Electronic Supplementary Material (ESI) for New Journal of Chemistry.
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6-gingerol derived semisynthetic analogs mitigates oxidative stress, reverses acrylamide induced neurotoxicity in zebrafish.

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General experimental information:

All reactions were performed with the commercially available starting materials without any further purifications. 6-gingerol was isolated from raw ginger purchased from the local market (potheri). Proton nuclear magnetic resonance (^{1}H NMR) spectra were recorded on a Bruker BBFO (500 & 400 MHz) spectrometer. Chemical shifts were recorded in parts per million (ppm, δ) relative to chloroform (δ = 7.26, singlet). ^{1}H NMR splitting patterns are designated as singlet (s), doublet (d), triplet (t), quartet (q), doublet of doublets (dd), multiplet, (m), etc. Carbon nuclear magnetic resonance (^{13}C NMR) spectra were recorded on a Bruker BBFO (126 & 100 MHz) spectrometer. ^{13}C NMR data are reported with the solvent peak ($CDCl_3$ = 77.16) as the internal standard. High-resolution mass spectral analysis (HRMS) was performed on Bruker Impact HD mass spectrometer. Analytical thin-layer chromatography (TLC) was carried out on Merck 60 F254 pre-coated silica gel plate (0.2 mm thickness). Visualization was performed using a UV lamp.

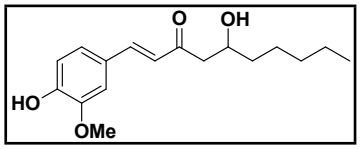
SC-XRD: The quality single crystals suitable for SC-XRD experiments of all the four compounds were obtained from acetonitrile (ACN) solvent by the slow evaporation method. The single-crystal X-ray diffraction measurements were performed to determine the crystal structure of compounds 1 and 2 at 273 K using APEX3 (Bruker, 2016; Bruker D8 Venture photon 100 CMOS detector) diffractometer having graphite-monochromatized (MoKα = 0.71073 Å). The X-ray generator was operated at 50 kV and 30 mA. A preliminary set of unit cell parameters and an orientation matrix were calculated from 36 frames, and the cell refinement was performed by SAINT-Plus (Bruker, 2016). An optimized strategy used for data collection consisted of different sets of φ and ω scans with 0.5º steps φ/ω. The data was collected with a time frame of 10 sec for the three components by setting the sample to detector distance fixed at 40 cm. The data points were corrected for Lorentzian, polarization, and absorption effects using SAINT-Plus and SADABS programs (Bruker, 2016). SHELXS-97 (Sheldrick, 2018) was used for structure solution and full-matrix least-squares refinement on F^2.[1] The program(s) used to refine the molecular structures of compounds 1-3 is SHELXL 2018/3 (Sheldrick, 2018). All non-hydrogen atoms were refined by the anisotropic method and hydrogen atoms were either refined or placed in calculated positions. The molecular graphics of ORTEP diagrams were performed by XP software. The crystal symmetry of the components was cross-checked by running the .cif file through PLATON (Spek, 2020) software and notified that no additional symmetry was observed.

Isolation of compound 1 and synthesis of compounds 3, 9 and 10.

Compound **1** was isolated from ginger and compounds **3, 9 and 10** were synthesized and reported by our group.[1]

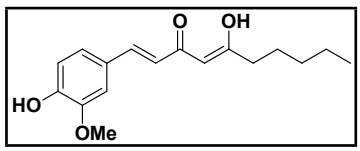
1.Manjunathan, T., Guru, A., Arokiaraj, J. and Gopinath, P., 2021. 6-Gingerol and Semisynthetic 6-Gingerdione Counteract Oxidative Stress Induced by ROS in Zebrafish. *Chemistry & Biodiversity*, *18*(12), p.e2100650.

Synthesis of compound 2



To a solution of 6-gingerol (500 mg, 1.7006 mmol, 1 eq) in 7 ml of THF at 0°C was stirred with DDQ (308 mg, 1.3605, 0.8 eq) was dissolved in 3 ml THF and added drop by drop then the solution was stirred for 30 min at 0°C, then the reaction mixture was warmed to room temperature and stirred until complete consumption of the starting material was observed (TLC) after 3 h. Then the reaction mixture was extracted with water and ethyl acetate (3 x 25 mL). The organic layers were combined, dried over anhydrous Na_2SO_4, and filtered. The solvent was evaporated under reduced pressure and the residue was purified over silica gel column chromatography (30%EtOAc/Hexane) afford desired product as a yellow syrup (330mg, 66 %). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.48 (d, *J* = 16.1 Hz, 1H), 7.12 – 7.04 (m, 1H), 7.03 (s, 1H), 6.94 – 6.87 (m, 1H), 6.56 (d, *J* = 16.1 Hz, 1H), 6.42 (s, 1H), 4.12-4.10 (m, 1H), 3.90 (s, 3H), 3.43 (s, 1H), 2.90 – 2.80 (m, 1H), 2.76-2.69 (m, 1H), 1.46-1.41 (m,2H), 1.39-1.24 (s, 6H), 0.88 (d, *J* = 6.4 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 201.16, 148.77, 147.14, 144.08, 126.71, 124.11, 123.83, 115.11, 109.74, 68.19, 56.04, 46.58, 36.62, 31.88, 31.00, 25.31, 22.71, 14.13.

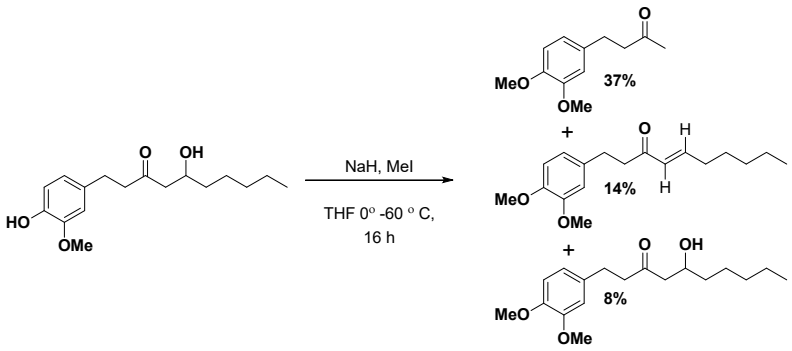
Synthesis of compound 4



To a solution of 6-gingerdione (100 mg, 0.3424 mmol, 1 eq) in 2 ml of THF at 0°C DDQ (63 mg, 0.2739 mmol, 0.8 eq) was dissolved in 1ml THF and added drop by drop then the solution was stirred for 30 min at 0°C, then the reaction mixture was warmed to room temperature and stirred until complete consumption of the starting material was observed (TLC) after 3 h. Then the reaction mixture was extracted with water and ethyl acetate (3 x 25 mL). The organic layers were combined, dried over anhydrous Na_2SO_4, and filtered. The solvent was evaporated under reduced pressure and the residue was purified over silica gel column chromatography (30%EtOAc/Hexanes) afford compound **4** as a yellow solid (33 mg, 33 %), mp 78 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 15.54 (bs, 1H), 7.52 (d, *J* = 15.8 Hz, 1H), 7.08 (dd,

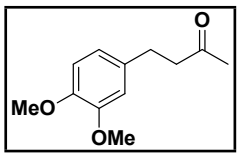
J = 2.2, 1.5 Hz, 1H), 7.01 (d, *J* = 1.4 Hz, 1H), 6.92 (d, *J* = 8.2 Hz, 1H), 6.34 (d, *J* = 15.8 Hz, 1H), 5.88 (s, 1H), 5.62 (s, 1H), 3.93 (s, 3H), 2.37 (t, *J* = 7.6 Hz, 2H), 1.67-1.63 (m, 2H), 1.36 – 1.24 (m, 4H), 0.90 (t, *J* = 6.7 Hz, 3H).^{13}C NMR (101 MHz, $CDCl_3$) δ 200.34, 178.20, 147.80, 146.92, 139.97, 127.87, 122.75, 120.71, 114.94, 109.61, 100.27, 56.08, 40.24, 31.61, 25.47, 22.59, 14.07. **HRMS (ESI)** Exact mass calcd. For $C_{17}H_{23}O_4$ $[M+H]^+$291.1591, found $[M+H]^+$291.1596.

Retro aldol reaction with O-alkylation

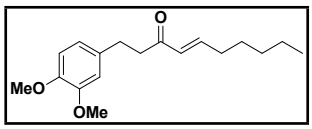


To a suspended solution of NaH in THF, 6-gingerol (1 eq) was added and stirred at at 0°C for 20 min. To this methyl iodide (1.5 eq) was added drop by drop then the solution allowed to stir 16 h at 60°C. Then the reaction mixture was extracted with water and ethyl acetate (3 x 100 mL). The organic layers were combined, dried over anhydrous Na_2SO_4, and filtered. The solvent was evaporated under reduced pressure and the residue was purified over silica gel column chromatography to obtain the products **5**, **6** and **7** in 37%, 14% and 8% yields respectively.

Compound 5



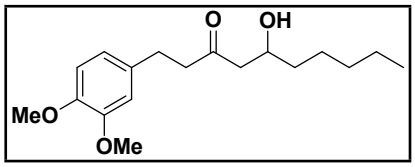
^{1}H NMR (500 MHz, $CDCl_3$) δ 6.76 (dd, *J* = 5.9, 2.7 Hz, 1H), 6.69 (s, 2H), 3.84 (s, 3H), 3.82 (s, 3H), 2.82 (t, *J* = 7.6 Hz, 2H), 2.72 (t, *J* = 7.4 Hz, 2H), 2.12 (s, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 208.26, 148.94, 147.44, 133.70, 120.14, 111.77, 111.37, 56.00, 55.90, 45.52, 30.23, 29.46. **(Yellow oil).**



Compound 6

^{1}H NMR (500 MHz, CDCl3) δ 6.84-6.72 (m, 4H), 6.09 (dt, *J* = 15.9, 1.4 Hz, 1H), 3.86 (s, 3H), 3.84 (s, 3H), 2.87 – 2.83 (m, 2H), 2.21-2.16 (m, 2H), 1.47-1.41 (m, 2H), 1.29-1.24 (m, 6H), 0.88 (t, *J* = 6.1 Hz, 3H).^{13}C NMR

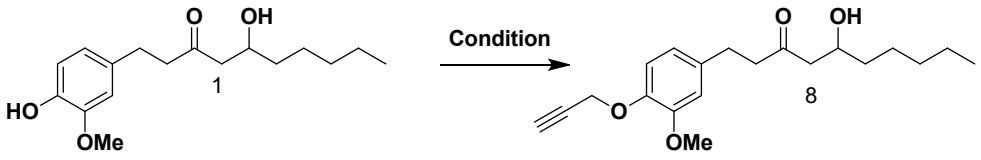
(126 MHz, $CDCl_3$) δ 201.16, 148.77, 147.14, 144.08, 126.71, 124.11, 123.83, 115.11, 109.74, 68.19, 56.04, 46.58, 36.62, 31.88, 31.00, 25.31, 22.71, 14.13. **(Yellow oil).**



Compound 7

Pale yellow solid mp (63 °C) ^{1}H NMR (500 MHz, $CDCl_3$) δ 6.78 (d, *J* = 8.7 Hz, 1H), 6.71 – 6.69 (m, 2H), 4.04-3.98 (s, 1H), 3.86 (s, 3H), 3.84 (s, 3H), 2.96 (bs, 1H), 2.85 (t, *J* = 7.5 Hz, 2H), 2.74 (t, *J* = 7.3 Hz, 2H), 2.58-2.46 (m, 2H), 1.32 – 1.27 (m, 8H), 0.88 (t, *J* = 5.4 Hz, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 211.5, 162.6, 148.9, 147.5, 133.4, 120.1, 111.7, 111.3, 67.6, 55.9, 49.4, 45.3, 36.4, 31.6, 29.2, 25.2, 22.5, 14.1.

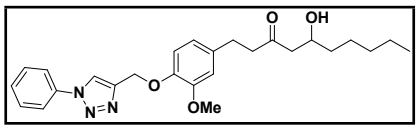
Propargylation of 6-gingerol[2]



K_2CO_3 (2 eq), Propargyl bromide (1.1 eq), DMF rt 5 h 84 %,

2. de Lima Silva, W.C., Conti, R., de Almeida, L.C., Morais, P.A., Borges, K.B., Júnior, V.L., Costa-Lotufo, L.V. and de Souza Borges, W., 2020. Novel [6]-gingerol triazole derivatives and their antiproliferative potential against tumor cells. *Current Topics in Medicinal Chemistry*, *20*(2), pp.161-169

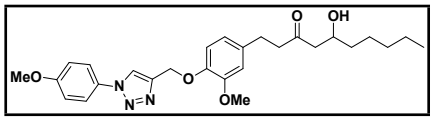
Compound 11



To a solution of **compound 8** (300 mg, 0.9036 mmol, 1 eq) in 7 ml of DMF to this $CuSO_4$ (112 mg, 0.4518 mmol, 0.5 eq), sodium ascorbate (179 mg, 0.9036 mmol, 1 eq) and phenyl azide (161 mg, 1.3554 mmol, 1.5 eq) was added and stirred at ambient condition till the consumption of starting material monitored by (TLC) for 30 min. To this saturated ammonium chloride (10 ml) was added and stirred for 10 min then the solution was extracted with EtOAc and concentrated under reduced pressure. Finally, the crude was subjected to column chromatography (50%EtOAc/Hexanes) afford compound **11** as a white solid (273 mg, 67%), mp 67-69 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.09 (s, 1H), 7.71 (d, *J* = 7.6 Hz, 2H), 7.51 (t, *J* = 7.7 Hz, 2H), 7.43 (t, *J* = 7.4 Hz, 1H), 6.98 (d, *J* = 8.1 Hz, 1H), 6.75 – 6.67 (m, 2H), 5.33 (s, 2H), 4.05 – 3.99 (m, 1H), 3.85 (s, 3H), 2.87 – 2.82 (m, 3H), 2.76 – 2.71(m, 2H), 2.59 – 2.45 (m, 2H), 1.38 – 1.24 (m, 8H), 0.86 (t, *J* = 6.7 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 211.39, 149.60, 145.97, 145.17, 137.03, 134.78, 129.88 (2C), 129.01, 121.29,

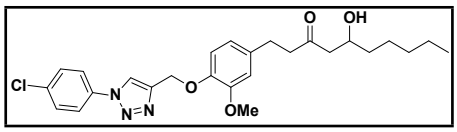
120.69 (2C), 120.26, 114.55, 112.27, 67.76, 63.29, 55.98, 49.44, 45.29, 36.55, 31.82, 29.22, 25.22, 22.69, 14.13. **HRMS (ESI)** Exact mass calcd. For $C_{26}H_{34}N_3O_4$ $[M+H]^+$452.2544, found $[M+H]^+$452.2537.

Synthesis of compound 12



To a solution of **compound 8** (200 mg, 0.6024 mmol, 1 eq) in 4 ml of DMF to this $CuSO_4$ (75 mg, 0.3012 mmol, 0.5 eq), sodium ascorbate (120 mg, 0.6024 mmol, 1 eq) and OMe-phenyl azide (135 mg, 0.9036 mmol, 1.5 eq) was added and stirred at ambient condition till the consumption of starting material monitored by (TLC) for 30 min. To this saturated ammonium chloride (10 ml) was added and stirred for 10 min then the solution was extracted with EtOAc and concentrated under reduced pressure. Finally, the crude was subjected to column chromatography (50%EtOAc/Hexanes) afford compound **12** as a pale brown solid (178 mg, 61%), mp 81-83 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.01 (s, 1H), 7.60 (d, *J* = 8.9 Hz, 2H), 7.01 (s, 1H), 6.98 (d, *J* = 8.2 Hz, 2H), 6.74 – 6.67 (m, 2H), 5.32 (s, 2H), 4.05 – 4.00 (m, 1H), 3.85 (s, 6H), 2.87 – 2.82 (m, 3H), 2.75 – 2.72 (m, 2H), 2.59 – 2.45 (m, 2H), 1.48 – 1.33 (m, 2H), 1.29 – 1.24 (m, 6H), 0.87 (t, *J* = 6.6 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 211.41, 160.04, 149.58, 145.99, 134.75, 130.45, 130.34, 122.35 (2C), 121.52, 120.26, 114.90 (2C), 114.53, 112.25, 67.76, 63.27, 55.98, 55.74, 49.45, 45.24, 36.55, 31.83, 29.23, 25.23, 22.69, 14.06. **HRMS (ESI)** Exact mass calcd. For $C_{27}H_{36}N_3O_5$ $[M+H]^+$ 482.2649, found $[M+H]^+$482.2646.

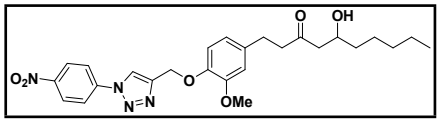
Synthesis of compound 13



To a solution of **compound 8** (200 mg, 0.6024 mmol, 1 eq) in 4 ml of DMF to this $CuSO_4$ (75 mg, 0.3012 mmol, 0.5 eq), sodium ascorbate (120 mg, 0.6024 mmol, 1 eq) and Cl-phenyl azide (140 mg, 0.9036 mmol, 1.5 eq) was added and stirred at ambient condition till the consumption of starting material monitored by (TLC) for 30 min. To this saturated ammonium chloride (10 ml) was added and stirred for 10 min then the solution was extracted with EtOAc and concentrated under reduced pressure. Finally, the crude was subjected to column chromatography (50%EtOAc/Hexanes) afford compound **13** as a yellow solid (145 mg, 50%), mp 75-77 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.06 (s, 1H), 7.67 (d, *J* = 8.7 Hz, 2H), 7.48 (d, *J* = 8.7 Hz, 2H), 6.96 (d, *J* = 8.1 Hz, 1H), 6.72 – 6.67 (m, 2H), 5.32 (s, 2H), 4.05 – 4.00 (m, 1H), 3.85 (s, 3H), 2.86 – 2.82 (m, 3H), 2.76 – 2.72 (m, 2H), 2.55 – 2.45

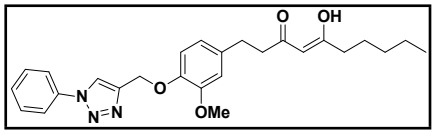
(m, 2H), 1.34 – 1.25 (m, 8H), 0.87 (t, J = 6.6 Hz, 3H).^{13}C NMR (101 MHz, $CDCl_3$) δ 211.38, 149.60, 145.95, 145.54, 135.55, 134.85, 134.76, 130.07 (2C), 121.84 (2C), 121.14, 120.27, 114.49, 112.28, 67.77, 63.28, 55.99, 49.45, 45.29, 36.56, 31.83, 29.23, 25.24, 22.70, 14.10. **HRMS (ESI)** Exact mass calcd. For $C_{26}H_{33}ClN_3O_4$ $[M+H]^+$486.2154, found $[M+H]^+$486.2153.

Synthesis of compound 14



To a solution of **compound 8** (200 mg, 0.6024 mmol, 1 eq) in 4 ml of DMF to this $CuSO_4$ (75 mg, 0.3012 mmol, 0.5 eq), sodium ascorbate (120 mg, 0.6024 mmol, 1 eq) and NO_2-phenyl azide (120 mg, 0.9036 mmol, 1.5 eq) was added and stirred at ambient condition till the consumption of starting material monitored by (TLC) for 30 min. To this saturated ammonium chloride (10 ml) was added and stirred for 10 min then the solution was extracted with EtOAc and concentrated under reduced pressure. Finally, the crude was subjected to column chromatography (50%EtOAc/Hexane) afford compound **14** as a yellow solid (133 mg, 45 %), mp 93-95 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.41 (d, J = 9.0 Hz, 2H), 8.20 (s, 1H), 7.97 (d, J = 9.0 Hz, 2H), 6.96 (d, J = 8.1 Hz, 1H), 6.77 – 6.69 (m, 2H), 5.34 (s, 2H), 4.04 – 4.00 (m, 1H), 3.87 (s, 3H), 2.88 – 2.83 (m, 3H), 2.76 – 2.73 (m, 2H), 2.59 – 2.45 (m, 2H), 1.48 – 1.38 (m, 2H), 1.29 – 1.25 (m, 6H), 0.88 – 0.86 (m, 3H). ^{13}C NMR (101 MHz, CDCl3) δ 211.23, 149.65, 147.35, 146.30, 145.85, 141.19, 135.10, 125.64(2C), 121.04, 120.61(2C), 120.29, 114.61, 112.38, 67.78, 63.25, 55.97, 49.47, 45.21, 36.59, 31.81, 29.21, 25.22, 22.67, 14.11. **HRMS (ESI)** Exact mass calcd. For $C_{26}H_{33}N_4O_6$ $[M+H]^+$497.2395, found $[M+H]^+$497.2391.

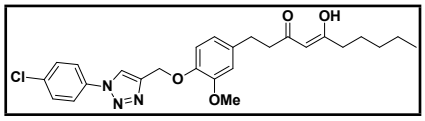
Synthesis of compound 15



To a solution of compound **11** (100 mg, 0.2217 mmol, 1 eq) in 5 ml of ethyl acetate were sequentially added DMP (188 mg, 0.4434 mmol, 2 eq). The reaction mixture was stirred for 20-30 min at RT. The reaction mixture was filtered and concentrated under reduced vacuum. The concentrated reaction mixture was applied to silica gel column chromatography (40%EtOAc/Hexanes) afford compound **15** (65 mg, 65 % as a pale white solid) after purification, mp 64-66 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 15.49 (s, 1H) enol, 8.10 (s, 1H), 7.72 (d, J = 7.6 Hz, 2H), 7.52 (t, J = 7.7 Hz, 2H), 7.44 (t, J = 7.4 Hz, 1H), 6.99 (d, J = 8.0 Hz, 1H), 6.74 – 6.68 (m, 2H), 5.46 (s, 1H), 5.35 (s, 2H), 3.85 (s, 3H), 3.53 (s, 1H), 2.90

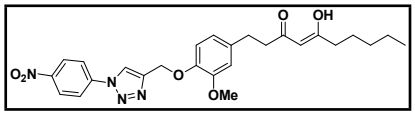
– 2.84 (m, 2H), 2.57 (t, J= 7.8 Hz, 2H), 2.25 (t, *J* = 7.8 Hz, 2H), 1.62 – 1.52 (m, 2H), 1.30 – 1.25 (m, 6H), 0.88 (t, *J* = 6.4 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 194.29, 193.65, 149.56, 145.98, 145.17, 137.01, 134.81, 129.91(2C), 129.09, 121.35, 120.72 (2C), 120.34, 114.51, 112.24, 99.53, 63.28, 55.97, 40.44, 38.30, 31.48, 31.31, 25.52, 22.50, 14.03. **HRMS (ESI)** Exact mass calcd. For $C_{26}H_{32}N_3O_4$ $[M+H]^+$450.2387, found $[M+H]^+$450.2387.

Synthesis of compound 16

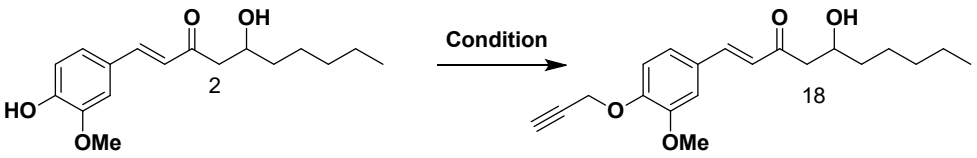


To a solution of compound **13** (100 mg, 0.2057 mmol, 1 eq) in 5 ml of ethyl acetate were sequentially added DMP (175 mg, 0.4115 mmol, 2 eq). The reaction mixture was stirred for 20-30 min at RT. The reaction mixture was filtered and concentrated under reduced vacuum. The concentrated reaction mixture was applied to silica gel column chromatography (40% EtOAc/Hexanes) afford compound **16** (52 mg, 52% as a yellow syrup) after purification. ^{1}H NMR (400 MHz, $CDCl_3$) δ 15.49 (s, 1H), 8.05 (s, 1H), 7.68 (d, *J* = 8.6 Hz, 2H), 7.50 (d, *J* = 8.7 Hz, 2H), 7.03 – 6.91 (m, 1H), 6.80 – 6.64 (m, 2H), 5.33 (s, 2H), 5.30 (s, 1H), 3.86 (s, 3H), 2.95 – 2.84 (m, 2H), 2.64 – 2.44 (m, 2H), 2.27-2.22 (m, 2H), 1.33 – 1.26 (m, 8H), 0.91 – 0.88 (m, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 194.26, 193.67, 149.59, 146.01, 145.60, 135.59, 134.85, 134.73, 130.06(2C), 121.83(2C), 121.09, 120.35, 114.48, 112.29, 99.51, 63.34, 55.98, 40.44, 38.29, 31.48, 31.30, 25.53, 22.49, 14.02. **HRMS (ESI)** Exact mass calcd. For $C_{26}H_{31}ClN_3O_4$ $[M+H]^+$484.1998, found $[M+H]^+$484.1666.

Synthesis of compound 17

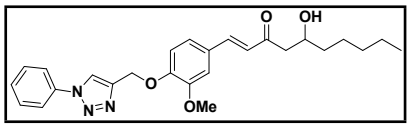


To a solution of **compound 14** (100 mg, 0.2016 mmol, 1 eq) in 5 ml of ethyl acetate were sequentially added DMP (171 mg, 0.4032 mmol, 2 eq). The reaction mixture was stirred for 20-30 min at RT. The reaction mixture was filtered and concentrated under reduced vacuum. The concentrated reaction mixture was applied to silica gel column chromatography (40%EtOAc/Hexanes) afford compound **17** (32 mg, 32% as a yellow solid) after purification, mp 83-85 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ **15.57 (s, 1H)enol**, 8.41 (d, *J* = 9.0 Hz, 2H), 8.20 (s, 1H), 7.97 (d, *J* = 9.0 Hz, 2H), 6.97 (d, *J* = 8.1 Hz, 1H), 6.75 – 6.71(m, 2H), 5.46 (s, 1H), 5.35 (s, 2H), 3.86 (s, 3H), 2.90 – 2.84 (m, 2H), 2.58 (t, *J* = 8 Hz, 2H), 2.25 (t, *J* = 8 Hz, 2H), 1.60 – 1.54 (m, 2H), 1.33 – 1.25 (m, 6H), 0.89 (t, *J* = 6.6 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 194.20, 193.72, 149.60, 147.40, 146.38, 145.86, 141.22, 135.08, 125.70 (2C), 121.01, 120.65 (2C), 120.37, 114.48, 112.37, 112.31, 99.52, 63.27, 55.99, 40.45, 38.28, 31.50, 31.30, 25.55, 22.51, 14.05.



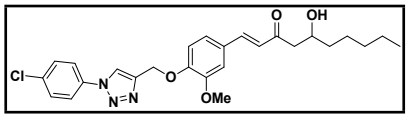
K_2CO_3 (2 eq), Propargyl bromide (1.1 eq), DMF rt 5 h 84 %,

Synthesis of compound 19



To a solution of **compound 18** (400 mg, 1.2121 mmol, 1 eq) in 7 ml of DMF to this $CuSO_4$ (151 mg, 0.6060 mmol, 0.5 eq), sodium ascorbate (240 mg, 1.2121 mmol, 1 eq) and phenyl azide (217 mg, 1.8181 mmol, 1.5 eq) was added and stirred at ambient condition till the consumption of starting material monitored by (TLC) for 30 min. To this saturated ammonium chloride (10 ml) was added and stirred for 10 min then the solution was extracted with EtOAc and concentrated under reduced pressure. Finally, the crude was subjected to column chromatography (40%EtOAc/Hexanes) afford compound **19** as a white solid (247 mg, 45 %), mp108-110 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.09 (s, 1H), 7.72 (d, *J* = 7.7 Hz, 2H), 7.54-7.49 (m, 3H), 7.47-7.43 (m, 1H), 7.13 (s, 2H), 7.09 (s, 1H), 6.61 (d, *J* = 16.1 Hz, 1H), 5.42 (s, 2H), 4.15 – 4.10 (m, 1H), 3.92 (s, 3H), 3.25 (s, 1H), 2.90-2.71 (m, 2H), 1.33-1.30 (m, 3H), 1.28-1.25 (m, 5H), 0.90 (t, *J* = 6.9 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 201.06, 150.22, 149.87, 144.55, 143.55, 137.08, 129.95, 129.12, 128.17(2C), 124.99, 123.20, 121.42, 120.78(2C), 113.78, 110.51, 68.14, 56.13, 46.80, 36.72, 31.96, 29.85, 25.38, 22.78, 14.19. **HRMS (ESI)** Exact mass calcd. For $C_{26}H_{32}N_3O_4$ $[M+H]^+$450.2387, found $[M+H]^+$450.2387.

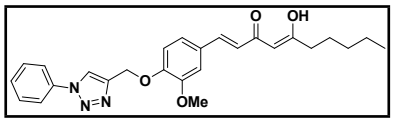
Synthesis of compound 20



To a solution of **compound 18** (200 mg, 0.6060 mmol, 1 eq) in 4 ml of DMF to this $CuSO_4$ (79 mg, 0.3030 mmol, 0.5 eq), sodium ascorbate (120 mg, 0.6060 mmol, 1 eq) and Cl-phenyl azide (140 mg, 0.9090 mmol, 1.5 eq) was added and stirred at ambient condition till the consumption of starting material monitored by (TLC) for 30 min. To this saturated ammonium chloride (10 ml) was added and stirred for 10 min then the solution was extracted with EtOAc and concentrated under reduced pressure. Finally, the crude was subjected to column chromatography (40%EtOAc/Hexanes) afford compound **20** as a pale-yellow solid (47 mg, 16 %), mp 114-116 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.42 (d, *J* = 9.1 Hz, 2H), 8.21 (s, 1H), 7.97 (d, *J* = 9.0

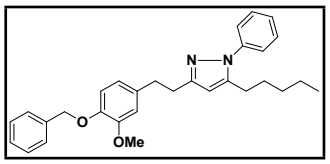
Hz, 2H), 7.51 (d, *J* = 16.1 Hz, 1H), 7.15-7.10 (m, 3H), 6.62 (d, *J* = 16.1 Hz, 1H), 5.43 (s, 2H), 4.15- 4.10 (m, 1H), 3.92 (s, 3H), 3.21 (s, 1H), 2.89 – 2.71 (m, 2H), 1.33 – 1.25 (m, 8H), 0.90 (t, *J* = 6.91 Hz, 3H).^{13}C NMR (101 MHz, $CDCl_3$) δ 200.95, 149.92, 149.84, 147.48, 145.53, 143.32, 141.11, 128.39(2C), 125.68, 125.09, 123.06, 121.24, 120.69(2C), 113.68, 110.53, 68.10, 62.83, 56.08, 46.86, 36.70, 31.91, 25.33, 22.73, 14.15.

Synthesis of compound 21



To a solution of **compound 19** (150 mg, 0.3325 mmol, 1 eq) in 7 ml of ethyl acetate were sequentially added DMP (282 mg, 0.6651 mmol, 2 eq). The reaction mixture was stirred for 20-30 min at RT. The reaction mixture was filtered and concentrated under reduced vacuum. The concentrated reaction mixture was applied to silica gel column chromatography (40%EtOAc/Hexanes) afford compound **21** (107 mg, 72 % as a white solid) after purification, mp 88-90 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ **15.51 (s, 1H)enol**, 8.09 (s, 1H), 7.72 (d, *J* = 7.8 Hz, 2H), 7.55 – 7.51 (m, 3H), 7.47-7.43 (m, 1H), 7.11 (s, 2H), 7.06 (s, 1H), 6.36 (d, *J* = 15.8 Hz, 1H), **5.63 (s, 1H)**, 5.41 (s, 2H), 3.92 (s, 3H), **3.86 (s, 1H),** 2.44 – 2.33 (m, 2H), 1.35 – 1.26 (m, 8H), 0.91 (t, *J* = 6.2 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 200.64, 171.80, 149.83, 149.43, 147.73, 139.53, 137.11, 129.94, 129.22, 129.08(2C), 122.10, 121.48, 121.38, 120.79(2C), 113.91, 110.53, 100.45, 63.07, 56.11, 31.60, 29.85, 25.42, 22.60, 14.03.

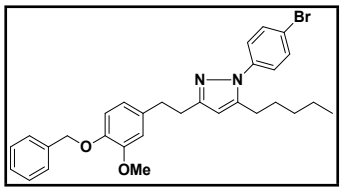
Synthesis of compound 22



To a solution of **compound 9** (150 mg, 0.3926 mmol, 1 eq) in 4 ml of ethanol to this phenyl hydrazine (47 µl, 0.4318 mmol, 1.1 eq) were added and the reaction mixture was stirred until complete consumption of the starting material was observed (TLC) for 12 h at 80°C. Then the reaction mixture was concentrated under reduced pressure. Finally, the crude was subjected to column chromatography (5%EtOAc/Hexanes) afford compound **22** as a white solid **(**60 mg, **34 %)**, mp 68-70 °C; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.43 – 7.41 (m, 2H), 7.40 – 7.38 (m, 2H), 7.37-7.35 (m, 2H), 7.34 – 7.33 (m, 1H), 7.32 (d, *J* = 1.25 Hz, 1H), 7.30-7.29 (m, 2H), 6.75 (d, *J* = 8.1 Hz, 1H), 6.59 – 6.52 (m, 2H), 6.05 (s, 1H), 5.11 (s, 2H), 3.81 (s, 3H), 2.95 – 2.87 (m, 2H), 2.81 - 2.78 (m, 2H), 2.66 – 2.61 (m, 2H), 1.71 - 1.65 (m, 2H), 1.41 – 1.34 (m, 4H), 0.91 (t, *J* = 7.0 Hz, 3H) ^{13}C NMR (126 MHz, $CDCl_3$) δ 153.95, 149.64, 146.72, 143.45, 140.04, 137.43, 134.15, 129.11, 128.65(2C), 127.91, 127.63(2C), 127.35 (2C), 125.55 (2C), 120.31,

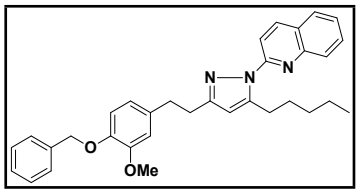
114.25, 112.32, 104.56, 71.26, 56.05, 35.01, 31.88, 29.64, 28.63, 28.46, 22.66, 14.22. **HRMS (ESI)** Exact mass calcd. For $C_{30}H_{35}N_2O_2$ $[M+H]^+$455.2693, found $[M+H]^+$455.2691.

Synthesis of compound 23



To a solution of **compound 9** (150 mg, 0.3926 mmol, 1 eq) in 4 ml of ethanol to this Br-phenyl hydrazine hydrochloride (97 mg, 0.4318 mmol, 1.1 eq) and to this triethyl amine (60 µl, 0.4318 mmol, 1.1 eq) was added and the reaction mixture was stirred until complete consumption of the starting material was observed (TLC) for 12 h at 80°C. Then the reaction mixture was concentrated under reduced pressure. Finally, the crude was subjected to column chromatography (5%EtOAc/Hexanes) afford compound **23** as a brown liquid (73 mg, 35 %). ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.58 – 7.57 (m, 2H), 7.44 (dd, *J* = 1.35, 0.5 Hz, 2H), 7.40 – 7.34 (m, 2H), 7.32 – 7.27 (m, 3H), 6.82 – 6.78 (m, 2H), 6.71 (dd, *J* = 2.0, 2.0 Hz, 1H), 6.01 (s, 1H), 5.13 (s, 2H), 3.86 (s, 3H), 2.92 (s, 4H), 2.62 – 2.54 (m, 2H), 1.60-1.54 (m, 2H), 1.26 (s, 4H), 0.87 (t, *J* = 6.5 Hz, 3H).^{13}C NMR (101 MHz, $CDCl_3$) δ 153.32, 149.63, 146.54, 144.71, 139.19, 137.58, 135.36, 132.30 (2C), 128.63 (2C), 127.87, 127.40 (2C), 126.86, 121.25, 120.37 (2C), 114.32, 112.50, 105.01, 71.34, 56.06, 35.75, 31.57, 30.63, 28.59, 26.44, 22.45, 14.06. **HRMS (ESI)** Exact mass calcd. For $C_{30}H_{34}BrN_2O_2$ $[M+H]^+$533.1793, found $[M+H]^+$533.1778

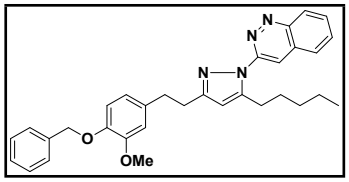
Synthesis of compound 24



To a solution of **compound 9** (300 mg, 0.785 mmol, 1 eq) in 4 ml of ethanol to this phenyl hydrazine (138 mg, 0.863 mmol, 1.1 eq) were added and the reaction mixture was stirred until complete consumption of the starting material was observed (TLC) for 12 h at 80°C. Then the reaction mixture was concentrated under reduced pressure. Finally, the crude was subjected to column chromatography (5%EtOAc/Hexanes) afford compound **24** as a yellow solid (80 mg, 20%), mp 92-94 °C; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.22 (d, *J* = 8.8 Hz, 1H), 8.10 (d, *J* = 8.8 Hz, 1H), 7.97 (d, *J* = 8.4 Hz, 1H), 7.82 (d, *J* = 8.0 Hz, 1H), 7.71 (t, *J* = 7.6 Hz, 1H), 7.51 (t, *J* = 7.5 Hz, 1H), 7.45 (d, *J* = 7.4 Hz, 2H), 7.37 (t, *J* = 7.4 Hz, 2H), 7.30 (t, *J* = 7.2 Hz, 1H), 6.82 (d, *J* = 2.0 Hz, 2H), 6.75 (d, *J* = 8.0 Hz, 1H), 6.07 (s, 1H), 5.14 (s, 2H), 3.88 (s, 3H), 3.28 (t, *J* = 7.7 Hz, 2H), 2.98 (s, 4H), 1.78-1.72 (m, 2H), 1.43 – 1.38 (m, 2H), 1.28 (d, *J* = 14.9 Hz, 2H), 0.91 (t, *J* = 7.0 Hz, 3H). ^{13}C NMR (126

MHz, $CDCl_3$) δ 153.90, 152.39, 149.59, 147.22, 146.49, 146.47, 138.33, 137.55, 135.28, 129.96, 128.67, 128.61, 127.84, 127.65 (2C), 127.38 (2C), 126.47, 125.95, 120.38, 115.58, 114.26, 112.46, 107.56, 71.28, 56.03, 35.44, 31.88, 30.72, 28.85, 28.47, 22.64, 14.23. **HRMS (ESI)** Exact mass calcd. For $C_{33}H_{36}N_3O_2$ $[M+H]^+$506.280, found $[M+H]^+$506.279.

Synthesis of compound 25



To a solution of **compound 9** (150 mg, 0.3926 mmol, 1 eq) in 4 ml of ethanol to this 1-hydrazino phthalazine (85 mg, 0.4319 mmol, 1.1 eq) and triethylamine (60 µl, 0.4319 mmol, 1.1 eq) was added and the reaction mixture was stirred until complete consumption of the starting material was observed (TLC) for 12 h at 80°C. Then the reaction mixture was concentrated under reduced pressure. Finally, the crude was subjected to column chromatography (25%EtOAc/Hexanes) afford compound **25** as a yellow solid (48 mg, 24 %), mp 144-146 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 10.09 (s, 1H), 8.35 - 8.32 (m, 1H), 7.74 (s, 1H), 7.64 – 7.58 (m, 2H), 7.49 – 7.42 (m, 3H), 7.36 (t, *J* = 7.4 Hz, 2H), 7.30 (d, *J* = 7.3 Hz, 1H), 6.85 - 6.81 (m, 2H), 6.73 (dd, *J* = 1.8, 1.8 Hz, 1H), 5.14 (s, 2H), 3.90 (s, 3H), 2.97 – 2.86 (m, 2H), 2.72 – 2.63 (m, 2H), 2.18 (s, 2H), 1.43 – 1.33 (m, 4H), 1.29 (s, 2H), 1.26 (s, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 164.37, 149.80, 147.22, 146.64, 146.51, 137.62, 137.53, 135.30, 131.73, 131.51, 128.62 (2C), 127.85, 127.45, 127.40, 127.31 (2C), 126.06, 124.60, 124.07, 120.26, 114.52, 12.46, 71.65, 56.16, 40.98, 32.60, 31.65, 31.49, 30.28, 29.84, 17.18.

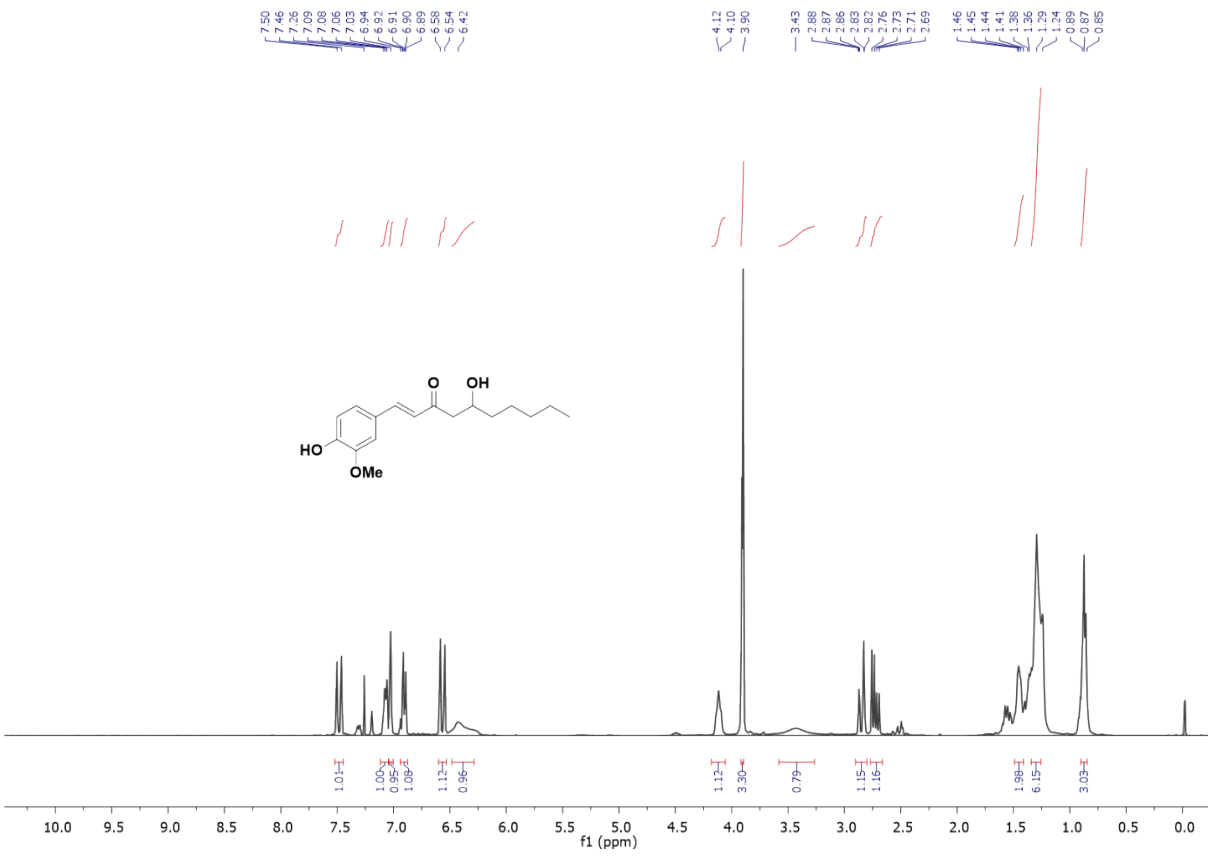


Fig.S1. ^{1}H NMR of compound 2

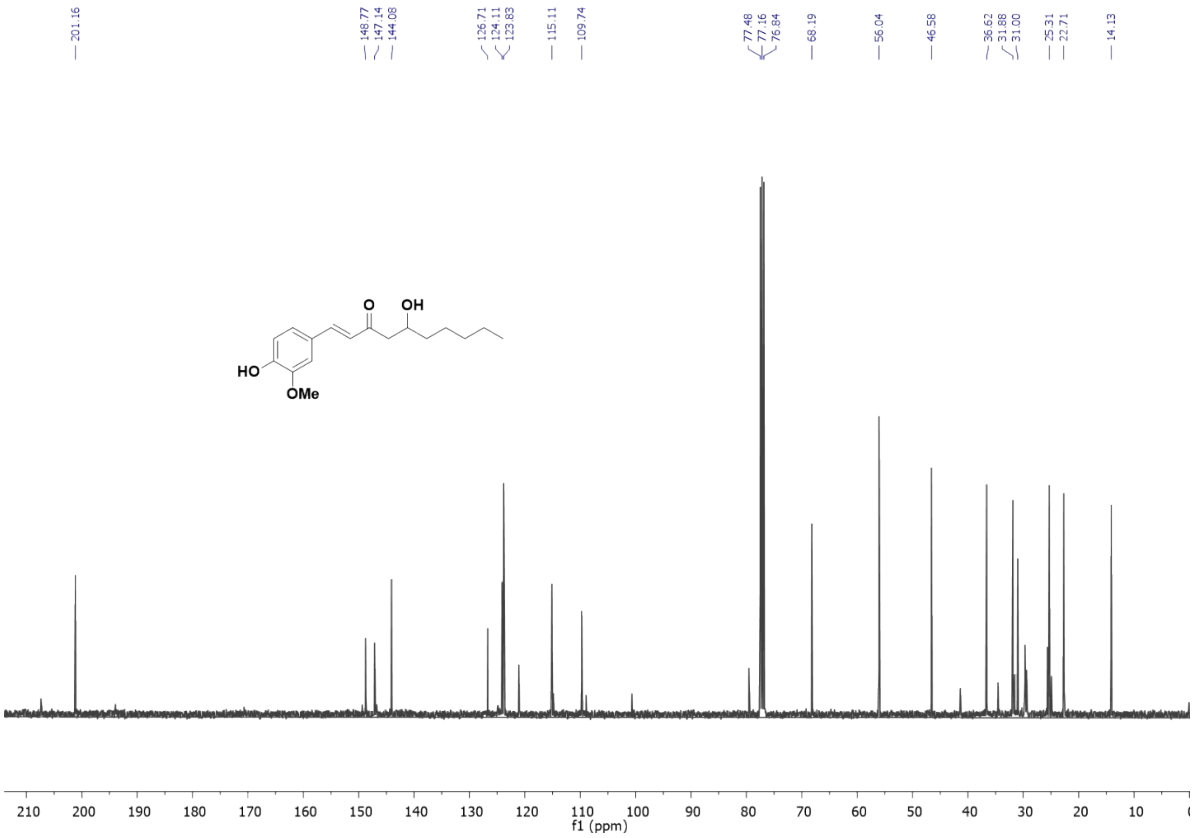


Fig.S2. ^{13}C NMR of compound 2

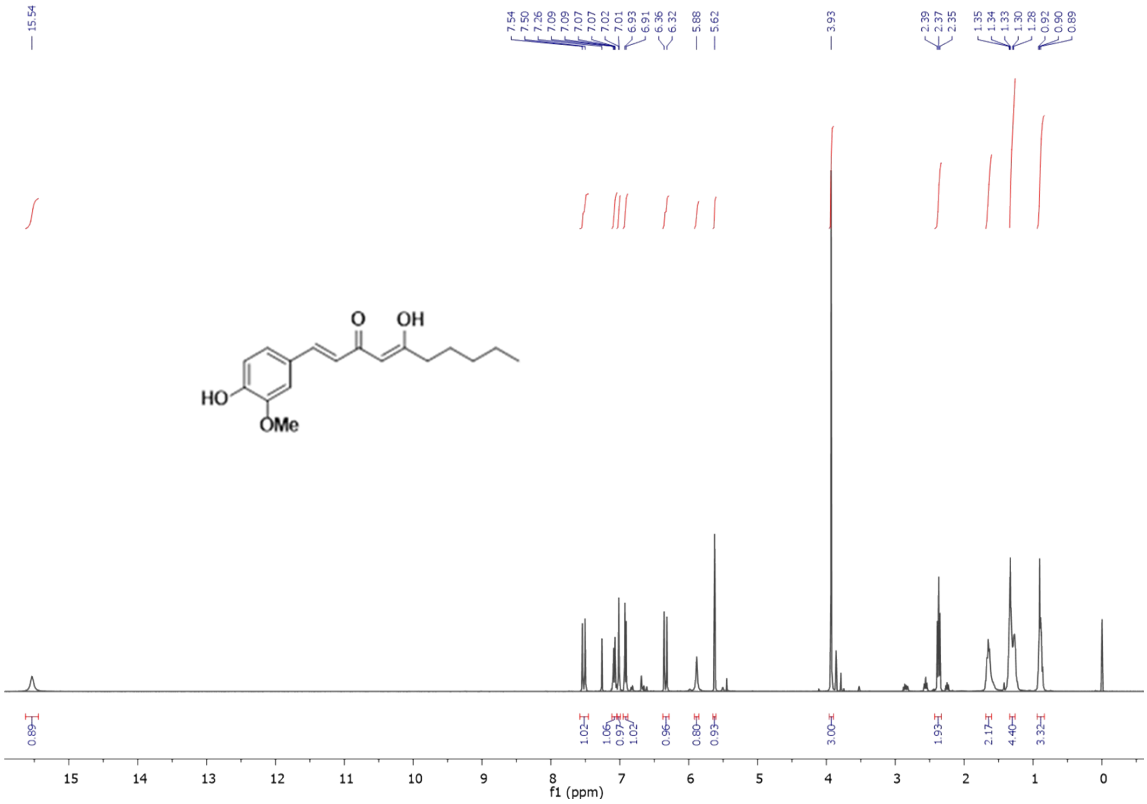


Fig.S3. ^{1}H NMR of compound 4

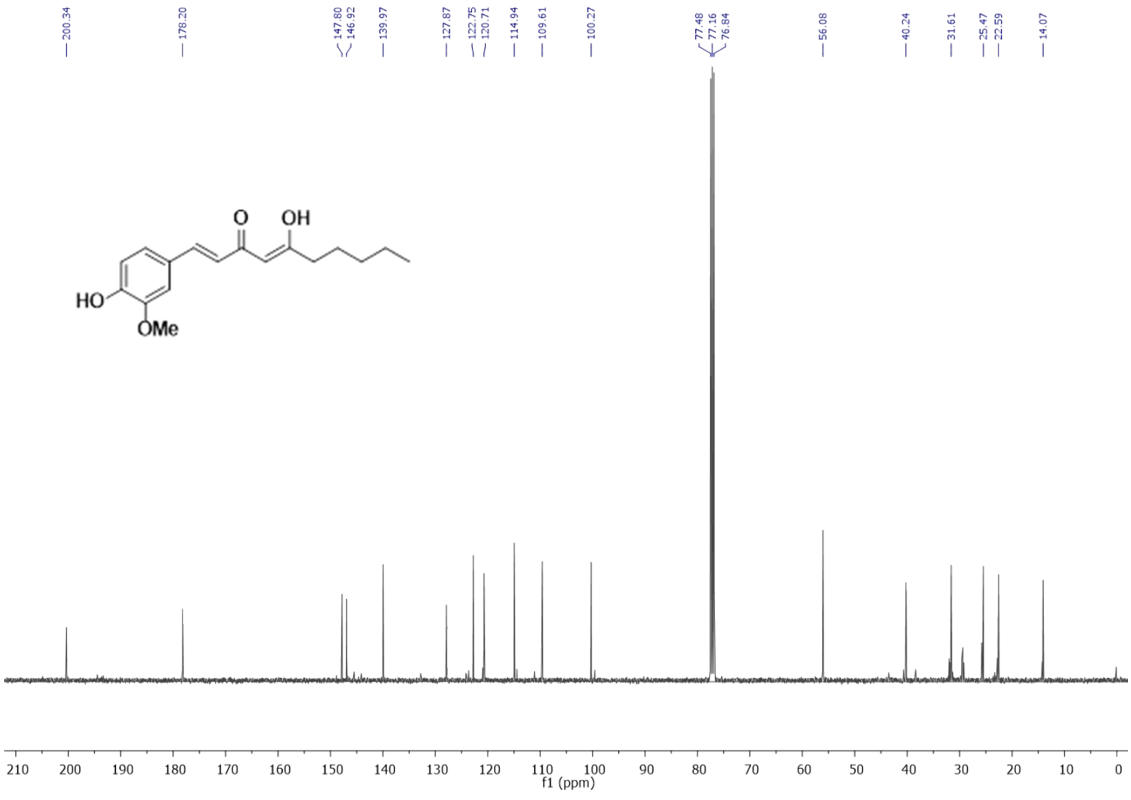


Fig.S4. ^{13}C NMR of compound 4

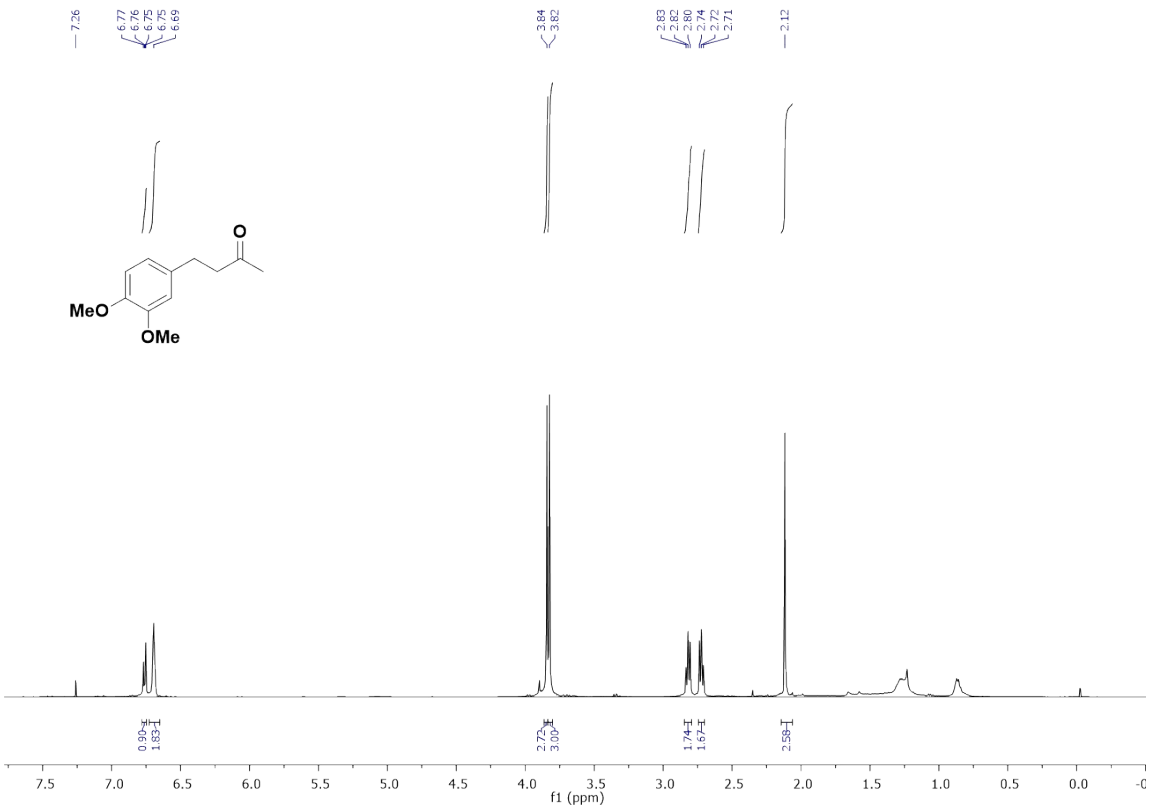


Fig.S5. ^{1}H NMR of compound 5

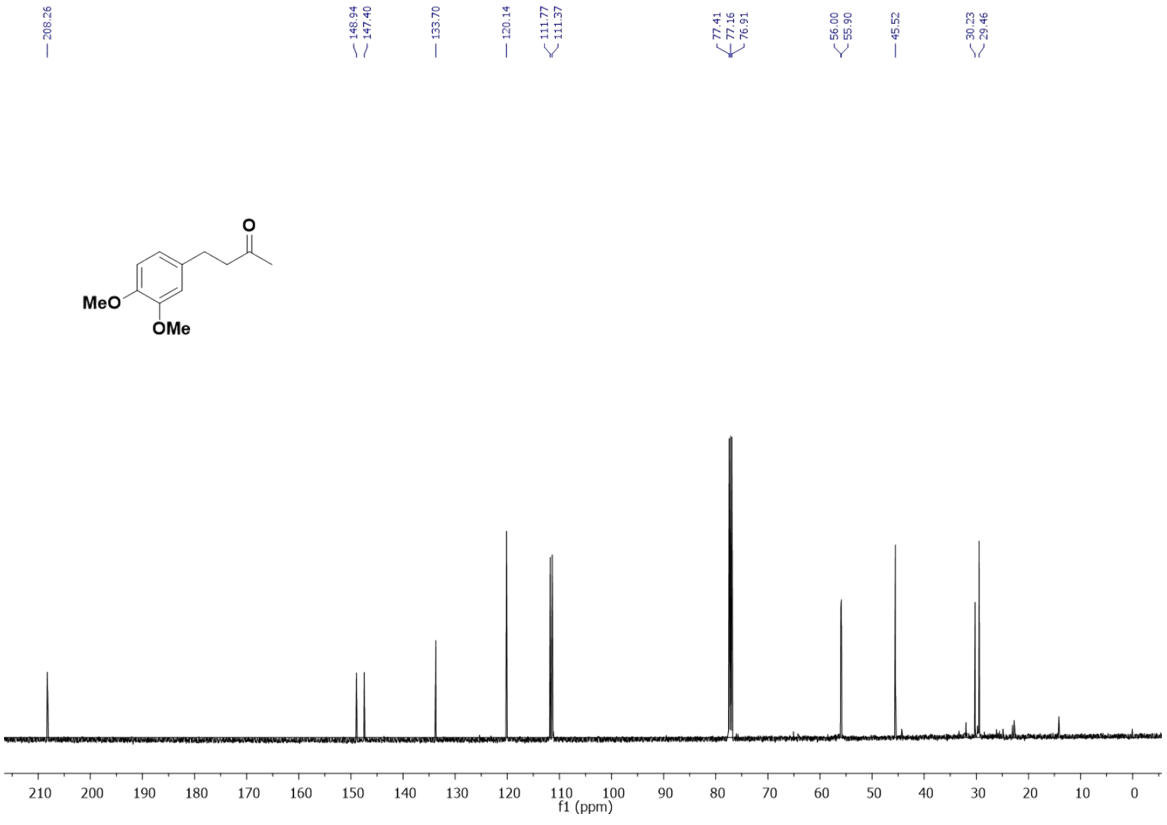


Fig.S6. ^{13}C NMR of compound 5

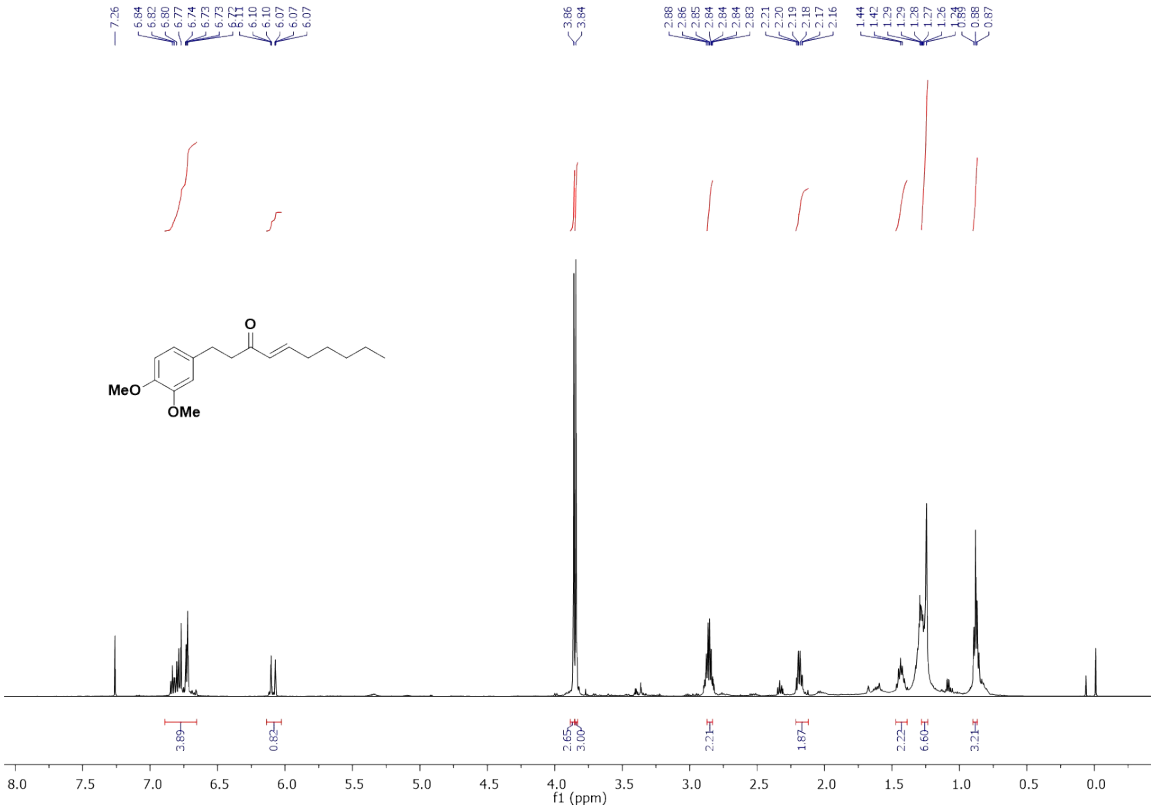


Fig.S7. ^{1}H NMR of compound 6

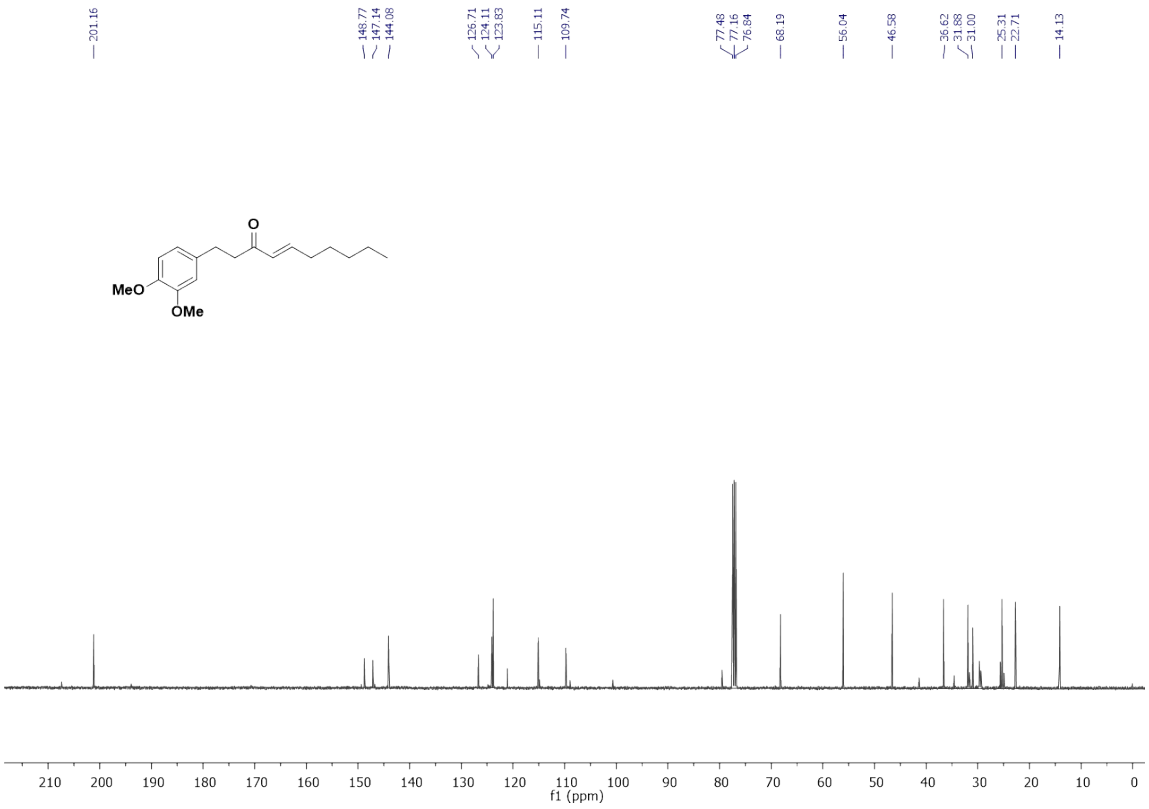


Fig.S8. ^{13}C NMR of compound 6

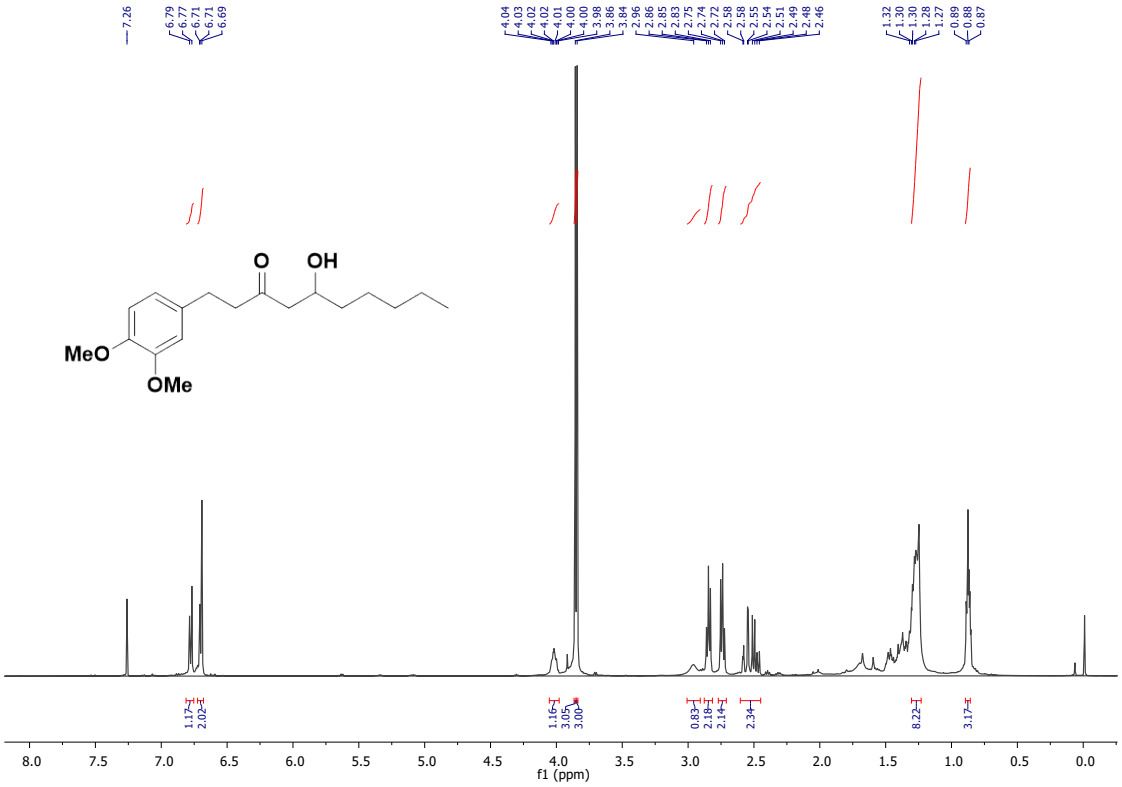


Fig.S9. ^{1}H NMR of compound 7

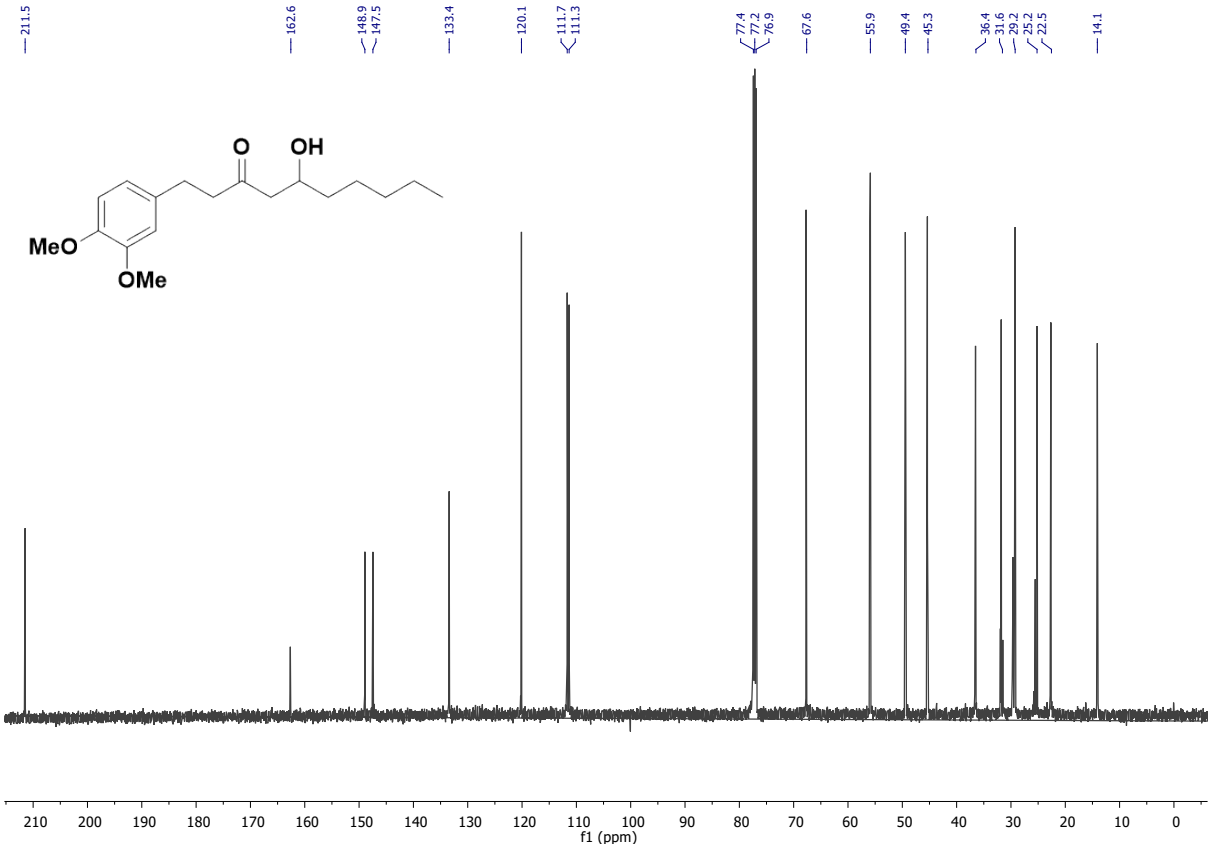


Fig.S10. ^{13}C NMR of compound 7

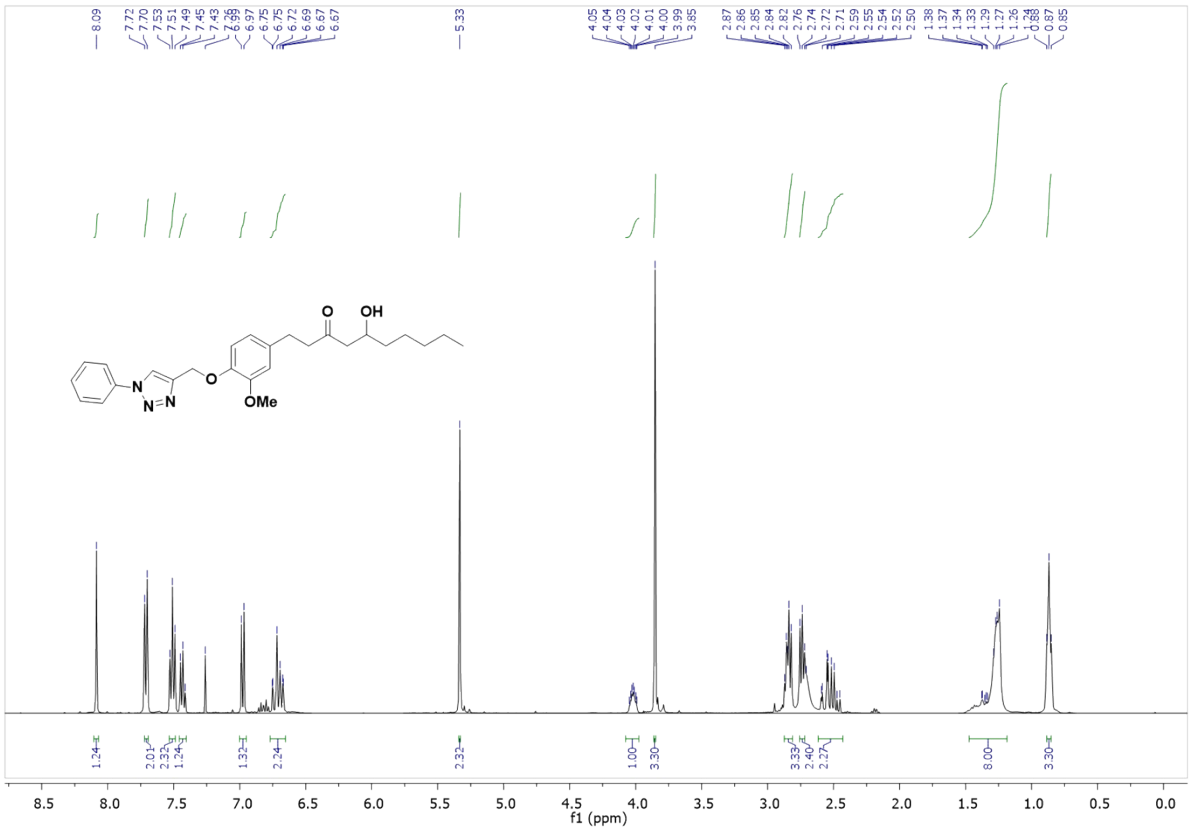


Fig.S11. ^{1}H NMR of compound 11

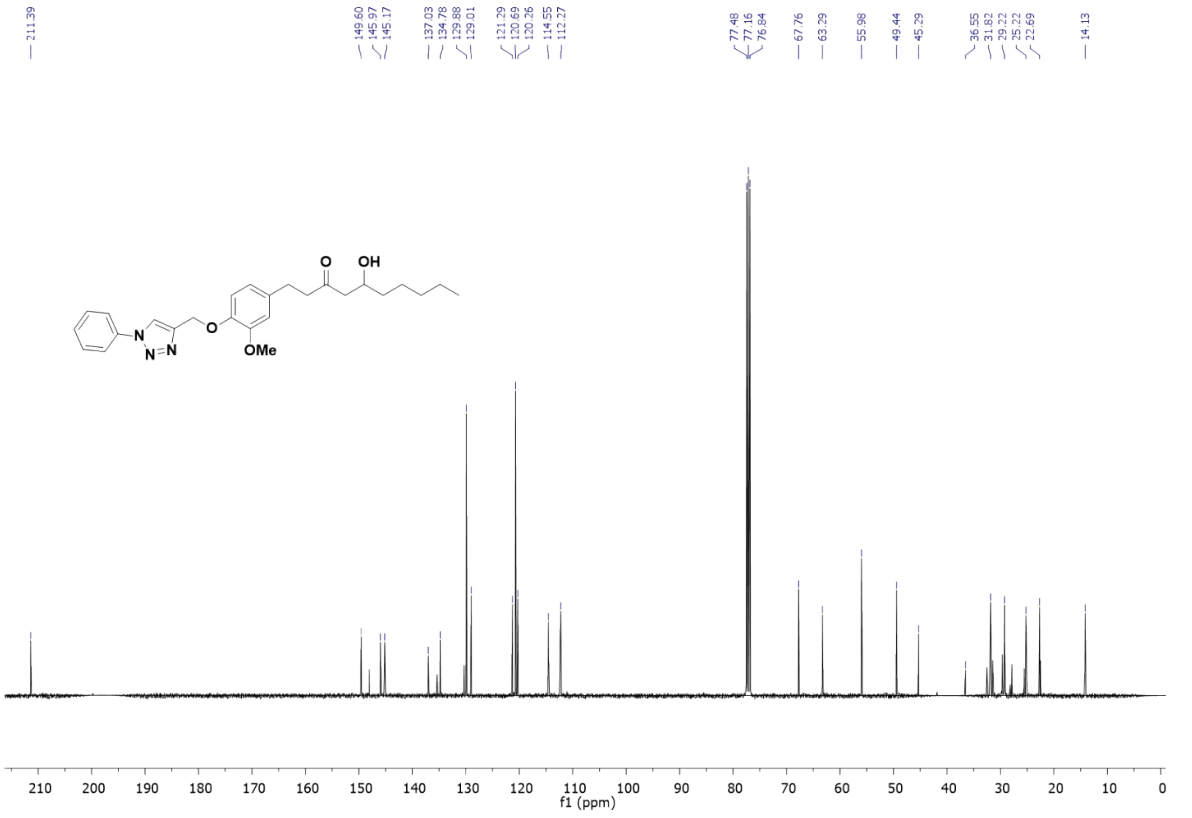


Fig.S12. ^{13}C NMR of compound 11

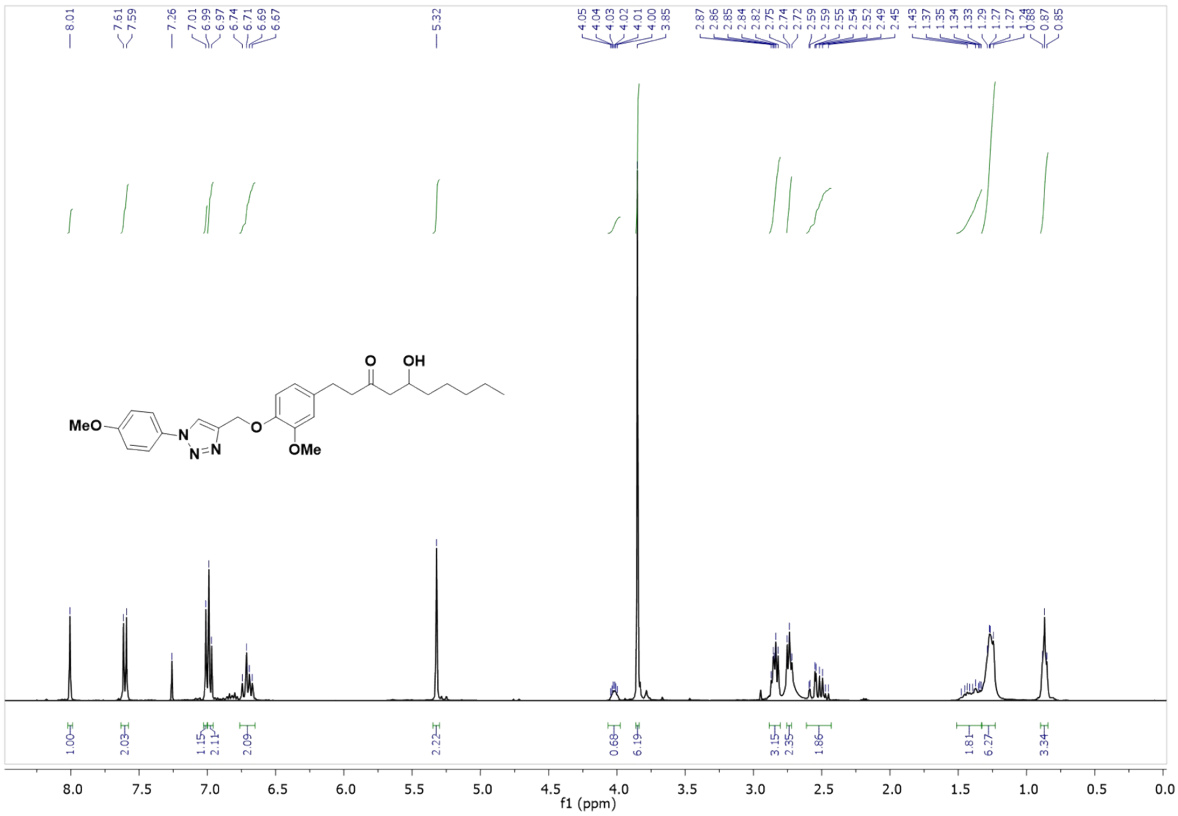


Fig.S13. ^{1}H NMR of compound 12

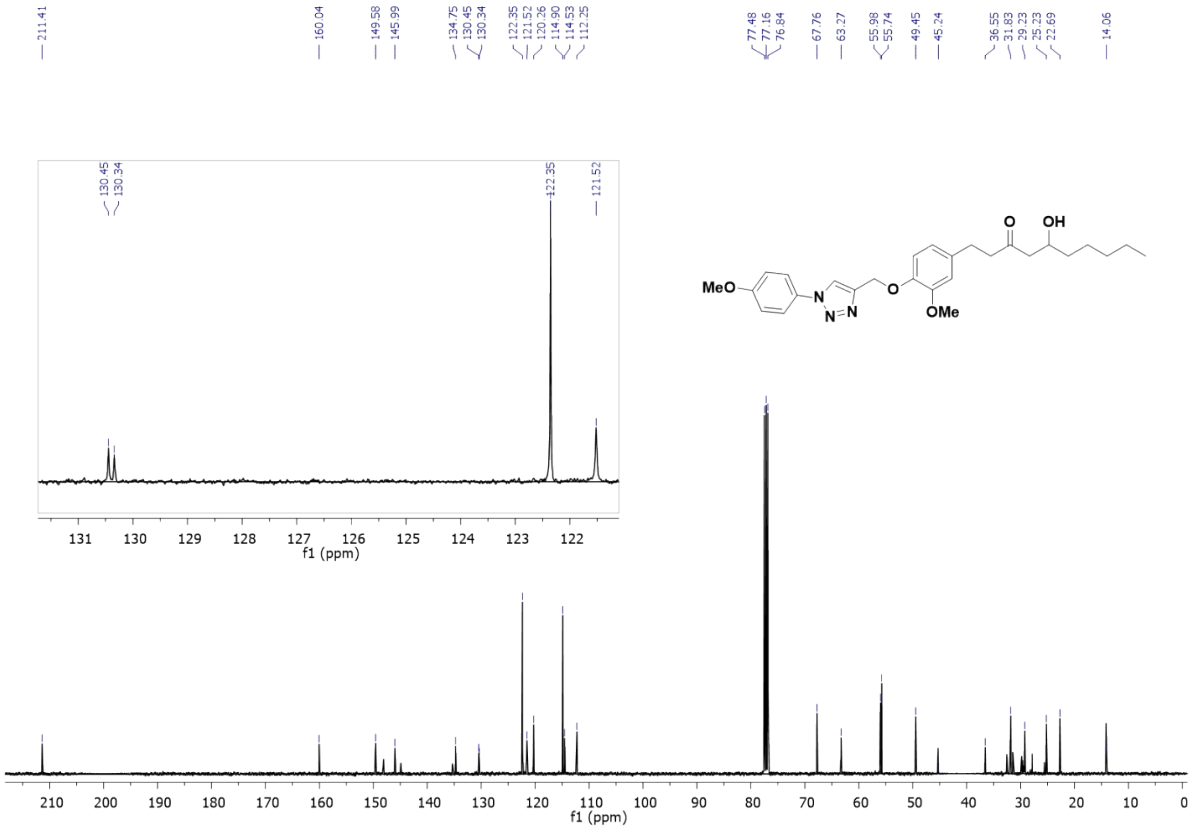


Fig.S14. ^{13}C NMR of compound 12

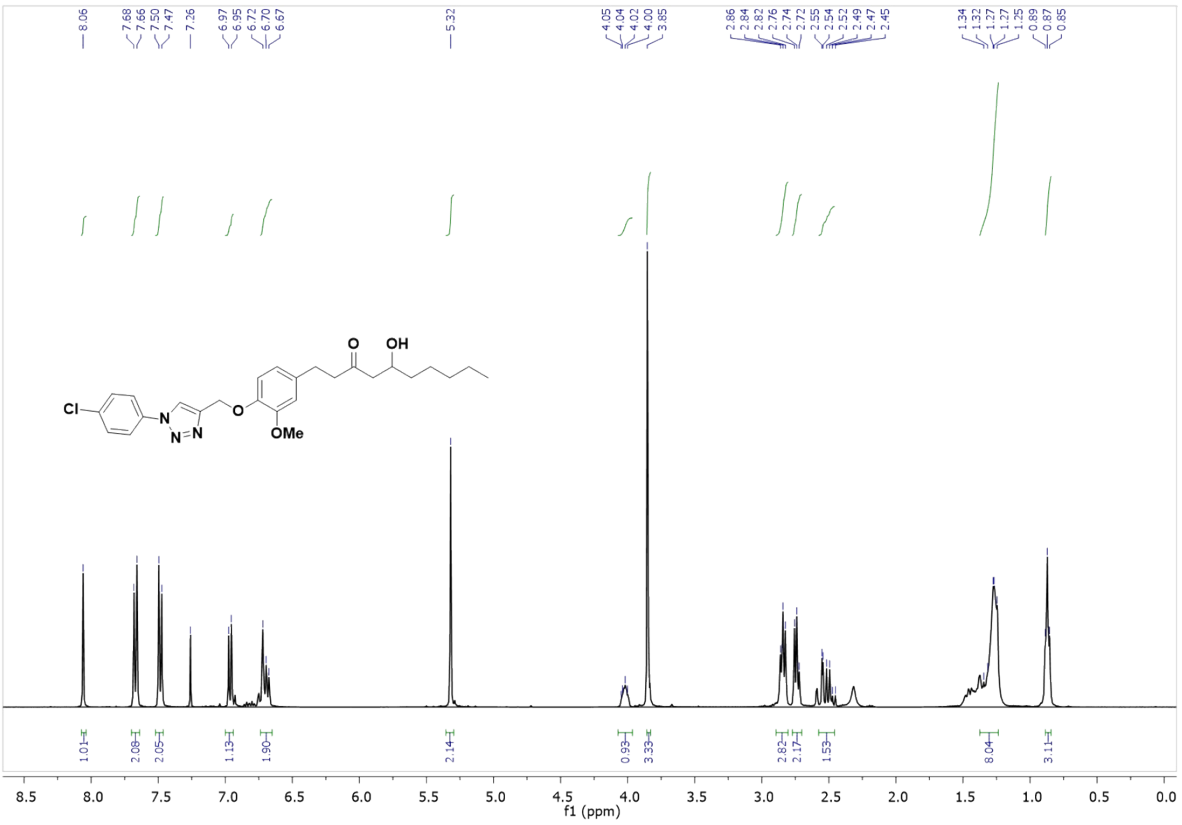


Fig.S15. ^{1}H NMR of compound 13

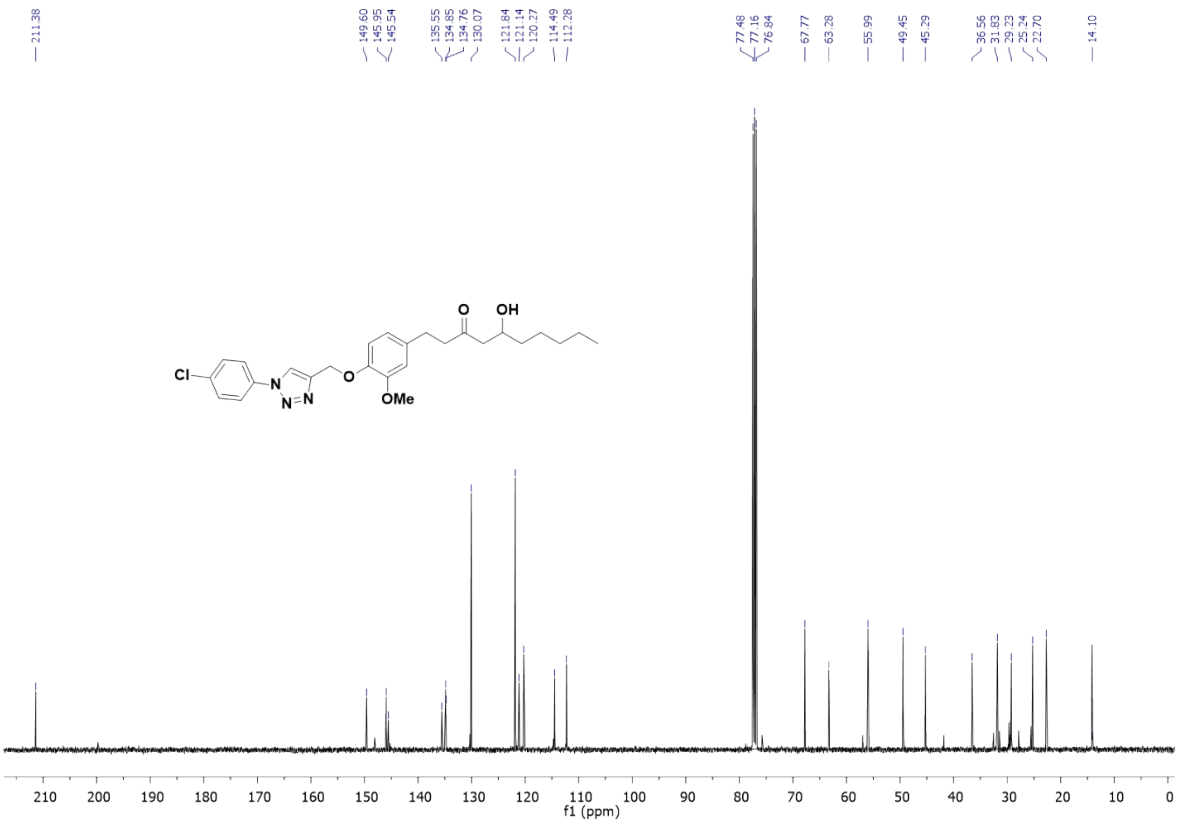


Fig.S16. ^{13}C NMR of compound 13

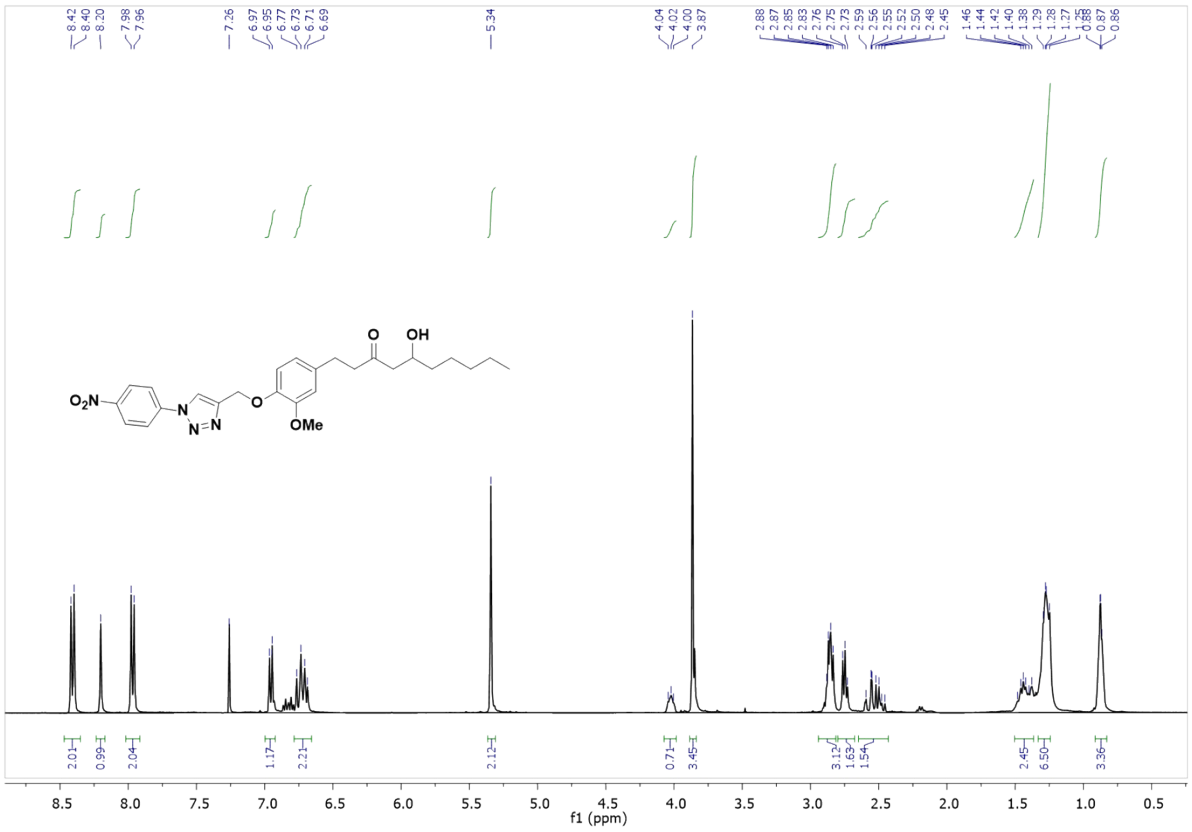


Fig.S17. ^{1}H NMR of compound 14

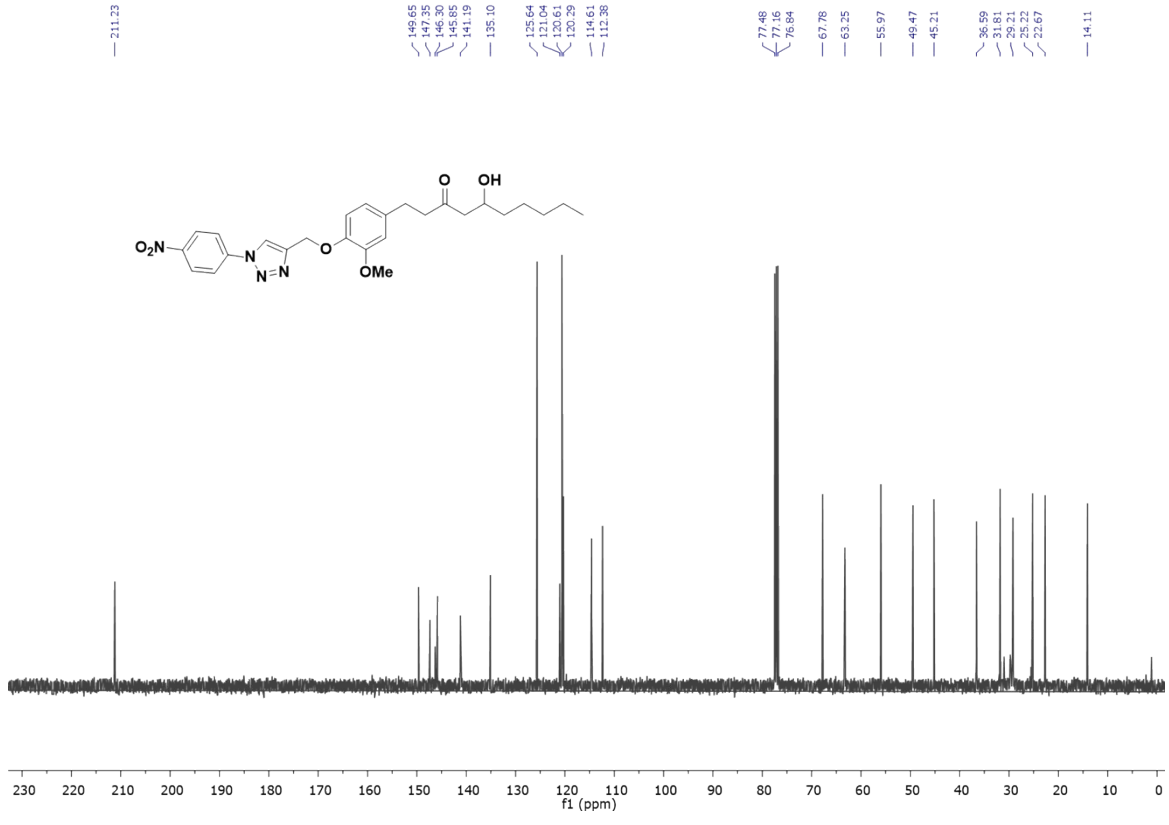


Fig.S18. ^{13}C NMR of compound 14

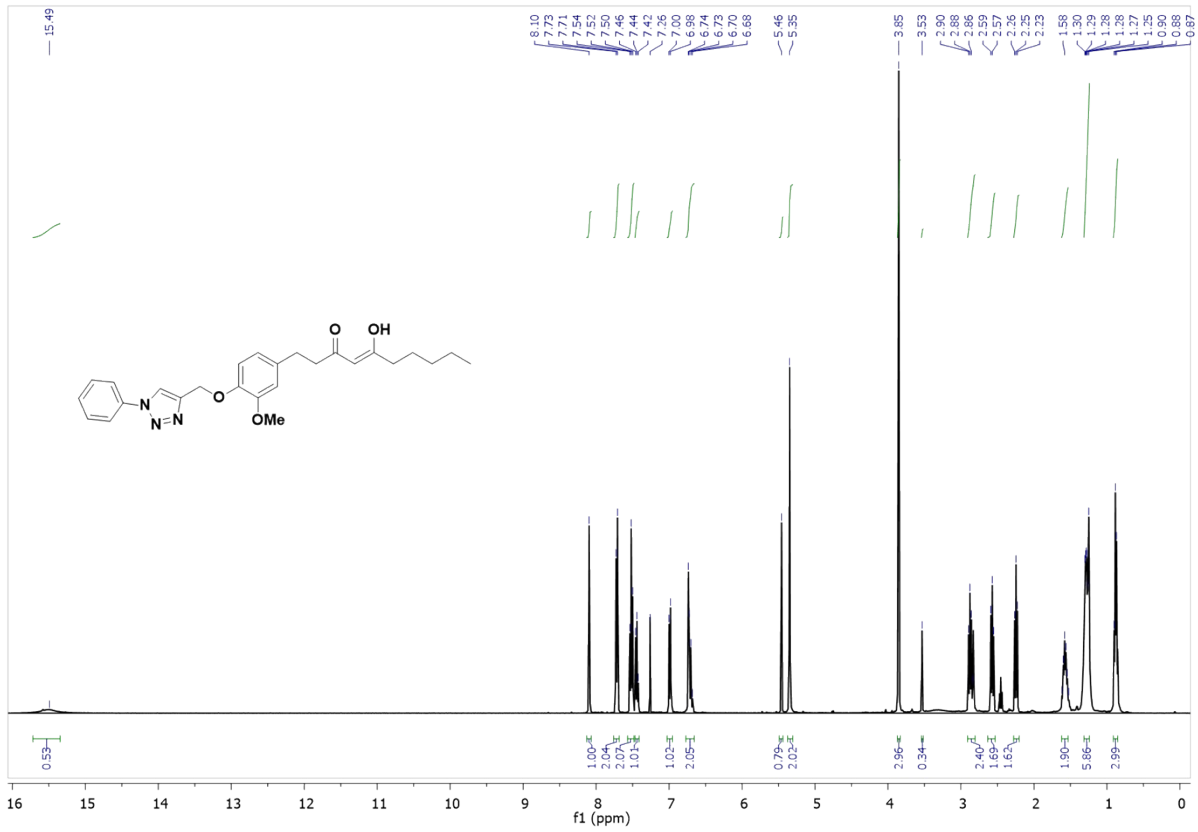


Fig.S19. ^{1}H NMR of compound 15

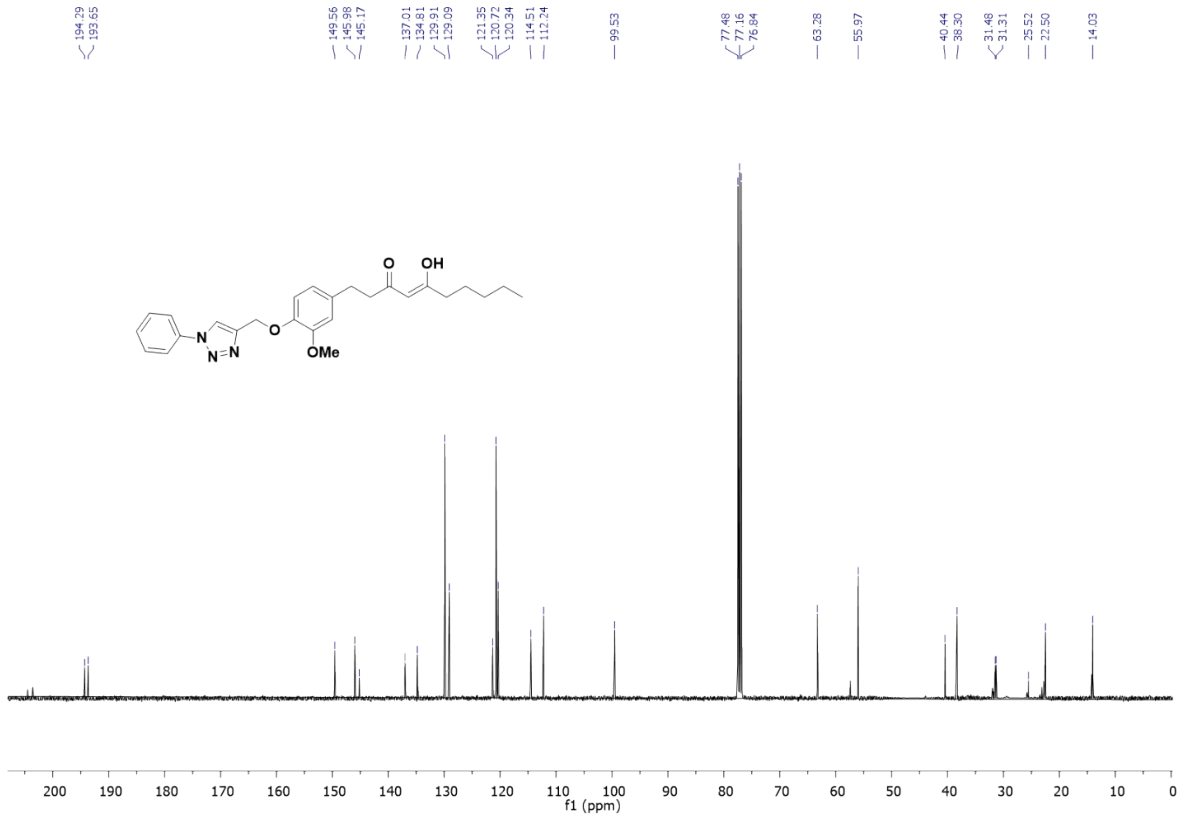


Fig.S20. ^{13}C NMR of compound 15

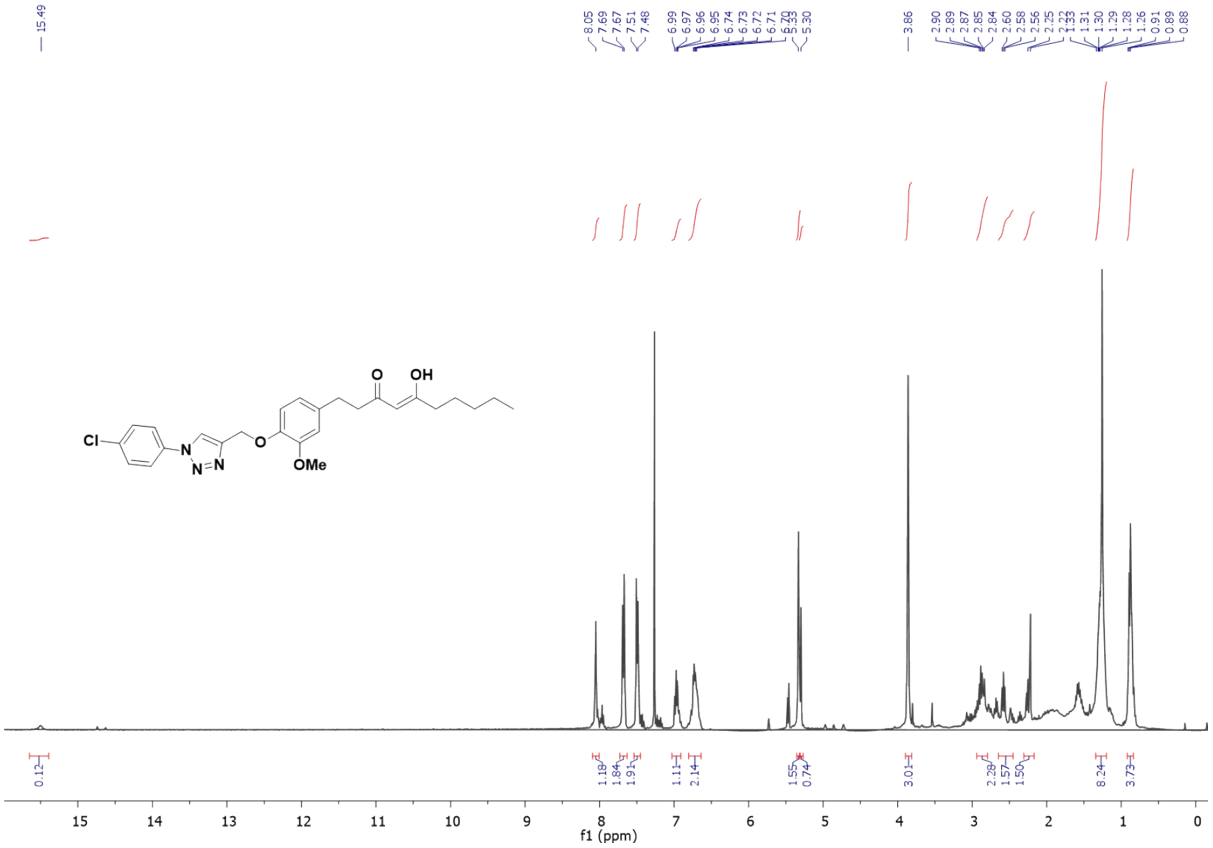


Fig.S21. ^{1}H NMR of compound 16

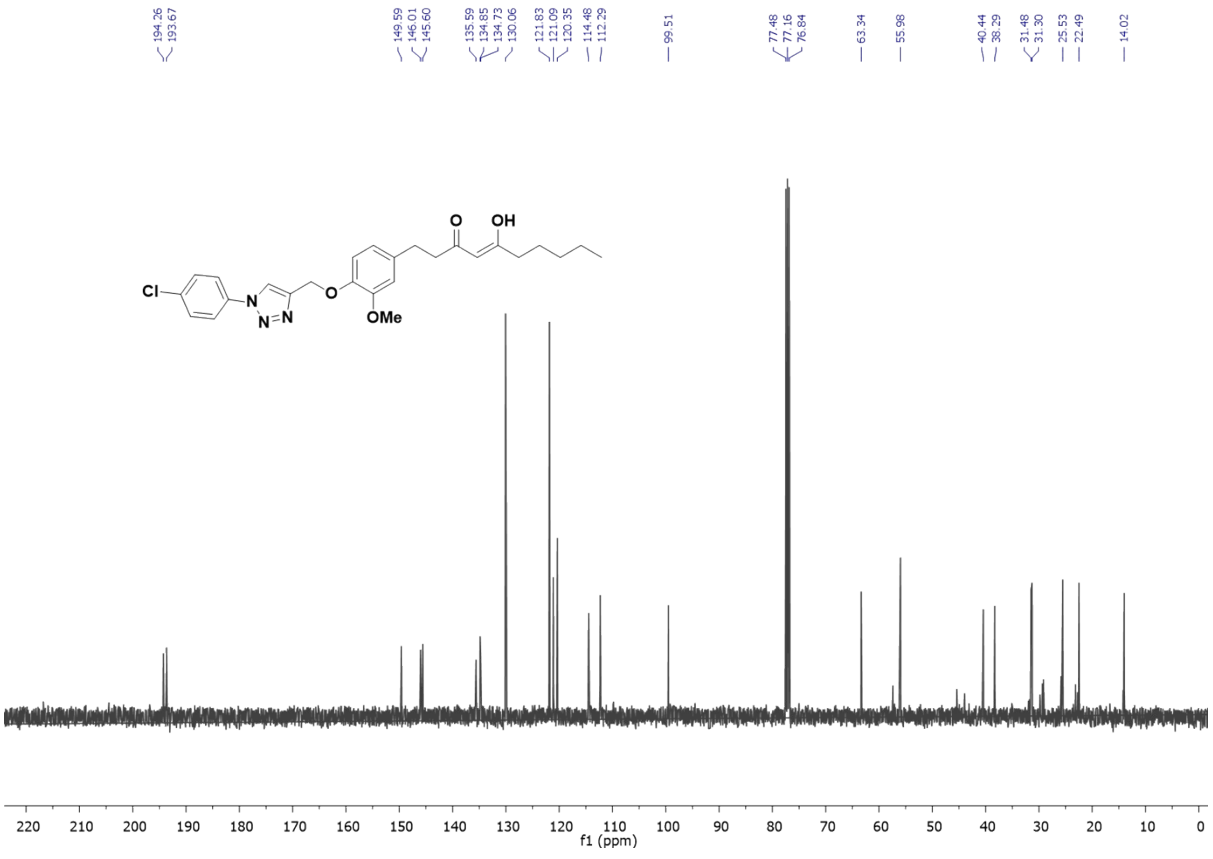


Fig.S22. ^{13}C NMR of compound 16

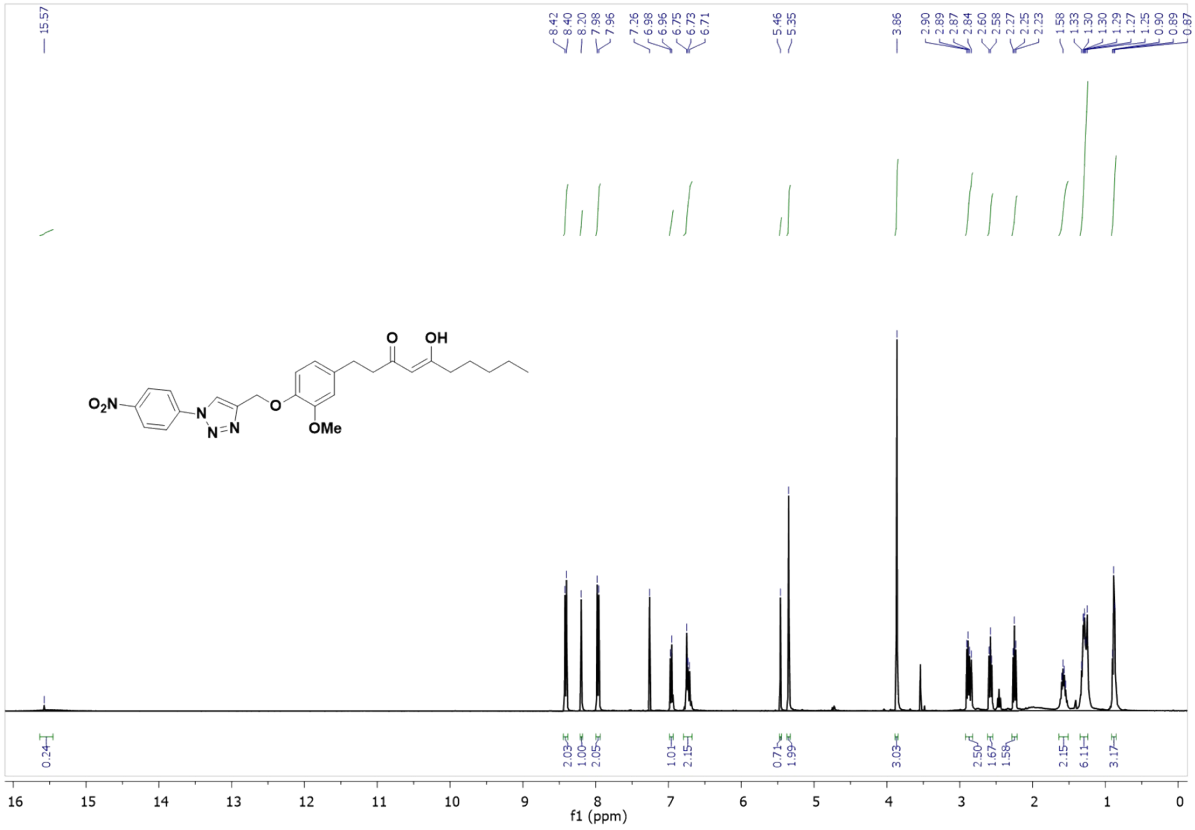


Fig.S23. ^{1}H NMR of compound 17

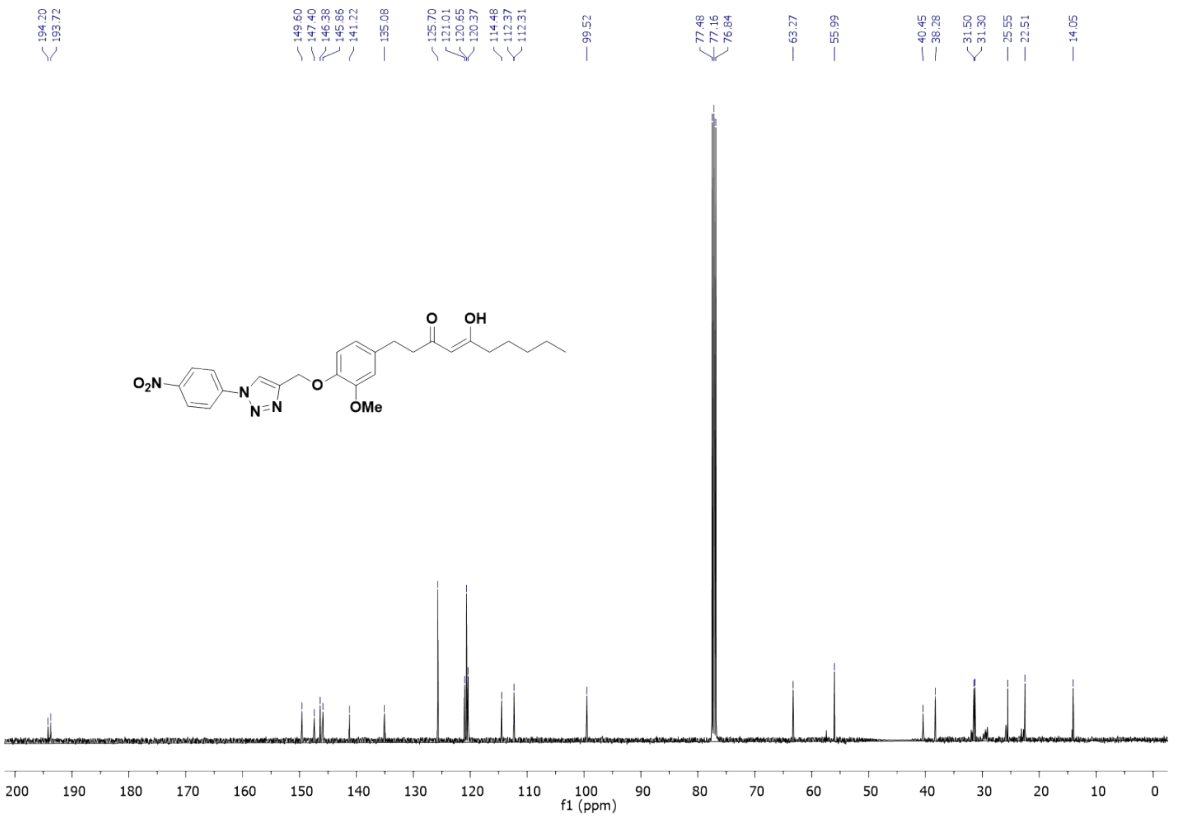


Fig.S24. ^{13}C NMR of compound 17

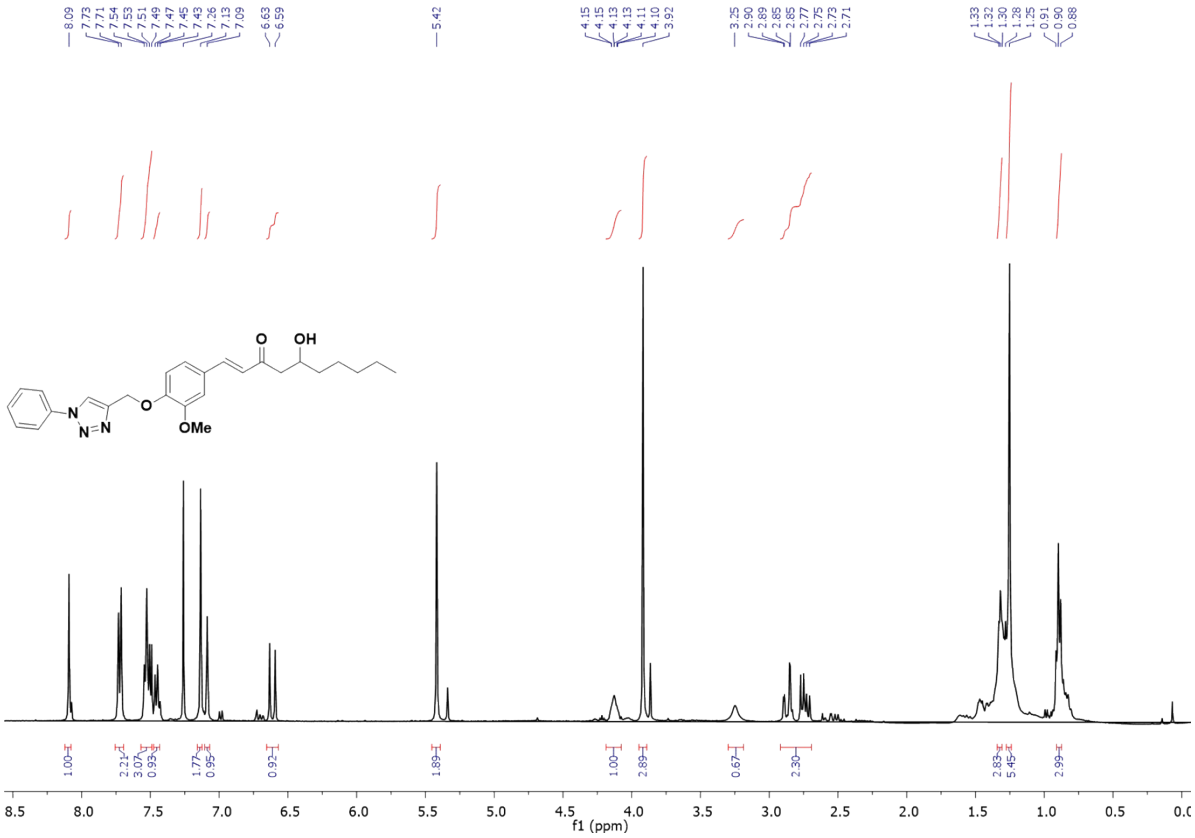


Fig.S25. ^{1}H NMR of compound 19

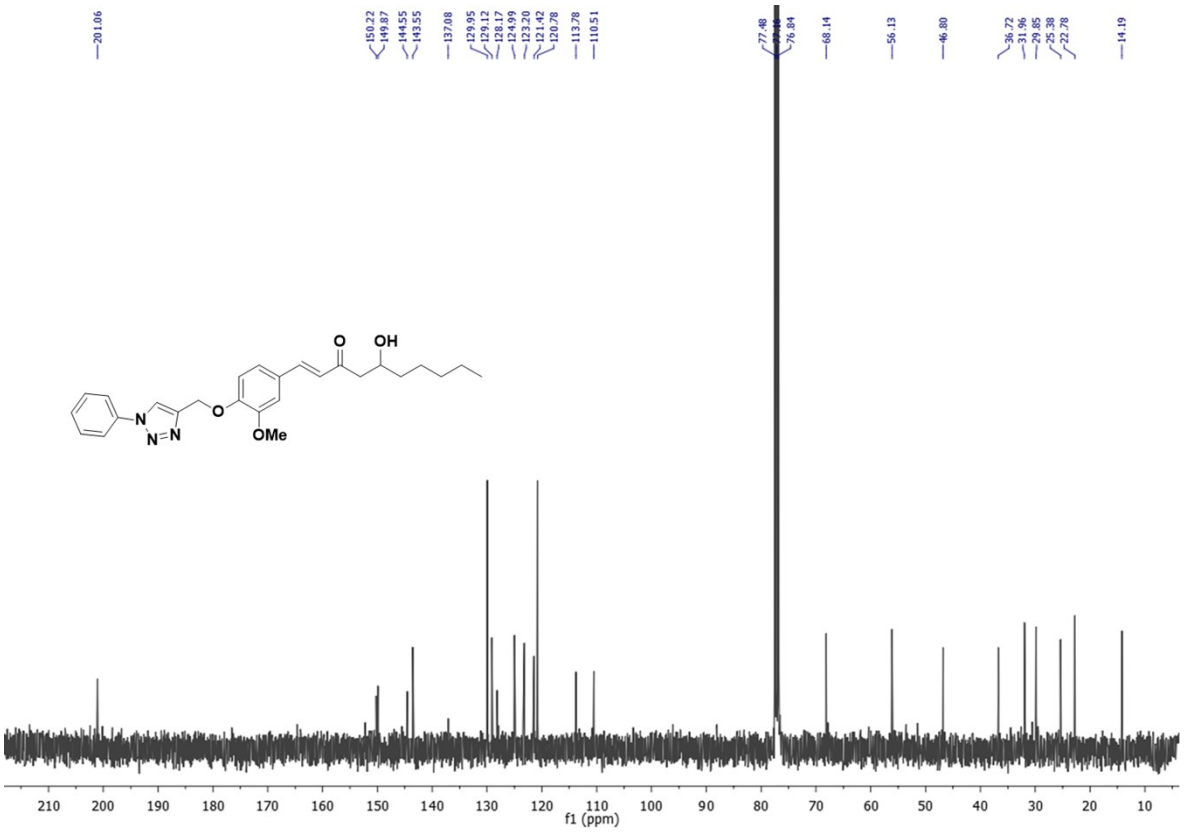


Fig.S26. ^{13}C NMR of compound 19

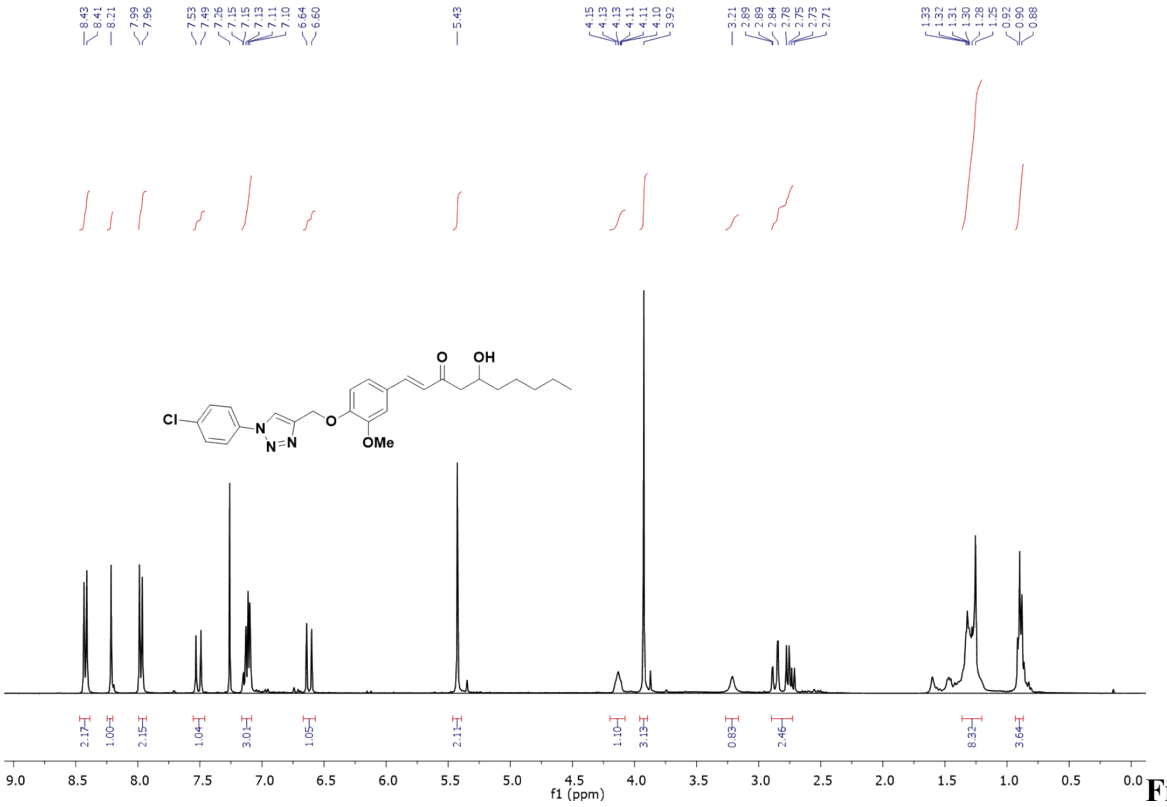


Fig. S27. ^{1}H NMR of compound 20

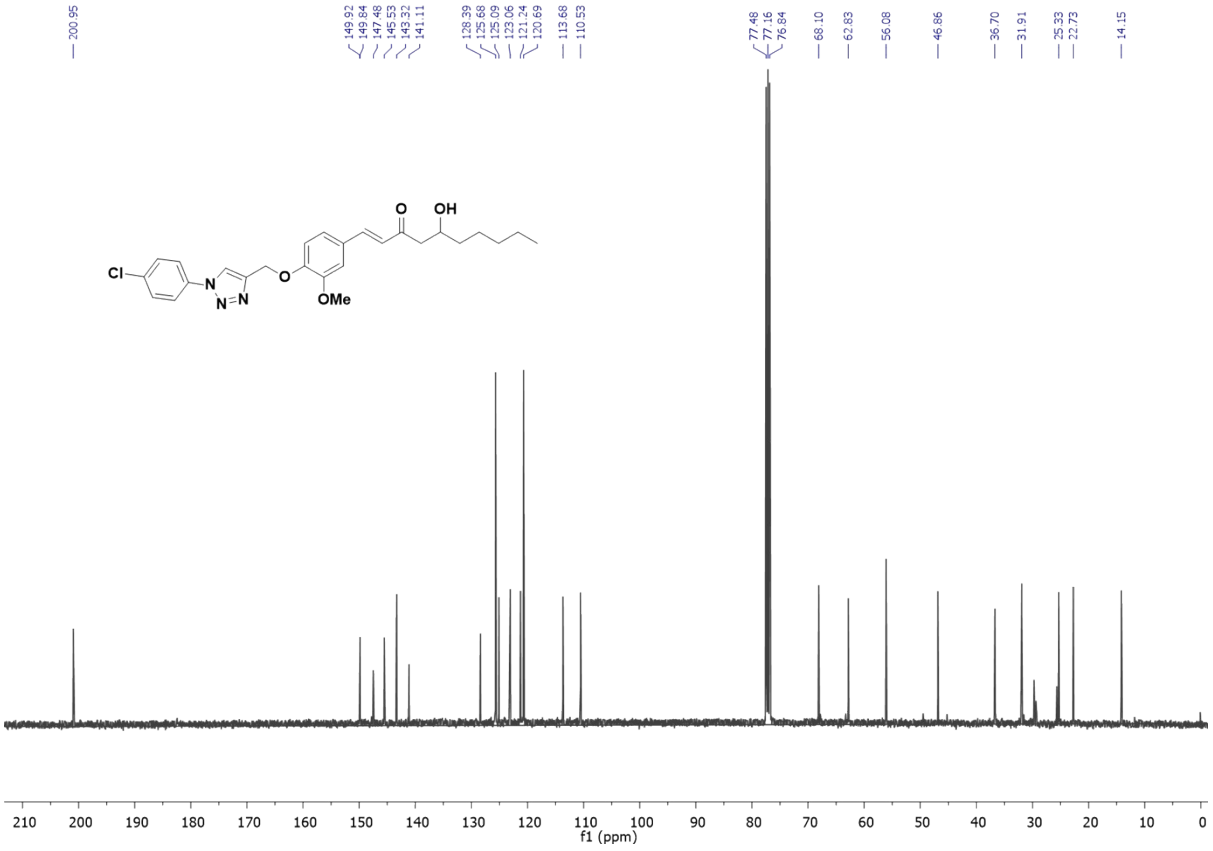


Fig.S28. ^{13}C NMR of compound 20

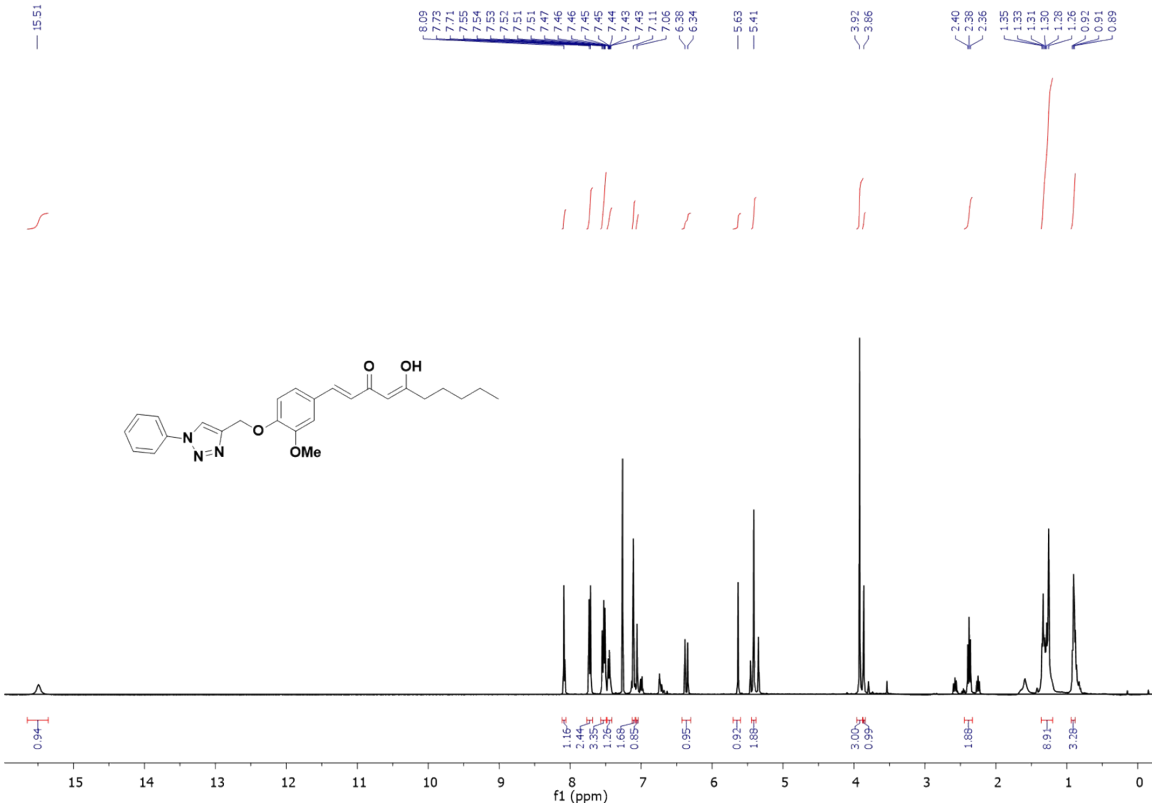


Fig.S29. ^{1}HNMR of compound 21

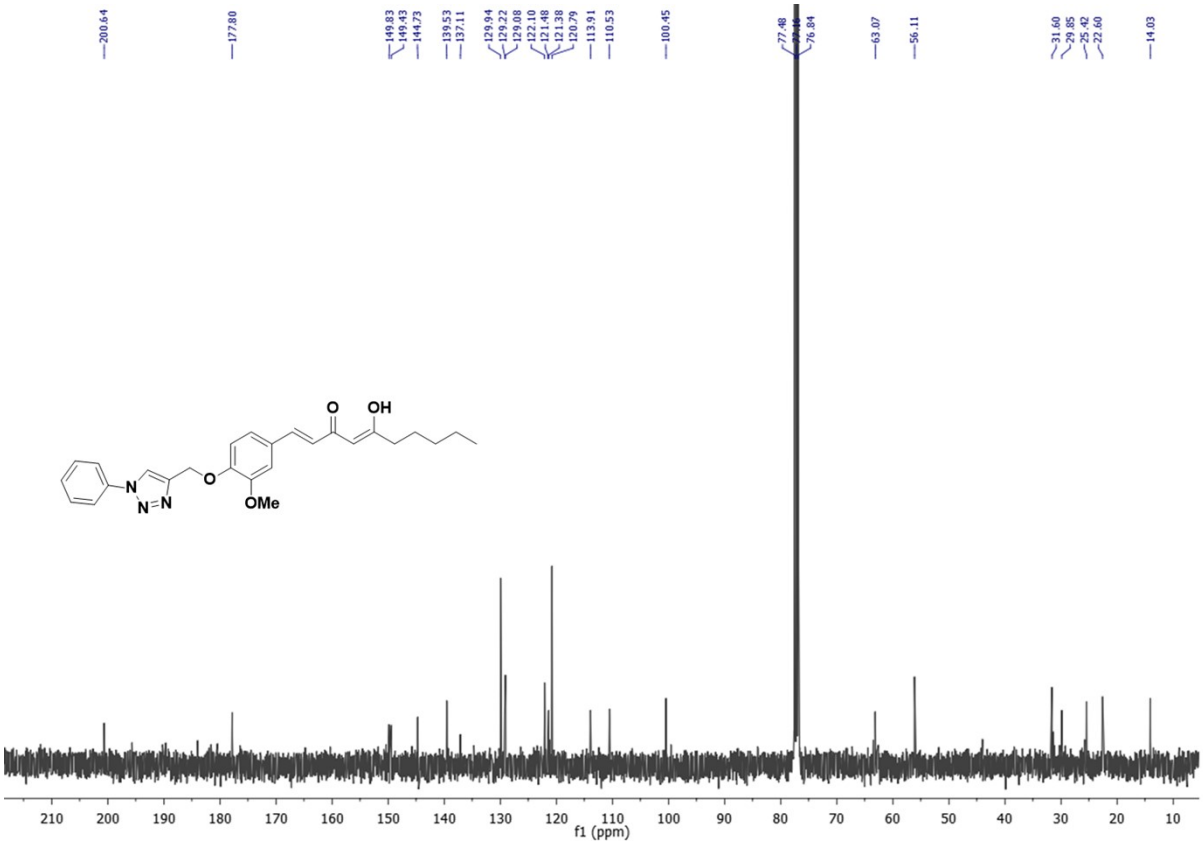


Fig.S30. ^{13}C NMR of compound 21

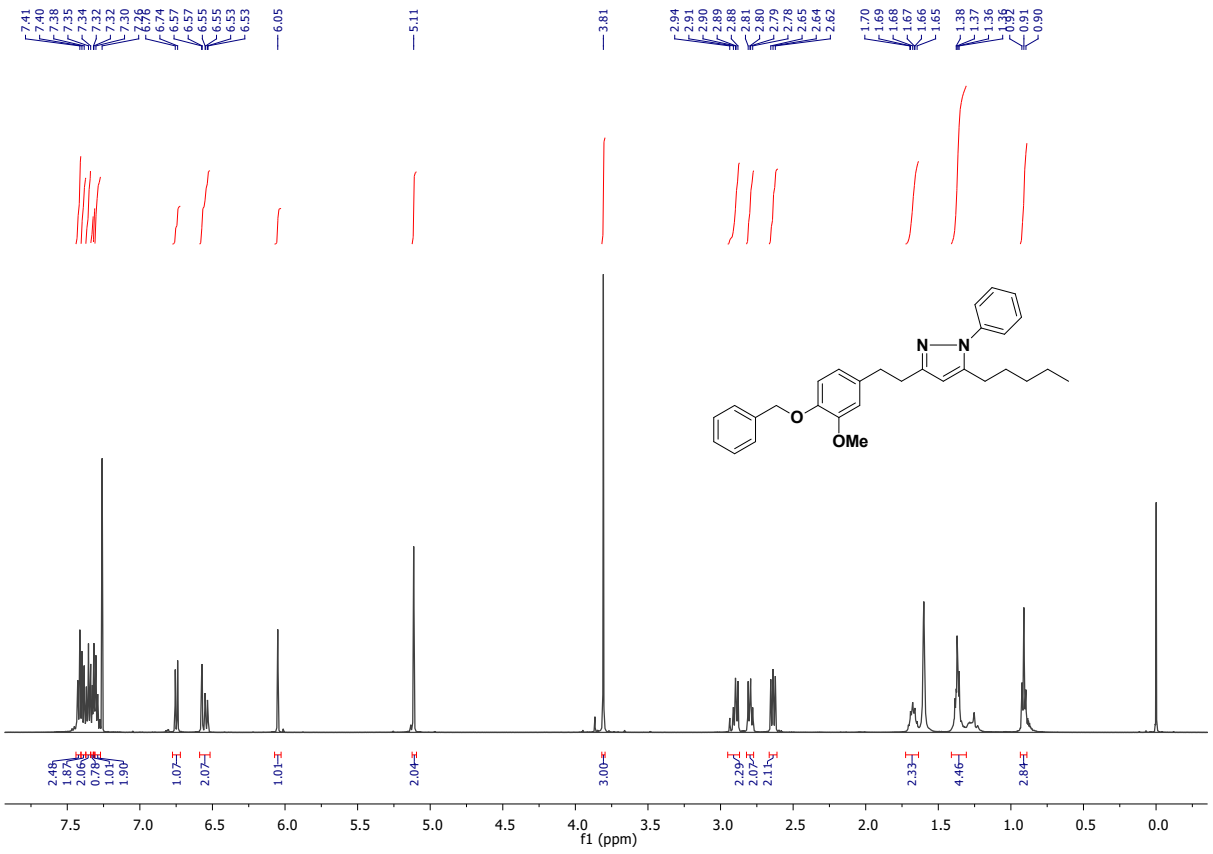


Fig.S31. ^{1}H NMR of compound 22

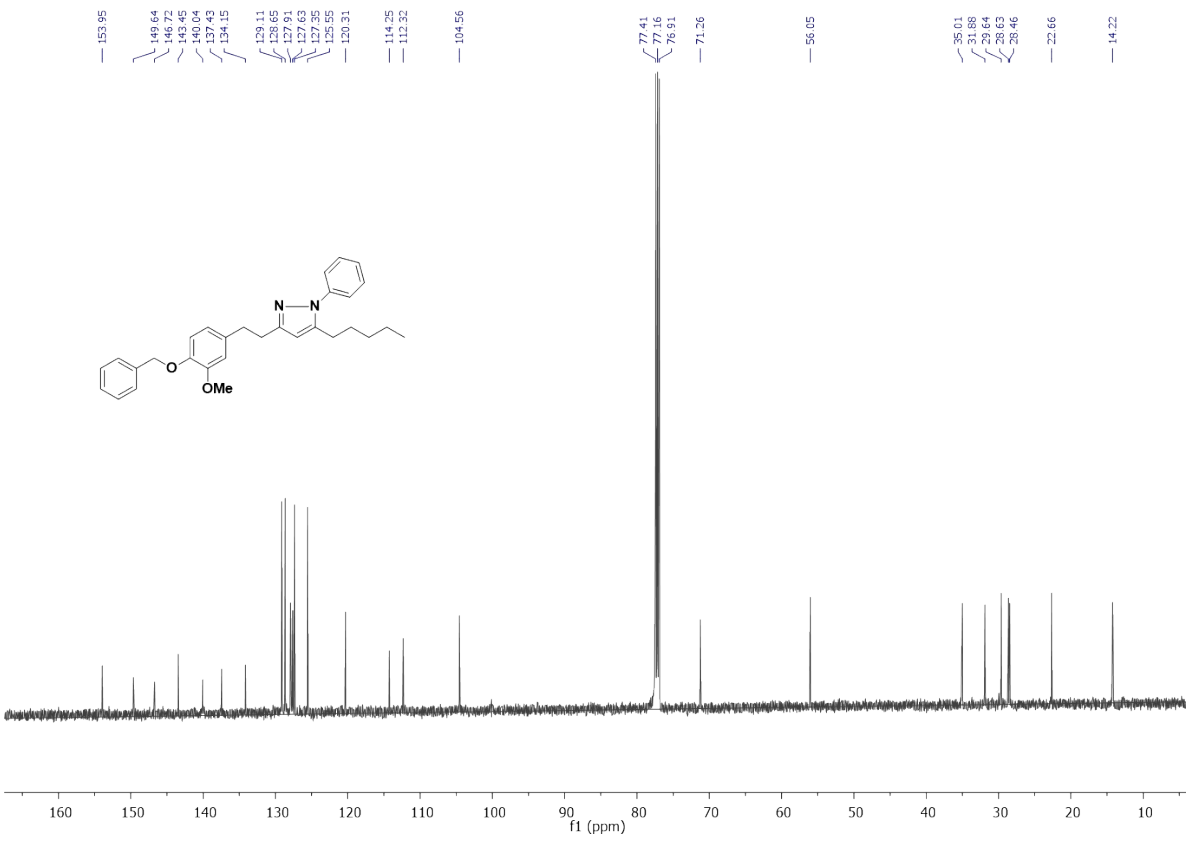


Fig.S32. ^{13}C NMR of compound 22

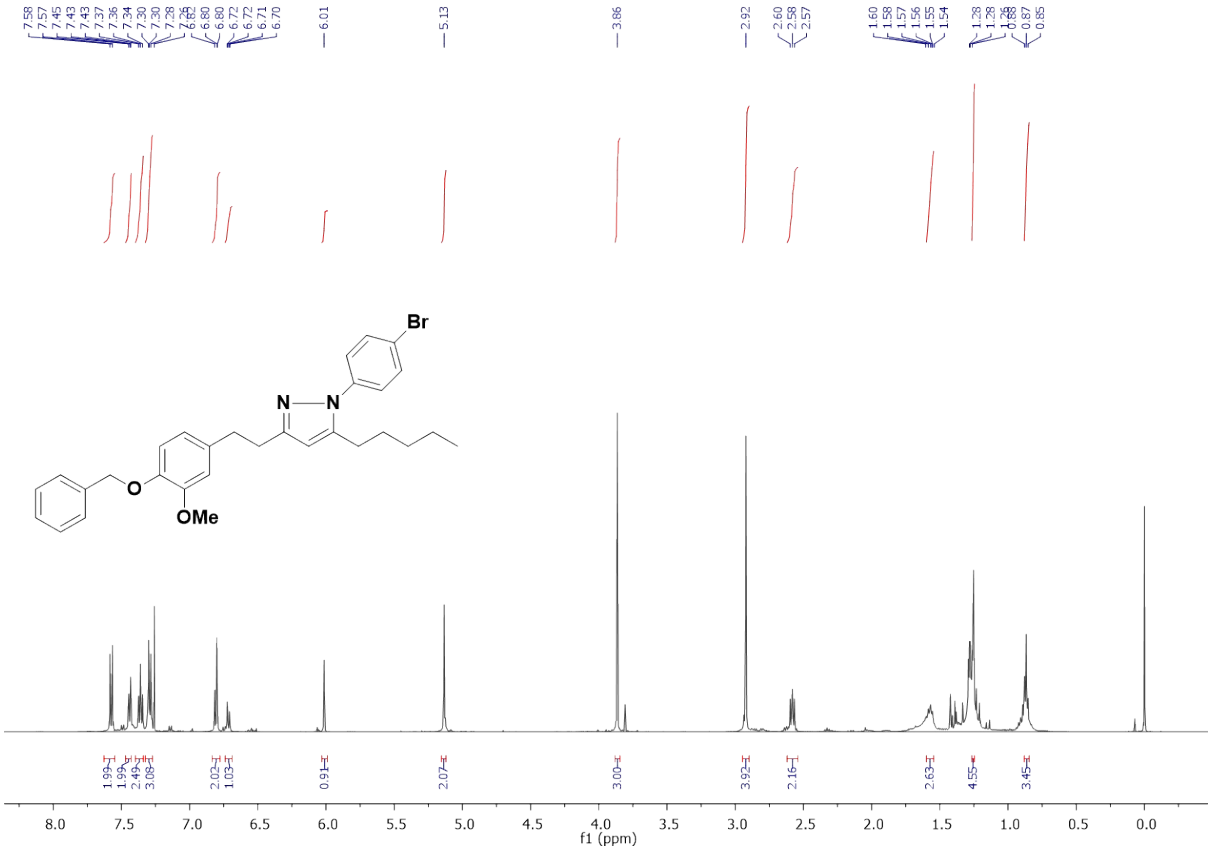


Fig.S33. ^{1}H NMR of compound 23

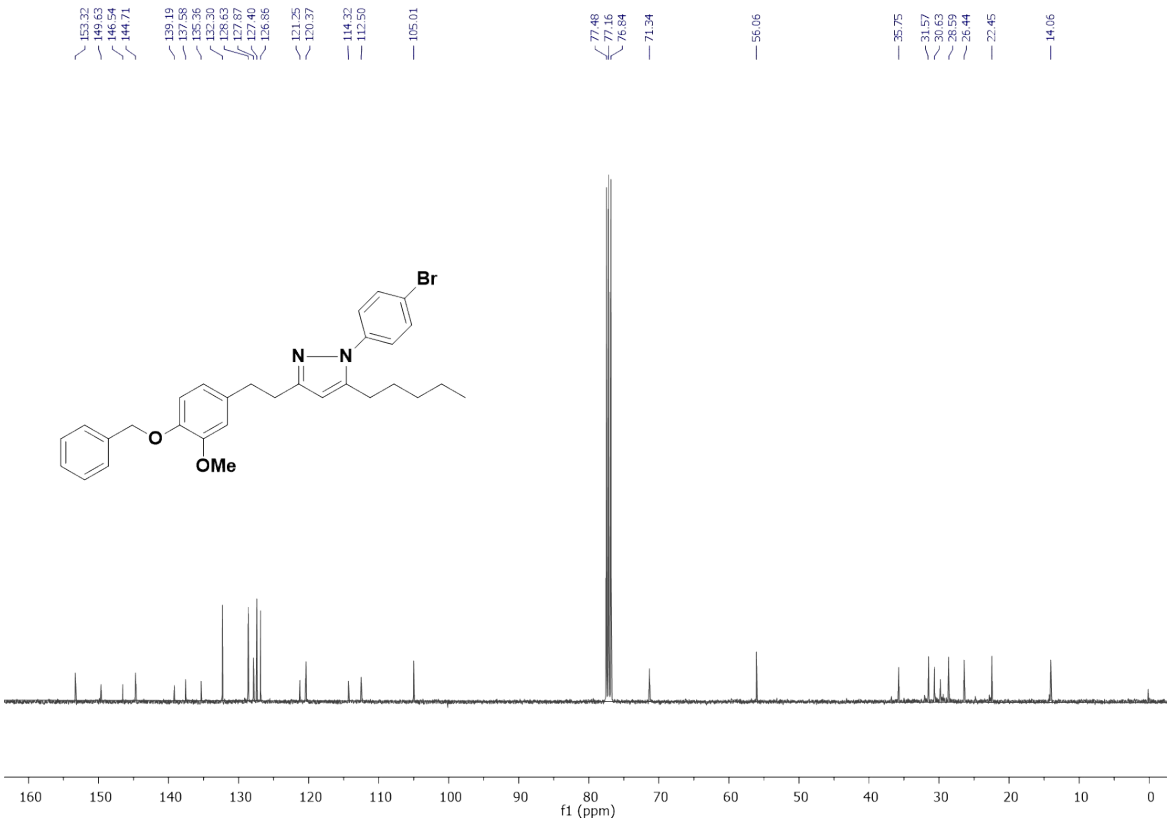


Fig.S34. ^{13}C NMR of compound 23

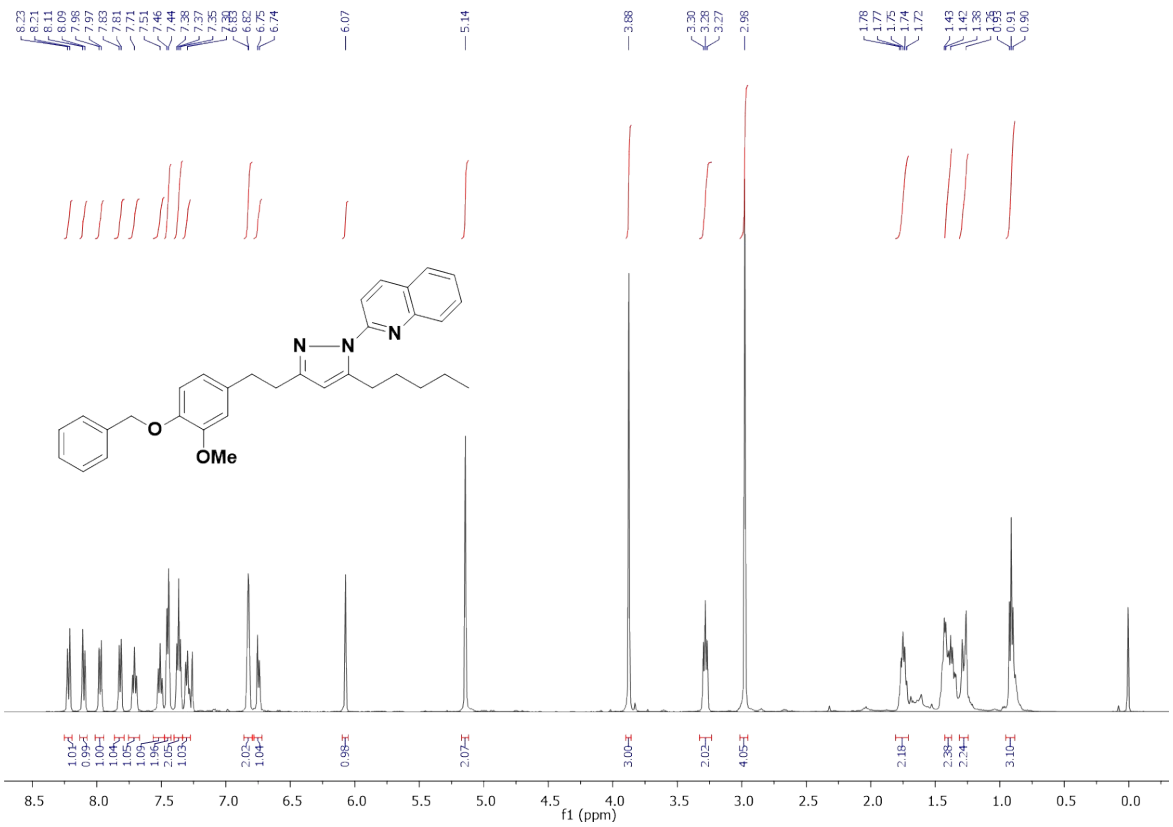


Fig.S35. ^{1}H NMR of compound 24

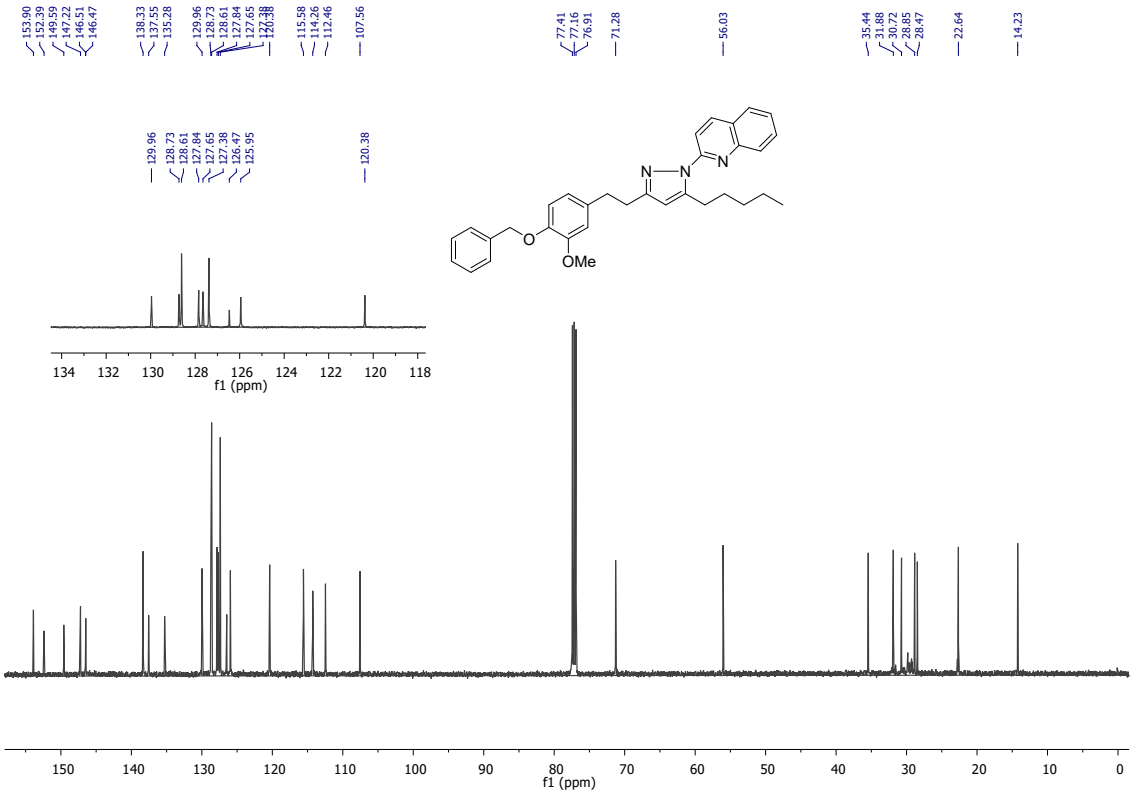


Fig.S36. ^{13}C NMR of compound 24

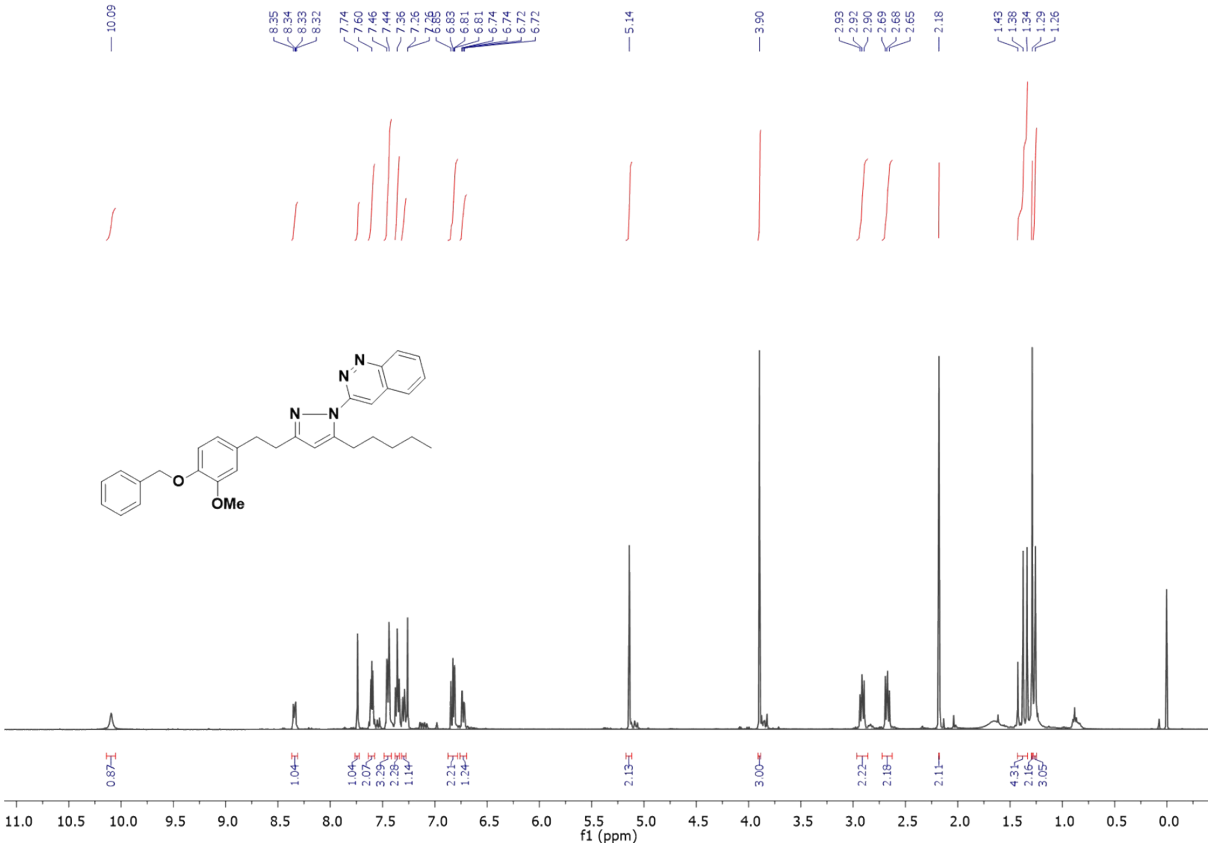


Fig.S37. ^{1}H NMR of compound 25

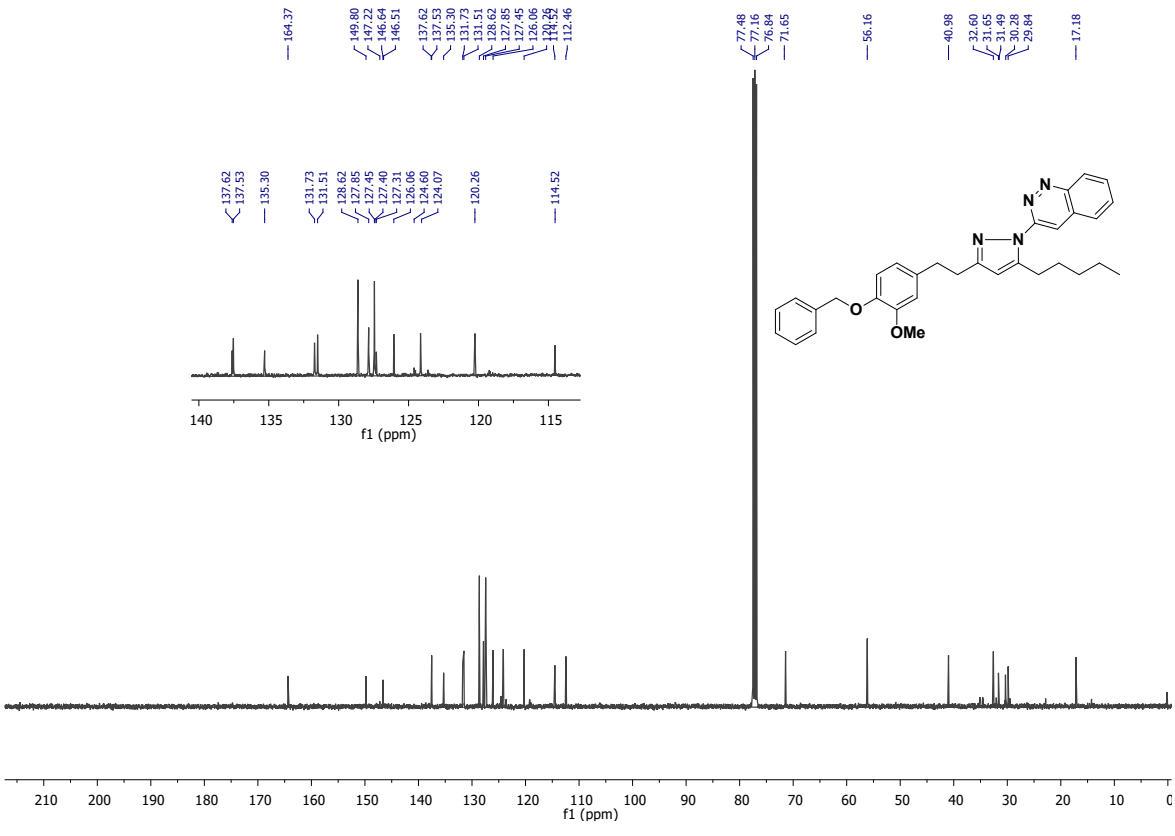


Fig.S38. ^{13}C NMR of compound 25

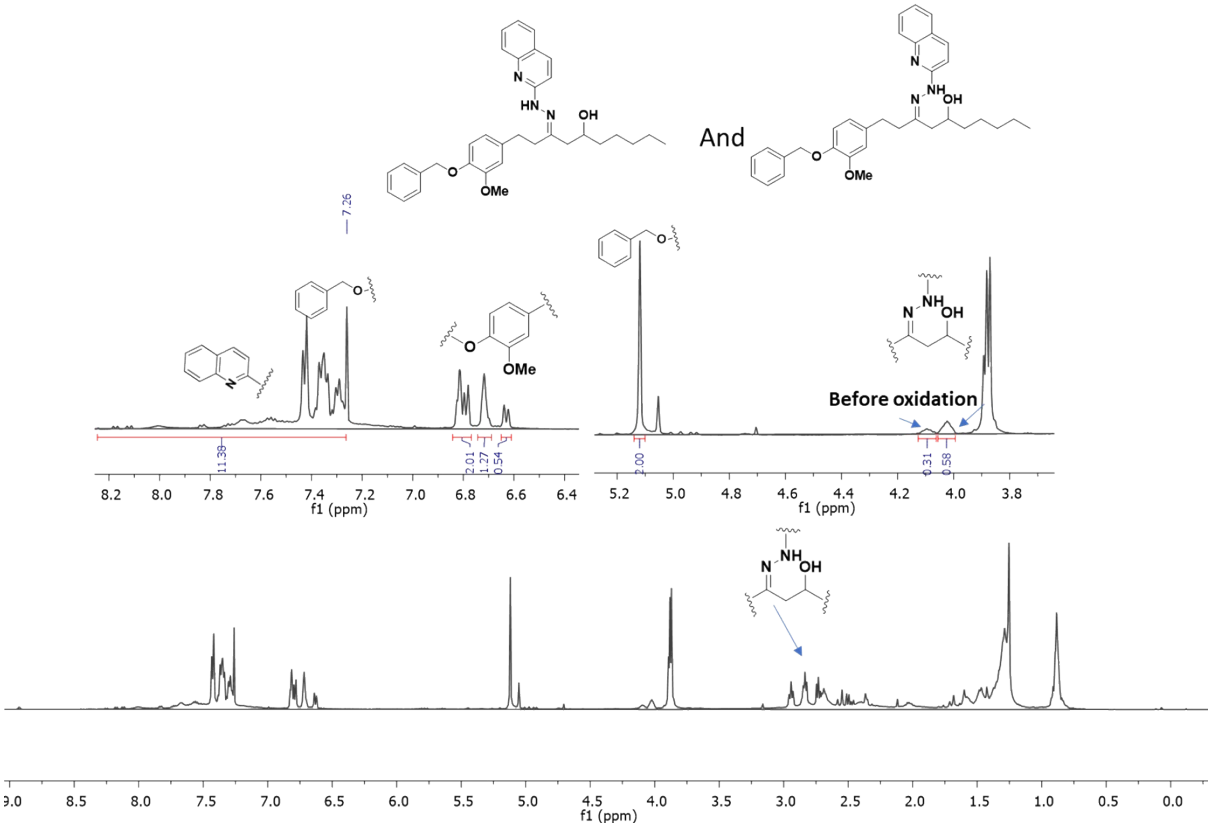


Fig.S39. ^{1}H NMR of intermediate Int-1

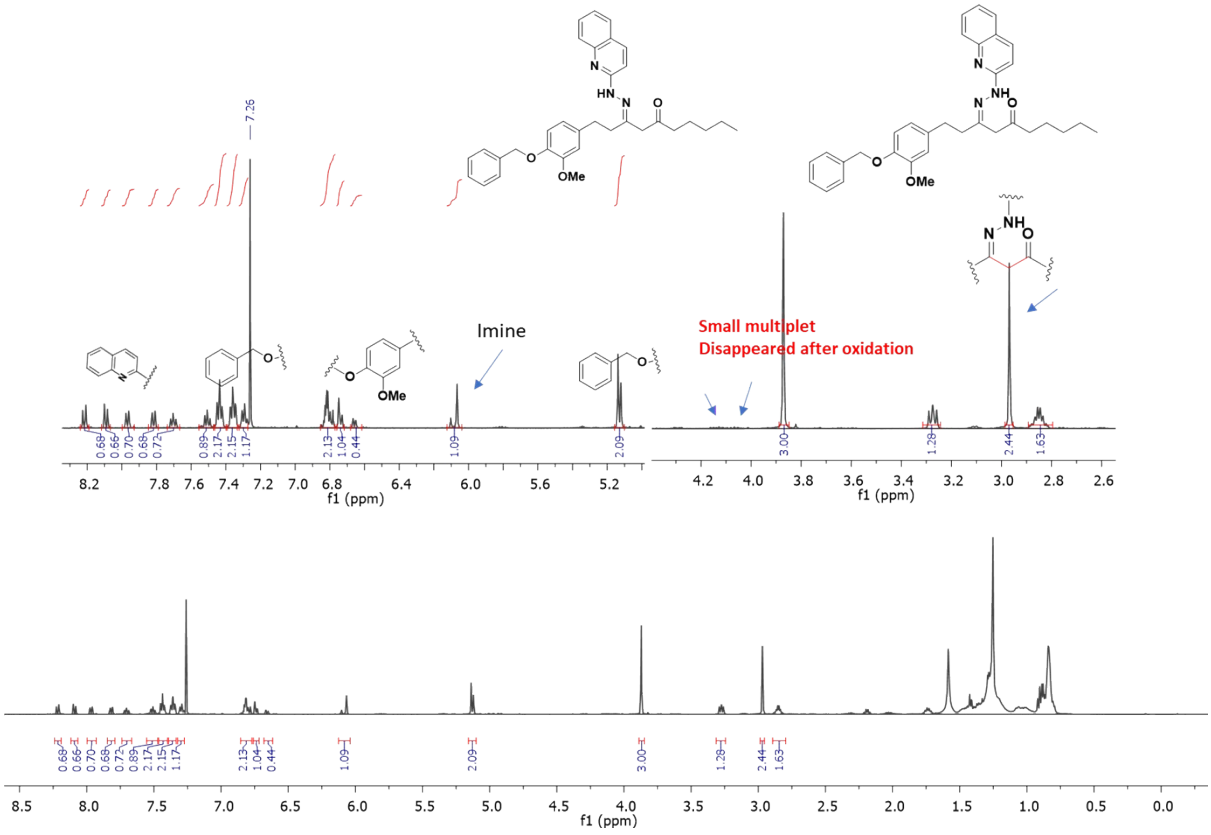


Fig.S40. ^{1}H NMR of intermediate Int-2

Crystallographic data

```
Bond precision:   C-C = 0.0059 A          Wavelength=0.71073

Cell:             a=5.650(3)        b=12.577(6)       c=20.32(1)
                  alpha=87.21(1)    beta=83.78(1)     gamma=89.06(1)
Temperature:      300 K

                  Calculated                  Reported
Volume            1433.7(12)                  1434.20(10)
Space group       P -1                        P -1
Hall group        -P 1                        -P 1
Moiety formula    C33 H35 N3 O2               C33 H35 N3 O2
Sum formula       C33 H35 N3 O2               C33 H35 N3 O2
Mr                505.64                      505.64
Dx,g cm-3         1.171                       1.171
Z                 2                           2
Mu (mm-1)         0.073                       0.073
F000              540.0                       540.0
F000'             540.21
h,k,lmax          7,16,26                     7,16,26
Nref              6709                        6605
Tmin,Tmax         0.981,0.984                 0.584,0.746
Tmin'             0.981

Correction method= # Reported T Limits: Tmin=0.584 Tmax=0.746
AbsCorr = MULTI-SCAN

Data completeness= 0.984          Theta(max)= 27.671

R(reflections)= 0.0929( 2562)                wR2(reflections)=
                                             0.2753( 6605)
S = 1.178                 Npar= 345
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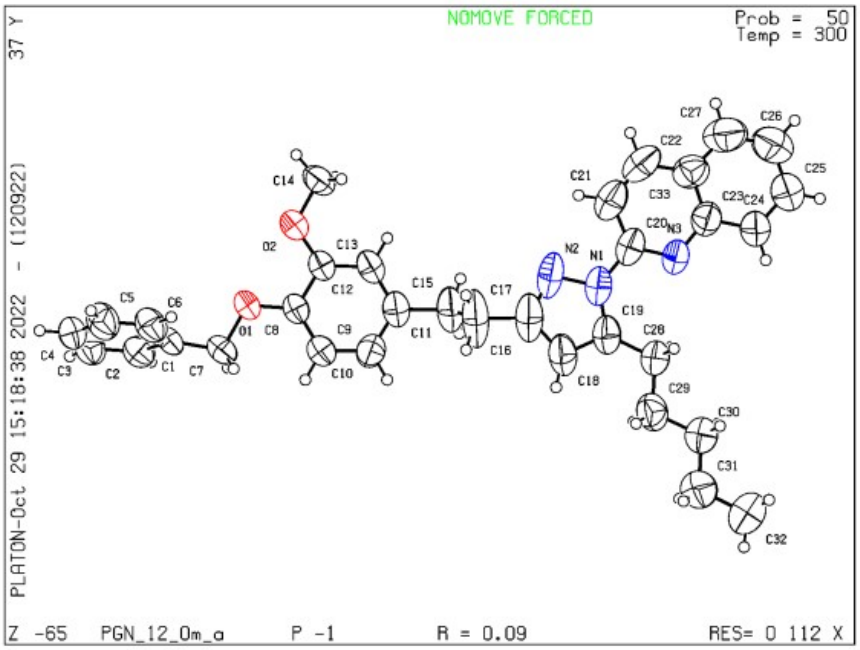

Fig.S41. Single crystal structure (ORTEP diagram) of 24.

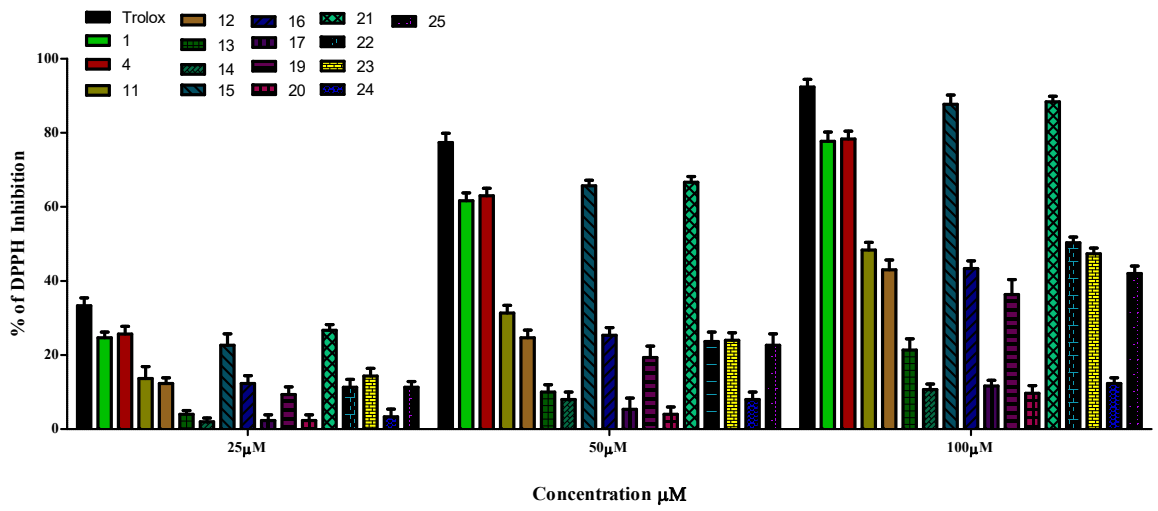

Fig.S42. DPPH free radical scavenging activity of compounds (**1, 4, 11, 12, 13, 14, 15, 16, 17, 19, 20, 21, 22, 23, 24** and **25**) was compared with control Trolox. The single asterisk (*) denotes the significant difference between the control and treatment group (25 µM, 50 µM, and 100 µM) at $p < 0.05$ level by one-way ANOVA followed by Duncan's multiple range test. The experiments were performed in triplicates and the values were provided in mean ± SD.

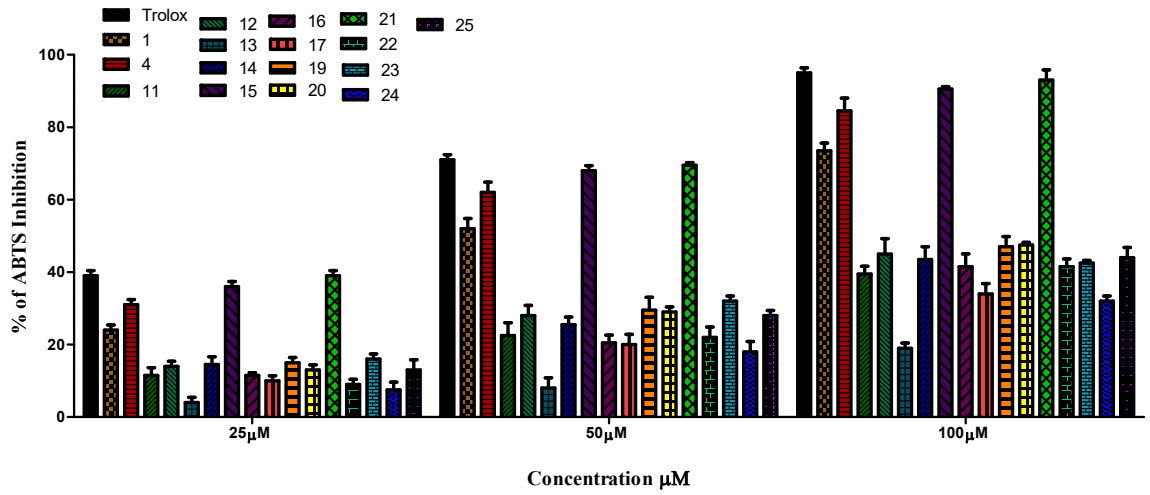

Fig.S43. ABTS free radical scavenging activity of compounds (**1, 4, 11, 12, 13, 14, 15, 16, 17, 19, 20, 21, 22, 23, 24** and **25**), was compared with control Trolox. The single asterisk (*) denotes the significant difference between the control and treatment group (25 µM, 50 µM, and 100 µM) at $p < 0.05$ level by one-way ANOVA followed by Duncan's multiple range test. The experiments were performed in triplicates and the values were provided in mean ± SD.

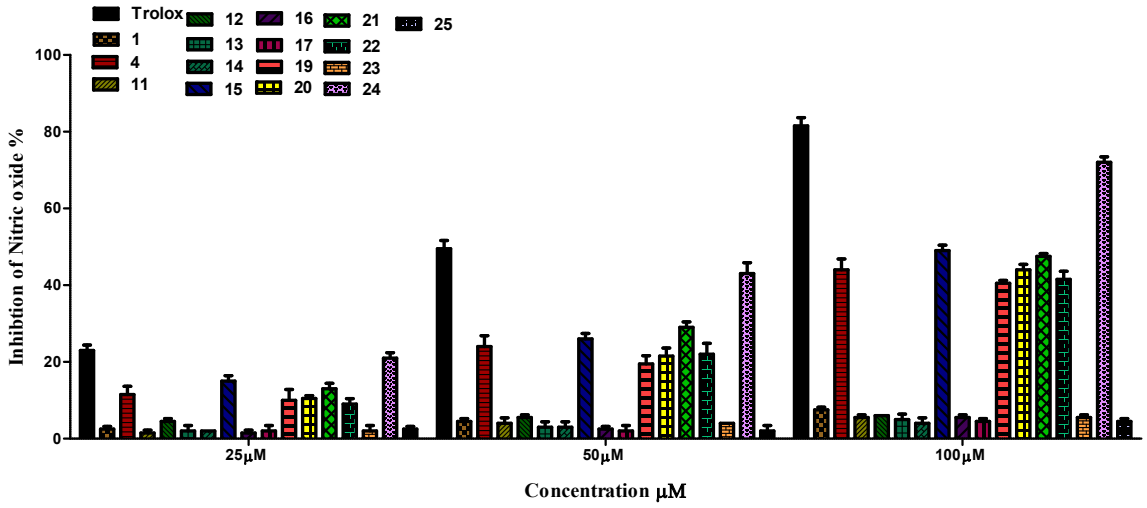

Fig.S44. NO free radical scavenging activity of compounds (**1, 4, 11, 12, 13, 14, 15, 16, 17, 19, 20, 21, 22, 23, 24** and **25**) was compared with control Trolox. The single asterisk (*) denotes the significant difference between the control and treatment group (25 µM, 50 µM, and 100 µM) at $p < 0.05$ level by one-way ANOVA followed by Duncan's multiple range test. The experiments were performed in triplicates and the values were provided in mean ± SD.