Populusene A, an Anti-inflammatory Diterpenoid with a Bicyclo[8,4,1]pentadecane Scaffold from *Populus euphratica* Resins

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Contents of Supporting Information

Table S1. ^{1}H (600 MHz) and ^{13}C NMR (150 MHz) Data of **1** (δ in ppm, J in Hz)

	1[a]			**1**[b]	
no.	δ_H	δ_C	**no.**	δ_H	δ_C
1		86.4, C	1		86.5, C
2	Ha: 2.22, dd (13.2, 4.0) Hb: 2.08, overlap	36.2, CH_2	2	Ha: 1.91, m Hb: 1.82, m	34.6, CH_2
3	2.64, dd (9.1, 4.0)	52.0, CH	3	2.30, overlap	50.5, CH
4		73.8, C	4		74.7, C
5	Ha: 2.10, m Hb: 2.05, m	41.1, CH_2	5	Ha: 1.84, overlap Hb: 1.84, overlap	40.2, CH_2
6	Ha: 2.83, m Hb: 1.48, m	24.6, CH_2	6	Ha: 2.30, overlap Hb: 1.22, m	23.5, CH_2
7	3.31, dd (8.2, 6.1)	60.5, CH	7	3.07, dd (9.2, 5.8)	60.0, CH
8		60.8, C	8		60.7, C
9	Ha: 2.08, overlap Hb: 1.07, brdd (12.4, 11.4)	39.0, CH_2	9	Ha: 2.10, m Hb: 0.97, m	38.1, CH_2
10	Ha: 2.33, q (11.4) Hb: 1.93, m	23.2, CH_2	10	Ha: 2.19, m Hb: 2.06, m	22.8, CH_2
11	5.03 dd (11.4, 5.5)	123.1, CH	11	5.08, ddd (11.5, 5.5, 1.6)	123.3, CH
12		137.1, C	12		135.6, C
13	Ha: 2.47, m Hb: 2.12, m	28.4, CH_2	13	Ha: 2.38, m Hb: 2.14, m	27.7, CH_2
14	Ha: 1.62, td (12.7, 6.0) Hb: 1.46, m	31.8, CH_2	14	Ha: 1.58, overlap Hb: 1.58, overlap	31.7, CH_2
15	1.98, m	37.4, CH	15	1.88, m	36.6, CH
16	1.01, d (6.9)	18.6, CH_3	16	0.96, d (6.8)	18.2, CH_3
17	0.92, d (6.9)	18.7, CH_3	17	0.95, d (6.8)	18.2, CH_3
18	1.43, s	26.0, CH_3	18	1.18, s	25.3, CH_3
19	1.36, s	18.4, CH_3	19	1.31, s	18.0, CH_3
20	4.91, s	76.5, CH	20	4.58, s	76.0, CH
4-OH	5.66, s				

[a] In pyridine-d_5; [b] In $CDCl_3$.

Table S2. HMBC and ROESY data of **1** in pyridine-d_5 (δ in ppm, J in Hz)

	1		
no.	**δ_H**	**HMBC (H→C)**	**ROESY**
2	Ha: 2.22, dd (13.2, 4.0)	C-1, C-3, C-4, C-14, C-15	H_3-17, H_3-18
	Hb: 2.08, overlap	C-3, C-4, C-14, C-20,	
3	2.64, dd (9.1, 4.0)	C-1, C-2, C-4, C-5, C-18	Ha-6, H-7, H-20, Ha-13
5	Ha: 2.10, m	C-3, C-4, C-7, C-6	
	Hb: 2.05, m	C-6, C-7	H-20, H_3-18
6	Ha: 2.83, m	C-4, C-5, C-7, C-8	4-OH, H-3
	Hb: 1.48, m	C-4, C-5, C-7, C-8	Ha-5, Ha-6
7	3.31, dd (8.2, 6.1)	C-6, C-8, C-9	Hb-9, H-3, H-20 (weak)
9	Ha: 2.08, overlap	C-7, C-8, C-11	
	Hb: 1.07, brdd (12.4, 11.4)	C-7, C-8, C-10, C-11, C-19	H-7, H-11
10	Ha: 2.33, q (11.4)	C-8, C-9, C-11, C-12	H-20, H_3-19
	Hb: 1.93, m	C-8, C-9, C-11, C-12	Ha-10
11	5.03 dd (11.4, 5.5)	C-9, C-11, C-13, C-20	Hb-9, Hb-13, Ha-10, Hb-10
13	Ha: 2.47, m	C-11, C-12, C-14	H-3, H-7 (weak), H-11 (weak)
	Hb: 2.12, m	C-1, C-11, C-12, C-14, C-20	H-11
14	Ha: 1.62, td (12.7, 6.0)	C-2, C-1, C-13	H_3-16, Ha-13, Hb-13, H-15
	Hb: 1.46, m	C-1, C-13	Ha-13
15	1.98, m	C-2, C-1, C-14, C-16, C-17	Ha-2, H_3-16, H_3-17
16	1.01, d (6.9)	C-17, C-15, C-1	Ha-14, Hb-14, H-15
17	0.92, d (6.9)	C-16, C-15, C-1	H_3-18
18	1.43, s	C-5, C-3, C-4	H-20, H_3-17
19	1.36, s	C-9, C-7, C-8	Ha-10, H-20 (weak)
20	4.91, s	C-13, C-2, C-3, C-4, C-1, C-11, C-12	H_3-18, Ha-11, H-3, H3-19 (weak), H-7 (weak)
4-OH	5.66, s	C-18, C-5, C-3, C-4	Ha-2, H-3, Ha-6

Table S3. ^{1}H (600 MHz) and ^{13}C NMR (150 MHz) Data of **2** (δ in ppm, J in Hz)

	2[a]			**2**[b]	
no.	δ_H	δ_C	**no.**	δ_H	δ_C
1		88.7, C	1		88.7, C
2	Ha: 2.27, dd (14.6, 10.5) Hb: 2.09, m	41.3, CH_2	2	Ha: 1.86, overlap Hb: 1.86, overlap	40.3, CH_2
3	4.23, m	70.1, CH	3	3.81, m	71.5, CH
4		80.2, C	4		79.1, C
5	Ha: 2.81, ddd (14.2, 5.4, 2.2) Hb: 1.90, td (14.2, 2.5)	37.2, CH_2	5	Ha: 2.36, overlap Hb: 1.70, m	37.0, CH_2
6	Ha: 2.70, ddd (16.0, 14.2, 2.2) Hb: 2.45, ddd (16.0, 5.5, 2.7)	37.3, CH_2	6	Ha: 2.40, m Hb: 2.36, overlap	36.1, CH_2
7		208.8, C	7		209.6, C
8		144.2, C	8		143.5, C
9	5.53, t (7.9)	124.5, CH	9	5.52, t (7.6)	124.2, CH
10	Ha: 2.54, m Hb: 1.72, ddd (12.5, 7.9, 1.5)	28.5, CH_2	10	Ha: 2.30, m Hb: 1.72, m	27.9, CH_2
11	3.60, brd (11.1)	81.6, CH	11	3.43, brd (11.1)	80.6, CH
12		86.4, C	12		86.1, C
13	Ha: 2.07, m Hb: 1.35, q (11.0)	33.8, CH_2	13	Ha: 2.07, m Hb: 1.40, m	33.1, CH_2
14	Ha: 2.36, q (11.0) Hb: 1.55, m	32.1, CH_2	14	Ha: 2.10, m Hb: 1.67, m	31.7, CH_2
15	1.57, m	40.5, CH	15	1.64, m	39.4, CH
16	0.90, d (6.7)	18.8, CH_3	16	0.88, overlap	18.3, CH_3
17	0.91, d (6.7)	18.9, CH_3	17	0.88, overlap	18.1, CH_3
18	1.52, s	18.6, CH_3	18	1.26, s	17.7, CH_3
19	1.85, s	20.3, CH_3	19	1.87, s	20.1, CH_3
20	1.09, s	24.9, CH_3	20	1.09, s	24.4, CH_3
3-OH	6.18, d (5.7)				

[a] In pyridine-d_5; [b] In $CDCl_3$.

Table S4. HMBC and ROESY data of **2** in pyridine-d_5 (δ in ppm, *J* in Hz)

	2		
no.	**δ_H**	**HMBC (H→C)**	**ROESY**
2	Ha: 2.27, dd (14.6, 10.5)	C-14, C-3, C-1, C-4, C-15	H_3-16, H_3-18, 3-OH (weak)
	Hb: 2.09, m	C-3, C-1, C-4, C-15	H_3-16, Hb-14
3	4.23, m	C-2, C-4, C-5, C-18	Ha-13, Ha-14
5	Ha: 2.81, ddd (14.2, 5.4, 2.2)	C-7, C-6, C-3, C-4	Hb-5, Ha-6, H_3-18 (weak)
	Hb: 1.90, td (14.2, 2.5)	C-18, C-6, C-7, C-3, C-4	Ha-5, H_3-18
6	Ha: 2.70, ddd (16.0, 14.2, 2.2)	C-4, C-5, C-7, C-8	H-3, Hb-5, Hb-6
	Hb: 2.45, ddd (16.0, 5.5, 2.7)	C-4, C-5, C-7	Ha-6, H_3-19
9	5.53, t (7.9)	C-7, C-10, C-11, C-19	H_3-19, Hb-10
10	Ha: 2.54, m	C-8, C-9, C-11, C-12	Hb-10, Ha-13
	Hb: 1.72, ddd (12.5, 7.9, 1.5)	C-8, C-9	Ha-10, H_3-20
11	3.60, brd (11.1)	C-9, C-4, C-12, C-13, C-10, C-20	H_3-18, Ha-10, H_3-20
13	Ha: 2.07, m	C-1, C-11, C-14	H-3, Ha-10, Hb-13, Hb-14
	Hb: 1.35, q (11.0)	C-11, C-12, C-14, C-20	Ha-13, Ha-14
14	Ha: 2.36, q (11.0)	C-1, C-2, C-13, C-15	H-3, Hb-14, Ha-13, Hb-13
	Hb: 1.55, m	C-12, C-15, C-13	H_3-17, Ha-14
15	1.57, m	C-16, C-17, C-14, C-2, C-1	H_3-16, H_3-17
16	0.90, d (6.7)	C-17, C-15, C-1	Ha-2, Hb-2
17	0.91, d (6.7)	C-16, C-15, C-1	Hb-14
18	1.52, s	C-3, C-4, C-5	Ha-2, Hb-5, H-11
19	1.85, s	C-7, C-8, C-9	Hb-6
20	1.09, s	C-11, C-12, C-13	Hb-10, H-11, Hb-13
3-OH	6.18, d (5.7)	C-2, C-3, C-4	Ha-2, Hb-2, Ha-5, H_3-18

Experimental Section

a. General experimental procedure

Column chromatography was performed by using silica gel (200-300 mesh; Qingdao Marine Chemical Inc., P.R. China), RP-18 silica gel (40–60 μm; Daiso Co., Japan), MCI gel CHP 20P (75-150 μm, Mitsubishi Chemical Industries, Tokyo, Japan), and Sephadex LH-20 (Amersham Pharmacia, Sweden). Optical rotations were measured on a Anton Paar MCP-100 digital polarimeter. UV spectra were obtained on a Shimadzu UV-2600 spectrometer (Shimadzu Corporation, Tokyo, Japan). CD spectra were measured on a Chirascan instrument (Agilent Technologies, Santa Clara, CA, USA). YMC-Pack ODS-A 250 mm × 9.4 mm, i.d., 5 μm, or a Phenomenex Kinetex (250 mm × 10 mm, i.d., 5 μm). NMR spectra were recorded on a Bruker AV-600 spectrometer, with TMS as an internal standard. HRESIMS were collected by a SCIEX X500R QTOF MS spectrometer.

b. Plant resins

The resins of *P. euphratica* were obtained from Bayin, Xinjiang Autonomous Region, November, 2011. A voucher specimen (CHYX0573), identified by Prof. Bin Qiu at Yunnan University of Traditional Chinese Medicine, is deposited at School of Pharmaceutical Sciences, Shenzhen University, P.R. China.

c. Extraction and isolation

The dried resins of the title plant (50 kg) was soaked with 95% EtOH (300 L × 3 × 24 h) to afford a crude extract, which was suspended in water and partitioned with EtOAc to afford an EtOAc soluble extract (12 kg). This extract was cut into eight fractions (Fr.A–Fr.H) by using a silica gel column with petroleum ether–acetone (50:1, 35:1, 20:1, 15:1, 10:1, 7:1, 3:1, 1:1) as solvents.

Fr. E (370 g) was separated via MCI gel CHP 20P eluted with aqueous MeOH (40%–100%) to provide seven portions (Fr.E.1–Fr.E.7). Fr.E.3 (57 g) was subjected to a RP-18 column eluted with aqueous MeOH (50%–100%) to provide three portions (Fr.E.3.1–Fr.E.3.3). Fr.E.3.3 (38 g) was separated via MPLC eluted with gradient aqueous MeOH (45%–55%) to yield eight fractions (Fr.E.3.3.1–Fr.E.3.3.8). Fr.E.3.3.3 (5.5 g) was separated via a RP-18 column eluted with aqueous MeOH (45%) to yield two portions Fr.E.3.3.3.1 and Fr.E.3.3.3.2. Fr.E.3.3.3.1 was separated via a RP-18 column eluted with gradient aqueous MeOH (30–58%) to yield eight fractions (Fr.E.3.3.3.1.1–Fr.E.3.3.3.1.8). Fr.E.3.3.3.1.6 (565 mg) was further separated via vacuum liquid chromatography on silica gel washed with CH_2Cl_2–acetone (1:0 and 50:1) to provide four portions (Fr.E.3.3.3.1.6.1–Fr.E.3.3.3.1.6.4). Fr.E.3.3.3.1.6.2 (71 mg) were subjected to preparative TLC (petroleum ether–EtOAc (3:1) to give Fr.E.3.3.3.1.6.2.1–Fr.E.3.3.3.1.6.2.5. Fr.E.3.3.3.1.6.2.2 (19 mg) was purified by semi-preparative HPLC on YMC-Pack ODS-A (aqueous MeCN, 49%) to yield **1** (3.57 mg, t_R = 19.24 min; flow rate: 3 mL/min). Fr.E.3.3.3.1.6.2.3 (27 mg) was purified by semi-preparative HPLC (aqueous MeCN, 59%) to yield **2** (5.68 mg, t_R = 13.12 min; flow rate: 3 mL/min) and Fr.E.3.3.3.1.6.2.3.1. Fr.E.3.3.3.1.6.2.3.1 (5.1 mg) was further purified by semi-preparative HPLC on Phenomenex Kinetex

(aqueous MeCN, 43%) to afford additional **1** (2.41 mg, t_R = 12.61 min, flow rate: 3 mL/min).

d. Physical constants and spectral data for 1 and 2

Compound 1: Colorless gums; UV (MeOH) λ_{max} (logε) 233 (2.72), 200 (3.80) nm; {$[\alpha]_D^{20}$ +24.3 (*c* 0.037, MeOH); CD (MeOH) $\Delta\varepsilon_{207}$ +1.61}; HRMS (ESI) *m*/*z*: $[M+H]^+$ 321.2423 calcd for $C_{20}H_{33}O_3$ 321.2420; ^1H and ^{13}C NMR data, see **Table 1**, **S5**, **6**, **S15**, and **16**.

Compound 2: Colorless crystals (MeOH); UV (MeOH) λ_{max} (logε) 273 (2.12), 220 (3.31), 206 (3.16) nm; {$[\alpha]_D^{20}$ –24.4 (*c* 0.041, MeOH); CD (MeOH) $\Delta\varepsilon_{208}$ +3.57}; HRMS (ESI) *m*/*z*: $[M+H]^+$ 337.2378 calcd for $C_{20}H_{33}O_4$ 337.2370; ^1H and ^{13}C NMR data, see **Tables S3**, **S30, S31, S39**, and **S40**.

e. Biological assays

Materials and Methods

1. Cell culture

RAW264.7, a mouse macrophage line (Procell Life Science & Technology Co., Wuhan, PR China), was cultured in high-glucose DMEM (C11995500BT, Gibco) supplemented with 10% fetal bovine serum (FBS) (2094468CP, Gibco), 100 U/mL penicillin and 100 μg/mL streptomycin at 37 °C in a humidified environment containing 5% CO_2.

2. Cell viability assay

Cell viability was assessed according to previous study briefly.[1] RAW264.7 cells (2× 10^4 cells/mL) were seeded into 96-well plates with completed DMEM. After overnight culture, cells were treated with various concentrations of compounds or DMSO for 24 h. Then Cell Count Kit-8 (CCK-8, Beyotime, Shanghai, PR China) was added into each well for 1 h at 37 °C. The absorbance of each well was recorded at 450 nm using a microplate reader (BioTek, USA).

3. ELISA of TNF-α and IL-6

The culture supernatants were collected and centrifuged from treated cells. The concentrations of TNF-α and IL-6 were measured using the ELISA Kit (Proteintech, USA) according to the manufacturer's instructions.

4. Western bloting

After lipopolysaccharide (LPS) treatment, total protein was extracted from the cell line using radioimmunoprecipitation assay (RIPA) buffer (Beyotime, PR China) containing protease cocktail (Roche, Germany) and quantified protein samples using the BCA assay (Thermo Scientific, USA). Equal amounts of protein extracts were separated by 10% SDS-PAGE and transferred to PVDF membranes. The membranes were blocked with 5% BSA, then with the indicated antibodies overnight at 4 °C, and followed the incubation with horseradish peroxidase (HRP)-conjugated secondary antibody at room temperature. The bands were visualized and measured via the ECL kit (Pierce, USA) and b analysis system (Bio-Rad, CA, USA). The densitometry analysis of the immunoblot results was performed using ImageJ software (NIH, USA).The system was used as previously described.[2]

5. Statistical analysis

All experimental data obtained from this study were performed in triplicate. The results represent mean ± SEM. Statistical analyses were performed by Graphpad prism 6 (GraphPad Software, San Diego, CA, USA) and Excel (Microsoft) with Student's *t*-test, one-way ANOVA test. Differences were considered significant when $*P \leq 0.05$, $**P \leq 0.01$, $***P \leq 0.001$, and $****P \leq 0.0001$.

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f. Crystal preparation and measurement for 2

Crystals of compound **2** were collected from methanol at room temperature. A suitable crystal was selected and placed on a XtaLAB AFC12 (RINC): Kappa single diffractometer. The crystal was kept at 99.98(10) K during data collection.

Using Olex2[1], the structure was solved with the ShelXT[2] structure solution program using Intrinsic Phasing and refined with the ShelXL[3] refinement package using Least Squares minimisation.

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(2) Sheldrick, G. M., *Acta Cryst.*, **2015**, *A71*, 3–8.

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g. X-ray crystallographic data of 2

Crystal Data for **2**: $C_{20}H_{32}O_4$ (M =336.45 g/mol): orthorhombic, space group $P2_12_12_1$ (no. 19), a = 6.43210(10) Å, b = 8.24480(10) Å, c = 34.7206(3) Å, V = 1841.28(4) Å^3, Z = 4, T = 99.98(10) K, μ(CuKα) = 0.659 mm^{-1}, *Dcalc* = 1.214 g/cm^3, 17952 reflections measured (10.19° ≤ 2Θ ≤ 148.672°), 3680 unique (R_{int} = 0.0296, R_{sigma} = 0.0217) which were used in all calculations. The final R_1 was 0.0314 (I > 2σ(I)) and wR_2 was 0.0780 (all data). The goodness of fit on F^2 was 1.036.

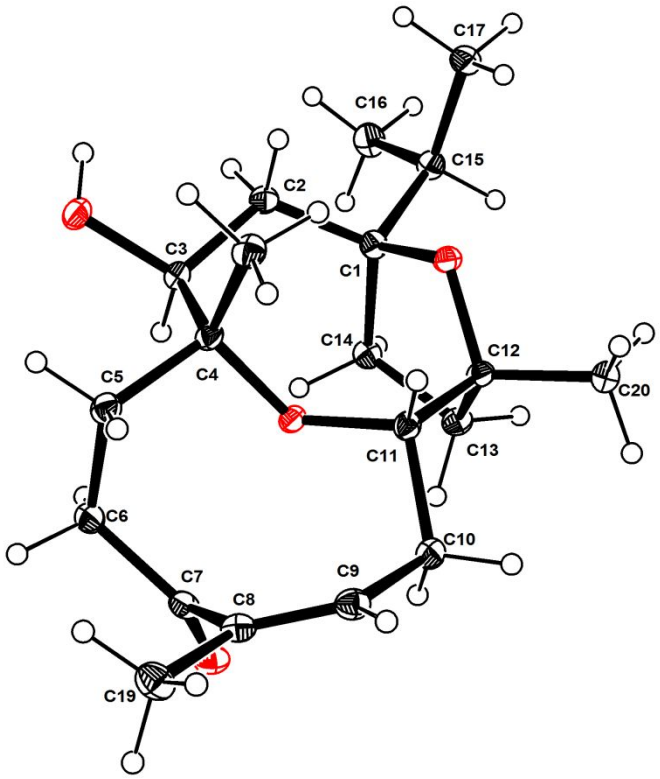

Figure S1. Plot of X-ray crystallographic data for **2**. Displacement ellipsoids are drawn at the 50% probability level.

h. ECD computational methods

Molecular Merck force field (MMFF) and DFT/TDDFT calculations were performed with Spartan'14 software package (Wavefunction Inc., Irvine, CA, USA) and Gaussian09 program package.[1] MMFF conformational search generated low-energy conformers within a 10 kcal/mol energy window were subjected to geometry optimization using DFT method at the B3LYP/6-311g (d,p) level. ECD calculations further were conducted at the B3LYP/6-311g (d,p) level with the PCM in MeOH. For comparisions of the calculated curves and experimental CD spectra, the program SpecDis 1.62[2] was used.

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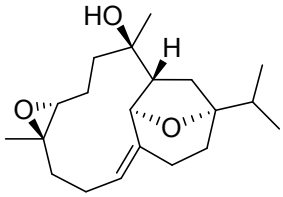
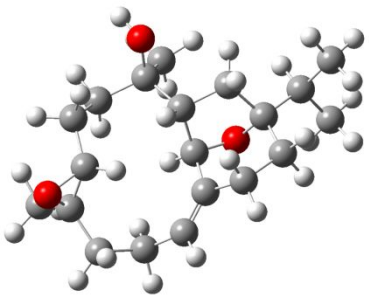
ECD calculations for compound 1.

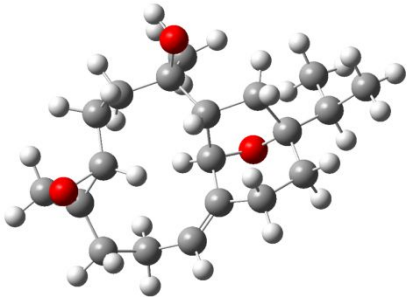


1

Figure S2. Model structures of **1** used for ECD calculations.



1a



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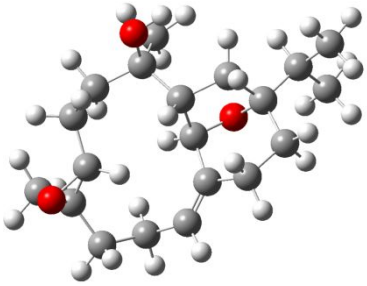
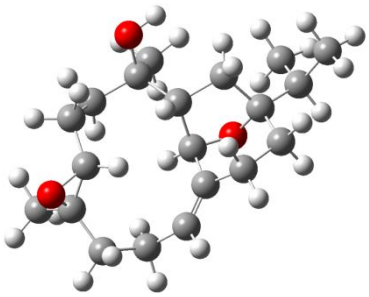


1c



1d

Figure S3. Optimized geometries of predominant conformers for compound **1** at the B3LYP/6-311g(d,p) level.

Table S5. Extracted heats and weighting factors of the optimized conformers of **1** at B3LYP/6-31G(d,p) level

	Conformer	Extracted heats	Population (%)
1	**1a**	-1006.85295	54.65%
	1b	-1006.85132	9.81%
	1c	-1006.85239	30.38%
	1d	-1006.85072	5.16%

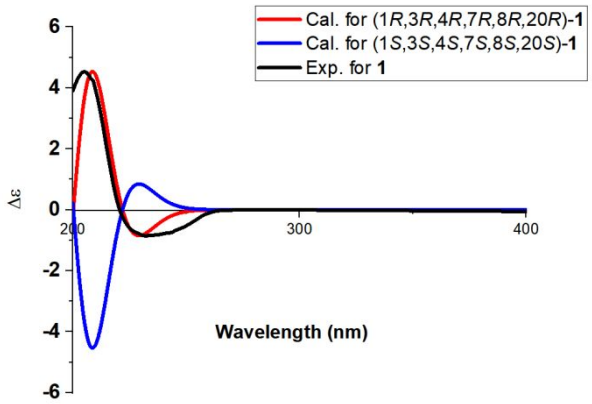


Figure S4. Comparison of B3LYP/6-311g (d,p) calculated ECD spectrum for (1*R*,3*R*,4*R*,7*R*,8*R*,20*R*)-**1**] with the experimental spectra of **1** in MeOH. σ= 0.4 eV; shift = 15 nm; scaling factor = 4.99.

Table S6. The Cartesian coordinates of the lowest energy conformers for **1a**, **1b**, **1c** and **1d**.

1a	X axis(Å)	Y axis(Å)	Z axis(Å)	**1b**	X axis(Å)	Y axis(Å)	Z axis(Å)
C	-3.9642	0.0687	0.4725	C	-4.0174	-0.4404	0.4871
C	-4.5236	-0.9804	1.4421	C	-5.0661	-0.2915	-0.6196
C	-5.0022	0.3673	-0.6131	C	-4.1628	0.7197	1.4806
C	-2.5749	-0.3114	-0.1018	C	-2.5736	-0.6114	-0.0589
C	-1.932	0.8383	-0.9013	C	-2.0418	0.5624	-0.8974
C	-0.4394	0.8058	-0.5146	C	-0.5423	0.6571	-0.5398
C	-0.3426	-0.4167	0.4082	C	-0.3453	-0.4863	0.4685
C	-0.1098	-1.7107	-0.352	C	0.0171	-1.7986	-0.2037
C	-1.1673	-2.011	-1.3946	C	-0.9868	-2.248	-1.2437
C	-2.5708	-1.6364	-0.901	C	-2.4265	-1.9643	-0.8036
O	-1.6525	-0.5108	0.9936	O	-1.6479	-0.678	1.0489
C	0.9241	-2.5568	-0.1737	C	1.1151	-2.5411	0.0393
C	0.0653	2.1272	0.1278	C	-0.1016	2.0547	-0.0236
C	-0.7174	2.5301	1.3928	C	-0.879	2.5227	1.2201
C	1.5759	2.1184	0.4882	C	1.4133	2.163	0.3002
O	-0.1354	3.1607	-0.8513	O	-0.3816	2.988	-1.0808
C	2.5656	1.8012	-0.6428	C	2.3989	1.8201	-0.8267
C	2.8377	0.3366	-0.8142	C	2.7846	0.3725	-0.864
C	3.5651	-0.5254	0.213	C	3.5979	-0.3211	0.2252
C	3.3726	-2.024	0.1725	C	3.5167	-1.8252	0.346
C	2.052	-2.4617	0.819	C	2.2212	-2.2855	1.0275
C	4.1151	0.0385	1.4909	C	4.129	0.4112	1.4231
O	4.2284	0.0022	-0.9472	O	4.1938	0.1319	-1.0013

H	-3.8328	0.9832	1.0679	H	-4.2614	-1.3468	1.0588
H	-3.8085	-1.2108	2.238	H	-5.0961	-1.1718	-1.2675
H	-5.4364	-0.608	1.9205	H	-6.0671	-0.1771	-0.1889
H	-4.7807	-1.9125	0.9295	H	-4.8709	0.5864	-1.2438
H	-4.6533	1.1431	-1.3011	H	-3.4216	0.6596	2.2832
H	-5.934	0.7264	-0.1623	H	-5.1524	0.6935	1.9511
H	-5.2434	-0.5252	-1.1989	H	-4.0657	1.6917	0.989
H	-2.3912	1.8024	-0.6645	H	-2.5681	1.4942	-0.681
H	-2.072	0.7045	-1.9806	H	-2.1871	0.3859	-1.9698
H	0.1284	0.6597	-1.4405	H	0.0183	0.4734	-1.4642
H	0.3771	-0.2879	1.2164	H	0.347	-0.2366	1.2725
H	-1.1613	-3.0687	-1.6844	H	-0.8861	-3.3163	-1.4699
H	-0.9358	-1.4413	-2.303	H	-0.783	-1.7145	-2.1805
H	-2.9146	-2.4544	-0.2561	H	-2.7339	-2.7747	-0.1287
H	-3.2505	-1.5871	-1.7584	H	-3.0774	-2.0104	-1.683
H	0.9726	-3.4444	-0.806	H	1.2479	-3.4587	-0.5357
H	-0.3484	3.4853	1.786	H	-0.5125	3.4993	1.5595
H	-1.7795	2.6925	1.1886	H	-1.9389	2.6771	1.006
H	-0.6236	1.7841	2.1876	H	-0.7816	1.8227	2.0549
H	1.7508	1.4607	1.3451	H	1.6426	1.5856	1.201
H	1.8314	3.1318	0.8305	H	1.6136	3.2133	0.5573
H	2.2149	2.2197	-1.5928	H	1.9936	2.1149	-1.8011
H	3.5093	2.3168	-0.4227	H	3.3022	2.4273	-0.6859
H	2.1727	-0.1324	-1.5265	H	2.1433	-0.2137	-1.5078
H	4.2023	-2.4926	0.7168	H	4.3697	-2.1712	0.9434
H	3.4406	-2.3967	-0.8579	H	3.6263	-2.3003	-0.6376
H	2.1877	-3.4699	1.2318	H	2.4159	-3.2406	1.5323
H	1.7956	-1.827	1.672	H	1.918	-1.5897	1.815
H	4.3579	1.1023	1.4097	H	4.2786	1.4779	1.231
H	3.3946	-0.0827	2.3055	H	3.4414	0.3126	2.2686
H	5.036	-0.4829	1.7727	H	5.0975	-0.0022	1.724
H	0.0993	4.0139	-0.4465	H	-0.1898	3.8846	-0.7547
1c	X axis(Å)	Y axis(Å)	Z axis(Å)	**1d**	X axis(Å)	Y axis(Å)	Z axis(Å)
C	-3.9691	0.0562	0.4761	C	-4.0179	-0.4497	0.4905
C	-4.5219	-1.0014	1.4402	C	-5.069	-0.301	-0.6139
C	-5.0091	0.3546	-0.6077	C	-4.1661	0.7064	1.4886
C	-2.5774	-0.3126	-0.1006	C	-2.5742	-0.6134	-0.0586
C	-1.941	0.8466	-0.8919	C	-2.0487	0.5664	-0.893
C	-0.4456	0.8173	-0.5129	C	-0.5473	0.6657	-0.5418
C	-0.3436	-0.4085	0.4076	C	-0.3448	-0.4797	0.4655
C	-0.1085	-1.7005	-0.3555	C	0.0185	-1.7911	-0.2079
C	-1.1621	-1.9986	-1.402	C	-0.983	-2.2403	-1.2497
C	-2.5667	-1.633	-0.9067	C	-2.4229	-1.9634	-0.8071

O	-1.6536	-0.507	0.9929	O	-1.6471	-0.6742	1.0467
C	0.9237	-2.5481	-0.1754	C	1.1152	-2.5347	0.0374
C	0.0696	2.1352	0.1313	C	-0.0993	2.0618	-0.0237
C	-0.702	2.5441	1.4012	C	-0.8686	2.5359	1.223
C	1.5801	2.1123	0.4927	C	1.4156	2.1587	0.3047
O	-0.0877	3.1861	-0.837	O	-0.3369	3.0155	-1.0729
C	2.5697	1.7973	-0.6394	C	2.4034	1.8191	-0.8216
C	2.839	0.3324	-0.8131	C	2.7873	0.371	-0.8617
C	3.5705	-0.5291	0.2115	C	3.6026	-0.3228	0.2256
C	3.3732	-2.0269	0.1765	C	3.5179	-1.8261	0.3498
C	2.049	-2.4566	0.8208	C	2.2191	-2.2811	1.0286
C	4.1284	0.0371	1.4849	C	4.1384	0.411	1.4206
O	4.2279	-0.0062	-0.9542	O	4.1954	0.1264	-1.0038
H	-3.843	0.9682	1.0765	H	-4.2575	-1.359	1.0594
H	-3.8048	-1.2325	2.234	H	-5.0963	-1.1789	-1.2652
H	-5.4362	-0.6365	1.9215	H	-6.0697	-0.1925	-0.1812
H	-4.7747	-1.9318	0.9224	H	-4.8785	0.5801	-1.235
H	-4.6647	1.1361	-1.2916	H	-3.4242	0.6453	2.2906
H	-5.9428	0.706	-0.1546	H	-5.1552	0.6754	1.9596
H	-5.2454	-0.5361	-1.1981	H	-4.072	1.6807	1.0011
H	-2.4063	1.8031	-0.6395	H	-2.5802	1.4919	-0.6654
H	-2.0859	0.7243	-1.9718	H	-2.1986	0.3968	-1.9658
H	0.1183	0.6728	-1.4415	H	0.0105	0.4834	-1.4682
H	0.3752	-0.2802	1.2166	H	0.3468	-0.2299	1.2698
H	-1.1514	-3.0542	-1.6989	H	-0.8787	-3.3073	-1.4802
H	-0.932	-1.4218	-2.3064	H	-0.7817	-1.7024	-2.1845
H	-2.9069	-2.4561	-0.2663	H	-2.7263	-2.7767	-0.134
H	-3.2466	-1.5824	-1.7638	H	-3.0748	-2.0094	-1.6857
H	0.9739	-3.4345	-0.8093	H	1.2492	-3.4521	-0.5378
H	-0.3385	3.5084	1.7765	H	-0.5134	3.5236	1.5413
H	-1.769	2.6905	1.2108	H	-1.9345	2.668	1.0241
H	-0.5914	1.8068	2.2021	H	-0.7501	1.848	2.0653
H	1.7518	1.45	1.3467	H	1.6415	1.577	1.2036
H	1.8405	3.1232	0.8392	H	1.619	3.2076	0.5662
H	2.2205	2.2175	-1.5892	H	2.0012	2.1162	-1.7965
H	3.5142	2.3109	-0.4182	H	3.3071	2.4251	-0.6773
H	2.1687	-0.135	-1.521	H	2.1426	-0.2133	-1.5033
H	4.1994	-2.4964	0.7253	H	4.3681	-2.1728	0.9506
H	3.4428	-2.4041	-0.8521	H	3.6291	-2.3039	-0.6323
H	2.1784	-3.4645	1.2363	H	2.4093	-3.2361	1.5354
H	1.7937	-1.8182	1.6715	H	1.9162	-1.583	1.8141
H	4.3728	1.1001	1.4002	H	4.2902	1.4769	1.2263
H	3.4121	-0.0811	2.3036	H	3.4527	0.3155	2.268

H	5.0499	-0.4854	1.7629	H	5.1067	-0.0042	1.72
H	-1.0193	3.4635	-0.8482	H	-1.2893	3.2066	-1.1113

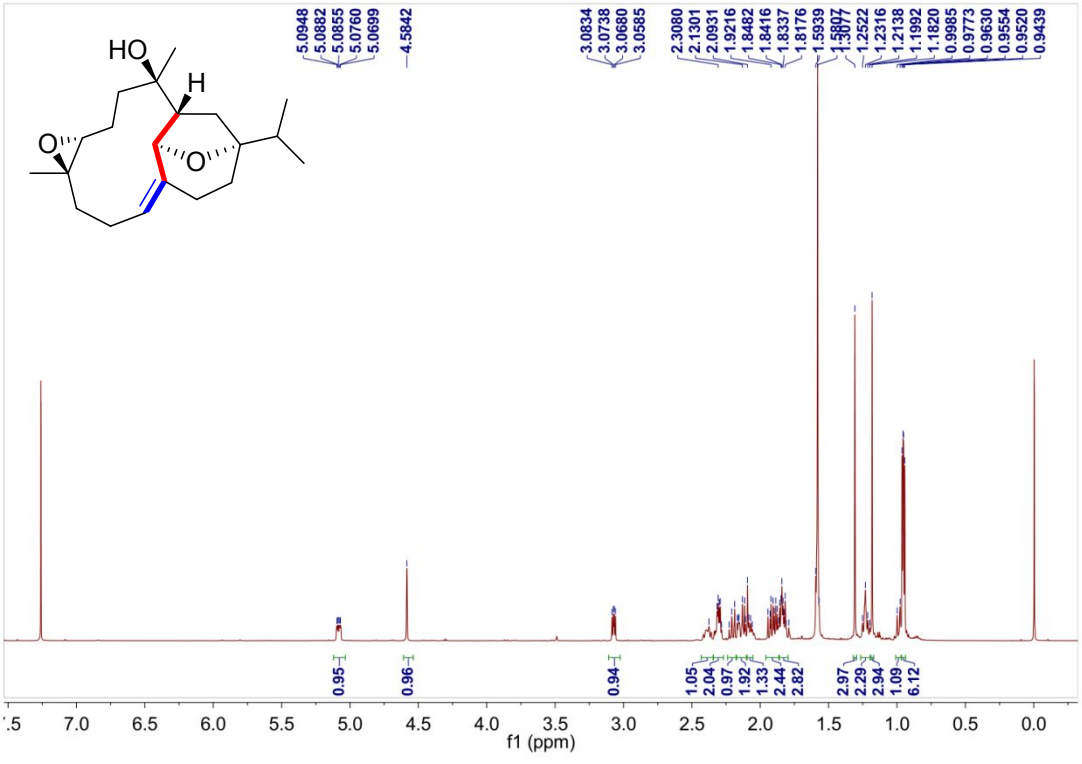


Figure S5. [1]H NMR (600 MHz) spectrum of **1** in $CDCl_3$.

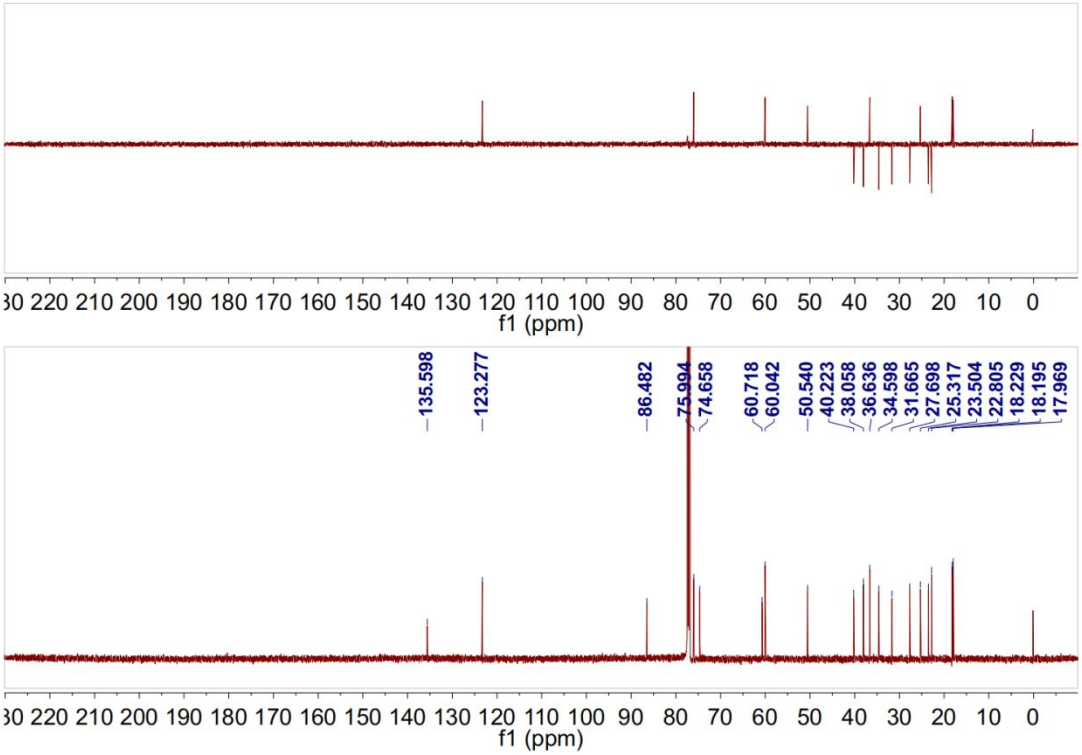


Figure S6. [13]C NMR and DEPT (150 MHz) spectra of **1** in $CDCl_3$.

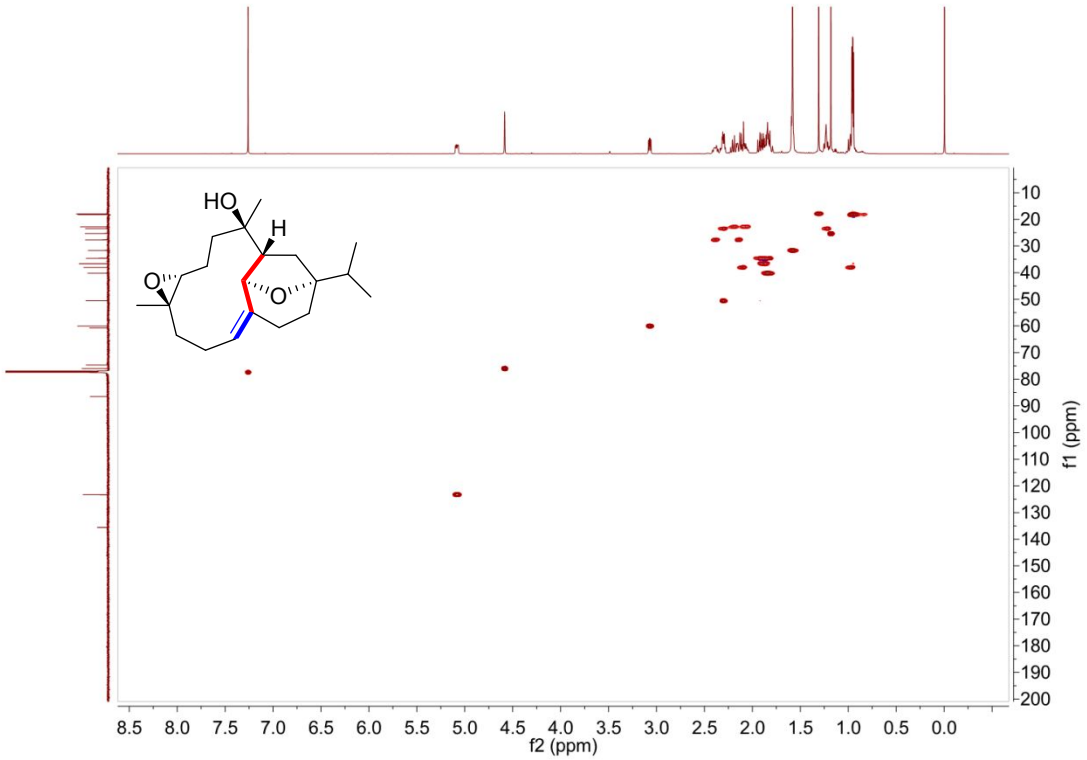


Figure S7. HSQC (600 MHz) spectrum of **1** in $CDCl_3$.

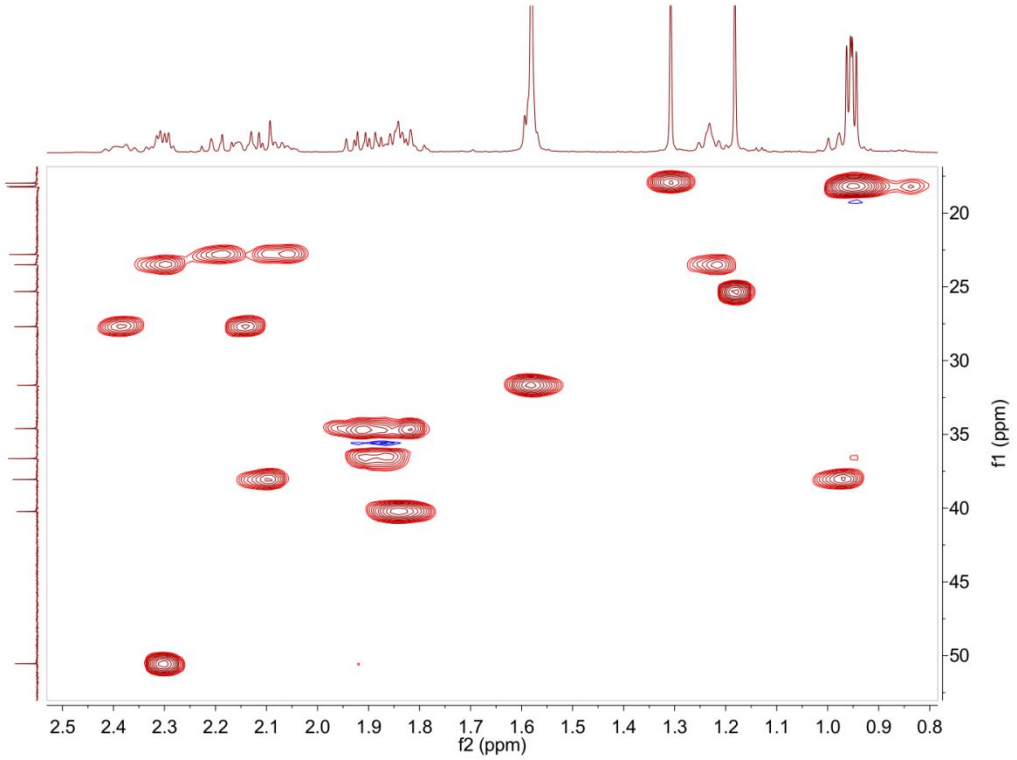


Figure S8. Enlarged HSQC (600 MHz) spectrum of **1** in $CDCl_3$.

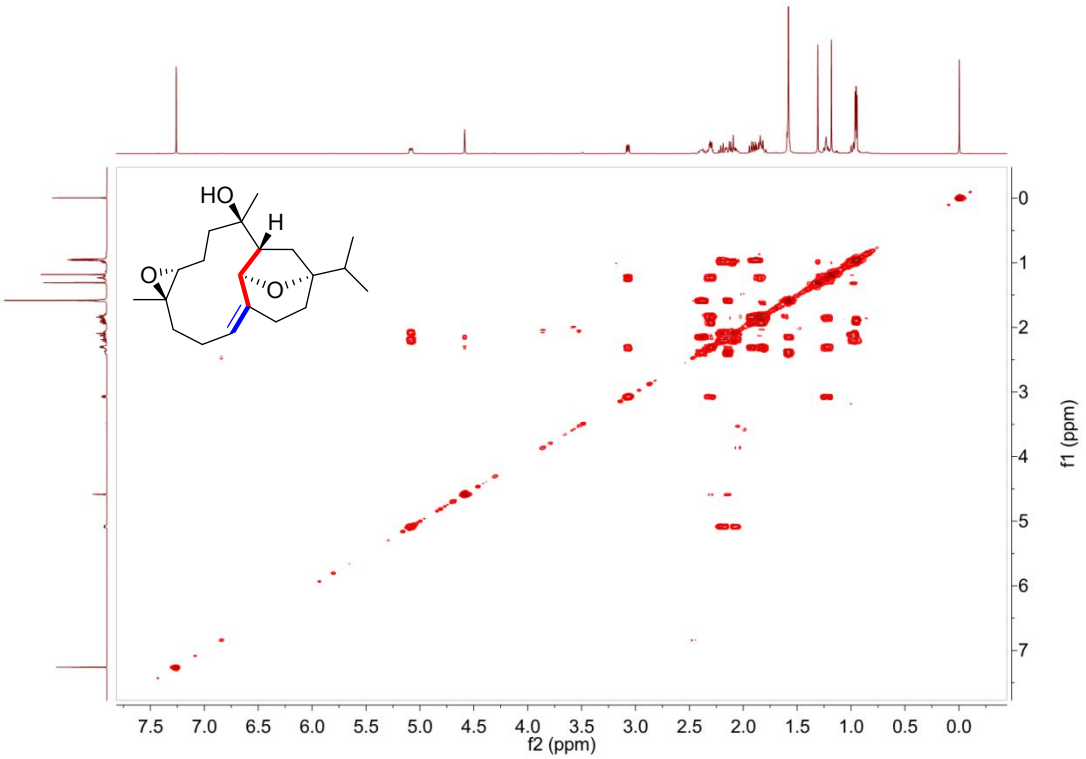


Figure S9. ^{1}H-^{1}H COSY (600 MHz) spectrum of **1** in $CDCl_3$.

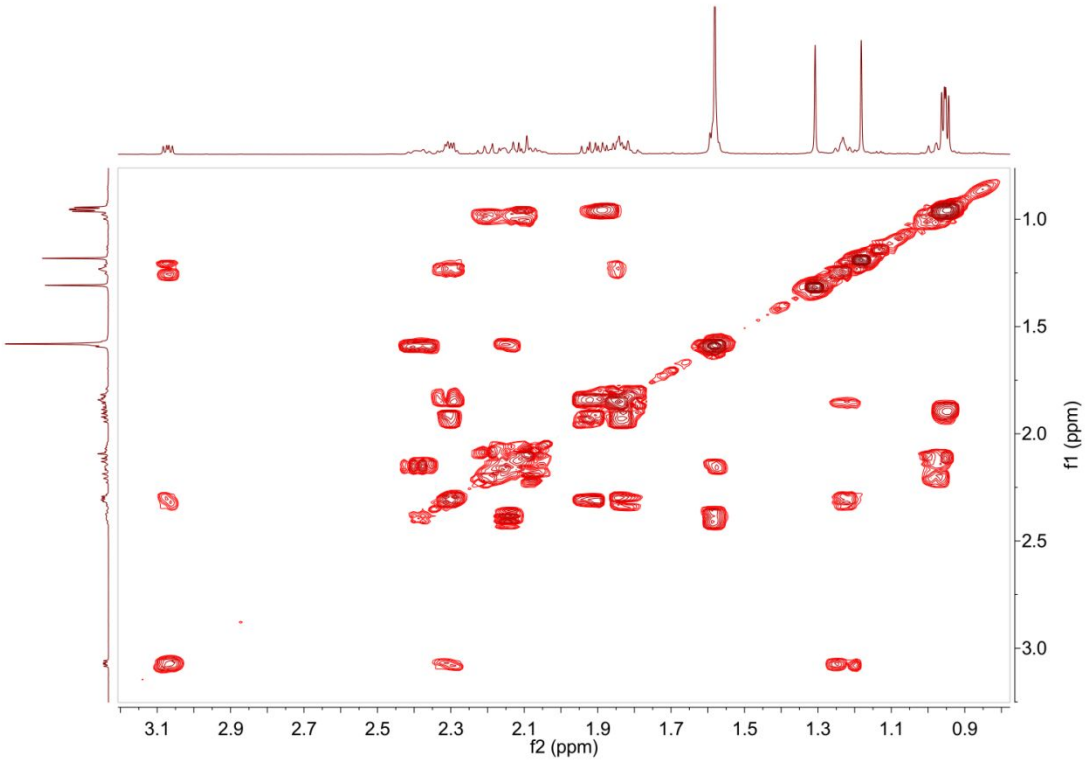


Figure S10. Enlarged ^{1}H-^{1}H COSY (600 MHz) spectrum of **1** in $CDCl_3$.

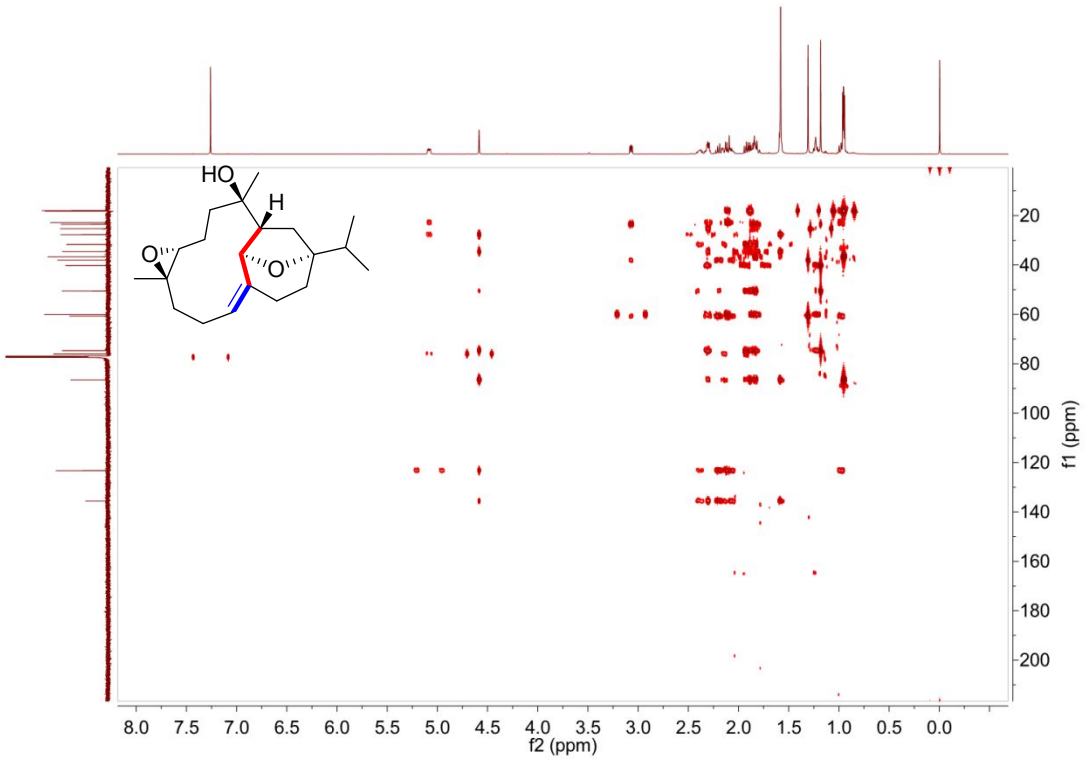


Figure S11. HMBC (600 MHz) spectrum of **1** in $CDCl_3$.

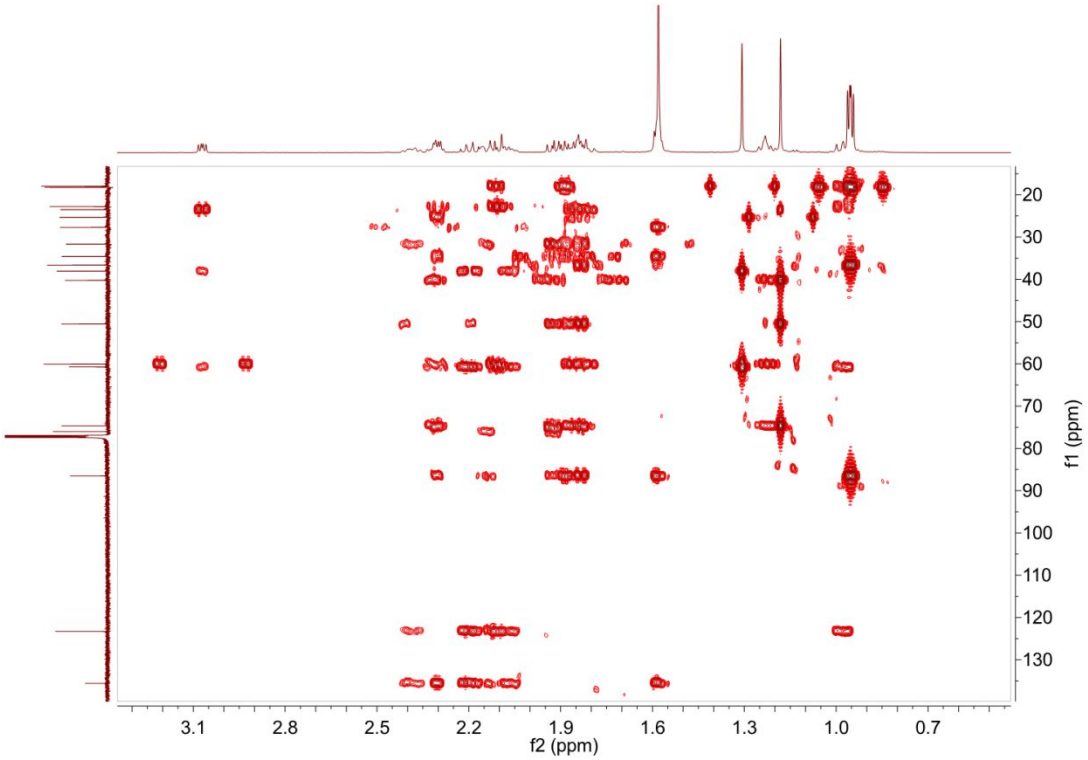


Figure S12. Enlarged HMBC (600 MHz) spectrum of **1** in $CDCl_3$.

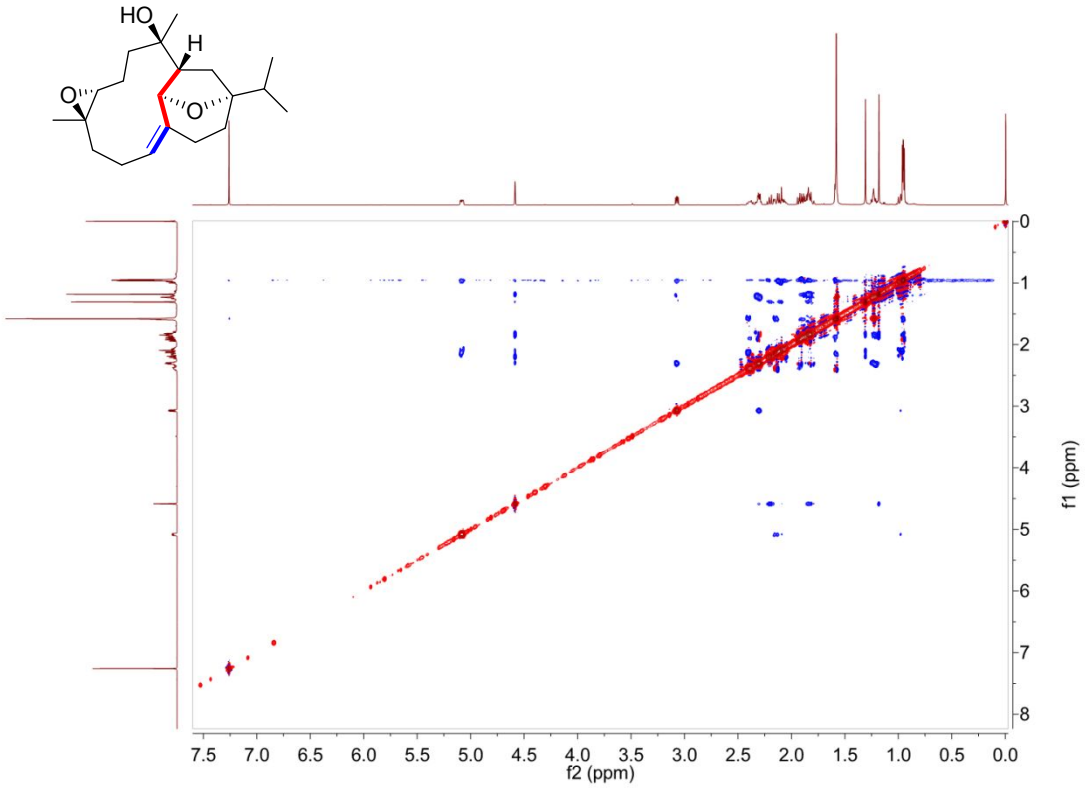


Figure S13. ROESY (600 MHz) spectrum of **1** in $CDCl_3$.

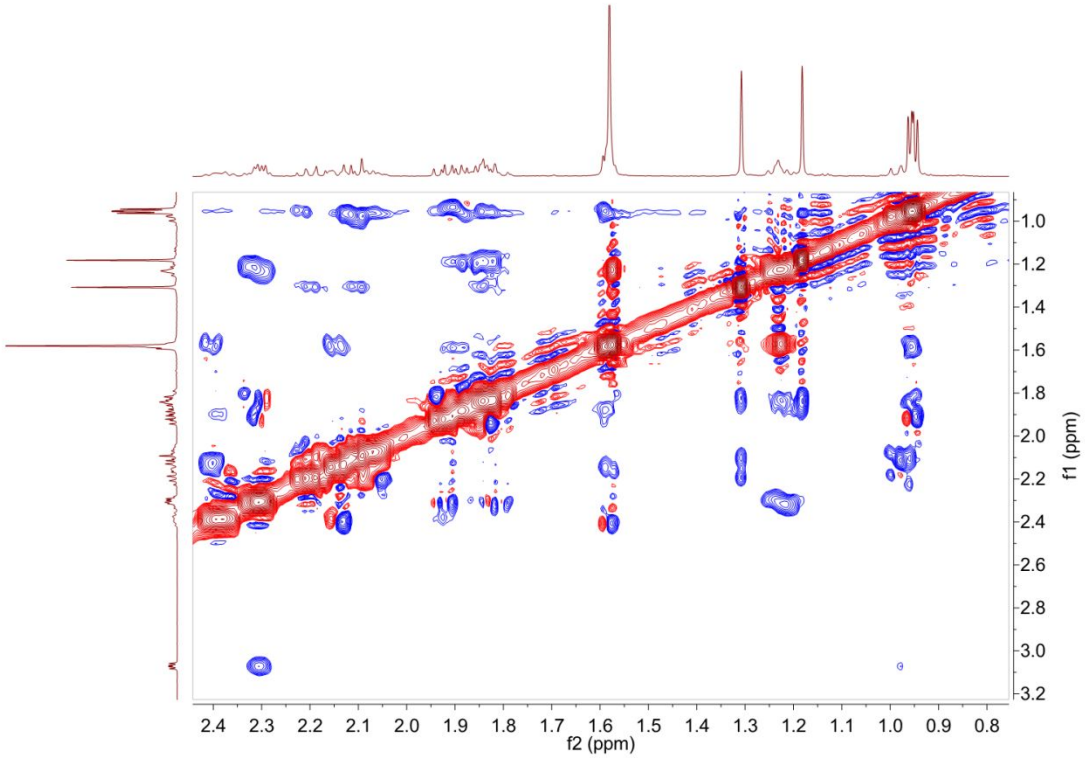


Figure S14. Enlarged ROESY (600 MHz) spectrum of **1** in $CDCl_3$.

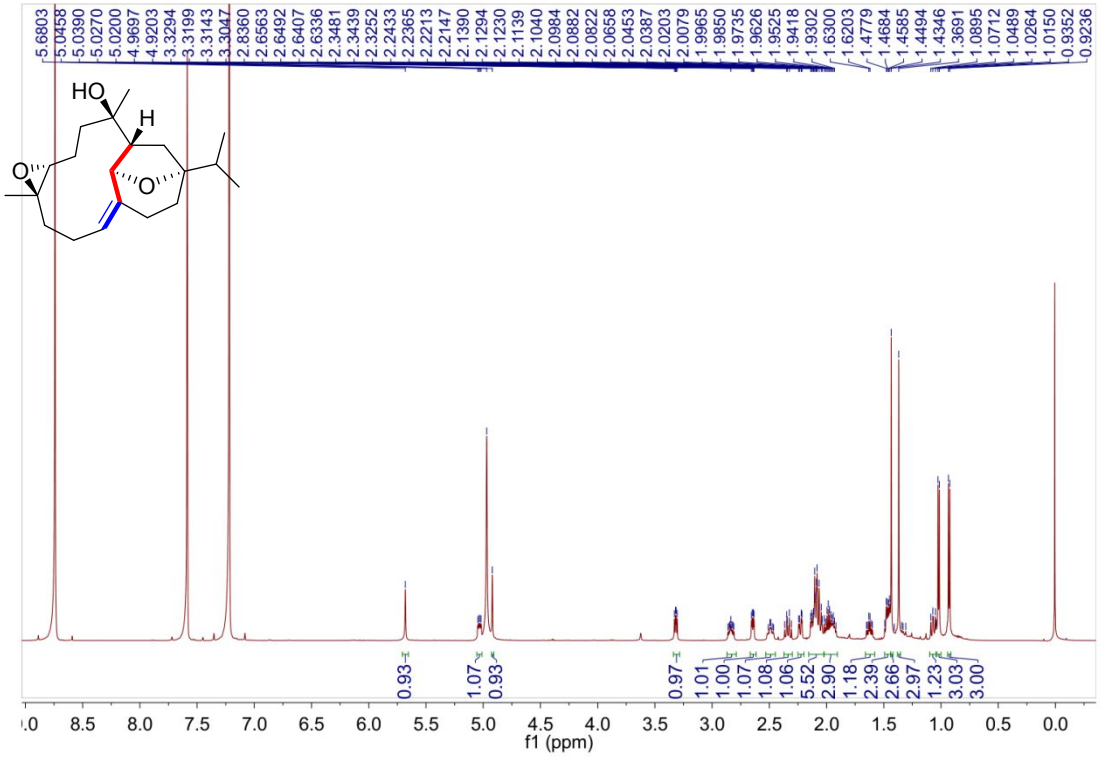


Figure S15. ^{1}H NMR (600 MHz) spectrum of **1** in pyridine-d_5.

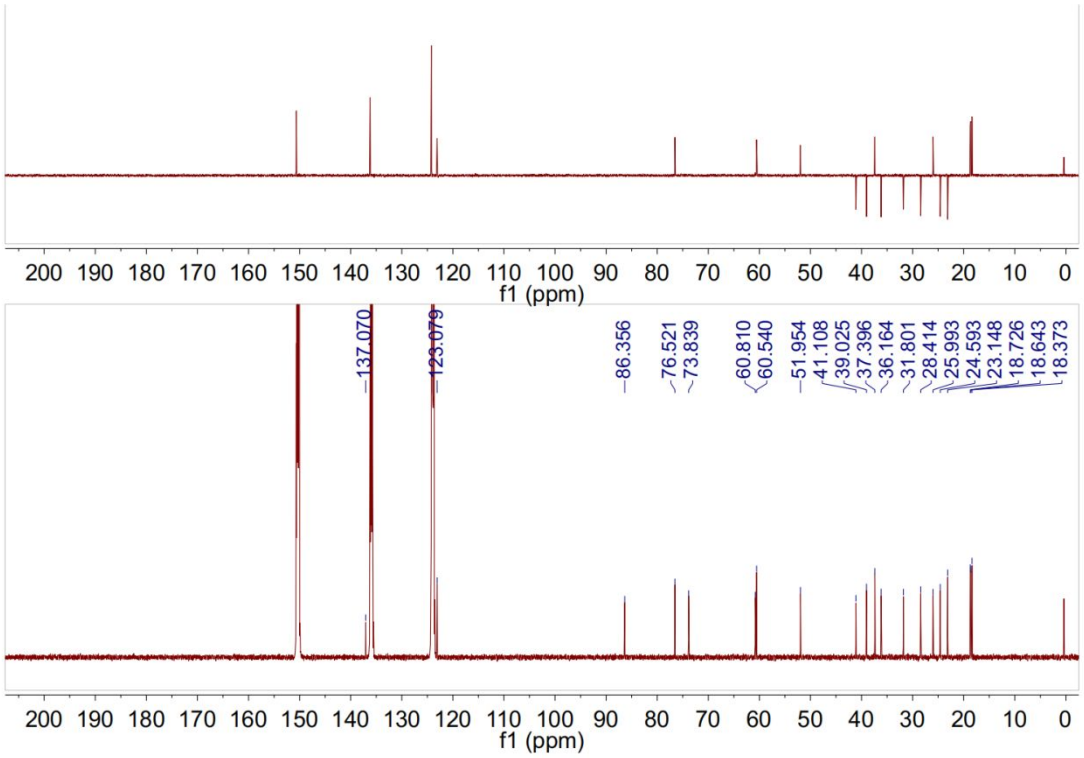


Figure S16. ^{13}C NMR and DEPT (150 MHz) spectra of **1** in pyridine-d_5.

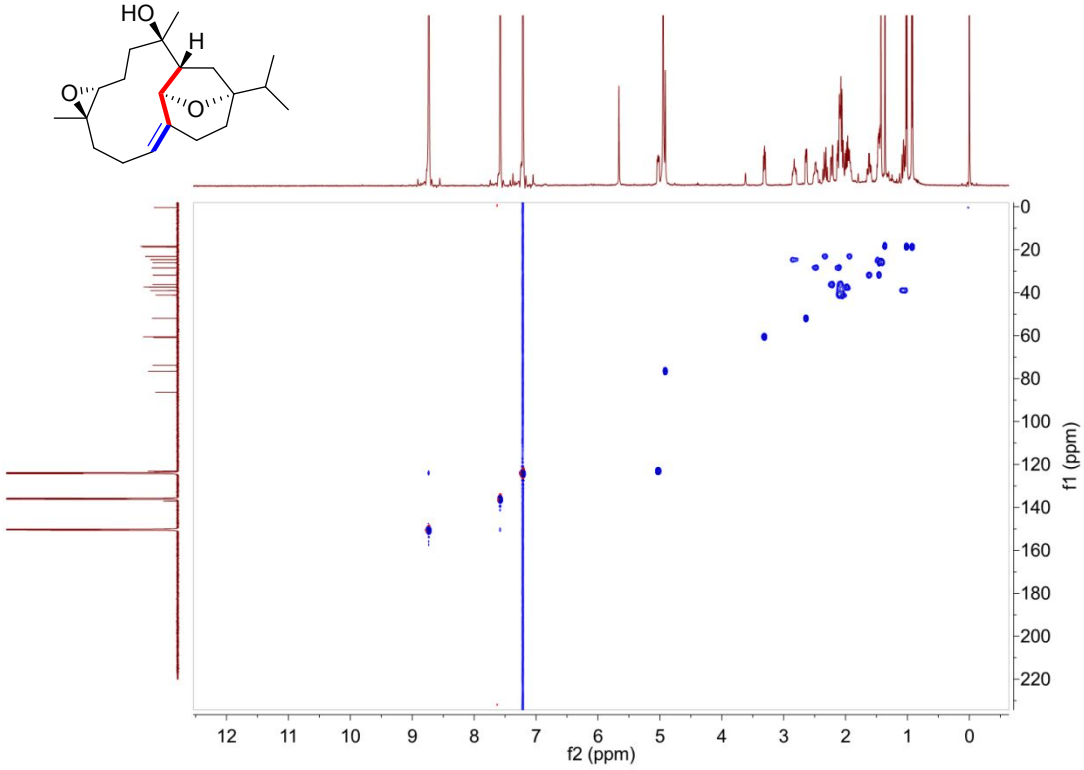


Figure S17. HSQC (600 MHz) spectrum of **1** in pyridine-d_5.

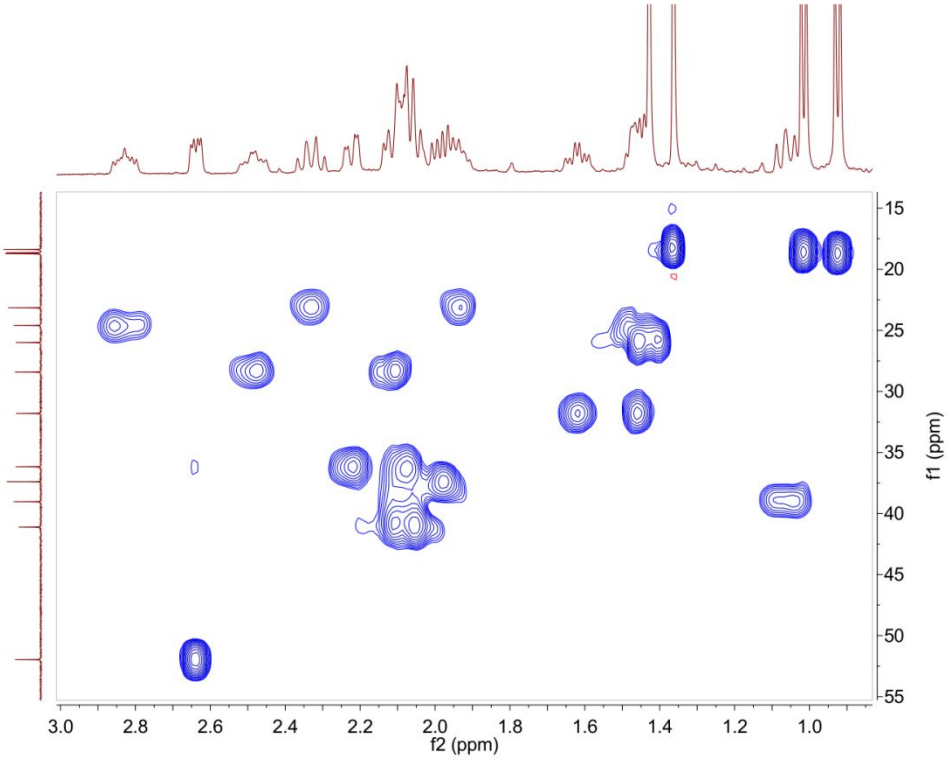


Figure S18. Enlarged HSQC (600 MHz) spectrum of **1** in pyridine-d_5.

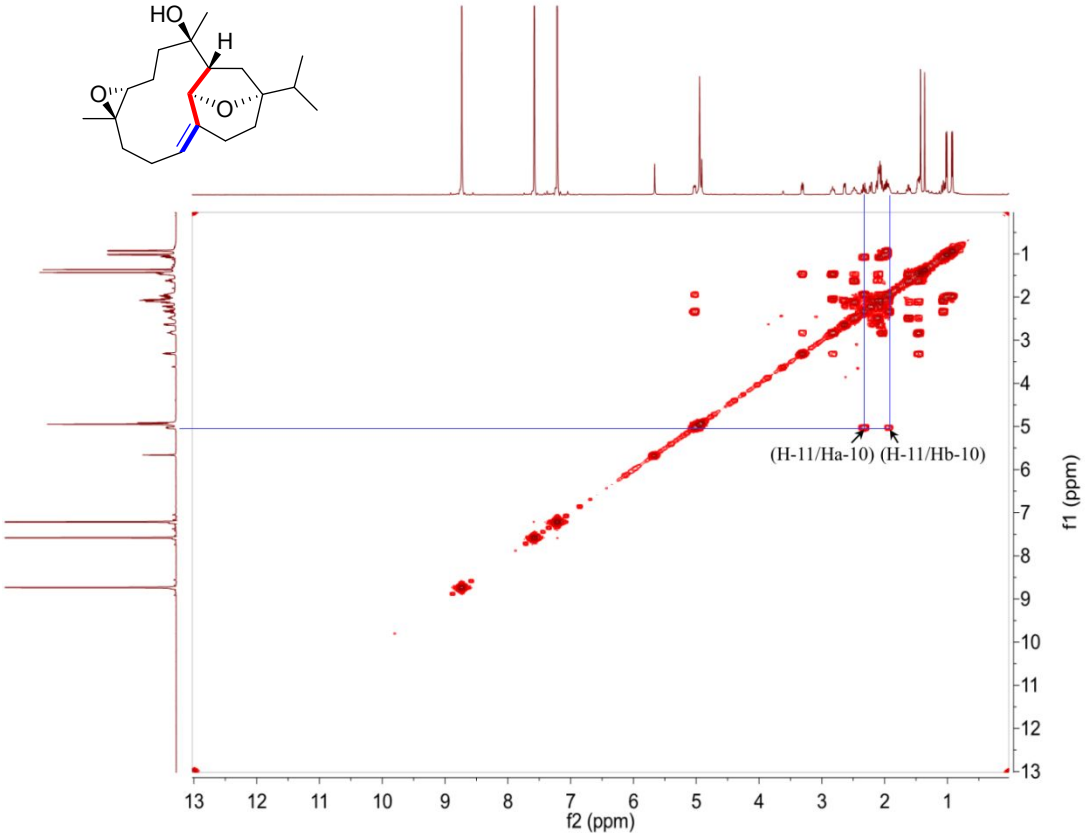


Figure S19. ^{1}H-^{1}H COSY (600 MHz) spectrum of **1** in pyridine-d_5.

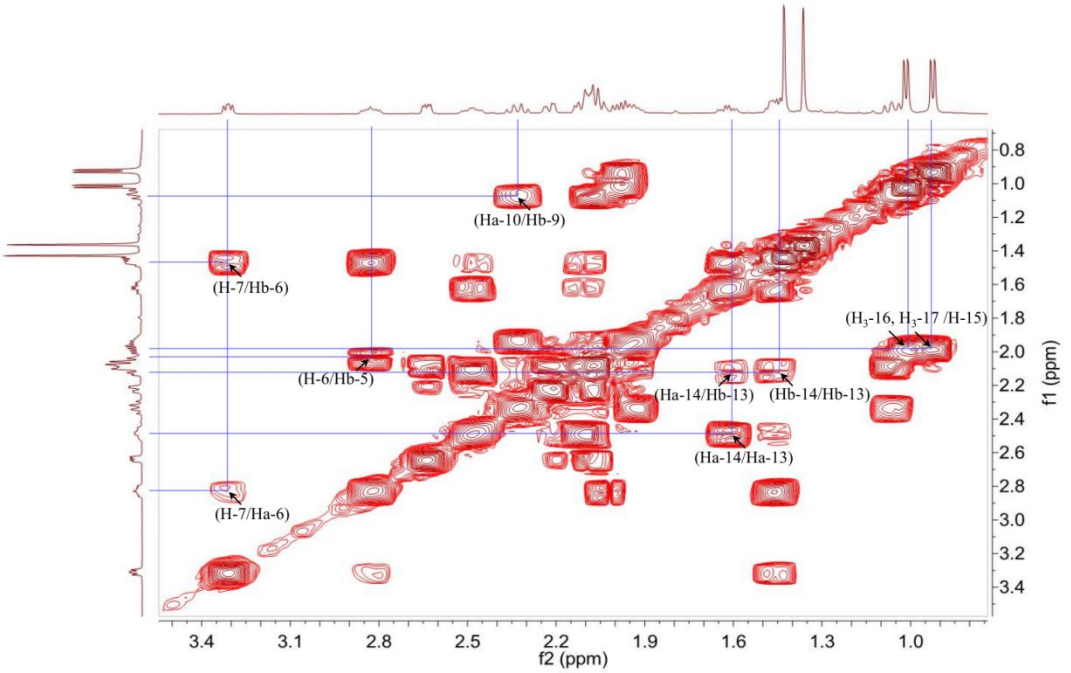


Figure S20. Enlarged ^{1}H-^{1}H COSY (600 MHz) spectrum of **1** in pyridine-d_5.

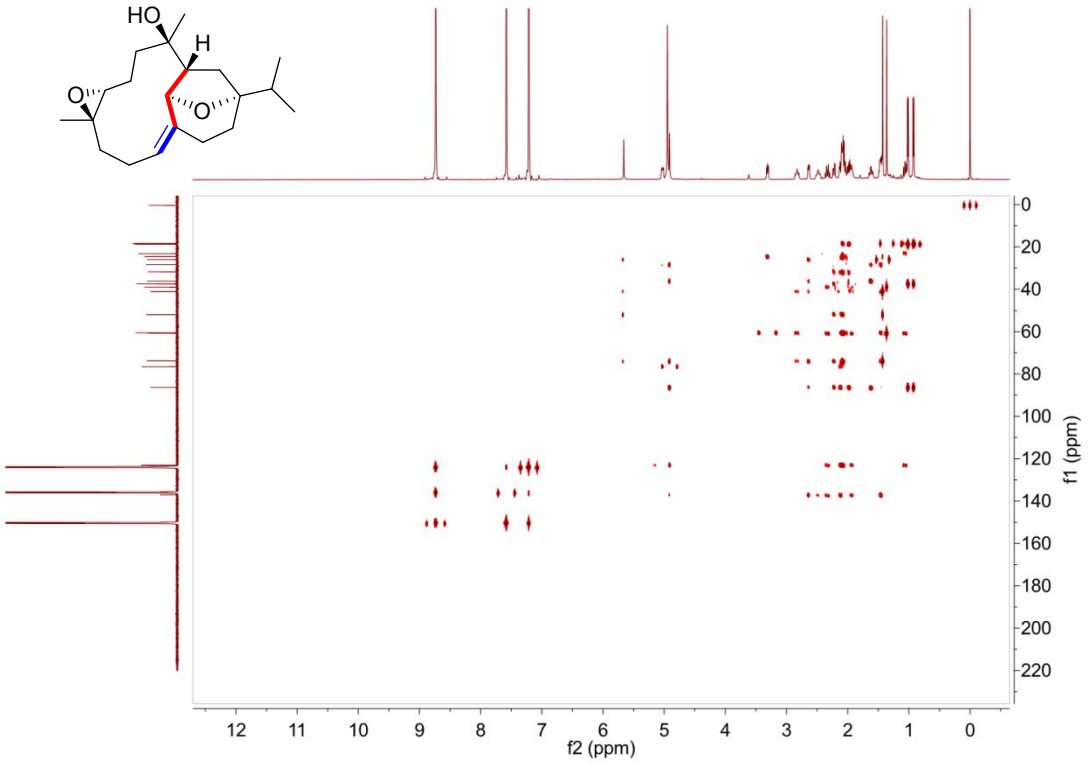


Figure S21. HMBC (600 MHz) spectrum of **1** in pyridine-d_5.

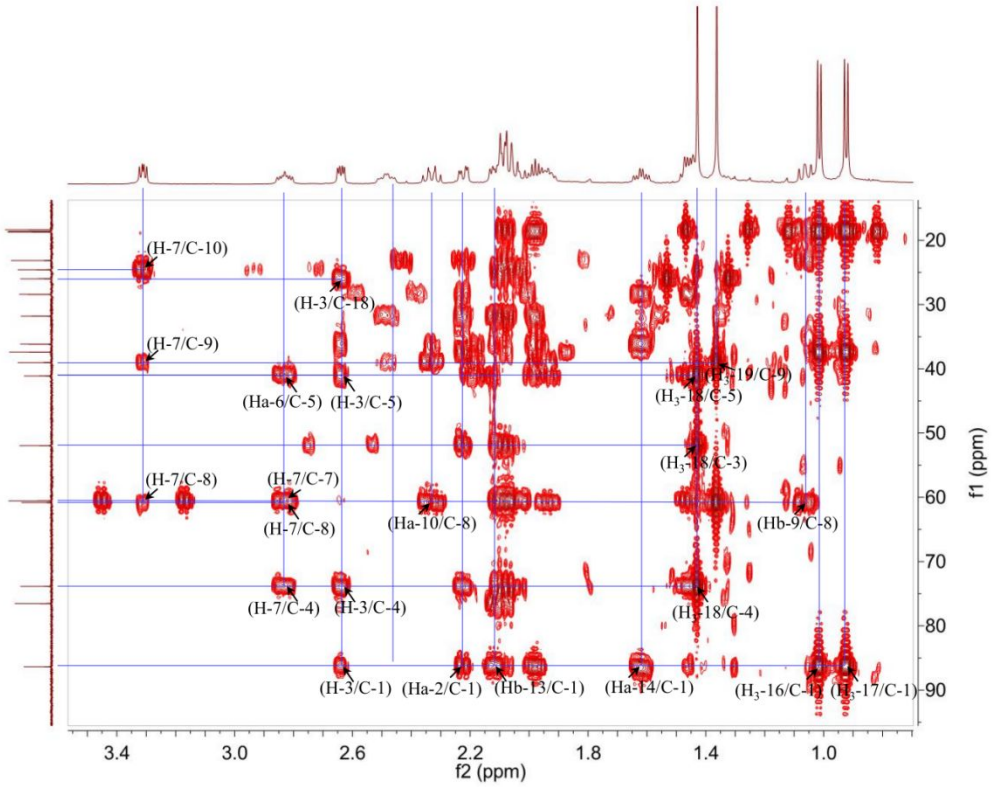


Figure S22. Enlarged HMBC (600 MHz) spectrum of **1** in pyridine-d_5.

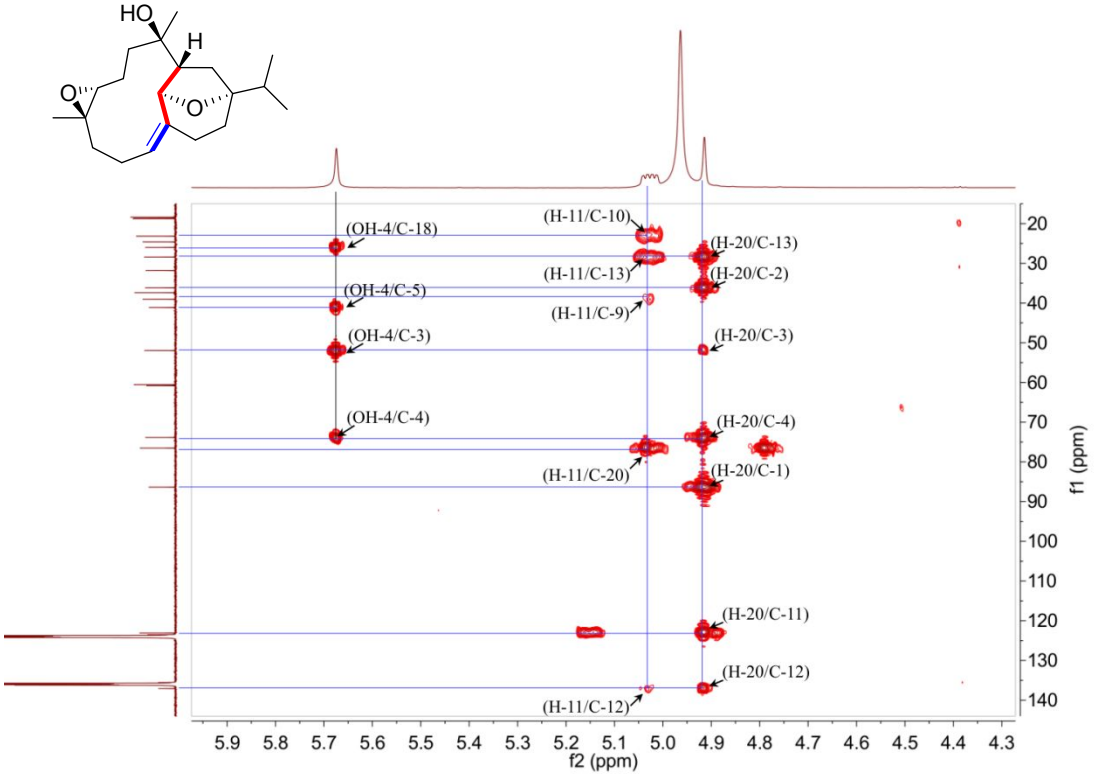


Figure S23. Enlarged HMBC (600 MHz) spectrum of **1** in pyridine-d_5.

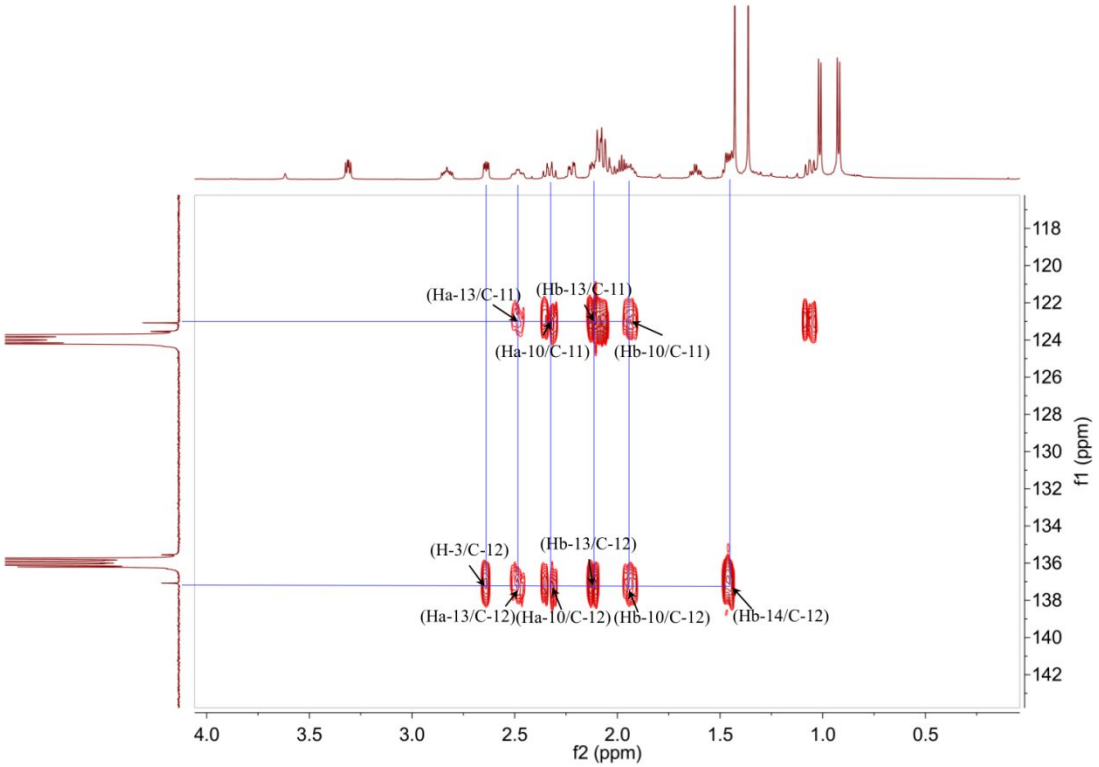


Figure S24. Enlarged HMBC (600 MHz) spectrum of **1** in pyridine-d_5.

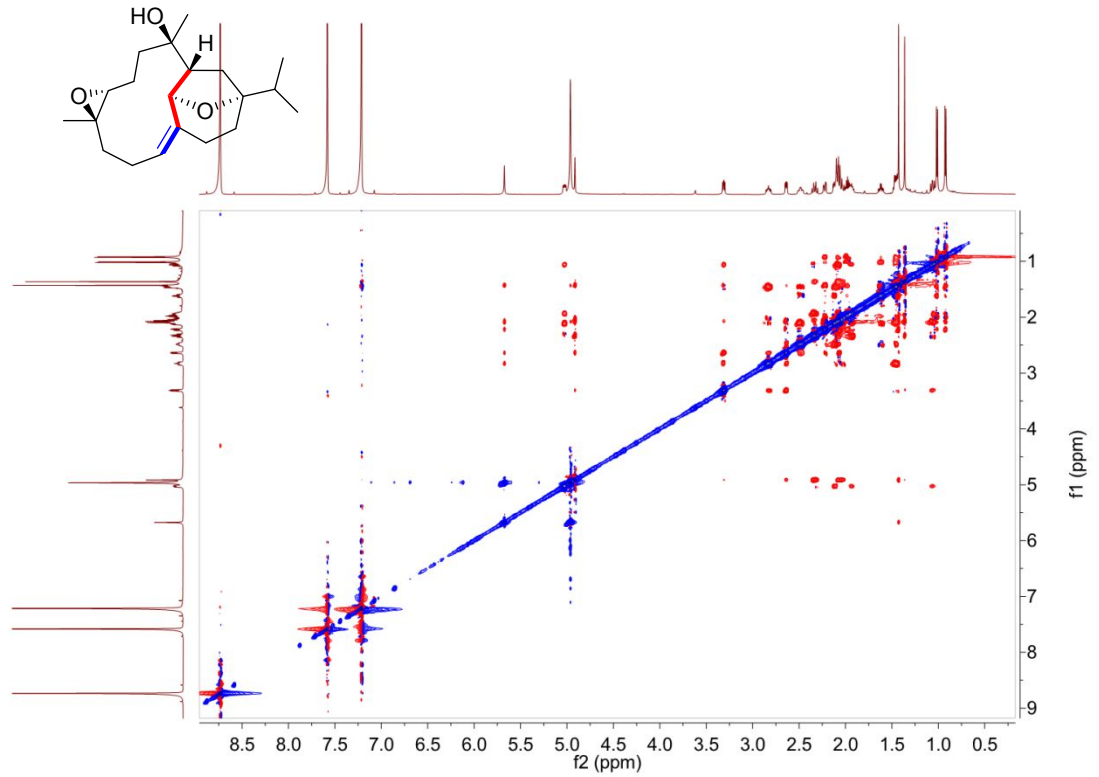


Figure S25. ROESY (600 MHz) spectrum of **1** in pyridine-d_5.

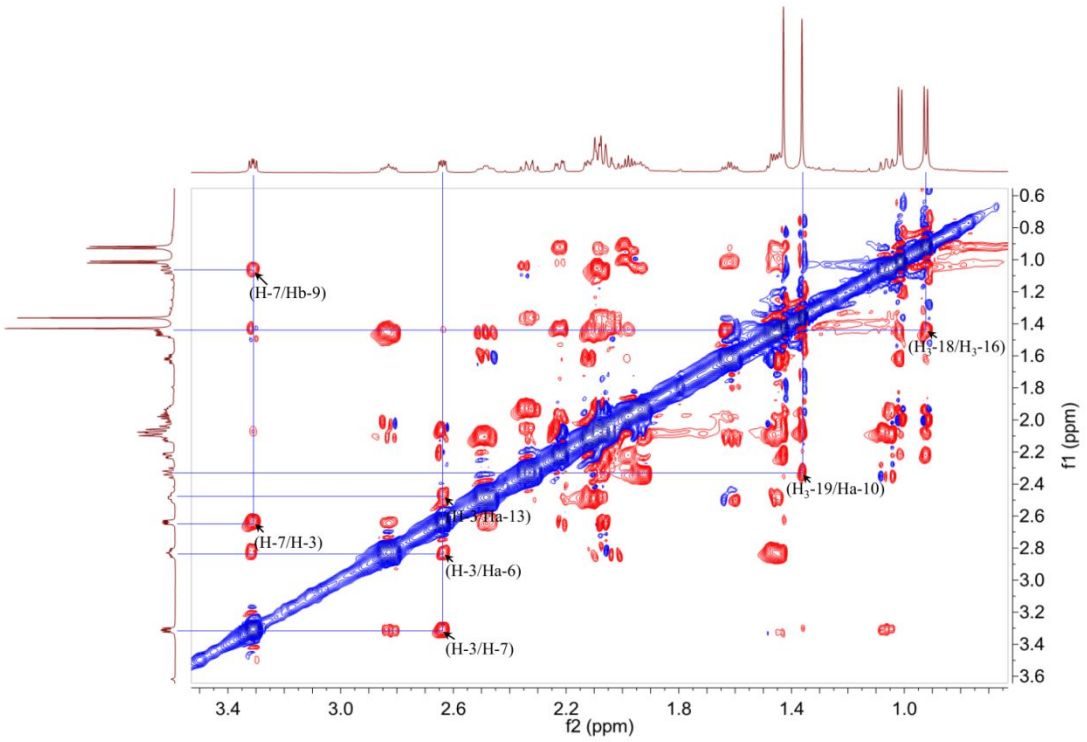


Figure S26. Enlarged ROESY (600 MHz) spectrum of **1** in pyridine-d_5.

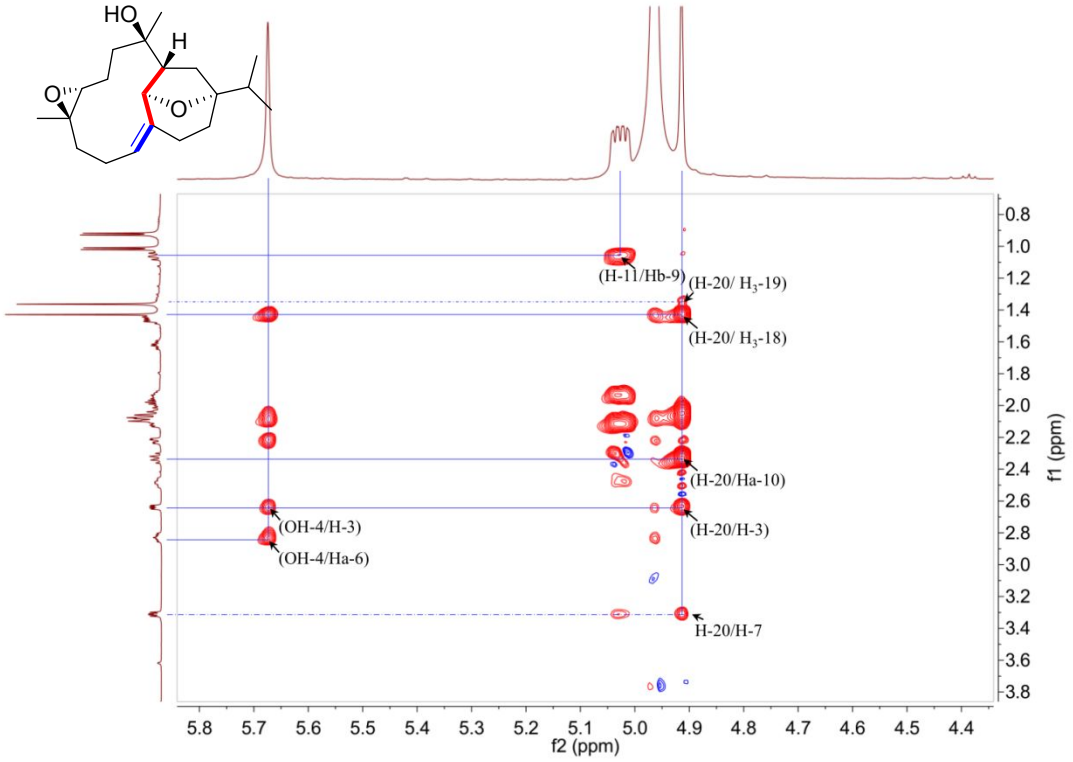


Figure S27. Enlarged ROESY (600 MHz) spectrum of **1** in pyridine-d_5.

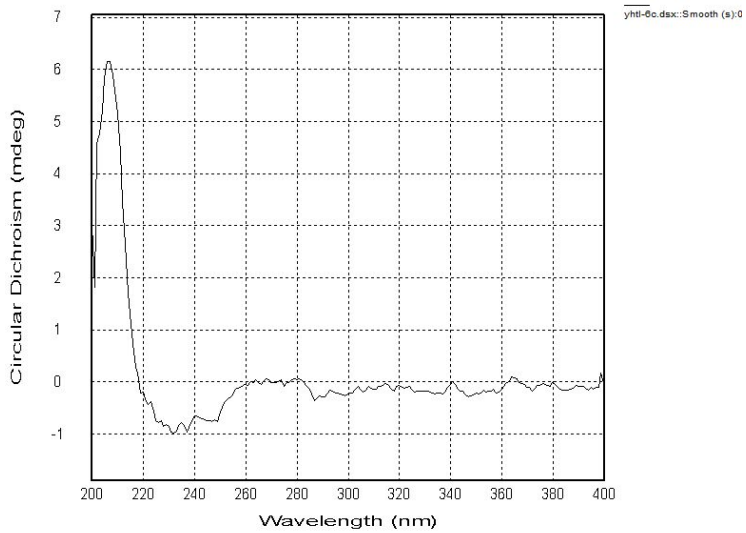


Figure S28. CD spectrum of **1**.

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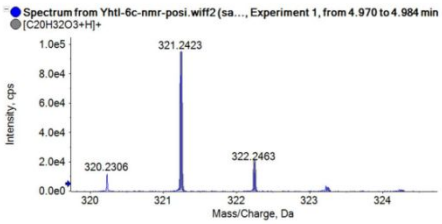
Acquisition Date	29/10/2020 10:34:13 AM	**Result Table**	Yhtl-6c
Acquisition Method	N/A	**Algorithm Used**	AutoPeak
Project	N/A	**Instrument Name**	X500 QTOF

Mass Spretra



#	Analyte Peak Name	Formula	Precursor Mass	Found At Mass	Mass Error (ppm)
1	Yhtl-6c	C20H32O3	321.2420	321.2423	-0.3

Figure S29. HRESIMS of **1**.

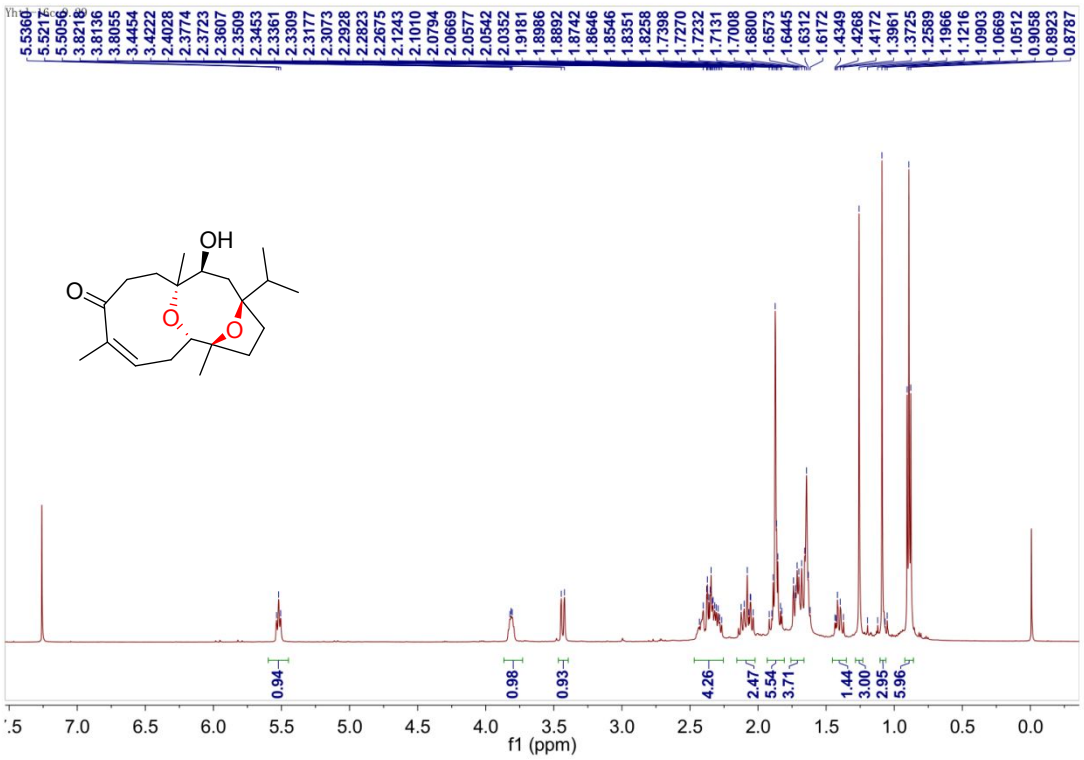


Figure S30. ^{1}H NMR (600 MHz) spectrum of **2** in $CDCl_3$.

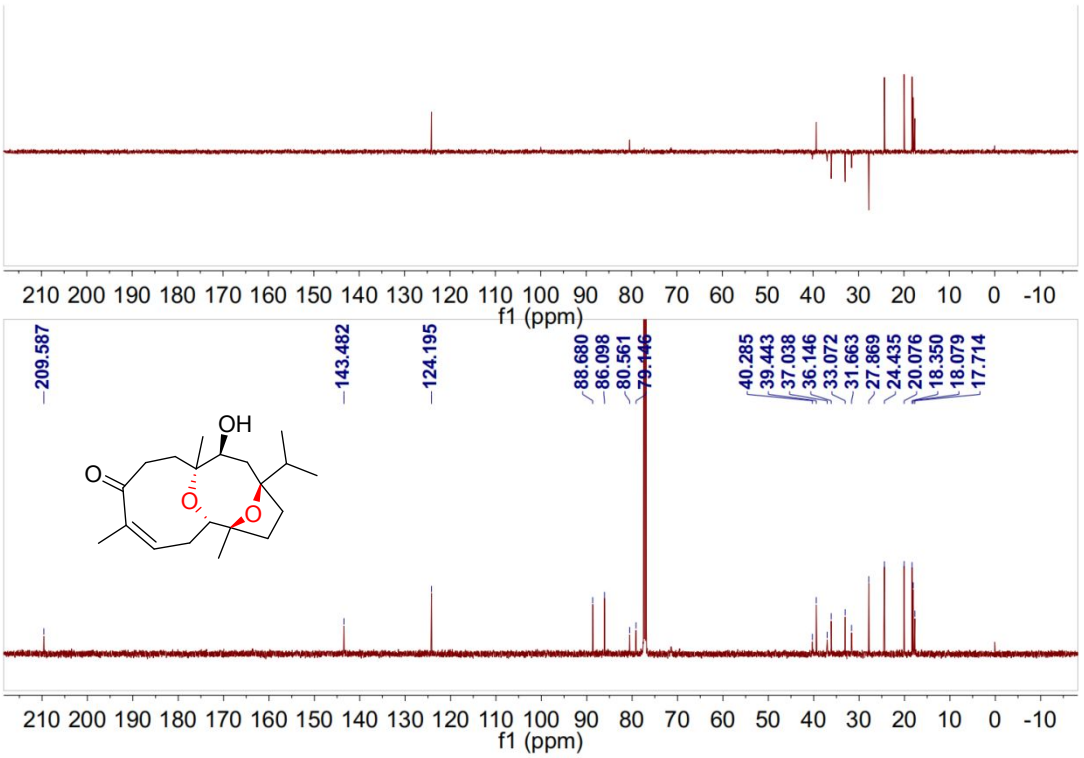


Figure S31. ^{13}C NMR and DEPT (150 MHz) spectra of **2** in $CDCl_3$.

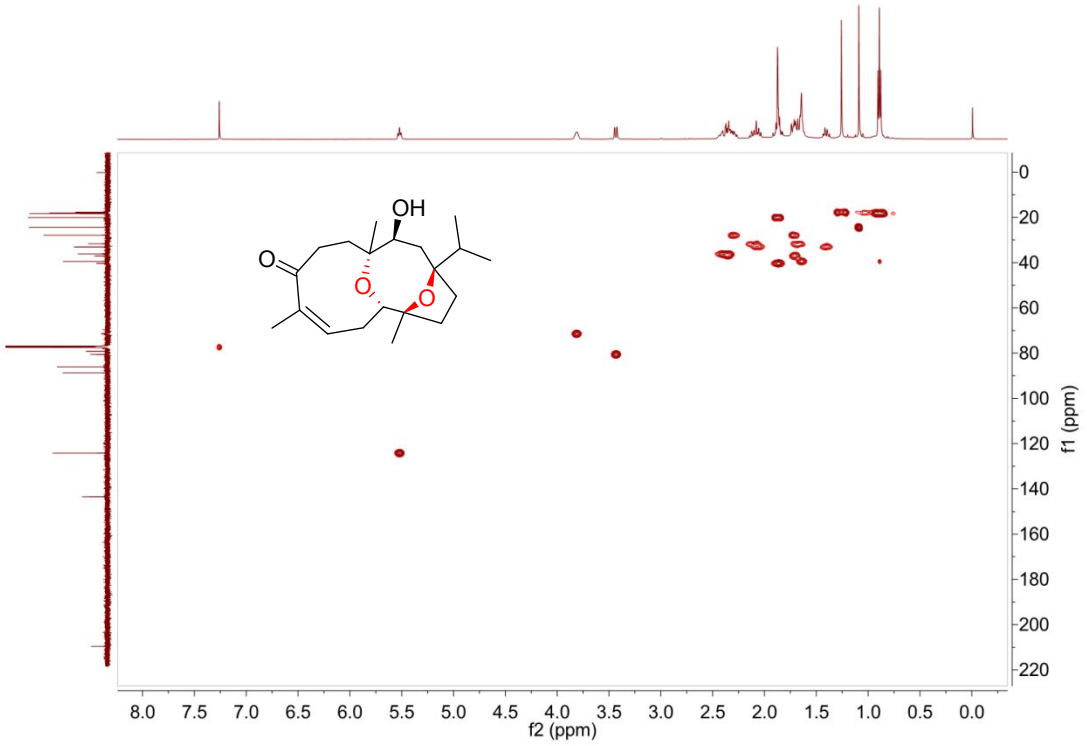


Figure S32. HSQC (600 MHz) spectrum of **2** in $CDCl_3$.

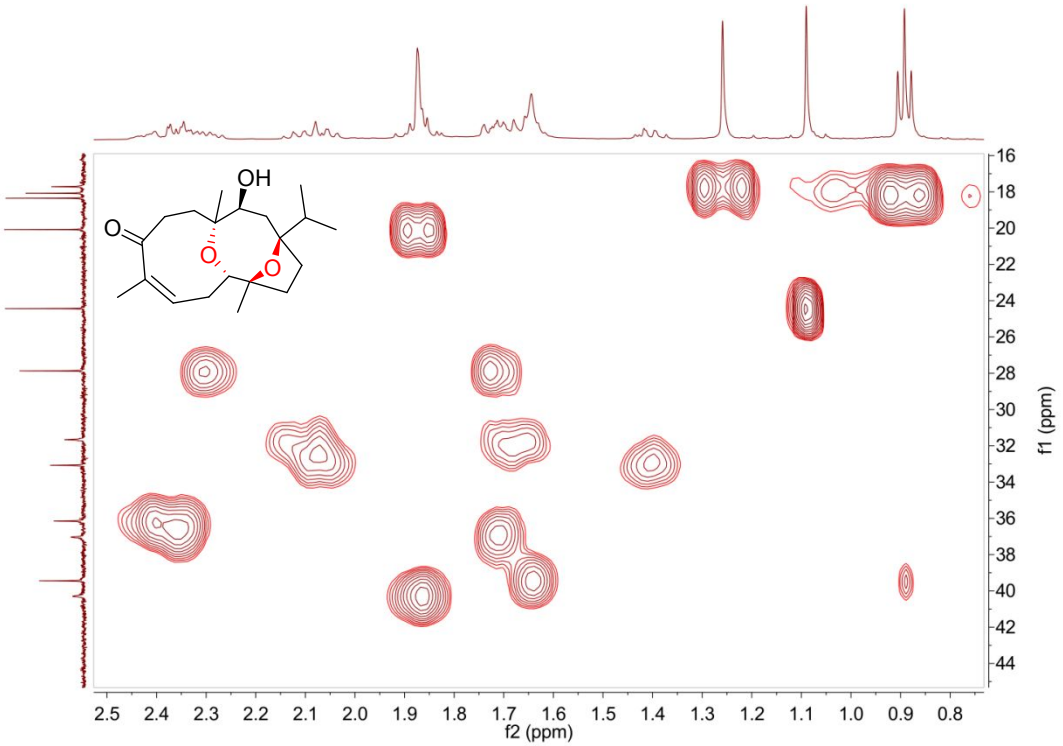


Figure S33. Enlarged HSQC (600 MHz) spectrum of **2** in $CDCl_3$.

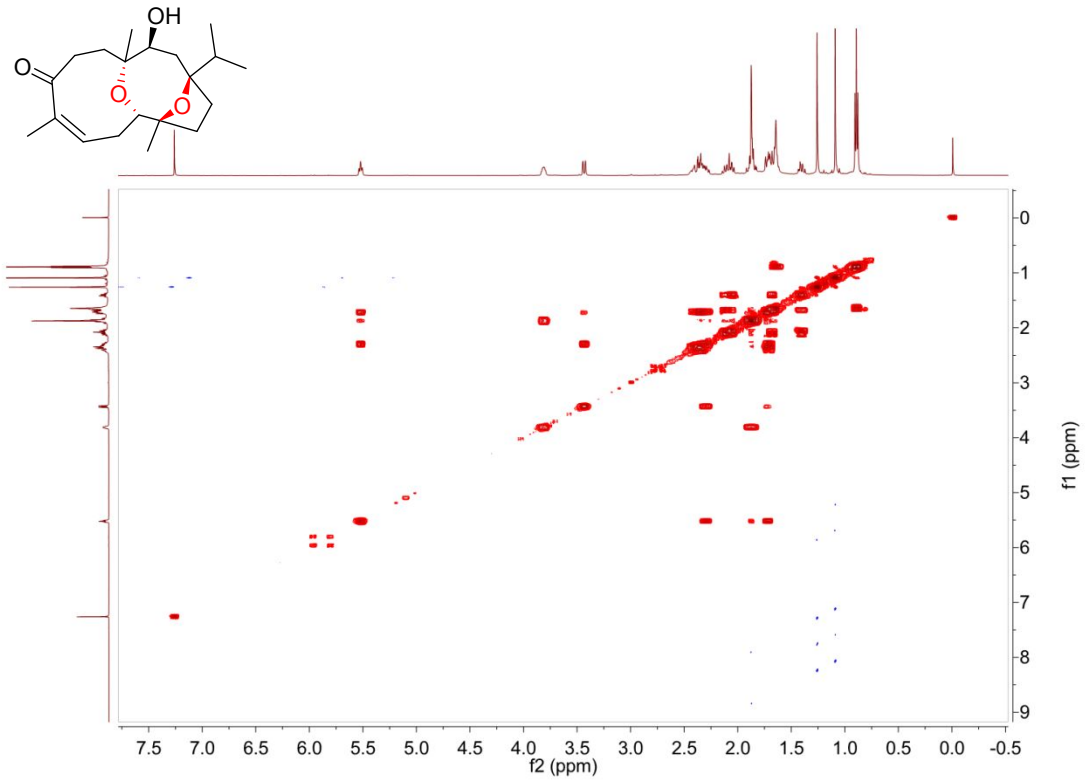


Figure S34. ^{1}H-^{1}H COSY (600 MHz) spectrum of **2** in $CDCl_3$.

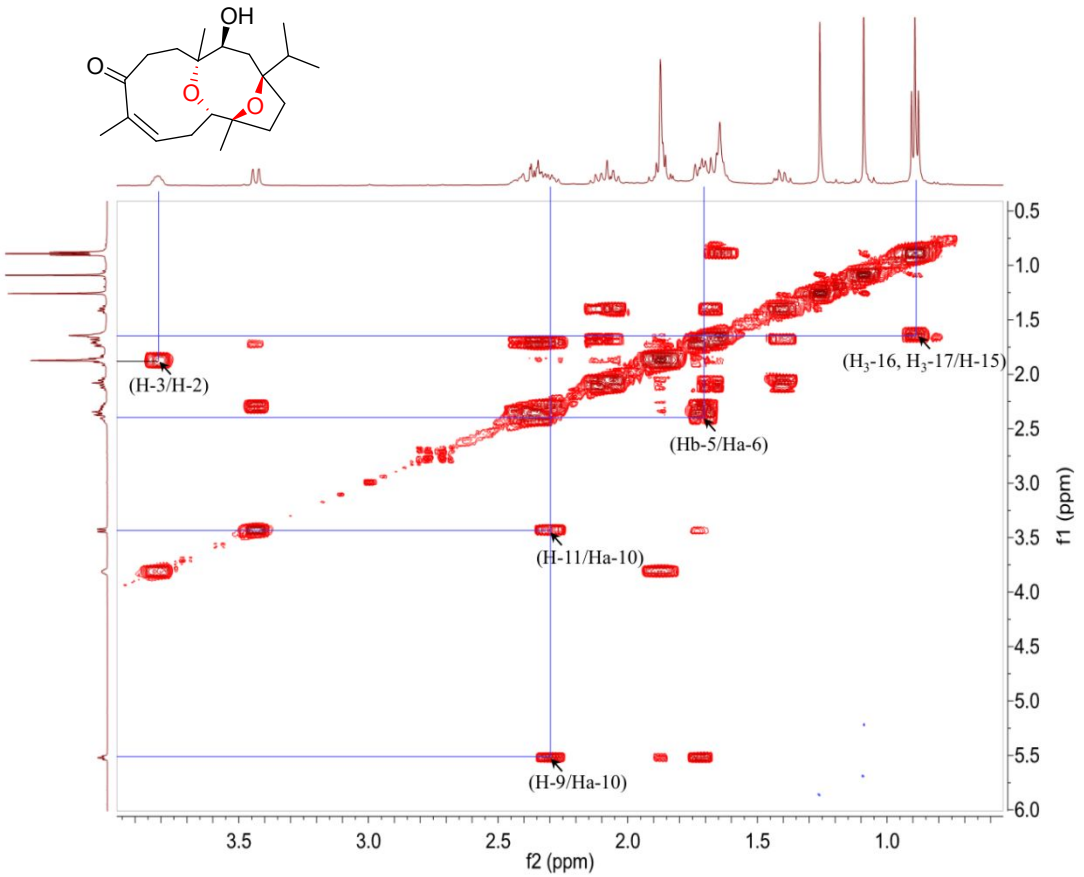


Figure S35. Enlarged ^{1}H-^{1}H COSY (600 MHz) spectrum of **2** in $CDCl_3$.

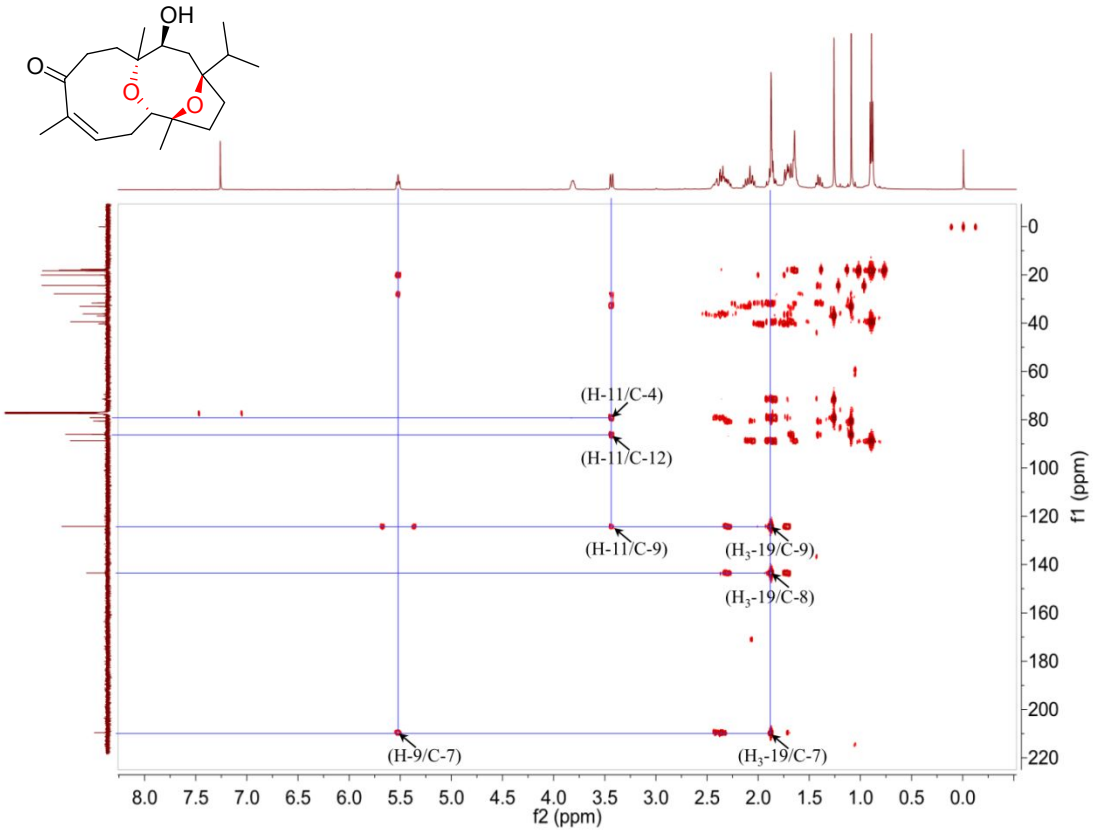


Figure S36. HMBC (600 MHz) spectrum of **2** in $CDCl_3$.

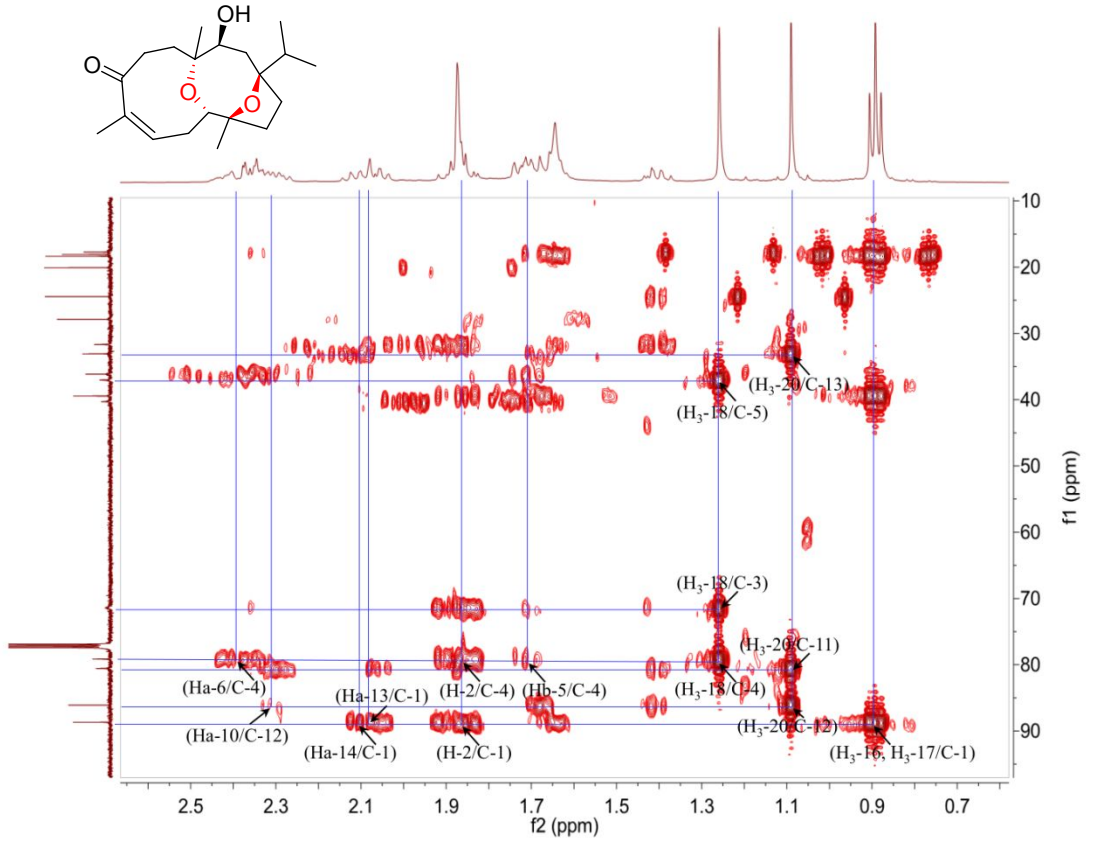


Figure S37. Enlarged HMBC (600 MHz) spectrum of **2** in $CDCl_3$.

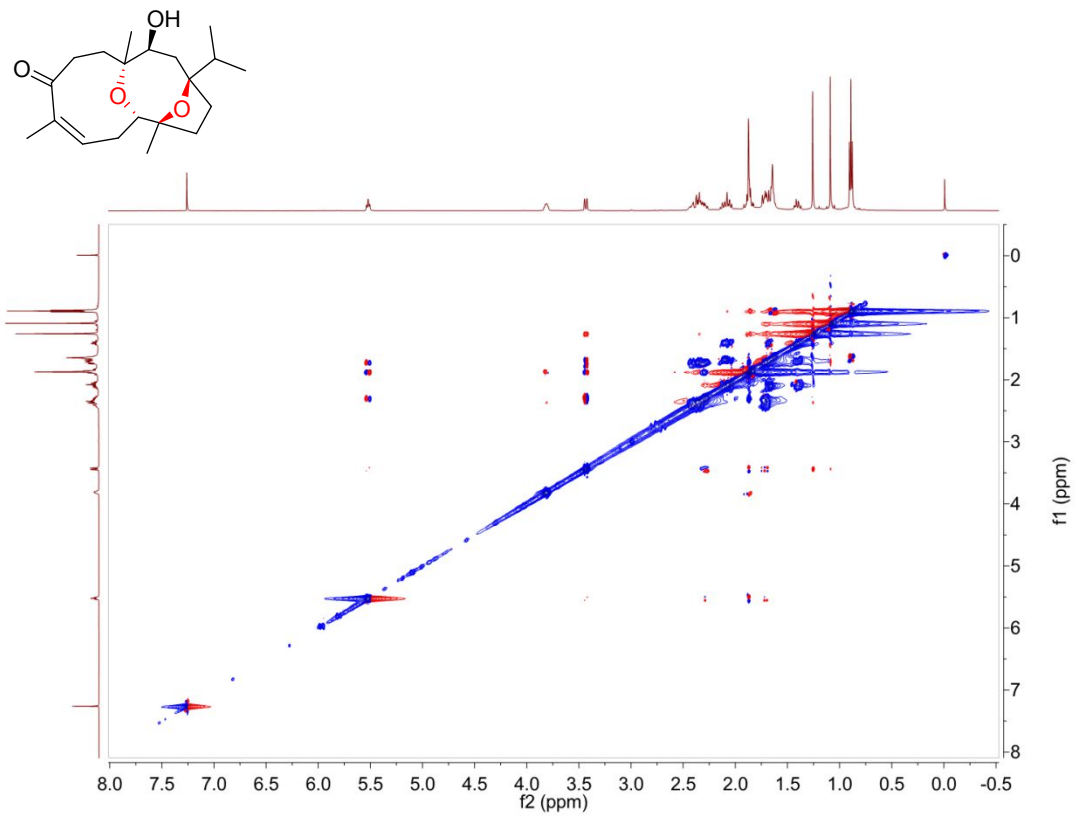


Figure S38. ROESY (600 MHz) spectrum of **2** in $CDCl_3$.

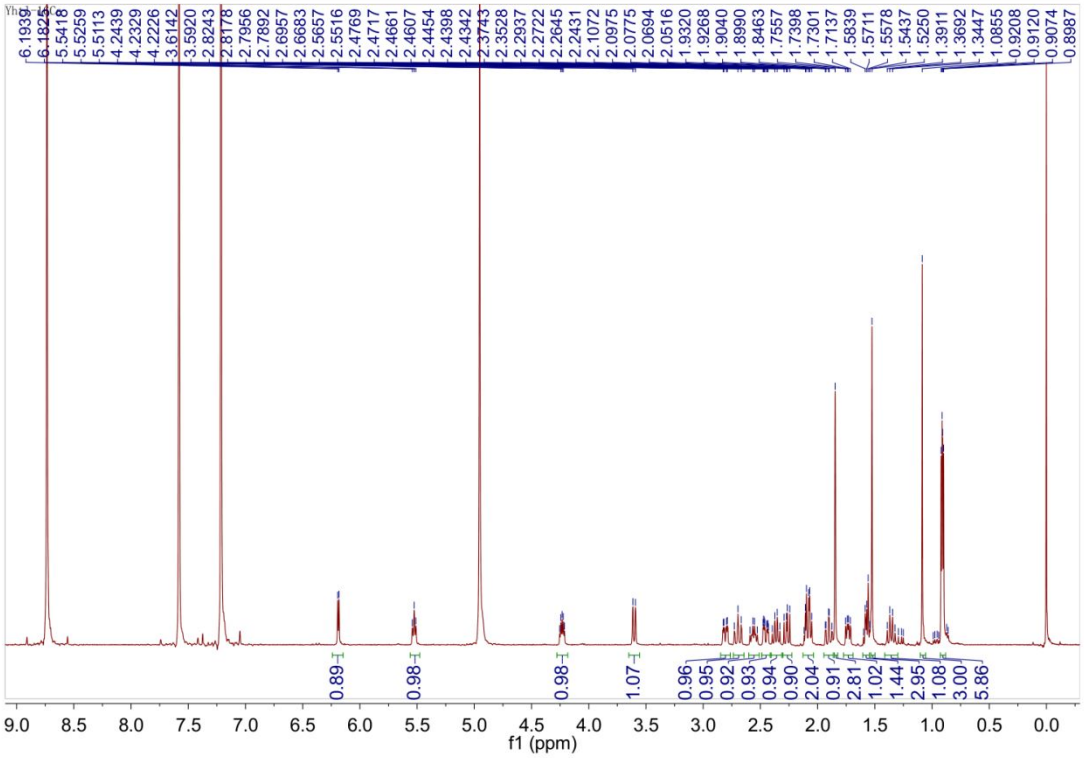


Figure S39. ^{1}H NMR (600 MHz) spectrum of **2** in pyridine-d_5.

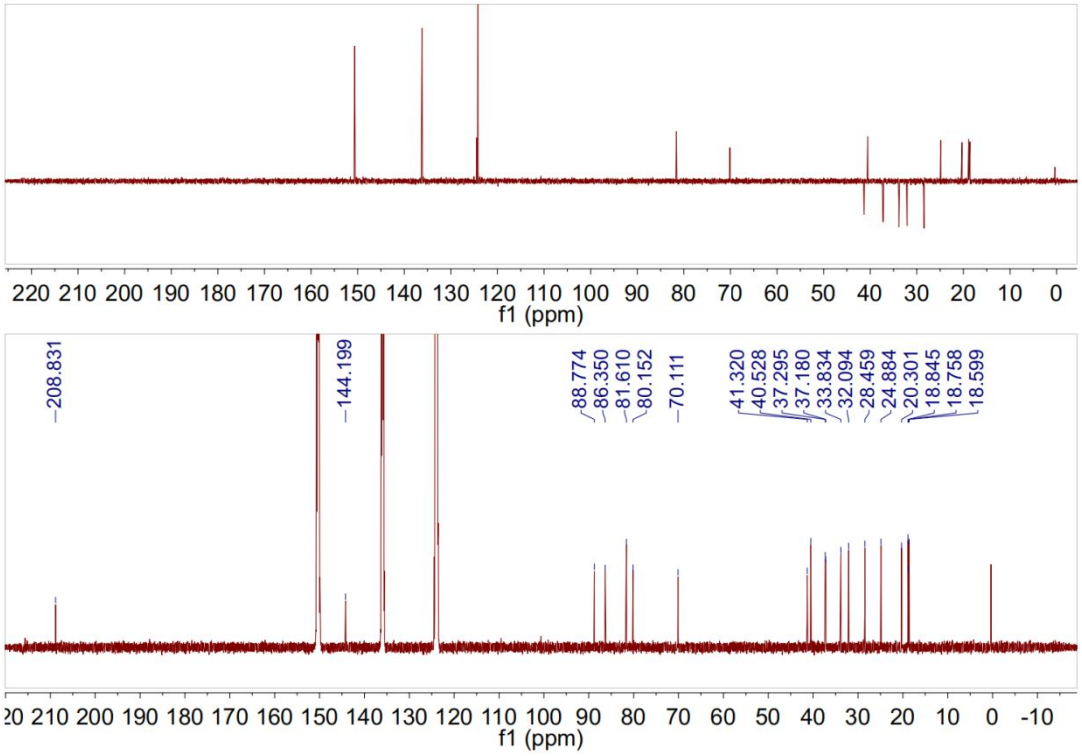


Figure S40. ^{13}C NMR and DEPT (150 MHz) spectra of **2** in pyridine-d_5.

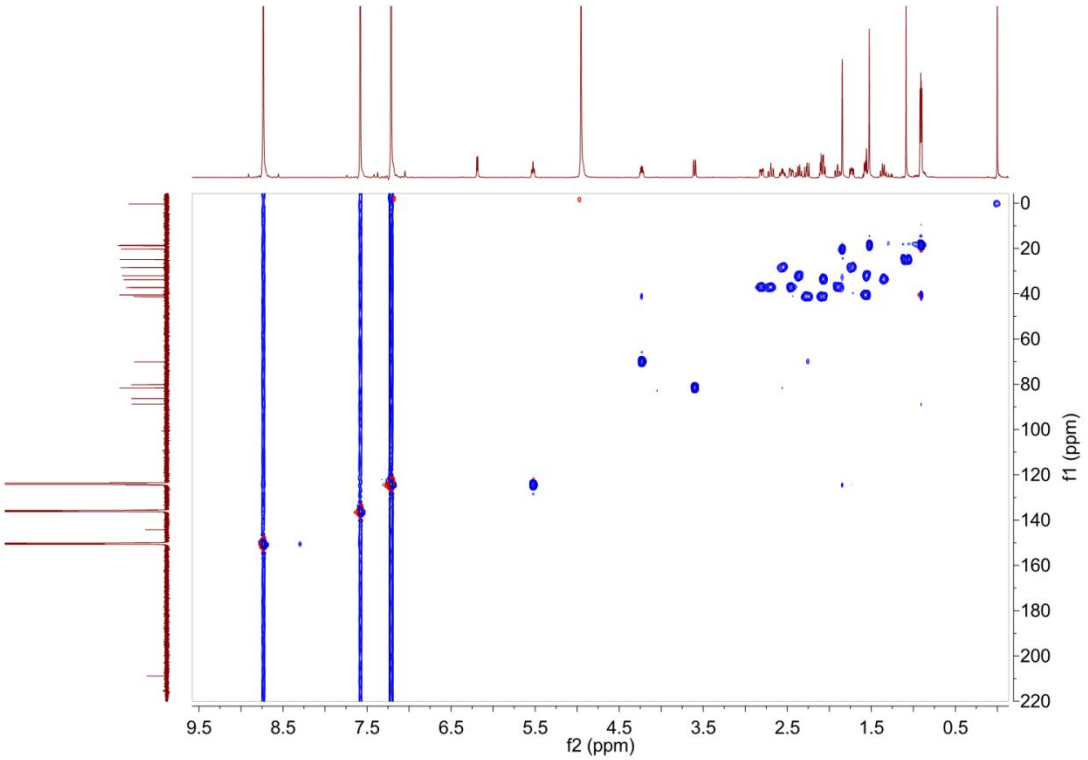


Figure S41. HSQC (600 MHz) spectrum of **2** in pyridine-d_5.

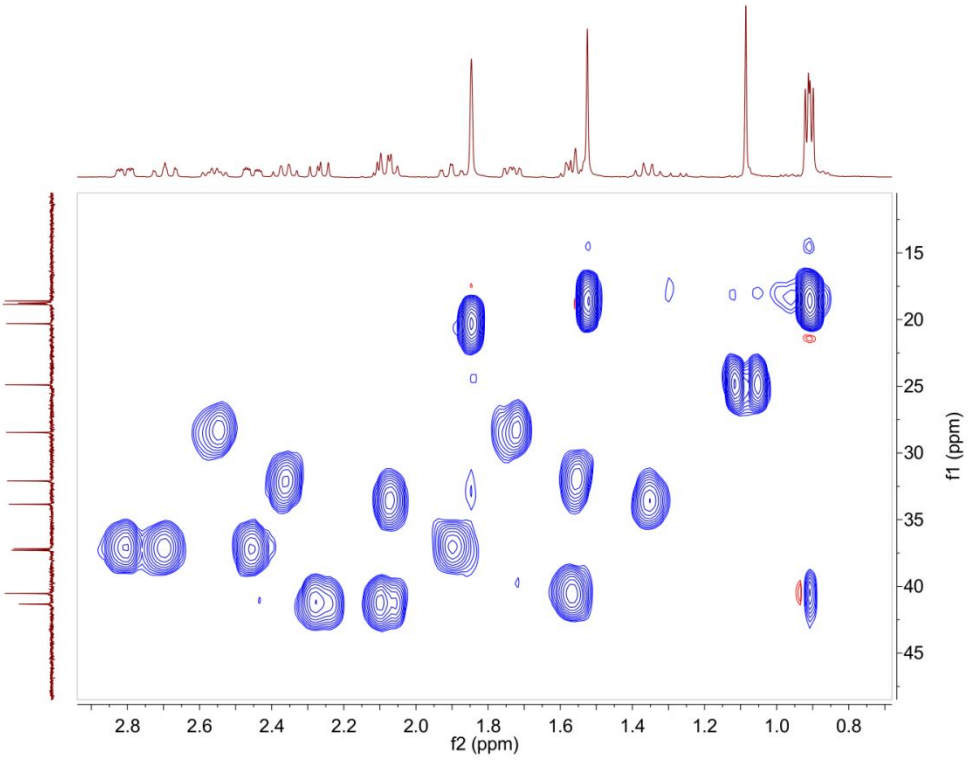


Figure S42. Enlarged HSQC (600 MHz) spectrum of **2** in pyridine-d_5.

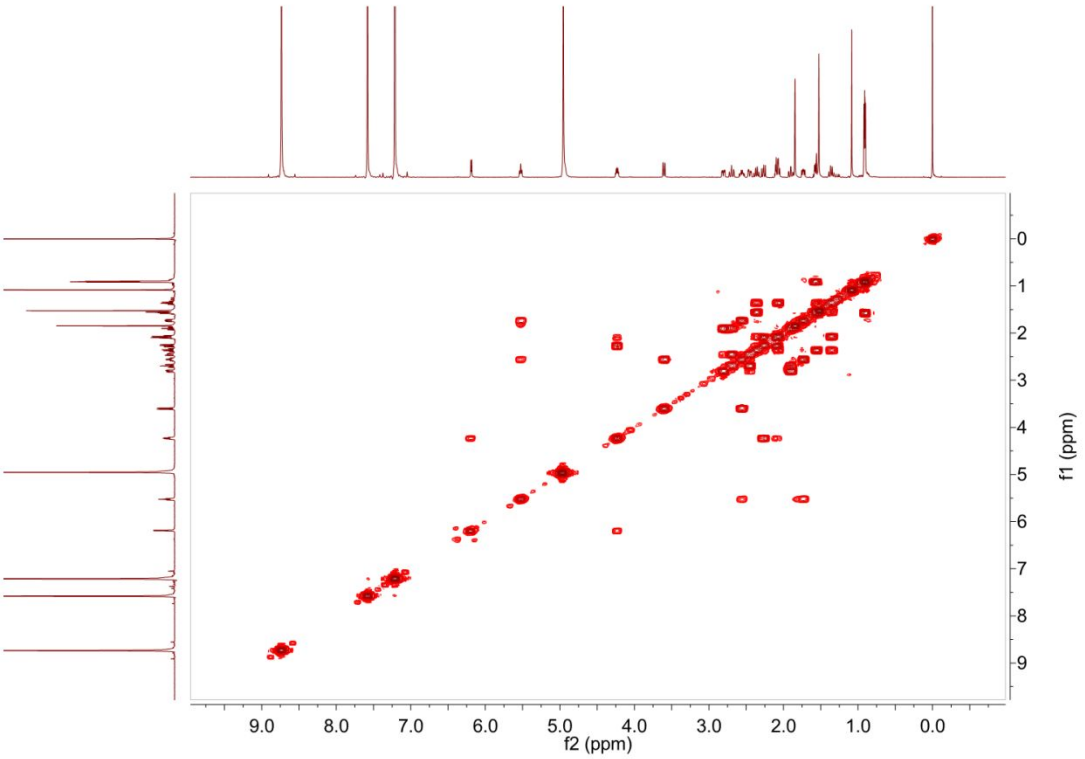


Figure S43. ^{1}H-^{1}H COSY (600 MHz) spectrum of **2** in pyridine-d_5.

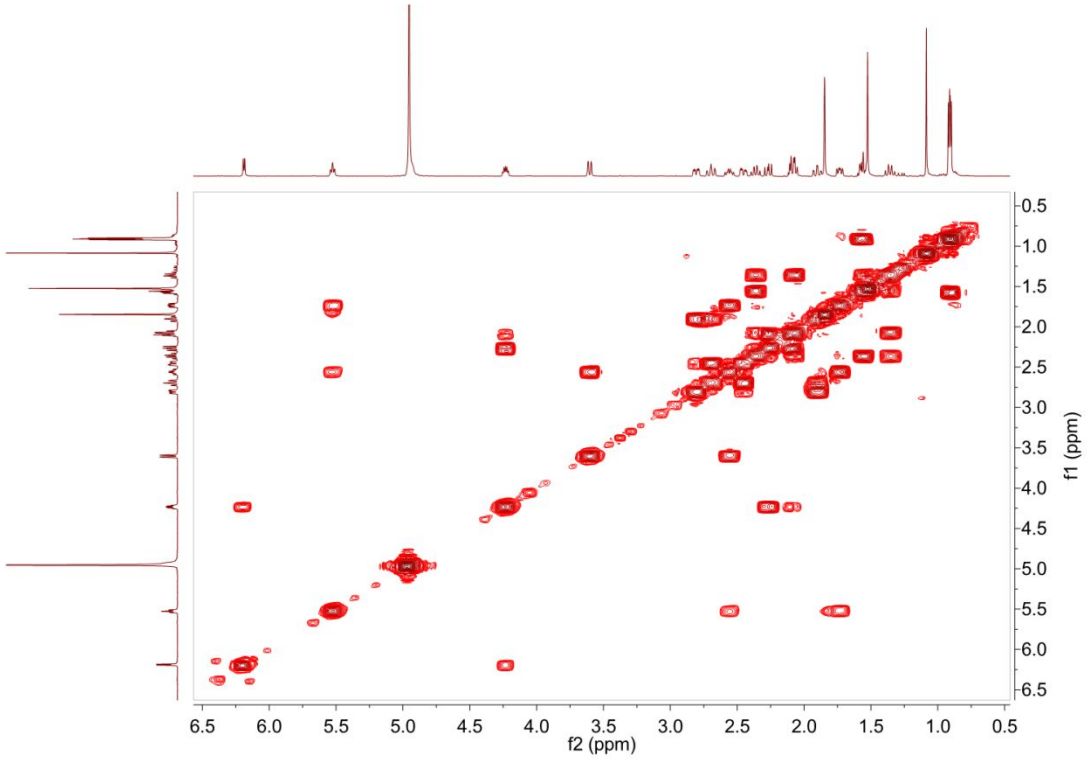


Figure S44. Enlarged ^{1}H-^{1}H COSY (600 MHz) spectrum of **2** in pyridine-d_5.

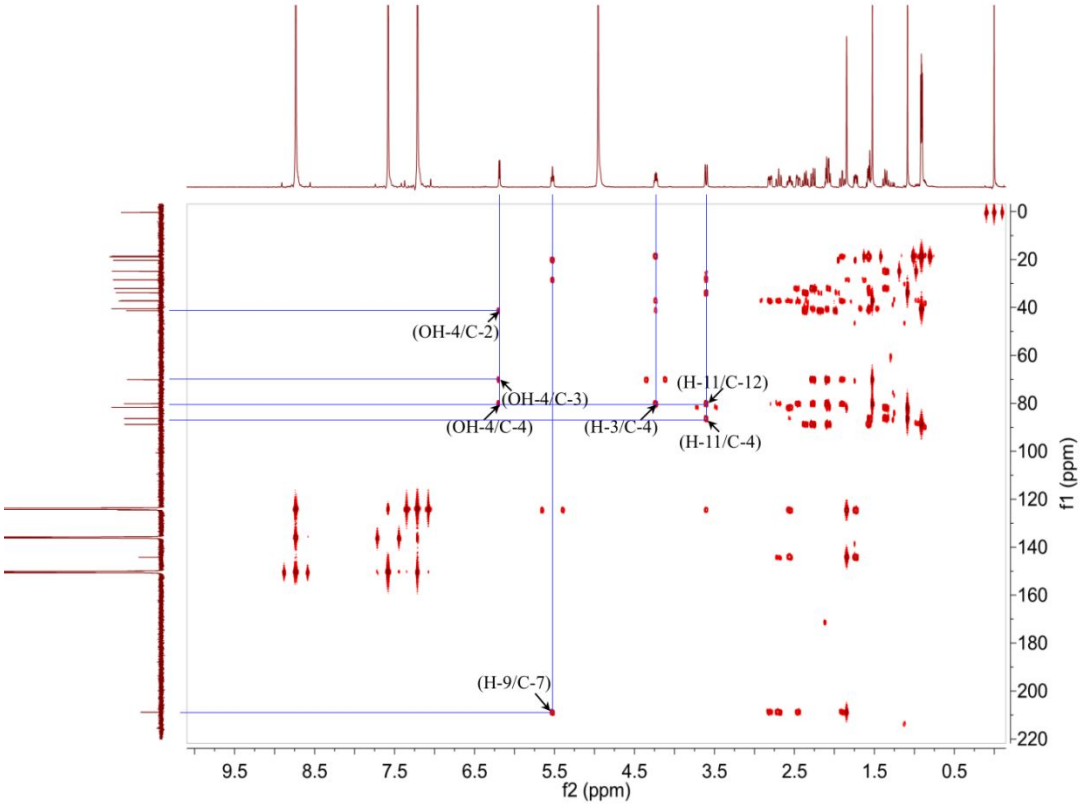


Figure S45. HMBC (600 MHz) spectrum of **2** in pyridine-d_5.

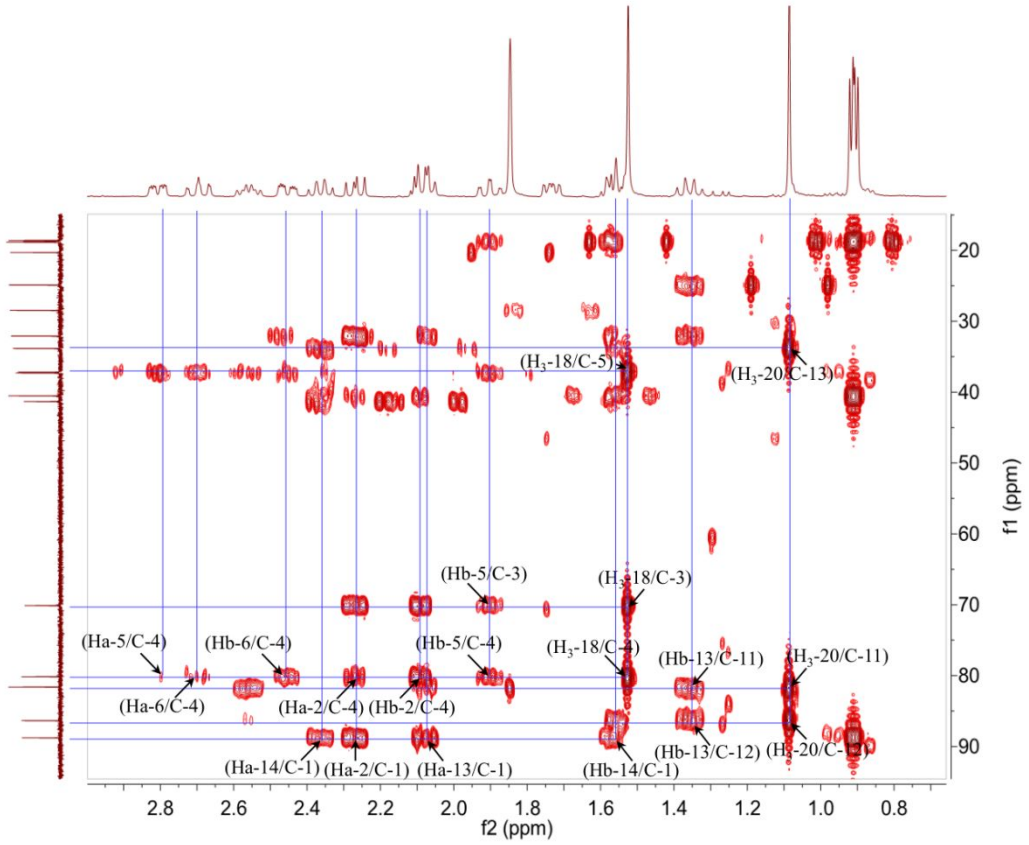


Figure S46. Enlarged HMBC (600 MHz) spectrum of **2** in pyridine-d_5.

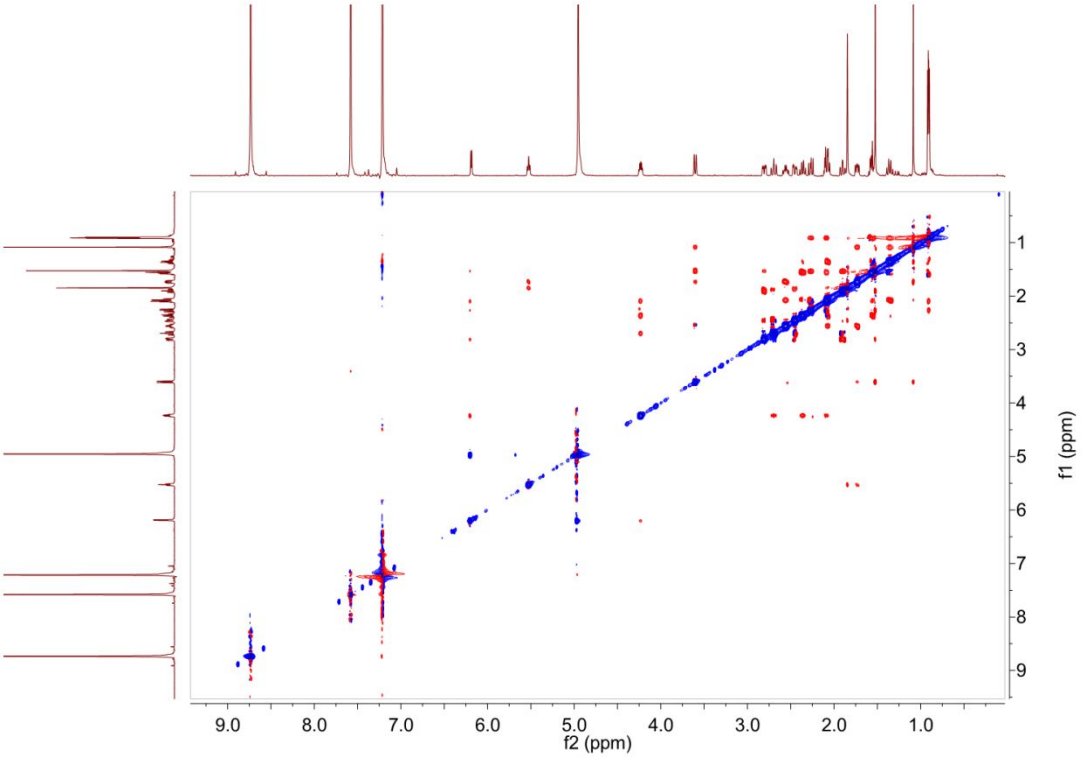


Figure S47. ROESY (600 MHz) spectrum of **2** in pyridine-d_5.

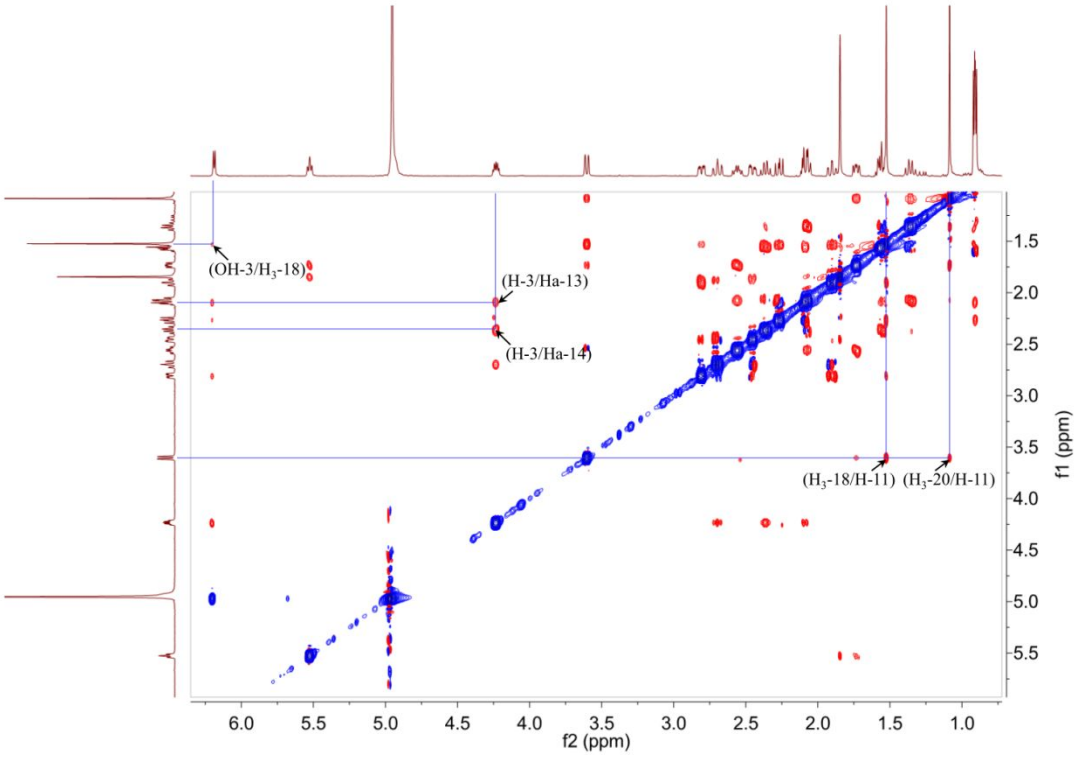


Figure S48. Enlarged ROESY (600 MHz) spectrum of **2** in pyridine-d_5.

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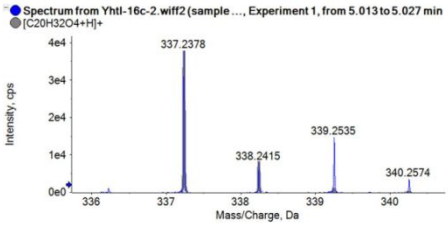
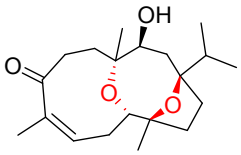
Acquisition Date	28/10/2020 2:55:16 PM	Result Table	Yhtl-16C
Acquisition Method	N/A	Algorithm Used	AutoPeak
Project	LYY	Instrument Name	X500 QTOF

Mass Spretra





#	Analyte Peak Name	Formula	Precursor Mass	Found At Mass	Mass Error (ppm)
1	Yhtl-16c	C20H32O4	337.2370	337.2378	1.4

Figure S49. HRESIMS of **2**.

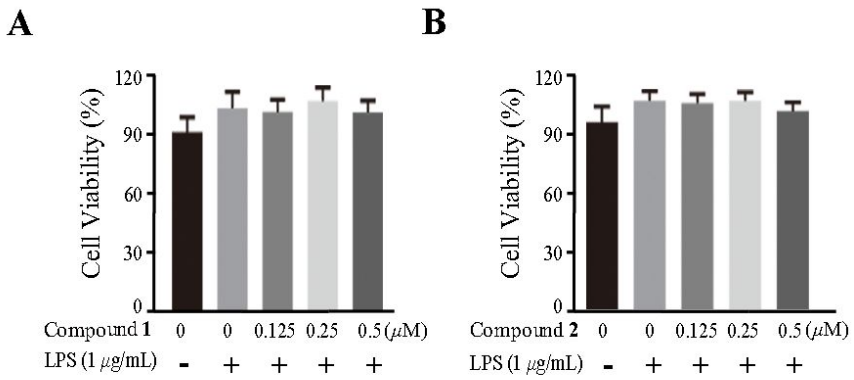


Figure S50. RAW264.7 cell proliferation in response to compound **1** (A) and compound **2** (B) at different doses assayed by CCK-8 assay. Data represent mean ± SEM values of three experiments.