

# Supporting Information

## A “Motif-Oriented” Total Synthesis of Nannocystin Ax. Preparation and Biological Assessment of Analogues

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Rolf Müller,<sup>‡</sup> and Alois Fürstner<sup>†\*</sup>

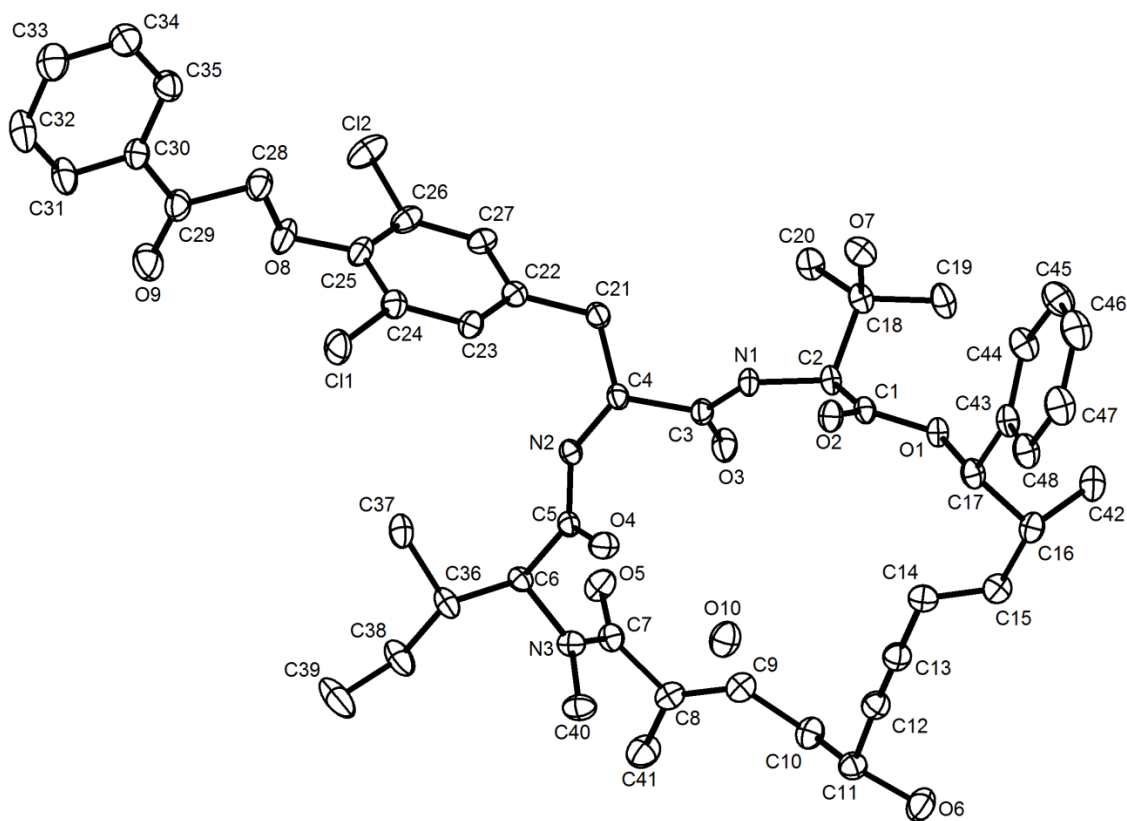
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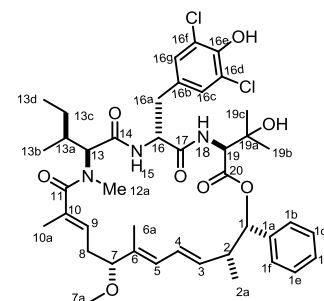
## Crystallographic Information



**Figure S1.** Structure of compound **26** monohydrate in the solid state with the crystallographic numbering scheme; anisotropic displacement parameters are shown at 50% probability level

**X-ray Crystal Structure Analysis of Compound 26:**  $C_{48}H_{55}Cl_2N_3O_9 \cdot H_2O$ ,  $M_r = 906.86 \text{ g} \cdot \text{mol}^{-1}$ , colorless needle, crystal size  $0.061 \times 0.041 \times 0.021 \text{ mm}^3$ , orthorhombic, space group  $P2_12_12_1$ ,  $a = 10.286(2) \text{ \AA}$ ,  $b = 13.899(5) \text{ \AA}$ ,  $c = 33.15(2) \text{ \AA}$ ,  $V = 4739(4) \text{ \AA}^3$ ,  $T = 80(2) \text{ K}$ ,  $Z = 4$ ,  $D_{calc} = 1.271 \text{ g} \cdot \text{cm}^3$ ,  $\lambda = 0.5636 \text{ \AA}$ ,  $\mu(22 \text{ keV}) = 0.110 \text{ mm}^{-1}$ , no absorption correction, merged data from three  $\phi$ -scans (SADABS/XPREP), P11 beamline at PETRA III, DESY Hamburg, equipped with Pilatus 6M detector,  $0.974 < \theta < 26.840^\circ$ , 279411 measured reflections, 14482 independent reflections, 12315 reflections with  $I > 2\sigma(I)$ ,  $R_{int} = 0.18$ , 99.9 % completeness to a resolution of  $0.7 \text{ \AA}$  with an average redundancy of over 18. The structure was solved by dual methods (SHELXT) and refined by full-matrix least-squares against  $F^2$  to  $R_1 = 0.046 [I > 2\sigma(I)]$ ,  $wR_2 = 0.126$ , 599 parameters. The H atoms on the amide N atoms and solvate water molecule were located on a difference Fourier map and refined using isotropic atomic displacement parameters (adp's), otherwise H atoms riding with adp's fixed at  $1.20 \times U_{C_{aromatic/methylene}}$  or  $1.50 \times U_{C_{methyl}}$ ,  $S = 1.091$ . Flack absolute structure parameter =  $0.02(3)$  [4899 quotients], residual electron density  $0.60 / -0.57 \text{ e} \cdot \text{\AA}^{-3}$ . **CCDC-1584397**.

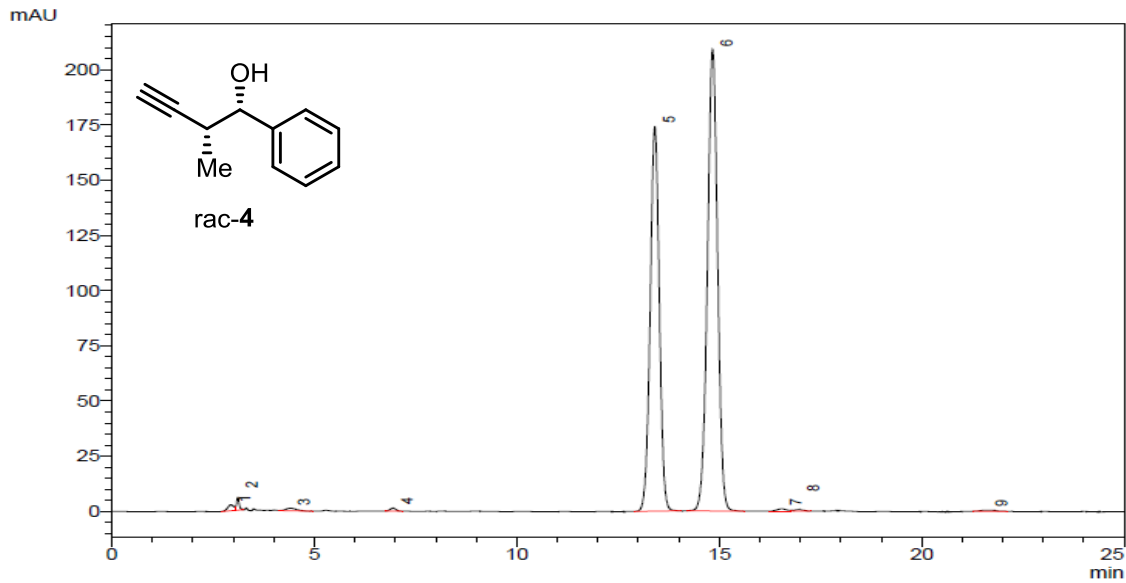
**Table S1. Comparison of  $^1\text{H}$  NMR data of Nannocystin Ax (1) in  $[\text{D}_6]$ -DMSO;  
Numbering Scheme as Shown in the Insert**



Position	Literature <small>[Fehler! Textmarke nicht definiert.]</small>		Synthetic sample		$\Delta\delta$
	$\delta$ (ppm)	$J$ (Hz)	$\delta$ (ppm)	$J$ (Hz)	
<b>18</b>	8.53	(d, $J = 10.0$ )	8.49	(d, $J = 9.8$ )	0.04
<b>15</b>	7.92	(d, $J = 9.6$ )	7.86	(d, $J = 9.6$ )	0.06
<b>1b</b>	7.54	(d, $J = 7.6$ )	7.53	(d, $J = 7.7$ )	0.01
<b>16g</b>	7.38	s	7.37	s	0.01
<b>1c</b>	7.32	(t, $J = 7.6$ )	7.32	(t, $J = 7.5$ )	0
<b>1d</b>	7.25	(t, $J = 7.6$ )	7.24	(t, $J = 7.4$ )	0.01
<b>4</b>	6.35	m	6.37–6.32	m	
<b>5</b>	6.02	m	6.04	(d, $J = 10.8$ )	
<b>3</b>	6.01	m	5.99	(dd, $J = 15.4, 4.6$ )	
<b>1</b>	5.89	br	5.88	s	
<b>19a-OH</b>	5.14	s	5.09	s	0.05
<b>9</b>	5.13	m	5.16–5.13	m	
<b>16</b>	4.70	m	4.74–4.70	m	
<b>19</b>	4.63	m	4.61	(d, $J = 9.0$ )	
<b>13</b>	4.51	(d, $J = 11.1$ )	4.52	(d, $J = 11.2$ )	
<b>7</b>	3.53	m	3.54	(dd, $J = 9.6, 4.1$ )	
<b>7a</b>	3.07	s	3.08	s	-0.01
<b>16a</b>	2.80	m	2.84–2.78	m	
<b>12a</b>	2.74	s	2.73	s	0.01
<b>2</b>	2.65	m	2.66–2.64	m	
<b>16a</b>	2.59	m	2.59–2.57	m	
<b>13a</b>	1.74	m	1.75–1.71	m	
<b>10a</b>	1.71	s	1.72	s	-0.01
<b>6a</b>	1.65	s	1.65	s	0
<b>13c</b>	1.22	m	1.23–1.18	m	
<b>19b</b>	1.10	s	1.10	s	0
<b>19c</b>	1.02	s	1.02	s	0
<b>2a</b>	0.93	(d = 6.7)	0.93	(d, $J = 6.8$ )	0
<b>13c</b>	0.89	m	0.86–0.84	m	
<b>13d</b>	0.76	(t, $J = 7.2$ )	0.77	(t, $J = 7.3$ )	-0.01
<b>13b</b>	0.41	(d, $J = 6.6$ )	0.43	(d, $J = 6.5$ )	-0.02

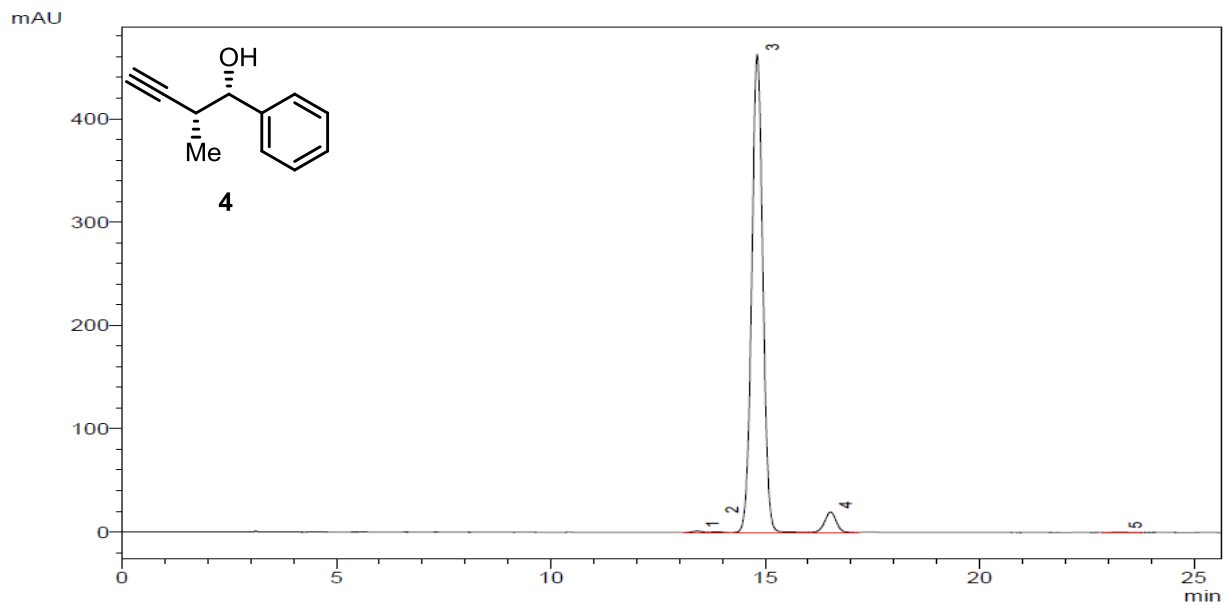
**Table S2. Comparison of  $^{13}\text{C}$  NMR data of Nannocystin Ax (1) in  $[\text{D}_6]$ -DMSO; Numbering Scheme as Shown in the Insert**

Literature <sup>[Fehler! Textmarke nicht definiert.]</sup> ( $\delta$ , ppm)	Synthetic sample ( $\delta$ , ppm)	$\Delta\delta$ (ppm)
172.8	172.8	0
170.7	170.7	0
170.5	170.5	0
169.1	169.1	0
147.3	147.3	0
139.7	139.7	0
137.3	137.3	0
133.9	133.9	0
133.5	133.5	0
130.9	130.9	0
129.6	129.6	0
128.5	128.4	0.1
127.8	127.8	0
127.0	127.0	0
126.1	126.1	0
125.1	125.1	0
124.8	124.8	0
121.6	121.6	0
84.9	84.9	0
78.9	78.9	0
71.7	71.7	0
59.3	59.3	0
58.9	58.9	0
55.0	55.0	0
52.9	52.9	0
41.7	41.7	0
36.5	36.5	0
31.7	31.7	0
31.3	31.1	0.2
30.4	30.4	0
28.1	28.1	0
24.5	24.5	0
24.0	24.0	0
14.8	14.8	0
14.4	14.4	0
11.1	11.1	0
10.2	10.2	0
10.0	10.1	0



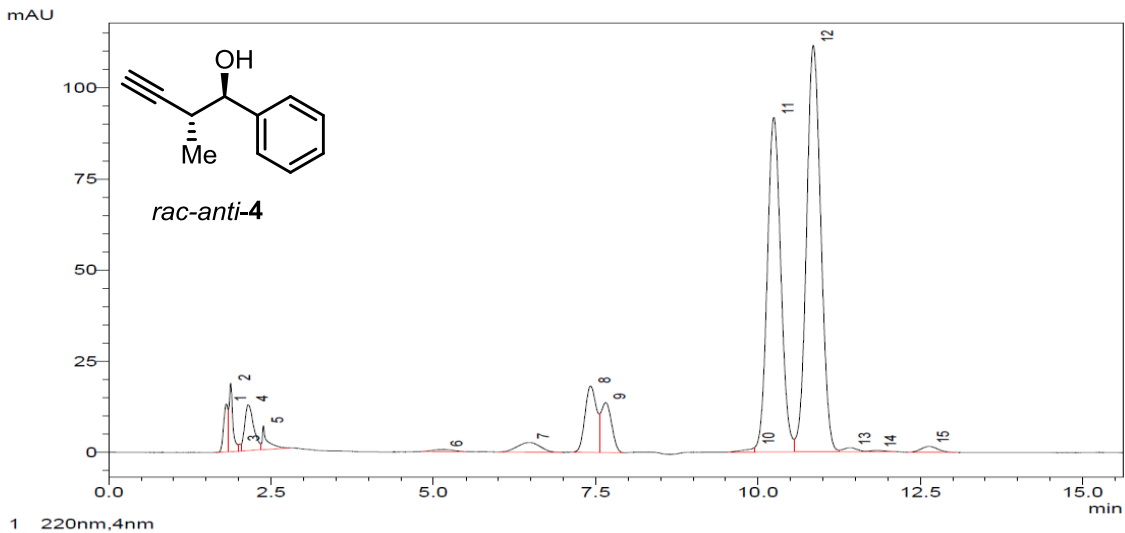
1 220nm,4nm

Peak #	Ret. Time	Area %	Name
1	2,94	0,43	
2	3,11	0,42	
3	4,40	0,33	
4	6,94	0,21	
5	13,41	41,99	1. Enantiomer
6	14,83	55,97	2. Enantiomer
7	16,54	0,28	
8	16,97	0,16	
9	21,62	0,21	
Total		100,00	

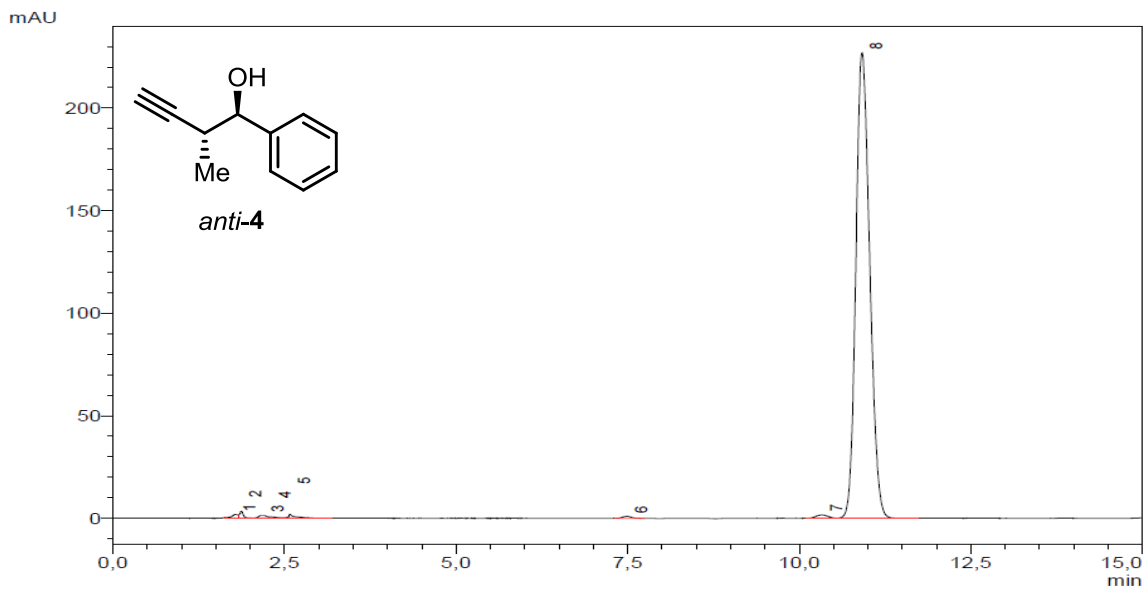


1 220nm,4nm

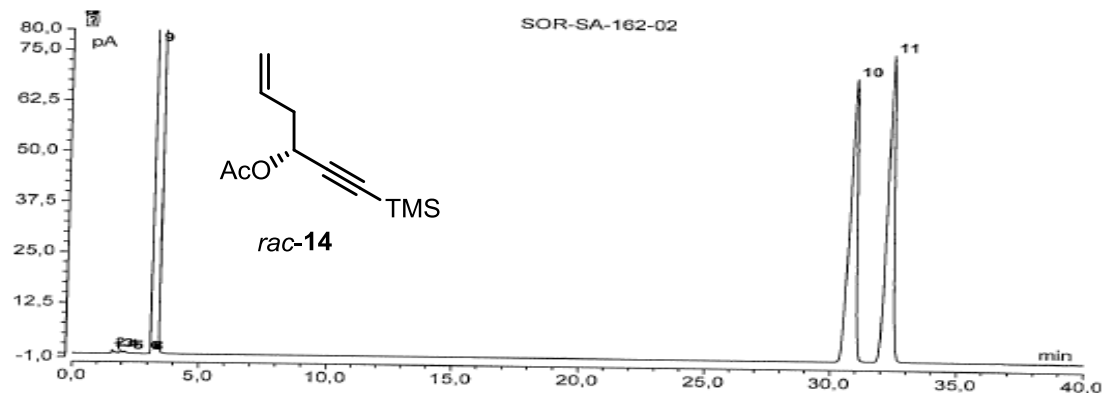
Peak #	Ret. Time	Area %	Name
1	13,42	0,26	1. Enantiomer
2	13,88	0,11	
3	14,81	95,28	2. Enantiomer 99.5 % ee
4	16,53	4,25	
5	23,28	0,10	
Total		100,00	



Peak #	Ret. Time	Area %	Name
1	1.81	1.56	
2	1.88	1.99	
3	2.01	0.14	
4	2.15	3.15	
5	2.38	0.95	
6	5.14	0.31	
7	6.47	1.68	
8	7.42	5.81	
9	7.65	4.12	
10	9.95	0.30	
11	10.24	34.48	1. Enantiomer
12	10.85	44.23	2. Enantiomer
13	11.43	0.42	
14	11.82	0.15	
15	12.63	0.75	
Total		100.00	



Peak #	Ret. Time	Area %	Name
1	1.79	0.40	
2	1.87	0.42	
3	2.19	0.36	
4	2.30	0.23	
5	2.58	0.47	
6	7.49	0.28	
7	10.33	0.64	1. Enantiomer
8	10.92	97.20	2. Enantiomer 98.7 % ee
Total		100.00	

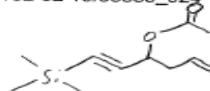


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 Sequenz: 95538 SOR-SA FK\_924 Measured: 13.11.15 12:46  
 Sequenz date: 13.11.15 Processing M.: MPI  
 Report-File: Verhältnis 162-02

chirale Messung, Racemat, ee-verhältnis

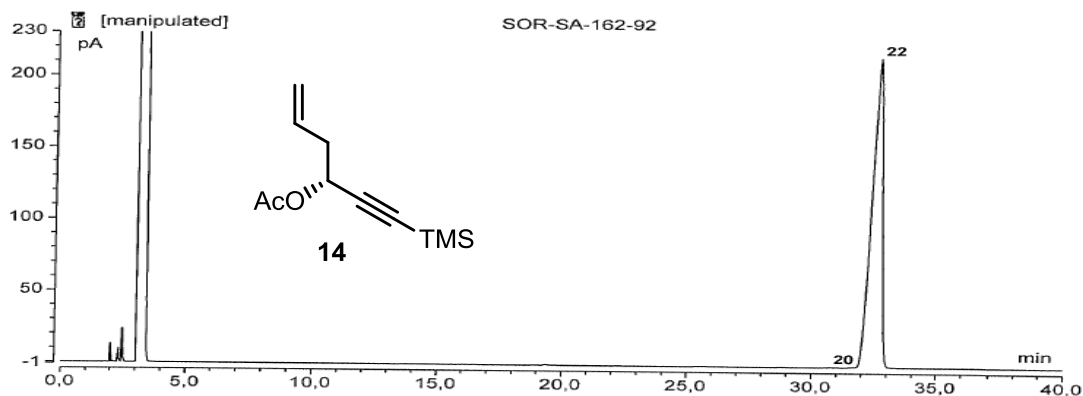
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No.	Ret.Time min	Rel.Area %	Peak Name
10	30,85	49,97	
11	32,29	50,03	



Instrument parameters:  
 Column: 30,0 m BGB-178/BGB-15, G/698  
 Temperature: 230/50min iso,80 4/min 220,5min iso/350  
 Gas: 0,50 bar H2  
 Sample size: 0,2 µL

F. Weller



Sample: SOR-SA-162-92 Instrument: GC\_122  
 Sequenz: 95538 SOR-SA FK\_924 Measured: 13.11.15 13:59  
 Sequenz date: 13.11.15 Processing M.: MPI  
 Report-File: Verhältnis 162-92

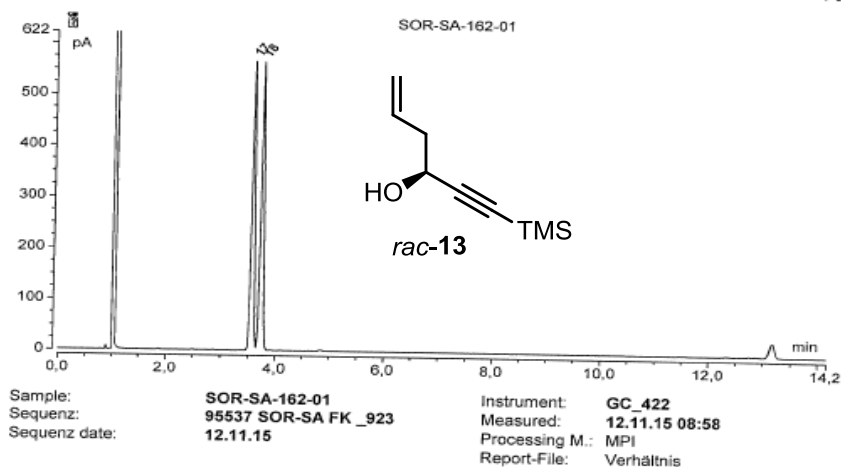
chirale Messung, Racemat, ee-verhältnis

Zuomung nach achiraler MSD-Messung w00702 SOR-SA-162-02 15/95538\_924

No.	Ret.Time min	Rel.Area %	Peak Name
20	30,75	0,07	
22	32,67	99,93	

Instrument parameters:  
 Column: 30,0 m BGB-178/BGB-15, G/698  
 Temperature: 230/50min iso,80 4/min 220,5min iso/350  
 Gas: 0,50 bar H2  
 Sample size: 0,2 µL

F. Weller



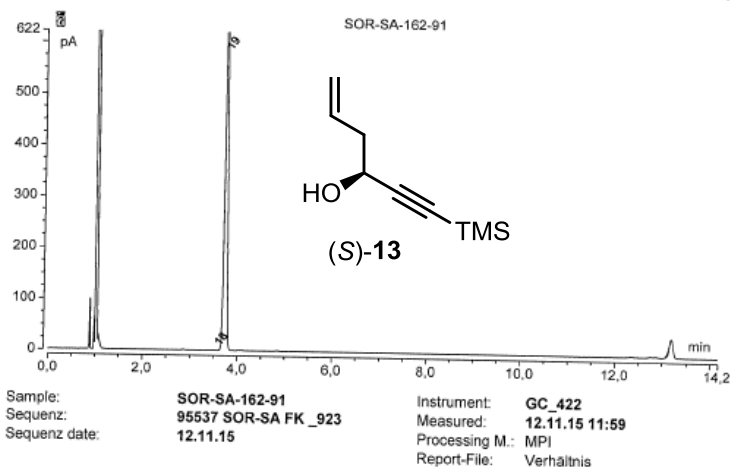
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No.	Ret.Time min	Rel.Area %	Peak Name	
17	3,57	49,93		 Racemat
18	3,74	50,07		

Instrument parameters:

Column:	24,5 m	Hydrodex-beta-TBDAC ; G/589
Temperature:	220/10min iso, 105 6/min 220,5min iso/350	
Gas:	0,80 bar	Hydrogen
Sample size:	0,2 µL	

F. Noll



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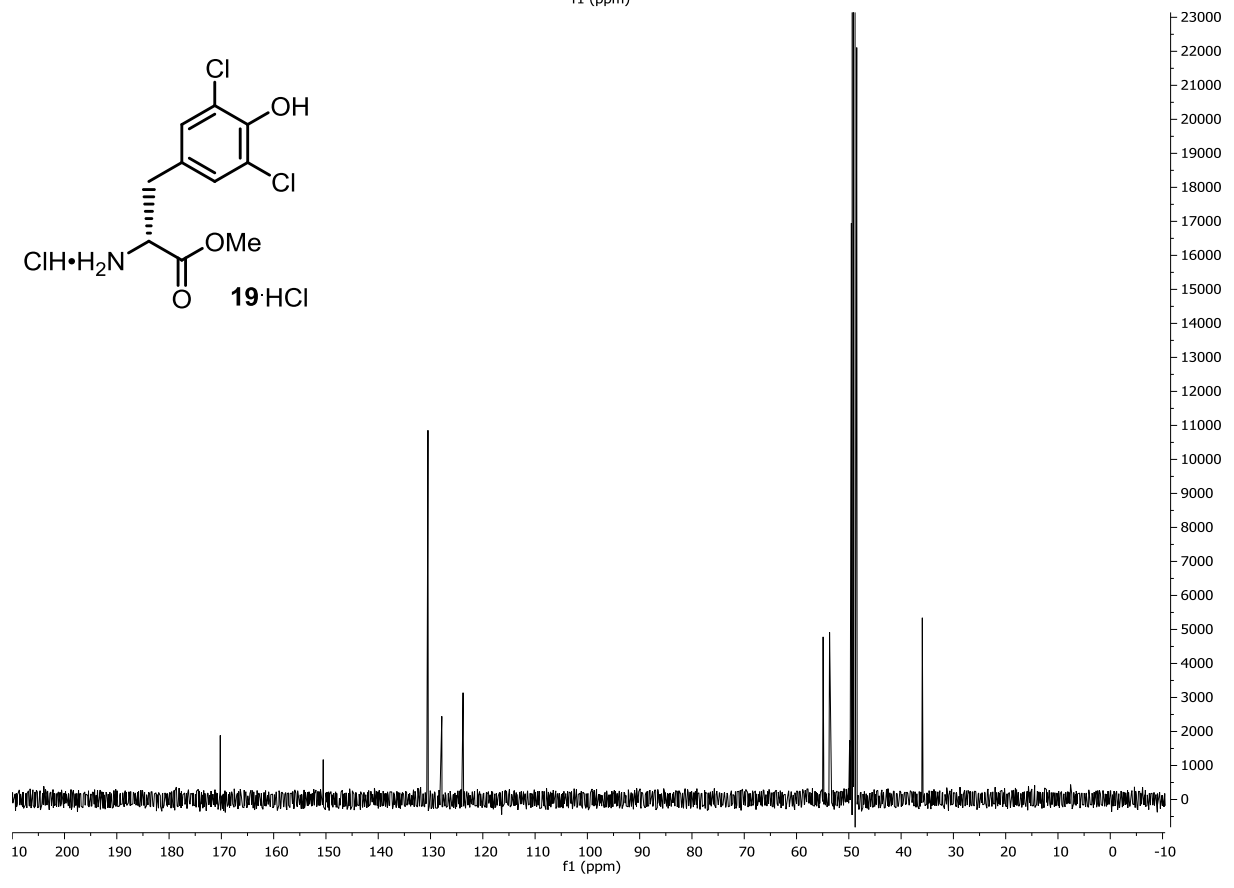
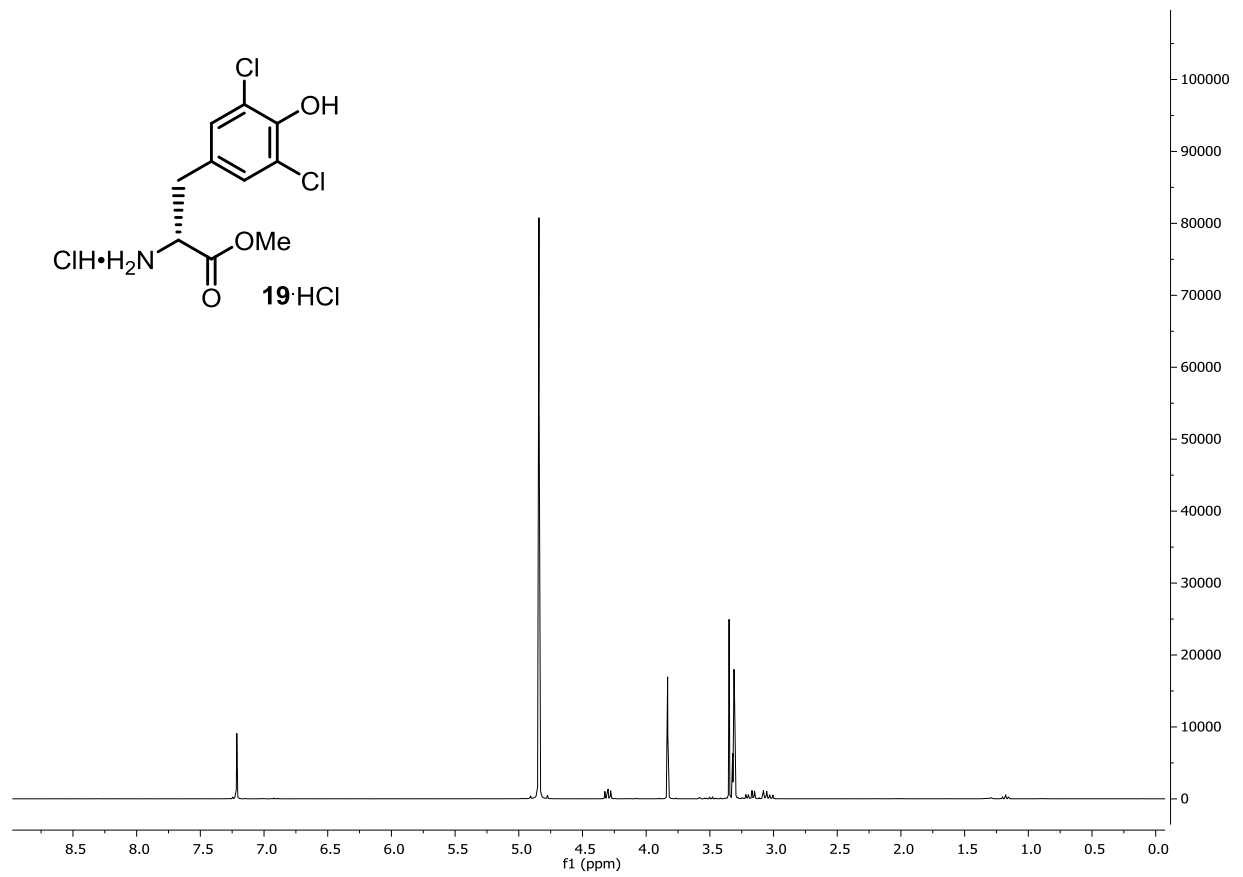
No.	Ret.Time min	Rel.Area %	Peak Name	
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19	3,73	99,99		

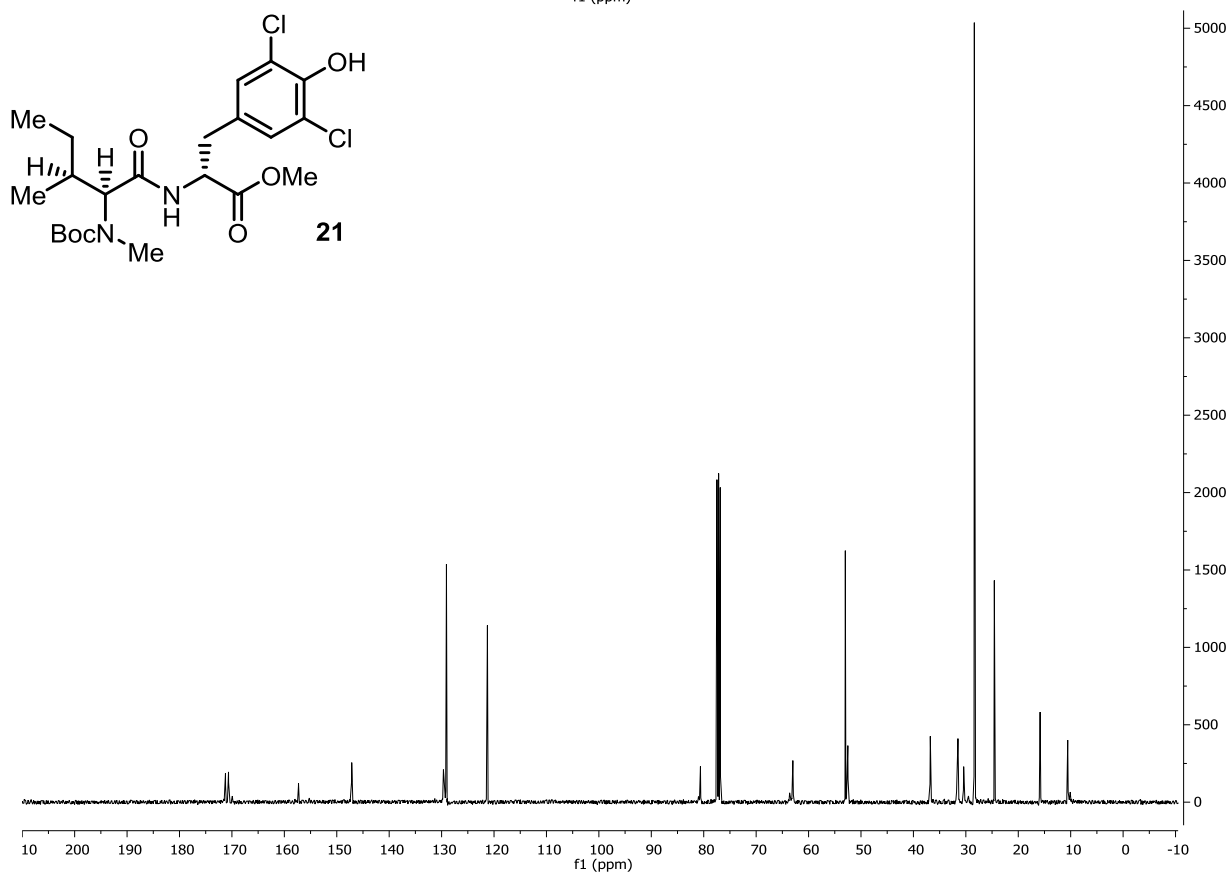
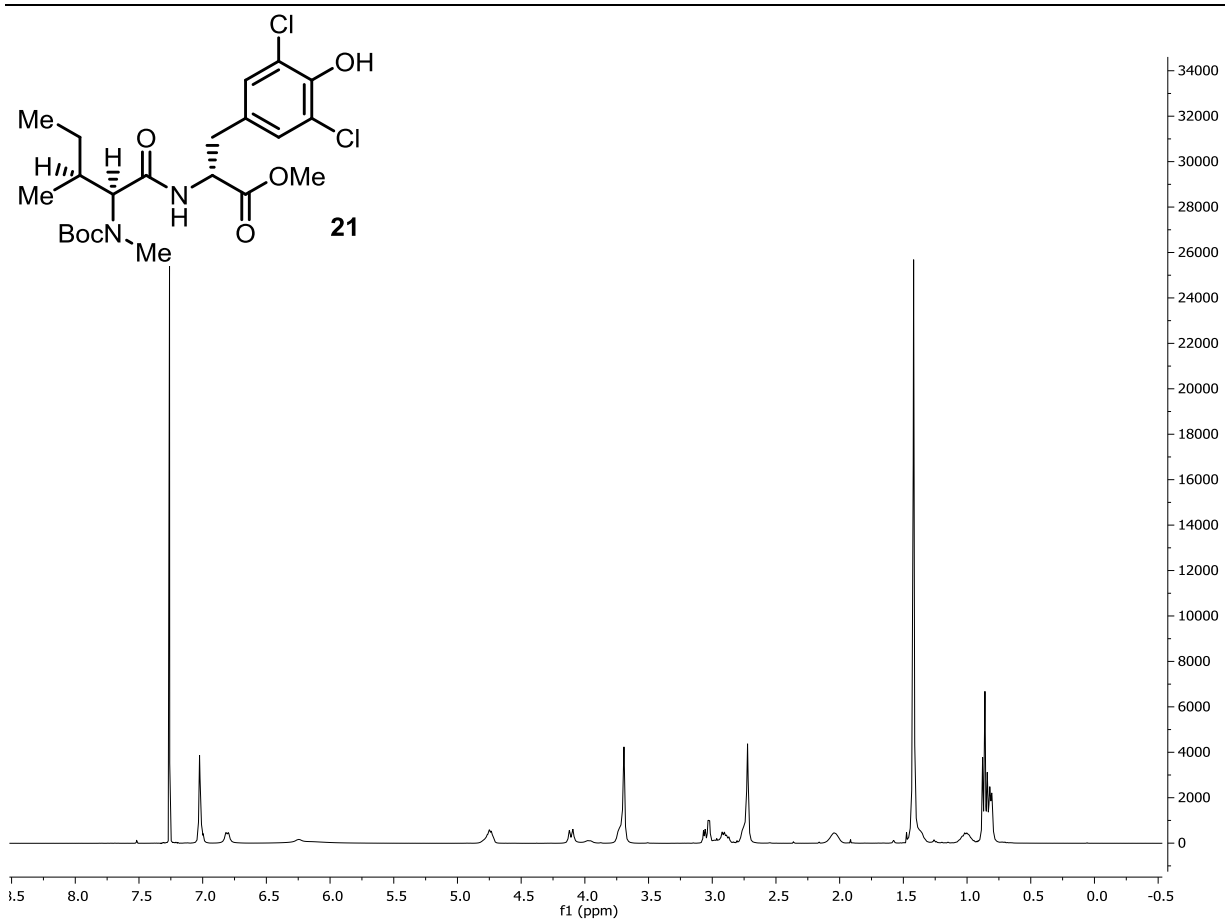
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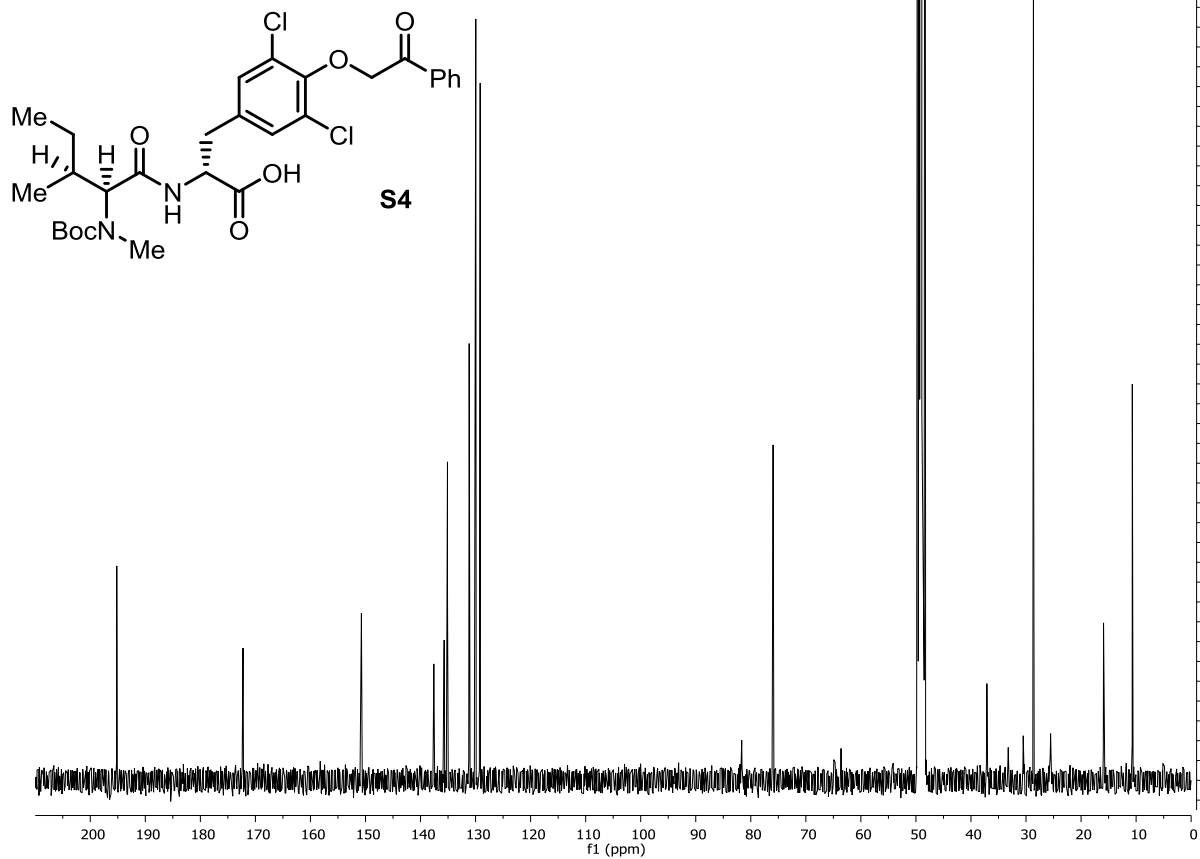
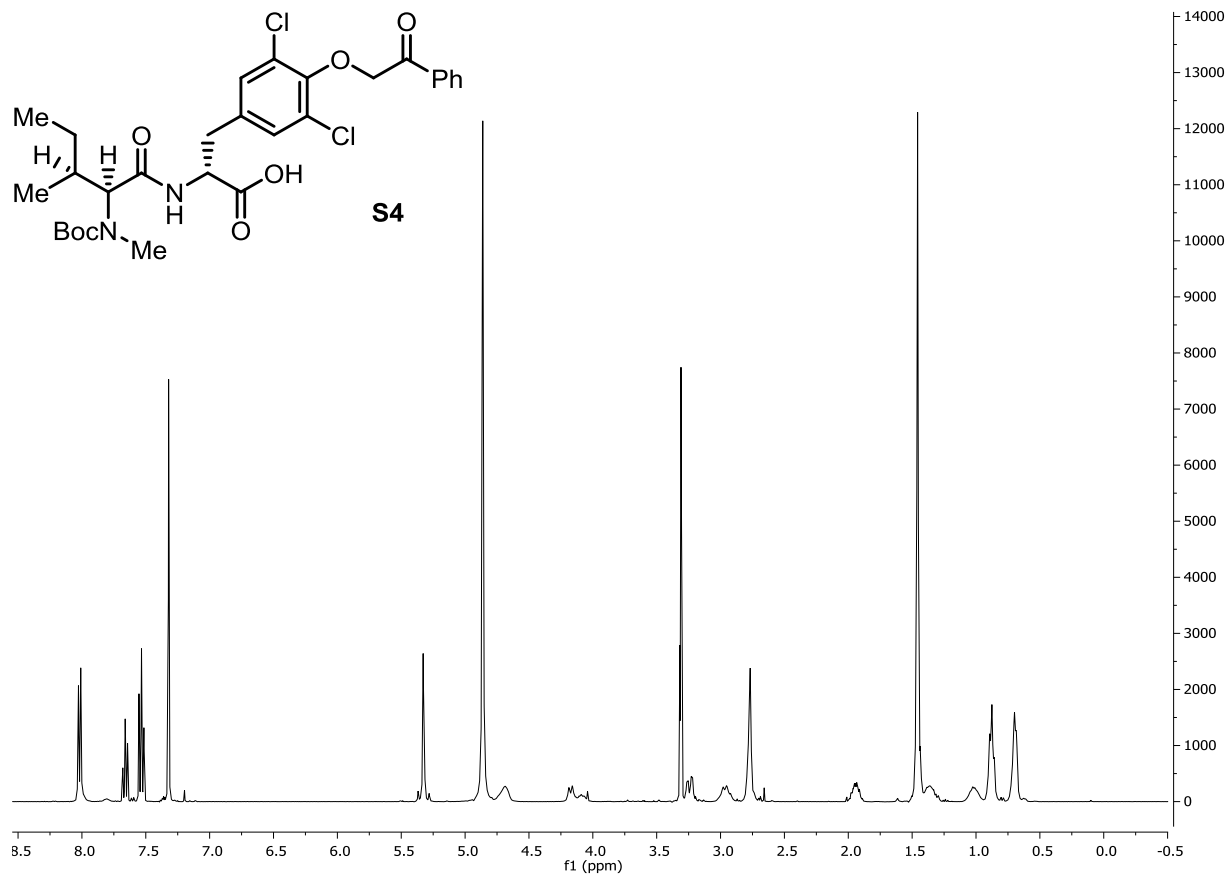
Column:	24,5 m	Hydrodex-beta-TBDAC ; G/589
Temperature:	220/10min iso, 105 6/min 220,5min iso/350	
Gas:	0,80 bar	Hydrogen
Sample size:	0,2 µL	

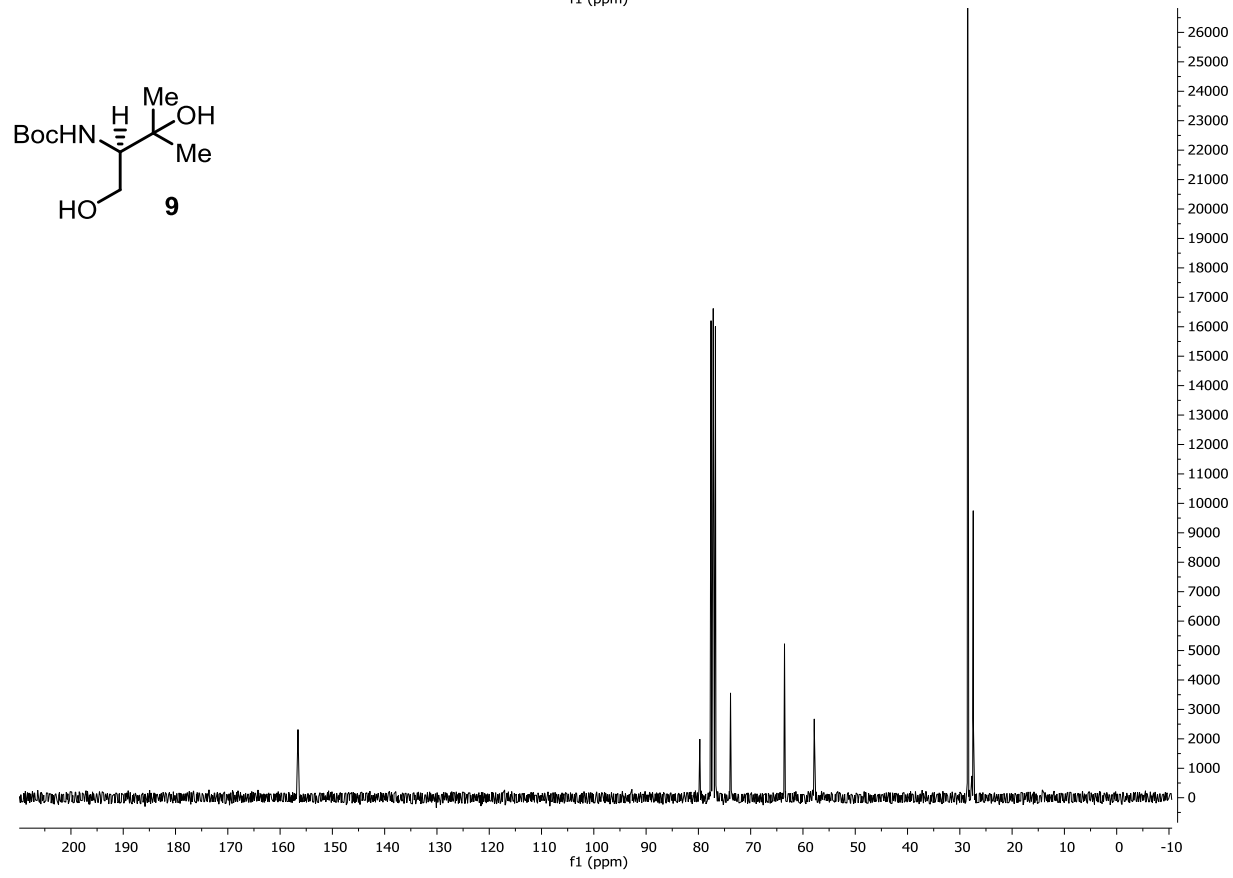
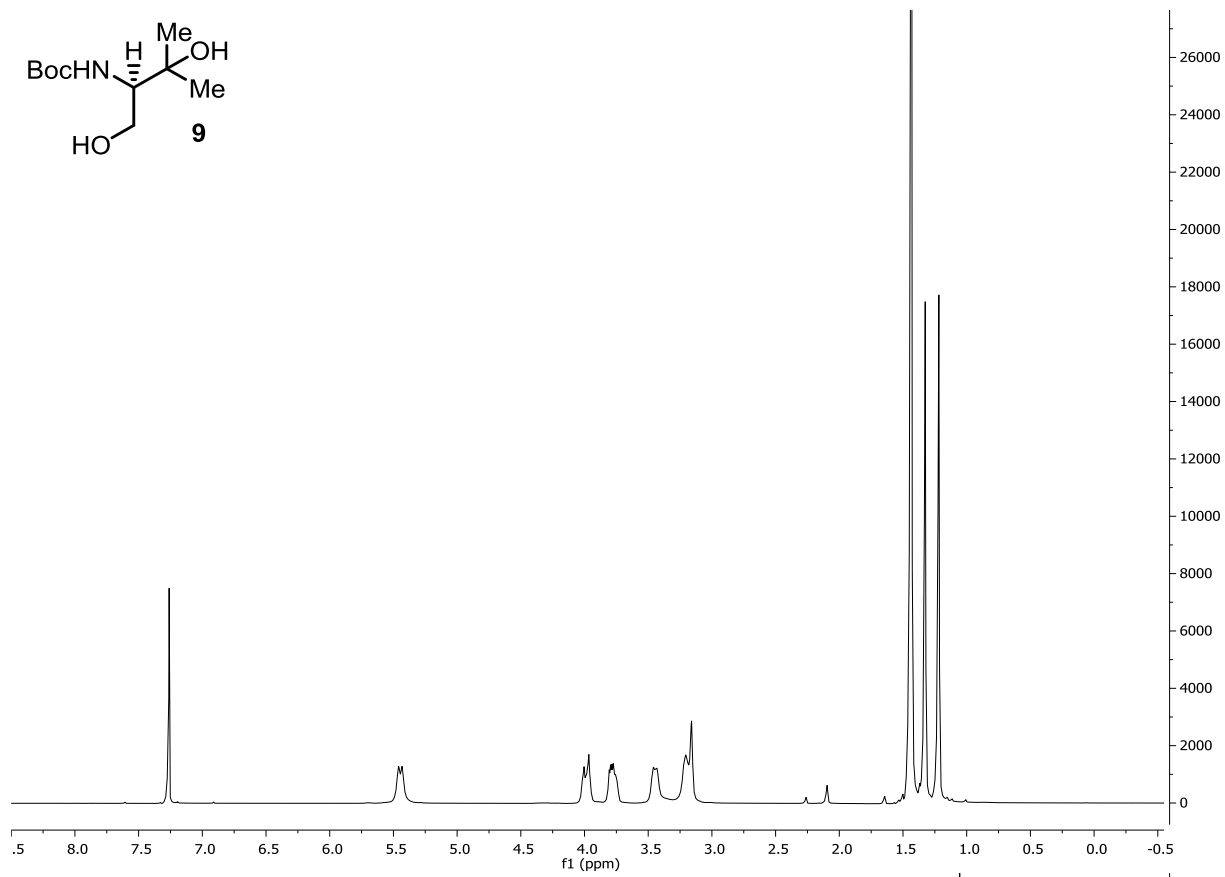
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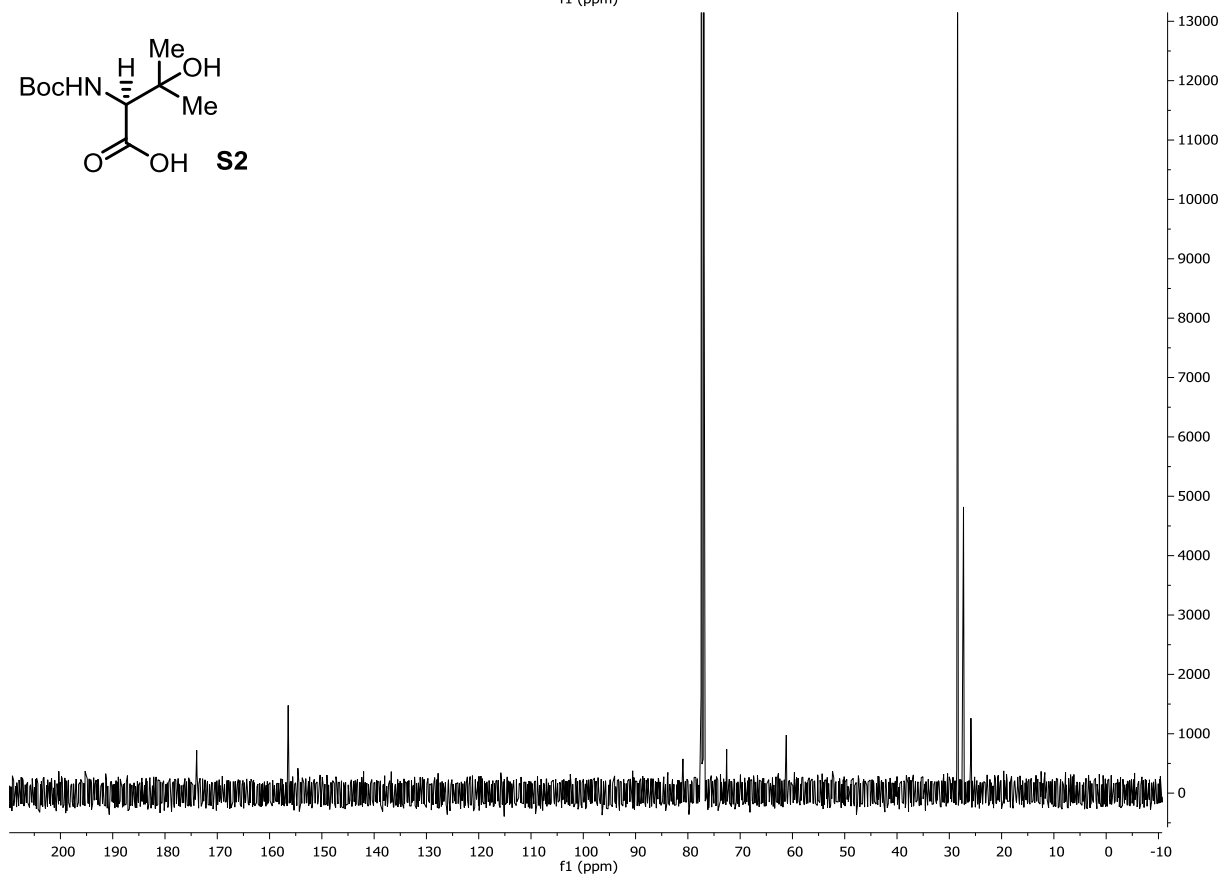
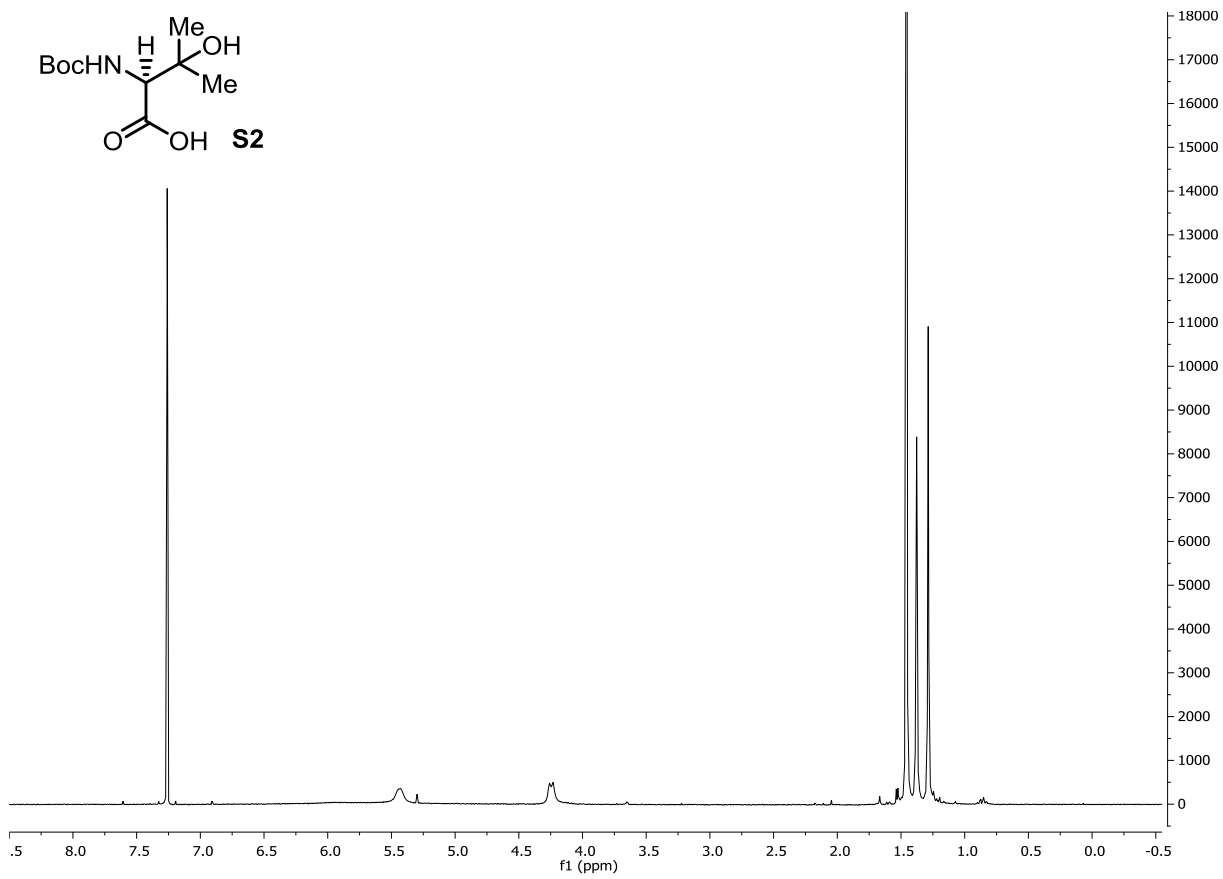


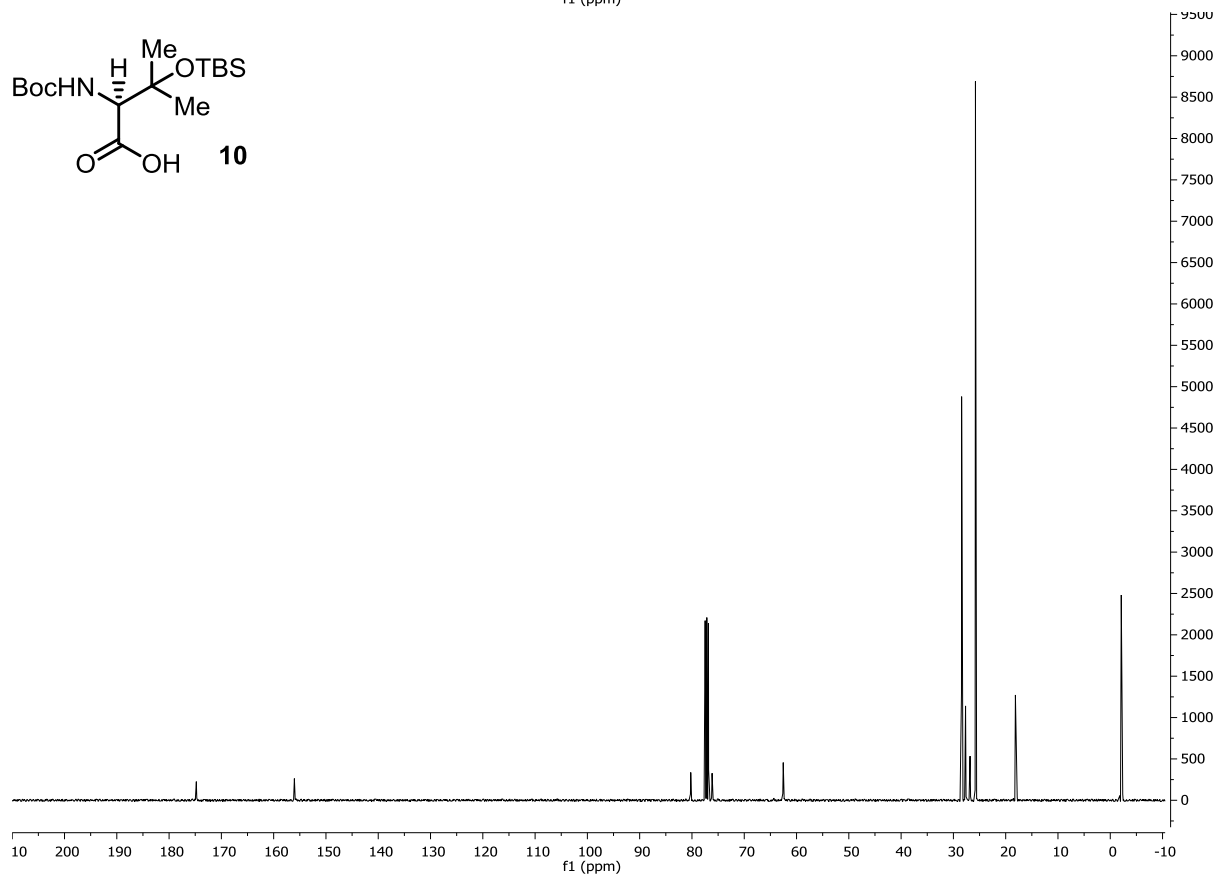
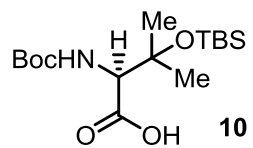
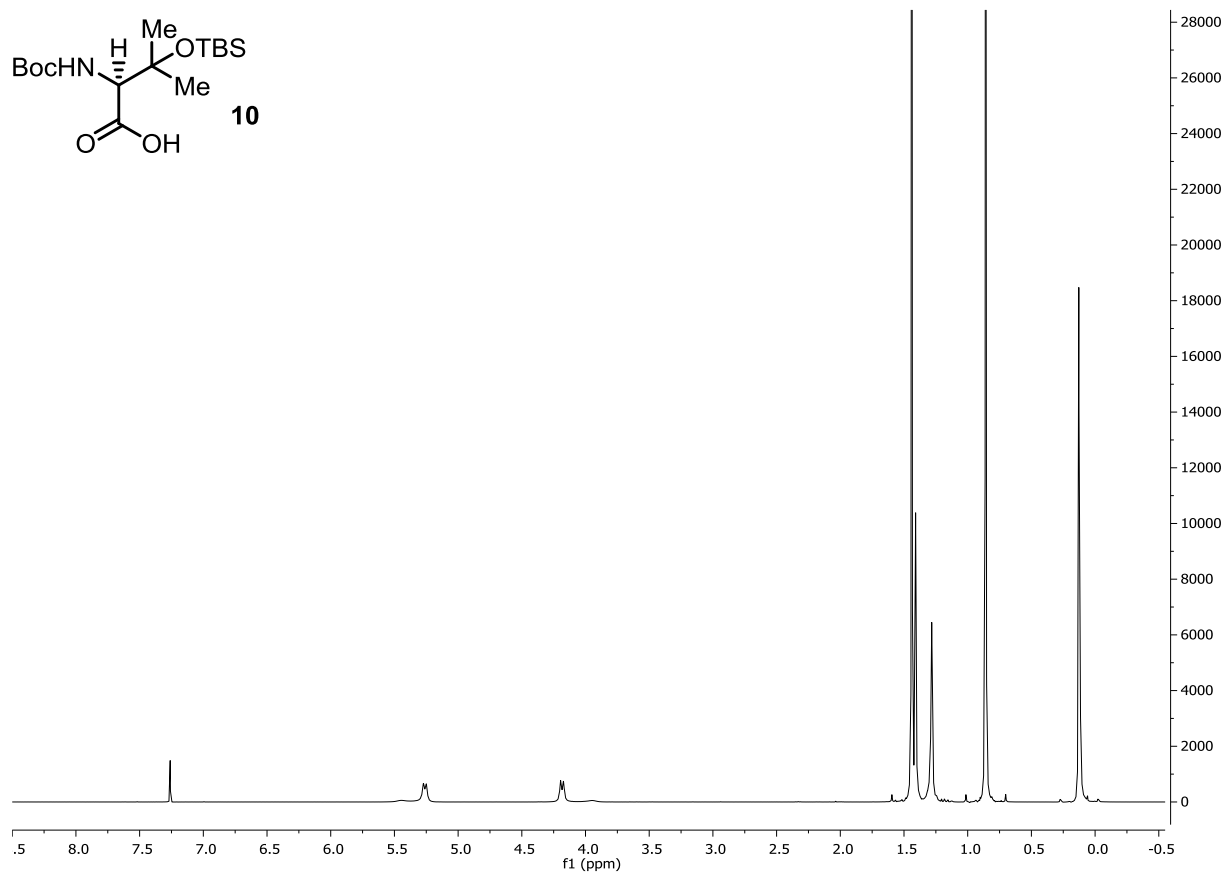
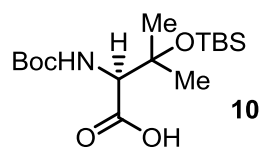


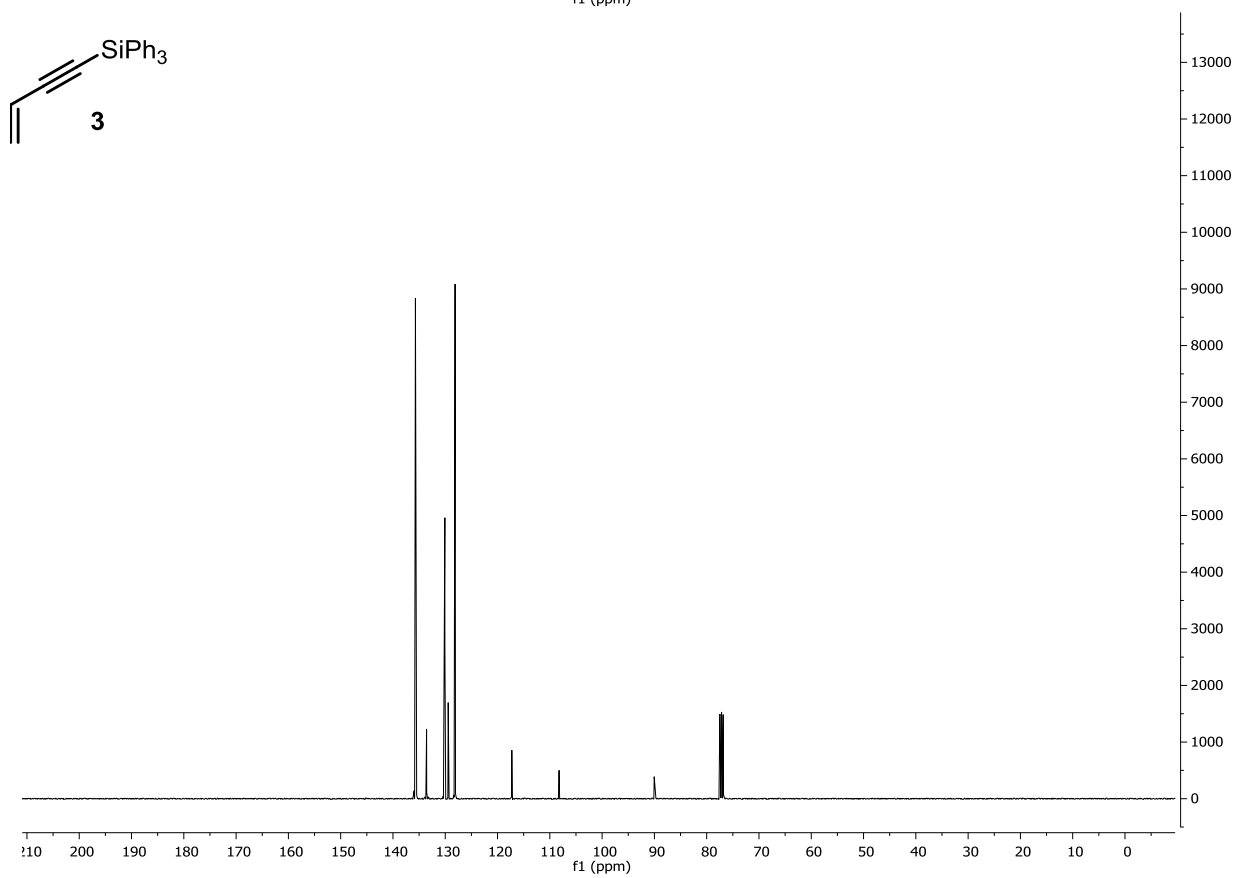
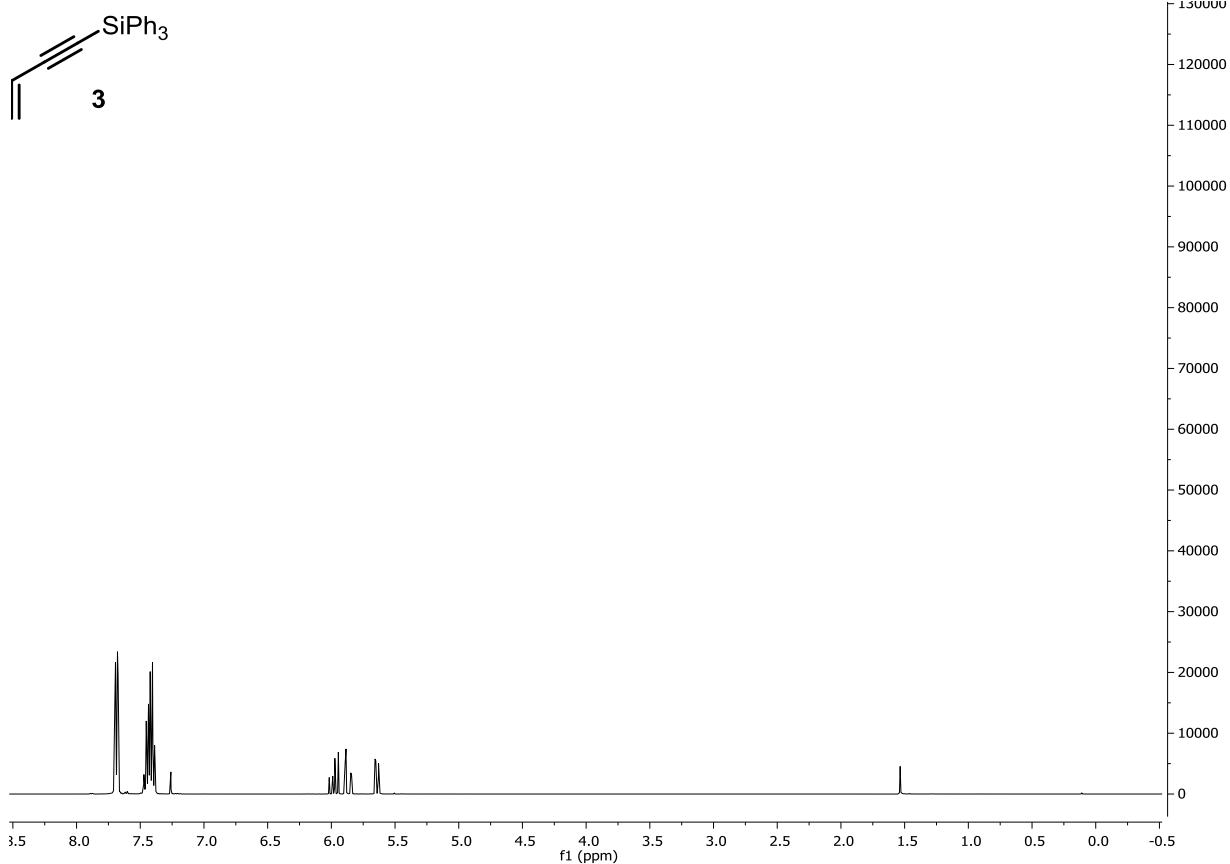


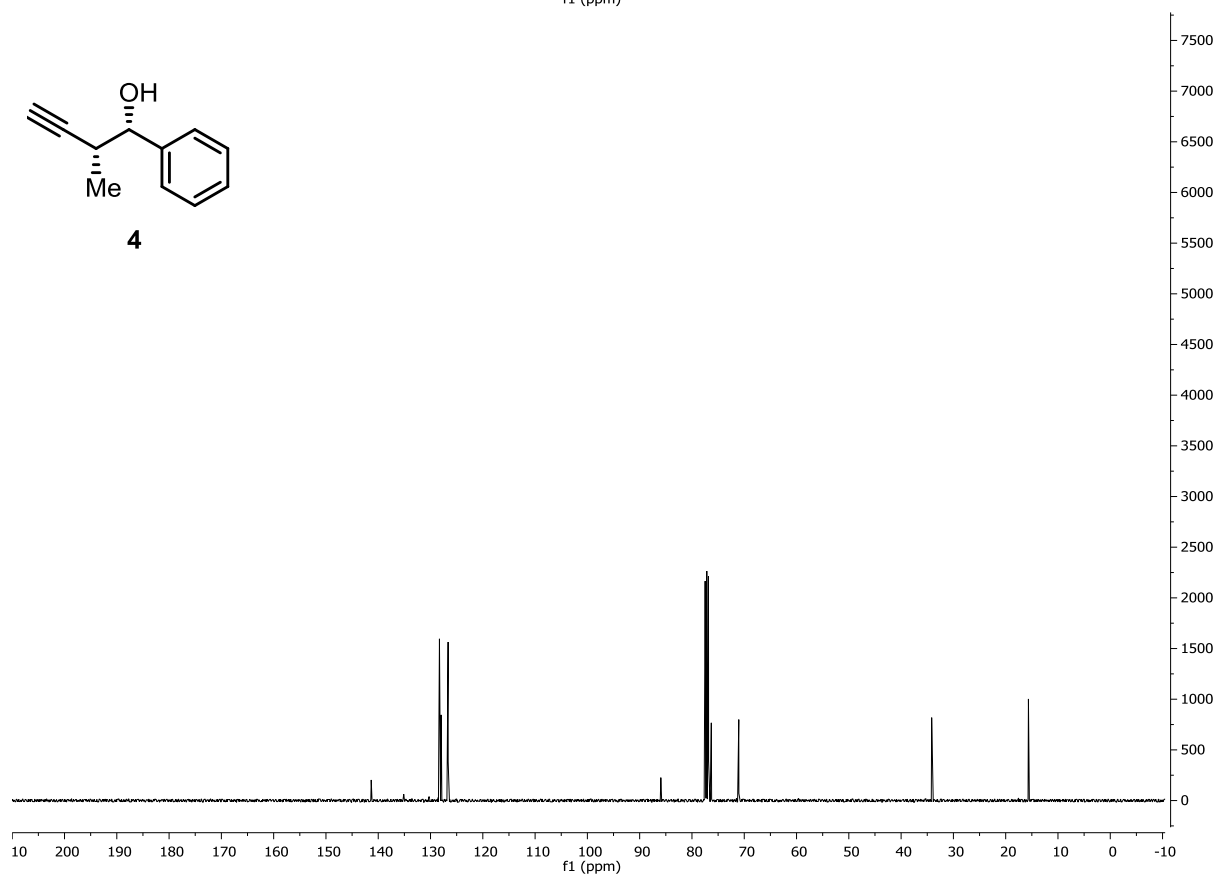
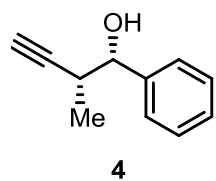
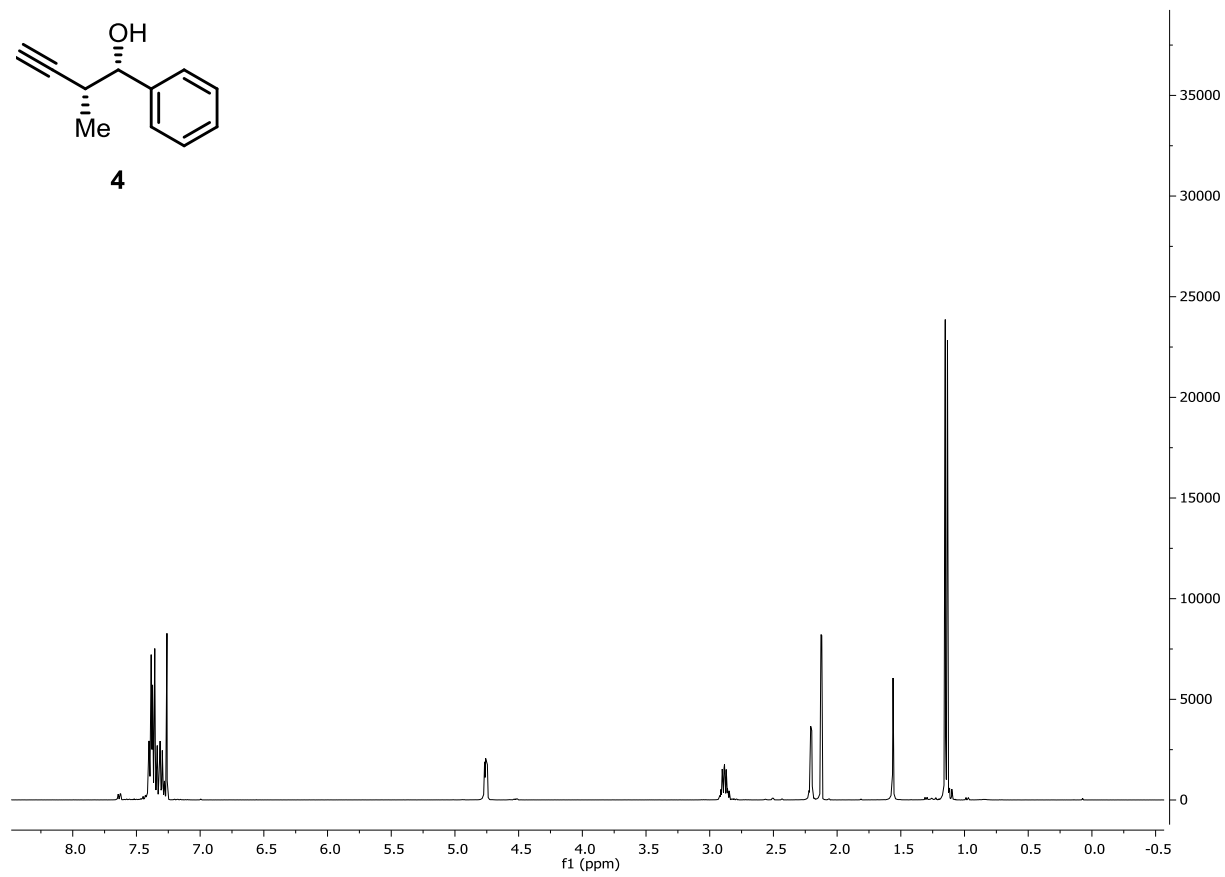
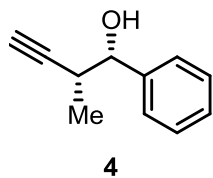




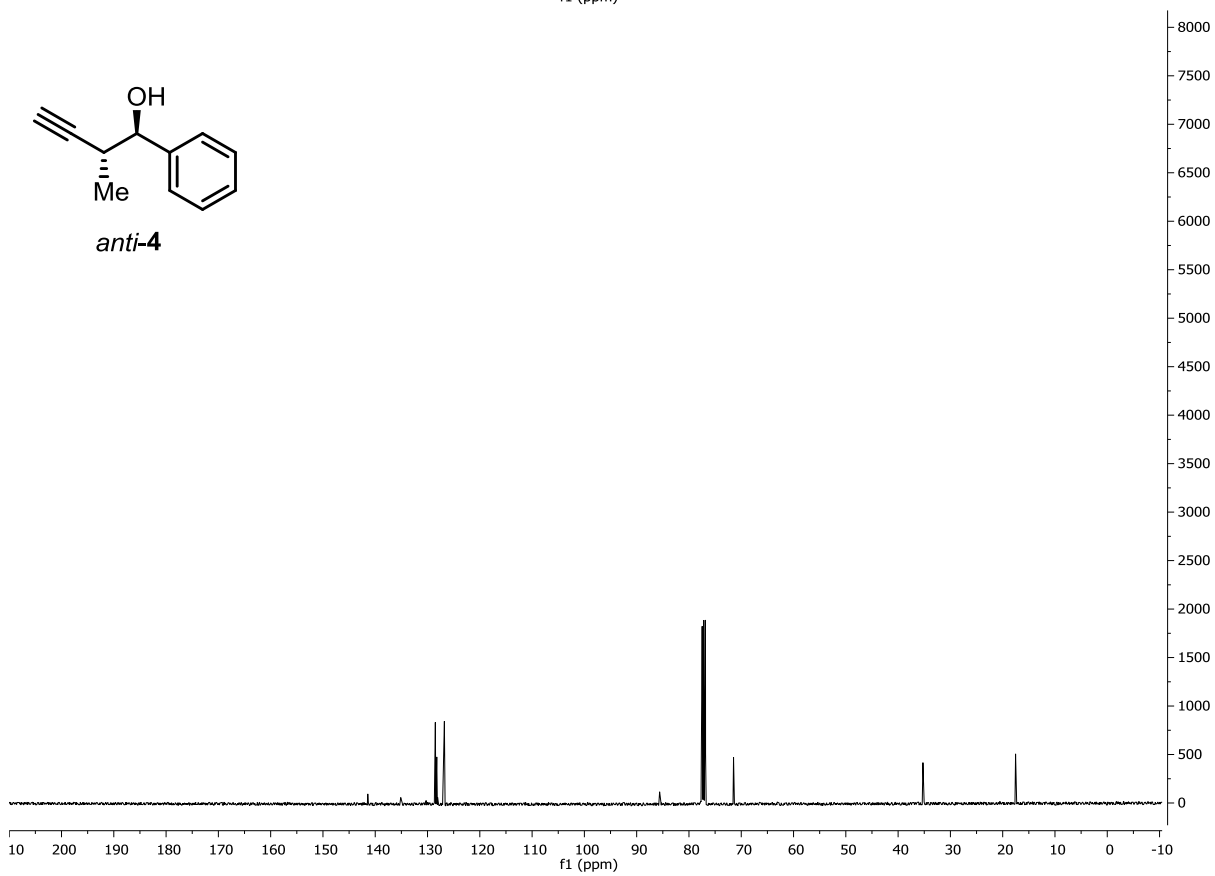
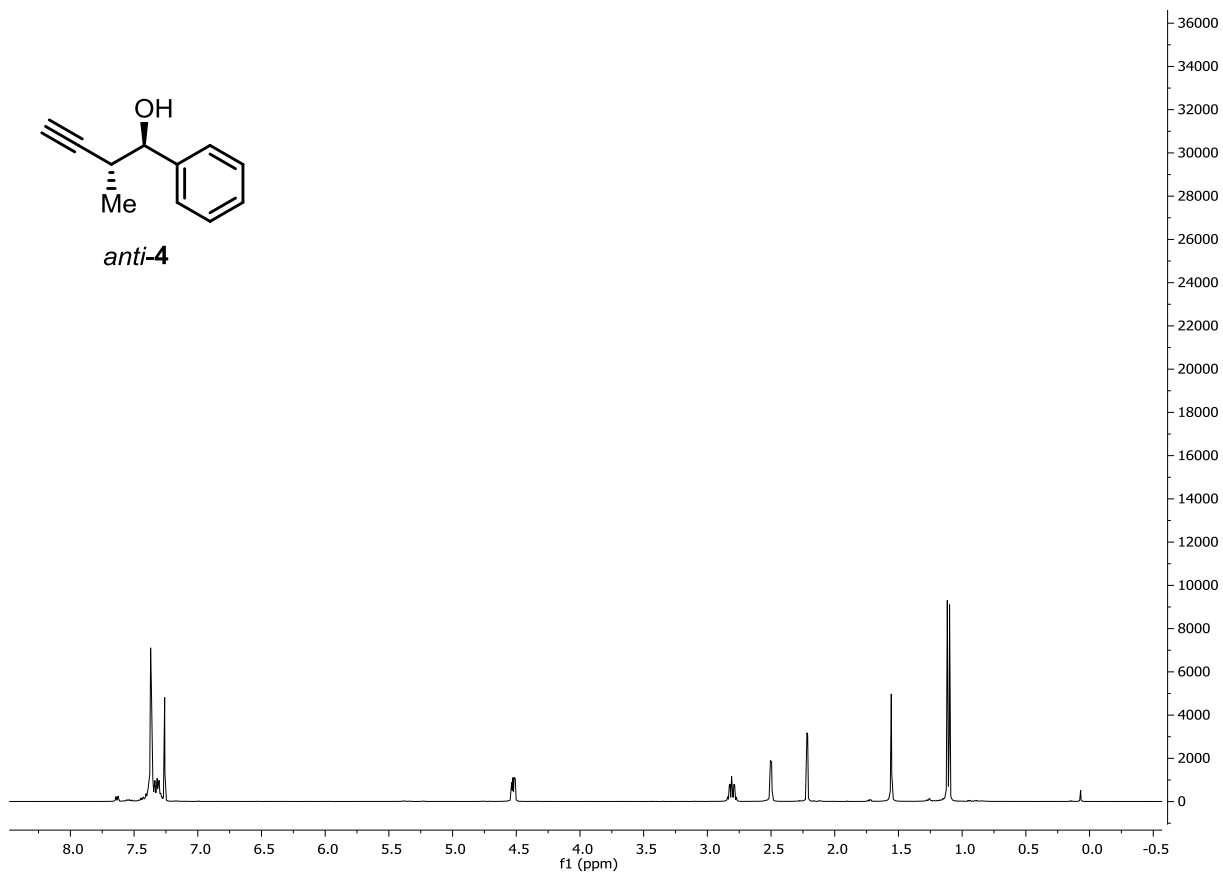


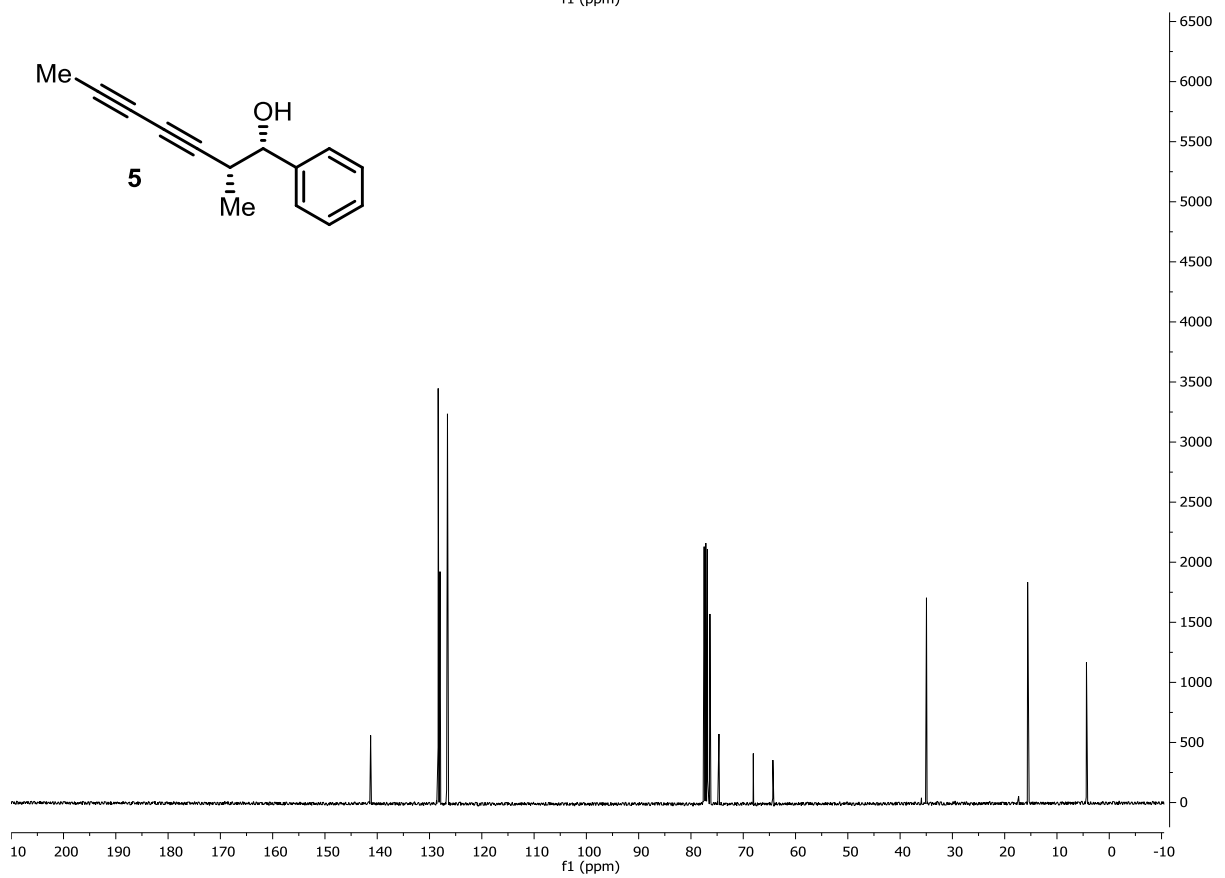
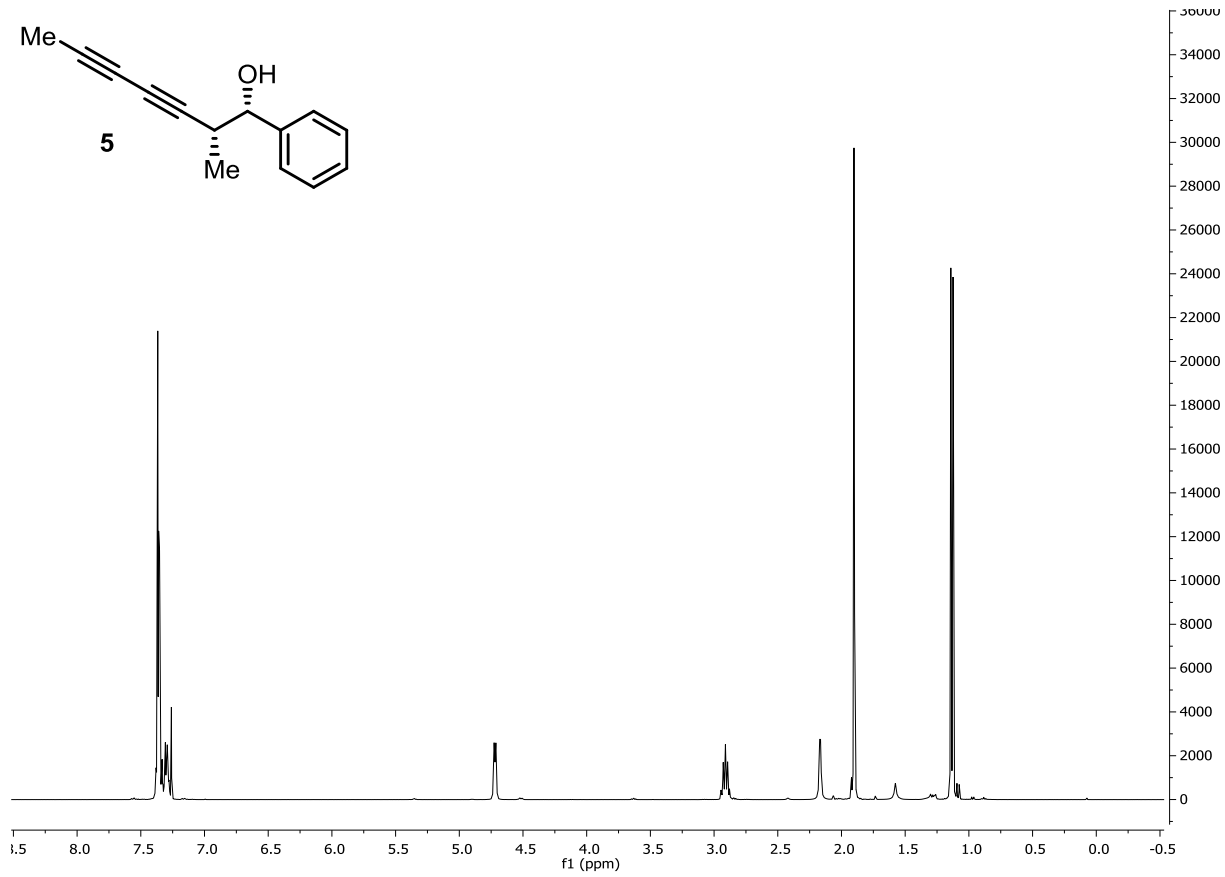


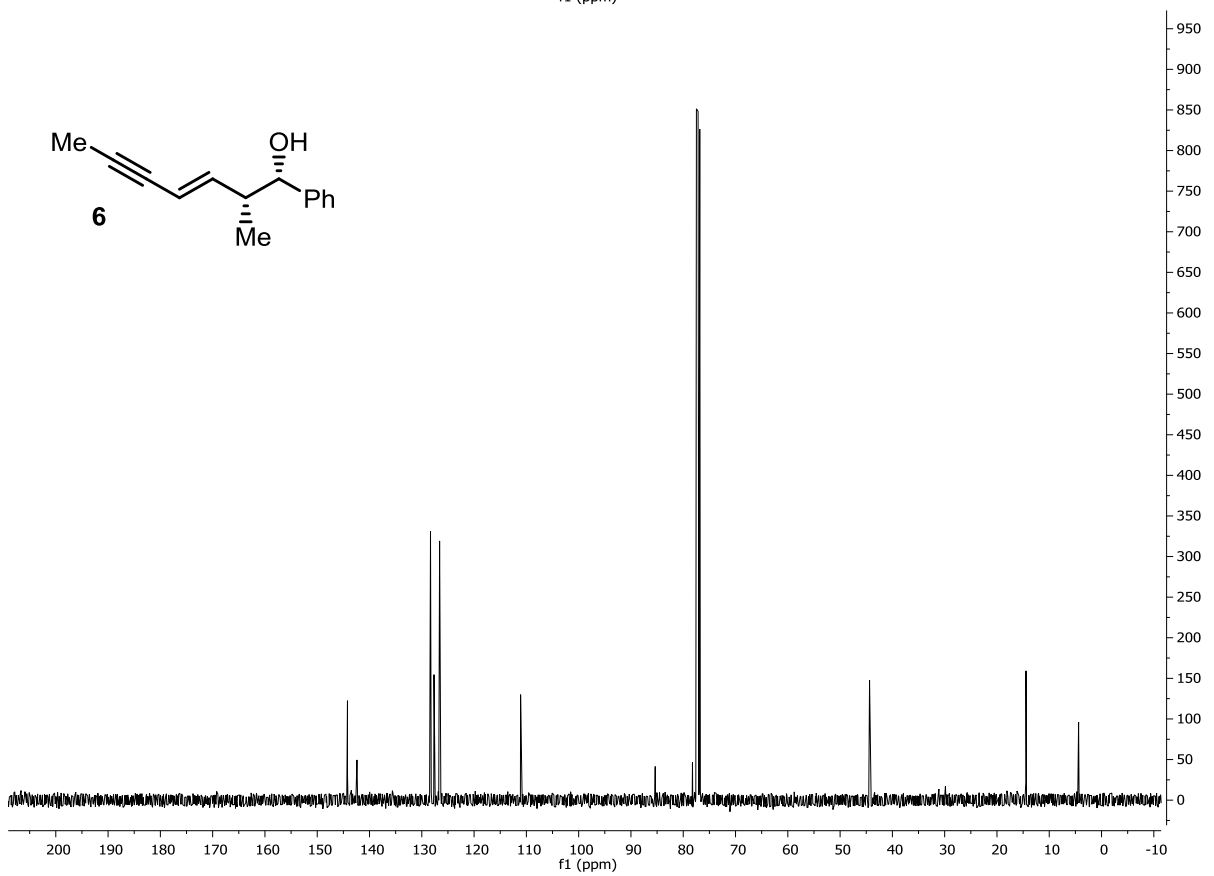
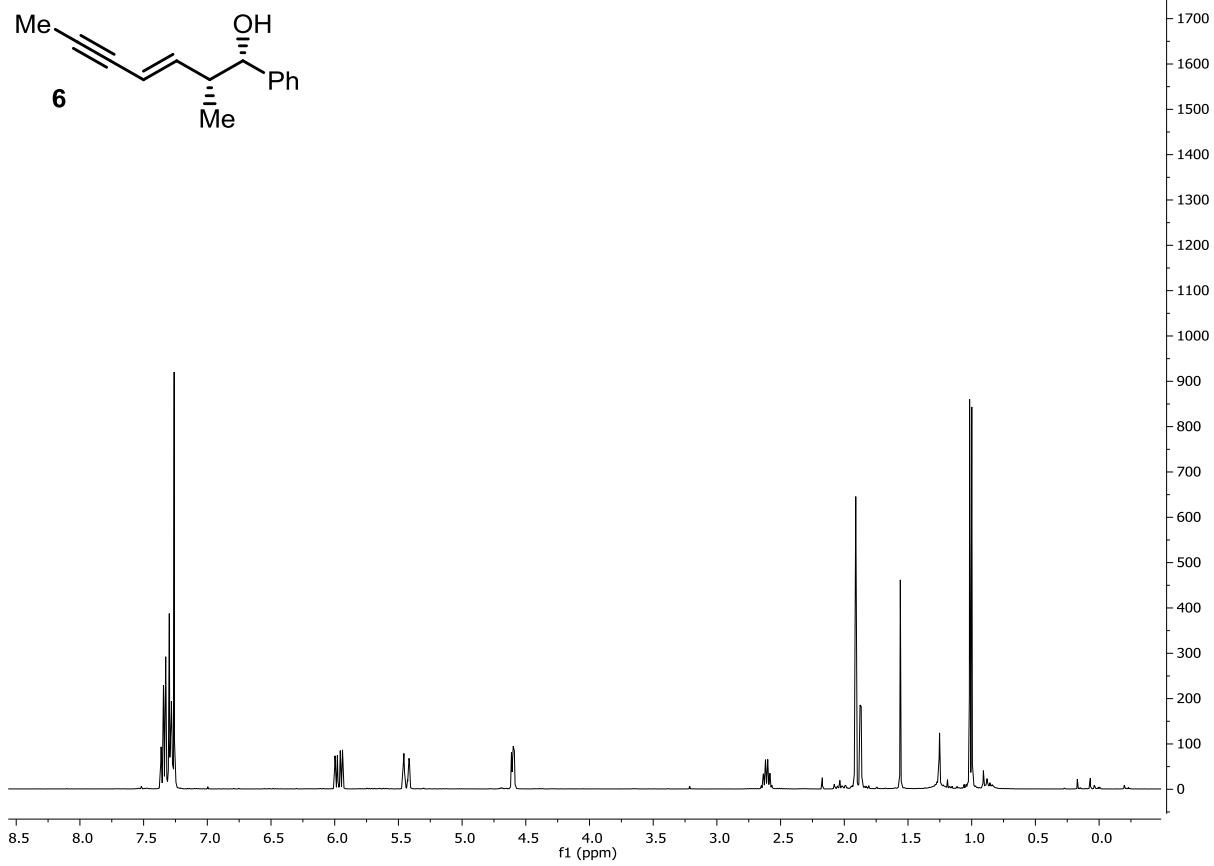


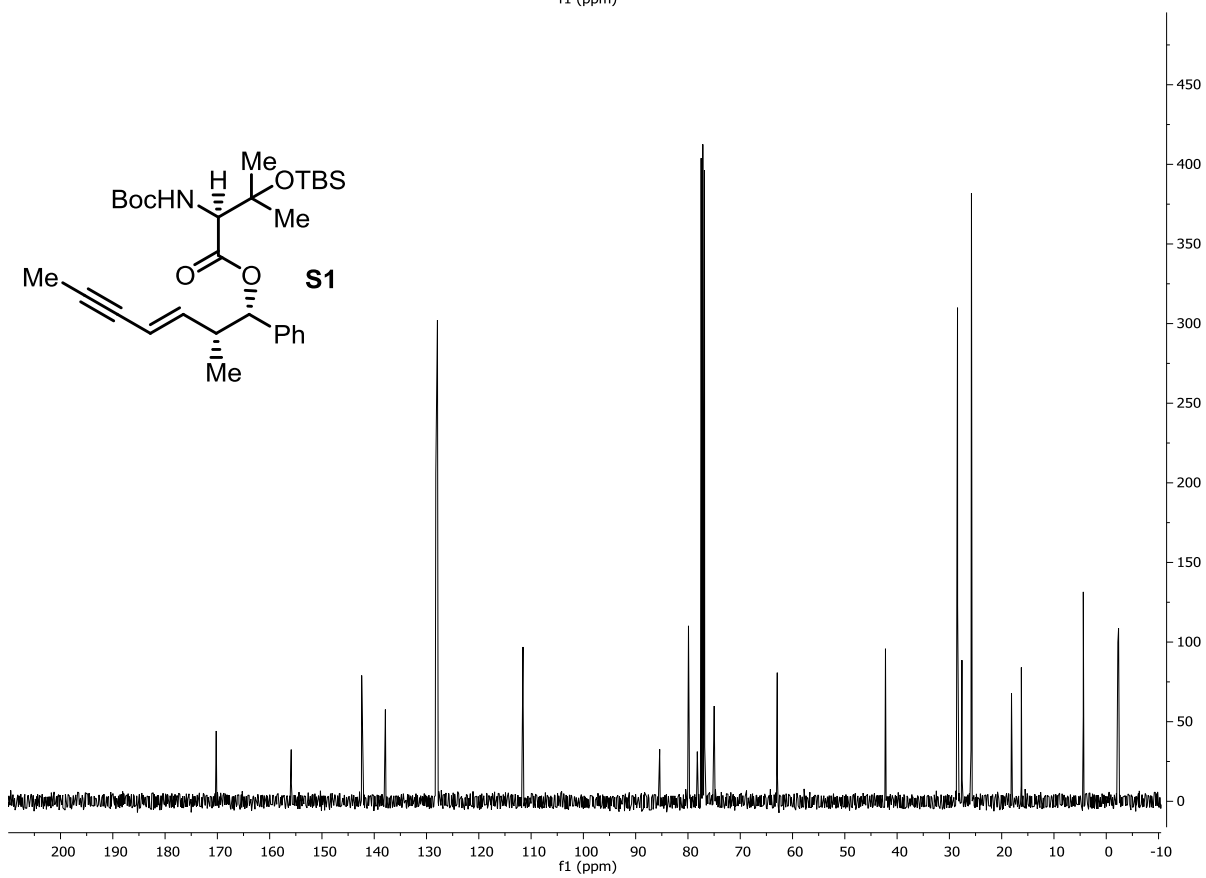
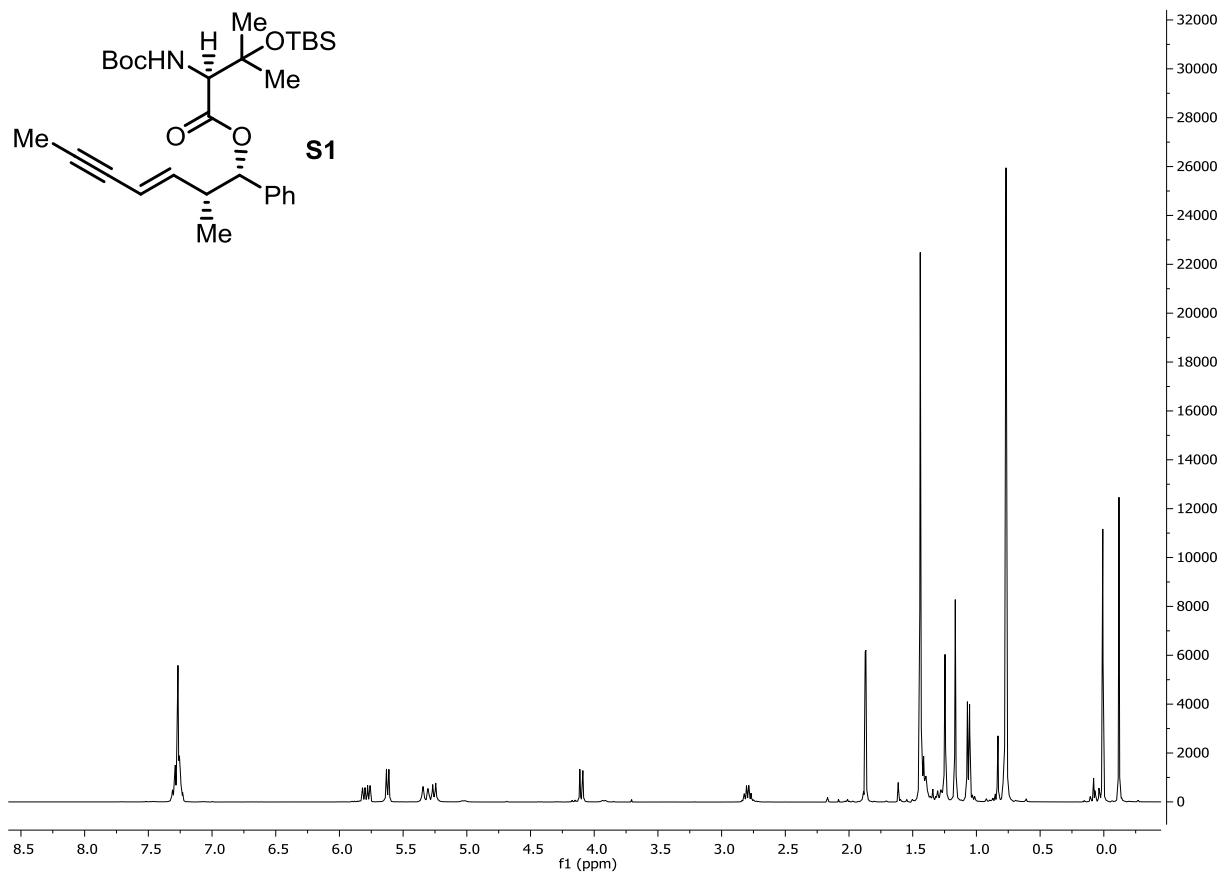


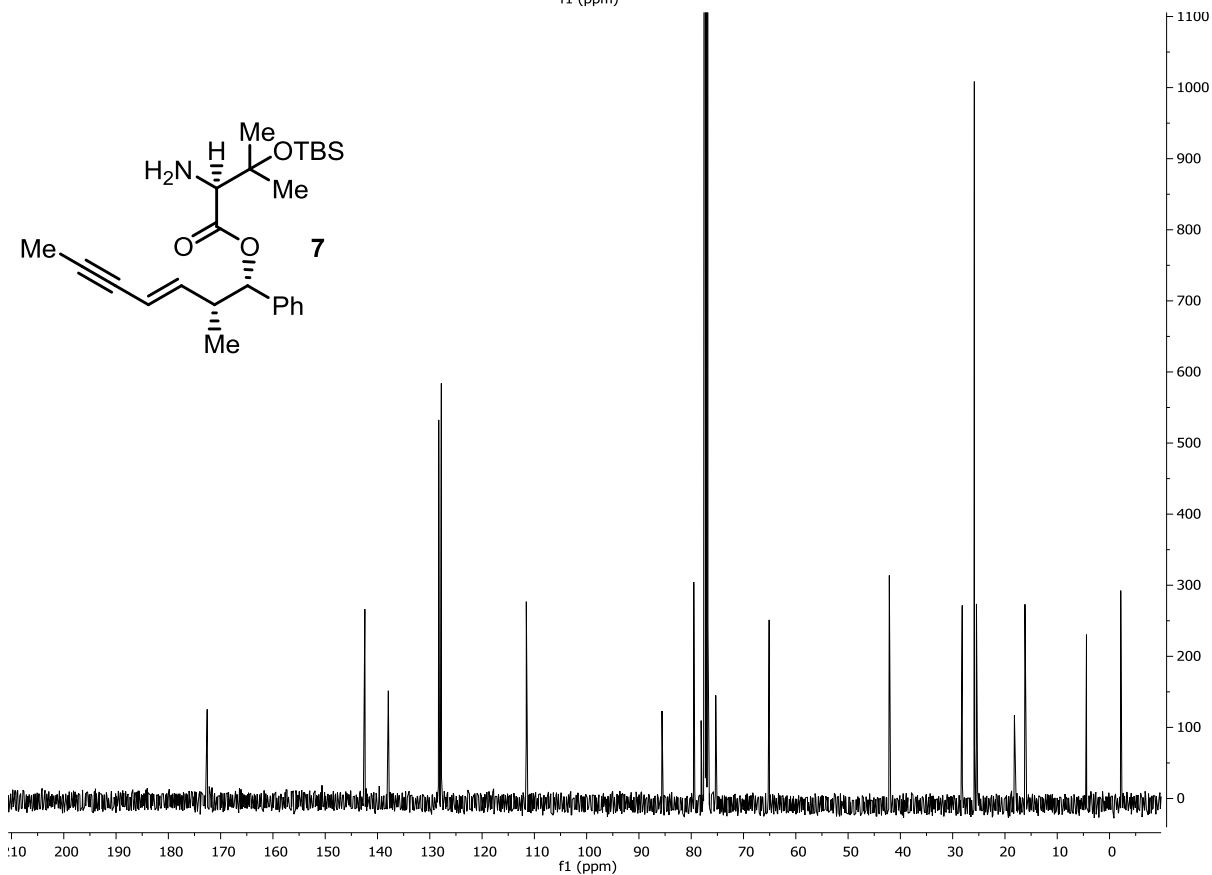
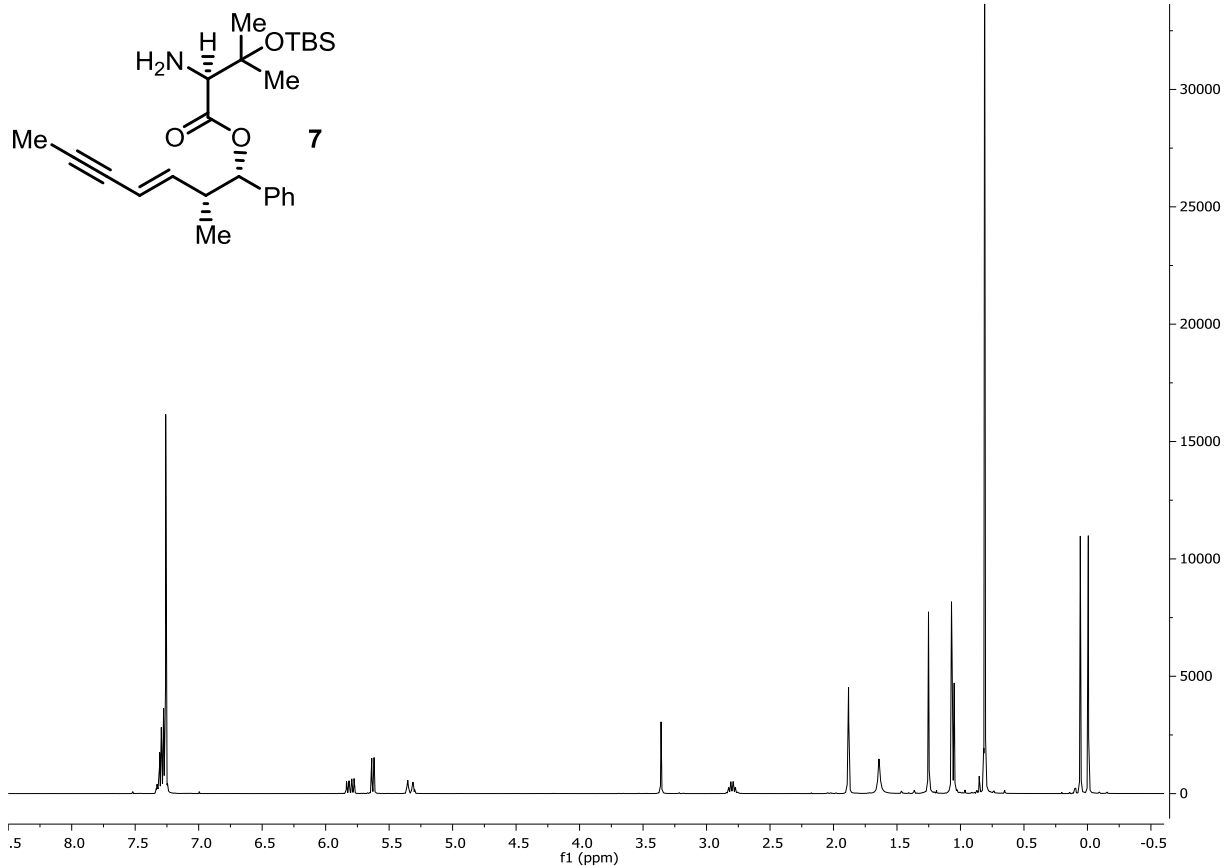


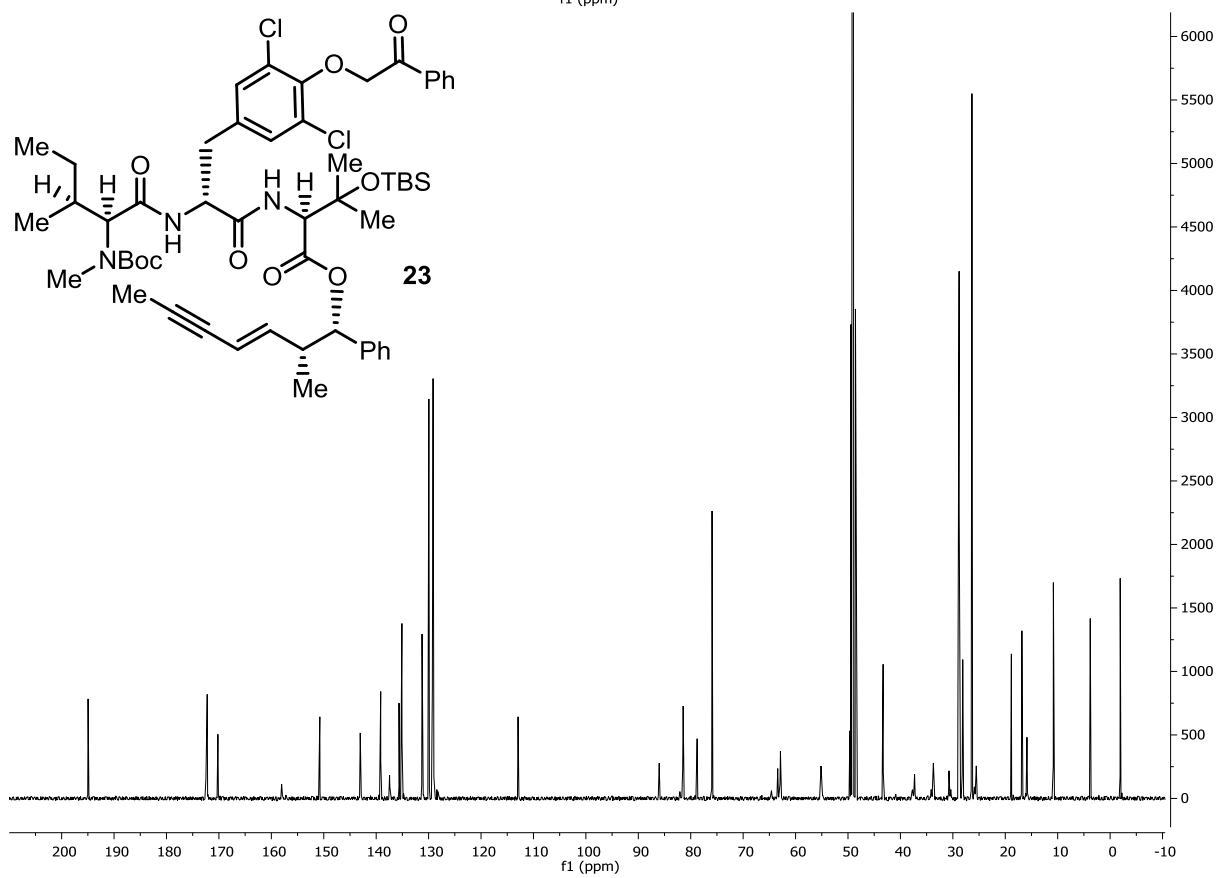
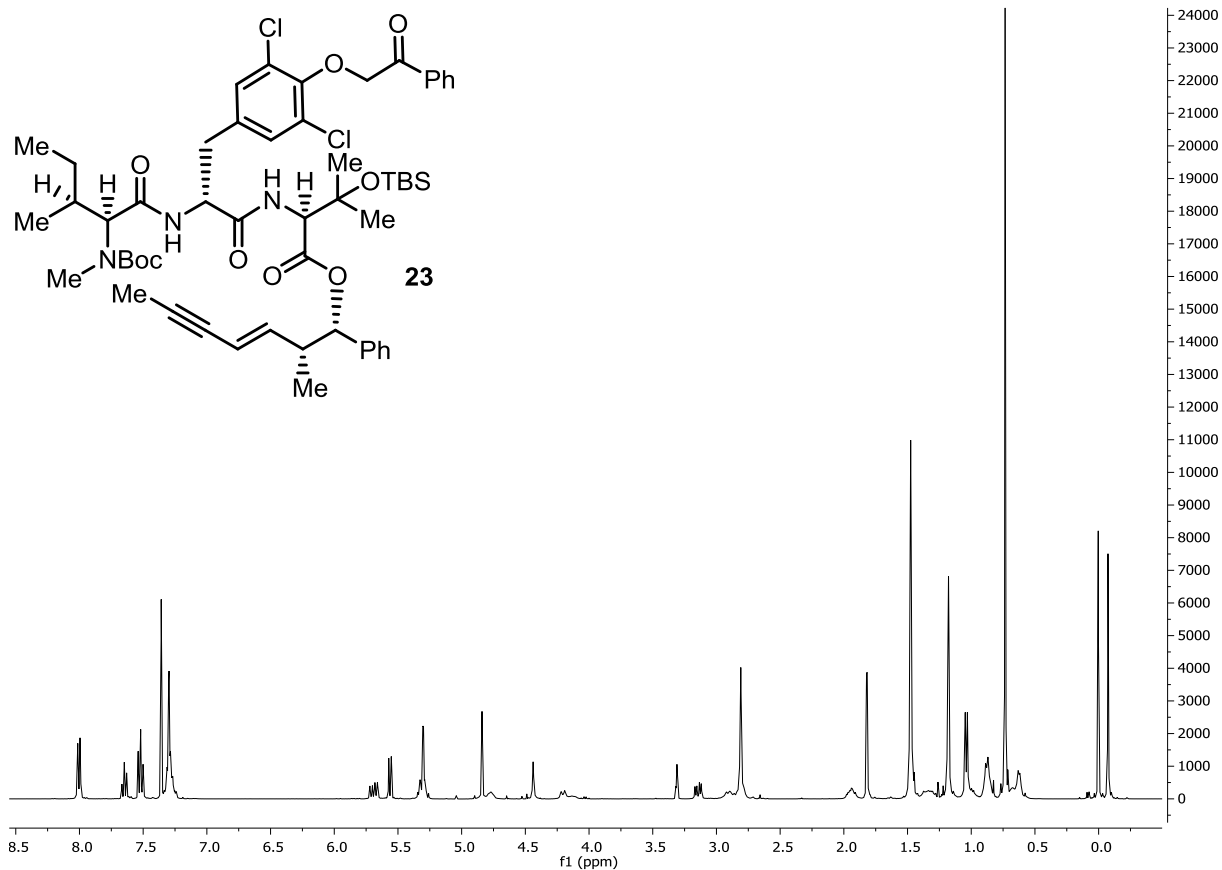


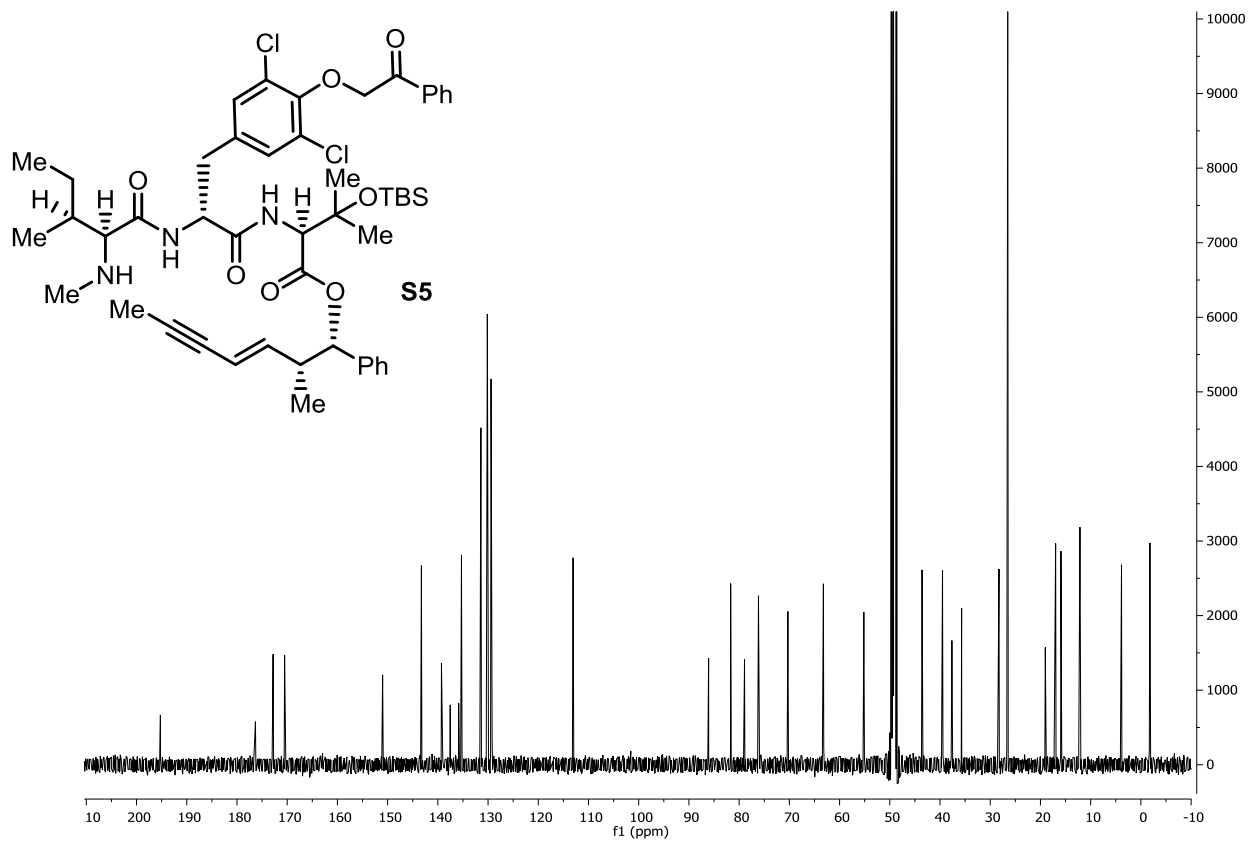
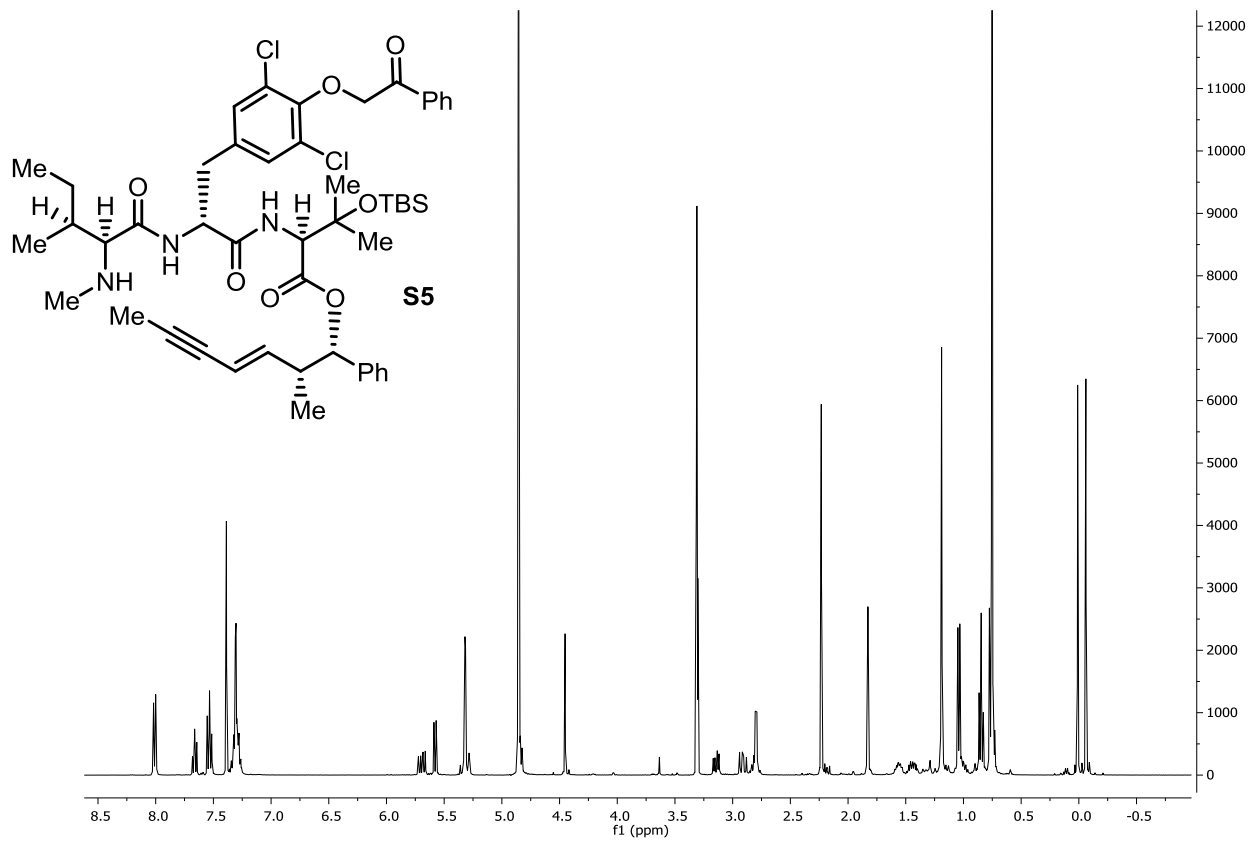


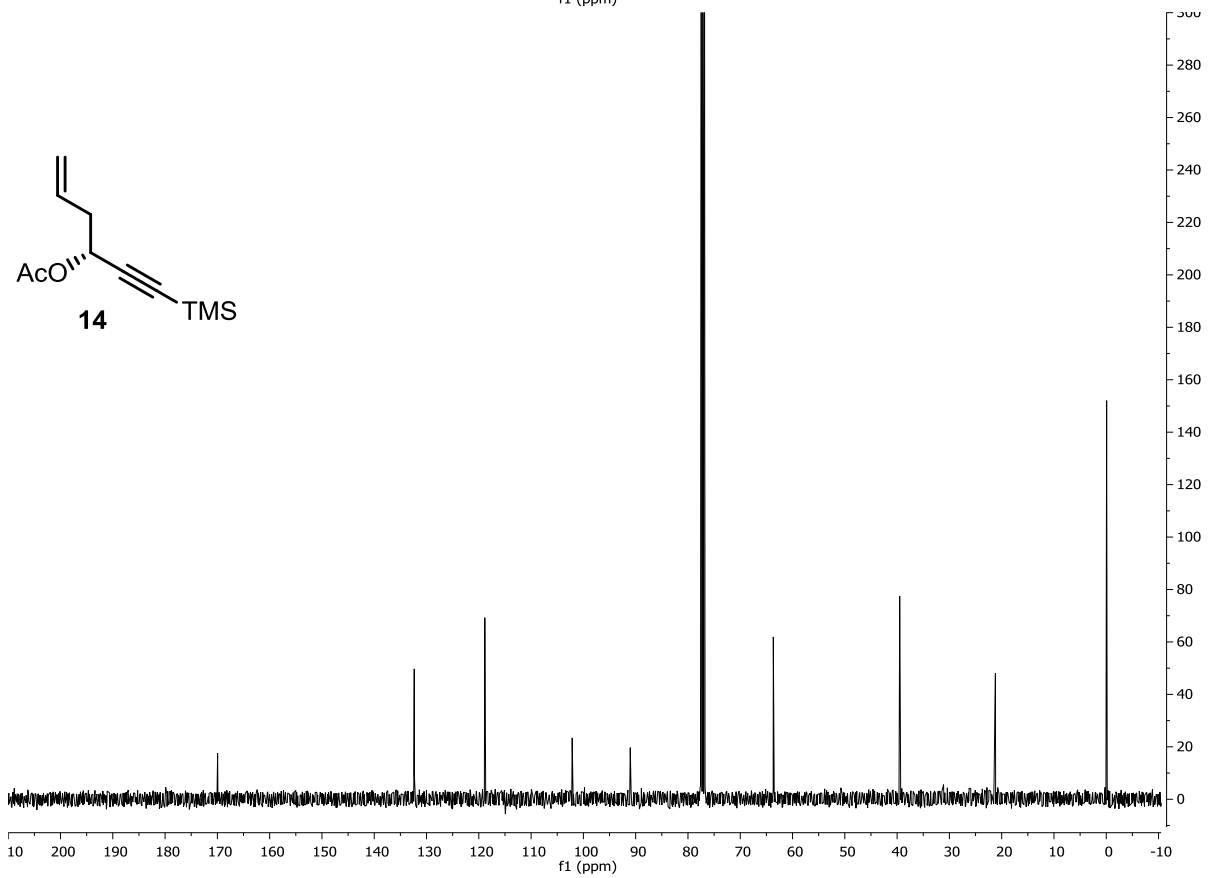
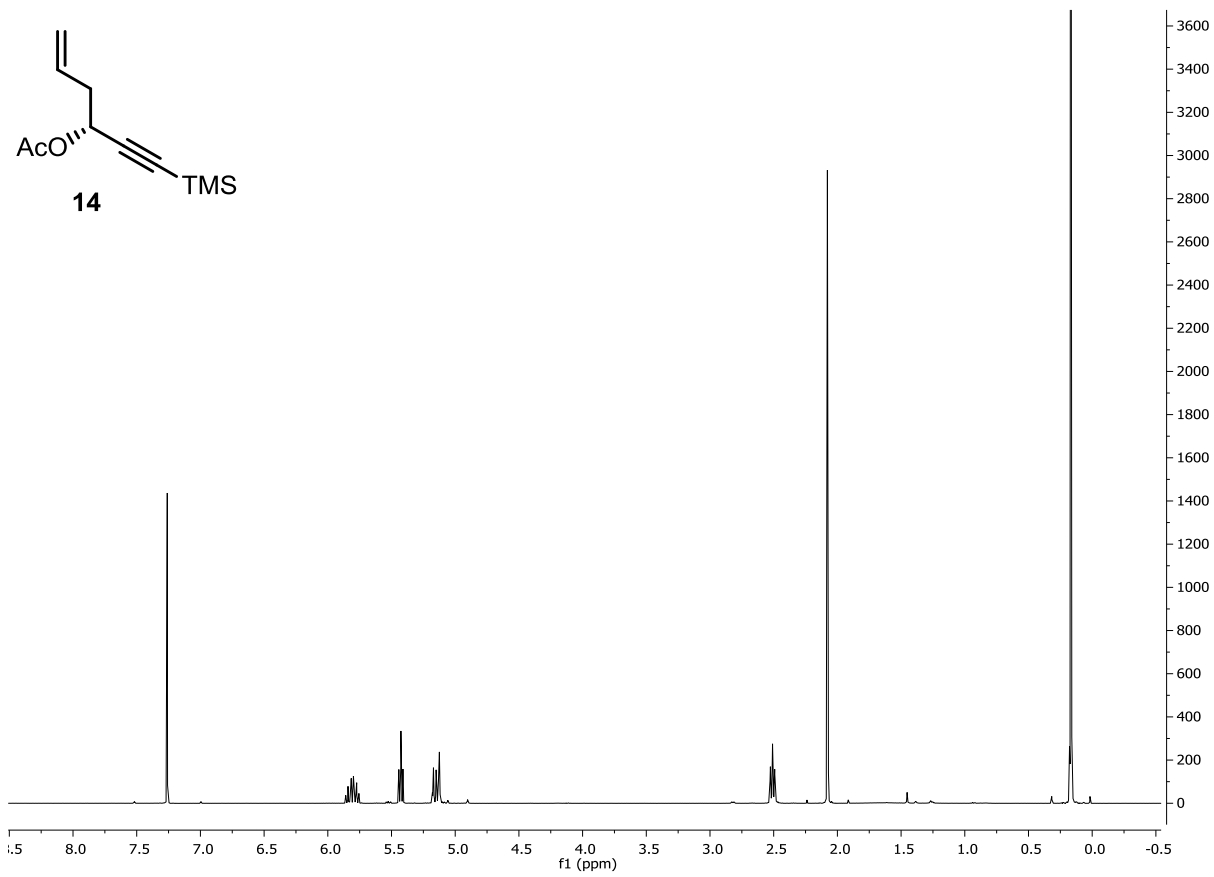




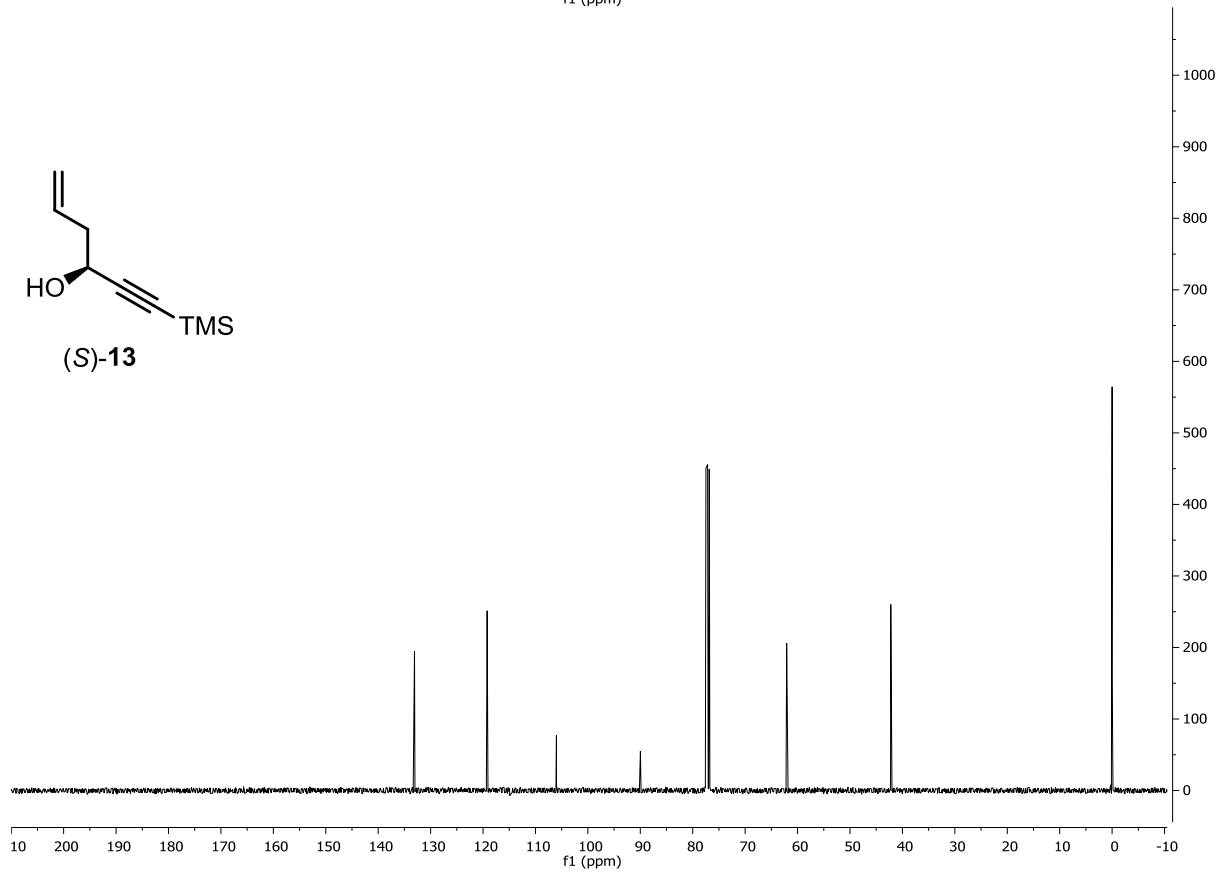
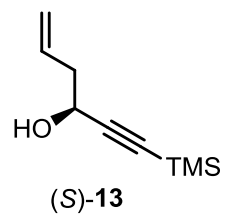
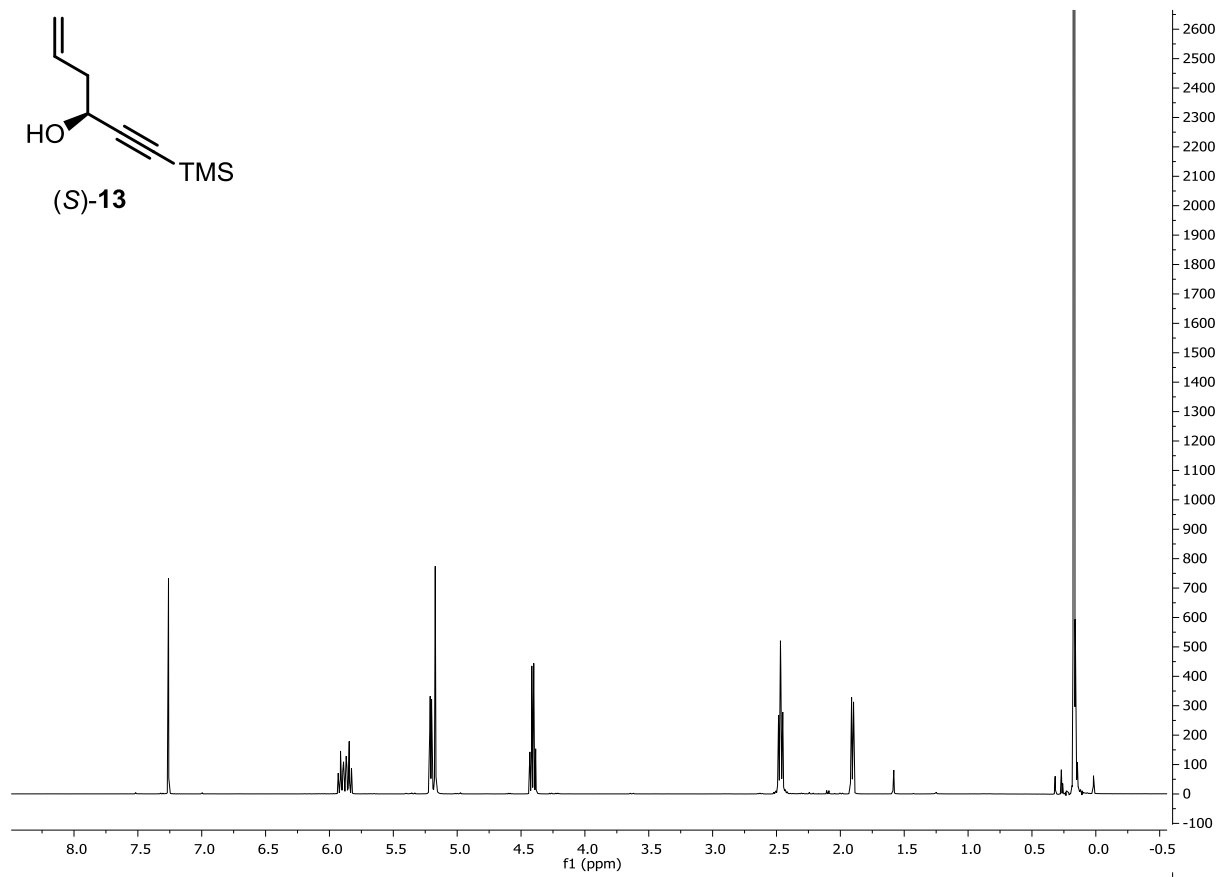
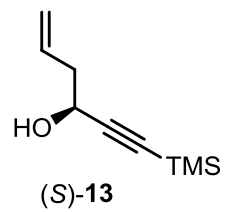


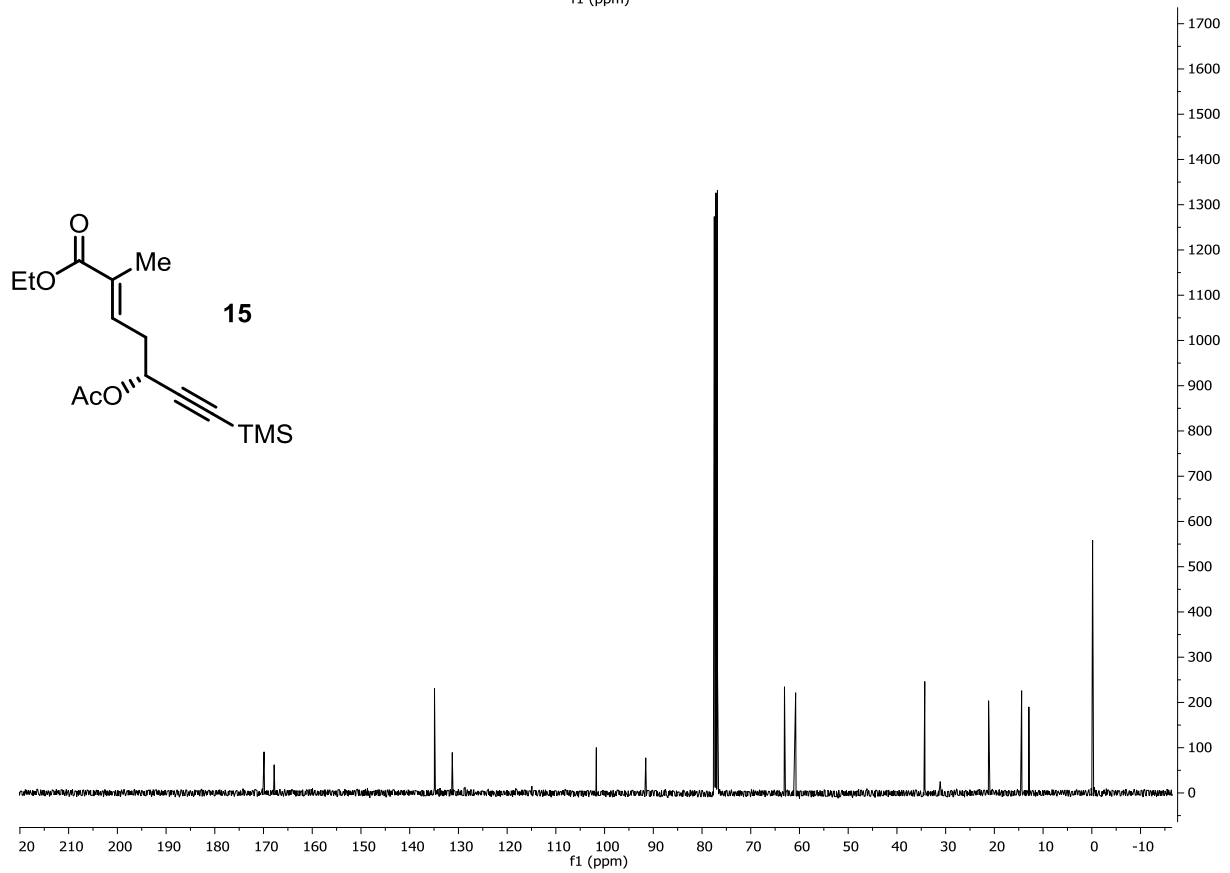
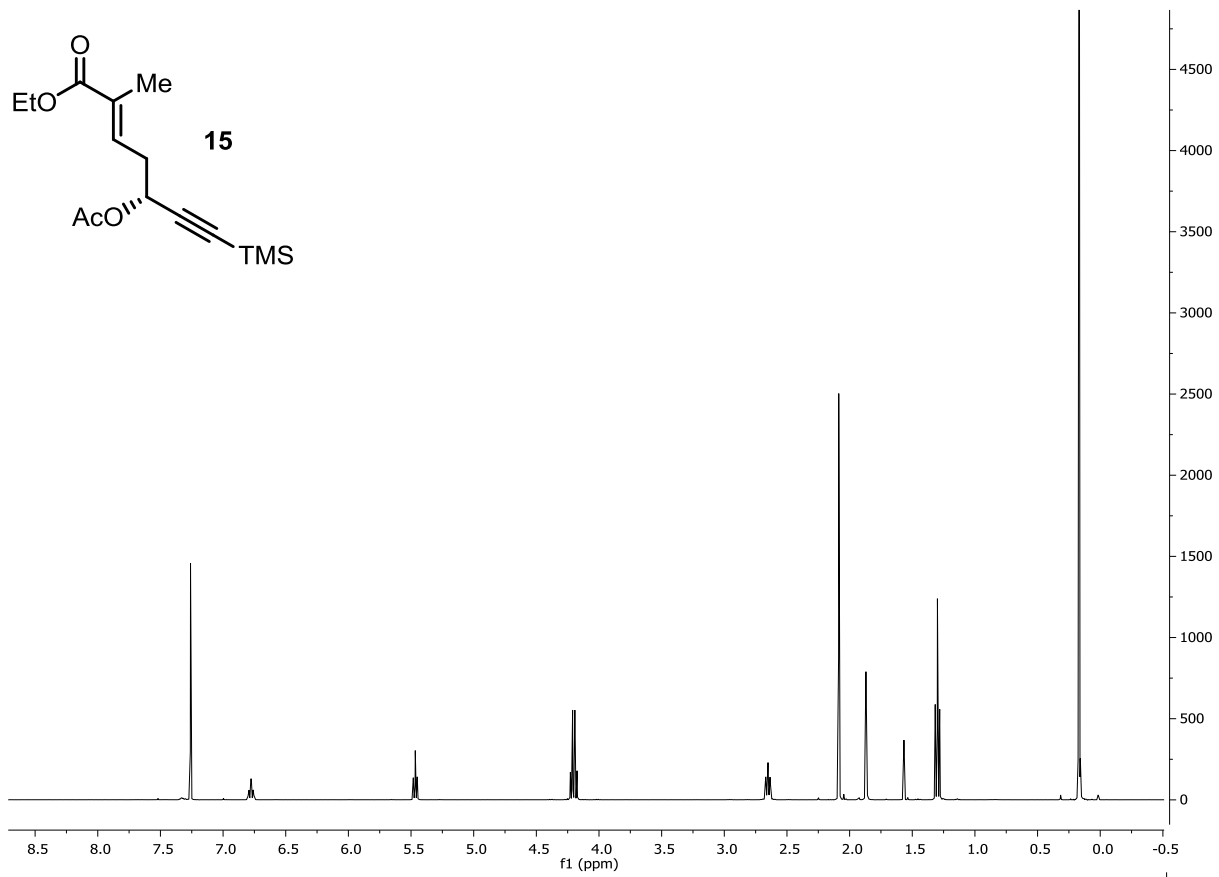


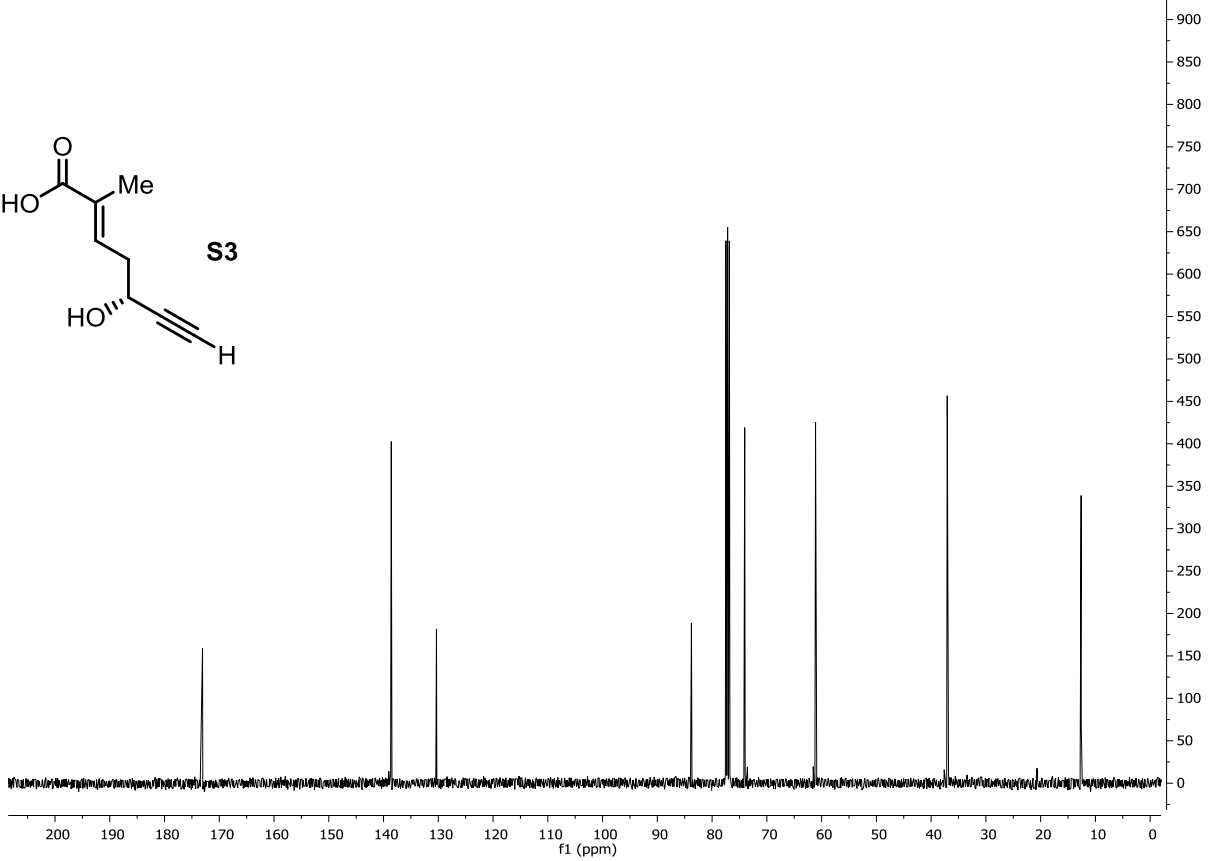
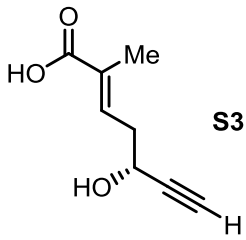
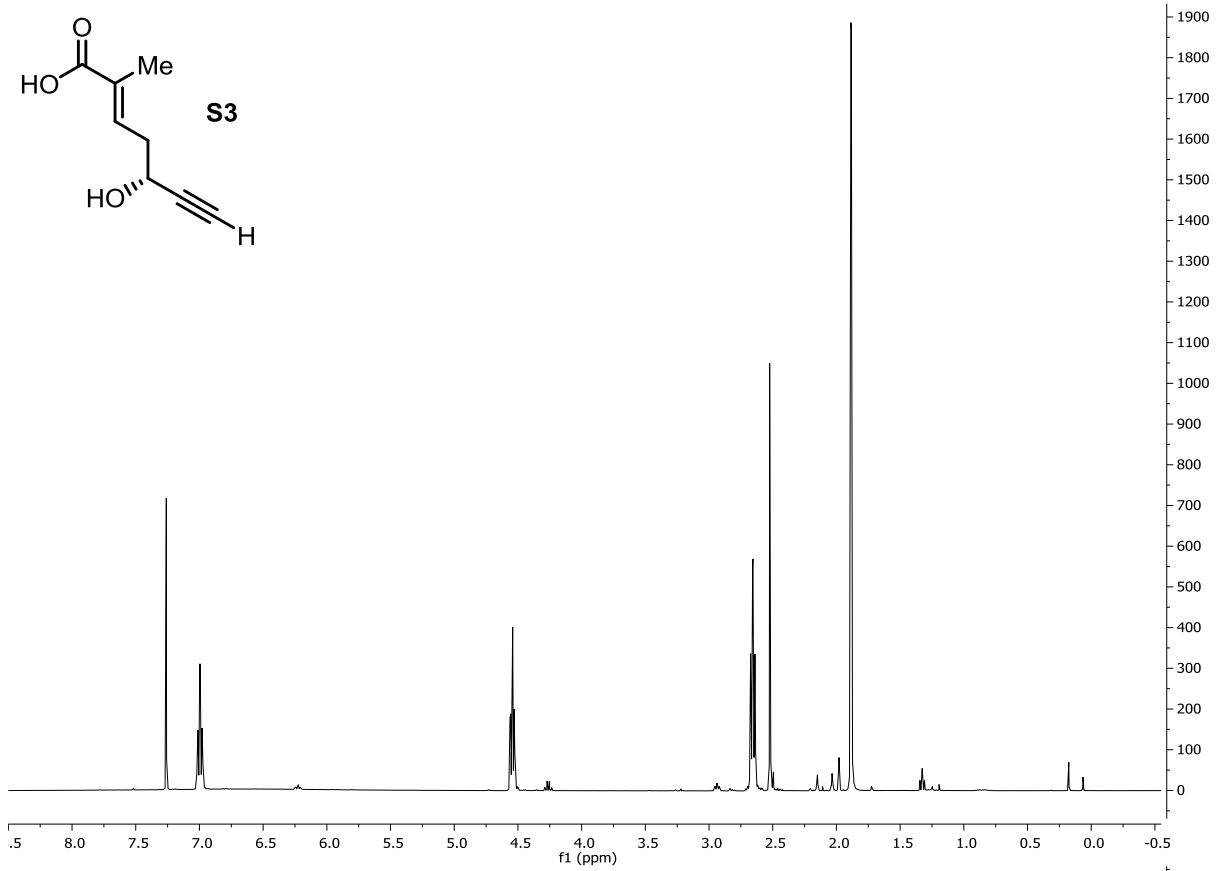
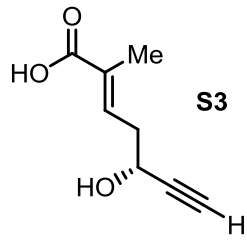






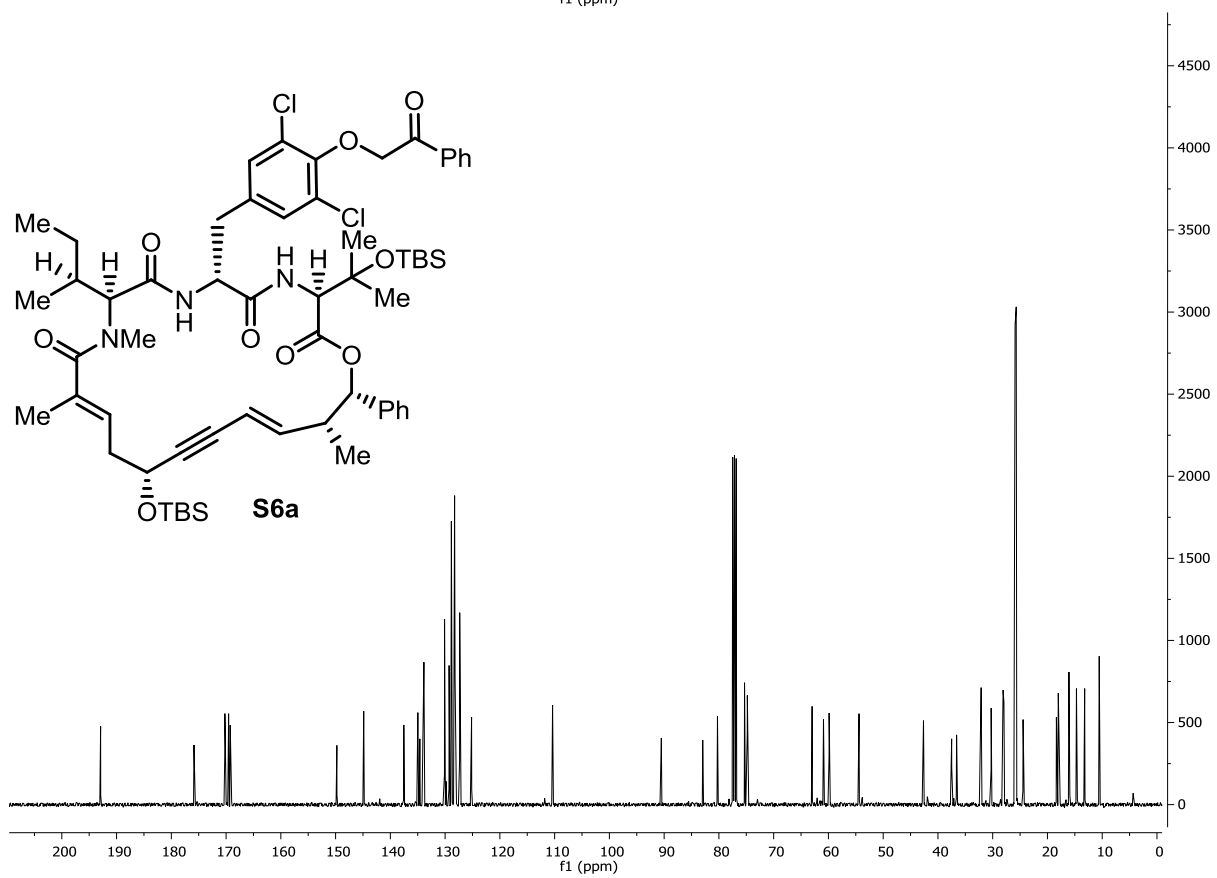
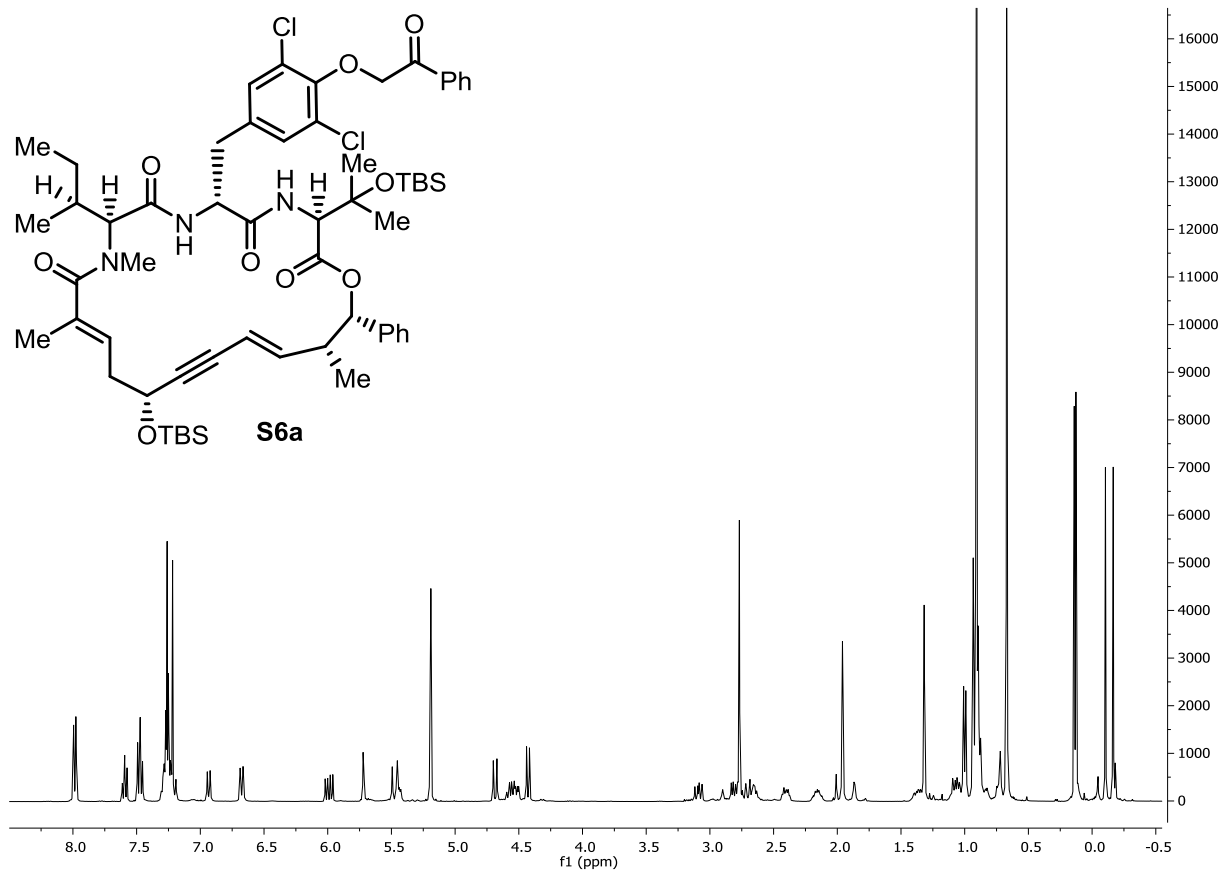


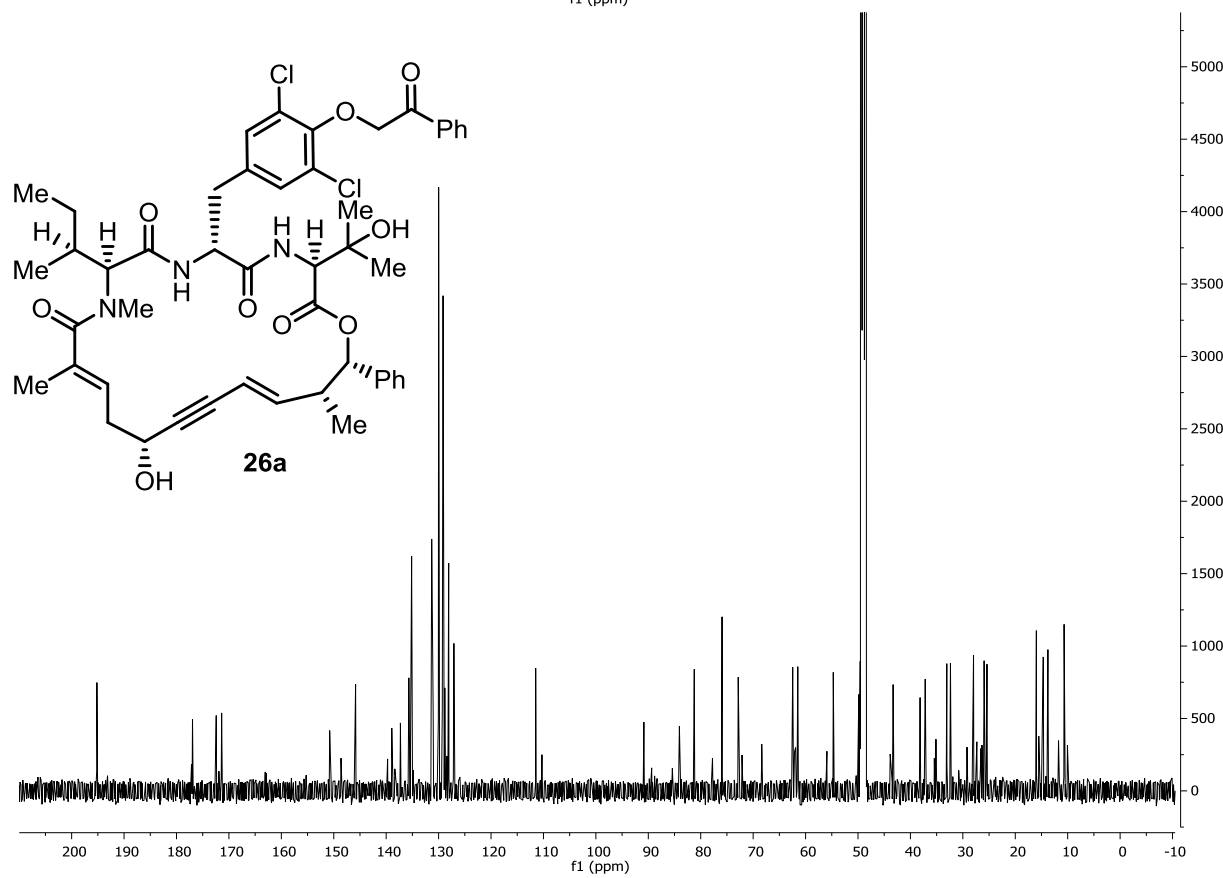
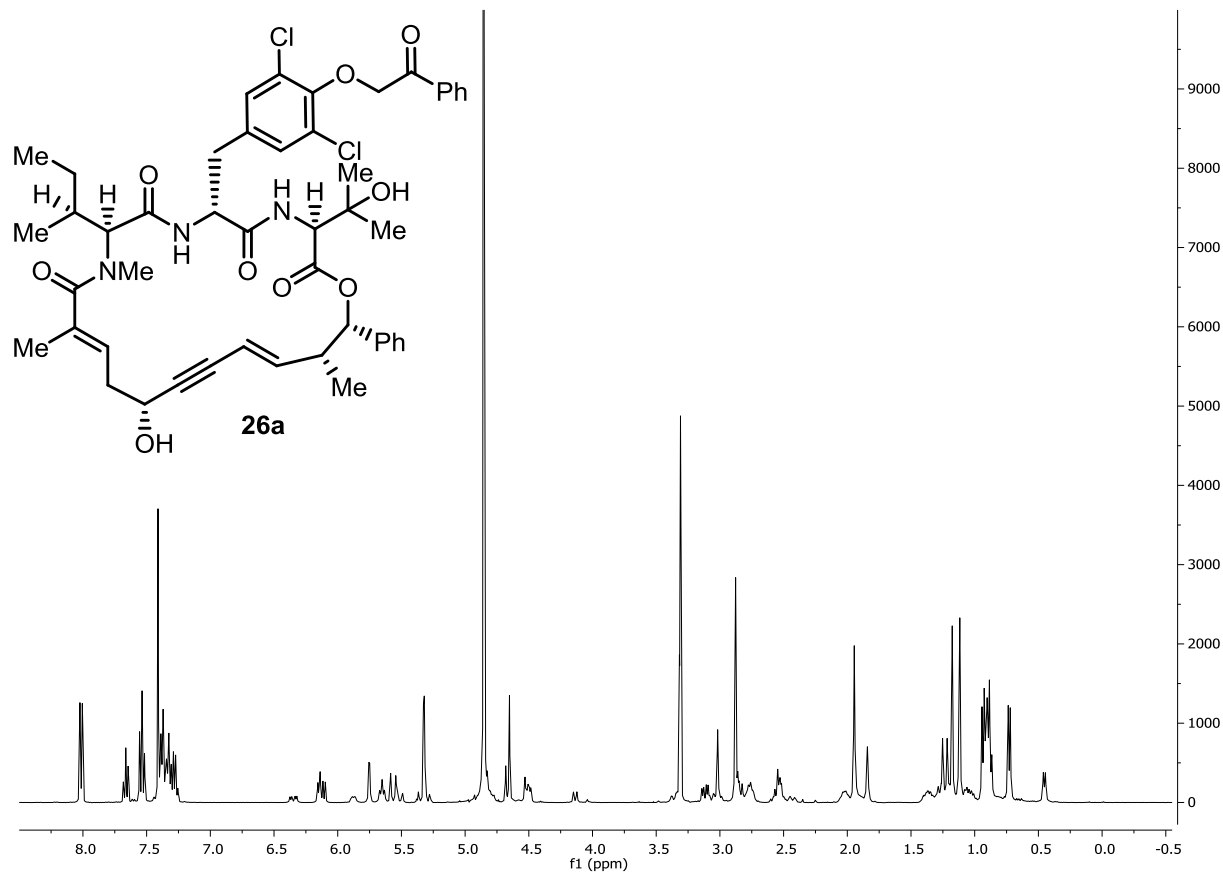


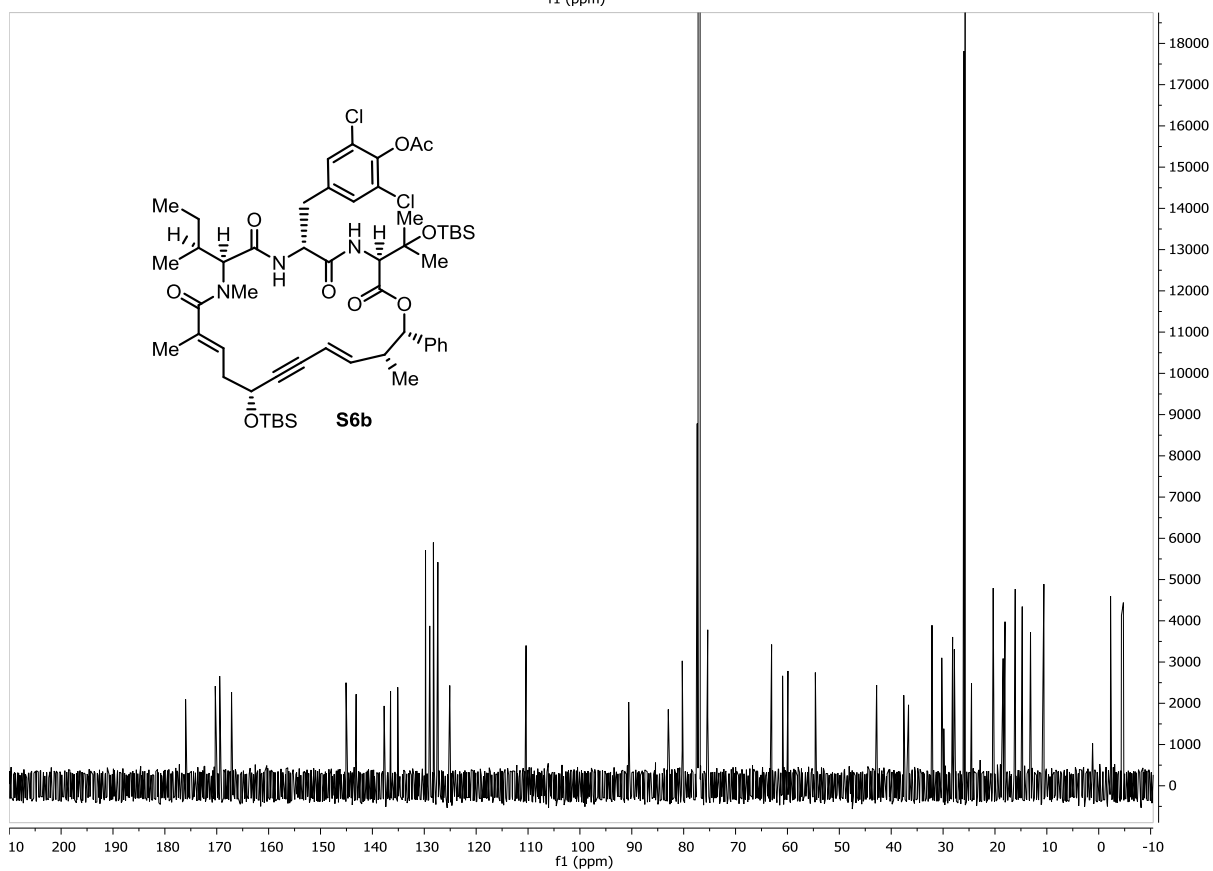
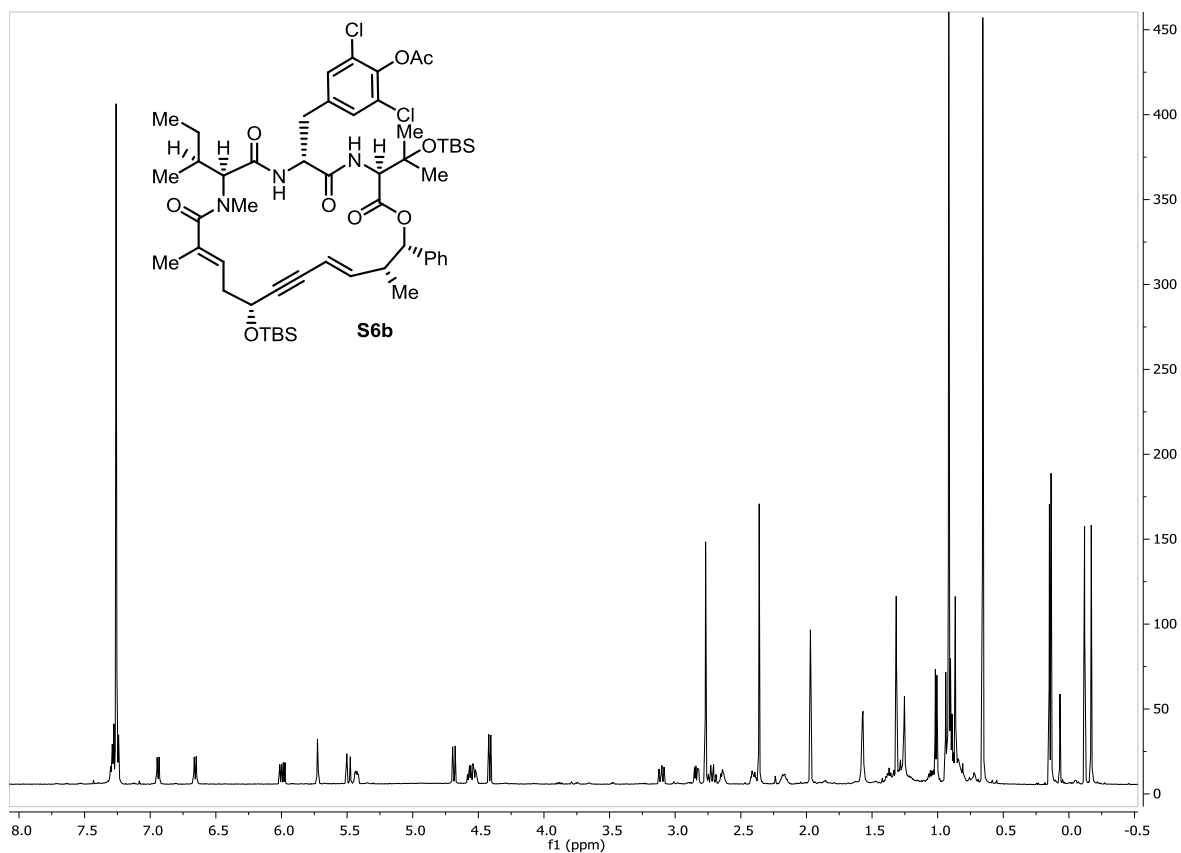




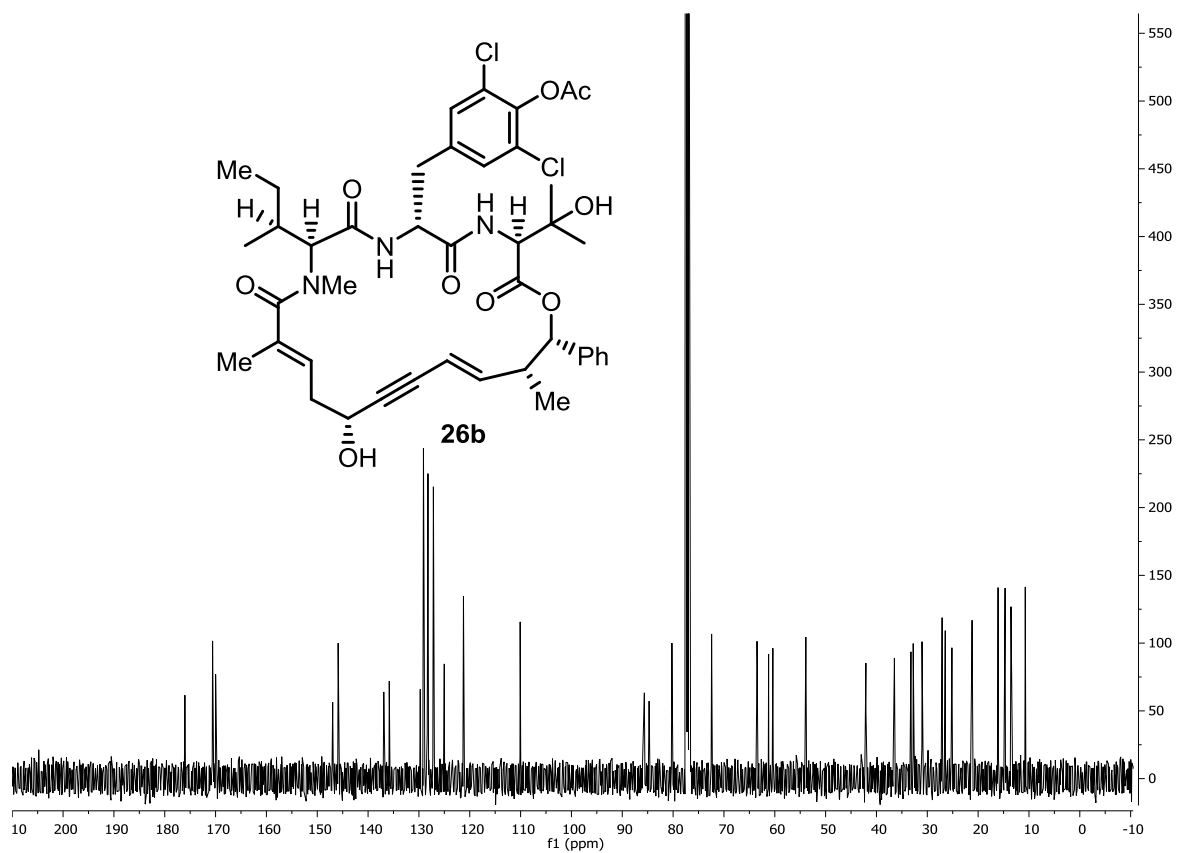
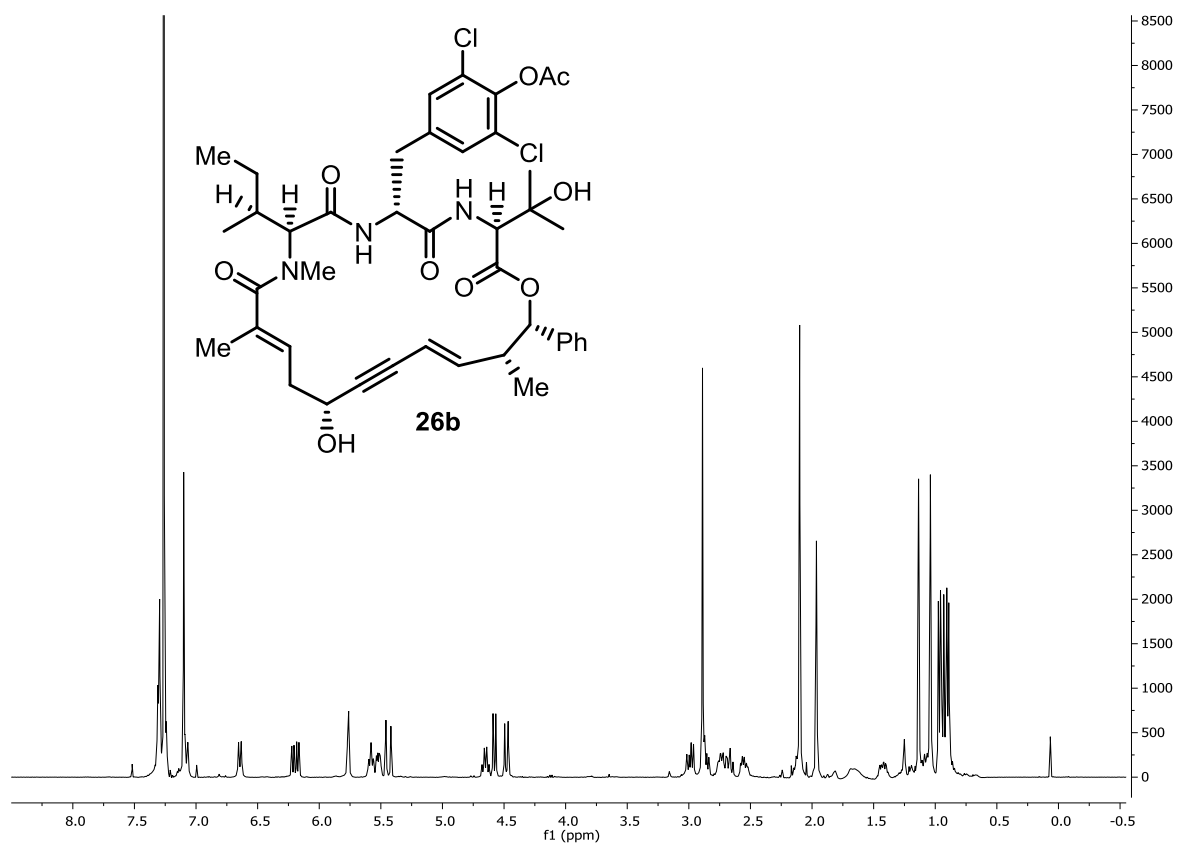






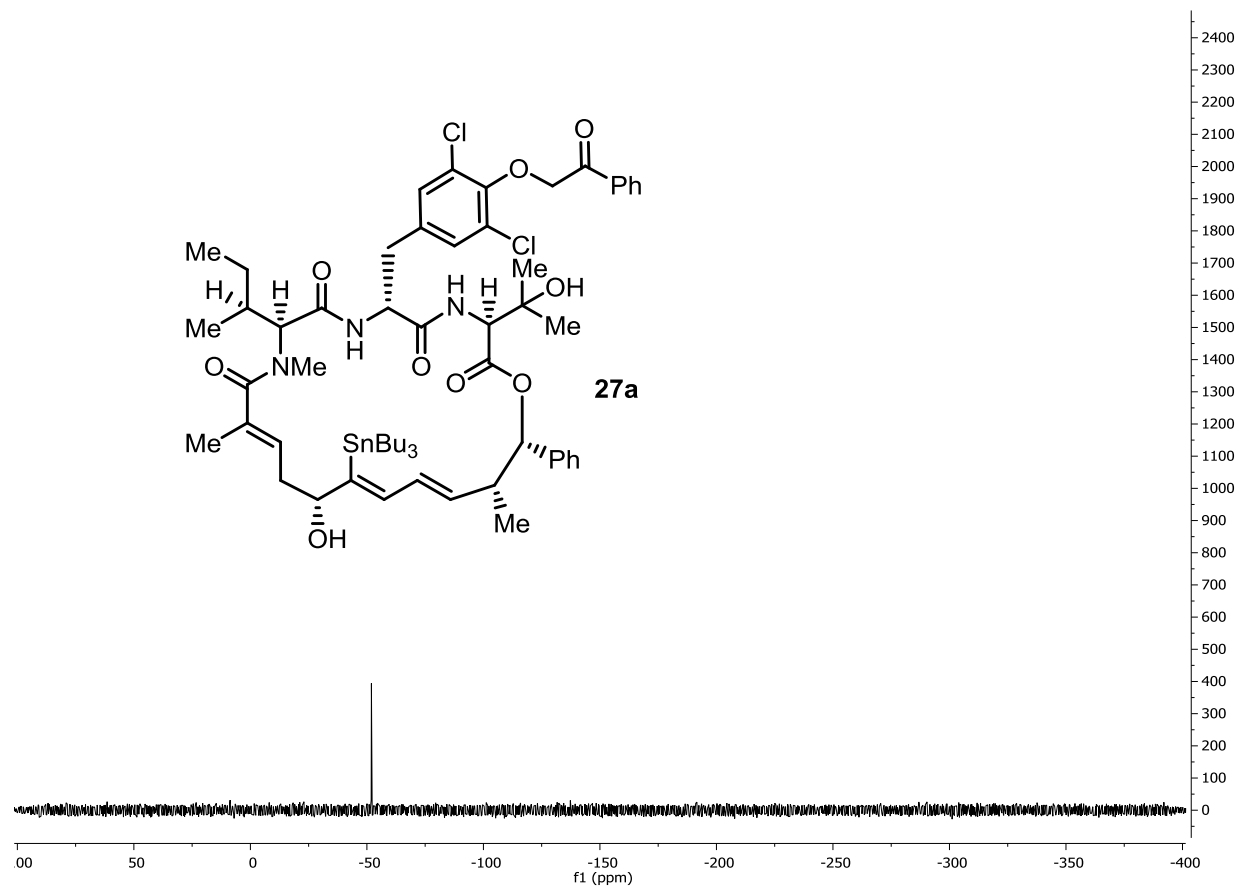


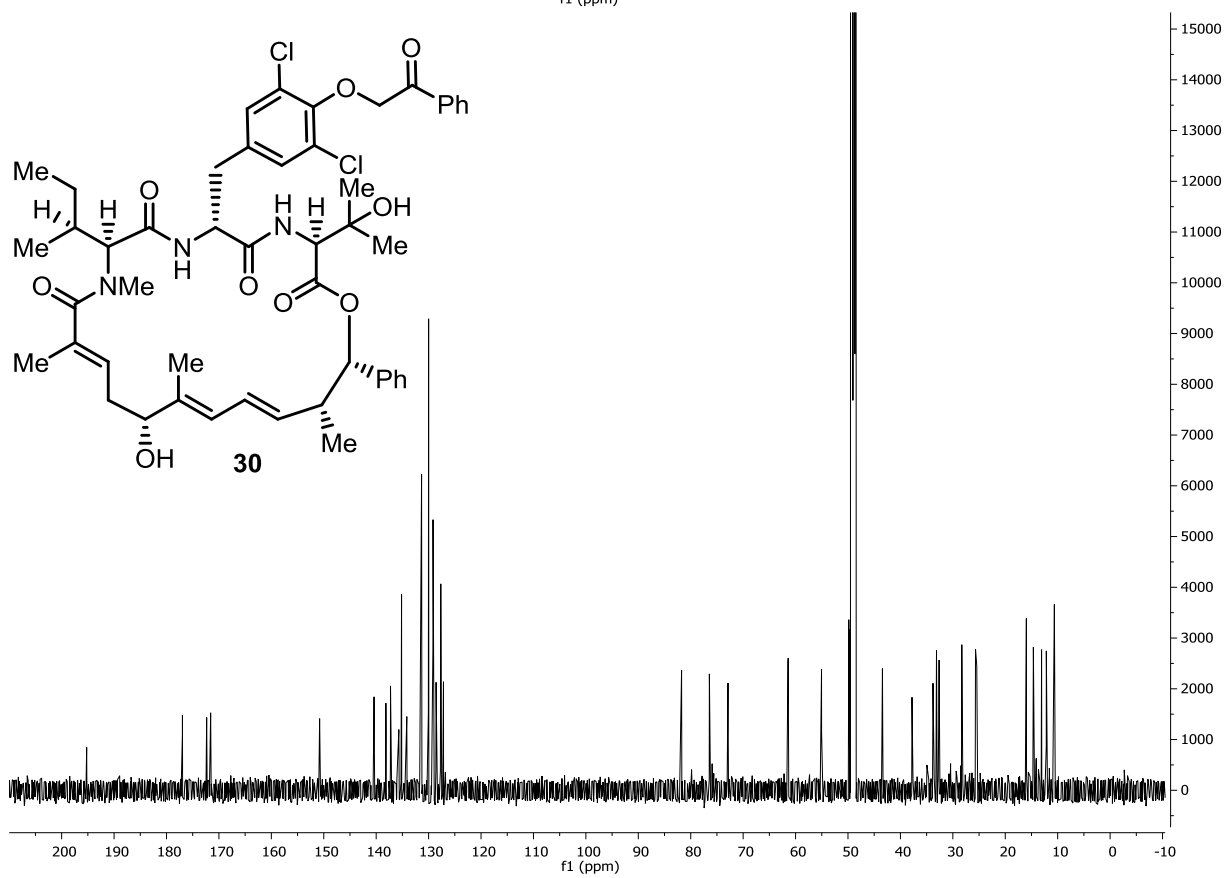
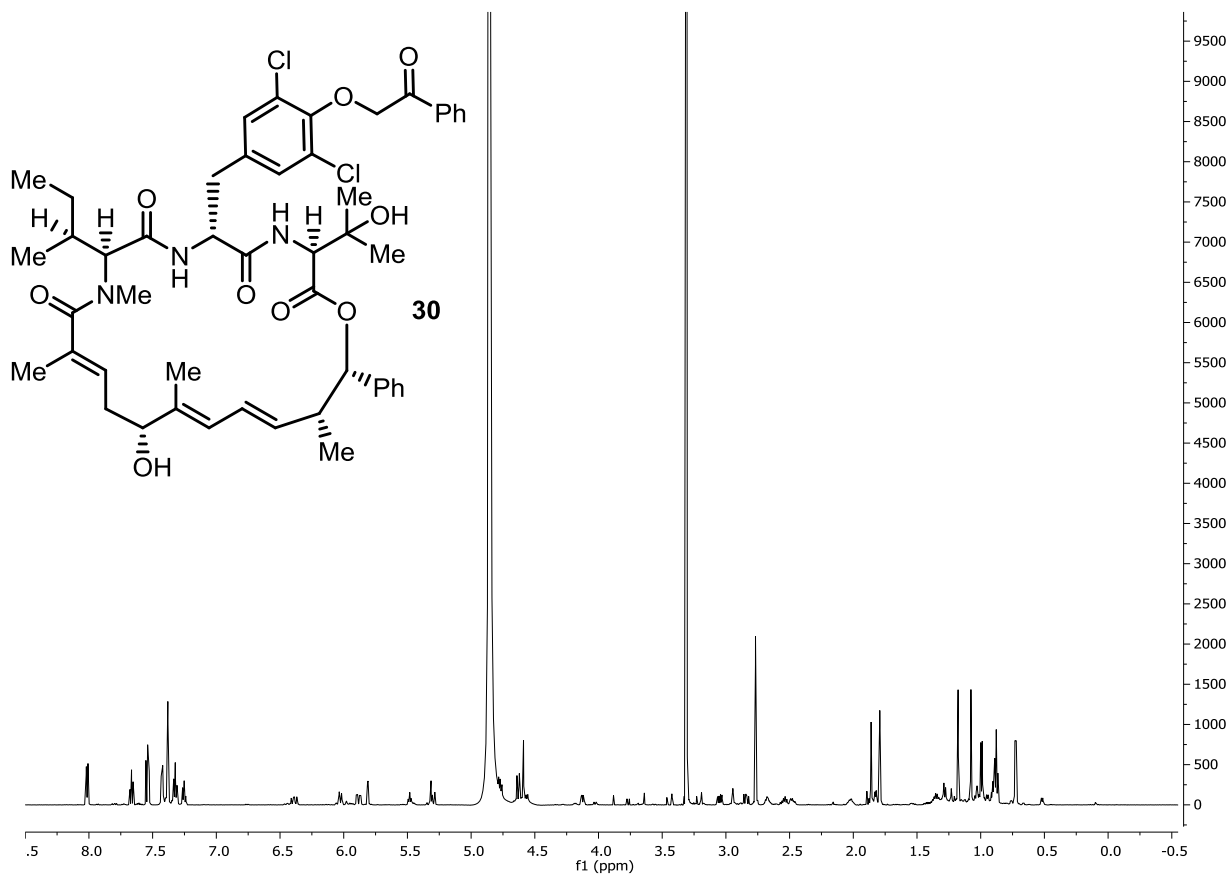


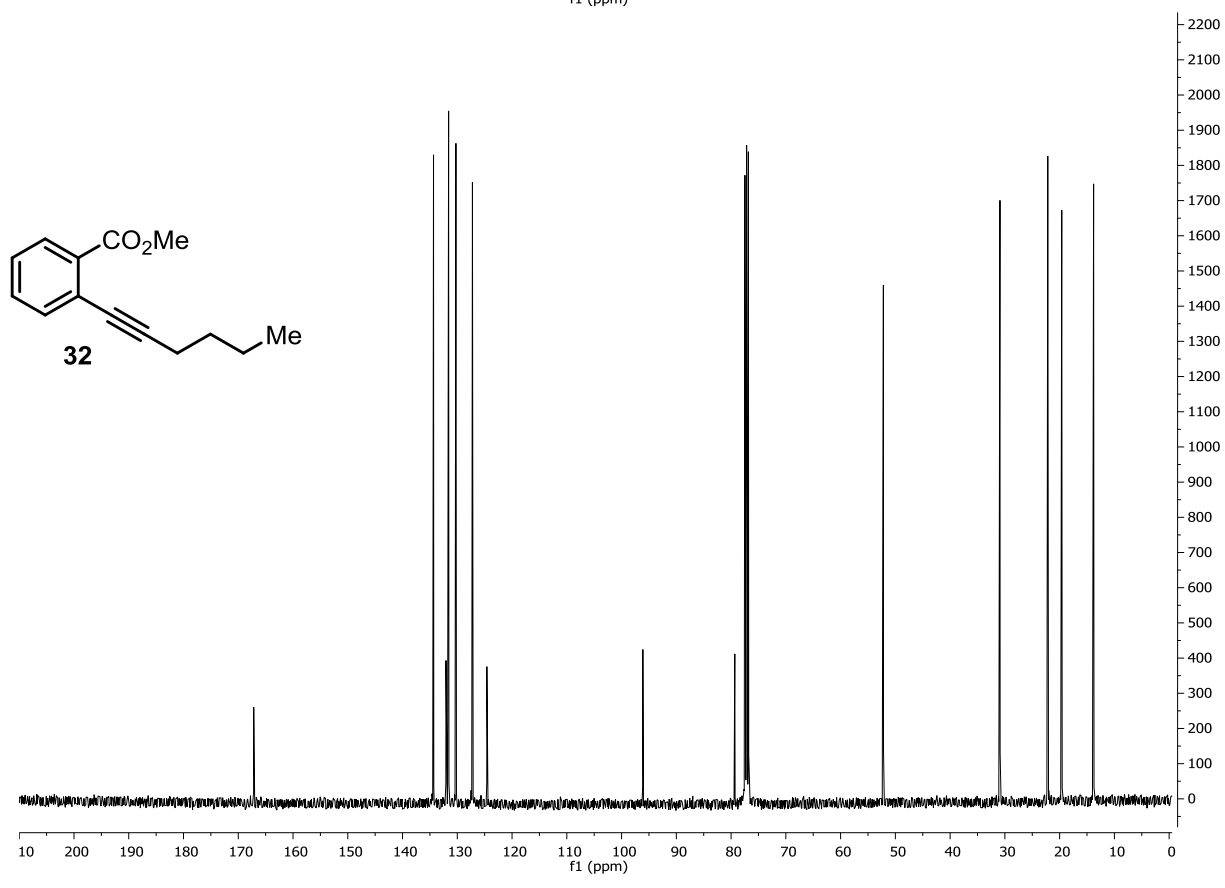
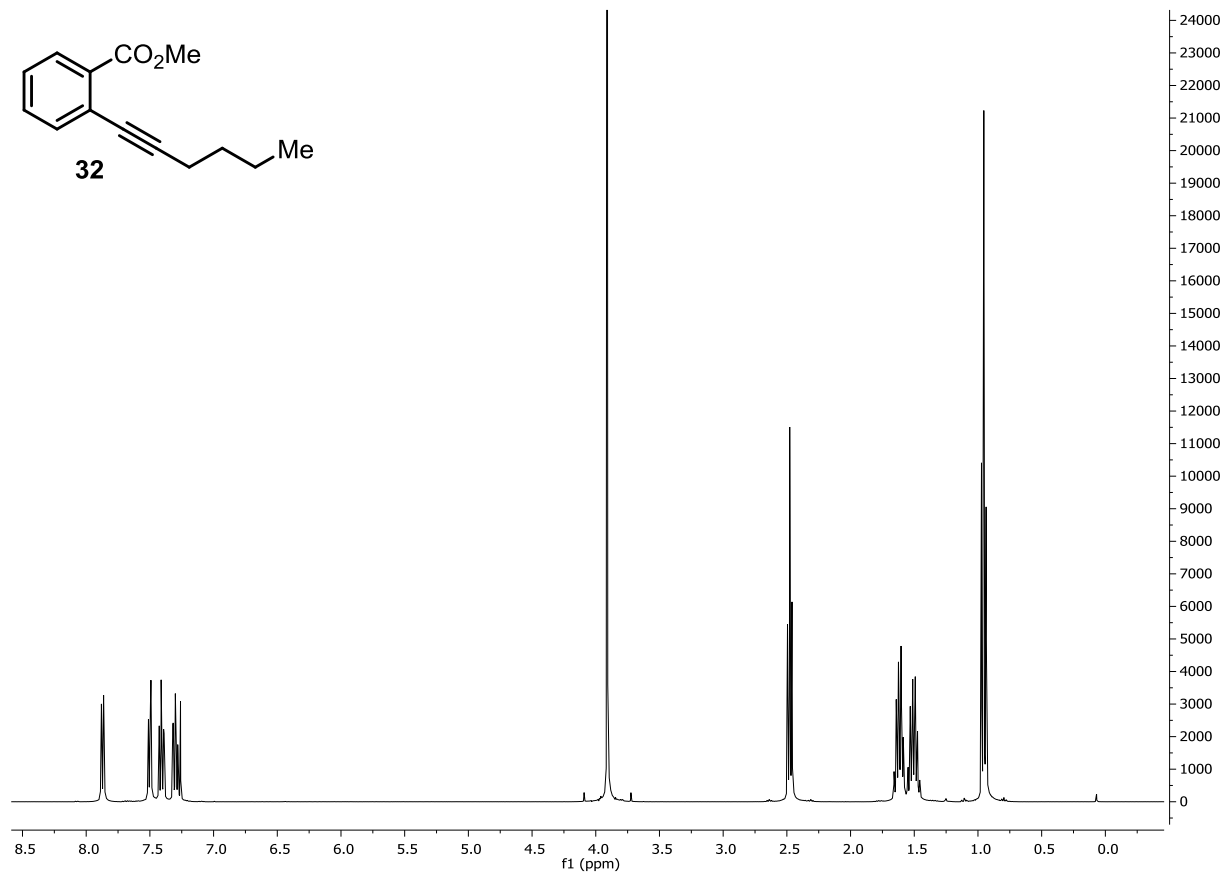


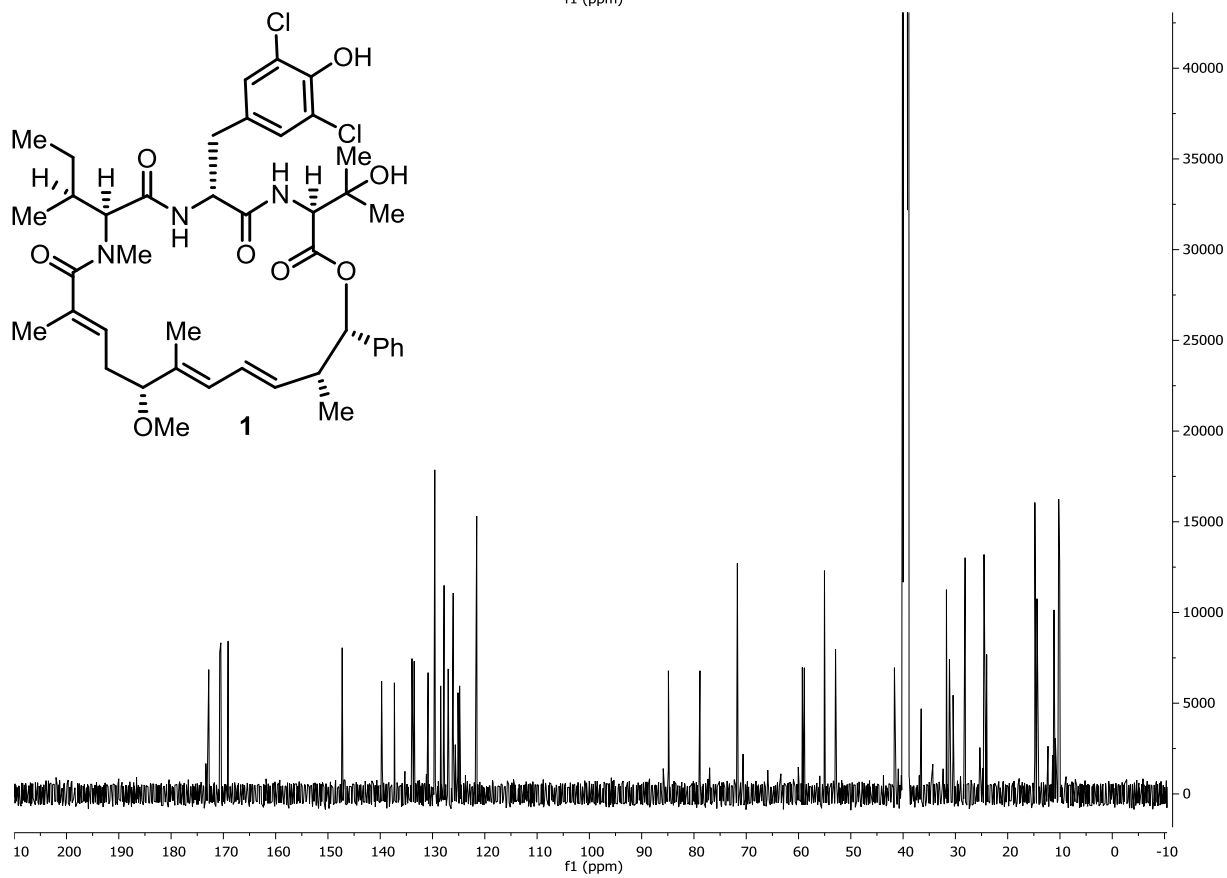
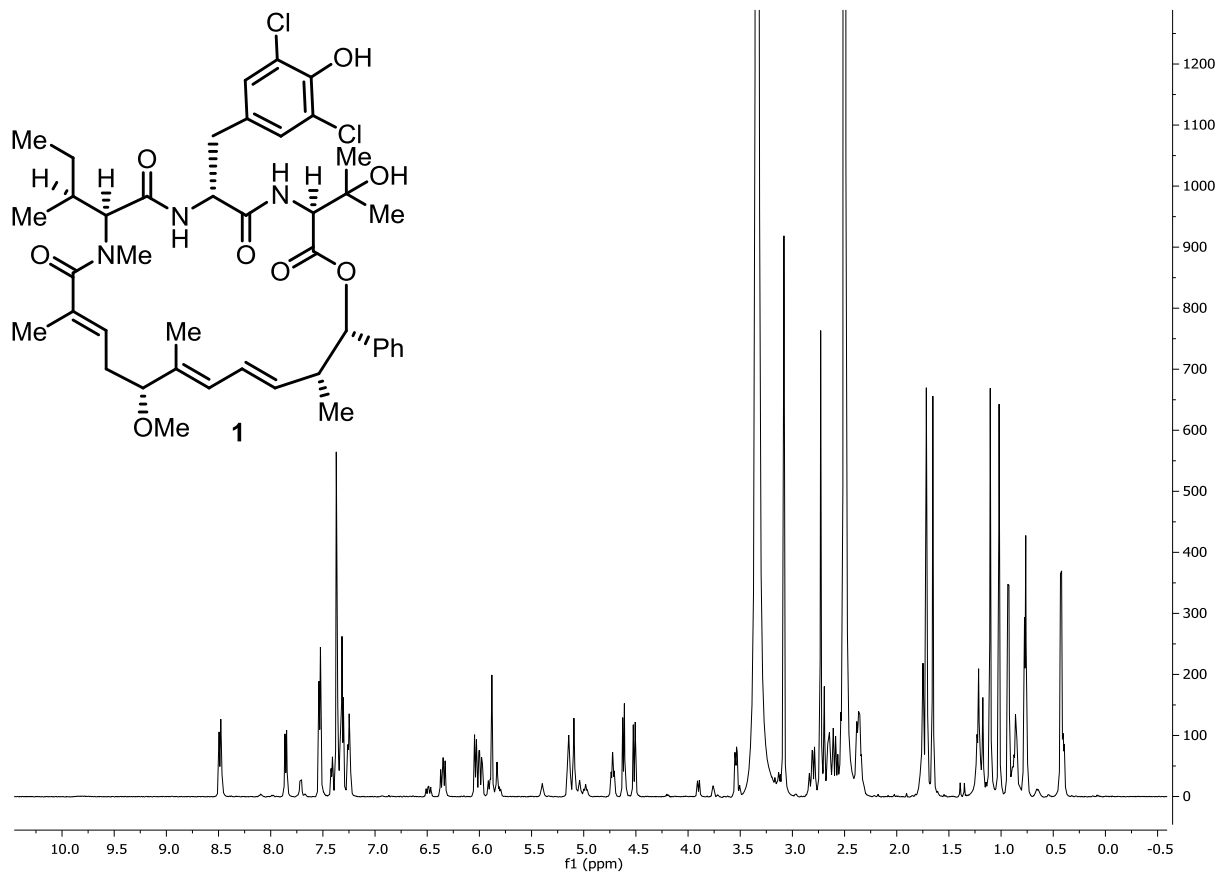


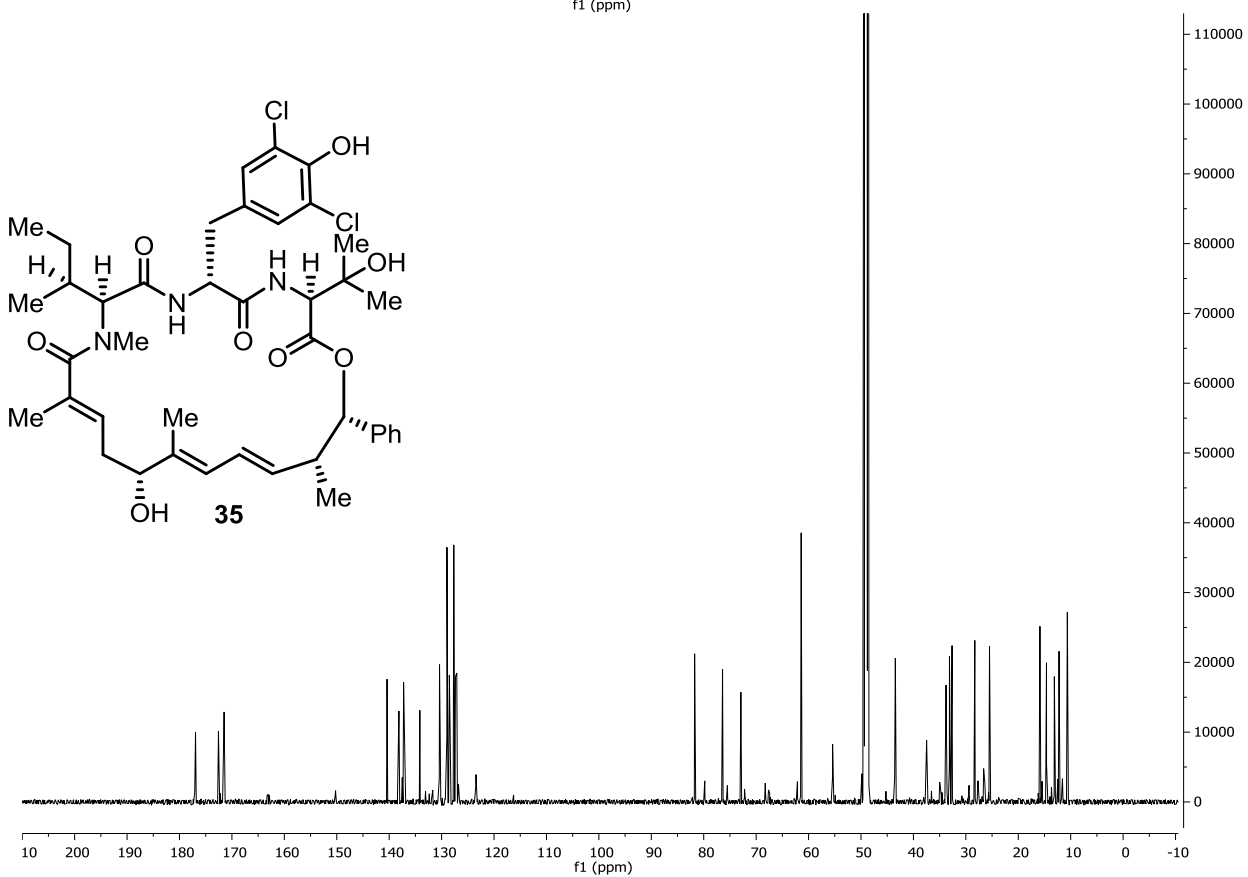
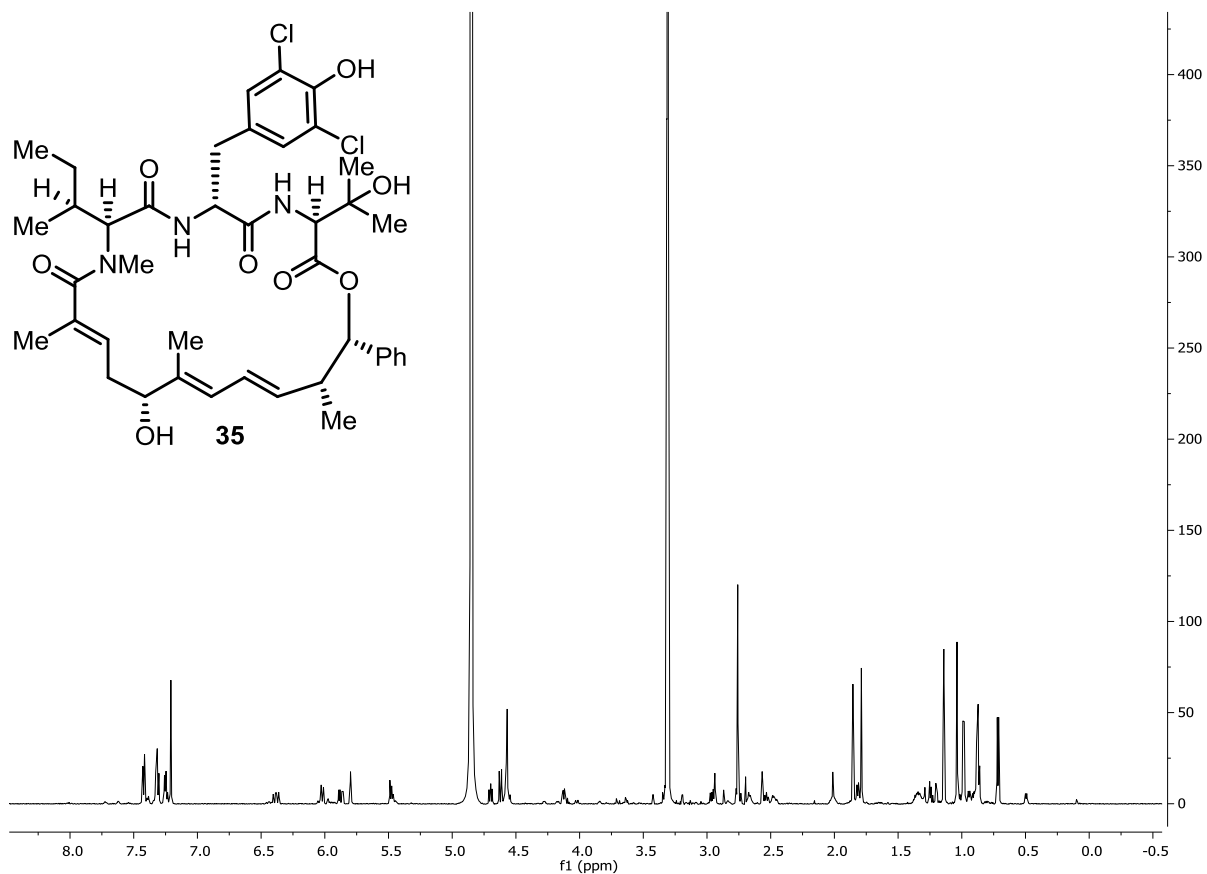
$^{119}\text{Sn}$  NMR (149 MHz,  $\text{CDCl}_3$ ):

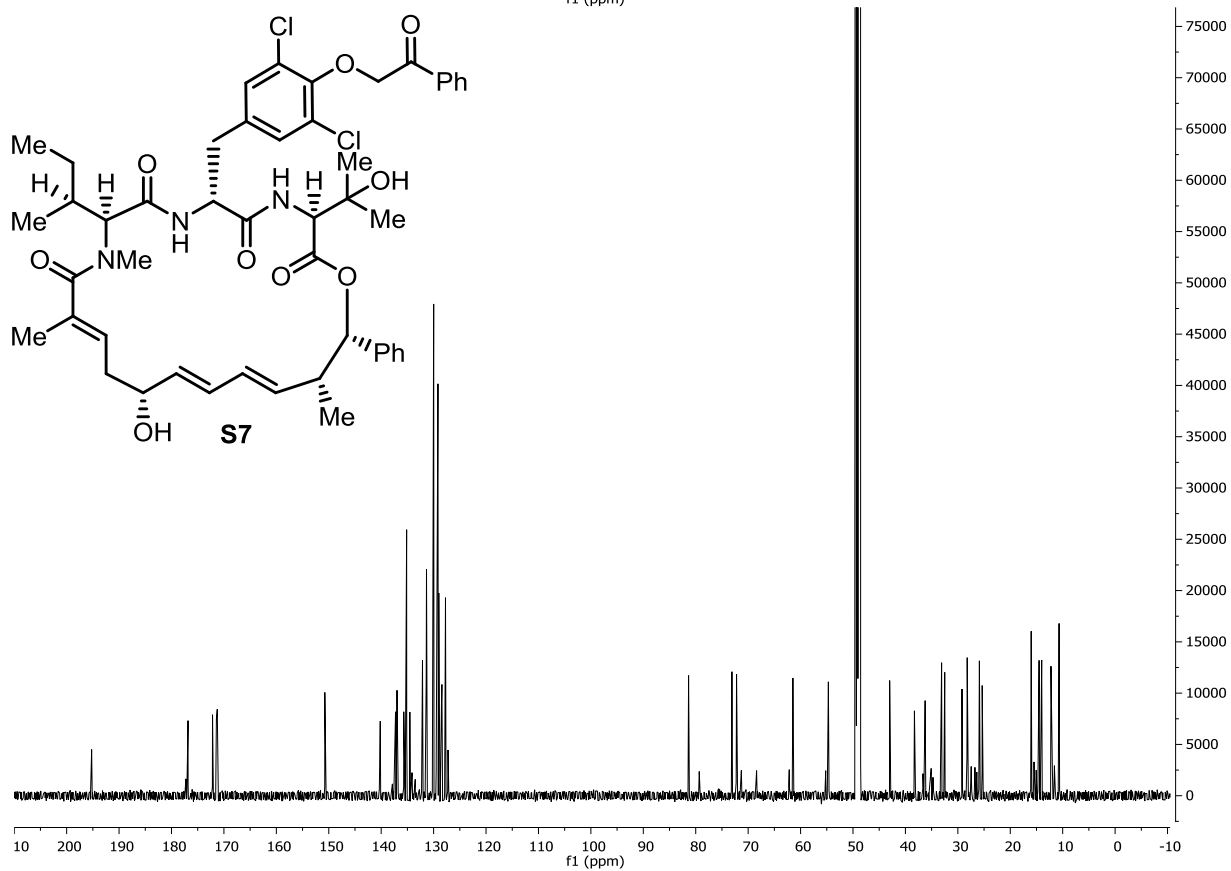
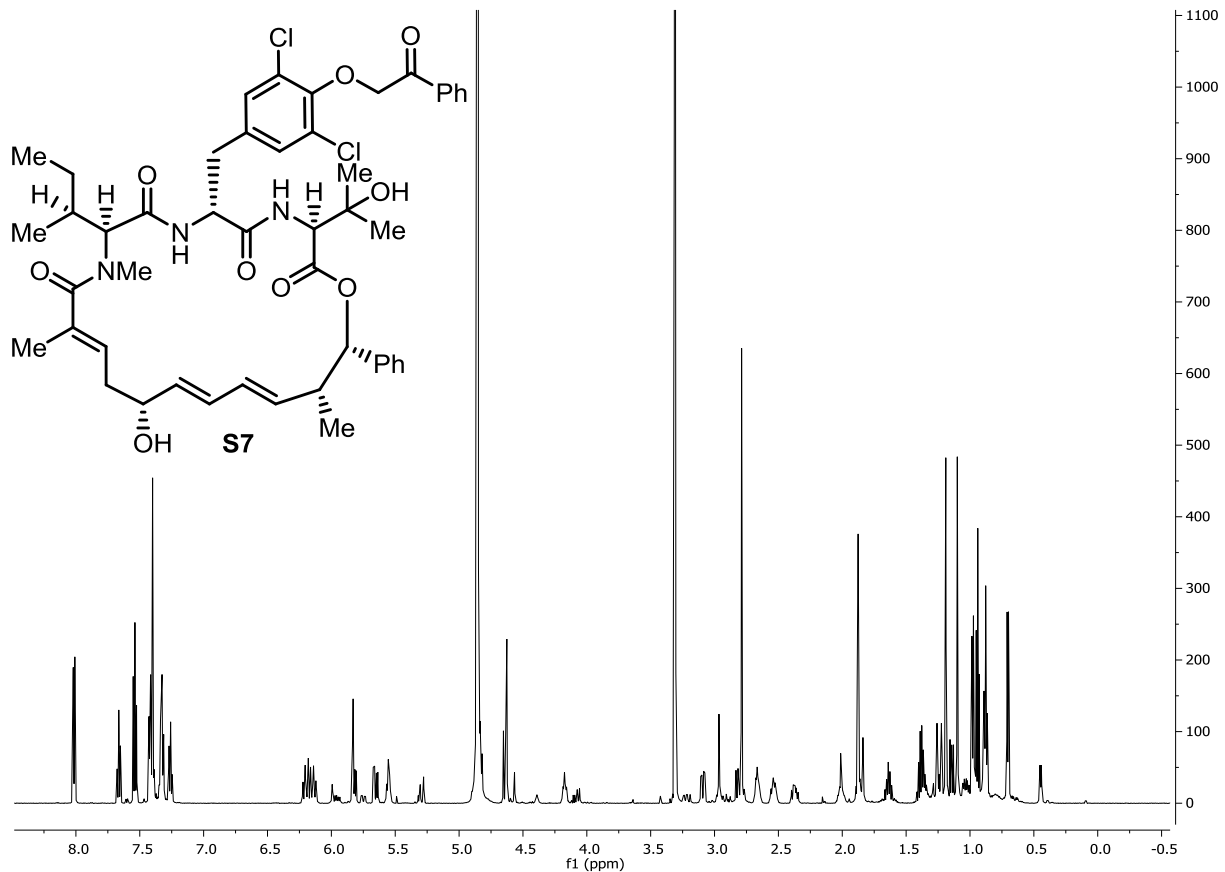




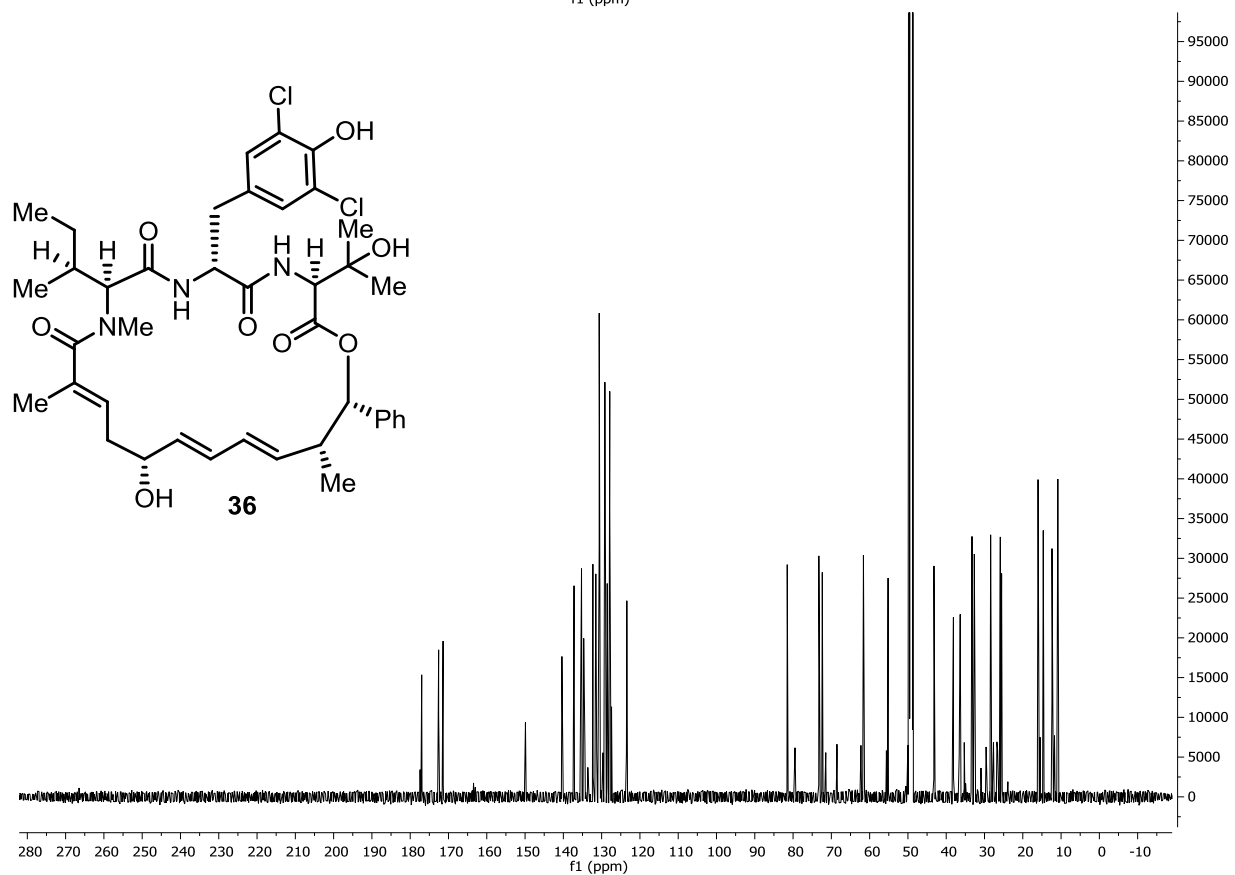
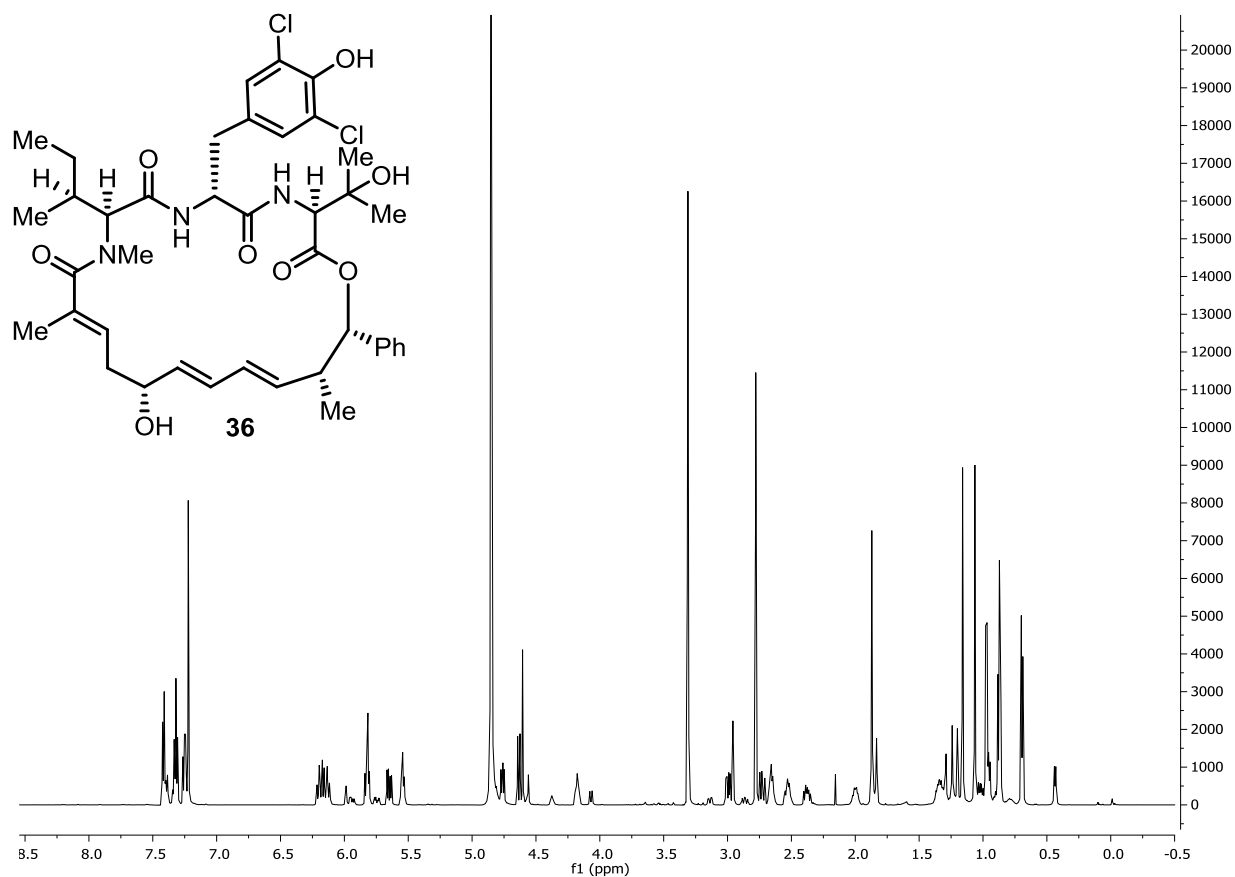


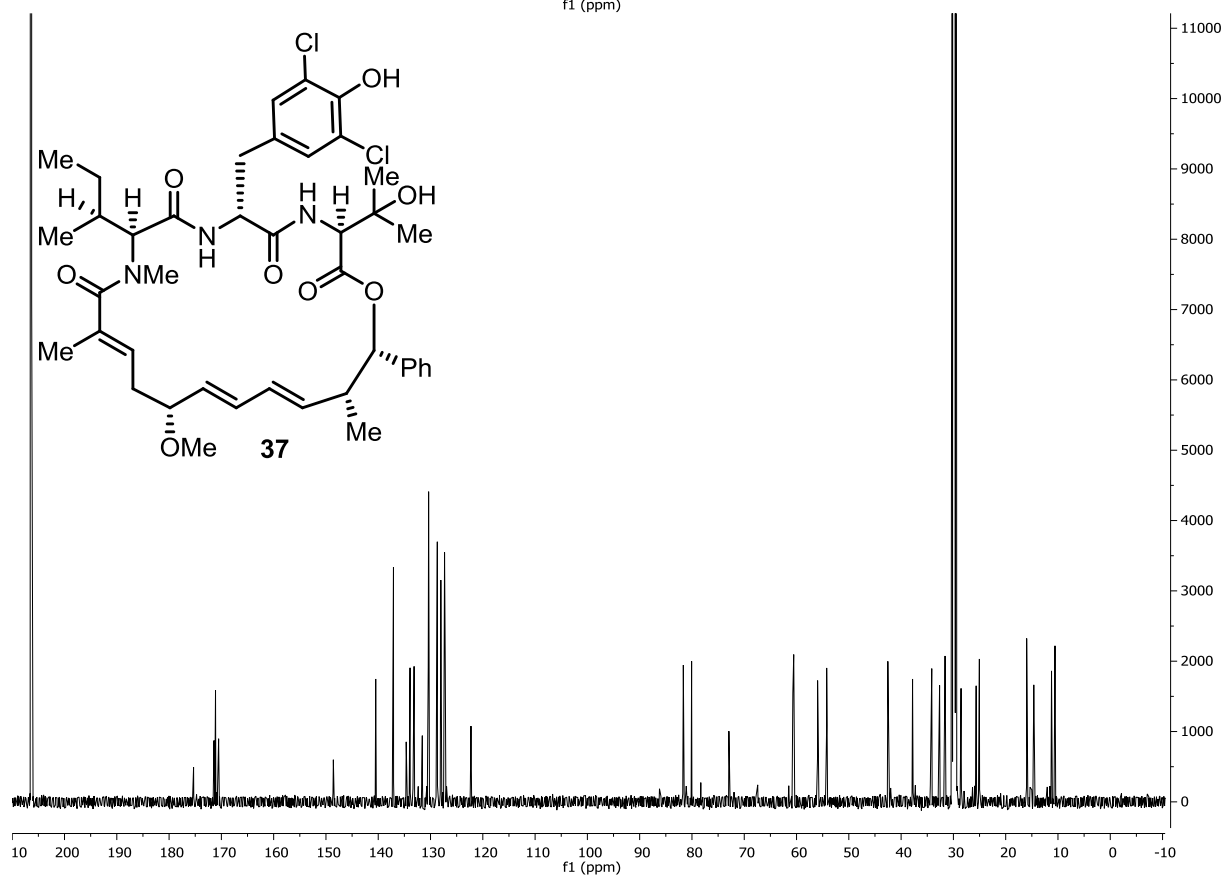
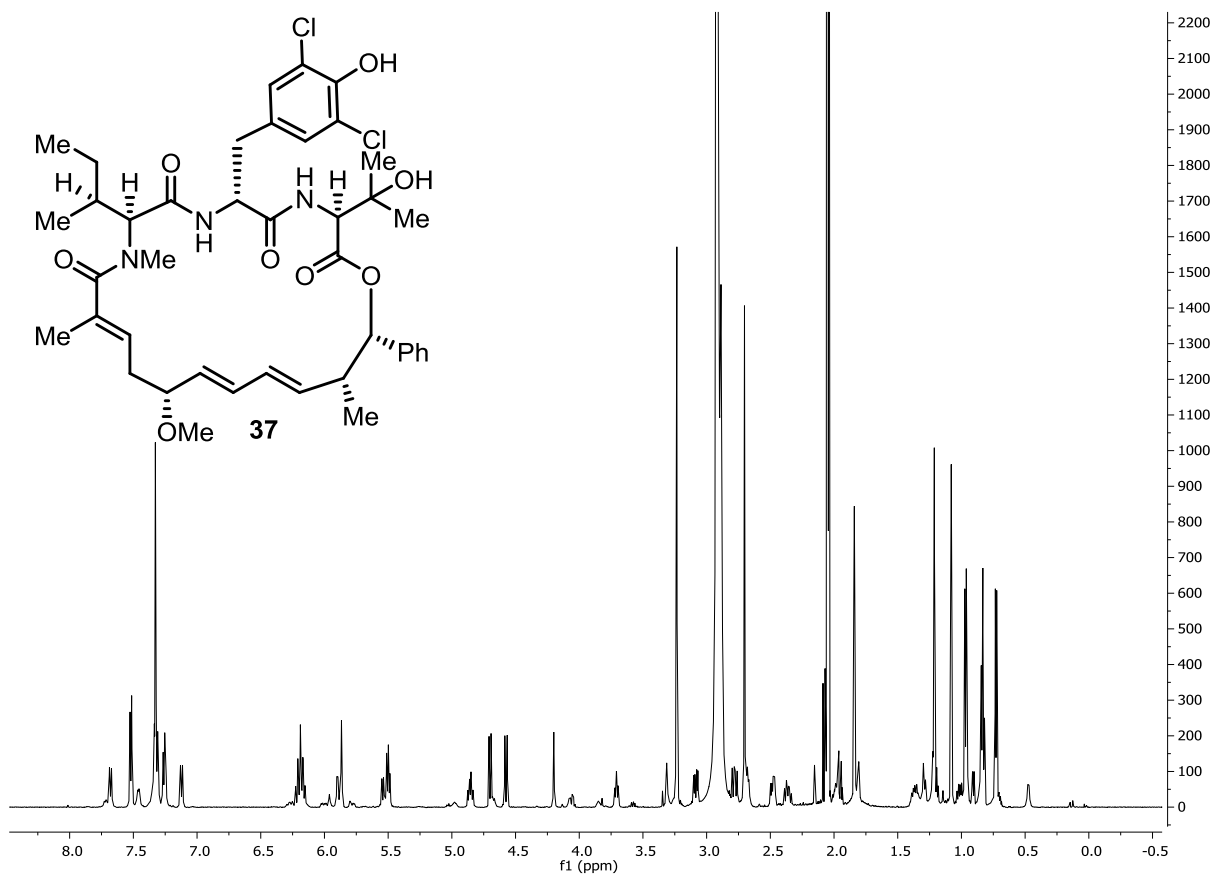


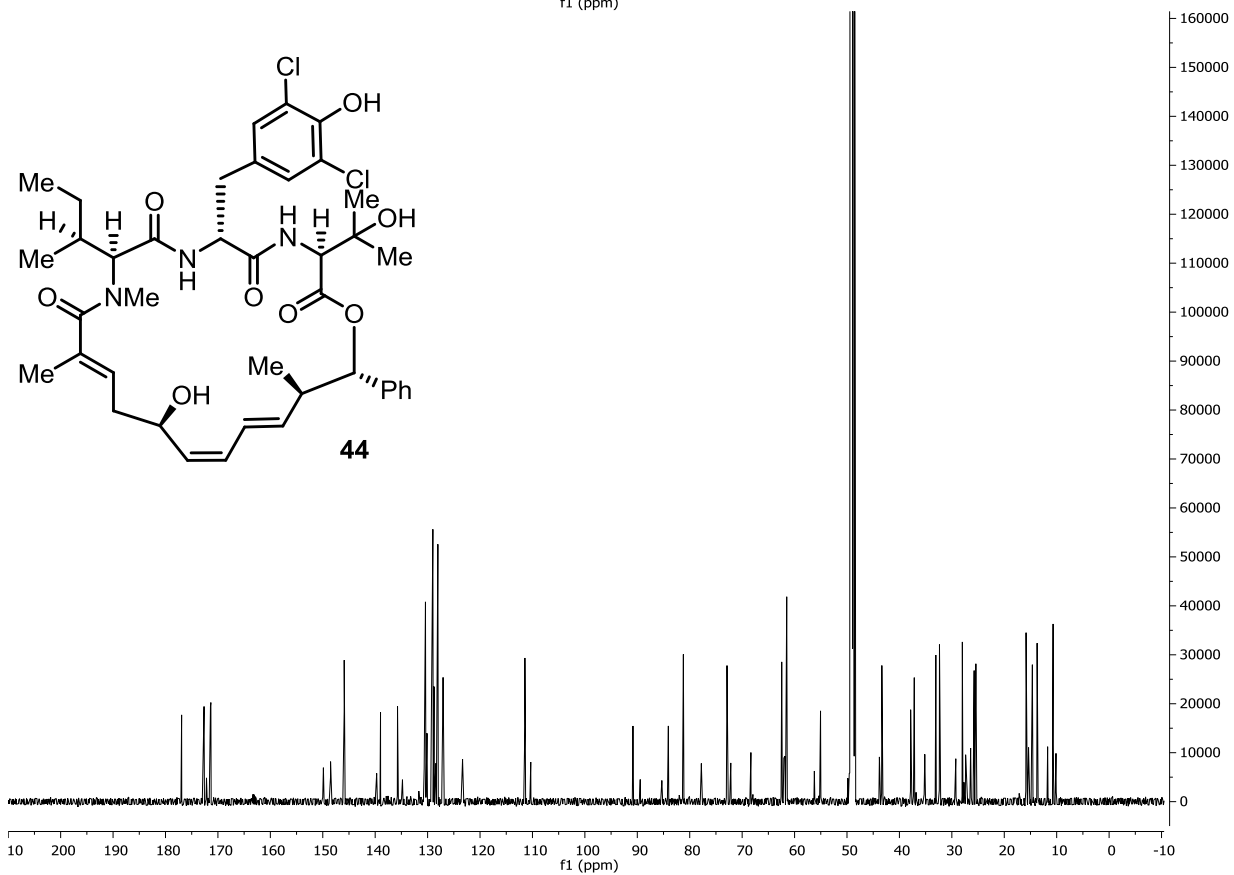
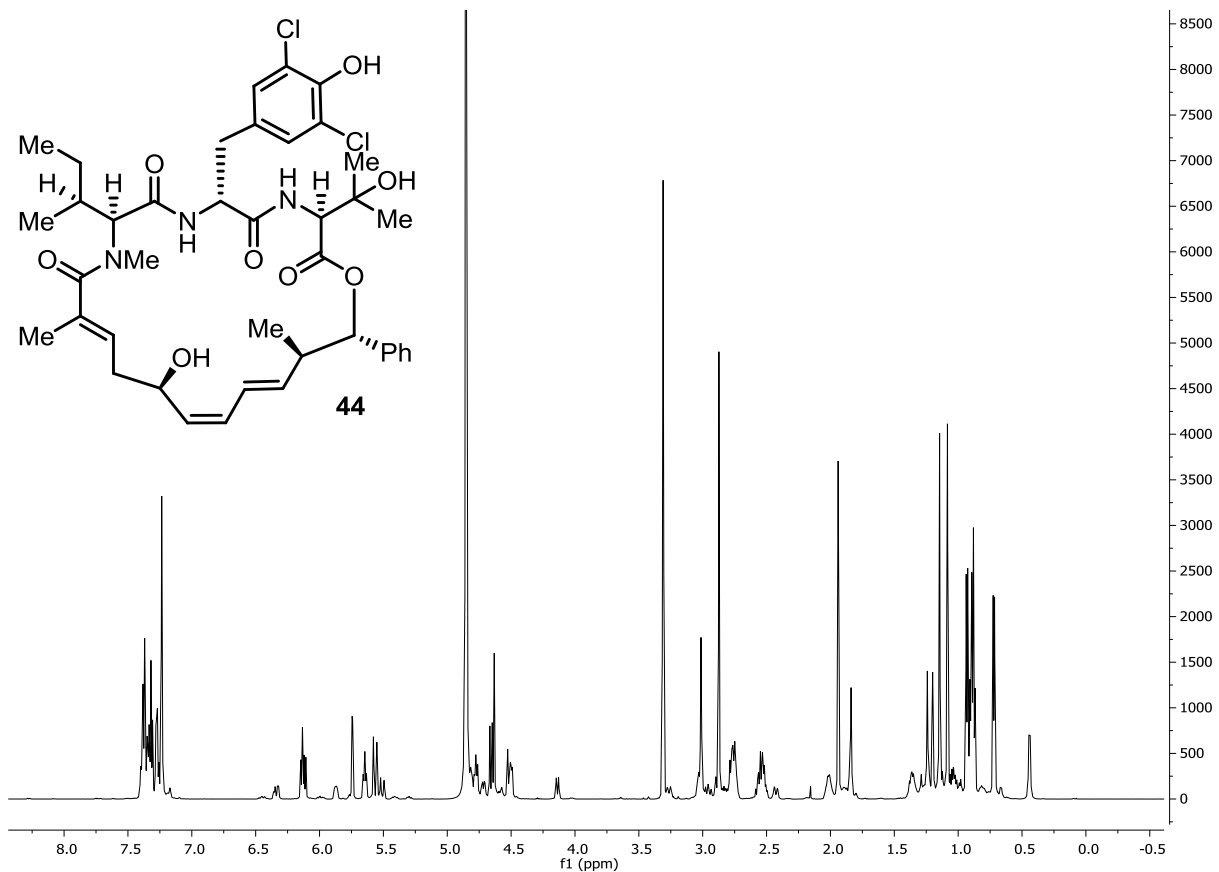


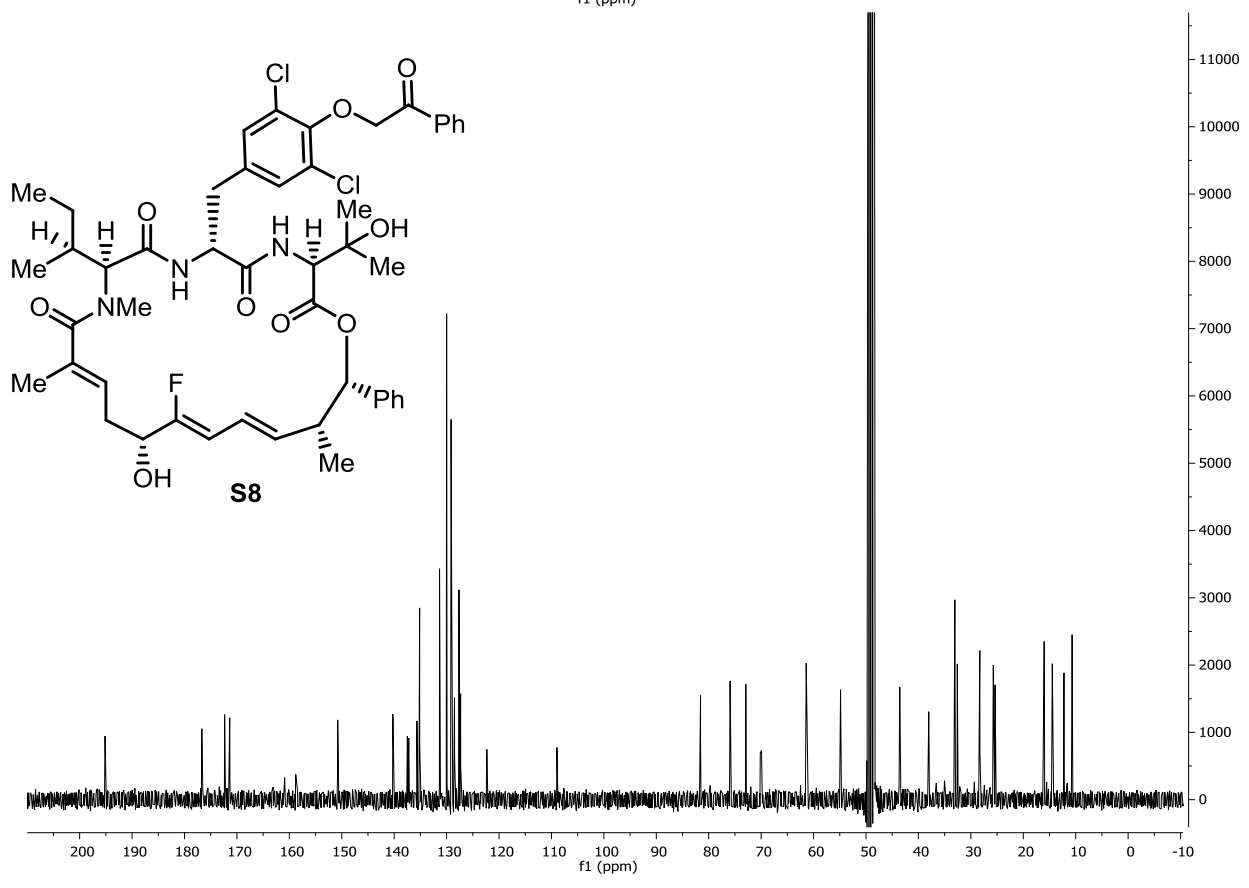
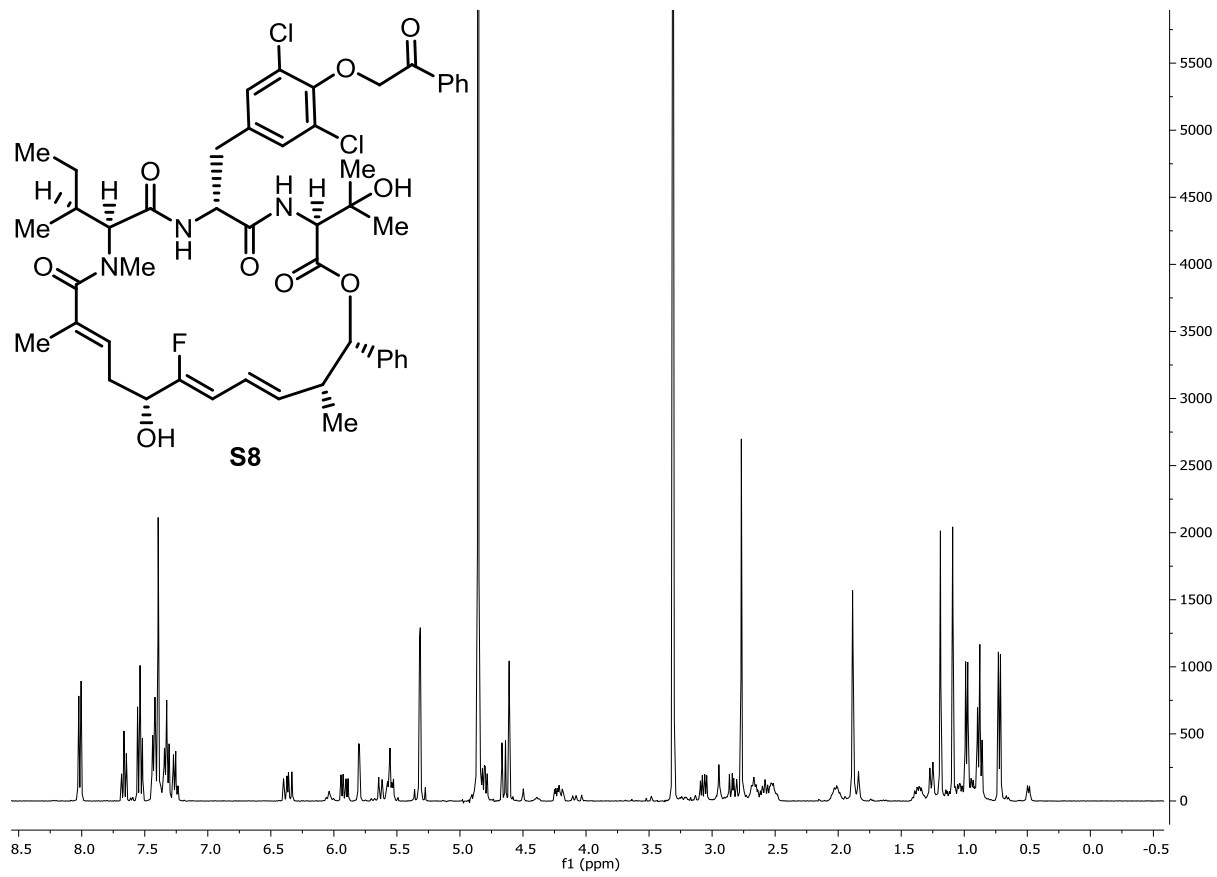




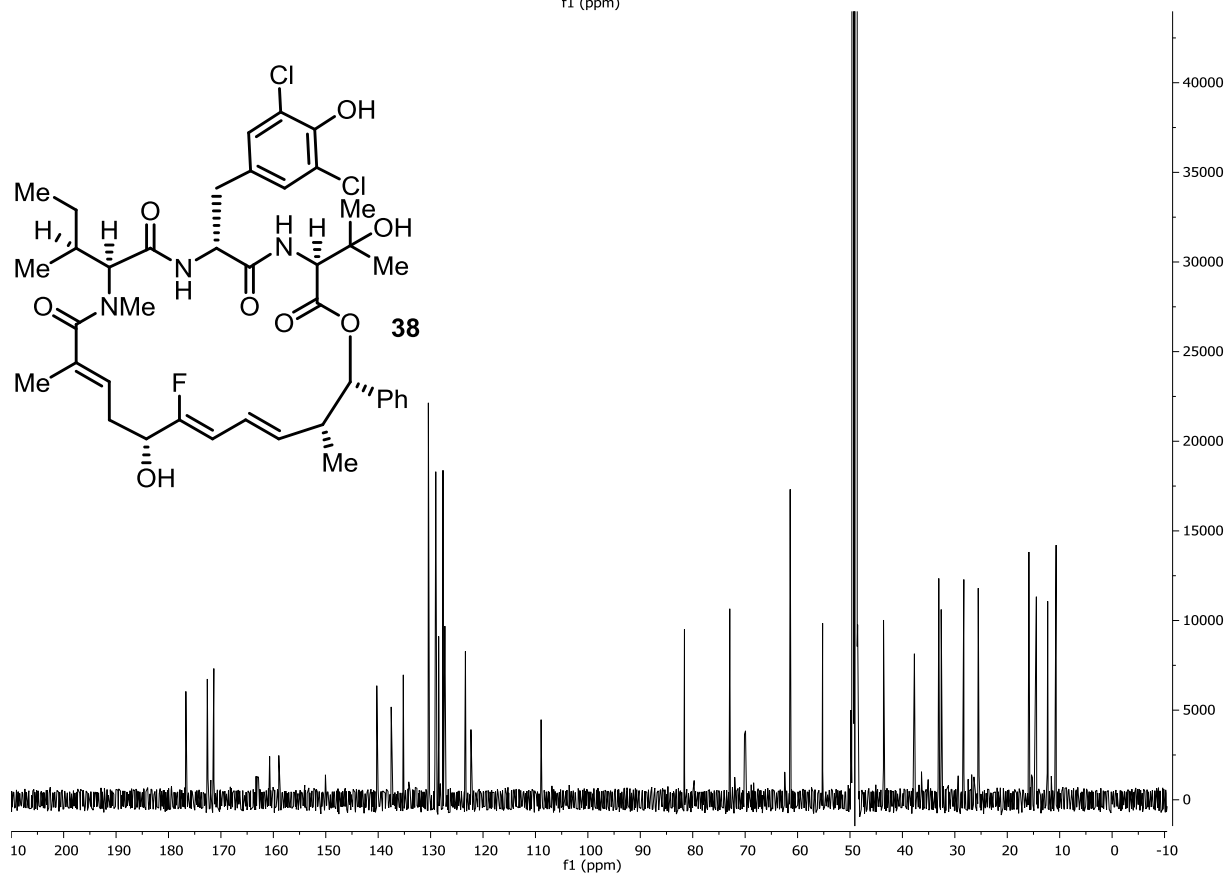
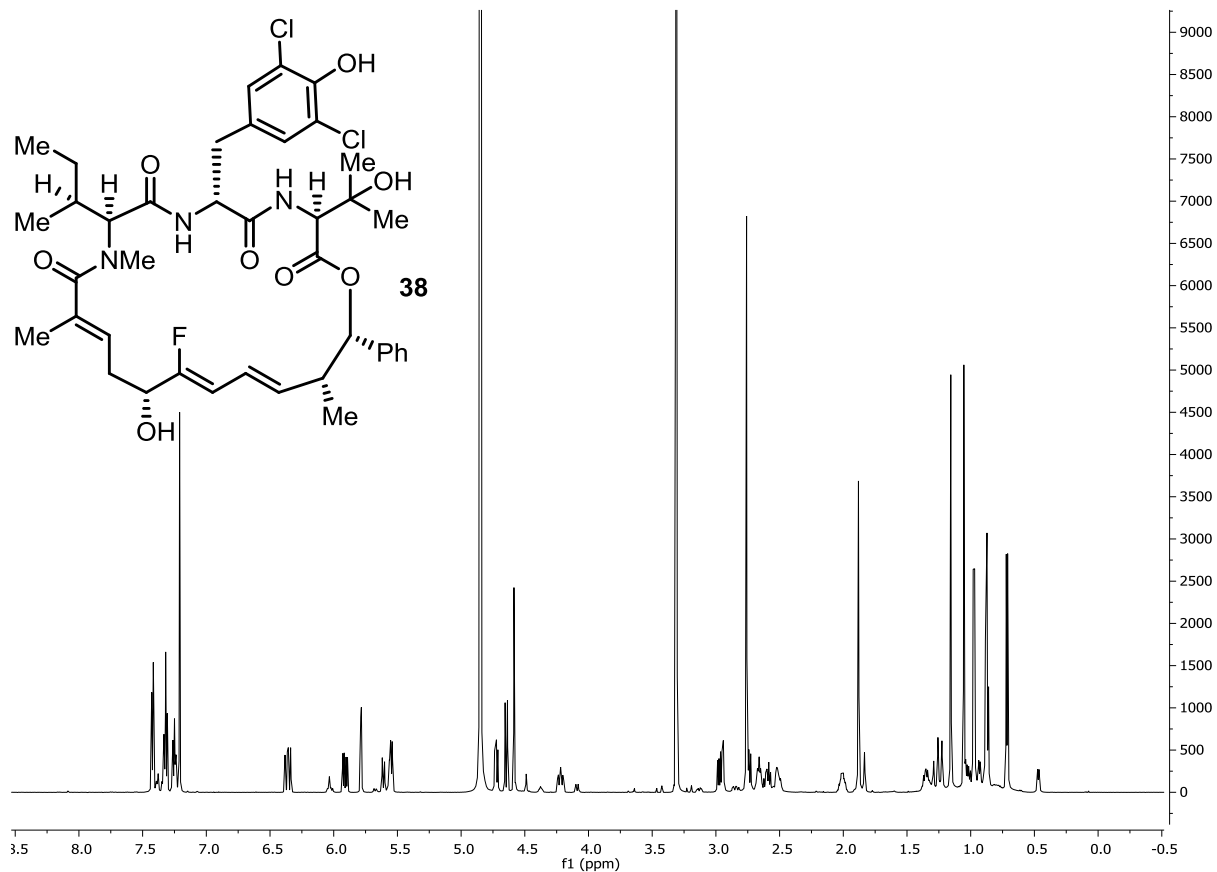


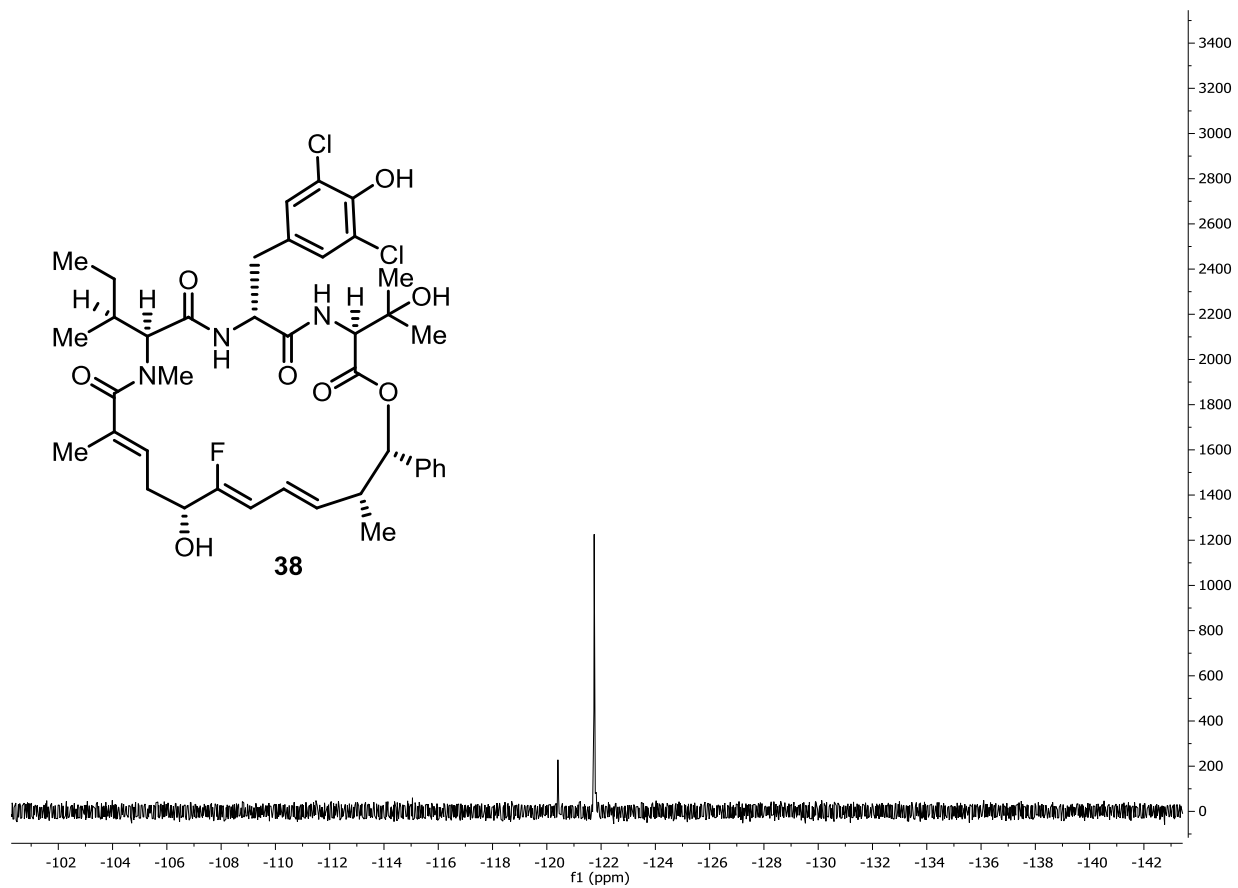


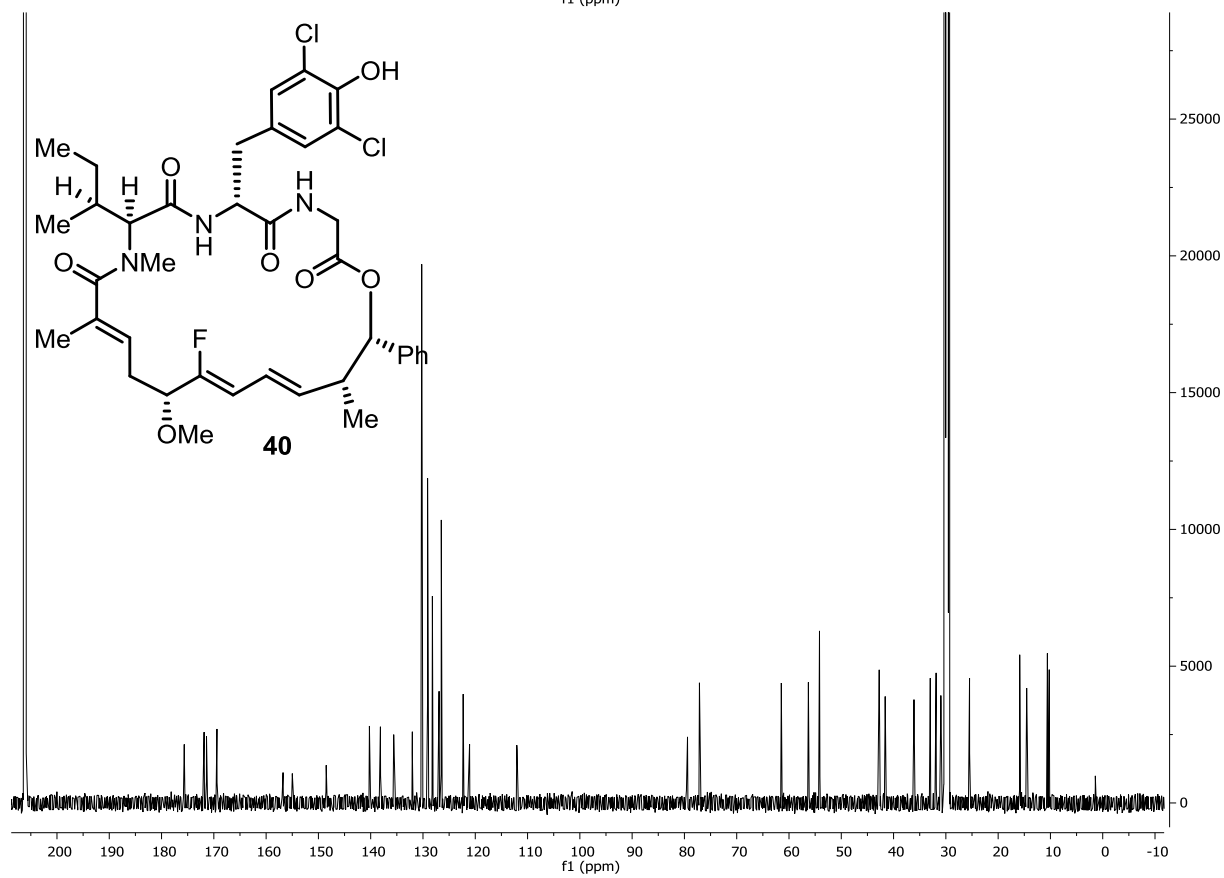
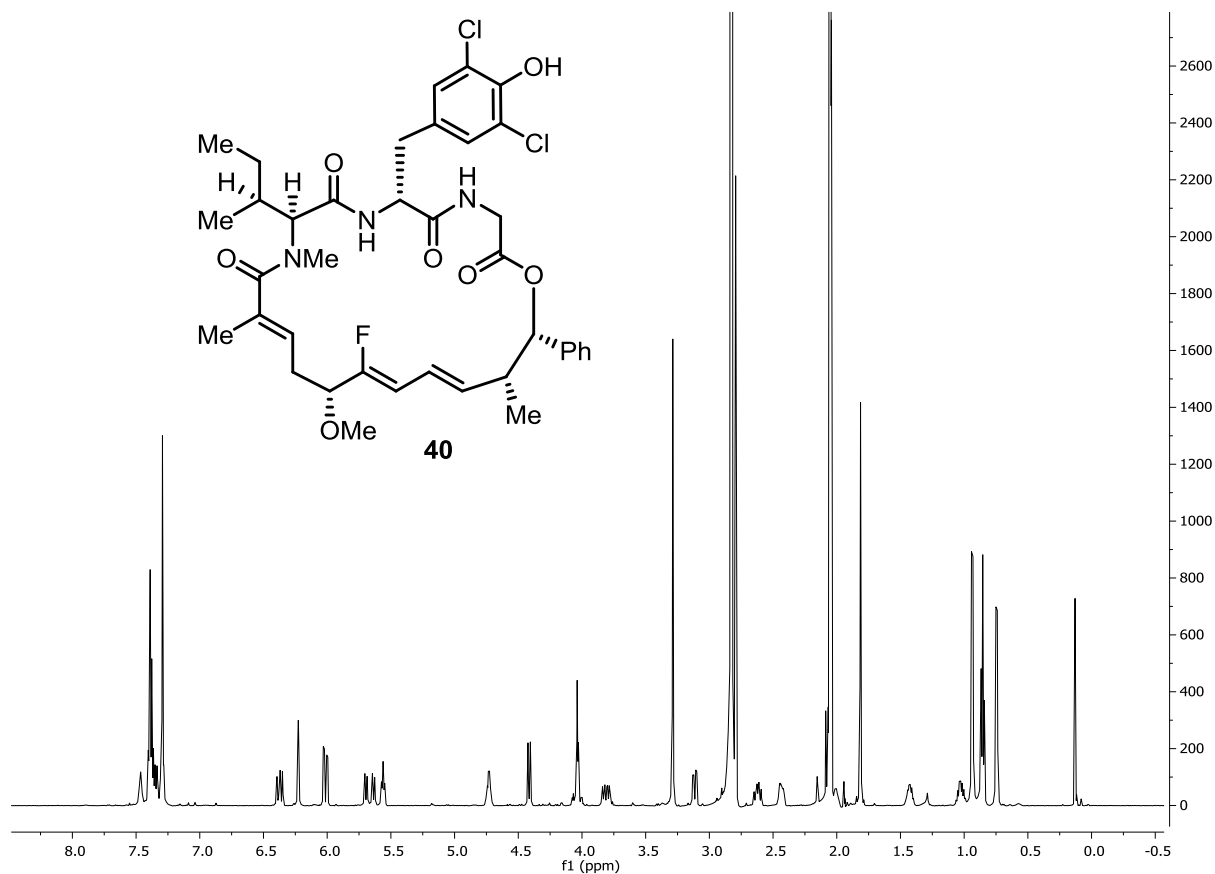




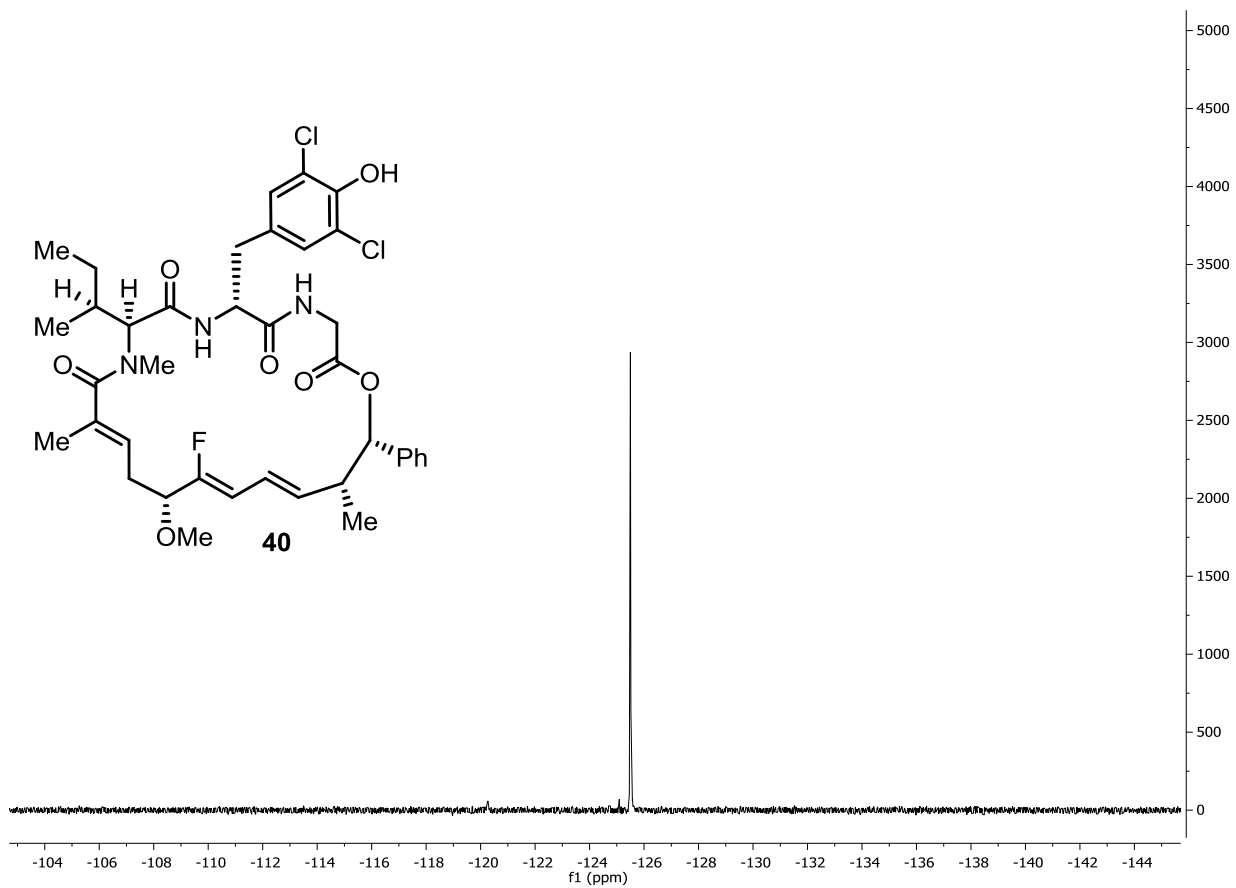


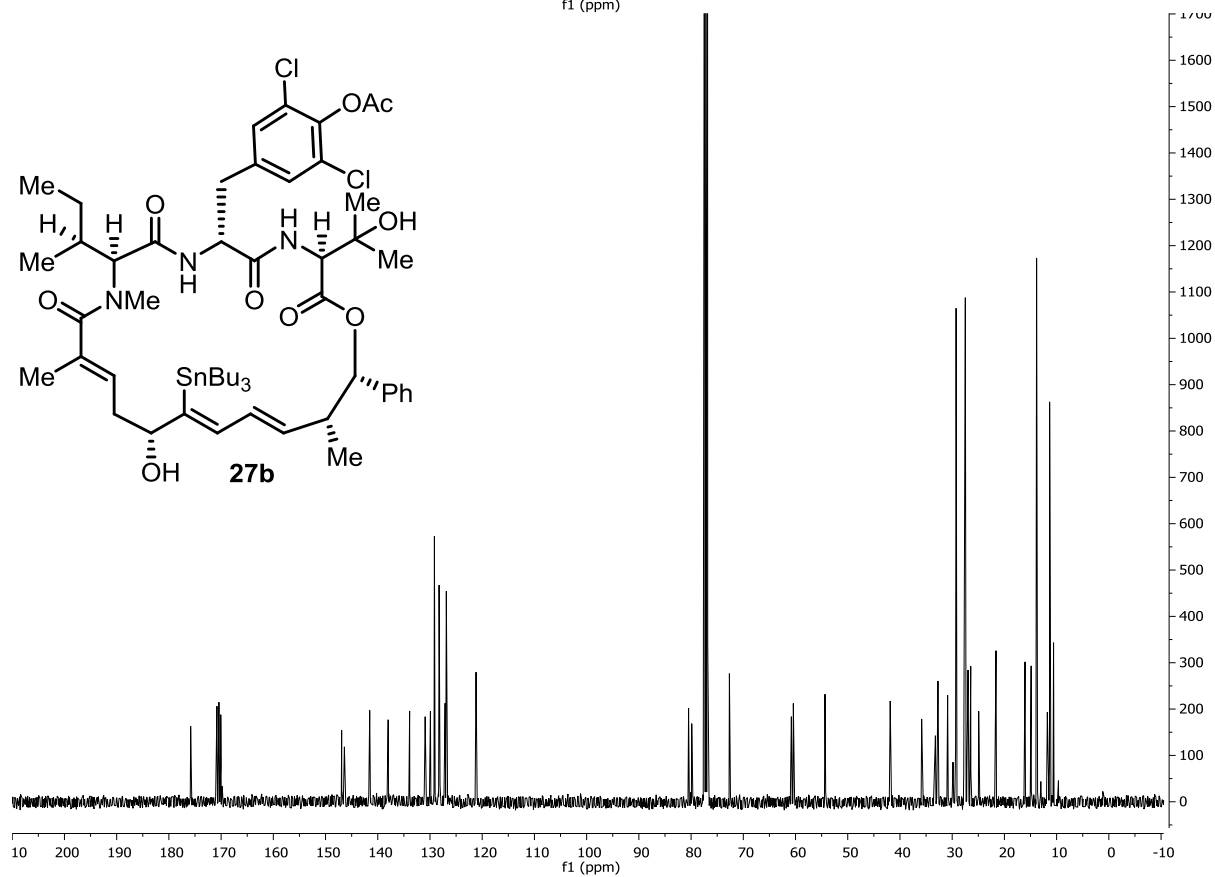
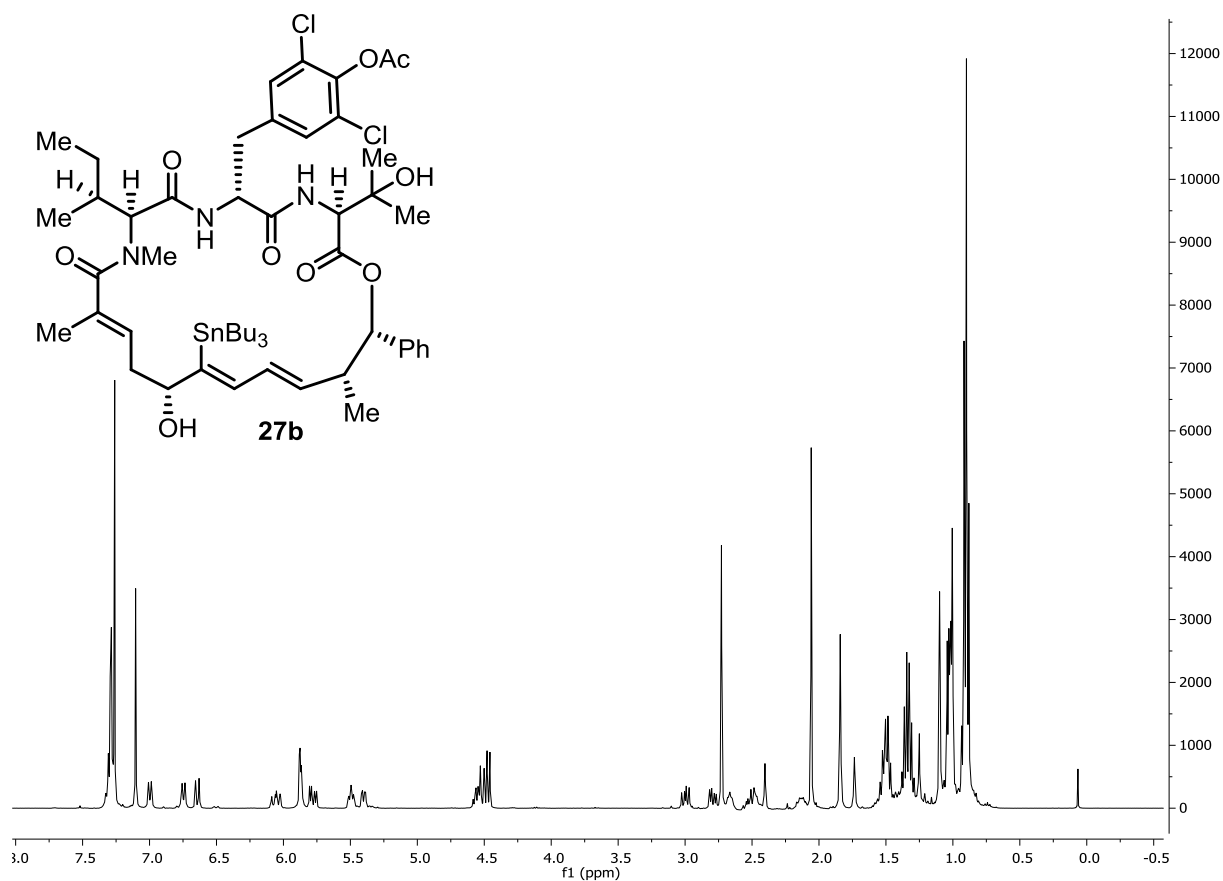




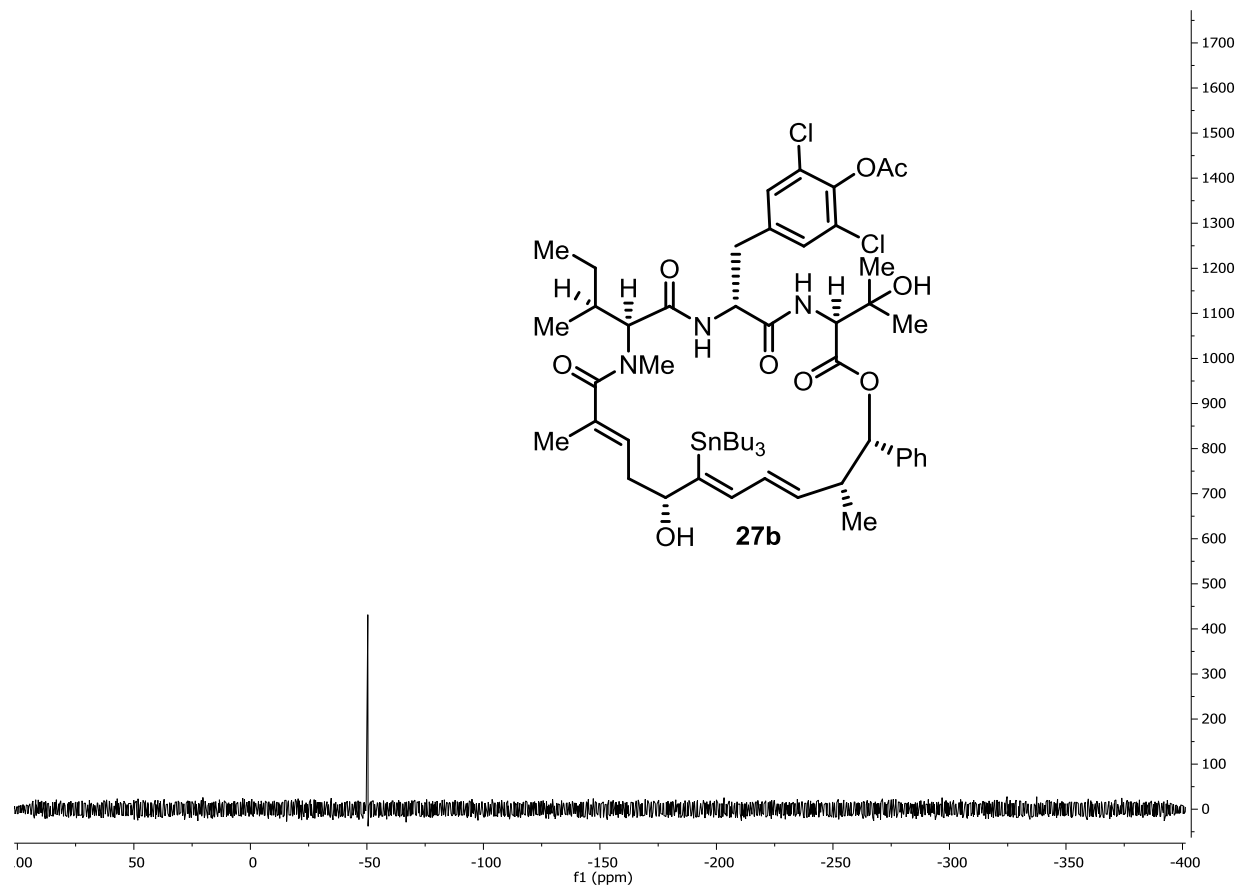


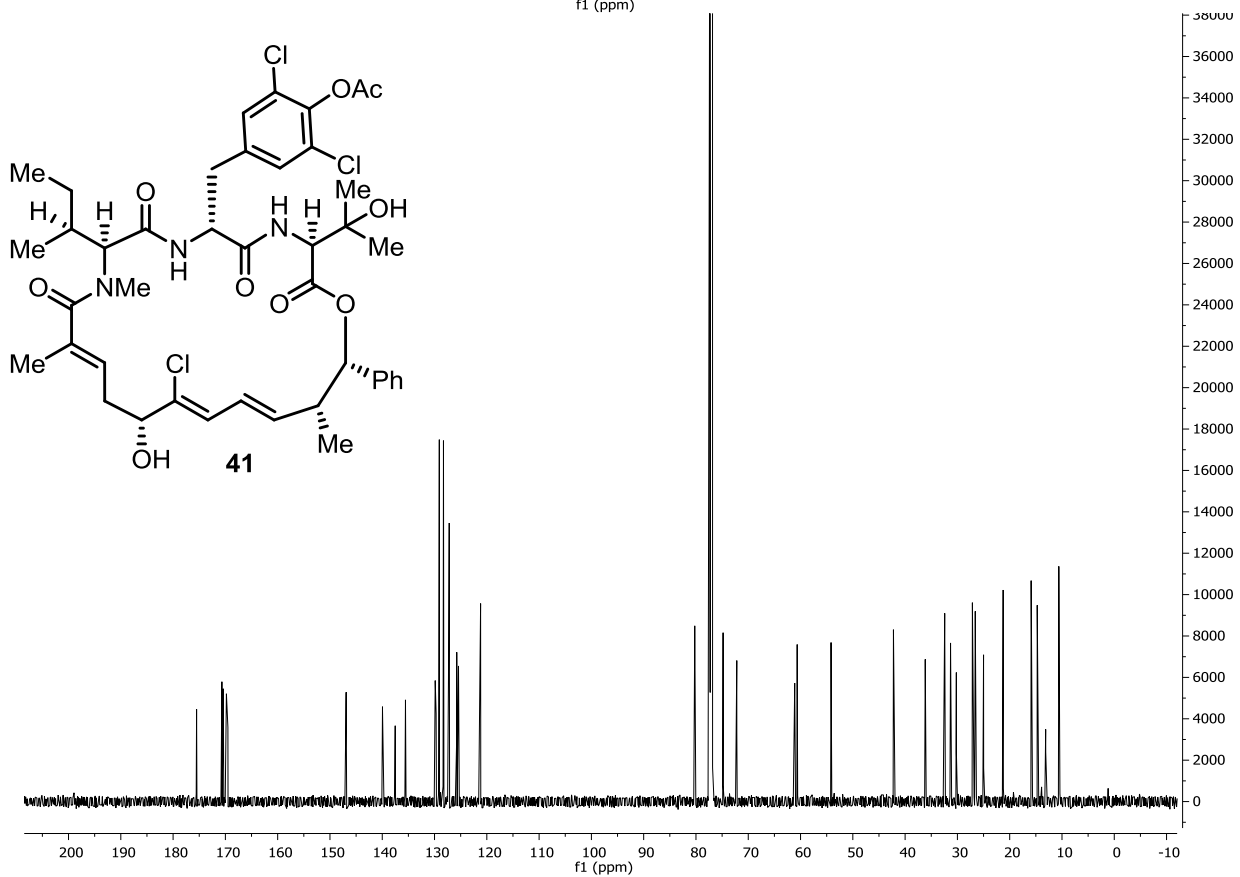
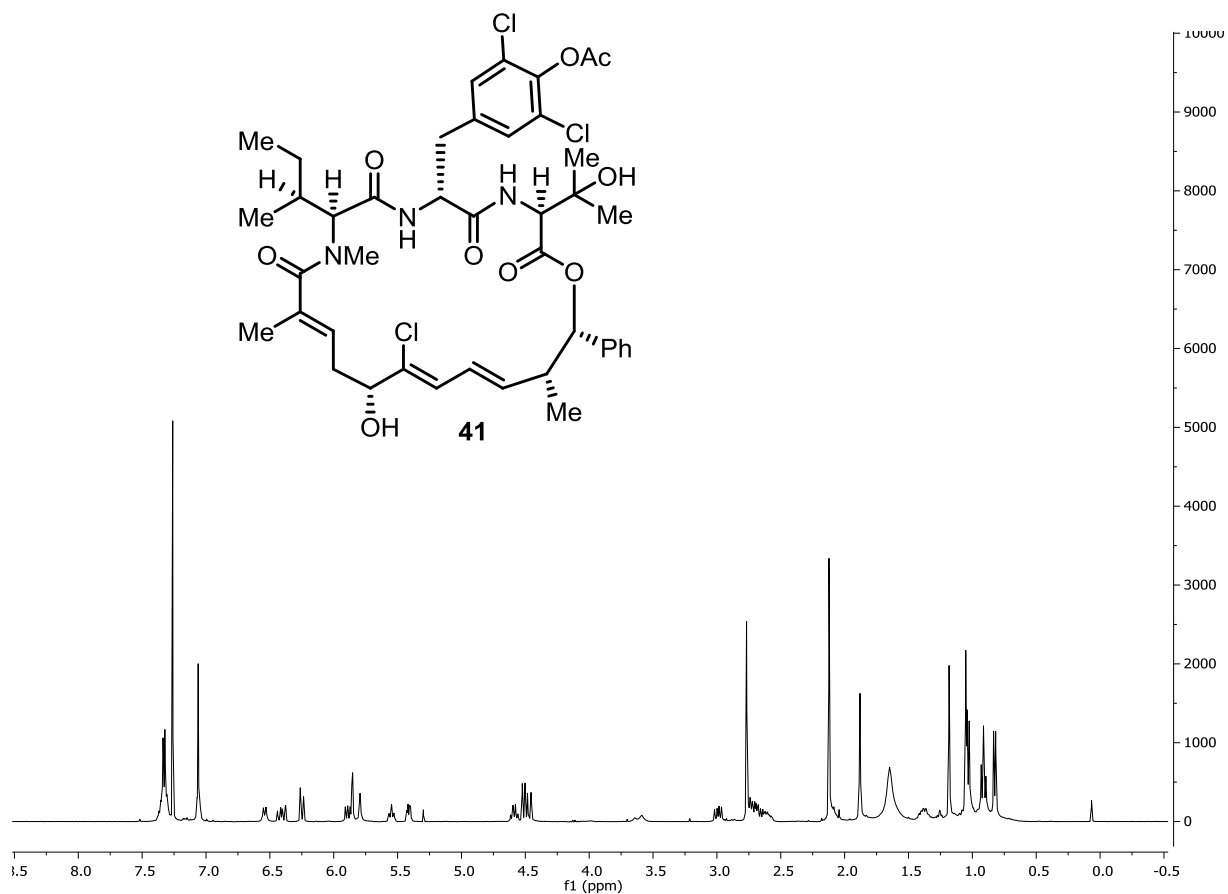


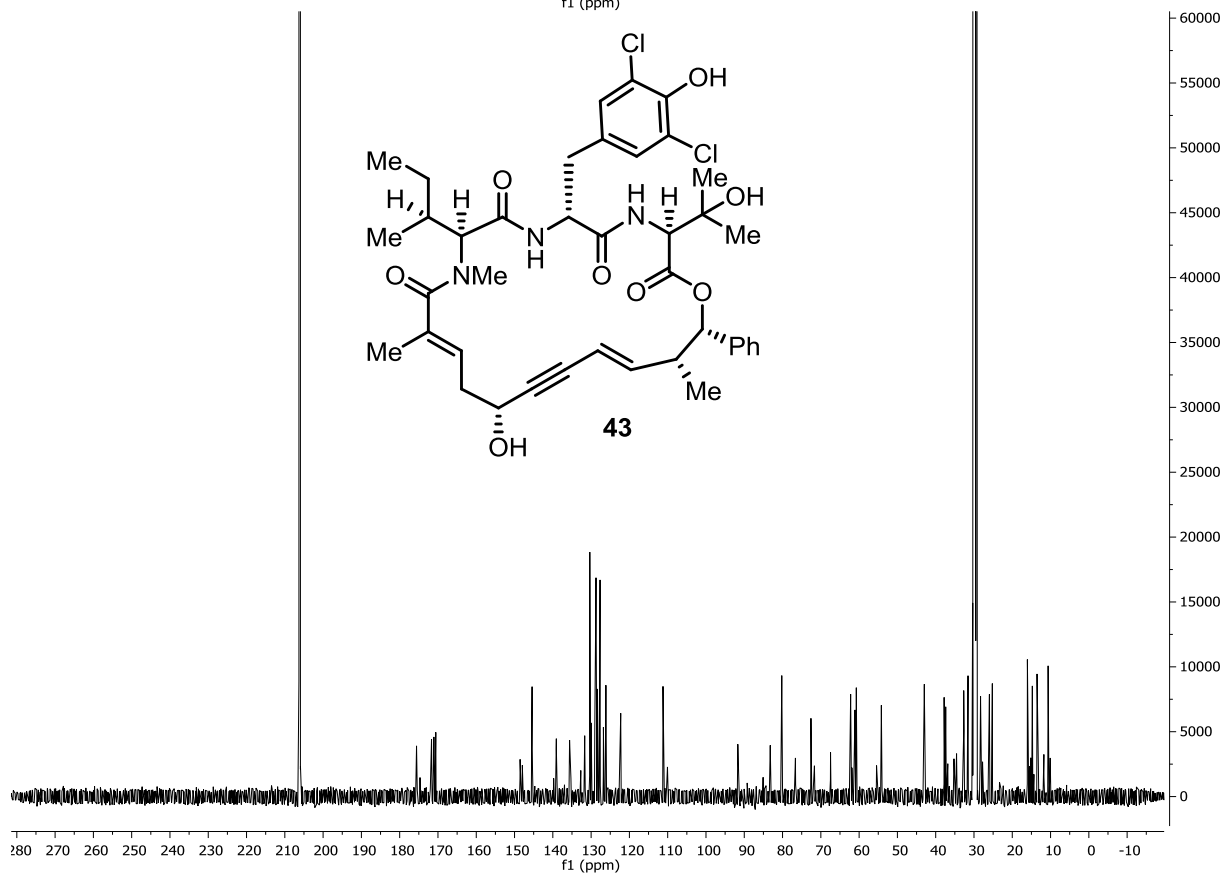
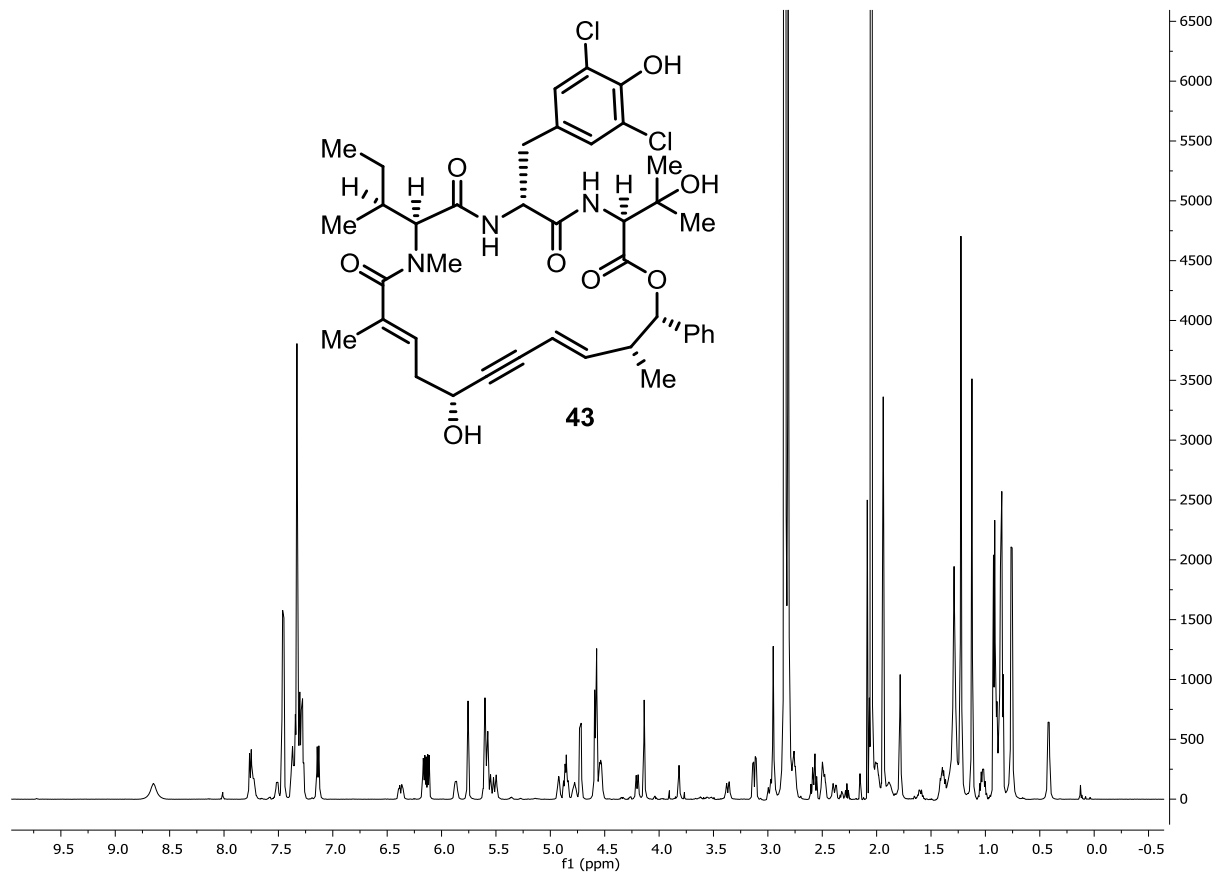


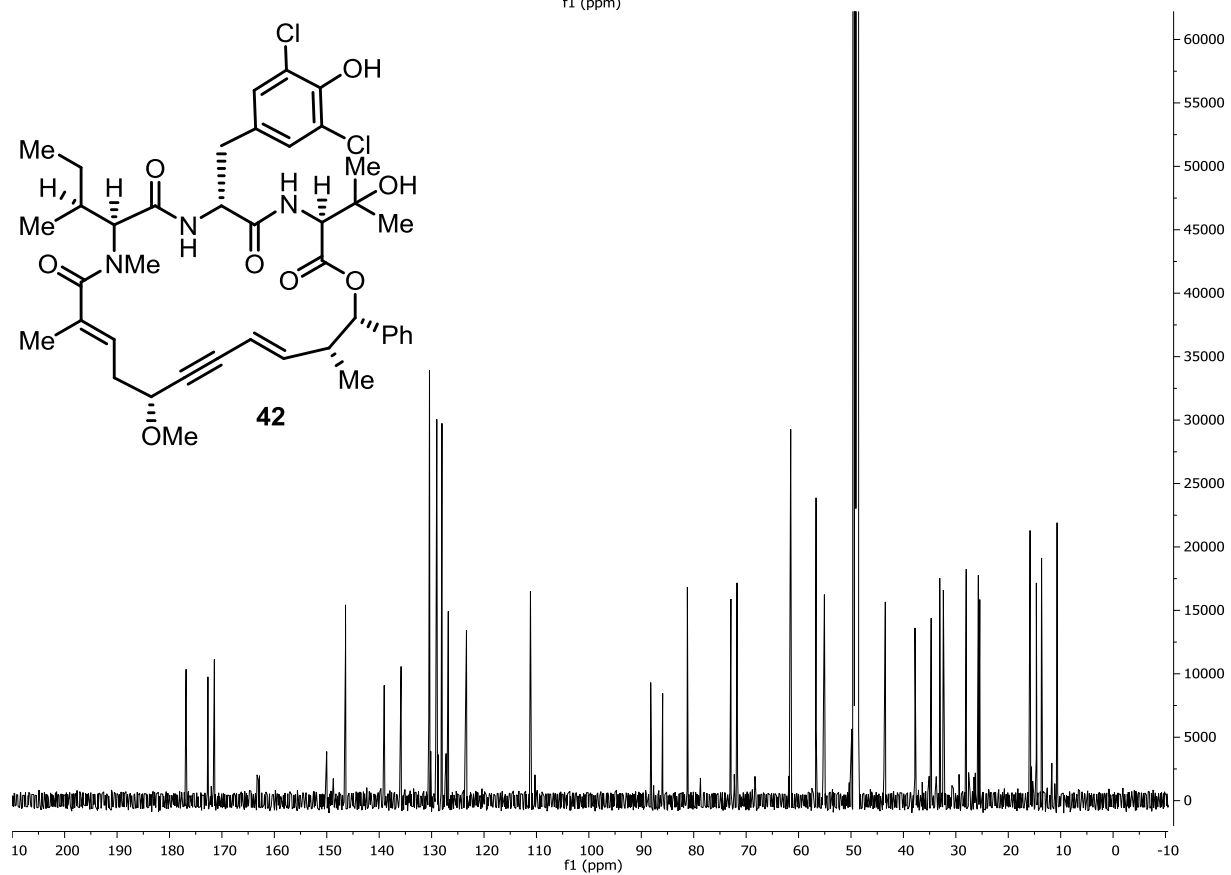
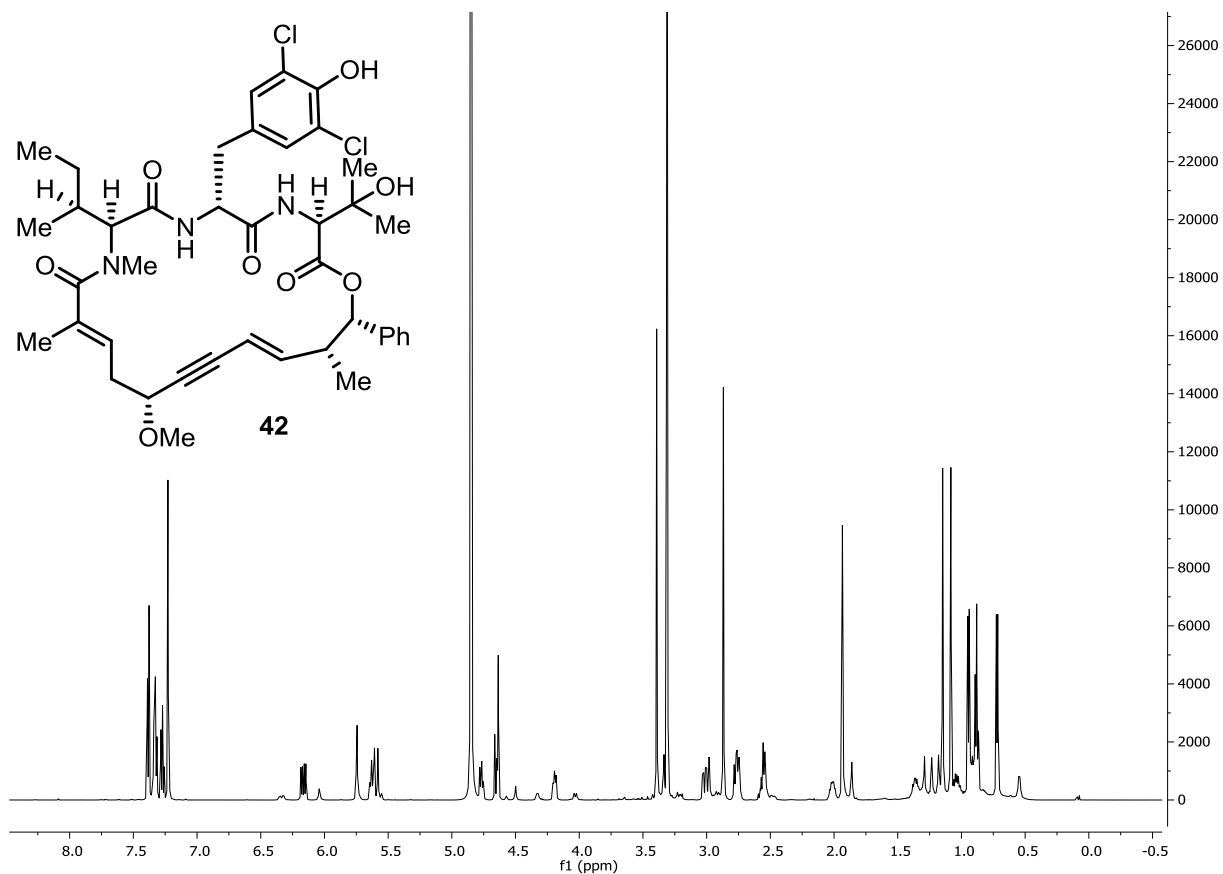


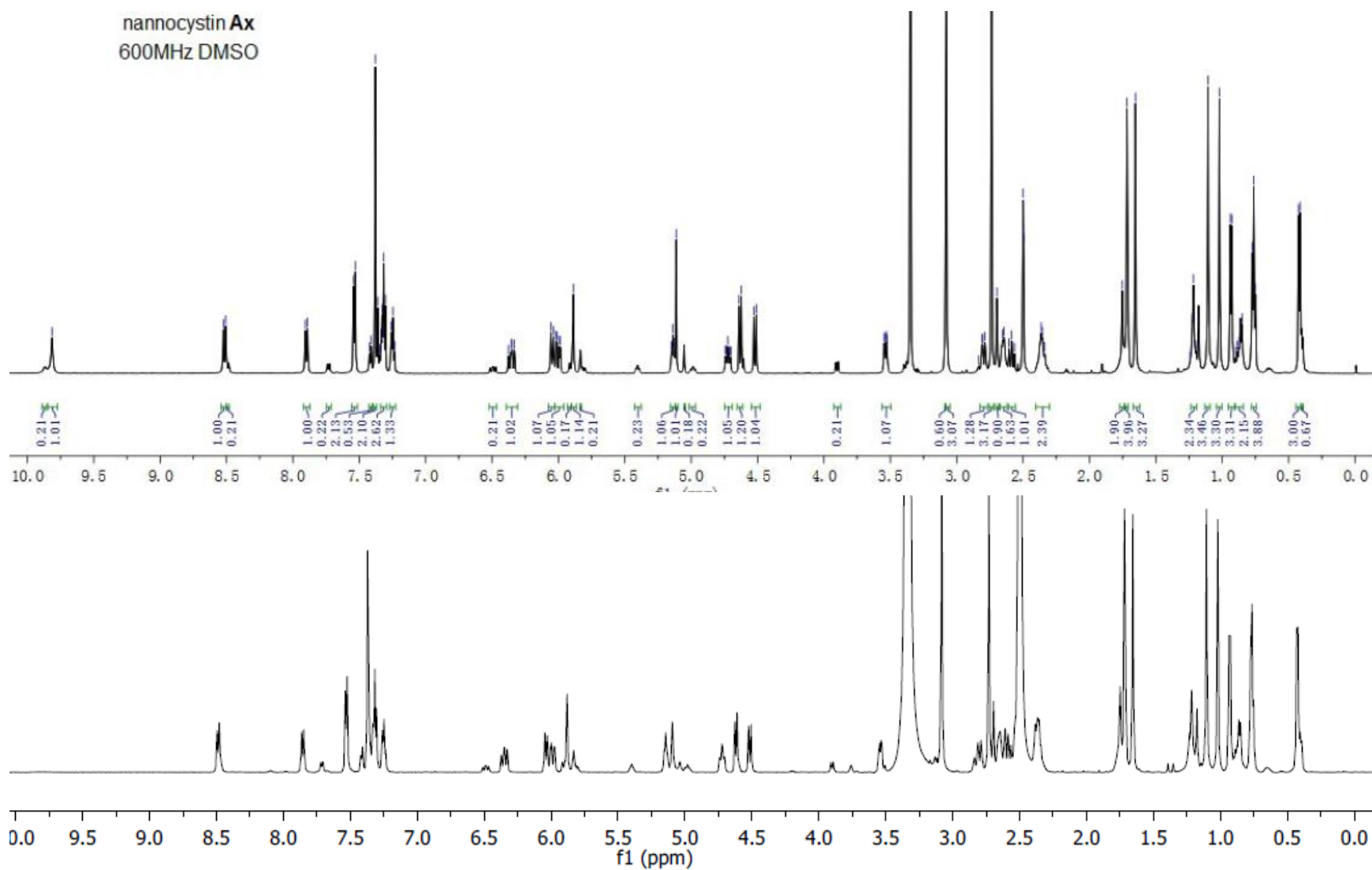
$^{119}\text{Sn}$  NMR (149 MHz,  $\text{CDCl}_3$ ):



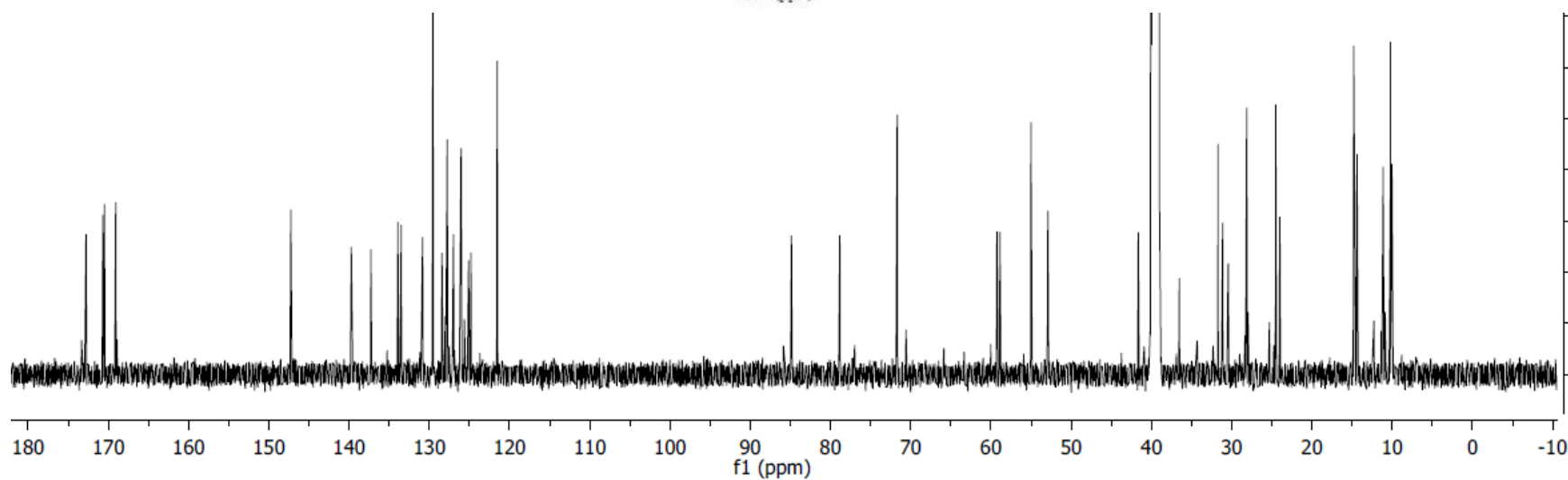
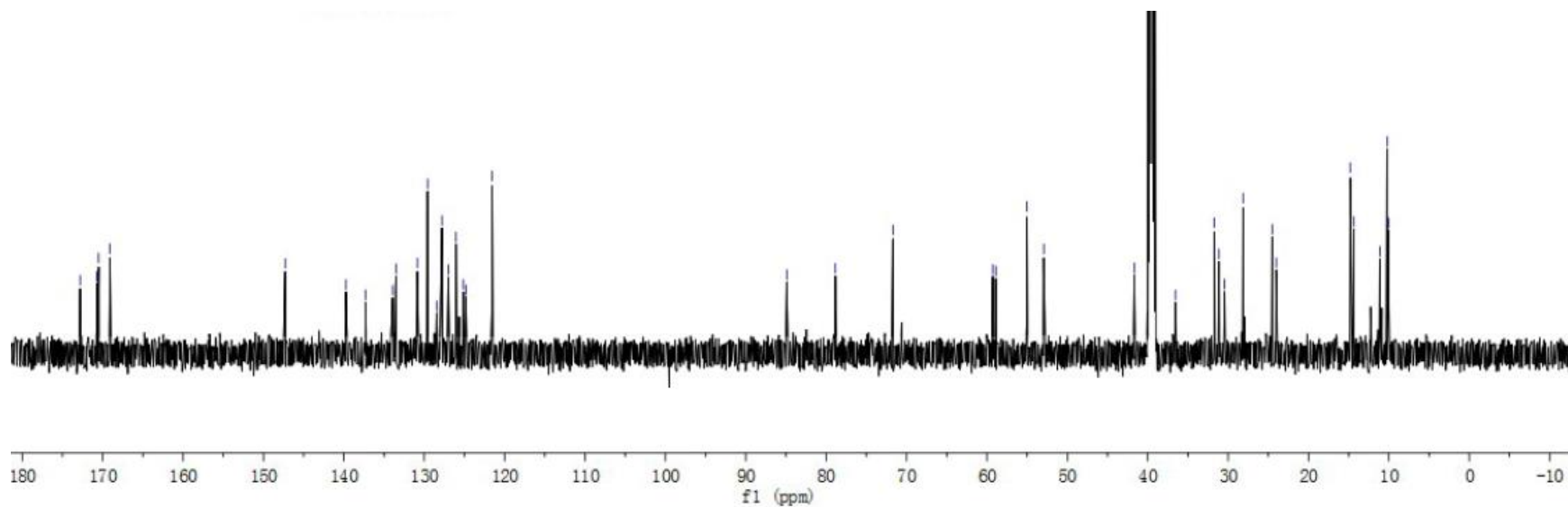








Comparison of the <sup>1</sup>H NMR spectra of Nannocystin Ax published by Liu (top) and the sample prepared in the Fürstner laboratory (bottom)



Comparison of the  $^{13}\text{C}$  NMR spectra of Nannocystin Ax published by Liu (top) and the sample prepared in the Fürstner laboratory (bottom)