

# **Supplementary Materials: A New Triterpenoid Glucoside from a Novel Acidic Glycosylation of Ganoderic Acid A via Recombinant Glycosyltransferase of *Bacillus subtilis***

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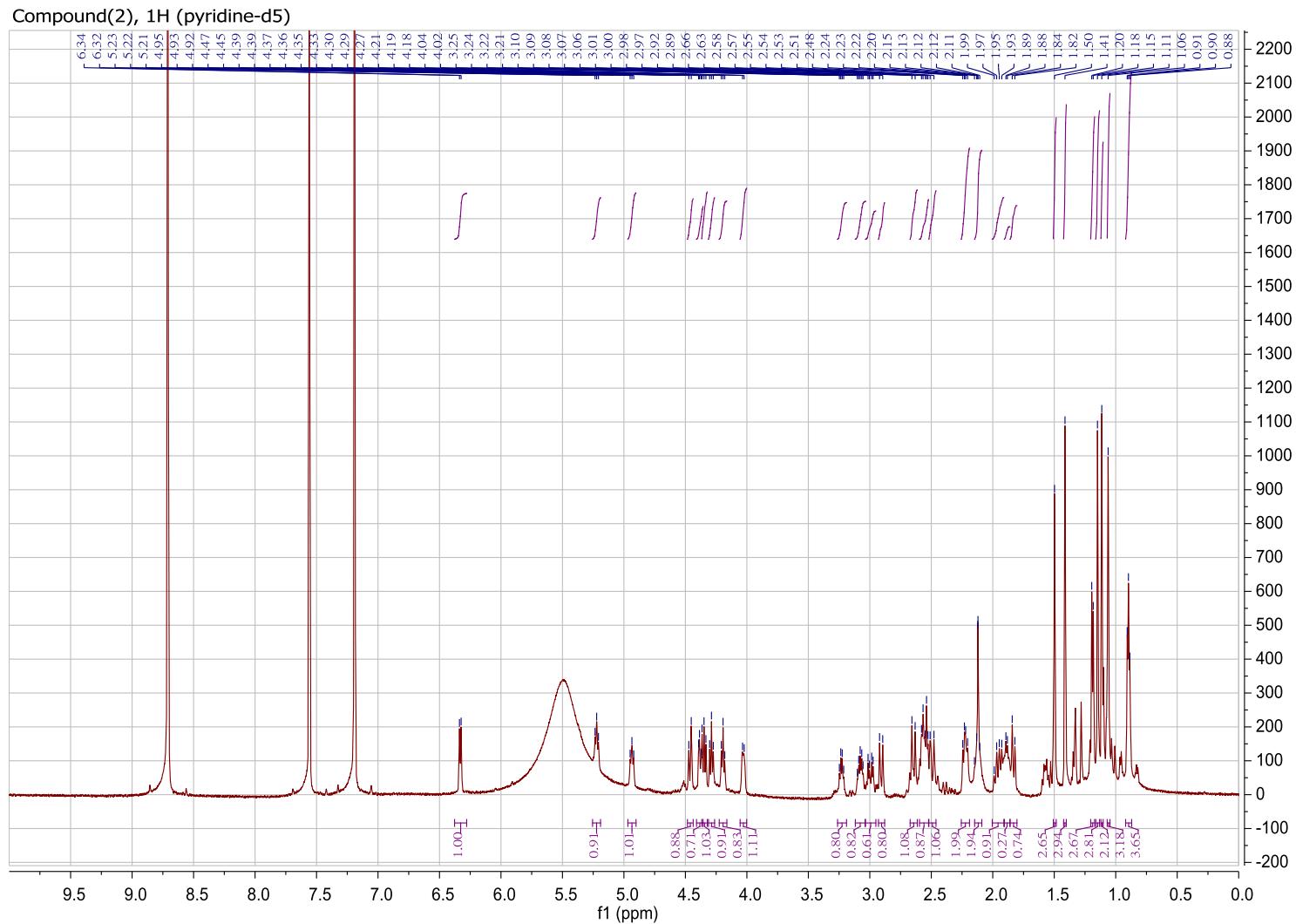
+ The two authors contributed equally

**Table S1.** NMR spectroscopic data for compound (**2**) in pyridine-*d*<sub>5</sub> (600MHz)

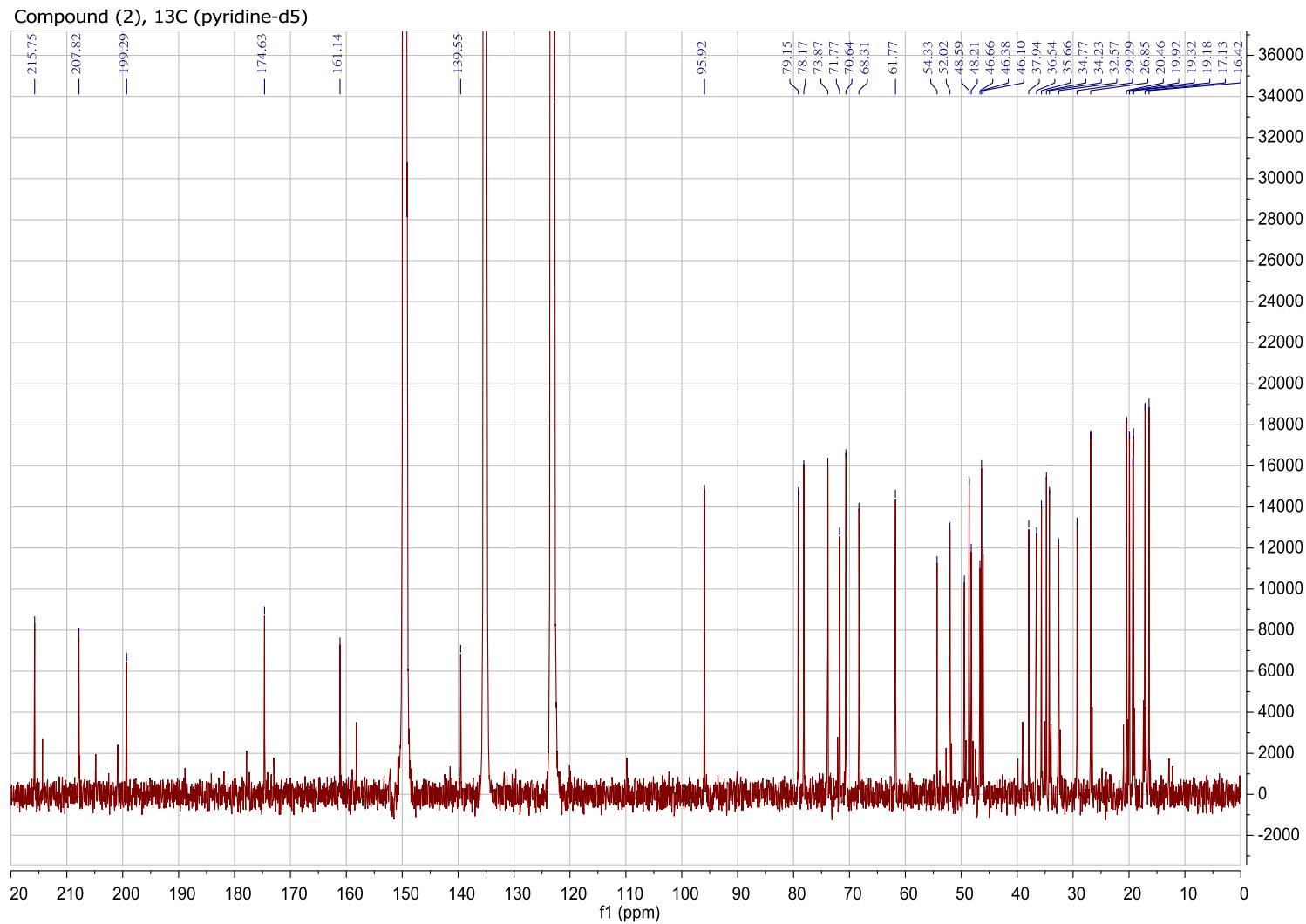
Compound	GAA-26-O- $\beta$ -glucoside Compound ( <b>2</b> )		
Position	$\delta_{\text{C}}$ ,	type	$\delta_{\text{H}}$ ( <i>J</i> in Hz)
GAA moiety			
1	35.7	CH <sub>2</sub>	1.57, m; 3.08, m
2	34.2	CH <sub>2</sub>	2.54, m 2.60, m
3	215.8	C	H-1, H-2, H-28, H-29
4	46.4	C	H-6, H-28, H-29
5	48.6	CH	H-1, H-6, H-19, H-29
6	29.3	CH <sub>2</sub>	1.96, m; 2.24, m
7	68.3	CH	4.93, t (8.8)
8	161.1	C	H-6, H-7, H-15, H-30
9	139.6	C	H-1, H-7, H-12, H-19
10	37.9	C	H-1, H-2, H-5, H-6, H-19
11	199.3	C	H-12,
12	52.0	CH <sub>2</sub>	2.64, d (15.8); 2.92, d (15.8)
13	46.7	C	H-12, H-17, H-18, H-30
14	54.3	C	H-12, H-15, H-16, H-18, H-30
15	71.8	CH	5.22, t (8.3)
16	36.5	CH <sub>2</sub>	1.89, m; 2.12, m
17	48.2	CH	1.88, m
18	17.1	CH <sub>3</sub>	1.06, s
19	19.2	CH <sub>3</sub>	1.41, s
20	32.6	CH	2.13, m
21	19.3	CH	0.90, t (6.5)
22	49.5	CH <sub>2</sub>	2.23, m; 2.49, m
23	207.8	C	H-22, H-24, H-25
24	46.1	CH <sub>2</sub>	2.55, m ; 2.99, m
25	34.8	CH	3.23, m
26	174.6	C	H-24, H-25, H-27, Glc-H-1'
27	16.4	CH <sub>3</sub>	1.19, d (7.3)
28	26.9	CH <sub>3</sub>	1.15, s
29	20.5	CH <sub>3</sub>	1.11, s
30	19.9	CH <sub>3</sub>	1.50, s
Glucose moiety			
1'	95.9		6.33, d (8.1)
2'	73.9		4.20, m
3'	78.2		4.29, m
4'	70.6		4.35, m
5'	79.1		4.03, m
6'	61.8	CH <sub>2</sub>	4.39, m ; 4.46, m

**Table S2.** BsGT110 sequence comparison with candidate triterpenoid-catalyzing GTs and flavonoid-catalyzing GTs.

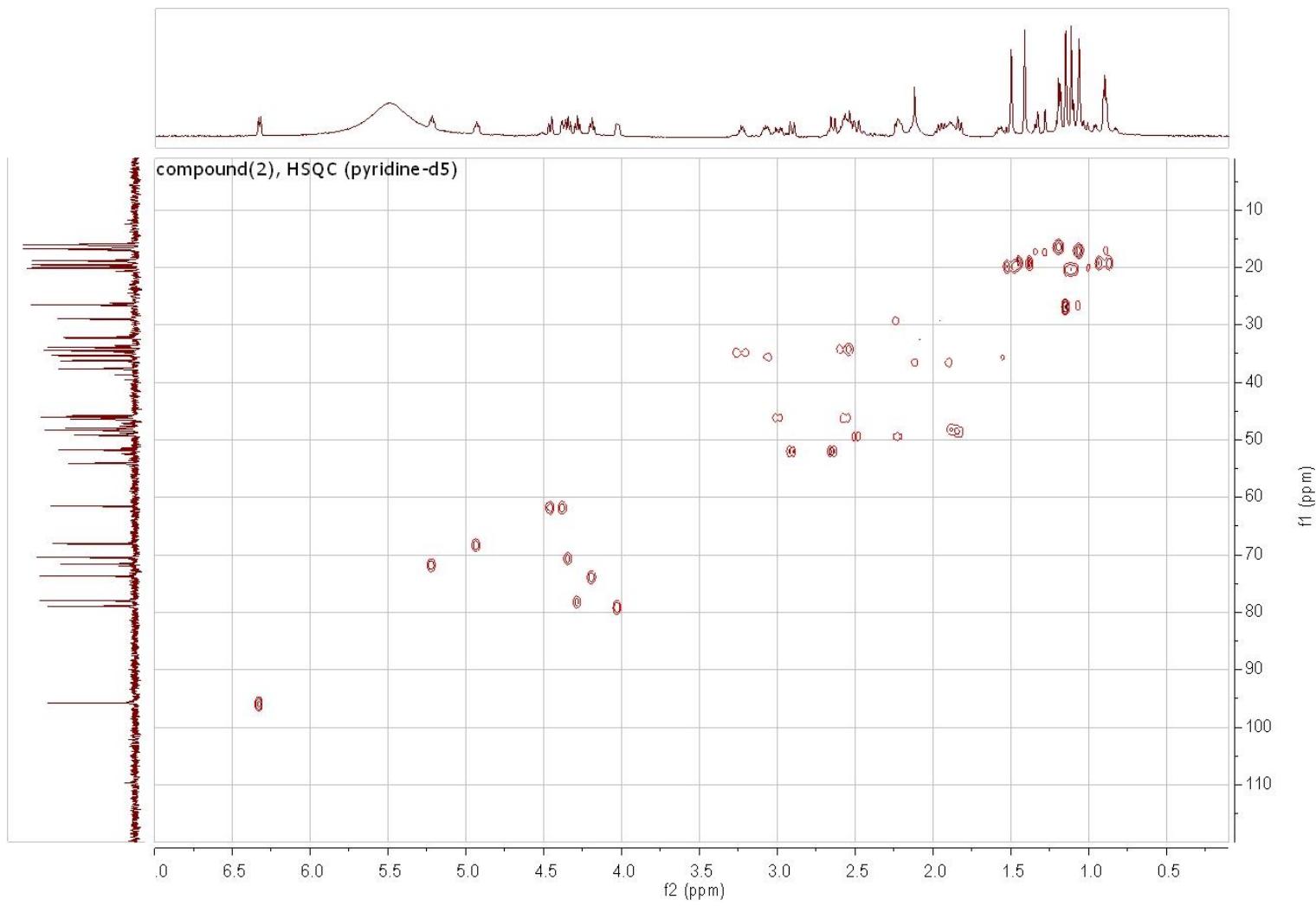
Candidate GTs	E-value	Score	Hit start	Hit end	Hit length	%Identity	%Gaps
BsYjiC_(NP_389104)	1.36E-63	515	5	364	360	<b>31.83</b>	5.57
BsGT1_(ANP92054)	1.36E-63	515	5	364	360	<b>31.83</b>	5.57
UGT109A1_(ASY97769)	2.41E-61	500	5	351	347	<b>31.59</b>	5.77
BsUGT489_(WP_003220489)	2.41E-61	500	5	351	347	<b>31.59</b>	5.77
BsUGT398_(WP_003225398)	3.09E-77	608	1	369	369	<b>33.87</b>	1.6
<b>BsGT110_(WP_003220110)*</b>	<b>0</b>	<b>2065</b>	<b>1</b>	<b>405</b>	<b>405</b>	<b>100</b>	<b>0</b>
BlYjiC_(AAU40842)	3.67E-60	492	5	369	365	<b>30.13</b>	3.2
BcGT1_(AAS41089)	1.46E-103	786	1	373	373	<b>40.69</b>	1.06
BcGT3_(AAS41737)	4.69E-88	682	1	371	371	<b>36.8</b>	1.87
OleD_(ABA42119)	1.42E-48	412	8	377	370	<b>26.87</b>	7.75
XcGT2_(AAM41712)	6.68E-08	100	202	348	147	<b>28.21</b>	8.97



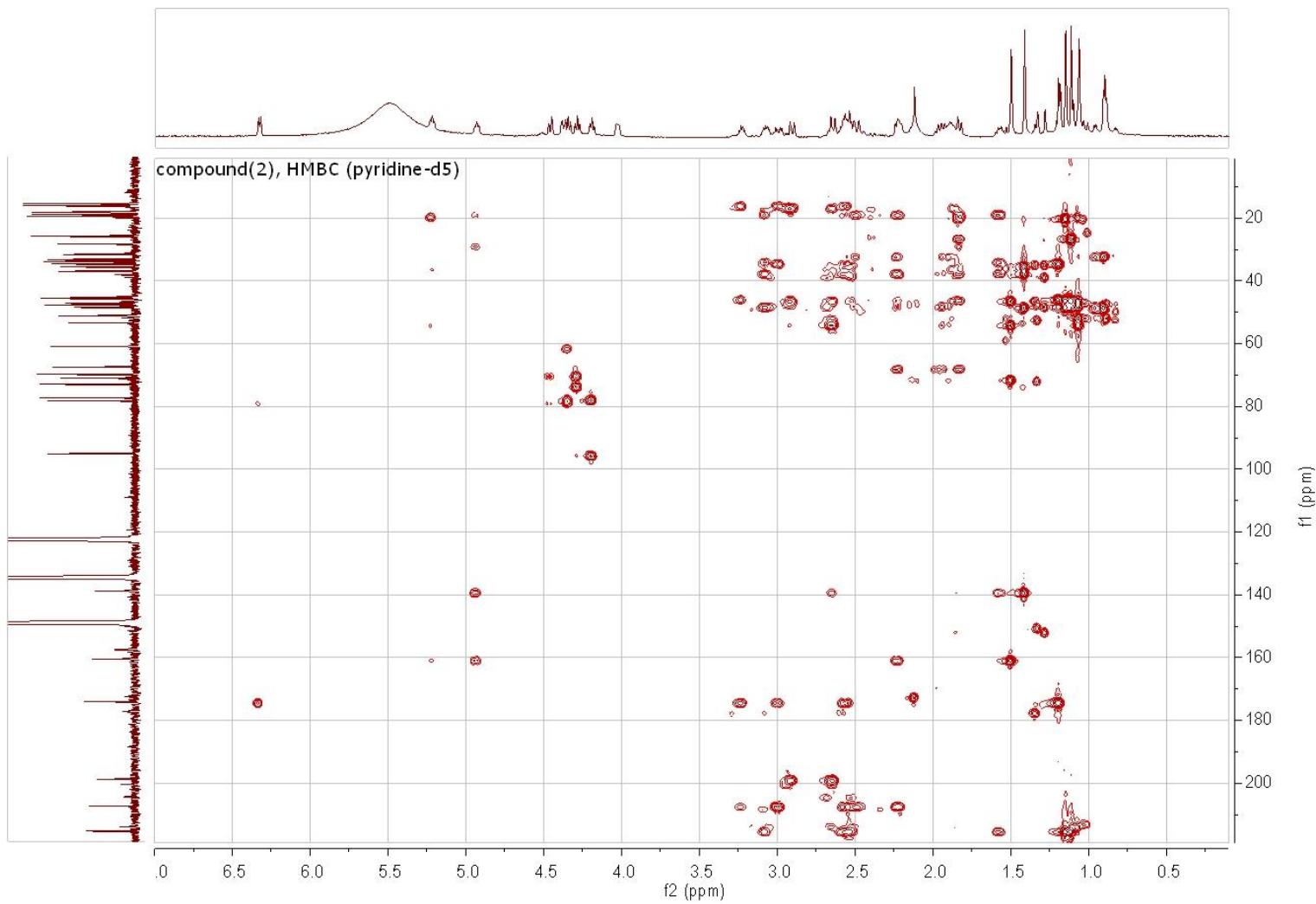
**Figure S1.** The  $^1\text{H}$ -NMR (600 MHz) spectrum of compound (2) in pyridine- $d_5$ .



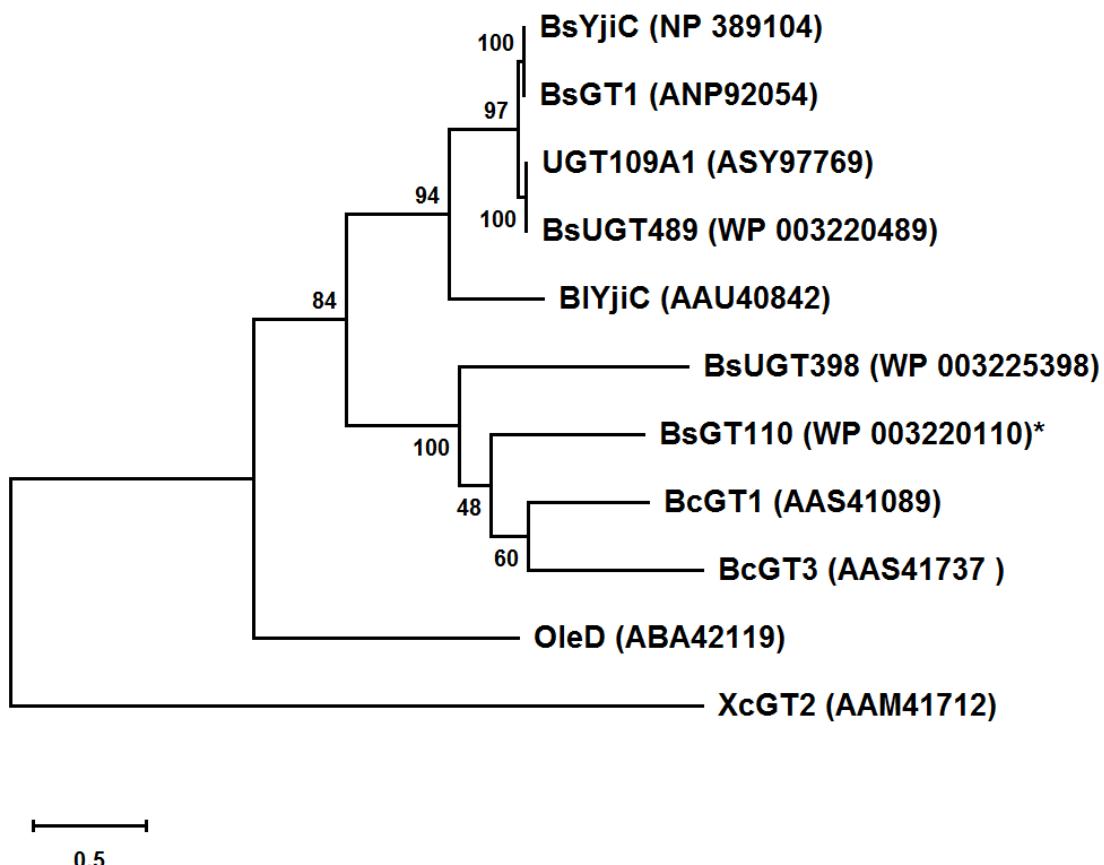
**Figure S2.** The  $^{13}\text{C}$ -NMR (150 MHz) spectrum of compound (2) in pyridine-d<sub>5</sub>.



**Figure S3.** The HSQC (600 MHz) spectrum of compound (2) in pyridine-*d*5.



**Figure S4.** The HMBC (600 MHz) spectrum of compound (**2**) in pyridine-*d*5.



**Figure S5.** Phylogenetic analysis using the Maximum Likelihood method. The evolutionary history was inferred by using the Maximum Likelihood method and General Reversible Mitochondrial model. The tree with the highest log likelihood (-7192.36) is shown. The percentage of trees in which the associated taxa clustered together is shown next to the branches. Initial tree(s) for the heuristic search were obtained automatically by applying Neighbor-Join and BioNJ algorithms to a matrix of pairwise distances estimated using a JTT model, and then selecting the topology with superior log likelihood value. The tree is drawn to scale, with branch lengths measured in the number of substitutions per site. This analysis involved 11 amino acid sequences. All positions with less than 95% site coverage were eliminated, i.e., fewer than 5% alignment gaps, missing data, and ambiguous bases were allowed at any position (partial deletion option). There were a total of 368 positions in the final dataset. Evolutionary analyses were conducted in MEGA X.



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