SUPPLEMENTARY MATERIAL

Two new sesquiterpenes derivatives from marine fungus *Leptosphaerulina Chartarum* sp. 3608

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Abstract: Two new sesquiterpenes, leptoterpenes A (**1**) and B (**2**) were isolated from the fungus *Leptosphaerulina Chartarum* sp. 3608, derived from a crinoid. It's the first chemical study on this species. The structures of these compounds were elucidated by spectroscopic methods including NMR and MS spectrometry. The absolute configurations of the new compounds were determined on the basis of the single-crystal X-ray diffraction and electronic circular dichroism data analysis. All compounds were tested for their anti-inflammatory activity and the inhibitory effects on Tyrosyl DNA phosphodiesterase II (TDP2).

Keywords: *Leptosphaerulina Chartarum*; cadinane-type sesquiterpenes; secondary metabolites

Supplementary Material List

Figure S1. ^{1}H (400 MHz) NMR Spectrum of (**1**) in Pyridine-d_5

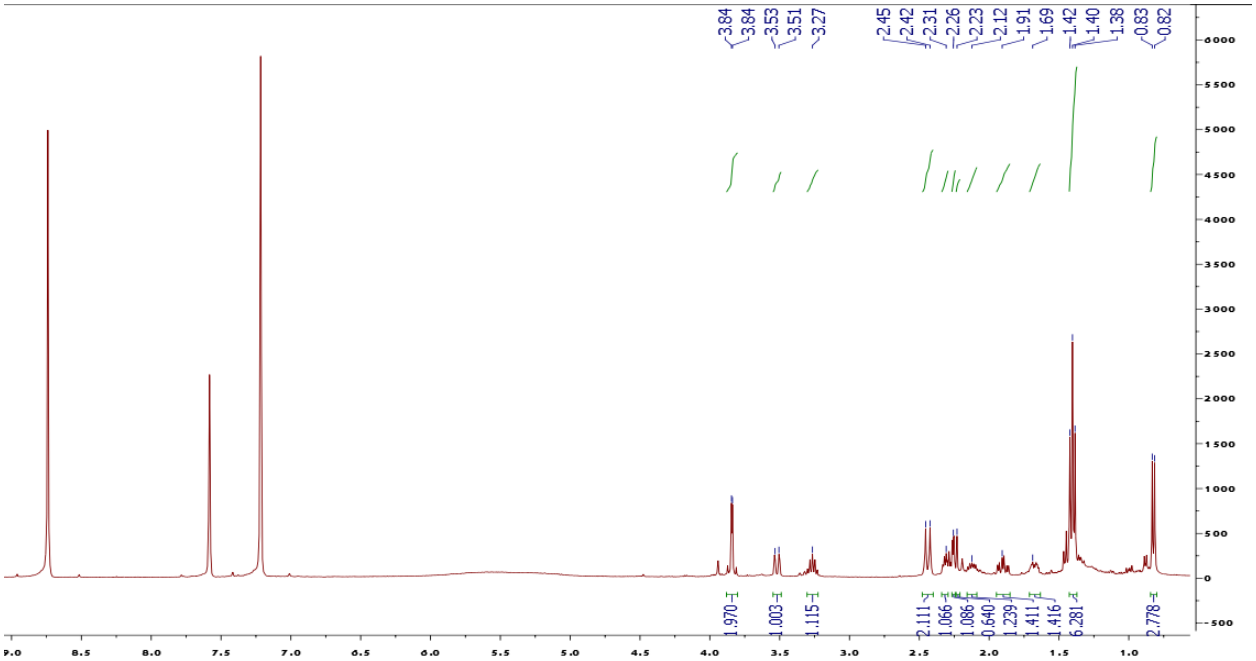


Figure S2. ^{13}C (100 MHz) NMR Spectrum of (**1**) in Pyridine-d_5

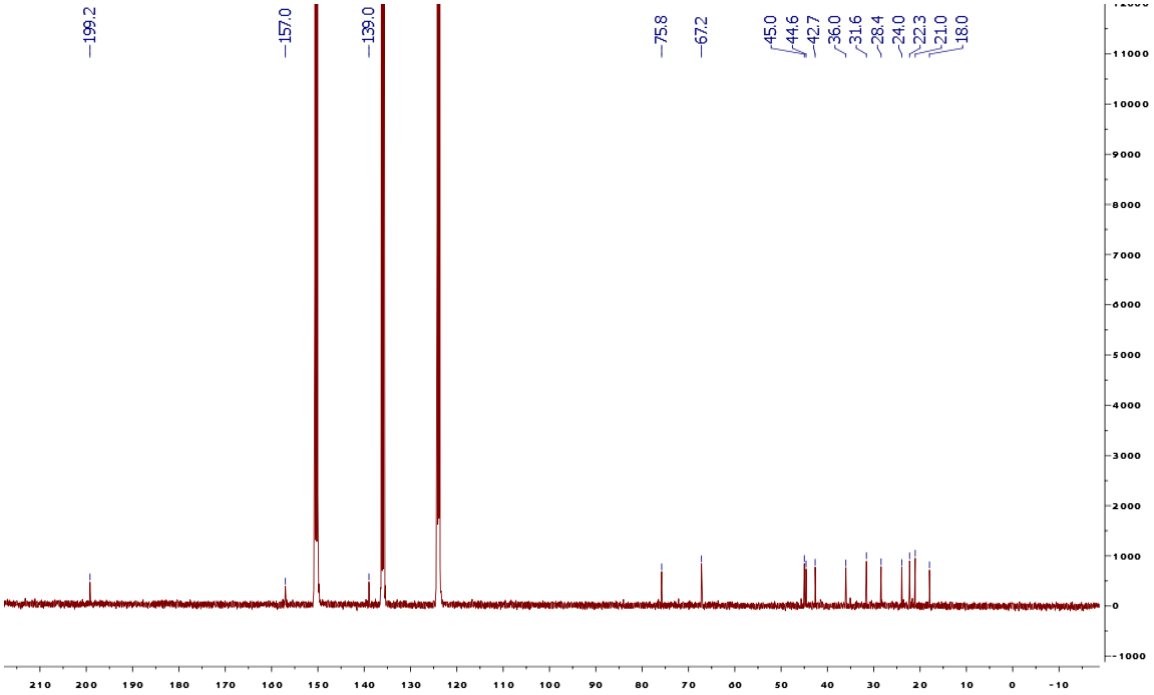


Figure S3. DEPT-90 Spectrum of (**1**) in Pyridine-d_5

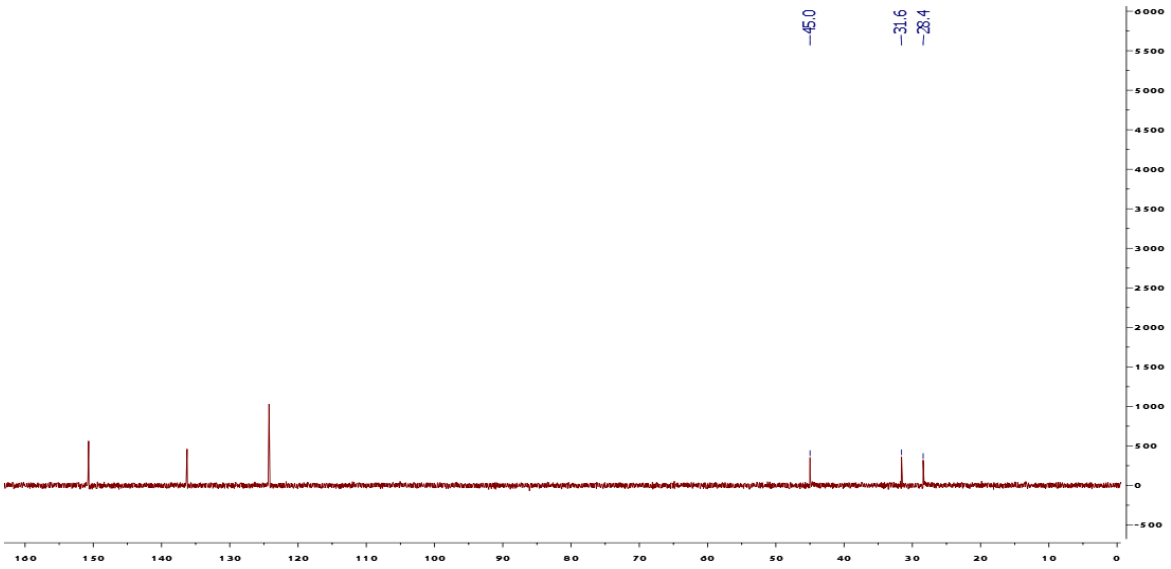


Figure S4. DEPT-135 Spectrum of (**1**) in Pyridine-d_5

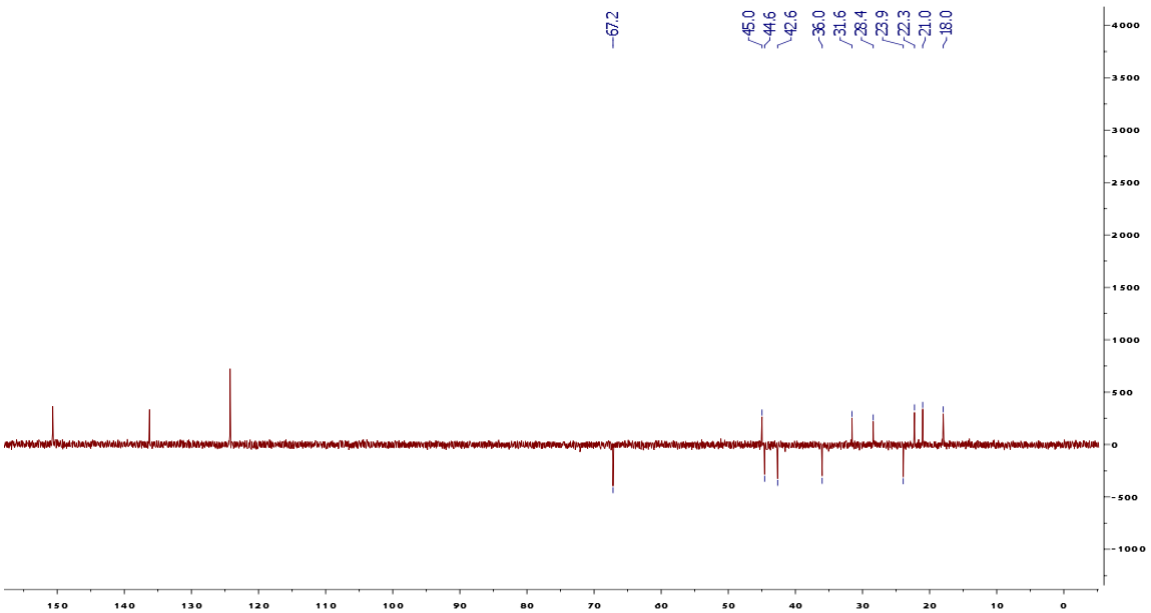


Figure S5. HSQC Spectrum of (**1**) in Pyridine-d_5

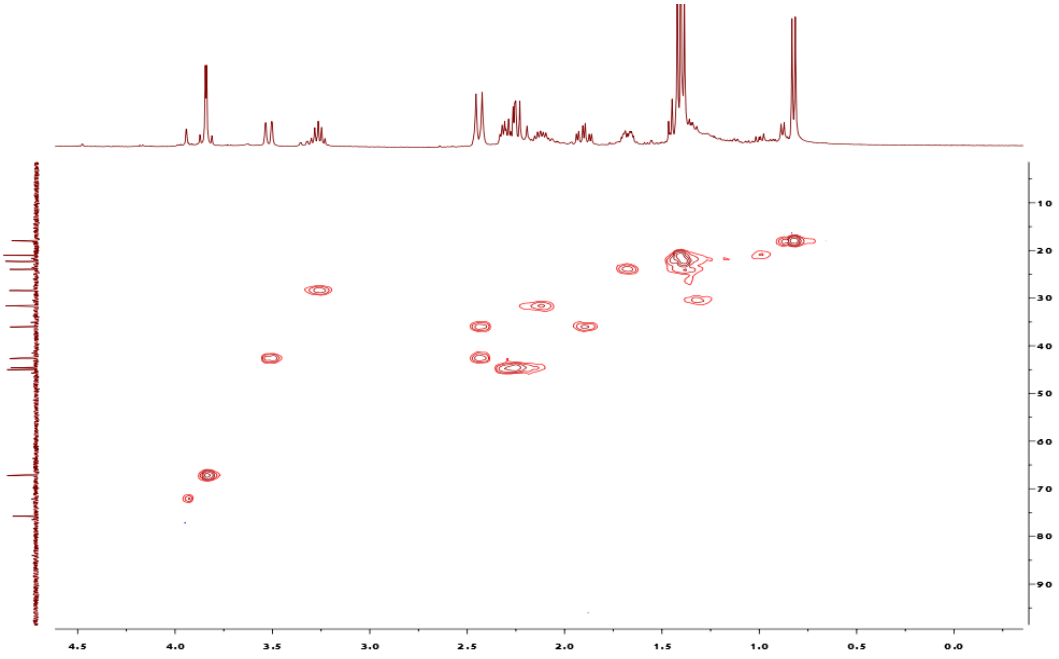


Figure S6. HMBC Spectrum of (**1**) in Pyridine-d_5

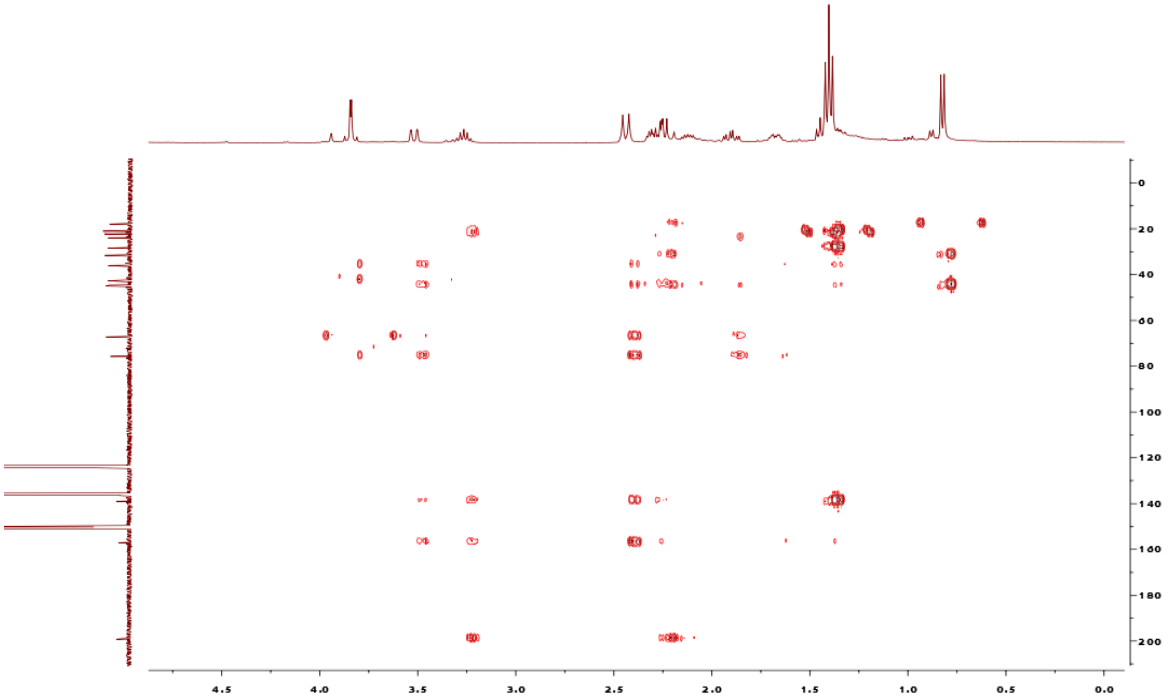


Figure S7. ^{1}H-^{1}H COSY Spectrum of (**1**) in Pyridine-d_5

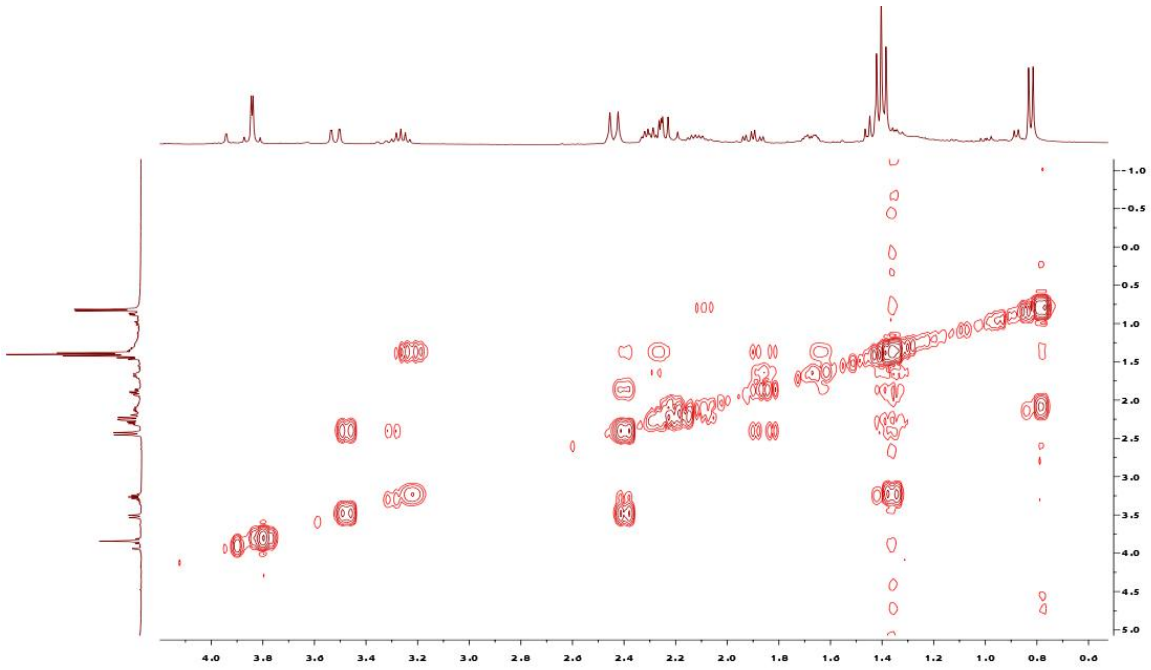


Figure S8. NOESY Spectrum of (**1**) in Pyridine-d_5

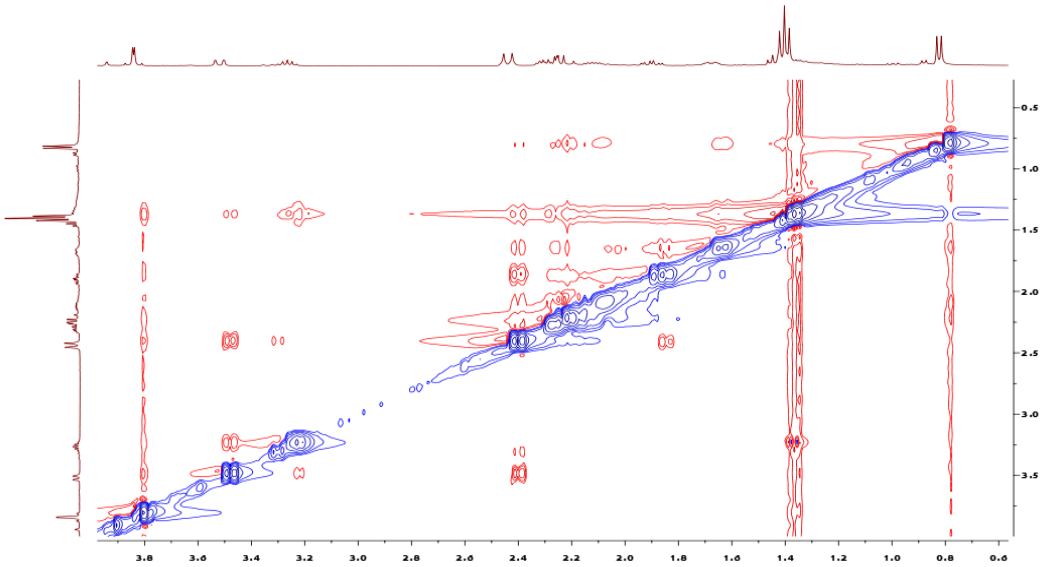
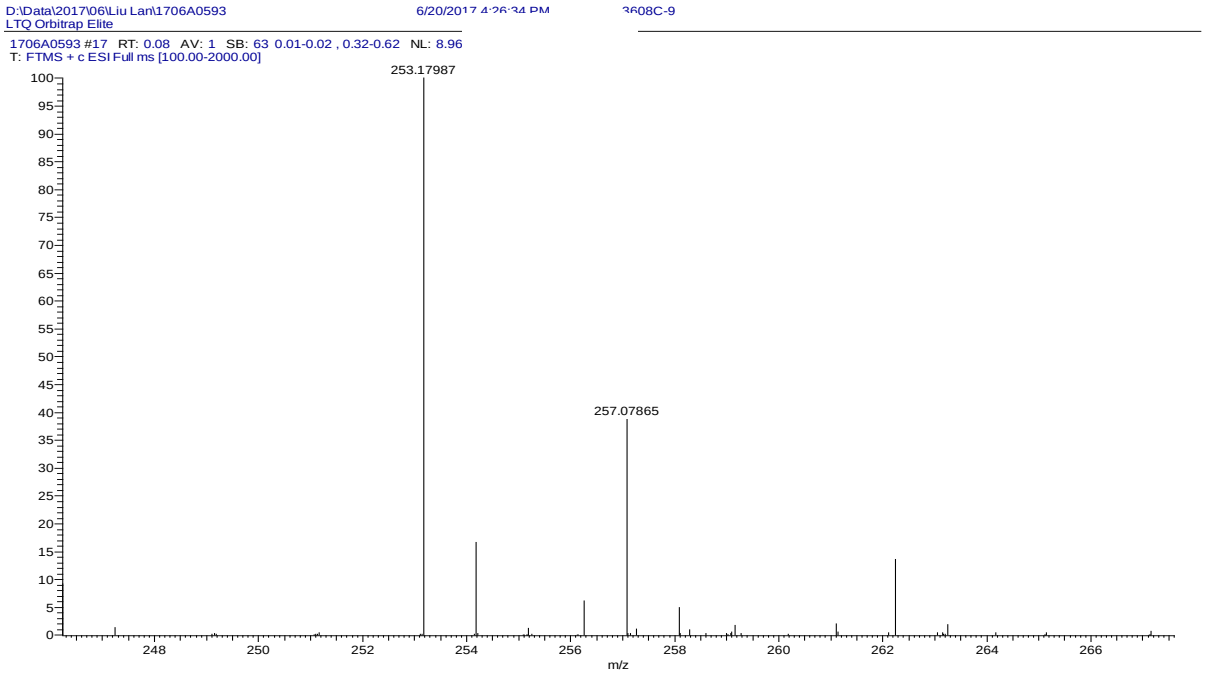


Figure S9. HR-ESIMS of (**1**)



SPECTRUM - simulation :				
m/z	Theo. Mass	Delta (ppm)	RDB equiv.	Composition
253.17987	253.17982	0.19	3.5	$C_{15}H_{25}O_3$

Figure S10. IR Spectrum of (**1**)

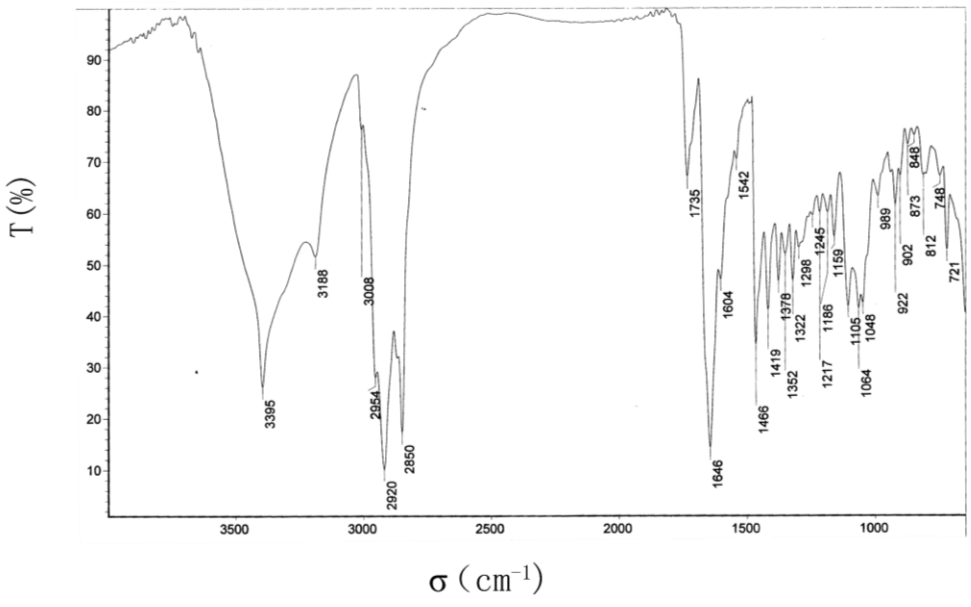


Figure S11. ^{1}H (400 MHz) NMR Spectrum of (**2**) in Pridine-d_5

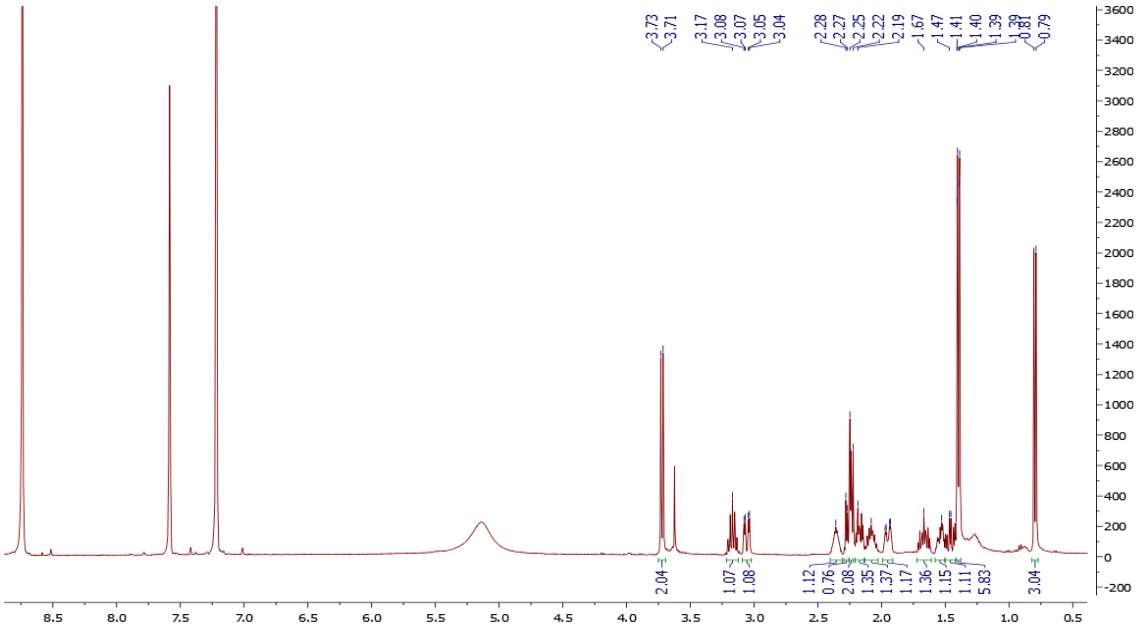


Figure S12. ^{13}C (100 MHz) NMR Spectrum of (**2**) in Pridine-d_5

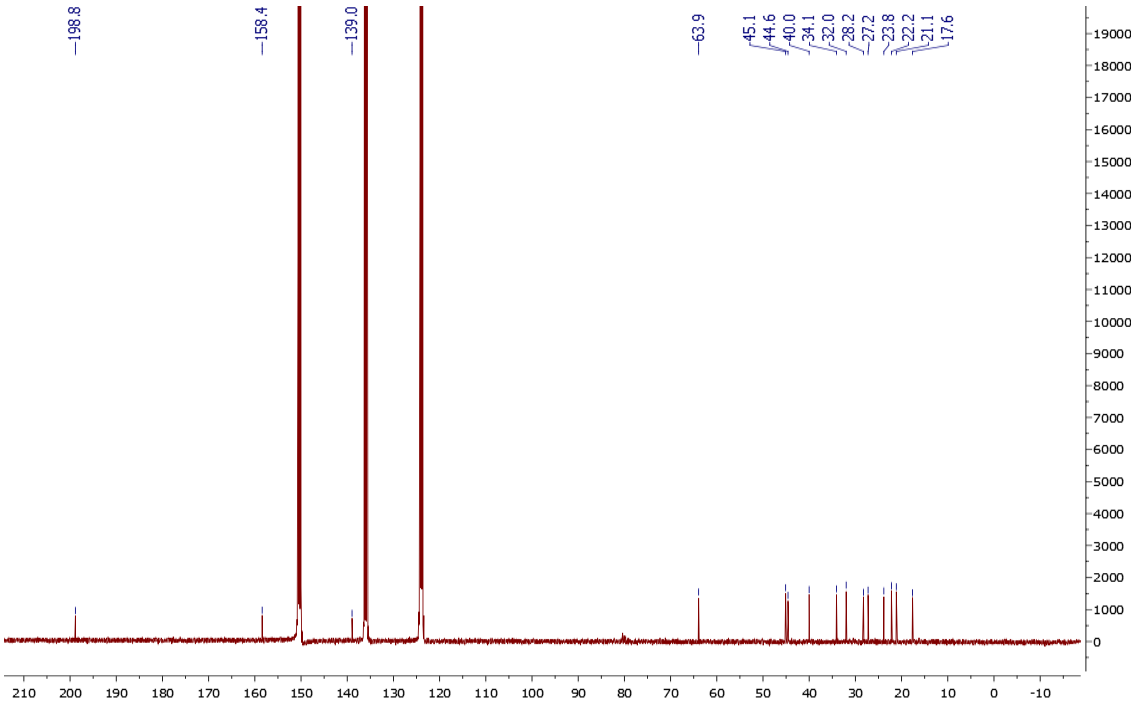


Figure S13. DEPT-90 Spectrum of (**2**) in Pridine-d_5

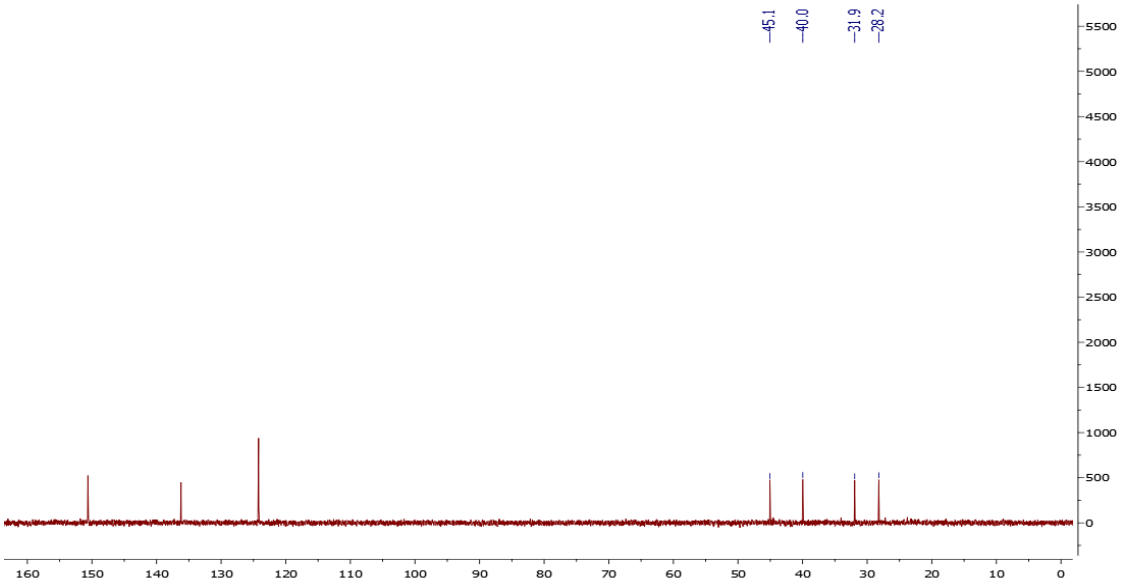


Figure S14. DEPT-135 Spectrum of (**2**) in Pridine-d_5

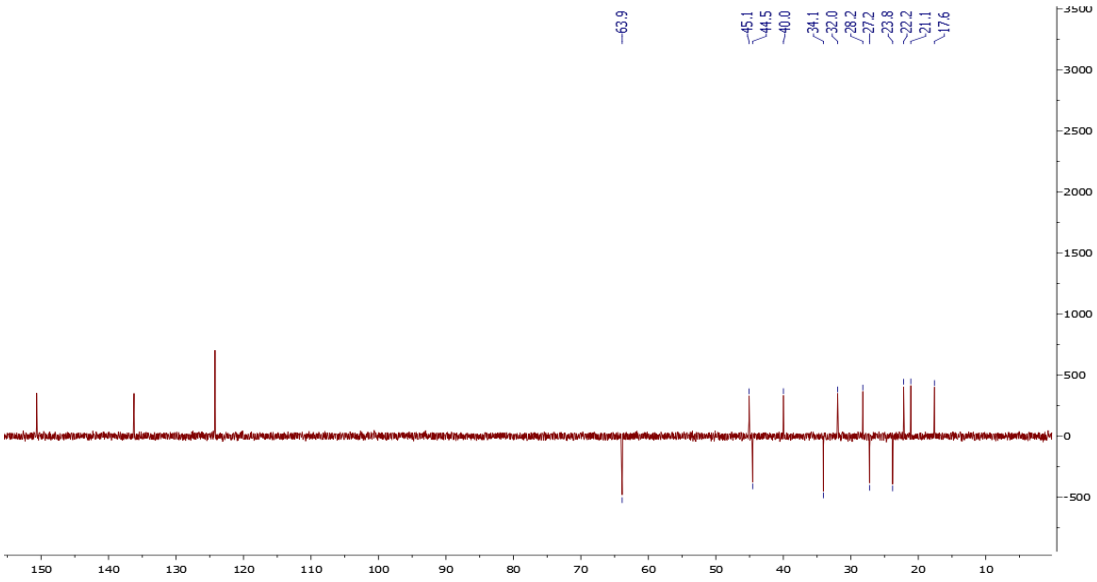


Figure S15. HSQC Spectrum of (**2**) in Pridine-d_5

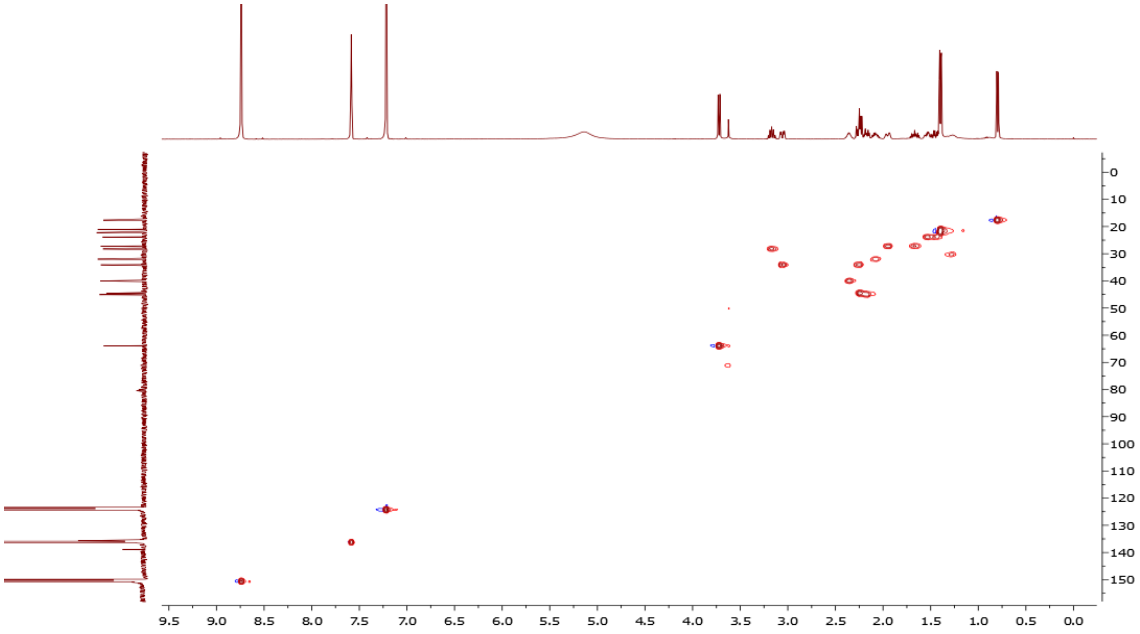


Figure S16. HMBC Spectrum of (**2**) in Pridine-d_5

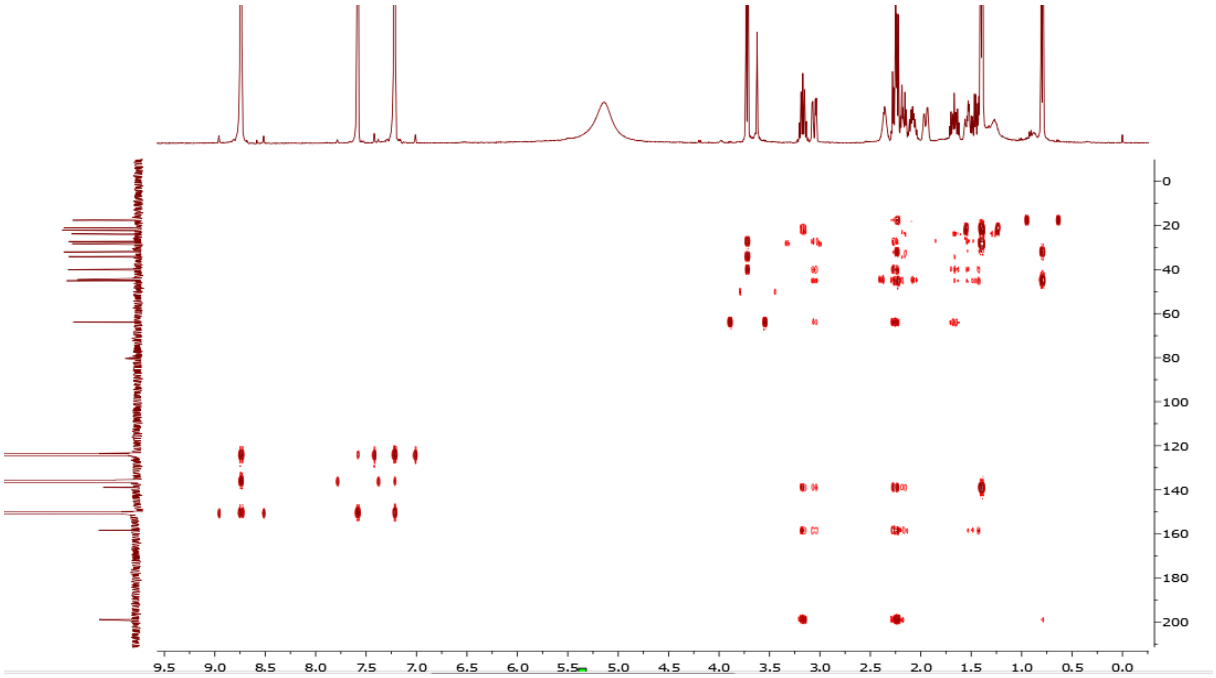


Figure S17. ^{1}H-^{1}H COSY Spectrum of (**2**) in Pridine-d_5

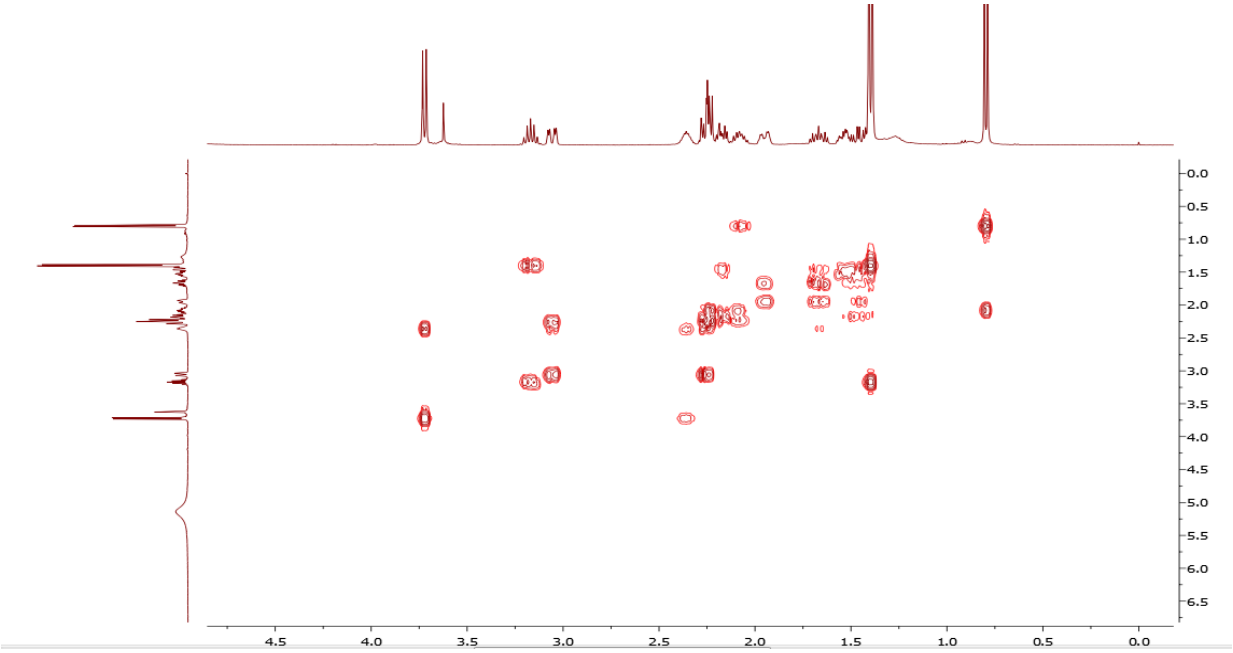


Figure S18. NOESY Spectrum of (**2**) in Pridine-d_5

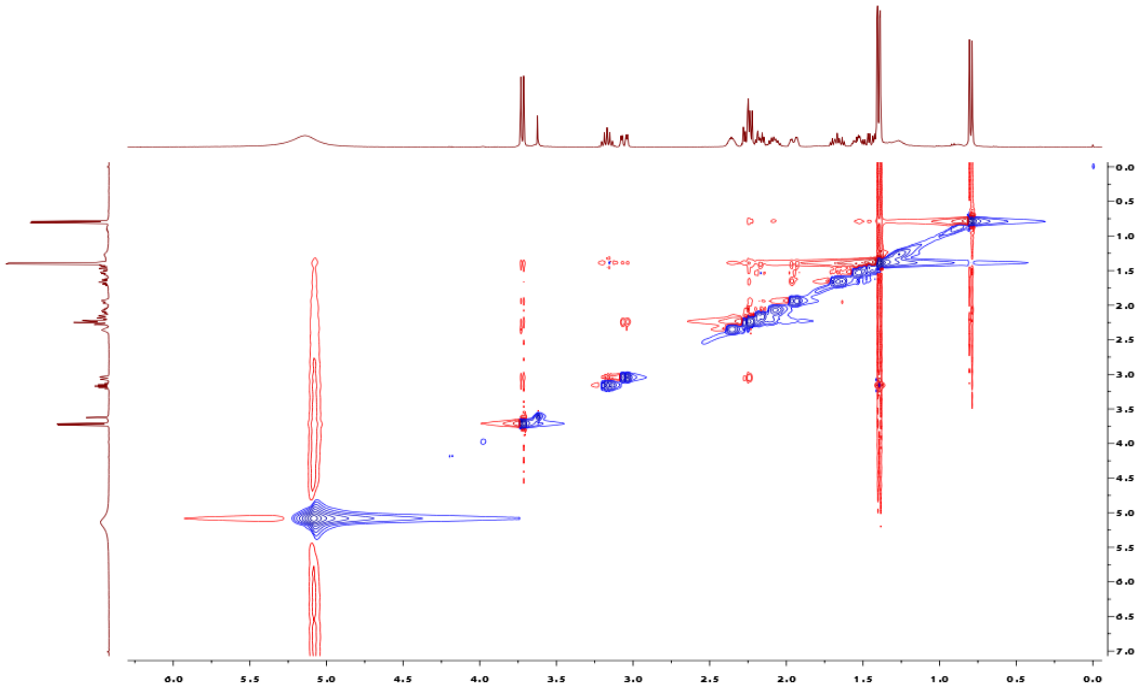
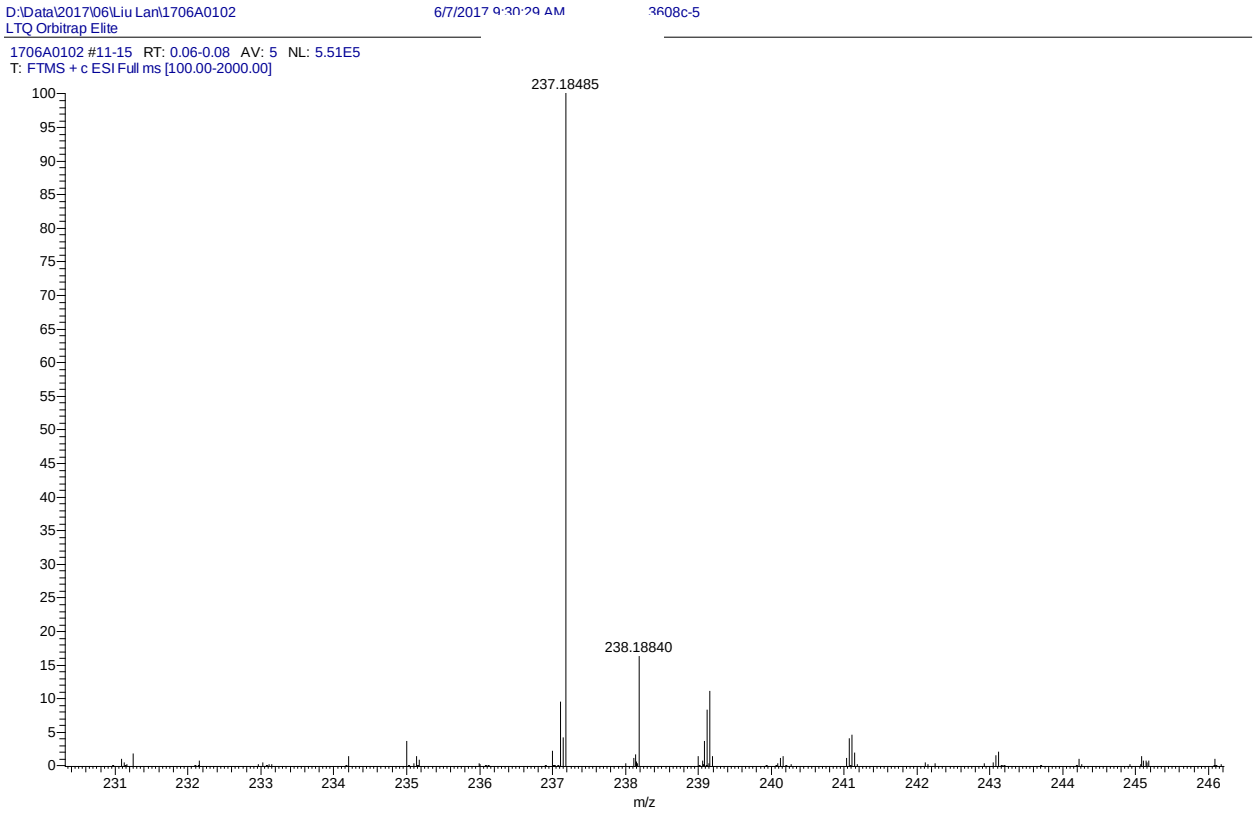


Figure S19. HR-ESIMS of (**2**)



SPECTRUM - simulation :				
m/z	Theo. Mass	Delta (ppm)	RDB equiv.	Composition
237.18485	237.18491	-0.24	3.5	$C_{15}H_{25}O_2$

Figure S20. IR spectrum of (**2**)

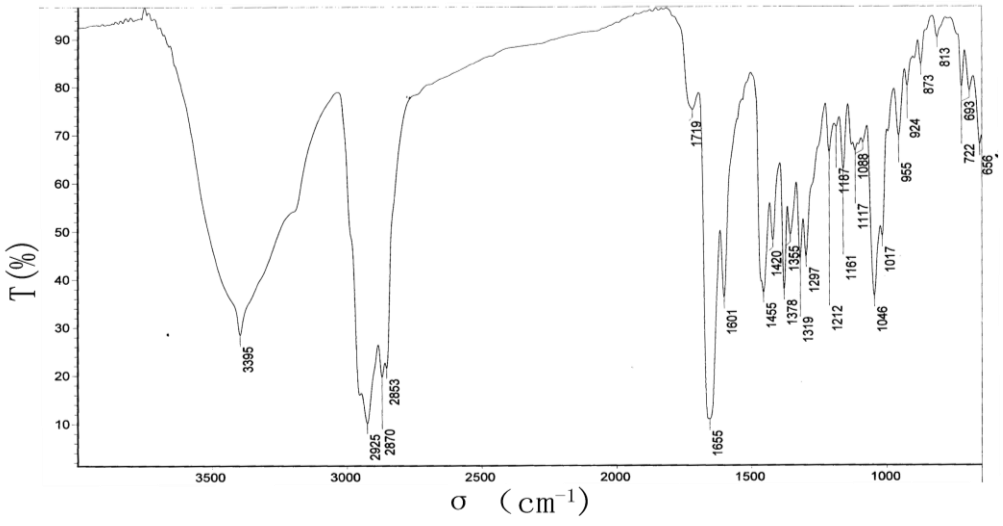


Figure S21. ECD spectra of **1** and 2

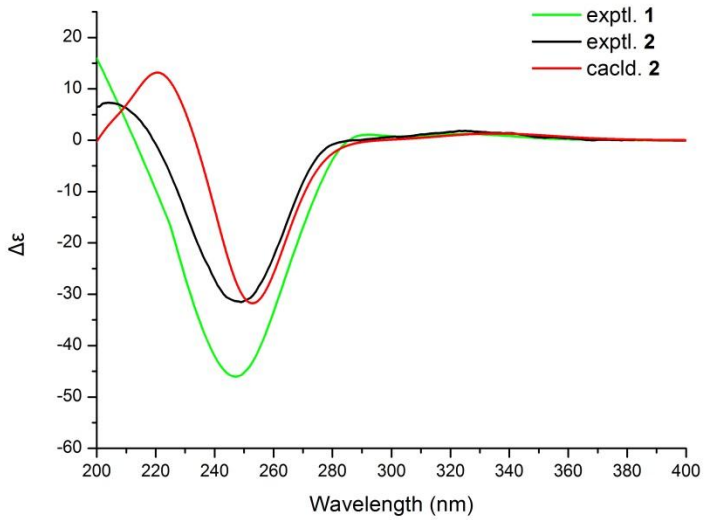


Figure S22. Key ^{1}H-^{1}H COSY (red lines), HMBC (blue arrow) and NOE (dashed arrow) correlations of **1-2**.

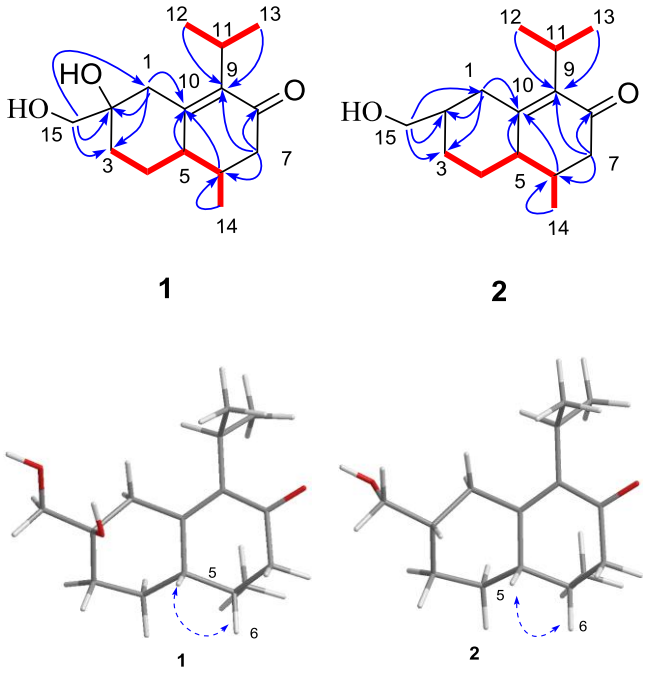


Table S1. ^{1}H (400 MHz) and ^{13}C NMR (100 MHz) Data of **1** and **2** in Pridine-d_5

No.	**1**		**2**	
	δ_C, type	δ_H (J in Hz)	δ_C, type	δ_H (J in Hz)
1.0	42.7, CH_2	3.52, dd (12.7, 1.6); 2.44, d (12.7)	34.1, CH_2	3.06, dd (13.2, 3.7); 2.25, m
2	75.8, C		40.0, CH	2.34, m
3.0	36.0, CH_2	2.44, d (12.7); 1.90, td (5.1, 13.3)	27.3, CH_2	1.94, m; 1.67, m
4.0	24.0, CH2	1.68, m; 1.40, t(7.3)	23.8, CH2	1.53, m; 1.46, m
5.0	45.0, CH	2.31, m	45.1, CH	2.17, m
6.0	31.6, CH	2.12, m	32.0, CH	2.08, m
7.0	44.6, CH_2	2.25, m	44.6, CH_2	2.25, m
8	199.2, C		198.8, C	
9.0	139.0, C		139.0, C	
10.0	157.0, C		158.4, C	
11.0	28.4, CH	3.26, m	28.2, CH	3.17, m
12.0	22.3, CH_3	1.40, d(7.3）	22.2, CH_3	1.40, d (6.9)
13.0	21.1, CH_3	1.40, d(7.3)	21.1, CH_3	1.40, d (6.9)
14.0	18.0, CH_3	0.82, d(6.8)	17.6, CH_3	0.80, d (6.8)
15.0	67.2, CH_2	3.83, m	63.9, CH_2	3.72, d (7.4)

Computational details

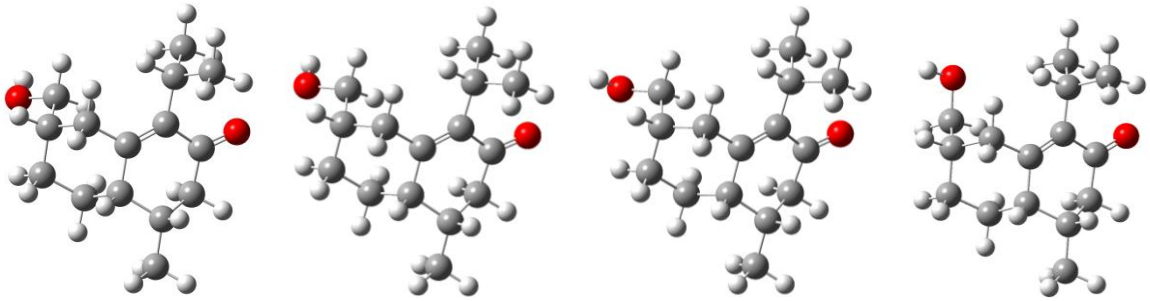
Molecular Merck force field (MMFF) and DFT/TD-DFT calculations were carried out with Spartan' 14 software (Wavefunction Inc., Irvine, CA, USA) and Gaussian 09 program, respectively (Bruhn et al. 2013). Conformers within 10 kcal/mol energy window were generated and optimized using DFT calculations at B3LYP/6-31G(d) level. Conformers with Bolzmann distribution over 1% were chosen for ECD calculations in methanol at B3lYP/6-311+g(2d,p) level. The IEF-PCM solvent model for MeOH was used. ECD spectra were generated using the program SpecDis 3.0 (University of Würzburg, Würzburg, Germany) and OriginPro 8.5 (OriginLab, Ltd., Northampton, MA, USA) from dipole-length rotational strengths by applying Gaussian band shapes with

sigma = 0.30 ev. All calculations were performed by High Performance Computing Platform in Sun Yat-sen University.

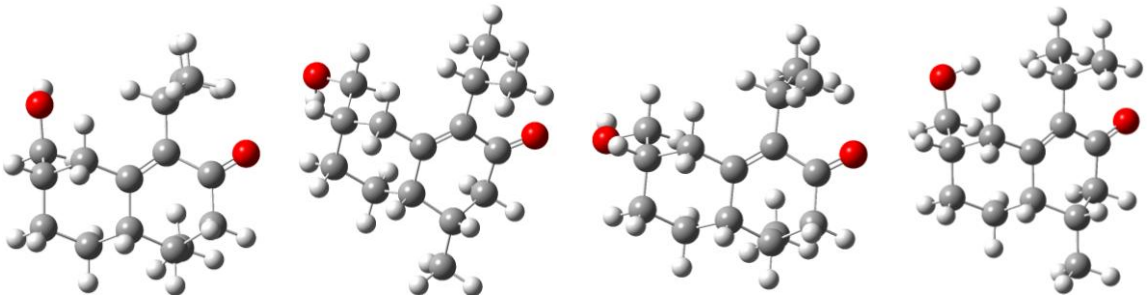
Table S2. Energy Analysis for the Conformers of (2*S*,5*S*,7*S*)-**2**.

compound	Conformation	G (Hartree)	G (Kcal/mol)	ΔG (Kcal/mol)	Boltzmann Dist (%)
(2*S*,5*S*,7*S*)-**2**	**2**-1	- 736.4911087	-462155.1445	0.613578759	0.102134797
(2*S*,5*S*,7*S*)-**2**	**2**-2	-736.4911087	-462155.1445	0.613578759	0.102134797
(2*S*,5*S*,7*S*)-**2**	**2**-3	-736.4916961	-462155.5132	0.244954596	0.190350772
(2*S*,5*S*,7*S*)-**2**	**2**-4	-736.4920865	-462155.7581	0	0.287889335
(2*S*,5*S*,7*S*)-**2**	**2**-5	-736.4890875	-462153.8762	1.881907173	0.011991451
(2*S*,5*S*,7*S*)-**2**	**2**-6	-736.4904237	-462154.7147	1.043410195	0.049419472
(2*S*,5*S*,7*S*)-**2**	**2**-7	-736.4885532	-462153.541	2.217154106	0.006807264
(2*S*,5*S*,7*S*)-**2**	**2**-8	-736.4918517	-462155.6108	0.147332948	0.224470266
(2*S*,5*S*,7*S*)-**2**	**2**-9	-736.4891228	-462153.8984	1.859737263	0.01244896
(2*S*,5*S*,7*S*)-**2**	**2**-10	-736.4891155	-462153.8938	1.864324357	0.012352888

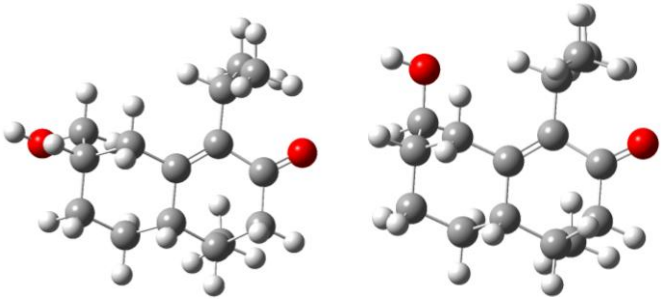
Fig. S23 B3LYP/6-31G(d) optimized low-energy conformers of **2.**



2-1 **2**-2 **2**-3 **2**-4



2-5 **2**-6 **2**-7 **2**-8



2-9 **2**-10

Reference:

Bruhn T, Schaumlöffel ANU, Hemberger Y, Bringmann G. 2013. SpecDis : Quantifying the Comparison of Calculated and Experimental Electronic Circular Dichroism Spectra. 249:243–249.