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Supporting Information for

Cephaloliverols A and B, two sterol-hybrid meroterpenoids from *Cephalotaxus oliveri*

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General experimental procedures

Optical rotations were recorded on Anton Par MCP 200 polarimeter. UV spectra were measured on a Shimadzu UV-2400 spectrophotometer. ECD spectrum was recorded on Bio-Logic Science MOS-450 spectropolarimeter. HRESIMS data were measured on a Bruker micro-TOFQ-Q mass spectrometer. NMR spectroscopic data were recorded on Bruker AV-600 NMR spectrometer with tetramethylsilane as an internal standard. ODS (50 μm, YMC Co. Ltd., Kyoto, Japan), MCI gel (Mitsubishi Chemical Industries Ltd., Tokyo, Japan) and Sephadex LH-20 (Pharmacia Biotech, Uppsala, Sweden) were used for column chromatography (CC). Semi-preparative HPLC was conducted on a YMC ODS-A column (250 × 10 mm I.D., 5 μm) equipped with a LC-6AD pump and a Shimadzu SPD-20A UV-vis detector (Shimadzu Co., Ltd., Tokyo, Japan). TLC analysis was performed on silica gel plates (GF254, Qingdao Haiyang Chemical Co., Ltd., Qingdao, PR China).

Plant material

The twigs and leaves of *C. oliveri* were collected in October 2019 from Longhui County, Shaoyang City of Hunan Province, PR China, and authenticated by Mr. Hongqiang Zhang of Kunming GenPHYTech Co. Ltd, Kunming, PR China. A voucher specimen (No. 20191105) was deposited at School of Traditional Chinese Materia Medica, Shenyang Pharmaceutical University, Shenyang, PR China.

Extraction and isolation

The air–dried twigs and leaves of *C. oliveri* (50 kg) were extracted with 95% EtOH twice and 75% EtOH once. The crude EtOH extract was suspended in water and then partitioned with petroleum ether (PE), dichloromethane (CH_2Cl_2), and *n*–butanol (*n*–BuOH) successively to afford PE, CH_2Cl_2, and *n*–BuOH extracts, respectively. The CH_2Cl_2 extract (70 g) was subjected to an MCI column eluted with CH_3OH–H_2O (30% to 100% CH_3OH) to yield nine fractions F1–F9.

F5 was separated subsequently by ODS (30% to 70% CH_3OH–H_2O) to obtain four subfractions F5A–F5D. F5C was chromatographed over Sephadex LH–20, and then purified by semi–preparative HPLC (CH_3OH–H_2O 65:35, v/v; flow rate, 2.0 mL/min) to obtain **4** (3.0 mg, t_R = 49.8 min). F8 was subjected to

silica gel CC (petroleum ether–ethyl acetate 100:0–100:50, v/v) to get four subfractions F8A–F8D. F8A was purified by Sephadex LH–20 (CH_3OH) to furnish **3** (5.1 mg). F8B was further separated by ODS, Sephadex LH–20 (CH_3OH) CC and semi–preparative HPLC (CH_3OH–H_2O 91:9, v/v; flow rate, 2.0 mL/min) to yield **1** (6.0 mg, t_R = 77.8 min) and **2** (6.0 mg, t_R = 87.4 min).

Cephaloliverol A (**1**). white solid; $[\alpha]_D^{20}$ –37 (*c* 0.3, CH_3OH); UV (CH_3OH) λ_{max} (log ε): 253 (3.45), 284 (3.31), 335 (3.32) nm; ECD (CH_3OH) λ_{max} (log ε): 253 (+68.30), 280 (+102.10), 335 (–69.76) nm; IR (KBr) υ_{max}: 3383, 2958, 2871, 1714, 1635, 1600, 1467, 1340, 1328, 1271, 1176, 1018 cm^{-1}; HRESIMS (*m/z*): 769.5042 $[M–H]^-$ (calcd for $C_{49}H_{69}O_7$, 769.5043).

Cephaloliverol B (**2**). white solid; $[\alpha]_D^{20}$ +25 (*c* 0.3, CH_3OH); UV (CH_3OH) λ_{max} (log ε): 253 (3.93), 285 (3.74), 334 (3.74) nm; ECD (CH_3OH) λ_{max} (log ε): 227 (+25.96), 279 (+82.60) nm; IR (KBr) υ_{max}: 3377, 2958, 2870, 1714, 1635, 1600, 1462, 1340, 1272, 1178, 1022 cm-1; HRESIMS (*m/z*): 769.5074 $[M–H]^-$ (calcd for $C_{49}H_{69}O_7$, 769.5043).

The other known metabolites isolated from this extract include sugiol, 3β-hydroxysugiol, hinokione, 3β,12,16-trihydroxy-6,8,11,13-abietatrien, salviniol, hinokiol, 8β-hydroxy-9(11),13-abietadien-12-one, abieta-6,8,11,13-tetraene-3β,12-diol, dehydrouomifoliol, epiloliolide, kaempferol, 3',4',5',5,7-pentamethoxyflavone, homoeriodictyol, naringenin, epicatechin, catechin, and 5,7-dihydroxychromone.

Cell culture and determination of NO

RAW 264.7 macrophages were cultured in Dulbecco's modified eagle medium (DMEM) (Hyclone, Logan, USA) with 10% fetal bovine serum (FBS, Gibco, Gaithersburg, USA) in a humidified atmosphere containing 5% CO_2 at 37 °C for 48 h. Cells were treated with different concentrations of compounds (10, 20, and 40 μM) and incubated for 1 h. After incubation, lipopolysaccharide (LPS) (1 μg/mL) was introduced to cells. The culture medium was collected and used

for measuring NO production by NO Test Kit. And indomethacin (10 μM) was used as the positive drug.

Electronic circular dichroism (ECD) caculation

The conformation of the simplified structure of **1** was used to search by Spartan 14.0 (Wavefunction Inc., Irvine, CA, USA) under MMFF force field. The low-energy conformers of them were optimized in the gas phase by semi-empirical method in Gaussian 09 program package, which were further reoptimized and analysed, using the density functional theory (DFT) at the RB31YP/6-31 G (d, p) level, resulted in no imaginary frequencies. Then the ECD spectra were calculated using TD-DFT-B3LYP/6-31 G (d, p) level in methanol solution. Finally, the calculated low-energy conformational results were Boltzmann averaged by software SpecDis1.51 to yield the depicted ECD spectra of the simplified structure of **1**. The ECD spectra of the simplified structure of **2** were calculated in the same method.

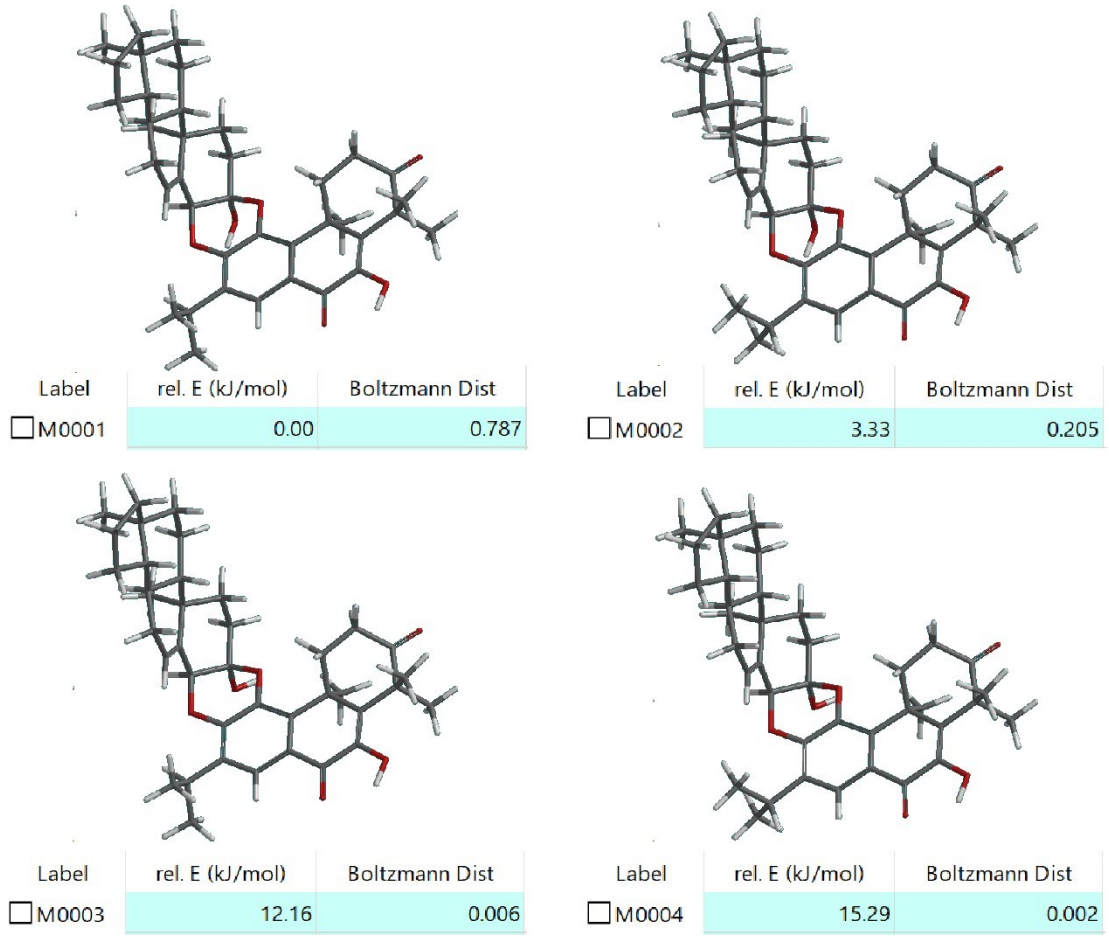


Figure S1 The optimized conformers of the simplified structure of **1**.

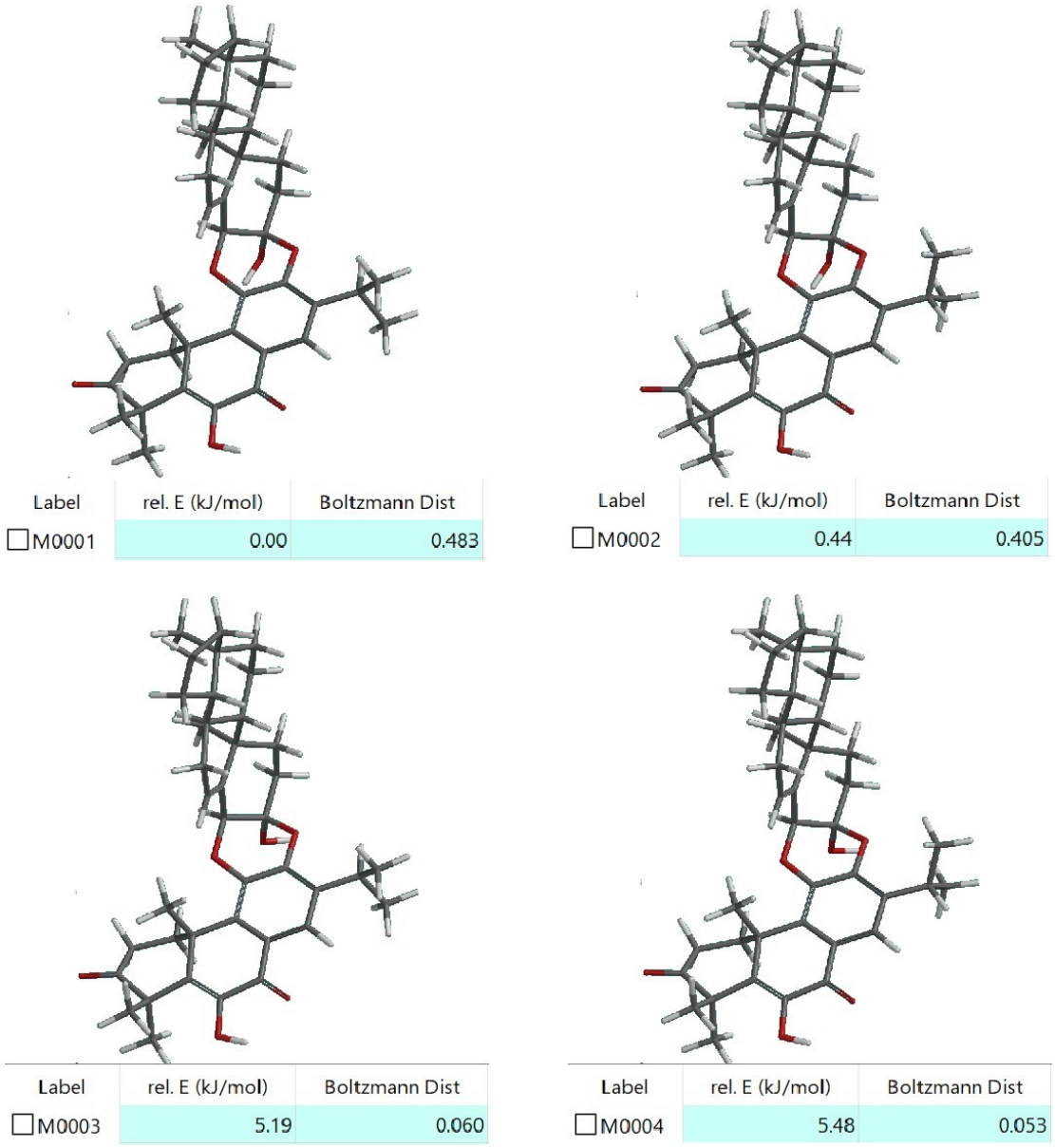


Figure S2 The optimized conformers of the simplified structure of **2**.

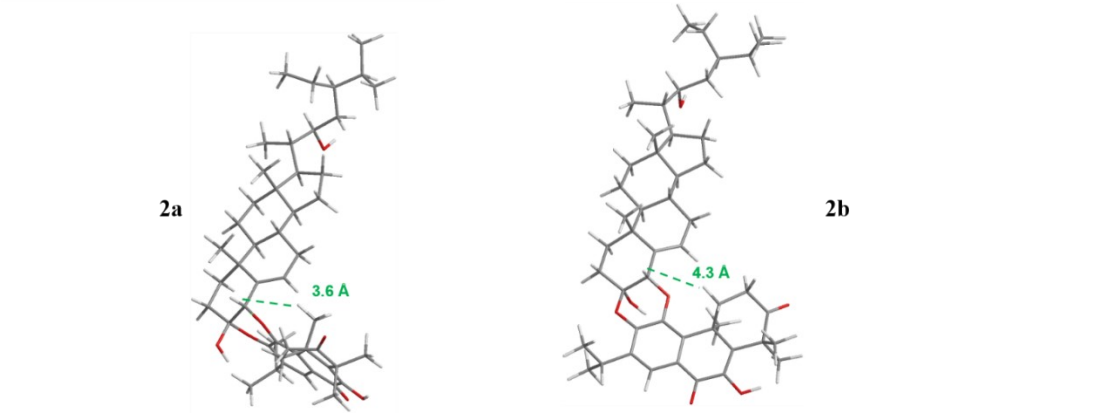


Figure S3 The optimized 3D structures of **2** with predicted interproton distance between H-4 and H_3-20′.

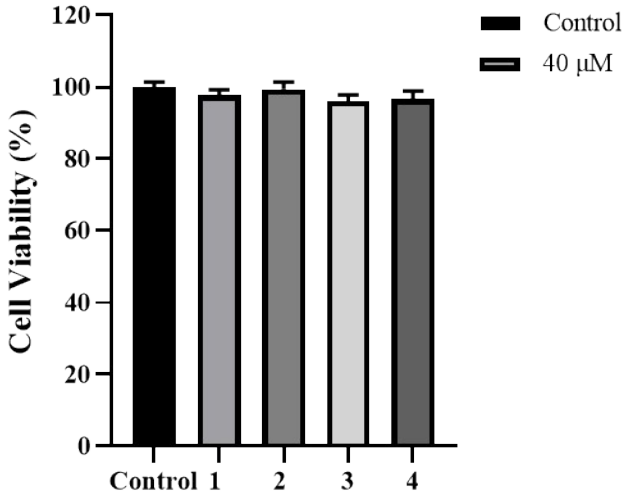


Figure S4 The viability of RAW 264.7 cells incubated with compounds **1**-**4** for 24 h.

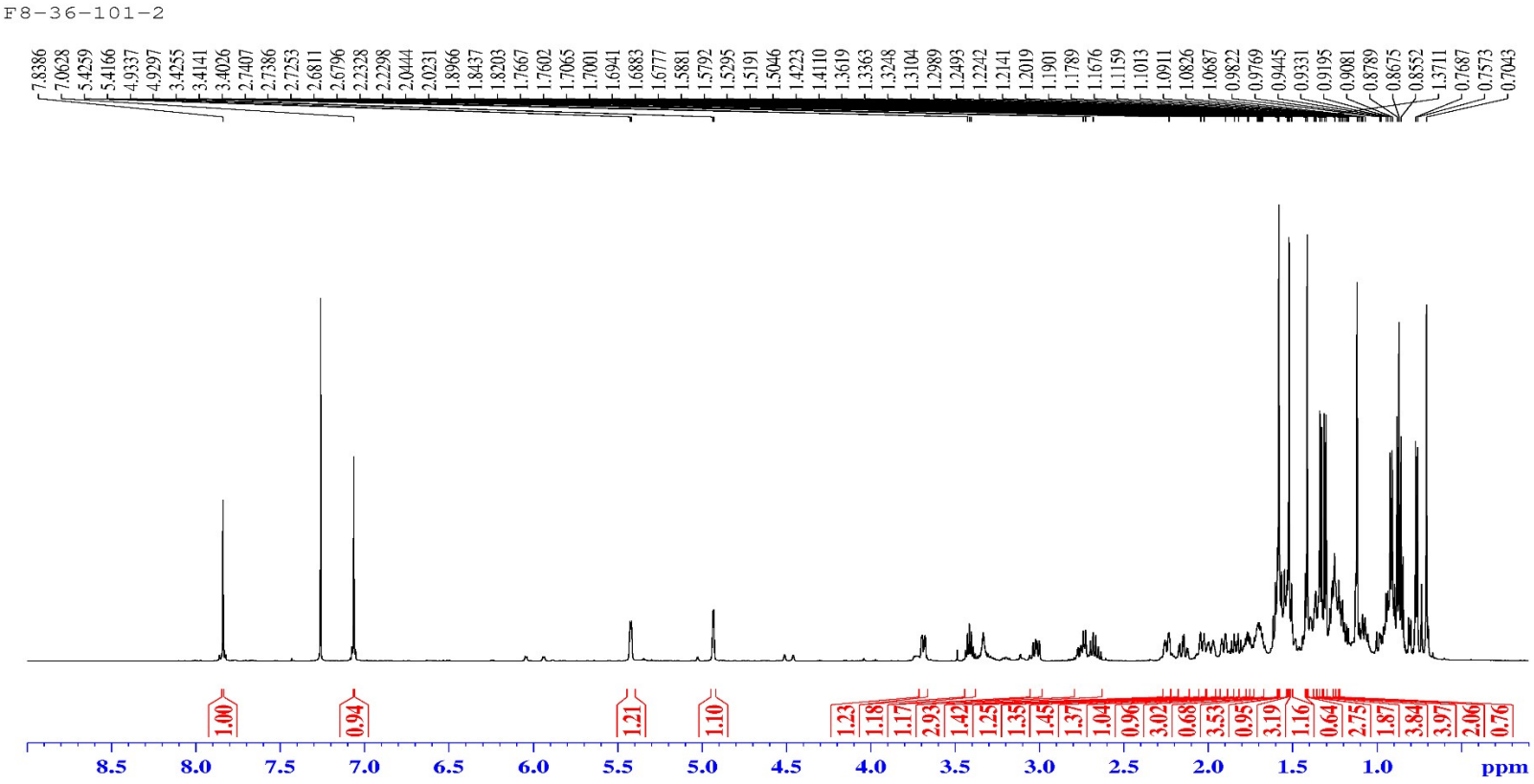
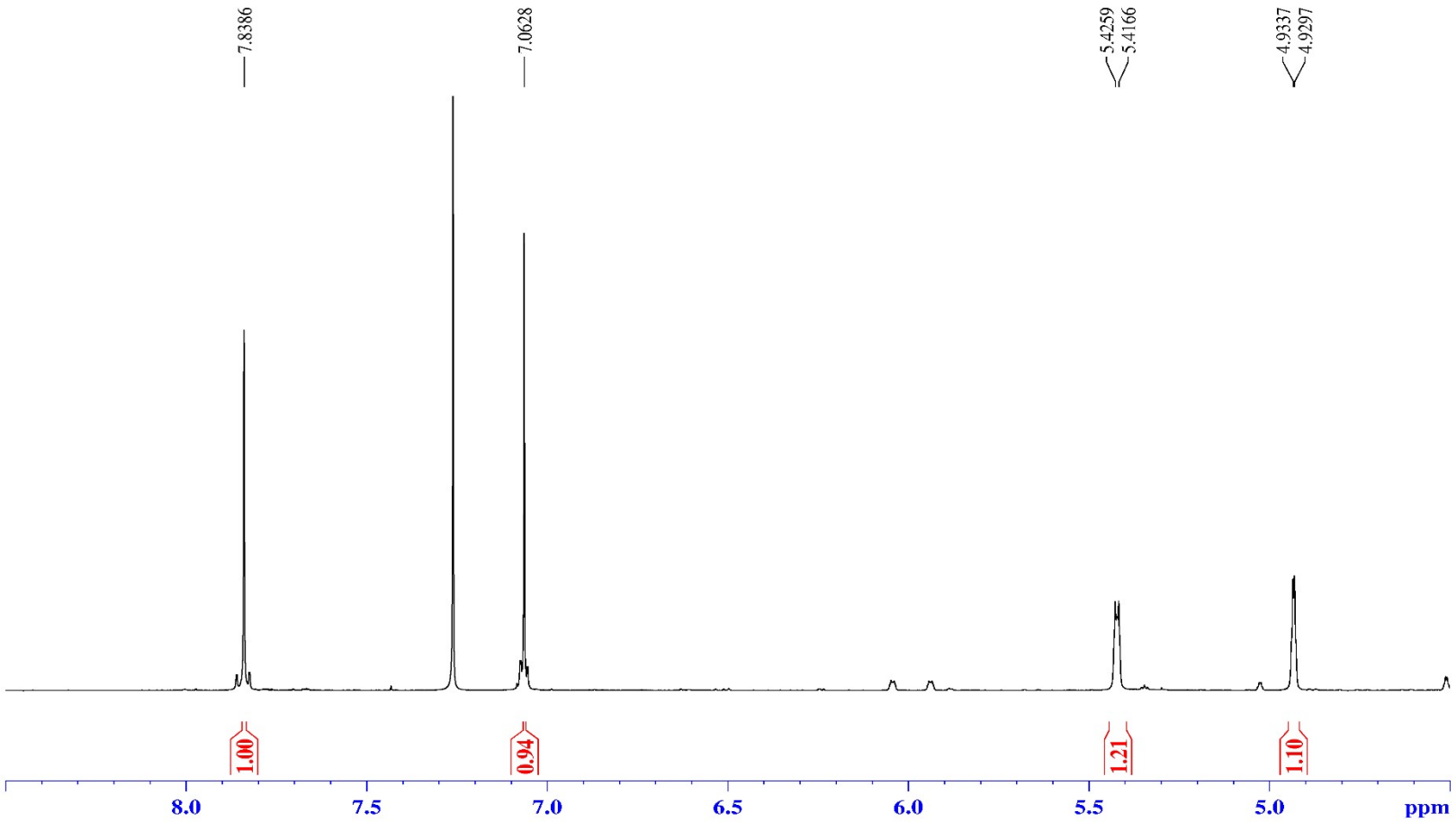


Figure S5 ^{1}H NMR spectrum (600 MHz, $CDCl_3$) of cephaloliverol A (**1**)

F8-36-101-2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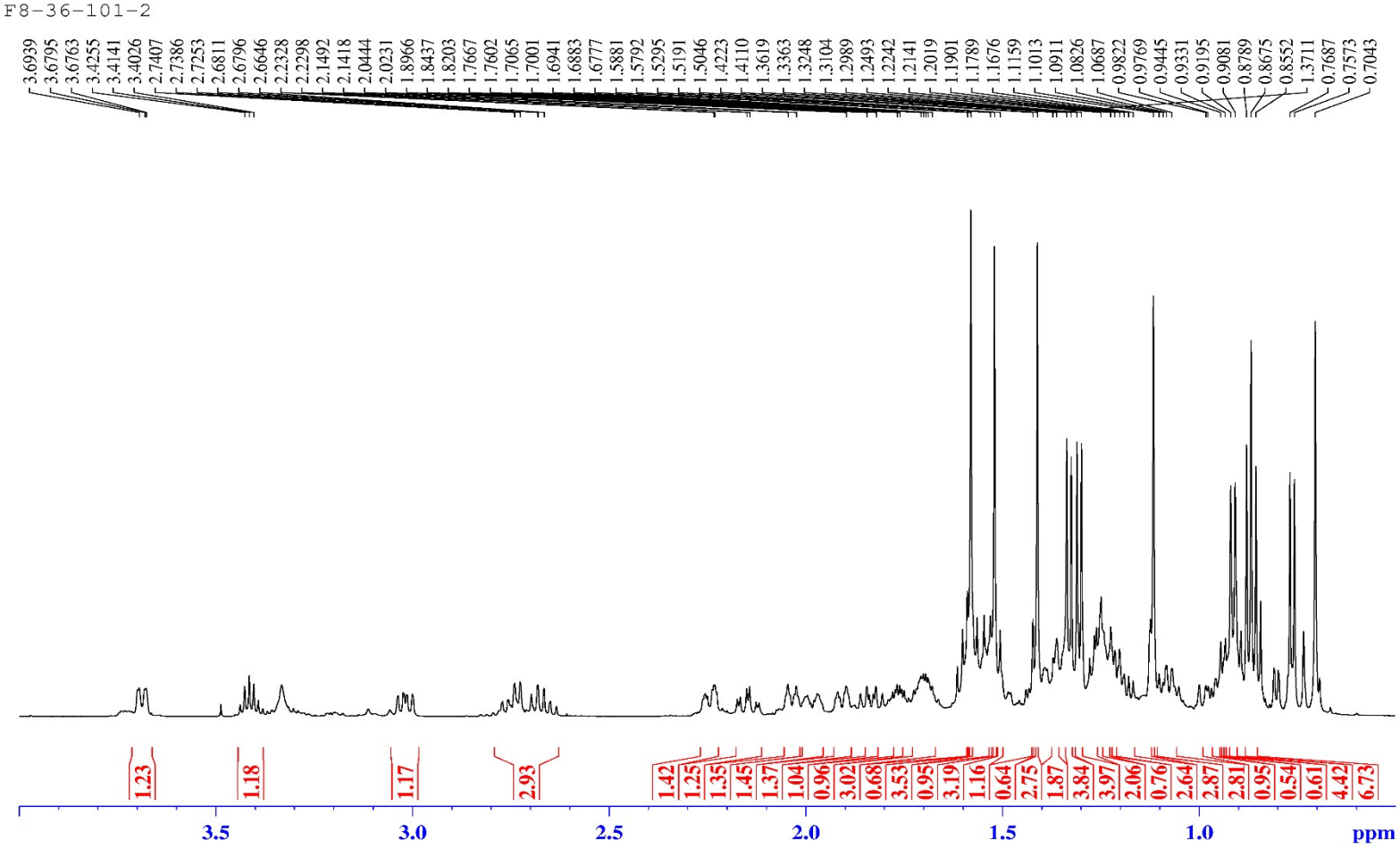
5.5
5.0
ppm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1.37
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0.95
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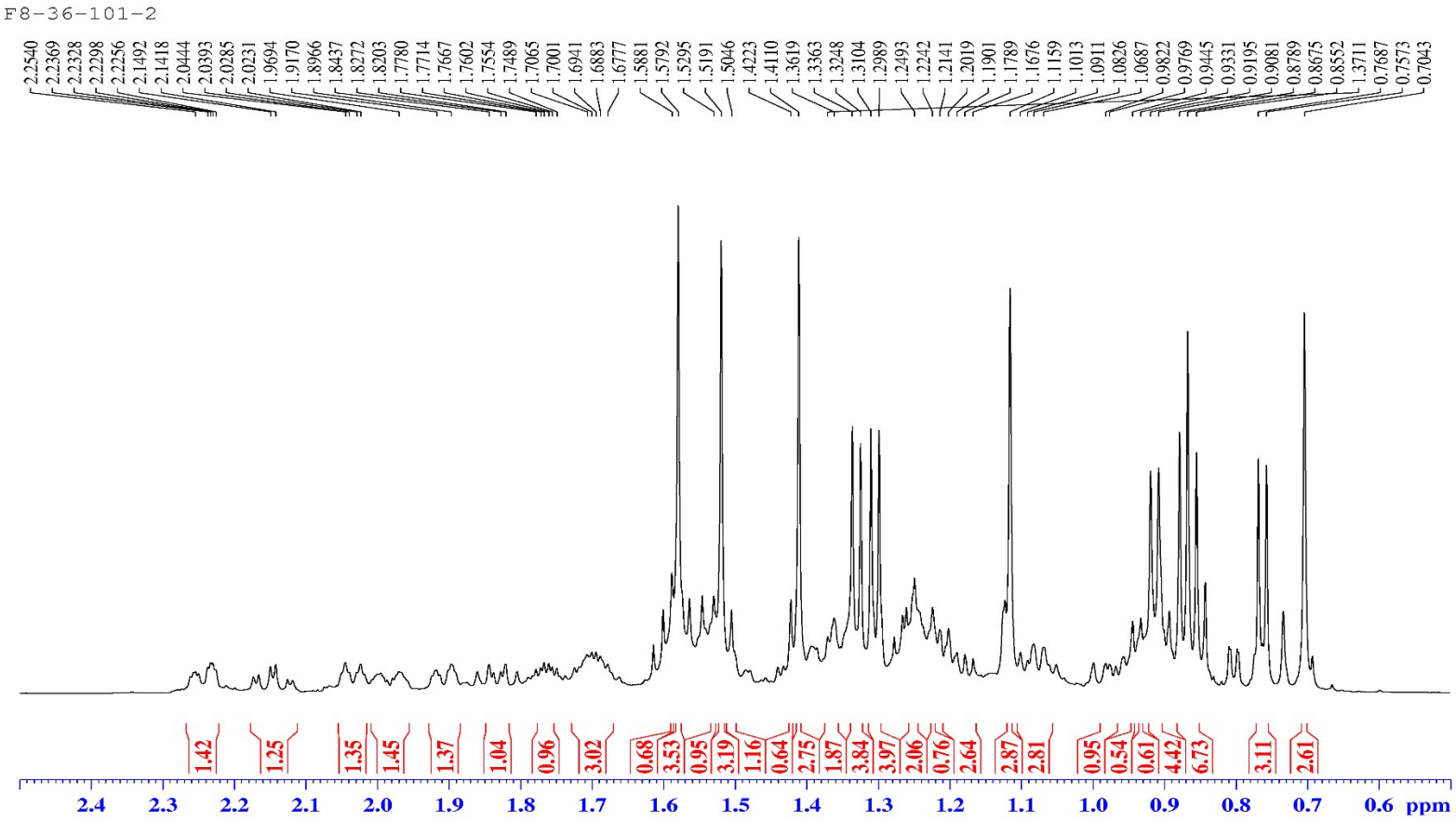

Figure S6 Enlarged ^{1}H NMR spectrum (600 MHz, $CDCl_3$) of cephaloliverol A (**1**)

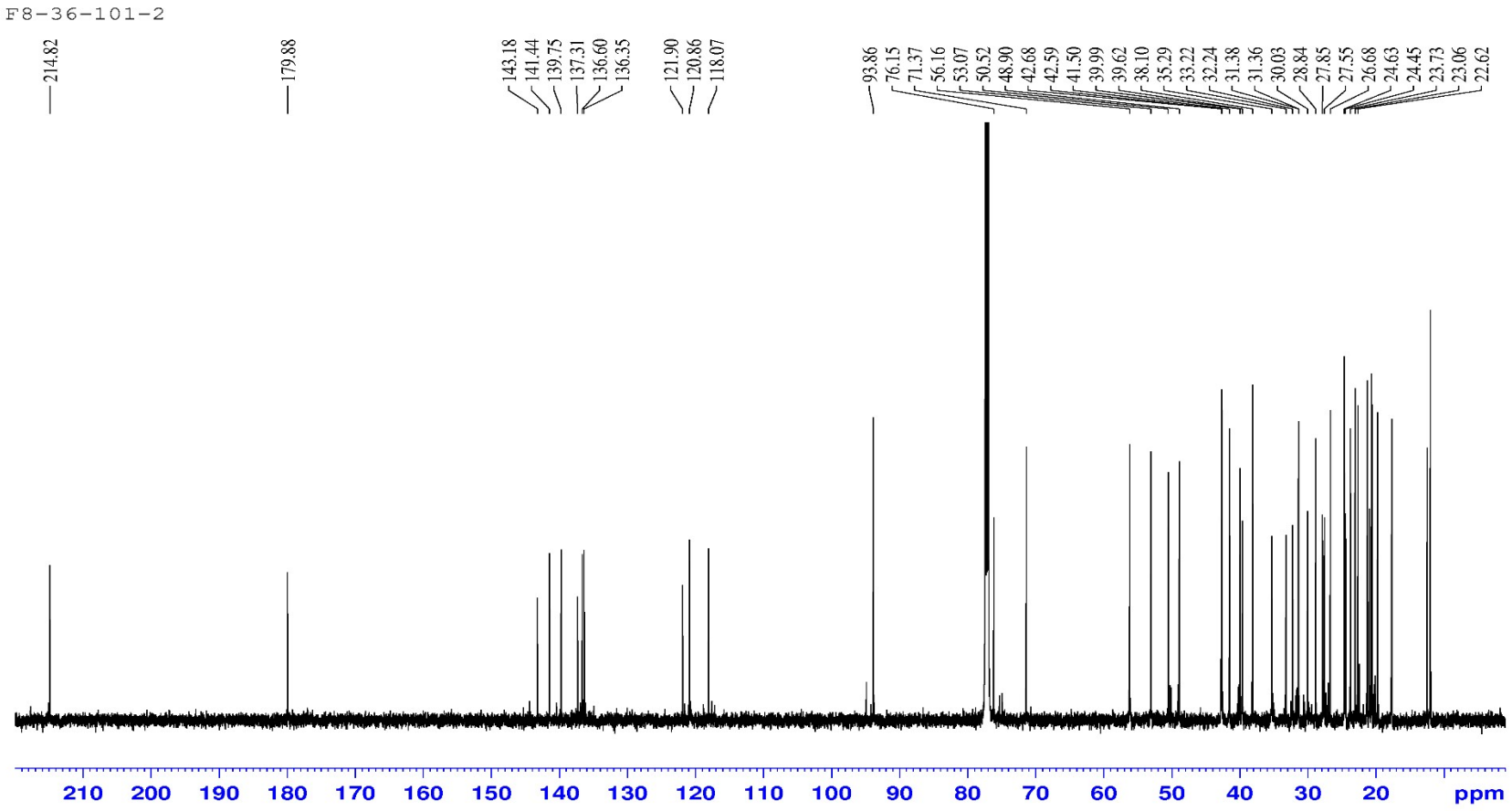

Figure S7 ^{13}C NMR spectrum (150 MHz, $CDCl_3$) of cephaloliverol A (**1**)

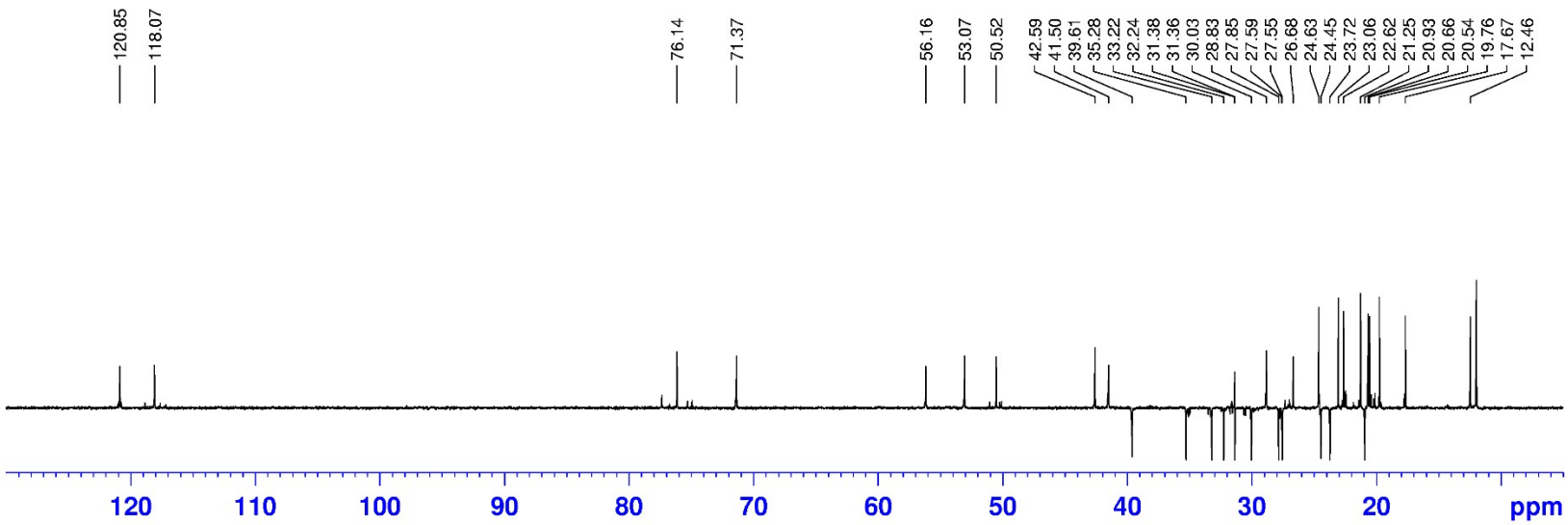

Figure S8 DEPT spectrum (150 MHz, $CDCl_3$) of cephaloliverol A (**1**)

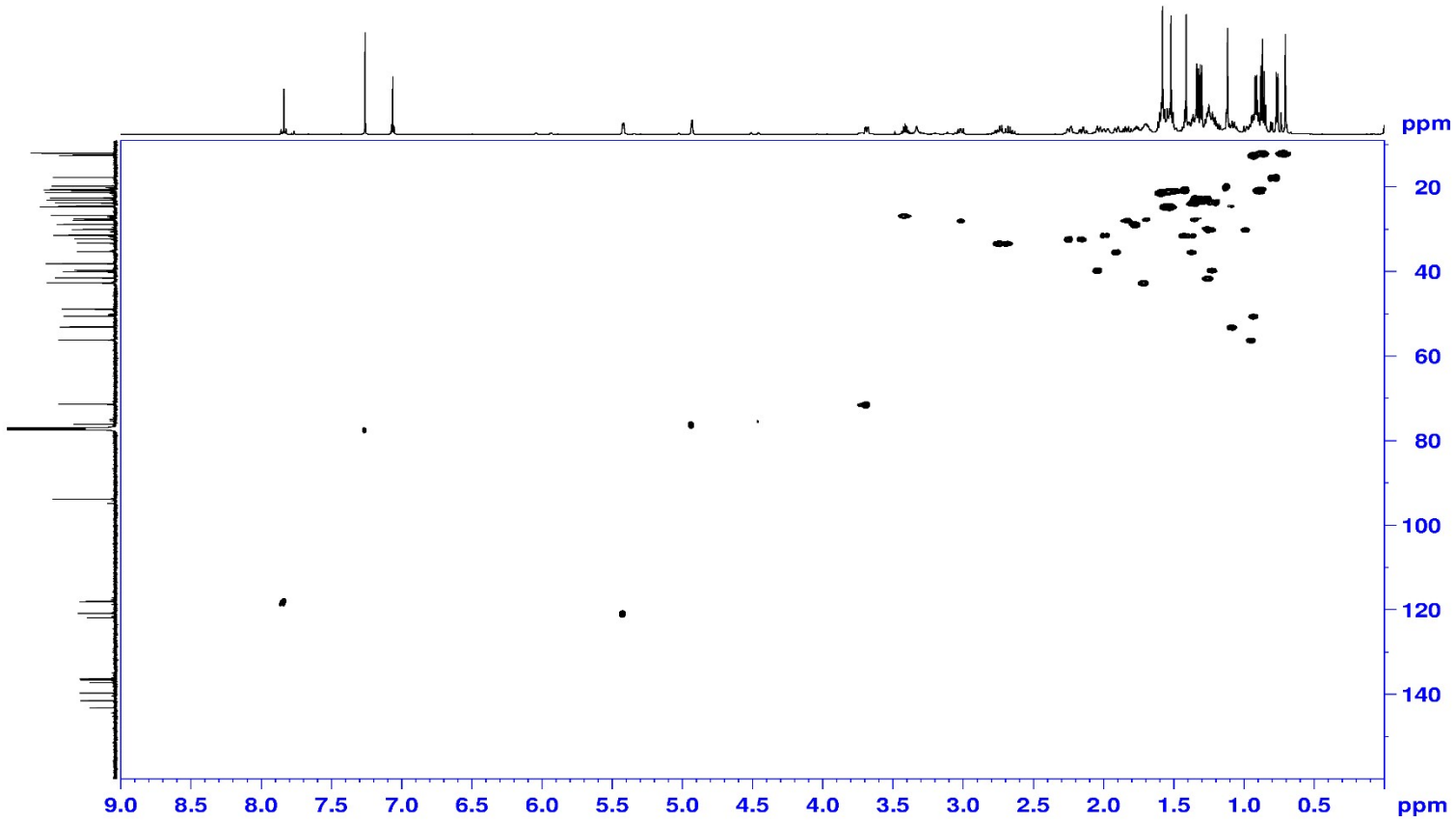

Figure S9 HSQC spectrum of cephaloliverol A (**1**)

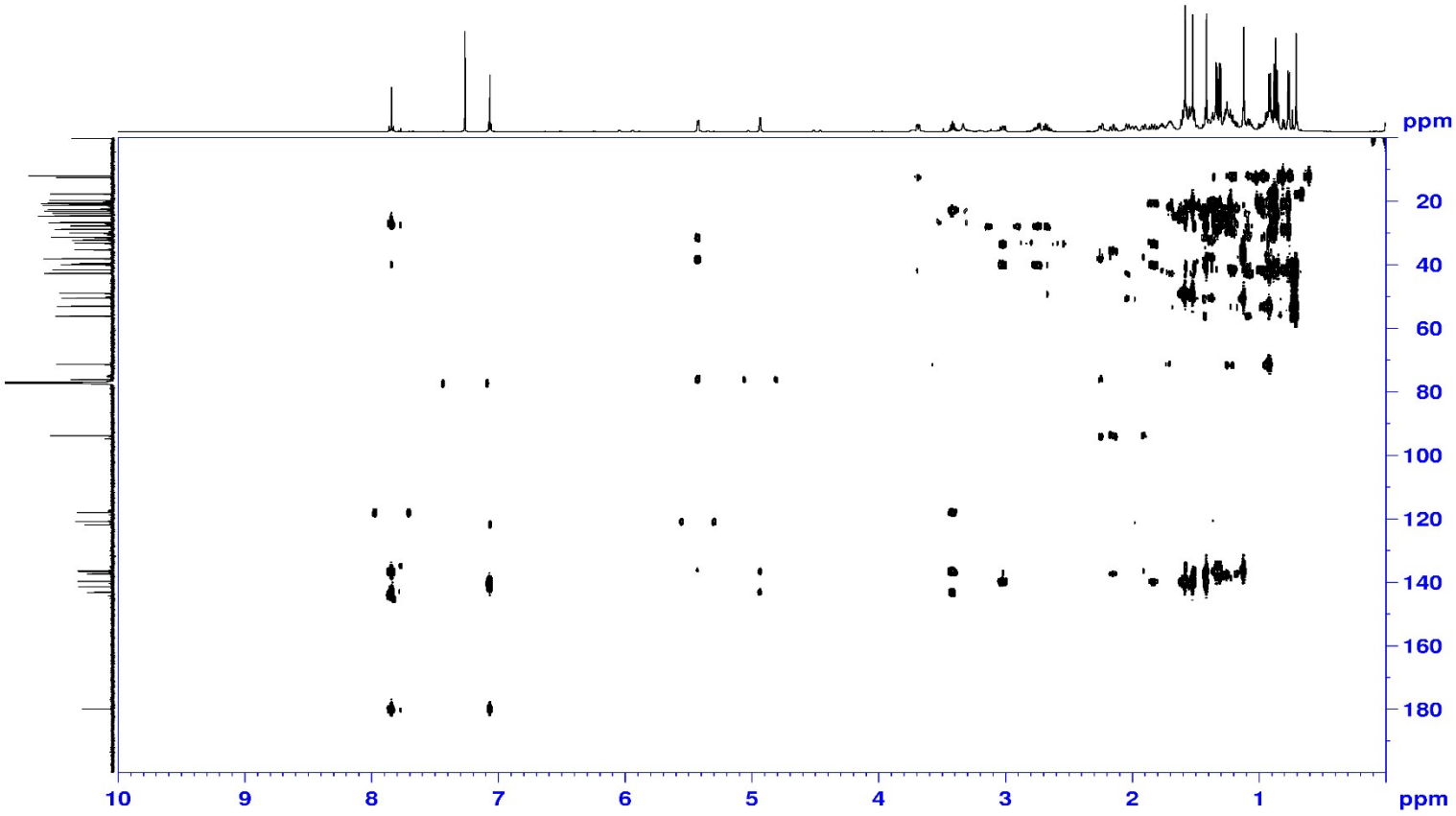

Figure S10 HMBC spectrum of cephaloliverol A (**1**)

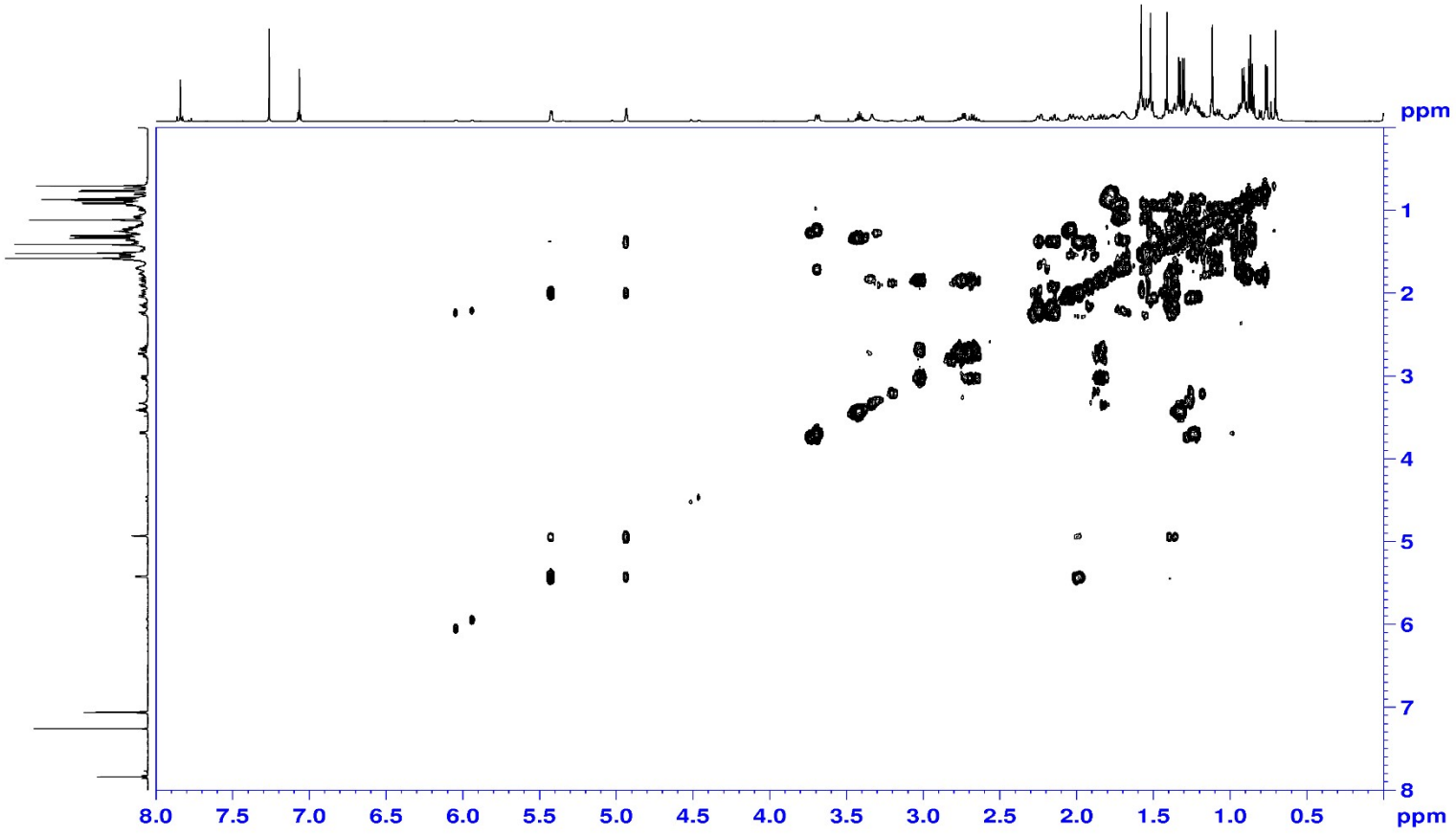

Figure S11 ^{1}H-^{1}H COSY spectrum of cephaloliverol A (**1**)

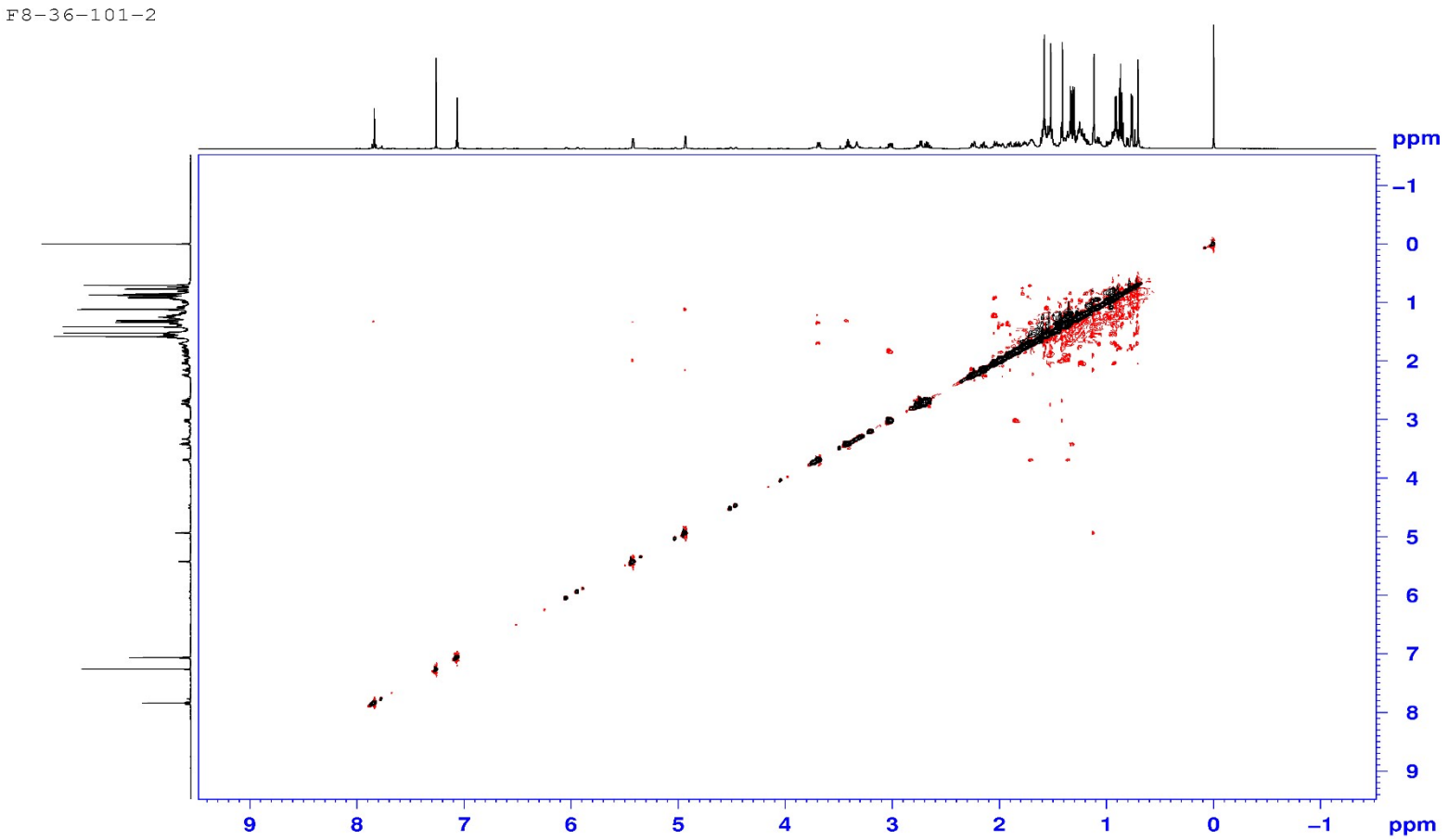

Figure S12 ROESY spectrum of cephaloliverol A (**1**)

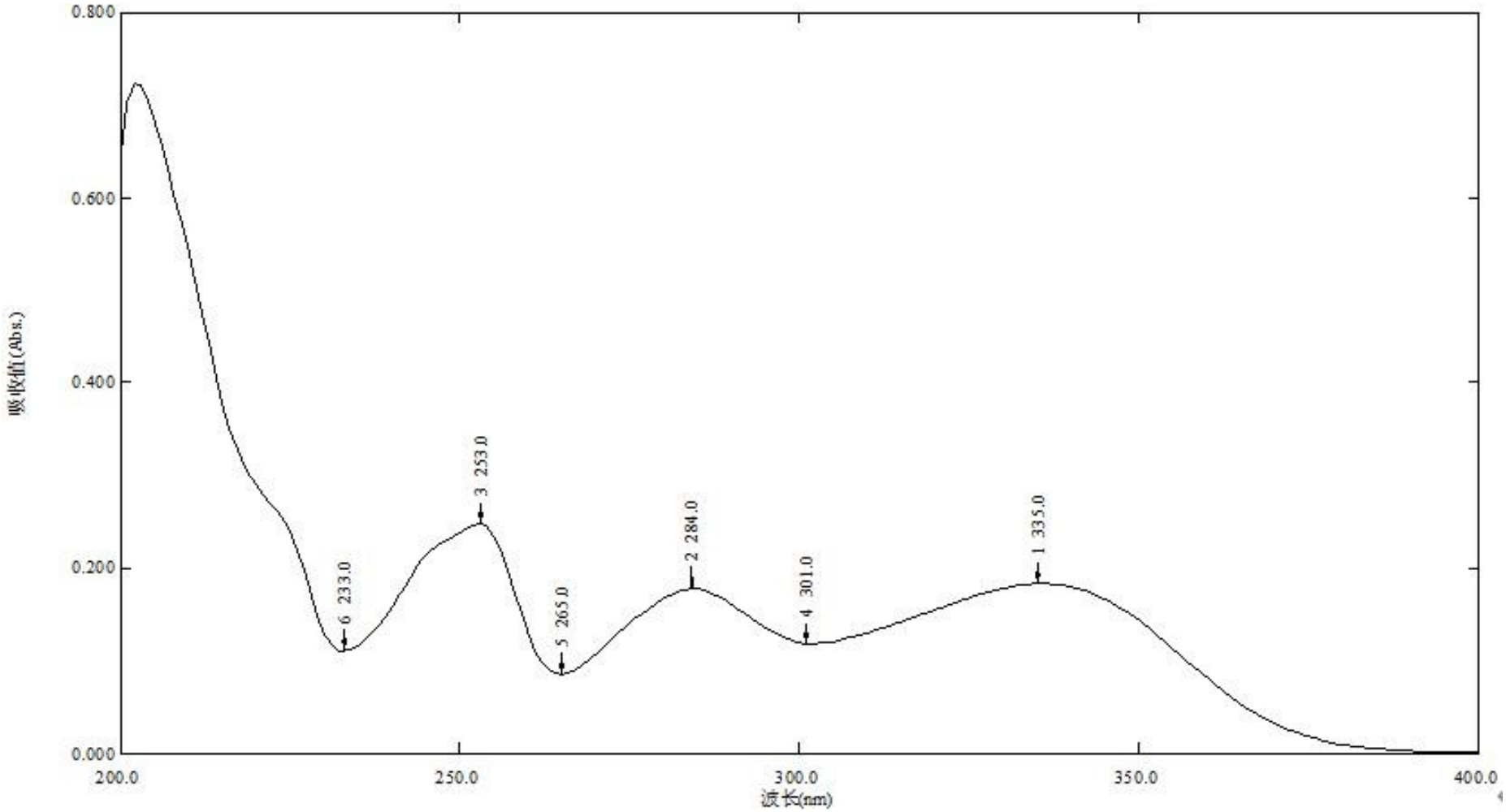

Figure S13 UV spectrum in CH_3OH of cephaloliverol A (**1**)

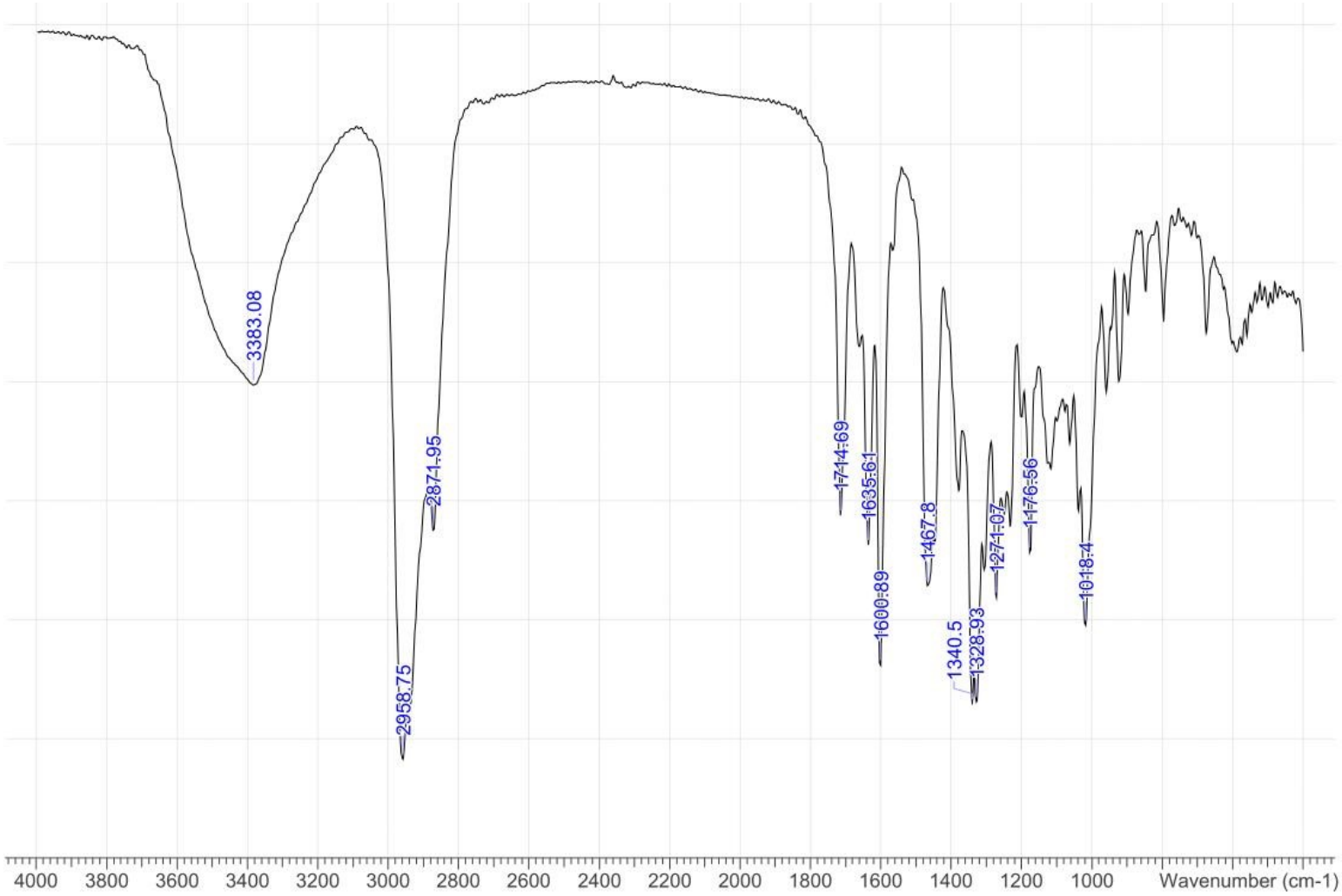

Figure S14 IR spectrum in KBr of cephaloliverol A (**1**)

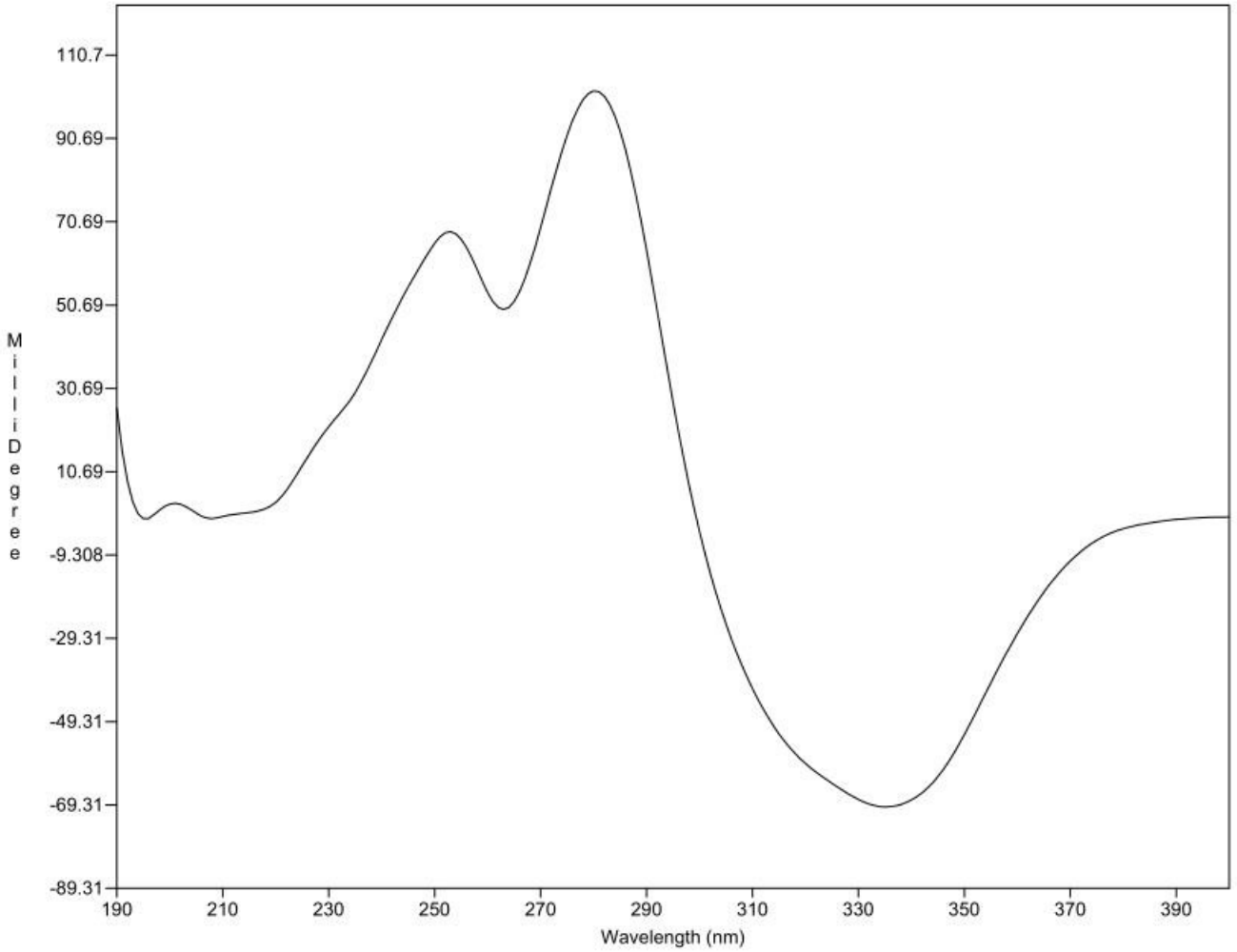

Figure S15 Experimental ECD spectrum in CH_3OH of cephaloliverol A (**1**)

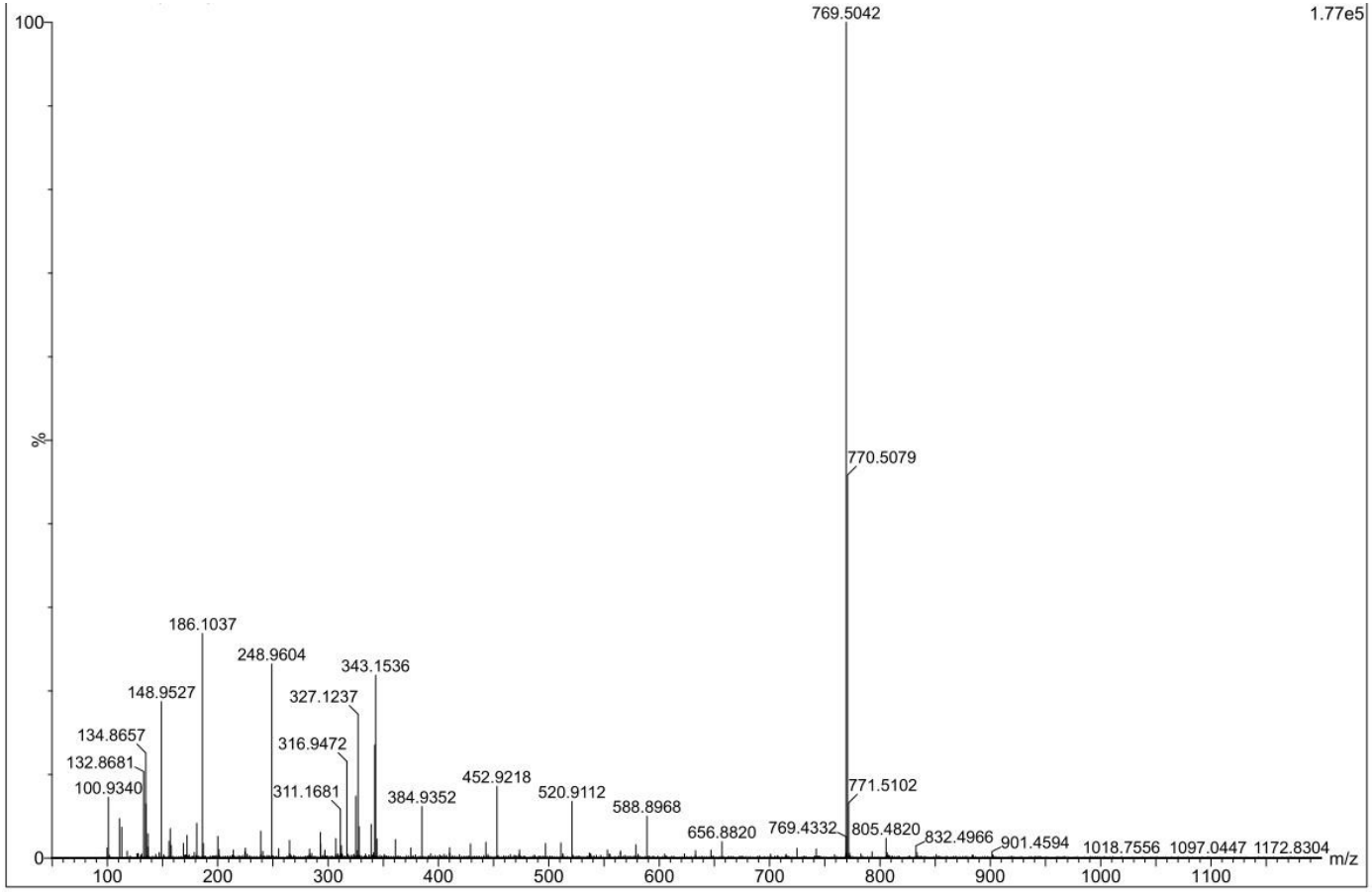

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Figure S16 HRESIMS of cephaloliverol A (**1**)

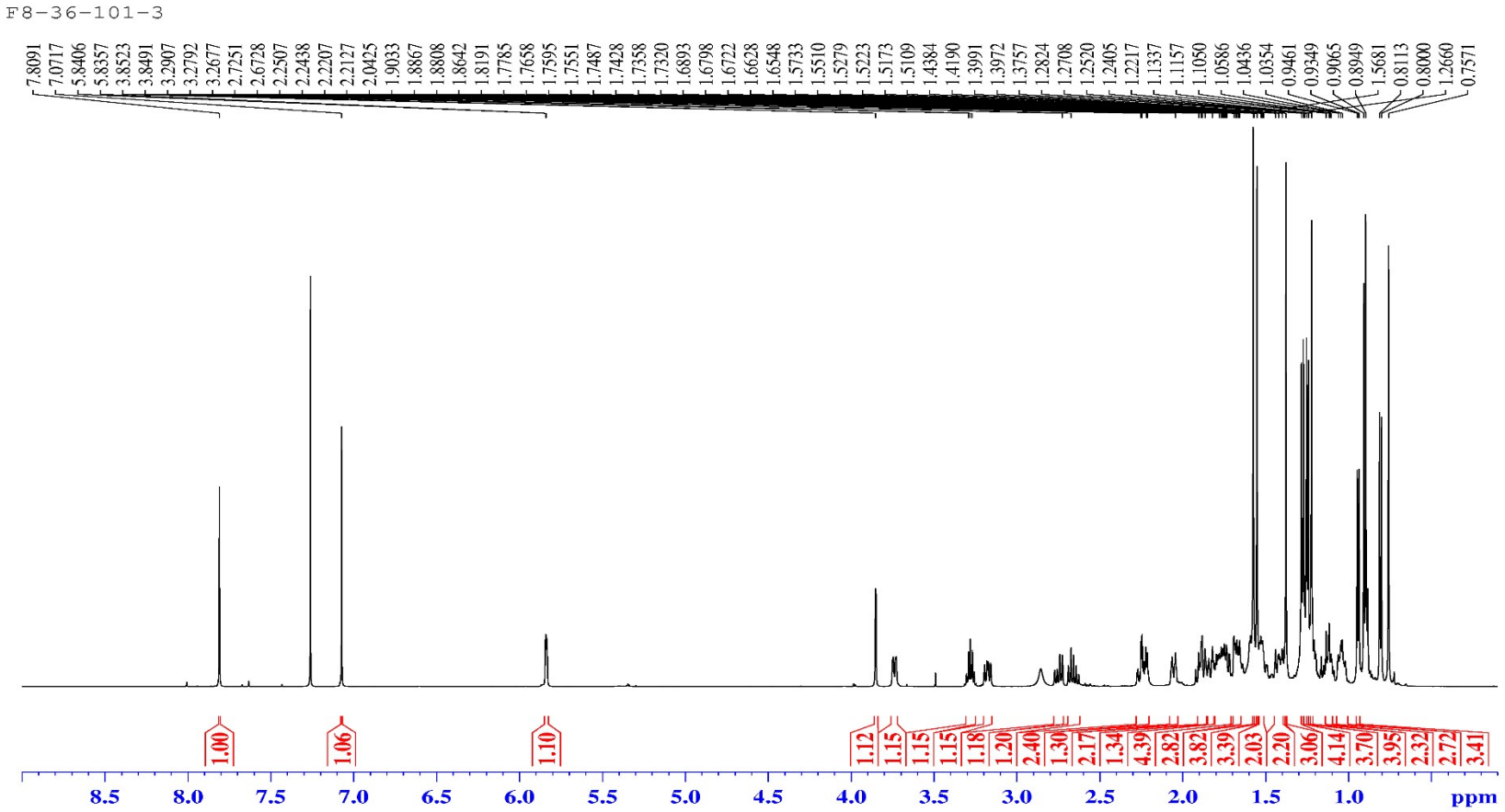
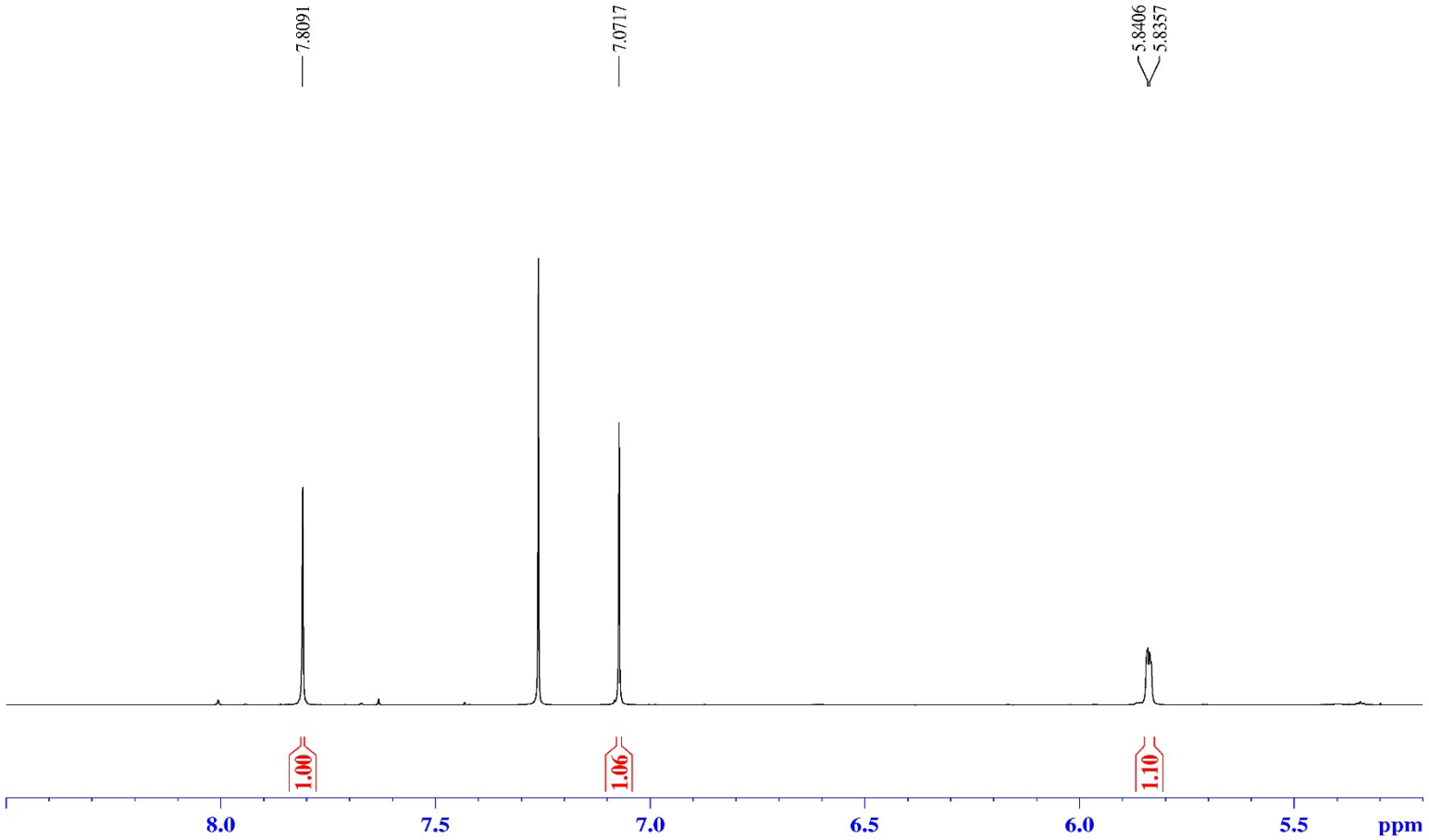
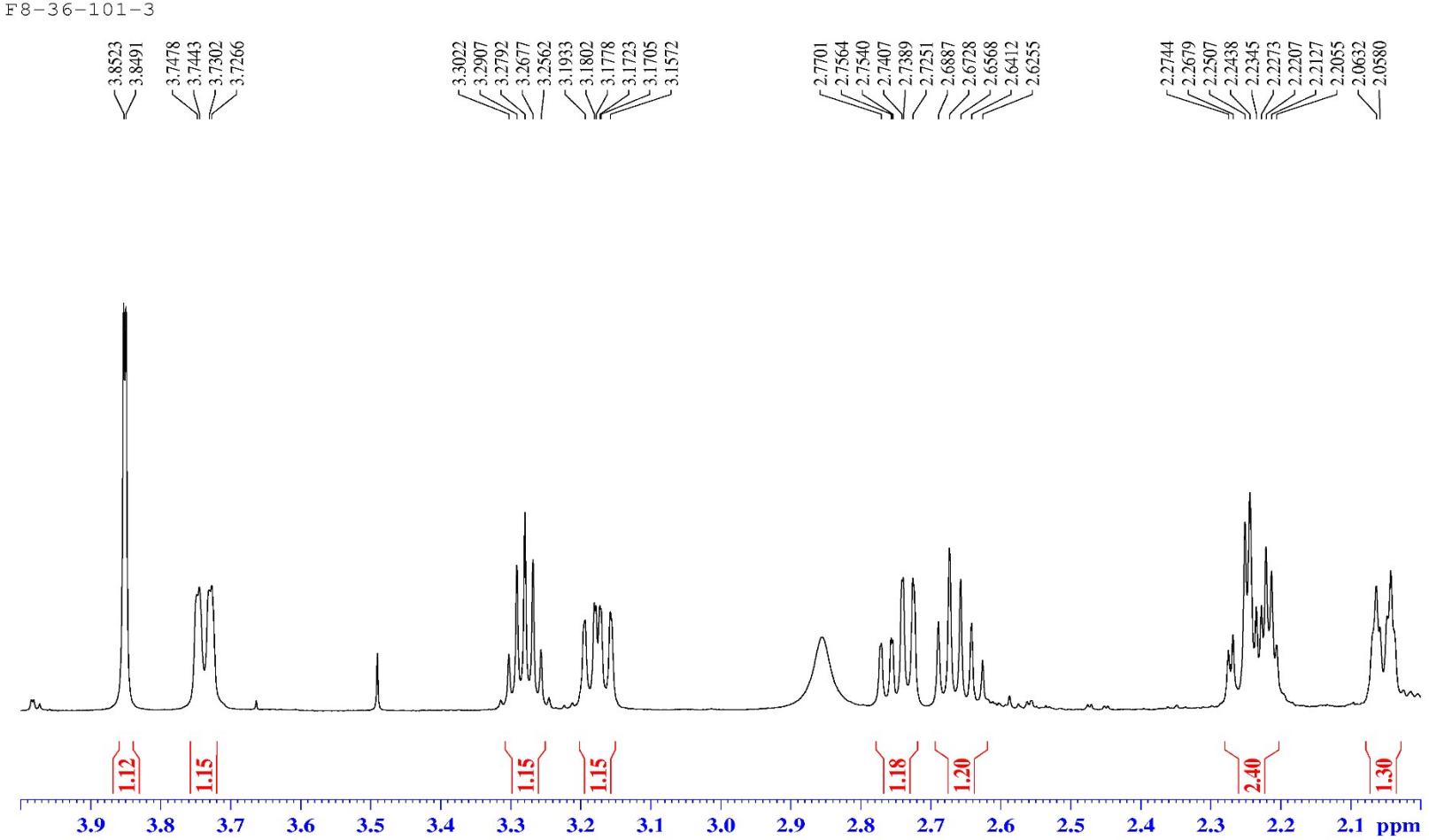


Figure S17 ^{1}H NMR spectrum (600 MHz, $CDCl_3$) of cephaloliverol B (**2**)

F8-36-101-3

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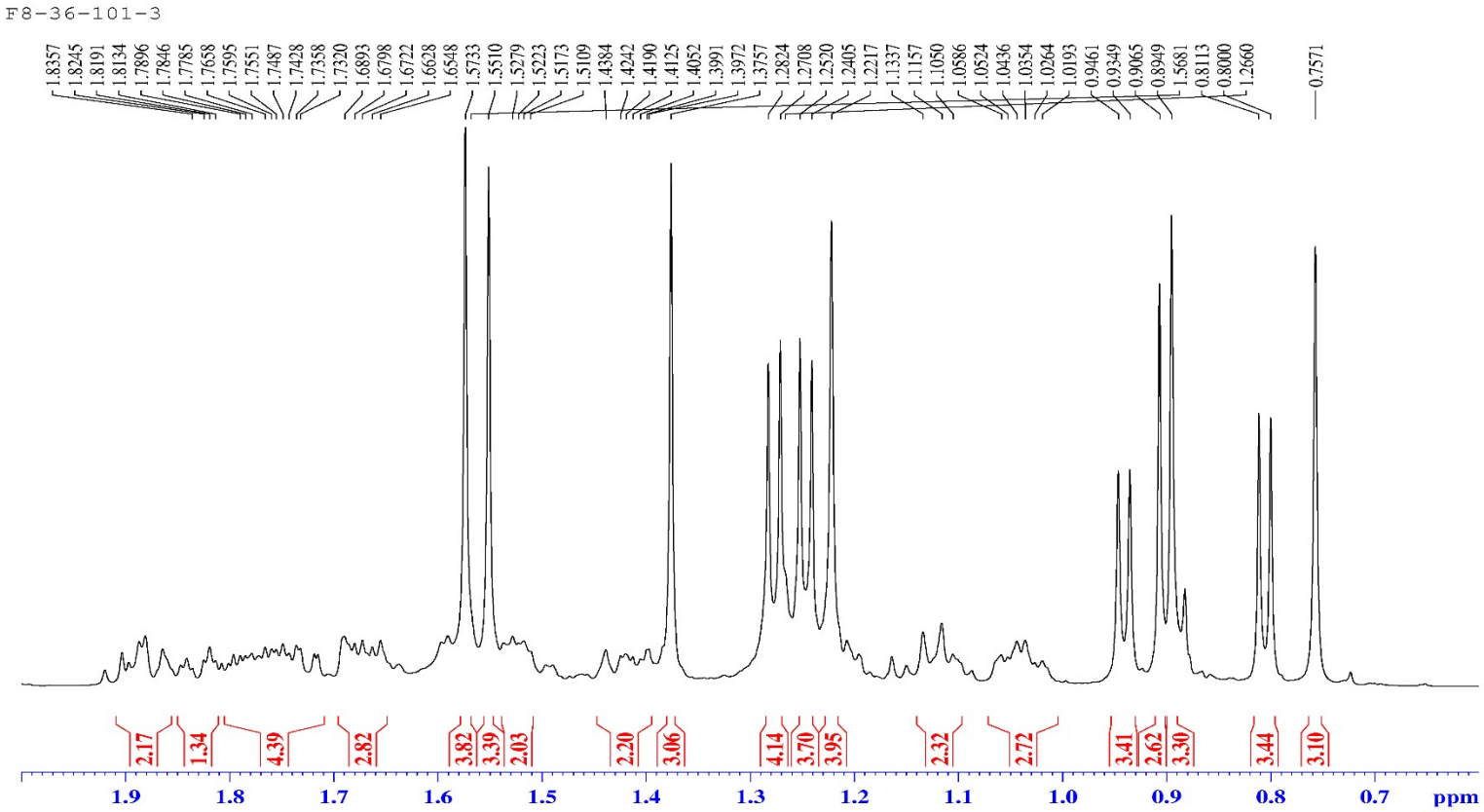


Figure S18 Enlarged ^{1}H NMR spectrum (150 MHz, $CDCl_3$) of cephaloliverol B (**2**)

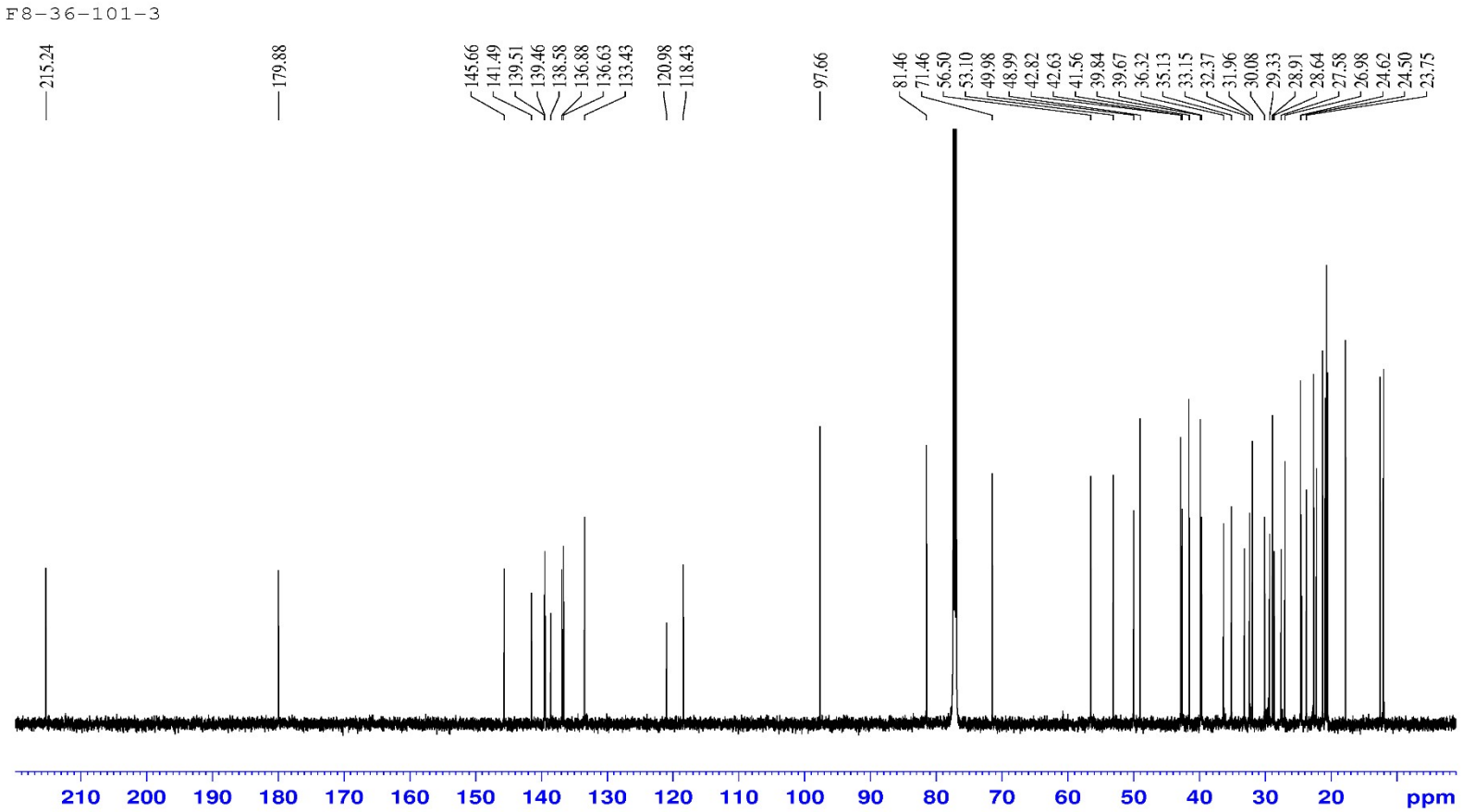


Figure S19 ^{13}C NMR spectrum (150 MHz, $CDCl_3$) of cephaloliverol B (**2**)

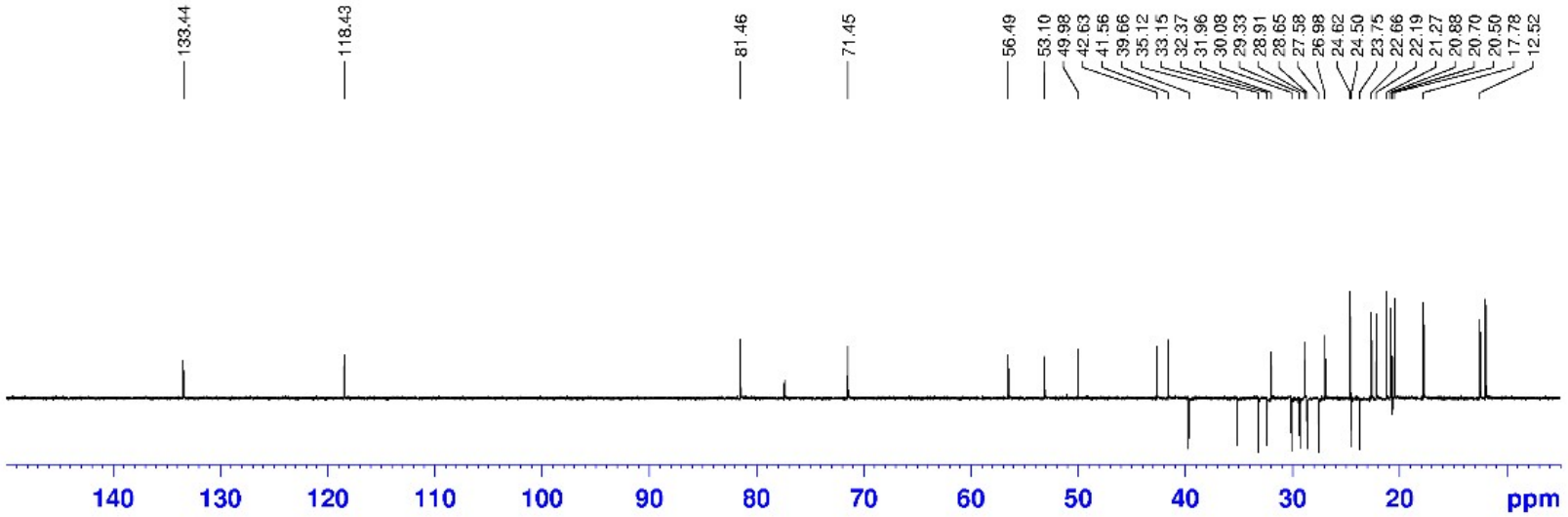


Figure S20 DEPT spectrum (150 MHz, $CDCl_3$) of cephaloliverol B (**2**)

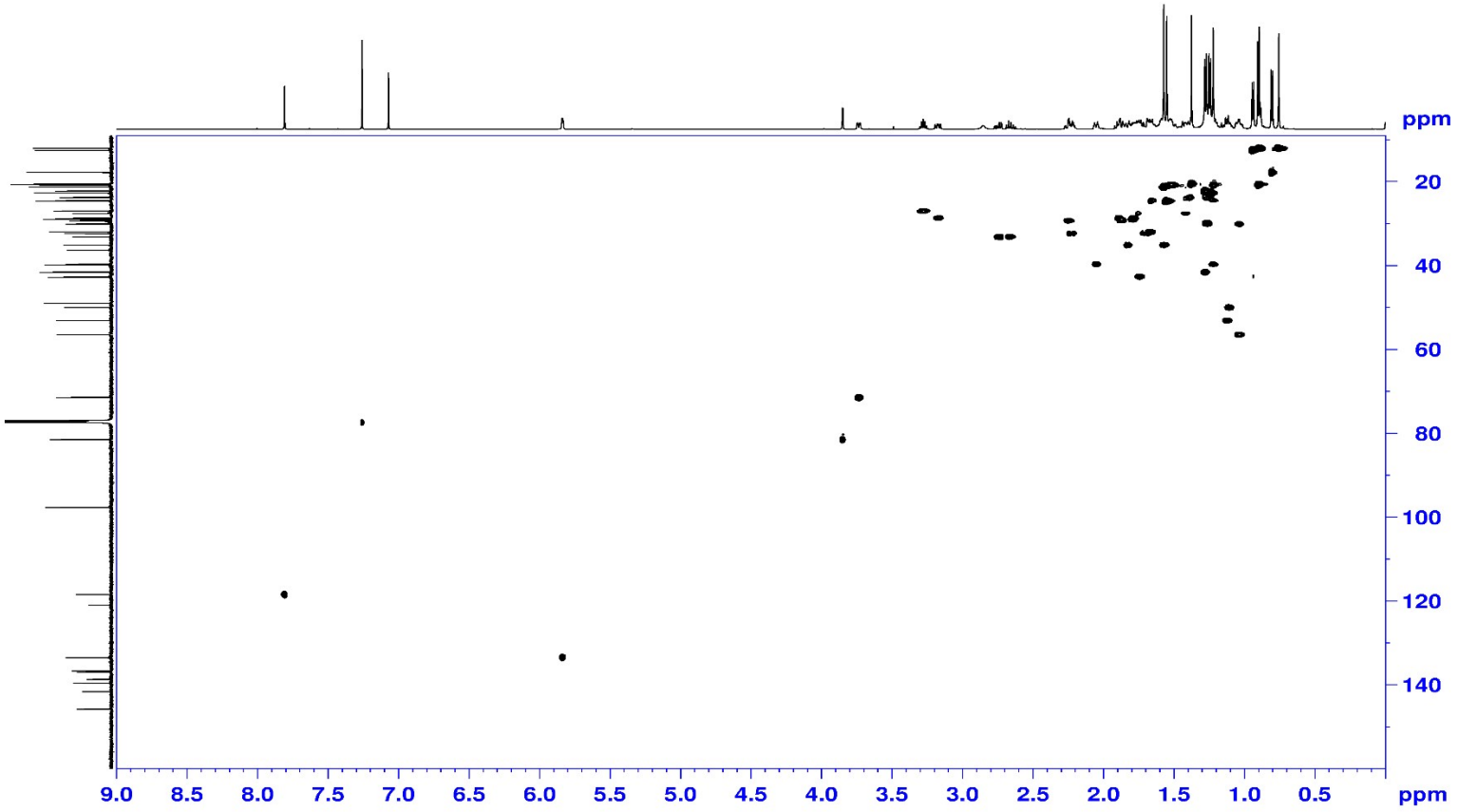


Figure S21 HSQC spectrum of cephaloliverol B (**2**)

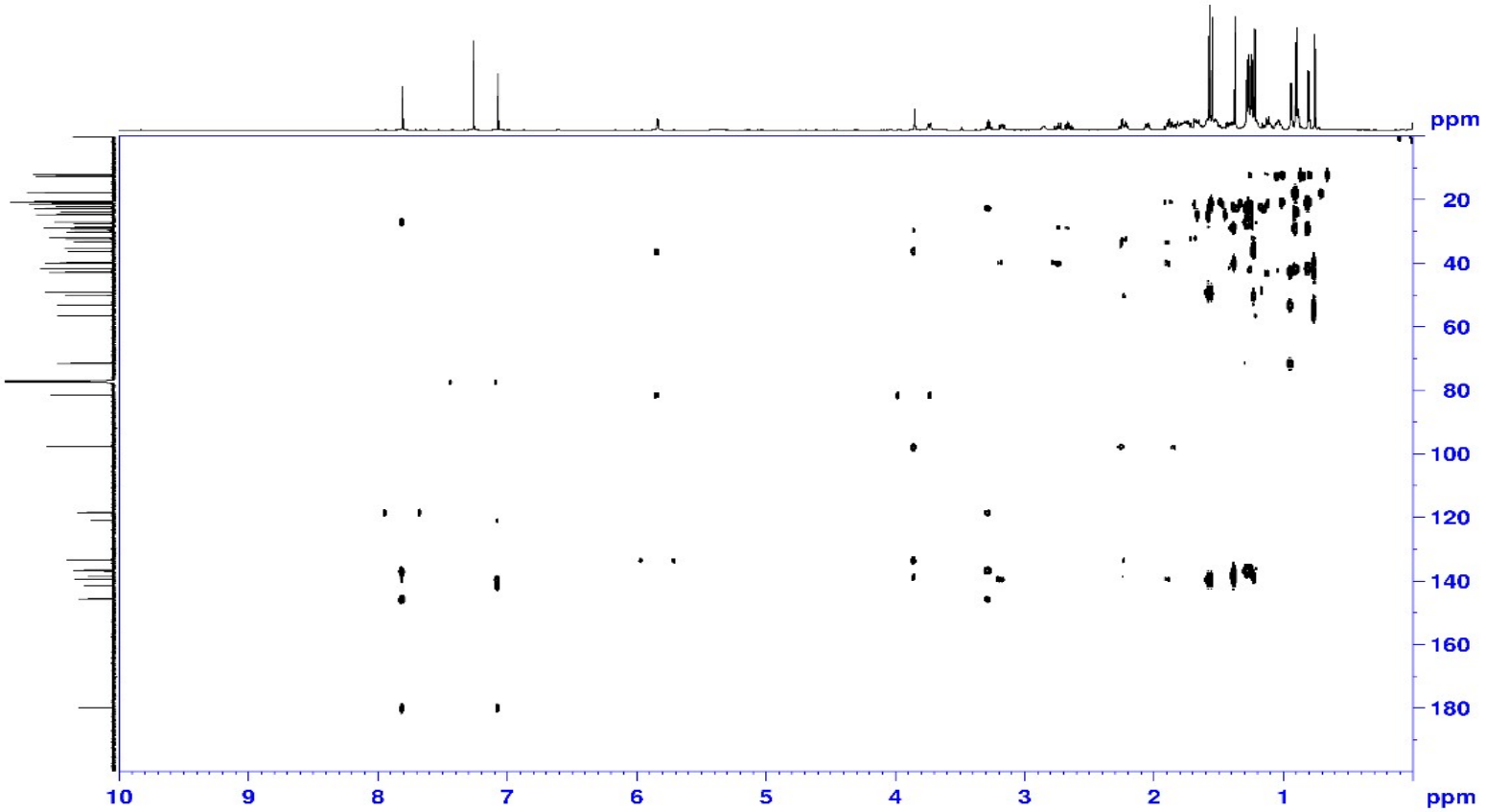


Figure S22 HMBC spectrum of cephaloliverol B (**2**)

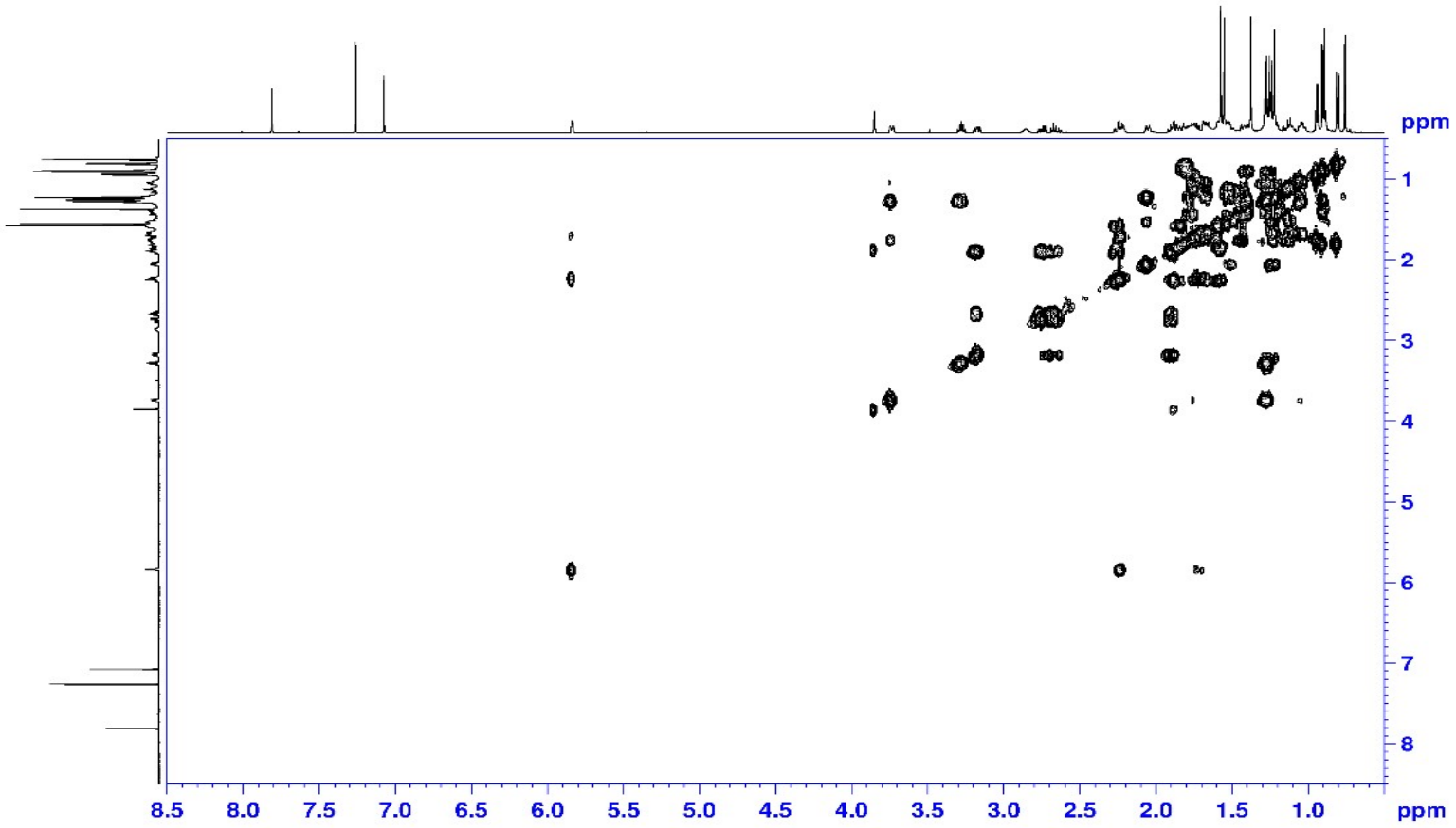


Figure S23 ^{1}H-^{1}H COSY spectrum of cephaloliverol B (**2**)

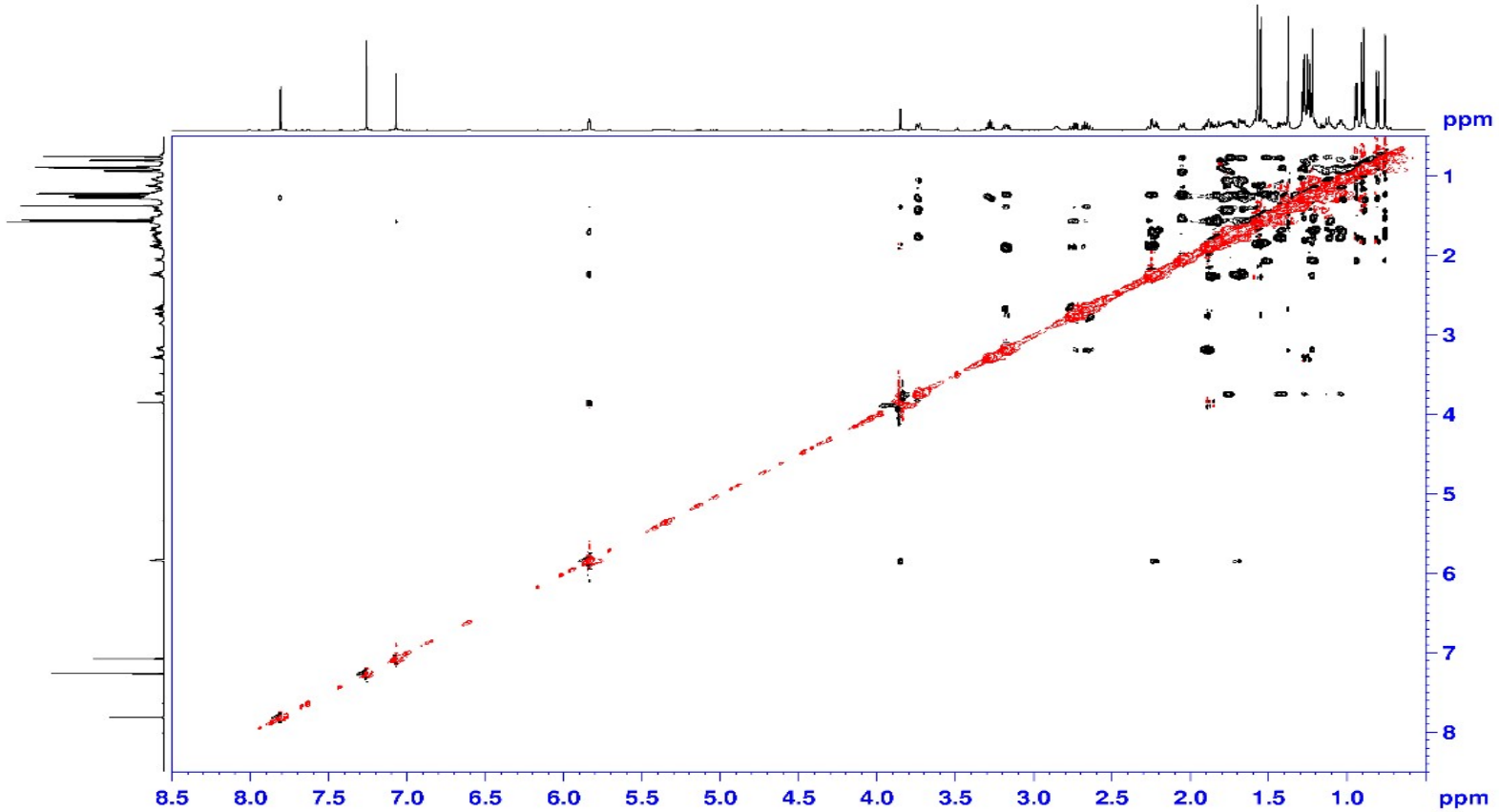


Figure S24 ROESY spectrum of cephaloliverol B (**2**)

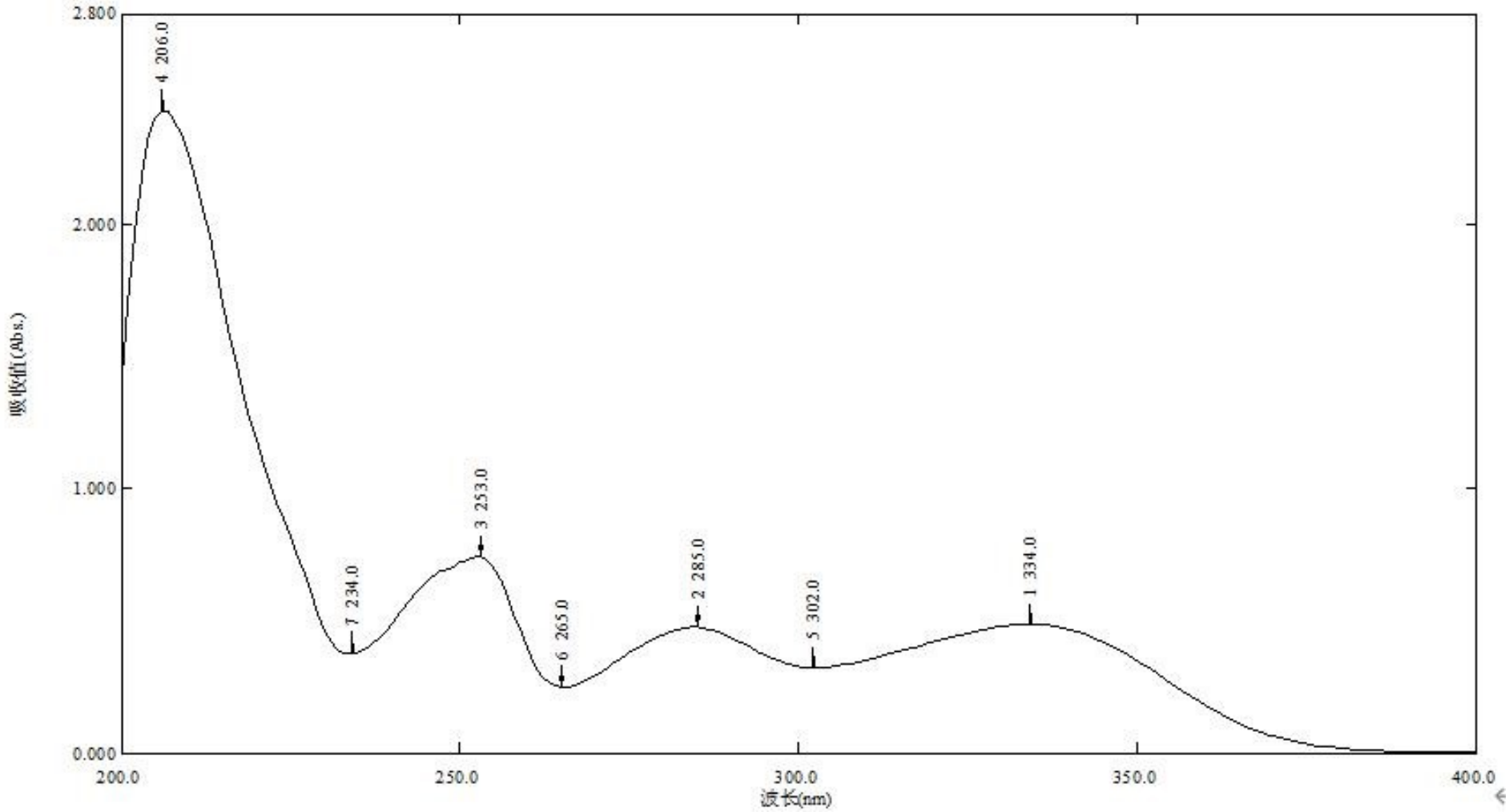


Figure S25 UV spectrum in CH_3OH of cephaloliverol B (**2**)

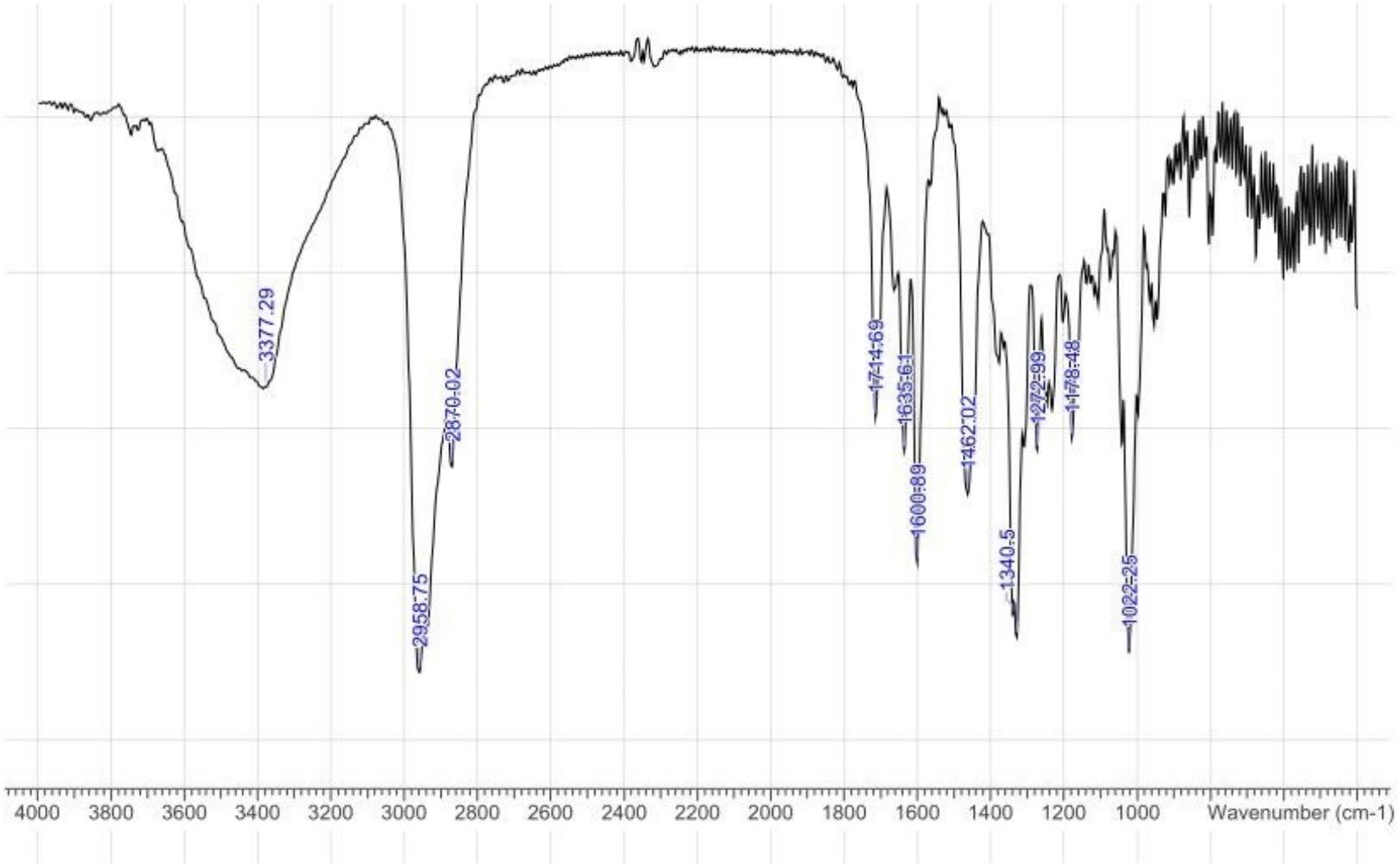


Figure S26 IR spectrum in KBr of cephaloliverol B (**2**)

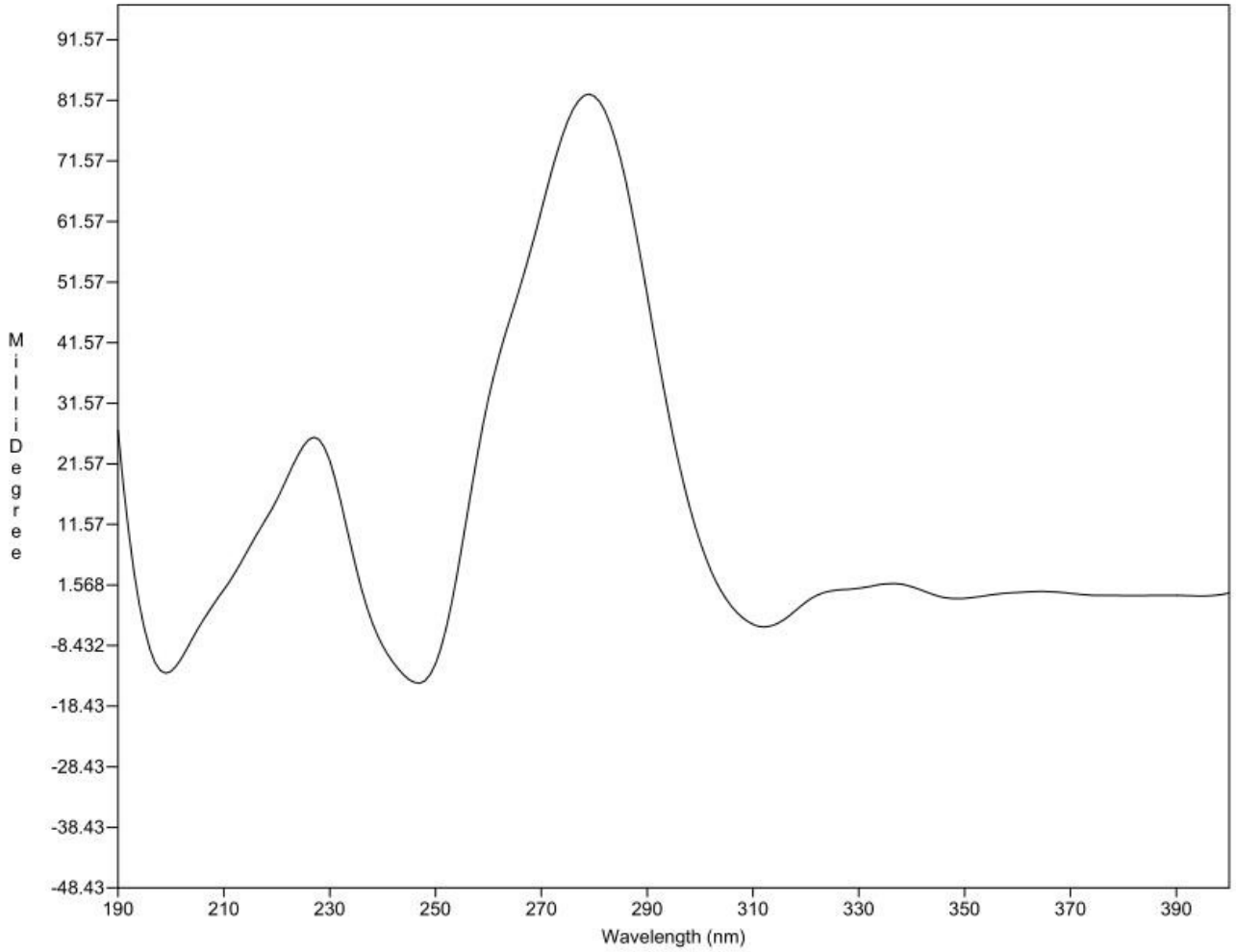
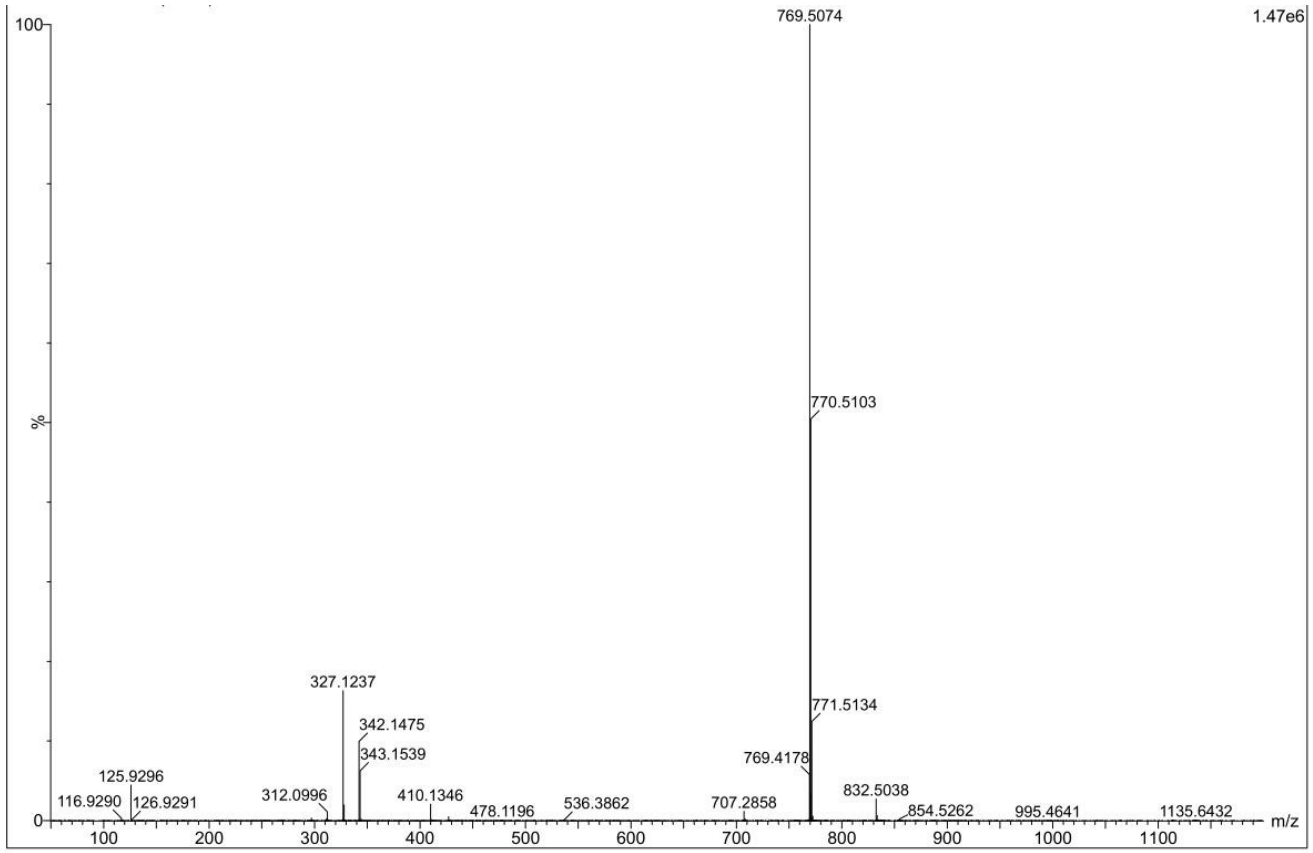


Figure S27 Experimental ECD spectrum in CH_3OH of cephaloliverol B (**2**)



Mass	Calc. Mass	mDa	PPM	DBE	i-FIT	Norm	Conf(%)	Formula
769.5074	769.5043	3.1	4.0	15.5	446.8	2.325	9.78	C49 H69 O7

Figure S28 HRESIMS of cephaloliverol B (**2**)