Supplementary Information

Spoxazomicin D and Oxachelin C, Potent Neuroprotective Carboxamides from the Appalachian Coal Fire-Associated Isolate *Streptomyces* sp. RM-14-6

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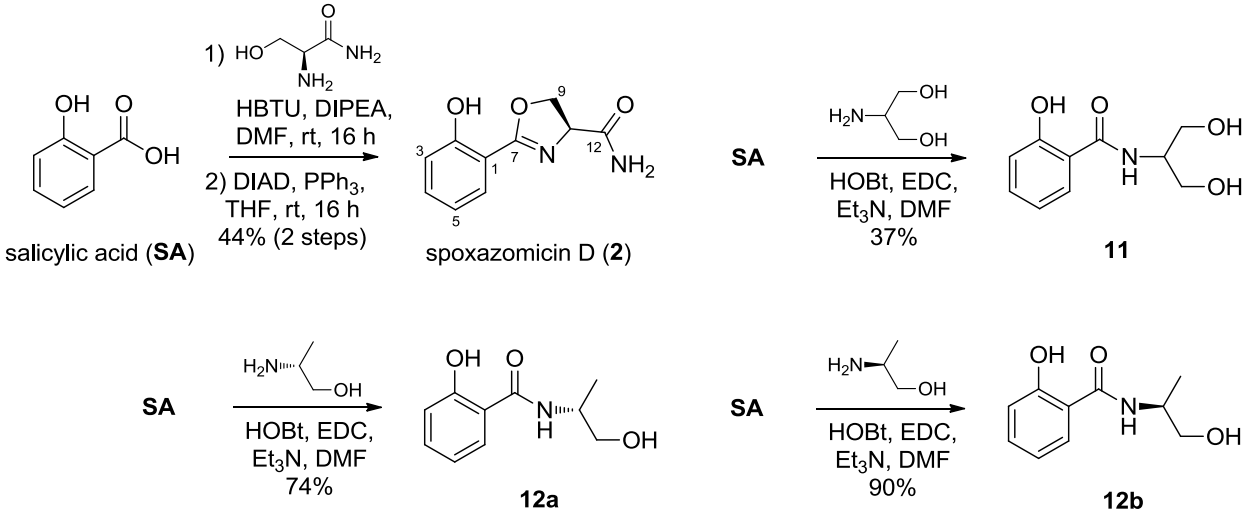
Synthesis of Compounds 2, 11 and 12a-b



Scheme S1. Synthesis of compounds **2**, **11** and **12a-b**

Spoxazomicin D (2). To a solution of salicylic acid (145 mg, 1.05 mmol), L-serine amide hydrochloride (140 mg, 1.0 mmol), DIPEA (360 uL, 2 mmol) in anhydrous DMF (5 mL), and HBTU (410 mg, 1.10 mmol) was added (Scheme S1). The resulting mixture was stirred at rt overnight. After evaporating the volatiles, the residue was purified via silica gel chromatography using a gradient of 20:1-15:1 CH_2Cl_2/MeOH to afford the crude product as a colorless solid (157 mg, 70%). The crude product and PPh_3 (211 mg, 0.8 mmol) was dissolved in anhydrous THF (14 mL) and the mixture cooled to 0 ^oC to which diisopropyl azodicarboxylate (152 uL, 0.77 mmol) was added in dropwise fashion over 10 min with stirring. The resulting mixture was allowed to warm to rt and continued with stirring overnight. After evaporating the volatiles, the residue was partially purified via silica gel using 1:1 *n*-hexane/EtOAc to afford a pale yellow oil containing the desired product and residual triphenylphosphine oxide. The product was further purified via Sephadex LH-20 (MeOH) to give compound **2** as a colorless solid (63 mg, 44%, 2 steps). Determined ^1H NMR, ^{13}C NMR, optical rotation and HRMS was consistent with the isolated natural product, spoxazomicin D (see main text file for NMR and MS data).

***N*-salicyloyl-2-aminopropan-1,3-diol (11).** To a solution of salicylic acid (0.10 g, 0.72 mmol) and serinol (0.65 g, 0.72 mmol) in DMF (10 mL), Et_3N (253 µL, 1.8 mmol), HOBt (0.15 g, 1.09 mmol) and EDC (0.21 g, 1.09 mmol) were added (Scheme S1). The reaction mixture was stirred at room temperature for 6 h. The crude mixture was subsequently concentrated *in vacuo* and

purified via silica column chromatography using a gradient of 1:1–9:1 hexane/EtOAc to afford desired product (0.06 g, 0.18 mmol, 37%). Determined ^{1}H NMR, ^{13}C NMR, optical rotation and HRMS was consistent with previously reported data for the isolated natural product **11**.[1]

(*R*)-(−)-*N*-salicyloyl-2-aminopropan-1-ol (12a). Following the protocol described for **11**, a reaction containing salicylic acid (0.10 g, 0.72 mmol), (*R*)-**(−)**-2-amino-1-propanol (0.054 g, 0.72 mmol), Et_3N (253 µL, 1.8 mmol), HOBt (0.15 g, 1.09 mmol) and EDC (0.21 g, 1.09 mmol) afforded 0.105 g desired product (0.54 mmol, 74%) (Scheme S1). Determined ^{1}H NMR, ^{13}C NMR, optical rotation and HRMS was consistent with previously reported data for the isolated natural product **12a**.[1,2]

(*S*)-(+)-*N*-salicyloyl-2-aminopropan-1-ol (12b). Following the protocol described for **11**, a reaction containing salicylic acid (0.10 g, 0.72 mmol), (*S*)- **(+)**-2-amino-1-propanol (0.054 g, 0.72 mmol), Et_3N (253 µL, 1.8 mmol), HOBt (0.15 g, 1.09 mmol) and EDC (0.21 g, 1.09 mmol) gave 0.128 g desired product (0.66 mmol, 90%) (Scheme S1). $[\alpha]^{25}_{D}$ = + 18.3° *(c* =1.0, MeOH); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 12.23 (brs, 1 H), 7.30 - 7.42 (m, 2 H), 6.97 (dd, *J* = 8.2, 0.8 Hz, 1 H), 6.76 - 6.90 (m, 1 H), 6.28 - 6.55 (m, 1 H), 4.29 (brs, 1 H), 3.80 (dd, *J* = 11.0, 3.5 Hz, 1 H), 3.67 (dd, *J* = 11.0, 4.7 Hz, 1 H), 1.59 (brs, 1 H), 1.30 (d, *J* = 6.7 Hz, 3 H); ^{13}C NMR ($CDCl_3$, 100 MHz): δ 170.2, 161.2, 134.4, 126.1, 119.0, 118.5, 114.5, 66.2, 47.6, 17.1 ppm.

EtOH damage neuroprotection assay

Organotypic Hippocampal Slice Culture Preparation. Entire brains were collected from eight-day-old Sprague-Dawley rats (Harlan Laboratories; Indianapolis, IN) and placed in chilled dissecting medium composed of Minimum Essential Medium (MEM; Invitrogen, Carlsbad, CA), 25 mM HEPES (Sigma, St. Louis, MO), and 50 µM streptomycin/penicillin (Invitrogen). Bilateral hippocampi were extracted, cleaned of excess tissue using a dissecting microscope, and sectioned at 200 µM using a McIlwain Tissue Chopper (Mickle Laboratory Engineering Co. Ltd., Gomshall, UK). Four intact hippocampi were plated onto Millicell-CM 0.4 µM biopore membrane inserts containing 1 mL of pre-incubated culture medium (dissecting medium, distilled water, 36 mM glucose [Fisher, Pittsburg, PA], 25% Hanks' Balanced Salt Solution [HBSS; Invitrogen], 25% v/v heat-inactivated horse serum [HIHS; Sigma], and 0.05% streptomycin/penicillin [Invitrogen]). Hippocampi were maintained in an incubator at 37 °C with a gas composition of 5% CO_2/95% air

for 5 days before any experiments were conducted. Care of all animals was carried out in accordance with the National Institutes of Health Guide for the Care and Use of Laboratory Animals (NIH Publications No. 80-23, revised 1996) and the University of Kentucky's Institutional Animal Care and Use Committee.

Ethanol and Compounds Treatment. At 5 DIV, slices were randomly transferred to new plates containing either 1 mL of standard culture medium (control) or culture medium with a calculated ethanol concentration of 100 mM. In an attempt to minimize ethanol evaporation, all plates containing ethanol in the medium were surrounded by 50 mL of double-distilled water containing ethanol (at a concentration of 100 mM) in topless polypropylene containers, and plates devoid of ethanol in the medium were surrounded by 50 mL of double-distilled water. Containers were placed into plastic bags, filled to capacity with 5% CO_2/95% air, sealed and placed in a water-jacketed CO_2 incubator for 48 hrs. Prior work has demonstrated that rapid evaporation of ethanol occurs during the preparation such that the actual starting concentration of ethanol in medium is approximately 90 mM.[3,4]

In cultures exposed to compounds **1**-**5**, with or without the addition of ethanol, the compound was diluted in dimethyl sulfoxide to a final working concentration of 0.01% dimethyl sulfoxide in cell culture medium. Ethanol-exposed and ethanol-naïve cultures were also exposed to 0.1% dimethyl sulfoxide in cell culture medium. Each 1 mL of cell culture medium, for all groups, also contained the nucleic acid intercalating agent, propidium iodide (3.74 μM). Propidium iodide is a polar compound that is only able to enter cells with compromised membranes, after which it binds to DNA and is able to fluoresce when excited at 515-560 nm. Propidium iodide uptake is highly correlated with multiple other measures of cytotoxicity in cell culture.[5] After 48 hrs of incubation, all cultures were removed from incubators and imaged to assess the intensity of propidium iodide uptake.

Fluorescent Microscopy and Statistical Analyses. Fluorescent intensity of propidium iodide was measured using densitometric measurement of the entire hippocampal slice. Images were taken using SPOT Advanced version 4.0.2 software for Windows (W. Nuhsbaum Inc., McHenry, IL, USA) with a 5× objective on an inverted Leica DMIRB microscope (W. Nuhsbaum Inc.) fitted for fluorescence detection (mercury-arc lamp) and connected to a personal computer via a SPOT 7.2 color mosaic camera (W. Nuhsbaum Inc.). Propidium iodide has a maximum excitation

wavelength of 536 nm and was excited using a band-pass filter that excites the wavelengths between 515 and 560 nm. The emission of PI in the visual range is 620 nm. Fluorescent intensity was analyzed ImageJ software (National Institutes of Health, Bethesda, MD, USA) for the entirety of each hippocampal slice culture. Raw fluorescent values were analyzed using a one-way analysis of variance ANOVA to assess effects of ethanol and treatment of compounds on propidium iodide uptake. When appropriate, Tukey's post-hoc analyses were conducted. The significance level was set at $P < 0.05$. For purposes of graphical illustration, all data were converted to percentage of ethanol-naïve control values.

Unfolded Protein Response (UPR) Assays and Screening

General. The ASGR-Cluc primary screen (below) was performed in a single replicate at 1 μM in 1536-well plate format using forskolin as a positive control and vehicle (DMSO) as the negative control. Preliminary hits compounds were subsequently subjected for confirmation and counterscreen assays. Confirmation assays were performed in triplicate using the same format/protocol as the primary screen in the absence and presence of 200 nM tunicamycin (**Tm**), a natural product inhibitor of *N*-linked glycosylation that induces UPR and leads to cell cycle arrest in the G1 phase (Figure S1). As described below, we also assessed the ability of confirmed hit compounds to improve rat insulinoma (INS-1E) β cell viability and basal insulin secretion in the absence and presence of thapsigargin [**Tg**, a potent chemical stressor that induces ER stress and UPR by non-competitively inhibit the sarco/endoplasmic reticulum Ca^{2+} ATPase (SERCA); Figure S2].

ASGR-Cluc Assays. ASGR-Cluc cells (HEK293 cell line) that were maintained in growth medium (Dulbecco's modified Eagle's medium (DMEM) containing antibiotics and 10% fetal bovine serum (FBS) were detached using trypsin. After removing excess trypsin by gentle centrifugation (800 rpm for 5 min), the cells were resuspended in growth medium containing 2% FBS at a density of 500 cells/μL. Using an automated liquid dispenser, 5 μL of this solution was then dispensed to each well of 1536-well plates (Greiner, Monroe, NC, USA), which were pre-spotted with 20 nL of compound (1 μM final concentration). The plates were incubated for 48 h at 37°C with constant supply of 5% CO_2, and 95% humidity. After the incubation period, 2 μL of Cluc reagent (Cypridina Luciferase Assay Kit; BioLux, Vancouver, BC, Canada) was added and the amount of secreted ASGR-Cluc determined by luminescence using Envision (0.1 second/well,

Perkin-Elmer, San Jose, CA, USA). As a counterscreen, ASGR-Cluc cells (2500 cells/4 µL/well) were incubated at 37°C with constant supply of 5% CO_2, and 95% humidity for 24 h. After the incubation period, cells were supplemented with 1 µL of growth medium (2% FBS) containing 1 µM **Tm** (200 nM final concentration) using an automated liquid dispenser, incubated for an additional 24 h and secreted ASGR-Cluc determined as described.

Insulin Secretion and Cell Viability Assays. INS-1E β cells that were maintained in growth medium were detached using trypsin. After removing excess trypsin by gentle centrifugation (800 rpm for 5 min), the cells were resuspended in growth medium at a density of 250 cells/µL. Aliquots (40 µL) of this solution were dispensed to each well of 384-well plates (Greiner, Monroe, NC, USA) pre-spotted with 20 nL of test agents at various concentrations. The plates were incubated for 24 h at 37°C with constant supply of 5% CO_2, and 95% humidity. After the incubation period, cells were supplemented with 10 µL of growth medium or 10 µL of growth medium containing 150 nM thapsigargin (**Tg**, 30 nM final concentration), incubated for an additional 24 h and secreted insulin (in a 2 µL sample) determined via the HTRF Insulin Assay Kit (Cisbio Assay, Bedford, MA, USA) using FRET as per manufacturer instructions. Cell viability of corresponding cells was accomplished in parallel using CellTiter-Glo (Promega, Fitchburg, WI, USA) as per manufacturer instructions.

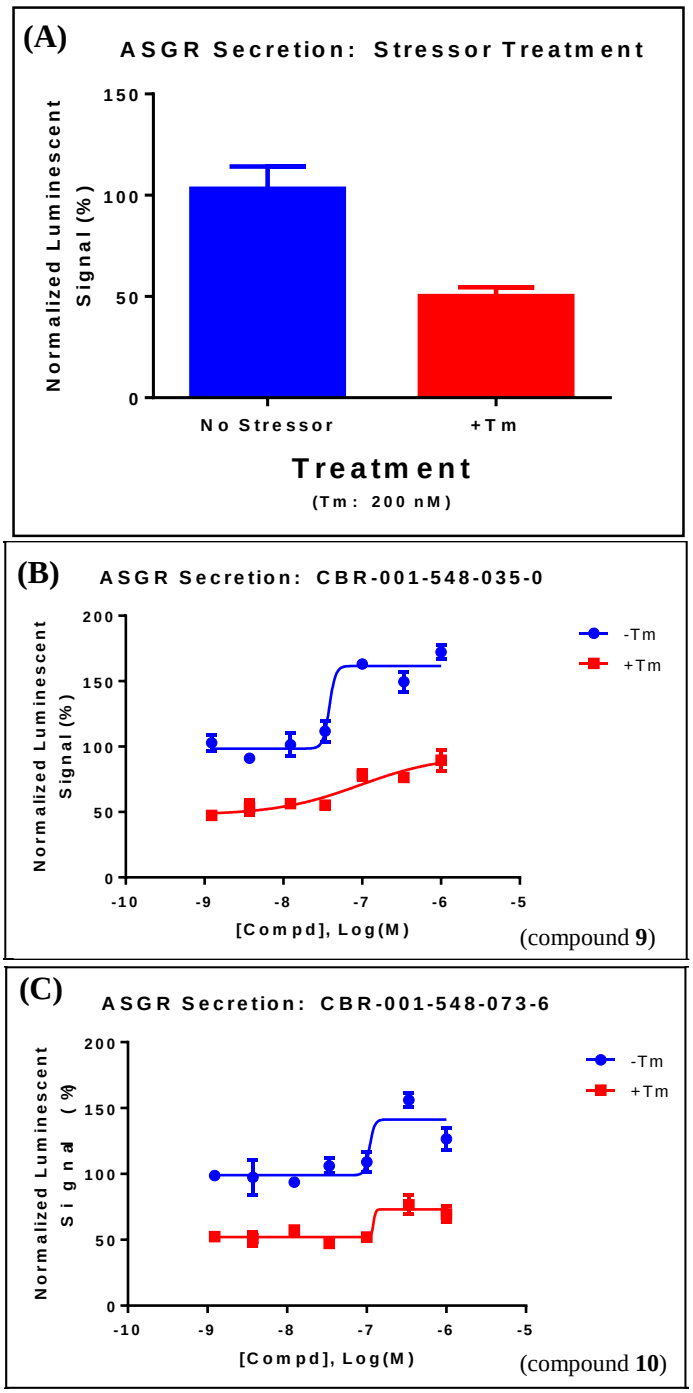


Figure S1. ASGR-Cluc assay in the absence and presence of Tunicamycin (**Tm**). **(A)** Effect of **Tm** on ASGR-Cluc secretion (200 nM **Tm** treatment for 24 h). **(B)** and **(C)** Dose response curves for hit compounds **9** and **10**, respectively, in the absence and presence of 200 nM **Tm**.

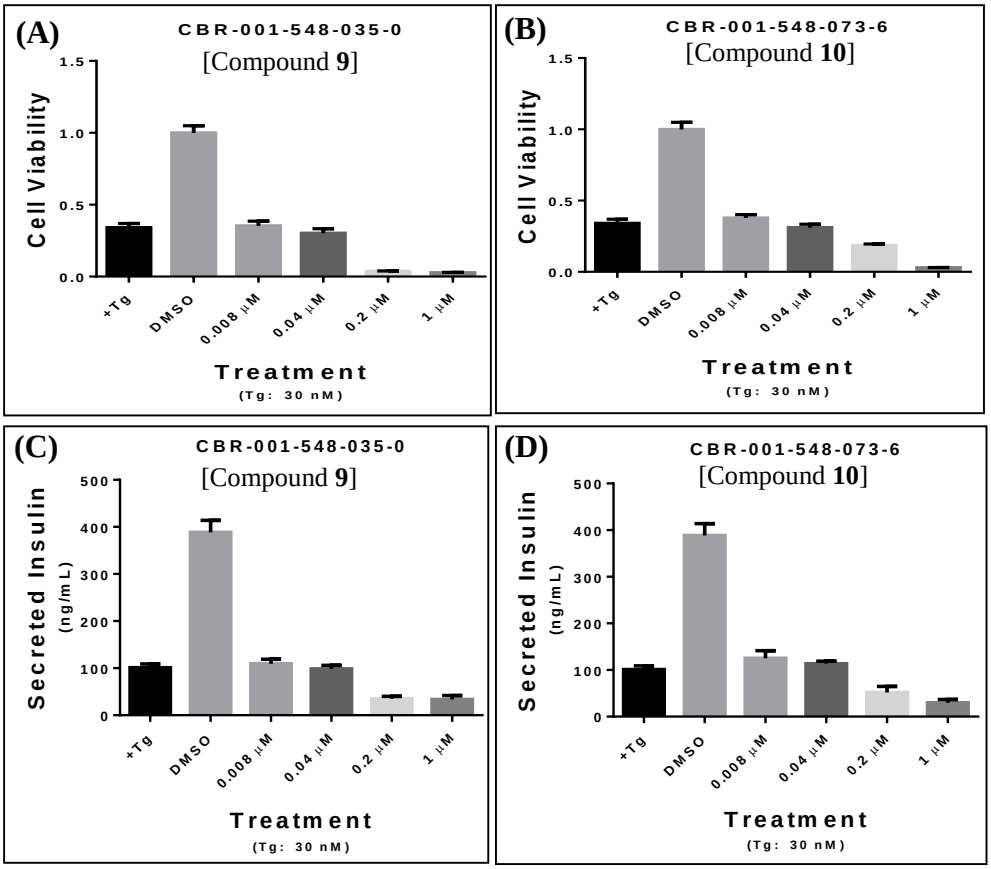


Figure S2. Modulation of INS-1E cell viability and insulin secretion. Cells were treated with various concentrations of hit compounds for 24 h. **Tg** (30 nM final concentration) was subsequently added and the cells incubated for additional 24 h. **(A)** and **(B)** INS-1E cell viability. **(C)** and **(D)** INS-1E cell insulin secretion.

Table S1. Determined EC_{50}s of compounds **9** and **10** from the ASGR-Cluc assay.

Compound	EC_{50} without **Tm** (nM)	EC_{50} with **Tm** (nM)
Lenoremycin (**9**)	38	93
Lenoremycin sodium salt (**10**)	109	122

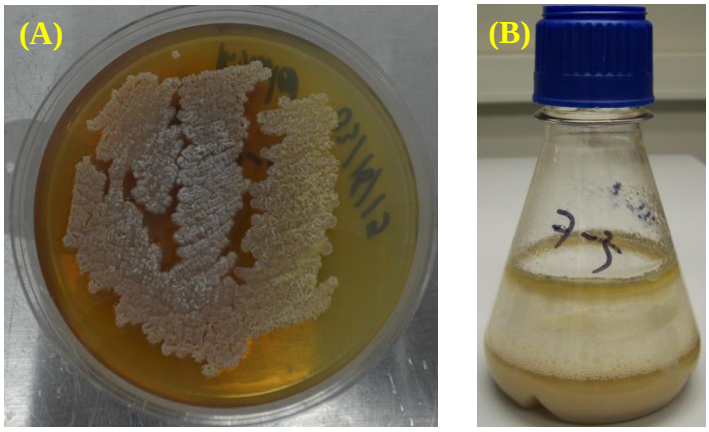


Figure S3. *Streptomyces* sp. RM-14-6 photographs. **(A)** Fully sporulated strain grown on a M_2-agar; **(B)** Representative liquid culture in A-medium after inoculation and cultivation at 28 °C and 210 rpm for 7 days.

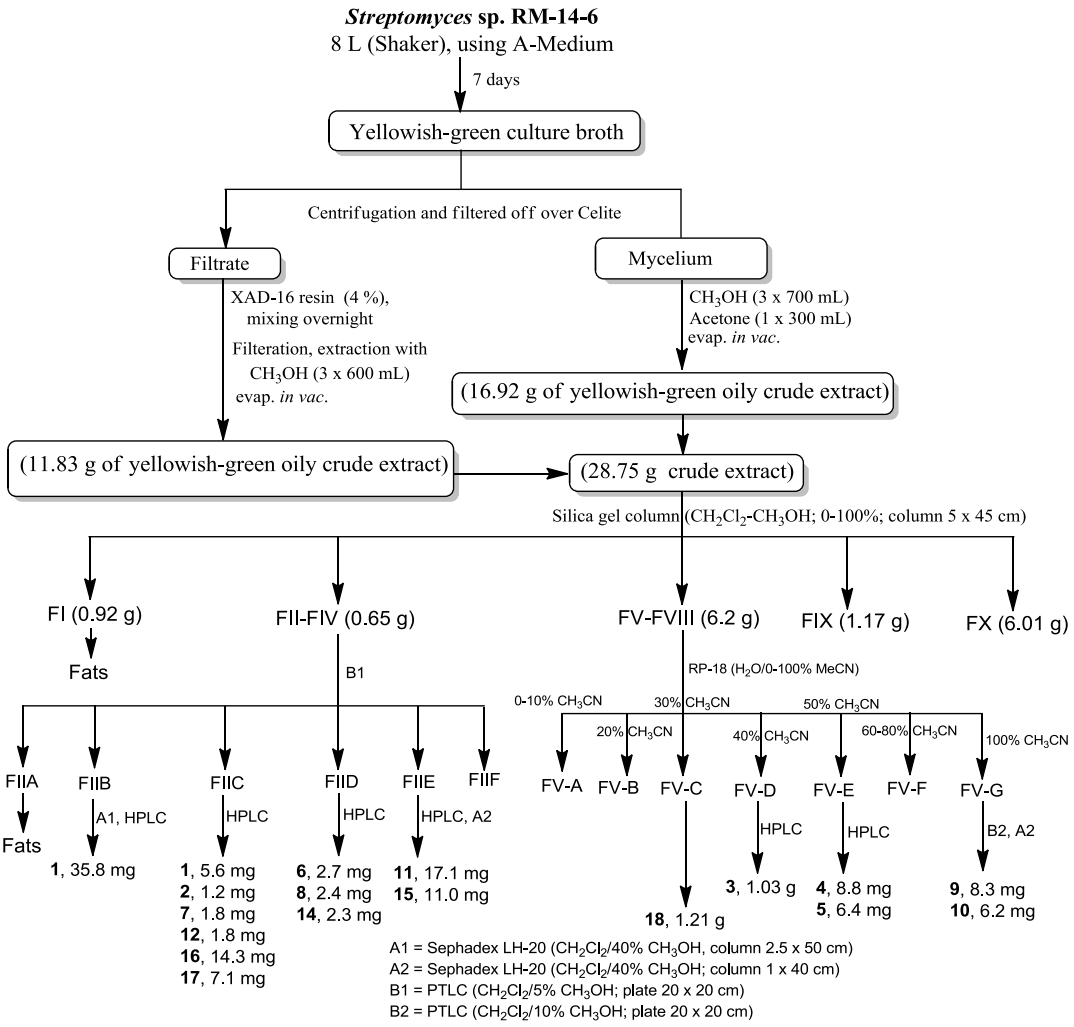
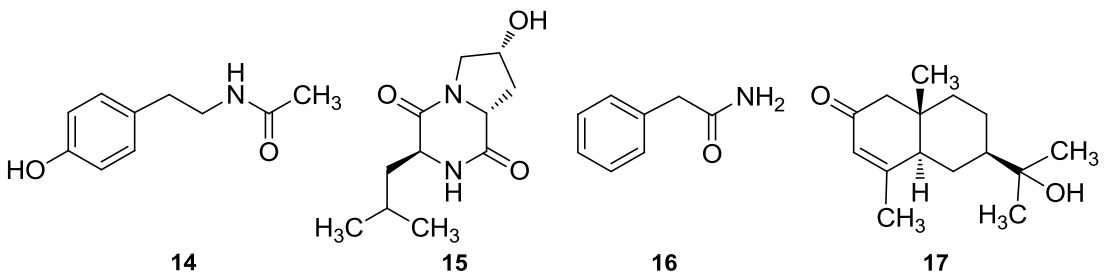


Figure S4. Work-up scheme of *Streptomyces* sp. RM-14-6.



14 15 16 17

Figure S5. Chemical structures of compounds **14-17** produced by *Streptomyces* sp. RM-14-6.

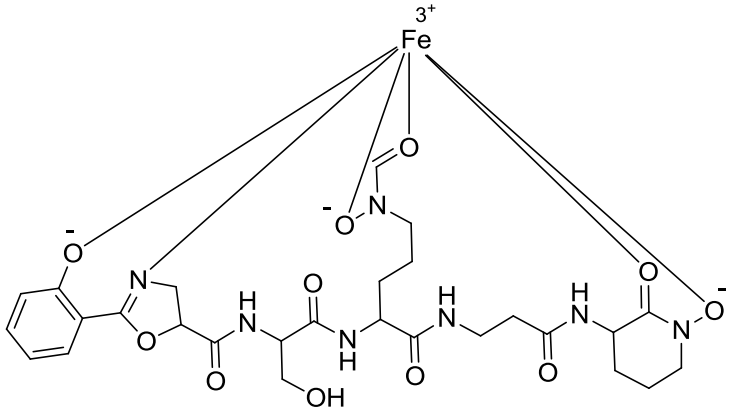


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Figure S6. Chemical structure of oxachelin-Fe-complex (**18**) based on HPLC/MS/UV data (Figure S34) and comparison with the reported X-ray crystal structure of the closely related amychelin-Fe-complex.[6]

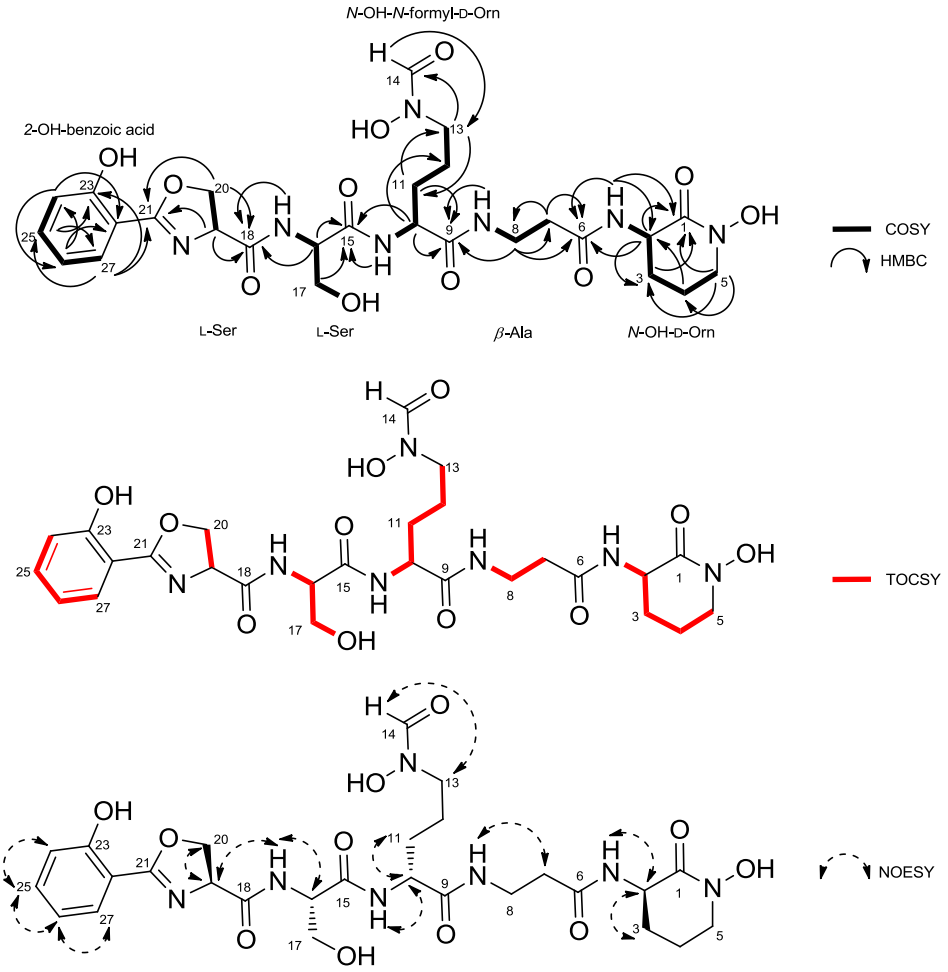

Figure S7. ^{1}H,^{1}H-COSY (▬), TOCSY (▬), selected HMBC (→) and NOESY (⋰⋱) correlations in oxachelin (**3**).

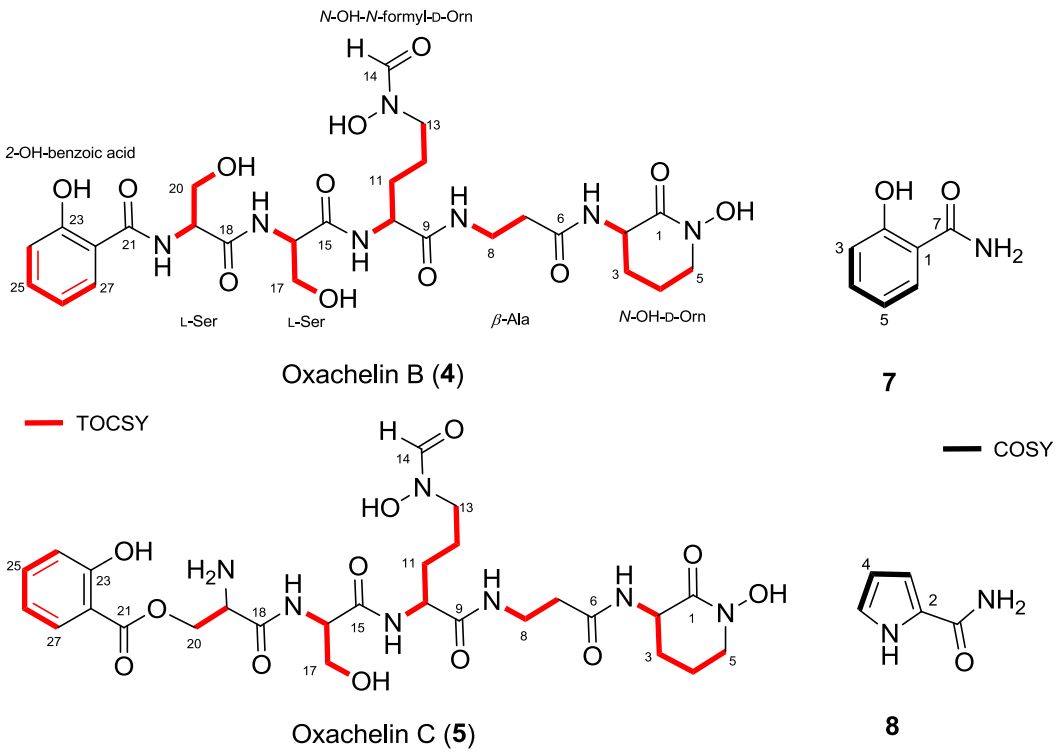

Figure S8. TOCSY (—) correlations in oxachelins B-C (**4**-**5**) and ^{1}H,^{1}H-COSY (—) correlations in compounds **7** and **8**.

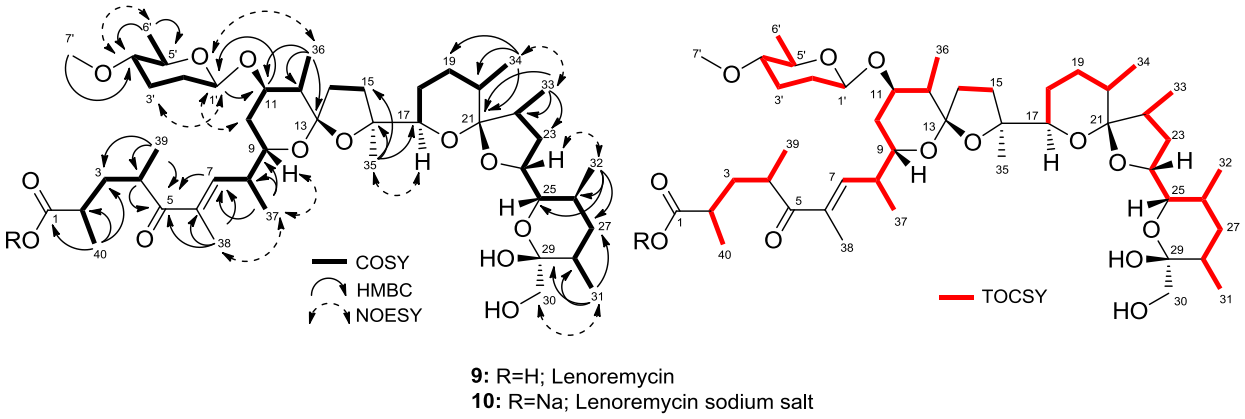

Figure S9. ^{1}H,^{1}H-COSY (—), TOCSY (—), selected HMBC (→) and NOESY (⋯) correlations in compounds **9** and **10**.

Table S2. ^{13}C and ^{1}H NMR data of lenoremycin (**9**) and lenoremycin sodium salt (**10**)[†]

Position	**10** (Literature)[7]	Lenoremycin (**9**)[a)]		Lenoremycin sodium salt (**10**)[a)]		
	δ_C[(b, c)], type	δ_C[(b, c)], type	δ_H[(b, d)], mult (*J* in Hz)	δ_C[(b, c)], type	δ_C[(c, e)], type	δ_H[(d, e)], mult (*J* in Hz)
1	181.3, C	181.6, C		178.1, C	181.8, C	
2	39.5, CH	39.7, CH	2.39, m	39.4, CH	40.5, CH	1.80, m
3	41.2, CH_2	41.3, CH_2	1.85, m, 0.87, m	40.0, CH_2	39.6, CH_2	1.80, m, 1.64, m
4	37.3, CH	37.9, CH	3.44, m	37.9, CH	38.7, CH	3.46, m
5	207.4, C	207.7, C		207.5, C	208.6, C	
6	134.0, C	134.3, C		137.5, C	137.5, C	
7	146.2, CH	146.3, CH	7.41, d (10.5)	145.1, CH	147.0, CH	6.60, d (9.5)
8	41.2, CH	41.4, CH	2.47, m	39.9, CH	40.3, CH	2.63, m
9	68.0, CH	68.3, CH	4.56, td (11.5, 1.5)	67.7, CH	68.7, CH	3.99, m
10	28.1, CH_2	28.2, CH_2	1.95, m, 1.78, m	28.2, CH_2	30.9, CH_2	1.85, m, 1.30, m
11	73.1, CH	73.2, CH	3.57, m	72.4, CH	74.5, CH	3.88, m
12	36.4, CH	36.7, CH	1.30, m	36.8, CH	39.9, CH	1.33, m
13	108.9, C	109.1, C		109.5, C	110.6, C	
14	35.6, CH_2	35.8, CH_2	1.95, m, 1.78, m	36.4, CH_2	37.2, CH_2	2.00, m, 1.80, m
15	32.2, CH_2	32.4, CH_2	1.95, m, 1.78, m	33.6, CH_2	35.2, CH_2	2.02, m, 1.90, m
16	85.8, C	86.0, C		86.7, C	87.4, C	
17	80.9, CH	81.1, CH	3.28, m	81.2, CH	81.6, CH	3.30, m
18	17.5, CH_2	17.7, CH_2	1.65, m, 1.20, m	17.8, CH_2	19.2, CH_2	1.60, m, 1.50, m
19	26.1, CH_2	26.3, CH_2	2.11, m, 1.11, m	25.9, CH_2	28.9, CH_2	1.65, m, 1.18, m
20	39.6, CH	39.9, CH	1.83, m	38.9, CH	39.9, CH	2.23, m
21	111.1, C	111.3, C		111.0, C	112.6, C	
22	35.1, CH	35.3, CH	2.57, m	35.2, CH	36.2, CH	2.63, m
23	29.8, CH_2	29.9, CH_2	2.40, m, 1.30, m	29.9, CH_2	31.9, CH_2	1.80, m, 1.55, m
24	79.4, CH	79.6, CH	4.37, ddd (11.5, 4.5, 2.0)	79.2, CH	80.0, CH	4.16, m
25	73.1, CH	73.3, CH	3.85, brdd (10.5, 1.5)	73.6, CH	78.7, CH	3.55, m
26	33.0, CH	33.2, CH	1.30, m	33.3, CH	31.4, CH	1.95, m
27	36.3, CH_2	36.5, CH_2	2.11, m, 1.30, m	36.4, CH_2	38.6, CH_2	1.50, m, 1.30, m
28	36.4, CH	36.7, CH	1.30, m	37.5, CH	34.8, CH	1.80, m
29	98.5, C	98.8, C		98.8, C	98.9, C	
30	64.1, CH_2	64.4, CH_2	4.03, brd (12.0, H_a), 3.15, brd (11.5, H_b)	66.0, CH_2	67.3, CH_2	3.45, m
31	17.1, CH_3	17.4, CH_3	0.91, d (6.5)	18.0, CH_3	16.8, CH_3	0.89, d (6.5)
32	17.8, CH_3	18.0, CH_3	0.83, d (6.5)	18.0, CH_3	18.9, CH_3	0.98, d (6.0)
33	15.3, CH_3	15.5, CH_3	0.97, d (7.0)	15.4, CH_3	16.5, CH_3	0.94, d (7.0)
34	13.9, CH_3	14.2, CH_3	1.05, d (6.5)	13.8, CH_3	14.5, CH_3	1.06, d (7.0)
35	27.0, CH_3	27.3, CH_3	1.45, s	27.2, CH_3	23.8, CH_3	1.32, s
36	13.9, CH_3	14.2, CH_3	1.12, d (7.0)	14.3, CH_3	13.8, CH_3	1.00, d (7.0)
37	14.6*, CH_3	17.3, CH_3	1.08, d (7.0)	17.3, CH_3	16.1, CH_3	1.09, d (6.0)
38	11.2, CH_3	11.5, CH_3	1.72, s	12.0, CH_3	12.4, CH_3	1.80, s
39	17.1*, CH_3	14.9, CH_3	1.02, d (6.5)	18.8, CH_3	19.7, CH_3	1.04, d (7.0)
40	20.3, CH_3	20.5, CH_3	1.08, d (7.0)	19.8, CH_3	19.0, CH_3	1.11, d (7.0)
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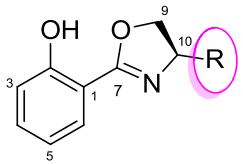
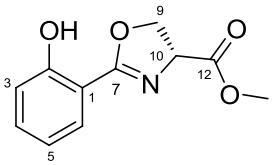
3'	27.5, CH_2	27.7, CH_2	2.40, m, 1.30, m	27.5, CH_2	28.1, CH_2	2.00, m, 1.30, m
4'	79.4, CH	79.6, CH	2.92, ddd (13.5, 11.0, 4.0)	80.5, CH	81.7, CH	2.85, m
5'	76.1, CH	76.3, CH	3.27, m	75.0, CH	76.3, CH	3.28, m
6'	18.3, CH_3	18.5, CH_3	1.42, d (6.0)	18.5, CH_3	18.9, CH_3	1.25, d (6.0)
7'	56.7, CH_3	57.0, CH_3	3.32, s	57.0, CH_3	57.2, CH_3	3.35, s

[a)]See figures S82-90 and S93-102 for the NMR spectra; [b)]$CDCl_3$; [c)]100 MHz; [d)]500 MHz; [e)]CD_3OD; *Wrongly assigned in literature. [†]The reported ^{13}C NMR data for **10** are closely related to our isolated **9** (not **10**), which might be incorrect reported.



1: R=CH_2OH; Spoxazomicin C
2: R=$CONH_2$; Spoxazomicin D



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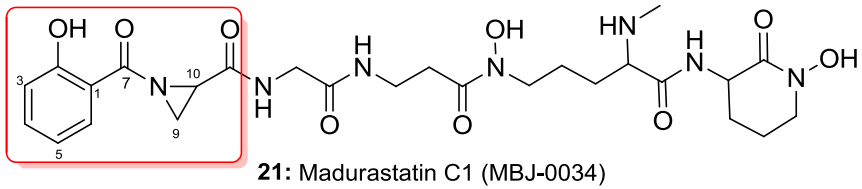


21: Madurastatin C1 (MBJ-0034)

Table S3. ^{1}H NMR spectroscopic data of isolated compounds **1-2** in comparison with the reported data of **1**, aziridine moiety of madurastatin C1 (**21**), and **13** (mult., *J* in [Hz]).

	Spoxazomicin C (Literature)[8]	Madurastatin C1 (aziridine moiety)[9]	**1**			**2**[a)]	**13**[a)] (Literature)[10]
Position	δ_H (300 MHz)[b)]	δ_H (400 MHz)[b)]	δ_H (400 MHz)[b)]	δ_H (500 MHz)[a)]	δ_H (400 MHz)[c)]	δ_H (400 MHz)	δ_H (400 MHz)
2-OH				11.92, brs	12.27, brs	11.28, brs	11.66, brs
3	6.93, dd (8.6, 0.9)	6.89, dd (8.3, 0.6)	6.93, dd (8.4, 0.8)	6.99, brd (7.5)	6.98, dd (7.6, 0.8)	7.02, d (8.5)	7.01, dd (8.5, 1.5)
4	7.37, ddd (8.4, 7.2, 1.5)	7.34, dt (8.2, 1.1)	7.37, ddd (8.4, 7.2, 1.6)	7.37, dt (8.5, 1.5)	7.34, ddd (8.8, 7.2, 1.6)	7.40, ddd (8.5, 7.0, 2.0)	7.38, ddd (8.5, 7.5, 1.5)
5	6.87, ddd (7.9, 7.4, 0.9)	6.82, dt (7.4, 0.5)	6.87, ddd (8.0, 7.2, 0.8)	6.86, ddd (7.5, 7.5, 0.8)	6.92, dt (8.0, 1.2)	6.91, ddd (8.0, 7.0, 1.0)	6.87, ddd (8.0, 7.5, 1.5)
6	7.63, dd (7.9, 1.5)	7.61, dd (7.7, 1.1)	7.64, dd (8.0, 2.0)	7.65, dd (8.0, 1.5)	7.61, dd (7.6, 1.6)	7.68, dd (8.0, 1.5)	7.65, dd (8.0, 1.5)
9	4.50, m	4.60, t (9.8)	4.48, m	4.49, m	4.49, dd (9.6, 8.0)	4.67, d (9.5, 2H)	4.67, dd (9.0, 7.5),
	4.33, m	4.53, t (8.4)	4.33, t (6.4)	4.35, t (6.0)	4.39, dd (7.2, 6.8)		4.56, dd (10.5, 9.0)
10	4.44, m	4.97, m	4.44, m	4.49, m	4.41, m	4.93 (t, 9.5)	4.97, dd (10.5, 7.5)
12	3.70, d (4.5)		3.70, d (4.0)	3.88, dd (11.0, 3.0), 3.69, dd (11.0, 3.0)	3.56, brs		
12-OCH_3							3.80, s
12-OH					4.96, brs		
12-NH_2						6.35 (brs), 5.61 (brs)	

[a)]($CDCl_3$); [b)]CD_3OD; [c)] DMSO-d_6); See figures S13-24 and S28-36 for the NMR spectra. Assignments supported by HSQC and HMBC experiments.

Table S4. *In vitro* antimicrobial activities of compounds **1-12** and **14-18**

Organism	**Compound** [MIC in μM (μg/mL)][a)]			
	1	**3**	**9**	**10**
Staphylococcus aureus ATCC 6538	60 (11.6)	>60 (>38.1)	0.75 (0.6)	3 (2.6)
Micrococcus luteus NRRL B-287	30 (5.8)	15 (9.5)	0.1 (0.1)	0.5 (0.4)
Escherichia coli NRRL B-3708	>60 (>11.6)	>60 (>38.1)	>60 (>51)	>60 (>52.4)
Salmonella enterica ATCC 10708	>60 (>11.6)	>60 (>38.1)	>60 (>51)	>60 (>52.4)
Saccharomyces cerevisiae ATCC 204508	>60 (>11.6)	>60 (>38.1)	0.5 (0.4)	2 (1.7)

[a)] Values are based on three independent replicates. Kanamycin and ampicillin (*S. aureus*, *M. luteus*, *S. enterica* and *E. coli*) and amphotericin B (*S. cerevisiae*), were used as positive controls. Compounds **2**, **4-8**, **11**, **12a-b** and **14-18** were inactive against all tested microorganisms up to 60 µM concentration.

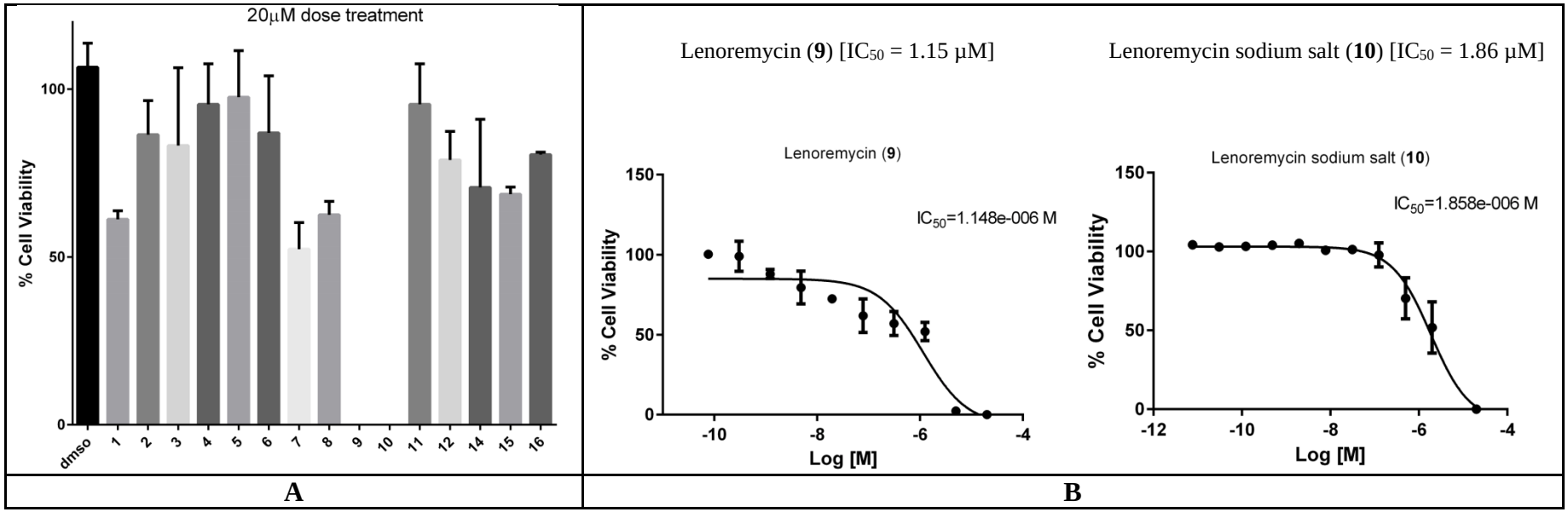
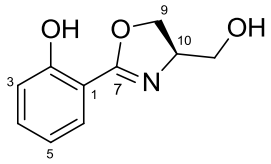


Figure S10. A) Viability of A549 (lung) human cancer cell line at 20 µM treatment for compounds **1-12,** and **14-16** after 48h. B) Dose–response curve of lenoremycin (**9**) and lenoremycin sodium salt (**10**) in A549 (lung) human cancer cell line at 48h (actinomycin D was used as a positive control).



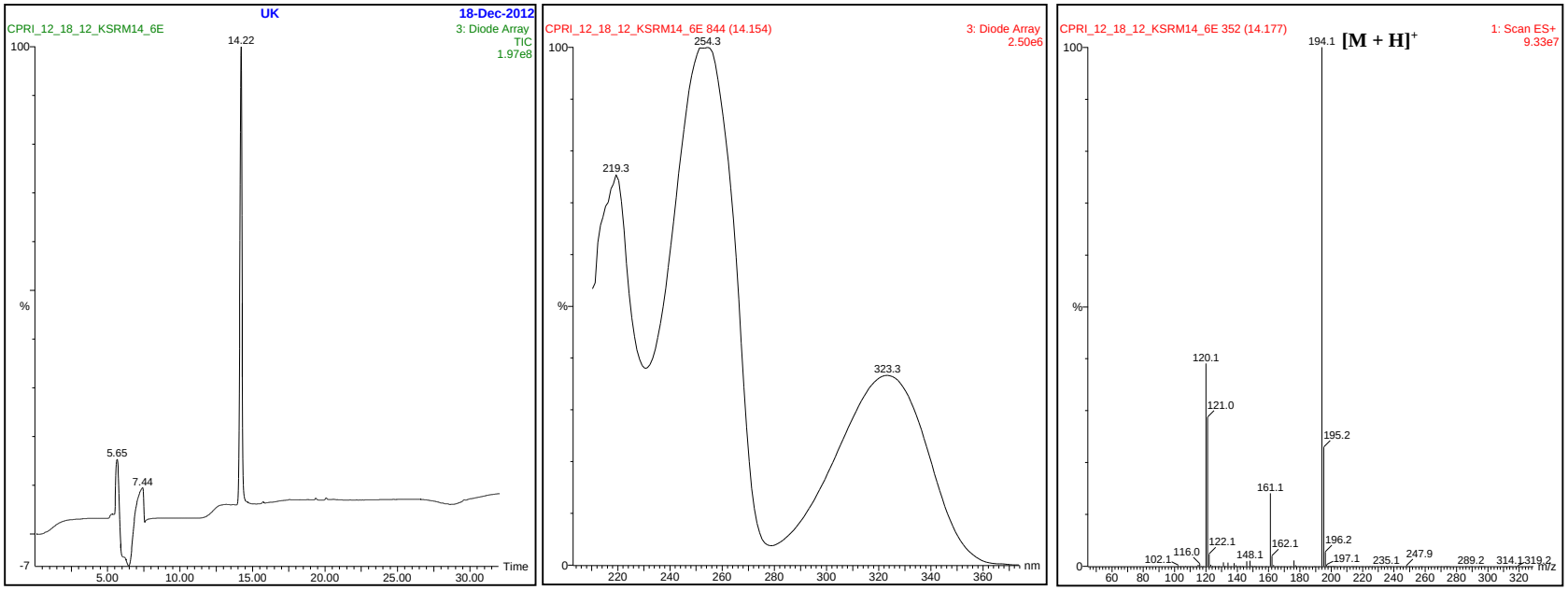
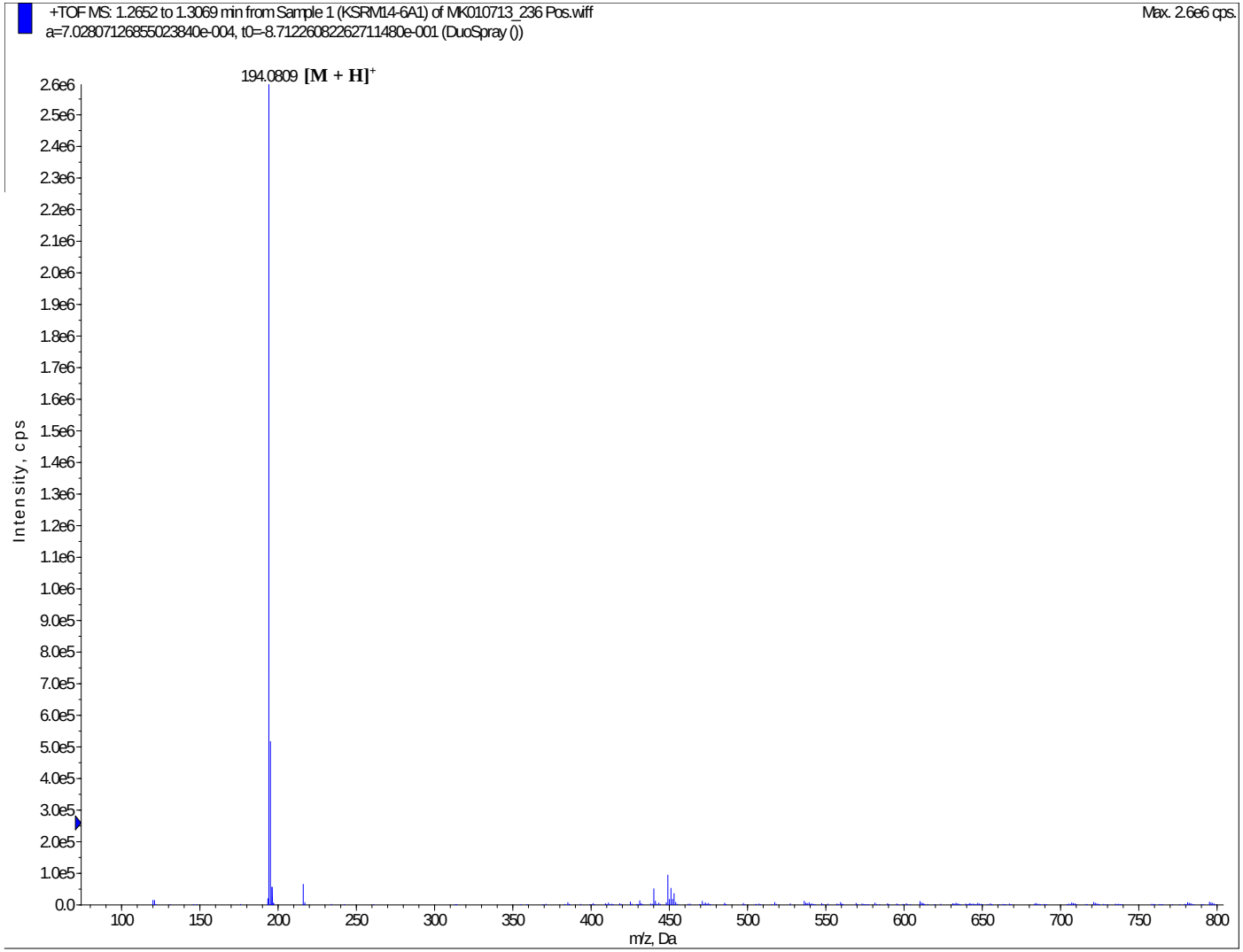


Figure S11. HPLC/UV/MS analyses of the purified spoxazomicin C (**1**). Detection wavelength: 210-550 nm; **solvent A:** H_2O/0.1% Formic acid; **solvent B:** CH_3CN/0.1% Formic acid; flow rate: 0.5 mL min^{-1}; 0-4 min, 10% B; 4-22 min, 10-100% B; 22-27 min, 100% B; 27-29 min, 100%-10% B; 29-35 min, 10 % B.



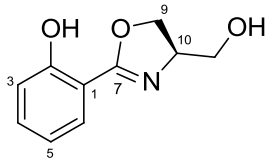


Figure S12. (+)-HRESI-MS spectrum of spoxazomicin C (**1**)

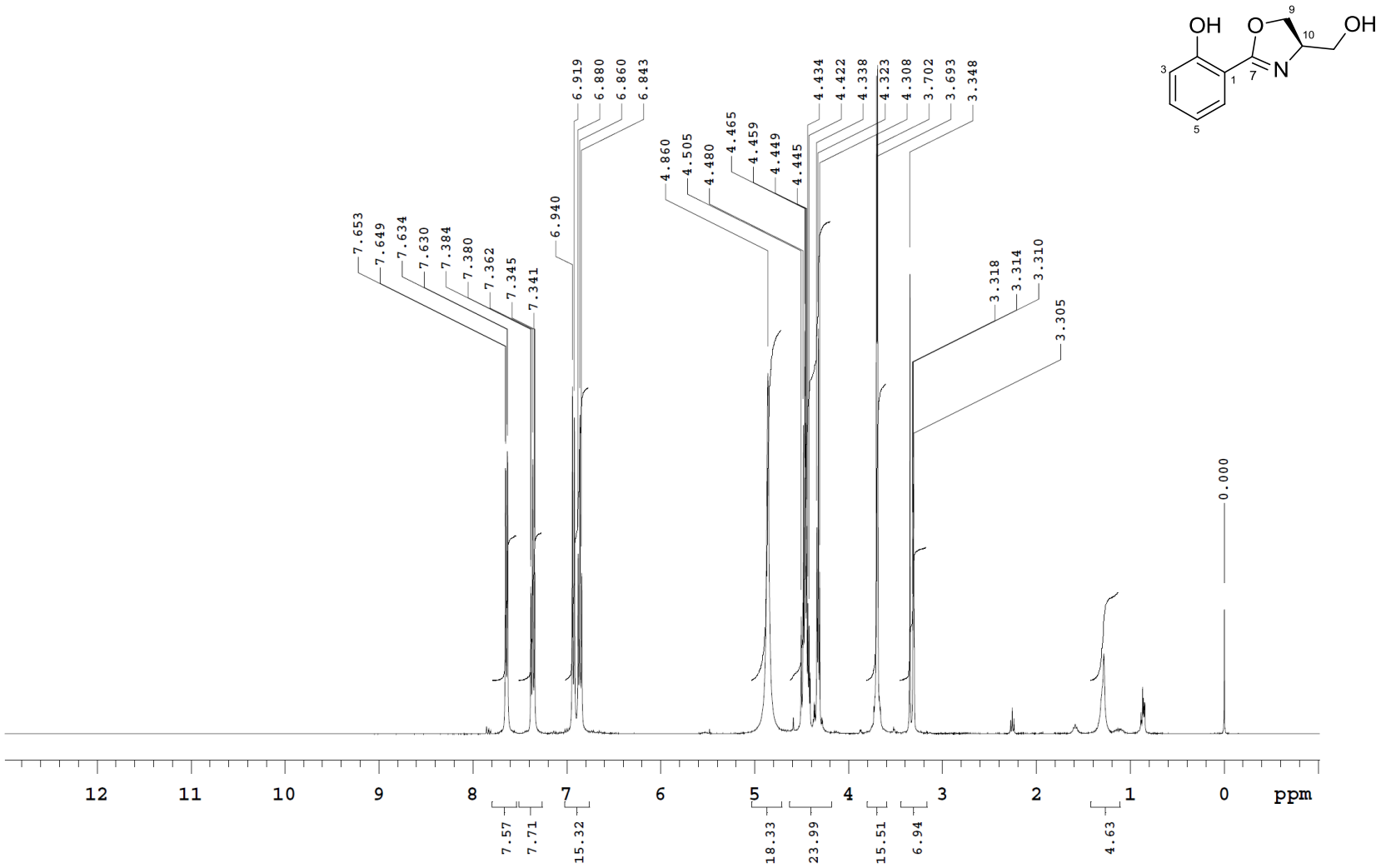


Figure S13. ^{1}H NMR spectrum (CD_3OD, 400 MHz) of spoxazomicin C (**1**)

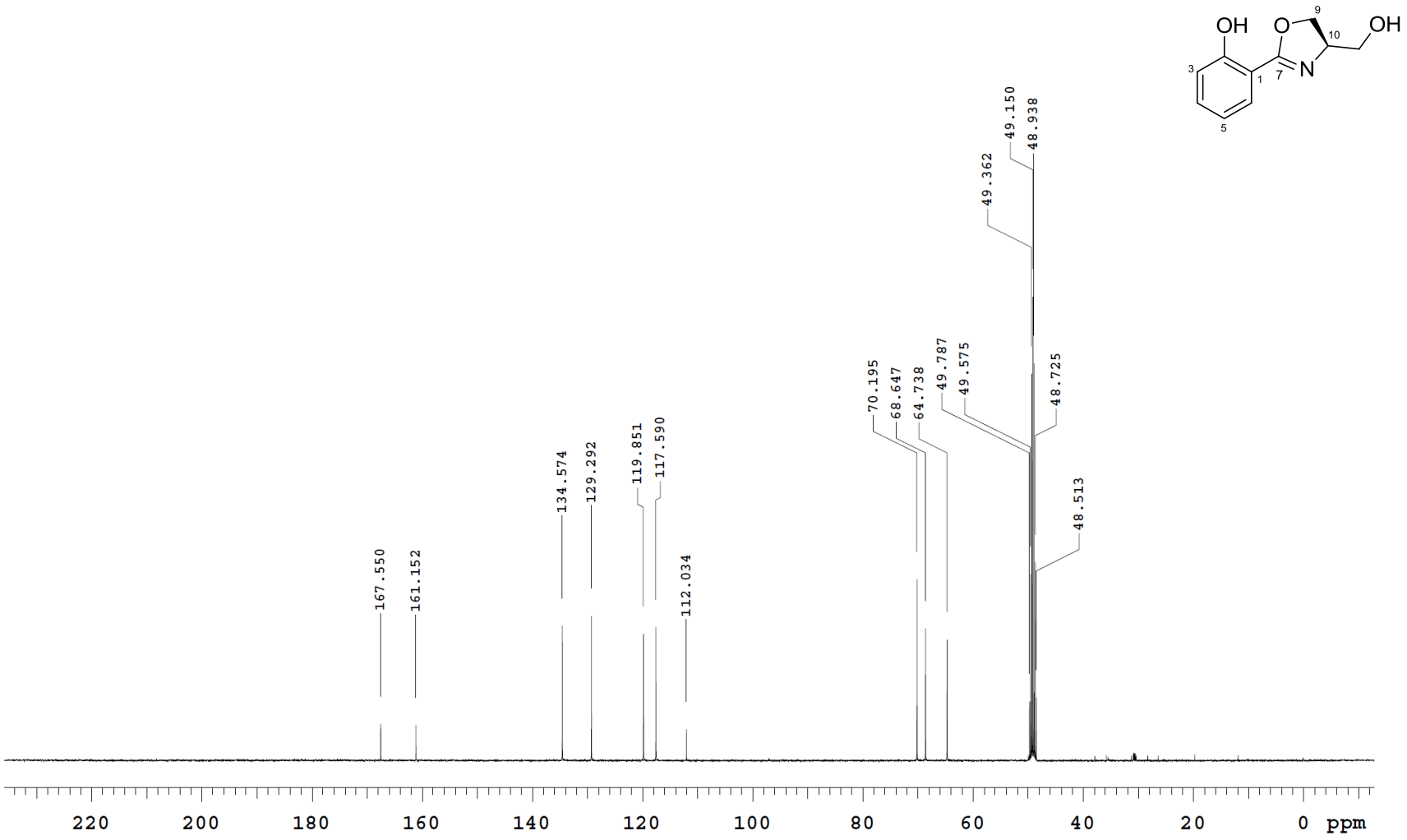


Figure S14. ^{13}C NMR spectrum (CD_3OD, 100 MHz) of spoxazomicin C (**1**)

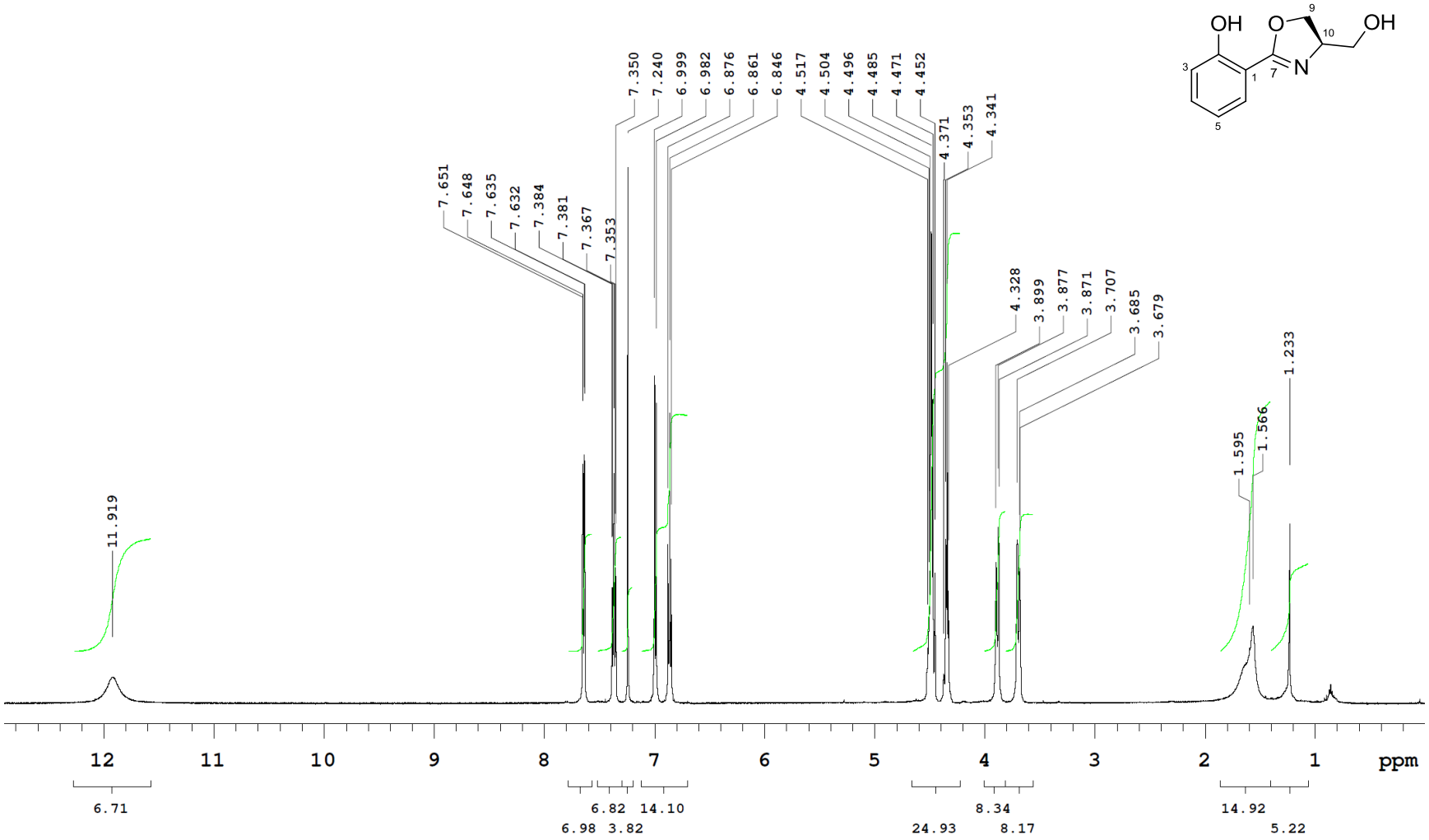


Figure S15. ^{1}H NMR spectrum ($CDCl_3$, 500 MHz) of spoxazomicin C (**1**)

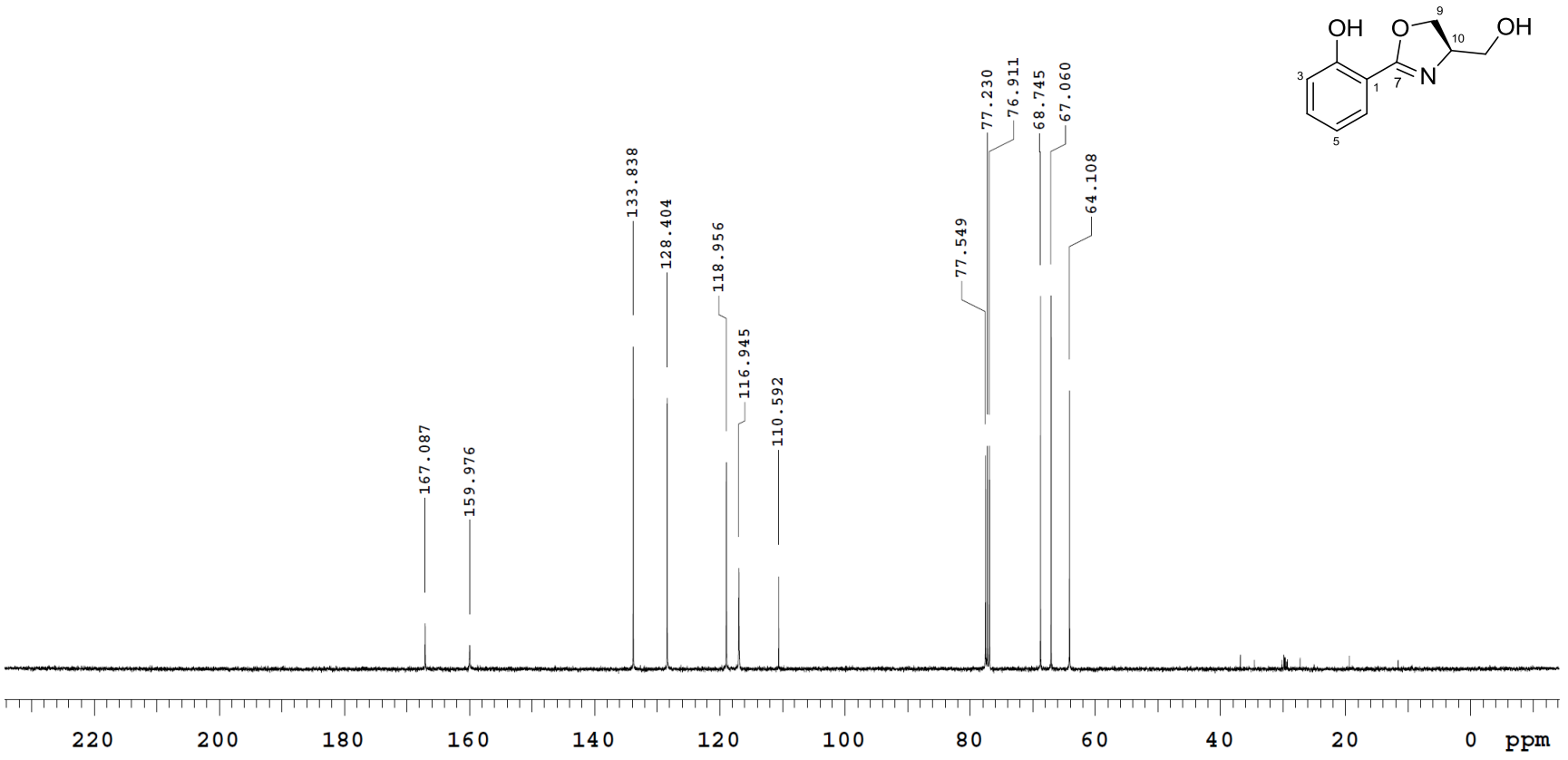


Figure S16. ^{13}C NMR spectrum ($CDCl_3$, 100 MHz) of spoxazomicin C (**1**)

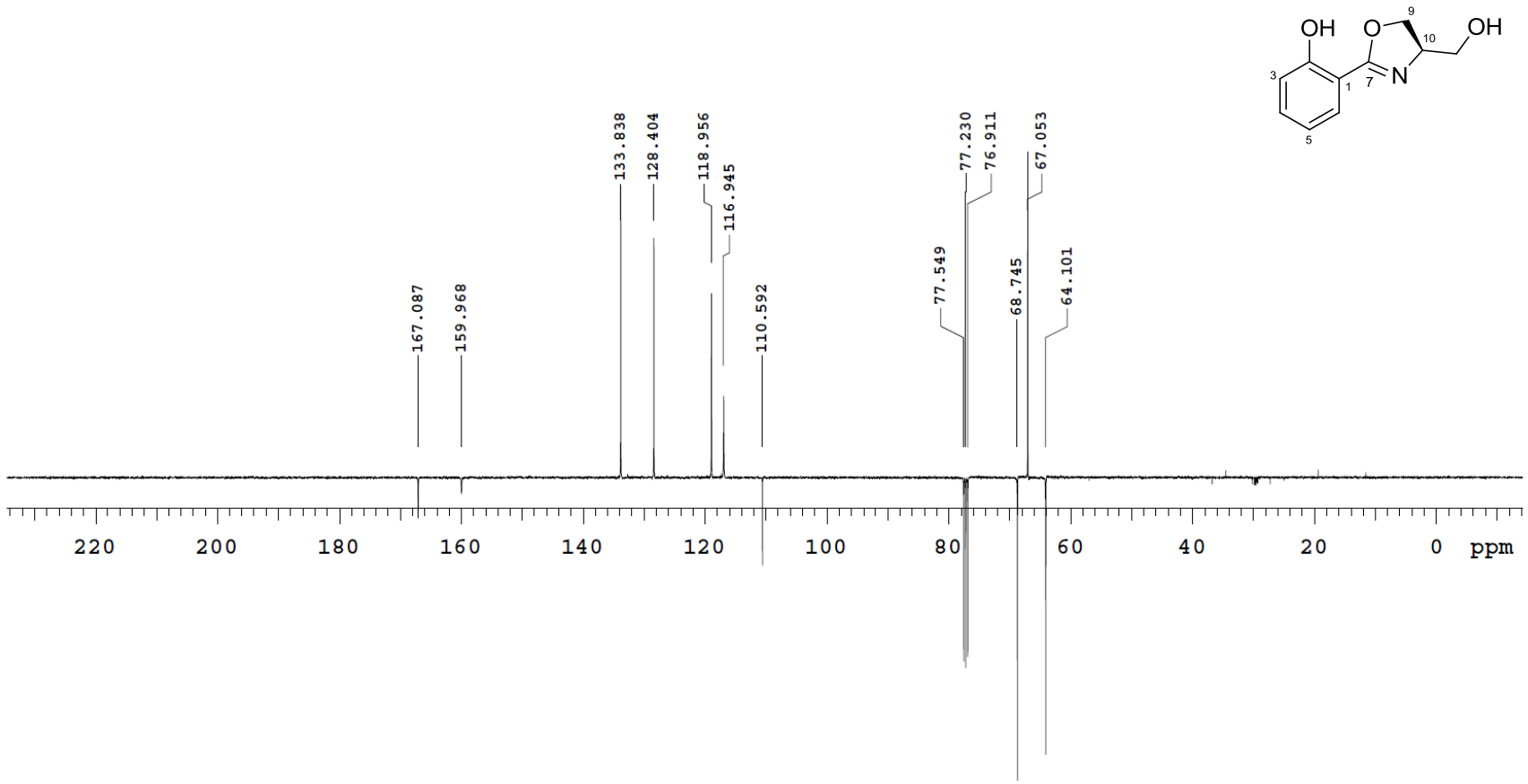


Figure S17. APT NMR spectrum ($CDCl_3$, 100 MHz) of spoxazomicin C (**1**)

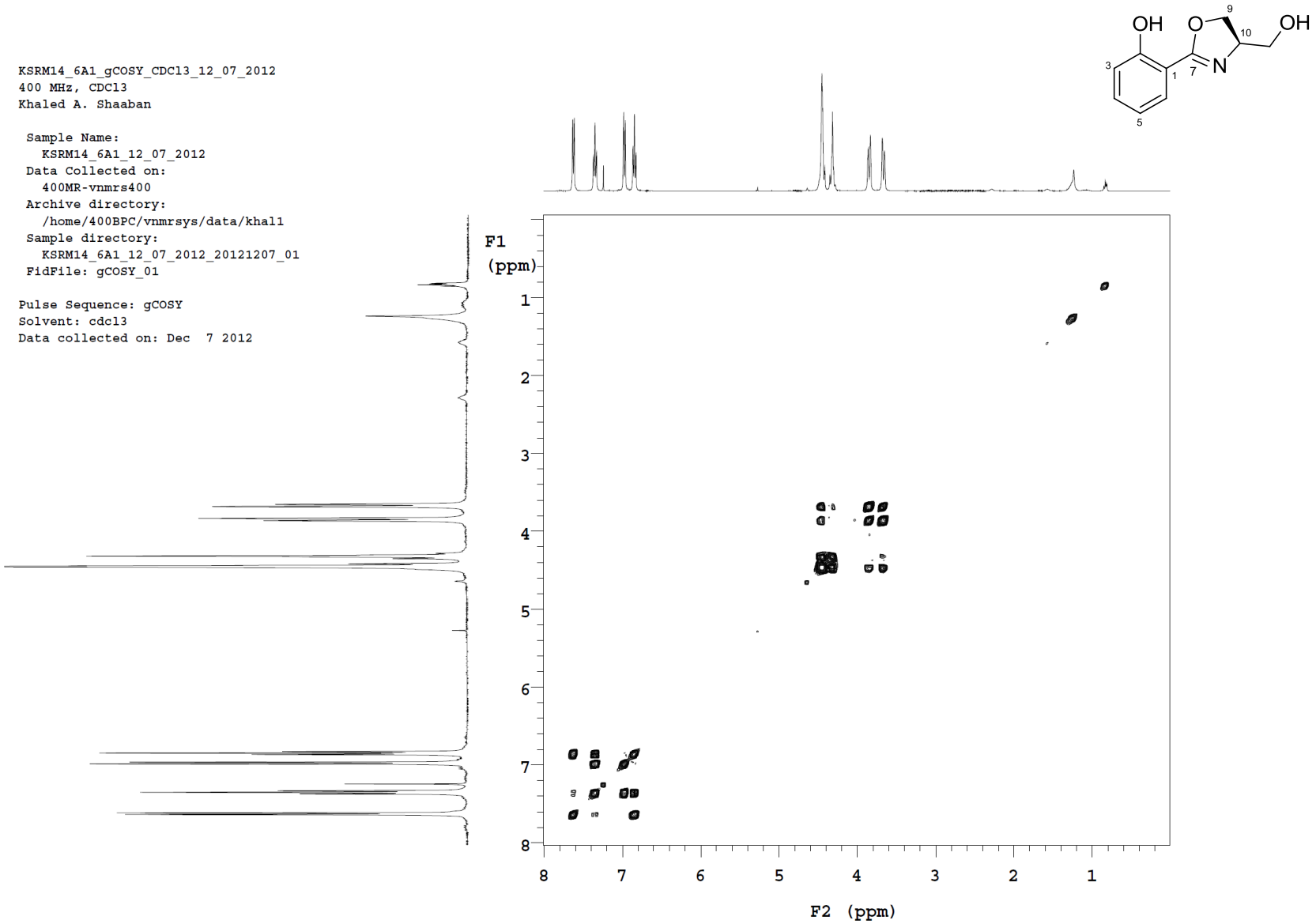


Figure S18. $^{1}H,^{1}H$-COSY spectrum ($CDCl_3$, 400 MHz) of spoxazomicin C (**1**)

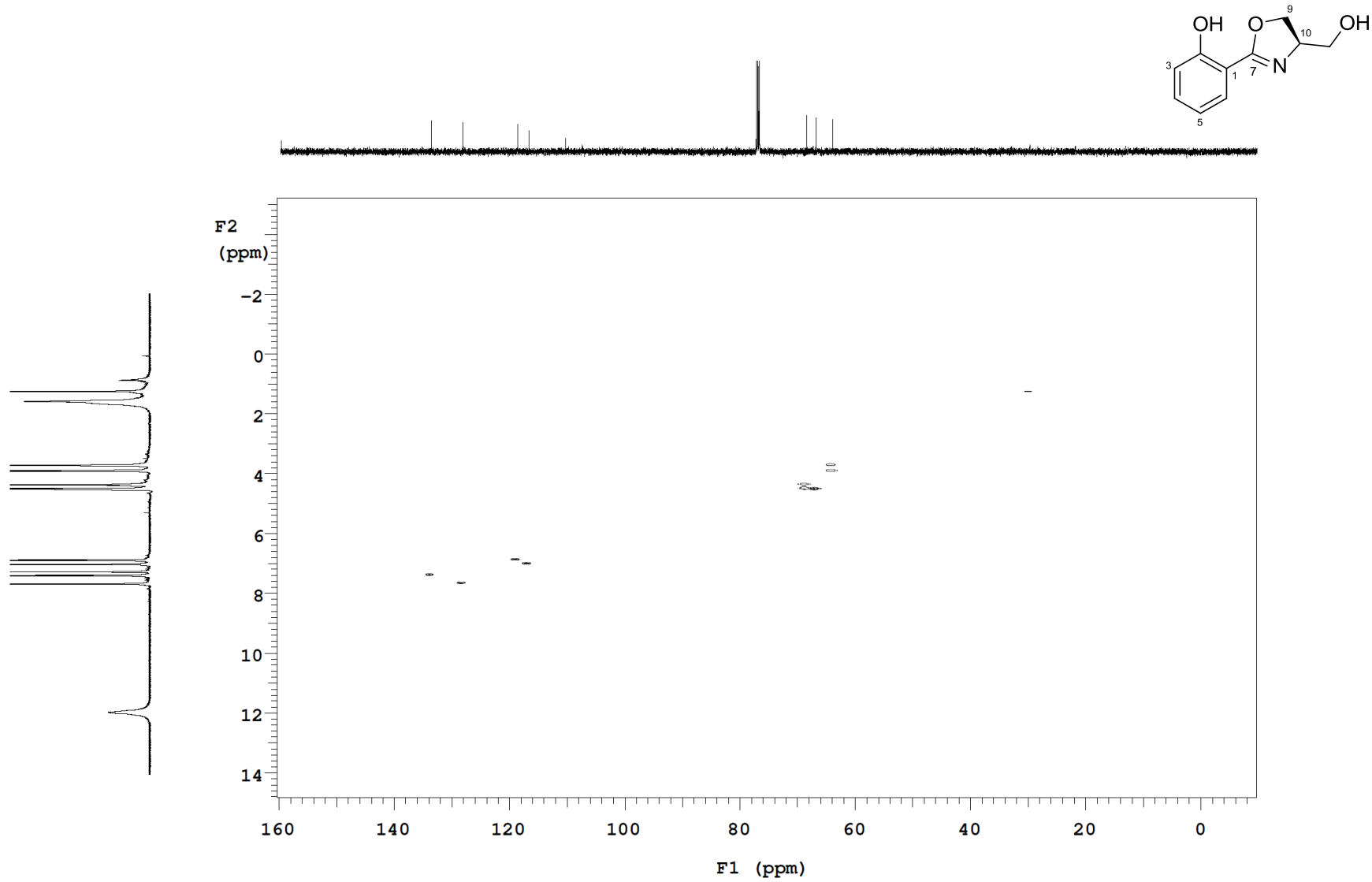


Figure S19. HSQC spectrum ($CDCl_3$, 500 MHz) of spoxazomicin C (**1**)

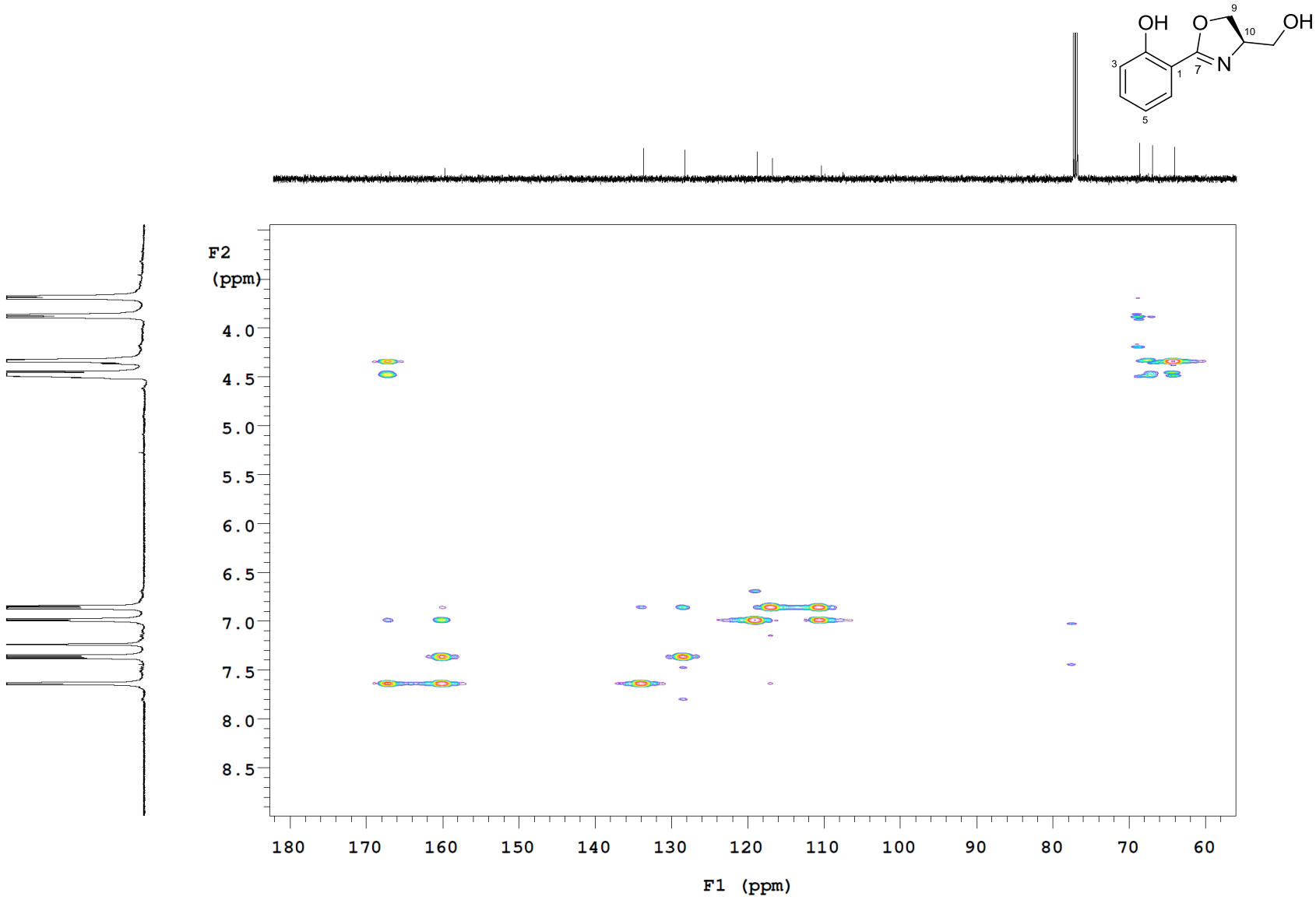


Figure S20. HMBC spectrum ($CDCl_3$, 500 MHz) of spoxazomicin C (**1**)

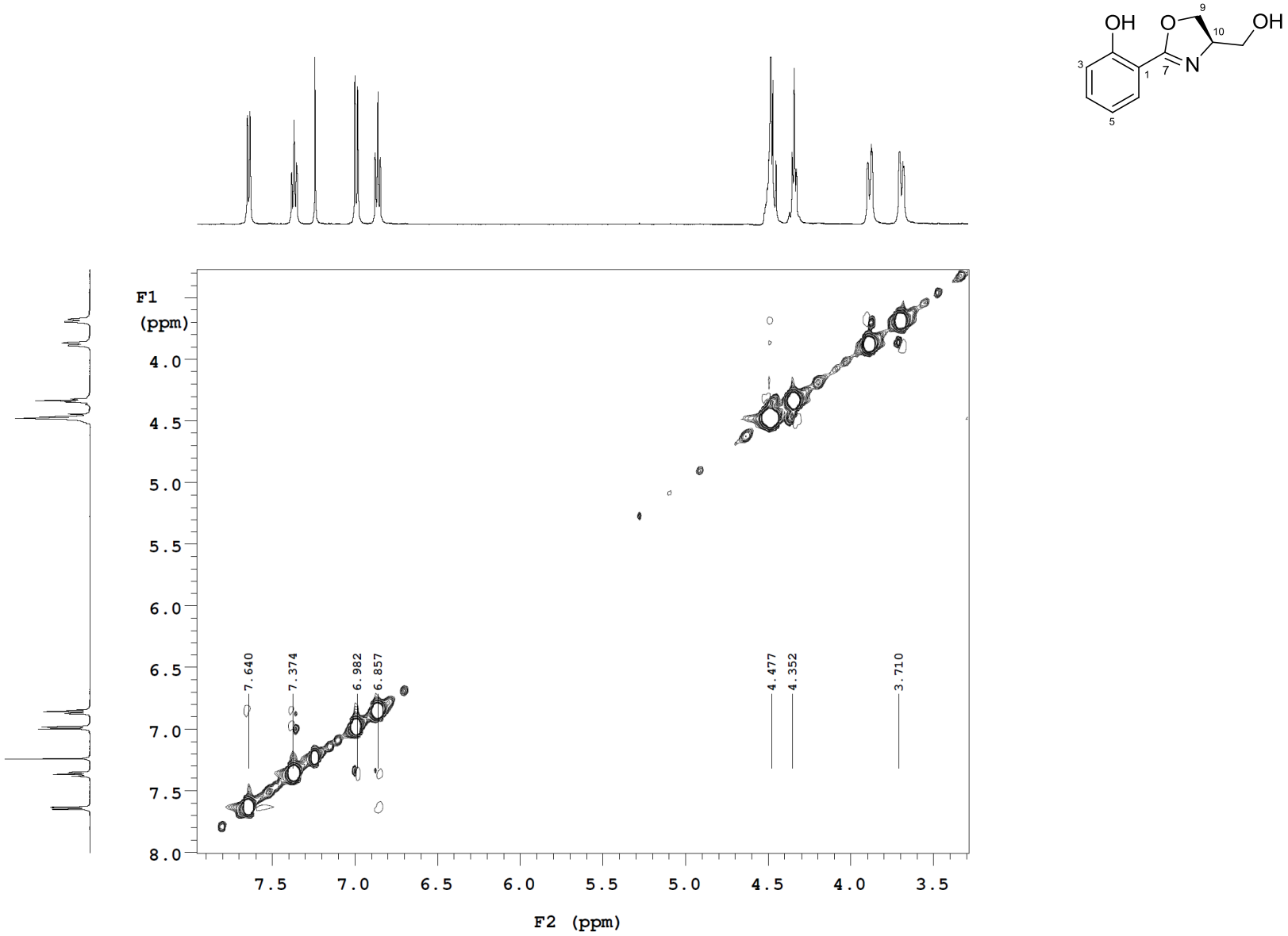


Figure S21. NOESY spectrum ($CDCl_3$, 500 MHz) of spoxazomicin C (**1**)

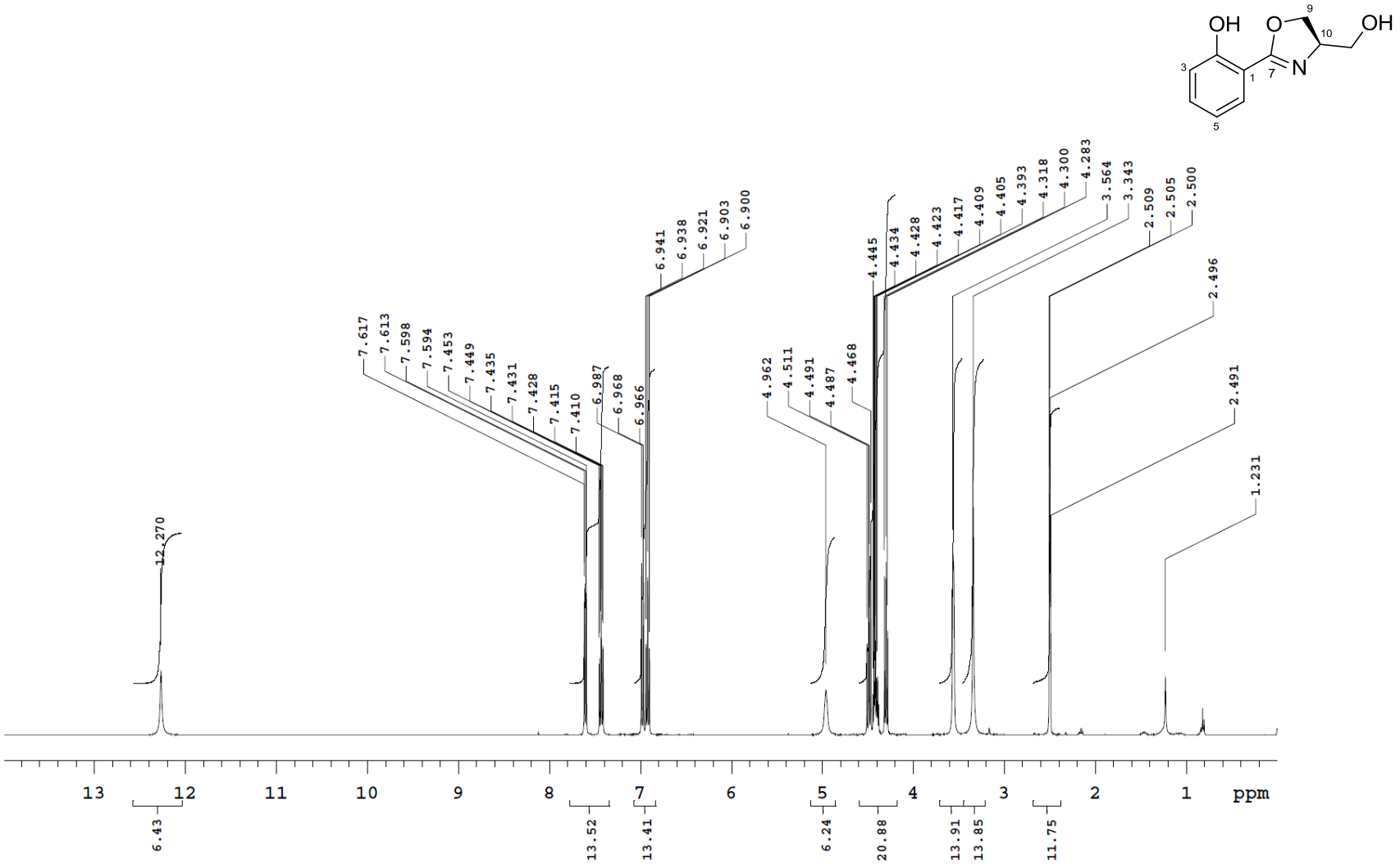


Figure S22. ^{1}H NMR spectrum (DMSO-d_6, 400 MHz) of spoxazomicin C (**1**)

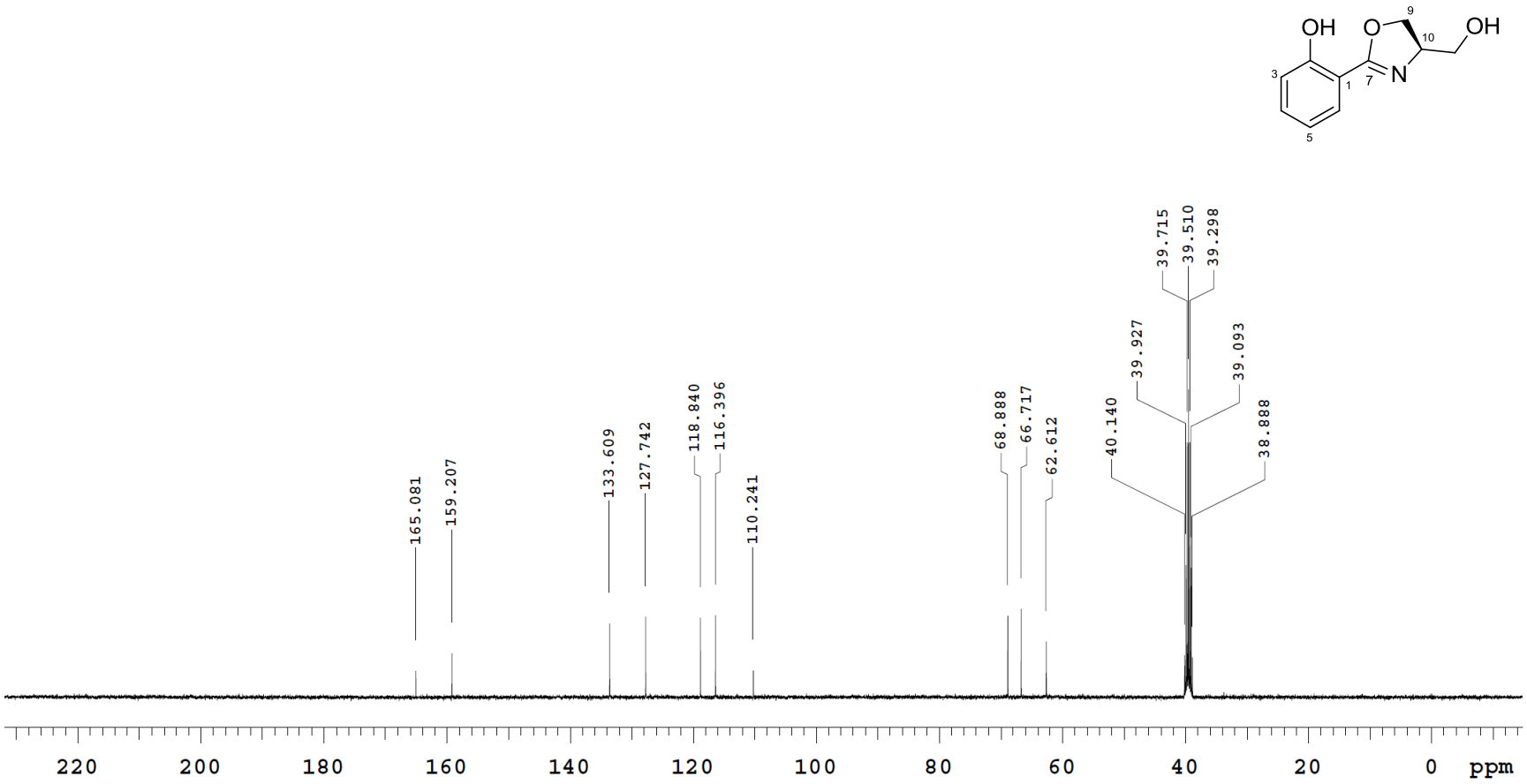


Figure S23. ^{13}C NMR spectrum (DMSO-d_6, 100 MHz) of spoxazomicin C (**1**)

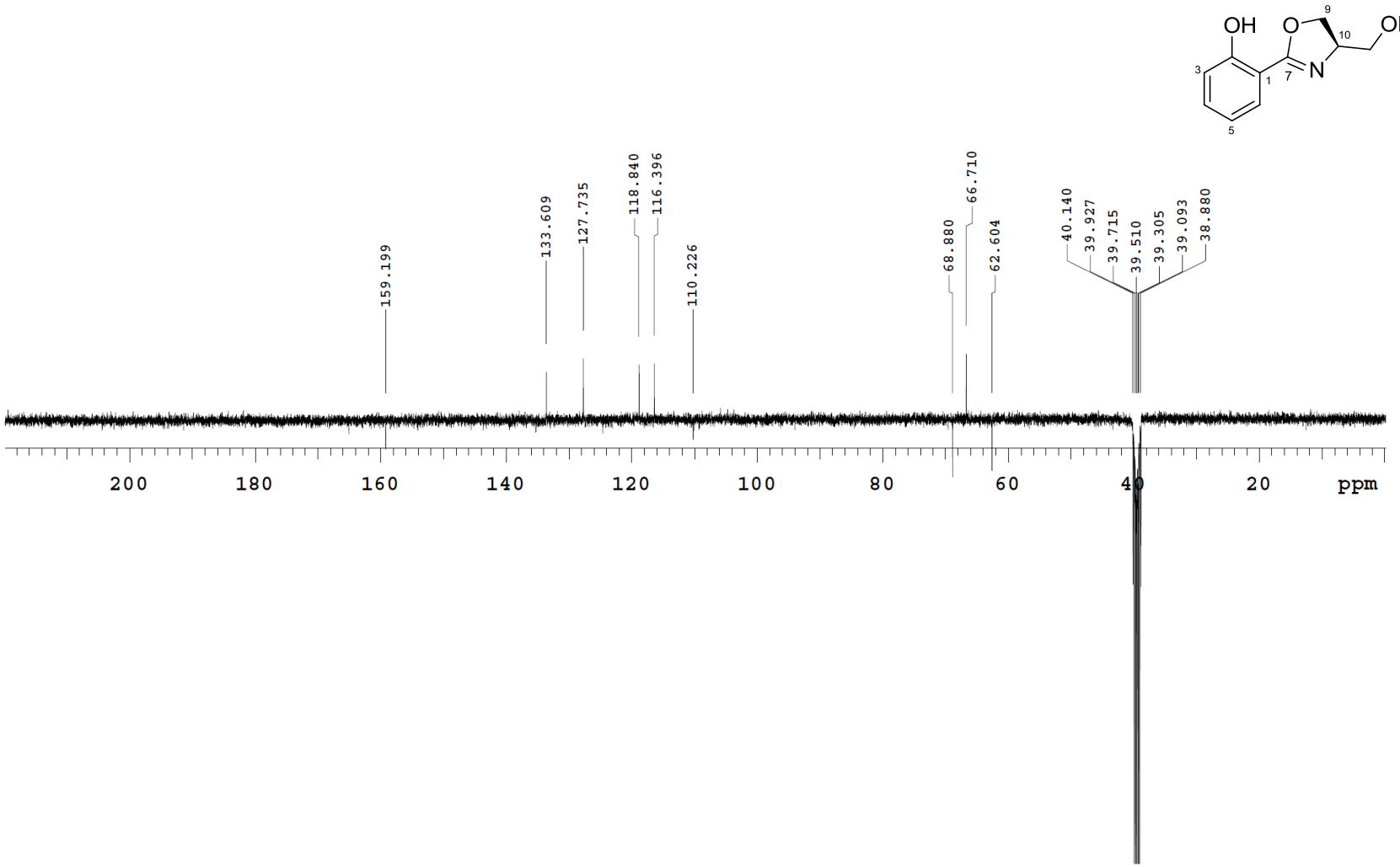


Figure S24. APT NMR spectrum (DMSO-d_6, 100 MHz) of spoxazomicin C (**1**)

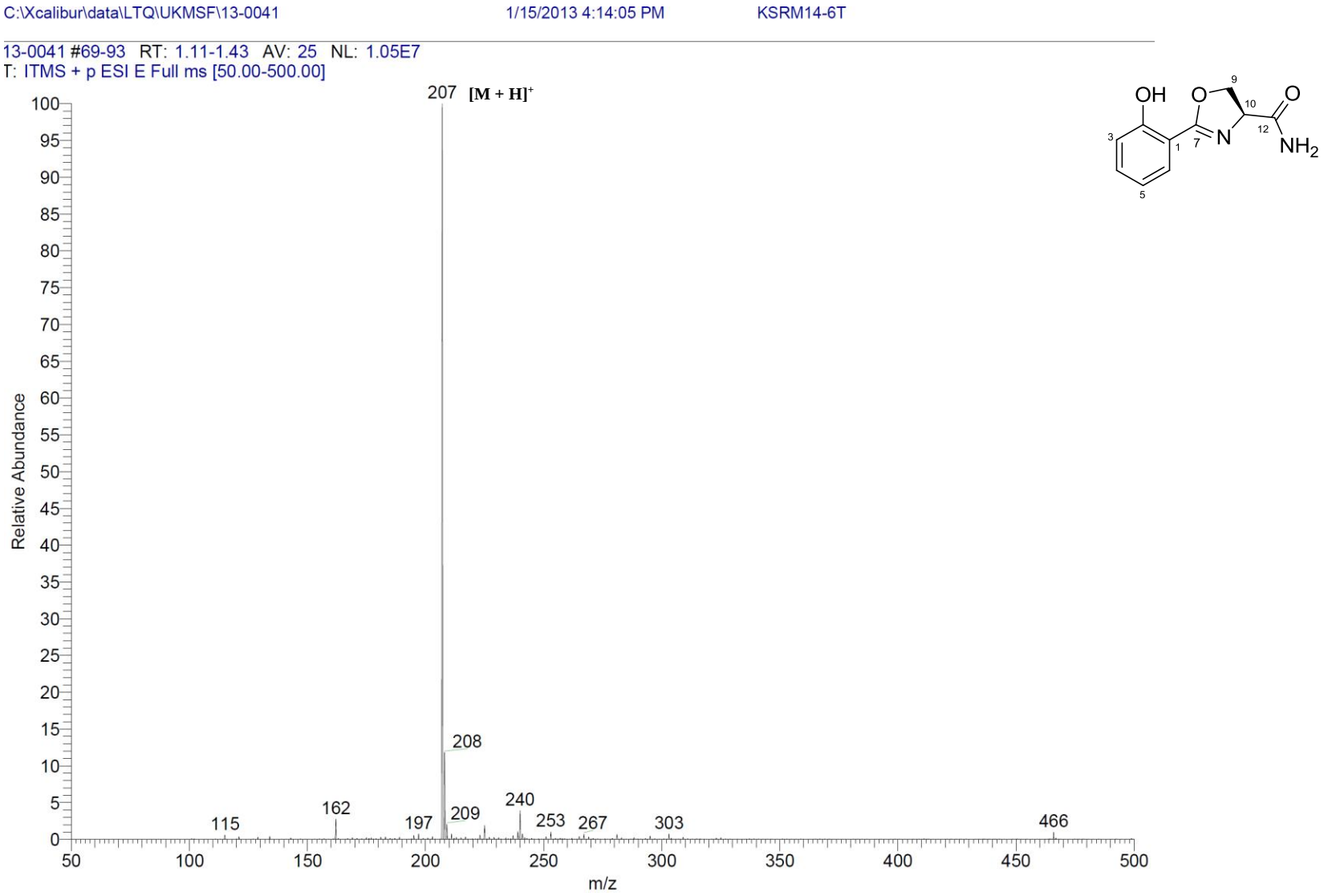


Figure S25. (+)-ESI-MS spectrum of spoxazomicin D (**2**)

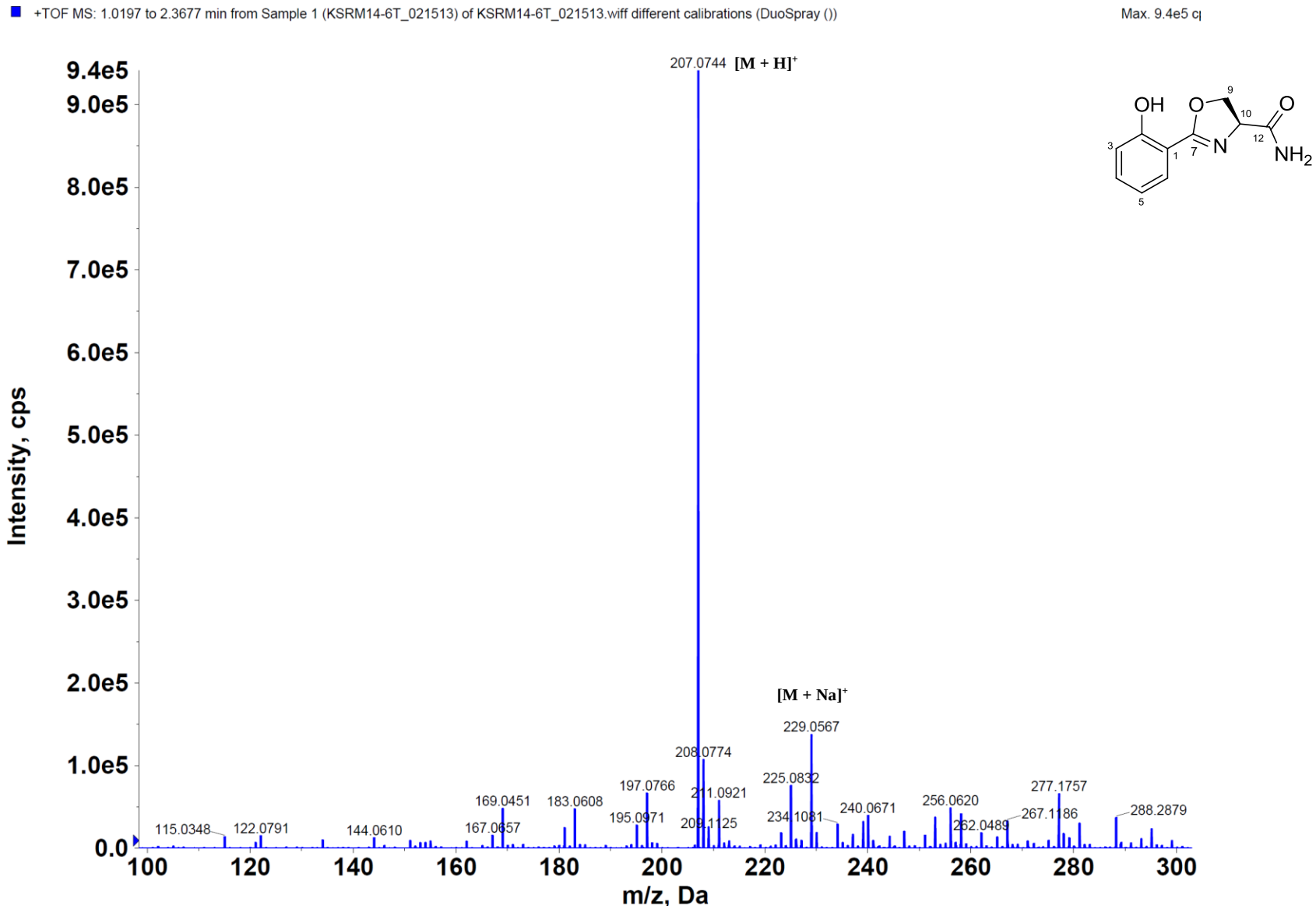


Figure S26. (+)-HRESI-MS spectrum of spoxazomicin D (**2**)

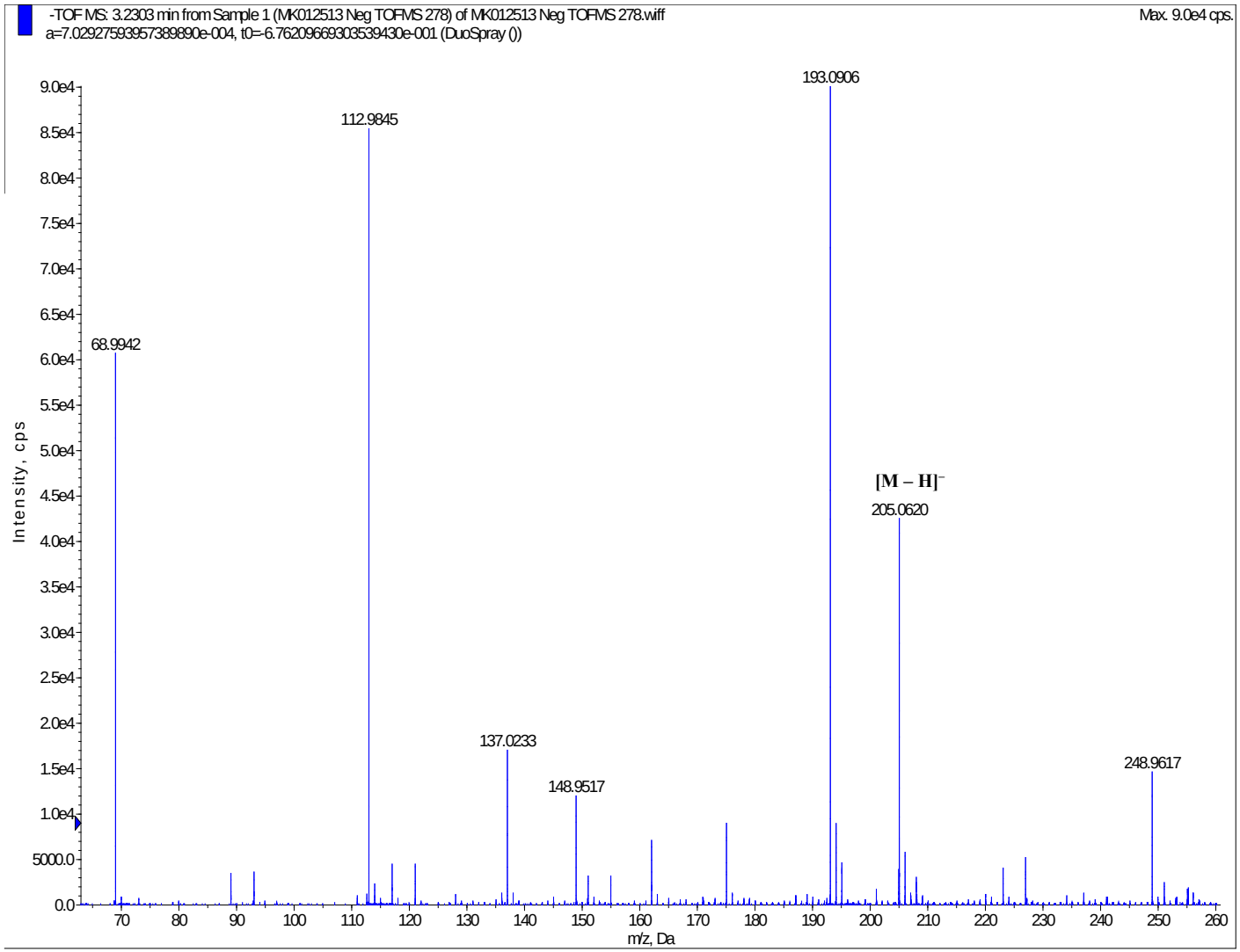


Figure S27. (–)-HRESI-MS spectrum of spoxazomicin D (**2**)

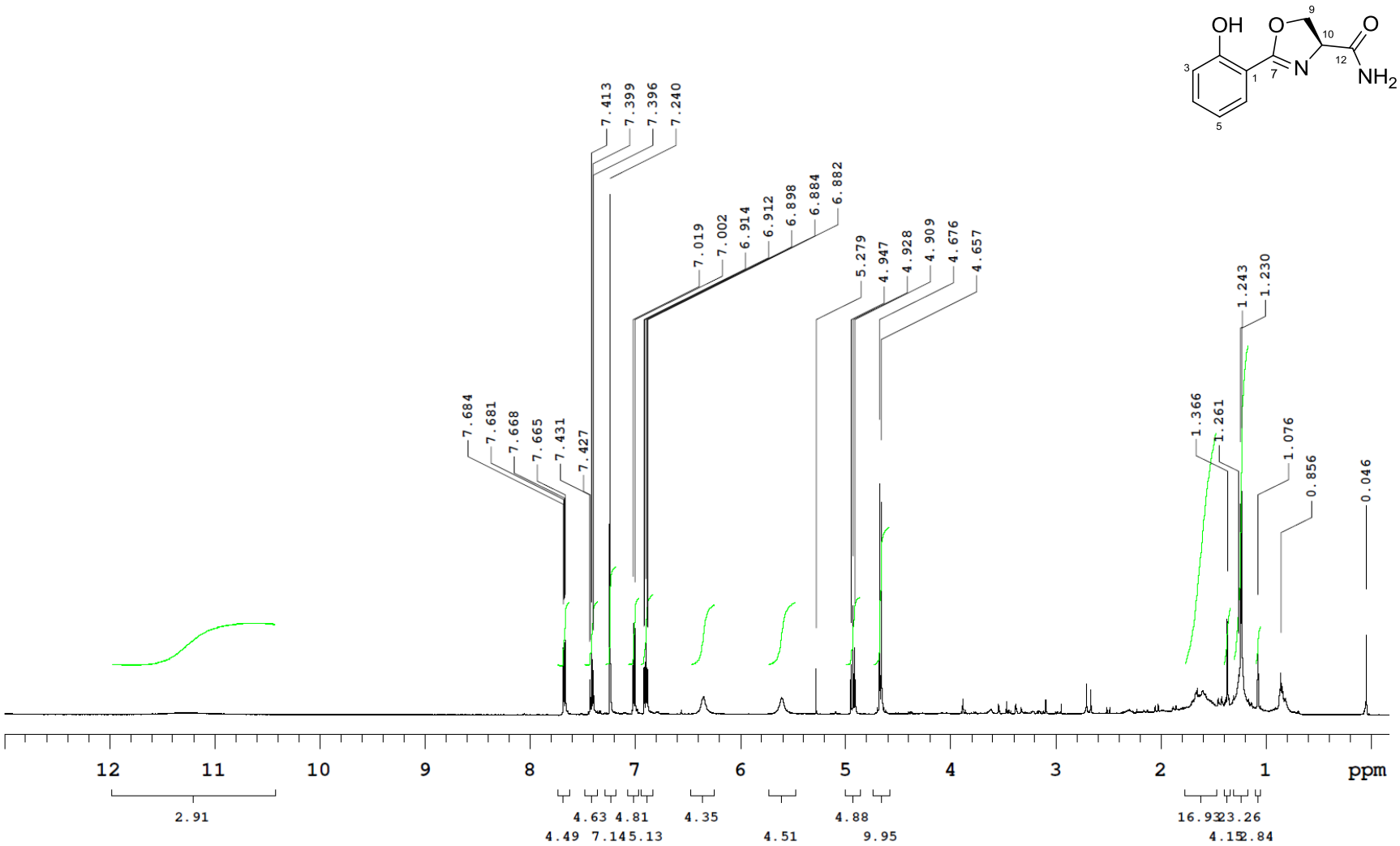


Figure S28. ^{1}H NMR spectrum ($CDCl_3$, 500 MHz) of spoxazomicin D (**2**)

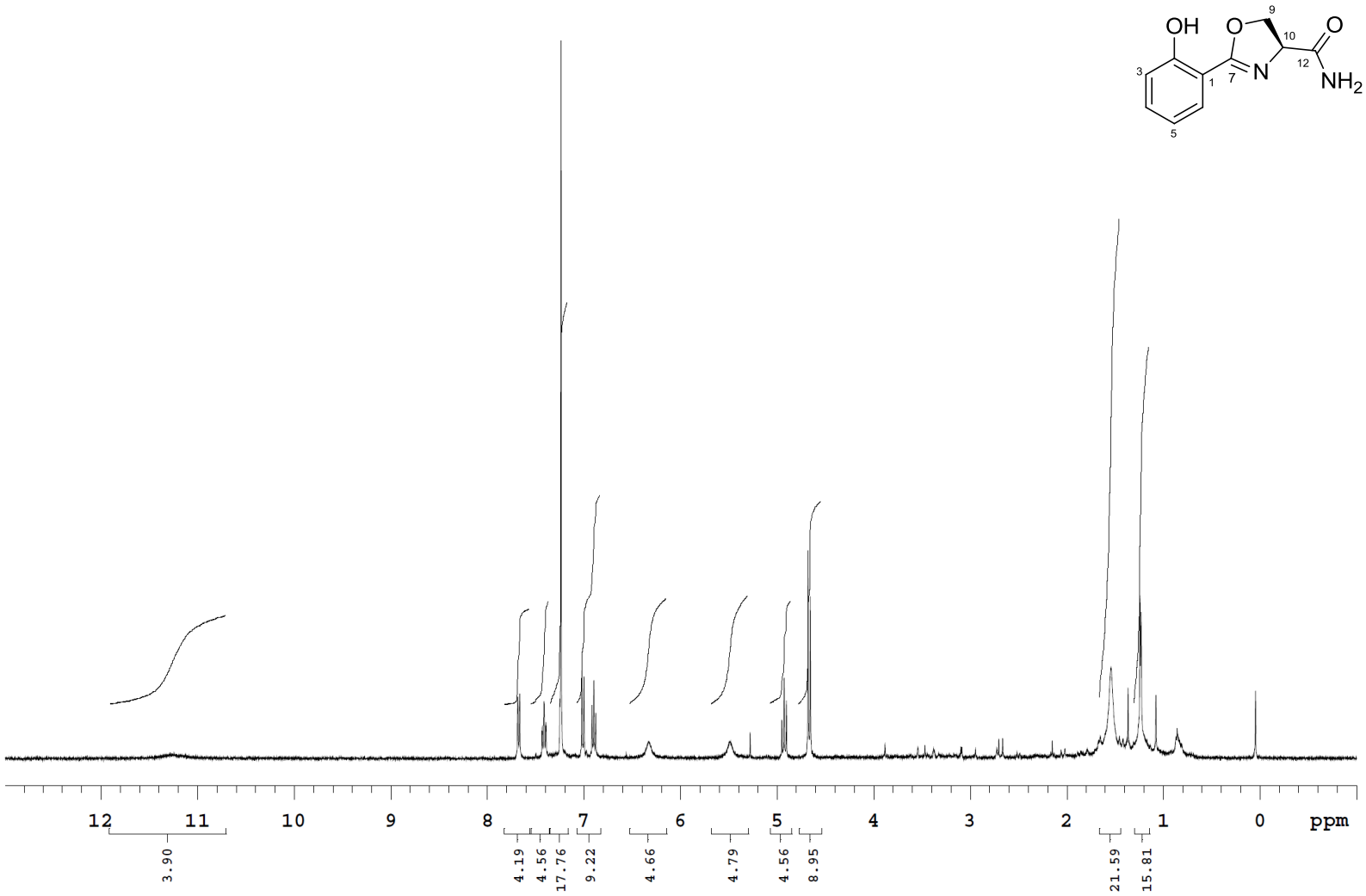


Figure S29. ^{1}H NMR spectrum ($CDCl_3$, 400 MHz) of spoxazomicin D (**2**)

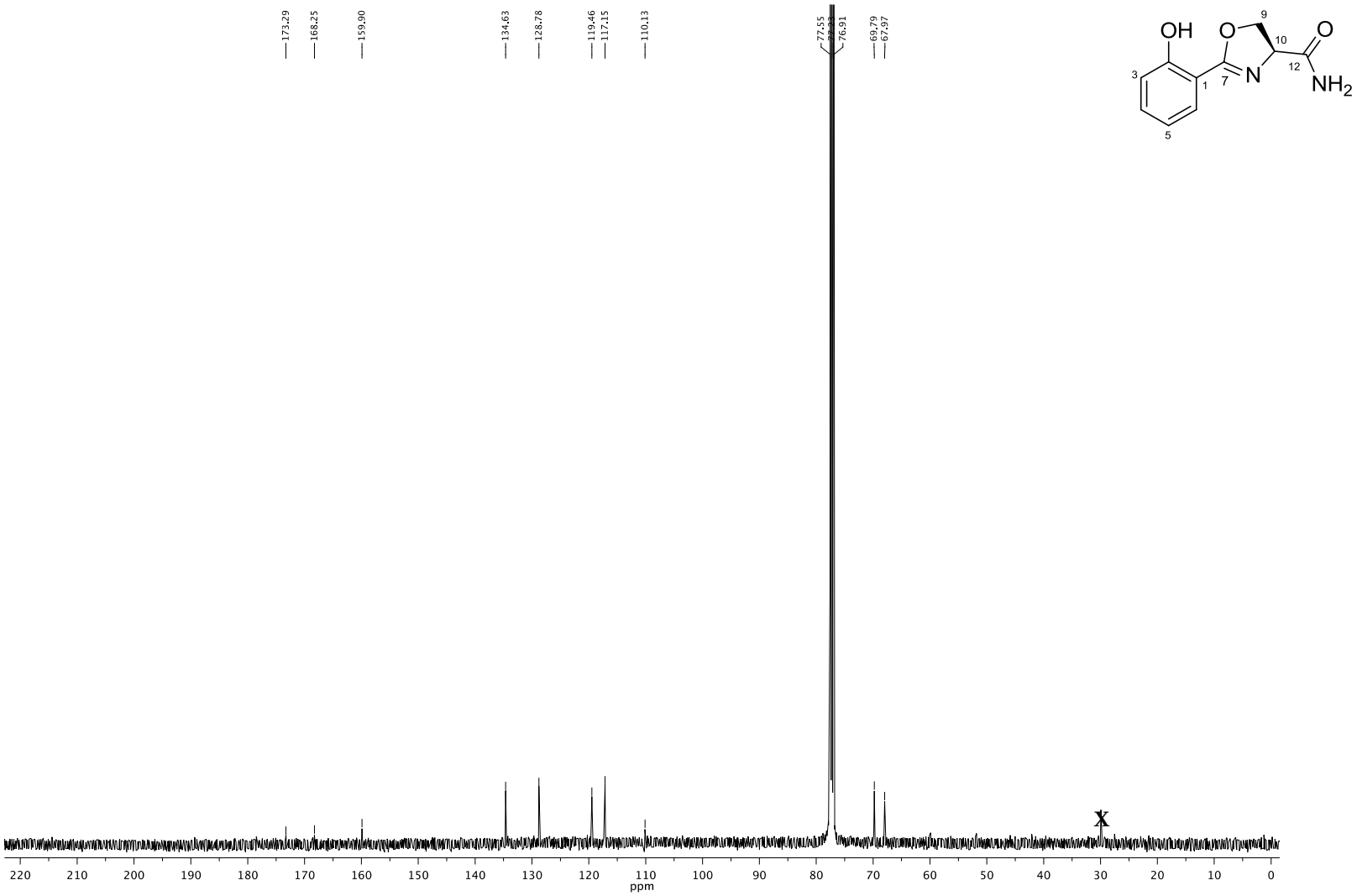


Figure S30. ^{13}C NMR spectrum ($CDCl_3$, 100 MHz) of spoxazomicin D (**2**)

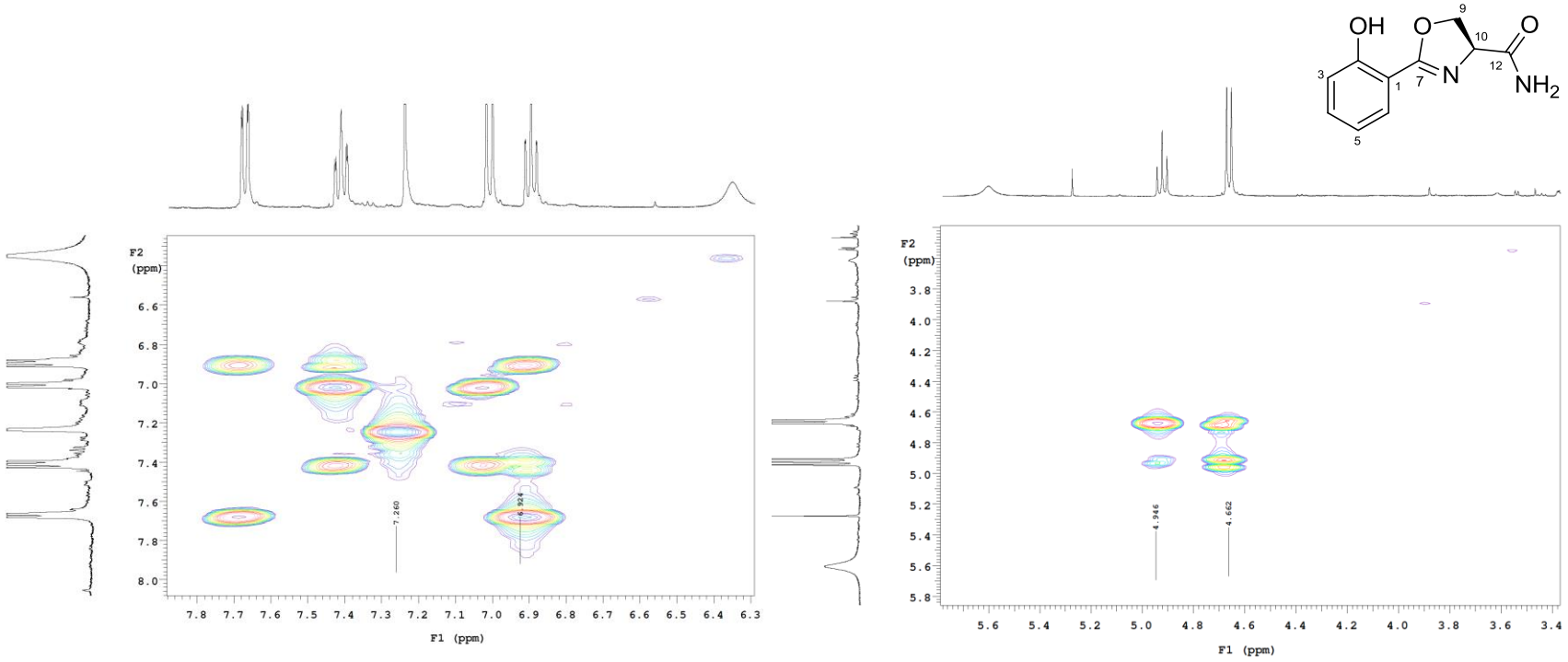


Figure S31. Enlarged $^{1}H,^{1}H$-COSY spectrum ($CDCl_3$, 400 MHz) of spoxazomicin D (**2**)

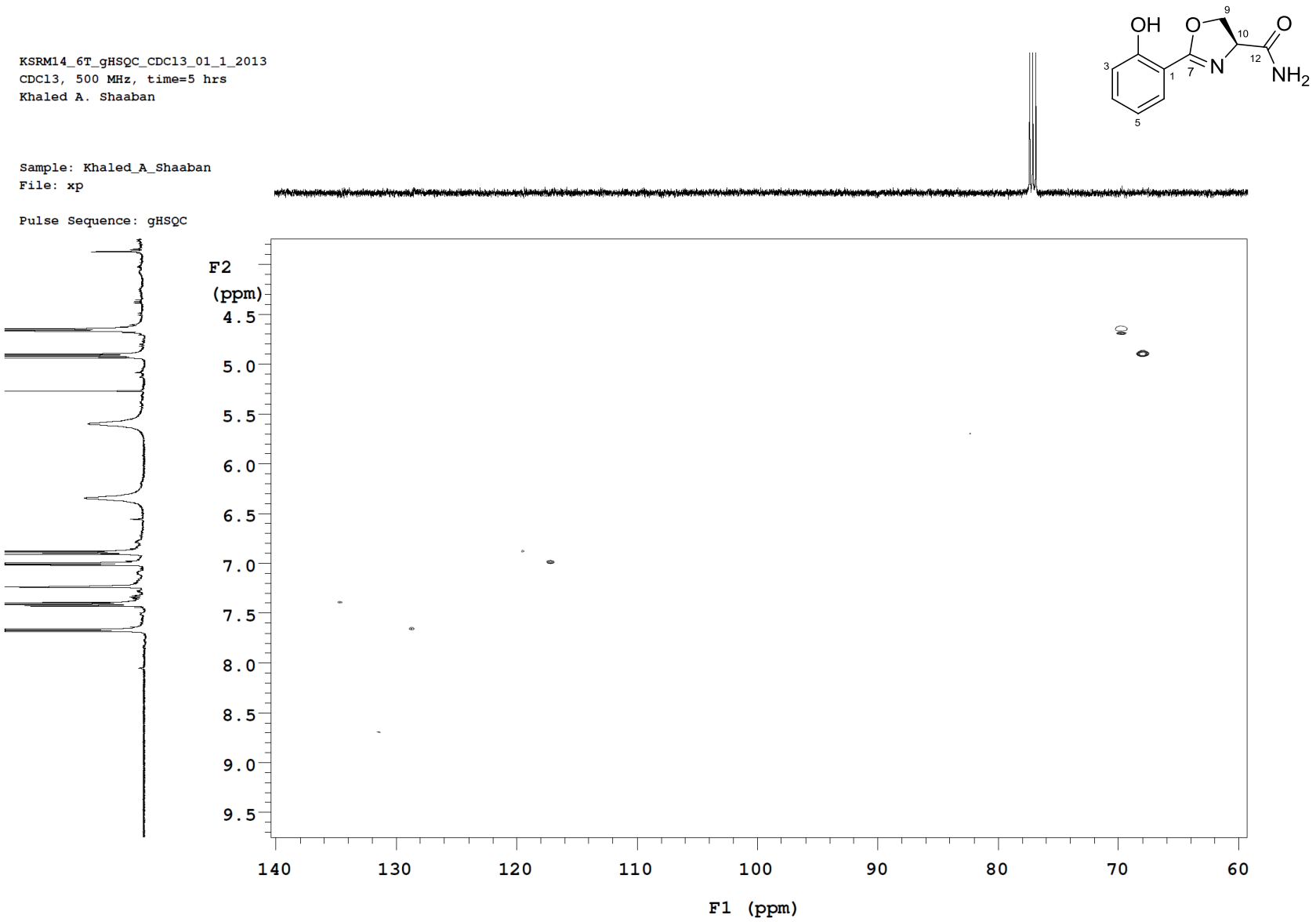


Figure S32. HSQC spectrum ($CDCl_3$, 500 MHz) of spoxazomicin D (**2**)

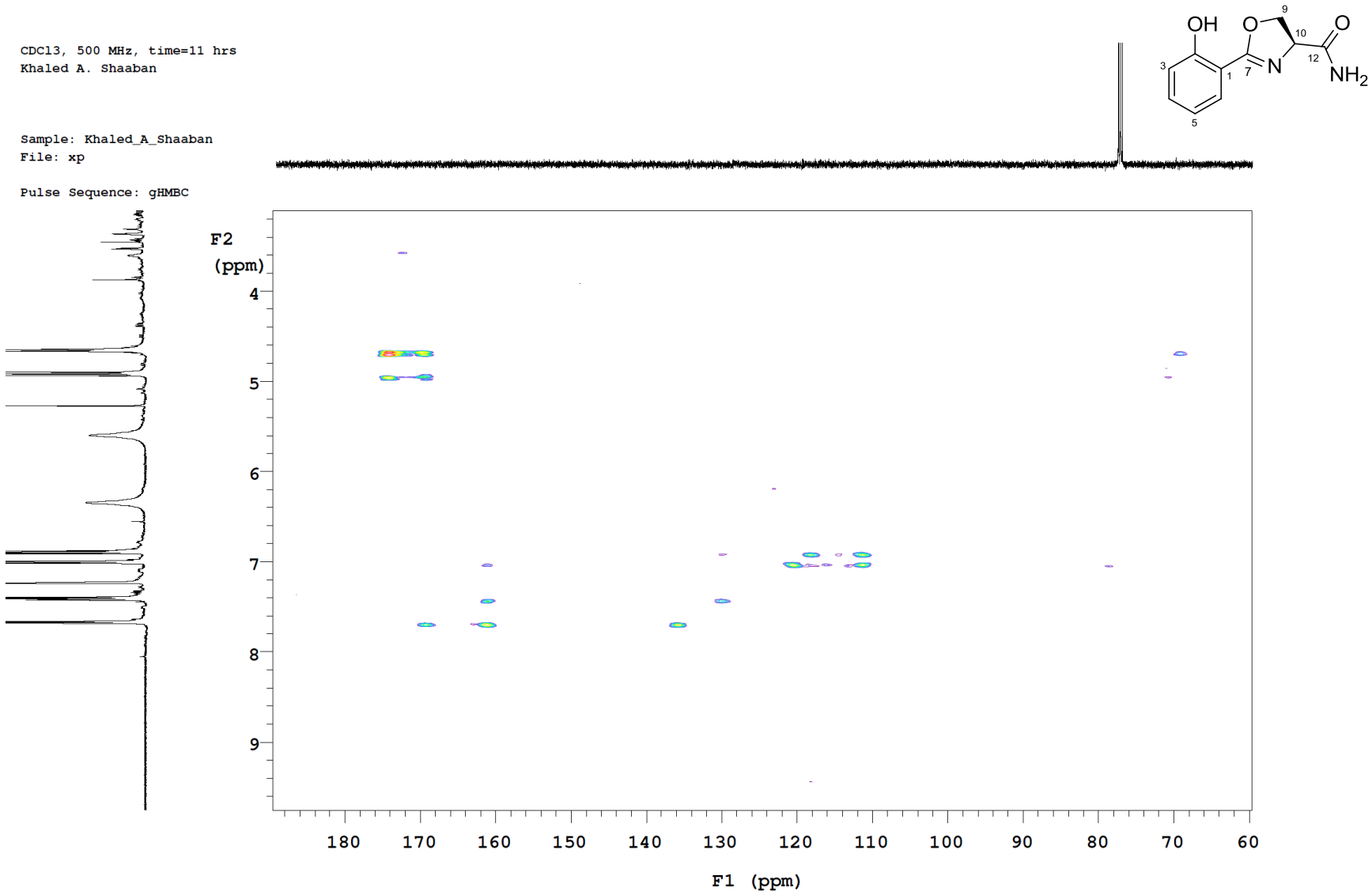


Figure S33. HMBC spectrum ($CDCl_3$, 500 MHz) of spoxazomicin D (**2**)

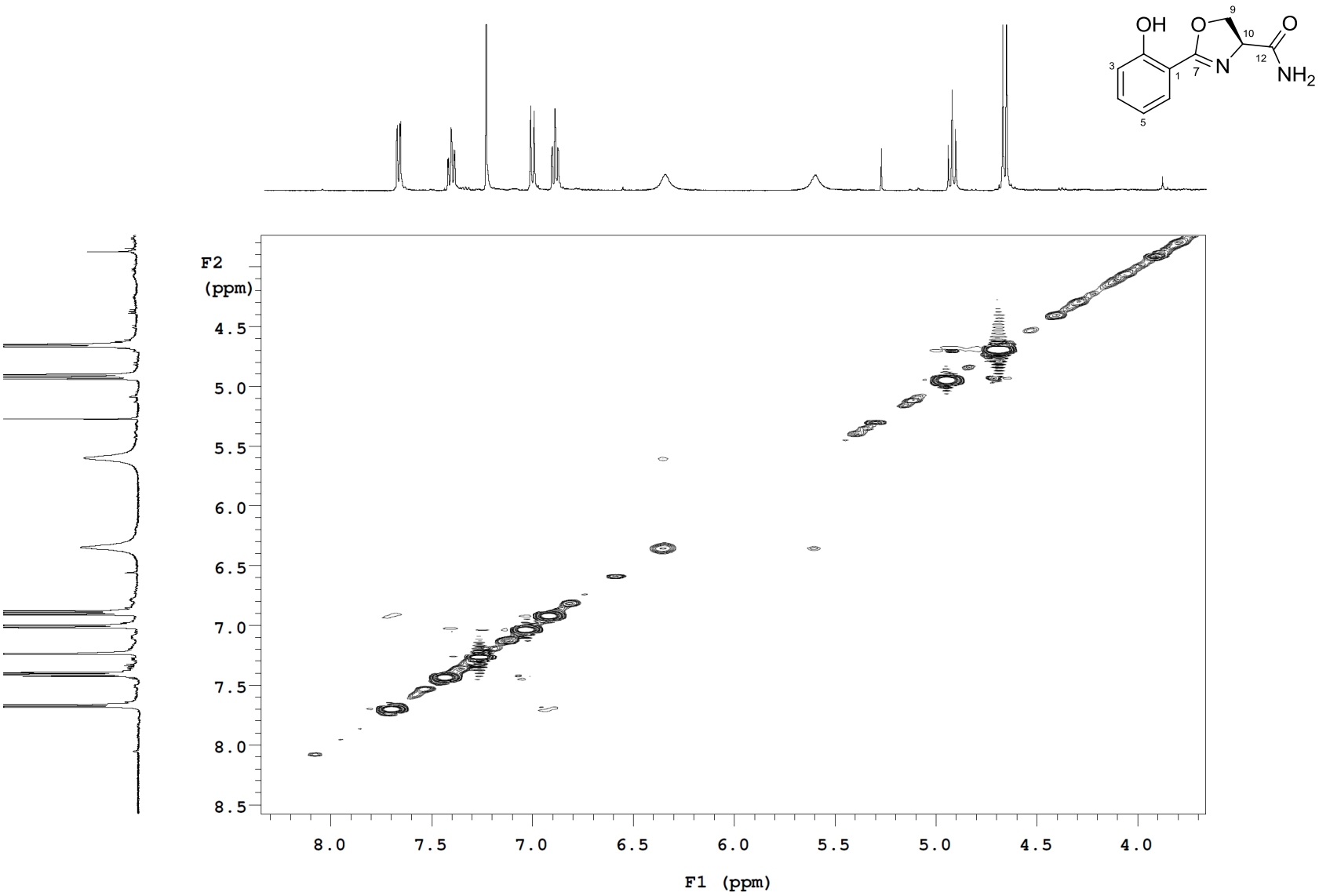


Figure S34. NOESY spectrum ($CDCl_3$, 500 MHz) of spoxazomicin D (**2**)

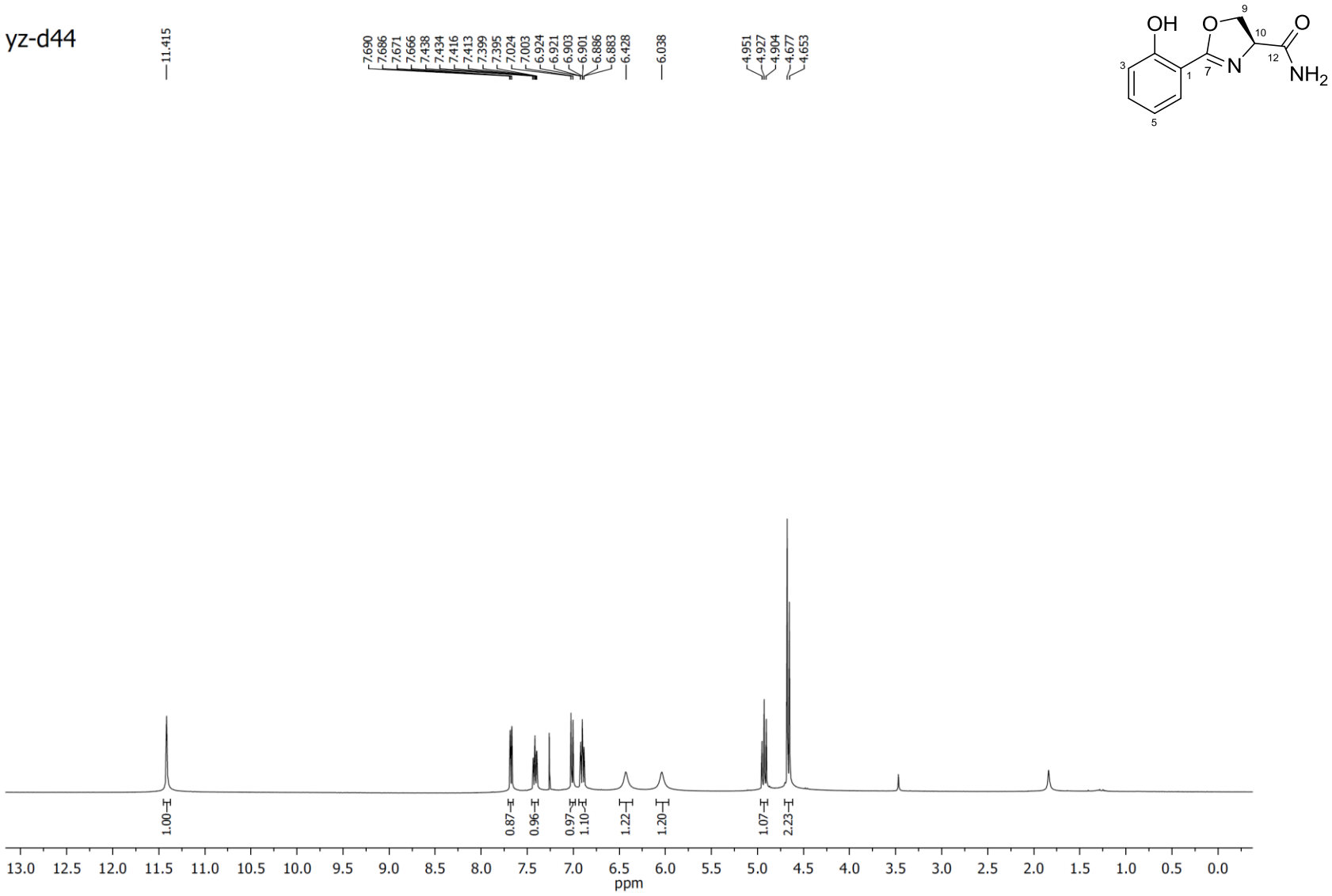


Figure S35. ^{1}H NMR spectrum ($CDCl_3$, 400 MHz) of synthesized spoxazomicin D (**2**)

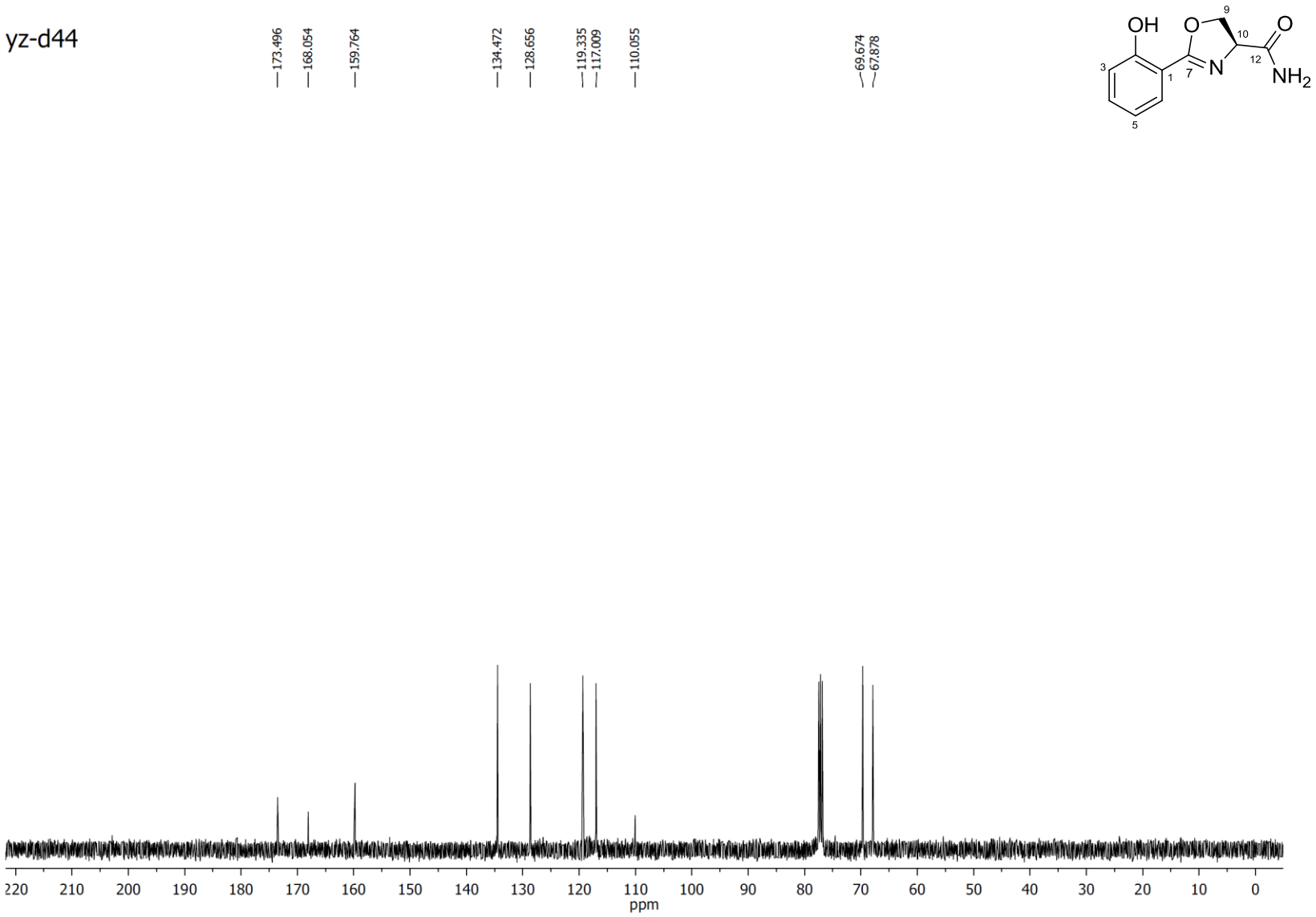


Figure S36. ^{13}C NMR spectrum ($CDCl_3$, 100 MHz) of synthesized spoxazomicin D (**2**)

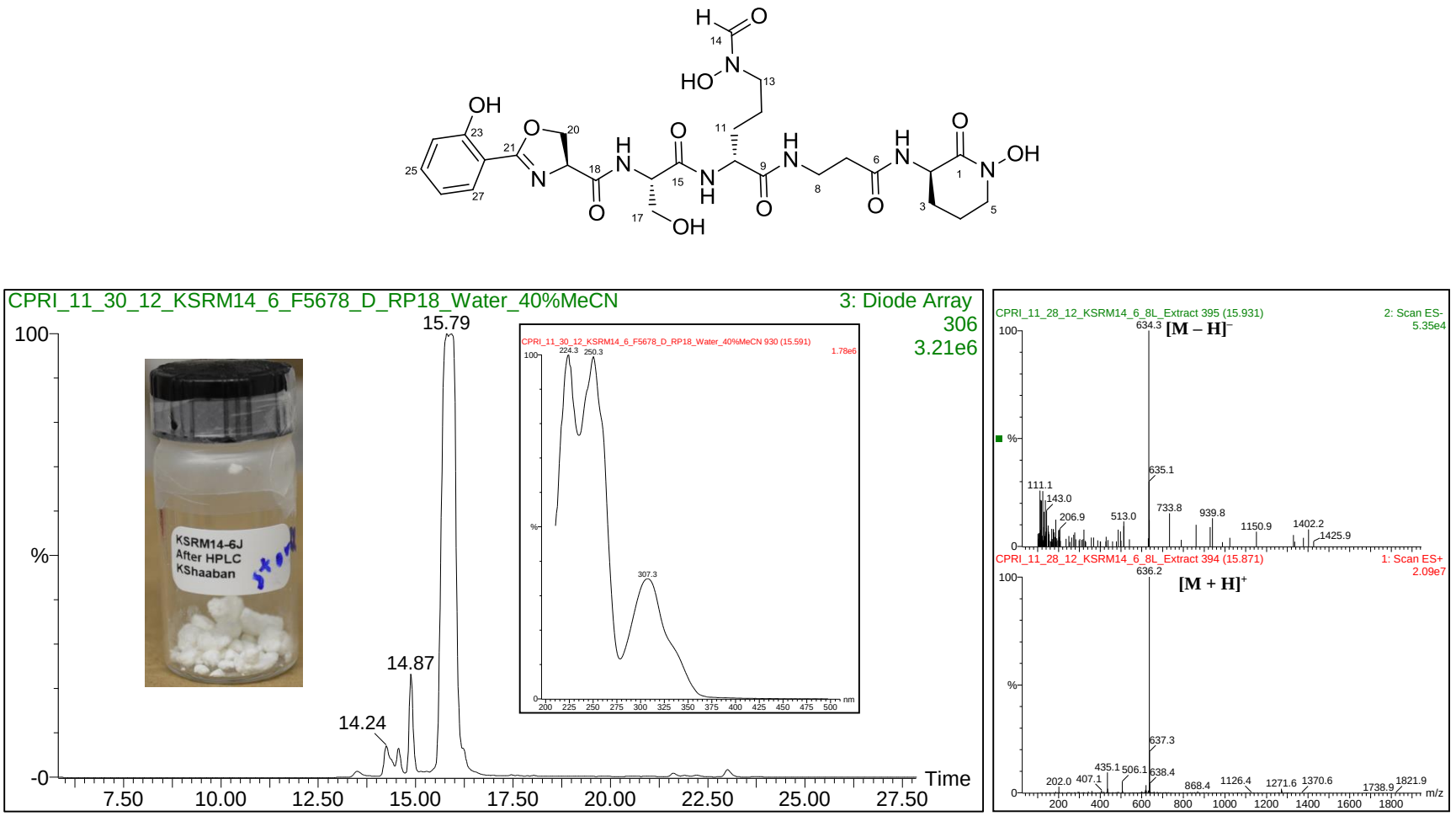


Figure S37. HPLC/UV/MS analyses of the purified oxachelin (**3**). Detection wavelength: 306 nm; **solvent A:** H_2O/0.1% Formic acid; **solvent B:** CH_3CN/0.1% Formic acid; flow rate: 0.5 mL min^{-1}; 0-4 min, 10% B; 4-22 min, 10-100% B; 22-27 min, 100% B; 27-29 min, 100%-10% B; 29-35 min, 10 % B.

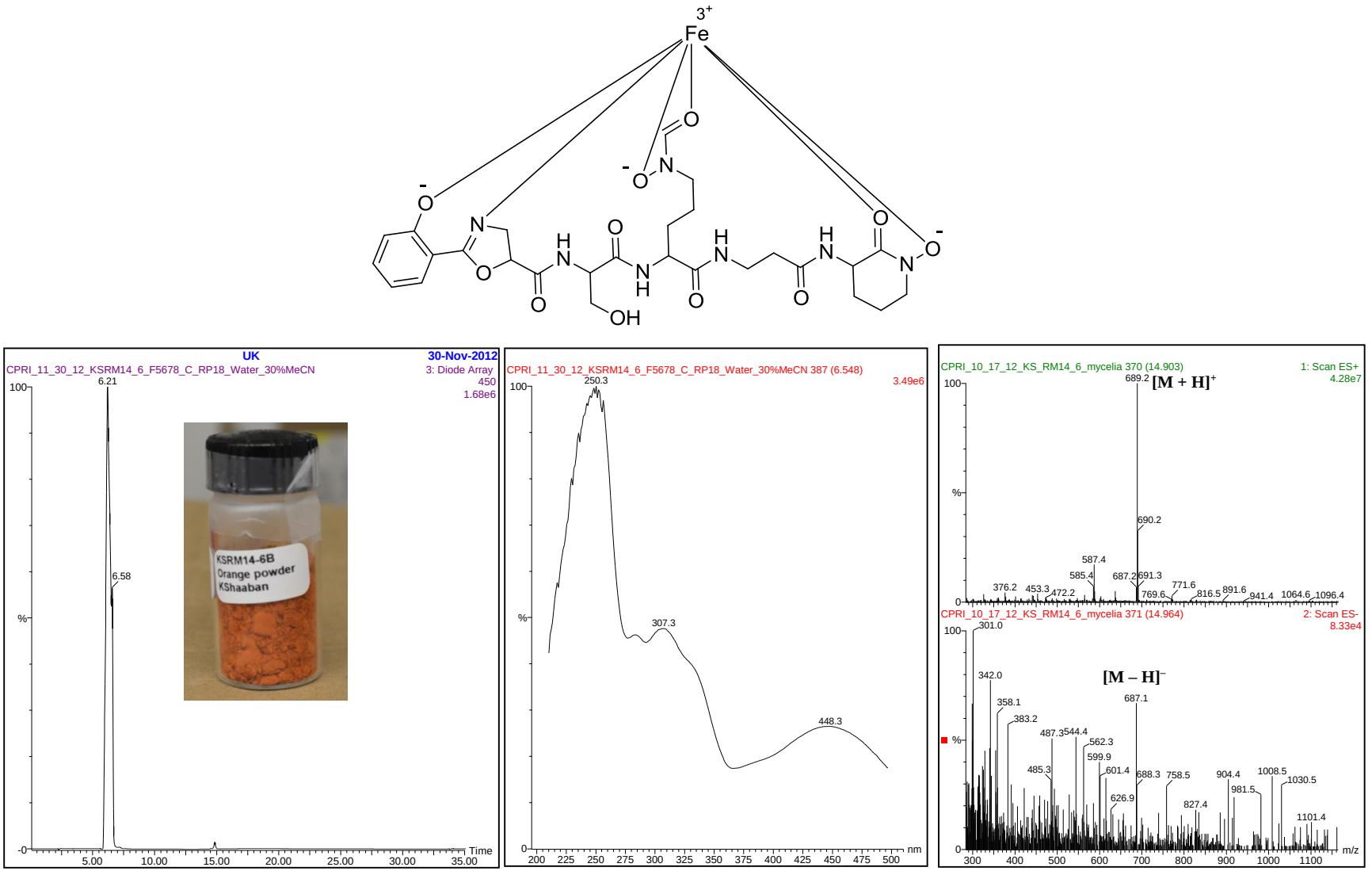


Figure S38. HPLC/UV/MS analyses of the purified oxachelin-Fe-complex (**18**). Detection wavelength: 450 nm; **solvent A:** H_2O/0.1% Formic acid; **solvent B:** CH_3CN/0.1% Formic acid; flow rate: 0.5 mL min^{-1}; 0-4 min, 10% B; 4-22 min, 10-100% B; 22-27 min, 100% B; 27-29 min, 100%-10% B; 29-35 min, 10 % B.

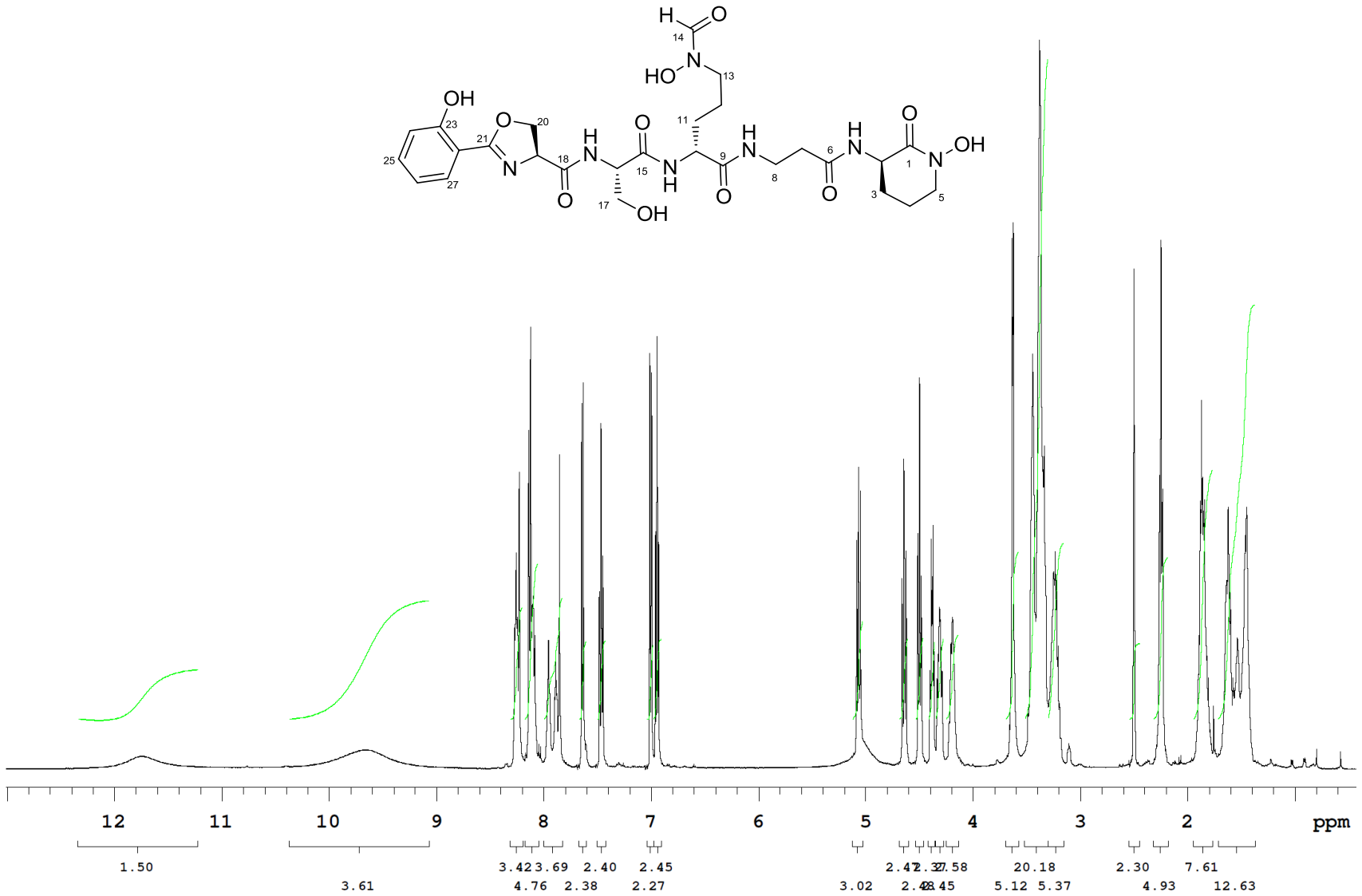


Figure S39. ^{1}H NMR spectrum (DMSO-d_6, 500 MHz) of oxachelin (**3**)

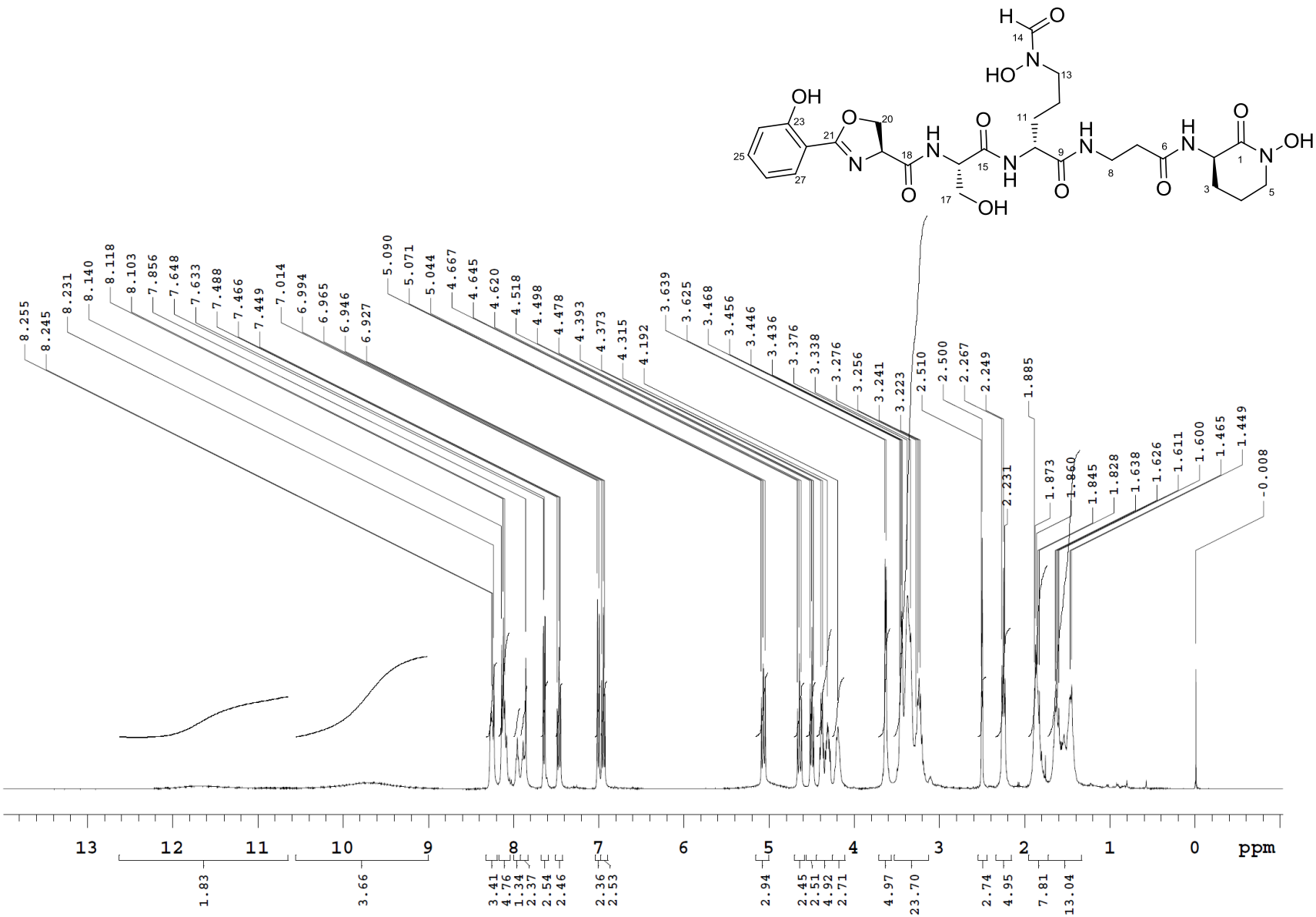


Figure S40. ^{1}H NMR spectrum (DMSO-d_6, 400 MHz) of oxachelin (**3**)

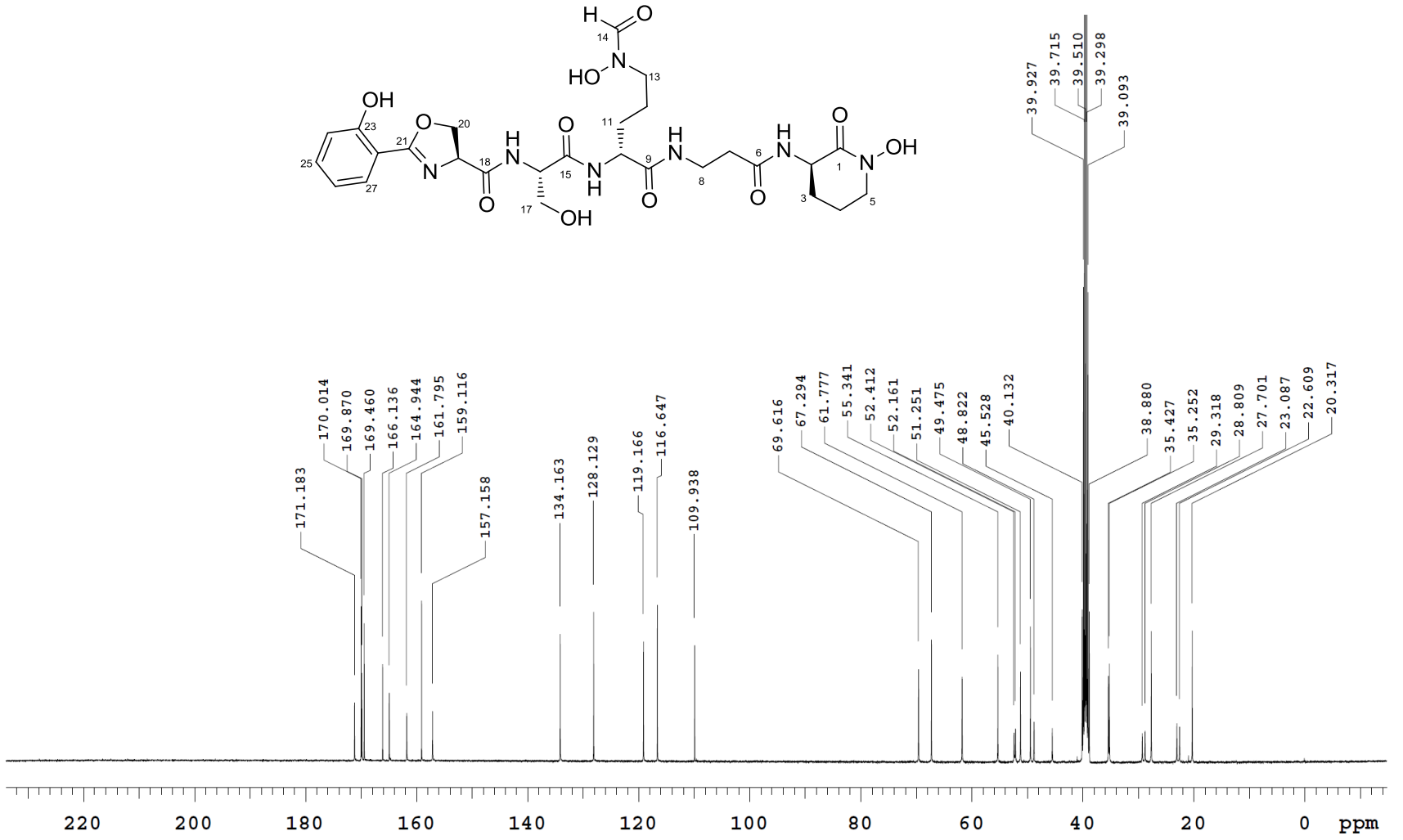


Figure S41. ^{13}C NMR spectrum (DMSO-d_6, 100 MHz) of oxachelin (**3**)

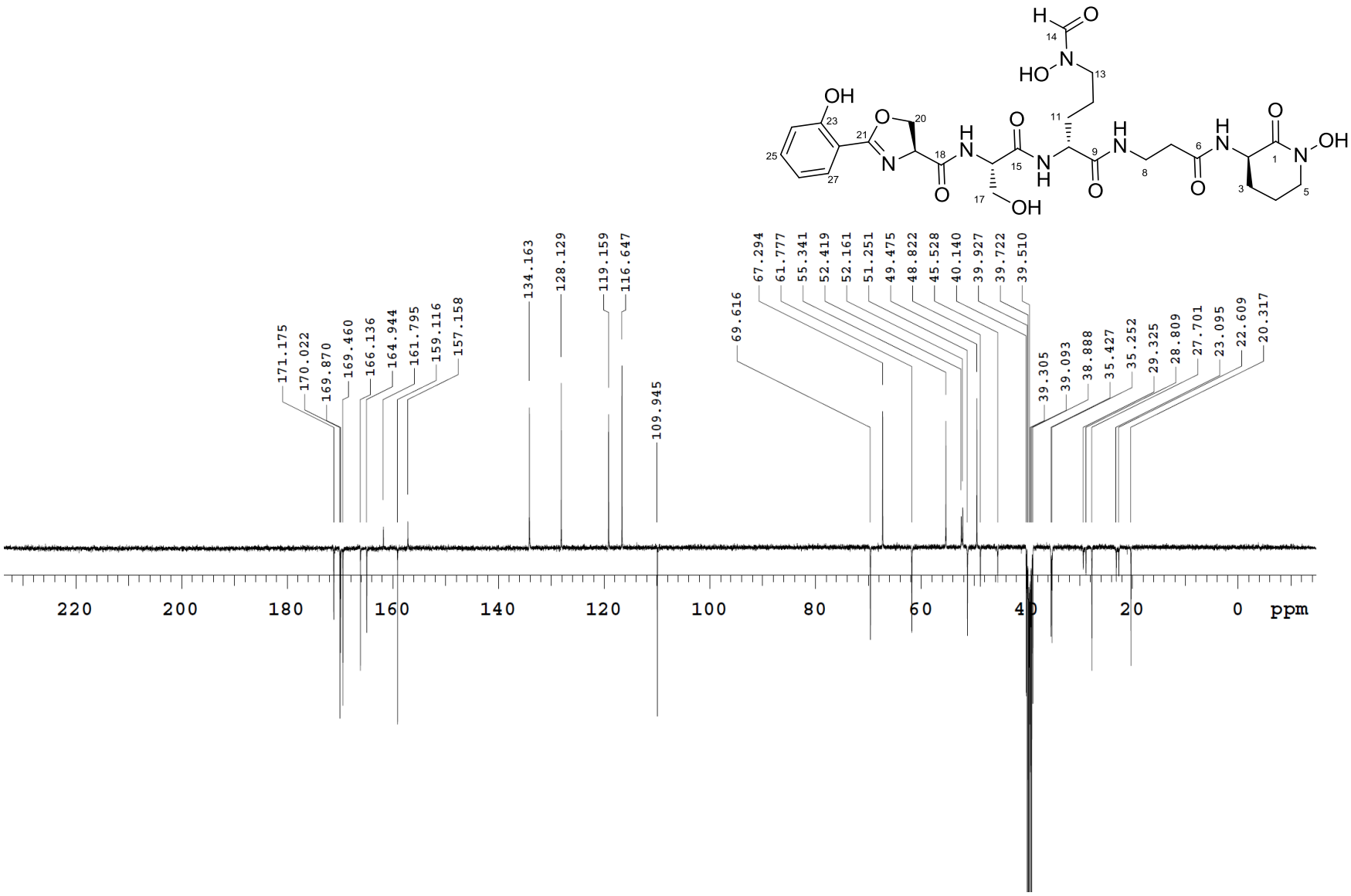


Figure S42. APT NMR spectrum (DMSO-d_6, 100 MHz) of oxachelin (**3**)

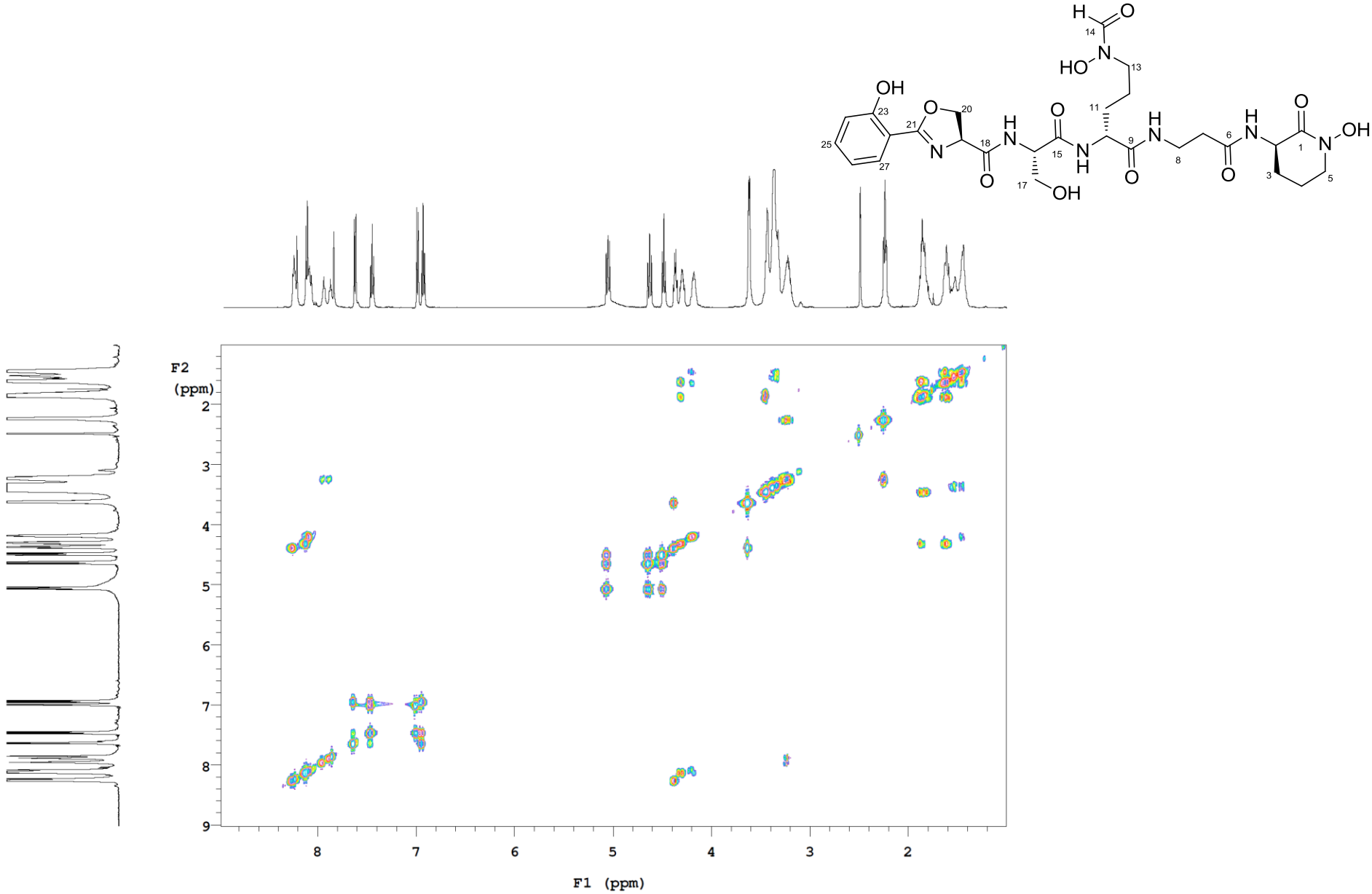


Figure S43. ^{1}H,^{1}H-COSY spectrum (DMSO-d_6, 500 MHz) of oxachelin (**3**)

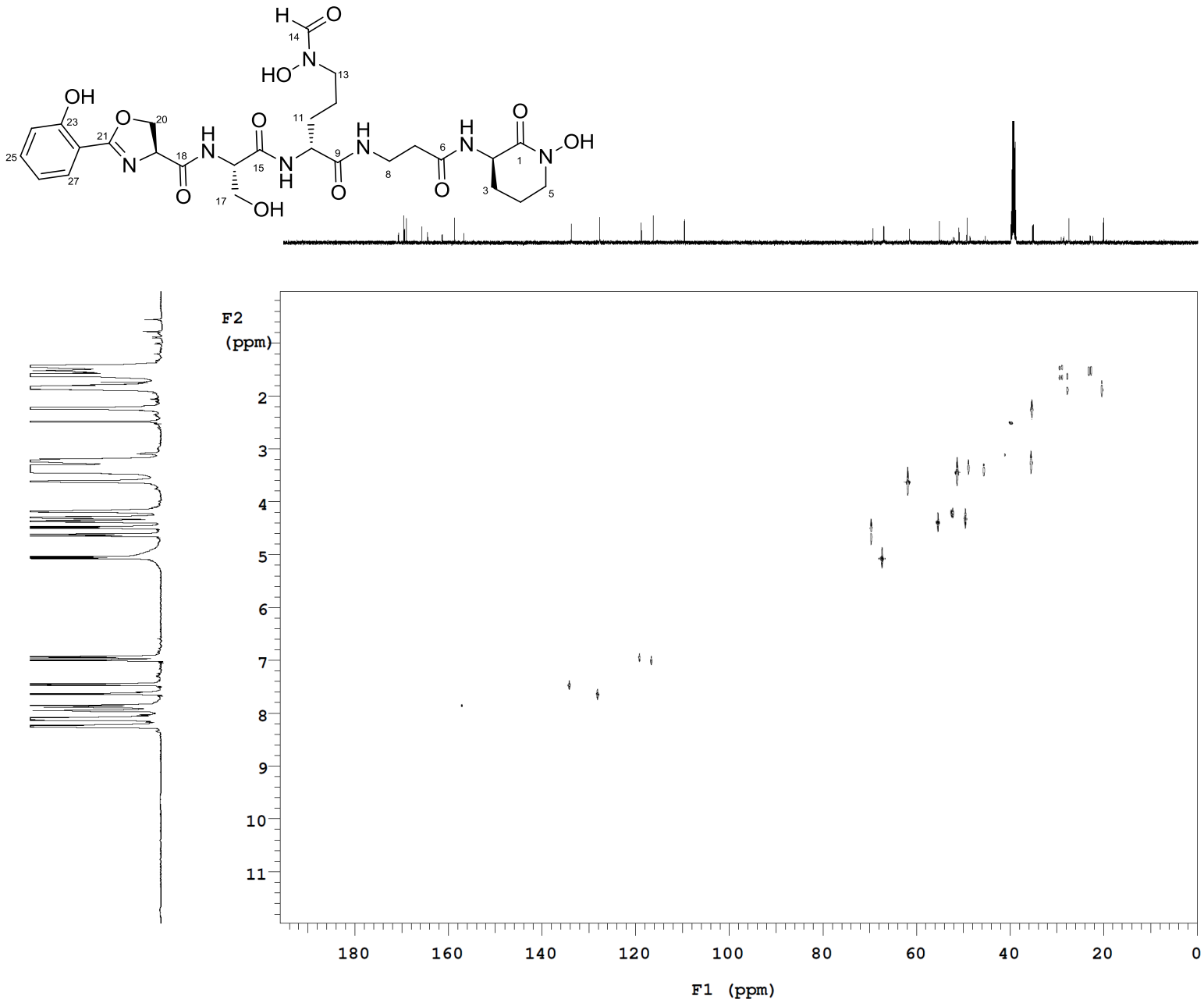


Figure S44. HSQC spectrum (DMSO-d_6, 500 MHz) of oxachelin (**3**)

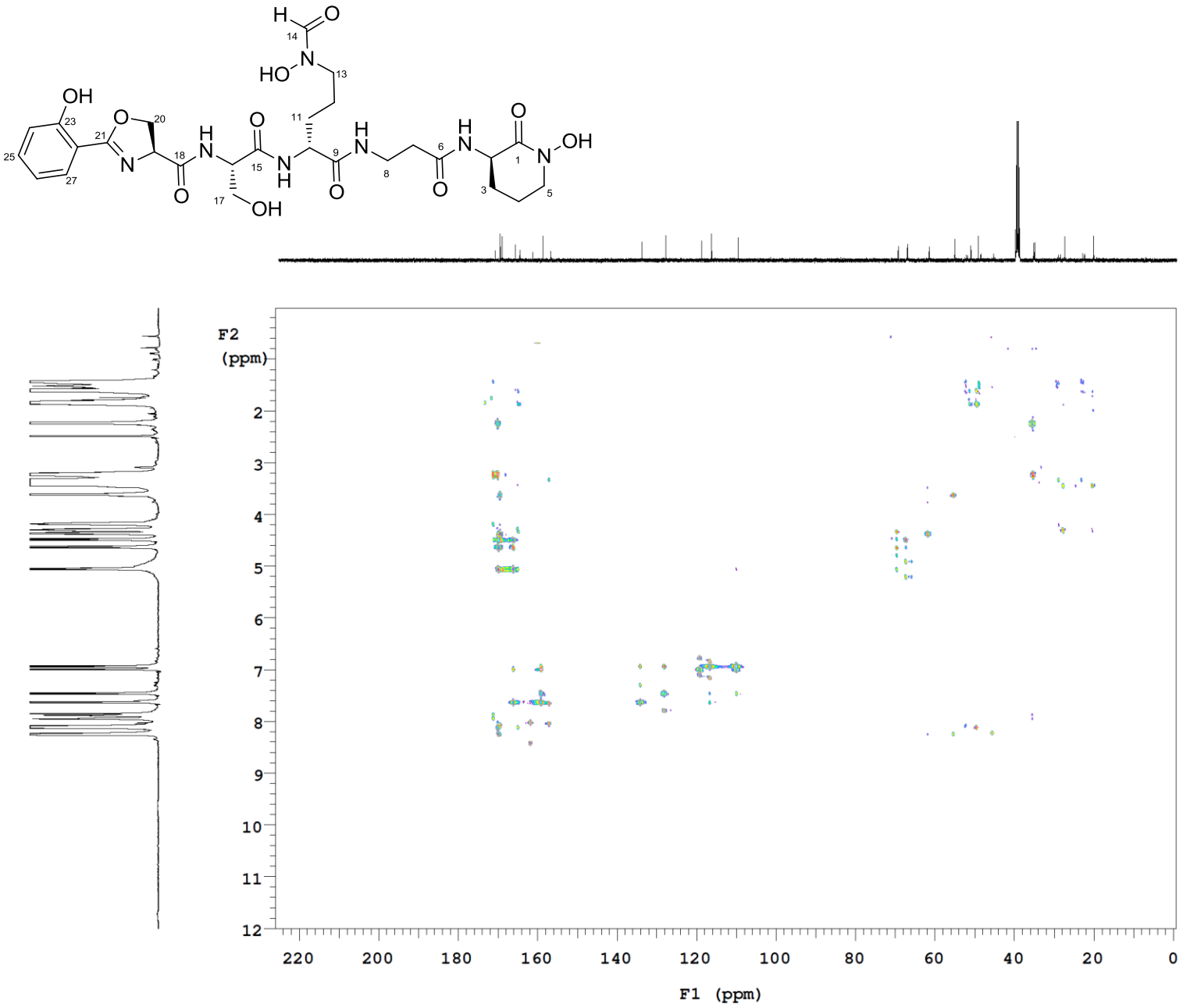


Figure S45. HMBC spectrum (DMSO-d_6, 500 MHz) of oxachelin (**3**)

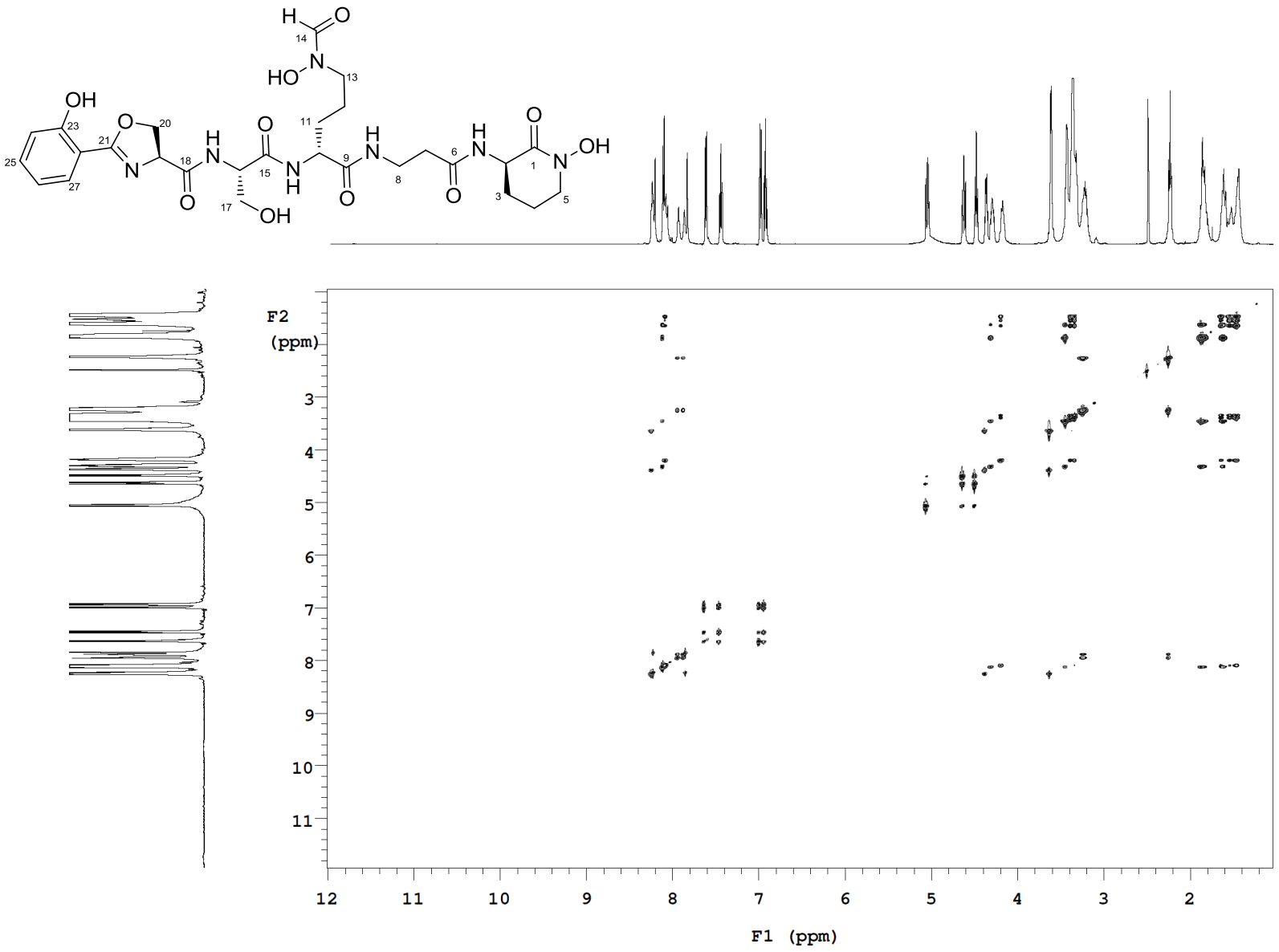


Figure S46. TOCSY spectrum (DMSO-d_6, 500 MHz) of oxachelin (**3**)

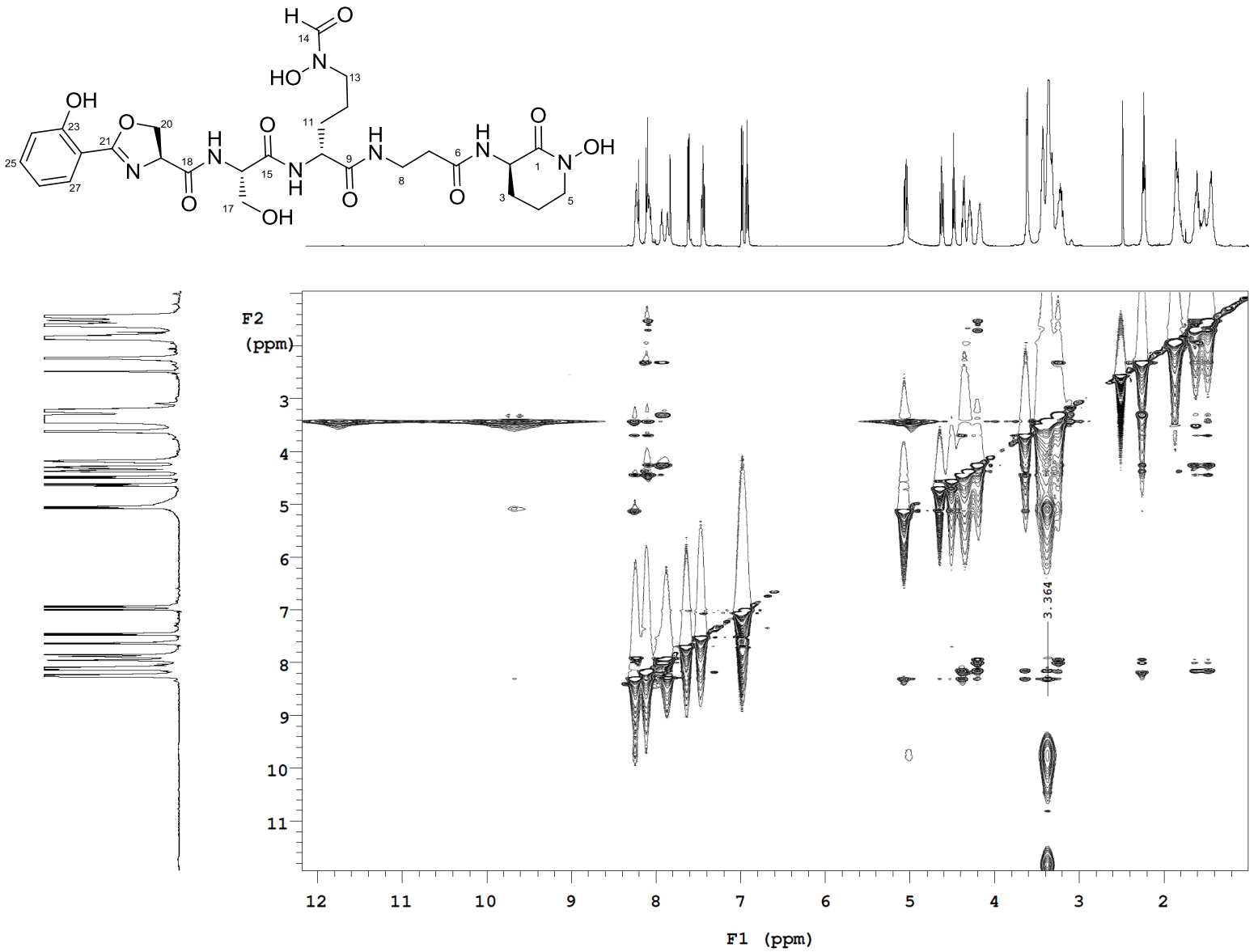


Figure S47. NOESY spectrum (DMSO-d_6, 500 MHz) of oxachelin (**3**)

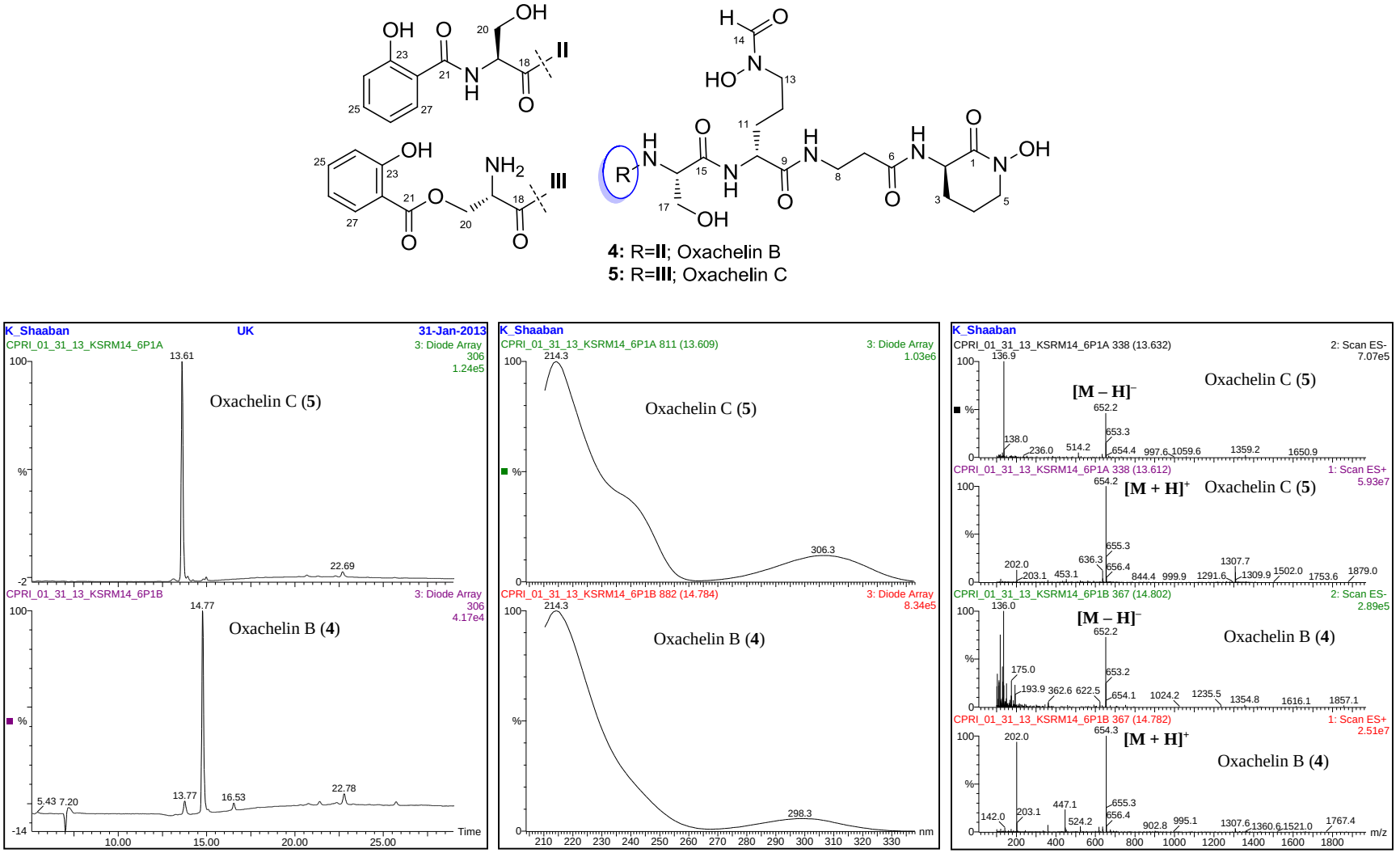


Figure S48. HPLC/UV/MS analyses of the purified oxachelins B-C (**4**-**5**). Detection wavelength: 306 nm; **solvent A:** H_2O/0.1% Formic acid; **solvent B:** CH_3CN/0.1% Formic acid; flow rate: 0.5 mL min^{-1}; 0-4 min, 10% B; 4-22 min, 10-100% B; 22-27 min, 100% B; 27-29 min, 100%-10% B; 29-35 min, 10 % B.

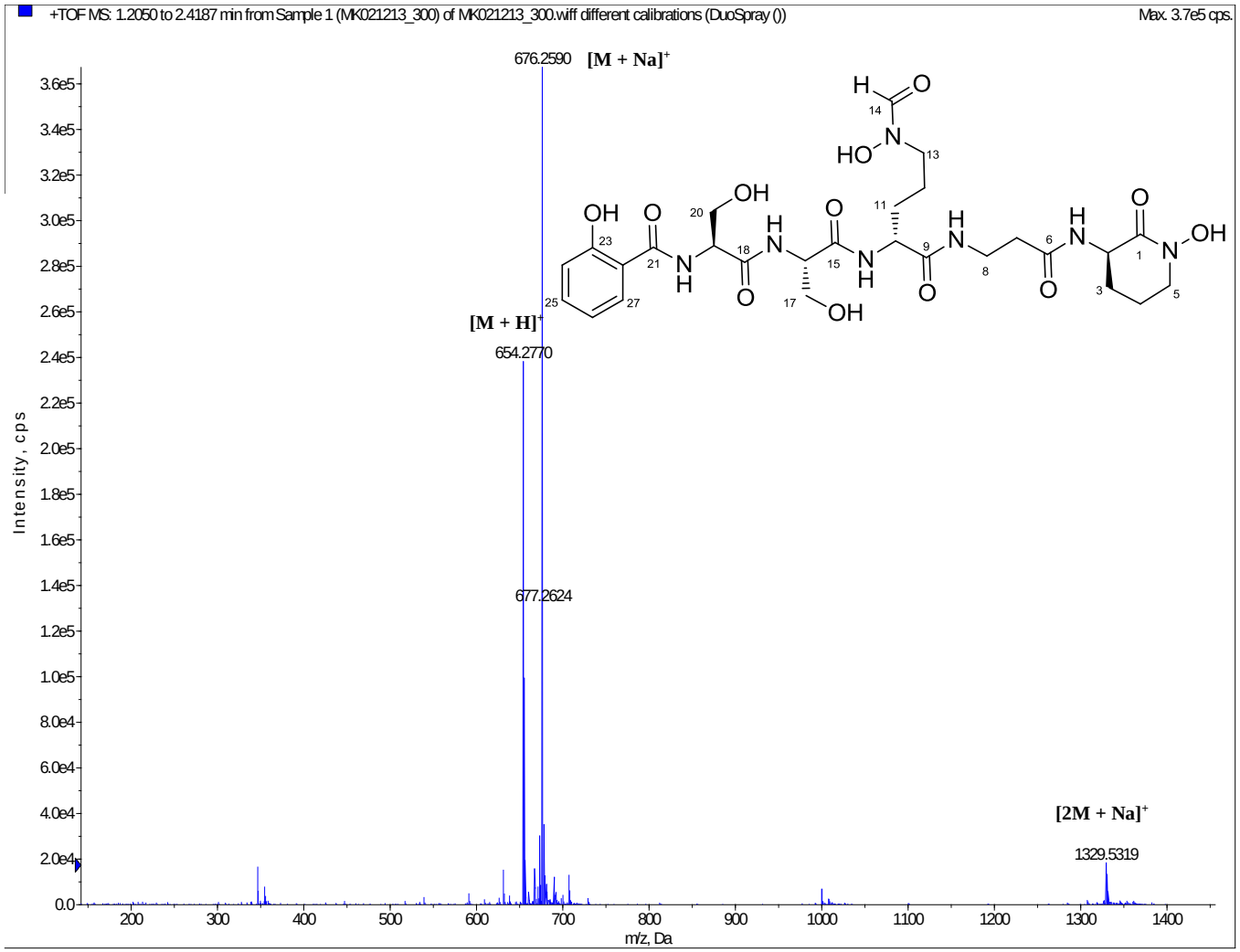


Figure S49. (+)-HRESI-MS spectrum of oxachelin B (**4**)

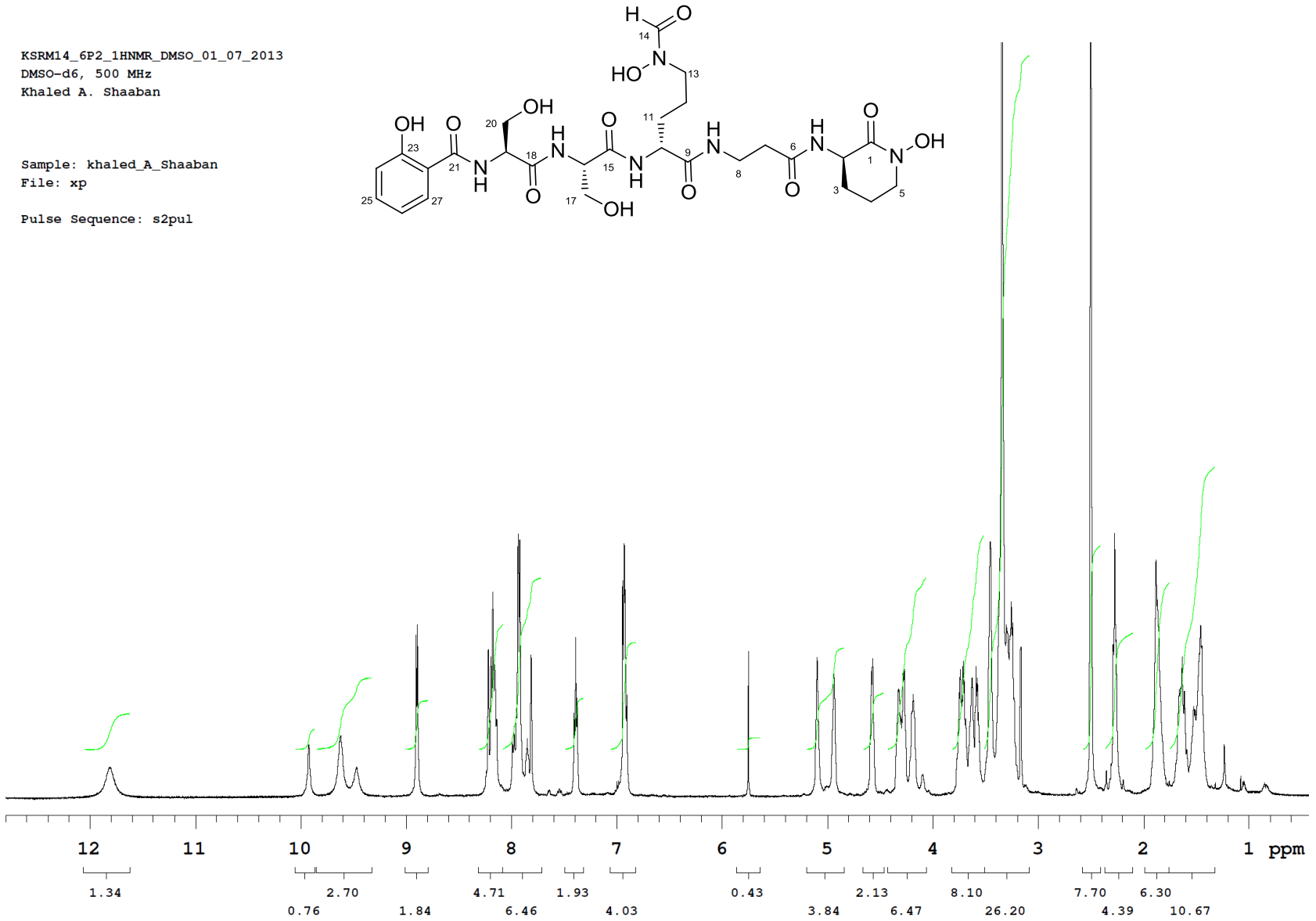


Figure S50. ^{1}H NMR spectrum (DMSO-d_6, 500 MHz) of oxachelin B (**4**)

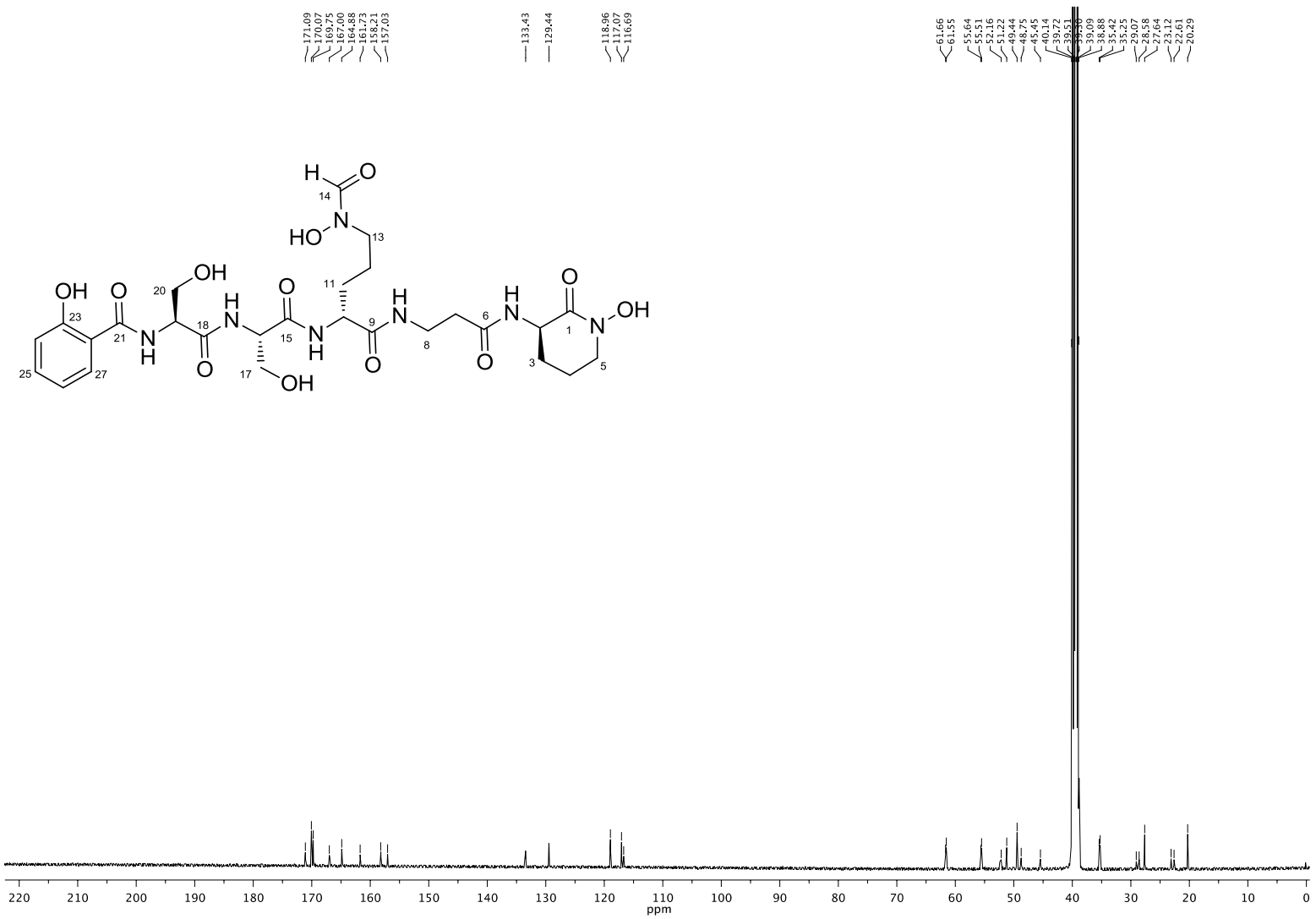


Figure S51. ^{13}C NMR spectrum (DMSO-d_6, 100 MHz) of oxachelin B (**4**)

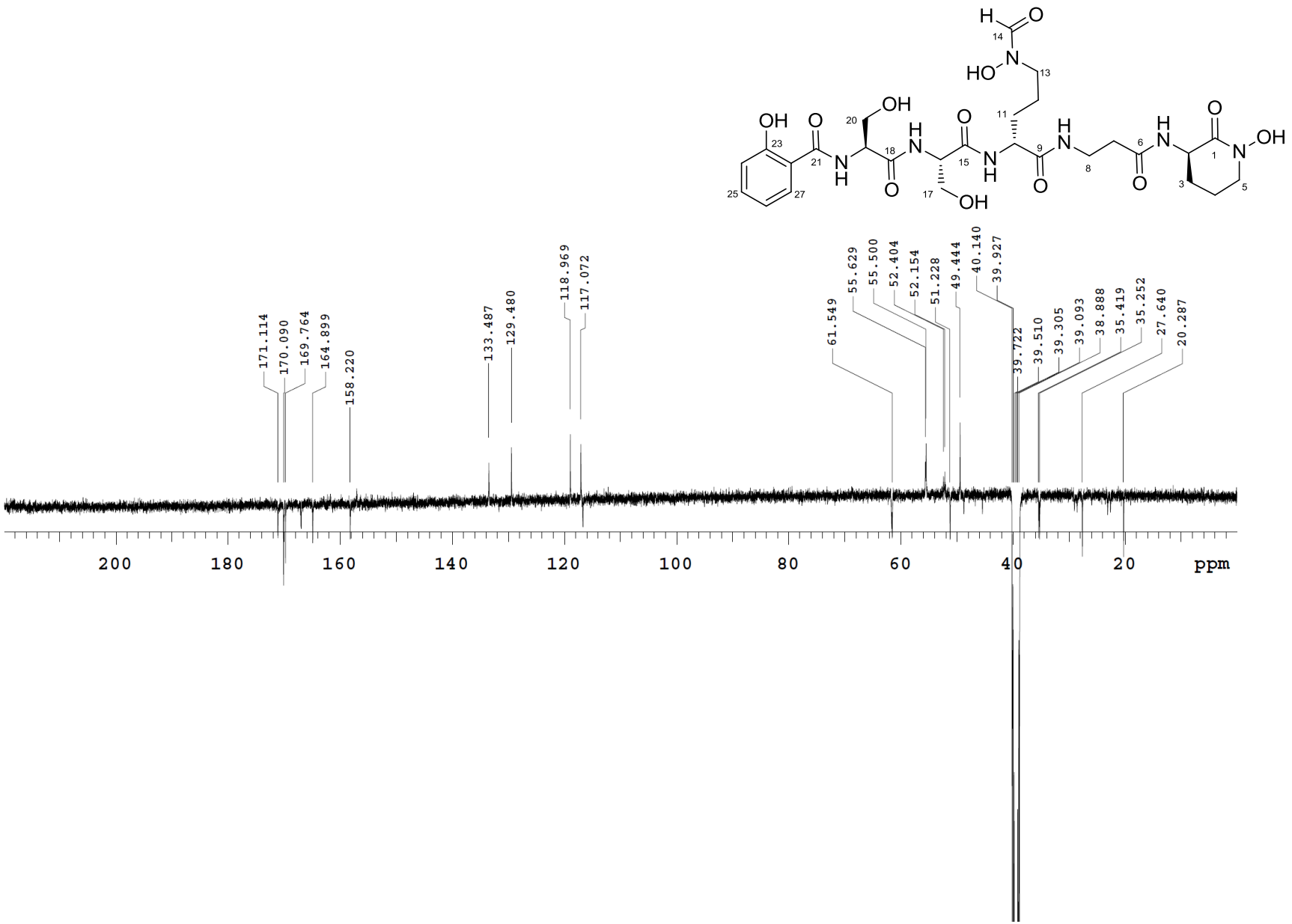


Figure S52. APT NMR spectrum (DMSO-d_6, 100 MHz) of oxachelin B (**4**)

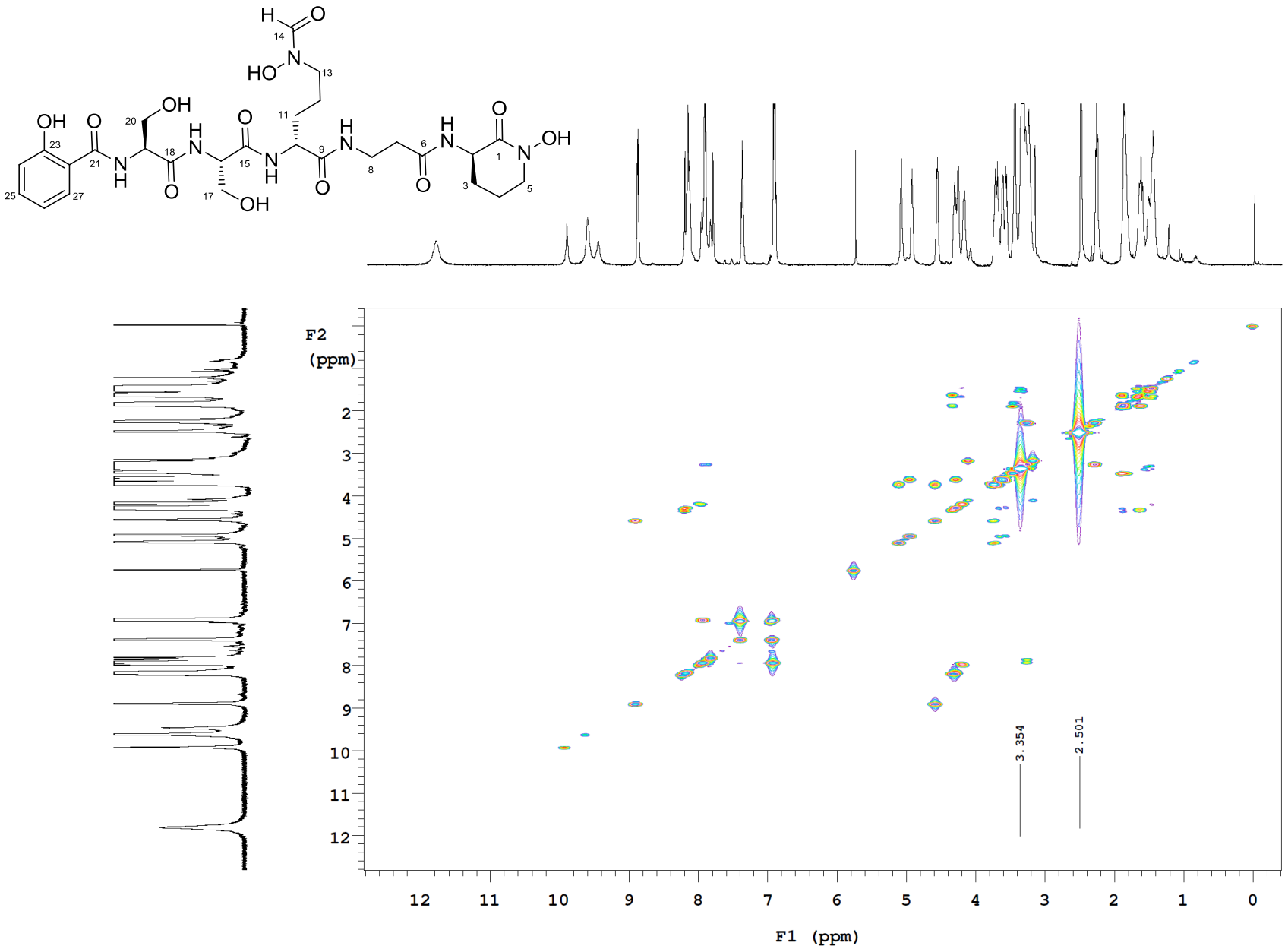


Figure S53. ^{1}H,^{1}H-COSY spectrum (DMSO-d_6, 500 MHz) of oxachelin B (**4**)

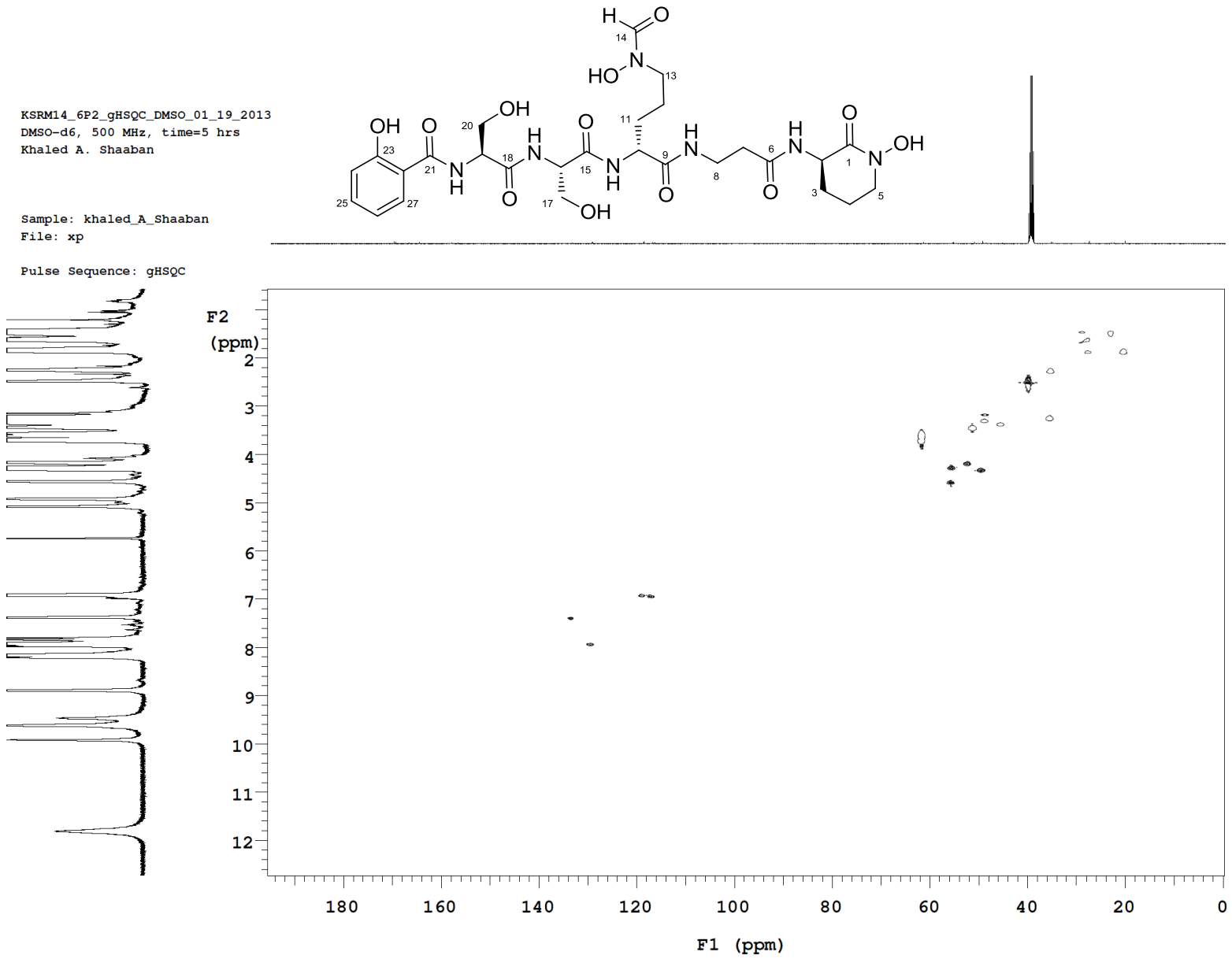


Figure S54. HSQC spectrum (DMSO-d_6, 500 MHz) of oxachelin B (**4**)

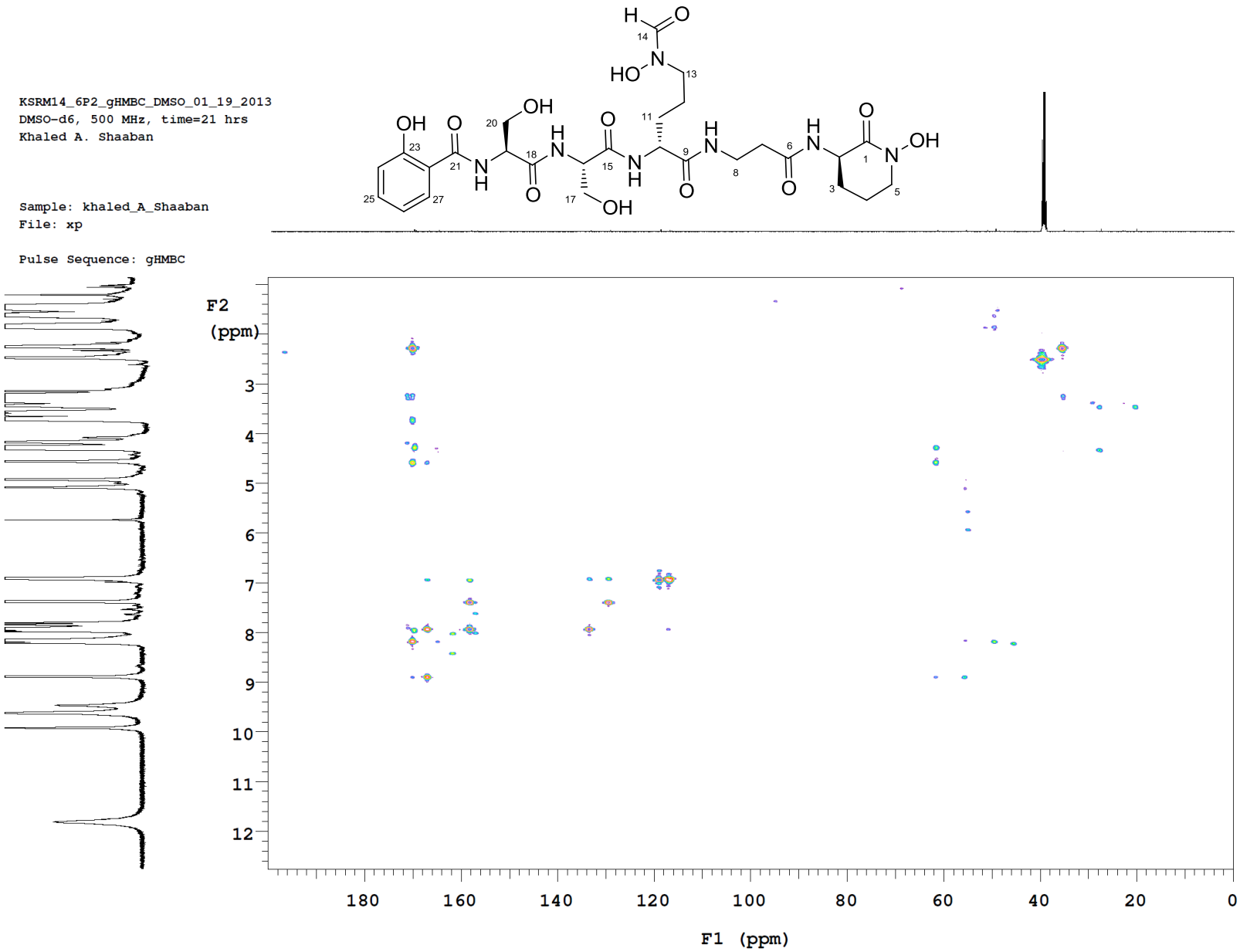


Figure S55. HMBC spectrum (DMSO-d_6, 500 MHz) of oxachelin B (**4**)

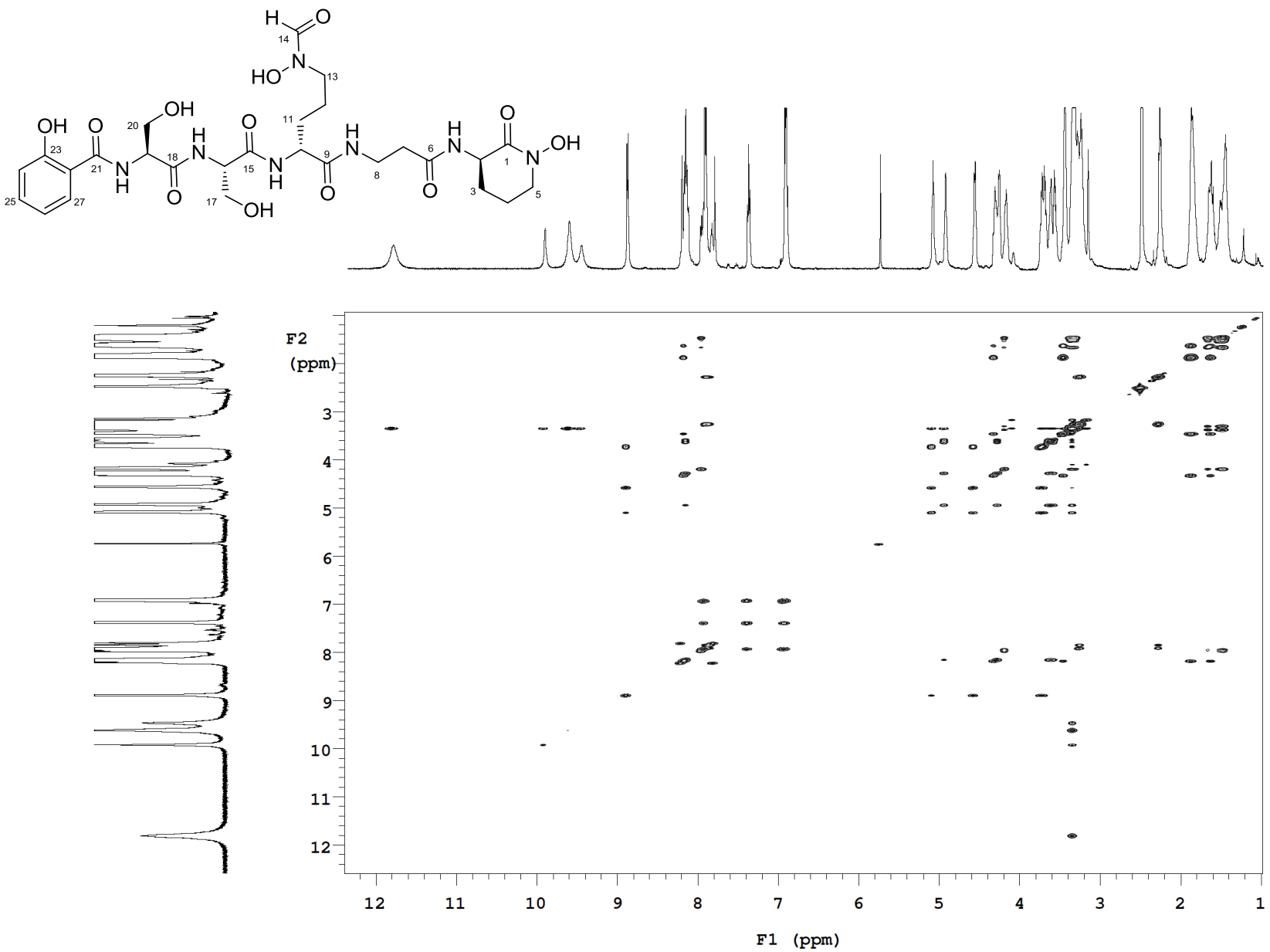


Figure S56. TOCSY spectrum (DMSO-d_6, 500 MHz) of oxachelin B (**4**)

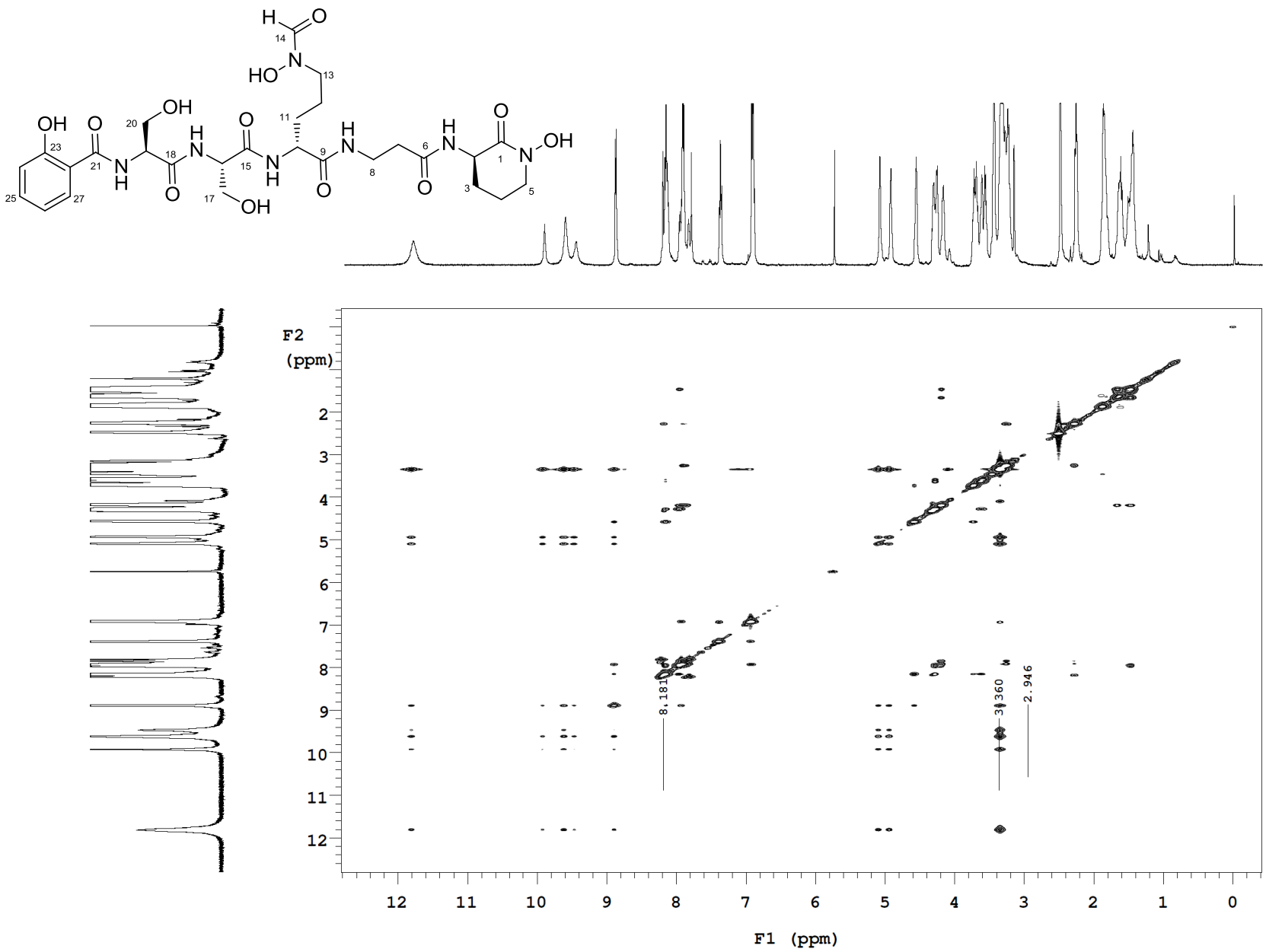


Figure S57. NOESY spectrum (DMSO-d_6, 500 MHz) of oxachelin B (**4**)

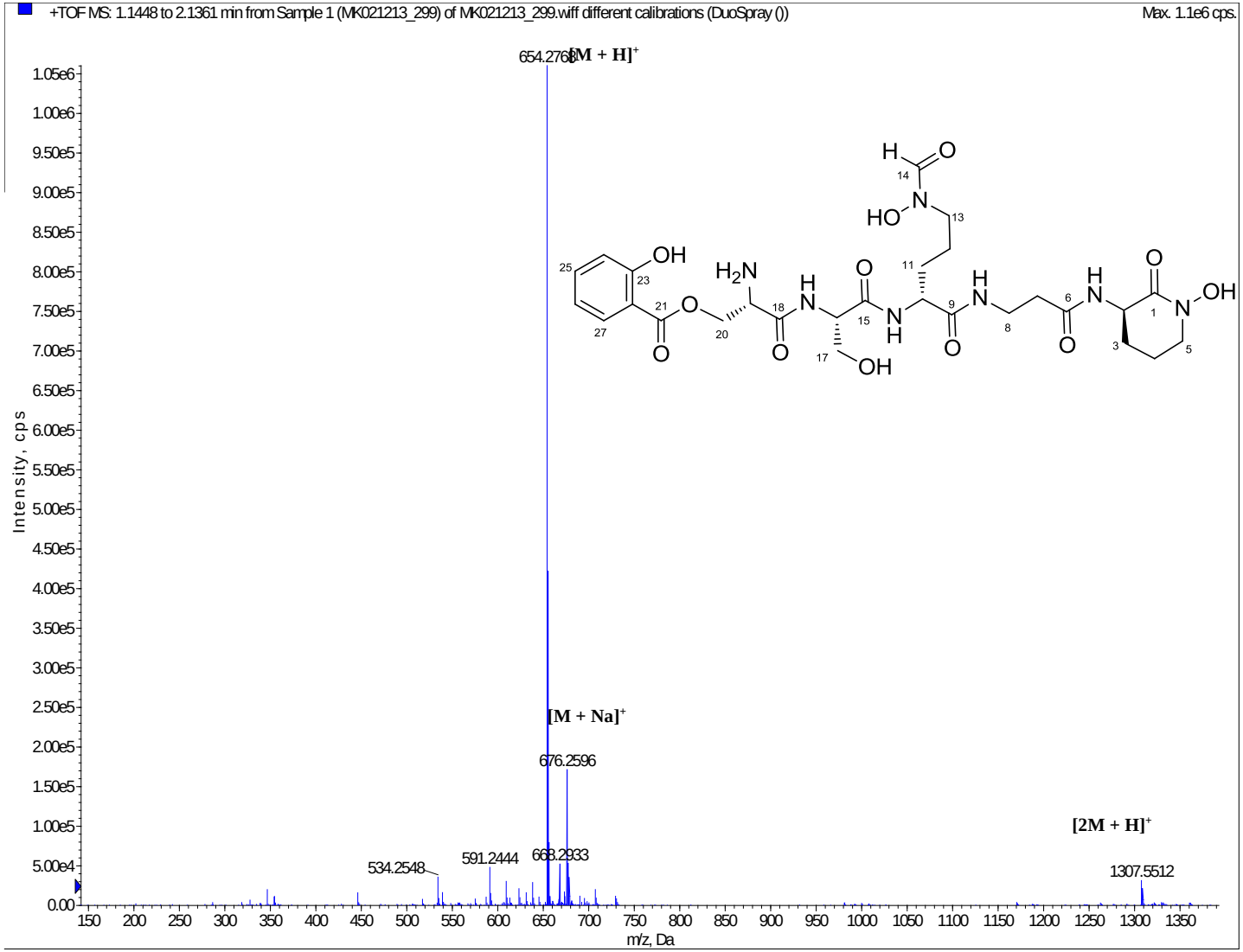


Figure S58. (+)-HRESI-MS spectrum of oxachelin C (**5**)

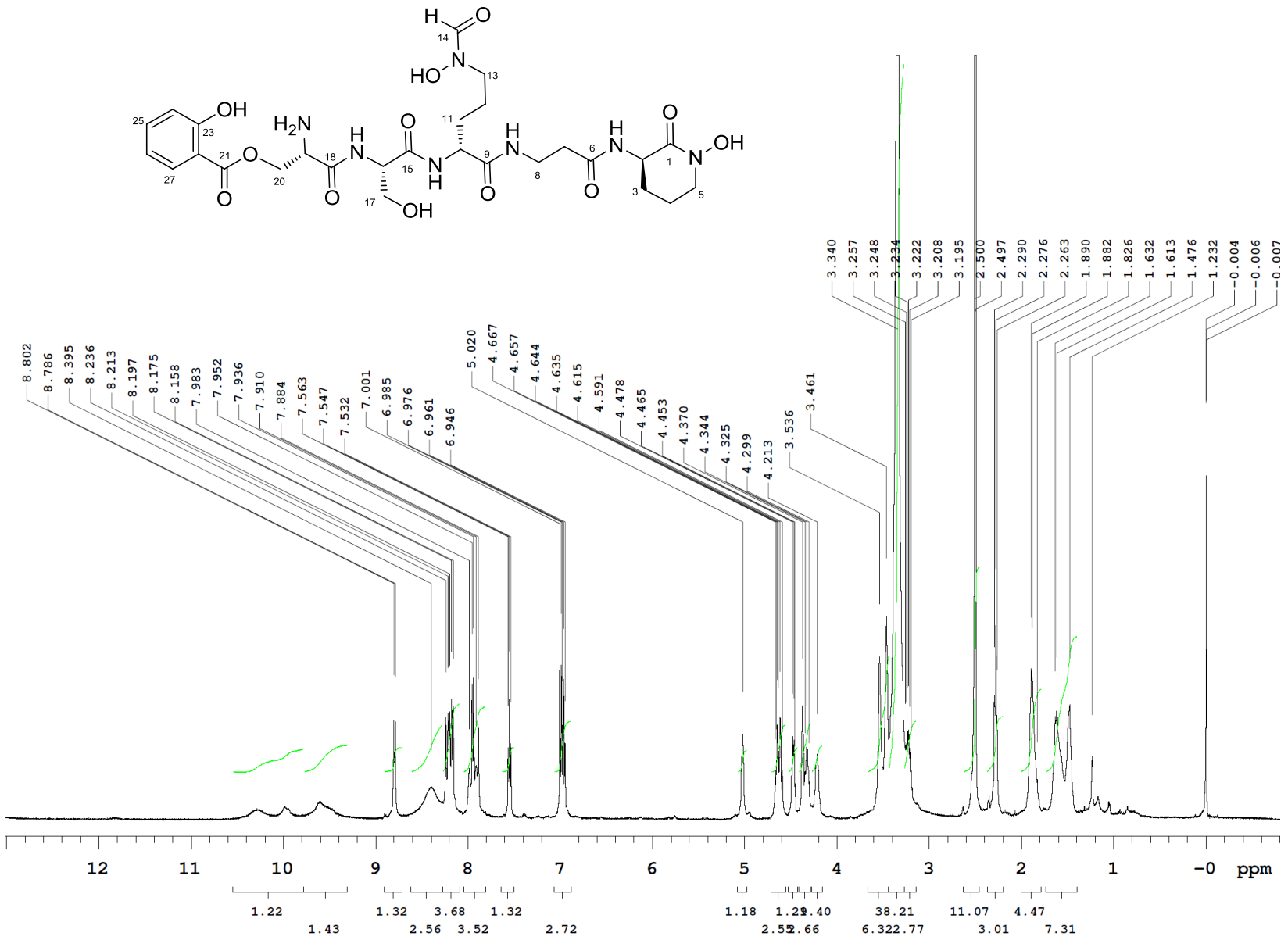


Figure S59. ^{1}H NMR spectrum (DMSO-d_6, 500 MHz) of oxachelin C (**5**)

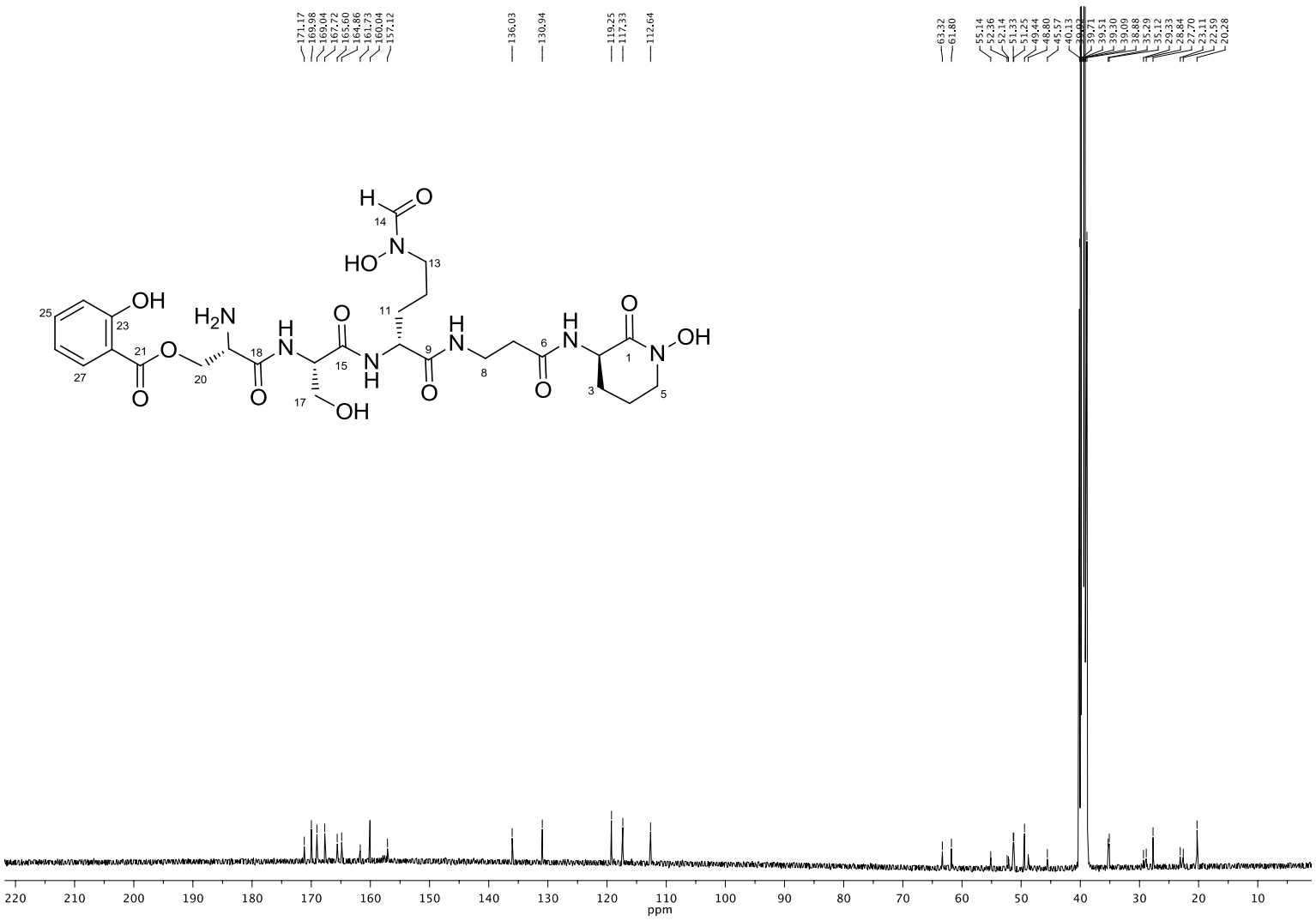


Figure S60. ^{13}C NMR spectrum (DMSO-d_6, 100 MHz) of oxachelin C (**5**)

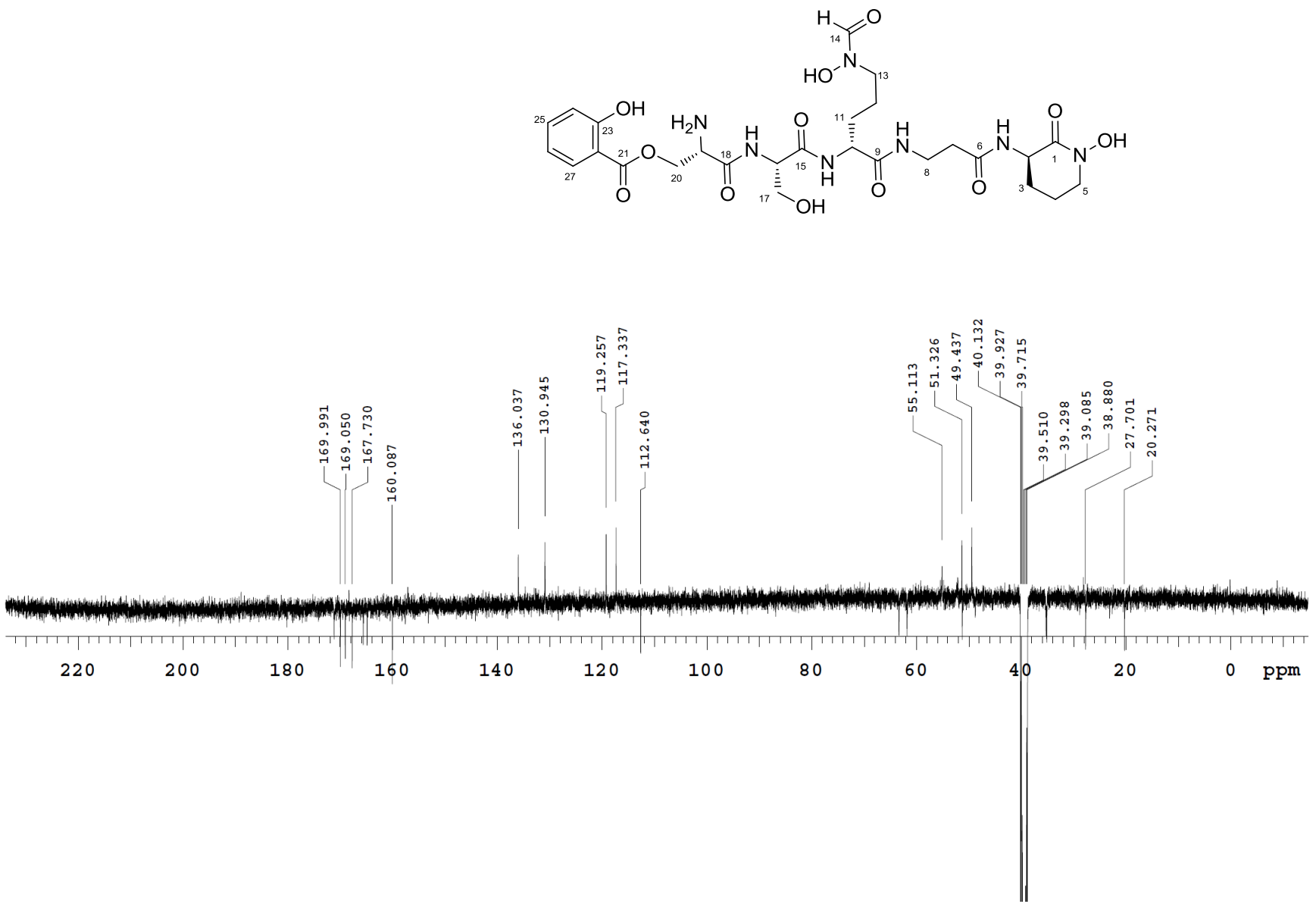


Figure S61. APT NMR spectrum (DMSO-d_6, 100 MHz) of oxachelin C (**5**)

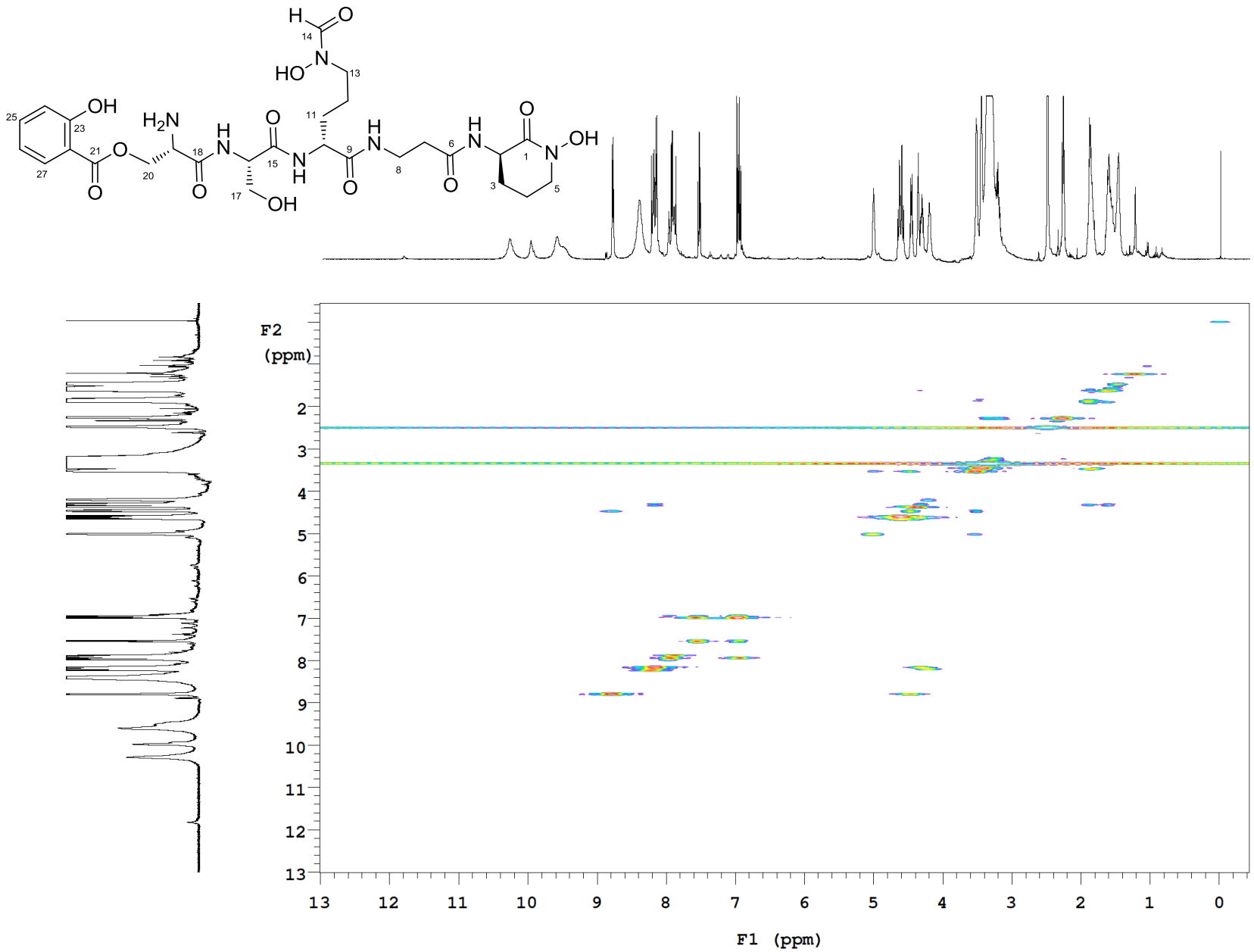


Figure S62. ^{1}H,^{1}H-COSY spectrum (DMSO-d_6, 500 MHz) of oxachelin C (**5**)

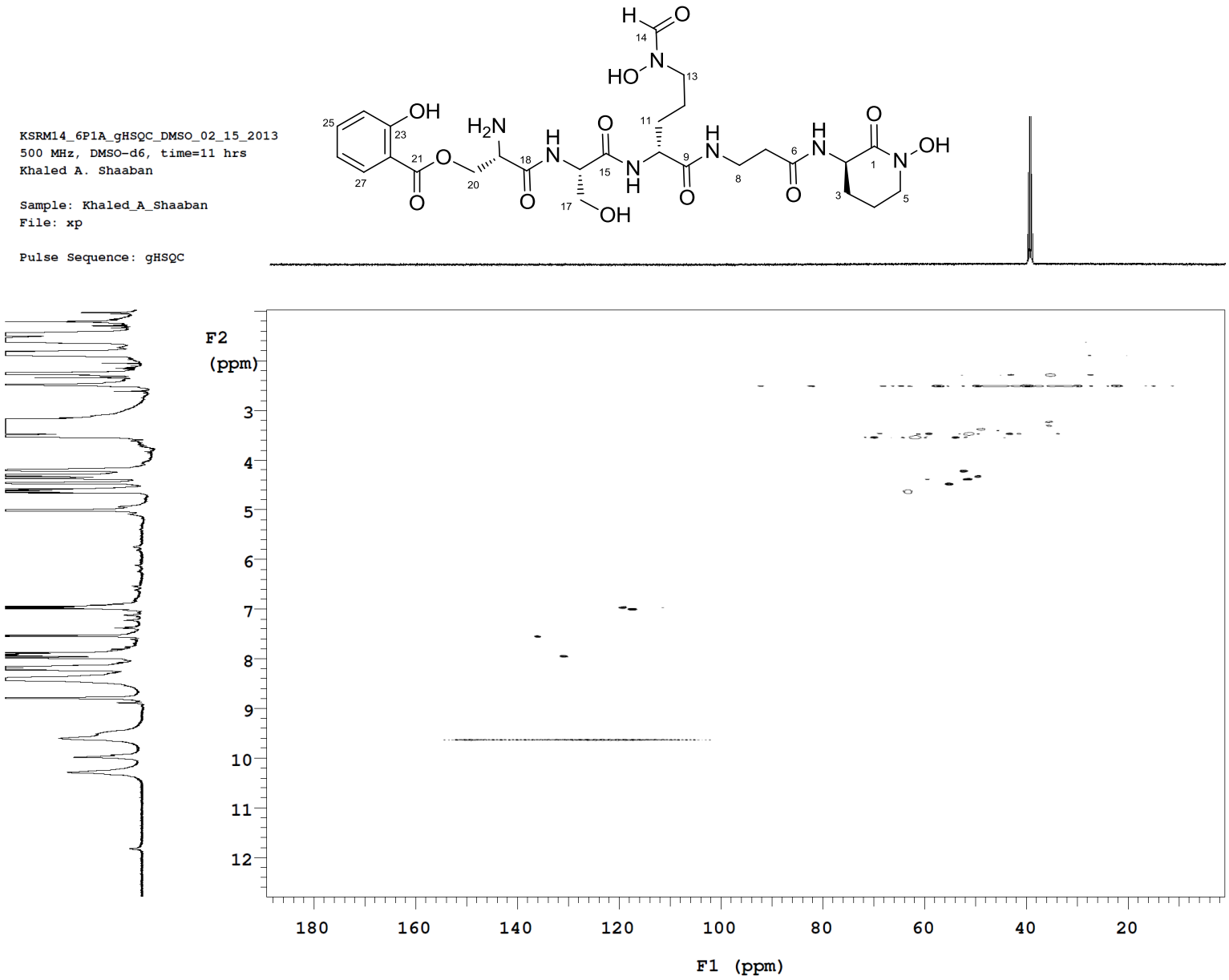


Figure S63. HSQC spectrum (DMSO-d_6, 500 MHz) of oxachelin C (**5**)

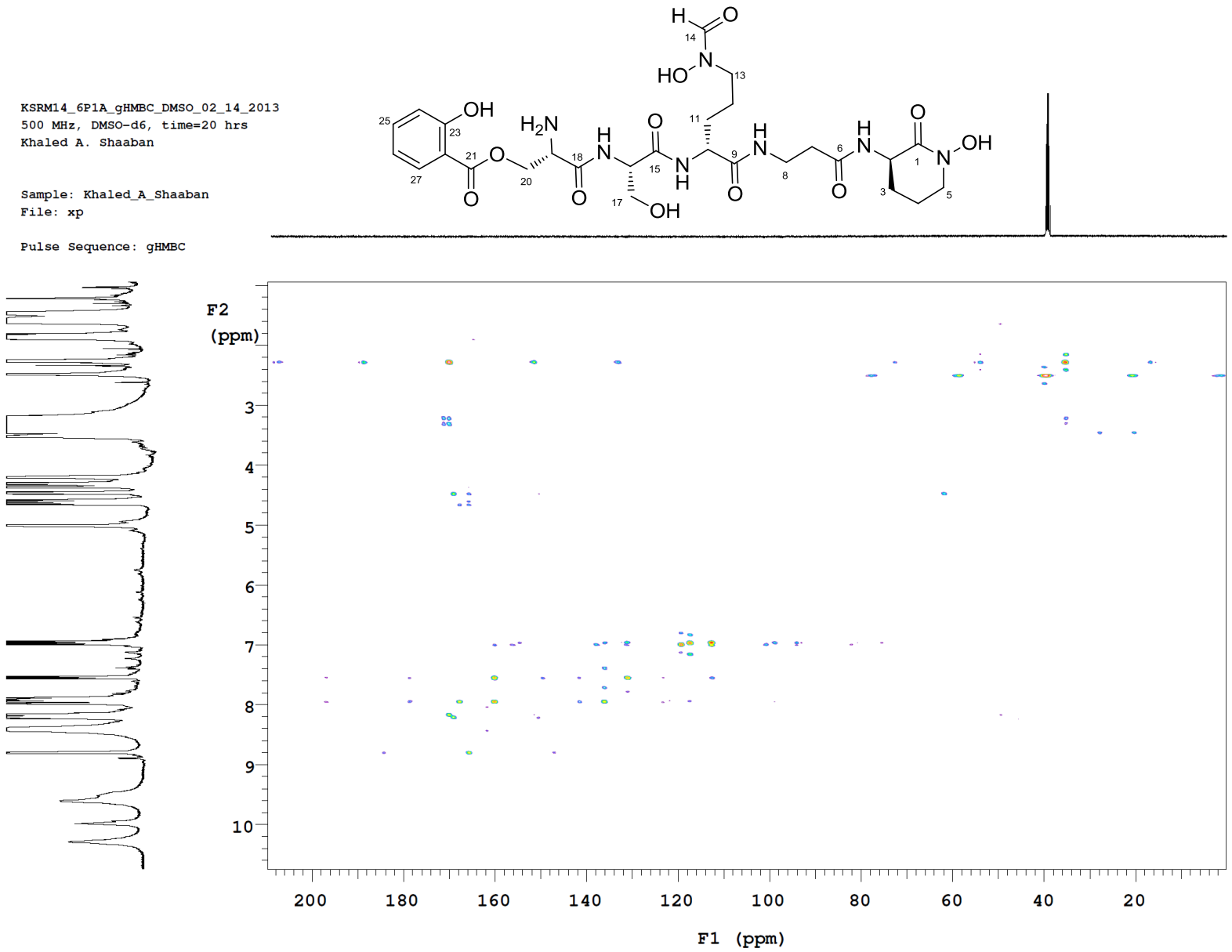


Figure S64. HMBC spectrum (DMSO-d_6, 500 MHz) of oxachelin C (**5**)

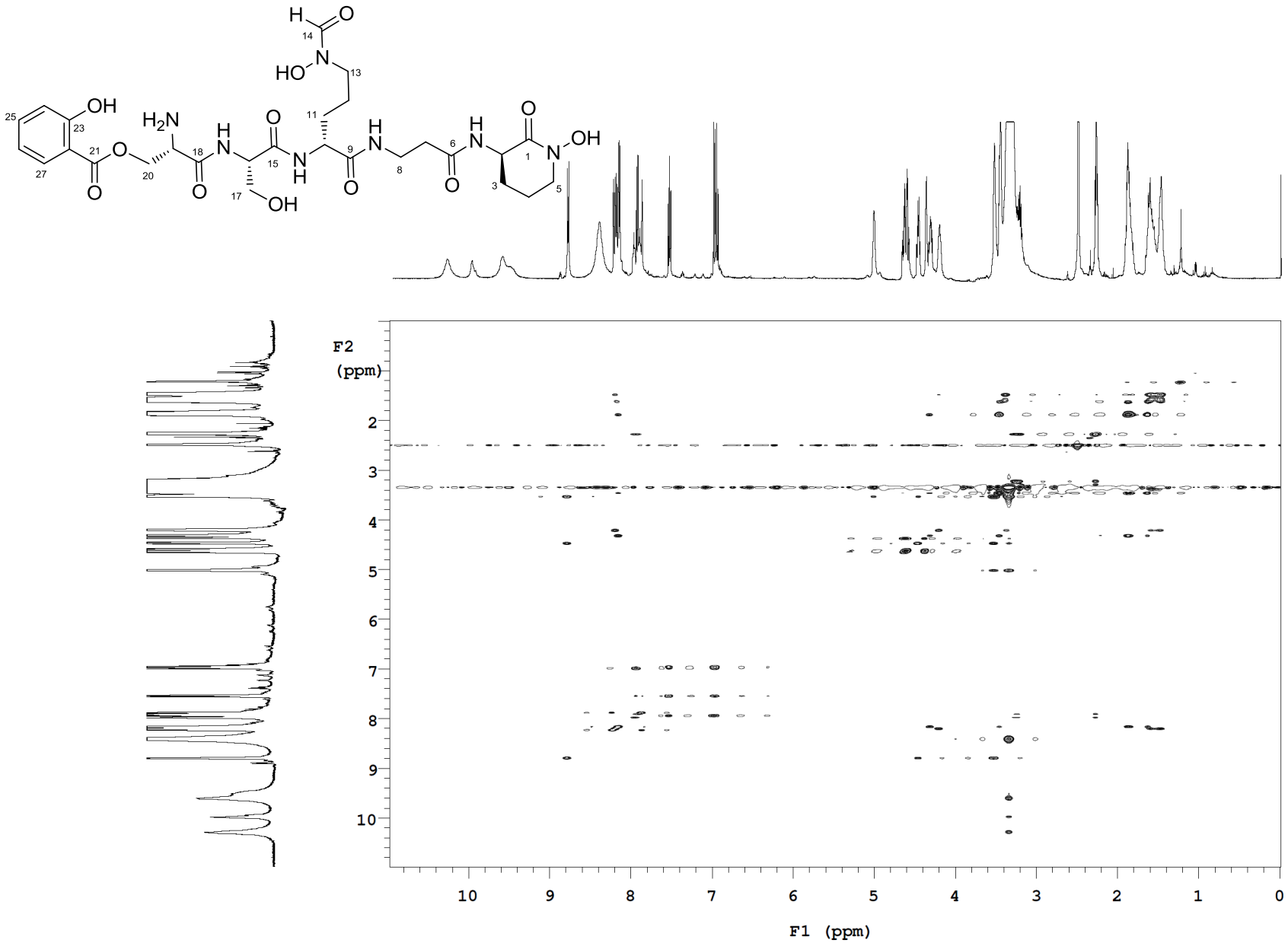


Figure S65. TOCSY spectrum (DMSO-d_6, 500 MHz) of oxachelin C (**5**)

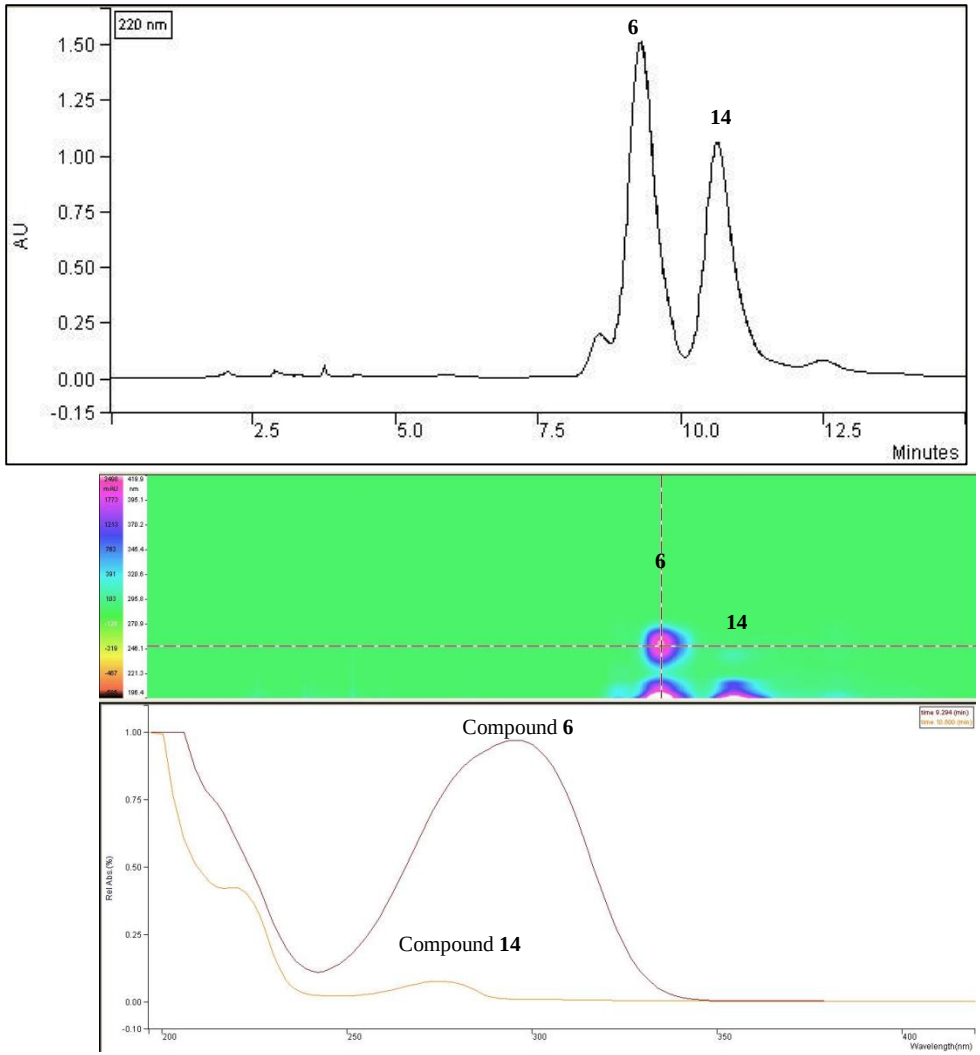


Figure S66. HPLC/UV purification of compounds **6** and **14**. Detection wavelength: 220 nm; **solvent A:** H_2O; **solvent B:** CH_3CN; flow rate: 5 mL min^{-1}; 0-4 min, 10% B; 4-22 min, 10-100% B; 22-27 min, 100% B; 27-29 min, 100%-10% B; 29-35 min, 10 % B.

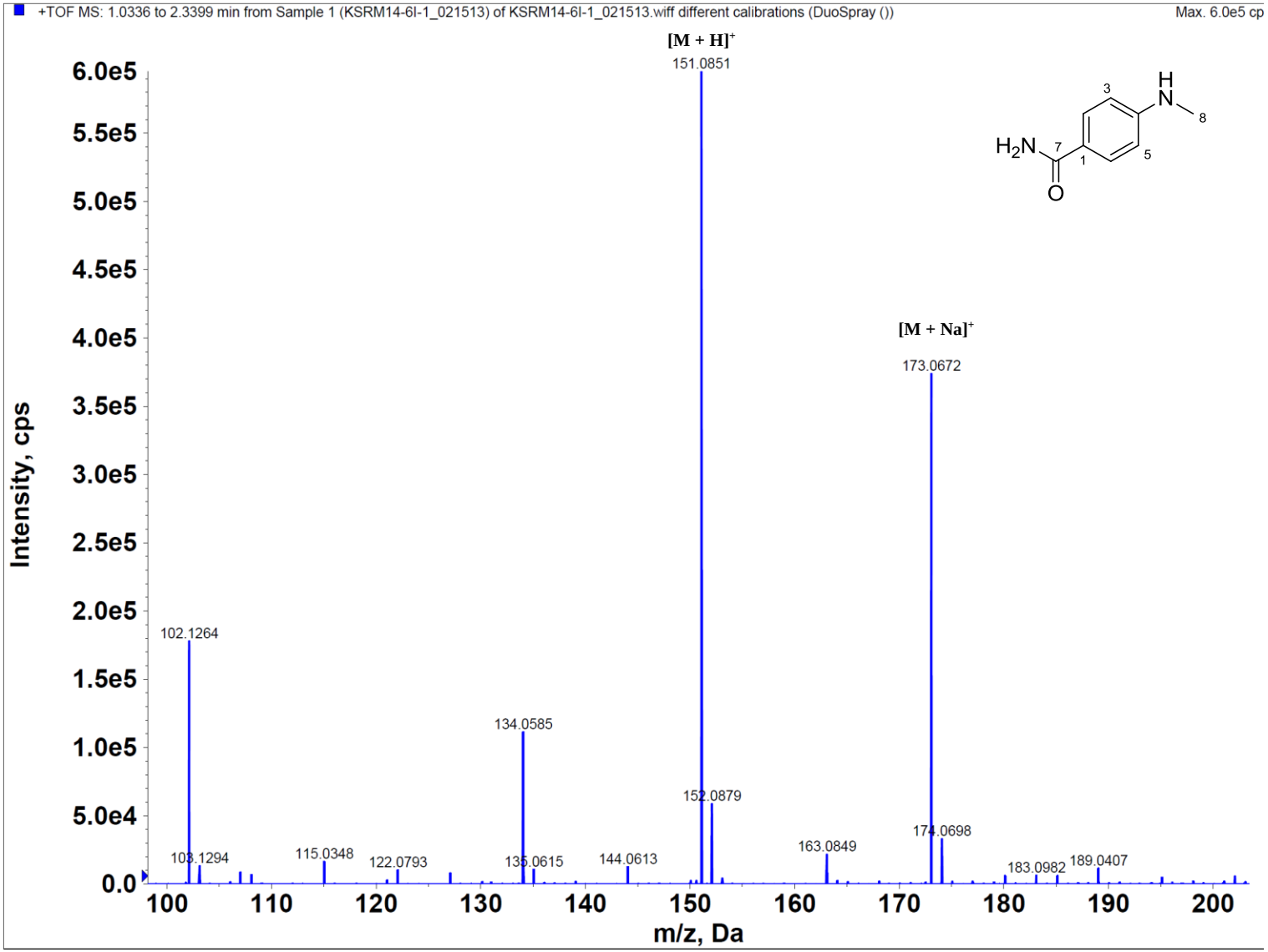


Figure S67. (+)-HRESI-MS spectrum of 4-(methylamino)benzamide (**6**)

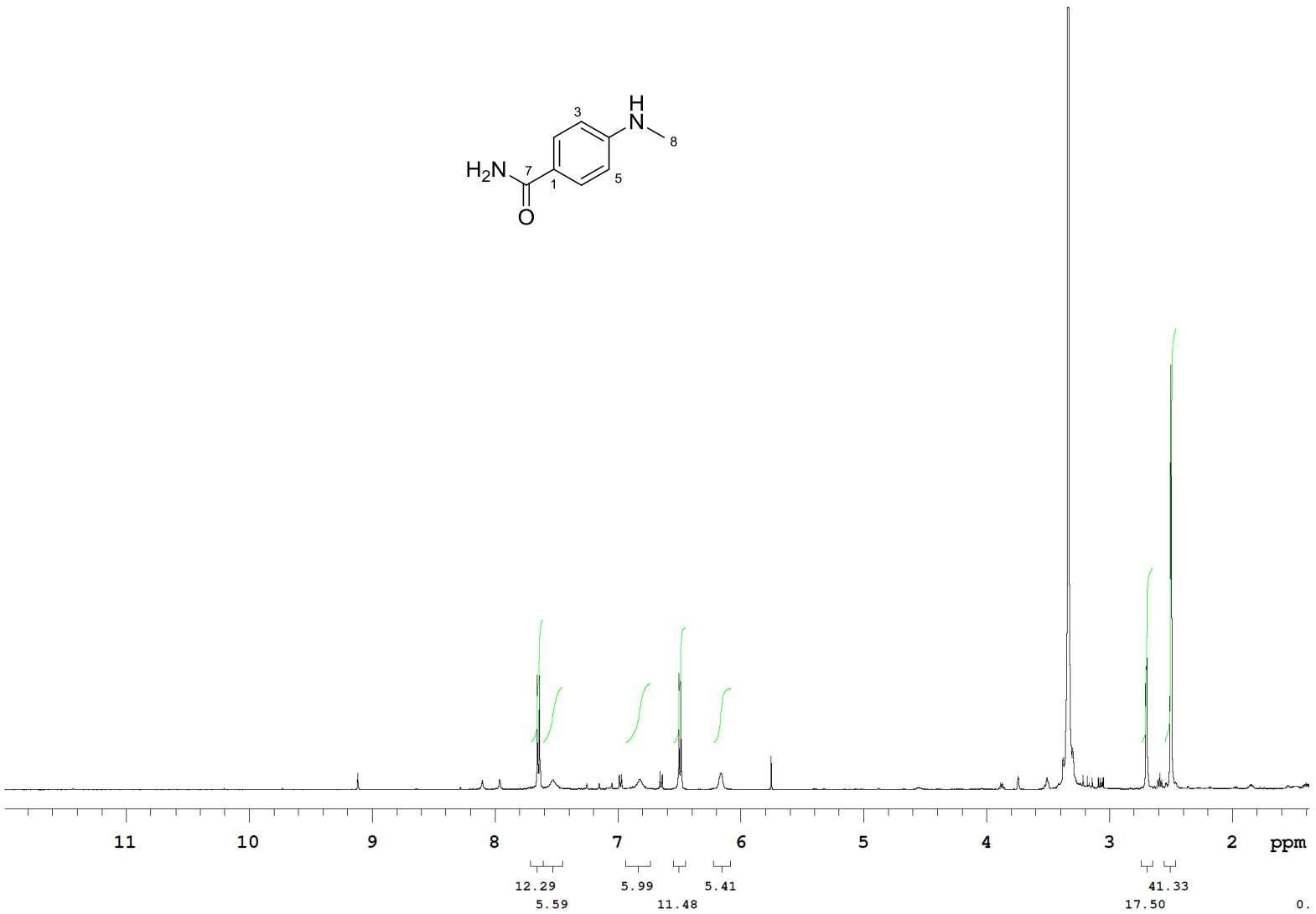


Figure S68. ^{1}H NMR spectrum (DMSO-d_6, 500 MHz) of 4-(methylamino)benzamide (**6**)

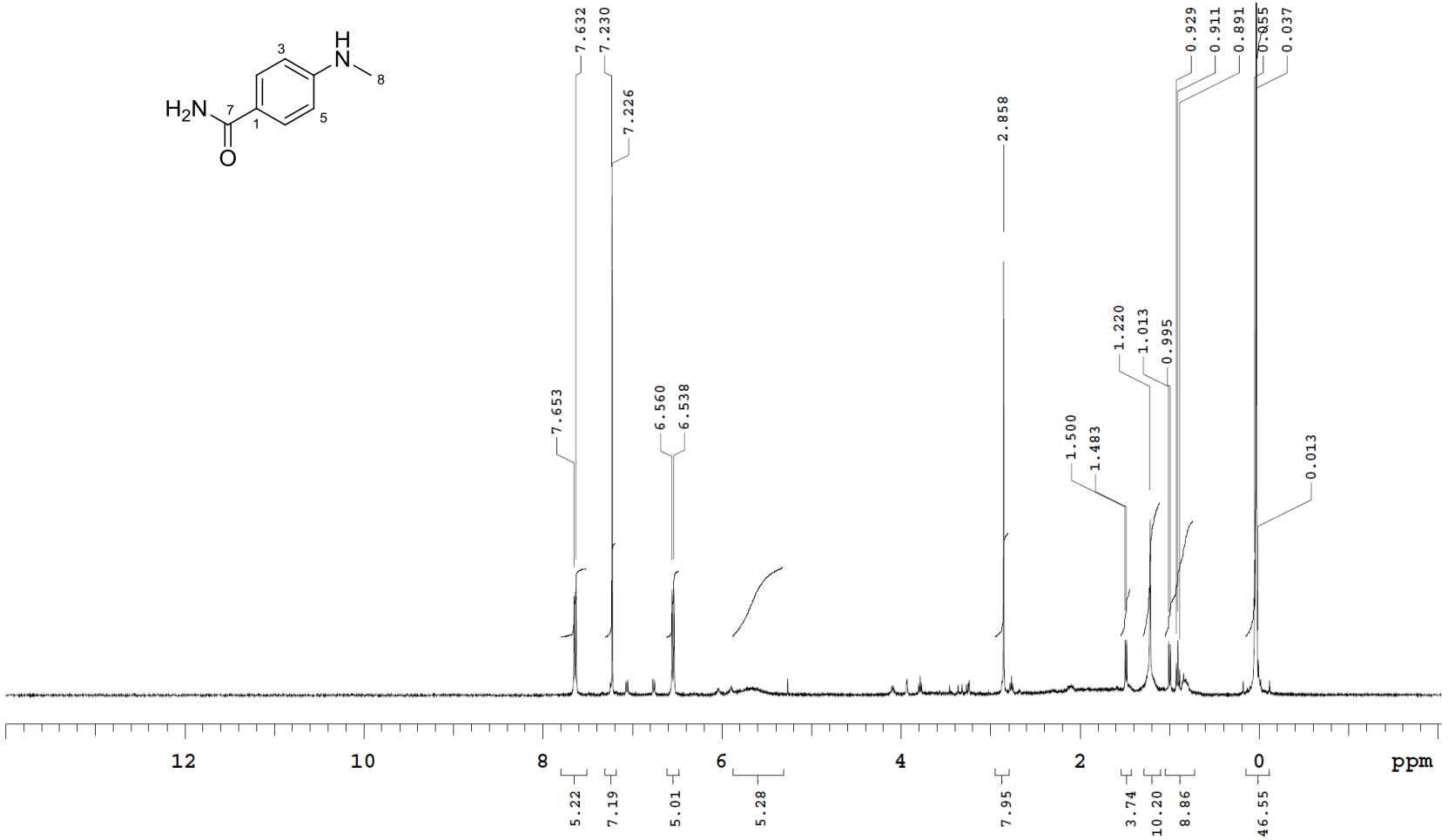


Figure S69. ^{1}H NMR spectrum ($CDCl_3$, 400 MHz) of 4-(methylamino)benzamide (**6**)

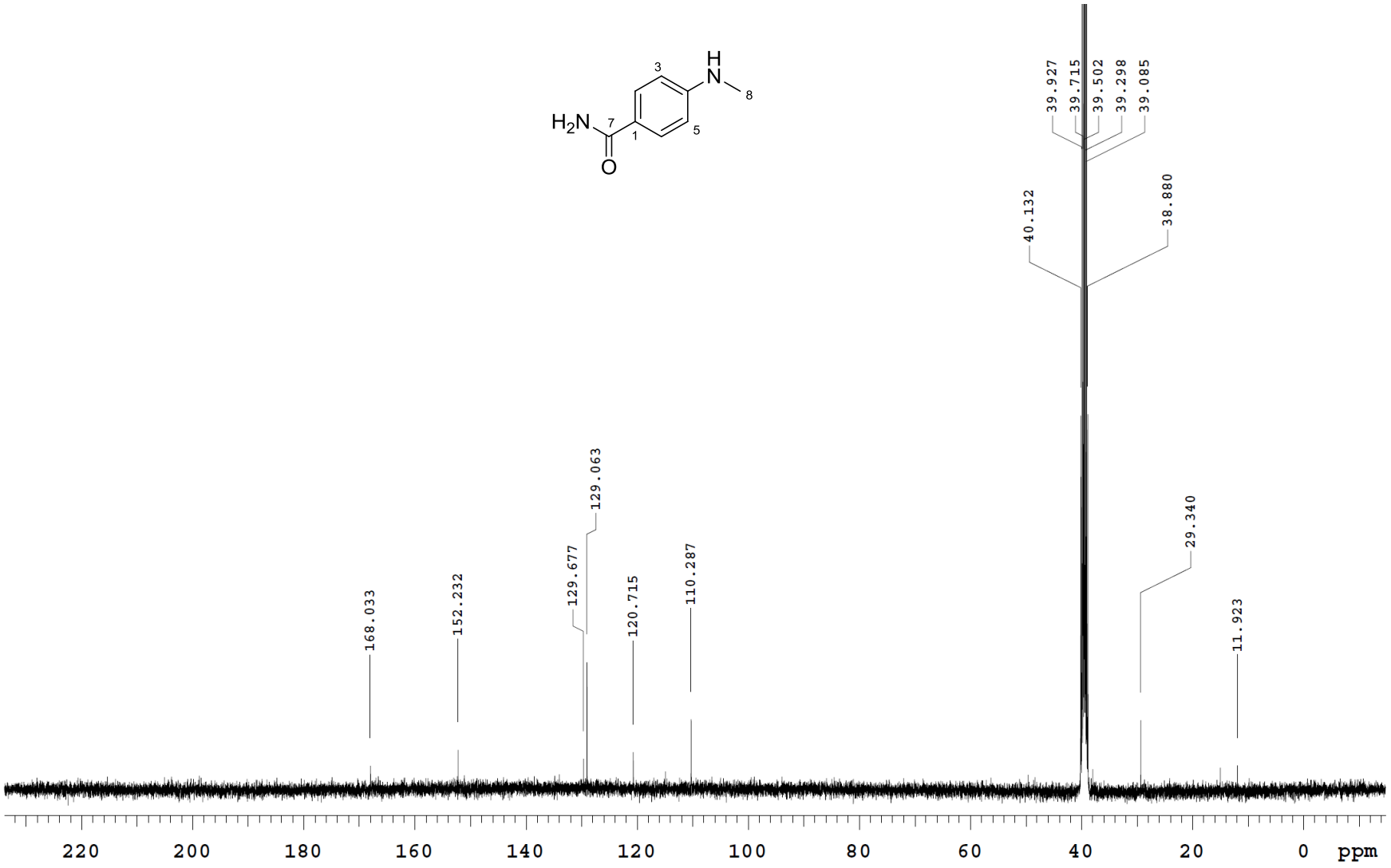
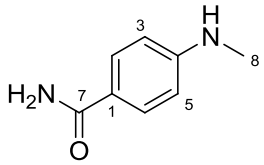


Figure S70. ^{13}C NMR spectrum (DMSO-d_6, 100 MHz) of 4-(methylamino)benzamide (**6**)



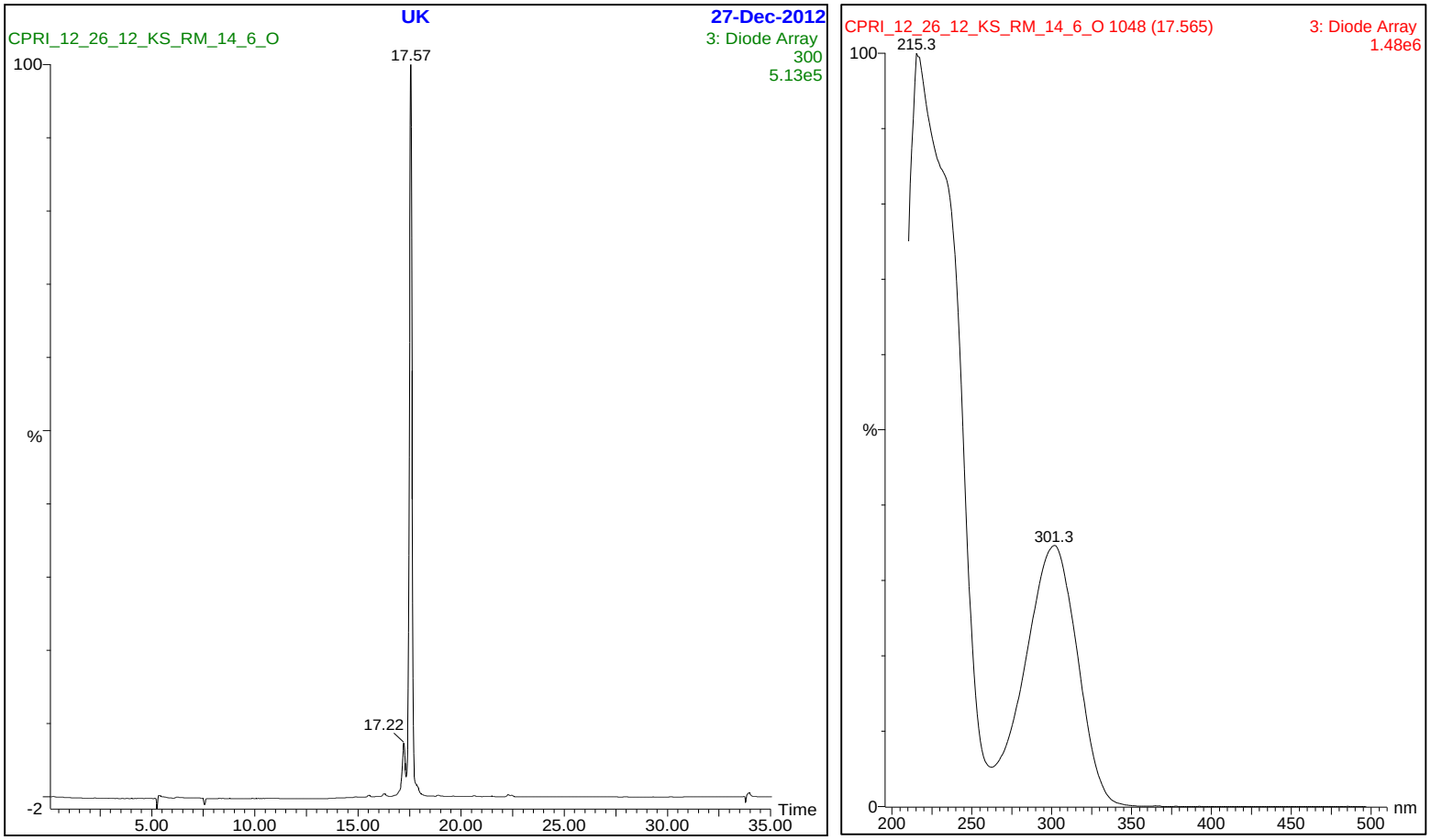


Figure S71. HPLC/UV analyses of the purified 2-hydroxy-benzamide (**7**). Detection wavelength: 300 nm; **solvent A:** H_2O/0.1% Formic acid; **solvent B:** CH_3CN/0.1% Formic acid; flow rate: 0.5 mL min^{-1}; 0-4 min, 10% B; 4-22 min, 10-100% B; 22-27 min, 100% B; 27-29 min, 100%-10% B; 29-35 min, 10 % B.

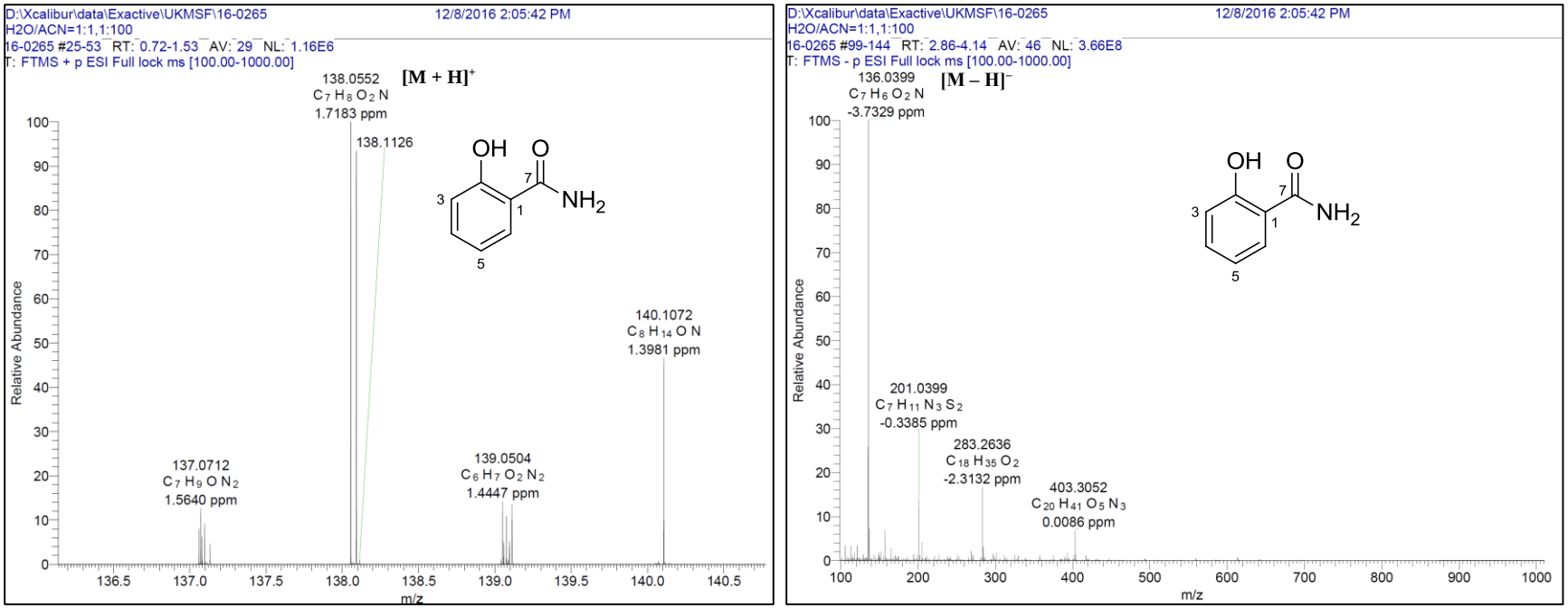


Figure S72. (+)- and (–)-HRESI-MS spectra of 2-hydroxy-benzamide (**7**)

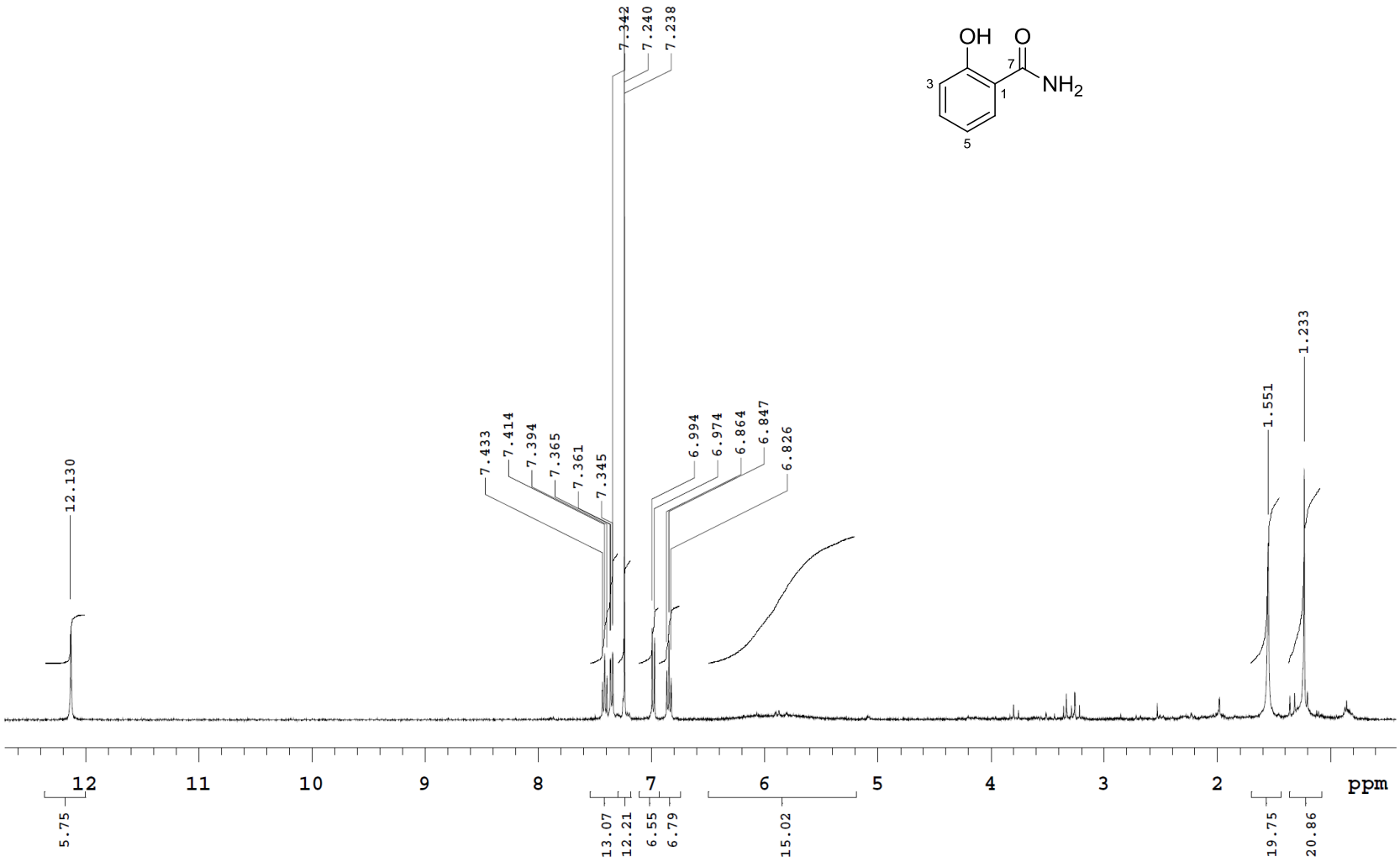


Figure S73. ^{1}H NMR spectrum ($CDCl_3$, 400 MHz) of 2-hydroxy-benzamide (**7**)

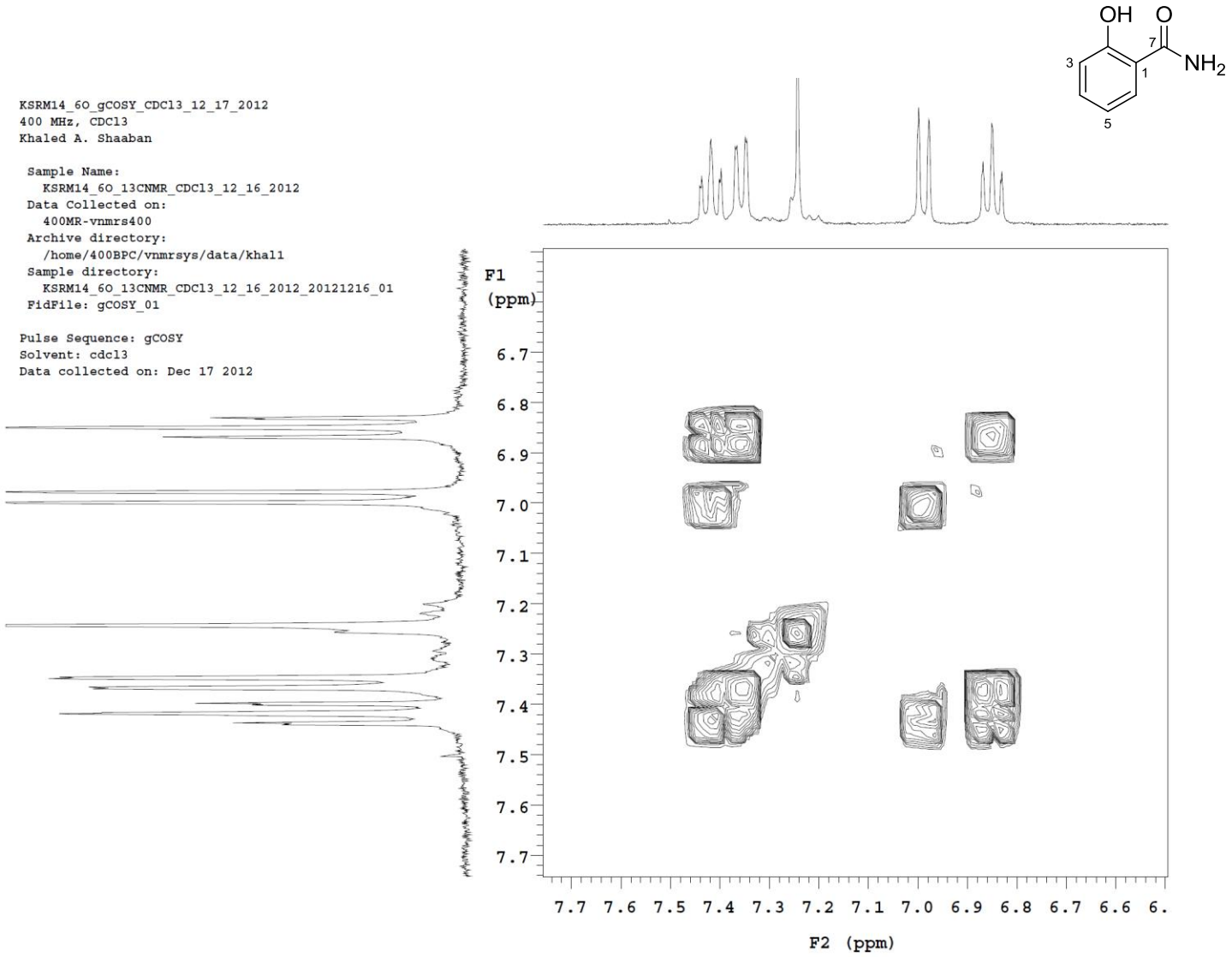


Figure S74. $^{1}H,^{1}H$-COSY spectrum ($CDCl_3$, 400 MHz) of 2-hydroxy-benzamide (**7**)

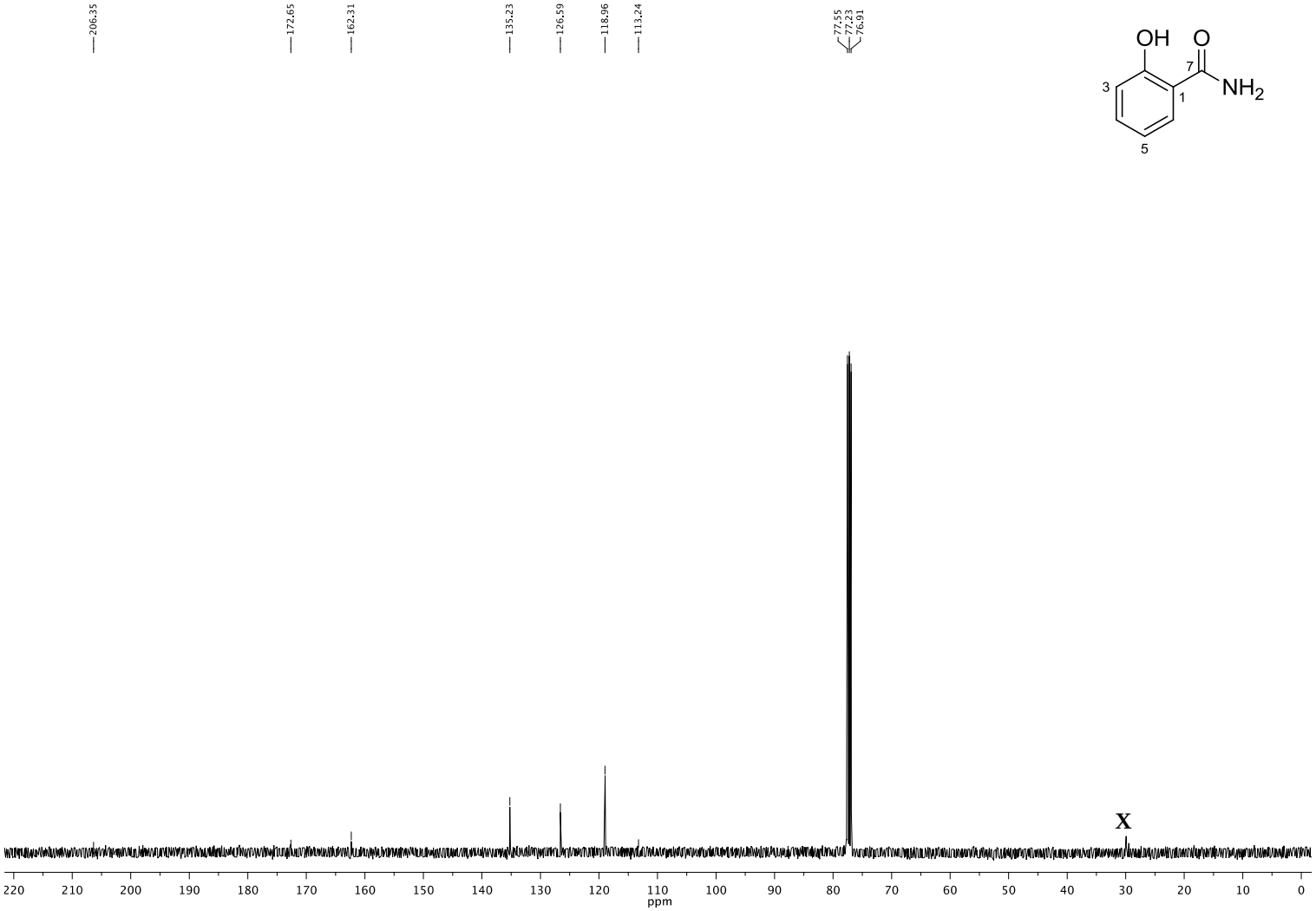


Figure S75. ^{13}C NMR spectrum ($CDCl_3$, 100 MHz) of 2-hydroxy-benzamide (**7**)

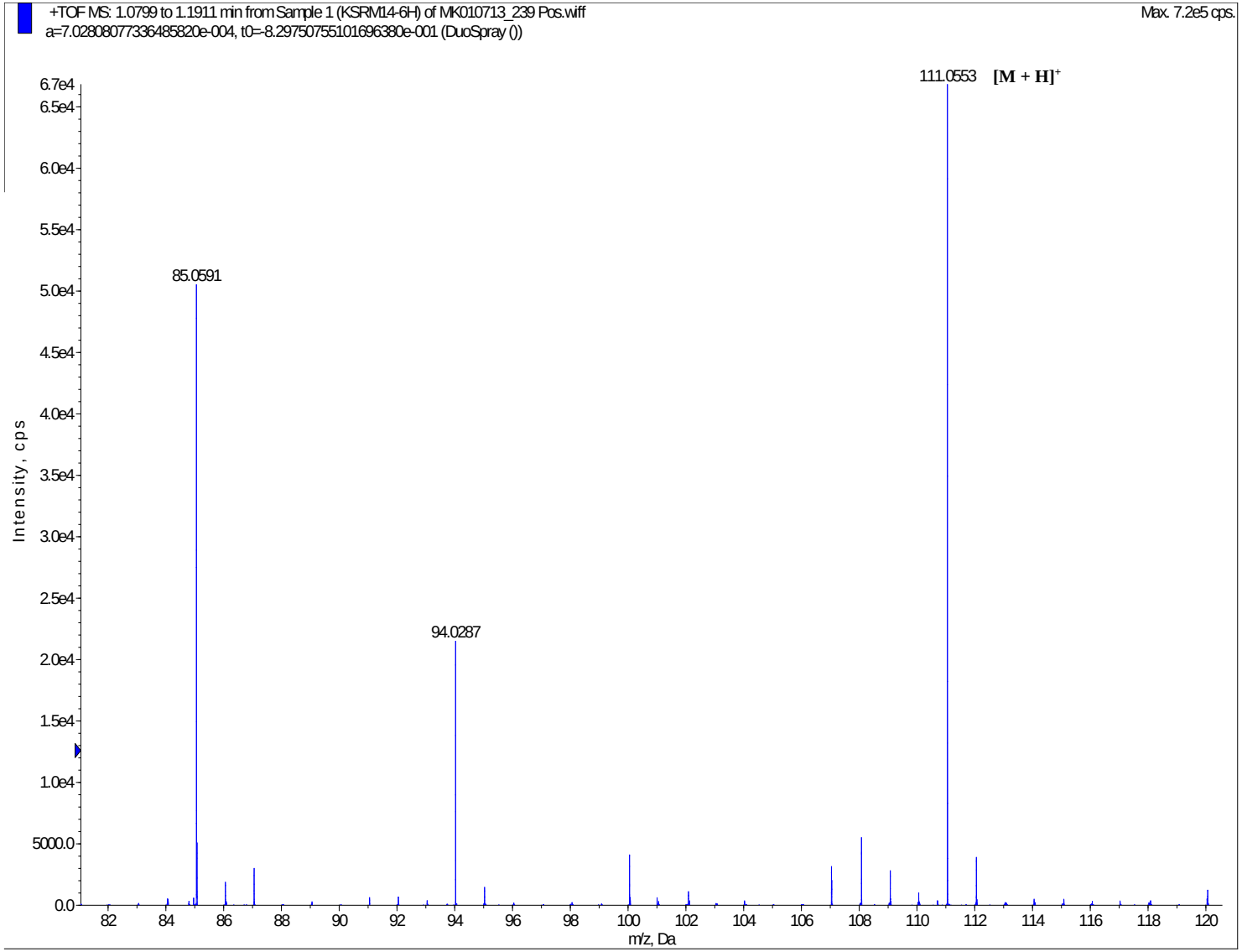


Figure S76. (+)-HRESI-MS spectrum of pyrrole-2-carboxamide (**8**)

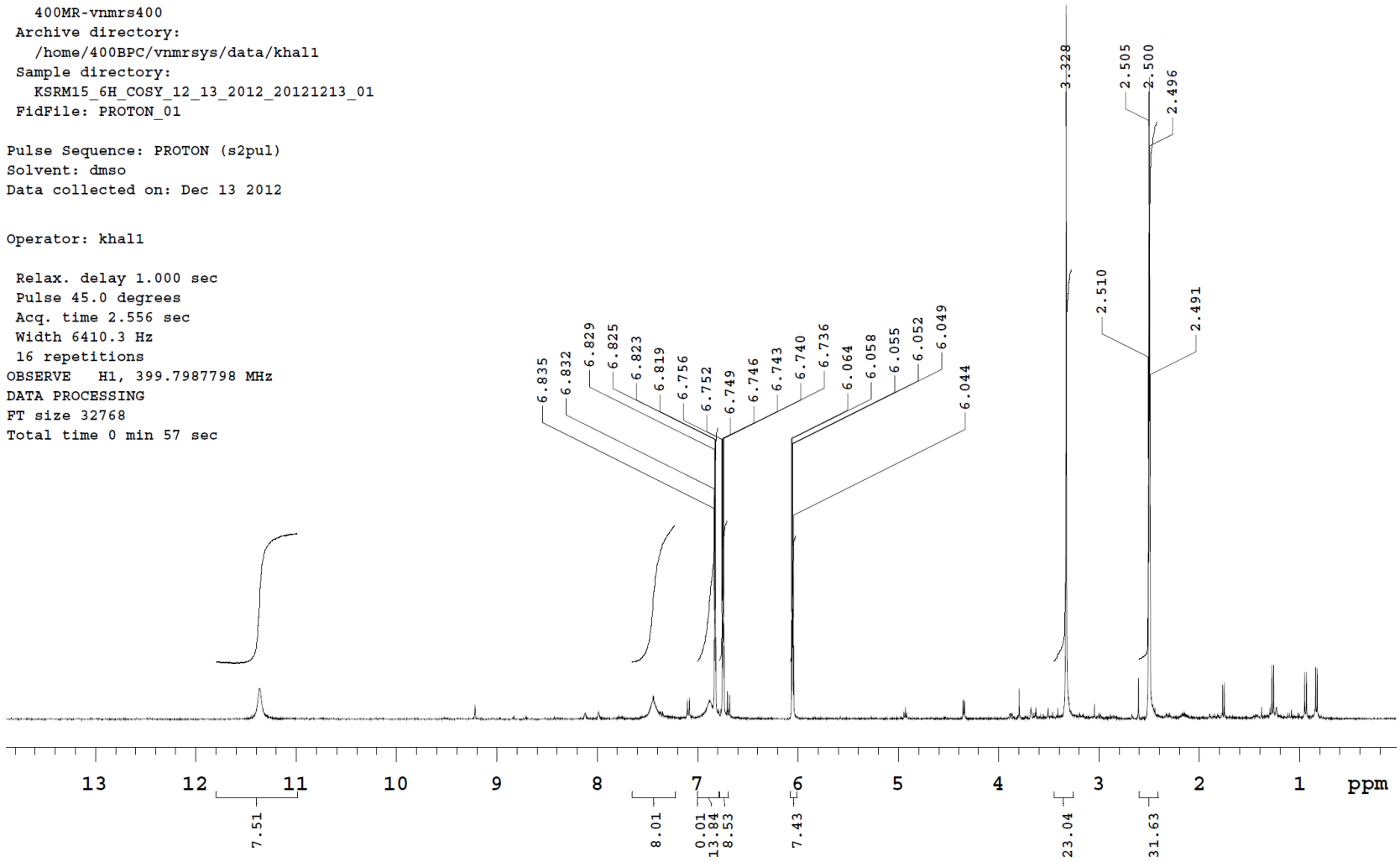


Figure S77. ^{1}H NMR spectrum (DMSO-d_6, 400 MHz) of pyrrole-2-carboxamide (**8**)

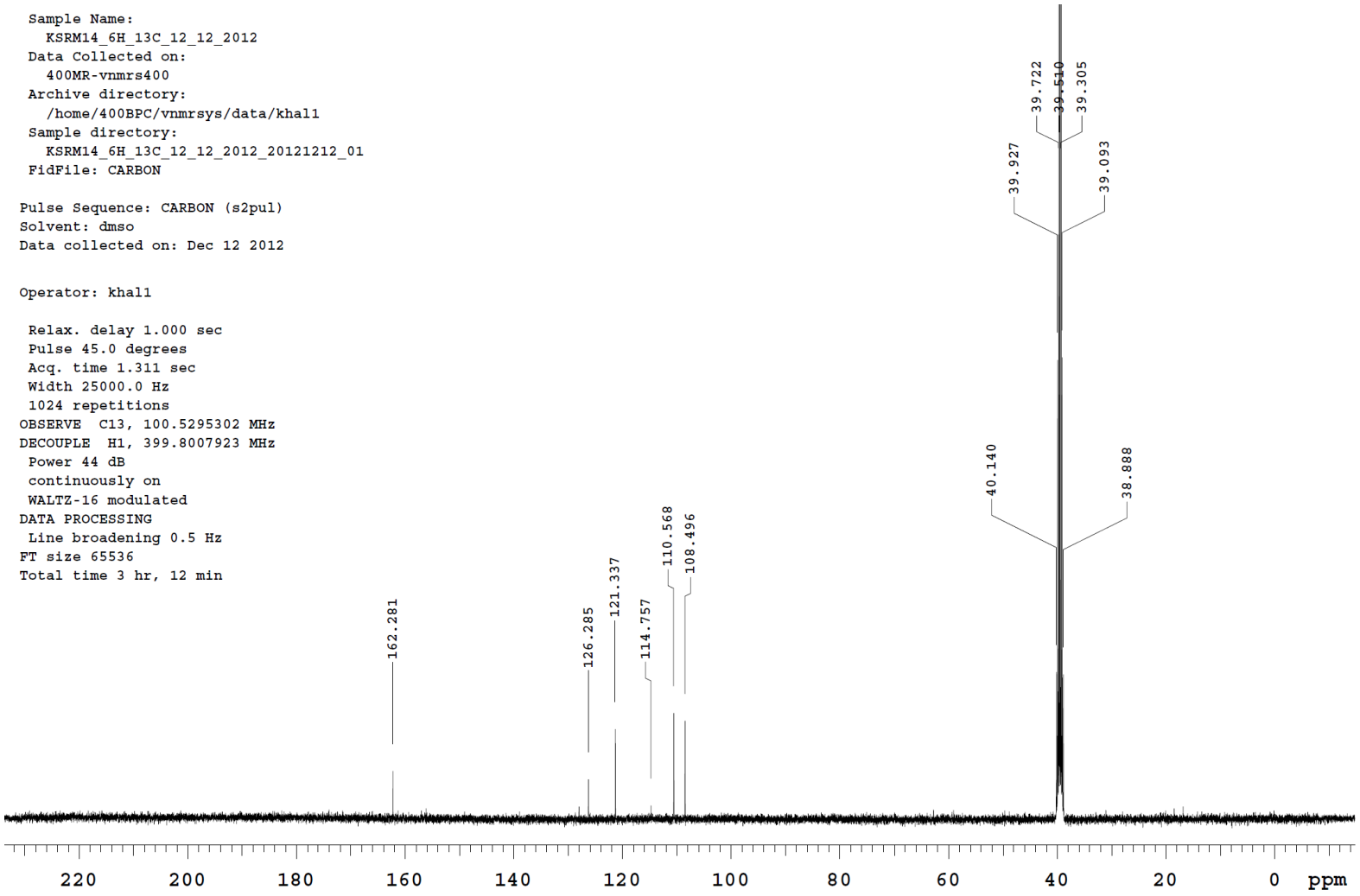


Figure S78. ^{13}C NMR spectrum (DMSO-d_6, 100 MHz) of pyrrole-2-carboxamide (**8**)

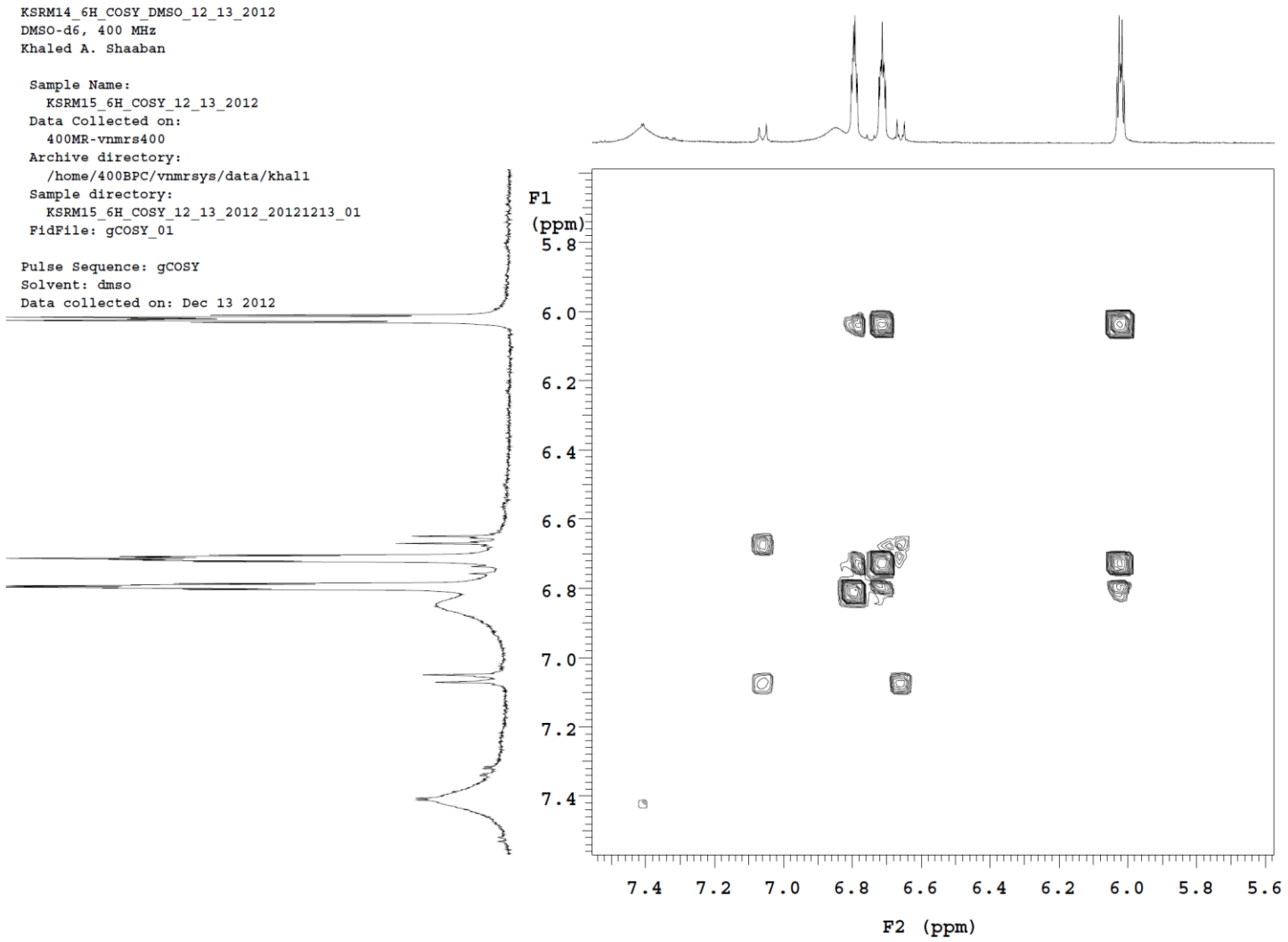
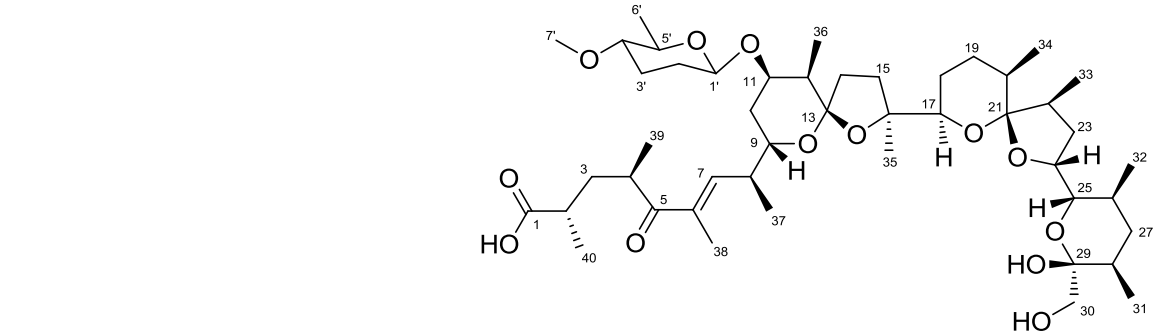
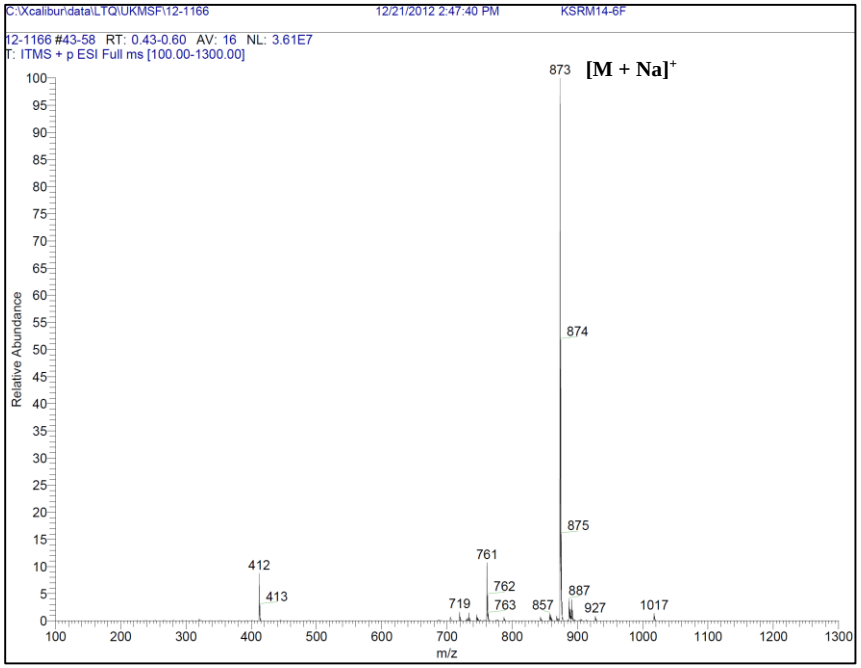


Figure S79. ^{1}H,^{1}H-COSY spectrum (DMSO-d_6, 100 MHz) of pyrrole-2-carboxamide (**8**)





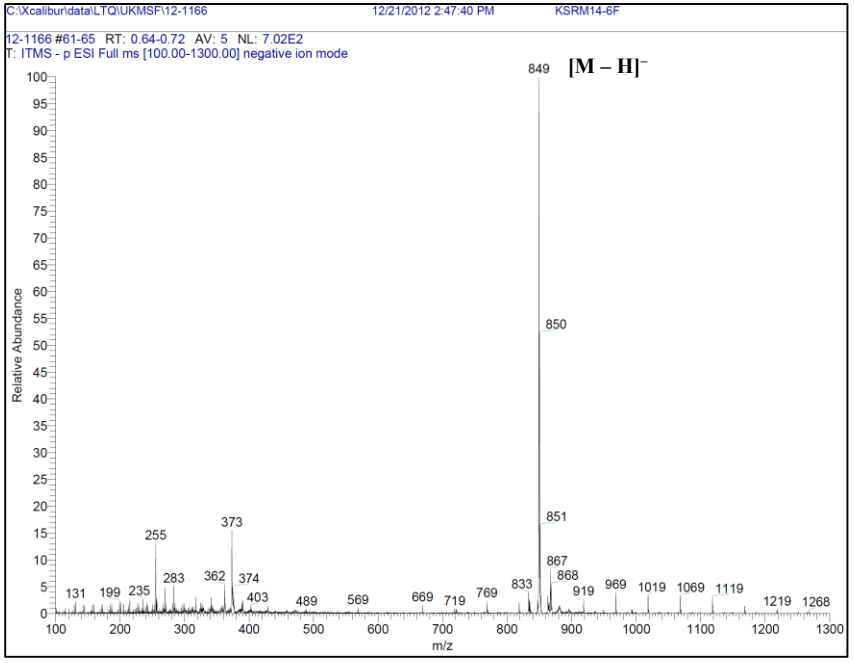


Figure S80. (+) and (–)-ESI-MS spectra of lenoremycin (**9**)

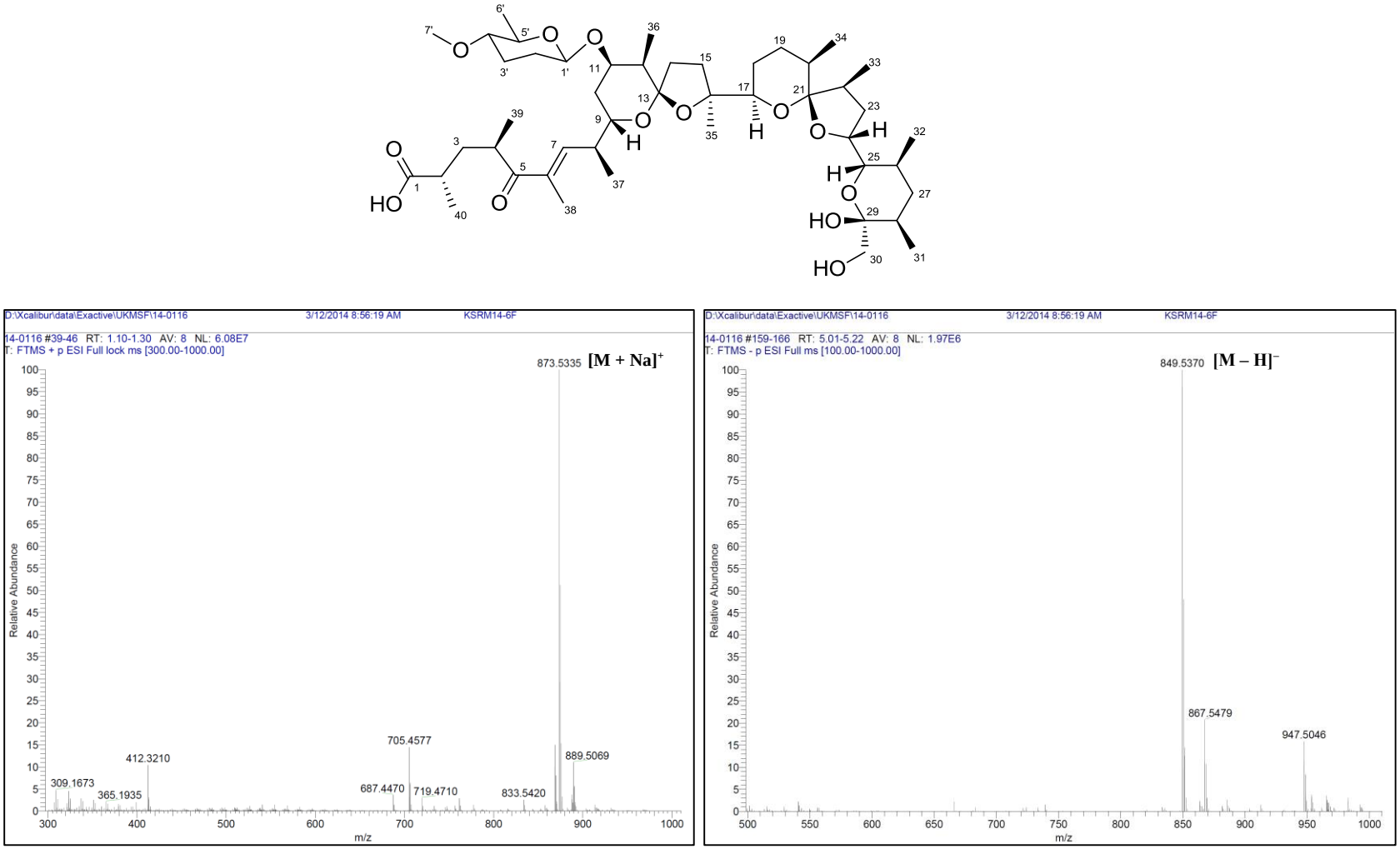


Figure S81. (+) and (–)-HRESI-MS spectra of lenoremycin (**9**)

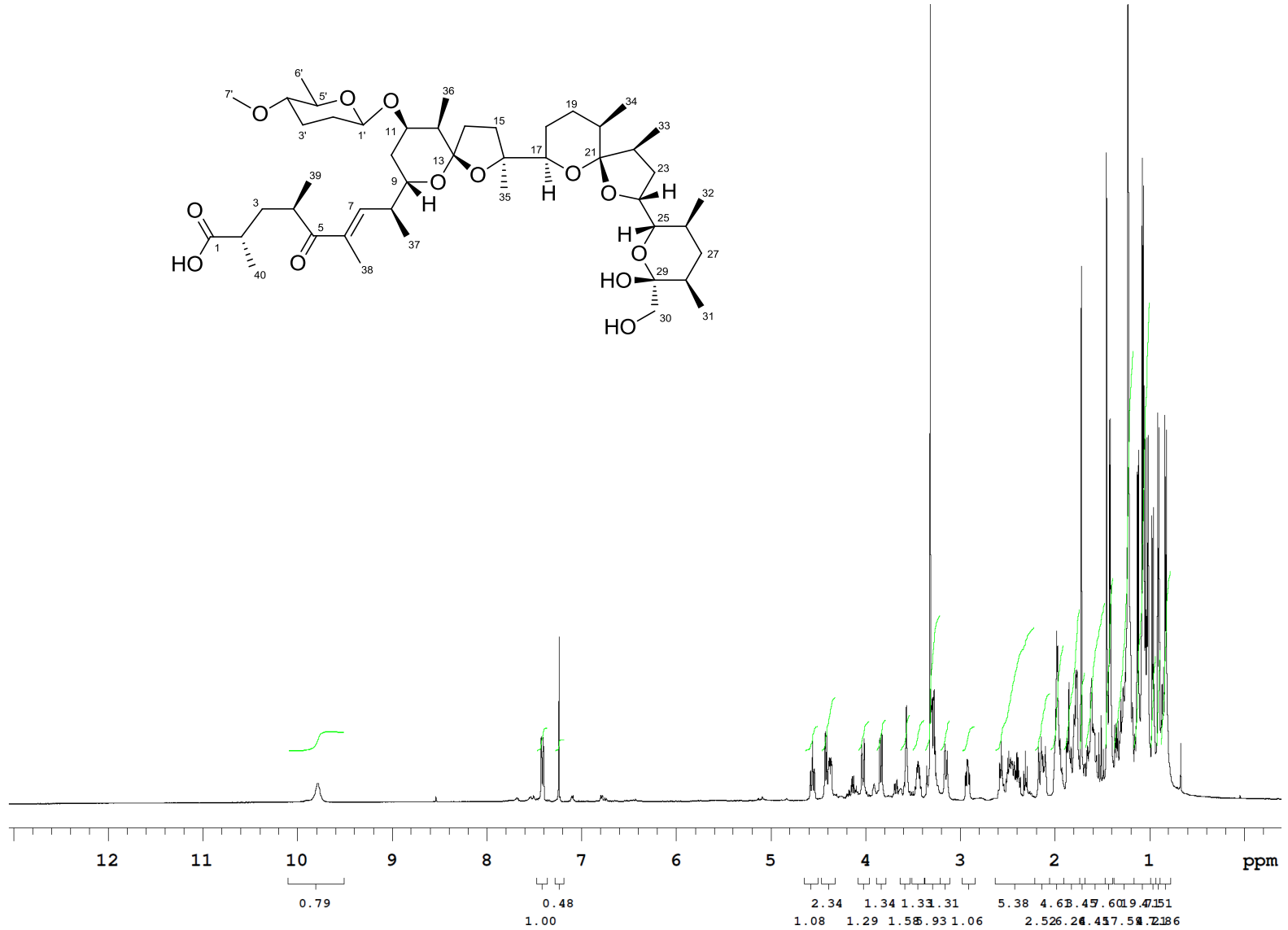


Figure S82. ^{1}H NMR spectrum ($CDCl_3$, 500 MHz) of lenoremycin (**9**)

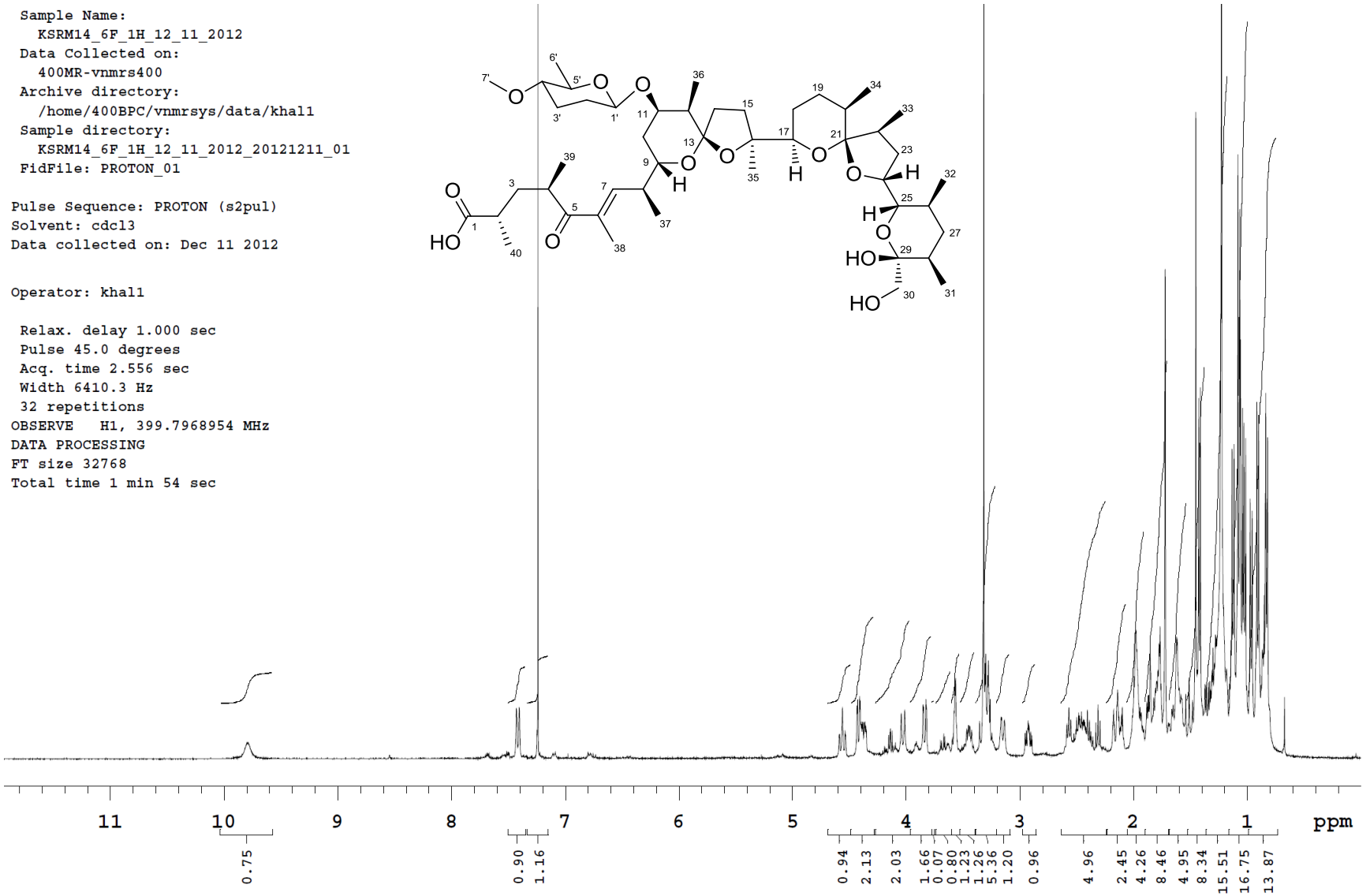
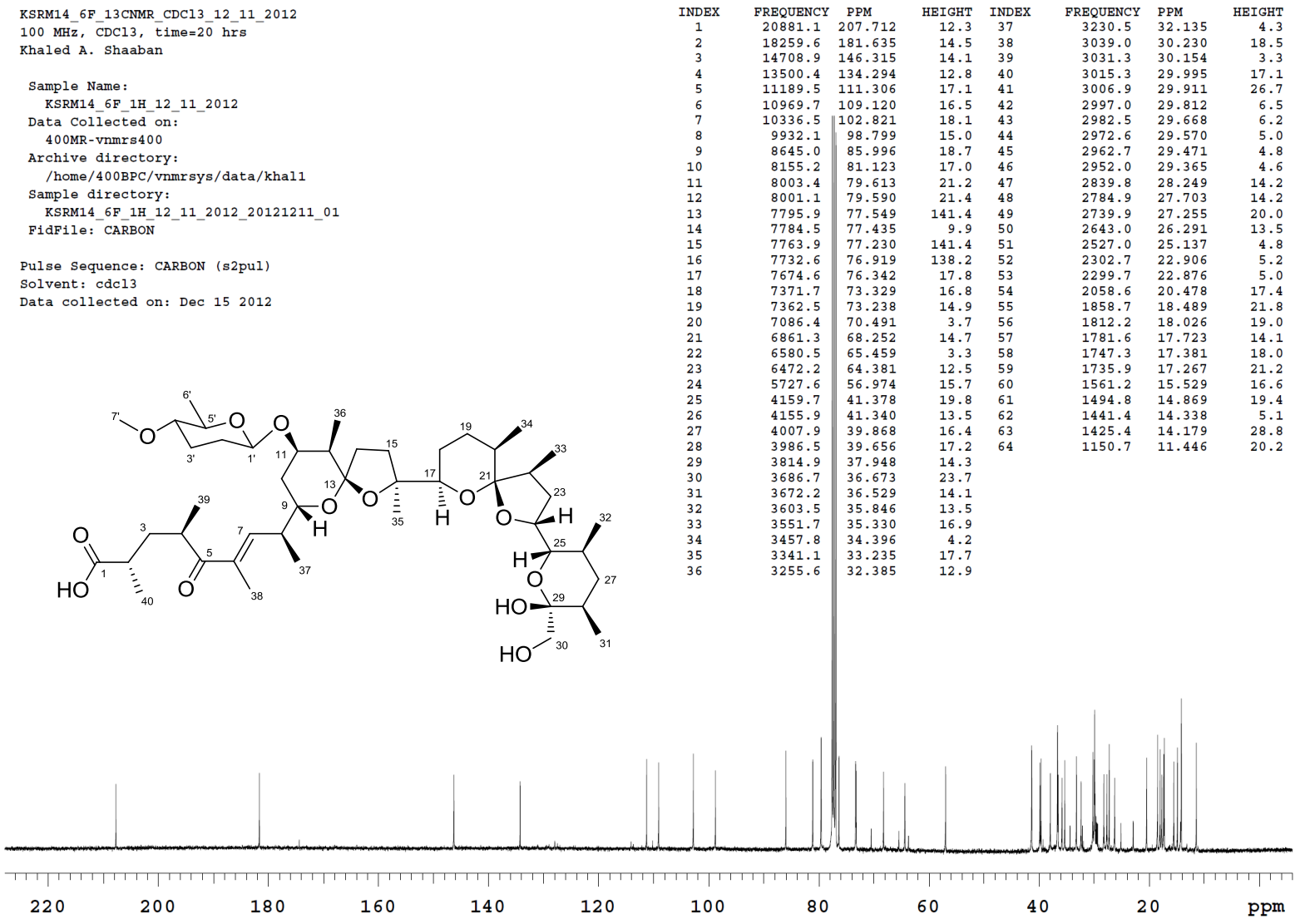


Figure S83. ^{1}H NMR spectrum ($CDCl_3$, 400 MHz) of lenoremycin (**9**)



INDEX	FREQUENCY	PPM	HEIGHT	INDEX	FREQUENCY	PPM	HEIGHT
1	20881.1	207.712	12.3	37	3230.5	32.135	4.3
2	18259.6	181.635	14.5	38	3039.0	30.230	18.5
3	14708.9	146.315	14.1	39	3031.3	30.154	3.3
4	13500.4	134.294	12.8	40	3015.3	29.995	17.1
5	11189.5	111.306	17.1	41	3006.9	29.911	26.7
6	10969.7	109.120	16.5	42	2997.0	29.812	6.5
7	10336.5	102.821	18.1	43	2982.5	29.668	6.2
8	9932.1	98.799	15.0	44	2972.6	29.570	5.0
9	8645.0	85.996	18.7	45	2962.7	29.471	4.8
10	8155.2	81.123	17.0	46	2952.0	29.365	4.6
11	8003.4	79.613	21.2	47	2839.8	28.249	14.2
12	8001.1	79.590	21.4	48	2784.9	27.703	14.2
13	7795.9	77.549	141.4	49	2739.9	27.255	20.0
14	7784.5	77.435	9.9	50	2643.0	26.291	13.5
15	7763.9	77.230	141.4	51	2527.0	25.137	4.8
16	7732.6	76.919	138.2	52	2302.7	22.906	5.2
17	7674.6	76.342	17.8	53	2299.7	22.876	5.0
18	7371.7	73.329	16.8	54	2058.6	20.478	17.4
19	7362.5	73.238	14.9	55	1858.7	18.489	21.8
20	7086.4	70.491	3.7	56	1812.2	18.026	19.0
21	6861.3	68.252	14.7	57	1781.6	17.723	14.1
22	6580.5	65.459	3.3	58	1747.3	17.381	18.0
23	6472.2	64.381	12.5	59	1735.9	17.267	21.2
24	5727.6	56.974	15.7	60	1561.2	15.529	16.6
25	4159.7	41.378	19.8	61	1494.8	14.869	19.4
26	4155.9	41.340	13.5	62	1441.4	14.338	5.1
27	4007.9	39.868	16.4	63	1425.4	14.179	28.8
28	3986.5	39.656	17.2	64	1150.7	11.446	20.2
29	3814.9	37.948	14.3				
30	3686.7	36.673	23.7				
31	3672.2	36.529	14.1				
32	3603.5	35.846	13.5				
33	3551.7	35.330	16.9				
34	3457.8	34.396	4.2				
35	3341.1	33.235	17.7				
36	3255.6	32.385	12.9				

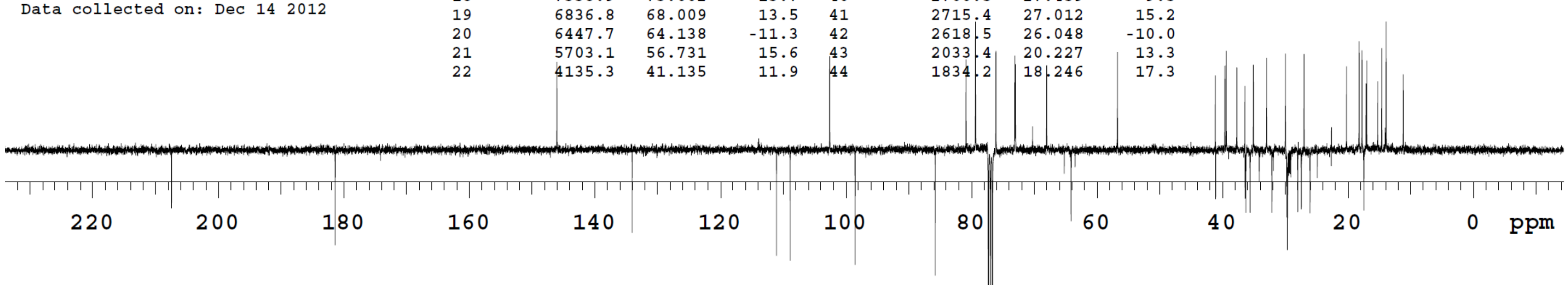
Figure S84. ^{13}C NMR spectrum ($CDCl_3$, 100 MHz) of lenoremycin (**9**)

KSRM14_6F_APT_CDCl3_12_12_2012
100 MHz, CDCl3, time=6 hrs
Khaled A. Shaaban

Sample Name:
KSRM14_6F_1H_12_11_2012
Data Collected on:
400MR-vnmrs400
Archive directory:
/home/400BPC/vnmrsys/data/khal1
Sample directory:
KSRM14_6F_1H_12_11_2012_20121211_01
FidFile: APT_01

Pulse Sequence: APT
Solvent: cdcl3
Data collected on: Dec 14 2012

INDEX	FREQUENCY	PPM	HEIGHT	INDEX	FREQUENCY	PPM	HEIGHT	INDEX	FREQUENCY	PPM	HEIGHT
1	20856.6	207.469	-9.3	23	4131.5	41.097	-7.8	45	1787.7	17.783	15.9
2	18235.2	181.392	-15.1	24	3983.4	39.625	13.4	46	1757.2	17.479	-9.6
3	14683.7	146.064	14.0	25	3962.1	39.412	15.8	47	1722.9	17.138	10.6
4	13475.9	134.050	-13.2	26	3791.2	37.712	13.2	48	1710.6	17.016	14.3
5	11165.0	111.062	-16.8	27	3662.2	36.430	10.2	49	1536.7	15.286	10.9
6	10945.3	108.877	-17.5	28	3660.0	36.407	-7.7	50	1469.6	14.618	16.2
7	10312.0	102.578	15.0	29	3647.8	36.286	-10.0	51	1400.9	13.935	20.4
8	9907.7	98.555	-18.3	30	3579.1	35.603	-9.9	52	1126.2	11.203	12.1
9	8620.6	85.752	-19.9	31	3527.2	35.086	13.6				
10	8131.5	80.888	14.5	32	3433.4	34.153	-5.0				
11	7979.0	79.370	17.8	33	3316.6	32.992	14.7				
12	7976.7	79.347	20.5	34	3231.2	32.142	-9.9				
13	7771.4	77.305	-123.3	35	3014.5	29.986	15.4				
14	7739.4	76.987	-127.1	36	2990.9	29.751	-10.7				
15	7707.4	76.668	-121.7	37	2981.7	29.660	-15.9				
16	7650.1	76.099	15.8	38	2977.9	29.622	-7.2				
17	7347.2	73.086	15.1	39	2816.1	28.013	-9.9				
18	7338.9	73.002	13.7	40	2760.5	27.459	-9.5				
19	6836.8	68.009	13.5	41	2715.4	27.012	15.2				
20	6447.7	64.138	-11.3	42	2618.5	26.048	-10.0				
21	5703.1	56.731	15.6	43	2033.4	20.227	13.3				
22	4135.3	41.135	11.9	44	1834.2	18.246	17.3				



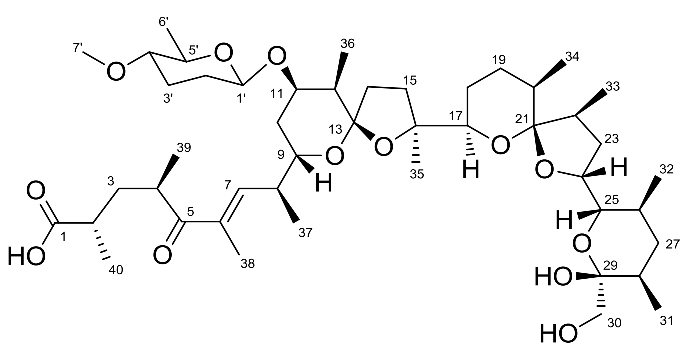


Figure S85. APT NMR spectrum ($CDCl_3$, 100 MHz) of lenoremycin (**9**)

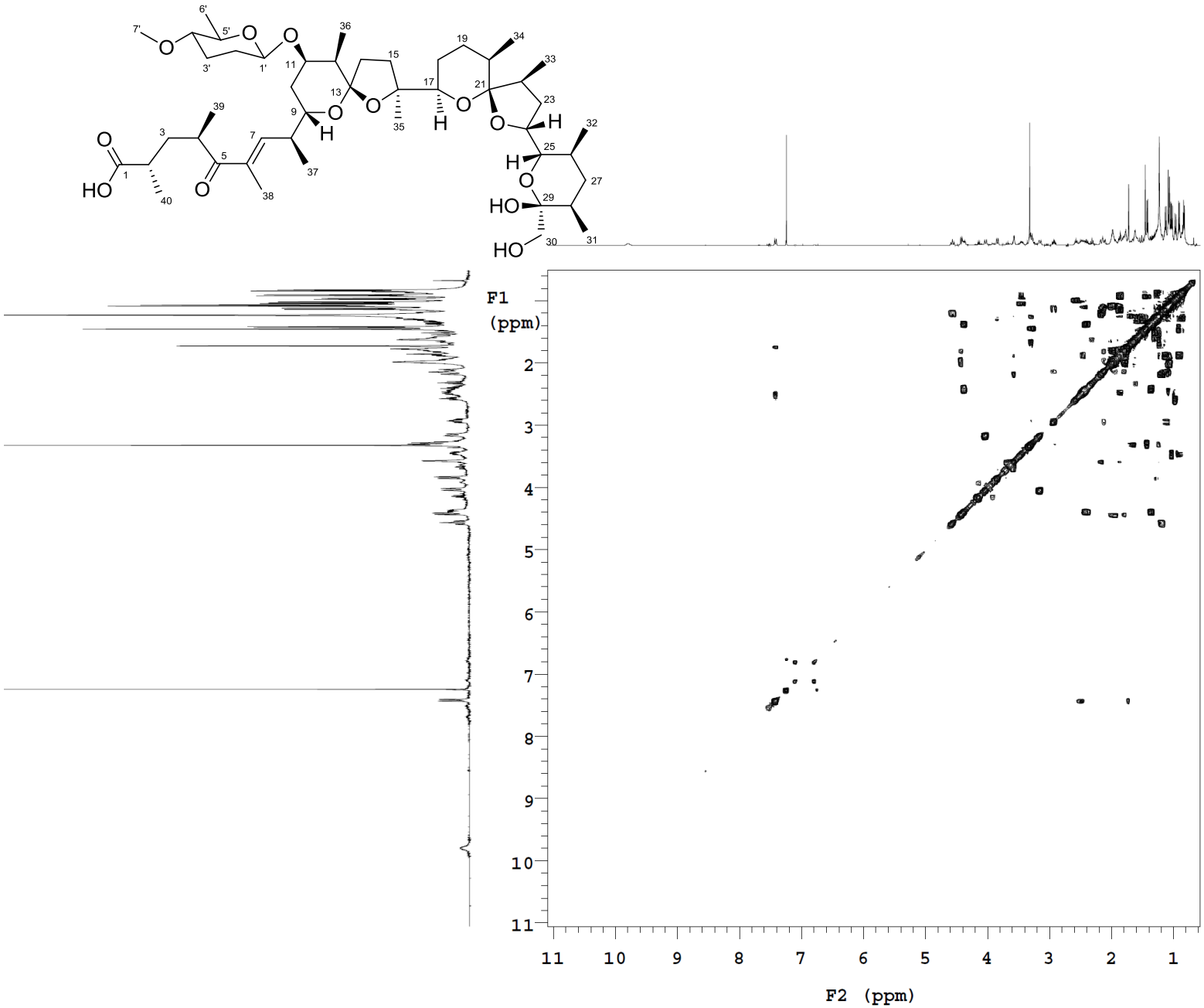


Figure S86. $^{1}H,^{1}H$-COSY spectrum ($CDCl_3$, 400 MHz) of lenoremycin (**9**)

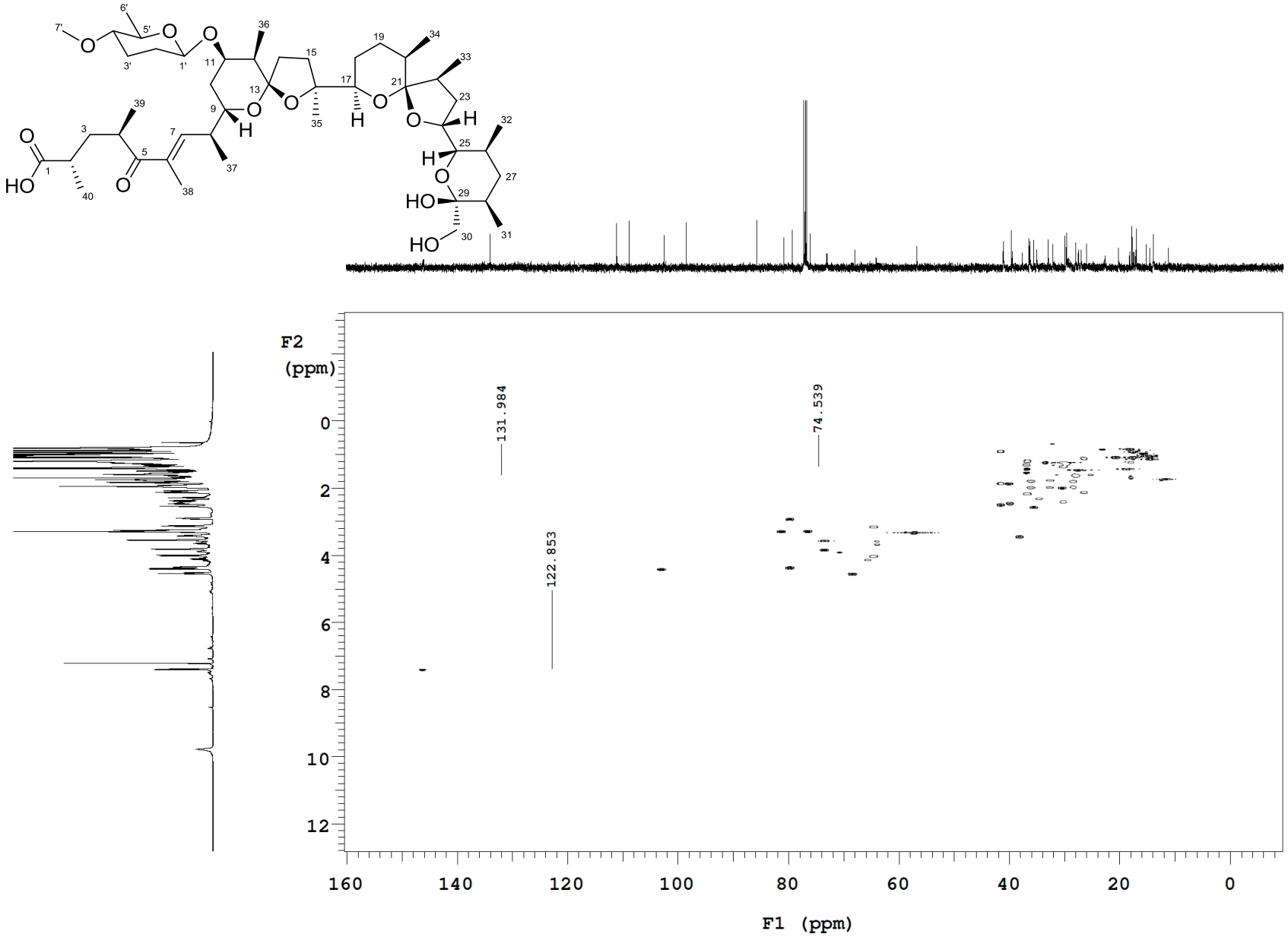


Figure S87. HSQC spectrum ($CDCl_3$, 500 MHz) of lenoremycin (**9**)

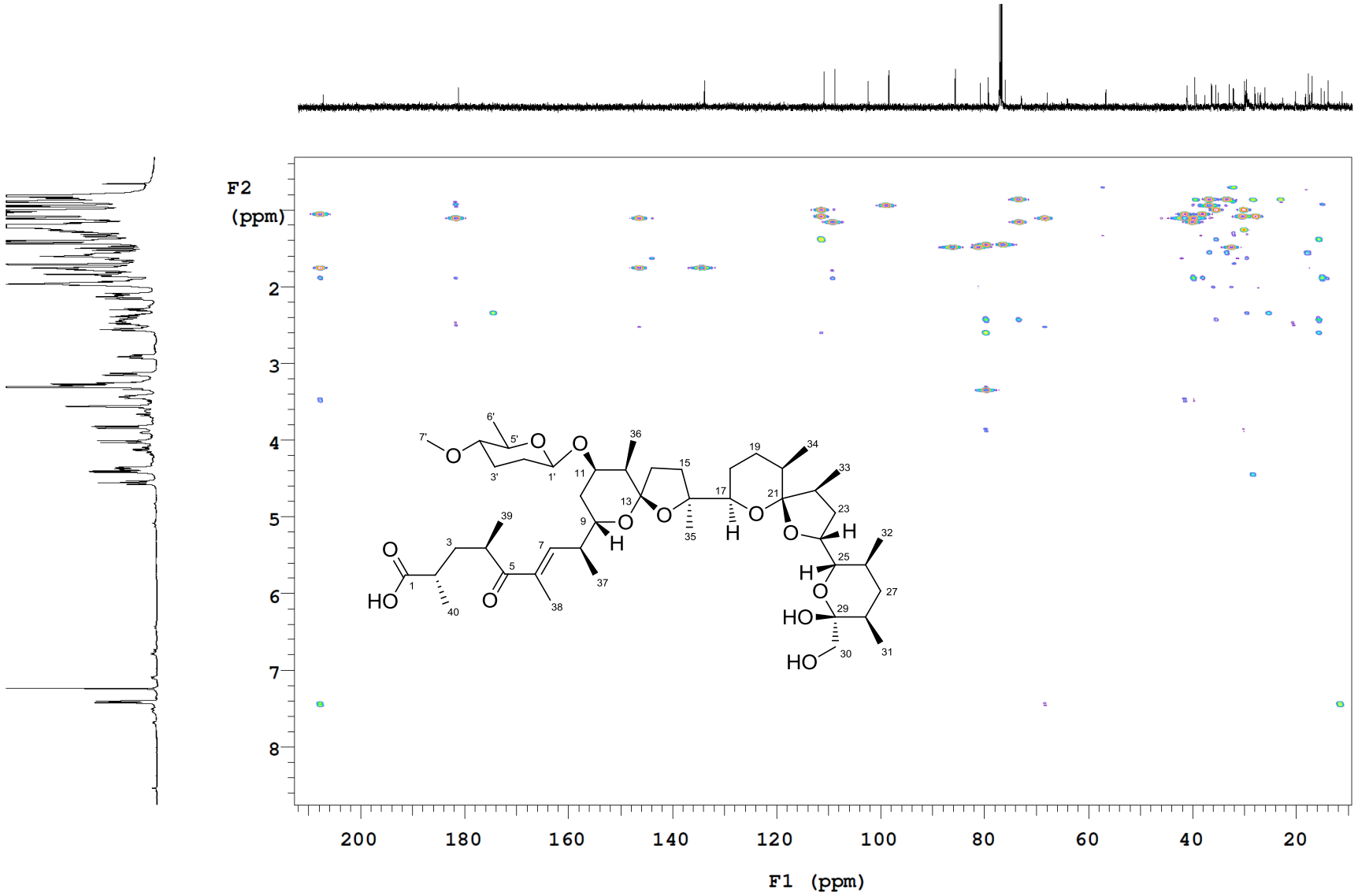


Figure S88. HMBC spectrum ($CDCl_3$, 500 MHz) of lenoremycin (**9**)

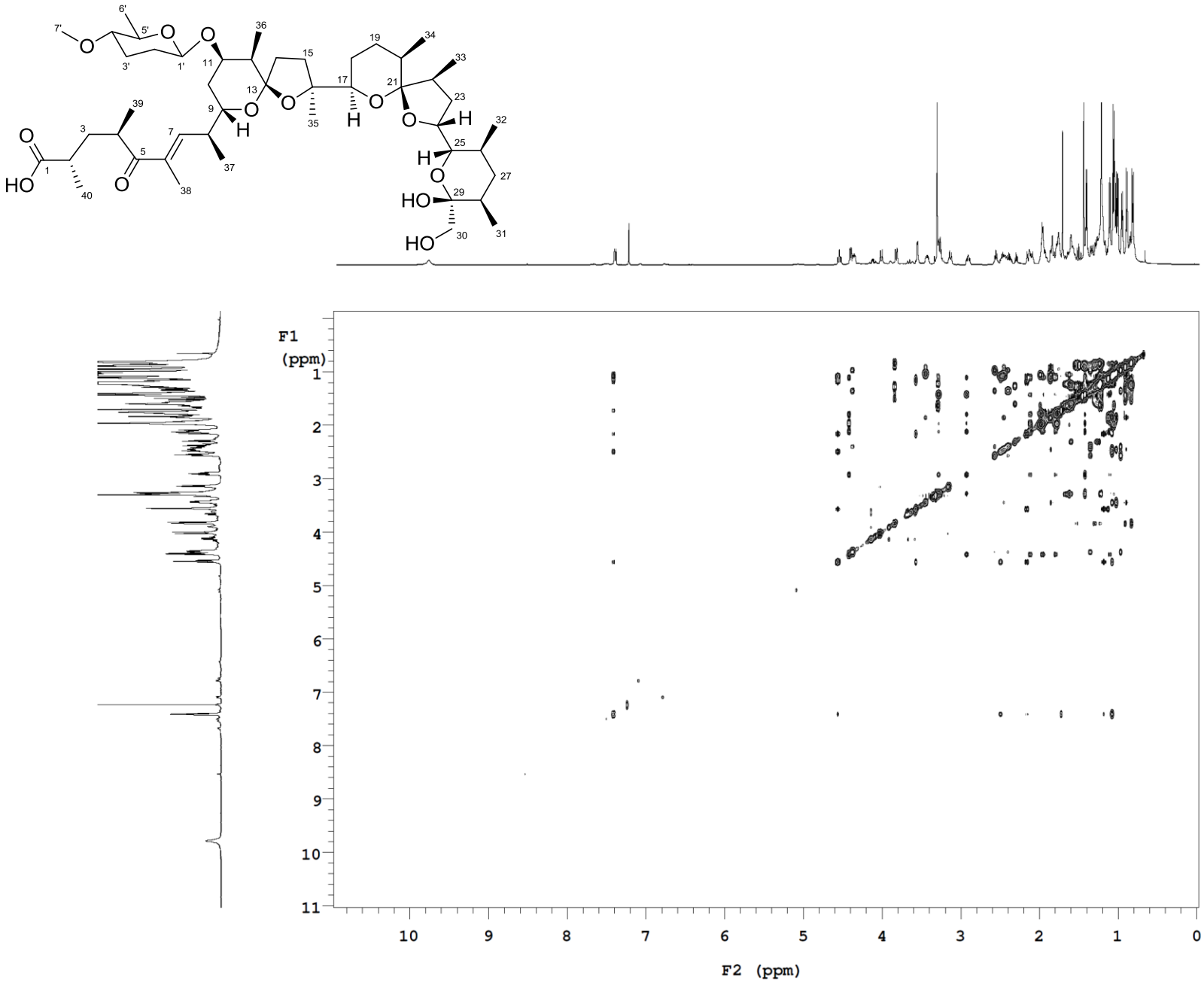


Figure S89. TOCSY spectrum ($CDCl_3$, 500 MHz) of lenoremycin (**9**)

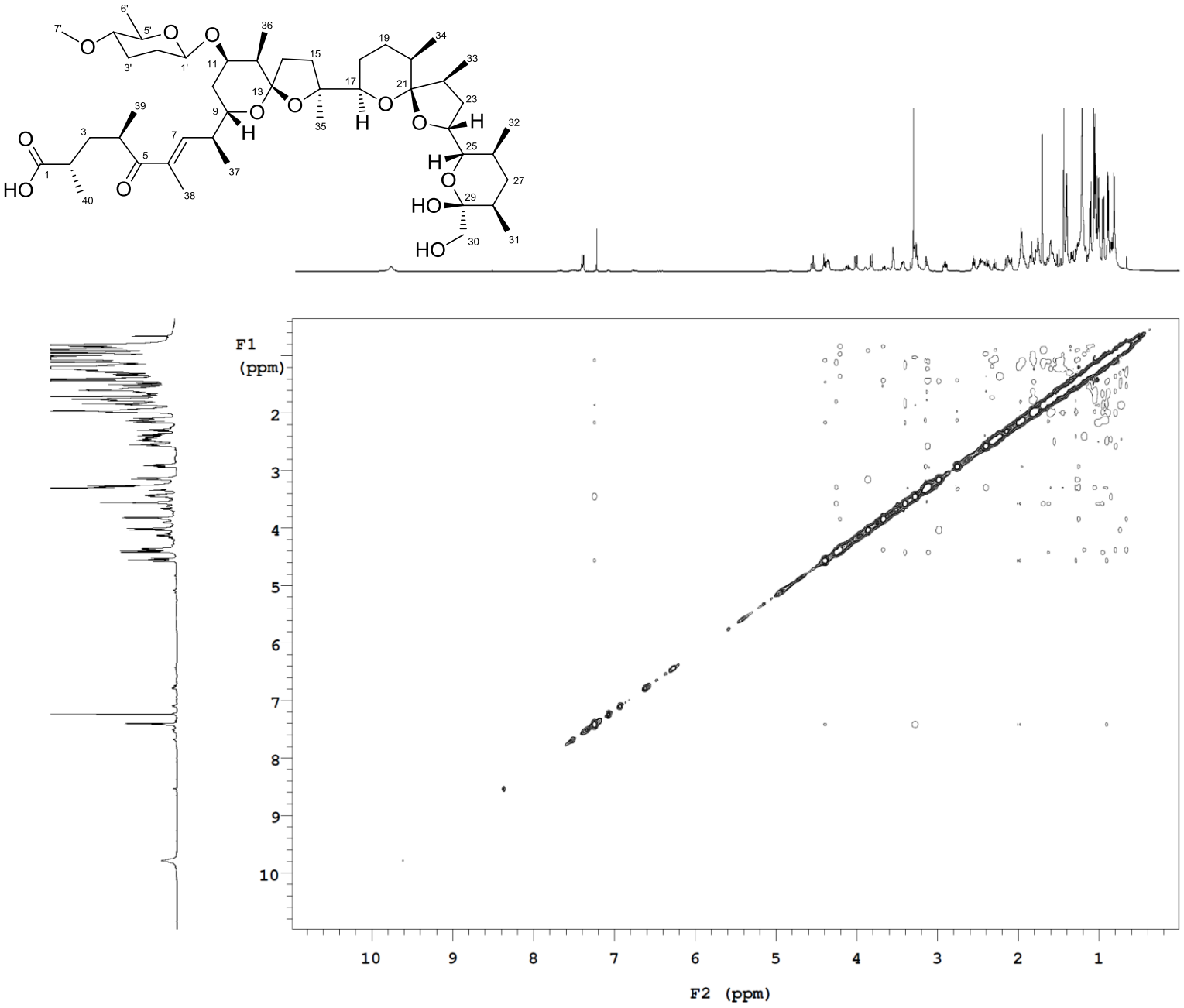


Figure S90. NOESY spectrum ($CDCl_3$, 500 MHz) of lenoremycin (**9**)

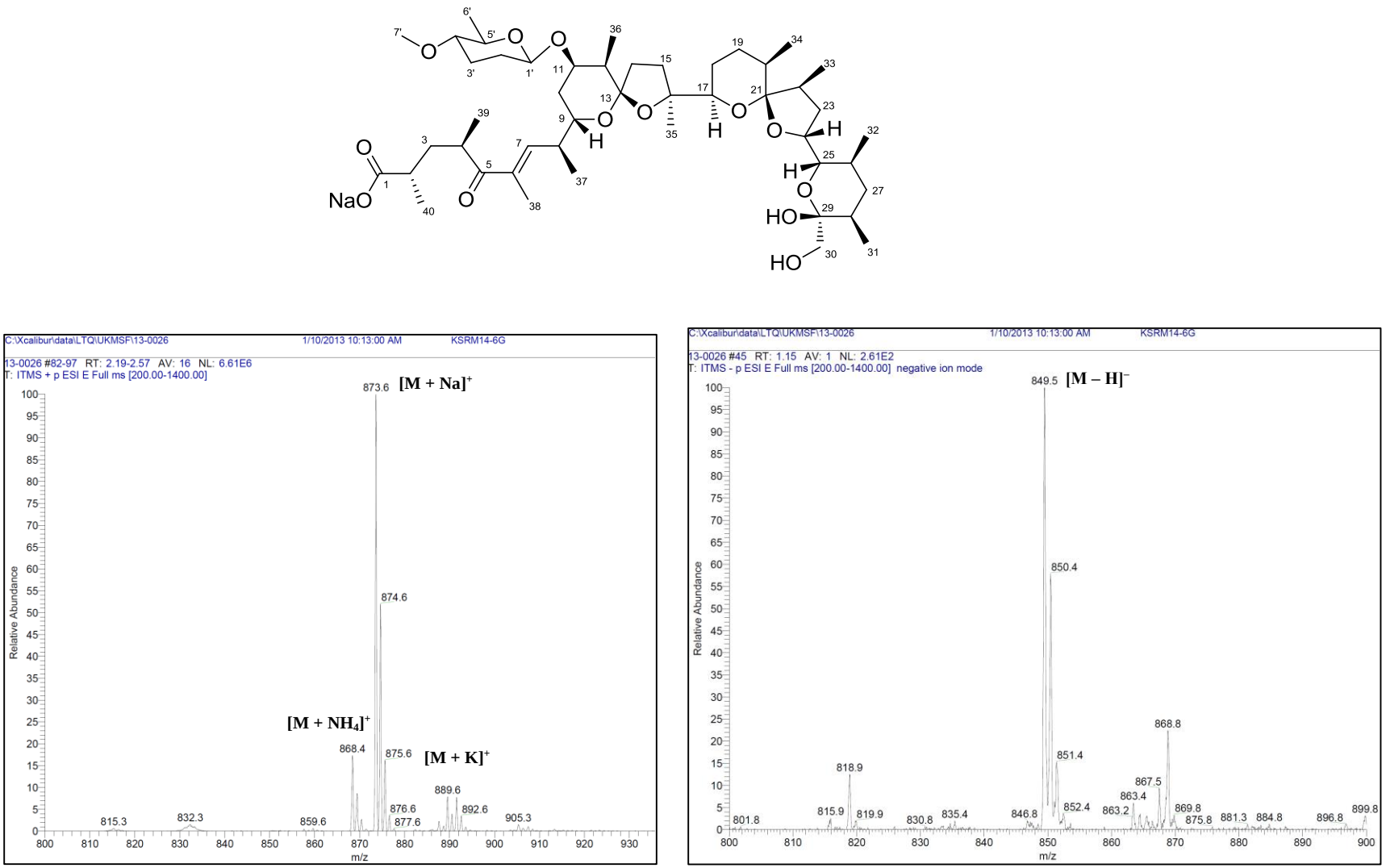
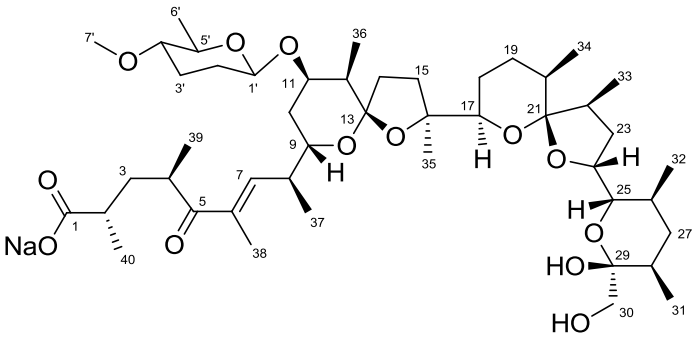
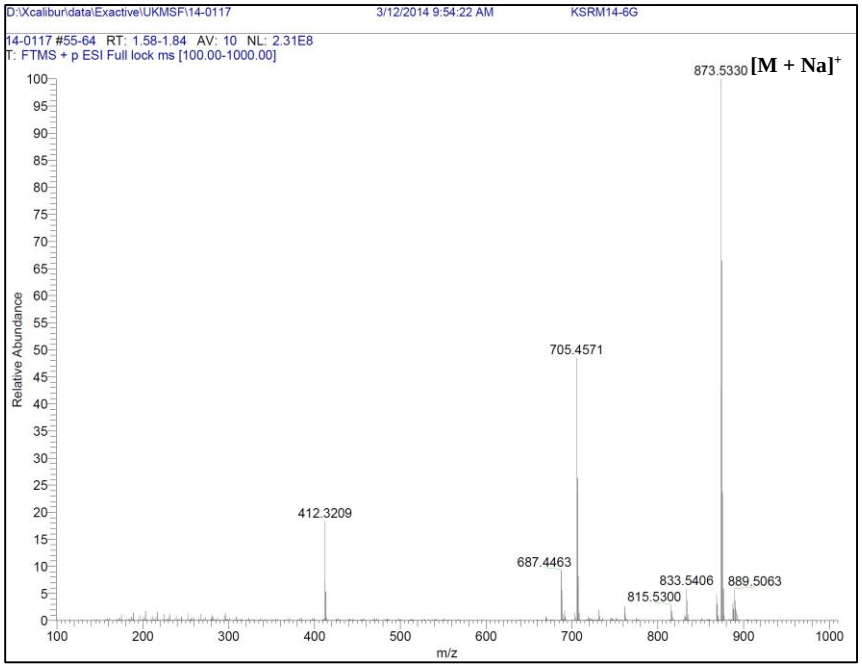


Figure S91. (+) and (–)-ESI-MS spectra of lenoremycin sodium salt (**10**) reflecting free acid form in solution [with the same mass and molecular formula as lenoremycin (**9**)].





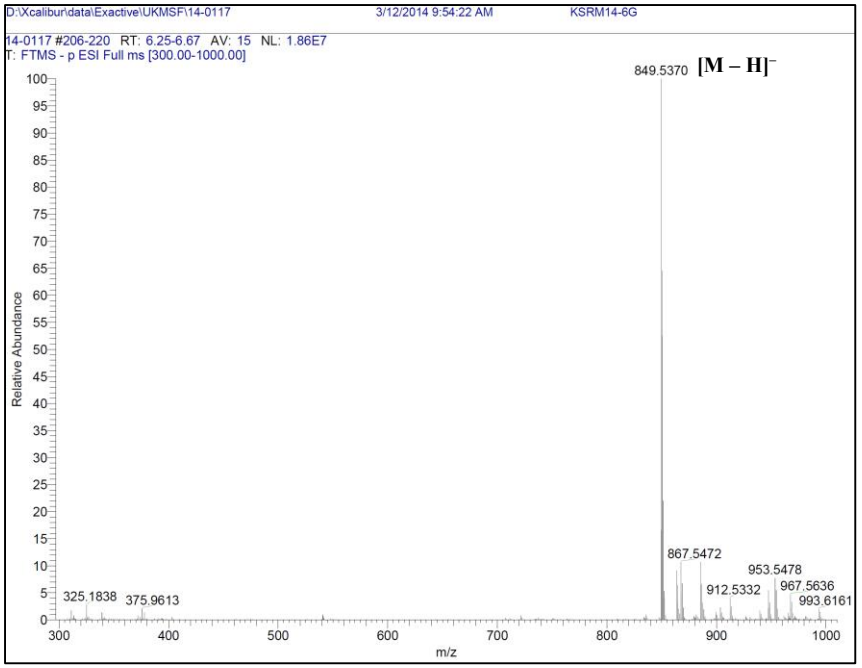


Figure S92. (+) and (–)-HRESI-MS spectra of lenoremycin sodium salt (**10**) reflecting free acid form in solution [with the same mass and molecular formula as lenoremycin (**9**)].

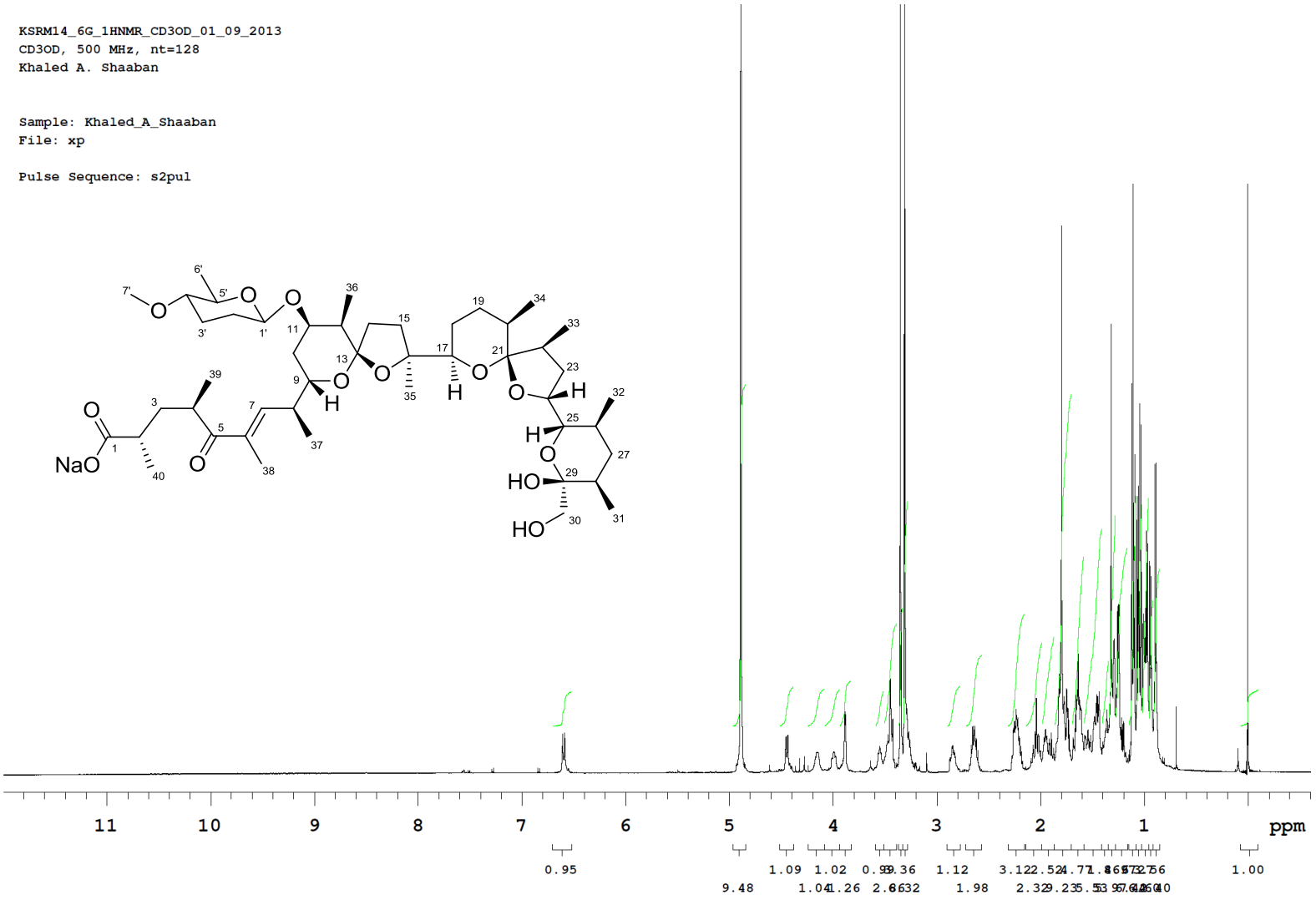


Figure S93. ^{1}H NMR spectrum (CD_3OD, 500 MHz) of lenoremycin sodium salt (**10**)

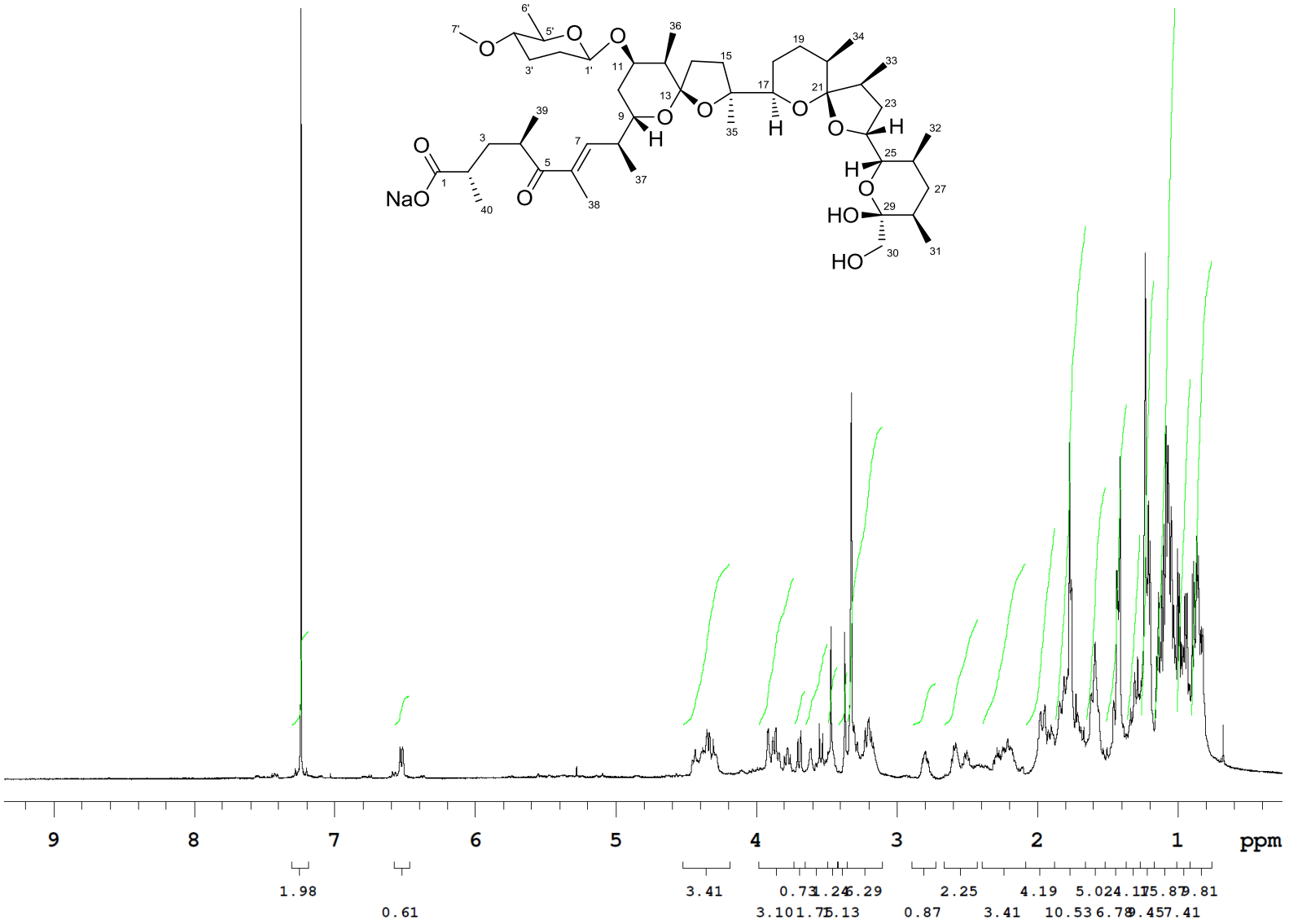


Figure S94. ^{1}H NMR spectrum ($CDCl_3$, 500 MHz) of lenoremycin sodium salt (**10**)

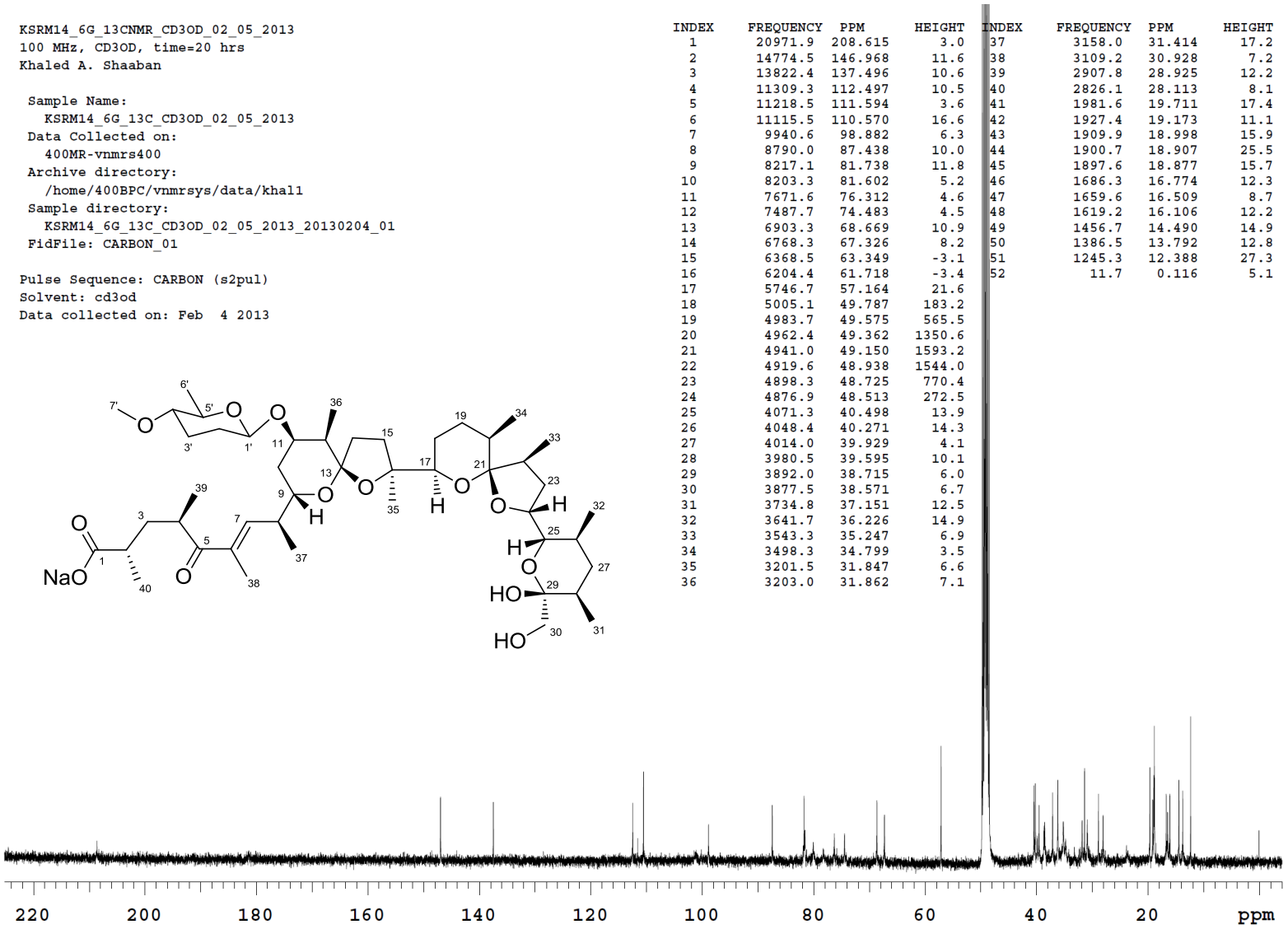


Figure S95. ^{13}C NMR spectrum (CD_3OD, 100 MHz) of lenoremycin sodium salt (**10**)

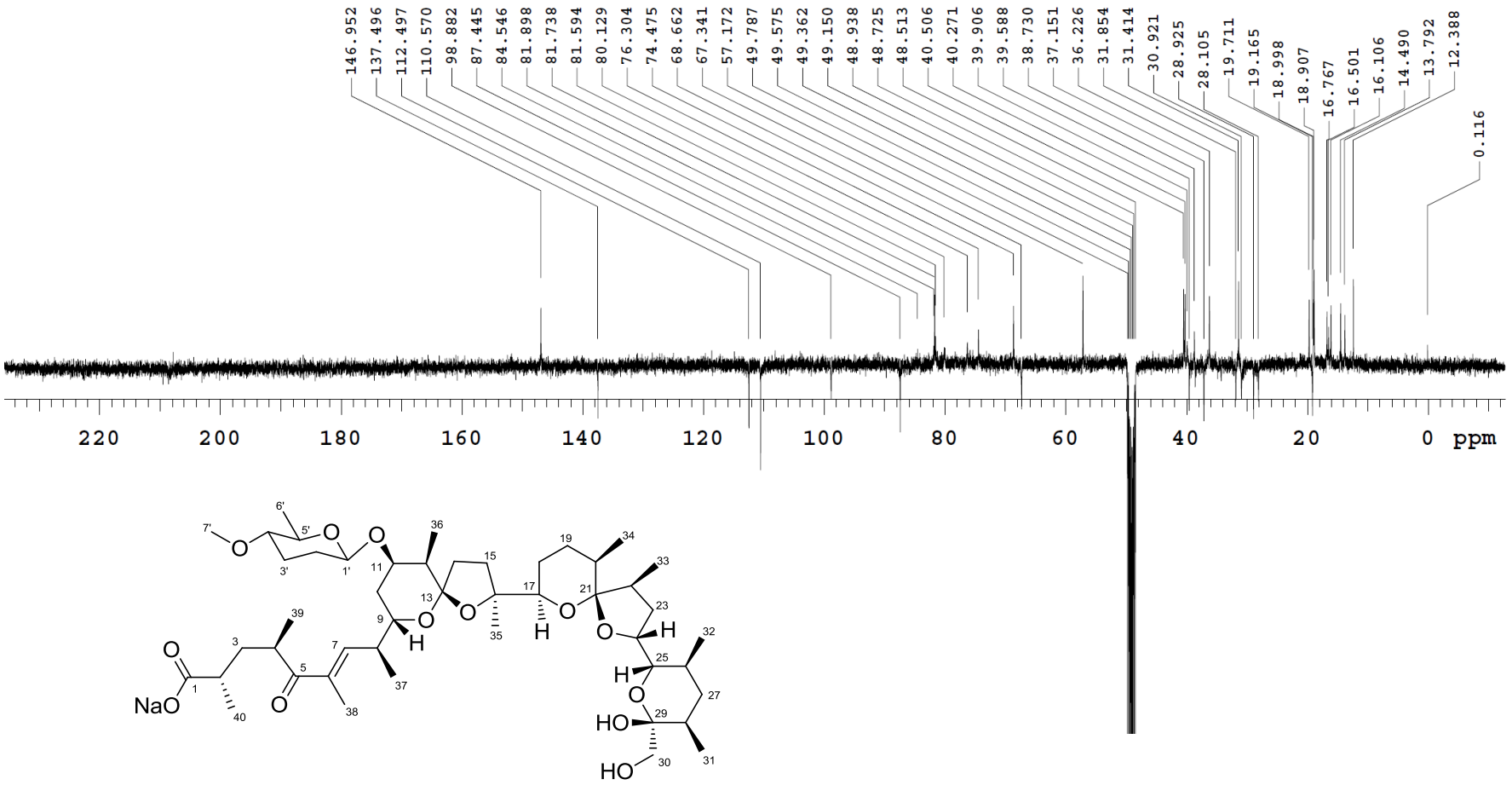
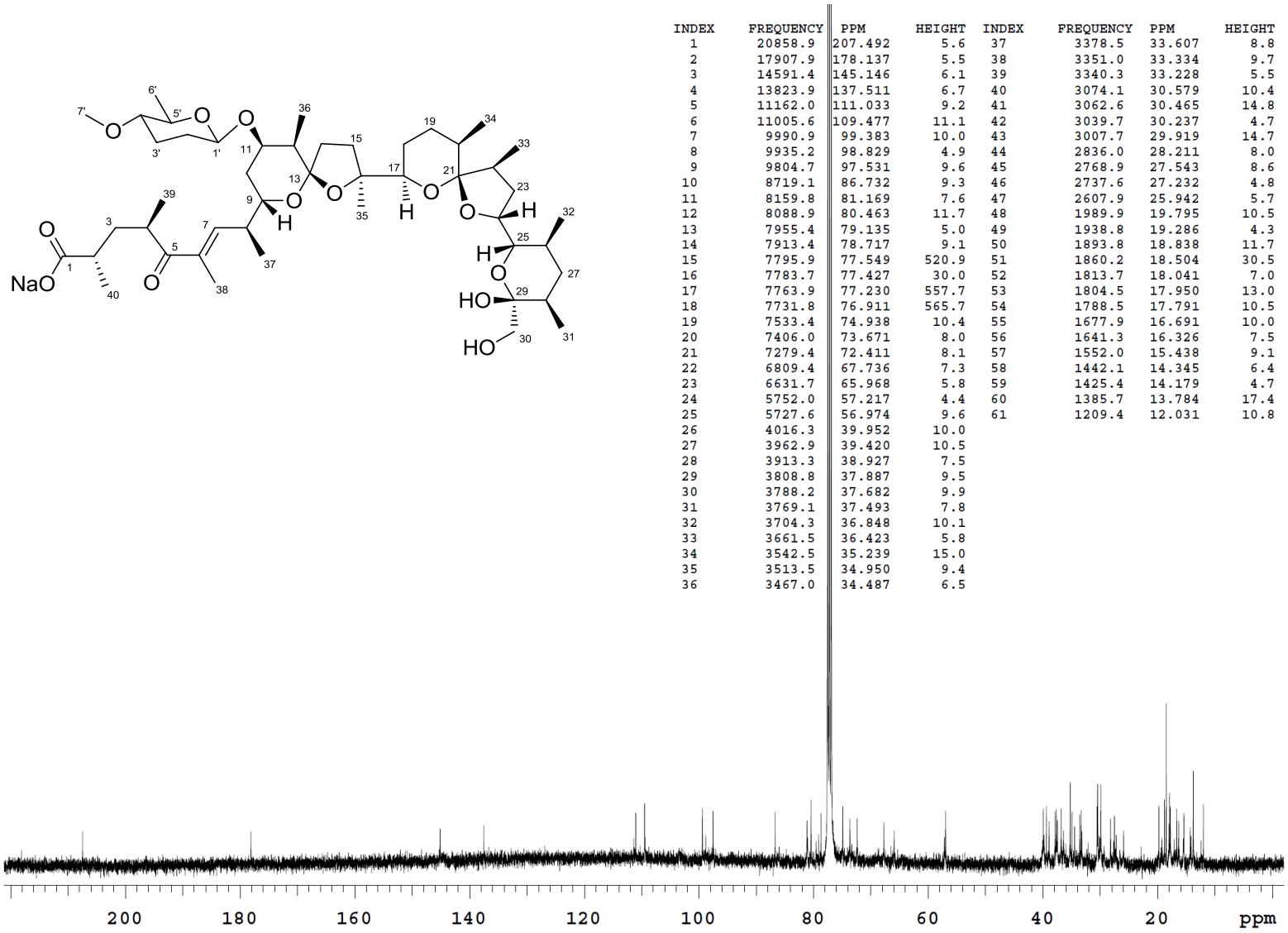


Figure S96. APT NMR spectrum (CD_3OD, 100 MHz) of lenoremycin sodium salt (**10**)



INDEX	FREQUENCY	PPM	HEIGHT	INDEX	FREQUENCY	PPM	HEIGHT
1	20858.9	207.492	5.6	37	3378.5	33.607	8.8
2	17907.9	178.137	5.5	38	3351.0	33.334	9.7
3	14591.4	145.146	6.1	39	3340.3	33.228	5.5
4	13823.9	137.511	6.7	40	3074.1	30.579	10.4
5	11162.0	111.033	9.2	41	3062.6	30.465	14.8
6	11005.6	109.477	11.1	42	3039.7	30.237	4.7
7	9990.9	99.383	10.0	43	3007.7	29.919	14.7
8	9935.2	98.829	4.9	44	2836.0	28.211	8.0
9	9804.7	97.531	9.6	45	2768.9	27.543	8.6
10	8719.1	86.732	9.3	46	2737.6	27.232	4.8
11	8159.8	81.169	7.6	47	2607.9	25.942	5.7
12	8088.9	80.463	11.7	48	1989.9	19.795	10.5
13	7955.4	79.135	5.0	49	1938.8	19.286	4.3
14	7913.4	78.717	9.1	50	1893.8	18.838	11.7
15	7795.9	77.549	520.9	51	1860.2	18.504	30.5
16	7783.7	77.427	30.0	52	1813.7	18.041	7.0
17	7763.9	77.230	557.7	53	1804.5	17.950	13.0
18	7731.8	76.911	565.7	54	1788.5	17.791	10.5
19	7533.4	74.938	10.4	55	1677.9	16.691	10.0
20	7406.0	73.671	8.0	56	1641.3	16.326	7.5
21	7279.4	72.411	8.1	57	1552.0	15.438	9.1
22	6809.4	67.736	7.3	58	1442.1	14.345	6.4
23	6631.7	65.968	5.8	59	1425.4	14.179	4.7
24	5752.0	57.217	4.4	60	1385.7	13.784	17.4
25	5727.6	56.974	9.6	61	1209.4	12.031	10.8
26	4016.3	39.952	10.0				
27	3962.9	39.420	10.5				
28	3913.3	38.927	7.5				
29	3808.8	37.887	9.5				
30	3788.2	37.682	9.9				
31	3769.1	37.493	7.8				
32	3704.3	36.848	10.1				
33	3661.5	36.423	5.8				
34	3542.5	35.239	15.0				
35	3513.5	34.950	9.4				
36	3467.0	34.487	6.5				

Figure S97. ^{13}C NMR spectrum ($CDCl_3$, 100 MHz) of lenoremycin sodium salt (**10**)

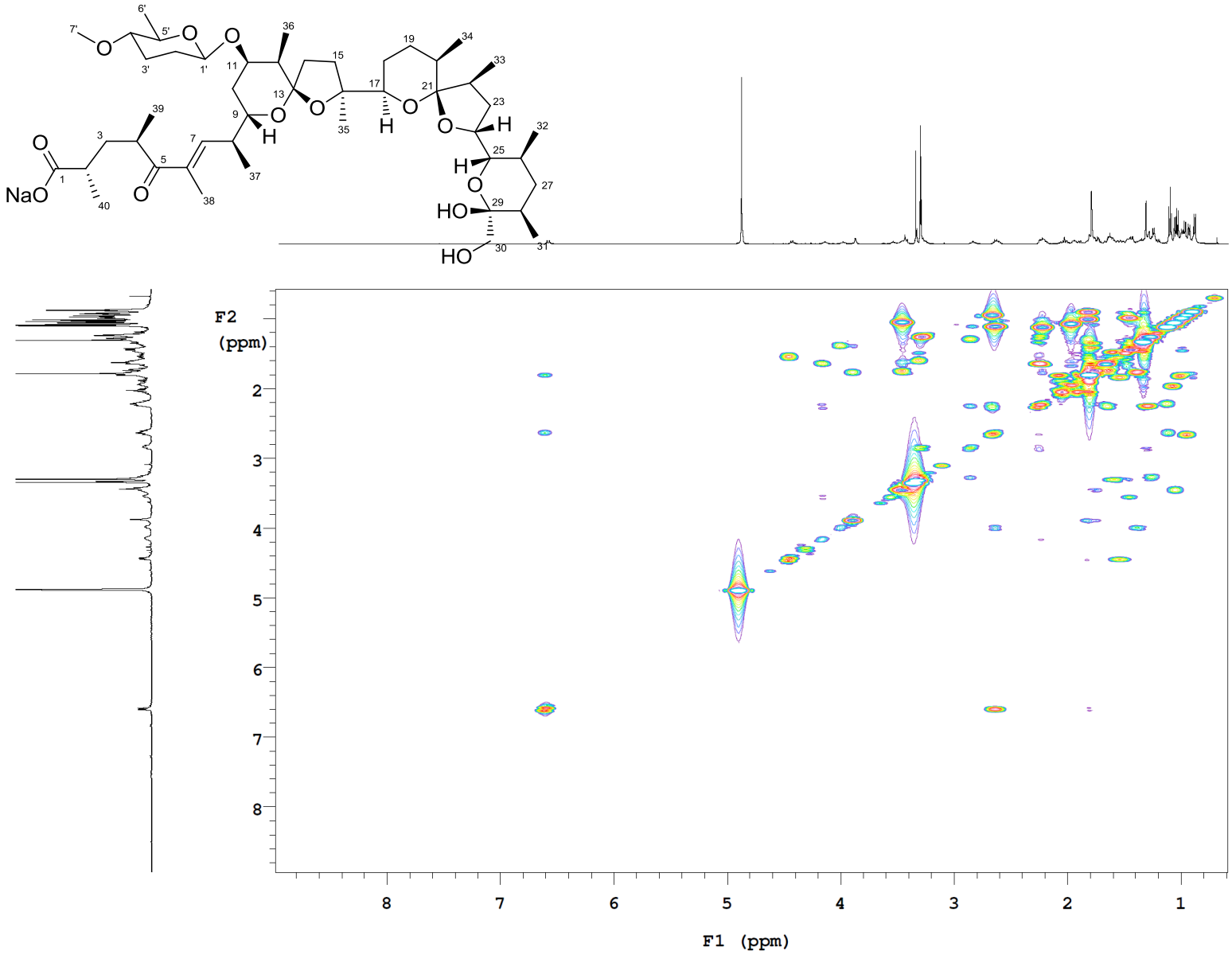


Figure S98. $^{1}H,^{1}H$-COSY spectrum (CD_3OD, 500 MHz) of lenoremycin sodium salt (**10**)

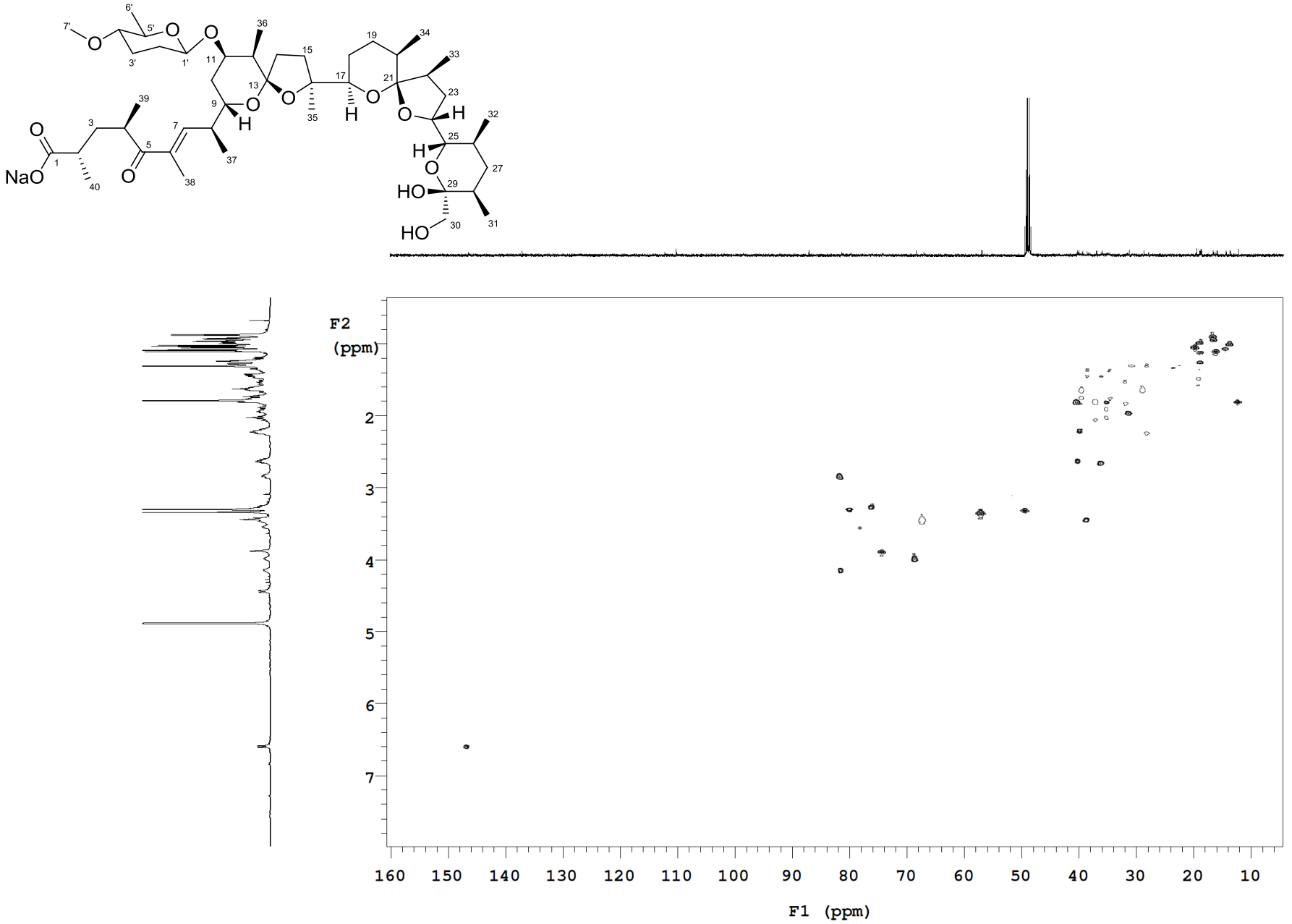


Figure S99. HSQC spectrum (CD_3OD, 500 MHz) of lenoremycin sodium salt (**10**)

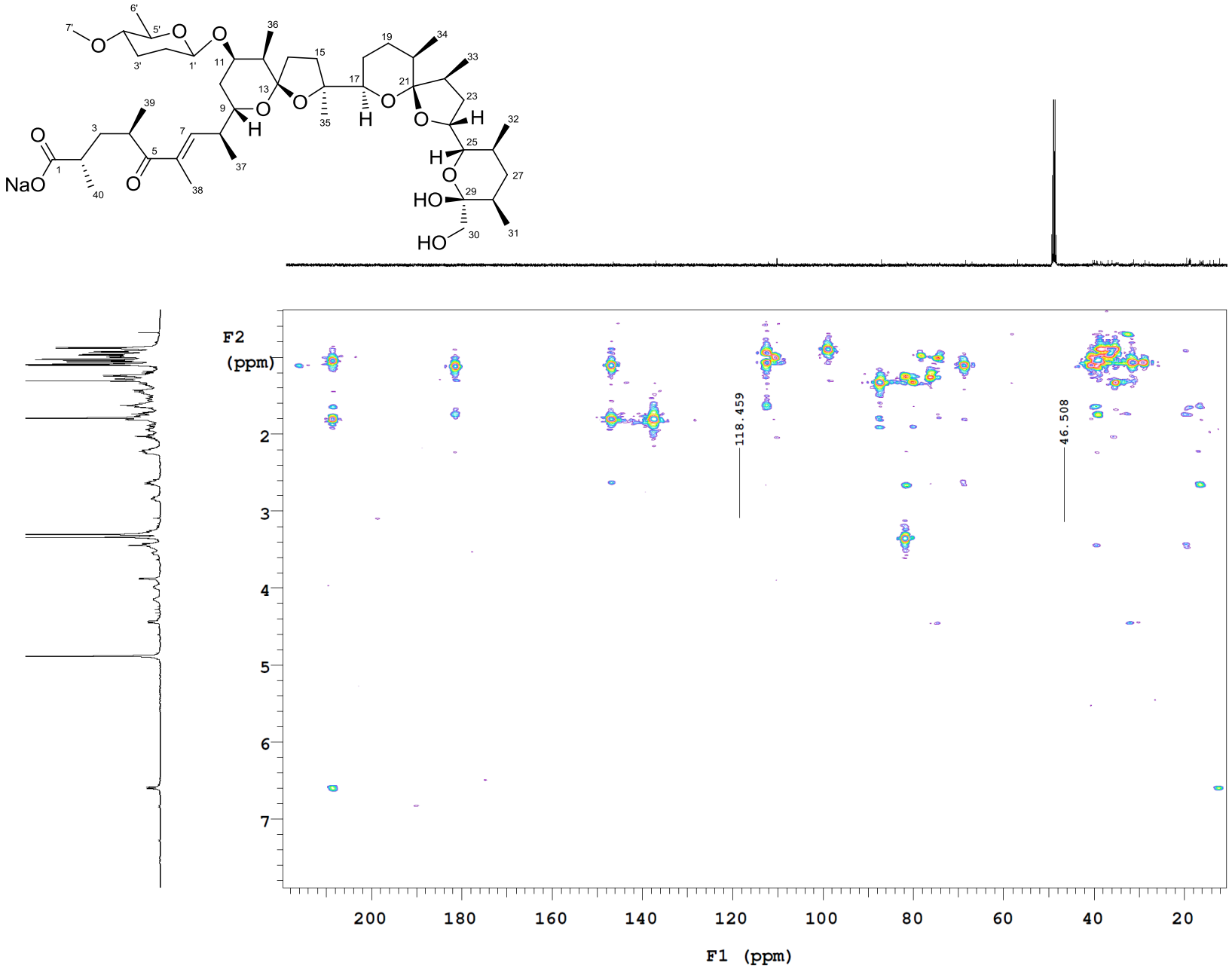


Figure S100. HMBC spectrum (CD_3OD, 500 MHz) of lenoremycin sodium salt (**10**)

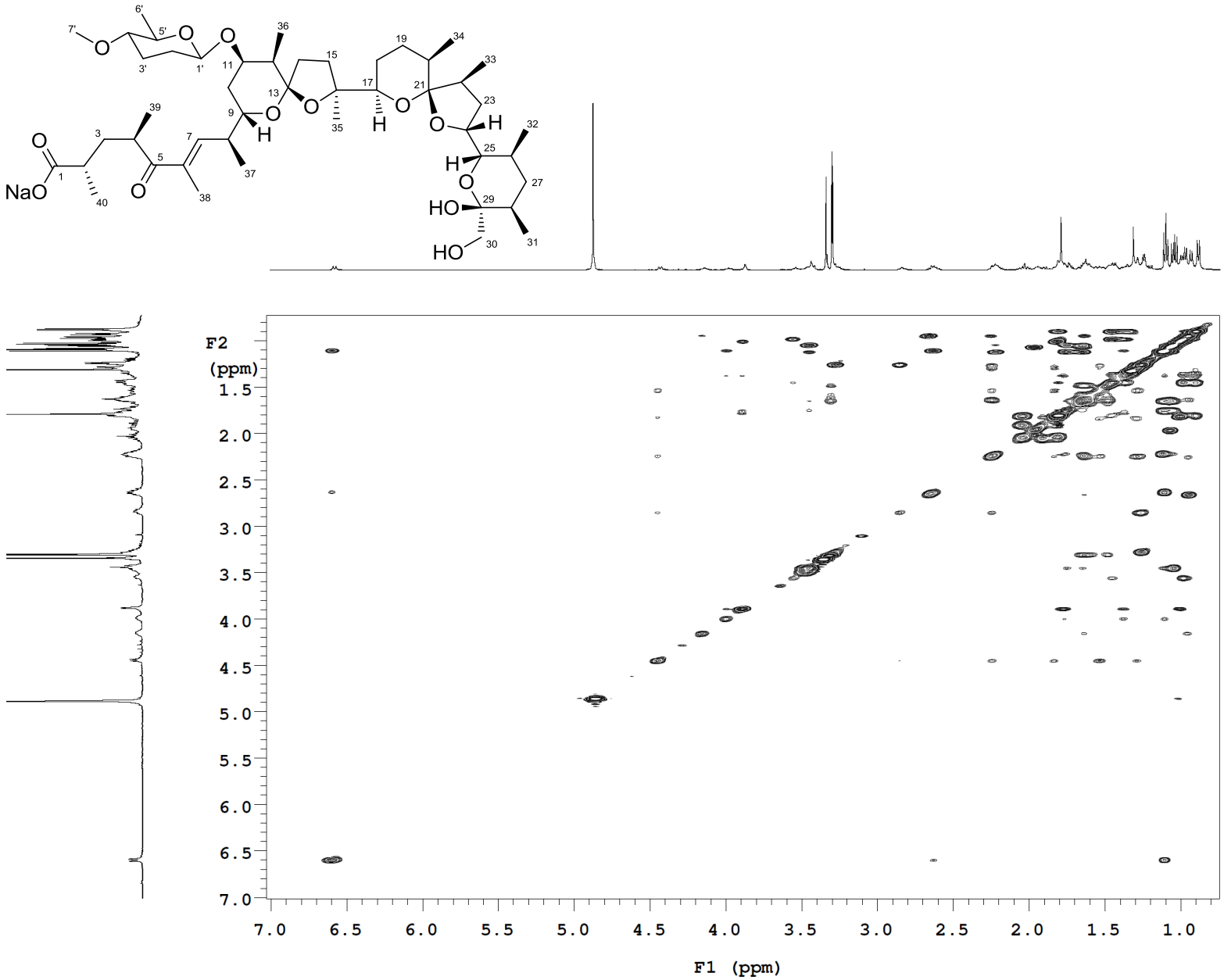


Figure S101. TOCSY spectrum (CD_3OD, 500 MHz) of lenoremycin sodium salt (**10**)

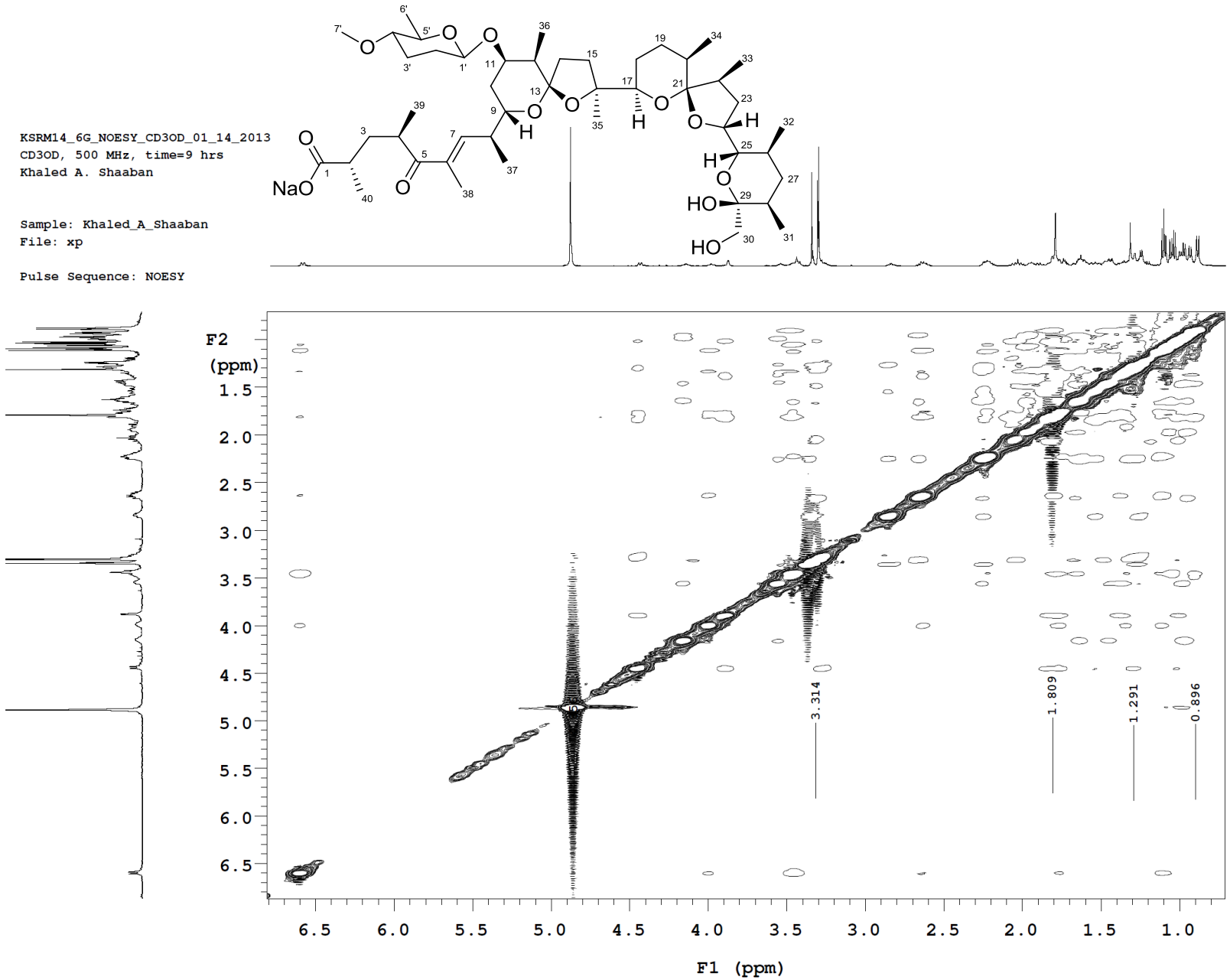


Figure S102. NOESY spectrum (CD_3OD, 500 MHz) of lenoremycin sodium salt (**10**)

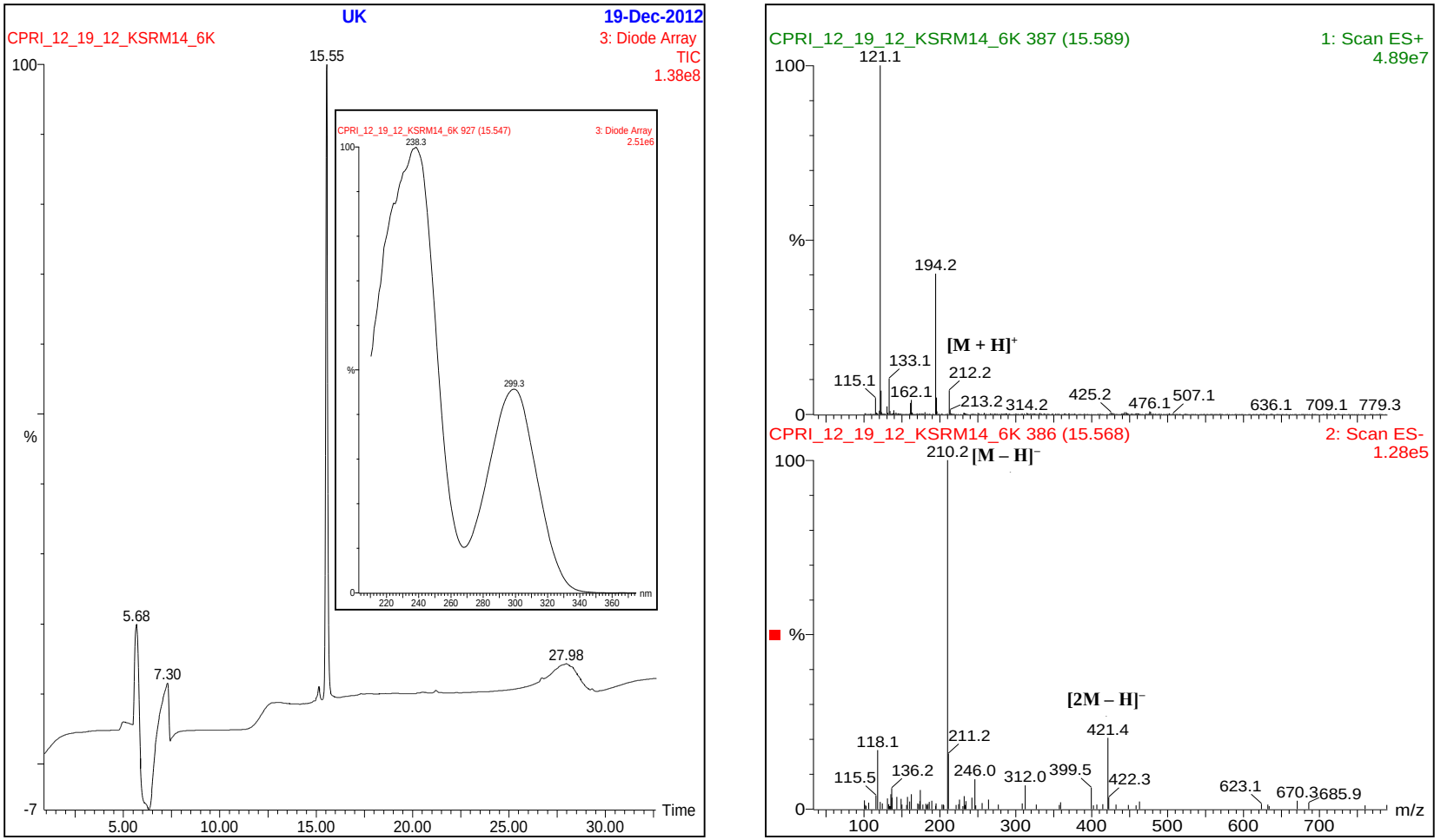


Figure S103. HPLC/UV/MS analyses of the purified *N*-salicyloyl-2-aminopropan-1,3-diol (**11**). Detection wavelength: 210-550 nm; **solvent A:** H_2O/0.1% Formic acid; **solvent B:** CH_3CN/0.1% Formic acid; flow rate: 0.5 mL min^{-1}; 0-4 min, 10% B; 4-22 min, 10-100% B; 22-27 min, 100% B; 27-29 min, 100%-10% B; 29-35 min, 10 % B.

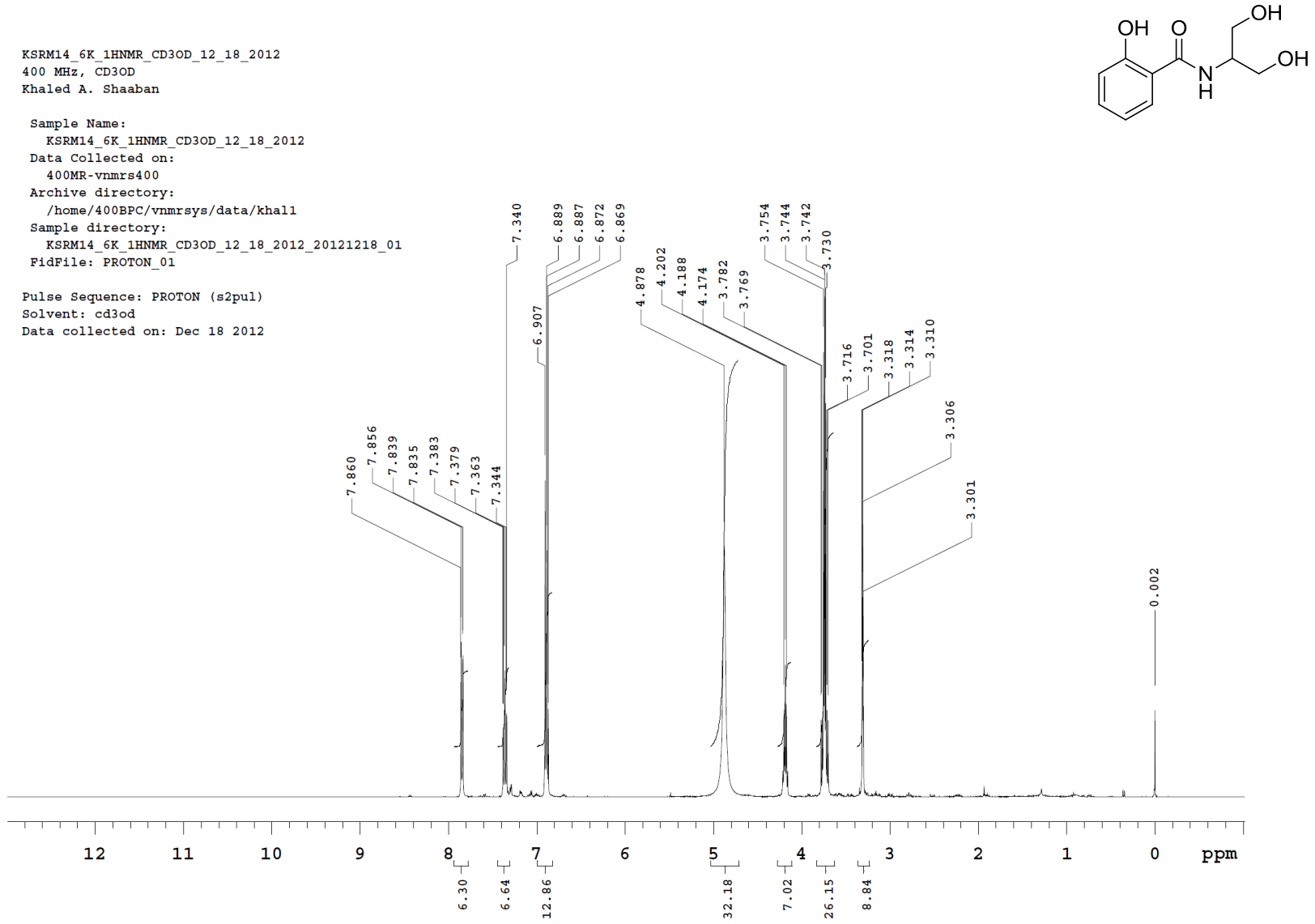


Figure S104. ^{1}H NMR spectrum (CD_3OD, 400 MHz) of *N*-salicyloyl-2-aminopropan-1,3-diol (**11**)

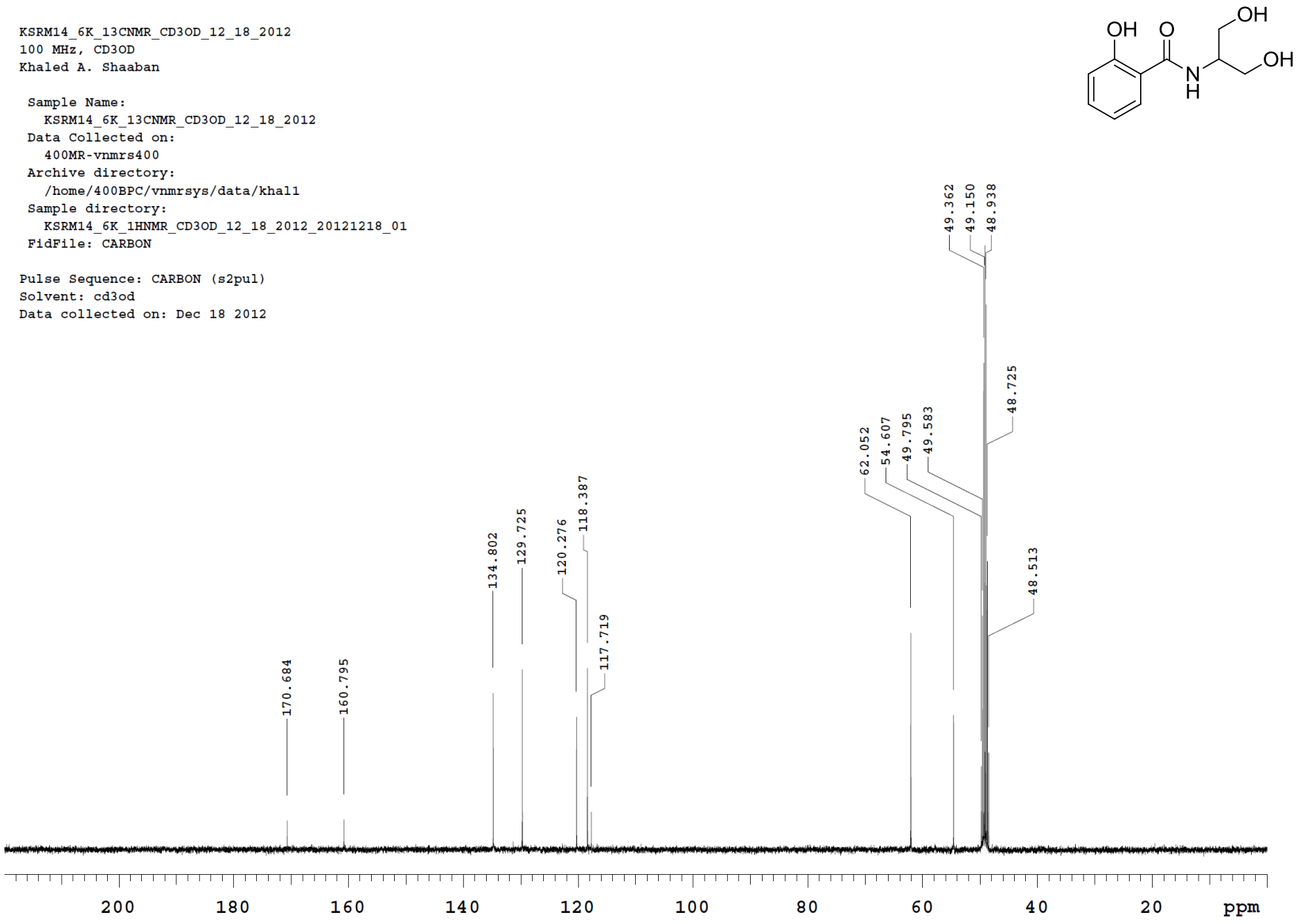


Figure S105. ^{13}C NMR spectrum (CD_3OD, 100 MHz) of *N*-salicyloyl-2-aminopropan-1,3-diol (**11**)

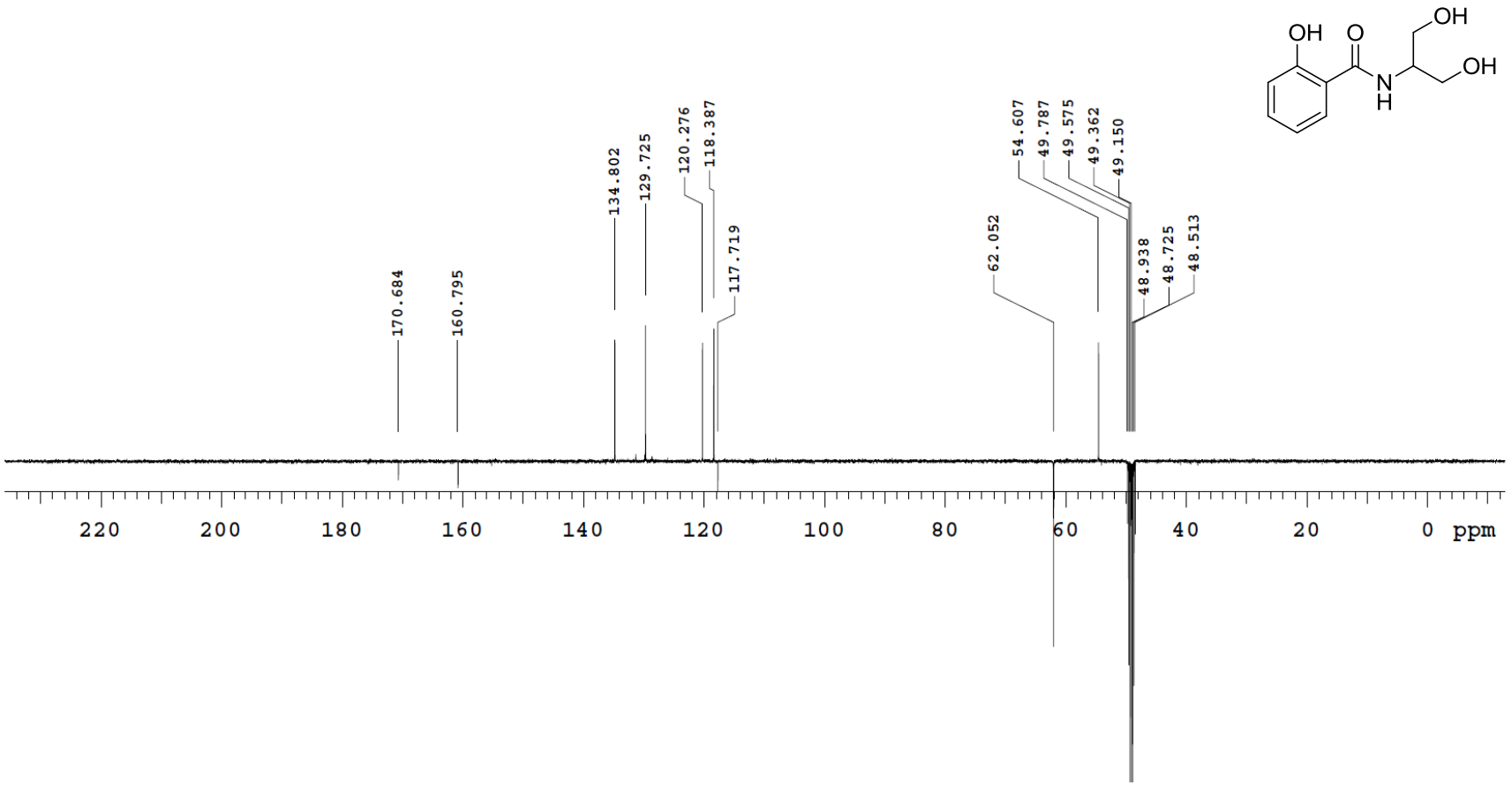


Figure S106. APT NMR spectrum (CD_3OD, 100 MHz) of *N*-salicyloyl-2-aminopropan-1,3-diol (**11**)

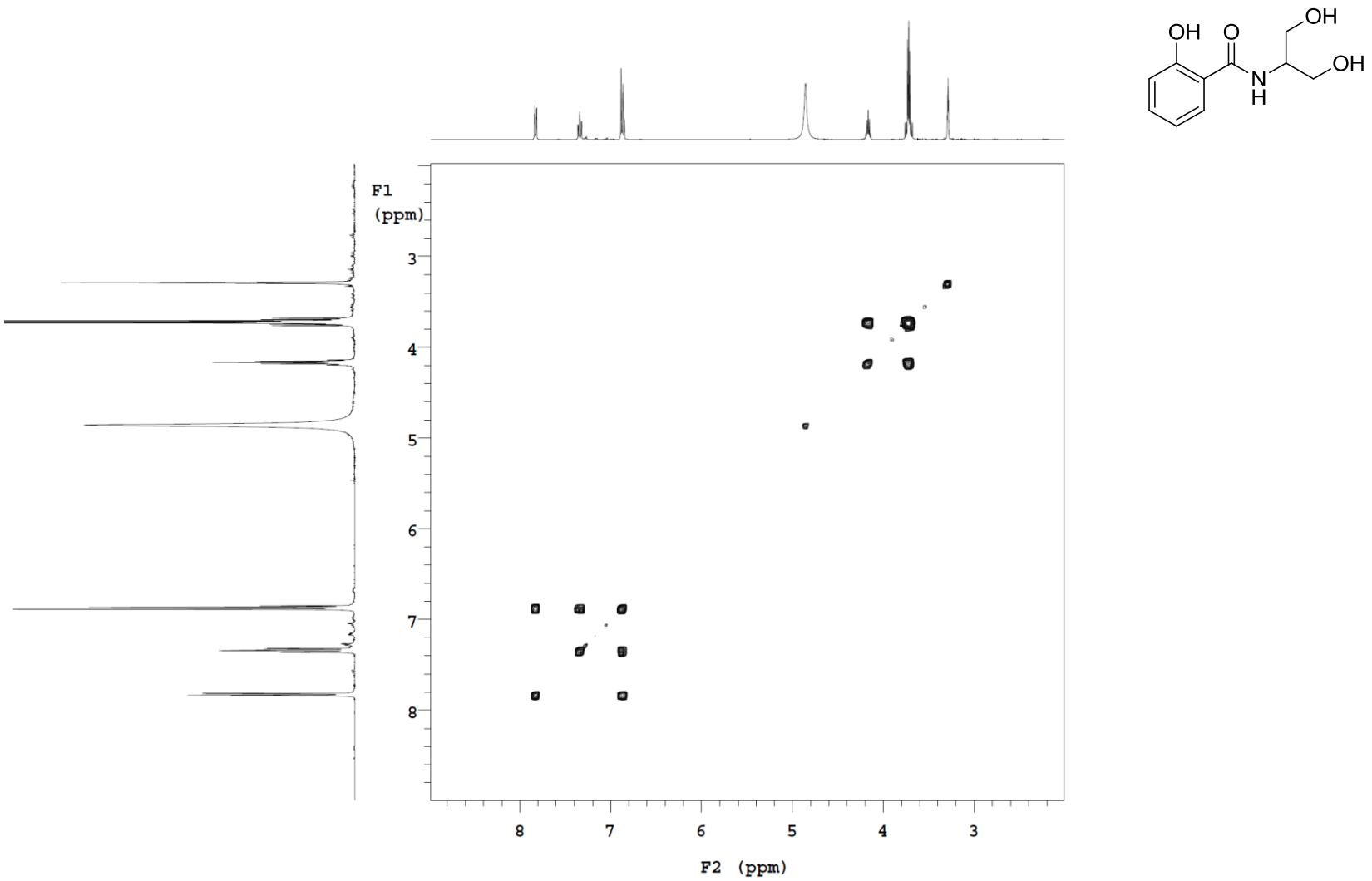


Figure S107. ^{1}H,^{1}H-COSY spectrum (CD_3OD, 100 MHz) of *N*-salicyloyl-2-aminopropan-1,3-diol (**11**)

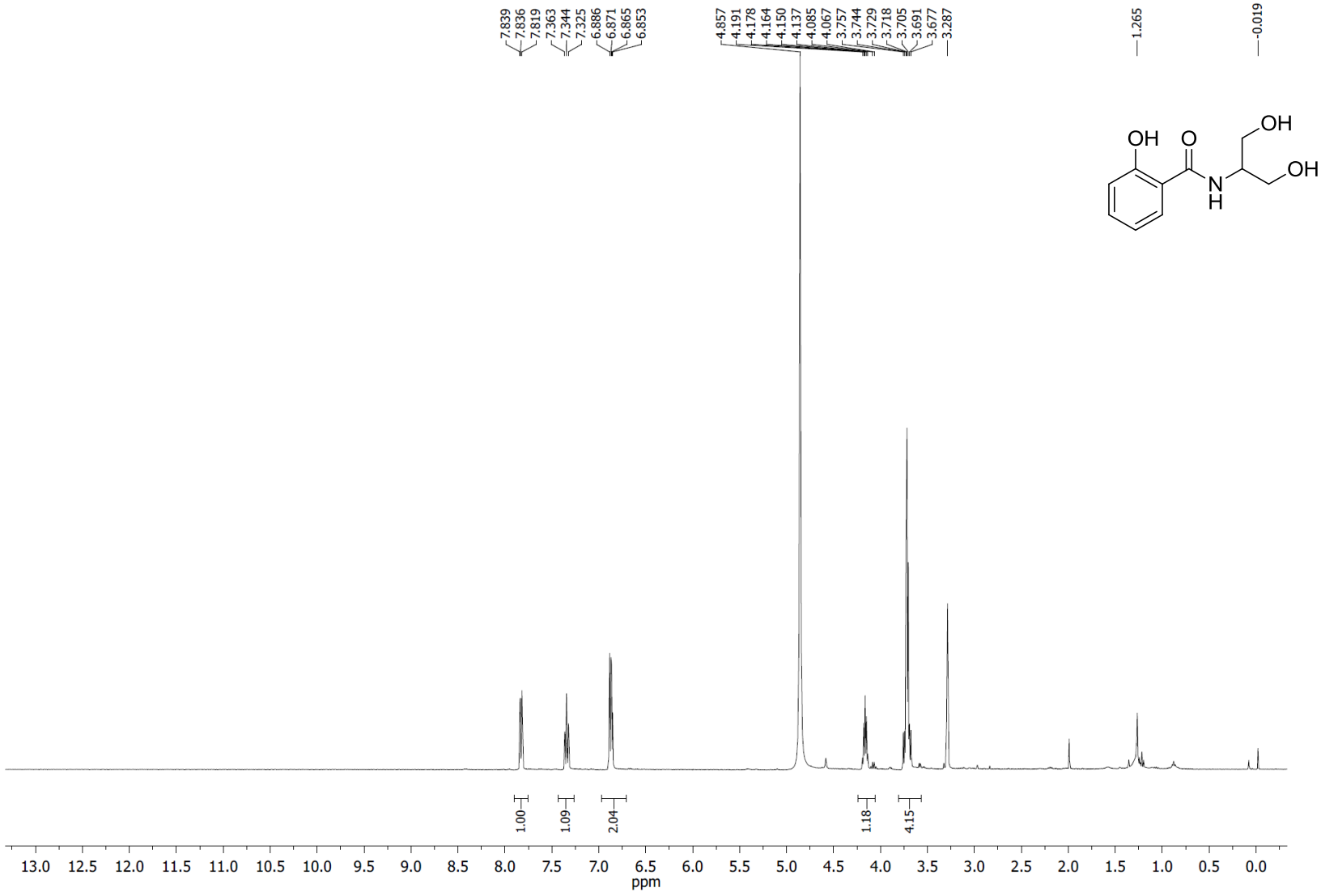


Figure S108. ^{1}H NMR spectrum (CD_3OD, 400 MHz) of synthesized *N*-salicyloyl-2-aminopropan-1,3-diol (**11**)

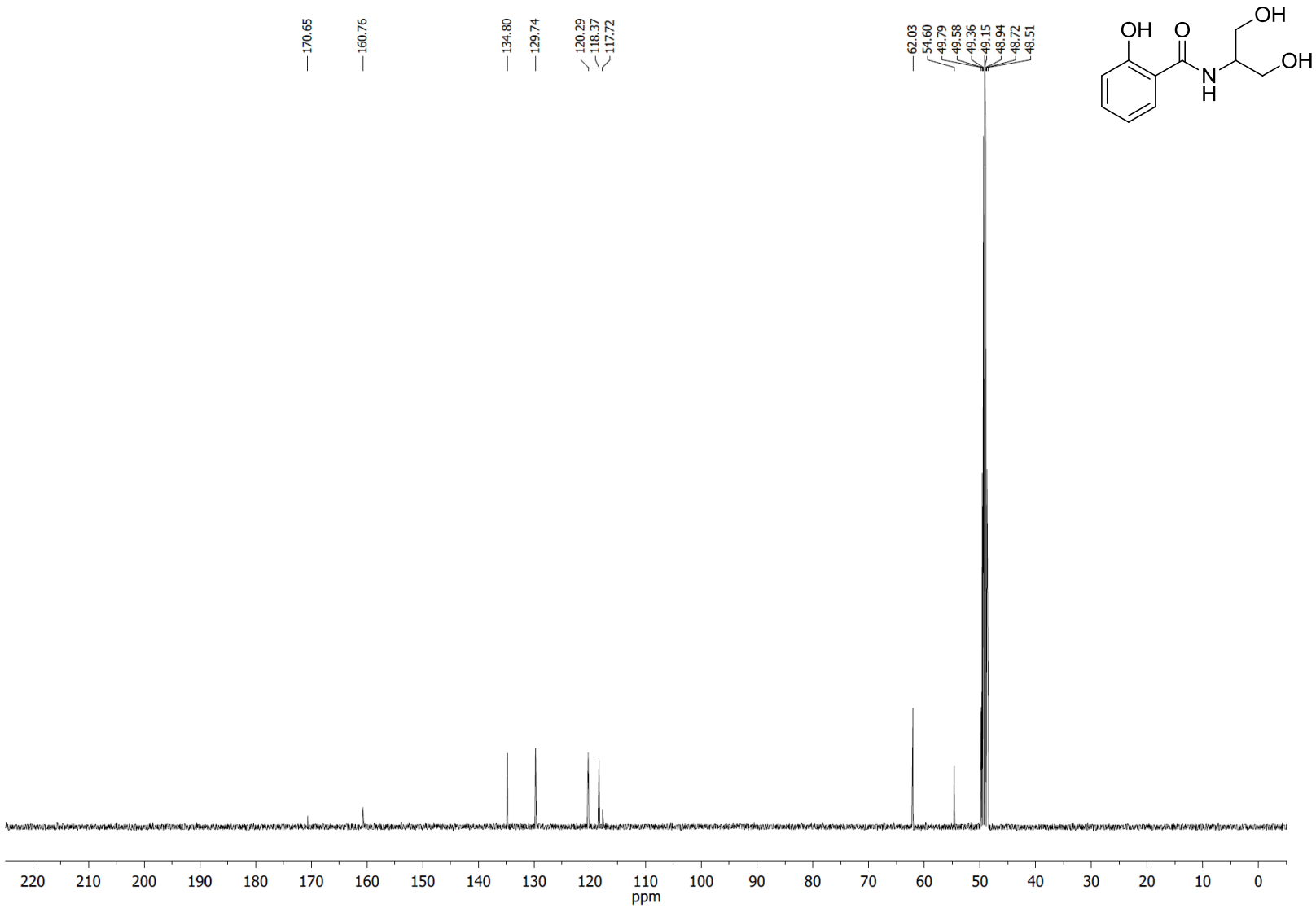


Figure S109. ^{13}C NMR spectrum (CD_3OD, 100 MHz) of synthesized *N*-salicyloyl-2-aminopropan-1,3-diol (**11**)

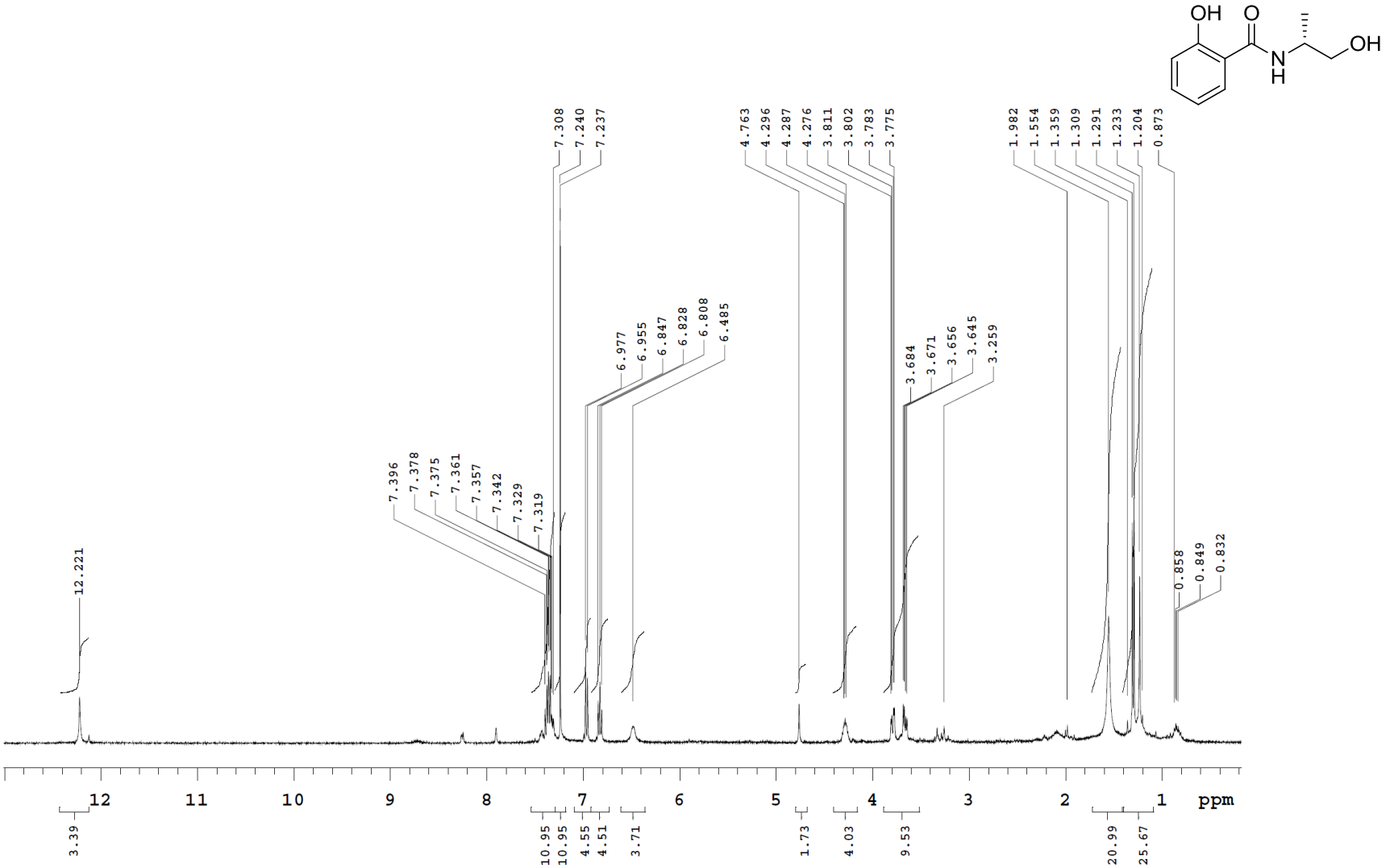


Figure S110. ^{1}H NMR spectrum ($CDCl_3$, 400 MHz) of (*R*)-(−)-*N*-salicyloyl-2-aminopropan-1-ol (**12a**)

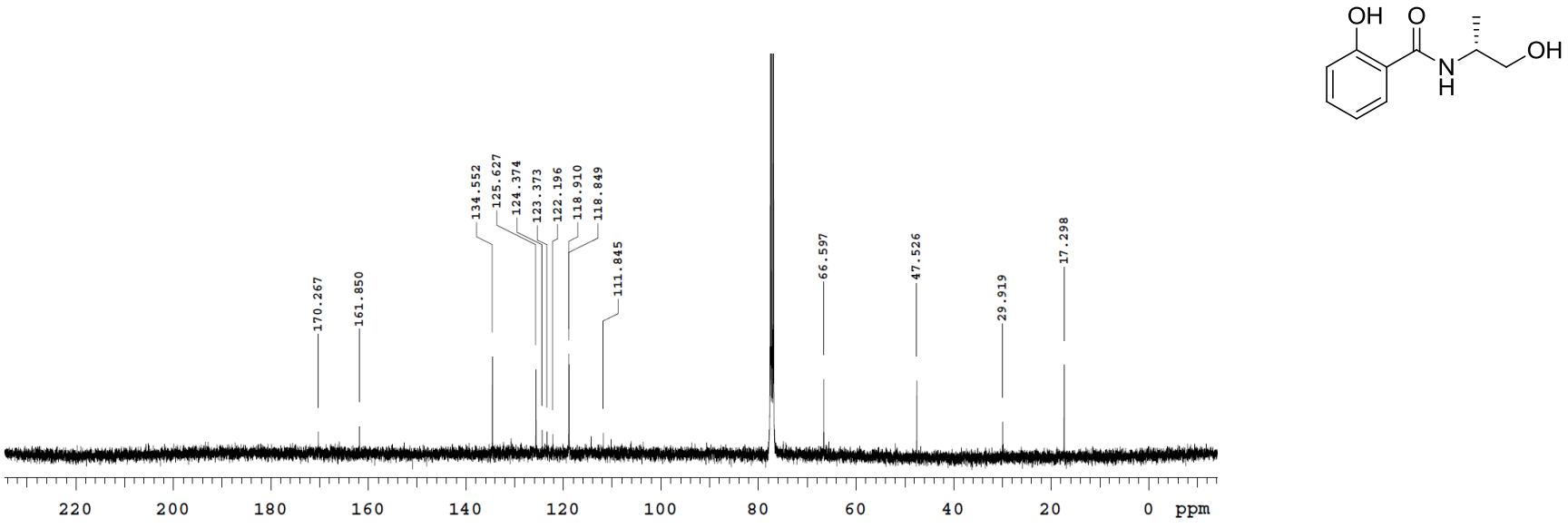


Figure S111. ^{13}C NMR spectrum ($CDCl_3$, 100 MHz) of (*R*)-(−)-*N*-salicyloyl-2-aminopropan-1-ol (**12a**)

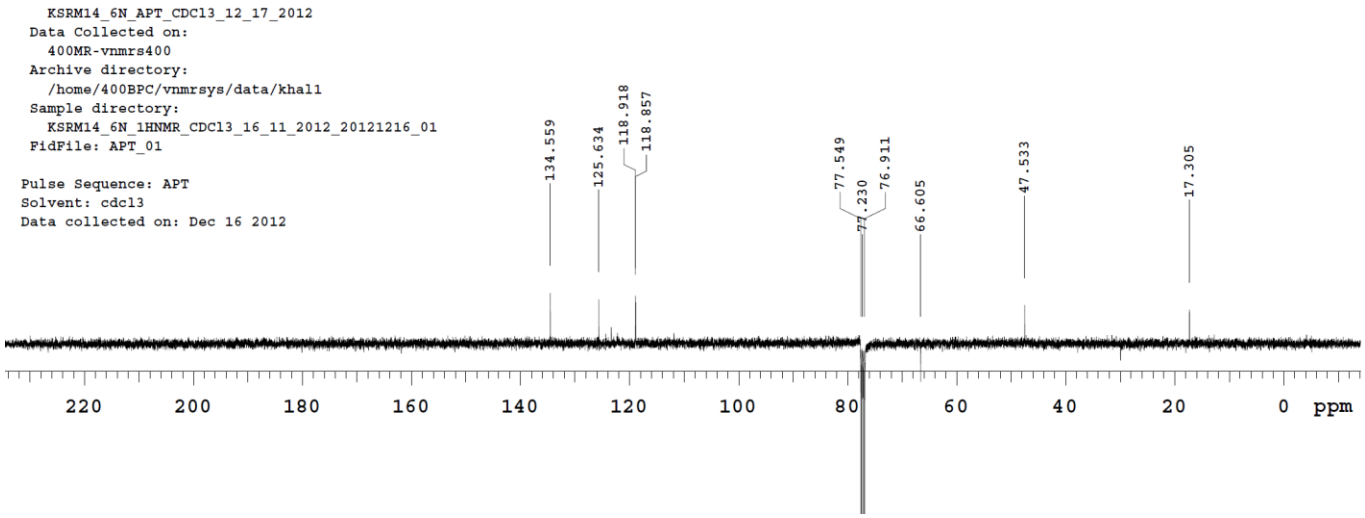


Figure S112. APT NMR spectrum ($CDCl_3$, 100 MHz) of (*R*)-(−)-*N*-salicyloyl-2-aminopropan-1-ol (**12a**)

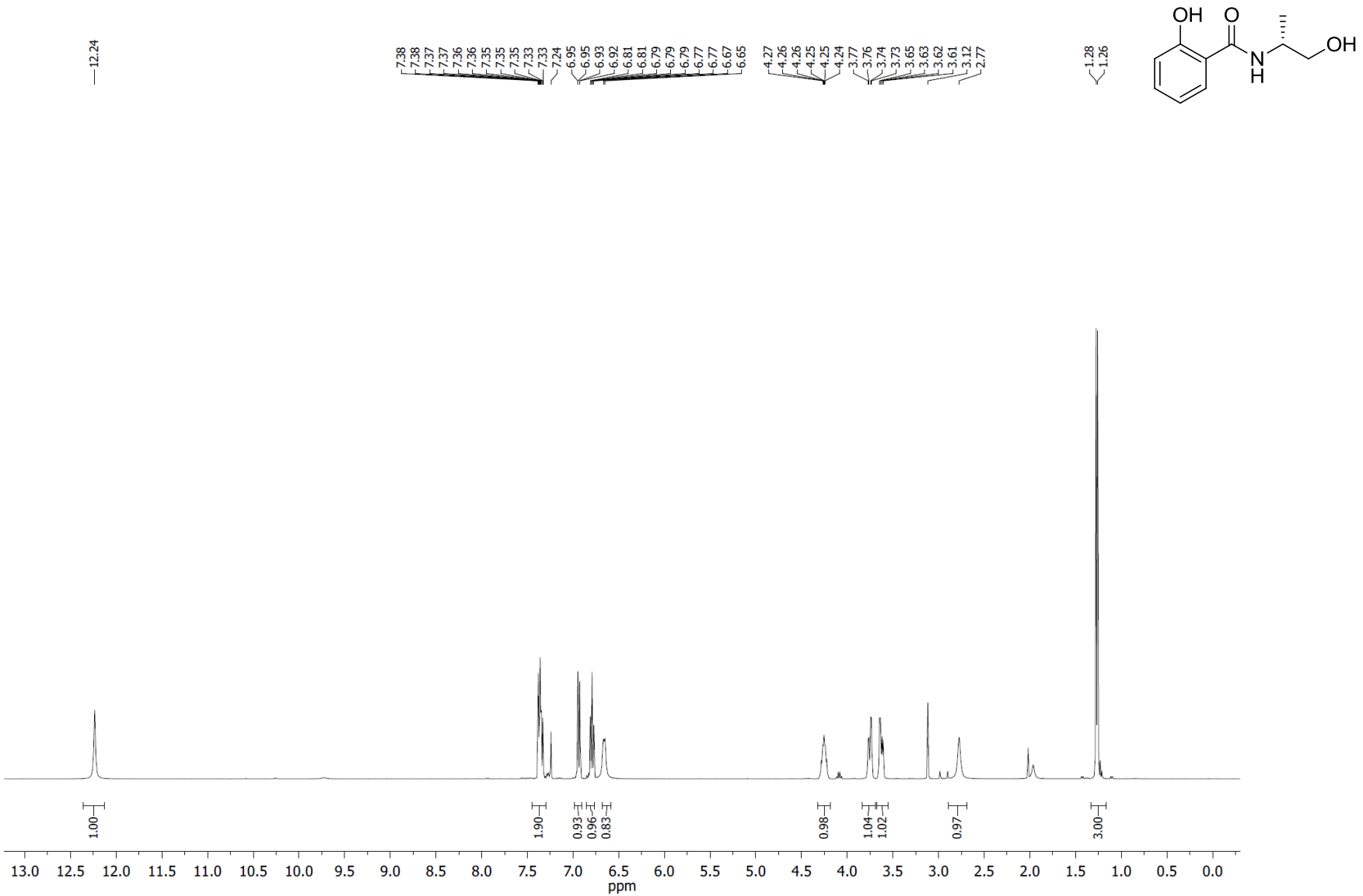


Figure S113. ^{1}H NMR spectrum ($CDCl_3$, 400 MHz) of synthesized (*R*)-(−)-*N*-salicyloyl-2-aminopropan-1-ol (**12a**)

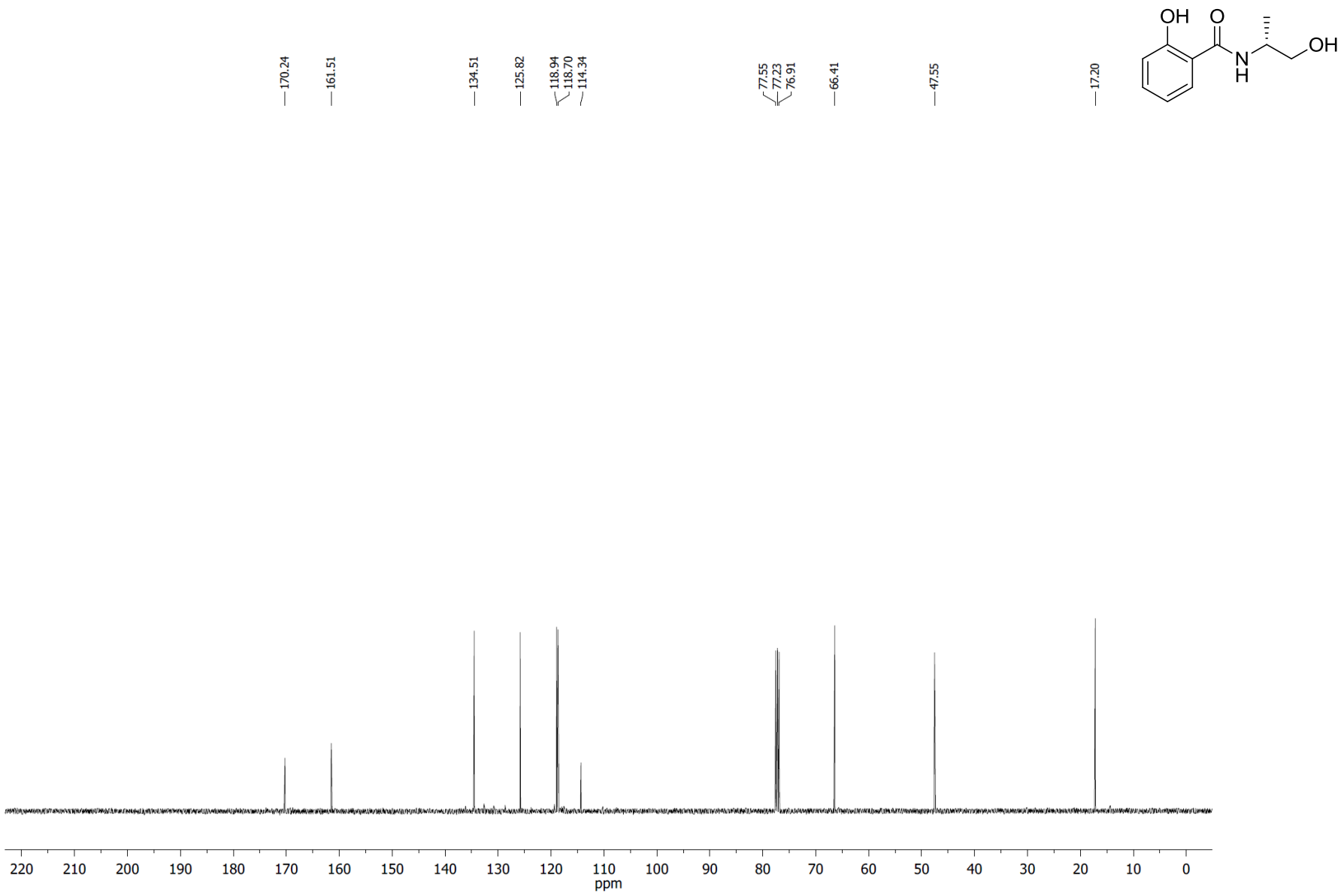


Figure S114. ^{13}C NMR spectrum ($CDCl_3$, 100 MHz) of synthesized (*R*)-(−)-*N*-salicyloyl-2-aminopropan-1-ol (**12a**)

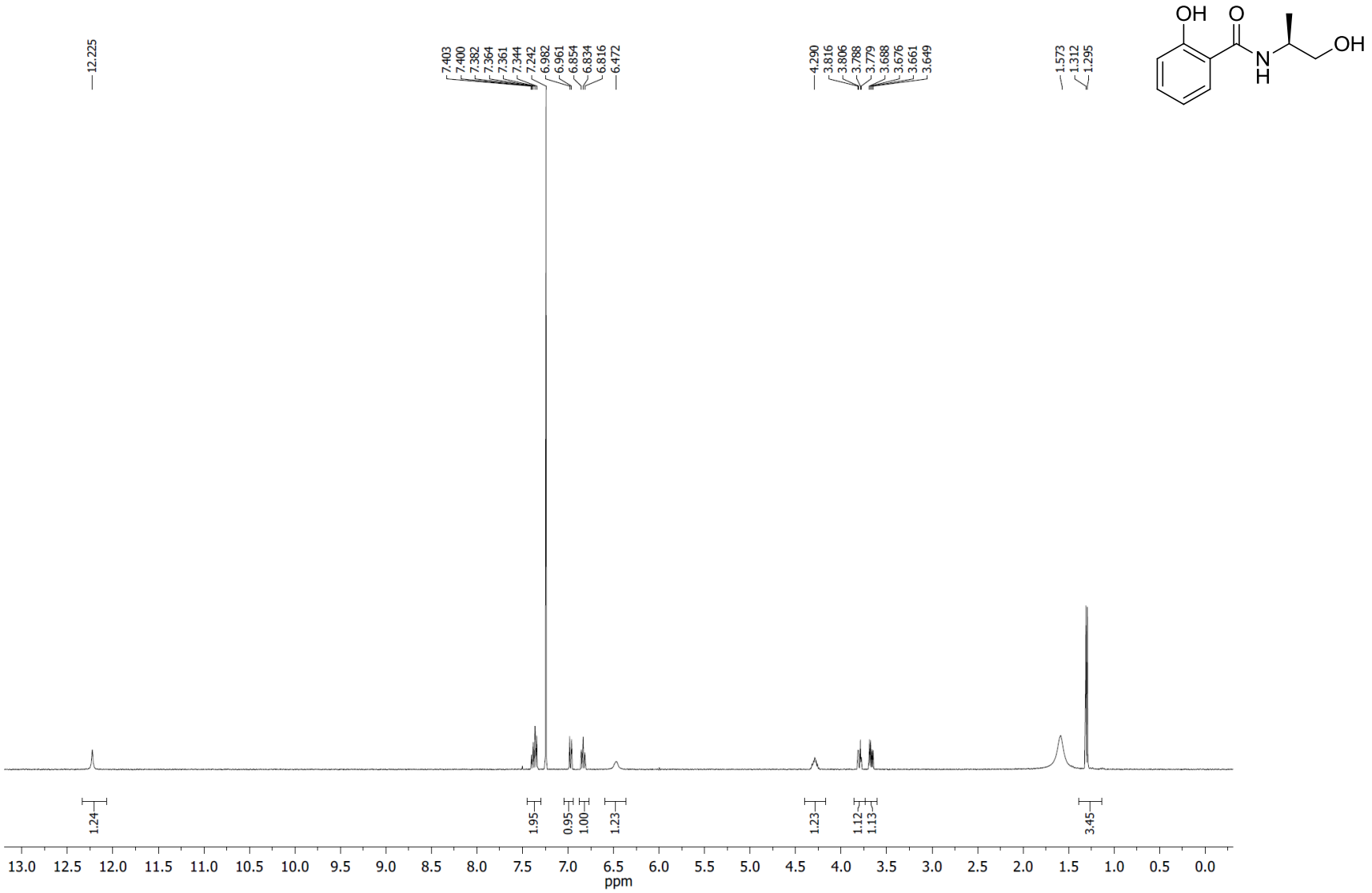


Figure S115. ^{1}H NMR spectrum ($CDCl_3$, 400 MHz) of synthesized (*S*)-(+)-*N*-salicyloyl-2-aminopropan-1-ol (**12b**)

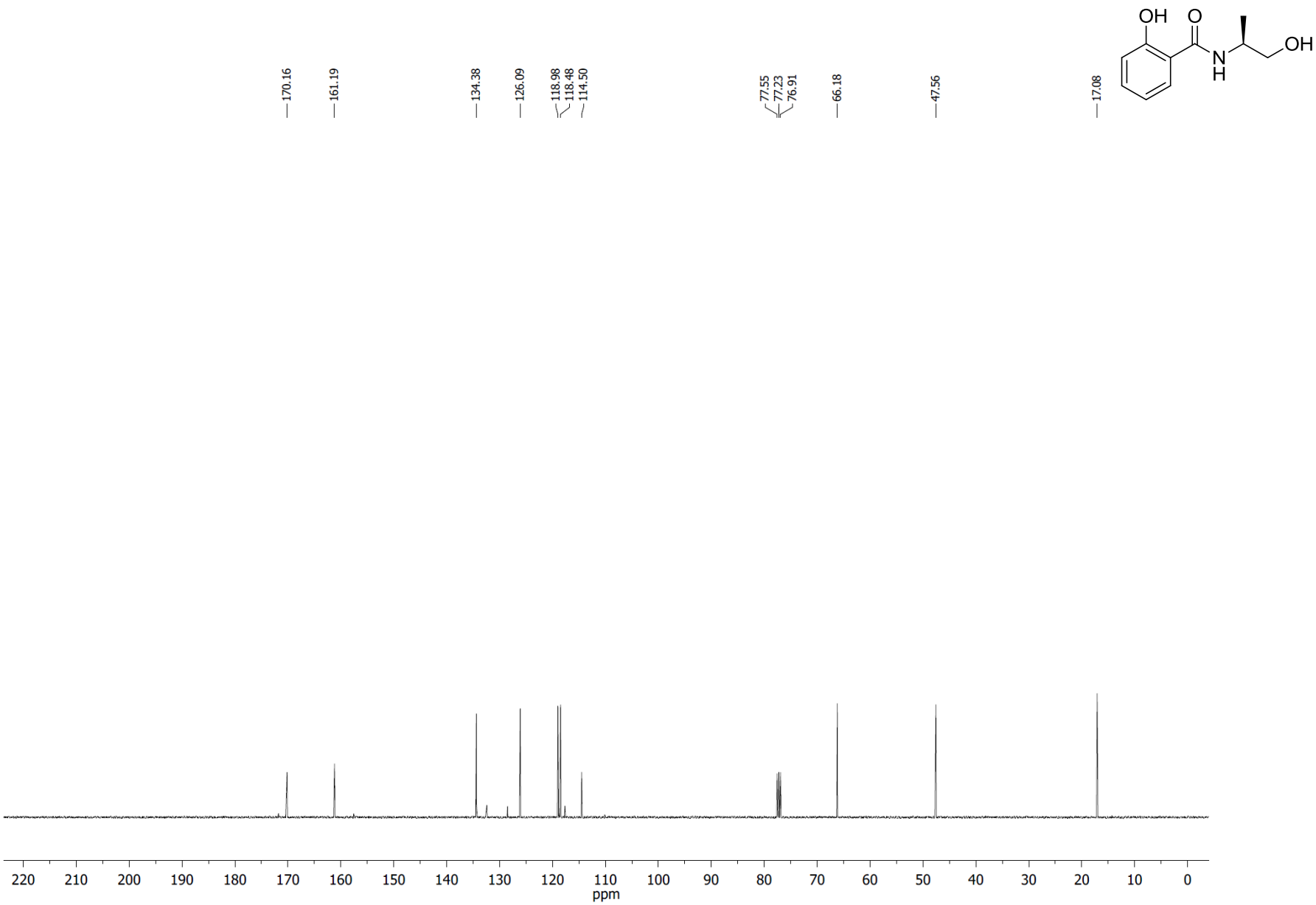


Figure S116. ^{13}C NMR spectrum ($CDCl_3$, 100 MHz) of synthesized (*S*)-(+)-*N*-salicyloyl-2-aminopropan-1-ol (**12b**)

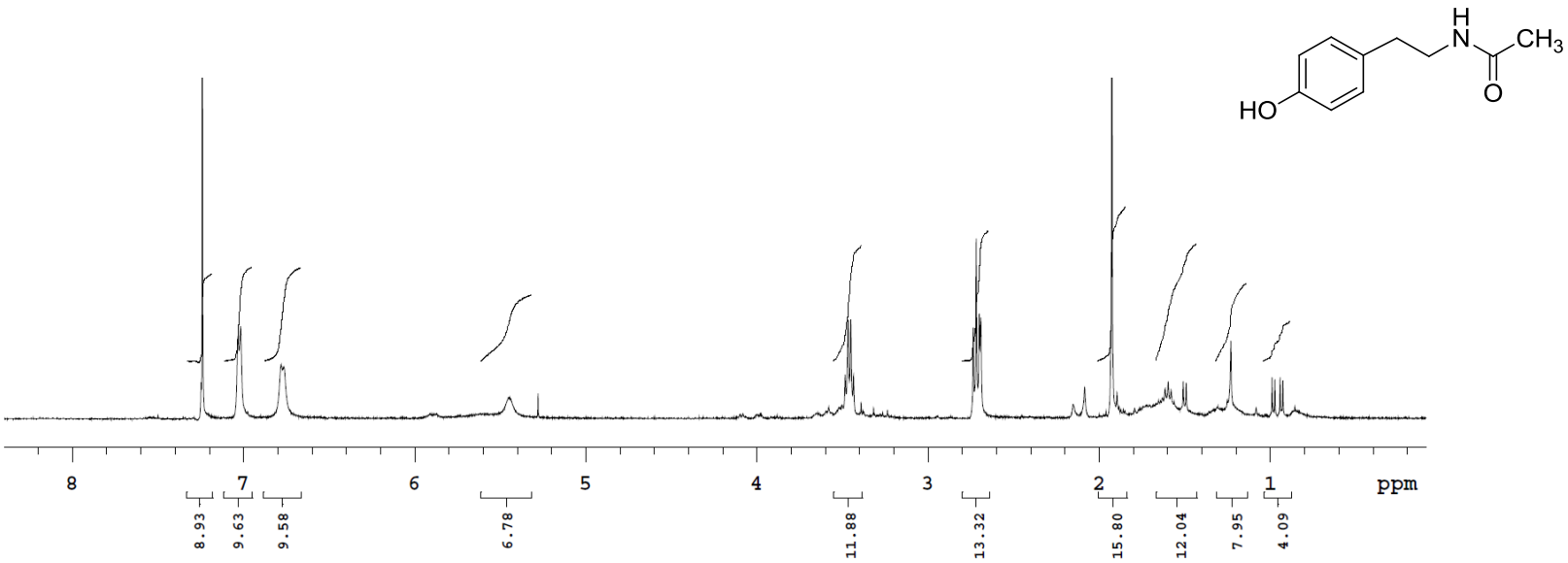


Figure S117. ^{1}H NMR spectrum ($CDCl_3$, 400 MHz) of *N*-acetyl-tyramine (**14**)

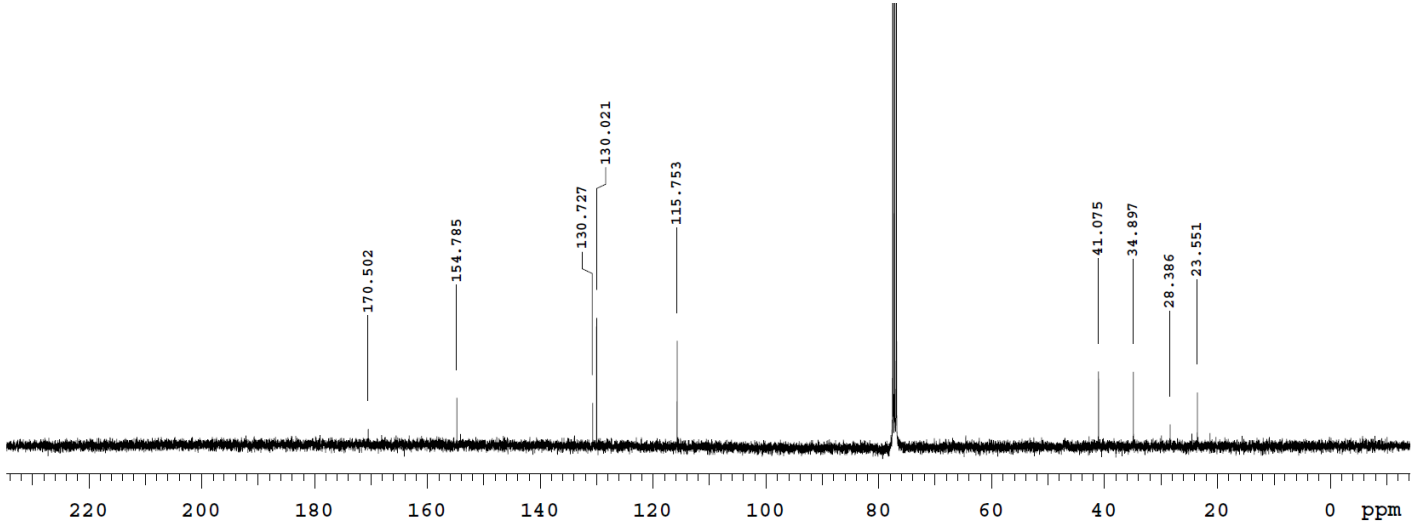


Figure S118. ^{13}C NMR spectrum ($CDCl_3$, 100 MHz) of *N*-acetyl-tyramine (**14**)

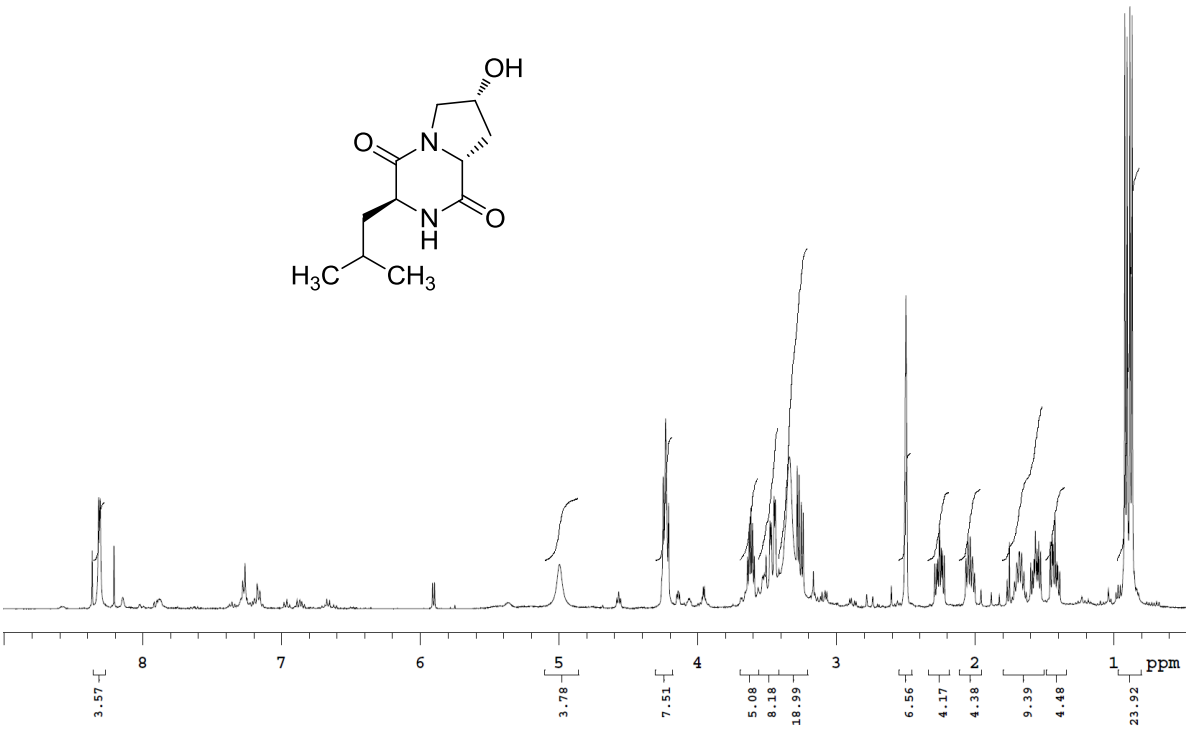


Figure S119. ^{1}H NMR spectra (DMSO-d_6, 400 MHz) of cyclo(D-cis-Hyp-L-Leu) (**15**)

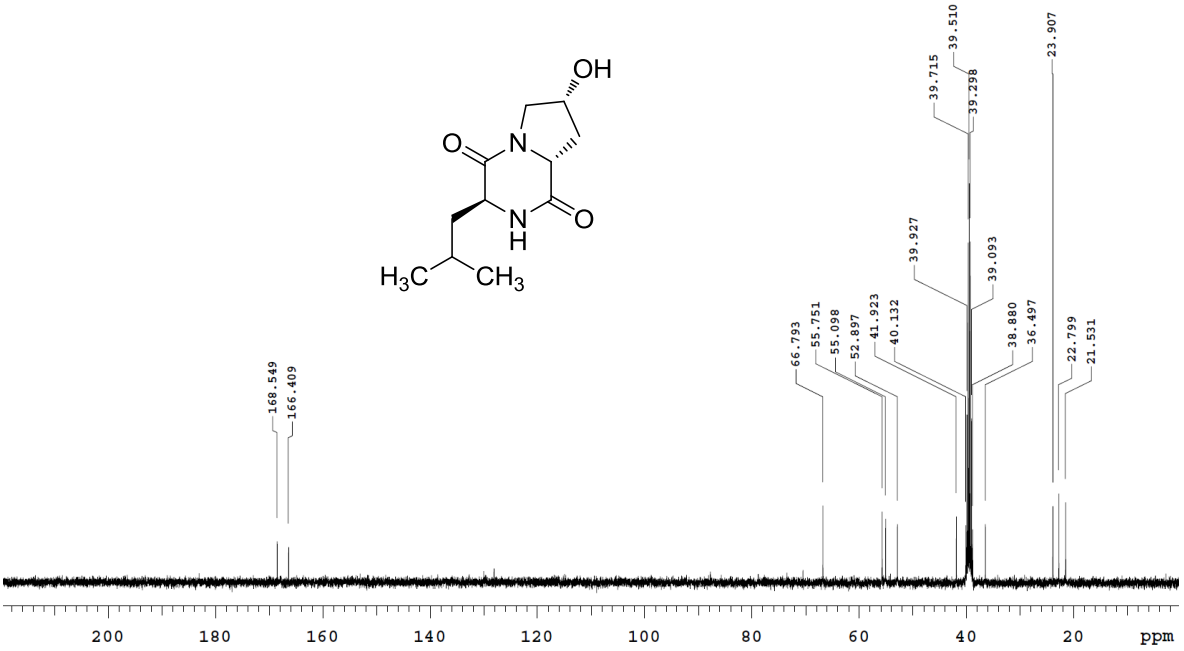


Figure S120. ^{13}C NMR spectra (DMSO-d_6, 100 MHz) of cyclo(D-cis-Hyp-L-Leu) (**15**)

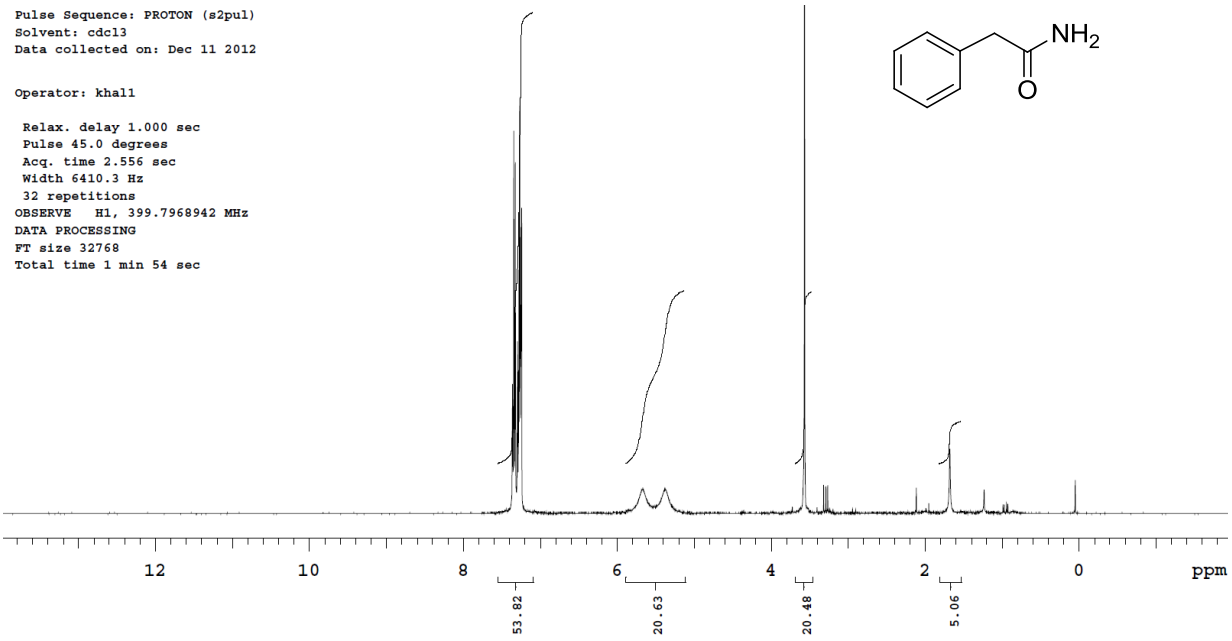


Figure S121. ^{1}H NMR spectrum ($CDCl_3$, 400 MHz) of 2-phenylacetamide (**16**)

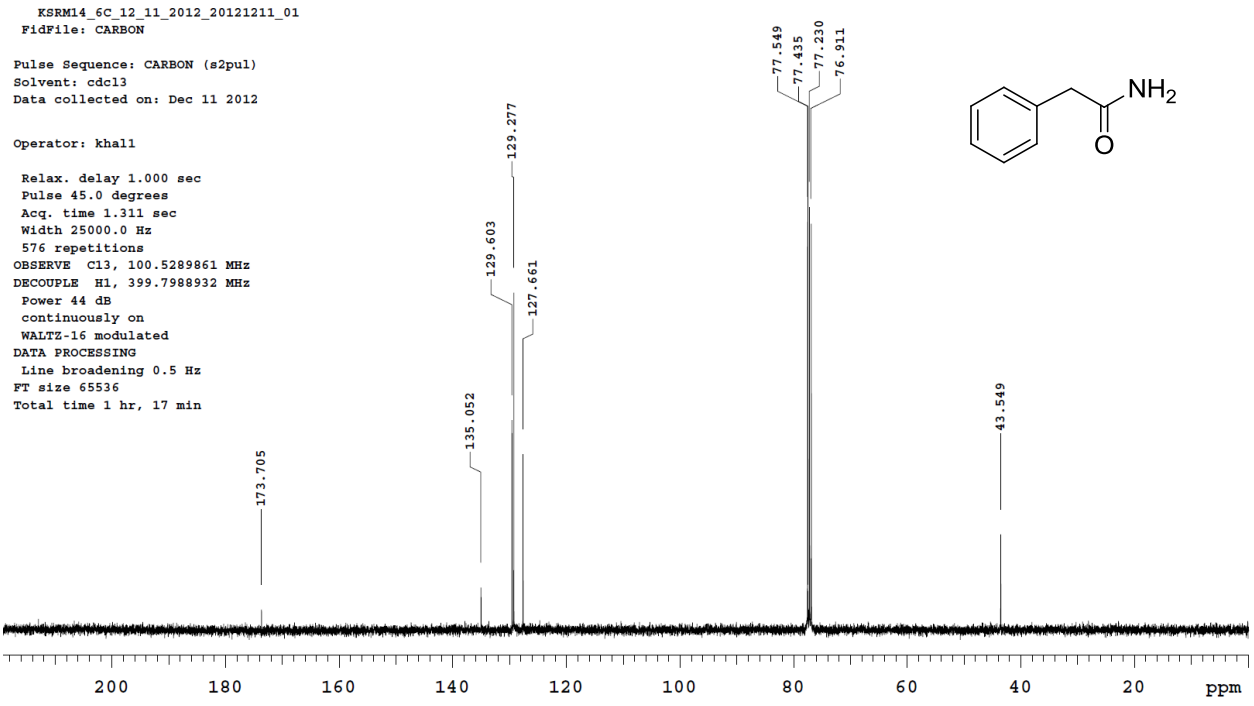


Figure S122. ^{13}C NMR spectrum ($CDCl_3$, 100 MHz) of 2-phenylacetamide (**16**)

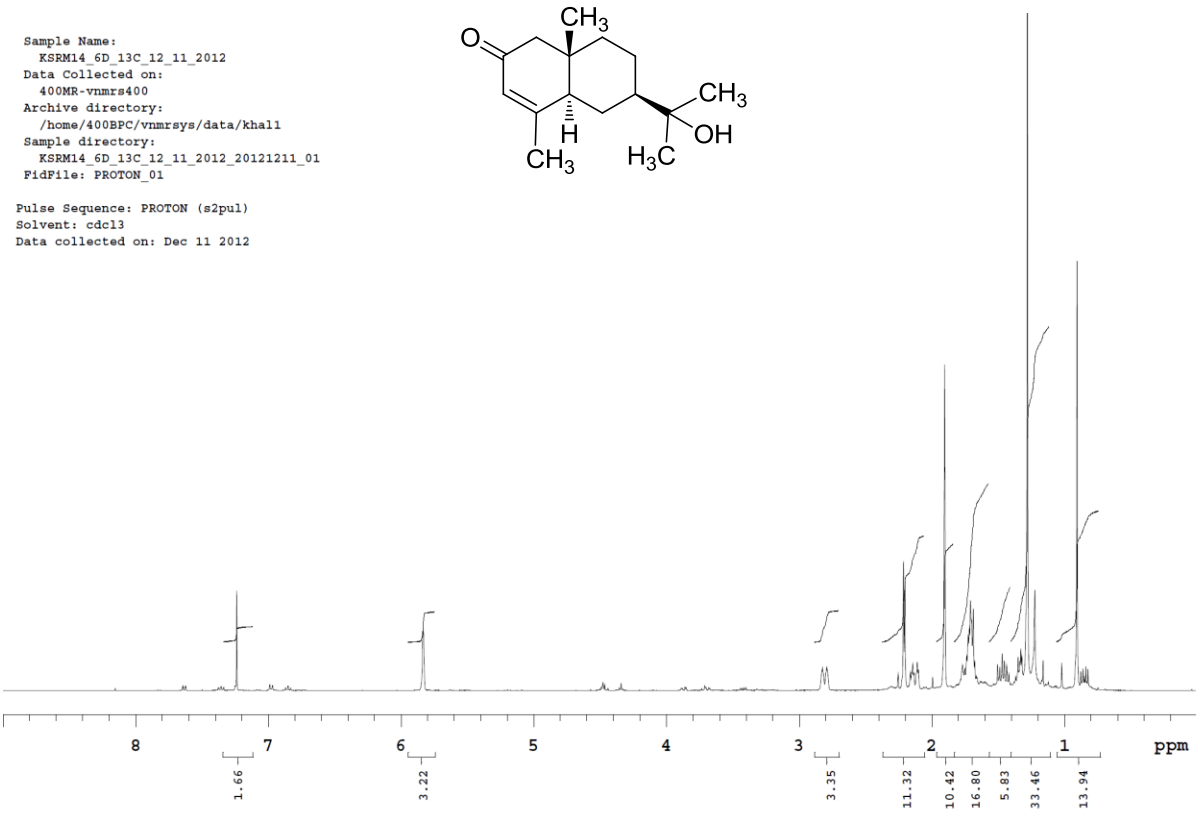


Figure S123. ^{1}H NMR spectrum ($CDCl_3$, 400 MHz) of isopterocarpolone (**17**)

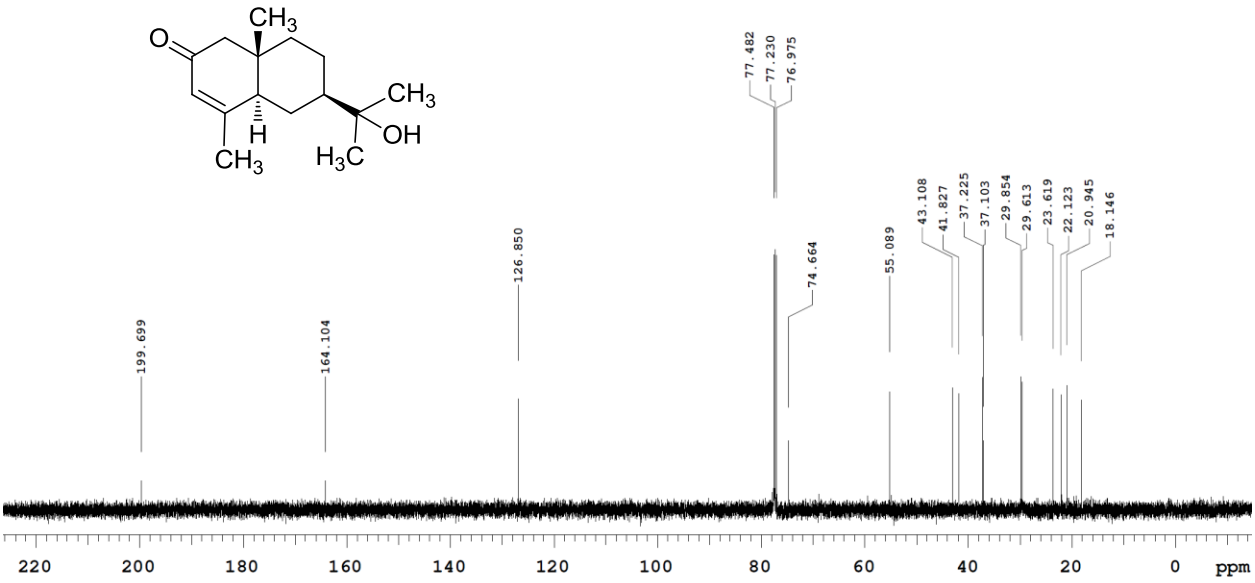


Figure S124. ^{13}C NMR spectrum ($CDCl_3$, 100 MHz) of isopterocarpolone (**17**)

Supplementary References

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