Supplementary Information

Synthesis and evaluation of the antibacterial activities of 9-substituted berberine derivatives

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1. Experimental section

1.1. Chemistry

Unless otherwise stated, all chemicals and solvents were obtained from commercial sources and used without any additional purification. The reactions were monitored via thin-layer chromatography (0.20 mm silica gel 60 F-254 glass plates, Qingdao Haiyang Chemical, China), with visualization achieved under UV light and/or I_2 in silica gel. Column chromatography was performed on silica gel (300-400 mesh, Qingdao Haiyang Chemical, China). ^{1}H NMR and ^{13}C NMR spectra (solvent DMSO-d_6 or Methanol-d_4) were recorded on a Zhongke-Niujin 400 NMR spectrometer at room temperature using tetramethylsilane (TMS) as an internal standard. The melting points of all products were determined using a Fisher-Johns melting apparatus and were left uncorrected. Mass spectrometry data were collected using an Agilent 1260 infinity electrospray ionization-mass spectrometry (ESI-MS). The purity of all final compounds was greater than 95%, as determined using high-performance liquid chromatography (HPLC).

*1.2. General procedure for the synthesis of intermediate **1** and **2***

Berberine hydrochloride (5 mmol) was placed in a round-bottom flask and stirred under reduced pressure at 200 °C for 30 min. After the reaction, the mixture was cooled to room temperature, dissolved by adding an appropriate amount of absolute ethanol, concentrated by vacuum rotary evaporator, and purified by column chromatography to obtain intermediate **1**. The synthesis method of intermediate **2** was the same as above, and the raw material Berberine can be replaced by Palmatine.

*1.2.1. 9-hydroxy-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**1**).* Red solid, 85% yield; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.09 (s, 1H), 8.00 (s, 1H), 7.60 (s, 1H), 7.23 (d, J = 7.9 Hz, 1H), 6.95 (s, 1H), 6.39 (d, J = 7.8 Hz, 1H), 6.09 (s, 2H), 4.48 (t, J = 5.9 Hz, 2H), 3.73 (s, 3H), 3.03 (t, J = 5.9 Hz, 2H).

*1.2.2. 9-hydroxy-2,3,10-trimethoxyprotopalmatine chloride (**2**).* Red solid, 86% yield; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.10 (s, 1H), 8.07 (s, 1H), 7.51 (s, 1H), 7.23 (d, J = 7.9 Hz, 1H), 6.97 (s, 1H), 6.39 (d, J = 7.8 Hz, 1H), 4.50 (t, J = 6.0 Hz, 2H), 3.89 (s, 3H), 3.82 (s, 3H), 3.73 (s, 3H), 3.06 (t, J = 6.0 Hz, 2H).

*1.3. General procedure for the synthesis of compounds **B1**–**B4***

The reaction bottle containing intermediate **1** (0.56 mmol) was dissolved by adding 10 mL acetonitrile, followed by adding K_2CO_3 (1.68 mmol) and halocarbons (0.84 mmol), stirring at 60 ℃ for 6-8 h, and TLC monitoring (CH_2Cl_2: MeOH=10:1). After the reaction, acetonitrile was removed under vacuum, and the residues were purified by column chromatography (eluent: dichloromethane/methanol) to obtain the target products **B1**–**B4**.

*1.3.1. 9-(3-chloropropoxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium bromide (**B1**).* Yellow solid, 68% yield, m.p: 236.2–237.1 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.82 (s, 1H), 8.97 (s, 1H), 8.20 (d, J = 9.2 Hz, 1H), 8.00 (d, J = 9.1 Hz, 1H), 7.80 (s, 1H), 7.09 (s, 1H), 6.17 (s, 2H), 4.96 (t, J = 6.1 Hz, 2H), 4.42 (t, J = 6.1 Hz, 2H), 4.06 (s, 3H), 3.95 (t, J = 6.5 Hz, 2H), 3.21 (t, J = 6.1 Hz, 2H), 2.38 – 2.30 (m, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 150.75,

150.32, 148.17, 145.74, 142.96, 137.99, 133.51, 131.19, 127.15, 124.02, 121.96, 120.93, 120.72, 108.91, 105.94, 102.59, 71.70, 57.59, 55.75, 42.67, 33.11, 26.81. ESI-MS *m/z*: 398.2 $[M–Br]^+$. HPLC purity: 95.91%.

*1.3.2. 9-((5-bromopentyl)oxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium bromide (**B2**).* Yellow solid, 61% yield, m.p: 218.7–219.5 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.77 (s, 1H), 8.95 (s, 1H), 8.16 (d, J = 9.1 Hz, 1H), 7.98 (d, J = 9.1 Hz, 1H), 7.76 (s, 1H), 7.07 (s, 1H), 6.16 (s, 2H), 4.97 (t, J = 5.8 Hz, 2H), 4.28 (t, J = 6.5 Hz, 2H), 4.04 (s, 3H), 3.70 (t, J = 6.5 Hz, 1H), 3.60 (t, J = 6.6 Hz, 1H), 3.21 (t, J = 5.4 Hz, 2H), 1.96 – 1.79 (m, 4H), 1.68 – 1.57 (m, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 150.87, 150.25, 148.11, 145.74, 143.24, 137.82, 133.46, 131.10, 127.02, 123.84, 122.08, 120.99, 120.68, 108.88, 105.90, 102.58, 74.50, 57.51, 55.80, 45.92, 32.28, 29.21, 26.80, 23.24. ESI-MS *m/z*: 470.1 $[M–Br]^+$. HPLC purity: 97.31%.

*1.3.3. 10-methoxy-9-(3-methoxypropoxy)-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium bromide (**B3**).* Yellow solid, 62% yield, m.p: 228.8–229.3 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.79 (s, 1H), 8.97 (s, 1H), 8.19 (d, J = 9.1 Hz, 1H), 8.00 (d, J = 9.1 Hz, 1H), 7.80 (s, 1H), 7.09 (s, 1H), 6.17 (s, 2H), 4.96 (t, J = 6.0 Hz, 2H), 4.35 (t, J = 6.6 Hz, 2H), 4.05 (s, 3H), 3.57 (t, J = 6.2 Hz, 2H), 3.29 (s, 3H), 3.21 (t, J = 5.4 Hz, 2H), 2.12 (p, J = 6.4 Hz, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 150.88, 150.30, 148.17, 145.82, 143.24, 137.96, 133.51, 131.18, 127.16, 123.91, 122.12, 120.96, 120.75, 108.91, 105.95, 102.58, 72.31, 69.20, 58.53, 57.55, 55.79,

30.25, 26.83. ESI-MS *m/z*: 394.2 $[M–Br]^+$. HPLC purity: 98.43%.

*1.3.4. 9-(2-(1,3-dioxolan-2-yl)ethoxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium bromide (**B4**).* Yellow solid, 56.2% yield, m.p: 235.6–236.3 °C. ^{1}H NMR (400 MHz, MeOD-d_4) δ 9.81 (s, 1H), 8.70 (s, 1H), 8.12 (d, J = 8.2 Hz, 1H), 8.01 (d, J = 8.4 Hz, 1H), 7.66 (s, 1H), 6.99 (s, 1H), 6.13 (s, 2H), 5.32 – 5.13 (m, 1H), 5.05 – 4.94 (m, 2H), 4.57 (t, J = 5.4 Hz, 2H), 4.12 (s, 3H), 4.09 – 3.83 (m, 4H), 3.34 (t, J = 5.7 Hz, 2H), 2.43 – 2.21 (m, 2H). ^{13}C NMR (101 MHz, MeOD-d_4) δ 150.78, 150.65, 148.53, 145.27 (2C), 143.32, 138.22, 133.74, 130.46, 126.52, 123.19, 122.14, 120.46, 120.06, 108.02, 105.16, 102.28, 70.14, 64.64 (2C), 56.25, 56.00, 34.10, 26.85. ESI-MS *m/z*: 422.3 $[M–Br]^+$. HPLC purity: 98.73%.

*1.4. General procedure for the synthesis of compounds **B5**–**B8***

B1 or **B2** (0.5 mmol) was dissolved with 10.0 mL acetonitrile, appropriate nucleophiles (0.75 mmol) and K_2CO_3 (1.0 mmol) were added, stirred at 60 ℃ for 8 h, and TLC monitoring was performed (CH_2Cl_2 : MeOH=10:1). After the reaction, acetonitrile was removed under vacuum, and the residues was purified by column chromatography (eluent: CH_2Cl_2/MeOH) to obtain the target products **B5**–**B8**.

*1.4.1. 9-(3-((3-carboxypropyl)amino)propoxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B5**).* Yellow solid, 46% yield, m.p: 216.7–217.3 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.82 (s, 1H), 8.96 (s, 1H), 8.18 (d, J = 9.2 Hz, 1H), 7.99 (d, J = 9.2 Hz, 1H), 7.79 (s, 1H), 7.08 (s, 1H), 6.16 (s, 2H), 4.96 (t, J = 5.6 Hz, 2H), 4.37 (t, J = 6.5 Hz, 2H), 4.04 (s, 3H), 3.66 (t, J =

6.1 Hz, 2H), 3.23 – 3.18 (m, 2H), 2.03 – 1.98 (m, 2H), 1.24 – 1.18 (m, 4H), 0.94 – 0.75 (m, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 161.63, 150.90, 150.29, 148.16, 145.86, 143.42, 137.92, 133.55, 131.16, 130.13, 127.20, 123.82, 122.16, 120.72, 108.91, 105.95, 102.56, 72.43, 58.08, 57.93, 57.58, 55.83, 33.27, 29.55, 29.49, 26.84. ESI-MS *m/z*: 466.0 $[M–Cl]^+$. HPLC purity: 97.96%.

*1.4.2. 9-(3-(1H-1,2,4-triazol-1-yl)propoxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B6**).* Yellow solid, 54% yield, m.p: 218.2–219.0 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.84 (s, 1H), 8.93 (s, 1H), 8.60 (s, 1H), 8.19 (d, J = 9.2 Hz, 1H), 8.02 (s, 1H), 7.99 (d, J = 9.1 Hz, 1H), 7.79 (s, 1H), 7.09 (s, 1H), 6.17 (s, 2H), 4.96 (t, J = 5.9 Hz, 2H), 4.52 (t, J = 6.8 Hz, 2H), 4.28 (t, J = 6.1 Hz, 2H), 4.03 (s, 3H), 3.22 (t, J = 5.9 Hz, 2H), 2.44 – 2.36 (m, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 152.00, 150.83, 150.35, 148.20, 145.81, 144.75, 142.88, 138.02, 133.51, 131.21, 127.12, 124.10, 122.00, 120.93, 120.72, 108.93, 105.92, 102.60, 71.56, 57.49, 55.79, 46.10, 30.49, 26.83. ESI-MS *m/z*: 431.2 $[M–Cl]^+$. HPLC purity: 98.97%.

*1.4.3. 9-((5-(1H-1,2,4-triazol-1-yl)pentyl)oxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B7**).* Yellow solid, 55% yield, m.p: 101.8–102.6 °C. ^{1}H NMR (400 MHz, MeOD-d_4) δ 9.68 (s, 1H), 8.70 (s, 1H), 8.53 (s, 1H), 8.33 (s, 1H), 8.12 (d, J = 9.1 Hz, 1H), 8.00 (d, J = 3.0 Hz, 1H), 7.66 (s, 1H), 6.98 (s, 1H), 6.13 (s, 2H), 4.99 (t, J = 6.3 Hz, 2H), 4.42 (t, J = 6.4 Hz, 2H), 4.36 (t, J = 6.9 Hz, 2H), 4.11 (s, 3H), 3.30 (t, J = 5.6 Hz, 2H), 2.09 – 2.03 (m, 2H), 2.02

– 1.96 (m, 2H), 1.64 – 1.56 (m, 2H). ^{13}C NMR (101 MHz, MeOD-d_4) δ 150.79, 150.72, 150.64, 148.51, 144.79, 143.73, 143.45, 138.23, 133.78, 130.51, 126.50, 123.08, 122.14, 120.45, 120.16, 108.01, 105.16, 102.29, 74.02, 56.23, 55.93, 49.13, 29.17, 29.12, 26.83, 22.60. ESI-MS *m/z*: 459.3 $[M–Cl]^+$. HPLC purity: 96.93%.

*1.4.4. 9-(3-(4-ethylpiperazin-1-yl)propoxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B8**).* Yellow solid, 41% yield, m.p: 160.5–161.3 °C. ^{1}H NMR (400 MHz, MeOD-d_4) δ 9.73 (s, 1H), 8.71 (s, 1H), 8.12 (d, *J* = 9.1 Hz, 1H), 8.01 (d, *J* = 9.1 Hz, 1H), 7.66 (s, 1H), 6.97 (s, 1H), 6.11 (s, 2H), 4.96 (t, *J* = 6.0 Hz, 2H), 4.45 (t, *J* = 6.3 Hz, 2H), 4.11 (s, 3H), 3.27 (t, *J* = 5.3 Hz, 2H), 2.77 – 2.70 (m, 4H), 2.64 – 2.53 (m, 4H), 2.20 – 2.14 (m, 2H), 2.10 – 1.90 (m, 2H), 1.30 – 1.29 (m, 2H), 1.16 (t, *J* = 7.2 Hz, 3H). ^{13}C NMR (101 MHz, MeOD-d_4) δ 150.78, 150.64, 148.53, 144.92, 143.42, 138.27, 133.78, 130.53, 126.46, 123.21, 122.14, 120.49, 120.18, 108.02, 105.18, 102.30, 72.63, 56.24, 55.89, 54.58, 51.98 (2C), 51.82 (3C), 26.98, 26.83, 10.11. ESI-MS *m/z*: 476.4 $[M–Cl]^+$. HPLC purity: 99.16%.

*1.5. General procedure for the synthesis of compounds **B9**–**B28***

Synthesis method 1: A solution of various substituted carboxylic acids (0.7 mmol) in 5.0 mL anhydrous CH_2Cl_2 was stirred at 0 °C for 5 min, followed by the addition of oxaloyl chloride (2.1 mmol) and anhydrous DMF (1-2 drops). The reaction was continued at 0 °C for 1 h, and the system was dried, then 5mL anhydrous acetonitrile was added to dissolve, and the intermediate **1** (0.2 mmol) and 2-3 drops of pyridine were added and stirred at room temperature for 0.5-1 h. TLC monitoring was

performed (CH_2Cl_2 : MeOH=10:1). After the reaction, the solid precipitates were filtered and washed with DCM three times, and then dried to obtain the target product **B10**–**B12**, **B16**, **B17** and **B23**.

Synthesis method 2: The intermediate **1** (0.2 mmol) was dissolved with 5.0 mL anhydrous acetonitrile, various substituted acyl chloride (0.3 mmol) and 2-3 drops of pyridine were added and stirred at room temperature for 0.5-1 h. TLC monitoring was performed (CH_2Cl_2 : MeOH=10:1). After the reaction, the solid precipitates were filtered and washed with DCM three times, and then dried to obtain the target product **B9**, **B13–B15**, **B18–B23**, **B24–B28.**

*1.5.1. 9-((cyclopropanecarbonyl)oxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B9**).* Yellow solid, 79% yield, m.p: >250.0 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.88 (s, 1H), 9.08 (s, 1H), 8.27 (d, J = 9.3 Hz, 1H), 8.21 (d, J = 9.2 Hz, 1H), 7.81 (s, 1H), 7.08 (s, 1H), 6.17 (s, 2H), 4.98 (t, J = 6.1 Hz, 2H), 4.03 (s, 3H), 3.22 (t, J = 6.1 Hz, 2H), 2.15 – 2.08 (m, 1H), 1.23 – 1.18 (m, 4H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 172.16, 150.87, 150.44, 148.17, 144.77, 138.51, 134.07, 133.41, 131.32, 127.15, 126.36, 121.60, 121.13, 120.82, 108.89, 106.05, 102.61, 57.78, 55.79, 26.67, 13.29, 10.16 (2C). ESI-MS *m/z*: 390.2 $[M–Cl]^+$. HPLC purity: 99.77%.

*1.5.2. 9-((cyclopentanecarbonyl)oxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B10**).* Yellow solid, 80% yield, m.p: 232.6–232.2 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.94 (s, 1H), 9.07 (s, 1H), 8.27 (d,

J = 9.3 Hz, 1H), 8.20 (d, *J* = 9.2 Hz, 1H), 7.80 (s, 1H), 7.08 (s, 1H), 6.17 (s, 2H), 4.98 (t, *J* = 6.0 Hz, 2H), 4.02 (s, 3H), 3.22 (t, *J* = 5.8 Hz, 2H), 2.09 – 2.01 (m, 4H), 1.77 – 1.64 (m, 5H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 173.91, 150.81, 150.45, 148.19, 144.90, 138.51, 134.37, 133.41, 131.33, 127.05, 126.35, 121.64, 121.26, 120.85, 108.90, 106.03, 102.62, 57.78, 55.82, 43.44, 29.93, 29.90, 26.68, 25.91 (2C) . ESI-MS *m/z*: 418.3 $[M–Cl]^+$. HPLC purity: 98.89%.

*1.5.3. 9-(2-cyclopentylacetoxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B11**).* Yellow solid, 86% yield, m.p: 230.4–231.1 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.05 (s, 1H), 9.08 (s, 1H), 8.27 (d, *J* = 9.2 Hz, 1H), 8.20 (d, *J* = 9.2 Hz, 1H), 7.80 (s, 1H), 7.08 (s, 1H), 6.17 (s, 2H), 5.00 (t, *J* = 5.8 Hz, 2H), 4.02 (s, 3H), 3.22 (t, *J* = 6.0 Hz, 2H), 2.89 (d, *J* = 7.3 Hz, 2H), 2.40 – 2.30 (m, 1H), 1.97 – 1.85 (m, 2H), 1.73 – 1.63 (m, 2H), 1.61 – 1.51 (m, 2H), 1.38 – 1.28 (m, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 170.54, 150.84, 150.43, 148.17, 144.93, 138.53, 134.12, 133.42, 131.31, 127.13, 126.35, 121.66, 121.09, 120.85, 108.89, 106.04, 102.61, 57.69, 55.89, 55.62, 36.33, 32.38 (2C), 26.67, 25.16 (2C). ESI-MS *m/z*: 432.3 $[M–Cl]^+$. HPLC purity: 98.94%

*1.5.4. 9-((3-cyclopentylpropanoyl)oxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B12**).* Yellow solid, 84% yield, m.p: 219.2–220.1 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.00 (s, 1H), 9.06 (s, 1H), 8.27 (d, *J* = 9.3 Hz, 1H), 8.21 (d, *J* = 9.2 Hz, 1H), 7.81 (s, 1H), 7.09 (s, 1H), 6.17 (s, 2H), 4.98 (t, *J* = 5.9 Hz, 2H), 4.02 (s, 3H), 3.22 (t, *J* = 6.1 Hz, 2H), 2.88

(t, J = 7.6 Hz, 2H), 1.98 – 1.89 (m, 1H), 1.87 – 1.79 (m, 2H), 1.79 – 1.72 (m, 2H), 1.67 – 1.59 (m, 2H), 1.58 – 1.49 (m, 2H), 1.23 – 1.13 (m, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 171.20, 150.86, 150.45, 148.19, 144.93, 138.55, 134.10, 133.42, 131.33, 127.15, 126.36, 121.66, 121.08, 120.85, 108.91, 106.03, 102.62, 57.72, 55.73, 33.05, 32.44 (3C), 30.76, 26.67, 25.23 (2C). ESI-MS *m/z*: 446.3 $[M–Cl]^+$. HPLC purity: 96.51%.

*1.5.5. 9-((furan-2-carbonyl)oxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B13**).* Yellow solid, 89% yield, m.p: 225.8–226.4 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.98 (s, 1H), 9.13 (s, 1H), 8.33 (d, J = 9.3 Hz, 1H), 8.28 (d, J = 9.2 Hz, 1H), 8.23 (s, 1H), 7.82 (s, 1H), 7.74 (d, J = 3.5 Hz, 1H), 7.08 (s, 1H), 6.94 – 6.89 (m, 1H), 6.17 (s, 2H), 4.93 (t, J = 6.0 Hz, 2H), 4.04 (s, 3H), 3.21 (t, J = 6.0 Hz, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 155.44, 150.95, 150.49, 149.77, 148.19, 144.87, 142.68, 138.70, 133.50, 133.10, 131.36, 127.66, 126.36, 121.99, 121.70, 121.18, 120.81, 113.59, 108.90, 106.07, 102.63, 57.81, 55.77, 26.65. ESI-MS m/z: 416.2 $[M–Cl]^+$. HPLC purity: 98.50%.

*1.5.6. 10-methoxy-9-((thiophene-2-carbonyl)oxy)-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B14**).* Yellow solid, 83% yield, m.p: 221.6–222.3 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.99 (s, 1H), 9.16 (s, 1H), 8.33 (d, J = 9.3 Hz, 1H), 8.29 (d, J = 9.2 Hz, 1H), 8.23 (d, J = 5.8 Hz, 1H), 8.18 (d, J = 3.7 Hz, 1H), 7.83 (s, 1H), 7.44 – 7.38 (m, 1H), 7.08 (s, 1H), 6.17 (s, 2H), 4.94 (t, J = 6.1 Hz, 2H), 4.04 (s, 3H), 3.20 (t, J = 6.1 Hz, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 159.33, 151.01, 150.47, 148.18, 144.85, 138.66, 136.75 (2C), 133.64, 133.48, 131.36, 131.14,

129.42, 127.65, 126.36, 121.71, 121.21, 120.82, 108.89, 106.09, 102.62, 57.85, 55.76, 26.65. ESI-MS *m/z*: 432.2 $[M–Cl]^+$. HPLC purity: 97.38%.

*1.5.7. 10-methoxy-9-(2-(thiophen-2-yl)acetoxy)-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B15**).* Yellow solid, 86% yield, m.p: >250 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.22 (s, 1H), 9.07 (s, 1H), 8.27 (d, J = 9.2 Hz, 1H), 8.21 (d, J = 9.2 Hz, 1H), 7.81 (s, 1H), 7.50 (d, J = 4.8 Hz, 1H), 7.18 (d, J = 2.1 Hz, 1H), 7.09 (s, 1H), 7.08 – 7.03 (m, 1H), 6.17 (s, 2H), 5.01 (d, J = 5.5 Hz, 2H), 4.63 (s, 2H), 4.00 (s, 3H), 3.27 – 3.20 (m, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 168.59, 150.88, 150.45, 148.18, 144.99, 138.63, 134.84, 133.78, 133.41, 131.34, 128.21, 127.51, 127.33, 126.50, 126.34, 121.55, 121.06, 120.83, 108.92, 106.02, 102.62, 57.72, 55.66, 34.63, 26.66. ESI-MS *m/z*: 446.1 $[M–Cl]^+$. HPLC purity: 99.01%.

*1.5.8. 10-methoxy-9-((3-(thiophen-2-yl)propanoyl)oxy)-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B16**).* Yellow solid, 80% yield, m.p: 219.7–220.5 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.08 (s, 1H), 9.07 (s, 1H), 8.26 (d, J = 9.3 Hz, 1H), 8.21 (d, J = 9.2 Hz, 1H), 7.80 (s, 1H), 7.39 (d, J = 5.0 Hz, 1H), 7.08 (s, 1H), 7.04 – 7.00 (m, 2H), 6.16 (s, 2H), 5.00 (t, J = 5.9 Hz, 2H), 3.99 (s, 3H), 3.32 – 3.27 (m, 4H), 3.22 (t, J = 5.9 Hz, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 170.12, 150.88, 150.44, 148.17, 144.88, 142.94, 138.57, 133.83, 133.41, 131.31, 127.52, 127.29, 126.36, 125.54, 124.57, 121.58, 121.10, 120.83, 108.90, 106.04, 102.61, 57.67, 55.68, 35.42, 26.67, 24.77. ESI-MS *m/z*: 460.2 $[M–Cl]^+$. HPLC purity: 99.32%.

*1.5.9. 10-methoxy-9-((5-methylthiophene-2-carbonyl)oxy)-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B17**).* Yellow solid, 79% yield, m.p: 195.6–196.3 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.94 (s, 1H), 9.13 (s, 1H), 8.31 (d, J = 9.3 Hz, 1H), 8.27 (d, J = 9.2 Hz, 1H), 8.00 (d, J = 3.7 Hz, 1H), 7.82 (s, 1H), 7.13 (d, J = 3.6 Hz, 1H), 7.07 (s, 1H), 6.17 (s, 2H), 4.94 (t, J = 6.1 Hz, 2H), 4.03 (s, 3H), 3.20 (t, J = 6.1 Hz, 2H), 2.62 (s, 3H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 159.11, 151.24, 151.03, 150.46, 148.17, 144.80, 138.62, 137.23, 133.64, 133.46, 131.34, 128.35, 128.25, 127.54, 126.34, 121.75, 121.19, 120.81, 108.88, 106.07, 102.62, 57.82, 55.77, 26.65, 16.06. ESI-MS *m/z*: 446.2 $[M–Cl]^+$. HPLC purity: 98.87%.

*1.5.10. 9-((5-chlorothiophene-2-carbonyl)oxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B18**).* Yellow solid, 85% yield, m.p: 219.3–220.2 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.04 (s, 1H), 9.17 (s, 1H), 8.33 (d, J = 9.3 Hz, 1H), 8.30 (d, J = 9.3 Hz, 1H), 8.08 (d, J = 4.1 Hz, 1H), 7.83 (s, 1H), 7.48 (d, J = 4.1 Hz, 1H), 7.08 (s, 1H), 6.17 (s, 2H), 4.94 (t, J = 6.0 Hz, 2H), 4.04 (s, 3H), 3.21 (t, J = 6.0 Hz, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 158.21, 150.93, 150.48, 148.18, 144.83, 138.72, 138.33, 136.99, 133.48, 133.16, 131.36, 129.76, 129.68, 127.86, 126.33, 121.55, 121.22, 120.81, 108.89, 106.11, 102.63, 57.89, 55.75, 26.64. ESI-MS m/z: 466.2 $[M–Cl]^+$. HPLC purity: 96.90%.

*1.5.11. 9-((4-fluorobenzoyl)oxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B19**).* Yellow solid, 90% yield, m.p: >250 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.03 (s, 1H), 9.14 (s, 1H), 8.38 – 8.32 (m,

3H), 8.29 (d, *J* = 9.2 Hz, 1H), 7.83 (s, 1H), 7.58 – 7.51 (m, 2H), 7.08 (s, 1H), 6.17 (s, 2H), 4.93 (t, *J* = 5.8 Hz, 2H), 4.03 (s, 3H), 3.20 (t, *J* = 5.8 Hz, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 167.63, 165.11, 162.98, 150.86, 150.45, 148.17, 144.97, 138.61, 134.14, 133.94, 133.46, 131.32, 127.55, 126.31, 125.08, 121.63, 121.16, 120.81, 116.93, 116.71, 108.89, 106.07, 102.63, 57.79, 55.72, 26.63. ESI-MS *m/z*: 444.2 [M–Cl]$^+$. HPLC purity: 97.72%.

*1.5.12. 9-((4-chlorobenzoyl)oxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B20**).* Yellow solid, 88% yield, m.p: 227.2–228.0 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.03 (s, 1H), 9.13 (s, 1H), 8.34 (d, *J* = 9.3 Hz, 1H), 8.31 – 8.26 (m, 3H), 7.83 (s, 1H), 7.78 (d, *J* = 8.6 Hz, 2H), 7.08 (s, 1H), 6.17 (s, 2H), 4.92 (t, *J* = 6.1 Hz, 2H), 4.03 (s, 3H), 3.20 (t, *J* = 6.1 Hz, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 163.14, 150.86, 150.49, 148.20, 144.98, 140.07, 138.67, 133.82, 133.49, 132.76 (2C), 131.34, 129.79 (2C), 127.62, 127.34, 126.36, 121.61, 121.16, 120.82, 108.90, 106.08, 102.63, 57.83, 55.75, 26.65. ESI-MS *m/z*: 460.2 [M–Cl]$^+$. HPLC purity: 97.46%.

*1.5.13. 9-((4-bromobenzoyl)oxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B21**).* Yellow solid, 87% yield, m.p: 215.4–216.1 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.01 (s, 1H), 9.09 (s, 1H), 8.33 (d, *J* = 9.3 Hz, 1H), 8.28 (d, *J* = 9.2 Hz, 1H), 8.19 (d, *J* = 8.5 Hz, 2H), 7.92 (d, *J* = 8.5 Hz, 2H), 7.82 (s, 1H), 7.08 (s, 1H), 6.17 (s, 2H), 4.91 (t, *J* = 5.7 Hz, 2H), 4.02 (s, 3H), 3.20 (t, *J* = 5.1 Hz, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 163.33, 150.85, 150.49, 148.20,

144.97, 138.67, 133.78, 133.46, 132.81 (2C), 132.76 (2C), 131.36, 129.29, 127.70, 127.63, 126.34, 121.60, 121.13, 120.81, 108.91, 106.05, 102.64, 57.81, 55.76, 26.64. ESI-MS m/z: 504.1 $[M–Cl]^+$. HPLC purity: 95.64%.

*1.5.14. 10-methoxy-9-((4-methylbenzoyl)oxy)-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B22**).* Yellow solid, 86% yield, m.p: >250.0 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.98 (s, 1H), 9.12 (s, 1H), 8.33 (d, J = 9.3 Hz, 1H), 8.27 (d, J = 9.2 Hz, 1H), 8.16 (d, J = 8.1 Hz, 2H), 7.83 (s, 1H), 7.50 (d, J = 8.1 Hz, 2H), 7.08 (s, 1H), 6.17 (s, 2H), 4.92 (t, J = 6.0 Hz, 2H), 4.02 (s, 3H), 3.22 – 3.16 (m, 2H), 2.48 (s, 3H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 163.90, 150.92, 150.45, 148.18, 145.74, 144.96, 138.58, 134.14, 133.46, 131.34, 130.99 (2C), 130.12 (2C), 127.39, 126.34, 125.70, 121.75, 121.15, 120.83, 108.90, 106.06, 102.63, 57.77, 55.73, 26.64, 21.86. ESI-MS *m/z*: 440.2 $[M–Cl]^+$. HPLC purity: 97.45%.

*1.5.15. 9-((6-fluoronicotinoyl)oxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B23**).* Yellow solid, 86% yield, m.p: 203.7–204.4 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.08 (s, 1H), 9.15 (s, 1H), 9.10 (s, 1H), 8.84 – 8.74 (m, 1H), 8.35 (d, J = 9.3 Hz, 1H), 8.29 (d, J = 9.3 Hz, 1H), 7.83 (s, 1H), 7.55 (d, J = 8.6 Hz, 1H), 7.09 (s, 1H), 6.18 (s, 2H), 4.91 (t, J = 5.5 Hz, 2H), 4.04 (s, 3H), 3.21 (t, J = 5.3 Hz, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 161.81, 151.49, 150.90, 150.54, 148.23, 145.02, 144.84, 138.78, 133.56, 133.35, 131.37, 127.83, 126.41, 123.65, 121.51, 121.15, 120.82, 111.26, 110.89, 108.92, 106.08, 102.65, 57.88, 55.79, 26.65. ESI-MS *m/z*: 445.2 $[M–Cl]^+$. HPLC purity: 97.34%.

*1.5.16. 10-methoxy-9-((morpholine-4-carbonyl)oxy)-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B24**).* Yellow solid, 79.9% yield, m.p: 221.7–222.3 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.93 (s, 1H), 9.06 (s, 1H), 8.25 (d, J = 9.2 Hz, 1H), 8.18 (d, J = 9.2 Hz, 1H), 7.80 (s, 1H), 7.08 (s, 1H), 6.17 (s, 2H), 5.02 (t, J = 6.1 Hz, 2H), 4.04 (s, 3H), 3.83 – 3.75 (m, 4H), 3.74 – 3.67 (m, 2H), 3.56 – 3.44 (m, 2H), 3.22 (t, J = 6.1 Hz, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 152.19, 151.22, 150.41, 148.17, 144.93, 138.40, 134.92, 133.30, 131.31, 126.77, 126.38, 122.04, 121.08, 120.87, 108.90, 106.02, 102.62, 66.28 (2C), 57.78, 55.77, 45.63, 44.78, 26.69. ESI-MS *m/z*: 435.2 $[M–Cl]^+$. HPLC purity: 96.57%.

*1.5.17. 10-methoxy-9-((thiophen-2-ylsulfonyl)oxy)-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B25**).* Yellow solid, 80% yield, m.p: 228.5–229.3 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.53 (s, 1H), 9.19 (s, 1H), 8.33 (d, J = 8.4 Hz, 1H), 8.31 (s, 1H), 8.26 (d, J = 9.3 Hz, 1H), 7.93 (d, J = 3.8 Hz, 1H), 7.83 (s, 1H), 7.35 (t, J = 4.4 Hz, 1H), 7.10 (s, 1H), 6.18 (s, 2H), 4.93 (t, J = 6.1 Hz, 2H), 3.81 (s, 3H), 3.21 (t, J = 6.1 Hz, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 152.49, 150.68, 148.24, 144.25, 139.09, 138.86, 137.98, 133.88, 133.45, 131.45, 131.39, 129.32, 129.19, 126.59, 122.21, 121.56, 120.63, 108.94, 106.14, 102.69, 57.68, 56.19, 26.71. ESI-MS *m/z*: 468.2 $[M–Cl]^+$. HPLC purity: 97.20%.

*1.5.18. 9-(((5-chlorothiophen-2-yl)sulfonyl)oxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B26**).* Yellow solid, 81% yield, m.p: 219.4–220.0 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.66 (s, 1H),

9.19 (s, 1H), 8.33 (d, *J* = 9.3 Hz, 1H), 8.28 (d, *J* = 9.4 Hz, 1H), 7.86 (d, *J* = 4.2 Hz, 1H), 7.84 (s, 1H), 7.47 (d, *J* = 4.2 Hz, 1H), 7.11 (s, 1H), 6.19 (s, 2H), 4.97 (t, *J* = 6.1 Hz, 2H), 3.86 (s, 3H), 3.22 (t, *J* = 6.1 Hz, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 152.27, 150.70, 148.25, 144.24, 140.57, 139.24, 138.18, 133.99, 131.64, 131.49, 131.17, 129.64, 129.48, 126.53, 122.16, 121.62, 120.63, 108.94, 106.16, 102.70, 57.65, 56.15, 26.68. ESI-MS *m/z*: 502.1 $[M–Cl]^+$. HPLC purity: 96.97%.

*1.5.19. 9-(((4-fluorophenyl)sulfonyl)oxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B27**).* Yellow solid, 82% yield, m.p: 230.1–230.6 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.62 (s, 1H), 9.12 (s, 1H), 8.28 (d, *J* = 8.9 Hz, 1H), 8.22 (d, *J* = 8.4 Hz, 1H), 8.06 (d, *J* = 7.9 Hz, 2H), 7.82 (s, 1H), 7.58 (d, *J* = 7.4 Hz, 2H), 6.19 (s, 2H), 4.97 (t, *J* = 6.1 Hz, 2H), 3.74 (s, 3H), 3.22 (t, *J* = 6.0 Hz, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 166.55, 152.14, 151.33, 148.78, 143.56, 139.27, 134.13, 131.82, 131.72, 131.42, 131.09, 130.04, 128.92, 125.92, 122.85, 121.30, 119.67, 116.79, 116.66, 108.42, 105.65, 102.35, 56.75 (2C), 27.12. ESI-MS *m/z*: 480.2 $[M–Cl]^+$. HPLC purity: 96.94%.

*1.5.20. 9-(((4-chlorophenyl)sulfonyl)oxy)-10-methoxy-5,6-dihydro-[1,3]dioxolo[4,5-g]isoquinolino[3,2-a]isoquinolin-7-ium chloride (**B28**).* Yellow solid, 82% yield, m.p: 210.3–211.6 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.61 (s, 1H), 9.17 (s, 1H), 8.30 (d, *J* = 9.1 Hz, 1H), 8.22 (d, *J* = 9.3 Hz, 1H), 7.98 (d, *J* = 8.6 Hz, 2H), 7.86 – 7.80 (m, 3H), 7.11 (s, 1H), 6.18 (s, 2H), 4.96 (t, *J* = 6.2 Hz, 2H), 3.73 (s, 3H),

3.22 (t, J = 6.2 Hz, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 152.04, 150.73, 148.28, 144.39, 141.17, 139.23, 133.95, 133.89, 131.51, 131.34, 131.07 (2C), 130.41 (2C), 129.18, 126.56, 122.23, 121.52, 120.64, 108.97, 106.09, 102.71, 57.38, 56.21, 26.68. ESI-MS *m/z*: 496.1 $[M–Cl]^+$. HPLC purity: 95.77%.

*1.6. General procedure for the synthesis of compounds **C1**–**C7***

*1.6.1. 9-((5-bromopentyl)oxy)-2,3,10-trimethoxy-5,6-dihydroisoquinolino[3,2-a]isoquinolin-7-ium bromide (**C1**).* Similar to the synthesis route of **B2**, intermediate **2** replaced intermediate **1**. Yellow solid, 61% yield, m.p: 223.4–224.7 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.75 (s, 1H), 9.07 (s, 1H), 8.20 (d, J = 9.1 Hz, 1H), 8.04 (d, J = 9.1 Hz, 1H), 7.72 (s, 1H), 7.10 (s, 1H), 4.97 (t, J = 5.7 Hz, 2H), 4.30 (t, J = 6.5 Hz, 2H), 4.05 (s, 3H), 3.94 (s, 3H), 3.87 (s, 3H), 3.70 (t, J = 6.5 Hz, 1H), 3.60 (t, J = 6.6 Hz, 1H), 3.23 (t, J = 6.0 Hz, 2H), 1.95 – 1.82 (m, 4H), 1.67 – 1.58 (m, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 151.93, 150.74, 149.19, 145.73, 143.22, 138.09, 133.63, 129.05, 127.10, 123.76, 122.07, 120.44, 119.41, 111.76, 109.22, 74.48, 57.51, 56.73, 56.36, 56.00, 45.92, 32.28, 29.22, 26.46, 23.25. ESI-MS *m/z*: 486.2 $[M–Br]^+$. HPLC purity: 95.14%.

*1.6.2. 9-((5-(1H-1,2,4-triazol-1-yl)pentyl)oxy)-2,3,10-trimethoxy-5,6-dihydroisoquinolino[3,2-a]isoquinolin-7-ium chloride (**C2**).* Similar to the synthesis route of **B7**, intermediate **2** replaced intermediate **1**. Yellow solid, 62% yield, m.p: 215.4–215.2 °C. ^{1}H NMR (400 MHz, MeOD-d_4) δ 9.56 (s, 1H), 8.72 (s, 1H), 8.44 (s, 1H), 8.01 (d, J = 9.1 Hz, 1H), 7.93 (d, J = 9.1 Hz, 1H), 7.90 (s, 1H), 7.56 (s, 1H), 6.96 (s, 1H), 4.90 (t, J = 6.2 Hz, 2H), 4.31 (t, J = 6.4 Hz, 2H), 4.26 (t, J = 6.9 Hz, 2H), 4.00

(s, 3H), 3.92 (s, 3H), 3.86 (s, 3H), 3.24 – 3.20 (m, 2H), 2.00 – 1.93 (m, 2H), 1.92 – 1.86 (m, 2H), 1.54 – 1.46 (m, 2H). ^{13}C NMR (101 MHz, MeOD-d_4) δ 152.42, 150.73, 150.53, 149.50, 144.76, 143.63, 143.40, 138.37, 133.90, 128.68, 126.45, 123.04, 122.09, 119.96, 119.06, 110.84, 108.55, 73.98, 56.19, 56.08, 55.65, 55.29, 49.12, 29.19, 29.08, 26.43, 22.61. ESI-MS *m/z*: 476.3 $[M–Br]^+$. HPLC purity: 96.29%.

*1.6.3. 9-((5-chlorothiophene-2-carbonyl)oxy)-2,3,10-trimethoxy-5,6-dihydroisoquinolino[3,2-a]isoquinolin-7-ium chloride (**C3**).* Similar to the synthesis routes of **B18**, intermediate **2** replaced intermediate **1**. Yellow solid, 79% yield, m.p: 180.1–181.5 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.01 (s, 1H), 9.29 (s, 1H), 8.33 – 8.31 (m, 2H), 8.09 (d, J = 4.1 Hz, 1H), 7.76 (s, 1H), 7.49 (d, J = 4.1 Hz, 1H), 7.08 (s, 1H), 4.95 (t, J = 6.0 Hz, 2H), 4.04 (s, 3H), 3.95 (s, 3H), 3.86 (s, 3H), 3.23 (t, J = 6.1 Hz, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 158.25, 152.14, 150.77, 149.22, 144.77, 138.94, 138.37, 136.99, 133.57, 133.09, 129.84, 129.69, 129.23, 127.76, 126.27, 121.48, 120.94, 119.27, 111.73, 109.40, 57.83, 56.73, 56.38, 55.94, 26.27. ESI-MS *m/z*: 482.2 $[M–Cl]^+$. HPLC purity: 97.10%.

*1.6.4. 9-((4-fluorobenzoyl)oxy)-2,3,10-trimethoxy-5,6-dihydroisoquinolino[3,2-a]isoquinolin-7-ium chloride (**C4**).* Similar to the synthesis routes of **B19**, intermediate **2** replaced intermediate **1**. Yellow solid, 78% yield, m.p: 193.4–194.6 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.02 (s, 1H), 9.30 (s, 1H), 8.39 – 8.35 (m, 2H), 8.34 – 8.32 (m, 2H), 7.77 (s, 1H), 7.58 – 7.52 (m, 2H), 7.08 (s, 1H), 4.94 (t, J = 6.1 Hz, 2H), 4.02 (s, 3H), 3.96 (s, 3H), 3.86 (s, 3H), 3.22 (t, J = 6.1 Hz, 2H). ^{13}C NMR (101 MHz, DMSO-

d_6) δ 167.64, 165.12, 163.01, 152.11, 150.71, 149.22, 144.91, 138.84, 134.04, 133.94, 133.59, 129.20, 127.47, 126.30, 125.13, 121.58, 120.93, 119.30, 116.94, 116.72, 111.73, 109.39, 57.76, 56.73, 56.37, 55.89, 26.28. ESI-MS *m/z*: 460.2 $[M–Cl]^+$. HPLC purity: 97.99%.

*1.6.5. 9-((4-chlorobenzoyl)oxy)-2,3,10-trimethoxy-5,6-dihydroisoquinolino[3,2-a]isoquinolin-7-ium chloride (**C5**).* Similar to the synthesis routes of **B20**, intermediate **2** replaced intermediate **1**. Yellow solid, 76% yield, m.p: 218.7–219.2 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.03 (s, 1H), 9.30 (s, 1H), 8.35 – 8.32 (m, 2H), 8.28 (d, *J* = 8.5 Hz, 2H), 7.81 – 7.76 (m, 3H), 7.08 (s, 1H), 4.94 (t, *J* = 5.9 Hz, 2H), 4.02 (s, 3H), 3.96 (s, 3H), 3.86 (s, 3H), 3.22 (t, *J* = 5.9 Hz, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 163.17, 152.12, 150.68, 149.22, 144.92, 140.07, 138.85, 133.74, 133.59, 132.77 (2C), 131.62, 129.80 (2C), 129.20, 127.58, 127.35, 126.29, 121.53, 120.92, 119.29, 111.73, 109.39, 57.77, 56.73, 56.38, 55.90, 26.28. ESI-MS *m/z*: 476.2 $[M–Cl]^+$. HPLC purity: 99.55%.

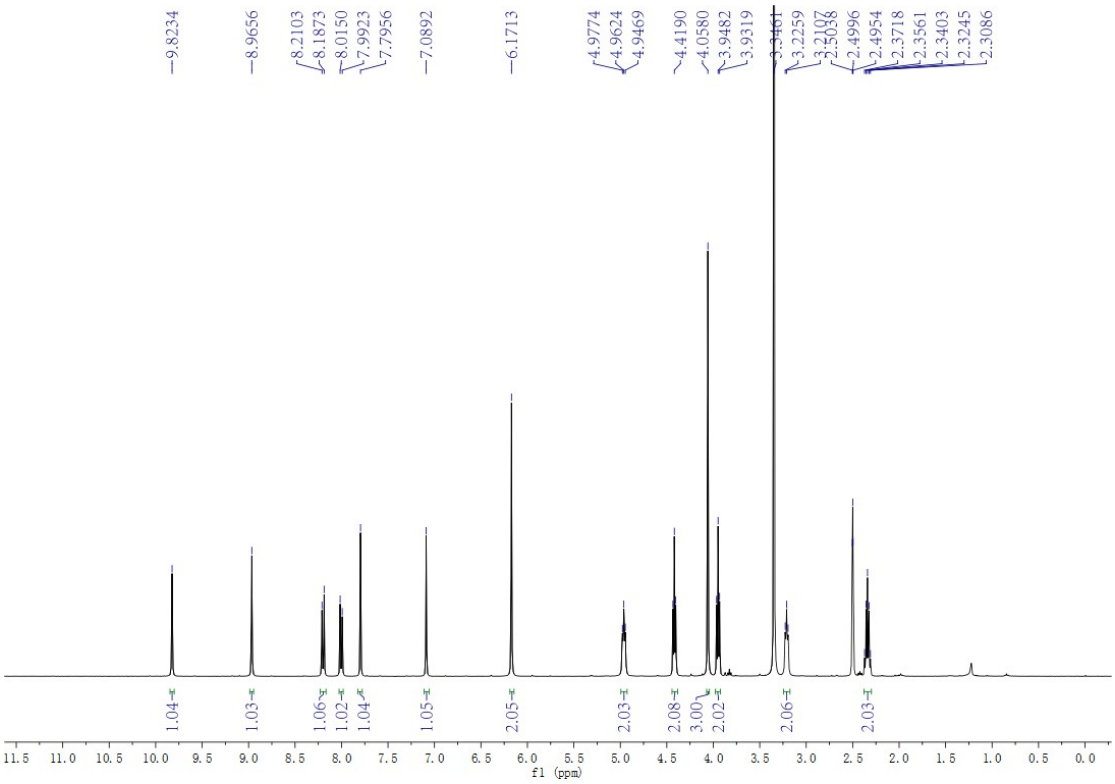
*1.6.6. 2,3,10-trimethoxy-9-((4-methylbenzoyl)oxy)-5,6-dihydroisoquinolino[3,2-a]isoquinolin-7-ium chloride (**C6**).* Similar to the synthesis routes of **B22**, intermediate **2** replaced intermediate **1**. Yellow solid, 78% yield, m.p: 118.5–119.9 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.97 (s, 1H), 9.29 (s, 1H), 8.35 – 8.29 (m, 2H), 8.16 (d, *J* = 8.1 Hz, 2H), 7.77 (s, 1H), 7.51 (d, *J* = 8.1 Hz, 2H), 7.09 (s, 1H), 4.94 (t, *J* = 6.1 Hz, 2H), 4.01 (s, 3H), 3.96 (s, 3H), 3.86 (s, 3H), 3.22 (t, *J* = 6.1 Hz, 2H), 2.48 (s, 3H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 163.93, 152.10, 150.76, 149.22, 145.72, 144.91, 138.78,

134.10, 133.58, 130.99 (2C), 130.12 (2C), 129.21, 127.30, 126.31, 125.73, 121.68, 120.92, 119.32, 111.73, 109.39, 57.73, 56.73, 56.37, 55.90, 26.29, 21.86. ESI-MS *m/z*: 456.3 $[M–Cl]^+$. HPLC purity: 95.27%.

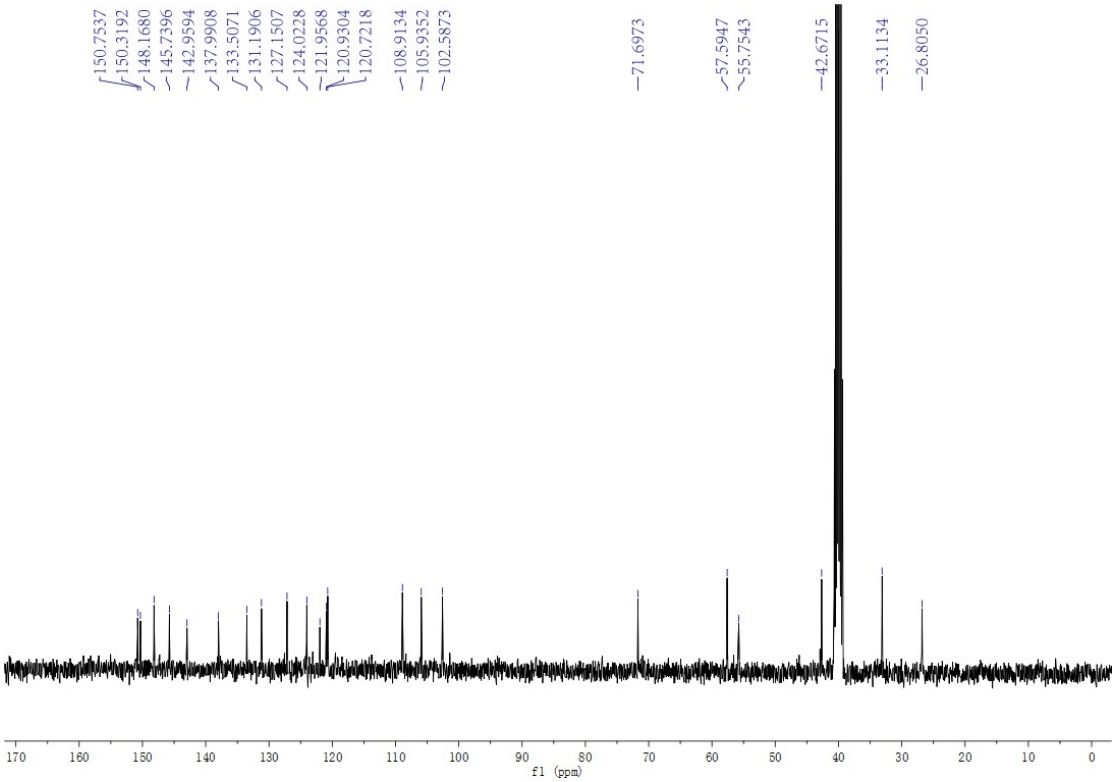
*1.6.7. 9-(((5-chlorothiophen-2-yl)sulfonyl)oxy)-2,3,10-trimethoxy-5,6-dihydroisoquinolino[3,2-a]isoquinolin-7-ium chloride (**C7**).* Similar to the synthesis routes of **B26**, intermediate **2** replaced intermediate **1**. Yellow solid, 76% yield, m.p: >250.0 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.64 (s, 1H), 9.36 (s, 1H), 8.37 (d, J = 9.3 Hz, 1H), 8.28 (d, J = 9.4 Hz, 1H), 7.87 (d, J = 4.2 Hz, 1H), 7.78 (s, 1H), 7.48 (d, J = 4.2 Hz, 1H), 7.12 (s, 1H), 4.99 (t, J = 6.2 Hz, 2H), 3.95 (s, 3H), 3.88 (s, 3H), 3.86 (s, 3H), 3.24 (t, J = 6.2 Hz, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ 152.32, 152.12, 149.26, 144.27, 140.57, 139.44, 138.19, 134.09, 131.63, 131.10, 129.65, 129.38, 129.32, 126.47, 122.06, 121.37, 119.09, 111.76, 109.49, 57.59, 56.76, 56.40, 56.31, 26.32. ESI-MS *m/z*: 518.2 $[M–Cl]^+$. HPLC purity: 95.73%.

2. ^{1}H- and ^{13}C-NMR spectra

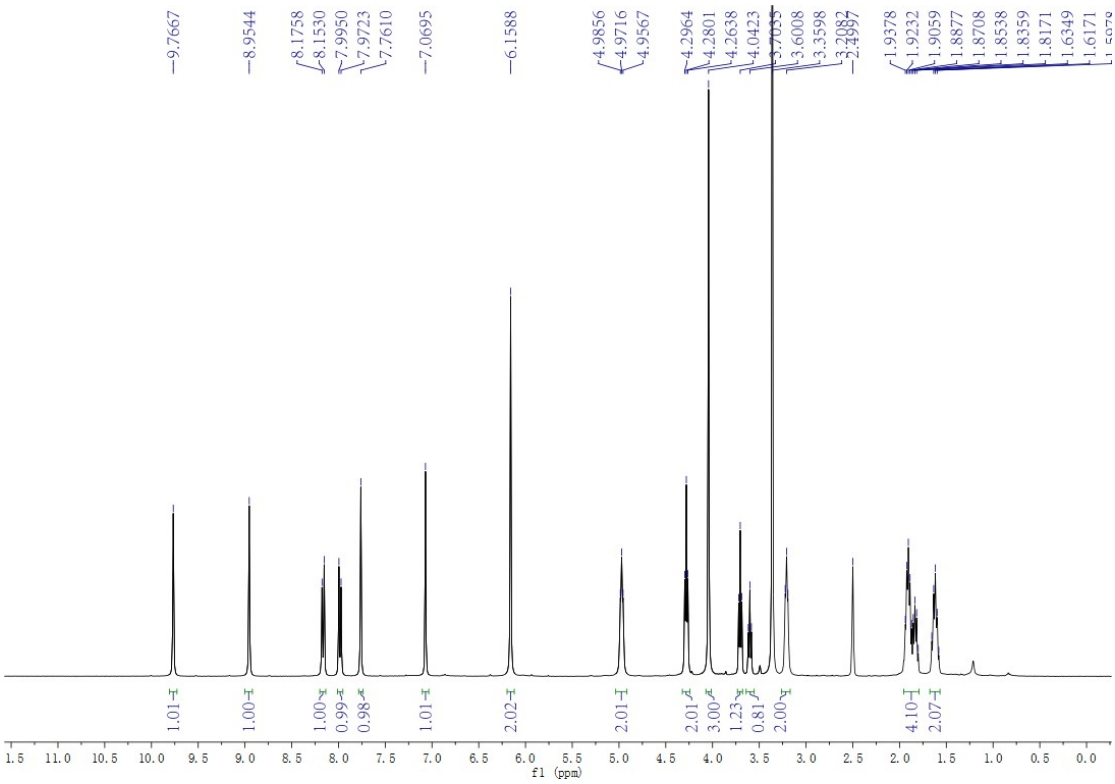
^{1}H NMR Spectra of Compound **B1**



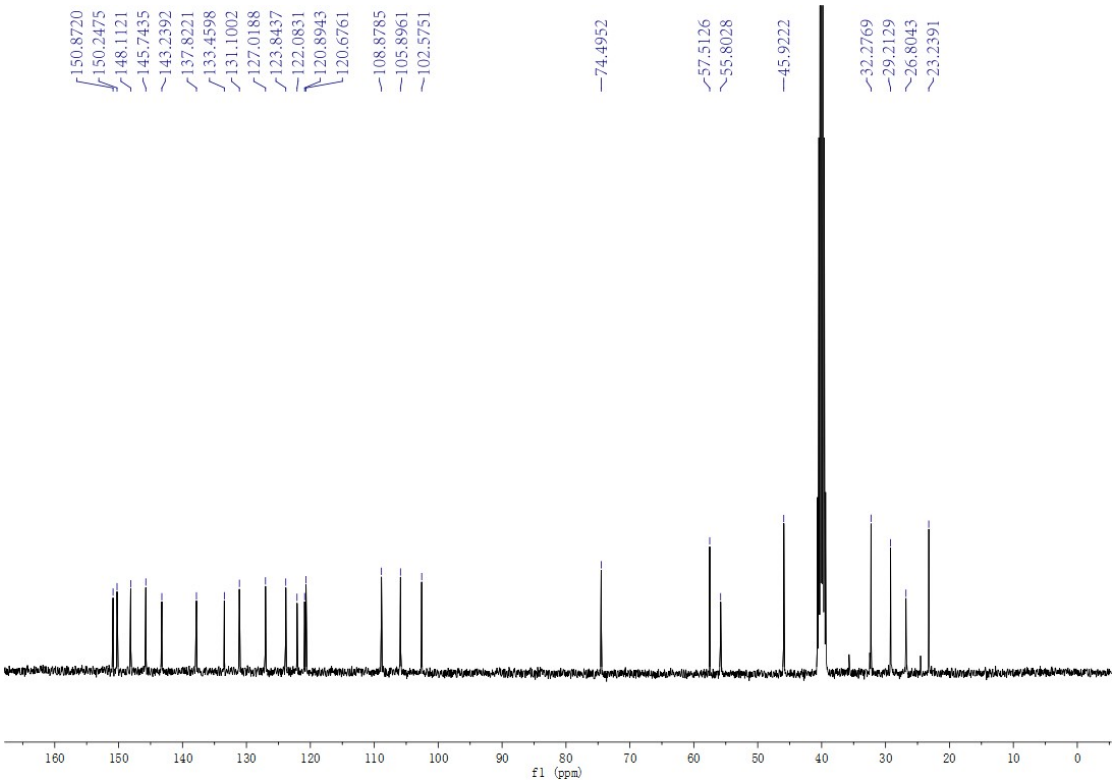
^{13}C NMR Spectra of Compound **B1**



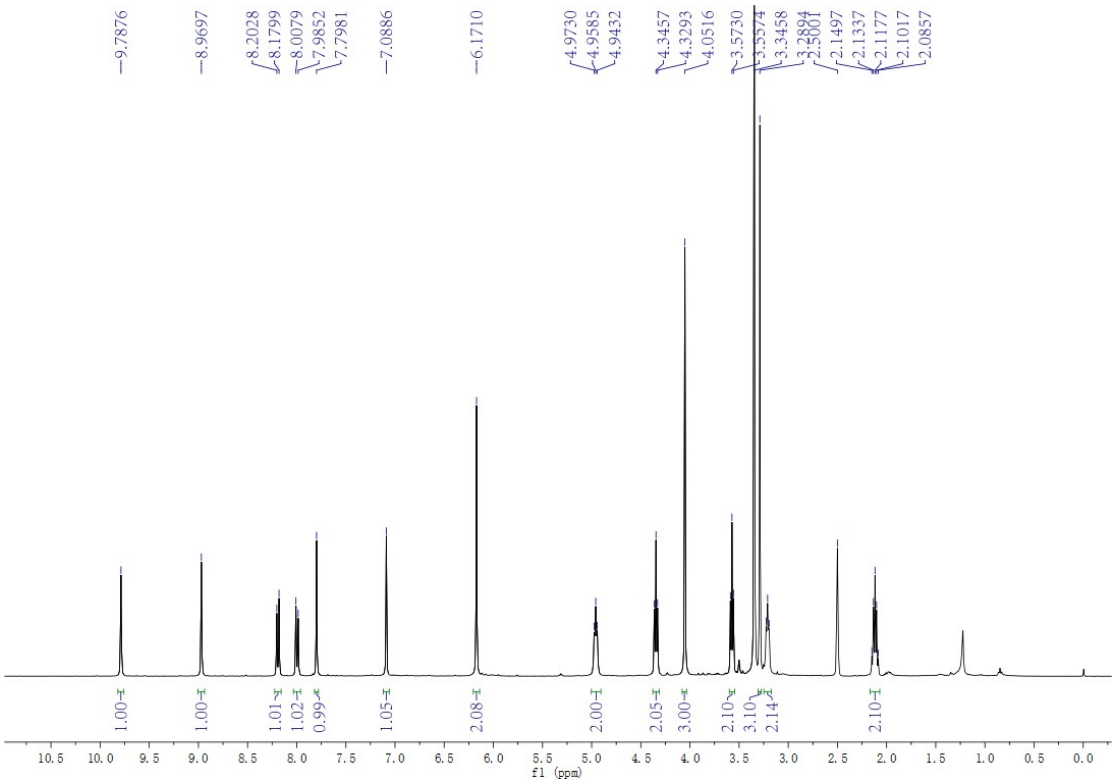
^{1}H NMR Spectra of Compound **B2**



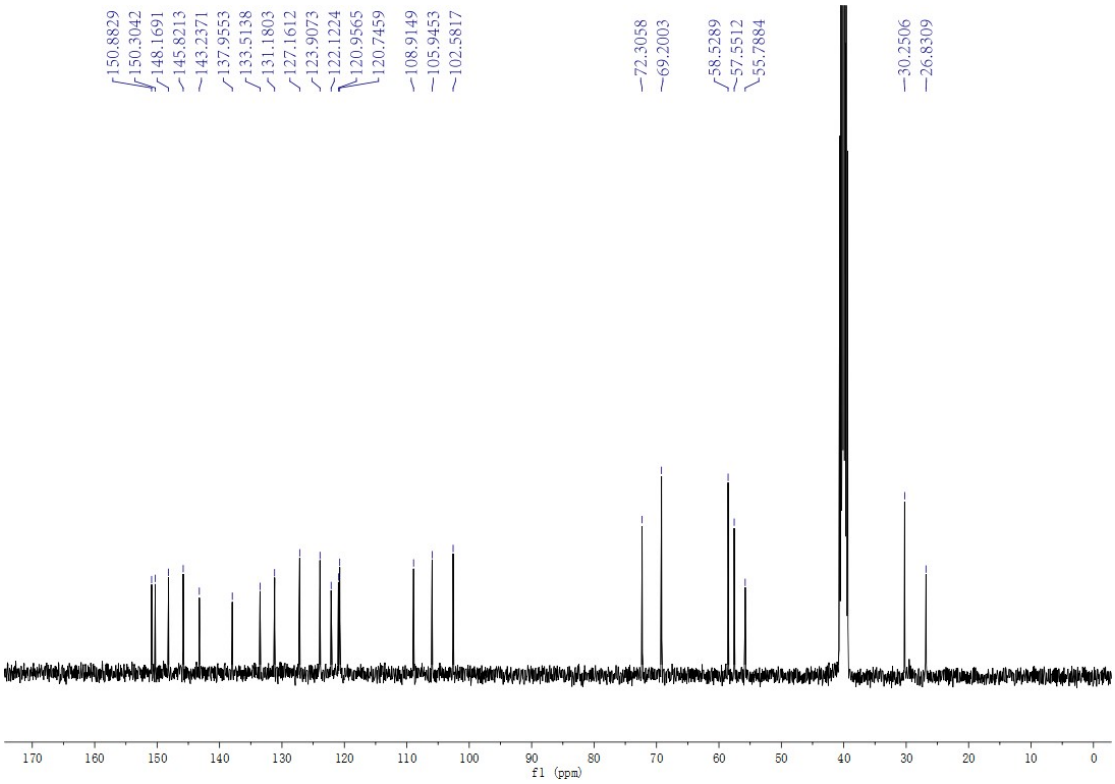
^{13}C NMR Spectra of Compound **B2**



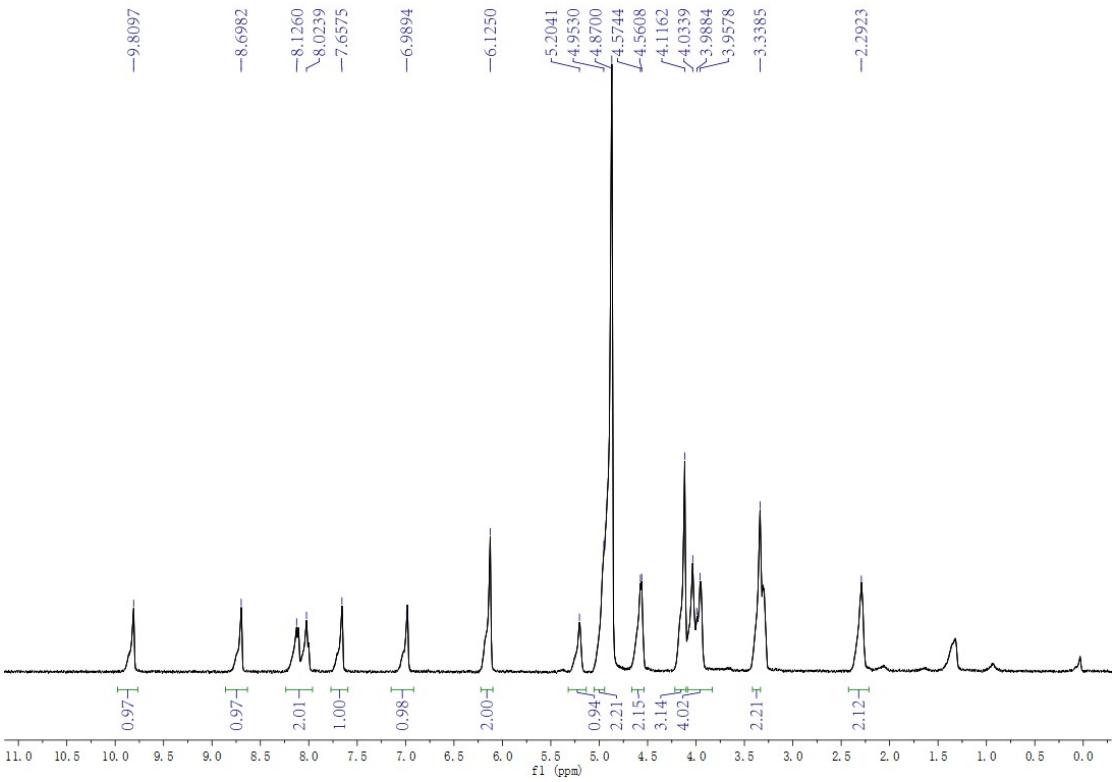
^{1}H NMR Spectra of Compound **B3**



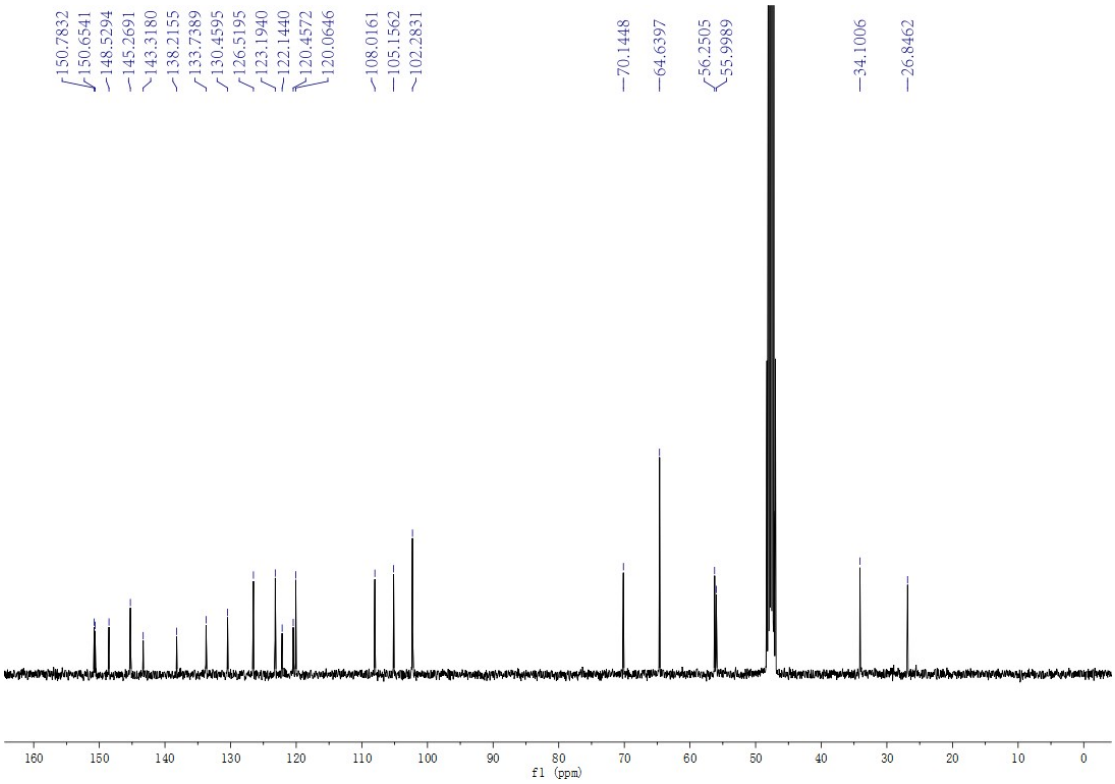
^{13}C NMR Spectra of Compound **B3**



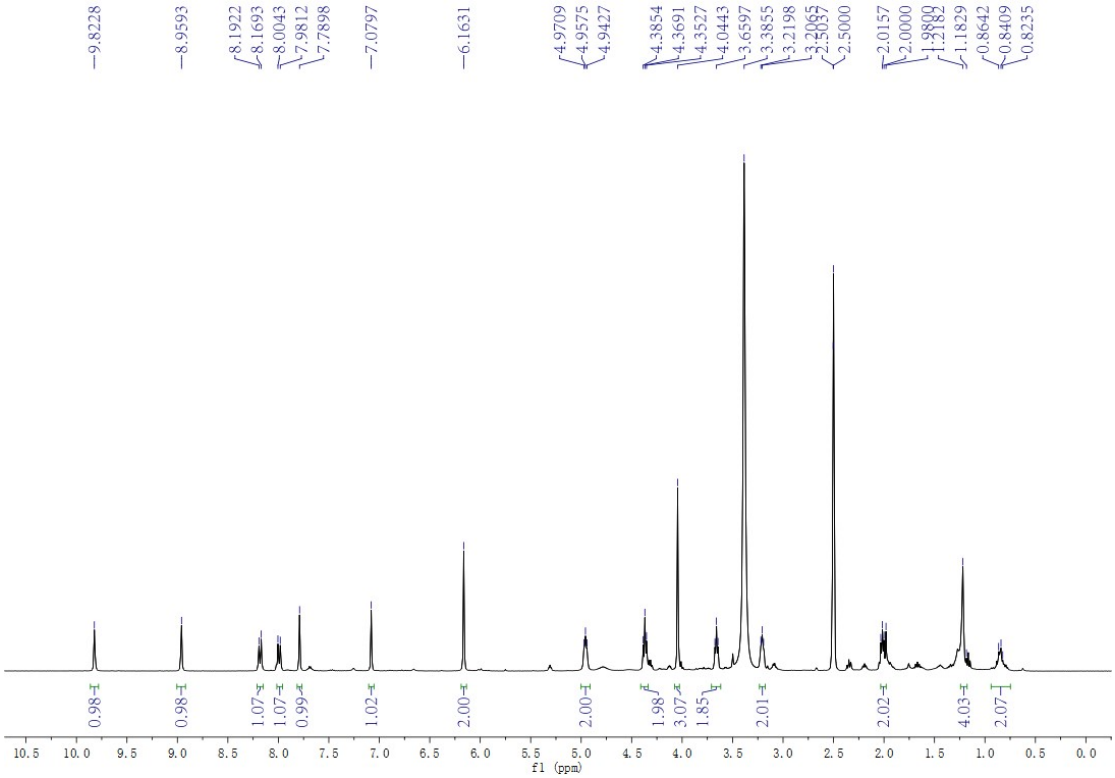
^{1}H NMR Spectra of Compound **B4**



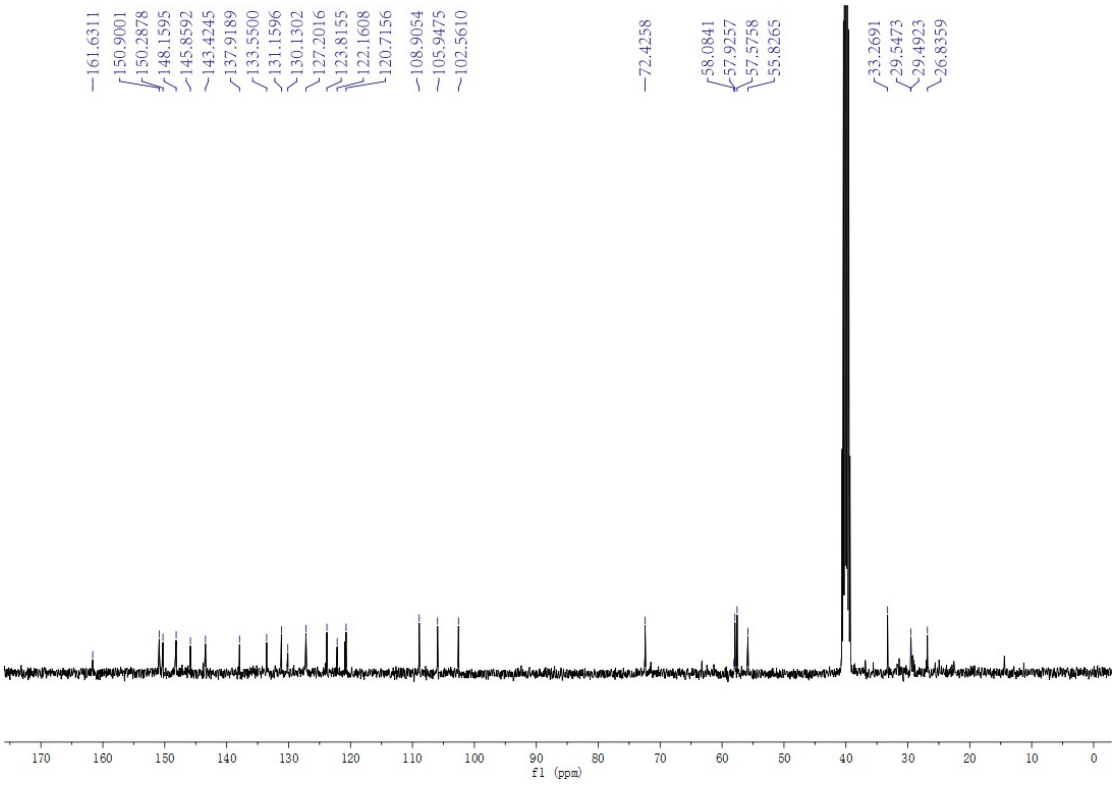
^{13}C NMR Spectra of Compound **B4**



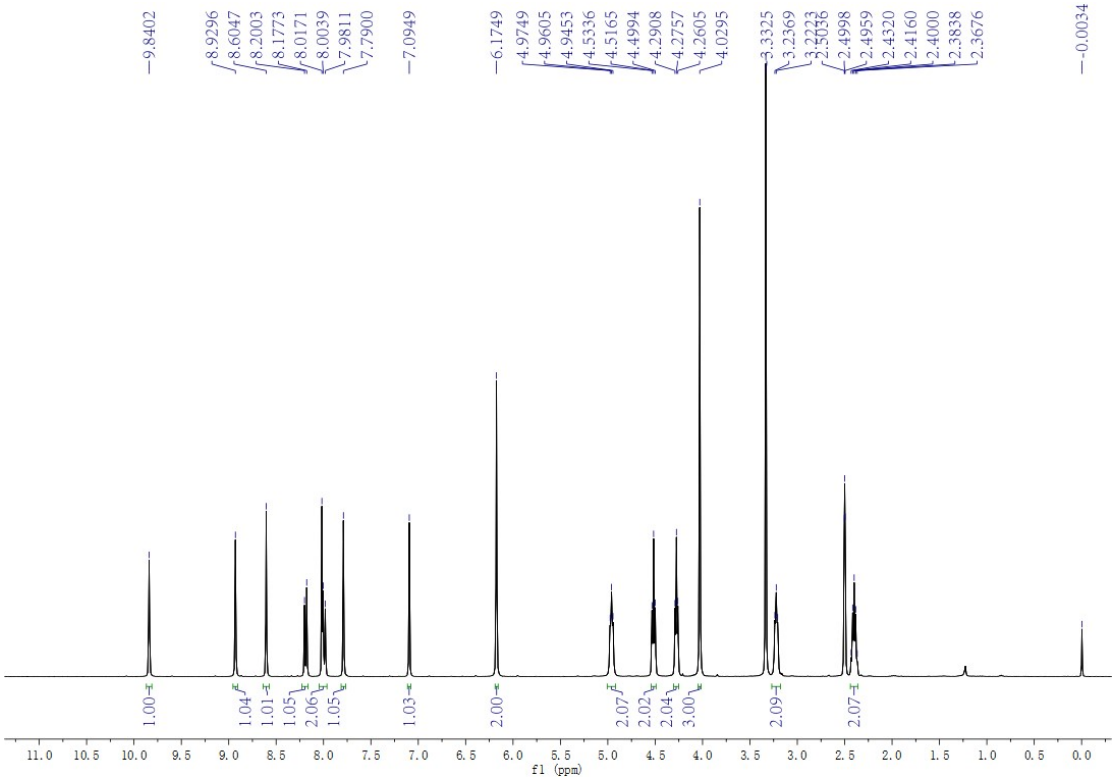
^{1}H NMR Spectra of Compound **B5**



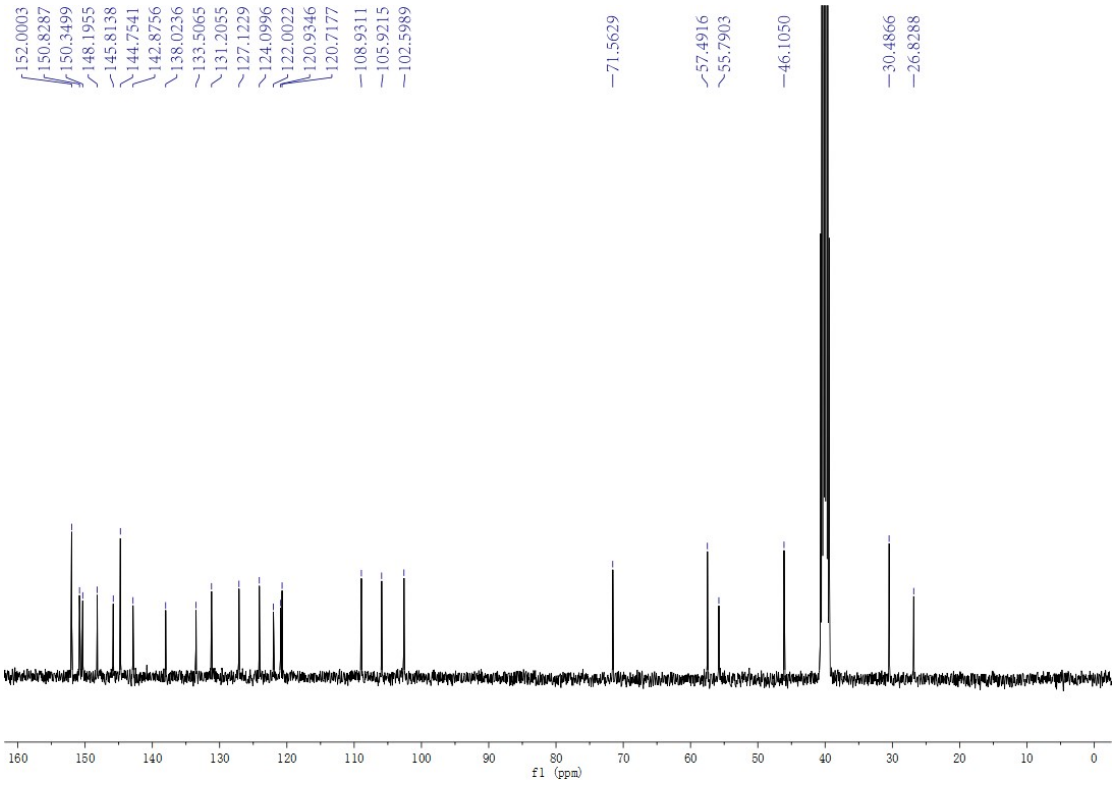
^{13}C NMR Spectra of Compound **B5**



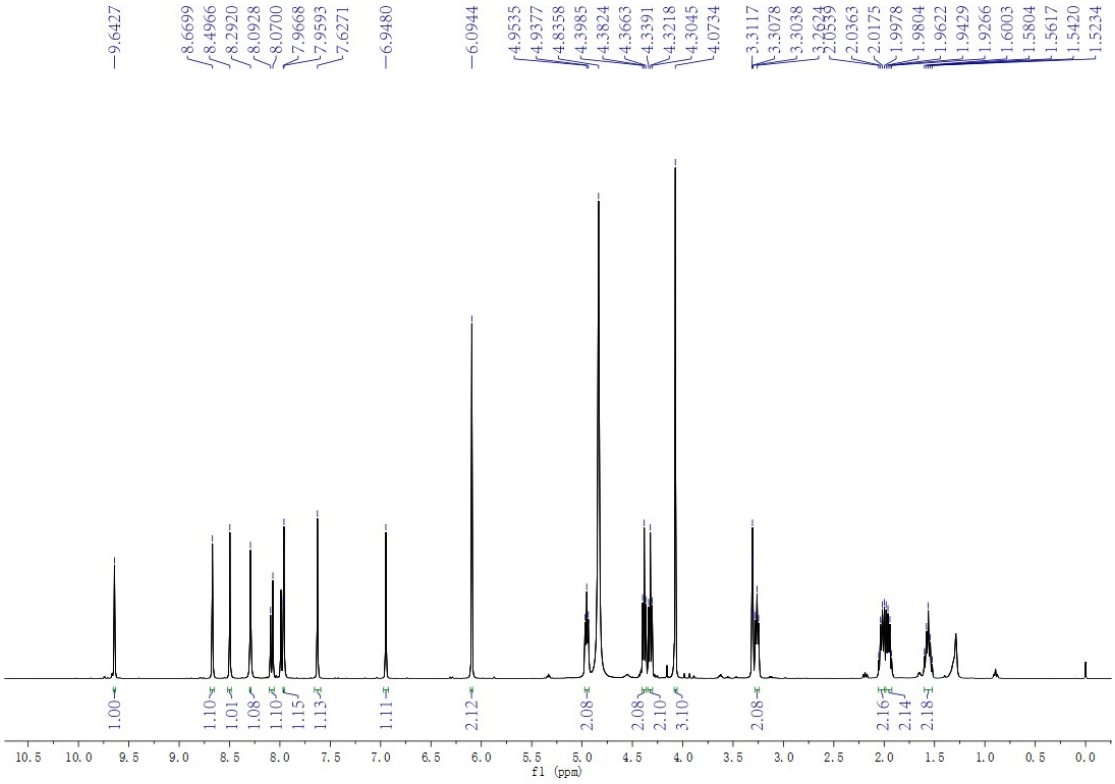
^{1}H NMR Spectra of Compound **B6**



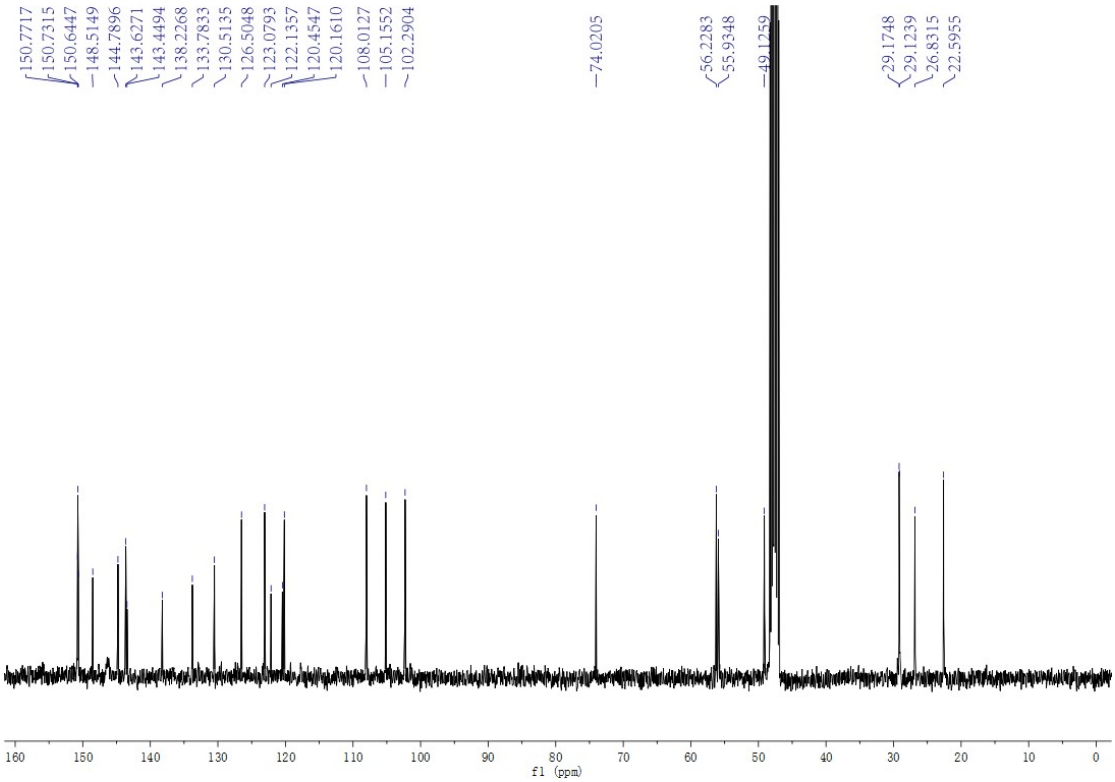
^{13}C NMR Spectra of Compound **B6**



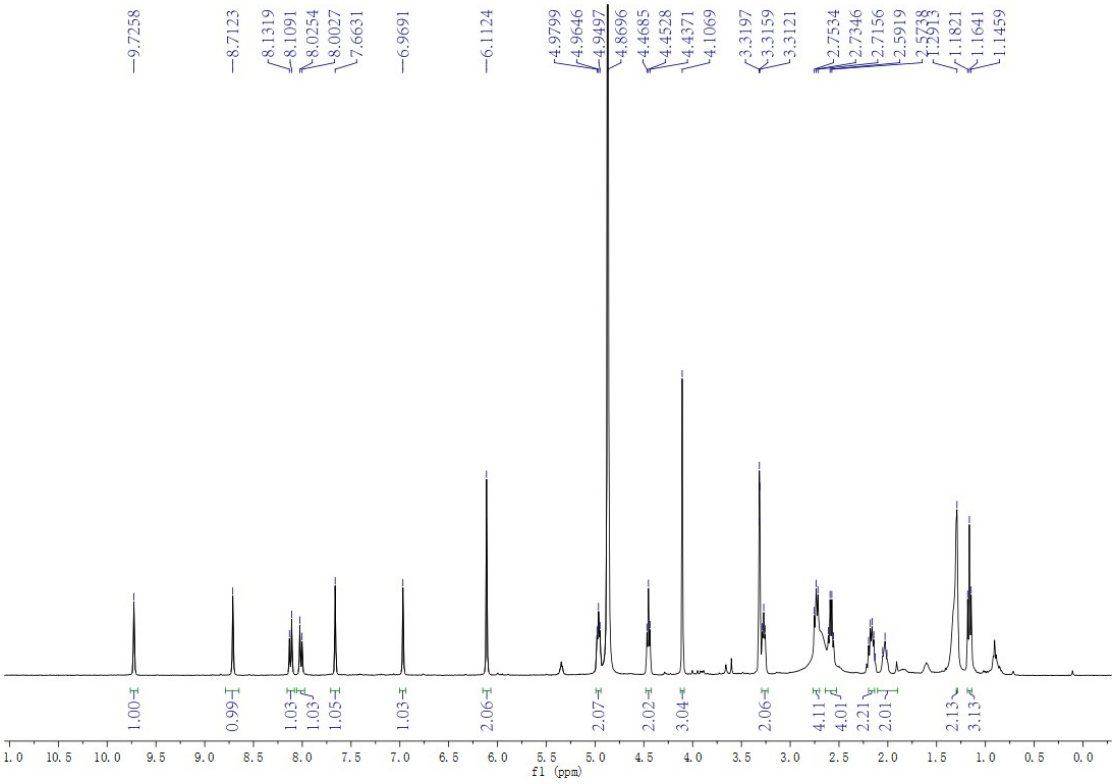
^{1}H NMR Spectra of Compound **B7**



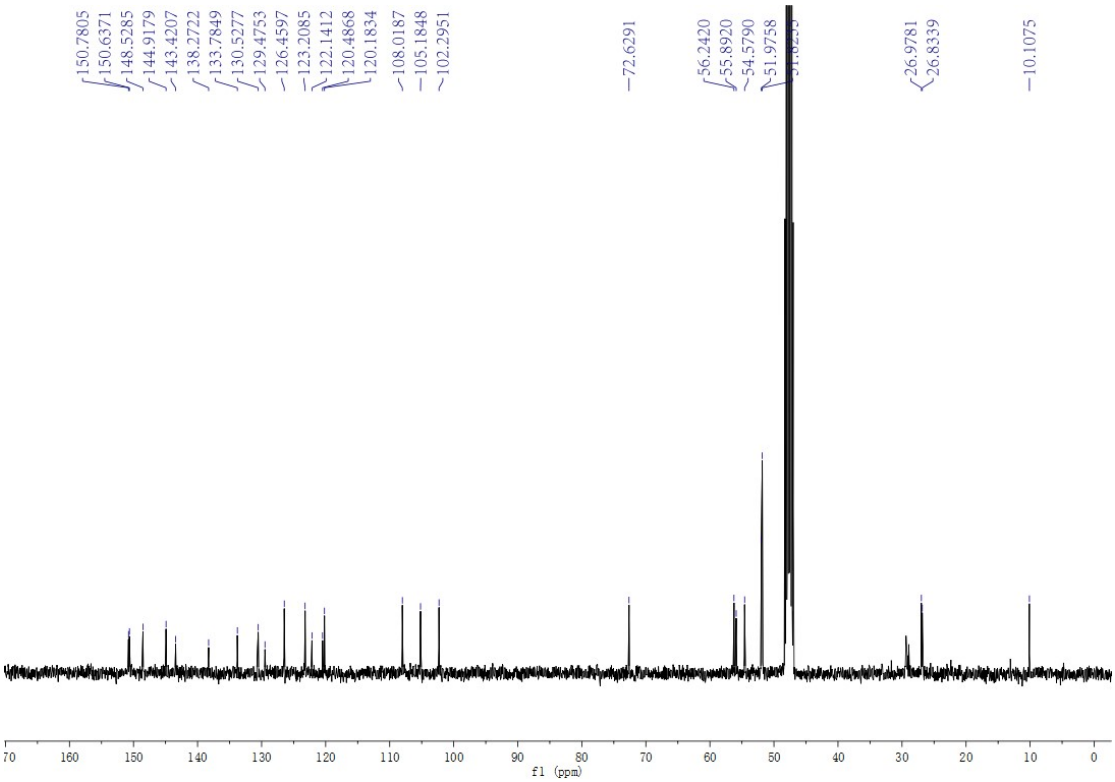
^{13}C NMR Spectra of Compound **B7**



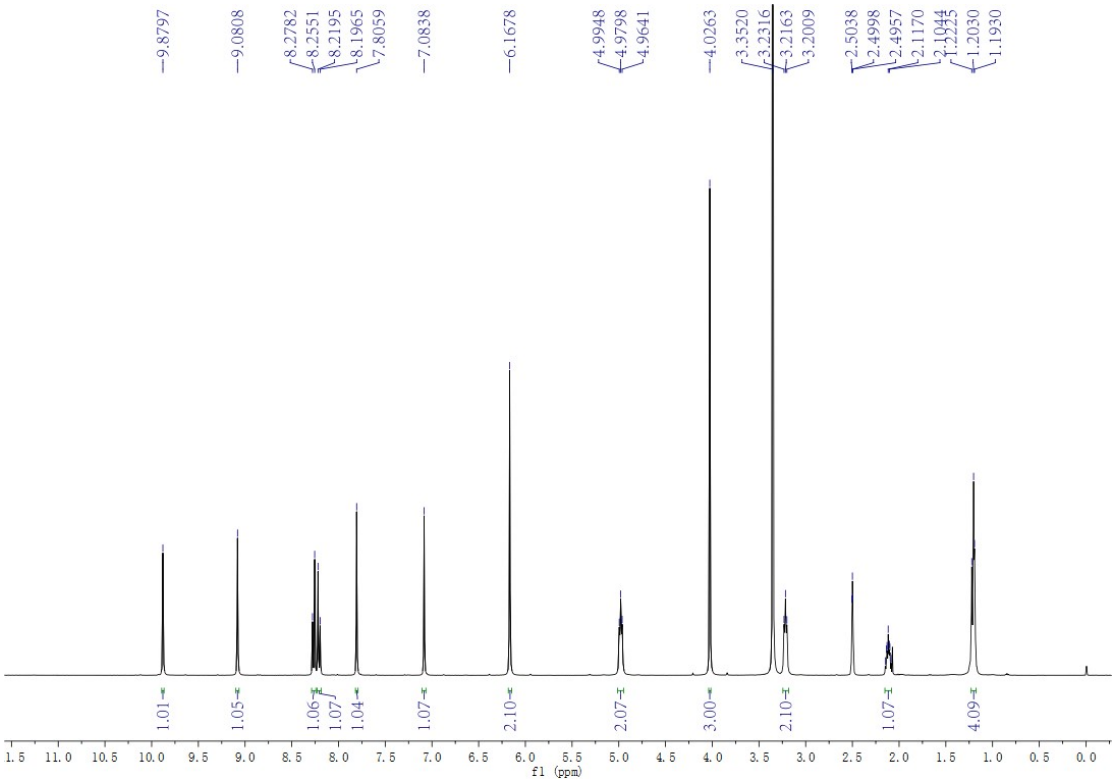
^{1}H NMR Spectra of Compound **B8**



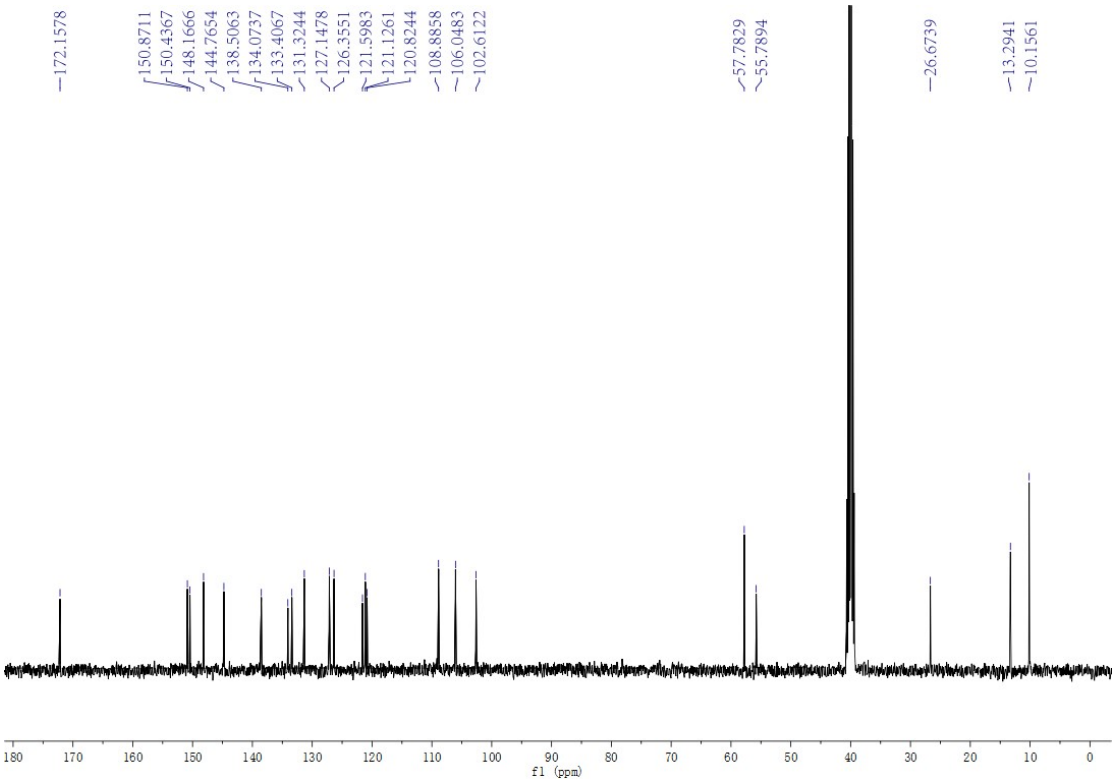
^{13}C NMR Spectra of Compound **B8**



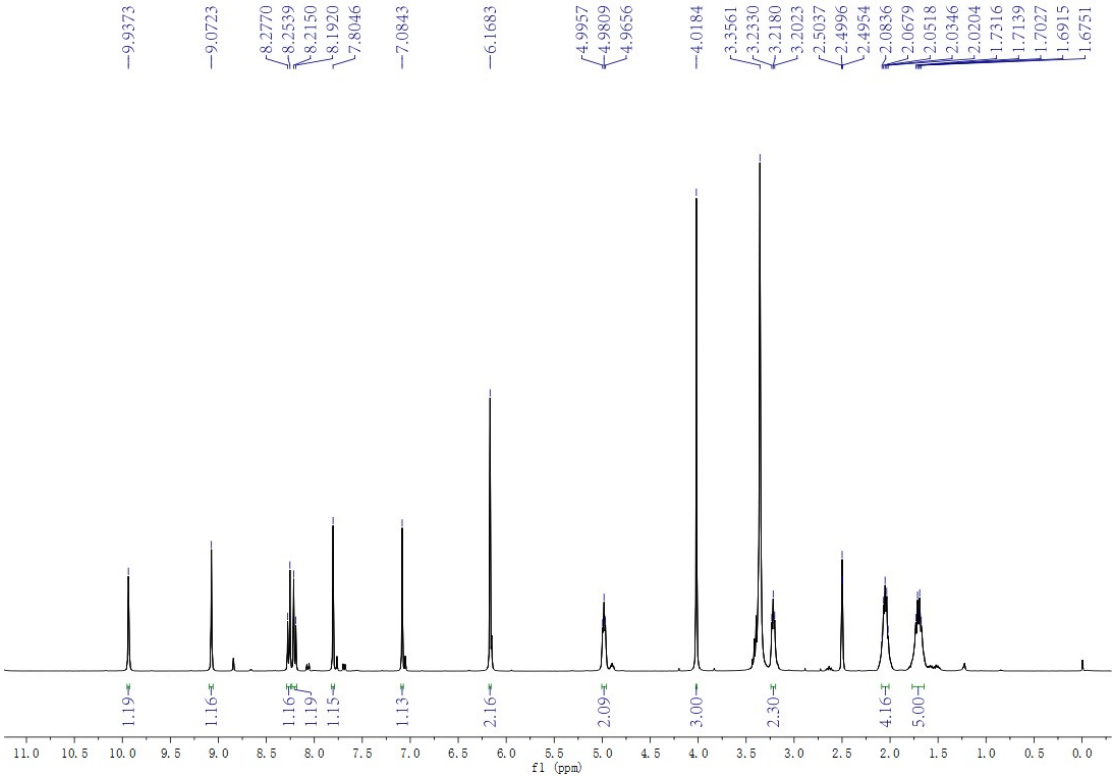
^{1}H NMR Spectra of Compound **B9**



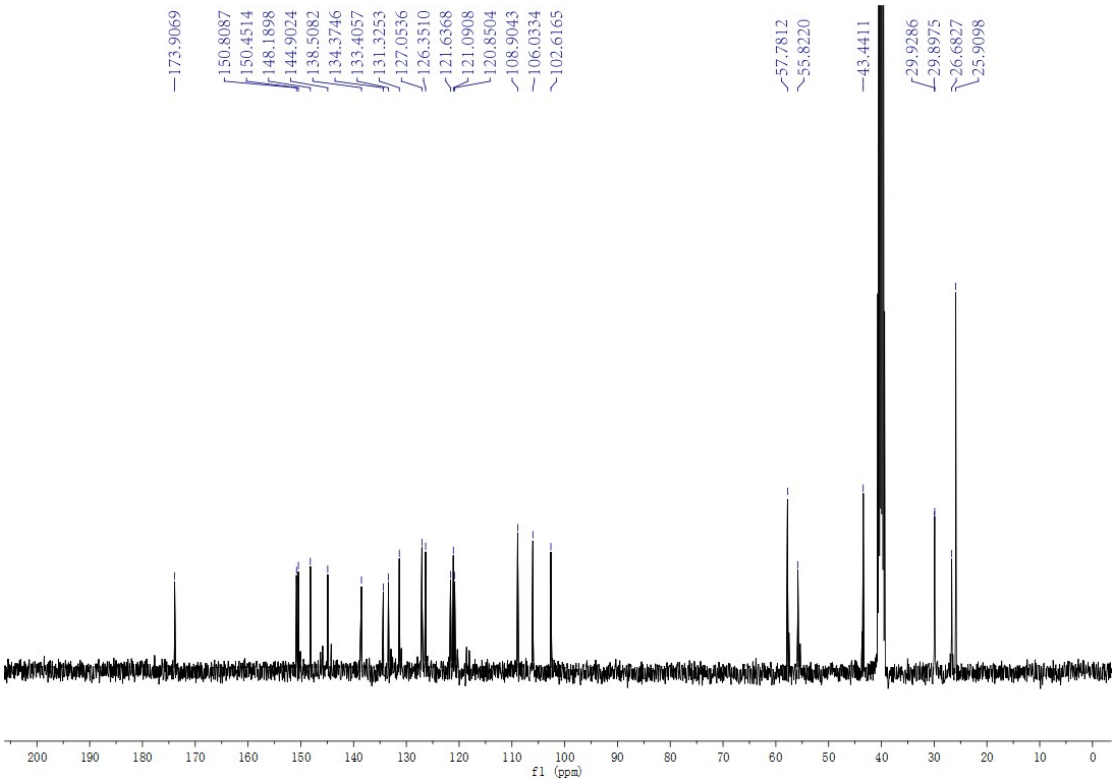
^{13}C NMR Spectra of Compound **B9**



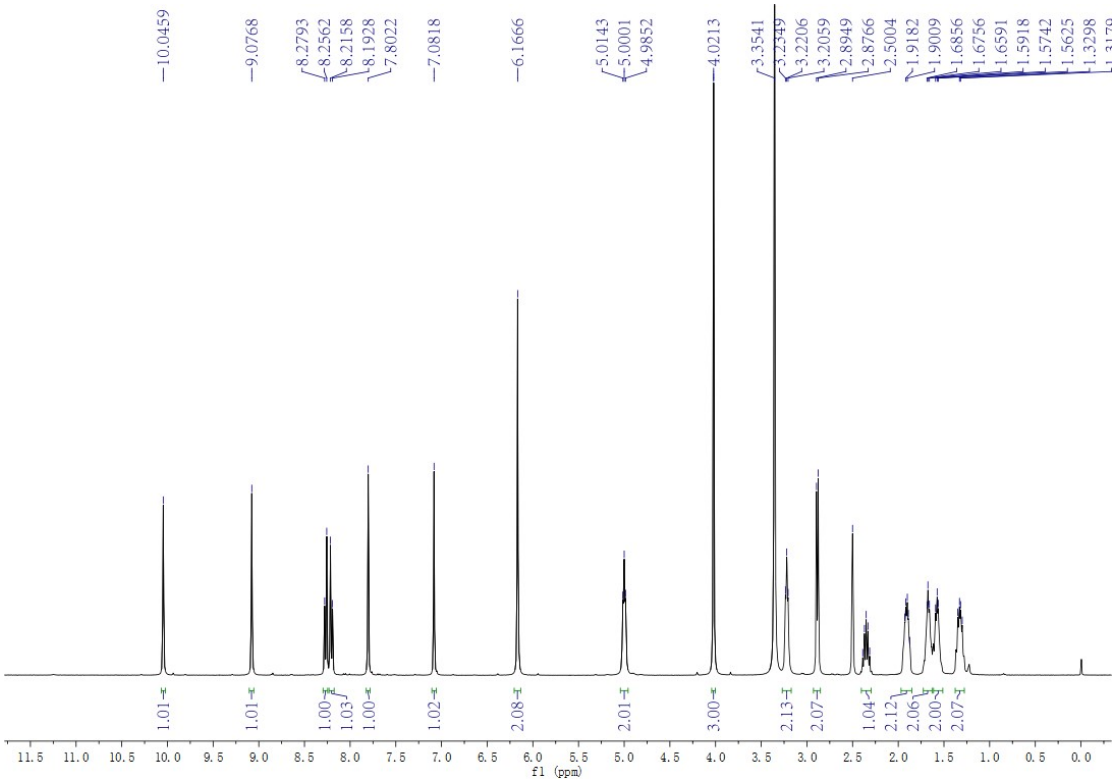
^{1}H NMR Spectra of Compound **B10**



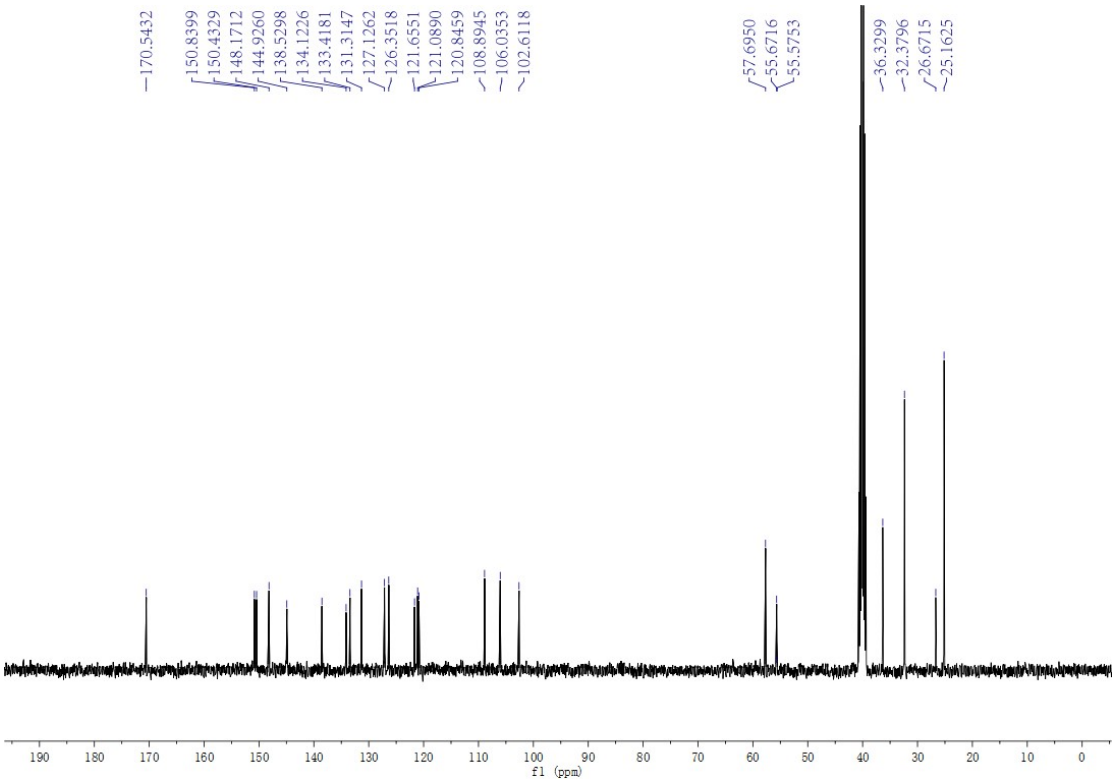
^{13}C NMR Spectra of Compound **B10**



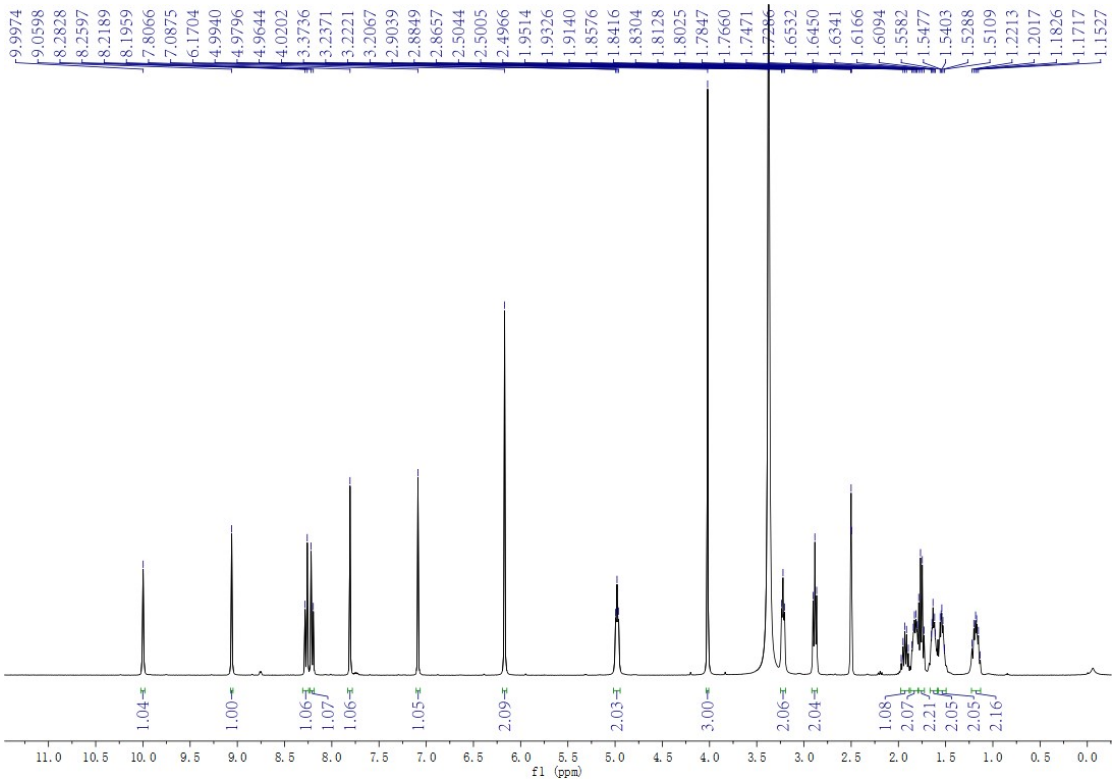
[1]H NMR Spectra of Compound **B11**



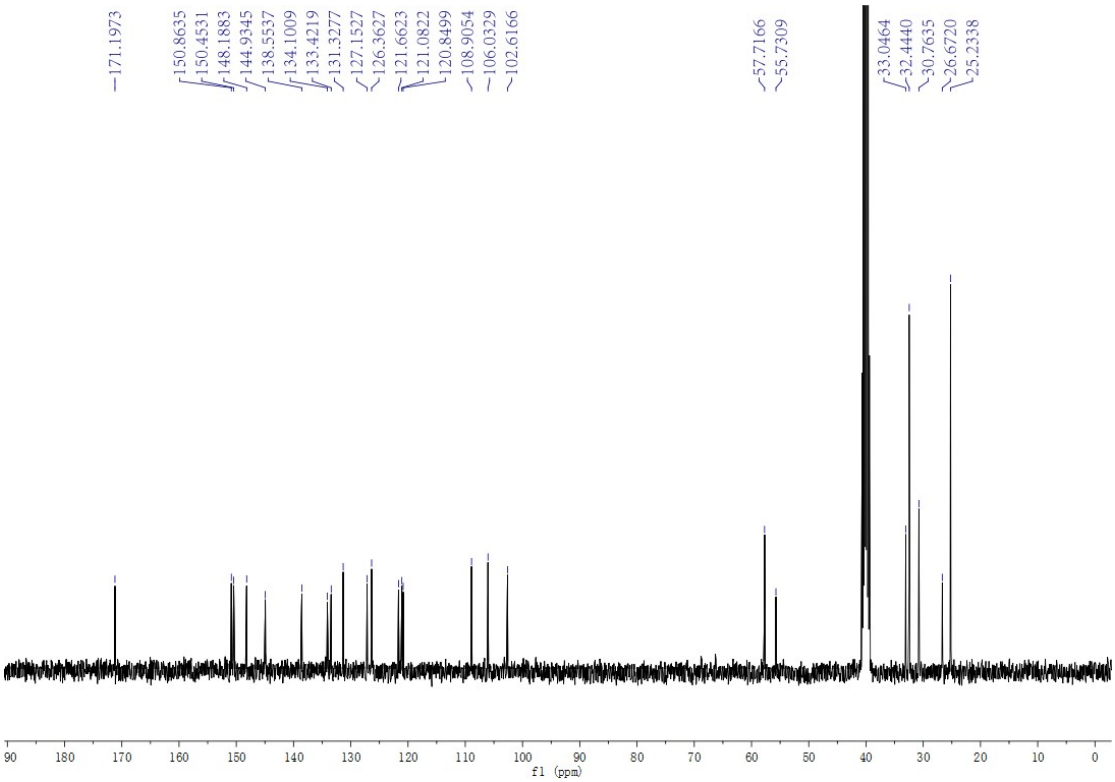
[13]C NMR Spectra of Compound **B11**



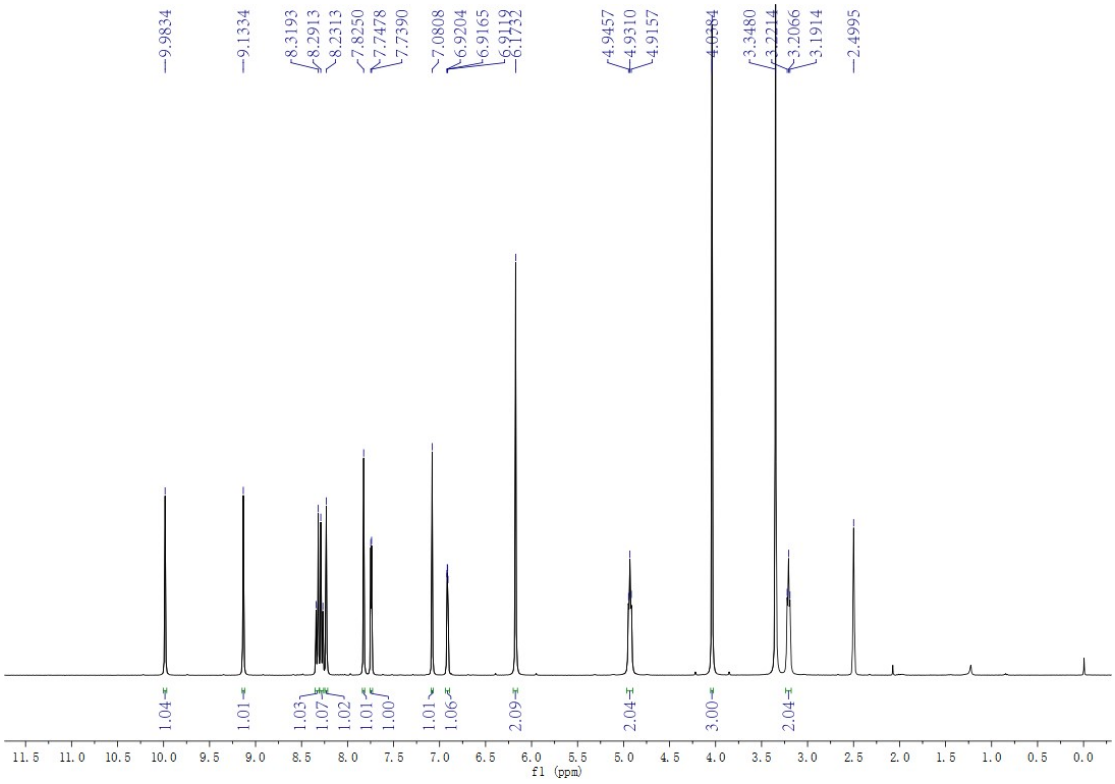
^{1}H NMR Spectra of Compound **B12**



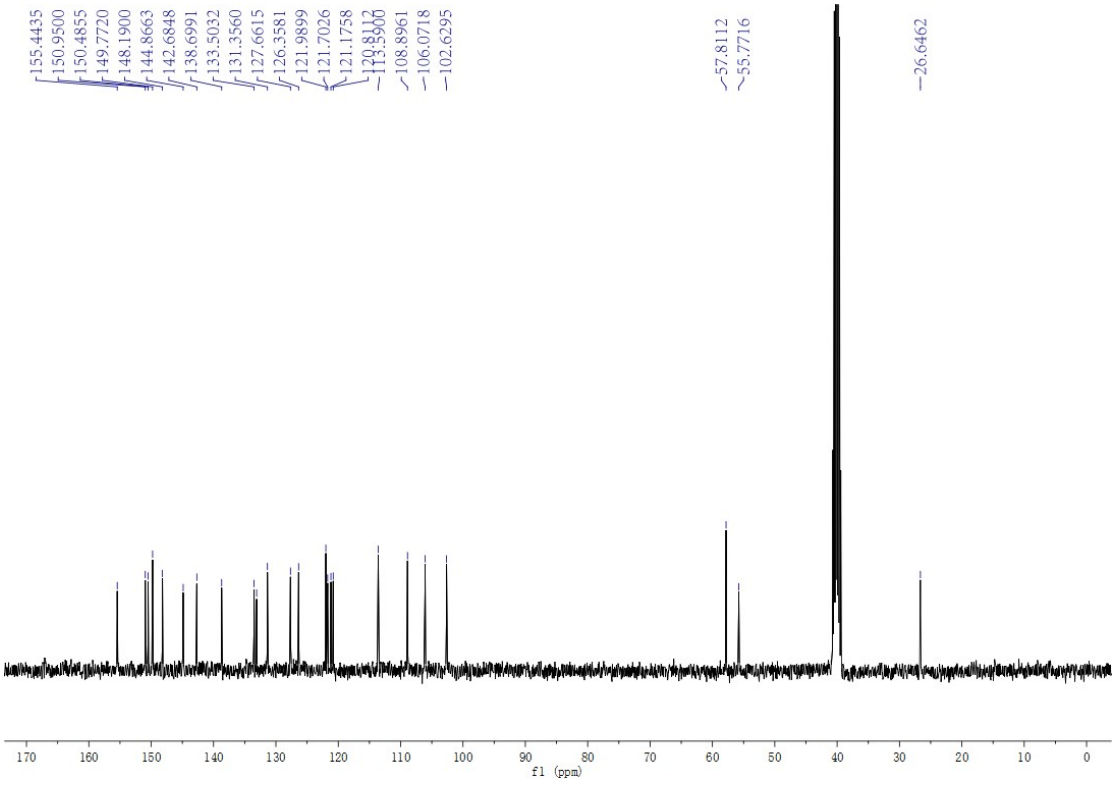
^{13}C NMR Spectra of Compound **B12**



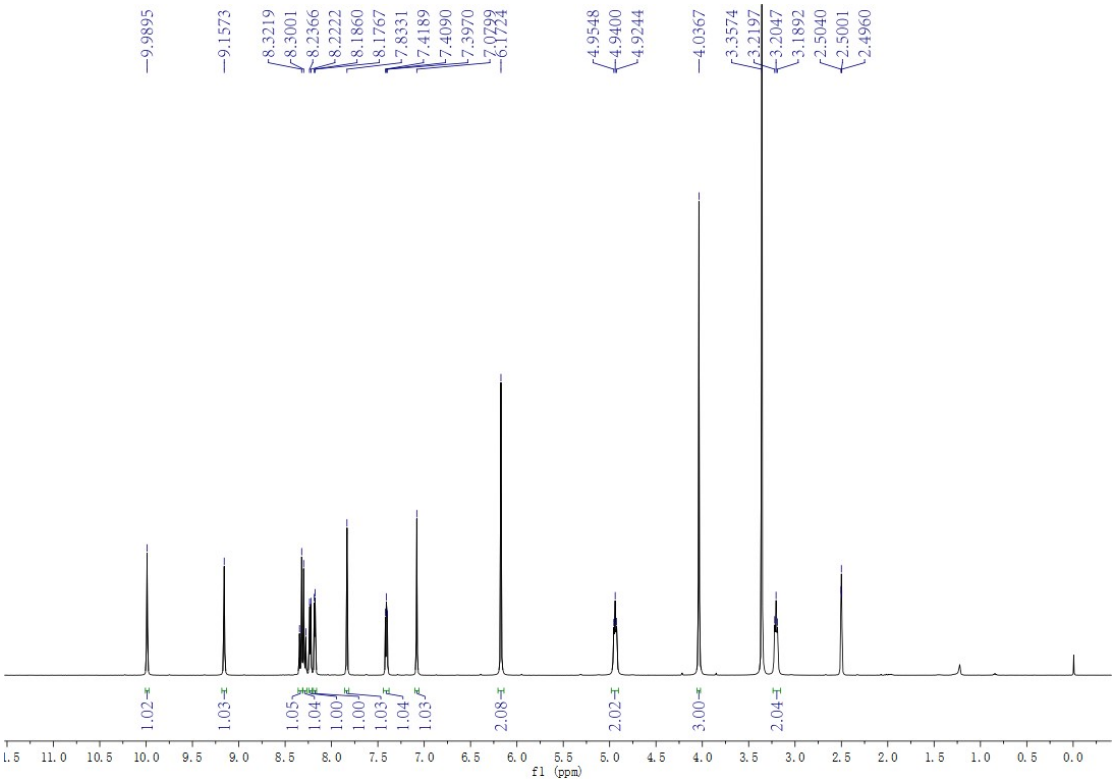
^{1}H NMR Spectra of Compound **B13**



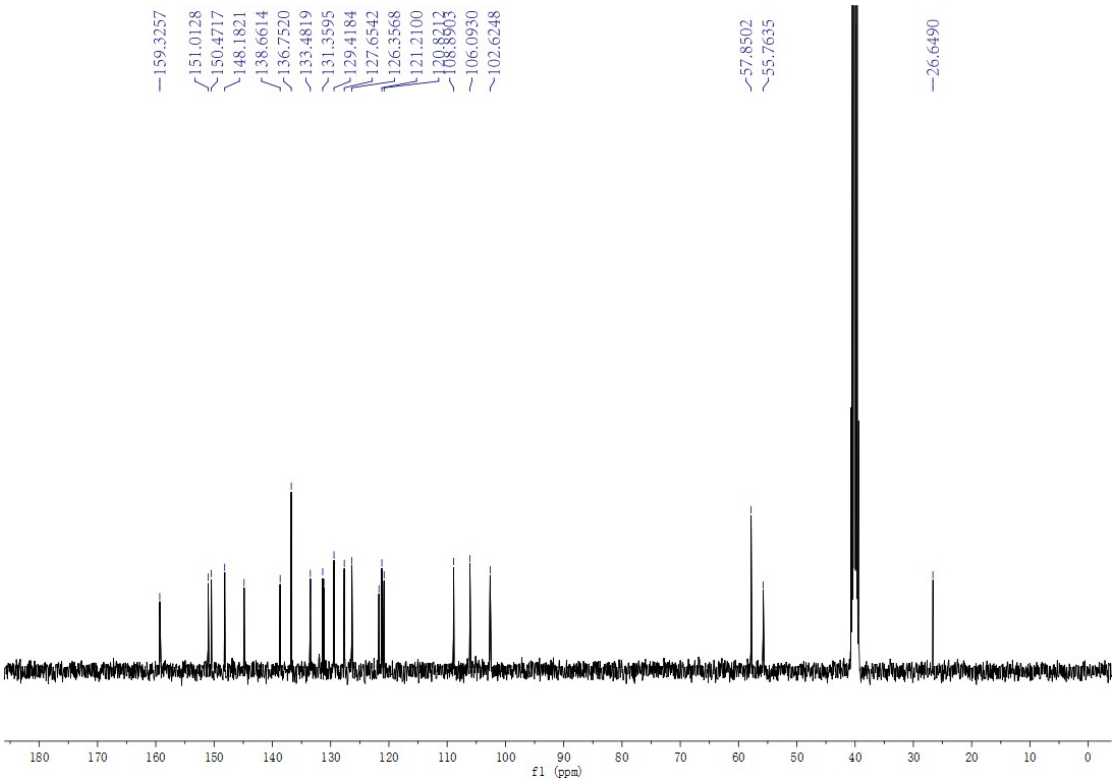
^{13}C NMR Spectra of Compound **B13**



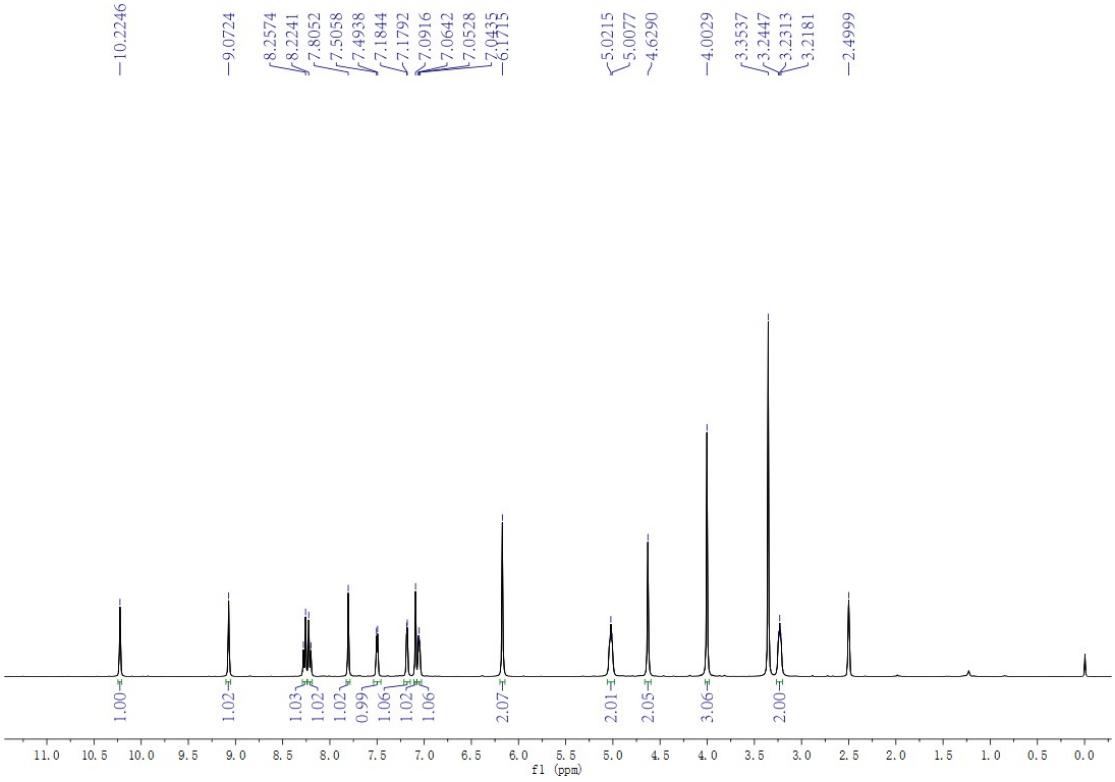
^{1}H NMR Spectra of Compound **B14**



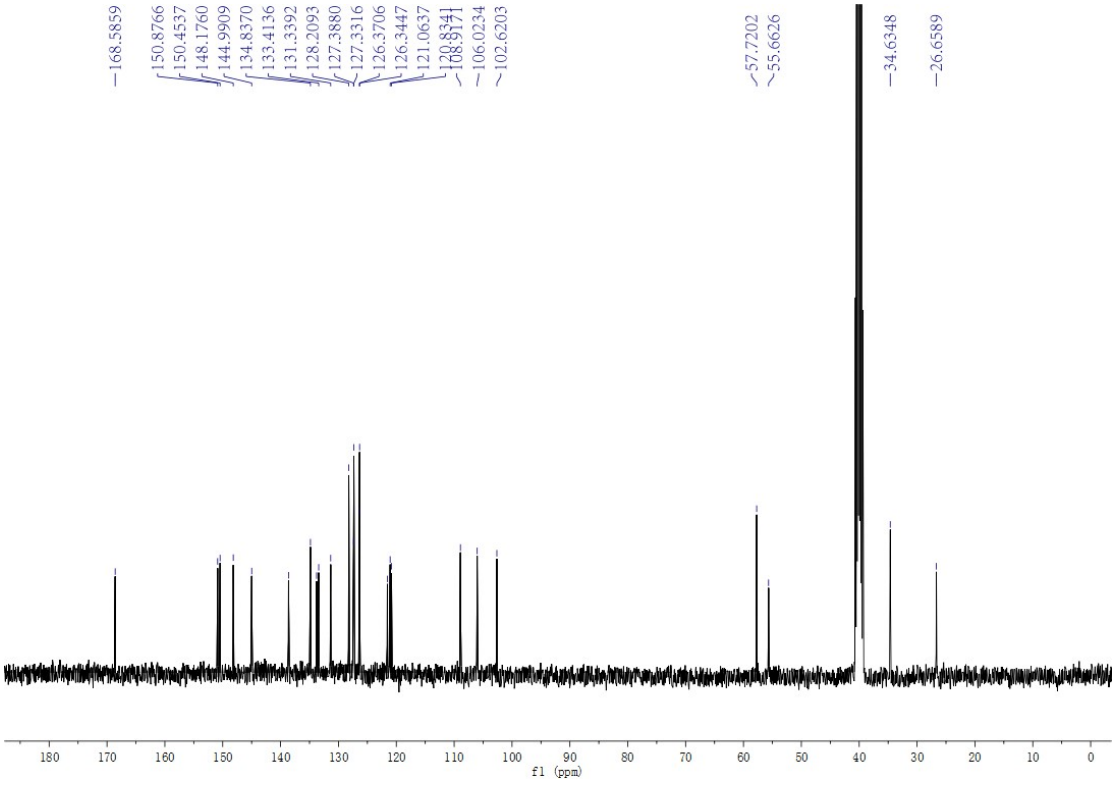
^{13}C NMR Spectra of Compound **B14**



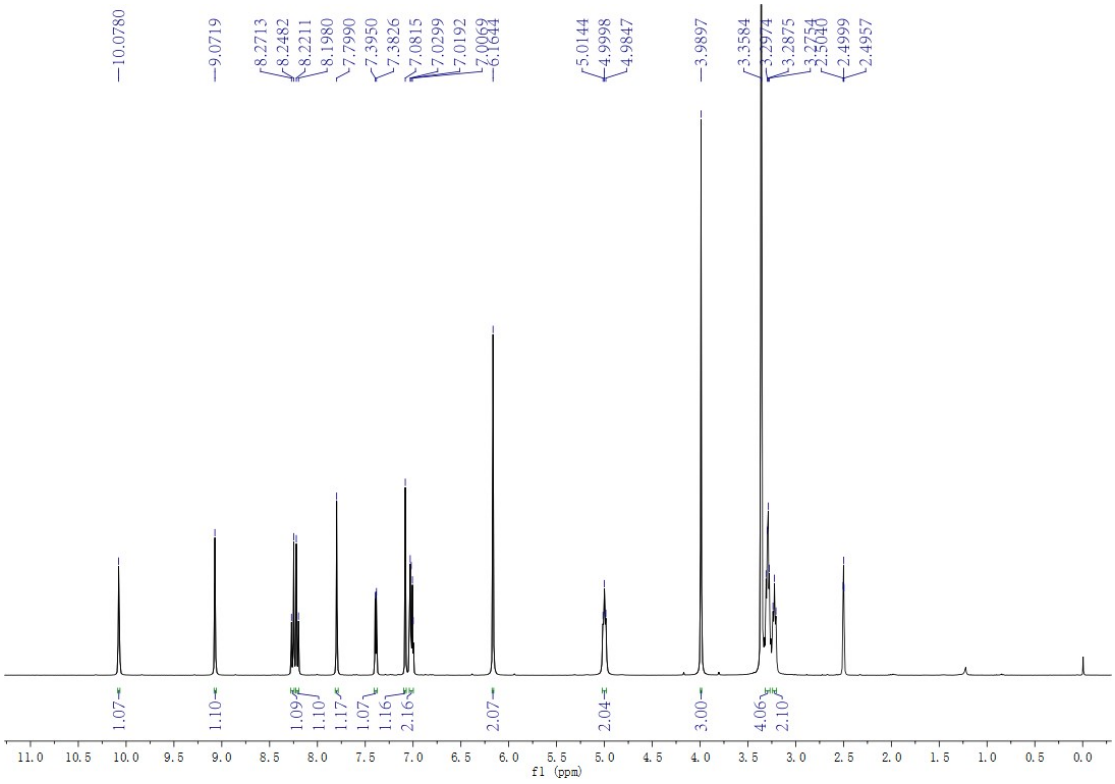
^{1}H NMR Spectra of Compound **B15**



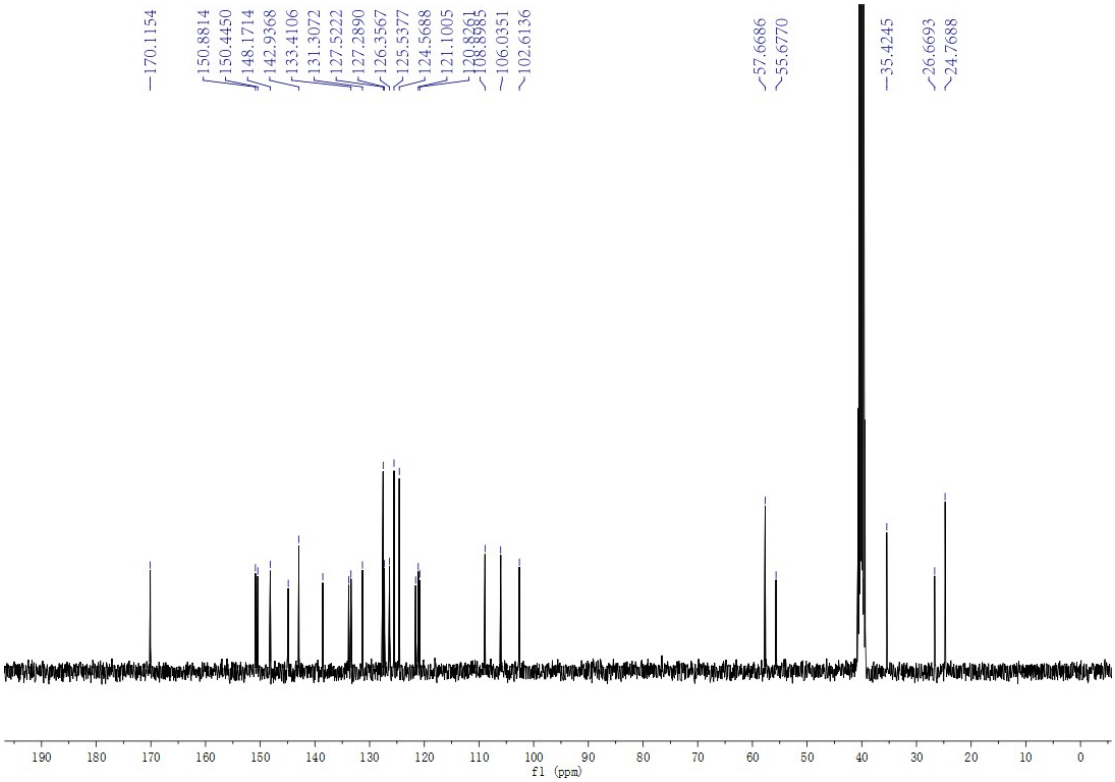
^{13}C NMR Spectra of Compound **B15**



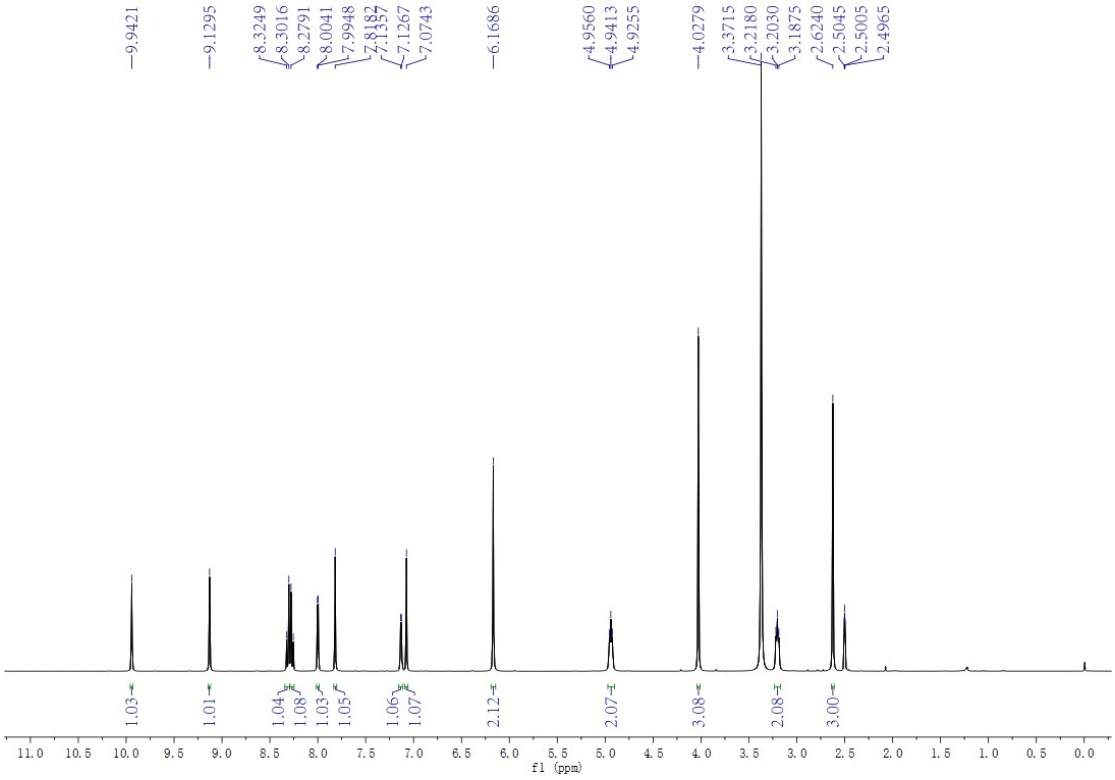
^{1}H NMR Spectra of Compound **B16**



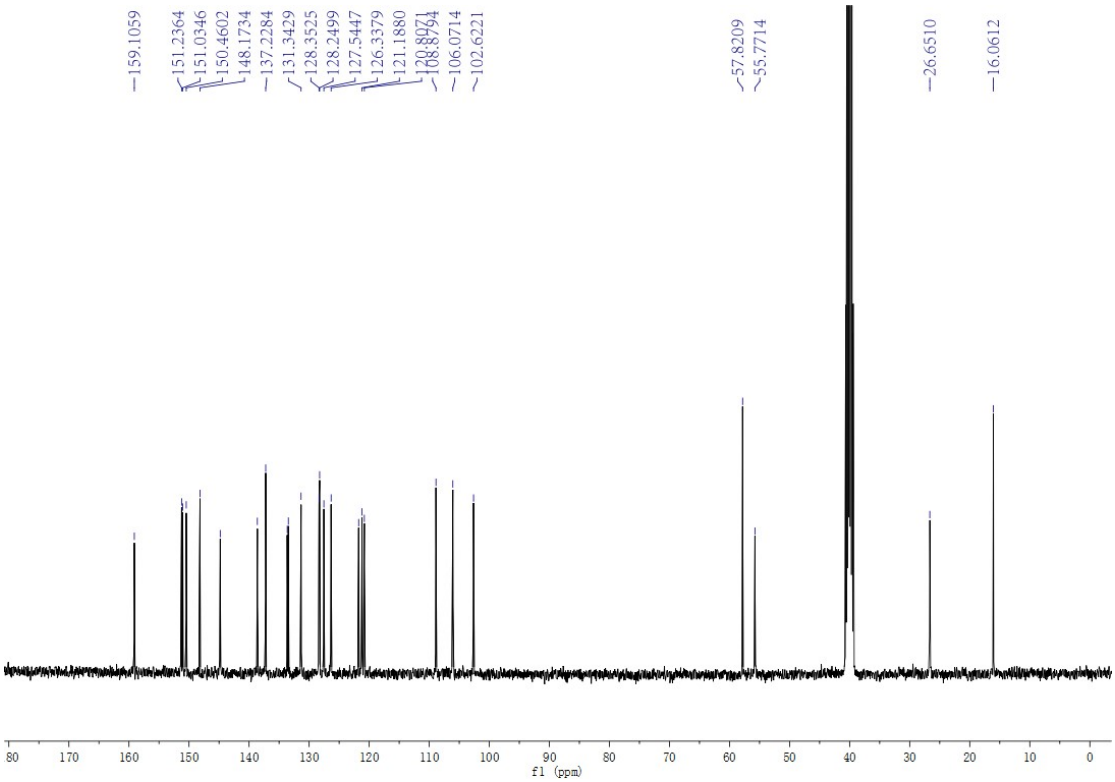
^{13}C NMR Spectra of Compound **B16**



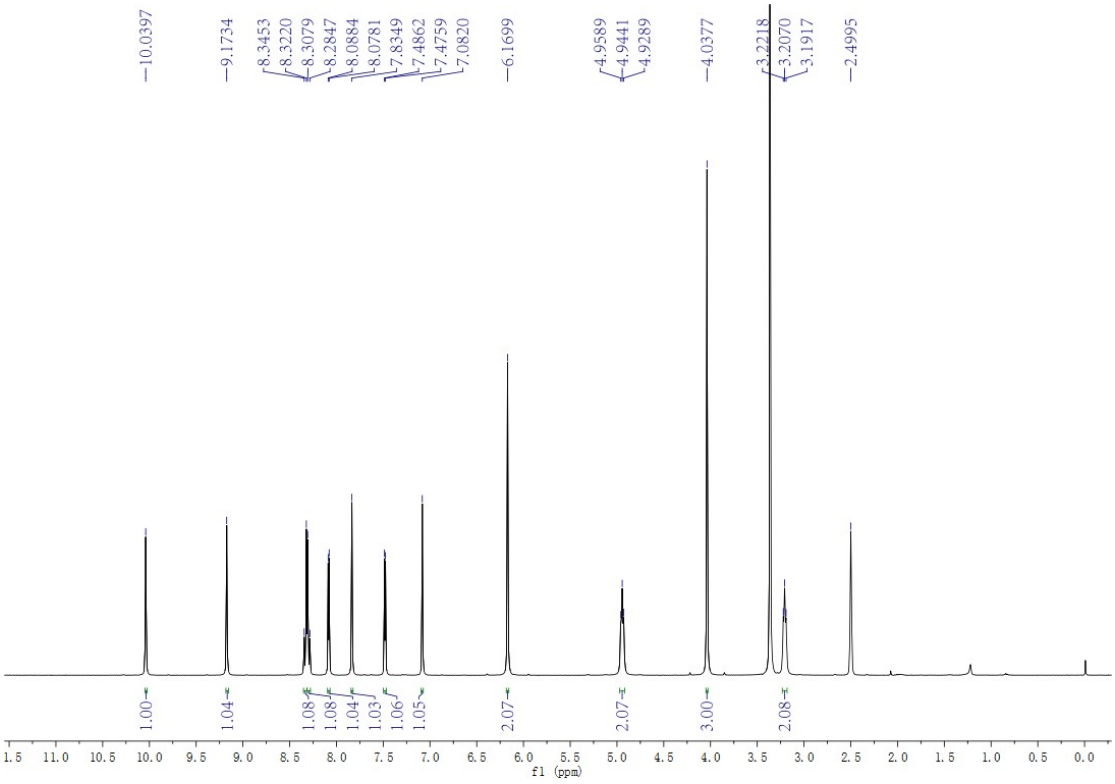
^{1}H NMR Spectra of Compound **B17**



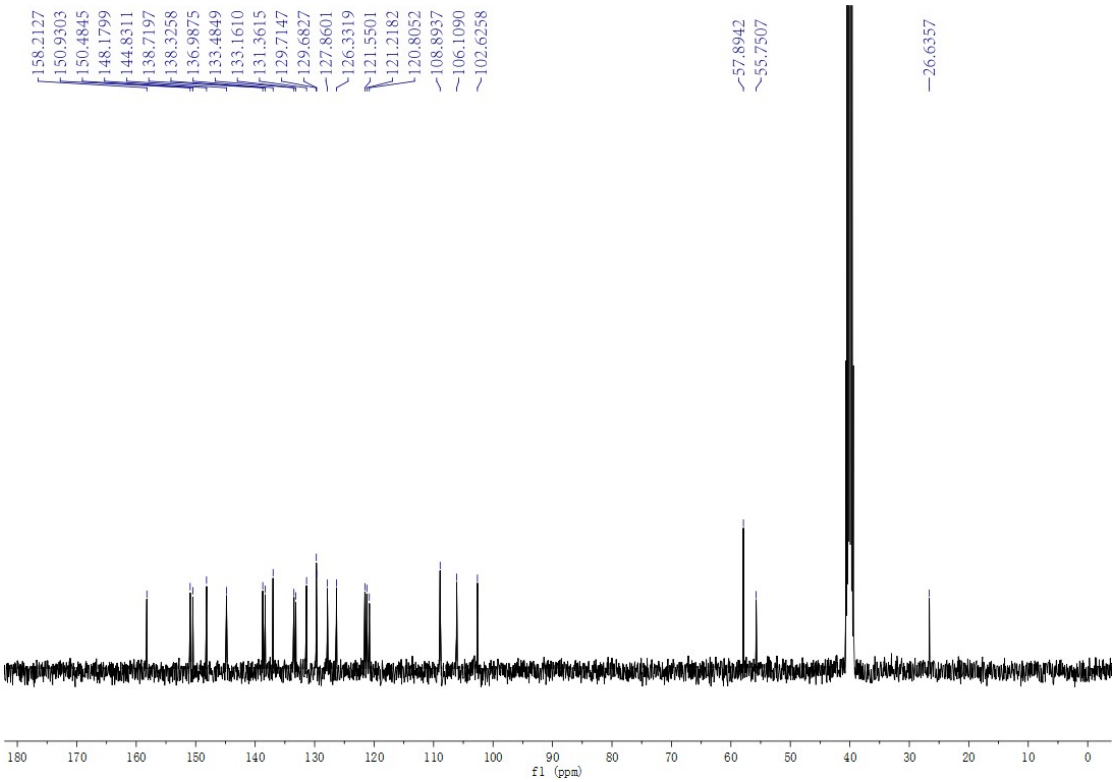
^{13}C NMR Spectra of Compound **B17**



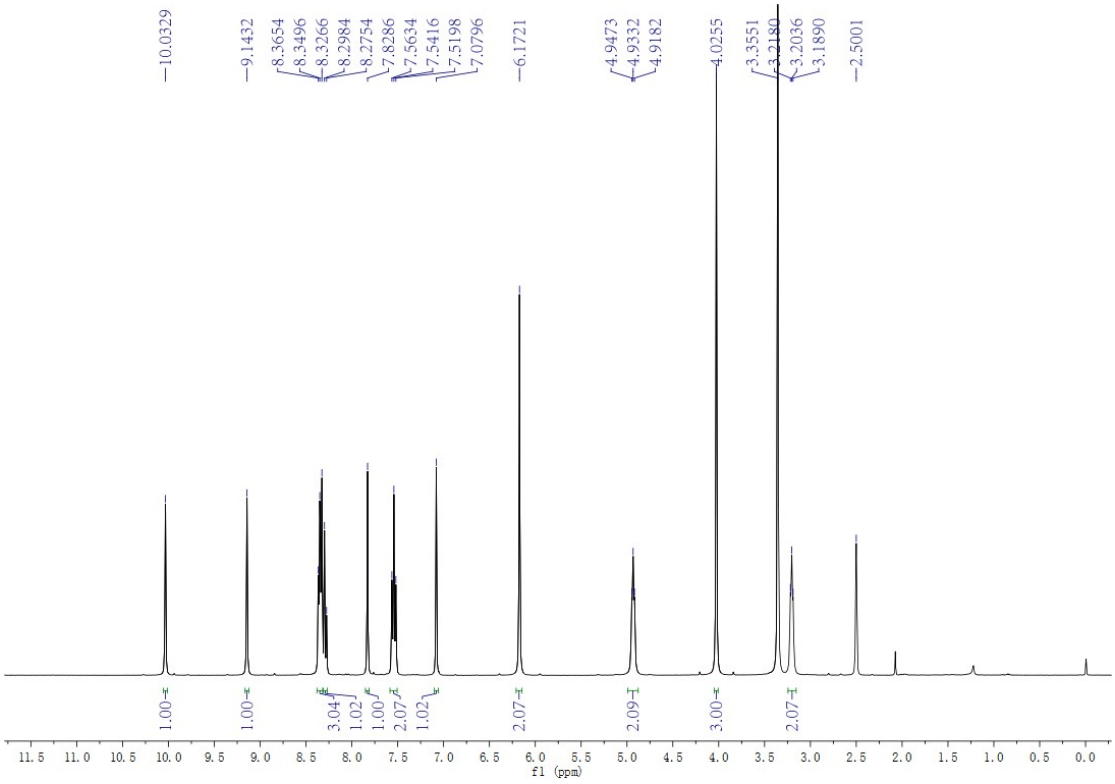
^{1}H NMR Spectra of Compound **B18**



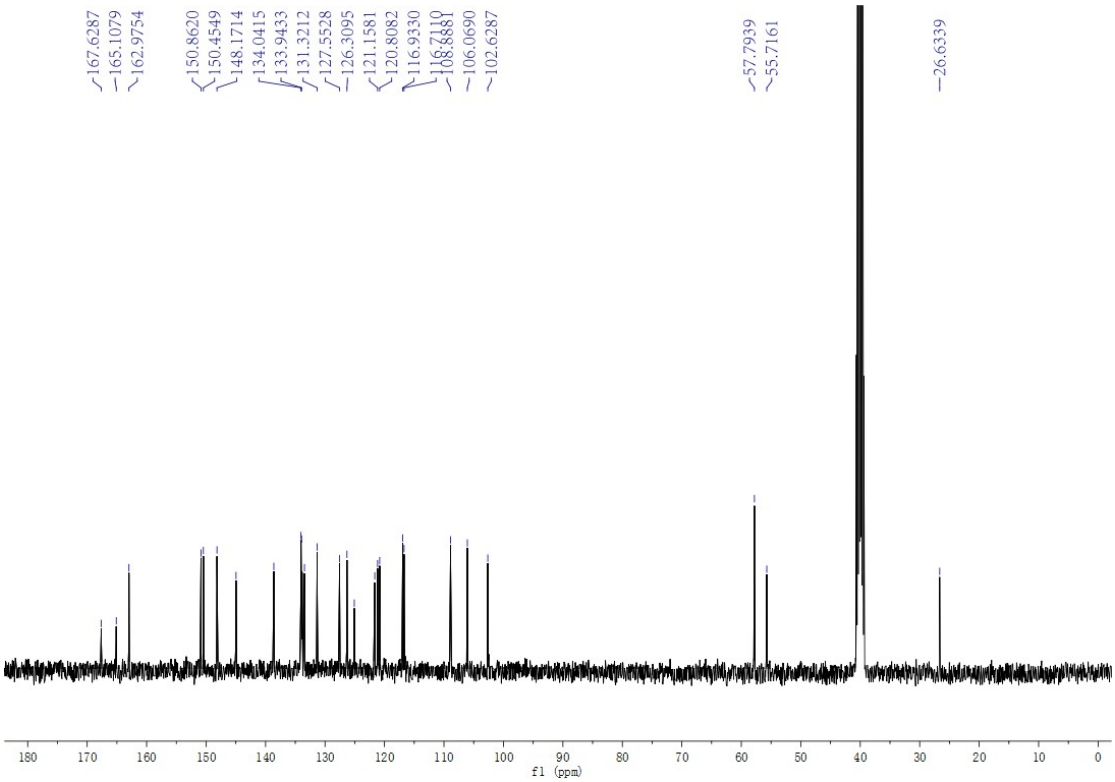
^{13}C NMR Spectra of Compound **B18**



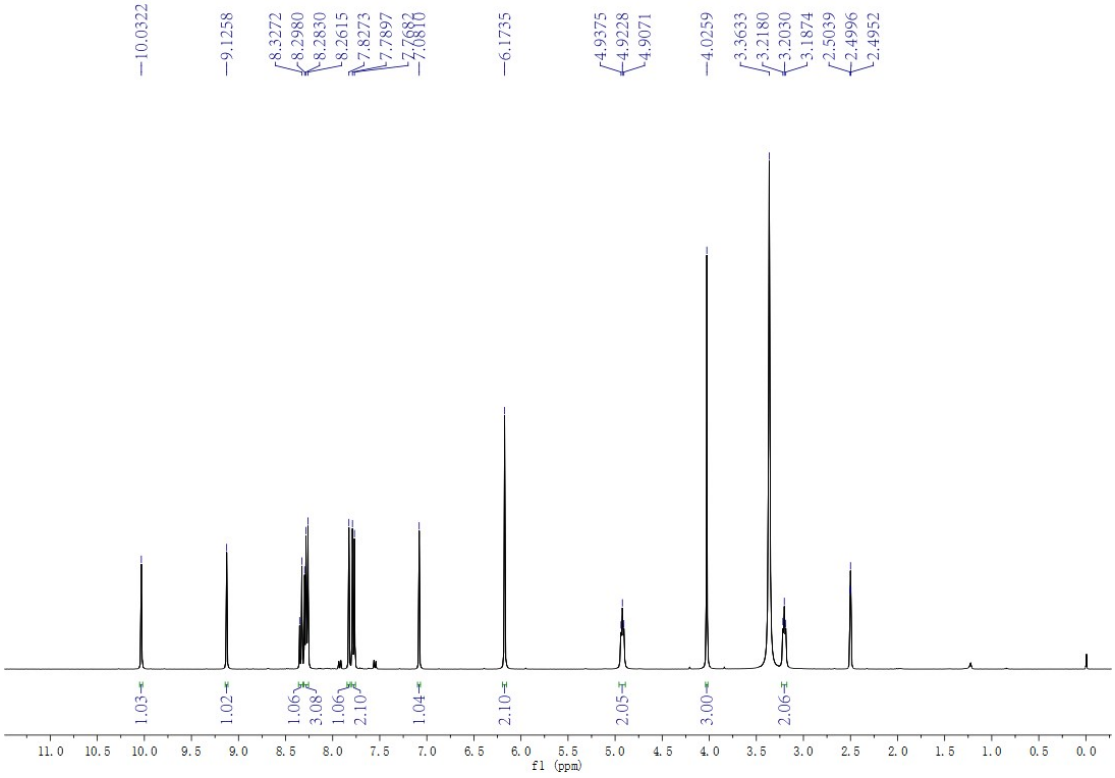
^{1}H NMR Spectra of Compound **B19**



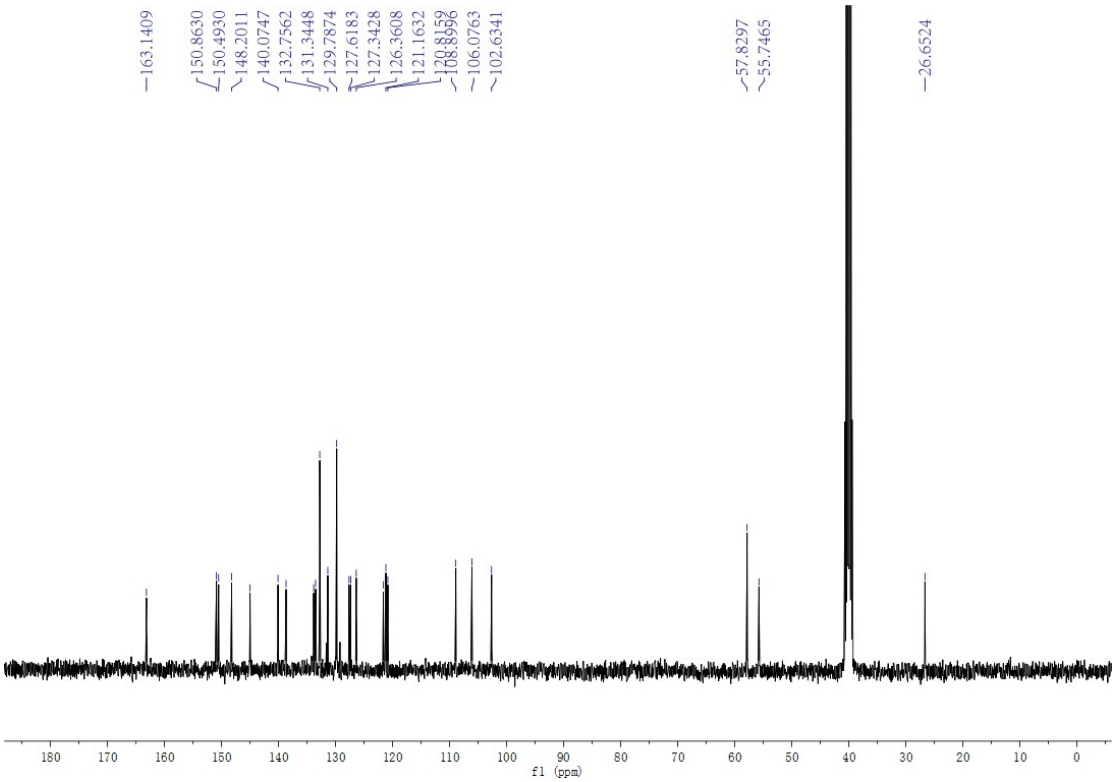
^{13}C NMR Spectra of Compound **B19**



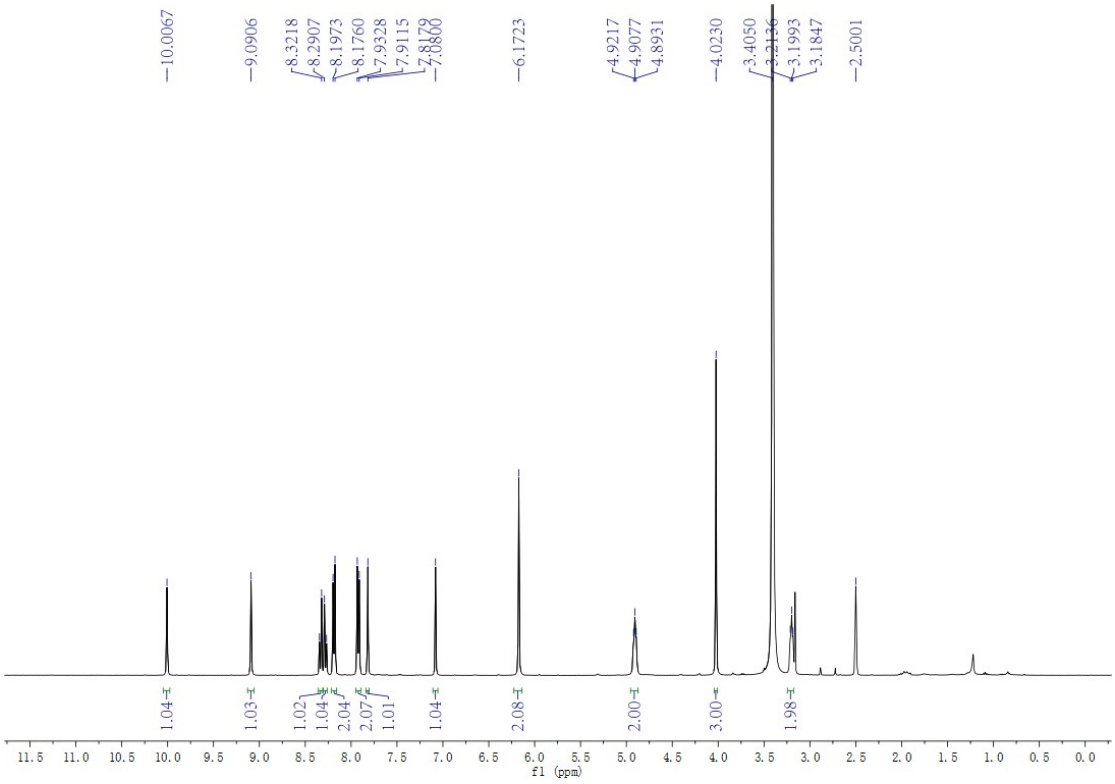
^{1}H NMR Spectra of Compound **B20**



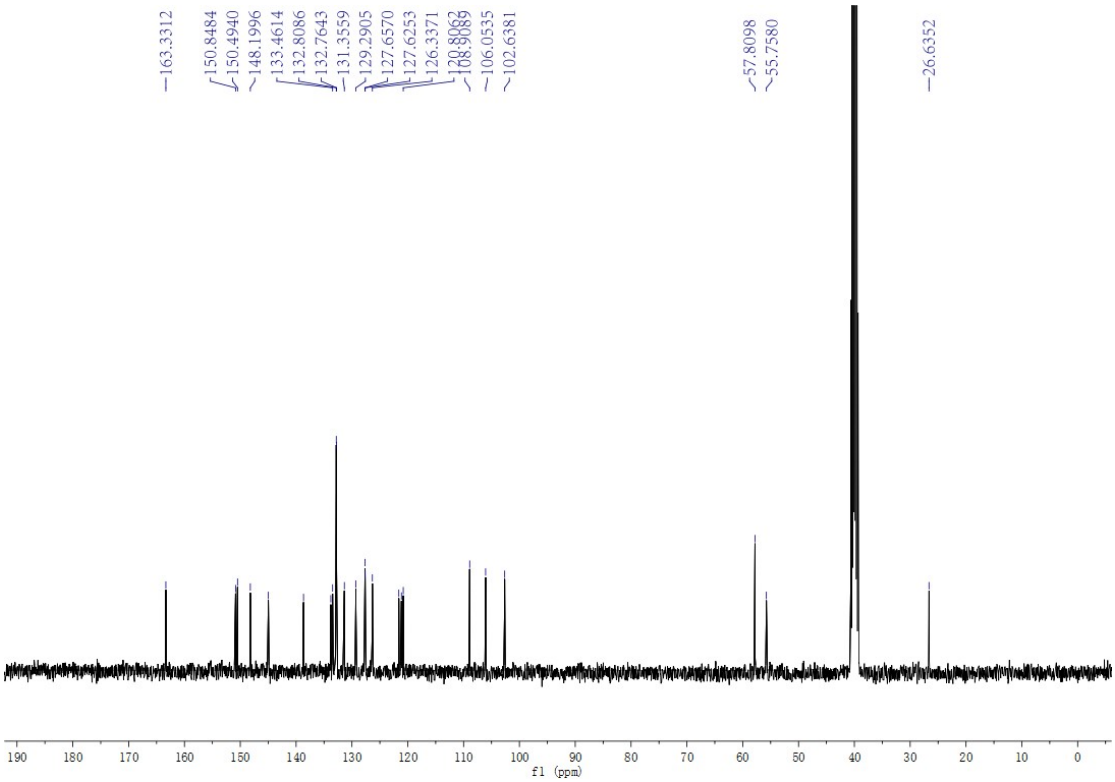
^{13}C NMR Spectra of Compound **B20**



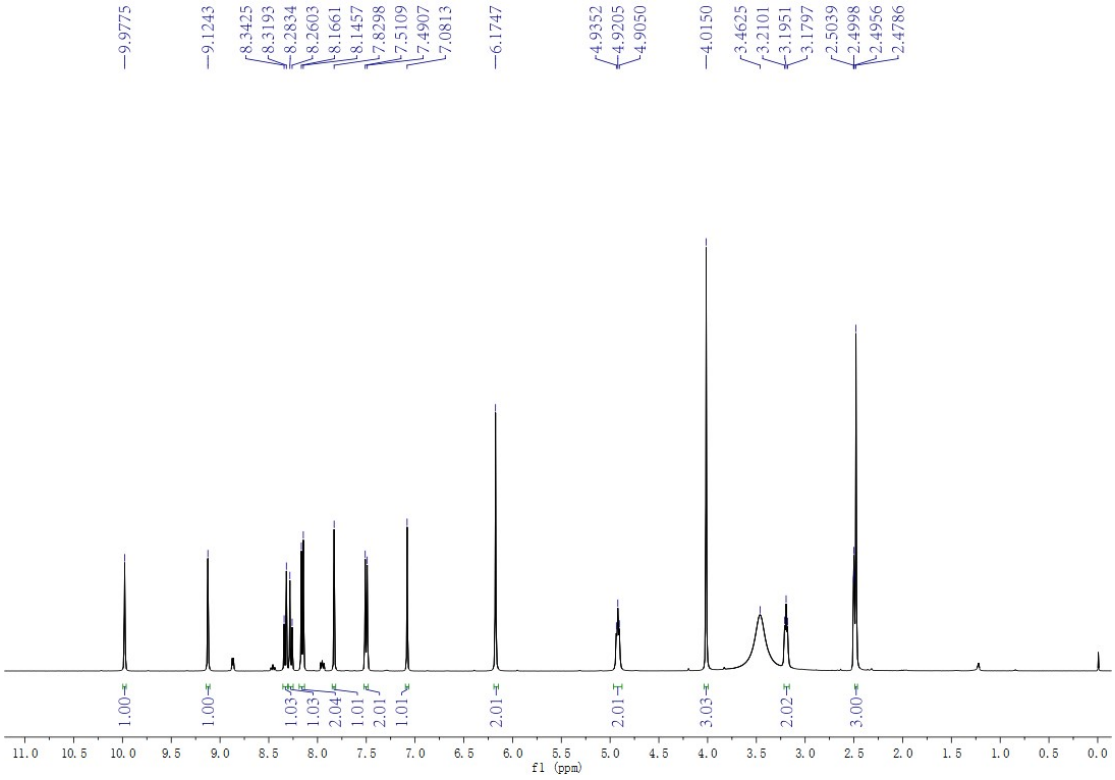
[1]H NMR Spectra of Compound **B21**



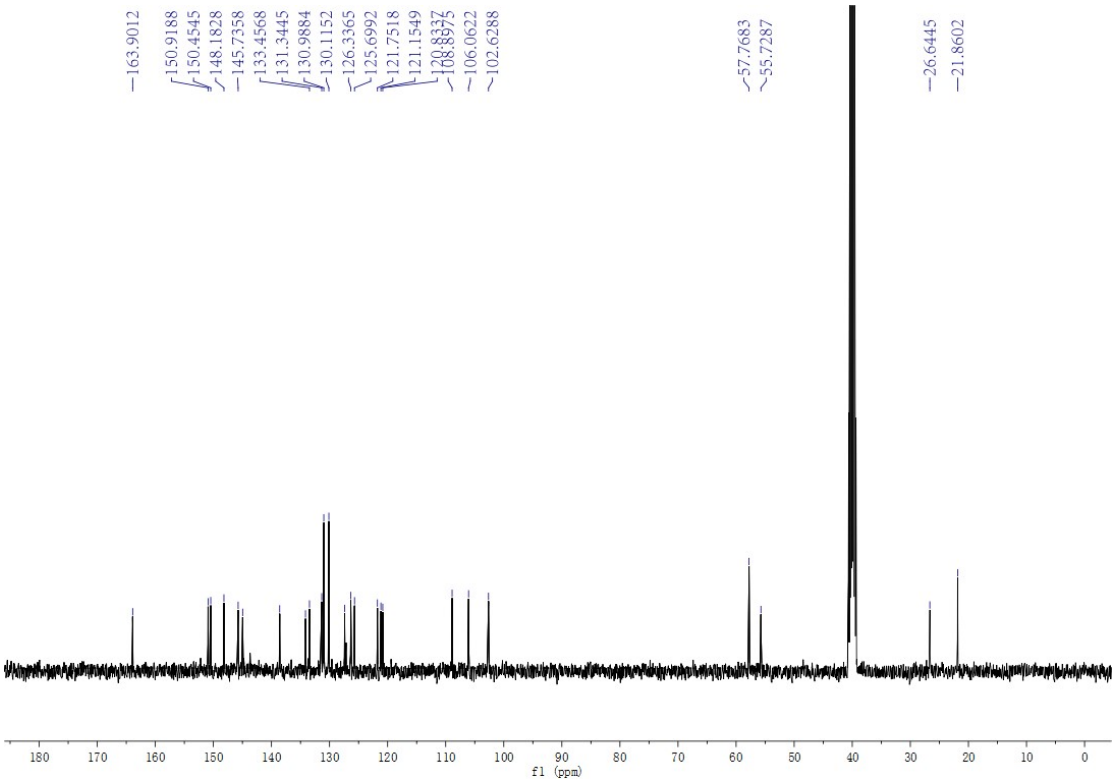
[13]C NMR Spectra of Compound **B21**



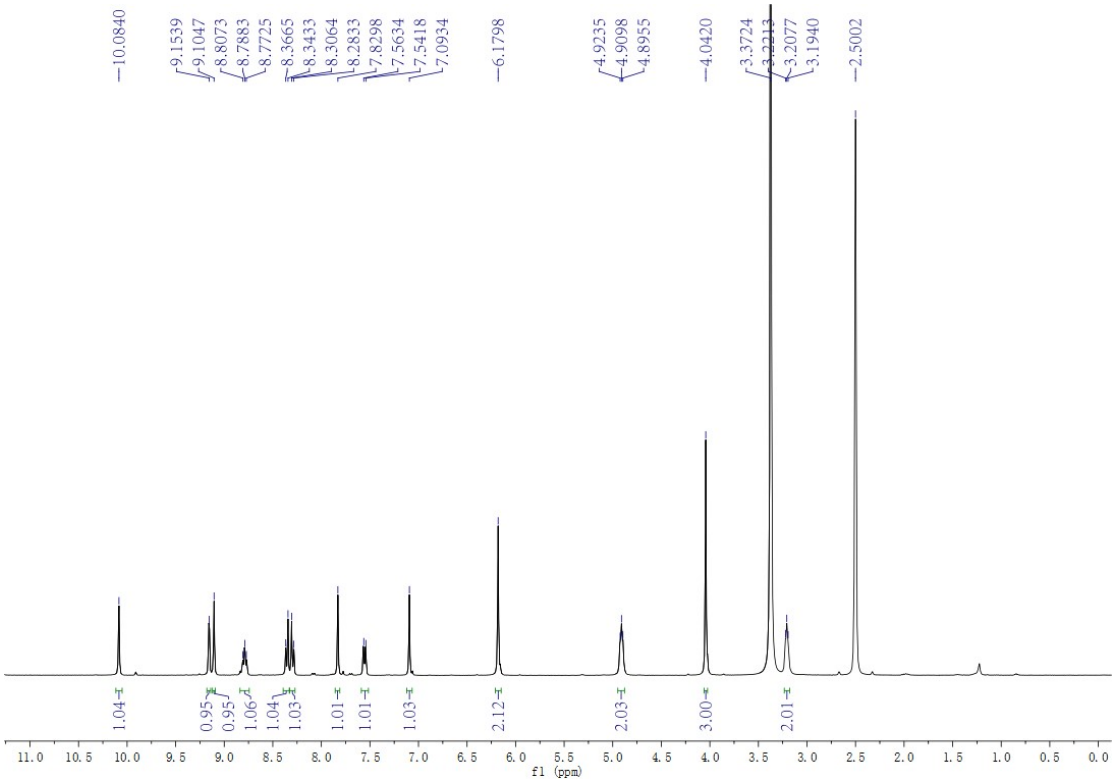
^{1}H NMR Spectra of Compound **B22**



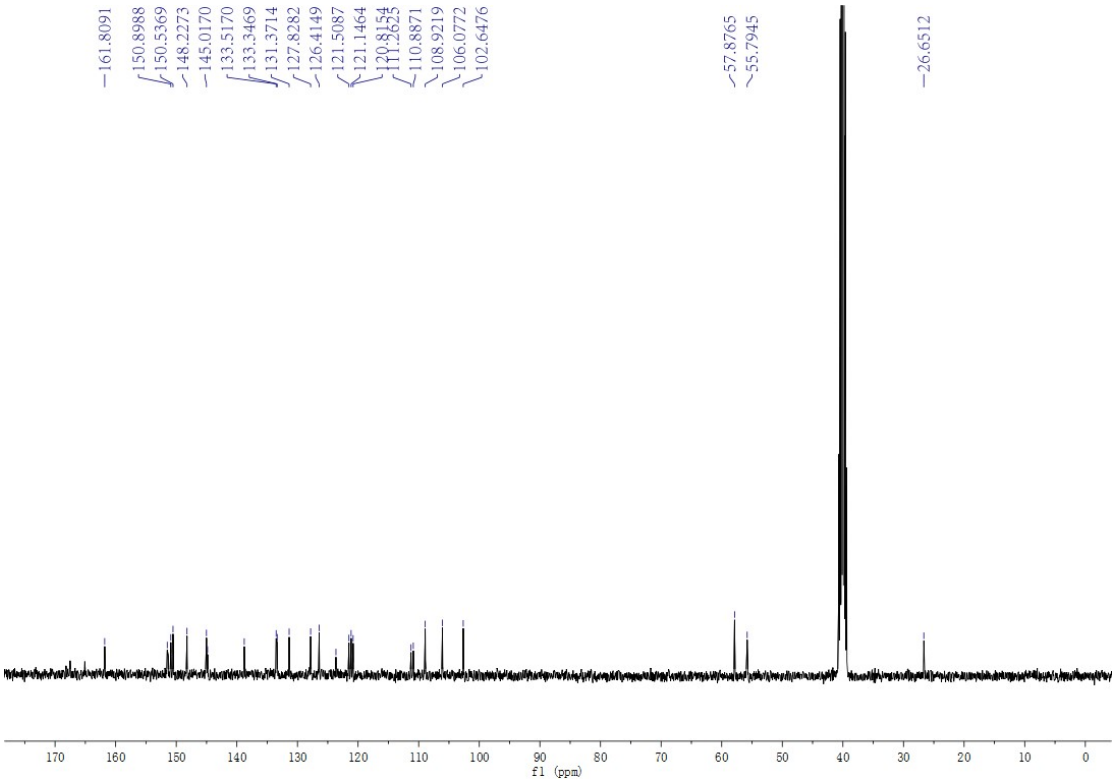
^{13}C NMR Spectra of Compound **B22**



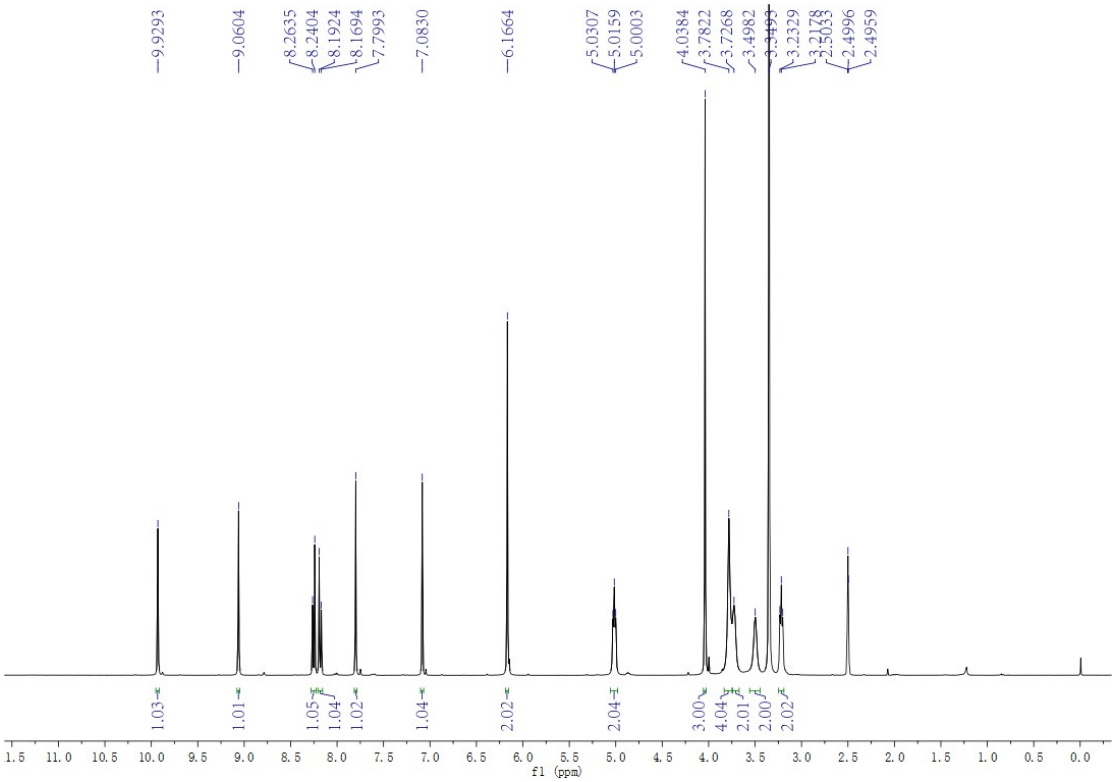
^{1}H NMR Spectra of Compound **B23**



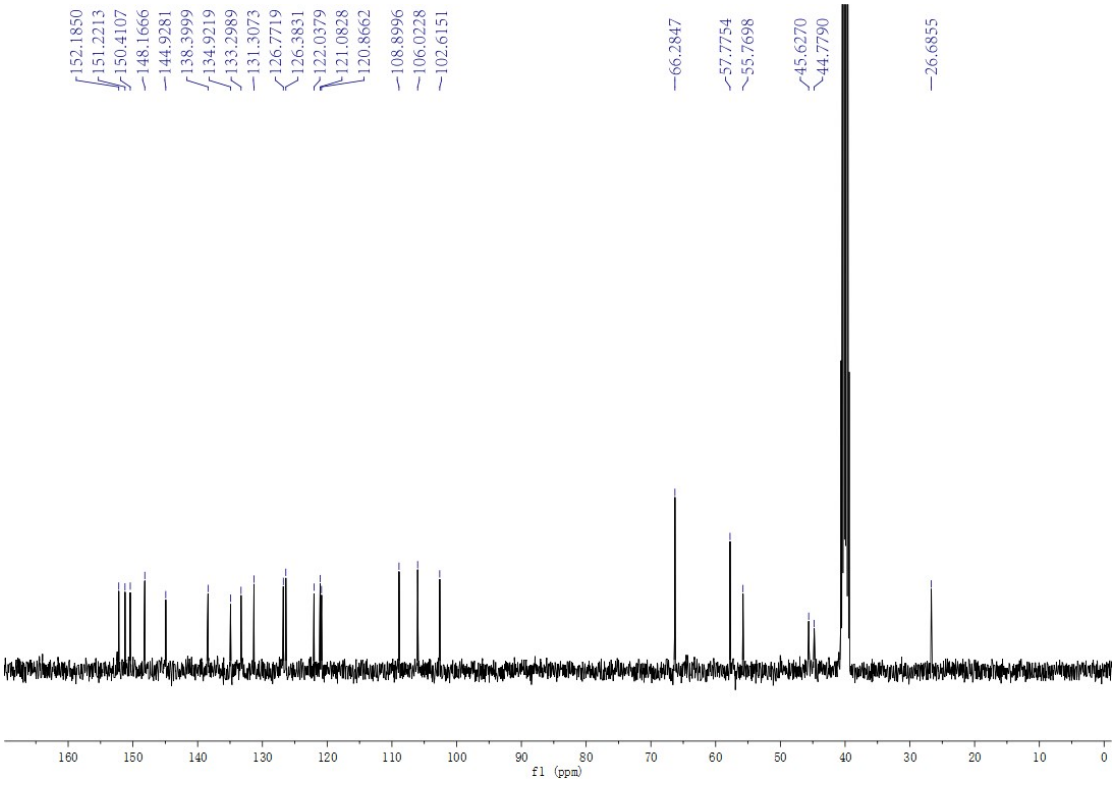
^{13}C NMR Spectra of Compound **B23**



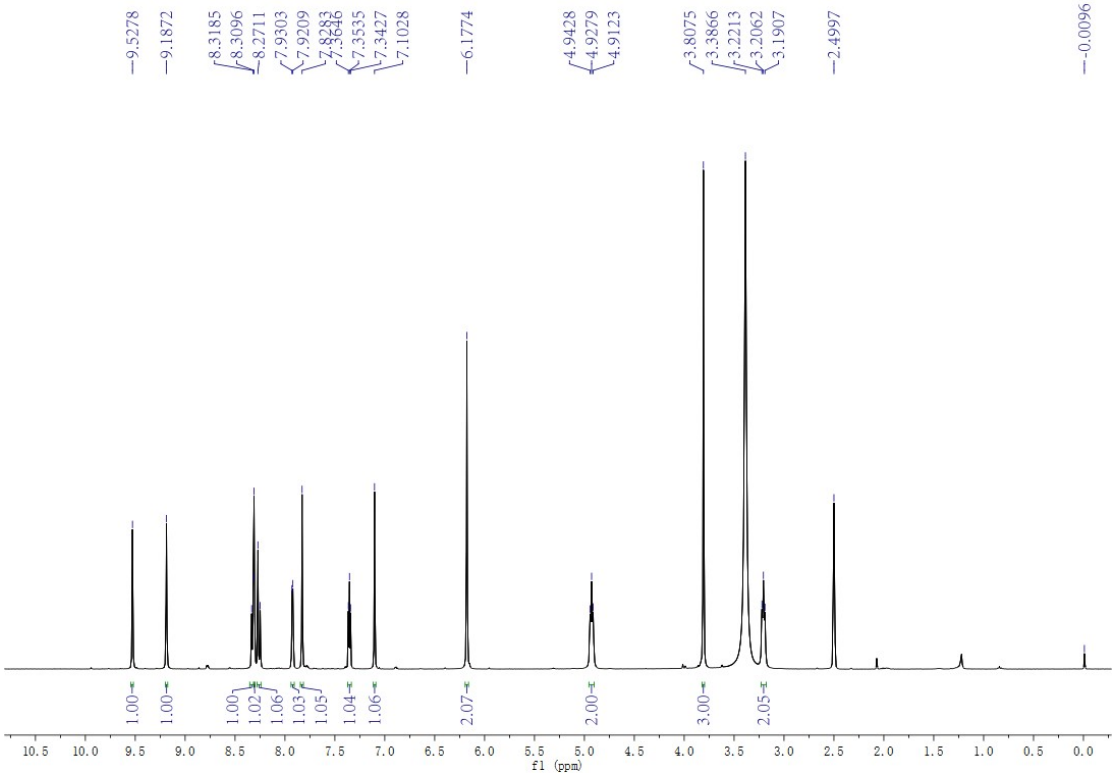
^{1}H NMR Spectra of Compound **B24**



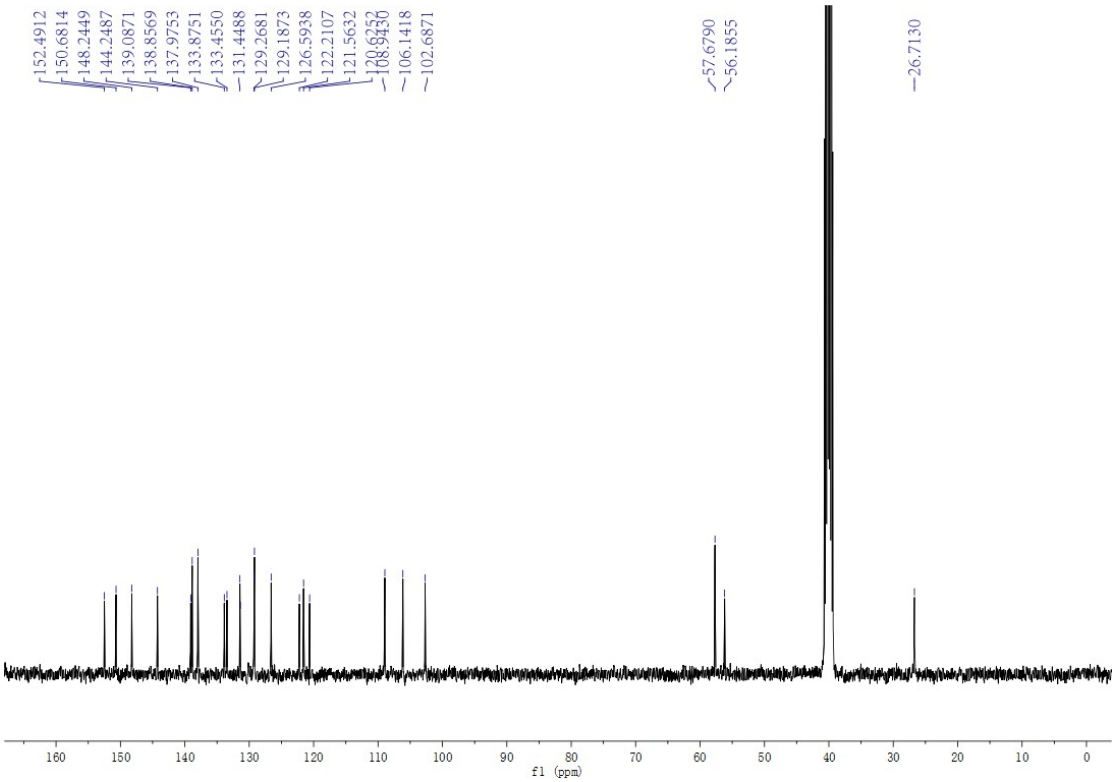
^{13}C NMR Spectra of Compound **B24**



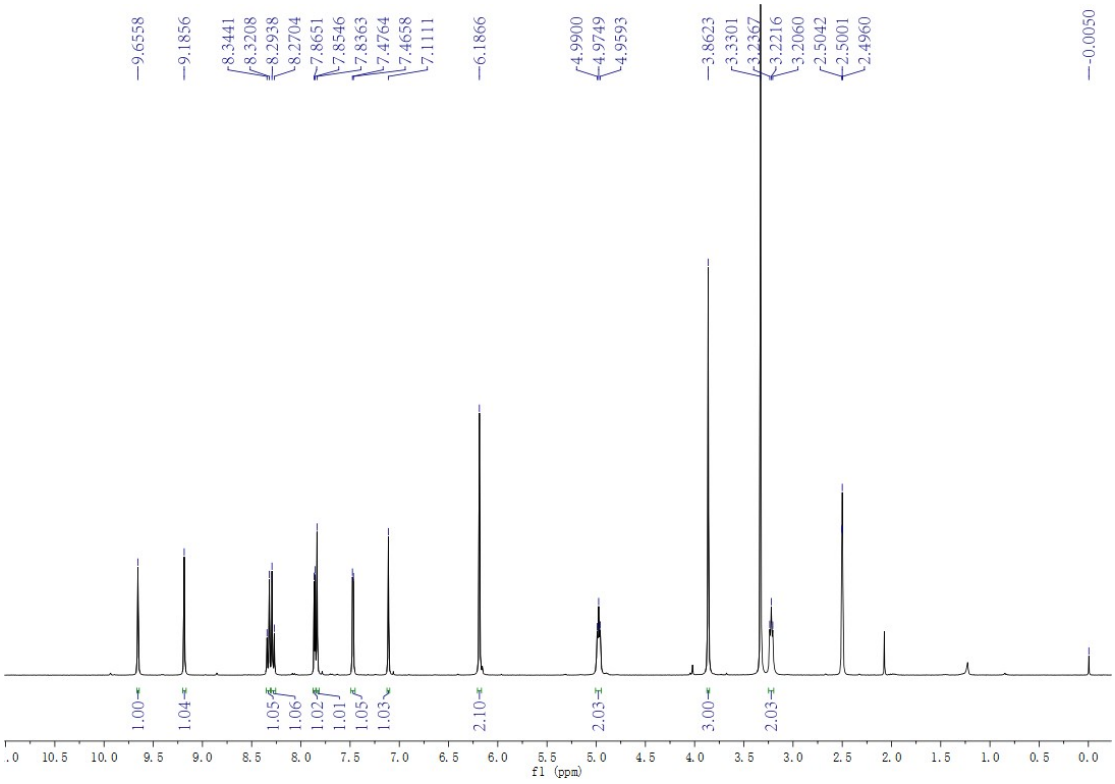
^{1}H NMR Spectra of Compound **B25**



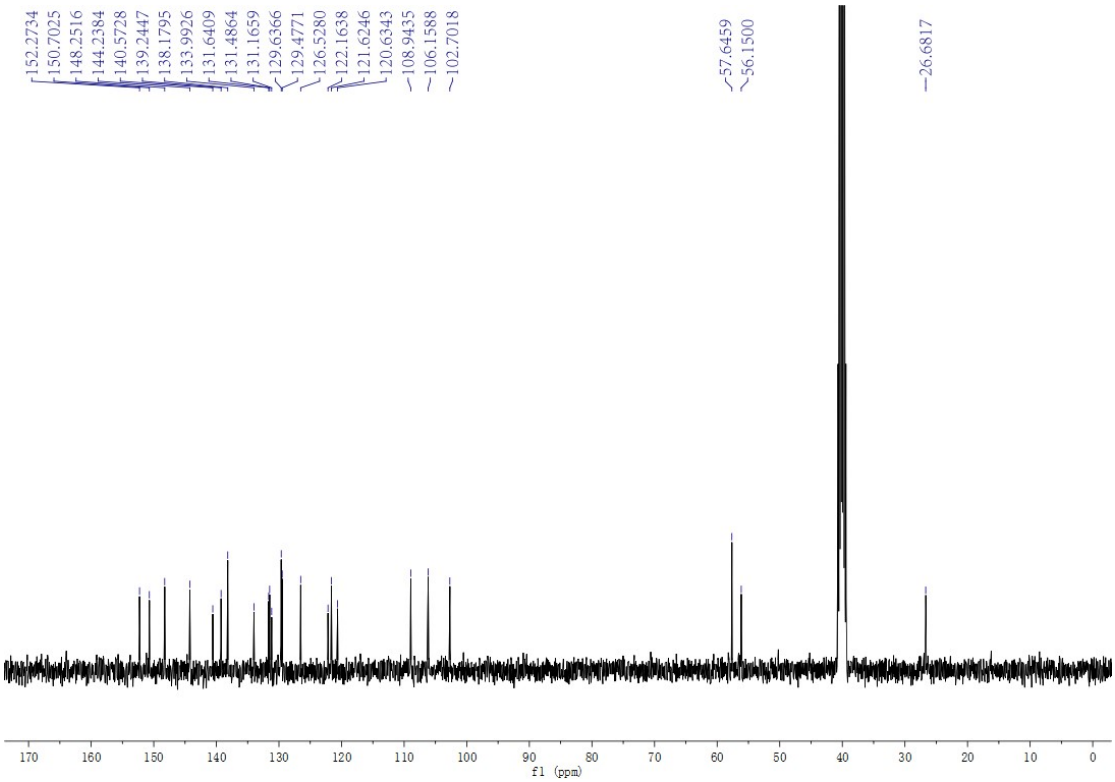
^{13}C NMR Spectra of Compound **B25**



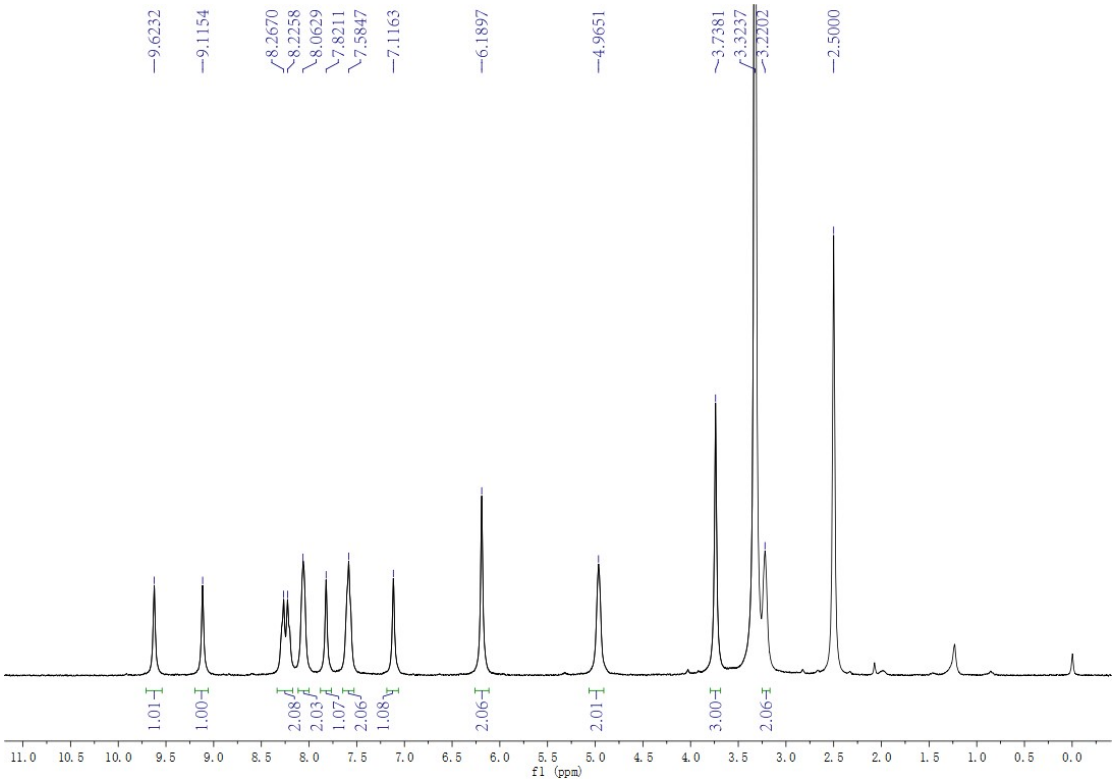
^{1}H NMR Spectra of Compound **B26**



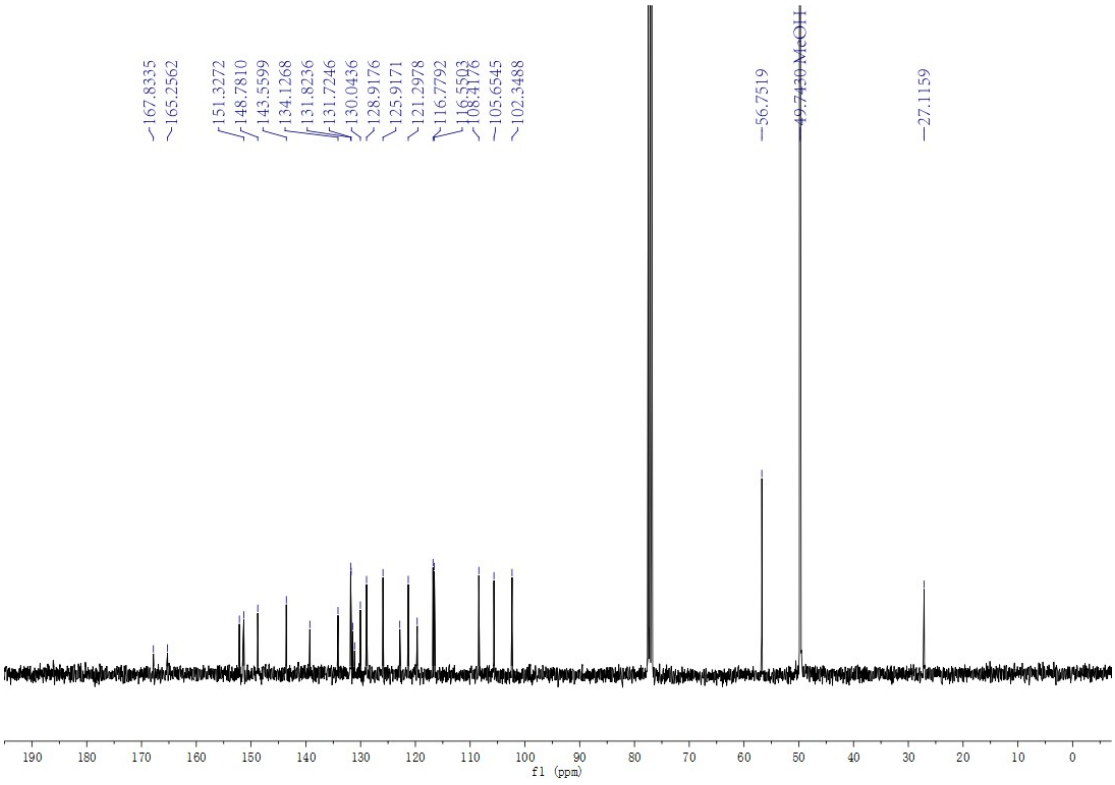
^{13}C NMR Spectra of Compound **B26**



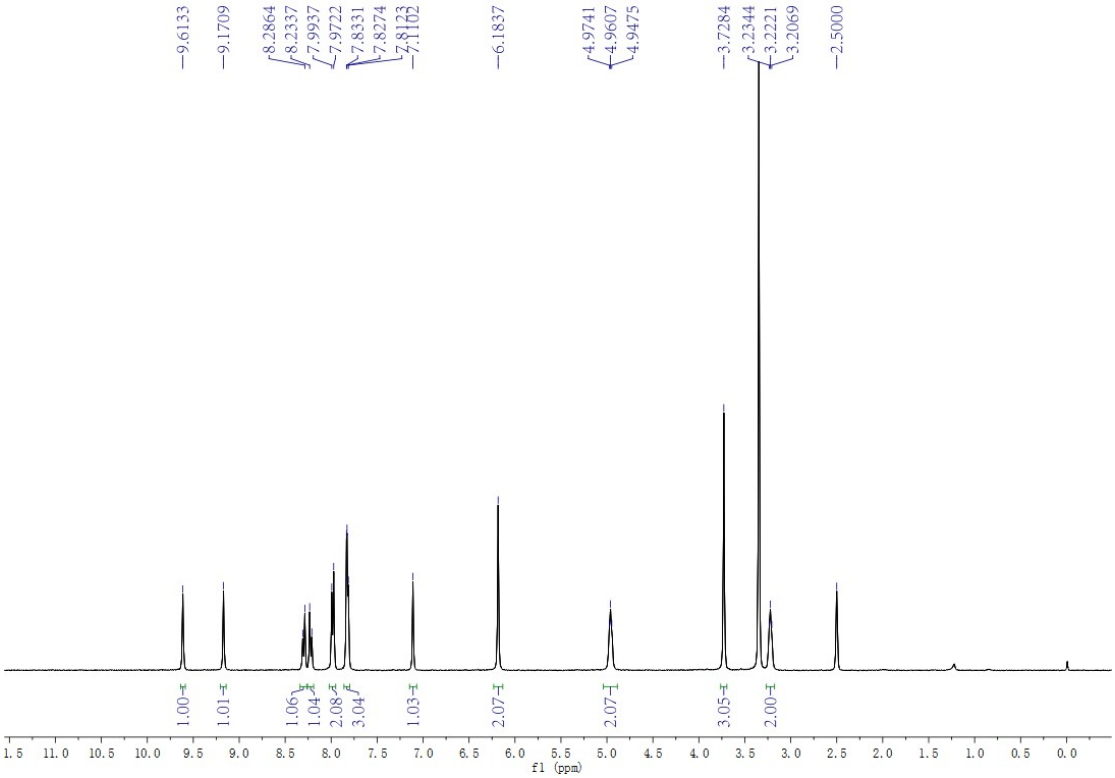
^{1}H NMR Spectra of Compound **B27**



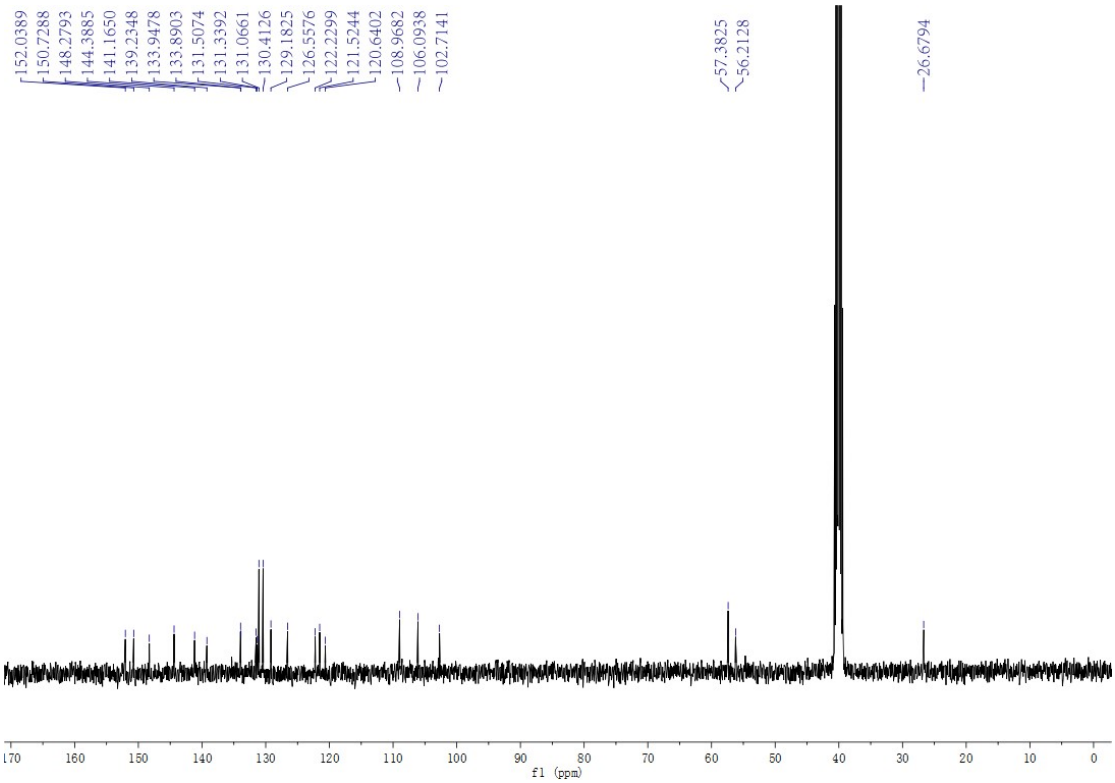
^{13}C NMR Spectra of Compound **B27**



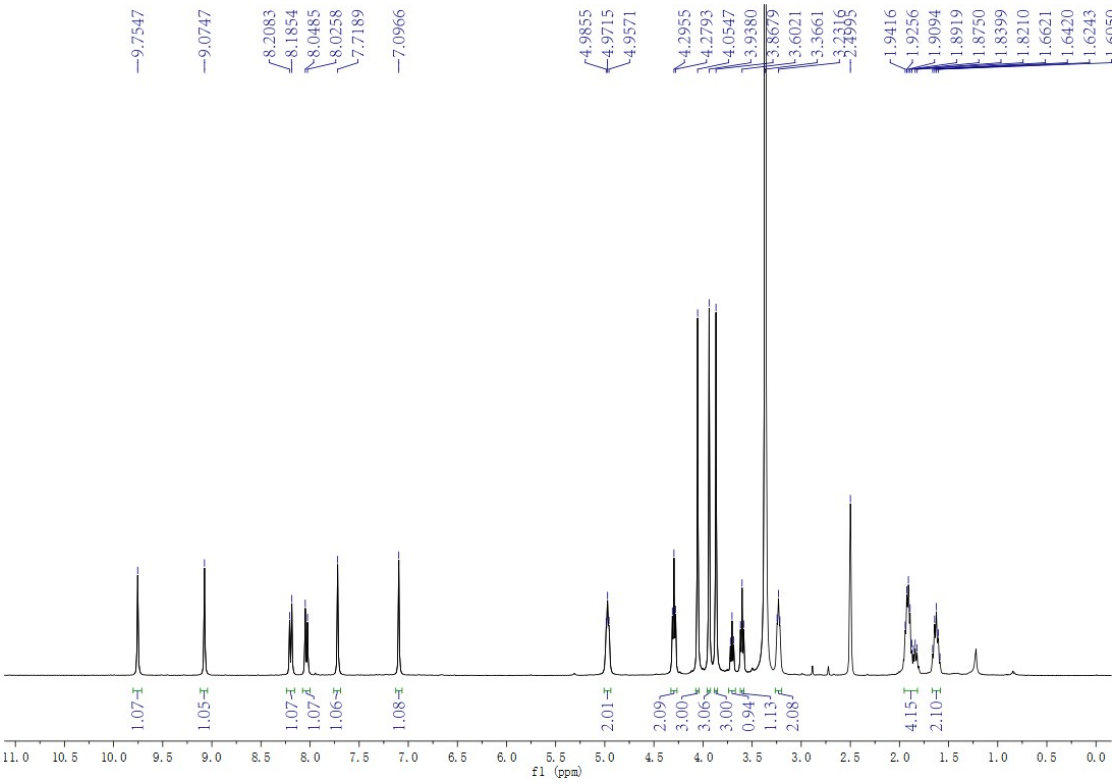
^{1}H NMR Spectra of Compound **B28**



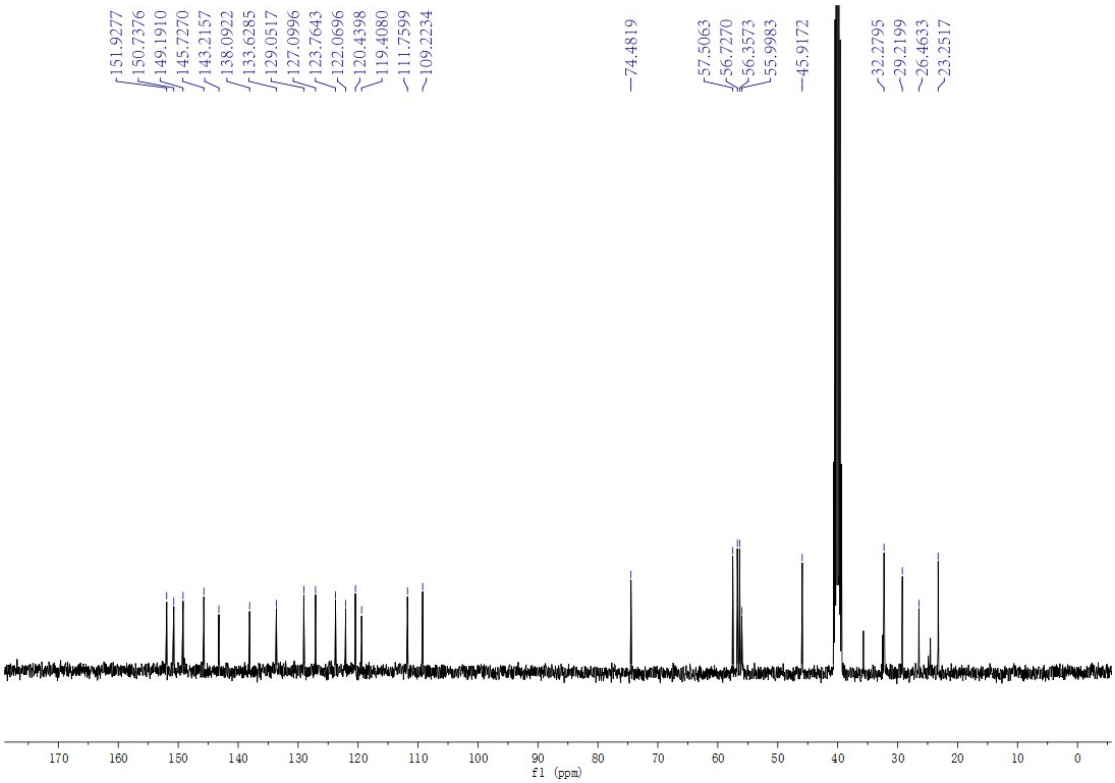
^{13}C NMR Spectra of Compound **B28**



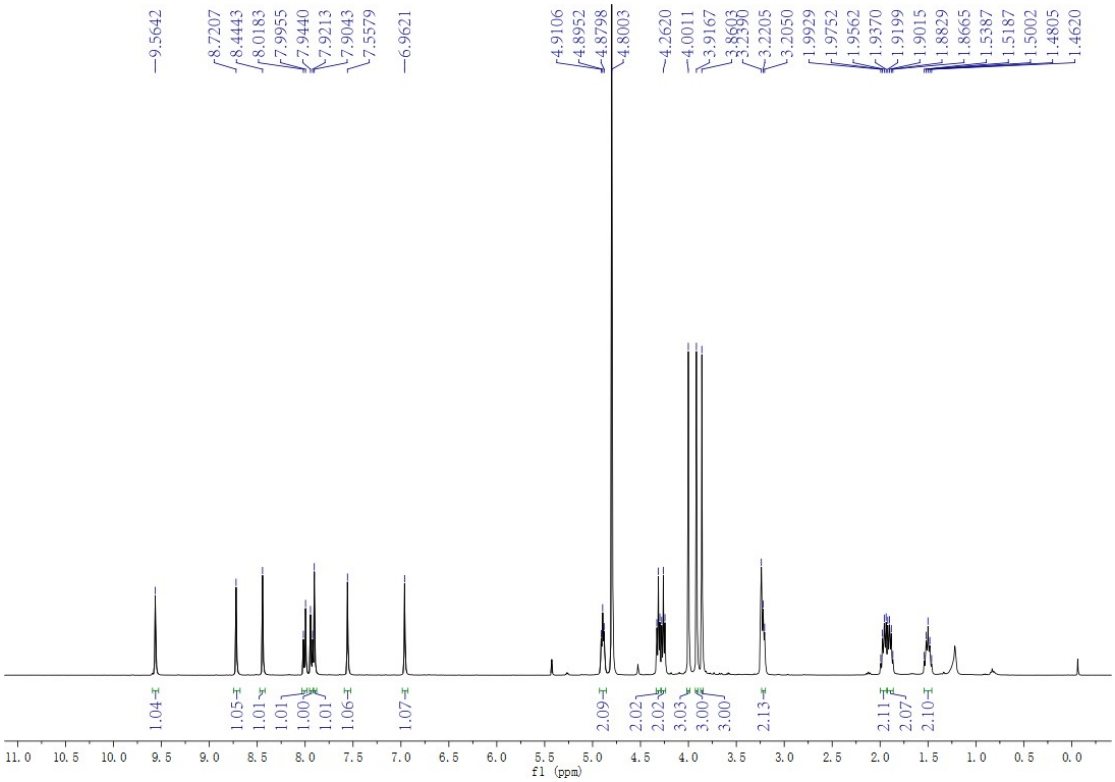
^{1}H NMR Spectra of Compound **C1**



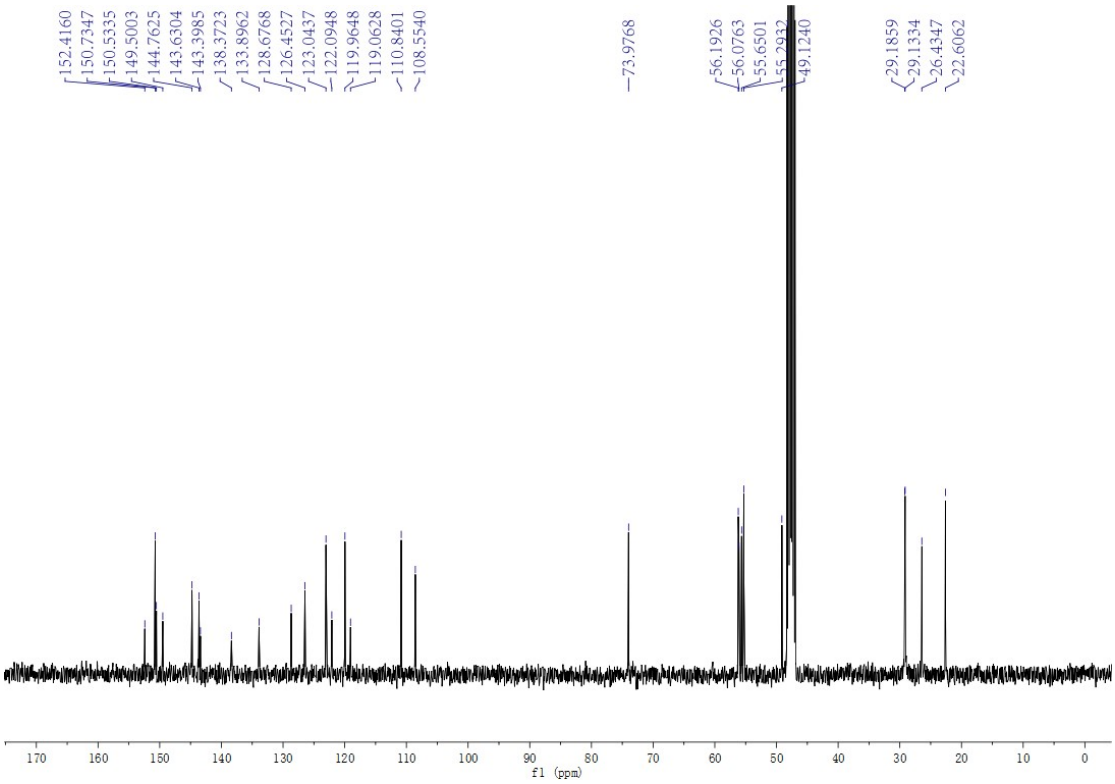
^{13}C NMR Spectra of Compound **C1**



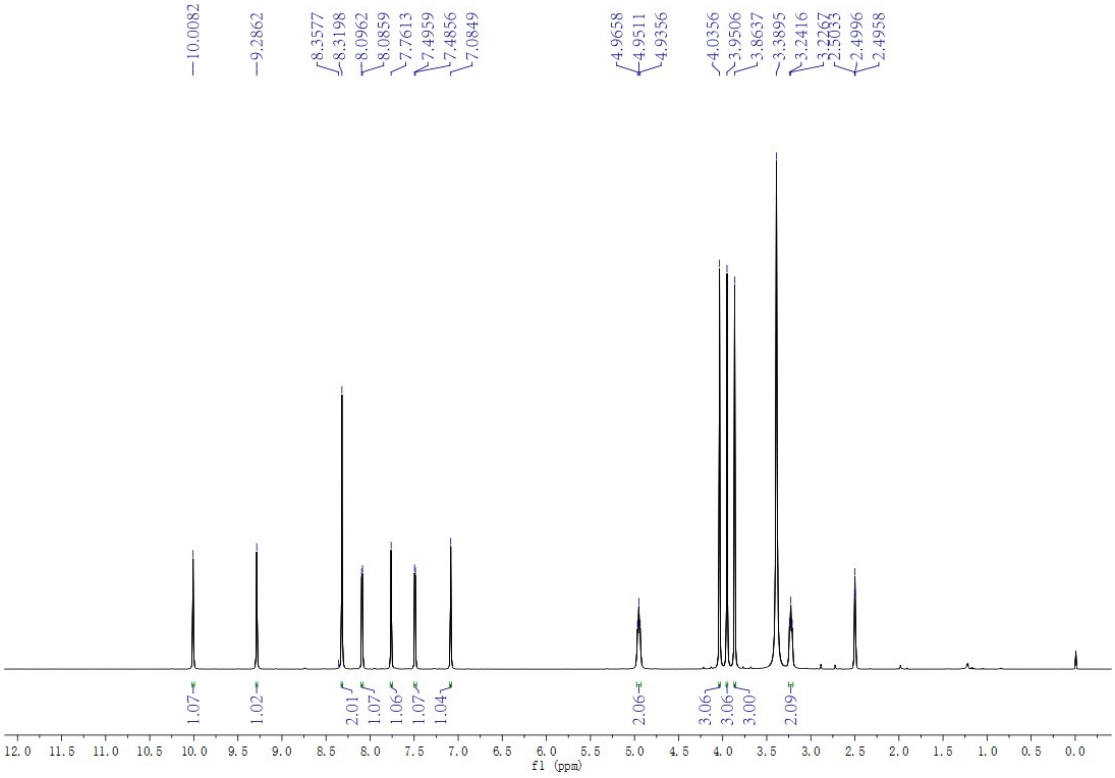
^{1}H NMR Spectra of Compound **C2**



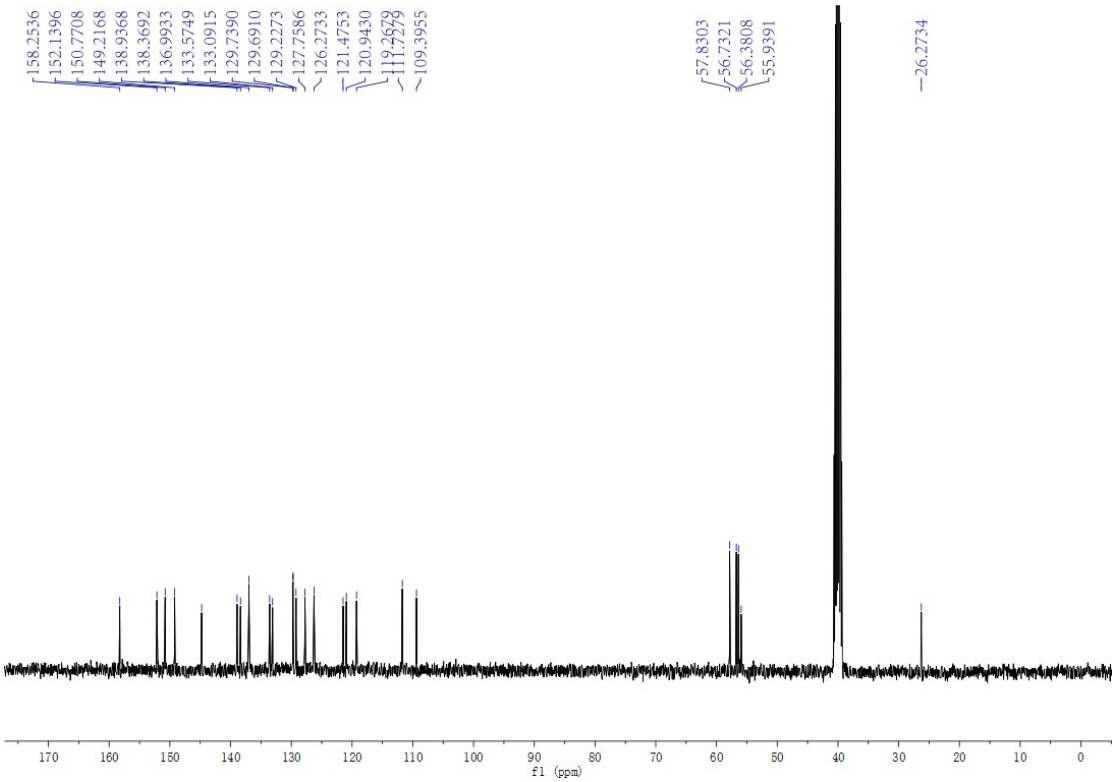
^{13}C NMR Spectra of Compound **C2**



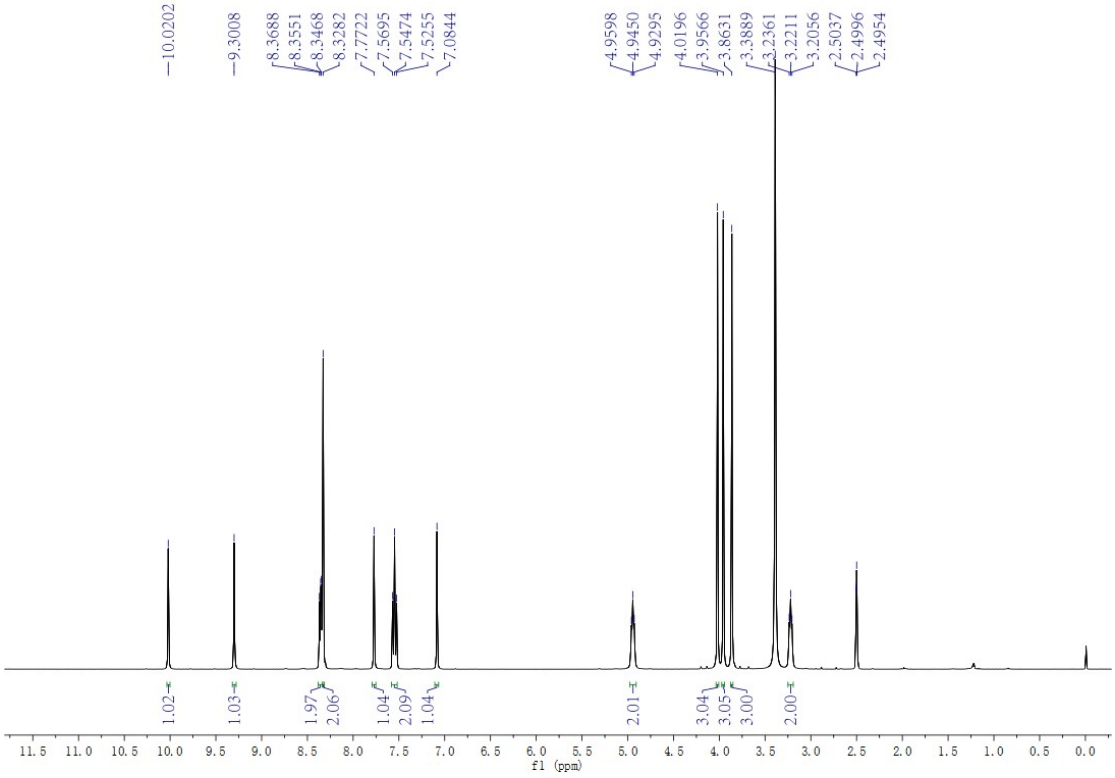
^{1}H NMR Spectra of Compound **C3**



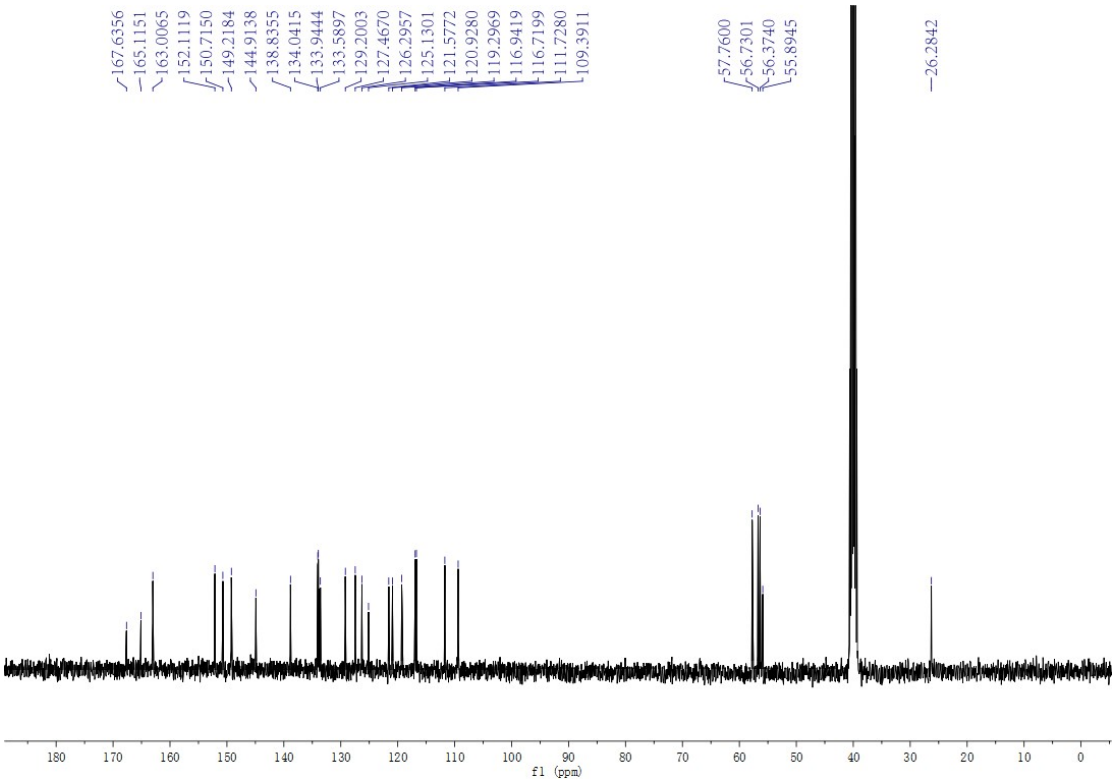
^{13}C NMR Spectra of Compound **C3**



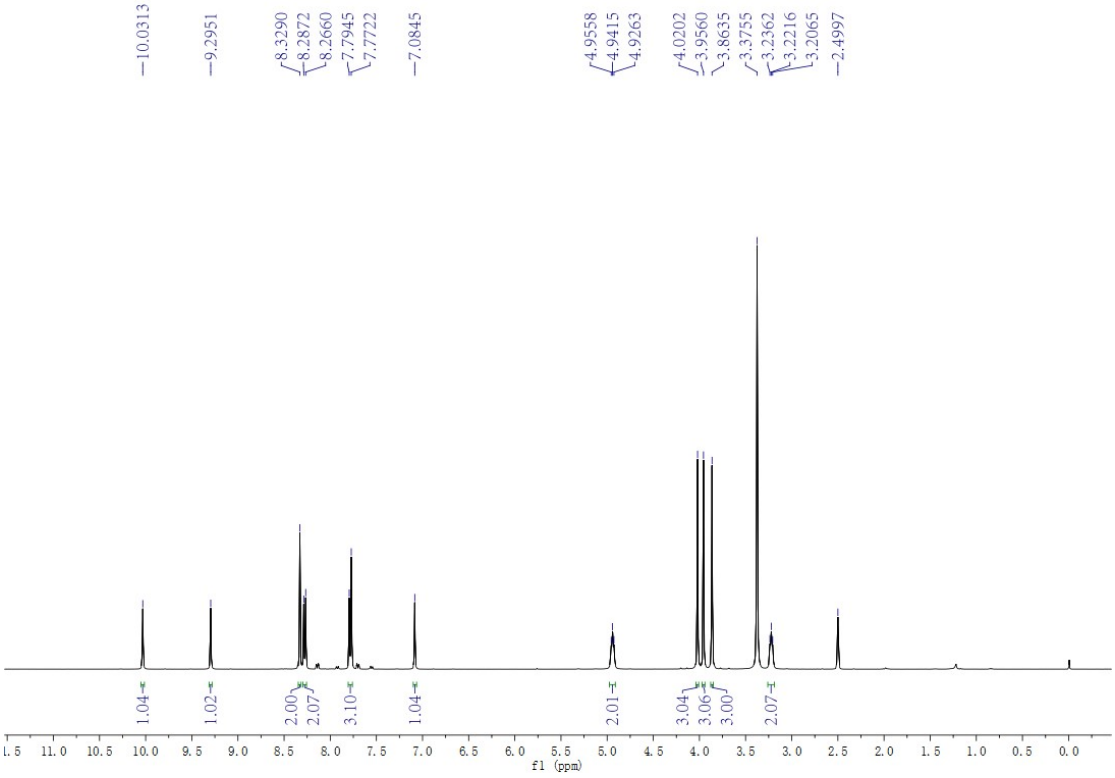
^{1}H NMR Spectra of Compound **C4**



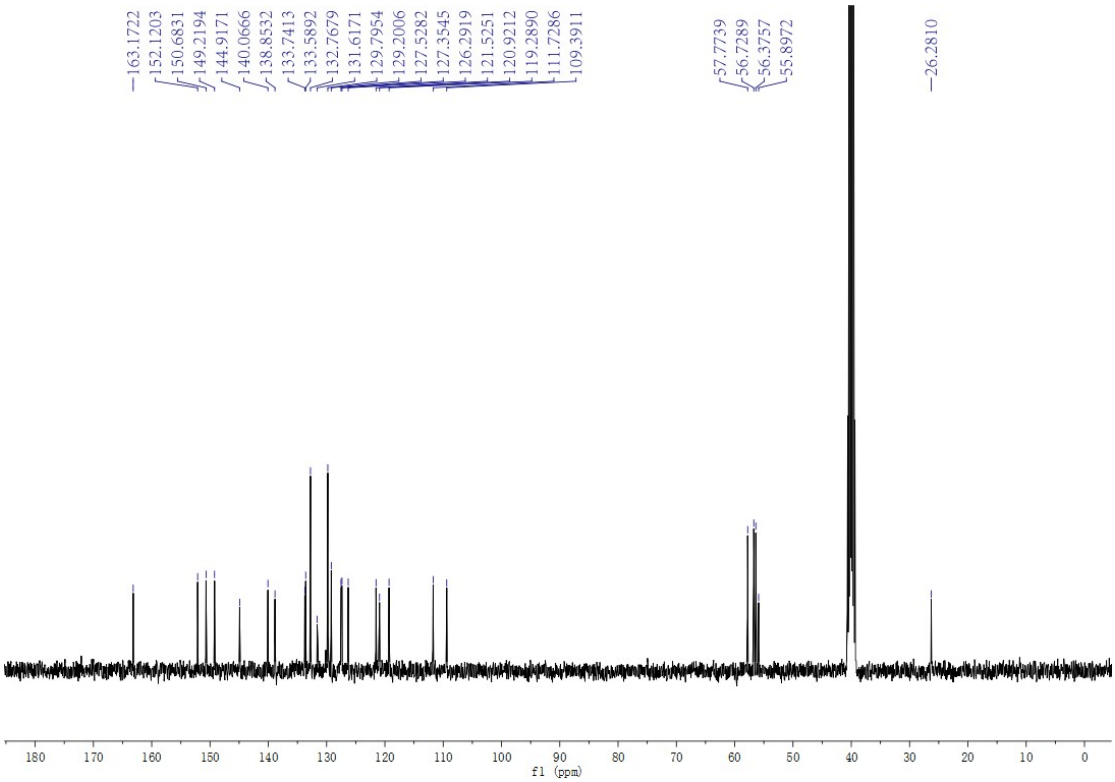
^{13}C NMR Spectra of Compound **C4**



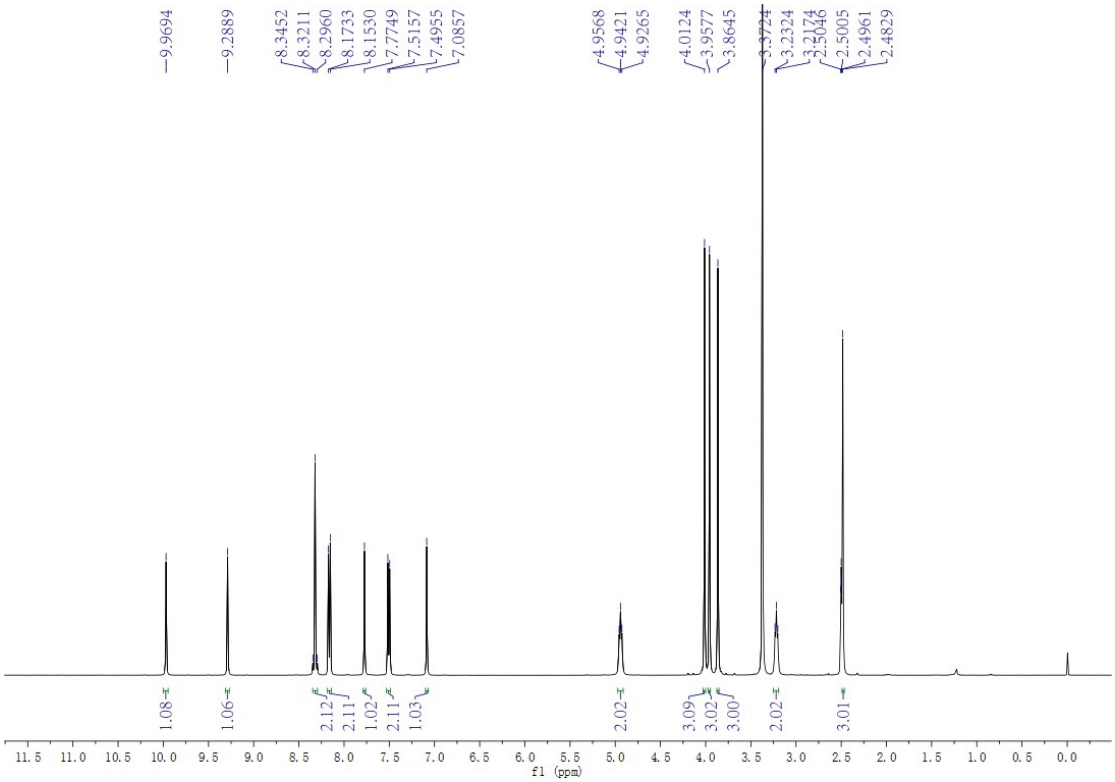
[1]H NMR Spectra of Compound **C5**



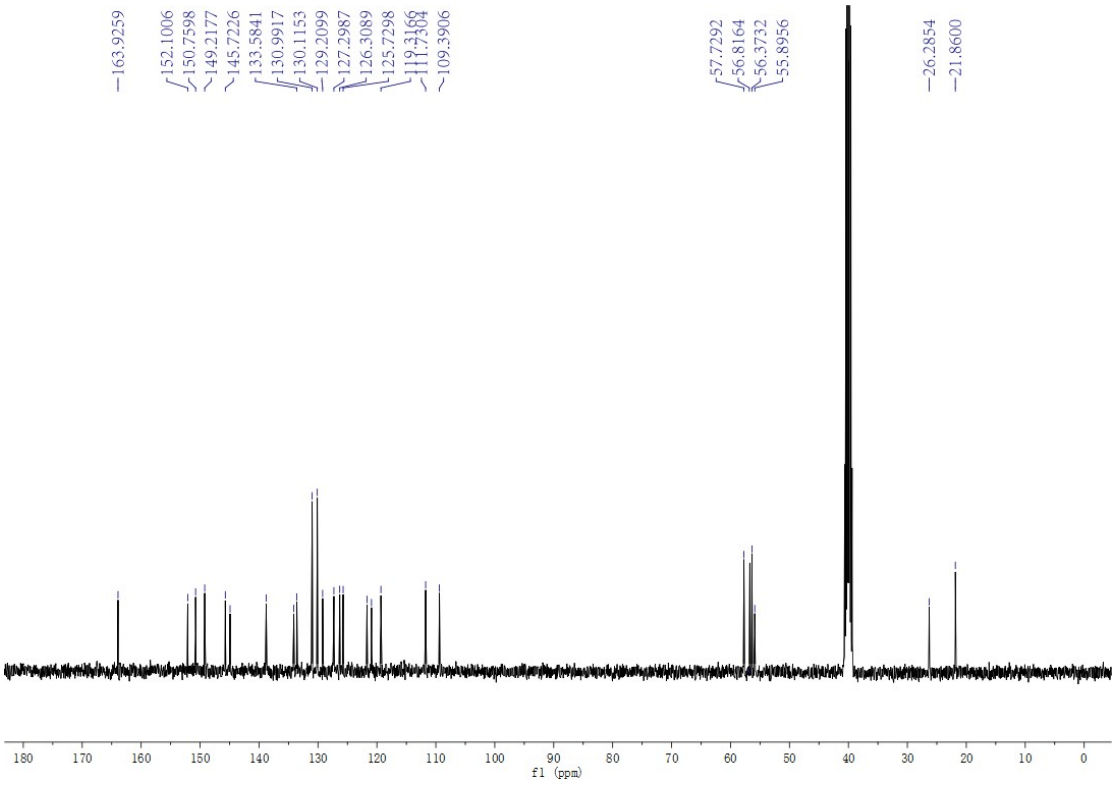
[13]C NMR Spectra of Compound **C5**



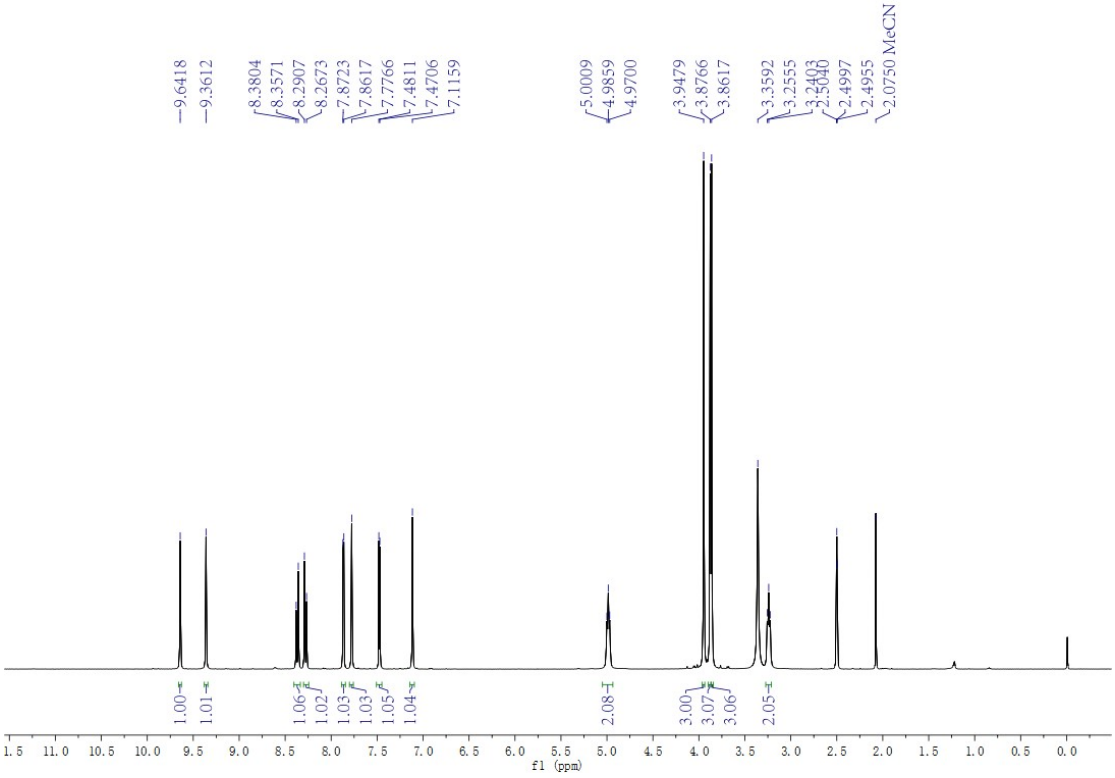
^{1}H NMR Spectra of Compound **C6**



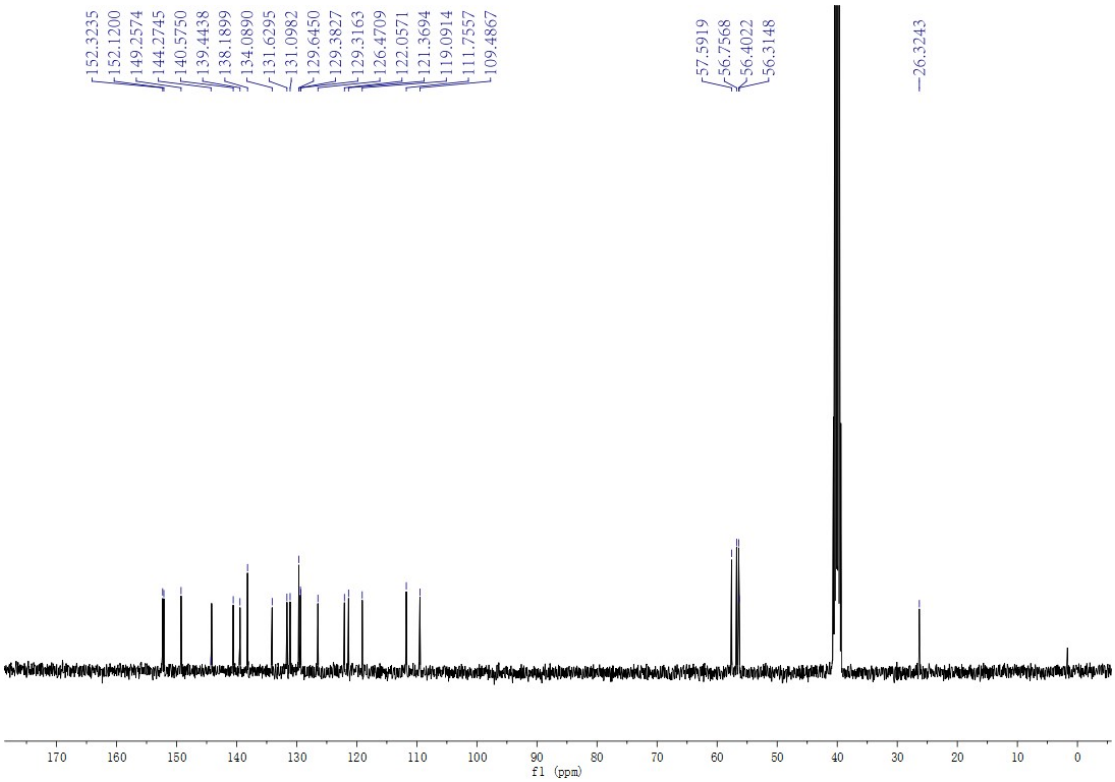
^{13}C NMR Spectra of Compound **C6**



^{1}H NMR Spectra of Compound **C7**



^{13}C NMR Spectra of Compound **C7**



3. HPLC spectra

We provided the spectra of HPLC assays as below.

HPLC: Agilent 1260

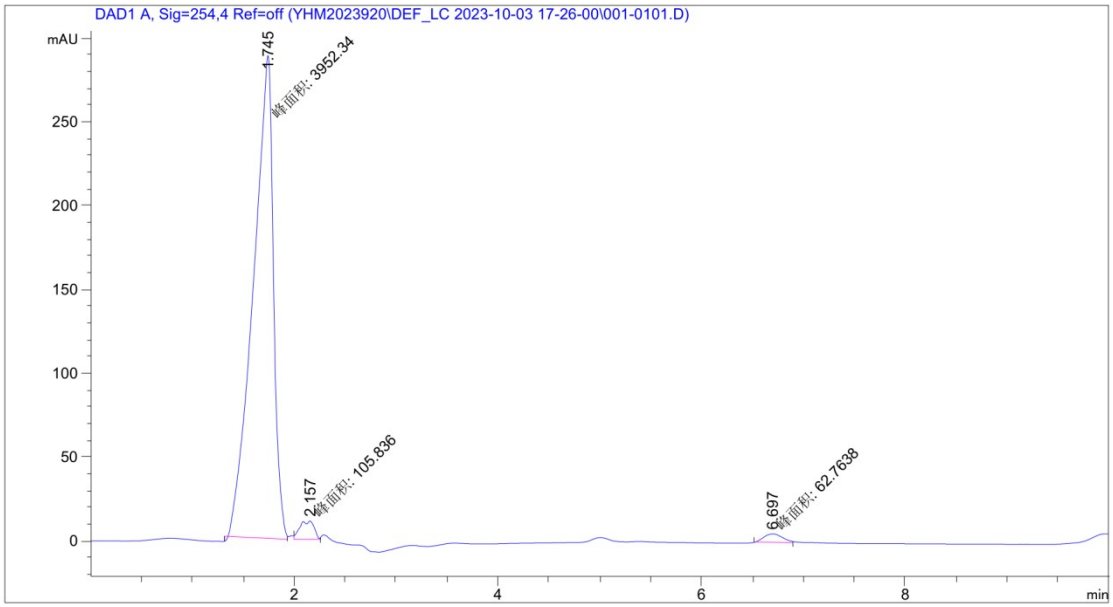
Column: Inertex C_{18} (150 mm × 4.6 mm × 5 μm);

Mobile phase: Acetonitrile-Phosphate buffer (pH=3~3.5) 80:20;

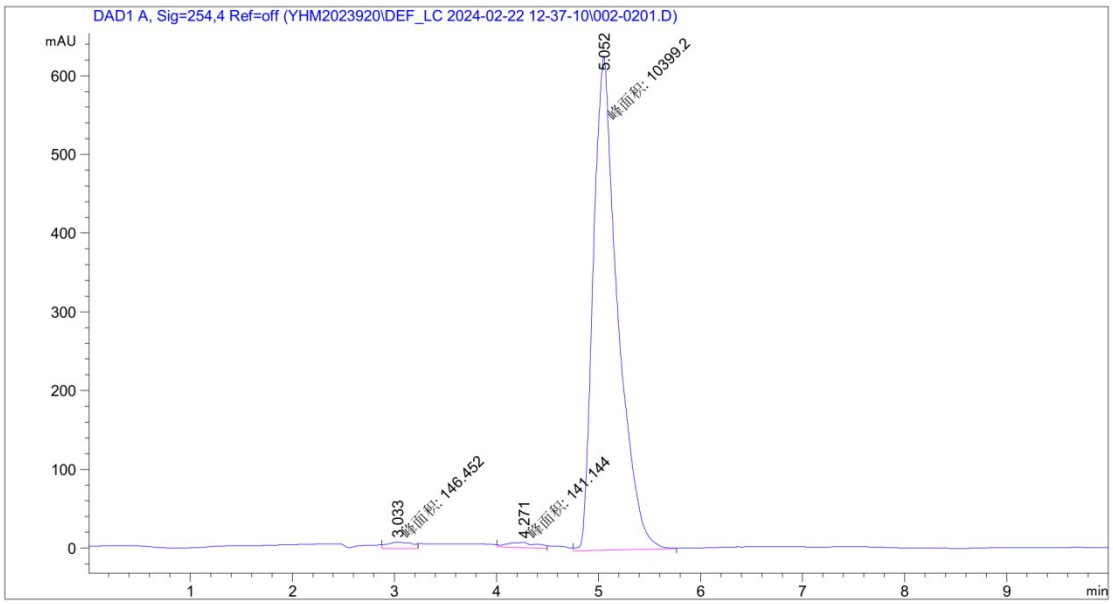
Wavelength: 254 nm;

Rate: 1.0 mL/min.

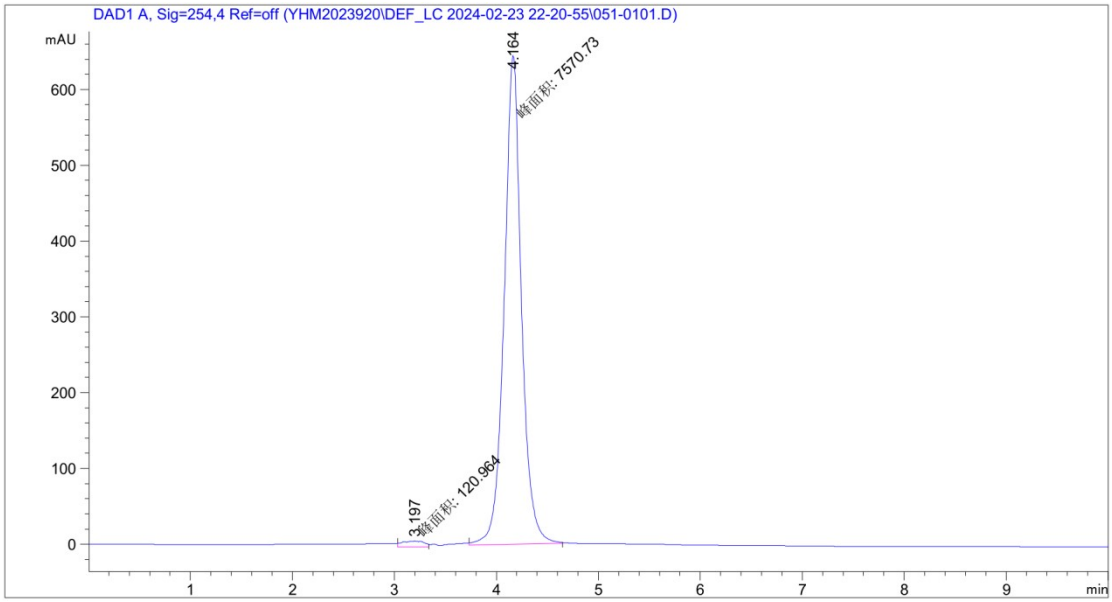
B1, 95.91%, t_R =1.745 min



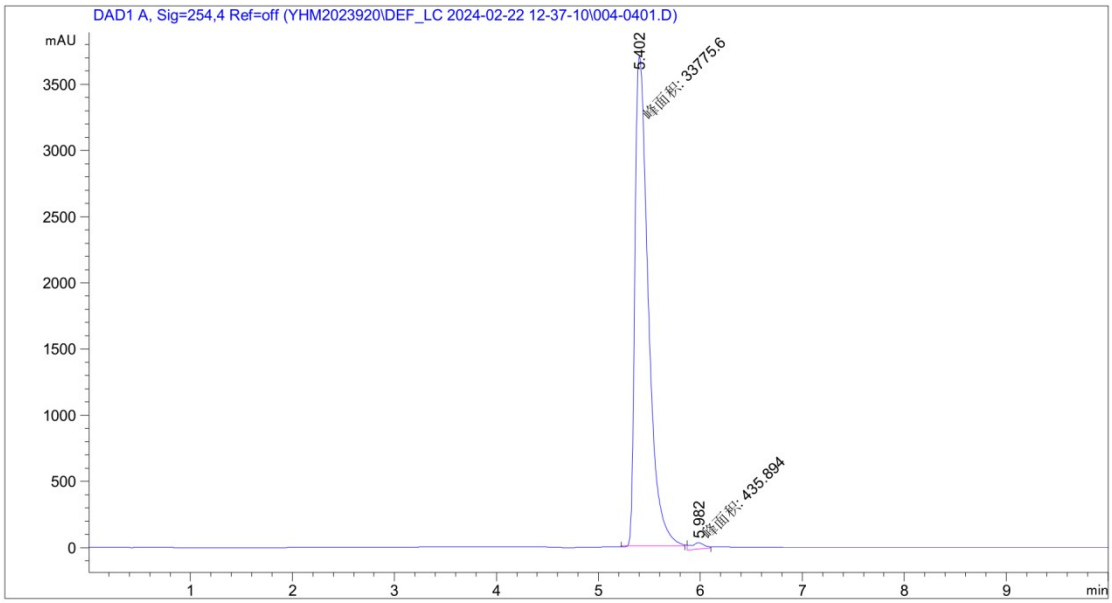
B2, 97.31%, t_R =5.052 min



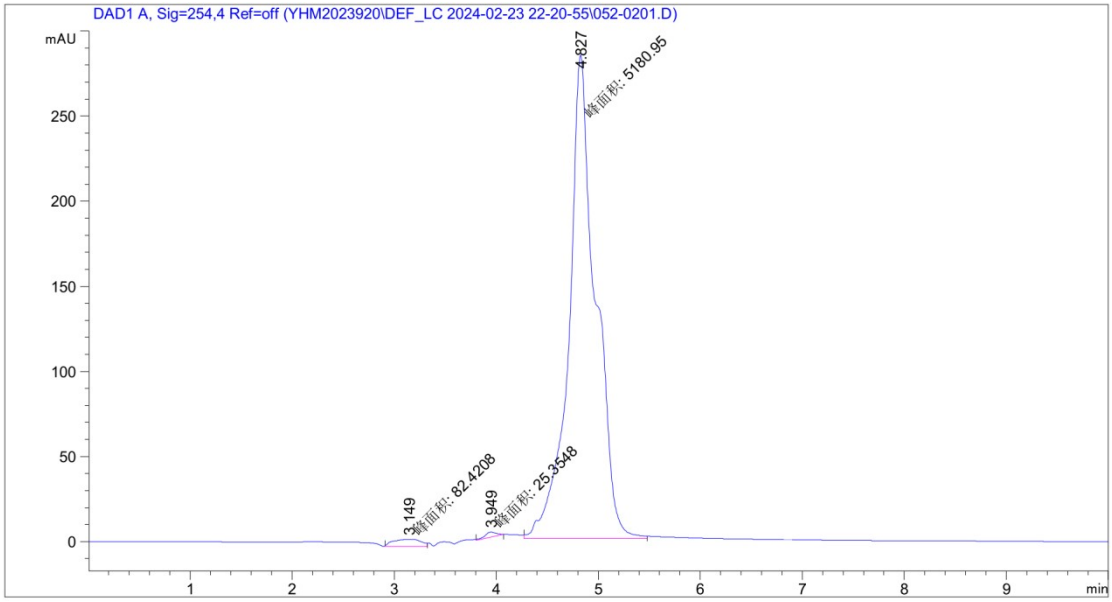
B3, 98.43%, t_R =4.164 min



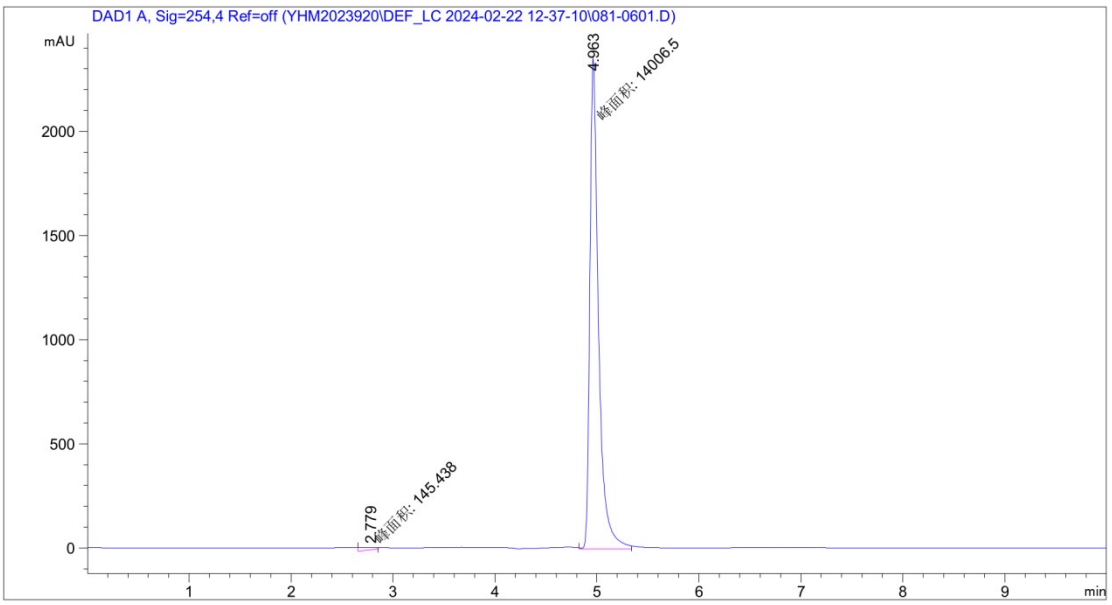
B4, 98.73%, t_R =5.402 min



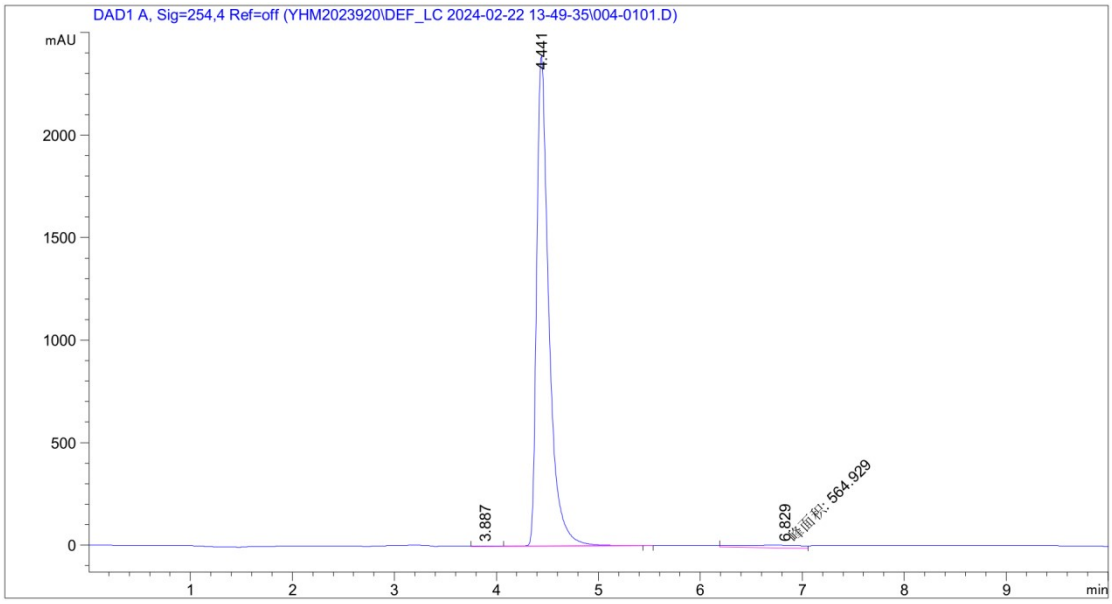
B5, 97.96%, t_R =4.827 min



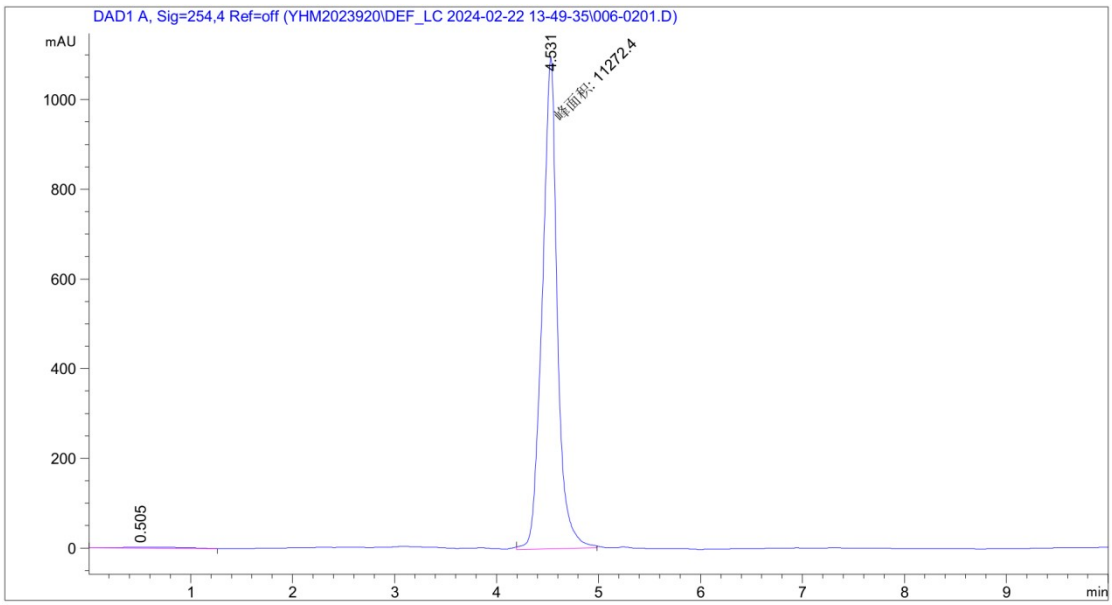
B6, 98.97%, t_R =4.963 min



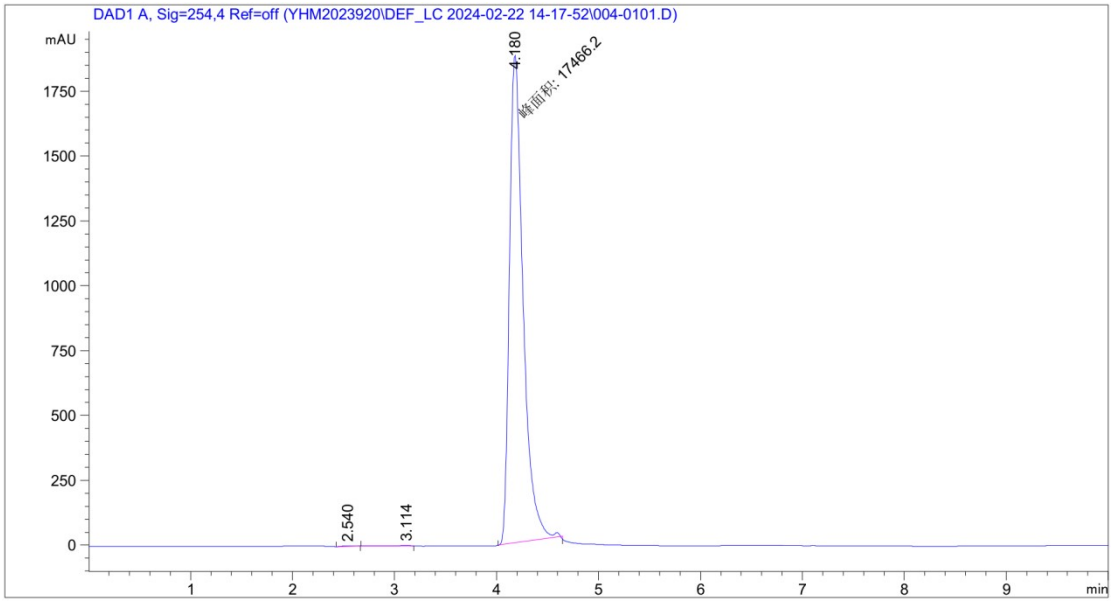
B7, 96.93%, t_R =4.441 min



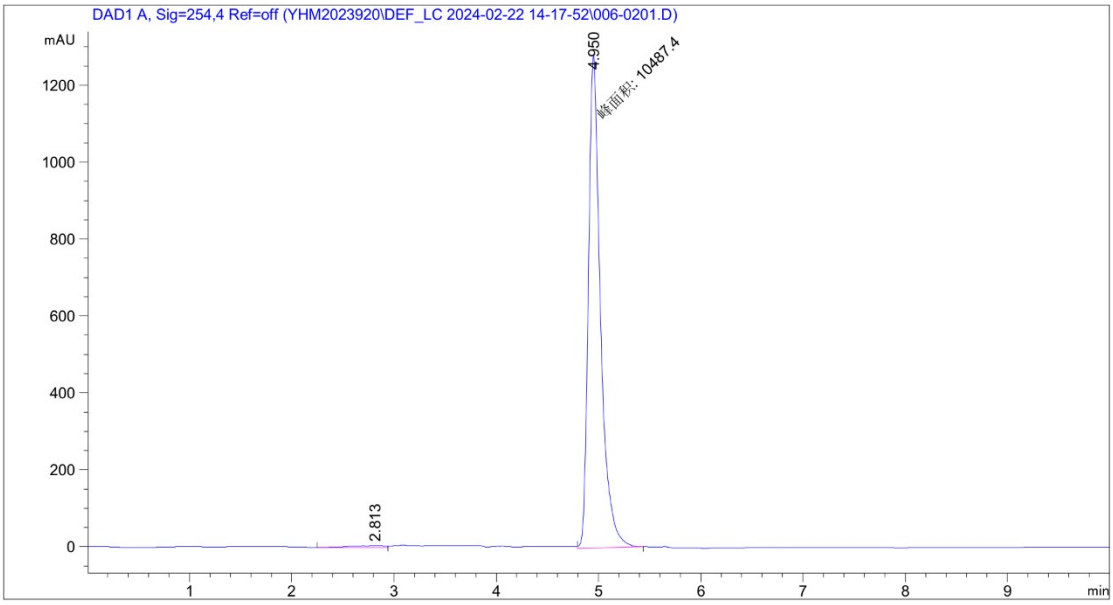
B8, 99.16%, t_R =4.531 min



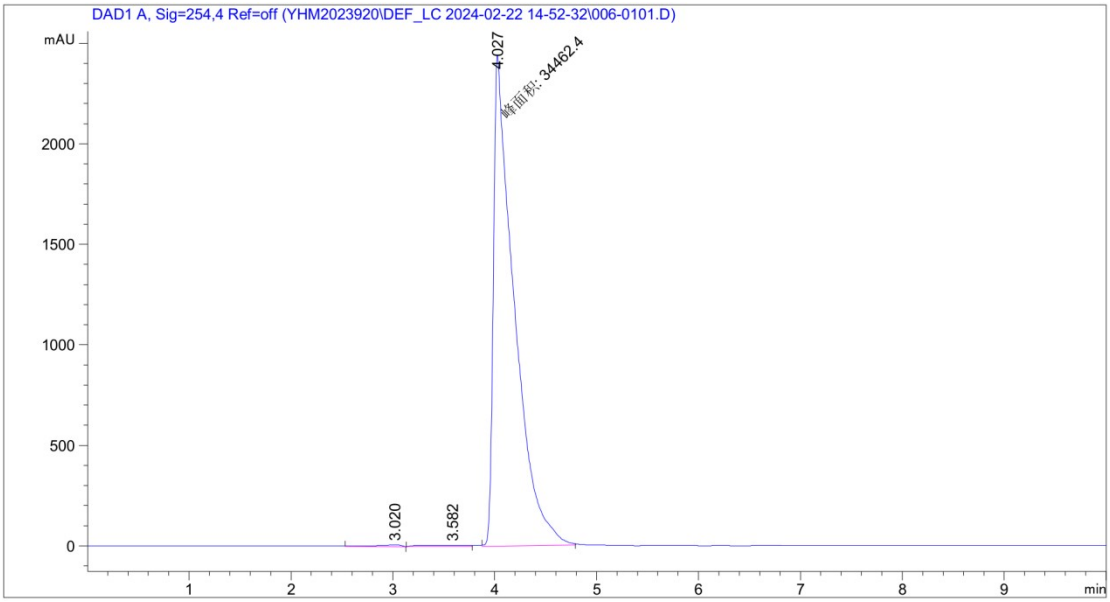
B9, 99.77%, t_R =4.180 min



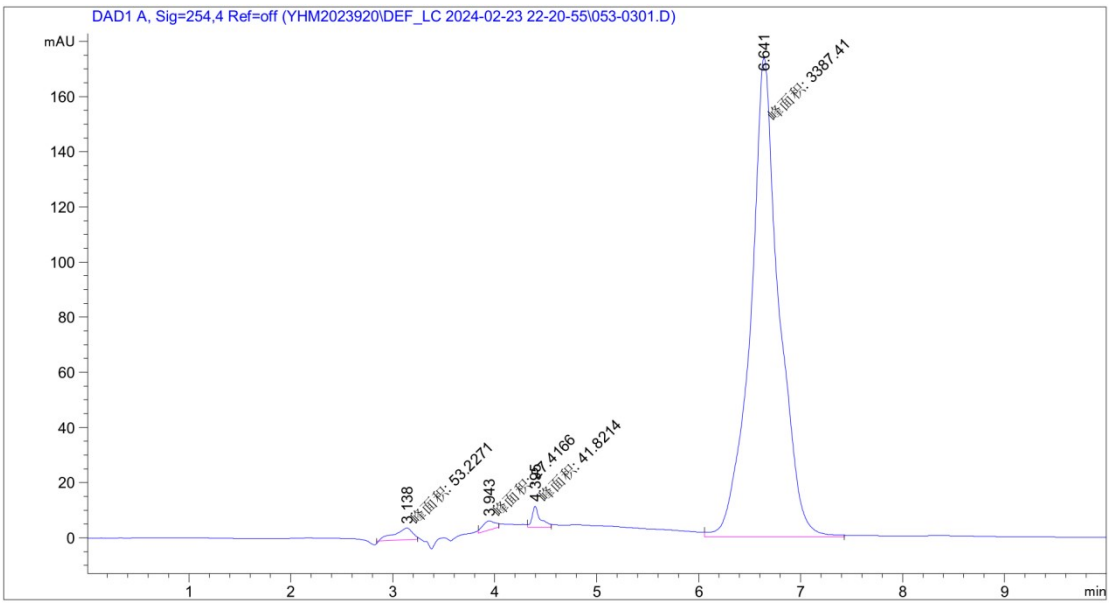
B10, 98.89%, t_R =4.950 min



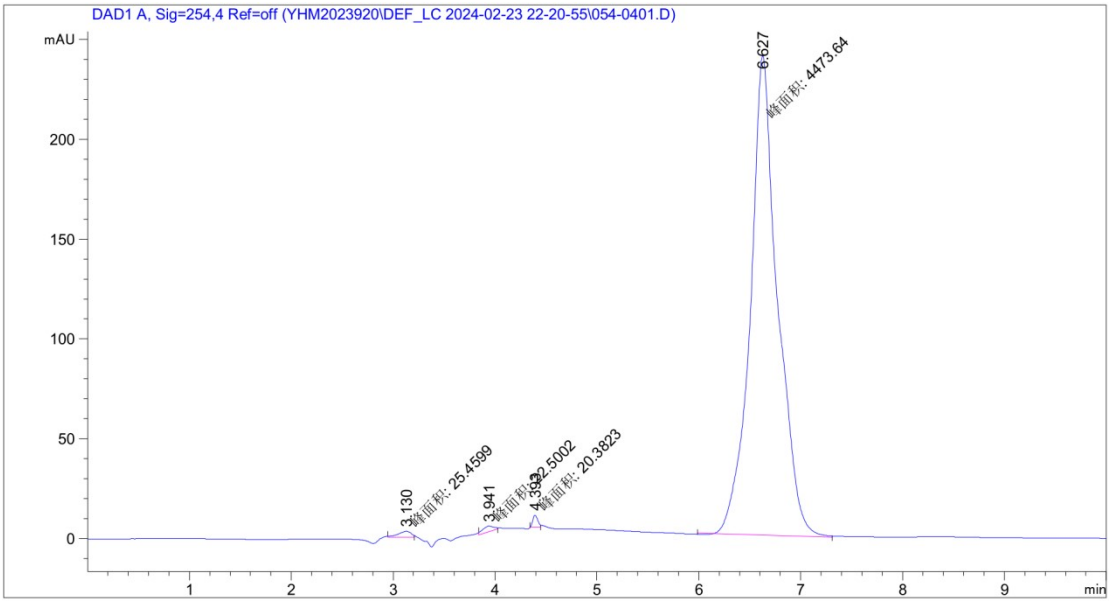
B11, 98.94%, t_R =4.027 min



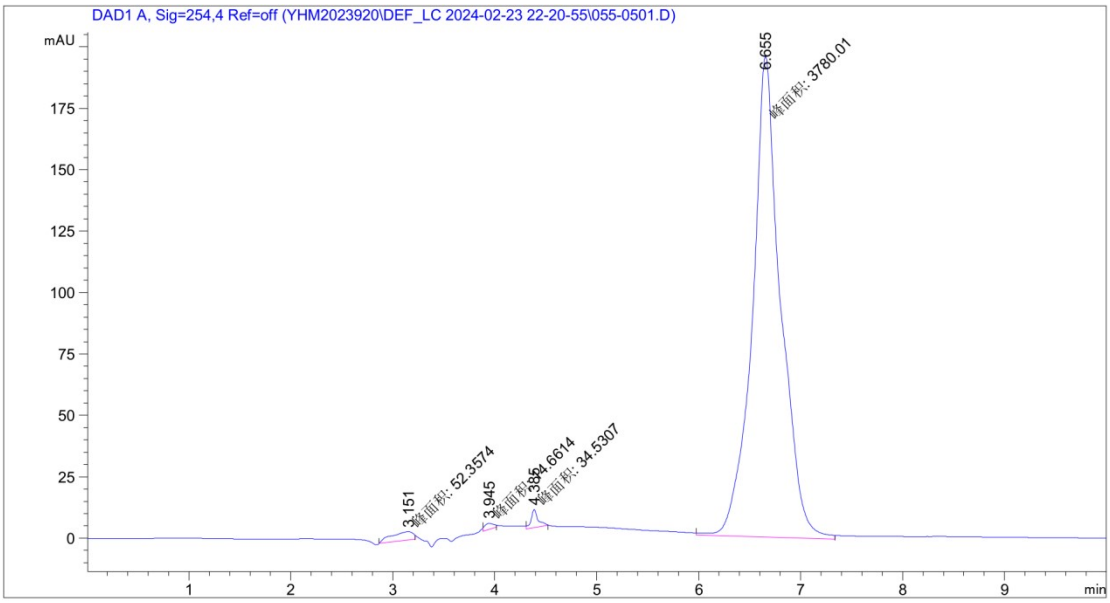
B12, 96.51%, t_R =6.641 min



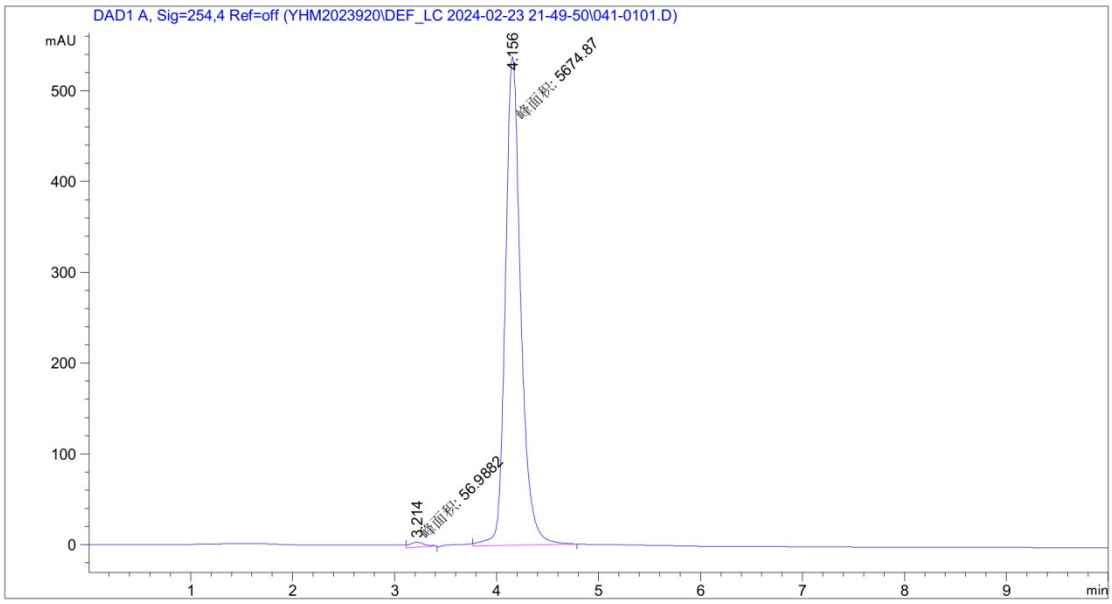
B13, 98.50%, t_R =6.627 min



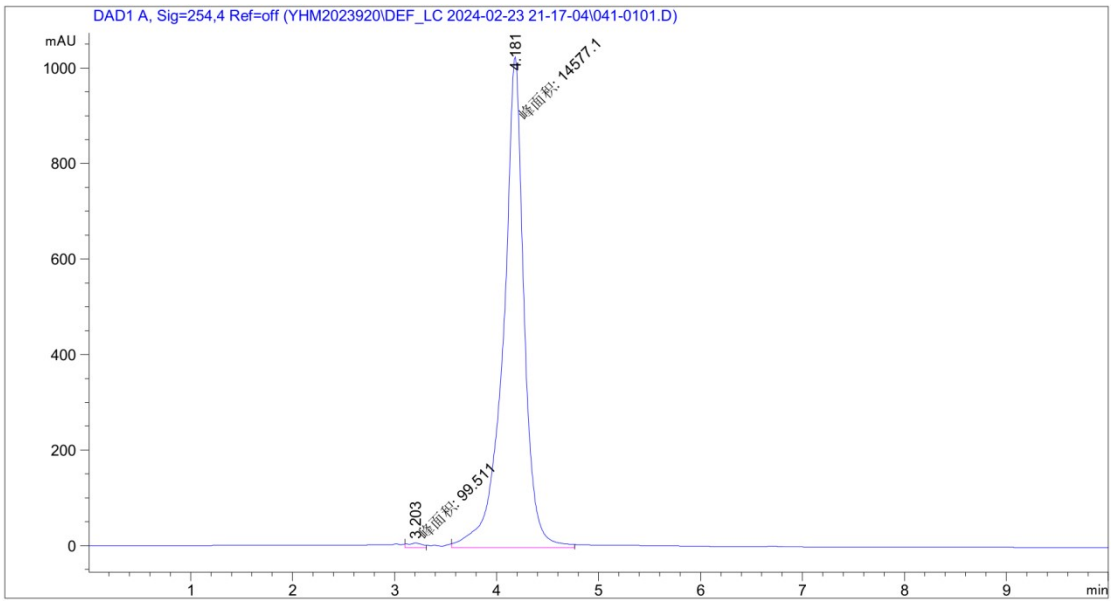
B14, 97.38%, t_R =6.655 min



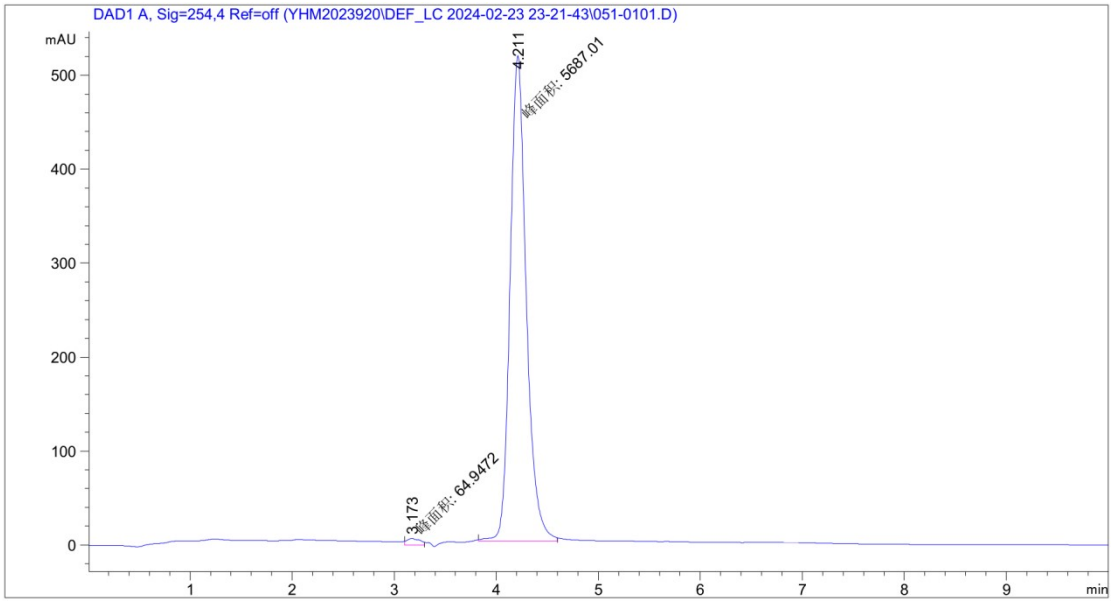
B15, 99.01%, t_R =4.156 min



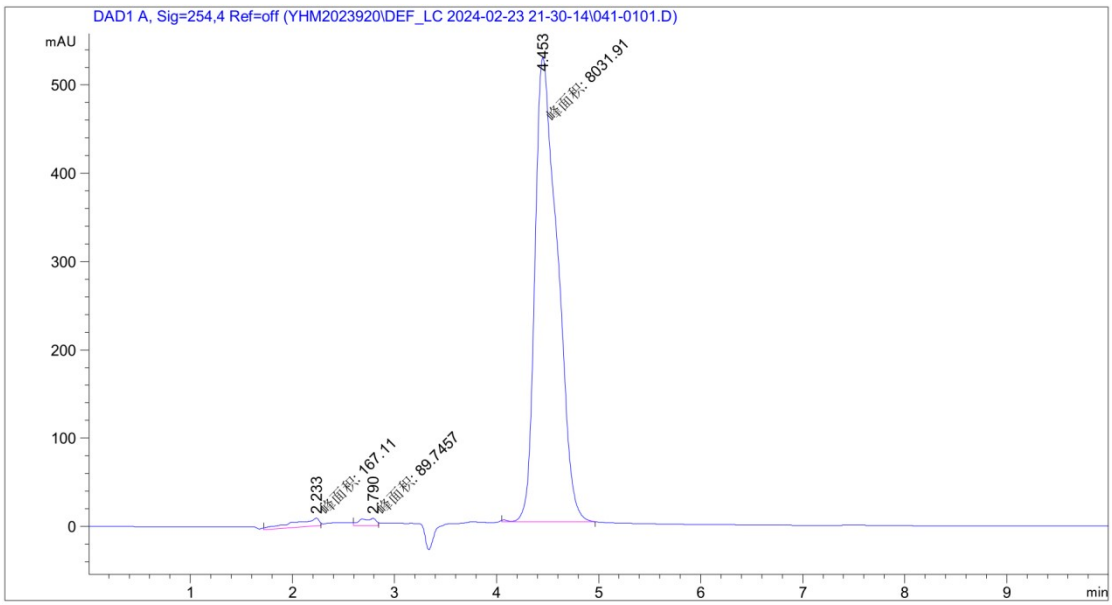
B16, 99.32%, t_R =4.181 min



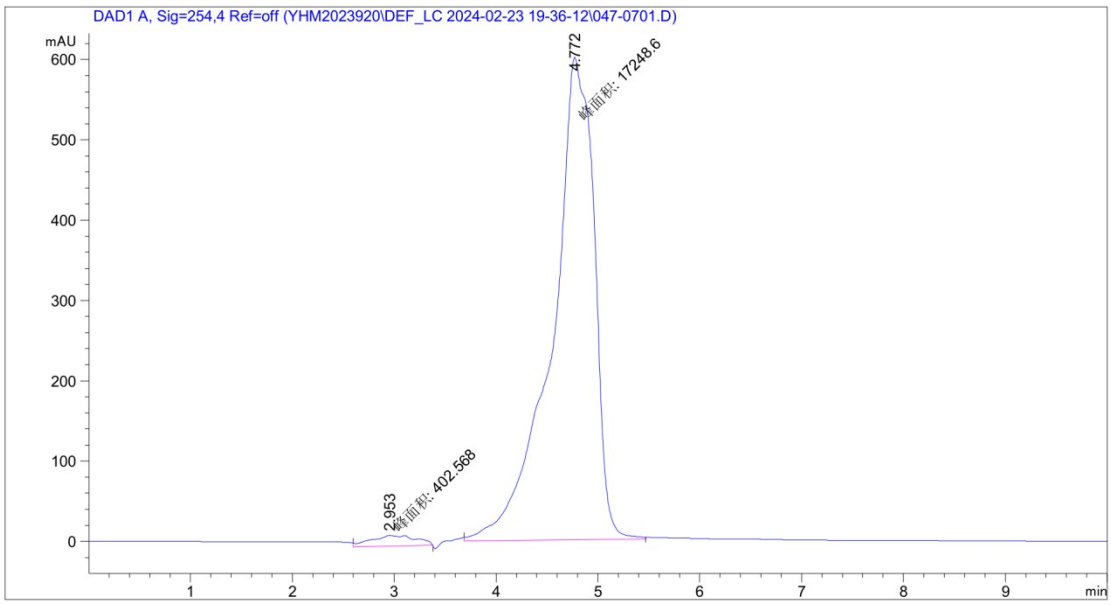
B17, 98.87%, t_R =4.211 min



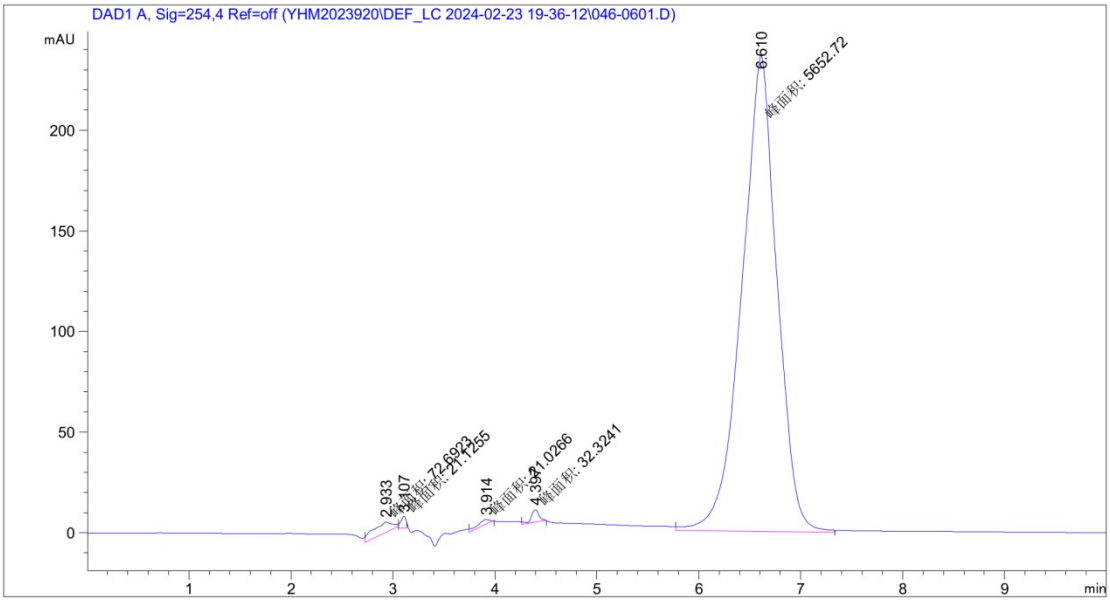
B18, 96.90%, t_R =4.453 min



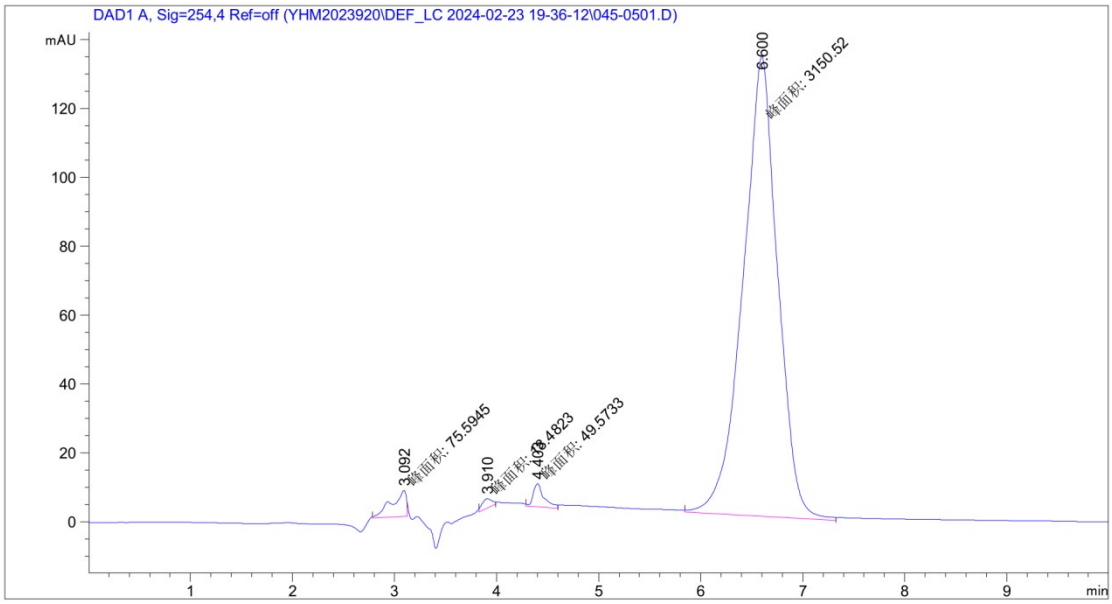
B19, 97.72%, t_R =4.772 min



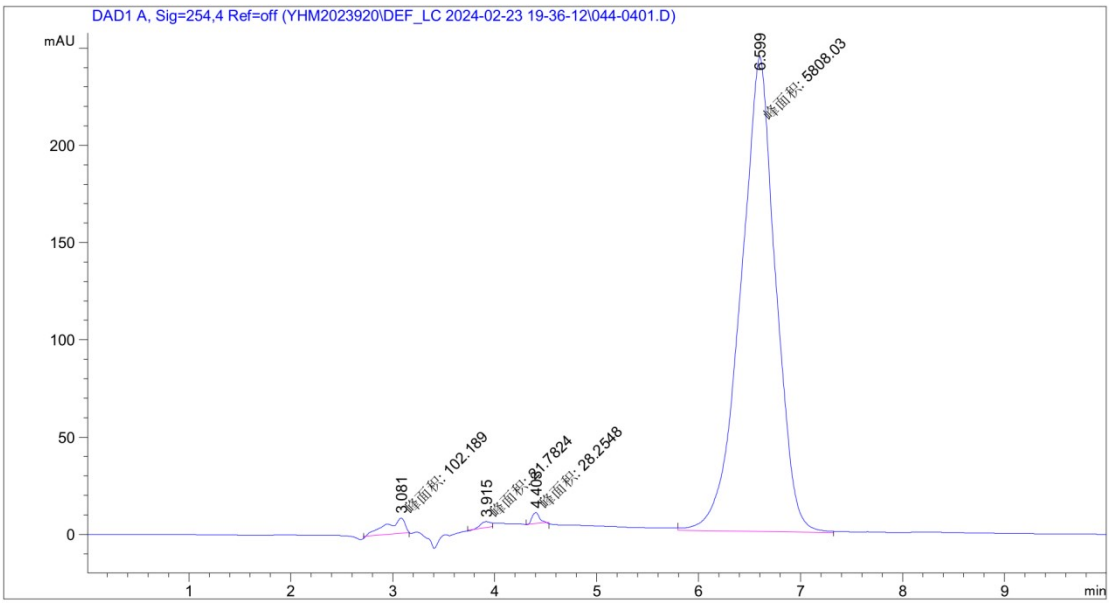
B20, 97.46%, t_R =6.610 min



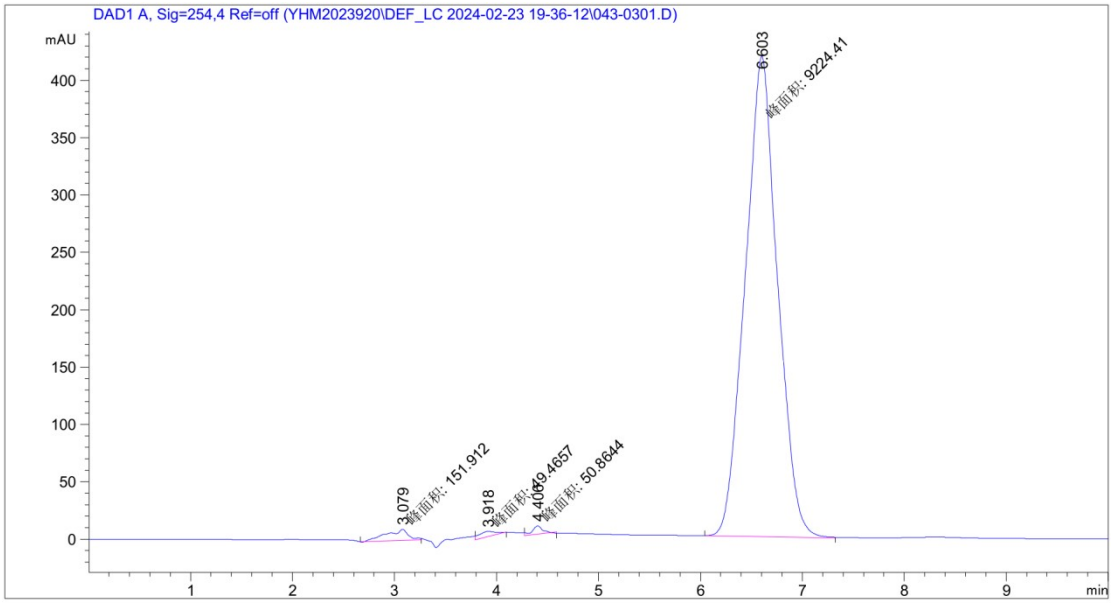
B21, 95.64%, t_R =6.600 min



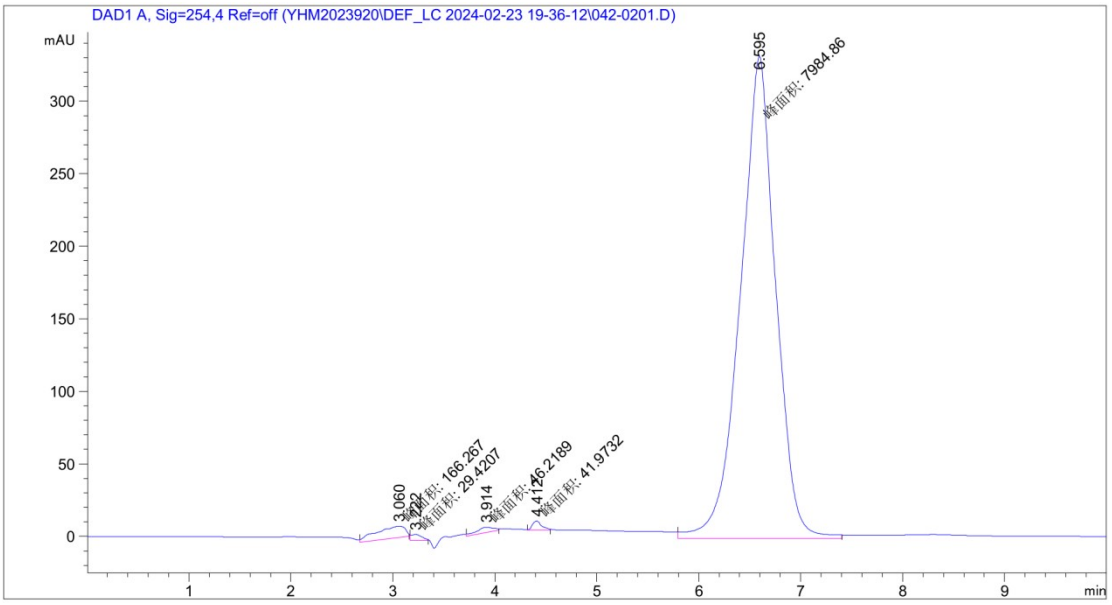
B22, 97.45%, t_R =6.599 min



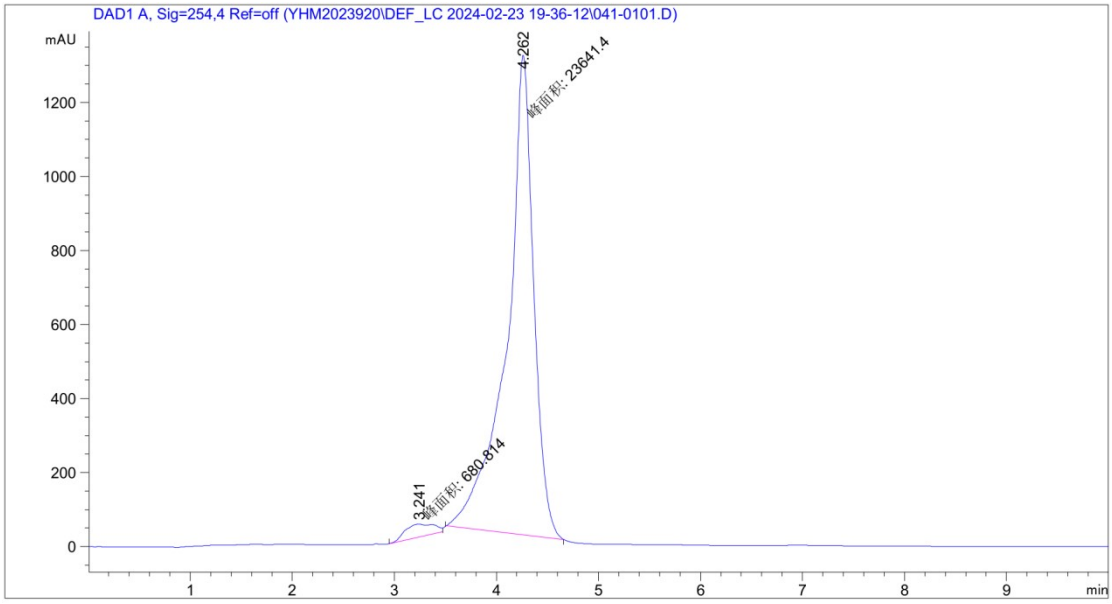
B23, 97.34%, t_R =6.603 min



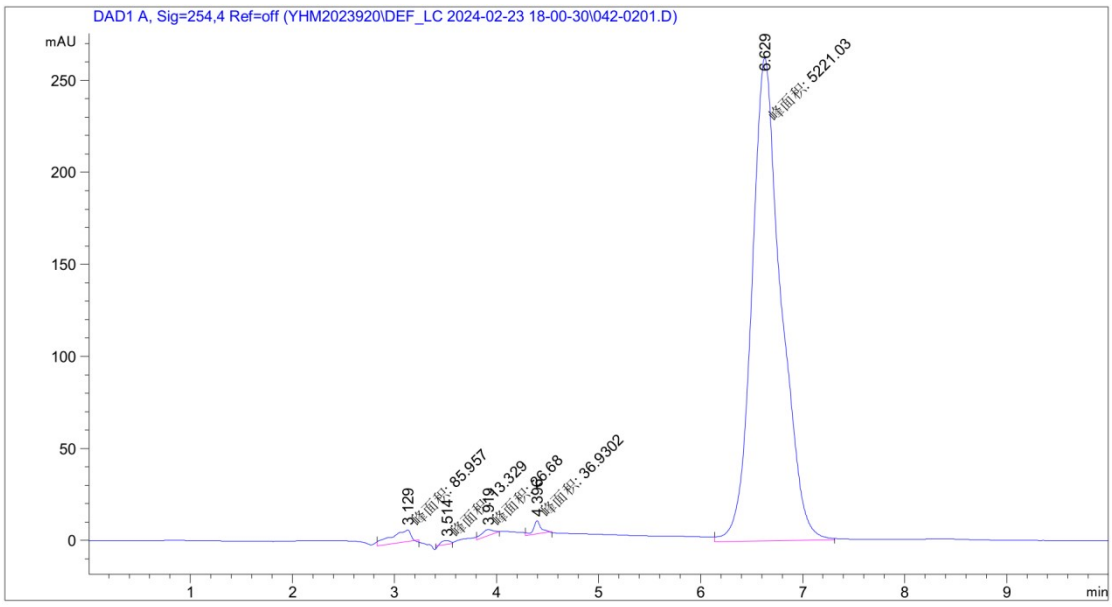
B24, 96.57%, t_R =6.595 min



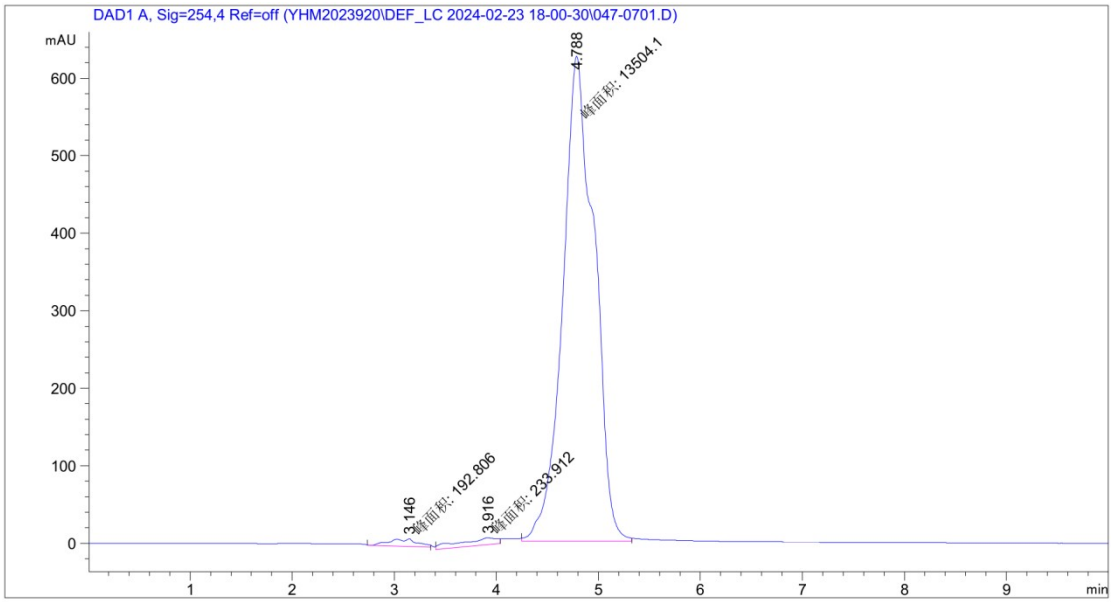
B25, 97.20%, t_R =4.262 min



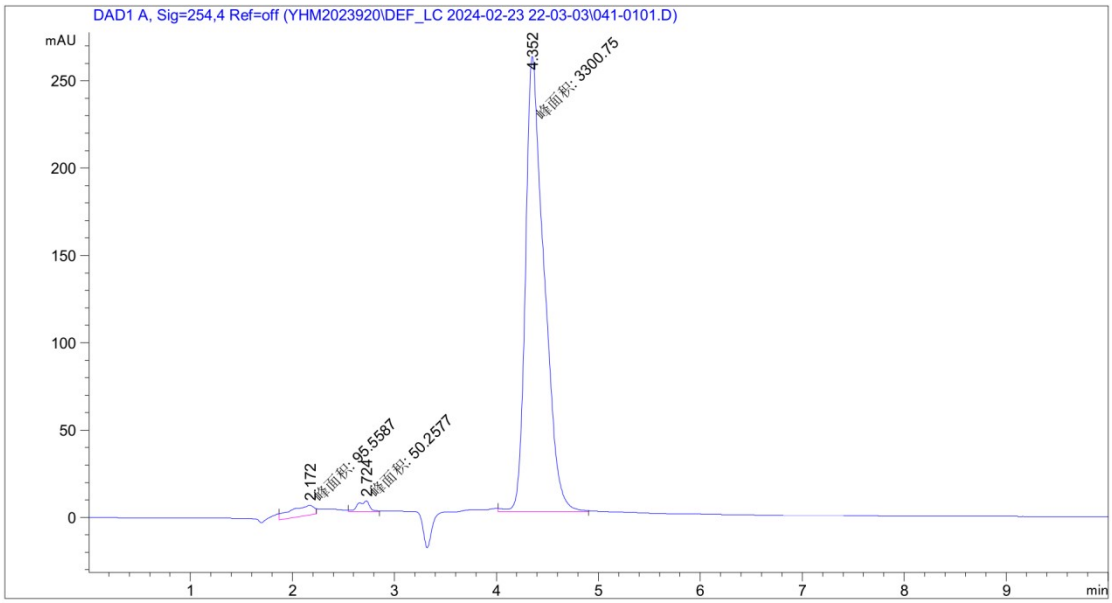
B26, 96.97%, t_R =6.629 min



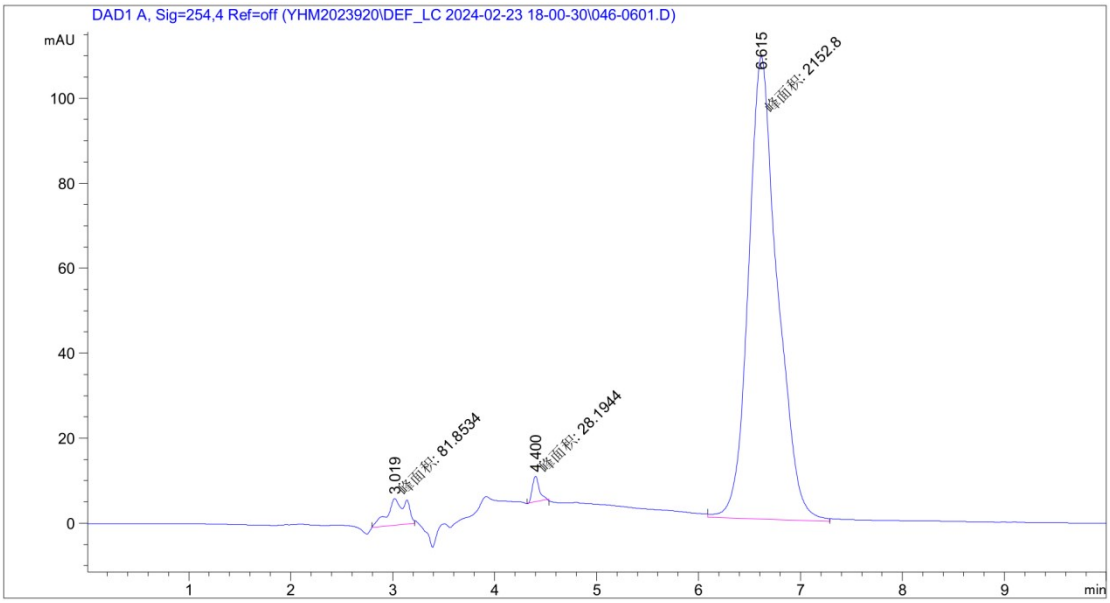
B27, 96.94%, t_R =4.788 min



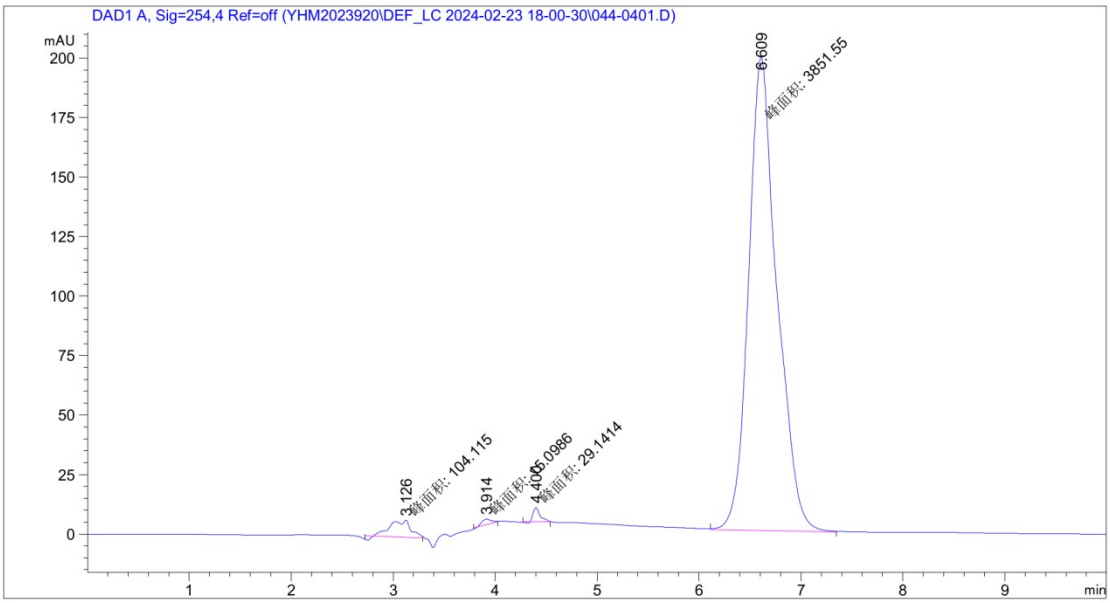
B28, 95.77%, t_R =4.352 min



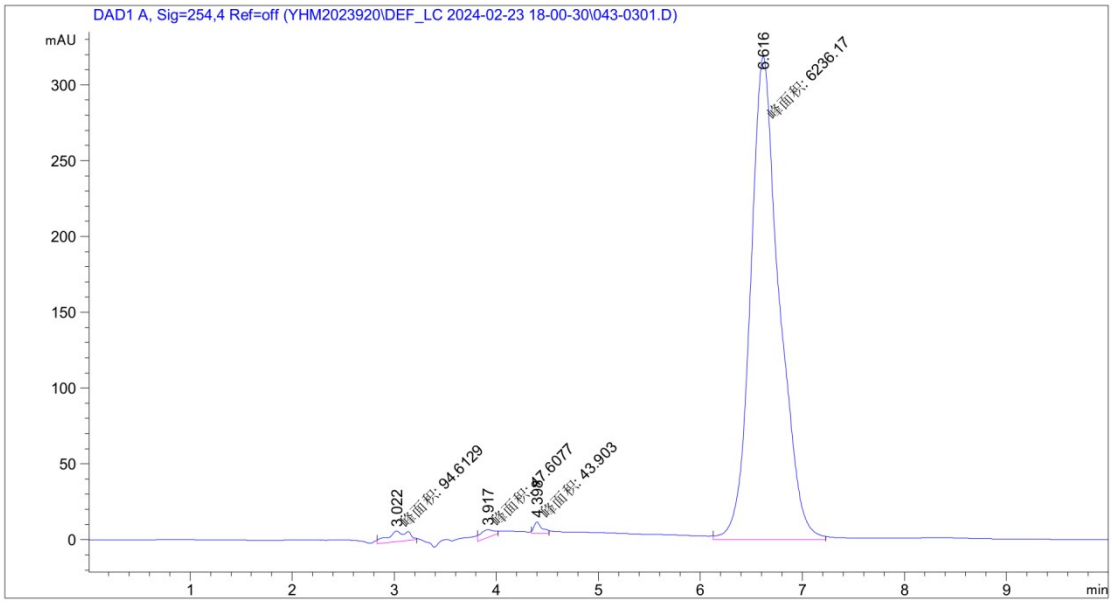
C1, 95.14%, t_R =6.615 min



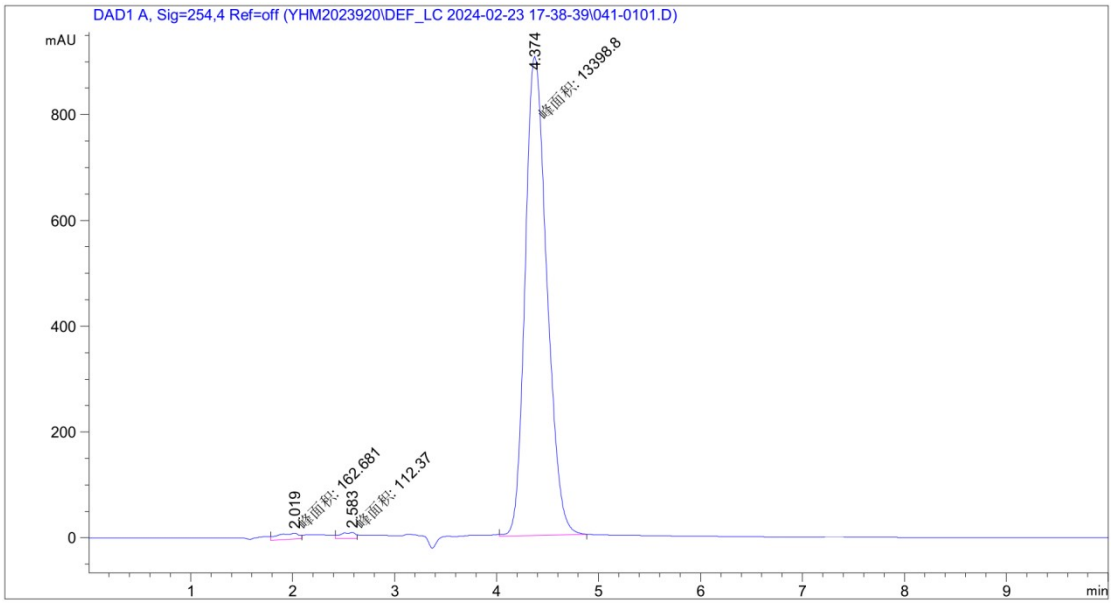
C2, 96.29%, t_R =6.609 min



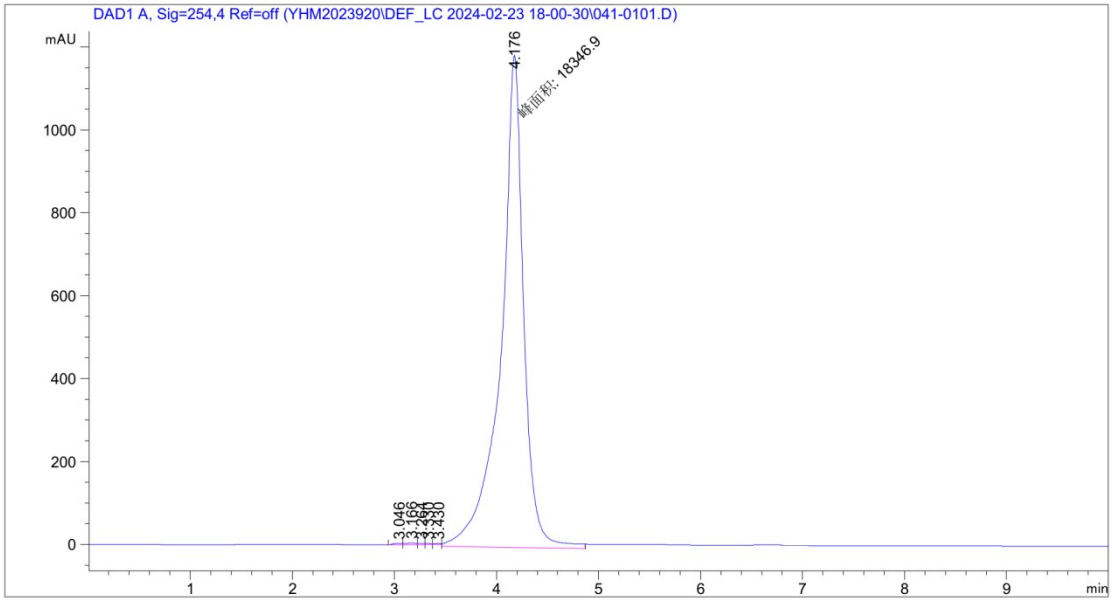
C3, 97.10%, t_R =6.616 min



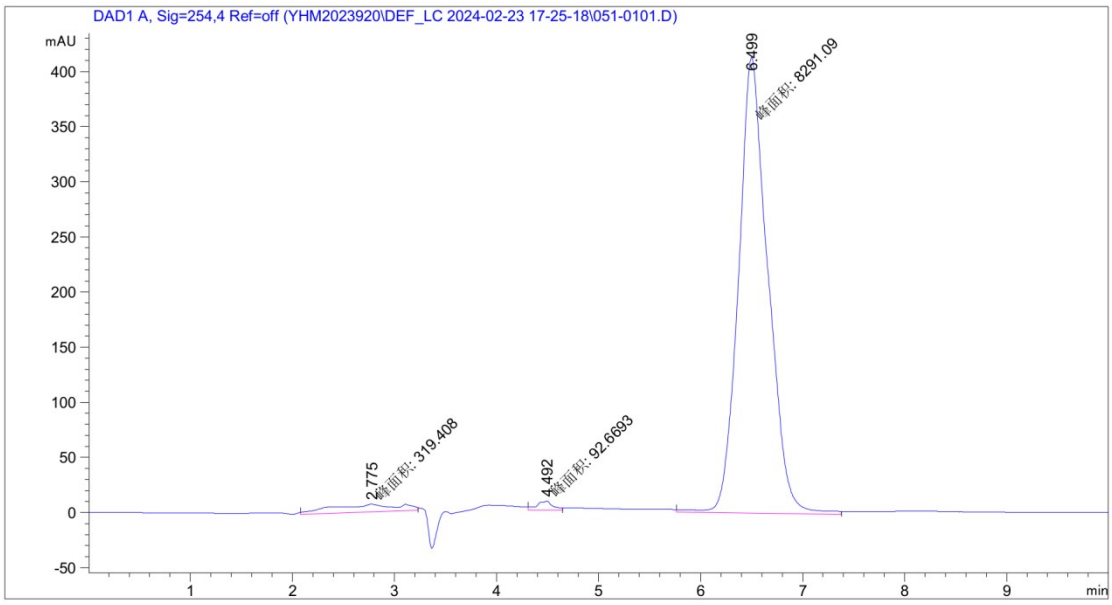
C4, 97.99%, t_R =4.374 min



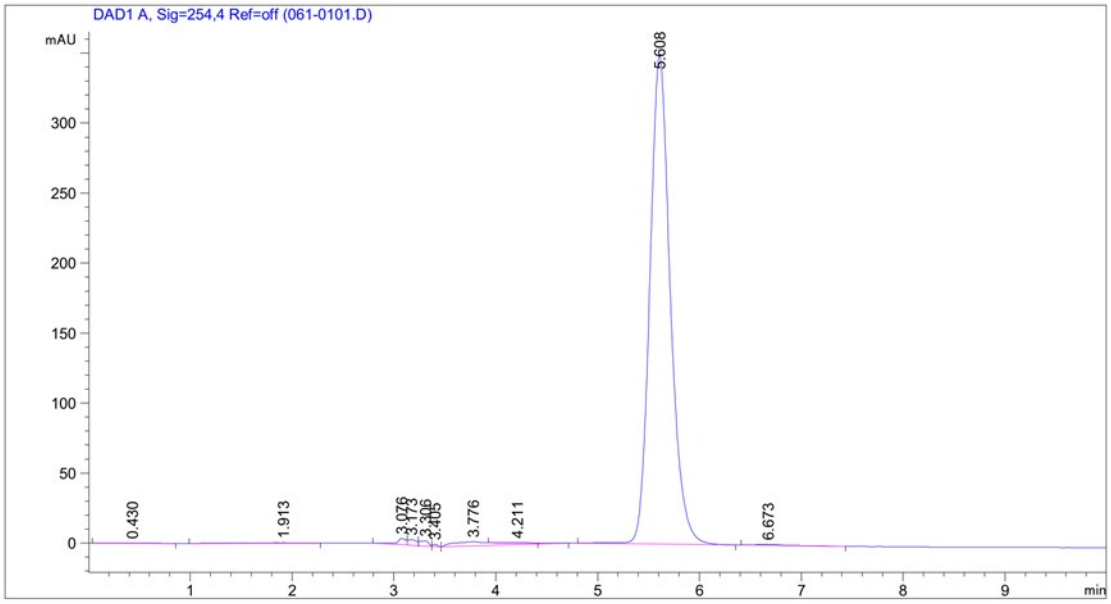
C5, 99.55%, t_R =4.176 min



C6, 95.27%, t_R =6.499 min



C7, 95.73%, t_R =5.608 min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3.173
3.306
3.405
3.776
4.211
5.608
6.673
1
2
3
4
5
6
7
8
9
min

4. Table

Table S1 Lung injury pathological scoring standards.

Pathological features	Score 0	Score 1	Score 2	Score 3
Inflammatory cell infiltration	None	Mild	Moderate	Severe
Alveolar hemorrhage	None	Mild, <25% of the observing field	Moderate, 25–50% of the observing field	Severe, >50% of the observing field
Alveolar edema	None	Mild, <25% of the observing field	Moderate, 25–50% of the observing field	Severe, >50% of the observing field
Pulmonary interstitial edema	None	Mild, <25% of the observing field	Moderate, 25–50% of the observing field	Severe, >50% of the observing field

Table S2 Primer sequences for quantitative real-time PCR.

Gene name	Forward Primer (5'-3')	Reverse Primer (5'-3')
TNF-α	CAGGGGCCACCACGCTCTTC	TTTGTGAGTGTGAGGGTCTGG
IL-6	GAGGATACCACTCCCAACAGACC	AAGTGCATCATCGTTGTTCATAC
β-actin	CCGTGAAAAGATGACCCAGA	TACGACCAGAGGCATACAG

5. Figure

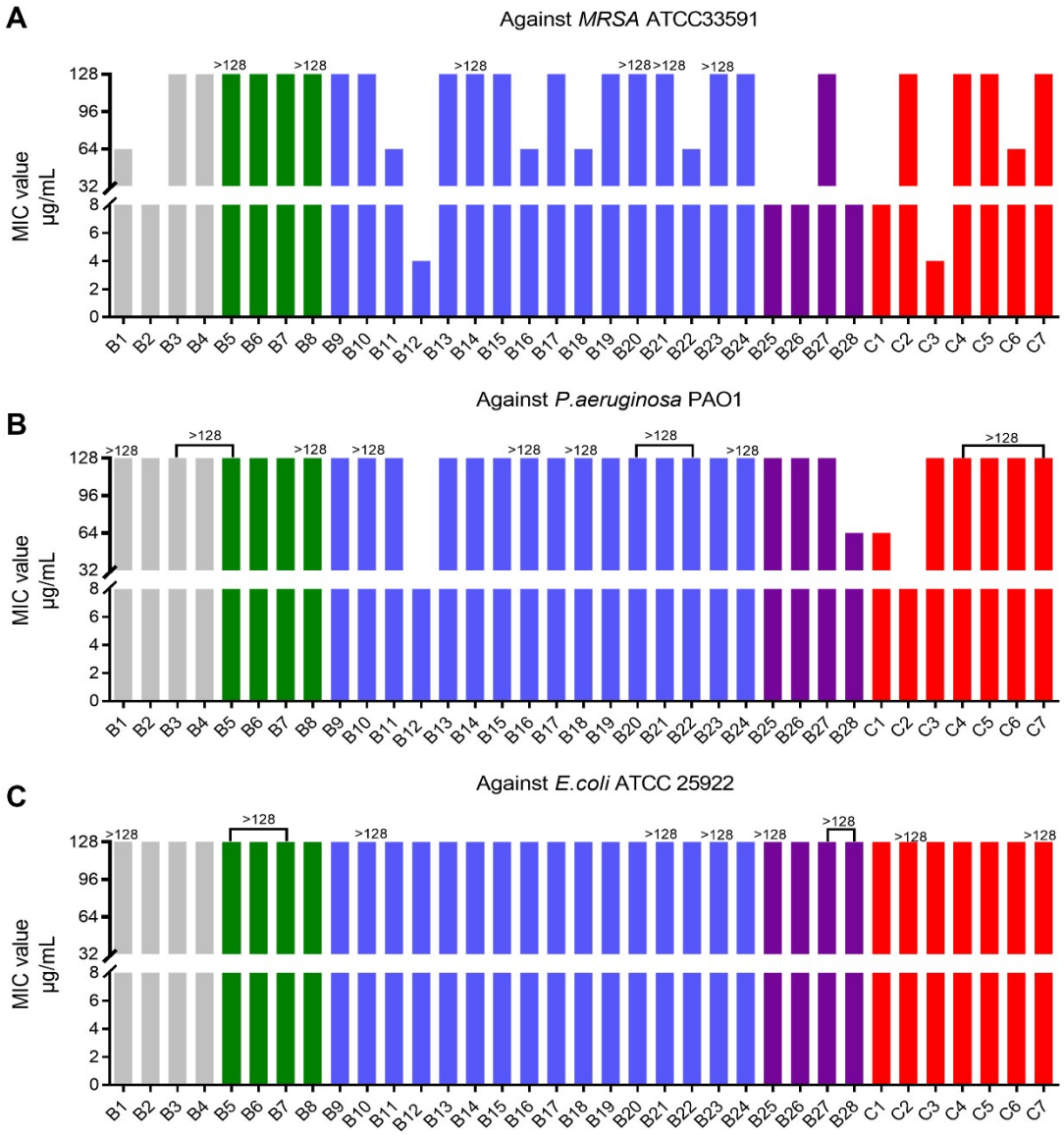

Fig. S1. Structure-activity relationship analysis of berberine derivatives against *MRSA* (A), *P. aeruginosa* (B), and *E. coli* (C).

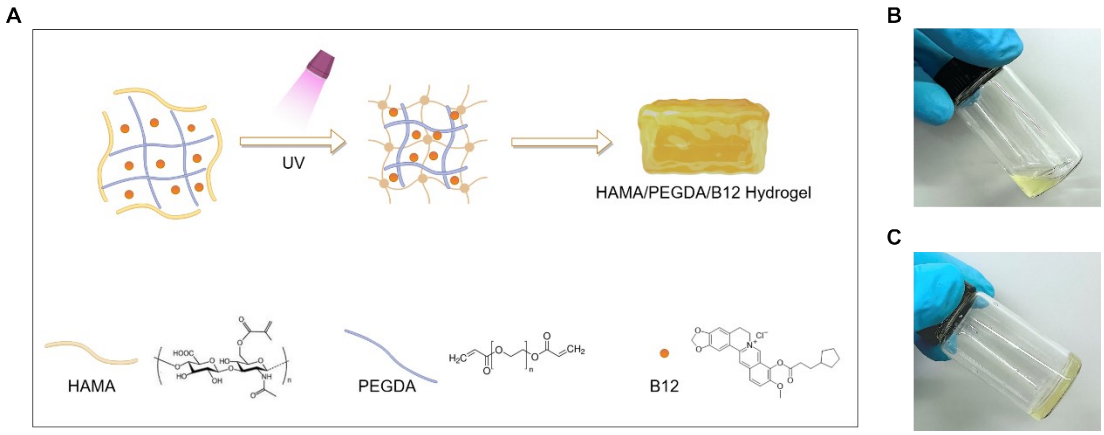

Fig. S2. Preparation of hydrogels. (A) Diagram of HAMA/PEGDA/**B12** hydrogel synthesis. (B-C) Photographs of UV-induced HAMA/PEGDA/**B12** hydrogels.

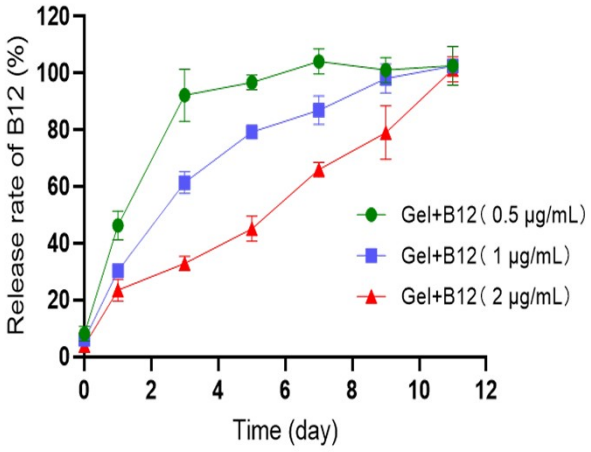

Fig. S3. In *vitro* release profile of **B12** from the fabricated hydrogel over time. Data are presented as mean ± SD (n=3).