Check for updates
Natural Product Sciences
29(1) : 17-23 (2023)
https://doi.org/10.20307/nps.2023.29.1.17

Meliasendanins E-J, Nor-neolignan Constituents from *Melia toosendan* and their Anti-inflammatory Activity

Jin Woo Lee*

College of Pharmacy, Duksung Women's University, Seoul 01369, Republic of Korea

Abstract – A phytochemical investigation of the fruits extract of *Melia toosendan* afforded the isolation of two new nor-neolignans, meliasendanins E (**1**) and F (**2**), as well as twelve known compounds (**3 - 14**) using various separation technique such as Diaion HP20, silica, RP-18 gel column chromatography and semi-preparative HPLC. Their chemical structures were elucidated by extensive NMR spectroscopic data including 2D-NMR, and HR-ESI-MS as well as ECD data. Among the twelve known compounds, the absolute structures of **3 - 6** were determined first, and given the trivial names as meliasendanins G-J (**3 - 6**). Based on the evaluation of anti-inflammatory activity, compounds **7 - 8** exhibited inhibitory effects on LPS-induced nitric oxide production in RAW 264.7 macrophages with IC_{50} values of 34.6 and 39.5 μM, respectively.
Keywords – *Melia toosendan*, Meliaceae, Nor-neolignans, Anti-inflammatory, Limonoid

Introduction

Melia toosendan Sieb. et Zucc. (Meliaceae) is widely distributed in the tropical and subtropical region such as southwestern China. The fruit of *Melia* sp. has been known to contain mainly limonoids,[1] triterpenes,[2] sterols,[3] lignans and neolignans.[4] These structures isolated from *M. toosendan* have been reported to show antibacterial,[5] antiviral,[6] and antifeedant activities.[7] The fruit of *M. toosendan* traditionally used as anthelmintics, antimalarials, and antipyretics in China and Africa. In the course of phytochemical search of Meliaceae family, two new nor-neolignans, meliasendanins E (**1**) and F (**2**), along with twelve known compounds (**3 - 14**) (Fig. 1), were isolated from the methanol extract of the fruit of *M. toosendan*. Herein, this paper describes the purification of all isolated compounds using various separation technique such as Diaion HP20 gel column chromatography, normal phase (NP) silica gel column chromatography, reversed phase (RP) silica gel column chromatography, and semi-preparative HPLC. The absolute structure elucidation of new compounds (**1** and **2**) and four known neolignans (**3-6**), which had been only reported the relative configurations of C-7 and C-8 in the lignan skeleton, has been determined based on the spectroscopic data interpretation, especially 1D- and 2D-NMR data such as HSQC, HMBC, COSY and NOESY, HR-ESI-MS, and ECD data.

*Author for correspondence
Jin Woo Lee, College of Pharmacy, Duksung Women's University, Seoul 01369, Republic of Korea.
Tel: +82-2-901-8841; E-mail: jwlee22@duksung.ac.kr

Experimental

General experimental procedures – TLC: Pre-coated silica gel 60 F_{254} (SiO_2, 0.25 mm; Merck); spots were visualized under UV light and by spraying with 10% vanillin-H_2SO_4 in water reagent followed by heating. Column chromatography (CC): silica gel (SiO_2, 70-230 mesh; Merck), Lichroprep RP-18 (40-63 μm, Merck), Diaion HP-20 ion exchange resin (Mitsubishi Chemical Corporation). Medium pressure liquid chromatography (MPLC): Biotage Isolera chromatography system (Biotage). Semi-preparative high-performance liquid chromatography (HPLC) was performed on Waters instruments (pump 515, UV detector 2996) equipped with YMC J'sphere ODS-H80 column (4 μm, 150 × 20 mm i.d.) at a flow rate of 6 mL/min. Optical rotations were measured on a JASCO DIP-1000 polarimeter. UV spectra were recorded on a JASCO UV-550 spectrophotometer, and IR spectra were measured on a JASCO FT-IR 4100 spectrometer. ECD spectra were obtained on a JASCO J-715 spectrometer. 1D- and 2D-NMR spectra were taken on a Bruker AVANCE 400 and 500 MHz spectrometers using CD_3OD as a solvent. ESI-MS and HR-ESI-MS were obtained with LCQ Fleet and maXis 4G mass spectrometers, respectively.

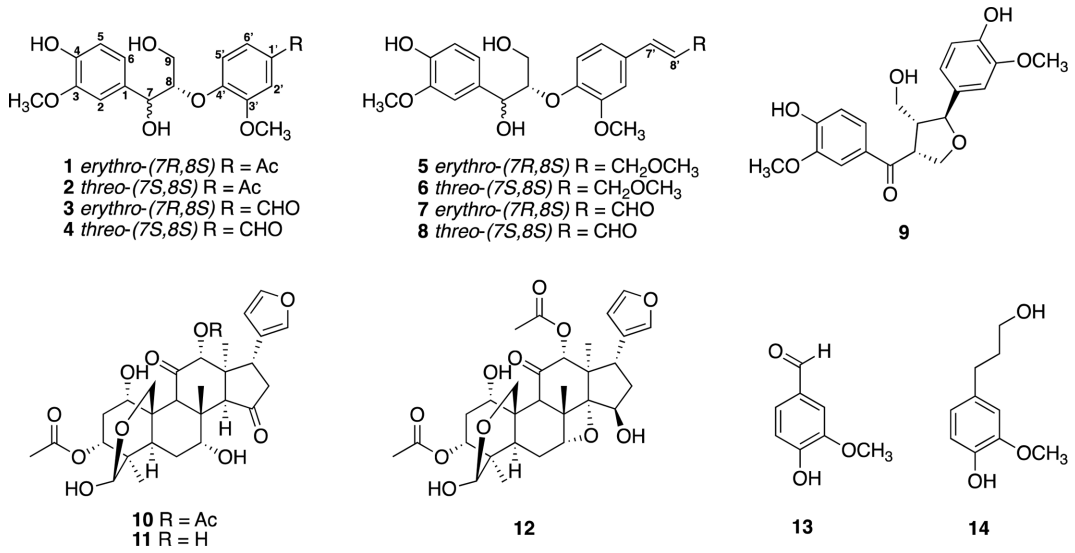


Fig. 1. Structures of compounds **1** - **14**.

Plant materials – The dried fruits of *M. toosendan* were purchased from Kyung-dong herbal market, Seoul, Korea, in April 2022 and authenticated by Prof. Jin Woo Lee. A voucher specimen (2022-DFMT) has been deposited at the Laboratory of Pharmacognosy, College of Pharmacy, Duksung Women's University, Seoul, Republic of Korea.

Extraction and isolation – The dried and powdered fruits of *M. toosendan* (1 kg) were extracted with 100% MeOH. The crude extract (100 g) was fractionated into five fractions, fractions A (100% water), B (30% MeOH), C (60% MeOH), D (80% MeOH) and E (100% MeOH), by Diaion HP20 gel column chromatography and eluted with H_2O-MeOH gradient system (100:0 to 0:100). Fraction D (28 g) was chromatographed on a silica gel column and eluted with CH_2Cl_2-MeOH gradient system (100:0 to 0:100) to give eight fractions, MTD1 – MTD8. Compound **13** (1 mg) was purified from MTD1 fraction (55 mg) by semi-preparative HPLC (MeCN-H_2O, 40:60 to 60:40). MTD4 fraction (2.0 g) was chromatographed on MPLC (RP-18) and eluted with MeOH-H_2O (20:80 to 100:0) to obtain nine fractions, MTD4-1 - MTD4-9. MTD4-2 fraction (160 mg) was further separated by semi-preparative HPLC (MeCN-H_2O, 30:70 to 50:50) to yield compounds **3** (1.8 mg) and **4** (2.3 mg). MTD4-3 fraction (120 mg) was purified by semi-preparative HPLC (MeCN-H_2O, 30:70 to 60:40) to obtain compounds **1** (0.9 mg) and **2** (1.4 mg). MTA4-4 fraction (150 mg) was further separated by semi-preparative HPLC (MeCN-H_2O, 40:60 to 60:40) to afford compounds **7** (5.6 mg), and **8** (6.3 mg). Compound **9** (2.2 mg) was purified from MTD4-5 fraction (40 mg) by semi-preparative HPLC (MeCN-H_2O, 40:60 to 70:30). MTD4-7 fraction (140 mg) was further purified by semi-preparative HPLC (MeCN-H_2O, 50:50 to 70:30) to yield compounds **5** (1.5 mg), **6** (1.9 mg) and **10** (5.0 mg). MTD5 fraction (2.8 g) was chromatographed on MPLC (RP-18) and eluted with MeOH-H_2O (20:80 to 100:0) to afford eleven fractions, MTD5-1 – MTD5-11. MTD5-3 fraction (150 mg) was further purified by semi-preparative HPLC (MeCN-H_2O, 50:50 to 70:30) to obtain compounds **12** (15.2 mg) and **14** (1.5 mg). Compound **11** (6.1 mg) was purified from MTD5-6 fraction (80 mg) by semi-preparative HPLC (MeCN-H_2O, 70:30 to 100:0).

Meliasendanin E (1) – Colorless gum; $[\alpha]_D^{25}$: -18 (*c* 0.1, MeOH); UV (MeOH): 280 (1.25); ECD (*c* 0.02, MeOH): 236 (+0.56), 298 (-0.16); ^{1}H-NMR (CD_3OD, 500 MHz) and ^{13}C-NMR (CD_3OD, 125 MHz), see Table 1; HR-ESI-MS: 385.1258 $[M + Na]^+$, $C_{19}H_{22}NaO_7$, calcd. 385.1258).

Meliasendanin F (2) – Colorless gum; $[\alpha]_D^{25}$: -16 (*c* 0.1, MeOH); UV (MeOH): 279 (0.85); ECD (*c* 0.02, MeOH): 240 (+0.71), 290 (-0.14); ^{1}H-NMR (CD_3OD, 500 MHz) and ^{13}C-NMR (CD_3OD, 125 MHz), see Table 1; HR-ESI-MS: 385.1258 $[M + Na]^+$, $C_{19}H_{22}NaO_7$, calcd. 385.1258).

Meliasendanin G (3) – Colorless gum; $[\alpha]_D^{25}$: -16 (*c* 0.1, MeOH); ECD (*c* 0.02, MeOH): 240 (+0.29), 298 (-0.19); ^{1}H-NMR (CD_3OD, 400 MHz): δ 9.78 (1H, d, J = 8.0 Hz, H-7'), 7.44 (1H, dd, J = 8.4, 2.0 Hz, H-6'), 7.42 (1H, d, J = 2.0 Hz, H-2'), 7.14 (1H, d, J = 8.0 Hz, H-5'), 7.07 (1H, d, J = 2.0 Hz, H-2), 6.87 (1H, dd, J = 8.0, 2.0 Hz, H-6), 6.70 (1H, d, J = 8.0 Hz, H-5), 4.83 (1H, d,

Table 1. ^{1}H- (500 MHz) and ^{13}C-NMR (125 MHz) spectroscopic data for **1** and **2** in CD_3OD

Position	**1**		**2**	
	δ_C (125 MHz)	δ_H (500 MHz)	δ_C (125 MHz)	δ_H (500 MHz)
1	132.4	-	132.3	-
2	110.5	7.06 d (2.0)	110.1	7.05 d (2.0)
3	147.2	-	147.4	-
4	145.6	-	145.7	-
5	114.1	6.71 d (8.0)	114.3	6.76 d (8.0)
6	119.7	6.87 dd (8.0, 2.0)	119.1	6.87 dd (8.0, 2.0)
7	72.6	4.82 d (5.5)	72.3	4.91 m
8	83.6	4.64 m	84.2	4.60 m
9	61.0	3.87 m	60.6	3.81 dd (12.0, 5.0) 3.57 dd (12.0, 7.0)
1'	130.2	-	130.3	-
2'	110.8	7.50 d (2.0)	110.8	7.57 d (2.0)
3'	149.7	-	149.6	-
4'	152.9	-	153.1	-
5'	114.2	7.04 d (8.5)	114.1	7.11 d (8.5)
6'	123.0	7.57 dd (8.5, 2.0)	123.1	7.61 dd (8.5, 2.0)
7'	198.0	-	198.0	-
CH_3CO	24.9	2.55 s	24.9	2.57 s
3-OCH_3	54.8	3.82 s	54.8	3.83 s
3'-OCH_3	55.0	3.85 s	55.0	3.93 s

J = 6.0 Hz, H-7), 4.66 (1H, m, H-8), 3.89 (2H, m, H-9), 3.87 (3H, s, 3'-OCH_3), 3.82 (3H, s, 3-OCH_3); ^{13}C-NMR (CD_3OD, 100 MHz): δ 191.5 (C-7'), 154.1 (C-4'), 150.4 (C-3'), 147.3 (C-3), 145.7 (C-4), 132.4 (C-1), 130.3 (C-1'), 125.7 (C-6'), 119.8 (C-6), 114.5, 114.2 (C-5,5'), 110.7, 110.2 (C-2,2'), 83.7 (C-8), 72.6 (C-7), 61.1 (C-9), 55.0, 54.9 (3,3'-OCH_3); HR-ESI-MS: 371.1100 $[M + Na]^+$, $C_{18}H_{20}NaO_7$, calcd. 371.1101).

Meliasendanin H (4) – Colorless gum; $[\alpha]_D^{25}$: -18 (*c* 0.1, MeOH); ECD (*c* 0.02, MeOH): 236 (+0.49), 278 (-0.15); ^{1}H-NMR (CD_3OD, 500 MHz): δ 9.81 (1H, d, *J* = 8.0 Hz, H-7'), 7.49 (1H, d, *J* = 2.0 Hz, H-2'), 7.48 (1H, dd, *J* = 8.5, 2.0 Hz, H-6'), 7.20 (1H, d, *J* = 8.0 Hz, H-5'), 7.06 (1H, d, *J* = 2.0 Hz, H-2), 6.88 (1H, dd, *J* = 8.0, 2.0 Hz, H-6), 6.77 (1H, d, *J* = 8.0 Hz, H-5), 4.93 (1H, m, H-7), 4.65 (1H, m, H-8), 3.94 (3H, s, 3'-OCH_3), 3.84 (3H, s, 3-OCH_3), 3.81 (1H, m, H-9), 3.60 (1H, m, H-9); ^{13}C-NMR (CD_3OD, 125 MHz): δ 191.5 (C-7'), 154.3 (C-4'), 150.2 (C-3'), 147.4 (C-3), 145.8 (C-4), 132.3 (C-1), 130.3 (C-1'), 125.9 (C-6'), 119.2 (C-6), 114.4, 114.2 (C-5, 5'), 110.2, 110.0 (C-2, 2'), 84.1 (C-8), 72.4 (C-7), 60.7 (C-9), 55.1, 54.9 (3,3'-OCH_3); HR-ESI-MS: 371.1100 $[M + Na]^+$, $C_{18}H_{20}NaO_7$, calcd. 371.1101).

Meliasendanin I (5) – Yellowish gum; $[\alpha]_D^{25}$: -24 (*c* 0.1, MeOH); ECD (*c* 0.02, MeOH): 236 (+0.49), 296 (-0.34); ^{1}H-NMR (CD_3OD, 400 MHz): δ 7.03 (2H, m, H-5',6'), 6.89 (2H, m, H-2,2'), 6.86 (1H, dd, *J* = 8.0, 2.0 Hz, H-6), 6.75 (1H, d, *J* = 8.0 Hz, H-5), 6.56 (1H, d, *J* = 16.0 Hz, H-7'), 6.20 (1H, dt, *J* = 16.0, 6.5 Hz, H-8'), 4.83 (1H, m, H-7), 4.39 (1H, m, H-8), 4.03 (2H, dd, *J* = 6.5, 1.5 Hz, H-9'), 3.84 (2H, m, H-9), 3.83, 3.82 (6H, s, 3,3'-OCH_3), 3.38 (3H, s, 9'-OCH_3); ^{13}C-NMR (CD_3OD, 100 MHz): δ 150.5 (C-3'), 147.8 (C-4'), 147.3 (C-3), 145.6 (C-4), 132.7, 132.4 (C-1,1'), 131.3 (C-7'), 123.8 (C-8'), 119.7, 119.4 (C-6, 6'), 117.4, 114.3 (C-5,5'), 110.5, 110.0 (C-2,2'), 84.8 (C-8), 72.8 (C-7), 72.7 (C-9'), 60.8 (C-9), 56.6 (9'-OCH_3), 55.1, 54.9 (3,3'-OCH_3); HR-ESI-MS: 413.1579 $[M + Na]^+$, $C_{21}H_{26}NaO_7$, calcd. 413.1571).

Meliasendanin J (6) – Yellowish gum; $[\alpha]_D^{25}$: -14 (*c* 0.1, MeOH); ECD (*c* 0.02, MeOH): 239 (+1.28), 290 (-1.51); ^{1}H-NMR (CD_3OD, 500 MHz): δ 7.09 (1H, d, *J* = 2.0 Hz, H-2), 7.05 (1H, d, *J* = 2.0 Hz, H-2'), 7.02 (1H, d, *J* = 8.5 Hz, H-5), 6.95 (1H, dd, *J* = 8.0, 2.0 Hz, H-6'), 6.88 (1H, dd, *J* = 8.5, 2.0 Hz, H-6), 6.77 (1H, d, *J* = 8.0 Hz, H-5'), 6.58 (1H, d, *J* = 16.0 Hz, H-7'), 6.22 (1H, dt, *J* = 16.0, 6.5 Hz, H-8'), 4.89 (1H, m, H-7), 4.32 (1H, m, H-8), 4.09 (2H, dd, *J* = 6.5, 1.5 Hz, H-9'), 3.90, 3.84 (6H, s, 3,3'-OCH_3), 3.75 (1H, m, H-9), 3.50 (1H, m, H-9), 3.39 (3H, s, 9'-OCH_3); ^{13}C-NMR (CD_3OD, 100 MHz): δ 150.3 (C-3'), 148.0 (C-4'), 147.4 (C-3), 145.8 (C-4), 132.4 (C-1,1'),

131.3 (C-7'), 123.8 (C-8'), 119.6, 119.3 (C-6,6'), 117.2, 114.4 (C-5,5'), 110.2, 109.8 (C-2,2'), 85.6 (C-8), 72.8 (C-7), 72.6 (C-9'), 60.4 (C-9), 56.7 (9'-OCH_3), 55.1, 54.9 (3,3'-OCH_3); HR-ESI-MS: 413.1571 $[M + Na]^+$, $C_{21}H_{26}NaO_7$, calcd. 413.1571).

Fordiane A (7) – Yellowish gum; $[\alpha]_D^{25}$: +28 (*c* 0.1, MeOH); ECD (*c* 0.02, MeOH): 242 (+0.60), 300 (-0.19); ^{1}H-NMR (CD_3OD, 400 MHz): δ 9.60 (1H, d, *J* = 8.0 Hz, H-9'), 7.59 (1H, d, *J* = 16.0, H-7'), 7.24 (1H, d, *J* = 2.0 Hz, H-2'), 7.18 (1H, dd, *J* = 8.4, 2.0 Hz, H-6'), 7.06 (1H, d, *J* = 2.0 Hz, H-2), 7.03 (1H, d, *J* = 8.4 Hz, H-5'), 6.87 (1H, dd, *J* = 8.0, 2.0 Hz, H-6), 6.72 (1H, d, *J* = 8.0 Hz, H-5), 6.68 (1H, d, *J* = 16.0, 8.0 Hz, H-8'), 4.83 (1H, d, *J* = 6.0 Hz, H-7), 4.57 (1H, m, H-8), 3.86 (2H, m, H-9), 3.85, 3.82 (6H, s, 3,3'-OCH_3); ^{13}C-NMR (CD_3OD, 100 MHz): δ 194.8 (C-9'), 154.2 (C-7'), 151.3 (C-4'), 150.3 (C-3'), 147.2 (C-3), 145.7 (C-4), 132.5 (C-1), 127.8 (C-1'), 126.2 (C-8'), 123.0 (C-6'), 119.7 (C-6), 115.6, 114.2 (C-5,5'), 111.2, 110.5 (C-2,2'), 83.9 (C-8), 72.6 (C-7), 61.0 (C-9), 55.1, 54.9 (3,3'-OCH_3); ESI-MS: 397 $[M + Na]^+$.

Fordiane B (8) – Yellowish gum; $[\alpha]_D^{25}$: -24 (*c* 0.1, MeOH); ECD (*c* 0.02, MeOH): 240 (+0.49), 308 (-0.12); ^{1}H-NMR (CD_3OD, 400 MHz): δ 9.61 (1H, d, *J* = 8.0 Hz, H-9'), 7.62 (1H, d, *J* = 16.0, H-7'), 7.31 (1H, d, *J* = 2.0 Hz, H-2'), 7.22 (1H, dd, *J* = 8.4, 2.0 Hz, H-6'), 7.11 (1H, d, *J* = 8.4 Hz, H-5'), 7.05 (1H, d, *J* = 2.0 Hz, H-2), 6.88 (1H, dd, *J* = 8.0, 2.0 Hz, H-6), 6.76 (1H, d, *J* = 8.0 Hz, H-5), 6.71 (1H, d, *J* = 16.0, 8.0 Hz, H-8'), 4.89 (1H, m, H-7), 4.53 (1H, m, H-8), 3.93, 3.83 (6H, s, 3,3'-OCH_3), 3.80 (1H, m, H-9), 3.55 (1H, m, H-9); ^{13}C-NMR (CD_3OD, 100 MHz): δ 194.7 (C-9'), 154.0 (C-7'), 151.6 (C-4'), 150.3 (C-3'), 147.4 (C-3), 145.8 (C-4), 132.4 (C-1), 128.0 (C-1'), 126.3 (C-8'), 123.2 (C-6'), 119.3 (C-6), 115.8, 114.5 (C-5,5'), 111.3, 110.3 (C-2,2'), 84.7 (C-8), 72.5 (C-7), 60.7 (C-9), 55.3, 55.0 (OCH_3); ESI-MS: 397 $[M + Na]^+$.

Measurement of LPS-induced NO production and cell viability – RAW 264.7 cells were obtained from the American Type Culture Collection (Manassas, VA, USA), and were cultured in Dulbecco's modified Eagle's medium (DMEM, Gibco-BRL. Louis, MO, USA) with 10% heat-inactivated fetal bovine serum (FBS) and penicillin/streptomycin (100 U/mL) at 37 °C humidified air containing 5% CO_2. RAW264.7 cells were plated at 2×10^5 cells/well in 96-well culture dishes and incubated with or without LPS (1 μg/mL) in the absence or presence of indicate concentration of the samples for 24 h. The NO production was measured according to the Griess reaction. Cell viability of the remaining cells was determined by MTT (Sigma Chemical Co., St. Louis, MO, USA) based colorimetric assay.

Result and Discussion

Meliasendanin E (**1**) was obtained as a colorless gum. Its HR-ESI-TOF-MS spectrum revealed a molecular formula of $C_{19}H_{22}O_7$ based on the ion peak at *m/z* 385.1258 $[M + Na]^+$ (calcd. for $C_{19}H_{22}NaO_7$, 385.1258). In the ^{1}H NMR spectrum of **1**, six aromatic proton signals at δ_H 7.57 (1H, dd, *J* = 8.5, 2.0 Hz, H-6'), 7.50 (1H, d, *J* = 2.0 Hz, H-2'), 7.06 (1H, d, *J* = 2.0 Hz, H-2), 7.04 (1H, d, *J* = 8.5 Hz, H-5'), 6.87 (1H, dd, *J* = 8.0, 2.0 Hz, H-6) and 6.71 (1H, d, *J* = 8.0 Hz, H-5) were observed, corresponding to a couple of ABX trisubstituted aromatic ring protons, and two methoxy group signals at δ_H 3.85 (3H, s, 3'-OCH_3) and 3.82 (3H, s, 3-OCH_3), and one acetyl group signal at δ_H 2.55 (3H, s, 1'-Ac) were detected as well. The ^{13}C NMR data of **1** displayed 19 carbon resonances including one carbonyl carbon signal (δ_C 198.0, C-7'), twelve olefinic carbon singnals (δ_C 152.9, 149.7, 147.2, 145.6, 132.4, 130.2, 123.0, 119.7, 114.2, 114.1, 110.8, 110.5), two oxymethine carbon signals (δ_C 83.6, 72.6), one oxymethylene carbon signal (δ_C 61.0), two methoxy group signals (δ_C 55.0, 54.8) and one methyl group signal (δ_C 24.9) (Table 1). Based on the interpretation of 1D and 2D NMR spectra, **1** was deduced to be biosynthesized with C_6-C_3 and C_6-C_2. The key HMBC correlation between H-8 and C-4' suggested that the two units of **1** were connected with an oxygen bond, indicating that **1** was thought to be an atypical lignan scaffold, called by nor-neolignan (Fig. 2). The two methoxy and the acetyl moieties were attached to C-3, C-3', and C-1', respectively, based on the HMBC correlations of 3-OCH_3/C-3, 3'-OCH_3/C-3', and CH_3(CO)/C-1'. The relative structure and the absolute configuration of **1** were determined by the difference of the chemical shift in the NMR spectrum, and a comparison of ECD analysis with the literatures. According to the literature reporting about the possible staggered conformers of 8-*O*-4' neolignan diastereomers, the large and small *J* values of H-7 and 8 are used to determine the relative configuration of C-7 and 8 like *threo* form and *erythro* form, respectively.[8] In the ^{1}H NMR spectrum of **1**, the relative configuration of C-7 and 8 was confirmed as *erythro* form by a small coupling constant between H-7 and 8 ($J_{H7\text{-}H8}$ = 5.5 Hz). The absolute configuration of C-8 was established as *S* by a positive Cotton effect at 230-240 nm in its ECD spectrum (Fig. 3).[9,10] Thus, the absolute configuration of meliasendanin E (**1**) was confirmed to be 7*R*, 8*S*, and the structure of **1** was turned out as an enantiomer of lycocernuaside D, which has been previously reported in 2017.[11]

Meliasendanin F (**2**) was isolated as a colorless gum

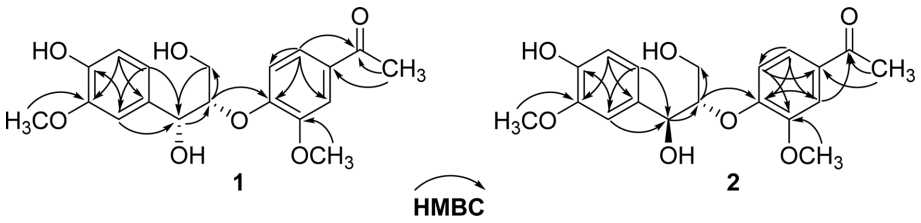


Fig. 2. Key HMBC correlations of compounds **1** and **2**.

Table 2. Comparison of the ^{1}H- and ^{13}C-NMR chemical shift of *erythro* and *threo*-form in C-7 and C-8

	erythro (C-7,8)				*threo* (C-7,8)			
Compound	**1**	**3**	**5**	**7**	**2**	**4**	**6**	**8**
H-9	3.87 (2H)	3.89 (2H)	3.84 (2H)	3.86 (2H)	3.81 3.57	3.81 3.60	3.75 3.50	3.80 3.55
pattern	overwrapped				separated			
C-7	72.6	72.6	72.8	72.6	72.3	72.4	72.8	72.5
C-8	83.6	83.7	84.8	83.8	84.2	84.1	85.6	84.7
$\Delta\delta_{C8-C7}$	11.0	11.1	12.0	11.2	11.9	11.7	12.8	11.8

with the same molecular formula as that of **2**. The interpretation of 1D and 2D NMR experiment suggested that the planar structure of **2** was identical to that of **1**. However, since the J_{H7-H8} value in the ^{1}H NMR spectrum of **2** was difficult and ambiguous to obtain, we applied the $\Delta\delta_{C8-C7}$ values eliminating the effect of systematic errors [$\Delta\delta_{C8-C7}$ (*threo*) > $\Delta\delta_{C8-C7}$ (*erythro*)]. These values are only able to be applied to differentiate *threo* and *erythro* aryl glycerols without substituents at C-7 or/and C-8 in the same solvent.[9,12] The $\Delta\delta_{C8-C7}$ of **2** (11.9) was larger than that of **1** (11.0) in CD_3OD, indicating **2** was determined as *threo* form, and **1** was *erythro*, supported by the pattern of the proton signal chemical shifts of H-9. While the two protons of C-9 in the *threo* forms of these molecules such as compounds **2**, **4**, **6** and **8** were shown separately in their ^{1}H-NMR spectra (δ_H 3.81, 3.57 in **2**, δ_H 3.81, 3.60 in **4**, δ_H 3.75, 3.50 in **6**, δ_H 3.80, 3.55 in **8**), those of the *erythro* forms (**1**, **3**, **5**, **7**) were overwrapped (δ_H 3.87 in **1**, δ_H 3.89 in **3**, δ_H 3.84 in **5**, δ_H 3.86 in **7**) (Table 2). In addition, the absolute configuration of C-8 in **2** was determined as *S* by a positive Cotton effect at 230-240 nm in its ECD spectrum. Therefore, the absolute configuration of **2** was confirmed as 7*S*, 8*S* (Fig. 3).

Meliasendanins G (**3**), H (**4**), I (**5**) and J (**6**) were previously reported as *erytrho*-guaiacylglycerol-8-vanillic acid ether,[13] *threo*-guaiacylglycerol-8-vanillic acid ether,[13] *erythro*-guaiacylglycerol-*β*-9'-methylconiferyl ether ether,[14] and *threo*-guaiacylglycerol-*β*-9'-methylconiferyl ether ether,[14] respectively, only with relative configurations of C-8 and C-9. To determine their relative and absolute configurations, the afore-mentioned methods were applied. Based on the $\Delta\delta_{C8-C7}$ values (11.1 for **3** < 11.7 for **4**, and 12.0 for **5** < 12.8 for **6**) (Table 2), the pattern of H_2-9 in their ^{1}H-NMR spectra, and Cotton effects at 230-240 nm (Fig. S13), the absolute configurations of meliasendanins G-J (**3**-**6**) were established as 7*R*,8*S* (**3**), 7*S*,8*S* (**4**), 7*R*,8*S* (**5**), and 7*S*,8*S* (**6**), respectively.

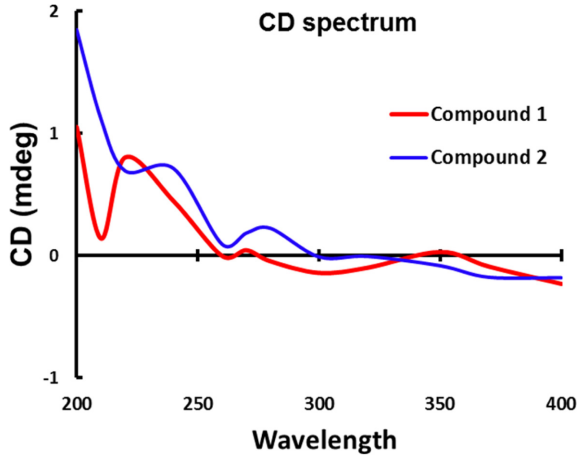


Fig. 3. ECD spectrum of compounds **1** and **2**.

The eight known compounds were identified as fordiane A (**7**),[14,15] fordiane B (**8**),[14,15] vladinol D (**9**),[16] isotoosendanin (**10**),[17] 12-hydroxyamoorastatone (**11**),[18] mesendanin H (**12**),[19] vanillin (**13**),[20] and dihydroconiferyl alcohol (**14**),[21] respectively, by comparing their physicochemical and spectroscopic data with those of published values.

Table 3. Inhibitory effects of compounds **1 - 14** on LPS-induced NO production in macrophage RAW 264.7 Cells[a]

Compound	IC_{50} (μM)	Compound	IC_{50} (μM)
1	>100	**8**	39.5
2	>100	**9**	>100
3	>100	**10**	>100
4	>100	**11**	>100
5	>100	**12**	>100
6	>100	**13**	>100
7	34.6	**14**	>100

Aminoguanidine was used as the positive control (IC_{50} = 15.8 μM)
[a] Results are expressed as the mean IC_{50} values in μM from triplicate experiments.

Nitric oxide (NO) generated by inducible NO synthase (iNOS) in macrophage is involved in the action of mechanisms of inflammation. Excessive NO production may cause various inflammatory diseases such as rheumatism and asthma.[22] Therefore, inhibition of NO production has been thought to be one of potential therapeutic strategies to control inflammatory diseases. Several neolignans isolated from medicinal plants have been reported to show inhibitory effect of NO production. For example, ficusal, isolated from the seeds of *Prunus tomentosa*, has been reported to show inhibitory activity of NO with IC_{50} value of 4.7 μM.[23] Other examples include myrislignan (IC_{50}: 21.1 μM) and machilin D (IC_{50}: 4.7 μM), isolated from the seeds of *Myristica fragrans*, piperkadsin C (IC_{50}: 14.6 μM) and futoquinol (IC_{50}: 16.8 μM), isolated from *Piper kadsura*.[24,25]

To find anti-inflammatory compounds, all isolated compounds (**1 - 14**) were tested for their anti-inflammatory activity on LPS-induced NO production in RAW 264.7 macrophage cells and aminoguanidine was used as positive control (IC_{50}: 15.8 μM). Among them, only fordianes A (**7**) and B (**8**), possessing an α,β-unsaturated aldehyde group at C-1', showed mild inhibitory effects with IC_{50} values of 34.6 and 39.5 μM (Table 3). Although the bioassay results are not enough to mention their structure-activity relationship, because a few aldehyde-bearing neolignans have been previously reported, it is necessary to develop the structures or find more potential nor-neolignan molecules.[26]

Conflicts of Interest

The authors declare that they have no conflicts of interest.

References

(1) Jin, Q.; Lee, C.; Lee, J. W.; Choi, J. Y.; Hong, J. T.; Kim, Y.; Lee, M. K.; Hwang, B. Y. *Helv. Chim. Acta* **2014**, *97*, 1152–1157.
(2) Chiang, C. K.; Chang, F. C. *Tetrahedron* **1973**, *29*, 1911–1929.
(3) Tan, Q.-G.; Li, X.-N.; Chen, H.; Feng, T.; Cai, X.-H.; Luo, X.-D. *J. Nat. Prod.* **2010**, *73*, 693–697.
(4) Wang, L.; Li, F.; Yang, C. Y.; Khan, A. A.; Liu, X.; Wang, M. K. *Fitoterapia* **2014**, *99*, 92–98.
(5) Zhang, Q.; Shi, Y.; Liu, X. T.; Liang, J. Y.; Ip, N. Y.; Min, Z. D. *Planta Med.* **2007**, *73*, 1298–1303.
(6) Petrera, E.; Coto, C. E. *Phytother. Res.* **2009**, *23*, 1771–1777.
(7) Carpinella, M. C.; Defago, M. T.; Valladares, G.; Palacios, S. M. *J. Agric. Food Chem.* **2003**, *51*, 369–374.
(8) Braga, A. C. H.; Zacchino, S.; Badano, H.; Sierra, M. G.; Rúveda, E. A. *Phytochemistry* **1984**, *23*, 2025–2028.
(9) Xiong, L.; Zhu, C.; Li, Y.; Tian, Y.; Lin, S.; Yuan, S.; Hu, J.; Hou, Q.; Chen, N.; Yang, Y.; Shi, J. *J. Nat. Prod.* **2011**, *74*, 1188–1200.
(10) Liao, S. G.; Wu, Y.; Yue, J. M. *Helv. Chim. Acta* **2006**, *89*, 73–80.
(11) Li, J.; Xu, P.-S.; Tan, L.-H.; Zou, Z.-X.; Wang, Y.-K.; Long, H.-P.; Zhou, G.; Li, G.; Xu, K.-P.; Tan, G.-S. *Fitoterapia* **2017**, *119*, 45–50.
(12) Gan, M.; Zhang, Y.; Lin, S.; Liu, M.; Song, W.; Zi, J.; Yang, Y.; Fan, X.; Shi, J.; Hu, J.; Sun, J.; Chen, N. *J. Nat. Prod.* **2008**, *71*, 647–654.
(13) Sakushima, A.; Ohno, K.; Maoka, T.; Coskun, M.; Guvenc, A.; Erdurak, C. S.; Ozkan, A. M.; Seki, K. I.; Ohkura, K. *Phytochem. Anal.* **2003**, *14*, 48–53.
(14) Zhao, C.; Chen, J.; Shao, J.; Shen, J.; Li, K.; Gu, W.; Li, S.; Fan, J. *J. Agric. Food Chem.* **2018**, *66*, 10421–10430.
(15) Lee, D.-Y.; Song, M.-C.; Yoo, K.-H.; Bang, M.-H.; Chung, I.-S.; Kim, S.-H.; Kim, D.-K.; Kwon, B.-M.; Jeong, T.-S.; Park, M.-H.; Baek, N.-I. *Arch. Pharm. Res.* **2007**, *30*, 402–407.
(16) Tan, R.; Jakupovic, J.; Jia, Z. J. *Planta Med.* **1990**, *56*, 475–477.
(17) Nakatani, M. *ChemInform* **1999**, *30*.
(18) Ahn, J.-W.; Yoo, G.-S.; No, Z.; Lee, S.-H. *Bull. Korean Chem. Soc.* **1993**, *14*, 554–556.
(19) Dong, S.-H.; Zhang, C.-R.; He, X.-F.; Liu, H.-B.; Wu, Y.; Yue, J.-M. *J. Nat. Prod.* **2010**, *73*, 1344–1349.
(20) Ito, J.; Chang, F. R.; Wang, H. K.; Park, Y. K.; Ikegaki, M.; Kilgore, N.; Lee, K. H. *J. Nat. Prod.* **2001**, *64*, 1278–1281.
(21) Borg-Karlson, A.-K.; Nordlander, G.; Mudalige, A.; Nordenhem, H.; Unelius, C. R. *J. Chem. Ecol.* **2006**, *32*, 943–957.
(22) Minhas, R.; Bansal, Y.; Bansal, G. *Med. Res. Rev.* **2020**, *40*, 823–855.
(23) Liu, Q. B.; Huang, X. X.; Bai, M.; Chang, X. B.; Yan, X. J.; Zhu, T.; Zhao, W.; Peng, Y.; Song, S. J. *J. Agric. Food Chem.* **2014**, *62*, 7796–7803.
(24) Cao, G. Y.; Xu, W.; Yang, X. W.; Gonzalez, F. J.; Li, F. *Food Chem.* **2015**, *173*, 231–237.
(25) Kim, K. H.; Choi, J. W.; Ha, S. K.; Kim, S. Y.; Lee, K. R. *Bioorg. Med. Chem. Lett.* **2010**, *20*, 409–412.

(26) Jiao, W.-H.; Gao, H.; Zhao, F.; He, F.; Zhou, G.-X.; Yao, X.-S. *Chem. Biodivers.* **2011**, *8*, 1163–1169.

Received January 11, 2023
Revised February 12, 2023
Accepted February 13, 2023

Supplementary data

Meliasendanins E-J, Nor-neolignan Constituents from *Melia toosendan* and their Anti-inflammatory Activity

Jin Woo Lee*

College of Pharmacy, Duksung Women's University, Seoul 01369, Republic of Korea

*Corresponding to: Jin Woo Lee, College of Pharmacy, Duksung Women's University, Seoul 01369, Republic of Korea. Tel: +82-2-901-8841; e-mail: jwlee22@duksung.ac.kr

List of supplementary data

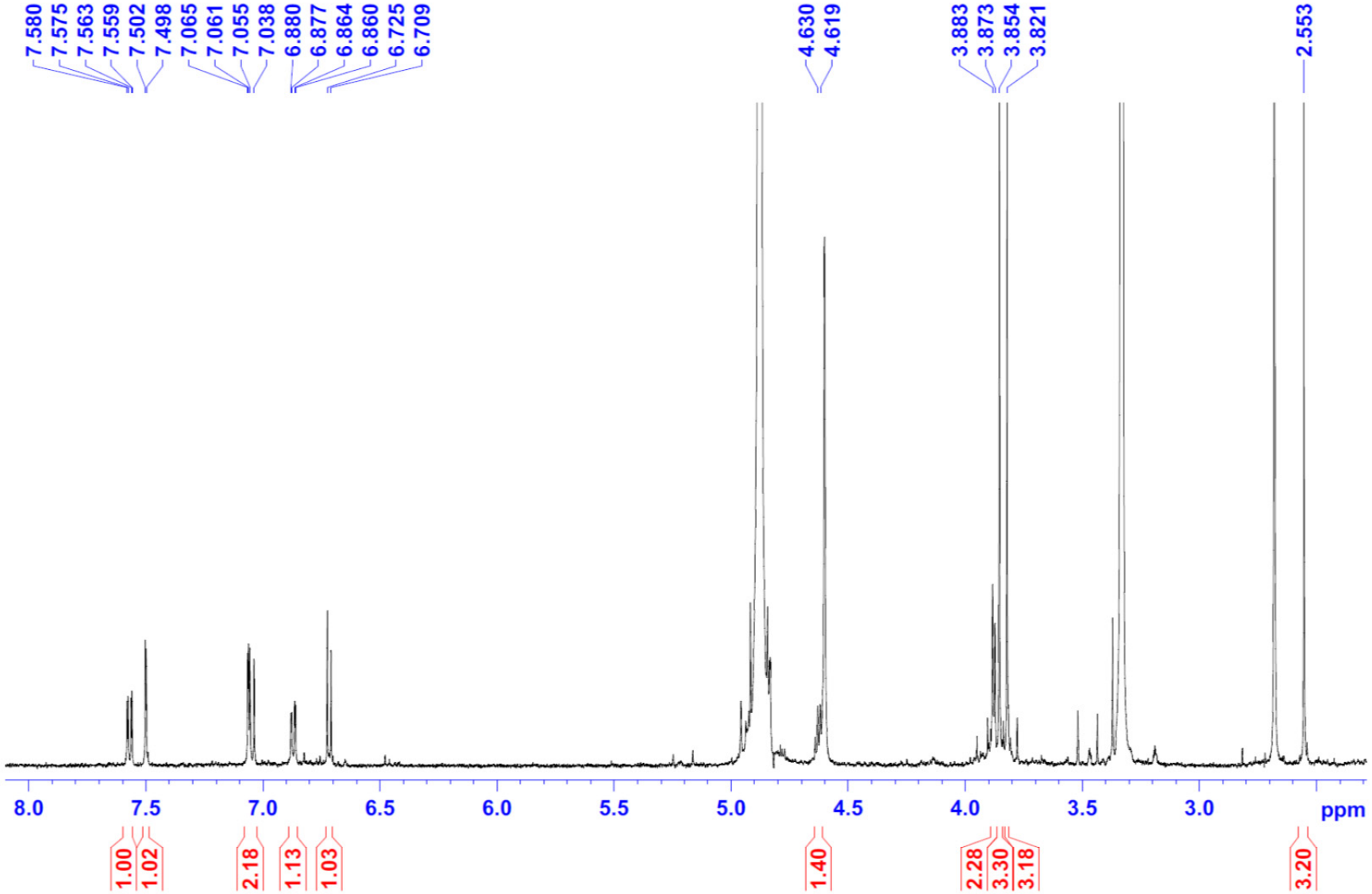


Figure S1. ^{1}H NMR spectrum (500 MHz, CD_3OD) of compound **1**

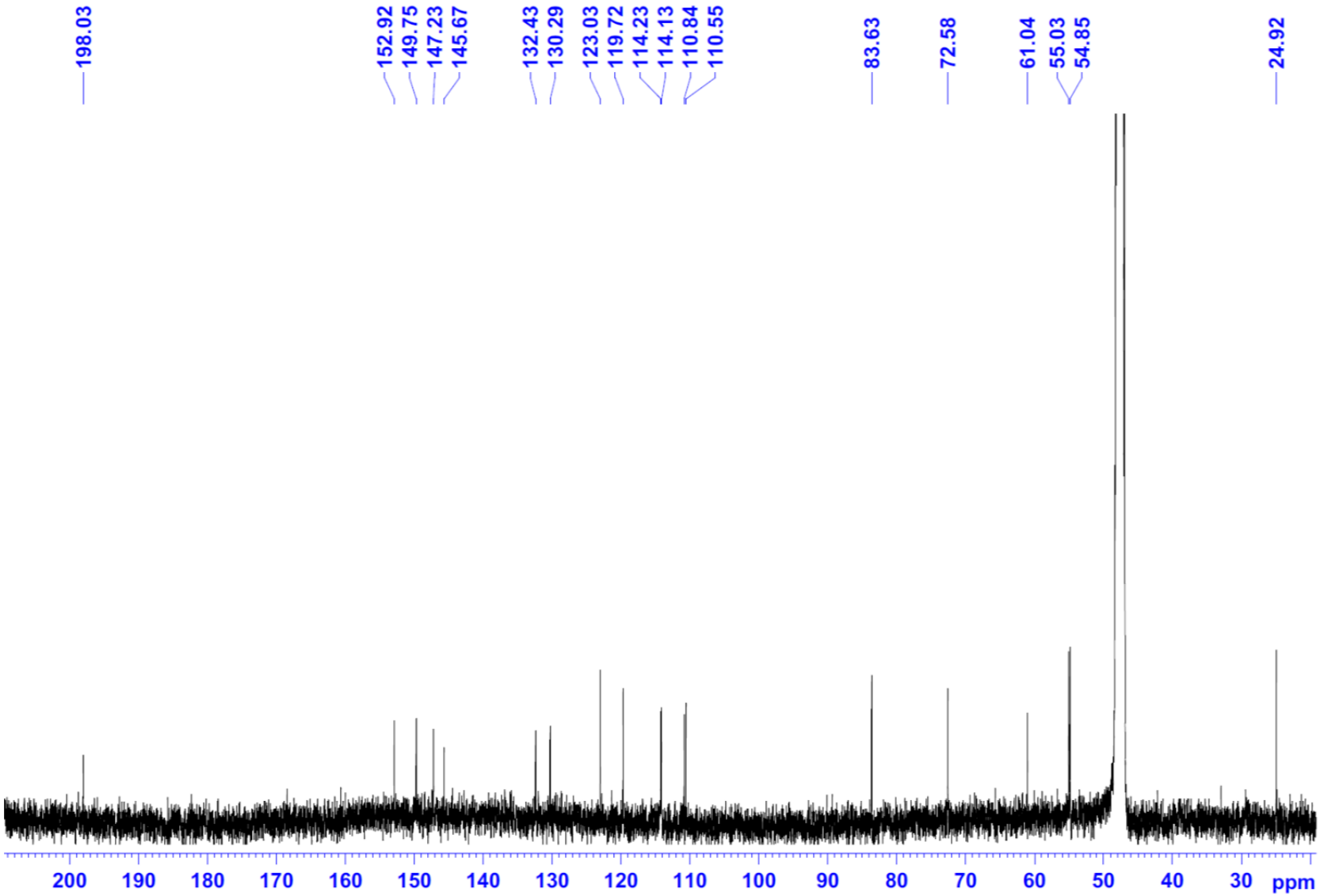


Figure S2. ^{13}C NMR spectrum (125 MHz, CD_3OD) of compound **1**

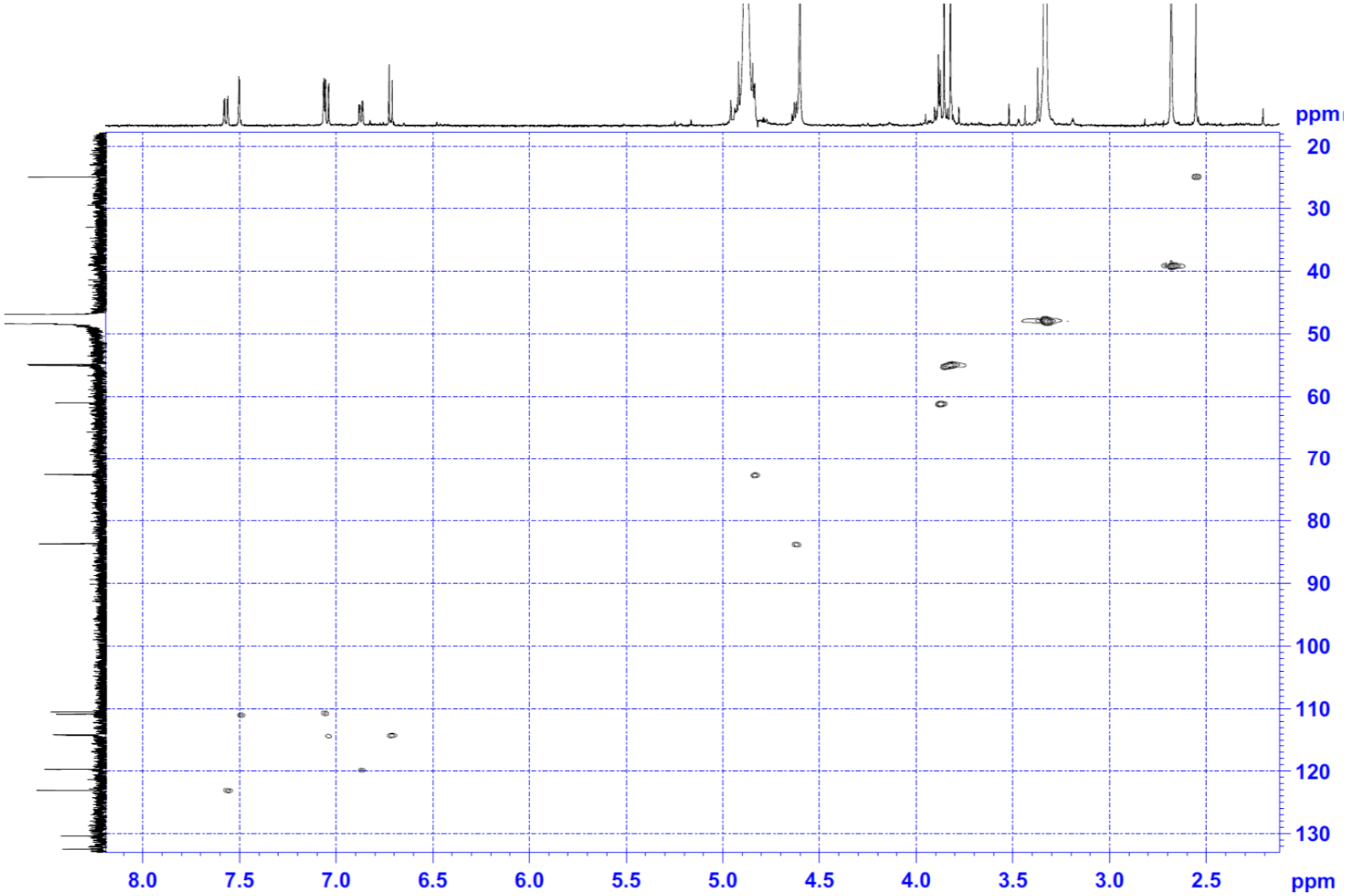


Figure S3. HSQC NMR spectrum (CD_3OD) of compound **1**

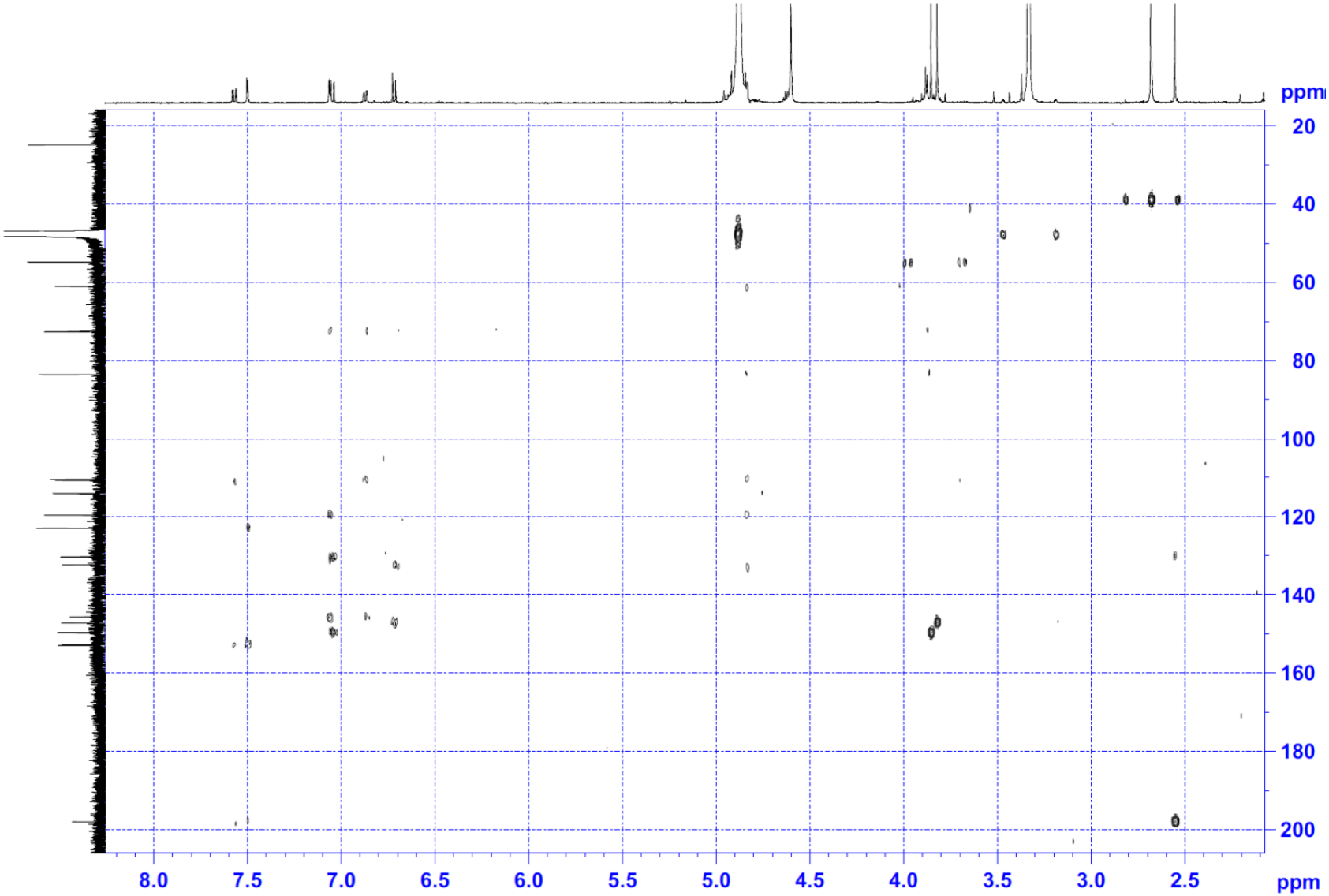


Figure S4. HMBC NMR spectrum (CD_3OD) of compound **1**

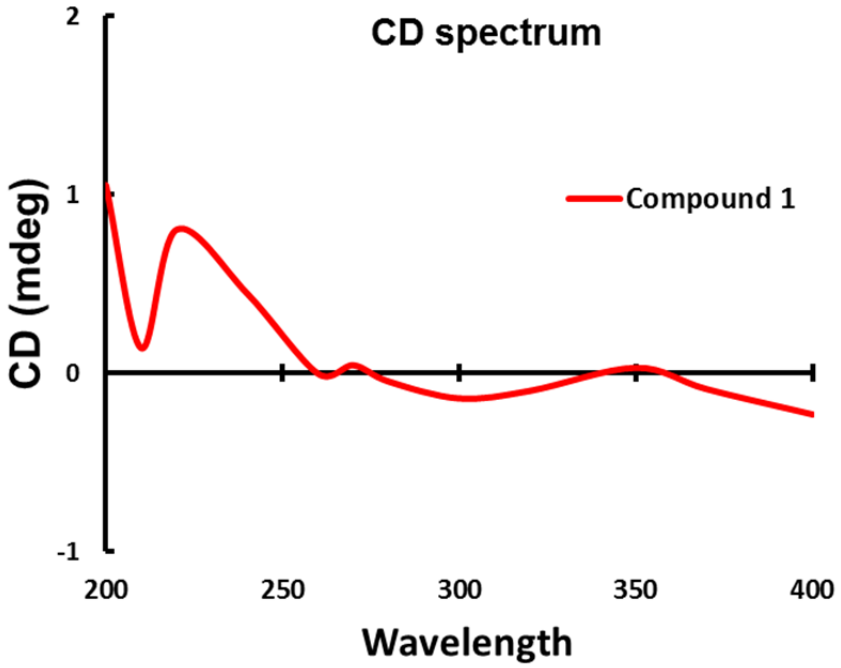
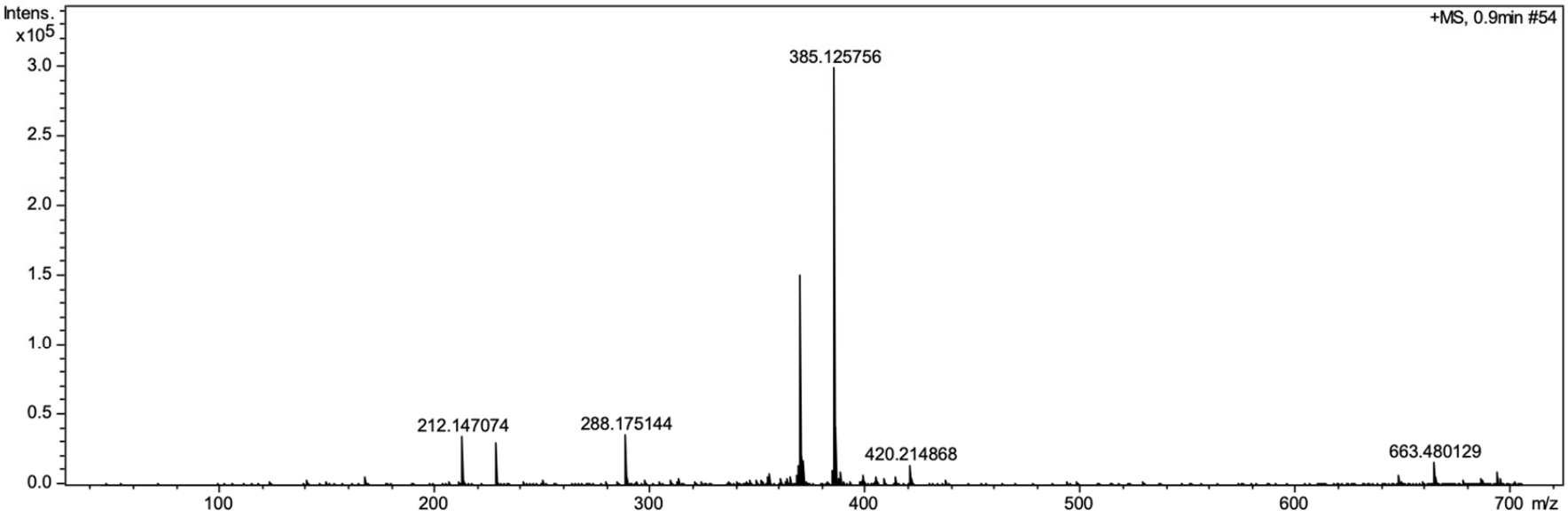


Figure S5. CD spectrum of compound **1**



Measured m/z 385.125756 Tolerance 2 ppm Charge 1

#	Mol. Formula	m/z	err [mDa]	|err| [ppm]	err [ppm]	mean err [ppm]	mSigma	Sigma Rank	rdb	N rule	e⁻
1	C 19 H 22 Na O 7	385.125774	0.02	0.0	0.0	0.0	40.0	2	8.5	ok	even

Figure S6. HRESIMS spectrum of compound **1**

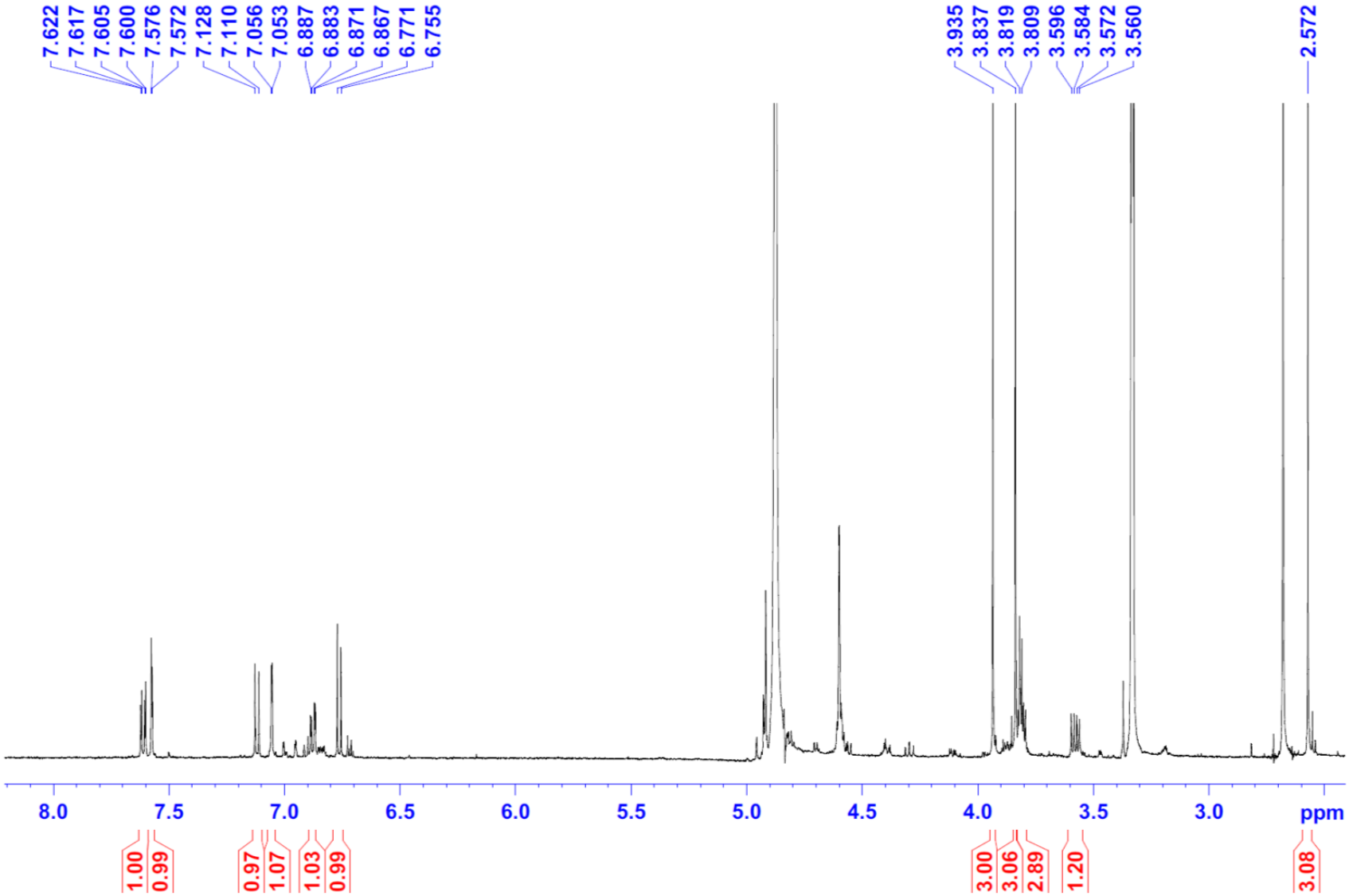


Figure S7. ^{1}H NMR spectrum (500 MHz, CD_3OD) of compound **2**

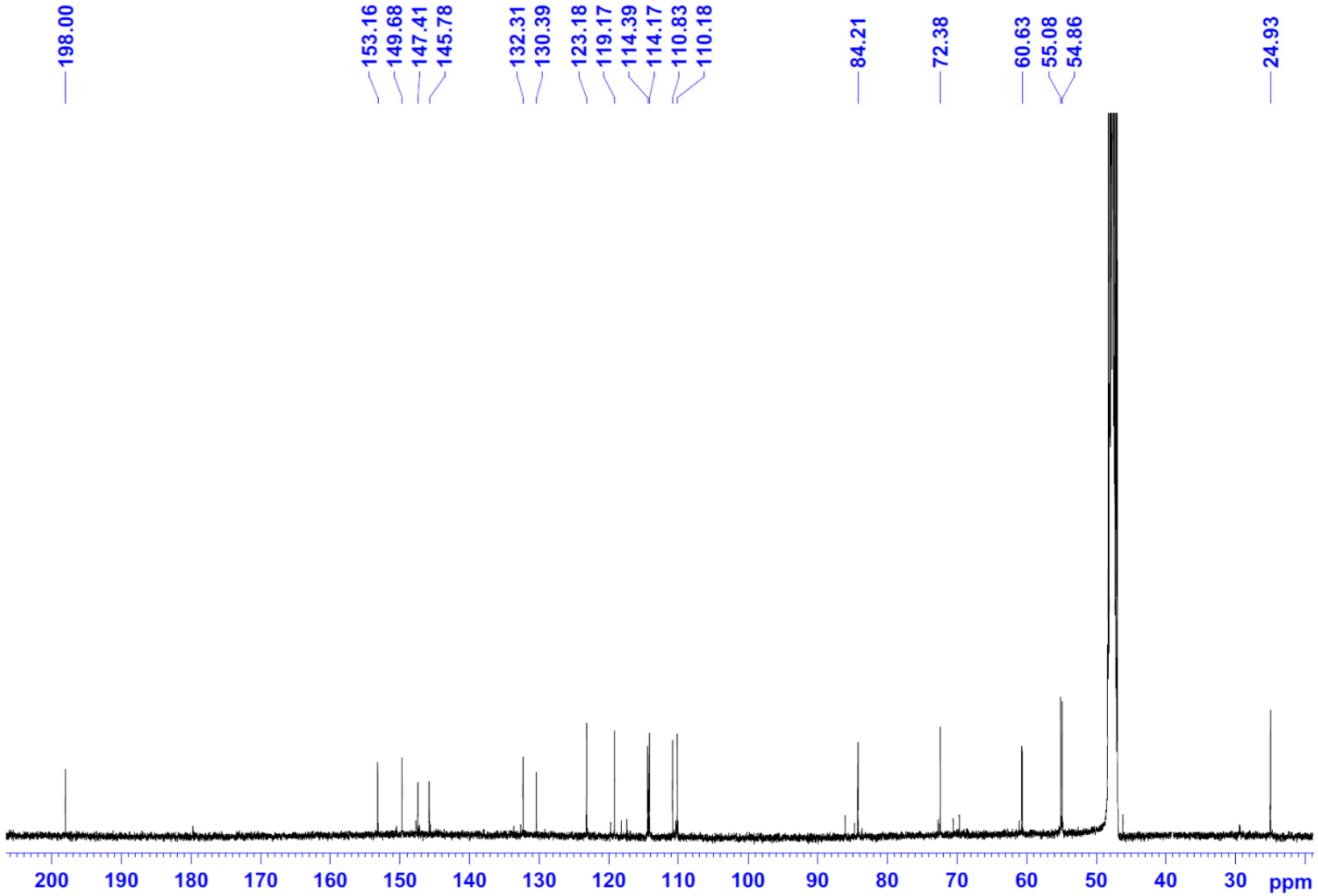


Figure S8. ^{13}C NMR spectrum (125 MHz, CD_3OD) of compound **2**

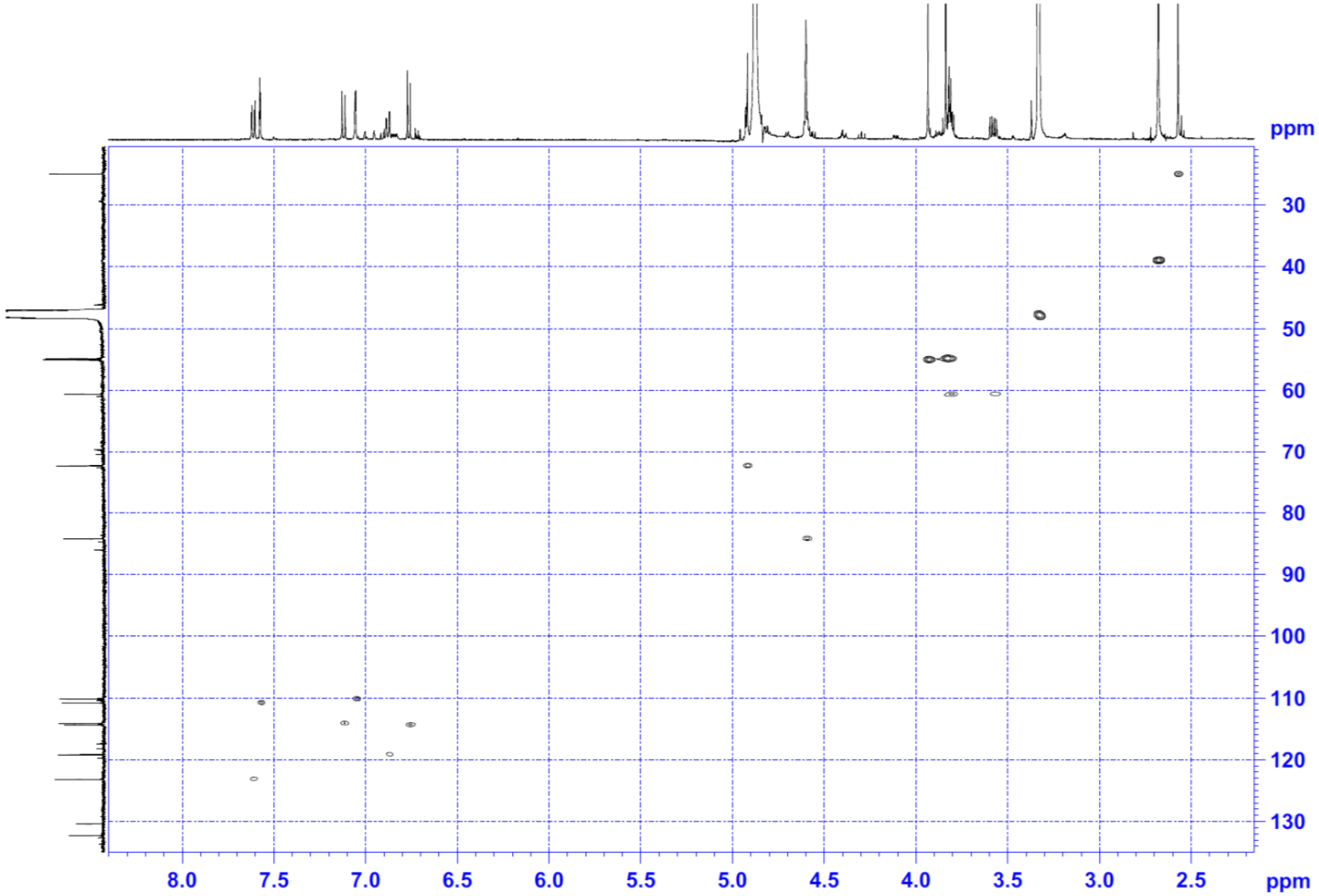


Figure S9. HSQC NMR spectrum (CD_3OD) of compound **2**

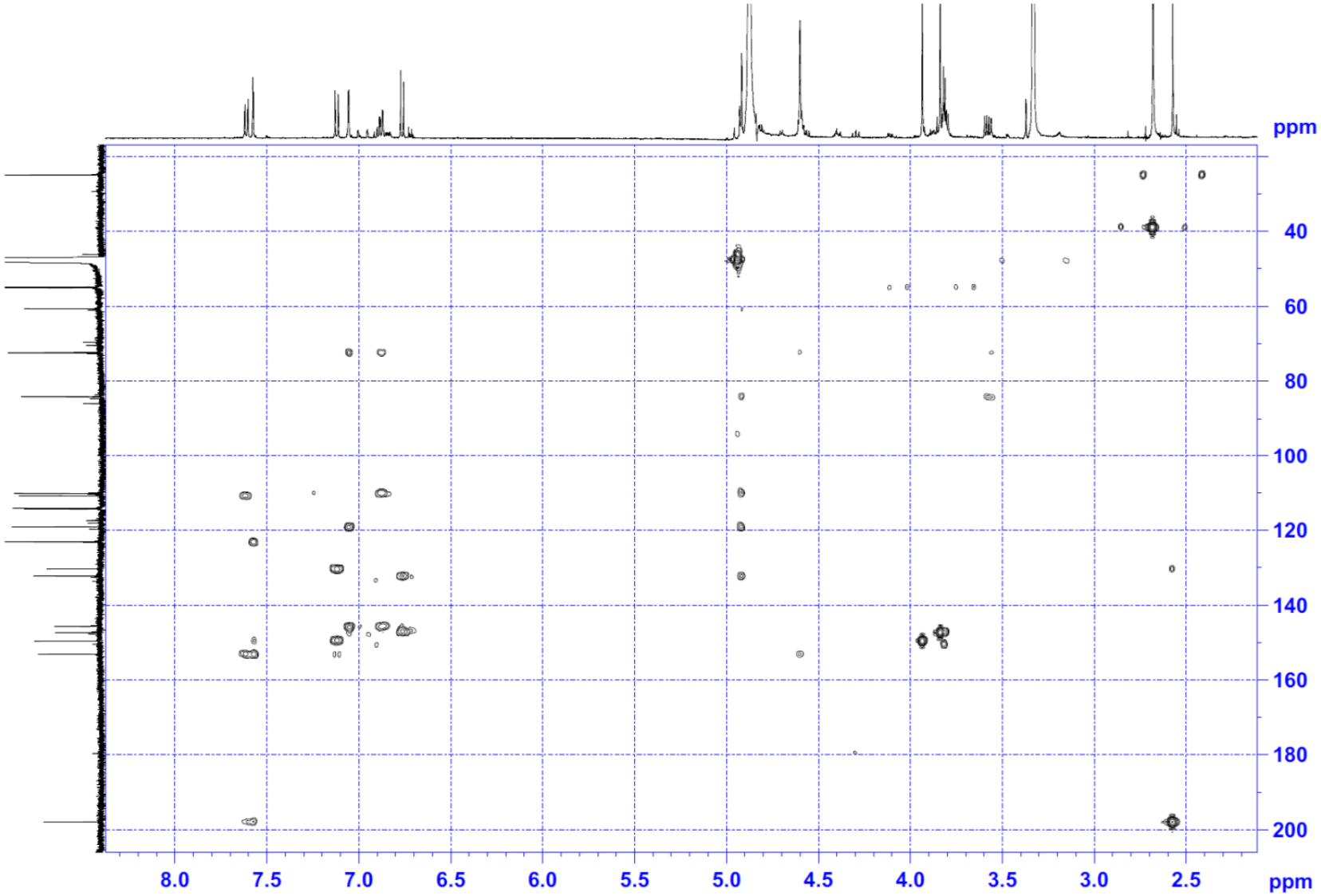


Figure S10. HMBC NMR spectrum (CD_3OD) of compound **2**

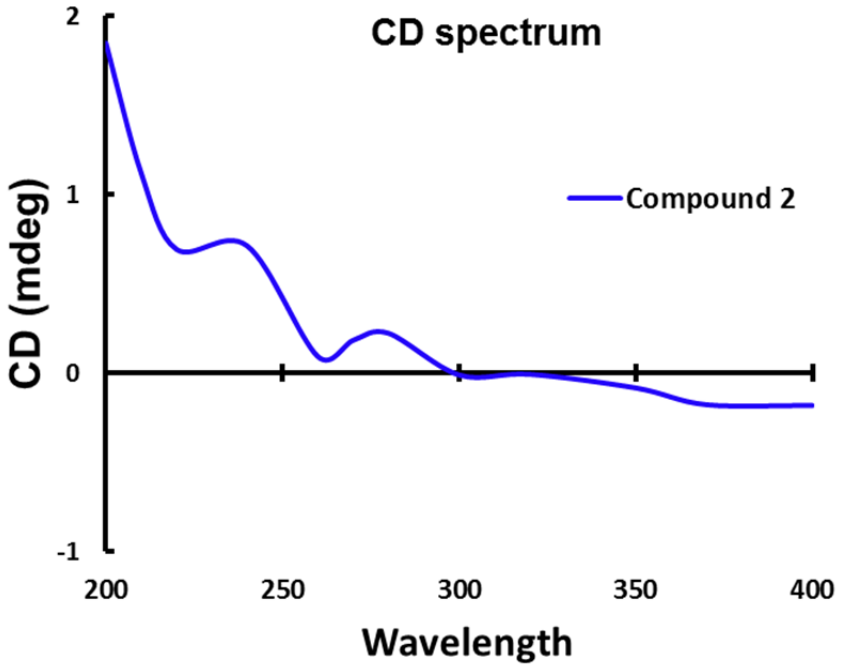


Figure S11. CD spectrum of compound **2**

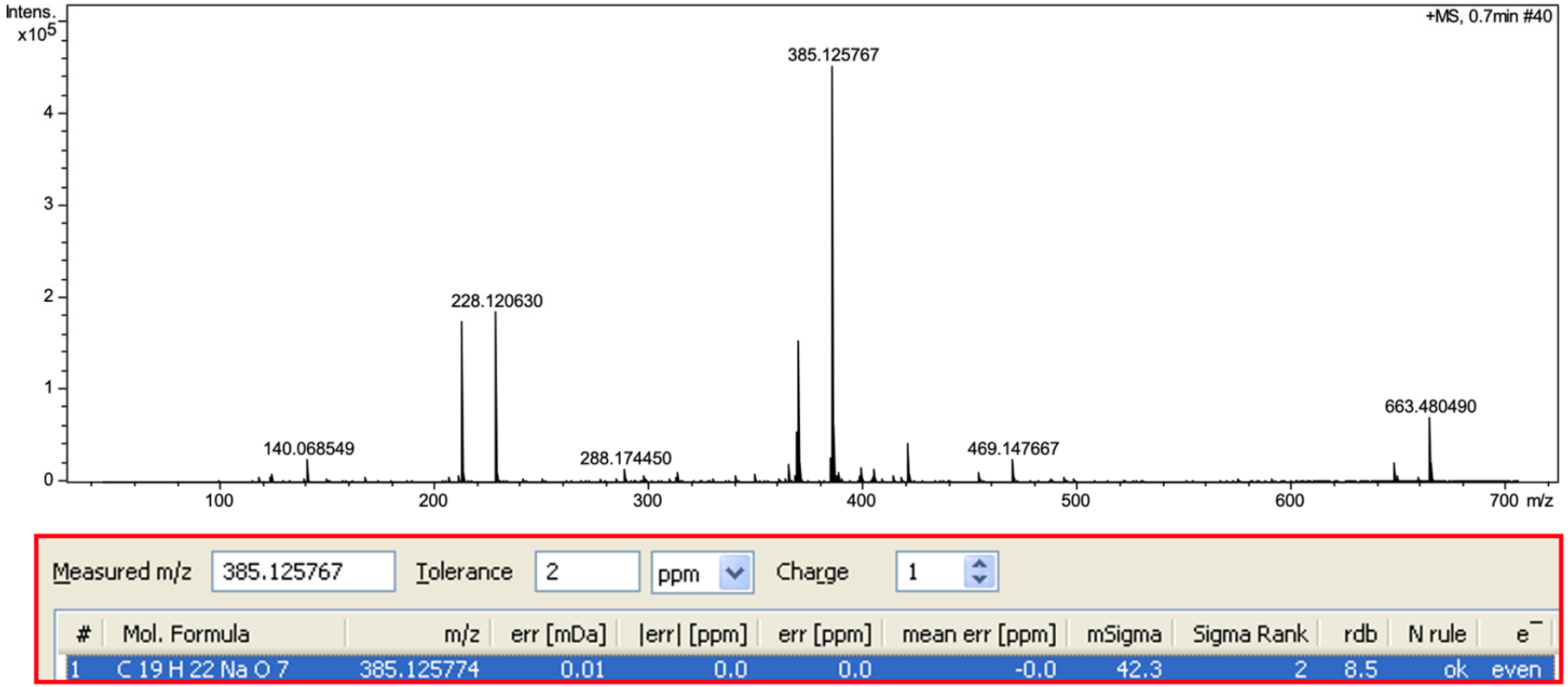


Figure S12. HRESIMS spectrum of compound **2**

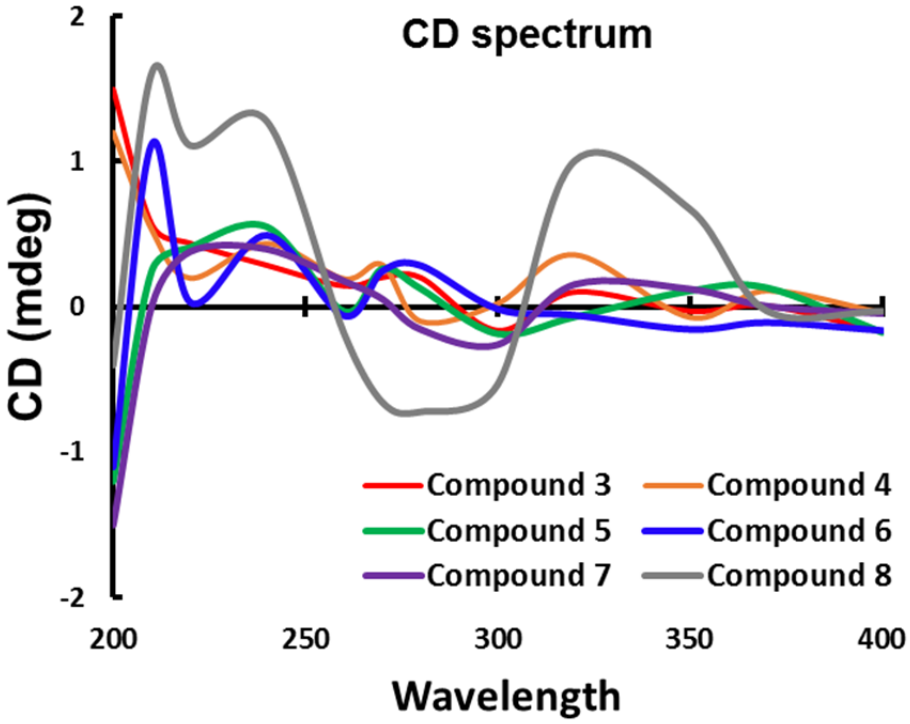


Figure S13. CD spectrum of compounds **3-8**

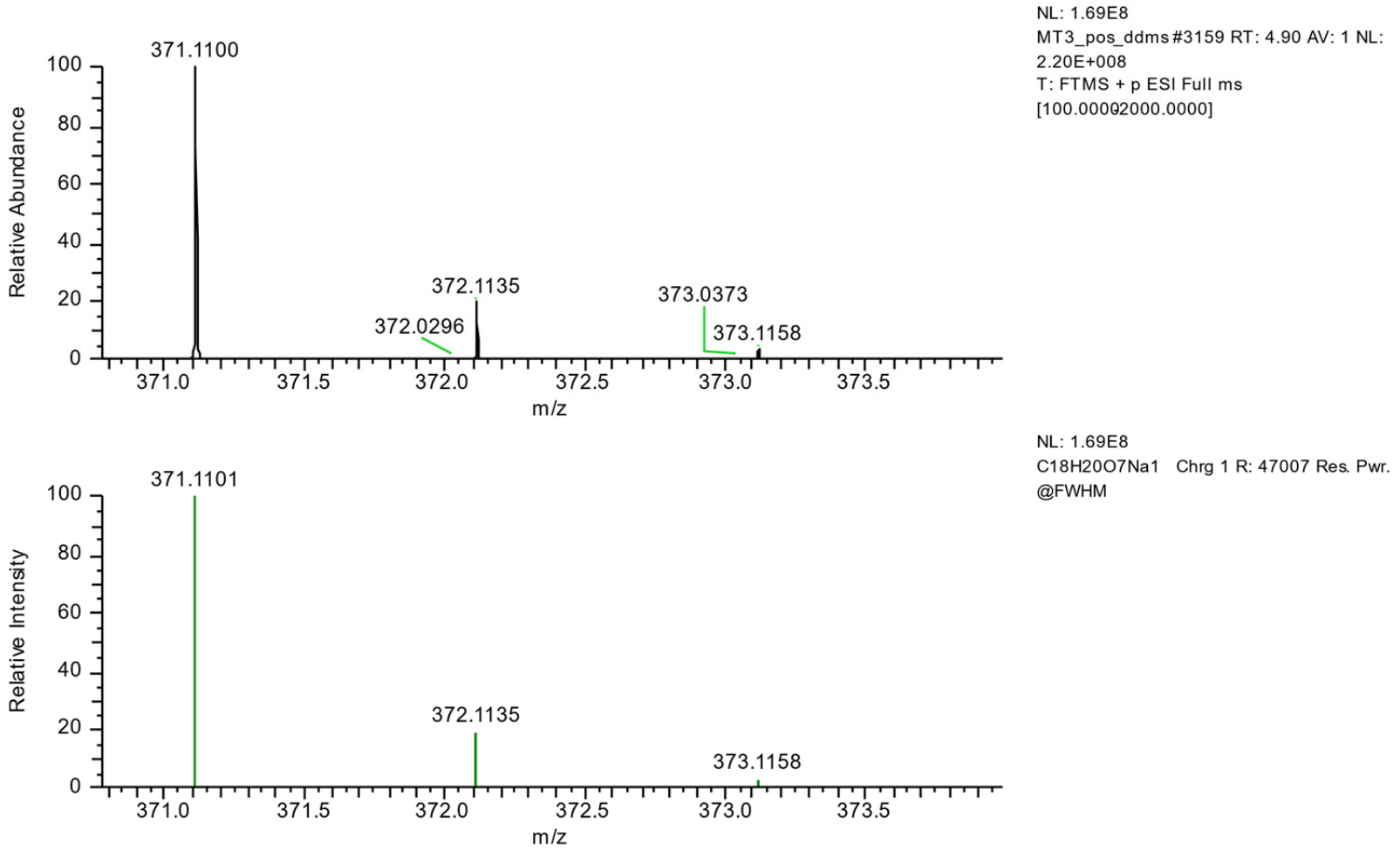


Figure S14. HRESIMS spectrum of compound **3**

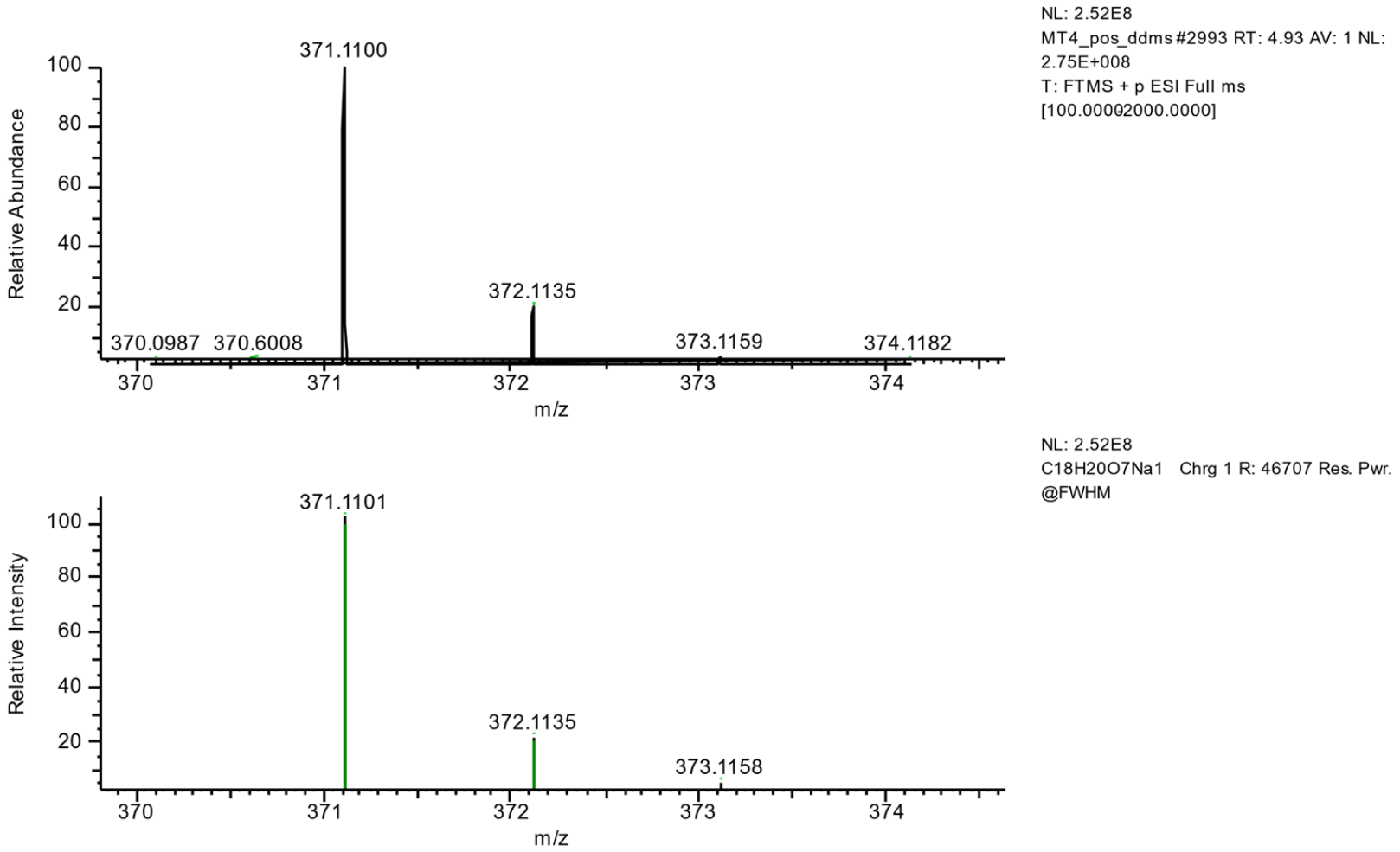


Figure S15. HRESIMS spectrum of compound **4**

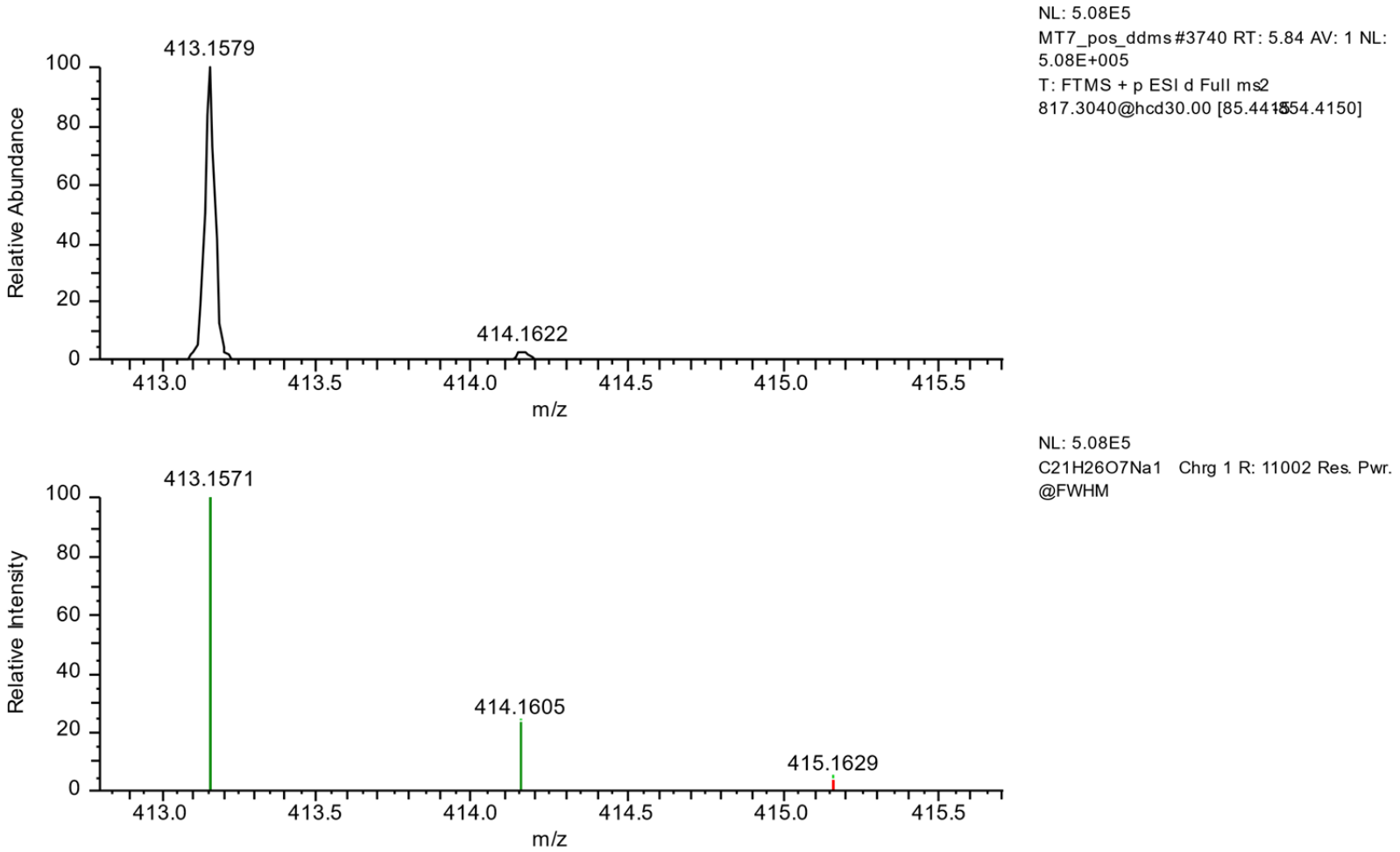


Figure S16. HRESIMS spectrum of compound **5**

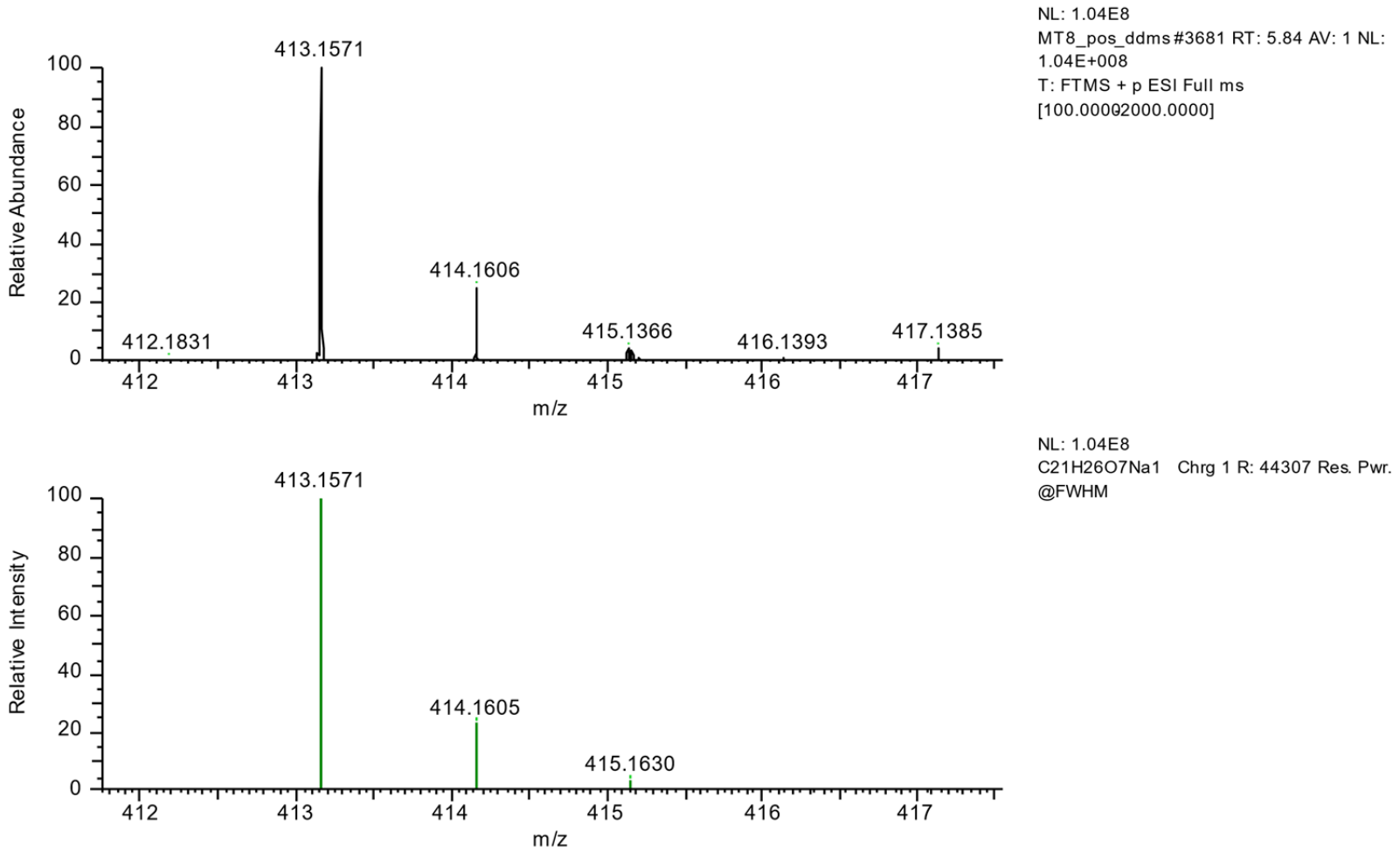


Figure S17. HRESIMS spectrum of compound **6**