Supporting Information

Intercepting Bacterial Indole Signaling with Flustramine Derivatives.

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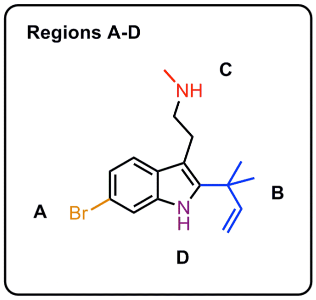
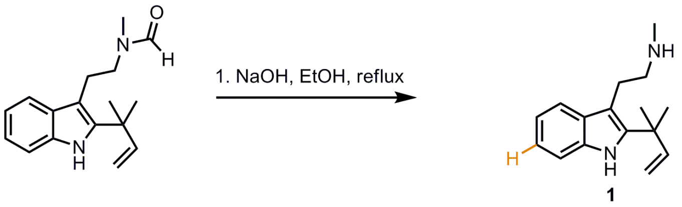
Synthesis: All reagents used for chemical synthesis were purchased from commercially available sources and used without further purification. Chromatography was performed using 60 mesh standard silica gel from Sorbtech. NMR solvents were obtained from Cambridge Isotope Labs and were used as is. ^{1}HNMR (300 MHz or 400 MHz) and ^{13}C NMR (75 MHz or 100 MHz) spectra were recorded at 25 °C on Varian Mercury spectrometers. Chemical shifts (δ) are given in ppm relative to the respective NMR solvents; coupling constants (*J*) are in hertz (Hz). Abbreviations used are s = singlet, bs = broad singlet, d = doublet, dd= doublet of doublets, t = triplet, td = triplet of doublets, m = multiplet, d_{ab} = ab doublet, dd_{ab} = ab doublet of doublet. Mass spectra were obtained at the NCSU Department of Chemistry Mass Spectrometry Facility. Infrared spectra were obtained on a FT/IR-4100 spectrophotometer (ν_{max} in cm^{-1}). UV absorbance was recorded on a G*enesys 10* scanning UV/visible spectrophotometer (λ_{max} in nm).

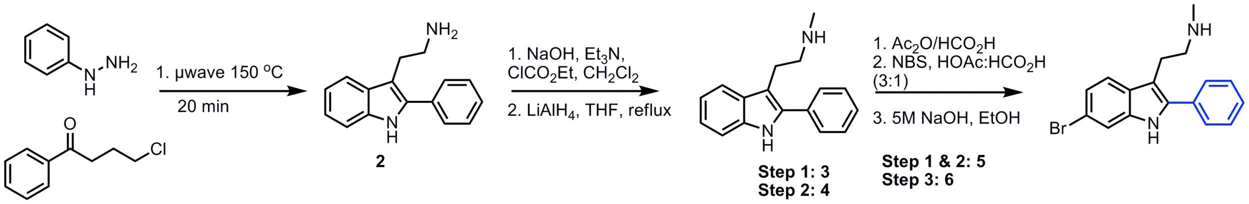
Synthesis of Regions A-D

Region A

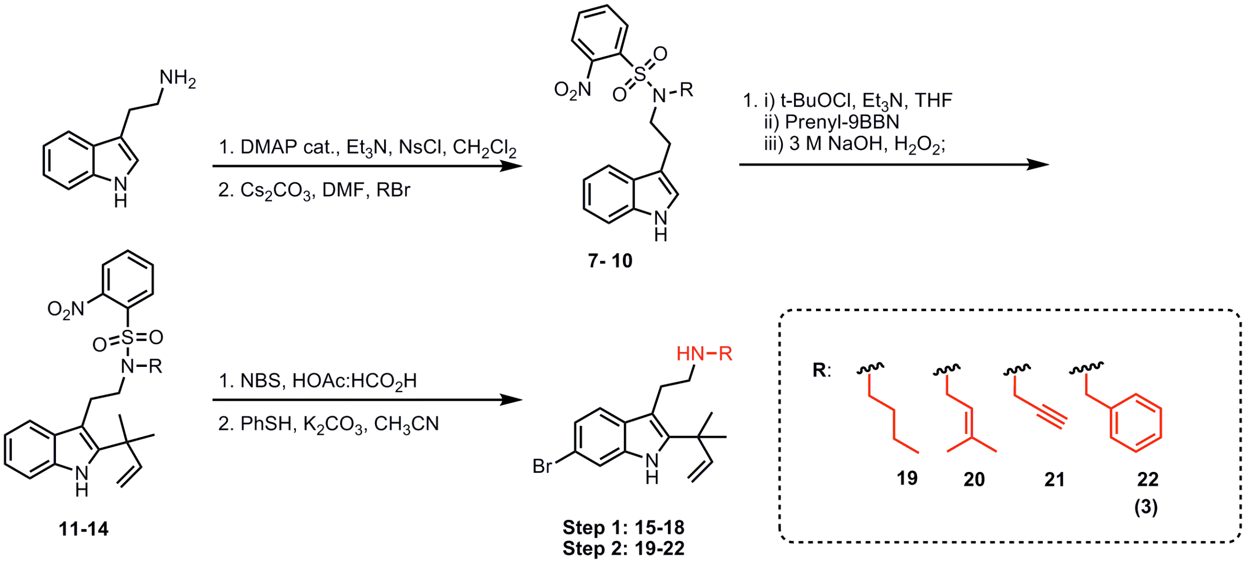




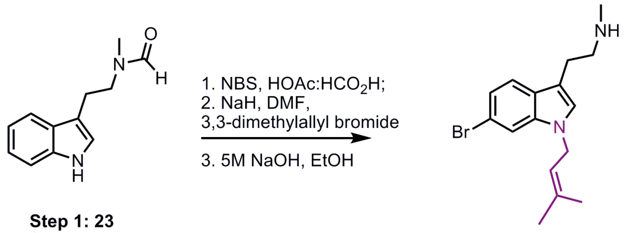
Region B



Region C

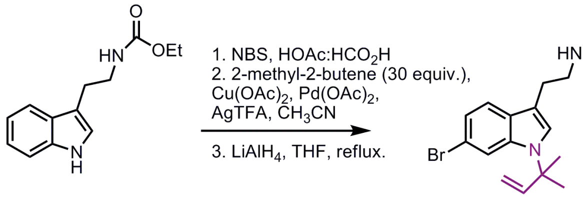


Region D



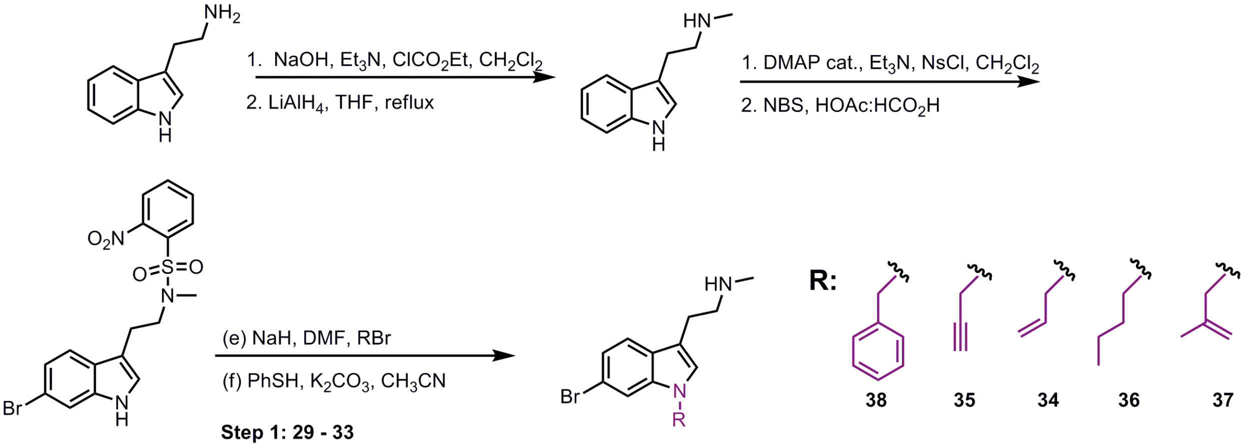
Step 1: 23
Step 2: 24
Step 3: 25

Desformylflustramine



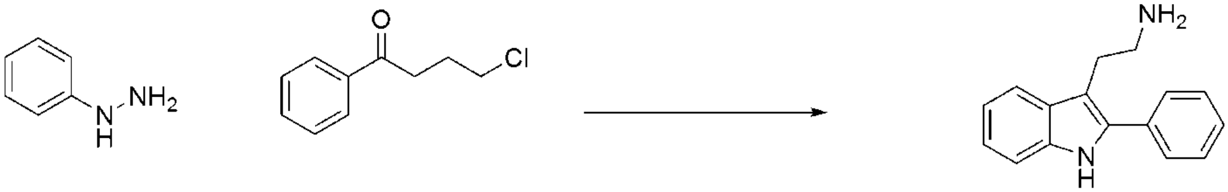
Step 1: 26
Step 2: 27
Step 3: 28 (4)

(4)

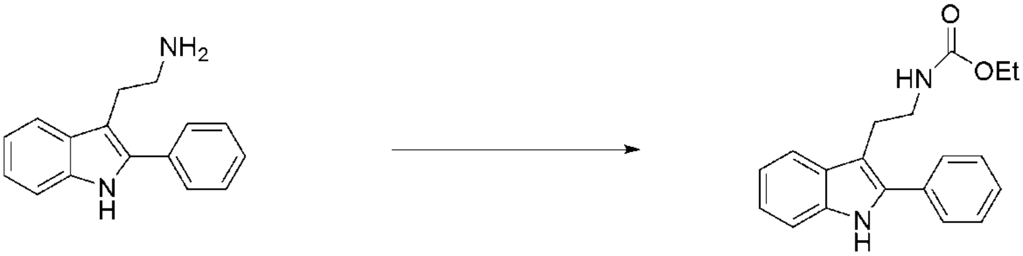


Step 1: 29 - 33
Step 2: 34 - 38

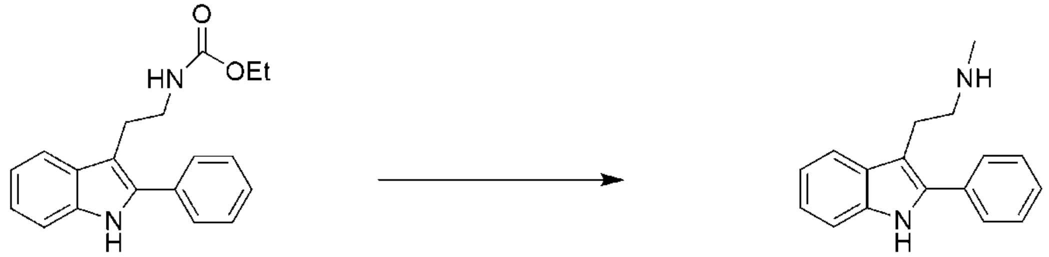
Synthesis and characterization for novel compounds



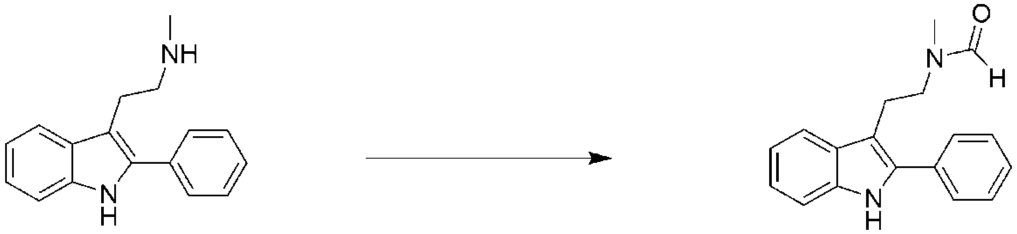
SI-2: was prepared by similar synthetic methods to those developed by Slade et al.[1,2] to afford the product SI-2 (170 mg, 20% yield) as a dark green foam. ^{1}H NMR (300 MHz, CDCl$_3$) δ 8.42 (bs, 1H), 7.64 (d, 1H, J = 6.0 Hz), 7.60-7.57 (m, 2H), 7.48-7.43 (m, 2H), 7.40-7.34 (m, 2H), 7.26-7.20, (m, 1H), 7.20-7.12 (m, 1H), 3.06 (s, 4H); ^{13}C NMR (75 MHz, CDCl$_3$) δ 136.1, 135.5, 133.4, 129.4, 129.0, 128.3, 127.8, 122.4, 119.7, 119.3, 111.1, 110.6, 34.1, 29.0; HRMS (ESI) calcd for $C_{16}H_{17}N_2$ (M$^+$H)+ 237.1383, found 237.1383; IR ν_{max}/cm^{-1}3406, 1637; λ_{max}: 226 nm, 302 nm.



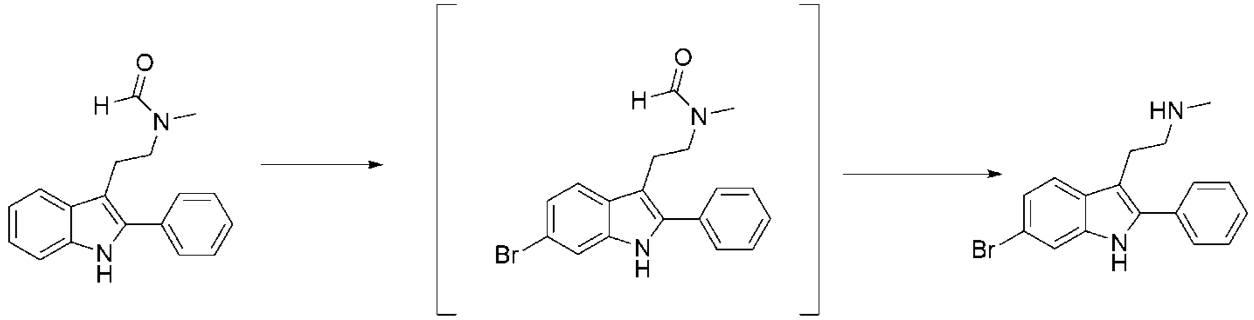
SI-3: was prepared by similar synthetic methods to those developed by Sutcliffe.[3] The crude residue was purified by flash chromatography (10-20% Acetone/Hexanes) to afford the product SI-3 (170 mg, 65% yield) as a brown oil. ^{1}H NMR (300 MHz, CDCl$_3$) δ 8.37 (bs, 1H), 7.67 (d, 1H, *J* = 7.8 Hz), 7.56-7.54 (m, 2H), 7.48-7.42 (m, 2H), 7.39-7.36 (m, 2H), 7.23-7.14, (m, 2H), 4.76 (bs, 1H), 4.09-4.03 (m, 2H), 3.50-3.48 (m, 2H), 3.12 (t, 2H, *J* = 6.6 Hz), 1.20 (t, 2H, *J* = 6.9 Hz); ^{13}C NMR (75 MHz, CDCl$_3$) δ 156.9, 136.1, 135.1, 135.6, 133.1, 129.1, 129.0, 128.2, 127.9, 122.5, 119.9, 111.1, 109.7, 60.8, 41.6, 25.3, 14.8; HRMS (ESI) calcd for $C_{19}H_{20}N_2NaO_2$ (M$^+$Na)+ 331.1417, found 331.1416; IR ν_{max}/cm^{-1}3377, 2937, 1679, 1406, 756; λ_{max}: 250 nm, 297 nm.



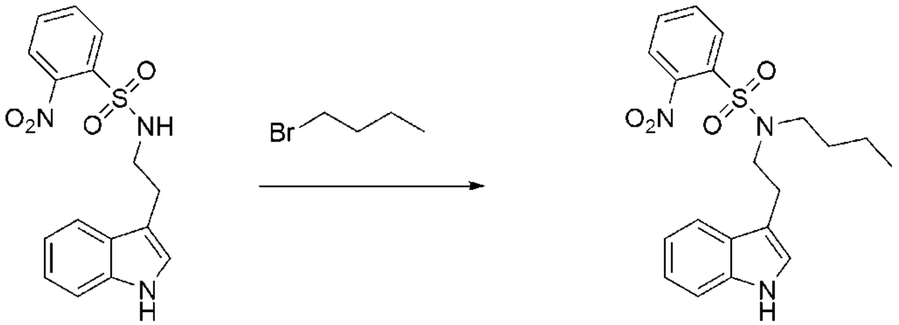
SI-4: was prepared by similar synthetic methods to those developed by Sutcliffe.[3] The crude residue was purified by flash chromatography (5-15% sat. NH_3-MeOH/CH_2Cl_2) to afford the product SI-4 (113 mg, 50% yield) as a brown oil. ^{1}H NMR (300 MHz, CDCl$_3$) δ 8.56 (bs, 1H), 7.67 (d, 1H, *J* = 7.5 Hz), 7.58-7.55 (m, 2H), 7.47-7.42 (m, 2H), 7.38-7.35 (m, 2H), 7.22-7.12, (m, 2H), 3.13 (t, 2H, J = 7.5 Hz), 2.95 (t, 2H, J = 7.2 Hz), 2.40 (s, 3H); ^{13}C NMR (75 MHz, CDCl$_3$) δ 136.1, 135.3, 133.3, 129.4, 133.1, 129.1, 128.2, 127.9, 122.5, 119.9, 119.4, 111.1, 110.9, 52.7, 36.5, 25.1; HRMS (ESI) calcd for $C_{17}H_{19}N_2$ (M$^+$H)+ 251.1543, found 251.1545; IR ν_{max}/cm^{-1} 3400, 2937, 1511; λ_{max}: 242 nm, 302 nm.



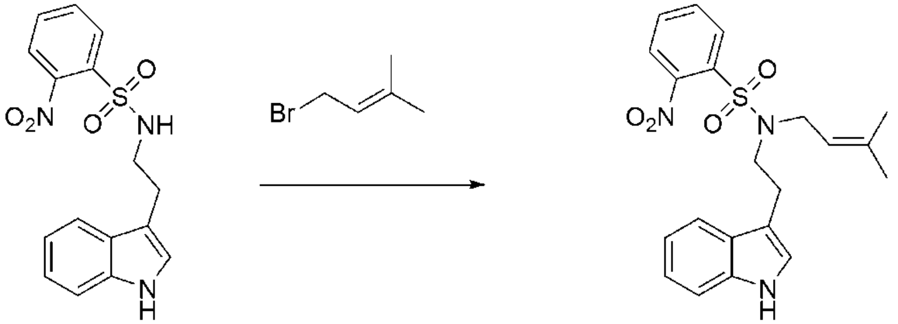
SI-5 was prepared by similar synthetic methods to those developed by Lindell.[4] The crude residue was purified by flash chromatography (50-60% EtOAc:Hexanes) to afford the product SI-5 (51 mg, 74% yield) as a brown oil. ^{1}H NMR (300 MHz, CDCl$_3$) δ 8.69 (bs, 1H), 7.61 (m, 2H), 7.43-7.29 (m, 5H), 7.23-7.15 (m, 2H), 3.46 (t, 2H, J = 6.6 Hz), 3.15 (t, 2H, J = 6.6 Hz), 2.61 (s, 3H); ^{13}C NMR (75 MHz, CDCl$_3$) δ 162.7, 136.4, 133.0, 129.1, 129.1, 128.4, 127.9 128.0, 122.6, 119.1, 118.4, 111.6, 109.7, 49.9 (45.5), 29.7 (35.3), 23.7 (22.5); HRMS (ESI) calcd for $C_{18}H_{19}N_2O$ (M$^+$H)+ 279.1493, found 279.1493; IR ν_{max}/cm^{-1} 2923, 2874, 1686, 1504; λ_{max}: 240 nm, 300 nm.



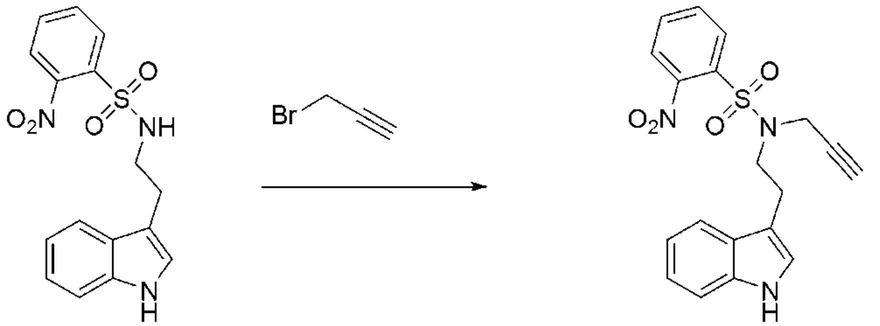
SI-6: was prepared by similar synthetic methods to those developed by Lindel.[4] The crude residue was purified by flash chromatography (5-15% sat. NH_3-MeOH/CH_2Cl_2) to afford the product SI-6 (10 mg, 92% yield) as a brown oil. ^{1}H NMR (300 MHz, CD3OD) δ 7.64-7.7.58 (m, 2H,), 7.52-7.46 (m, 3H), 7.40-7.36 (m, 2H), 7.22-7.18 (m, 1H), 3.12-3.-5 (m, 2H), 2.97-2.90 (m, 2H), 2.42 (s, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 136.8, 135.9, 132.7, 129.3, 128.2, 123.4, 123.2, 121.9, 120.6, 116.0, 113.9, 112.5, 110.7, 52.4, 36.2, 24.7; HRMS (ESI) calcd for $C_{17}H_{18}N_2Br$ $(M^+H)+$ 329.0648, found 329.0649; IR ν_{max}/cm^{-1} 3405, 1532, ; λ_{max}: 237 nm, 308 nm.



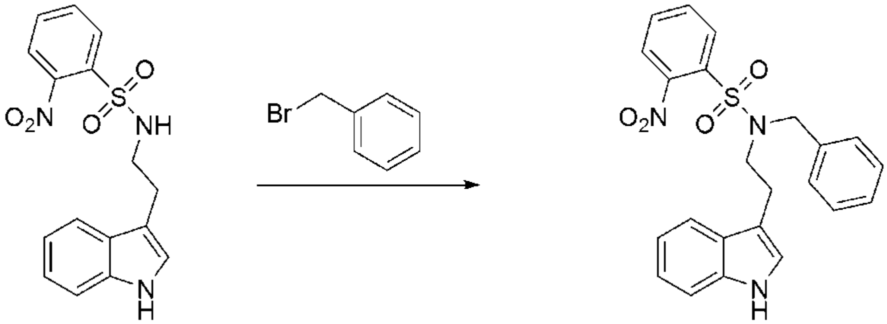
SI-7 was prepared by similar synthetic methods developed by Bowman.[5] **SI- 7** was prepared and purified to provide SI-19 (512 mg, 92% yield), as a yellow oil. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.99 (br s, 1H), 7.91 (d, 1H, *J* = 7.5 Hz), 7.61-7.50 (m, 4H), 7.35-7.03 (m, 5H), 3.61 (t, 2H, *J* = 7.8 Hz), 3.43 (t, 2H, *J* = 7.5 Hz), 3.04 (t, 2H, *J* = 7.2 Hz), 1.61 (quintet, 2H, *J* = 7.5 Hz), 1.37-1.28 (m, 2H), 0.92 (t, 3H, *J* = 6.0 Hz). ^{13}C NMR (100 MHz, $CDCl_3$) δ 136.7, 133.6, 131.9, 131.0, 127.6, 124.5, 122.8, 122.6, 120.0, 118.9, 112.6, 111.8, 48.1, 47.9, 30.8, 25.1, 20.3, 14.2. HRMS (ESI) calcd for $C_{20}H_{23}N_3O_4SNa$ $(M^+Na)+$ 424.1301, found 424.129; IR ν_{max}/cm^{-1} 3405, 2923, 1532, 1350, 1148, 749; λ_{max}: 236 nm, 278 nm.



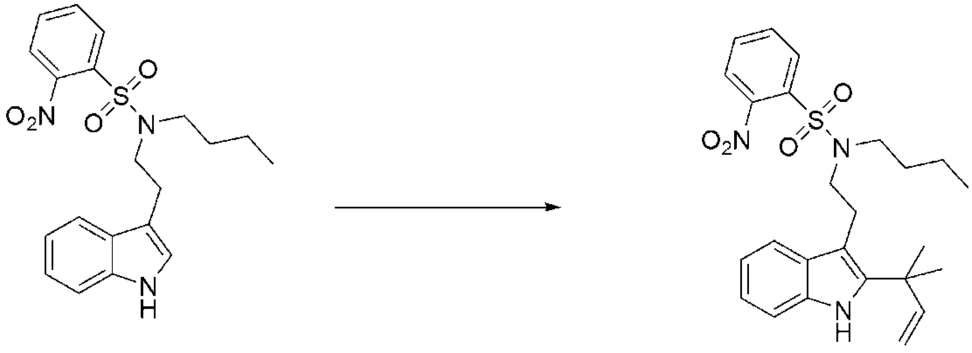
SI-8 was prepared and purified in a similar manner to **SI-7** to provide **SI-8** (595 mg, 99% yield), as a yellow oil. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.99 (br s, 1H), 7.94 (d, 1H, *J* = 8.7 Hz), 7.62-7.51 (m, 4H), 7.35-7.32 (m, 1H), 7.22-7.08 (m, 2H), 7.02 (d, 1H, *J* = 2.4 Hz), 5.14 (tt, 1H, *J* = 7.2 Hz, *J* = 1.2 Hz), 4.02 (d, 1H, *J* = 7.2 Hz), 3.56 (t, 2H, *J* = 7.5 Hz), 3.01 (t, 2H, *J* = 7.5 Hz), 1.71 (s, 3H), 1.65 (s, 3H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 148.1, 137.9, 136.4, 134.0, 133.4, 131.7, 130.8, 127.4, 124.3, 122.6, 122.3, 119.6, 119.3, 118.7, 112.4, 111.5, 47.7, 45.6, 26.1, 24.9, 18.1. HRMS (ESI) calcd for $C_{21}H_{24}N_3O_4S$ $(M^+H)+$ 414.1482, found 414.1409; IR ν_{max}/cm^{-1} 3426, 2923, 1539, 1448, 1336, 1148, 749, 574; λ_{max}: 246 nm, 281 nm.



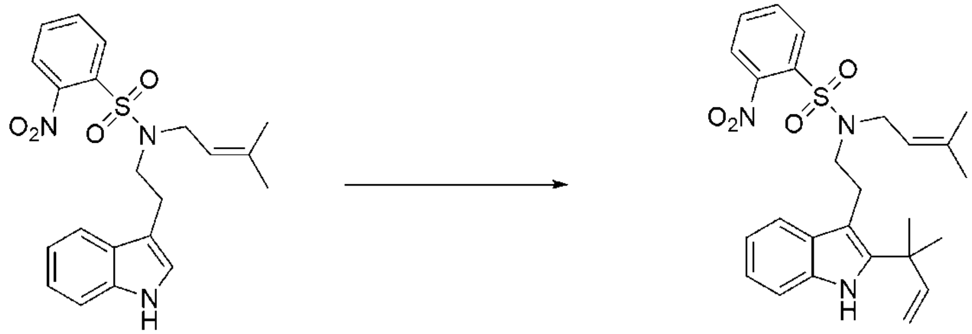
SI-9 was prepared and purified in a similar manner to **SI-7** to provide **SI-9** (415 mg, 75% yield), as a yellow oil. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.99 (br s, 1H), 7.93 (d, 1H, J = 8.1 Hz), 7.59-7.50 (m, 4H), 7.34-7.05 (m, 5H), 4.29 (s, 2H), 3.75 (t, 2H, J = 7.2 Hz), 3.09 (t, 2H, J = 7.8 Hz), 2.25 (t, 1H, J = 2.4 Hz). ^{13}C NMR (100 MHz, $CDCl_3$) δ 136.7, 133.9, 133.2, 132.1, 131.1, 127.6, 124.6, 122.9, 122.6, 120.0, 119.0, 112.2, 111.8, 74.5, 47.7, 37.2, 24.5. HRMS (ESI) calcd for $C_{19}H_{17}N_3NaO_4S$ $(M^+Na)+$ 406.0832, found 406.0821; IR ν_{max}/cm^{-1} 3412, 3272, 1909, 1539, 1357, 1161, 581; λ_{max}: 246 nm, 280 nm.



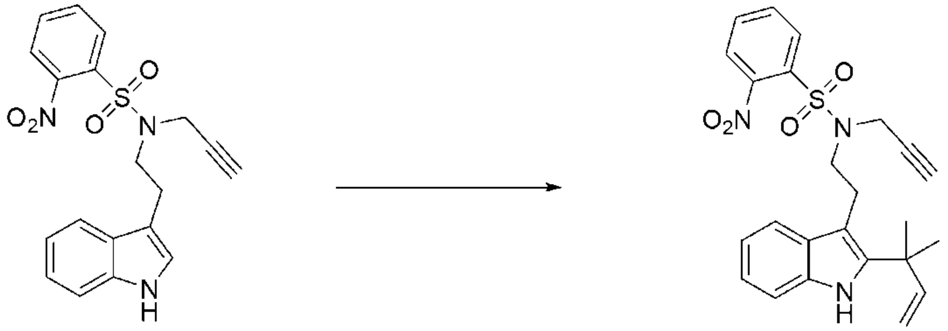
SI-10:was prepared and purified in a similar manner to **SI-7** to provide **SI-10** (272 mg, 72% yield) as a yellow oil. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.90-7.7.87 (m, 2H), 7.60-7.57 (m, 2H), 7.48-7.45 (m, 1H), 7.36-7.30 (m, 3H), 7.28-7.25 (m, 2H), 7.14 (dt, 2H, J = 7.1 Hz, J = 0.9 Hz), 7.03 (dt, 2H, J = 7.8 Hz, J = 1.2 Hz), 6.90 (d, 1H, J = 7.2 Hz) 4.64 (s, 2H), 3.52 (t, 2H, J = 8.1 Hz), 2.86 (t, 2H, J = 7.6 Hz); ^{13}C NMR (100 MHz, $CDCl_3$) δ 136.3, 135.9, 133.9, 133.4, 131.8, 130.9, 129.0, 128.7, 128.3, 127.2, 124.3, 122.5, 122.3, 119.6, 118.7, 112.2, 111.4, 51.7, 47.8, 24.3; HRMS (ESI) calcd for $C_{23}H_{21}N_3NaO_4S$ $(M^+Na)+$ 458.1145, found 458.1135; IR ν_{max}/cm^{-1} 3426, 2930, 1539, 1441, 1357, 1148, 742; λ_{max}: 242 nm, 278 nm.



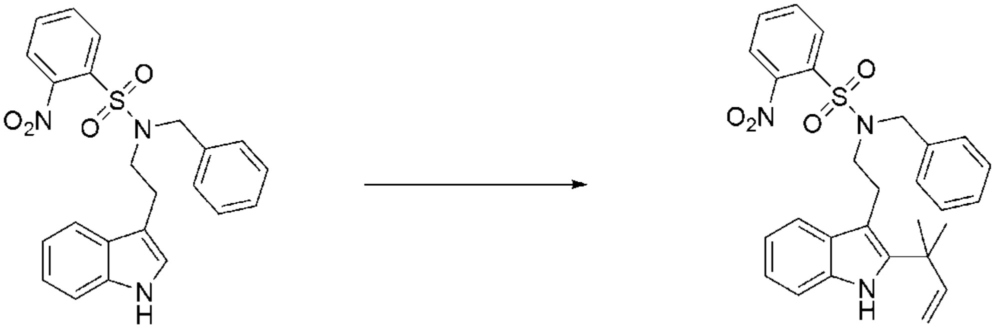
SI-11: was prepared by similar synthetic methods developed by Lindel.[4] The crude residue was purified by flash chromatography (5-20% EtOAc:Hexanes) to afford the product **SI-11** (100 mg, 53% yield) as a yellow oil. ^{1}H NMR (300 MHz, CDCl$_3$) δ 8.03-8.01 (m, 1H), 7.88 (br s, 1H), 7.70-7.61 (m, 3H), 7.50 (d, 1H, *J* = 7.5 Hz), 7.29 (d, 1H, *J* = 7.2 Hz), 7.14-7.08 (m, 2H), 6.10 (dd, 1H, *J* = 17.4 Hz, *J* = 10.2 Hz), 5.17 (dd, 1H, *J* = 17.2 Hz, *J* = 0.9 Hz), 5.14 (dd, 1H, *J* = 10.2 Hz, *J* = 0.9 Hz), 3.47-3.40 (m, 4H), 3.16-3.10 (m, 2H), 1.67-1.59 (m, 2H),1.56 (s, 6H), 1.40-1.26 (m, 2H), 0.91 (t, 1H, *J* = 7.2 Hz). ^{13}C NMR (100 MHz, CDCl$_3$) δ 148.2, 145.9, 140.3, 134.2, 134.1, 133.4, 131.7, 130.9, 129.5, 124.4, 121.8, 119.7, 118.1, 112.3, 110.7, 107.1, 48.3, 39.0, 31.1, 27.8, 25.2, 20.1, 14.0. HRMS (ESI) calcd for $C_{25}H_{31}N_3O_4S$ (M^+Na)+ 492.1927, found 492.1910; IR ν_{max}/cm^{-1} 3433, 2958, 1539, 1469, 1329, 1168, 742; λ_{max}: 233 nm, 280 nm.



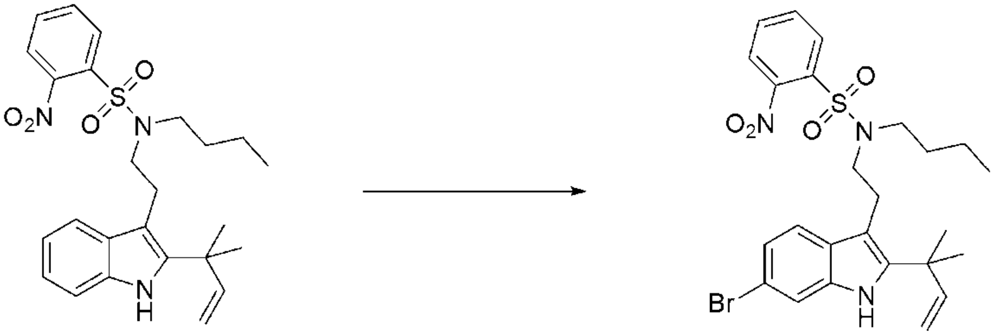
SI-12 was prepared and purified in a similar manner to **SI-11** to provide **SI-12** (450 mg, 65% yield), as a clear oil. ^{1}H NMR (300 MHz, CDCl$_3$) δ 8.03-8.01 (m, 1H), 7.87 (br s, 1H), 7.66-7.60 (m, 3H), 7.59 (d, 1H, *J* = 7.5 Hz), 7.28 (d, 1H, *J* = 7.5 Hz), 7.16-7.04 (m, 2H), 6.10 (dd, 1H, *J* = 17.4 Hz, *J* = 10.2 Hz), 5.25-5.18 (m, 1H), 5.15 (dd, 1H, *J* = 17.4 Hz, *J* = 0.9 Hz), 5.14 (dd, 1H, *J* = 10.2 Hz, *J* = 0.9 Hz), 4.06 (d, 2H, J = 6.6 Hz), 3.45-3.40 (m, 2H), 3.18-3.12 (m, 2H), 1.75 (s, 3H), 1.70 (s, 3H), 1.52 (s, 6H). ^{13}C NMR (75 MHz, CDCl$_3$) δ 148.3, 145.8, 140.4, 137.4, 134.2, 134.1, 133.4, 131.7, 131.0, 129.7, 124.4, 121.7, 119.9, 119.6, 118.2, 112.6, 110.7, 107.1, 48.0, 46.1, 39.0, 27.8, 26.1, 25.4, 18.2. HRMS (ESI) calcd for $C_{26}H_{31}N_3O_4S$ (M^+H)+ 482.2108, found 482.2100; IR ν_{max}/cm^{-1}3412, 2329, 1539, 1453, 1148, 749; λ_{max}: 237 nm, 285 nm.



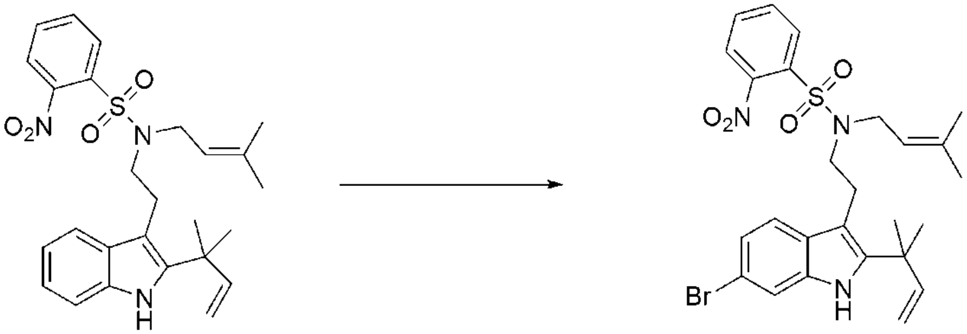
SI-13 was prepared and purified in a similar manner to **SI-11** to provide **SI-13** (267 mg, 55% yield), as a clear oil. ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.03-8.00 (m, 1H), 7.91 (br s, 1H), 7.68-7.61 (m, 3H), 7.55 (d, 1H, *J* = 7.8 Hz), 7.29 (d, 1H, *J* = 7.2 Hz), 7.17-7.06 (m, 2H), 6.13 (dd, 1H, *J* = 17.4 Hz, *J* = 10.8 Hz), 5.14 (d, 1H, *J* = 17.4 Hz, *J* = 0.6 Hz), 5.10 (d, 1H, *J* = 10.8 Hz, *J* = 0.6 Hz), 4.34 (d, 2H, *J* = 2.1 Hz), 3.62-3.57 (m, 2H), 3.22-3.17 (m, 2H), 2.27 (t, 1H, *J* = 2.7 Hz),1.57 (s, 6H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 148.2, 145.8, 140.5, 134.2, 133.8, 133.2, 131.9, 130.8, 129.6, 124.6, 121.8, 119.8, 118.2, 112.6, 110.7, 107.8, 74.2, 47.4, 39.1, 31.0, 27.9, 24.4. HRMS (ESI) calcd for $C_{24}H_{25}N_3O_4S$ (M^+Na)+ 474.1458, found 474.1447; IR ν_{max}/cm^{-1}3433, 2930, 1553, 1448, 1364, 1155, 749; λ_{max}: 233 nm, 283.



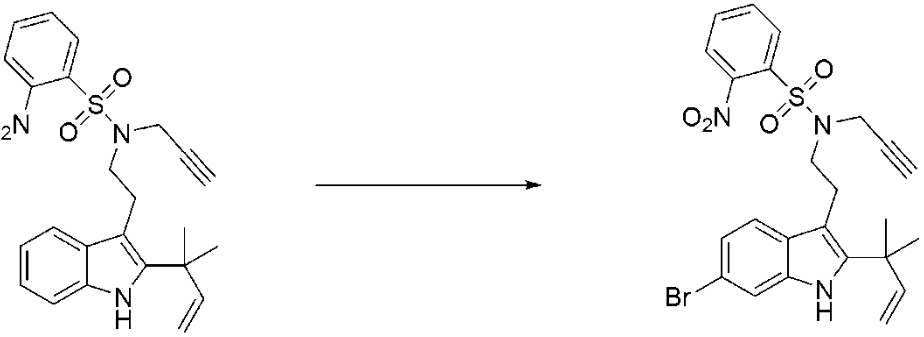
SI-14 was prepared and purified in a similar manner to **SI-11** to provide **SI-14** (205 mg, 59% yield), as a yellow oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.04 (d, 1H, J = 8.4 Hz), 7.80 (br s, 1H), 7.70-7.61 (m, 3H), 7.40-7.30 (m, 5H), 7.22 (d, 1H, *J* = 8.8 Hz), 7.10-7.06 (m, 2H), 7.00- 6.95 (m, 1H), 5.97 (dd, 1H, *J* = 17.6 Hz, *J* = 10.4 Hz), 5.04 (dd, 1H, *J* = 17.6 Hz, *J* = 1.0 Hz), 5.03 (dd, 1H, *J* = 10.4 Hz, *J* = 1.0 Hz), 4.67 (s, 2H), 3.43-3.38 (m, 2H), 2.98-2.93 (m, 2H), 1.37 (s, 6H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 145.7, 140.3, 136.2, 134.1, 134.0, 133.6, 131.9, 131.3, 129.5, 128.9, 128.5, 128.3, 124.6, 121.6, 119.5, 118.1, 112.5, 110.6, 106.9, 52.5, 48.4, 38.9, 27.7, 25.0. HRMS (ESI) calcd for $C_{28}H_{29}N_3O_4S$ (M^+H)+ 504.1952, found 504.1944; IR ν_{max}/cm^{-1}3433, 2916, 1546, 1455, 1336, 1161, 749; λ_{max}: 233 nm, 285 nm.



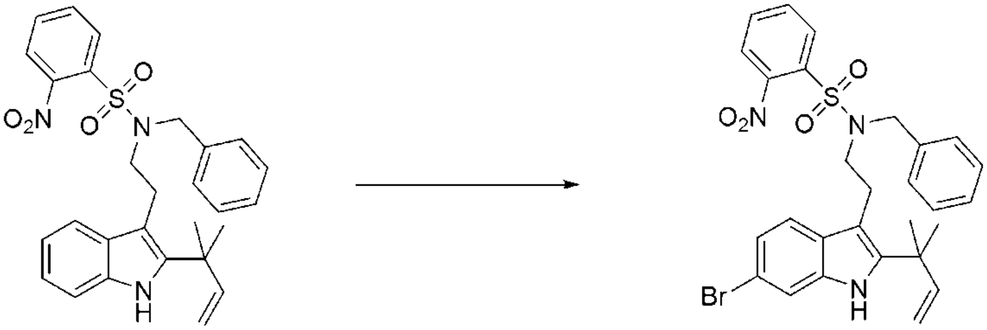
SI-15 was prepared by similar synthetic methods developed by Lindel.[4] The crude residue was purified by flash chromatography (10-30% EtOAc:Hexanes) to afford the product **SI-15** (130 mg, 62% yield) as a yellow oil. ^{1}H NMR (300 MHz, CDCl$_3$) δ 8.02-7.99 (m, 1H), 7.87 (br s, 1H), 7.66-7.61 (m, 3H), 7.43 (d, 1H, J = 1.2 Hz), 7.32 (d, 1H, J = 8.4 Hz), 7.18 (dd, 1H, J = 8.4 Hz, J = 1.2 Hz), 5.94 (dd, 1H, J = 17.4 Hz, J = 10.2 Hz), 5.05 (d, 1H, J = 17.4 Hz), 5.04 (d, 1H, J = 10.8 Hz), 3.39 (t, 4H, J = 8.1 Hz), 3.14-3.08 (m, 2H), 1.64-1.58 (m, 2H), 1.54 (s, 6H), 1.37-1.30 (m, 2H), 0.92 (t, 3H, J = 7.5 Hz). ^{13}C NMR (100 MHz, CDCl$_3$) δ 148.2, 145.6, 141.0, 135.1, 133.9, 133.6, 131.8, 130.7, 128.5, 124.4, 122.9, 119.4, 115.0, 113.7, 112.5, 107.4, 48.2, 40.0, 31.0, 27.8, 27.7, 25.2, 20.1, 13.9. HRMS (ESI) calcd for $C_{25}H_{30}N_3O_4SBr$ (M^+Na)+ 570.1033, found 570.1022; IR ν_{max}/cm^{-1} 3433, 2916, 1539, 1455, 1350, 1161; λ_{max}: 233 nm, 285 nm.



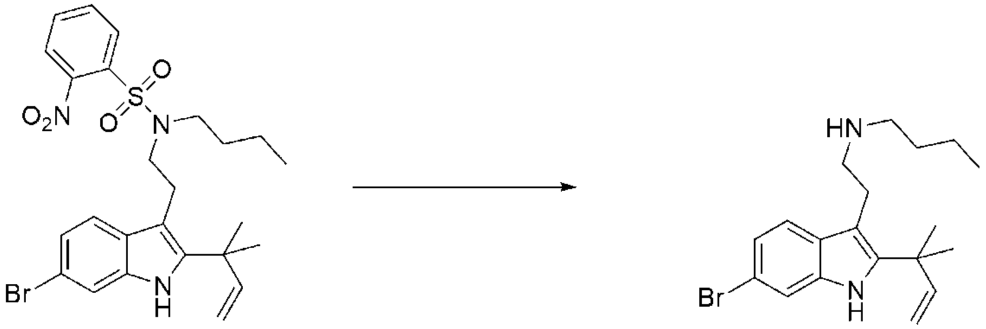
SI-16 was prepared and purified in a similar manner to **SI-15** to provide **SI-16** (105 mg, 53% yield), as a clear oil. ^{1}H NMR (300 MHz, CDCl$_3$) δ 8.02-8.00 (m, 1H), 7.88 (s, 1H), 7.69-7.60 (m, 3H), 7.42 (d, 1H, J = 1.8 Hz), 7.40 (d, 1H, J = 8.4 Hz), 7.18 (dd, 1H, J = 8.7 Hz, J = 1.8 Hz), 6.09 (dd, 1H, J = 17.4 Hz, J = 10.5 Hz), 5.18 (dd, 1H, J = 17.4 Hz, J = 0.9 Hz), 5.17 (dd, 1H, J = 10.5 Hz, J = 0.9 Hz), 4.30 (d, 2H, J = 2.4 Hz), 3.577-3.52 (m, 2H,), 3.18-3.13 (m, 2H), 2.26 (t, 1H, J = 2.4 Hz), 1.52 (s, 6H). ^{13}C NMR (75 MHz, CDCl$_3$) δ 148.0, 145.4, 141.1, 135.1, 133.8, 133.1, 131.9, 130.8, 128.5, 124.6, 123.1, 119.5, 115.2, 113.7, 112.8, 107.1, 74.3, 47.3, 39.1, 37.1, 27.8, 24.3. HRMS (ESI) calcd for $C_{26}H_{30}N_3O_4S$ (M^+Na)+ 582.1033, found 582.1020; IR ν_{max}/cm^{-1}3433, 3240, 1554, 1448, 1364, 1155; λ_{max}: 231 nm, 283 nm.



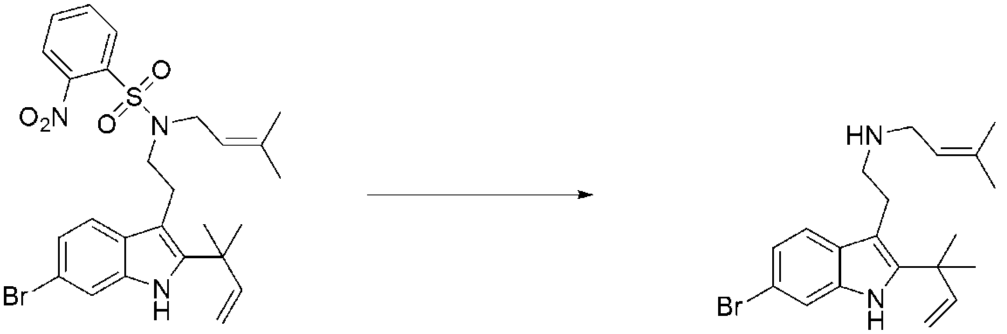
SI-17 was prepared and purified in a similar manner to **SI-15** to provide **SI-17** (32 mg, 66% yield), as a yellow oil. ^{1}H NMR (300 MHz, CDCl$_3$) δ 8.02-8.00 (m, 1H), 7.88 (s, 1H), 7.69-7.60 (m, 3H), 7.42 (d, 1H, *J* = 1.8 Hz), 7.40 (d, 1H, J = 8.4 Hz), 7.18 (dd, 1H, *J* = 8.7 Hz, *J* = 1.8 Hz), 6.09 (dd, 1H, *J* = 17.4 Hz, *J* = 10.5 Hz), 5.18 (dd, 1H, *J* = 17.4 Hz, J = 0.9 Hz), 5.17 (dd, 1H, *J* = 10.5 Hz, *J* = 0.9 Hz), 4.30 (d, 2H, *J* = 2.4 Hz), 3.577-3.52 (m, 2H,), 3.18-3.13 (m, 2H), 2.26 (t, 1H, *J* = 2.4 Hz), 1.52 (s, 6H). ^{13}C NMR (75 MHz, CDCl$_3$) δ 148.0, 145.4, 141.1, 135.1, 133.8, 133.1, 131.9, 130.8, 128.5, 124.6, 123.1, 119.5, 115.2, 113.7, 112.8, 107.1, 74.3, 47.3, 39.1, 37.1, 27.8, 24.3. HRMS (ESI) calcd for $C_{24}H_{24}N_3O_4SBr$ $(M^+H)+$ 530.0744, found 530.0733; IR ν_{max}/cm^{-1} 3433, 3287, 2916, 1525, 1344, 1148; λ_{max}: 230 nm, 281 nm.



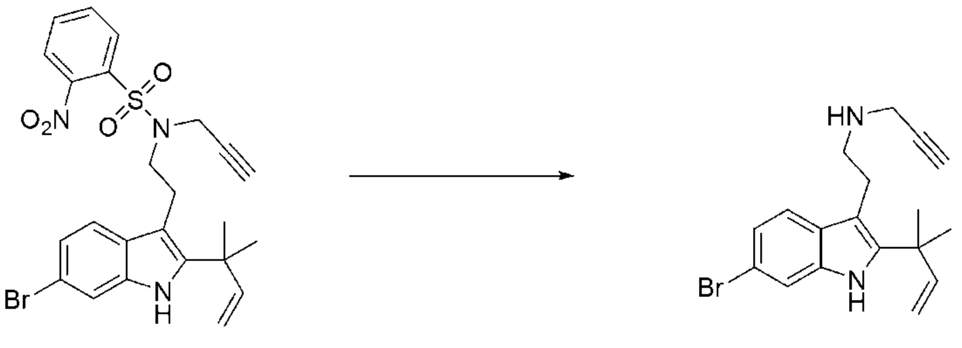
SI-18 was prepared and purified in a similar manner to **SI-15** to provide **SI-18** (117 mg, 49% yield), as a yellow oil. ^{1}H NMR (300 MHz, CDCl$_3$) δ 8.02 (d, 1H, J = 8.4 Hz), 7.77 (s, 1H), 7.70-7.62 (m, 3H), 7.36-7.33 (m, 6H), 7.05 (dd, 1H, *J* = 8.4 Hz, *J* = 1.5 Hz), 6.88 (d, 1H, J = 8.4 Hz), 5.94 (dd, 1H, J = 17.4 Hz, J = 10.8 Hz), 5.17 (dd, 1H, *J* = 17.4 Hz, *J* = 1.0 Hz), 5.14 (dd, 1H, *J* = 10.8 Hz, *J* = 1.0 Hz), 4.63 (s, 2H), 3.39-3.33 (m, 2H), 2.95-2.89 (m, 2H), 1.35 (s, 6H). ^{13}C NMR (75 MHz, CDCl$_3$) δ 148.1, 145.4, 140.9, 136.1, 134.9, 133.8, 133.7, 131.9, 131.2, 129.3, 129.1, 128.6, 128.4, 128.4, 124.6, 122.8, 119.4, 115.0, 113.5, 112.8, 107.2, 52.6, 48.3, 38.9, 27.8, 25.0. HRMS (ESI) calcd for $C_{28}H_{28}N_3O_4SBr$ $(M^+Na)+$ 604.0876, found 604.0864; IR ν_{max}/cm^{-1}3440, 2357, 1532, 679; λ_{max}: 239 nm, 280 nm.



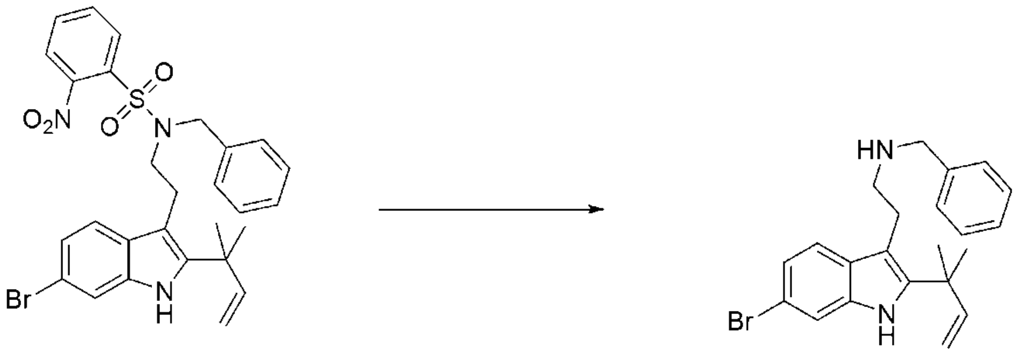
SI-19 (83 mg, 0.151 mmol) was placed in a vial and dissolved in 4 mL of CH_3CN. Then K_2CO_3 (83 mg, 0.604 mmol) was added to the vial, then thiophenol (33 mg, 0.302 mmol) was added dropwise and the reaction was allowed to stir for 16 h at room temperature.[6,7] The reaction was then concentrated under reduced pressure to afford crude product. Purification by flash chromatography (1-10% sat. NH_3-MeOH/CH_2Cl_2) afforded **SI-19** as a light brown oil (43 mg, 78% yield). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.83 (s, 1H), 7.42 (d, 1H, J = 1.2 Hz), 7.41 (d, 1H, J = 8.4 Hz), 7.51 (dd, 2H, J = 8.4 Hz, J = 1.2 Hz), 6.10 (dd, 1H, *J* = 17.2 Hz, *J* = 10.4 Hz), 5.16 (d, 1H, *J* = 17.2 Hz, *J* = 0.8 Hz), 5.15 (d, 1H, *J* = 10.4 Hz, *J* = 0.8 Hz), 3.01 (t, 2H, *J* = 8.4 Hz), 2.85 (t, 2H, *J* = 8.4 Hz), 2.65 (t, 2H, *J* = 7.6 Hz), 1.53 (s, 6H), 1.51-1.45 (m, 2H), 1.36-1.31 (m, 2H), 0.91 (t, 3H, *J* = 7.6 Hz). ^{13}C NMR (75 MHz, $CDCl_3$) δ 145.8, 140.3, 135.1, 128.9, 122.6, 119.8, 114.9, 113.5, 112.5, 109.4, 51.0, 49.9, 39.2, 32.4, 27.8, 25.9, 20.7, 14.2. HRMS (ESI) calcd for $C_{19}H_{27}N_2Br$ $(M^+H)+$ 362.1430, found 362.1420; IR ν_{max}/cm^{-1} 3400, 2944, 1455; λ_{max}: 231 nm, 278 nm.



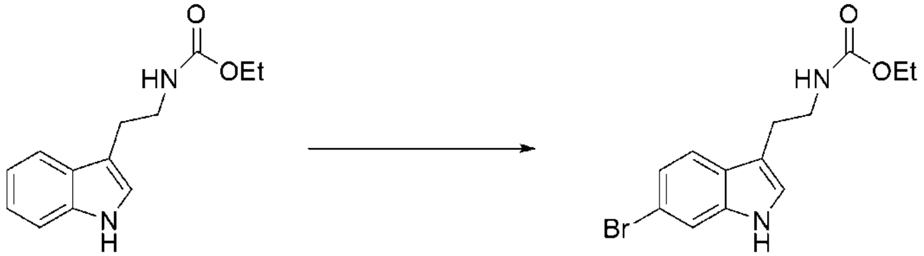
SI-20 was prepared and purified in a similar manner to **SI-19** to provide **SI-20** (35 mg, 50% yield), as a clear oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.84 (s, 1H), 7.42 (d, 1H, *J* = 2.0 Hz), 7.01 (d, 1H, *J* = 8.4 Hz), 7.17 (dd, 1H, *J* = 8.4 Hz, *J* = 2.0 Hz), 6.11 (dd, 1H, *J* = 17.2 Hz, *J* = 10.4 Hz), 5.25 (t, 1H, *J* = 7.2 Hz) 5.16 (dd, 1H, *J* = 17.2 Hz, J = 0.8 Hz), 5.15 (dd, 1H, *J* = 10.4 Hz, *J* = 0.8 Hz), 3.26 (d, 2H, *J* = 7.2 Hz), 3.02-3.00 (m, 2H), 2.87-2.83 (m, 2H), 1.71 (s, 3H), 1.63 (s, 3H), 1.53 (s, 6H). ^{13}C NMR (75 MHz, $CDCl_3$) δ 145.8, 140.3, 135.1, 134.5, 128.9, 123.1, 122.6, 119.8, 114.9, 113.5, 112.5, 109.4, 50.5, 47.5, 39.2, 29.9, 27.8, 26.0, 18.2. HRMS (ESI) calcd for $C_{20}H_{27}N_2Br$ $(M^+H)+$ 375.1430, found 375.1414; IR ν_{max}/cm^{-1} 3272, 2923, 1616, 1462; λ_{max}: 233 nm, 285 nm.



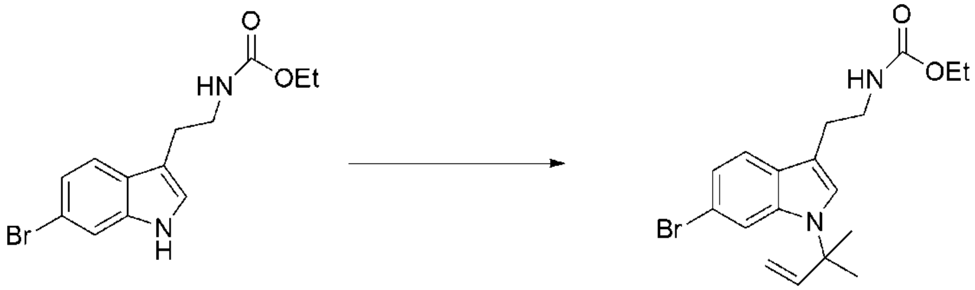
SI-21 was prepared and purified in a similar manner to **SI-19** to provide **SI-21** (32 mg, 71% yield), as a yellow oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.85 (s, 1H), 7.42 (d, 1H, *J* = 2.0 Hz), 7.42 (d, 1H, *J* = 8.4 Hz), 7.16 (dd, 1H, *J* = 8.4 Hz, *J* = 2.0 Hz), 6.11 (dd, 1H, *J* = 17.6 Hz, *J* = 10.0 Hz), 5.18 (dd, 1H, *J* = 4.0 Hz, *J* = 1.2 Hz), 5.15 (dd, 1H, *J* = 2.8 Hz, *J* = 0.8 Hz), 3.50-3.46 (m, 2H), 3.03-2.94 (m, 4H), 2.20 (t, 1H, *J* = 2.8 Hz), 1.53 (s, 6H). ^{13}C NMR (100 MHz, $CDCl_3$) δ 145.8, 140.5, 135.1, 129.0, 124.3, 122.9, 121.2, 119.8, 114.9, 112.6, 112.1, 109.4, 71.6, 49.6, 39.2, 38.3, 27.9, 25.5. HRMS (ESI) calcd for $C_{18}H_{21}N_2Br$ $(M^+H)+$ 345.0961, found 345.0948; IR ν_{max}/cm^{-1} 3300, 2916, 1462; λ_{max}: 233 nm, 287 nm.



SI-22 (3) was prepared and purified in a similar manner to **SI-19** to provide **SI-22 (3)** (26 mg, 62% yield), as a clear oil. ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.83 (s, 1H), 7.41 (br s, 1H), 7.36 (d, 1H, *J* = 8.4 Hz), 7.31-7.29 (m, 5H), 7.15 (d, 1H, *J* = 8.4 Hz), 6.08 (dd, 1H, *J* = 17.2 Hz, *J* = 10.2 Hz), 5.14 (dd, 1H, *J* = 17.2 Hz, *J* = 1.0 Hz), 5.13 (d, 1H, *J* = 10.2 Hz, *J* = 1.0 Hz), 3.83 (s, 2H), 3.02 (t, 2H, *J* = 8.0 Hz), 2.87 (t, 2H, *J* = 8.0 Hz), 1.50 (s, 6H). ^{13}C NMR (75 MHz, CDCl$_3$) δ 145.8, 140.4, 135.1, 128.9, 128.7, 128.6, 128.2, 127.1, 122.6, 119.8, 114.9, 113.5, 112.4, 109.4, 54.1, 50.3, 39.2, 27.8, 25.9. HRMS (ESI) calcd for $C_{22}H_{25}N_2Br$ (M$^+$H)+ 397.1274, found 397.1266; IR ν_{max}/cm^{-1} 3300, 1916, 2357, 1448; λ_{max}: 240 nm, 280 nm.

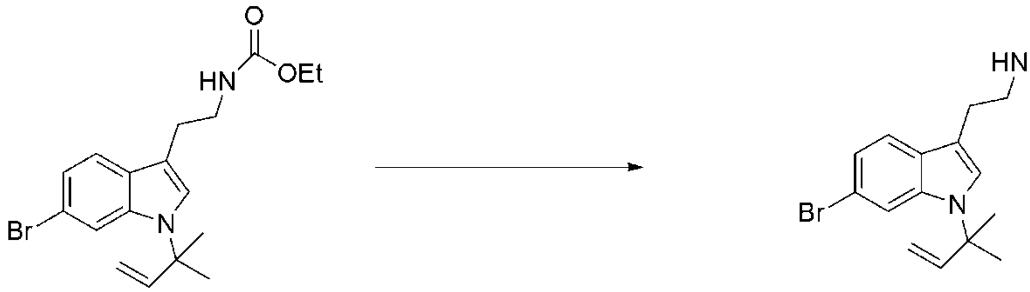


SI-26: was prepared by similar synthetic methods developed by Lindell.[4] The crude residue was purified by flash chromatography (10-50% EtOAc:Hexanes) to afford the product **SI-26** (110 mg, 10% yield) as a brown oil.^{1}H NMR (300 MHz, DMSO-*d*$_6$) δ 11.10 (bs, 1H), 7.68 (s, 1H), 7.32-7.28 (m, 1H), 7.18-7.14 (m, 2H), 4.01-3.94 (m, 2H), 3.20-3.17 (m, 2H), 2.78 (t, 2H, *J* = 7.5 Hz), 1.15 (t, 2H, *J* = 7.2 Hz); ^{13}C NMR (100 MHz, CDCl$_3$) δ 156.9, 143.9, 135.1, 129.3, 124.9, 123.6, 121.4, 112.9, 112.7, 61.0, 41.4, 25.8, 14.8; HRMS (ESI) calcd for $C_{13}H_{16}N_2O_2Br$ (M$^+$H)+ 311.0390, found 311.0381; IR ν_{max}/cm^{-1} 3328, 2923, 1707, 1542, 1252. λ_{max}: 240 nm, 300 nm.

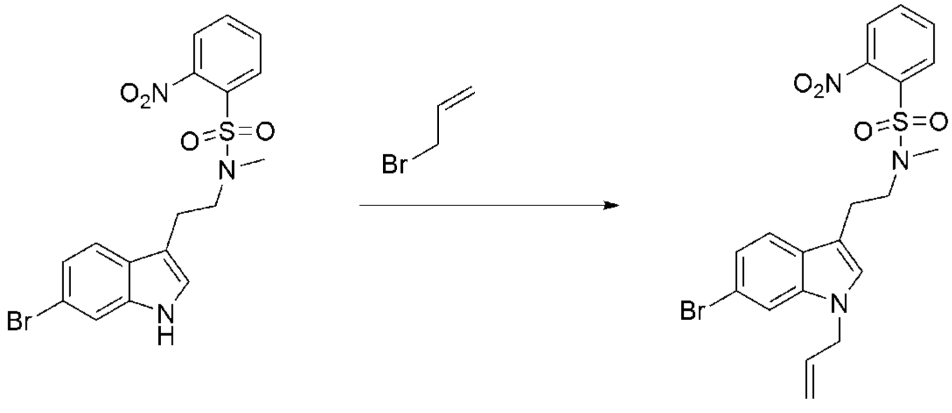


SI-27: was prepared by similar synthetic methods developed by Baran.[8] The crude residue was purified by flash chromatography (10-20% EtOAc:Hexanes) to afford the product **SI-27** (53 mg, 43% yield) as a brown oil.^{1}H NMR (300 MHz, CDCl$_3$) δ 7.68 (d, 1H, *J* = 1.8 Hz), 7.36 (d, 1H, *J* = 9.3 Hz), 7.20-7.13 (m, 1H), 6.09 (dd, 1H, *J* = 17.4, Hz, *J* = 10.5 Hz), 5.19 (dd, 2H, *J* = 21.6, Hz, *J* = 10.8 Hz), 4.17-4.09 (m, 2H), 3.49-3.46 (m, 2H), 2.90 (t, 2H, *J* = 6.9

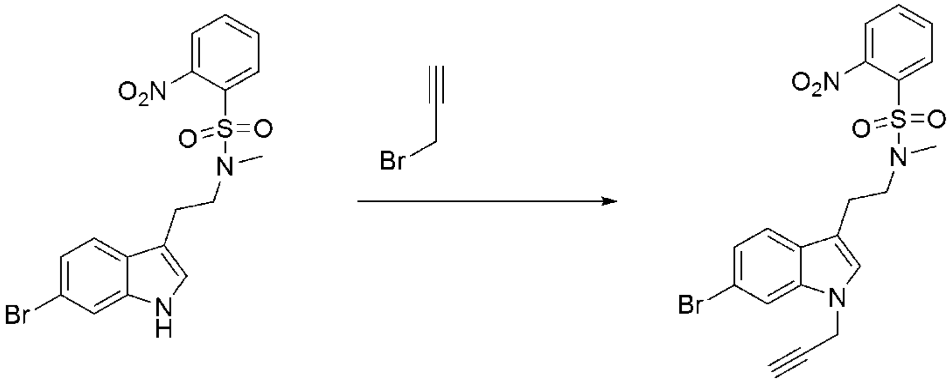
Hz), 1.71 (s, 6H), 1.24 (t, 2H, *J* = 6.9 Hz); ^{13}C NMR (100 MHz, $CDCl_3$) δ 156.8, 143.9, 134.5, 131.2, 124.5, 123.8, 121.5, 115.4, 113.9, 112.5, 110.7, 60.9, 59.3, 41.5, 28.0, 25.9, 14.9; HRMS (ESI) calcd for $C_{18}H_{24}N_2O_2Br$ $(M^{+}H)+$ 379.1016, found 379.1015; IR ν_{max}/cm^{-1} 2909, 1679, 1505; λ_{max}: 245 nm, 297 nm.



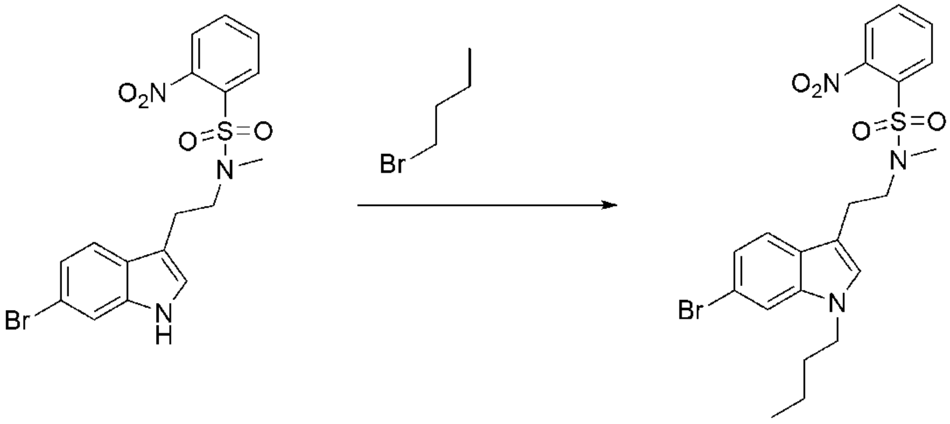
SI-28 (4): was prepared by similar synthetic methods developed by Sutcliffe.[3] The crude residue was purified by flash chromatography (1-10% sat. NH_3-MeOH/CH_2Cl_2) to afford the product **SI-28 (4)** (24 mg, 57% yield) as a brown oil.^{1}H NMR (300 MHz, $CDCl_3$) δ 7.69 (d, 1H, *J* = 2.1 Hz), 7.33 (d, 1H, *J* = 8.7 Hz), 7.17-7.11 (m, 1H), 6.07 (dd, 1H, *J* = 17.4, Hz, *J* = 10.8 Hz), 5.16 (dd, 2H, *J* = 18.9 Hz, *J* = 10.5 Hz), 2.91-2.88 (m, 4H), 2.45 (s, 3H), 1.70 (s, 6H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 144.4, 134.5, 131.2, 124.5, 123.7, 123.4, 120.9, 119.1, 113.9, 112.4, 110.9, 59.2, 52.0, 36.3, 28.0, 25.5; HRMS (ESI) calcd for $C_{16}H_{22}N_2Br$ $(M^{+}H)+$ 321.0961, found 321.0967; IR ν_{max}/cm^{-1} 2937, 1512; λ_{max}: 242 nm, 308 nm.



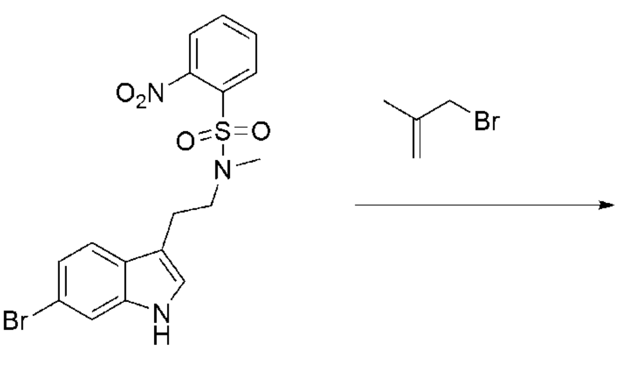
SI-29: was prepared by similar synthetic methods developed by Glennon.[6] The crude residue was purified by flash chromatography (10-30% Acetone:Petroleum Ether) to afford the product **SI-29** (234 mg, 86% yield) as a yellow oil. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.95-7.92 (m, 1H), 7.67-7.55 (m, 4H), 7.27-7.18 (m, 2H), 7.17-7.11 (m, 1H), 5.95-5.86 (m, 1H), 5.31 (s, 1H), 5.16 (d, 1H, *J* = 5.7 Hz), 4.92 (d, 1H, *J* = 19.8 Hz), 4.79 (m, 1H), 3.49 (t, 2H, *J* = 8.7 Hz), 3.12-3.07 (m, 2H), 3.05 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 148.1, 136.4, 133.5, 132.8, 131.7, 130.7, 127.2, 124.2, 122.3, 120.2, 118.2, 117.1, 113.3, 111.1, 109.9, 49.9, 47.2, 35.2, 24.8; HRMS (ESI) calcd for $C_{20}H_{21}N_3O_4SBr$ $(M^{+}H)+$ 478.0431, found 478.0426; IR ν_{max}/cm^{-1} 3426, 1644, 1350, 749; λ_{max}: 280 nm, 291 nm, 306 nm.



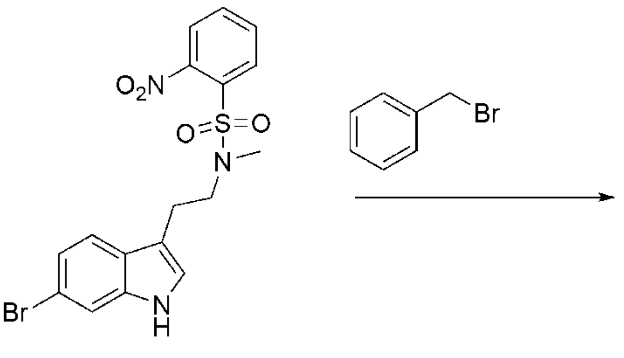
SI-30: was prepared and purified in a similar manner to **SI-29** to provide **SI-30.** The crude residue was purified by flash chromatography (10-30% EtOAc:Hexanes) to afford the product SI-10 (178 mg, 85% yield) as a green oil. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.91-7.88 (m, 1H), 7.61-7.52 (m, 4H), 7.37 (d, 1H, *J* = 8.1 Hz), 7.27-7.21 (m, 1H), 7.17-7.12 (m, 1H), 4.90 (s, 2H), 3.46 (t, 2H, *J* = 8.1 Hz), 3.07 (t, 2H, *J* = 8.1 Hz), 3.03 (s, 3H), 2.29 (t, 1H, *J* = 2.4 Hz); ^{13}C NMR (100 MHz, $CDCl_3$) δ 148.1, 136.3, 133.4, 132.9, 131.7, 130.9, 127.5, 124.2, 122.8, 120.8, 118.5, 112.8, 112.2, 109.9, 72.9, 49.9, 35.2, 34.6, 24.8; HRMS (ESI) calcd for $C_{20}H_{19}N_3O_4SBr$ (M^+H)+ 475.0274, found 476.0264; IR ν_{max}/cm^{-1} 2916, 1707, 1344; λ_{max}: 277 nm, 306 nm.



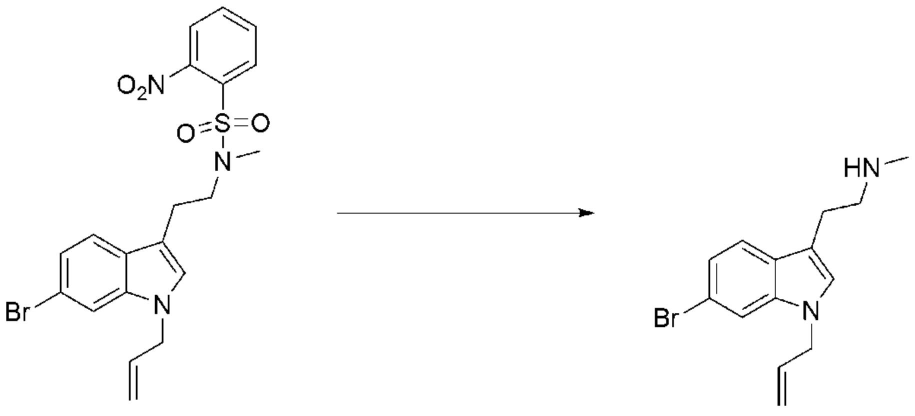
SI-31: was prepared and purified in a similar manner to **SI-29** to provide **SI-31**. The crude residue was purified by flash chromatography (10-25% Acetone:Petroleum Ether) to afford the product SI-11 (221 mg, 78% yield) as a yellow oil. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.91-7.89 (m, 1H), 7.62-7.54 (m, 4H), 7.29 (d, 1H, *J* = 6.0 Hz), 7.21 (t, 1H, *J* = 5.7 Hz), 7.13 (t, 1H, *J* = 5.7 Hz), 4.14 (t, 2H, *J* = 5.4 Hz), 3.49 (t, 2H, *J* = 5.4 Hz), 3.09 (t, 2H, *J* = 5.4 Hz), 3.05 (s, 3H), 1.73 (quintet, 2H, *J* = 5.7 Hz), 1.38 (sextet, 2H, *J* = 5.4 Hz), 0.96 (t, 3H, *J* = 5.7 Hz) ; ^{13}C NMR (100 MHz, $CDCl_3$) δ 148.0, 136.2, 133.4, 132.7, 131.7, 130.6, 127.1, 124.1, 122.0, 119.9, 118.1, 113.3, 110.5, 109.7, 49.9, 44.9, 35.1, 32.1, 24.7, 20.2, 14.0; HRMS (ESI) calcd for $C_{21}H_{25}N_3O_4SBr$ (M^+H)+ 494.0744, found 494.0729; IR ν_{max}/cm^{-1} 2930, 1546, 1350, 728; λ_{max}: 229 nm, 284 nm.



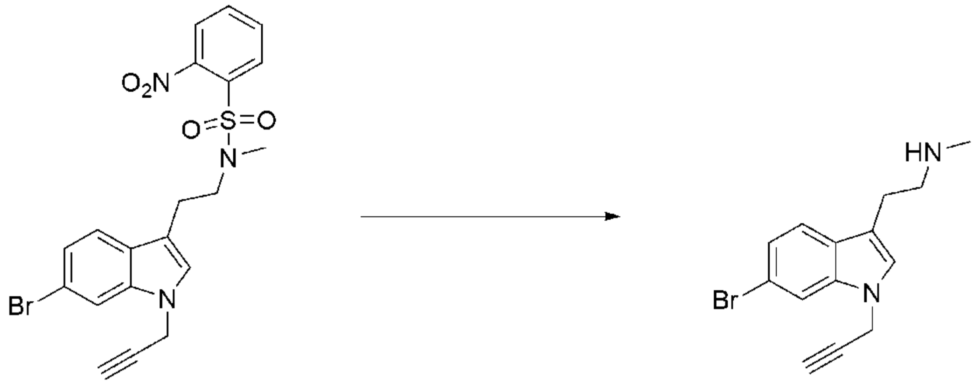
SI-32: was prepared and purified in a similar manner to **SI-29** to provide **SI-32**. The crude residue was purified by flash chromatography (10-25% EtOAc:Hexanes) to afford the product SI-12 (92 mg, 33% yield) as a yellow oil.^{1}H NMR (400 MHz, $CDCl_3$) δ 7.93-7.91 (m, 1H), 7.64-7.55 (m, 4H), 7.26-4.12 (m, 3H), 4.85 (s, 1H), 4.67 (s, 2H), 4.45 (s, 1H), 3.48 (t, 2H, J = 7.6 Hz), 3.07 (t, 2H, J = 7.6 Hz), 3.03 (s, 3H), 1.70 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 140.7, 136.9, 133.8, 133.2, 132.0, 131.1, 127.5, 124.5, 122.6, 120.6, 118.5, 114.1, 112.4, 111.3, 110.4, 50.9, 50.3, 35.5, 25.2, 20.4; HRMS (ESI) calcd for $C_{21}H_{22}N_3O_4SBr$ (M^+H)+ 492.0587, found 492.0577; IR ν_{max}/cm^{-1}3454, 2923, 1539, 1455, 1344, 1155; λ_{max}: 231 nm, 283 nm.



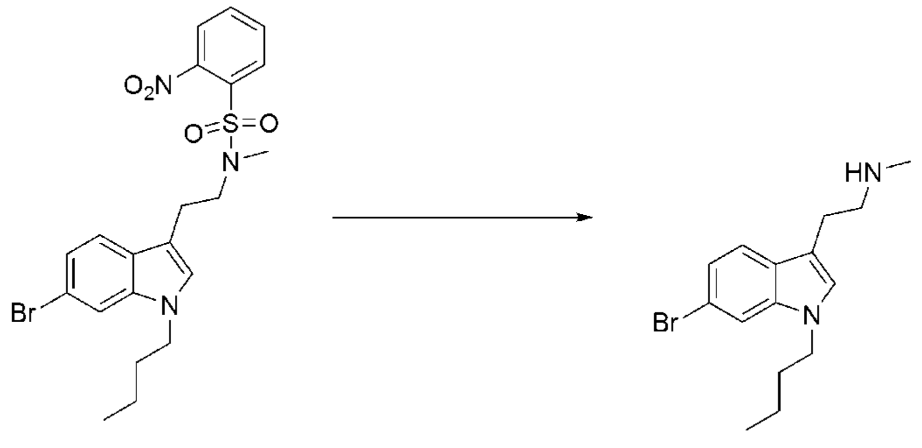
SI-33: was prepared and purified in a similar manner to **SI-29** to provide **SI-33**. The crude residue was purified by flash chromatography (10-30% Acetone:Petroleum Ether) to afford the product SI-13 (245 mg, 81% yield) as a yellow crystals. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.93-7.91 (m, 1H), 7.62-7.54 (m, 4H), 7.28-7.21 (m, 4H), 7.16-7.13 (m, 2H), 7.07 (d, 2H, J = 6.4 Hz), 5.38 (s, 2H), 3.51 (t, 2H, J = 9.6 Hz), 3.13 (t, 2H, J = 9.6 Hz), 3.05 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 148.1, 137.1, 136.7, 133.4, 132.8, 131.7, 130.8, 129.9, 128.9, 127.7, 127.3, 126.6, 124.2, 122.5, 120.4, 118.3, 113.7, 111.4, 110.1, 49.9, 48.4, 35.2, 24.9; HRMS (ESI) calcd for $C_{24}H_{22}N_3O_4SBr$ (M^+H)+ 528.0587, found 528.0581; IR ν_{max}/cm^{-1} 3448, 1630, 1539, 1336; λ_{max}: 208 nm, 282 nm.



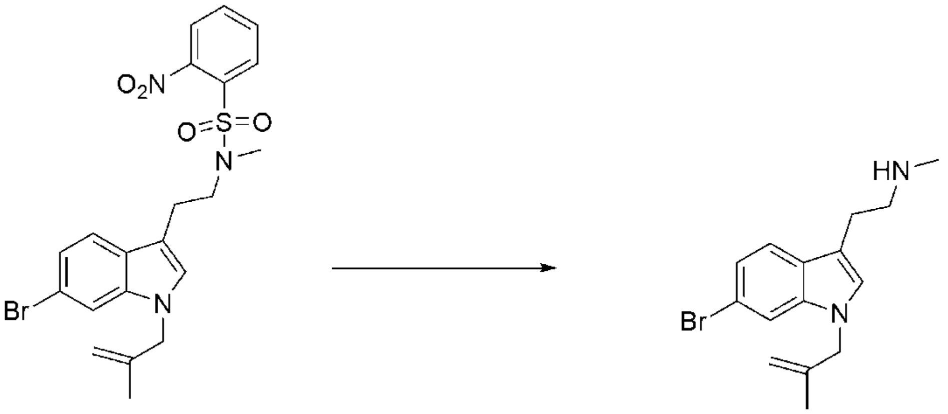
SI-34 (110 mg, 0.230 mmol) was placed in a vial and dissolved in 4 mL of CH_3CN. Then K_2CO_3 (127 mg, 0.920 mmol) was added to the vial, then thiophenol (110.2 mg, 0.920 mmol) was added dropwise and the reaction was allowed to stir for 16 h at room temperature.[7,8] The reaction was then concentrated under reduced pressure to afford crude product. Purification by flash chromatography (1-10% sat. NH_3-MeOH/CH_2Cl_2) afforded **SI-34** as yellow oil (45 mg, 67% yield). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.61 (d, 1H, J = 7.8 Hz), 7.28-7.26 (m, 1H), 7.20 (t, 1H, J = 7.2 Hz), 7.12 (t, 1H, J = 6.9 Hz), 5.99-5.86 (m, 1H), 5.16 (d, 1H, J = 10.5 Hz), 4.94 (d, 1H, J = 17.1 Hz), 4.8 (d, 2H, J = 5.1 Hz), 3.04 (t, 2H, J = 6.9 Hz), 2.93 (t, 2H, J = 6.9 Hz), 2.70 (br s, 1H), 2.50 (s, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 136.6, 132.9, 127.6, 122.2, 120.1, 118.6, 117.2, 113.3, 112.7, 109.9, 51.7, 47.3, 36.3, 25.7; HRMS (ESI) calcd for $C_{14}H_{18}N_2Br$ (M^+H)+ 293.0648, found 293.0646; IR ν_{max}/cm^{-1} 3412, 2923, 1651, 1462, 728; λ_{max}: 208 nm, 286 nm.



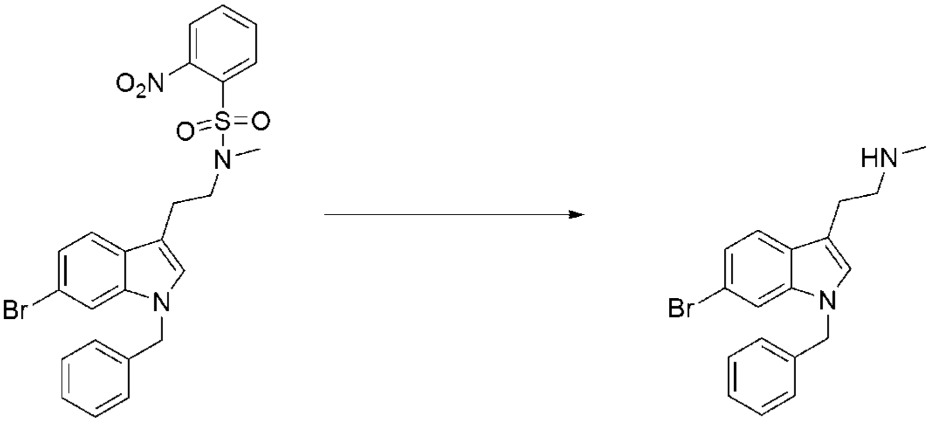
SI-35 was prepared and purified in a similar manner to **SI-34** to provide **SI-35** (8.9 mg, 65% yield), as a yellow oil. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.58 (d, 1H, J = 5.7 Hz), 7.41-7.39 (m, 1H), 7.27-7.23 (m, 1H), 7.15 (t, 1H, J = 6.0 Hz), 4.95 (d, 2H, J = 1.8 Hz), 2.99 (t, 2H, J = 5.4 Hz), 2.90 (t, 2H, J = 4.8 Hz), 2.47 (s, 3H), 2.29 (t, 1H, J = 1.5 Hz), 2.02 (br s, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 129.2, 122.3, 120.6, 115.7, 115.4, 111.5, 105.9, 105.6, 102.7, 65.7, 43.8, 28.4, 27.5, 17.8; HRMS (ESI) calcd for $C_{14}H_{16}N_2Br$ (M^+H)+ 291.0491, found 291.0484; IR ν_{max}/cm^{-1} 3420, 1644, 1015; λ_{max}: 208 nm, 282 nm.



SI-36 was prepared and purified in a similar manner to **SI-34** to provide **SI-36** (47 mg, 53% yield), as a yellow oil. ^{1}H NMR (300 MHz, CDCl$_3$) δ 7.61-7.58 (m, 1H), 7.30-7.26 (m, 1H), 7.21-7.16 (m, 1H), 7.12-7.07 (m, 1H), 4.16 (t, 2H, *J* = 7.5 Hz), 3.35 (br s, 1H), 3.07-3.02 (m, 2H), 2.94-2.89 (m, 2H), 2.50 (s, 3H), 1.74 (quintet, 2H, *J* = 7.8 Hz), 1.38 (sextet, 2H, *J* = 7.8 Hz), 0.95 (t, 3H, *J* = 7.2 Hz); ^{13}C NMR (100 MHz, CDCl$_3$) δ 136.4, 127.5, 121.9, 119.8, 118.5, 113.4, 112.1, 109.7, 51.6, 45.0, 36.2, 32.3, 25.6, 20.4, 14.1; HRMS (ESI) calcd for $C_{15}H_{22}N_2Br$ (M$^+$H)+ 309.0961, found 309.0961; IR ν_{max}/cm^{-1} 2937, 1350, 742; λ_{max}: 208 nm, 284 nm.



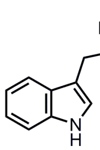
SI-37 was prepared and purified in a similar manner to **SI-34** to provide **SI-37** (33 mg, 61% yield), as a yellow oil. ^{1}H NMR (300 MHz, CDCl$_3$) δ 7.62-7.59 (m, 1H), 7.20-7.08 (m, 3H), 5.3 (s, 1H), 4.85 (s, 1H), 4.70 (s, 2H), 4.47 (s, 1H), 3.04-2.92 (br s, 3H), 2.92-2.50 (m, 2H), 2.49-2.41 (m, 2H), 1.71 (s, 3H); ^{13}C NMR (75 MHz, CDCl$_3$) δ 140.6, 136.7, 127.4, 122.1, 120.0, 118.4, 113.1, 112.3, 112.1, 110.1, 50.6, 25.6, 20.2; HRMS (ESI) calcd for $C_{15}H_{19}N_2Br$ (M$^+$H)+ 307.0804, found 307.0798; IR ν_{max}/cm^{-1} 3412, 2930, 1455, 1322, 1127, 735; λ_{max}: 233 nm, 276 nm.



SI-38 was prepared and purified in a similar manner to **SI-34** to provide **SI-38** (11 mg, 34% yield), as a yellow oil. ^{1}H NMR (300 MHz, $CDOD_3$) δ 7.63-7.60 (m, 1H), 7.35-7.32 (m, 1H), 7.28-7.21 (m, 3H), 7.16-7.11 (m, 2H), 7.09-7.04 (m, 2H), 5.48 (s, 2H), 3.11-3.07 (m, 4H), 2.59 (s, 3H); ^{13}C NMR (100 MHz, $CDOD_3$) δ 138.9, 138.4, 129.8, 128.6, 128.5, 127.7, 123.7, 121.4, 119.1, 114.7, 112.1, 111.5, 51.2, 34.9, 24.9; HRMS (ESI) calcd for $C_{18}H_{20}N_2Br$ $(M^+H)+$ 343.0804, found 343.0801; IR ν_{max}/cm^{-1} 2930, 1714, 1511; λ_{max}: 221 nm, 281 nm.

Inhibition of *E. coli and S. aureus* Biofilms with dfBr Analogues

Biofilm Activity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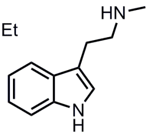
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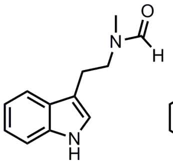
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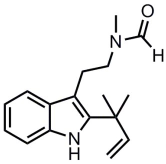
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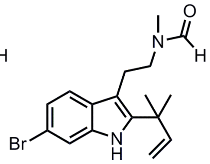
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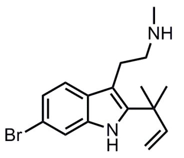
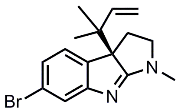
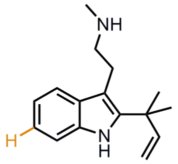

dfBr (44)


Flustramine C (45)

Compound	*E. coli* K 12 ER 2738	*S. aureus* ATCC 29213
39	< 5%	NA
40	< 5%	NA
41	Inducer	NA
42	< 5%	NA
43	Inducer	NA
44	IC $_{50}$ = 174.0 μM*	IC$_{50}$ = 70.0μM*
45	20% @ 100 μM	15% @ 150 μM

Table 1. Biological evaluation of flustramine C (**45**) intermediates. Inducer indicates increase of biofilm formation in comparison to controls. <5% inhibition was measured at 150 μM. * = Indicates inhibition of biofilm formation through a toxic mechanism as determined by growth curve analysis. Compounds 39 -45 were synthesized through the Lindel procedure.[4]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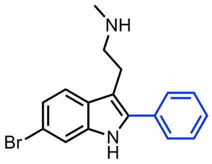
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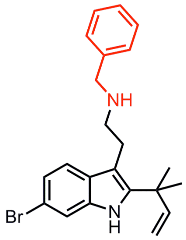
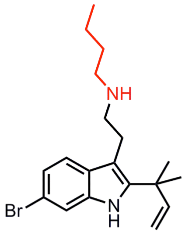

22 (3)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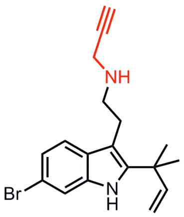
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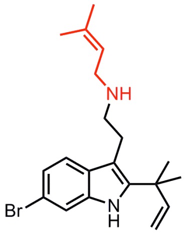
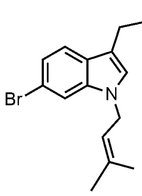
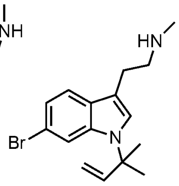

20

Compound	*E. coli*	*S. aureus*
1	< 5%	35% @ 150 μM
6	20% @ 200 μM	15% @ 200 μM
22 (3)	IC$_{50}$ = 53.0 μM	IC$_{50}$ = 5.9 μM
19	IC$_{50}$ = 101.5 μM	IC$_{50}$ = 12.7 μM
21	IC$_{50}$ = 85.8 μM	IC$_{50}$ = 37.4 μM**
20	IC$_{50}$ = 87.5 μM	IC$_{50}$ = 48.8 μM**

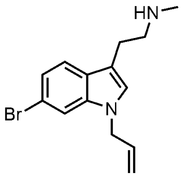
Table 2. Biological activity of region A ,B,C. <5% inhibition was measured at 150 μM. * = Indicates inhibition of biofilm formation through a toxic mechanism as determined by growth curve analysis. ** = Indicates not complete toxicity, bacterial growth delay is noted in the first 4 hours, bacterial density is identical at 6, 8, and 24 hours.



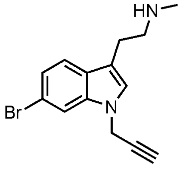
25



28 (4)



34



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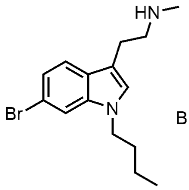

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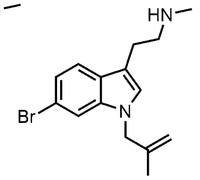
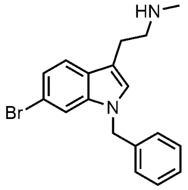


37



38

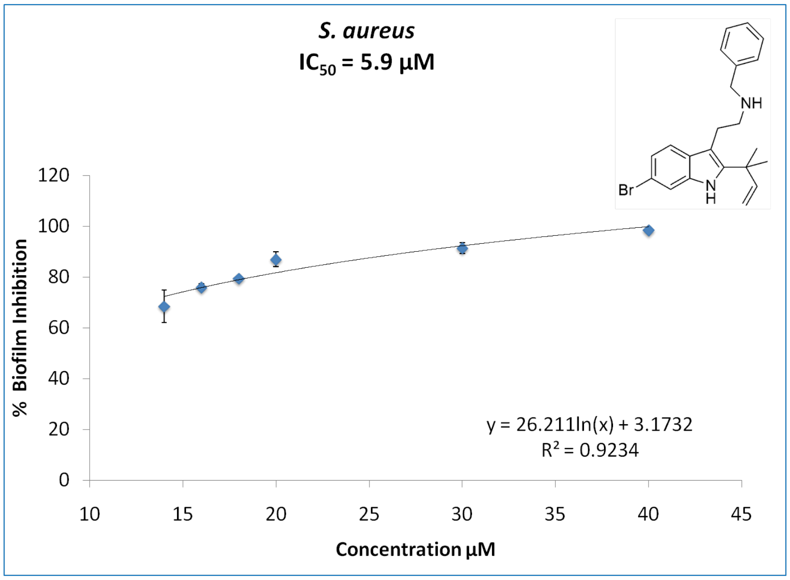
Compound	*E. coli*	*S. auerus*
25	IC_{50} = 121.5 μM*	IC_{50} = 85.6 μM**
28 (4)	IC_{50} = 79.8 μM	IC_{50} = 64.7 μM
37	< 5%	< 5%
36	IC_{50} = 97.7 μM	IC_{50} = 74.9 μM
34	30% @ 100 μM	< 5%
35	< 5%	IC_{50} = 94.3 μM*
38	IC_{50} = 102.9 μM	IC_{50} = 85.8 μM

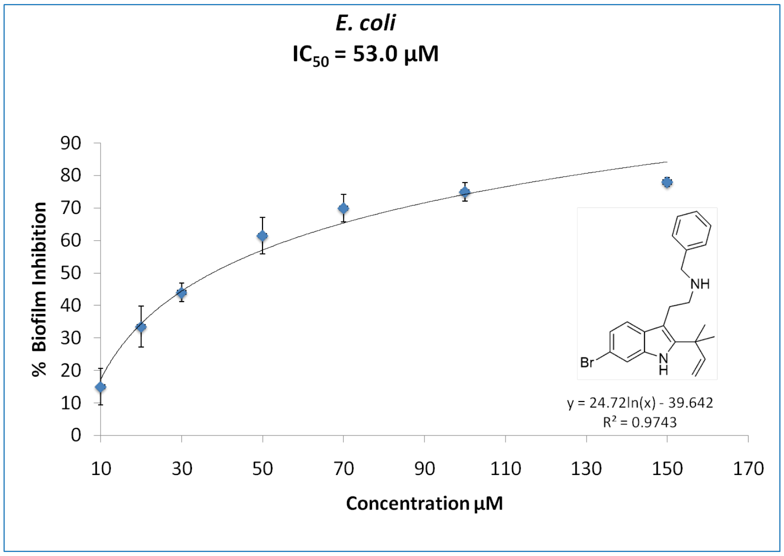
Table 3. Biological activity of region D. <5% inhibition was measured at 150 μM. * = Indicates inhibition of biofilm formation through a toxic mechanism as determined by growth curve analysis. ** = Indicates not complete toxicity, bacterial growth delay is noted in the first 4 hours, bacterial density is identical at 6, 8, and 24 hours.

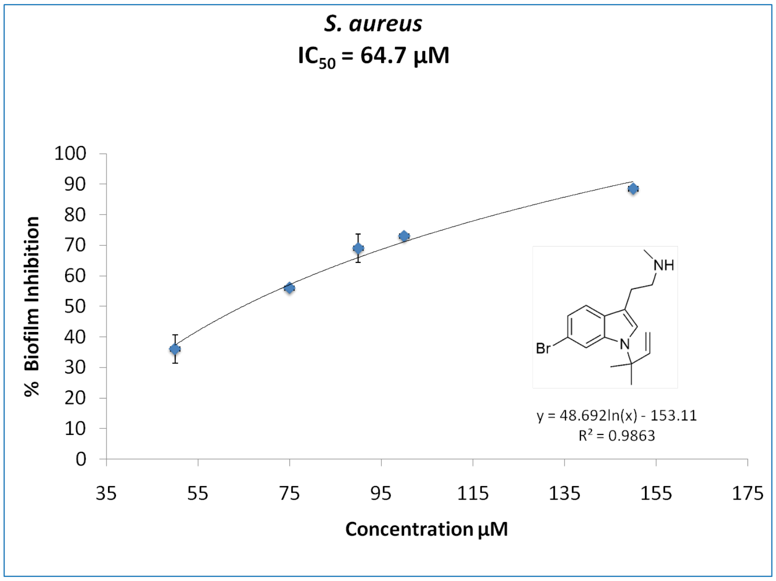
Biofilm Inhibition Procedure (Dose Reponse Curve): to determine the inhibitory effects of test compounds against *E. coli* (K12 ER2718) and *S. aureus* (ATCC 29213) biofilm formation. Inhibition assays were performed by taking an overnight culture and sub-culturing it to an OD_{600} of 0.01 into the necessary media; Luria-Bertani (LB) for *E coli*, and Tryptic Soy Broth with 5% Glucose (TSBG) for *S. aureus*. Solutions of predetermined concentrations of the test compounds were made from DMSO (biology grade) stock solutions. These solutions were aliquoted (100 μL) into the wells of a 96-well PVC microtiter plate. Sample plates were wrapped in GLAD Press n' Seal® followed by incubation under stationary conditions for 24 h at 37 °C. After incubation the media was discarded and the plates were washed with water. The sample plates were then stained with 110 μL of 0.1% aqueous solution of crystal violet (CV) and then incubated at ambient temperature for 30 min. The CV stain was then discarded and the plates were washed with water. The remaining stain was solubilized with 200 μL of 95% ethanol. After the stain was dissolved (10 minutes) 125 μL of the solubilized CV was transferred from each well into the corresponding wells of a polystyrene microtiter dish. Biofilm inhibition was quantified by measuring the OD_{540} of each well in which a negative control lane wherein no biofilm was formed served as a background and was subtracted out. The percent inhibition was calculated by the comparison of the OD_{540} for biofilm grown in the absence of compound (control) versus biofilm grown in the presence of compound under identical conditions.

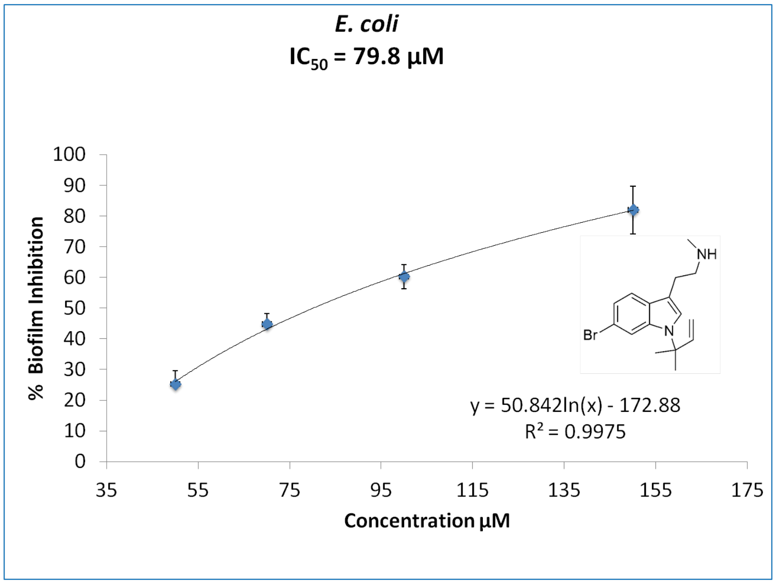
Representative Biofilm Inhibition Data (Dose Response Curve)

All dose-response curves for IC_{50} value determinations were run in triplicate or more







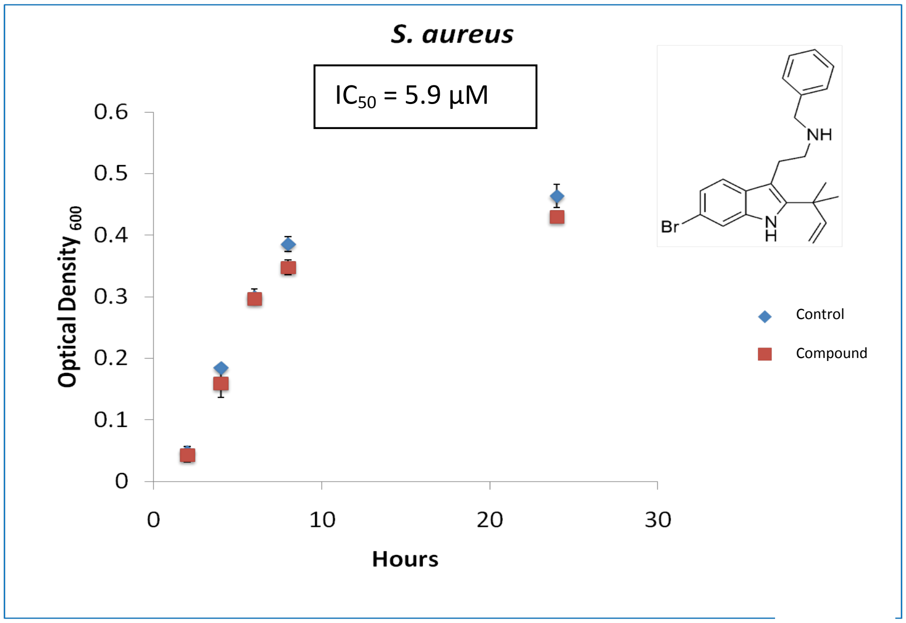


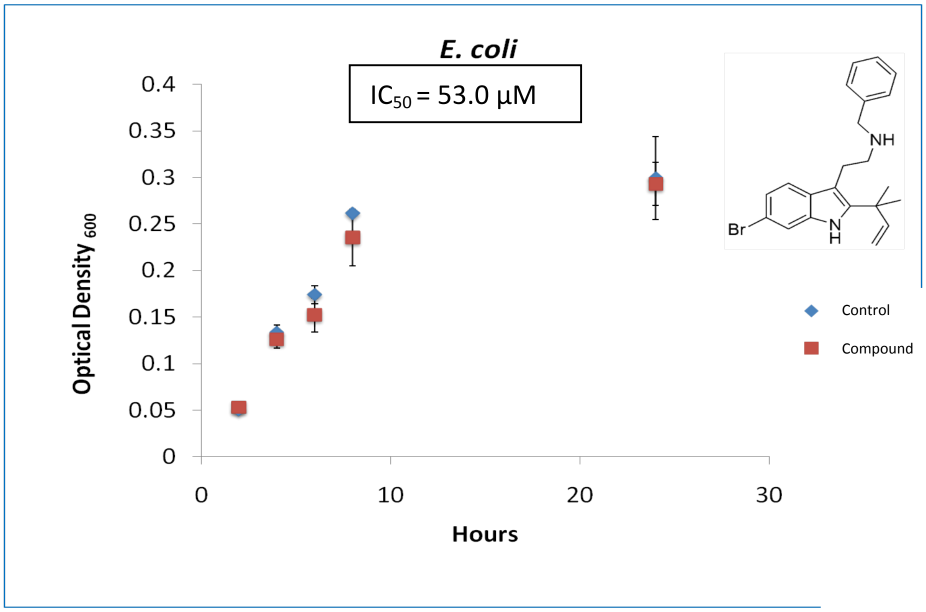
Growth Curve Analysis: to determine the toxicity of the test compound at its IC_{50} value. In the growth curve assay, bacterial cell density was evaluated in the appropriate media by taking its optical density (OD_{600}). The bacteria were cultured overnight and the OD_{600} adjusted to 0.01. A control tube containing only media and bacteria, and a tube containing media, bacteria and the IC_{50} concentration of compound were prepared in duplicate. Cultures were incubated at 37 ˚C and the OD_{600} was determined at time (t) = 2, 4, 6, 8, 24 hours. A compound was determined to be non-toxic if comparable OD_{600} readings to the control were observed at the same time points.

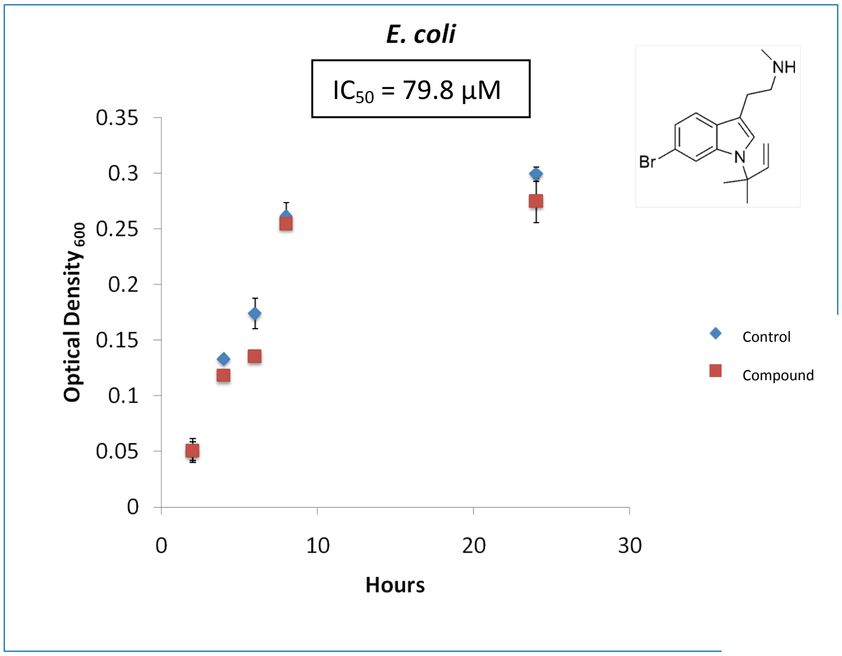
Representative Growth Curves

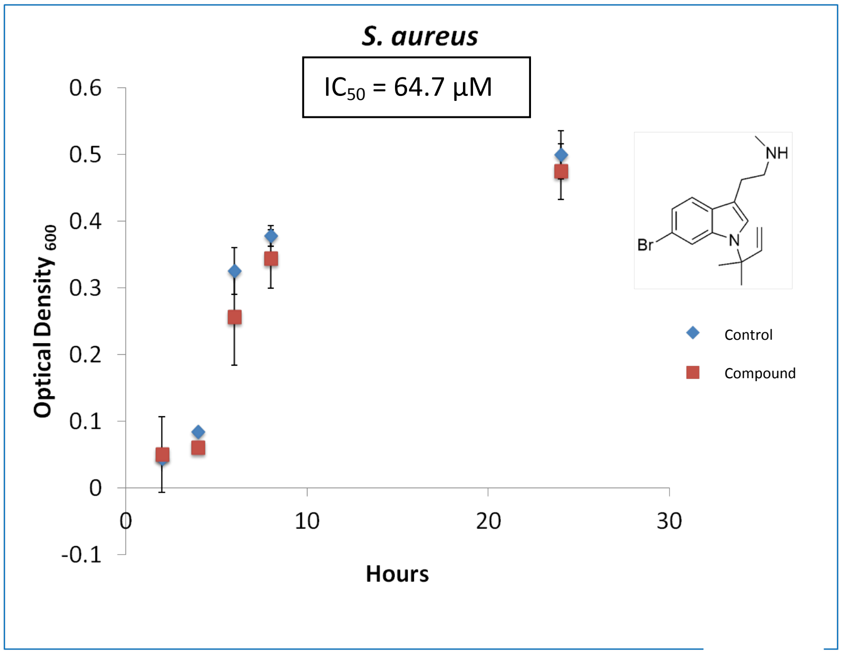
All growth curves performed in duplicate.

Control is in blue and compound is in red.






E. coli
IC_{50} = 79.8 µM
NH
Br
N
Optical Density $_{600}$
0.35
0.3
0.25
0.2
0.15
0.1
0.05
0
0
10
20
30
Hours
Control
Compound


S. aureus
IC_{50} = 64.7 µM
NH
Br
N
Optical Density $_{600}$
0.6
0.5
0.4
0.3
0.2
0.1
0
-0.1
0
10
20
30
Hours
Control
Compound

Temperature Dependent Biofilm Assays with *E. coli* Strains

E. coli strains BW25113 (parent strain), JW1901-5 (*sdiA* knockout) and JW3686-7 (*tnaA* knockout), part of the Keio collection,[10] were obtained from the Coli Genetic Stock Center (CGSC) at Yale University. Strains were cultured overnight in LB containing 25 µg/mL kanamycin, bacteria were sub-cultured to an OD_{600} of 0.05 in Luria-Bertani (LB) . Bacteria were incubated in polystyrene 96-well plates at 25 and 37 ˚C for 16 h in the presence and absence of indole and compound **22**.[11] The same protocol as described above was used to measure the amount of biofilm formed. Each data point was averaged from at least 24 replicate wells (twelve wells from two independent cultures). The control (biofilm formed in the absence of indole and compound **22**) was normalized to an OD_{540} of 1.

References

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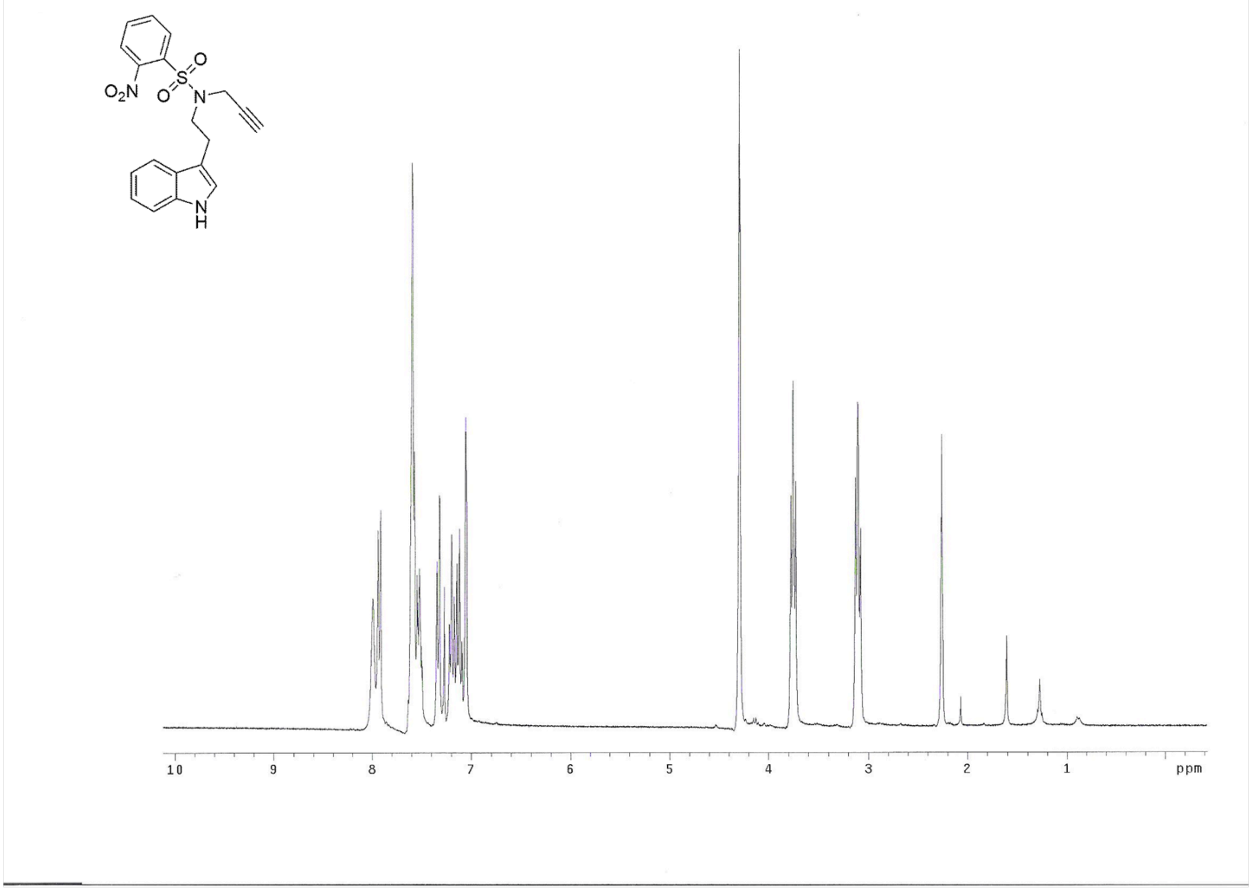
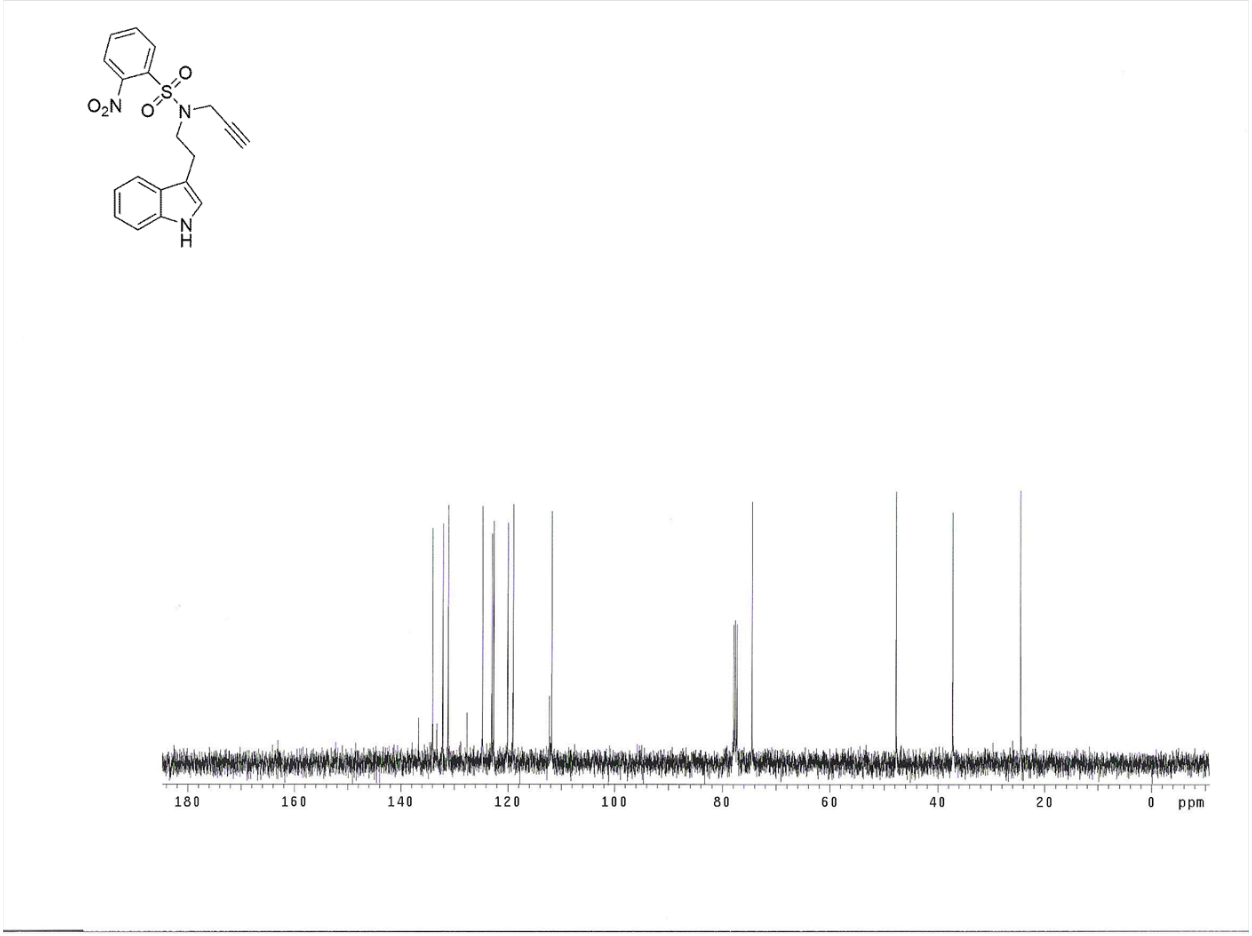
NMR Spectra: ^{1}H & ^{13}C

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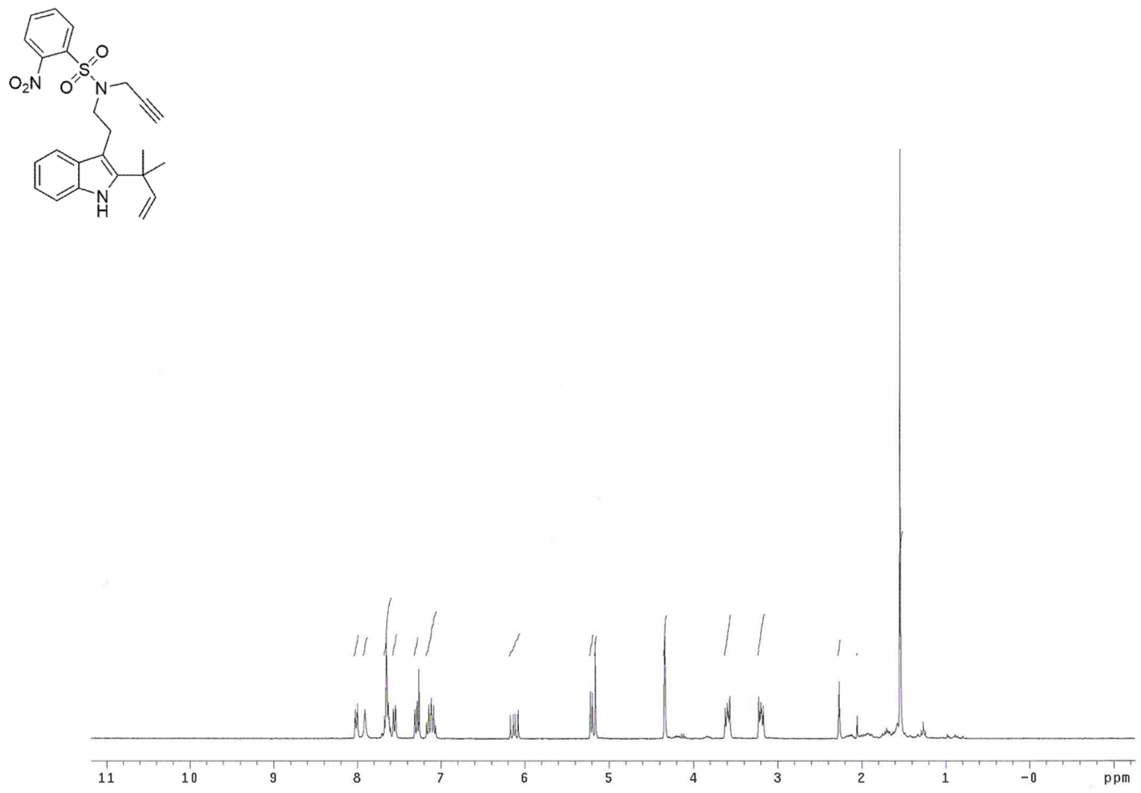
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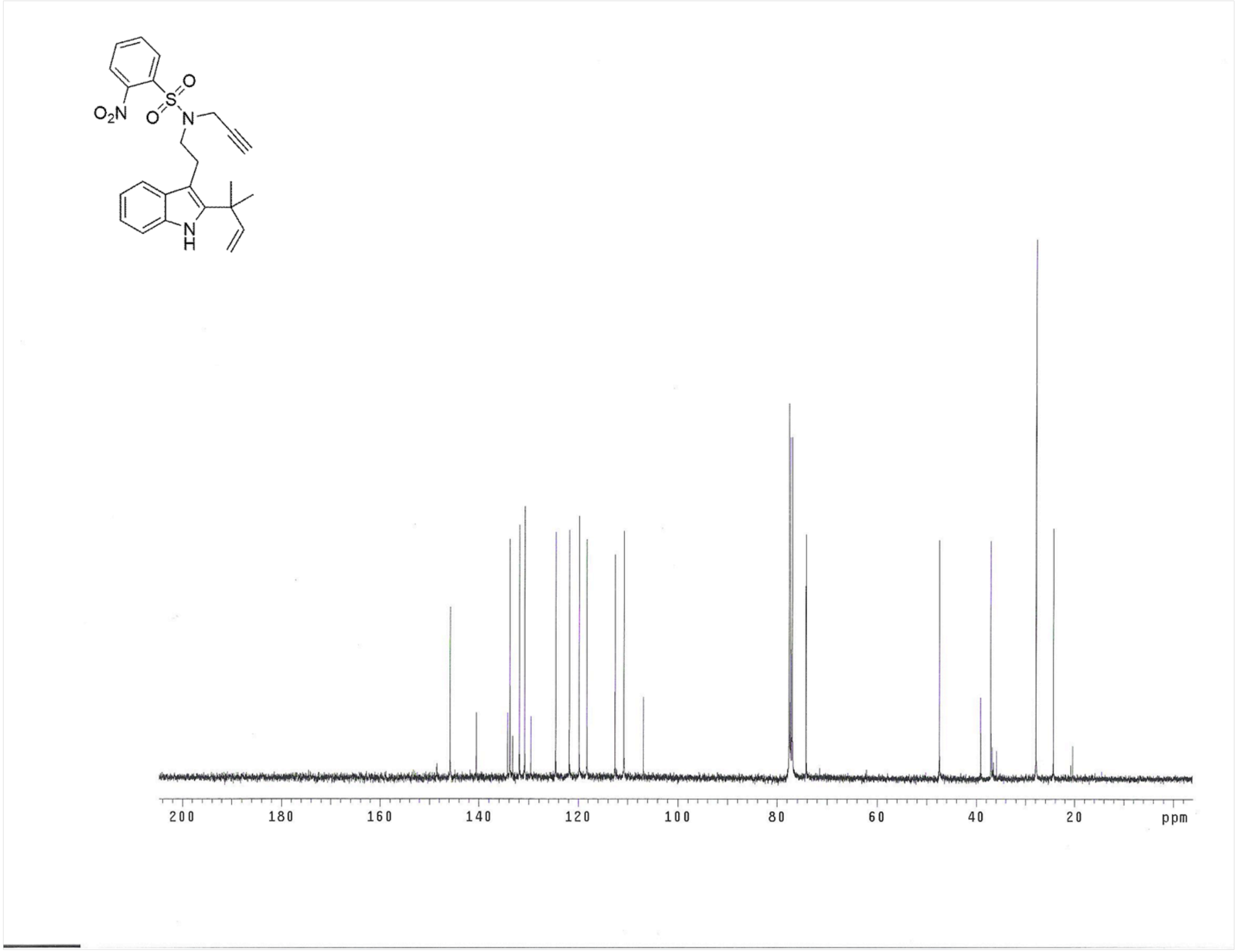
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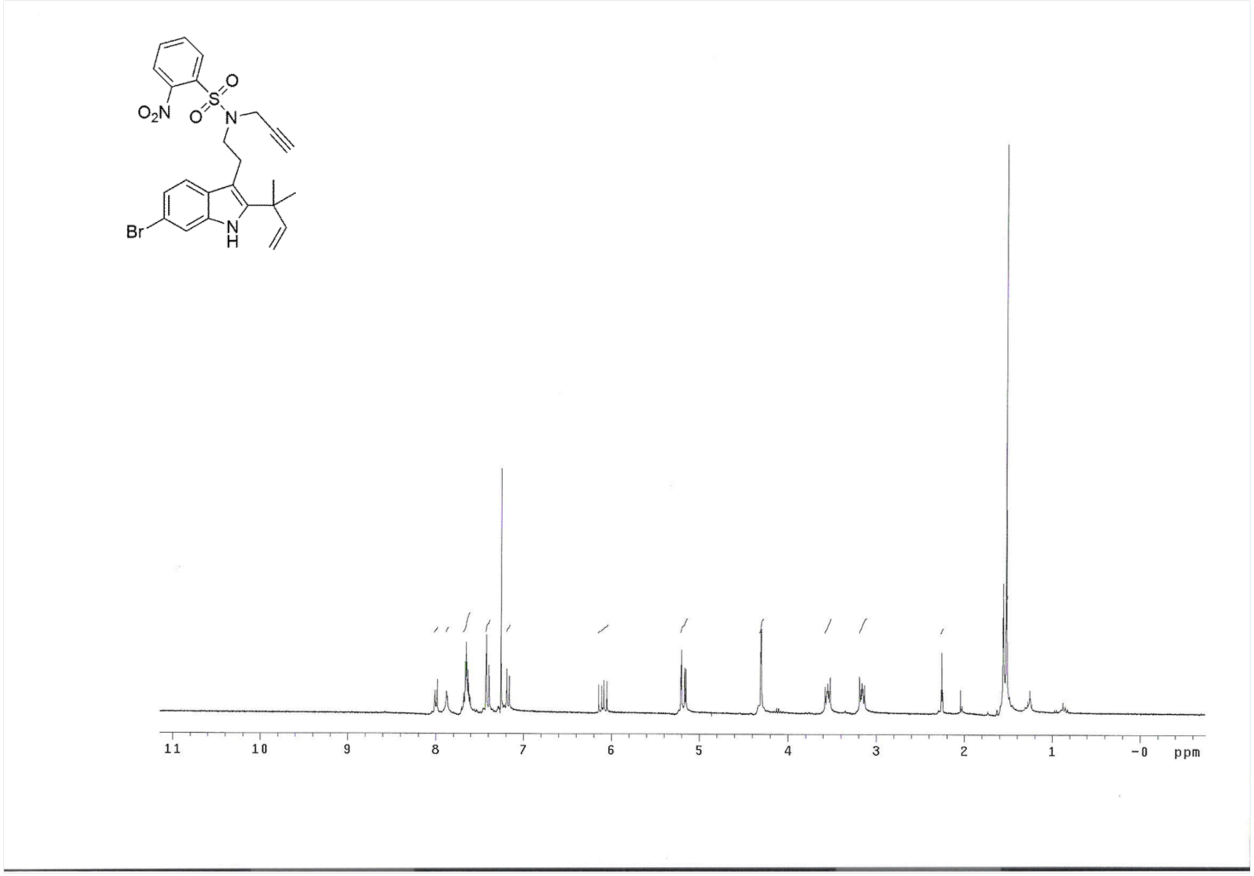
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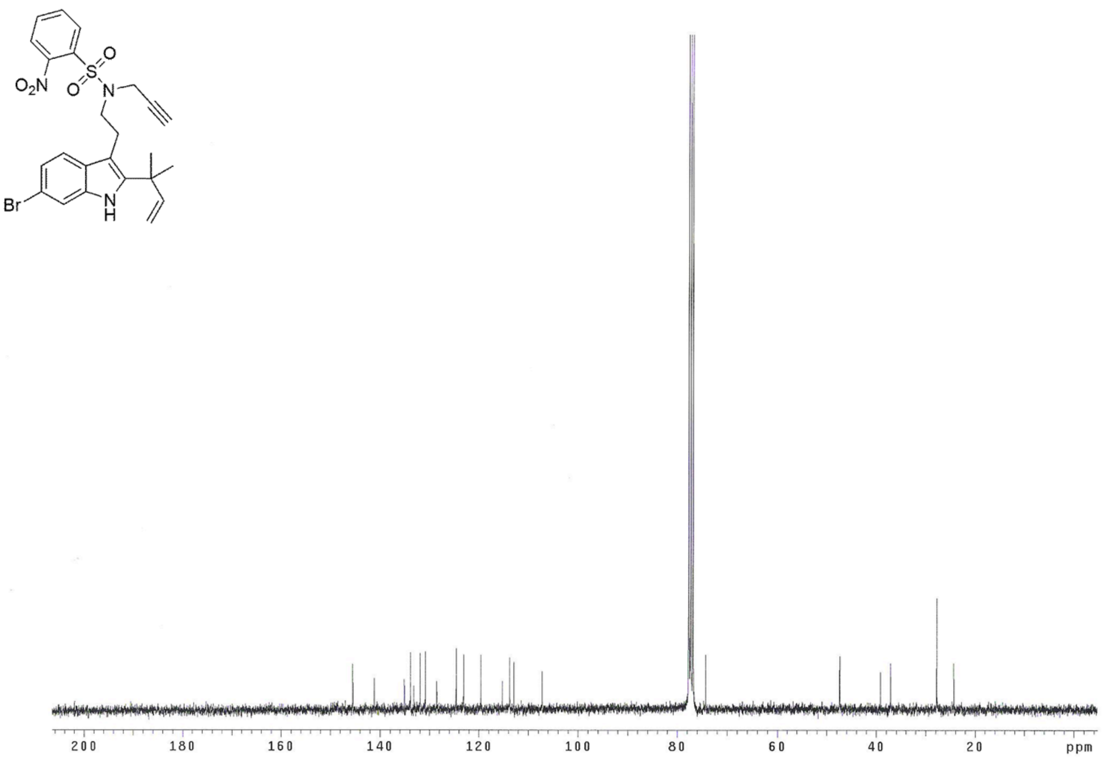

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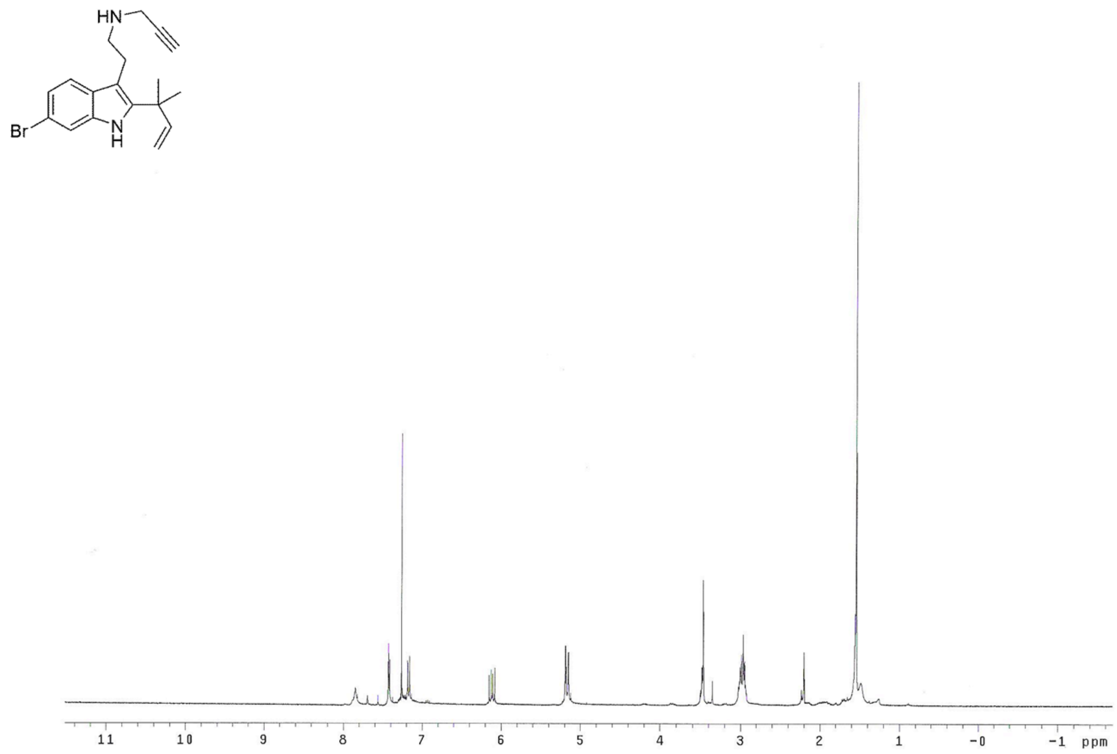
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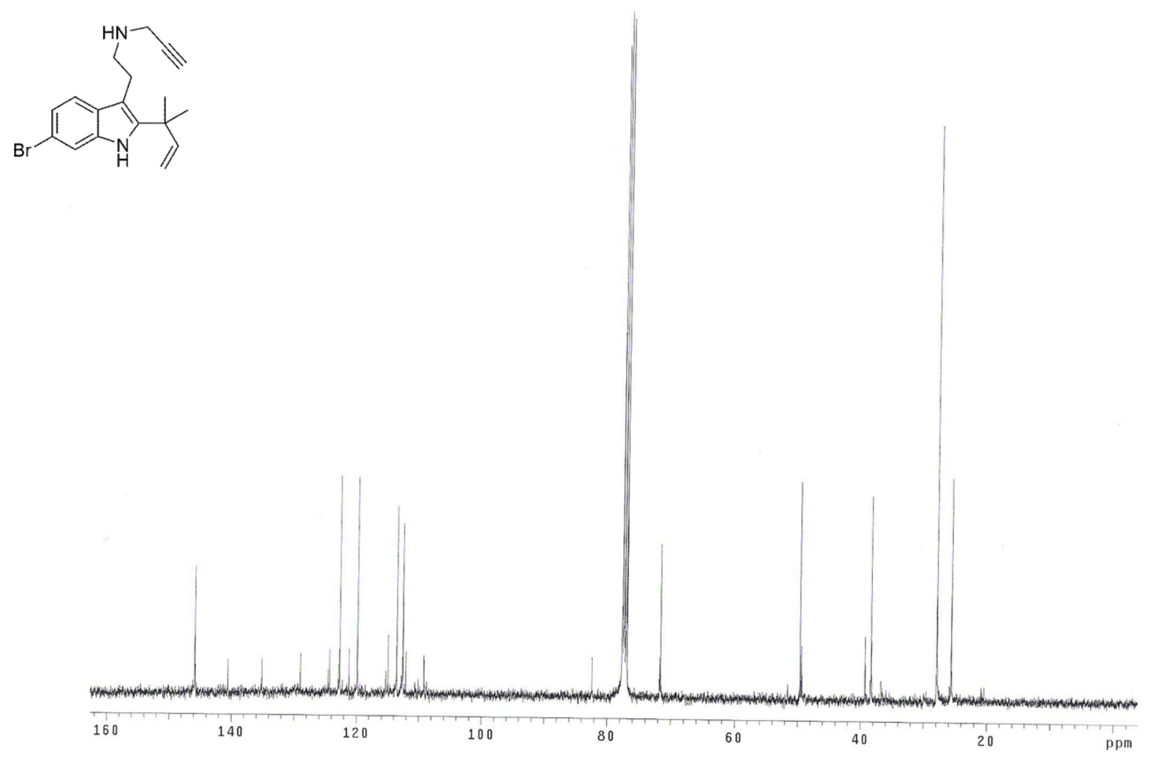
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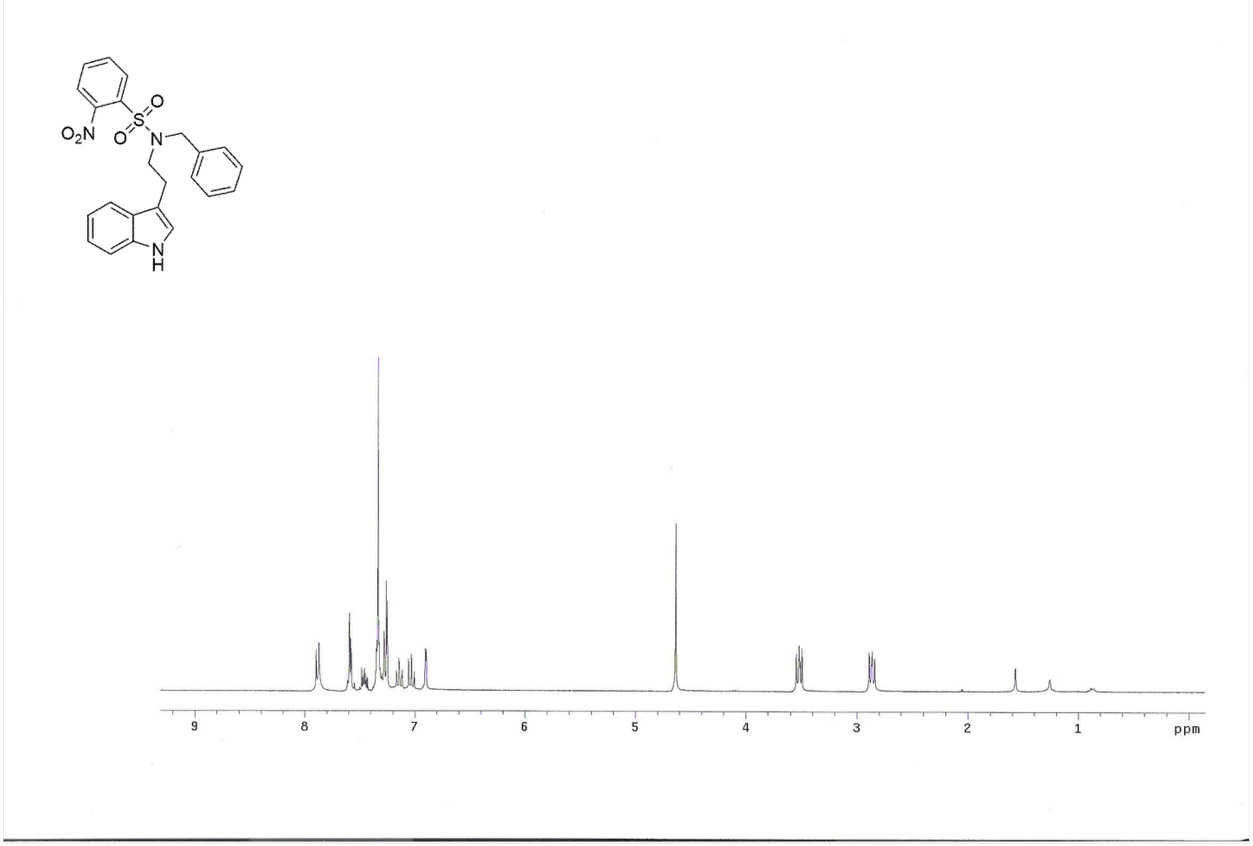
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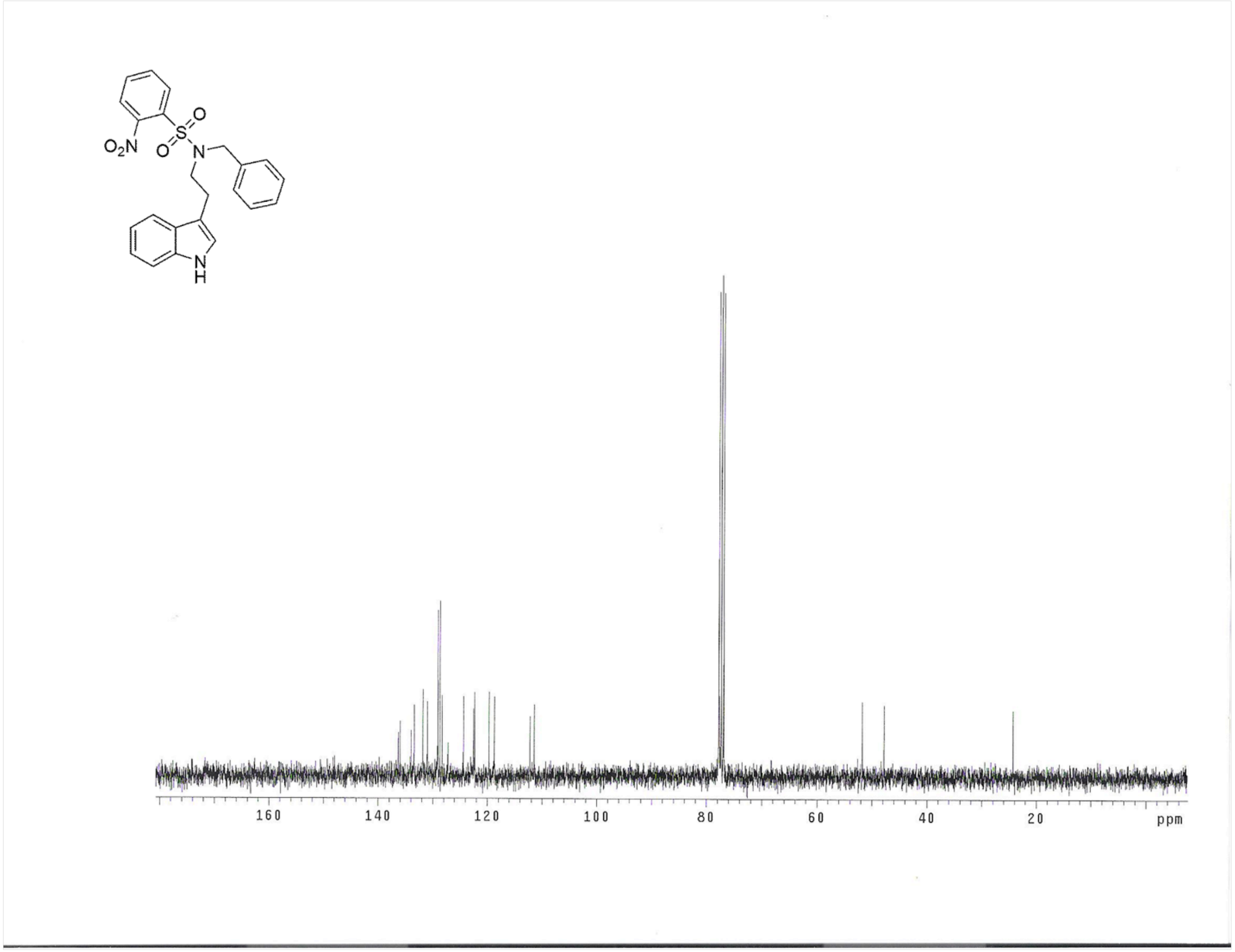
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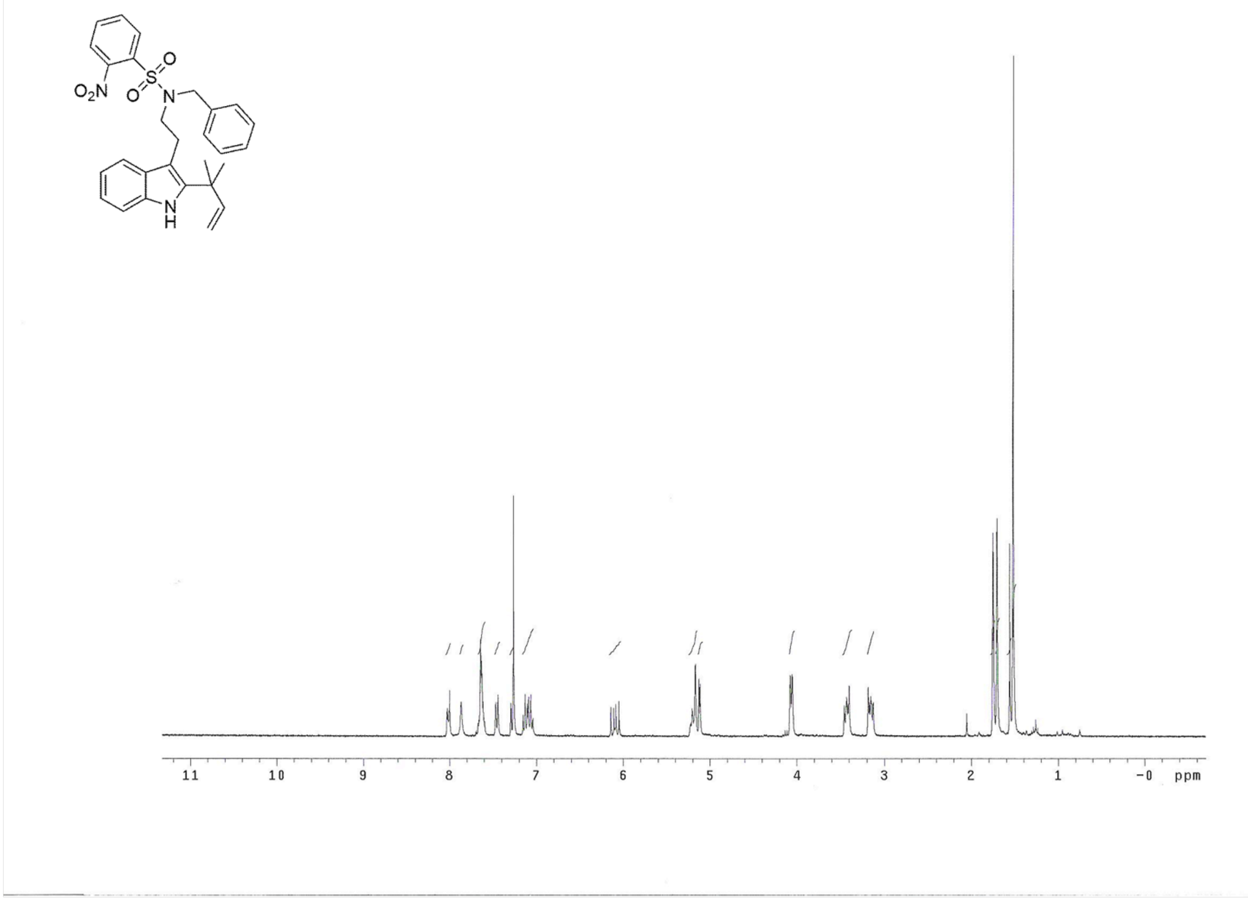
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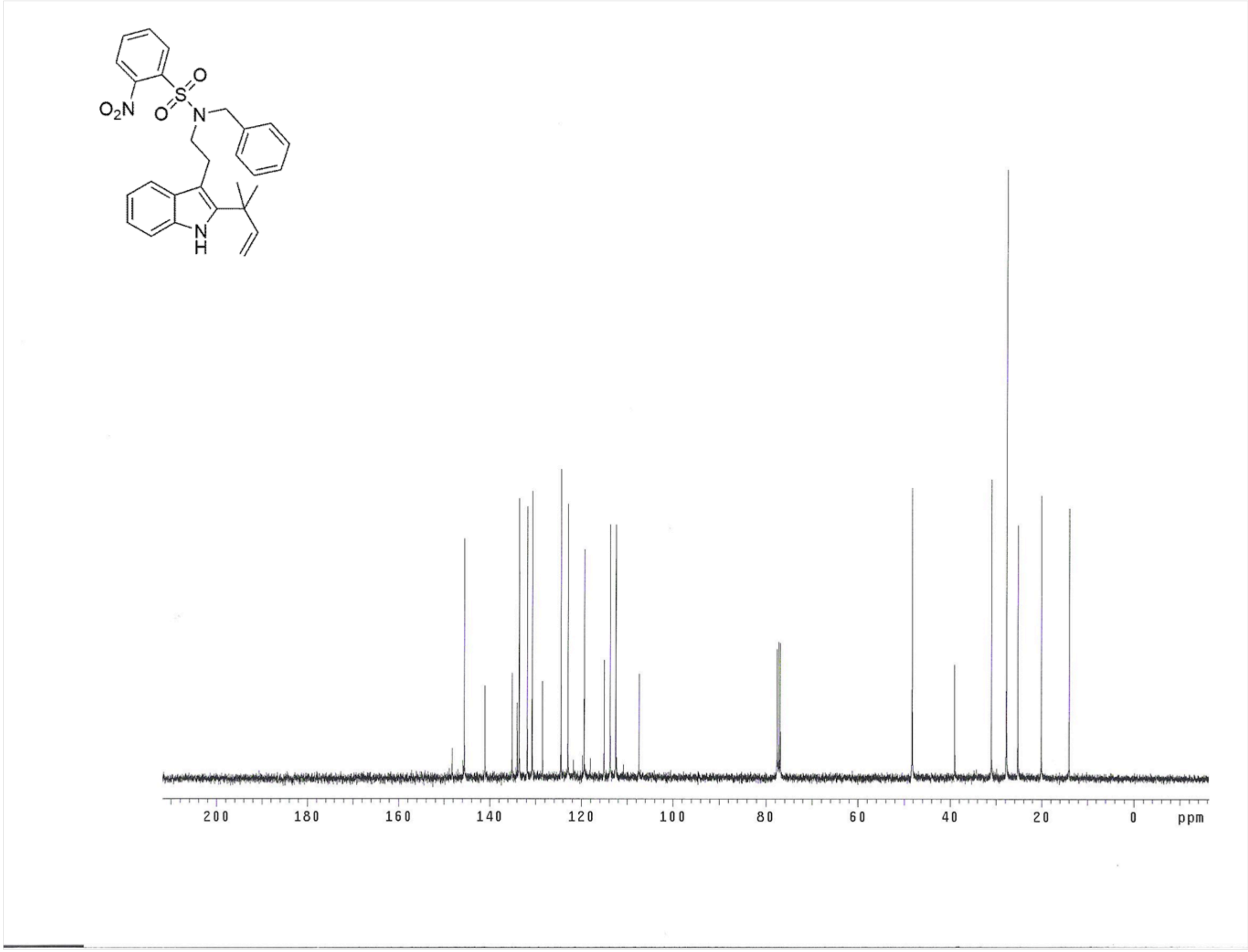
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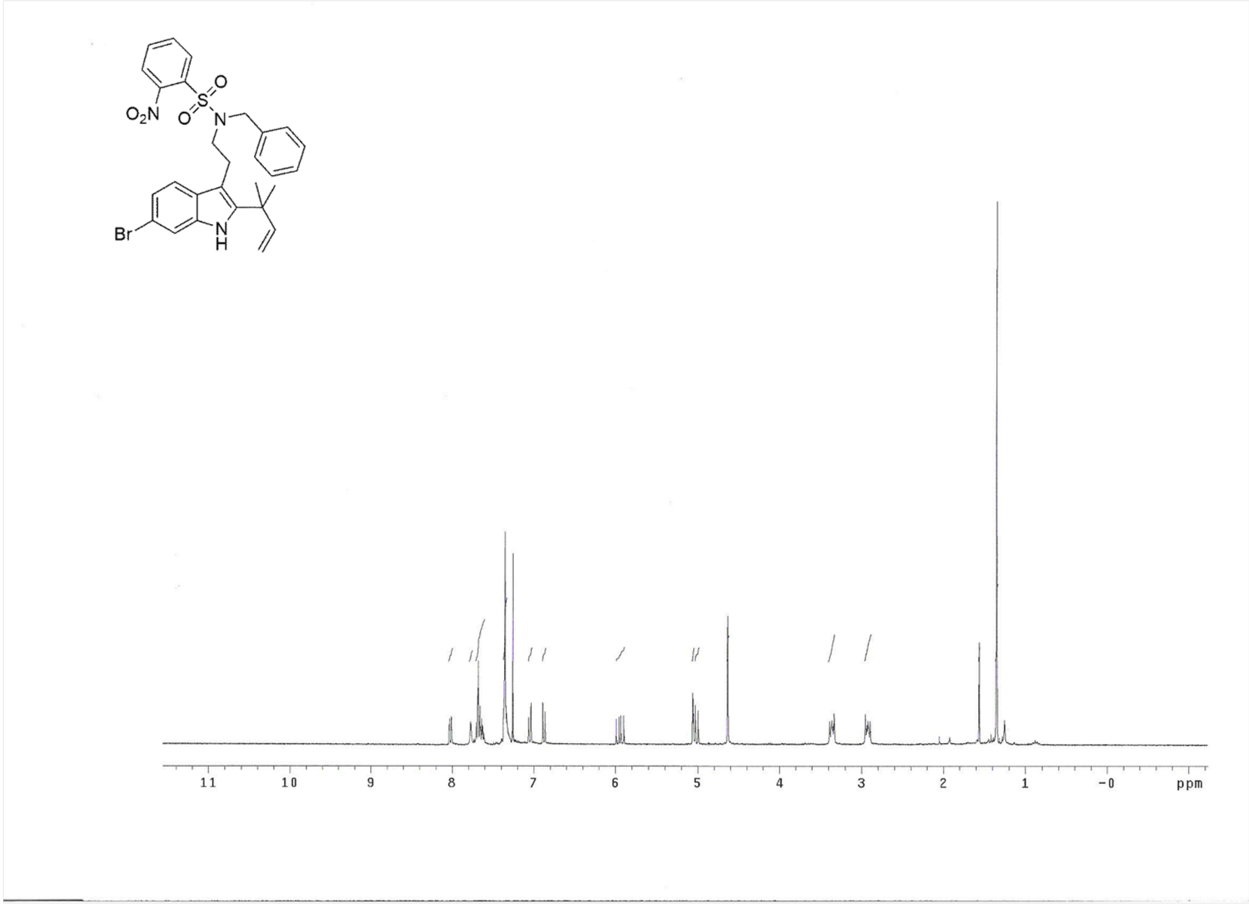
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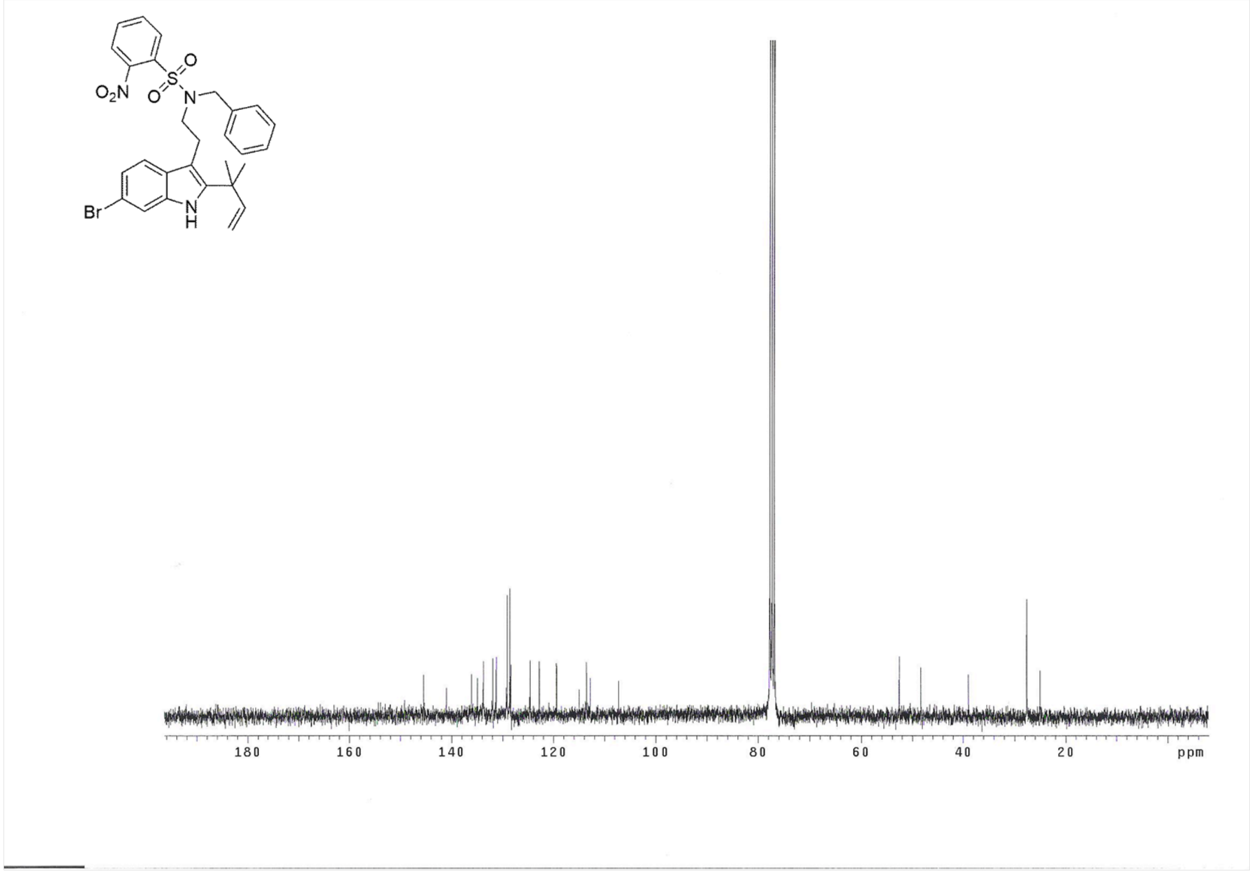
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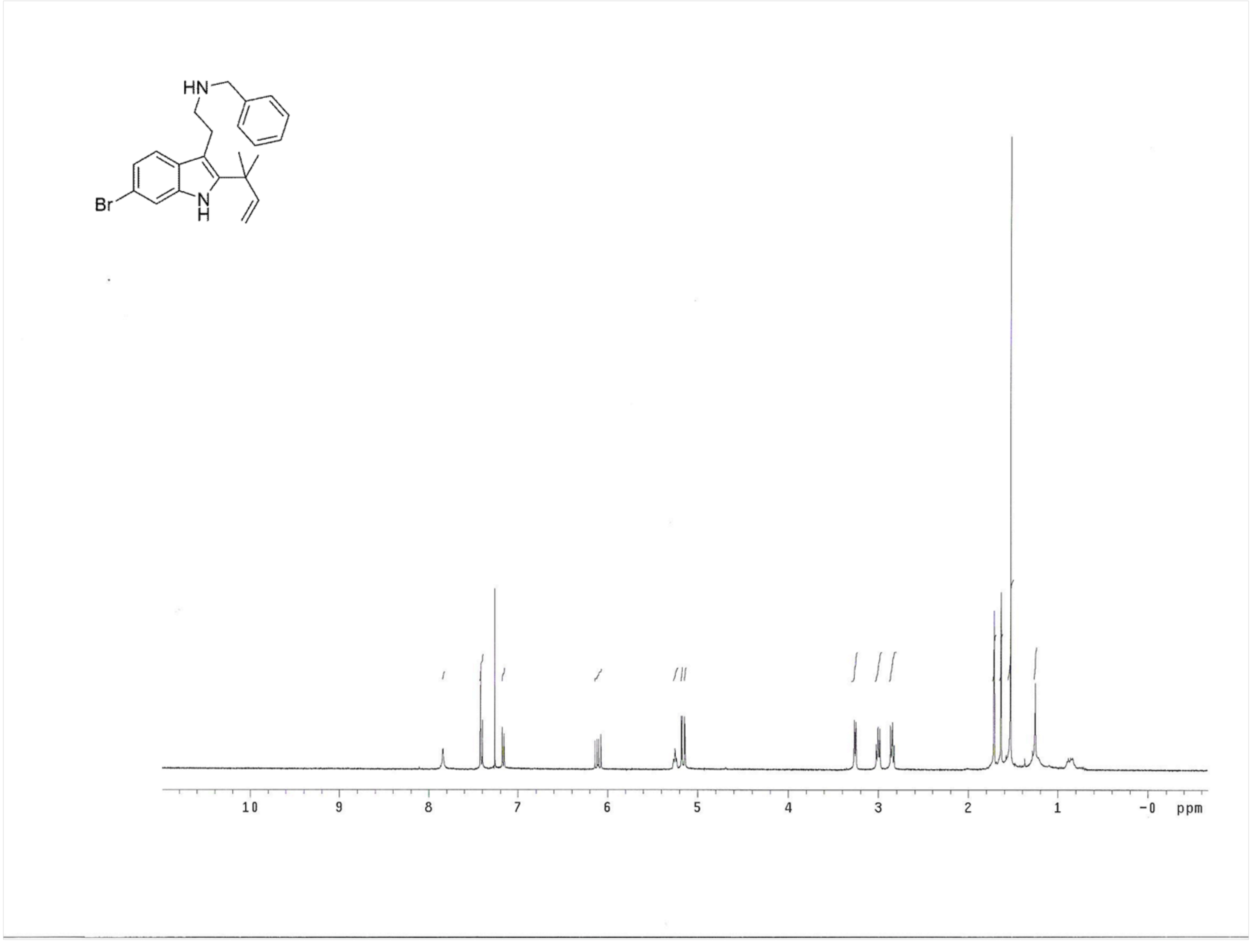
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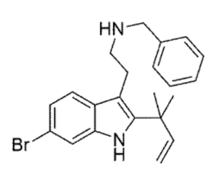

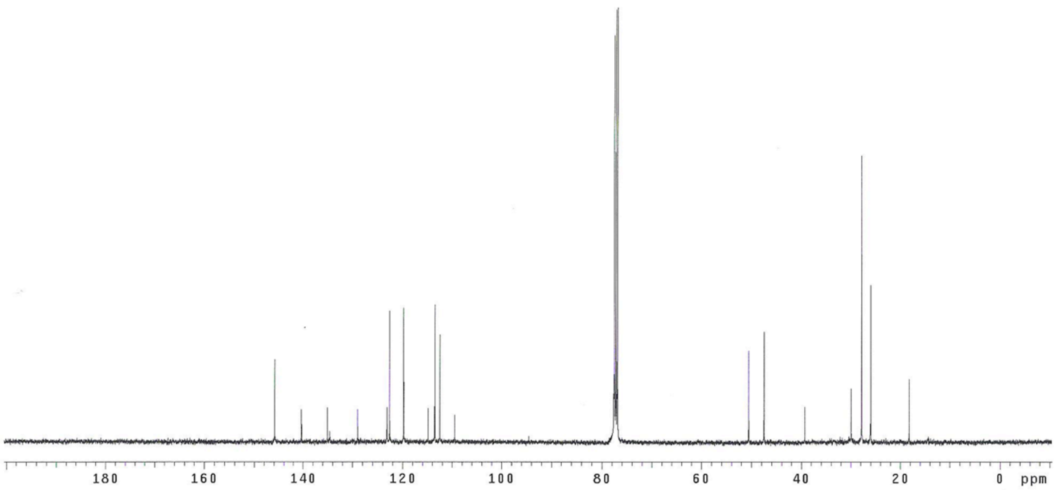
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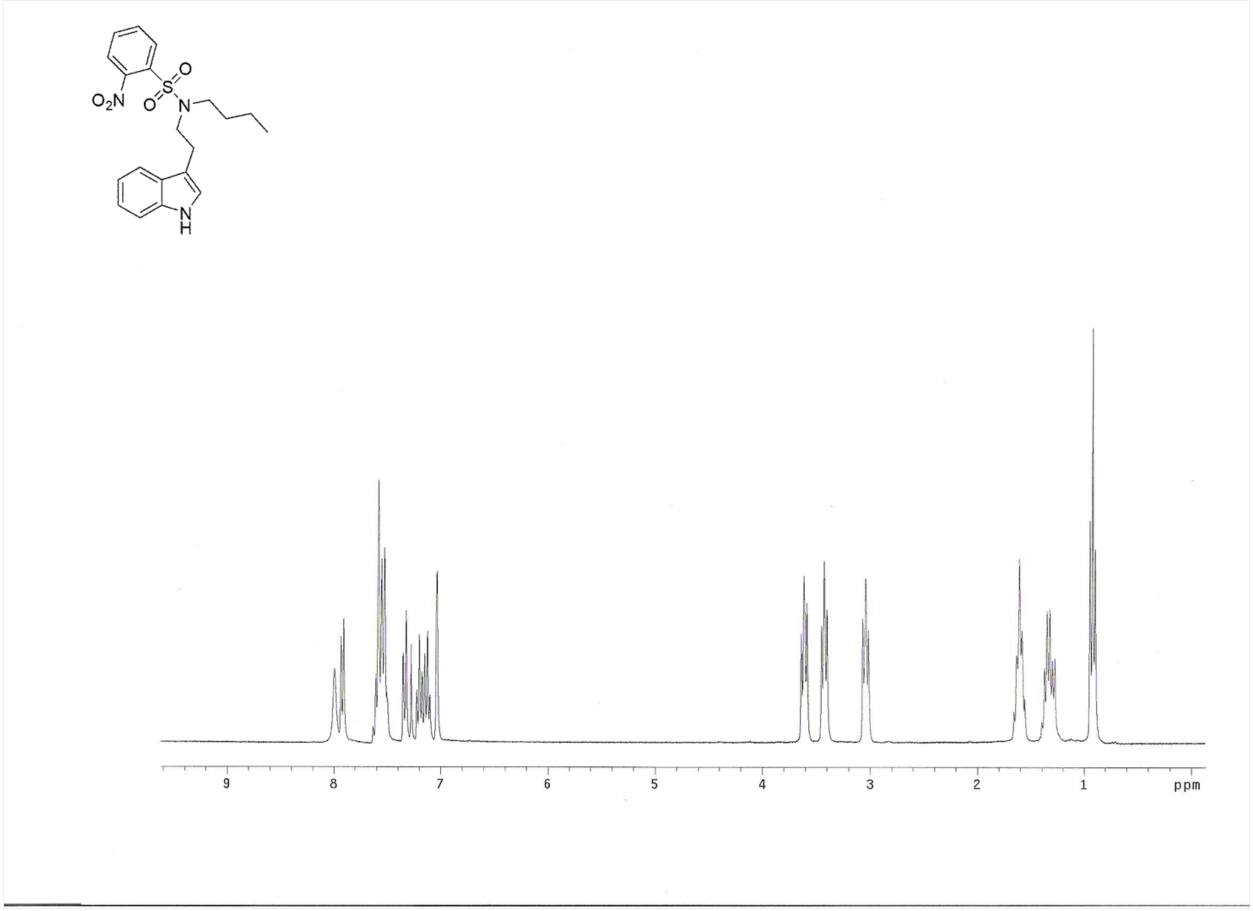
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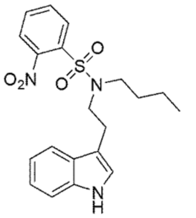
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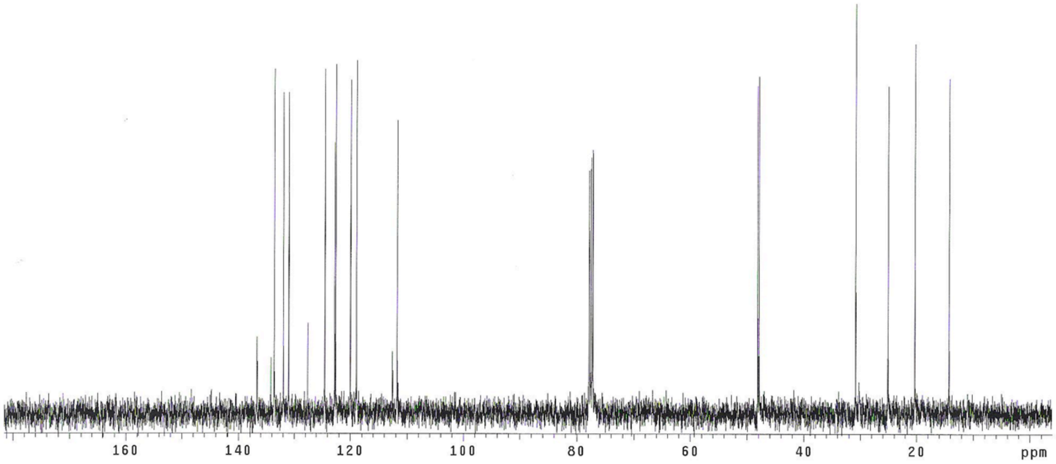
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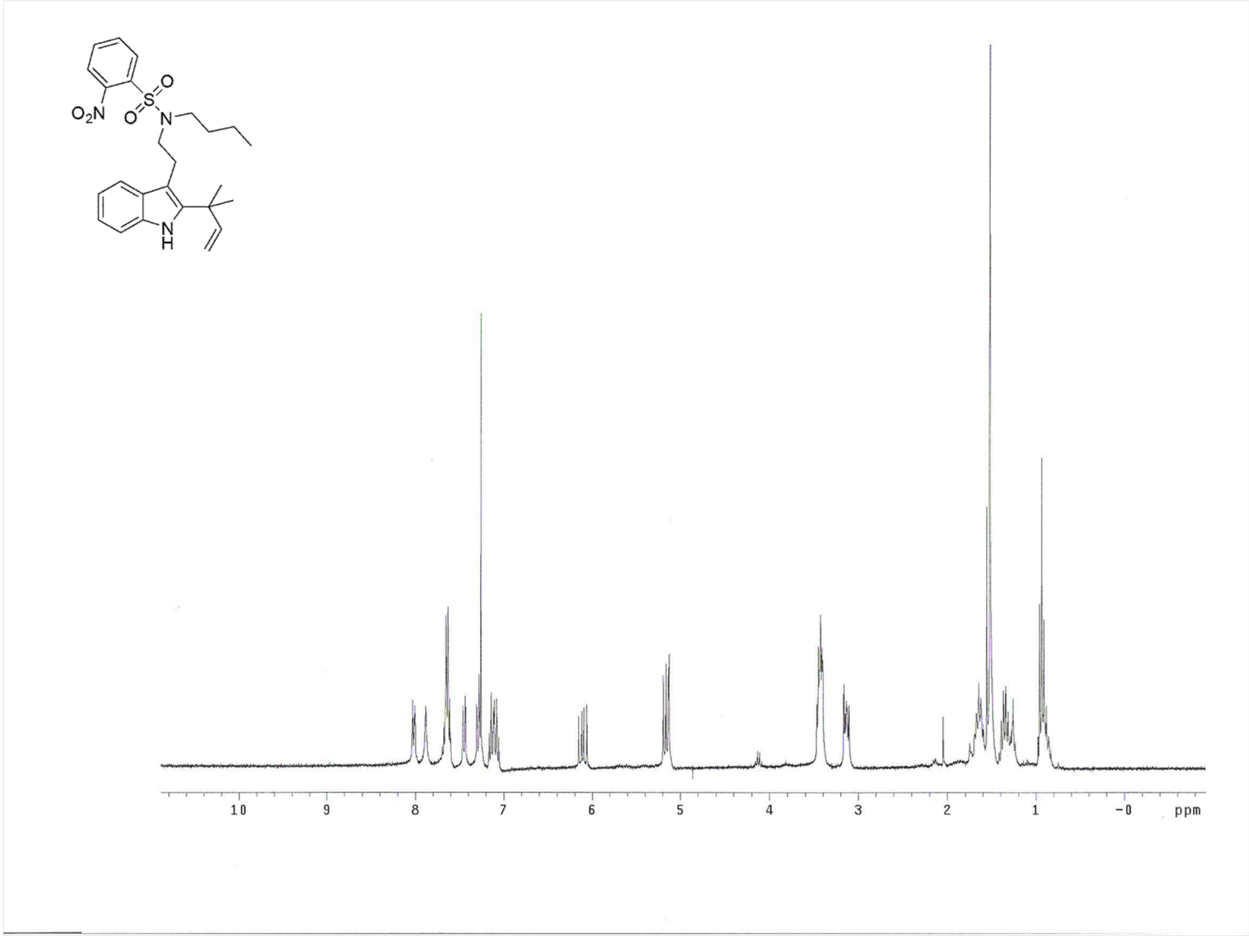
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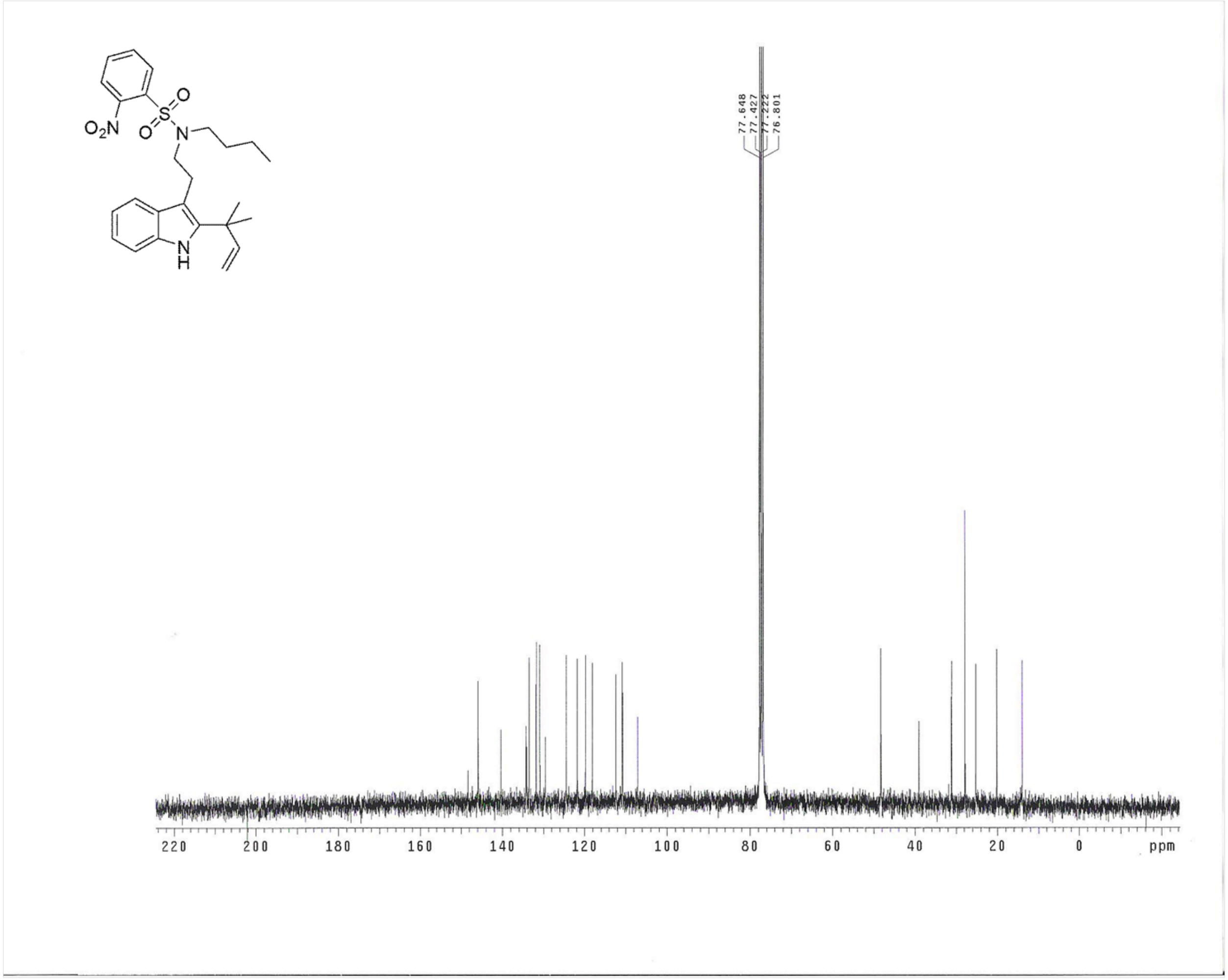
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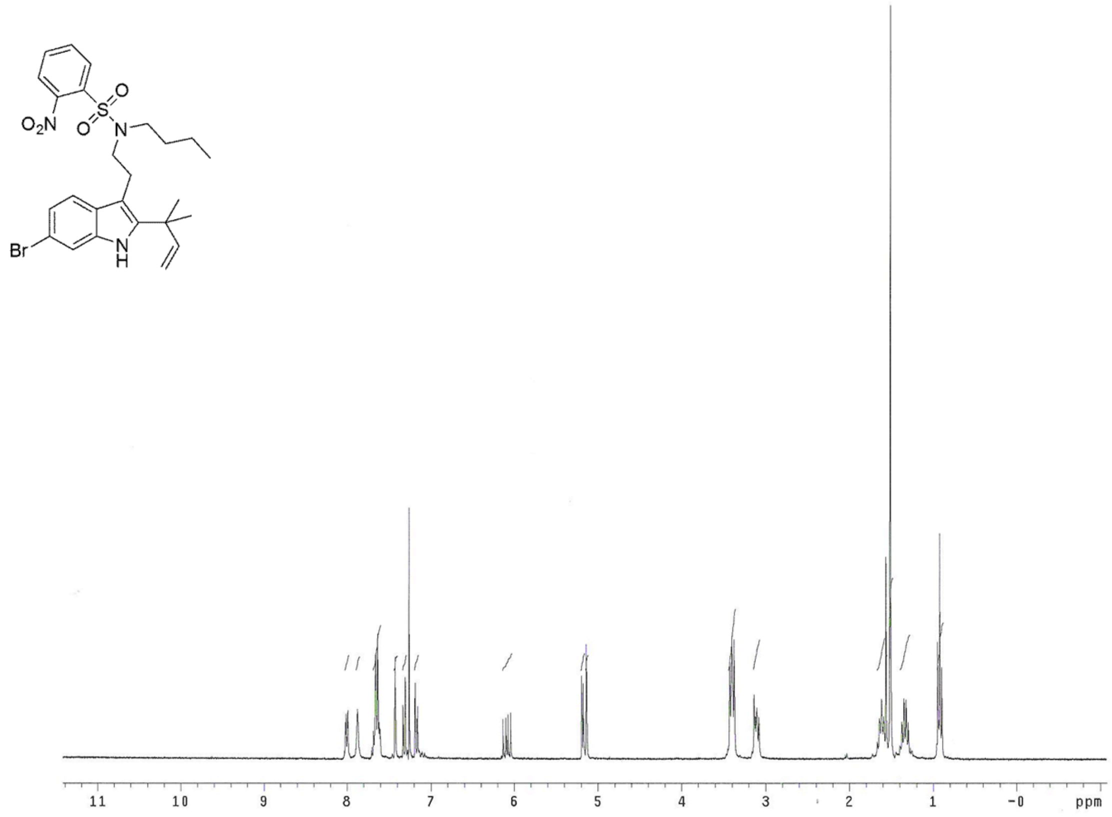

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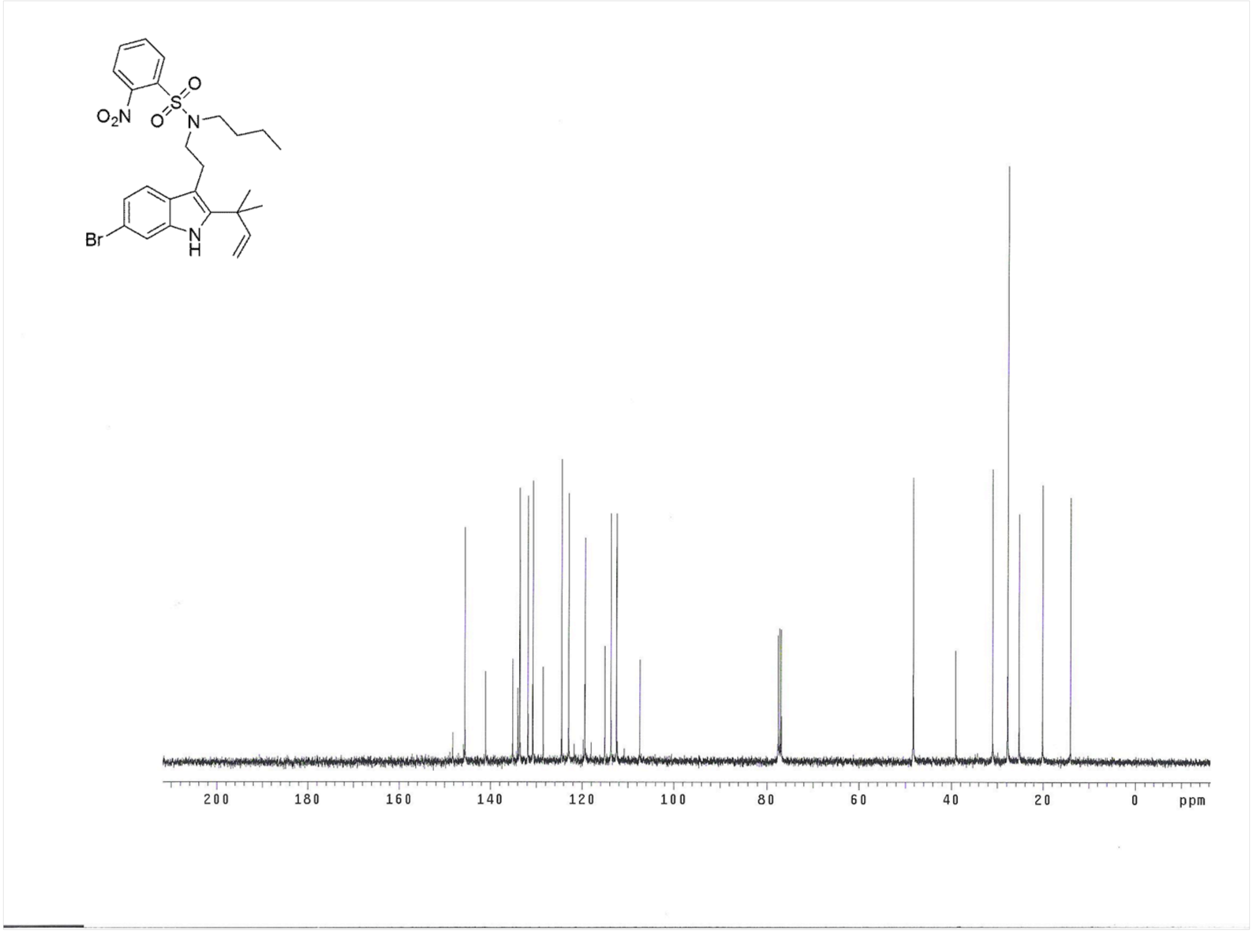
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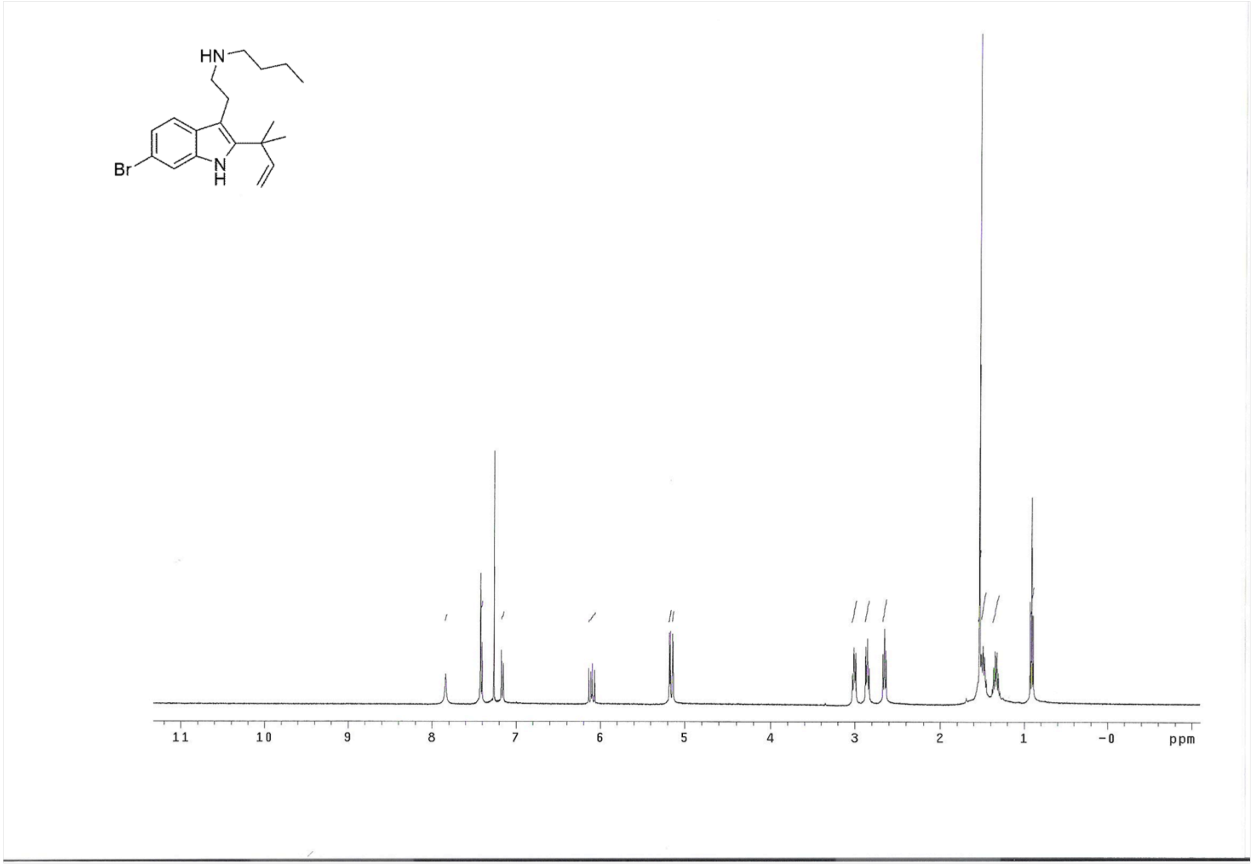
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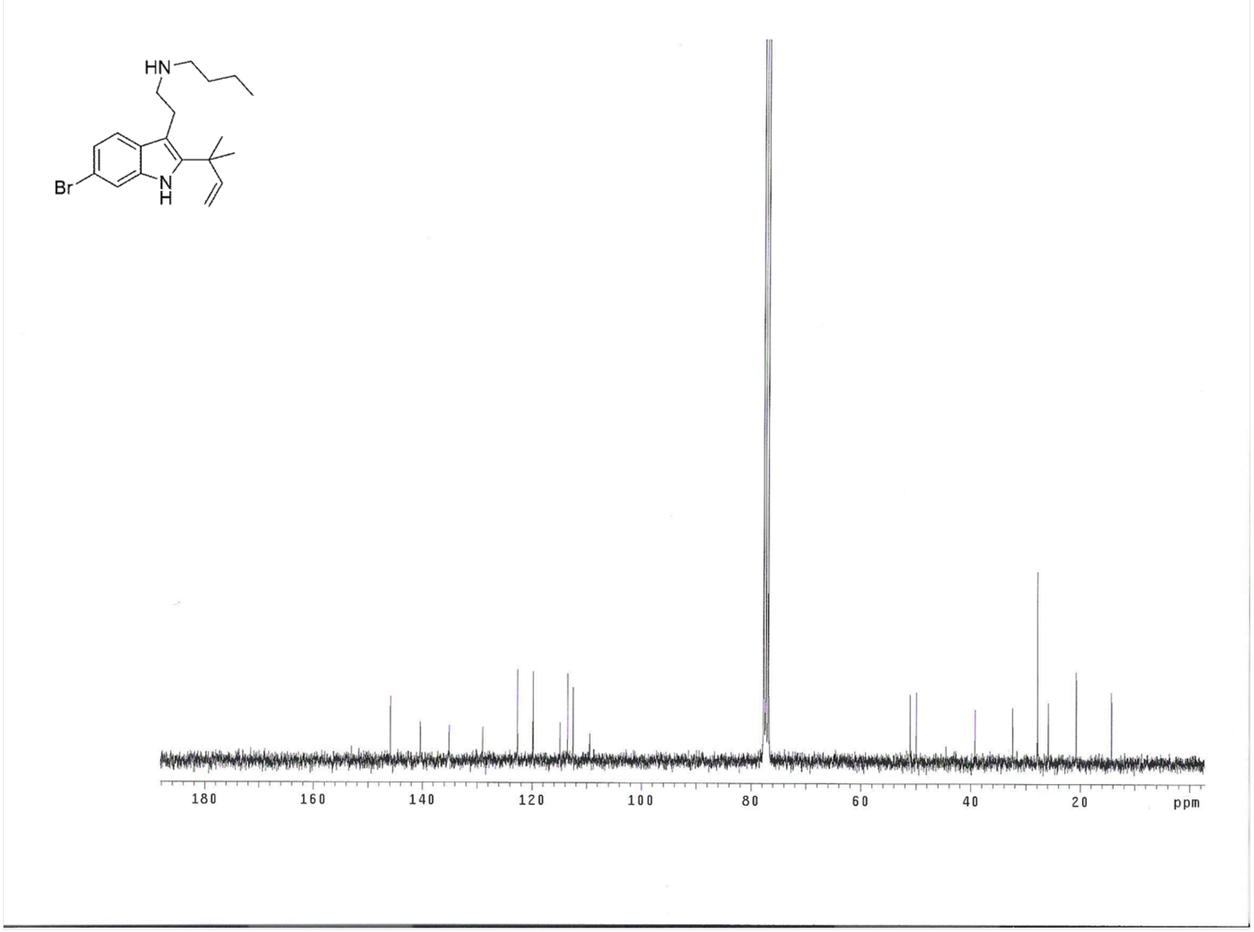
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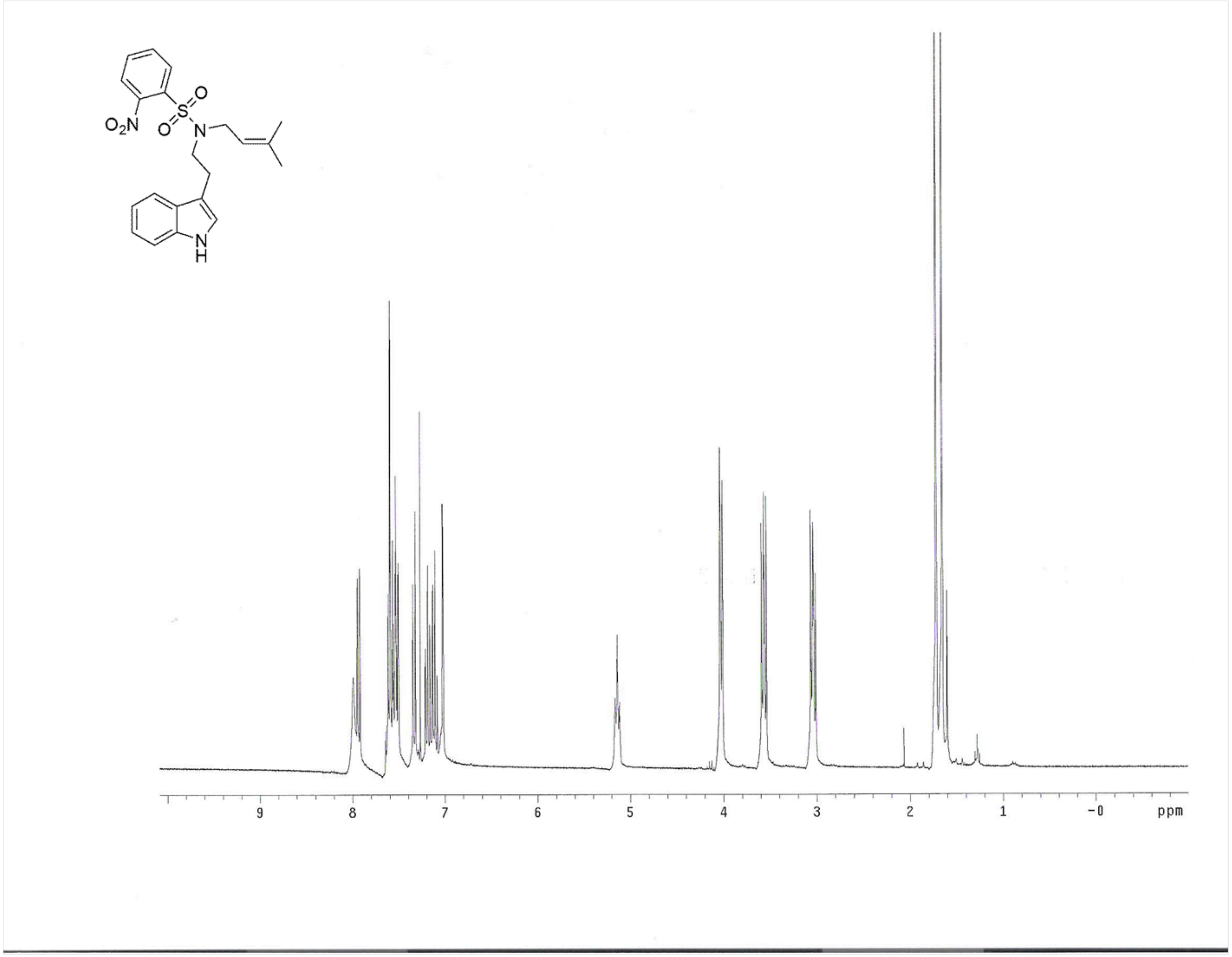
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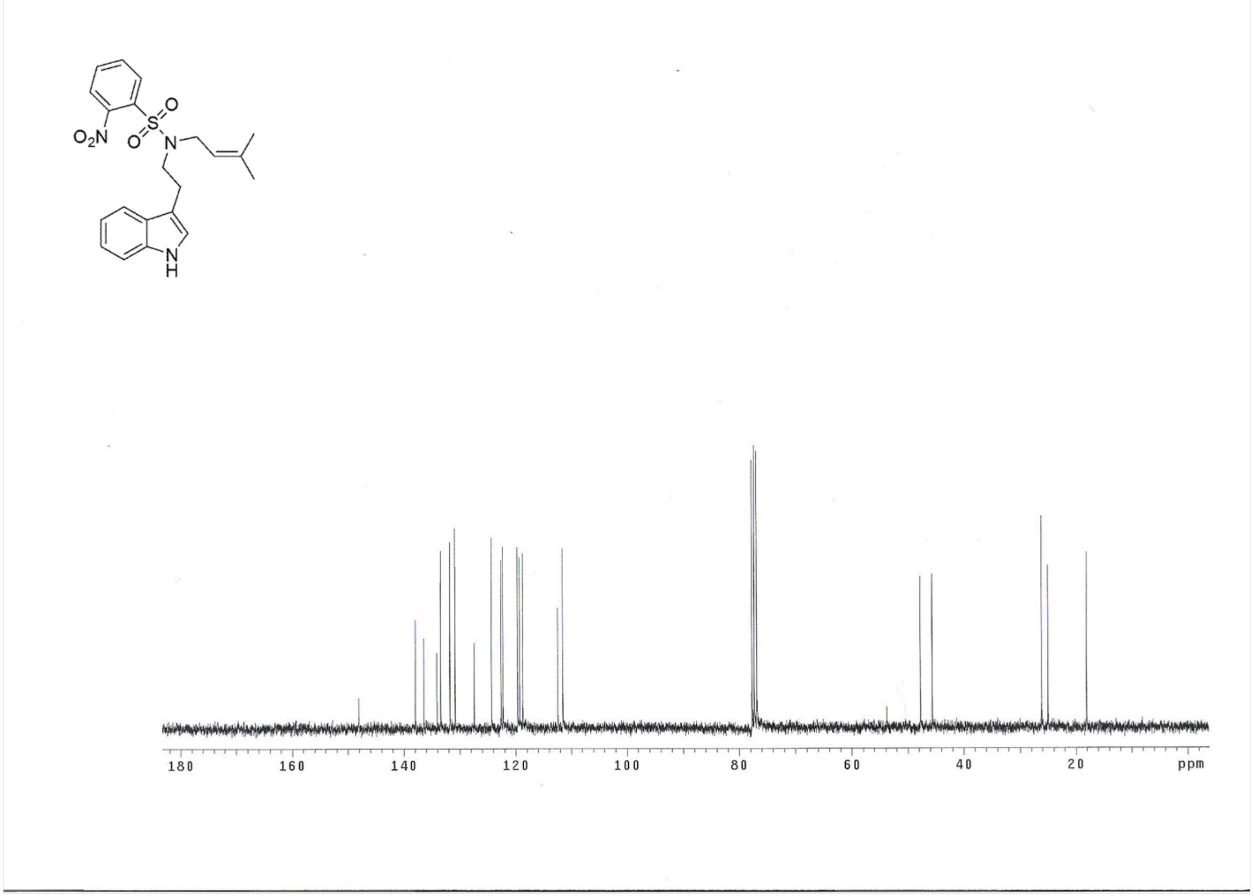
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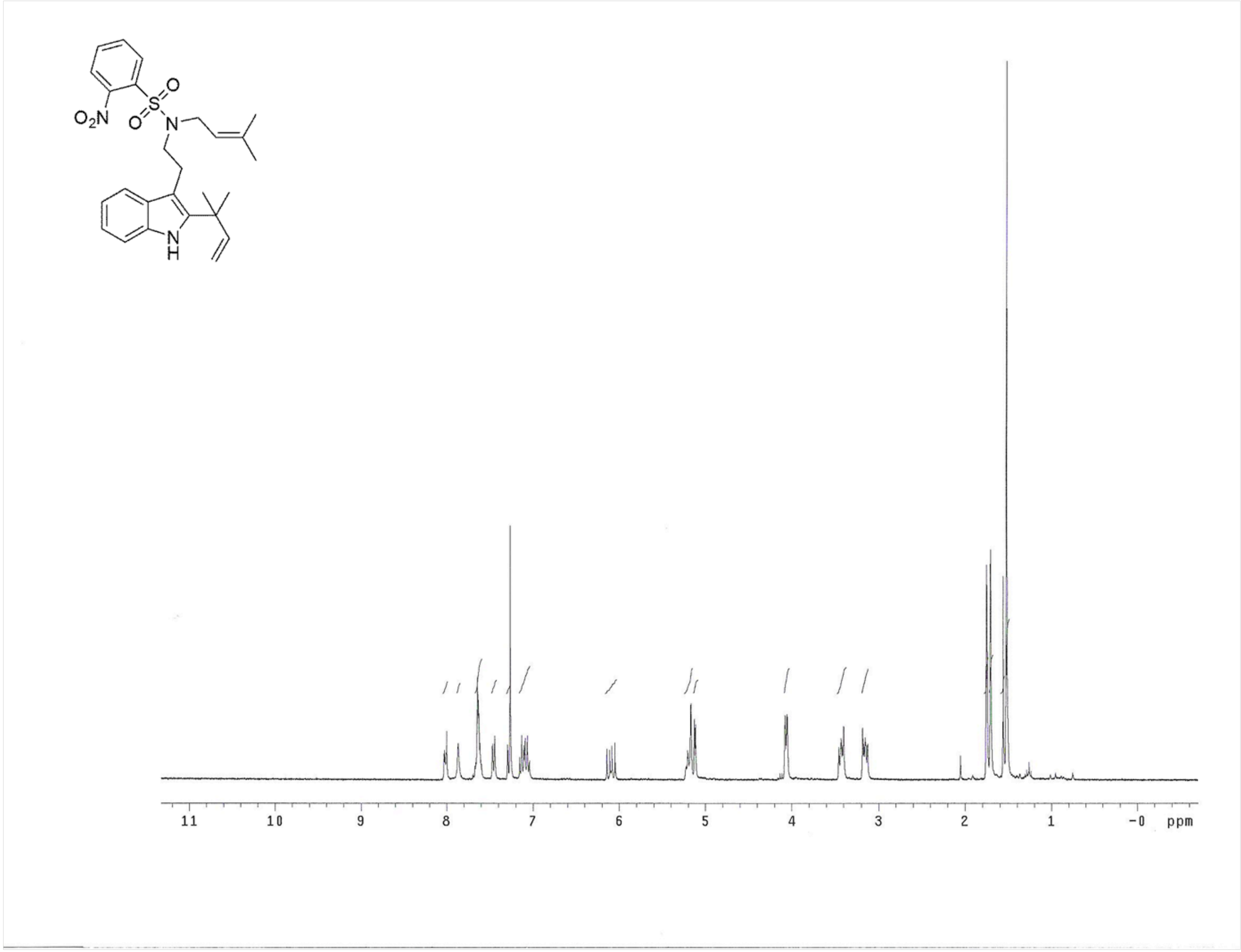
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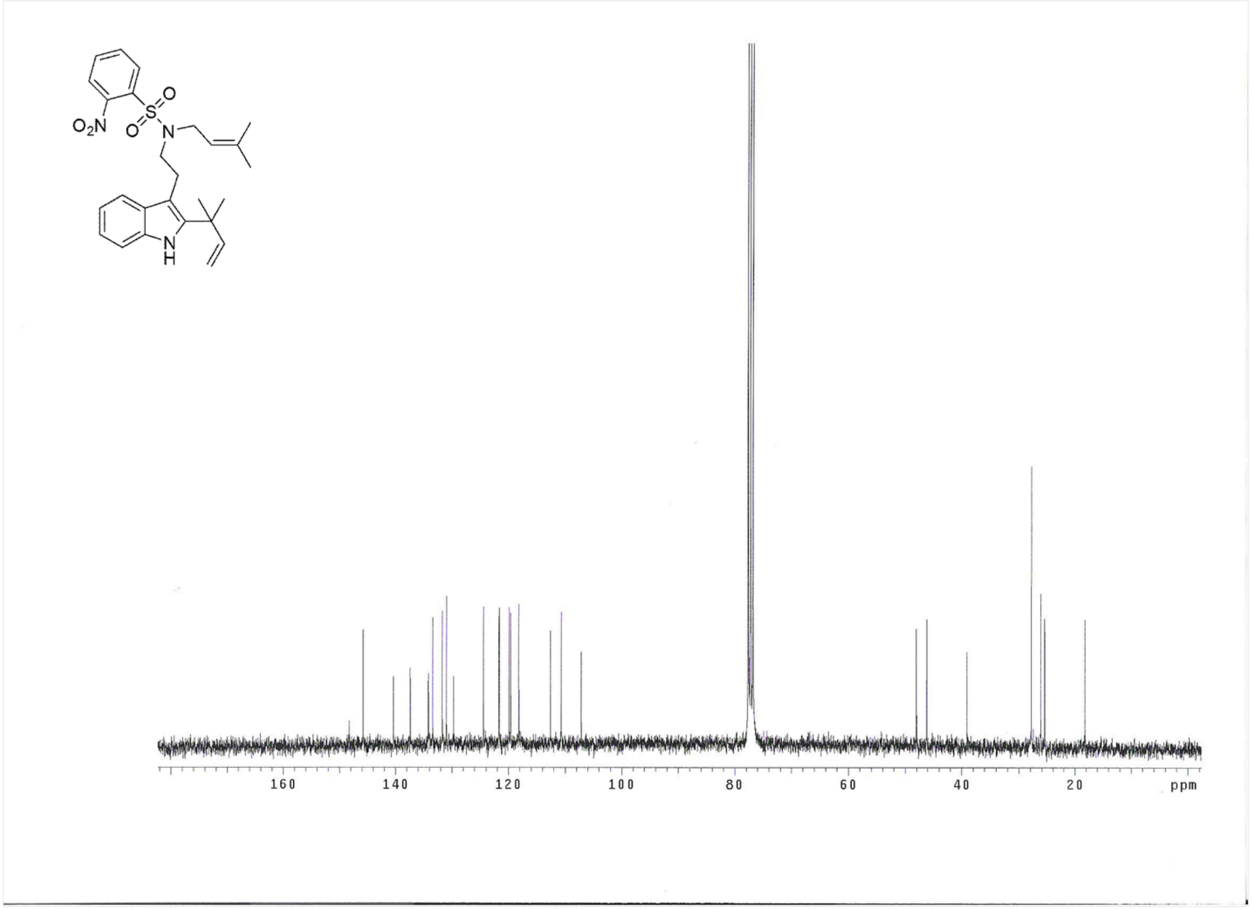
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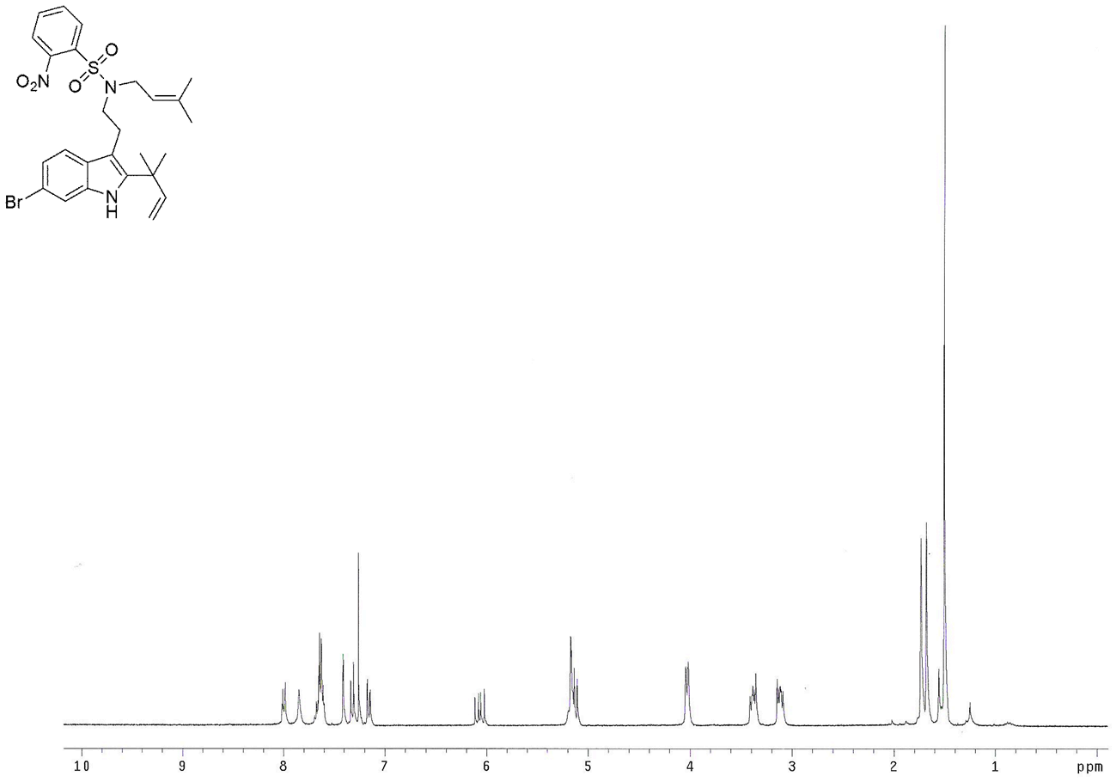
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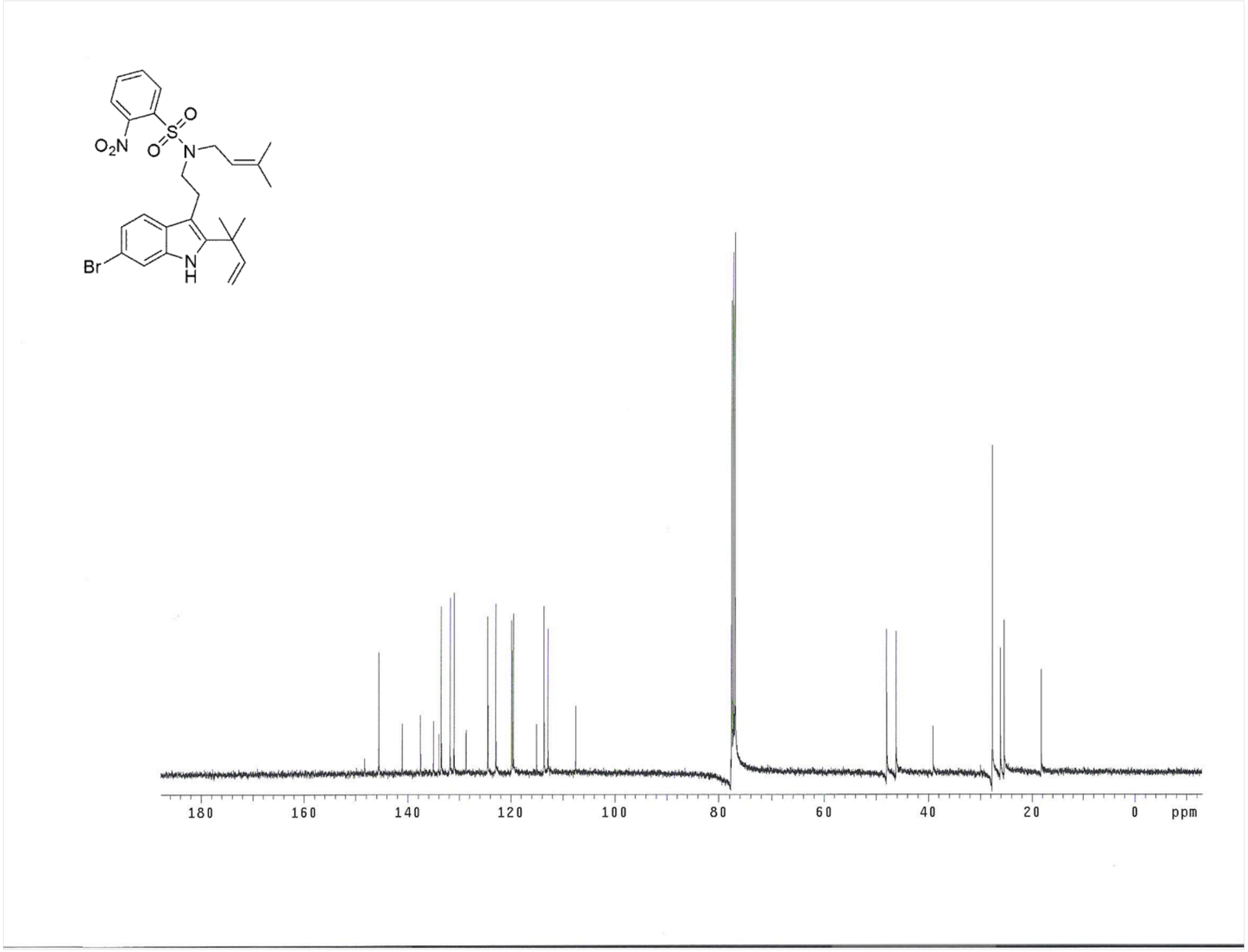
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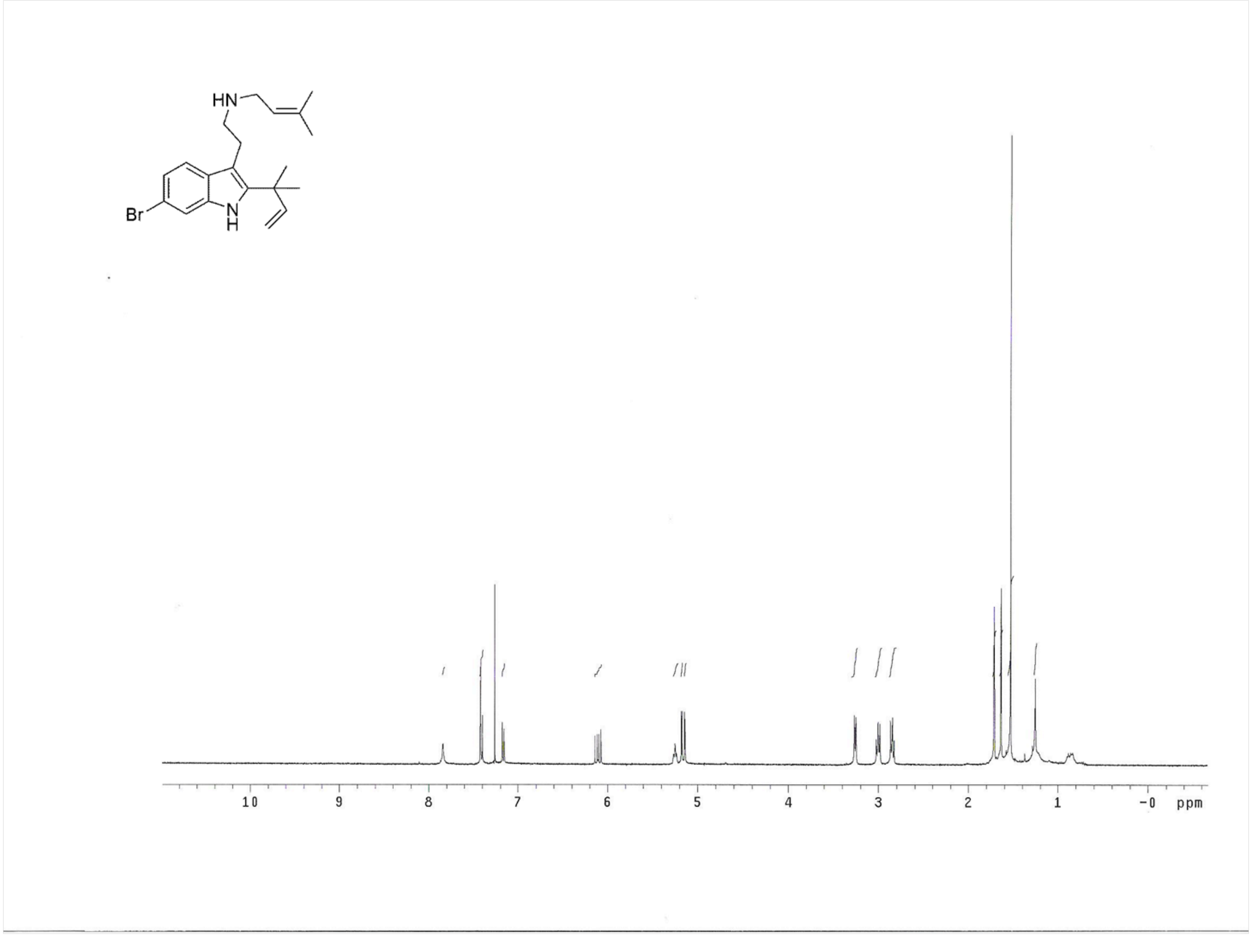
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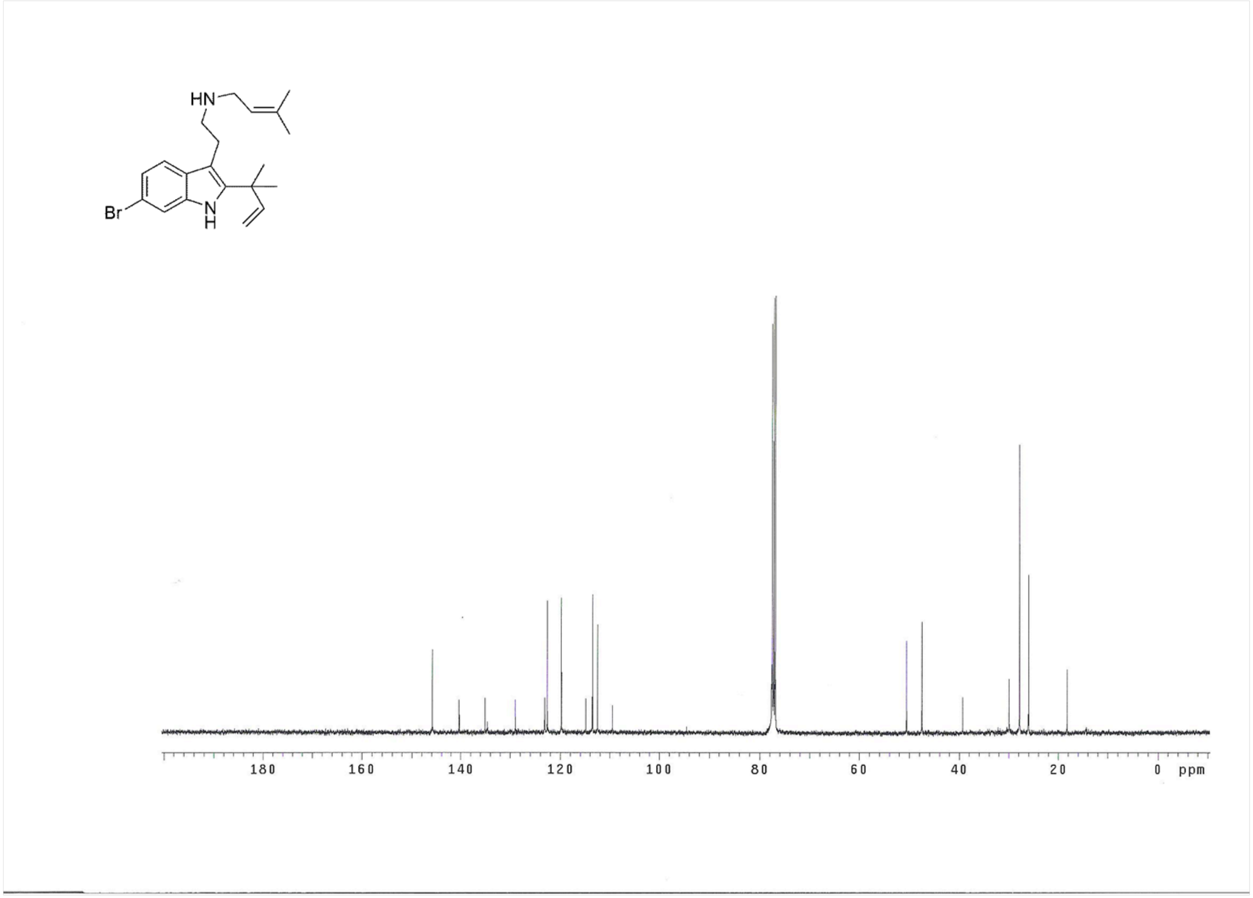
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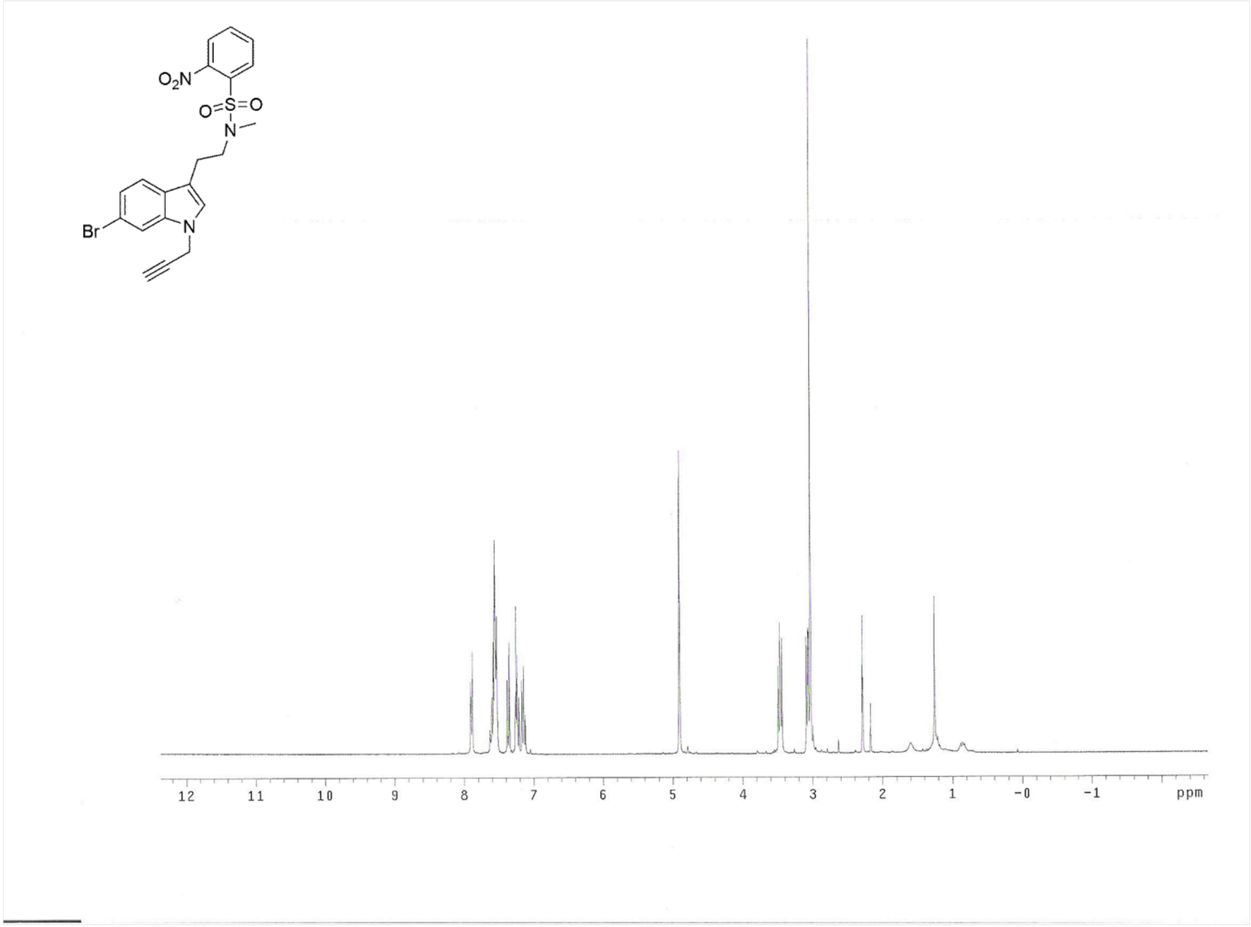
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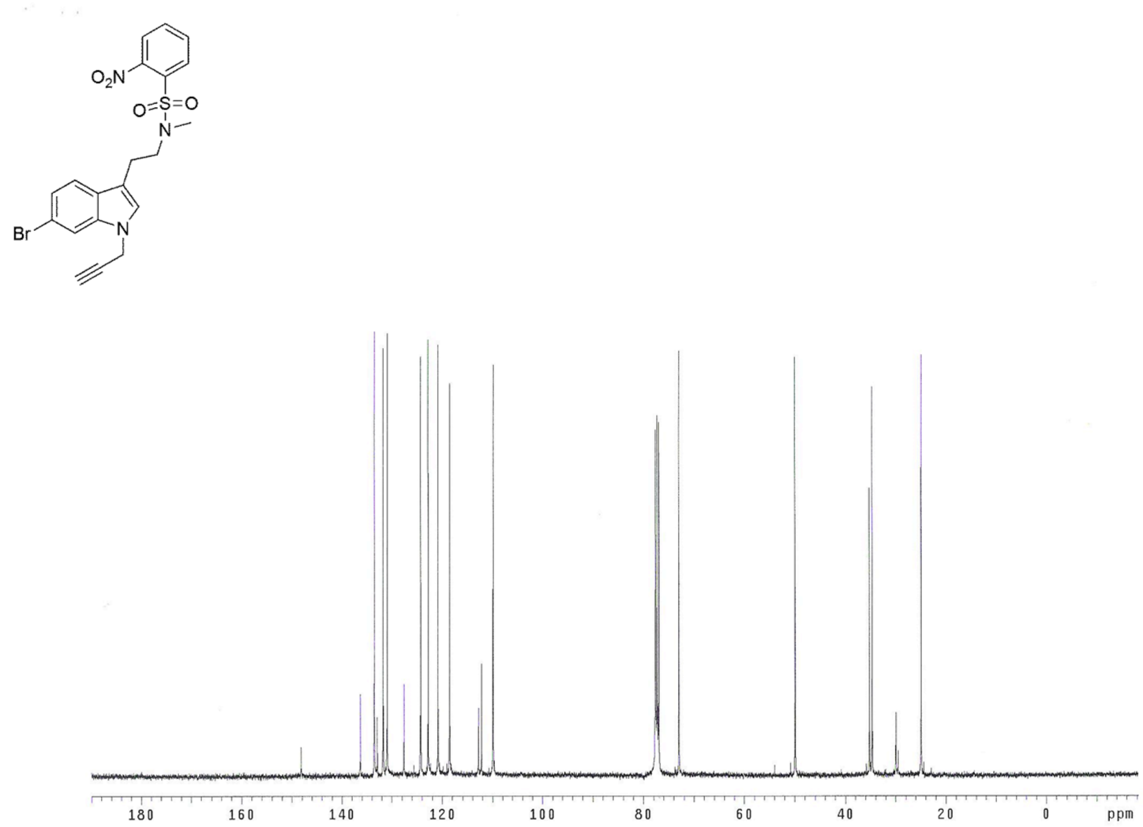
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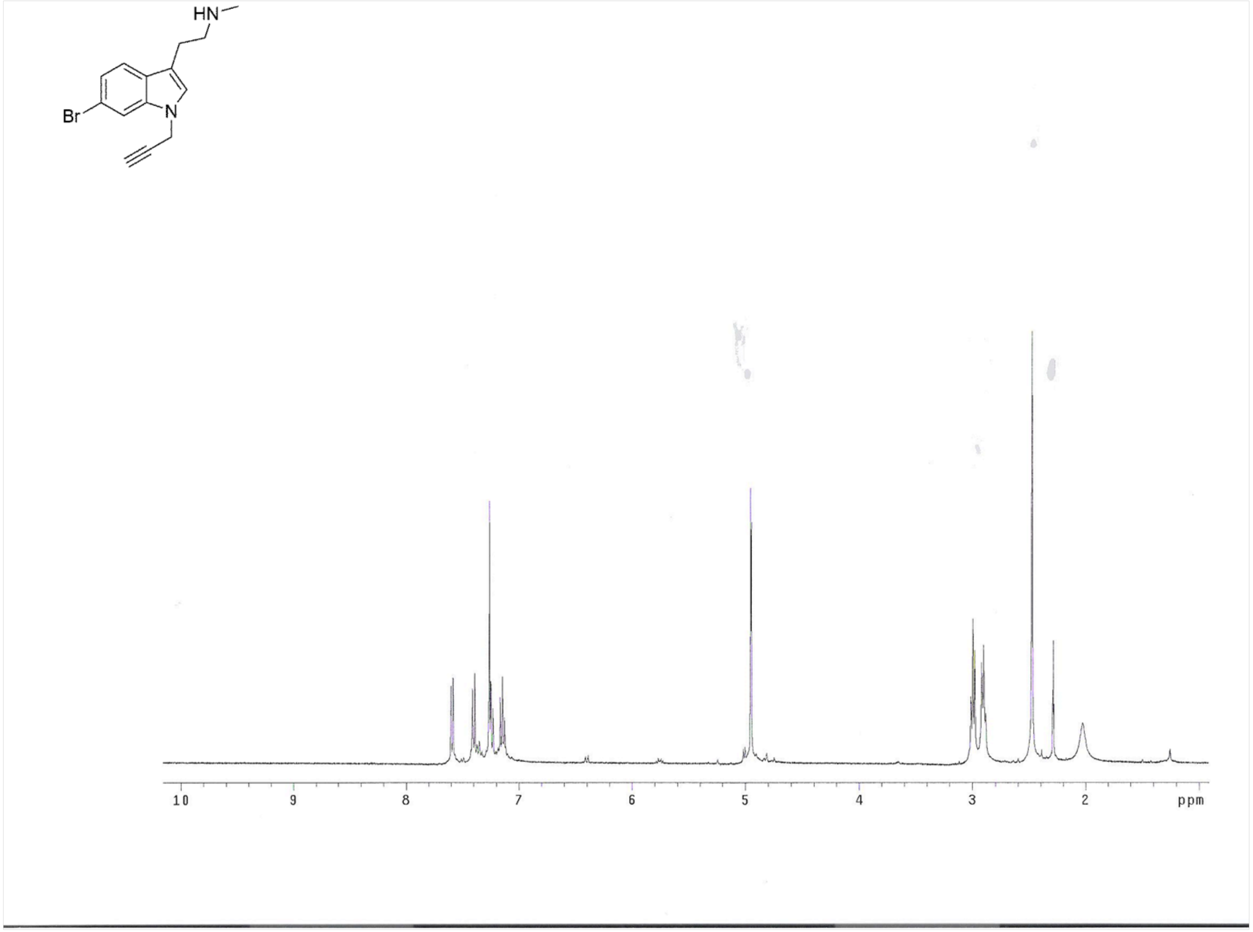
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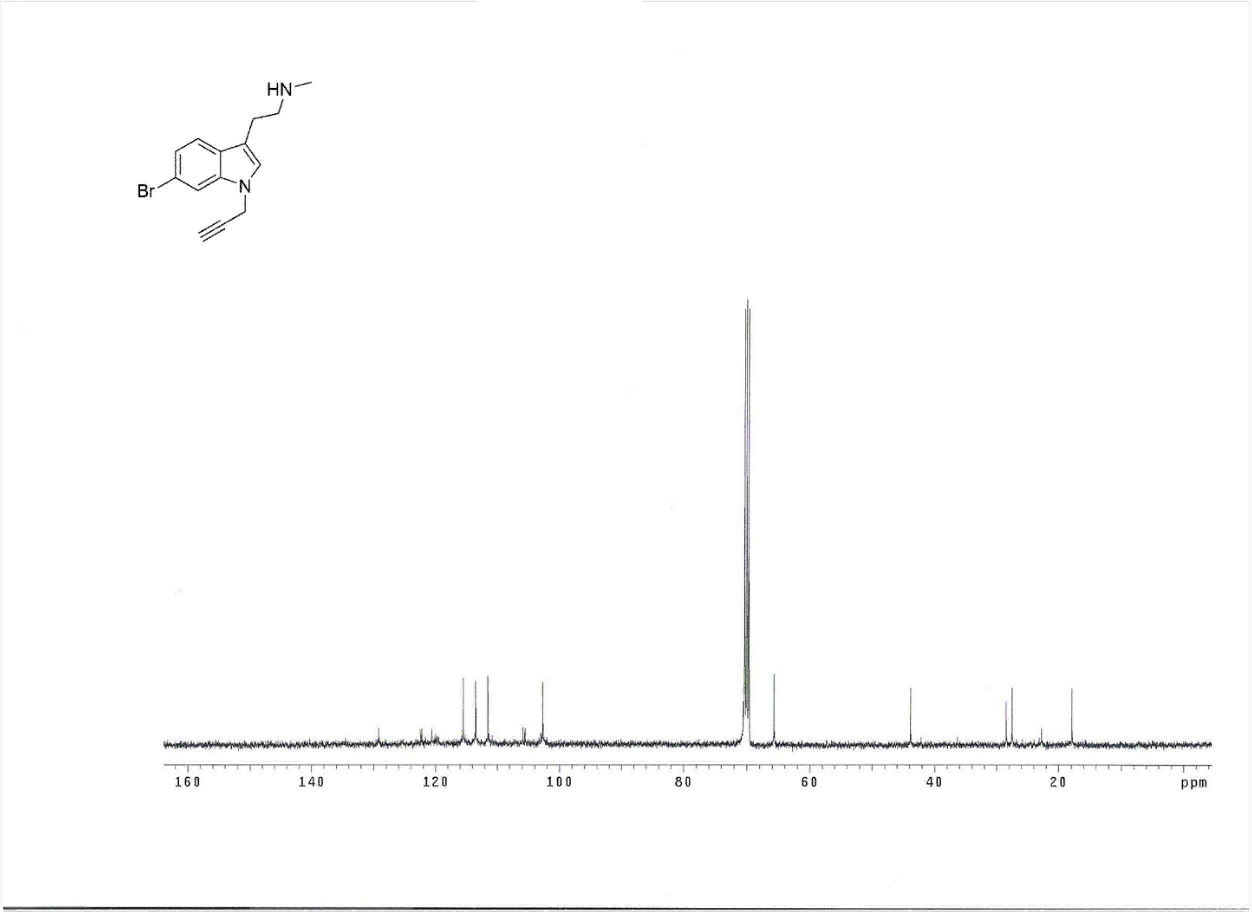
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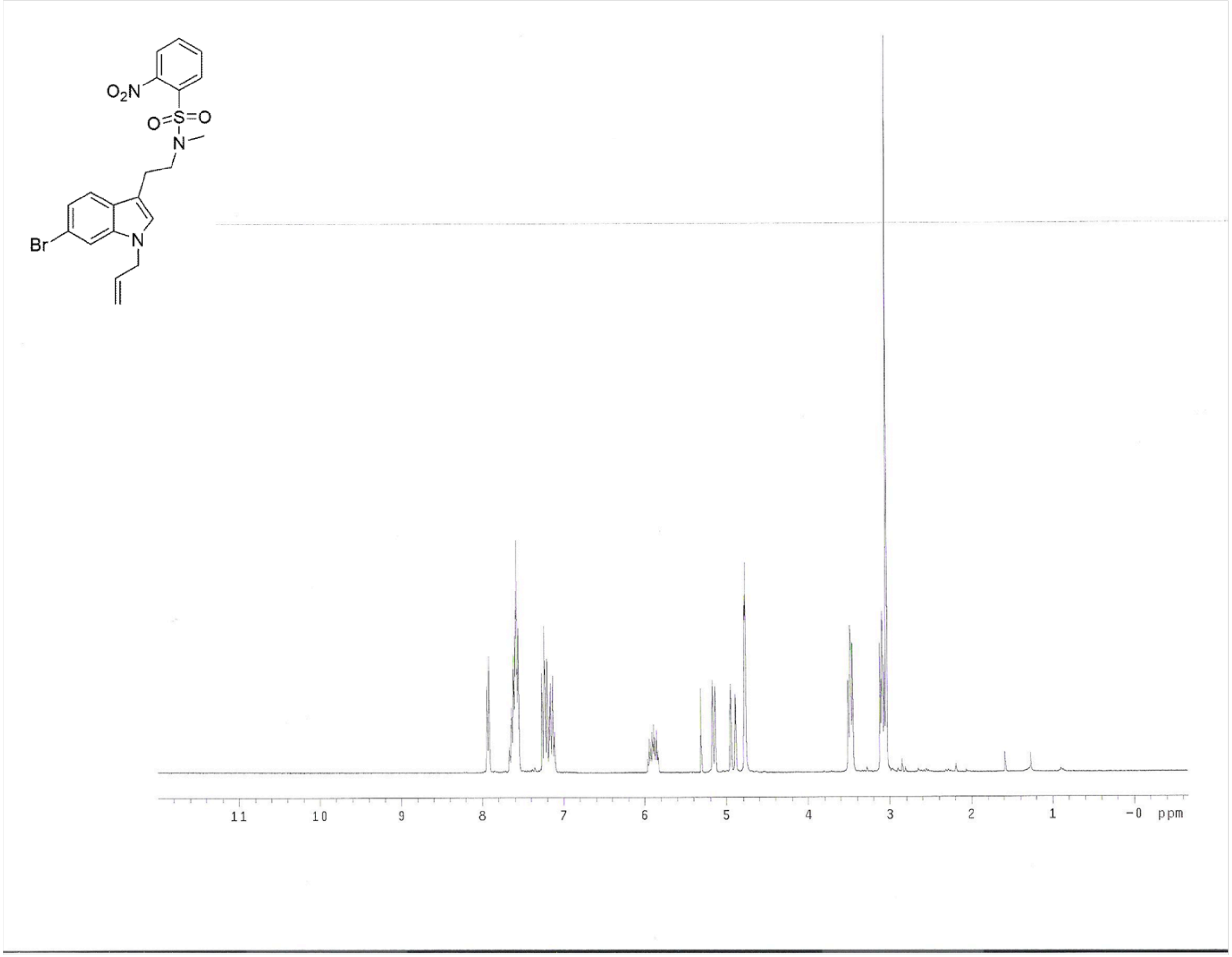
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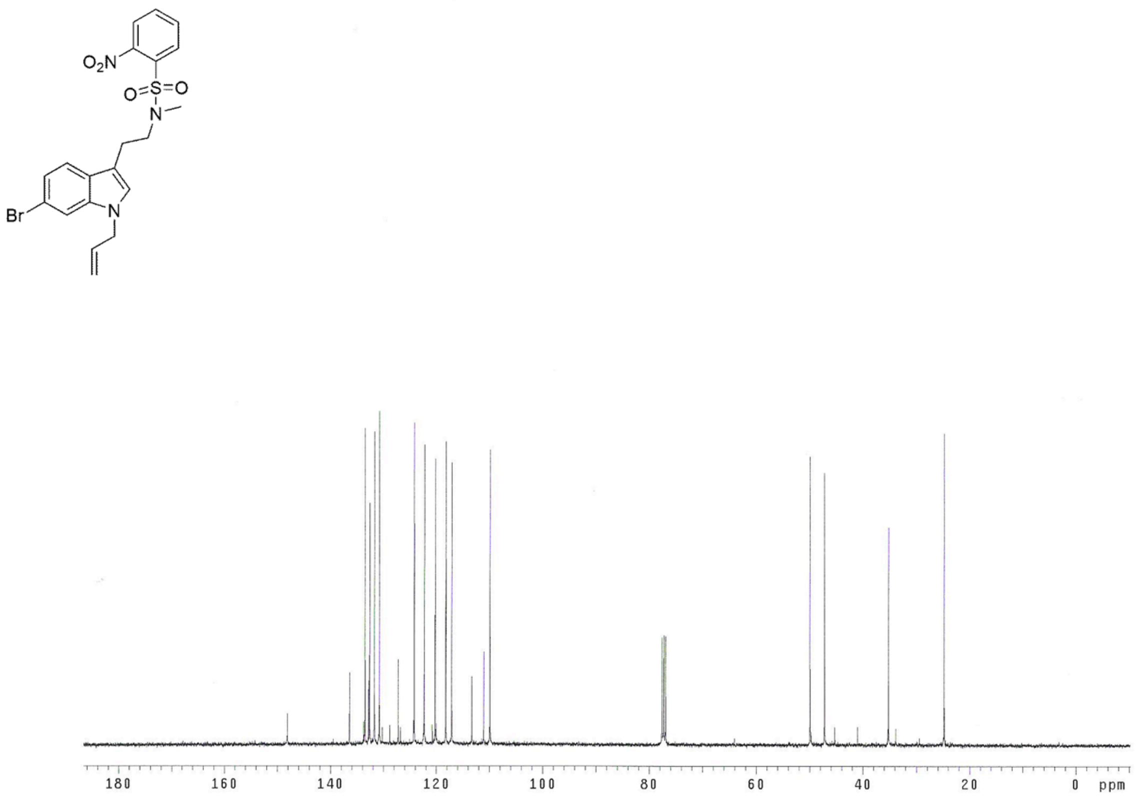
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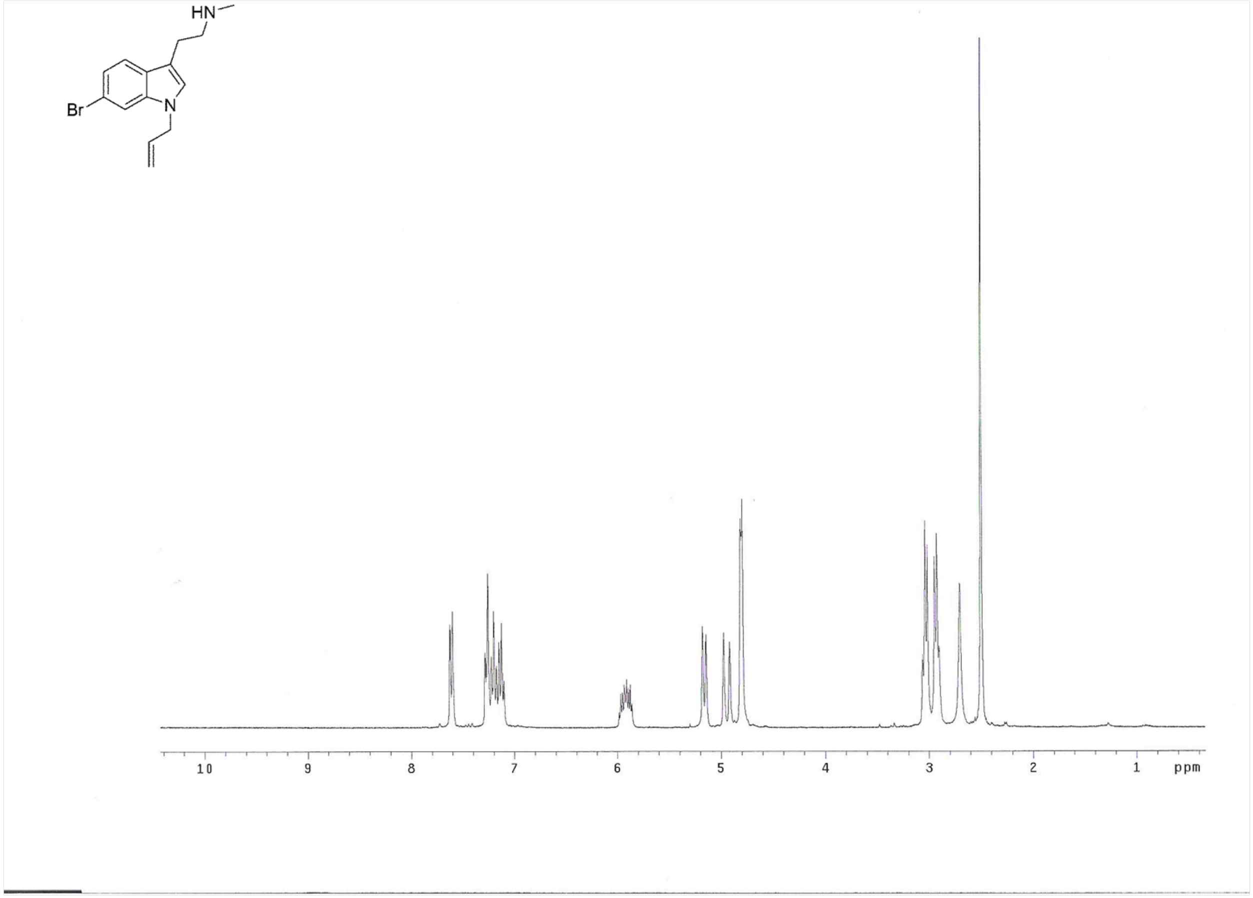
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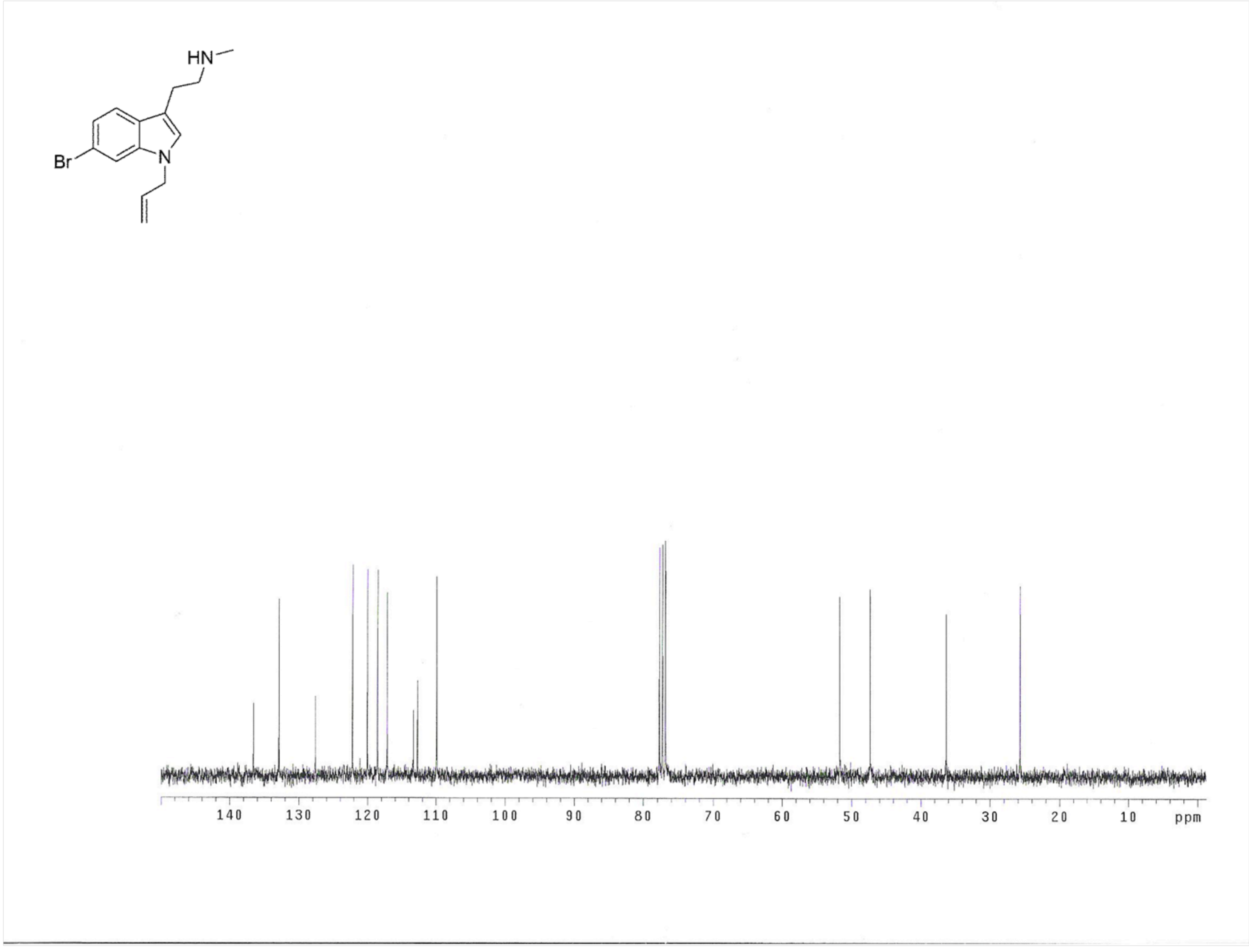
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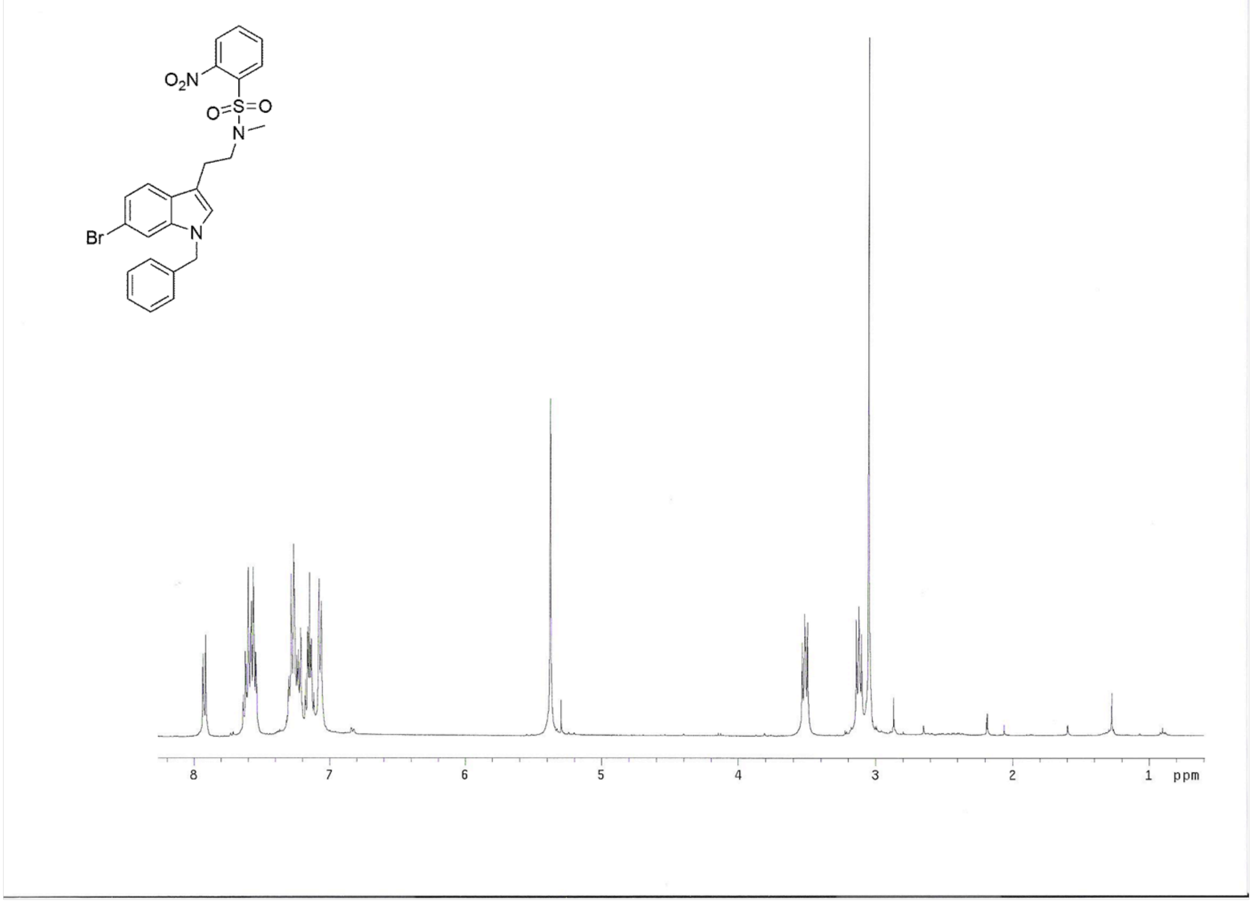
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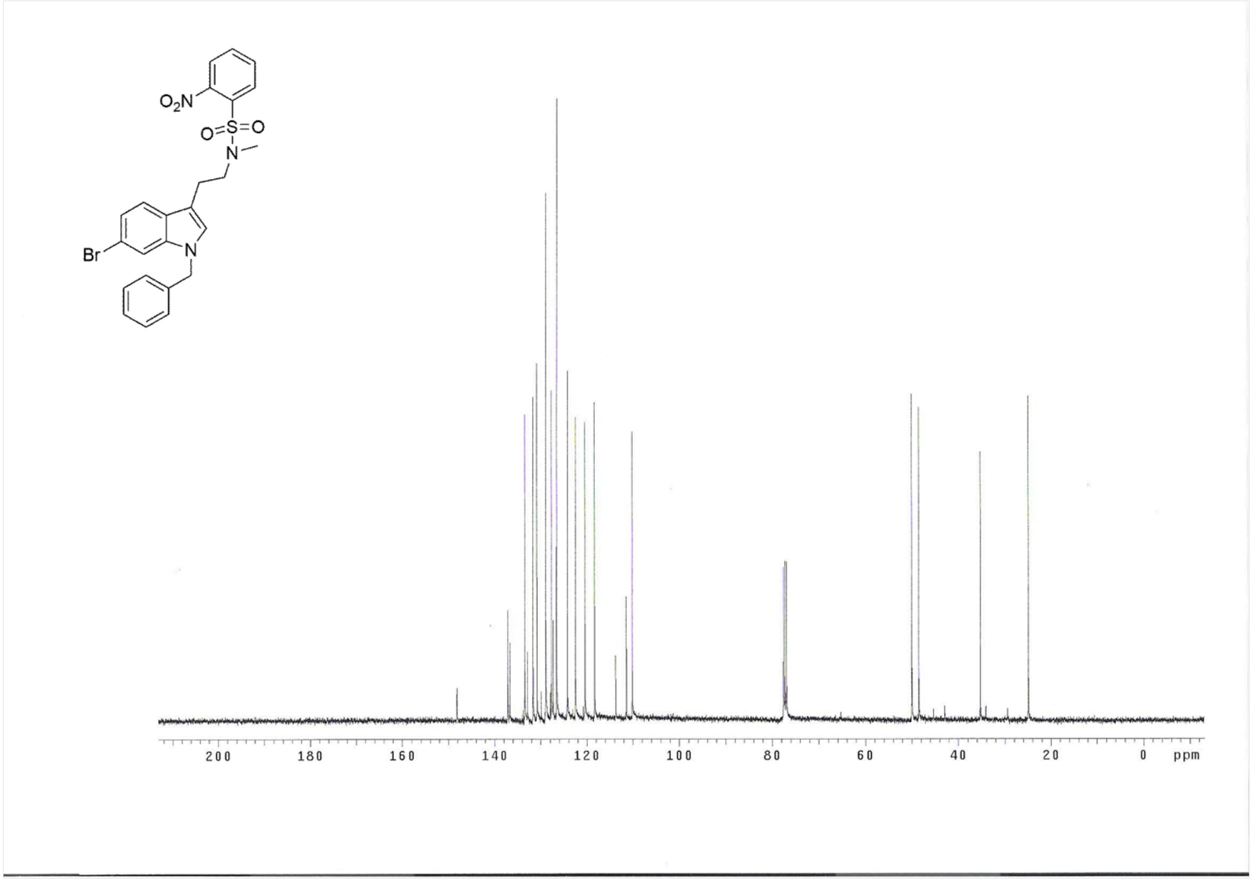

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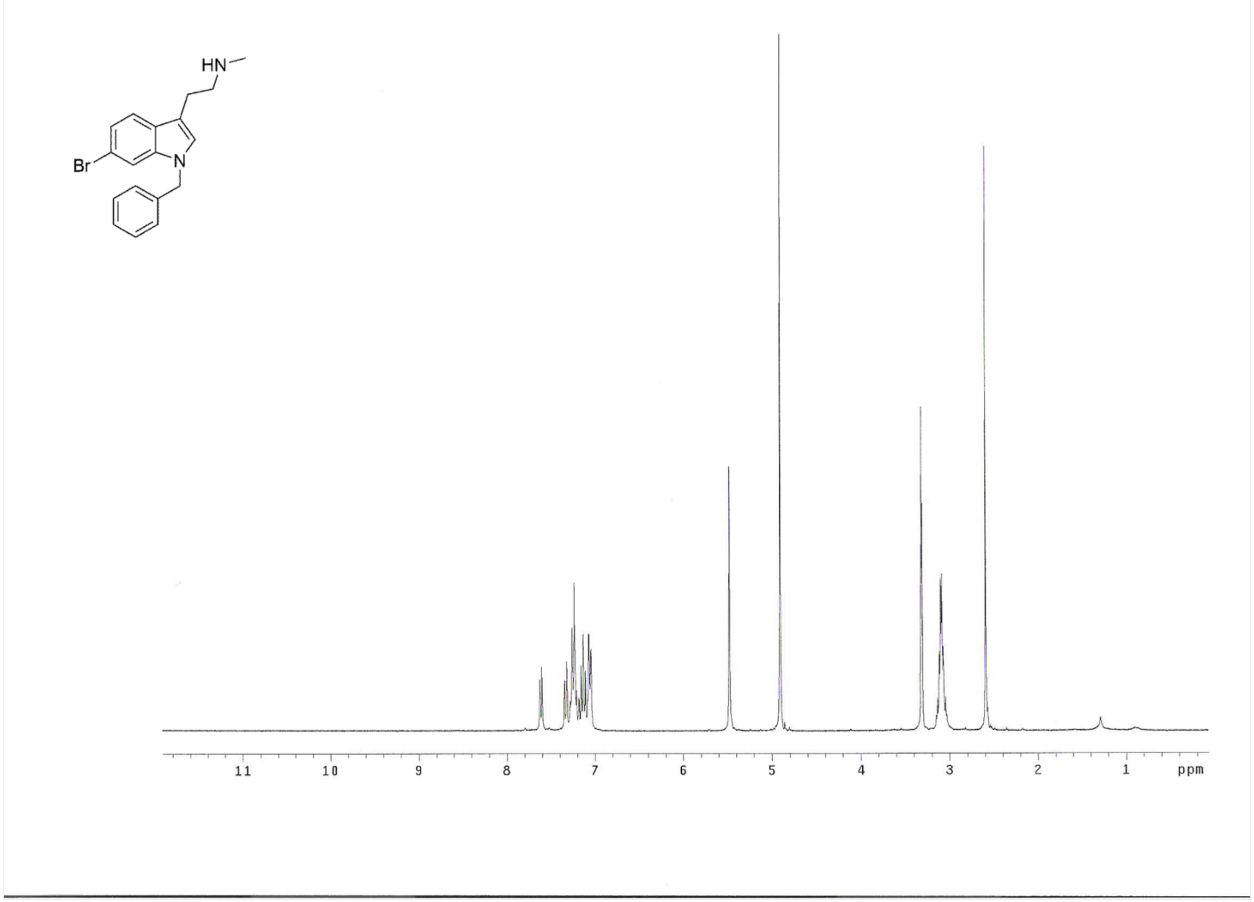
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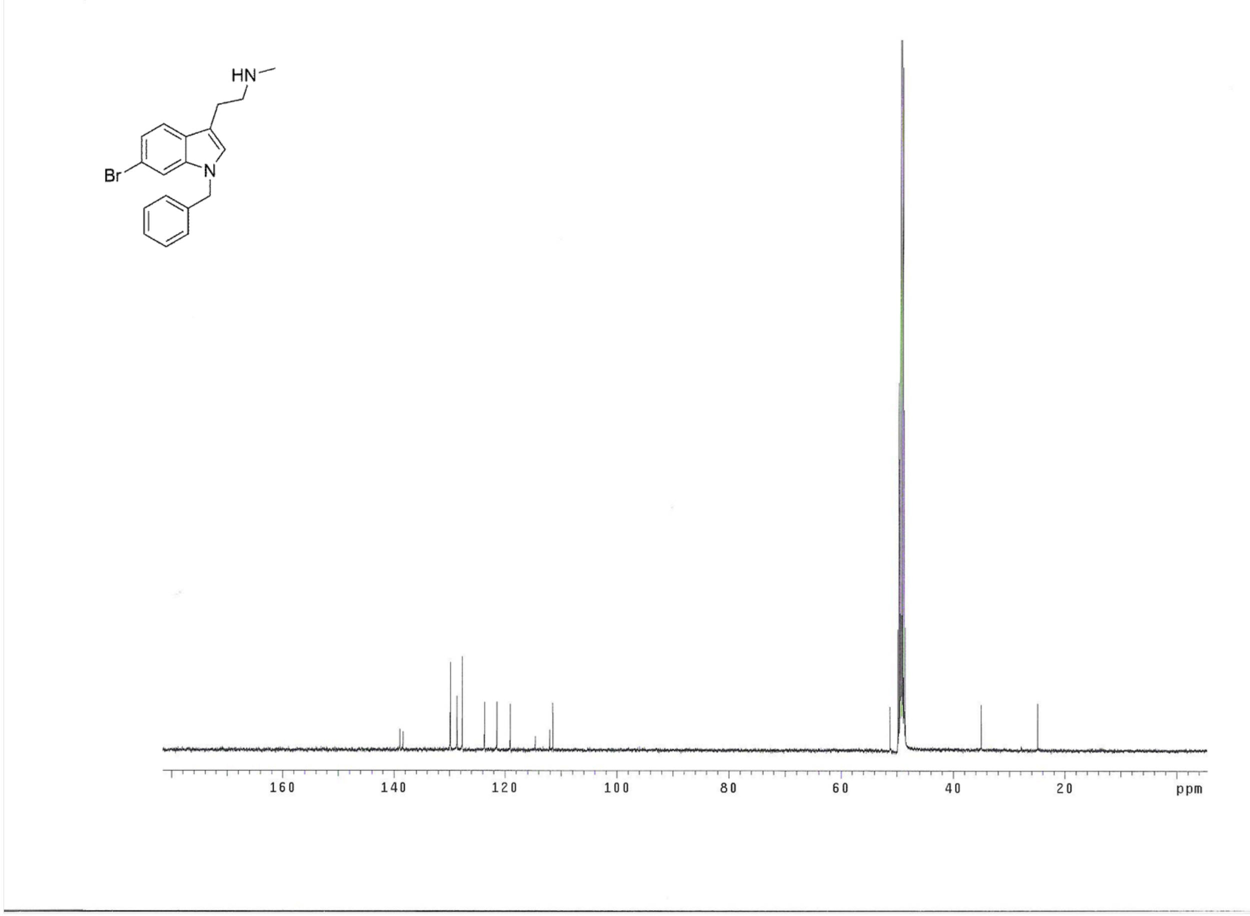
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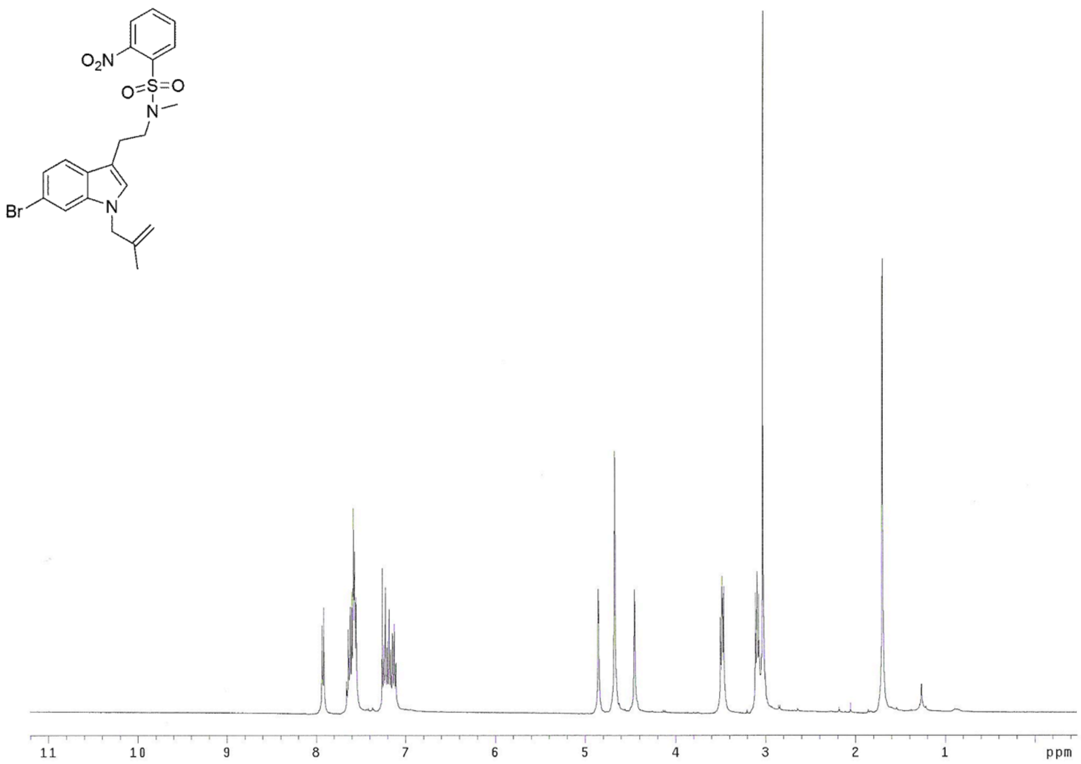
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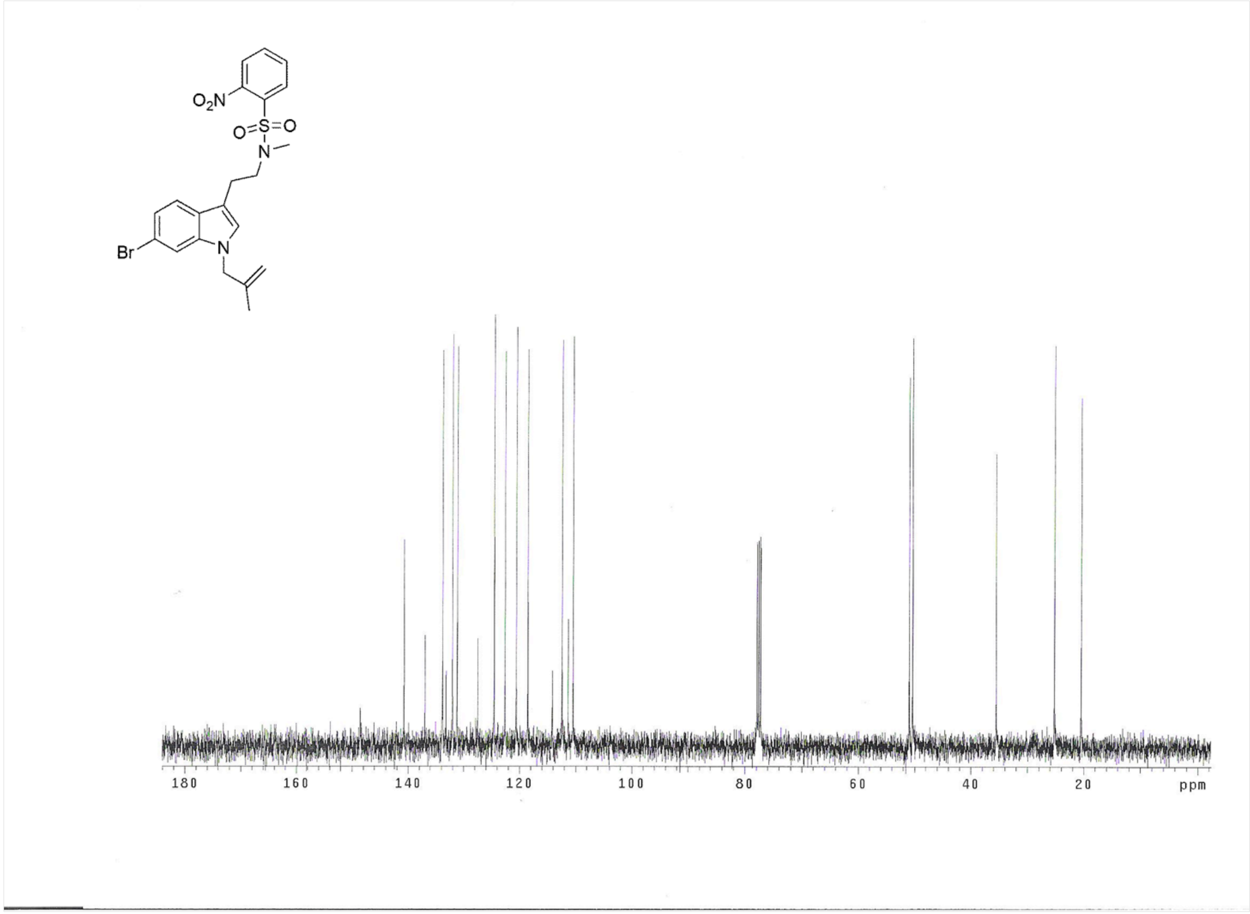
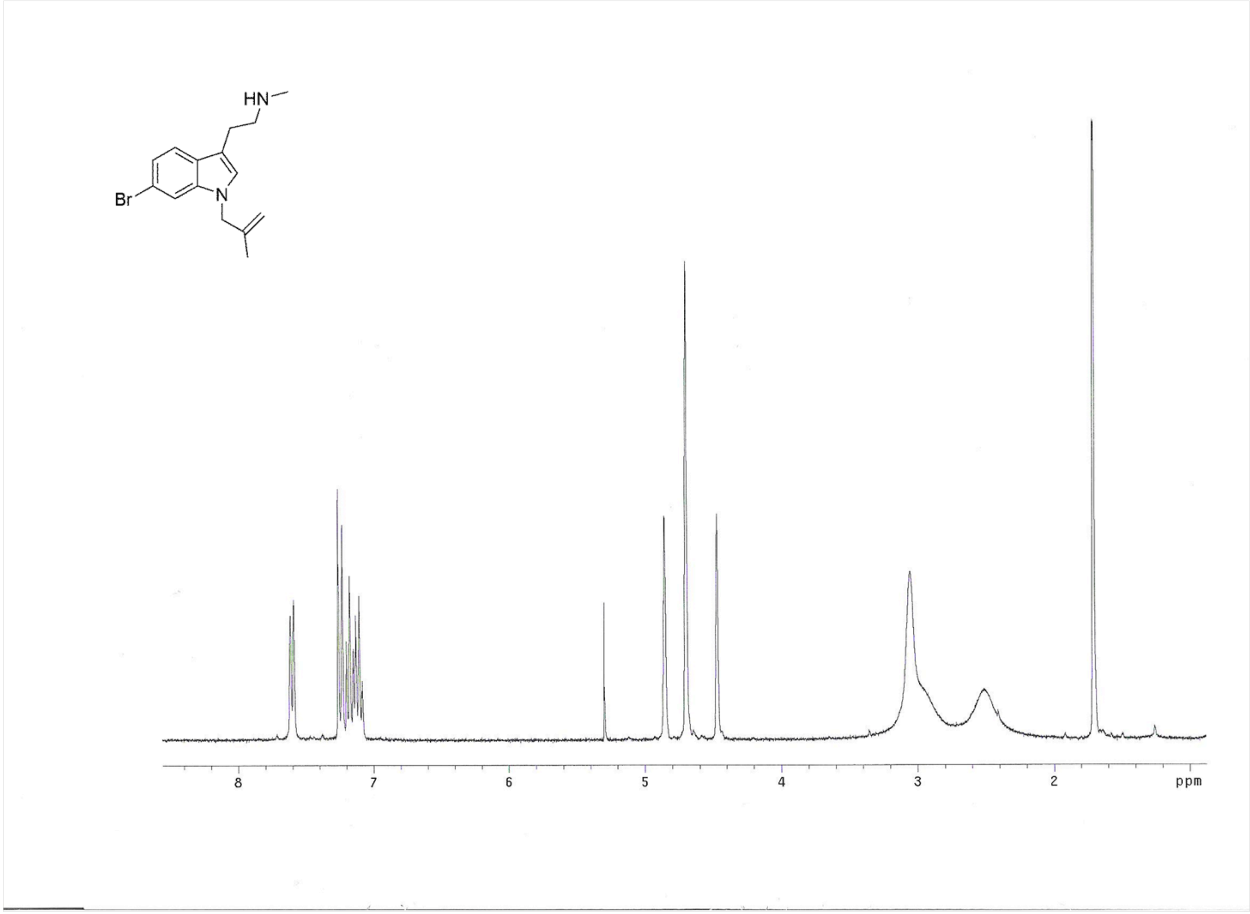
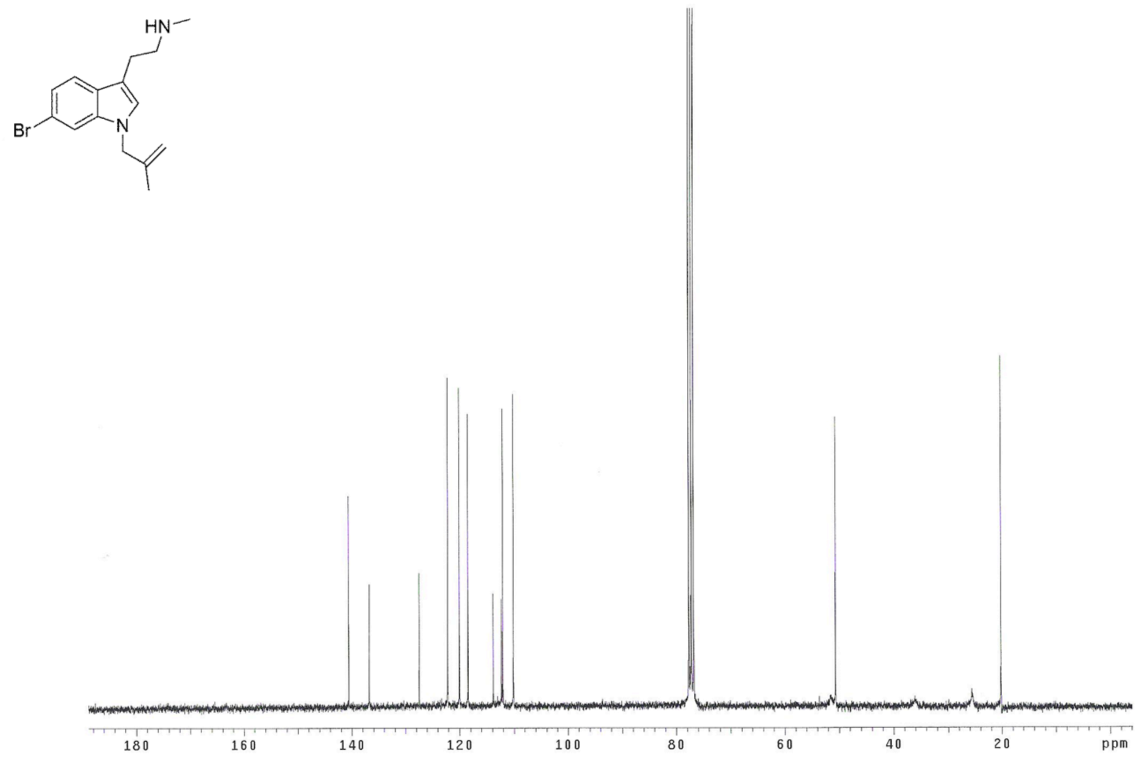
ppm


O2N
S
O
O
N
Br
N
180
160
140
120
100
80
60
40
20
ppm


HN
Br
N
8
7
6
5
4
3
2
ppm


HN
Br
N
180
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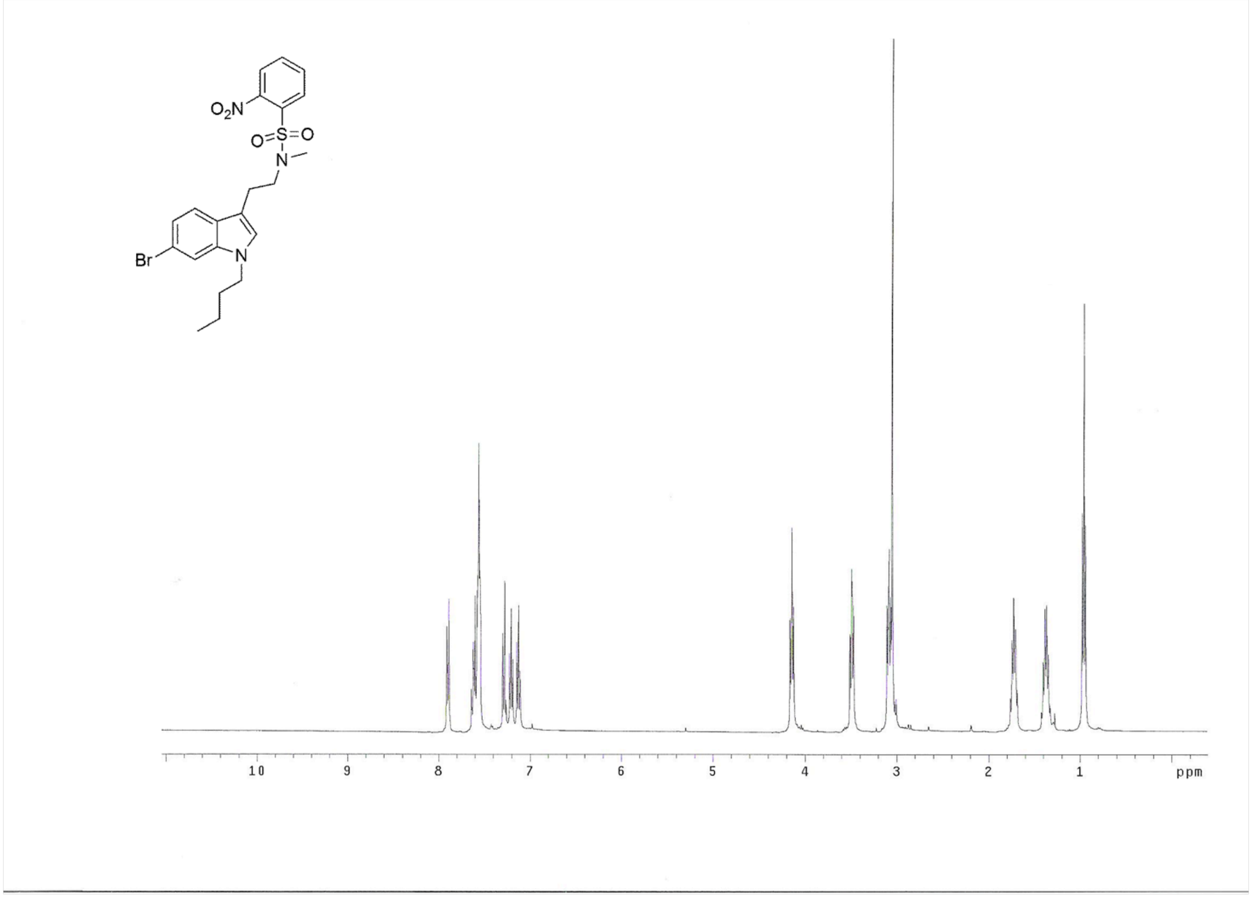
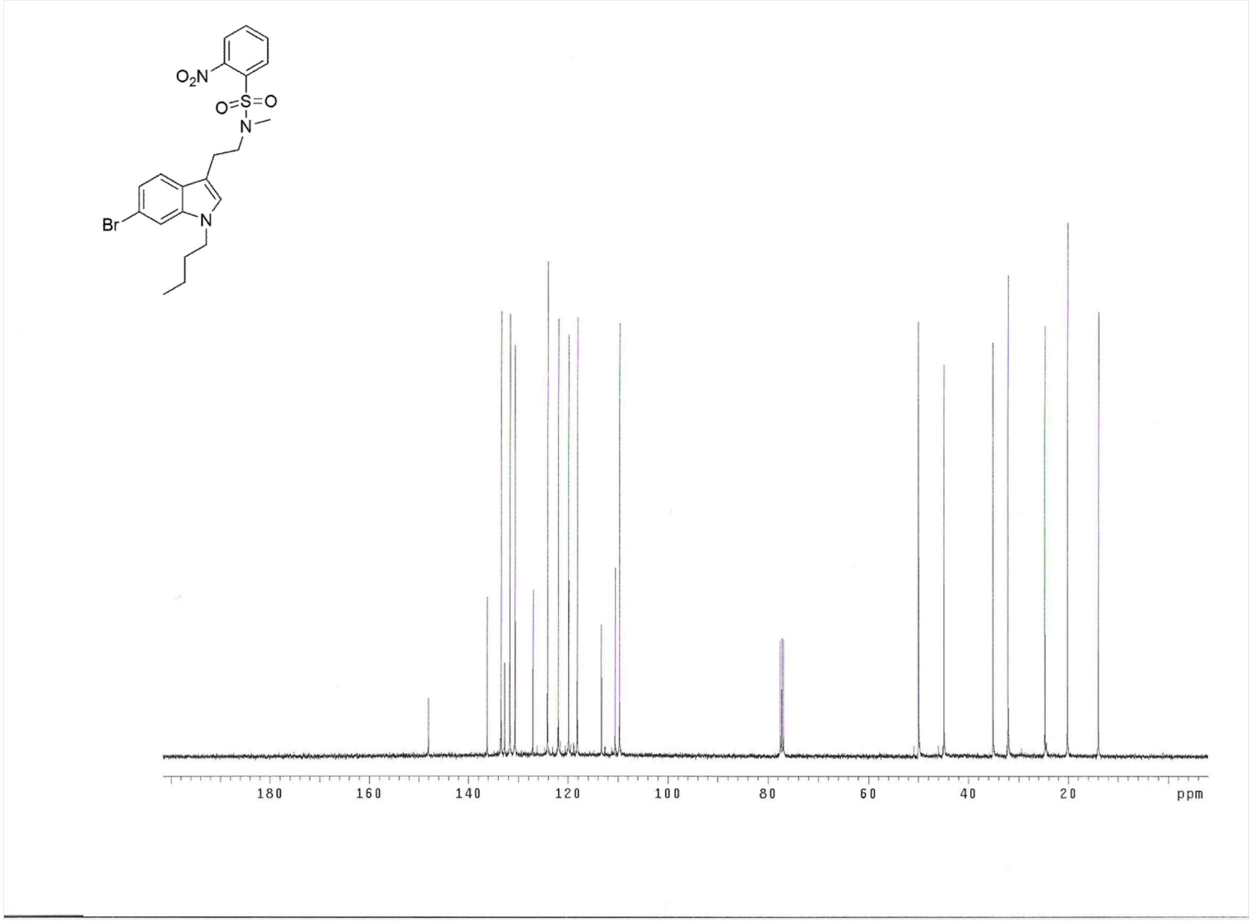
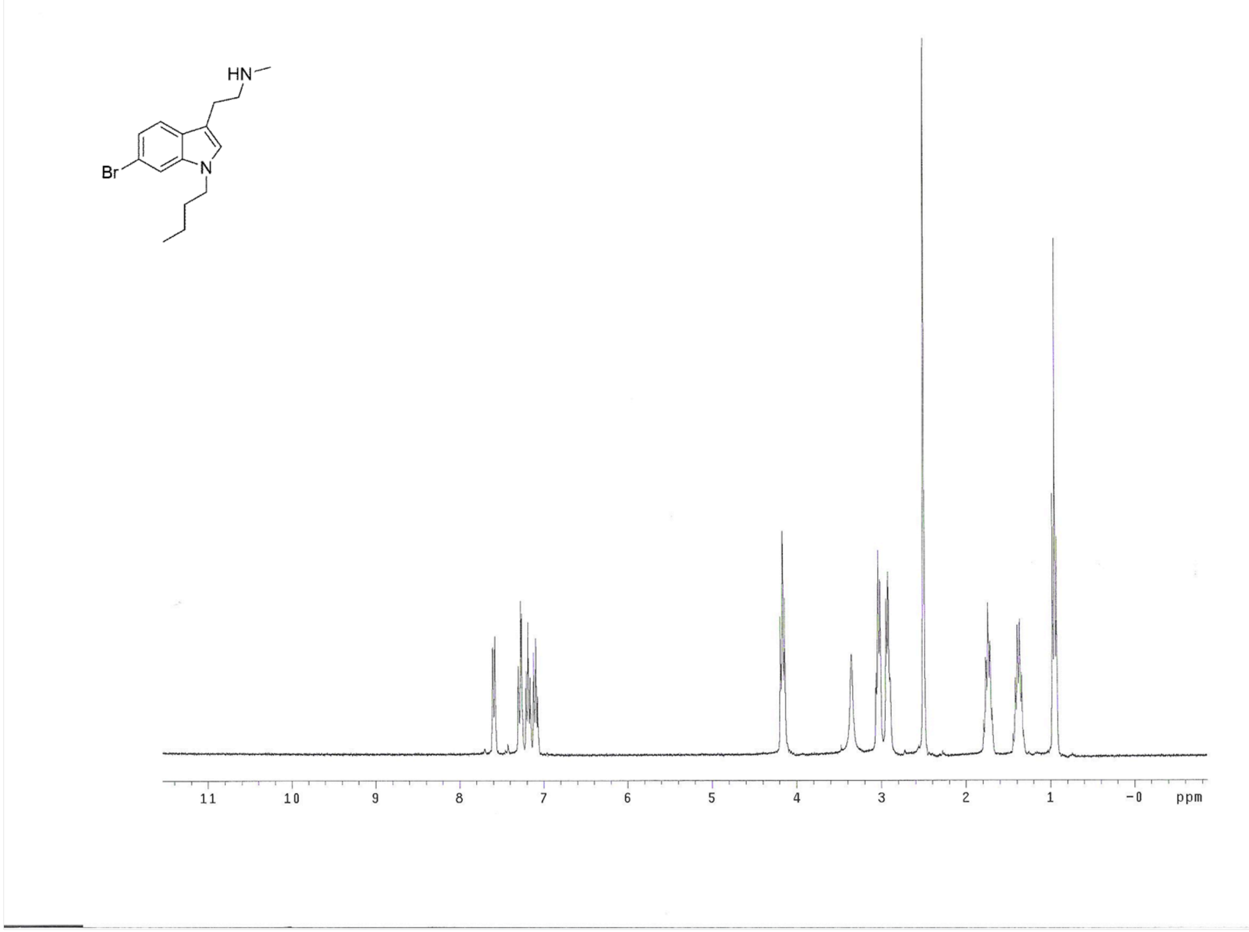
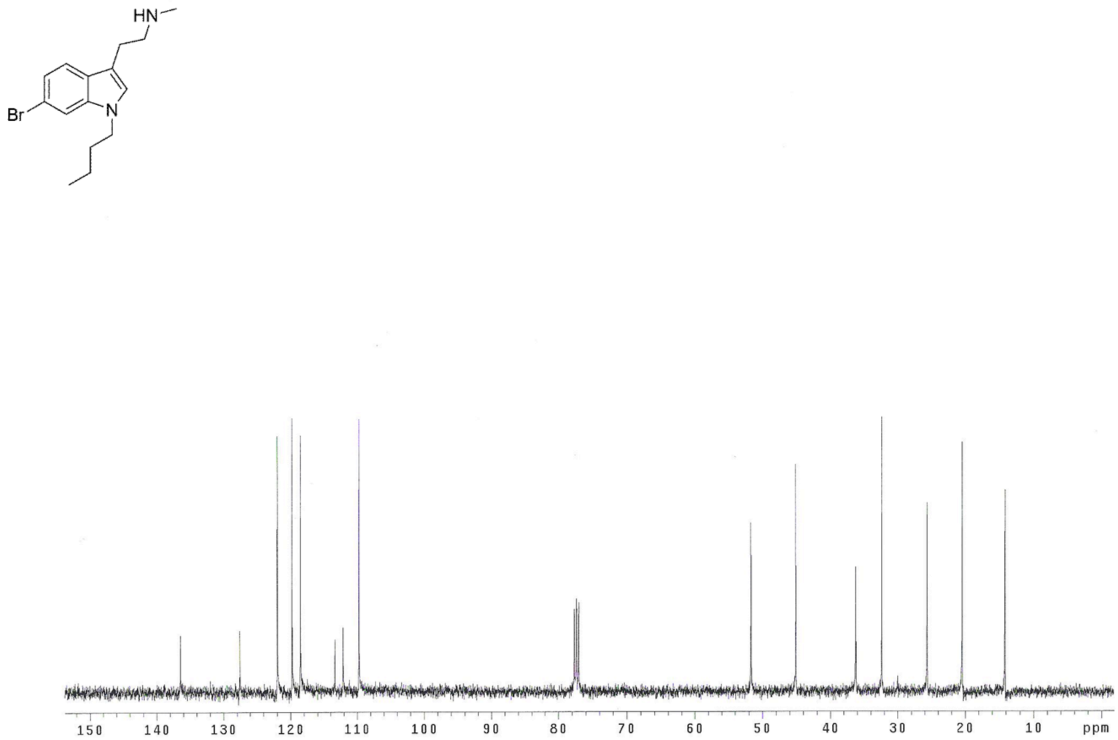
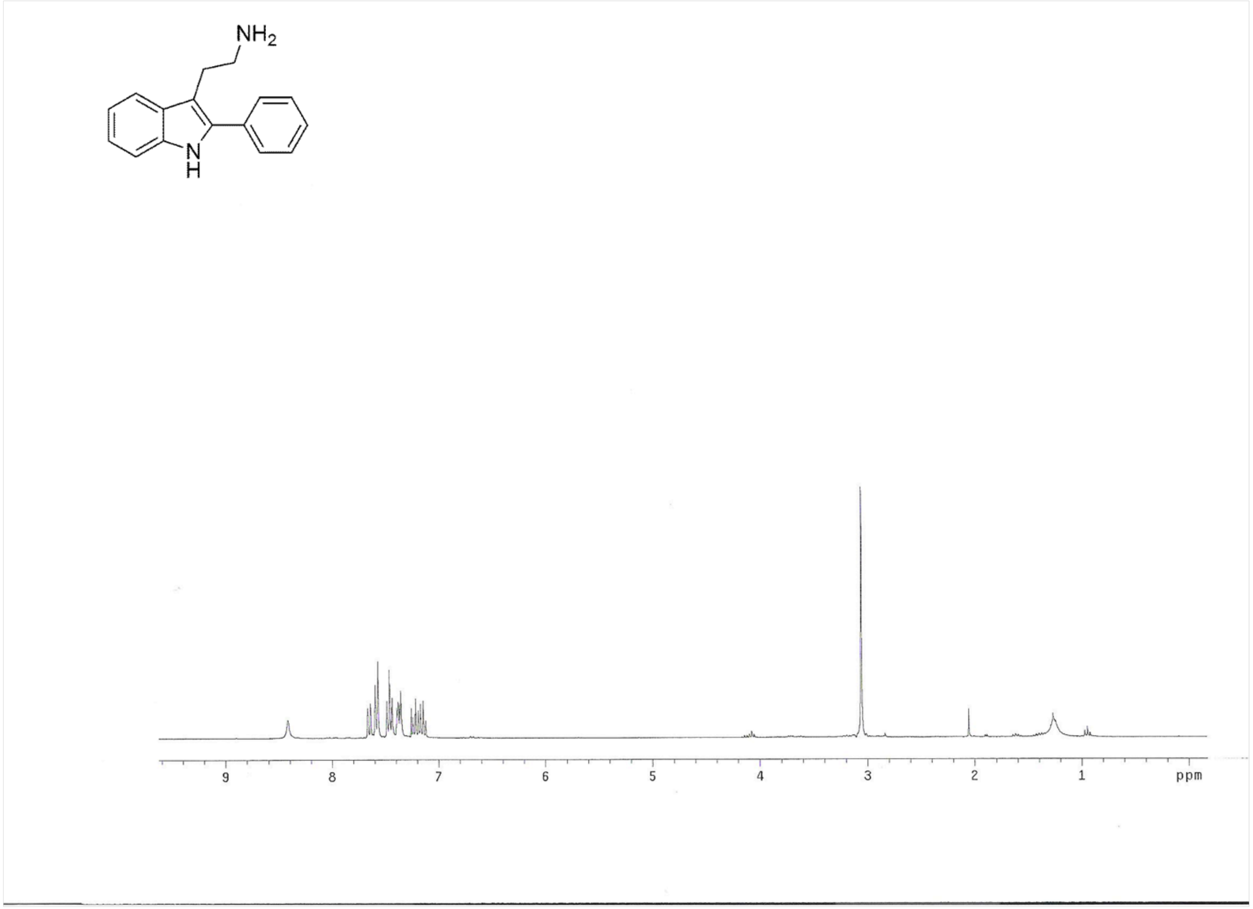
140
120
100
80
60
40
20
ppm

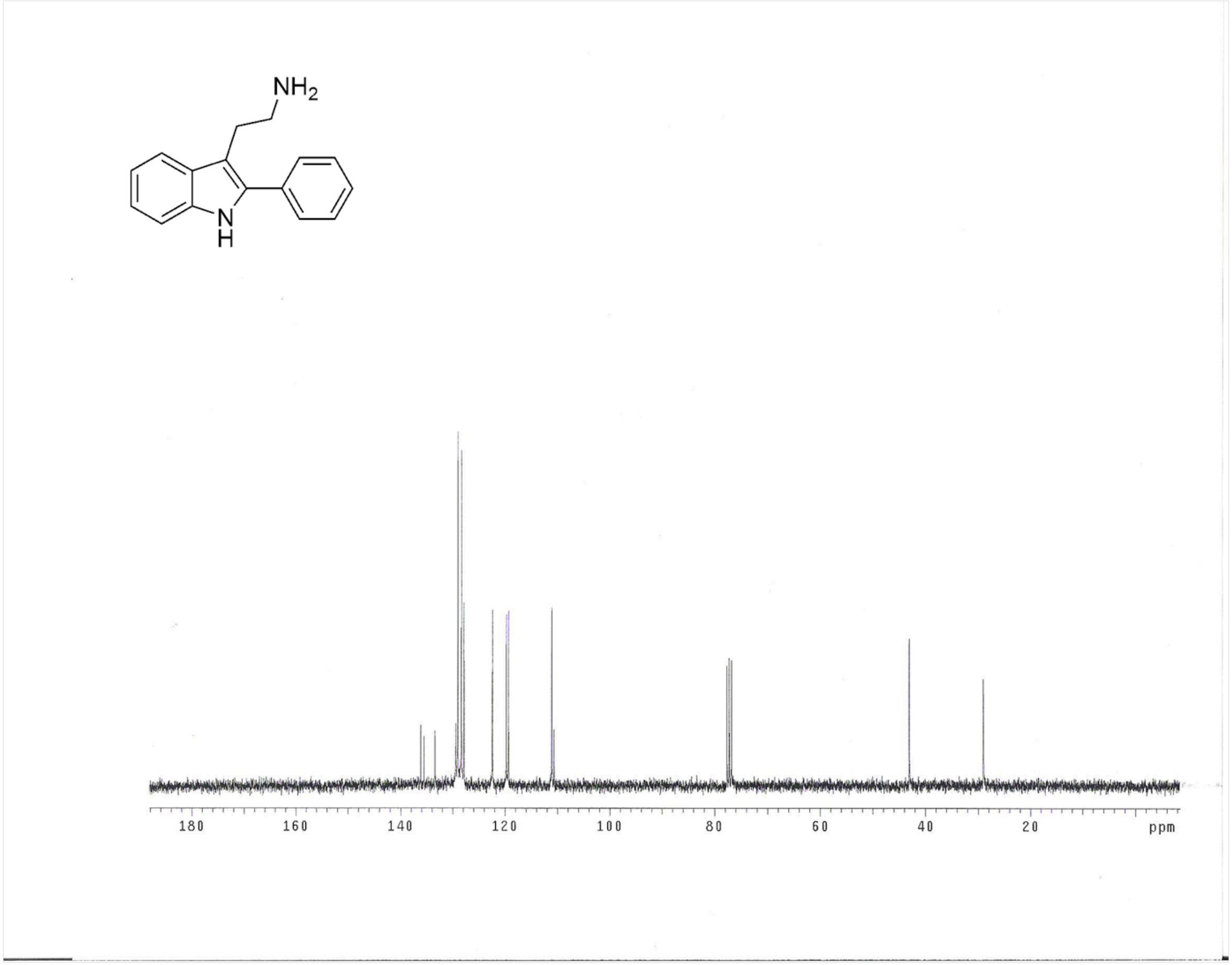

O2N
O=S=O
N
Br
N
10
9
8
7
6
5
4
3
2
1
ppm

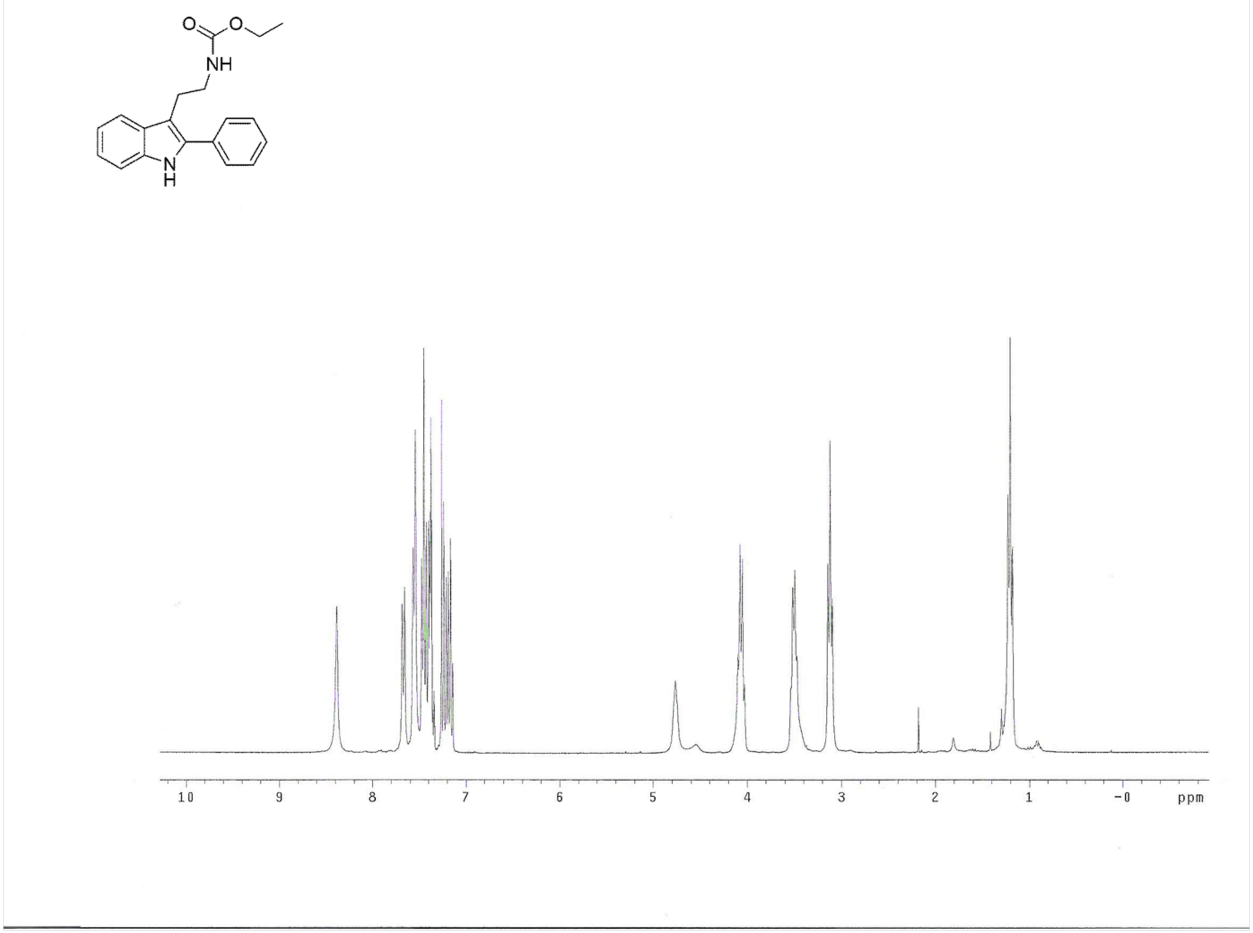

O2N
O=S=O
N
Br
N
180
160
140
120
100
80
60
40
20
ppm


HN
Br
N
11
10
9
8
7
6
5
4
3
2
1
-0
ppm


HN
Br
N
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
ppm


NH2
N
H
9
8
7
6
5
4
3
2
1
ppm


NH2
N
H
180
160
140
120
100
80
60
40
20
ppm



O
O
NH
N
H
10
9
8
7
6
5
4
3
2
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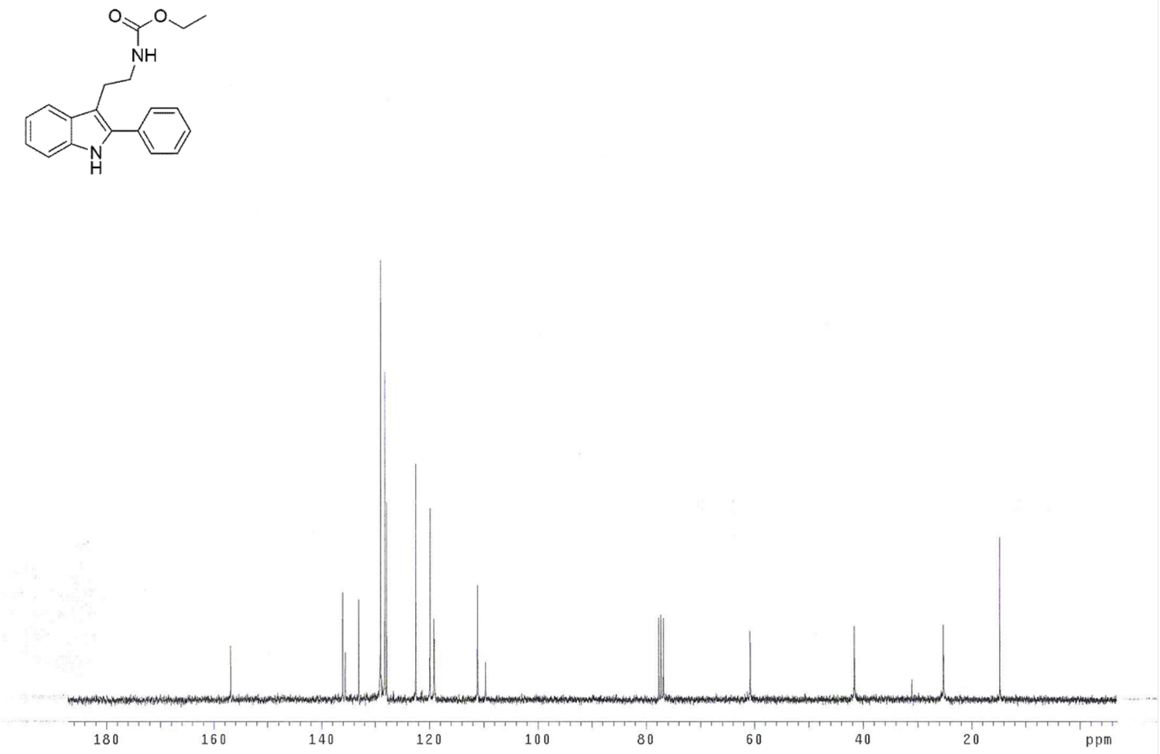
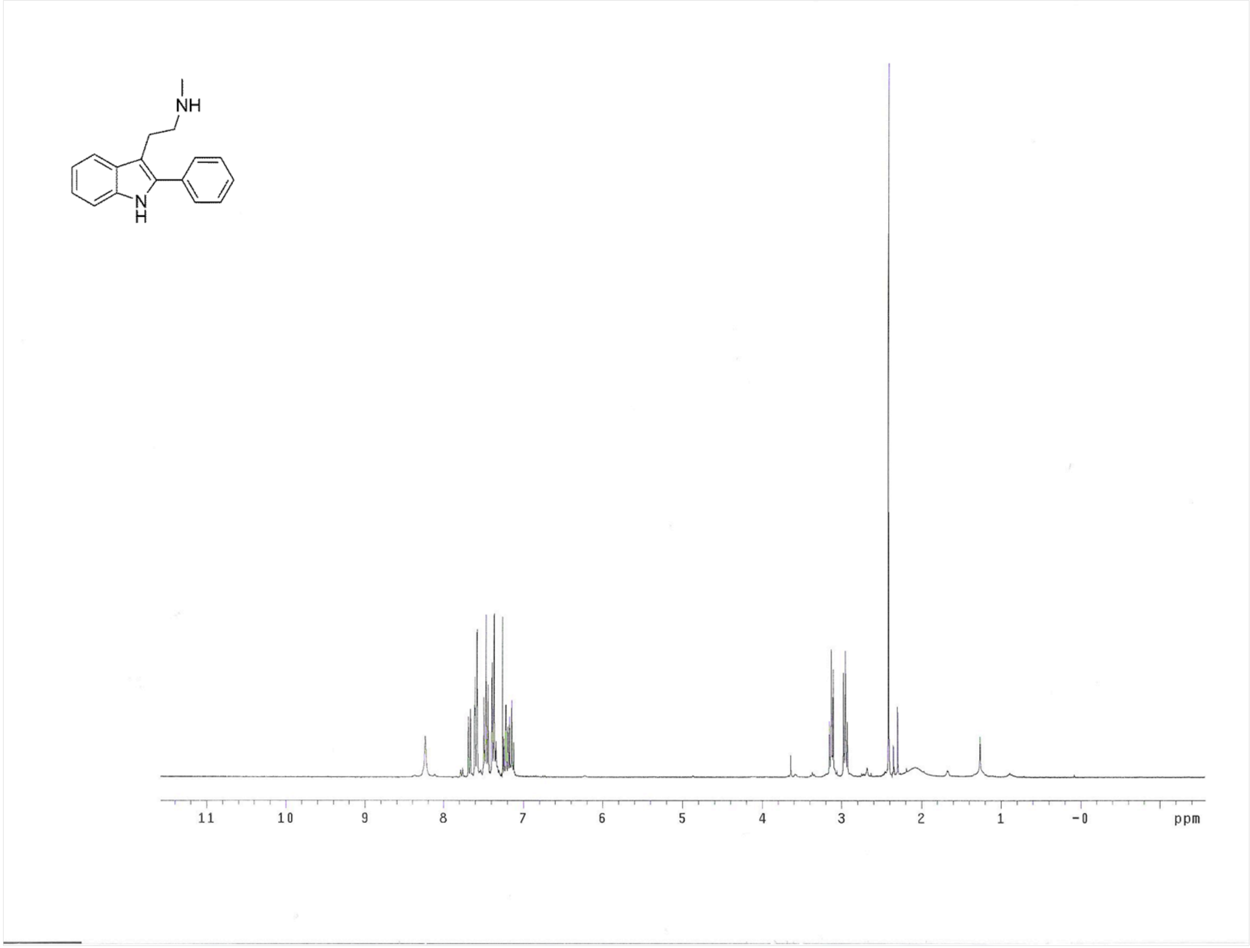
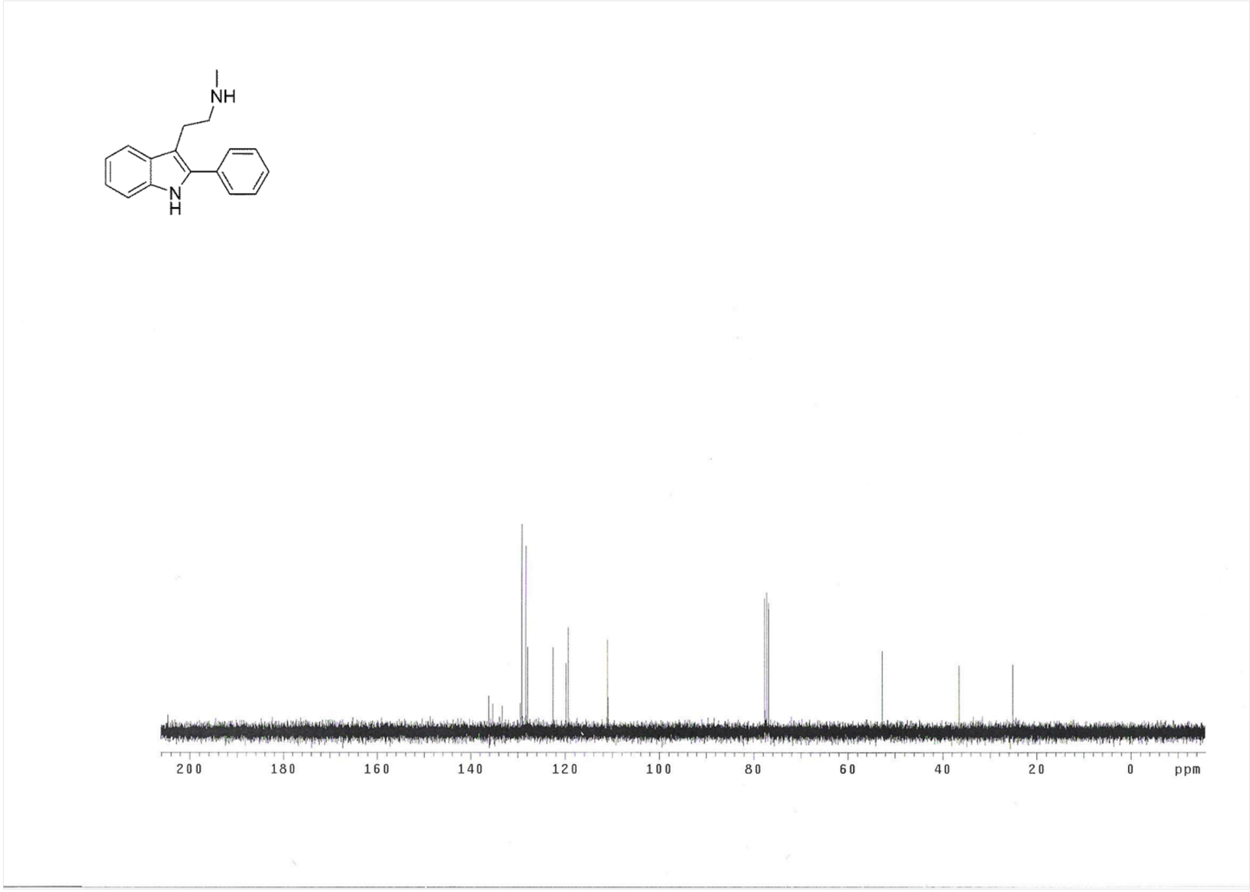
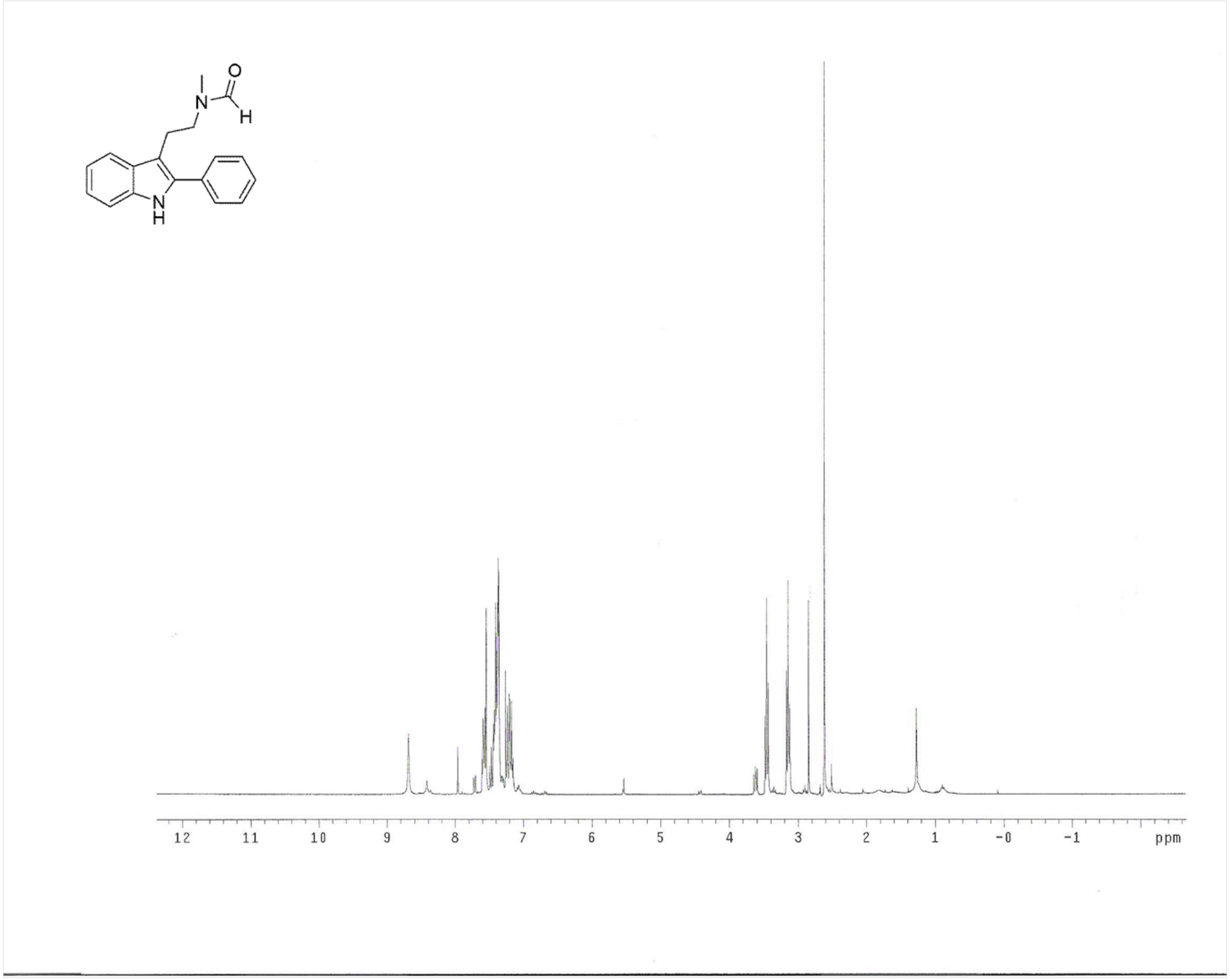
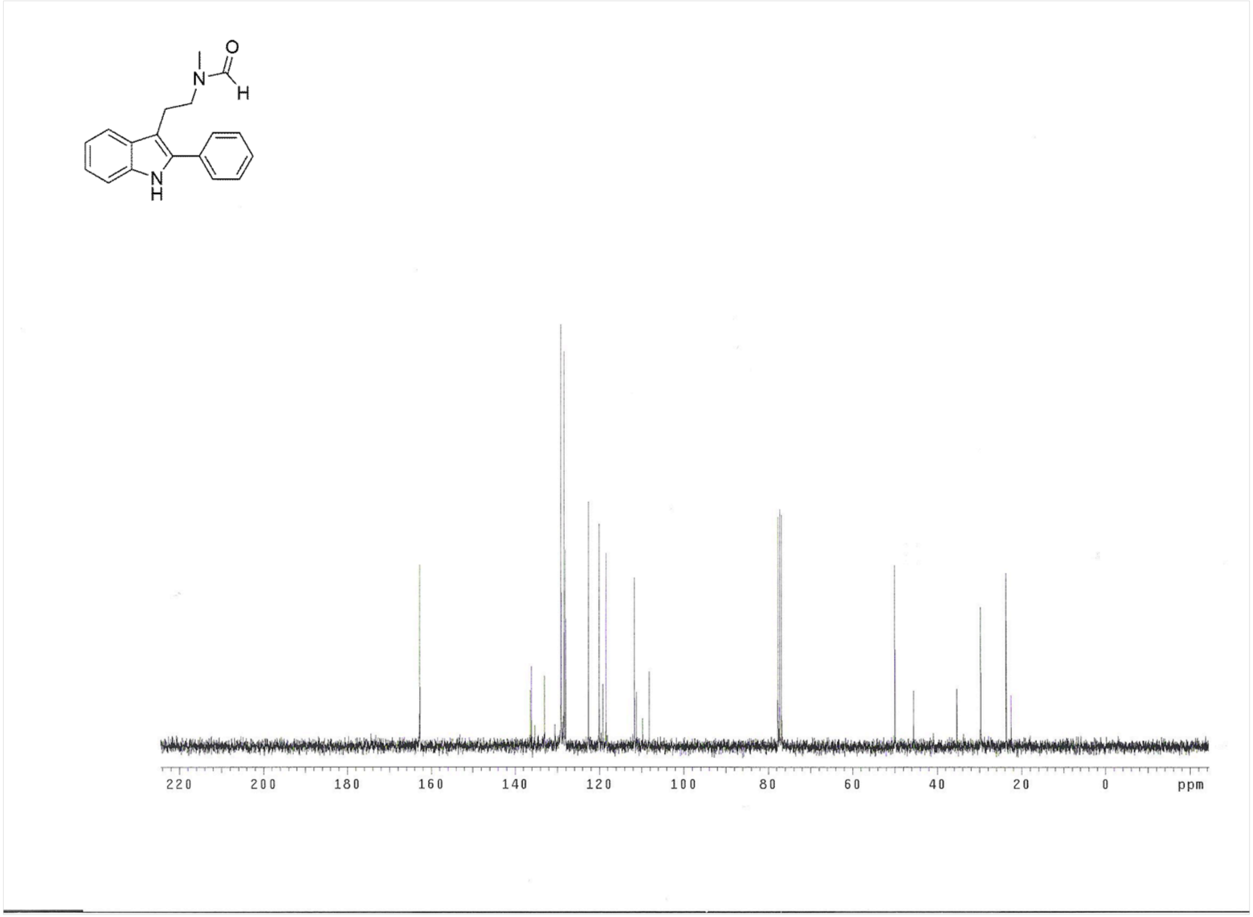
-0
ppm

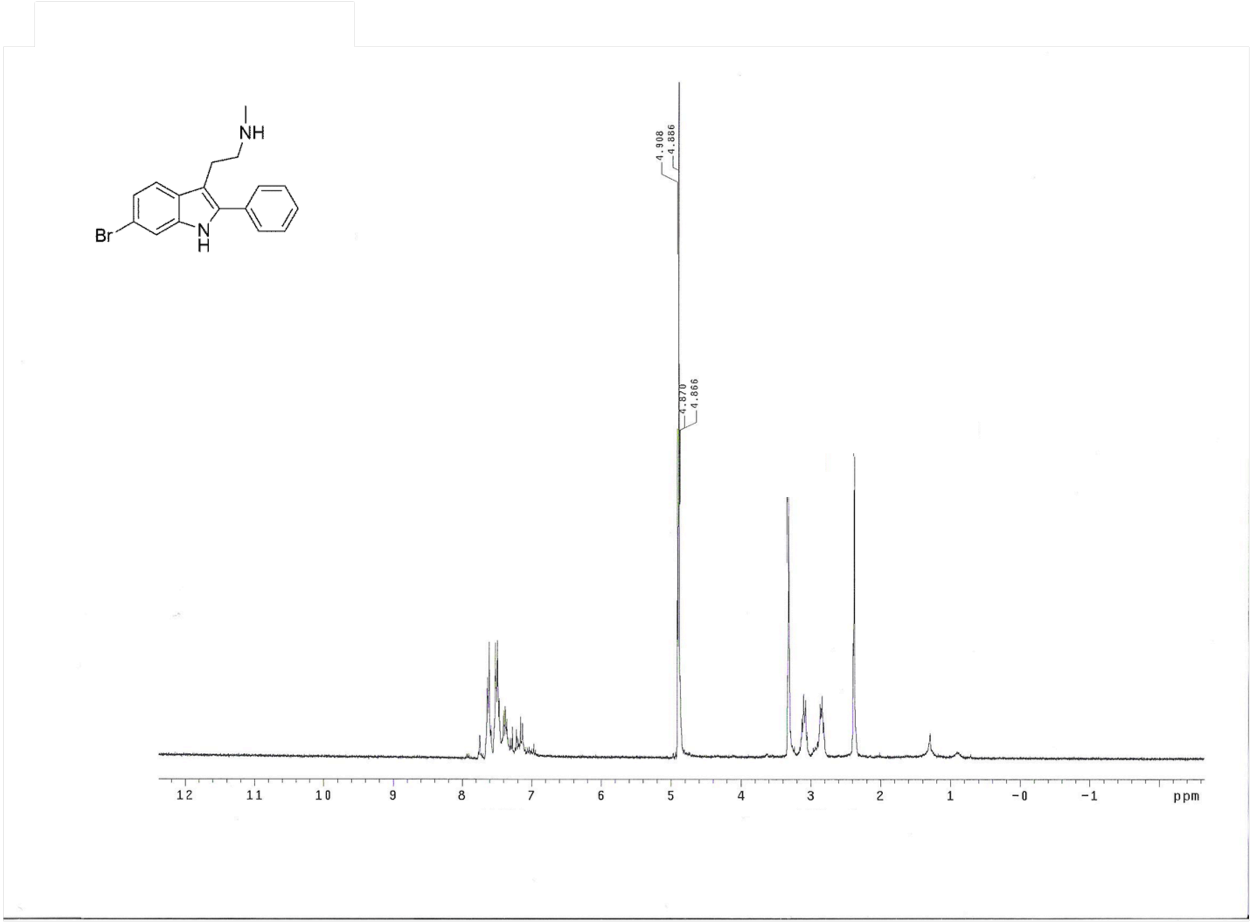

O
O
NH
N
H
180
160
140
120
100
80
60
40
20
ppm

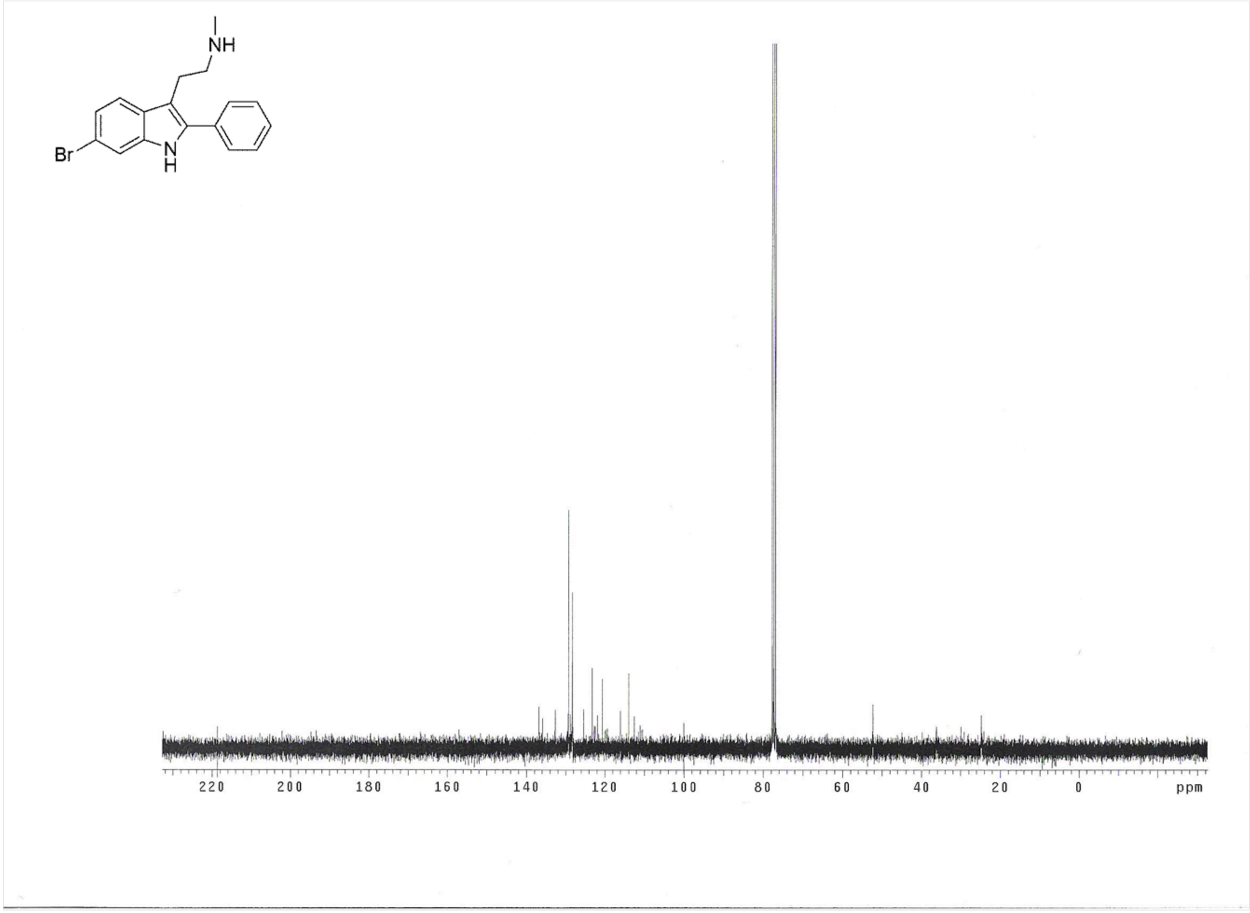

NH
N
H
11
10
9
8
7
6
5
4
3
2
1
-0
ppm

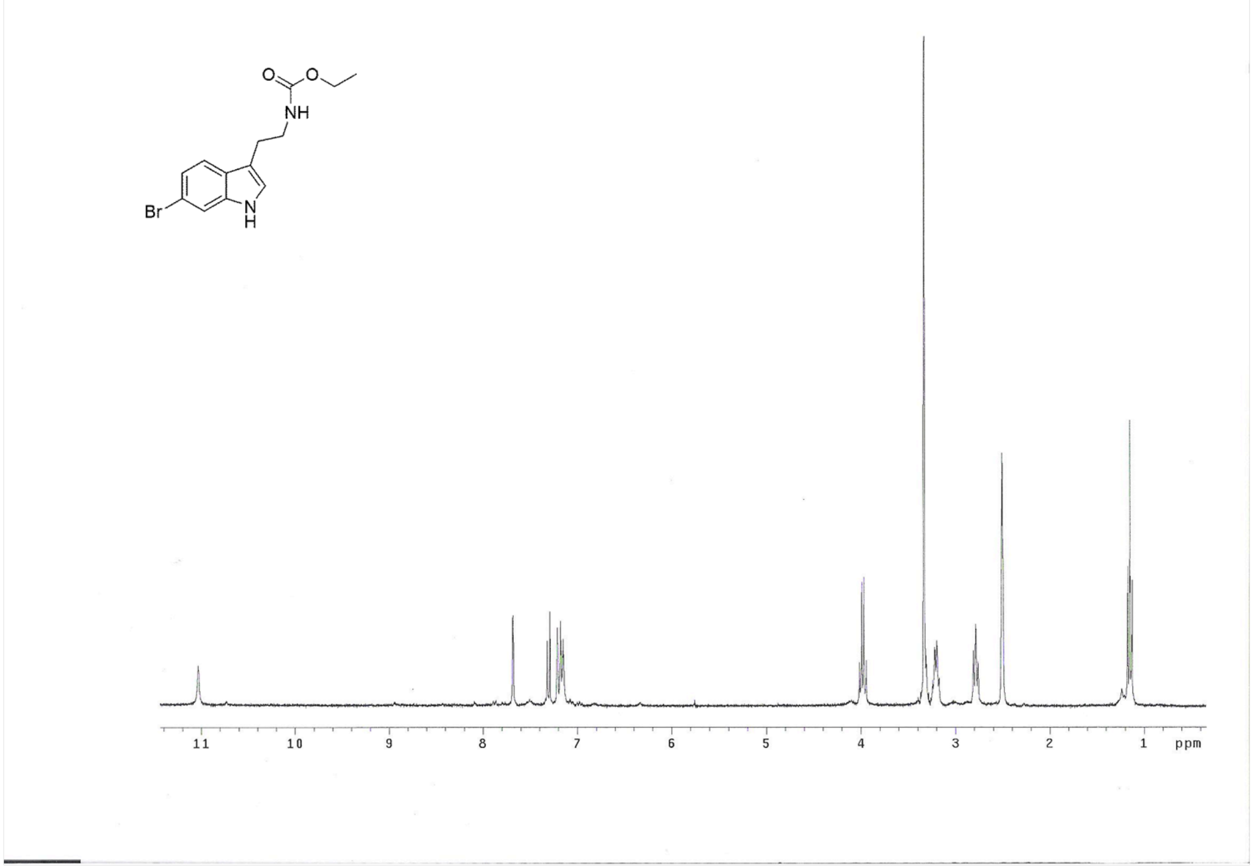

NH
N
H
200
180
160
140
120
100
80
60
40
20
0
ppm


N
O
H
N
H
12
11
10
9
8
7
6
5
4
3
2
1
-0
-1
ppm

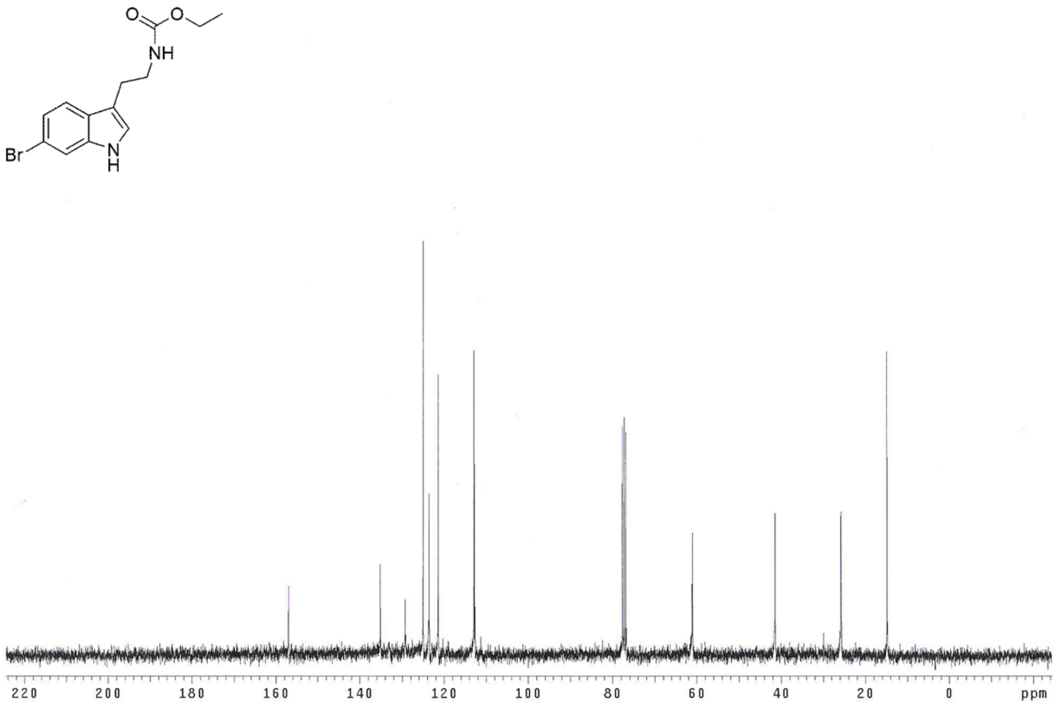

N
O
H
N
H
220
200
180
160
140
120
100
80
60
40
20
0
ppm

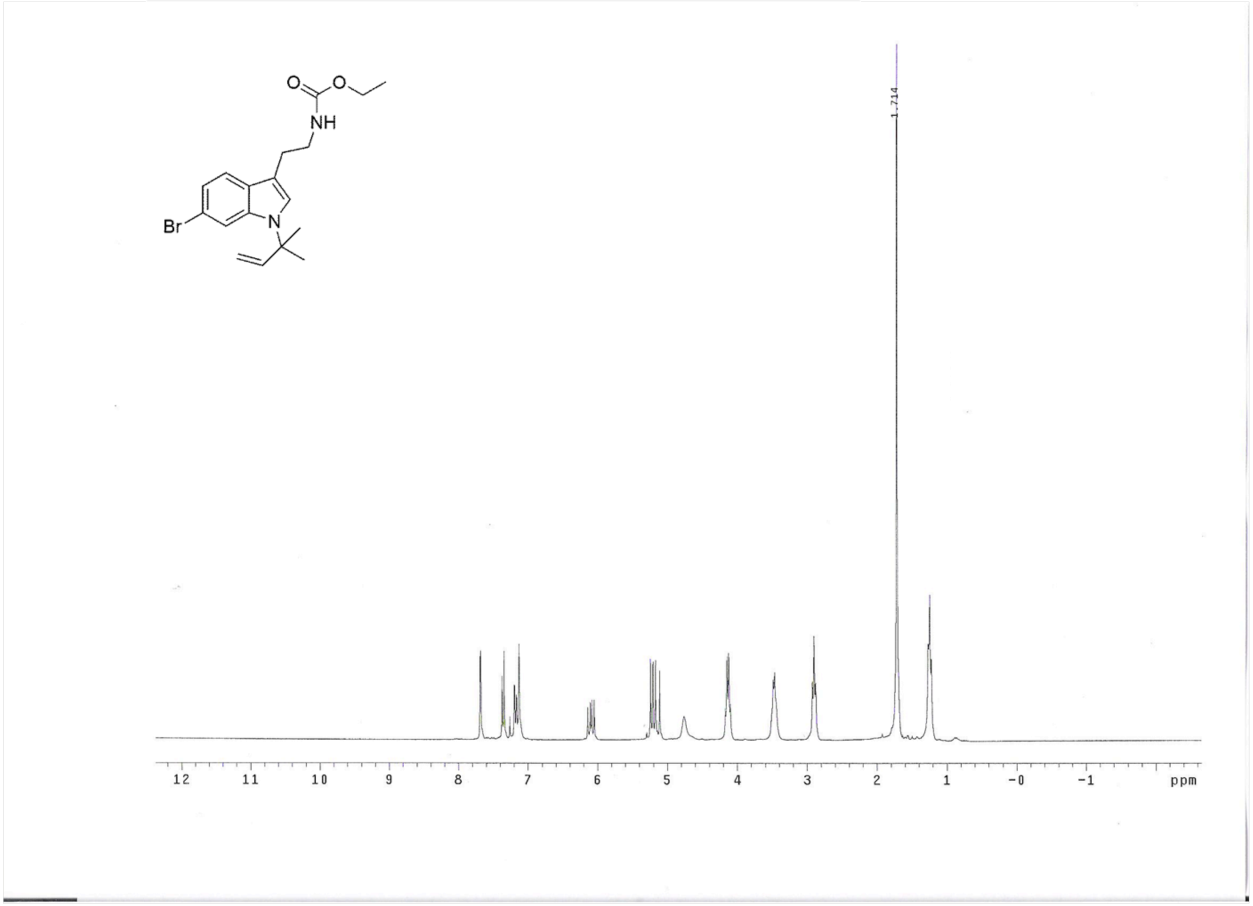

NH
Br
H
4.908
4.886
4.870
4.866
12
11
10
9
8
7
6
5
4
3
2
1
-0
-1
ppm

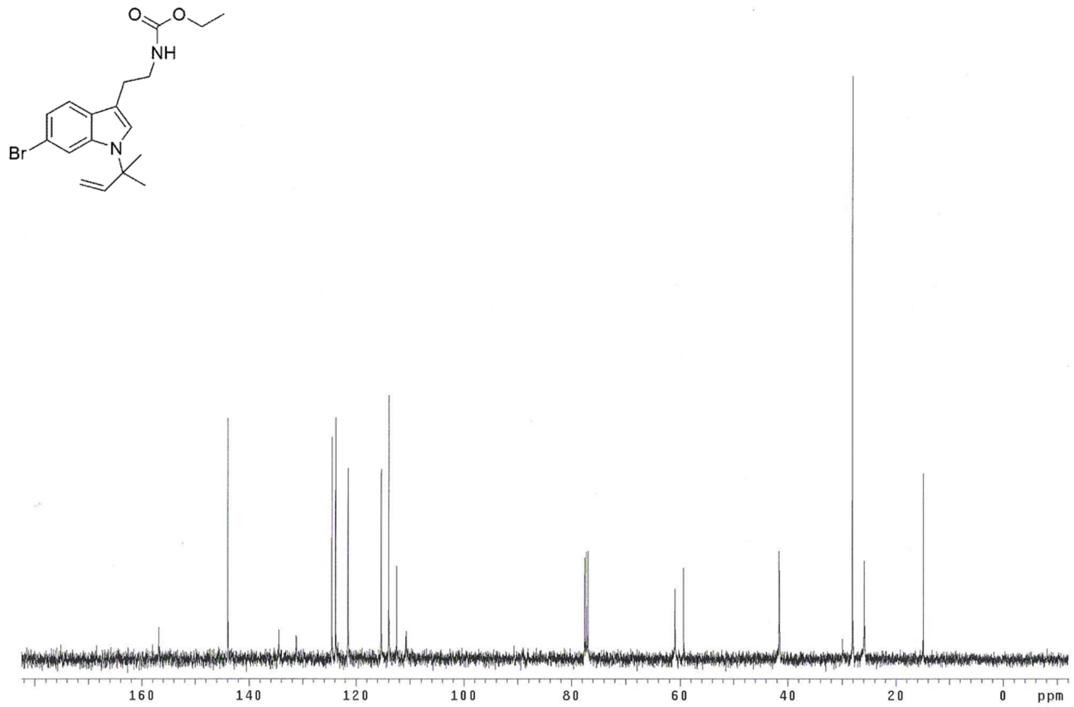

NH
Br
N
H
220
200
180
160
140
120
100
80
60
40
20
0
ppm



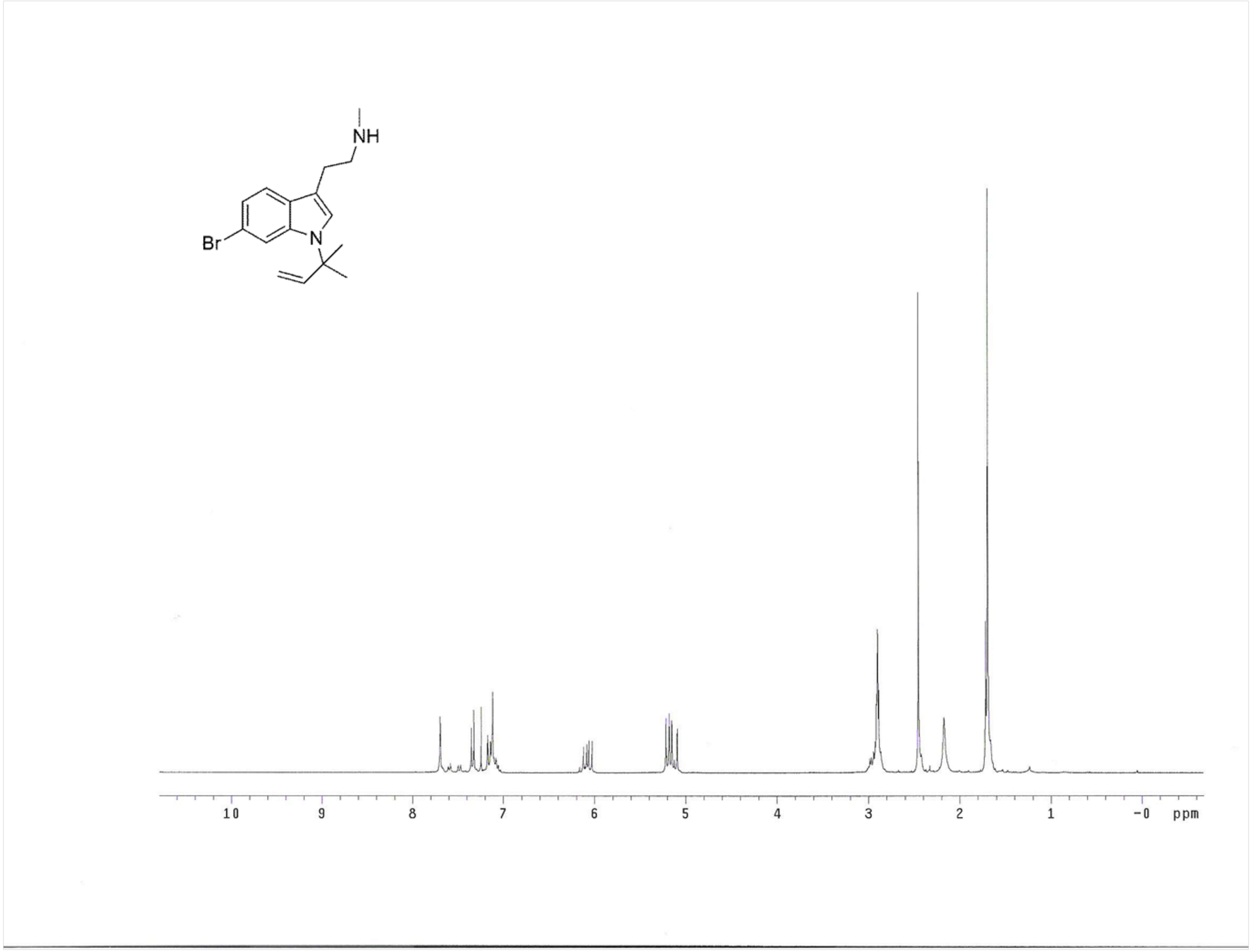
O
O
NH
Br
N
H
11
10
9
8
7
6
5
4
3
2
1
ppm

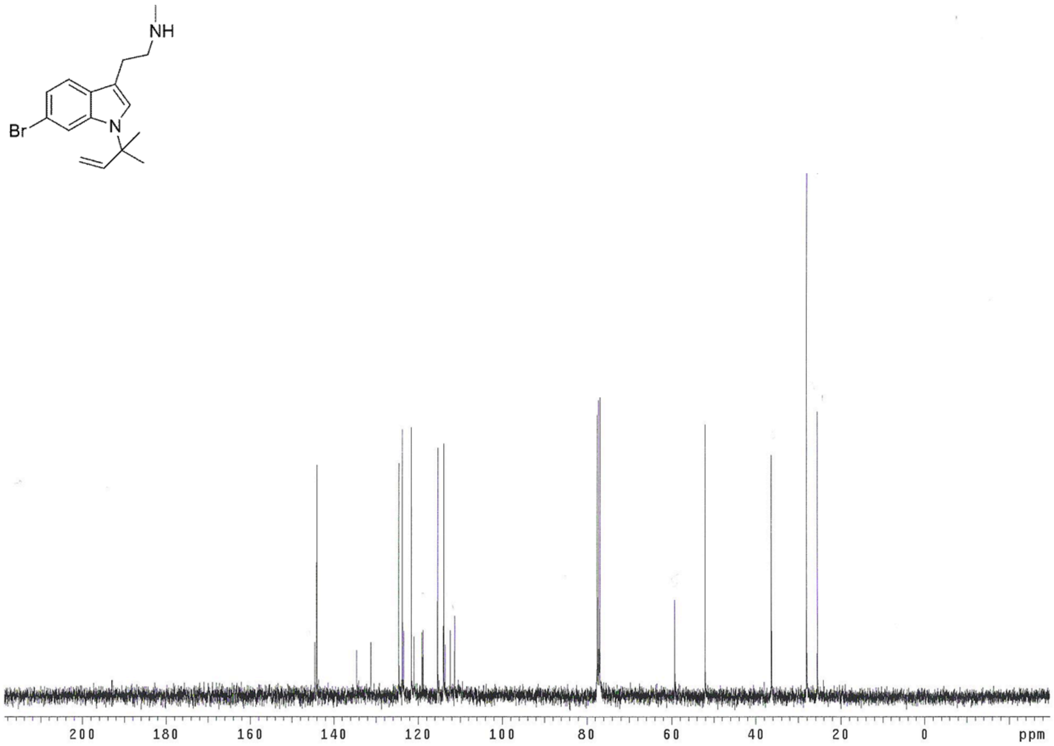

O
O
NH
Br
N
H
220
200
180
160
140
120
100
80
60
40
20
0
ppm


O
O
NH
Br
N
1.714
12
11
10
9
8
7
6
5
4
3
2
1
-0
-1
ppm



O
O
NH
Br
N
160
140
120
100
80
60
40
20
0
ppm


NH
Br
N
10
9
8
7
6
5
4
3
2
1
-0
ppm


NH
Br
N
200
180
160
140
120
100
80
60
40
20
0
ppm