Supplementary material

Synthesis and Pharmacological Evaluation of Donepezil and Chromone Hybirds as New Multipotent Cholinesterase/Monoamine Oxidase Inhibitors for the Potential Treatment of Alzheimer's Disease

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Contents

1. Experimental section

1.1. Chemistry

All chemicals (reagent grade) used were purchased from Sino pharm Chemical Reagent Co., Ltd. (China). Reaction progress was monitored using analytical thin layer chromatography (TLC) on precoated silica gel GF254 (Qingdao Haiyang Chemical Plant, Qing-Dao, China) plates and the spots were detected under UV light (254 nm). Melting point was measured on an XT-4 micromelting point instrument and uncorrected. IR (KBr-disc) spectra were recorded by Bruker Tensor 27 spectrometer. ^{1}H NMR and ^{13}C NMR spectra were measured on a Bruker ACF-500 spectrometer at 25°C and referenced to TMS. Chemical shifts are reported in ppm (δ) using the residual solvent line as internal standard. Splitting patterns are designed as s, singlet; d, doublet; t, triplet; m, multiplet. The purity of all compounds was confirmed to be higher than 95% through analytical HPLC performed with Agilent 1200 HPLC System. Mass spectra were obtained on a MS Agilent 1100 Series LC/MSD Trap mass spectrometer (ESI-MS) and a Mariner ESI-TOF spectrometer (HRESIMS), respectively. Column chromatography was performed on silica gel (90-150 μm; Qingdao Marine Chemical Inc.).

1.1.1. General procedures for the preparation of intermediate 3-Formylchromones *2a-g*

2-Acetylphenol analogues **1a-h** (10 mmol) were dissolved in 10 mL DMF and cooled to 0°C. $POCl_3$ (20 mmol) was added dropwise to the reaction and the resulting mixture was incubated at room temperature for 18 h. The reaction was poured into

ice-water, the resulting precipitate was collected by filtration and dried at 60 °C overnight.

1.1.2. General procedures for the preparation of intermediate 3a-3g

3-Formylchromones **2a**-**2h** (2 mmol) and NH_2SO_3H (10 mmol) were dissolved in a mixture of 12 mL water and 10 mL CH_2Cl_2. The reaction was cooled to 0 °C and a solution of $NaClO_2$ (8 mmol) in 6 mL water was added dropwise. The reaction was stirred for a further 30 min at 0 °C and the organic phase was separated. The aqueous phase was extracted to CH_2Cl_2 (2 × 15 mL) and the combined organic phases were washed with water (30 mL) and brine (30 mL). The organic phase was dried over Na_2SO_4, removed in vacuo and the residue was recrystallized from ethyl acetate.

1.1.2.1 4-oxo-4H-chromene-3-carboxylic acid (3a)

The title compound (**3a**) was prepared from **2a** following the general procedure as light white solid in 75% yield.

1.1.2.2 6-methoxy-4-oxo-4H-chromene-3-carboxylic acid (3b)

The title compound (**3b**) was prepared from **2b** following the general procedure as light white solid in 68% yield. ^{1}H NMR (500 MHz, DMSO-d_6) δ 13.35 (s, 1H), 9.12 (s, 1H), 7.78 (d, J = 8.5 Hz, 1H), 7.54 (t, J = 3.9 Hz, 1H), 7.52 (s, 1H), 3.91 (s, 3H).

1.1.2.3 6-(benzyloxy)-4-oxo-4H-chromene-3-carboxylic acid (3c)

The title compound (**3c**) was prepared from **2c** following the general procedure as light white solid in 65% yield. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.15 (s, 1H), 8.91 (s, 1H), 7.75 (d, J = 9.1 Hz, 1H), 7.62 (d, J = 3.0 Hz, 1H), 7.56 (dd, J = 9.1, 3.1 Hz, 1H), 7.51 (d, J = 7.4 Hz, 2H), 7.43 (t, J = 7.5 Hz, 2H), 7.37 (t, J = 7.3 Hz, 1H), 5.27 (s,

2H).

1.1.2.4 6-methyl-4-oxo-4H-chromene-3-carboxylic acid (3d)

The title compound (**3d**) was prepared from **2d** following the general procedure as light white solid in 75% yield. ^{1}H NMR (500 MHz, DMSO-d_6) δ 9.12 (s, 1H), 7.98 (s, 1H), 7.77 (dd, J = 8.6, 1.7 Hz, 1H), 7.73 (s, 1H), 7.71 (s, 1H), 2.49 (s, 3H).

1.1.2.5 6-bromo-4-oxo-4H-chromene-3-carboxylic acid (3e)

The title compound (**3f**) was prepared from **2f** following the general procedure as light white solid in 56% yield.

1.1.2.6 7-methoxy-4-oxo-4H-chromene-3-carboxylic acid (3f)

The title compound (**3g**) was prepared from **2g** following the general procedure as light white solid in 25% yield. ^{1}H NMR (500 MHz, DMSO-d_6) δ 13.47 (s, 1H), 9.11 (s, 1H), 8.10 (d, J = 8.9 Hz, 1H), 7.34 (d, J = 2.0 Hz, 1H), 7.22 (dd, J = 8.9, 2.1 Hz, 1H), 3.96 (s, 4H).

1.1.2.7 7-bromo-4-oxo-4H-chromene-3-carboxylic acid (3g)

The title compound (**3h**) was prepared from **2h** following the general procedure as light white solid in 20% yield. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.09 (s, 1H), 8.91 (s, 1H), 8.12 (d, J = 1.4 Hz, 1H), 8.04 (d, J = 8.5 Hz, 1H), 7.76 (dd, J = 8.5, 1.7 Hz, 1H).

1.1.3. General procedures for the preparation of compounds 4a-4h and 5a-5h

To 3 mL anhydrous dichloromethane were added oxalyl chloride (1 mmol) and DMF (cat). The solution was stirred for 0.5 h at room temperature and was added **3a-3h** (0.5 mmol). The mixture was heated to 40 °C for 1 h. All solvents were removed

under reduced pressure to get acyl chloride **3**, which was dissolved in dichloromethane (4 mL). This solution was added to 1-benzylpiperidin-4-amine or 2-(1-benzylpiperidin-4-yl) ethanamine (0.5 mmol) and K_2CO_3 (1 mmol) in dichloromethane (2 mL) by dripping at 0 °C. When the reaction was completed, it was diluted with dichloromethane, washed with water, followed by brine solution. The organic layer was dried over anhydrous Na_2SO_4 and concentrated under reduced pressure to give crude product, which was purified by chromatography (PE: EtOAc: Et_3N = 20: 10: 1) on silica gel to afford **4a-4h** and **5a-5h** as a solid.

1.1.3.1 N-(1-benzylpiperidin-4-yl)-4-oxo-4H-chromene-3-carboxamide (4a)

Intermediate **3a** was treated with 1-benzylpiperidin-4-amine according to the general procedure to give the desired product **4a** as a pale white solid, yield 68%, m.p. 125-127 °C; IR (KBr) *v* 3474, 1661, 1615, 1566, 1544, 1464, 1391, 852, 770, 698; ^{1}H NMR (500 MHz, DMSO-d_6) δ 9.21 (s, 1H), 9.04 (s, 1H), 8.18 (d, J = 7.7 Hz, 1H), 7.91 (t, J = 7.3 Hz, 1H), 7.78 (d, J = 8.4 Hz, 1H), 7.61 (t, J = 7.5 Hz, 1H), 7.40 (s, 5H), 3.96 (s, 2H), 1.78-2.21 (m, 8H), 1.40 (s, 1H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 177.19, 162.39 (2C), 162.31, 156.12, 134.71, 130.50 (2C), 128.89 (2C), 126.40, 126.22, 124.23, 118.42 (2C), 115.64, 61.75 (2C), 51.06, 44.93, 29.69 (2C).ESI-MS *m/z*: 363.16 $[M + H]^+$; HRMS: calcd for $C_{22}H_{23}N_2O_3$ $[M + H]^+$, 363.1703, found 363.1704.

1.1.3.2 N-(1-benzylpiperidin-4-yl)-6-methoxy-4-oxo-4H-chromene-3-carboxamide (4b)

Intermediate **3b** was treated with 1-benzylpiperidin-4-amine according to the general procedure to give the desired product **4b** as a pale white solid, yield 72%, m.p.

198-200 °C; IR (KBr) *v* 3449, 1639, 1548, 1483, 1400, 1308, 1175, 669; ^{1}H NMR (500 MHz, $CDCl_3$) *δ* 9.40 (s, 1H), 8.94 (s, 1H), 7.61 (d, *J* = 2.8 Hz, 1H), 7.48 (dd, *J* = 9.1 Hz, 2.5 Hz, 1H), 7.30-7.40 (m, 5H), 7.28 (d, *J* = 9.0 Hz, 1H), 4.06 (s, 1H), 3.92 (s, 3H), 3.58 (s, 2H), 2.85 (s, 2H), 2.31 (s, 2H), 2.02 (s, 2H), 1.72 (s, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) *δ* 177.24, 162.37, 162.10, 157.93, 151.17, 129.45 (2C), 128.46 (2C), 127.37, 125.18, 124.81, 119.97, 115.29, 105.30 (2C), 63.11, 56.14, 52.03 (2C), 46.12, 31.79 (2C).ESI-MS *m/z*: 393.17 $[M + H]^+$; HRMS: calcd for $C_{23}H_{25}N_2O_4$ $[M + H]^+$, 393.1809, found 393.1807.

1.1.3.3 6-(benzyloxy)-N-(1-benzylpiperidin-4-yl)-4-oxo-4H-chromene-3-carboxamide

(4c)

Intermediate **3c** was treated with 1-benzylpiperidin-4-amine according to the general procedure to give the desired product **4c** as a pale yellow solid, yield 48%, m.p. 160-162 °C; IR (KBr) *v* 3454, 1669, 1567, 1549, 1481, 1449, 845, 805, 774, 733; ^{1}H NMR (500 MHz, $CDCl_3$) *δ* 9.44 (t, *J* = 33.6 Hz, 1H), 8.94 (s, 1H), 7.71 (d, *J* = 3.0 Hz, 1H), 7.50 (d, *J* = 9.1 Hz, 1H), 7.46 (d, *J* = 7.3 Hz, 2H), 7.26-7.43 (m, 9H), 5.17 (s, 2H), 4.06 (s, 1H), 3.61 (S, 2H), 2.88 (s, 2H), 2.34 (s, 2H), 2.13 – 1.97 (m, 2H), 1.76 (s, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) *δ* 177.04, 162.27, 161.99, 156.89, 151.12, 135.97, 129.44 (2C), 128.76 (2C), 128.38, 127.66 (2C), 127.41, 125.14 (2C) 125.05, 124.93，119.93, 115.16, 106.52, 70.79, 62.87, 51.84 (2C), 45.83, 31.54 (2C)；ESI-MS *m/z*: 469.20 $[M + H]^+$; HRMS: calcd for $C_{29}H_{29}N_2O_4$ $[M + H]^+$, 469.2122, found 469.2119.

1.1.3.4 N-(1-benzylpiperidin-4-yl)-6-methyl-4-oxo-4H-chromene-3-carboxamide

(4d)

Intermediate **3d** was treated with 1-benzylpiperidin-4-amine according to the general procedure to give the desired product **4d** as a pale white solid, yield 75%, m.p. 163-165 °C; IR (KBr) *v* 3454, 1673, 1548, 1484, 1398, 1165, 979, 807, 787, 697; ^{1}H NMR (500 MHz, $CDCl_3$) δ 9.37 (s, 1H), 8.94 (s, 1H), 8.05 (s, 1H), 7.55 (dd, *J* = 8.6, 1.9 Hz, 1H), 7.44 (d, *J* = 8.5 Hz, 1H), 7.40 – 7.26 (m, 5H), 4.05 (s, 1H), 3.54 (s, 2H), 2.81 (s, 2H), 2.49 (s, 3H), 2.27 (s, 2H), 2.01 (d, *J* = 10.5 Hz, 2H), 1.69 (d, *J* = 9.8 Hz, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 177.40, 162.19, 162.16, 154.49, 136.48, 135.82, 129.18 (2C), 128.27 (2C), 127.09, 125.47, 124.02, 118.18, 115.76, 63.12, 52.01 (2C), 46.24, 31.96 (2C), 21.05; ESI-MS *m/z*: 377.18 $[M + H]^+$; HRMS: calcd for $C_{23}H_{25}N_2O_3$ $[M + H]^+$, 377.1860, found 377.1859.

1.1.3.5 N-(1-benzylpiperidin-4-yl)-6-bromo-4-oxo-4H-chromene-3-carboxamide (4e)

Intermediate **3e** was treated with 1-benzylpiperidin-4-amine according to the general procedure to give the desired product **4e** as a pale white solid, yield 72%, m.p. 139-141 °C; IR (KBr) *v* 3443, 1631, 1551, 1513, 1454, 1400, 804, 740, 721, 696; ^{1}H NMR (500 MHz, $CDCl_3$) δ 9.21 (s, 1H), 8.95 (s, 1H), 8.40 (d, *J* = 2.3 Hz, 1H), 7.83 (dd, *J* = 8.9, 2.4 Hz, 1H), 7.45 (d, *J* = 8.9 Hz, 1H), 7.30-7.37 (m, 5H), 4.04 (s, 1H), 3.55 (s, 2H), 2.82 (s, 2H), 2.27 (s, 2H), 2.00 (s, 2H), 1.70 (s, 2H);^{13}C NMR (126 MHz, $CDCl_3$)δ 177.19, 162.39 (2C), 162.31, 156.12 (2C), 134.71, 130.50 (2C), 128.89 (2C), 126.40, 126.22, 124.23, 118.42, 115.64, 61.75, 51.06 (2C), 44.93, 29.69 (2C). ESI-MS *m/z*: 441.07 $[M + H]^+$; HRMS: calcd for $C_{22}H_{22}BrN_2O_3$ $[M + H]^+$, 441.0808,

found 441.0809.

1.1.3.6 N-(1-benzylpiperidin-4-yl)-7-methoxy-4-oxo-4H-chromene-3-carboxamide (4f)

Intermediate **3f** was treated with 1-benzylpiperidin-4-amine according to the general procedure to give the desired product **4f** as a pale white solid, yield 46%, m.p. 166-168 °C; IR (KBr) *v* 3451, 1641, 1549, 1513, 1399, 696, 668, 644; ^{1}H NMR (500 MHz, $CDCl_3$) δ 9.57 (s, 1H), 8.84 (s, 1H), 8.15 (d, J = 9.0 Hz, 1H), 7.55 (s, 2H), 7.51 – 7.30 (m, 3H), 7.05 (dd, J = 9.0, 2.2 Hz, 1H), 6.91 (d, J = 2.1 Hz, 1H), 4.12 (s, 1H), 3.96 (s, 1H), 3.92 (s, 3H), 3.12 (s, 2H), 2.0-2.3 (m, 6H);^{13}C NMR (126 MHz, $CDCl_3$) δ 164.95, 162.68 (2C), 161.85, 158.03, 130.76, 129.02 (2C), 127.57 (2C), 117.88, 115.84, 115.44, 100.57 (2C), 63.50, 61.91, 56.05 (2C), 31.92, 29.71 (2C). ESI-MS *m/z*: 393.17 $[M + H]^+$; HRMS: calcd for $C_{23}H_{25}N_2O_4$ $[M + H]^+$, 393.1809, found 393.1809.

1.1.3.7 N-(1-benzylpiperidin-4-yl)-7-bromo-4-oxo-4H-chromene-3-carboxamide (4g)

Intermediate **3g** was treated with 1-benzylpiperidin-4-amine according to the general procedure to give the desired product **4g** as a pale white solid, yield 38%, m.p. 172-174 °C; IR (KBr) *v* 3454, 1689, 1607, 1550, 1420, 979, 869, 855, 792, 695, 668; ^{1}H NMR (500 MHz, $CDCl_3$) δ 9.22 (s, 1H), 8.92 (s, 1H), 8.14 (d, J = 8.6 Hz, 1H), 7.75 (d, J = 1.4 Hz, 1H), 7.62 (dd, J = 8.6, 1.5 Hz, 1H), 7.42 – 7.29 (m, 4H), 7.23 (s, 1H), 4.03 (s, 1H), 3.56 (s, 2H), 2.83 (s, 2H), 2.27 (s, 2H), 2.00 (s, 2H), 1.70 (s, 2H); 13C NMR (126 MHz, CDCl3) δ 176.65, 162.29, 162.24 (2C), 156.14, 129.97 (2C), 129.08, 128.26, 127.55 (2C), 123.23, 121.59 (2C), 116.32, 53.56 (2C), 37.03, 36.03

(2C), 33.38, 31.83.ESI-MS *m/z*: 441.07 $[M + H]^+$; HRMS: calcd for $C_{22}H_{22}BrN_2O_3$ $[M + H]^+$, 441.0808, found 441.0809.

1.1.3.8 N-(1-benzylpiperidin-4-yl)-4-oxo-4H-chromene-2-carboxamide (4h)

Intermediate **3h** was treated with 1-benzylpiperidin-4-amine according to the general procedure to give the desired product **4h** as a pale white solid, yield 78%, m.p. 185-187 °C; IR (KBr) *v* 3447, 1640, 1546,1397, 1066, 668, 419; ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.94 (d, J = 7.9 Hz, 1H), 8.07 (dd, J = 7.9, 1.1 Hz, 1H), 8.00 – 7.85 (m, 1H), 7.79 (d, J = 8.4 Hz, 1H), 7.56 (t, J = 7.5 Hz, 1H), 7.42 – 7.29 (m, 4H), 7.28 (d, J = 6.7 Hz, 1H), 6.85 (s, 1H), 3.91 – 3.69 (m, 1H), 3.50 (s, 2H), 2.86 (d, J = 10.8 Hz, 2H), 2.04 (t, J = 11.3 Hz, 2H), 1.81 (d, J = 10.8 Hz, 2H), 1.62-1.75 (m, 2H); ^{13}C NMR (126 MHz, DMSO-d_6) δ 177.79, 158.84, 156.29, 155.63, 139.13, 135.39, 129.18 (2C), 128.65 (2C), 127.35, 126.47, 125.39, 124.15, 119.39, 111.03, 62.49, 52.58 (2C), 47.83, 31.64 (2C); ESI-MS *m/z*: 363.17 $[M + H]^+$; HRMS: calcd for $C_{22}H_{23}N_2O_3$ $[M + H]^+$, 363.1703, found 363.1703.

1.1.3.9 N-(2-(1-benzylpiperidin-4-yl)ethyl)-4-oxo-4H-chromene-3-carboxamide (5a)

Intermediate **3a** was treated with 2-(1-benzylpiperidin-4-yl) ethanamine according to the general procedure to give the desired product **5a** as a pale white solid, yield 74%, m.p. 101-103 °C; IR (KBr) *v* 3448, 2928, 1671, 1541, 1467, 1390, 1142, 859, 768, 695; ^{1}H NMR (500 MHz, DMSO-d_6) δ 9.16 (s, 1H), 9.05 (s, 1H), 8.20 (d, J = 7.9 Hz, 1H), 7.92 (s, 1H), 7.79 (d, J = 8.4 Hz, 1H), 7.62 (s, 1H), 7.39 – 7.27 (m, 4H), 7.26 (d, J = 6.8 Hz, 1H), 3.45 (s, 2H), 3.37 (d, J = 6.9 Hz, 2H), 2.80 (d, J = 10.9 Hz, 2H),

1.92 (t, *J* = 11.1 Hz, 2H), 1.69 (d, *J* = 12.2 Hz, 2H), 1.48-1.52 (m, 2H), 1.33-1.38 (m, 1H), 1.16-1.24 (m, 2H); ^{13}C NMR (126 MHz, DMSO-d_6) δ 176.41, 162.57, 161.70, 155.57, 138.52, 135.07, 128.66 (2C), 128.00 (2C), 126.69, 126.42, 125.41, 123.56, 118.59, 115.45, 62.38, 53.11 (2C), 36.16, 35.74, 32.96 (2C), 31.70； ESI-MS *m/z*: 391.19 $[M + H]^+$; HRMS: calcd for $C_{24}H_{27}N_2O_3$ $[M + H]^+$, 391.2016, found 391.2017.

1.1.3.10 N-(2-(1-benzylpiperidin-4-yl)ethyl)-6-methoxy-4-oxo-4H-chromene-3-carboxamide (5b)

Intermediate **3b** was treated with 2-(1-benzylpiperidin-4-yl) ethanamine according to the general procedure to give the desired product **5b** as a pale white solid, yield 65%, m.p. 165-167 °C; IR (KBr) *v* 3444, 2987, 2376, 2344, 1630, 1548, 1397, 1056, 668, 646; ^{1}H NMR (500 MHz, $CDCl_3$) δ 9.33 (s, 1H), 8.92 (s, 1H), 7.58 (d, *J* = 3.0 Hz, 1H), 7.54 (d, *J* = 3.9 Hz, 2H), 7.48 (d, *J* = 9.2 Hz, 1H), 7.38-7.42 (m, 3H), 7.33 (dd, *J* = 9.2, 3.1 Hz, 1H), 3.91 (s, 3H), 3.46-3.50 (m, 2H), 3.30 (s, 2H), 2.52 (s, 2H), 1.90 (s, 2H), 1.62-1.66 (m, 2H), 1.54 (s, 1H), 1.22-1.26 (m, 2H);13C NMR (126 MHz, CDCl3) δ 177.09, 163.13, 161.97 (2C), 157.85, 151.03, 131.09, 129.07 (2C), 125.00, 124.67, 119.85 (2C), 115.00, 105.29 (2C), 61.25, 56.01 (2C), 52.36, 36.41 (2C), 35.32, 31.94, 29.25. ESI-MS *m/z*: 421.20 $[M + H]^+$; HRMS: calcd for $C_{25}H_{29}N_2O_4$ $[M + H]^+$, 421.2122, found 421.2122.

1.1.3.11 6-(benzyloxy)-N-(2-(1-benzylpiperidin-4-yl)ethyl)-4-oxo-4H-chromene-3-carboxamide (5c)

Intermediate **3c** was treated with 2-(1-benzylpiperidin-4-yl) ethanamine according to the general procedure to give the desired product **5c** as a pale yellow solid, yield

66%, m.p. 142-144 °C; IR (KBr) *v* 3450, 2931, 1674, 1565, 1533, 1482, 1451, 1392, 1301, 833, 800, 739, 698; ^{1}H NMR (500 MHz, $CDCl_3$) δ 9.29 (s, 1H), 8.95 (s, 1H), 7.71 (d, *J* = 3.0 Hz, 1H), 7.50 (d, *J* = 9.1 Hz, 1H), 7.48 – 7.42 (m, 5H), 7.40 (d, *J* = 2.8 Hz, 1H), 7.30-7.37 (m, 5H), 5.17 (s, 2H), 3.53 (s, 2H), 3.51 – 3.43 (m, 2H), 2.01 (s, 2H), 1.74 (d, *J* = 10.6 Hz, 2H), 1.58-1.62 (m, 2H), 1.40 (s, 2H), 1.26 (s, 2H), 0.88 (s, 1H);^{13}C NMR (126 MHz, $CDCl_3$) δ 177.07, 162.90, 161.97 (2C), 156.86, 151.14, 135.98, 128.76, 128.39 (2C), 128.30 (3C), 127.65 (3C), 125.08 (2C), 119.94 (2C), 115.19, 106.60 (2C), 70.79 (2C), 63.20, 53.57, 36.94, 36.05 ,33.37, 29.71. ESI-MS *m/z*: 497.24 $[M + H]^+$; HRMS: calcd for $C_{31}H_{33}N_2O_4$ $[M + H]^+$, 497.2435, found 497.2433.

1.1.3.12 N-(2-(1-benzylpiperidin-4-yl)ethyl)-6-methyl-4-oxo-4H-chromene-3-carboxamide (5d)

Intermediate **3d** was treated with 2-(1-benzylpiperidin-4-yl) ethanamine according to the general procedure to give the desired product **5d** as a pale white solid, yield 68%, m.p. 137-139 °C; IR (KBr) *v* 3448, 2923, 1664, 1545, 1484, 1400, 1312, 1159,805, 650; ^{1}H NMR (500 MHz, $CDCl_3$) δ 9.29 (s, 1H), 8.94 (s, 1H), 8.05 (s, 1H), 7.55 (dd, *J* = 8.6, 1.9 Hz, 1H), 7.44 (d, *J* = 8.6 Hz, 1H), 7.28-7.31 (m, 4H), 7.21-7.25 (m, 1H), 3.65 – 3.28 (m, 4H), 2.88 (d, *J* = 11.5 Hz, 2H), 2.49 (s, 3H), 1.97 (t, *J* = 11.0 Hz, 2H), 1.72 (d, *J* = 12.6 Hz, 2H), 1.57-1.61 (m, 2H), 1.47 – 1.20 (m, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 177.37, 162.85, 162.11, 154.48, 138.46, 136.42, 135.80, 129.25 (2C), 128.13 (2C), 126.92, 125.45, 124.00, 118.16, 115.73, 63.43, 53.71 (2C), 36.98, 36.13 , 33.55 (2C), 32.16, 21.02; ESI-MS *m/z*: 405.27 $[M + H]^+$; HRMS: calcd for

$C_{25}H_{29}N_2O_3$ $[M + H]^+$, 405.2173, found 405.2173.

1.1.3.13 N-(2-(1-benzylpiperidin-4-yl)ethyl)-6-bromo-4-oxo-4H-chromene-3-carboxamide (5e)

Intermediate **3e** was treated with 2-(1-benzylpiperidin-4-yl) ethanamine according to the general procedure to give the desired product **5e** as a pale white solid, yield 58%, m.p. 117-119 °C; IR (KBr) *v* 3453, 2923, 1641, 1398, 1057, 696, 668, 519; ^{1}H NMR (500 MHz, $CDCl_3$) δ 9.17 (s, 1H), 9.01 (s, 1H), 8.45 (d, J = 2.4 Hz, 1H), 7.88 (dd, J = 8.9, 2.4 Hz, 1H), 7.50 (d, J = 8.9 Hz, 1H), 7.34-7.36 (m, 4H), 7.30 – 7.26 (m, 1H), 3.51-3.55 (m, 4H), 2.94 (d, J = 10.7 Hz, 2H), 2.01-2.09 (m, 2H), 1.77 (d, J = 11.2 Hz, 2H), 1.62-1.66 (m, 2H), 1.31-1.41 (m, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 176.18, 162.53, 162.38, 155.07, 137.75, 129.45, 129.00 (2C), 128.32 (2C), 127.14, 125.81, 120.47, 120.01, 116.30, 63.53, 53.82 (2C), 37.22, 36.21, 33.68 (2C), 32.22; ESI-MS *m/z*: 469.10 $[M + H]^+$; HRMS: calcd for $C_{24}H_{26}BrN_2O_3$ $[M + H]^+$, 469.1121, found 469.1120.

1.1.3.14 N-(2-(1-benzylpiperidin-4-yl)ethyl)-7-methoxy-4-oxo-4H-chromene-3-carboxamide (5f)

Intermediate **3f** was treated with 2-(1-benzylpiperidin-4-yl) ethanamine according to the general procedure to give the desired product **5f** as a pale white solid, yield 46%, m.p. 165-167 °C; ^{1}H NMR (500 MHz, $CDCl_3$) δ 9.39 (s, 1H), 8.92 (s, 1H), 8.21 (d, J = 9.0 Hz, 1H), 7.41 (d, J = 7.0 Hz, 2H), 7.37 (t, J = 7.2 Hz, 2H), 7.09 (dd, J = 8.9, 2.3 Hz, 1H), 6.96 (d, J = 2.2 Hz, 1H), 3.97 (dd, 2H), 3.65 (dd, 1H), 3.51 (dd, J = 13.3, 6.7 Hz, 2H), 3.00 (m, 2H), 2.11 (m, J = 25.9 Hz, 2H), 1.80 (m, J = 9.5 Hz, 2H), 1.68-

1.61 (m, 2H), 1.47 (m, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 176.67, 164.85, 162.96 (2C), 161.78, 158.06, 129.72, 129.34, 128.38, 127.51 (2C), 118.00, 115.73 (2C), 100.51 (2C), 56.02, 55.35, 53.41, 45.88, 36.83, 35.95, 33.15, 29.71. HRMS: calcd for $C_{25}H_{28}N_2O_4$ $[M + H]^+$, 469.2122, found 469.2120.

1.1.3.15 N-(2-(1-benzylpiperidin-4-yl)ethyl)-7-bromo-4-oxo-4H-chromene-3-carboxamide (5g)

Intermediate **3g** was treated with 2-(1-benzylpiperidin-4-yl) ethanamine according to the general procedure to give the desired product **5g** as a pale white solid, yield 22%, m.p. 132-134 °C; IR (KBr) *v* 3454, 2923, 1640, 1400, 663; ^{1}H NMR (500 MHz, $CDCl_3$) δ 9.14 (s, 1H), 8.92 (s, 1H), 8.13 (d, J = 8.6 Hz, 1H), 7.75 (d, J = 1.5 Hz, 1H), 7.61 (dd, J = 8.6, 1.6 Hz, 1H), 7.34 – 7.28 (m, 4H), 7.23-7.25 (m, 1H), 3.59 – 3.33 (m, 4H), 2.90 (d, J = 8.8 Hz, 2H), 1.99 (s, 2H), 1.72 (d, J = 10.6 Hz, 2H), 1.56-1.60 (m, 2H), 1.26-1.37 (m, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 176.56, 162.45, 161.12, 159.15, 152.40, 129.37 (2C), 128.20 (2C), 127.08, 125.70, 123.01, 120.69, 115.44, 111.28, 111.08, 63.30, 53.61 (2C), 37.07 (2C), 36.09, 33.51, 32.01.ESI-MS *m/z*: 469.10 $[M + H]^+$; HRMS: calcd for $C_{24}H_{26}BrN_2O_3$ $[M + H]^+$, 469.1121, found 469.1121.

1.1.3.16 N-(2-(1-benzylpiperidin-4-yl)ethyl)-4-oxo-4H-chromene-2-carboxamide (5h)

Intermediate **3h** was treated with 2-(1-benzylpiperidin-4-yl) ethanamine according to the general procedure to give the desired product **5h** as a pale white solid, yield 76%, m.p. 154-156 °C; IR (KBr) *v* 3450, 2925, 1641, 1530, 1462, 1399, 1267, 1108,

691; ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.22 (dd, J = 7.9, 1.4 Hz, 1H), 7.80 - 7.68 (m, 1H), 7.52 (d, J = 8.4 Hz, 1H), 7.45 (t, J = 7.5 Hz, 1H), 7.39 – 7.28 (m, 5H), 7.15 (s, 1H), 6.88 (s, 1H), 3.48-3.66 (m, 4H), 2.92 (d, J = 10.6 Hz, 2H), 2.00 (s, 2H), 1.73 (d, J = 8.8 Hz, 2H), 1.68 – 1.58 (m, 2H), 1.38 (s, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 178.10, 159.24, 155.29, 154.77, 134.48, 129.35 (2C), 128.25 (2C), 127.16, 126.19, 125.97, 124.41, 118.03, 112.14, 63.29, 53.58 (2C), 37.82, 36.14, 33.55, 31.99 (2C); ESI-MS *m/z*: 391.19 $[M + H]^+$; HRMS: calcd for $C_{24}H_{27}N_2O_3$ $[M + H]^+$, 391.2016, found 391.2018.

1.2. Inhibitory activity against AChE and BuChE

Butyrylcholinesterase (BuChE, E.C. 3.1.1.8, from equine serum), acetylcholinesterase (AChE, E.C. 3.1.1.7, from electric eel), 5, 5'-dithiobis-(2-nitrobenzoic acid) (Ellman's reagent, DTNB), acetylthiocholine iodide (ATCI), *S*-butyrylthiocholine iodide (BTCI) and donepezil hydrochloride were purchased from Sigma-Aldrich. The capacity of the test compounds (**4a**-**4h** and **5a**-**h**) to inhibit BuChE and AChE activities were assessed by Ellman's method. Stock solution of test compounds was dissolved in a minimum volume of DMSO (1%) and was diluted using the buffer solution (50 mM Tris-HCl, *p*H= 8.0, 0.02 M $MgCl_2 \cdot 6H_2O$, 0.1 M NaCl). In 96-well plates, 50 µL of BuChE (0.12 U/mL prepared in 50 mM Tris-HCl, *p*H=8.0, 0.1% w/v BSA) or 50 µL of AChE (0.22 U/mL prepared in 50 mM Tris-HCl, *p*H =8.0, 0.1% w/v bovine serum albumin, BSA), 160 µL of 1.5 mM DTNB were incubated with 10 µL of various concentrations of test compounds (0.001-100 µM) at 37 °C for 6 min followed by the addition of the substrates (30 µL) *S*-

butyrylthiocholine iodide (15 mM) or acetylthiocholine iodide (15 mM) and the absorbance was measured at different time intervals (0, 60, 120, and 180 s) at a wavelength of 405 nm. The concentration of compound producing 50% of enzyme activity inhibition (IC_{50}) was calculated by nonlinear regression analysis of the response-concentration (log) curve, using the Graph-Pad Prism program package (Graph Pad Software; San Diego, CA). Results are expressed as the mean ± SEM of at least three different experiments performed in triplicate.

1.3. Inhibitory activity against h-MAOB and h-MAOA

Monoamine Oxidases (*h*MAO-B, *h*MAO-A, E.C. 1.4.3.4), *p*-tyramine horseradish peroxidase (E.C. 1.11.1.7) and Amplex Red were bought from Sigma-Aldrich. Firstly, MAOs activity was adjusted to obtain in our experimental conditions the same reaction velocity in the presence of both isoforms (i.e., to oxidize (in the control group) the same concentration of substrate: 165 pmol of *p*-tyramine/min(*h*MAO-B: 7.5µg protein; specific activity: 22 nmol of *p*-tyramine transformed/min/mg protein ; *h*-MAOA: 1.1µg protein; specific activity: 150 nmol of *p*-tyramine oxidized to *p*-hydroxyphenylacetaldehyde/min/mg protein). The DMSO (10 mM) was added to compounds and diluted in 0.05 M KH_2PO_4/K_2HPO_4 buffer (*p*H = 7.4) to the desired final concentration. All the compounds are soluble at the tested concentration. Test drugs (20 µL) and MAO (80 µL) were incubated at 37 °C for 15 min in a flat-black-bottom 96-well microtest plate in dark. The reaction was started by adding 2 U/mL horseradish peroxidase, 2 mM *p*-tyramine and 200 µM Amplex Red reagent for *h*MAO at 37 °C for 20 min. The reaction was quantified in a multidetection

microplate fluorescence reader based on the fluorescence generated (excitation, 545 nm; emission, 590 nm). The specific fluorescence emission was calculated after subtraction of the background activity by a sodium phosphate buffer solution. The percent inhibition was calculated by the following expression: $(1 - I_{Fi}/I_{Fc}) \times 100$ in which I_{Fi} and I_{Fc} are the fluorescence intensities obtained for *h*-MAO in the presence and absence of inhibitors after subtracting the respective background.

1.4. Kinetic study of ChEs

Three concentrations of **5c** were selected for the kinetic studies: 10.5, 5.2 and 2.6 μM for the analysis of BuChE inhibition, and 0.74, 0.37 and 0.185 μM for the analysis of AChE inhibition, respectively. The initial catalytic rates of the ChEs-catalyzed hydrolysis of substrates (BTCI for the BuCh, EATCI for the AChE) at 6 diverse substrate concentrations (50, 75, 100, 150, 200 and 500 μM) in the absence and presence of 3 diverse concentrations of **5c** were measured. The plots were assessed by a weighted least-squares analysis that assumed the variance of velocity (v) to be a constant percentage of v for the entire data set. Slopes of these reciprocal plots were then plotted against the concentration of **5c** in a weighted analysis and *Ki* was determined as the intercept on the negative x-axis. Data analysis was performed with GraphPad Prism 4.03 software (GraphPad Software Inc.).

1.5. Kinetic study of h-MAOB

Firstly, compounds **5c** and pargyline were prepared at a concentration of 100 × IC_{50} and 10 × IC_{50} value for the reversibility and irreversibility study. Test compounds and *h*-MAOB (0.75 mg/ml) were incubated at 37 °C for 30 min in a flat-

black-bottom 96-well microtest plate in dark. Control incubations were conducted in the absence of inhibitor, and DMSO (4%) was added as co-solvent to all preincubations. The reactions were subsequently diluted 100-fold to yield final concentrations of compounds equal to 0.1 × IC_{50} and 1 × IC_{50}. And a similar method to the one described above (in section Inhibitory activity against *h*-MAOA and *h*-MAOB) was used for the subsequent measurement of activity of *h*-MAOB. All measurements were carried out in triplicate and are expressed as mean ± SD.

Then, three concentrations of **5c** (0.54, 0.27 and 0.13 μM) were selected for the kinetic study. The initial rates of the MAOB-catalyzed oxidation of *p*-tyramine at six different substrate concentrations in the absence and in the presence of three different concentrations (0.05, 0.1, 0.25, 0.33, 0.5, and 1.0 mM) of **5c** were measured. The plots were assessed by a weighted least-squares analysis that assumed the variance of velocity (*v*) to be a constant percentage of *v* for the entire data set. Slopes of these reciprocal plots were then plotted against the concentration of **5c** in a weighted analysis and *Ki* was determined as the intercept on the negative *x*-axis. Data analysis was performed with GraphPad Prism 4.03 software (GraphPad Software Inc.).

1.6. Molecular modeling studies of compound 5c with AChE, h-MAOB and h-MAOA

Molecular modeling calculations and docking studies were performed using Molecular Operating Environment (MOE) software version 2008.10 (Chemical Computing Group, Montreal,Canada). The X-ray crystallographic structure of human MAOB in complexed with 7-(3-chlorobenzyloxy)-4-formylcoumarin (PDB code

2V60), Crystal structure of human monoamine oxidase A with Harmine (PDB code 2Z5Y) and AChE in complexed with donepezil (PDB code 1EVE) were obtained from the Protein Data Bank. The compound **5c** was built using the MOE program and energy minimized using MMFF94x forcefield. Then the **5c** was docked into the active site of the protein by the "Triangle Matcher" method, which generated poses by aligning the ligand triplet of atoms with the triplet of alpha spheres in cavities of tight atomic packing. The Dock scoring in MOE software was done using ASE scoring function and forcefield was selected as the refinement method. The best 10 poses of molecules were retained and scored. After docking, the geometry of resulting complex was studied using the MOE's pose viewer utility.

1.7. In vitro blood–brain barrier permeation assay

Brain penetration of compounds was evaluated using a parallel artificial membrane permeation assay (PAMPA). The 96-well UV plate (COSTAR@) was from Corning Incorporated. Commercial drugs were purchased from Alfa Aesar and Sigma. The porcine brain lipid (PBL) was obtained from Avanti Polar Lipids. The acceptor microplate and the donor microplate (PVDF membrane, pore size 0.45 mm) were both from Millipore. The acceptor 96-well microplate was filled with 300 μL of PBS/EtOH (7:3), and the filter membrane was impregnated with 4 μL of PBL in dodecane (20 mg/mL). The compound solutions (DMSO at 5 mg/mL) were diluted 50-fold in PBS/EtOH (7:3) to achieve 100 mg/mL, 200 μL of which were added to the donor wells. The acceptor filter plate was laid on the donor plate, which was left undisturbed for 16 h at 25 °C. After incubation, the donor plate was removed and the

concentration of compound in the acceptor wells was determined using a UV plate reader (Flexsta-tion@ 3). In each experiment, 9 quality control standards of known BBB permeability were included to validate the analysis set. Every sample was analyzed at five wavelengths, in four wells, in at least three independent runs, and the results are given as the mean ± standard deviation.

1.8. Rat pheochromocytoma (PC12) cell toxicity

The toxicity effect of compounds on the rat pheochromocytoma (PC12) cells (PC12 cells come from Cell Resource Center, Shanghai Institute of Life Sciences, Chinese Academy of Sciences). was examined. The PC12 cells were routinely grown at 37 °C in a humidified incubator with 5% CO_2 in Dulbecco's modified Eagle's medium (DMEM) supplemented with 100 units/mL of streptomycin, 100 units/mL penicillin, and 10% bovine calf serum. Cells were subcultured in 96-well plates at a seeding density of 10,000 cells per well and allowed to grow and adhere. When cells reached the required confluence, they were placed into serum-free medium and treated with compound **5c**. 24 h later the survival of cells was determined by MTT assay. In a few words, after incubation with 20 μL of MTT at 37 °C for four hours, living cells containing MTT formazan crystals were dissolved in 200 μL DMSO. The absorbance of each well was measured using a microculture plate reader with a reference wavelength of 630 nm and a test wavelength of 570 nm. Results are expressed as the mean ± SD of 3 independent experiments.

2. The ^{1}H NMR , ^{13}C NMR and HRMS spectra of representative compounds (compounds 4a, 4b, 4c, 4d, 4e, 4f, 4g, 4h, 5a, 5b, 5c, 5d, 5e, 5f, 5g, 5h)

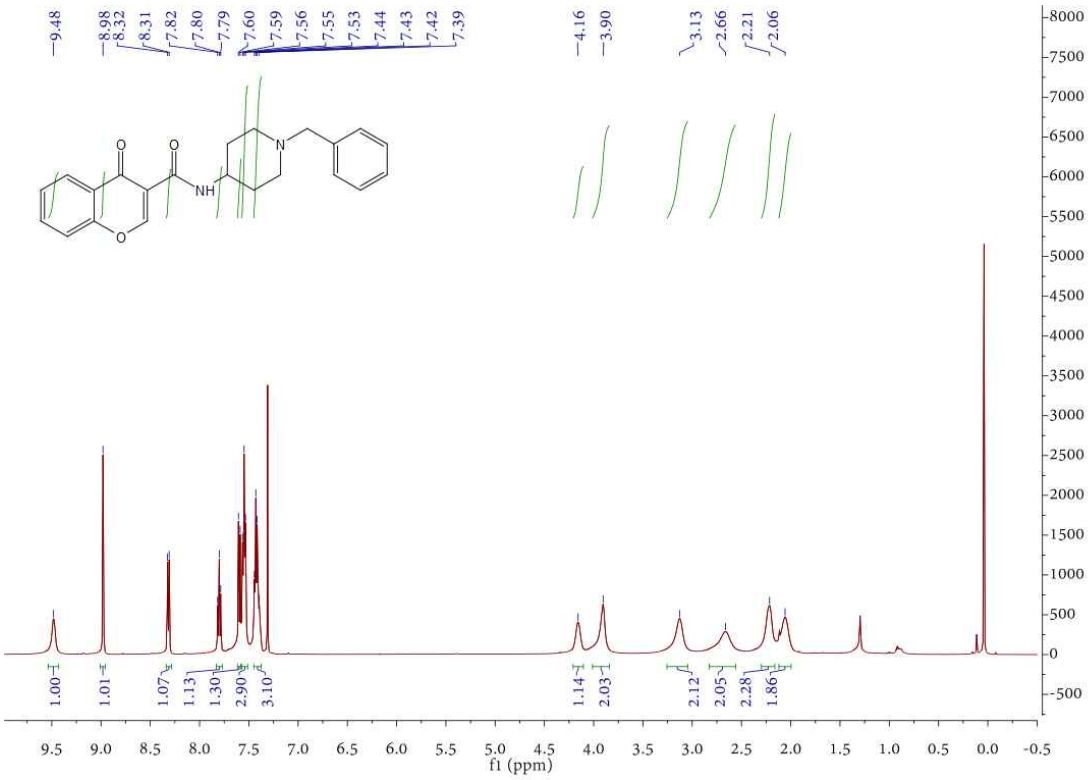


Figure 1. ^{1}H NMR Spectrum of **4a** in DMSO

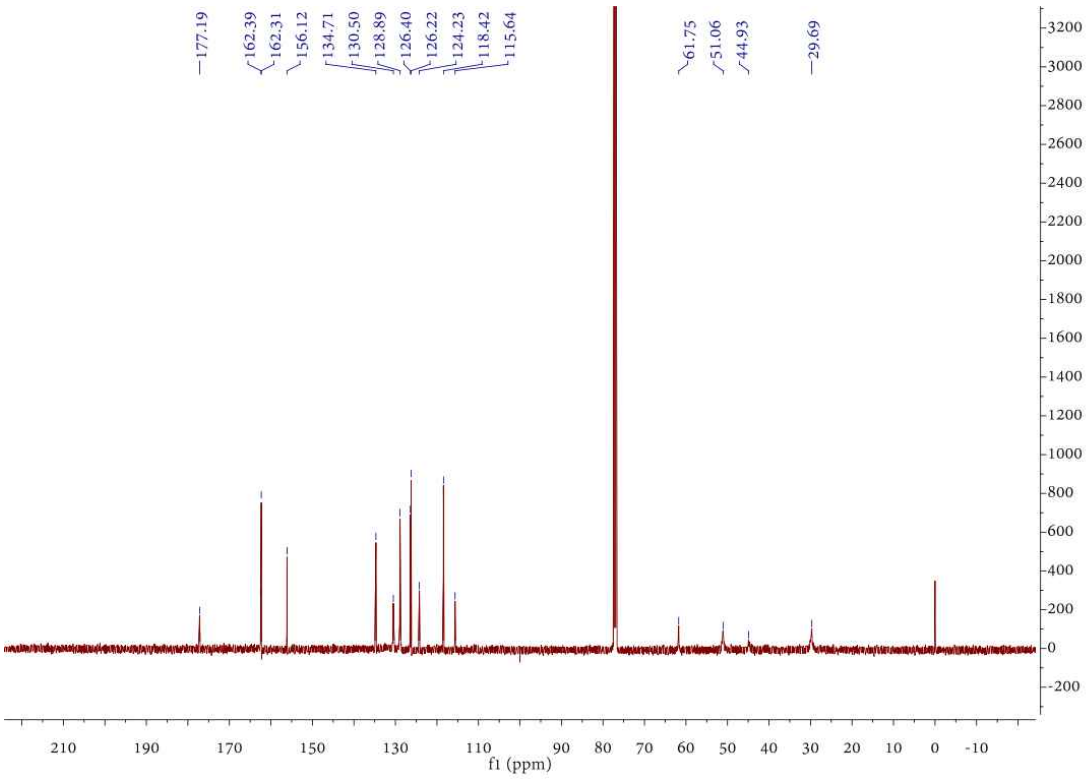
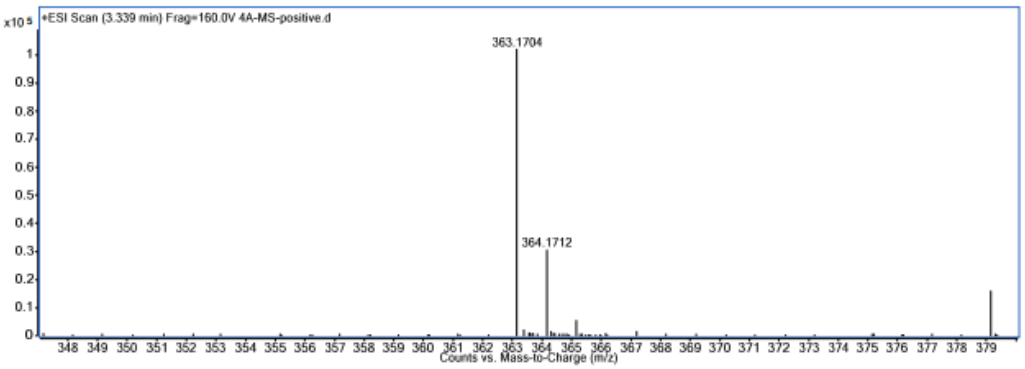


Figure 2. ^{13}C NMR Spectrum of **4a** in DMSO



Elemental Composition Calculator

Target m/z:	363.1704	**Result type:**	Positive ions	**Species:**	$[M+H]^+$
Elements:		C (0-80); H (0-120); O (0-30); N(0-5)			
Ion Formula		**Calculated m/z**		**PPM Error**	
C22H23N2O3		363.1703		-0.13	

Agilent Technologies

Figure 3. HRMS Spectrum of **4a**

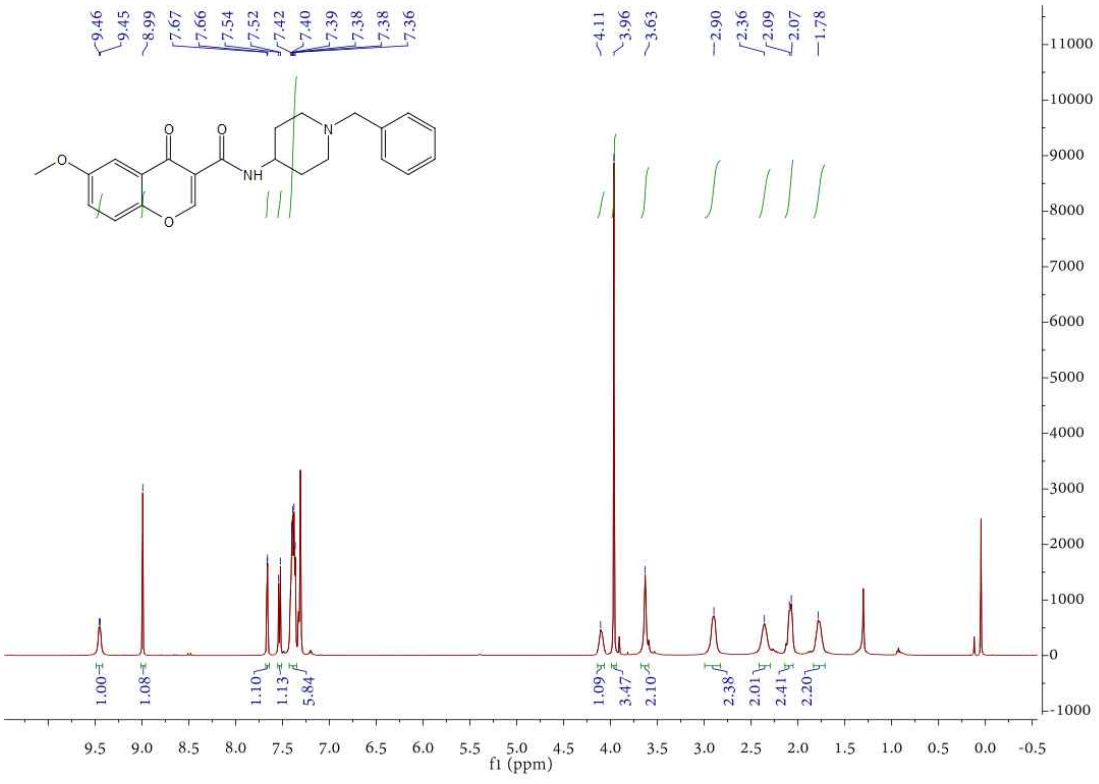


Figure 4. ^{1}H NMR Spectrum of **4b** in $CDCl_3$

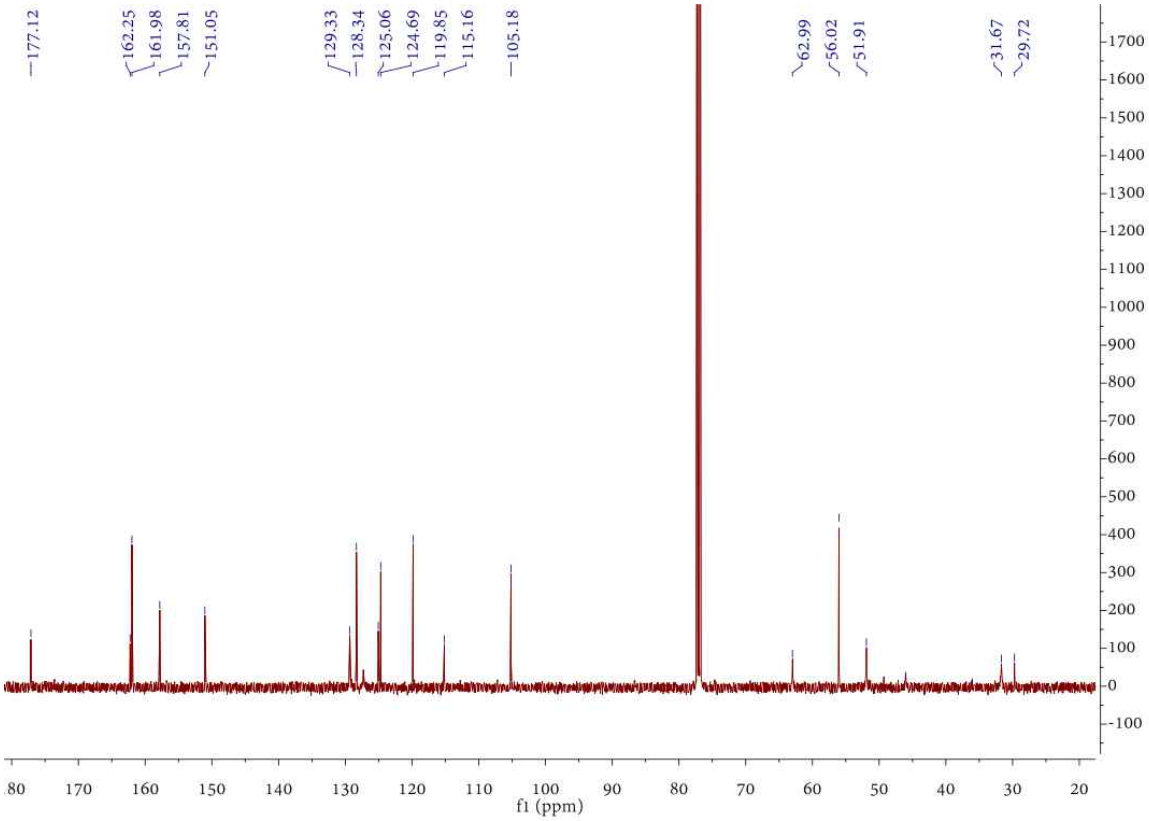
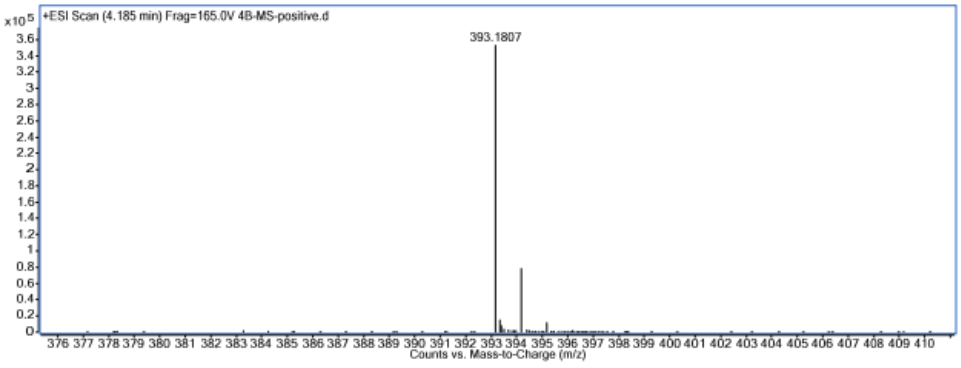


Figure 5. ^{13}C NMR Spectrum of **4b** in $CDCl_3$



Elemental Composition Calculator

Target m/z:	393.1807	**Result type:**	Positive ions	**Species:**	$[M+H]^+$
Elements:		C (0-80); H (0-120); O (0-30); N(0-5)			
Ion Formula		**Calculated m/z**		**PPM Error**	
C23H25N2O4		393.1809		0.51	

Agilent Technologies

Figure 6. HRMS Spectrum of **4b**

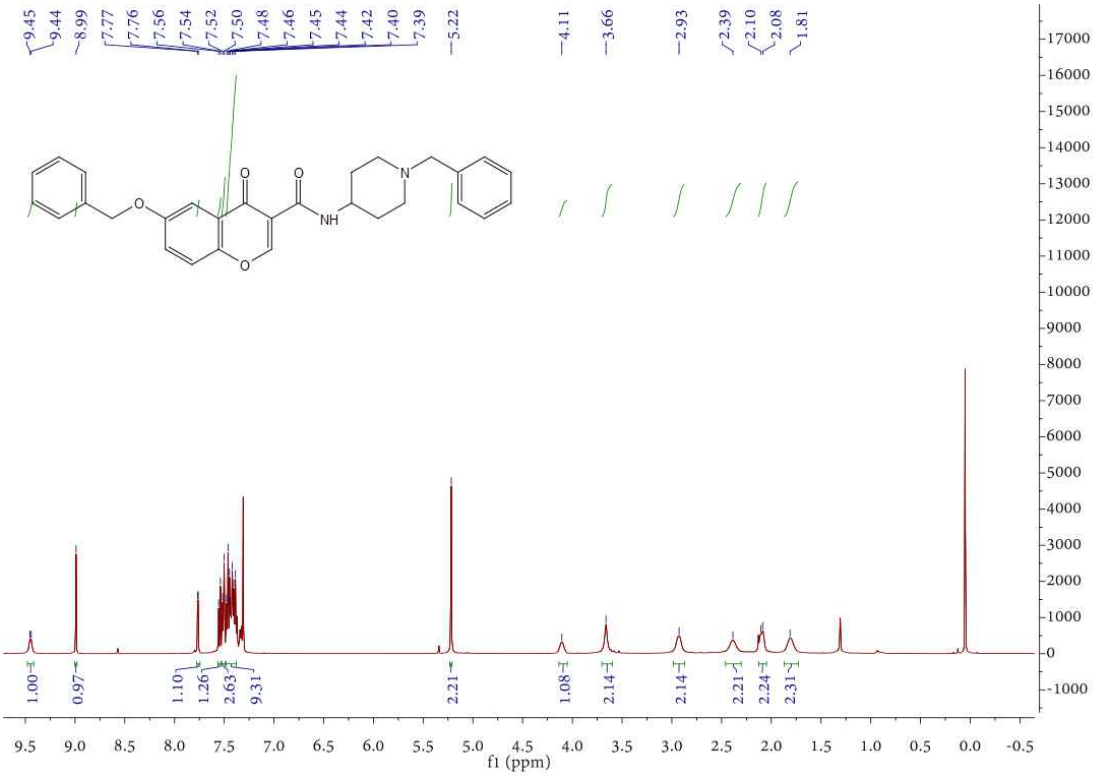


Figure 7. ^{1}H NMR Spectrum of **4c** in $CDCl_3$

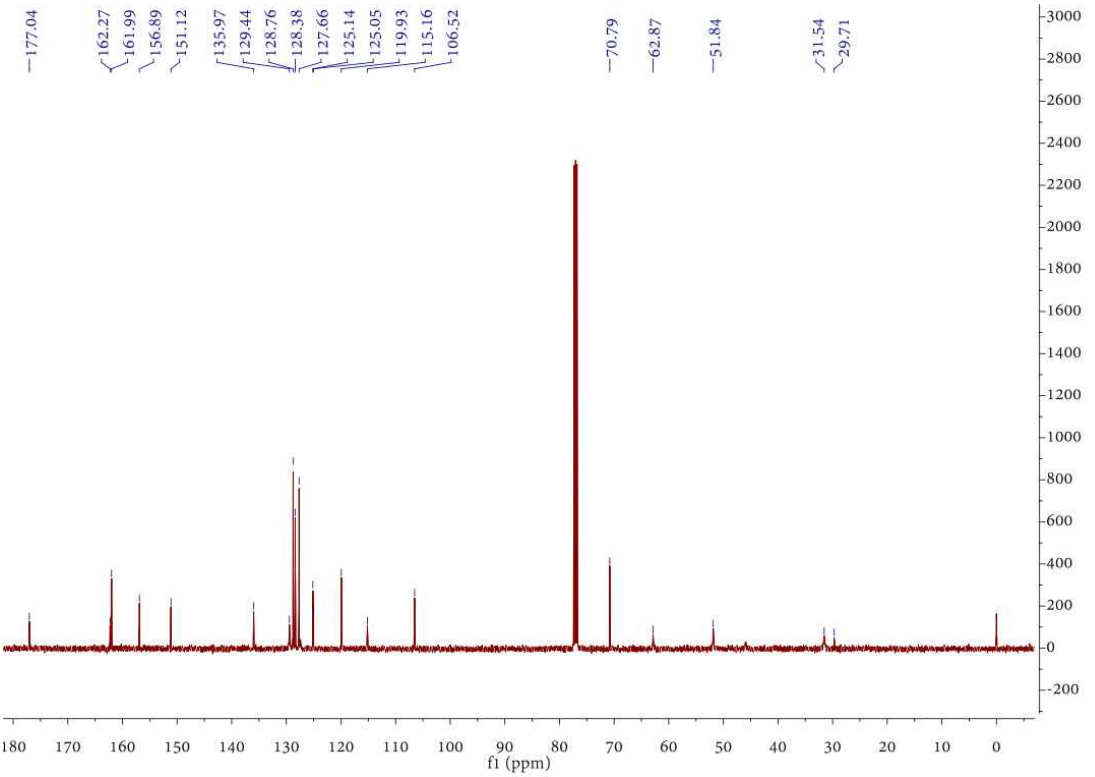
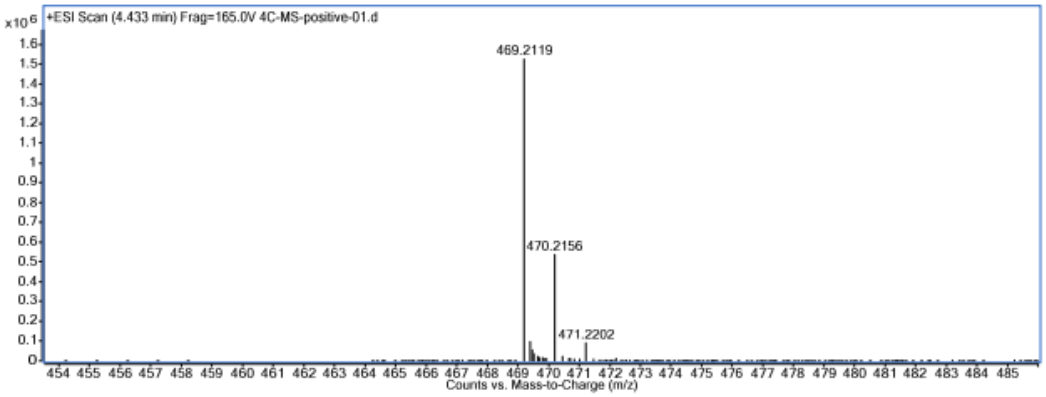


Figure 8. ^{13}C NMR Spectrum of **4c** in $CDCl_3$



Elemental Composition Calculator

Target m/z:	469.2119	**Result type:**	Positive ions	**Species:**	$[M+H]^+$
Elements:		C (0-80); H (0-120); O (0-30); N(0-5)			
Ion Formula		**Calculated m/z**		**PPM Error**	
$C_{29}H_{29}N_2O_4$		469.2122		0.52	

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Figure 9. HRMS Spectrum of **4c**

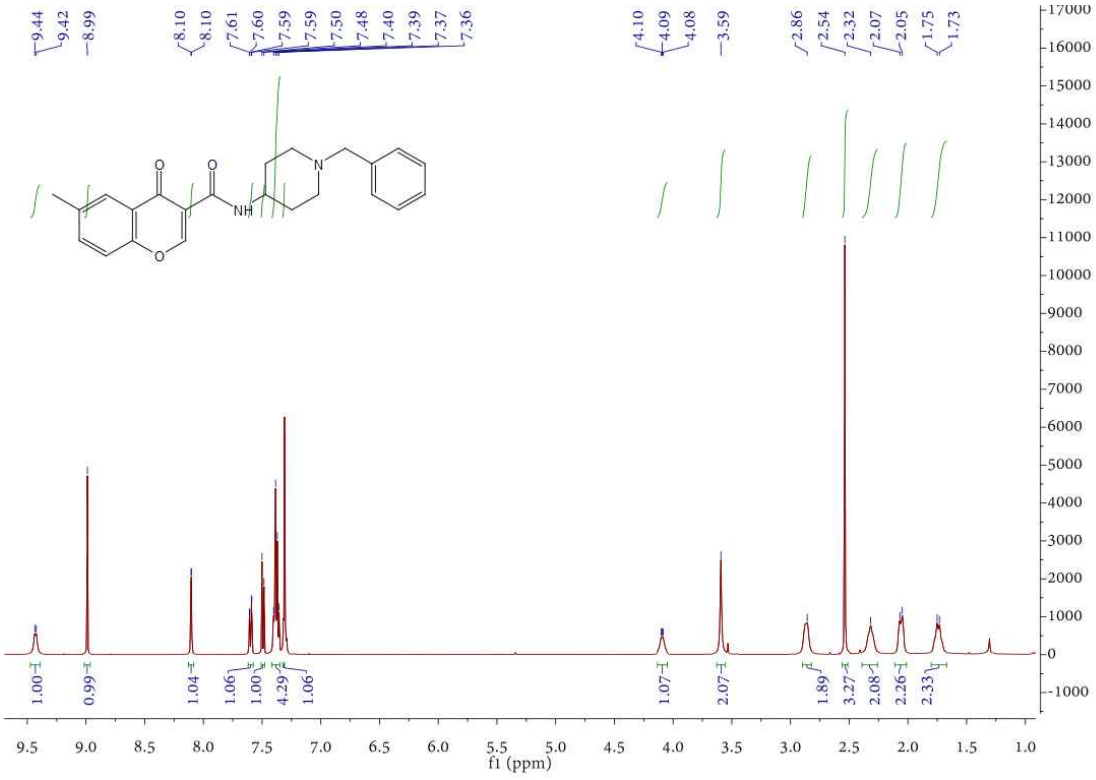


Figure 10. ^1H NMR Spectrum of **4d** in $CDCl_3$

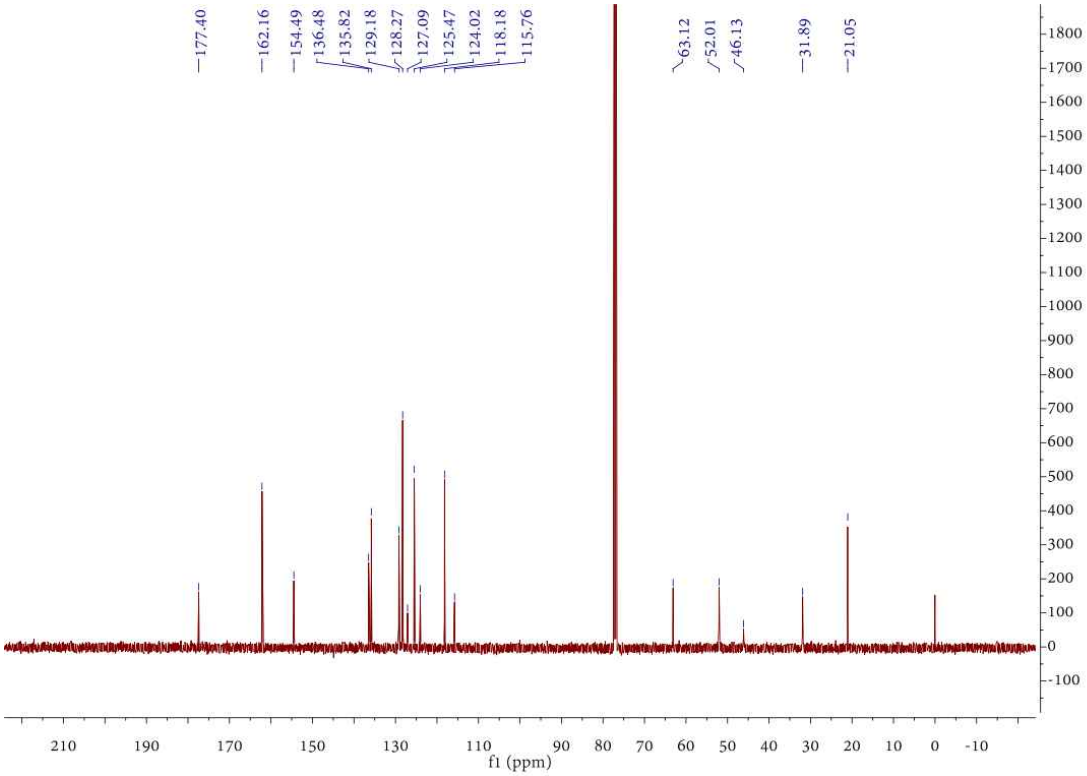
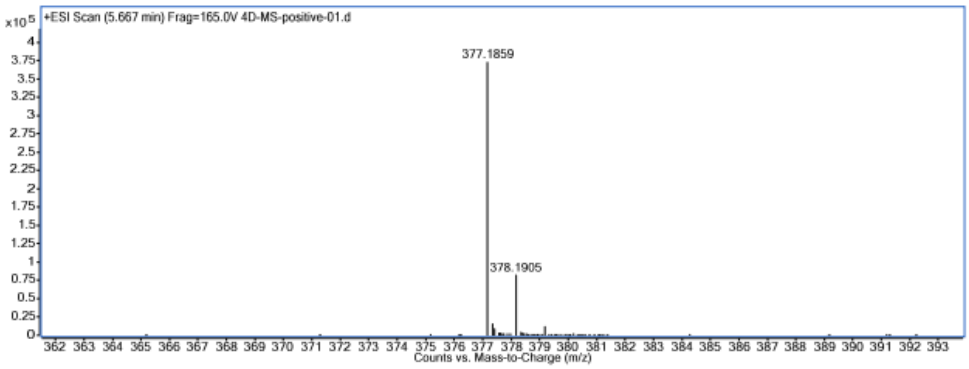


Figure 11. ^{13}C NMR Spectrum of **4d** in $CDCl_3$



Elemental Composition Calculator

Target m/z:	377.1859	**Result type:**	Positive ions	**Species:**	$[M+H]^+$
Elements:		C (0-80); H (0-120); O (0-30); N(0-5)			
Ion Formula		**Calculated m/z**		**PPM Error**	
C23H25N2O3		377.1860		0.18	

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Figure 12. HRMS Spectrum of **4d**

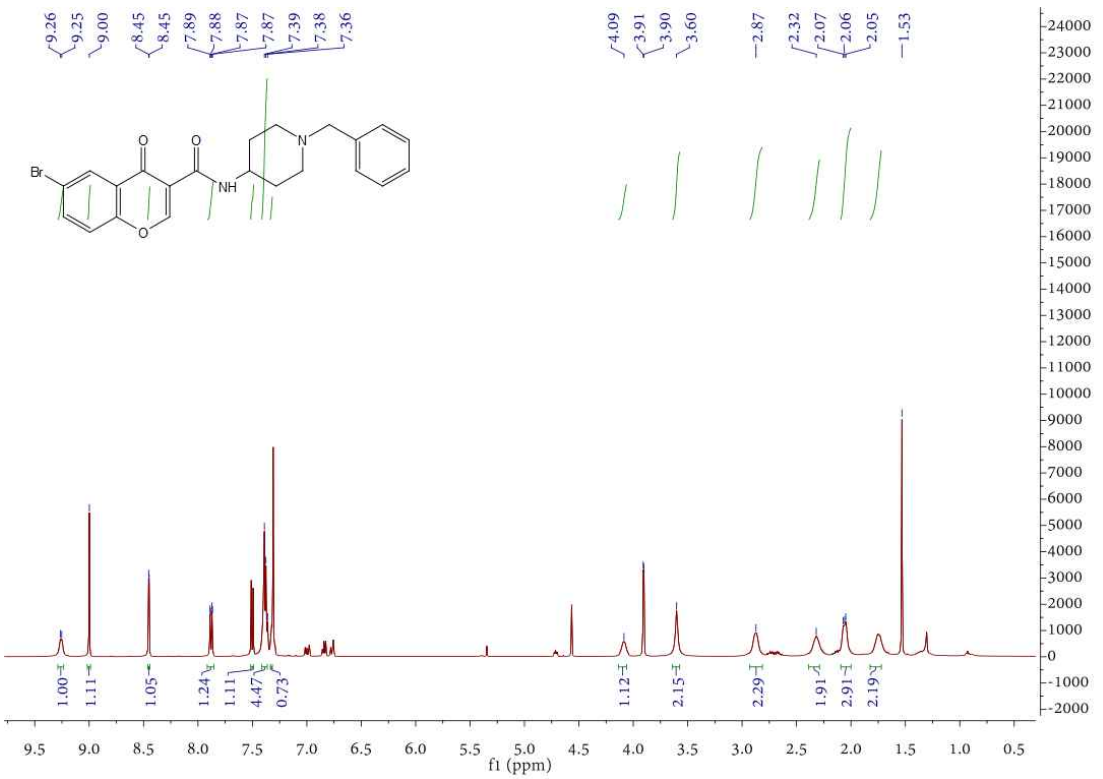


Figure 13. ^{1}H NMR Spectrum of **4e** in $CDCl_3$

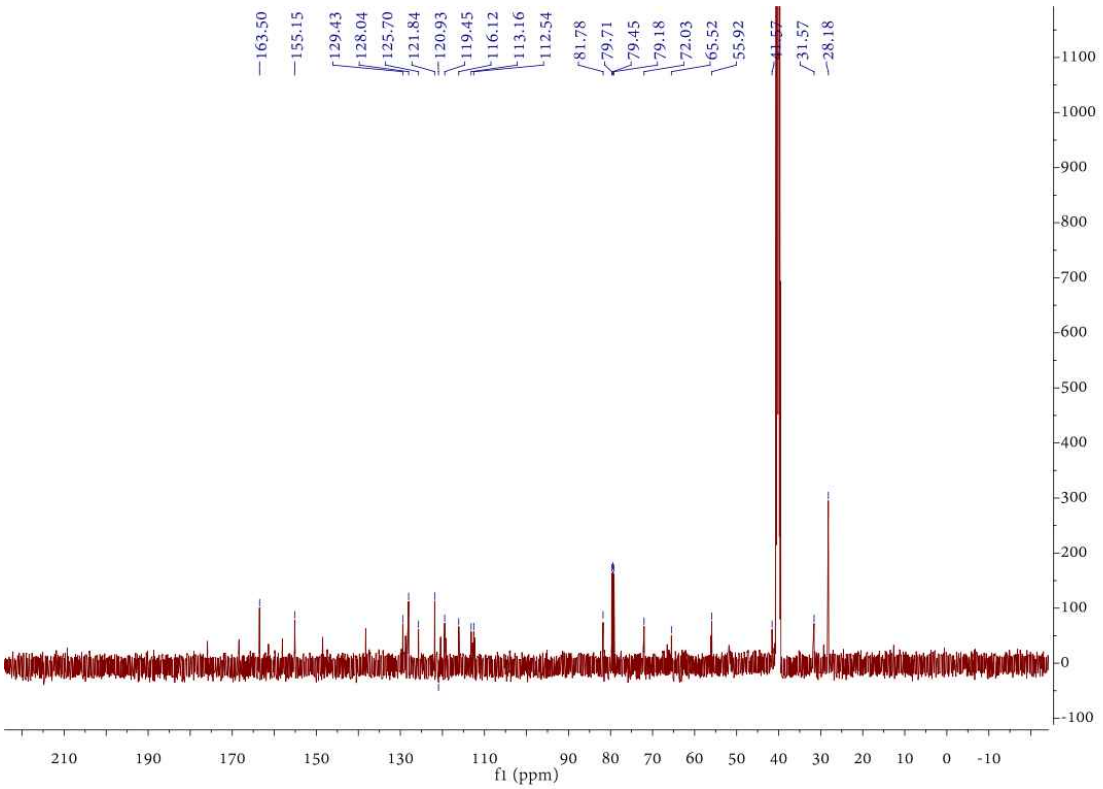
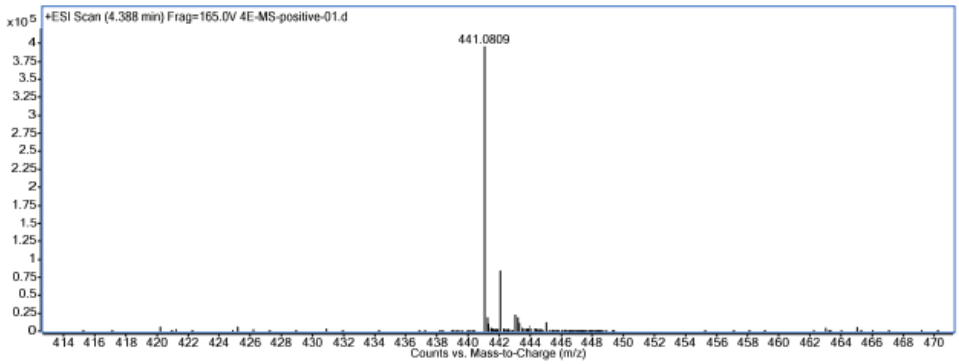


Figure 14. ^{13}C NMR Spectrum of **4e** in $CDCl_3$



Elemental Composition Calculator

Target m/z:	441.0809	**Result type:**	Positive ions	**Species:**	$[M+H]^+$
Elements:		C (0-80); H (0-120); O (0-30); N(0-5); Br(0-5)			
Ion Formula		**Calculated m/z**		**PPM Error**	
C22H22BrN2O3		441.0808		-0.10	

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Figure 15. HRMS Spectrum of **4e**

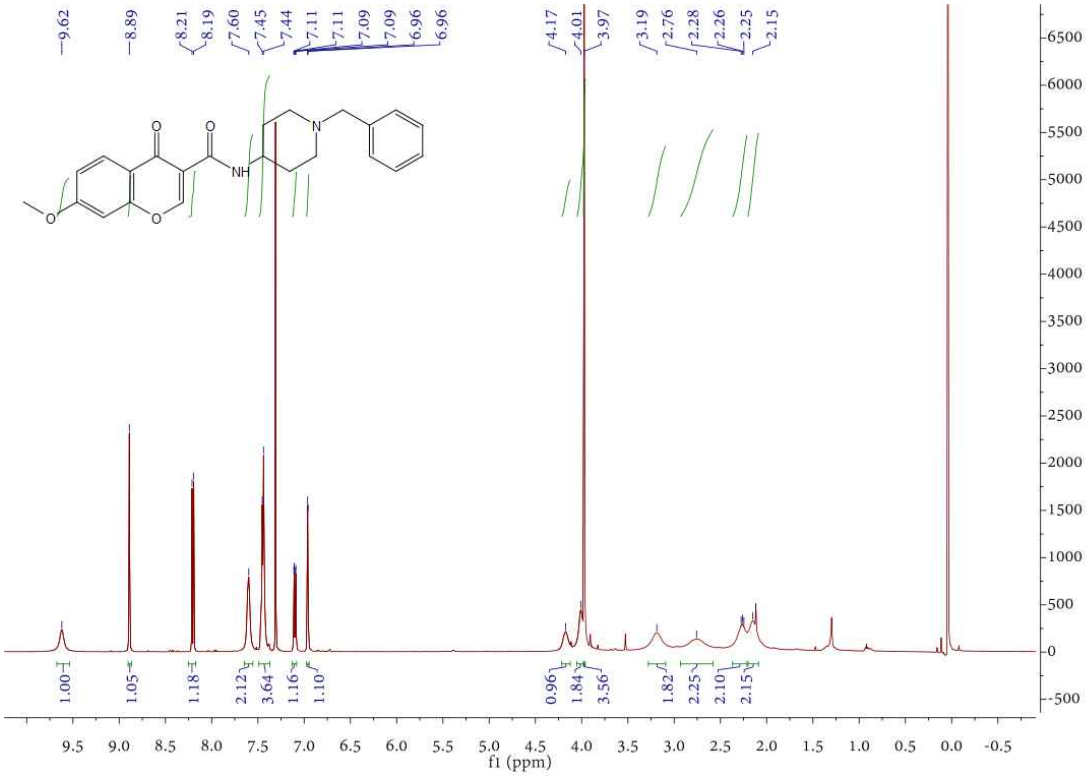


Figure 16. ^{1}H NMR Spectrum of **4f** in $CDCl_3$

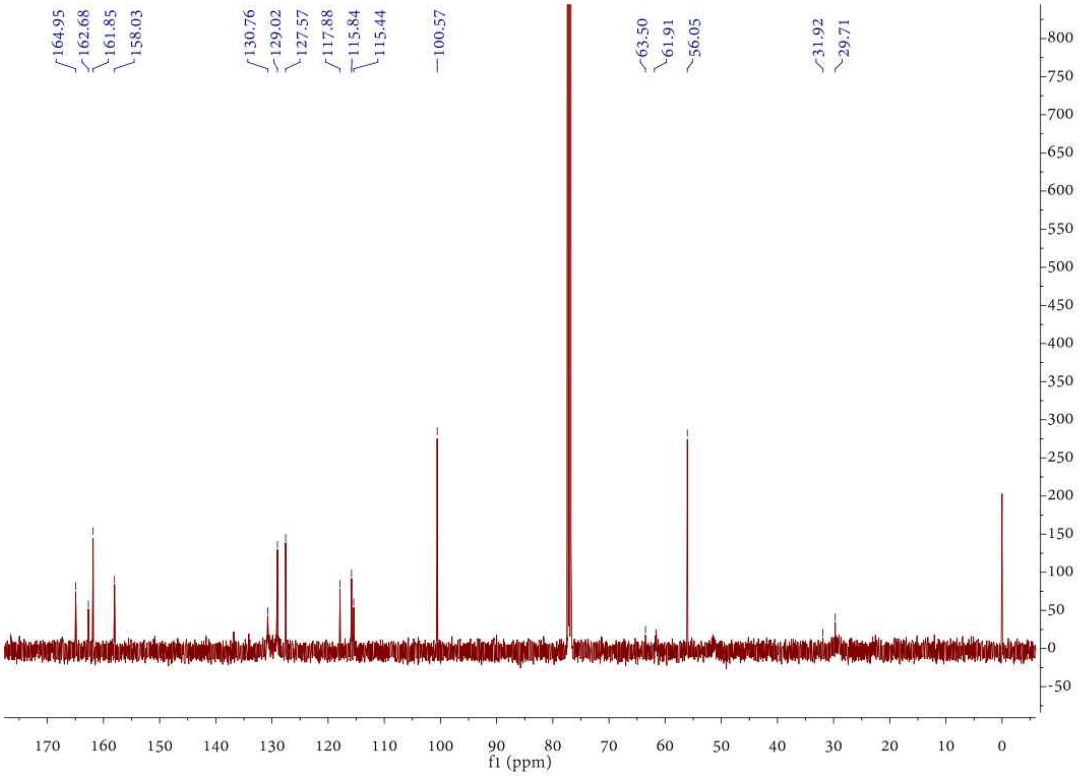
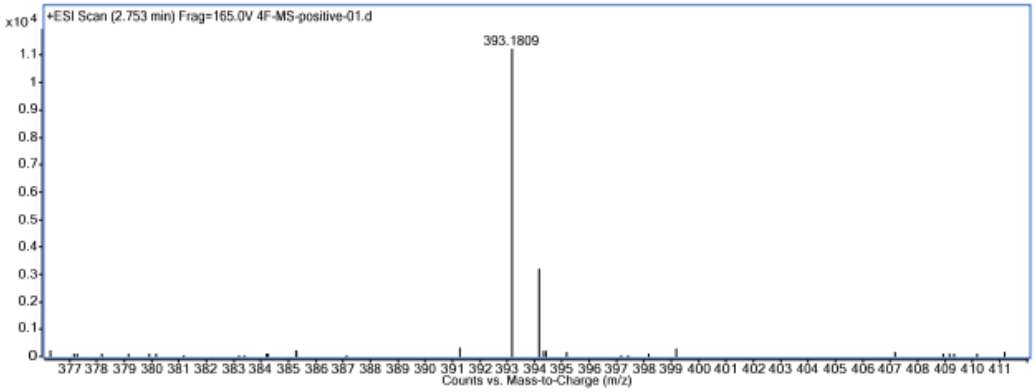


Figure 17 ^{13}C NMR Spectrum of **4f** in $CDCl_3$



Elemental Composition Calculator

Target m/z:	393.1809	**Result type:**	Positive ions	**Species:**	$[M+H]^+$
Elements:		C (0-80); H (0-120); O (0-30); N(0-5)			
Ion Formula		**Calculated m/z**		**PPM Error**	
C23H25N2O4		393.1809		-0.07	

Agilent Technologies

Figure 18. HRMS Spectrum of **4f**

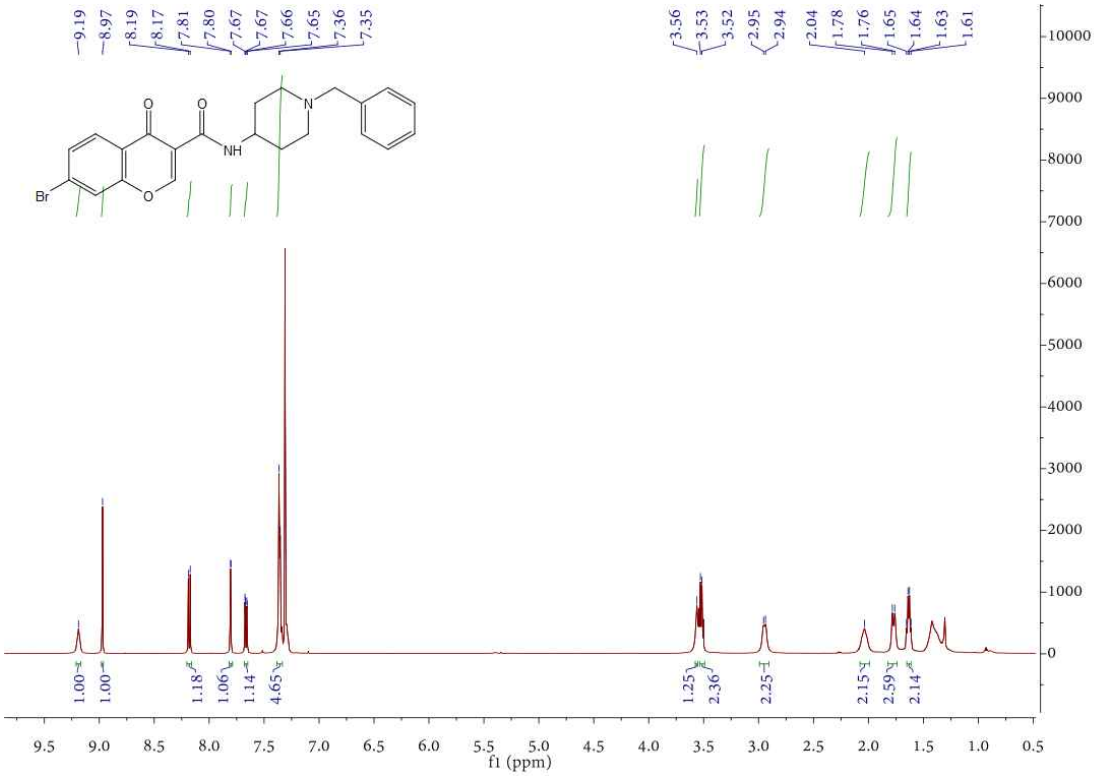


Figure 19. ^{1}H NMR Spectrum of **4g** in $CDCl_3$

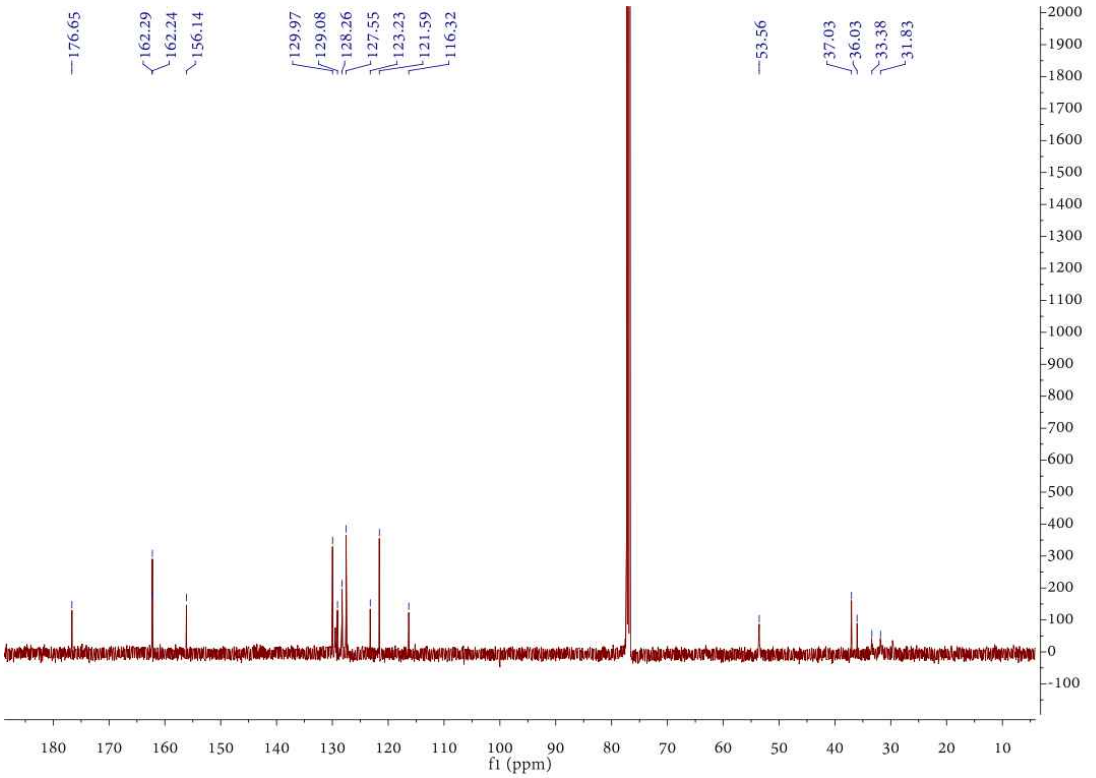
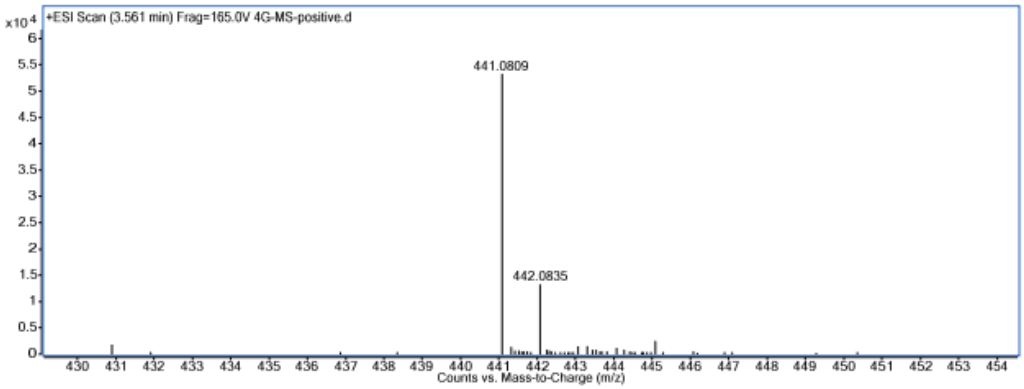


Figure 20. ^{13}C NMR Spectrum of **4g** in $CDCl_3$



Elemental Composition Calculator

Target m/z:	441.0809	**Result type:**	Positive ions	**Species:**	$[M+H]^+$
Elements:		C (0-80); H (0-120); O (0-30); N(0-5); Br(0-5)			
Ion Formula		**Calculated m/z**		**PPM Error**	
C22H22BrN2O3		441.0808		-0.11	

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Figure 21. HRMS Spectrum of **4g**

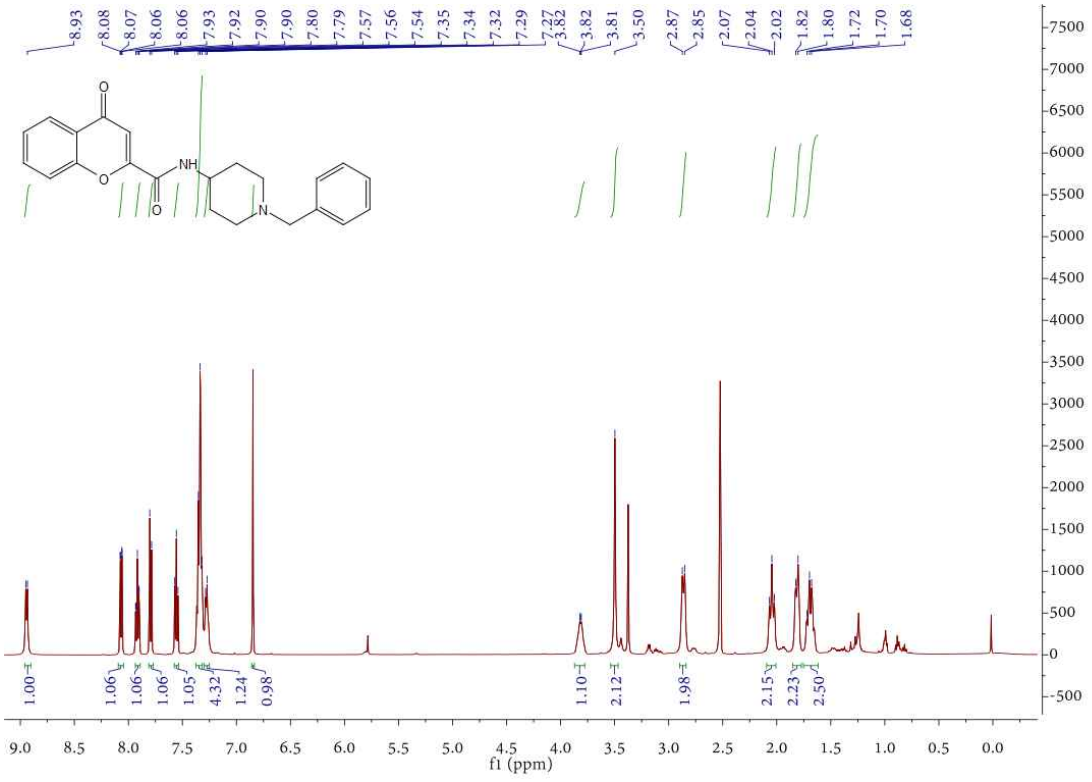


Figure 22. ^{1}H NMR Spectrum of **4h** in DMSO

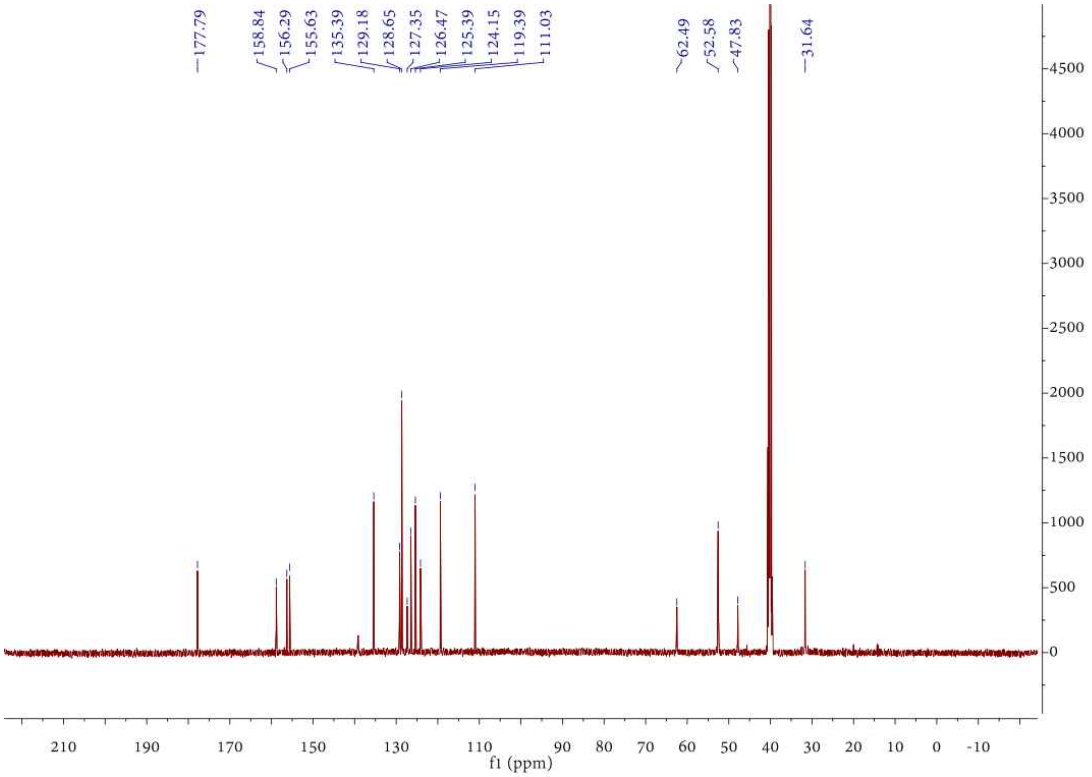
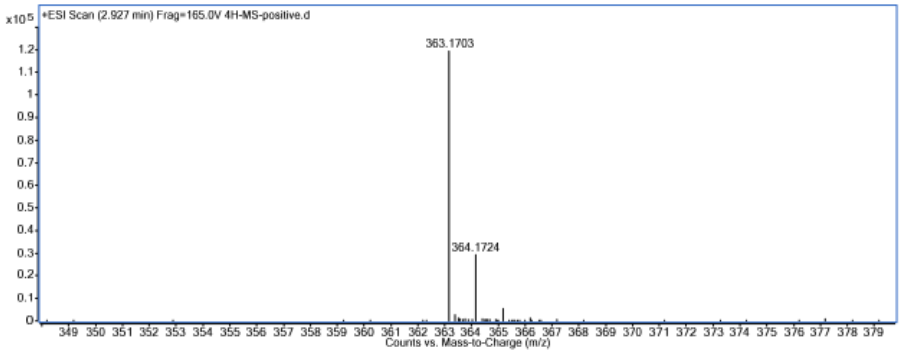


Figure 23. ^{13}C NMR Spectrum of **4h** in DMSO



Elemental Composition Calculator

Target m/z:	363.1703	**Result type:**	Positive ions	**Species:**	$[M+H]^+$
Elements:		C (0-80); H (0-120); O (0-30); N(0-5)			
Ion Formula		**Calculated m/z**		**PPM Error**	
C22H23N2O3		363.1703		0.04	

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Figure 24. HRMS Spectrum of **4h**

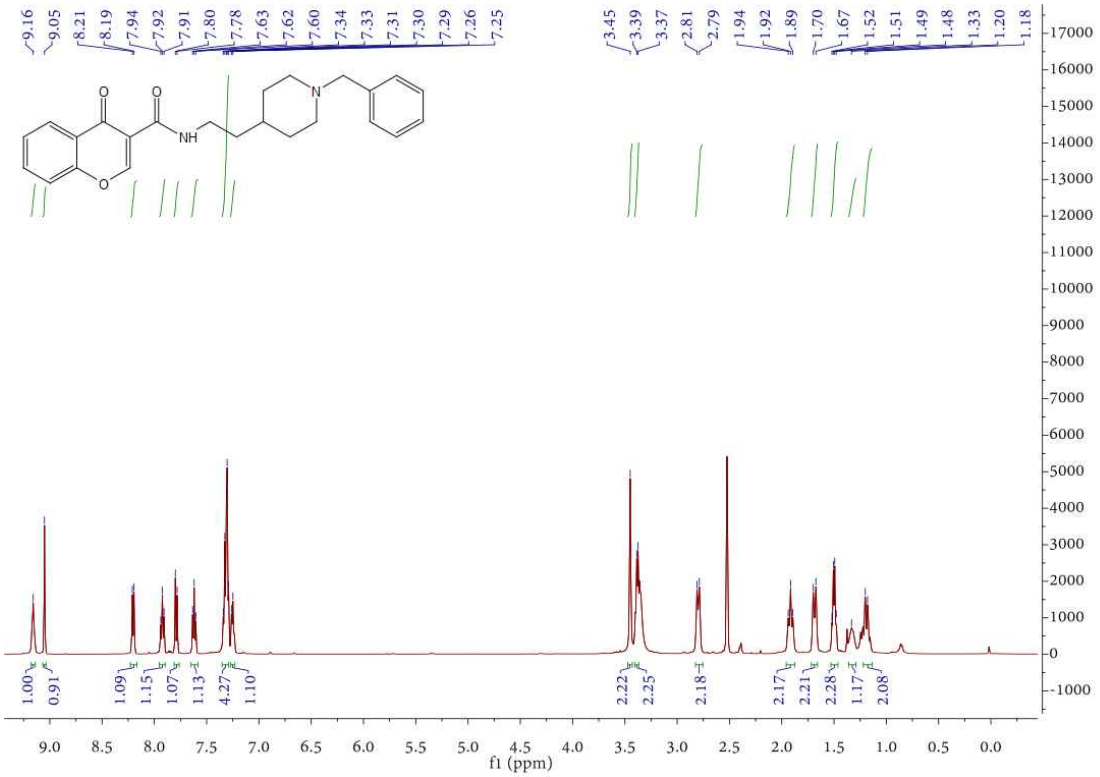


Figure 25. ^{1}H NMR Spectrum of **5a** in DMSO

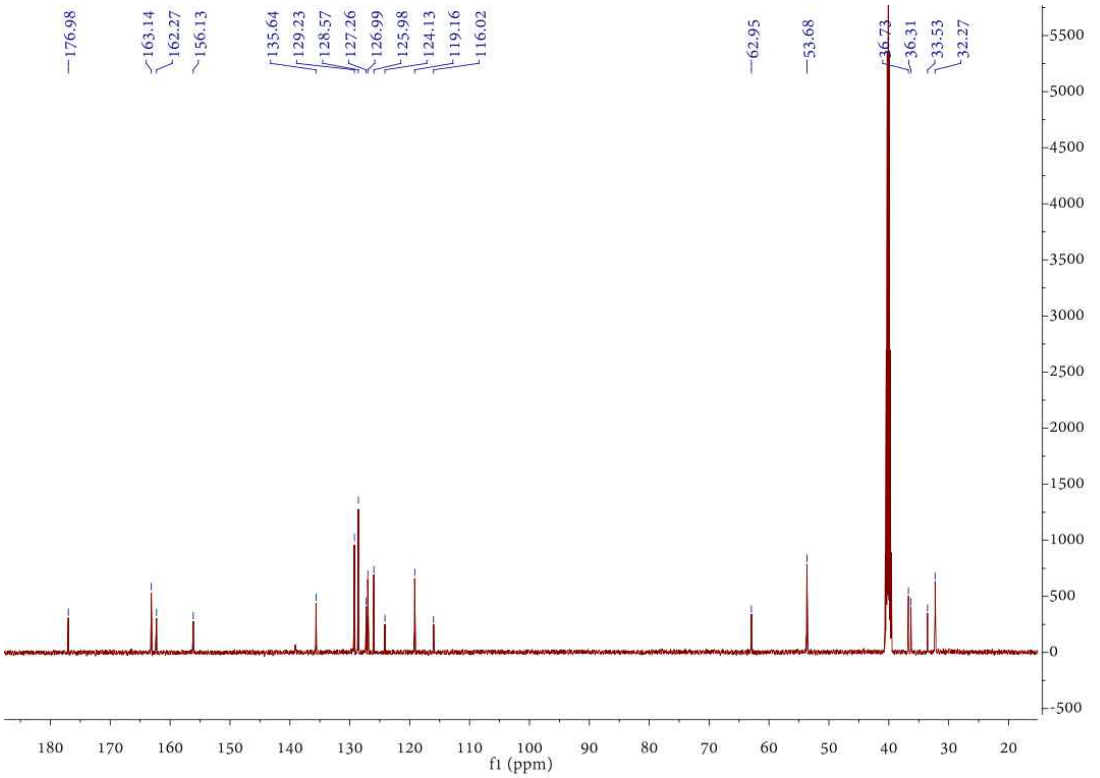
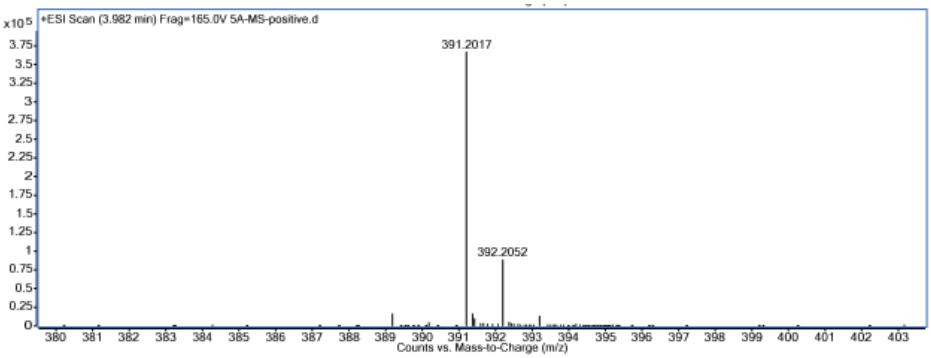


Figure 26. ^{13}C NMR Spectrum of **5a** in DMSO



Elemental Composition Calculator

Target m/z:	391.2017	**Result type:**	Positive ions	**Species:**	$[M+H]^+$
Elements:		C (0-80); H (0-120); O (0-30); N(0-5)			
Ion Formula		**Calculated m/z**		**PPM Error**	
C24H27N2O3		391.2016		-0.23	

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Figure 27 HRMS Spectrum of **5a**

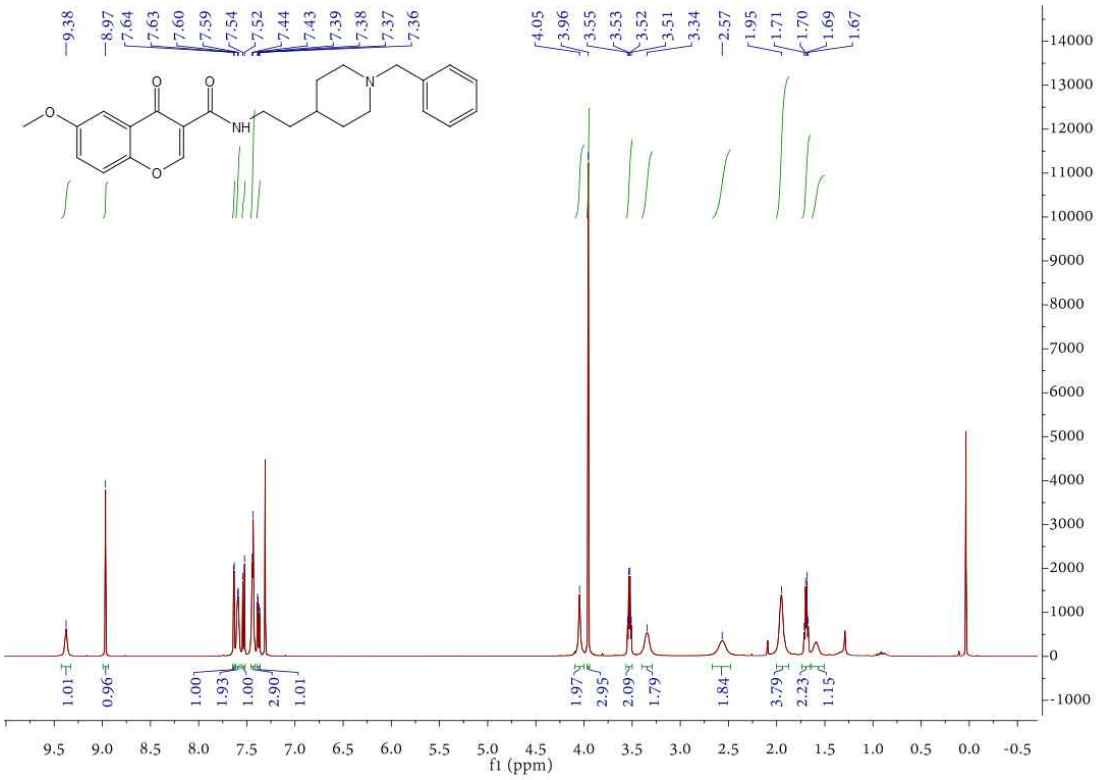


Figure 28. ^{1}H NMR Spectrum of **5b** in $CDCl_3$

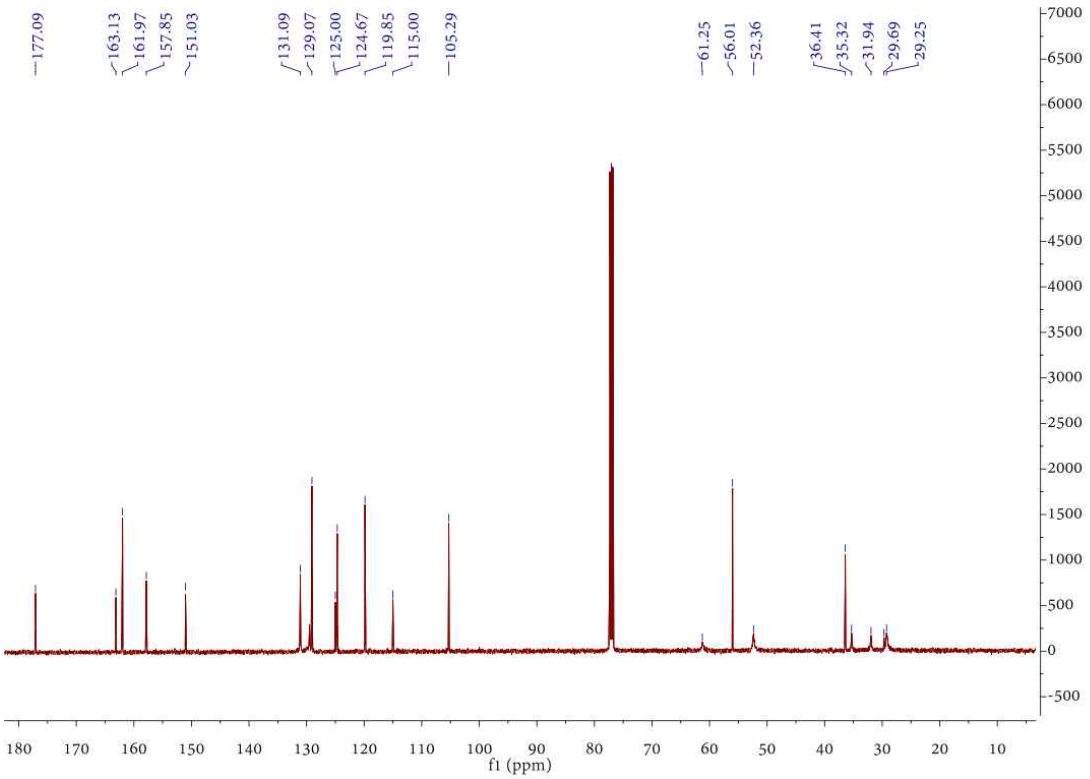
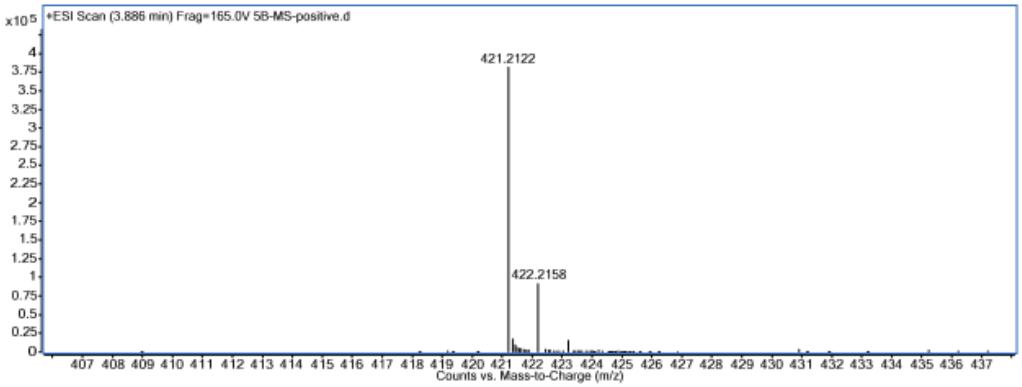


Figure 29. ^{13}C NMR Spectrum of **5b** in $CDCl_3$



Elemental Composition Calculator

Target m/z:	421.2122	**Result type:**	Positive ions	**Species:**	$[M+H]^+$
Elements:		C (0-80); H (0-120); O (0-30); N(0-5)			
Ion Formula		**Calculated m/z**		**PPM Error**	
$C_{25}H_{29}N_2O_4$		421.2122		0.03	

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Figure 30 HRMS Spectrum of **5b**

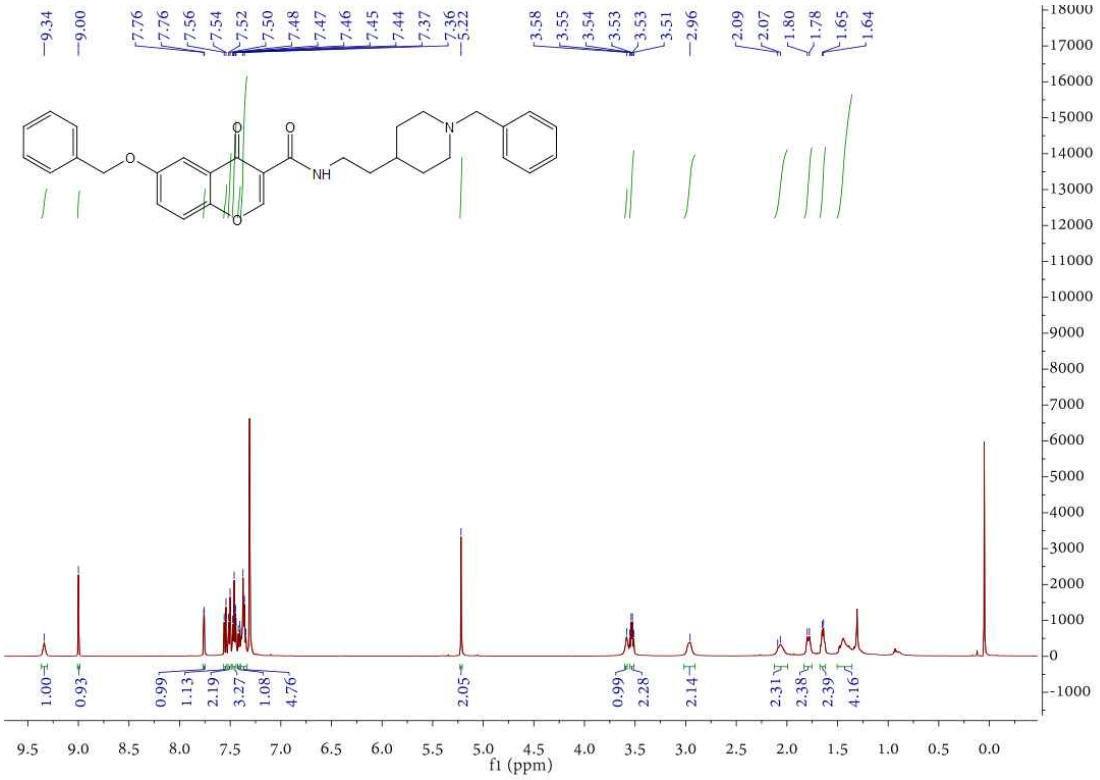


Figure 31. ^{1}H NMR Spectrum of **5c** in $CDCl_3$

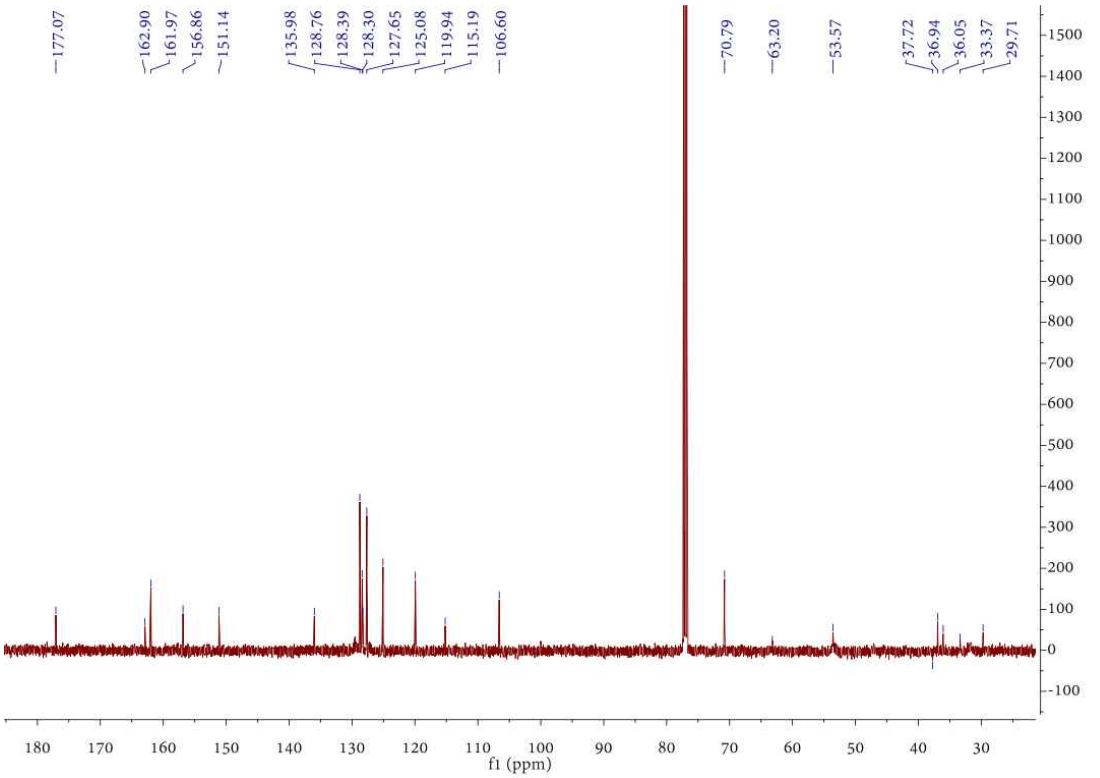
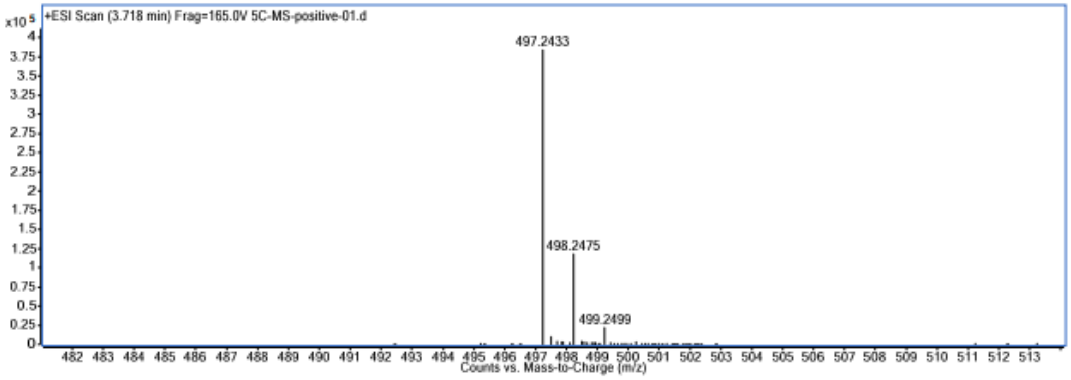


Figure 32. ^{13}C NMR Spectrum of **5c** in $CDCl_3$



Elemental Composition Calculator

Target m/z:	497.2433	**Result type:**	Positive ions	**Species:**	$[M+H]^+$
Elements:		C (0-80); H (0-120); O (0-30); N(0-5)			
Ion Formula		**Calcalated m/z**		**PPM Error**	
C31H33N2O4		497.2435		0.38	

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Figure 33 HRMS Spectrum of **5c**

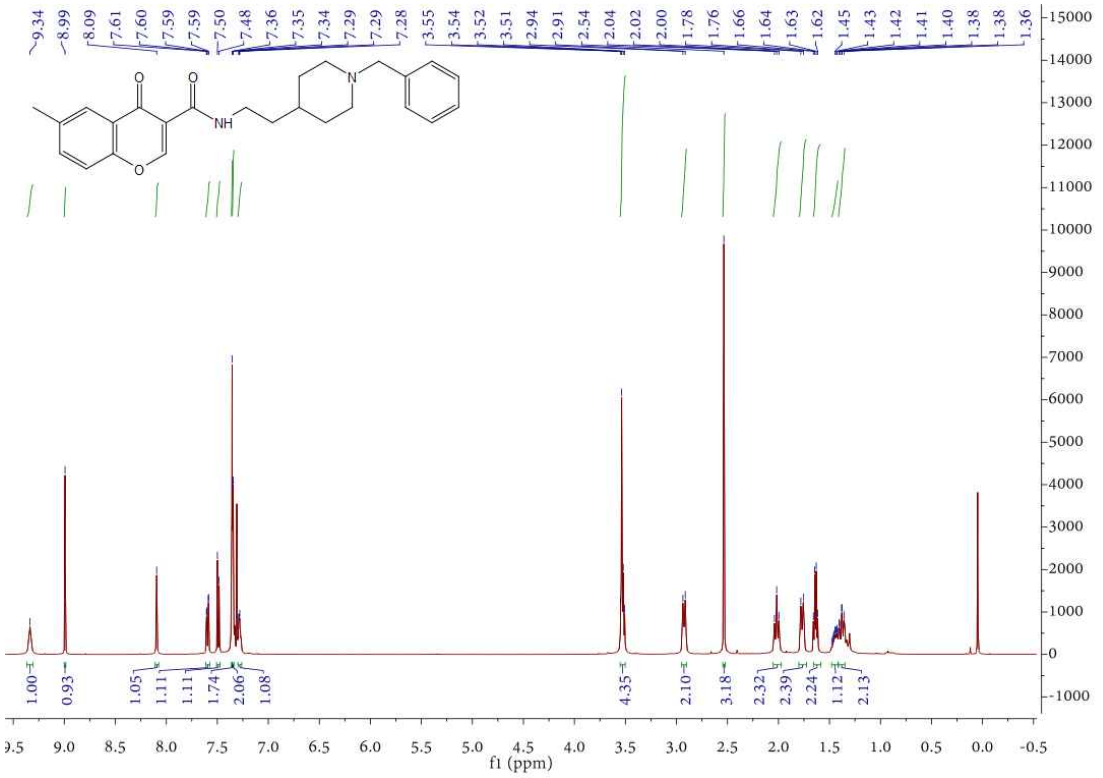


Figure 34. ^{1}H NMR Spectrum of **5d** in $CDCl_3$

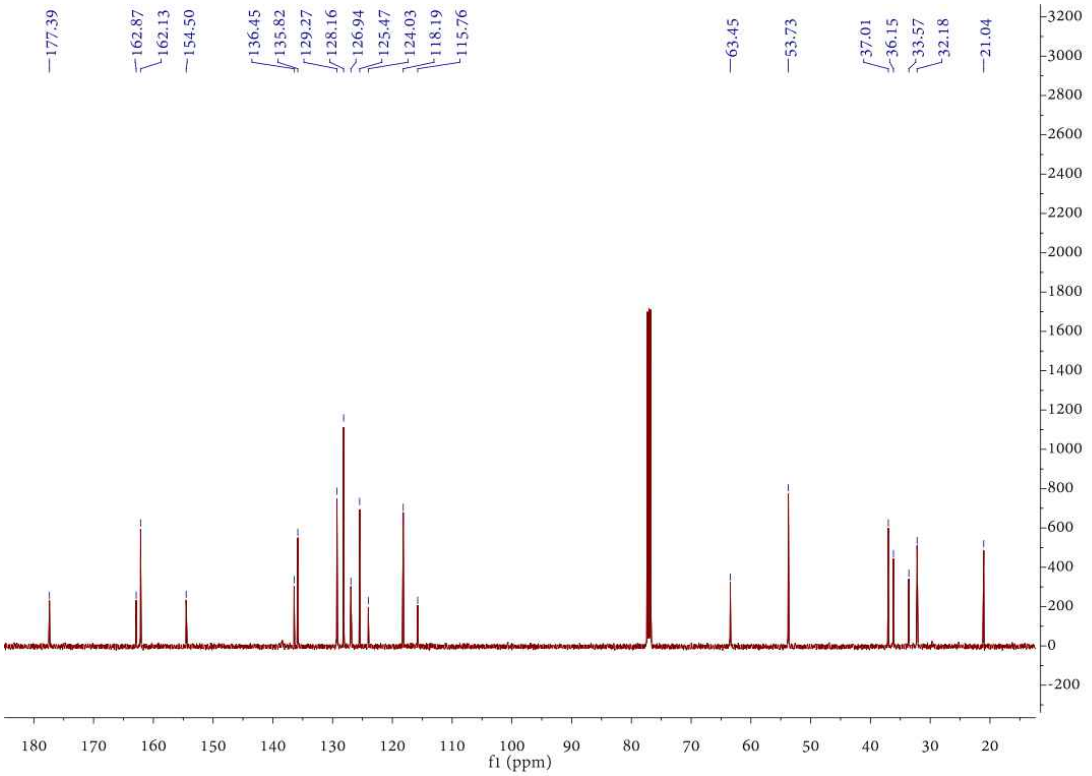
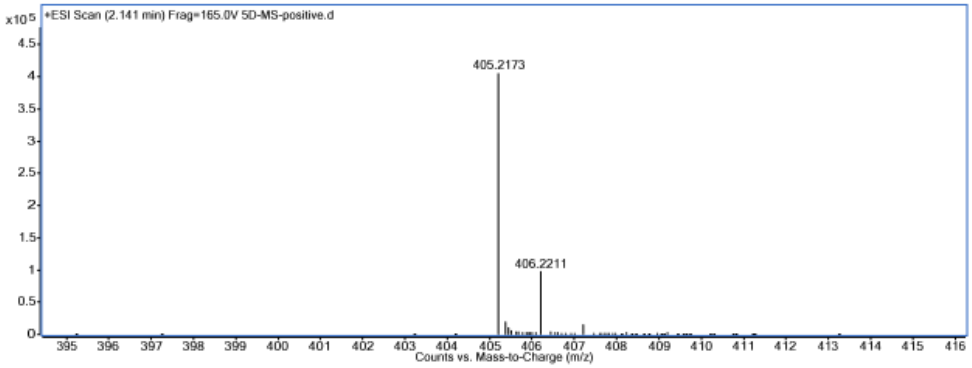


Figure 35. ^{13}C NMR Spectrum of **5d** in $CDCl_3$



Elemental Composition Calculator

Target m/z:	405.2173	**Result type:**	Positive ions	**Species:**	$[M+H]^+$
Elements:		C (0-80); H (0-120); O (0-30); N(0-5)			
Ion Formula		**Calculated m/z**		**PPM Error**	
C25H29N2O3		405.2173		-0.19	

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Figure 36 HRMS Spectrum of **5d**

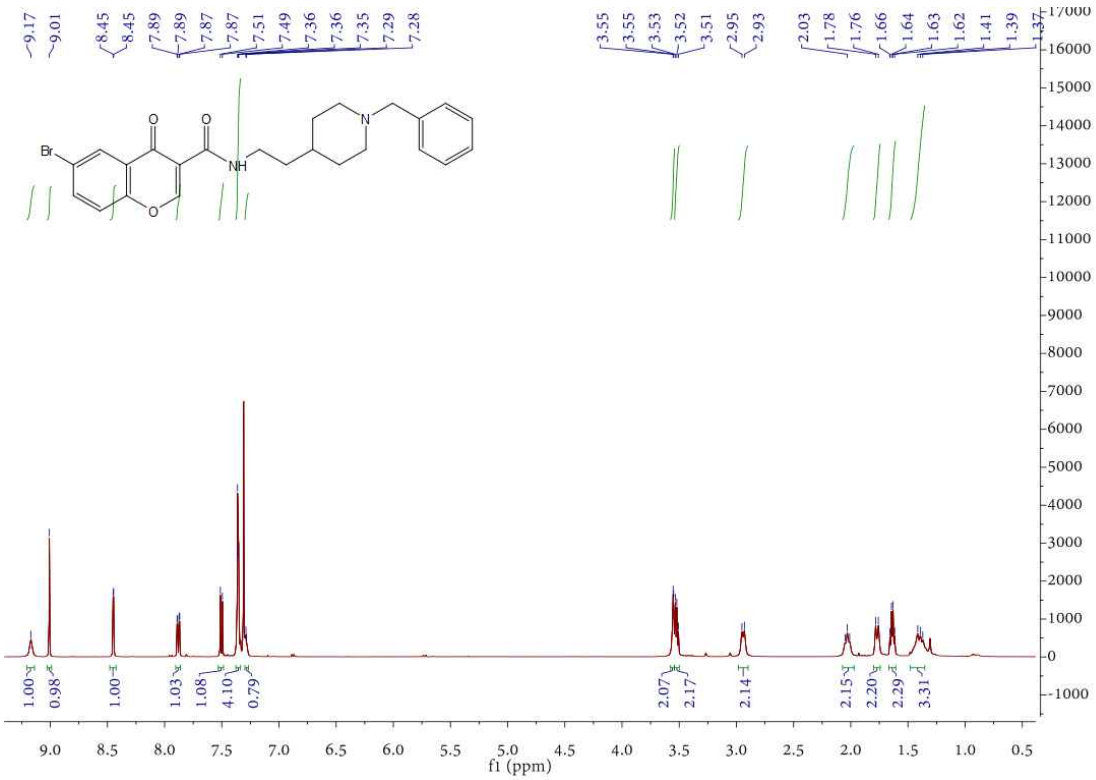


Figure 37. ^{1}H NMR Spectrum of **5e** in $CDCl_3$

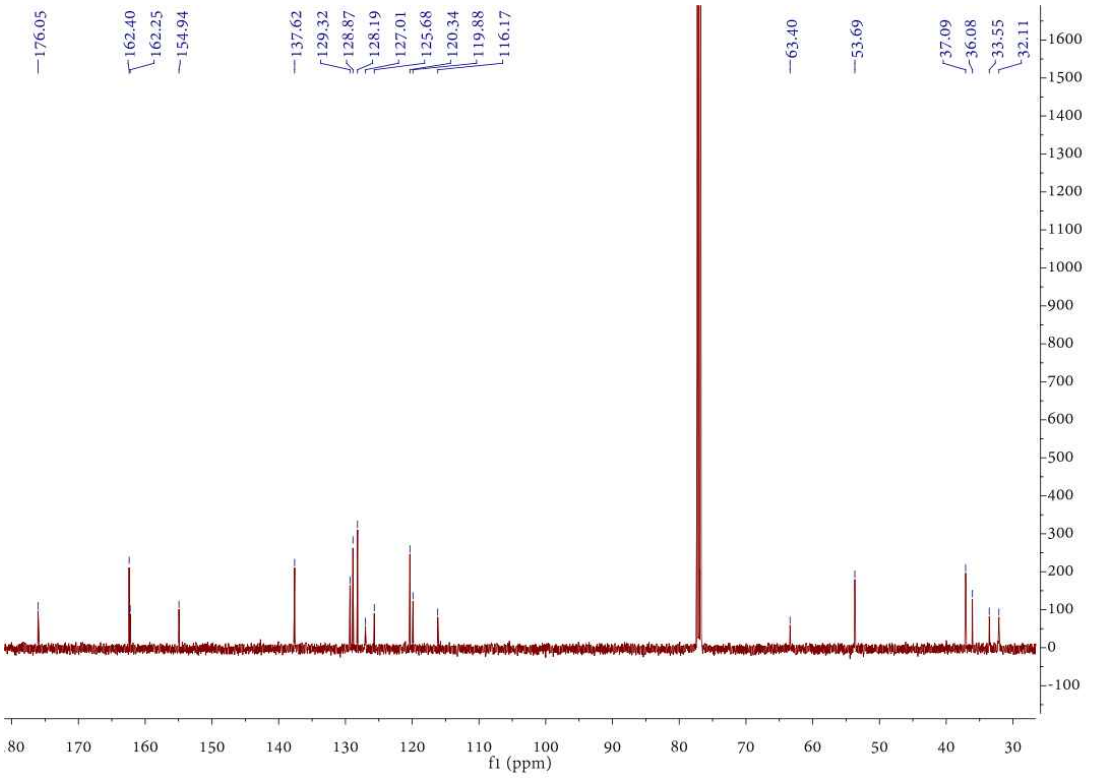
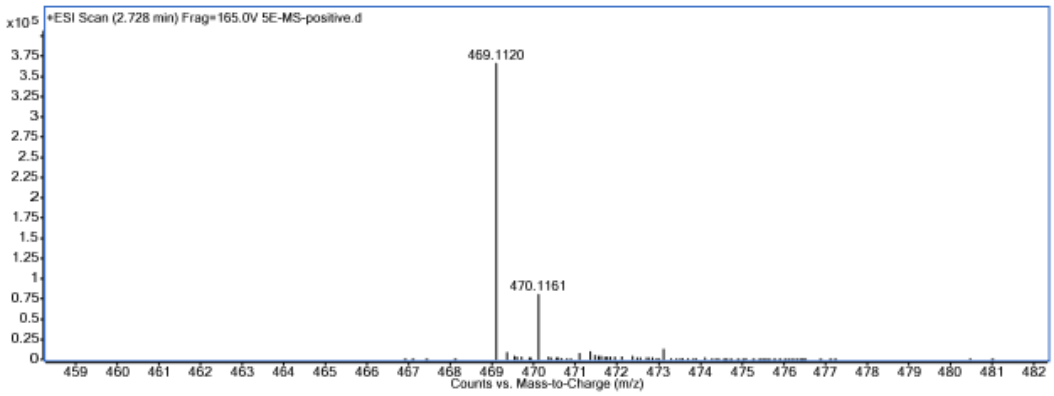


Figure 38. ^{13}C NMR Spectrum of **5e** in $CDCl_3$



Elemental Composition Calculator

Target m/z:	469.1120	**Result type:**	Positive ions	**Species:**	$[M+H]^+$
Elements:		C (0-80); H (0-120); O (0-30); N (0-5); Br (0-5)			
Ion Formula		**Calculated m/z**		**PPM Error**	
C24H26BrN2O3		469.1121		0.36	

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Figure 39 HRMS Spectrum of **5e**

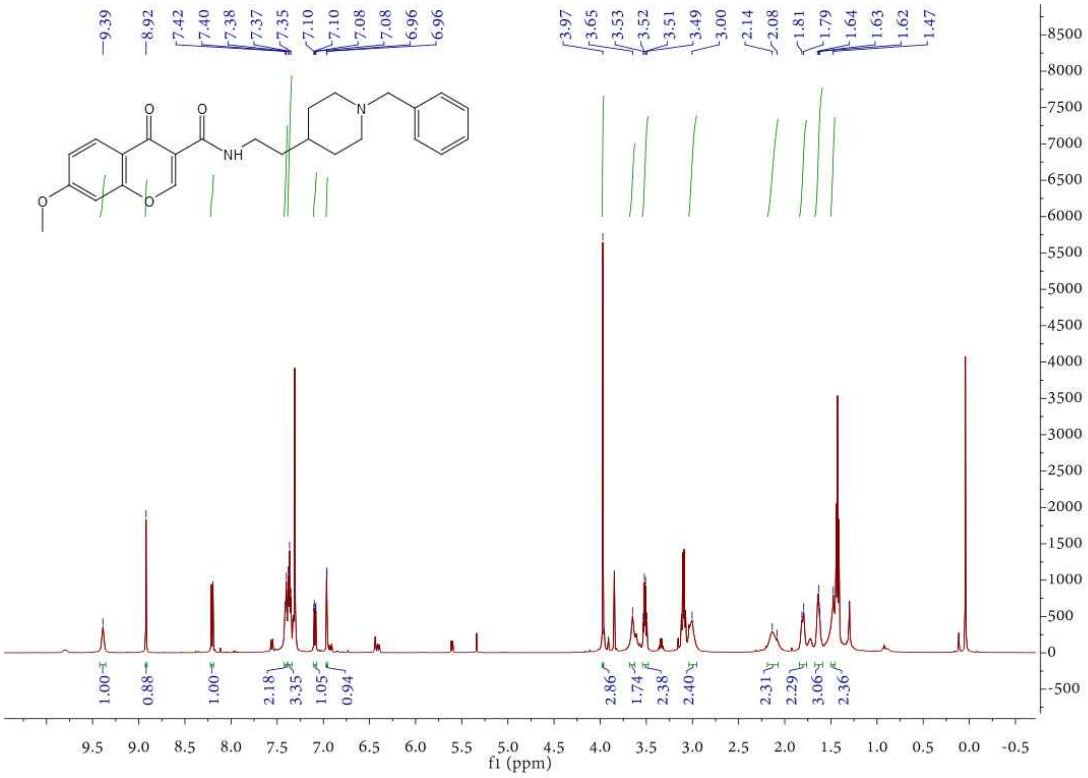


Figure 40. ^{1}H NMR Spectrum of **5f** in $CDCl_3$

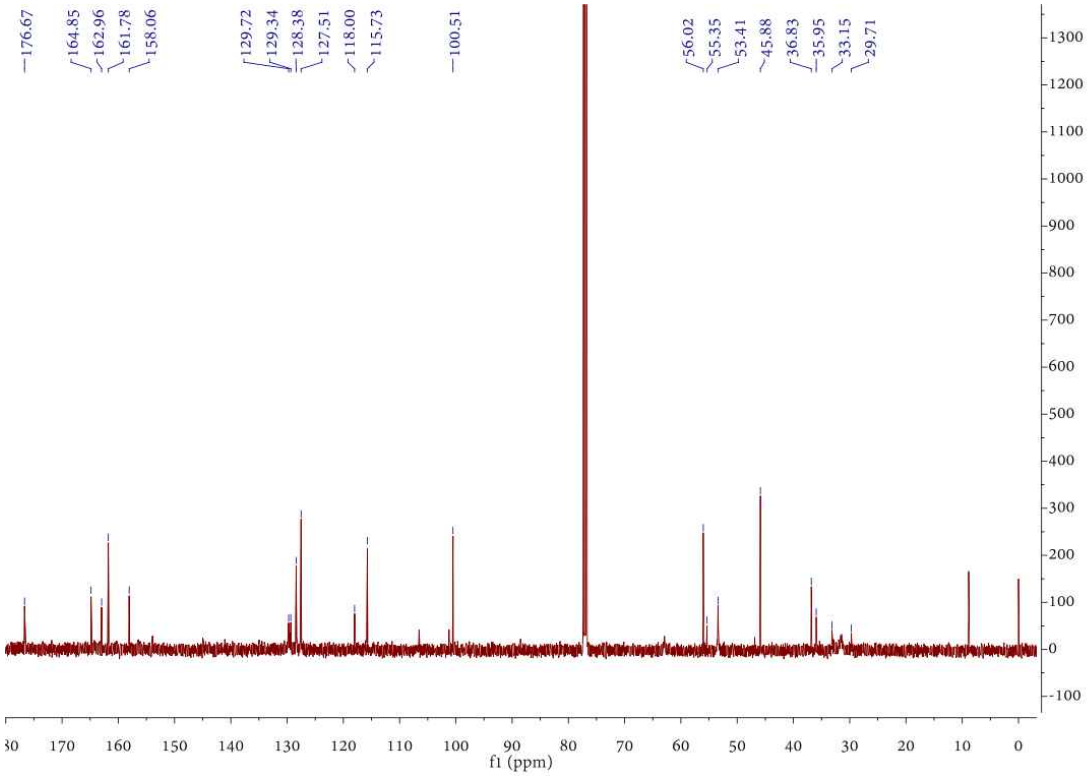
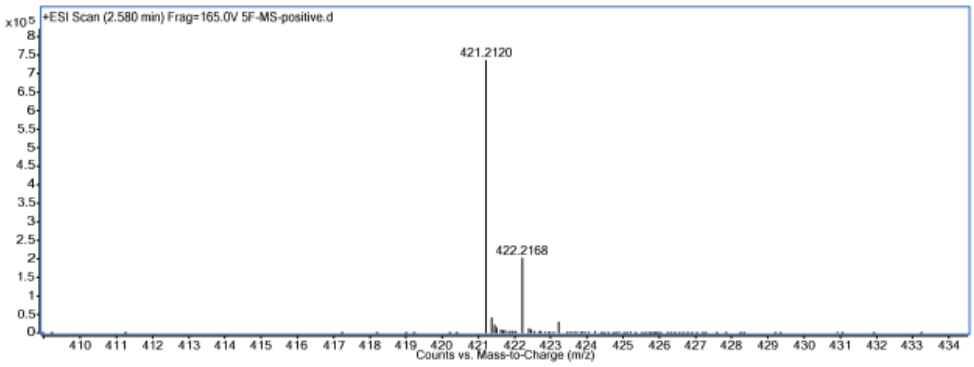


Figure 41. ^{13}C NMR Spectrum of **5f** in $CDCl_3$



Elemental Composition Calculator

Target m/z:	421.2120	**Result type:**	Positive ions	**Species:**	$[M+H]^+$
Elements:		C (0-80); H (0-120); O (0-30); N (0-5)			
Ion Formula		**Calculated m/z**		**PPM Error**	
C25H29N2O4		421.2122		0.32	

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Figure 42 HRMS Spectrum of **5f**

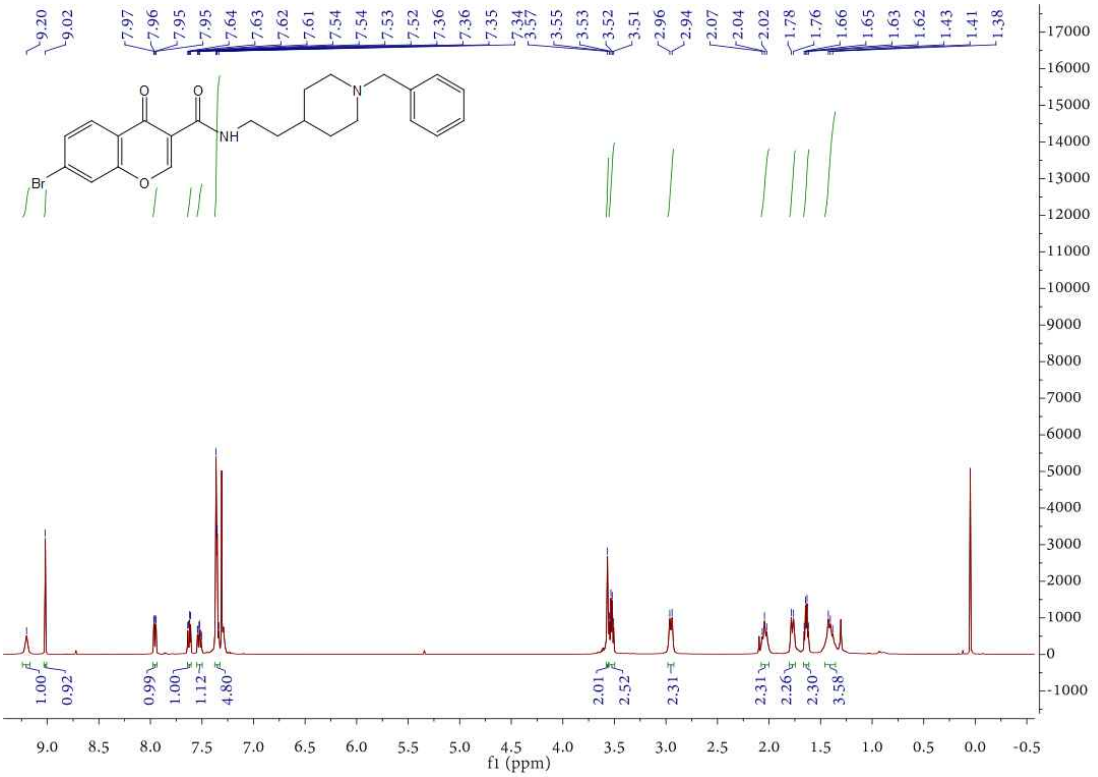


Figure 43. ^{1}H NMR Spectrum of **5g** in $CDCl_3$

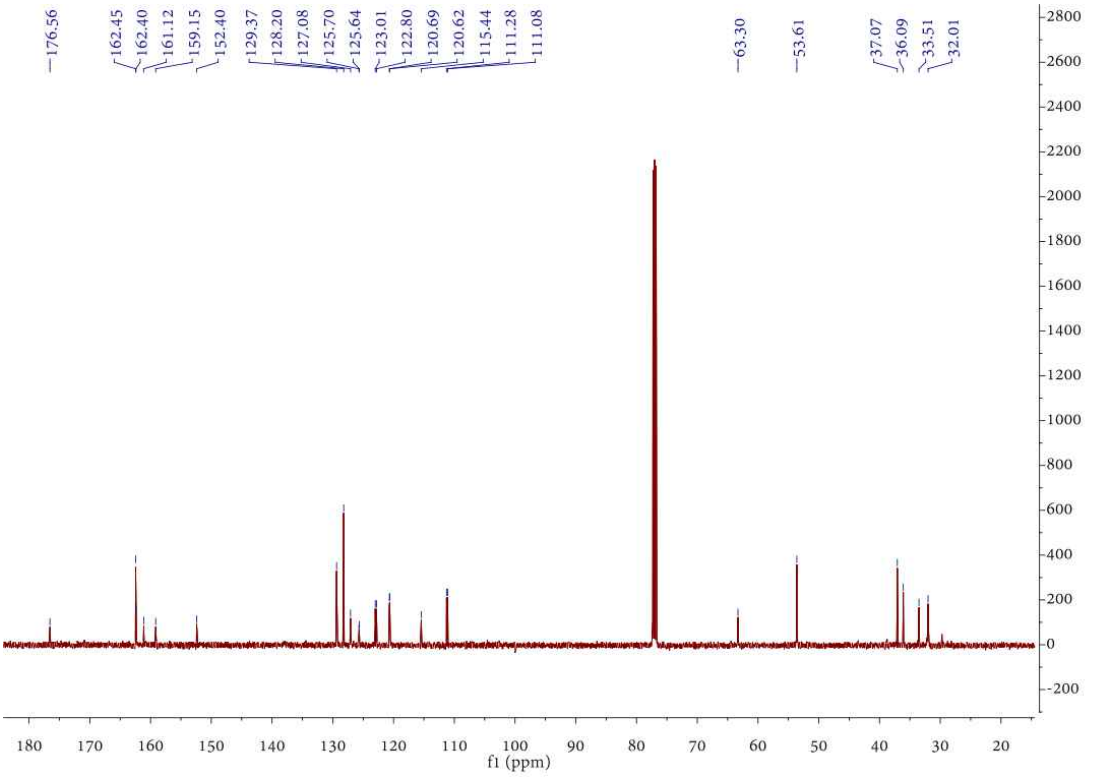
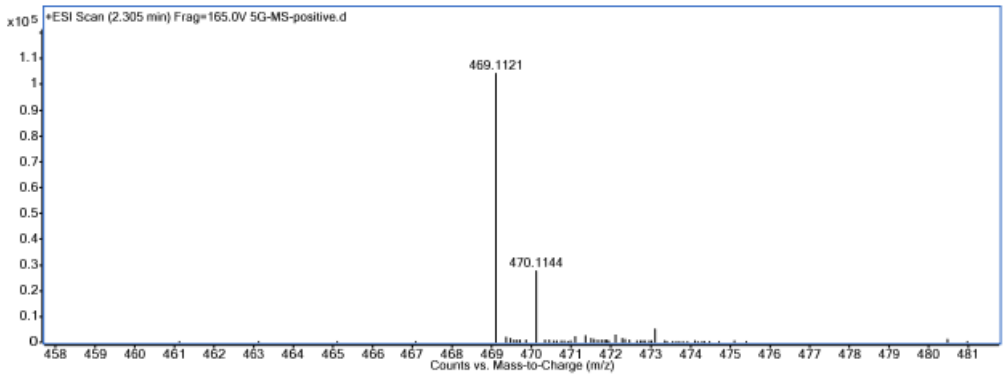


Figure 44. ^{13}C NMR Spectrum of **5g** in $CDCl_3$



Elemental Composition Calculator

Target m/z:	469.1121	**Result type:**	Positive ions	**Species:**	$[M+H]^+$
Elements:		C (0-80); H (0-120); O (0-30); N (0-5); Br (0-5)			
Ion Formula		**Calculated m/z**		**PPM Error**	
C24H26BrN2O3		469.1121		0.16	

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Figure 45 HRMS Spectrum of **5g**

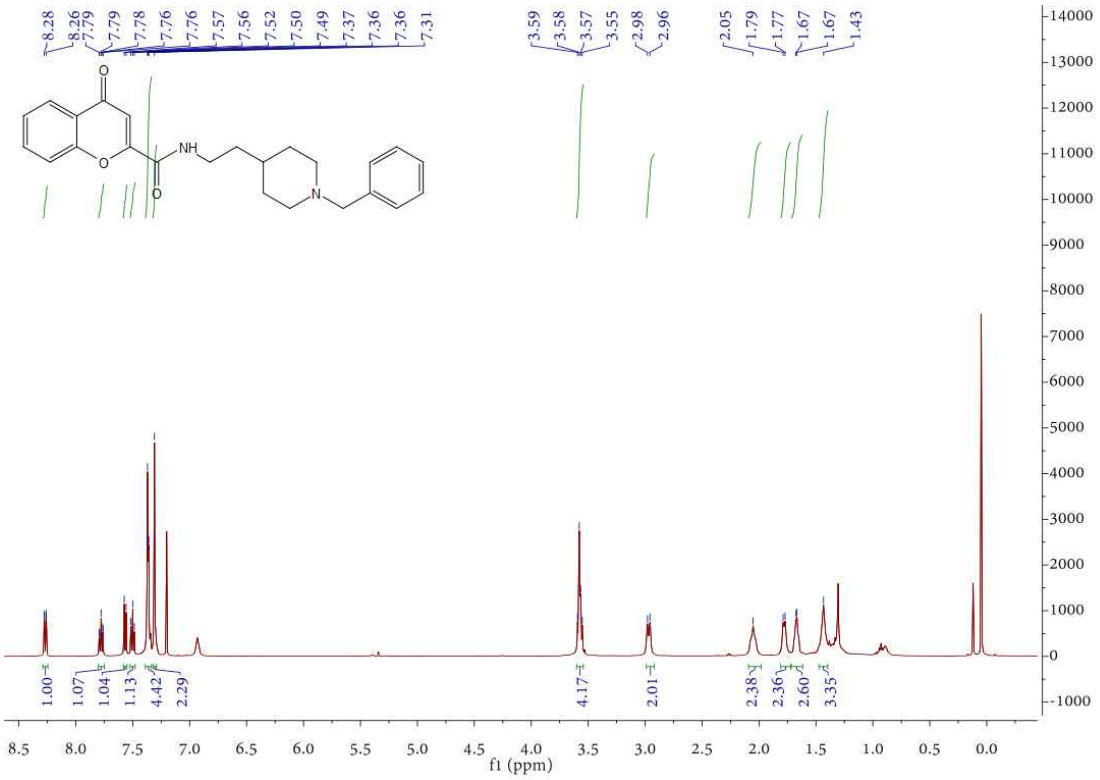


Figure 46. ^{1}H NMR Spectrum of **5h** in $CDCl_3$

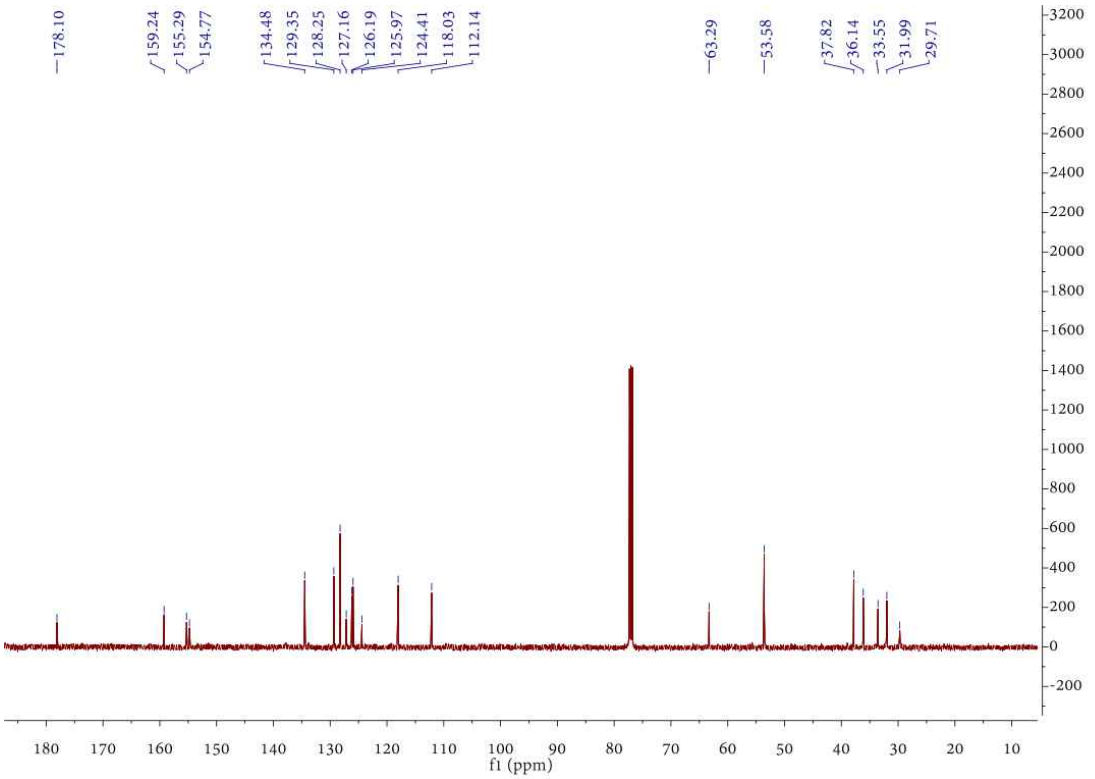
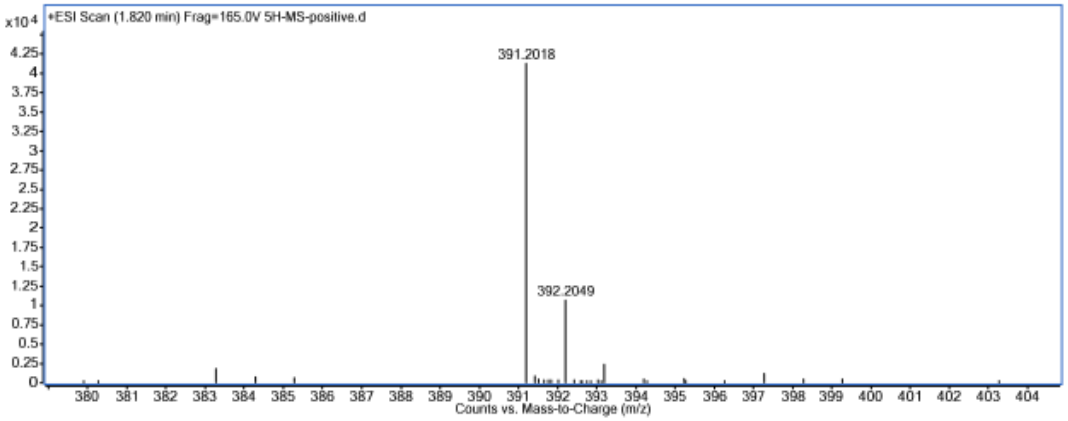


Figure 47. ^{13}C NMR Spectrum of **5h** in $CDCl_3$



Elemental Composition Calculator

Target m/z:	391.2018	**Result type:**	Positive ions	**Species:**	$[M+H]^+$
Elements:		C (0-80); H (0-120); O (0-30); N (0-5)			
Ion Formula		**Calculated m/z**		**PPM Error**	
C24H27N2O3		391.2016		-0.55	

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Figure 48 HRMS Spectrum of **5h**