

Supplementary Information

Design, synthesis and comparison of water-soluble phthalocyanine/ porphyrin analogues on inhibition of A β ₄₂ fibrillization

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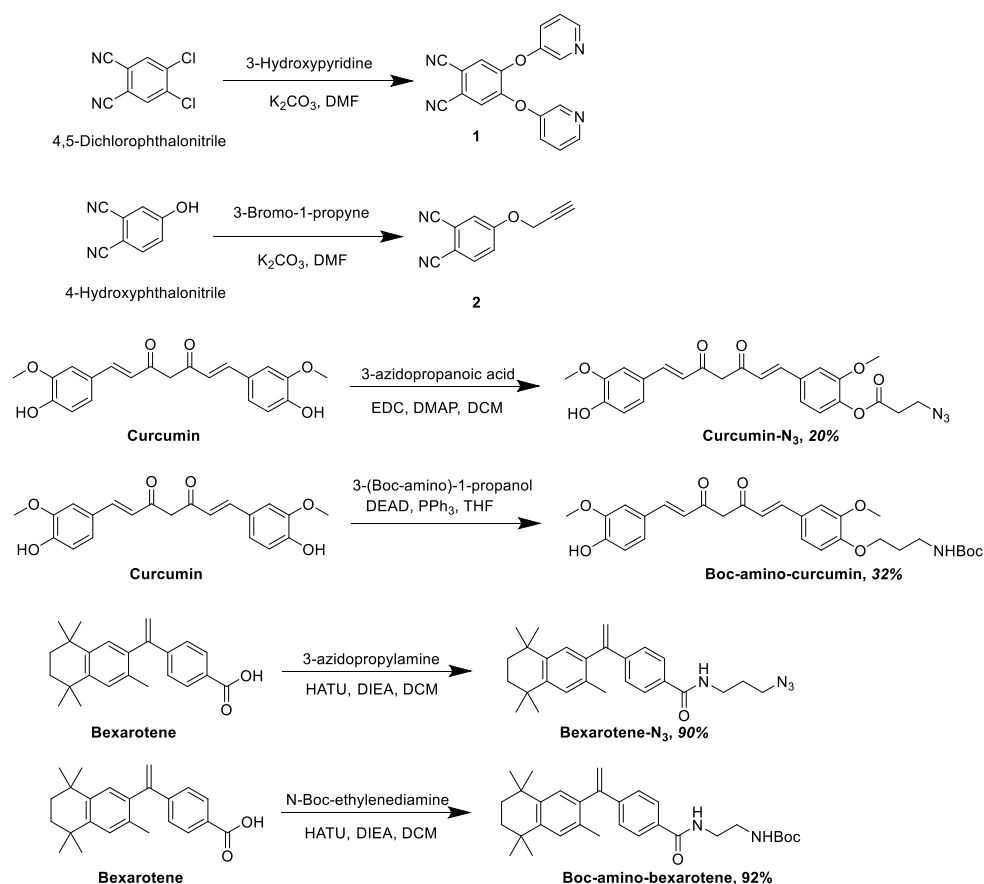
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Materials

All solvents and reagents were obtained from commercial suppliers and used without further purification unless otherwise stated. Curcumin (95%), 3-bromopropionic acid (98%), 3-bromopropylamine hydrobromide (98%), EDCI (99%), HATU (98%), DIEA (99%), 6-chloropyridine-3-carboxaldehyde (98%), 1-Boc-piperazine (98%), 4-nitrophthalonitrile (99%), 3-bromo-1-propyne (99%), 1,8-diazabicyclo(5.4.0)undec-7-ene (DBU, 98%), 3-hydroxypyridine (99%), iodomethane (98%), zinc acetate anhydrous (99%) and n-pentanol (98%) were purchased from Dieckmann. Diethylamine (99%), sodium azide (99%), 2,4-pentanedione (99%), trifluoroacetic acid (TFA, 99%), boron trifluoride ethyl etherate (46%) were provided by Sigma-Aldrich. 1,2,3,4-Tetrahydroisoquinoline (95%) was bought from Tokyo Chemical Industry. Bexarotene (99%) was purchased from Aladdin Chemical.

Synthetic procedures



Scheme S1. Synthetic routes of phthalonitrile **1**, phthalonitrile **2**, Curcumin-N₃, Boc-amino-

curcumin, Bexarotene-N₃ and Boc-amino-bexarotne.

Phthalonitrile 1

4, 5-Dichlorophthalonitrile (2 g, 10 mmol), 3-hydroxypyridine (3.86 g, 40 mmol) were dissolved in 32 mL DMF and heated to 90 °C. Potassium carbonate (8.41 g, 60 mmol) was added in 5 times within 2 hours, and the mixture was heated overnight. The product was purified by flash chromatography (Al₂O₃, CH₂Cl₂/MeOH = 30/1 as eluent) after solvent was evaporated under reduced pressure. At last, 2.33 g product **1** was given with a yield of 73%. ¹H NMR (400 MHz, DMSO-d₆) δ (ppm): 7.46 (2H, dd, J = 8.48 Hz, J = 4.76 Hz), 7.60 (2H, ddd, J = 8.42 Hz, J = 2.98 Hz, J = 1.40 Hz), 8.05 (2H, s), 8.44 (4H, m). ¹³C NMR (100 MHz, DMSO-d₆): δ (ppm): 112.19, 115.67, 125.35, 126.30, 126.36, 140.99, 146.39, 150.79, 152.23. ESI-MS (m/z): [M+Na]⁺ C₁₈H₁₀N₄NaO₂ 337.07; found 337.14.

Phthalonitrile 2

4-Hydroxyphthalonitrile (0.5 g, 3.5 mmol) was mixed with propargyl bromide (0.62 mg, 5.2 mmol) and potassium carbonate (0.96 g, 7 mmol) in 5 mL DMF, and then heated to around 80 °C for 3 h. After the reaction was completed, the mixture was poured into 100 mL water and extracted with ethyl acetate (50 mL × 3). The organic layers were collected and dried with anhydrous sodium sulfate, which was then evaporated under vacuum and dissolved in methanol (10 mL) for further recrystallization with water. The resulted solid was filtered and washed with water, and then dried under vacuum, finally given 0.56 g product **2** (yield at 89%). ¹H NMR (400 MHz, DMSO-d₆) δ (ppm): 3.73 (1H, t, J = 2.44 Hz), 5.03 (2H, d, J = 2.40 Hz), 7.52 (1H, dd, J = 2.72 Hz, J = 8.82 Hz), 7.82 (1H, d, J = 2.64 Hz), 8.11 (1H, d, J = 8.80 Hz). ¹³C NMR (100 MHz, DMSO-d₆) δ (ppm): 57.16, 78.20, 80.10, 107.23, 116.12, 116.55, 116.67, 120.95, 121.12, 136.28, 160.92. ESI-MS (m/z): [M+Na]⁺ calcd. for C₁₁H₆N₂NaO 205.04; found 204.96.

Curcumin-N₃

3-Azidopropionic acid was prepared according to the reported procedures.² 3-Bromopropionic acid (1.53 g, 10 mmol) and sodium azide (1.30 g, 20 mmol) were dissolved in water (15 mL). After constant stir for 24 h, the aqueous solution was adjusted to pH 1 with hydrochloric acid and then extracted with diethyl ether (30 mL × 3). After that, organic layer was washed with 30 mL saturated brine and dried with anhydrous sodium sulfate, which was followed by evaporation under vacuum. ¹H NMR (400 MHz, D₂O) δ (ppm): 2.64 (2H, t, J = 6.4 Hz), 3.59 (2H, t, J = 6.4 Hz).

Curcumin (500 mg, 1.36 mmol) was mixed with 3-azidopropionic acid (156 mg, 1.36 mmol) and then dissolved in 20 mL DCM, and afterwards EDC (264 μL, 1.50 mmol) and DMAP (42 mg, 0.34 mmol) were added and stirred for further 24 hours. The product was harvested by separation using a silica column with EA/DCM/n-hexane = 1/1/2 as eluent, finally giving 128 mg product with yield at 20%. ¹H NMR (400 MHz, CDCl₃) δ (ppm): 2.88 (2H, t, J = 6.56 Hz), 3.71 (2H, t, J = 6.52 Hz), 3.88 (3H, s), 3.95 (3H, s), 5.83 (1H, s), 5.88 (1H, s), 6.49 (1H, d, J = 15.76 Hz), 6.55 (1H, d, J = 15.76 Hz), 6.94 (1H, d, J = 8.2 Hz), 7.12 (5H, m), 7.59 (1H, d, J = 3.96 Hz), 7.63 (1H, d, J = 3.92 Hz). ¹³C NMR (100 MHz, CDCl₃) δ (ppm): 33.83, 46.72, 55.98, 101.50, 109.69, 111.46, 114.86, 120.92, 121.80, 123.03, 123.12, 124.43, 127.57, 134.35, 139.25, 140.86, 141.42, 146.81, 148.01, 151.25, 168.77, 181.71, 184.54. ESI-MS (*m/z*): [M+H]⁺ calcd. for C₂₄H₂₄N₃O₇ 466.15; found 466.00.

Boc-amino-curcumin

Curcumin (0.5 g, 1.36 mmol) was mixed with triphenylphosphine (0.43 g, 1.63 mmol) and 3-(Boc-amino)-1-propanol (0.24 g, 1.36 mmol) in 15 mL dry THF at 0 °C, and then DEAD (0.25 g, 1.43 mmol) was dissolved in 5 mL dry THF and added into the mixture dropwise. Afterwards, the reaction was recovered to room temperature and further stirred overnight. After evaporating the solvent, the residue was loaded onto a silica

column with DCM/EA = 10/1 as eluent to give the final product (220 mg, 32%). ¹H NMR (400 MHz, CDCl₃) δ (ppm): 1.48 (9H, s), 2.04 (2H, t, J= 5.96 Hz), 3.38 (2H, d, J= 6.08 Hz), 3.93 (6H, s), 4.13 (2H, t, J= 5.84 Hz), 5.54 (1H, s), 5.82 (1H, s), 6.21 (1H, br), 6.50 (2H, s), 6.85 (1H, d, J= 7.00 Hz), 6.93 (1H, d, J= 7.52 Hz), 7.07 (4H, m), 7.60 (2H, d, J= 14.40 Hz). ¹³C NMR (100 MHz, CDCl₃) δ (ppm): 28.50, 29.23, 29.70, 38.93, 55.80, 55.95, 68.16, 79.04, 101.26, 109.76, 110.01, 112.40, 114.94, 121.72, 122.13, 122.45, 122.89, 127.61, 128.37, 140.31, 140.68, 146.92, 148.01, 149.51, 150.17, 156.19, 183.04, 183.50. ESI-MS (m/z): [M+Na]⁺ calcd. for C₂₉H₃₅NO₈Na 548.23; found 548.29.

Bexarotene-N₃

3-Azido-1-propanamine was prepared according to the reported steps.³ Simply, 3-bromopropan-1-amine was mixed with sodium azide in 1/3 molar ratio and then dissolved in water. After stirring for 3 days, the solution was adjusted with NaOH solution until pH was higher than 10, and then the product was extracted with DCM for 3 times. The organic phases were combined and washed with saline before evaporation under vacuum. The finally given product was used for next step without any more purification. ¹H NMR (400 MHz, D₂O) δ (ppm): 1.97 (2H, m), 3.12 (2H, t, J = 7.52 Hz), 3.53 (2H, t, J = 6.44 Hz).

Bexarotene is commercially available, and 50 mg of this product (0.14 mmol) was dissolved in 5 mL DCM, and 3-azido-1-propanamine (30 mg, 0.22 mmol), HATU (110 mg, 0.28 mmol) and DIEA (60 μL, 0.35 mmol) were added subsequently. After further stirring for 3 hours, the reaction mixture was concentrated and then loaded onto a silica column and purified with EA/PE (1/4), finally acquiring 56 mg product (yield at 90%). ¹H NMR (400 MHz, CDCl₃) δ (ppm): 1.27 (6H, s), 1.30 (6H, s), 1.70 (4H, s), 1.92 (2H, q, J= 6.56 Hz), 1.94 (3H, s), 3.45 (2H, t, J= 6.52 Hz), 3.56 (2H, q, J= 6.32 Hz), 5.30 (1H, d, J= 1.36 Hz), 5.78 (1H, d, J= 1.36 Hz), 6.31 (1H, s), 7.07 (1H, s), 7.12 (1H, s),

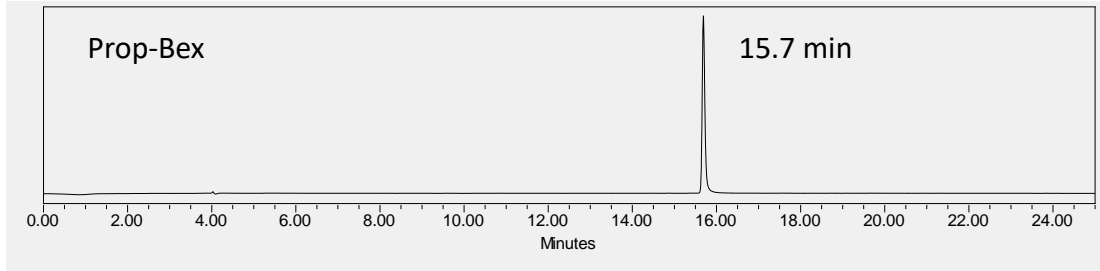
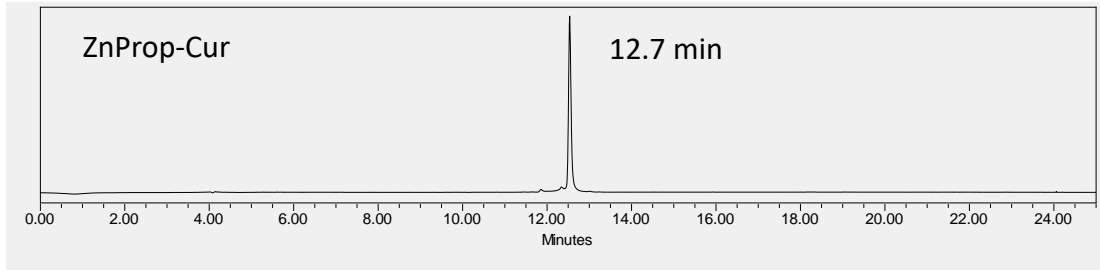
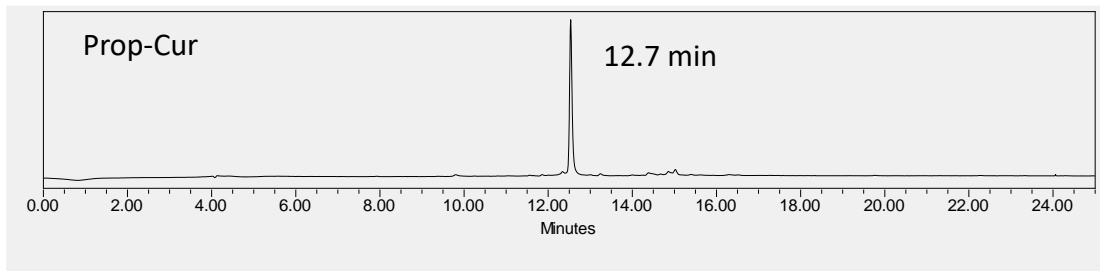
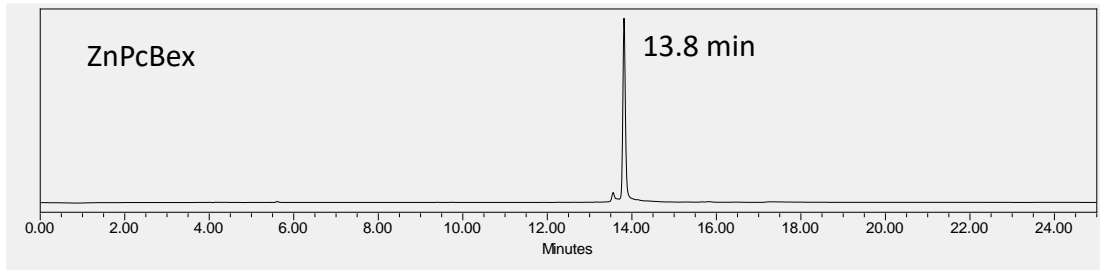
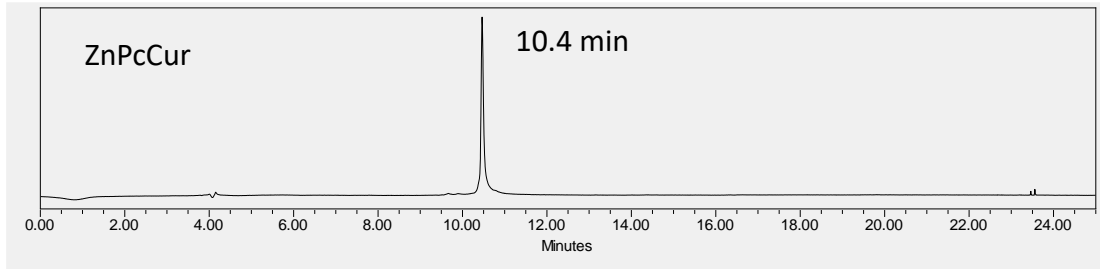
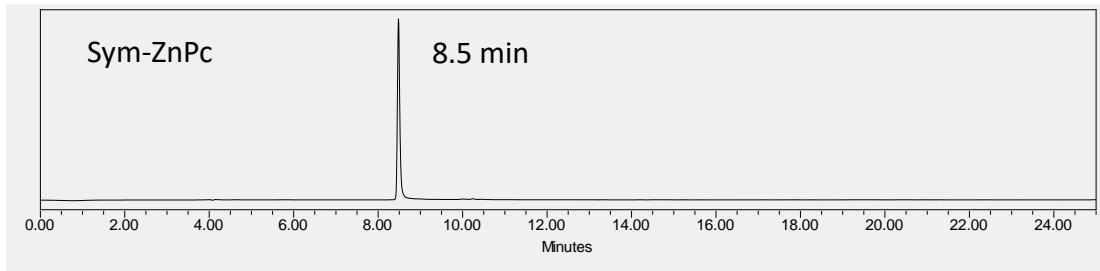
7.34 (2H, d, J= 8.44 Hz), 7.68 (2H, d, J= 8.44 Hz). ¹³C NMR (100 MHz, CDCl₃) δ (ppm): 19.92, 28.79, 31.88, 31.93, 33.89, 33.99, 35.20, 37.84, 49.68, 116.37, 126.82, 126.88, 128.01, 128.04, 132.70, 133.08, 138.04, 142.30, 144.32, 149.04. ESI- MS (*m/z*): [M+H]⁺ calcd. for C₂₇H₃₅N₄O 431.27; found 431.40.

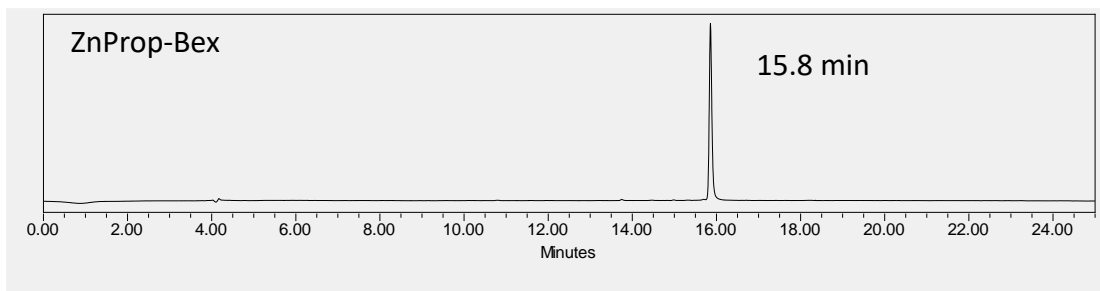
Boc-amino-bexarotene

Bexarotene (50 mg, 0.14 mmol), N-Boc-ethylenediamine (23 mg, 0.14 mmol) and HATU (110 mg, 0.28 mmol) were mixed in 10 mL DCM and DIEA (60 μL, 0.35 mmol) was added subsequently, which was followed by constant stir for 2 hours at room temperature. The mixture was purified using a silica column (DCM/EA = 20/1) to give 63 mg product (92% yield). ¹H NMR (400 MHz, CDCl₃) δ (ppm): 1.29 (6H, s), 1.32 (6H, s), 1.43 (9H, s), 1.72 (4H, s), 1.95 (3H, s), 3.41 (2H, d, J = 5.72 Hz), 3.57 (2H, q, J = 5.44 Hz), 5.30 (1H, d, J = 1.40 Hz), 5.79 (1H, d, J = 1.44 Hz), 7.09 (1H, s), 7.14 (1H, s), 7.34 (2H, d, J = 8.36 Hz), 7.76 (2H, d, J = 8.12 Hz). ¹³C NMR (400 MHz, CDCl₃) δ (ppm): 19.92, 28.34, 31.89, 31.93, 33.89, 33.99, 35.21, 35.23, 38.61, 40.04, 41.94, 79.91, 116.24, 126.71, 127.06, 128.01, 128.06, 132.74, 132.92, 138.13, 142.26, 144.27, 149.14, 157.44. ESI-MS (*m/z*): [M+H]⁺ calcd. for C₃₁H₄₃N₂O₃ 491.32; found 491.35.

HPLC analysis of ZnPc/prophyrin analogues

Liquid chromatography analysis was performed using a Waters 1525 HPLC Binary Gradient Module System with UV detection at 350 nm. Water containing 0.05 % trifluoroacetic acid and pure acetonitrile were adopted as mobile phase A and B respectively. The instrument method started with 90 % A / 10 % B with a flow rate of 1 mL/min, and the portion of B was increased gradually to 100 % over 20 min, which was followed with equilibrium with 90 % A / 10 % B for 5 min before next injection.





ICP-OES analysis

The final products of ZnPc conjugates and metallic porphyrin analogues were examined using Agilent 710 inductive coupled plasma optical emission spectrometer (ICP-OES) to give the exact contents of metals (zinc or copper) for determining the true contents for each compound. Generally, 1 mg of each compound was weighed out using a sartorius balance and prepared to be 100 mL aqueous solution containing 2 % nitric acid. The zinc and copper references were prepared from Zn(OAc)₂ and CuCl₂, respectively, as 2 % nitric acid solutions of concentration gradients from 100, 200, 300, 500, to 1000 ppb. The absorptions at 324.75 nm and 213.85 nm were adopted for analyzing levels of copper and zinc respectively. Then the metal contents of each compound were given referring to the standard curve. According to the recorded zinc or copper levels of various compound solutions, the true contents (W) of the conjugates can be calculated through the following equation,

$$W(\%) = \frac{C_m V m.w.}{W} \times 100\%,$$

C_m is the molar level of zinc or copper of tested compound measured by ICP-OES; V is the volume of tested aqueous solution; $m.w.$ indicates the molecular weight of the specific compound; W is the mass of the compound weighed out by a balance.

Tables and figures

Table 1. Determination of contents of compounds.

	Amount (mg)	M.W.	Theoretical level (ppb)	True level (ppb)	Content (%)
sym-ZnPc	1.0	2346.87	278.59	225.38	80.9
ZnPcCur	1.0	2424.14	269.70	205.00	76.0
ZnPcBex	1.0	2389.27	273.64	178.69	65.3
sym-ZnPorp	1.0	1192.24	536.80	475.61	88.6
ZnProp-Cur	1.0	1572.90	415.67	399.00	96.0
ZnProp-Bex	1.0	1537.50	416.26	268.07	64.4

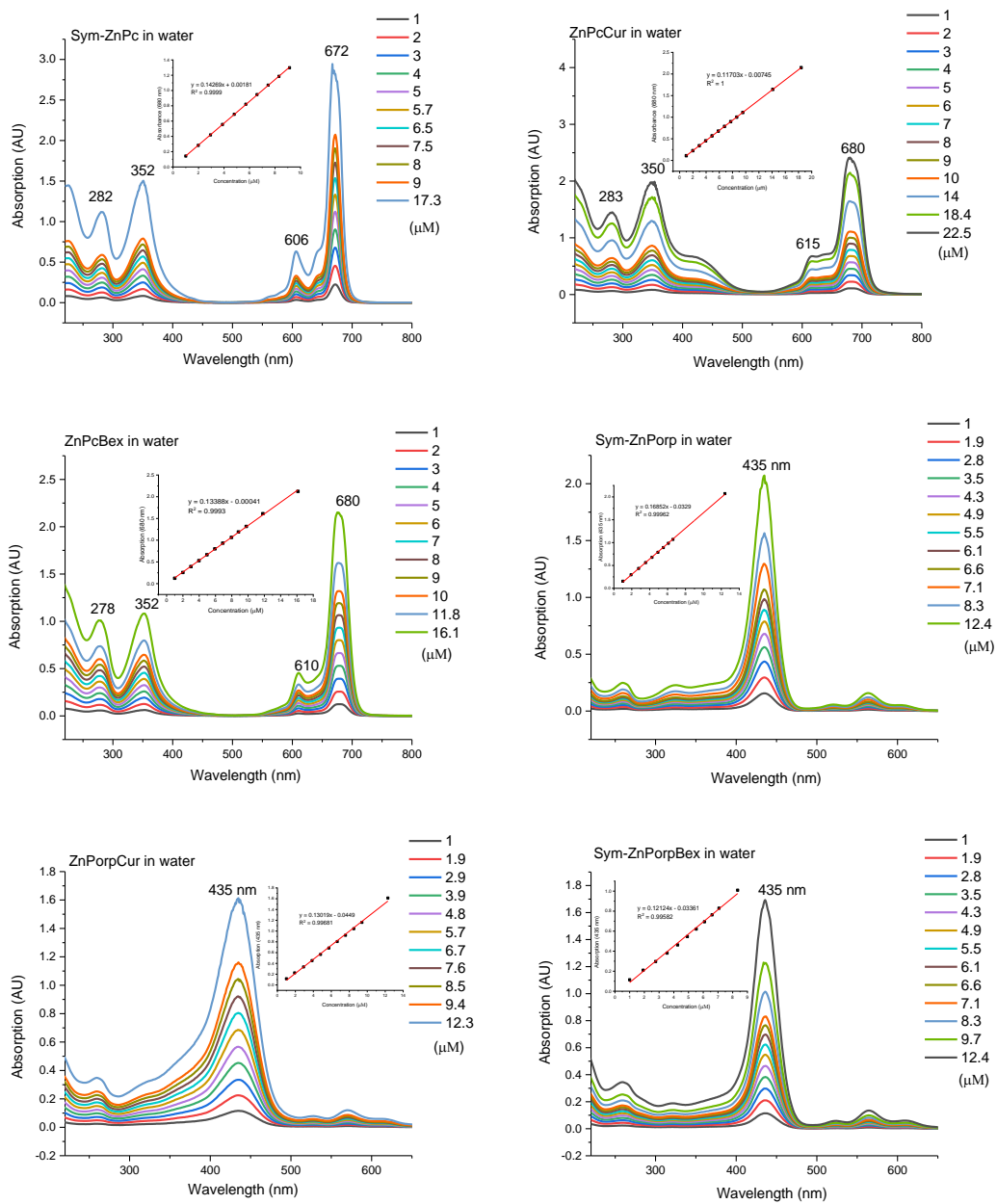


Figure S1. UV-vis absorption spectra of ZnPc and ZnPc compounds in water.

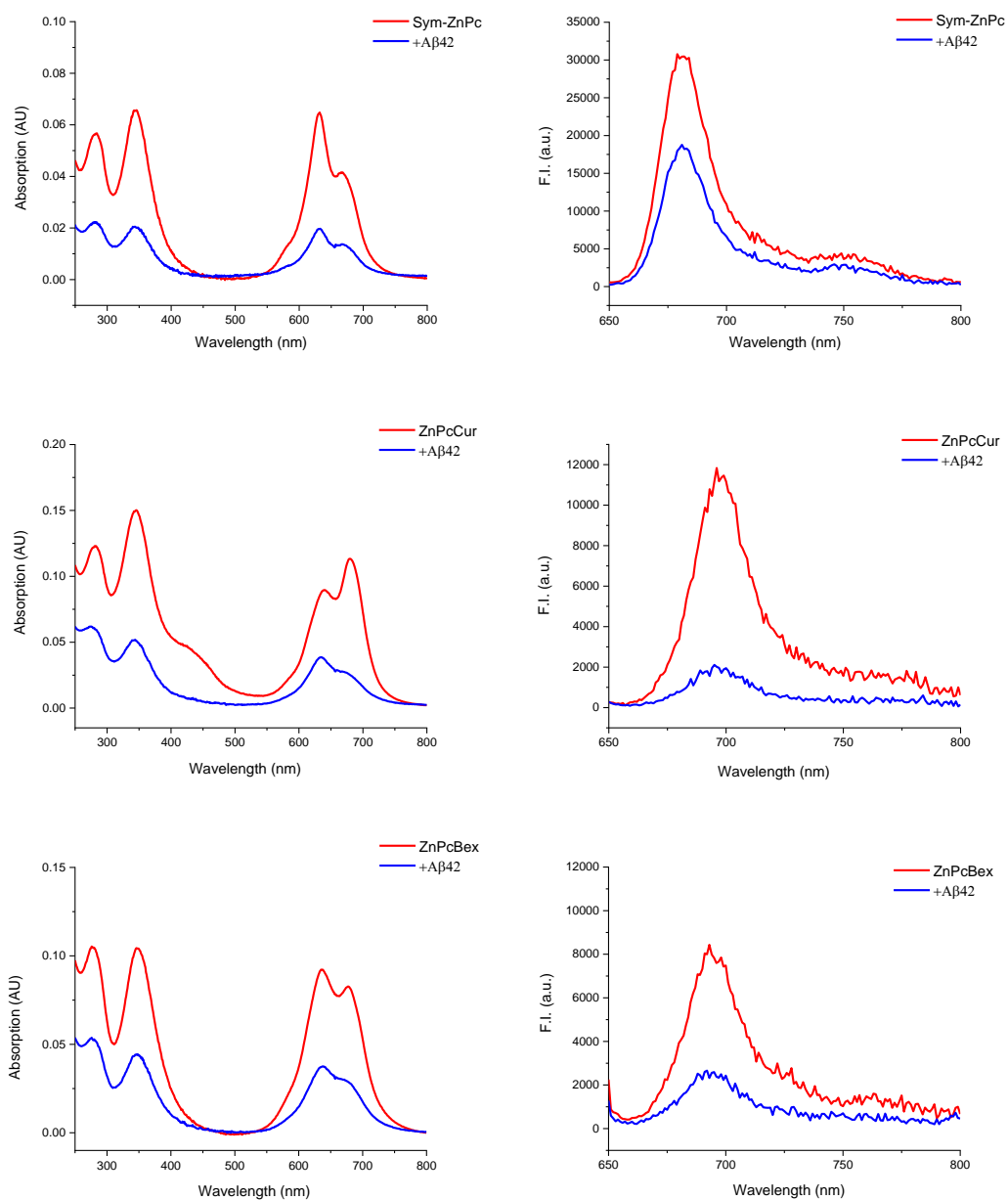


Figure S2. UV-vis absorption and fluorescence emission spectra of ZnPc analogues before and after incubation with A β ₄₂ in buffer (20 mM sodium phosphate, 272 mM NaCl, pH 7.4). 2 μ M of various compounds were incubated with 5 μ M of A β ₄₂ for 24 hours at 37 ° C. The excitation wavelength was fixed at 635 nm.

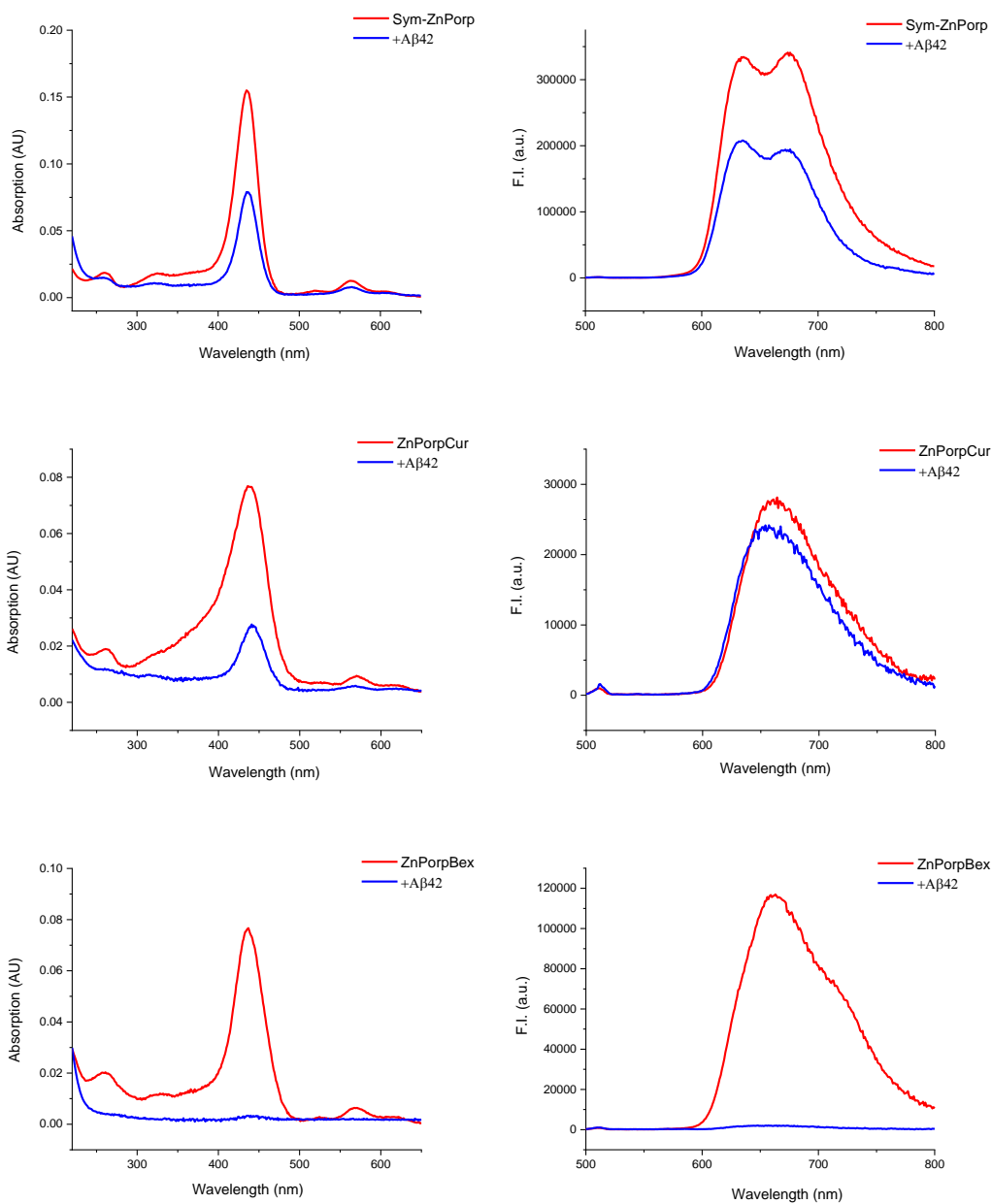


Figure S3. Uv-vis absorption and fluorescence emission spectra of ZnPorp analogues before and after incubation with A β ₄₂ in buffer (20 mM sodium phosphate, 272 mM NaCl, pH 7.4). 2 μ M of various compounds were incubated with 5 μ M of A β ₄₂ for 24 hours at 37 ° C. The excitation wavelength was fixed at 435 nm.

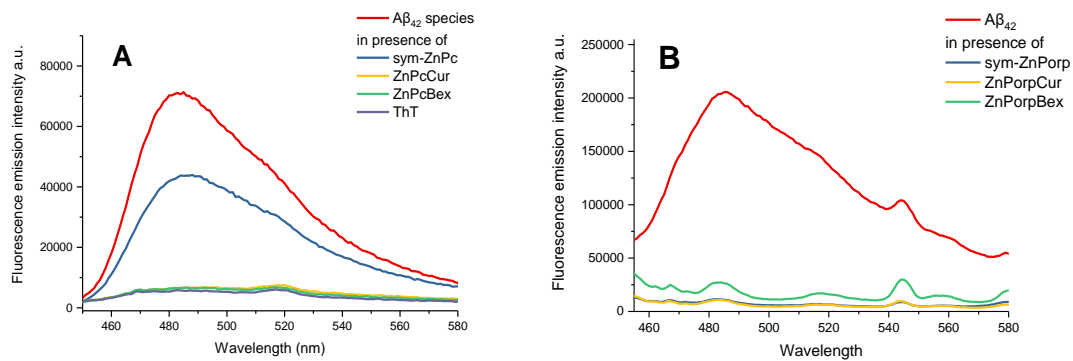


Figure S4. Incubation results of Aβ₄₂ peptide (10 μM) with equal equivalent of ZnPc or ZnPorp analogues respectively. A) ThT emission spectra of Aβ₄₂ species after 24 hours of incubation in presence of ZnPc analogues. B) ThT emission spectra of Aβ₄₂ species after 24 hours of incubation in presence of ZnPorp analogues.

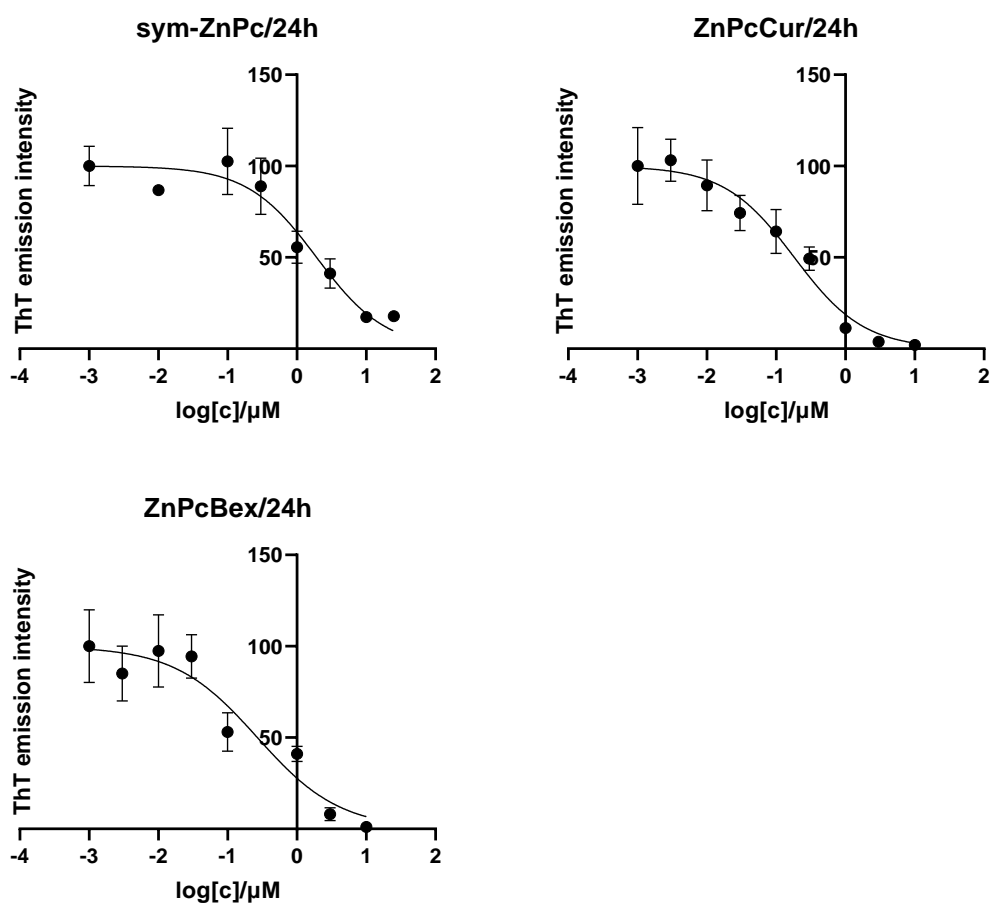


Figure S5. Evaluation of inhibition on Aβ₄₂ fibrillization in presence of sym-ZnPc, ZnPcCur, and ZnPcBex respectively by using ThT assay. After 24 hours of incubation, fluorescence of ThT was measured using a BMG LABTECH ClarioStar microplate reader using excitation and emission

wavelengths of 440 nm and 485 nm respectively. The acquired data were analyzed using GraphPad Prism to obtain IC_{50} values using $\log(\text{concentration})$ vs. normalized response-variable slope.

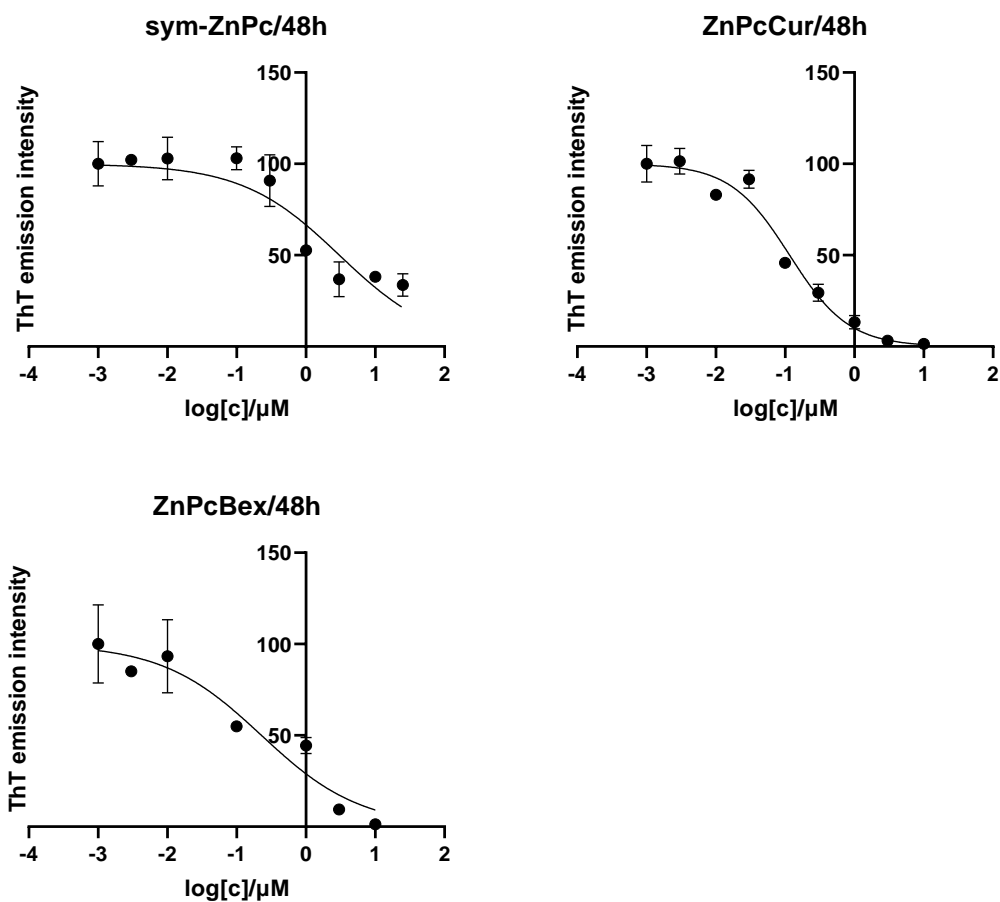


Figure S6. Inhibition study results of $A\beta_{42}$ in presence of various concentrations of ZnPc compounds after 48 hours of incubation. The extents of fibrillization were examined using ThT fluorescence, and the data were acquired and treated using methods as described in Figure S4.

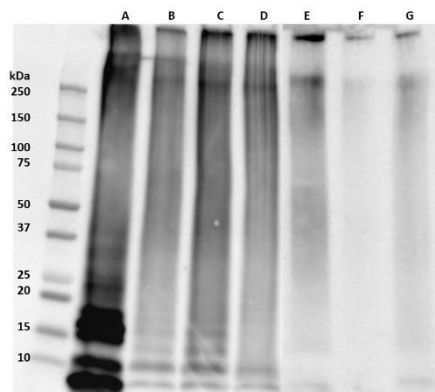


Figure S7. Western blot results of 10 μM of $\text{A}\beta_{42}$ monomer incubated with 10 μM of various compounds in fibril growing buffer (20 mM sodium phosphate, 272 mM NaCl, pH 7.4) after 24 hours of incubation. Each lane represents the following items, A) $\text{A}\beta_{42}$ only, B) sym-ZnPc, C) ZnPcCur, D) ZnPcBex, E) sym-ZnPorp, F) ZnPorpCur, G) ZnPorpBex.

NMR and ESI-MS spectra

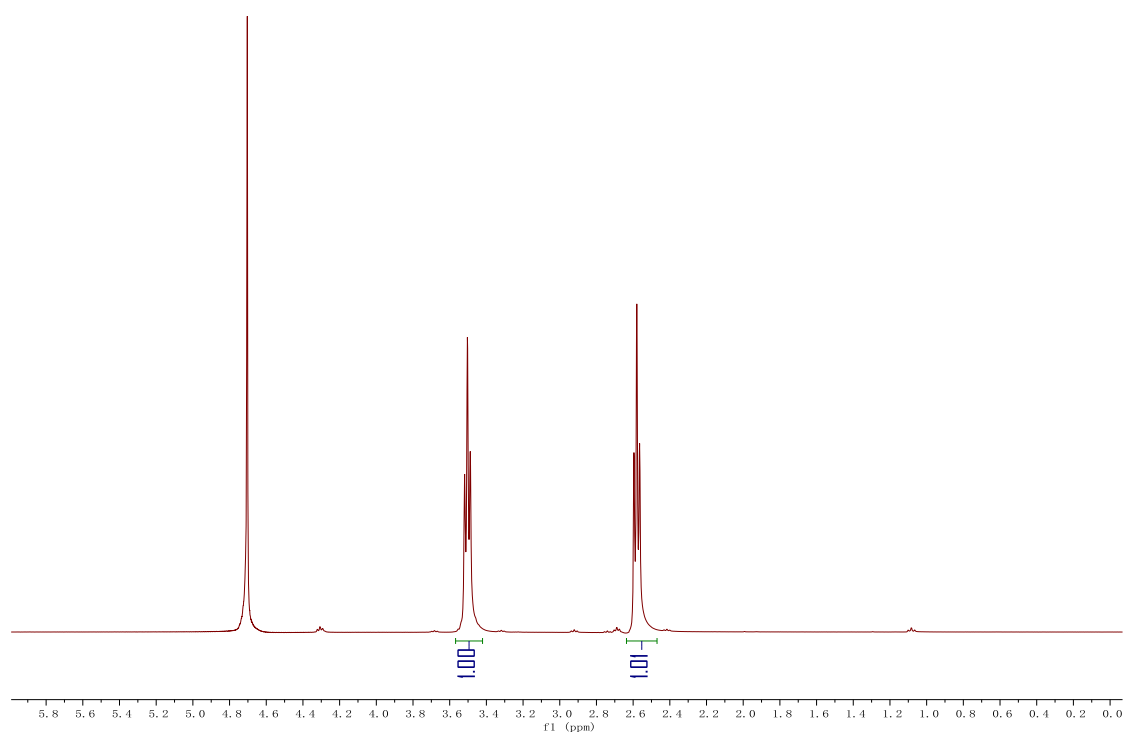


Figure S6. ^1H NMR of 3-azidopropanoic acid in D_2O .

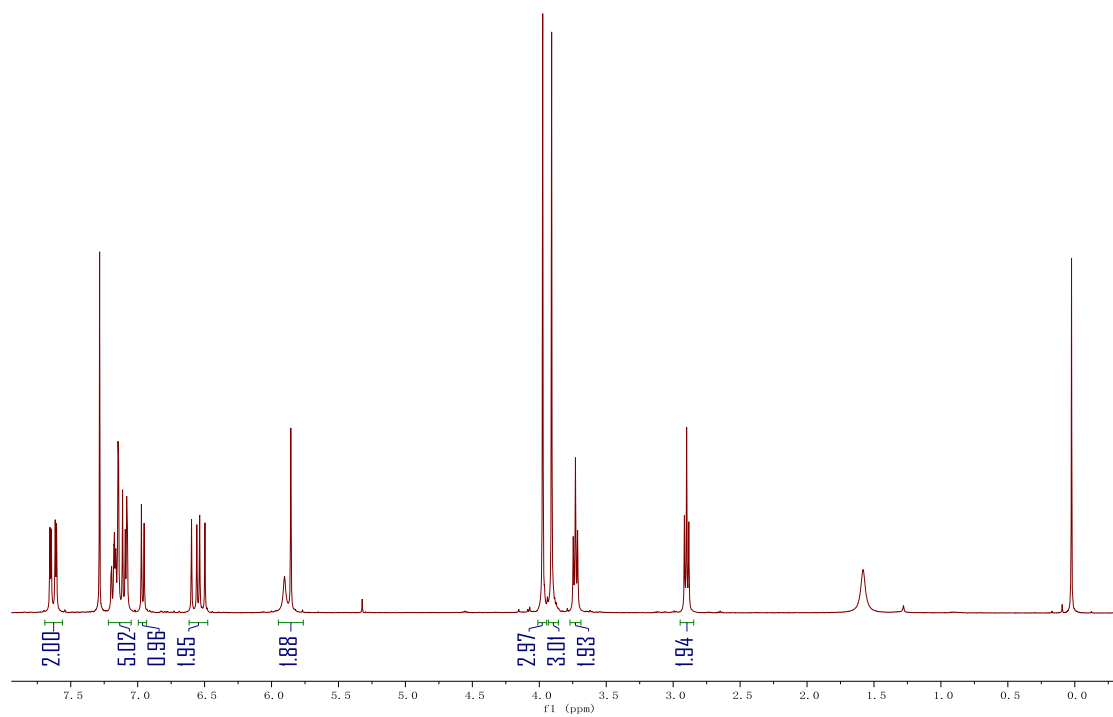


Figure S7. ^1H NMR of curcumin- N_3 in CDCl_3 .

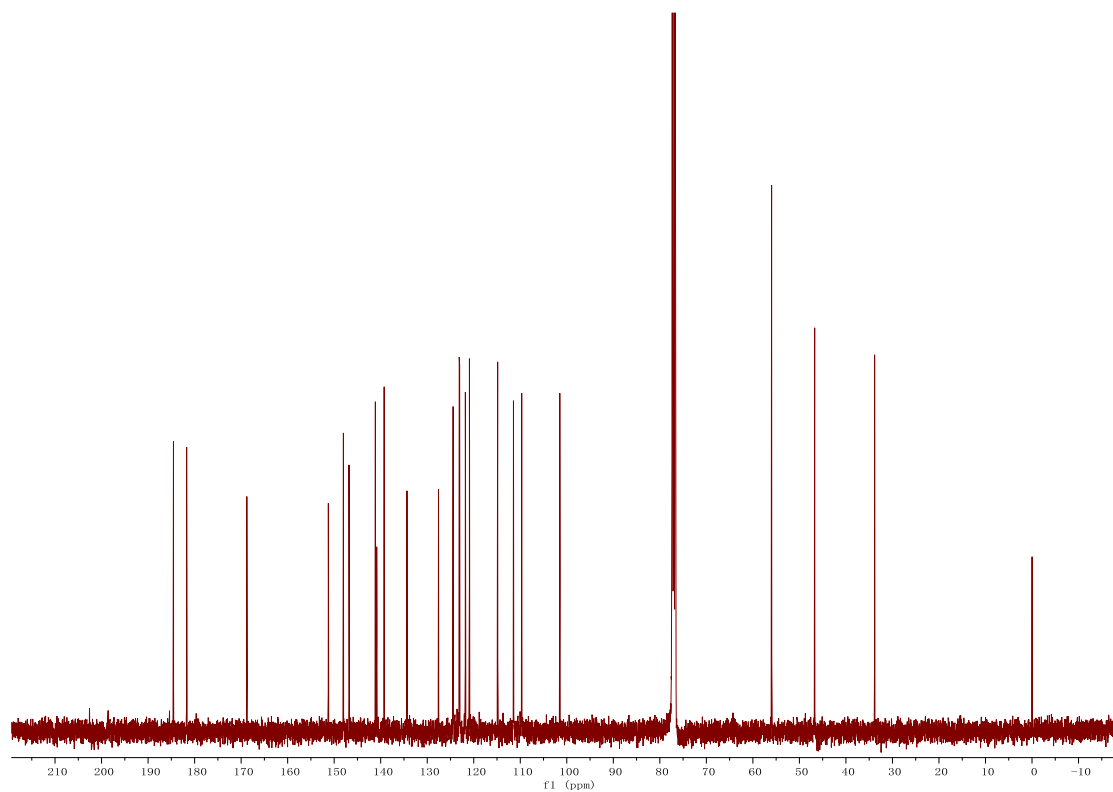


Figure S8. ^{13}C NMR of curcumin- N_3 in CDCl_3 .

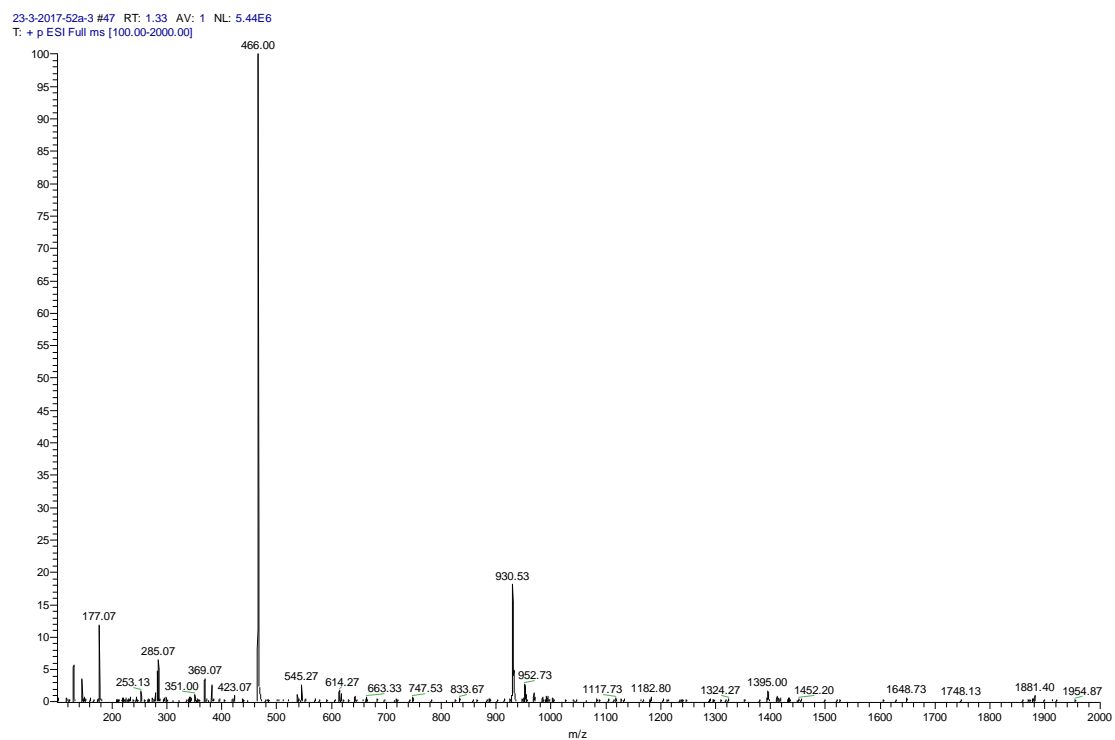


Figure S9. ESI-MS of curcumin- N_3 . m/z : 466.00 $[M+H]^+$, 930.53 $[2M+H]^+$.

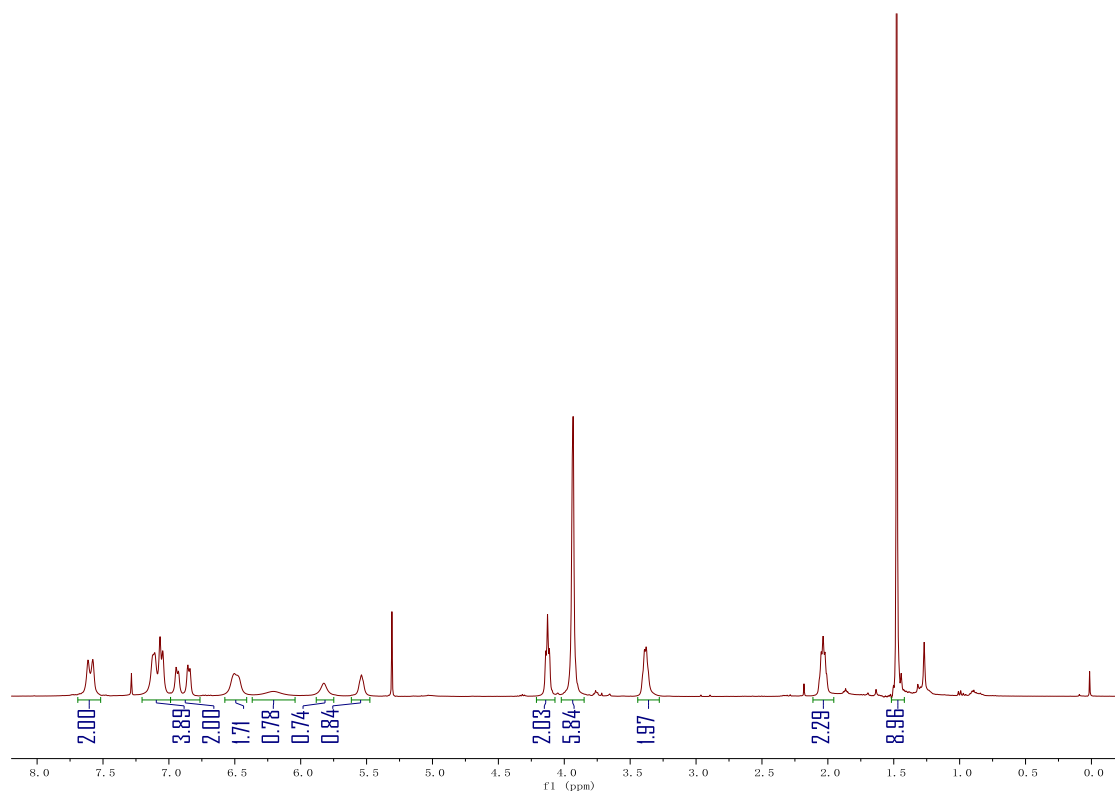


Figure S10. 1H NMR of Boc-amino-curcumin in $CDCl_3$.

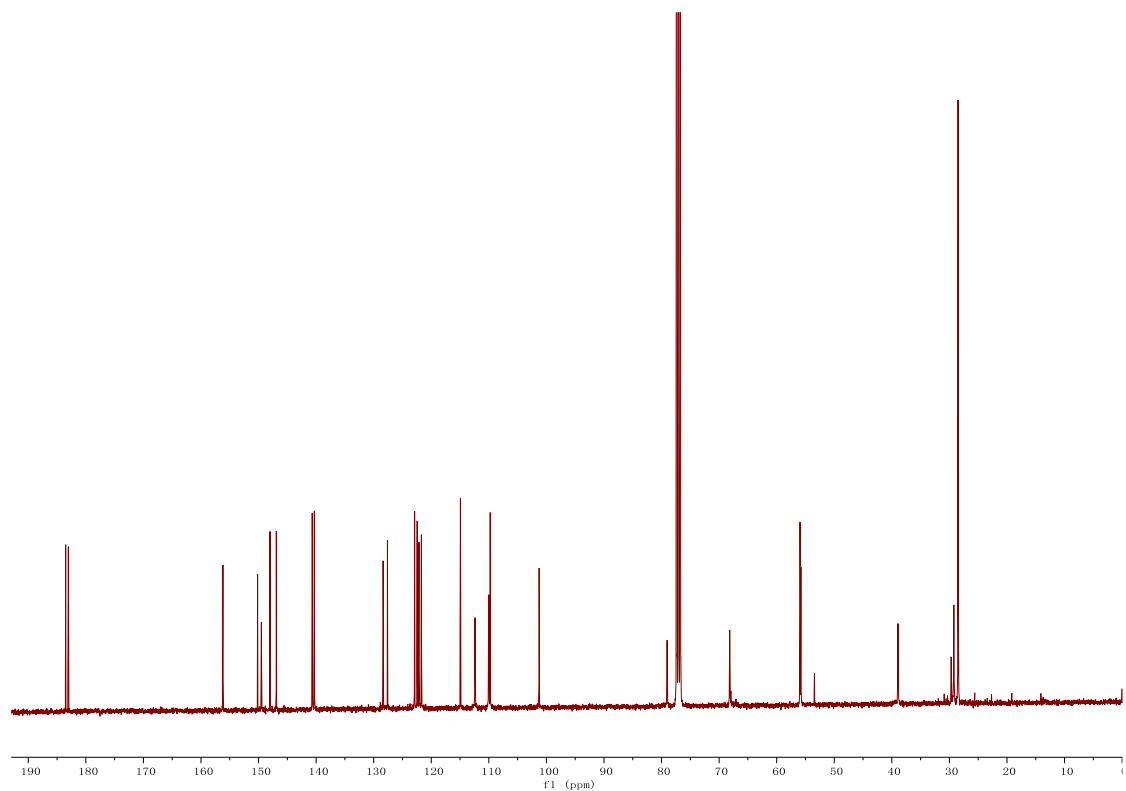


Figure S11. ^{13}C NMR of Boc-amino-curcumin in CDCl_3 .

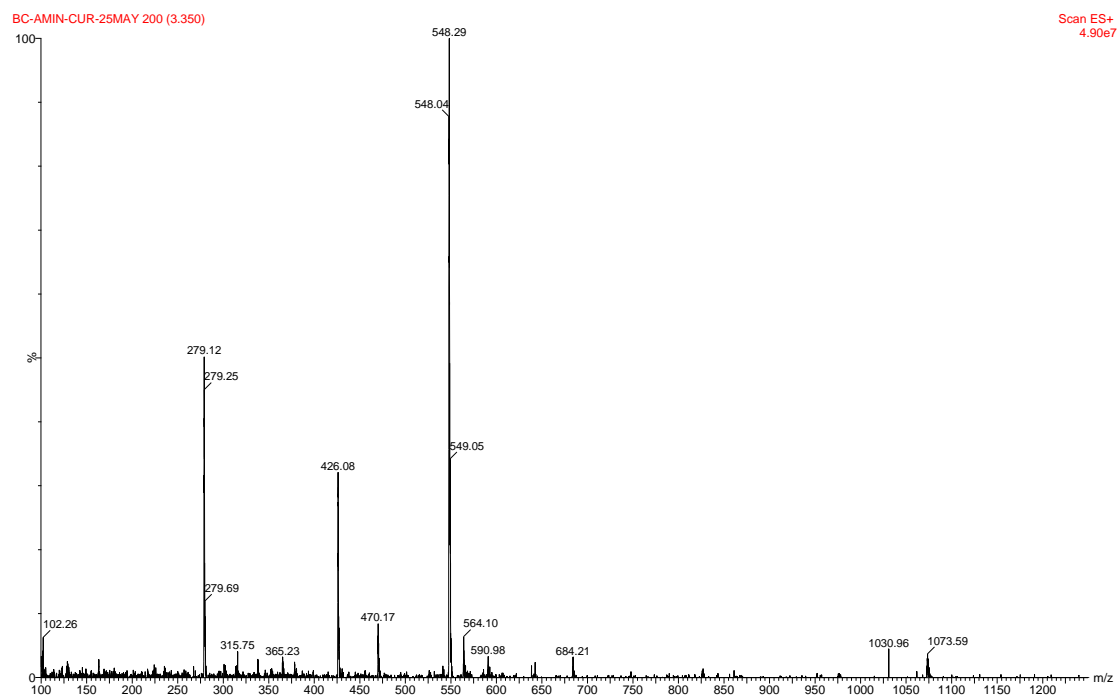


Figure S12. ESI-MS of Boc-amino-curcumin. m/z : 548.29 $[\text{M}+\text{Na}]^+$.

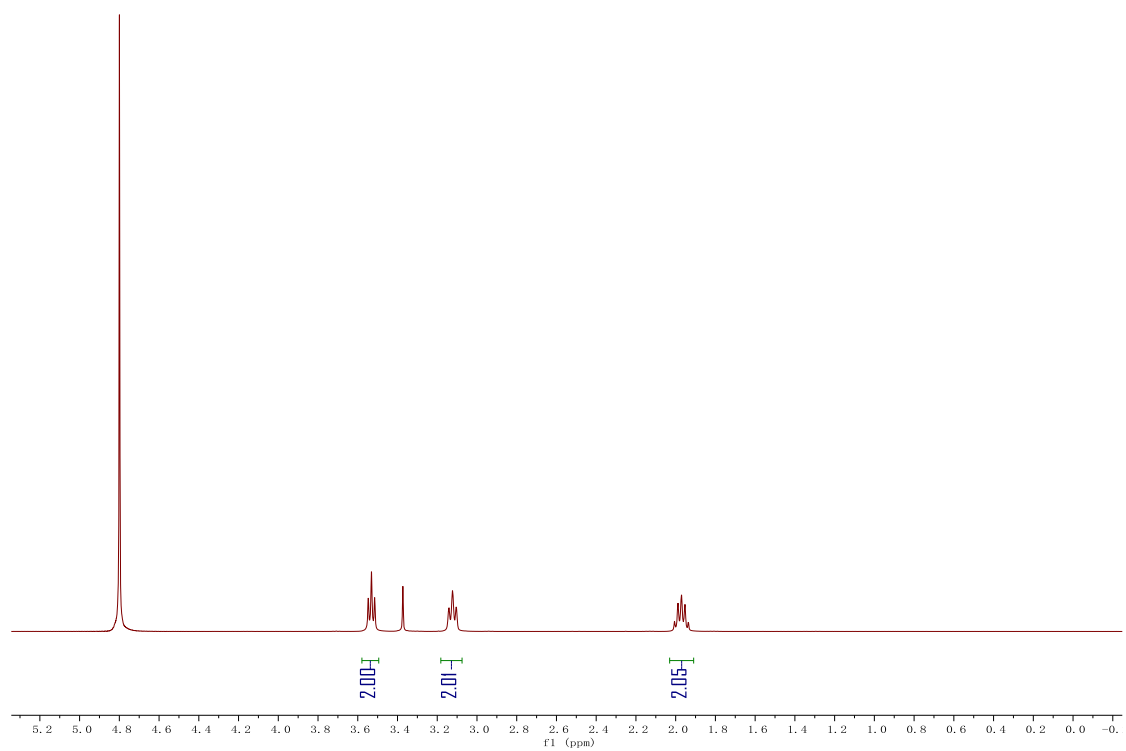


Figure S13. ^1H NMR of 3-azidopropylamine in D_2O .

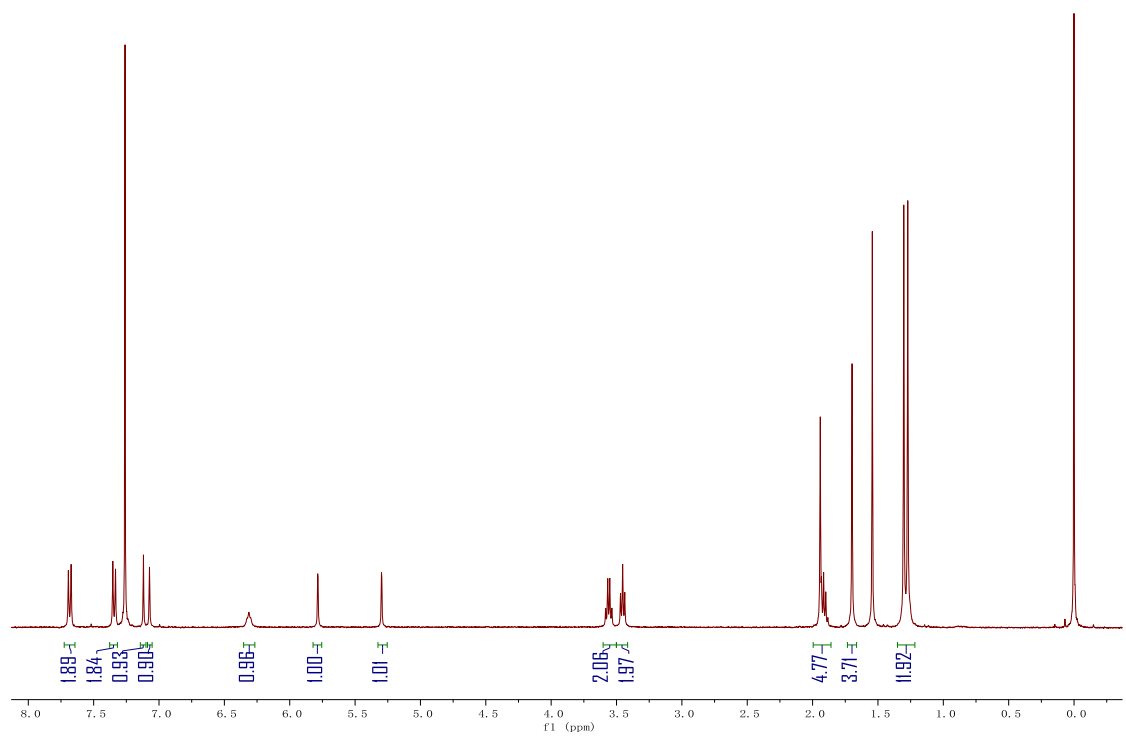


Figure S14. ^1H NMR of bexarotene- N_3 in CDCl_3 .

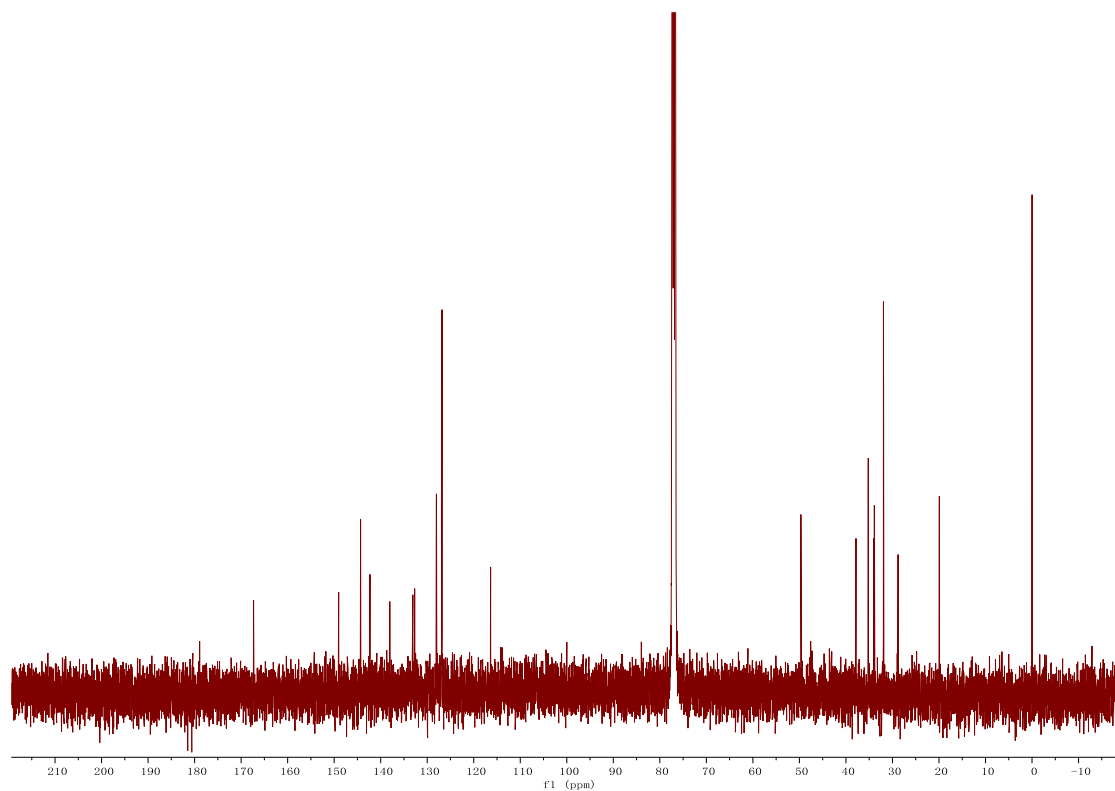


Figure S15. ^{13}C NMR of bexarotene- N_3 in CDCl_3 .

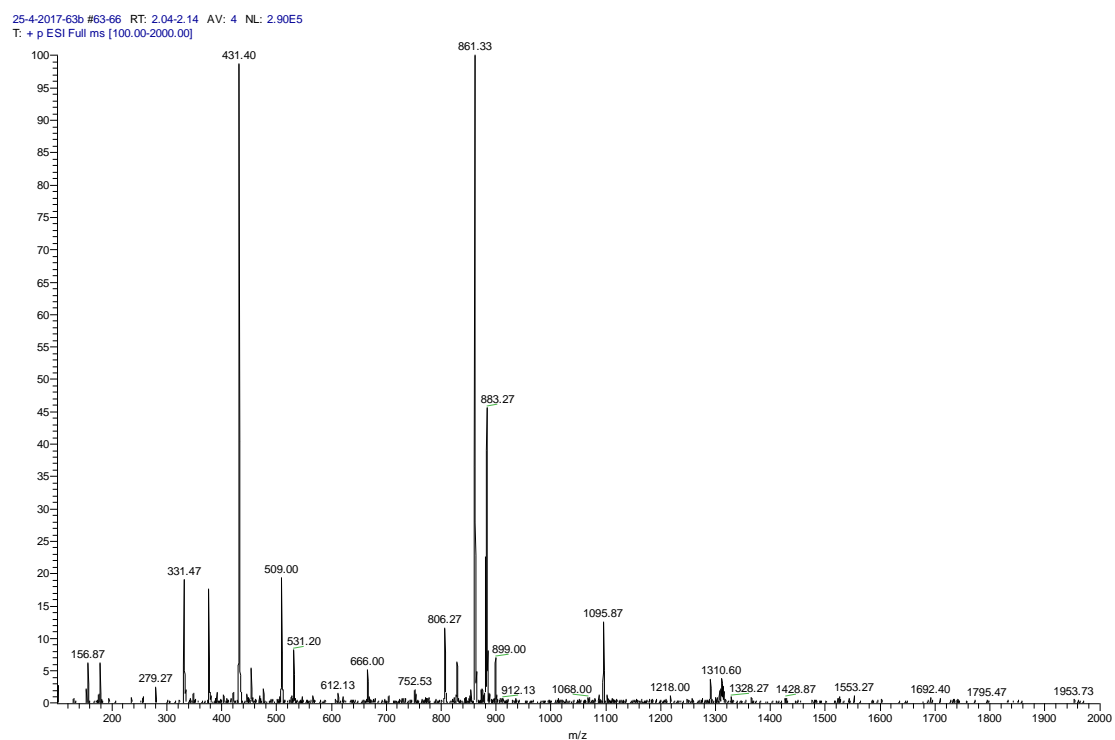


Figure S16. ESI-MS of bexarotene- N_3 . m/z : 431.40 $[\text{M}+\text{H}]^+$, 861.33 $[2\text{M}+\text{H}]^+$, 883.27 $[2\text{M}+\text{Na}]^+$.

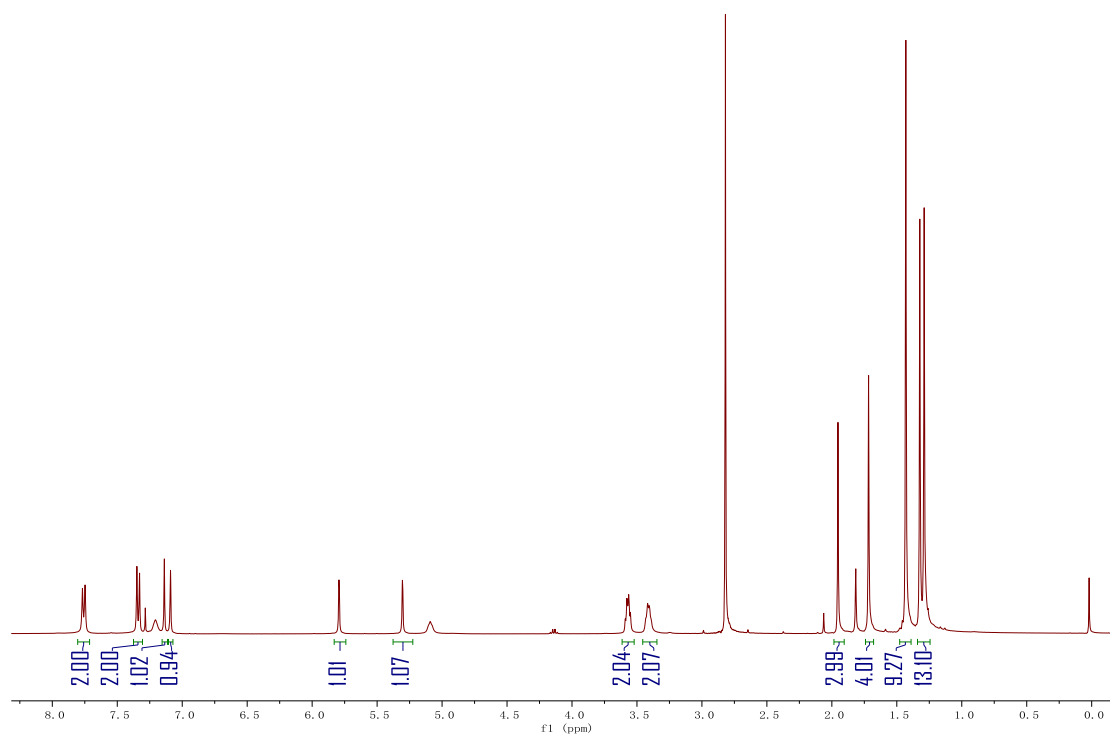


Figure 17. ¹H NMR of Boc-amino-bexarotene in CDCl₃.

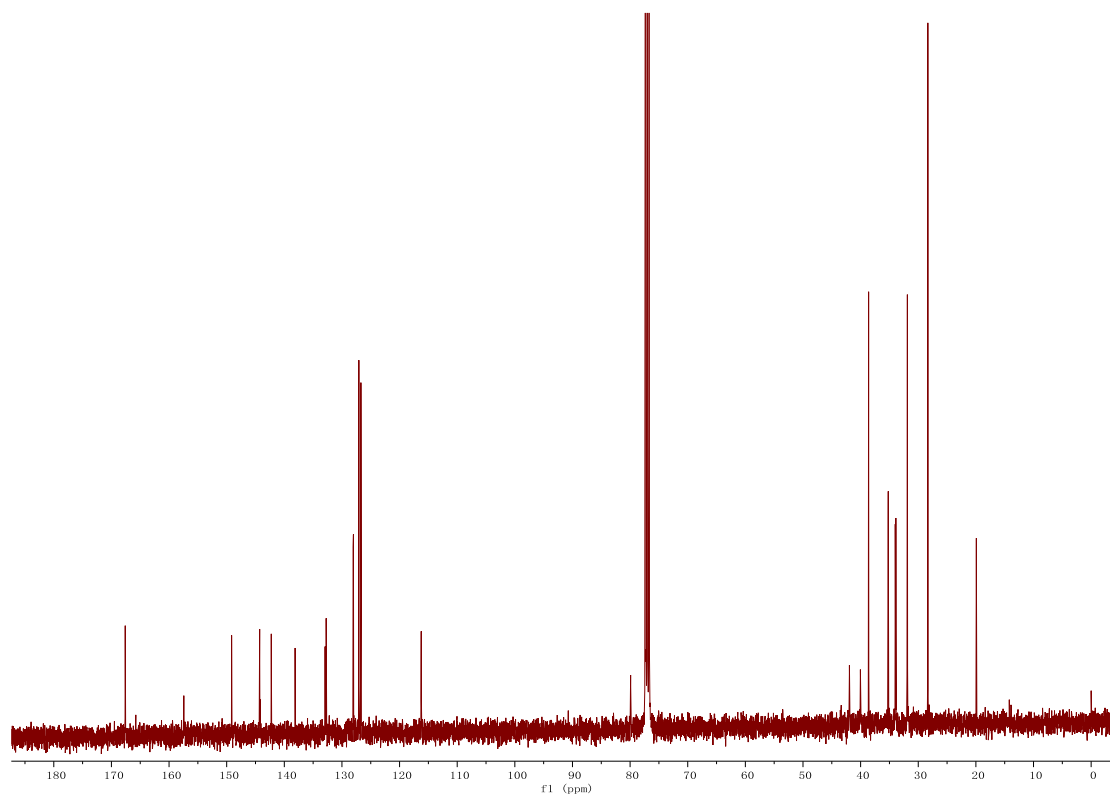


Figure 18. ¹³C NMR of Boc-amino-bexarotene in CDCl₃.

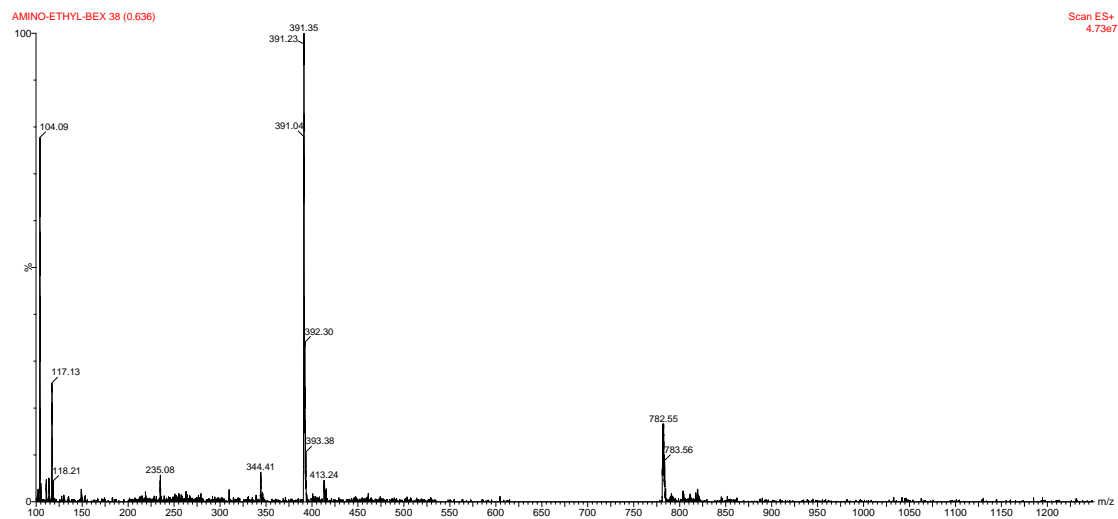


Figure 19. ESI-MS of Boc-amino-bexarotene. $m/z = 391.35$ $[M+H]^+$.

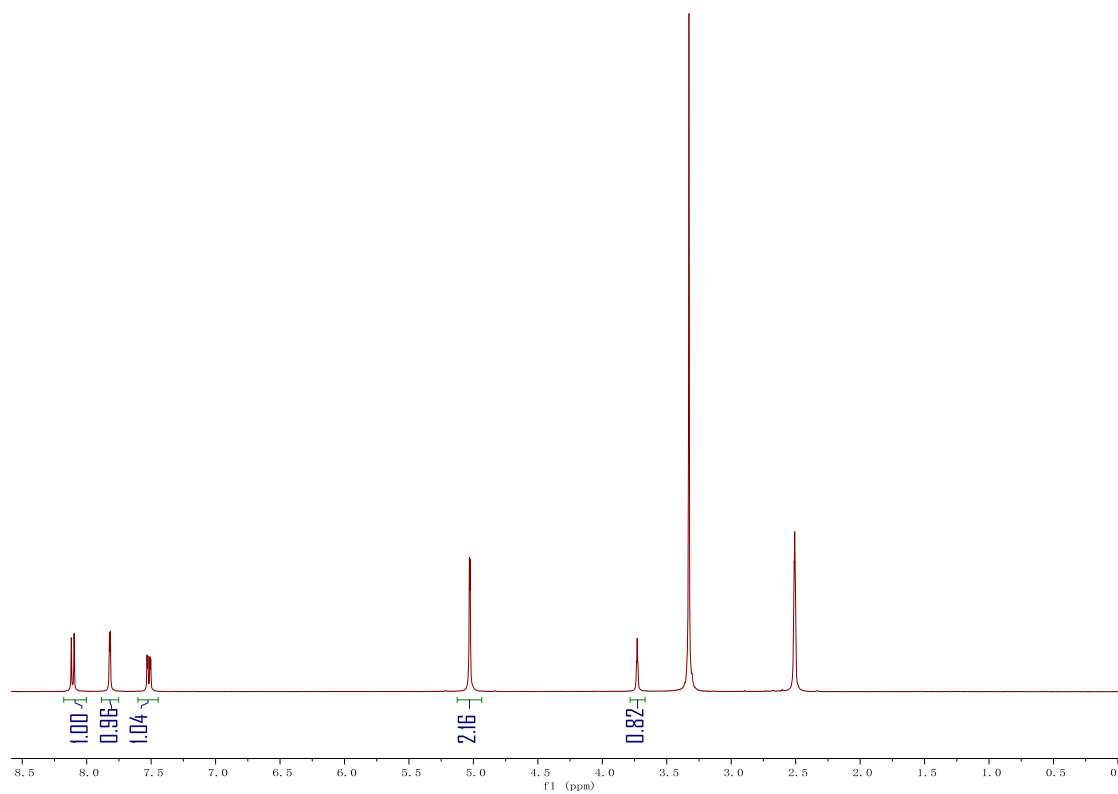


Figure S20. ^1H NMR of phthalonitrile **2** in DMSO-d_6 .

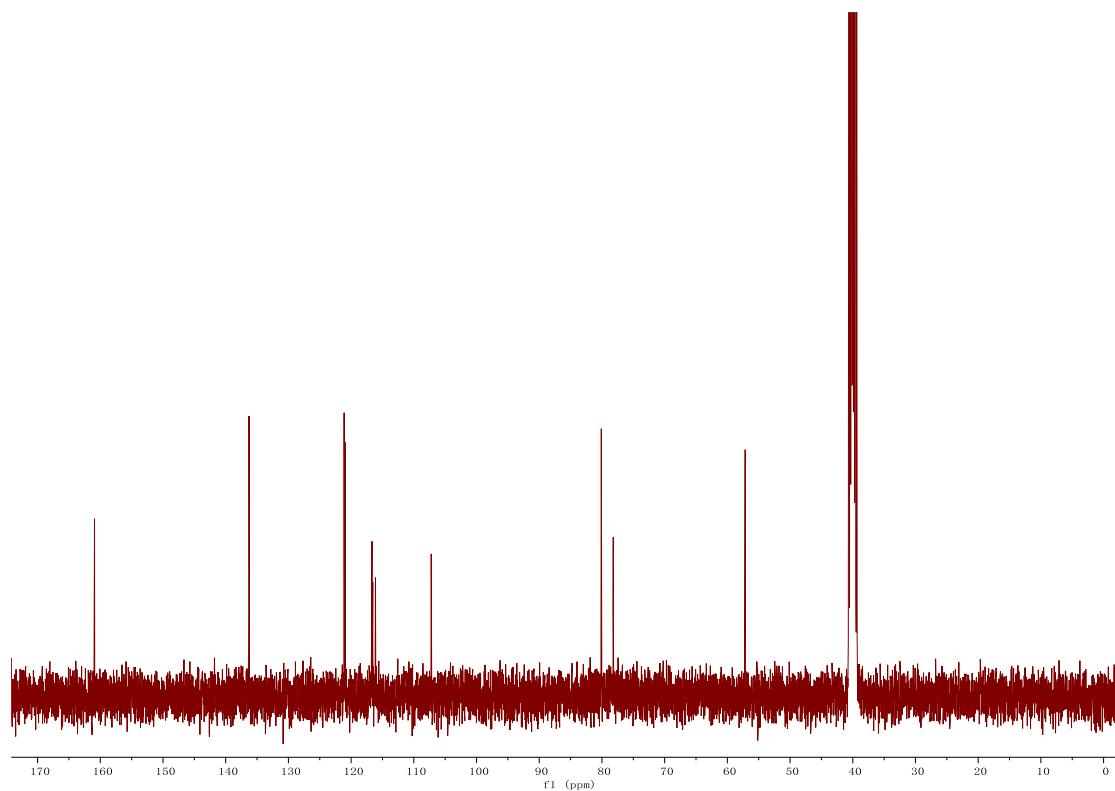


Figure S21. ^{13}C NMR of phthalonitrile **2** in $\text{DMSO-}d_6$.

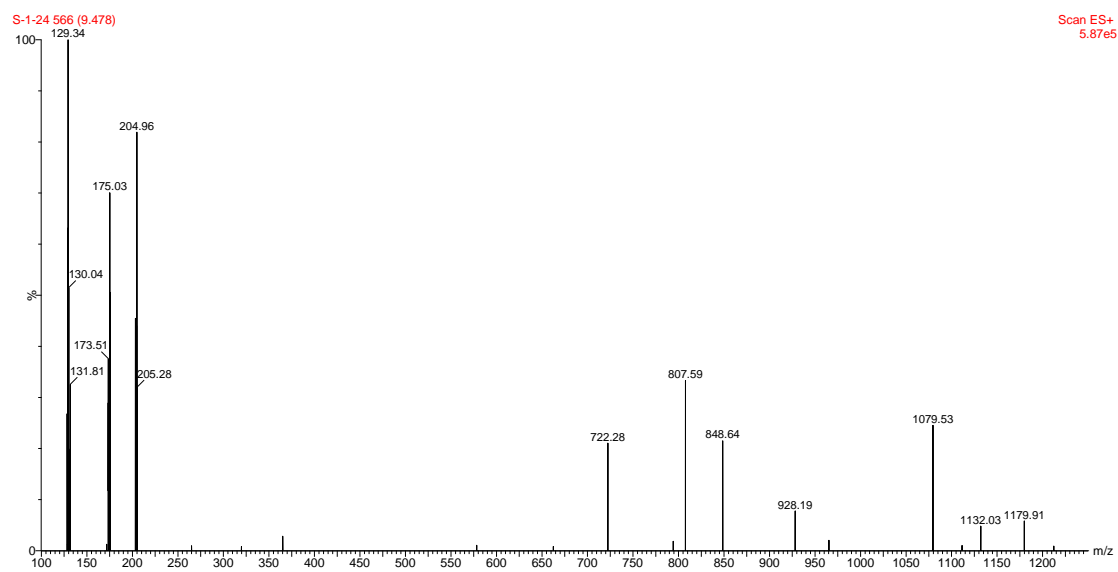


Figure S22. ESI-MS of phthalonitrile **2**, m/z : 204.96 $[\text{M}+\text{Na}]^+$.

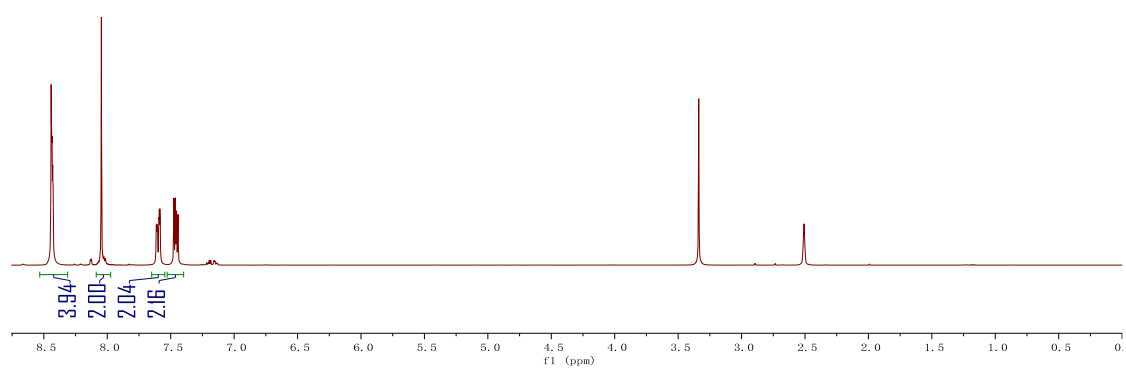


Figure S23. ^1H NMR of phthalonitrile **1** in DMSO-d_6 .

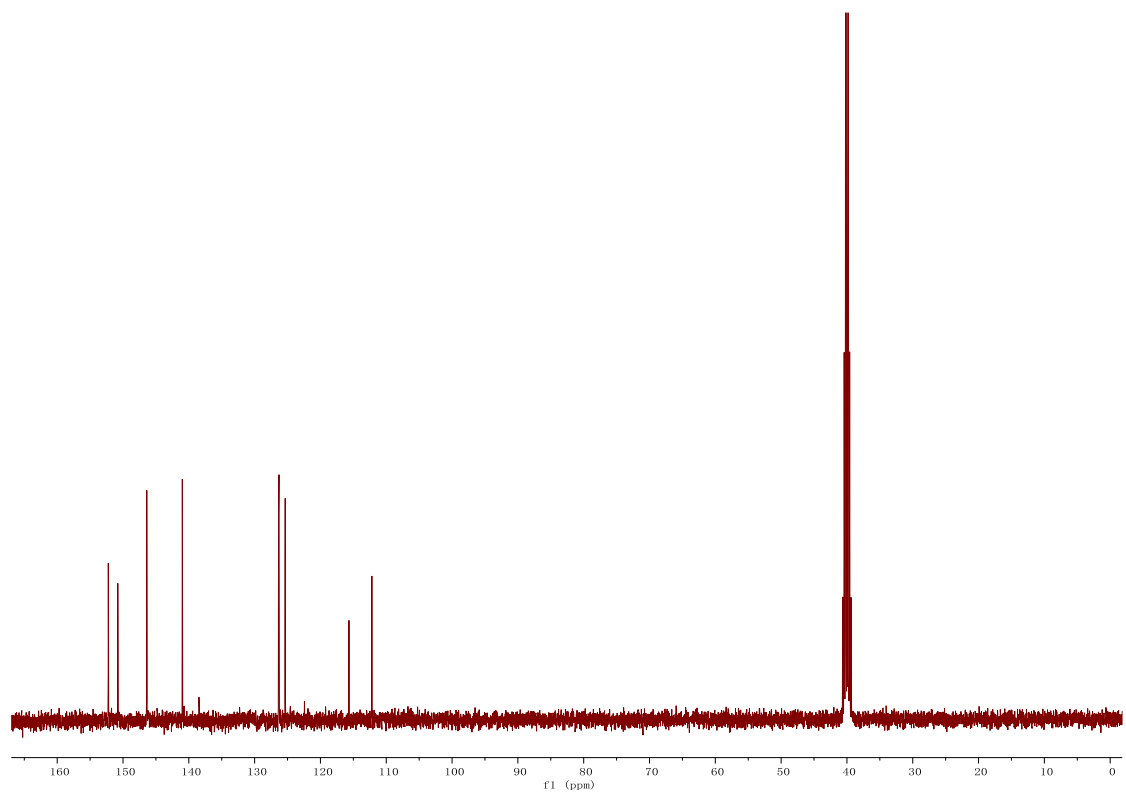


Figure S24. ^{13}C NMR of phthalonitrile **1** in DMSO-d_6 .

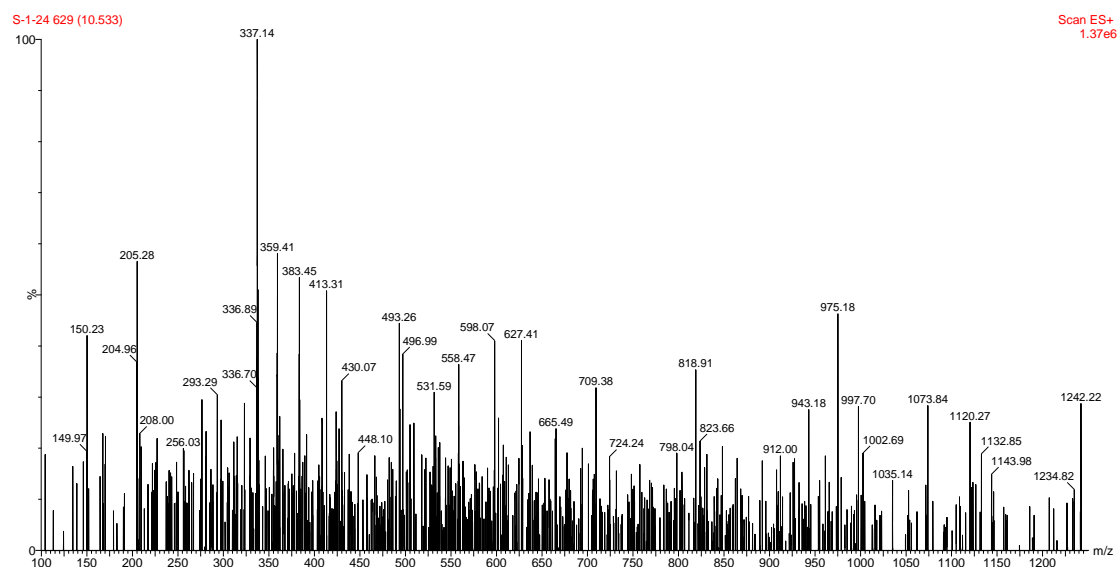


Figure S25. ESI-MS of phthalonitrile **1**, m/z : 337.14 $[M+Na]^+$.

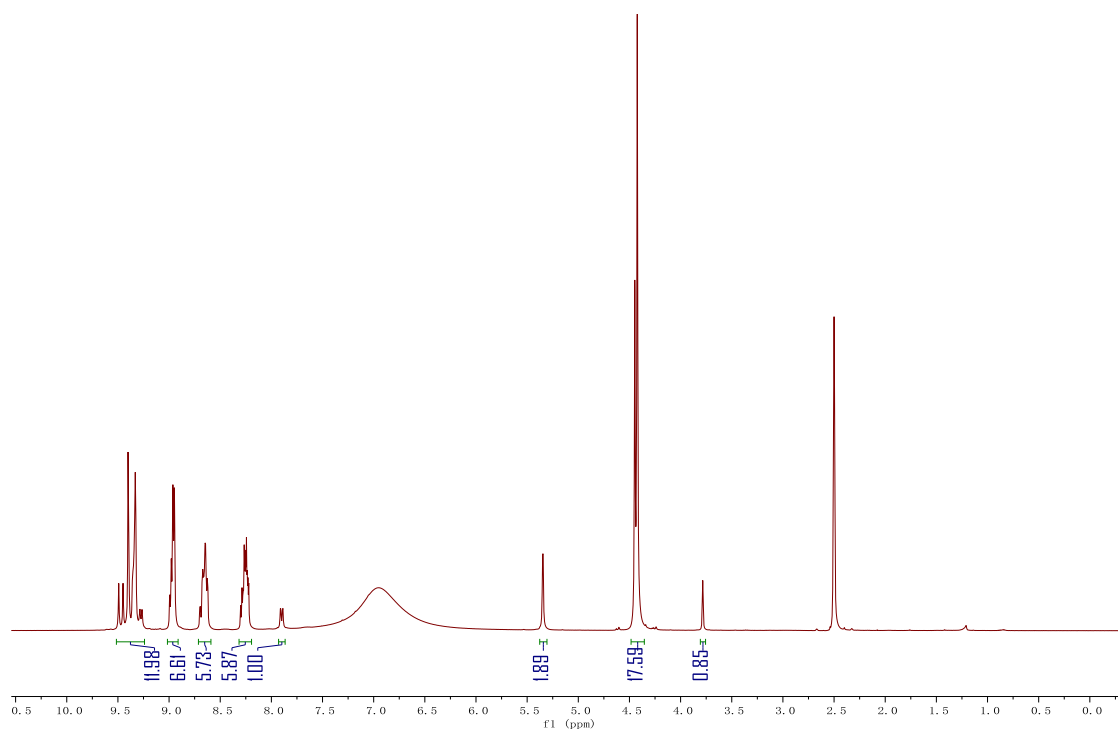


Figure S26. 1H NMR of asy-ZnPc in DMSO- d_6 . The anion is CF_3COO^- .

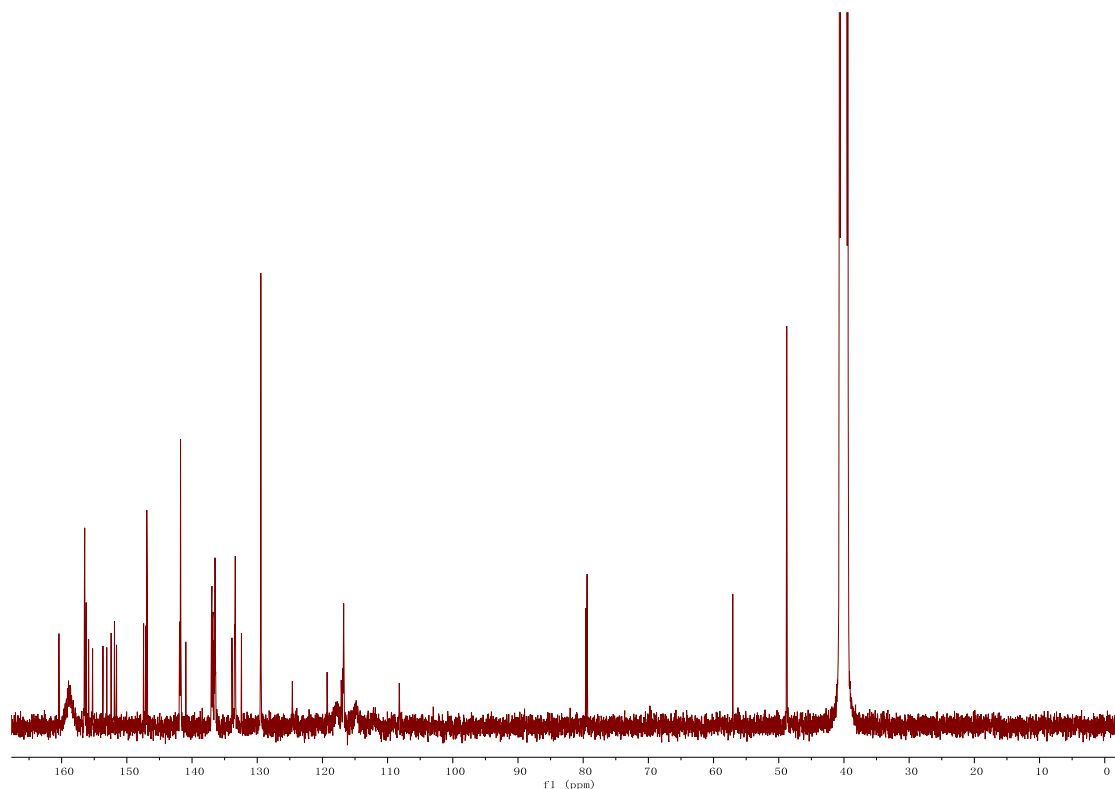


Figure S27. ^{13}C NMR of asy-ZnPc in DMSO-d_6 . The anion is CF_3COO^- .

15Dec2016-41b-4 #11-13 RT: 0.34-0.39 AV: 3 NL: 1.04E6
T: + p ESI Full ms [150.00-2000.00]

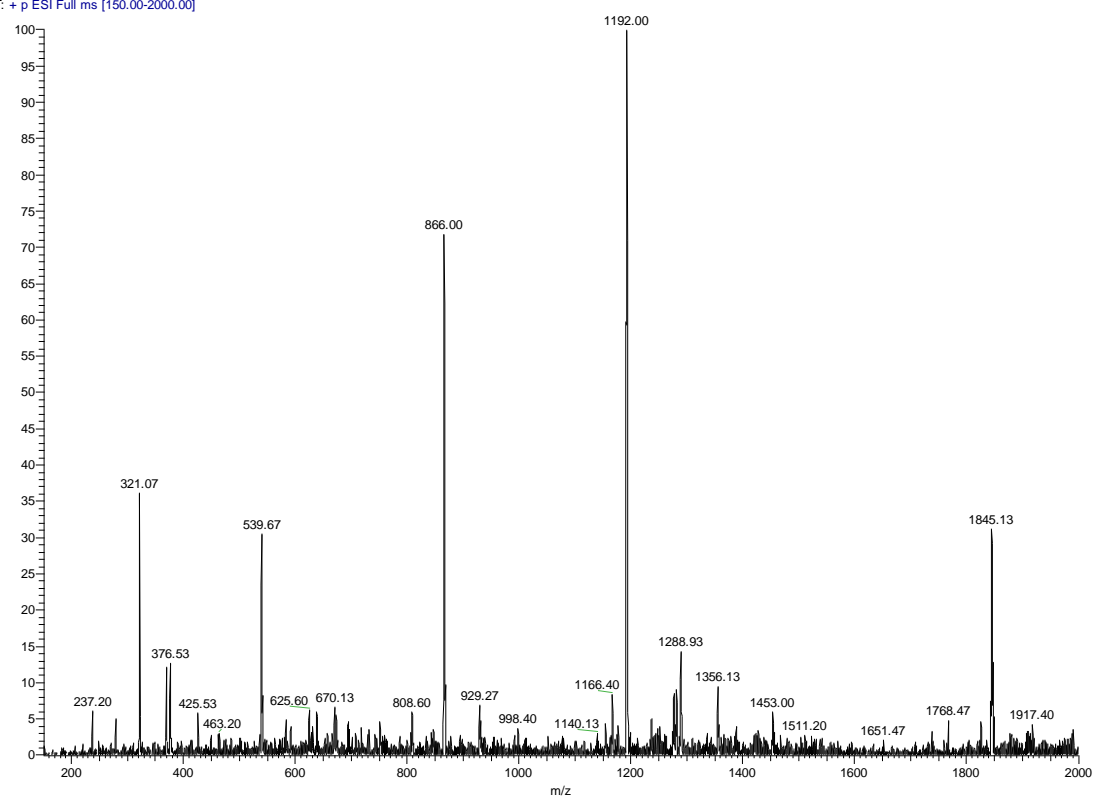


Figure S28. ESI-MS of sym-ZnPc, m/z : 321.07 $[2\text{M}+3\text{CF}_3\text{COO}]^{9+}$, 539.67 $[\text{M}+3\text{CF}_3\text{COO}]^{3+}$, 866.00 $[\text{M}+4\text{CF}_3\text{COO}]^{2+}$, 1192.00 $[2\text{M}+9\text{CF}_3\text{COO}]^{3+}$, 1845.13 $[\text{M}+5\text{CF}_3\text{COO}]^+$.

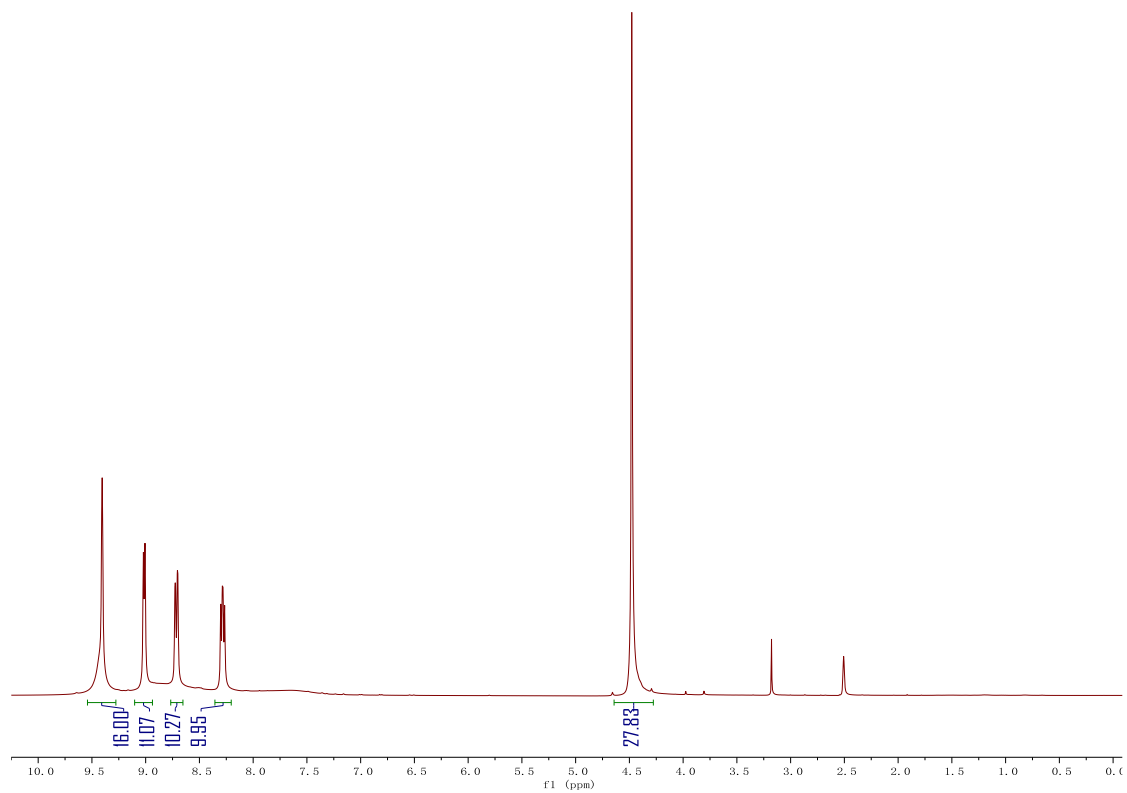


Figure S29. ^1H NMR of sym-ZnPc in DMSO- d_6 . The anion is CF_3COO^- .

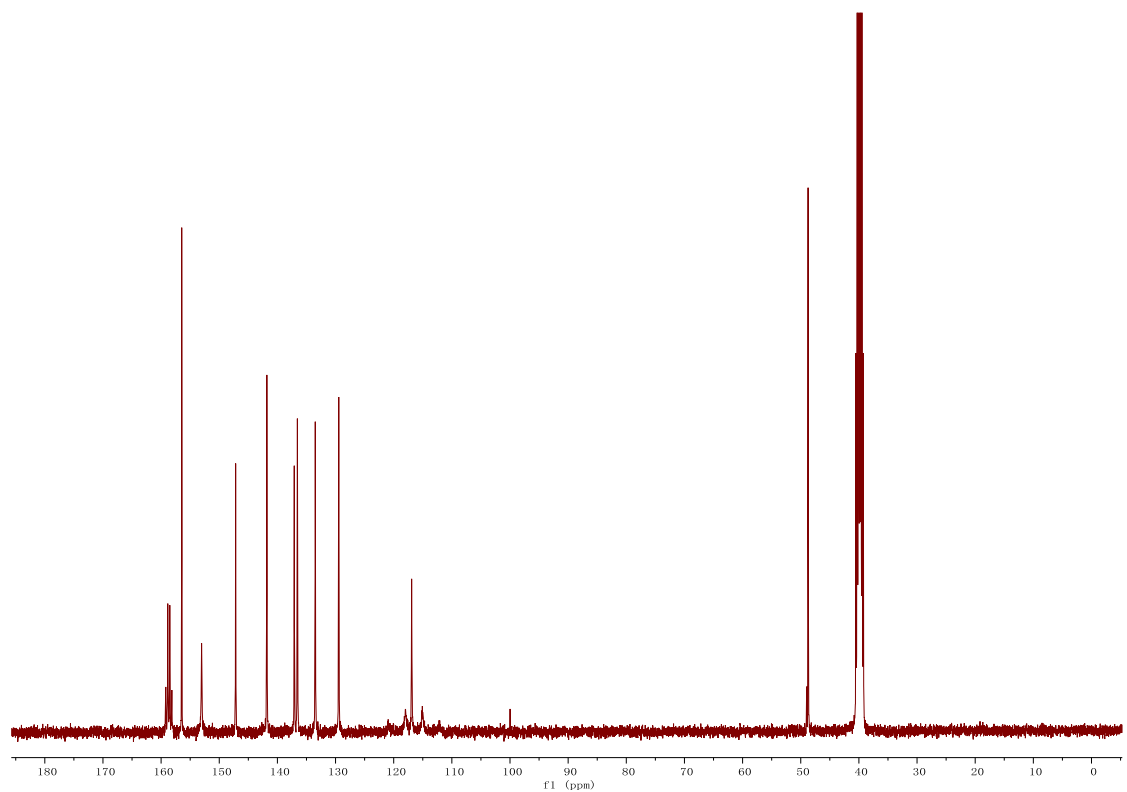


Figure S30. ^{13}C NMR of sym-ZnPc in DMSO- d_6 . The anion is CF_3COO^- .

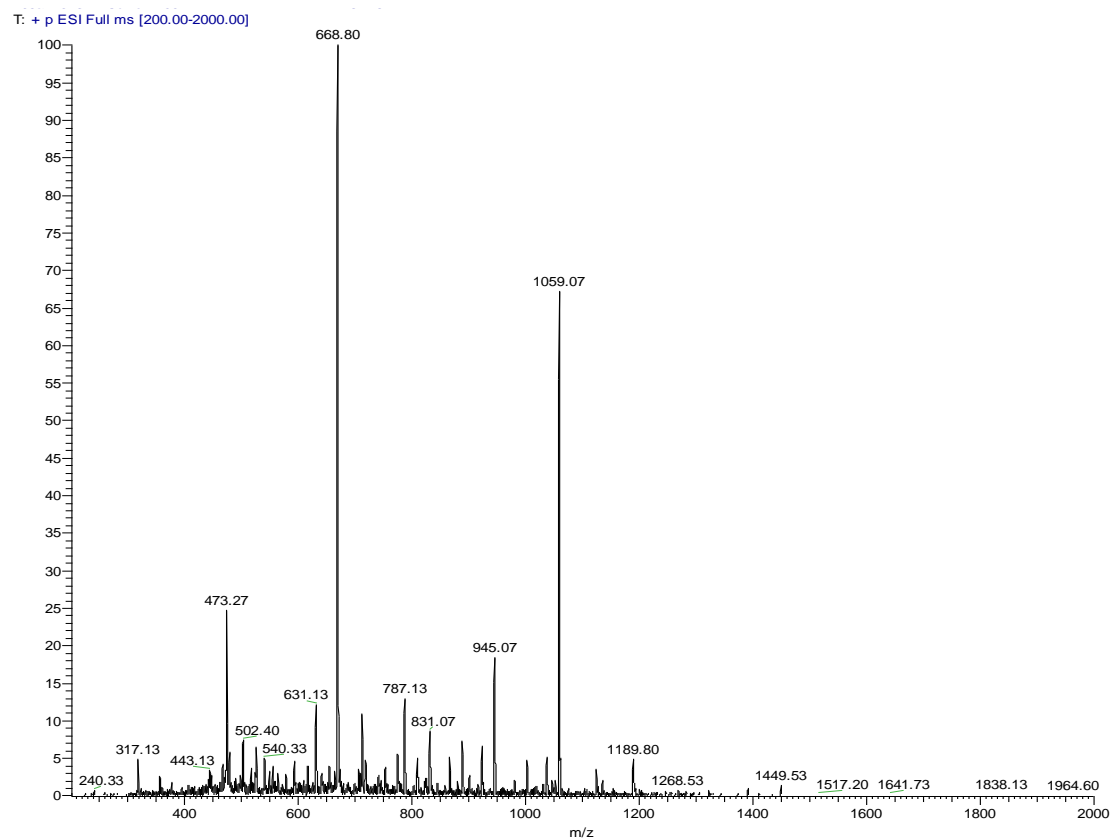


Figure S31. ESI-MS of sym-ZnPc. m/z: 668.80 $[M+5CF_3COO]^3+$, 1059.07 $[M+6CF_3COO]^2+$.

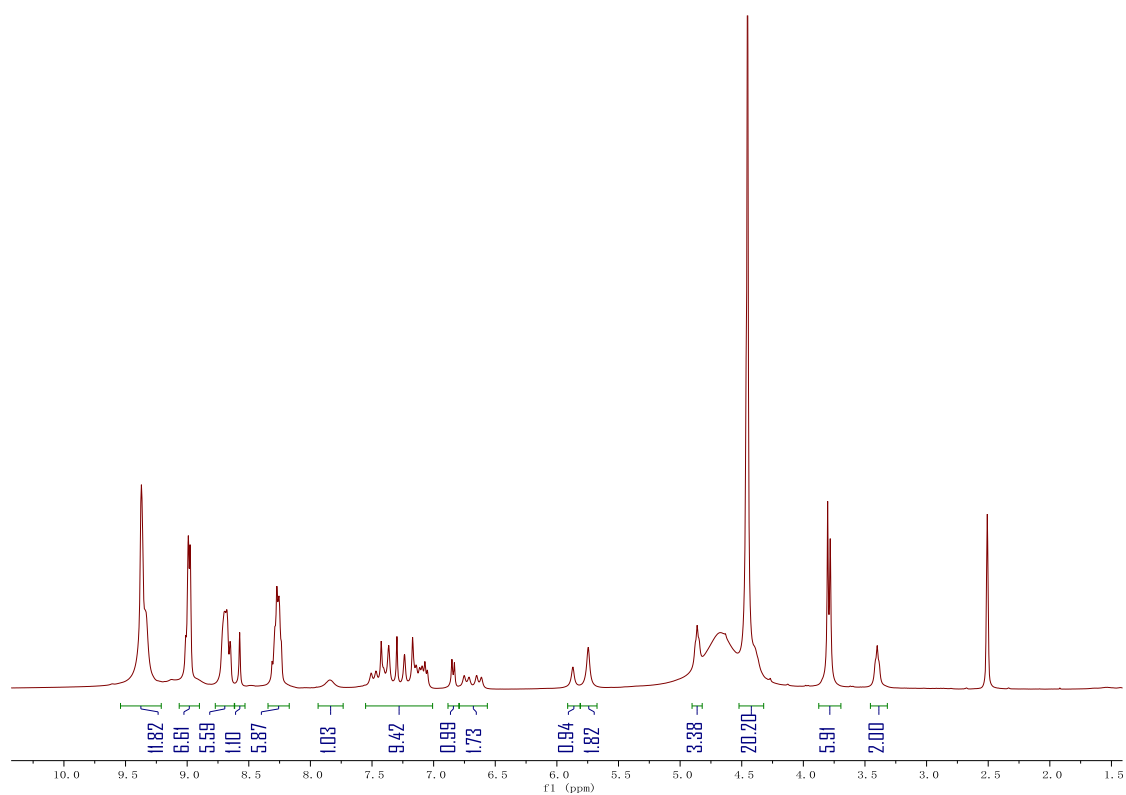


Figure S32. 1H NMR of ZnPcCur in DMSO- d_6 . The anion is CF_3COO^- .

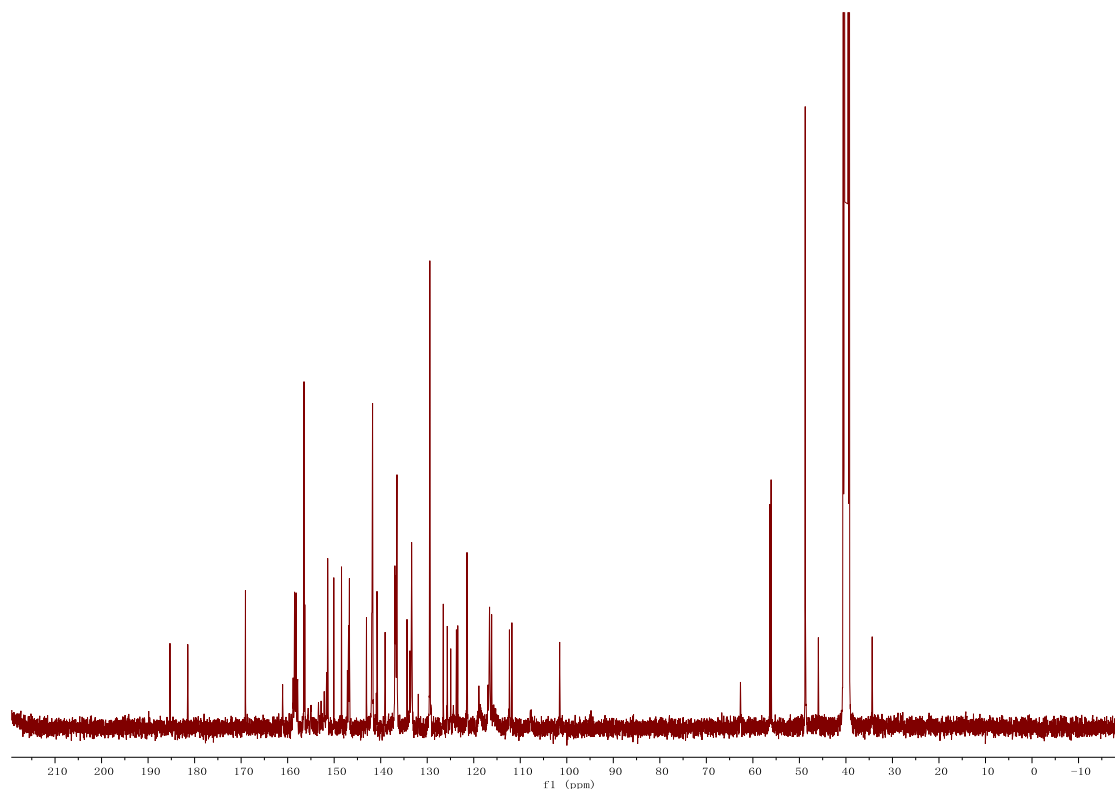


Figure S33. ^{13}C NMR of ZnPcCur in DMSO- d_6 . The anion is CF_3COO^- .

25April2017-ZnPcCurcumin-1 #82 RT: 2.75 AV: 1 NL: 2.80E5
T: + p ESI Full ms [100.00-2000.00]

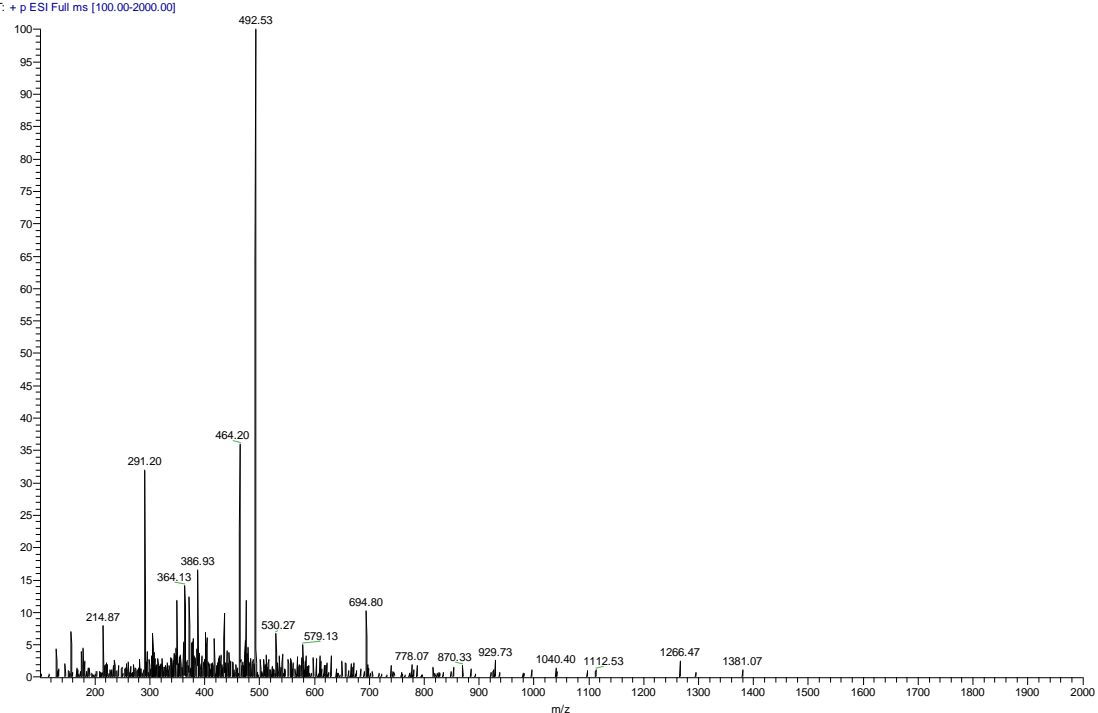


Figure S34. ESI-MS of ZnPc-Curcumin. m/z : 291.20 $[\text{M}]^{6+}$, 464.20 $[\text{M}-\text{H}+\text{CF}_3\text{COO}]^{4+}$, 492.53 $[\text{M}+2\text{CF}_3\text{COO}]^{4+}$, 694.80 $[\text{M}+3\text{CF}_3\text{COO}]^{3+}$.

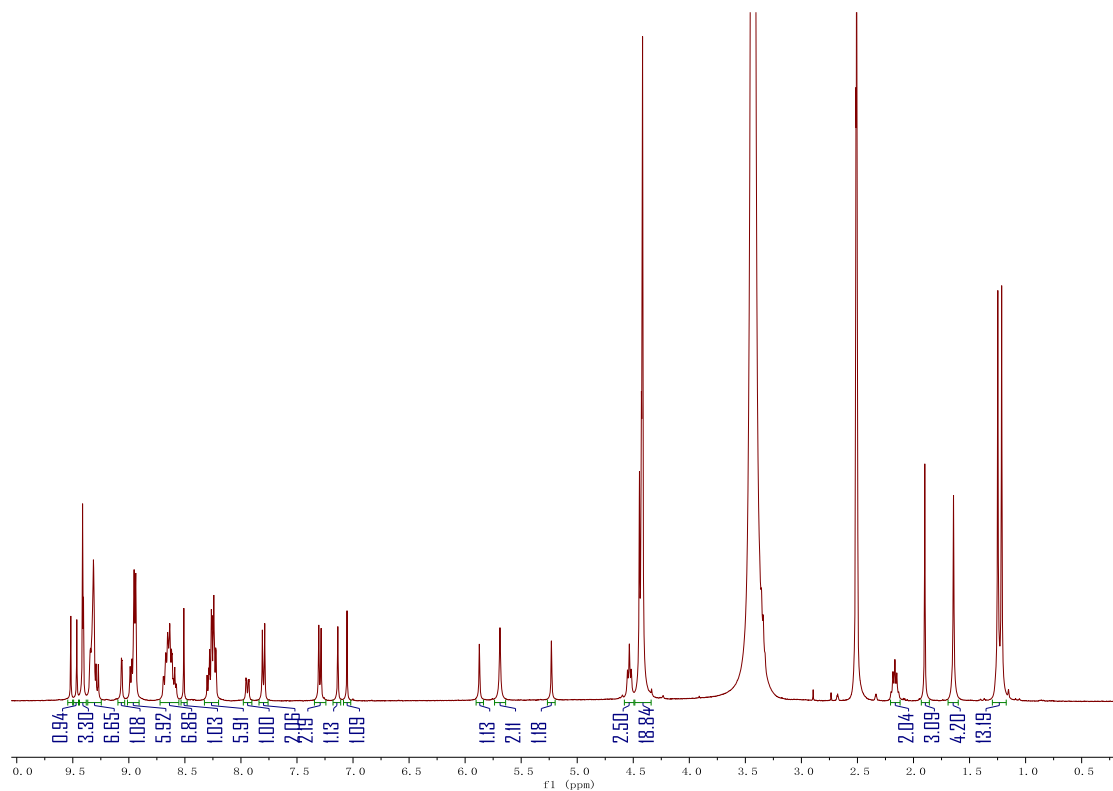


Figure S35. ^1H NMR of ZnPcBex in DMSO- d_6 . The anion is CF_3COO^- .

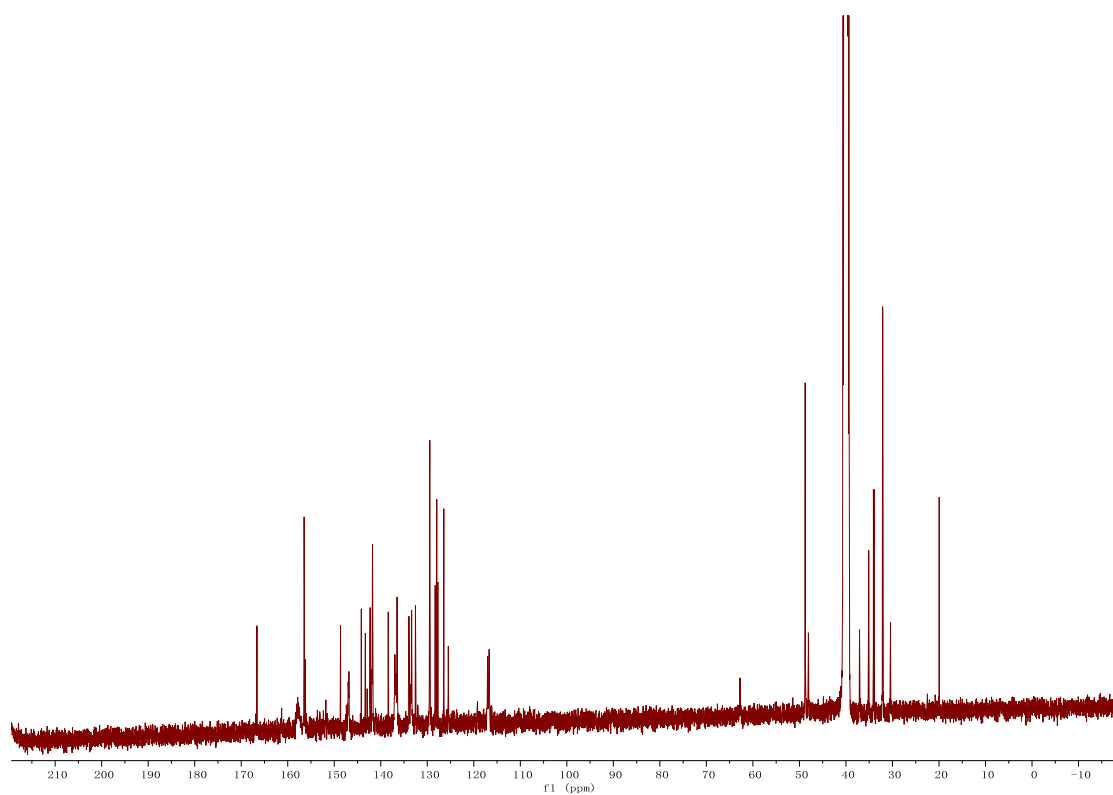


Figure S36. ^{13}C NMR of ZnPcBex in DMSO- d_6 . The anion is CF_3COO^- .

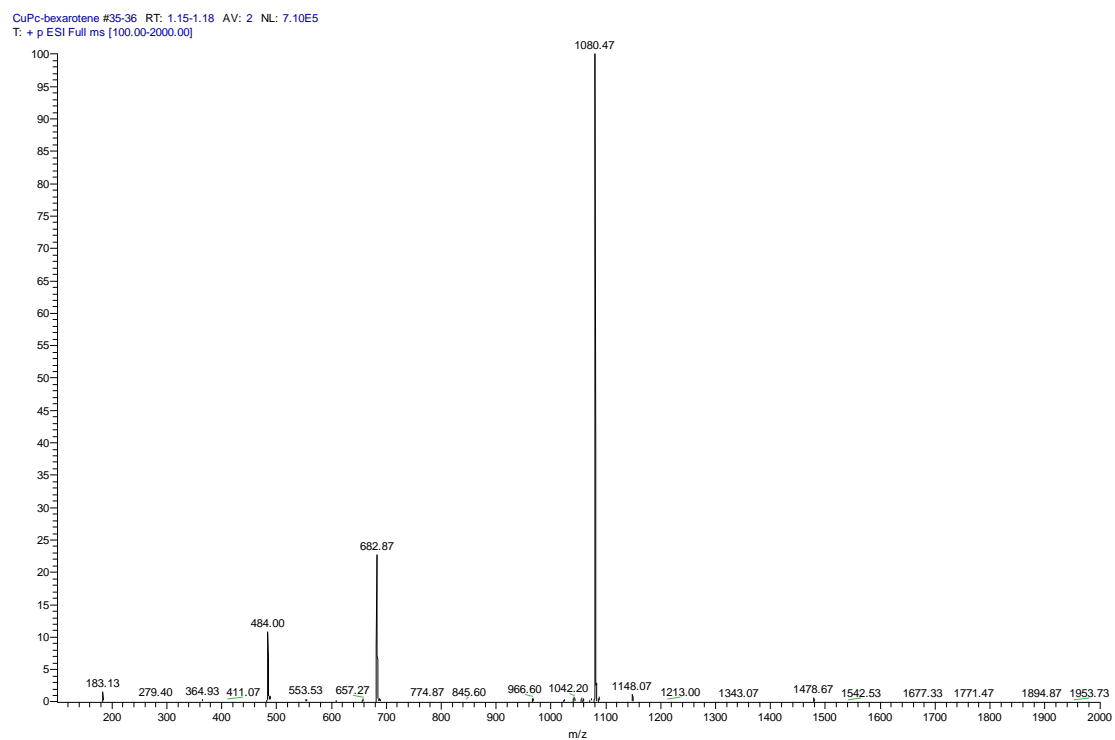


Figure S37. ESI-MS of ZnPc-Bexarotene. m/z : 484.00 $[M+2CF_3COO]^{4+}$, 682.87 $[M+3CF_3COO]^{3+}$, 1080.47 $[M+4CF_3COO]^{2+}$.

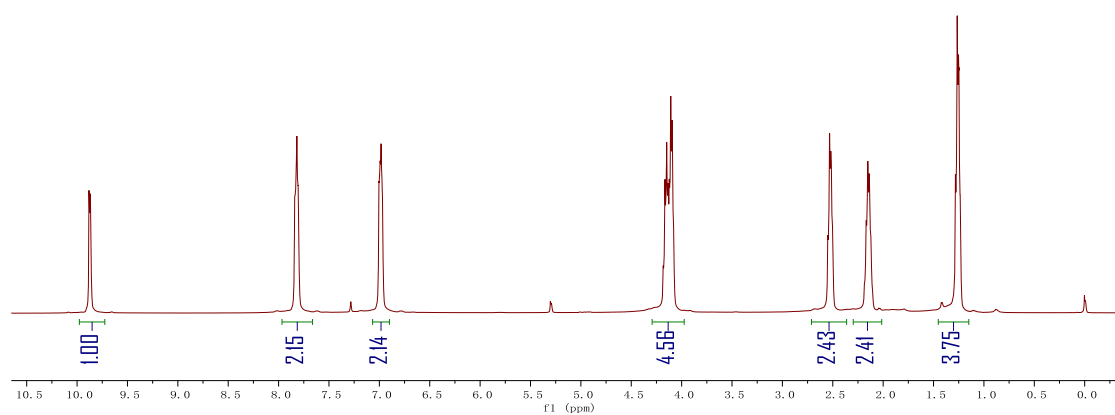


Figure S38. 1H NMR of compound **4** in $CDCl_3$.

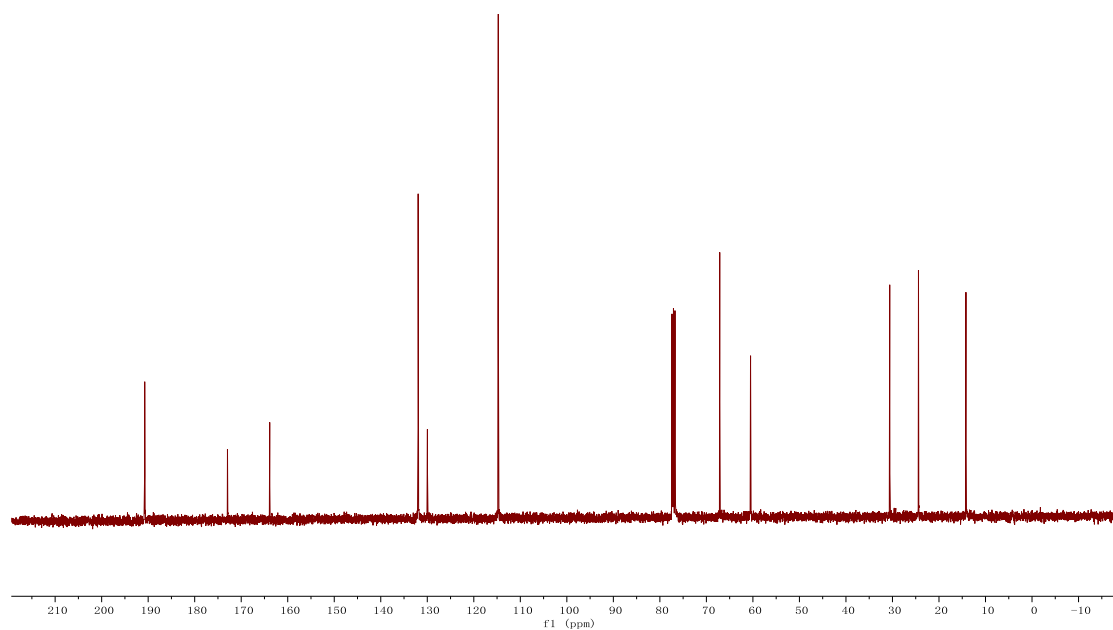


Figure S39. ^{13}C NMR of compound **4** in CDCl_3 .

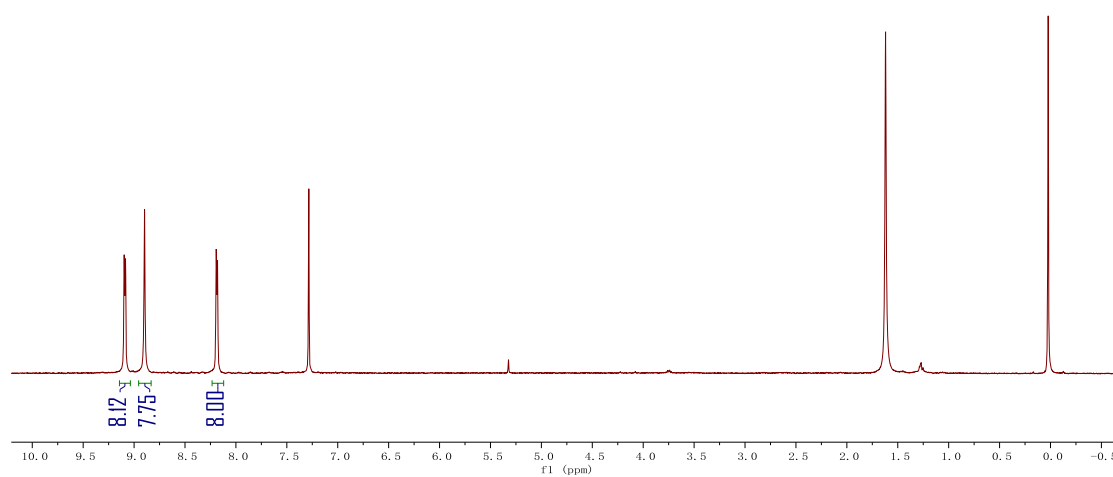


Figure S40. ^1H NMR of porphyrin **8** in CDCl_3 .

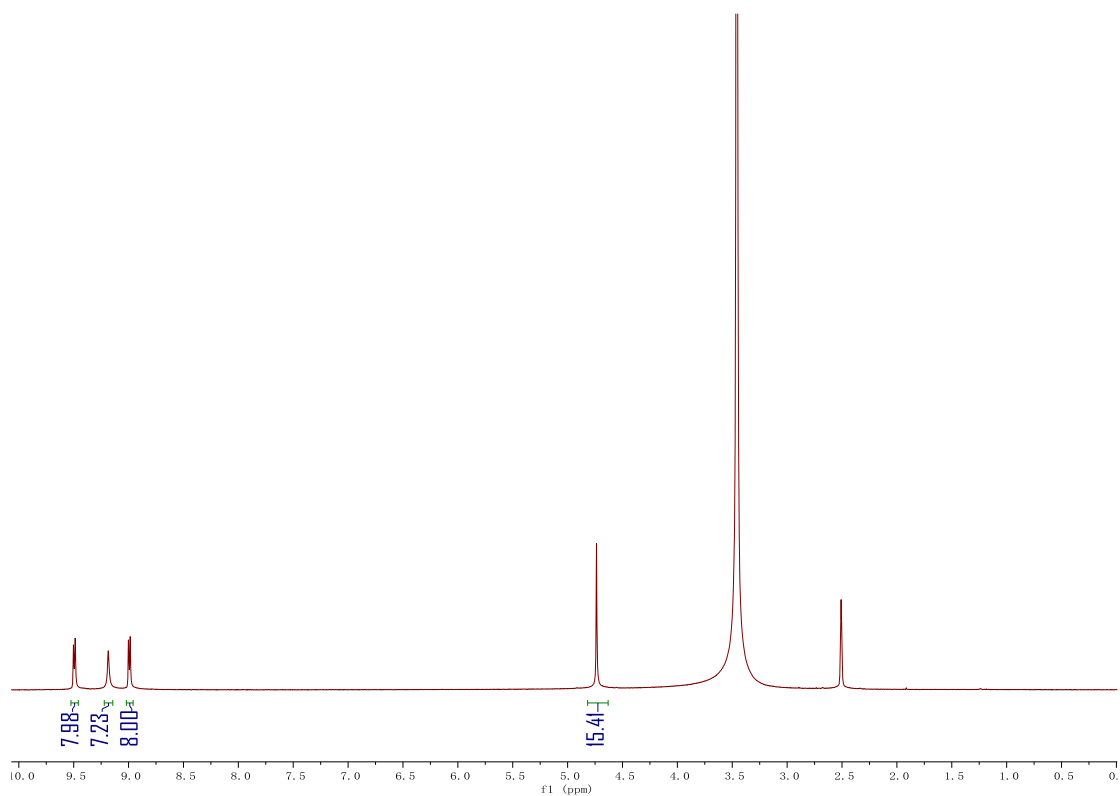


Figure 41. ^1H NMR of sym-Porp in DMSO-d_6 .

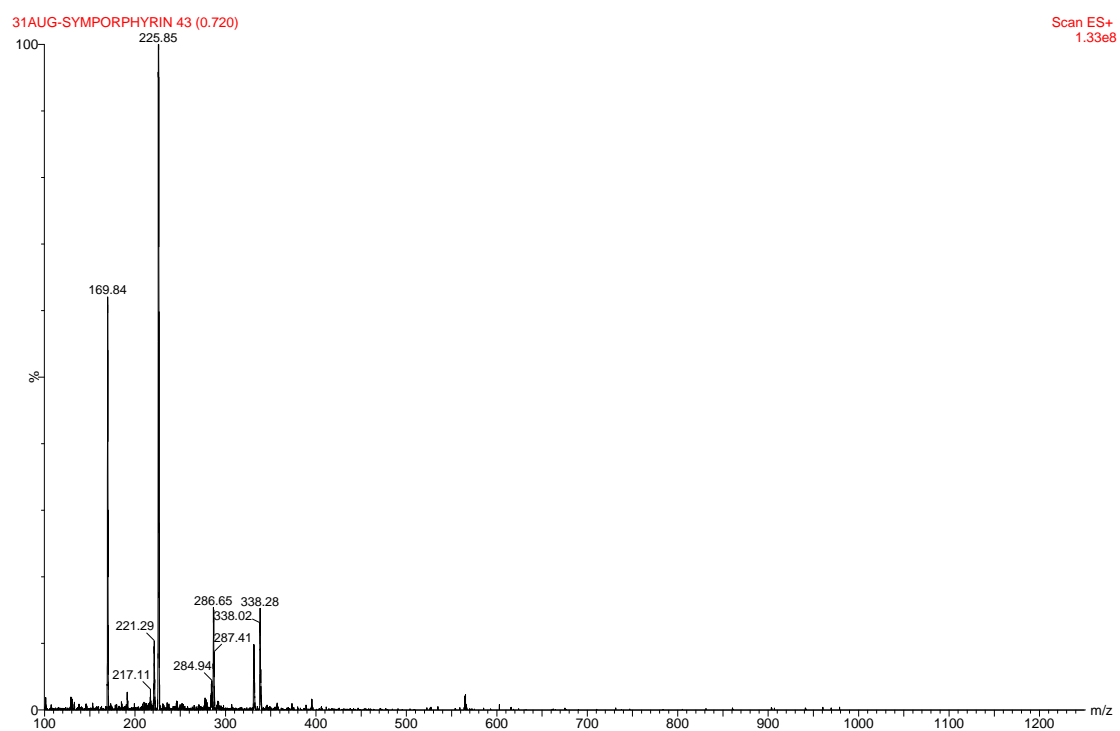


Figure S42. ESI-MS of sym-Porp. $m/z = 169.84$ $[\text{M}]^{4+}$, 225.85 $[\text{M-H}]^{3+}$.

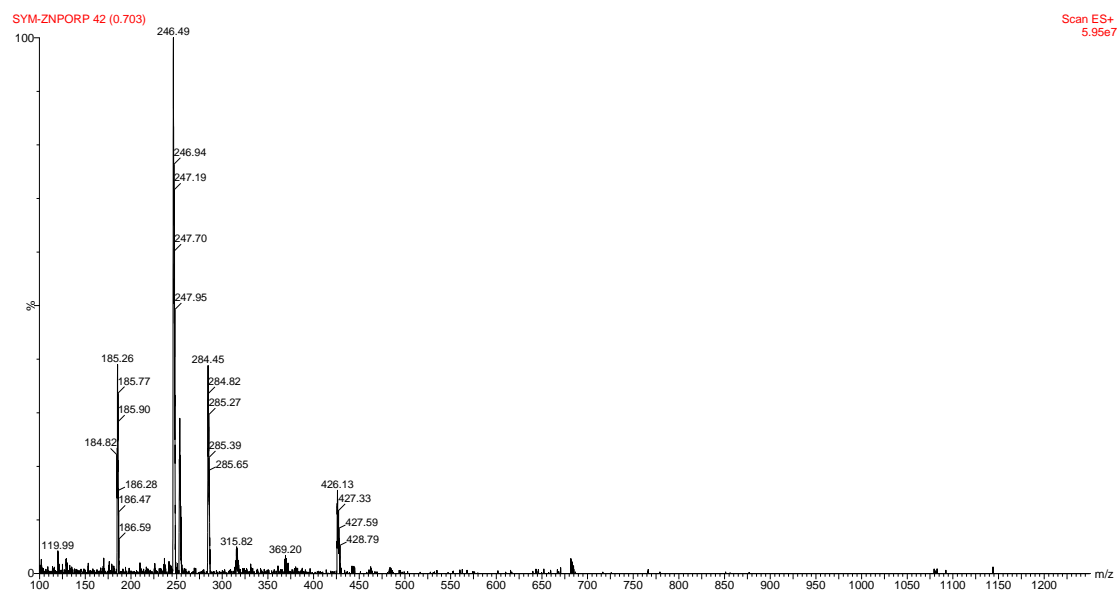


Figure S43. ESI-MS of sym-ZnPorph. $m/z = 246.49$ $[M-H]^{3+}$.

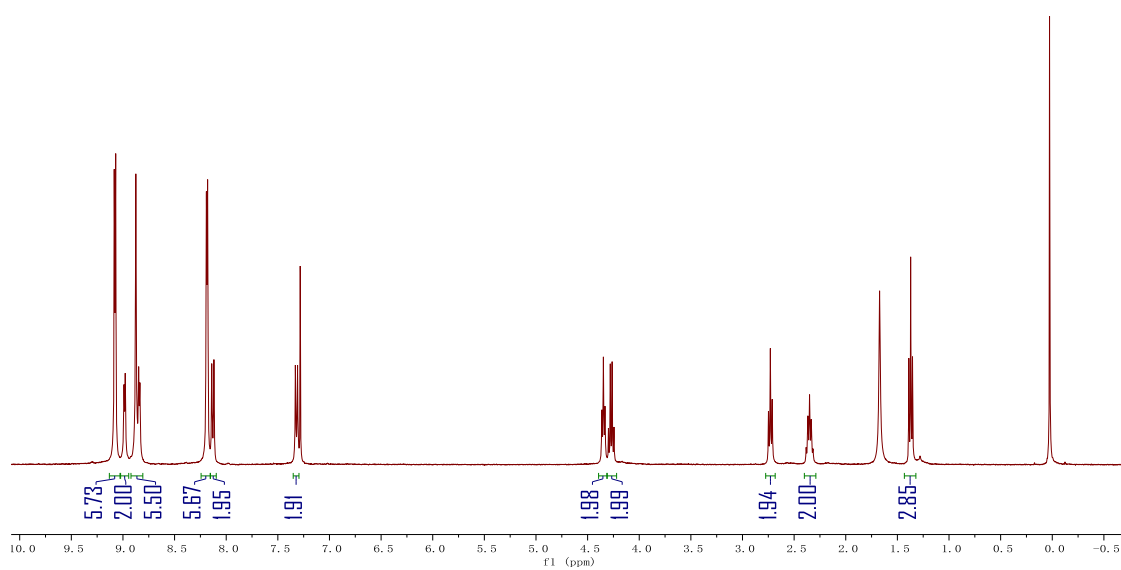


Figure S44. 1H NMR of porphyrin 7 in $CDCl_3$.

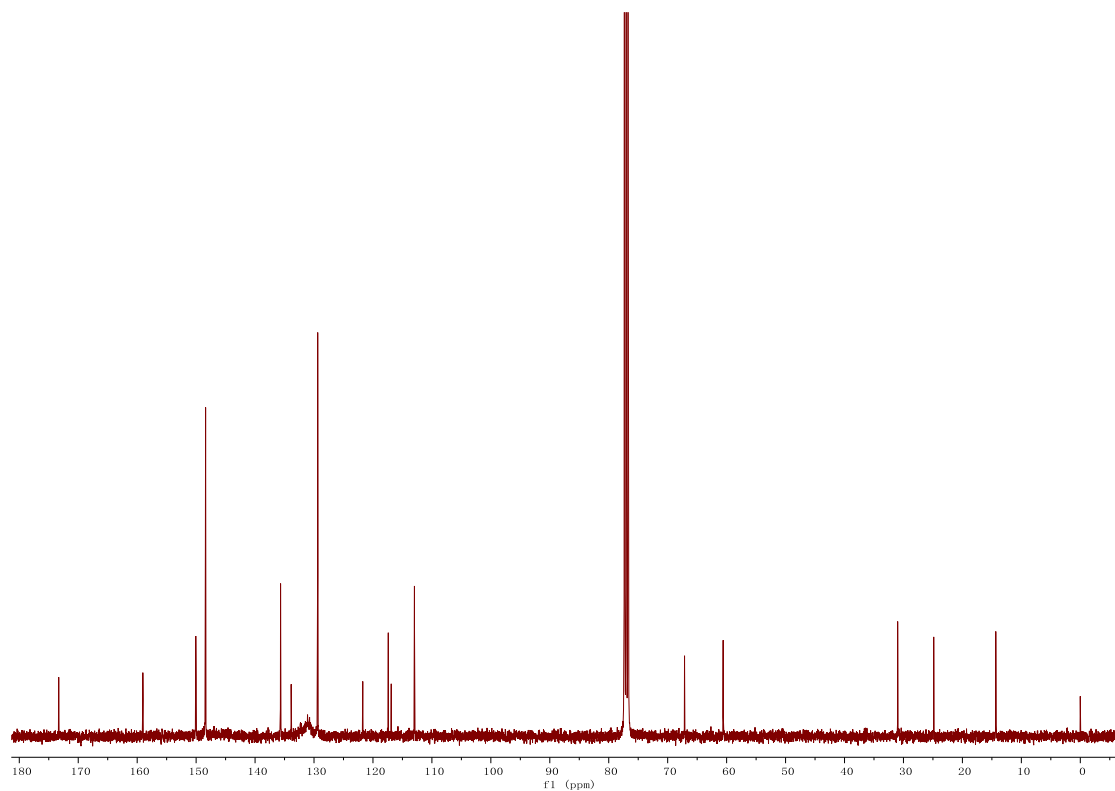


Figure S45. ^{13}C NMR of compound 7 in CDCl_3 .

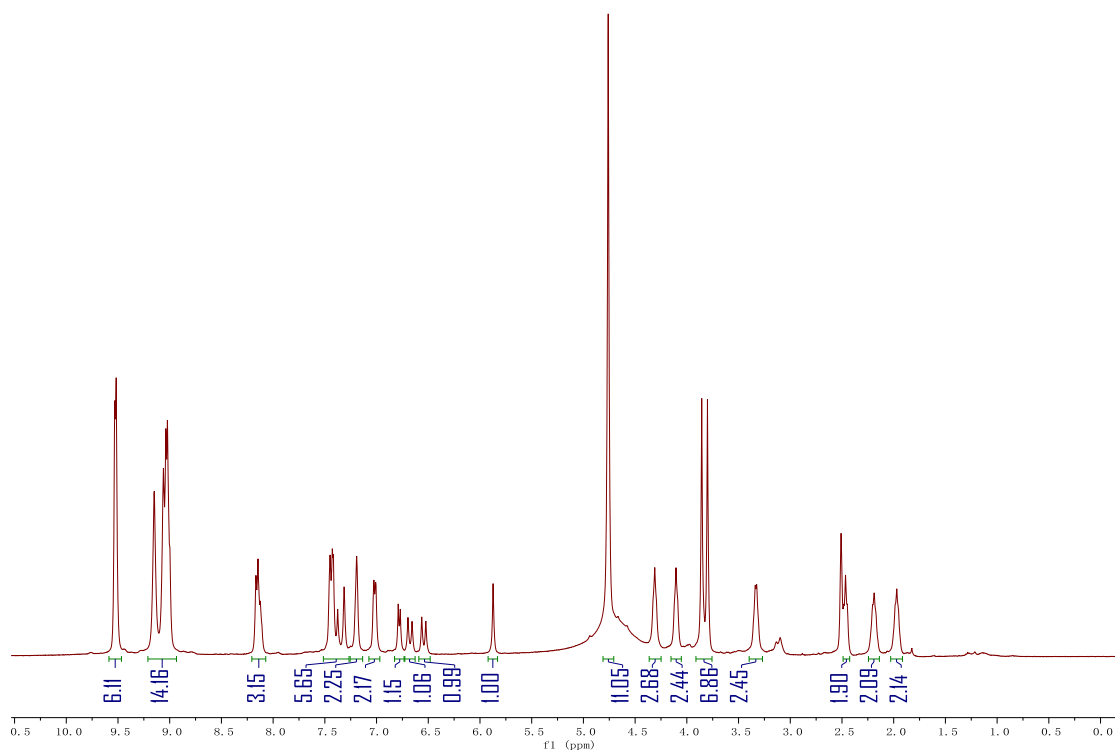


Figure S46. ^1H NMR of Prop-Cur in DMSO-d_6 .

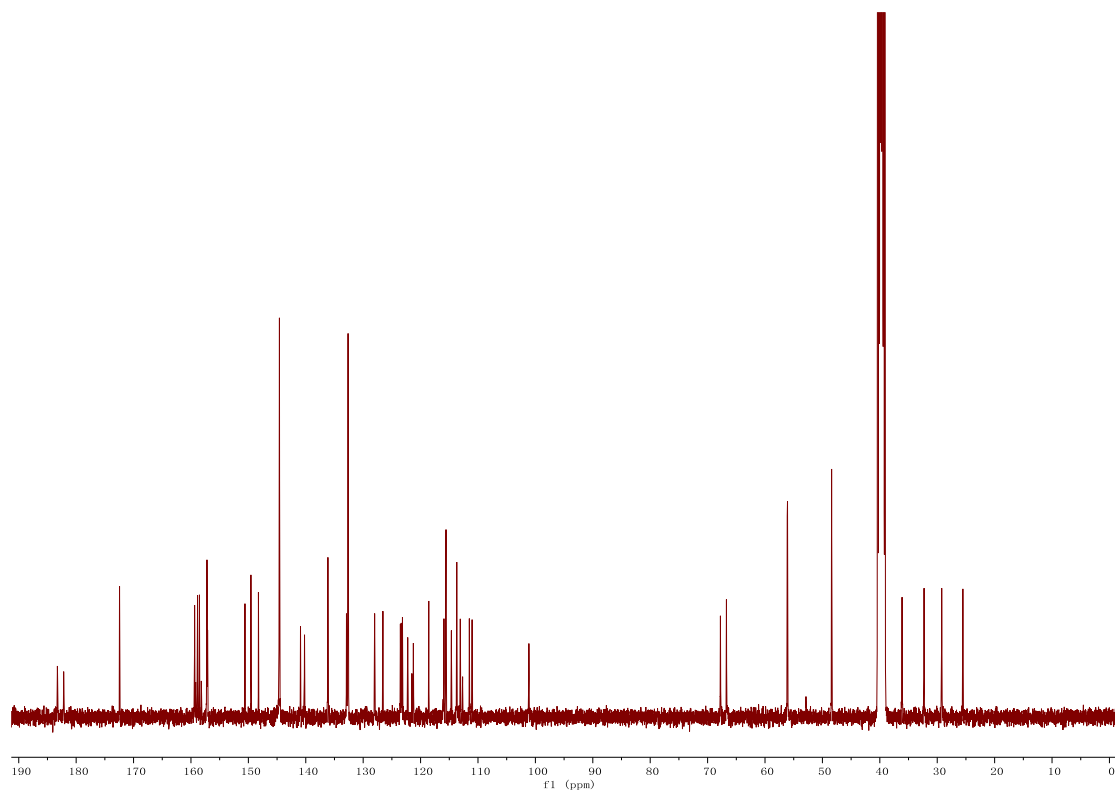


Figure S47. ^{13}C NMR of Prop-Cur in DMSO-d_6 .

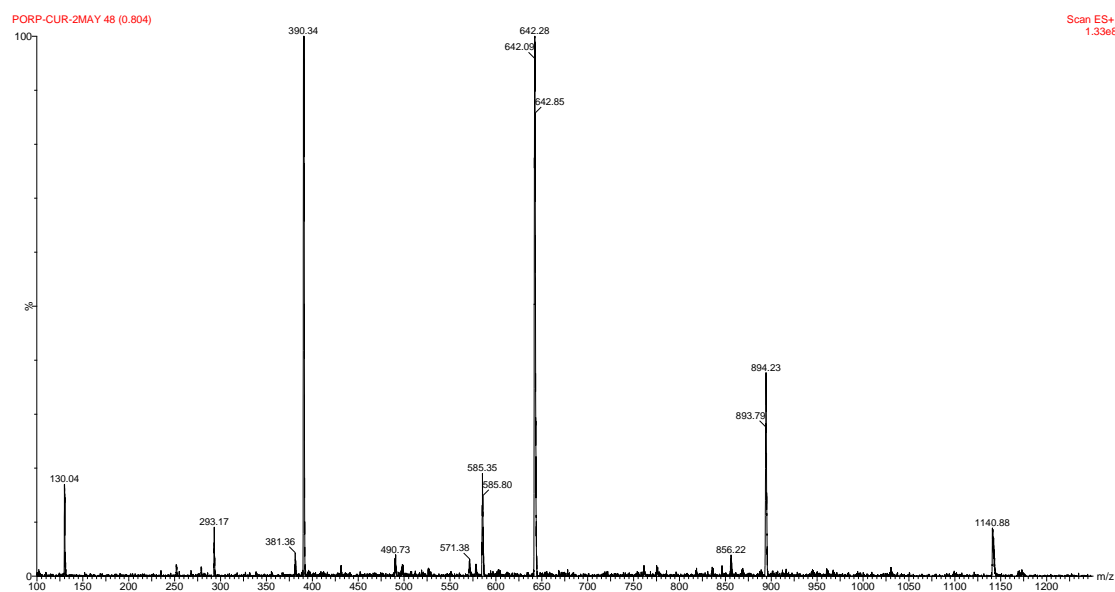


Figure S48. ESI-MS of prop-Cur. m/z : 390.34 $[\text{M}]^{3+}$, 642.28 $[\text{M}+\text{CF}_3\text{COO}]^{2+}$.

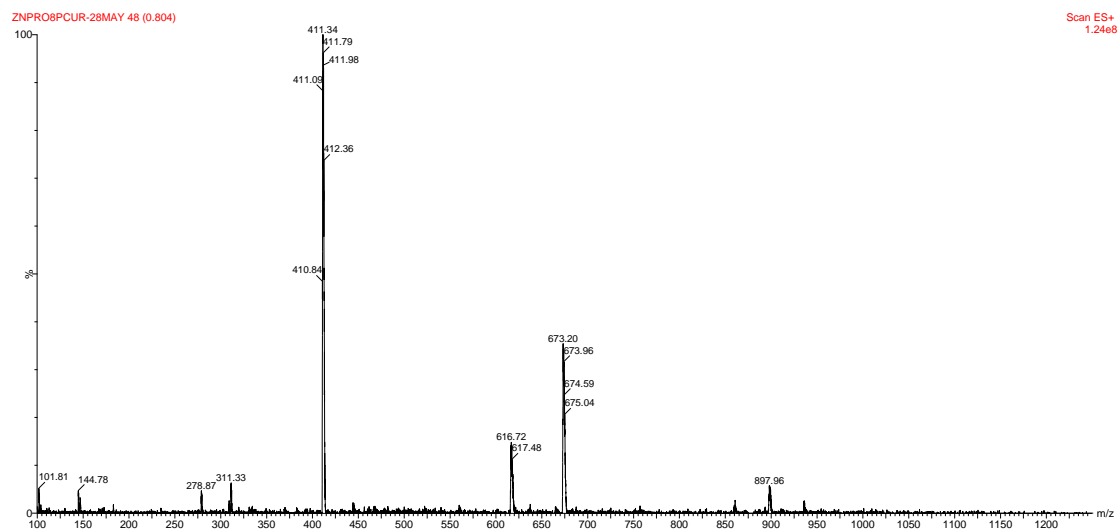


Figure S49. ESI-MS of ZnPorp-Cur. m/z : 411.34 $[M]^{3+}$, 673.20 $[M+CF_3COO]^{2+}$.

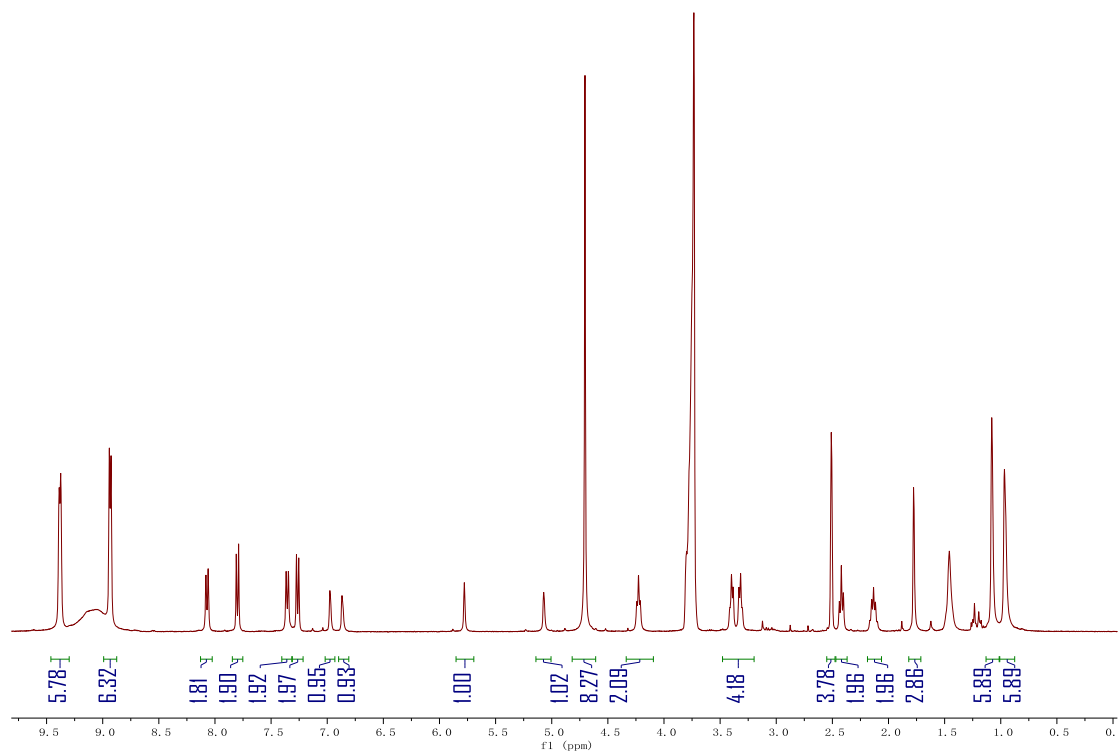


Figure S50. 1H NMR of Prop-Bex in $DMSO-d_6$.

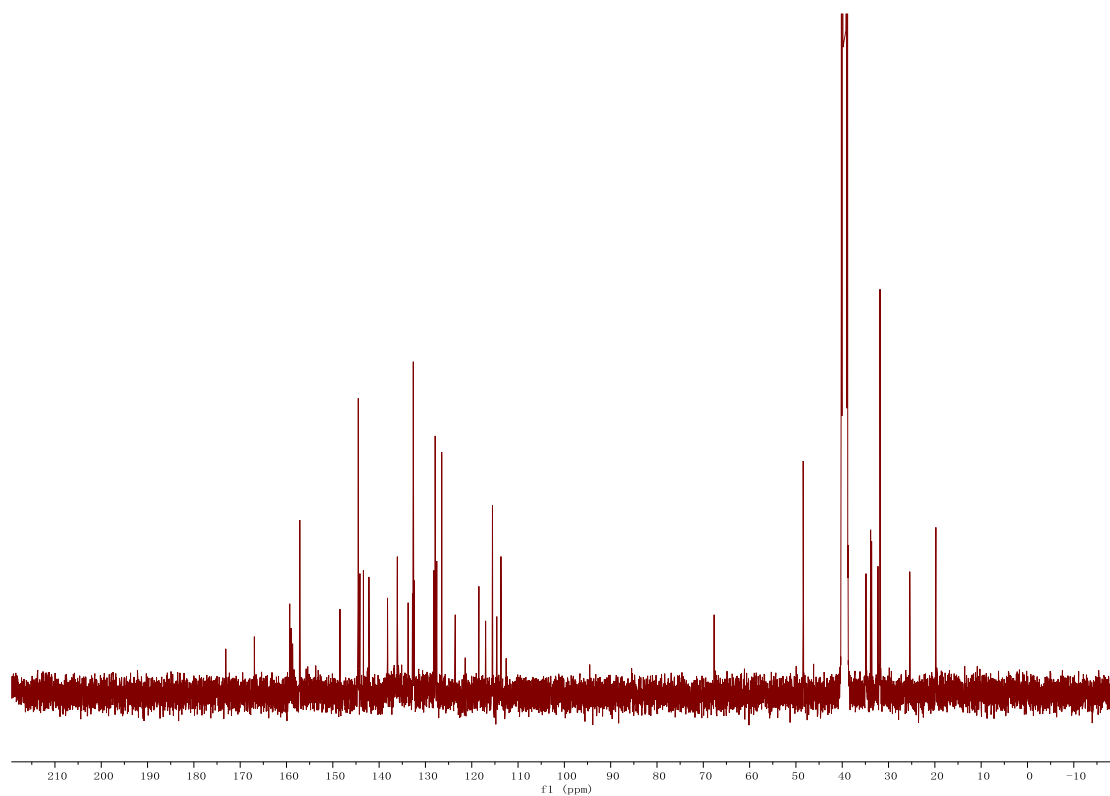


Figure S51. ^{13}C NMR of Prop-Bex in DMSO-d_6 .

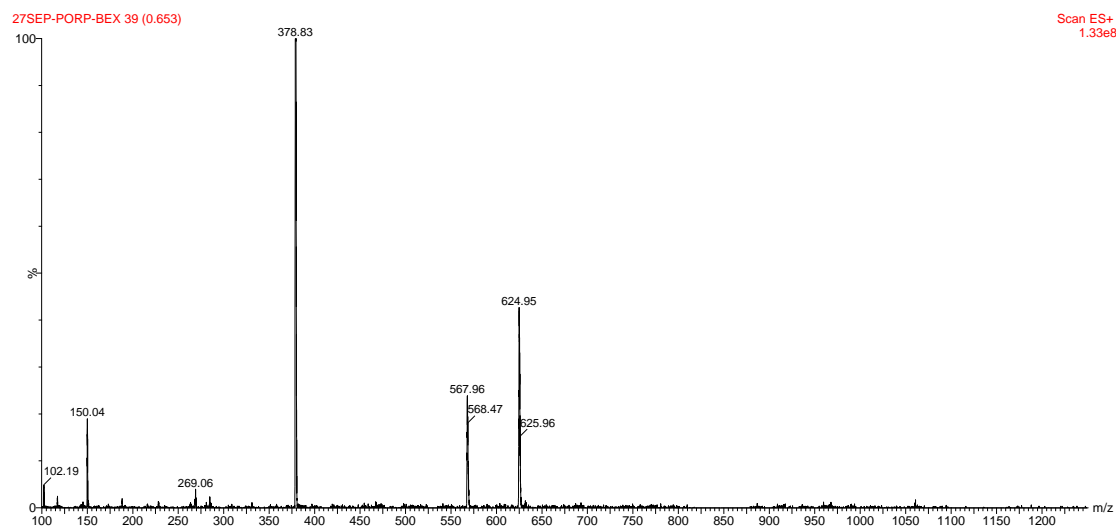


Figure S52. ESI-MS of Prop-Bex. m/z : 378.83 $[\text{M}]^{3+}$, 567.96 $[\text{M-H}]^{2+}$, 624.95 $[\text{M}+\text{CF}_3\text{COO}]^{2+}$.

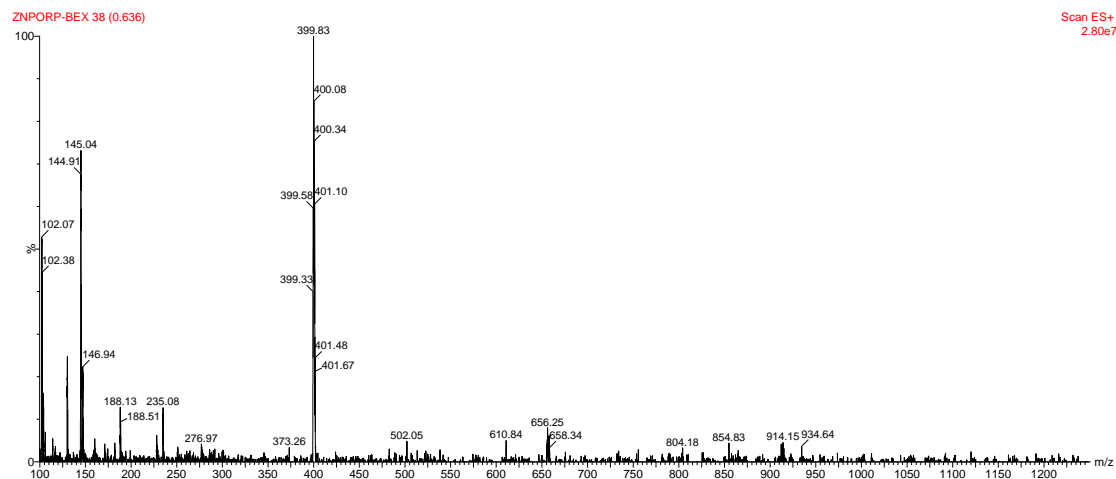


Figure S53. ESI-MS of ZnPorp-Bex. m/z : 399.83 $[M]^{3+}$.

Reference

1. Li, H.; Jensen, T. J.; Fronczek, F. R.; Vicente, M. G. H., Syntheses and Properties of a Series of Cationic Water-Soluble Phthalocyanines. *Journal of Medicinal Chemistry* **2008**, *51* (3), 502-511.
2. Schmitz, J.; Li, T.; Bartz, U.; Gütschow, M., Cathepsin B Inhibitors: Combining Dipeptide Nitriles with an Occluding Loop Recognition Element by Click Chemistry. *ACS Medicinal Chemistry Letters* **2016**, *7* (3), 211-216.
3. Uchiyama, S.; Tsuji, T.; Kawamoto, K.; Okano, K.; Fukatsu, E.; Noro, T.; Ikado, K.; Yamada, S.; Shibata, Y.; Hayashi, T.; Inada, N.; Kato, M.; Koizumi, H.; Tokuyama, H., A Cell-Targeted Non-Cytotoxic Fluorescent Nanogel Thermometer Created with an Imidazolium-Containing Cationic Radical Initiator. *Angewandte Chemie* **2018**, *130* (19), 5511-5515.