Supplementary Material

Bioassay-Guided Isolation of Two Eudesmane Sesquiterpenes from *Lindera strychnifolia* Using Centrifugal Partition Chromatography

Ji Hoon Kim [1,†], Je-Seung Jeon [1,†], Jung Hoon Kim [1], Eun Ju Jung [1], Yun Jung Lee [1], En Mei Gao [1], Ahmed Shah Syed [2], Rak Ho Son [1,3] and Chul Young Kim [1,*]

[1] College of Pharmacy and Institute of Pharmaceutical Science and Technology, Hanyang University, Ansan 15588, Korea; gg890718@gmail.com (J.H.K.); jsjeoncy@gmial.com (J.-S.J.); evveje@naver.com (J.H.K.); jejs2@naver.com (E.J.J.); sopihya@naver.com (Y.J.L.); rhdmsal@hanyang.ac.kr (E.M.G.); sonnaco@huons.com (R.H.S.)

[2] Department of Pharmacognosy, Faculty of Pharmacy, University of Sindh, Jamshoro 76080, Pakistan; shahahmed454@gmail.com

[3] R&D Center, Huons Co. Ltd., Ansan 15588, Korea

* Correspondence: chulykim@hanyang.ac.kr; Tel.: +82-31-400-5809

† These authors contributed equally to this work.

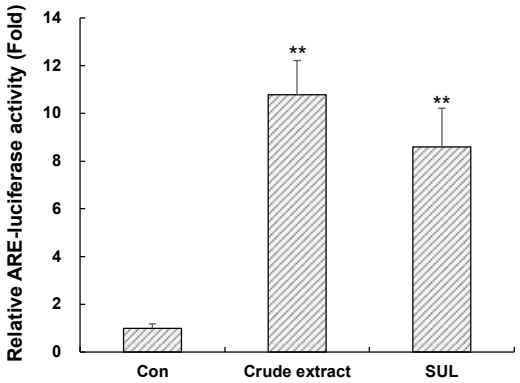

Figure S1. The relative ARE-luciferase activity of *Lindera strychnifolia* extract. ARE induction activity was evaluated in ARE-HepG2 cells at concentration of 30 μg/mL. Sulforaphane (SUL, 5μM) was used as positive control. Data are presented as means ± S.E. (n = 3). **, $P < 0.01$ (compared with the vehicle-treated control).

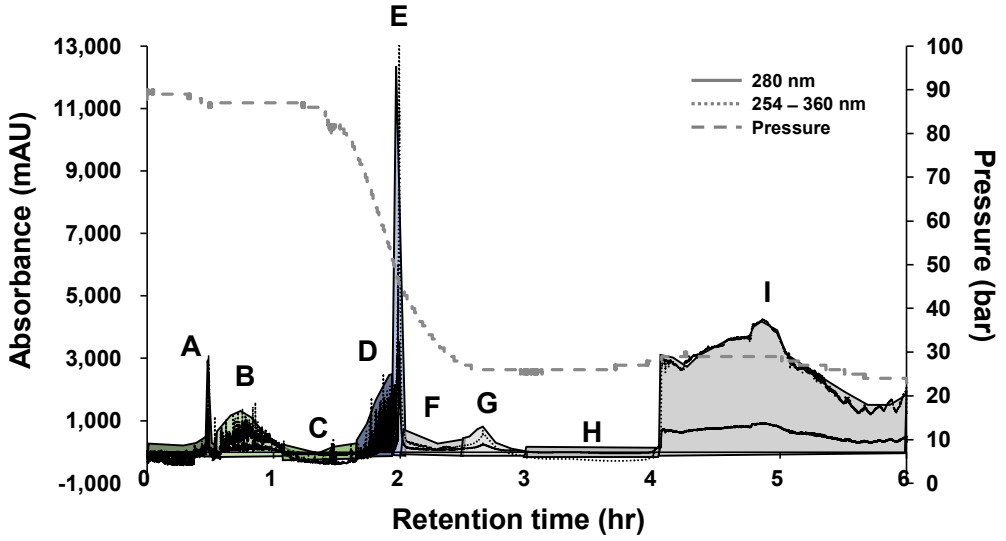

Figure S2. CPC chromatogram of crude *L. strychnifolia* extract. CPC operation condition was as follows; The lower layer of n-hexane–acetonitrile–water (10:2:8, v/v/v) was used as the stationary phase and upper layer of A (n-hexane–acetonitrile–water, 10:2:8, v/v/v), B (upper layer of ethyl acetate–acetonitrile–water, 10:2:8, v/v/v), and C (upper layer of water-saturated *n*-butanol–acetonitrile–water, 10:2:8, v/v/v) were subsequently eluted as mobile phase with a flow rate of 10 mL/min. Step gradient elution of mobile phase was as follows: 0–120 min (100% A), 120–180 min (100% A–100% B), 180–240 min (100% B), 240–300 min (100% B–100% C), and 300–360 min (100% C). The rotation speed of the rotor was 900 rpm. Following that, all the remaining samples were recovered by elution of methanol at 50 mL/min. One gram of crude sample was dissolved in 20 mL of mixed upper and lower phases and subjected to the CPC system.

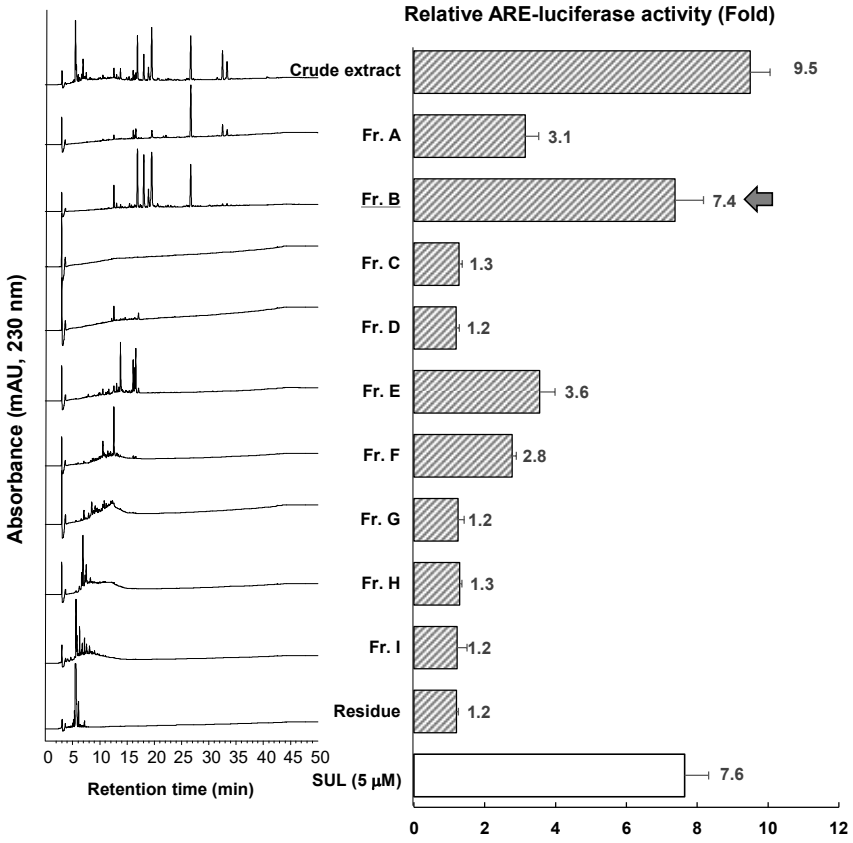

Figure S3. HPLC chromatograms and relative ARE-luciferase activity of CPC-fractions (**A**–**R**). Each CPC fraction was analyzed by HPLC, and its ARE induction activity was evaluated in ARE-HepG2 cells at concentration applied at each assigned weight ratio (based on 30 μg/ml CME). Data are presented as means ± S.E. (n = 3).

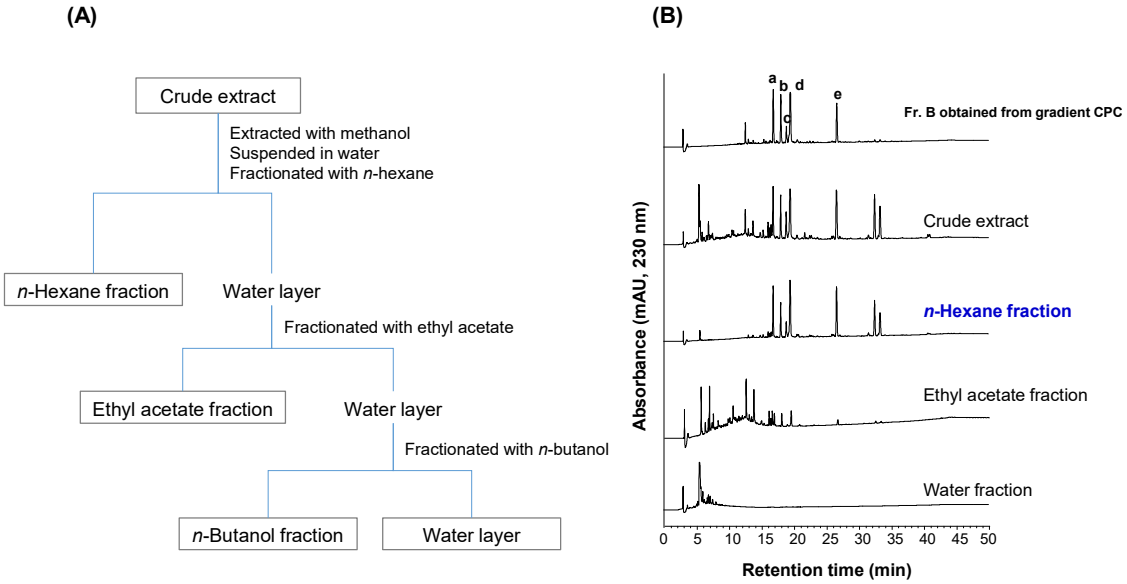

Figure S4. Comparison of solvent fraction and Fr. B obtained from gradient CPC. Scheme of solvent fractionation (**A**) and HPLC analysis of Fr. B (active fraction) and solvent extracts (**B**). HPLC conditions are as follows; column: Capcellpak UG120 C18 column (4.6 × 250 mm, 5 μm, Shiseido, Tokyo, Japan), mobile phase: acetonitrile containing 0.1% formic acid (**A**) and water containing 0.1% formic acid (**B**), gradient elution: 0–10 min, 10–50 % A; 10–45 min, 50–95% A; and 50 min, 95% A, column temperature: 40°C, mobile phase flow rate: 1 mL/min, the detection wavelength: 230 nm, and the injection volume: 10 μL.

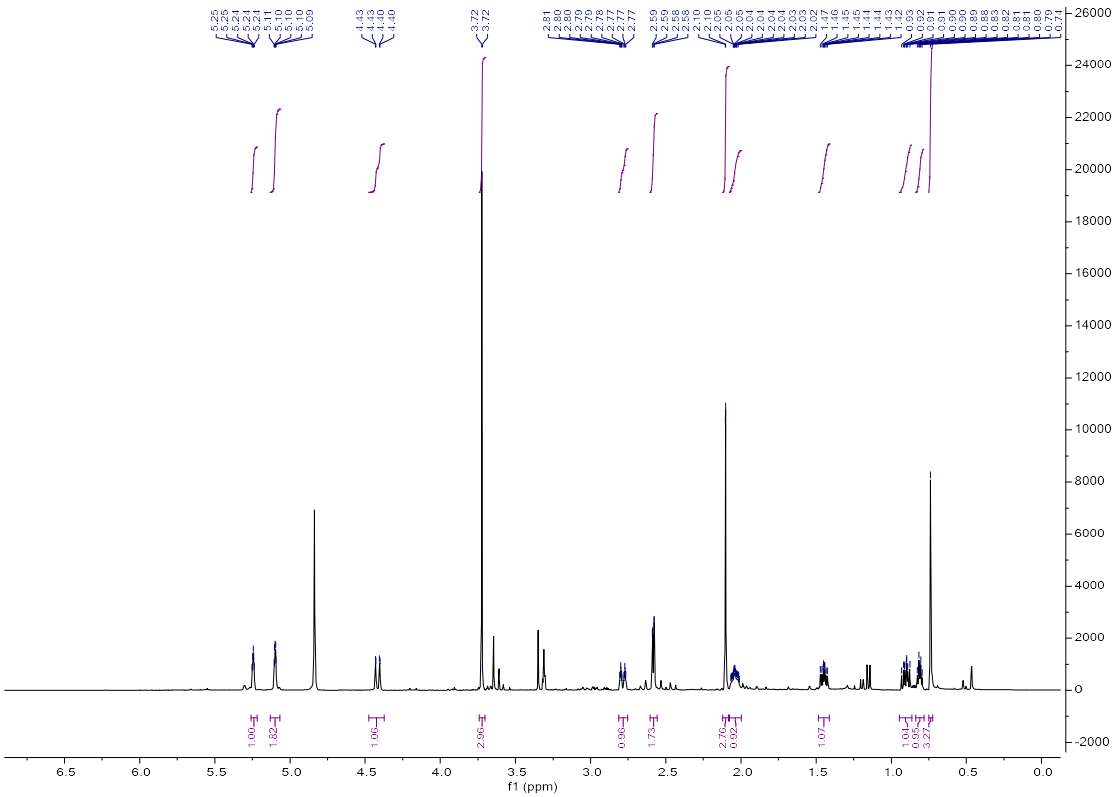

Figure S5. ^{1}H NMR spectrum of **1** (400 MHz) in CD_3OD.

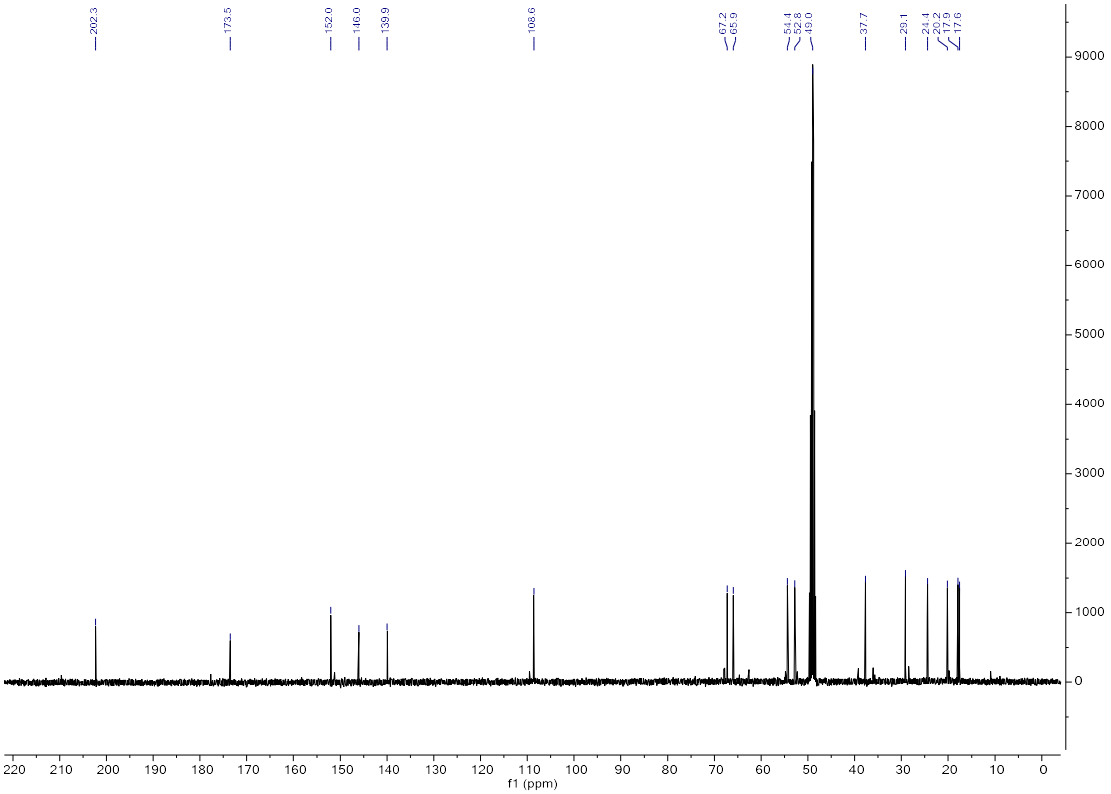

Figure S6. ^{13}C NMR spectrum of **1** (100 MHz) in CD_3OD.

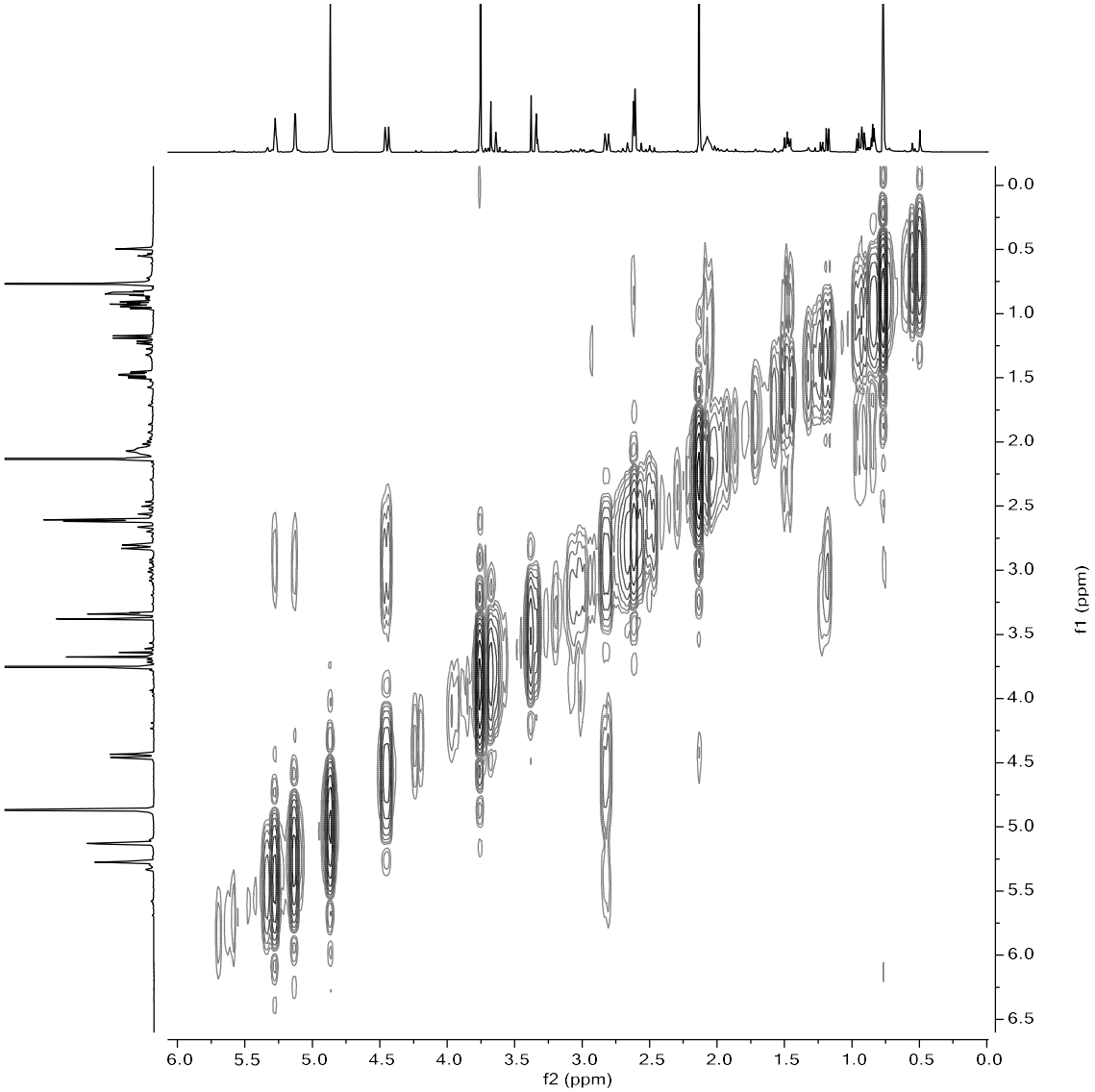

Figure S7. ^{1}H-^{1}H COSY spectrum of **1** in CD_3OD.

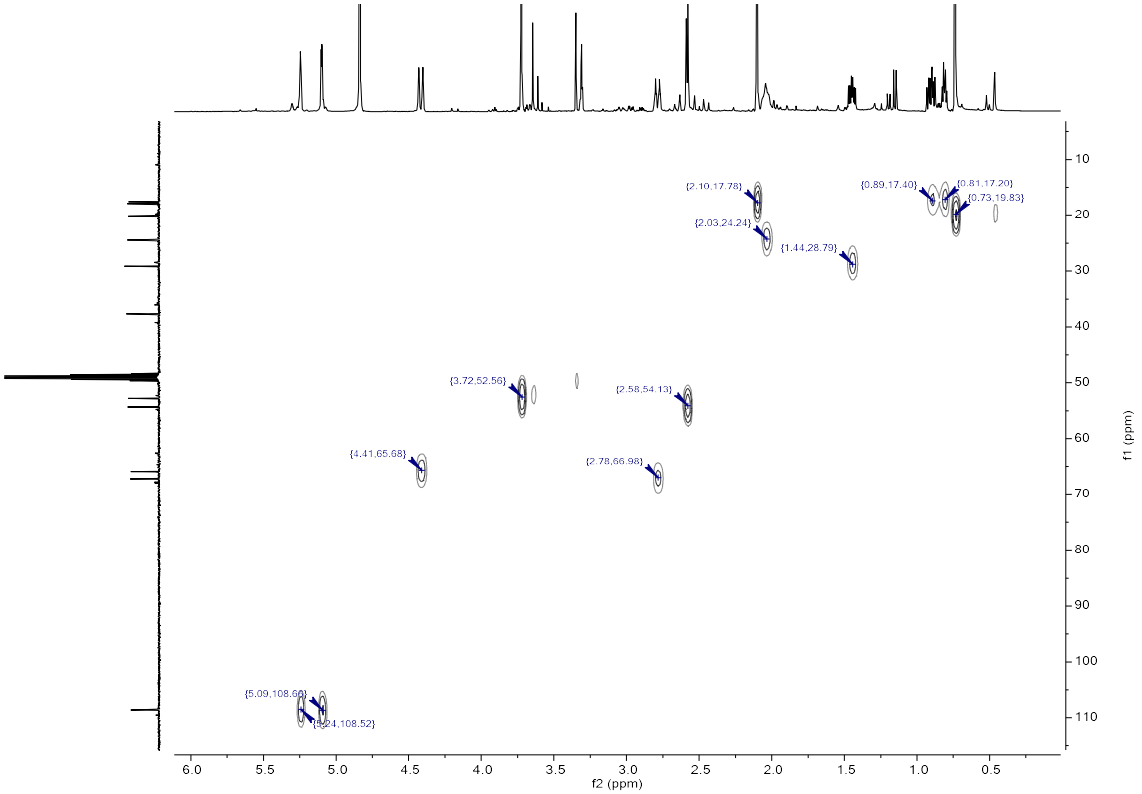

Figure S8. HSQC spectrum of **1** in CD_3OD.

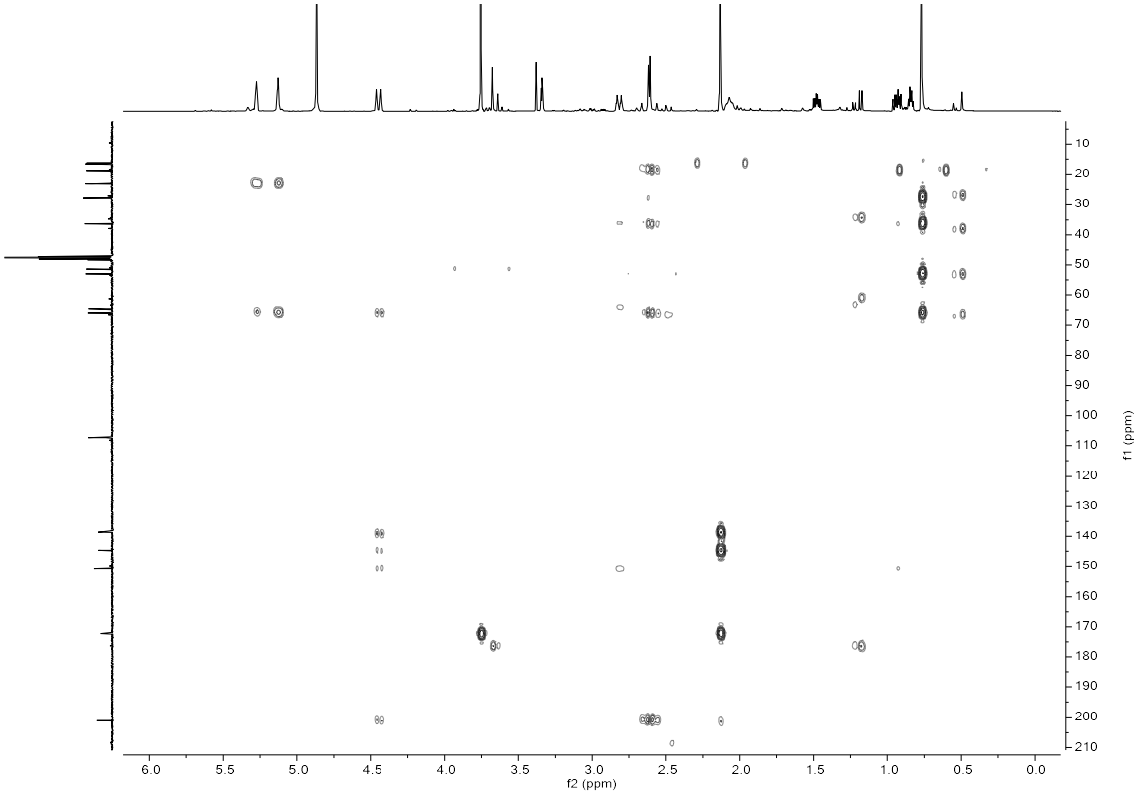

Figure S9. HMBC spectrum of **1** in CD_3OD.

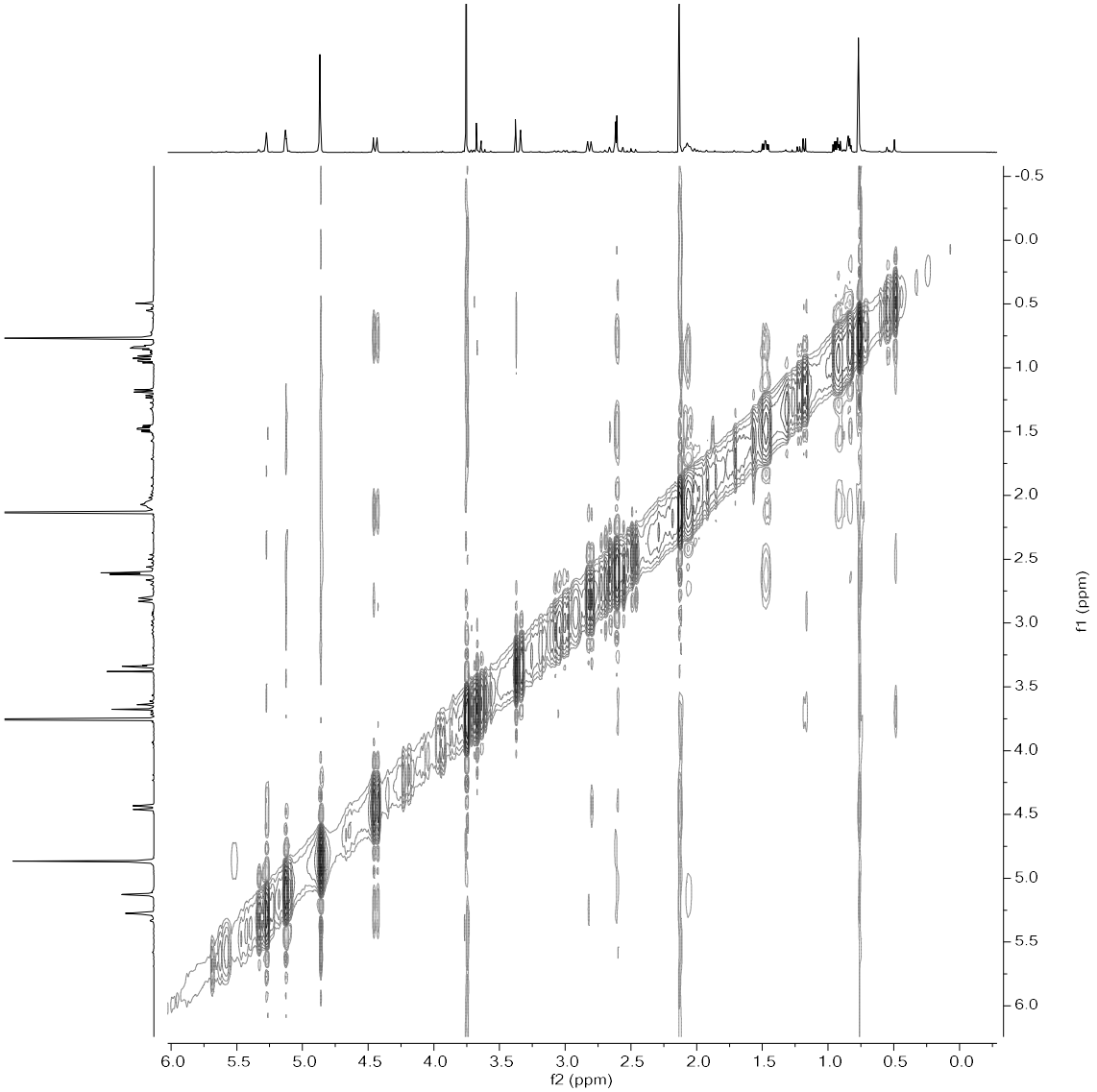

Figure S10. NOESY spectrum of **1** in CD_3OD.

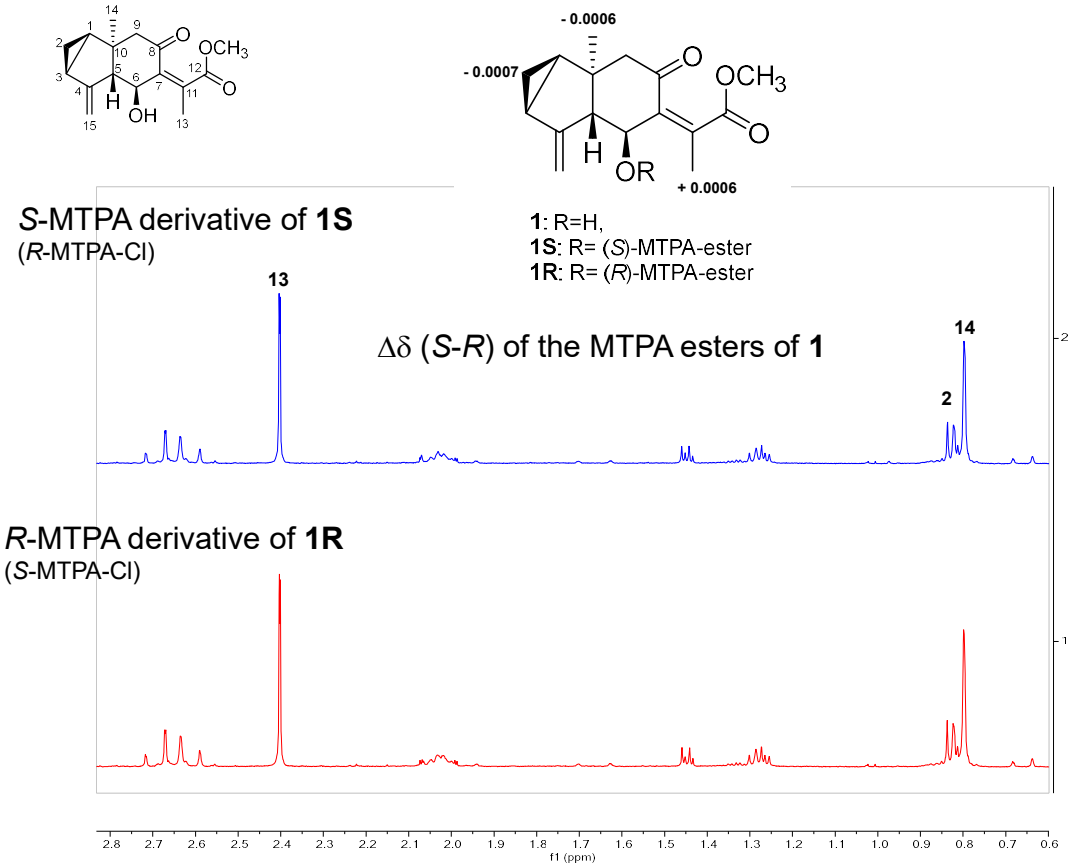

Figure S11. ^{1}H NMR spectra of MTPA esters **1S** and **1R** and $\Delta\delta(S\text{-}R)$ values for (*S*)- and (*R*)-MPTA esters (**1S** and **1R**).

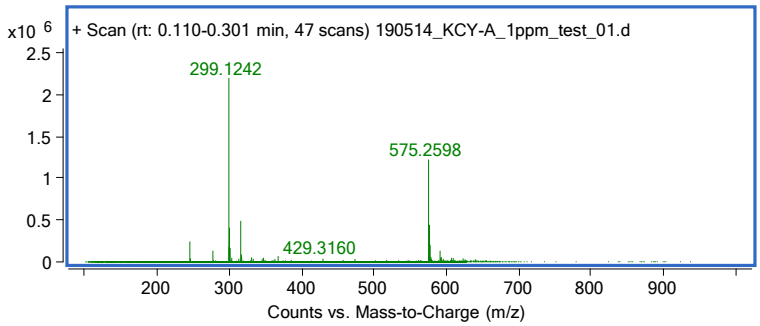

Figure S12. HRESI (qTOF) mass spectrum of **1**.

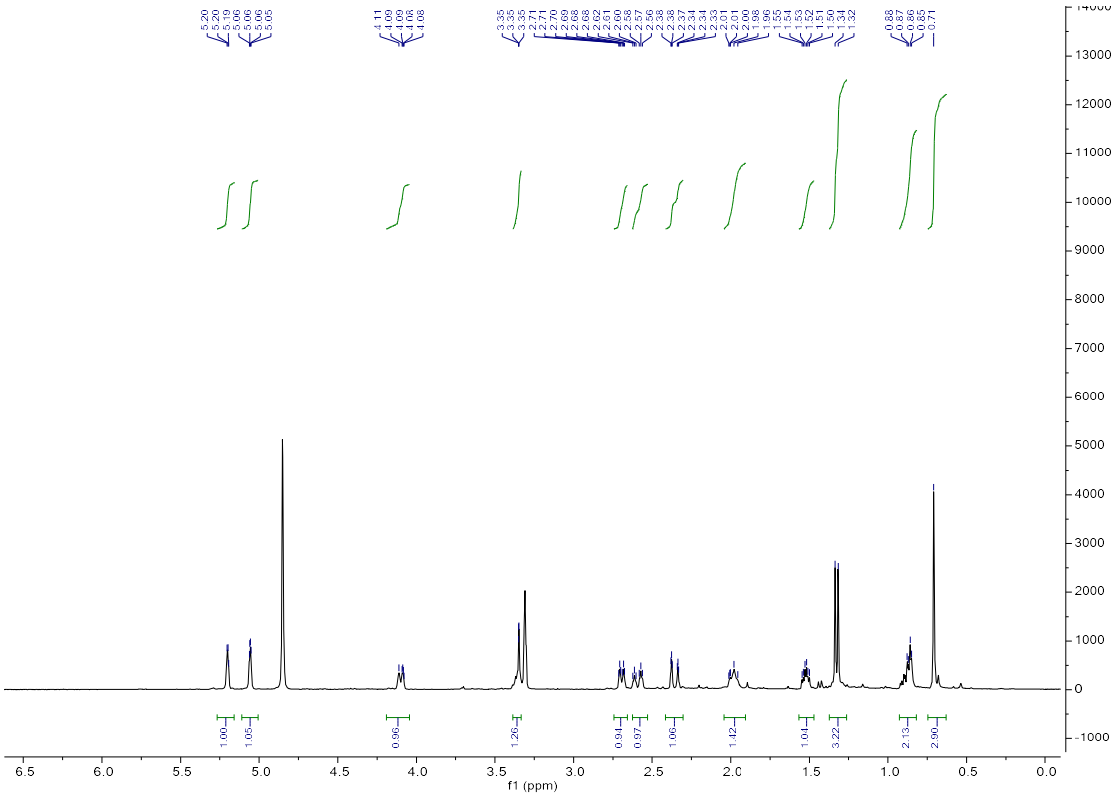

Figure S13. ^{1}H NMR spectrum of **2** (400 MHz) in CD_3OD.

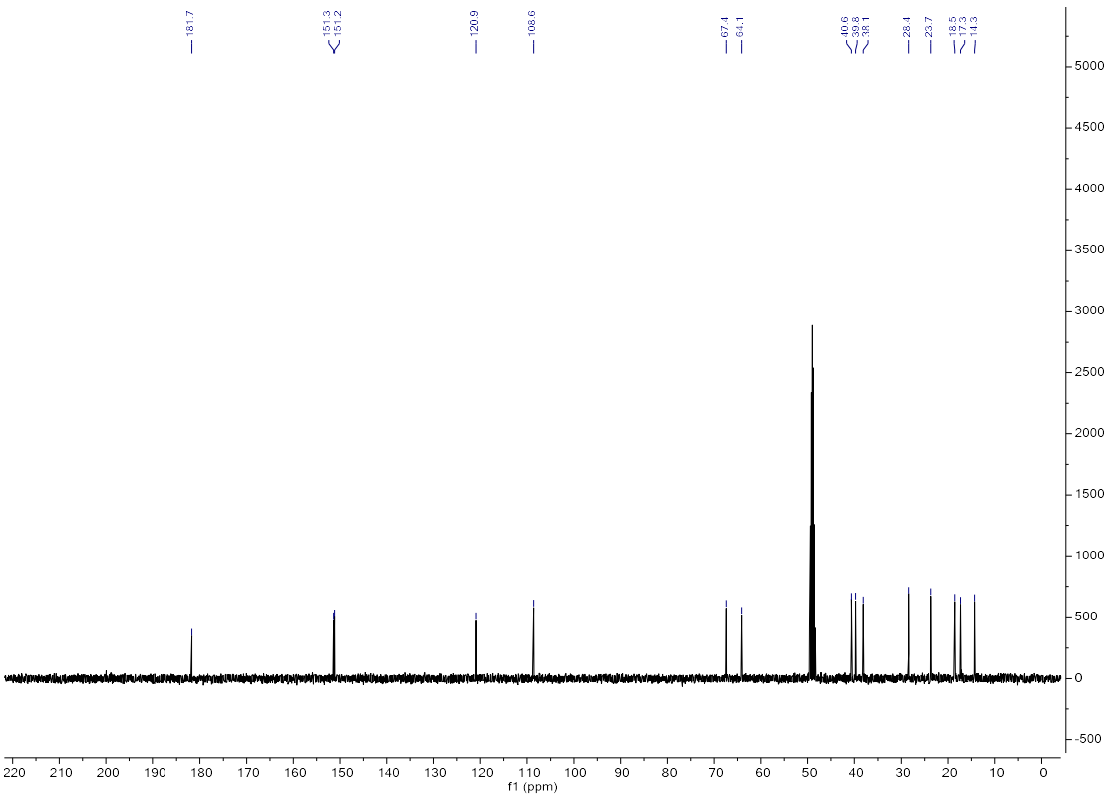

Figure S14. ^{13}C NMR spectrum of **2** (100 MHz) in CD_3OD.

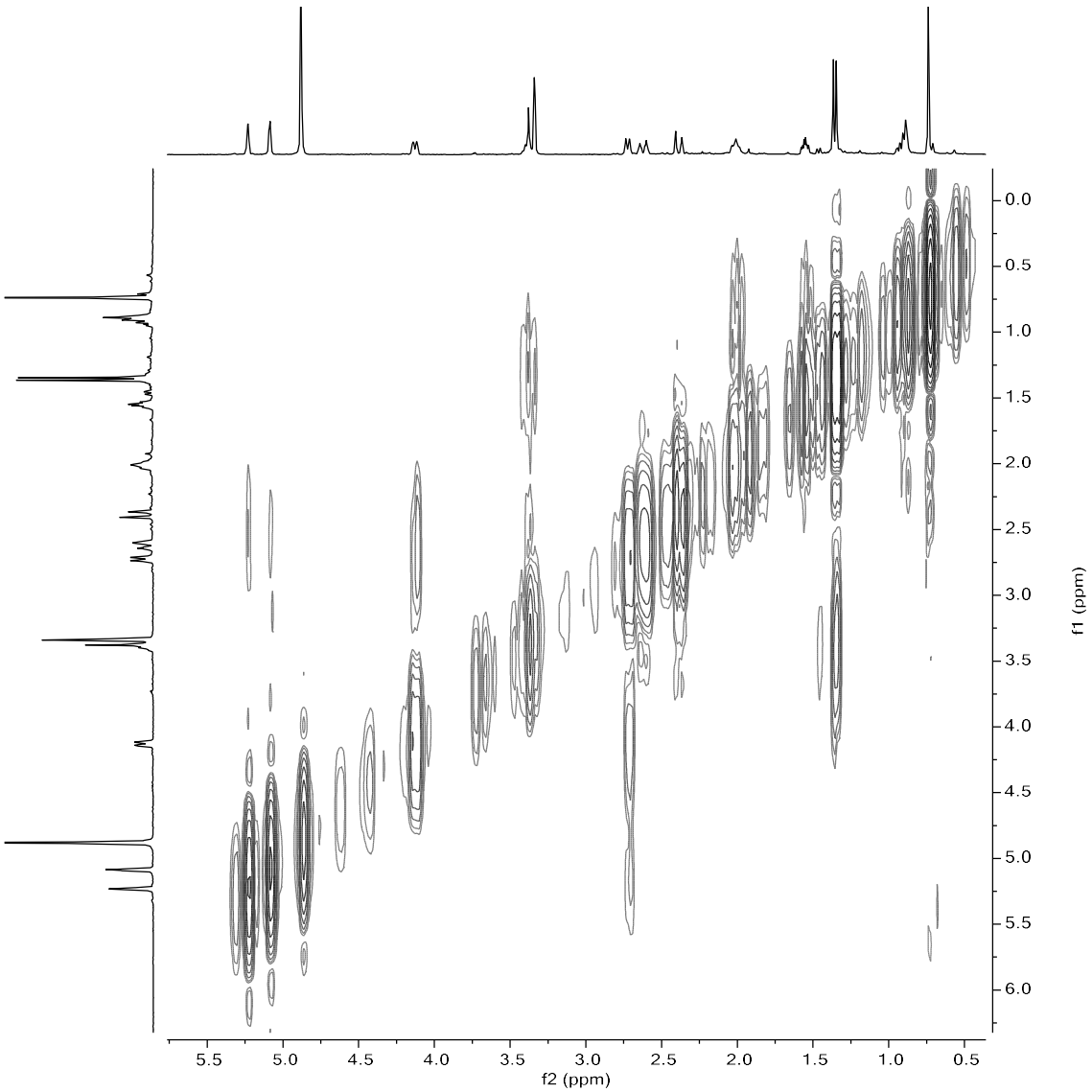

Figure S15. ^{1}H-^{1}H COSY spectrum of **2** in CD_3OD.

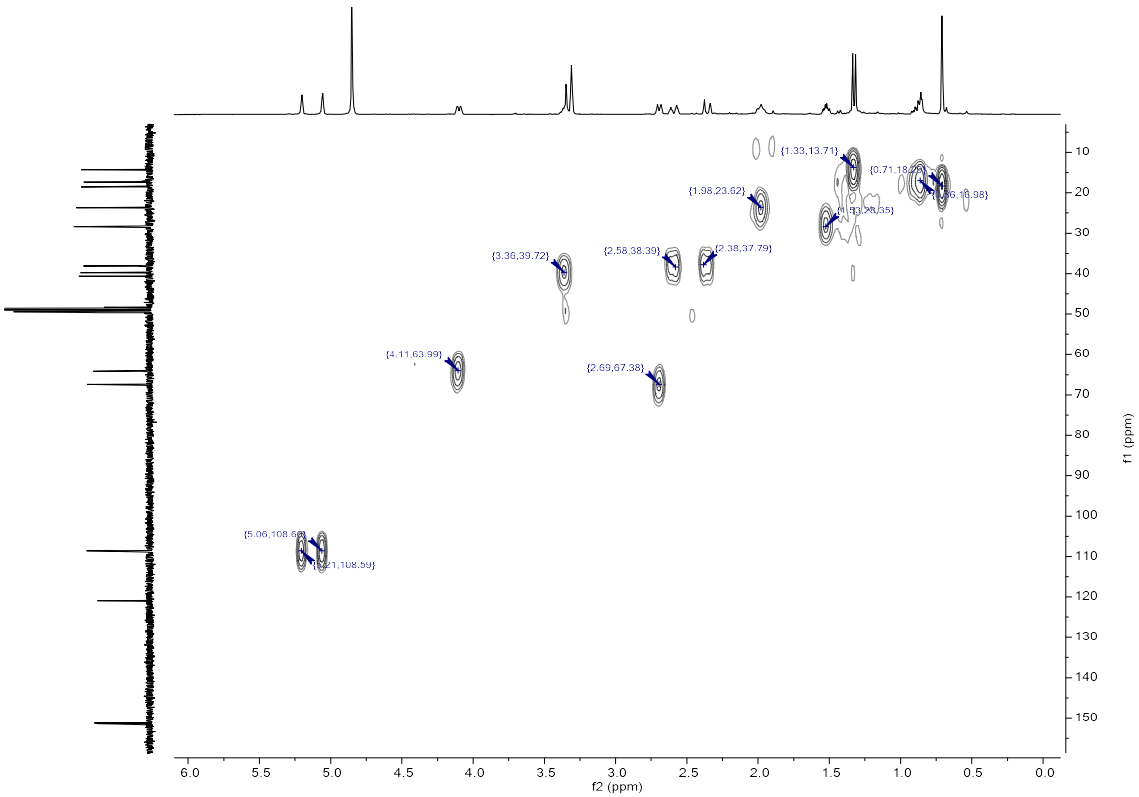

Figure S16. HSQC spectrum of **2** in CD_3OD.

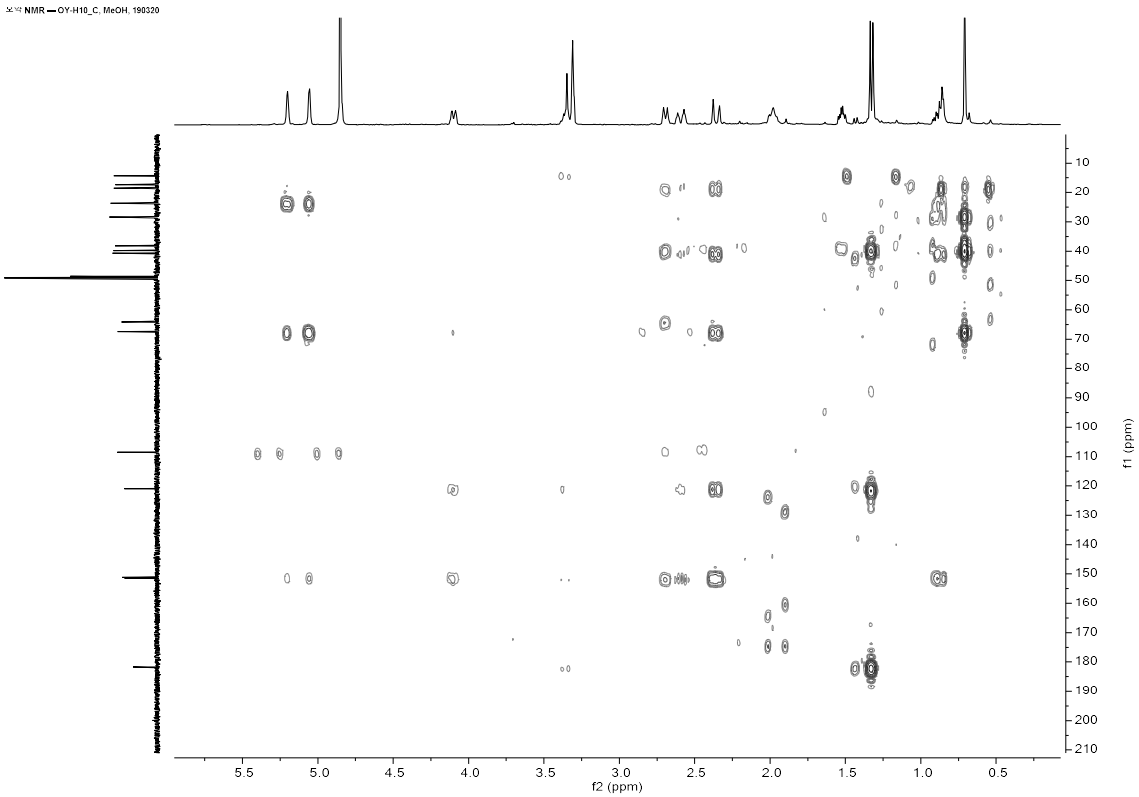

Figure S17. HMBC spectrum of **2** in CD_3OD.

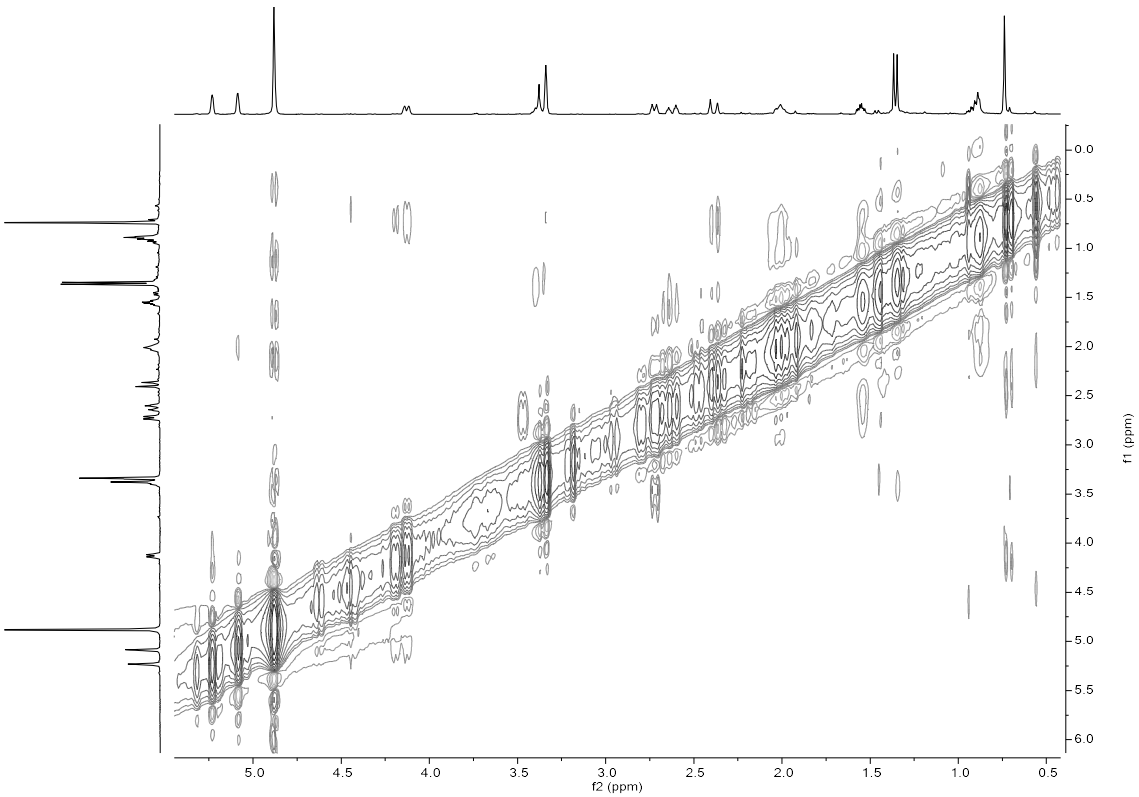

Figure S18. NEOSY spectrum of **2** in CD_3OD.

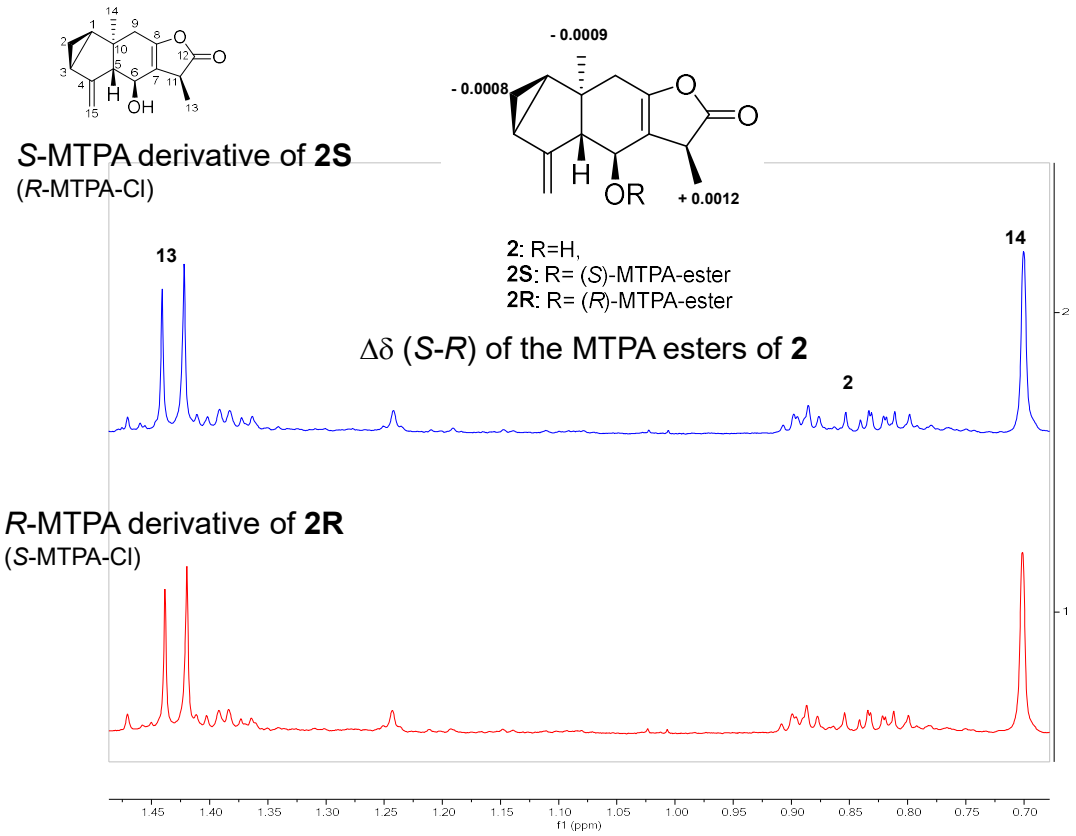

Figure S19. ^{1}H NMR spectra of MTPA esters **2S** and **2R** and $\Delta\delta$ (*S*-*R*) values for (*S*)- and (*R*)-MPTA esters (**2S** and **2R**).

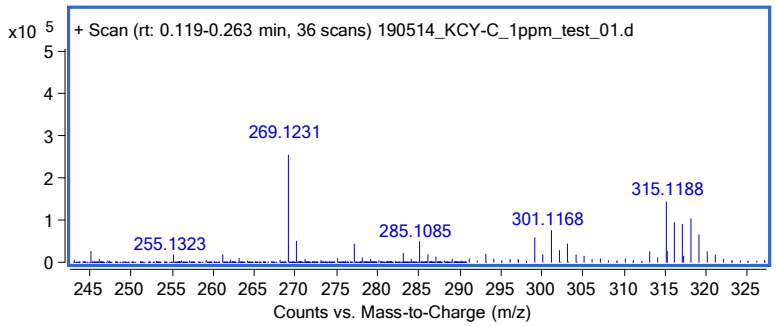

Figure S20. HRESI (qTOF) mass spectrum of **2**.