

First-In-Class Inhibitors Targeting the Interaction between Bacterial RNA Polymerase and Sigma Initiation Factor Affect the Viability and Toxin Release of *Streptococcus pneumoniae*

Jiqing Ye ^{1,†} Adrian Jun Chu ^{2,†} Lin Lin ² Xiao Yang ^{2,*} and Cong Ma ^{1,*}

¹ State Key Laboratory of Chemical Biology and Drug Discovery, Department of Applied Biology and Chemical Technology, The Hong Kong Polytechnic University, Kowloon, Hong Kong

² Department of Microbiology, The Chinese University of Hong Kong, Prince of Wales Hospital, Shatin, Hong Kong

* Correspondence: cong.ma@polyu.edu.hk (C.M.); xiaoyang@cuhk.edu.hk (X.Y.)

† These authors contributed equally to this work.

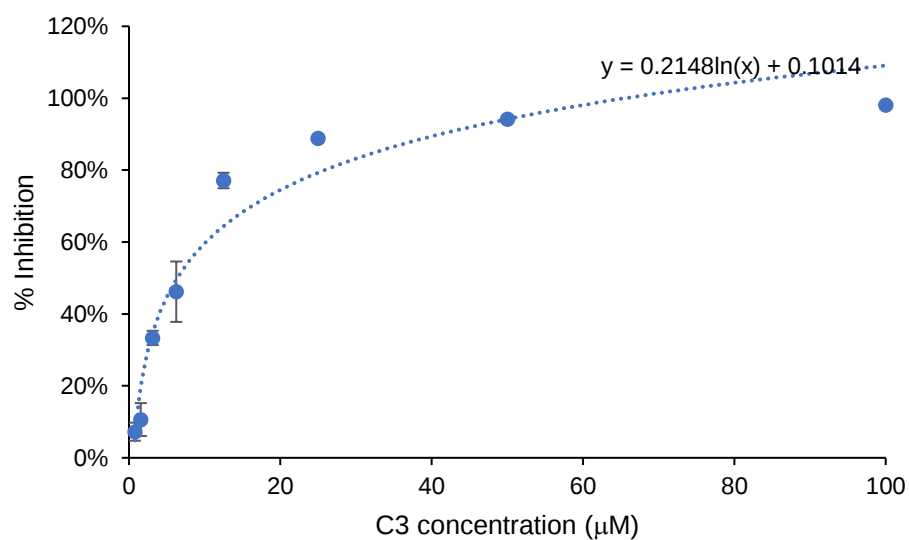


Figure 1. IC₅₀ measurement of C3 against the protein-protein interaction between RNAP CH-σ at 0.05 μM.

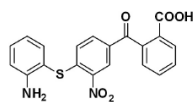
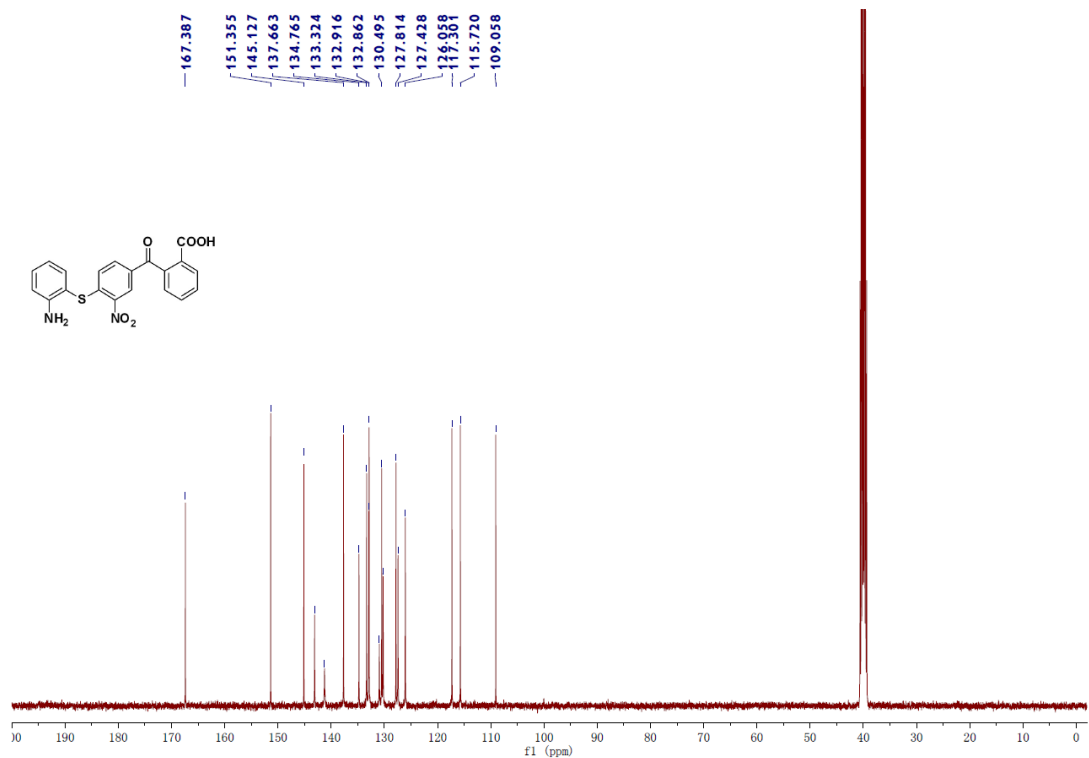
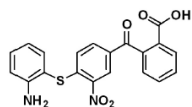
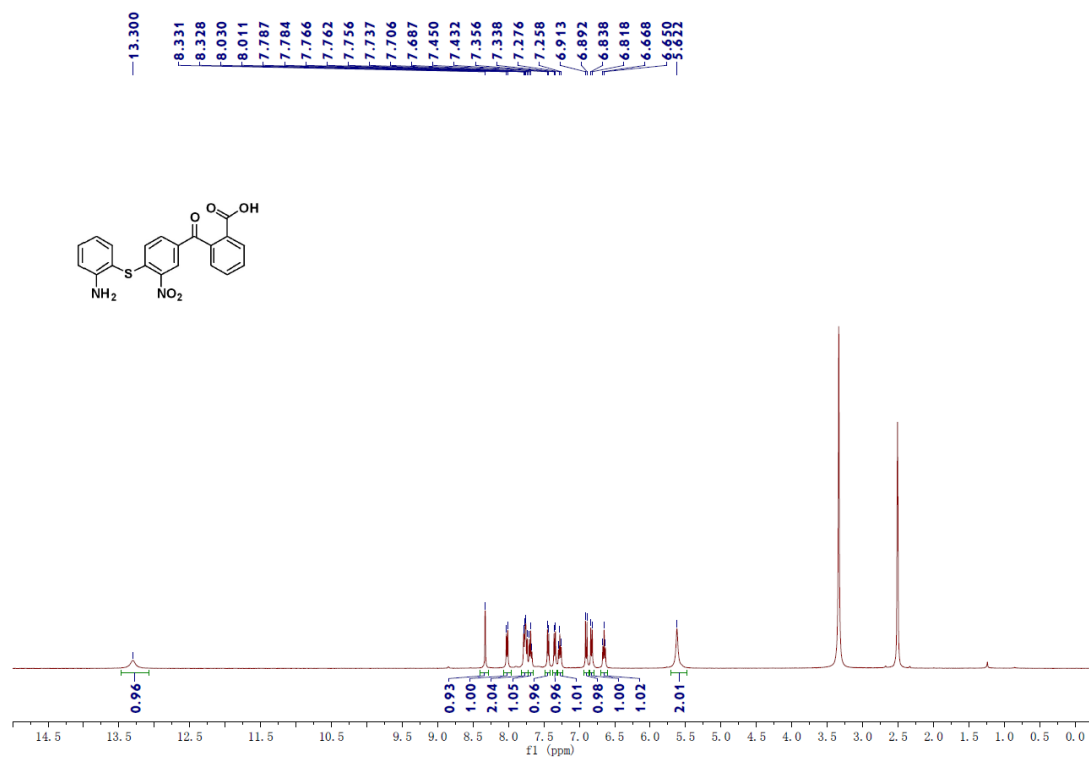
Table S1. The luminescence data of C3 inhibiting the RNAP CH – σ protein-protein interaction.

Triplicate A	Triplicate B	Triplicate C	Average	% Inhibition	% SE ^a	C3 concentration (μM)
25819	33863	27577	29086	100%	8.39%	0
632	614	434	560	98.07%	0.22%	100
1848	1776	1422	1682	94.22%	0.45%	50
3074	3836	2850	3253	88.81%	1.03%	25
6800	7662	5510	6657	77.11%	2.15%	12.5
14861	20218	11868	15649	46.20%	8.40%	6.25
18575	20504	19085	19388	33.34%	1.98%	3.125
27429	27207	23336	25991	10.64%	4.57%	1.5625
25793	28308	26815	26972	7.27%	2.51%	0.78125

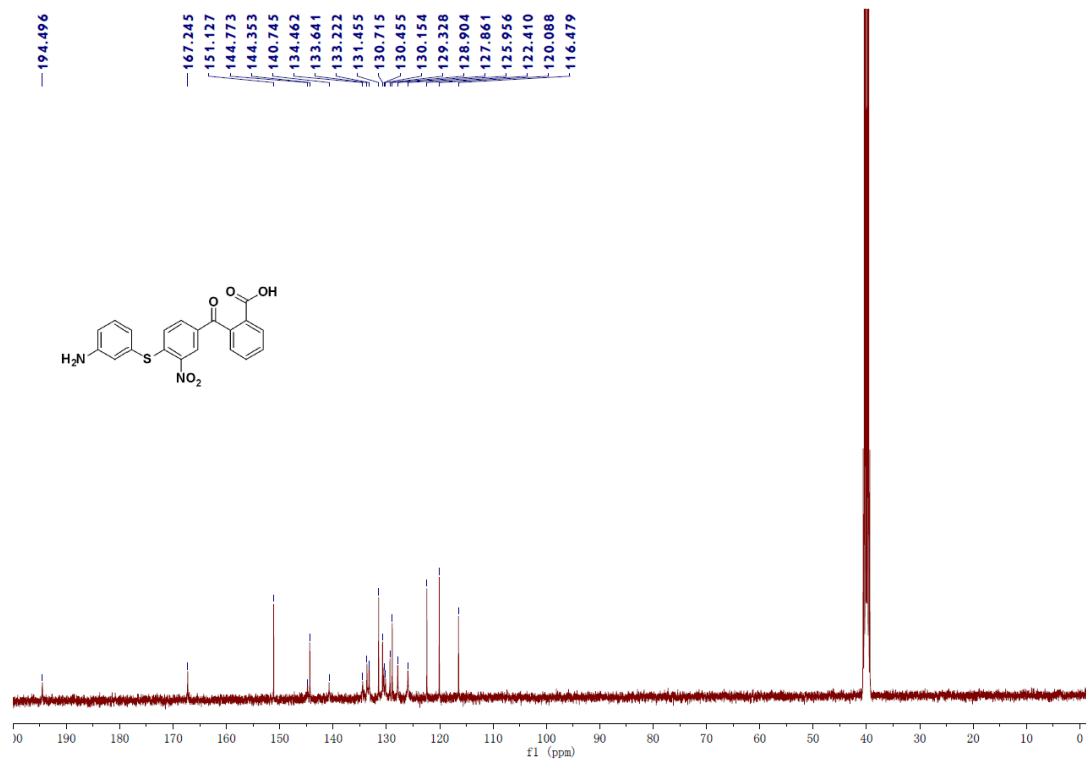
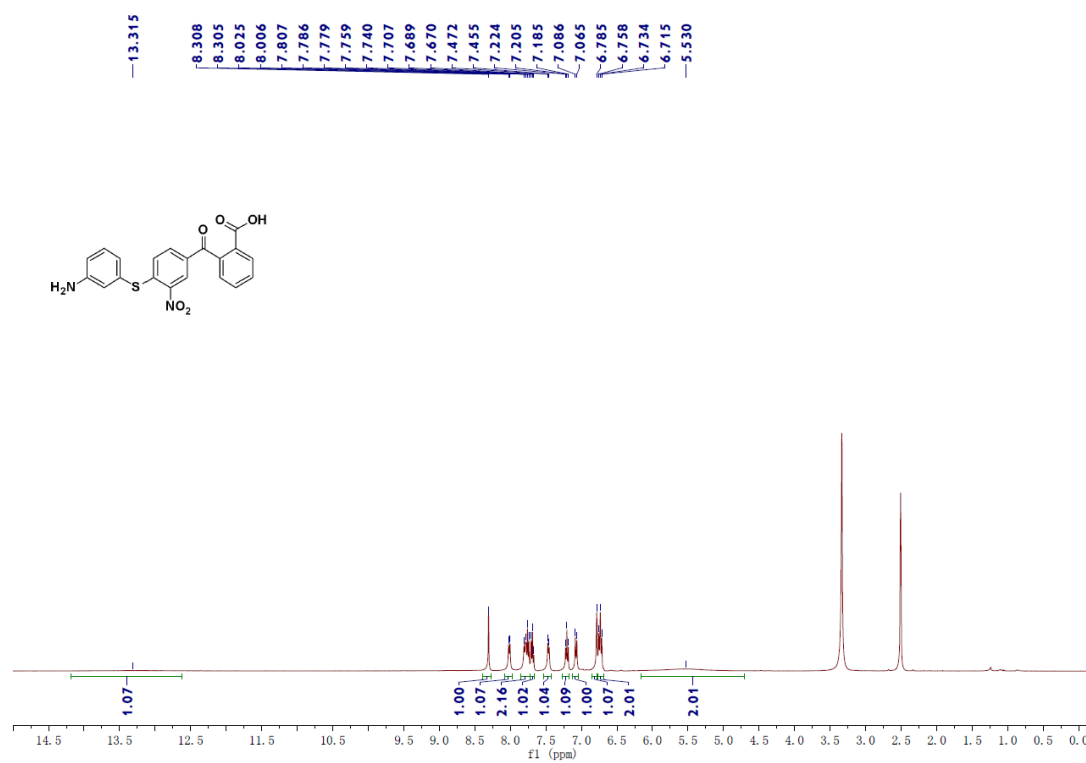
^a Standard error

¹H NMR and ¹³C NMR spectra.

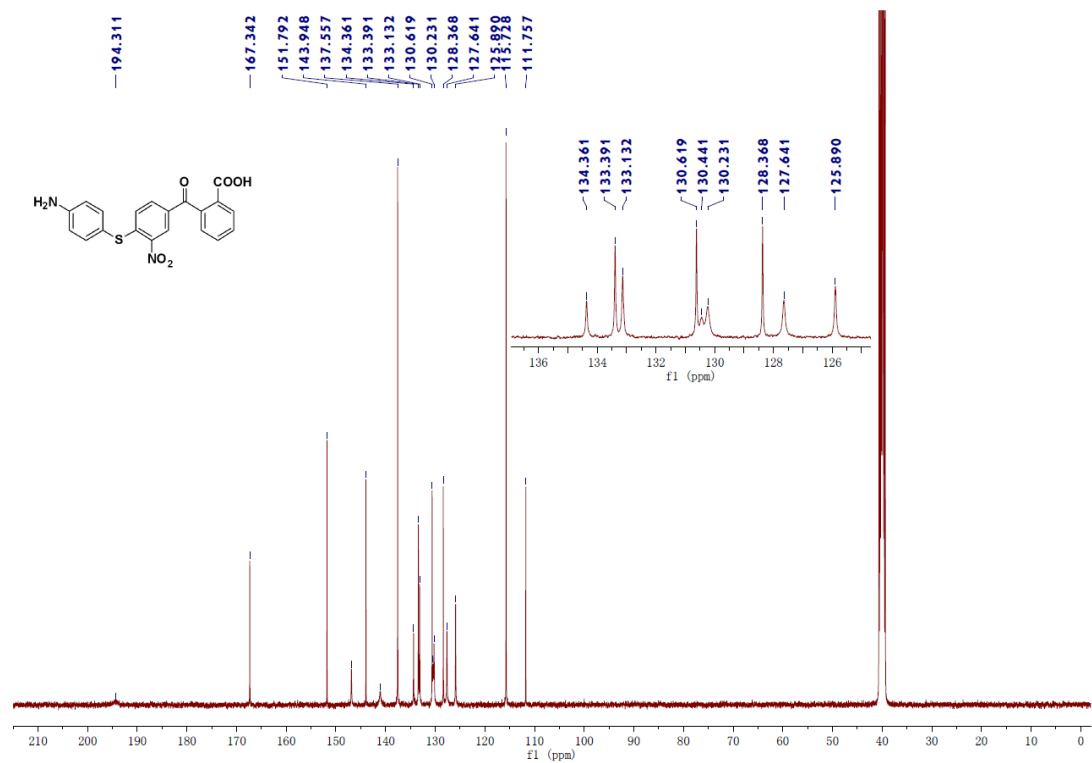
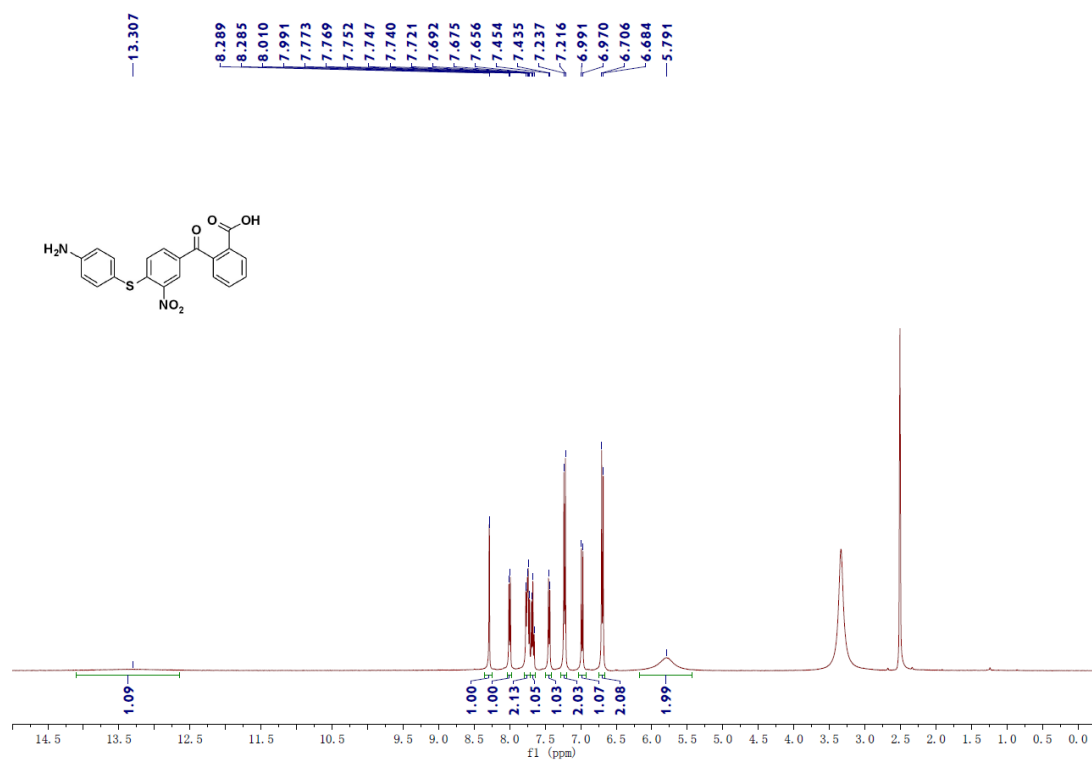
C3-001



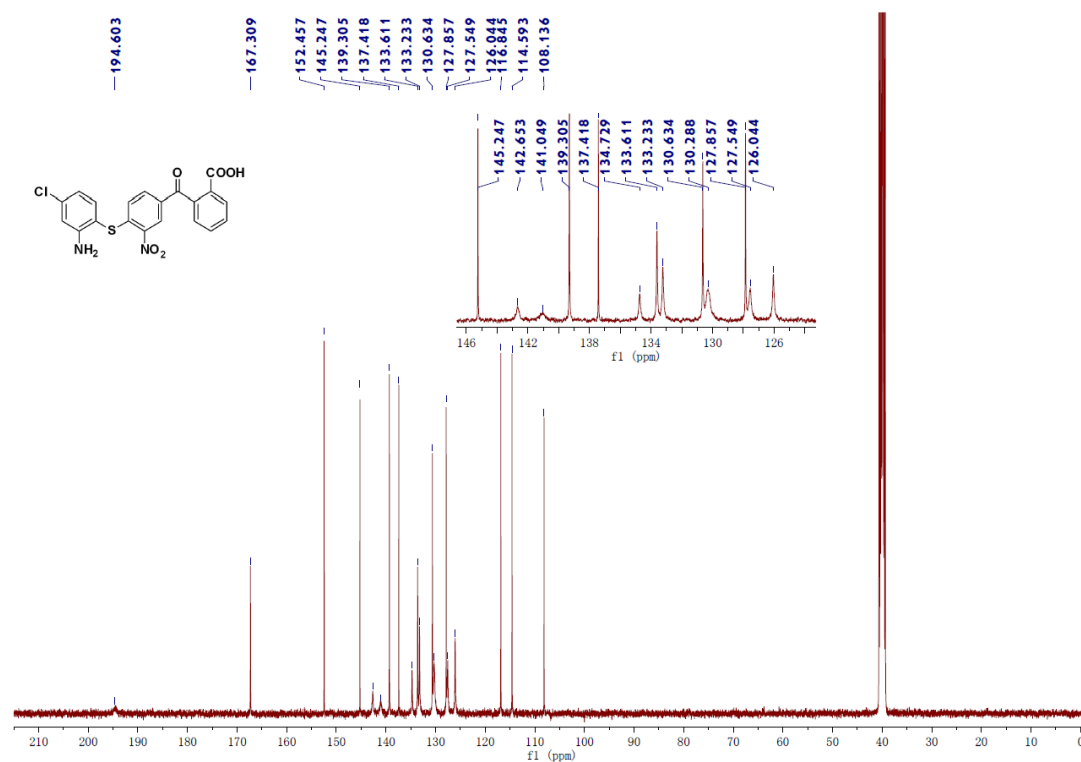
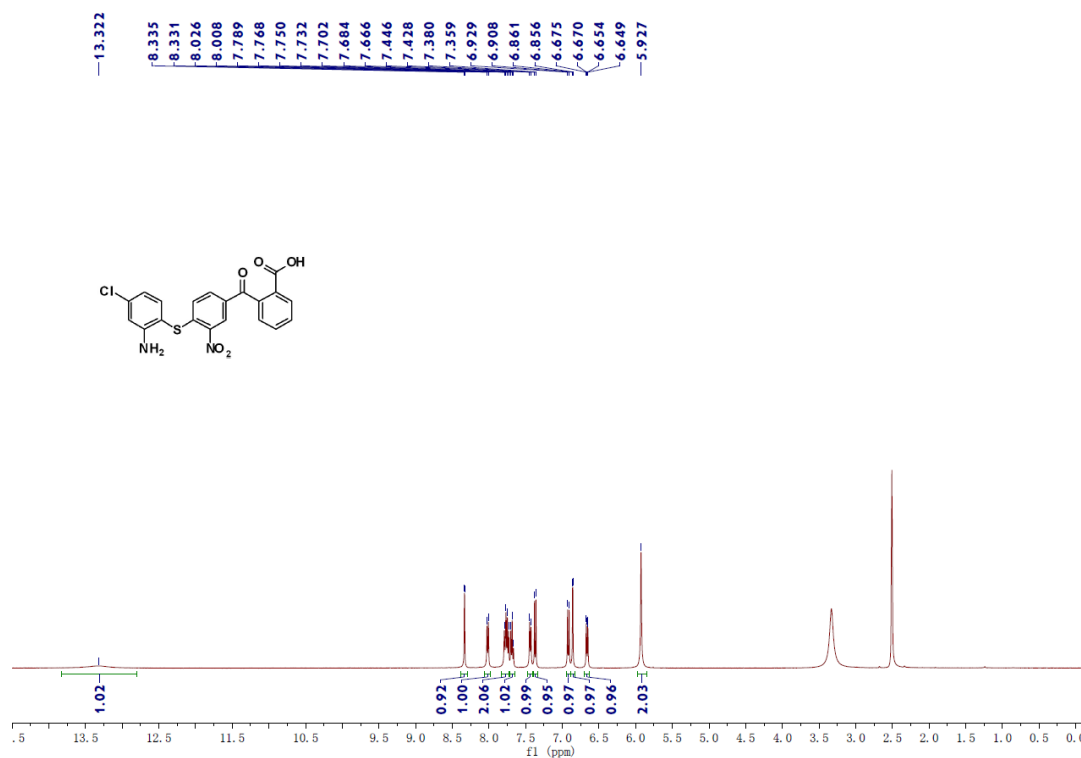
C3-002



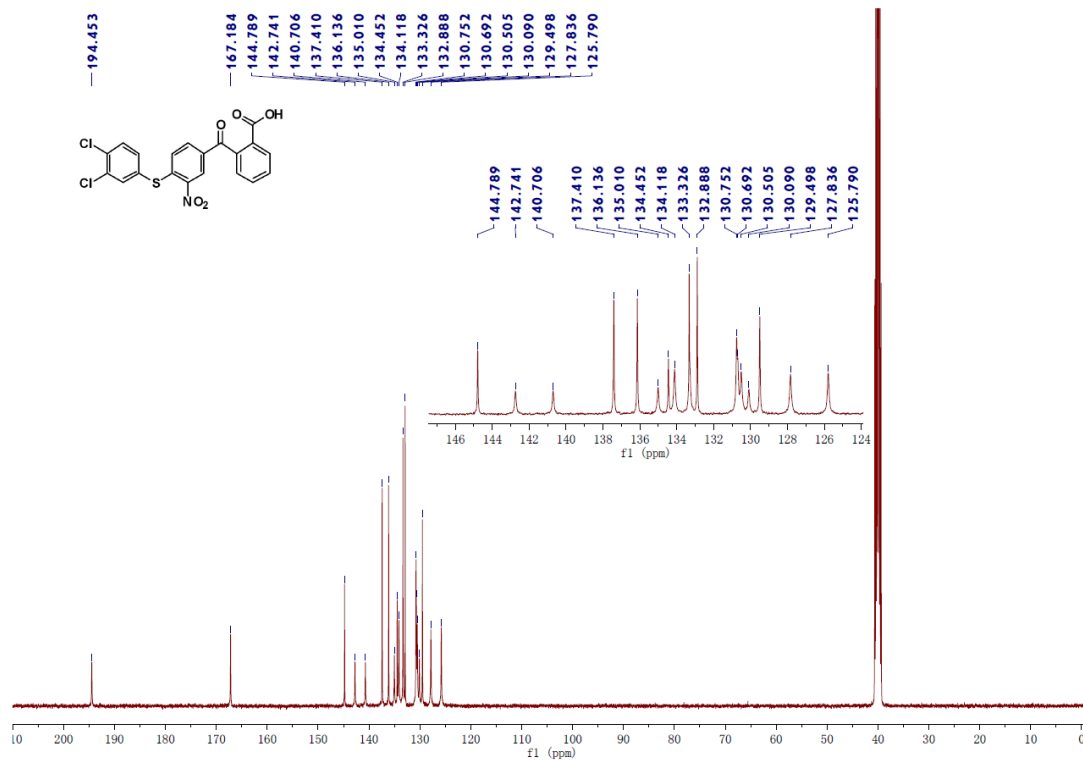
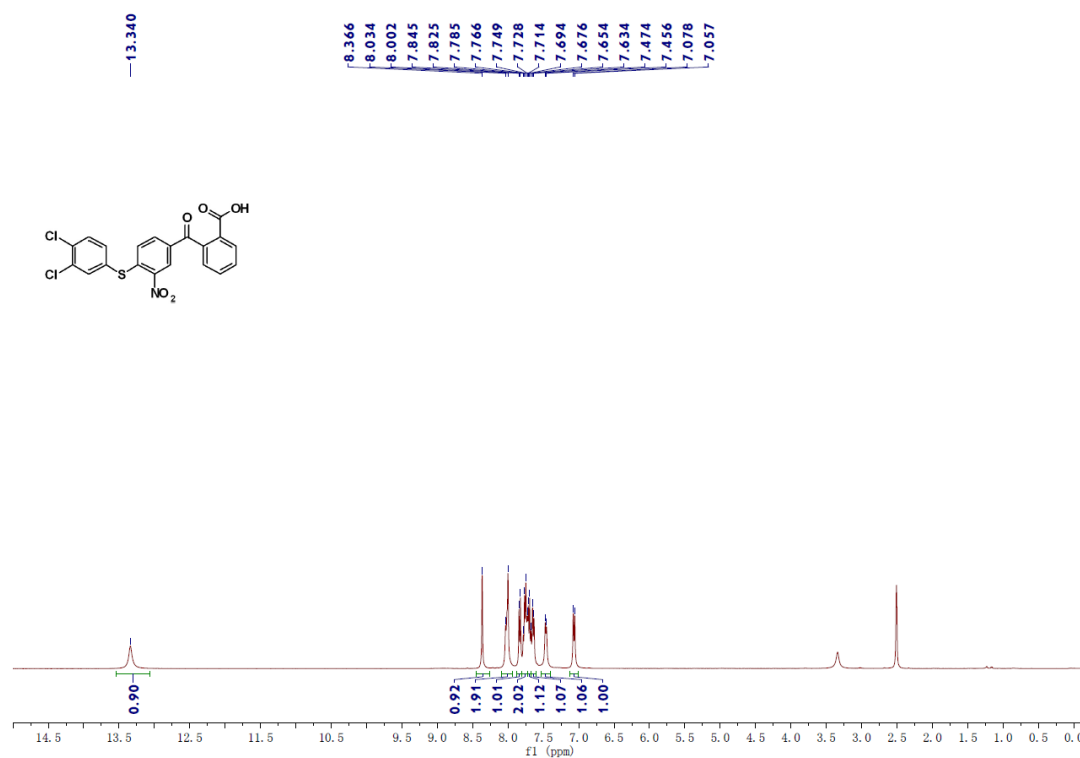
C3-003



C3-004



C3-005



HPLC spectra**HPLC analysis of final compounds**

System: Agilent 1260 Infinity

Mobile phase: Mobile phase A: MeCN; mobile phase B: H₂O

Detector: MWD at 254 nm

Column: Agilent ZORBAX Eclipse plus C18 (4.6 × 100 mm, 5 μm)

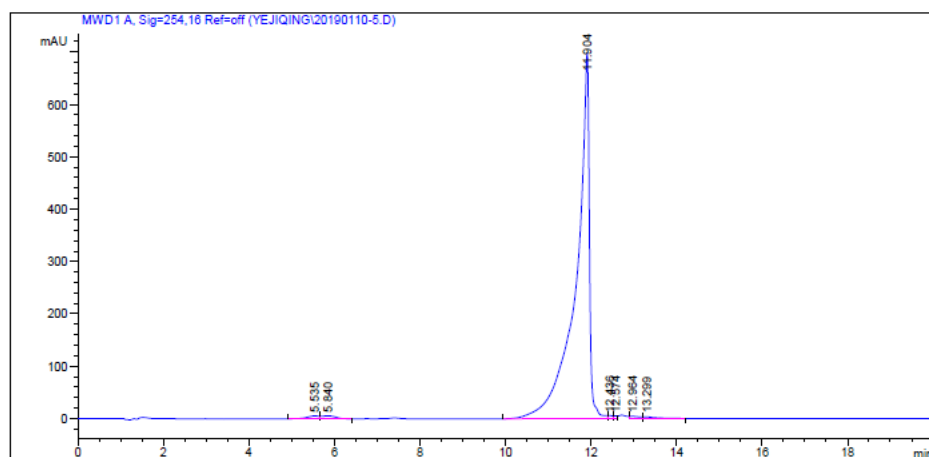
Flow rate: 1.000 mL/min

Using gradients elution.

C3-001

Data File C:\CHEM32\1\DATA\YEJIQING\20190110-5.D
Sample Name: 20190110-5

```
=====
Acq. Operator   : QYY
Acq. Instrument : Instrument 1          Location : Vial 3
Injection Date  : 1/10/2019 5:45:01 PM
Acq. Method     : C:\CHEM32\1\METHODS\YEJIQING.M
Last changed    : 1/10/2019 5:44:11 PM by QYY
Analysis Method : C:\CHEM32\1\METHODS\WU HAICUI.M
Last changed    : 1/11/2019 8:19:24 PM by yejiqing
                  (modified after loading)
Sample Info     : C3-18-001
=====
```



Area Percent Report

```
=====
Sorted By      : Signal
Multiplier     : 1.0000
Dilution       : 1.0000
Sample Amount  : 1.00000 [ng/ul] (not used in calc.)
Use Multiplier & Dilution Factor with ISTDs
=====
```

Signal 1: MWD1 A, Sig=254,16 Ref=off

Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
1	5.535	BV	0.2844	114.39557	5.89585	0.7429
2	5.840	VB	0.3480	131.97195	5.87640	0.8570

Instrument 1 1/11/2019 8:19:41 PM yejiqing

Page 1 of 2

Data File C:\CHEM32\1\DATA\YEJIQING\20190110-5.D
Sample Name: 20190110-5

Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
3	11.904	BV	0.2759	1.49381e4	697.45142	97.0034
4	12.436	VV	0.1152	46.18891	5.65991	0.2999
5	12.574	VV	0.0730	27.20704	5.20154	0.1767
6	12.964	VV	0.2077	73.63541	4.62694	0.4782
7	13.299	VB	0.2561	68.06255	3.39128	0.4420

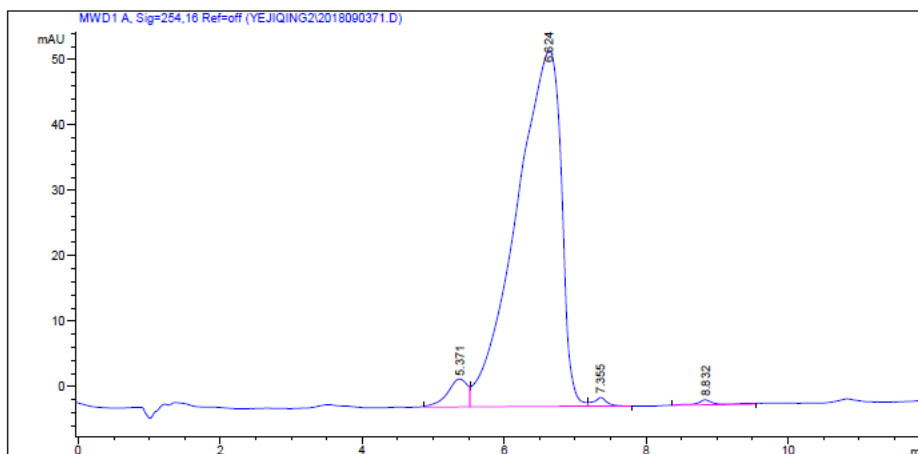
Totals : 1.53995e4 728.10334

*** End of Report ***

C3-002

DATA File C:\CHEM32\1\DATA\YEJIQING2\20180903\1.D
Sample Name: 2018090371

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=====
Acq. Operator   : YeJiqing
Acq. Instrument : Instrument 1          Location : Vial 1
Injection Date  : 9/6/2018 11:43:04 AM
Acq. Method     : C:\CHEM32\1\METHODS\YEJIQING.M
Last changed    : 9/6/2018 11:42:43 AM by YeJiqing
Analysis Method : C:\CHEM32\1\METHODS\YEJIQING.M
Last changed    : 12/12/2018 9:27:24 PM by SKY
                  (modified after loading)
Sample Info     : C3-18-031
=====
```



Area Percent Report

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=====
Sorted By      : Signal
Multiplier     : 1.0000
Dilution       : 1.0000
Sample Amount  : 1.00000 [ng/ul] (not used in calc.)
Use Multiplier & Dilution Factor with ISIDs
=====
```

Signal 1: MWD1 A, Sig=254.16 Ref=off

Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
1	5.371	BV	0.2961	85.72115	4.30946	3.4111
2	6.624	VV	0.6259	2393.43896	54.33510	95.2416

Instrument 1 12/12/2018 9:27:30 PM SKY

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Data File C:\CHEM32\1\DATA\YEJIQING2\2018090371.D
Sample Name: 2018090371

Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
3	7.355	VB	0.1956	17.92930	1.34427	0.7135
4	8.832	BB	0.2822	15.92837	7.93498e-1	0.6338

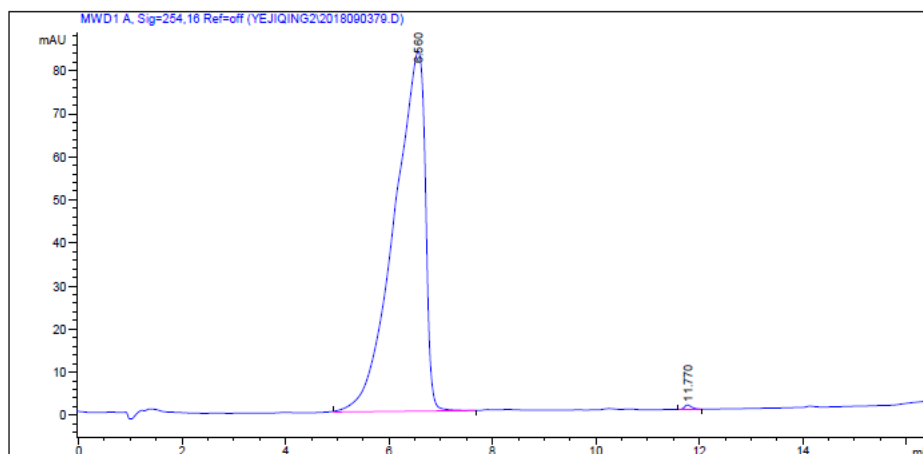
Totals : 2513.01778 60.78234

*** End of Report ***

C3-003

Data File C:\CHEM32\1\DATA\YEJING2\2018090379.D
Sample Name: 2018090379

```
=====
Acq. Operator   : YeJiqing
Acq. Instrument : Instrument 1          Location : Vial 1
Injection Date  : 9/6/2018 3:58:52 PM
Acq. Method     : C:\CHEM32\1\METHODS\YEJING.M
Last changed    : 9/6/2018 3:56:38 PM by YeJiqing
Analysis Method : C:\CHEM32\1\METHODS\YEJING.M
Last changed    : 12/12/2018 9:39:33 PM by SXY
                  (modified after loading)
Sample Info     : C3-18-032
=====
```



Area Percent Report

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=====
Sorted By      : Signal
Multiplier     : 1.0000
Dilution       : 1.0000
Sample Amount  : 1.00000 [ng/ul] (not used in calc.)
Use Multiplier & Dilution Factor with ISTDs
=====
```

Signal 1: MWD1 A, Sig=254.16 Ref=off

Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
1	6.560	BB	0.5863	3484.63843	83.59244	99.7268
2	11.770	BB	0.1470	9.54592	1.00294	0.2732

Instrument 1 12/12/2018 9:40:00 PM SXY

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Data File C:\CHEM32\1\DATA\YEJING2\2018090379.D
Sample Name: 2018090379

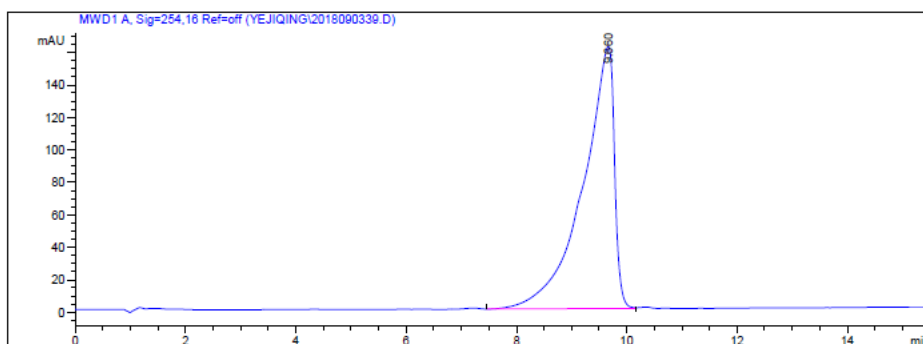
Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
----- ----- ----- ----- ----- -----						
Totals :				3494.18435	84.59538	

*** End of Report ***

C3-004

Data File C:\CHEM32\1\DATA\YEJQING\2018090339.D
Sample Name: 2018090339

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Acq. Operator   : YeJiqing
Acq. Instrument : Instrument 1          Location : Vial 1
Injection Date  : 9/4/2018 7:38:29 PM
Acq. Method     : C:\CHEM32\1\METHODS\YEJQING.M
Last changed    : 9/4/2018 7:36:05 PM by YeJiqing
Analysis Method : C:\CHEM32\1\METHODS\YEJQING.M
Last changed    : 9/4/2018 8:20:16 PM by YeJiqing
                  (modified after loading)
Sample Info     : C3-18-063
=====
```



Area Percent Report

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=====
Sorted By      : Signal
Multiplier     : 1.0000
Dilution       : 1.0000
Sample Amount  : 1.00000 [ng/ul] (not used in calc.)
Use Multiplier & Dilution Factor with ISIDs
=====
```

Signal 1: MWD1 A, Sig=254.16 Ref=off

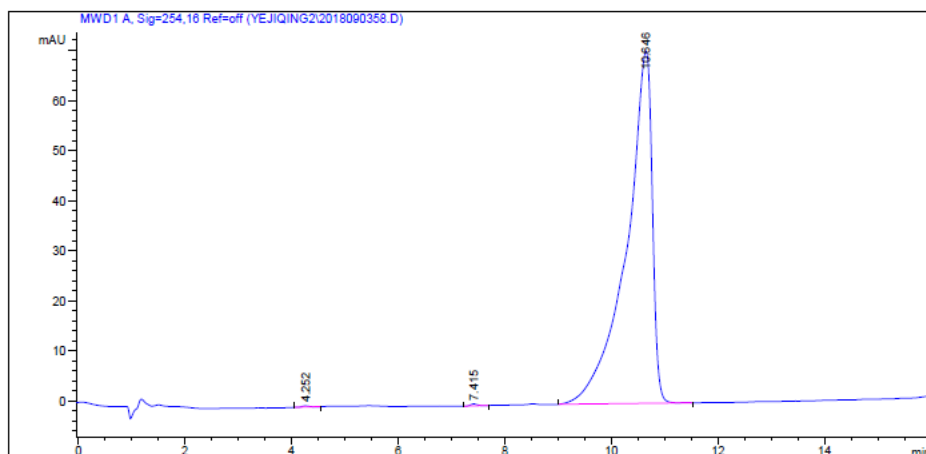
Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
1	9.660	VV	0.5424	6398.52100	161.43578	100.0000
Totals :				6398.52100	161.43578	

```
=====
*** End of Report ***
=====
```

C3-005

Data File C:\CHEM32\1\DATA\YEJIQING2\2018090358.D
Sample Name: 2018090358

```
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Acq. Operator   : YeJiqing
Acq. Instrument : Instrument 1          Location : Vial 1
Injection Date  : 9/5/2018 2:41:28 PM
Acq. Method     : C:\CHEM32\1\METHODS\YEJIQING.M
Last changed    : 9/5/2018 2:40:21 PM by YeJiqing
Analysis Method : C:\CHEM32\1\METHODS\YEJIQING.M
Last changed    : 12/12/2018 9:15:23 PM by SKY
                  (modified after loading)
Sample Info     : C3-18-005H
=====
```



Area Percent Report

```
=====
Sorted By      : Signal
Multiplier     : 1.0000
Dilution       : 1.0000
Sample Amount  : 1.00000 [ng/ul] (not used in calc.)
Use Multiplier & Dilution Factor with ISTDs
=====
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Signal 1: MWD1 A, Sig=254,16 Ref=off

Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
1	4.252	BB	0.1669	2.47320	2.31038e-1	0.1045
2	7.415	BB	0.1459	3.93987	4.03477e-1	0.1665

Instrument 1 12/12/2018 9:15:28 PM SKY

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Data File C:\CHEM32\1\DATA\YEJIQING2\2018090358.D
Sample Name: 2018090358

Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
3	10.646	BB	0.4735	2360.16724	70.33060	99.7290

Totals : 2366.58031 70.96512

*** End of Report ***