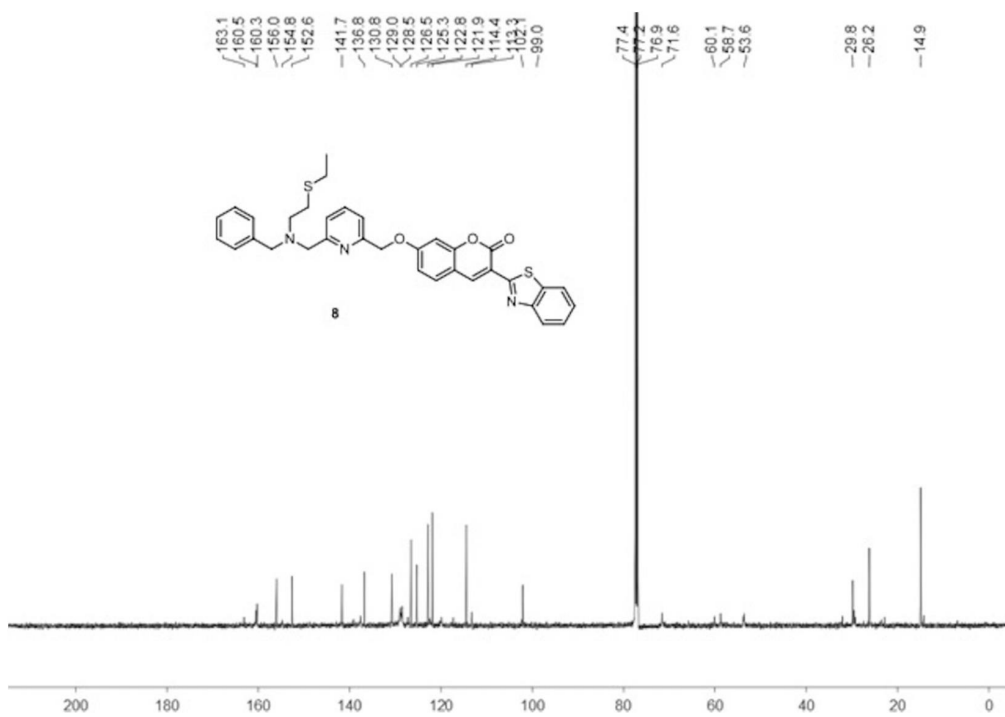
Figure S59. ^{13}C{^{1}H} NMR (126 MHz, CDCl$_3$, 298K) of **8**.
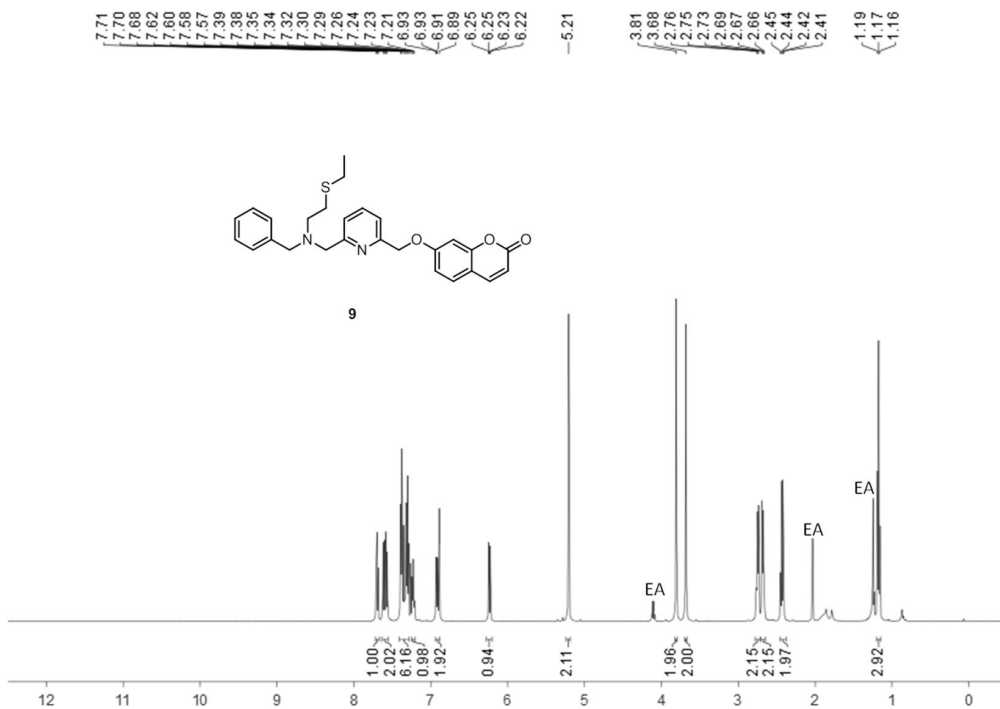
Figure S60. ^{1}H NMR (500 MHz, CDCl$_3$, 298K) of **9**.
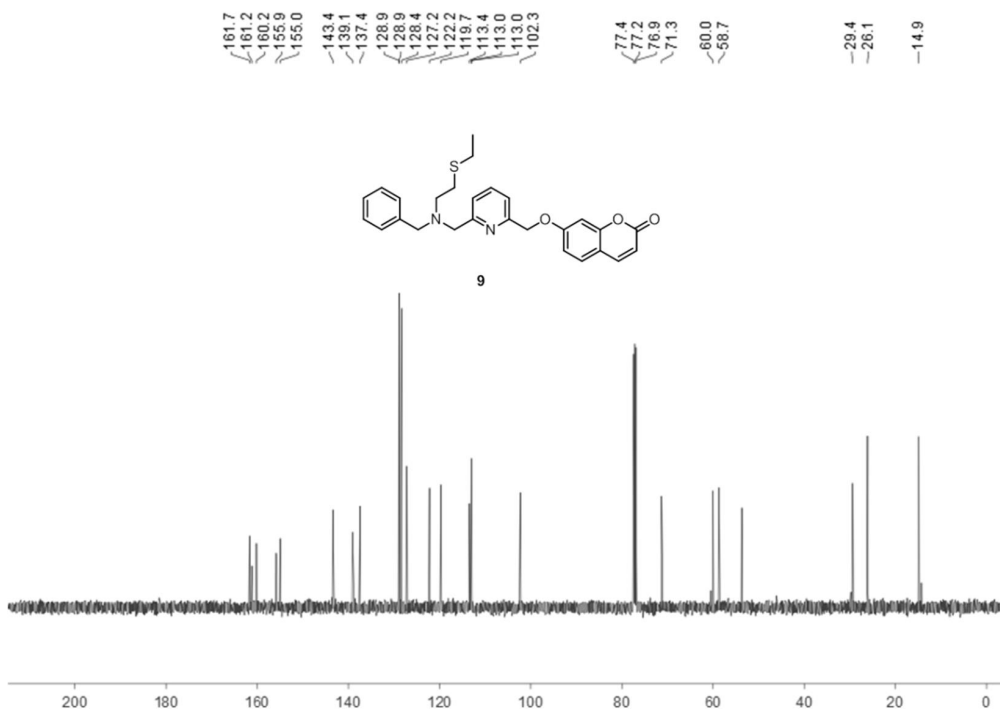
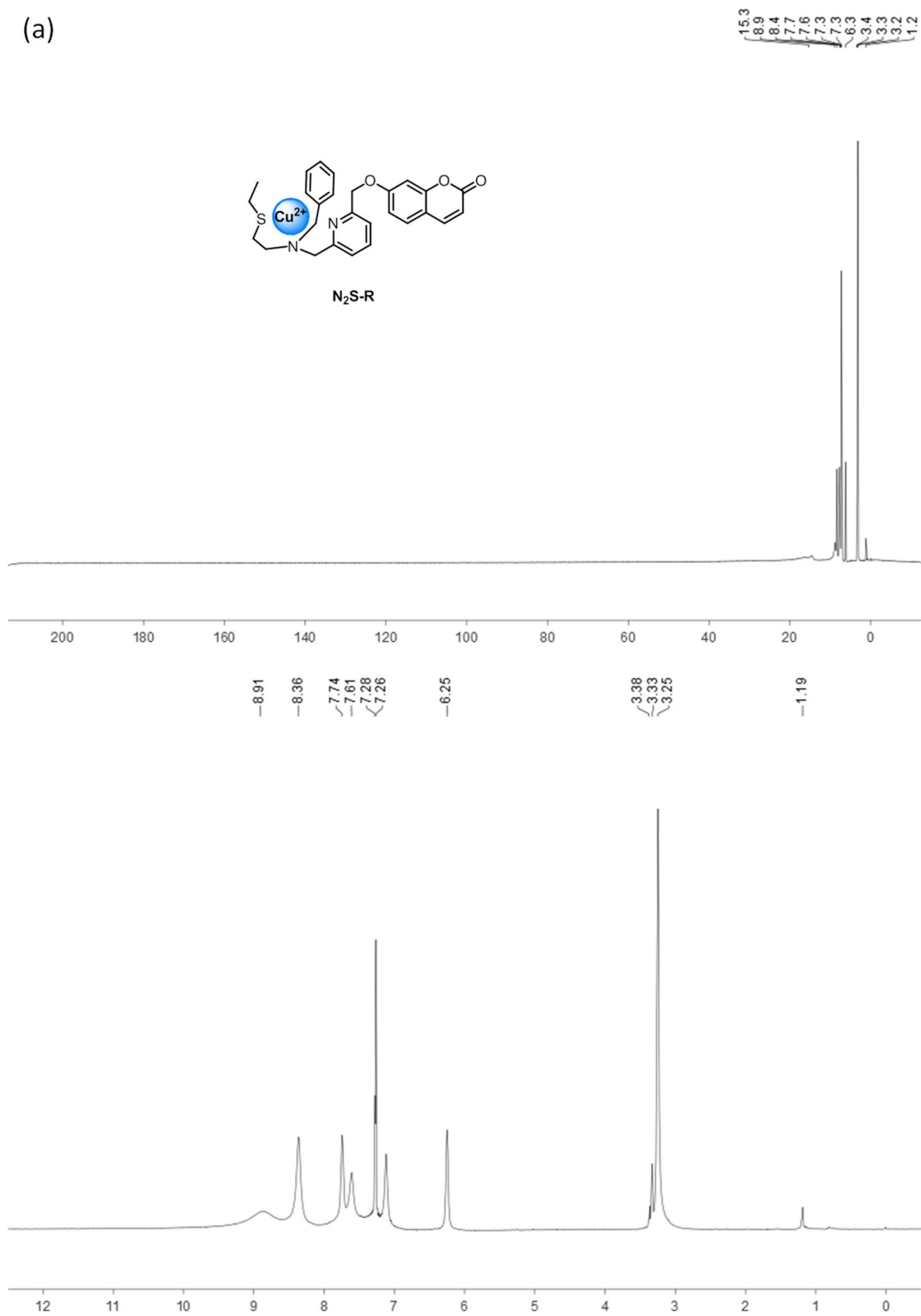**Figure S61.** ^{13}C{^{1}H} NMR (126 MHz, CDCl$_3$, 298K) of **9**.

(a)
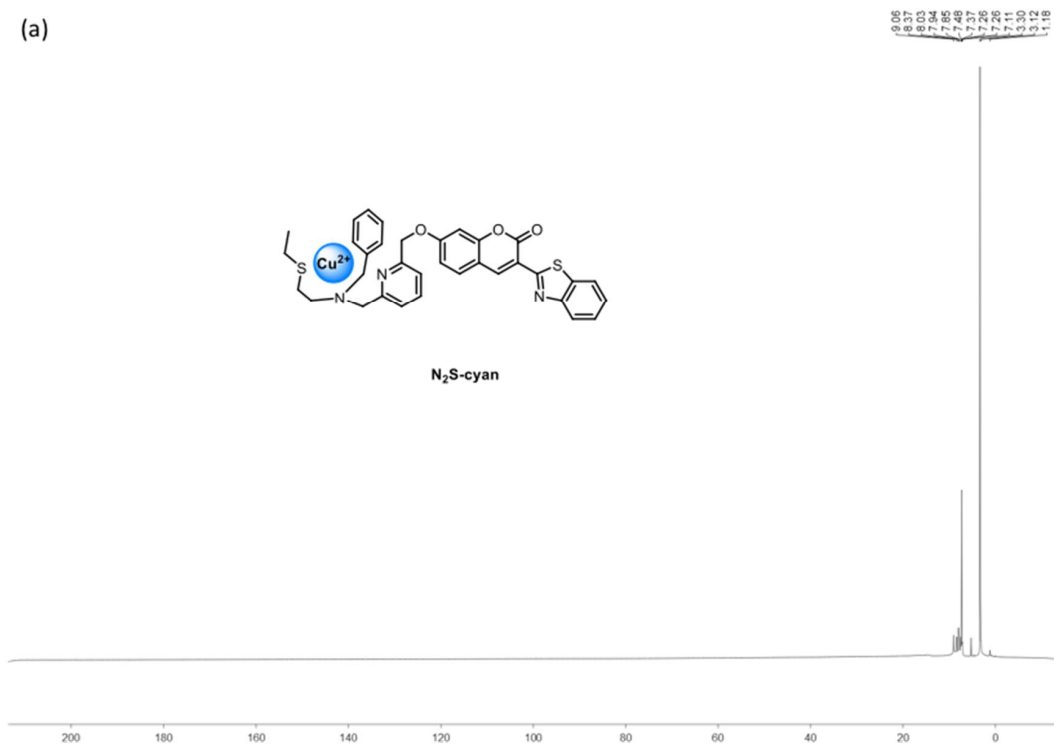
N₂S-cyan

(b)
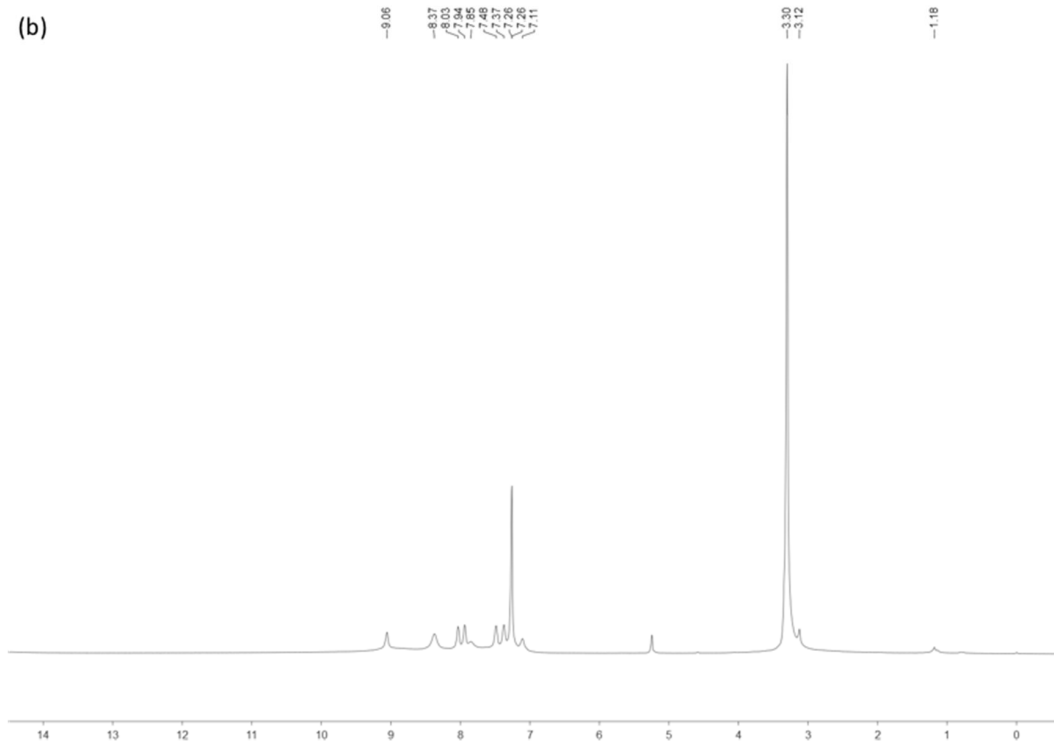
Figure S62. ¹H NMR (500 MHz, 9:1 CDCl₃/MeOD, 298K) of **N₂S-cyan** recorded at (a) -12 to 210 ppm; (b) 0 to 14 ppm.
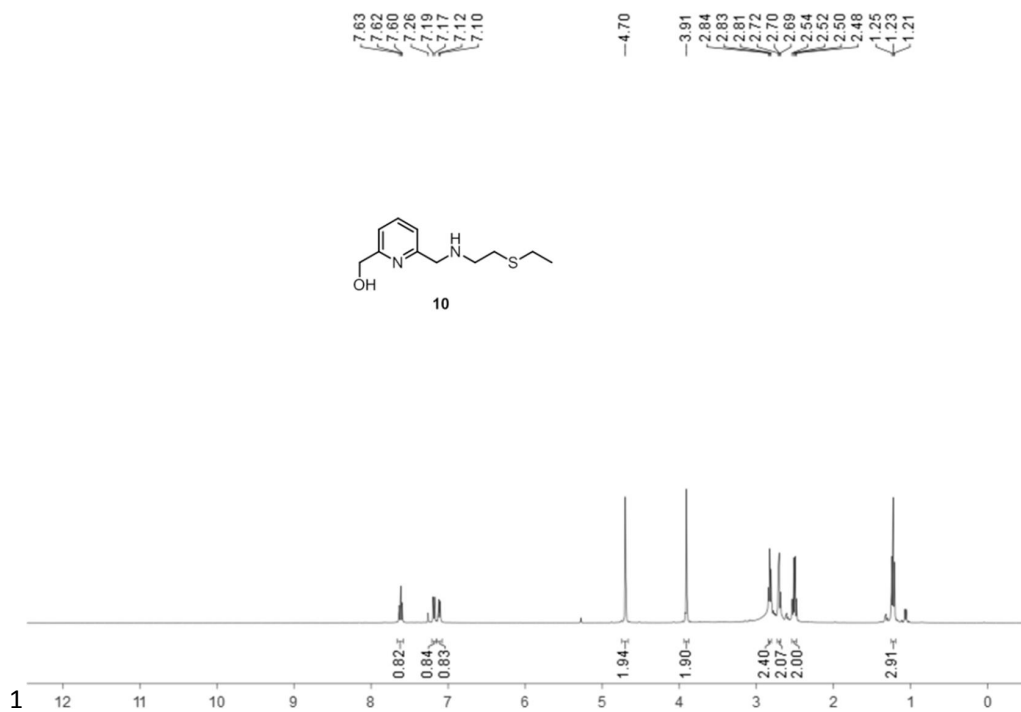
(a)

15.3
8.9
8.4
7.7
7.6
7.3
7.3
6.3
3.4
3.3
3.2
1.2

N₂S-R

200   180   160   140   120   100   80   60   40   20   0

—8.91   —8.36   7.74 7.61   7.28 7.26   —6.25   3.38 3.33 3.25   —1.19

12   11   10   9   8   7   6   5   4   3   2   1   0

Figure S63. ^{1}H NMR (500 MHz, 9:1 CDCl$_3$/MeOD, 298K) of **N₂S-R** recorded at (a) -12 to 210 ppm; (b) 0 to 14 ppm.

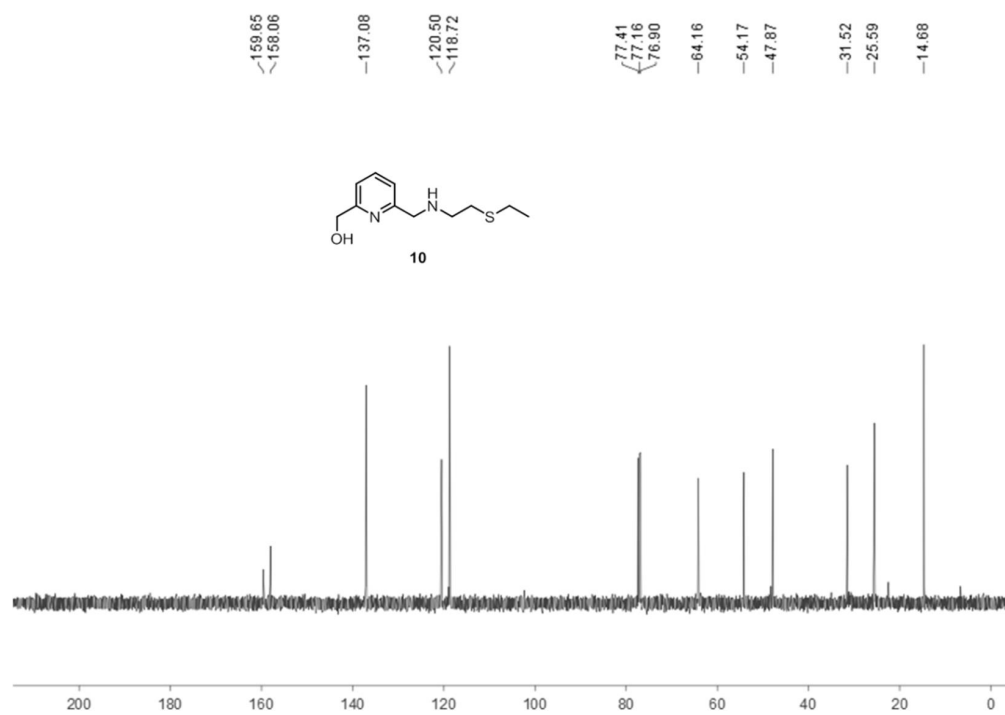**Figure S64.** ^{1}H NMR (400 MHz, CDCl$_3$, 298K) of **10**

Figure S65. ^{13}C{^{1}H} NMR (126 MHz, CDCl$_3$, 298K) of **10**
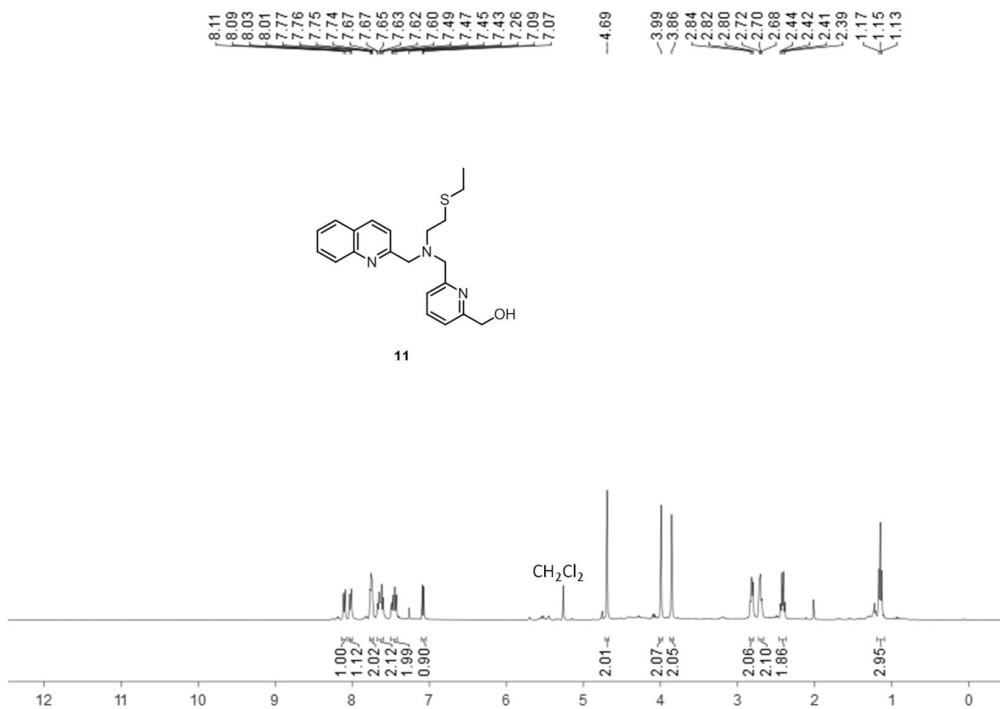
Figure S66. ^{1}H NMR (400 MHz, CDCl$_3$, 298K) of **11**
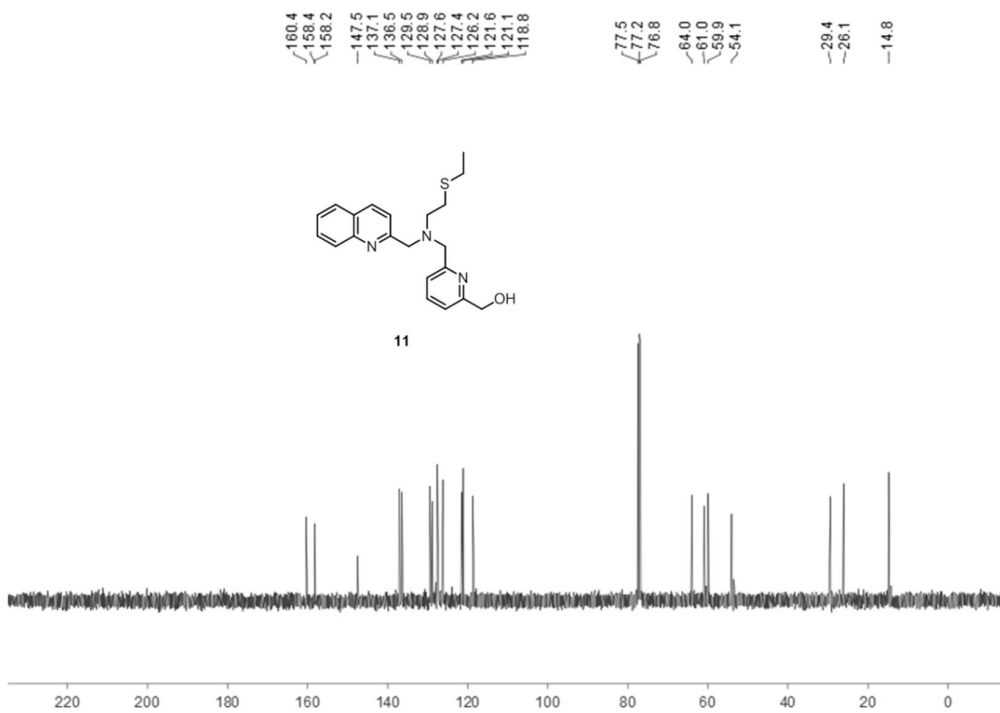
Figure S67. ^{13}C{^{1}H} NMR (101 MHz, CDCl$_3$, 298K) of **11**
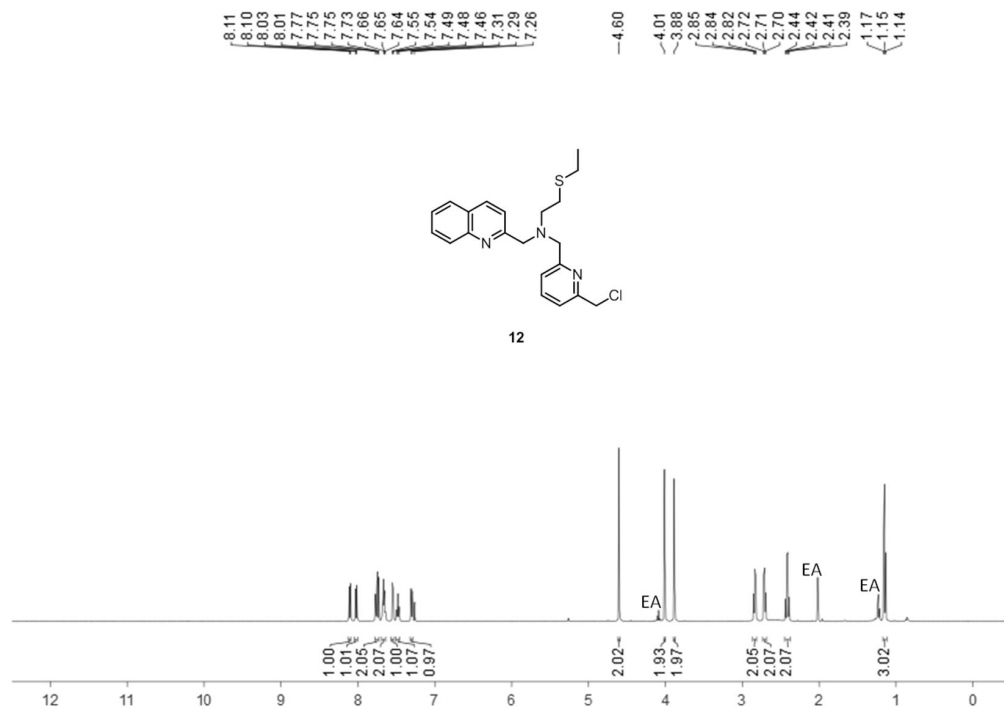
Figure S68. ^{1}H NMR (500 MHz, CDCl$_3$, 298K) of **12**
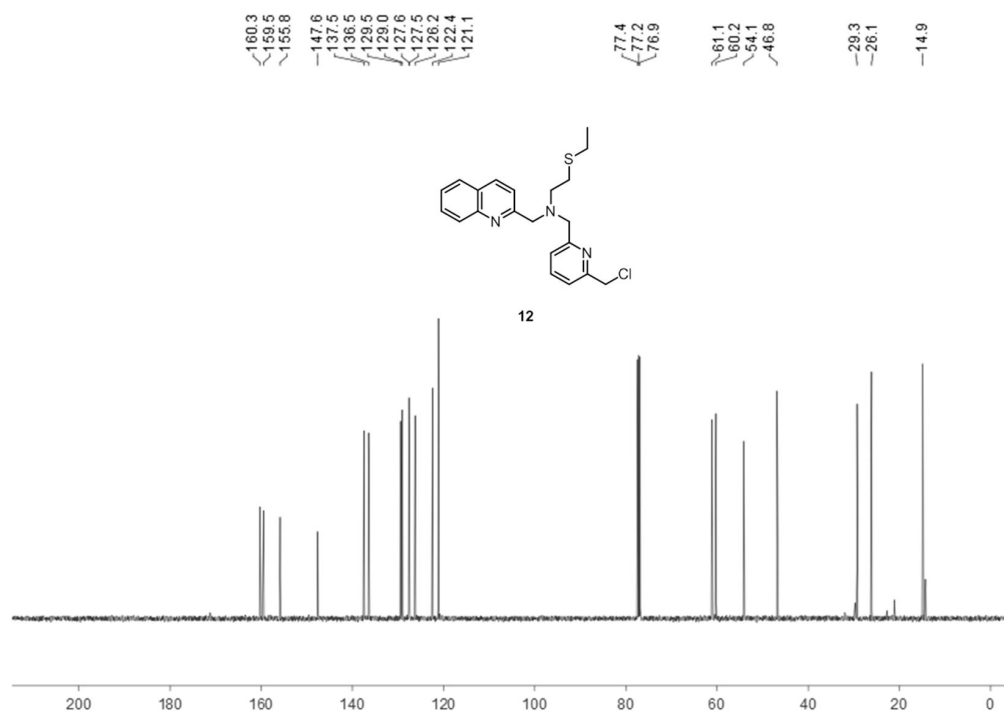
Figure S69. ^{13}C{^{1}H} NMR (126 MHz, CDCl$_3$, 298K) of **12**
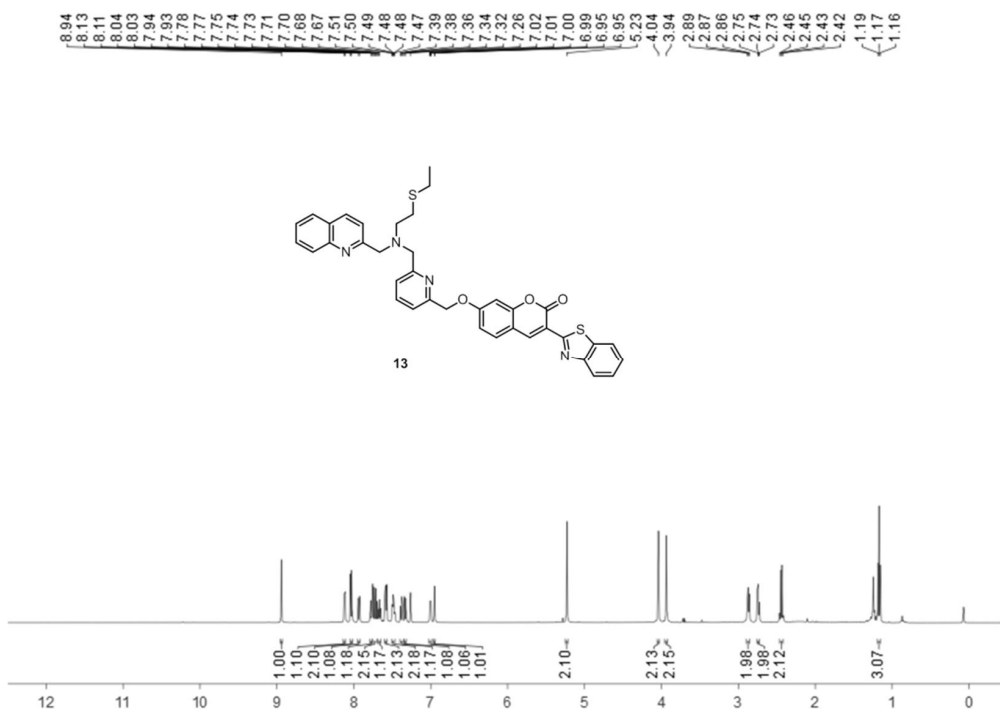
Figure S70. ^{1}H NMR (500 MHz, CDCl$_3$, 298K) of **13**.
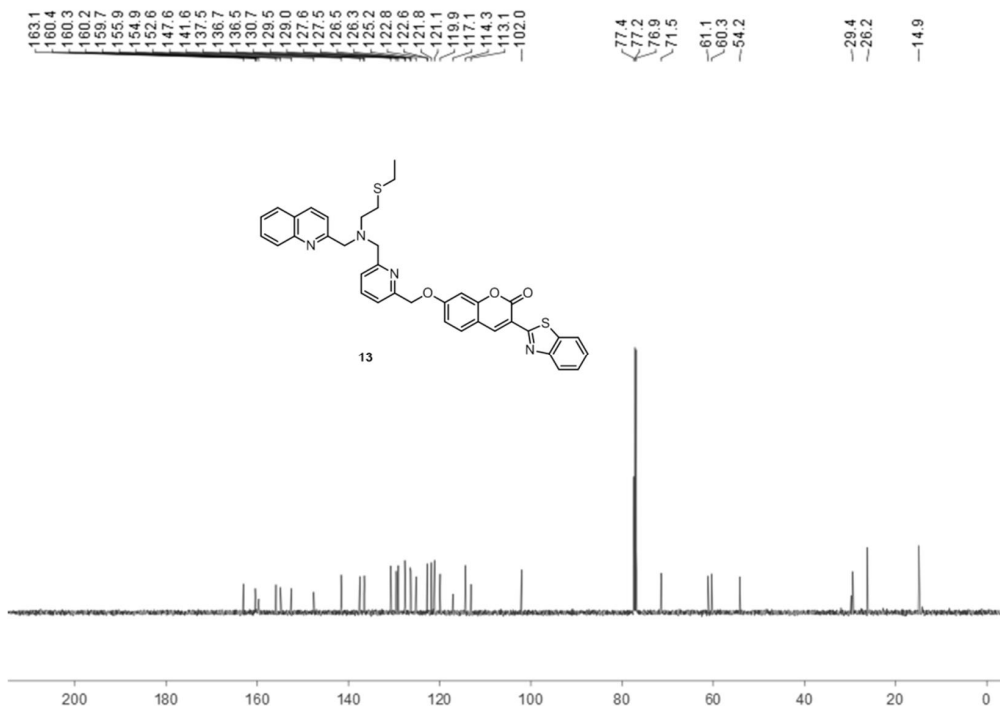
Figure S71. ^{13}C{^{1}H} NMR (126 MHz, CDCl$_3$, 298K) of **13**

(a)

N3qS-cyan

(b)
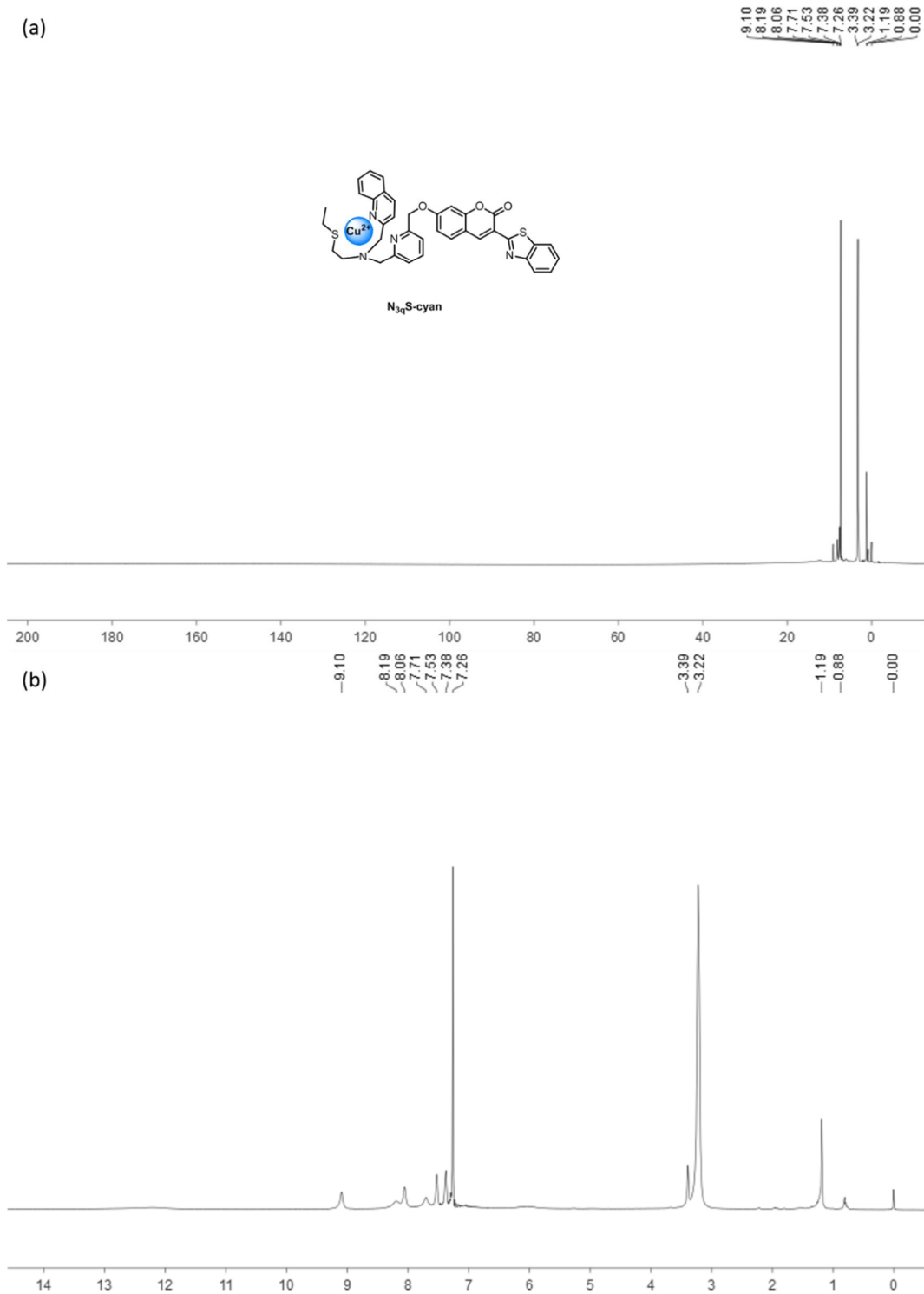
Figure S72. ^{1}H NMR (500 MHz, 9:1 CDCl$_3$/MeOD, 298K) of **N$_{3q}$S-cyan** recorded at (a) -12 to 210 ppm; (b) 0 to 14 ppm.

Figure S73. ^{1}H NMR (400 MHz, CDCl$_3$, 298K) of **14**.

Figure S74. ^{13}C{^{1}H} NMR (101 MHz, CDCl$_3$, 298K) of **14**.

(a)

11.89
10.97
7.27
7.26
3.40
3.28
2.11
1.18
0.75

N₃qS

200 180 160 140 120 100 80 60 40 20 0

—11.89
—10.97
7.27
7.26
3.40
3.37
3.28
—2.11
—1.18
—0.75

14 13 12 11 10 9 8 7 6 5 4 3 2 1 0
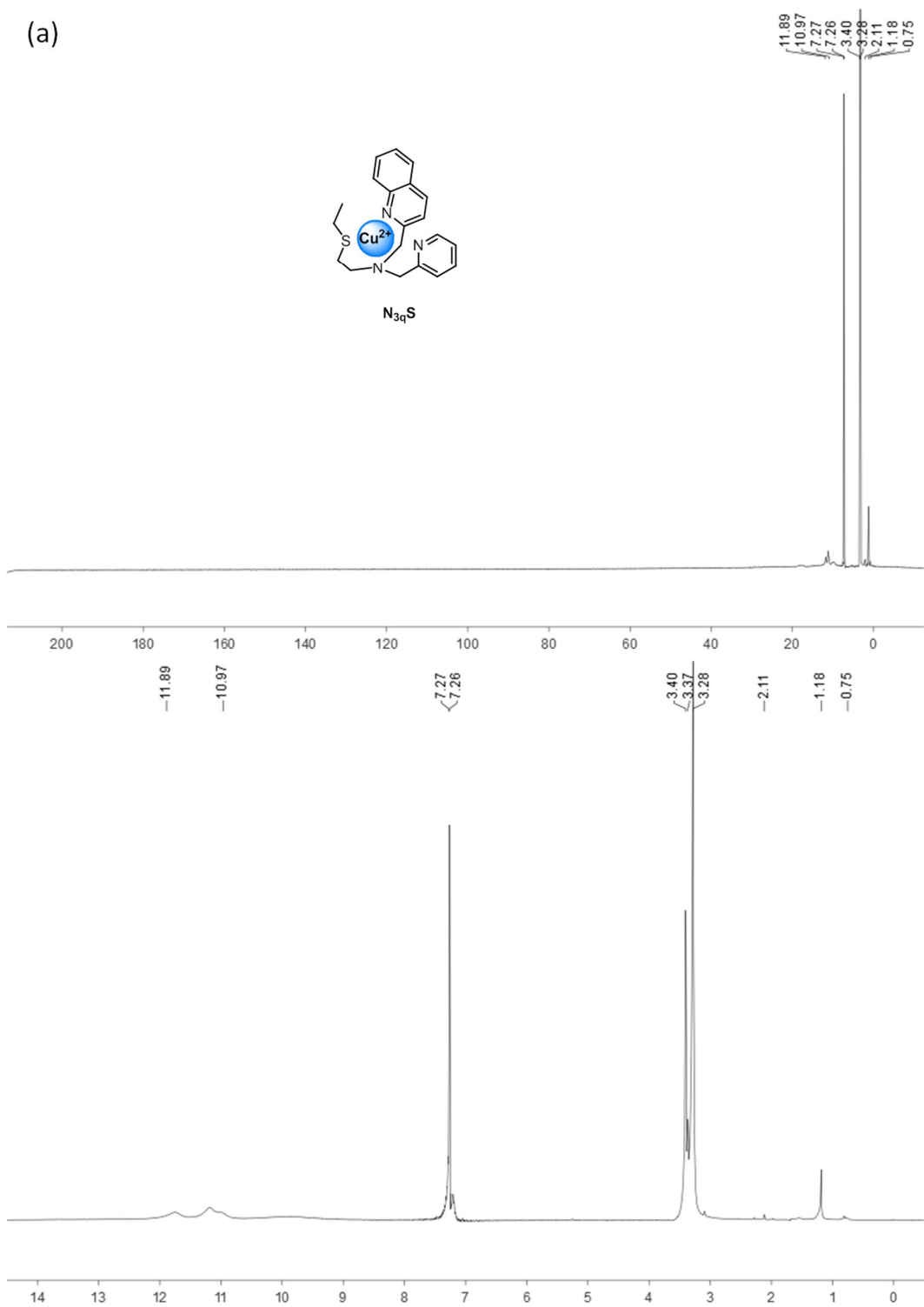
Figure S75. ^{1}H NMR (500 MHz, 9:1 CDCl₃/MeOD, 298K) of **N₃qS** recorded at (a) -12 to 210 ppm; (b) 0 to 14 ppm.
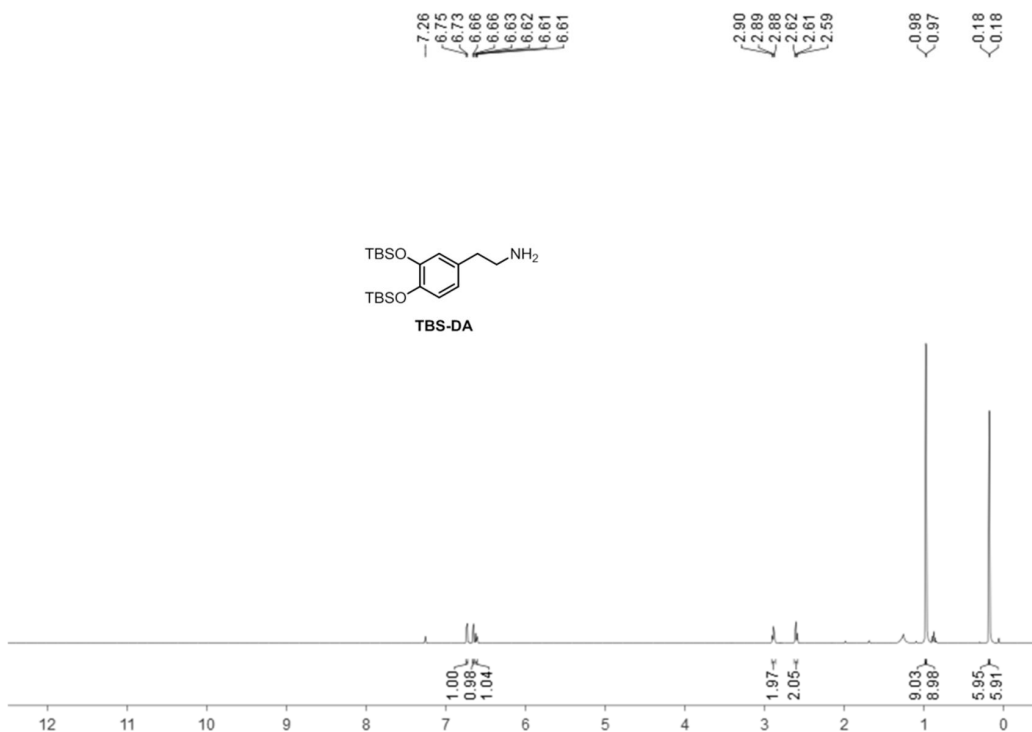
Figure S76. ^{1}H NMR (500 MHz, CDCl$_3$, 298K) of **TBS-DA**.
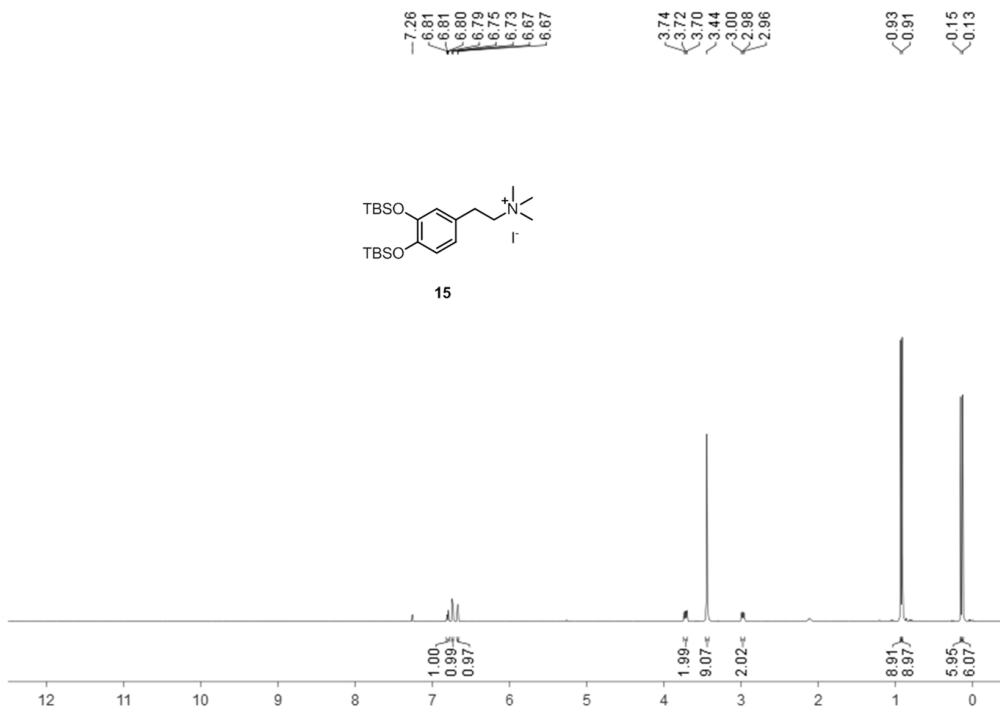
Figure S77. ^{1}H NMR (500 MHz, CDCl$_3$, 298K) of **15**.
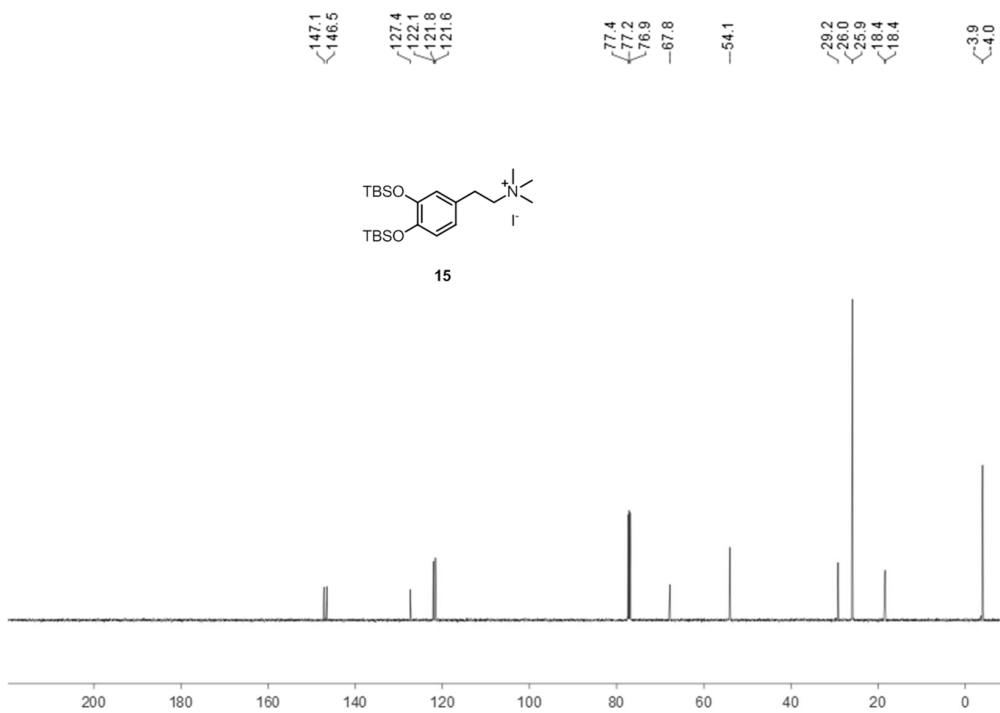
Figure S78. ^{13}C{^{1}H} NMR (126 MHz, CDCl$_3$, 298K) of **15**.
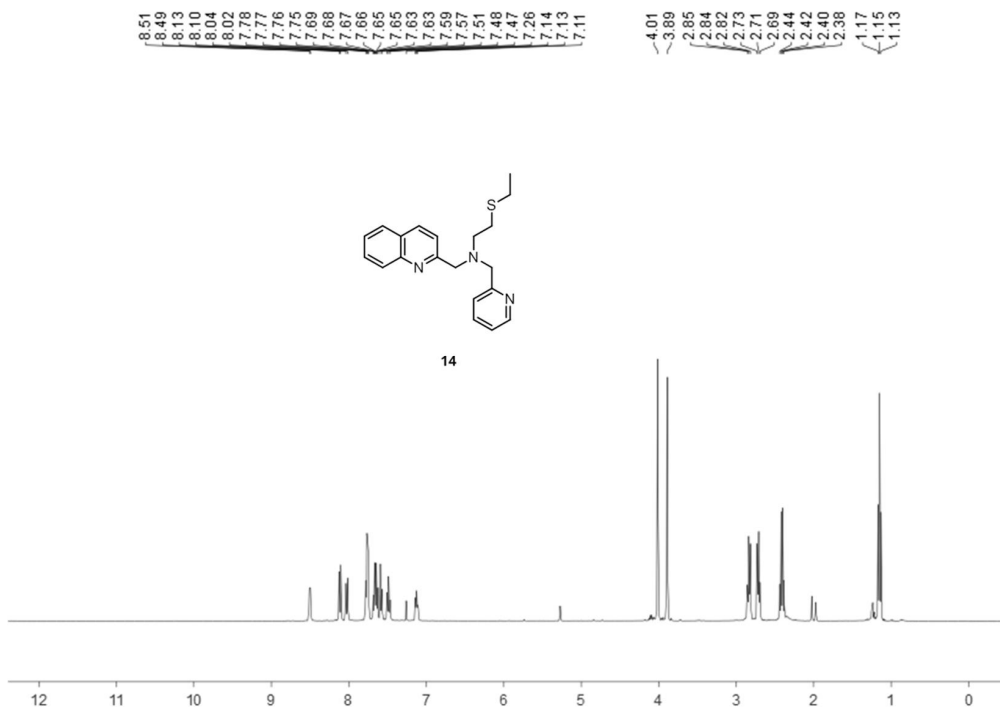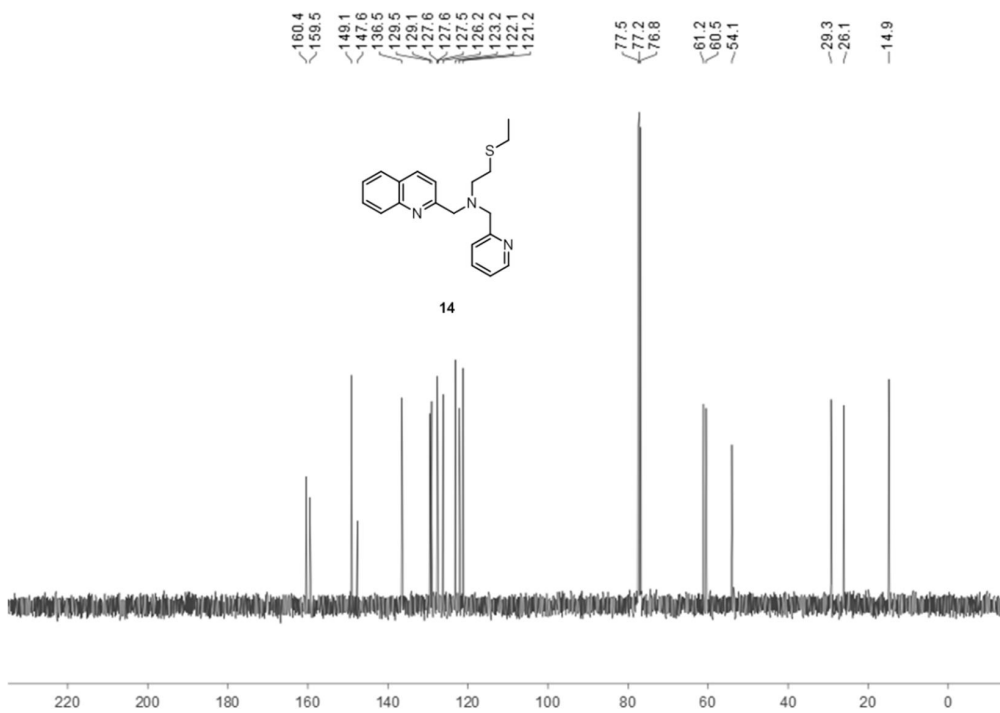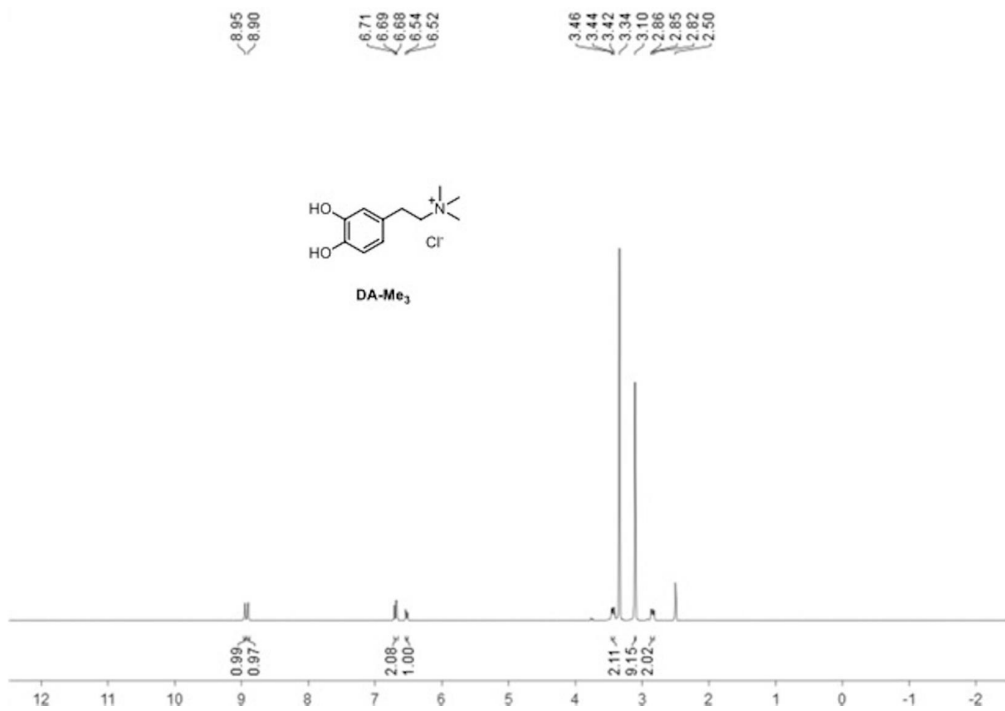
Figure S79. ^{1}H NMR (400 MHz, DMSO-d$_6$, 298K) of **DA-Me$_3$**.
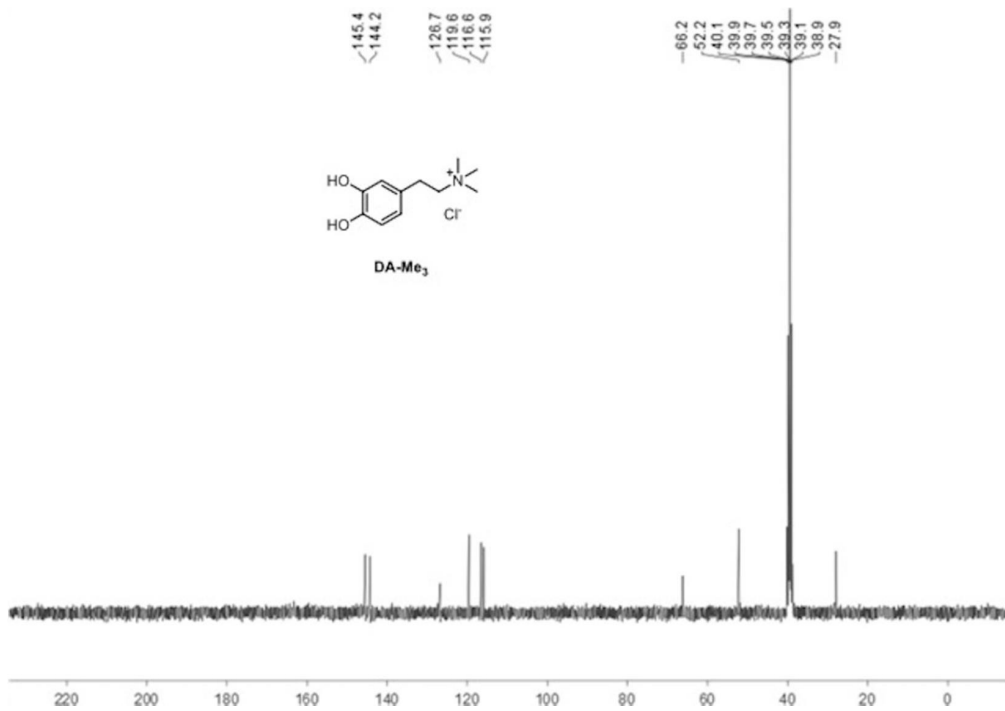
Figure S80. ^{13}C{^{1}H} NMR (101 MHz, DMSO-d$_6$, 298K) of **DA-Me$_3$**.
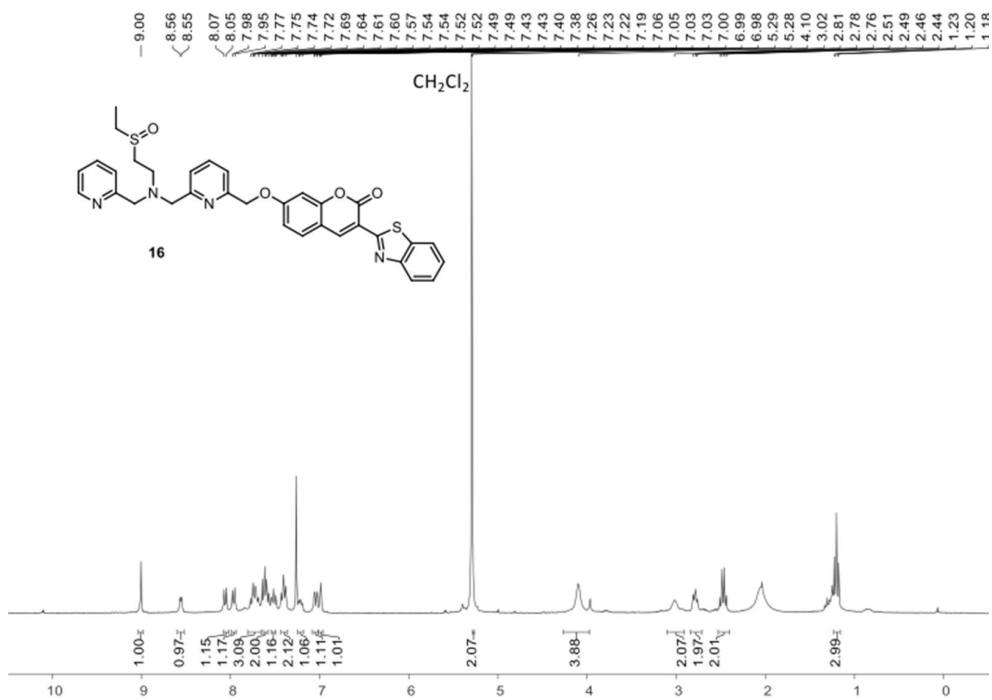
Figure S81. ^{1}H NMR (300 MHz, CDCl$_3$, 298K) and ESI-MS of oxidized product **16**.
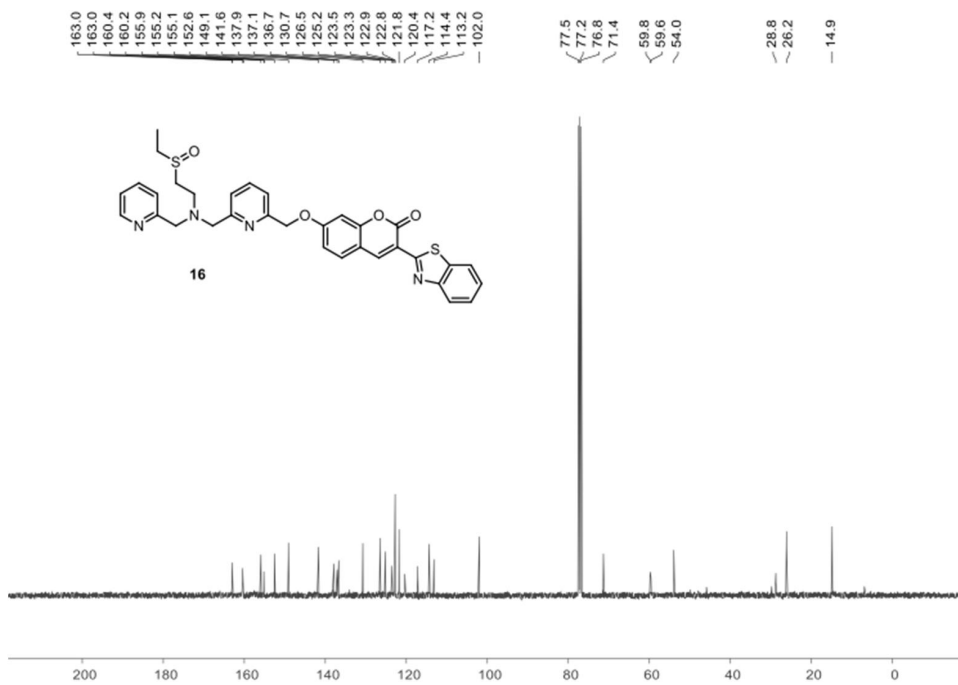
Figure S82. ^{13}C{^{1}H} NMR (101 MHz, CDCl$_3$, 298K) of oxidized product **16**.

X-Ray Diffraction Analysis
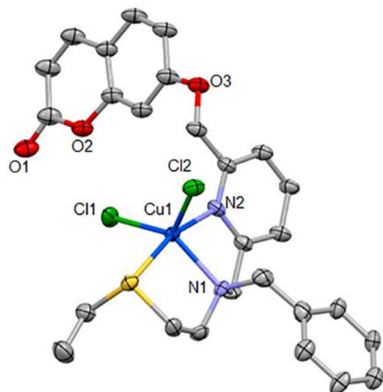
Figure S83. Molecular structure of [Cu^{II}(**2**)Cl]$^{+}$. Thermal ellipsoids represent 50% probability surfaces, solvents, [CuCl$_4$]$^{2-}$ and hydrogen atoms are omitted for clarity. Selected bond length (Å) and angles (°): Cu1–N1 2.016(8), Cu1–N2 2.038(8), Cu1–N3 2.240(8), Cu1–S1 2.340(5), Cu1–Cl1 2.239(5), N1–Cu1–N2 81.1(3), N1–Cu1–N3 112.4(3), N2–Cu1–N3 79.7(3), N1–Cu1–S1 137.5(2), N2–Cu1–S1 88.0(2), N3–Cu1–S1 105.7(2), N2–Cu1–Cl1 173.7(2).
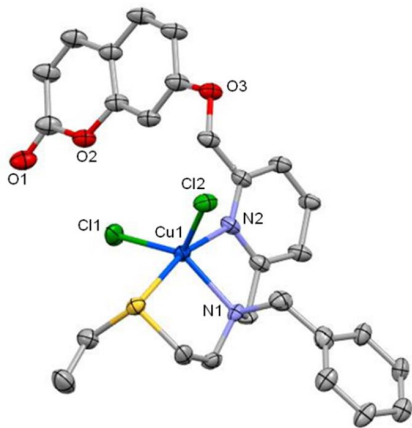
Figure S84. Molecular structure of [Cu^{II}(**9**)Cl$_2$]. Thermal ellipsoids represent 50% probability surfaces, solvents, hydrogen atoms are omitted for clarity. Selected bond length (Å) and angles (°): Cu1–N1 2.131(7), Cu1–N2 2.08(3), Cu1–S1 2.369(2), N1–Cu1–N2 79.5(13), N1–Cu1–S1 93.49(9), N2–Cu1–S1 164.3(12).
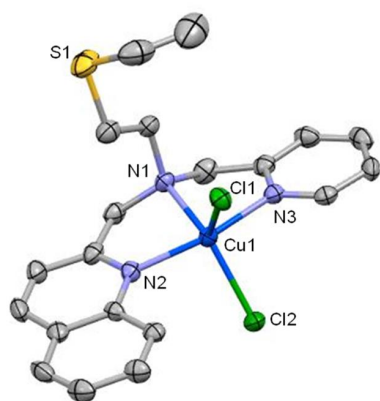
Figure S85. Molecular structure of [Cu^{II}(**14**)Cl$_2$]. Thermal ellipsoids represent 50% probability surfaces, solvents, hydrogen atoms are omitted for clarity. Selected bond length (Å) and angles (°): Cu1–N1 2.113(4), Cu1–N2 2.017(4), Cu1–N3 1.995(4), N1–Cu1–N2 82.43(16), N1–Cu1–N3 80.66(16), N2–Cu1–N3 161.68(16).

Compound	[Cu^{II}(**2**)]$_2$CuCl$_4$·(MeOH)$_2$	
Empirical formula	C$_{54}$H$_{62}$Cl$_6$Cu$_3$N$_6$O$_8$S$_2$	
Formula weight	1390.53	
Temperature / K	173	
Wavelength / Å	0.71073	
Crystal system	Triclinic	
Space group	P- $\bar{1}$	
Unit cell dimension		
a / Å α / degree	13.36(3)	88.34(3)
b / Å β / degree	13.86(3)	84.94(3)
c / Å γ / degree	15.84(3)	87.51(3)
Volume / Å^3	2917(10)	
Z	2	
Density (calcd) / Mgm^{-3}	1.583	
Absorption coeff. / mm^{-1}	1.488	
F(000)	1426.0	
Crystal size / mm^3	0.11 x 0.04 x 0.03	
θ range for data collection	2.4 to 25.0°	
Index ranges	-15<=h<=15	
	-16<=k<=16	
	-18<=l<=18	
Reflection collected	24596	
Independent reflections	10207 [R(int) = 0.120]	
Completeness to θ	θ = 25.208, 97.2 %	
Absorption correction	Multi-scan	
Max. and min. transmission	0.745 and 0.636	
Refinement method	Full-matrix least-squares on F^2	
Data/ restraints / parameters	10207 / 45 / 724	
Goodness-of-fit on F^2	1.07	
Final R indices [$I \geq 2\sigma(I)$]	R1 = 0.094, wR2 = 0.166	
R indices (all data)	R1 = 0.094, wR2 = 0.1395	
Largest diff. peak and hole	1.48 and -1.14 eÅ^{-3}	

Compound	$Cu^{II}(\mathbf{9})Cl_2$
Empirical formula	$C_{27}H_{28}Cl_2CuN_2O_3S$
Formula weight	595.01
Temperature / K	173
Wavelength / Å	0.71073
Crystal system	Triclinic
Space group	P-$\bar{1}$
Unit cell dimension	

a / Å	α / degree	9.163(3)	65.072(9)
b / Å	β / degree	12.719(4)	89.457(8)
c / Å	γ / degree	13.205(5)	73.991(7)

Volume / Å^3	1336.9(8)
Z	2
Density (calcd) / Mgm^{-3}	1.478
Absorption coeff. / mm^{-1}	1.127
F(000)	614.0
Crystal size / mm^3	0.15 x 0.14 x 0.04
θ range for data collection	2.690 to 25.180°
Index ranges	-10<=h<=10
	-15<=k<=13
	-15<=l<=15
Reflection collected	12495
Independent reflections	4692 [R(int) = 0.151]
Completeness to θ	θ = 25.180, 98.0 %
Absorption correction	Multi-scan
Max. and min. transmission	0.595 and 0.745
Refinement method	Full-matrix least-squares on F^2
Data/ restraints / parameters	4692 / 205 / 385
Goodness-of-fit on F^2	1.10
Final R indices [$I \geq 2\sigma(I)$]	R1 = 0.105, wR2 = 0.185
R indices (all data)	R1 = 0.181, wR2 = 207
Largest diff. peak and hole	0.60 and -0.61 eÅ^{-3}

Compound	$Cu^{II}(\textbf{14})Cl_2$	
Empirical formula	$C_{20}H_{23}Cl_2CuN_3S$	
Formula weight	471.91	
Temperature / K	173	
Wavelength / Å	0.71073	
Crystal system	Monoclinic	
Space group	$P2_1/n$	
Unit cell dimension		
a / Å α / degree	9.1168(14)	
b / Å β / degree	13.355(3)	97.889(5)
c / Å γ / degree	16.904(3)	
Volume / Å^3	2038.7(7)	
Z	4	
Density (calcd) / Mgm^{-3}	1.537	
Absorption coeff. / mm^{-1}	1.446	
F(000)	972.0	
Crystal size / mm^3	0.32 x 0.22 x 0.10	
θ range for data collection	2.411 to 26.387°	
Index ranges	-11<=h<=11	
	-16<=k<=14	
	-20<=l<=21	
Reflection collected	17019	
Independent reflections	4143 [R(int) = 0.056]	
Completeness to θ	θ = 26.387, 99.1 %	
Absorption correction	Multi-scan	
Max. and min. transmission	0.609 and 0.745	
Refinement method	Full-matrix least-squares on F^2	
Data/ restraints / parameters	4143 / 19 / 256	
Goodness-of-fit on F^2	1.06	
Final R indices [$I\geq 2\sigma(I)$]	R1 = 0.055, wR2 = 0.113	
R indices (all data)	R1 = 0.105, wR2 = 0.140	
Largest diff. peak and hole	1.06 and -0.96 eÅ^{-3}	

The X-ray intensity data were measured on a *PHOTON100 CMOS* detector system equipped with a compact optics monochromator and a Mo Kα microfocus IμS (λ = 0.71073 Å). The frames were integrated with the Bruker SAINT software package using a narrow-frame algorithm. The structure was solved and refined using the Bruker SHELXTL Software Package.

References

1. S. T. Prigge, B. A. Eipper, R. E. Mains and L. M. Amzel, *Science* 2004, **304**, 864.
2. J. Jia, L. Wu, Y. Ding, C. Huang, W. Zhu, Y. Xu and X. Qian, *Dalton Trans.* 2016, **45**,9402.
3. W. Lin, L. Long and W. Tan, *Chem. Commun.* 2010, **46**, 1503.
4. J. I. MacDonald, H. K. Munch, T. Moore and M. B. Francis, *Nat. Chem. Biol.* 2015, **11**, 326.
5. S. S. More and R. Vince, *J. Med. Chem.* 2008, **51**, 4581.
6. E. W. Miller, O. Tulyathan, E. Y. Isacoff and C. J. Chang, *Nat. Chem. Biol.* 2007, **3**, 263.
7. Z. H. Yu, C. Y. S. Chung, F. K. Tang, T. F. Brewer and H. Y. Au-Yeung, *Chem. Commun.* 2017, **53**, 10042.
8. A. Rukavishnikov, K. R. Gee, I. Johnson and S. Corry, *Anal. Biochem.* 2011, **419**, 9.
9. J. Wang and G. Evano, *Org. Lett.* 2016, **18**, 3542.
10. J. T. Moon, S. H. Ha, S. H. Lee, T. H. Kwon, C. R. Oh, Y. D. Kim, J. Kim, D. J. Choo and J. Y. Lee, *Bioorg. Med. Chem. Lett.* 2010, **20**, 52.
11. X. F. Yang and X. Q. Guo, *Analyst* 2001, **126**, 928.
12. A. Greer, *Acc. Chem. Res.* 2006, **39**, 797.
13. K. M. Robinson and J. S. Beckman, *Methods Enzymol.* 2005, **396**, 207.
14. T. A. Little, *Biopharm. Int.* 2015, **28**, 49.
15. I. Zablocka, M. Wysocka-Zolopa, and K. Winkler, *Int. J. Mol. Sci.* 2019, **20**, 111.
16. J. Zhao, Y. Y. Cheng, W. Fan, C. B. Yang, S. F. Ye, W. Cui, W. Wei, L. X. Lao, J. Cai, Y. F. Han and J. Rong, *CNS Neurosci. Ther.* 2015, **21**, 61.
17. Y. Cheng, C. Yang, J. Zhao, H. F. Tse, J. Rong, *J. Nutr. Biochem.* 2015, **26**, 146.
18. J. Zhao, Y. Cheng, C. Yang, S. Lau, L. Lao, B. Shuai, J. Cai and J. Rong, *Mol. Neurobiol.* 2016, **53**, 2200.
19. V. Najmanová, L. Rambousek, K. Syslová, V. Bubeníková, R. Šlamberová, K. Valeš, and P. Kačer, P. *Chromatographia* 2011, **73**, 143.