

## Supporting information for journal *Marine drugs*

# Four New C9 Metabolites from the Sponge-associated Fungus *Gliomastix* sp. ZSDS1-F7-2

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## Table of contents

<b>Table S1.</b> <sup>1</sup> H and <sup>13</sup> C NMR data of compounds <b>2</b> and prelactone C	Page 3	<b>Figure S20.</b> <sup>1</sup> H NMR (500 MHz, CD <sub>3</sub> OD) of compound <b>4</b> .	Page 23
<b>Table S2.</b> Anti-fouling and cytotoxicity activities of compounds <b>1–4</b>	3	<b>Figure S21.</b> <sup>13</sup> C NMR (75 MHz, CD <sub>3</sub> OD) of compound <b>4</b>	24
<b>Figure S1.</b> The conformers, relative energies (ΔG) and conformer populations of the minimized conformers of (4 <i>R</i> , 5 <i>S</i> )- <b>1</b> within an energy range of 3 kcal/mol.	4	<b>Figure S22.</b> <sup>1</sup> H- <sup>1</sup> H COSY (500 MHz, CD <sub>3</sub> OD) of compound <b>4</b>	25
<b>Figure S2.</b> The conformers, relative energies (ΔG) and conformer populations of the minimized conformers of (4 <i>R</i> , 5 <i>R</i> )- <b>1</b> within an energy range of 3 kcal/mol.	5	<b>Figure S23.</b> HSQC (500 MHz, CD <sub>3</sub> OD) of compound <b>4</b> .	26
<b>Figure S3.</b> <sup>1</sup> H NMR (300 MHz, CDCl <sub>3</sub> ) of compound <b>1</b>	6	<b>Figure S24.</b> HMBC (500 MHz, CD <sub>3</sub> OD) of compound <b>4</b> .	27
<b>Figure S4.</b> <sup>13</sup> C NMR (75 MHz, CDCl <sub>3</sub> ) of compound <b>1</b>	7	<b>Figure S25.</b> HRMS of compound <b>1</b> .	28
<b>Figure S5.</b> <sup>1</sup> H- <sup>1</sup> H COSY (300 MHz, CDCl <sub>3</sub> ) of compound <b>1</b>	8	<b>Figure S26.</b> HRMS of compound <b>2</b> .	29
<b>Figure S6.</b> HSQC (300 MHz, CDCl <sub>3</sub> ) of compound <b>1</b> .	9	<b>Figure S27.</b> HRMS of compound <b>3</b> .	30
<b>Figure S7.</b> HMBC (300 MHz, CDCl <sub>3</sub> ) of compound <b>1</b> .	10	<b>Figure S28.</b> HRMS of compound <b>4</b> .	31
<b>Figure S8.</b> <sup>1</sup> H NMR (300 MHz, CD <sub>3</sub> OD) of compound <b>2</b> .	11	<b>Figure S29.</b> IR of compound <b>1</b> .	32
<b>Figure S9.</b> <sup>13</sup> C NMR (75 MHz, CD <sub>3</sub> OD) of compound <b>2</b> .	12	<b>Figure S30.</b> IR of compound <b>2</b> .	33
<b>Figure S10.</b> <sup>1</sup> H- <sup>1</sup> H COSY (300 MHz, CD <sub>3</sub> OD) compound <b>2</b>	13	<b>Figure S31.</b> IR of compound <b>3</b> .	34
<b>Figure S11.</b> HSQC (300 MHz, CD <sub>3</sub> OD) of compound <b>2</b> .	14	<b>Figure S32.</b> IR of compound <b>4</b> .	35
<b>Figure S12.</b> HMBC (300 MHz, CD <sub>3</sub> OD) of compound <b>2</b> .	15		
<b>Figure S13.</b> NOESY (300 MHz, CD <sub>3</sub> OD) of compound <b>2</b> .	16		
<b>Figure S14.</b> <sup>1</sup> H NMR (300 MHz, CD <sub>3</sub> OD) of compounds <b>3</b> and <b>4</b> .	17		
<b>Figure S15.</b> <sup>1</sup> H NMR (500 MHz, CD <sub>3</sub> OD) of compound <b>3</b> .	18		
<b>Figure S16.</b> <sup>13</sup> C NMR (75 MHz, CD <sub>3</sub> OD) of compound <b>3</b>	19		
<b>Figure S17.</b> <sup>1</sup> H- <sup>1</sup> H COSY (500 MHz, CD <sub>3</sub> OD) of compound <b>3</b>	20		
<b>Figure S18.</b> HSQC (500 MHz, CD <sub>3</sub> OD) of compound <b>3</b> .	21		
<b>Figure S19.</b> HMBC (500 MHz, CD <sub>3</sub> OD) of compound <b>3</b> .	22		

Table S1. <sup>1</sup>H and <sup>13</sup>C NMR data of compounds **2** and **prelactone C**<sup>d</sup>

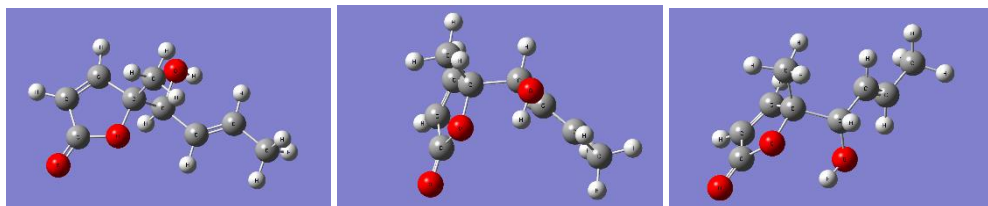
No	<b>2</b> <sup>a</sup>		<b>prelactone C</b> <sup>b</sup>		<b>prelactone C</b> <sup>c</sup>	
	$\delta_{\text{H}}$ (mult., J in Hz)	$\delta_{\text{C}}$	$\delta_{\text{H}}$ (mult., J in Hz)	$\delta_{\text{C}}$	$\delta_{\text{H}}$ (mult., J in Hz)	$\delta_{\text{C}}$
1	-	173.4	-	-	-	170.2
2	2.42 (dd, J = 18.3, 9.0) 2.81 (dd, J = 18.3, 6.6)	36.4	2.40 (dd, J = 17, 7) 2.91 (dd, J = 17, 6)	39.1	2.50 (dd, J = 17, 8) 2.94 (dd, J = 17, 6)	39.1
3	4.20 (m)	67.4	3.72 (ddq, J = 7,7,6)	69.6	3.79 (m)	69.6
4	2.16 (m)	38.9	1.60 (ddq, J = 10, 7, 6)	41.5	1.68 (m)	41.5
5	4.81 (dd, J = 6.9, 2.1)	83.2	4.30 (dd, J = 10, 8)	84.1	4.21 (m)	84.1
6	5.66 (dd, J = 15.3, 6.9)	128.6	5.46 (ddq, J = 15, 8, 2)	127.6	5.46 (ddq, J = 15, 8, 2)	127.6
7	5.84 (dq, J = 15.3, 6.3)	130.6	5.83 (ddq, J = 15, 6, 1)	132.6	5.81 (ddq, J = 15, 6, 1)	132.6
8	1.75 (d, J = 6.3)	17.9	1.75 (dd, J = 6.5, 2)	17.7	1.76 (dd, J = 6.5, 2)	17.7
9	0.92 (d, J = 7.2)	5.9	1.04 (d, J = 6.5)	13.7	1.04 (d, J = 7.0)	13.7

<sup>a</sup>: Recorded for <sup>1</sup>H NMR at 300MHz/ <sup>13</sup>C NMR at 75MHz in CD<sub>3</sub>OD; <sup>b</sup>: Recorded for <sup>1</sup>H NMR at 500MHz in CD<sub>3</sub>OD; <sup>c</sup>: Recorded for <sup>1</sup>H NMR at 200MHz / <sup>13</sup>C NMR at 50MHz in CDCl<sub>3</sub>; <sup>d</sup>: The NMR data of Prelactone C were taken from the literature, Bindseil, Kai U., and Axel Zeeck. "Metabolic products of microorganisms. Part 265. Prelactones C and B, oligoketides from Streptomyces producing concanamycins and bafilomycins." *Helvetica chimica acta* 76.1 (1993): 150-157.

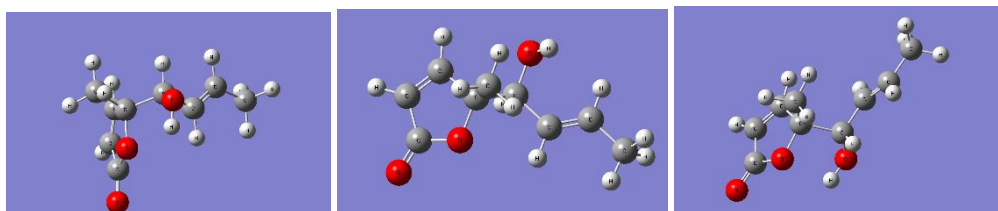
Table S2. Anti-fouling and cytotoxicity activities of compounds **1–4**

Compounds	Antifouling ( <i>Balanus amphitrite</i> )			Cytotoxicity (HeLa)
	EC <sub>50</sub> (μg/mL)	LC <sub>50</sub> (μg/mL)	LC <sub>50</sub> /EC <sub>50</sub>	IC <sub>50</sub> (μg/mL)
<b>1</b>	12.8	>25	>1.9	>25
<b>2</b>	>25	>25	-	>25
<b>3</b>	>25	>25	-	>25
<b>4</b>	>25	>25	-	>25

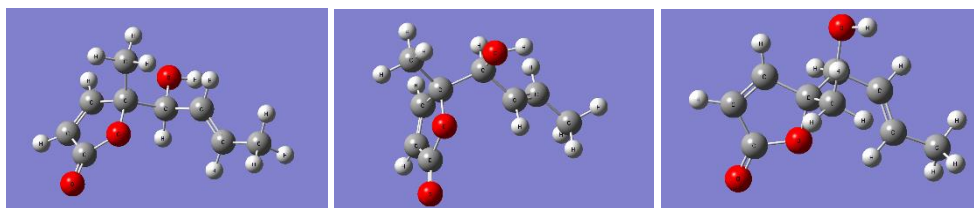
- Not applicable



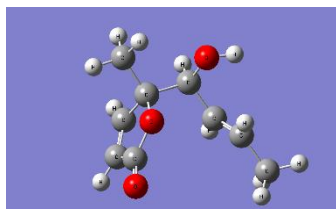
$\Delta G = 0$  kcal/mol, 21%;  $\Delta G = 0.14$  kcal/mol, 17%;  $\Delta G = 0.16$  kcal/mol, 16%;



$\Delta G = 0.22$  kcal/mol, 15%;  $\Delta G = 0.36$  kcal/mol, 11%;  $\Delta G = 0.39$  kcal/mol, 11%;

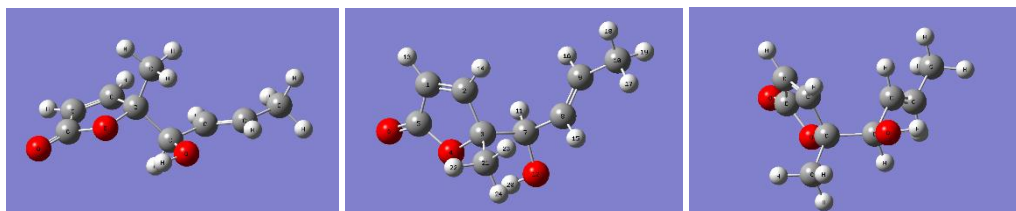


$\Delta G = 0.54$  kcal/mol, 8%;  $\Delta G = 2.00$  kcal/mol, 0.7%;  $\Delta G = 2.85$  kcal/mol, 0.15%;

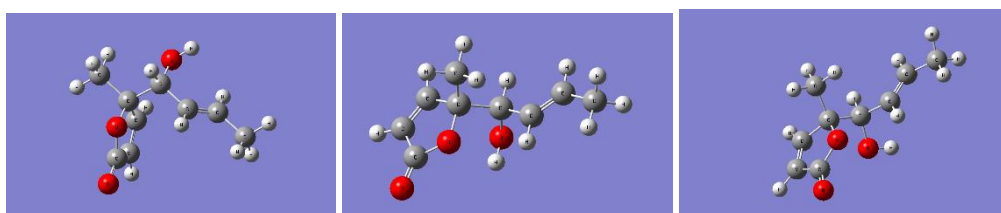


$\Delta G = 2.87$  kcal/mol, 0.15%;

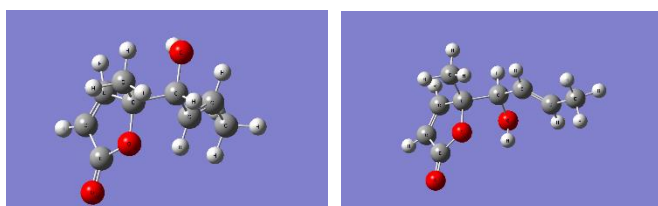
**Figure S1.** The conformers, relative energies ( $\Delta G$ ) and conformer populations of the minimized conformers of (4*R*, 5*S*)-1 within an energy range of 3 kcal/mol.



$\Delta G = 0$  kcal/mol, 43%;     $\Delta G = 0.09$  kcal/mol, 37%;     $\Delta G = 1.09$  kcal/mol, 7%;



$\Delta G = 1.25$  kcal/mol, 5%;     $\Delta G = 1.41$  kcal/mol, 4%;     $\Delta G = 2.02$  kcal/mol, 2%;



$\Delta G = 2.07$  kcal/mol, 1%;     $\Delta G = 2.31$  kcal/mol, 1%.

**Figure S2.** The conformers, relative energies ( $\Delta G$ ) and conformer populations of the minimized conformers of (4*R*, 5*R*)-1 within an energy range of 3 kcal/mol.

Figure S3. <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) of compound **1**.

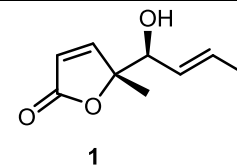
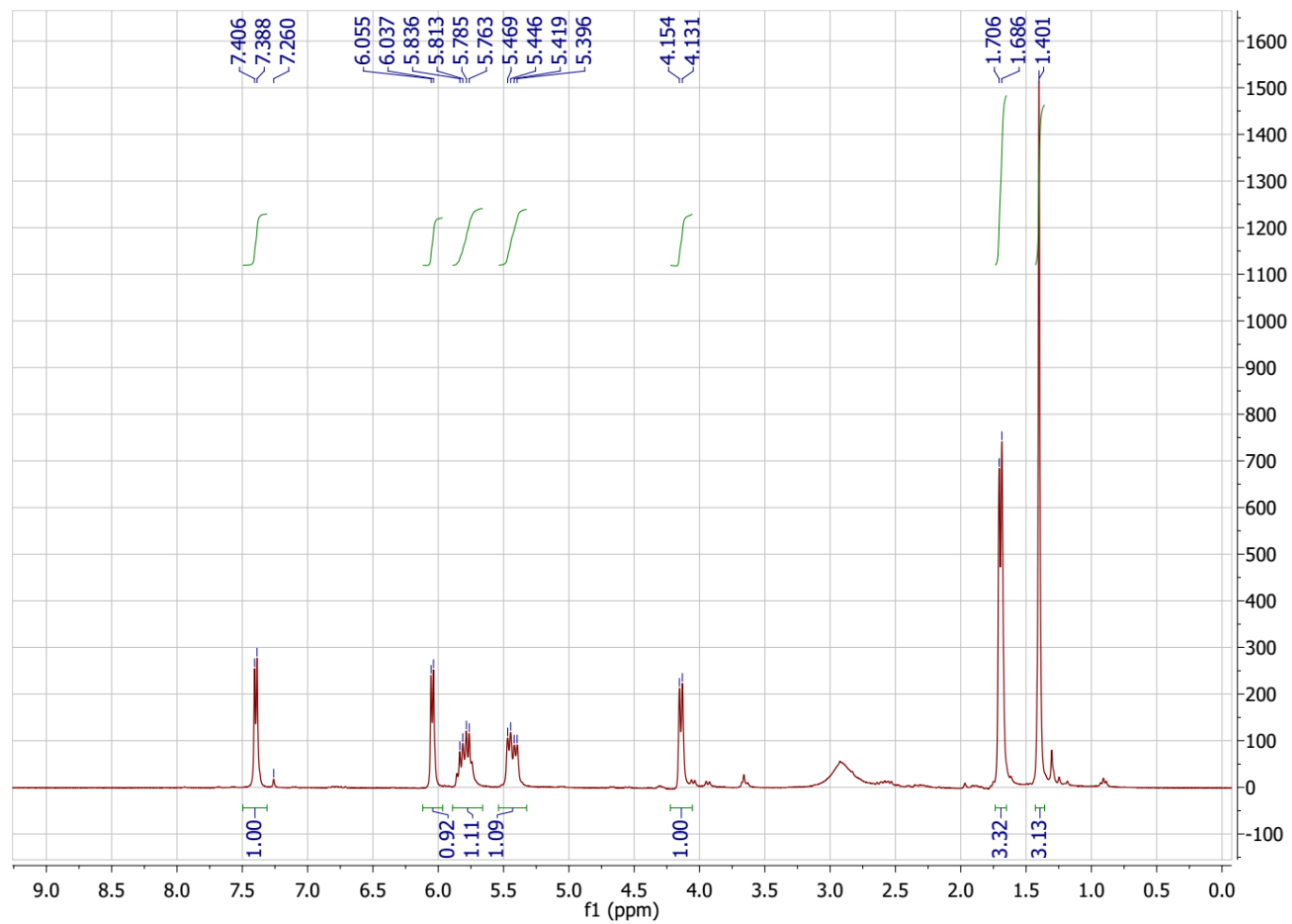


Figure S4.  $^{13}\text{C}$  NMR (75 MHz,  $\text{CDCl}_3$ ) of compound **1**.

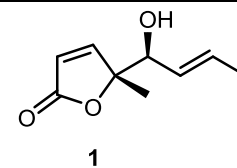
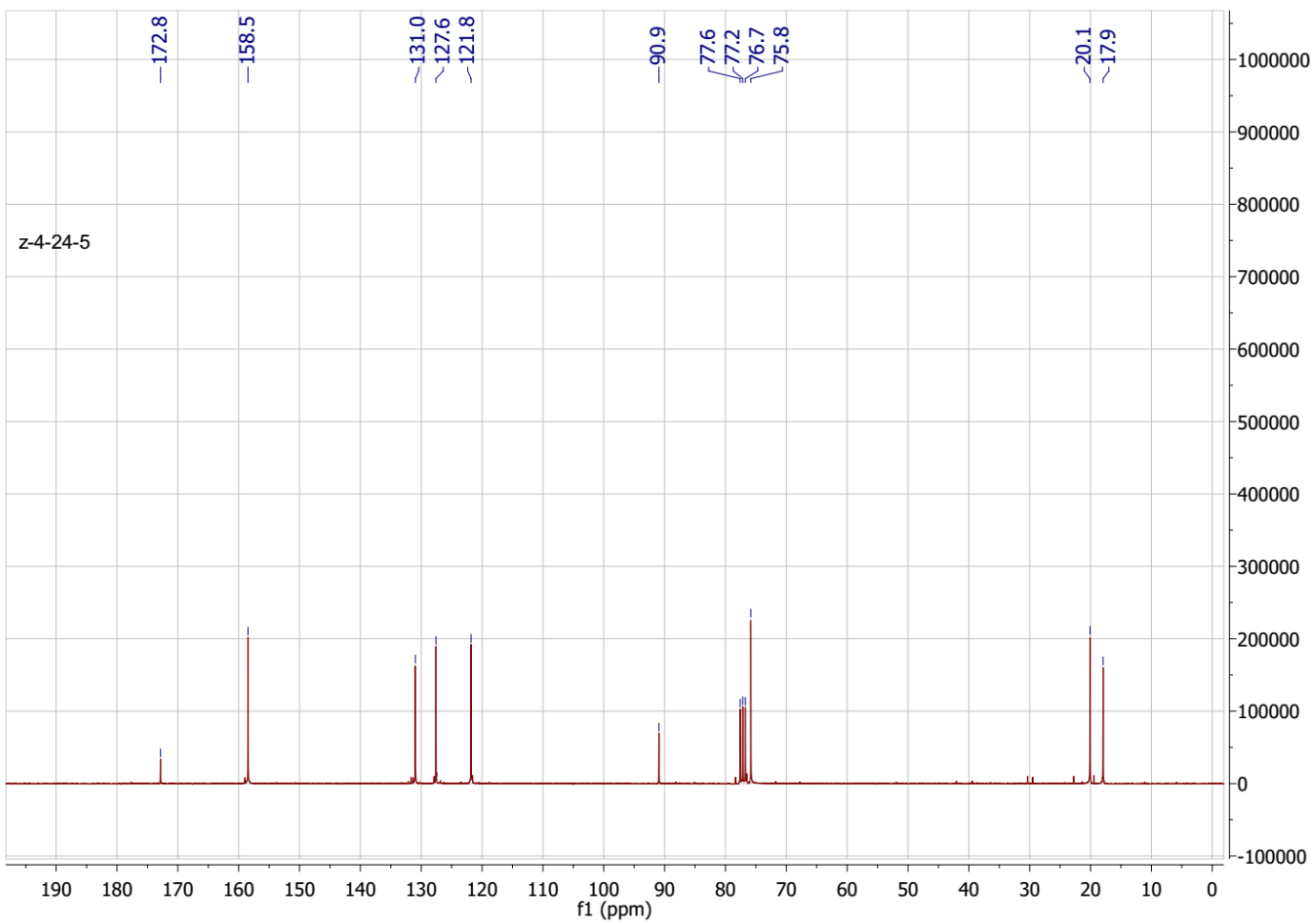


Figure S5.  $^1\text{H}$ - $^1\text{H}$  COSY (300 MHz,  $\text{CDCl}_3$ ) of compound **1**

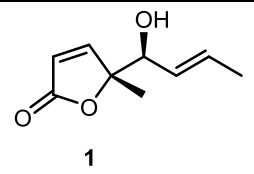
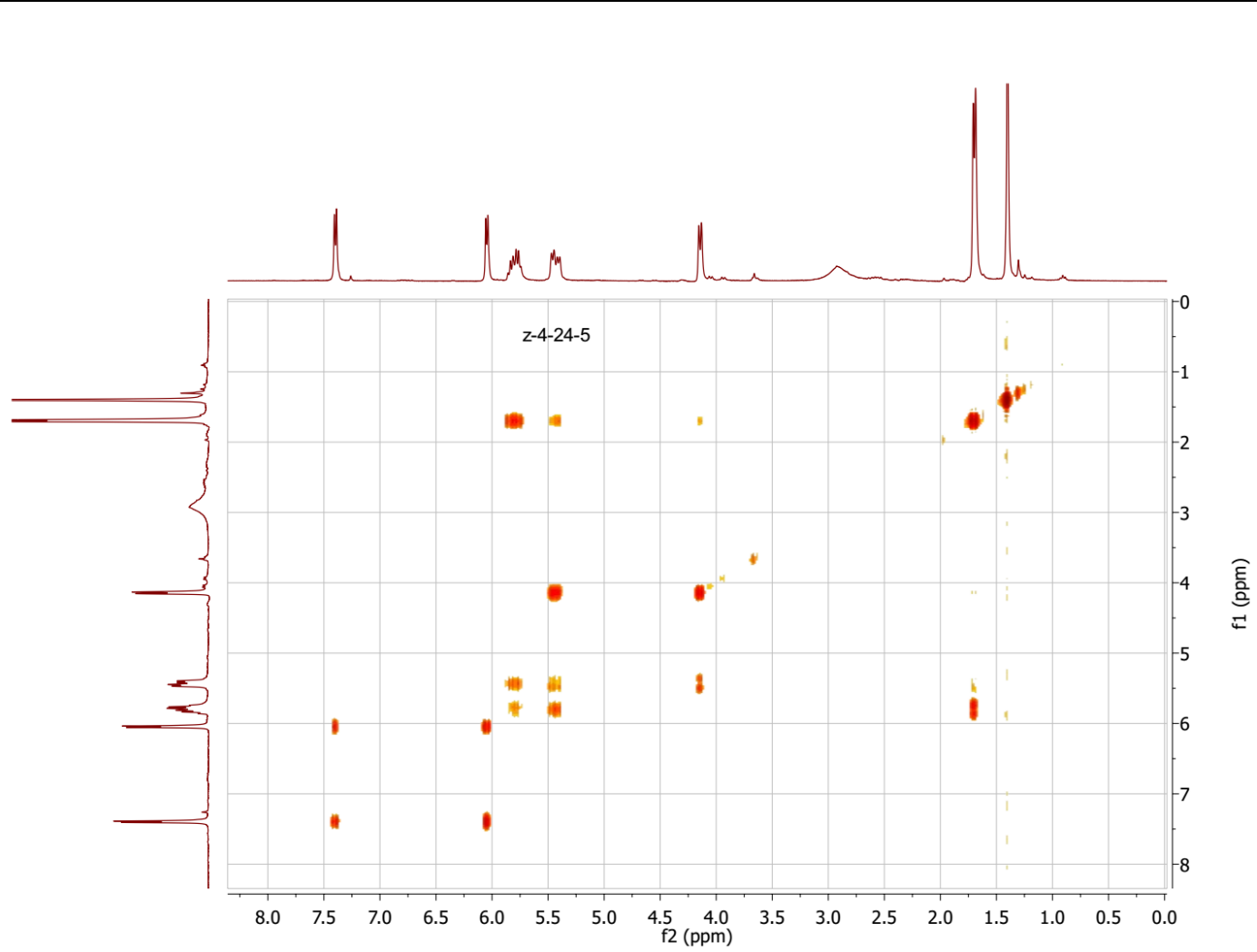




Figure S6. HSQC (300 MHz, CDCl<sub>3</sub>) of compound **1**.

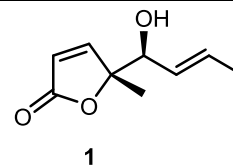
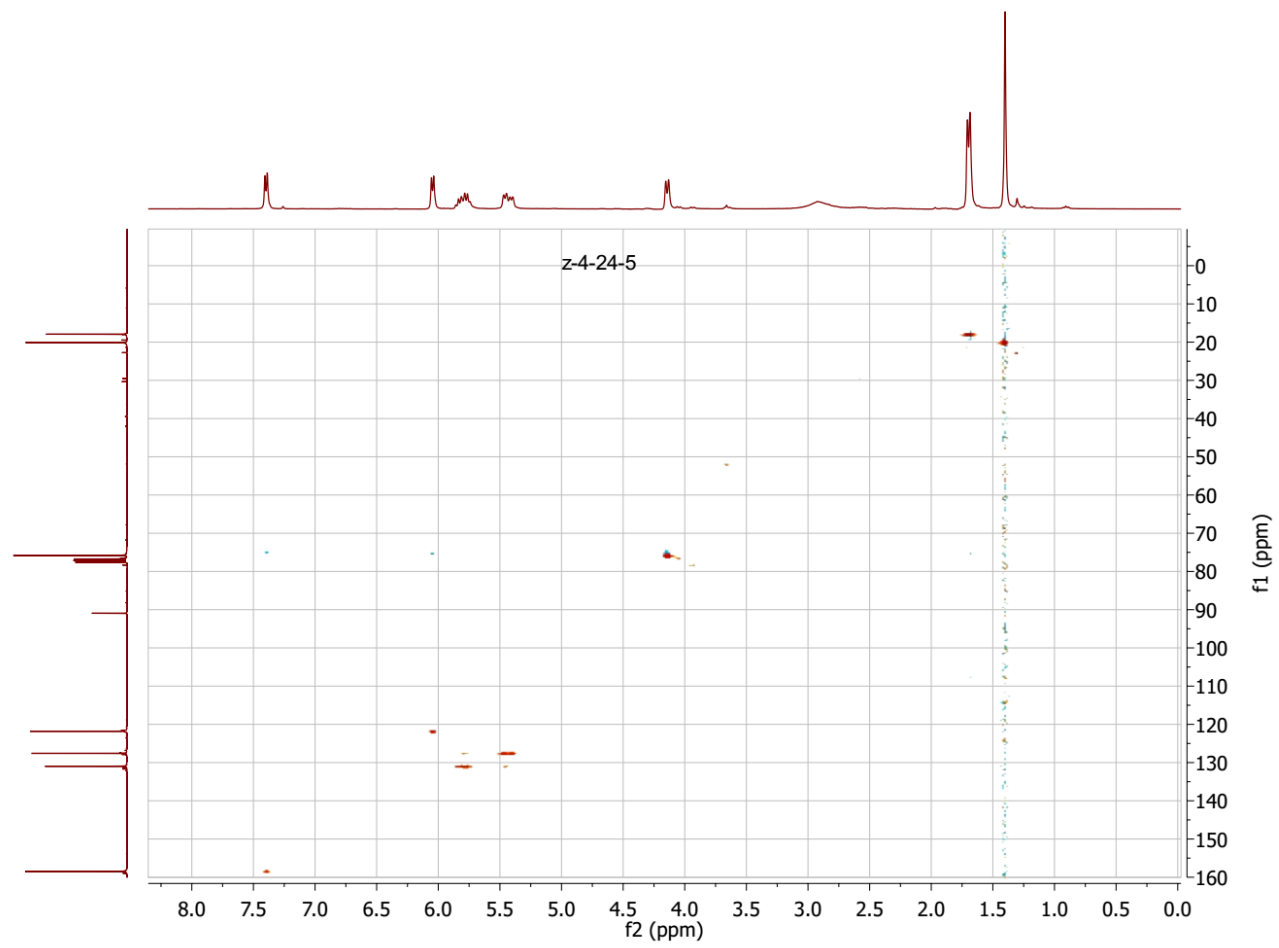


Figure S7. HMBC (300 MHz, CDCl<sub>3</sub>) of compound 1.

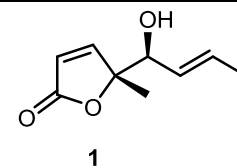
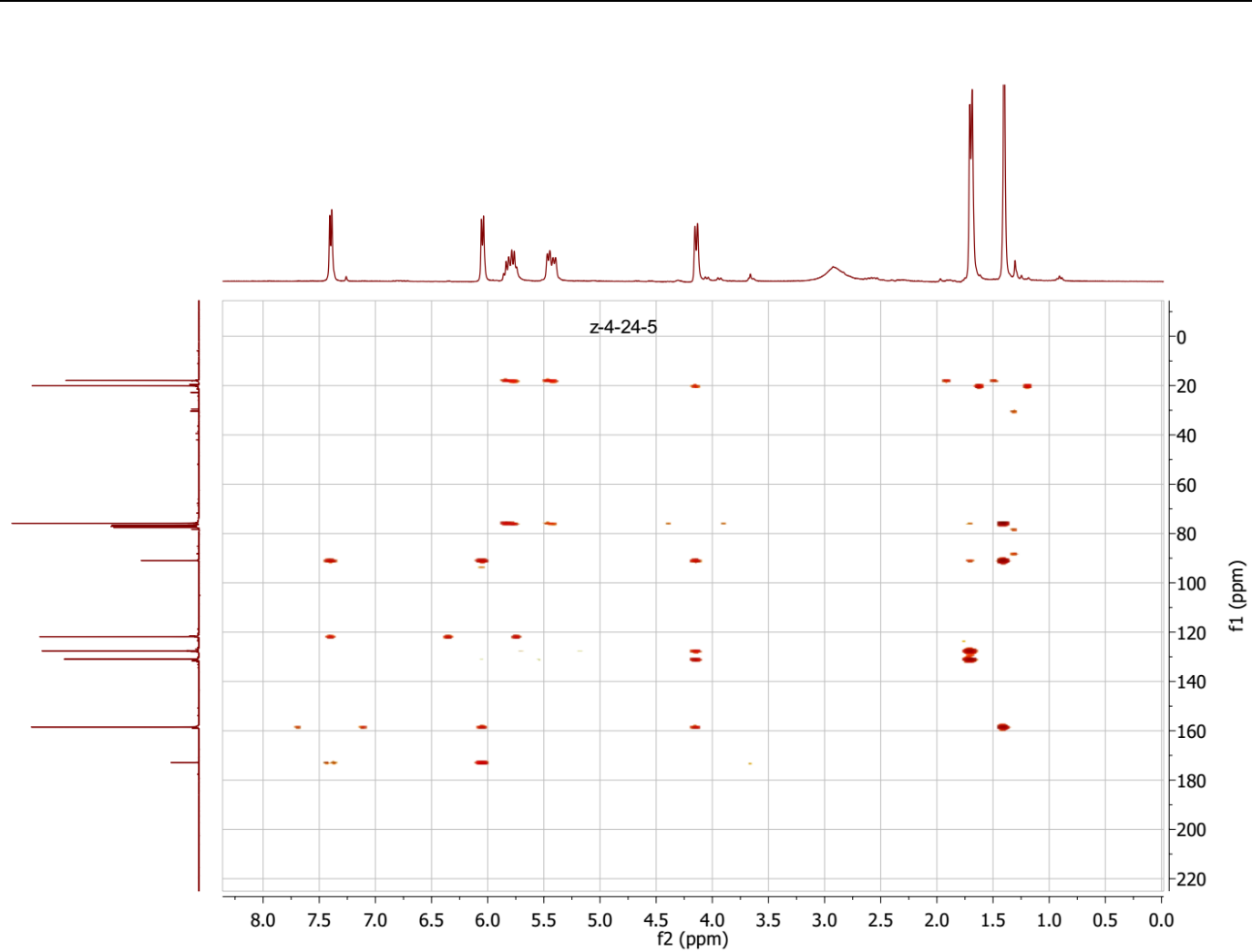
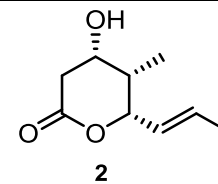
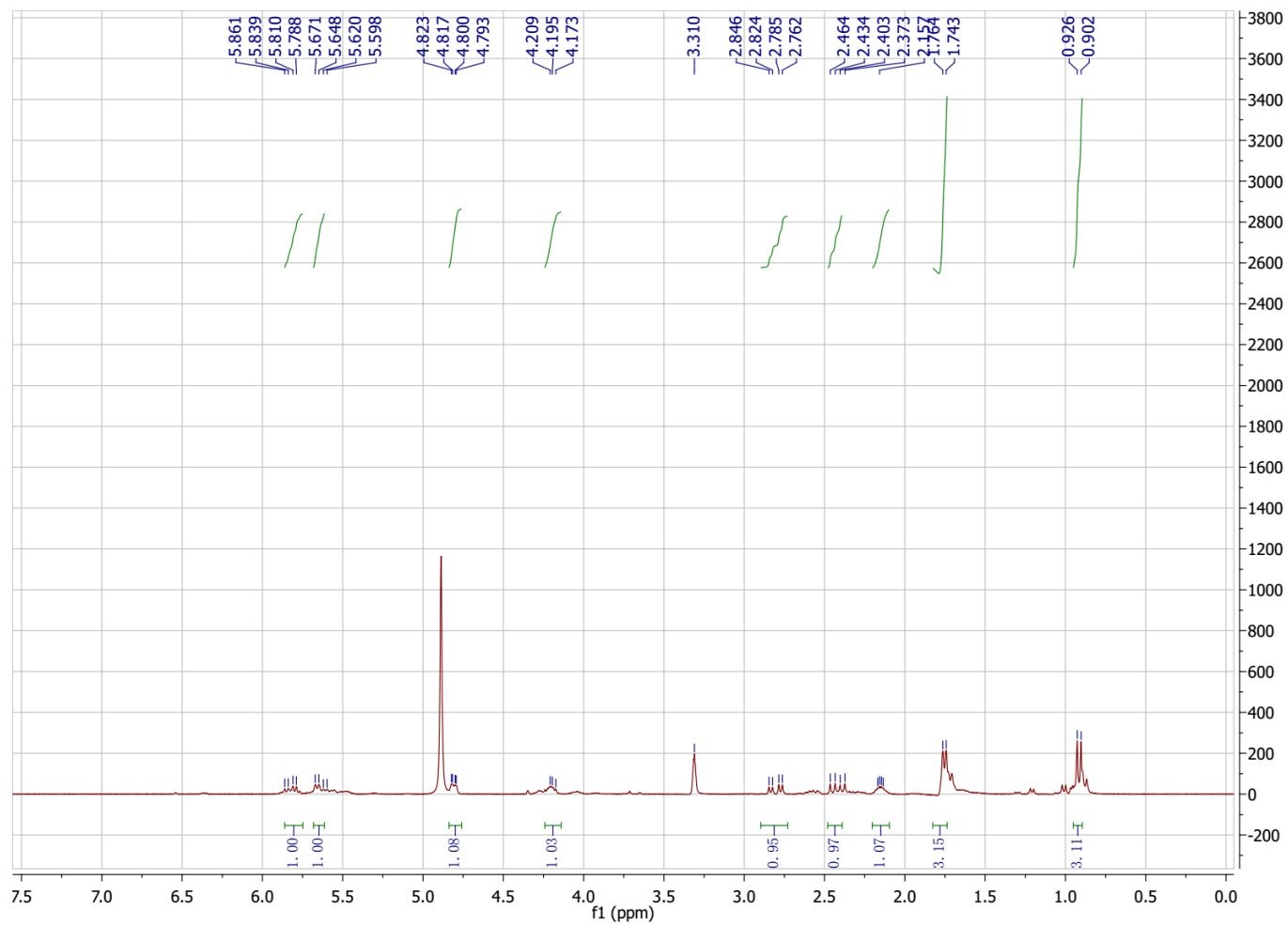


Figure S8. <sup>1</sup>H NMR (300 MHz, CD<sub>3</sub>OD) of compound 2.



**Figure S9.**  $^{13}\text{C}$  NMR (75 MHz,  $\text{CD}_3\text{OD}$ ) of compound **2**.

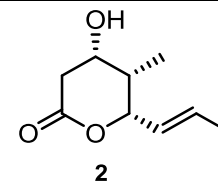
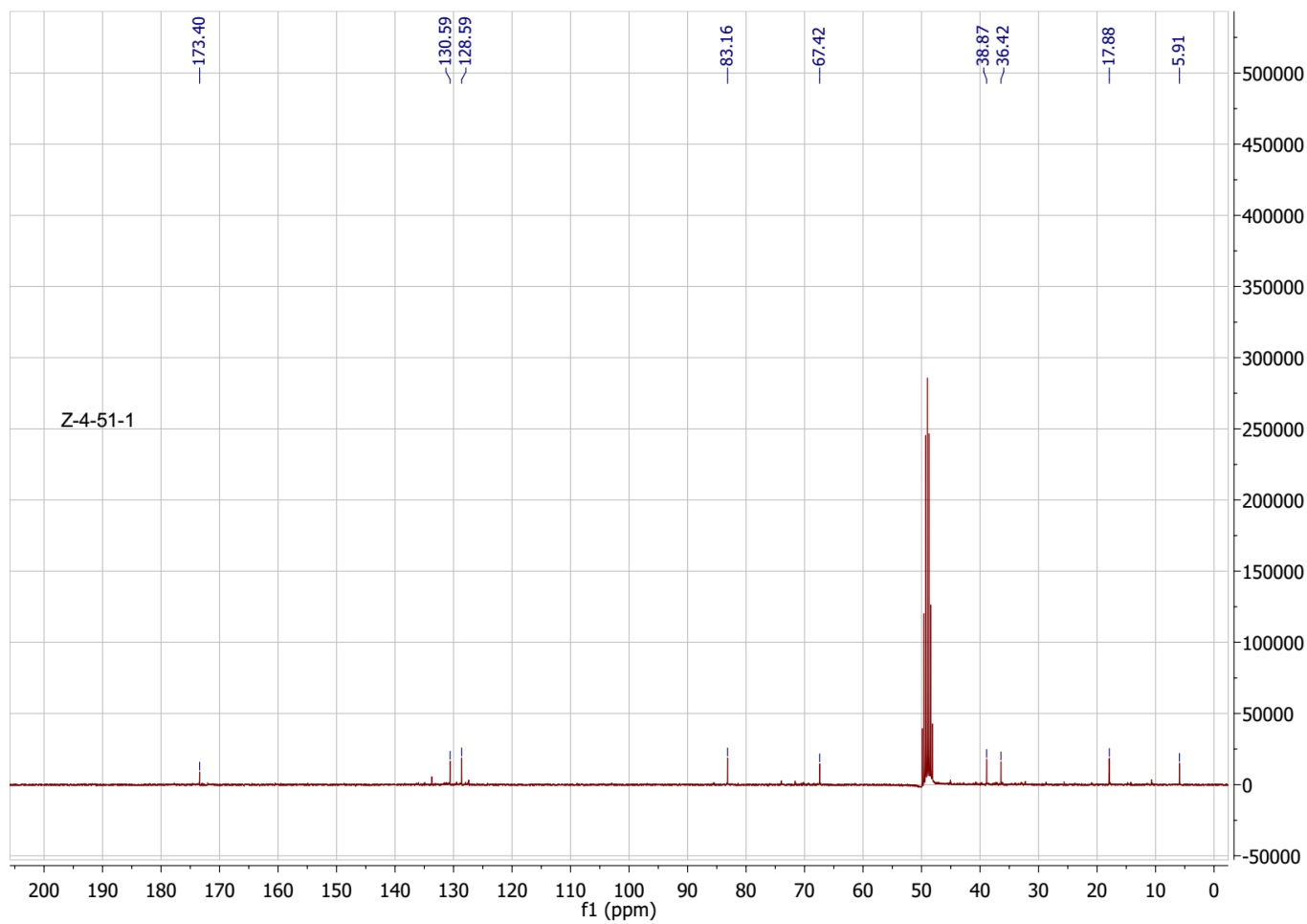


Figure S10. <sup>1</sup>H-<sup>1</sup>H COSY (300 MHz, CD<sub>3</sub>OD) compound 2

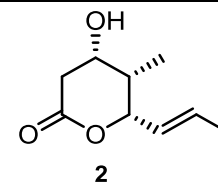
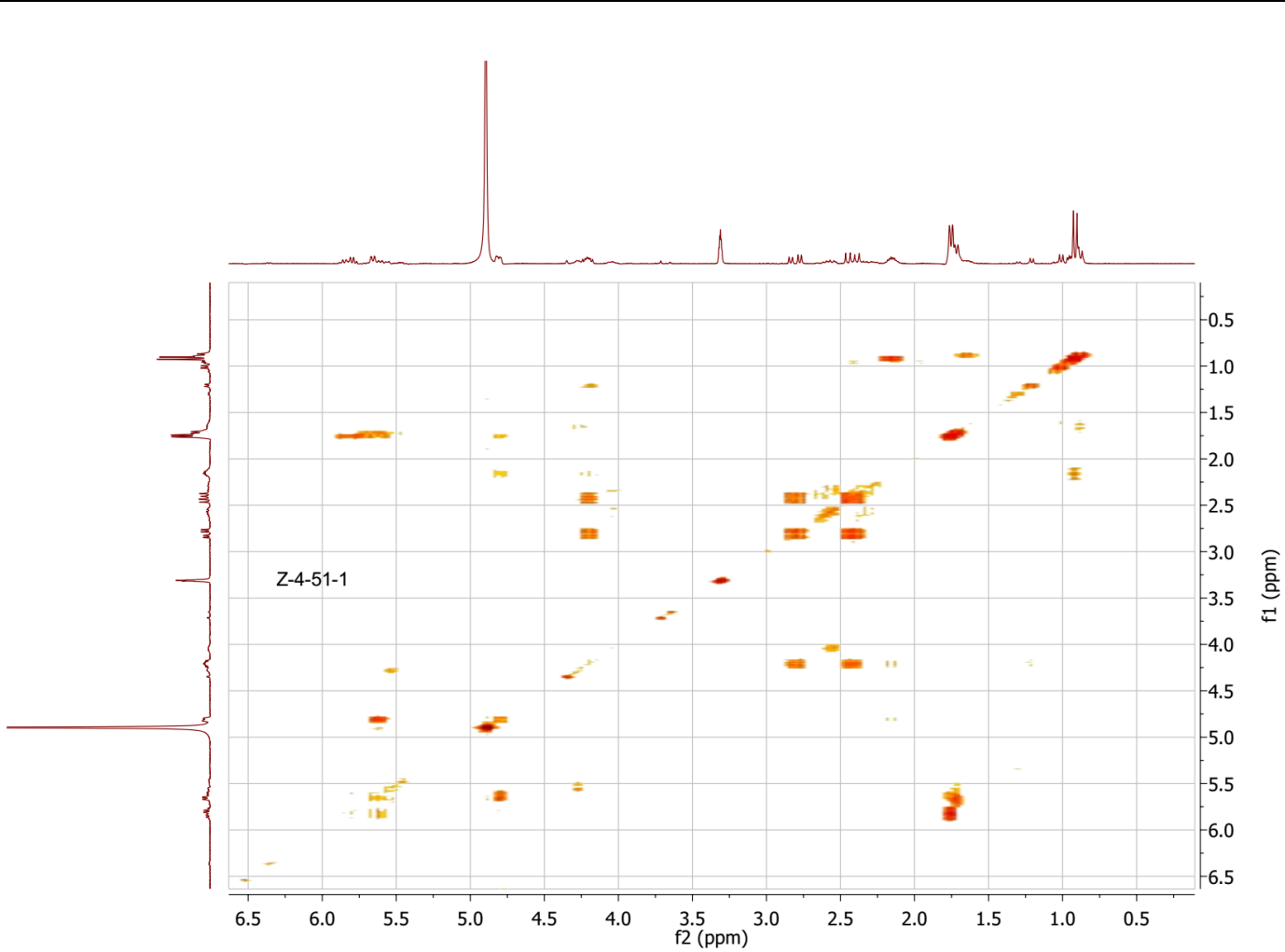


Figure S11. HSQC (300 MHz, CD<sub>3</sub>OD) of compound 2.

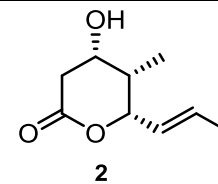
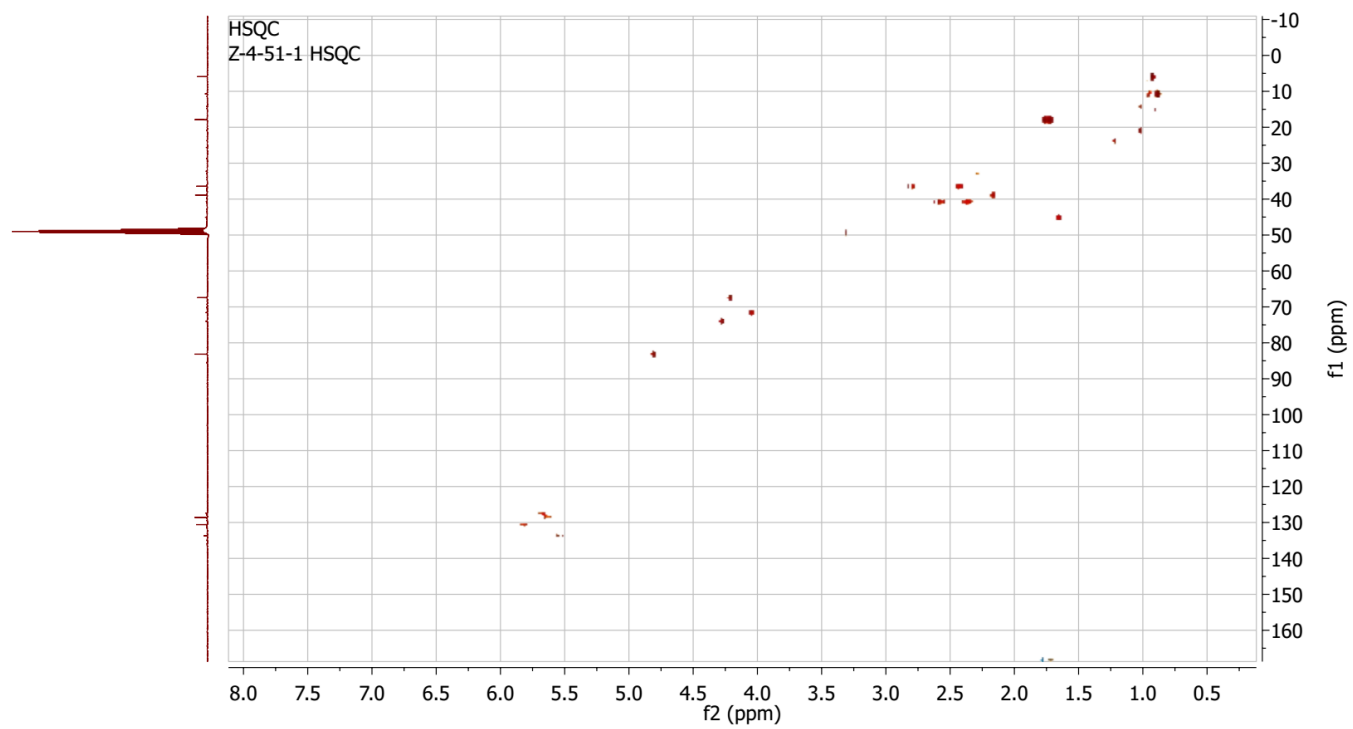




Figure S13. NOESY (300 MHz, CD<sub>3</sub>OD) of compound 2.

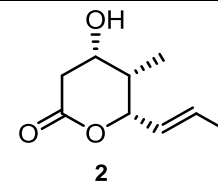
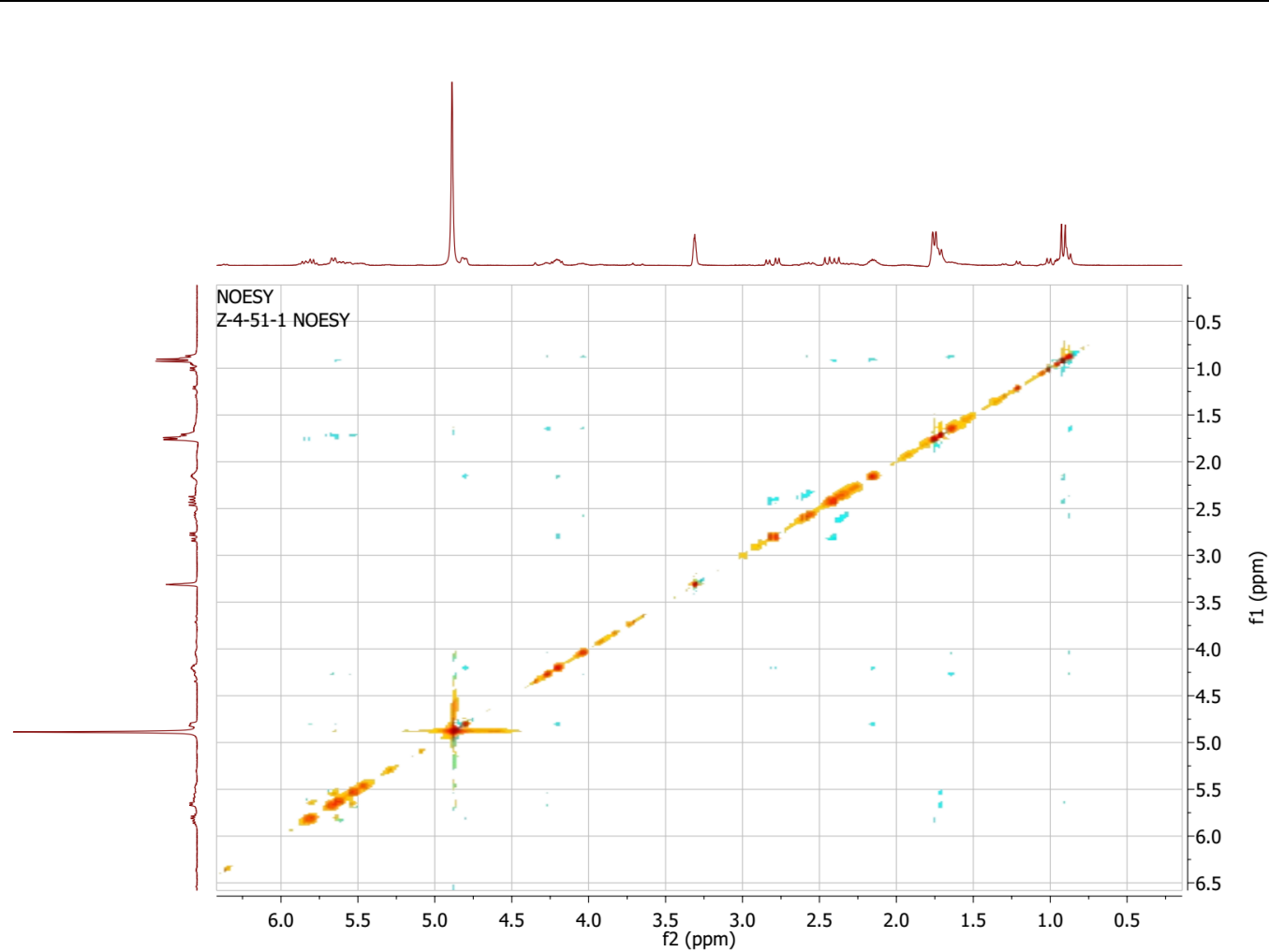




Figure S14. <sup>1</sup>H NMR (500 MHz, CD<sub>3</sub>OD) of compounds **3** and **4** (1:1 ratio).

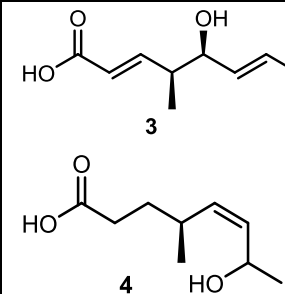
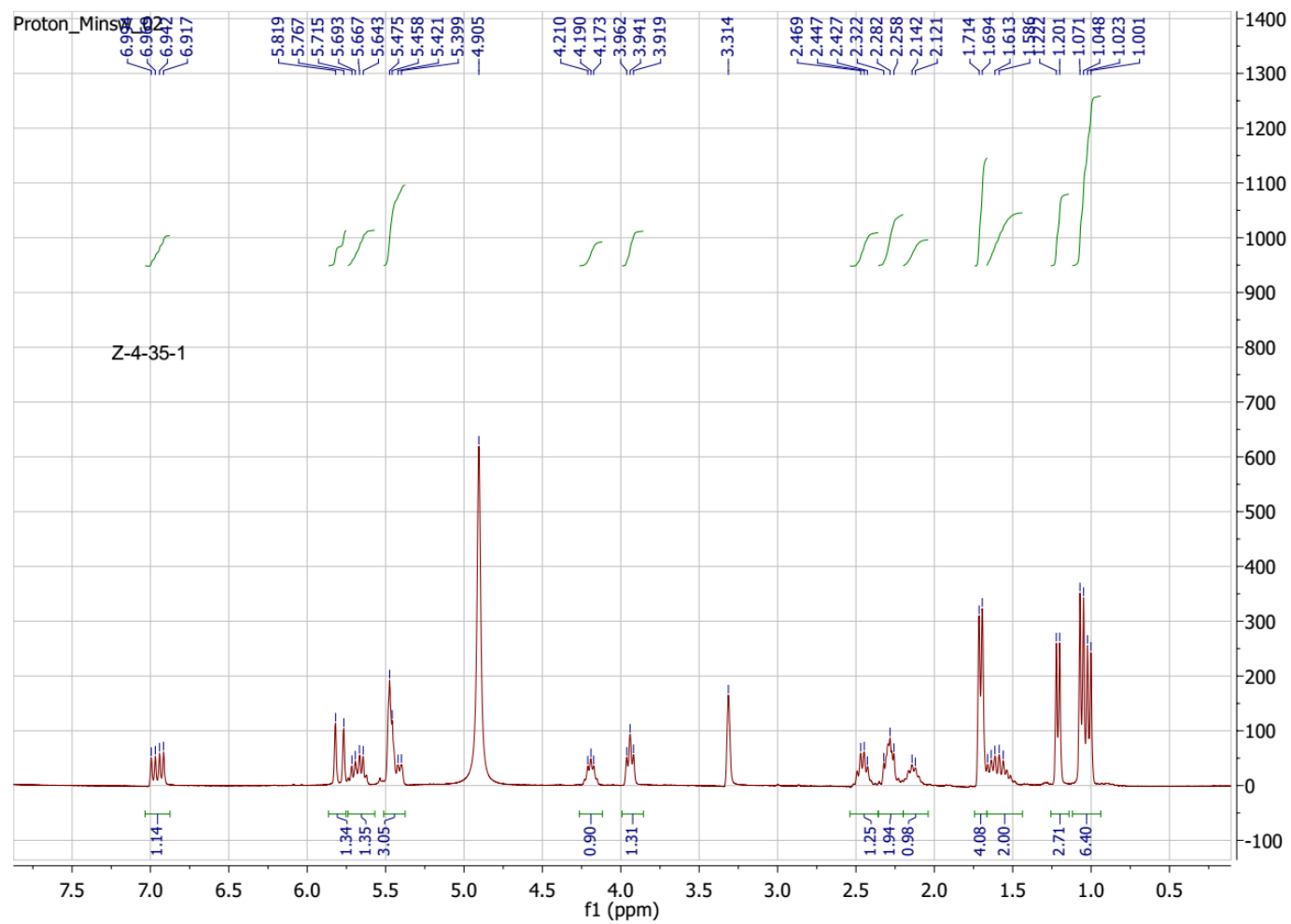


Figure S15. <sup>1</sup>H NMR (500 MHz, CD<sub>3</sub>OD) of compound 3.

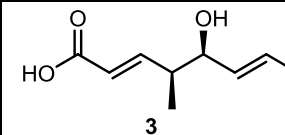
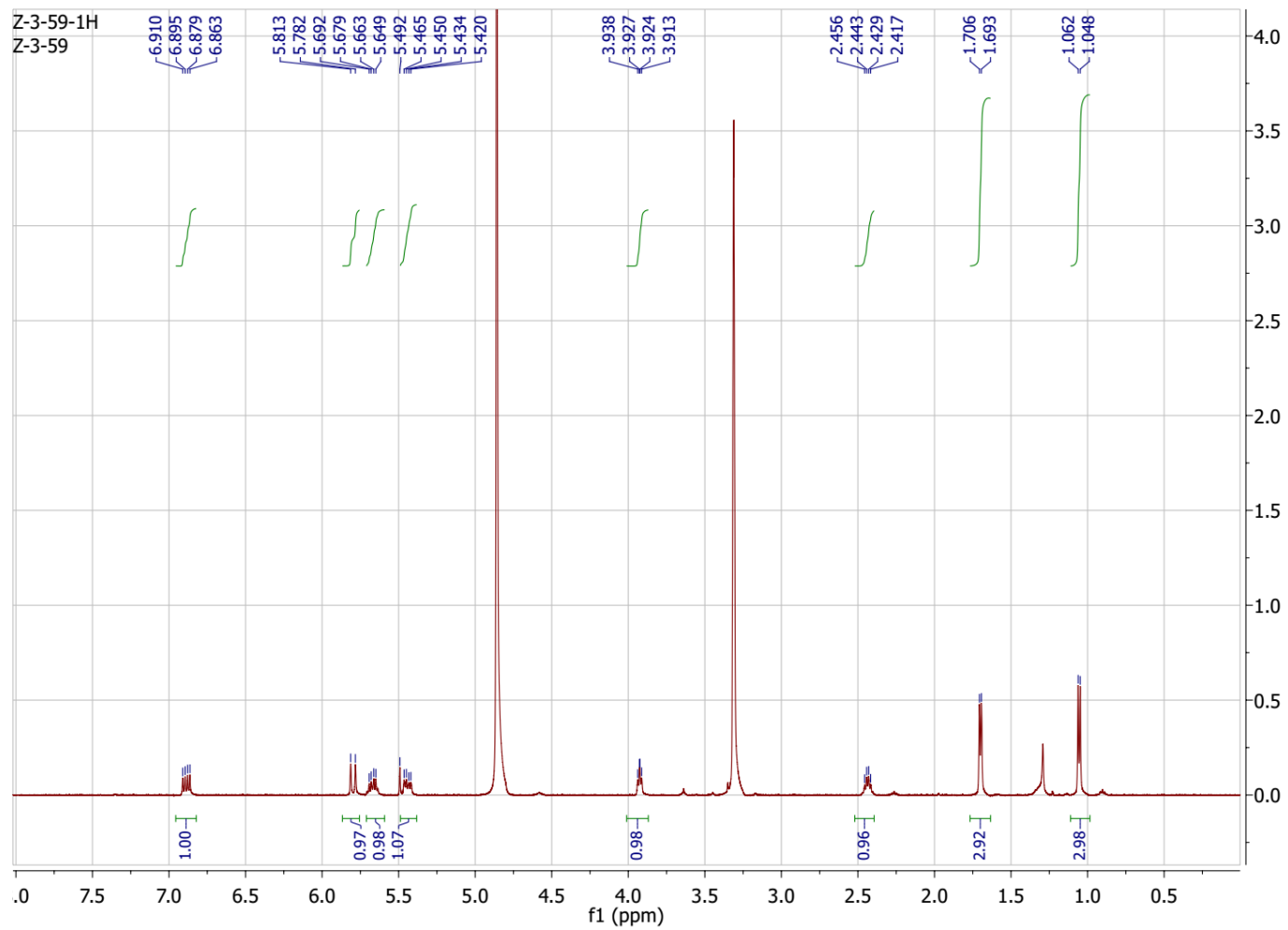


Figure S16. <sup>13</sup>C NMR (75 MHz, CD<sub>3</sub>OD) of compound 3.

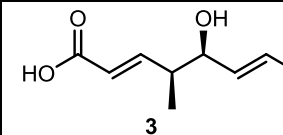
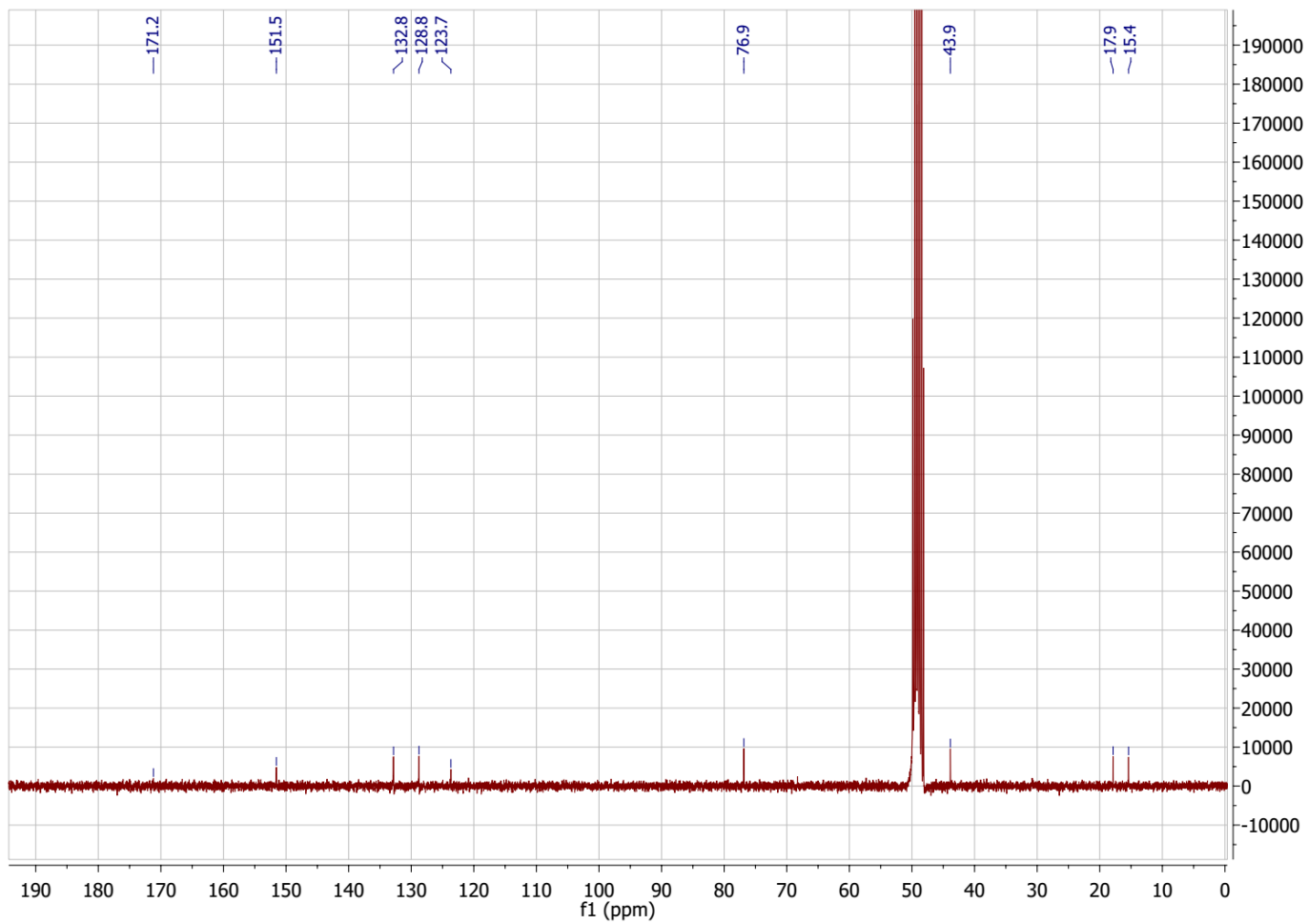


Figure S17.  $^1\text{H}$ - $^1\text{H}$  COSY (500 MHz,  $\text{CD}_3\text{OD}$ ) of compound **3**

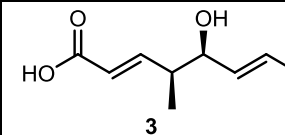
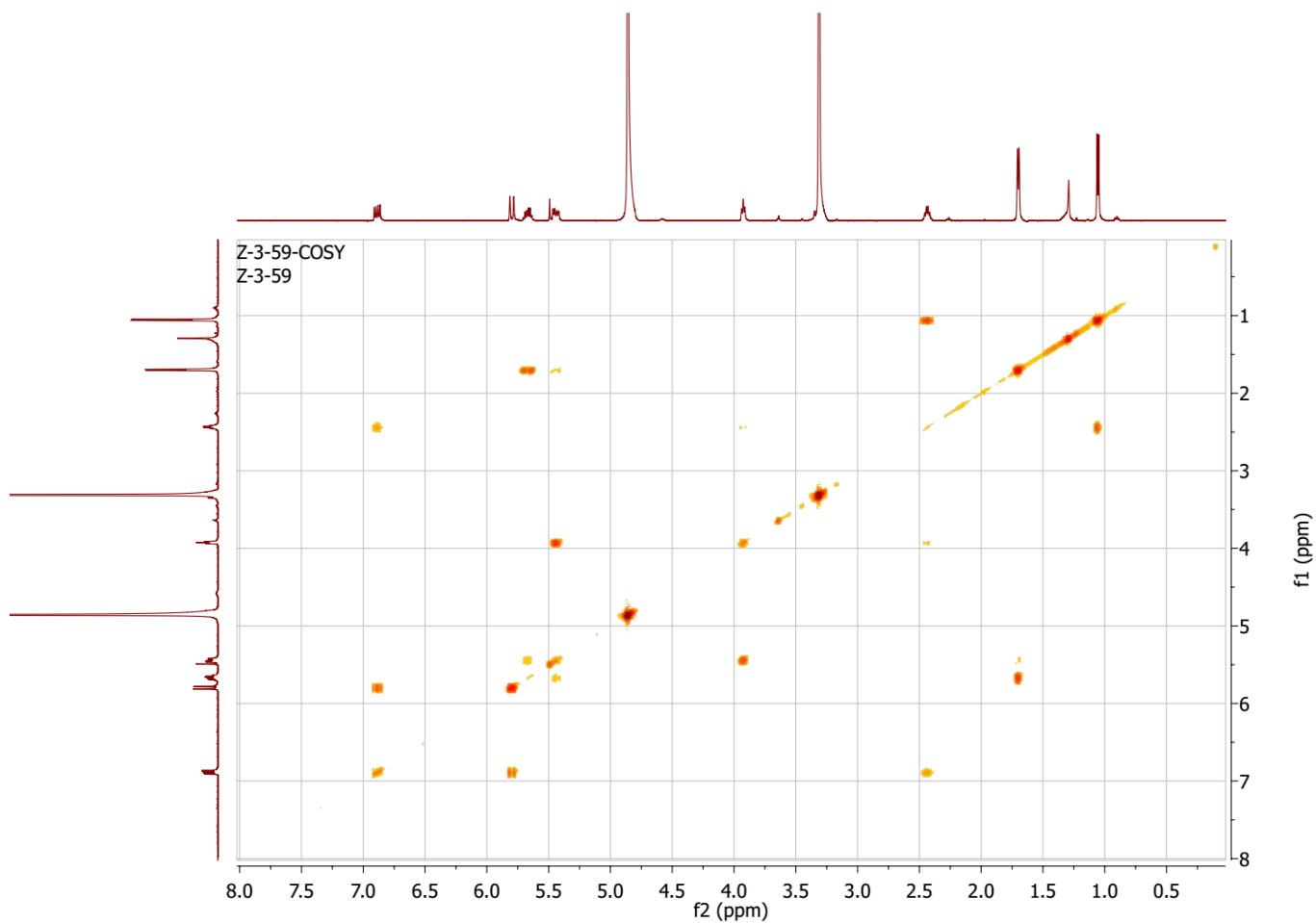


Figure S18. HSQC (500 MHz, CD<sub>3</sub>OD) of compound **3**

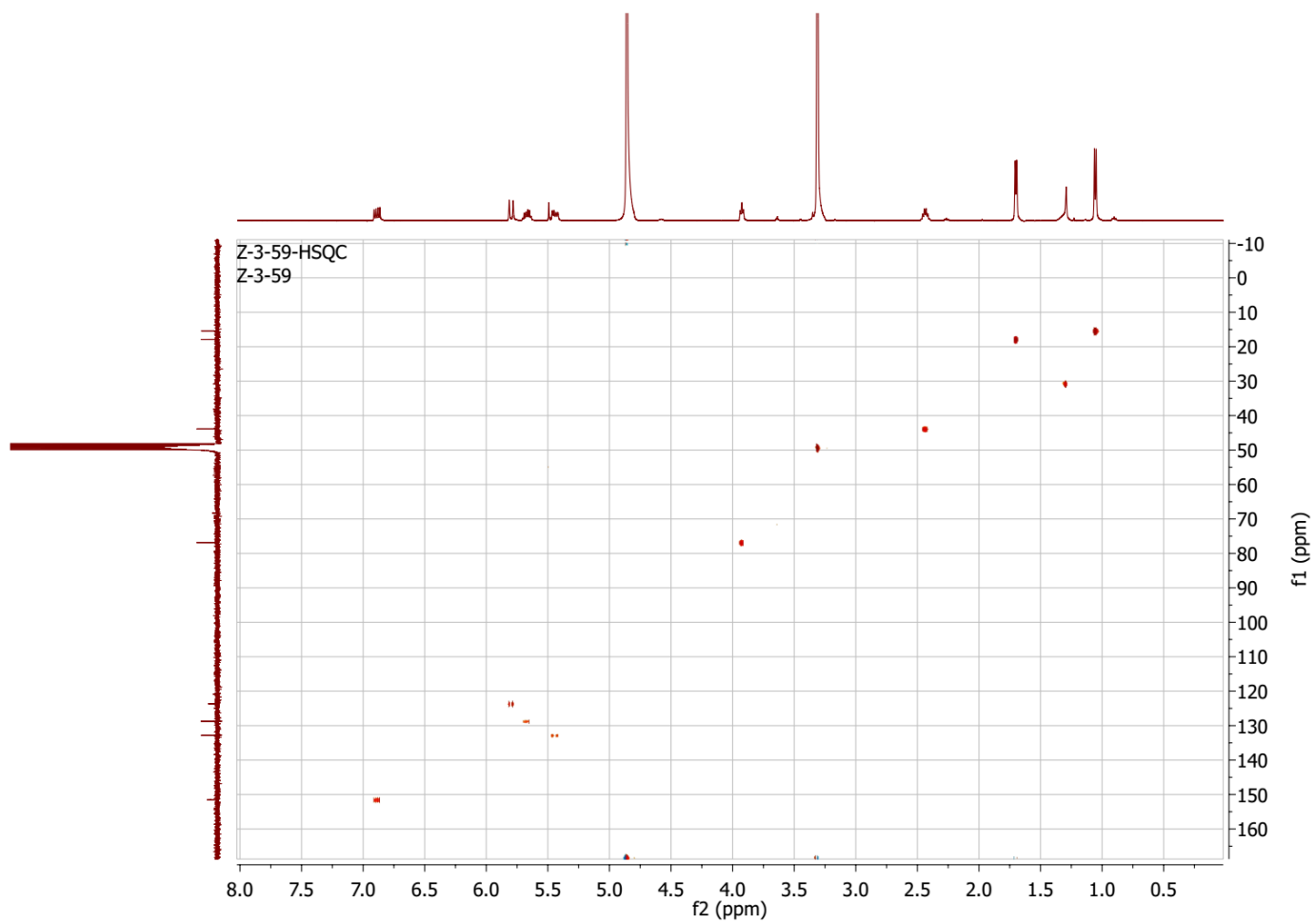
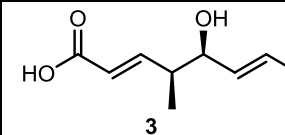


Figure S19. HMBC (500 MHz, CD<sub>3</sub>OD) of compound 3.

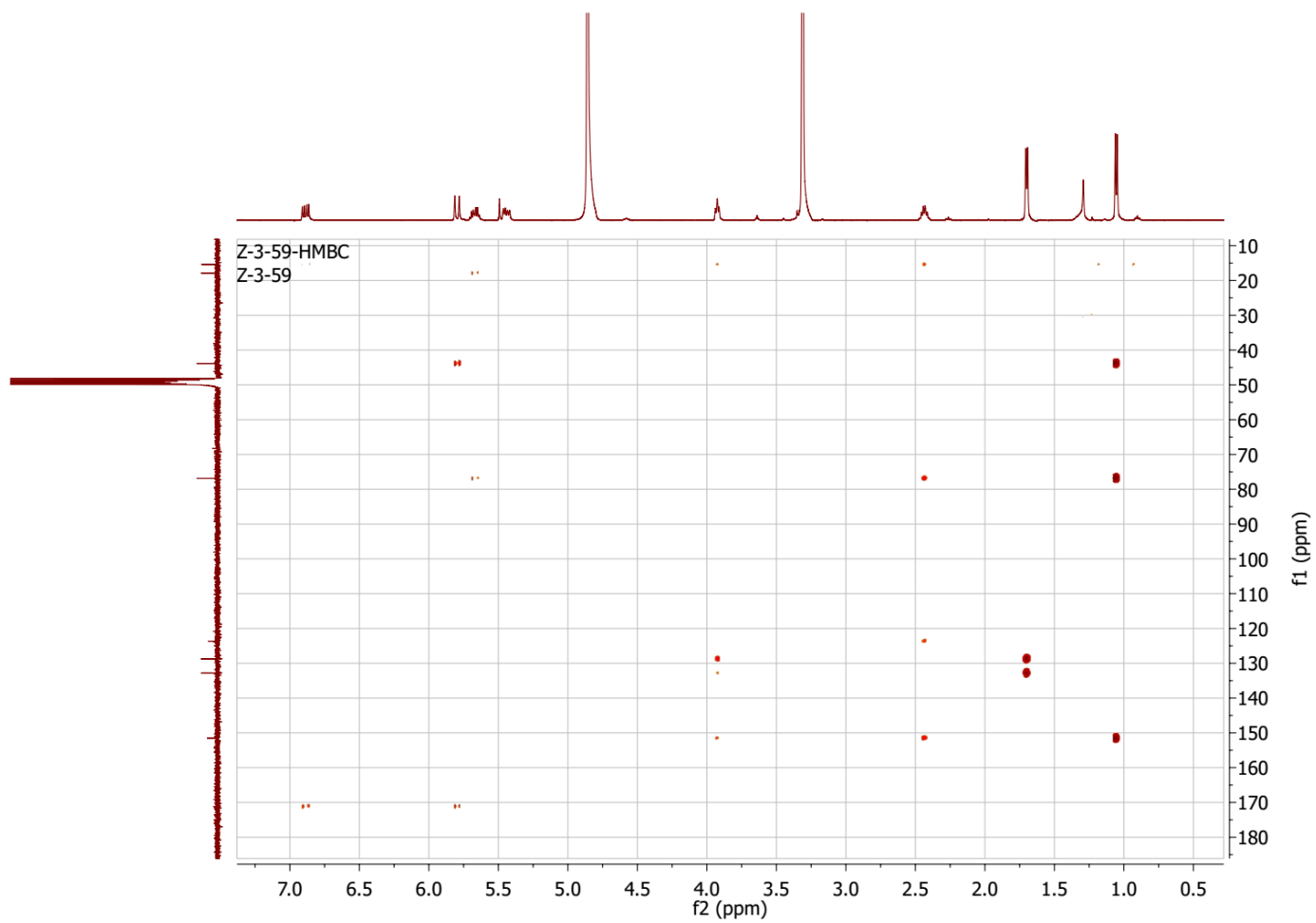
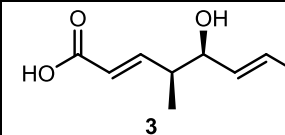


Figure S20. <sup>1</sup>H NMR (500 MHz, CD<sub>3</sub>OD) of compound 4.

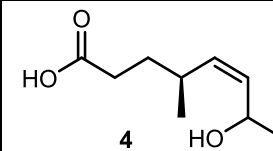
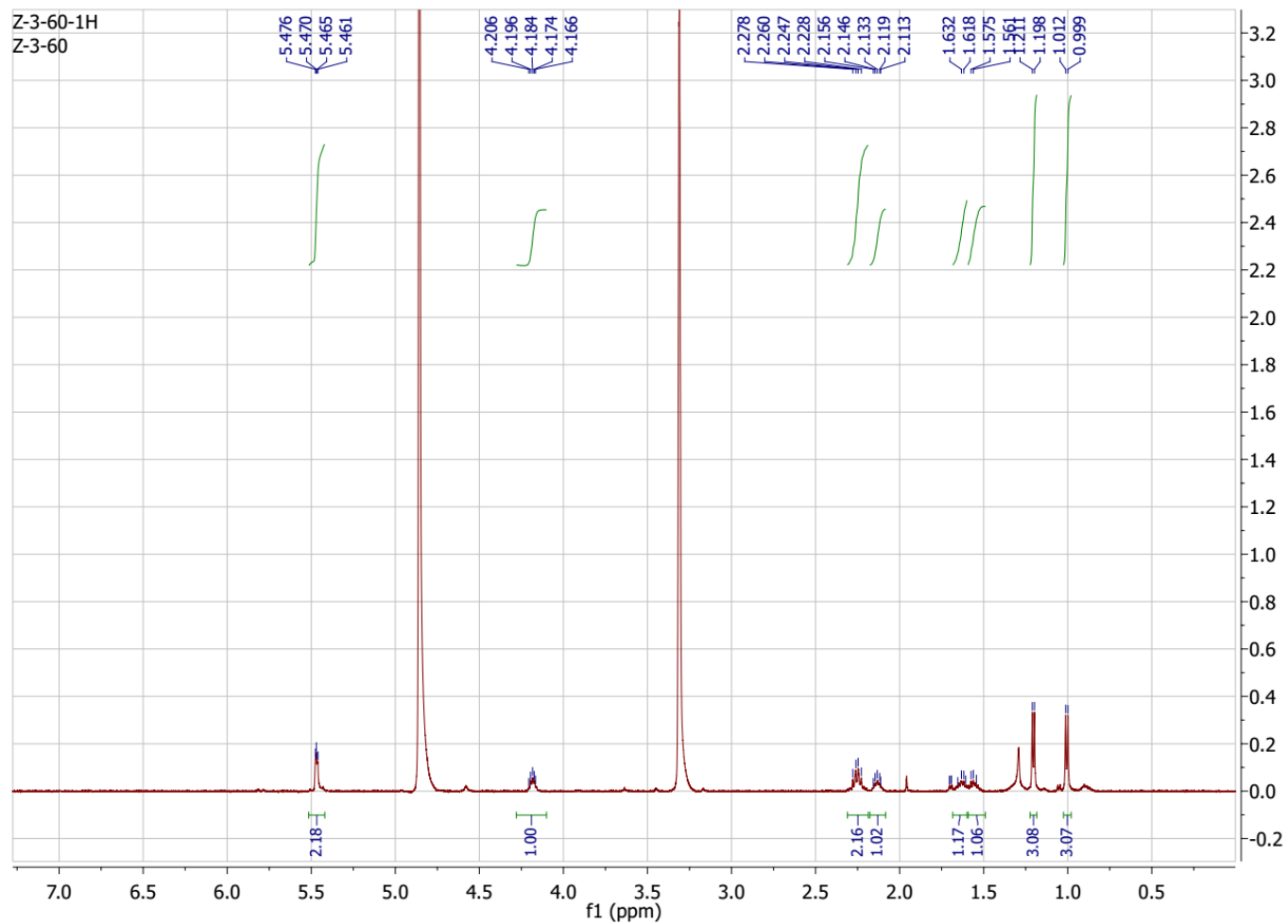


Figure S21.  $^{13}\text{C}$  NMR (75 MHz,  $\text{CD}_3\text{OD}$ ) of compound 4

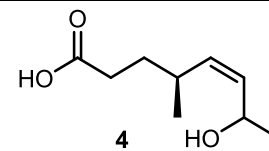
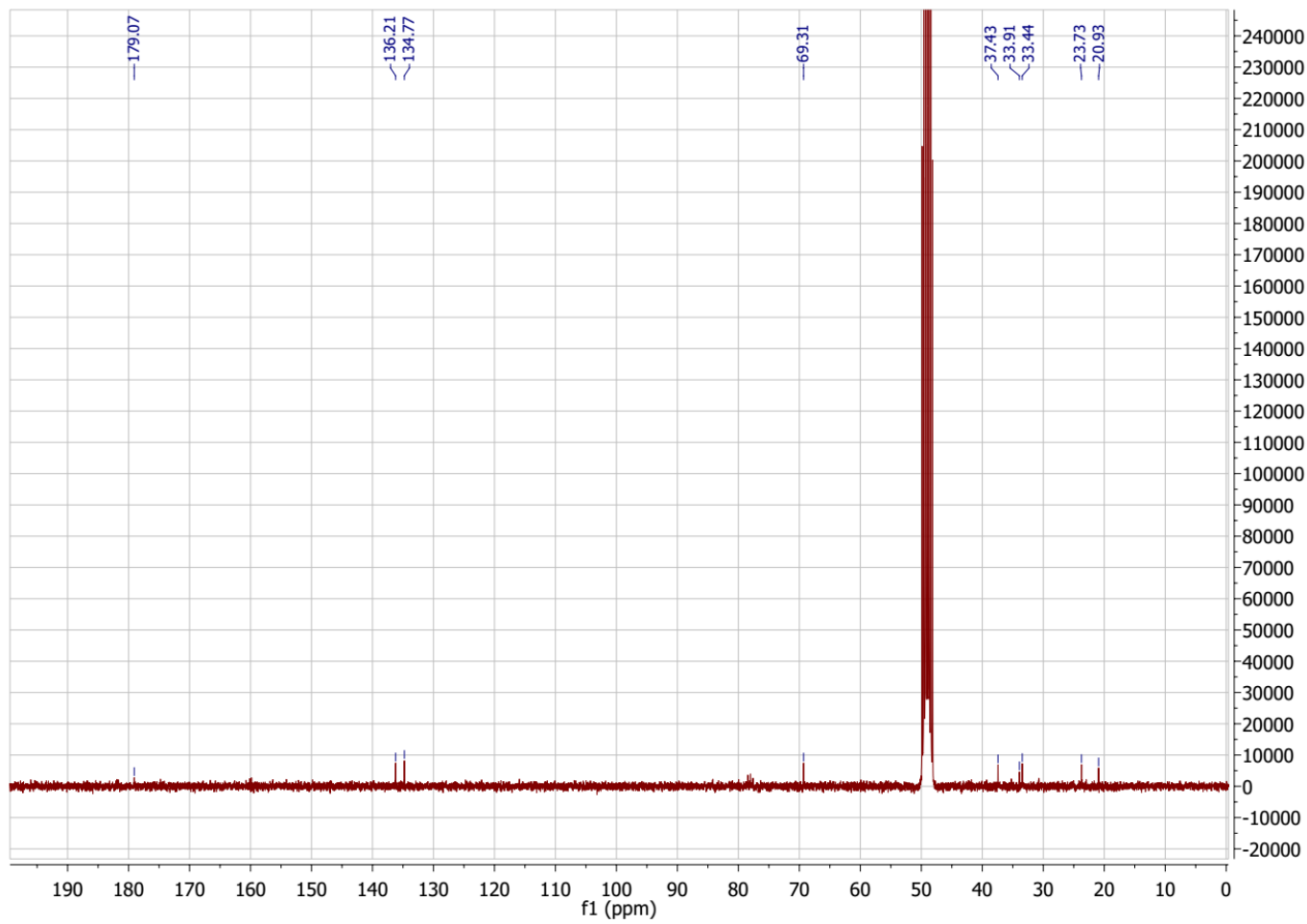




Figure S22. <sup>1</sup>H-<sup>1</sup>H COSY (500 MHz, CD<sub>3</sub>OD) of compound 4

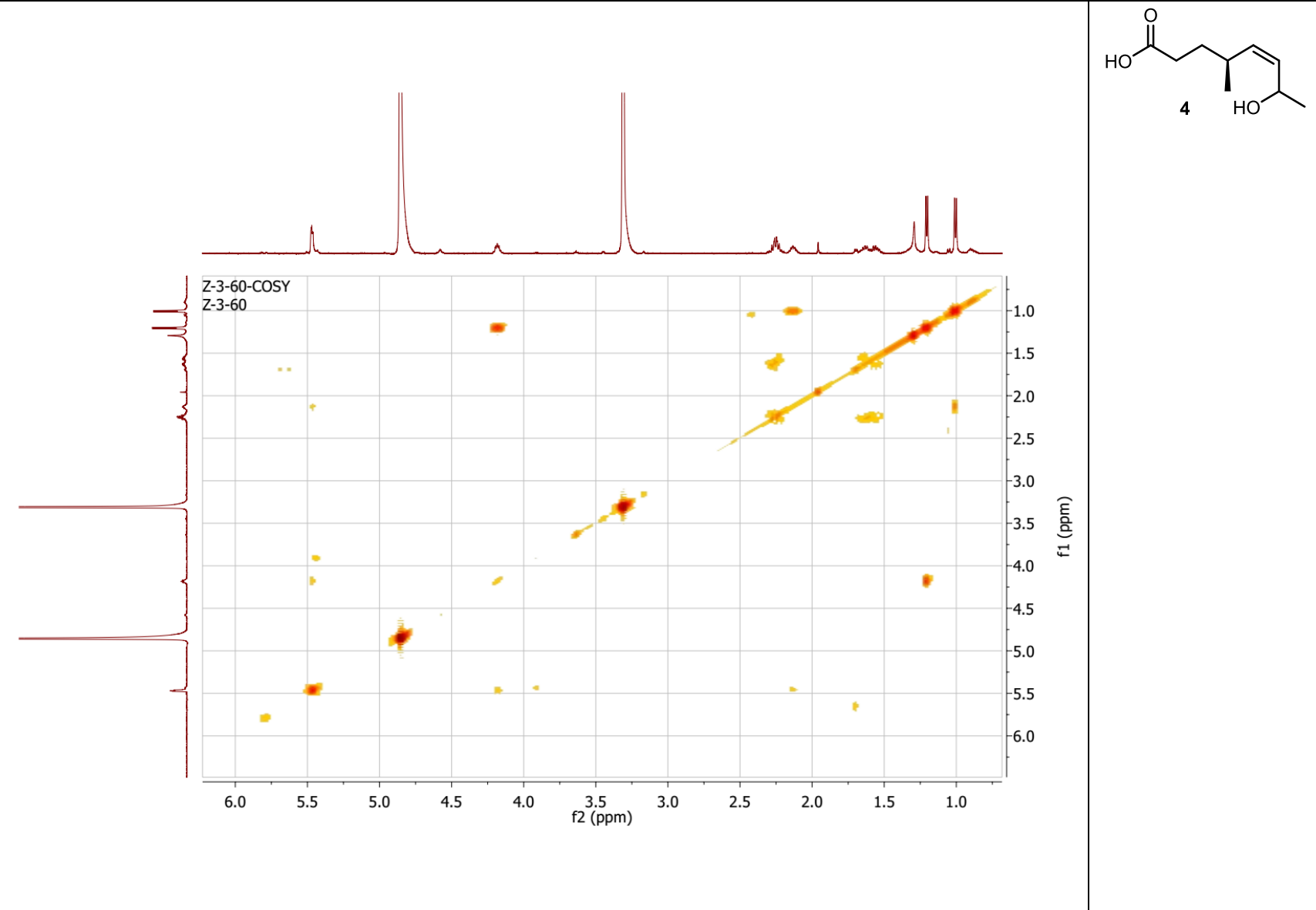


Figure S23. HSQC (500 MHz, CD<sub>3</sub>OD) of compound 4.

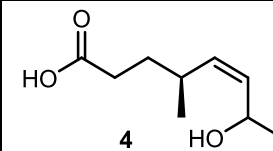
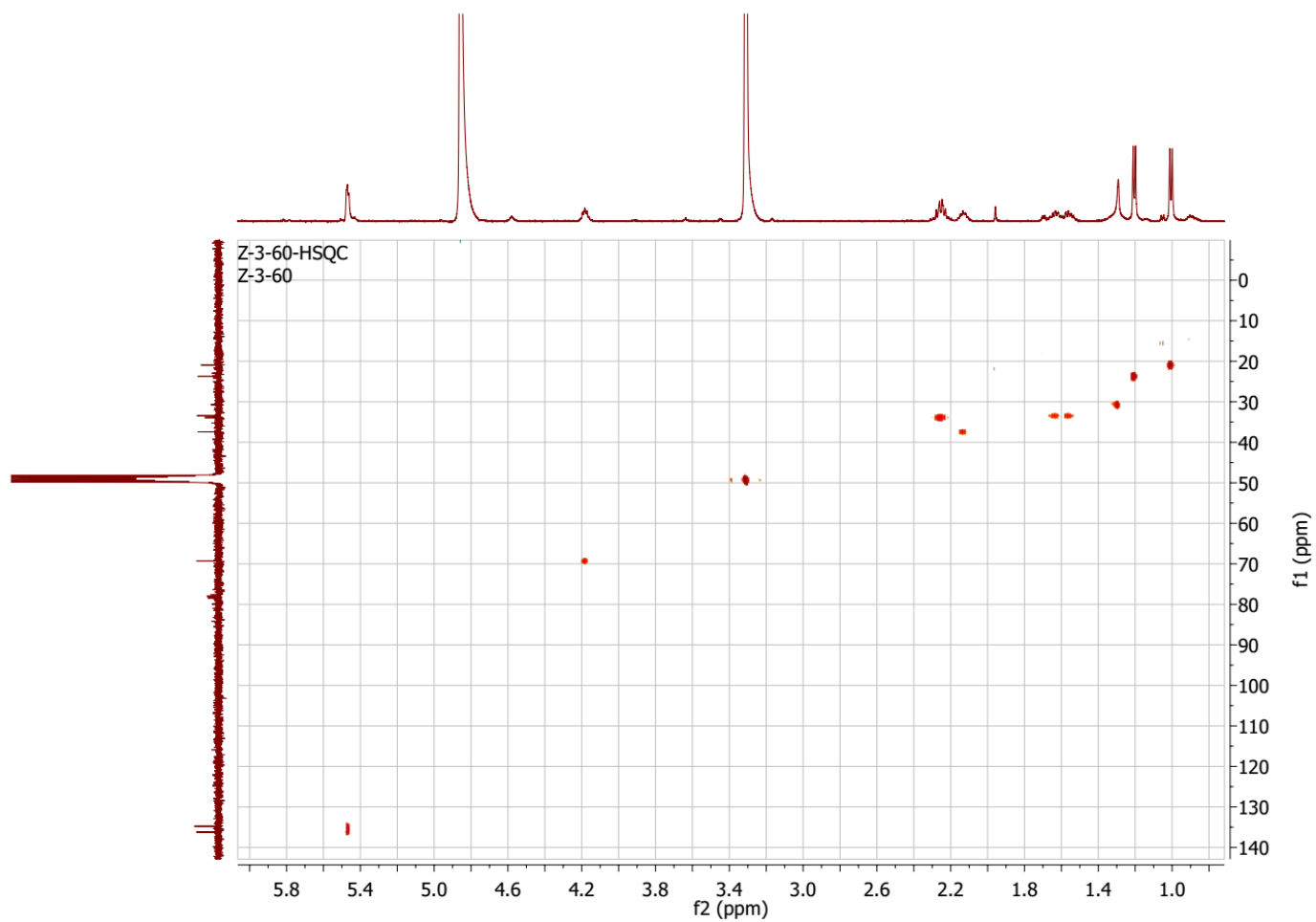


Figure S24. HMBC (500 MHz, CD<sub>3</sub>OD) of compound 4.

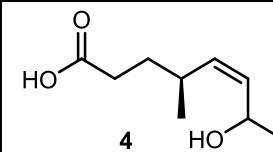
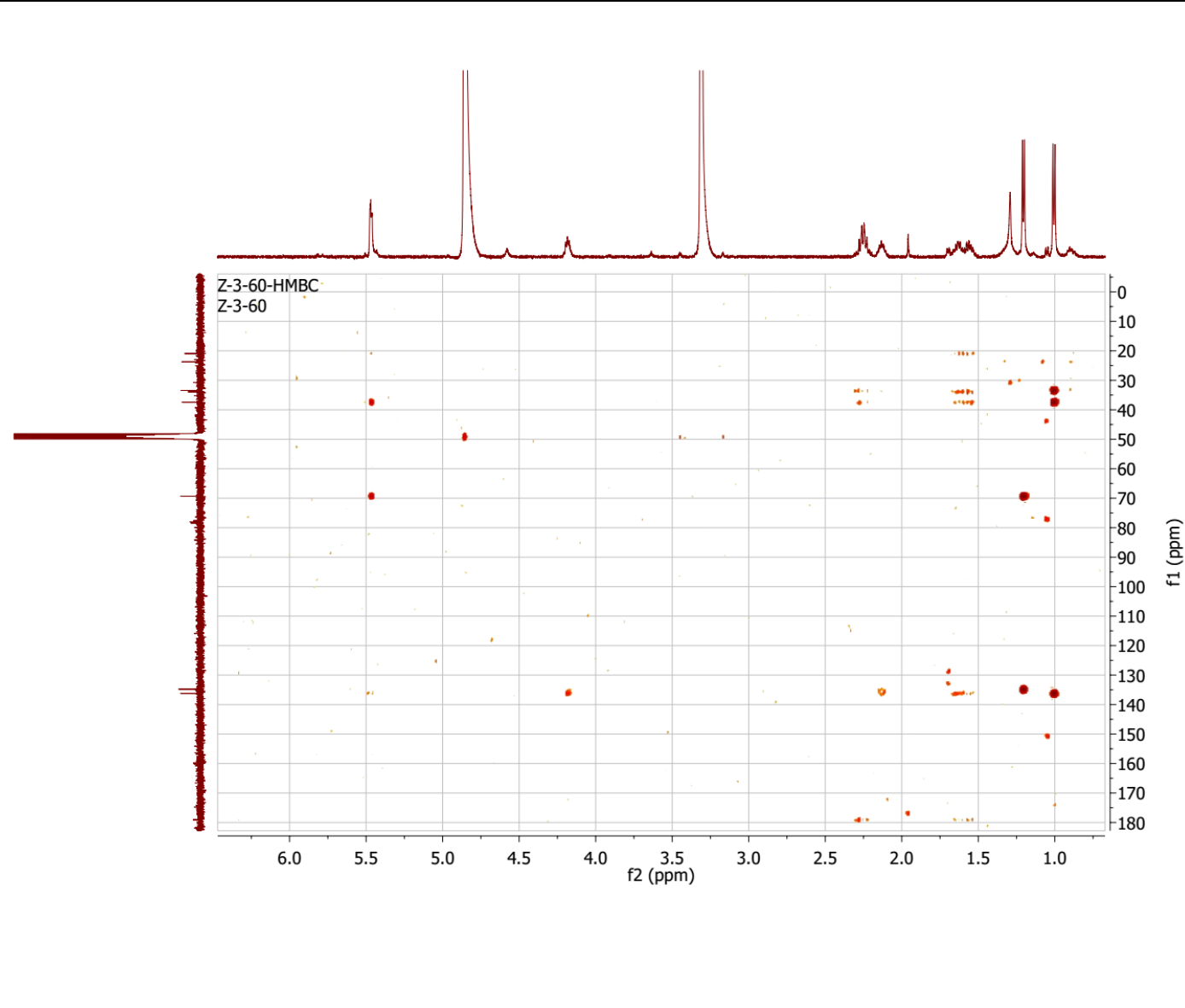


Figure S25. HRMS of compound 1.

z-4-24-5, MW=168, CH4

bri140312\_3 61 (1.017) Cm (58:61-1:33)

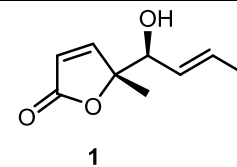
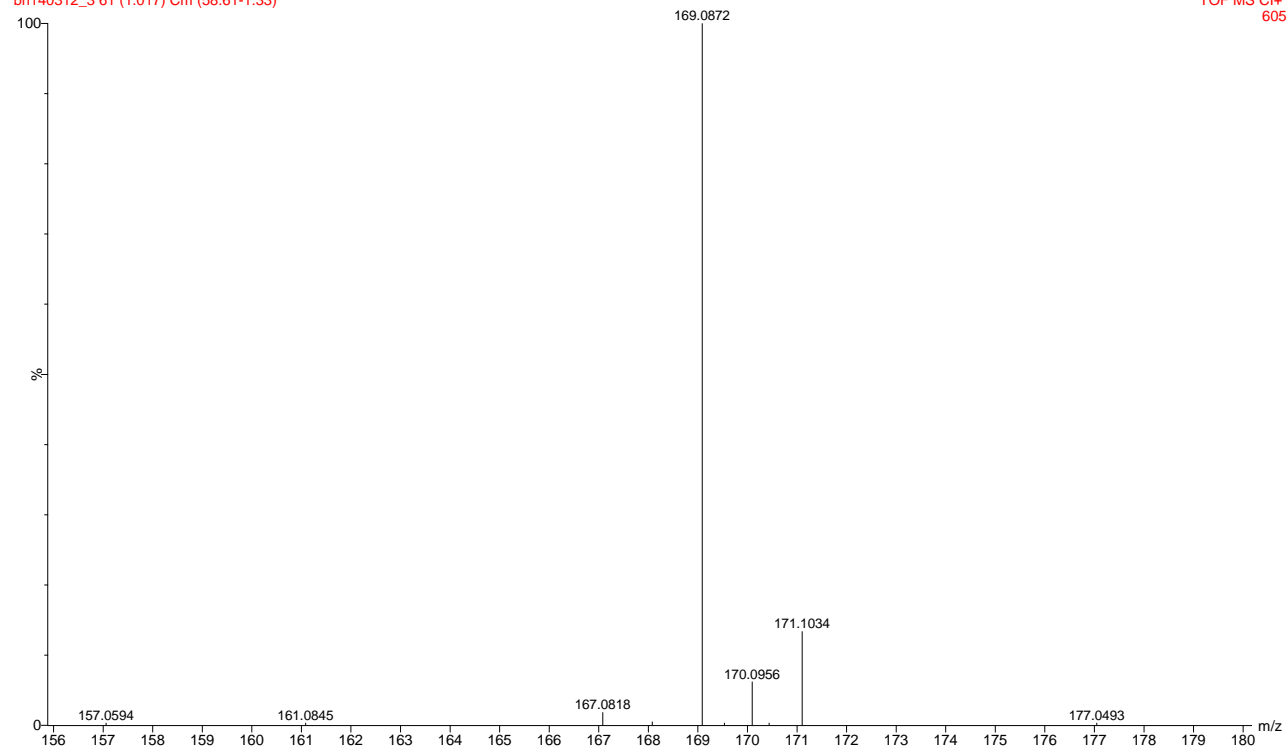
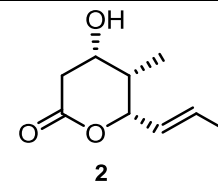


Figure S26. HRMS of compound 2.



z-4-51-1, MW=170, NH3

br1140312\_4\_2\_60 (1.000) Cm (56:60-1:47)

TOF MS Cl<sup>+</sup>  
5.10e3

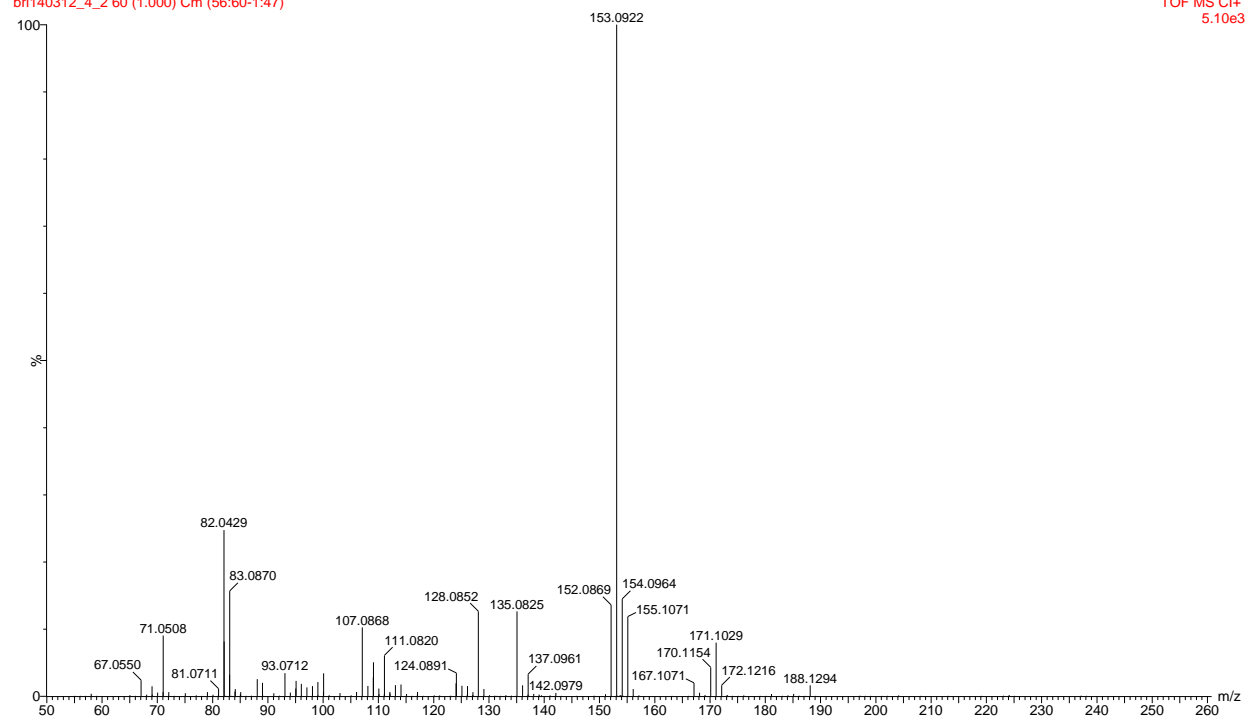
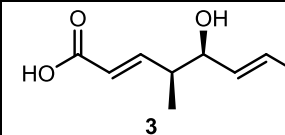


Figure S27. HRMS of compound 3.



z-4-35-1, MW=170/172, NH3  
br140312\_5\_2 99 (1.650) Cm (99:103-1:43)

TOF MS Cl+  
3.90e4

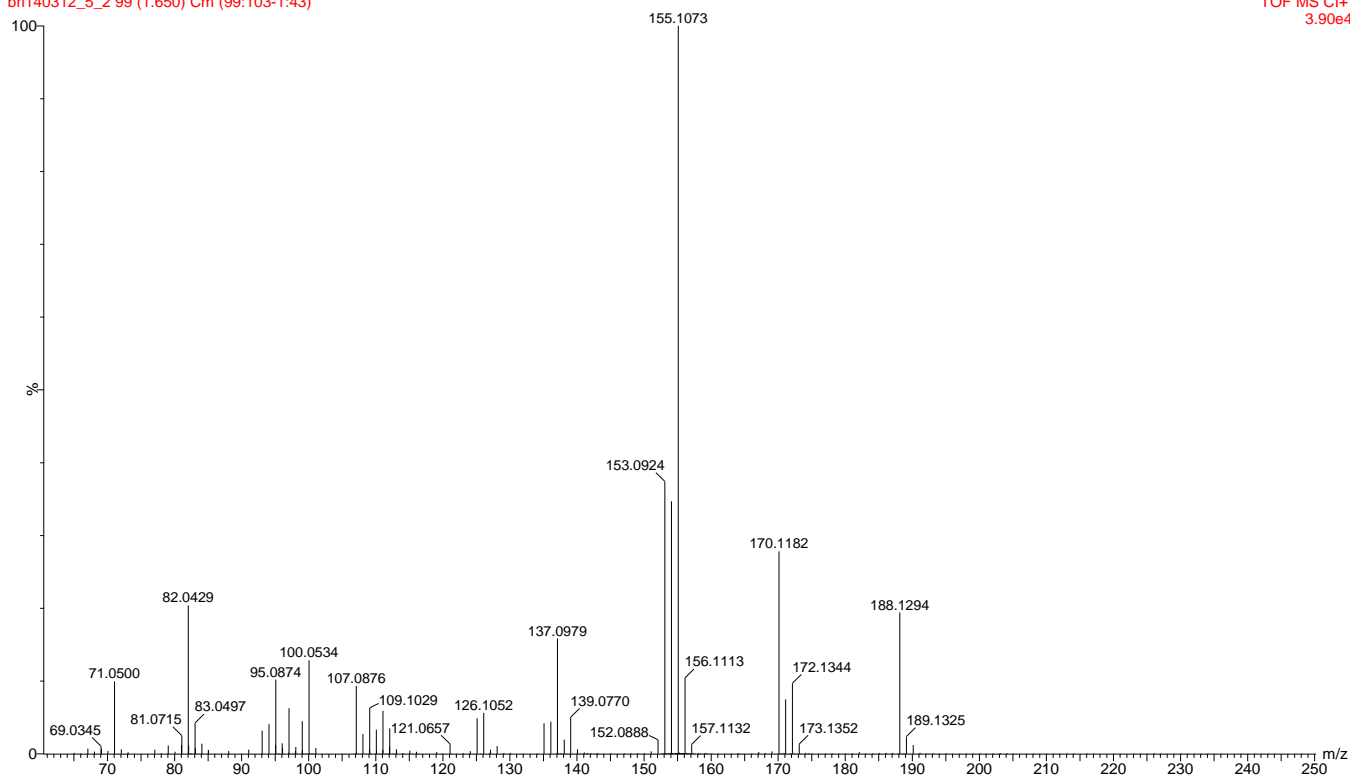


Figure S28. HRMS of compound 4.

z-4-35-1, MW=170/172, NH3  
bri140312\_5\_2 99 (1.650) Cm (99:103-1:43)

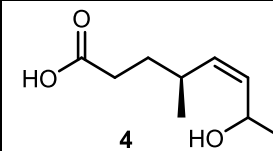
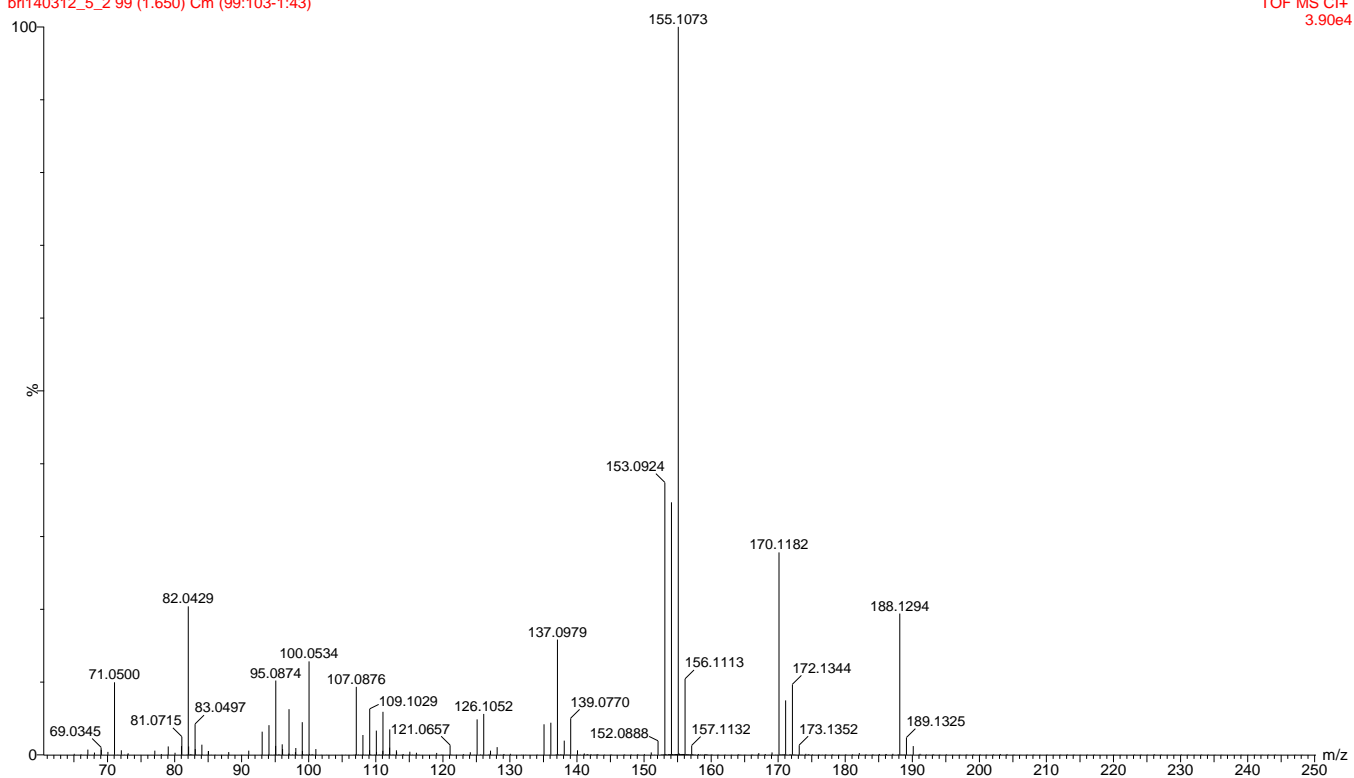


Figure S29. IR of compound 1.

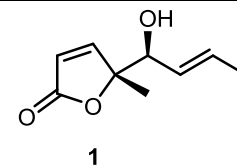
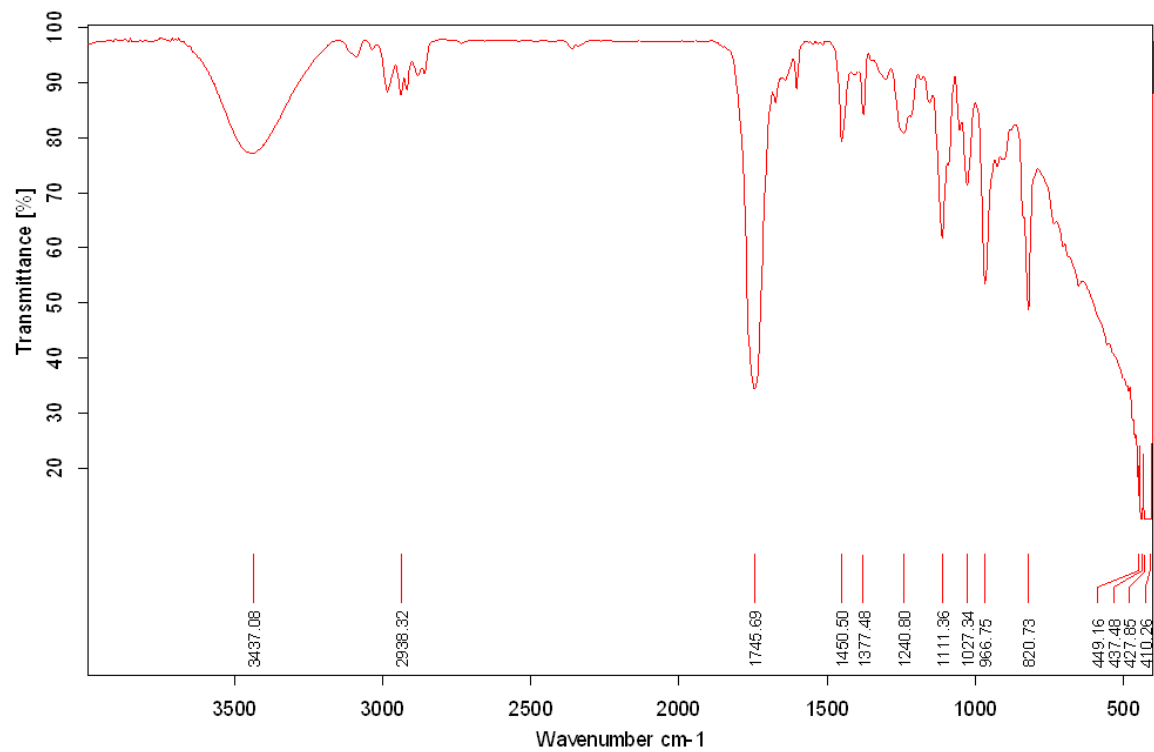




Figure S30. IR of compound 2.

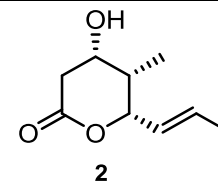
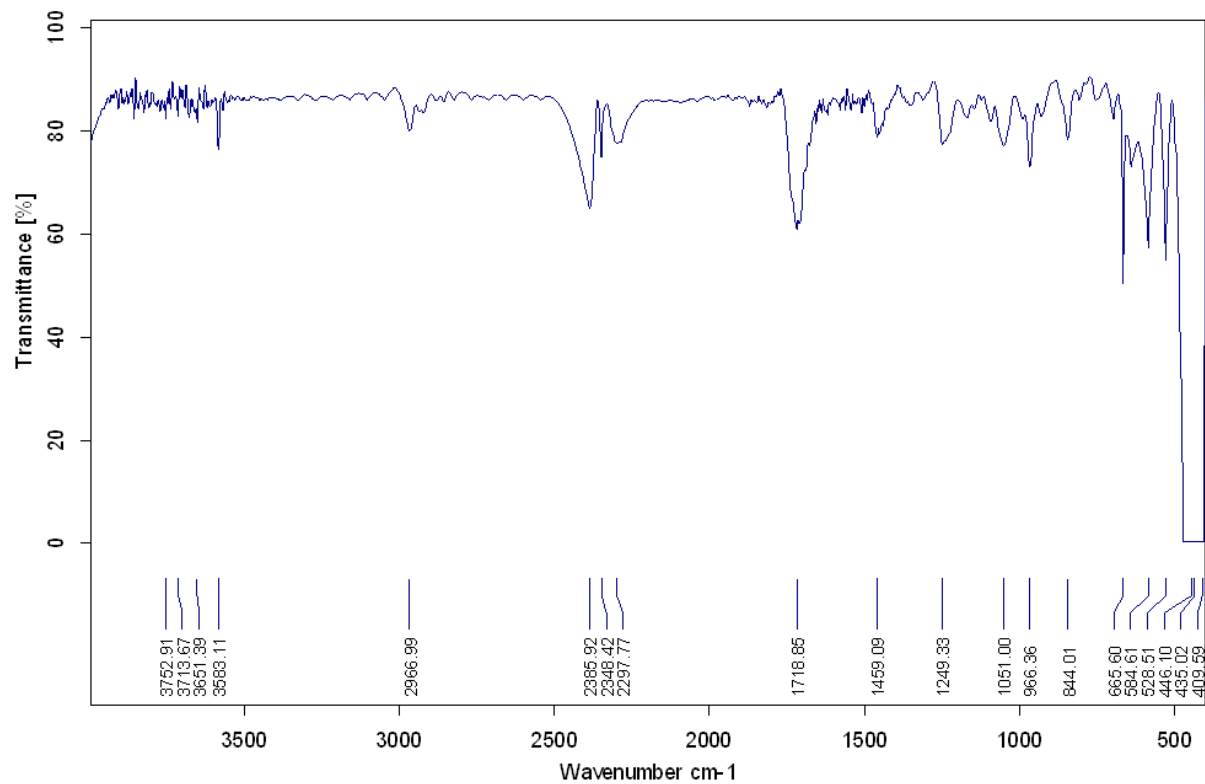


Figure S31. IR of compound 3.

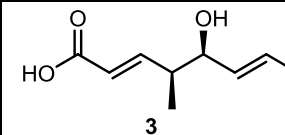
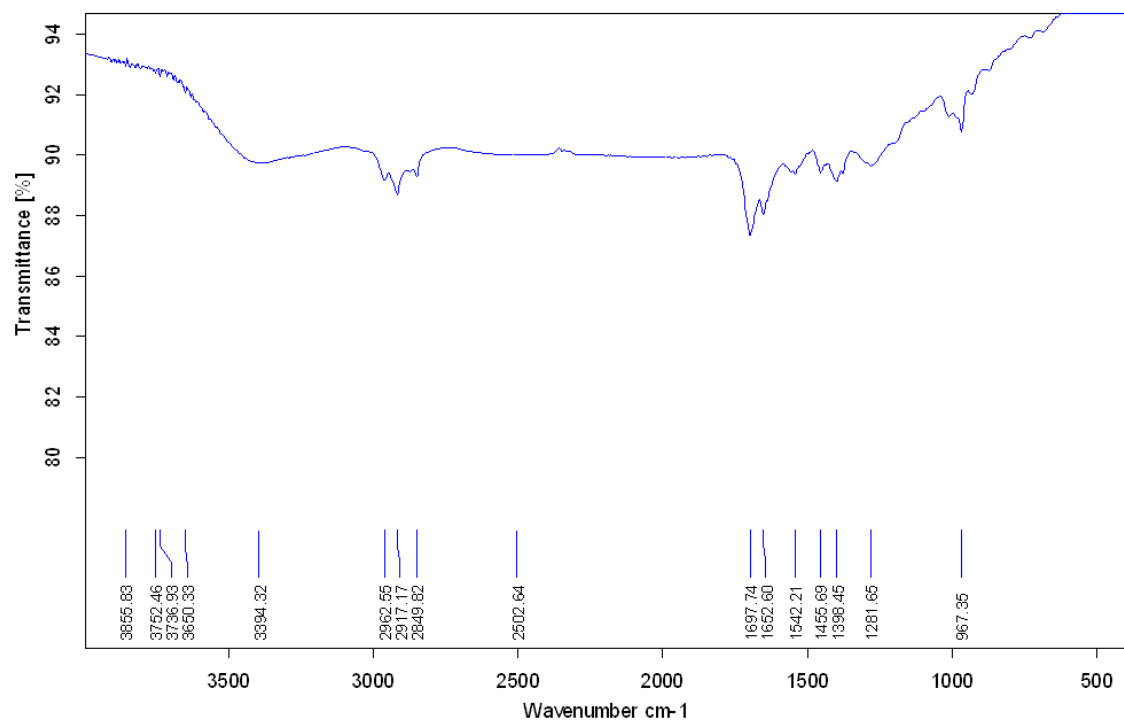


Figure S32. IR of compound 4.

