Supporting Information

Discovery of Phytoalexin Camalexin Derivatives as Novel Antiviral and Anti-Phytopathogenic-Fungus Agents

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Contents

Chemicals.

Reagents were purchased from commercial sources and were used as received. All anhydrous solvents were dried and purified by standard techniques prior to use.

Instruments.

The melting points of the products were determined on an X-4 binocular microscope (Gongyi Yuhua Instrument Co., China) and the thermometer was not corrected. NMR spectra were acquired with a Bruker 400 MHz (100 MHz for ^{13}C) instrument at room temperature. Chemical shifts were measured relative to residual solvent peaks of $CDCl_3$ (^1H: δ = 7.26 ppm; ^{13}C: δ = 77.0 ppm) or DMSO-d_6 (^1H: δ = 2.50 ppm; ^{13}C: δ = 39.5 ppm) with tetramethylsilane as internal standard. The following abbreviations were used to designate chemical shift multiplicities: s = singlet, d = doublet, t = triplet, m = multiplet, and bs = broad singlet. HRMS data were obtained with an FT-ICR MS spectrometer (Ionspec, 7.0 T). The *in vitro* TMV rod assembly inhibition and 20S CP disk assembly inhibition was tested via transmission electron microscopy (Tecnai G2 F20).

Procedures for the preparation of compounds 3a–3d

2,2,2-Trichloroethyl chloroformate (1.15 mmol) was slowly added to a solution of the thiazole (1 mmol) in 1,2-dichloroethane (7 mL) with magnetic stirring at 0 °C. Immediately after that, indole (1 mmol) was added. Then gradually, during 30 min, triethylamine (1 mmol), dissolved in 1,2-dichloroethane (2 mL), was added. The reaction mixture was stirred for 1 h at 0 °C. After completion of the reaction, the mixture was transferred to a separatory funnel with dichloromethane (10 mL) and was

consecutively extracted with equal volumes of aqueous HCl (10%), Na_2CO_3 (5%), and brine. The combined organic layers were dried (Na_2SO_4), and the solvent was removed under reduced pressure. The residue was purified by flash chromatography on a silica gel using petroleum ether and ethyl acetate (v/v = 5: 1) as the eluent to give corresponding product.[1]

2,2,2-Trichloroethyl 2-(1*H*-indol-3-yl)thiazole-3(2*H*)-carboxylate (**3a**): White solid, yield 91 %; mp 130–131 °C (lit.[1] 135–137 °C); ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.17 (s, 1H), 7.78 (dd, J = 18.7, 8.3 Hz, 1H), 7.39 (d, J = 8.0 Hz, 1H), 7.26 (d, J = 8.1 Hz, 2H), 7.19 (d, J = 7.8 Hz, 1H), 6.96 (d, J = 8.8 Hz, 1H), 6.74 (s, 1H), 5.91 (dd, J = 16.4, 4.8 Hz, 1H), 4.96–4.58 (m, 2H); $C_{14}H_{12}Cl_3N_2O_2S$ $[M+H]^+$ 376.9680, found (ESI^+) 376.9685.

2,2,2-Trichloroethyl 2-(5-methoxy-1*H*-indol-3-yl)thiazole-3(2*H*)-carboxylate (**3b**): White solid, yield 94 %; mp 152–153 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.17 (s, 1H), 7.33 – 7.07 (m, 3H), 6.88 (t, J = 9.7 Hz, 2H), 6.72 (s, 1H), 5.96–5.75 (m, 1H), 4.88–4.68 (m, 1H), 4.68–4.48 (m, 1H), 3.85 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 154.2, 150.8, 132.0, 123.8, 123.5, 121.1, 119.9, 112.5, 105.7, 105.4, 102.0, 75.2, 61.2, 60.1, 55.9; $C_{15}H_{14}Cl_3N_2O_3S$ $[M+H]^+$ 406.9785, found (ESI^+) 406.9787.

2,2,2-Trichloroethyl 2-(5-bromo-1*H*-indol-3-yl)thiazole-3(2*H*)-carboxylate (**3c**): White solid, yield 76 %; mp 189–190 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.22 (s, 1H), 7.86 (d, J = 14.5 Hz, 1H), 7.30 (d, J = 8.5 Hz, 1H), 7.24 (d, J = 12.2 Hz, 2H), 6.87 (d, J = 6.6 Hz, 1H), 6.72 (d, J = 4.7 Hz, 1H), 5.90 (d, J = 12.1 Hz, 1H), 4.94–4.56 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 150.8, 135.4, 125.6, 124.2, 122.5, 121.2, 120.0,

113.7, 113.1, 105.8, 105.5, 75.2, 60.8, 59.6; $C_{14}H_{11}BrCl_3N_2O_2S$ $[M+H]^+$ 454.8785, found (ESI^+) 454.8789.

2,2,2-Trichloroethyl 2-(6-bromo-1*H*-indol-3-yl)thiazole-3(2*H*)-carboxylate (**3d**): White solid, yield 60 %; mp 132–133 °C; ^1H NMR (400 MHz, $CDCl_3$) δ 8.23 (s, 1H), 7.61 (d, J = 16.1 Hz, 1H), 7.48 (d, J = 9.5 Hz, 1H), 7.23 (s, 1H), 7.17 (d, J = 15.2 Hz, 1H), 6.88 (s, 1H), 6.70 (s, 1H), 5.87 (d, J = 13.0 Hz, 1H), 4.80–4.59 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 150.8, 137.6, 123.6, 123.2, 121.1, 119.8, 117.1, 116.3, 114.6, 105.8, 105.5, 75.2, 60.7, 59.7; $C_{14}H_{11}BrCl_3N_2O_2S$ $[M+H]^+$ 454.8785, found (ESI^+) 454.8786.

Procedures for the preparation of compounds 4a–4d

The corresponding compound 3 (1 mmol) was dissolved in CH_3CN (15 mL/mmol), then DDQ (1 mmol) was added, and the reaction mixture was magnetically stirred at 0 °C. After completion of the reaction, the solvent was evaporated under reduced pressure. The residue was purified by flash chromatography on a silica gel using petroleum ether and ethyl acetate (v/v = 10: 1) as the eluent to give corresponding compounds **4a**–**4d**.

Camalexin (**4a**): White solid, yield 65 %; mp 142–143 °C (lit.[1] 137–138 °C); ^1H NMR (400 MHz, $CDCl_3$) δ 8.94 (s, 1H), 8.29 (d, J = 9.2 Hz, 1H), 7.87 (d, J = 8.7 Hz, 2H), 7.44 (d, J = 8.6 Hz, 1H), 7.32 (dd, J = 7.4, 3.7 Hz, 2H), 7.29 (s, 1H); $C_{11}H_9N_2S$ $[M+H]^+$ 201.0481, found (ESI^+) 201.0476.

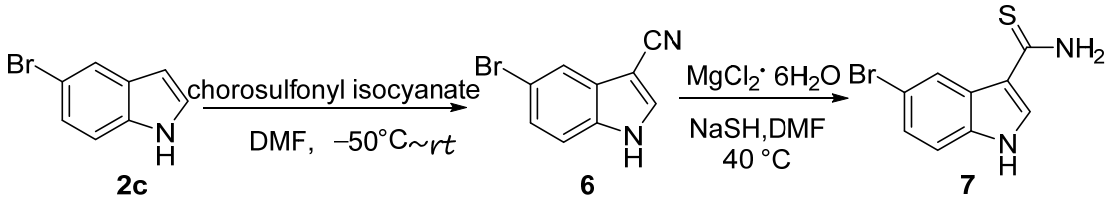
2-(5-Methoxy-1H-indol-3-yl)thiazole (**4b**): Red solid, yield 49 %; mp 114–115 °C (lit.[2] 112–112.5 °C); ^1H NMR (400 MHz, $CDCl_3$) δ 9.29 (s, 1H), 7.82 (s, 1H), 7.72 (d,

J = 5.6 Hz, 2H), 7.22 (d, *J* = 8.9 Hz, 2H), 6.91 (s, 1H), 3.89 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 163.96, 155.38, 142.24, 131.65, 125.57, 125.16, 115.82, 113.36, 112.68, 111.66, 102.11, 55.90; $C_{12}H_{11}N_2OS$ $[M+H]^+$ 231.0587, found (ESI^+) 231.0589.

2-(5-Bromo-1*H*-indol-3-yl)thiazole (**4c**): White solid, yield 76 %; mp 189–190 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.52 (s, 1H), 8.16 (d, *J* = 8.6 Hz, 1H), 7.84 (d, *J* = 3.3 Hz, 1H), 7.81 (d, *J* = 2.4 Hz, 1H), 7.59 (s, 1H), 7.39 (d, *J* = 8.6 Hz, 1H), 7.26 (s, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 162.6, 142.6, 135.0, 126.3, 126.2, 125.5, 123.3, 116.3, 114.9, 113.0, 112.0; $C_{11}H_8BrN_2S$ $[M+H]^+$ 278.9586, found (ESI^+) 278.9583.

2-(6-Bromo-1*H*-indol-3-yl)thiazole (**4d**): White solid, yield 53 %; mp 211–212 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.60 (s, 1H), 8.46 (s, 1H), 7.83 (d, *J* = 12.4 Hz, 2H), 7.37 (s, 1H), 7.30 (d, *J* = 8.7 Hz, 2H);^{13}C NMR (100 MHz, $CDCl_3$) δ 162.6, 142.7, 137.1, 124.9, 124.8, 123.6, 122.1, 116.8, 116.3, 114.5, 112.7; $C_{11}H_8Br\ N_2S$ $[M+H]^+$ 278.9586, found (ESI^+) 278.9589.

Procedures for the preparation of compounds 6–7



To a solution of indole **2c** (10 mmol) in DMF was added chlorosulfonyl isocyanate (12 mmol) at −50 °C in an argon atmosphere. The temperature was raised to −10 °C, and then the solution was brought to room temperature after it was stirred for 1.5 h.

The resulting solution was poured into ice water, kept there for 30 min, and filtered to give **6**.[3]

5-Bromo-1*H*-indole-3-carbonitrile (**6**): Yellow solid, yield 99%; mp 185–186 °C (lit.[3] 178–179 °C); ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.18 (s, 1H), 7.93 (s, 1H), 7.76 (s, 1H), 7.45 (d, J = 8.7 Hz, 1H), 7.39 (d, J = 8.7 Hz, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 135.9, 134.0, 128.4, 126.1, 120.7, 115.6, 115.0, 114.4, 84.0.

To a slurry of 70% sodium hydrosulfide hydrate (24 mmol) and magnesium chloride hexahydrate (8 mmol) in 10 mL of DMF was added the corresponding precursor material (**6**, 8 mmol) in one portion, and the mixture was stirred at 40 °C overnight. The resulting green slurry was poured into 200 mL of water, and the resulting precipitates were collected by filtration. The crude product was resuspended in 1 N HCl, stirred for 20 min, filtered, and washed with water to give compound 5-Bromo-1*H*-indole-3-carbothioamide (**7**): White solid, yield 89%; mp 145–147 °C (lit.[3] 141–142 °C); ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.00 (s, 1H), 9.05 (s, 1H), 8.95 (s, 1H), 8.91 (s, 1H), 8.15 (s, 1H), 7.43 (d, J = 8.5 Hz, 1H), 7.30 (d, J = 8.5 Hz, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 193.5, 136.0, 129.2, 128.5, 125.1, 124.6, 116.0, 114.5, 114.2.

Detailed biological assay methods

Phytotoxic Activity. The growing 3–5 leaf stage tobaccos (*Nicotiana tabacum var Xanthi nc*) were selected. The compound solution (500 μg/mL) was sprayed on the leaves and then tested the plant height and weight changes after 0, 3, 7 and 10 days respectively. There are three replicates for each compound.

Antiviral Biological Assay.[4]

Purification of Tobacco Mosaic Virus.

Using Gooding's method, [5] the upper leaves of *Nicotiana tabacum* L. inoculated with TMV were selected and ground in phosphate buffer and then filtered through double-layer pledget. The filtrate was centrifuged at 10000 g, treated with PEG twice, and centrifuged again. The whole experiment was processed at 4 °C. Absorbance value was estimated at 260 nm by ultraviolet spectrophotometer.

$$Virus\ concn = (A_{260} \times dilution\ ratio) / E_{1cm}^{0.1\%,\,260nm}$$

Protective Effect of Compounds against TMV in Vivo.

The solution (100 μg/mL or 500 μg/mL) of synthesized compounds or controls was sprayed on growing *Nicotiana. tabacum* L. leaves (at least 3 leaves) of the same age. In another pot, the leaves were sprayed with the solvent as a control. The leaves were then inoculated with the virus (6×10^{-3} μg/mL) after 12 h by rubbing emery. First, spread a layer of 600 mesh emery on the tested leaves, then brush the leaves 1 to 2 times along the direction of the vein with a brush stained with TMV (6×10^{-3} μg/mL), and then wash the leaves with clear water. The total local lesion numbers appearing on the leaves 3−4 days after inoculation were recorded.[5] There are three replicates for each compound.

Inactivation Effect of Compounds against TMV in Vivo.

To test viral inhibition, equal volumes of the virus (6×10^{-3} μg/mL) and the solution (100 μg/mL or 500 μg/mL) of synthesized compounds or controls were mixed together for 30 min. The mixture was then inoculated into the growing *N.*

tabacum L leaves of the same age by rubbing emery, and another pot was inoculated with the mixture of solvent and the virus (6×10^{-3} μg/mL) by rubbing emery as the control. Then, the leaves were washed with water and dried. The local lesion numbers were recorded 3−4 days after inoculation.[6] There are three replicates for each compound.

Curative Effect of Compounds against TMV in Vivo.

TMV (concentration of 6.0×10^{-3} μg/mL) was inoculated on the growing leaves of *N. tabacum* L. of the same age by rubbing emery. Then, the leaves were washed with water and dried. The solution (100 μg/mL or 500 μg/mL) of synthesized compounds or controls was smeared on the inoculated leaves, while inoculated leaves in another pot were smeared with the solvent as a control. The local lesion numbers were recorded 3−4 days after inoculation.[6]

There are three replicates for each compound. The *in vivo* inhibition rates of the compound were then calculated according to the following formula ("av" means average, and controls were not treated with compound).

Inhibition rate (%) = [(av local lesion no. of control − av local lesion no. of drug-treated)/av local lesion no. of control] × 100%

Detailed bioassay procedures for the fungicidal activities

In Vitro Antifungal Bioassay.[7] The fungicidal activities of compounds were evaluated in mycelial growth tests conducted in artificial media against 14 plant pathogens at a rate of 50 μg/mL. Each test compound was dissolved in a suitable amount of acetone and diluted with water containing 0.1% TW-80 to a concentration

of 500 μg/mL. To each petri dish was added 1 mL of the test solution and 9 mL of culture medium to make a 50 μg/mL concentration of the test compound, while in another petri dish was added 1 mL distilled water containing 0.1% TW-80 and 9 mL of culture medium as a blank control. A 4 mm diameter of hyphal growth was cut using a hole puncher on a growing fungal culture and the hyphae were moved to the petri dish containing the test compound. Each assay was performed three times. The dishes were stored in controlled environment cabinets (24±1°C) for 4 days, after which the diameter of mycelial growth was measured and the percentage inhibition was calculated using the following equation: *Percentage inhibition (%)* = (*averaged diameter of mycelia in blank controls – averaged diameter of mycelia in medicated tablets*) / (*averaged diameter of mycelia in blank controls* – 4 mm) × 100.

Mode of Action Studies

In vitro TMV rod assembly inhibition. TMV purification was performed according to the instructions by Leberman.[8] TMV RNA was purified by RNApure virus kit (CoWin Biosciences) and TMV capsid protein (TMV CP) was isolated using glacial acetic acid as described by Fraenkel-Conrat.[9] Before assembly, 20S CP Disk was prepared by incubating 23.5 mg/mL TMV CP in 0.1 M phosphate buffer (pH 7.0) at 20 °C for 12 h. After incubation, *in vitro* TMV reconstitution reactions were performed by adding 7.5 μL of phosphate buffer (0.1 M, pH 7.0), 2 μL of 20S Disk (23.5 mg/mL) and 0.5 μL of TMV RNA (2 μg/μL). The assembly reaction mixture was incubated at 20 °C for 12 h and could be then transferred into the copper grid for transmission electron microscopy (TEM) assay. The assembly reaction mixture (5 μL)

was mixed with 5 μL 0.1 M phosphate buffer (pH 7.0) and dropped onto the copper film waiting for 5 minutes. After the incubation, the droplet was removed by filter paper and negatively stained by 2% phosphotungstic acid (pH 7.0) for three minutes. After removing the staining agent, the copper was placed at 37 ℃ for 2 h for drying. The morphology of the reconstituted TMV rods was imaged at 200 keV on a CCD camera. For the inhibition tests with the compounds, in vitro TMV reconstitution inhibition reactions were performed by adding 7.4 μL of phosphate buffer (0.1 M, pH 7.0), 2 μL of 20S Disk (2 mg/mL), 0.5 μL of TMV RNA (2 μg/μL) and 0.1 μL of DMSO or the compound (10 μM). All treatments were repeated over time to validate the results.

In vitro 20S CP Disk assembly inhibition. For the inhibition tests with the compounds, TMV CP was first adjusted to 23.5 mg/mL with 0.1 M phosphate buffer (pH 7.0). *In vitro* 20S CP Disk assembly reactions were performed by adding 4.9 μL of phosphate buffer (0.1 M, pH 7.0), 5 μL TMV CP (23.5 mg/mL) and 0.1 μL DMSO or the compound (10 μM). The assembly reaction was incubated at 20 °C for 12 h. The morphology of the 20S CP Disk was imaged via TEM at 200 keV on a CCD camera. All treatments were repeated over time for confirmation.

Fluorescence Spectroscopy. TMV CP was purified according to operation instructions as reported previously by Fraenkel-Conrat.[9] Fluorescence spectroscopy measurement was performed on an F-2700 fluorescence spectroscopy instrument (Hitachi, Tokyo, Japan). Fluorescence emission spectra were recorded in a 1 cm quartz cell at an excitation wavelength of 278 nm with the wavelength ranging from

290 to 420 nm at 298 K, and the emission and excitation slit width was set at 5 nm. Two milliliter of 0.02 mM TMV-CP in SEC buffer solution (14 mM β-mercaptoethanol, 0.02 M phosphate buffer, 100 mM NaCl, pH 7.4) was added to a 1.0 cm quartz cell. Then, four microliters of 0.01 M **5a** or **10f** was manually titrated into the TMV-CP solution with trace syringe. Ribavirin (0.02 M) was used as the control. The ultimate antiviral compounds concentrations were 0, 0.2, 0.4, 0.6, 0.8, 10.0, 12.0, 14.0, 16.0, 18.0, and 20.0 μmol/L.[10]

Binding constant was calculated using fluorescence titration (FT). The relationship between fluorescence quenching intensity and the concentration of the compounds can be described by the modified Stern−Volmer equation: $\lg(F_0 - F)/F = \lg K_a + n \lg Cq$, where n is the number of binding sites per proteins; K_a (unit: L/mol) is the binding constant; Cq (unit: mol/L) is the total concentration of the selected compounds, and F_0 and F (unit: a.u.) are the intensities before and after the addition of the selected compounds, respectively.[10]

Calculation procedures for molecular docking research

The calculation procedures for molecular docking research consist of four steps.[11]

Receptor Preparation. The 3D crystal structure of TMV-CP (PDB code:1EI7) was downloaded from the protein data bank (PDB) and this was used as the receptor for molecular docking. Water molecules were removed from the target protein and hydrogen atoms were added using AutoDock Tools prior to molecular docking.

Ligand preparation. Target compounds are drawn using ChemOffice 2015 as ligands followed by management of its conformer and the minimisation process.

Molecular Docking Using AutoDock Vina. The input files for AutoDock Vina were prepared using AutoDock Tools. The protein was placed in a grid box (grid parameters: center x = 5, center y = -20, center z = 0.8, size x = 60, size y = 60, size z = 56), using AutoDock Vina at 1.00 Å to define the binding site. The docking procedure was performed using the instructed command prompts.

Analysing and Output Visualisation using PyMOL.

The docking poses were ranked according to their docking scores. The scoring function in Auto Dock was used to predict the binding affinity of one ligand to the receptor molecule. The conformation with the lowest binding affinity was selected for further analysis after the docking process. The docking results included the locations of hydrogen bonds and closely interacting residues were performed by PyMOL software.

Reference:

(1) Stremski, Y.; Statkova-Abeghe, S.; Angelov, P.; Ivanov, I. Synthesis of camalexin and related analogues. *J. Heterocyclic Chem.* **2018**, *55*,1589–1595.

(2) Ayer, W. A.; Craw, P. A.; Ma, Y.; Miao, S. Synthesis of camalexin and related phytoalexins. *Tetrahedron* **1992**, *48*, 2919–2924.

(3) Guo, J. C.; Hao, Y. N.; Ji, X. F.; Wang, Z. W.; Liu, Y. X.; Ma, D. J.; Li, Y. Q.; Pang, H. L.; Ni, J. P.; Wang, Q. M. Optimization, structure−activity relationship, and mode of Action of nortopsentin analogues containing thiazole and oxazole moieties. *J. Agric. Food Chem*. **2019**, *67*, 10018−10031.

(4) Wang, Z. W.; Wei, P.; Wang, L. Z.; Wang, Q. M. Design, synthesis, and

anti-tobacco mosaic virus (TMV) activity of phenanthroindolizidines and their analogues. *J. Agric. Food Chem*. **2012**, *60*, 10212−10219.

(5) Gooding, G. V., Jr.; Hebert, T. T. A simple technique for purification of tobacco mosaic virus in large quantities. *Phytopathology* **1967**, *57*, 1285−1290.

(6) Li, S. Z.; Wang, D. M.; Jiao, S. M. In *Pesticide Experiment Methods-Fungicide Sector*; Li, S. Z., Ed.; Agriculture Press of China: Beijing, China, **1991**; 93−94.

(7) Zhao, H. P.; Liu, Y. X.; Cui, Z. P.; Beattie, D.; Gu, Y. C.; Wang, Q. M. Design, synthesis, and biological activities of arylmethylamine substituted chlorotriazine and methylthiotriazine compounds. *J. Agric. Food Chem*. **2011**, *59*, 11711−11717.

(8) Leberman, R. Isolation of plant viruses by means of simple coacervates. *Virol* **1966**, *30*, 341–347.

(9) Fraenkel Conrat, H.; Williams, R. C. Reconstitution of active tobacco mosaic virus fromits inactive protein and nucleic acid components. *Proc. Natl. Acad. Sci.* U S A **1955**, *41*, 690–698.

(10) Wang, T.; Yang, S.; Li, H.; Lu, A.; Wang, Z.; Yao, Y.; Wang, Q. Discovery, structural optimization, and mode of action of essramycin alkaloid and its derivatives as anti-tobacco mosaic virus and anti-phytopathogenic fungus agents. *J. Agric. Food Chem.* **2020**, *68*, 471–484.

(11) Seyedi, S. S.; Shukri, M.; Hassandarvish, P.; Oo, A.; Muthu, S. E.; Abubakar, S.; Zandi, K. Computational approach towards exploring potential anti-chikungunya activity of selected flavonoids. *Sci. Rep.* **2016**, *6*, 24027.

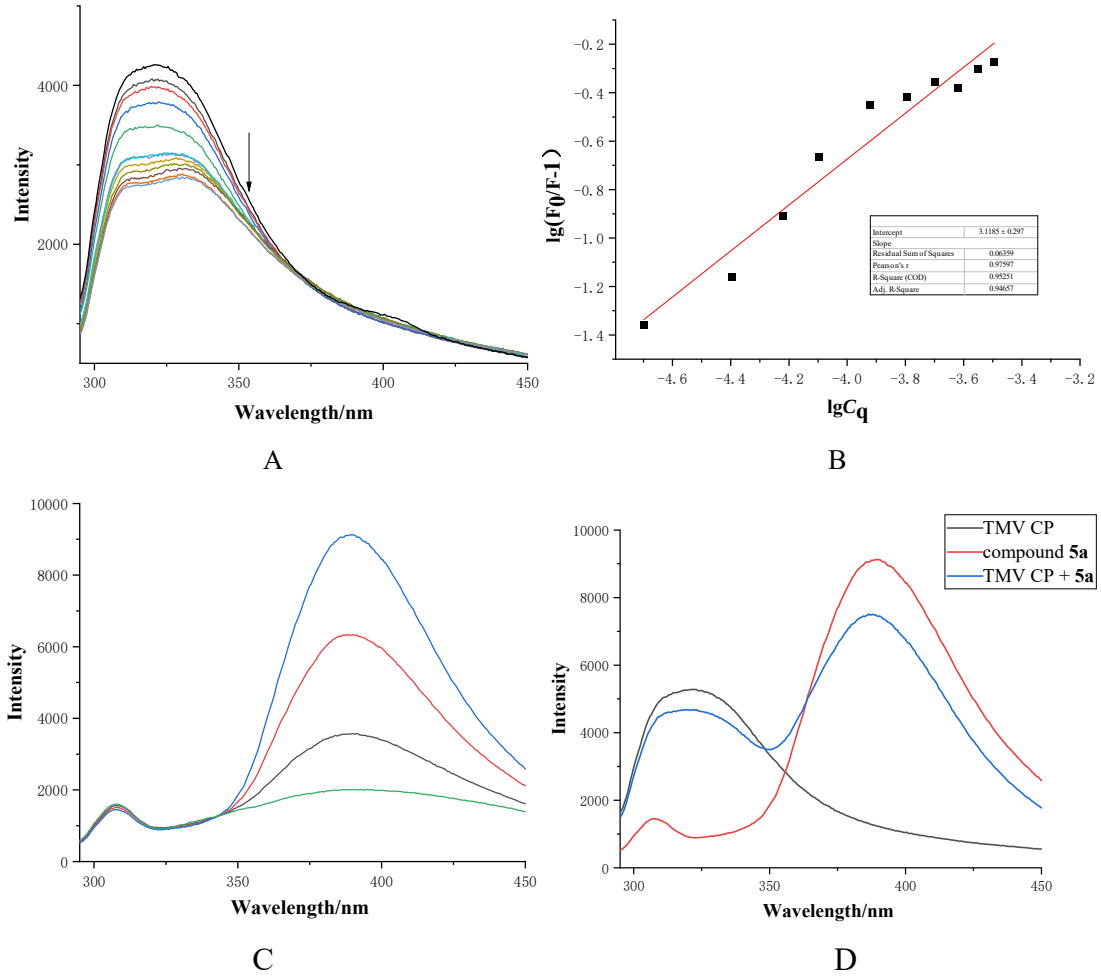
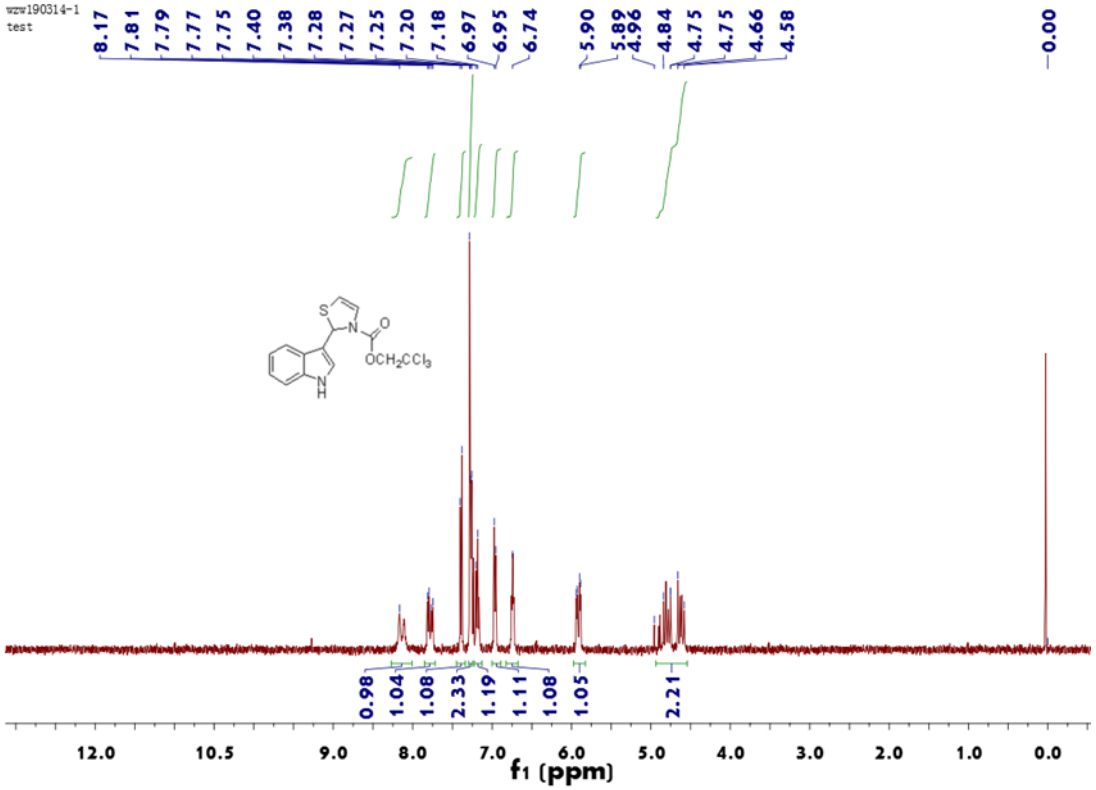
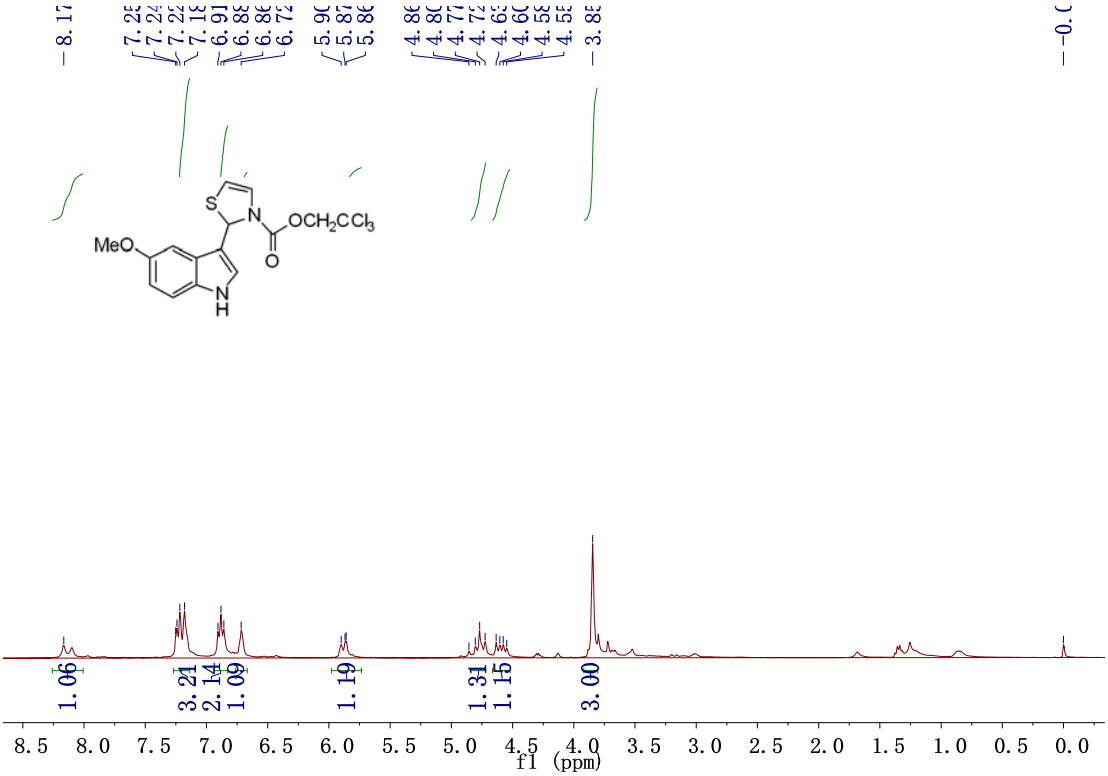


Figure S1 Fluorescence titration graph: (A)Fluorescence titration graph of TMV-CP in the presence of ribavirin with different concentrations (λ_{ex} = 278 nm); (B) The linear relationship for quenching TMV-CP by ribavirin; (C) Fluorescence intensities at 390 nm of **5a** with different concentrations (0.2–0.8 μM) (λ_{ex} = 278 nm); (D) Fluorescence spectra of TMV-CP (0.01 mM), compound **5a** (0.8 μM), and TMV-CP (0.01 mM) with **5a** (0.8 μM).



^{1}H NMR spectrum of **3a**



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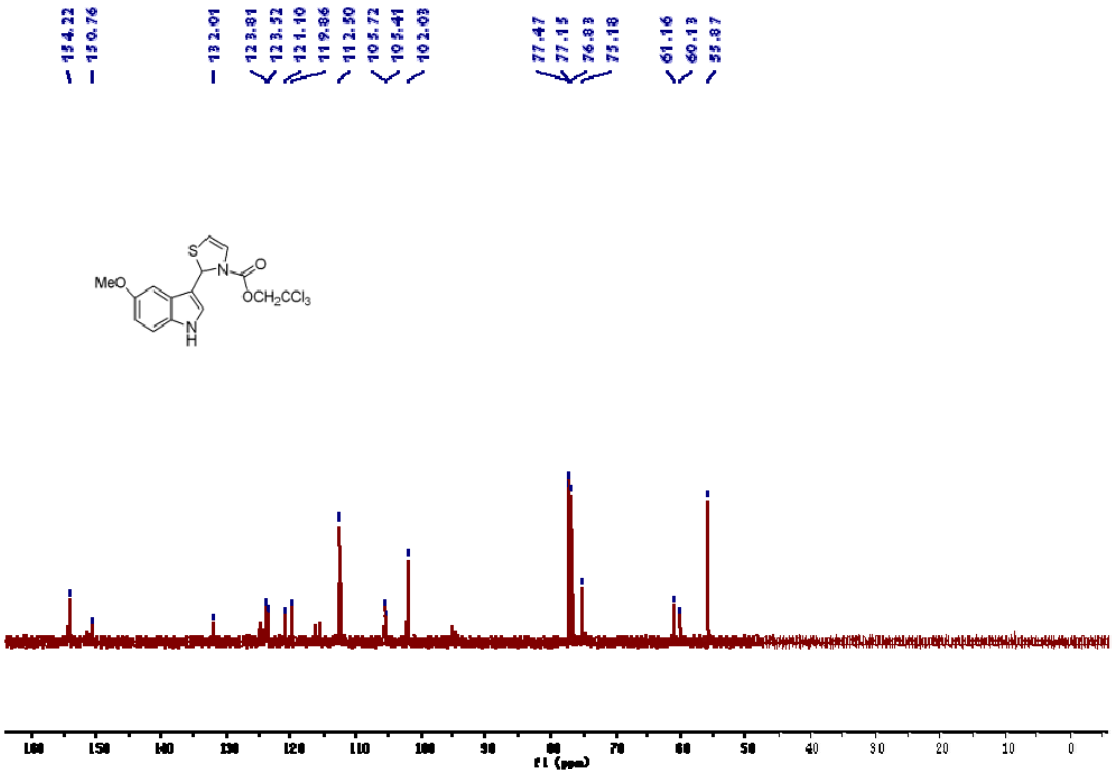
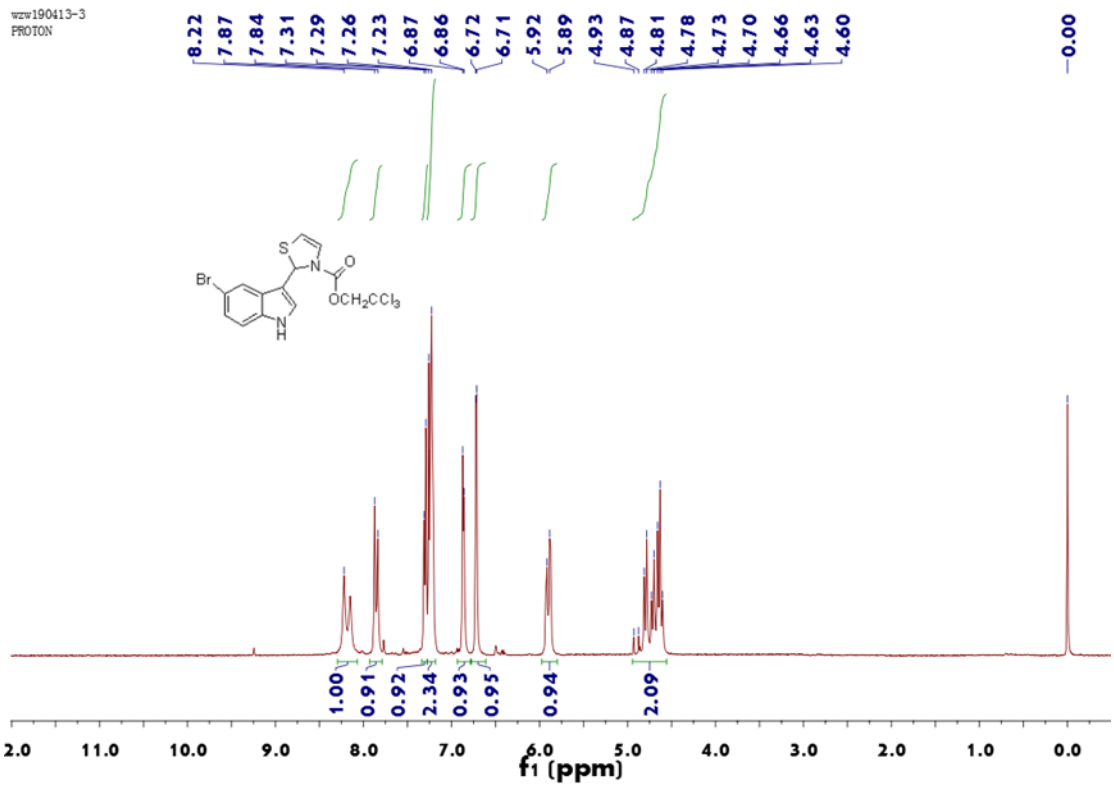
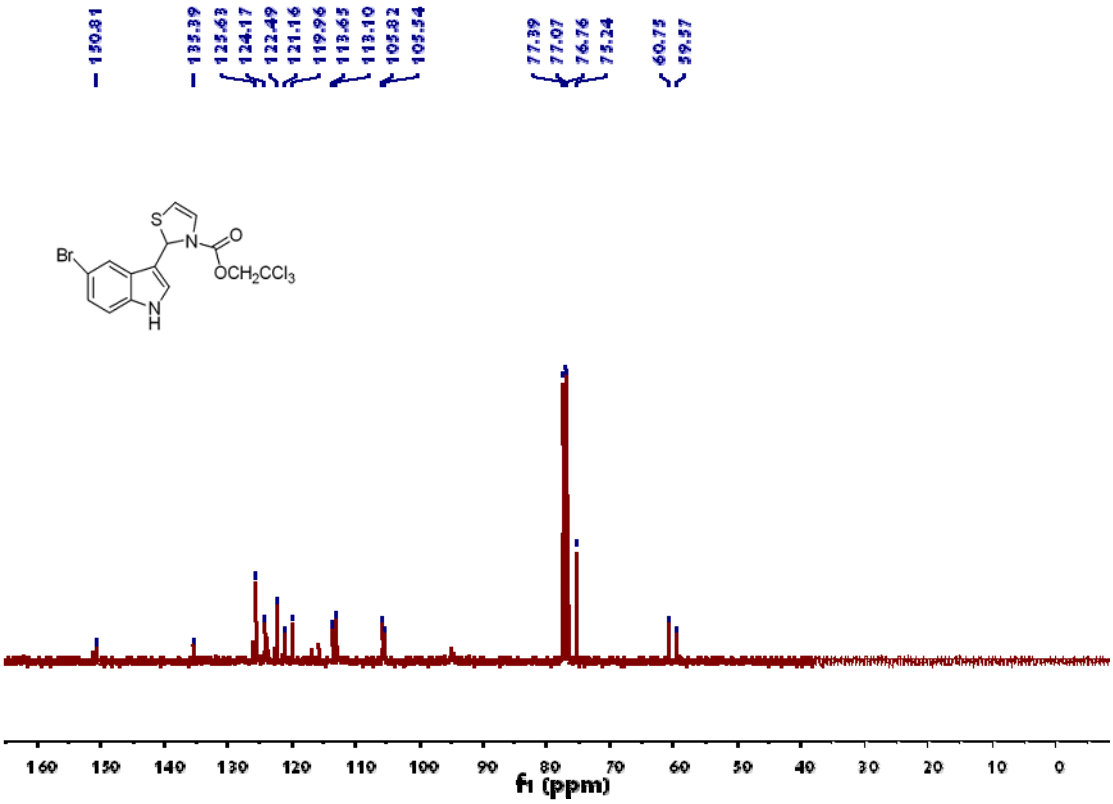


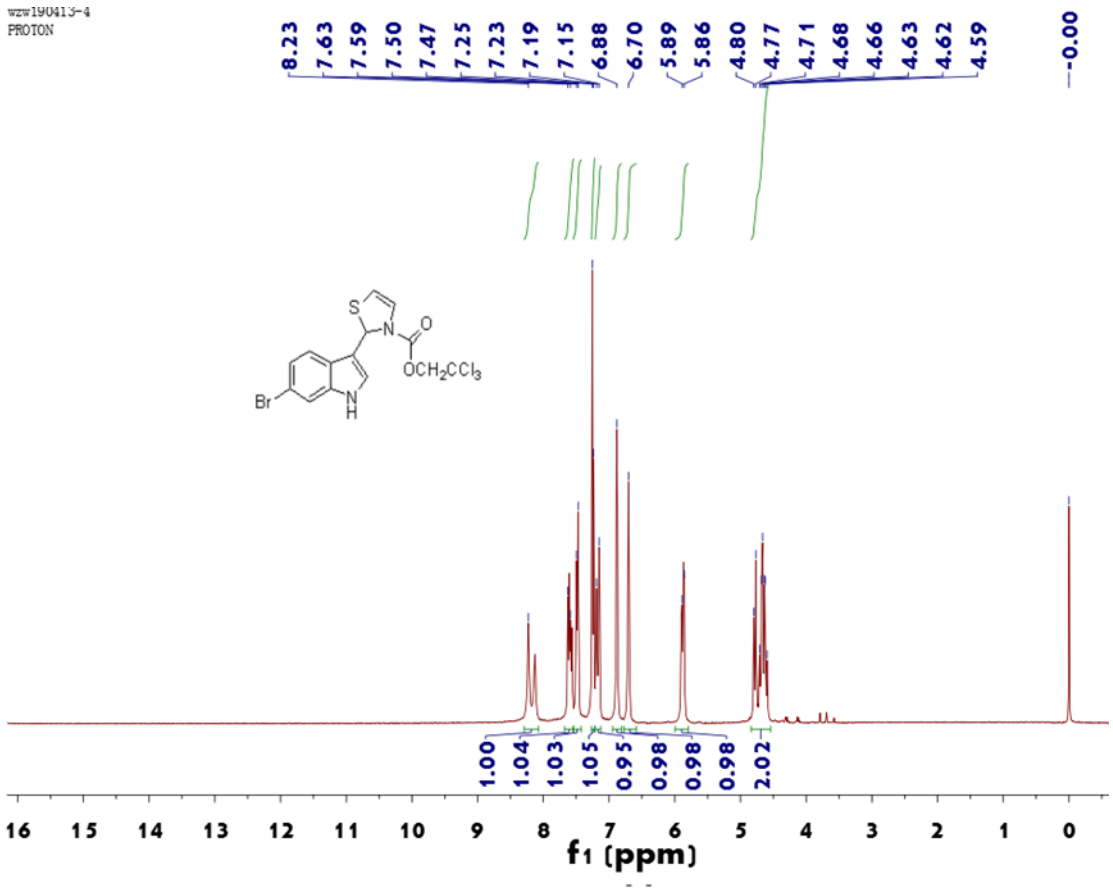
^{13}C NMR spectrum of **3b**



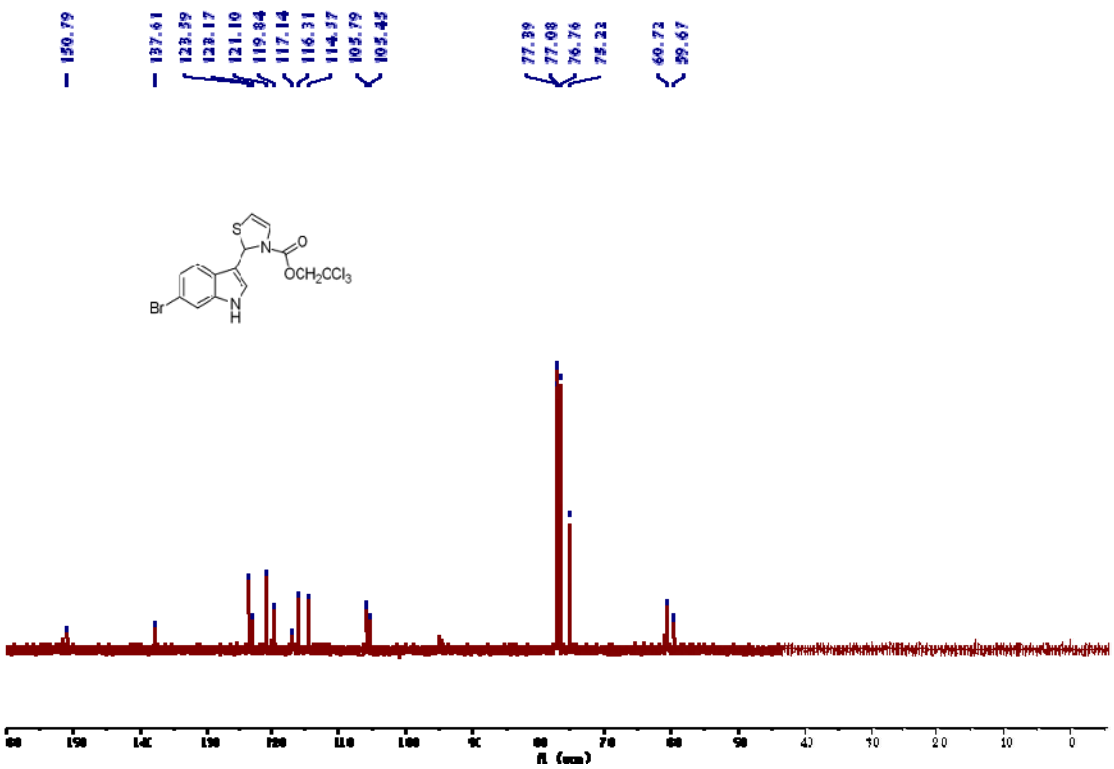
^{1}H NMR spectrum of **3c**



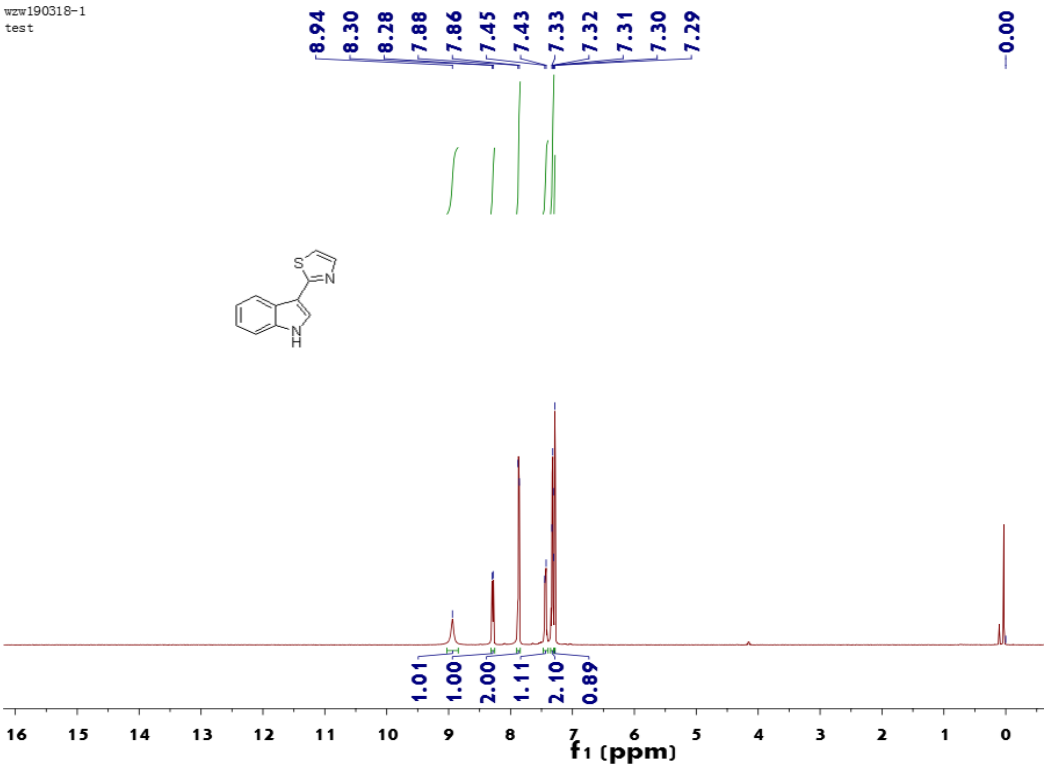
^{13}C NMR spectrum of **3c**



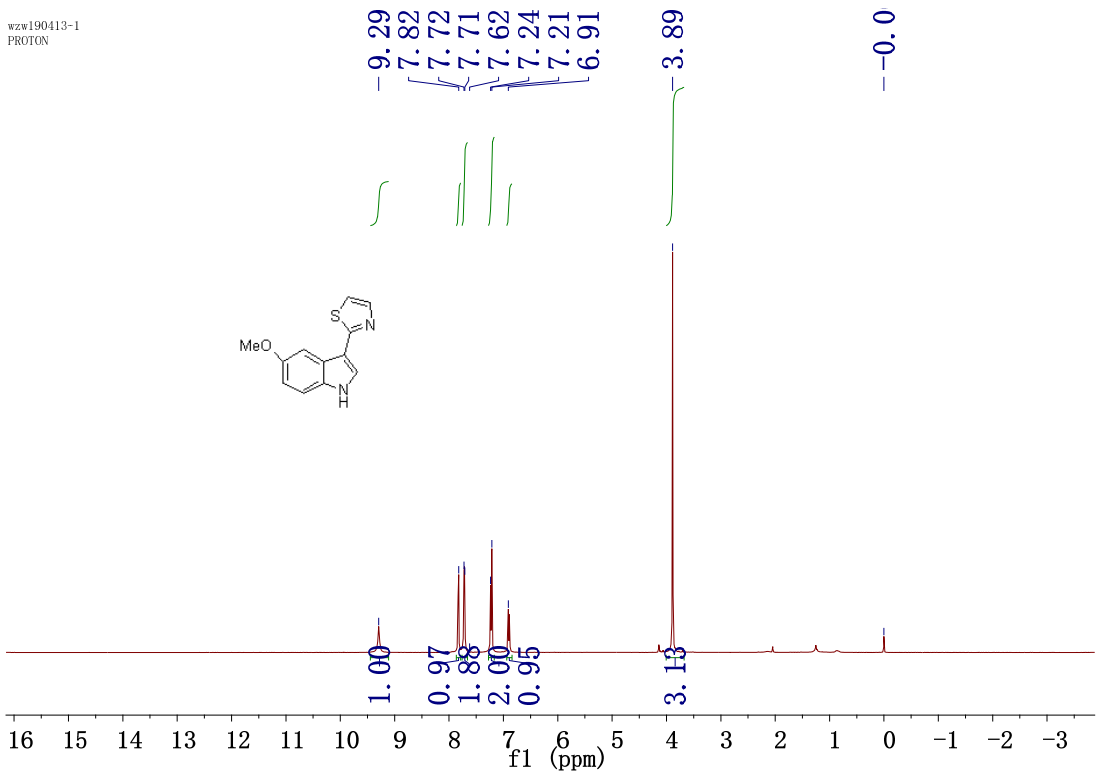
^{1}H NMR spectrum of **3d**



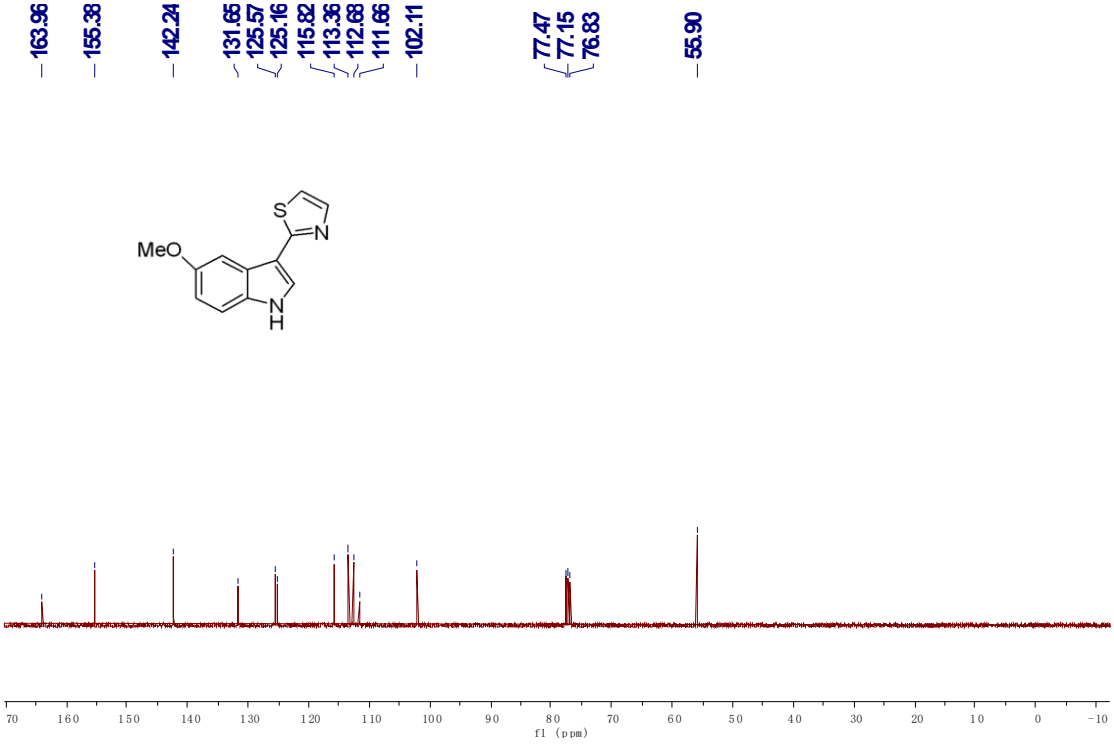
^{13}C NMR spectrum of **3d**



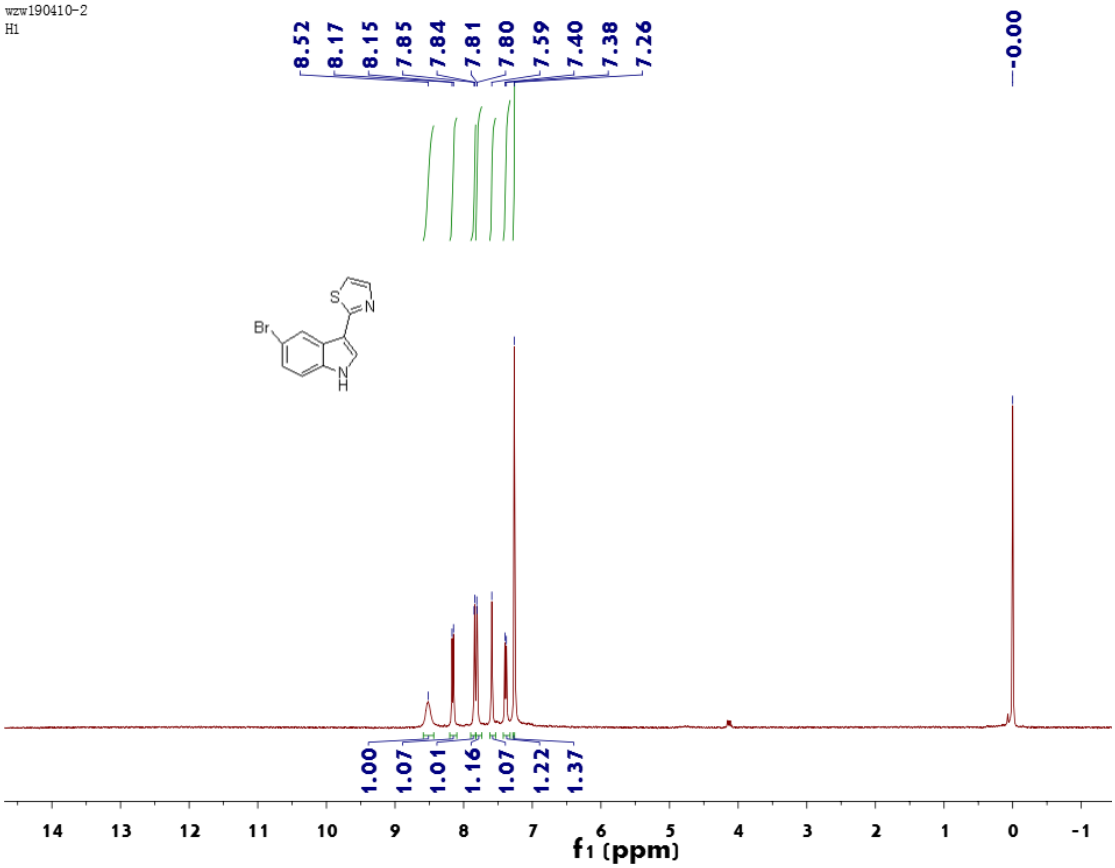
^{1}H NMR spectrum of **4a**



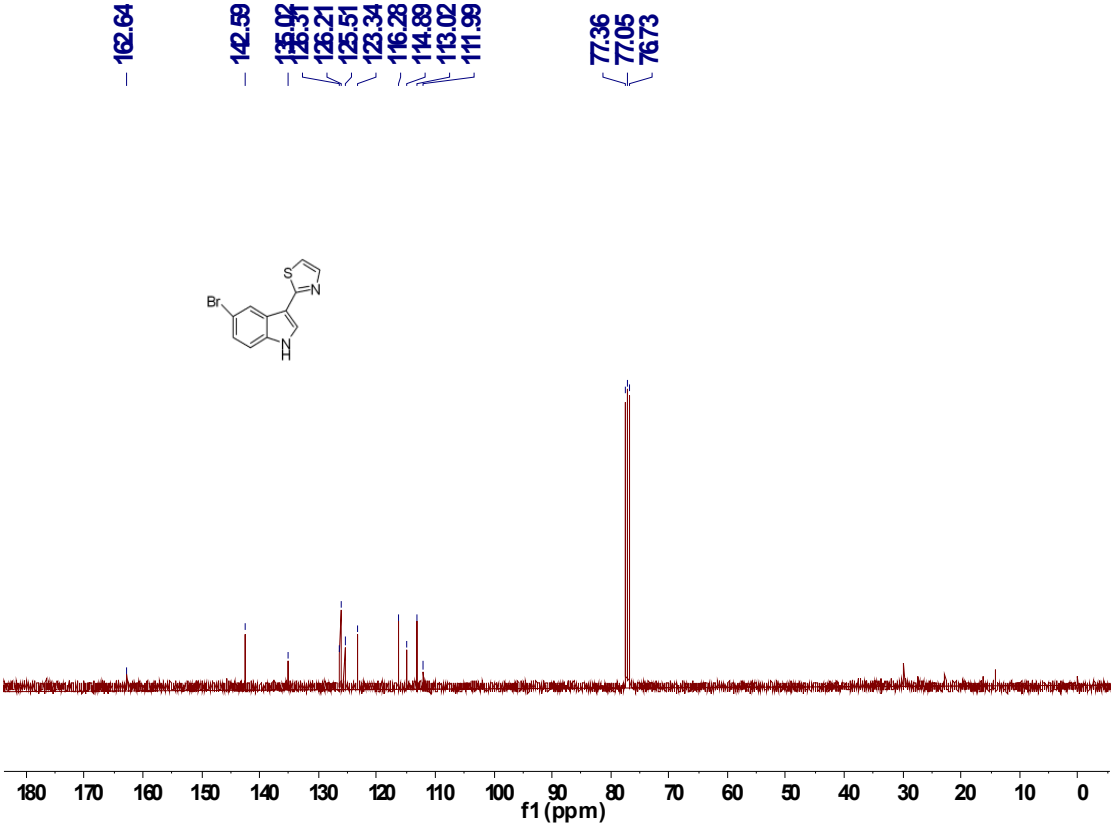
^{1}H NMR spectrum of **4b**



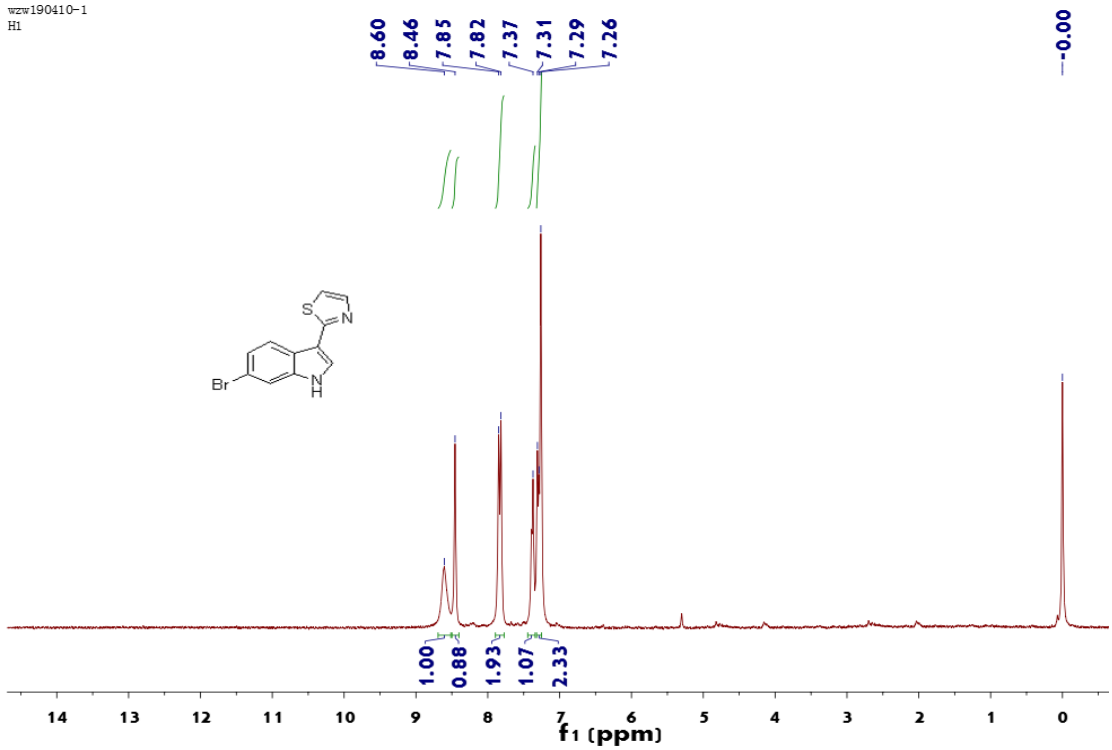
^{13}C NMR spectrum of **4b**



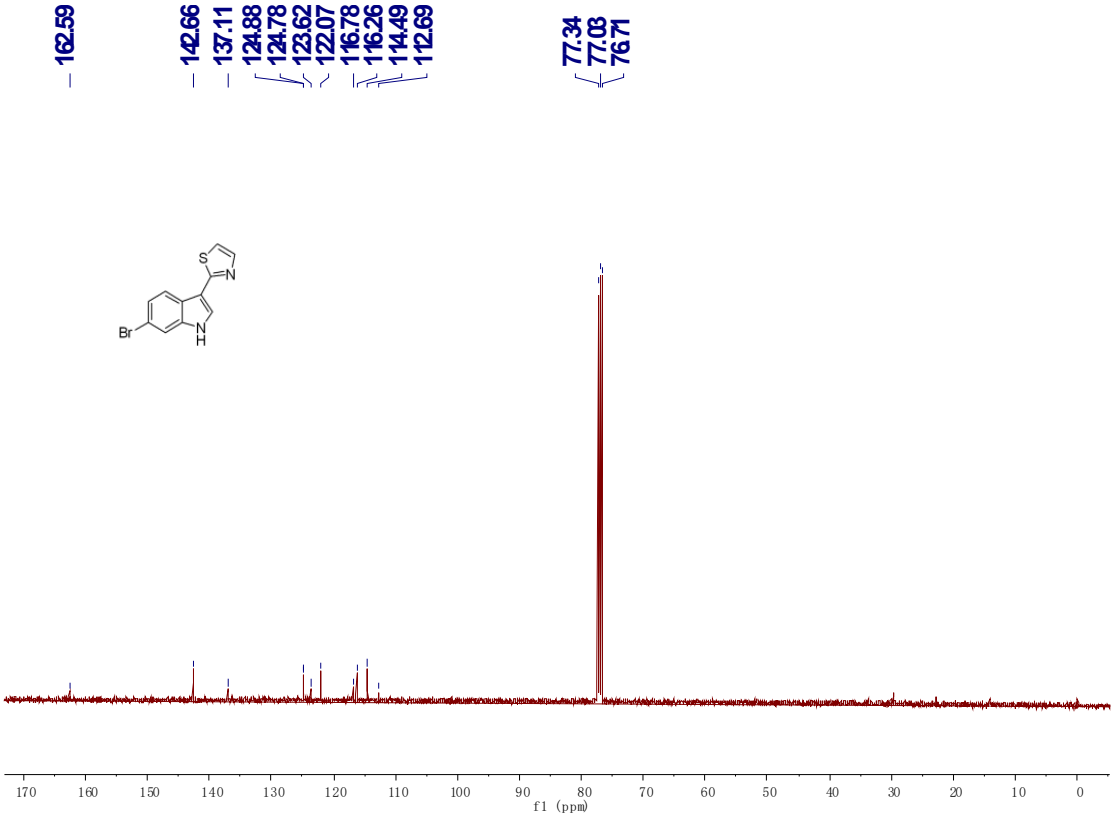
^{1}H NMR spectrum of **4c**



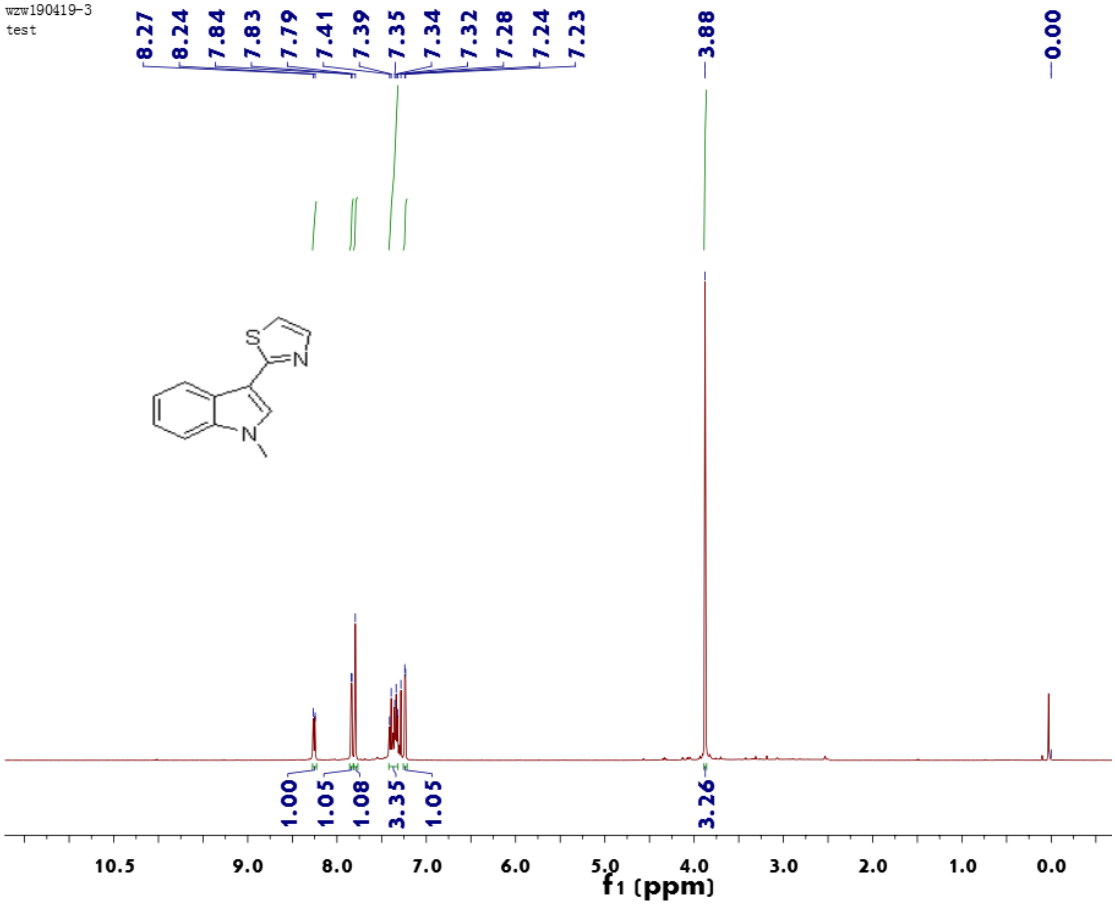
^{13}C NMR spectrum of **4c**



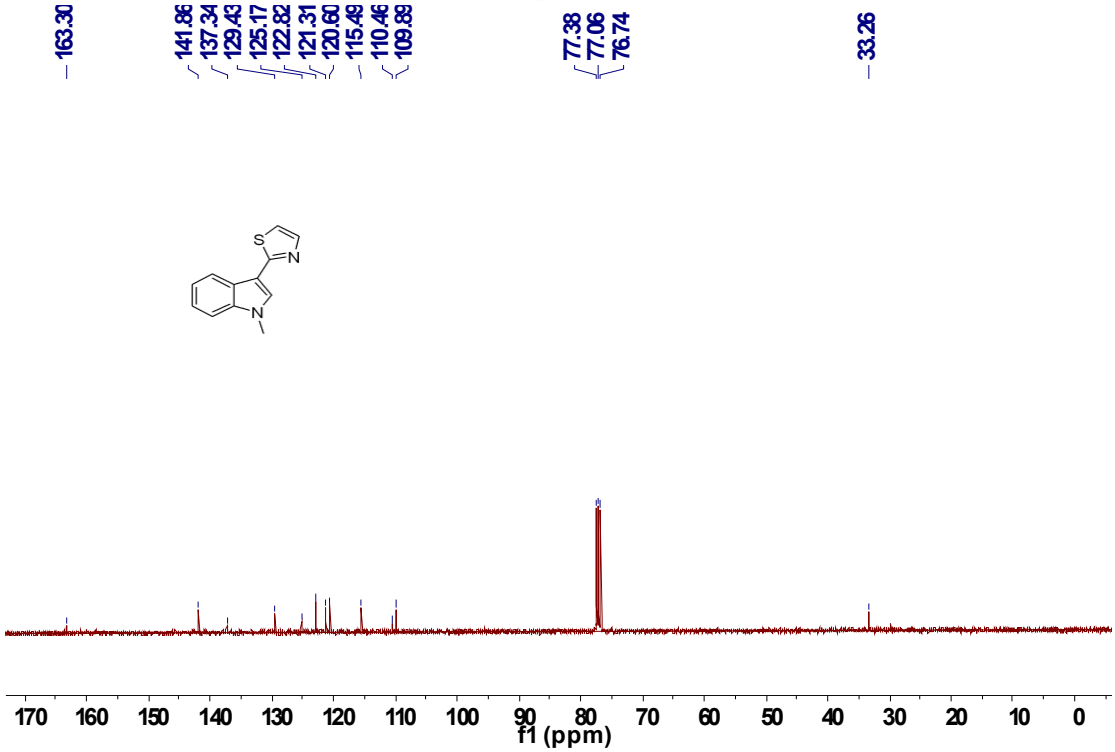
^{1}H NMR spectrum of **4d**



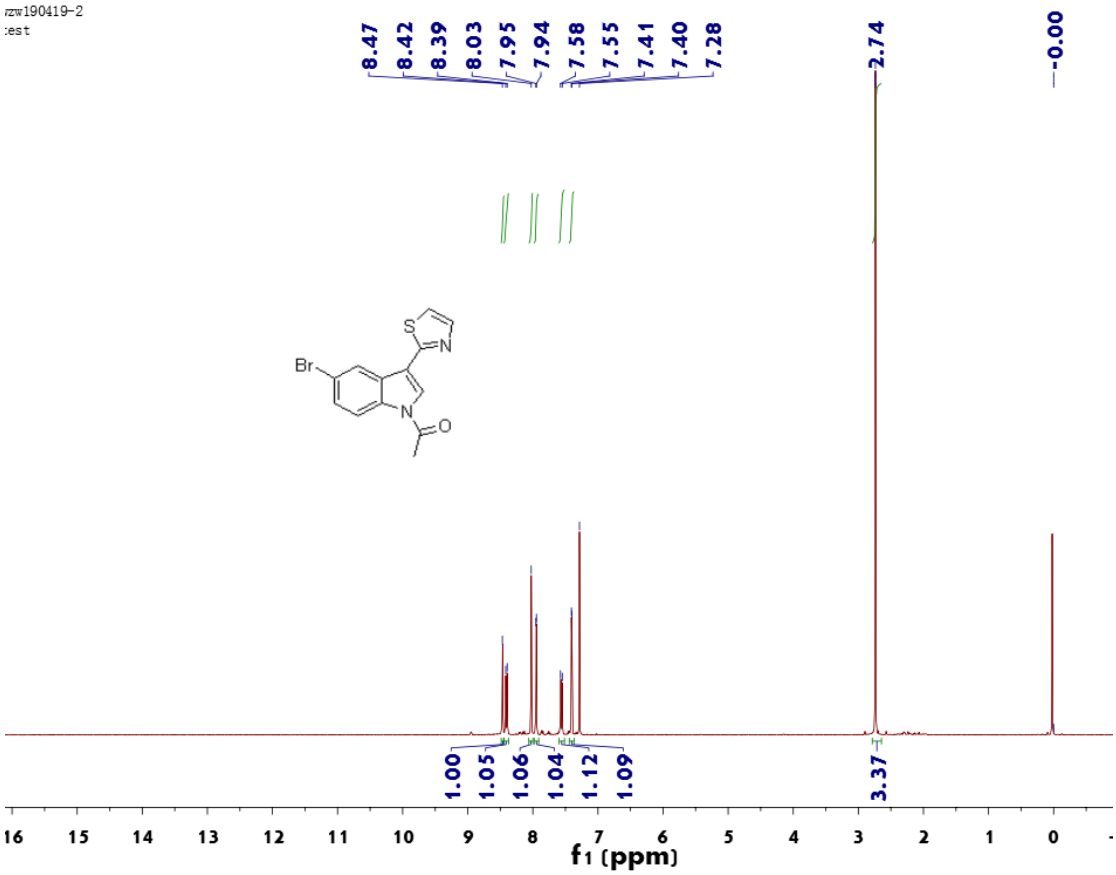
^{13}C NMR spectrum of **4d**



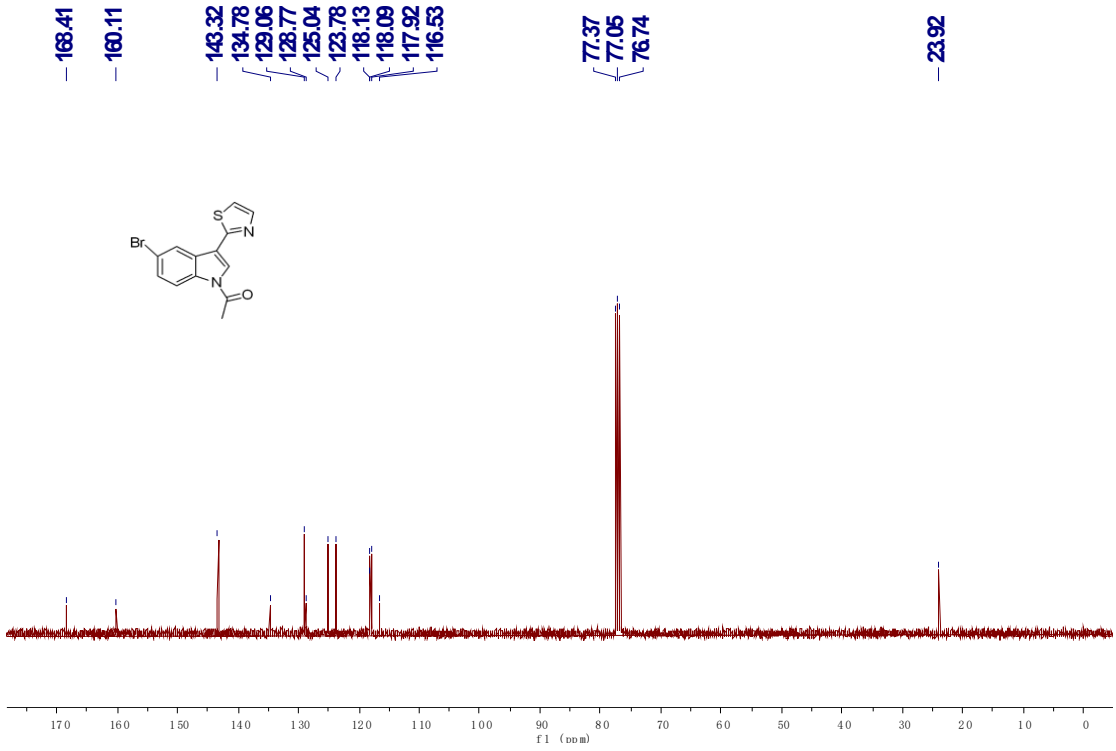
^{1}H NMR spectrum of **5a**



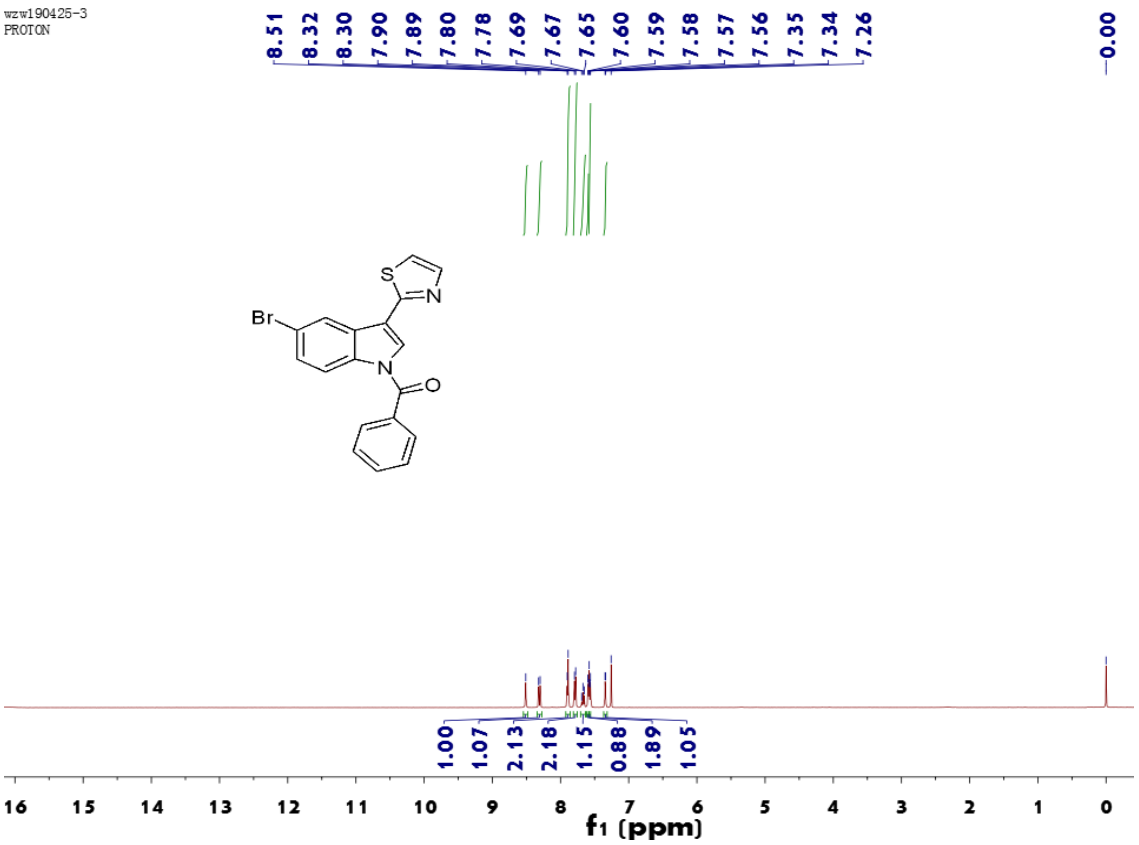
^{13}C NMR spectrum of **5a**



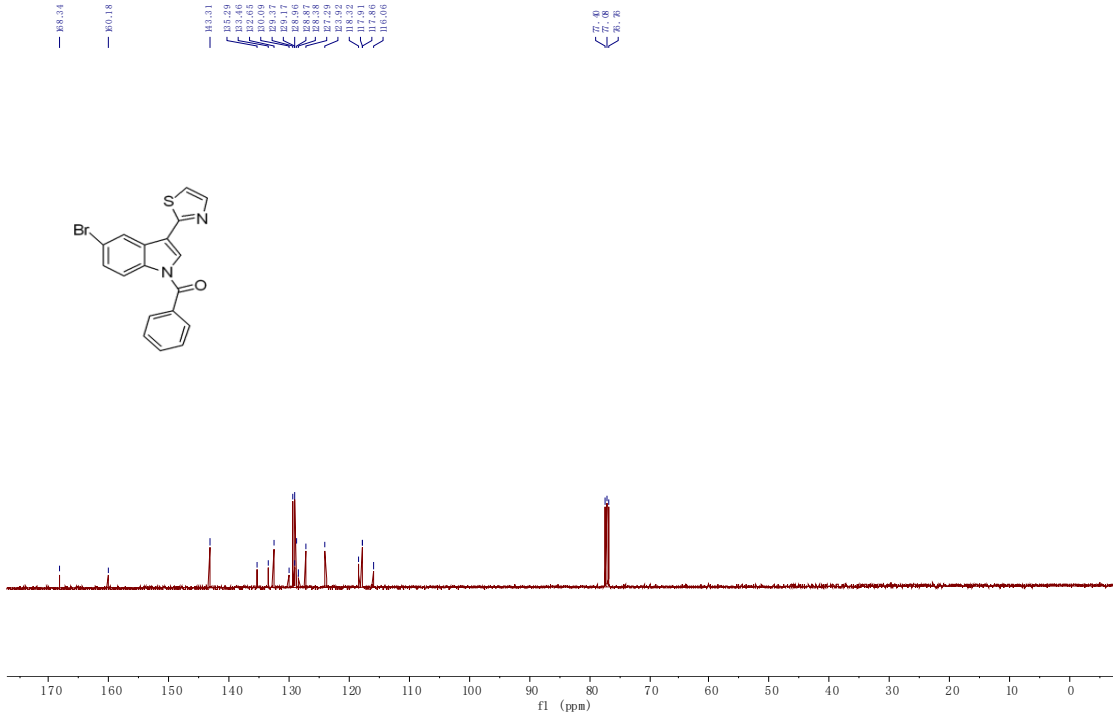
^{1}H NMR spectrum of **5b**



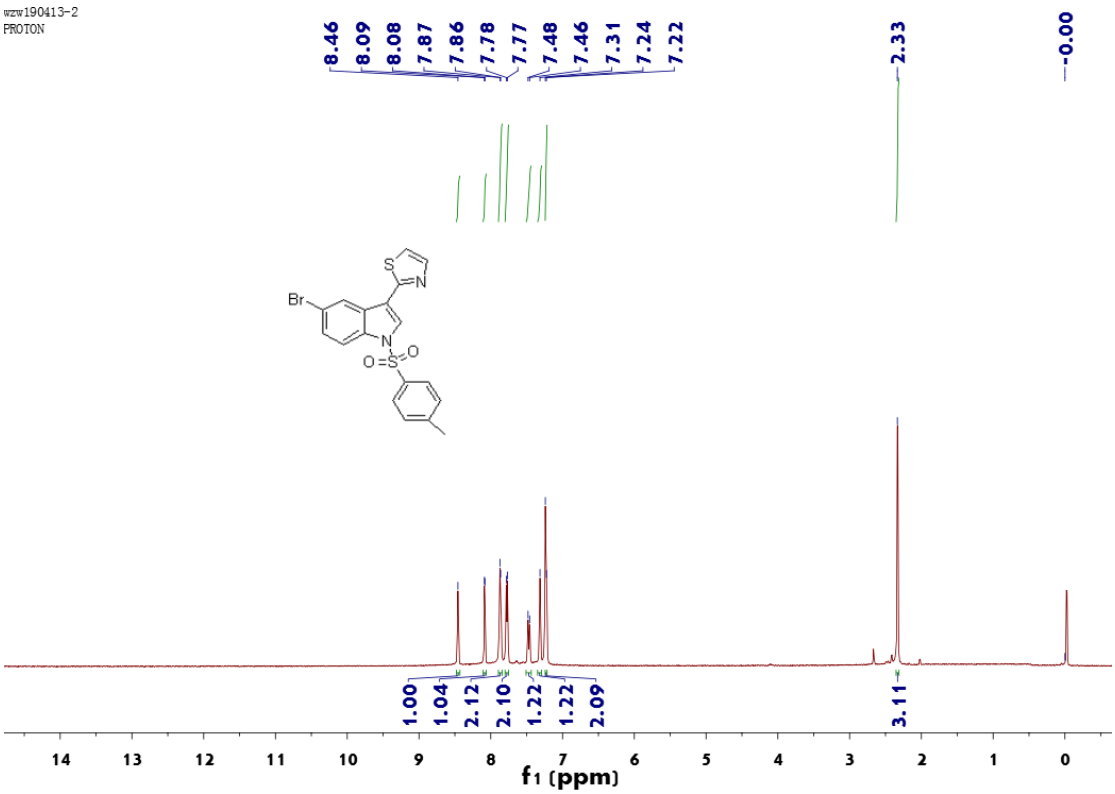
^{13}C NMR spectrum of **5b**



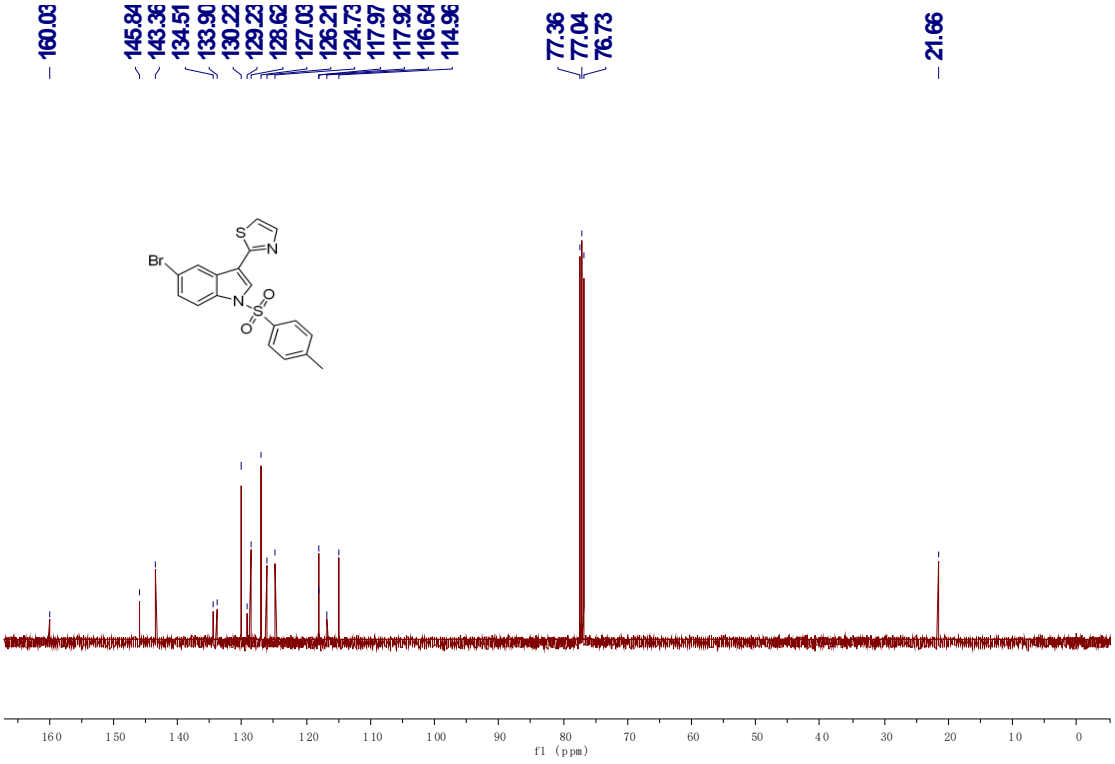
^{1}H NMR spectrum of **5c**



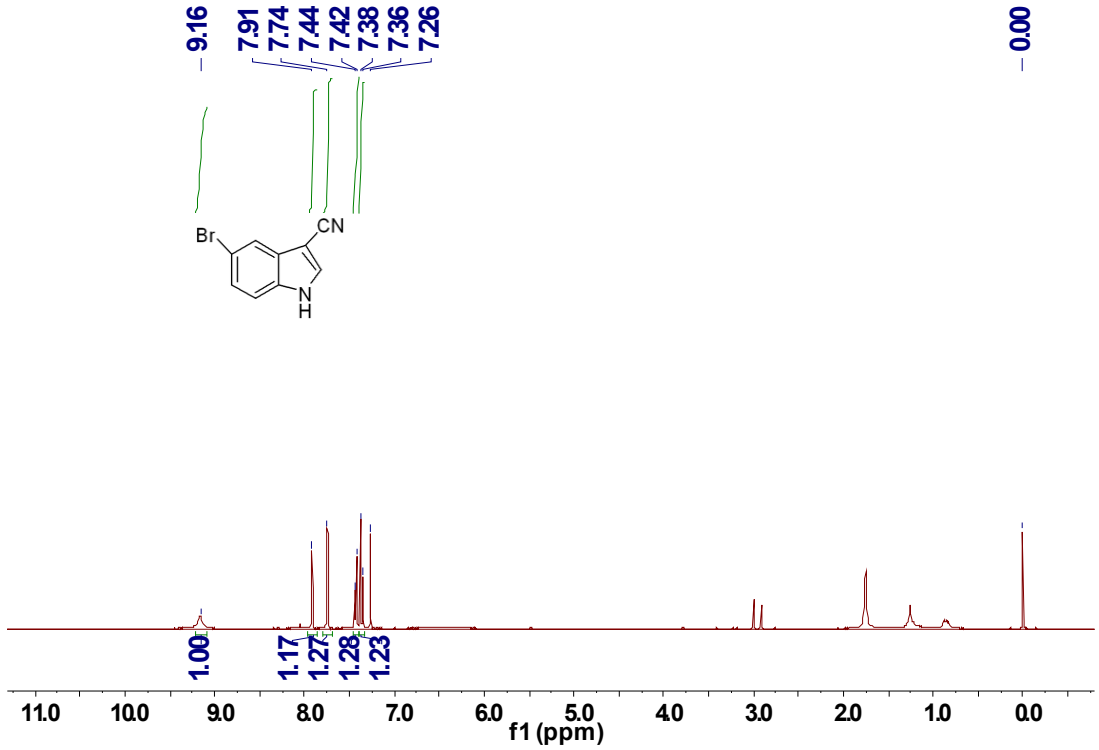
^{13}C NMR spectrum of **5c**



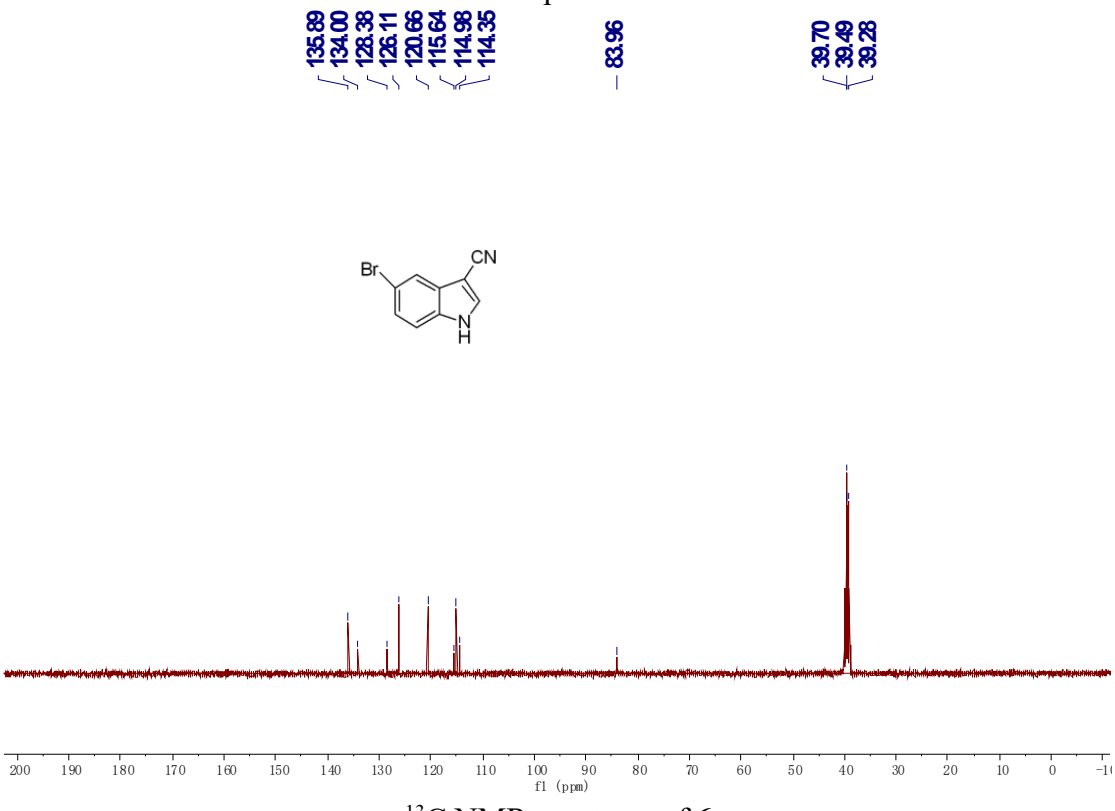
^{1}H NMR spectrum of **5d**



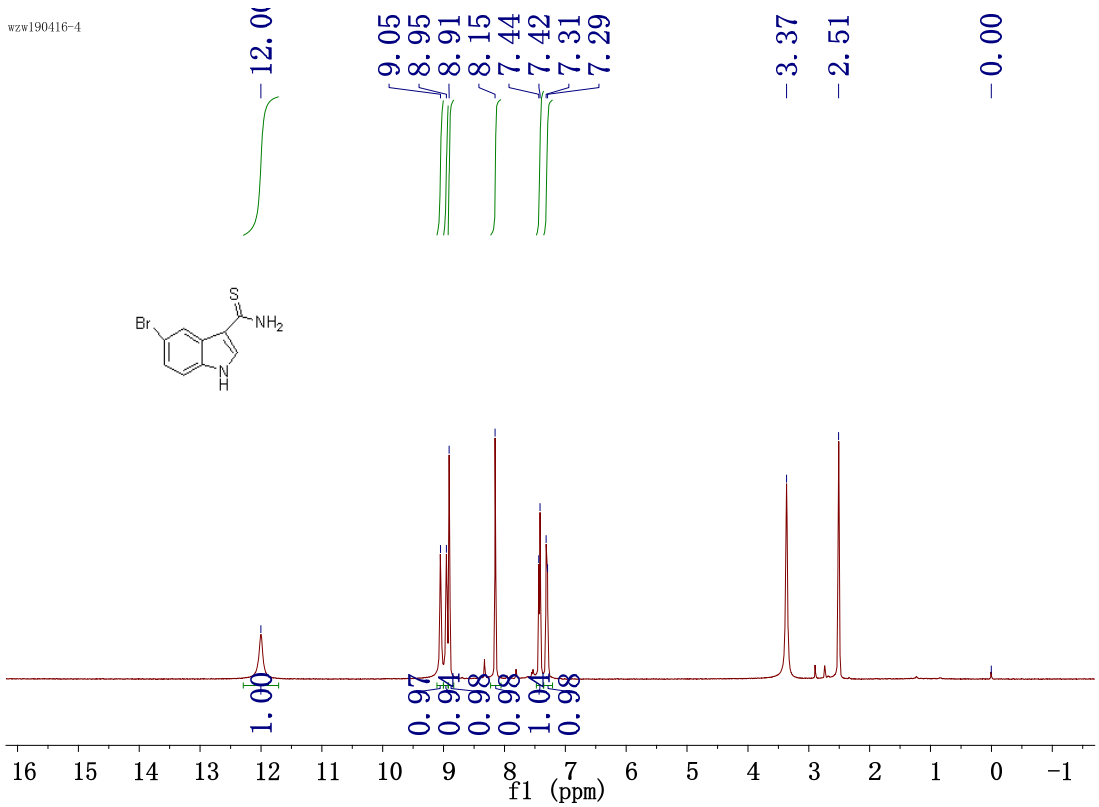
^{13}C NMR spectrum of **5d**



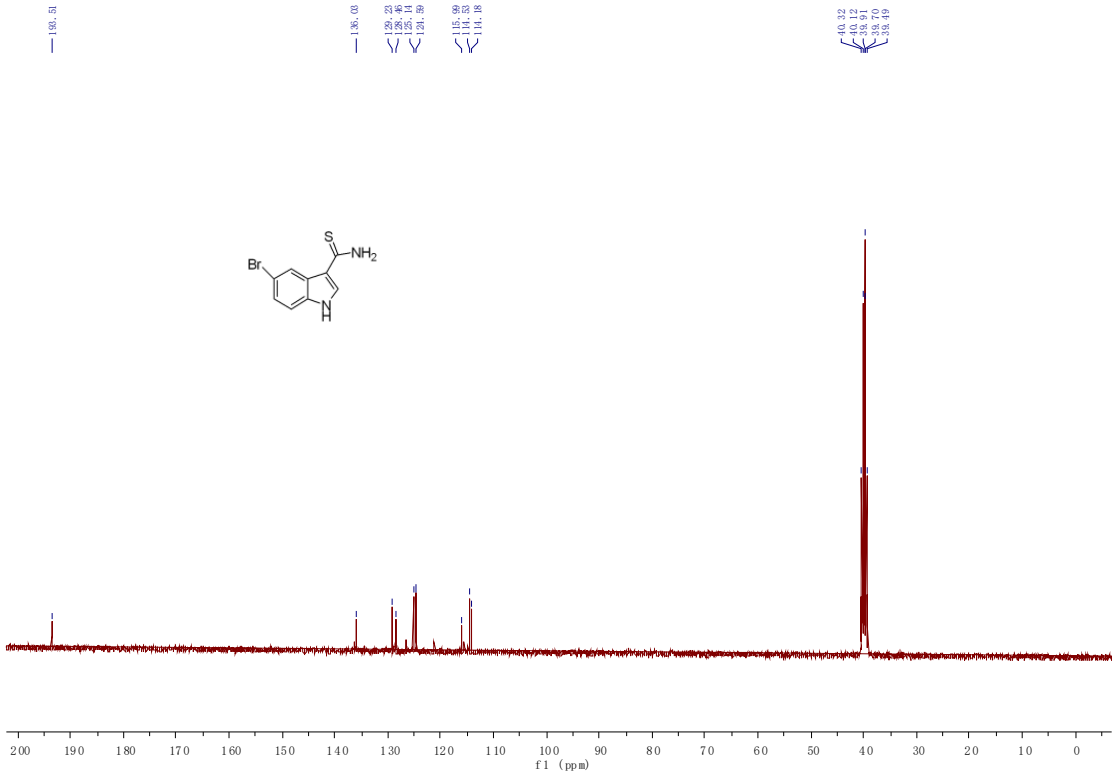
^{1}H NMR spectrum of **6**



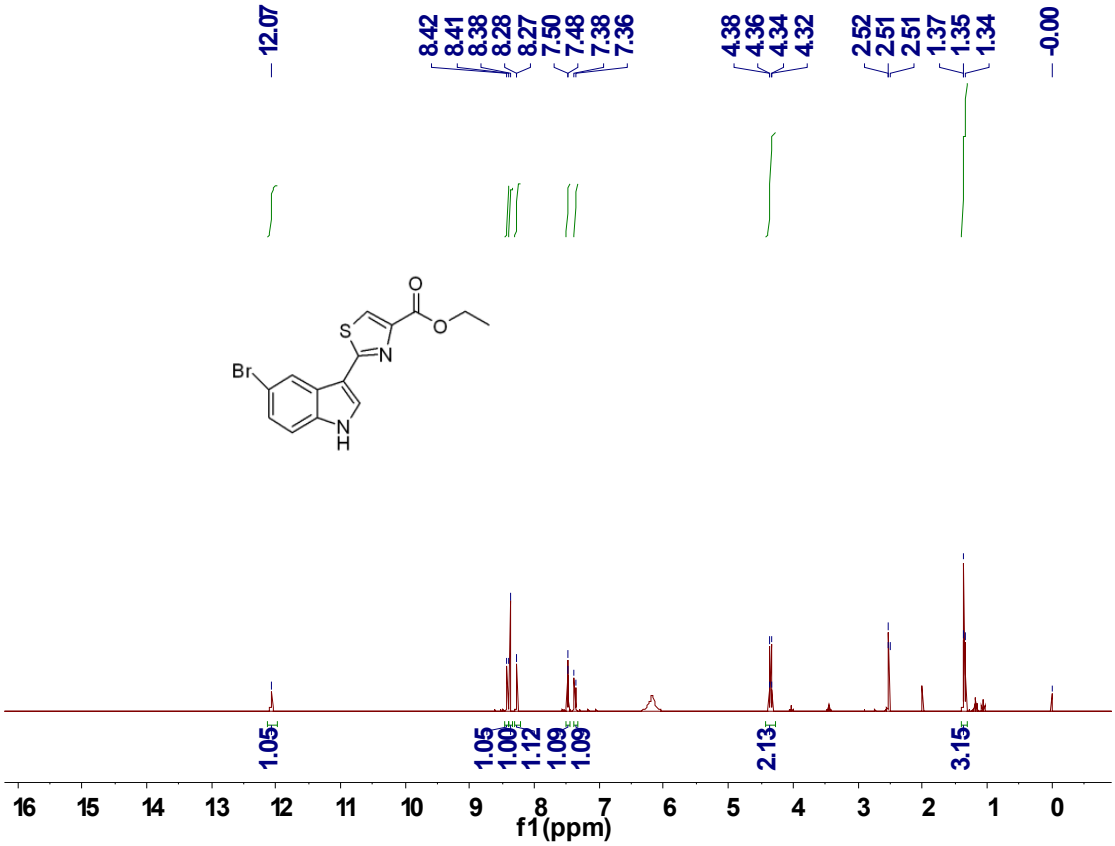
^{13}C NMR spectrum of **6**



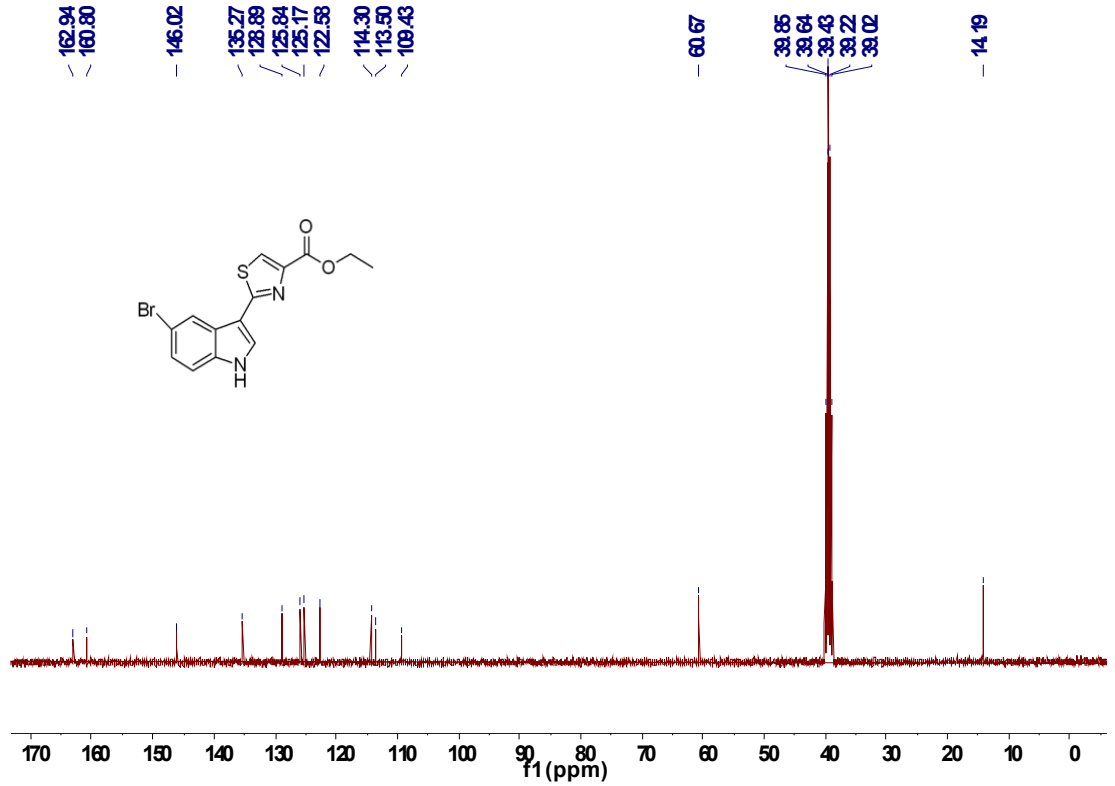
^{1}H NMR spectrum of **7**



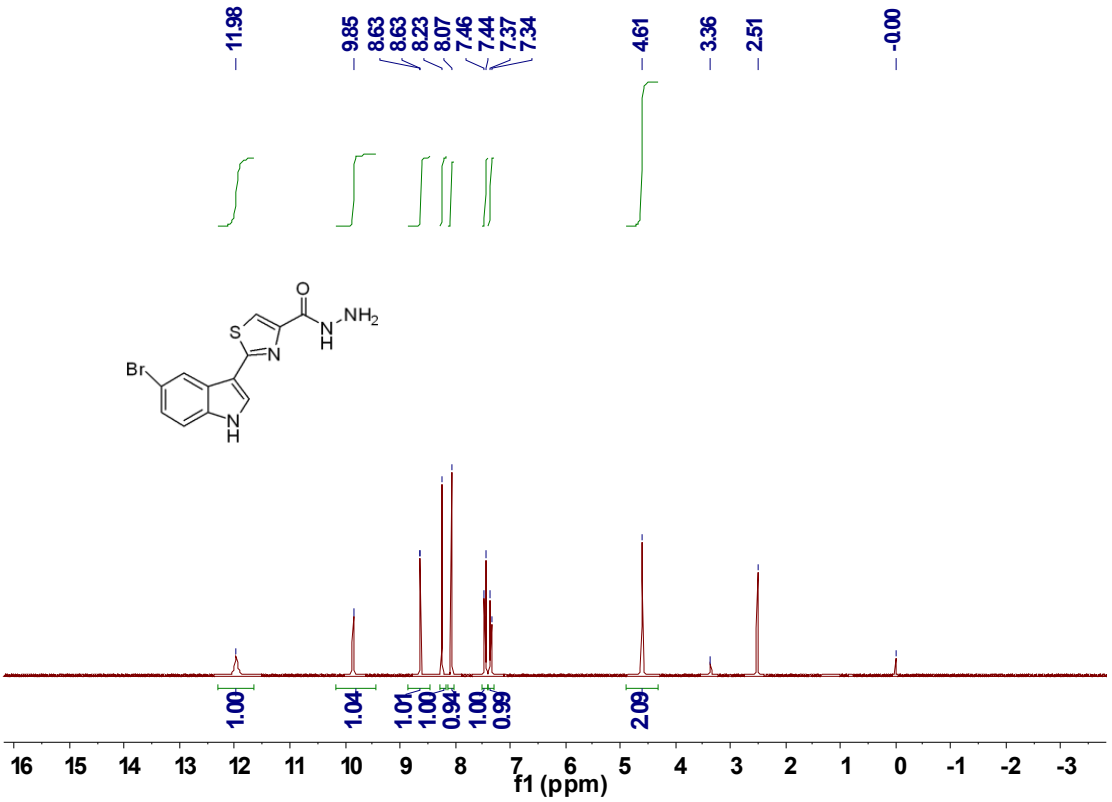
^{13}C NMR spectrum of **7**



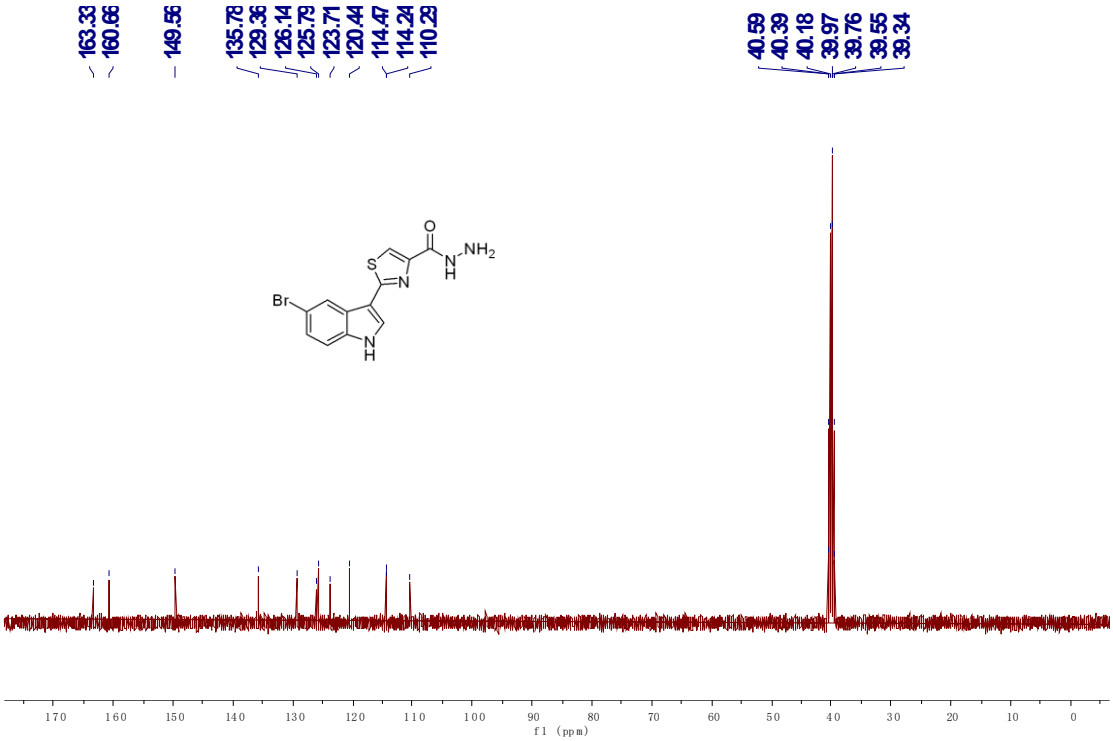
^{1}H NMR spectrum of **8**



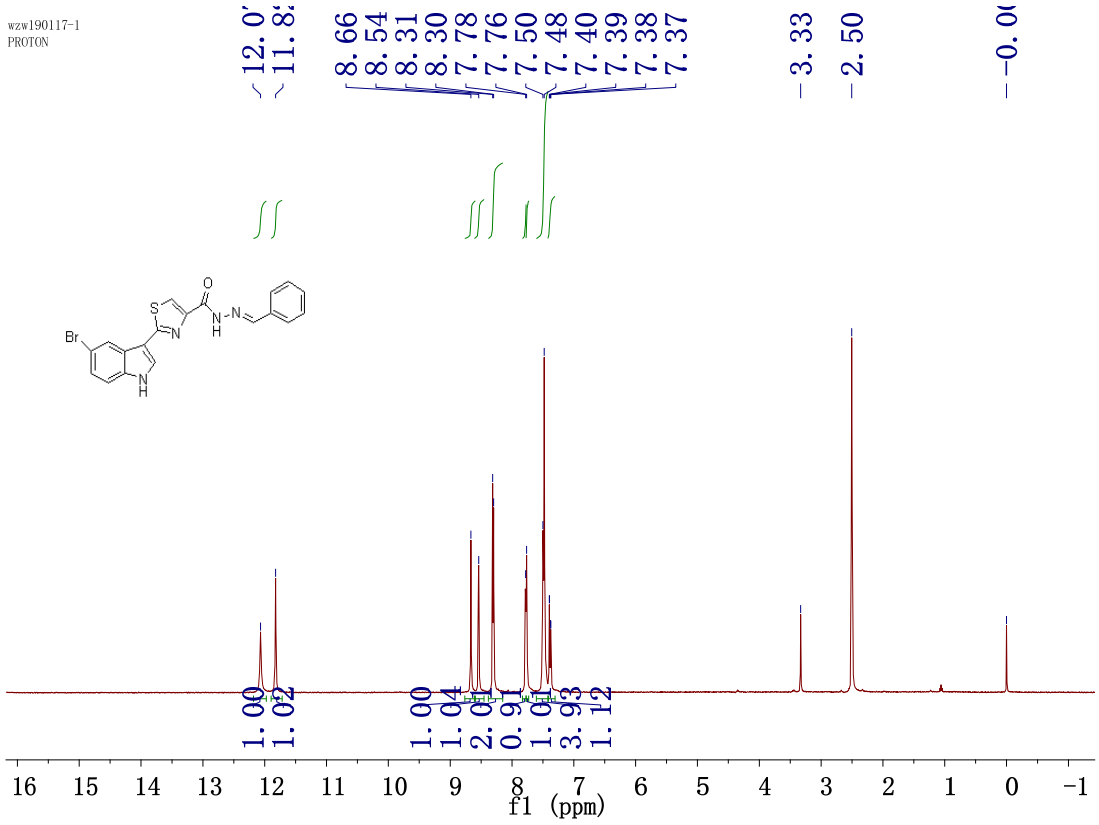
^{13}C NMR spectrum of **8**



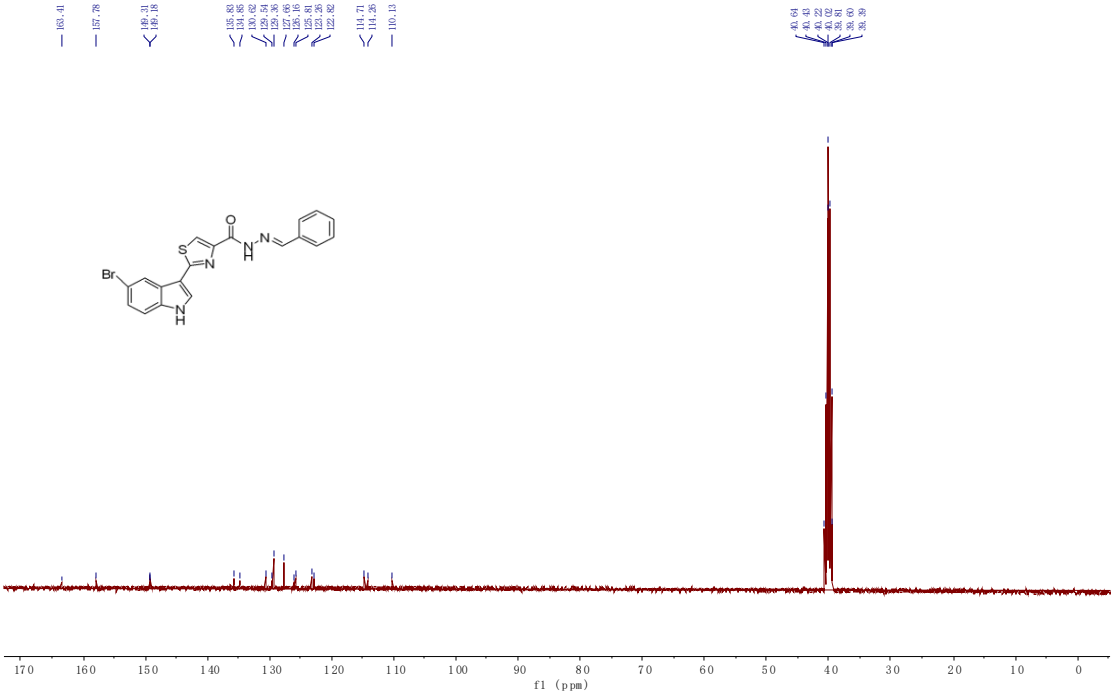
^{1}H NMR spectrum of **9**



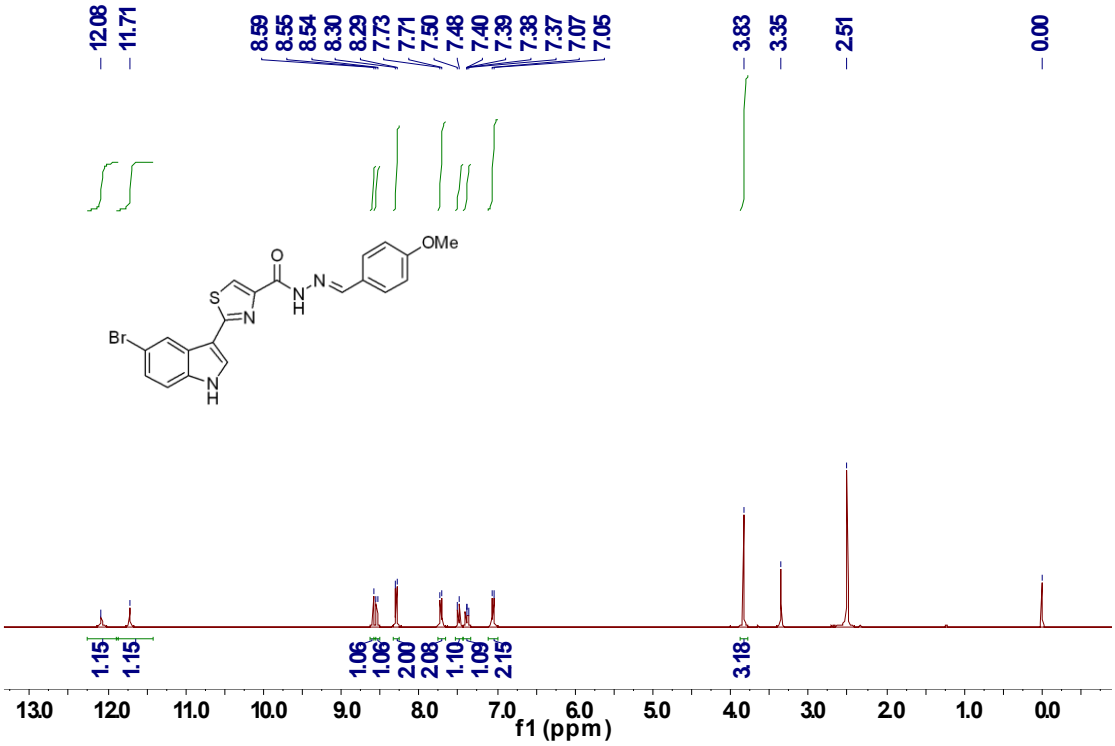
^{13}C NMR spectrum of **9**



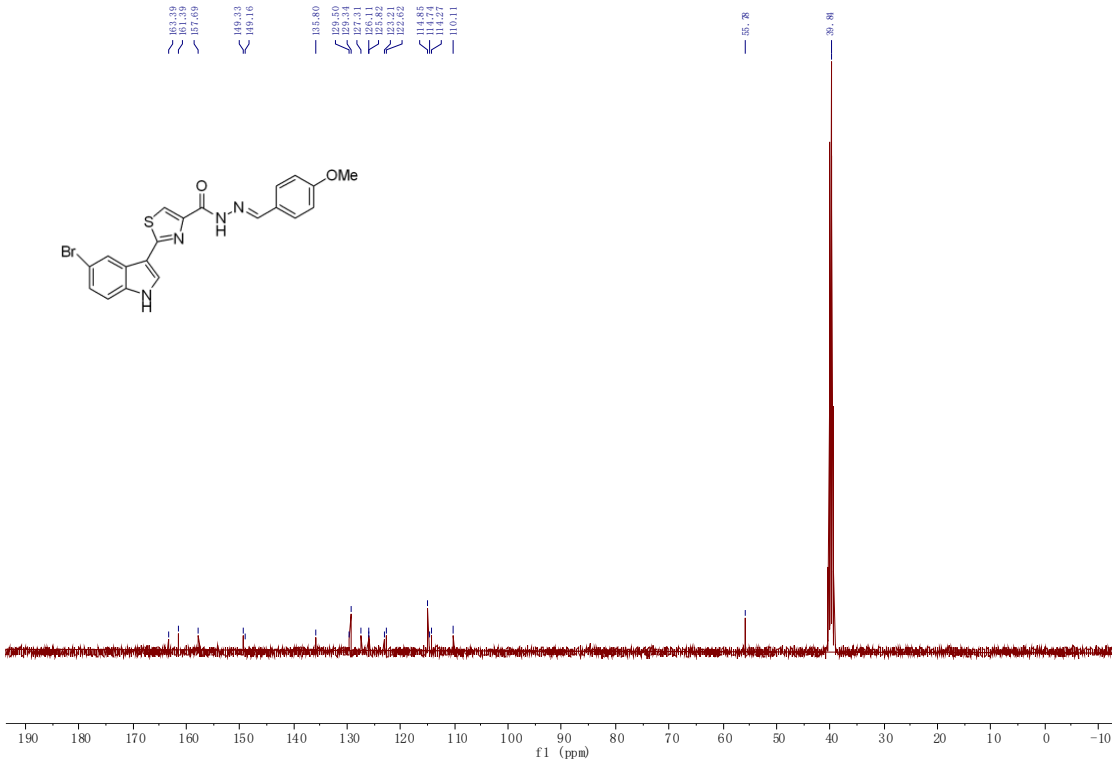
^{1}H NMR spectrum of **10a**



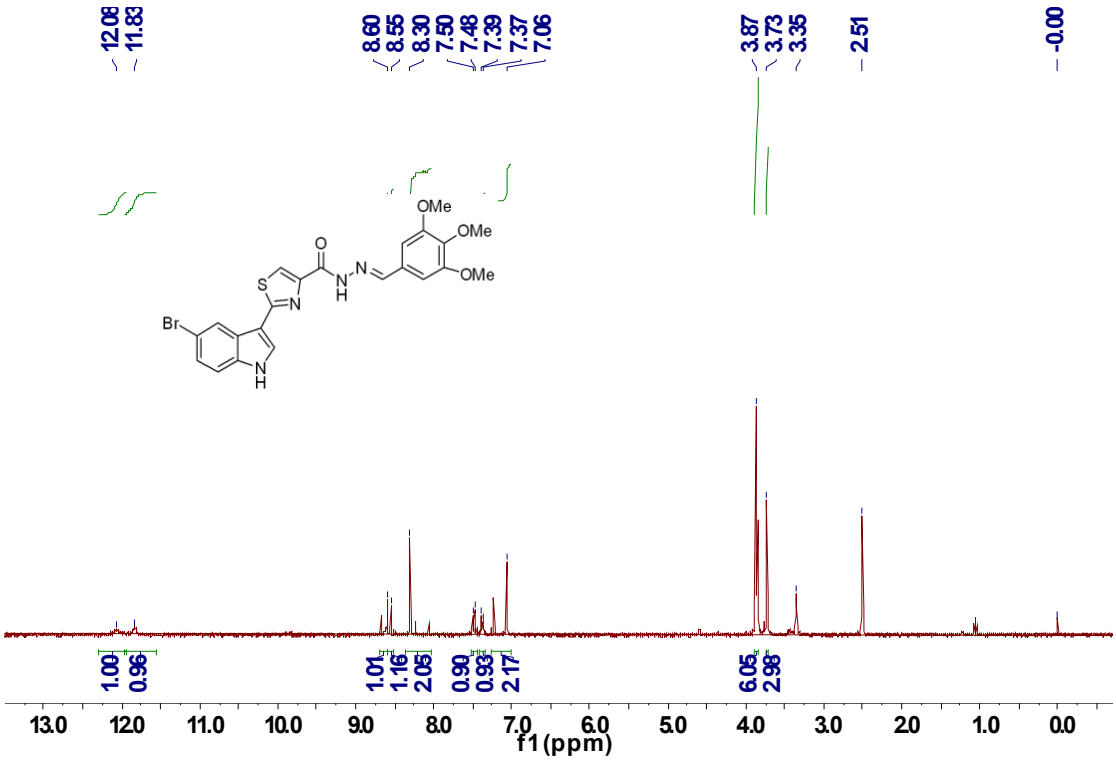
^{13}C NMR spectrum of **10a**



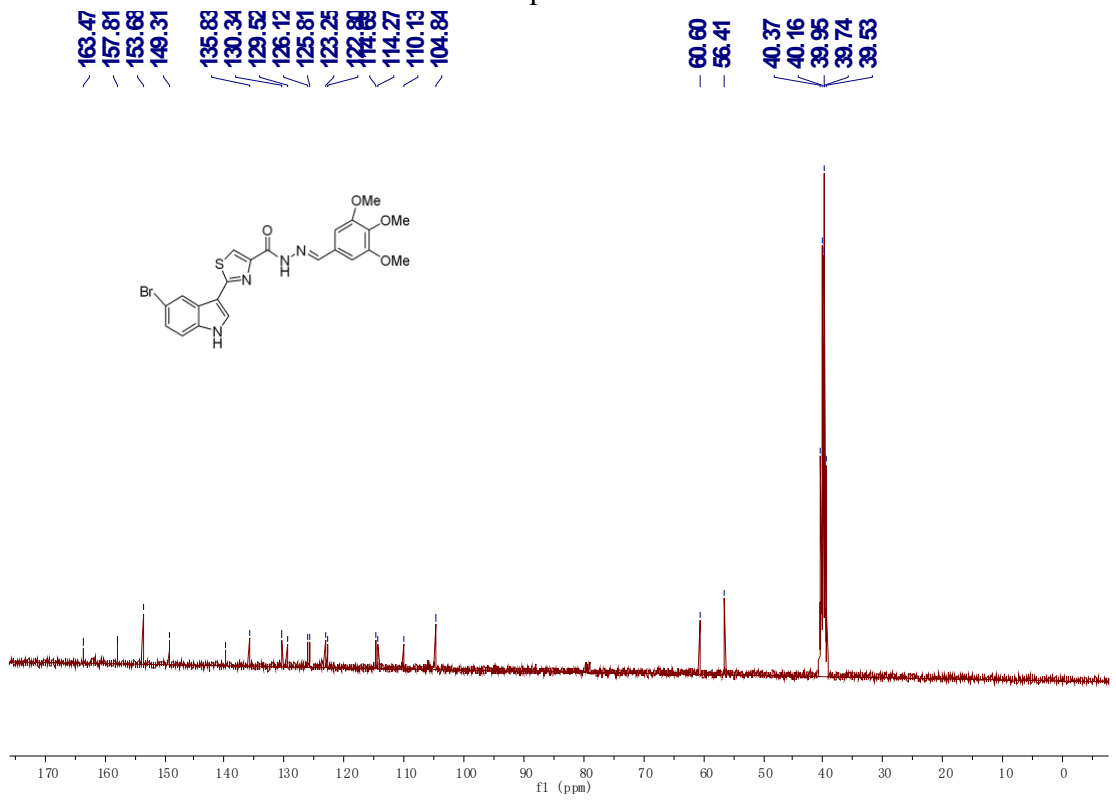
^{1}H NMR spectrum of **10b**



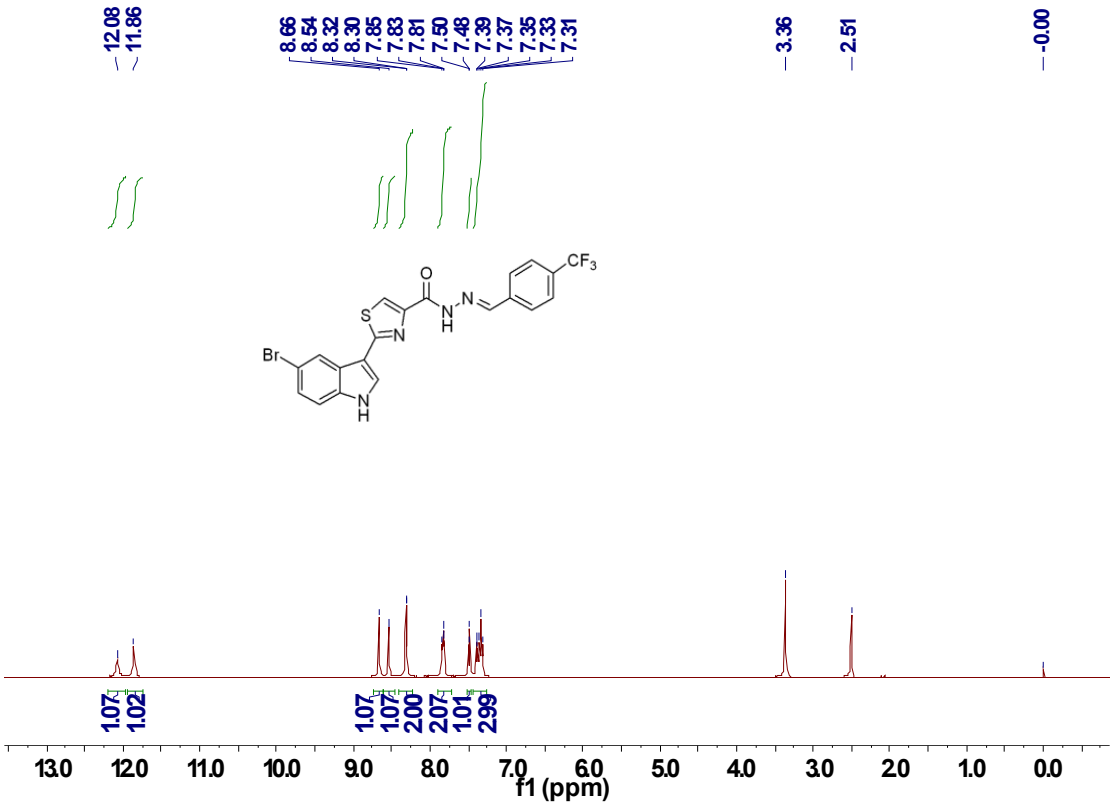
^{13}C NMR spectrum of **10b**



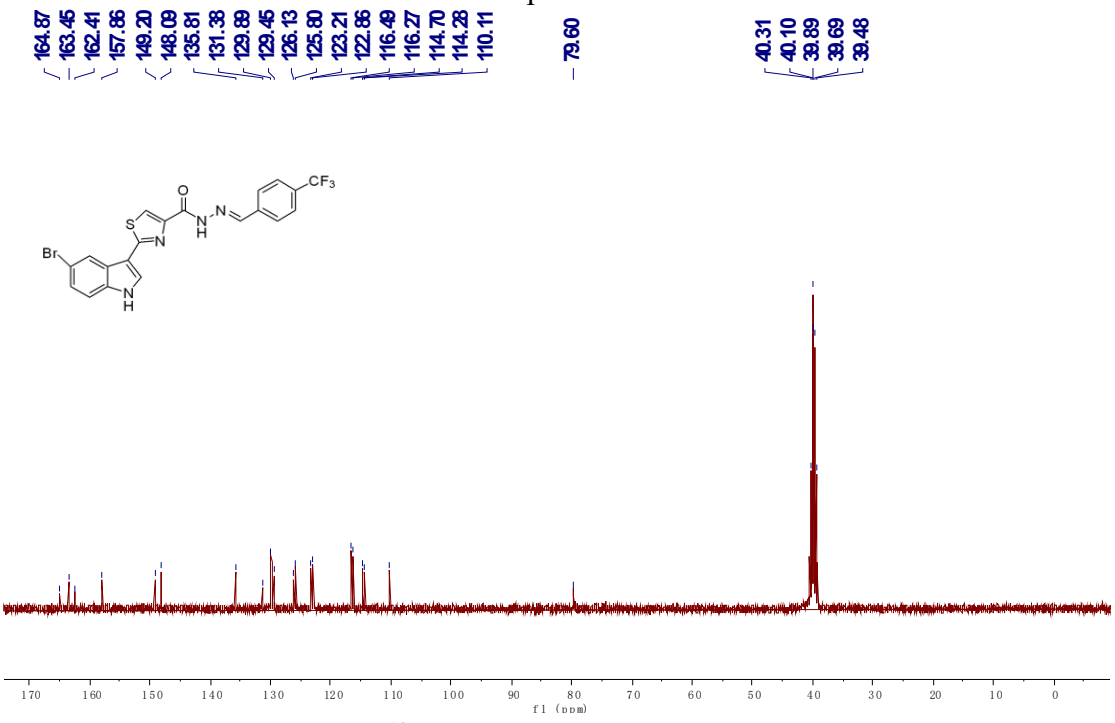
^{1}H NMR spectrum of **10c**



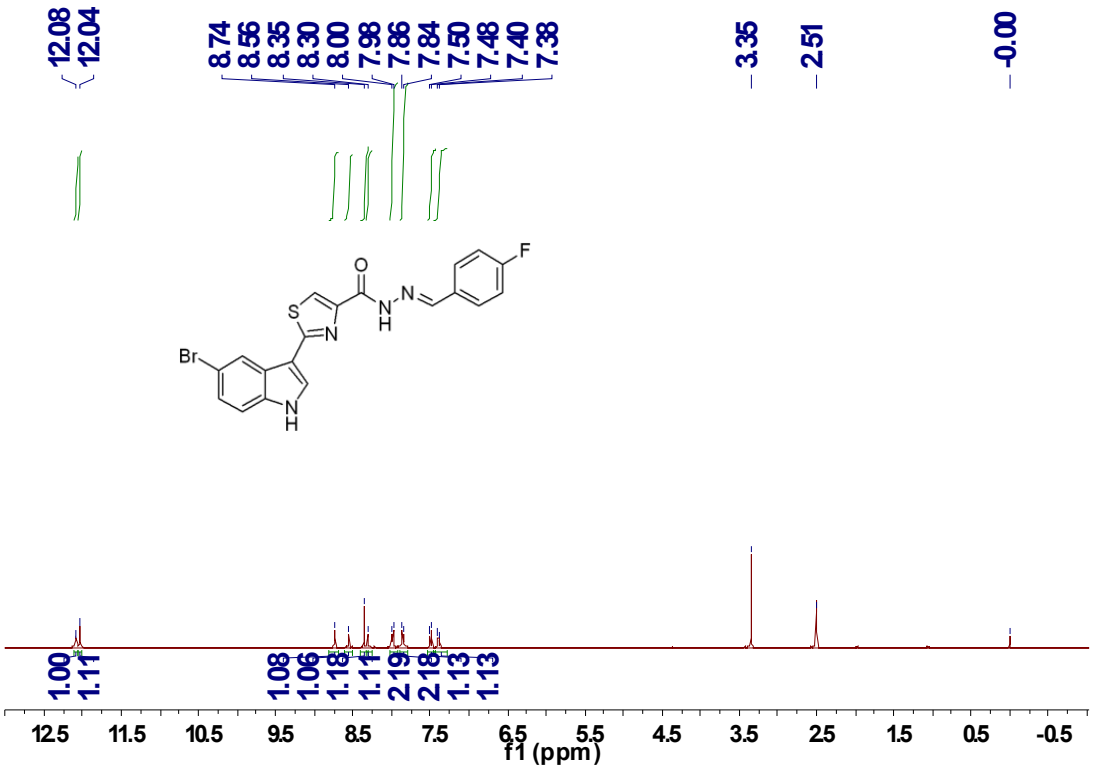
^{13}C NMR spectrum of **10c**



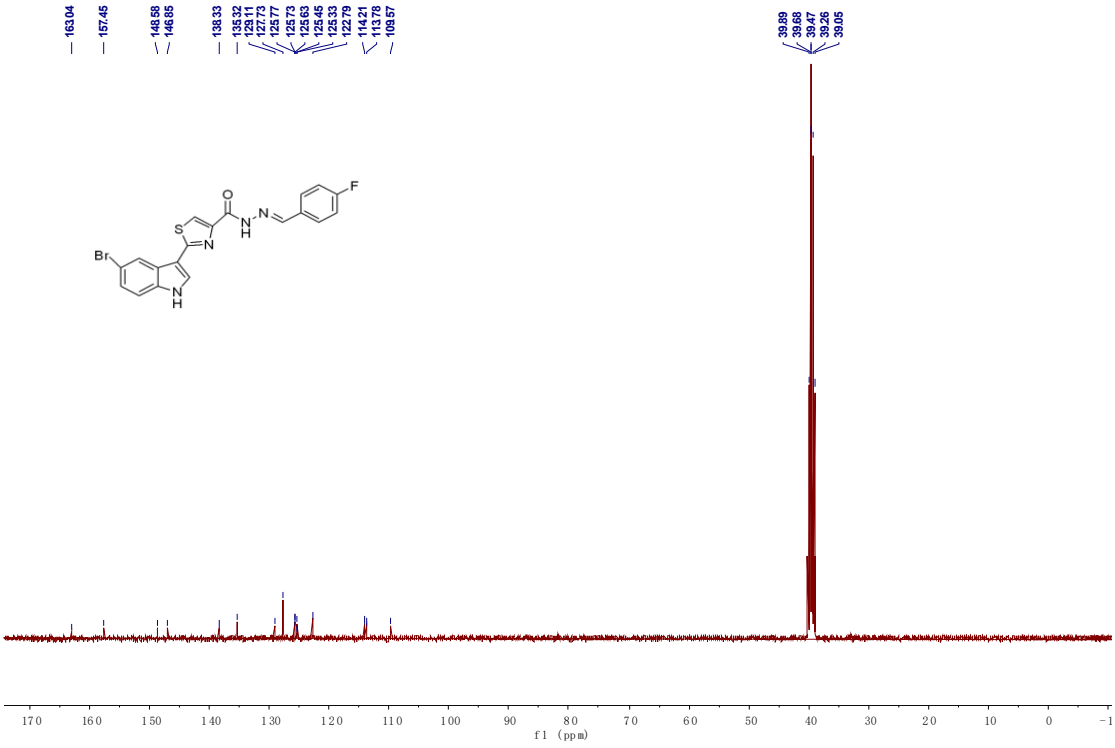
^{1}H NMR spectrum of **10d**



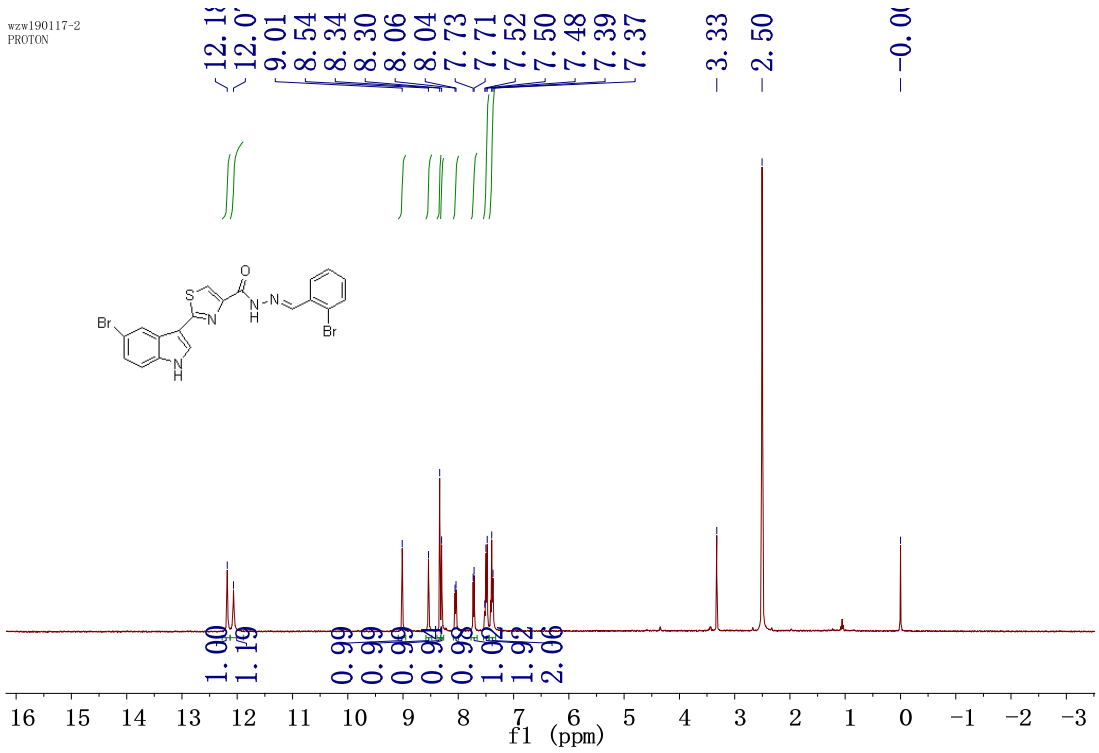
^{13}C NMR spectrum of **10d**



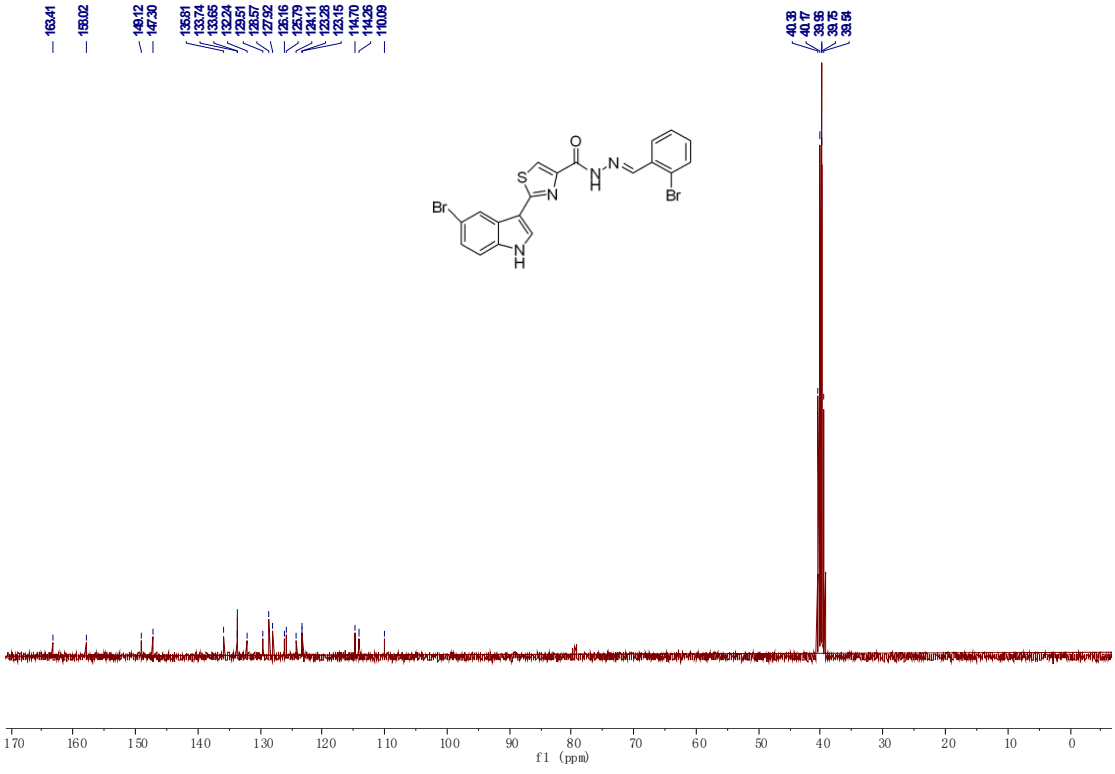
^{1}H NMR spectrum of **10e**



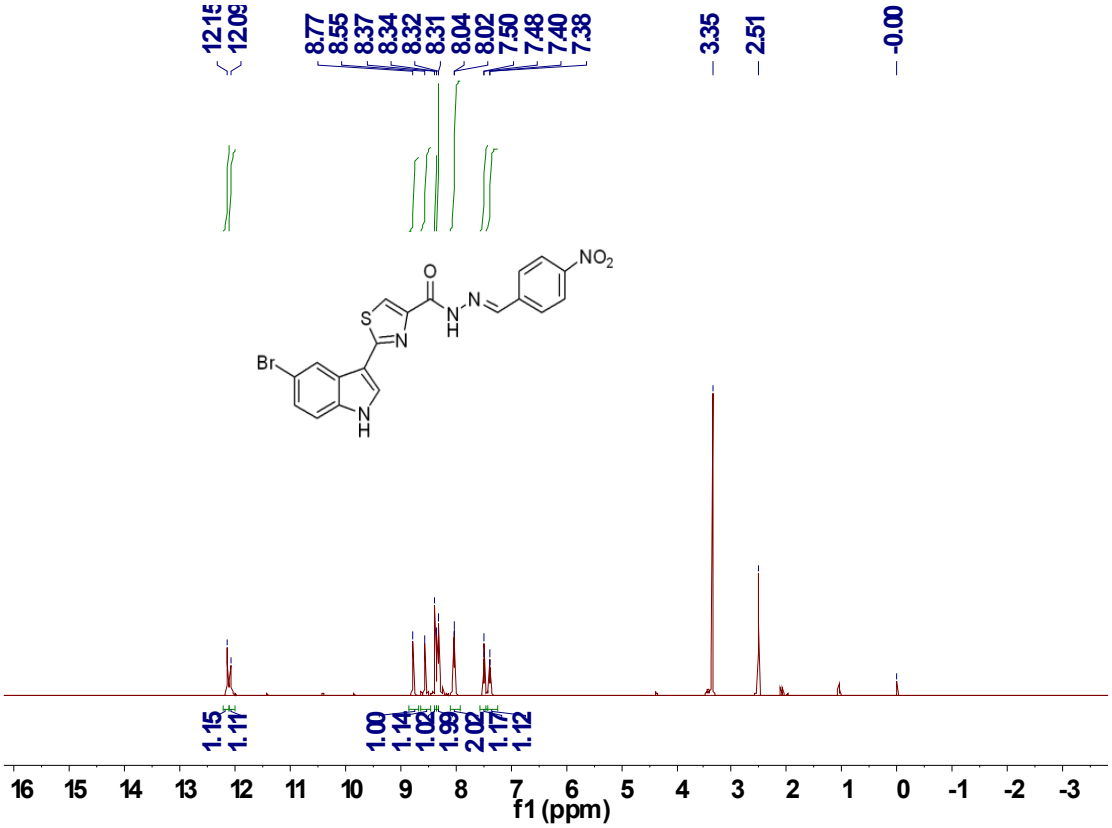
^{13}C NMR spectrum of **10e**



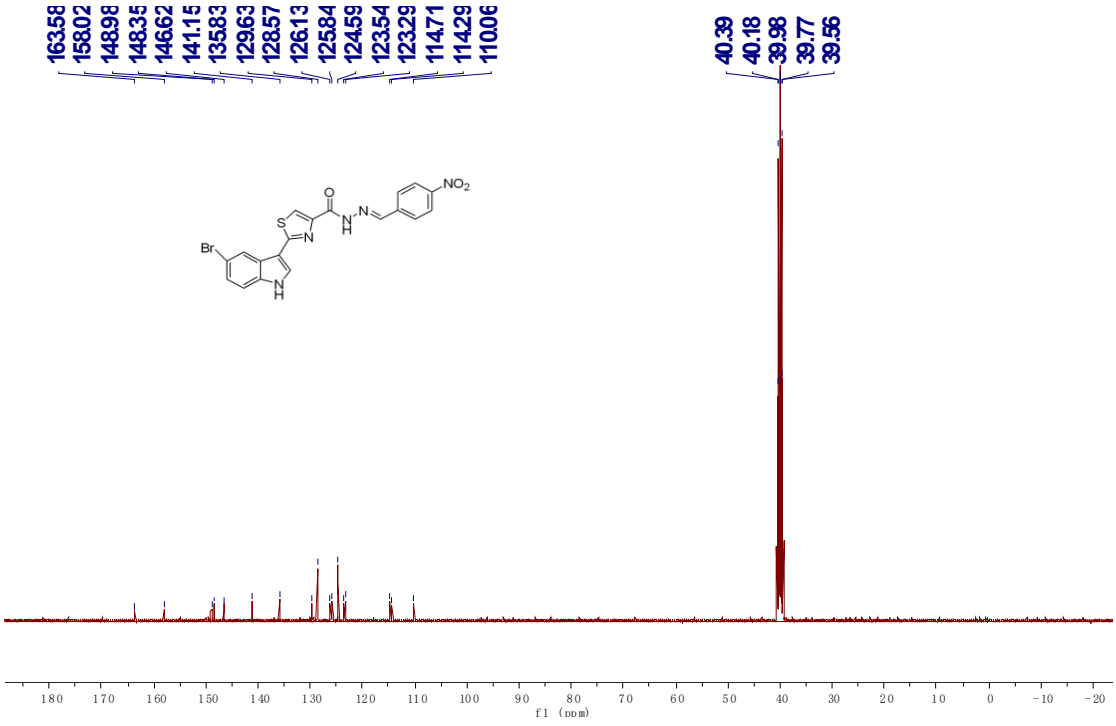
^{1}H NMR spectrum of **10f**



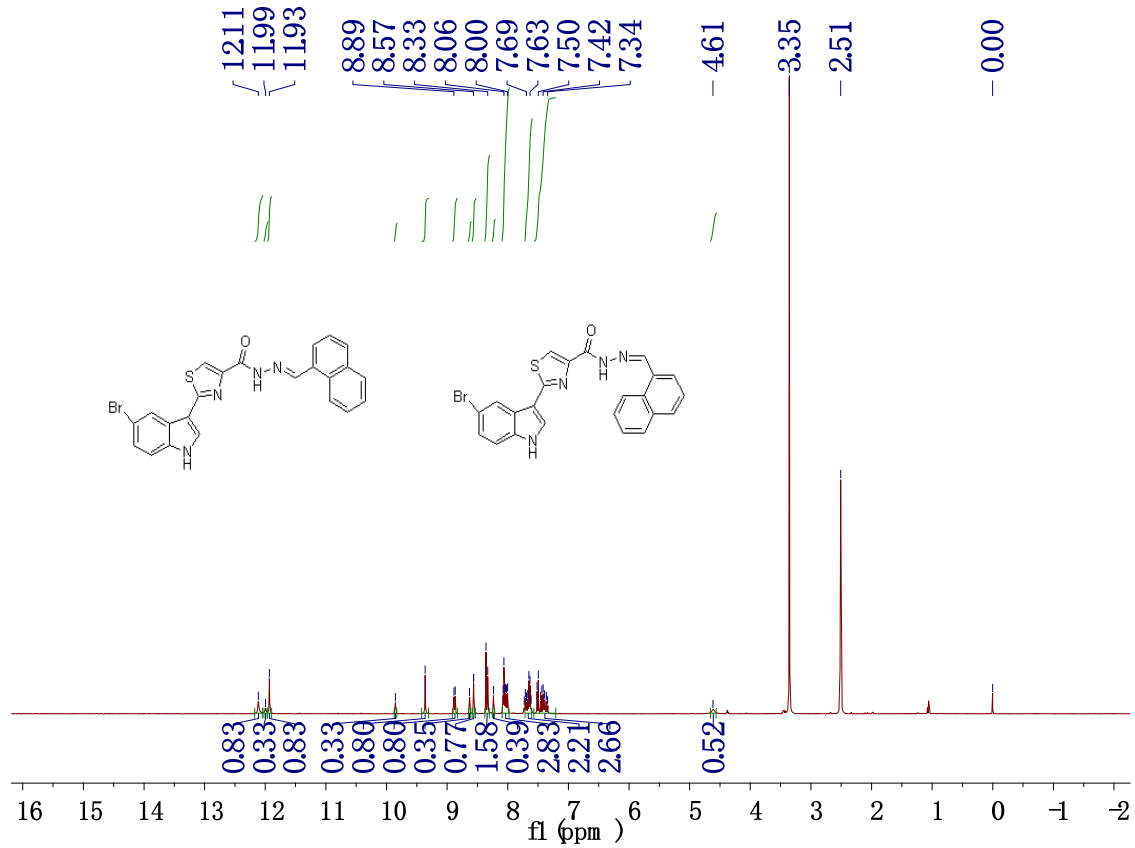
^{13}C NMR spectrum of **10f**



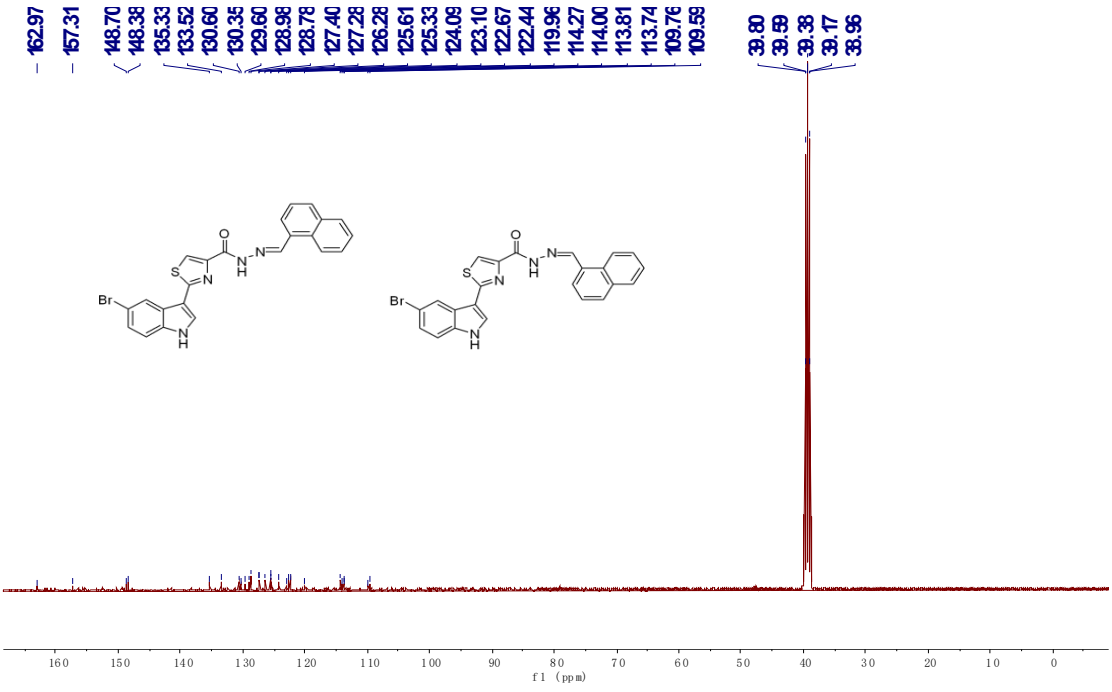
^{1}H NMR spectrum of **10g**



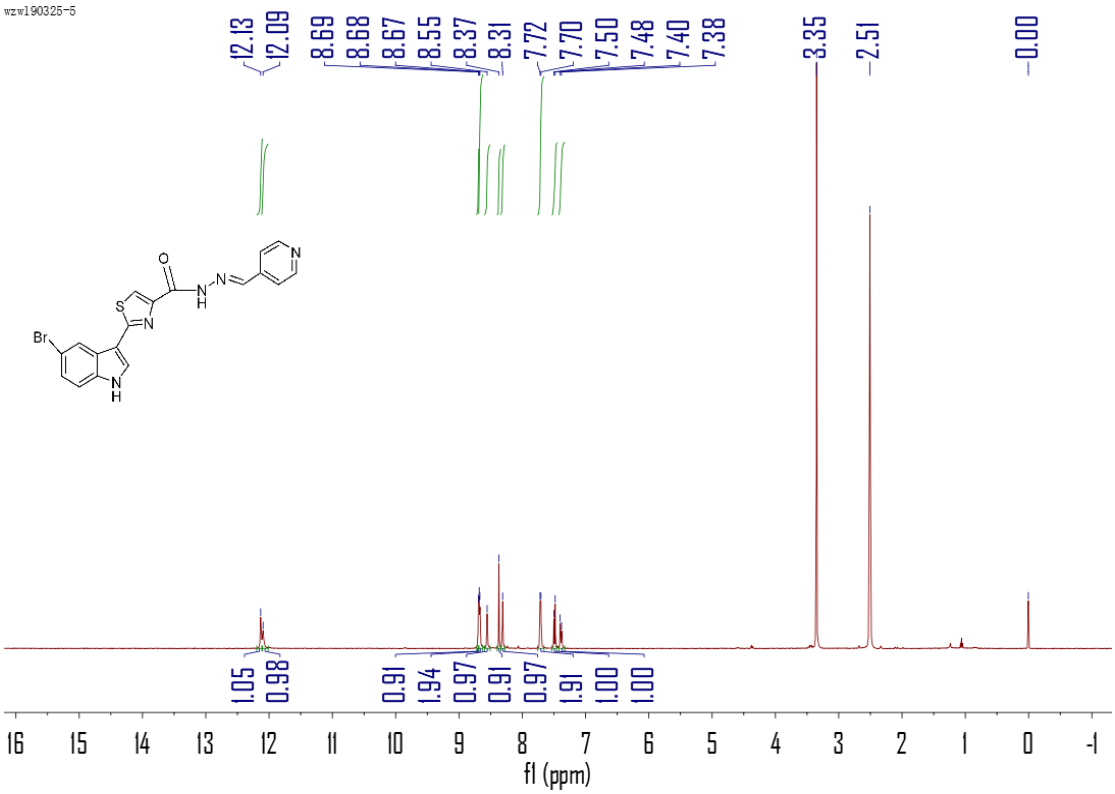
^{13}C NMR spectrum of **10g**



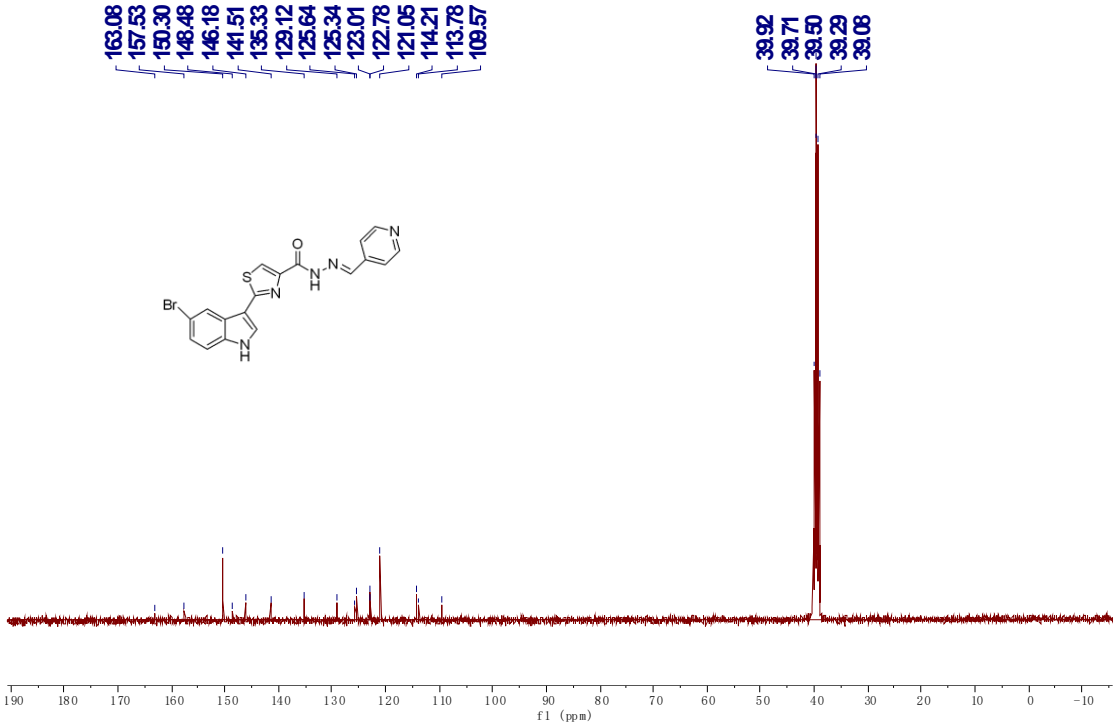
^{1}H NMR spectrum of **10h**



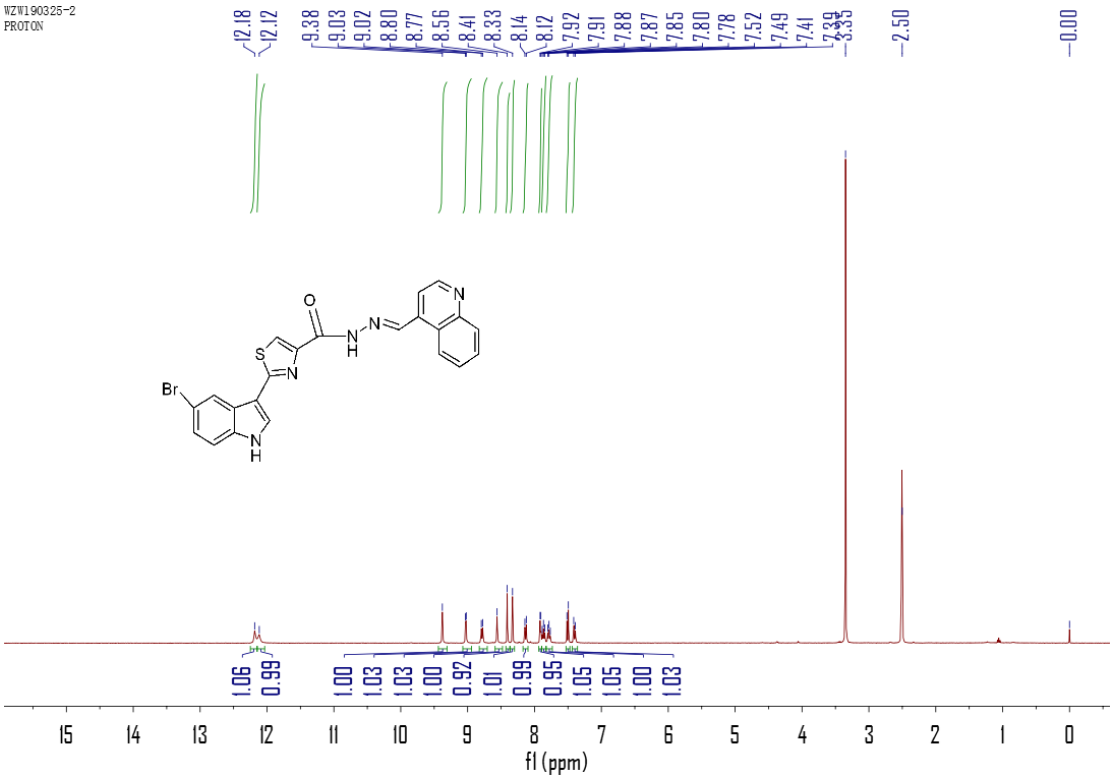
^{13}C NMR spectrum of **10h**



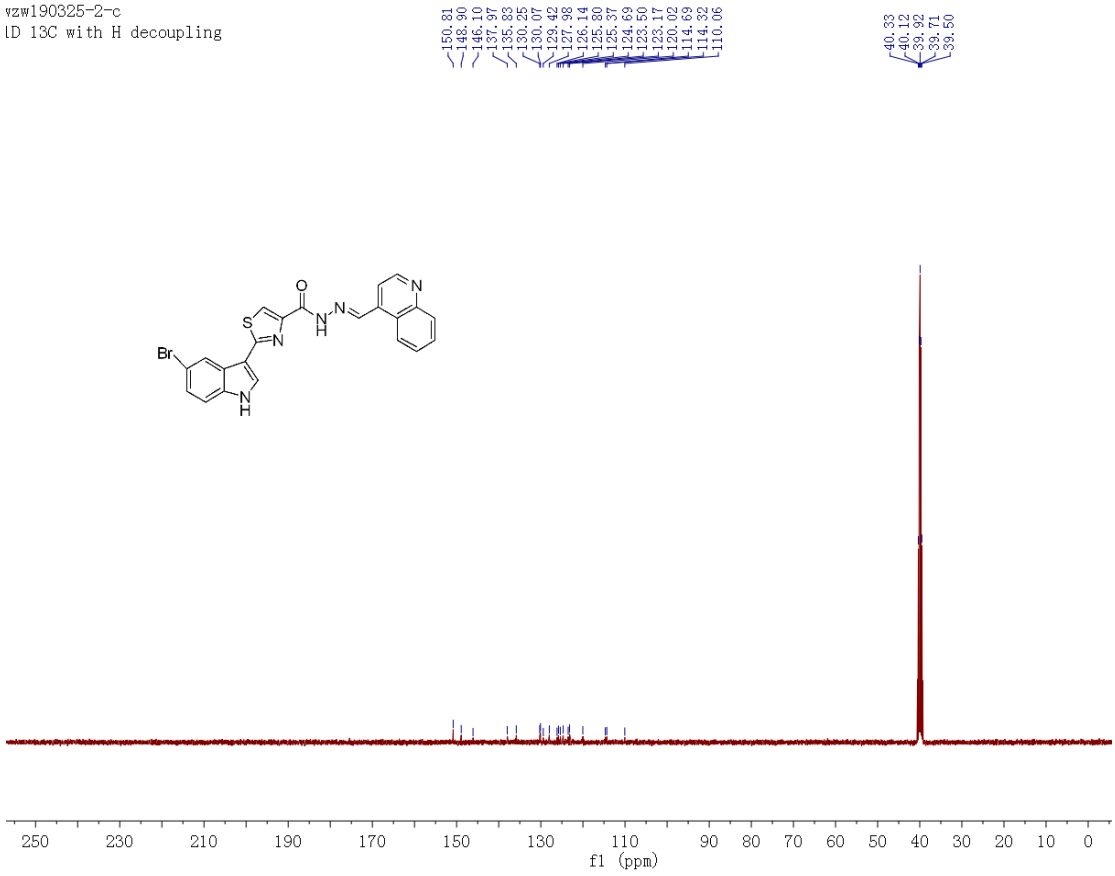
^{1}H NMR spectrum of **10i**



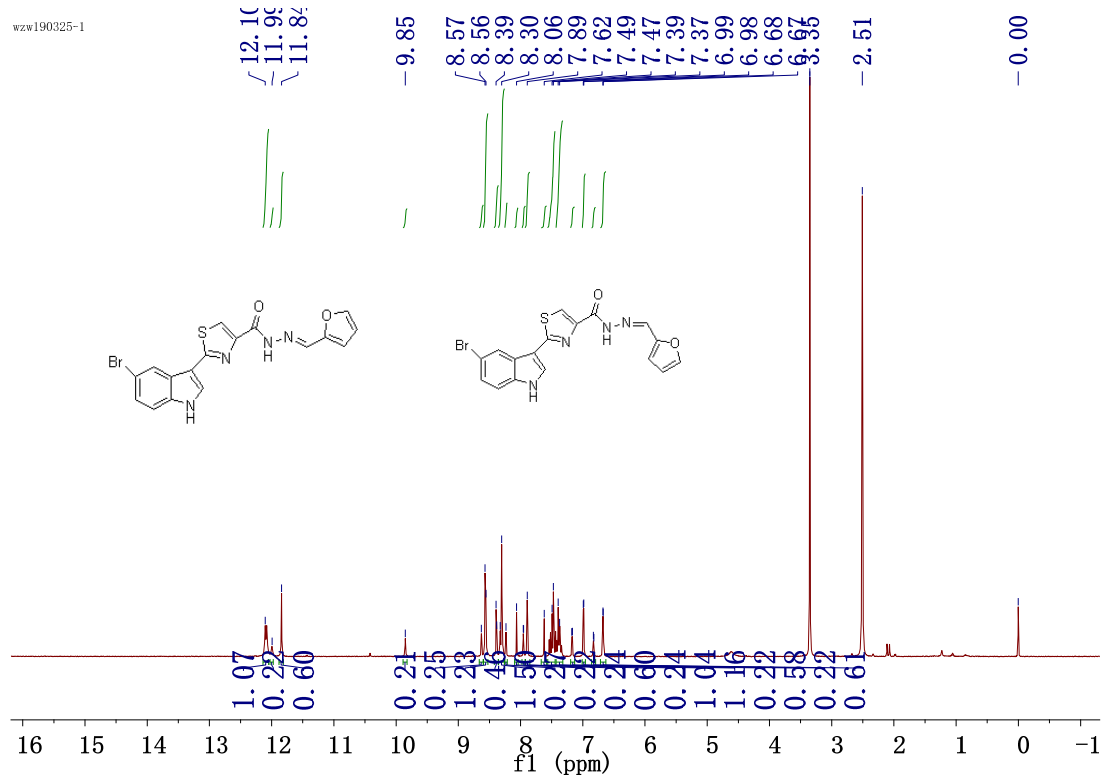
^{13}C NMR spectrum of **10i**



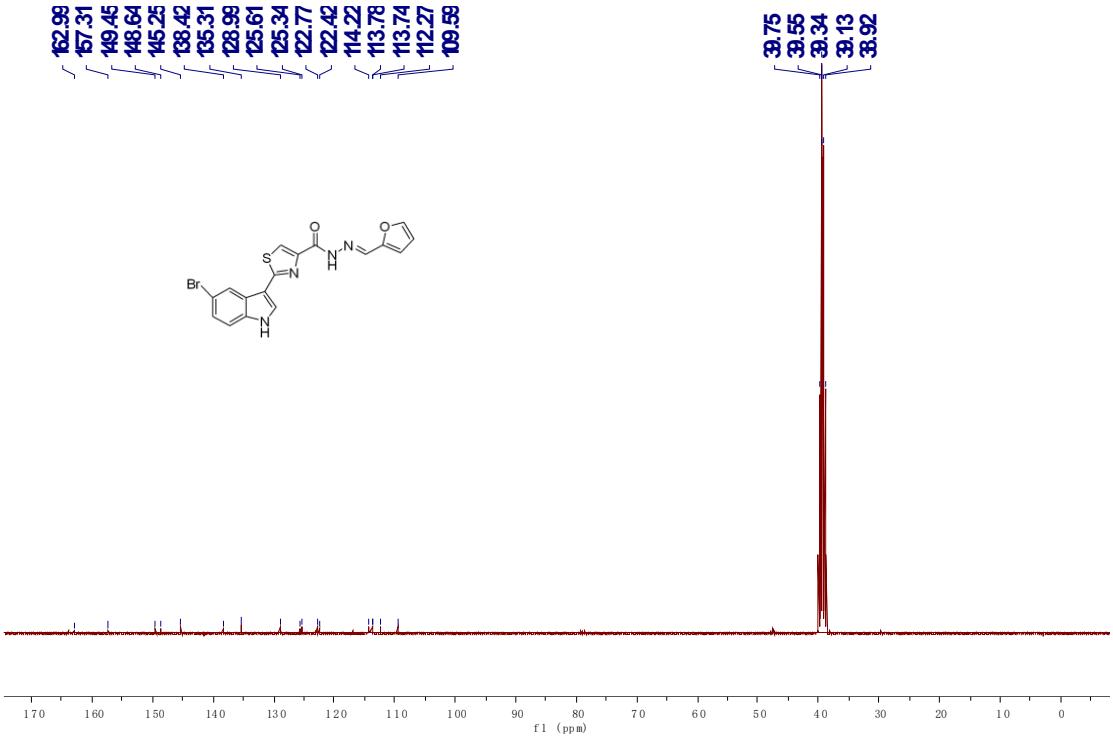
^{1}H NMR spectrum of **10j**



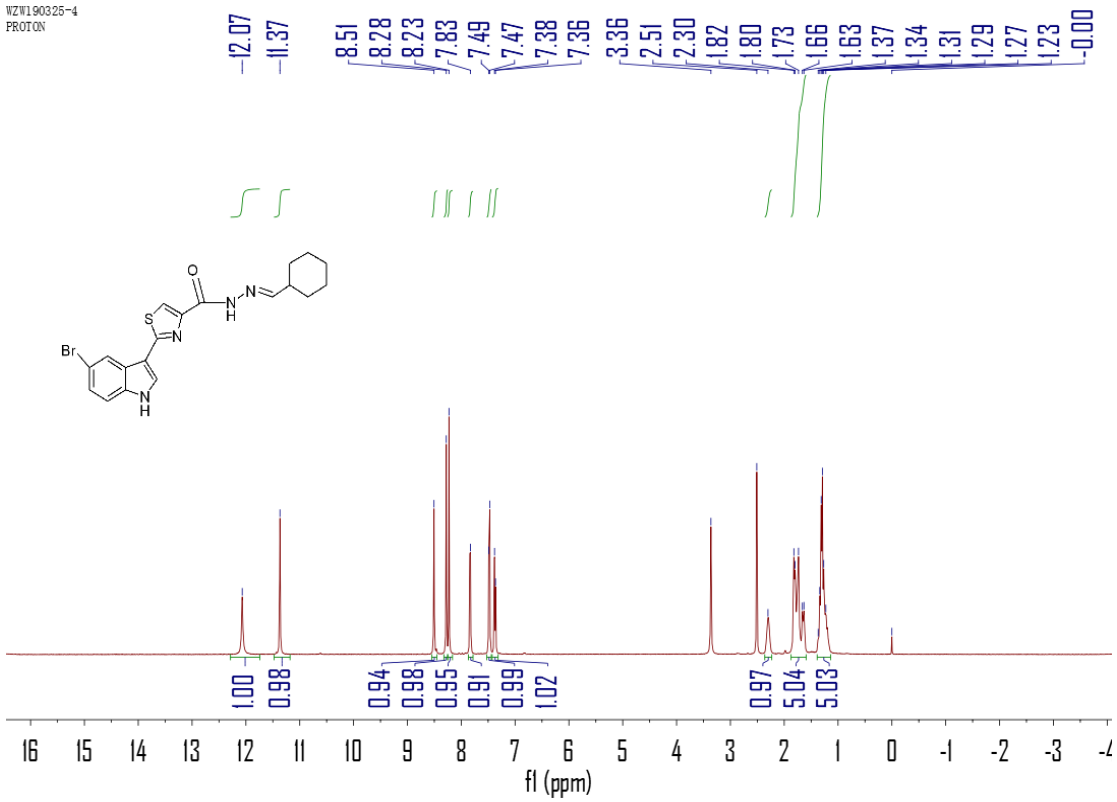
^{13}C NMR spectrum of **10j**



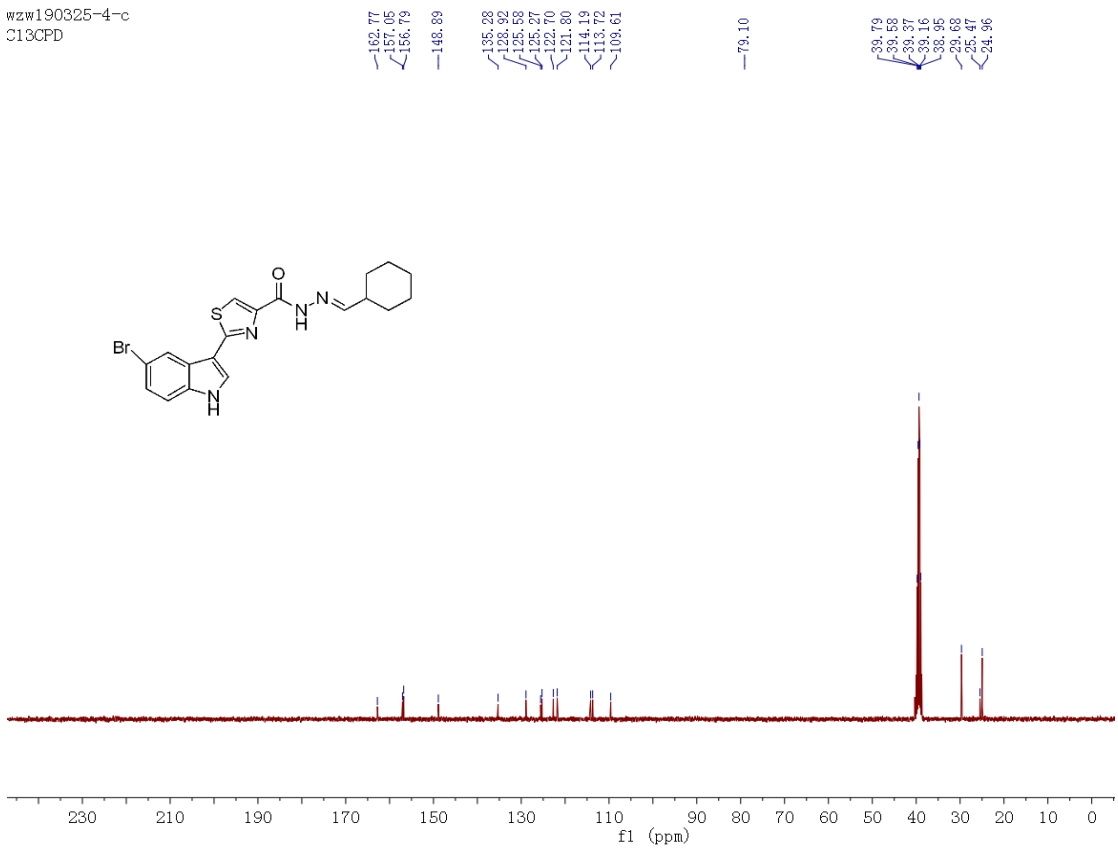
^{1}H NMR spectrum of **10k**



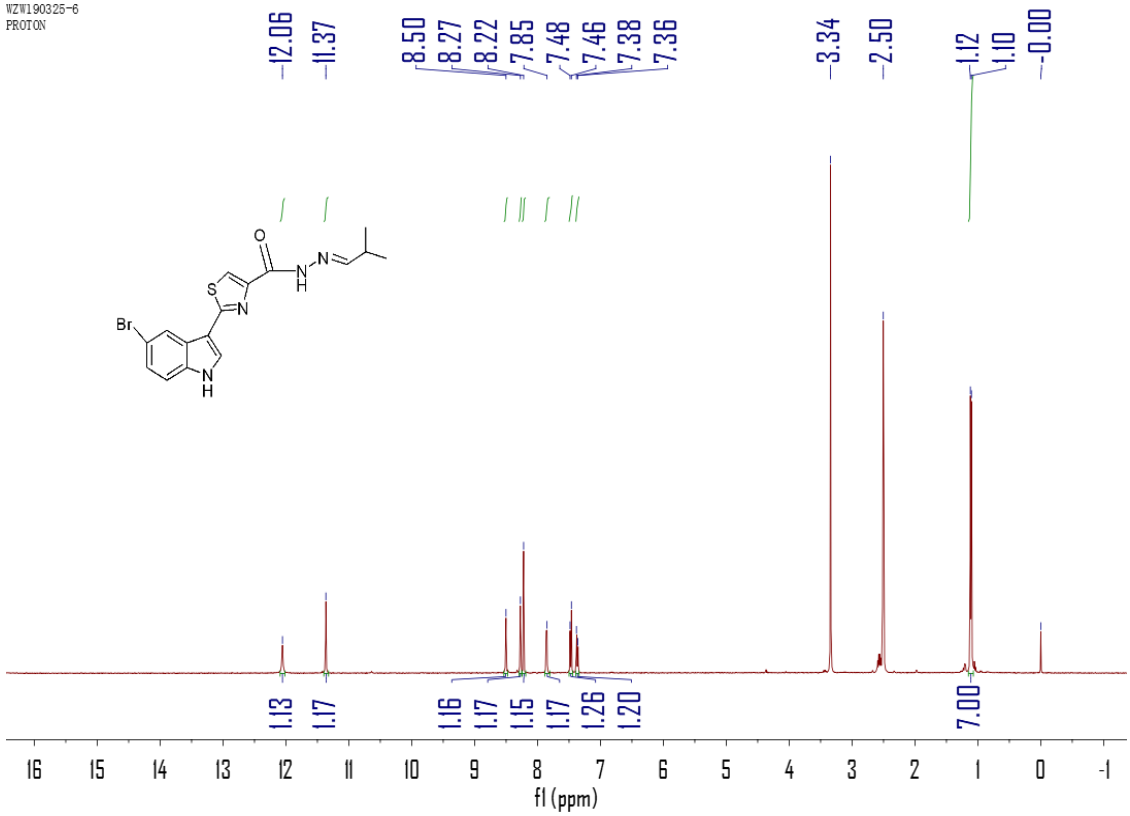
^{13}C NMR spectrum of **10k**



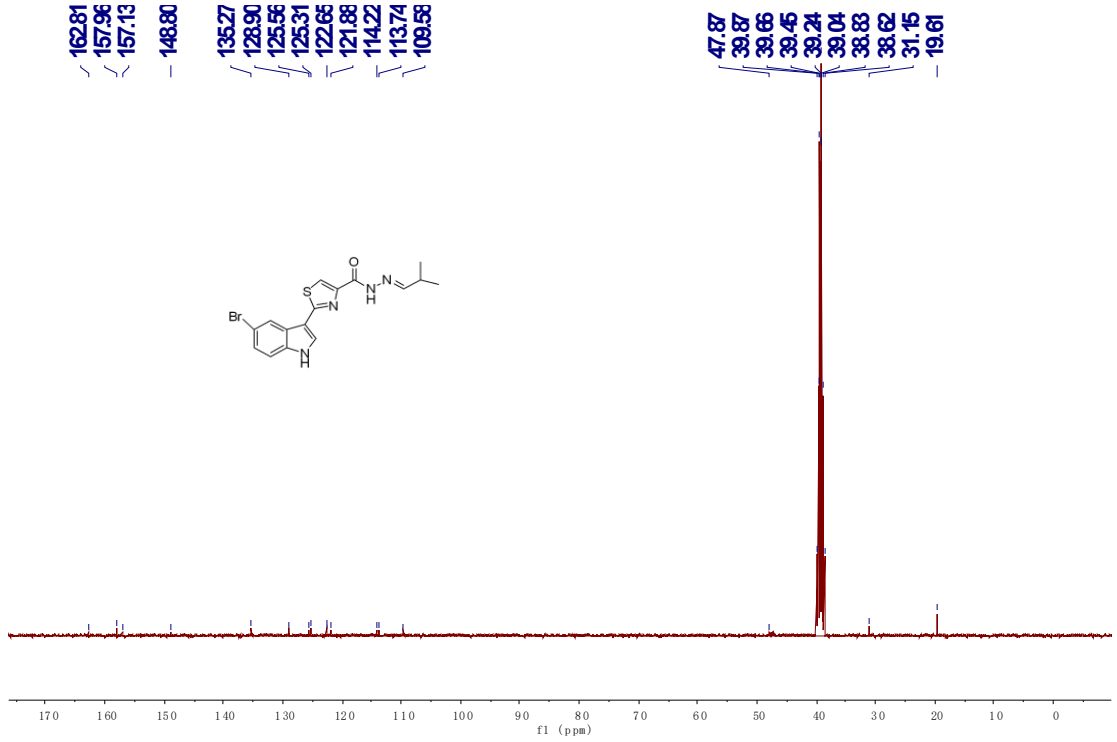
^{1}H NMR spectrum of **10l**



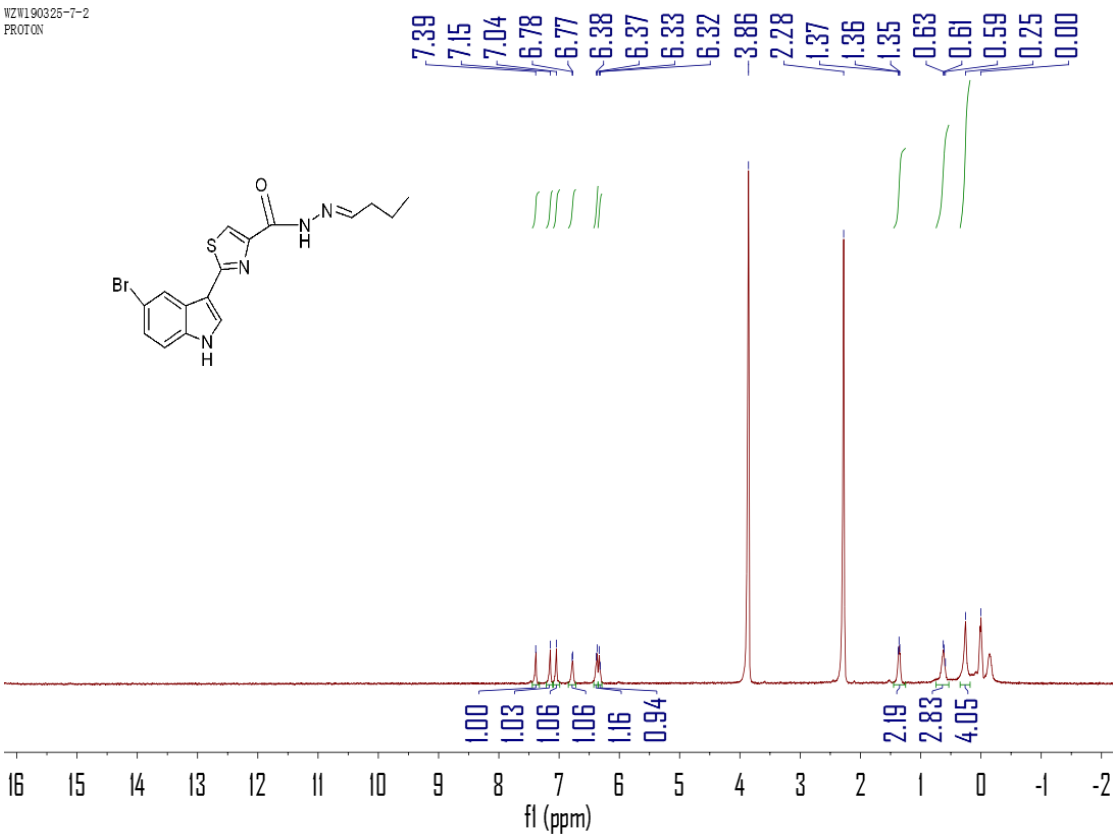
^{13}C NMR spectrum of **10l**



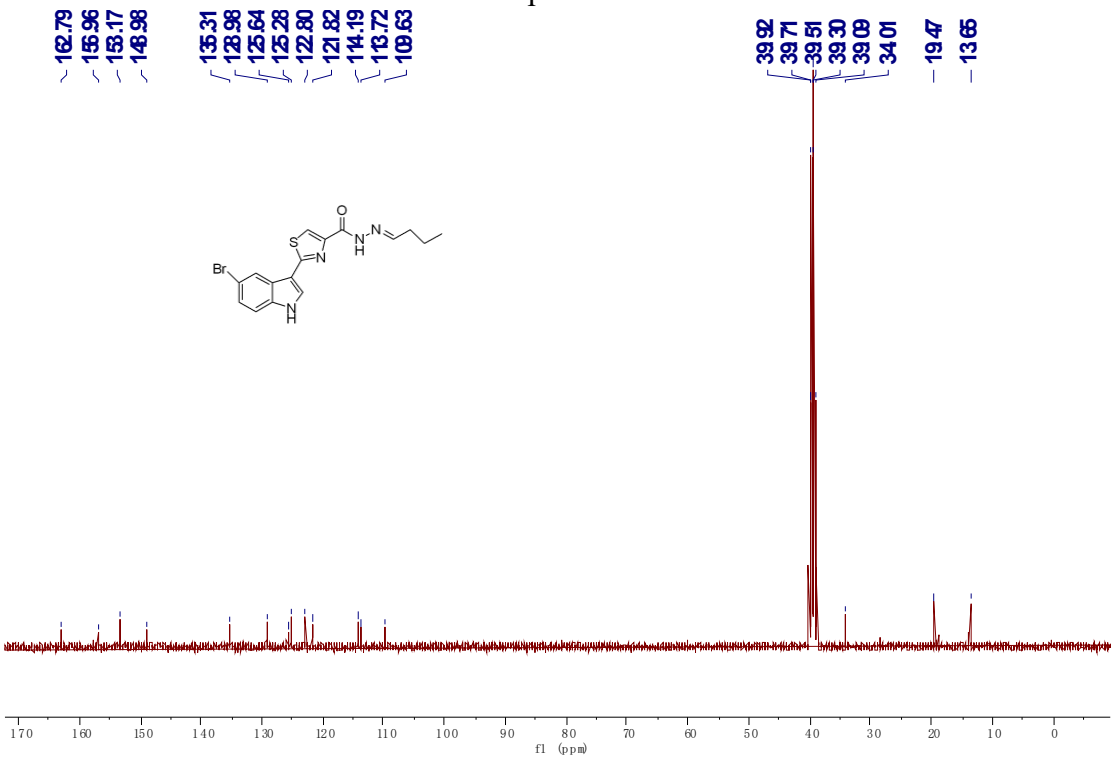
^{1}H NMR spectrum of **10m**



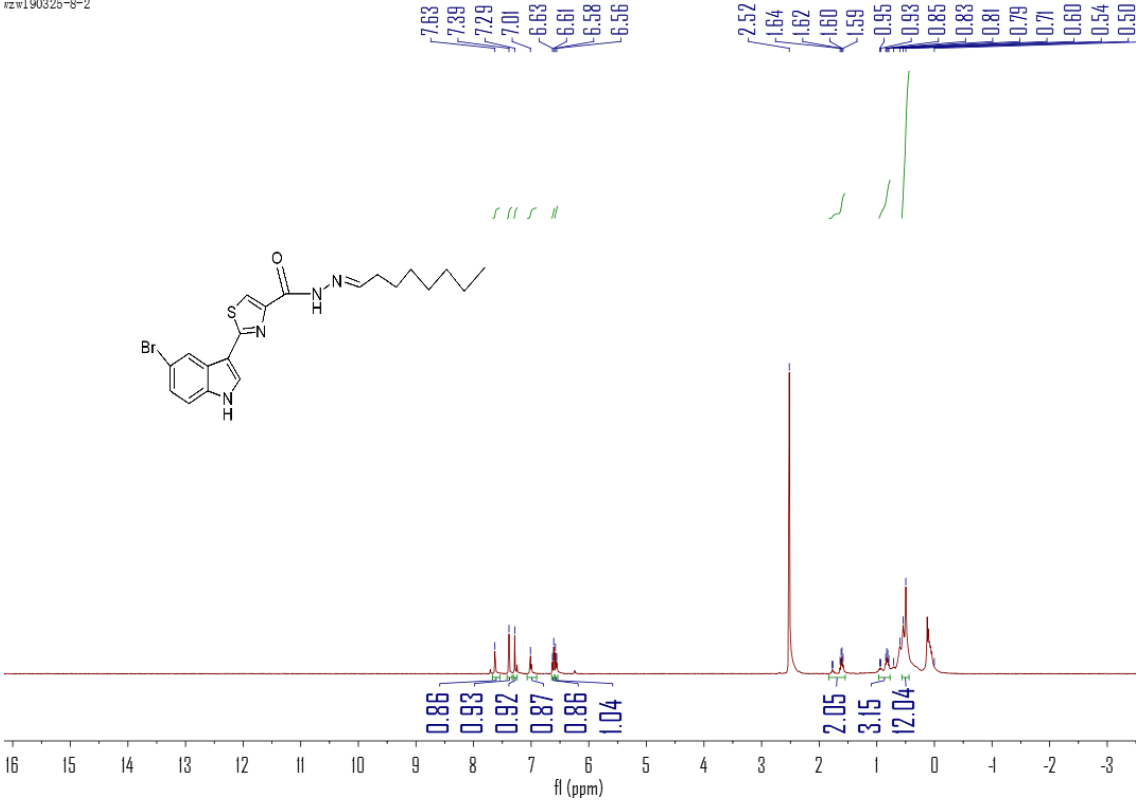
^{13}C NMR spectrum of **10m**



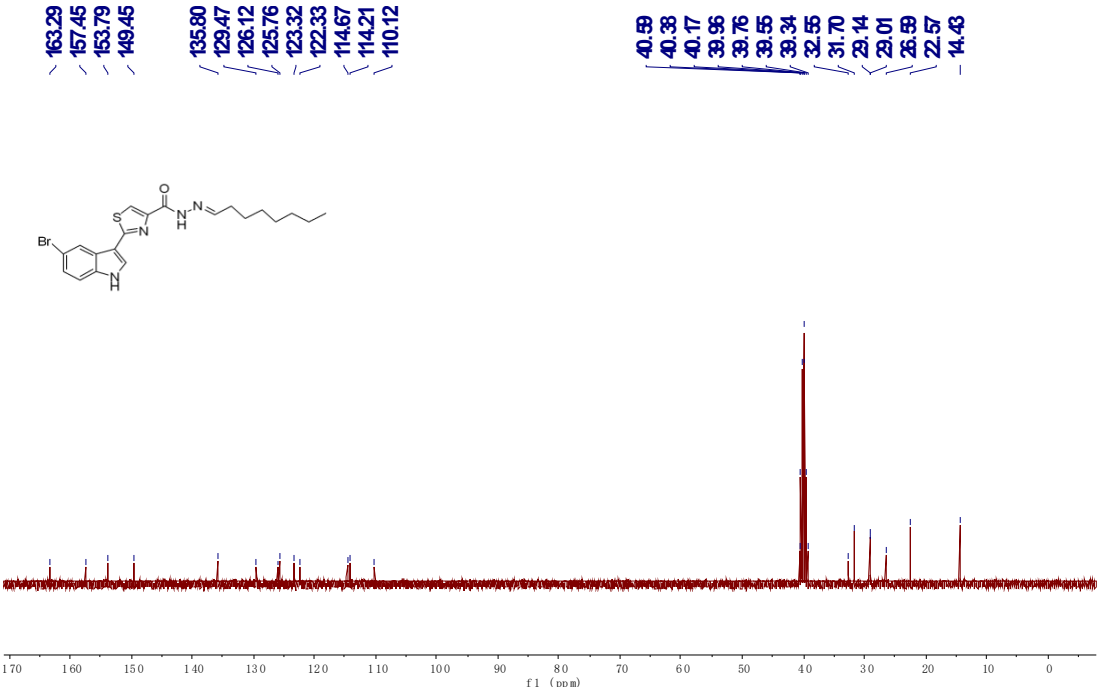
^{1}H NMR spectrum of **10n**



^{13}C NMR spectrum of **10n**



^{1}H NMR spectrum of **10o**



^{13}C NMR spectrum of **10o**