

## SUPPLEMENTARY MATERIAL

### Pregnane glycosides from *Gymnema inodorum* and their $\alpha$ -glucosidase inhibitory activity

Do Thi Trang<sup>a</sup>, Duong Thi Hai Yen<sup>a</sup>, Nguyen The Cuong<sup>c</sup>, Luu The Anh<sup>b</sup>, Nguyen Thi Hoai<sup>d</sup>, Bui Huu Tai<sup>a,e</sup>, Vu Van Doan<sup>a</sup>, Pham Hai Yen<sup>a</sup>, Tran Hong Quang<sup>a</sup>, Nguyen Xuan Nghiem<sup>a,e</sup>, Chau Van Minh<sup>a</sup>, Phan Van Kiem<sup>a,e\*</sup>

<sup>a</sup>Institute of Marine Biochemistry, Vietnam Academy of Science and Technology (VAST), 18 Hoang Quoc Viet, Cau Giay, Hanoi, Vietnam

<sup>b</sup>Central Institute for Natural Resources and Environmental Studies, Vietnam National National University, Hanoi, 19 Le Thanh Tong, Hoan Kiem, Hanoi, Vietnam

<sup>c</sup>Institute of Ecology and Biological Resources, VAST, 18 Hoang Quoc Viet, Cau Giay, Hanoi, Vietnam

<sup>d</sup>Faculty of Pharmacy, Hue University of Pharmacy and Medicine, Hue, Vietnam

<sup>e</sup>Graduate University of Science and Technology, VAST, 18 Hoang Quoc Viet, Cau Giay, Hanoi, Vietnam

**ABSTRACT:** Two new pregnane glycosides, gyminosides A and B (**1** and **2**) and three known, tinctoroside B (**3**), tinctoroside C (**4**), and gymnepregoside F (**5**) were isolated from the leaves of *Gymnema inodorum* (Lour.) Decne. Their structures were elucidated by physical and chemical methods and comparing with those reported in the literature. All these compounds were evaluated for  $\alpha$ -glucosidase assay. Compound **5** exhibited the most  $\alpha$ -glucosidase activity with inhibitory percentage of  $63.7 \pm 4.9\%$  at the concentration of 200  $\mu$ M. Compounds **1-4** showed significant  $\alpha$ -glucosidase activity with inhibitory percentage ranging from 40.0 to 52.1 % at the concentration of 200  $\mu$ M.

**Keywords:** *Gymnema inodorum*, Asclepiadaceae, pregnane,  $\alpha$ -glucosidase, gyminoside

## Content

<b>Table S1.</b>	NMR spectroscopic data for compounds <b>1</b> and <b>2</b> in CD <sub>3</sub> OD.....	1
<b>Figure S1.</b>	The key HMBC and COSY correlations of compounds <b>1</b> and <b>2</b> .....	3
<b>Figure S2.</b>	HR-ESI-MS of compound <b>1</b> .....	3
<b>Figure S3.</b>	<sup>1</sup> H-NMR spectrum of compound <b>1</b> .....	4
<b>Figure S4.</b>	<sup>13</sup> C-NMR spectrum of compound <b>1</b> .....	4
<b>Figure S5.</b>	HSQC spectrum of compound <b>1</b> .....	5
<b>Figure S6.</b>	HMBC spectrum of compound <b>1</b> .....	5
<b>Figure S7.</b>	COSY spectrum of compound <b>1</b> .....	6
<b>Figure S8.</b>	NOESY spectrum of compound <b>1</b> .....	6
<b>Figure S9.</b>	HR-ESI-MS of compound <b>2</b> .....	7
<b>Figure S10.</b>	<sup>1</sup> H-NMR spectrum of compound <b>2</b> .....	7
<b>Figure S11.</b>	<sup>13</sup> C-NMR spectrum of compound <b>2</b> .....	8
<b>Figure S12.</b>	HSQC spectrum of compound <b>2</b> .....	8
<b>Figure S13.</b>	HMBC spectrum of compound <b>2</b> .....	9
<b>Figure S14.</b>	COSY spectrum of compound <b>2</b> .....	9
<b>Figure S15.</b>	NOESY spectrum of compound <b>2</b> .....	10
<b>Figure S16.</b>	<sup>1</sup> H-NMR spectrum of compound <b>3</b> .....	10
<b>Figure S17.</b>	<sup>13</sup> C-NMR spectrum of compound <b>3</b> .....	11
<b>Figure S18.</b>	<sup>1</sup> H-NMR spectrum of compound <b>4</b> .....	11
<b>Figure S19.</b>	<sup>13</sup> C-NMR spectrum of compound <b>4</b> .....	12
<b>Figure S20.</b>	<sup>1</sup> H-NMR spectrum of compound <b>5</b> .....	12
<b>Figure S21.</b>	<sup>13</sup> C-NMR spectrum of compound <b>5</b> .....	13
<b>Figure S22.</b>	$\alpha$ -Glucosidase inhibitory activity of compounds <b>1–5</b> .....	13

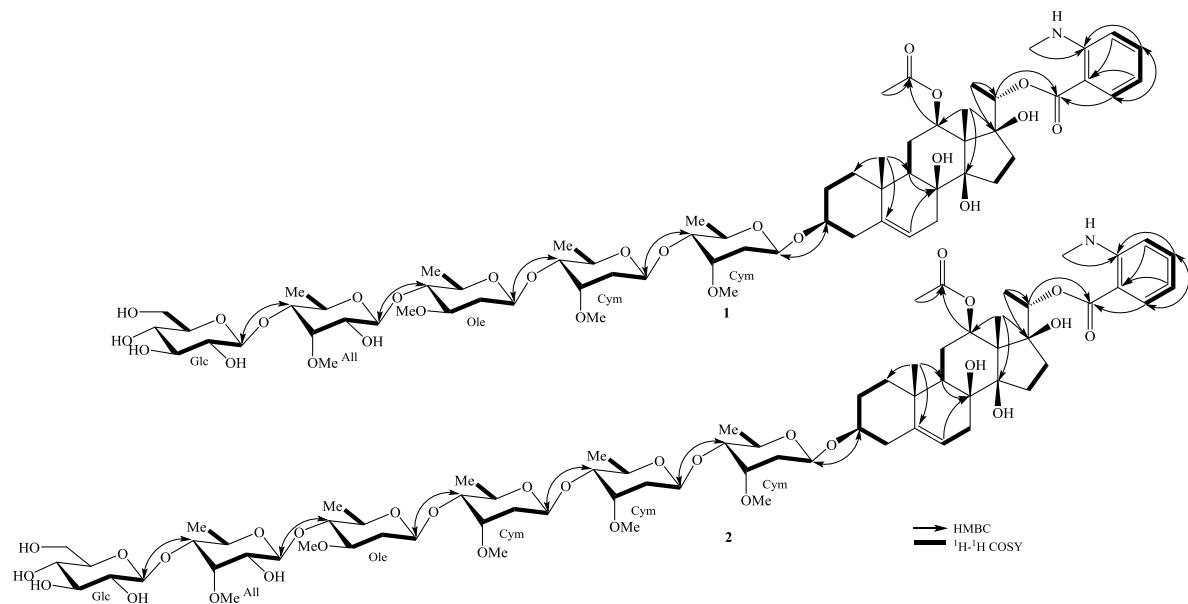
**Table S1.** NMR spectroscopic data for compounds **1** and **2** in CD<sub>3</sub>OD.

C	<b>1</b>		<b>2</b>	
	$\delta_{\text{C}}$	$\delta_{\text{H}}$ (mult., <i>J</i> , in Hz)	$\delta_{\text{C}}$	$\delta_{\text{H}}$ (mult., <i>J</i> , in Hz)
1	39.8	1.11 (m)/1.80 (m)	39.7	1.11 (m)/1.80 (m)
2	30.1	1.60 (m)/1.87 (m)	30.1	1.60 (m)/1.87 (m)
3	79.3	3.53 (m)	79.2	3.54 (m)
4	39.8	2.23 (m)/2.37 (m)	39.8	2.23 (m)/2.38 (m)
5	140.3	-	140.1	-
6	119.6	5.35 (br s)	119.8	5.34 (br s)
7	35.2	2.18 (m)/2.22 (m)	35.2	2.17 (m)/2.22 (m)
8	75.1	-	75.0	-
9	44.8	1.51 (m)	44.7	1.51 (m)
10	38.0	-	38.0	-
11	26.0	1.63 (m)/1.94 (m)	26.0	1.61 (m)/1.94 (m)
12	75.4	4.74 (m)	75.3	4.72 (m)
13	57.6	-	57.5	-
14	89.5	-	89.5	-
15	34.0	1.93 (m)/1.97 (m)	34.0	1.93 (m)/1.99 (m)
16	34.3	1.96 (m)	34.3	1.96 (m)
17	88.6	-	88.5	-
18	10.9	1.53 (s)	11.0	1.53 (s)
19	18.5	1.13 (s)	18.5	1.13 (s)
20	75.2	4.76 (m)	75.1	4.74 (m)
21	15.4	1.32 (d, 6.0)	15.4	1.32 (d, 6.0)
<b>12-Ac</b>				
1	173.1	-	173.1	-
2	21.9	1.85 (s)	21.9	1.85 (s)
<b>20-Ant</b>				
1	111.6	-	111.4	-
2	153.5	-	153.5	-
3	111.9	6.74 (d, 8.0)	111.9	6.74 (d, 8.0)
4	135.8	7.41 (ddd, 1.5, 8.0, 8.0)	135.8	7.41 (ddd, 1.5, 8.0, 8.0)
5	115.5	6.62 (ddd, 1.5, 8.0, 8.0)	115.5	6.62 (ddd, 1.5, 8.0, 8.0)
6	133.2	8.08 (dd, 1.5, 8.0)	133.2	8.07 (dd, 1.5, 8.0)
7	169.1	-	169.1	-
N-Me	29.6	2.91 (s)	29.6	2.91 (s)
<b>Cym I</b>				
1	97.3	4.87 (dd, 1.5, 9.5)	97.2	4.87 (dd, 1.5, 9.5)
2	36.8	1.59 (m)/2.06 (m)	36.6	1.60 (m)/2.07 (m)
3	78.5	3.86 (m)	78.5	3.86 (m)
4	83.9	3.24 (m)	83.9	3.24 (m)
5	69.9	3.81 (m)	69.8	3.82 (m)
6	18.5	1.20 (d, 6.5)	18.4	1.20 (d, 6.5)
3-OMe	58.5	3.45 (s)	58.5	3.45 (s)
<b>Cym II</b>				
1	101.1	4.82 (br d, 9.0)	101.3	4.82 (br d, 9.0)
2	36.6	1.62 (m)/2.13 (m)	36.4	1.62 (m)/2.15 (m)
3	78.6	3.86 (m)	78.6	3.86 (m)

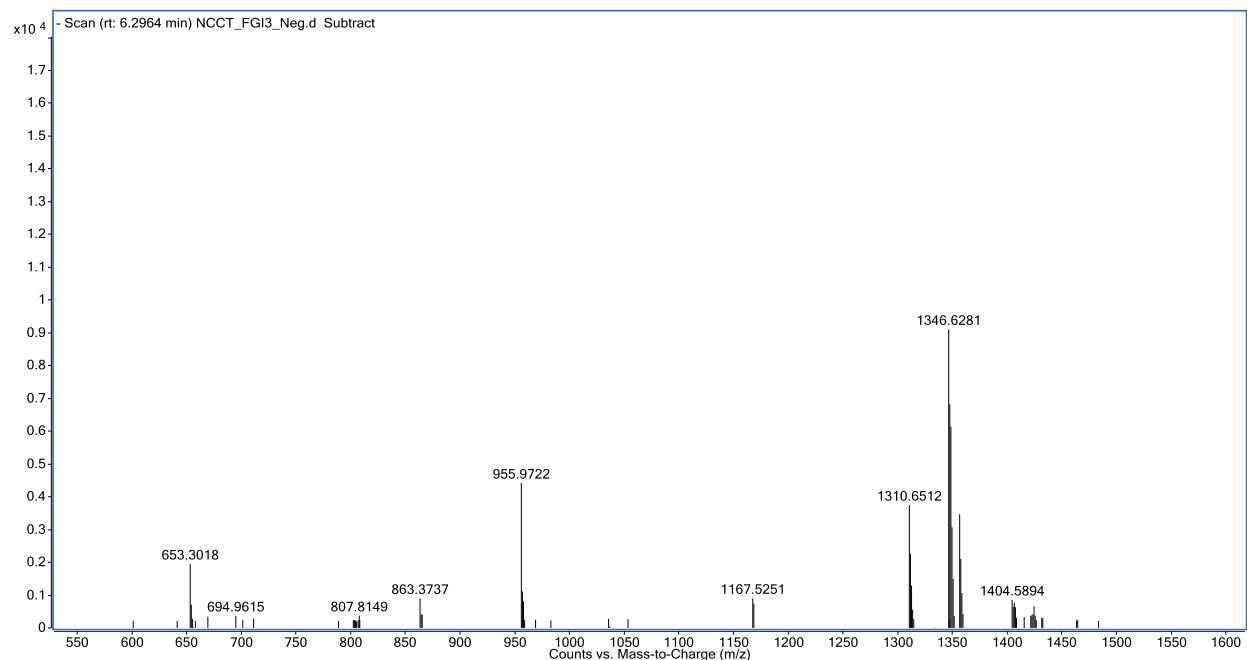
4	83.9	3.24 (m)		83.9	3.24 (m)
5	70.0	3.84 (m)		70.0	3.84 (m)
6	18.6	1.23 (d, 6.0)		18.6	1.23 (d, 6.0)
3-OMe	58.6	3.45 (s)		58.6	3.45 (s)
<b>Cym III</b>					
1			101.2	4.80 (dd, 1.5, 9.5)	
2			36.6	1.63 (m)/2.18 (m)	
3			78.7	3.95 (m)	
4			84.0	3.25 (m)	
5			70.0	3.76 (m)	
6			18.1	1.22 (d, 6.0)	
3-OMe			58.6	3.48 (s)	
<b>Ole</b>					
1	102.6	4.60 (dd, 1.5, 9.5)	102.6	4.60 (dd, 1.5, 9.5)	
2	37.6	1.45 (m)/2.34 (m)	37.5	1.44 (m)/2.34 (m)	
3	80.4	3.40 (m)	80.4	3.40 (m)	
4	83.7	3.28 (m)	83.8	3.29 (m)	
5	72.7	3.39 (m)	72.6	3.39 (m)	
6	19.0	1.38 (d, 6.0)	18.9	1.38 (d, 6.0)	
3-OMe	57.4	3.44 (s)	57.5	3.43 (s)	
<b>All</b>					
1	102.0	4.75 (d, 8.0)	102.1	4.72 (d, 8.0)	
2	73.1	3.35 (m)	73.0	3.31 (m)	
3	83.2	3.98 (t, 3.0)	83.1	3.93 (t, 3.0)	
4	83.8	3.35 (m)	83.8	3.36 (m)	
5	70.2	3.86 (m)	70.1	3.86 (m)	
6	18.1	1.33 (d, 6.5)	18.7	1.34 (d, 6.5)	
3-OMe	61.9	3.62 (s)	61.9	3.61 (s)	
<b>Glc</b>					
1	106.4	4.38 (d, 7.5)	106.2	4.37 (d, 7.5)	
2	75.6	3.23 (m)	75.4	3.23 (m)	
3	78.1	3.31 (m)	78.0	3.29 (m)	
4	72.0	3.30 (m)	71.8	3.28 (m)	
5	78.0	3.38 (m)	78.0	3.36 (m)	
6	63.2	3.70 (dd, 6.0, 12.0) 3.93 (dd, 2.0, 12.0)	63.0	3.67 (dd, 6.0, 12.0) 3.92 (dd, 2.0, 12.0)	

Assignments were done by HSQC, HMBC, and COSY experiments; Ac, acetyl; Cym,  $\beta$ -D-cymaropyranosyl; Ole,  $\beta$ -D-oleandropyranosyl; All, 6-deoxy-3-*O*-methyl- $\beta$ -D-allopyranosyl; glc,  $\beta$ -D-glucopyranosyl; Ant, *N*-methylantraniloyl

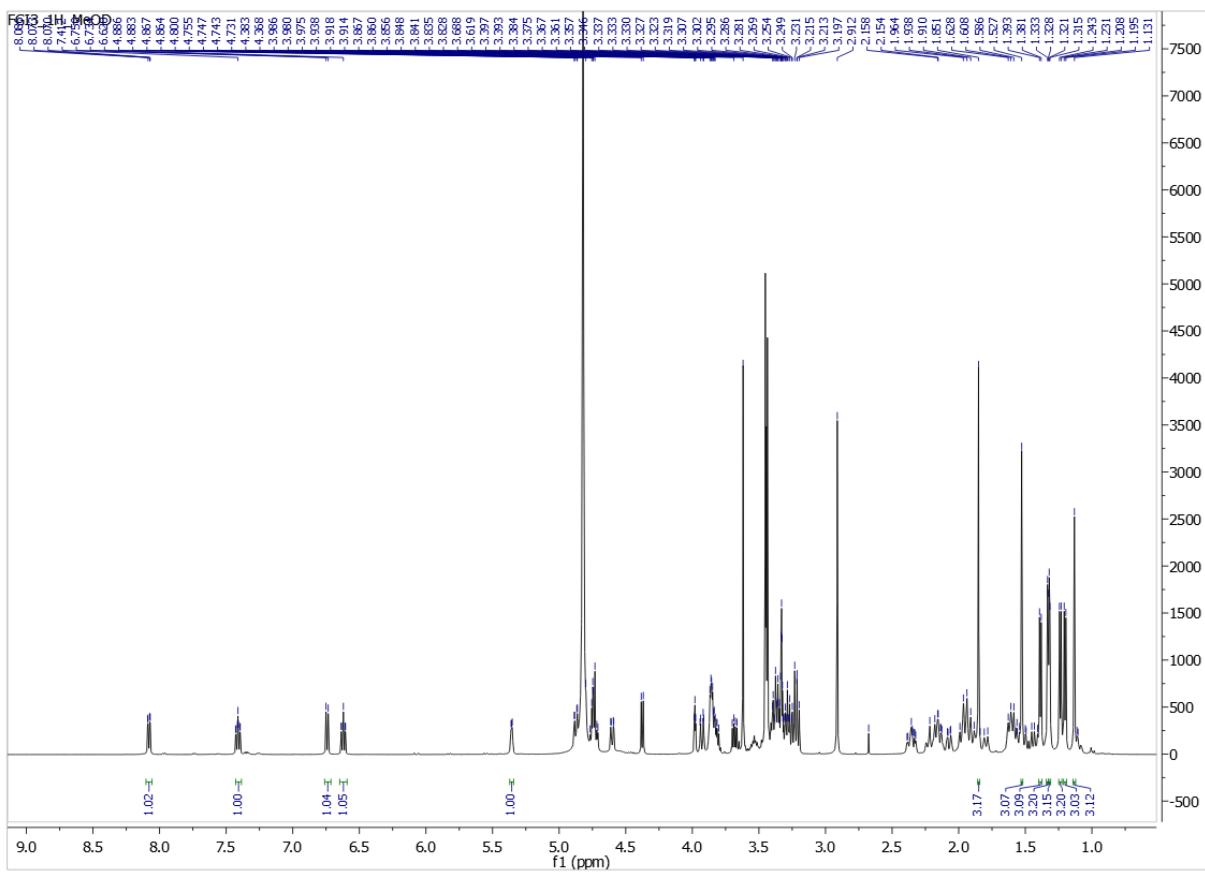
**Figure S1.** The key HMBC and COSY correlations of compounds **1** and **2**



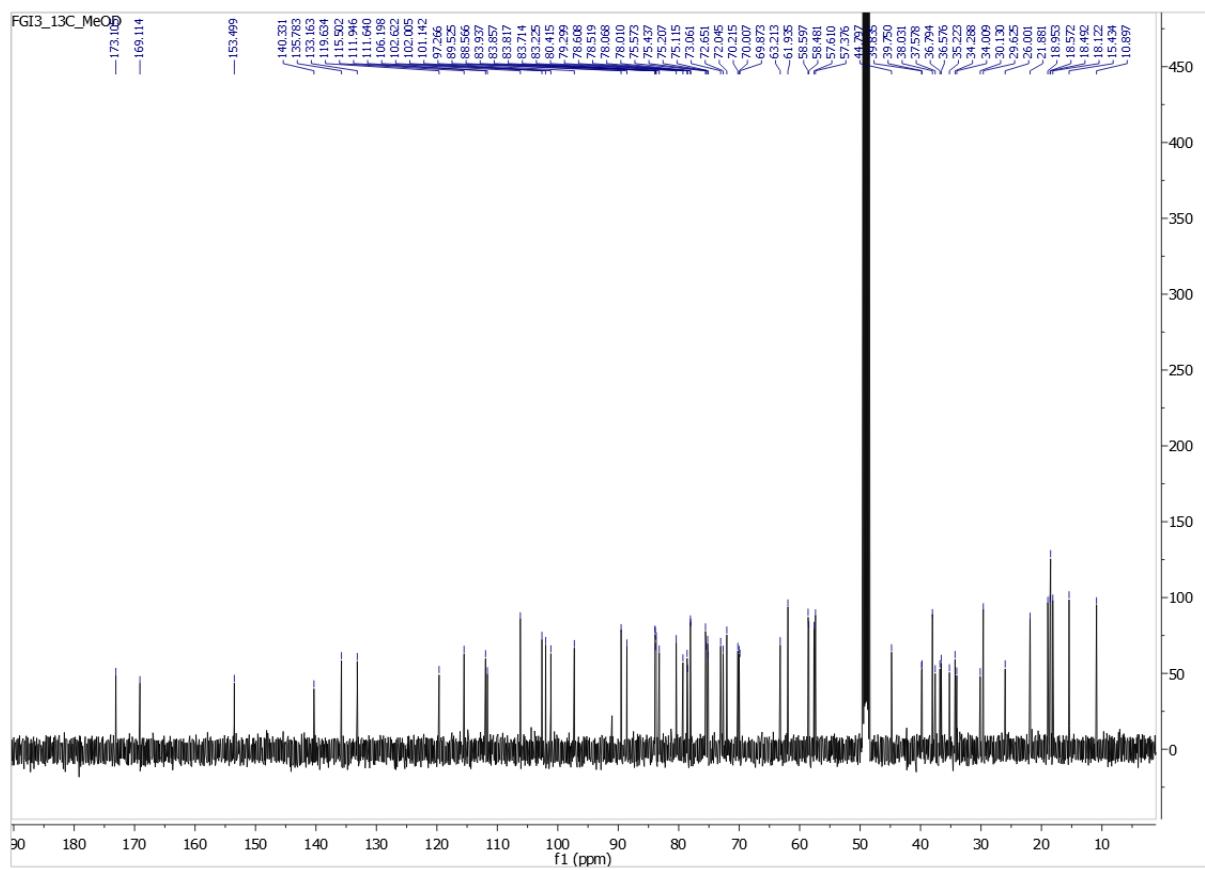
**Figure S2.** HR-ESI-MS of compound **1**



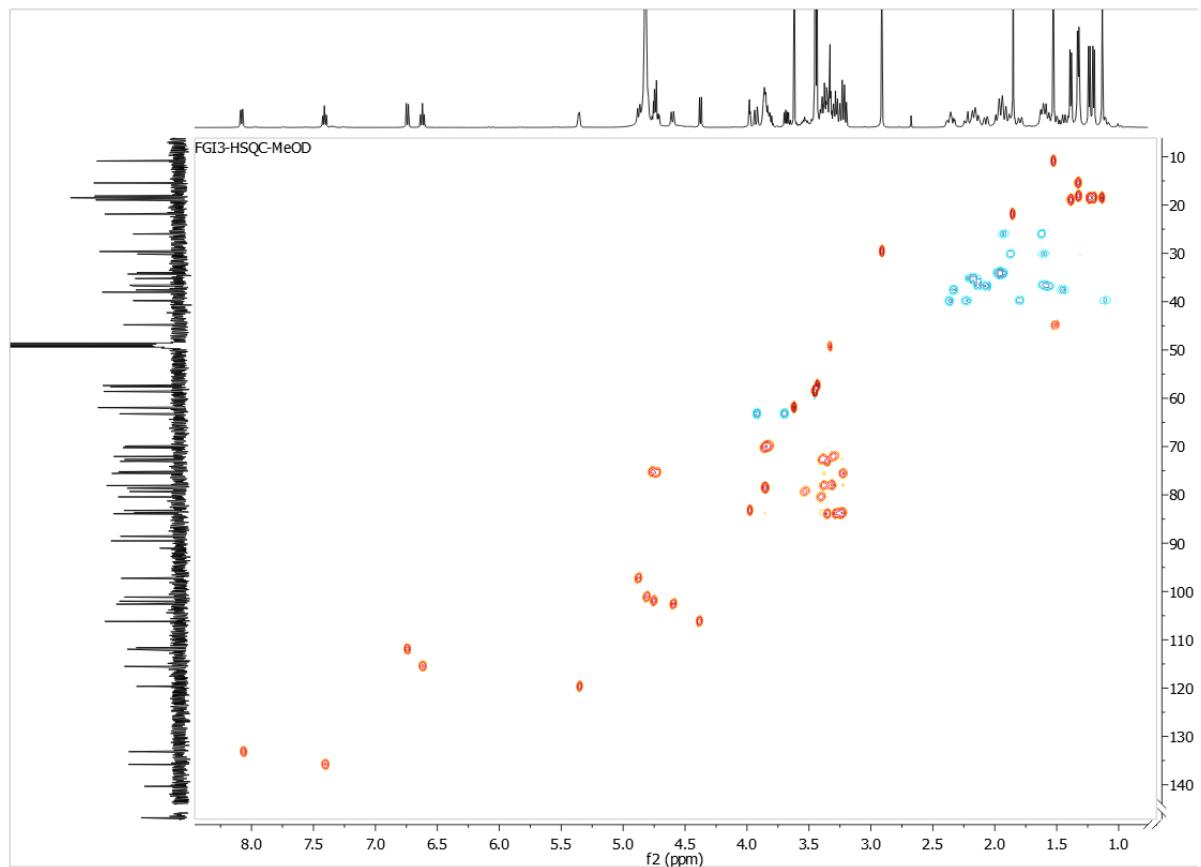
**Figure S3.**  $^1\text{H}$ -NMR spectrum of compound **1**



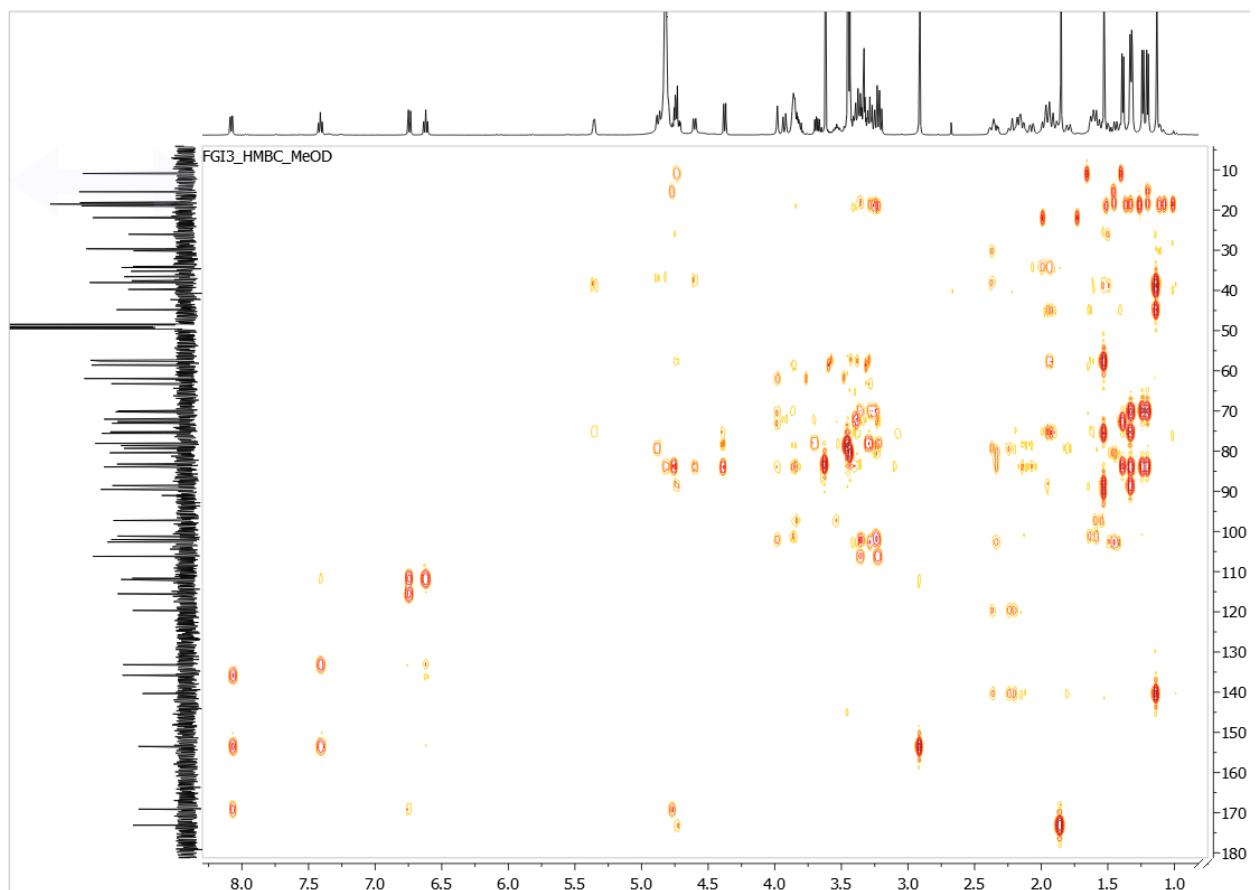
**Figure S4.**  $^{13}\text{C}$ -NMR spectrum of compound **1**



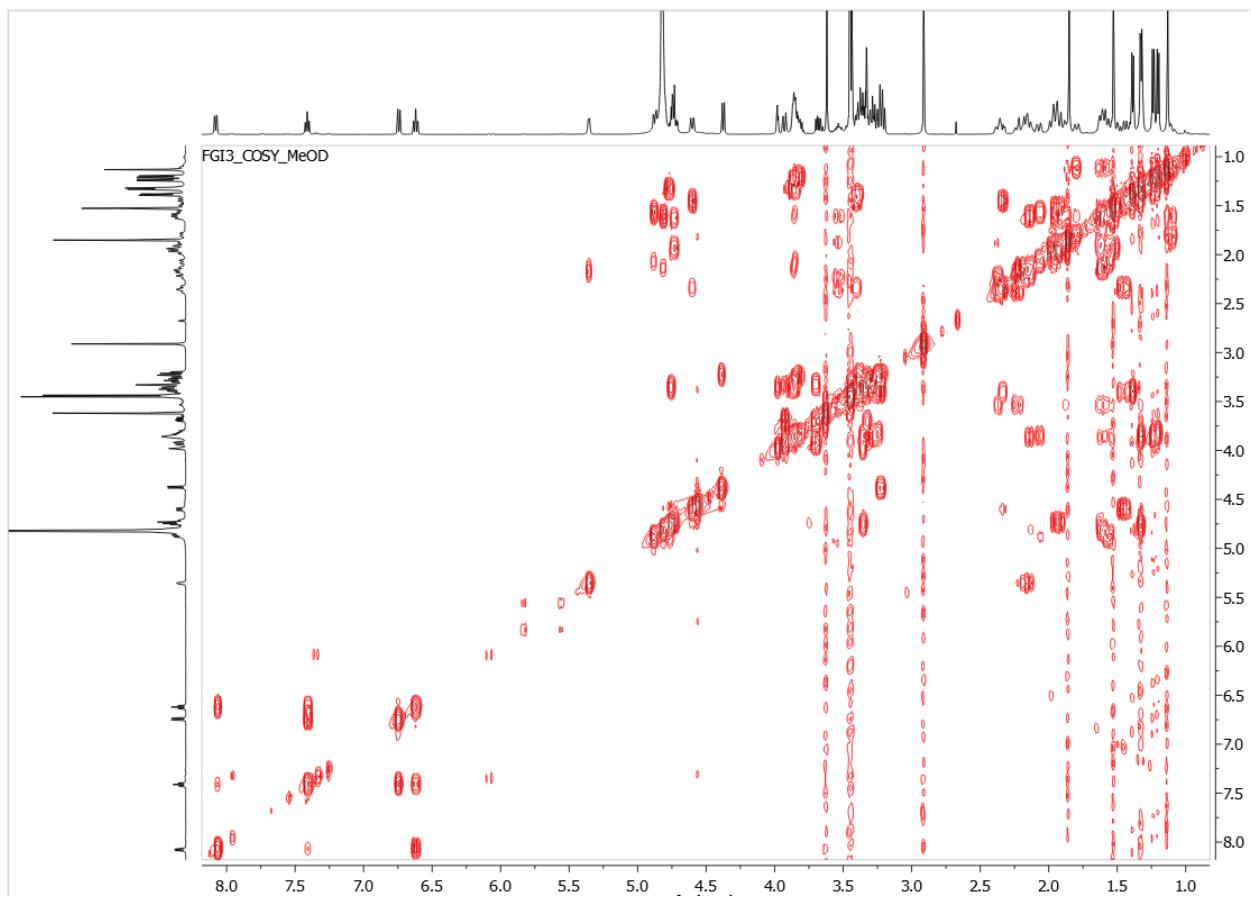
**Figure S5.** HSQC spectrum of compound **1**



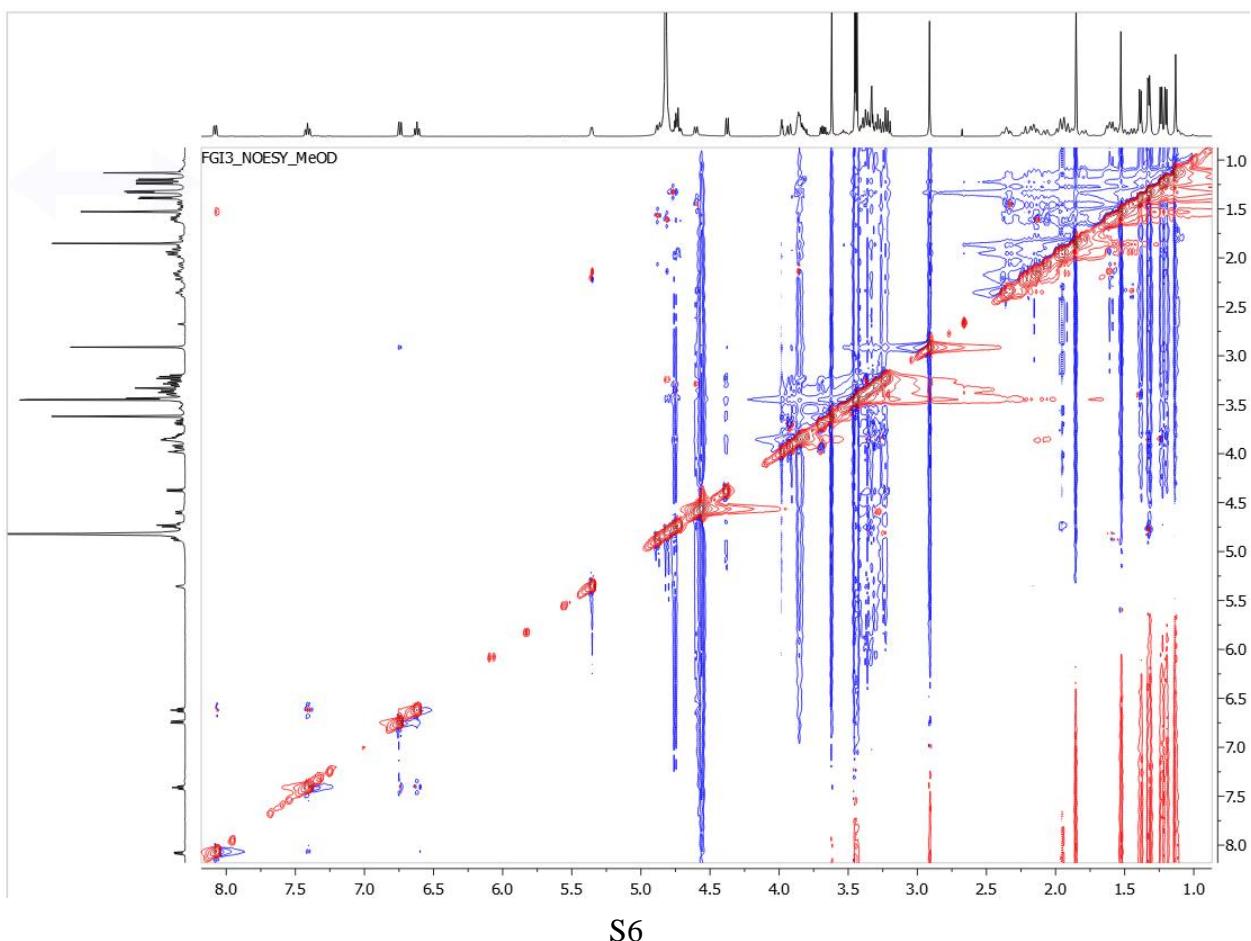
**Figure S6.** HMBC spectrum of compound **1**



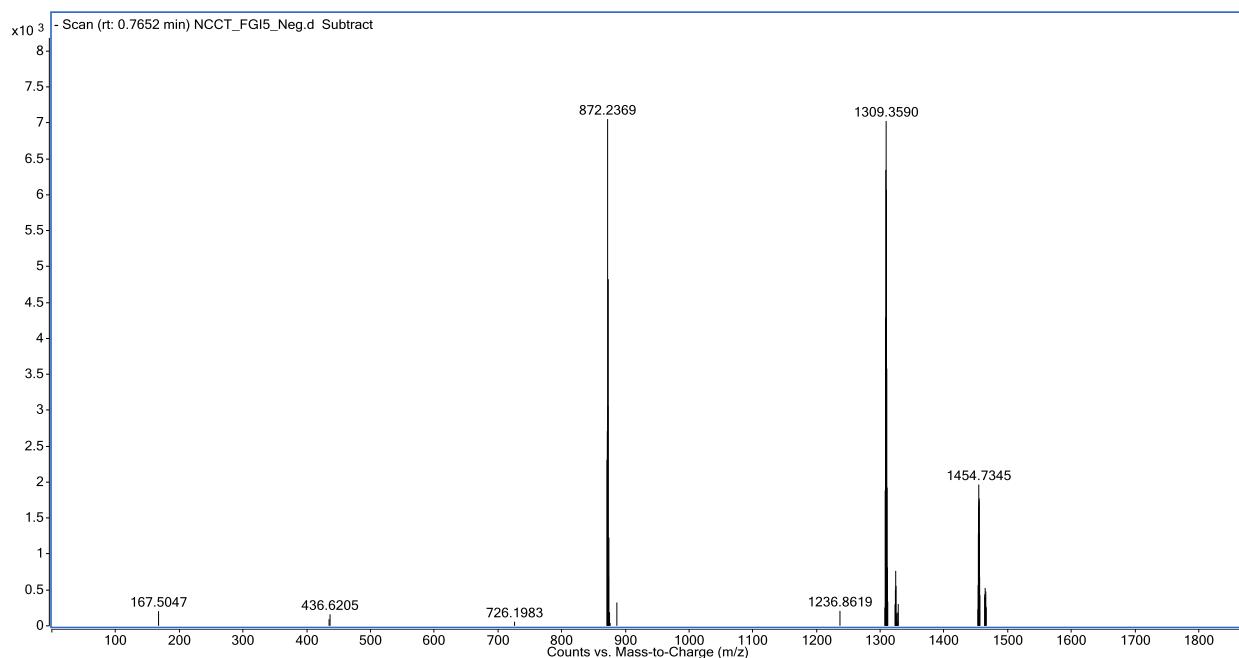
**Figure S7.** COSY spectrum of compound **1**



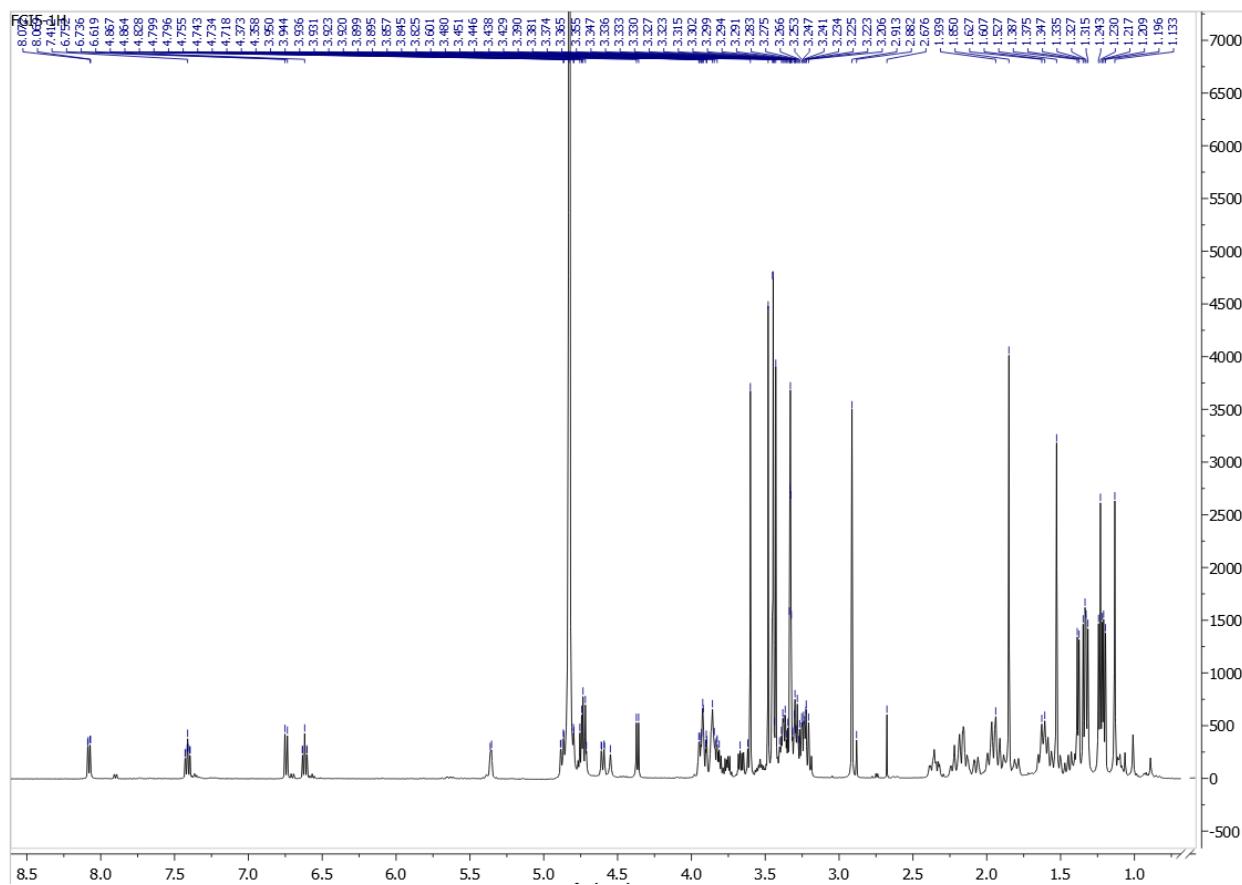
**Figure S8.** NOESY spectrum of compound **1**



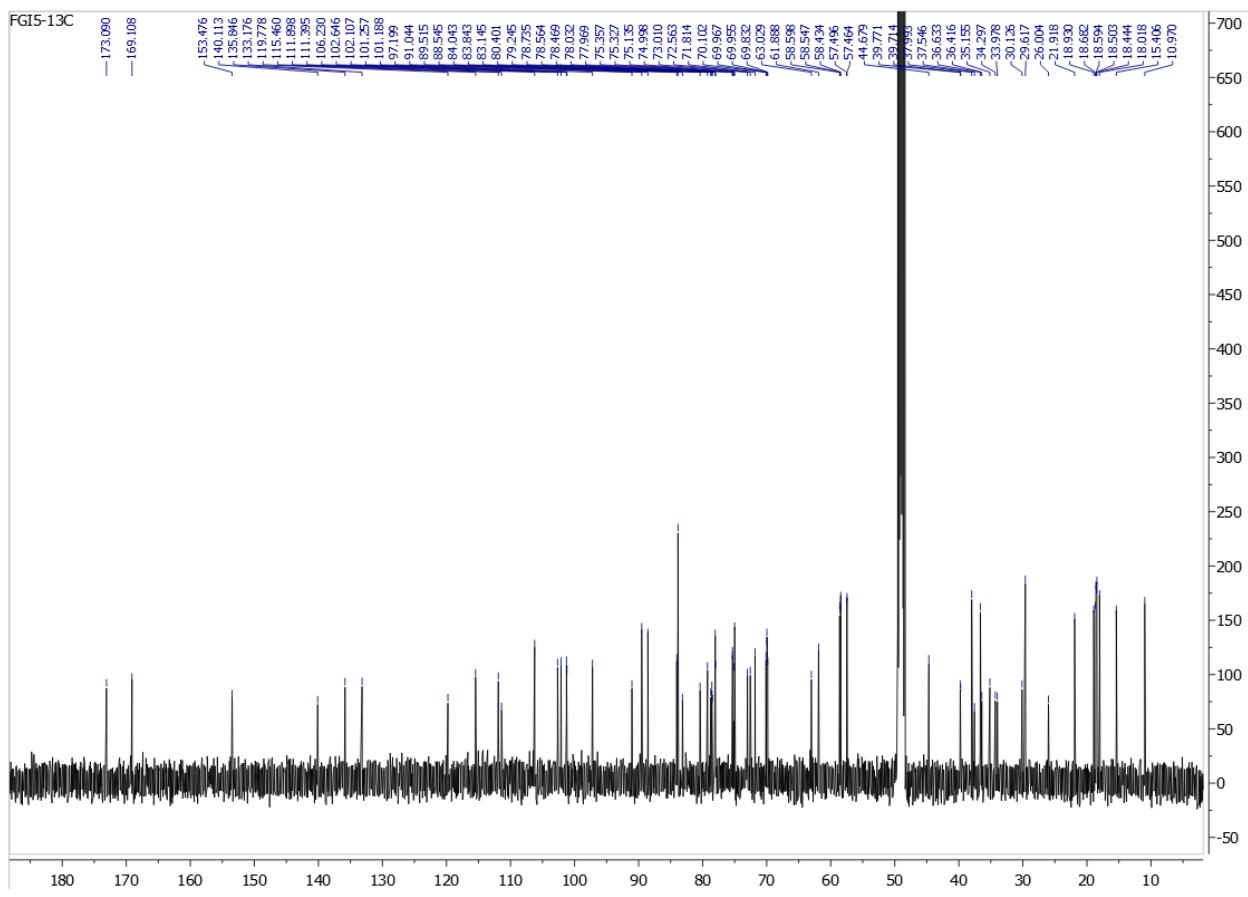
**Figure S9.** HR-ESI-MS of compound 2



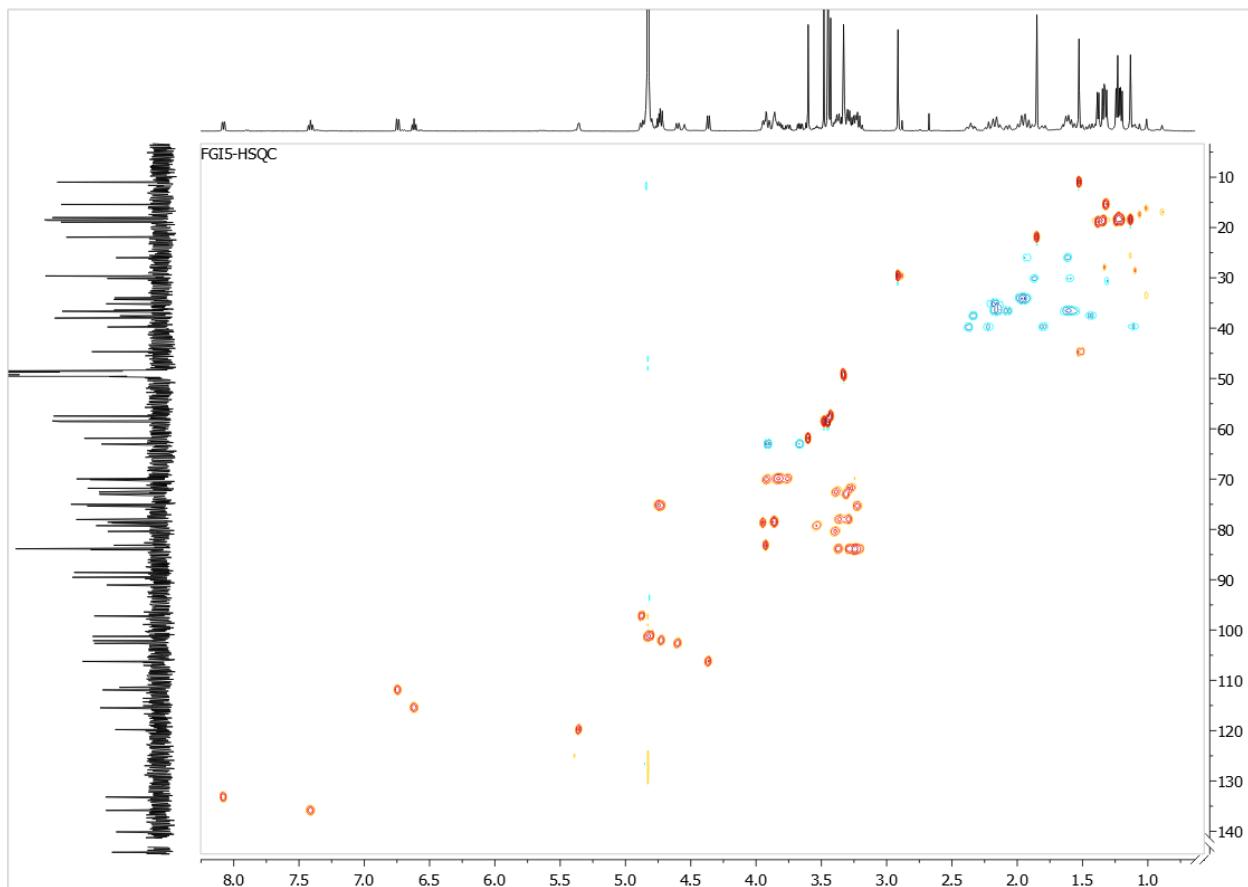
**Figure S10.**  $^1\text{H}$ -NMR spectrum of compound 2



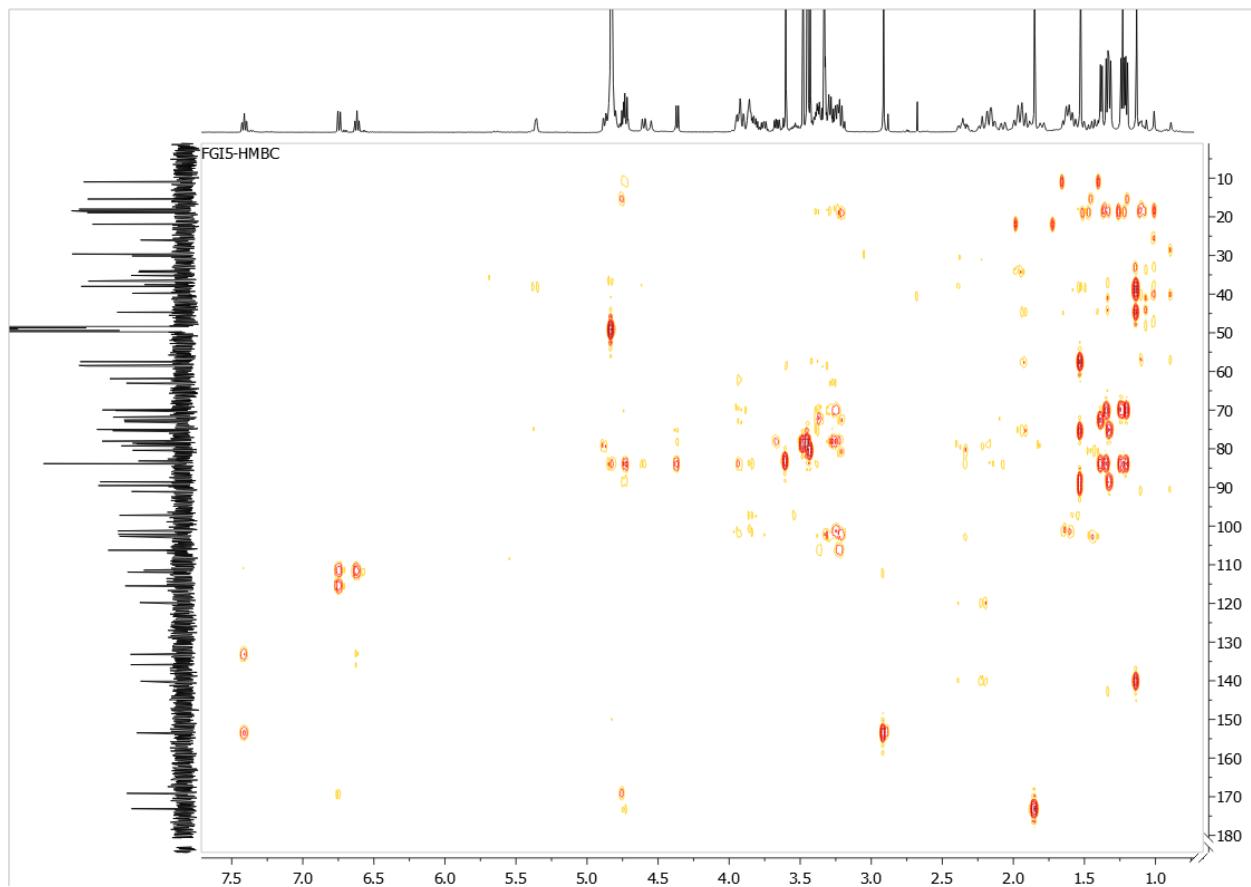
**Figure S11.**  $^{13}\text{C}$ -NMR spectrum of compound 2



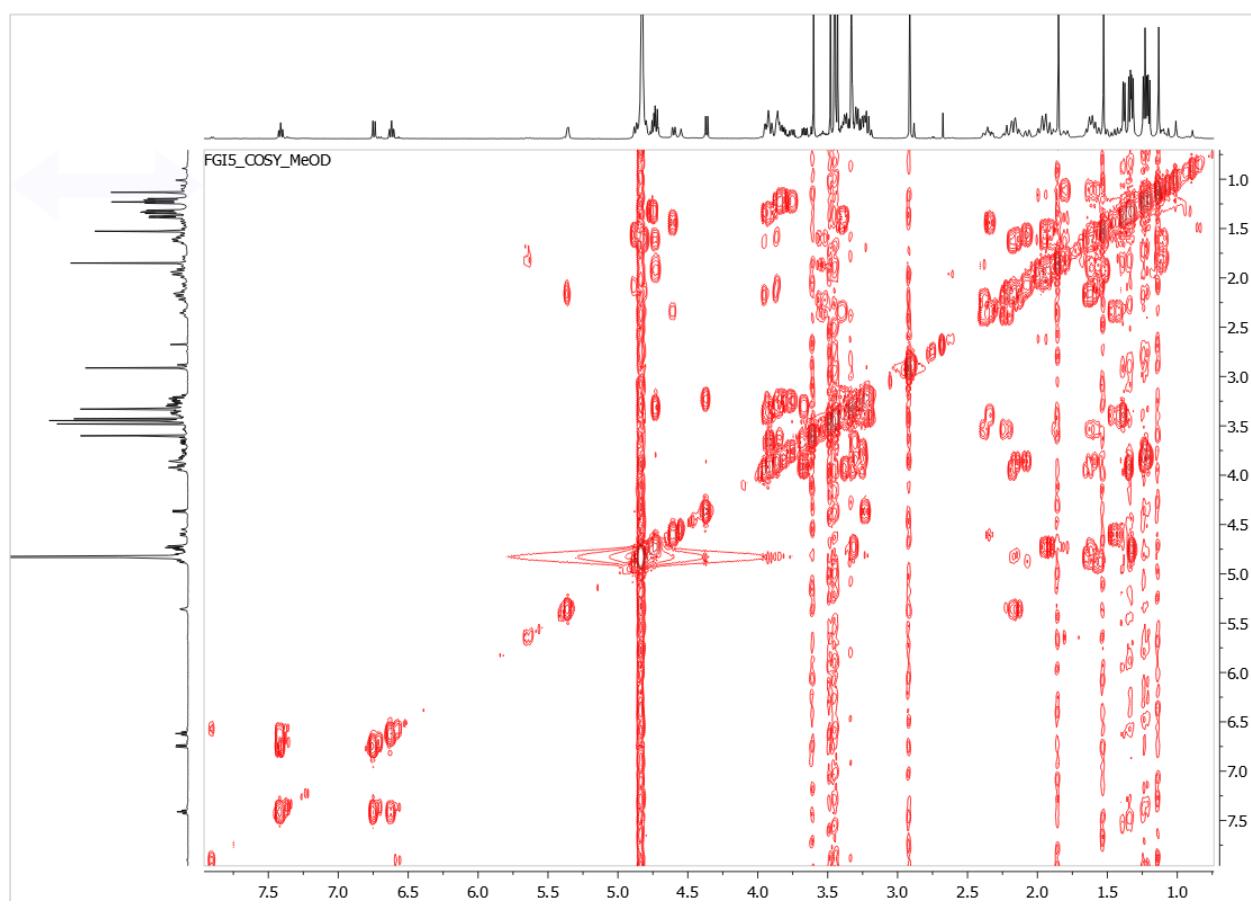
**Figure S12.** HSQC spectrum of compound 2



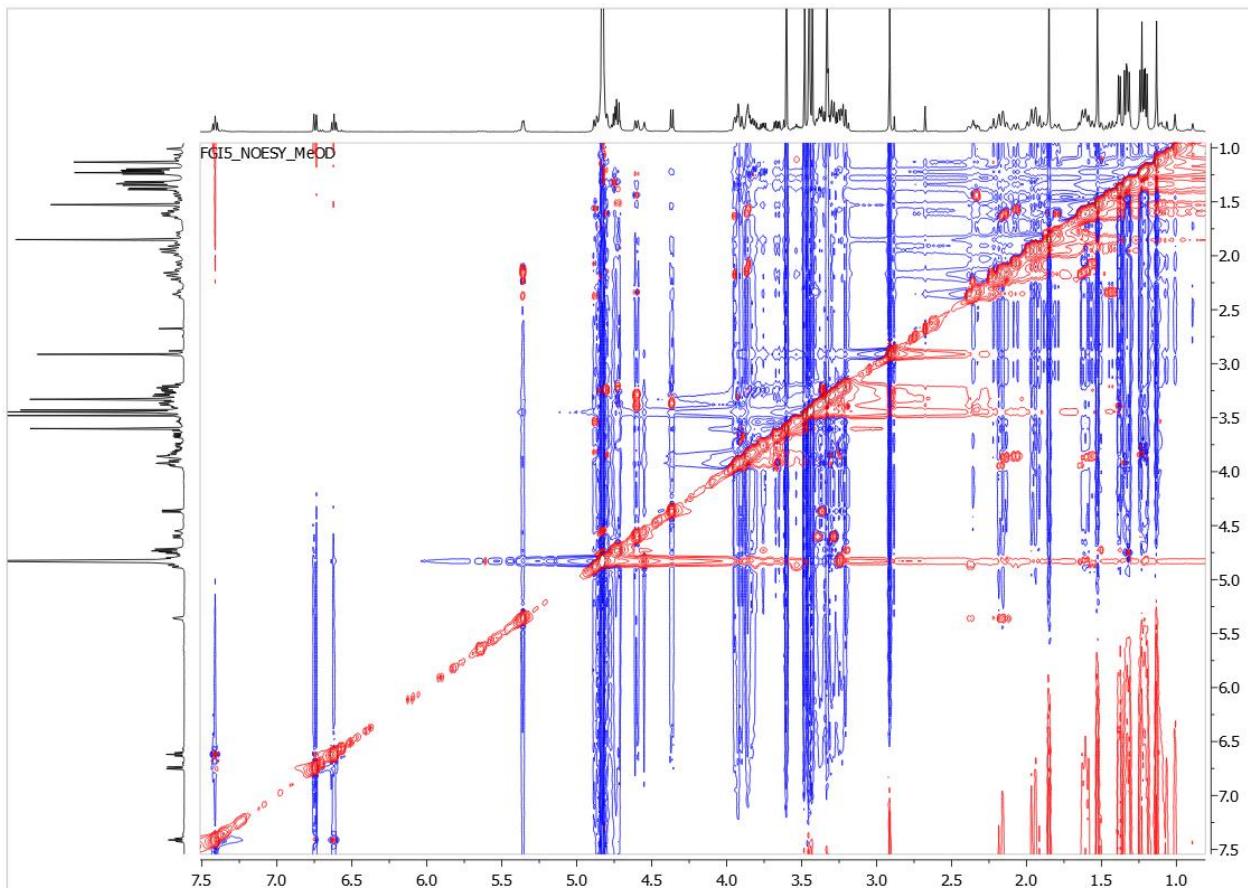
**Figure S13.** HMBC spectrum of compound 2



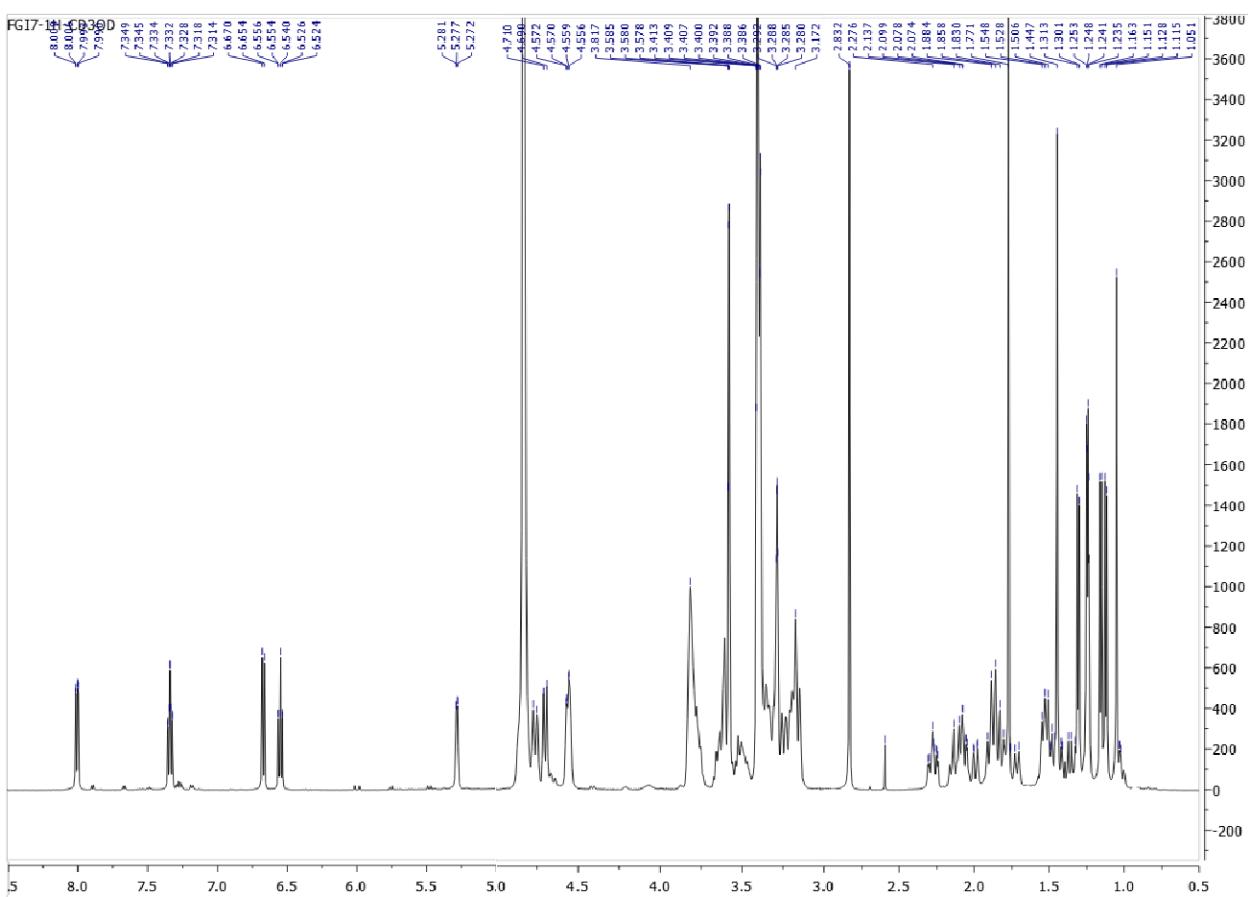
**Figure S14.** COSY spectrum of compound 2



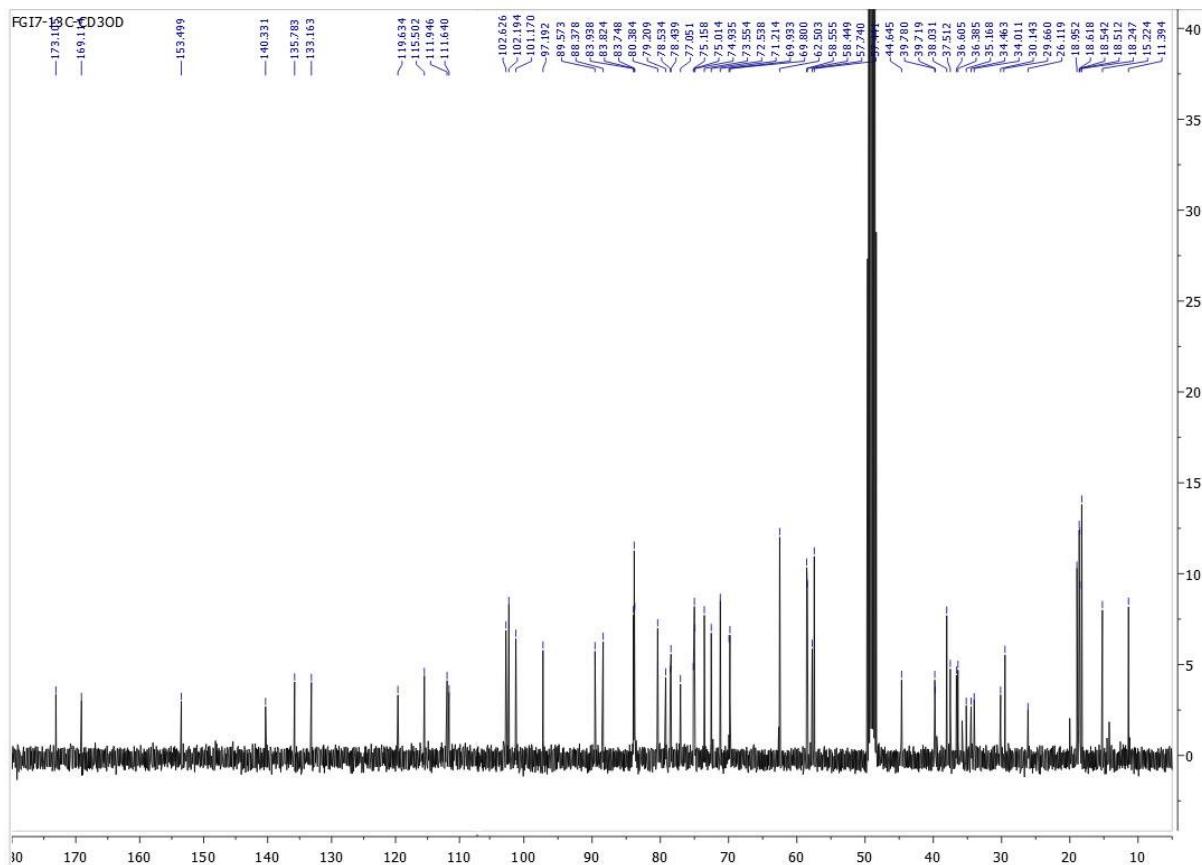
**Figure S15.** NOESY spectrum of compound 2



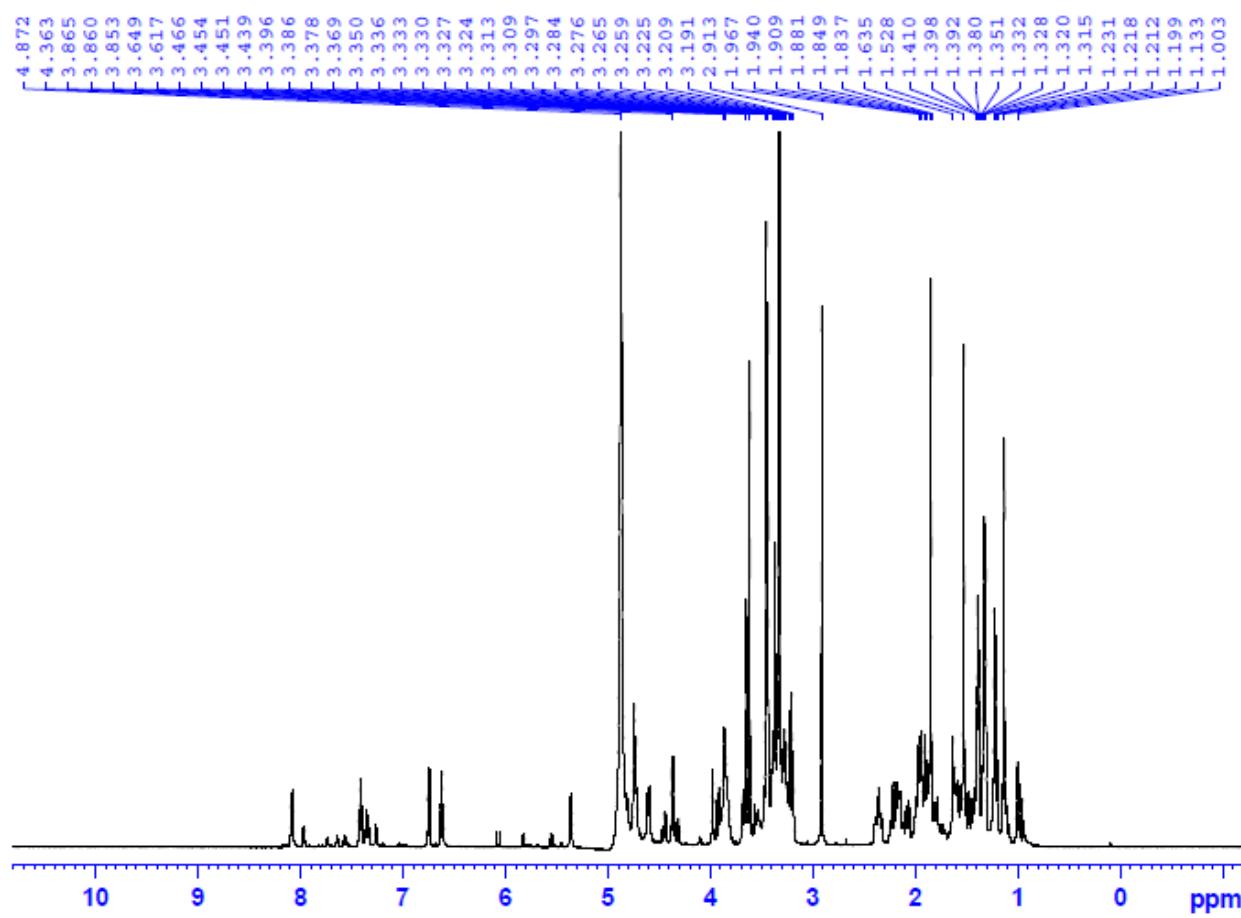
**Figure S16.**  $^1\text{H}$ -NMR spectrum of compound 3



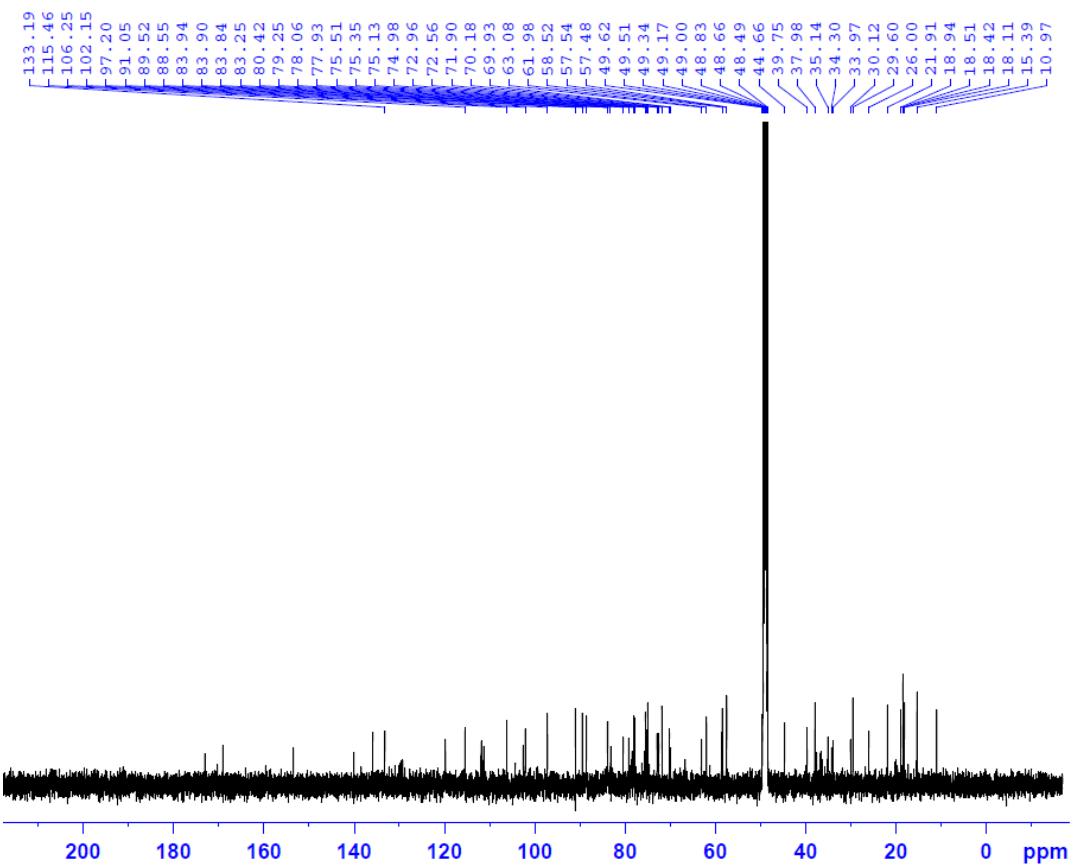
**Figure S17.**  $^{13}\text{C}$ -NMR spectrum of compound 3



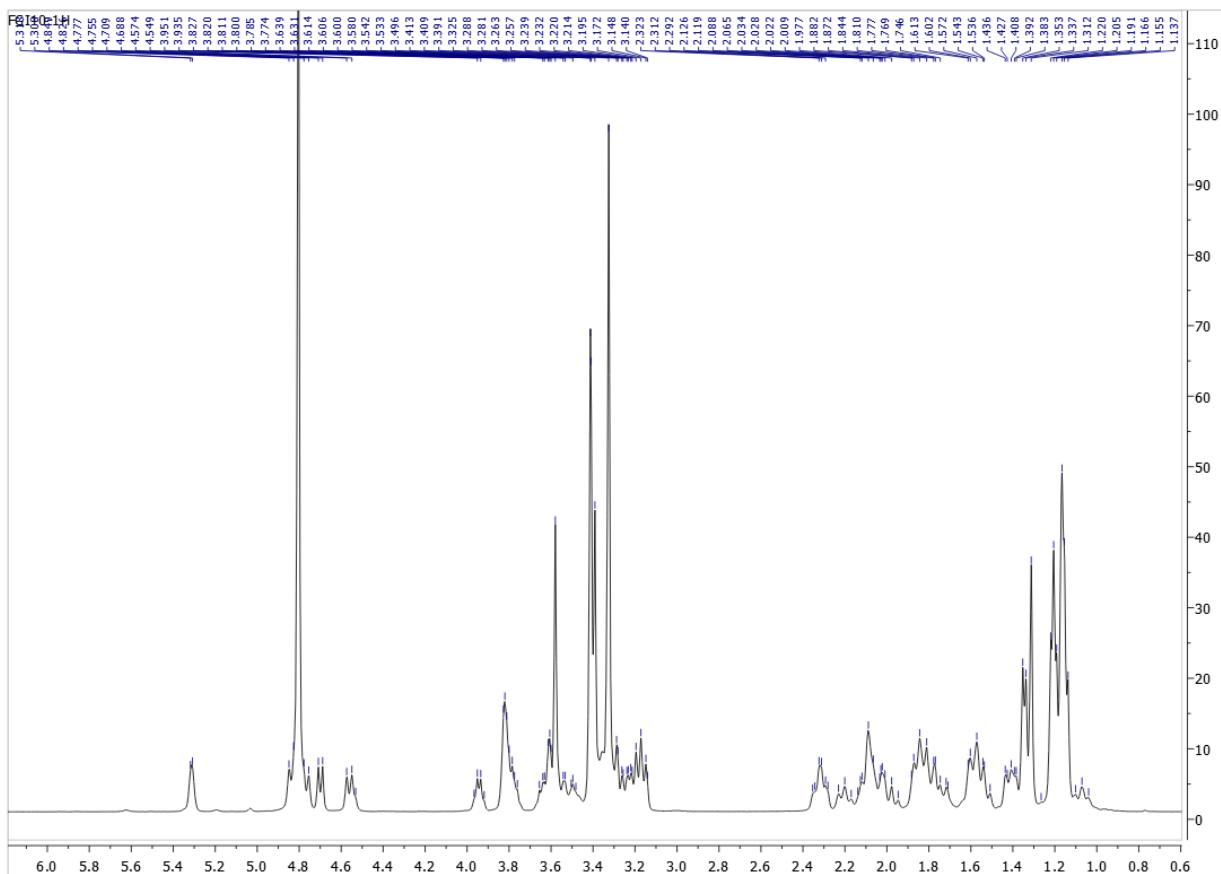
**Figure S18.**  $^1\text{H}$ -NMR spectrum of compound 4



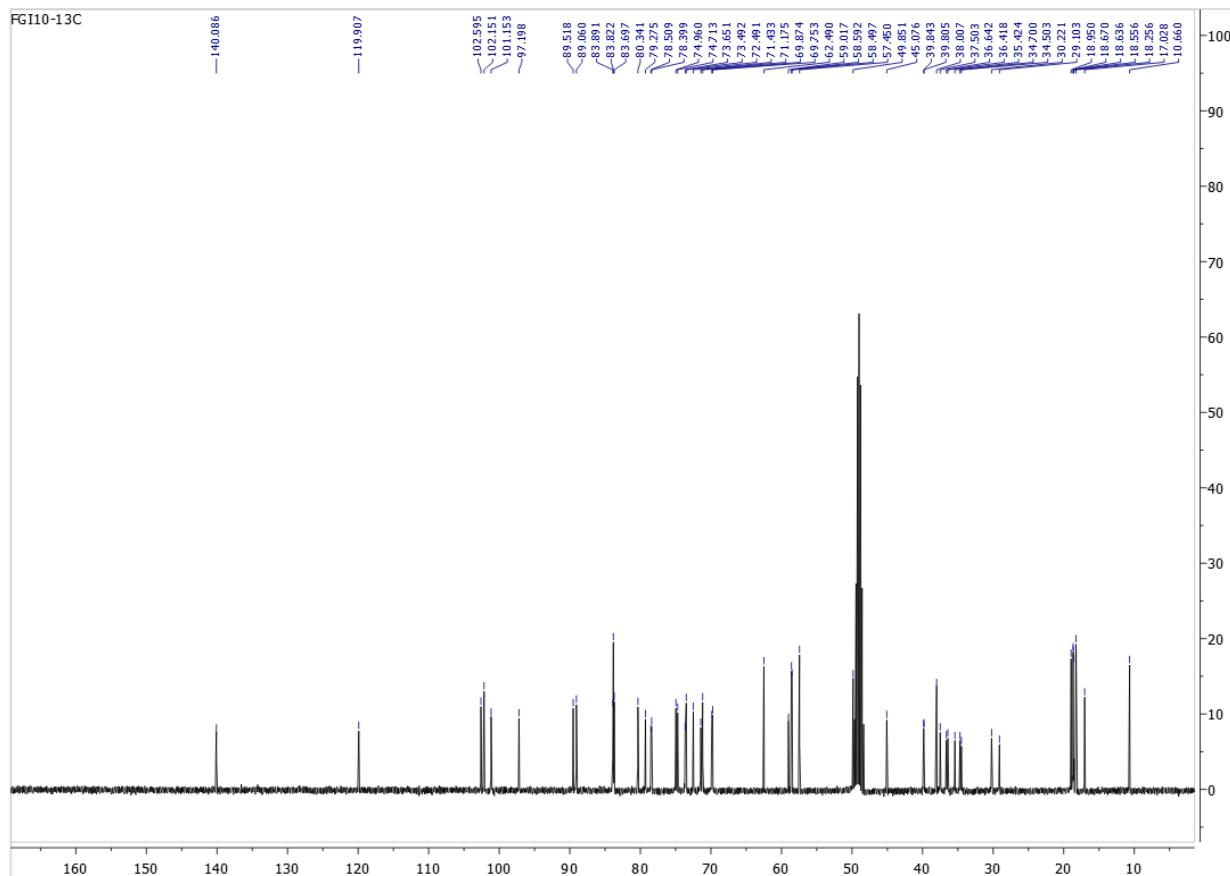
**Figure S19.**  $^{13}\text{C}$ -NMR spectrum of compound 4



**Figure S20.**  $^1\text{H}$ -NMR spectrum of compound 5



**Figure S21.**  $^{13}\text{C}$ -NMR spectrum of compound 5



**Figure S22.**  $\alpha$ -Glucosidase inhibitory activity of compounds 1–5

