

# Support Information

## Hybrids of Phenylsulfonylfuroxan and Coumarin as Potent Antitumor Agents

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## The Effect of Hemoglobin on the Proliferation of Cells.

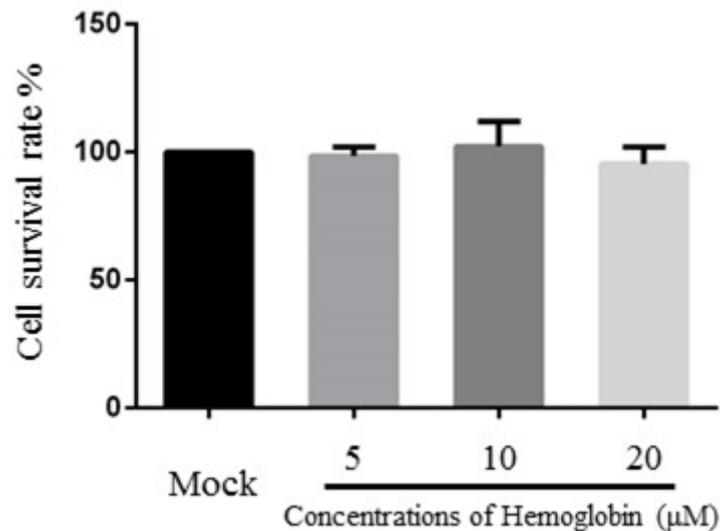


Figure S1. The effect of Hemoglobin on Cellular Proliferation.

Cells were treated with vehicle (PBS) and different concentrations of hemoglobin for 24h, and MTT assay was used to detect the cellular proliferation.

### Methods for MEK1 Inhibitory Activity<sup>1</sup>.

Caliper Mobility Shift assay (MSA) was used to test MEK1 inhibitory activity. The active MEK1 was obtained from Invitrogen and was tested at the concentration of 0.015 nM with 100 µM ATP, 0.05 µM unactive ERK1, 2 mM DTT and the reaction buffer contained 20 mM Ph 7.5 HEPES, 10 mM MgCl<sub>2</sub>, and 0.01% Triton X100. The kinase assay was carried out for 120 min at 28 °C and was terminated by addition 25 uL stop buffer (100 mM HEPES, pH 7.5, 0.015% Brij-35, 0.2% Coating Reagent #3, 50 mM EDTA). Then the data was collected on Caliper EZ Reader.

## High Resolution Mass Spectral Data for Target Compounds.

**Table S1.** High Resolution Mass Spectra for Target Compounds.

compd.	High resolution mass spectra			
	Chemical formula	Calculated	Measured	ppm error
<b>6a</b>	C <sub>17</sub> H <sub>10</sub> N <sub>2</sub> O <sub>7</sub> S + H	387.0287	387.0292	1.3
<b>6b</b>	C <sub>18</sub> H <sub>12</sub> N <sub>2</sub> O <sub>7</sub> S + H	401.0443	401.0446	0.7
<b>6c</b>	C <sub>19</sub> H <sub>14</sub> N <sub>2</sub> O <sub>7</sub> S + H	415.0600	415.0600	0
<b>8a</b>	C <sub>19</sub> H <sub>14</sub> N <sub>2</sub> O <sub>8</sub> S + H	431.0549	431.0557	1.8
<b>8b</b>	C <sub>20</sub> H <sub>16</sub> N <sub>2</sub> O <sub>8</sub> S + H	445.0706	445.0712	1.3
<b>8c</b>	C <sub>20</sub> H <sub>16</sub> N <sub>2</sub> O <sub>7</sub> S <sub>2</sub> + H	461.0477	461.0489	2.6
<b>8d</b>	C <sub>21</sub> H <sub>18</sub> N <sub>2</sub> O <sub>8</sub> S + H	459.0862	459.0864	0.4
<b>8e</b>	C <sub>22</sub> H <sub>20</sub> N <sub>2</sub> O <sub>8</sub> S + H	473.1019	473.1021	0.4
<b>8f</b>	C <sub>21</sub> H <sub>18</sub> N <sub>2</sub> O <sub>8</sub> S + H	459.0862	459.0864	0.4
<b>8g</b>	C <sub>20</sub> H <sub>17</sub> N <sub>3</sub> O <sub>7</sub> S + H	444.0865	444.0867	0.4
<b>10a</b>	C <sub>21</sub> H <sub>19</sub> N <sub>3</sub> O <sub>7</sub> S + H	458.1022	458.1033	2.4
<b>13a</b>	C <sub>21</sub> H <sub>16</sub> N <sub>2</sub> O <sub>10</sub> S + H	489.0604	489.0613	1.8
<b>13b</b>	C <sub>22</sub> H <sub>18</sub> N <sub>2</sub> O <sub>10</sub> S + H	503.0760	503.0762	0.4
<b>15</b>	C <sub>19</sub> H <sub>14</sub> N <sub>2</sub> O <sub>7</sub> S + H	415.0600	415.0604	0.9
<b>17a</b>	C <sub>21</sub> H <sub>18</sub> N <sub>2</sub> O <sub>8</sub> S + H	459.0862	459.0859	0.6
<b>17b</b>	C <sub>22</sub> H <sub>20</sub> N <sub>2</sub> O <sub>8</sub> S + H	473.1019	473.1011	1.7

## HPLC Assessment of Compound Purity.

As showed in Table W, all tested compounds (**6a-c**, **8a-g**, **10a**, **13a-b**, **15**, **17a-b** and **18**) with a purity of > 95% (HPLC analysis) were used for subsequent experiments. We provided the spectra of HPLC assays as below.

(**8g**, **8e**, **8b**, **10a**) utilized the Column: ODS-C<sub>18</sub> (150 mm×4.6 mm×3.5 μm); the rest compounds utilized the Column: ODS –C<sub>18</sub> (150 mm×4.6 mm×5 μm).

Mobile phase: acetonitrile-water (70: 30)

Wavelength: 254 nm

Rate: 1 mL/min

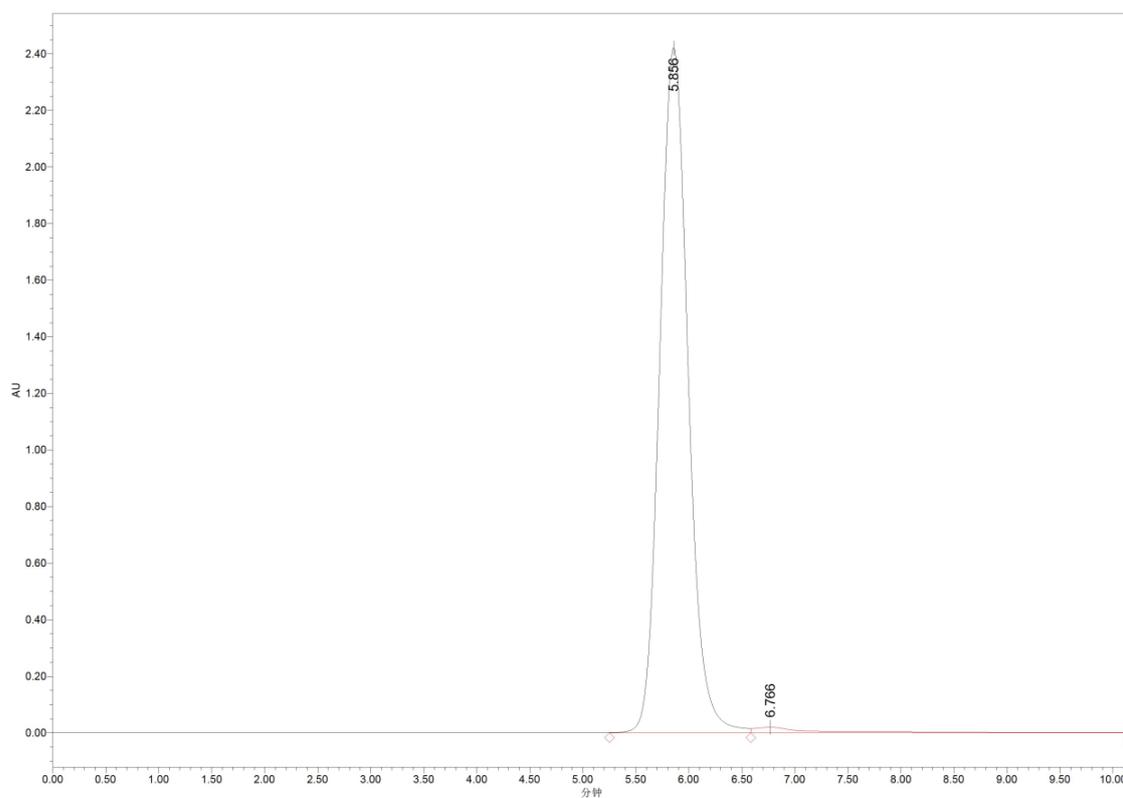
Temperature: 25 °C

**Table S2.** Purity of Target Compounds.

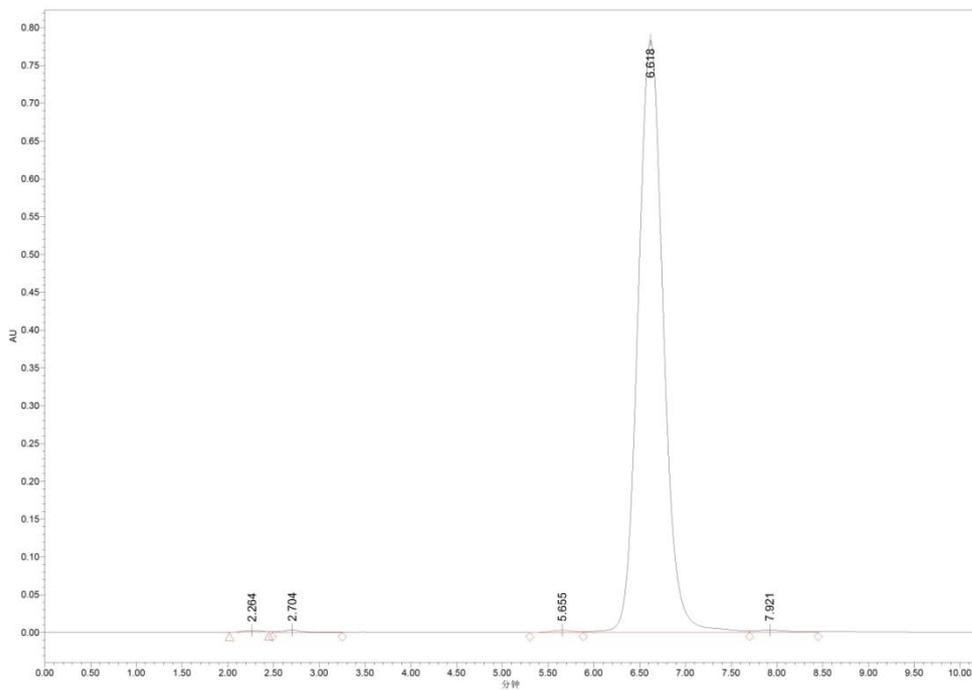
Compound	Purity (area %)	Retention time (min)
<b>6a</b>	98.24	5.856
<b>6b</b>	99.77	6.618
<b>6c</b>	99.76	7.376
<b>8a</b>	96.64	5.89
<b>8b</b>	97.82	5.33
<b>8c</b>	96.25	7.816
<b>8d</b>	97.09	7.432
<b>8e</b>	98.21	5.978
<b>8f</b>	99.65	7.031
<b>8g</b>	99.29	4.74
<b>10a</b>	98.06	5.799
<b>13a</b>	97.7	5.595
<b>13b</b>	99.02	6.036
<b>15</b>	99.33	6.597
<b>17a</b>	98.52	6.111
<b>17b</b>	98.69	7.293
<b>18</b>	97.01	3.350

The details are listed as below:

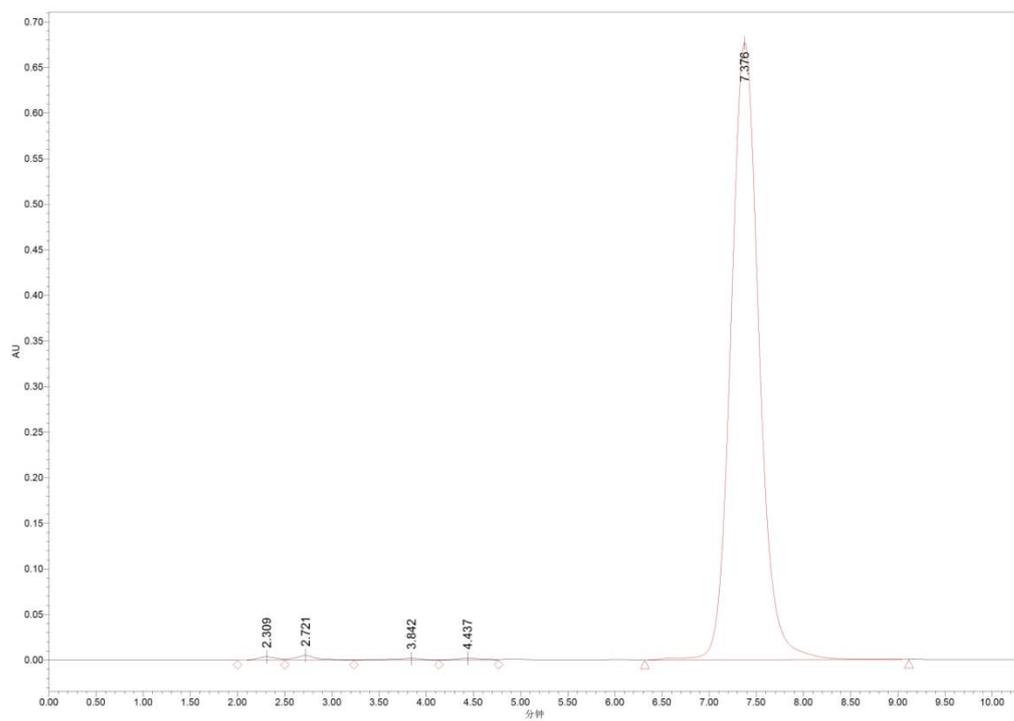
**6a**, 98.24%, retention time: 5.856 min.



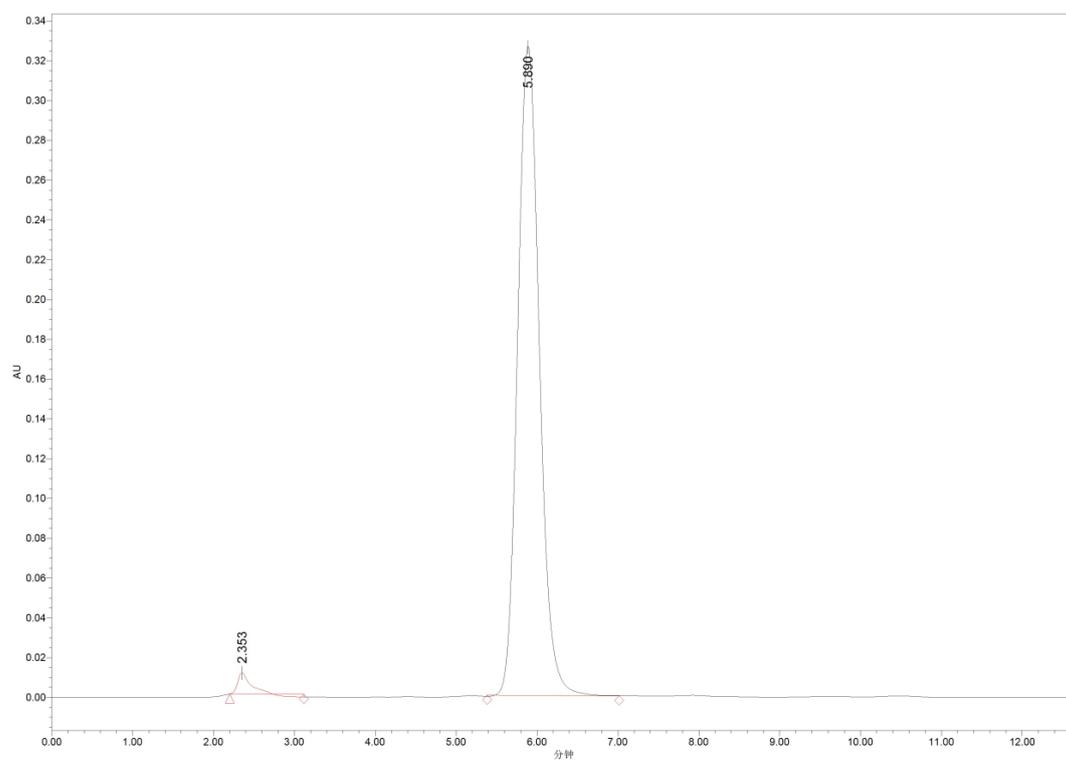
**6b**, 99.77%, retention time: 6.618 min.



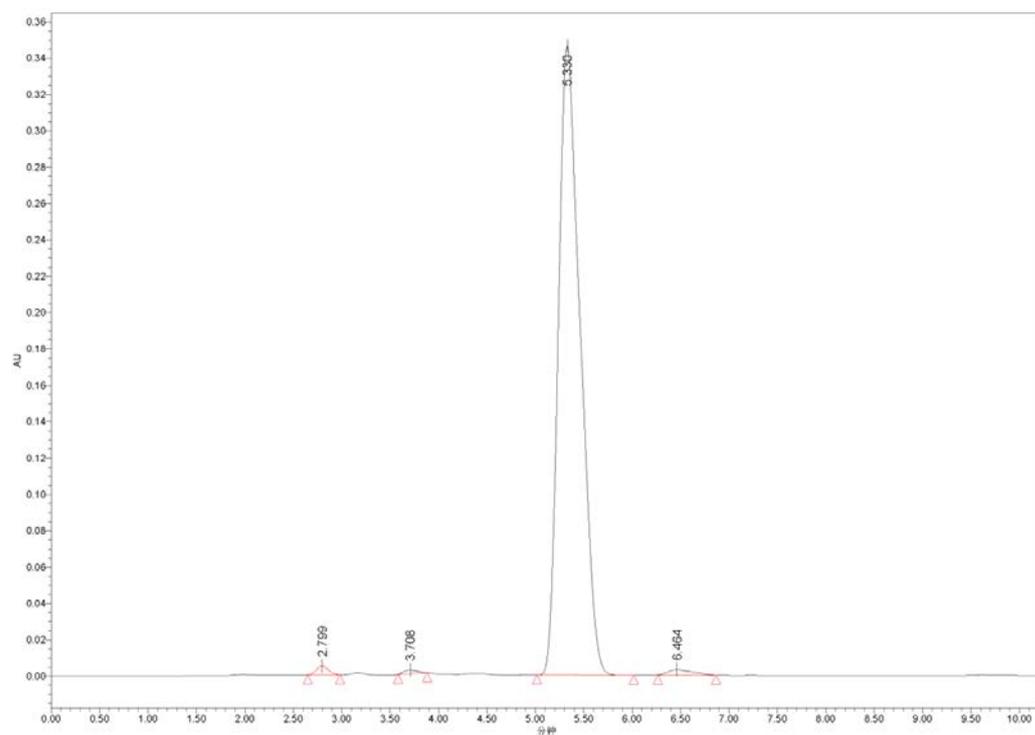
**6c**, 99.76%, retention time: 7.376 min.



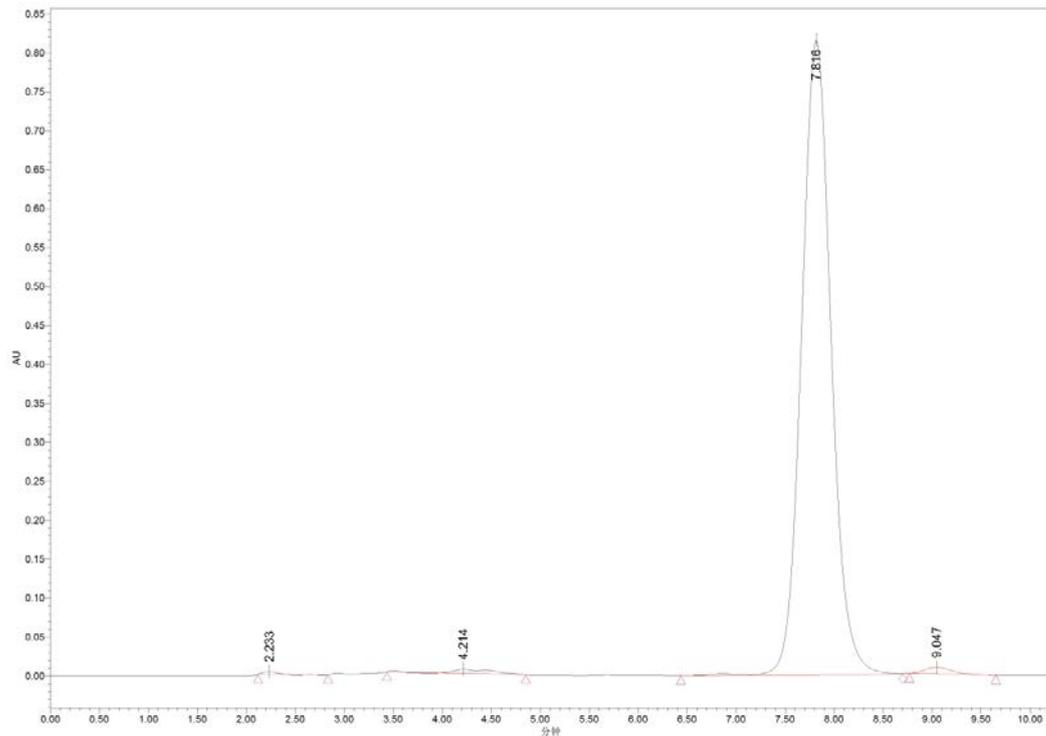
**8a**, 96.64%, retention time: 5.890 min.



**8b**, 97.82%, retention time: 5.330 min.

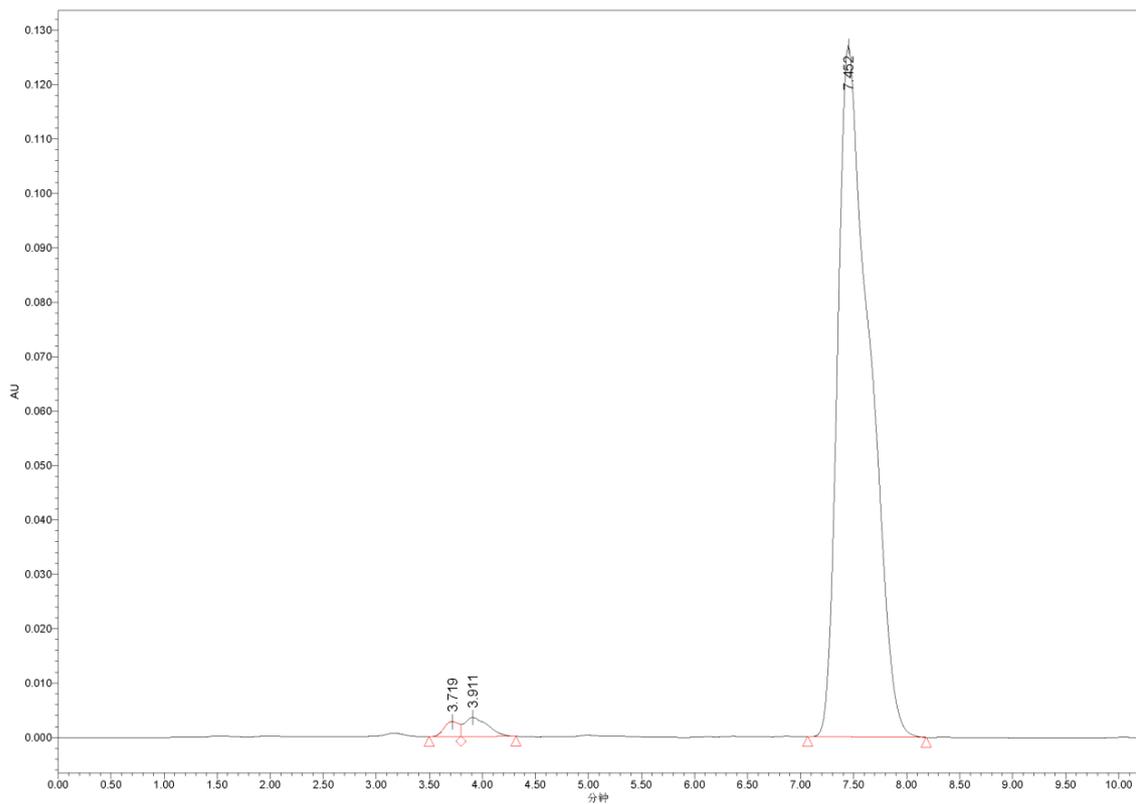


**8c**, 96.25%, retention time: 7.816 min.

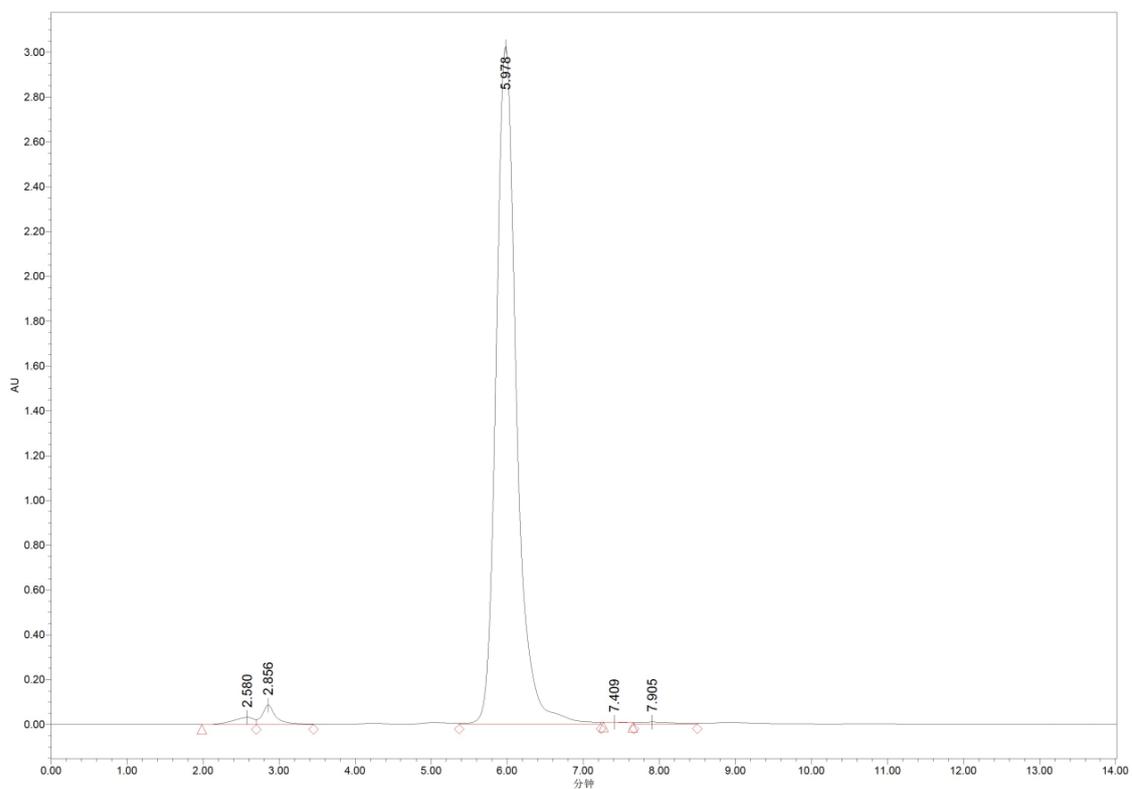


**8d**, 97.09%, retention time: 7.432 min.

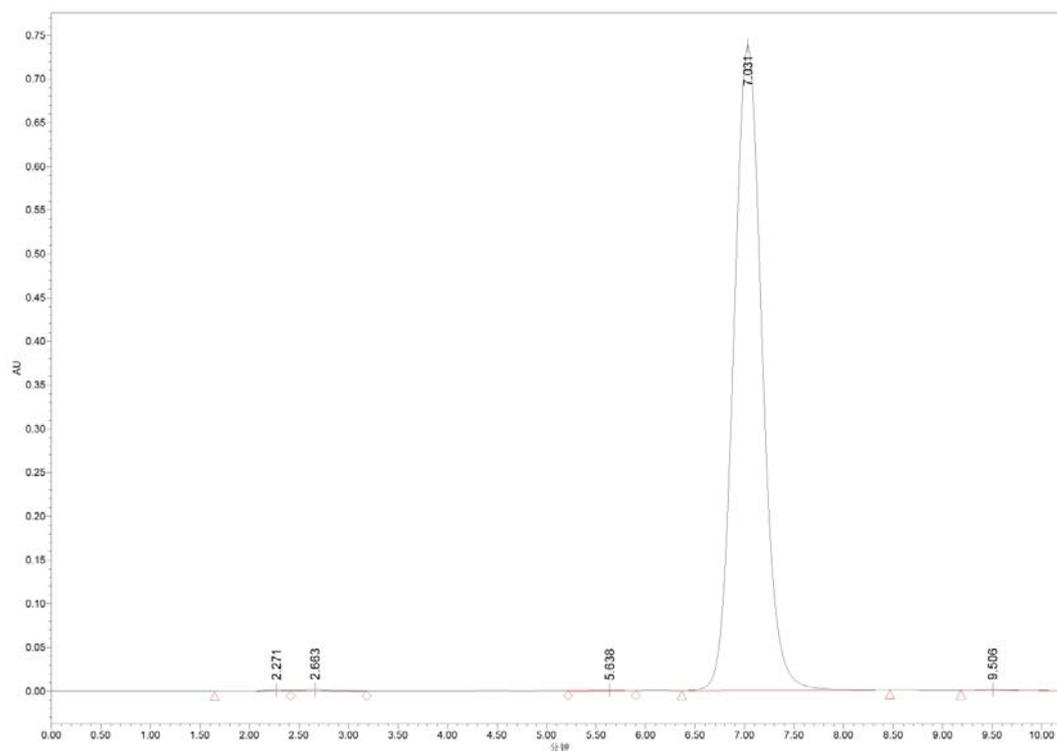
S7



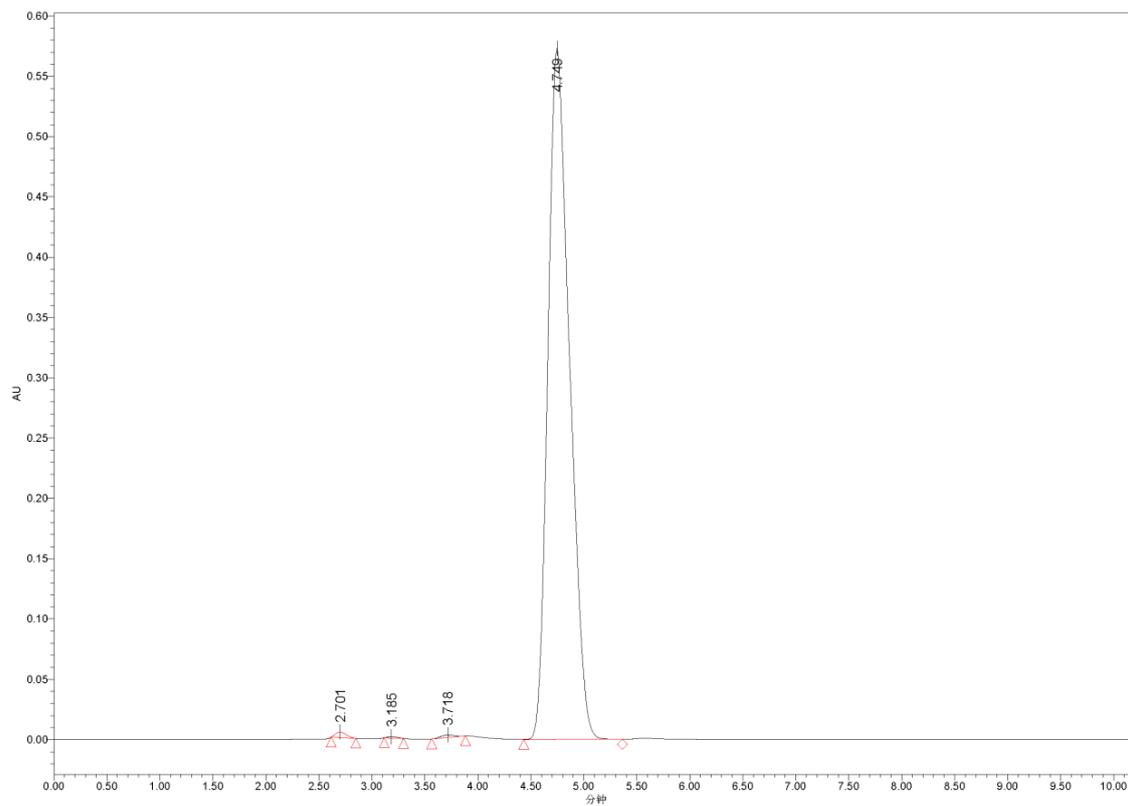
**8e**, 98.21%, retention time: 5.978 min.



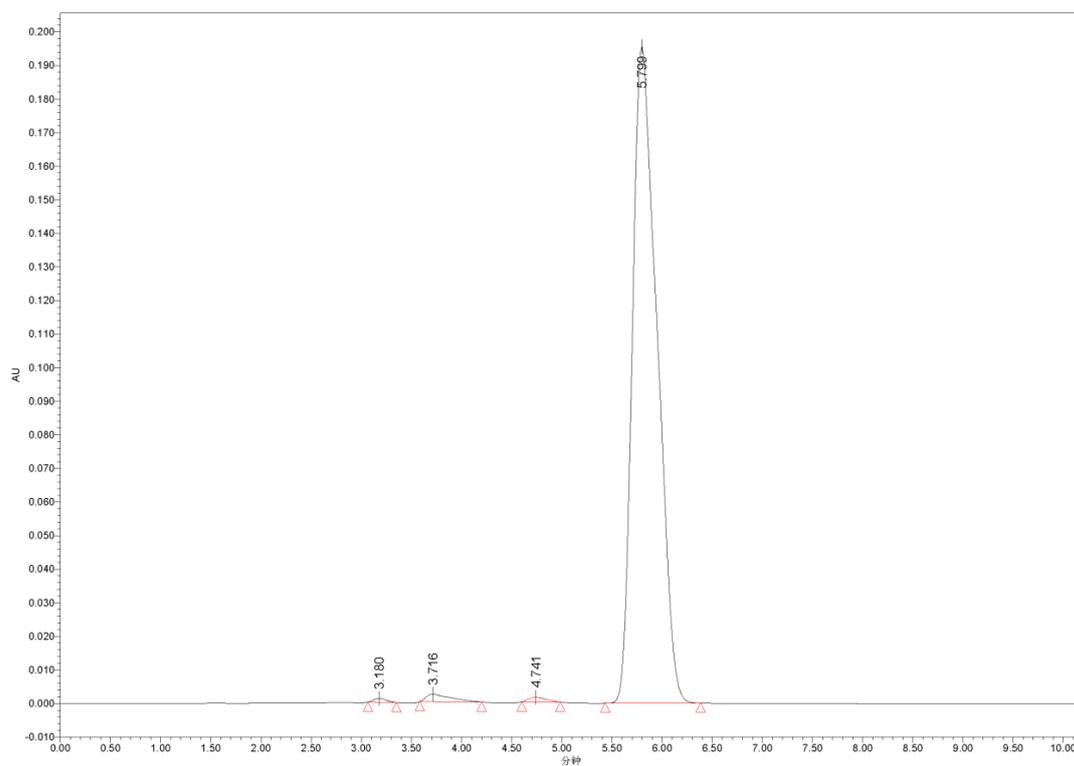
**8f**, 99.65%, retention time: 7.031 min.



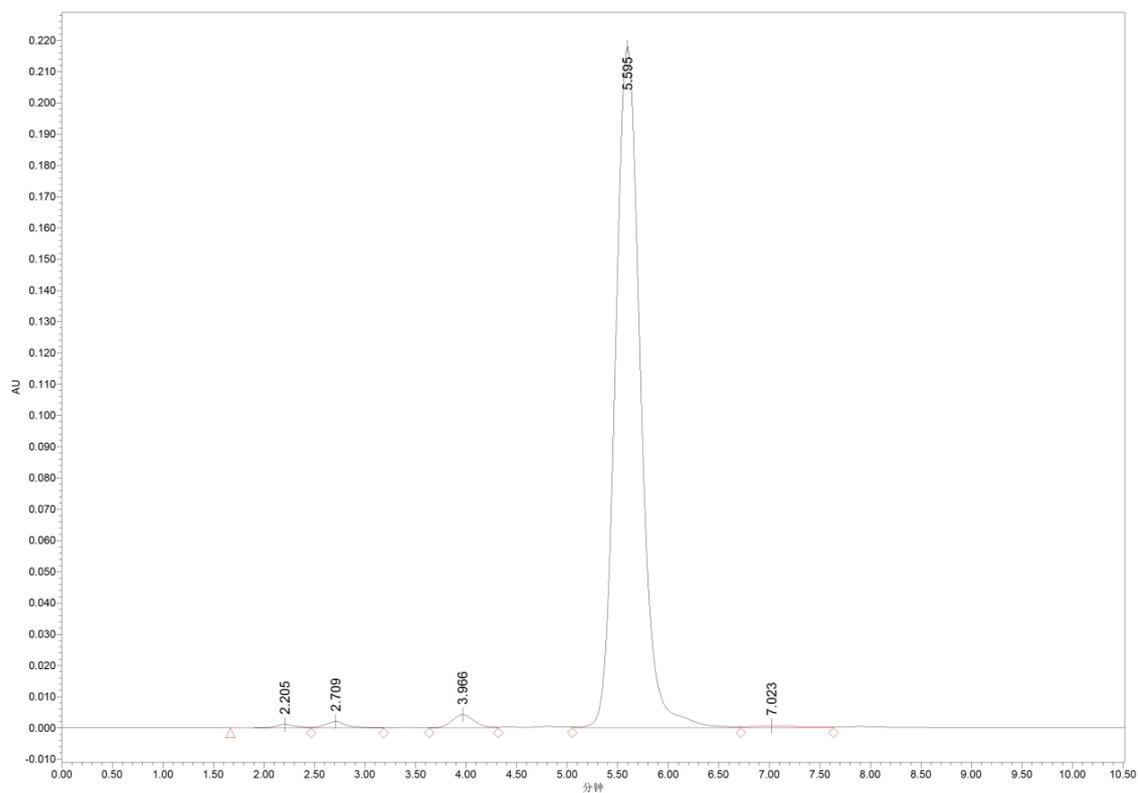
**8g**, 99.29%, retention time: 4.749 min.



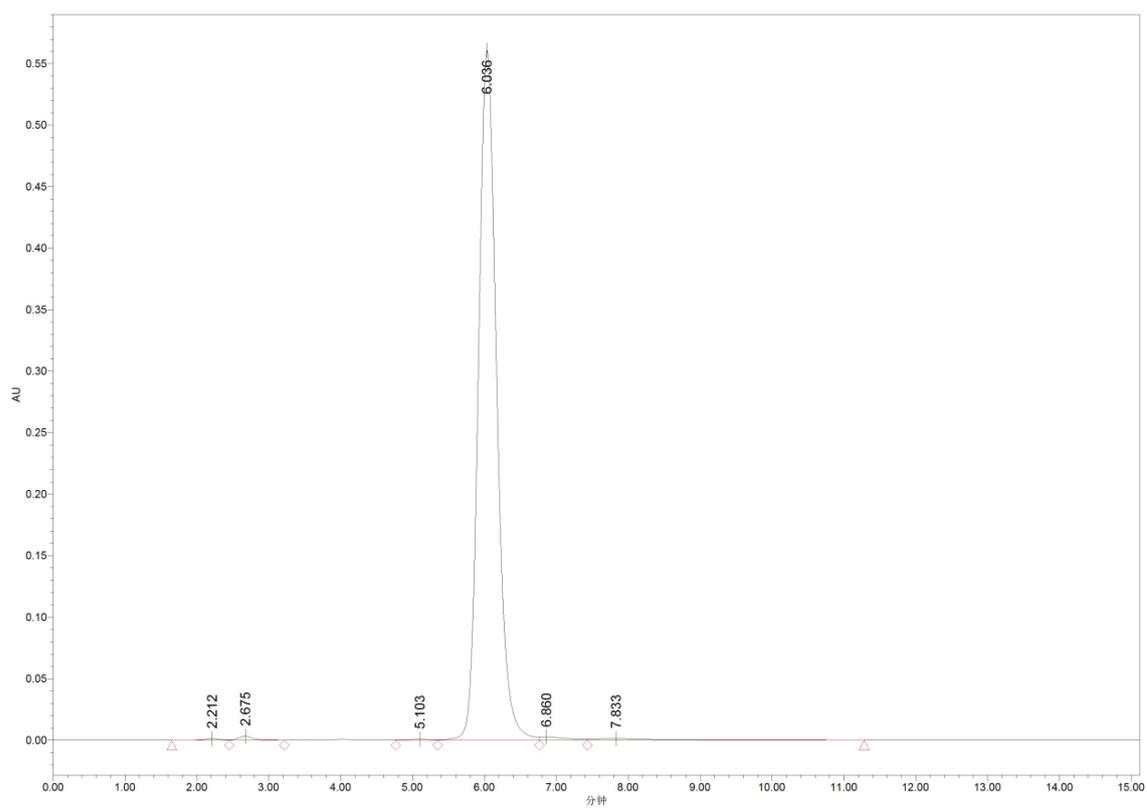
**10a**, 98.06%, retention time: 5.799 min.



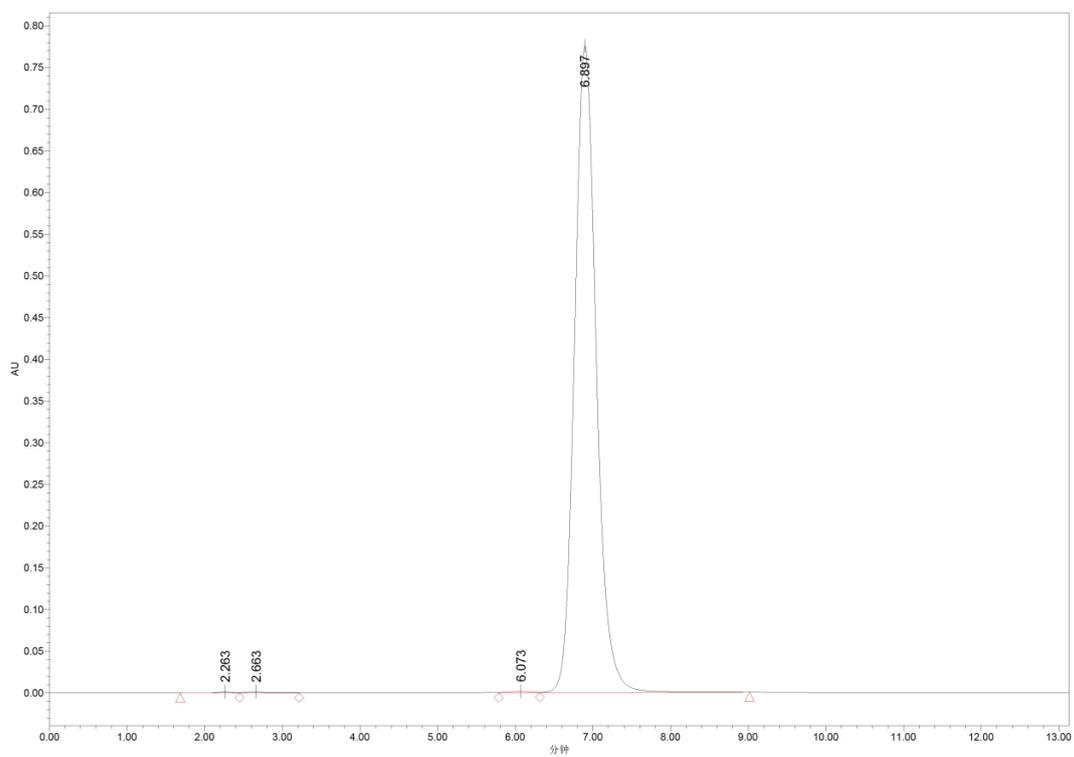
**13a**, 97.70%, retention time: 5.595 min.



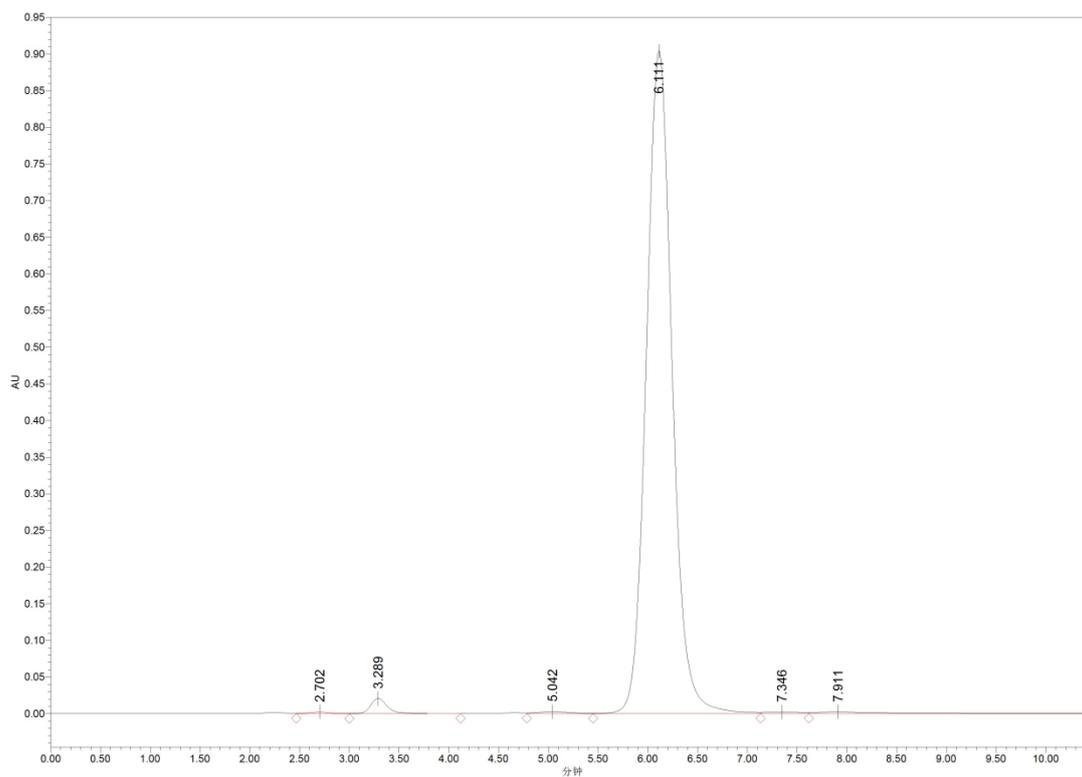
**13b**, 99.02%, retention time: 6.036 min.



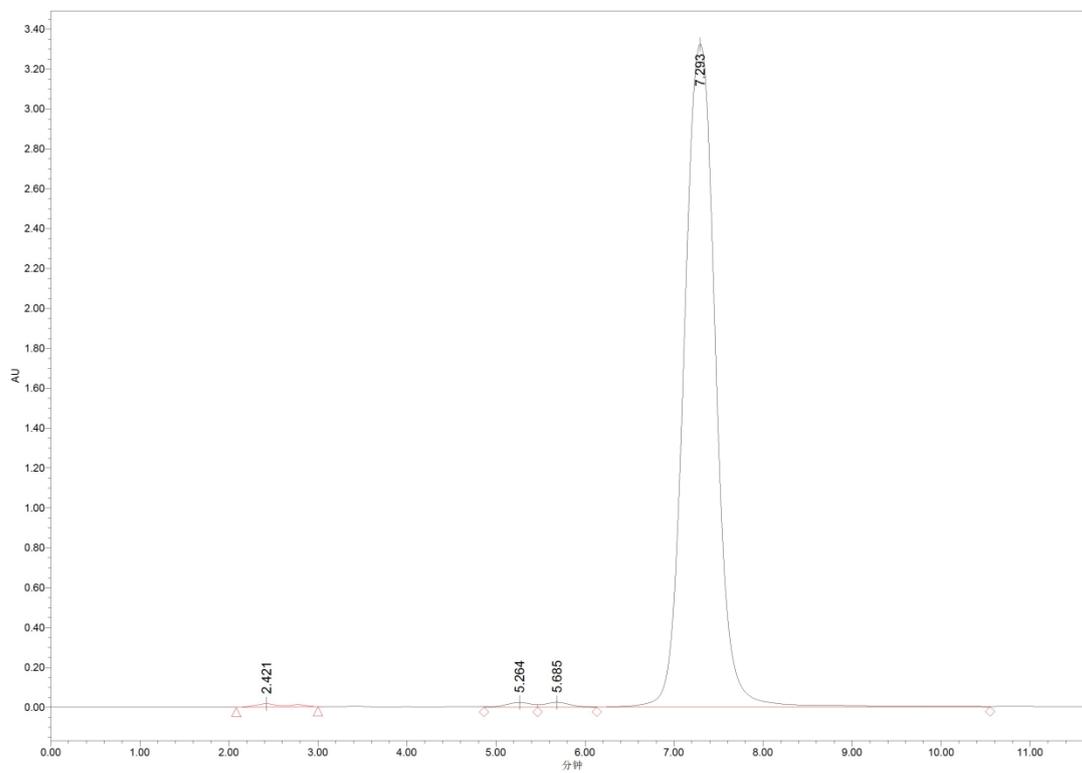
**15**, 99.33%, retention time: 6.597 min.



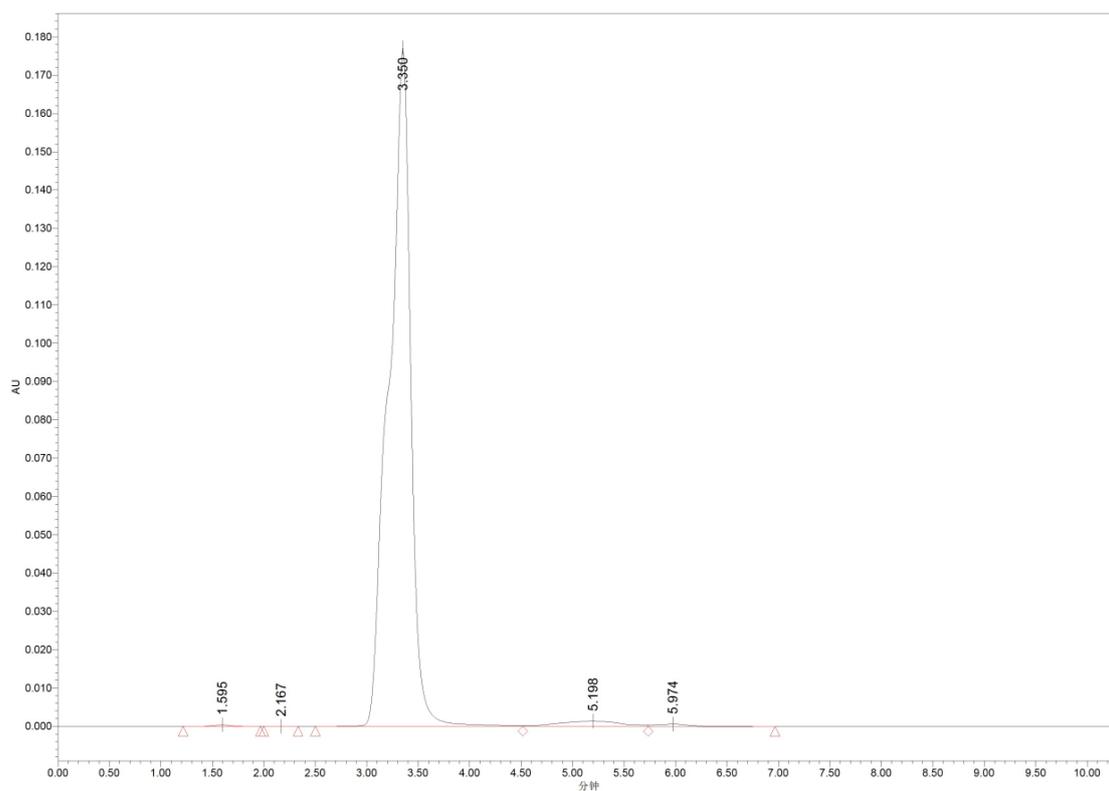
**17a**, 98.52%, retention time: 6.111 min.



**17b**, 98.69%, retention time: 7.293 min.

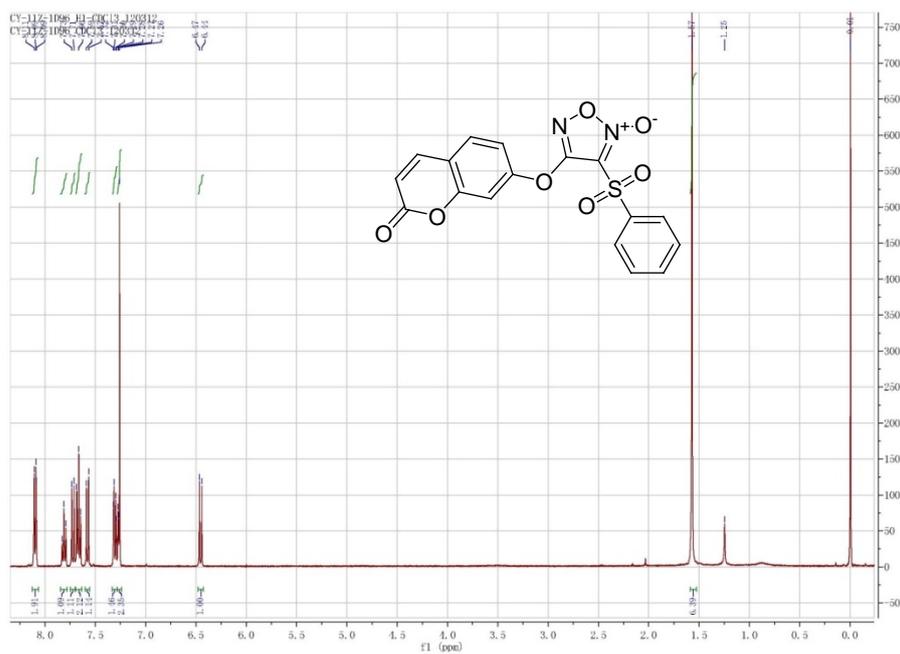


18, 97.01%, retention time: 3.350



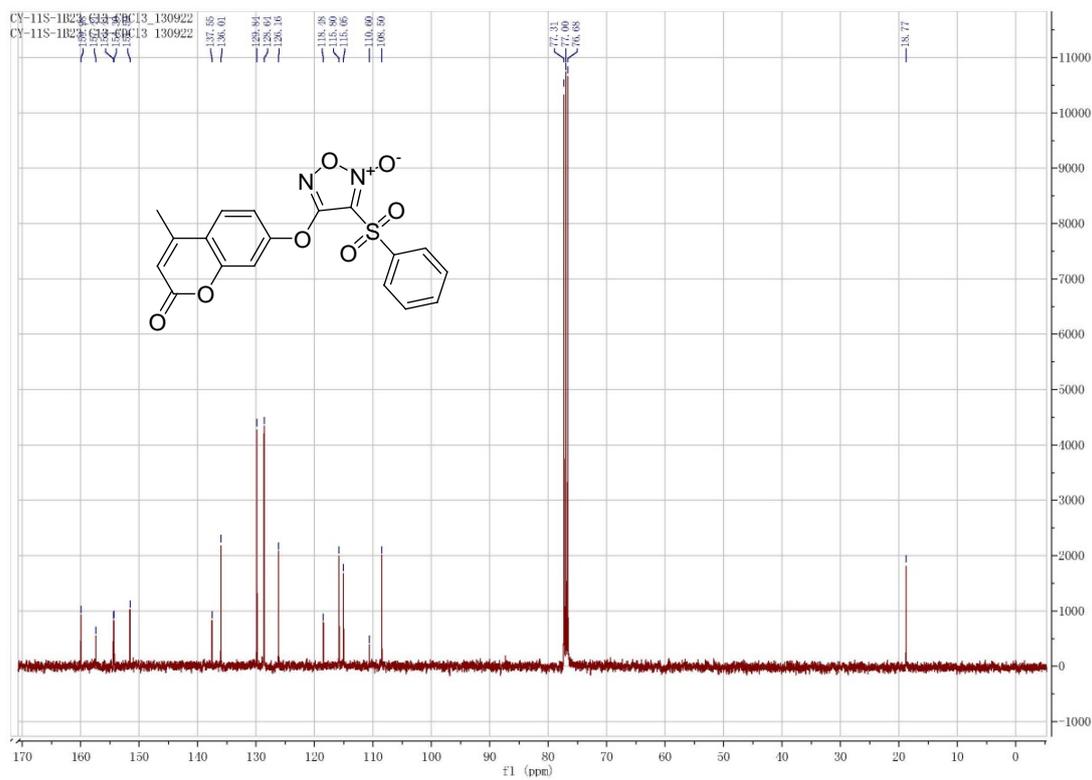
**<sup>1</sup>H NMR and <sup>13</sup>C NMR Data of Target Compounds 6a-c, 8a-g, 10a, 13a-b, 15, 17a-b.**

**Compound 6a: <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>)**

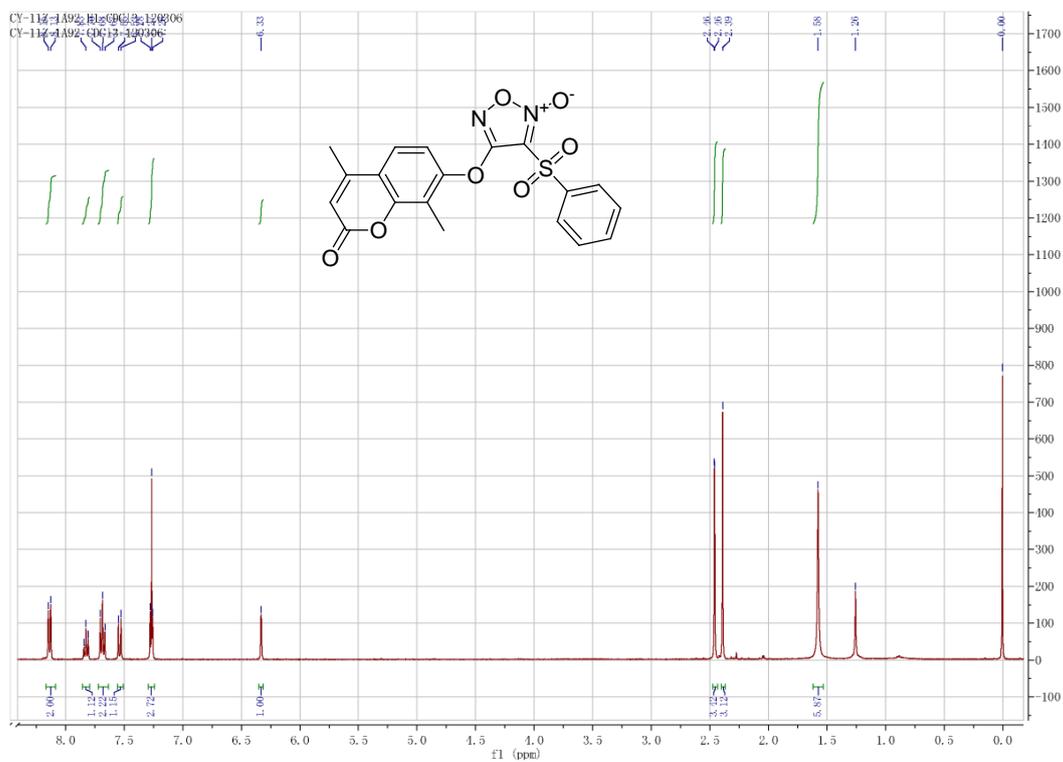




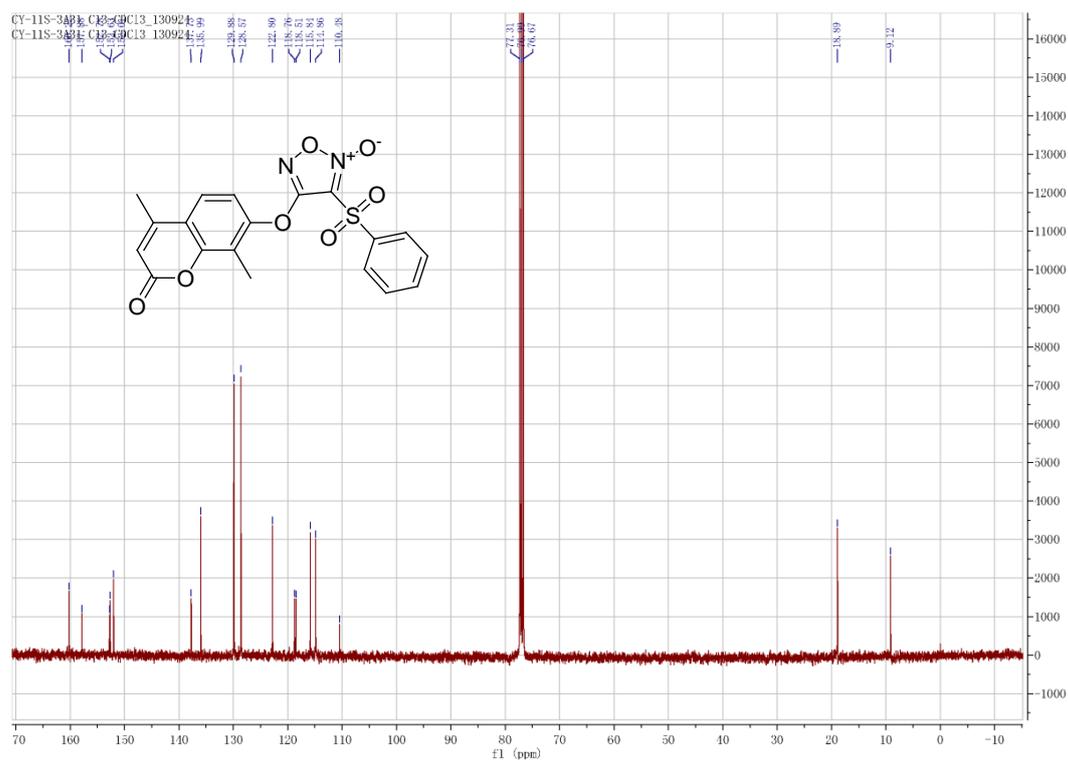
**Compound 6b:  $^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )**



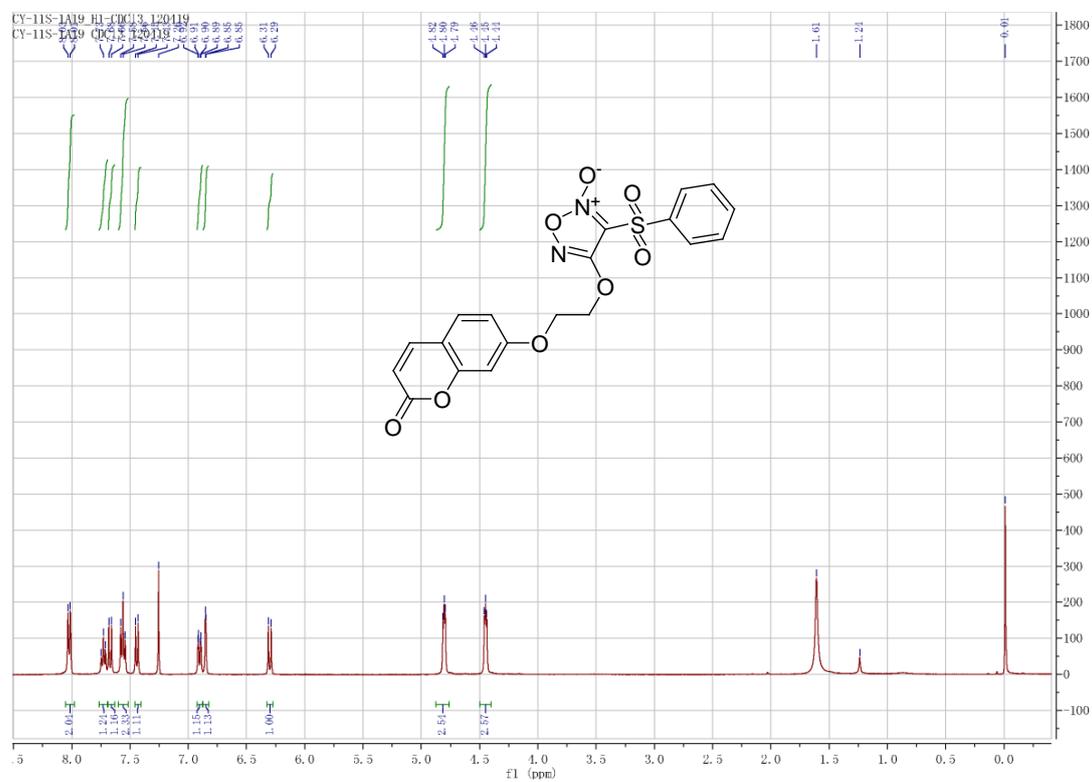
**Compound 6c:  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )**



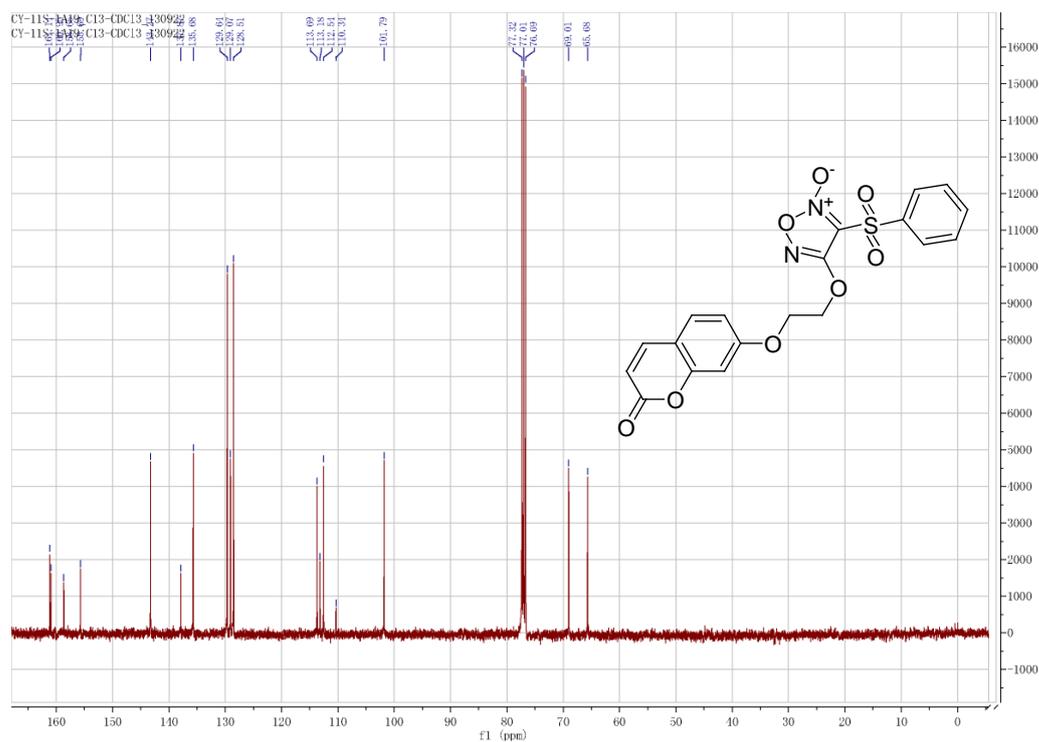
**Compound 6c:  $^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )**



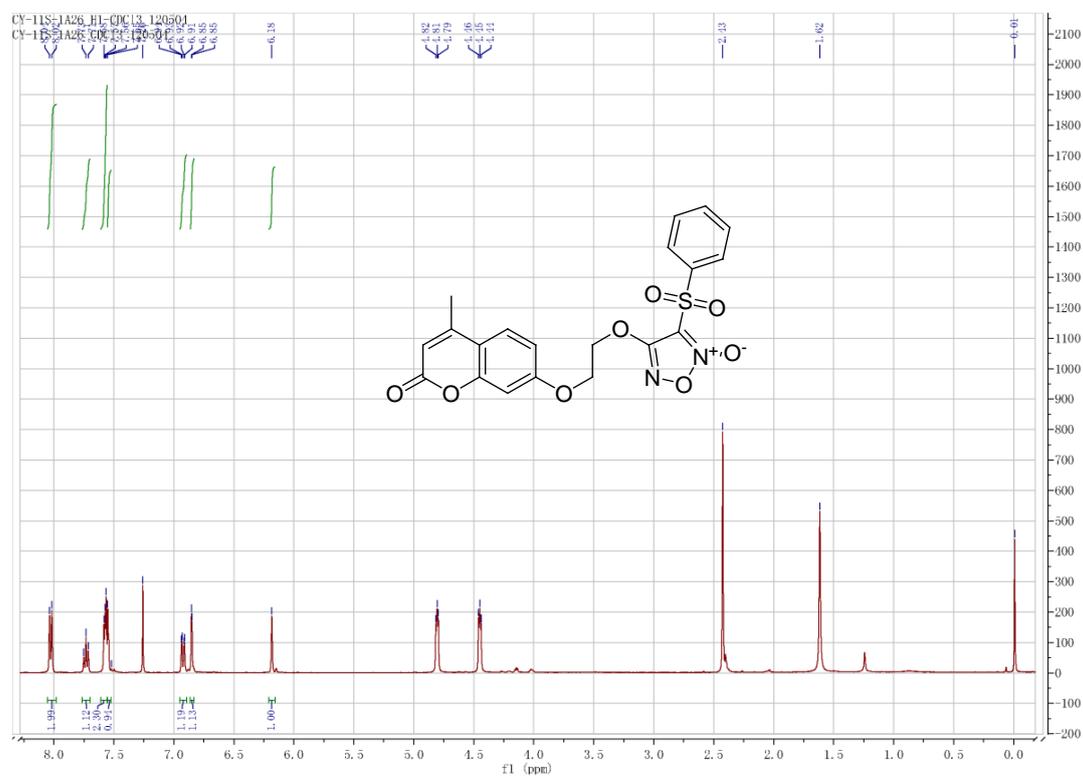
**Compound 8a:  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )**



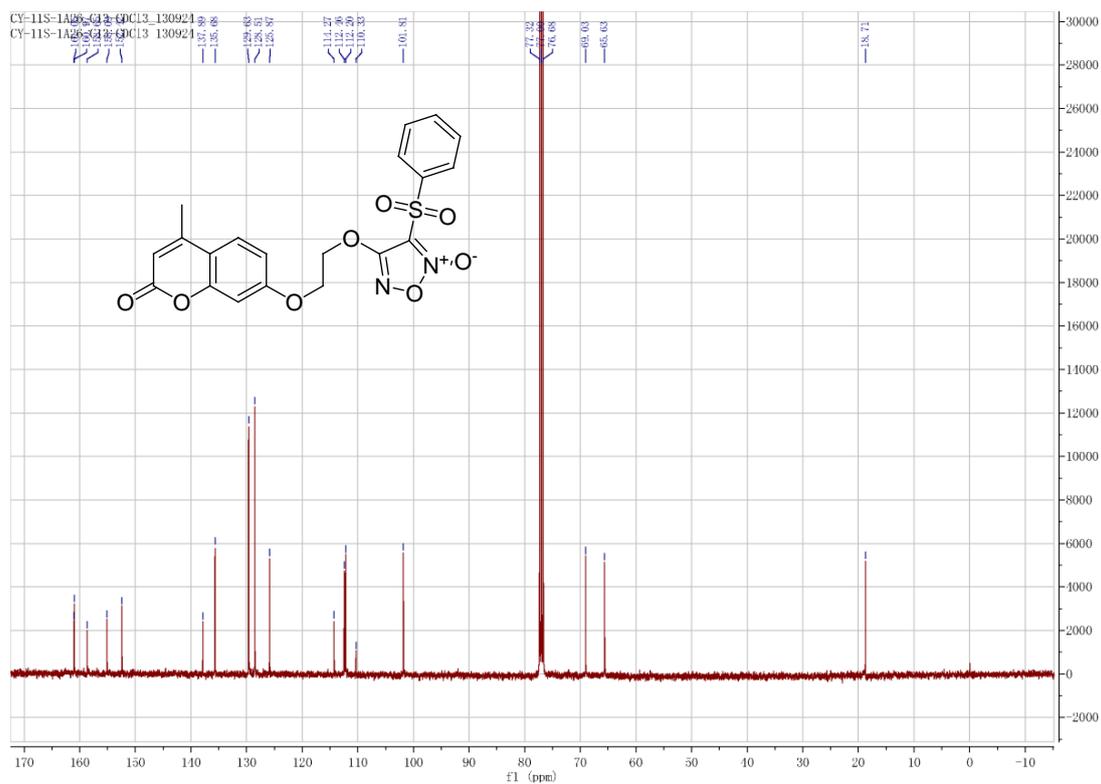
**Compound 8a:  $^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )**



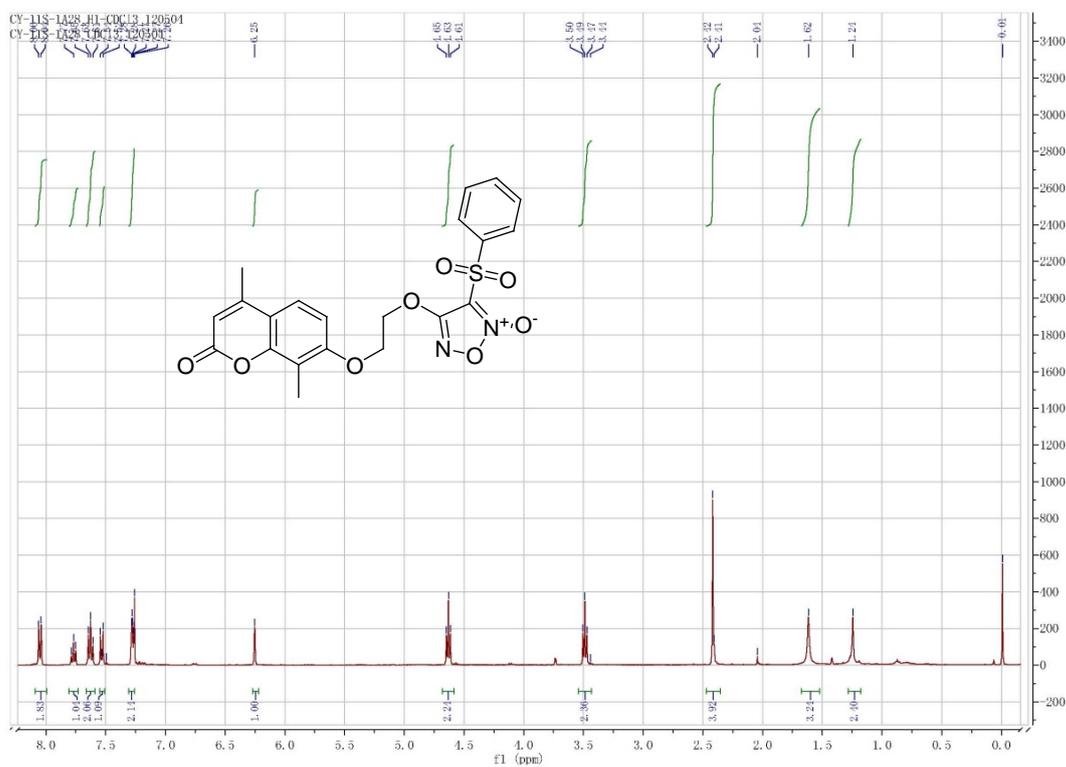
**Compound 8b:  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )**



**Compound 8b:  $^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )**

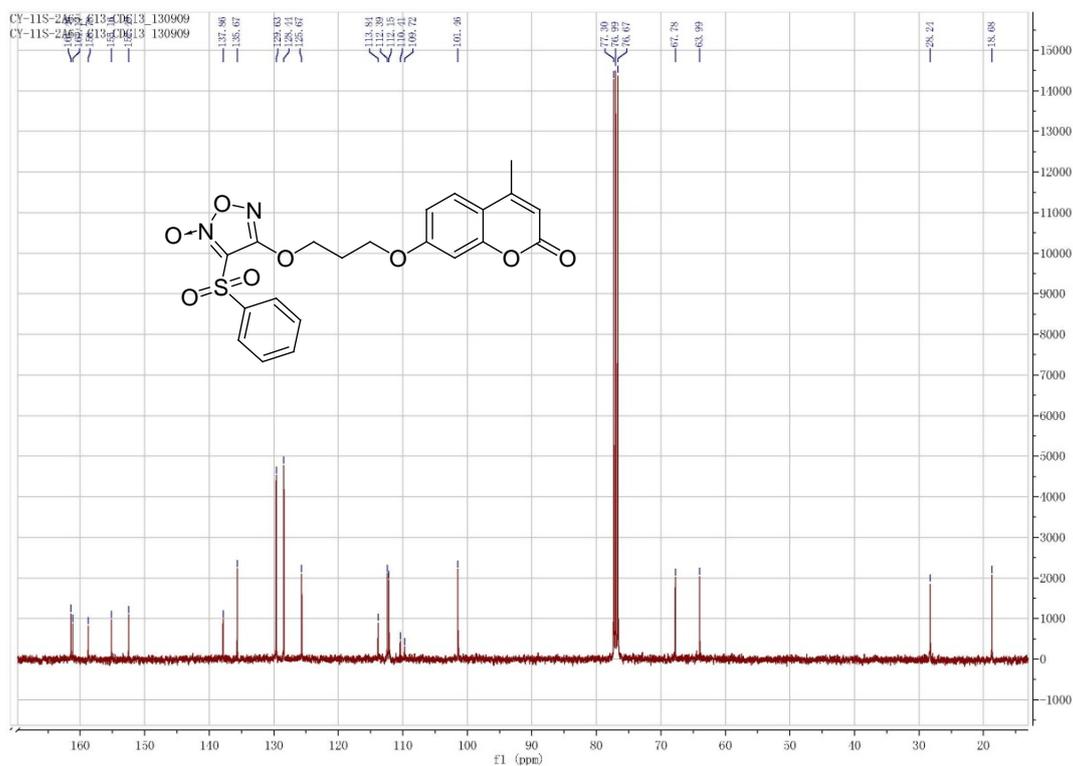


**Compound 8c:  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )**

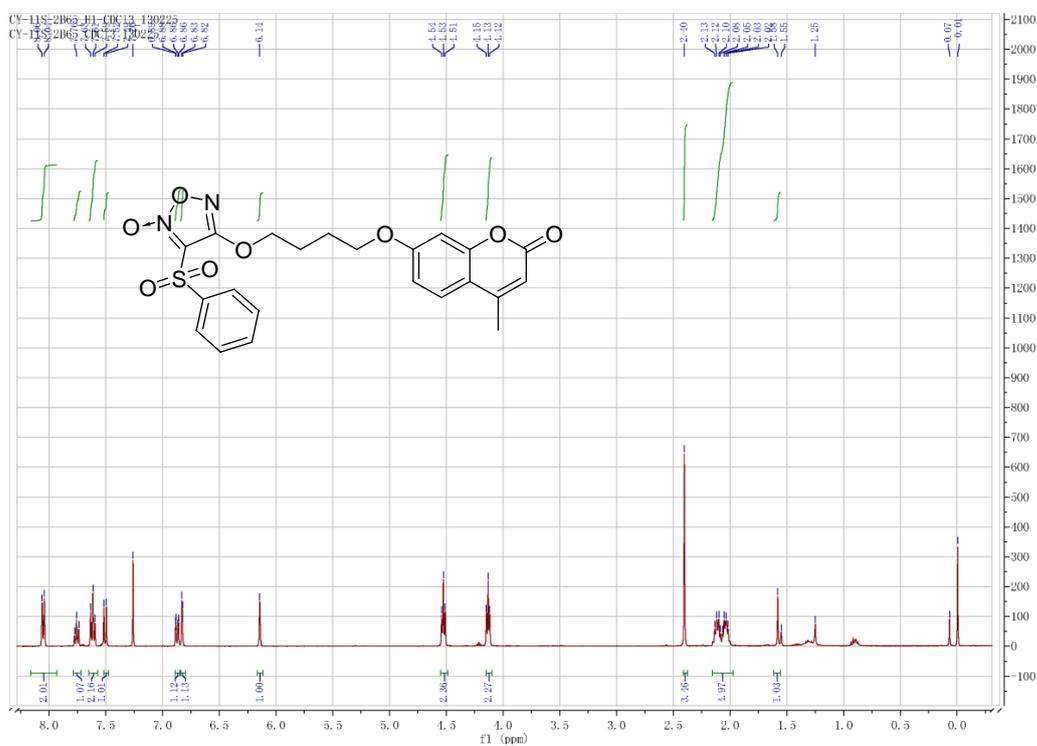




**Compound 8d:  $^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )**



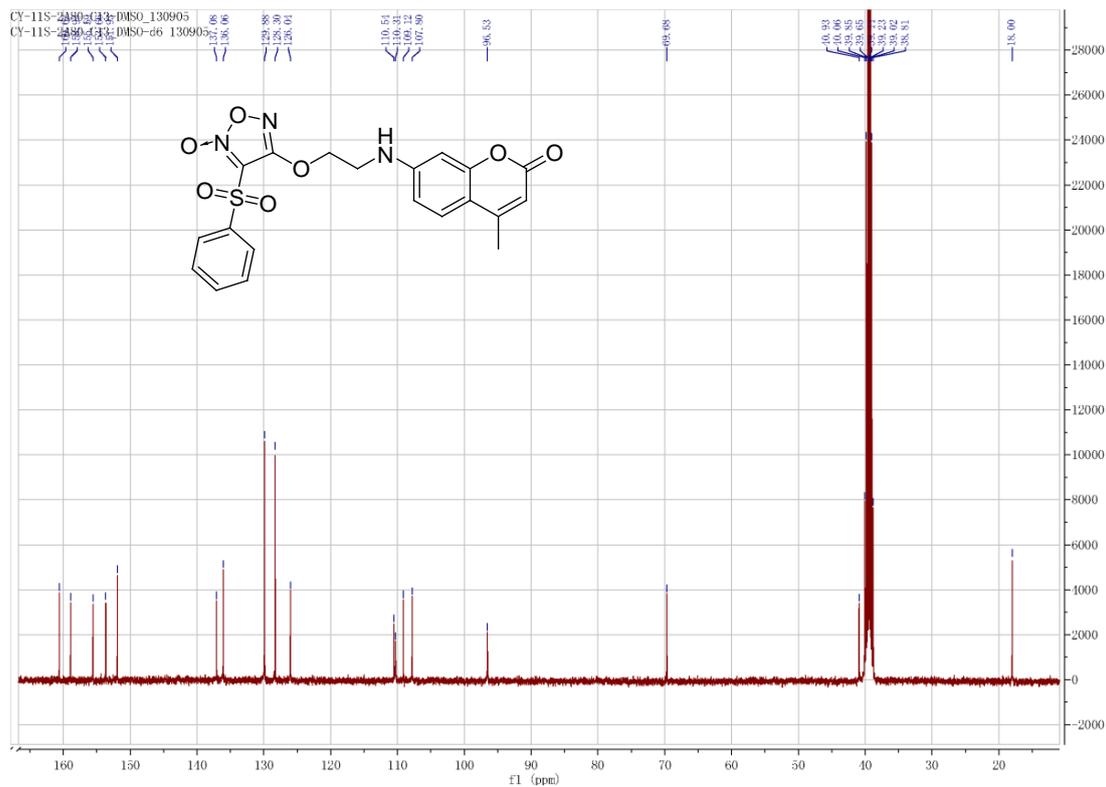
**Compound 8e:  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )**



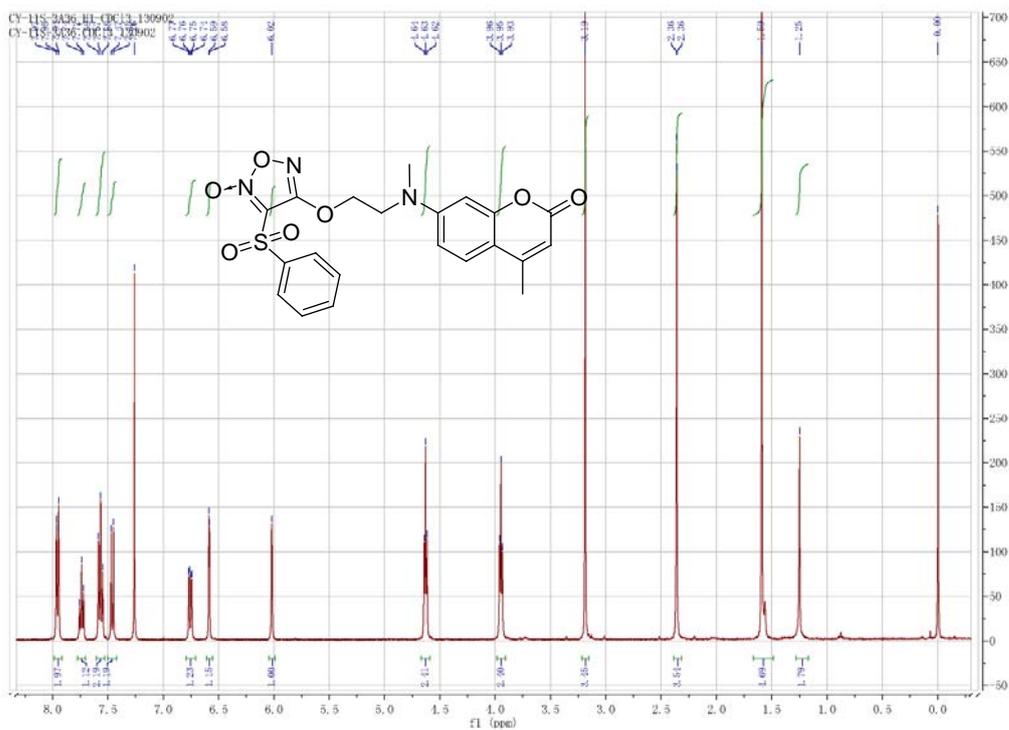




**Compound 8g:  $^{13}\text{C}$  NMR (101 MHz, DMSO- $d_6$ )**

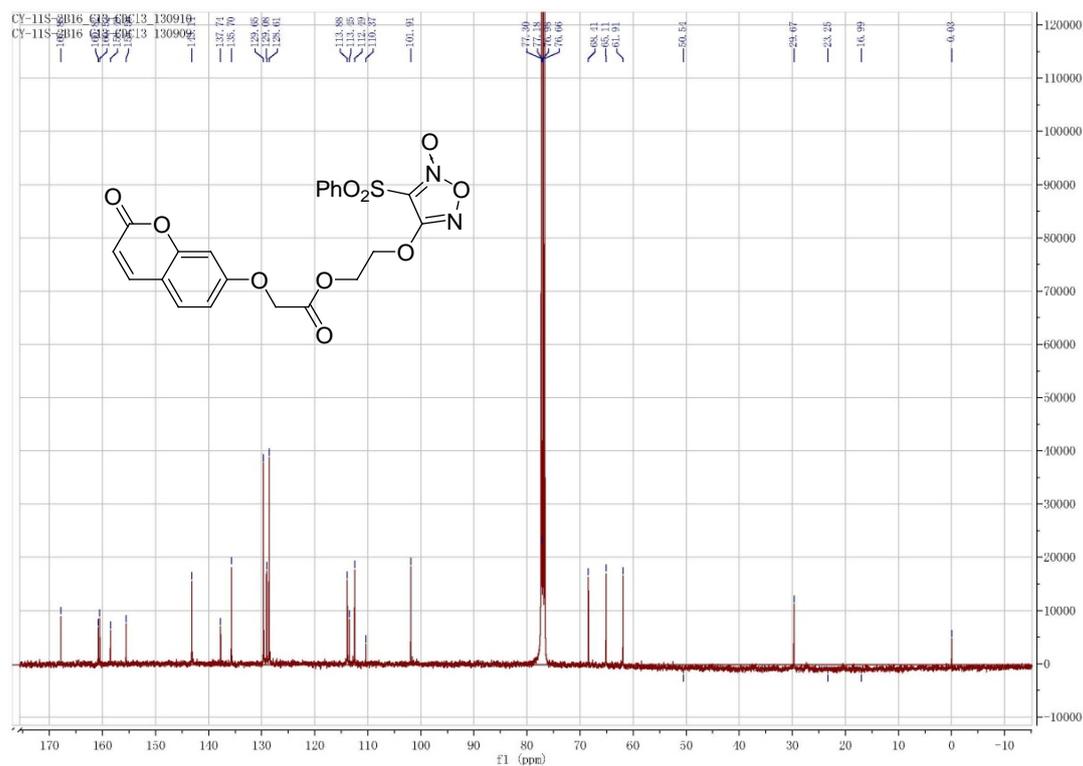


**Compound 10a:  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )**

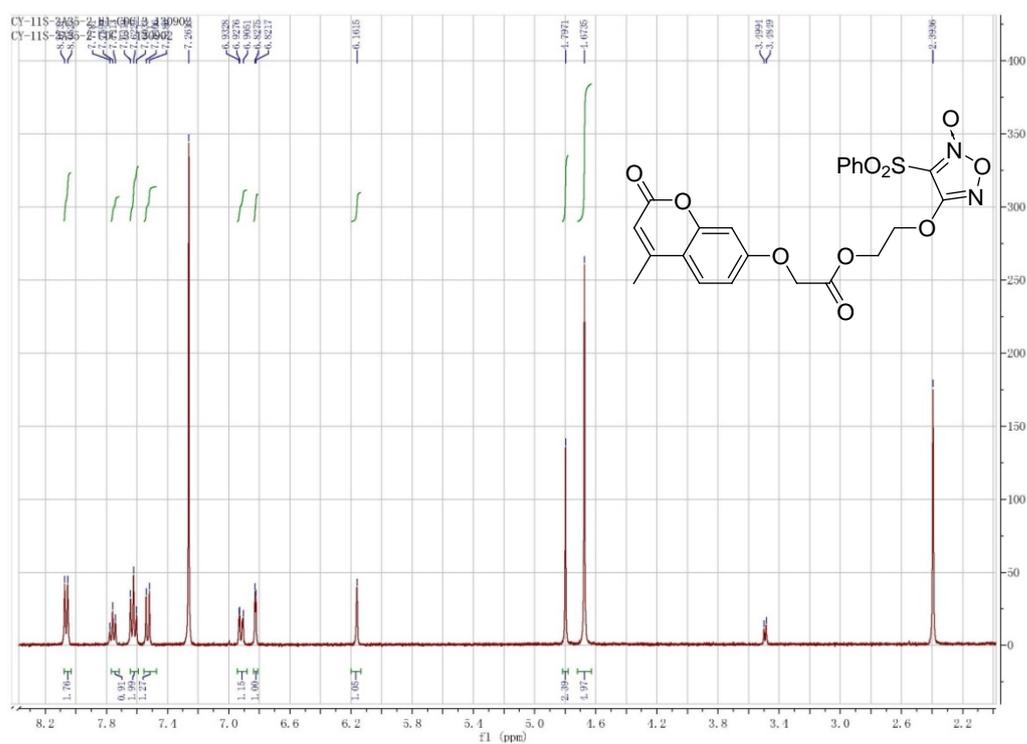




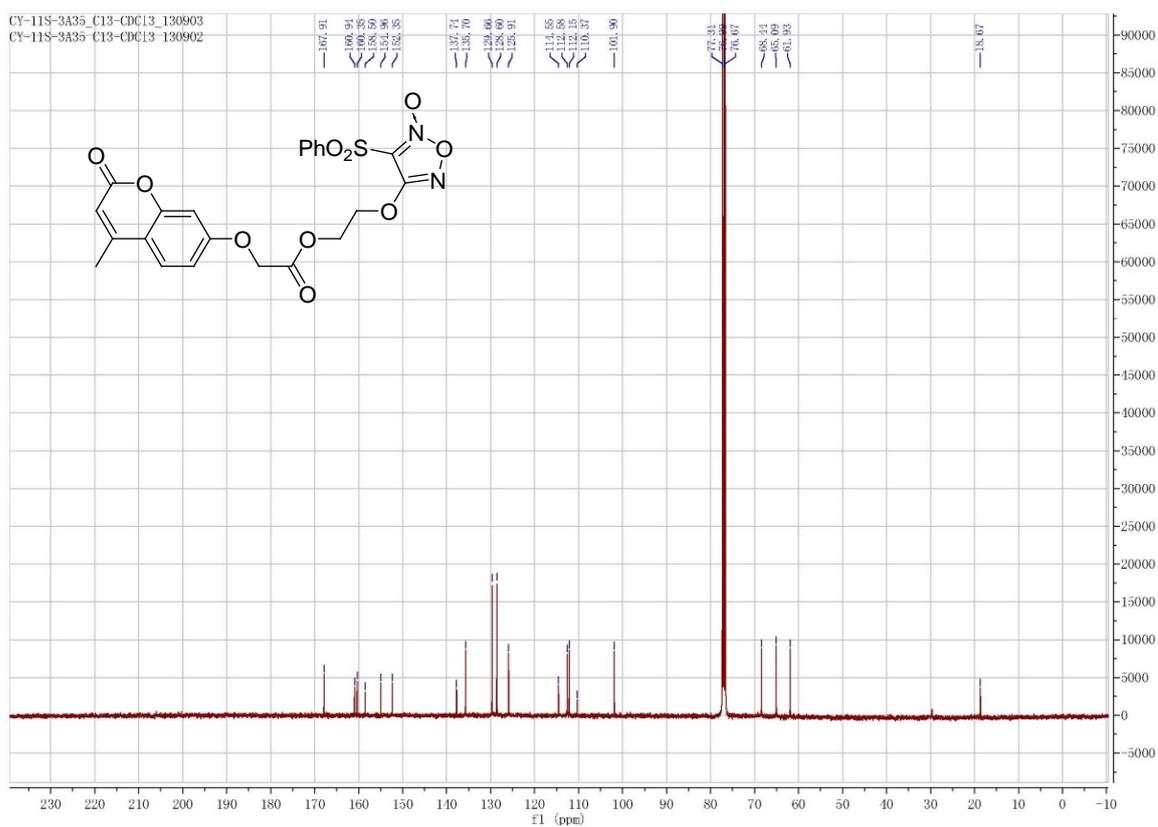
Compound 13a:  $^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )



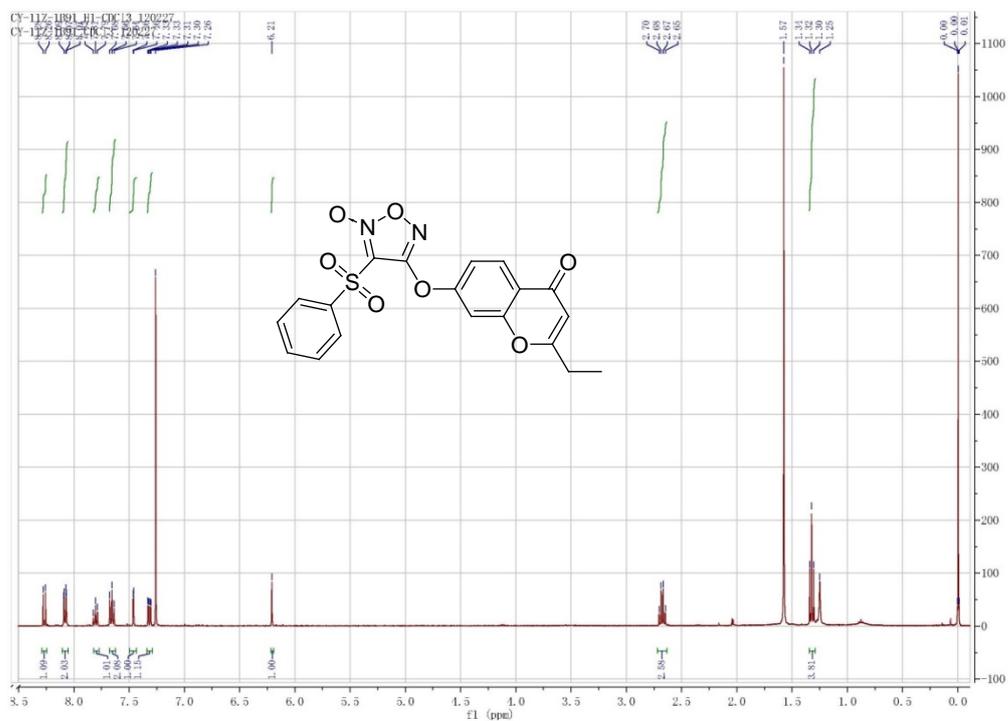
Compound 13b:  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )



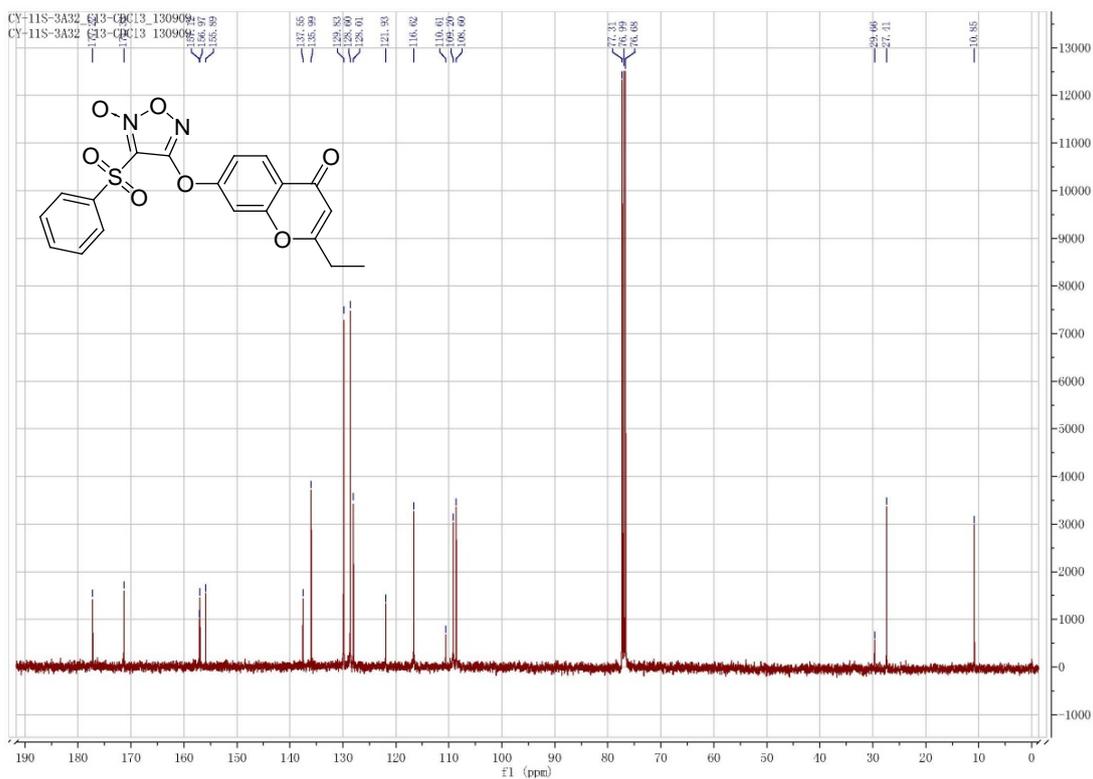
**Compound 13b:  $^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )**



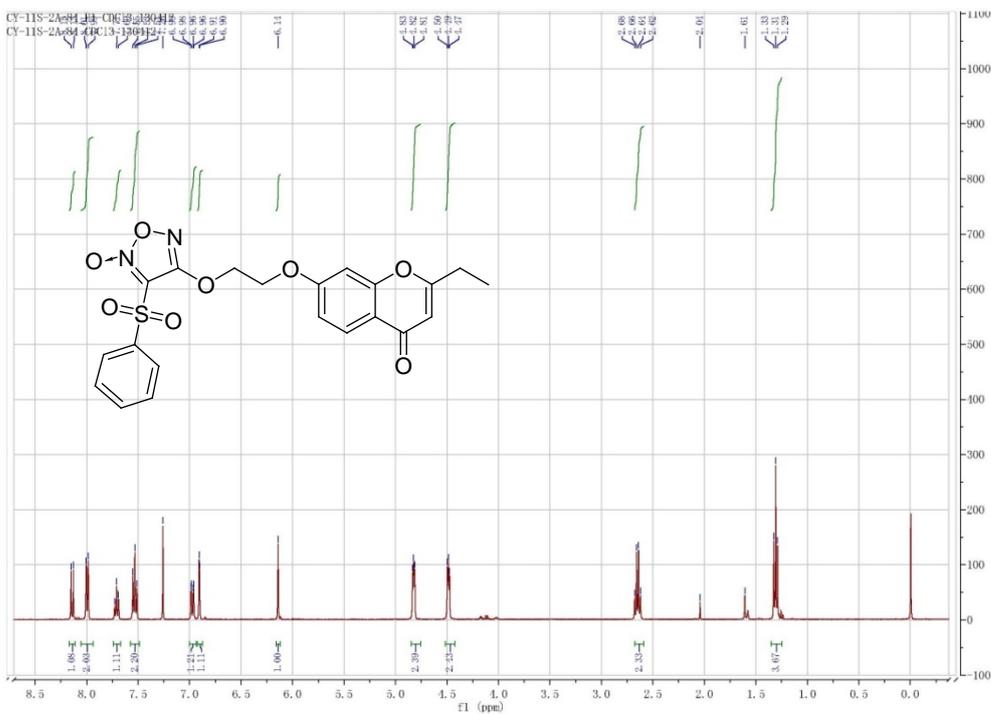
**Compound 15:  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )**



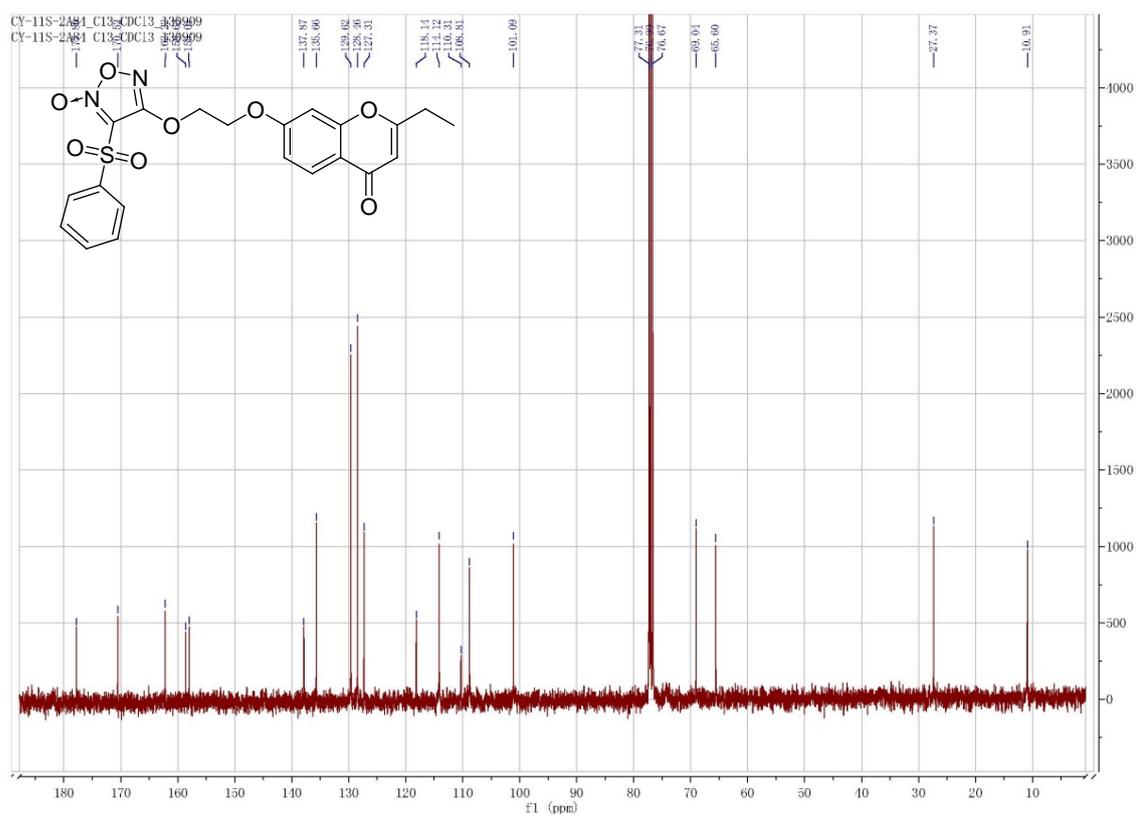
**Compound 15:  $^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )**



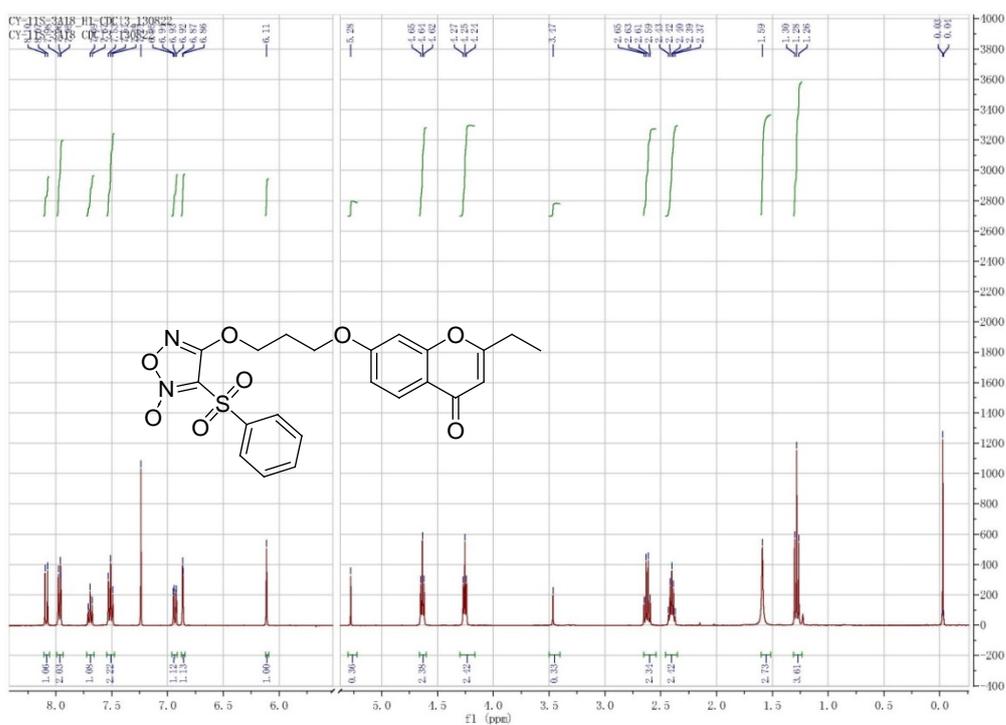
**Compound 17a:  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )**



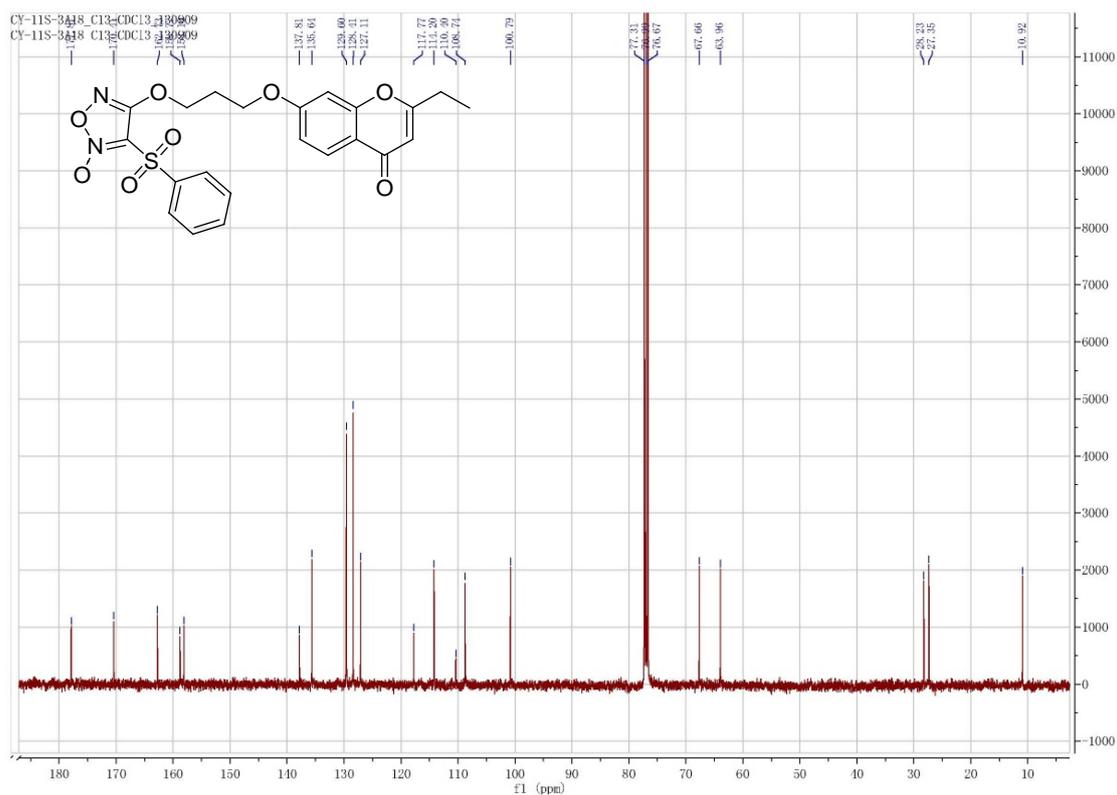
**Compound 17a:  $^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )**



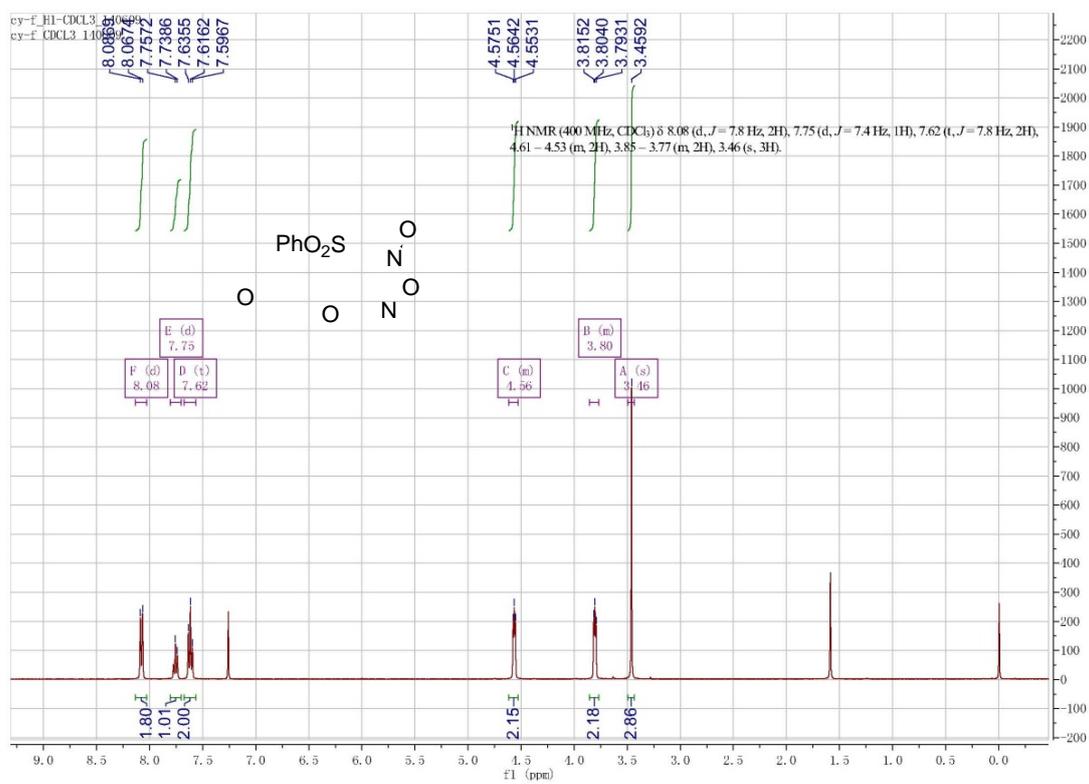
**Compound 17b:  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )**



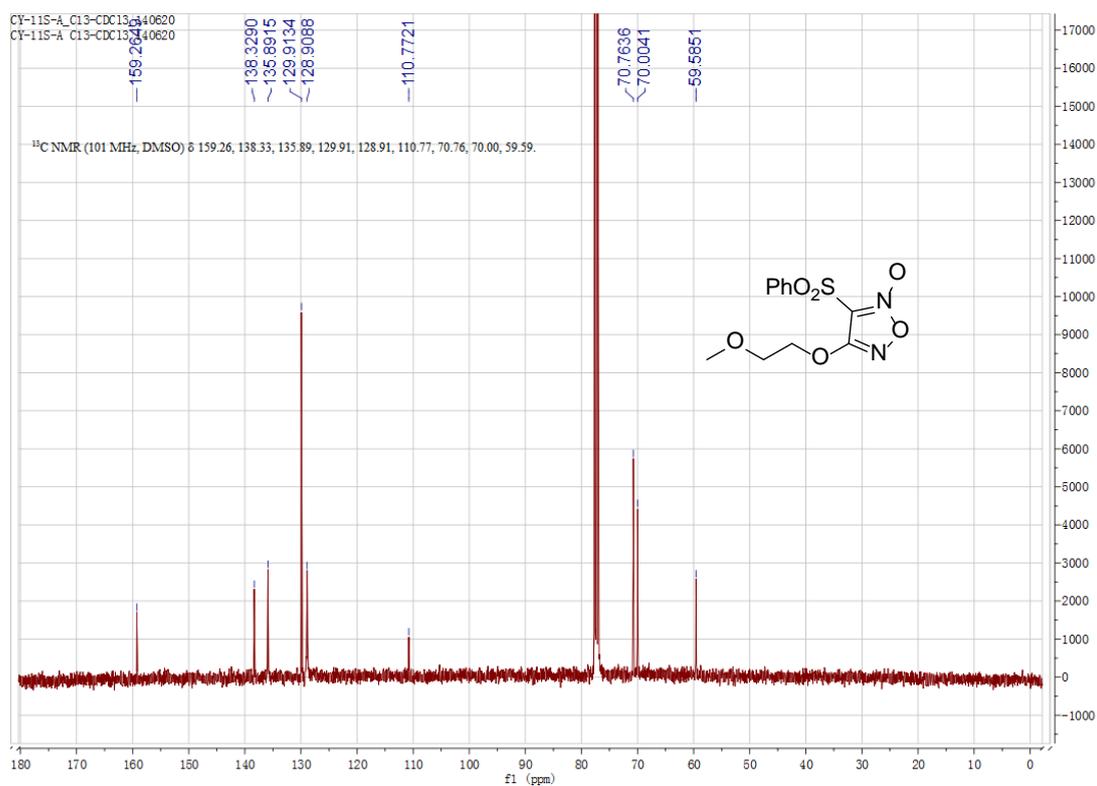
**Compound 17b:  $^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )**



**Compound 18:  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )**



**Compound 18:  $^{13}\text{C}$  NMR (101 MHz,  $\text{CDCl}_3$ )**



**REFERENCE**

1. Heald, R. A.; Jackson, P.; Savy, P.; Jones, M.; Gancia, E.; Burton, B.; Newman, R.; Boggs, J.; Chan, E.; Chan, J.; Choo, E.; Merchant, M.; Rudewicz, P.; Ultsch, M.; Wiesmann, C.; Yue, Q.; Belvin, M.; Price, S. Discovery of Novel Allosteric Mitogen-Activated Protein Kinase Kinase (MEK) 1,2 Inhibitors Possessing Bidentate Ser212 Interactions. *J. Med. Chem.* **2012**, *55*, 4594–4604.