Synthesis and reactivity of precolibactin 886.

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Supporting Information

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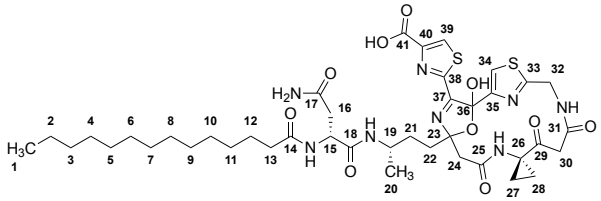
Supplementary Table 1: Comparison of ^{1}H NMR data of synthetic and natural precolibactin 886 (**1**).



precolibactin-886 (**1**)

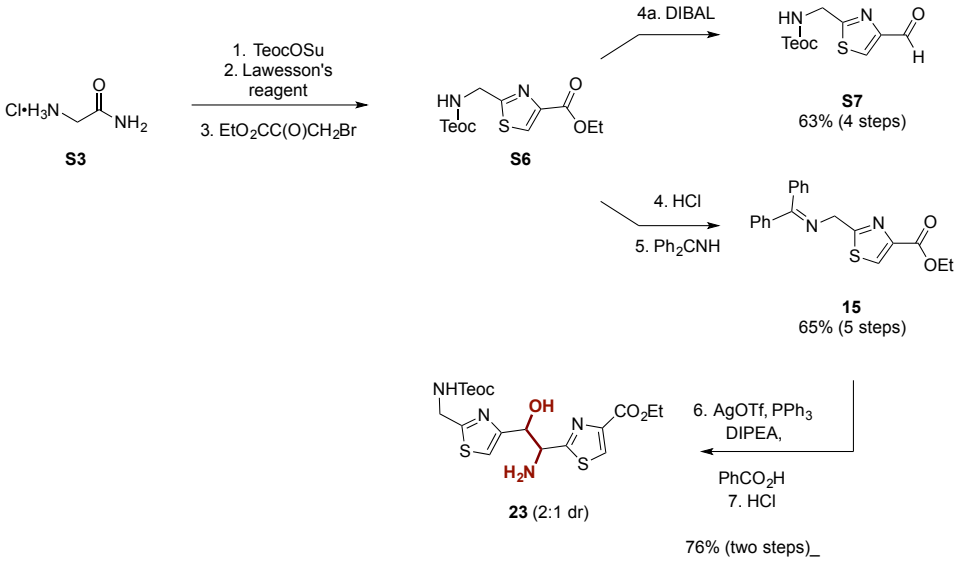
Position	δ_H (*J* in Hz)		δ_c	
	Synthetic[a]	Natural[b,c]	Synthetic[a] (HSQC/*HMBC*)[d]	Natural[b,c]
1	0.85 t (7.0)	0.84, t (6.8)	14.0 (H-1)	14.1
2	1.23, m	1.25, m	22.1 (H-2)	22.3
3	1.16–1.27, overlap	1.14–1.27, m	31.3 (H-3)	31.5
4-10	1.16–1.27, overlap	1.14–1.27, m	28.7–29.1 (H-4–H-10)	28.8–29.2
11	1.16–1.27, overlap	1.14–1.27, m	28.6 (H-11)	28.8
12	1.43, overlap	1.41, m	25.2 (H-12)	25.4
13	2.06, m	2.05, m	35.3 (H-13)	35.4
14	NH, 7.89, overlap	NH, 8.01, d (7.7)/8.03, d (7.7)	172.5 (*H-13*)	172.4/172.5
15	4.45, q (7.3)	4.46, m	49.9 (H-15)	50.2
16a	2.326, dd (15.1, 7.8)/2.332, dd (15.2, 7.8)	2.35, dd (15.3, 8.5)	37.5 (H-16)	37.7/37.8
16b	2.416, dd (15.1, 5.8)/2.421, dd (15.0, 5.6)	2.41, dd (15.3, 4.3)		
17a	NH, 6.81, s	NH, 6.84, s	171.4 (*H-15, H-16*)	171.6
17b	NH, 7.23, s	NH, 7.31, s		
18	NH, 7.55, d (8.4)/7.50, d (8.4)	NH, 7.58, d (7.7)/7.66, d (7.7)	170.3 (*H-15, H-16*)	170.5/170.6
19	3.71, m	3.70, m	44.5 (H-19)	44.7
20	0.990, d (6.5)/0.986, d (6.4)	0.99, d (6.0)	20.6 (H-20)	20.4
21a	1.42, m	1.44, m	30.3 (H-21)	30.3/30.4
21b	1.58, m	1.57, m		
22a	1.69, m/1.78, m	1.69, m/1.79, m	34.9 (H-22)	34.9/35.0
22b	1.78, m/1.89, m	1.79, m/1.87, m		
23	-	-	107.9 (*H-24*)	107.8

24a	2.62, overlap	2.60, m	45.8 (H-24)	45.8/46.1
24b	3.29, overlap	3.31, d (12.8)		
25	NH, 7.87, overlap	NH, 7.86, brs	169.8 (*H-24, NH-25*)	170.1/170.2
26	-	-	39.3 (*H-30*)	39.4
27a	0.87, m	0.87, m	n.d.	22.1
27b	1.10, m	1.10, m		
28a	0.87, m	0.87, m	n.d.	22.1
28b	1.10, m	1.10, m		
29	-	-	205.1 (*H-30*)	205.4
30a	3.10, d (14.7)	3.10, d (14.5)	48.3 (H-30)	48.4
30b	3.71, overlap	3.71, m		
31	NH, 9.00, t (6.1)	NH, 9.06, s	166.8 (*H-30, NH-31*)	166.9
32a	4.27, dd (17.2, 5.0)	4.27, d (14.5)	40.5 (H-32)	40.6
32b	4.79, dd (17.2, 7.1)	4.78, dd (17.0, 6.8)		
33	-	-	168.1 (*H-32, H-34*)	168.1
34	7.90 (s)/7.89 (s)	8.03, s	119.9 (H-34)	120.1
35	-		153.6 (*H-34*)	154.1
36	-		107.4 (*H-39*)	107.6
37	-		160.0 (*H-39*)	160.4
38	-		n.d.	156.3
39	8.16 (s)/8.15 (s)	8.17, s	n.d.	127.6
40	-		n.d.	156.1
41	-		n.d.	163.9

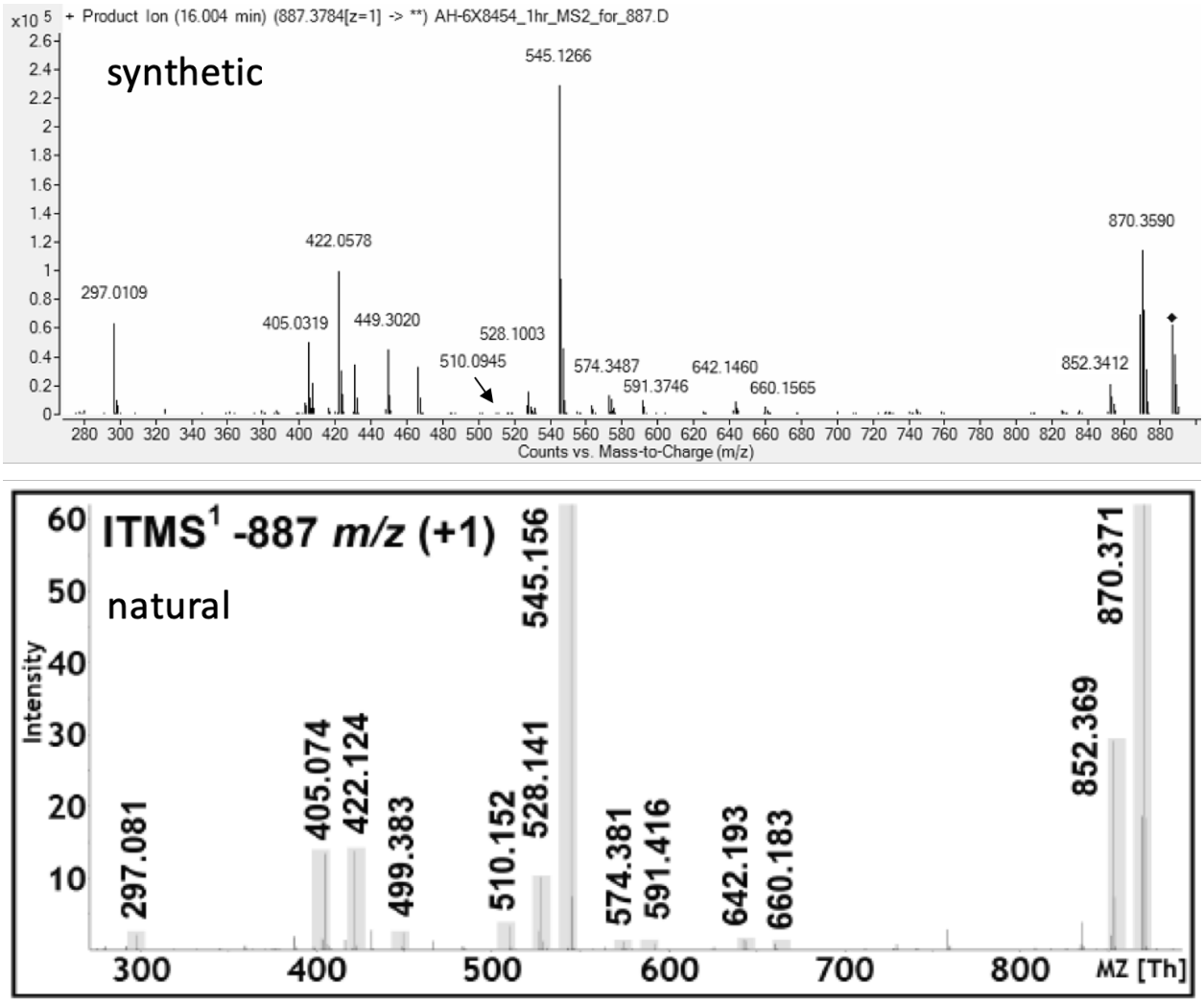
[a] NMR spectra were obtained in DMSO-d_6 at 600 MHz for ^{1}H and 150 MHz for ^{13}C. [b]NMR spectra were obtained in DMSO-d_6 at 850 MHz for ^{1}H and 212.5 MHz for ^{13}C. [c]Data for natural precolibactin-886 (**1**) were obtained from Li et al.[1] [d]Key HSQC (normal) and HMBC (italic) correlations for assignment of ^{13}C chemical shift.

Supplementary Table 2: Strain and plasmids.

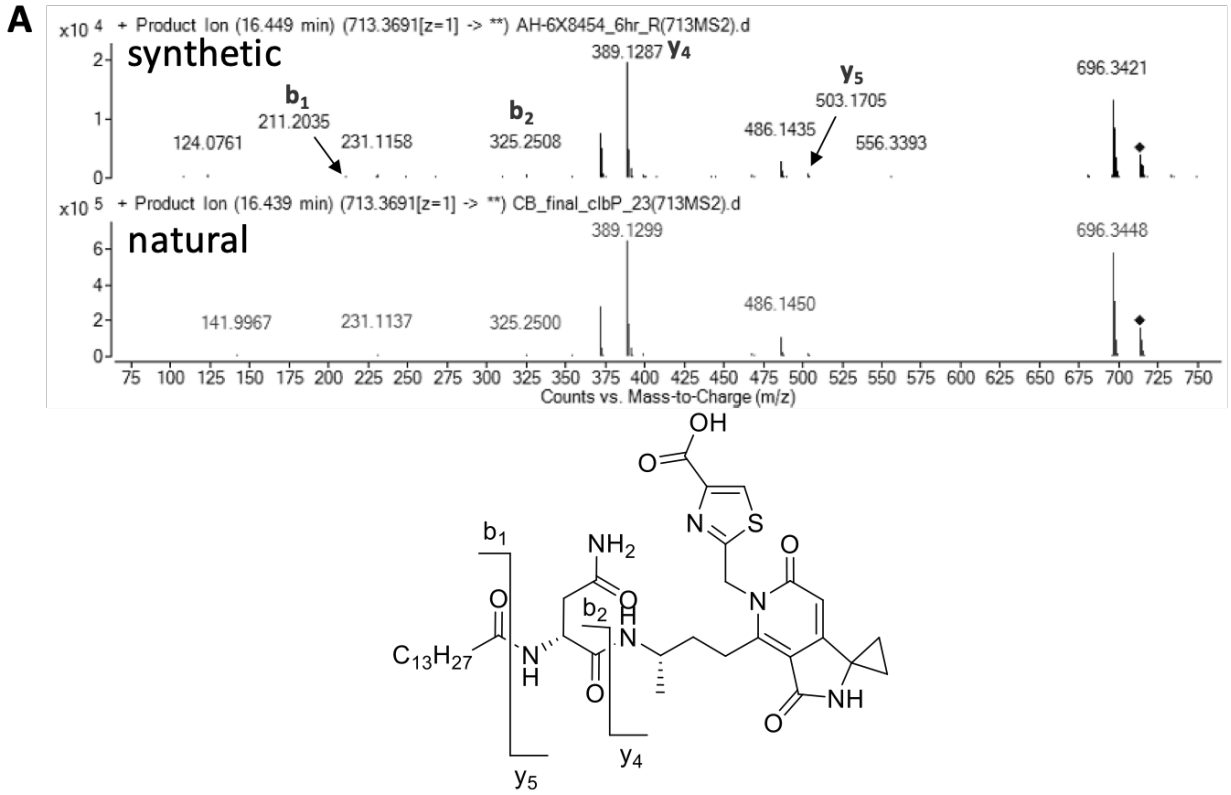
Strain	Genotype	Reference
E. coli DH10B	F- mcrA _(mcrBC-hsdRMS-mrr) [_80d_lacZ_M15] _lacX74 deoR recA1 endA1 araD139_(ara,leu)7697 galU galK _- rpsL nupG	Invitrogen
Plasmids		
pBAD18	Expression vector with araBAD promoter for induction	Guzman et al.[2]
pPEB018	*clbP* gene cloned into pBAD18	Vizcaino et al.[3]

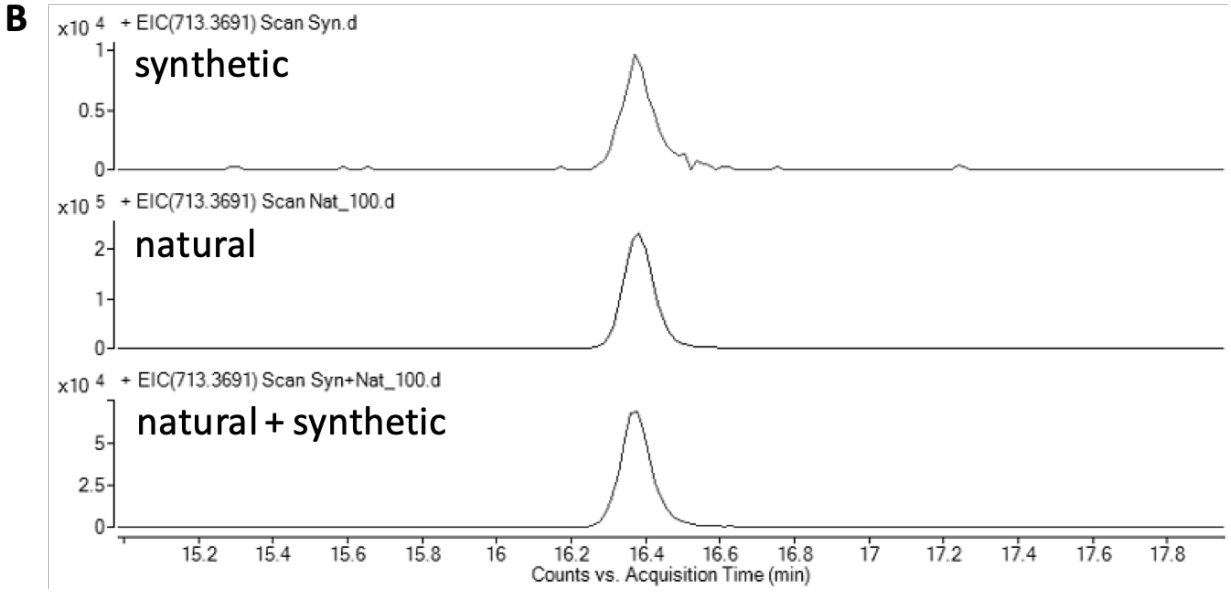


Supplementary Fig. 1. Synthesis of the aminoalcohol **23**.

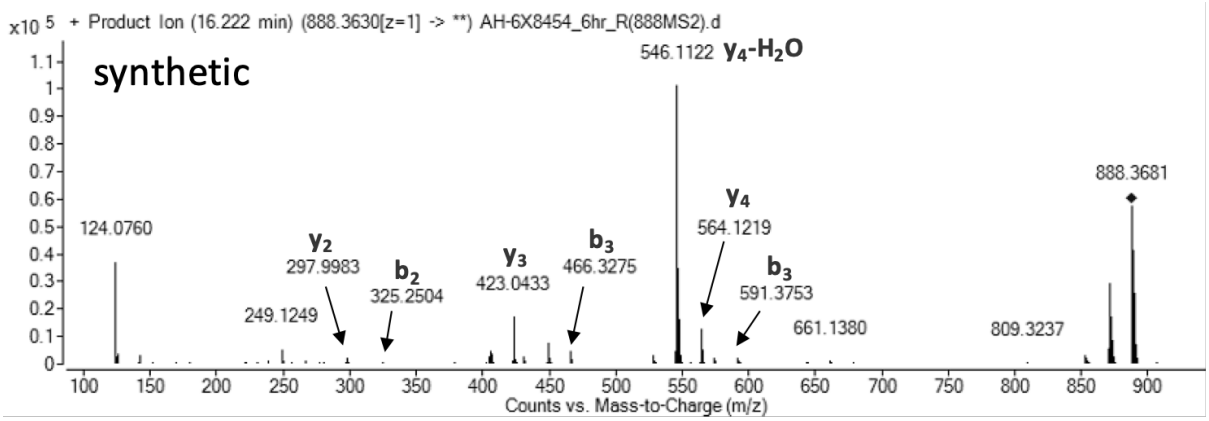


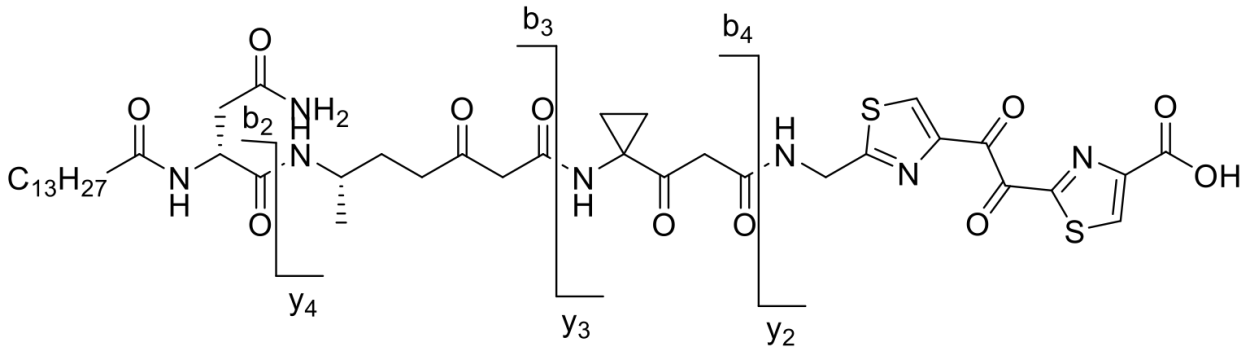
Supplementary Fig. 2. Tandem MS spectra of synthetic (top) and natural (bottom) precolibactin 886 (**1**). The tandem MS spectrum for natural precolibactin-886 (**1**) was obtained from Li et al. [1]



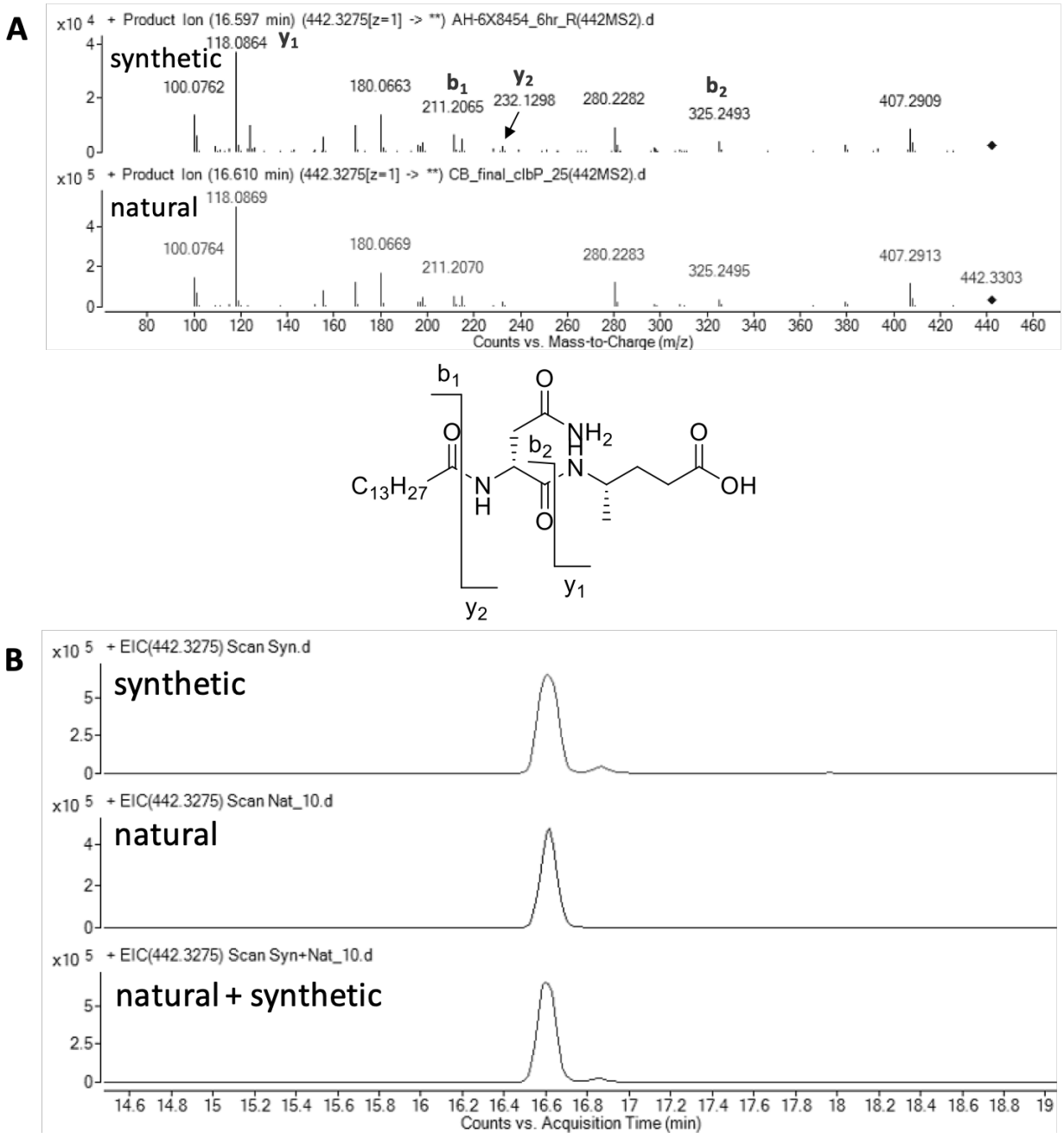


Supplementary Fig. 3. Structural identification of precolibactin B (7). A. Tandem MS spectra of synthetic (top) and natural (middle) **7**, and its fragmentation pattern (bottom). **B.** LC/HRMS analysis of precolibactin B (**7**): synthetic (top), natural (middle), and co-injection (bottom). Precolibactin B (**7**): HRMS (*m/z*): $[M + H]^+$ calcd for $C_{36}H_{53}N_6O_7S$, 713.3691; found, 713.3685.

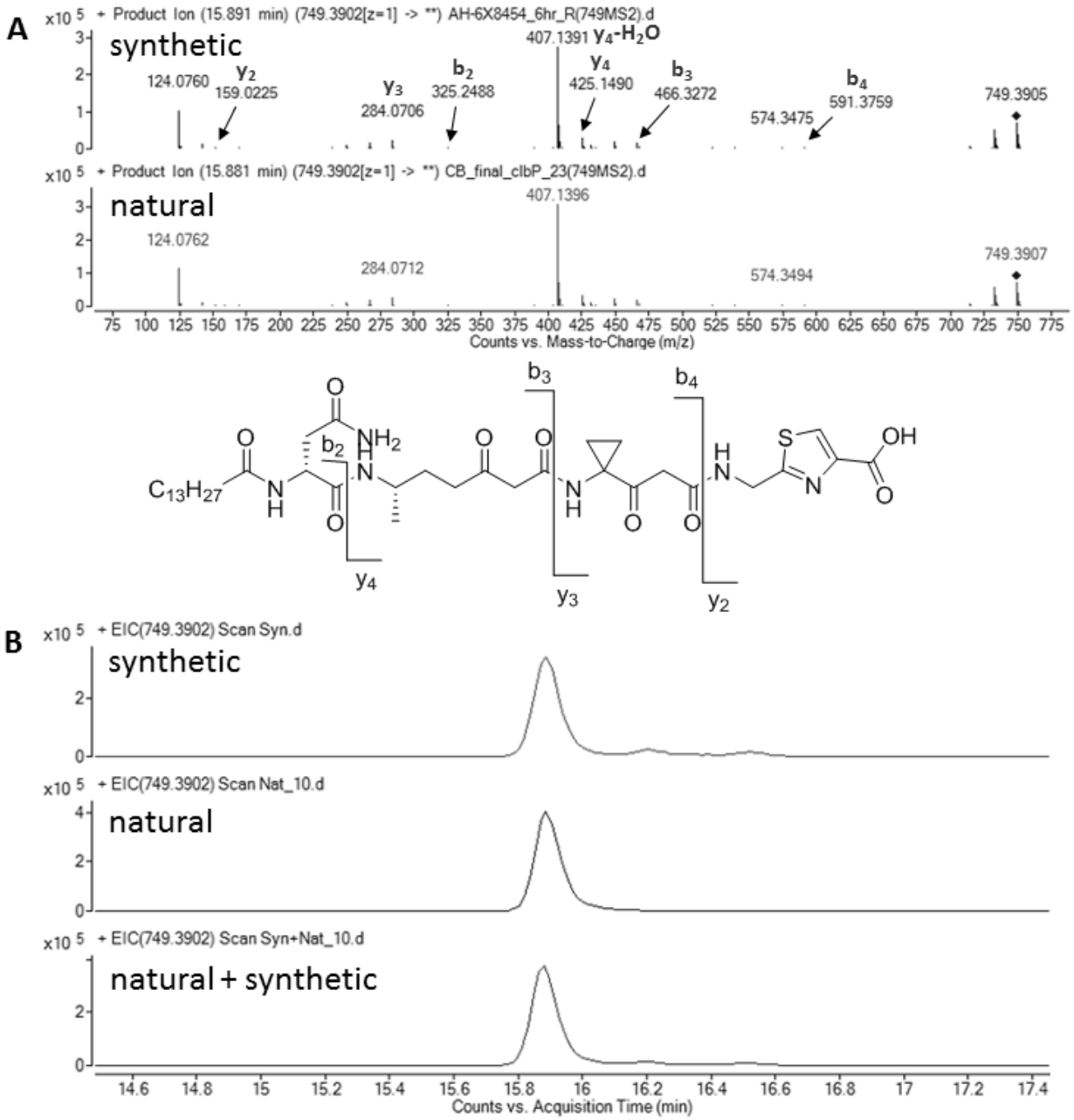




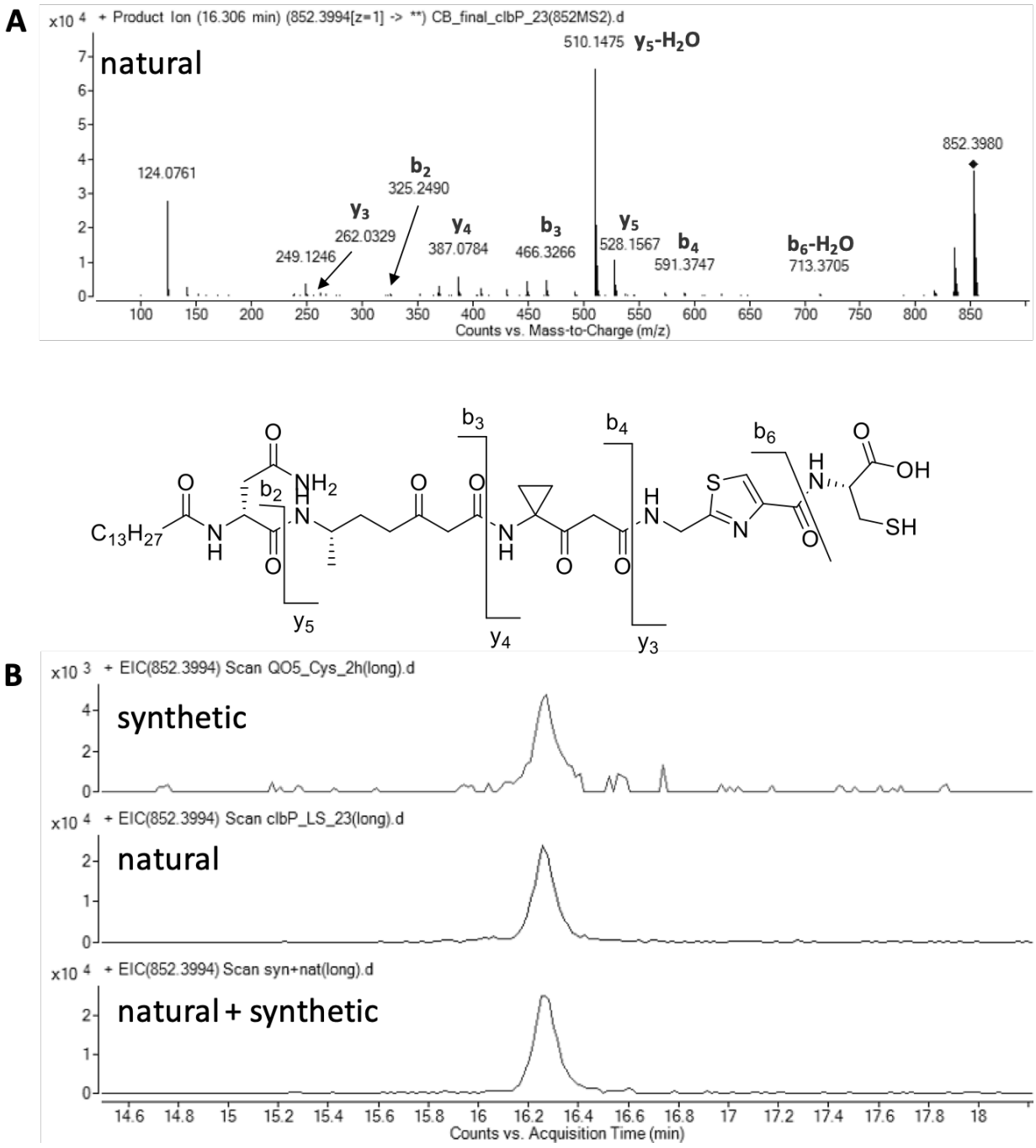
Supplementary Fig. 4. Structural identification of compound 22. Tandem MS spectra of synthetic **22** (top) and its fragmentation pattern (bottom). α-diketone **22**: HRMS (*m/z*): $[M + H]^+$ calcd for $C_{41}H_{58}N_7O_{11}S_2$, 888.3630; found, 888.3633.



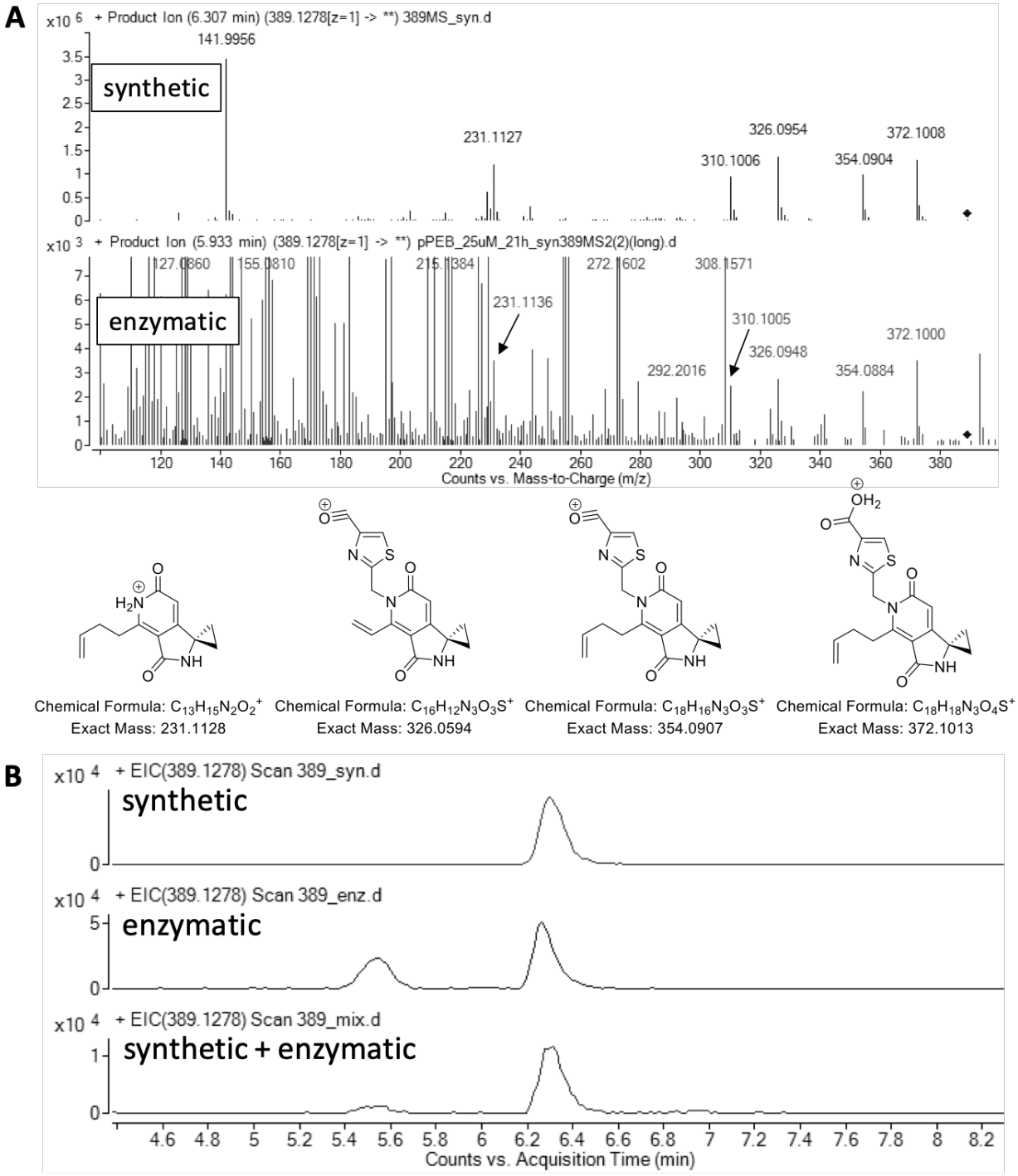
Supplementary Fig. 5. Structural identification of compound 31. A. Tandem MS spectra of synthetic (top) and natural (middle) **31**, and its fragmentation pattern (bottom). **B.** LC/HRMS analysis of synthetic (top), natural (middle), and coinjection (bottom) **31**. **31**: HRMS (*m/z*): $[M + H]^+$ calcd for $C_{23}H_{44}N_3O_5$, 442.3275; found, 442.3281.



Supplementary Fig. 6. Structural identification of compound 32. A. Tandem MS spectra of synthetic (top) and natural (middle) **32**, and its fragmentation pattern (bottom). **B.** LC/HRMS analysis of synthetic (top), natural (middle), and co-injection (bottom) **32**. **32**: HRMS (*m/z*): $[M + H]^+$ calcd for $C_{36}H_{57}N_6O_9S$, 749.3902; found, 749.3914.



Supplementary Fig. 7. Structural identification of compound 33. A. Tandem MS spectra of natural **33** (top) and its fragmentation pattern (bottom). **B.** LC/HRMS analysis of synthetic (top), natural (middle), and mixed (bottom) **33**.



Supplementary Fig. 8. Structural identification of compound 35. A. Tandem MS spectra of synthetic (top) and enzymatic (middle) **35**, and its fragmented ions (bottom). **B.** LC/HRMS analysis of synthetic (top), enzymatic (middle), and co-injection (bottom) **35**.

General Experimental Methods.

General Experimental Procedures. All reactions were performed in single-neck, flame-dried, round-bottomed flasks fitted with rubber septa under a positive pressure of nitrogen unless otherwise noted. Air- and moisture-sensitive liquids were transferred via syringe or stainless steel cannula, or were handled in a nitrogen-filled drybox (working oxygen level <10 ppm). Organic solutions were concentrated by rotary evaporation at 28–32 °C. Flash-column chromatography was performed as described by Still et al.,[4] employing silica gel (60 Å, 40–63 μm particle size) purchased from Sorbent Technologies (Atlanta, GA). Analytical thin-layered chromatography (TLC) was performed using glass plates pre-coated with silica gel (0.25 mm, 60 Å pore size) impregnated with a fluorescent indicator (254 nm). TLC plates were visualized by exposure to ultraviolet light (UV).

Materials. Commercial solvents and reagents were used as received with the following exceptions. Dichloromethane, ether and *N*,*N*-dimethylformamide were purified according to the method of Pangborn et al.[5] Triethylamine was distilled from calcium hydride under an atmosphere of argon immediately before use. Di-*iso*-propylamine was distilled from calcium hydride and was stored under nitrogen. Methanol was distilled from magnesium turnings under an atmosphere of nitrogen immediately before use. Tetrahydrofuran was distilled from sodium–benzophenone under an atmosphere of nitrogen immediately before use. Ethyl 2-(((*tert*-butoxycarbonyl)amino)methyl)thiazole-4-carboxylate (**13**), *S*-(*tert*-butyl) 3-(1-((*tert*-butoxycarbonyl)amino)cyclopropyl)-3-oxopropanethioate (**11**), and *S*-(*tert*-butyl) (*S*)-6-((*R*)-4-amino-4-oxo-2-tetradecanamidobutanamido)-3-oxoheptanethioate (**10**) were prepared according to published procedures.[6]

Instrumentation. Proton nuclear magnetic resonance spectra (^{1}H NMR) were recorded at 400, 500, or 600 MHz at 24 °C, unless otherwise noted. Chemical shifts are expressed in parts per million (ppm, δ scale) downfield from tetramethylsilane and are referenced to residual protium in the NMR solvent ($CDCl_3$, δ 7.26; CD_2HOD, δ 3.31; C_2D_5HSO, δ 2.50; $CDHCl_2$, δ 5.32). Data are represented as follows: chemical shift, multiplicity (s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet and/or multiple resonances, br = broad, app = apparent), coupling constant in Hertz, integration, and assignment. Proton-decoupled carbon nuclear magnetic resonance spectra (^{13}C NMR) were recorded at 100, 125 or 150 MHz at 24 °C, unless otherwise noted. Chemical shifts are expressed in parts per million (ppm, δ scale) downfield from tetramethylsilane and are referenced to the carbon resonances of the solvent ($CDCl_3$, δ 77.2; CD_3OD, δ 49.0; C_2D_6SO, δ 39.5; CD_2Cl_2, δ 53.8). Signals of protons and carbons were assigned, as far as possible, by using the following two dimensional NMR spectroscopy techniques: [^{1}H, ^{1}H] COSY (Correlation Spectroscopy), [^{1}H, ^{13}C] HSQC (Heteronuclear Single Quantum Coherence) and long range [^{1}H, ^{13}C] HMBC (Heteronuclear Multiple Bond Connectivity). Attenuated total reflectance Fourier transform infrared (ATR-FTIR) spectra were obtained using a Thermo Electron Corporation Nicolet 6700 FTIR spectrometer referenced to a polystyrene standard. Data are represented as follows: frequency of absorption (cm^{-1}), intensity of absorption (s = strong, m = medium, w = weak, br = broad). Analytical ultra high-performance liquid chromatography/mass spectrometry (UPLC/MS) was performed on a Waters UPLC/MS instrument equipped with a reverse-phase C_{18} column (1.7 μm particle size, 2.1 × 50 mm), dual atmospheric pressure chemical ionization (API)/electrospray (ESI) mass spectrometry detector, and photodiode array detector. Samples

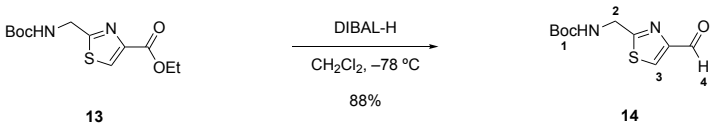
were eluted with a linear gradient of 5% acetonitrile–water containing 0.1% formic acid→100% acetonitrile containing 0.1% formic acid over 0.75 min, followed by 100% acetonitrile containing 0.1% formic acid for 0.75 min, at a flow rate of 800 μL/min. High-resolution mass spectrometry (HRMS) were obtained on either a Waters UPLC/HRMS instrument equipped with a dual API/ESI high-resolution mass spectrometry detector and photodiode array detector eluting over a reverse-phase C_{18} column (1.7 μm particle size, 2.1 × 50 mm) with a linear gradient of 5% acetonitrile–water containing 0.1% formic acid→95% acetonitrile–water containing 0.1% formic acid for 1 min, at a flow rate of 600 μL/min or an Agilent 6550A QTOF Hi Res LCMS equipped with a 1290 dual spray API source eluting over an Agilent Eclipse Plus C_{18} column (1.7 μm particle size, 4.5 × 50 mm) with a linear gradient of 5% acetonitrile–water containing 0.1% formic acid→95% acetonitrile–water containing 0.1% formic acid for 6 min, at a flow rate of 500 μL/min. Isolation of precolibactin 886 (**1**) was performed using an Agilent Prepstar HPLC system (Agilent) with a Phenomenex Luna C8 (2) 100 Å (250 × 10 mm) column (Phenomenex, Torrance, CA, USA) and a Phenomenex Luna C18 (2) 100 Å (250 × 10 mm) column (Phenomenex, Torrance, CA, USA).

Bacterial organic extract sample preparation. *E. coli* DH10B carrying pBAC *ΔclbP* was grown overnight at 37 °C with shaking (250 rpm) in LB + 12.5 μg/mL CAM. This overnight culture was inoculated into production media at a 1:200 dilution (Difco M9 + 2 mM $MgSO_4$ + 0.1 mM $CaCl_2$ + 5 g/L-casamino acids + 0.4% glucose + 12.5 μg/mL CAM + 1 g/L of each of the following amino acids: L-serine, L-cysteine, L-alanine, L-valine, and L-asparagine). Cultures were grown at 37 °C with 250 rpm shaking to an OD_{600} of 0.4-0.6. Cultures were cooled on ice for approximately 10 min before inducing with isopropyl β-D-1-galactopyranoside (IPTG) at a final concentration of 200 μM. Cultures were incubated at 25 °C for 42 h before extraction. To extract, 6.0 mL of ethyl acetate was added to the culture. Cultures were vortexed for 15-30 seconds. The layers then were separated by centrifugation (3000 rpm × 10 min). The top 5 mL of the ethyl acetate layer was removed and transferred to a glass vial. The ethyl acetate was removed in vacuo. The dried extracts were dissolved in 200 μL of methanol for LC/HRMS analysis.

Cleavage of precolibactin 886 (1) by ClbP. 5.0 μL of an overnight culture of pBAD18 (–ClbP) or pPEB018 (+ClbP) in LB + 100 μg/mL Amp was used to inoculate 1.0 mL of fresh LB + 100 μg/mL Amp. Cultures were incubated at 37 °C with 250 rpm to OD_{600} 0.4–0.6. To induce protein expression, L-arabinose (final concentration 0.01%) was added to the cultures. After another 30 min incubation, synthetic precolibactin 886 (**1**) was added (final concentration 25 μM). The cultures were extracted with 3.0 mL of *n*-butanol after 3, 8, or 21 h incubation. 2.5 mL of the organic layers were removed and dried in vacuo. The dried samples were dissolved in 50 μL of methanol for LC/HRMS analysis.

Synthetic Procedures.

*Synthesis of the aldehyde **14**.*



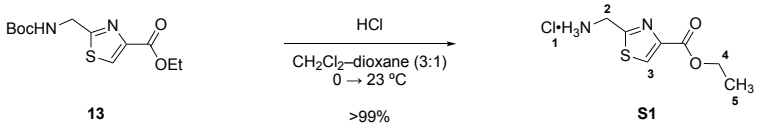
13 14

A solution of di-*iso*-butylaluminum hydride (DIBAL-H) in dichloromethane (1.0 M, 9.11 mL, 9.11 mmol, 3.00 equiv) was added dropwise via syringe to a solution of the ester **13** (870 mg, 3.04 mmol, 1 equiv) in dichloromethane (11 mL) at –78 ºC. The reaction mixture was stirred for 3 h at –78 ºC. Methanol (2.0 mL) was added dropwise to the reaction mixture. The resulting mixture was allowed to slowly warm to 23 °C over 1 h. The product mixture was diluted with saturated aqueous potassium sodium tartrate solution (30 mL). The diluted product mixture was stirred for 3 h at 23 °C and then transferred to a separatory funnel. The layers that formed were separated. The aqueous layer was extracted with dichloromethane (3 × 20 mL). The organic layers were combined and the combined organic layers were washed with saturated aqueous sodium chloride solution (30 mL). The washed organic layer was dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated. The residue obtained was purified by flash-column chromatography (eluting with 5% ethyl acetate–hexanes initially, grading to 50% ethyl acetate–hexanes) to provide the aldehyde **14** as a white solid (650 mg, 88%).

R_f = 0.42 (30% ethyl acetate–hexanes; UV). ^{1}H NMR (600 MHz, DMSO-d_6) δ 9.87 (s, 1H, H_4), 8.62 (s, 1H, H_3), 7.87 (t, J = 6.2 Hz, 1H, N*H*), 4.42 (d, J = 6.1 Hz, 2H, H_2), 1.41 (s, 9H, H_1). ^{13}C NMR (151 MHz, DMSO-d_6) δ 184.9 (CH), 172.9 (C), 155.8 (C), 154.2 (C), 132.1 (CH), 78.8 (C), 41.9 (CH_2), 28.1 (3 × CH_3). IR (ATR-FTIR), cm^{-1}: 3097 (w), 2977 (m), 2931 (w), 1696 (s), 1519 (m), 1488 (m), 1392 (w), 1367 (m), 1165 (s). HRMS-CI (m/z): [M + Na]$^+$ calcd for $C_{10}H_{14}N_2NaO_3S$, 265.0617; found, 265.0631.

*Synthesis of the benzophenone imine **15**.*

*Step 1: Synthesis of the amine **S1**.*

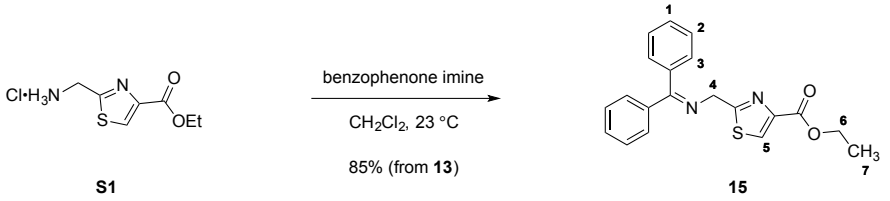


13 >99% **S1**

A solution of hydrogen chloride in 1,4-dioxane (4.0 N, 26.0 mL, 104 mmol, 10.0 equiv) was added dropwise via syringe pump over 30 min to a solution of the ester **13** (2.97 g, 10.4 mmol, 1 equiv) in dichloromethane (75 mL) at 0 °C. The resulting mixture was allowed to slowly warm to 23 °C. The reaction mixture was stirred for 16 h at 23 °C. The product mixture was concentrated to provide the amine **S1** as a white solid (2.31 g, >99%). The product **S1** obtained in this way was used directly in the following step.

R_f: Compound has no mobility on silica. ^{1}H NMR (600 MHz, DMSO-d_6) δ 8.80 (s, 3H, H_1), 8.61 (s, 1H, H_3), 4.46 (s, 2H, H_2), 4.32 (q, J = 7.1 Hz, 1H, H_4), 1.30 (t, J = 7.1 Hz, 2H, H_5). ^{13}C NMR (151 MHz, DMSO-d_6) δ 163.1 (C), 160.5 (C), 145.6 (C), 131.1 (CH), 60.9 (CH_2), 39.4 (CH_2, obscured by NMR solvent, detected indirectly by HSQC), 14.2 (CH_3).

*Step 2. Synthesis of the imine **15**.*

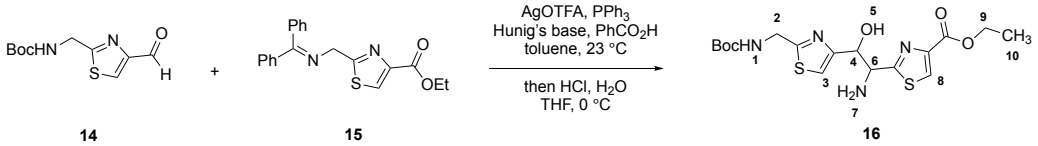


Benzophenone imine (1.81 g, 9.99 mmol, 1.10 equiv) was added to a solution of the amine **S1** (2.02 g, 9.08 mmol, 1 equiv) in dichloromethane (35 mL) at 23 °C. The resulting mixture was stirred for 16 h at 23 °C. The product mixture was filtered through a pad of Celite (2.5 × 4.5 cm). The filter cake was washed with dichloromethane (15 mL). The filtrates were combined and the combined filtrates were concentrated. The residue obtained was dissolved in ether (35 mL) and the resulting solution was filtered through a pad of Celite (2.5 × 4.5 cm). The filtrate was washed with water (35 mL). The washed organic layer was dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated to provide the imine **15** as a white solid (2.70 g, 85% from **13**).

R_f = 0.30 (20% ethyl acetate–hexanes; UV, $KMnO_4$). 1H NMR (500 MHz, DMSO-d_6) δ 8.47 (s, 1H, H_5), 7.69 – 7.62 (m, 2H, H_3), 7.61 – 7.54 (m, 3H, H_2, H_1), 7.53 – 7.48 (m, 1H, H_1), 7.49 – 7.42 (m, 2H, H_2), 7.41 – 7.14 (m, 2H, H_3), 4.77 (s, 2H, H_4), 4.27 (q, J = 7.1 Hz, 2H, H_6), 1.28 (t, J = 7.1 Hz, 3H, H_7). ^{13}C NMR (126 MHz, DMSO-d_6) δ 172.6 (C), 169.9 (C), 160.8 (C), 146.0 (C), 138.4 (C), 135.3 (C), 130.8 (CH), 129.2 (CH), 128.9 (2 × CH), 128.8 (CH), 128.4 (2 × CH), 128.2 (2 × CH), 127.3 (2 × CH), 60.6 (CH_2), 54.6 (CH_2), 14.1 (CH_3). IR (ATR-FTIR), cm^{-1}: 3063 (w), 2982 (w), 1731 (s), 1715 (s), 1236 (m), 1205 (s). HRMS-CI (m/z): $[M + H]^+$ calcd for $C_{20}H_{19}N_2O_2S$, 351.1162; found, 351.1167.

*Synthesis of the azide **17**.*

*Step 1: Synthesis of the amino alcohol **16**.*



A solution of triphenylphosphine in toluene (0.1 M, 7.00 mL, 700 μmol, 0.10 equiv) was added to a solution of silver trifluoroacetate (180 mg, 700 μmol, 0.10 equiv) in toluene (17 mL) at 23 °C. The resulting mixture was stirred for 20 min at 23 °C, with protection from light. The imine **15** (2.45 g, 7.00 mmol, 1 equiv) and the aldehyde **14** (2.54 g, 10.5 mmol, 1.50 equiv) were then added in sequence. In a separate flask, diisopropylethylamine (Hünig's base, 244 μL, 1.40 mmol, 0.20 equiv) and benzoic acid (42.7 mg, 0.35 mmol, 0.05 equiv) were dissolved in toluene (4.1 mL). The resulting solution was transferred to the flask containing the imine **15** and the aldehyde **14**. The resulting mixture was stirred for 24 h at 23 °C. The product mixture was concentrated. The residue obtained was partitioned between saturated aqueous sodium chloride solution (20 mL) and dichloromethane (20 mL). The layers that formed were separated, and the aqueous layer was extracted with dichloromethane (2 × 20 mL). The organic layers were combined and the combined organic layers were dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated.

The residue obtained was dissolved in tetrahydrofuran (80 mL) and the resulting solution was cooled to 0 °C. Aqueous hydrogen chloride solution (1.0 N, 14.0 mL, 14.0 mmol, 2.00 equiv) was then added. The reaction mixture was stirred for 1 h at 0 °C. The product mixture was partially concentrated to remove tetrahydrofuran. The partially concentrated solution was diluted with water (10 mL) and the diluted mixture was extracted with ether (3 × 20 mL). The aqueous layer was basified to pH ~8–9 by the slow addition of solid sodium hydrogen carbonate. The basified aqueous layer was extracted with dichloromethane (4 × 30 mL). The organic layers were combined and the combined organic layers were dried over sodium sulfate. The dried solution was filtered. The filtrate was concentrated to provide **16** as a yellow solid (~2:1 dr, stereochemistry not assigned). The product **16** obtained in this way was used directly in the following step.

R_f = 0.32 both diastereomers (10% methanol–dichloromethane; UV).

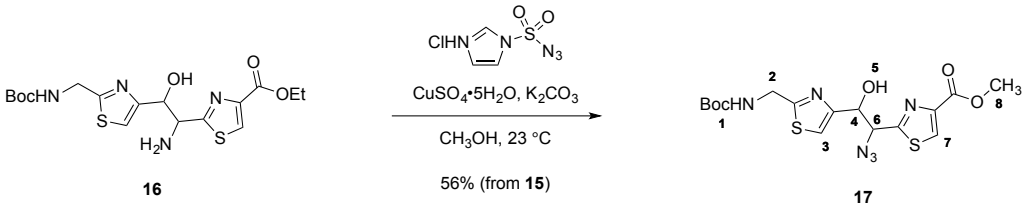
^{1}H NMR (500 MHz, DMSO-d_6, major diastereomer) δ 8.38 (s, 1H, H_8), 7.75 (t, J = 6.2 Hz, 1H, N*H*), 7.37 (s, 1H, H_3), 5.72 (d, J = 5.8 Hz, 1H, H_5), 5.42 – 5.10 (m, 1H, H_4), 4.42 (app d, J = 2.2 Hz, 1H, H_6), 4.38 (d, J = 6.2 Hz, 2H, H_2), 4.34 – 4.20 (m, 2H, H_9), 2.38 (bs, 3H, H_7), 1.42 (s, 9H, H_1), 1.34 – 1.26 (m, 3H, H_{10}). ^{13}C NMR (126 MHz, DMSO-d_6, major diastereomer) δ 178.9 (C), 170.6 (C), 161.1 (C), 157.9 (C), 155.7 (C), 146.0 (C), 128.9 (CH), 115.2 (CH), 78.5 (C), 72.5 (CH), 60.5 (CH_2), 57.3 (CH), 42.0 (CH_2), 28.2 (3 × CH_3), 14.2 (CH_3).

^{1}H NMR (500 MHz, DMSO-d_6, minor diastereomer) δ 8.33 (s, 1H, H_8), 7.70 (t, J = 6.2 Hz, 1H, N*H*), 7.24 (s, 1H, H_3), 5.86 (d, J = 5.2 Hz, 1H, H_5), 4.92 (t, J = 4.8 Hz, 1H, H_4), 4.50 (app d, J = 5.0 Hz, 1H, H_6), 4.32 (d, J = 6.1 Hz, 2H, H_2), 4.34 – 4.20 (m, 2H, H_9), 2.38 (bs, 3H, H_7), 1.40 (s, 9H, H_1), 1.34 – 1.26 (m, 3H, H_{10}). ^{13}C NMR (126 MHz, DMSO-d_6, minor diastereomer) δ 175.3

(C), 170.3 (C), 161.0 (C), 156.5 (C), 155.7 (C), 145.1 (C), 129.1 (CH), 115.9 (CH), 78.4 (C), 73.2 (CH), 60.5 (CH_2), 58.5 (CH), 41.9 (CH_2), 28.2 (3 × CH_3), 14.2 (CH_3).

IR (ATR-FTIR), cm^{-1}: 3342 (br), 2978 (m), 1710 (s), 1504 (m), 1239 (m), 1212 (m), 1164 (s). HRMS-CI (m/z): $[M + H]^+$ calcd for $C_{17}H_{25}N_4O_5S_2$, 429.1261; found, 429.1239.

*Step 2: Synthesis of the azide **17**.*



Potassium carbonate (2.42 g, 17.5 mmol, 2.50 equiv) and copper(II) sulfate pentahydrate (17.5 mg, 70.0 μmol, 0.01 equiv) were added in sequence to a solution of the amino alcohol **16** obtained in the preceding step (3.00 g, 7.00 mmol, 1 equiv) in methanol (40 mL) at 23 °C. 1*H*-Imidazole-1-sulfonyl azide hydrogen chloride (2.94 g, 14.0 mmol, 2.00 equiv) was then added. The resulting mixture was stirred for 10 h at 23 °C. The product mixture was concentrated. The residue obtained was partitioned between aqueous hydrogen chloride solution (1.0 M, 10 mL) and ethyl acetate (20 mL). The biphasic mixture was transferred to a separatory funnel and the layers that formed were separated. The aqueous layer was extracted with ethyl acetate (2 × 20 mL). The organic layers were combined and the combined organic layers were washed with saturated aqueous sodium chloride solution (30 mL). The washed organic layer was dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated. The residue obtained was purified by flash-column chromatography (eluting with hexanes initially, grading to 100% ethyl acetate) to provide the azide **17** as a colorless oil (1.73 g, 56% from **15**).

The azide **17** was isolated as an inconsequential mixture (2:1) of diastereomers.

R_f = 0.55 both diastereomers (10% methanol–dichloromethane; UV).

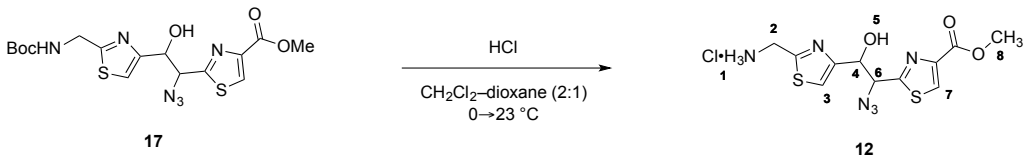
^{1}H NMR (600 MHz, DMSO-d_6, major diastereomer) δ 8.53 (s, 1H, H_7), 7.73 (t, J = 6.2 Hz, 1H, N*H*), 7.39 (s, 1H, H_3), 6.54 (d, J = 5.2 Hz, 1H, H_5), 5.37 (d, J = 4.9 Hz, 1H, H_6), 5.17 (app t, J = 5.1 Hz, 1H, H_4), 4.36 (d, J = 6.2 Hz, 2H, H_2), 3.82 (s, 3H, H_8), 1.40 (s, 9H, H_1). ^{13}C NMR (151 MHz, DMSO-d_6, major diastereomer) δ 171.1 (C), 165.4 (C), 161.1 (C), 155.7 (C), 155.2 (C), 145.1 (C), 130.7 (CH), 116.7 (CH), 78.5 (C), 71.5 (CH), 64.9 (CH), 52.1 (CH_3), 41.9 (CH_2), 28.2 (3 × CH_3).

^{1}H NMR (600 MHz, DMSO-d_6, minor diastereomer) δ 8.56 (s, 1H, H_7), 7.78 (t, J = 6.2 Hz, 1H, N*H*), 7.47 (s, 1H, H_3), 6.37 (d, J = 6.0 Hz, 1H, H_5), 5.37 (d, J = 3.3 Hz, 1H, H_6), 5.23 (ddd, J = 6.1, 3.6, 1.1 Hz, 1H, H_4), 4.30 (dd, J = 6.2, 3.3 Hz, 2H, H_2), 3.84 (s, 3H, H_8), 1.41 (s, 9H, H_1). ^{13}C NMR (151 MHz, DMSO-d_6, minor diastereomer) δ 171.3 (C), 167.6 (C), 161.1 (C), 155.9 (C), 155.7 (C), 145.4 (C), 130.2 (CH), 116.4 (CH), 78.5 (C), 72.3 (CH), 65.4 (CH), 52.1 (CH_3), 42.0 (CH_2), 28.2 (3 × CH_3).

IR (ATR-FTIR), cm^{-1}: 3345 (br), 2929 (w), 2115 (m), 2105 (s), 1717 (s), 1506 (m), 1247 (s), 1165 (m), 1043 (m), 761 (w). HRMS-CI (m/z): $[M + H]^+$ calcd for $C_{16}H_{20}N_6O_5S_2$, 441.1009; found, 441.1030.

*Synthesis of the ester **18**.*

*Step 1: Synthesis of the amine **12**.*



A solution of hydrogen chloride in 1,4-dioxane (4.0 N, 14.0 mL, 56.0 mmol, 13.2 equiv) was added dropwise via syringe pump over 30 min to a solution of the ester **17** (1.87 g, 4.25 mmol, 1 equiv) in dichloromethane (28 mL) at 0 °C. The resulting mixture was allowed to slowly warm to 23 °C. The reaction mixture was stirred for 3 h at 23 °C. The product mixture was concentrated to provide the amine **12** as a white solid (2.31 g, >99%). The product **12** obtained in this way was used directly in the following step.

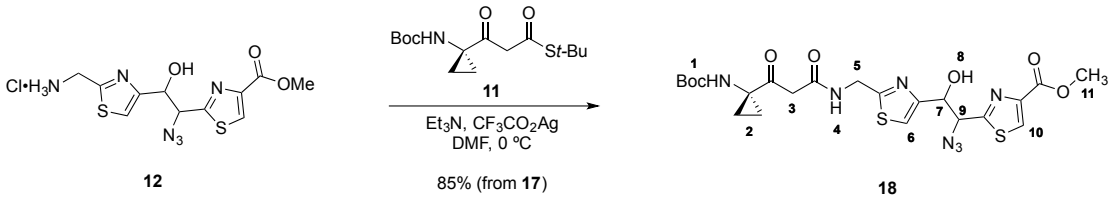
The amine **12** was isolated as an inconsequential mixture (2:1) of diastereomers.

R_f: Compound has no mobility on silica.

^{1}H NMR (600 MHz, DMSO-d_6, major diastereomer) δ 8.68 (bs, 3H, H_1), 8.60 (s, 1H, H_7), 7.69 (d, J = 1.1 Hz, 1H, H_3), 5.44 (d, J = 3.0 Hz, 1H, H_6), 5.30 (dd, J = 3.1, 1.2 Hz, 1H, H_4), 4.42 (app dp, J = 5.8, 2.9 Hz, 2H, H_2), 3.85 (s, 3H, H_8). ^{13}C NMR (151 MHz, DMSO-d_6, major diastereomer) δ 167.4 (C), 162.2 (C), 161.1 (C), 156.0 (C), 145.4 (C), 130.4 (CH), 118.6 (CH), 72.3 (CH), 66.4 (CH_2), 65.3 (CH), 52.2 (CH_3).

^{1}H NMR (600 MHz, DMSO-d_6, minor diastereomer) δ 8.63 (bs, 3H, H_1), 8.55 (s, 1H, H_7), 7.60 (d, J = 0.9 Hz, 1H, H_3), 5.40 (d, J = 4.8 Hz, 1H, H_4), 5.25 (dd, J = 4.8, 0.9 Hz, 1H, H_4), 4.35 (app dq, J = 7.9, 5.8 Hz, 2H, H_2), 3.82 (s, 3H, H_8). ^{13}C NMR (151 MHz, DMSO-d_6, minor diastereomer) δ 165.0 (C), 161.8 (C), 161.1 (C), 155.3 (C), 145.1 (C), 130.9 (CH), 119.2 (CH), 71.4 (CH), 66.4 (CH_2), 65.0 (CH), 52.1 (CH_3).

*Step 2: Synthesis of the ester **18**.*



Silver trifluoroacetate (1.50 g, 6.79 mmol, 1.60 equiv) was added to a solution of triethylamine (2.37 mL, 17.0 mmol, 4.00 equiv) and the amine **12** obtained in the preceding step (nominally 4.25 mmol, 1 equiv) in *N*,*N*-dimethylformamide (25 mL) at 0 °C. A solution of the β-ketothioester **11** (1.74 g, 5.52 mmol, 1.30 equiv) in *N*,*N*-dimethylformamide (10 mL) was then added dropwise via syringe to the reaction mixture. The reaction vessel was covered with foil to exclude light. The reaction mixture was stirred for 1 h at 0 °C. The heterogeneous product mixture was diluted with methanol (40 mL) and the diluted solution was filtered through a fritted funnel. The filtrate was collected and concentrated. The residue obtained was dissolved in ethyl acetate (60 mL) and the resulting solution was transferred to a separatory funnel. The organic layer was washed sequentially with saturated aqueous ammonium chloride solution (20 mL), saturated aqueous sodium hydrogen carbonate solution (20 mL), and saturated aqueous sodium chloride solution (20 mL). The washed organic layer was dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated. The residue obtained was purified by flash-column chromatography (eluting with 60% ethyl acetate–hexanes initially, grading to 100% ethyl acetate) to provide the ester **18** as a white solid (2.05 g, 85%).

The ester **18** was isolated as an inconsequential mixture (2:1) of diastereomers.

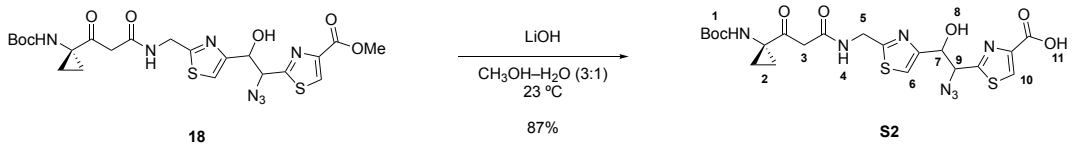
R_f = 0.41 both diastereomers (10% methanol–dichloromethane; UV).

^{1}H NMR (600 MHz, DMSO-d_6, major diastereomer) δ 8.88 (t, J = 6.0 Hz, 1H, H_4), 8.56 (s, 1H, H_{10}), 7.76 (bs, 1H, N*H*), 7.50 (d, J = 1.0 Hz, 1H, H_6), 6.38 (d, J = 6.0 Hz, 1H, H_8), 5.38 (d, J = 3.6 Hz, 1H, H_9), 5.24 (ddd, J = 6.0, 3.6, 1.1 Hz, 1H, H_7), 4.52 (d, J = 6.0 Hz, 2H, H_5), 3.84 (s, 3H, H_{11}), 3.55 (s, 2H, H_3), 1.41 (s, 9H, H_1), 1.40 – 1.33 (m, 2H, H_2), 1.11 – 1.01 (m, 2H, H_2). ^{13}C NMR (151 MHz, DMSO-d_6, major diastereomer) δ 204.7 (C), 169.6 (C), 167.6 (C), 166.7 (C), 161.1 (C), 156.0 (C), 155.8 (C), 145.4 (C), 130.2 (CH), 116.8 (CH), 78.6 (C), 72.3 (CH), 65.4 (CH), 52.1 (CH_3), 46.1 (CH_2), 41.2 (C), 40.5 (CH_2), 28.2 (3 × CH_3), 19.5 (2 × CH_2).

^{1}H NMR (600 MHz, DMSO-d_6, minor diastereomer) δ 8.85 (t, J = 6.1 Hz, 1H, H_4), 8.53 (s, 1H, H_{10}), 7.75 (bs, 1H, N*H*), 7.41 (d, J = 0.8 Hz, 1H, H_6), 6.55 (d, J = 5.2 Hz, 1H, H_8), 5.37 (d, J = 5.0 Hz, 1H, H_9), 5.18 (td, J = 5.1, 0.9 Hz, 1H, H_7), 4.46 (d, J = 6.1 Hz, 2H, H_5), 3.82 (s, 3H, H_{11}), 3.53 (s, 2H, H_3), 1.40 (s, 9H, H_1), 1.40 – 1.33 (m, 2H, H_2), 1.11 – 1.01 (m, 2H, H_2). ^{13}C NMR (151 MHz, DMSO-d_6, minor diastereomer) δ 204.7 (C), 169.4 (C), 166.6 (C), 165.4 (C), 161.1 (C), 156.0 (C), 155.0 (C), 145.1 (C), 130.7 (CH), 117.2 (CH), 78.6 (C), 71.5 (CH), 65.0 (CH), 52.1 (CH_3), 46.1 (CH_2), 41.2 (C), 40.5 (CH_2), 28.2 (3 × CH_3), 19.5 (2 × CH_2).

IR (ATR-FTIR), cm^{-1}: 3327 (br), 2976 (w), 2109 (s), 1706 (s), 1506 (m), 1248 (m), 1166 (m), 1096 (m), 756 (w). HRMS-CI (m/z): $[M + H]^+$ calcd for $C_{22}H_{28}N_7O_7S_2$, 566.1486; found, 566.1516.

*Synthesis of the acid **S2**.*



18 S2

Lithium hydroxide monohydrate (185 mg, 4.42 mmol, 5.00 equiv) was added to a solution of the ester **18** (500 mg, 0.88 mmol, 1 equiv) in methanol–water (3:1 v/v, 12 mL) at 23 °C. The resulting mixture was stirred for 3 h at 23 °C. The product mixture was partially concentrated to remove methanol. The partially concentrated solution was diluted with water (5.0 mL). The diluted solution was transferred to a separatory funnel and extracted with ethyl acetate (10 mL). The aqueous layer was acidified by the slow addition of 10% aqueous citric acid solution (w/v, 10 mL). The acidified mixture was extracted with ethyl acetate (3 × 30 mL). The organic layers were combined and the combined organic layers were dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated. The residue obtained was purified by flash-column chromatography (eluting with dichloromethane initially, grading to 6% methanol–dichloromethane) to provide the acid **S2** as a white solid (425 g, 87%).

The acid **S2** was isolated as an inconsequential mixture (2.5:1) of diastereomers.

R_f: Compound has no mobility on silica.

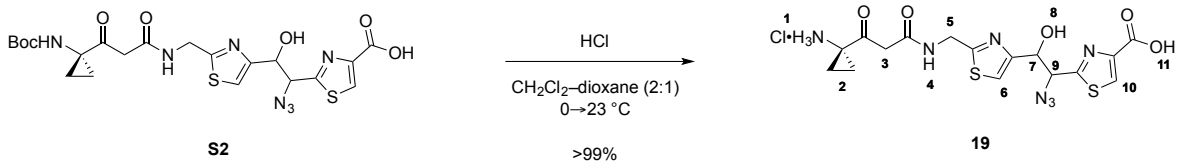
^{1}H NMR (600 MHz, DMSO-d_6, major diastereomer) δ 13.07 (s, 1H, H_{11}), 8.89 (t, J = 6.1 Hz, 1H, H_4), 8.46 (s, 1H, H_{10}), 7.76 (bs, 1H, N*H*), 7.50 (d, J = 1.1 Hz, 1H, H_6), 6.38 (bs, 1H, H_8), 5.36 (d, J = 3.5 Hz, 1H, H_9), 5.25 (d, J = 3.5 Hz, 1H, H_7), 4.53 (d, J = 6.0 Hz, 2H, H_5), 3.55 (s, 2H, H_3), 1.41 (s, 9H, H_1), 1.40 – 1.33 (m, 2H, H_2), 1.07 (app q, J = 4.3 Hz, 2H, H_2). ^{13}C NMR (151 MHz, DMSO-d_6, major diastereomer) δ 204.7 (C), 169.6 (C), 167.1 (C), 166.7 (C), 162.0 (C), 156.1 (C), 155.8 (C), 146.9 (C), 129.6 (CH), 116.8 (CH), 78.6 (C), 72.3 (CH), 65.4 (CH), 46.1 (CH_2), 41.2 (C), 40.5 (CH_2), 28.2 (3 × CH_3), 19.5 (2 × CH_2).

^{1}H NMR (600 MHz, DMSO-d_6, minor diastereomer) δ 13.07 (s, 1H, H_{11}), 8.85 (t, J = 6.1 Hz, 0H, H_4), 8.43 (s, 1H, H_{10}), 7.75 (bs, 1H, N*H*), 7.42 (d, J = 0.8 Hz, 1H, H_6), 6.53 (bs, 1H, H_8), 5.34 (d, J = 5.1 Hz, 1H, H_9), 5.17 (d, J = 5.1 Hz, 1H, H_7), 4.47 (app dd, J = 6.0, 2.1 Hz, 2H, H_5), 3.53 (s, 2H, H_3), 1.40 (s, 9H, H_1), 1.40 – 1.33 (m, 2H, H_2), 1.07 (app q, J = 4.3 Hz, 2H, H_2). ^{13}C NMR (151 MHz, DMSO-d_6, minor diastereomer) δ 204.7 (C), 169.4 (C), 166.6 (C), 165.0 (C), 162.0 (C), 156.1 (C), 155.1 (C), 146.5 (C), 130.1 (CH), 117.2 (CH), 78.6 (C), 71.5 (CH), 65.0 (CH), 46.1 (CH_2), 41.2 (C), 40.5 (CH_2), 28.2 (3 × CH_3), 19.5 (2 × CH_2).

IR (ATR-FTIR), cm^{-1}: 3343 (br), 2925 (m), 2852 (w), 2110 (m), 1973 (w), 1700 (s), 1653 (s), 1506 (m), 1259 (s), 1070 (m), 1023 (m), 797 (s), 696 (w). HRMS-CI (m/z): $[M + H]^+$ calcd for $C_{21}H_{26}N_7O_7S_2$, 552.1330; found, 552.1338.

*Synthesis of the ß-ketoamide **20**.*

*Step 1: Synthesis of the amine **19**.*



S2 >99% **19**

A solution of hydrogen chloride in 1,4-dioxane (4.0 N, 650 µL, 2.60 mmol, 14.4 equiv) was added dropwise via syringe pump over 20 min to a solution of the acid **S2** (100 mg, 180 µmol, 1 equiv) in dichloromethane (1.3 mL) at 0 °C. The resulting mixture was allowed to slowly warm to 23 °C. The reaction mixture was stirred for 3 h at 23 °C. The product mixture was concentrated to provide the amine **19** as a white solid (88.5 mg, >99%). The product **19** obtained in this way was used directly in the following step.

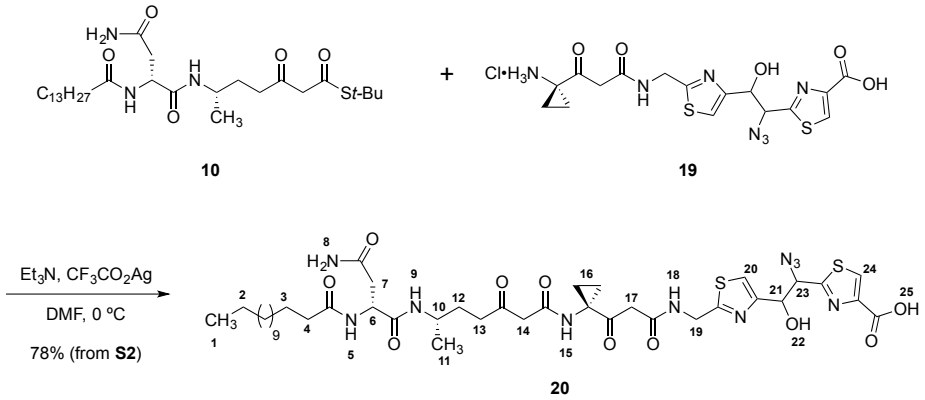
Amine **19** was isolated as an inconsequential mixture (2:1) of diastereomers.

R_f : Compounds has no mobility on silica.

^{1}H NMR (600 MHz, DMSO-d_6, major diastereomer) δ 9.10 (t, J = 6.0 Hz, 1H, H_4), 8.87 (s, 3H, H_1), 8.50 (s, 1H, H_{10}), 7.53 (d, J = 1.0 Hz, 1H, H_6), 5.35 (dd, J = 4.3, 2.5 Hz, 1H, H_9), 5.25 (dd, J = 3.4, 1.1 Hz, 1H, H_7), 4.54 (d, J = 6.0 Hz, 2H, H_5), 3.38 (s, 2H, H_3), 1.78 – 1.73 (m, 2H, H_2), 1.57 – 1.52 (m, 2H, H_2). ^{13}C NMR (151 MHz, DMSO-d_6, major diastereomer) δ 199.4 (C), 169.0 (C), 167.1 (C), 165.7 (C), 162.0 (C), 155.9 (C), 146.8 (C), 129.7 (CH), 116.9 (CH), 72.2 (CH), 65.4 (CH), 42.5 (CH_2), 42.0 (C), 40.5 (CH_2), 13.1 (CH_2).

^{1}H NMR (600 MHz, DMSO-d_6, minor diastereomer) δ 9.06 (t, J = 6.0 Hz, 1H, H_4), 8.87 (s, 3H, H_1), 8.44 (s, 1H, H_{10}), 7.43 (d, J = 0.9 Hz, 1H, H_6), 5.35 (dd, J = 4.3, 2.5 Hz, 1H, H_9), 5.19 (dd, J = 5.0, 0.9 Hz, 1H, H_7), 4.48 (d, J = 6.0 Hz, 2H, H_5), 3.36 (s, 2H, H_3), 1.78 – 1.73 (m, 2H, H_2), 1.57 – 1.52 (m, 2H, H_2). ^{13}C NMR (151 MHz, DMSO-d_6, minor diastereomer) δ 199.3 (C), 168.8 (C), 167.1 (C), 165.7 (C), 162.0 (C), 155.0 (C), 146.5 (C), 130.2 (CH), 117.3 (CH), 71.5 (CH), 65.0 (CH), 42.4 (CH_2), 42.0 (C), 40.5 (CH_2), 13.1 (CH_2).

*Step 2: Synthesis of the β-ketoamide **20**.*



10 19 20

Silver trifluoroacetate (54.8 mg, 250 μmol, 1.50 equiv) was added to a solution of triethylamine (92.3 μL, 180 μmol, 4.00 equiv) and the amine **19** obtained in the preceding step (nominally 180 μmol, 1.10 equiv) in *N*,*N*-dimethylformamide (1.0 mL) at 0 °C. A solution of the β-ketothioester **10** (92.0 mg, 170 μmol, 1 equiv) in *N*,*N*-dimethylformamide (1.0 mL) was then added dropwise via syringe. The reaction vessel was covered with foil to exclude light. The reaction mixture was stirred for 1 h at 0 °C. The heterogeneous product mixture was diluted with aqueous hydrogen chloride solution (1.0 N, 10 mL). The precipitate that formed was isolated by filtration and washed with water (5.0 mL). The washed precipitate was purified by flash-column chromatography (Si-Cyano; eluting with dichloromethane initially, grading to 6% methanol–dichloromethane) to provide the acid **20** as a white solid (118 mg, 78%).

The acid **20** was isolated as an inconsequential mixture (2:1) of diastereomers.

R_f: Compound does not move on silica.

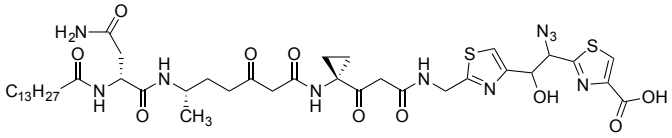
^{1}H NMR (600 MHz, DMSO-d_6, major diastereomer) δ 8.82 – 8.79 (m, 2H, H_{18}, H_{15}), 8.45 (s, 1H, H_{24}), 7.86 (d, J = 7.8 Hz, 1H, H_5), 7.49 (s, 1H, H_{20}), 7.48 (d, J = 8.3 Hz, 1H, H_9), 7.25 (bs, 1H, H_8), 6.83 (bs, 1H, H_8), 5.35 (d, J = 3.5 Hz, 1H, H_{21}), 5.25 (d, J = 3.4 Hz, 1H, H_{23}), 4.53 (d, J = 6.0 Hz, 2H, H_{19}), 4.43 (td, J = 7.6, 5.9 Hz, 1H, H_6), 3.77 – 3.65 (m, 1H, H_{10}), 3.59 (s, 2H, H_{17}), 3.35 (s, 2H, H_{14}), 2.54 – 2.51 (m, 1H, H_{13}), 2.50 – 2.44 (m, 1H, H_{13}), 2.43 (dd, J = 15.1, 5.9 Hz, 1H, H_7), 2.36 (dd, J = 15.3, 7.7 Hz, 1H, H_7), 2.10 (t, J = 7.5 Hz, 2H, H_4), 1.65 – 1.56 (m, 1H, H_{12}), 1.53 – 1.49 (m, 1H, H_{12}), 1.51 – 1.42 (m, 2H, H_3), 1.42 – 1.36 (m, 2H, H_{16}), 1.29 – 1.24 (m, 2H, H_2), 1.24 – 1.21 (m, 18H, myristoyl), 1.05 (app q, J = 3.9 Hz, 2H, H_{16}), 0.99 (d, J = 6.6 Hz, 3H, H_{11}), 0.85 (t, J = 6.9 Hz, 3H, H_1). ^{13}C NMR (151 MHz, DMSO-d_6, major diastereomer) δ 205.2 (C), 204.5 (C), 172.7 (C), 171.8 (C), 170.9 (C), 170.0 (C), 168.5 (C), 167.6 (C), 167.2 (C), 162.4 (C), 156.3 (C), 147.3 (C), 130.0 (CH), 117.2 (CH), 72.7 (CH), 65.8 (CH), 50.6 (CH_2), 50.4 (CH), 46.9 (CH_2), 44.1 (CH), 41.0 (C), 41.0 (CH_2), 39.5 (CH_2), 37.8 (CH_2), 35.7 (CH_2), 31.7 (CH_2), 30.1 (CH_2), 29.5 (4 × CH_2), 29.4 (CH_2), 29.3 (CH_2), 29.2 (CH_2), 29.1 (CH_2), 25.7 (CH_2), 22.5 (CH_2), 21.0 (CH_3), 19.8 (CH_2), 14.4 (CH_3).

^{1}H NMR (600 MHz, DMSO-d_6, minor diastereomer) δ 8.85 (t, J = 6.0 Hz, 1H, H_{18}), 8.81 (s, 1H, H_{15}), 8.42 (s, 1H, H_{24}), 7.86 (d, J = 7.8 Hz, 1H, H_5), 7.48 (d, J = 8.2 Hz, 1H, H_9), 7.41 (s, 1H, H_{20}), 7.25 (bs, 1H, H_8), 6.83 (bs, 1H, H_8), 5.34 (d, J = 5.2 Hz, 1H, H_{21}), 5.18 (d, J = 5.1 Hz, 1H, H_{23}),

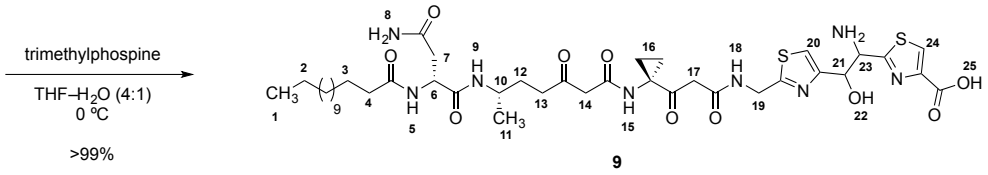
4.47 (d, J = 6.1 Hz, 2H, H_{19}), 4.43 (td, J = 7.6, 5.9 Hz, 1H, H_6), 3.77 – 3.65 (m, 1H, H_{10}), 3.57 (s, 2H, H_{17}), 3.34 (s, 2H, H_{14}), 2.54 – 2.51 (m, 1H, H_{13}), 2.50 – 2.44 (m, 1H, H_{13}), 2.43 (dd, J = 15.1, 5.9 Hz, 1H, H_7), 2.36 (dd, J = 15.3, 7.7 Hz, 1H, H_7), 2.10 (t, J = 7.5 Hz, 2H, H_4), 1.65 – 1.56 (m, 1H, H_{12}), 1.53 – 1.49 (m, 1H, H_{12}), 1.51 – 1.42 (m, 2H, H_3), 1.42 – 1.36 (m, 2H, H_{16}), 1.29 – 1.24 (m, 2H, H_2), 1.24 – 1.21 (m, 18H, myristoyl), 1.05 (app q, J = 3.9 Hz, 2H, H_{16}), 0.99 (d, J = 6.6 Hz, 3H, H_{11}), 0.85 (t, J = 6.9 Hz, 3H, H_1). ^{13}C NMR (151 MHz, DMSO-d_6, minor diastereomer) δ 205.2 (C), 204.5 (C), 172.7 (C), 171.8 (C), 170.9 (C), 169.8 (C), 168.5 (C), 167.2 (C), 165.44 (C), 162.4 (C), 155.5 (C), 146.9 (C), 130.5 (CH), 117.6 (CH), 71.9 (CH), 65.4 (CH), 50.6 (CH_2), 50.4 (CH), 46.9 (CH_2), 44.1 (CH), 41.0 (C), 41.0 (CH_2), 39.5 (CH_2), 37.8 (CH_2), 35.7 (CH_2), 31.7 (CH_2), 30.1 (CH_2), 29.5 (4 × CH_2), 29.4 (CH_2), 29.3 (CH_2), 29.2 (CH_2), 29.1 (CH_2), 25.7 (CH_2), 22.5 (CH_2), 21.0 (CH_3), 19.8 (CH_2), 14.4 (CH_3).

IR (ATR-FTIR), cm^{-1}: 3290 (m), 2919 (m), 2850 (m), 21103 (m), 1710 (m), 1662 (s), 1631 (s), 1541 (m), 1410 (w), 1347 (w), 1201 (m), 1134 (m), 1026 (m), 1005 (w), 800 (w), 720 (m), 587 (m). HRMS-CI (*m/z*): $[M + H]^+$ calcd for $C_{41}H_{61}N_{10}O_{10}S_2$, 917.4008; found, 917.4004.

*Synthesis of the amino alcohol **9**.*



20



9

A solution of trimethylphospine in tetrahydrofuran (1.0 N, 109 μL, 110 μmol, 2.50 equiv) was added to a solution of the acid **20** (40.0 mg, 43.6 μmol, 1 equiv) in tetrahydrofuran–water (4:1 v/v, 1.0 mL) at 23 °C. The reaction mixture was stirred for 40 min at 23 °C. The product mixture was concentrated to provide the amino alcohol **9** as a white solid (38.9 mg, >99%). The amino alcohol **9** obtained in this way was used directly in the following step.

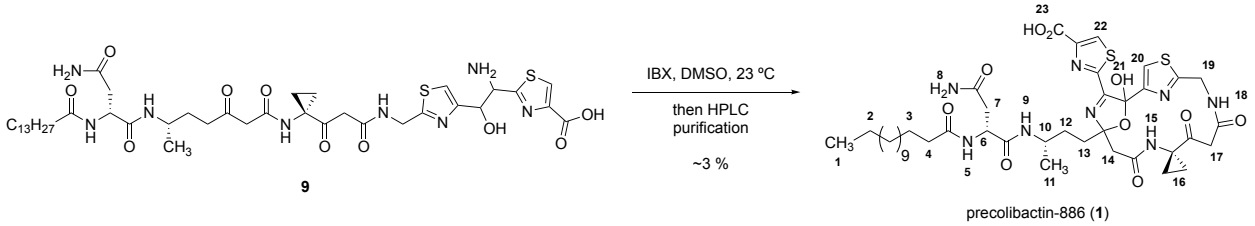
The amino alcohol **9** was isolated as an inconsequential mixture (2:1) of diastereomers.

^{1}H NMR (600 MHz, DMSO-d_6, major diastereomer) δ 8.84 (t, J = 6.0 Hz, 1H, H_{18}), 8.82 (s, 1H, H_{15}), 8.44 (s, 1H, H_{24}), 7.88 (d, J = 7.8 Hz, 1H, H_5), 7.51 (s, 1H, H_{20}), 7.50 (d, J = 8.2 Hz, 1H, H_9), 7.28 – 7.23 (m, 1H, H_8), 6.87 – 6.81 (m, 1H, H_8), 6.70 (d, J = 5.4 Hz, 1H, H_{22}), 5.08 (dd, J = 7.1, 2.9 Hz, 1H, H_{21}), 5.05 (d, J = 7.2 Hz, 1H, H_{23}), 4.52 (d, J = 6.5 Hz, 2H, H_{19}), 4.43 (td, J = 7.7, 5.9 Hz, 1H, H_6), 3.75 – 3.65 (m, 1H, H_{10}), 3.60 (s, 2H, H_{17}), 3.35 (s, 2H, H_{14}), 2.55 – 2.51 (m, 1H, H_{13}), 2.49 – 2.46 (m, 1H, H_{13}), 2.43 (dd, J = 15.1, 5.9 Hz, 1H, H_7), 2.36 (dd, J = 15.2, 7.8 Hz, 1H, H_7), 2.10 (t, J = 7.5 Hz, 2H, H_4), 1.64 – 1.56 (m, 1H, H_{12}), 1.55 – 1.47 (m, 1H, H_{12}), 1.50 – 1.42 (m, 2H, H_3), 1.40 – 1.37 (m, 2H, H_{16}), 1.30 – 1.24 (m, 2H, H_2), 1.23 (s, 18H, myristoyl), 1.06 (app q, J = 3.4 Hz, 2H, H_{16}), 1.00 (d, J = 6.6 Hz, 3H, H_{11}), 0.85 (t, J = 6.9 Hz, 3H, H_1). ^{13}C NMR (151 MHz, DMSO-d_6, major diastereomer) δ 204.83 (C), 204.13 (C), 172.23 (C), 171.41 (C), 170.54 (C), 168.06 (C), 166.9 (C), 166.83 (C), 161.80 (C), 156.3 (C), 146.05 (C), 131.19 (CH), 118.38 (CH), 70.02 (CH), 55.93 (CH), 50.22 (CH_2), 49.96 (CH), 46.43 (CH_2), 43.61 (CH), 40.56 (CH_2), 40.1 (C), 39.10 (CH_2), 37.37 (CH_2), 35.24 (CH_2), 31.30 (CH_2), 29.67 (CH_2), 29.1 (3 × CH_2), 29.0 (2 × CH_2), 28.9 (CH_2), 28.7 (2 × CH_2), 25.23 (CH_2), 22.10 (CH_2), 20.57 (CH_3), 19.40 (2 × CH_2), 13.97 (CH_3).

^{1}H NMR (600 MHz, DMSO-d_6, minor diastereomer) δ 8.86 (t, J = 6.2 Hz, 1H, H_{18}), 8.82 (s, 1H, H_{15}), 8.44 (s, 1H, H_{24}), 7.88 (d, J = 7.8 Hz, 1H, H_5), 7.50 (d, J = 8.2 Hz, 1H, H_9), 7.37 (s, 1H, H_{20}), 7.28 – 7.23 (m, 1H, H_8), 6.87 – 6.81 (m, 1H, H_8), 6.70 (d, J = 5.4 Hz, 1H, H_{22}), 5.23 (t, J = 4.5 Hz, 1H, H_{21}), 5.14 (d, J = 3.9 Hz, 1H, H_{23}), 4.52 (d, J = 6.5 Hz, 2H, H_{19}), 4.55 (d, J = 6.1 Hz, 1H, H_6), 3.75 – 3.65 (m, 1H, H_{10}), 3.60 (s, 2H, H_{17}), 3.35 (s, 2H, H_{14}), 2.55 – 2.51 (m, 1H, H_{13}), 2.49 – 2.46 (m, 1H, H_{13}), 2.43 (dd, J = 15.1, 5.9 Hz, 1H, H_7), 2.36 (dd, J = 15.2, 7.8 Hz, 1H, H_7), 2.10 (t, J = 7.5 Hz, 2H, H_4), 1.64 – 1.56 (m, 1H, H_{12}), 1.55 – 1.47 (m, 1H, H_{12}), 1.50 – 1.42 (m, 2H, H_3), 1.40 – 1.37 (m, 2H, H_{16}), 1.30 – 1.24 (m, 2H, H_2), 1.23 (s, 18H, myristoyl), 1.06 (app q, J = 3.4 Hz, 2H, H_{16}), 1.00 (d, J = 6.6 Hz, 3H, H_{11}), 0.85 (t, J = 6.9 Hz, 3H, H_1).

IR (ATR-FTIR), cm^{-1}: 2965 (w), 2918 (s), 2850 (m), 2360 (w), 2334 (w), 1733 (w), 1465 (w), 1376 (w), 1260 (w), 1177 (w), 1026 (w), 801 (w), 722 (w), 700 (w). HRMS-CI (*m/z*): $[M + H]^+$ calcd for $C_{41}H_{63}N_8O_{10}S_2$, 891.4103; found, 891.4105.

*Synthesis of precolibactin-886 (**1**).*



2-Iodoxybenzoic acid (IBX, 12.6 mg, 44.8 μmol, 4.00 equiv) was added to a solution of the amino alcohol **9** (10.0 mg, 11.2 μmol, 1 equiv) in dimethyl sulfoxide (250 μL) at 23 °C. The resulting mixture was stirred for 1 h at 23 °C. The product mixture was directly injected onto a semipreparative reverse phase HPLC system equipped with a Phenomenex Luna C8 (2) 100 Å column (250 × 10 mm, flow rate 4.0 mL/min, a gradient elution from 70 to 100% aqueous methanol with 0.01% trifluoroacetic acid over 30 min) using a 1 min fraction collection time window. Fraction 14 was isolated and repurified by a semipreparative reverse phase HPLC system with a Phenomenex Luna C18 (2) 100 Å column (250 × 10 mm, flow rate 4.0 mL/min, a gradient elution from 30 to 100% aqueous acetonitrile with 0.01% trifluoroacetic acid over 30 min) to give precolibactin 886 (**1**) as colorless oil (t_R = 18.8 min, ~0.3 mg, ~3%).

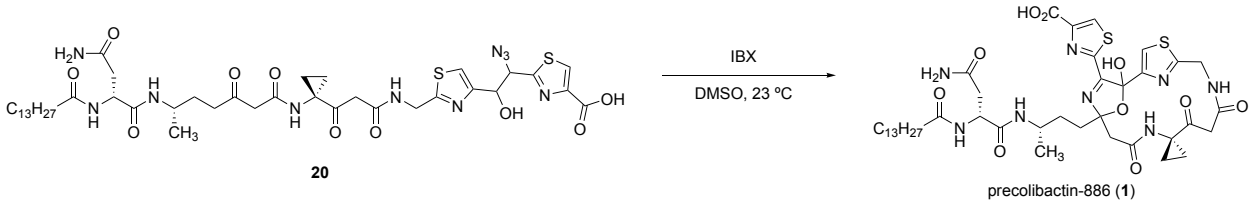
Synthetic precolibactin 886 (**1**) was isolated as a 1.9:1 mixture of C36 diastereomers.

^{1}H NMR (600 MHz, DMSO-d_6, major diastereomer) δ 9.00 (t, J = 6.1 Hz, 1H, H_{18}), 8.15 (s, 1H, H_{22}), 7.89 (overlap, 2H, H_{20}, H_5), 7.87 (overlap, 1H, H_{15}), 7.50 (d, J = 8.4 Hz, 1H, H_9), 7.23 (s, 1H, H_8), 6.81 (s, 1H, H_8), 4.79 (dd, J = 17.2, 7.1 Hz, 1H, H_{19}), 4.45 (q, J = 7.3 Hz, 1H, H_6), 4.27 (dd, J = 17.2, 5.0 Hz, 1H, H_{19}), 3.71 (overlap, 2H, H_{17}, H_{10}), 3.29 (overlap, 1H, H_{14}), 3.10 (d, J = 14.7 Hz, 1H, H_{17}), 2.62 (overlap, 1H, H_{14}), 2.416 (dd, J = 15.1, 5.8 Hz, 1H, H_7), 2.332 (dd, J = 15.2, 7.8 Hz, 1H, H_7), 2.06 (m, 2H, H_4), 1.89 (m, 1H, H_{13}), 1.78 (m, 1H, H_{13}), 1.58 (m, 1H, H_{12}), 1.39 – 1.48 (overlap, 3H, H_{21}, H_3), 1.27 – 1.21 (overlap, 20H), 1.10 (m, 2H, H_{16}), 0.990 (d, J = 6.5 Hz, 3H, H_{11}), 0.87 (m, 2H, H_{16}), 0.85 (t, J = 7.0 Hz, 3H, H_1).

^{1}H NMR (600 MHz, DMSO-d_6, minor diastereomer) δ 9.00 (t, J = 6.1 Hz, 1H, H_{18}), 8.16 (s, 1H, H_{22}), 7.90 (overlap, 2H, H_{20}, H_5), 7.87 (overlap, 1H, H_{15}), 7.55 (d, J = 8.4 Hz, 1H, H_9), 7.26 (s, 1H, H_8), 6.81 (s, 1H, H_8), 4.79 (dd, J = 17.2, 7.1 Hz, 1H, H_{19}), 4.45 (q, J = 7.3 Hz, 1H, H_6), 4.27 (dd, J = 17.2, 5.0 Hz, 1H, H_{19}), 3.71 (overlap, 2H, H_{17}, H_{10}), 3.29 (overlap, 1H, H_{14}), 3.10 (d, J = 14.7 Hz, 1H, H_{17}), 2.62 (overlap, 1H, H_{14}), 2.421 (dd, J = 15.0, 5.6 Hz, 1H, H_7), 2.326 (dd, J = 15.1, 7.8 Hz, 1H, H_7), 2.06 (m, 2H, H_4), 1.78 (m, 1H, H_{13}), 1.69 (m, 1H, H_{13}), 1.58 (m, 1H, H_{12}), 1.39 – 1.48 (overlap, 3H, H_{21}, H_3), 1.27 – 1.21 (overlap, 20H), 1.10 (m, 2H, H_{16}), 0.986 (d, J = 6.4 Hz, 3H, H_{11}), 0.87 (m, 2H, H_{16}), 0.85 (t, J = 7.0 Hz, 3H, H_1).

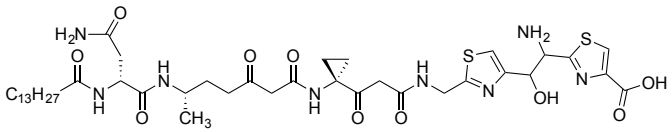
^{13}C NMR (151 MHz, DMSO-d_6, major and minor diastereomers) δ 205.1 (C), 172.5 (C), 171.4 (C), 170.3 (C), 169.8 (C), 168.1 (C), 166.8 (C), 160.0 (C), 153.6 (C), 119.9 (CH), 107.9 (C), 107.4 (C), 49.9 (CH), 48.3 (CH_2), 45.8 (CH_2), 44.5 (CH), 40.4 (CH_2), 39.3 (C), 37.5 (CH_2), 35.2 (CH_2), 34.9 (CH_2), 31.3 (CH_2), 30.3 (CH_2), 29.1 – 28.7 (8 × CH_2), 25.2 (CH_2), 22.1 (CH_2), 20.6 (CH_2), 14.0 (CH_3). HRMS-CI (m/z): $[M + H]^+$ calcd for $C_{41}H_{59}N_8O_{10}S_2$, 887.3790; found, 887.3794.

*Synthesis of precolibactin-886 (**1**) from the azide **20**.*

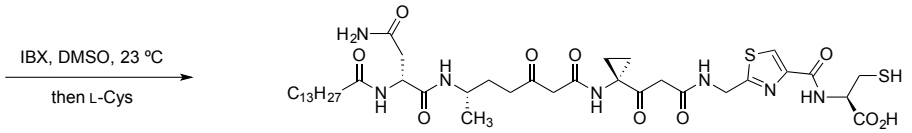


2-Iodoxybenzoic acid (IBX, 3.14 mg, 11.2 μmol, 4.00 equiv) was added to a solution of the azide **20** (2.5 mg, 2.8 μmol, 1 equiv) in dimethyl sulfoxide (250 μL) at 23 °C. The resulting mixture was stirred for 1 h at 23 °C. Production of precolibactin 886 (**1**) was observed by LC/HRMS analysis of the reaction mixture.

*Synthesis of linear precolibactin A (**33**) from the amino alcohol **9**.*



9



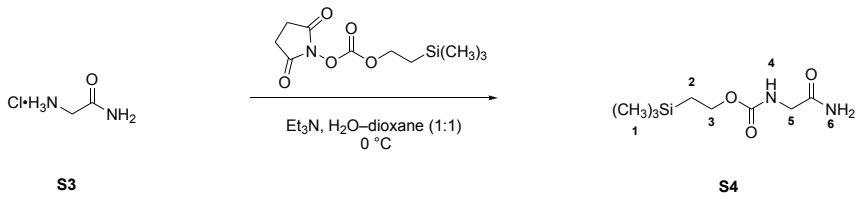
linear precolibactin A (**33**)

2-Iodoxybenzoic acid (IBX, 3.14 mg, 11.2 μmol, 4.00 equiv) was added to a solution of the amino alcohol **9** (2.50 mg, 2.81 μmol, 1 equiv) in dimethyl sulfoxide (250 μL) at 23 °C. The resulting mixture was stirred for 2 h at 23 °C. L-Cysteine (2.5 mg, 21.1 μmol, 7.50 equiv) was then added to the reaction mixture. The resulting mixture was stirred for 5 h at 23 °C and analyzed by LC/HRMS. The retention time of synthetic **33** was identical to that of natural **33**, the structure of which was confirmed by tandem MS.

33: HRMS-CI (*m/z*): $[M + H]^+$ calcd for $C_{39}H_{62}N_7O_{10}S_2$, 852.3994; found, 852.3989.

*Synthesis of thiazole **S6**.*

*Step 1: Synthesis of the amide **S4**.*

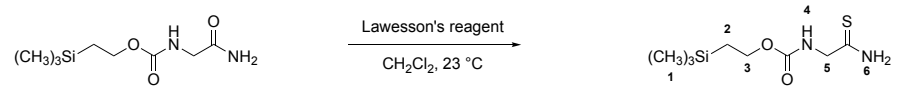


S3 **S4**

A solution of triethylamine (1.60 mL, 11.8 mmol, 2.60 equiv) in 1,4-dioxane (29 mL) was added dropwise via an addition funnel over 30 min to a solution of glycinamide hydrogen chloride (**S3**, 500 mg, 4.52 mmol, 1 equiv) in distilled water (29 mL) at 23 °C. 1-[2-(Trimethylsilyl)ethoxycarbonyloxy]pyrrolidin-2,5-dione (1.29 g, 4.97 mmol, 1.10 equiv) was then added in one portion. The reaction was stirred for 16 h at 23 °C. The product mixture was diluted with ethyl acetate (100 mL) and the aqueous layer was separated. The aqueous layer was extracted with ethyl acetate (2 × 40 mL). The organic layers were combined and the combined organic layers were washed sequentially with saturated aqueous sodium bicarbonate solution (80 mL) and saturated aqueous sodium chloride solution (80 mL). The washed organic layer was dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated to provide the amide **S4** as a white solid. The product **S4** obtained in this way was used directly in the following step.

R_f = 0.10 (50% hexanes–ethyl acetate; UV, $KMnO_4$). ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.22 (s, 1H, H_6), 7.07 (t, J = 5.8 Hz, 1H, H_4), 6.96 (s, 1H, H_6), 4.03 (t, J = 8.3 Hz, 2H, H_3), 3.50 (d, J = 6.1 Hz, 2H, H_5), 0.92 (t, J = 8.4 Hz, 2H, H_2), 0.02 (s, 9H, H_1). ^{13}C NMR (100 MHz, DMSO-d_6) δ 171.2 (C), 156.6 (C), 61.8 (CH_2), 43.2 (CH_2), 17.4 (CH_2), –1.4 (3 × CH_3).

*Step 2: Synthesis of the thioamide **S5**.*

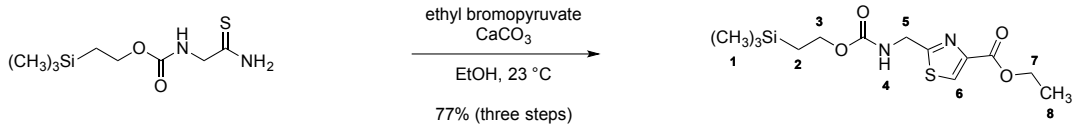


S4 **S5**

Lawesson's reagent (1.66 g, 4.12 mmol, 1.00 equiv) was added to a solution of the amide **S4** obtained in the preceding step (nominally 4.12 mmol, 1 equiv) in dichloromethane (41 mL) at 23 °C. The resulting mixture was stirred for 16 h at 23 °C. The product mixture was filtered through a pad of celite (2.5 × 4.5 cm). The filter cake was washed with dichloromethane (15 mL). The filtrates were combined, and the combined filtrates were concentrated. The residue obtained was dissolved in ethyl acetate (100 mL) and the resulting solution was washed sequentially with saturated aqueous sodium bicarbonate solution (50 mL) and saturated aqueous sodium chloride solution (50 mL). The washed organic layer was dried over sodium sulfate. The dried solution was filtered, and the filtrate was concentrated to provide the thioamide **S5** as a white solid. The product **S5** obtained in this way was used directly in the following step.

R_f = 0.45 (50% hexanes–ethyl acetate; UV, $KMnO_4$). ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.67 (br s, 1H, H_6), 9.01 (br s, 1H, H_6), 7.27 (t, J = 5.5 Hz, 1H, H_4), 4.04 (t, J = 8.4 Hz, 2H, H_3), 3.85 (d, J = 6.1 Hz, 2H, H_5), 0.93 (t, J = 8.4 Hz, 2H, H_2), 0.02 (s, 9H, H_1). ^{13}C NMR (100 MHz, DMSO-d_6) δ 203.8 (C), 156.4 (C), 62.0 (CH_2), 55.2 (CH_2), 17.3 (CH_2), –1.4 (3 × CH_3).

*Step 3: Synthesis of thiazole **S6**.*

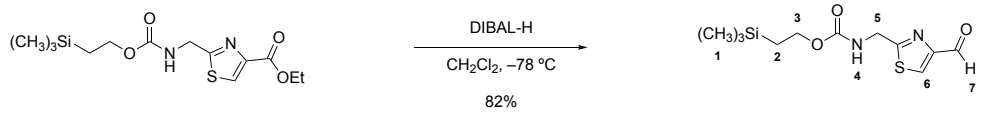


S5 **S6**

Ethyl bromopyruvate (770 μL, 6.15 mmol, 1.50 equiv) and calcium carbonate (410 mg, 4.10 mmol, 1.00 equiv) were added in sequence to a solution of the thiomaide **S5** obtained in the preceding step (nominally, 4.11 mmol, 1 equiv) in ethanol (16 mL) at 23 °C. The reaction mixture was stirred for 16 h at 23 °C. The product mixture was concentrated. The residue obtained was dissolved in chloroform (30 mL) and the resulting solution was washed with saturated aqueous sodium chloride solution (20 mL). The organic layer was dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated. The residue obtained was purified by flash-column chromatography (eluting with 10% ethyl acetate–hexanes initially, grading to 30% ethyl acetate–hexanes, linear gradient) to furnish the thiazole **S6** as a yellow solid (1.13 g, 77% over 3 steps).

R_f = 0.55 (50% hexanes–ethyl acetate; UV, $KMnO_4$). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.42 (s, 1H, H_6), 8.02 (t, J = 6.2 Hz, 1H, H_4), 4.45 (d, J = 6.2 Hz, 2H, H_5), 4.29 (q, J = 7.0 Hz, 2H, H_7), 4.09 (t, J = 8.3 Hz, 2H, H_3), 1.29 (t, J = 7.0 Hz, 3H, H_8), 0.94 (t, J = 8.3 Hz, 2H, H_2), 0.03 (s, 9H, H_1). ^{13}C NMR (100 MHz, DMSO-d_6) δ 171.5 (C), 160.7 (C), 156.6 (C), 145.7 (C), 129.1 (CH), 62.3 (CH_2), 60.7 (CH_2), 42.1 (CH_2), 17.3 (CH_2), 14.2 (CH_3), –1.4 (3 × CH_3). IR (ATR-FTIR), cm^{-1}: 3325 (w), 2972 (w), 1718 (s), 1716 (s), 1522 (m). HRMS-CI (m/z): $[M + H]^+$ calcd for $C_{13}H_{23}N_2O_4SSi$, 331.1142, mass; found, 331.1177.

*Synthesis of the aldehyde **S7**.*



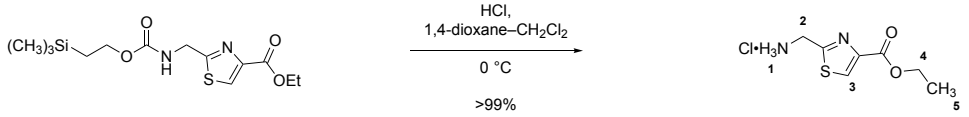
S6 **S7**

A solution of di-*iso*-butylaluminium hydride (DIBAL-H) in dichloromethane (1.0 M, 16.1 mL, 16.1 mmol, 3.01 equiv) was added dropwise over 30 min to a solution of the thiazole **S6** (1.77 g, 5.35 mmol, 1 equiv) in dichloromethane (35 mL) at –78 °C. The reaction was stirred for 3 h at –78 °C. The cold product mixture was diluted slowly with methanol (2.0 mL). The diluted product mixture was allowed to warm to 23 °C over 30 min. The warmed product mixture was diluted with saturated aqueous sodium potassium tartrate solution (40 mL). The resulting biphasic mixture was stirred vigorously for 4 h at 23 °C. The biphasic mixture was transferred to a separatory funnel and the layers that formed were separated. The aqueous layer was extracted with dichloromethane (2 × 30 mL). The organic layers were combined and the combined organic layers were washed with saturated aqueous sodium chloride solution (40 mL). The washed organic layer was dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated. The residue obtained was purified by flash-column chromatography (eluting with 10% ether–hexanes initially, grading to 20% ether–hexanes, linear gradient) to furnish the aldehyde **S7** as a yellow oil (1.25 g, 82%).

R_f = 0.40 (20% hexanes–ether; UV, $KMnO_4$). ^{1}H NMR (500 MHz, DMSO-d_6) δ 9.84 (s, 1H, H_7), 8.59 (s, 1H, H_6), 8.00 (t, J = 5.5 Hz, 1H, H_4), 4.44 (d, J = 6.1 Hz, 2H, H_5), 4.06 (app. t, J = 8.2 Hz, 2H, H_3), 0.91 (app. t, J = 8.2 Hz, 2H, H_2), –0.01 (s, 9H, H_1). ^{13}C NMR (125 MHz, DMSO-d_6) δ 184.9 (CH), 172.5 (C), 156.6 (C), 154.2 (C), 132.1 (CH), 62.3 (CH_2), 42.2 (CH_2), 17.3 (CH_3), –1.4 (3 × CH_3). IR (ATR-FTIR), cm^{-1}: 3323 (w), 2954 (m), 2923 (w), 1698 (s), 1529 (m), 1250 (s). HRMS-CI (m/z): $[M + Na]^+$ calcd for $C_{11}H_{18}N_2NaO_3SSi$, 309.0700; found, 309.0753

*Synthesis of the amino alcohol **23**.*

*Step 1: Synthesis of the ammonium ion **S8**.*

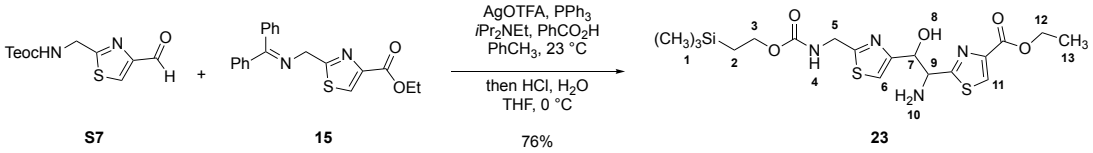


S6 **S8**

A solution of hydrogen chloride in 1,4-dioxane (4.0 N, 25 mL, 100 mmol, 11.0 equiv) was added dropwise via syringe to a solution of the thiazole **S6** (3.00 g, 9.08 mmol, 1 equiv) in dichloromethane (50 mL) at 0 °C. The reaction mixture was allowed to warm to 23 °C and was stirred at this temperature for 16 h. The product mixture was concentrated to provide the ammonium ion **S8** as a white solid (2.02 g, >99%). The product **S8** obtained in this way was used directly in the following step.

^{1}H NMR (400 MHz, DMSO-d_6) δ 8.70 (br s, 3H, H_1), 8.60 (s, 1H, H_3), 4.47 (br s, 2H, H_2), 4.32 (q, J = 7.0 Hz, 2H, H_4), 1.31 (t, J = 7.0 Hz, 3H, H_5). ^{13}C NMR (100 MHz, DMSO-d_6) δ 163.1 (C), 160.5 (C), 145.6 (C), 131.1 (CH), 60.9 (CH_2), 39.4 (CH_2, obscured by NMR solvent, detected indirectly by HSQC), 14.2 (CH_3).

*Step 2: Synthesis of the amino alcohol **23**.*



A solution of triphenylphosphine (260 mg, 990 μmol, 0.10 equiv) in toluene (10 mL) was added to a solution of silver trifluoromethanesulfonate (254 mg, 990 μmol, 0.10 equiv) in toluene (5.0 mL) at 23 °C. The resulting mixture was stirred for 20 min at 23 °C, with protection from light. A solution of the imine **15** (3.48 g, 9.92 mmol, 1 equiv) in toluene (10 mL) and a solution of the aldehyde **S7** (4.26 g, 14.9 mmol, 1.50 equiv) in toluene (10 mL) were then added in sequence. A solution of *N*,*N*-di-*iso*-propyl ethylamine (510 μL, 1.98 mmol, 0.20 equiv) and benzoic acid (60.0 mg, 50.0 μmol, 0.05 equiv) in toluene (1.2 mL) was prepared in a separate flask. This solution was then added to the flask containing the imine **15** and the aldehyde **S7**. The resulting mixture was stirred for 32 h at 23 °C. The product mixture was concentrated to dryness. The residue obtained was partitioned between saturated aqueous sodium chloride solution (40 mL) and dichloromethane (40 mL). The layers that formed were separated, and the aqueous layer was extracted with dichloromethane (2 × 40 mL). The organic layers were combined and the combined organic layers were dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated.

The residue obtained was dissolved in tetrahydrofuran (100 mL) and the resulting solution was cooled to 0 °C. Aqueous hydrogen chloride solution (1.0 N, 24 mL) was then added. The reaction mixture was stirred for 1 h at 0 °C. The product mixture was partially concentrated to remove tetrahydrofuran. The partially concentrated product mixture was diluted with water (20 mL) and the diluted solution was extracted with ether (3 × 50 mL). The pH of the aqueous layer was adjusted to ~8–9 by the slow addition of solid sodium hydrogen carbonate. The basified solution was extracted with dichloromethane (4 × 50 mL). The organic layers were combined and the combined organic layers were dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated to provide the amino alcohol **23** as a yellow solid (3.54 g, 76%).

The amino alcohol **23** was isolated as an inconsequential mixture (2:1) of diastereomers.

R_f = 0.17 (80% ethyl acetate–hexanes; UV, $KMnO_4$).

^{1}H NMR (400 MHz, DMSO-d_6, major diastereomer) δ 8.38 (s, 1H, H_{11}), 7.94 (t, J = 6.2 Hz, 1H, H_4), 7.38 (s, 1H, H_{11}), 5.72 (d, J = 5.7 Hz, 1H, H_8), 5.26 (app. d, J = 5.7 Hz, 1H, H_7), 4.43 (d, J = 6.2 Hz, 2H, H_5), 4.41 (d, J = 2.4 Hz, 1H, H_9), 4.34–4.25 (m, 2H, H_{12}), 4.10 (app. t, J = 8.3 Hz, 2H, H_3), 2.32 (br s, 2H, H_{10}), 1.31 (t, J = 7.1 Hz, 3H, H_{13}), 0.95 (app. t, J = 8.3 Hz, 2H, H_2), 0.03 (s, 9H, H_1). ^{13}C NMR (100 MHz, DMSO-d_6, major diastereomer) δ 178.9 (C), 170.2 (C), 161.1 (C), 157.9 (C), 156.5 (C), 146.1 (C), 128.95 (CH), 115.3 (CH), 72.5 (CH), 62.2 (CH_2), 60.5 (CH_2), 57.4 (CH), 42.2 (CH_2), 17.3 (CH_2), 14.2 (CH_3), –1.4 (3 × CH_3). IR (ATR-FTIR), cm^{-1}: 3360 (w), 3334 (w), 2954 (m), 1717 (s), 1712 (s), 1522 (m), 1249 (s). HRMS-CI (m/z): $[M + H]^+$ calcd for $C_{18}H_{29}N_4O_5S_2Si$, 473.1343; found, 473.1357.

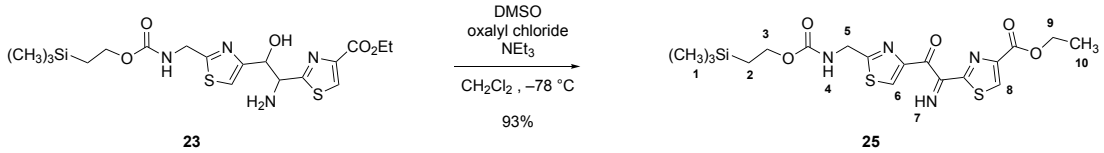
*Synthesis of the azide **24**.*

1*H*-Imidazole-1-sulfonyl azide hydrogen chloride (110 mg, 635 μmol, 3.00 equiv) was added to a solution of the amino alcohol **23** (100 mg, 212 μmol, 1 equiv), triethylamine (206 μL, 1.48 mmol, 7.00 equiv), and copper(II) sulfate pentahydrate (0.5 mg, 2.00 μmol, 0.01 equiv) in methanol (1.1 mL) at 23 °C. The resulting mixture was stirred for 2 h at 23 °C. The product mixture was concentrated to dryness. Aqueous hydrogen chloride solution (1.0 M, 10 mL) and ethyl acetate (20 mL) were then added in sequence. The resulting biphasic mixture was transferred to a separatory funnel and the layers that formed were separated. The aqueous layer was extracted with ethyl acetate (2 × 20 mL). The organic layers were combined and the combined organic layers were washed with saturated aqueous sodium chloride solution (20 mL). The washed organic layer was dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated. The residue obtained was purified by flash-column chromatography (eluting with hexanes initially, grading to 60% ethyl acetate–hexanes, linear gradient) to provide the azide **24** as a yellow oil (102 mg, 96%).

The azide **24** was isolated as a mixture (2:1) of diastereomers (stereochemistry not assigned).

R_f = 0.50 (60% ethyl acetate–hexanes; UV, $KMnO_4$). ^{1}H NMR (500 MHz, $CDCl_3$, major diastereomer) δ 8.17 (s, 1H, H_{10}), 7.33 (s, 1H, H_6), 5.55 – 5.28 (m, 3H, H_4, H_7, H_9), 4.62 (d, J = 6.0 Hz, 2H, H_5), 4.40 (q, J = 7.0 Hz, 2H, H_{11}), 4.22 – 4.15 (m, 2H, H_3), 1.39 (app. t, J = 7.0 Hz, 3H, H_{12}) 1.02 – 0.96 (m, 2H, H_2), 0.03 (s, 9H, H_1). ^{13}C NMR (126 MHz, $CDCl_3$, major diastereomer) δ 169.2 (C), 167.9 (C), 161.2 (C), 156.7 (C), 154.8 (C), 147.2 (C), 128.7 (CH), 116.7 (CH), 72.8 (CH), 66.1 (CH), 63.9 (CH_2), 61.7 (CH_2), 42.6 (CH_2), 17.8 (CH_2), 14.4 (CH_3), –1.3 (CH_3). IR (ATR-FTIR), cm^{-1}: 3353 (broad), 2954 (m), 2109 (s), 1717 (s), 1524 (m), 1249 (s). HRMS-CI (m/z): $[M + Na]^+$ calcd for $C_{18}H_{26}N_6O_5S_2SiNa$, 521.1068; found, 521.1073.

*Synthesis of the α-ketoimine **25**.*

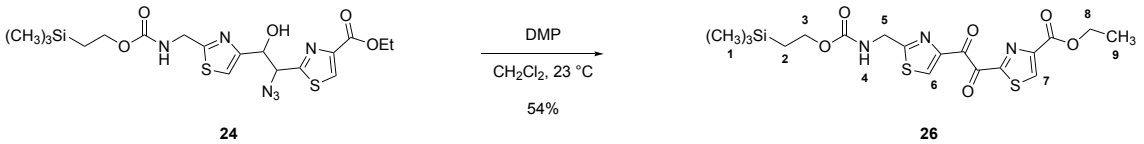


23 **25**

A solution of dimethyl sulfoxide (65.0 μL, 913 μmol, 4.36 equiv) in dichloromethane (1.0 mL) was added dropwise over 5 min to a solution of oxalyl chloride (38.0 μL, 454 μmol, 2.17 equiv) in dichloromethane (2.0 mL) at –78 °C. The resulting mixture was stirred for 15 min at –78 °C. A solution of the amino alcohol **23** (99.0 mg, 209 μmol, 1 equiv) in dichloromethane (2.0 mL) was then added dropwise via syringe. The resulting mixture was stirred for 2 h at –78 °C. Triethylamine (250 μL, 1.82 mmol, 8.70 equiv) was then added dropwise via syringe. The resulting mixture was warmed to –40 °C and stirred for 30 min at –40 °C. The product mixture was transferred to a separatory funnel that had been charged with saturated aqueous ammonium chloride solution (10 mL). The diluted product mixture was extracted with dichloromethane (2 × 10 mL). The organic layers were combined and the combined organic layers were dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated to provide the α-ketoimine **25** as a dark green solid (91.0 mg, nominally 93%). The α-ketoimine **25** was unstable toward chromatographic purification and was used without further purification.

^{1}H NMR (400 MHz, DMSO-d_6) δ 12.07 (s, 1H, H_7), 8.74 (s, 1H, H_8), 8.70 (s, 1H, H_6), 8.04 (t, J = 6.2 Hz, 1H, H_4), 4.41 (d, J = 6.2 Hz, 2H, H_5), 4.28 (q, J = 7.2 Hz, 2H, H_9), 4.09 (app q, J = 8.6 Hz, 2H, H_3), 1.27 (t, J = 7.2 Hz, 3H, H_{10}), 0.94 (app q, J = 8.6 Hz, 2H, H_2), 0.02 (s, 9H, H_1). ^{13}C NMR (100 MHz, DMSO-d_6) δ 184.8 (C), 172.6 (C), 167.9 (C), 166.6 (C), 160.2 (C), 156.6 (C), 151.08 (C), 147.5 (C), 132.5 (CH), 131.7 (CH), 62.4 (CH_2), 61.1 (CH_2), 42.1 (CH_2), 17.3 (CH_2), 14.2 (CH_3), –1.4 (3 × CH_3). IR (ATR-FTIR), cm^{-1}: 3311 (w), 3101 (w), 2961 (w), 1691 (s), 1644 (s), 1514 (m), 1244 (s). HRMS-CI (m/z): $[M + H]^+$ calcd for $C_{18}H_{25}N_4O_5S_2Si$, 469.1030; found, 469.1055.

*Synthesis of the diketone **26** from the azide **24**.*

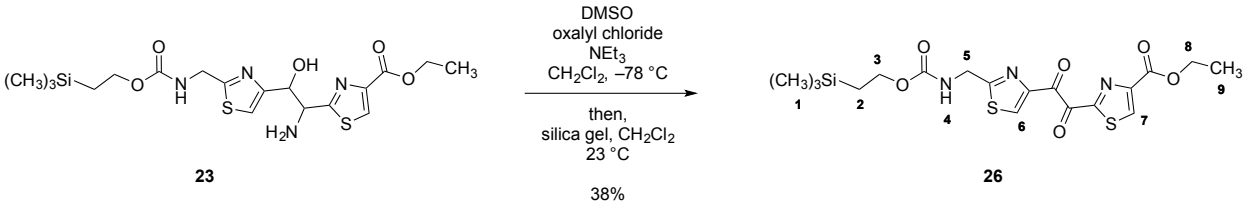


24 **26**

The Dess–Martin periodinane (280 mg, 660 μmol, 2.50 equiv) was added to a solution of the azide **24** (140 mg, 264 μmol, 1 equiv) in dichloromethane (9.0 mL) at 23 ºC. The resulting mixture was stirred for 30 min at 23 °C. The product mixture was concentrated. The residue obtained was dissolved in ether (10 mL) and the mixture was transferred to a separatory funnel that had been charged with saturated aqueous ammonium chloride solution (15 mL). The layers that formed were separated and the aqueous layer was extracted with ether (2 × 10 mL). The organic layers were combined and the combined organic layers were washed with saturated aqueous sodium chloride solution (30 mL). The washed organic layer was dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated. The residue obtained was purified by flash-column chromatography (eluting with 30% ethyl acetate–hexanes) to provide the diketone **26** as a bright yellow solid (71.0 mg, 54%).

R_f = 0.60 (60% ethyl acetate–hexanes; UV, $KMnO_4$). ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.09 (s, 1H, H_7), 8.97 (s, 1H, H_6), 8.02 (t, J = 5.7 Hz, 1H, H_4), 4.43 (d, J = 6.1 Hz, 2H, H_5), 4.32 (q, J = 7.1 Hz, 2H, H_8), 4.08 (t, J = 8.4 Hz, 2H, H_3), 1.29 (t, J = 7.1 Hz, 3H, H_9), 0.93 t, J = 8.4 Hz, 2H, H_2), 0.02 (s, 9H, H_1). ^{13}C NMR (100 MHz, DMSO-d_6) δ 185.4 (C), 184.6 (C), 173.3 (C), 162.2 (C), 159.9 (C), 156.6 (C), 149.1 (C), 148.6 (C), 136.9 (CH), 133.0 (CH), 62.4 (CH_2), 61.5 (CH_2), 42.1 (CH_2), 17.3 (CH_2), 14.1 (CH_3), –1.42 (3 × CH_3). IR (ATR-FTIR), cm^{-1}: 3370 (w), 3103 (w), 2953 (w), 1699 (s), 1693 (s), 1519 (m), 1238 (s). HRMS-CI (m/z): [M + Na]$^+$ calcd for $C_{18}H_{23}N_3NaO_6S_2Si$, 492.0690; found, 492.0730.

*Synthesis of the diketone **26** from the amino alcohol **23**.*

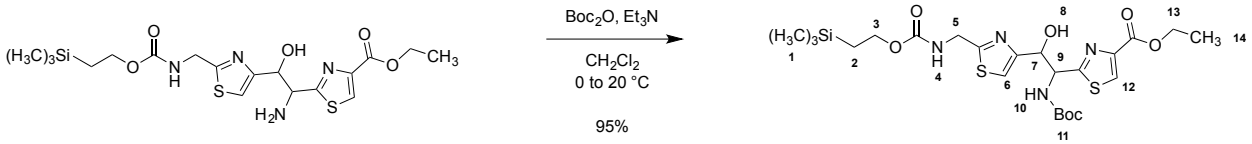


23 **26**

A solution of methyl sulfoxide (98.0 μL, 1.38 mmol, 4.36 equiv) in dichloromethane (2.0 mL) was added dropwise over 5 min to a solution of oxalyl chloride (59.0 μL, 688 μmol, 2.17 equiv) in dichloromethane (3.0 mL) at –78 °C. The resulting mixture was stirred for 15 min at –78 °C. A solution of the amino alcohol **23** (150 mg, 317 μmol, 1 equiv) in dichloromethane (2.0 mL) was then added dropwise via syringe. The resulting mixture was stirred for 2 h at –78 °C. Triethylamine (390 μL, 2.76 mmol, 8.70 equiv) was then added dropwise via syringe. The reaction mixture was warmed to –40 °C and the warmed reaction mixture was stirred for 30 min at –40 °C. The product mixture was transferred to a separatory funnel that had been charged with saturated aqueous ammonium chloride solution (15 mL). The diluted product mixture was extracted with dichloromethane (2 × 15 mL). The organic layers were combined, and the combined organic layers were dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated. The residue obtained was dissolved in dichloromethane (3.0 mL). Silica gel (200 mg) was added and the resulting mixture was stirred for 15 min at 24 °C. The suspension was loaded directly onto a flash-column and purified (eluting with 30% ethyl acetate–hexanes initially, grading to 60% ethyl acetate–hexanes, two steps) to furnish the diketone **26** as a bright yellow solid (57.0 mg, 38% from **23**).

Spectroscopic data for the diketone **26** obtained in this way were identical to that reported above.

*Synthesis of the carbamate alcohol **27**.*



23 **27**

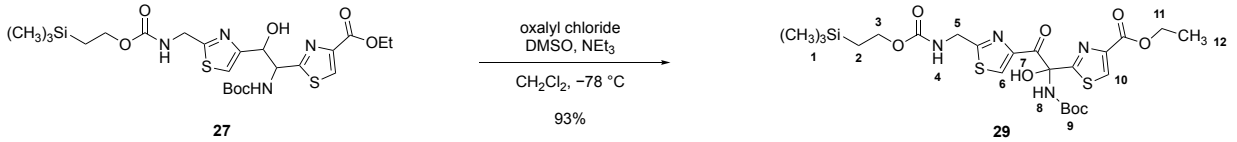
Di-*tert*-butyl dicarbonate (7.4 mg, 33.9 µmol, 1.6 equiv) was added to a solution of triethyl amine (6.0 µL, 42.3 µmol, 2.00 equiv) and the amino alcohol **23** (10.0 mg, 21.2 µmol, 1 equiv) in dichloromethane at 0 °C and the resulting mixture was stirred for 15 min at 0 °C. The reaction was then warmed to 20 °C and stirred for 12 h at 20 °C. The product mixture was then diluted with ethyl acetate (50 mL). The diluted product mixture was transferred to a separatory funnel. The organic layer was washed sequentially with saturated ammonium chloride aqueous solution (30 mL), saturated sodium bicarbonate aqueous solution (30 mL), and saturated sodium chloride aqueous solution (30 mL). The washed organic layer was dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated. The residue obtained was purified by automated flash-column chromatography (eluting with hexanes initially, grading to ethyl acetate) to provide the carbamate alcohol **27** as a colorless residue (11.5 mg, 95%).

The amino alcohol **27** was isolated as an inconsequential mixture (2.5:1) of diastereomers.

R_f = 0.30 (40% ethyl acetate–hexanes; UV, $KMnO_4$).

^{1}H NMR (600 MHz, DMSO-d_6, major diastereomer) δ 8.42 (s, 1H, H_{12}), 8.06 – 7.56 (m, 1H, H_4), 7.39 (s, 1H, H_6), 7.07 (d, J = 8.3 Hz, 1H, H_8), 5.93 (bs, 1H, H_{10}), 5.25 – 5.17 (m, 2H, H_7, H_9), 4.41 (d, J = 6.8 Hz, 2H, H_5), 4.31 (q, J = 7.1, 2H, H_{13}), 4.09 (t, J = 8.3 Hz, 2H, H_3), 1.32 (s, 9H, H_{11}), 1.32 – 1.28 (m, 3H, H_{14}), 0.97 – 0.89 (m, 2H, H_2), 0.03 (s, 9H, H_1). ^{13}C NMR (151 MHz, DMSO-d_6, major diastereomer) δ 173.7 (C), 170.4 (C), 160.8 (C), 156.5 (C), 155.8 (C), 155.2 (C), 146.0 (C), 128.9 (CH), 115.7 (CH), 78.9 (C), 71.6 (CH), 62.2 (CH_2), 60.7 (CH_2), 57.6 (CH), 42.2 (CH_2), 28.0 (3 × CH_3), 17.3 (CH_2), 14.2 (CH_3), –1.4 (3 × CH_3). LRMS (m/z): $[M + H]^+$ calcd for $C_{23}H_{37}N_4O_7S_2Si$, 573.19; found, 573.21.

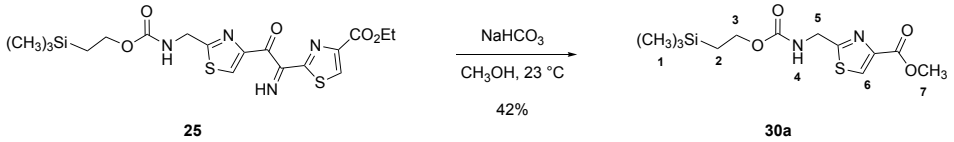
*Synthesis of the hemiaminal **29.***



A solution of dimethyl sulfoxide (65.0 μL, 913 μmol, 4.36 equiv) in dichloromethane (1.0 mL) was added dropwise over 5 min to a solution of oxalyl chloride (38.0 μL, 454 μmol, 2.17 equiv) in dichloromethane (2.0 mL) at –78 °C. The resulting mixture was stirred for 15 min at –78 °C. A solution of the carbamate alcohol **27** (120 mg, 209 μmol, 1 equiv) in dichloromethane (2.0 mL) was then added dropwise via syringe. The resulting mixture was stirred for 2 h at –78 °C. Triethylamine (250 μL, 1.82 mmol, 8.70 equiv) was then added dropwise via syringe. The resulting mixture was warmed to –40 °C and stirred for 30 min at –40 °C. The product mixture was transferred to a separatory funnel that had been charged with saturated aqueous ammonium chloride solution (10 mL). The diluted product mixture was extracted with dichloromethane (2 × 10 mL). The organic layers were combined and the combined organic layers were dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated to provide the hemiaminal **29** as a bright yellow semi-solid (109 mg, nominally 89%). The hemiaminal **29** was unstable toward chromatographic purification and was used without further purification

^{1}H NMR (500 MHz, DMSO-d_6) δ 8.63 (s, 1H, H_{10}), 8.48 (s, 1H, H_6), 7.93 (t, J = 6.2 Hz, 1H, H_4), 7.80 (bs, 1H, H_8), 4.42 – 4.37 (m, 2H, H_5), 4.23 (q, J = 7.4 Hz, 2H, H_{11}), 4.05 (t, J = 8.2 Hz, 2H, H_3), 3.56 (br s, 1H, H_7), 1.29 –1.20 (m, 12H, H_9, H_{12}), 0.90 (t, J = 8.2 Hz, 2H, H_2), -0.02 (s, 9H, H_1). ^{13}C NMR (125 MHz, DMSO-d_6) δ 186.6 (C), 172.9 (C), 170.9 (C), 161.1 (C), 157.0 (C), 149.9 (C), 146.6 (C), 145.7 (C), 131.5 (CH), 130.9 (CH), 79.7 (C), 62.7 (CH_2), 61.1 (CH_2), 42.5 (CH_2), 28.3 (3 × CH_3), 17.8 (CH_2), 14.6 (CH_3), –1.00 (3 × CH_3). IR (ATR-FTIR), cm^{-1}: 3364 (m), 3309 (w), 3117 (w), 2959 (w), 1693 (s), 1649 (s), 1520 (m), 1270 (s). HRMS-CI (m/z): [M + H]+ calcd for $C_{23}H_{35}N_4O_8S_2Si$, 587.1660; found, 587.1781.

*Synthesis of the methyl ester **30a** from the α-ketoimine **25**.*

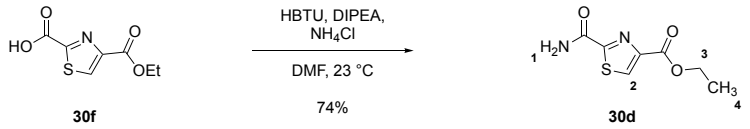


25 **30a**

Sodium bicarbonate (30.0 mg, 360 μmol, 9.47 equiv) was added to a solution of the α-ketoimine **25** (18.0 mg, 38.0 μmol, 1 equiv) in methanol (600 μL) at 23 ºC in a 1-dram vial. The vial was sealed and the mixture was stirred for 48 h at 23 °C. The product mixture was concentrated. The residue obtained was purified by preparative thin-layer chromatography (eluting with 60% ethyl acetate–hexanes) to provide the methyl ester **30a** as a colorless oil (5.1 mg, 42%).

R_f = 0.75 (60% ethyl acetate–hexanes; UV, $KMnO_4$). ^1H NMR (500 MHz, CD_3OD-d_4) δ 8.32 (s, 1H, H_6), 4.57 (s, 2H, H_5), 4.20 (t, J = 8.5 Hz, 2H, H_3), 3.91 (s, 3H, H_7), 1.02 (app. t, J = 8.5 Hz, 2H, H_2), 0.06 (s, 9H, H_1). ^{13}C NMR (150 MHz, CD_3OD-d_4) δ 172.4 (C), 161.5 (C), 157.7 (C), 145.8 (C), 128.1 (CH), 63.1 (CH_2), 51.3 (CH_3), 41.9 (CH_2), 17.2 (CH_2), –2.9 (3 × CH_3). IR (ATR-FTIR), cm^{-1}: 2953 (m), 2360 (m), 2343 (m), 1721 (s), 1718 (s), 1434 (m), 1249 (s), 1217 (s). HRMS-CI (m/z): $[M + H]^+$ calcd for $C_{12}H_{21}N_2O_4SSi$, 317.0986; found, 317.0991.

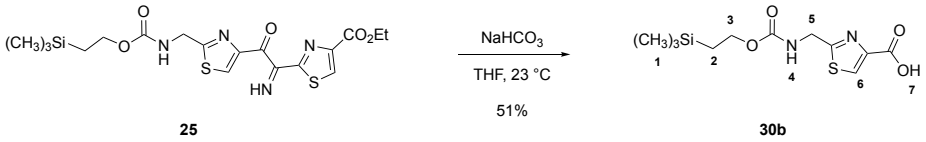
*Synthesis of the carboxamide **30d.***



Ammonium chloride (160 mg, 873 μmol, 3.00 equiv) was added in one portion to a solution of the carboxylic acid **30f** (58.6 mg, 291 μmol, 1 equiv), *N,N*-di-*iso*-propylethylamine (71.0 μL, 437 μmol, 1.50 equiv) and HBTU (133 mg, 350 μmol, 1.20 equiv) in *N,N*-dimethylformamide (4.0 mL) at 23 °C. The reaction mixture was stirred for 2 h at 23 °C. The product mixture was diluted sequentially with saturated aqueous sodium bicarbonate solution (10 mL) and ethyl acetate (15 mL). The layers that formed were separated, and the aqueous layer was extracted with ethyl acetate (2 × 15 mL). The organic layers were combined and the combined organic layers were washed with saturated aqueous sodium chloride solution (20 mL). The washed organic layer was dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated. The residue obtained was purified by flash-column chromatography (eluting with 30% ethyl acetate–hexanes initially, grading to 50% ethyl acetate–hexanes) to provide the carboxamide **30d** as a pale yellow solid (43.3 mg, 74%).

R_f = 0.59 (100% ethyl acetate; UV). ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.75 (s, 1H, H_2), 8.27 (s, 1H, H_1), 7.98 (s, 1H, H_1), 4.34 (q, J = 7.1 Hz, 3H, H_3), 1.32 (t, J = 7.1 Hz, 4H, H_4). ^{13}C NMR (125 MHz, DMSO-d_6) δ 165.2 (C), 160.41 (C), 160.40 (C), 146.8 (C), 133.8 (CH), 61.0 (CH_2), 14.2 (CH_3). IR (ATR-FTIR), cm^{-1}: 3366 (m), 3237 (w), 3130 (w) 1727 (s), 1688 (s), 1659 (s), 1590 (w). HRMS-CI (m/z): $[M + Na]^+$ calcd for $C_7H_8N_2NaO_3S$, 223.0148; found, 223.12.

*Synthesis of the carboxylic acid **30b** from the α-ketoimine **25**.*

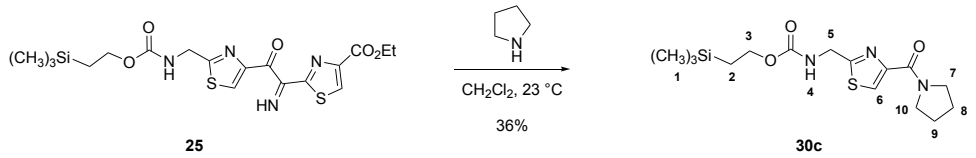


25 30b

Saturated aqueous sodium bicarbonate solution (500 μL) was added to a solution of the α-ketoimine **25** (12.3 mg, 26.2 μmol, 1 equiv) in tetrahydrofuran (500 μL) at 23 °C. The reaction mixture was stirred for 48 h at 23 °C. The pH of the product mixture was adjusted to ~9 by the addition of aqueous sodium hydroxide solution (20% w/v). The basified product mixture was extracted with ether (4 × 2.0 mL). The ether layers were collected and discarded. The pH of the aqueous layer was then adjusted to 3–4 by the dropwise addition of 1 N aqueous hydrogen chloride solution. The acidified aqueous layer was extracted with ether (4 × 2.0 mL). The organic layers were combined and the combined organic layers were dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated to furnish the carboxylic acid **30b** as a white solid (4.0 mg, 51%).

R_f = 0.20 (10% methanol/ethyl acetate; UV, $KMnO_4$). ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.96 (br. s, 1H, H_7), 8.35 (s, 1H, H_6), 8.02 (t, J = 6.1 Hz, 1H, H_4), 4.44 (d, J = 6.1 Hz, 2H, H_5), 4.09 (dd, J = 9.2, 7.4 Hz, 2H, H_3), 0.94 (dd, J = 9.2, 7.4 Hz, 2H, H_2), 0.02 (s, 9H, H_1). ^{13}C NMR (100 MHz, DMSO-d_6) δ 171.1 (C), 162.1 (C), 156.6 (C), 146.8 (C), 128.6 (CH), 62.3 (CH_2), 42.2 (CH_2), 17.3 (CH_2), –1.4 (3 × CH_3). IR (ATR-FTIR), cm^{-1}: 3327 (w), 3125 (w), 2955 (w), 2896 (w), 1723 (s), 1685 (s), 1534 (m), 1254 (s), 1235 (s). HRMS-CI (m/z): $[M + Na]^+$ calcd for $C_{11}H_{18}N_2NaO_4SSi$, 325.0649; found, 325.0682.

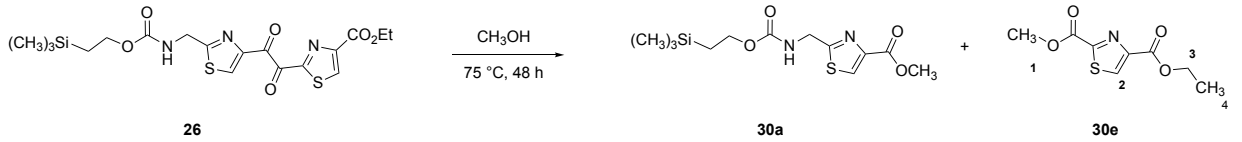
*Synthesis of the pyrrolidinyl amide **30c** from the α-ketoimine **25**.*



A solution of pyrrolidine (22.7 mg, 319 μmol, 11.5 equiv) in dichloromethane (150 μL) was added dropwise to a solution of the α-ketoimine **25** (13.0 mg, 27.7 μmol, 1 equiv) in dichloromethane (650 μL) at 23 °C. The resulting mixture was stirred for 48 h at 23 °C. The product mixture was concentrated. The residue obtained was purified by preparative thin-layered chromatography (eluting with ethyl acetate) to provide the amide **30c** as a colorless oil (3.5 mg, 36%).

R_f = 0.25 (ethyl acetate; UV, $KMnO_4$). ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.94 (s, 1H, H_6), 4.66 (d, J = 6.2 Hz, 2H, H_5), 4.20 (app t, J = 8.5 Hz, 2H, H_3), 3.84 (t, J = 6.6 Hz, 2H, H_7 or H_{10}), 3.65 (t, J = 6.6 Hz, 2H, H_7 or H_{10}), 1.97–1.88 (m, 4H, H_8 and H_9), 1.01 (app t, J = 8.6 Hz, 2H, H_2), 0.03 (s, 9H, H_1). ^{13}C NMR (150 MHz, $CDCl_3$) δ 167.5 (C), 161.7 (C), 156.7 (C), 151.5 (C), 124.9 (CH), 63.97 (CH_2), 49.2 (CH_2), 47.1 (CH_2), 42.8 (CH_2), 26.7 (CH_2), 24.2 (CH_2), 17.9 (CH_2), –1.3 (3 × CH_3). IR (ATR-FTIR), cm^{-1}: 2952 (w), 1716 (m), 1607 (m), 1504 (m), 1247 (s). HRMS-CI (m/z): $[M + H]^+$ calcd for $C_{15}H_{26}N_3O_3SSi$, 356.1459; found, 356.1482.

*Synthesis of the methyl ester **30a** and the diester **30e** from the α-diketone **26**.*



26 **30a** **30e**

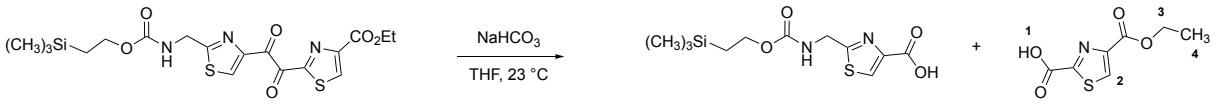
A solution of the α-diketone **26** (8.0 mg, 17.0 μmol, 1 equiv) in methanol (600 μL) was heated for 48 h at 75 °C in a sealed vial fitted with a Teflon-lined cap. The product mixture was cooled over 30 min to 23 °C. The cooled product mixture was concentrated. The residue obtained was purified by preparative thin-layer chromatography (eluting with 60% ethyl acetate–hexanes) to furnish separately the methyl ester **30a** (2.4 mg, 45%, colorless oil) and the diester **30e** (1.5 mg, 41%, colorless oil).

Spectroscopic data for the methyl ester **30a** obtained in this way were identical to that reported above.

Diester **30e**:

R_f = 0.20 (15% ethyl acetate–hexanes; UV, $KMnO_4$). ^1H NMR (500 MHz, CD_3OD-d_4) δ 8.67 (s, 1H, H_2), 4.41 (q, J = 7.1 Hz, 2H, H_3), 4.01 (s, 3H, H_1), 1.40 (t, J = 7.1 Hz, H_4). ^1H NMR (150 MHz, CD_3OD-d_4) δ 162.1 (C), 160.98 (C), 160.1 (C), 149.6 (C), 134.3 (CH), 62.8 (CH_2), 53.9 (CH_3), 14.5 (CH_3). IR (ATR-FTIR), cm^{-1}: 3096 (w), 2923 (w), 2851 (w), 1719 (s), 1716 (s), 1466 (m), 1237 (s), 1212 (s). HRMS-CI (m/z): $[M + H]^+$ calcd for $C_8H_{10}NO_4S$, 216.0325; found, 216.0376.

*Synthesis of the carboxylic acids **30b** and **30f** from the α-diketone **26**.*



26 **30b** **30f**

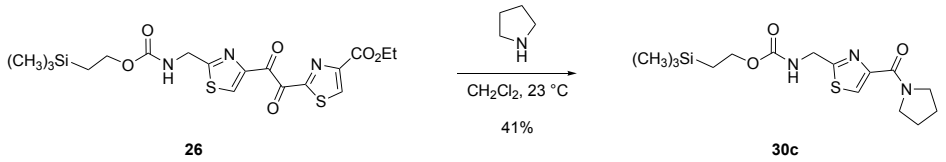
Saturated aqueous sodium bicarbonate solution (500 μL) was added to a solution of the α-diketone **26** (10.6 mg, 22.6 μmol mmol, 1 equiv) in tetrahydrofuran (500 μL) at 23 °C. The resulting mixture was stirred for 72 h at 23 °C. The product mixture was transferred to a separatory funnel and washed with ether (4 × 2.0 mL). The ether layers were collected and discarded. The pH of the aqueous layer was adjusted to 3–4 by the dropwise addition of 1 N aqueous hydrogen chloride solution. The acidified aqueous layer was extracted with ether (4 × 2.0 mL). The organic layers were combined and the combined organic layers were dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated to provide a 2:1 mixture of the carboxylic acids **30b** and **30f** (characterized by ^{1}H NMR analysis).

Spectroscopic data for the carboxylic acid **30b** were identical to that reported above.

Carboxylic acid **30f** (characterized in solution):

^{1}H NMR (500 MHz, CD_3OD-d_4) δ 8.62 (s, 1H, H_2), 4.39 (q, J = 7.1 Hz, 2H, H_3), 1.39 (t, J = 7.1 Hz, H_4). ^{1}H NMR (150 MHz, CD_3OD-d_4) δ 162.2 (C), 162.0 (C), 161.9 (C), 149.4 (C), 134.2 (CH), 62.8 (CH_2), 14.5 (CH_3). HRMS-CI (m/z): $[M + H]^+$ calcd for $C_7H_8NO_4S$, 202.0169; found, 202.0179.

*Synthesis of pyrrolidinyl amide **30c** from the α-diketone **26**.*

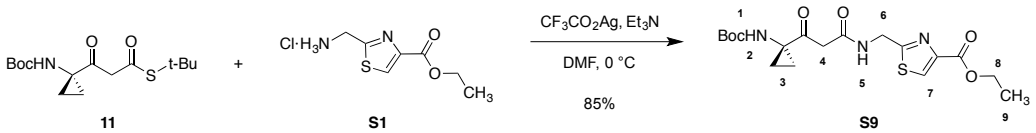


26 **30c**

Pyrrolidine (22.7 mg, 319 μmol, 14.0 equiv) was added dropwise via syringe to a stirred solution of the α-diketone **26** (10.8 mg, 23.0 μmol, 1 equiv) in dichloromethane (900 μL) at 23 °C. The reaction mixture was stirred for 48 h at 23 °C. The product mixture was concentrated and the residue obtained was purified by preparative thin-layered chromatography (eluting with ethyl acetate) to provide the amide **30c** as a colorless oil (3.4 mg, 41%).

Spectroscopic data for amide **30c** were identical to that reported above.

*Synthesis of β-ketoamide **S9**.*

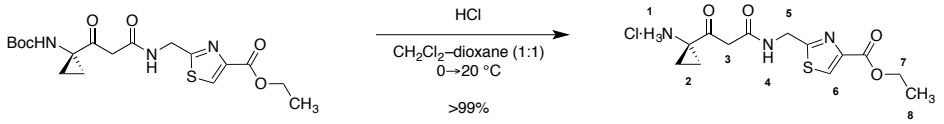


A solution of silver trifluoroacetate (30.3 mg, 140 μmol, 1.40 equiv) in *N,N*-dimethylformamide (400 μL) was added to a solution of triethylamine (55.0 μL, 390 μmol, 4.00 equiv), the ammonium ion **S1** (21.8 mg, 100 μmol, 1 equiv), and the β-ketothioester **11** (37.1 mg, 120 μmol, 1.20 equiv) in *N,N*-dimethylformamide (3.5 mL) at 0 °C. The reaction vessel was covered with foil to exclude light. The reaction mixture was stirred for 1 h at 0 °C. The heterogeneous product mixture was diluted with ethyl acetate (75 mL) and the diluted product mixture was transferred to a separatory funnel. The solution was washed sequentially with saturated aqueous ammonium chloride solution (3 × 40 mL) and saturated aqueous sodium chloride solution (50 mL). The washed organic layer was dried over sodium sulfate. The dried solution was filtered and the filtrate was concentrated. This residue obtained was purified by automated flash-column chromatography (eluting with hexanes initially, grading to ethyl acetate) to provide the ester **S9** as a colorless residue (34.2 mg, 85%).

R_f = 0.45 (80% ethyl–hexanes; UV, $KMnO_4$). ^{1}H NMR (600 MHz, dichloromethane-d_2) δ 8.10 (s, 1H, H_7), 7.61 – 7.54 (m, 1H, H_5), 5.68 (s, 1H, H_2), 4.74 (d, J = 6.0 Hz, 2H, H_6), 4.34 (q, J = 7.1 Hz, 2H, H_8), 3.67 (s, 2H, H_4), 1.60 – 1.56 (m, 2H, H_3), 1.42 (s, 9H, H_1), 1.36 (3, J = 7.1 Hz, 3H, H_9), 1.21 (m, 2H, H_3). ^{13}C NMR (151 MHz, dichloromethane-d_2) δ 206.4 (C), 169.3 (C), 167.0 (C), 161.6 (C), 156.6 (C), 147.3 (C), 128.5 (CH), 113.9 (C), 80.9 (C), 61.8 (CH_2), 45.9 (CH_2), 41.7 (CH_2), 28.6 (CH_3), 21.8 (CH_2), 14.6 (CH_3). IR (ATR-FTIR), cm^{-1}: 2975 (m), 1703 (s), 1662 (s), 1502 (m), 1367 (m), 1239 (s), 1212 (s), 1162 (s), 1064 (m), 1022 (m), 945 (w), 732 (w), 615 (w) 546 (w). HRMS-CI (m/z): $[M + H]^+$ calcd for $C_{18}H_{26}N_3O_6S$, 412.1537; found, 412.1552.

*Synthesis of pyridone **S12.***

*Step 1: Synthesis of the ammonium ion **S10**.*

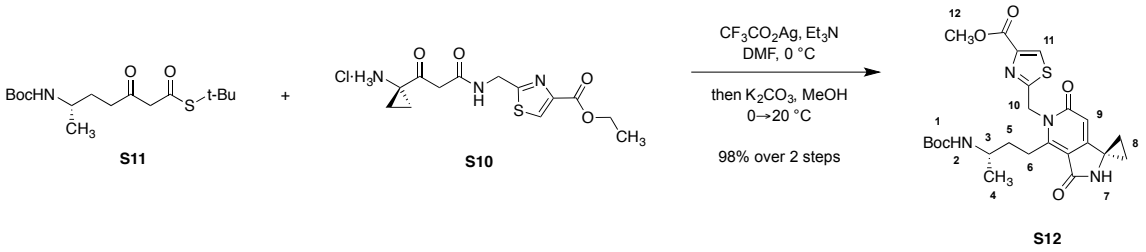


S9 **S10**

A solution of hydrogen chloride in 1,4-dioxane (4.0 N, 1.10 mL, 4.20 mmol, 100 equiv) was added dropwise via syringe to a solution of the ester **S9** (17.3 mg, 42.0 μmol, 1 equiv) in dichloromethane (1.1 mL) at 0 °C. The resulting mixture was immediately warmed to 20 °C. The reaction mixtures was stirred for 3 h at 20 °C. The product mixture was concentrated to provide the ammonium ion **S10** as a pure white solid (14.6 mg, >99%).

Compound has no mobility on silica. ^{1}H NMR (600 MHz, DMSO-d_6) δ 9.14 (t, J = 6.0 Hz, 1H, H_4), 8.78 (s, 3H, H_1), 8.46 (s, 1H, H_6), 4.58 (d, J = 5.8 Hz, 2H, H_5), 4.29 (q, J = 7.1 Hz, 2H, H_7), 3.38 (s, 2H, H_3), 1.82 – 1.74 (m, 2H, H_2), 1.56 – 1.47 (m, 2H, H_2), 1.30 (t, J = 7.1 Hz, 3H, H_8). ^{13}C NMR (151 MHz, DMSO-d_6) δ 199.4 (C), 169.7 (C), 165.9 (C), 160.7 (C), 145.5 (C), 129.4 (CH), 60.7 (CH_2), 42.3 (CH_2), 42.0 (C), 40.5 (CH_2), 14.2 (CH_3), 13.1 (CH_2). IR (ATR-FTIR), cm^{-1}: 3217 (w), 3128 (w), 2923 (m), 2854 (m), 2662 (w), 1734 (s), 1705 (s), 1683 (s), 1544 (m), 1295 (s), 1225 (s), 1121 (m), 1093 (m), 889 (w), 869 (s), 755 (m), 640 (m), 517 (w).

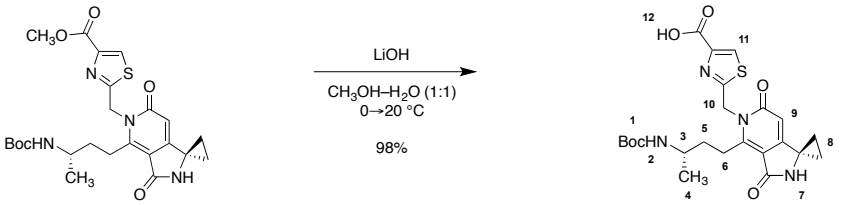
*Step 2: Synthesis of the pyridone **S12**.*



A solution of silver trifluoroacetate (13.0 mg, 59 μmol, 1.40 equiv) in *N,N*-dimethylformamide (200 μL) was added to a solution of triethylamine (23.0 μL, 170 μmol, 4.00 equiv), the ammonium ion **S10** (14.6 mg, 42.0 μmol, 1 equiv), and the β-ketothioester **S11** (16.7 mg, 50.0 μmol, 1.20 equiv) in *N*,*N*-dimethylformamide (1.2 mL) at 0 °C. The reaction vessel was covered with foil to exclude light. The reaction mixture was stirred for 30 min at 0 °C. Methanol (1.7 mL) and anhydrous potassium carbonate (58.0 mg, 420 μmol, 10.0 equiv) were then added in sequence. The reaction vessel was removed from the cooling bath and wrapped with aluminum foil to exclude light. The mixture was stirred for 12 h at 20 °C. The heterogeneous product mixture was diluted with ethyl acetate (75 mL). The diluted solution was washed sequentially with 1 N aqueous hydrogen chloride solution (40 mL) and saturated aqueous sodium chloride solution (50 mL). The washed organic layer was dried over sodium sulfate and the dried solution was filtered. The filtrate was concentrated. The residue obtained was purified via automated flash-column chromatography (eluting with dichloromethane initially, grading to 10% methanol–dichloromethane) to provide the pyridone **S12** as a white solid (20.7 mg, 98%).

R_f = 0.30 (ethyl acetate; UV, $KMnO_4$). ^{1}H NMR (600 MHz, dichloromethane-d_2) δ 8.16 (s, 1H, H_{11}), 6.38 (s, 1H, H_7), 6.00 (s, 1H, H_9), 5.63 (d, J = 15.1 Hz, 1H, H_{10}), 5.60 (s, 1H, H_2), 5.51 (d, J = 15.1 Hz, 1H, H_{10}), 3.89 (s, 3H, H_{12}), 3.80 – 3.70 (m, 1H, H_3), 3.46 (t, J = 8.0 Hz, 2H, H_6), 1.82 – 1.76 (m, 1H, H_5), 1.62 – 1.58 (m, 1H, H_5), 1.50 – 1.47 (m, 2H, H_8), 1.42 (s, 9H, H_1), 1.35 – 1.32 (m, 2H, H_8), 1.15 (d, J = 6.5 Hz, 3H, H_4). ^{13}C NMR (151 MHz, dichloromethane-d_2) δ 168.4 (C), 165.6, (C), 163.2 (C), 162.0 (C), 160.5 (C), 156.3 (C), 154.5 (C), 146.6 (C), 130.2 (CH), 110.4 (C), 103.8 (CH), 79.0 (C), 52.7 (CH_3), 47.1 (CH), 45.4 (CH_2), 40.6 (C), 36.2 (CH_2), 28.8 (CH_3), 25.5 (CH_2), 21.7 (CH_3), 16.3 (CH_2), 16.3 (CH_2). IR (ATR-FTIR), cm^{-1}: 2975 (m), 1696 (s), 1652 (s), 1574 (m), 1245 (m), 1183 (m), 1063 (w), 838 (w), 779 (m), 570 (m). HRMS-CI (m/z): [M + Na]$^+$ calcd for $C_{24}H_{30}N_4NaO_6S$, 525.1778; found, 525.1758.

*Synthesis of the carboxylic acid **S13**.*

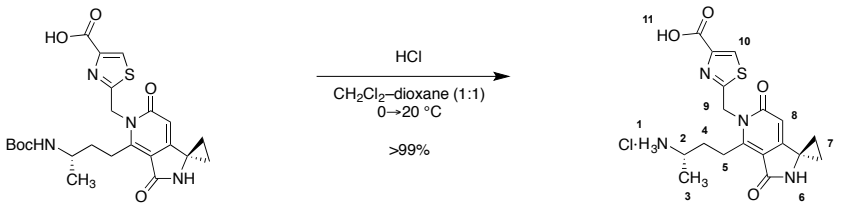


S12 **S13**

Lithium hydroxide (6.7 mg, 160 μmol, 10.0 equiv) was added to a solution of the pyridone **S12** (8.0 mg, 16.0 μmol, 1 equiv) in methanol (200 μL) and water (200 μL) at 0 °C. The reaction vessel was removed from the cooling bath. The mixture was stirred for 25 min at 20 °C. The product mixture was then cooled to 0 °C. The pH of the mixture was adjusted to ~ 2 by the slow addition of 1 N aqueous hydrogen chloride solution. The acidified solution was transferred to a separatory funnel and extracted with ethyl acetate (2 × 20 mL). The organic layers were combined and the combined organic layer was washed with saturated aqueous sodium chloride solution (20 mL). The washed solution was dried over sodium sulfate and the dried solution was filtered. The filtrate was concentrated to provide the carboxylic acid **S13** as a white solid (7.6 mg, 98%).

R_f = 0.29 (10% methanol–dichloromethane; UV, $KMnO_4$). ^{1}H NMR (600 MHz, DMSO-d_6) δ 13.01 (s, 1H, H_{12}), 8.46 (s, 1H, H_7), 8.39 (s, 1H, H_{11}), 6.77 (d, J = 8.3 Hz, 1H, H_2), 6.14 (s, 1H, H_9), 5.56 (d, J = 16.2 Hz, 1H, H_{10}), 5.45 (d, J = 16.1 Hz, 1H, H_{10}), 3.55 (p, J = 6.8 Hz, 1H, H_3), 3.51 – 3.41 (m, 1H, H_6), 3.17 – 3.08 (m, 1H, H_6), 1.62 (m, 2H, H_5), 1.38 – 1.36 (m, 2H, H_8), 1.35 – 1.33 (m, 2H, H_8), 1.32 (s, 9H, H_1), 1.04 (d, J = 6.6 Hz, 3H, H_4). ^{13}C NMR (151 MHz, DMSO-d_6) δ 166.7 (C), 165.7 (C), 161.8 (C), 161.7 (C), 159.8 (C), 155.1 (C), 153.0 (C), 146.6 (C), 129.5 (CH), 109.6 (C), 103.3 (CH), 77.4 (C), 46.0 (CH), 44.2 (CH_2), 40.1 (C), 35.5 (CH_2), 28.2 (CH_3), 24.3 (CH_2), 20.8 (CH_3), 15.2 (CH_2). IR (ATR-FTIR), cm^{-1}: 2971 (m), 2926 (m), 1693 (s), 1651 (s), 1573 (m), 1520 (m), 1365 (m), 1335 (m), 1248 (m), 1168 (m), 1025 (m), 995 (m), 837 (m), 762 (m), 728 (m), 570 (m). HRMS-CI (m/z): $[M + Na]^+$ calcd for $C_{23}H_{28}N_4NaO_6S$, 511.1622 found, 511.1625.

*Synthesis of the ammonium ion **35**.*



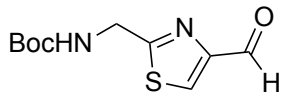
35

A solution of hydrogen chloride in 1,4-dioxane (4.0 N, 250 µL, 1.00 mmol, 489 equiv) was added dropwise via syringe to a solution of the carboxylic acid **S13** (1.0 mg, 2.0 µmol, 1 equiv) in dichloromethane (250 µL) at 0 °C. The reaction vessel was removed from the cooling bath and the mixture was stirred for 1 h at 20 °C. The product mixture was concentrated to provide the ammonium ion **35** as a white solid (0.8 mg, >99%).

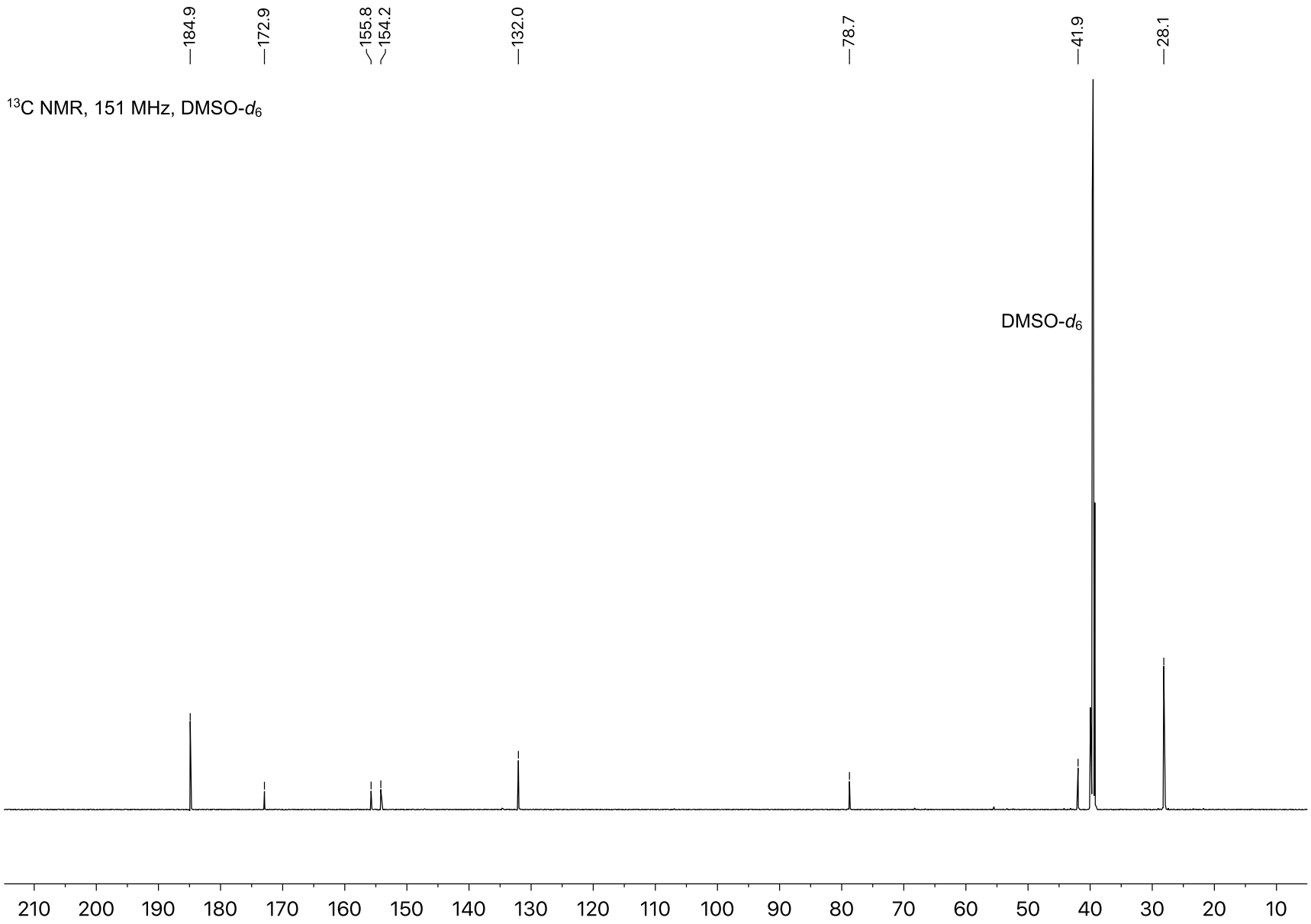
R_f: Compound has no mobility on silica. ^{1}H NMR (600 MHz, DMSO-d_6) δ 8.62 (s, 1H, H_6), 8.38 (s, 1H, H_{10}), 8.02 (s, 3H, H_1), 6.17 (s, 1H, H_8), 5.54 (d, J = 16.0 Hz, 1H, H_9), 5.50 (d, J = 15.9 Hz, 1H, H_9), 3.51 – 3.43 (m, 1H, H_5), 3.39 – 3.32 (m, 1H, H_5), 3.26 – 3.22 (m, 1H, H_2), 1.91 – 1.83 (m, 1H, H_4), 1.83 – 1.76 (m, 1H, H_4), 1.39 – 1.36 (m, 2H, H_7), 1.35 – 1.31 (m, 2H, H_7), 1.22 (d, J = 6.6 Hz, 3H, H_3). ^{13}C NMR (151 MHz, dmso) δ 166.9 (C), 165.5 (C), 162.0 (C), 161.7 (C), 159.6 (C), 151.7 (C), 146.4 (C), 129.9 (CH), 109.9 (C), 103.8 (CH), 46.5 (CH), 44.4 (CH_2), 40.1 (C), 33.4 (CH_2), 23.2, (CH_2) 18.1 (CH_3), 15.2 (CH_2). IR (ATR-FTIR), cm^{-1}: 3015 (w), 2975 (w), 2361 (w), 2337 (w), 1698 (s), 1652 (m), 1457 (w), 1412 (m), 1138 (s), 953 (m), 824 (m), 763 (m). HRMS-CI (m/z): $[M + H]^+$ calcd for $C_{18}H_{21}N_4O_4S$, 389.1278; found, 389.1280.

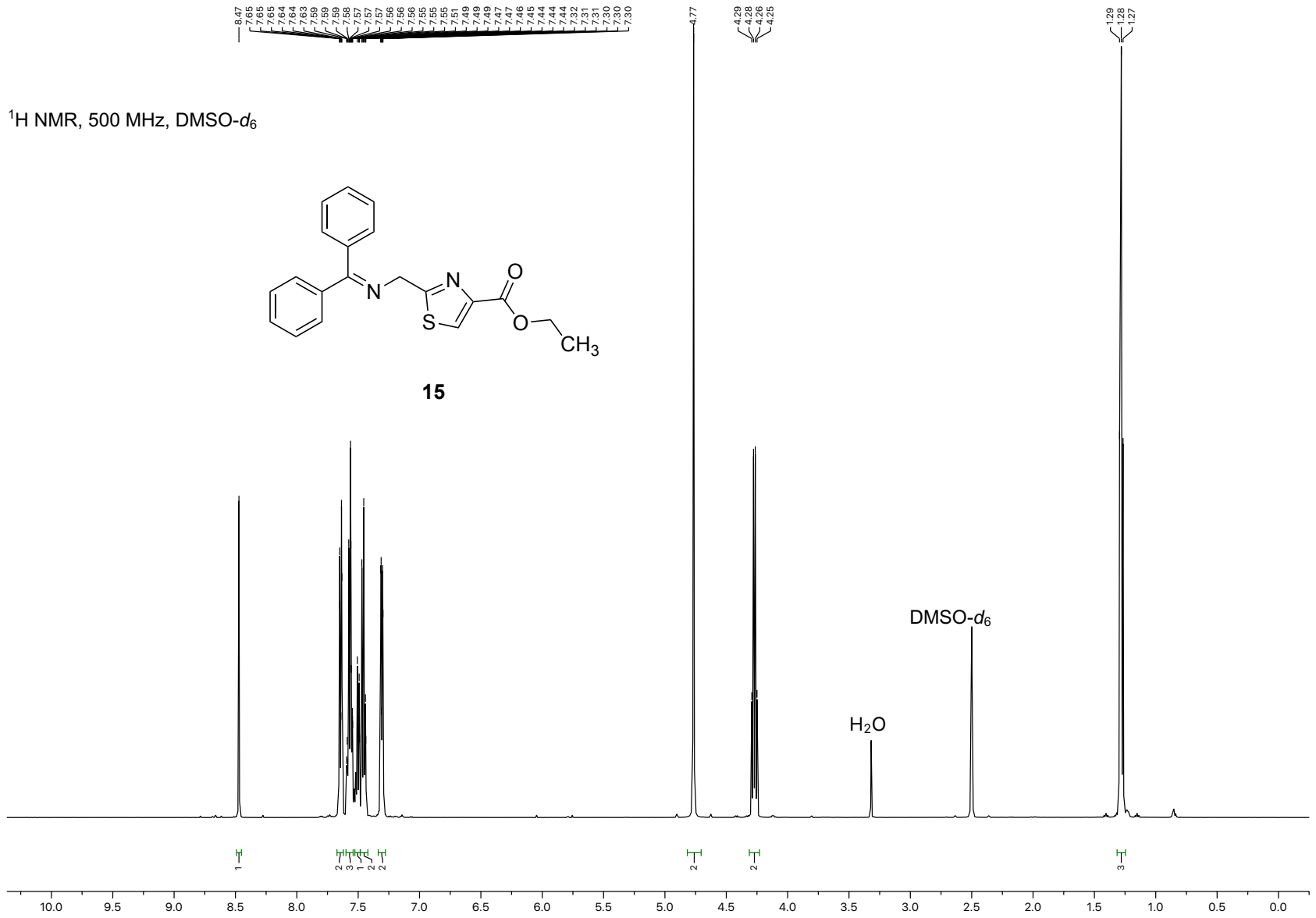
Catalog of nuclear magnetic resonance spectra.

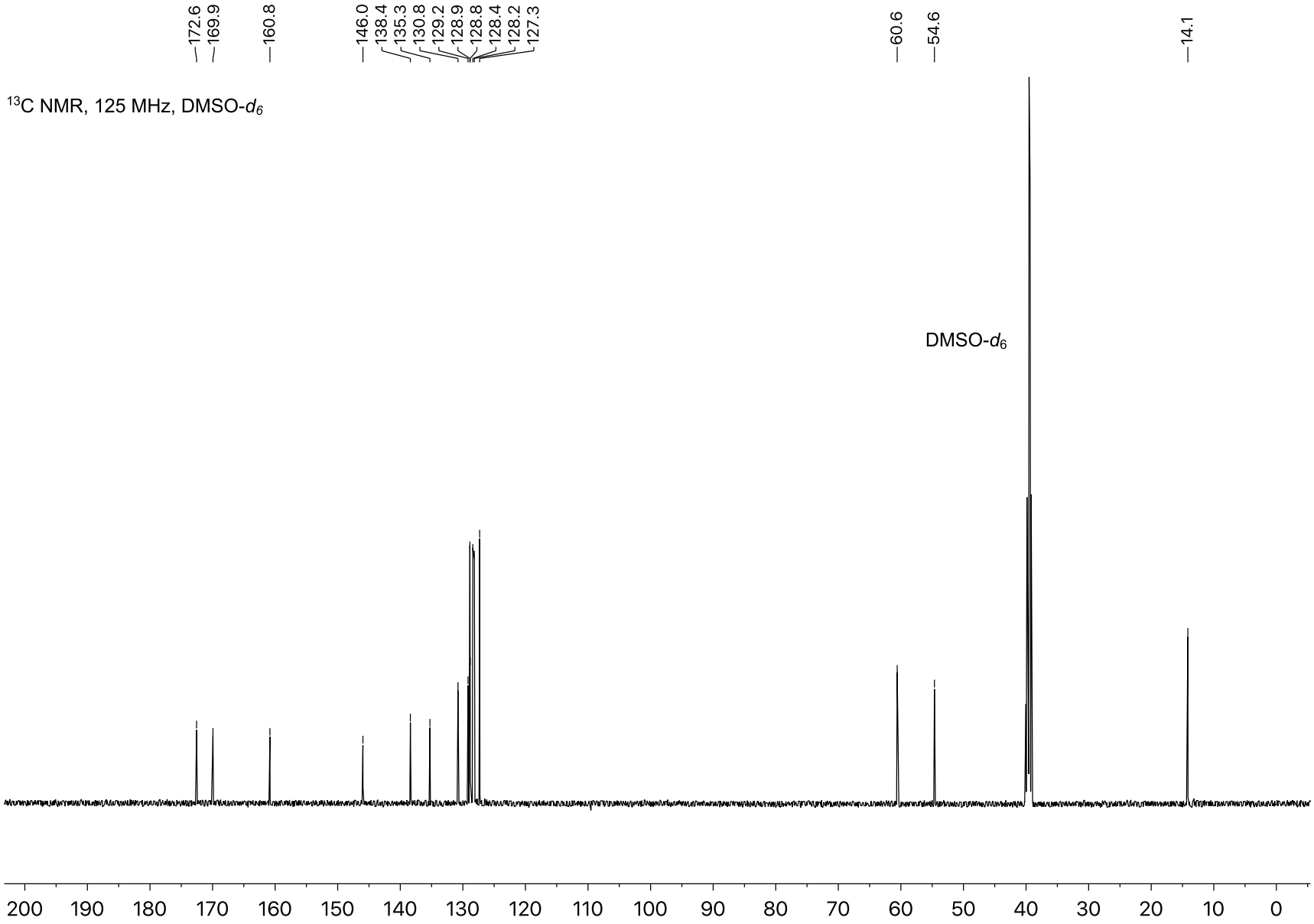
^{1}H NMR, 500 MHz, DMSO-d_6

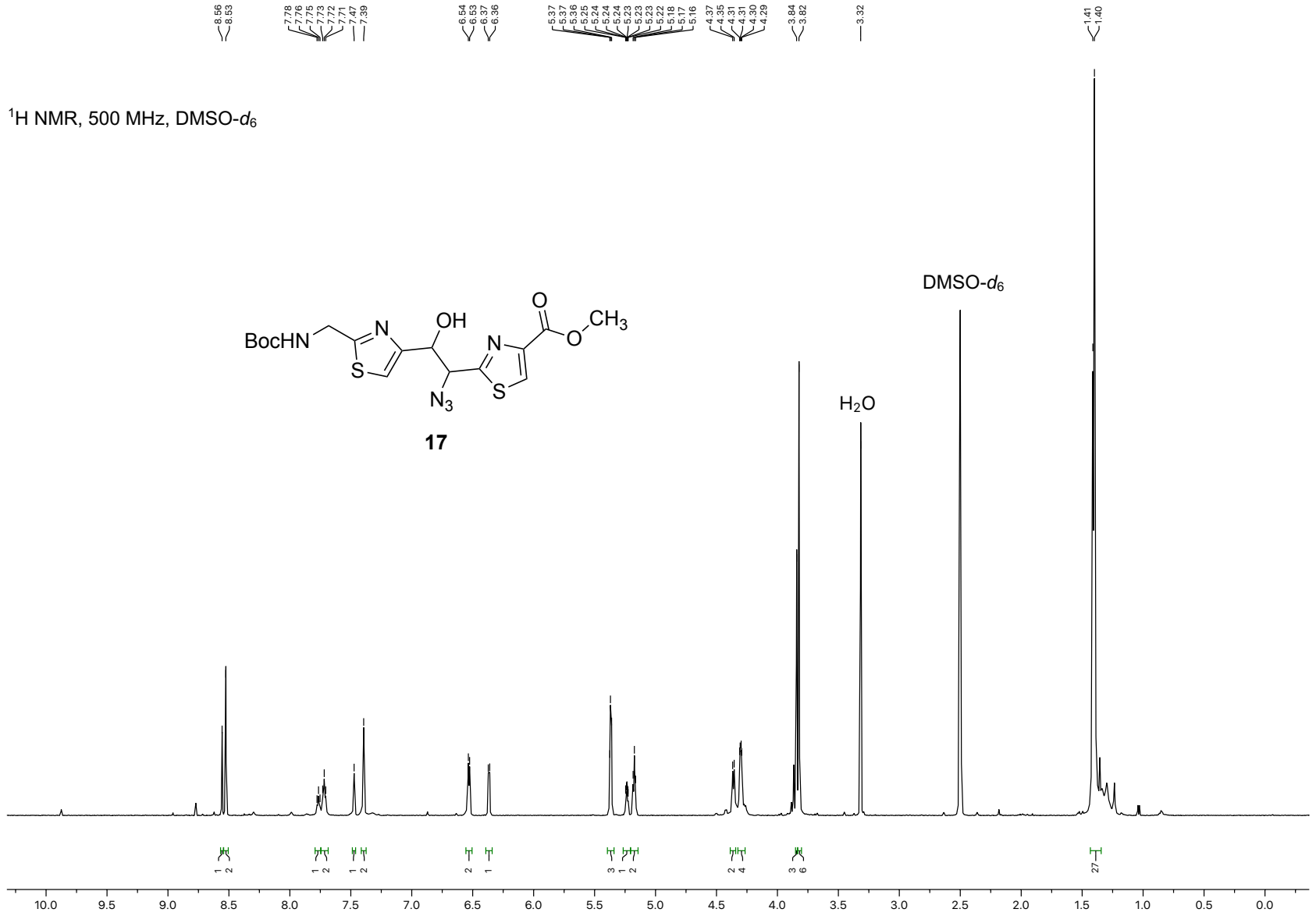


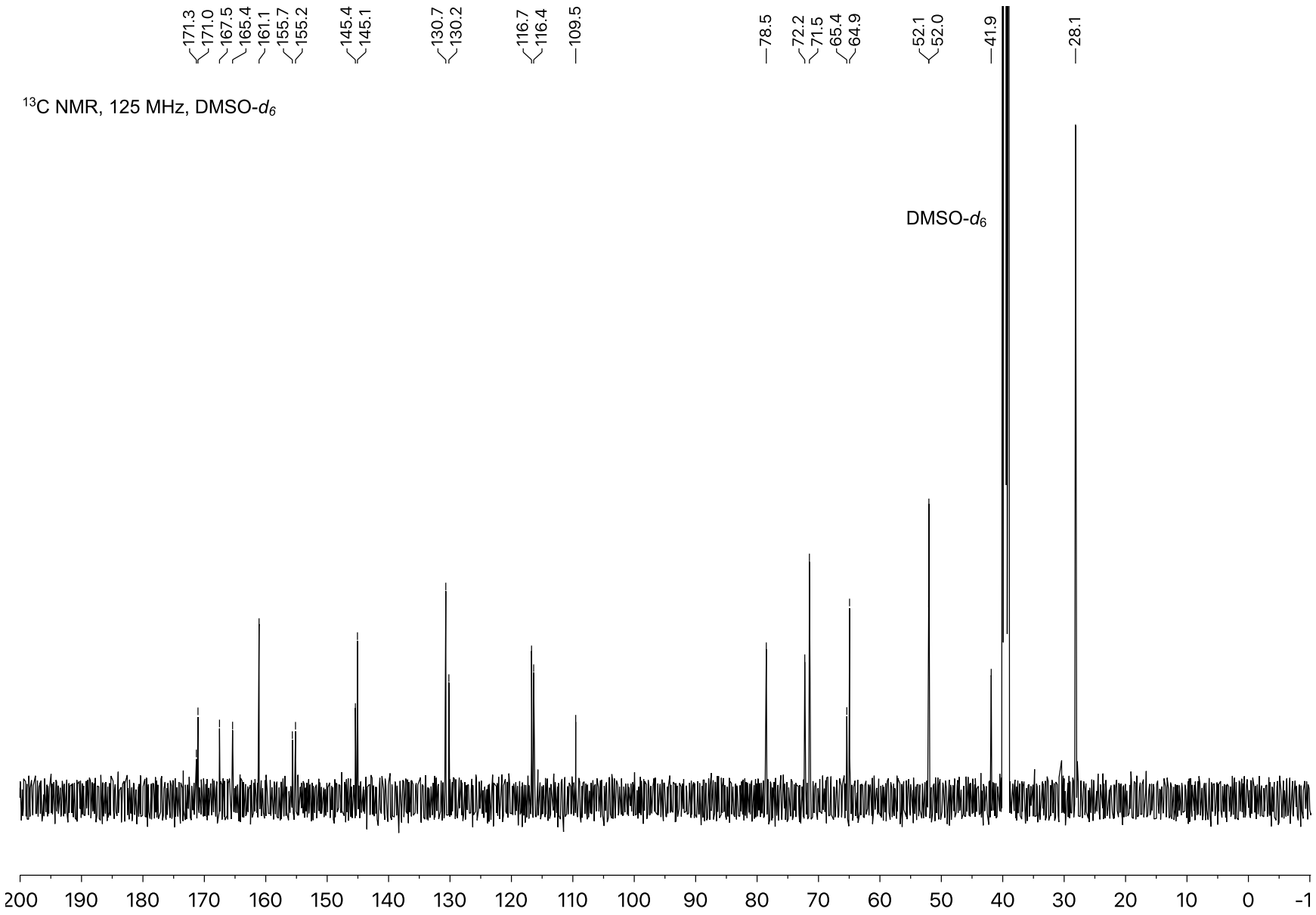
14

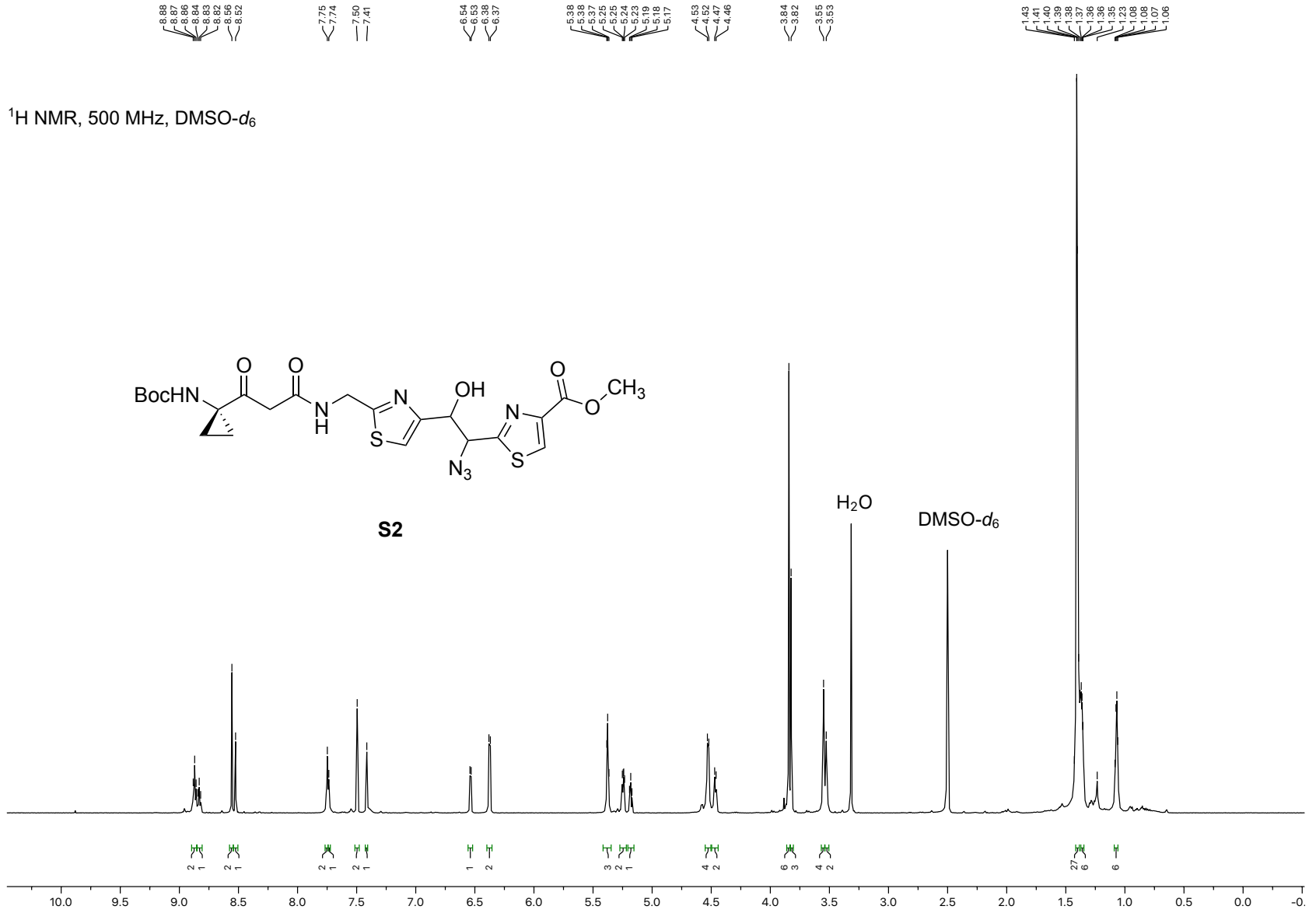

184.9
172.9
155.8
154.2
132.0
78.7
41.9
28.1
^{13}C NMR, 151 MHz, DMSO-d_6
DMSO-d_6
210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10

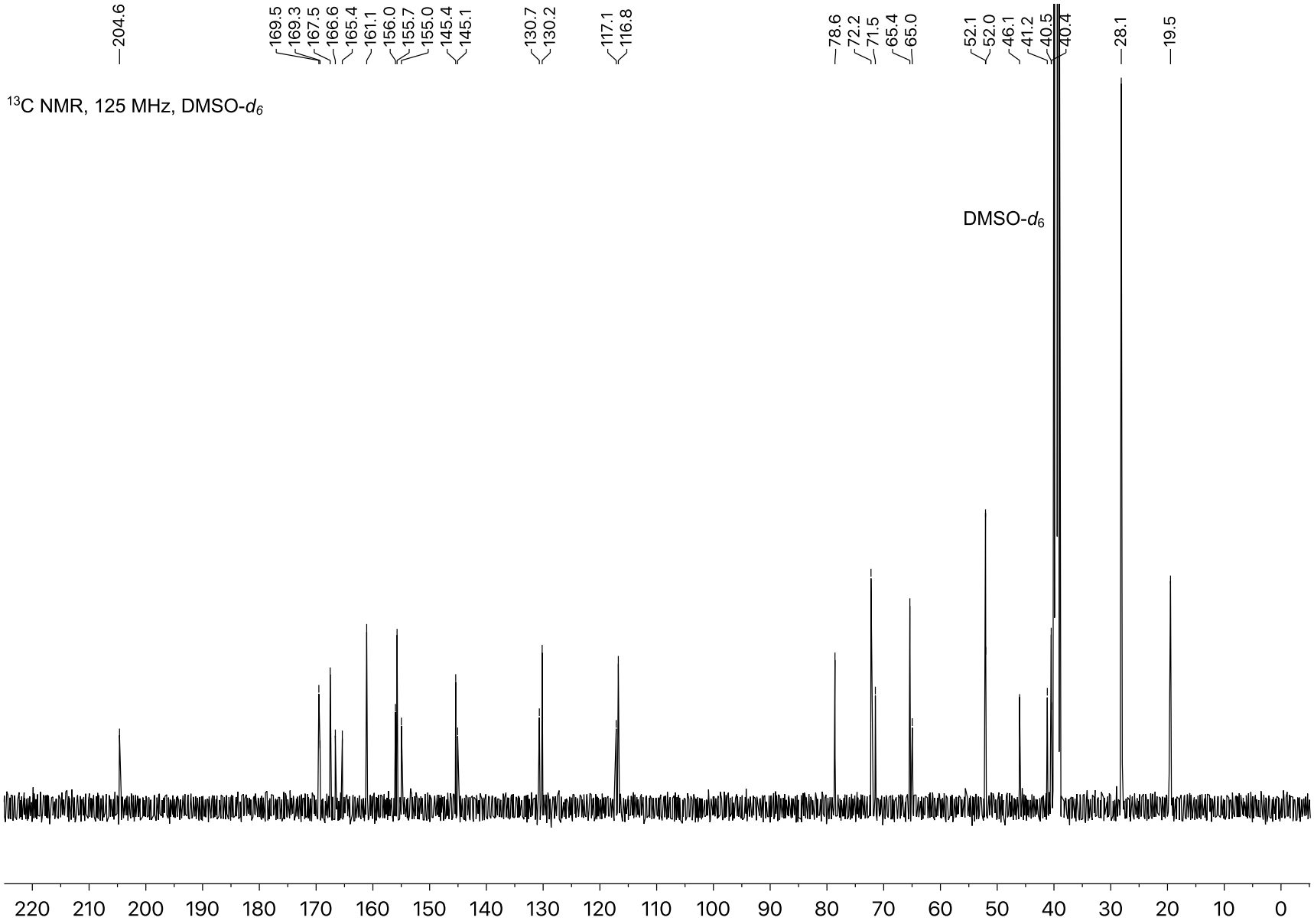

8.47
7.65
7.65
7.65
7.64
7.64
7.63
7.59
7.59
7.59
7.58
7.57
7.57
7.57
7.56
7.56
7.56
7.55
7.55
7.55
7.51
7.49
7.49
7.49
7.47
7.47
7.46
7.45
7.44
7.44
7.44
7.32
7.31
7.31
7.30
7.30
7.30
4.77
4.29
4.28
4.26
4.25
1.29
1.28
1.27
^{1}H NMR, 500 MHz, DMSO-d_6
N
N
S
O
O
CH_3
15
DMSO-d_6
H_2O
1
2
3
1
2
2
2
2
3
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0

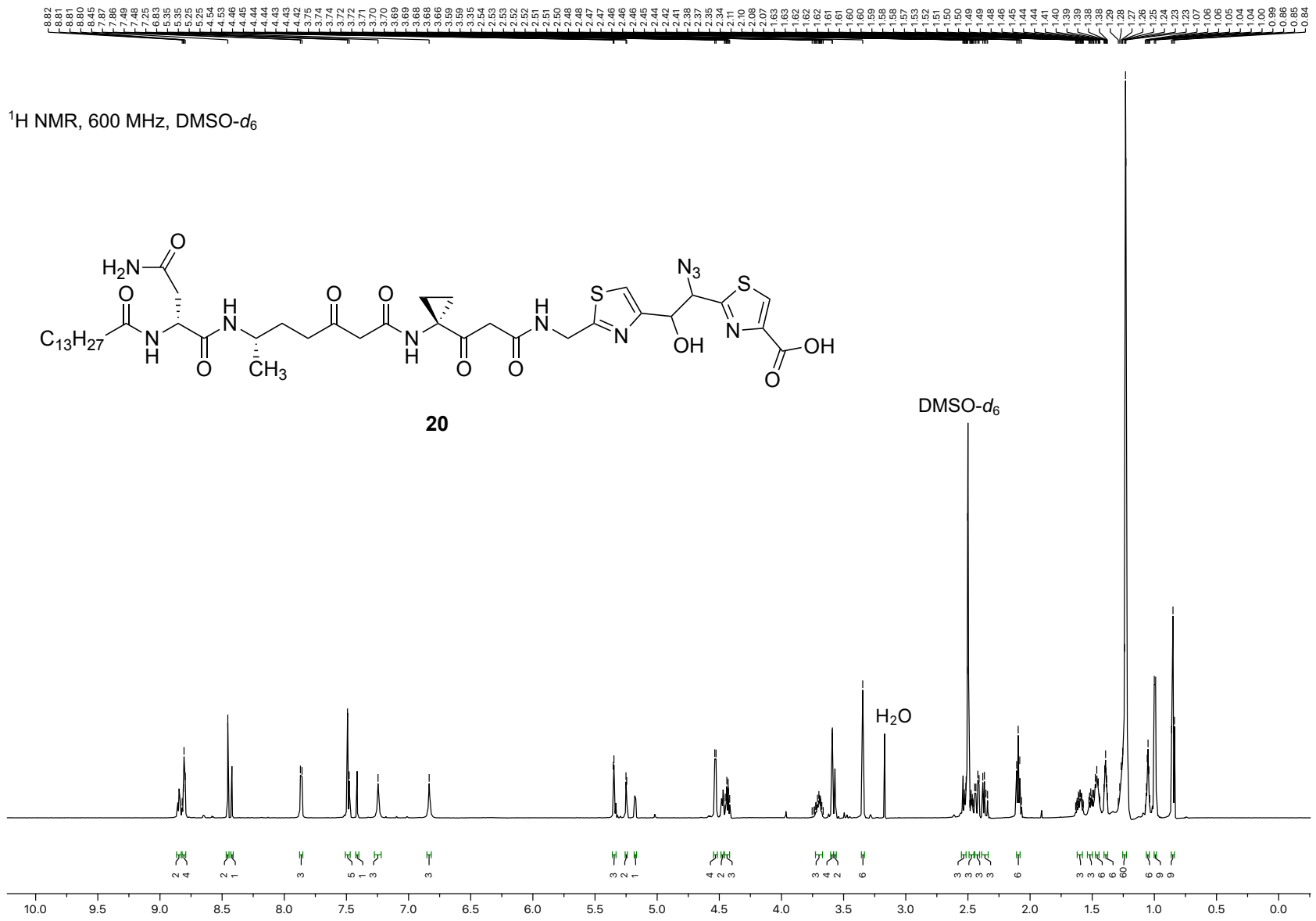

172.6
169.9
160.8
146.0
138.4
135.3
130.8
129.2
128.9
128.8
128.4
128.2
127.3
60.6
54.6
14.1
^{13}C NMR, 125 MHz, DMSO-d_6
DMSO-d_6
200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0

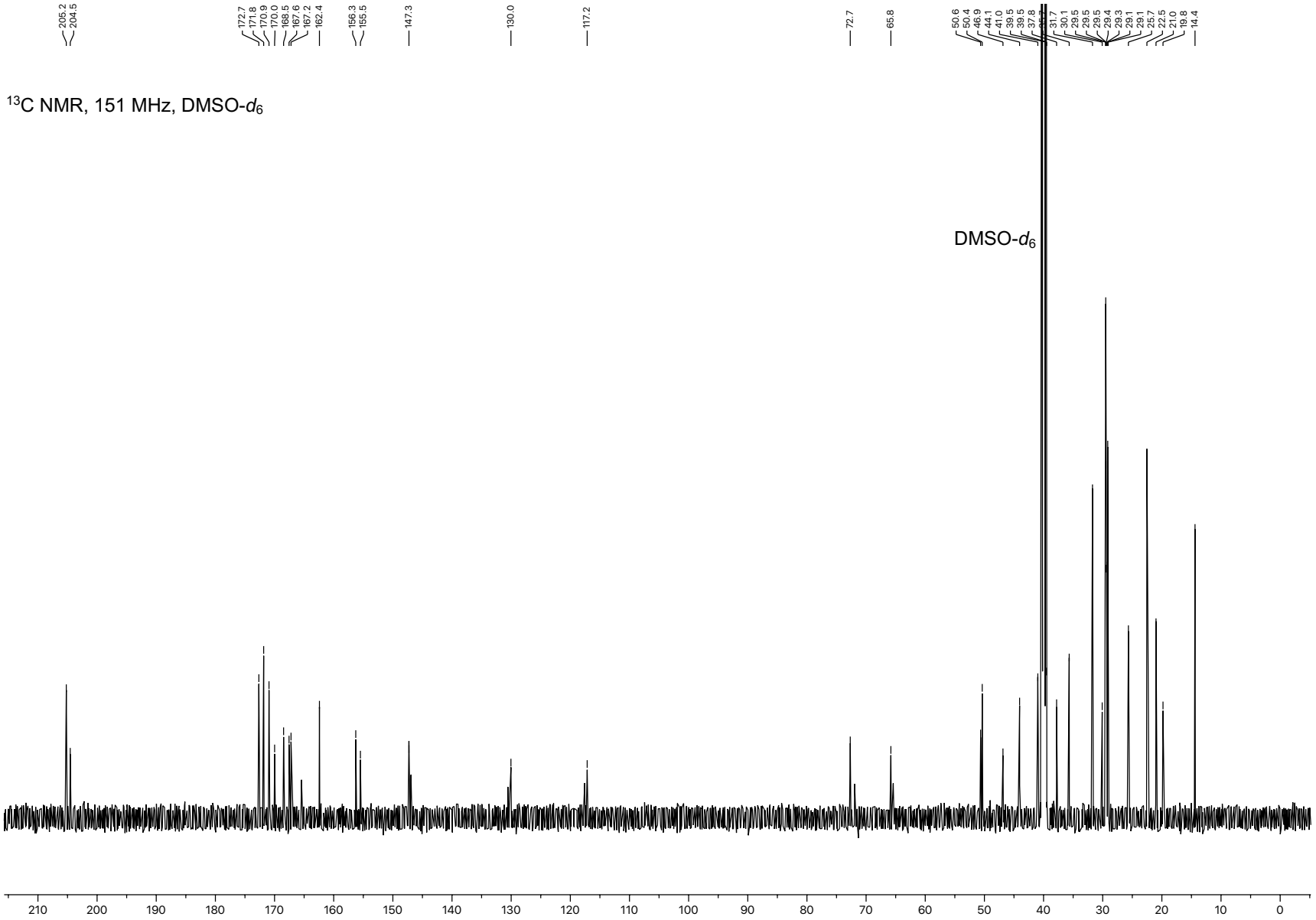

8.56
8.53
7.78
7.76
7.75
7.73
7.72
7.71
7.47
7.39
6.54
6.53
6.37
6.36
5.37
5.37
5.36
5.25
5.24
5.24
5.24
5.23
5.23
5.23
5.22
5.18
5.17
5.16
4.37
4.35
4.31
4.31
4.30
4.29
3.84
3.82
3.32
1.41
1.40
^{1}H NMR, 500 MHz, DMSO-d_6
BocHN
N
S
OH
N$_3$
N
S
O
O
CH$_3$
17
DMSO-d_6
H$_2$O
1 2
1 2
1 2
2 1
3 1 2
2 4
3 6
27
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0

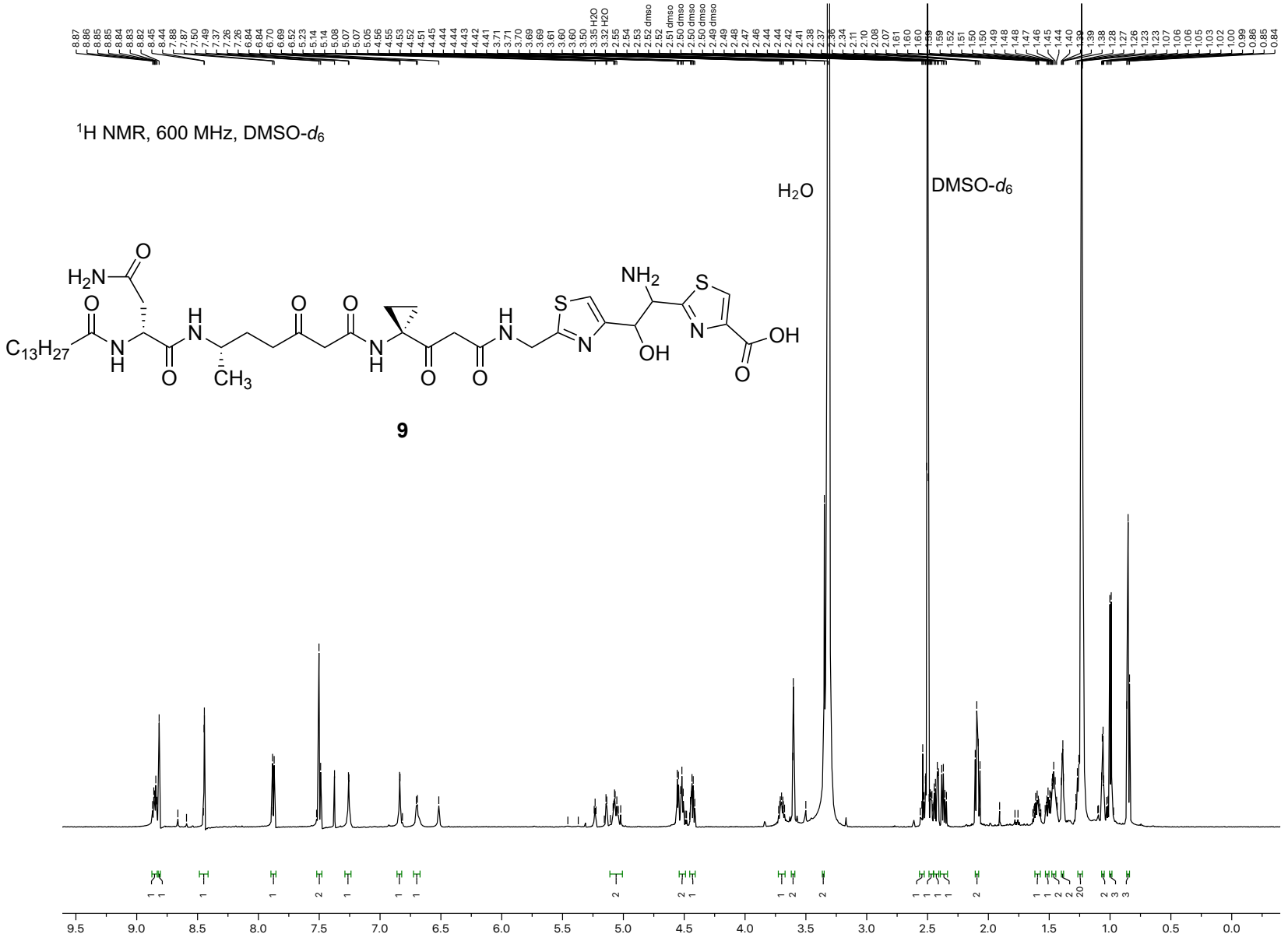

171.3
171.0
167.5
165.4
161.1
155.7
155.2
145.4
145.1
130.7
130.2
116.7
116.4
109.5
78.5
72.2
71.5
65.4
64.9
52.1
52.0
41.9
28.1
^{13}C NMR, 125 MHz, DMSO-d_6
DMSO-d_6
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-1

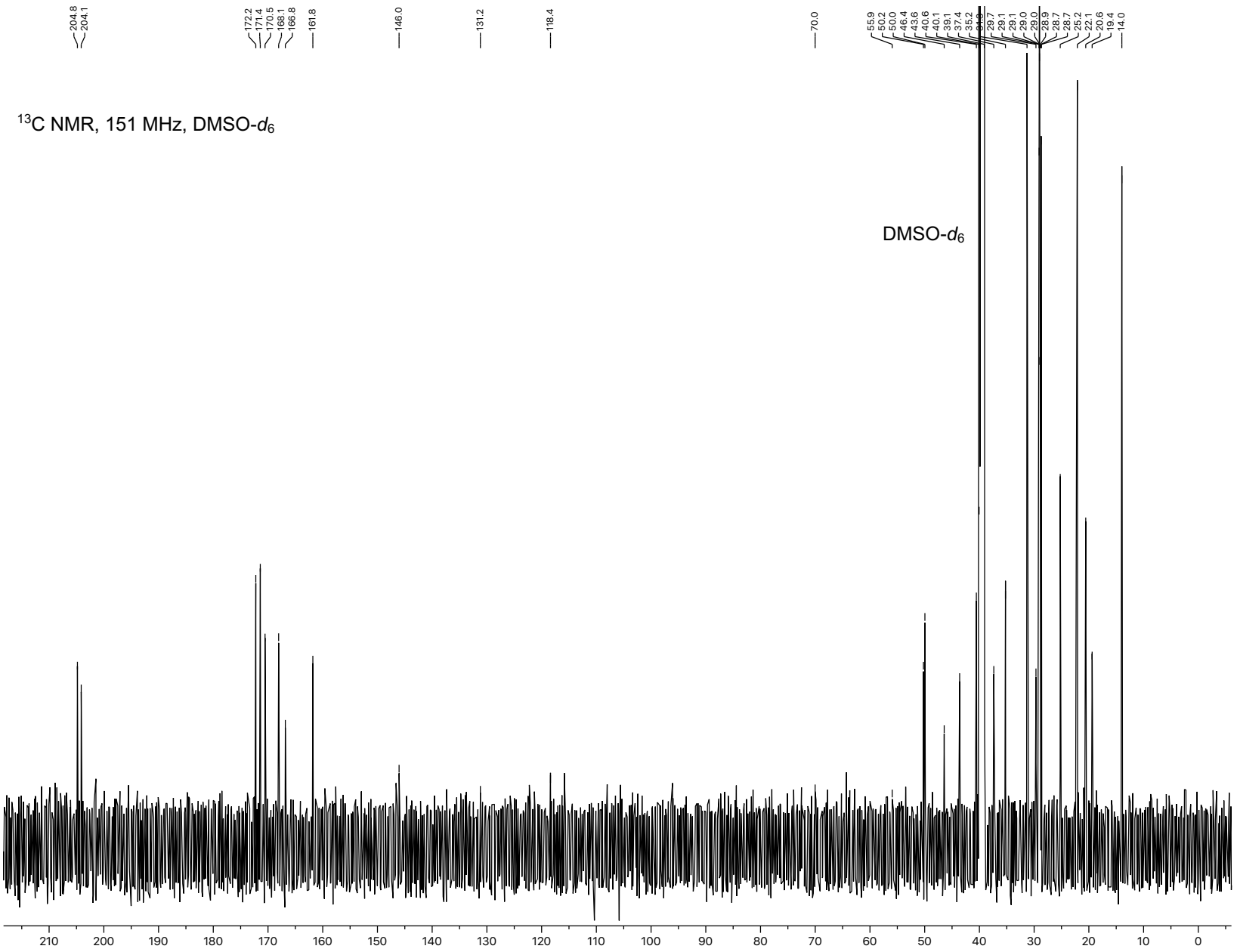

^{1}H NMR, 500 MHz, DMSO-d_6
BocHN
O
O
N
H
N
S
OH
N_3
N
S
O
O
CH_3
S2
H_2O
DMSO-d_6

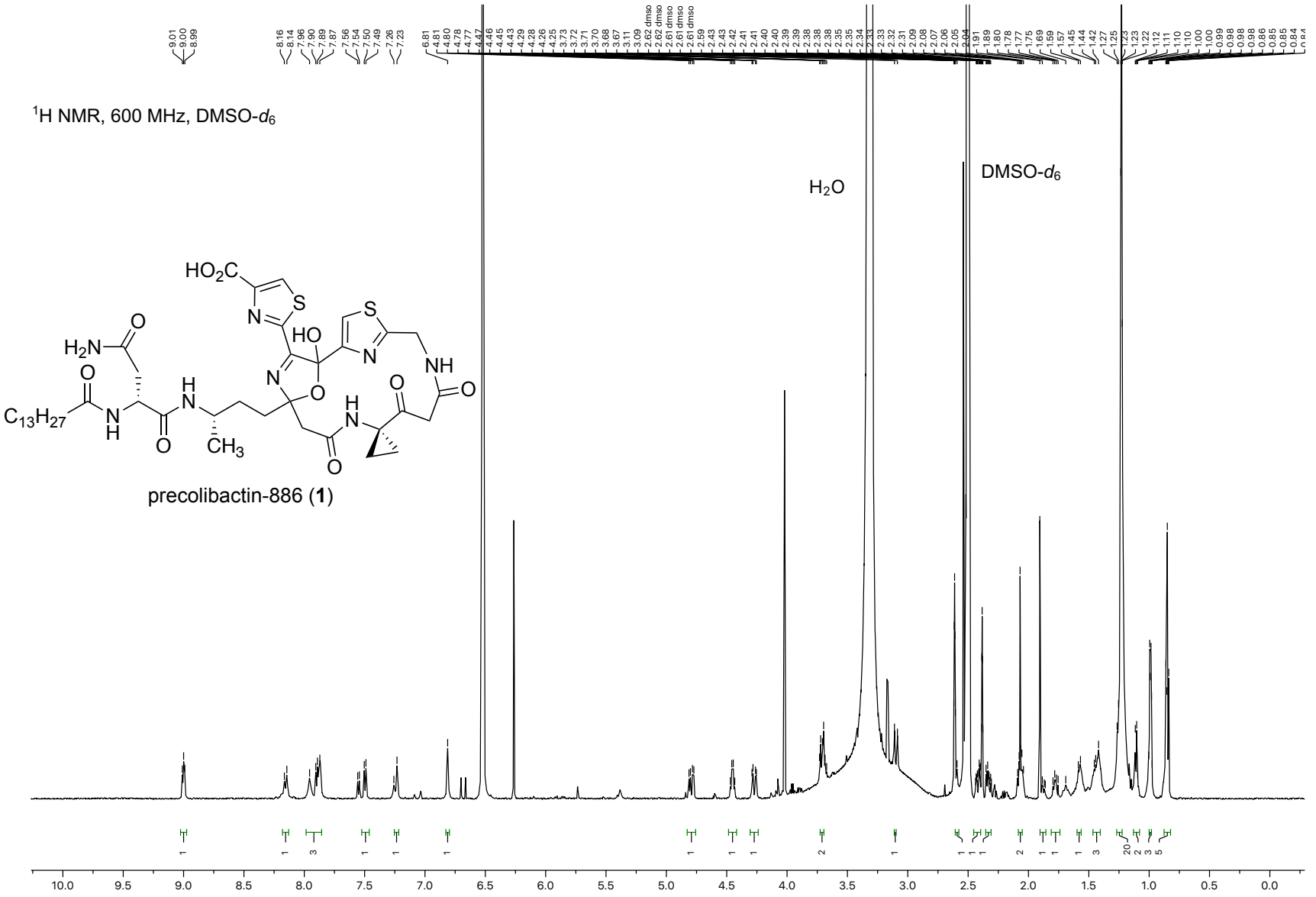

204.6
169.5
169.3
167.5
166.6
165.4
161.1
156.0
155.7
155.0
145.4
145.1
130.7
130.2
117.1
116.8
78.6
72.2
71.5
65.4
65.0
52.1
52.0
46.1
41.2
40.5
40.4
28.1
19.5
13C NMR, 125 MHz, DMSO-d6
DMSO-d6
220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0

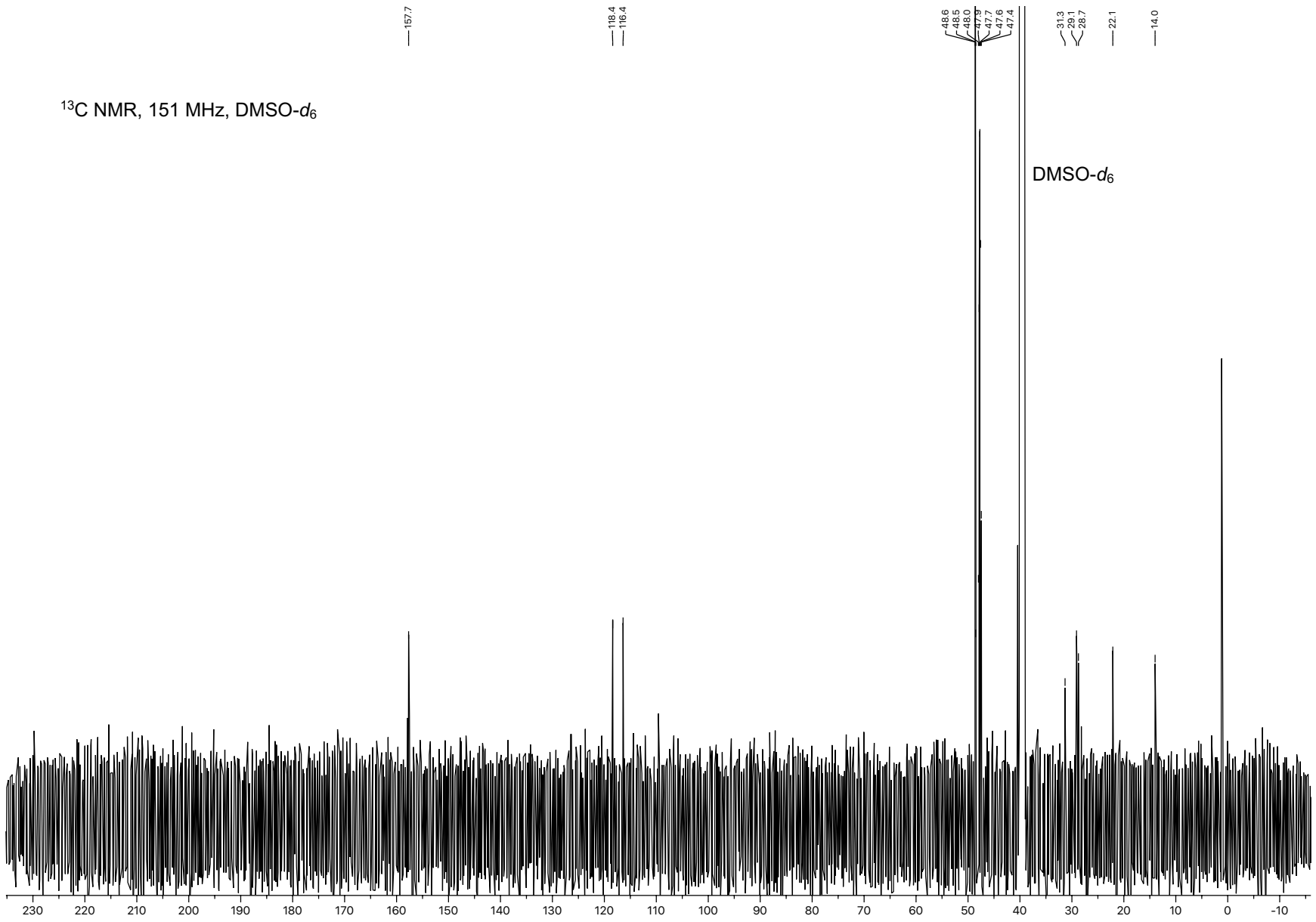

^{1}H NMR, 600 MHz, DMSO-d_6
20
DMSO-d_6
H_2O

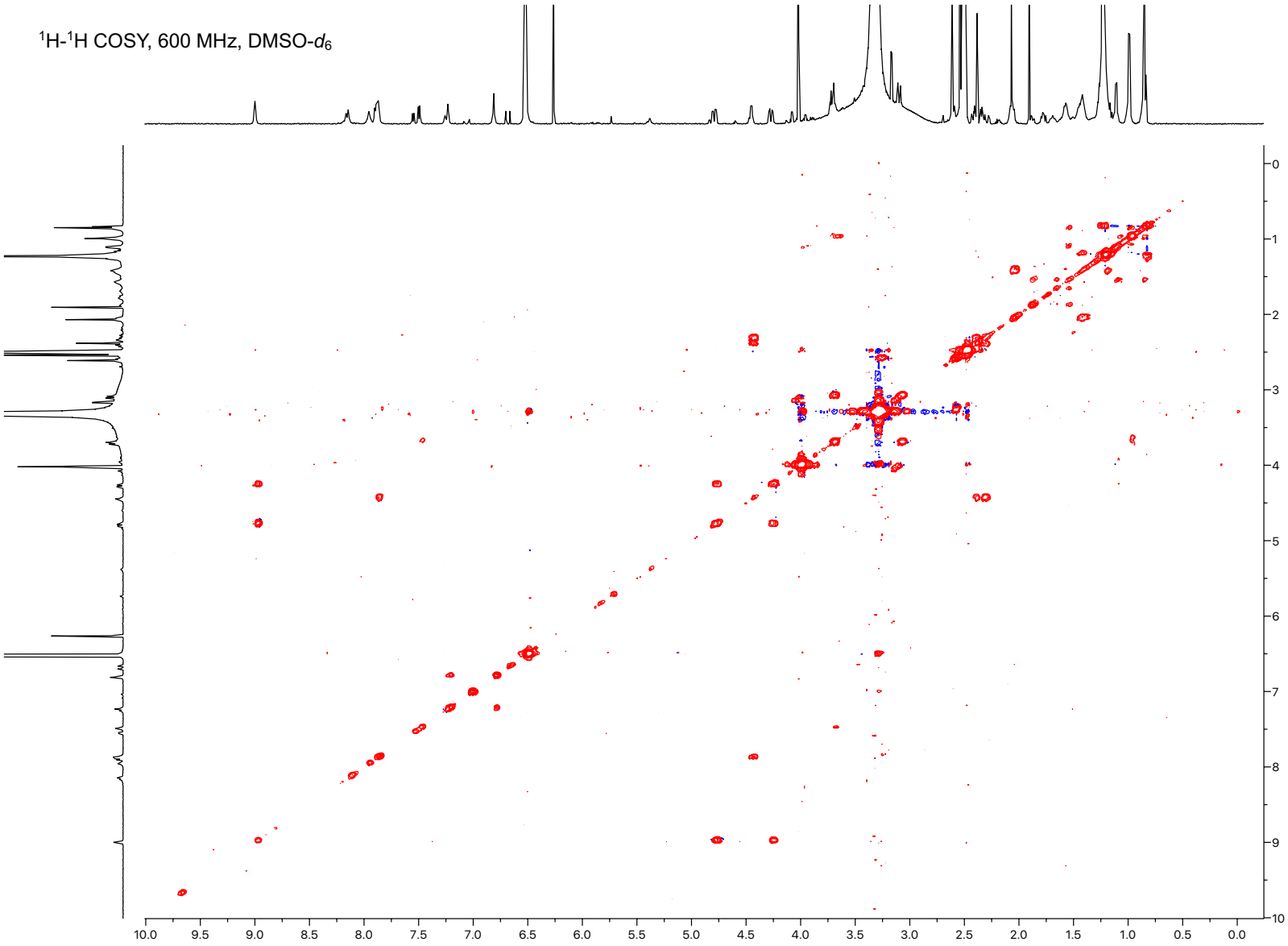

205.2
204.5
172.7
171.8
170.9
170.0
168.5
167.6
167.2
162.4
156.3
155.5
147.3
130.0
117.2
72.7
65.8
50.6
50.4
46.9
44.1
41.0
39.5
39.5
37.8
31.7
30.1
29.5
29.5
29.5
29.4
29.3
29.1
29.1
25.7
22.5
21.0
19.8
14.4
^{13}C NMR, 151 MHz, DMSO-d_6
DMSO-d_6
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0

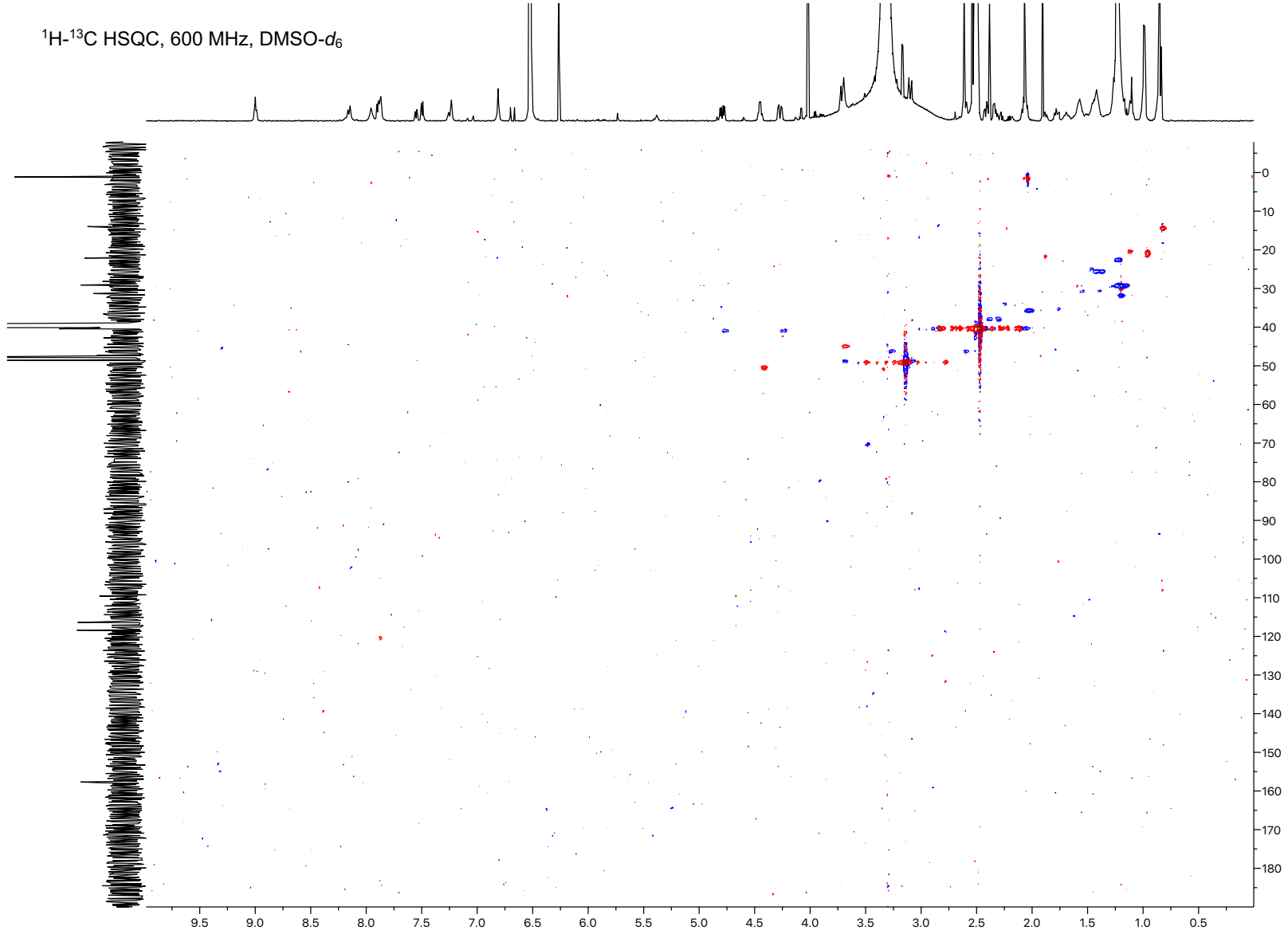

1H NMR, 600 MHz, DMSO-d6
H2O
DMSO-d6
9

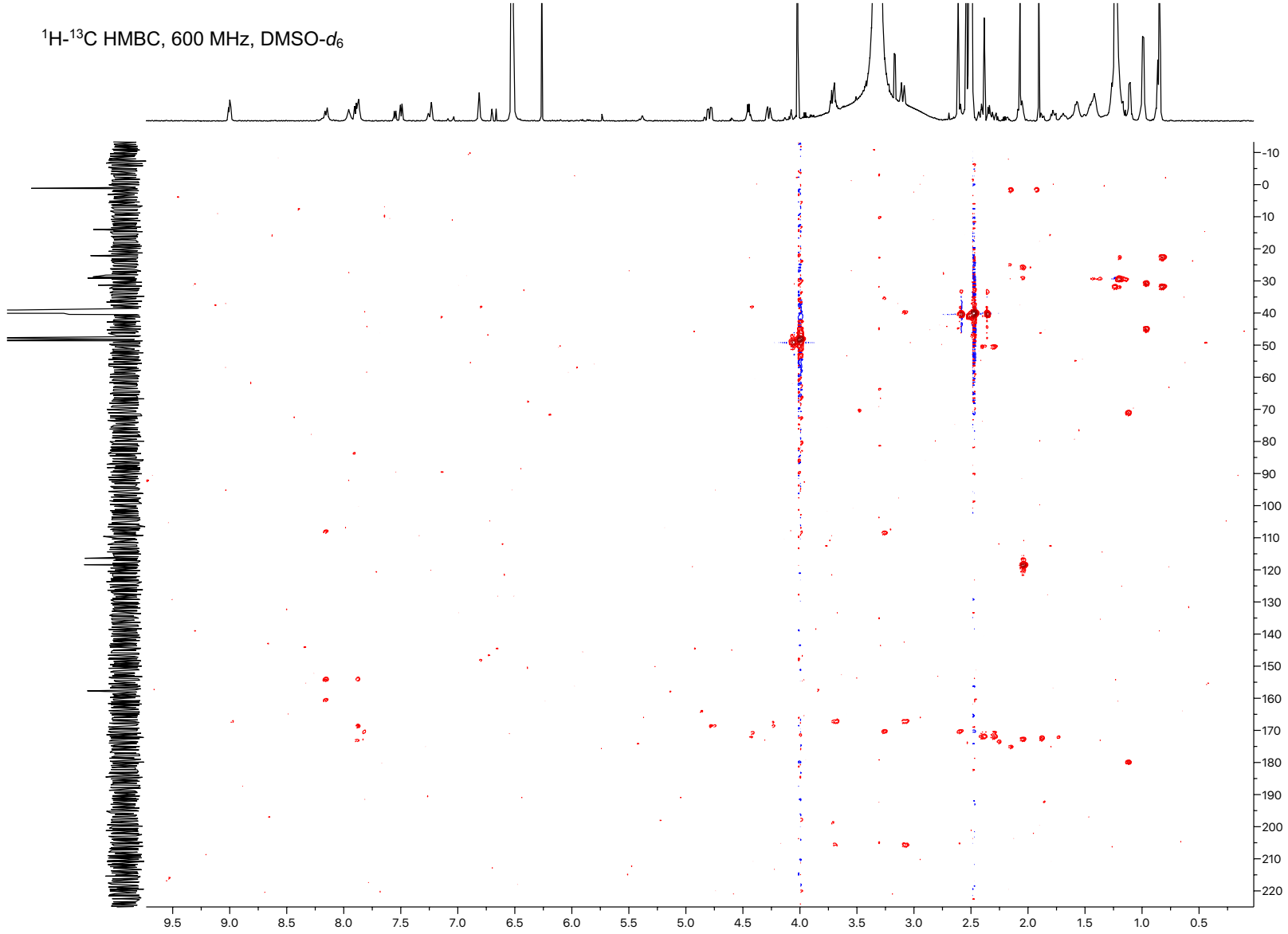

204.8
204.1
172.2
171.4
170.5
168.1
166.8
161.8
146.0
131.2
118.4
70.0
55.9
50.2
50.0
46.4
43.6
40.6
40.1
39.1
37.4
35.2
29.7
29.1
29.1
29.0
29.0
28.9
28.7
28.7
25.2
22.1
20.6
19.4
14.0
^{13}C NMR, 151 MHz, DMSO-d_6
DMSO-d_6
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0

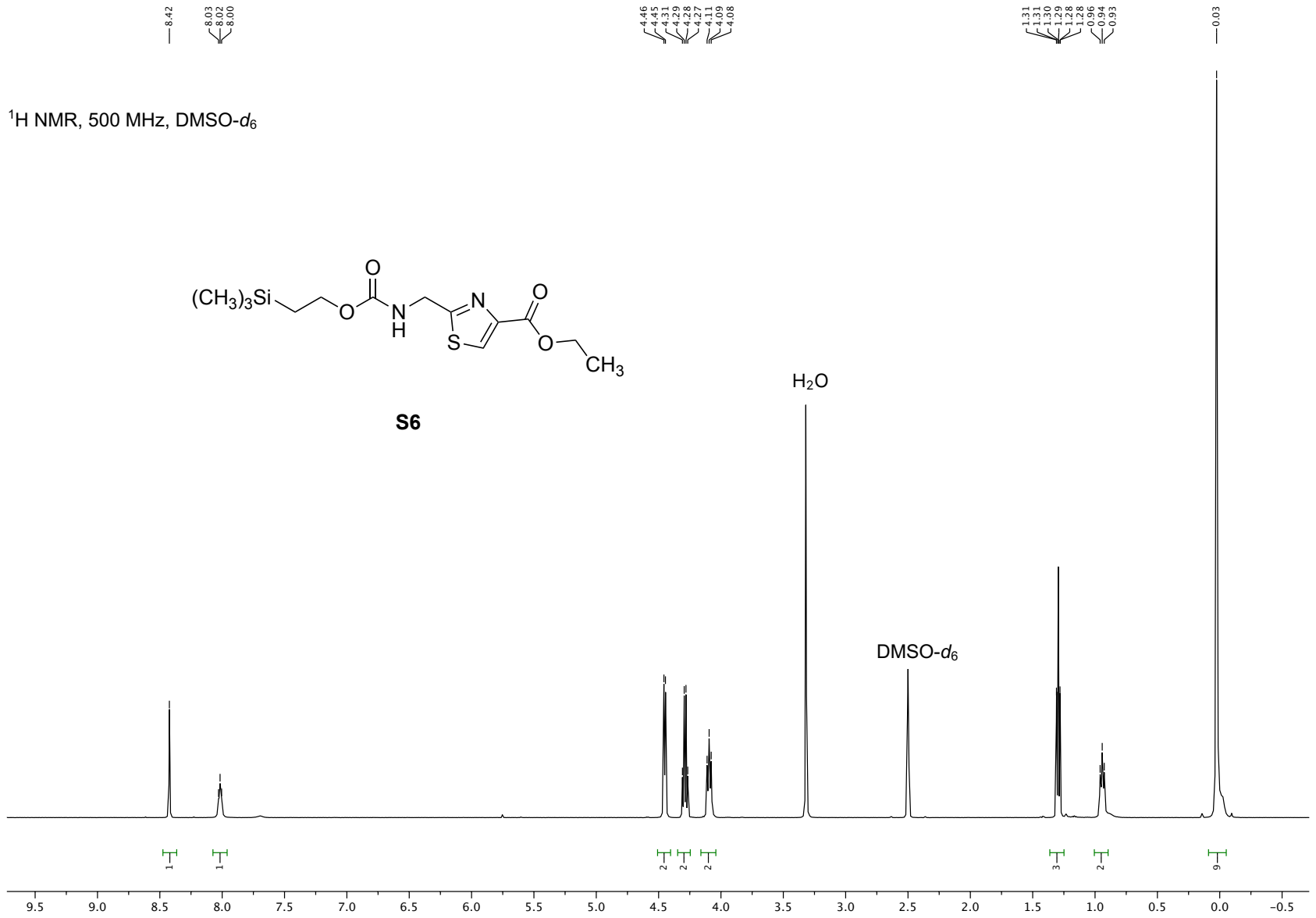

^{1}H NMR, 600 MHz, DMSO-d_6
H_2O
DMSO-d_6
precolibactin-886 (**1**)

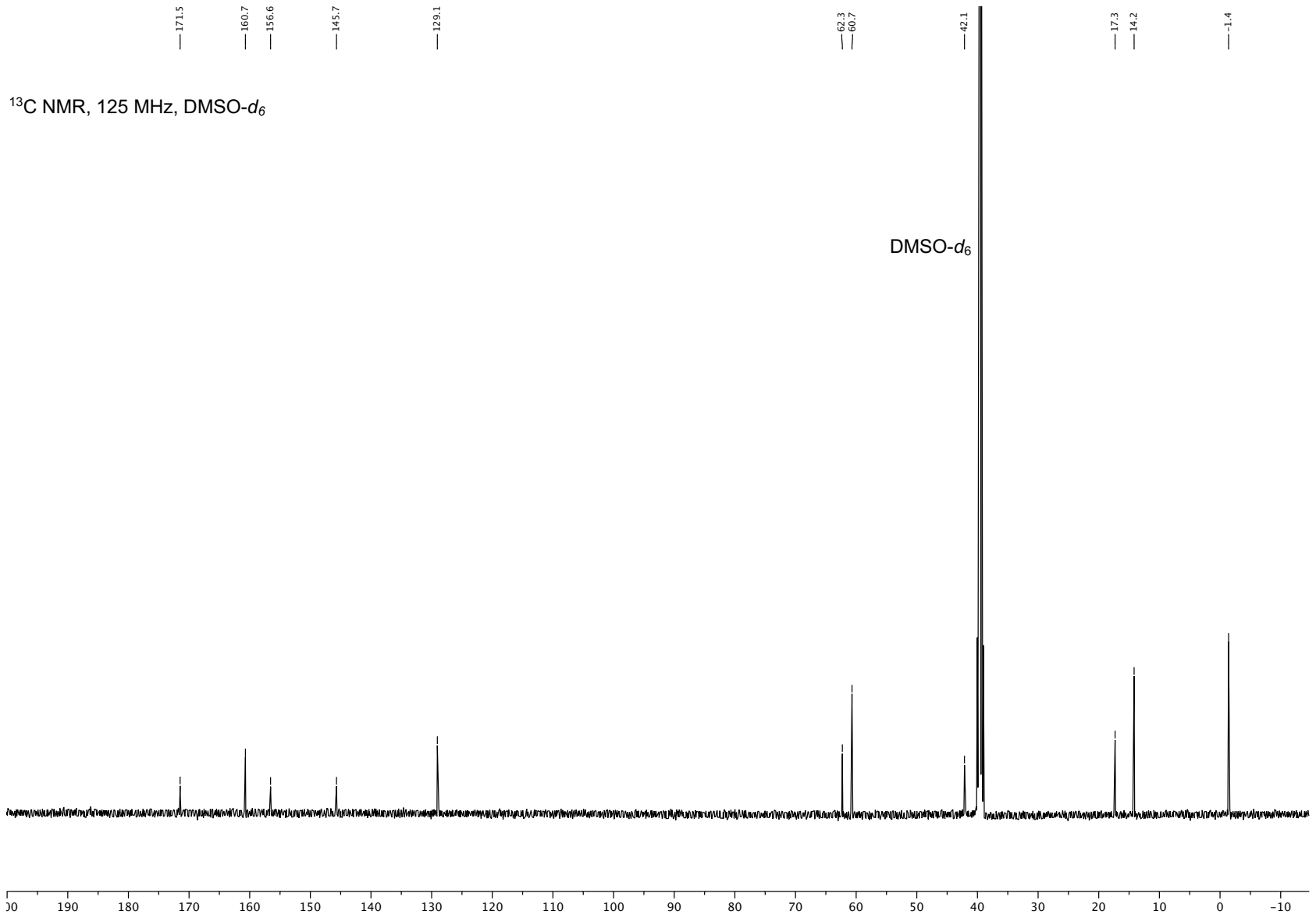

157.7
118.4
116.4
48.6
48.5
48.0
47.9
47.7
47.6
47.4
31.3
29.1
28.7
22.1
14.0
^{13}C NMR, 151 MHz, DMSO-d_6
DMSO-d_6
230 220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10

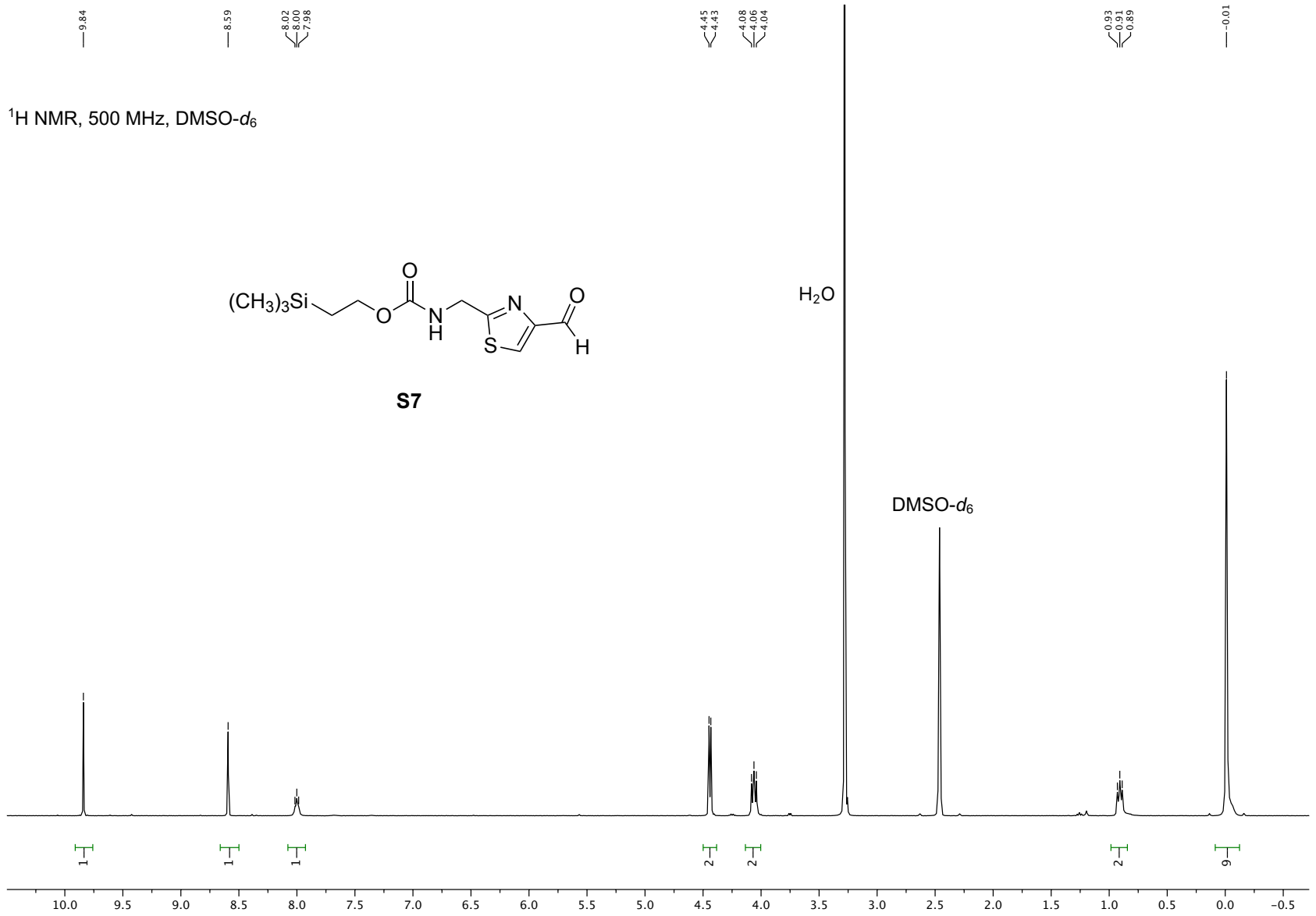

^{1}H-^{1}H COSY, 600 MHz, DMSO-d_6
0
1
2
3
4
5
6
7
8
9
10
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0

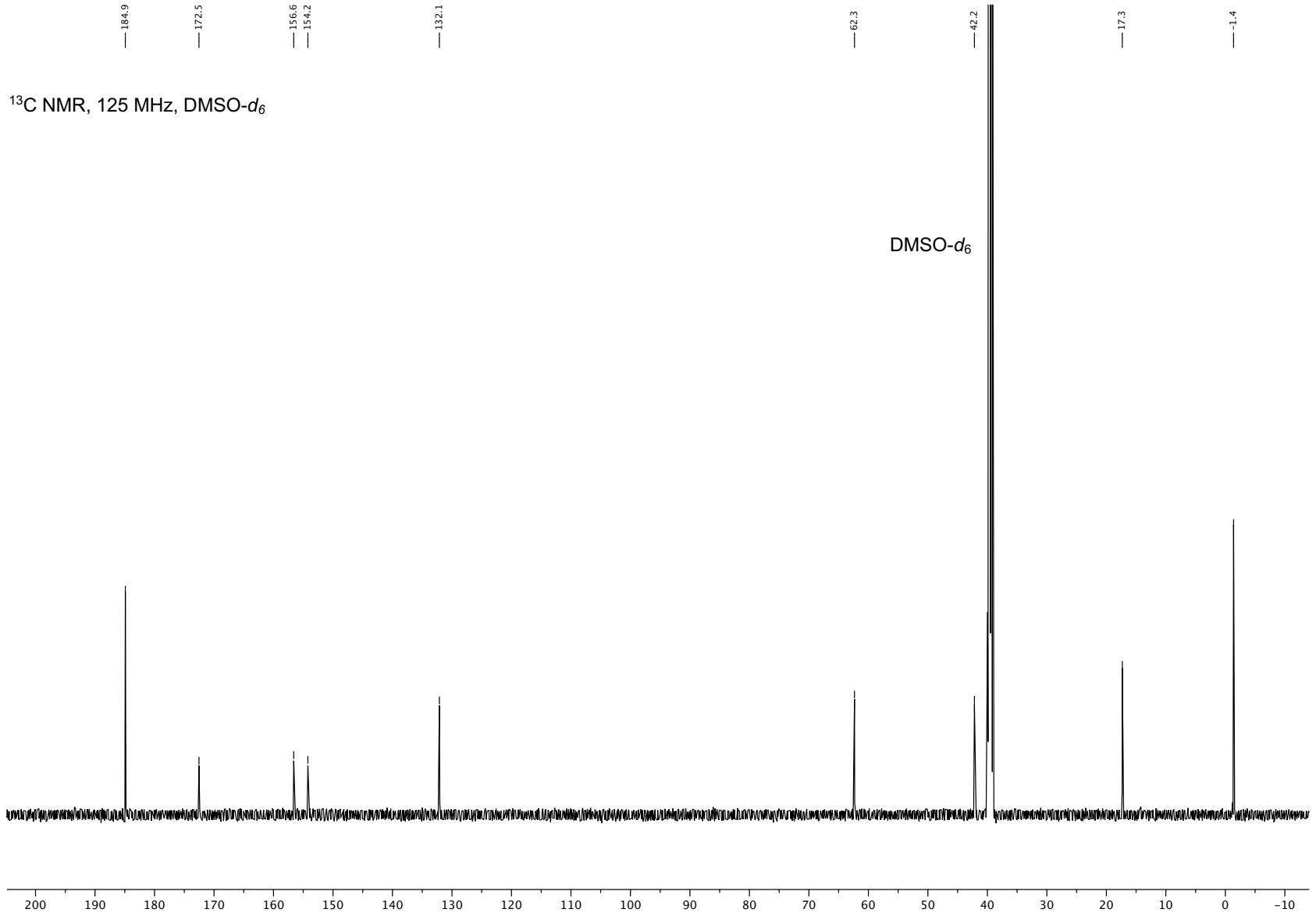

^{1}H-^{13}C HSQC, 600 MHz, DMSO-d_6
0
10
20
30
40
50
60
70
80
90
100
110
120
130
140
150
160
170
180
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5


^{1}H-^{13}C HMBC, 600 MHz, DMSO-d_6
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
-10
0
10
20
30
40
50
60
70
80
90
100
110
120
130
140
150
160
170
180
190
200
210
220

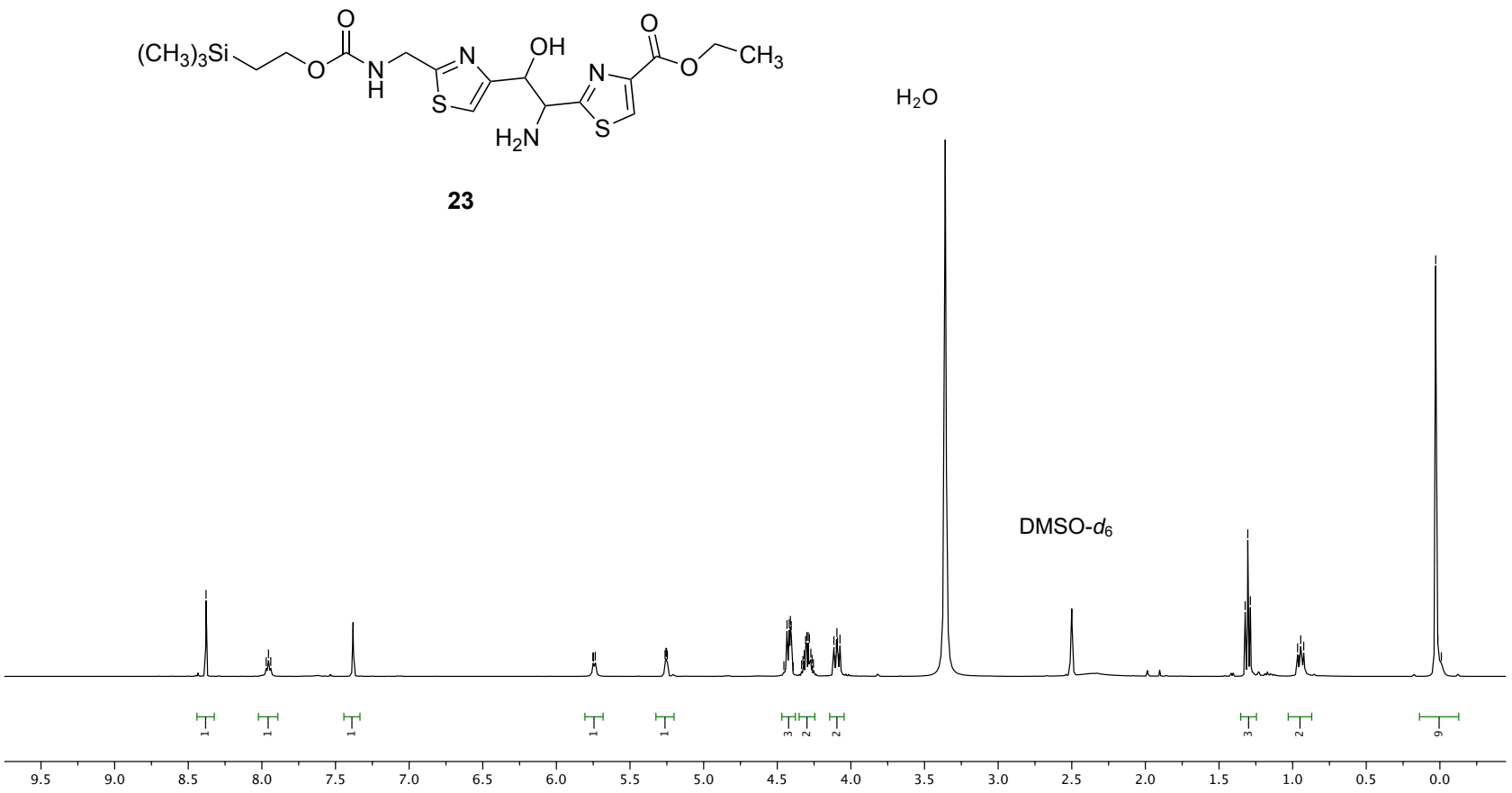

8.42
8.03
8.02
8.00
4.46
4.45
4.31
4.29
4.28
4.27
4.11
4.09
4.08
1.31
1.31
1.30
1.29
1.28
1.28
0.96
0.94
0.93
0.03
^{1}H NMR, 500 MHz, DMSO-d_6
$(CH_3)_3Si$
O
O
N
H
N
S
O
O
CH_3
S6
H_2O
DMSO-d_6
1
1
2
2
2
3
2
9
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5

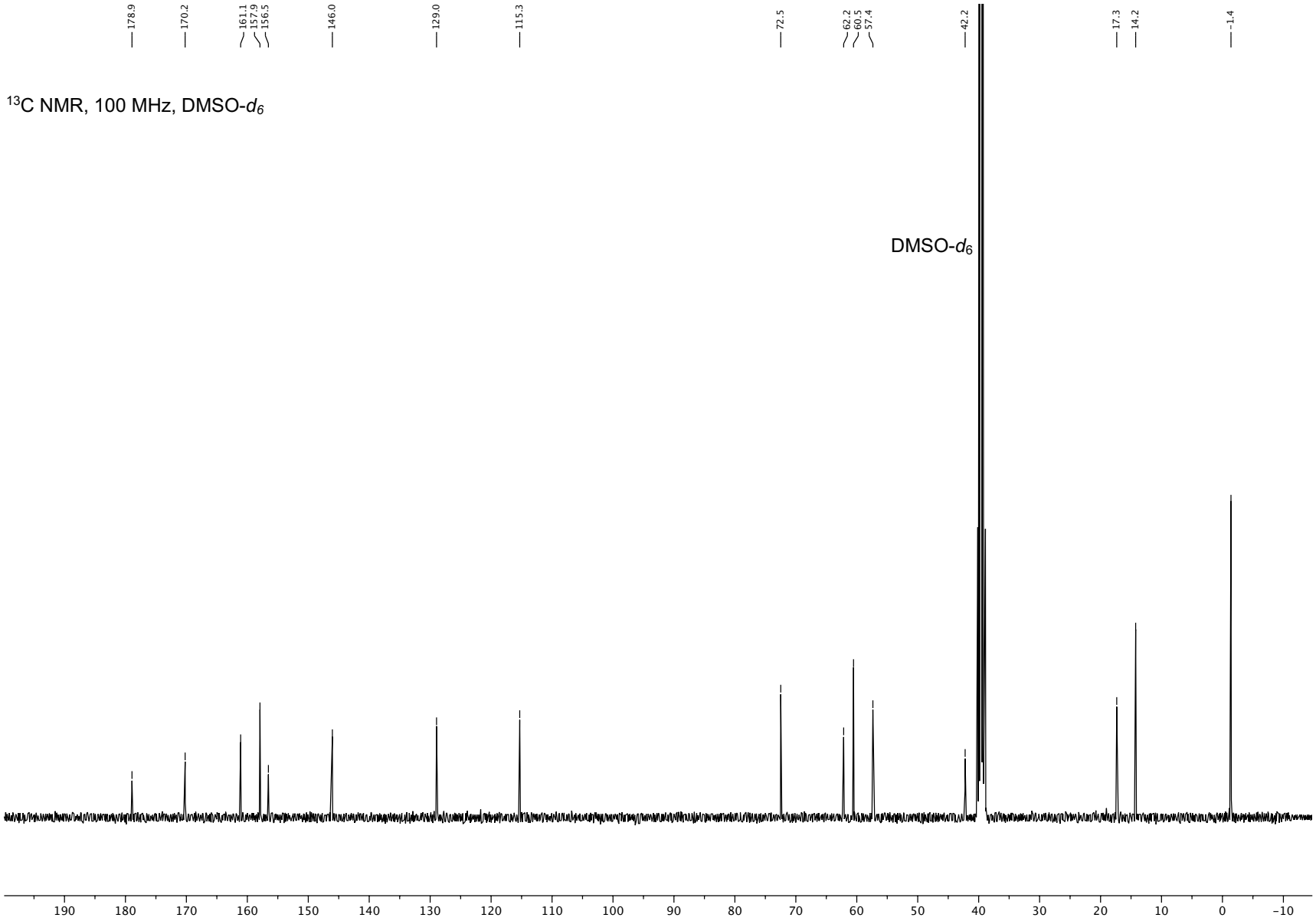

171.5
160.7
156.6
145.7
129.1
62.3
60.7
42.1
17.3
14.2
-1.4
^{13}C NMR, 125 MHz, DMSO-d_6
DMSO-d_6
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10


9.84
8.59
8.02
8.00
7.98
4.45
4.43
4.08
4.06
4.04
0.93
0.91
0.89
-0.01
^{1}H NMR, 500 MHz, DMSO-d_6
$(CH_3)_3Si$
O
O
N
H
N
S
O
H
S7
H_2O
DMSO-d_6
1
1
1
2
2
2
9
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5

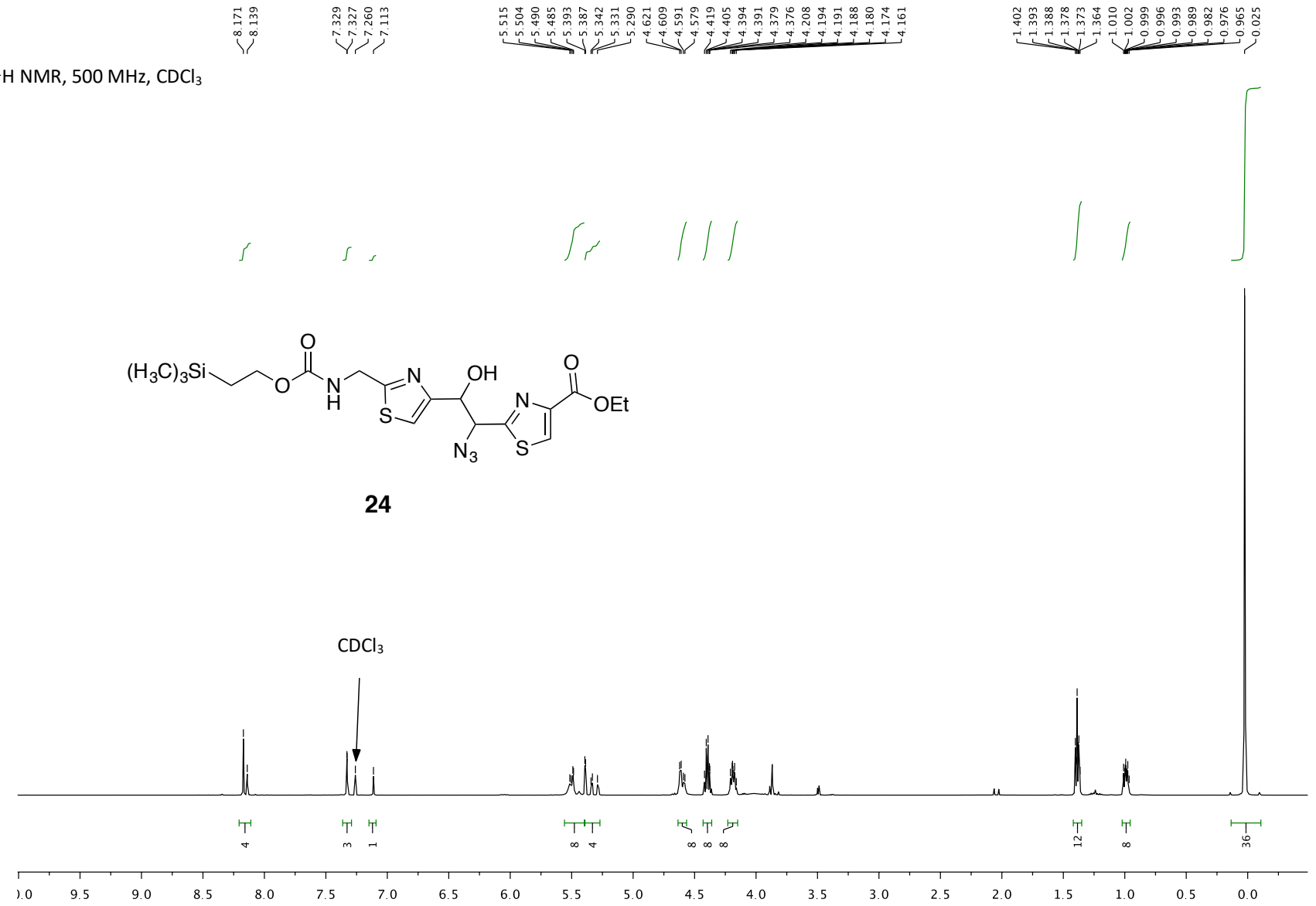

184.9
172.5
156.6
154.2
132.1
62.3
42.2
17.3
-1.4
^{13}C NMR, 125 MHz, DMSO-d_6
DMSO-d_6
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10

^{1}H NMR, 400 MHz, DMSO-d_6




178.9
170.2
161.1
157.9
156.5
146.0
129.0
115.3
72.5
62.2
60.5
57.4
42.2
17.3
14.2
-1.4
^{13}C NMR, 100 MHz, DMSO-d_6
DMSO-d_6
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10

^{1}H NMR, 500 MHz, $CDCl_3$



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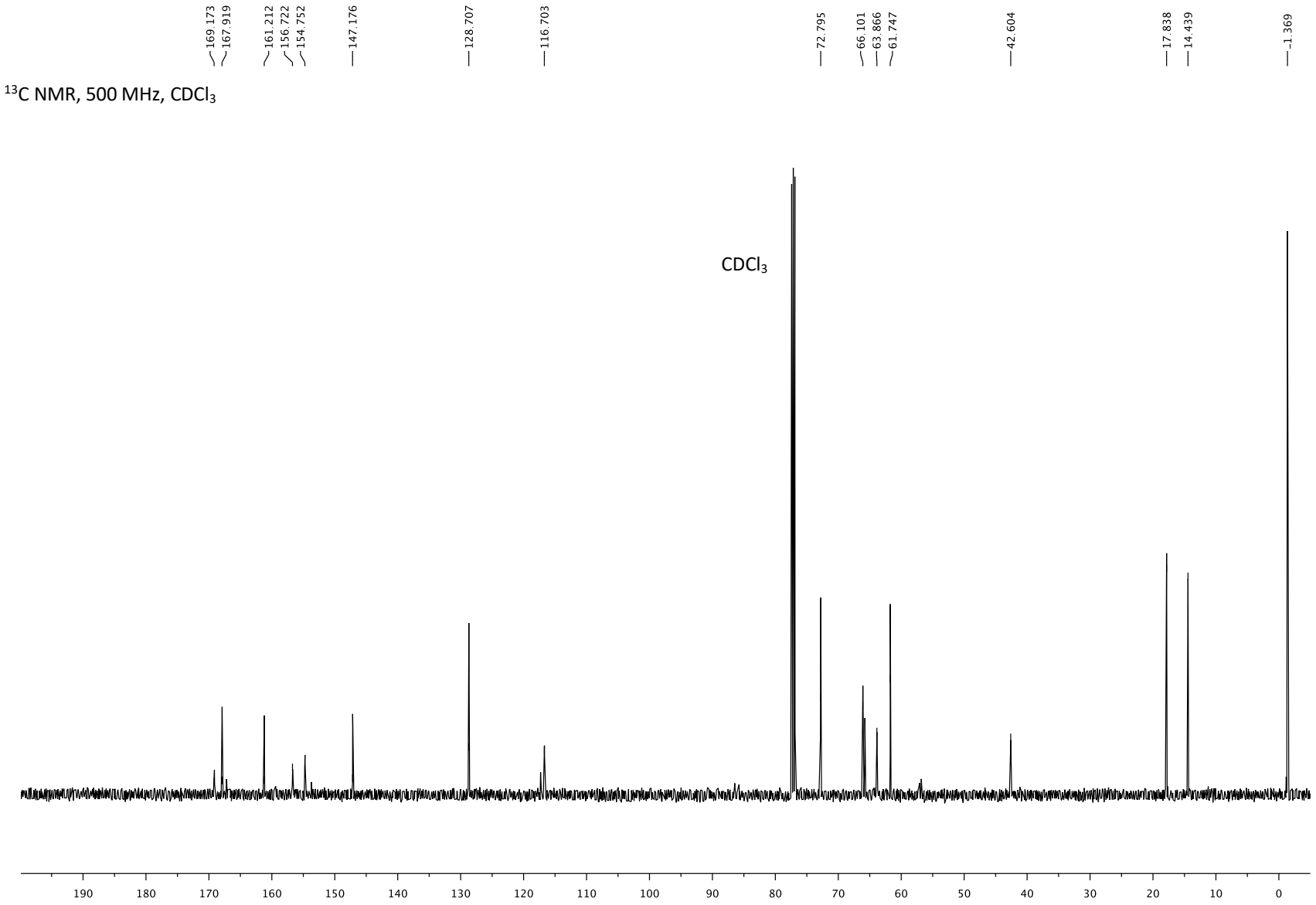
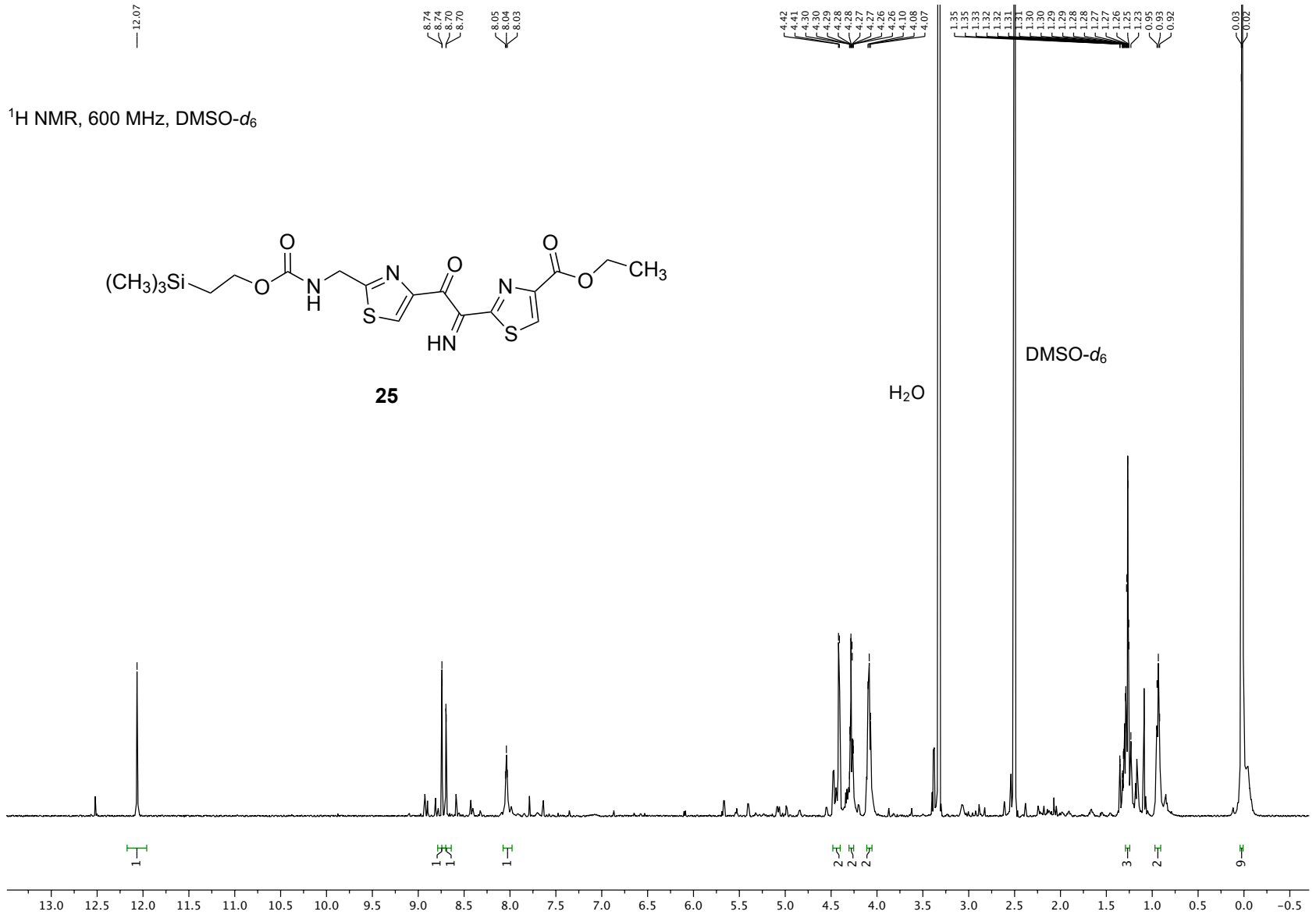
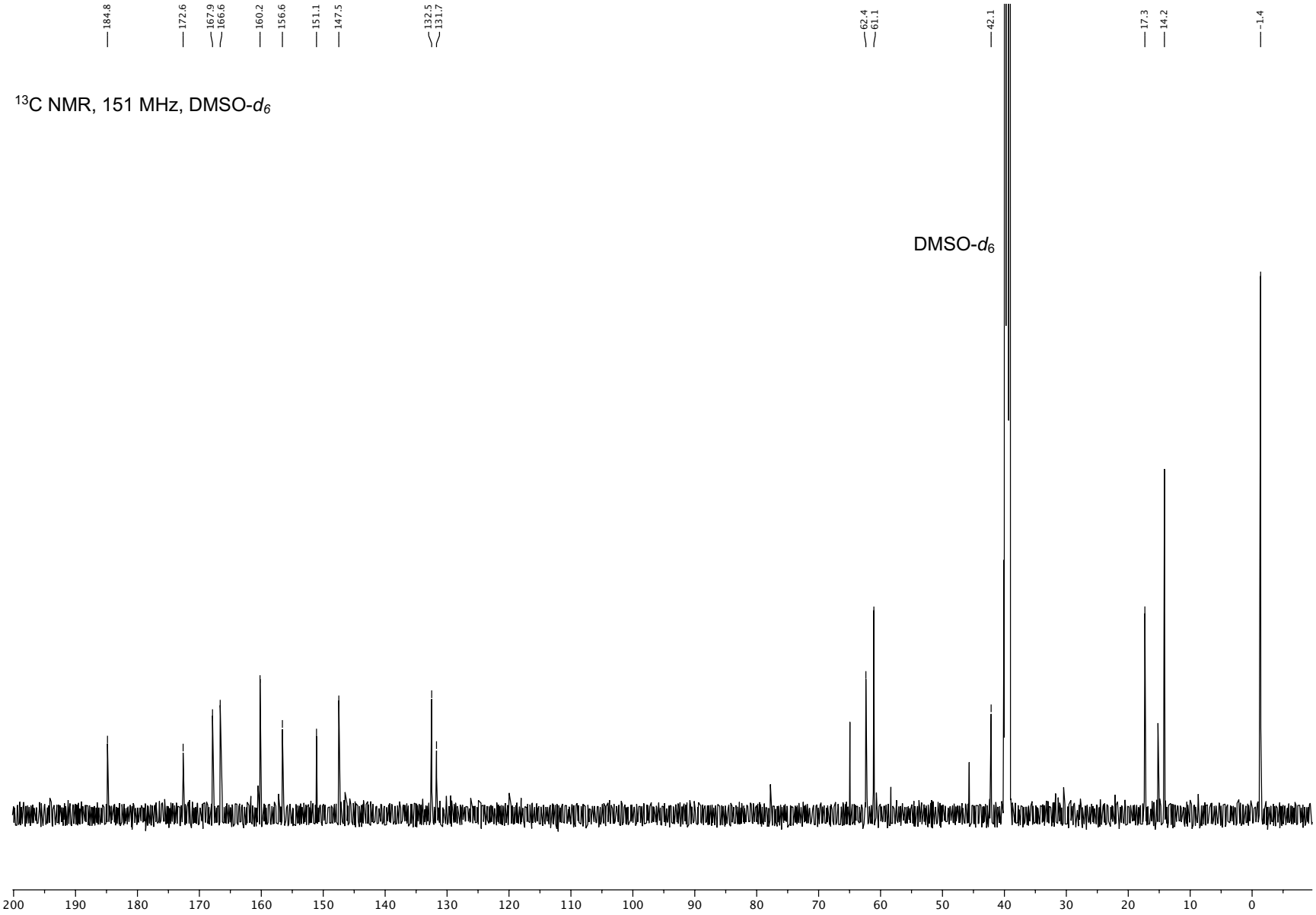
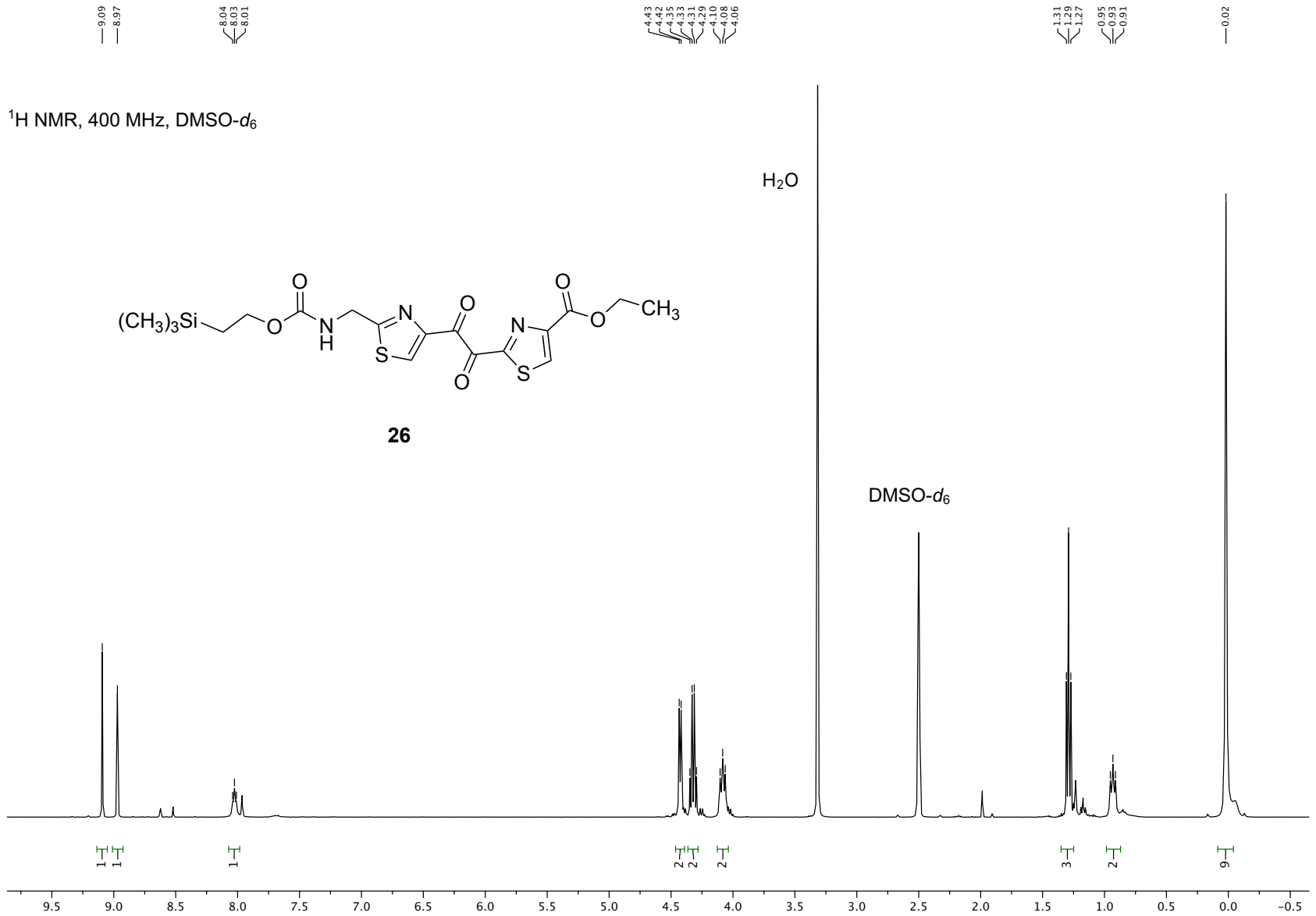
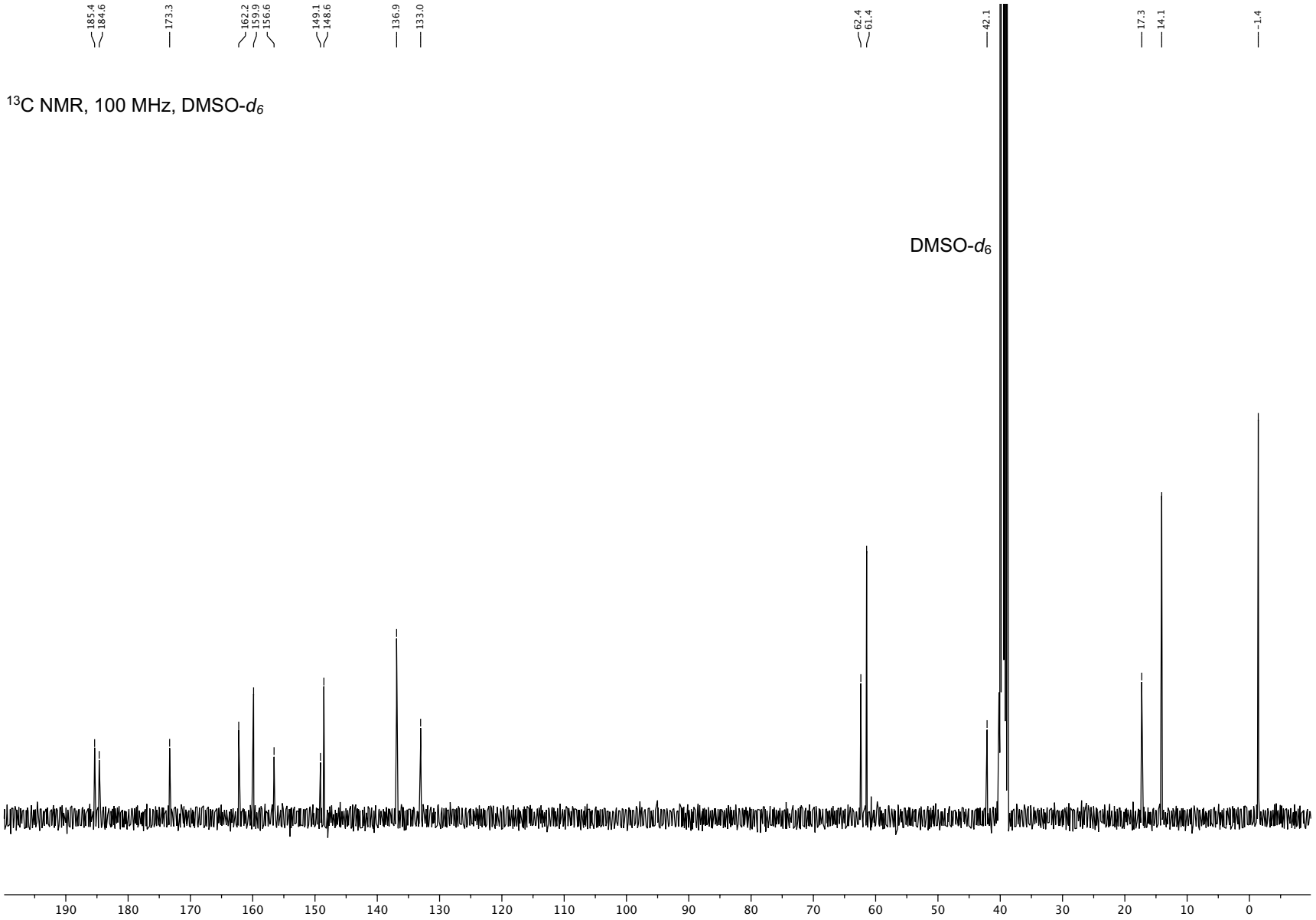

169.173
167.919
161.212
156.722
154.752
147.176
128.707
116.703
72.795
66.101
63.866
61.747
42.604
17.838
14.439
1.369
^{13}C NMR, 500 MHz, $CDCl_3$
$CDCl_3$
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0

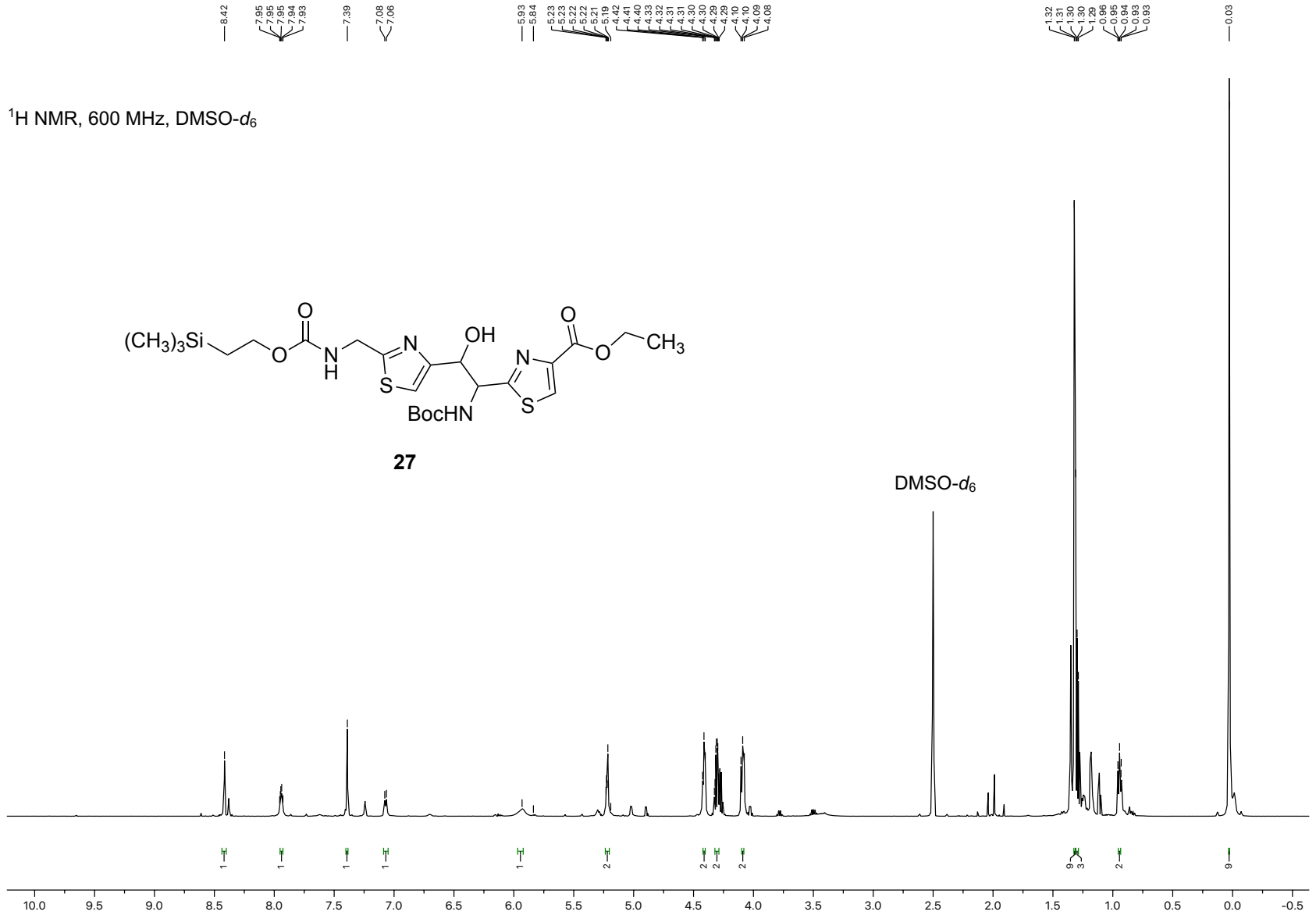

12.07
8.74
8.74
8.70
8.70
8.05
8.04
8.03
4.42
4.41
4.30
4.30
4.29
4.28
4.28
4.27
4.27
4.26
4.26
4.10
4.08
4.07
1.35
1.35
1.33
1.32
1.32
1.31
1.31
1.30
1.30
1.29
1.29
1.28
1.28
1.27
1.27
1.26
1.25
1.23
0.95
0.93
0.92
0.03
0.02
[1]H NMR, 600 MHz, DMSO-d6
(CH3)3Si
O
O
N
H
N
S
O
HN
N
S
O
O
CH3
25
H2O
DMSO-d6
1
1
1
2
2
2
3
2
9
13.0
12.5
12.0
11.5
11.0
10.5
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5

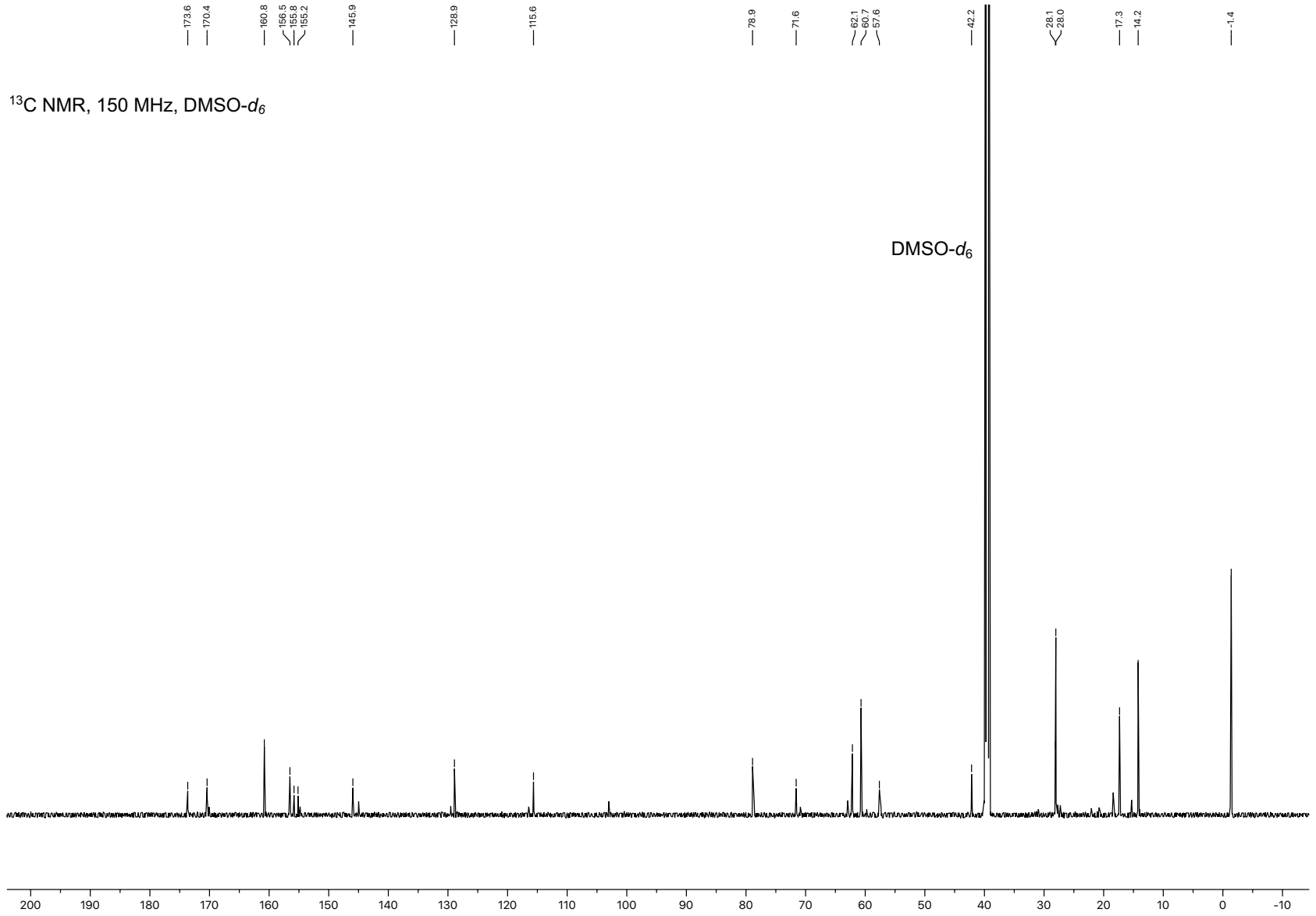

184.8
172.6
167.9
166.6
160.2
156.6
151.1
147.5
132.5
131.7
62.4
61.1
42.1
17.3
14.2
-1.4
^{13}C NMR, 151 MHz, DMSO-d_6
DMSO-d_6
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0

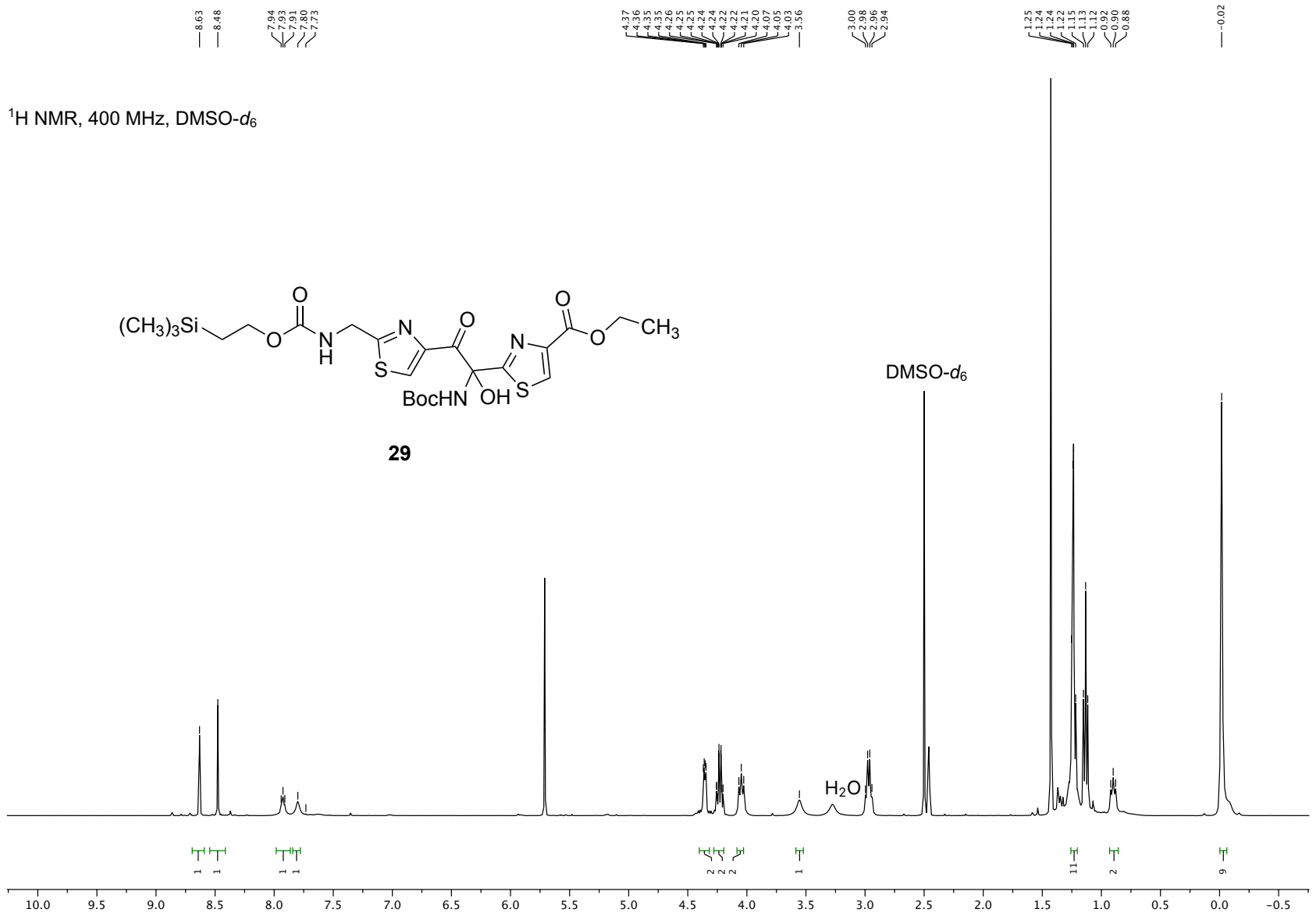

9.09
8.97
8.04
8.03
8.01
4.43
4.42
4.35
4.33
4.31
4.29
4.10
4.08
4.06
1.31
1.29
1.27
0.95
0.93
0.91
0.02
^{1}H NMR, 400 MHz, DMSO-d_6
H_2O
DMSO-d_6
$(CH_3)_3Si$
O
N
H
S
N
S
O
O
O
O
CH_3
1
1
1
2
2
2
3
2
9
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5

26

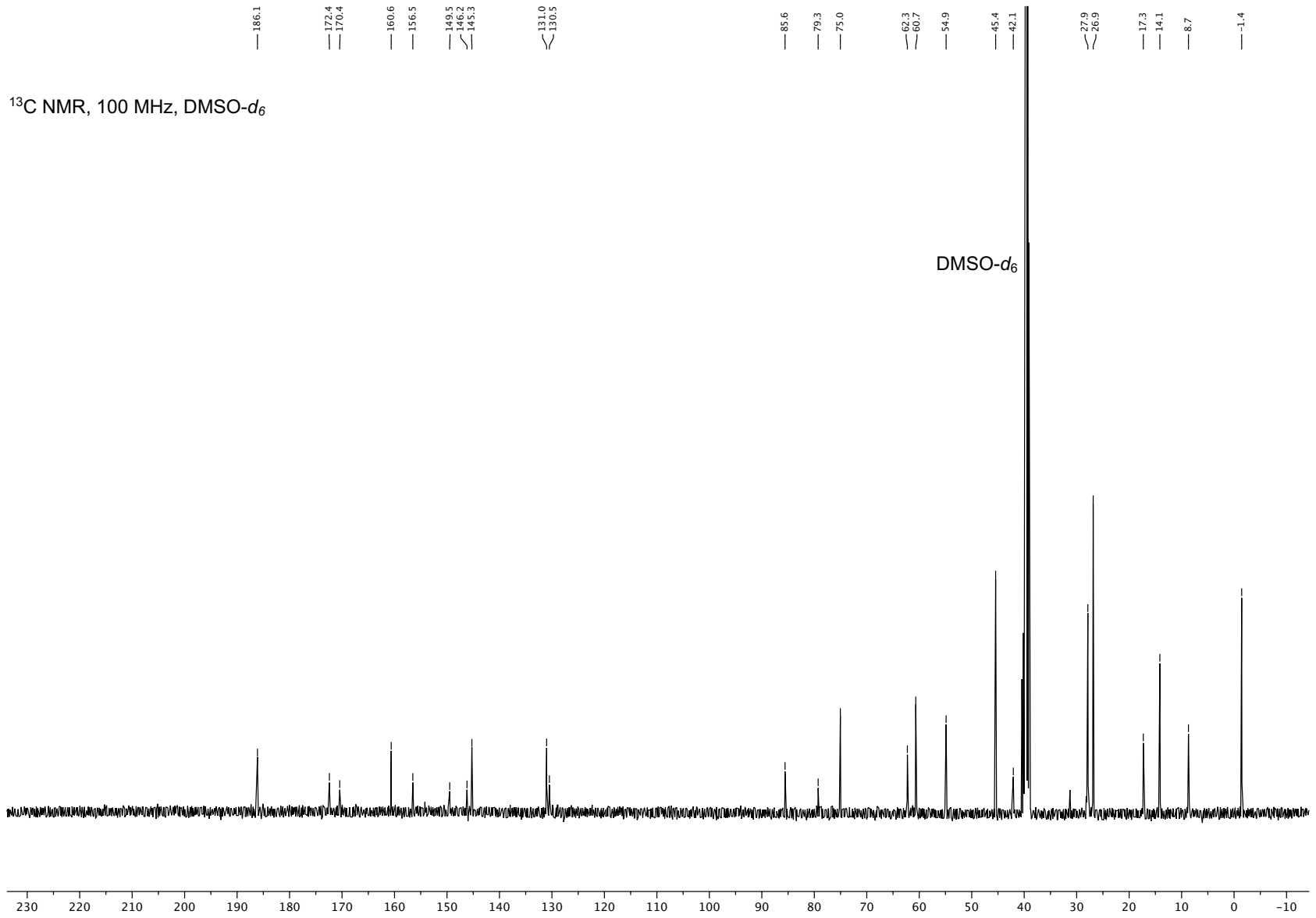

185.4
184.6
173.3
162.2
159.9
156.6
149.1
148.6
136.9
133.0
62.4
61.4
42.1
17.3
14.1
-1.4
^{13}C NMR, 100 MHz, DMSO-d_6
DMSO-d_6
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0

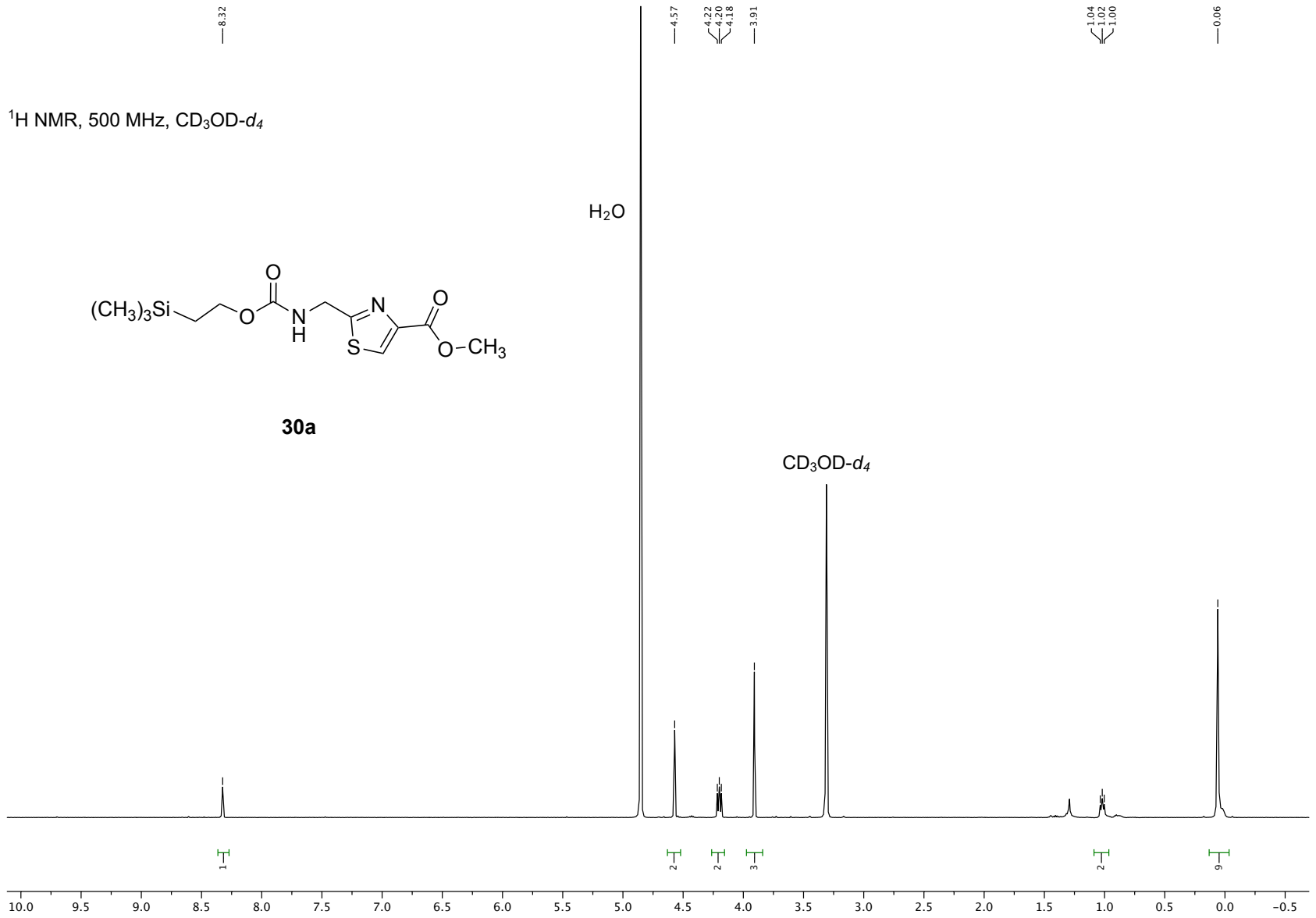

8.42
7.95
7.95
7.95
7.94
7.93
7.39
7.08
7.06
5.93
5.84
5.23
5.23
5.22
5.22
5.21
5.19
4.42
4.41
4.40
4.33
4.32
4.31
4.31
4.30
4.30
4.29
4.29
4.10
4.10
4.09
4.08
1.32
1.31
1.30
1.30
1.29
0.96
0.95
0.94
0.93
0.93
0.03
1H NMR, 600 MHz, DMSO-d6
(CH3)3Si
O
O
N
H
N
S
OH
BocHN
N
S
O
O
CH3
27
DMSO-d6
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5

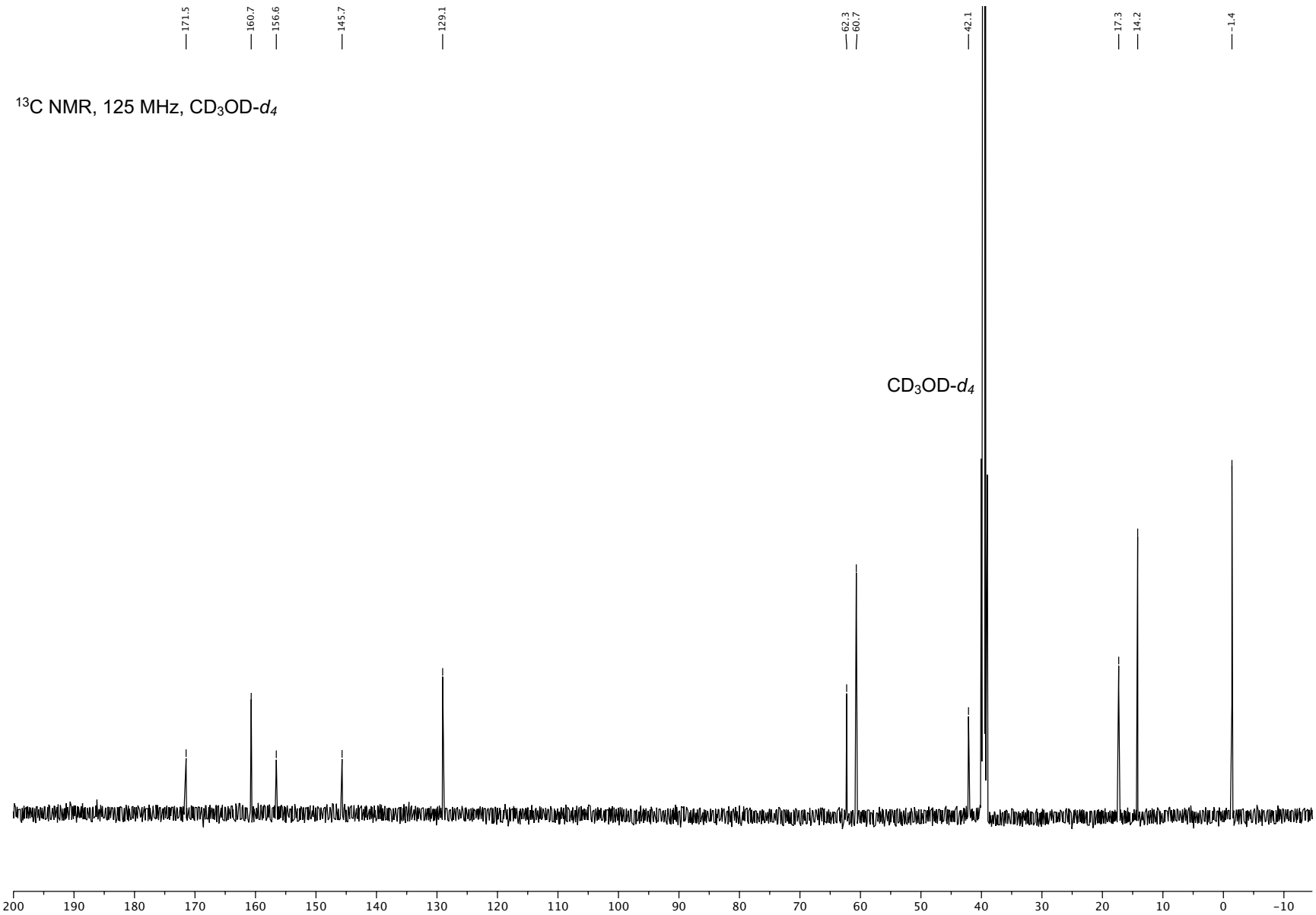

173.6
170.4
160.8
156.5
155.8
155.2
145.9
128.9
115.6
78.9
71.6
62.1
60.7
57.6
42.2
28.1
28.0
17.3
14.2
-1.4
13C NMR, 150 MHz, DMSO-d6
DMSO-d6
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10

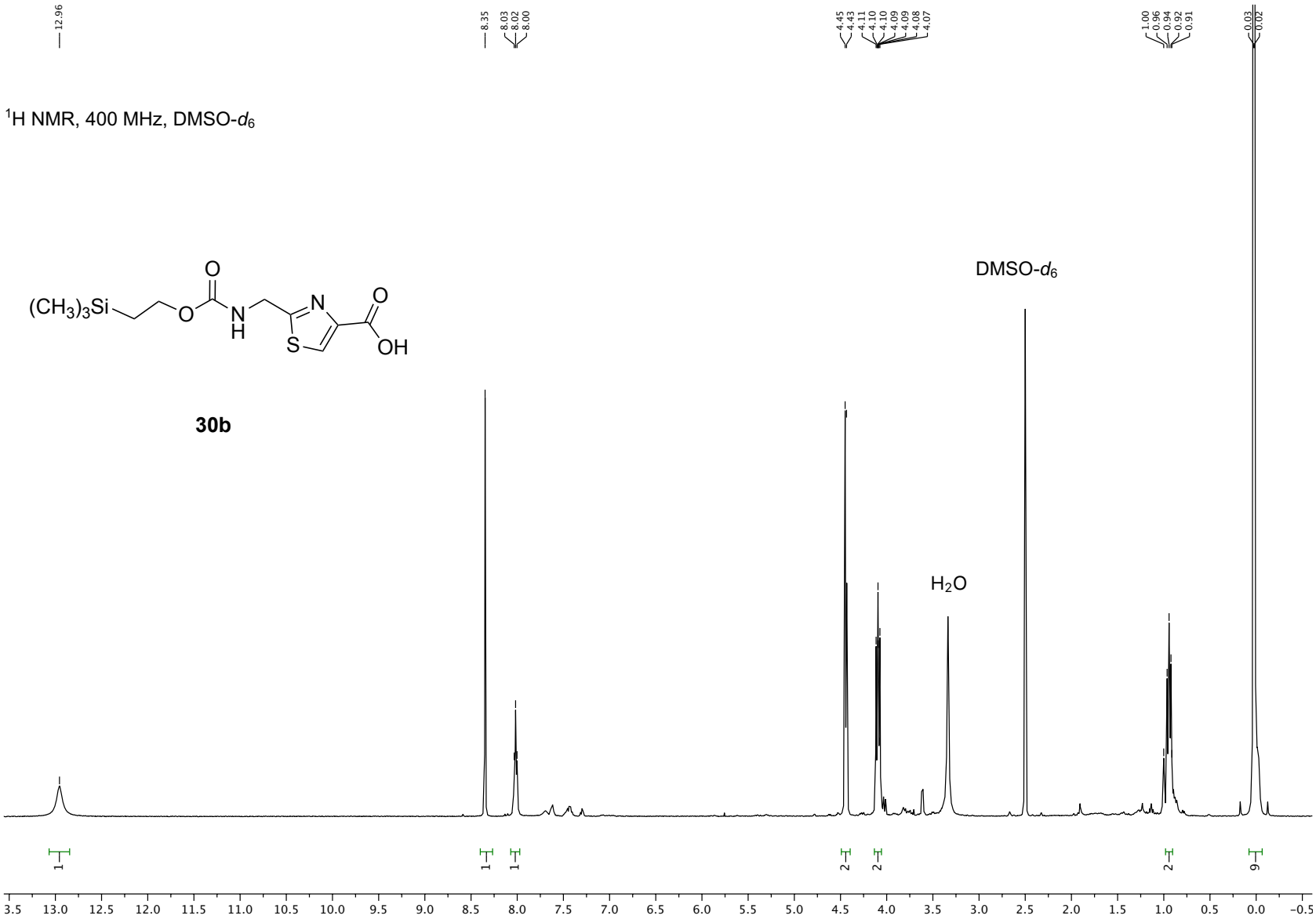

8.63
8.48
7.94
7.93
7.91
7.80
7.73
4.37
4.36
4.35
4.35
4.26
4.25
4.25
4.24
4.24
4.22
4.22
4.21
4.20
4.07
4.05
4.03
3.56
3.00
2.98
2.96
2.94
1.25
1.24
1.24
1.22
1.22
1.15
1.13
1.12
0.92
0.90
0.88
-0.02
^{1}H NMR, 400 MHz, DMSO-d_6
$(CH_3)_3Si$
O
O
N
H
N
S
O
N
S
O
O
CH_3
BocHN
OH
DMSO-d_6
H_2O
1
1
1
1
2
2
2
1
11
2
9
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5

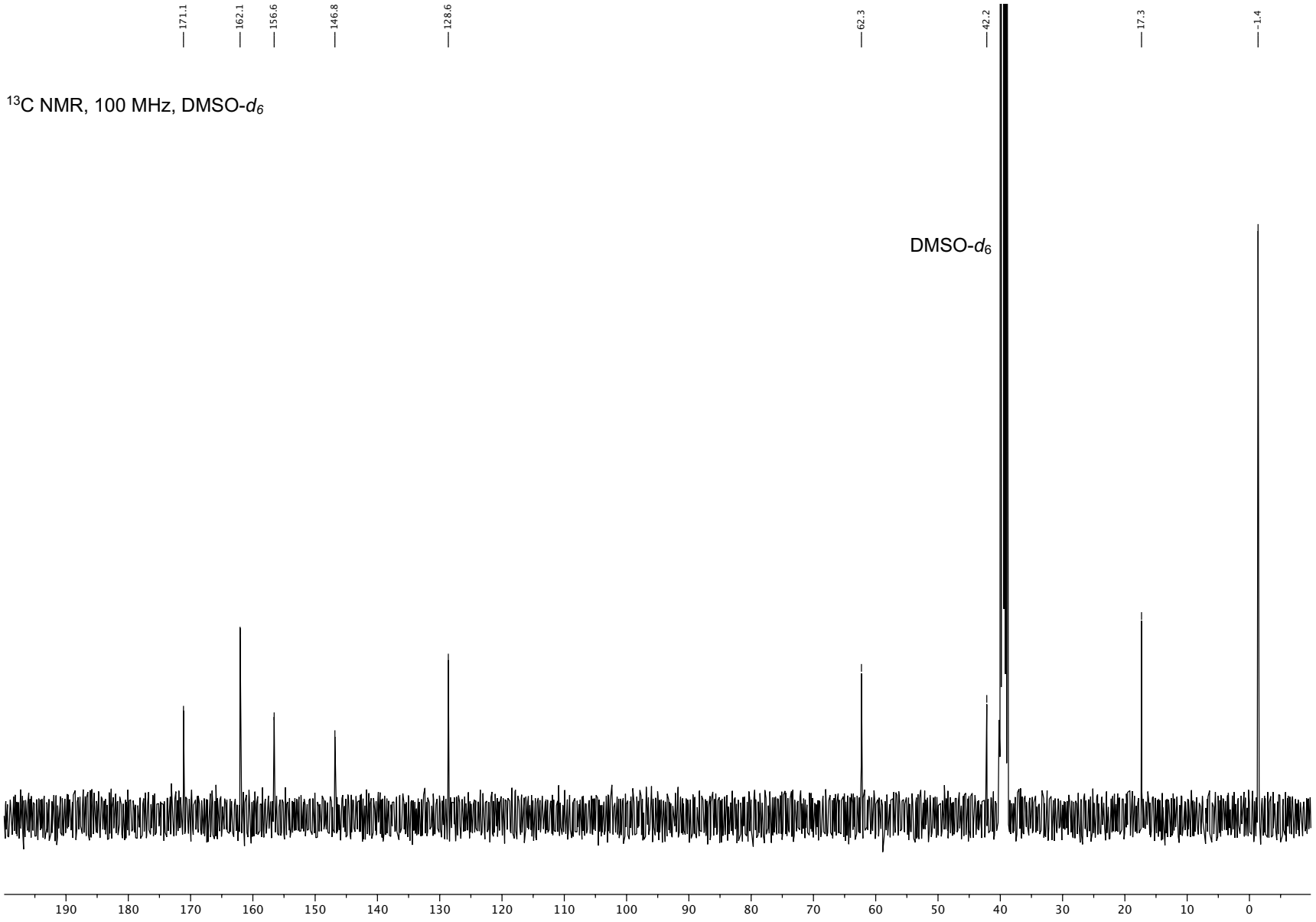
29

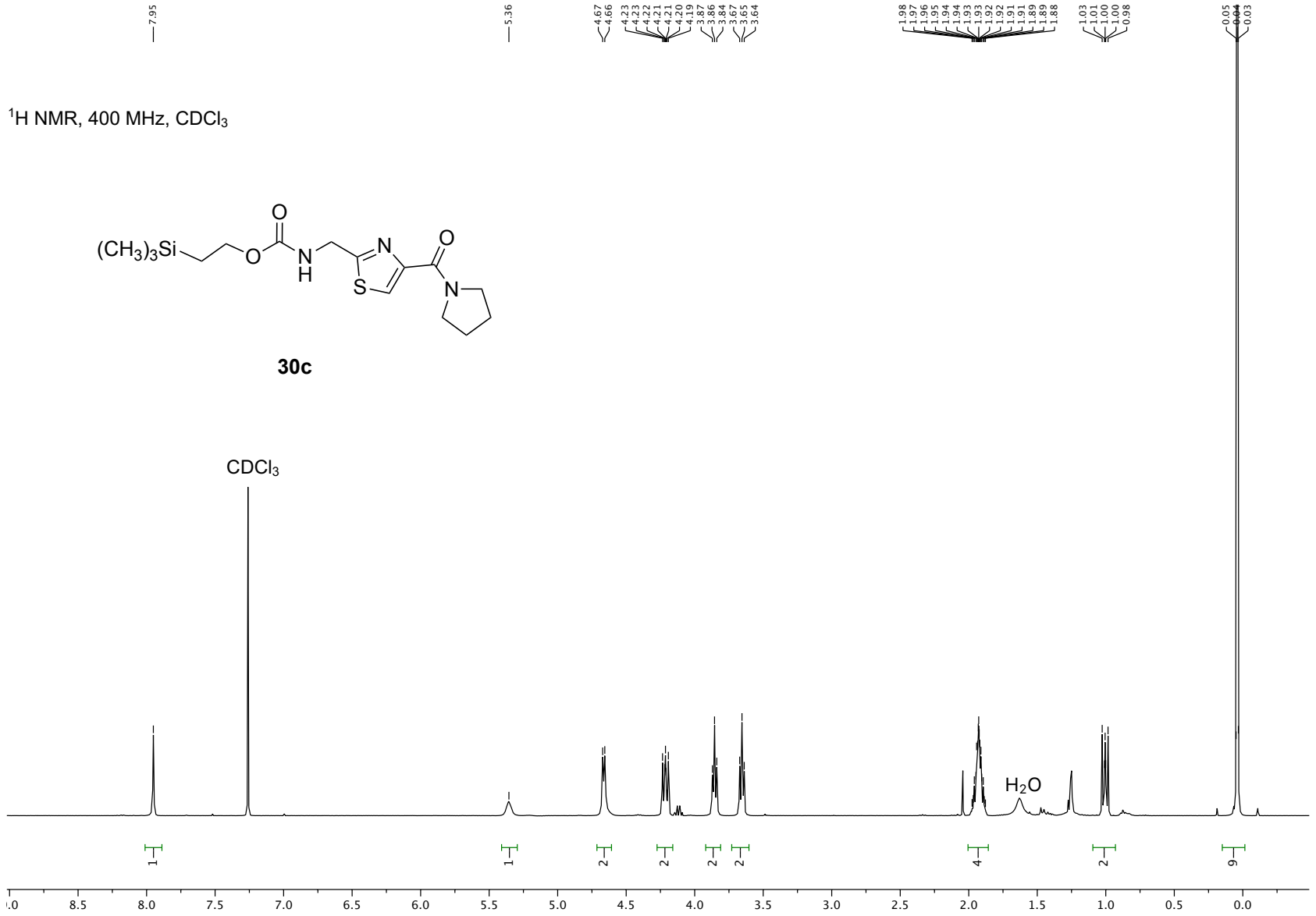

186.1
172.4
170.4
160.6
156.5
149.5
146.2
145.3
131.0
130.5
85.6
79.3
75.0
62.3
60.7
54.9
45.4
42.1
27.9
26.9
17.3
14.1
8.7
-1.4
13C NMR, 100 MHz, DMSO-d6
DMSO-d6
230 220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10

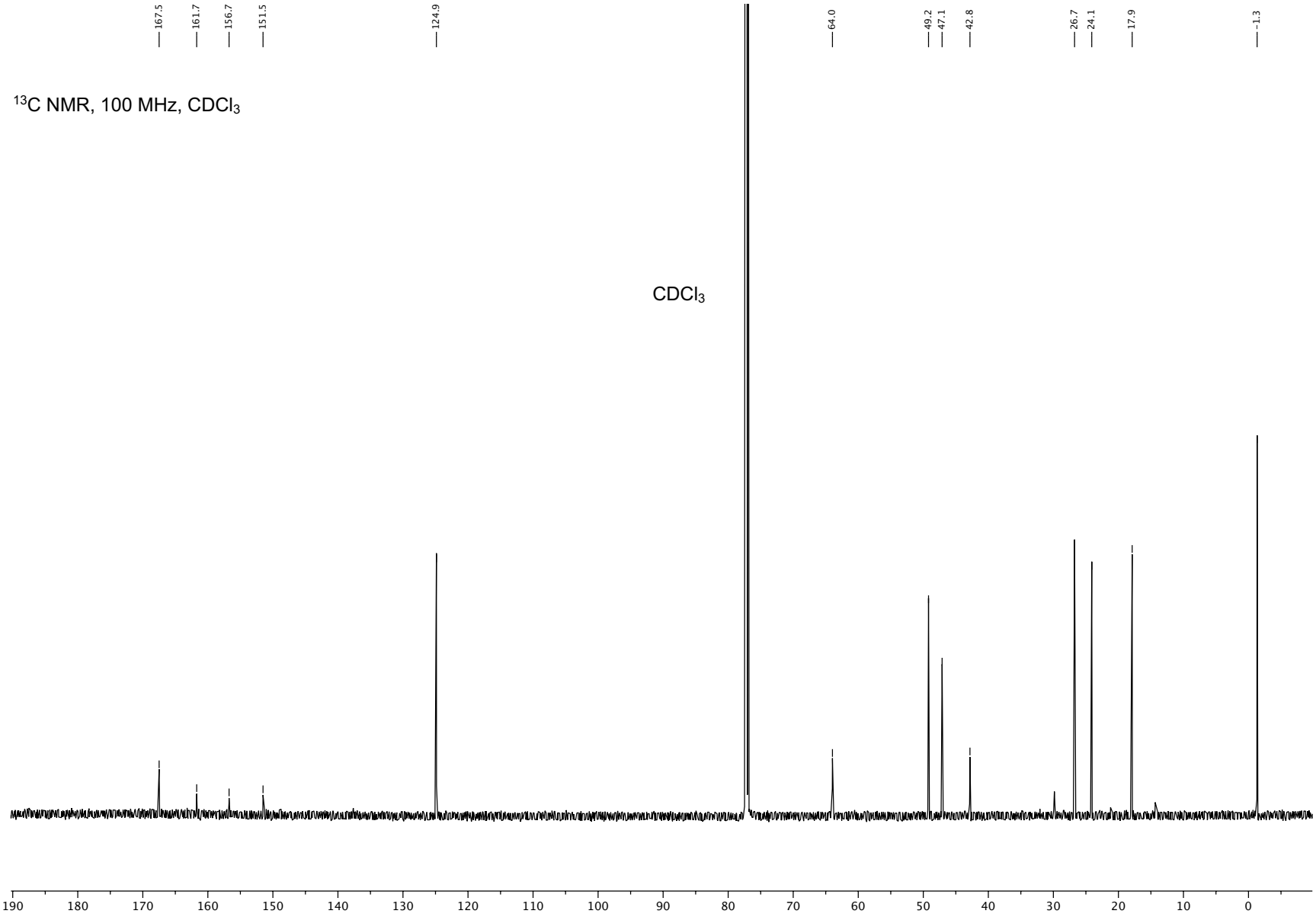

8.32
4.57
4.22
4.20
4.18
3.91
1.04
1.02
1.00
0.06
^{1}H NMR, 500 MHz, CD_3OD-d_4
H_2O
$(CH_3)_3Si$
O
N
H
S
N
O
O
$O-CH_3$
30a
CD_3OD-d_4
1
2
2
3
2
9
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5

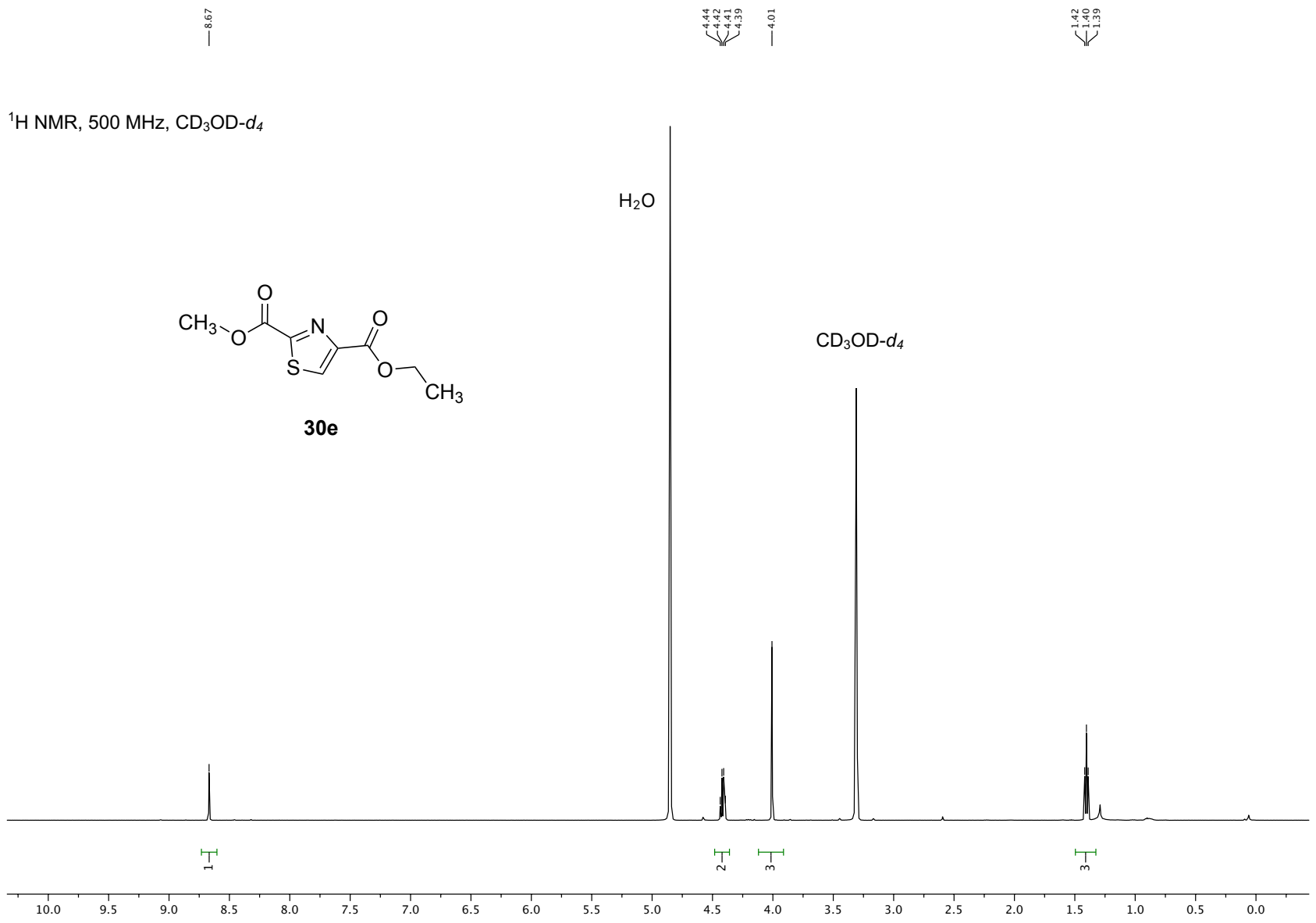

171.5
160.7
156.6
145.7
129.1
62.3
60.7
42.1
17.3
14.2
-1.4
^{13}C NMR, 125 MHz, CD_3OD-d_4
CD_3OD-d_4
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10

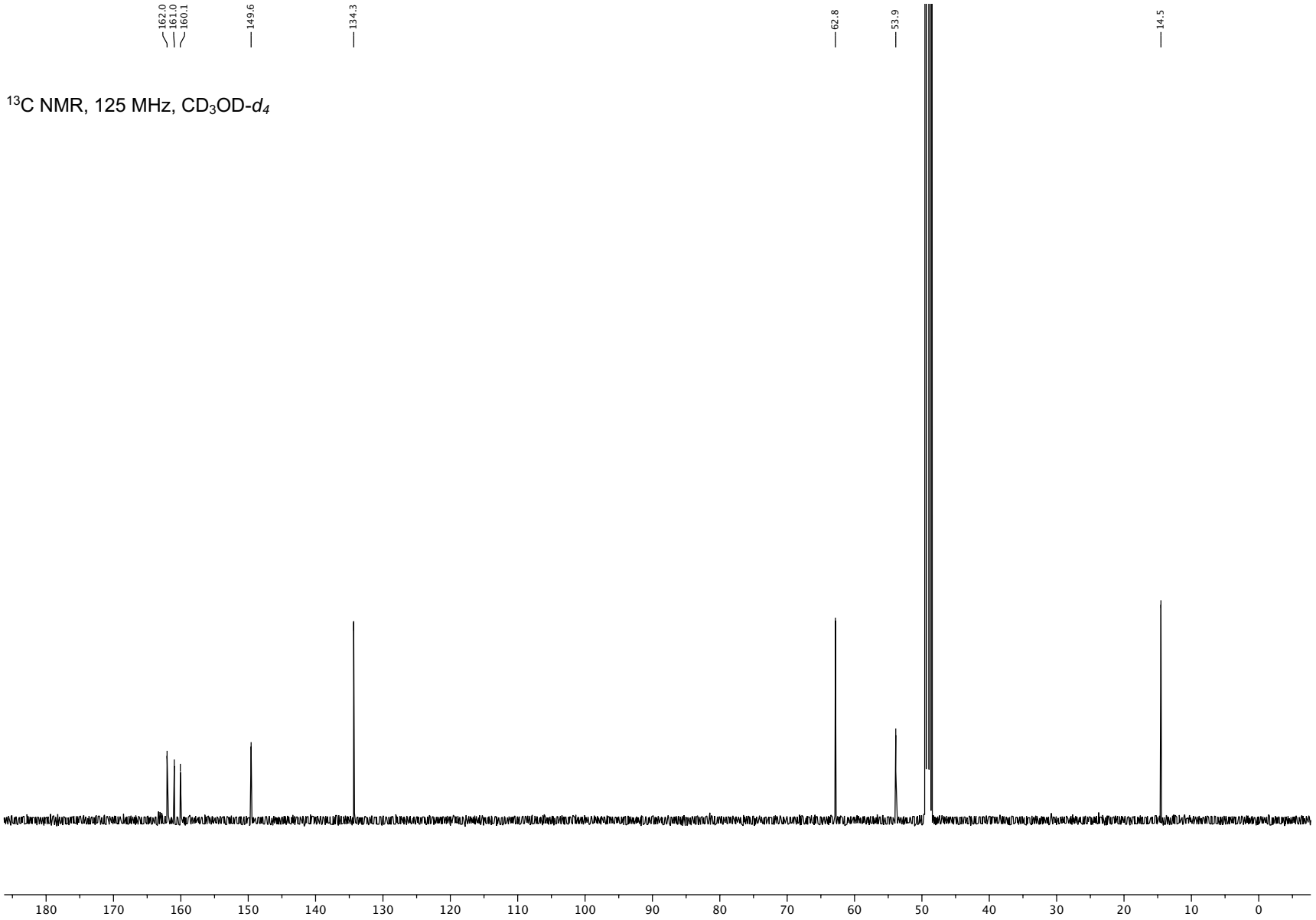

12.96
8.35
8.03
8.02
8.00
4.45
4.43
4.11
4.10
4.09
4.09
4.08
4.07
1.00
0.96
0.94
0.92
0.91
0.03
0.02
^{1}H NMR, 400 MHz, DMSO-d_6
$(CH_3)_3Si$
O
O
N
H
N
S
O
OH
30b
DMSO-d_6
H_2O
1
1
1
2
2
2
9
3.5 13.0 12.5 12.0 11.5 11.0 10.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0 -0.5

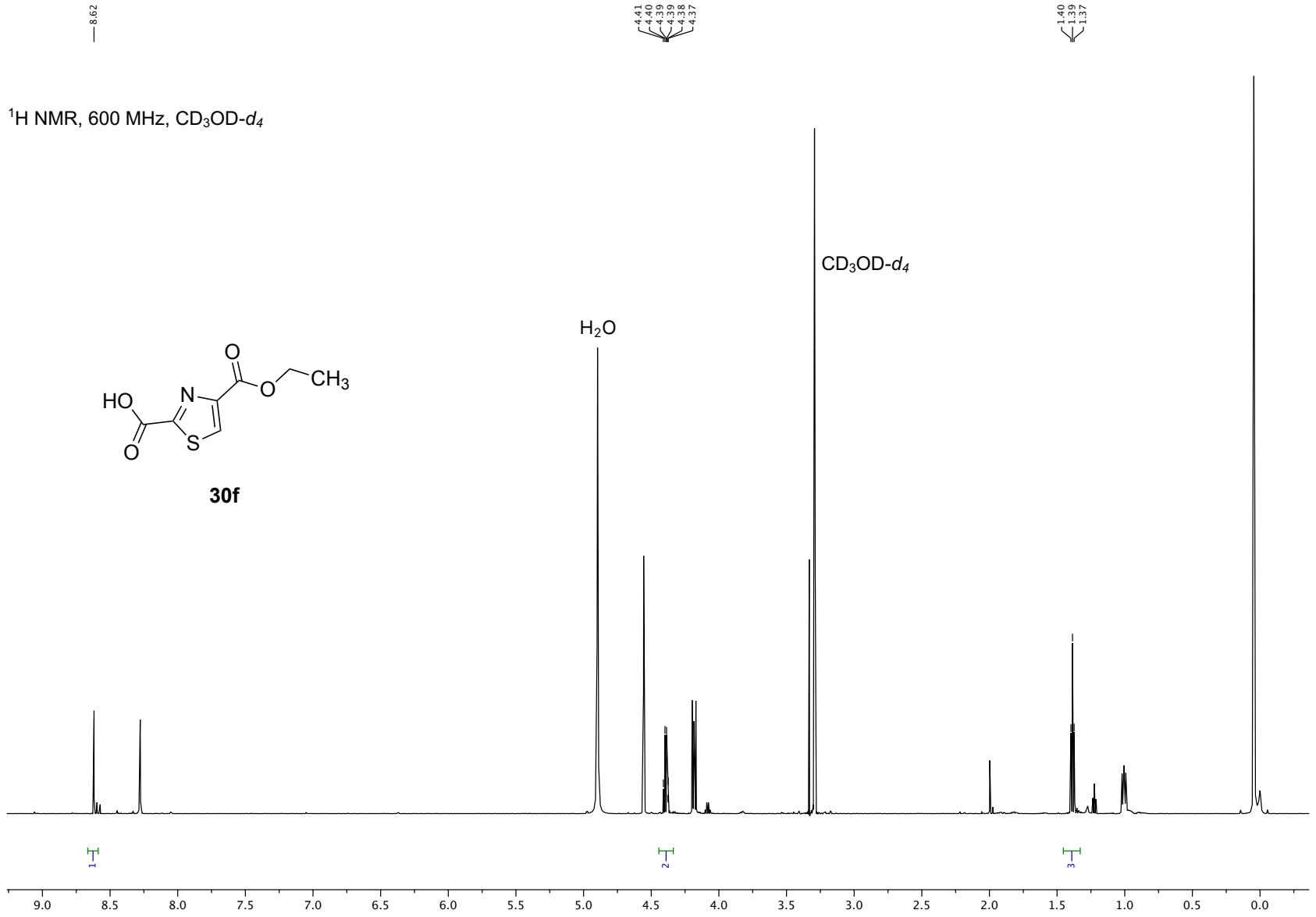

171.1
162.1
156.6
146.8
128.6
62.3
42.2
17.3
-1.4
^{13}C NMR, 100 MHz, DMSO-d_6
DMSO-d_6
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0

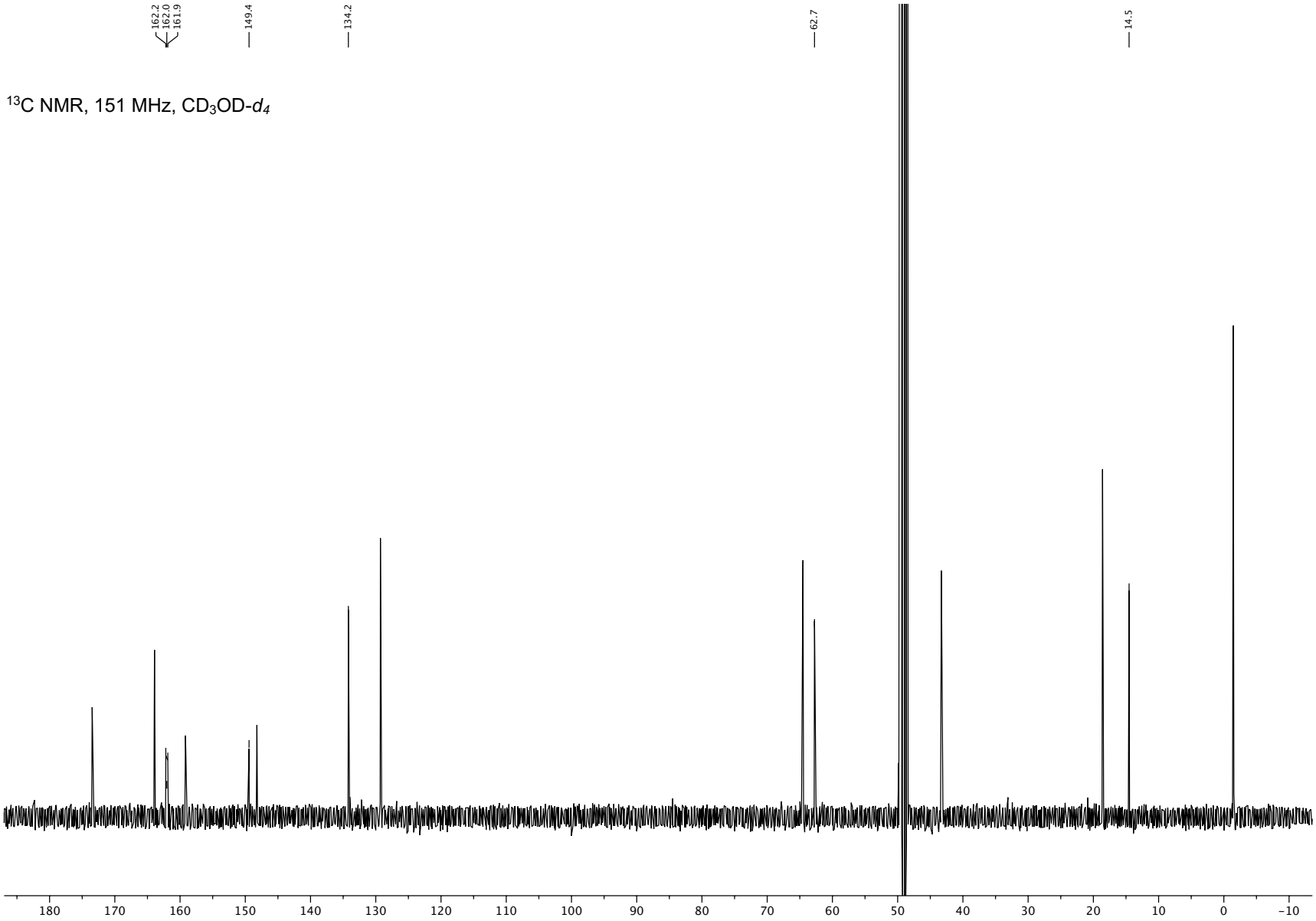

7.95
5.36
4.67
4.66
4.23
4.22
4.21
4.20
4.19
3.87
3.86
3.84
3.67
3.65
3.64
1.98
1.97
1.96
1.95
1.94
1.93
1.92
1.91
1.89
1.88
1.03
1.01
1.00
0.98
0.05
0.04
0.03
^{1}H NMR, 400 MHz, $CDCl_3$
$(CH_3)_3Si$
O
O
N
H
S
N
O
N
30c
$CDCl_3$
H_2O
1
1
2
2
2
2
4
2
9
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0

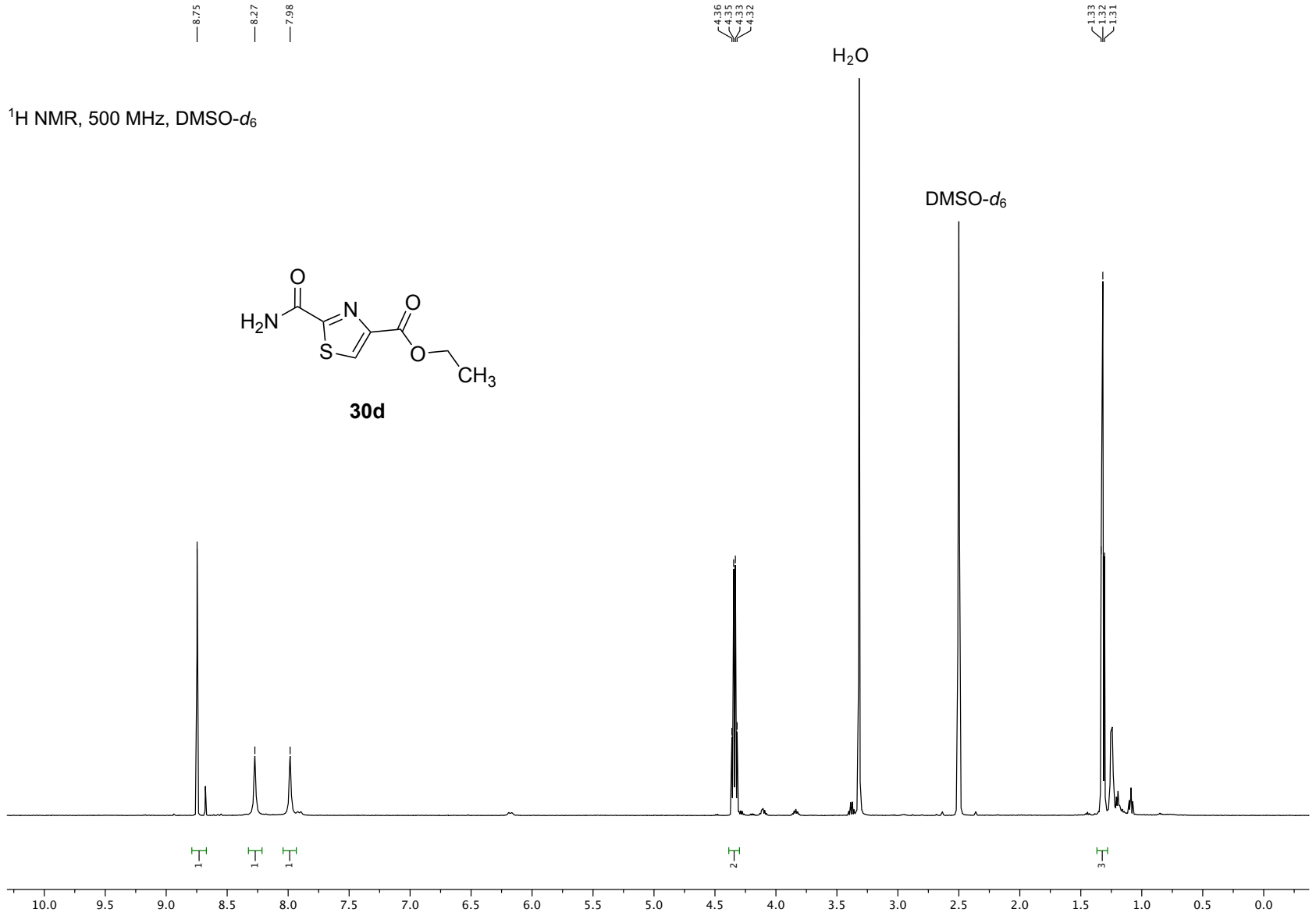

167.5
161.7
156.7
151.5
124.9
64.0
49.2
47.1
42.8
26.7
24.1
17.9
-1.3
^{13}C NMR, 100 MHz, $CDCl_3$
$CDCl_3$
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0

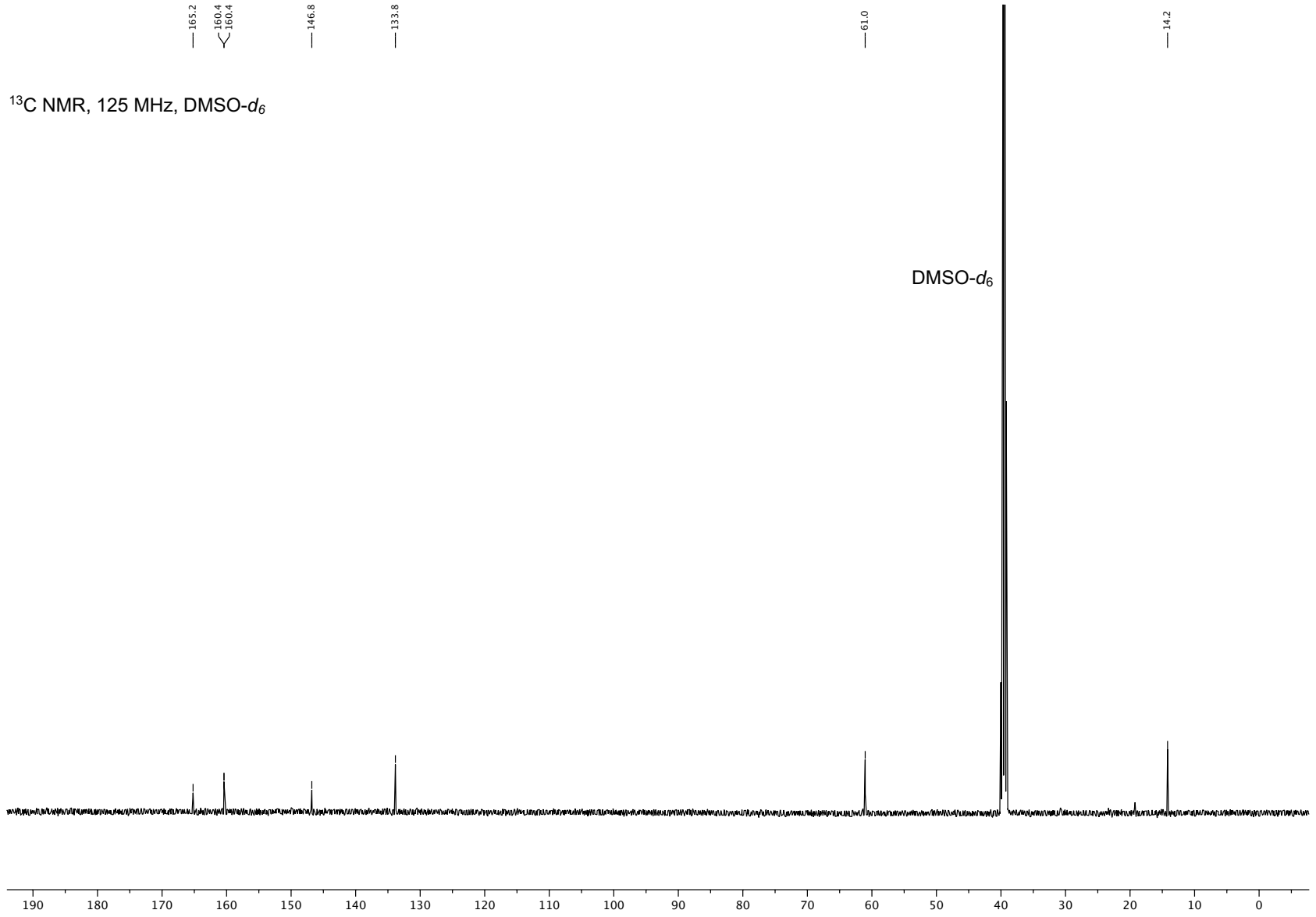

8.67
4.44
4.42
4.41
4.39
4.01
1.42
1.40
1.39
^{1}H NMR, 500 MHz, CD_3OD-d_4
H_2O
CD_3OD-d_4
O
CH_3
O
N
O
S
O
CH_3
30e
1
2
3
3
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0


162.0
161.0
160.1
149.6
134.3
62.8
53.9
14.5
^{13}C NMR, 125 MHz, CD_3OD-d_4
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0

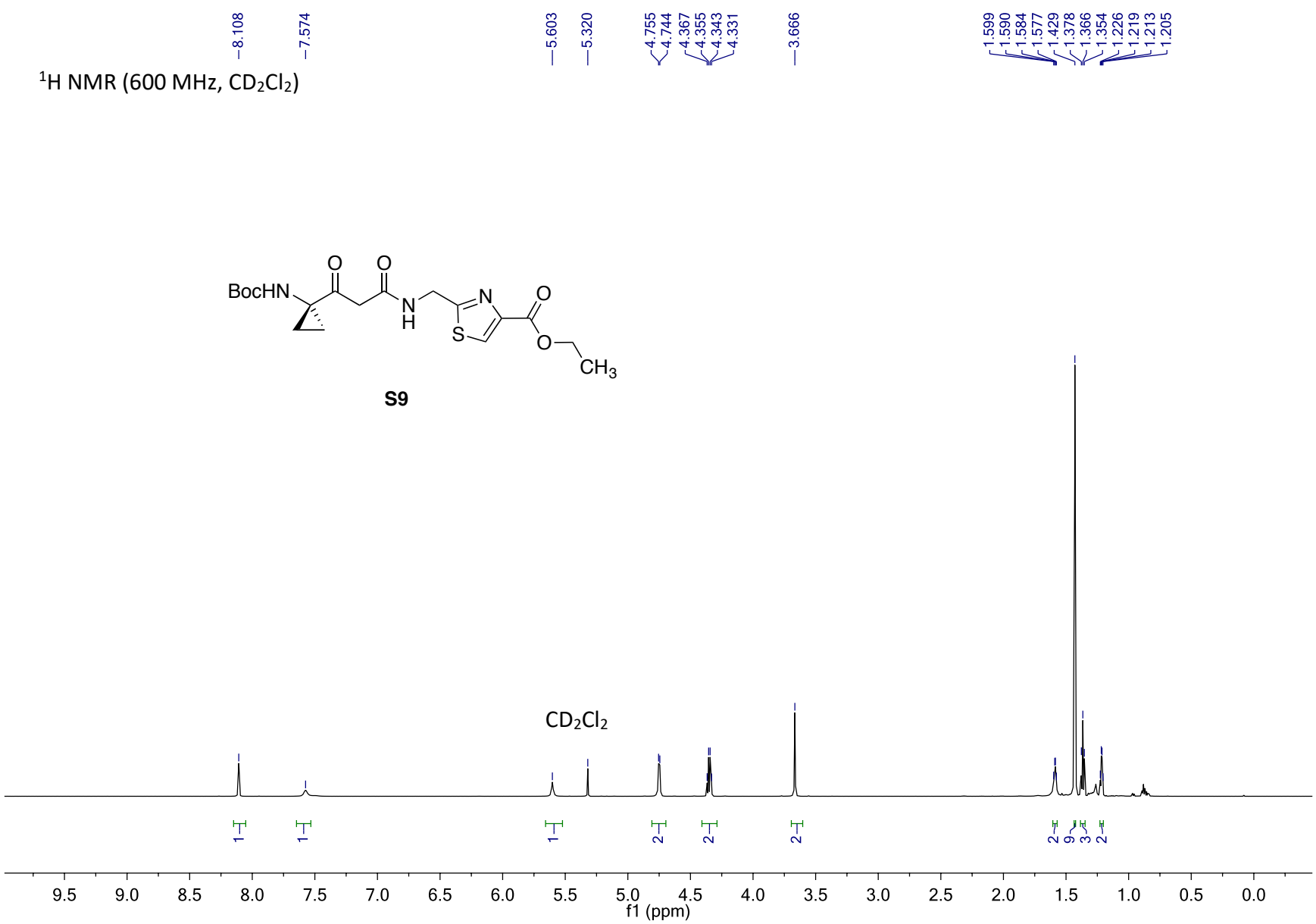

8.62
4.41
4.40
4.39
4.39
4.38
4.37
1.40
1.39
1.37
^{1}H NMR, 600 MHz, CD_3OD-d_4
H_2O
CD_3OD-d_4
HO
O
N
O
CH_3
S
O
30f
1
2
3
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0

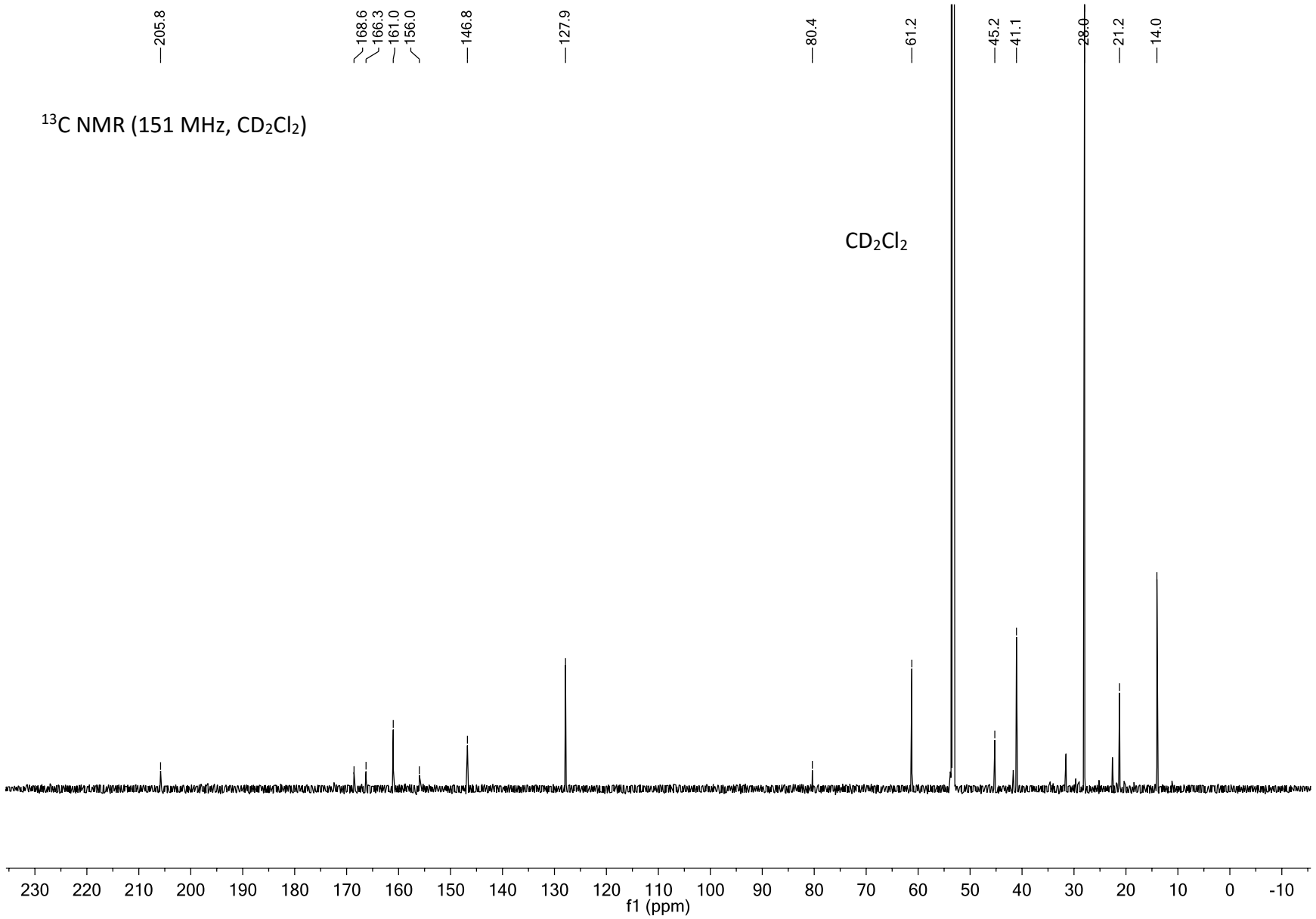

162.2
162.0
161.9
149.4
134.2
62.7
14.5
^{13}C NMR, 151 MHz, CD_3OD-d_4
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10

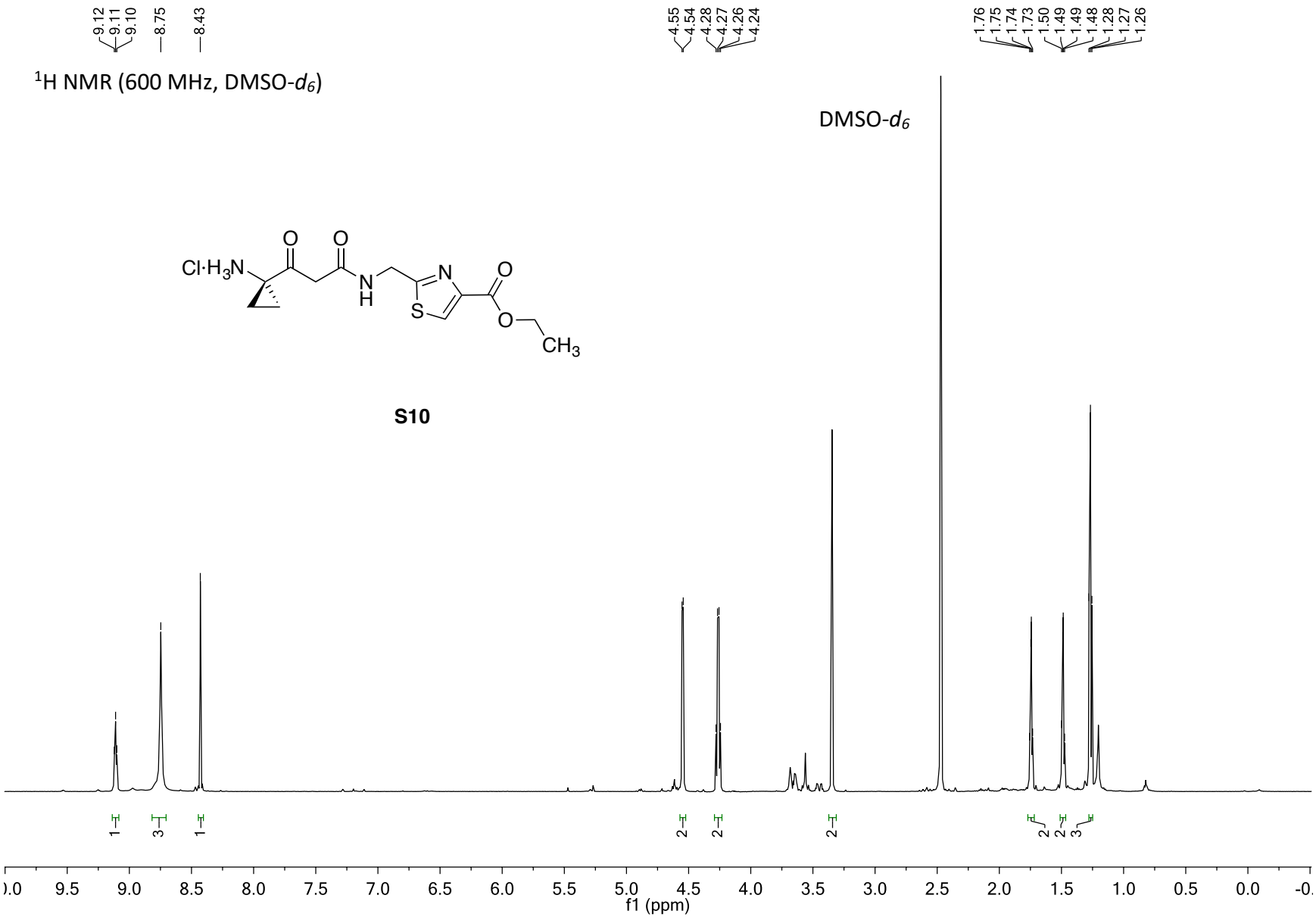

8.75
8.27
7.98
4.36
4.35
4.33
4.32
1.33
1.32
1.31
H2O
DMSO-d6
1H NMR, 500 MHz, DMSO-d6
O
H2N
N
O
S
O
CH3
30d
1
1
1
2
3
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0

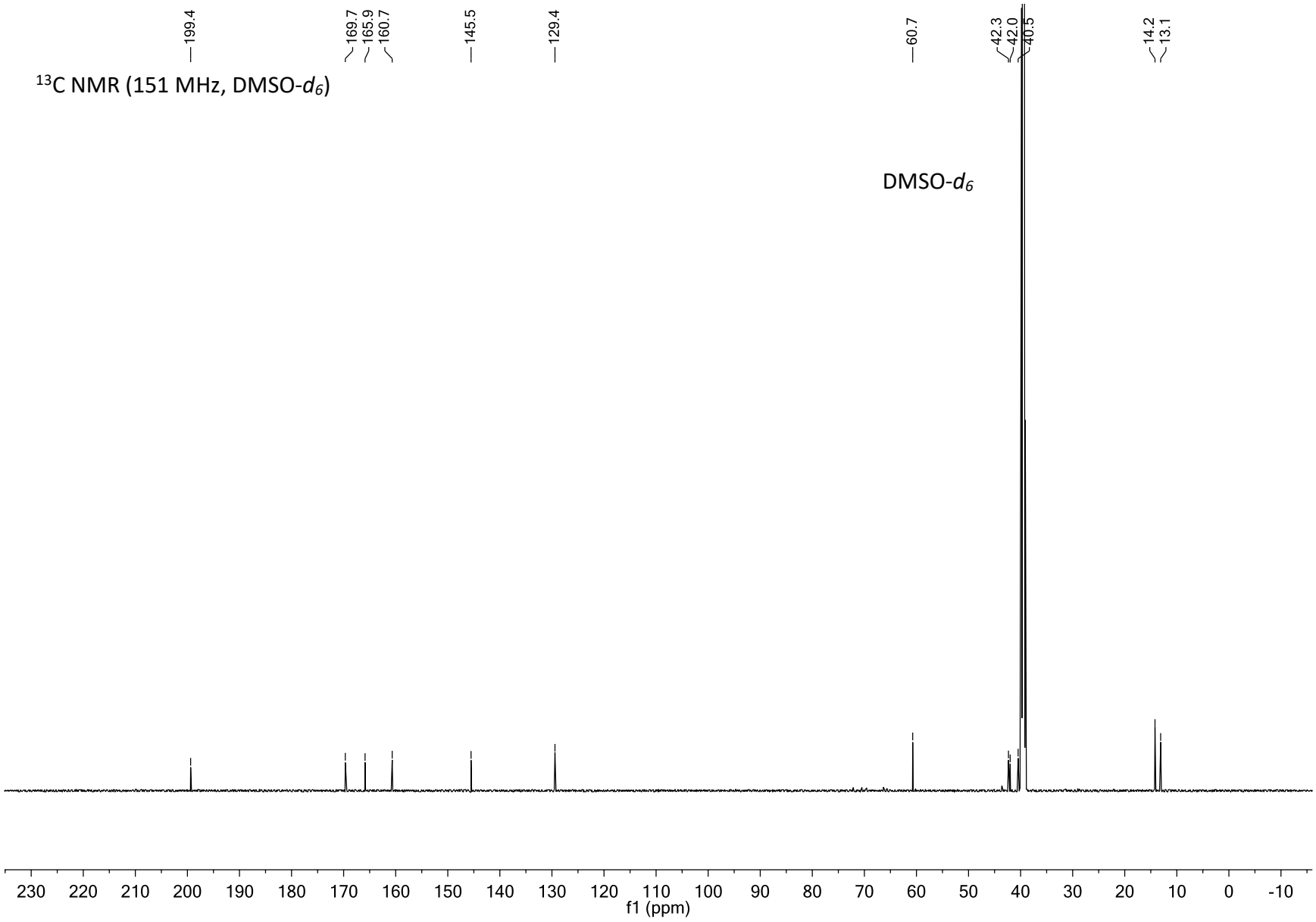

165.2
160.4
160.4
146.8
133.8
61.0
14.2
^{13}C NMR, 125 MHz, DMSO-d_6
DMSO-d_6
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0

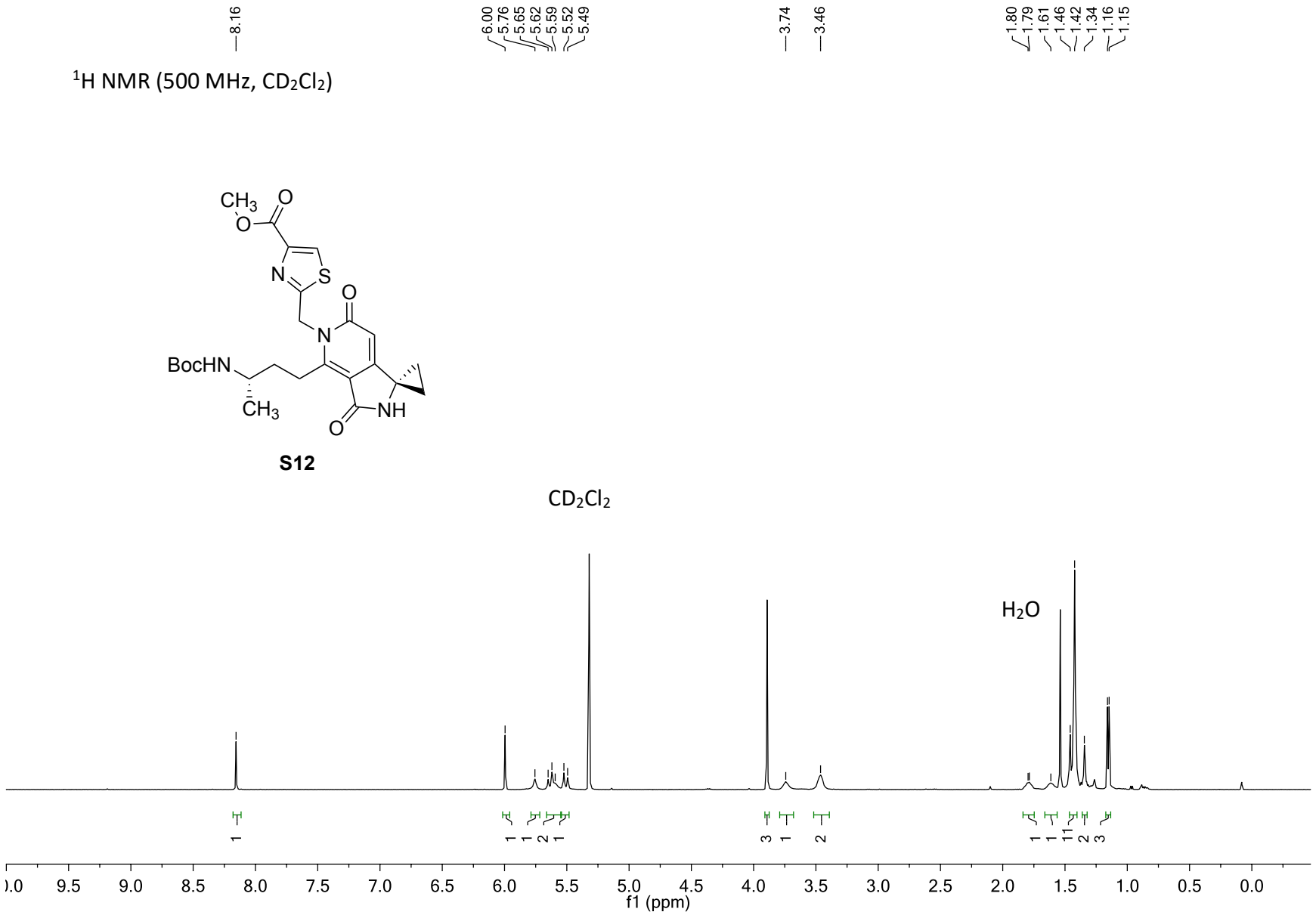
^{1}H NMR (600 MHz, CD_2Cl_2)

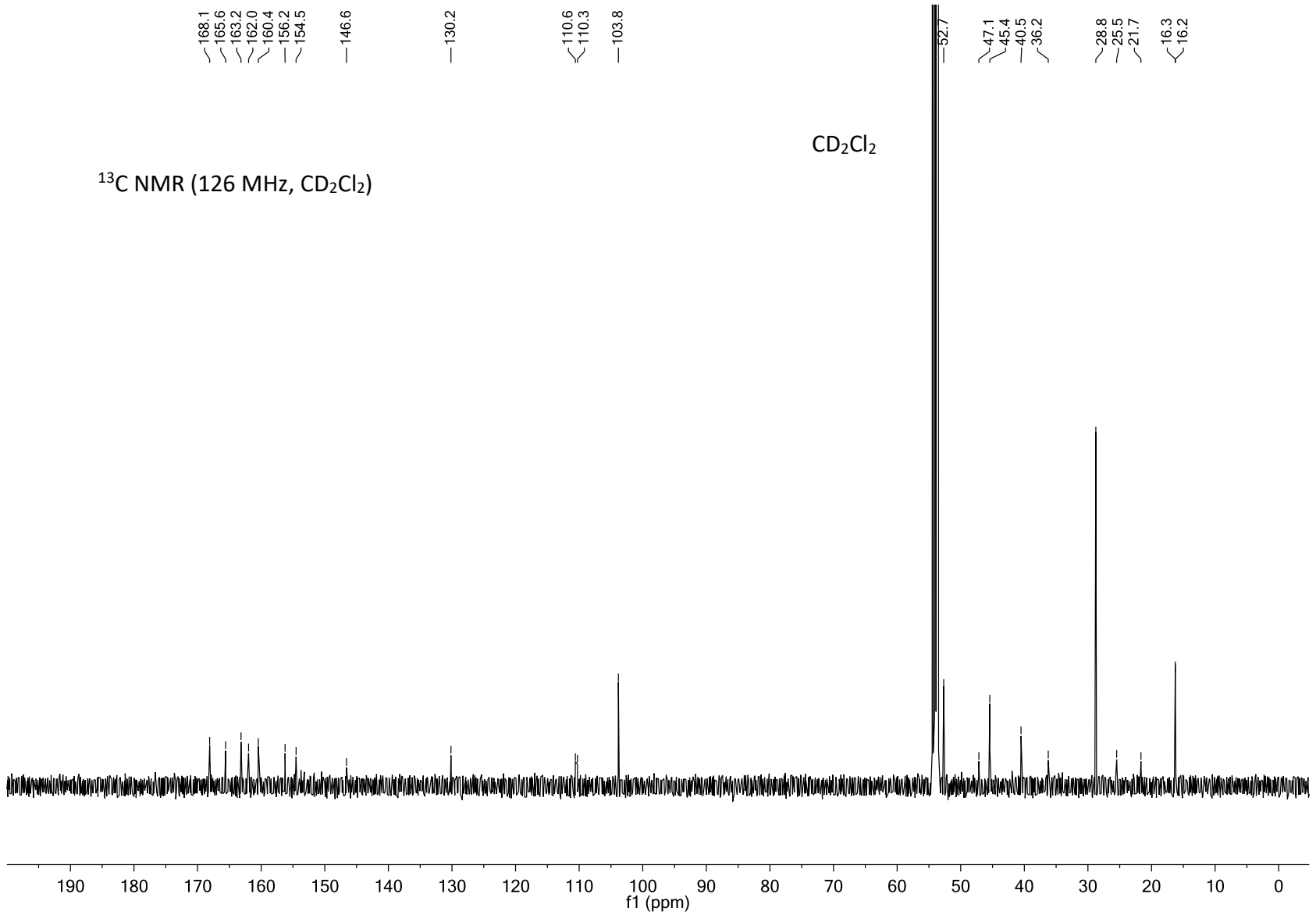



205.8
168.6
166.3
161.0
156.0
146.8
127.9
80.4
61.2
45.2
41.1
28.0
21.2
14.0
13C NMR (151 MHz, CD2Cl2)
CD2Cl2
230 220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10
f1 (ppm)

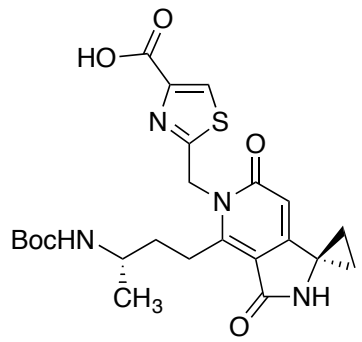

9.12
9.11
9.10
8.75
8.43
4.55
4.54
4.28
4.27
4.26
4.24
1.76
1.75
1.74
1.73
1.50
1.49
1.49
1.48
1.28
1.27
1.26
^{1}H NMR (600 MHz, DMSO-d_6)
DMSO-d_6
Cl·H$_3$N
O
O
N
H
N
S
O
O
CH$_3$
S10
1
3
1
2
2
2
2
2
3
f1 (ppm)


199.4
169.7
165.9
160.7
145.5
129.4
60.7
42.3
42.0
40.5
14.2
13.1
^{13}C NMR (151 MHz, DMSO-d_6)
DMSO-d_6
230 220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10
f1 (ppm)

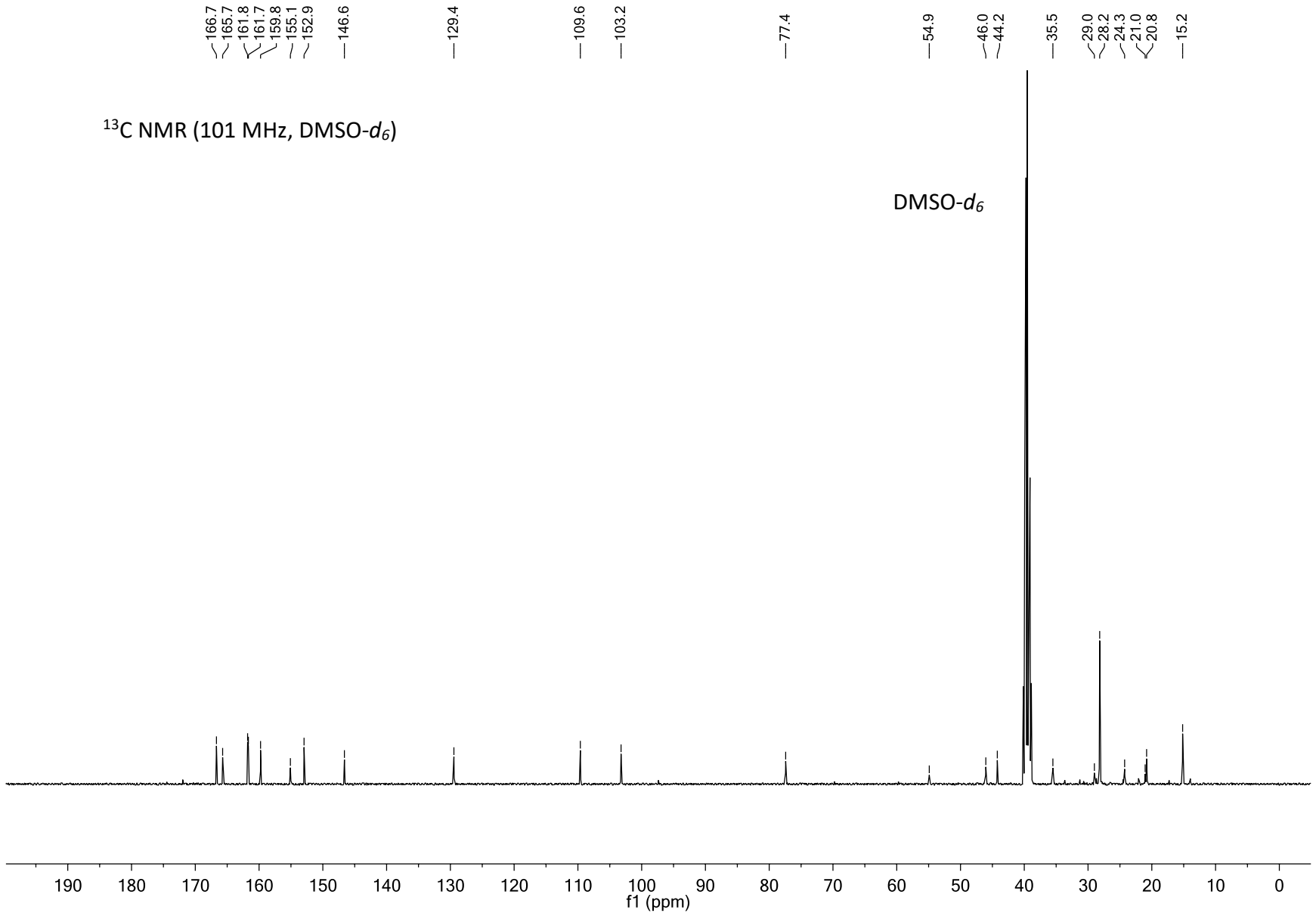

8.16
6.00
5.76
5.65
5.62
5.59
5.52
5.49
3.74
3.46
1.80
1.79
1.61
1.46
1.42
1.34
1.16
1.15
^{1}H NMR (500 MHz, CD_2Cl_2)
CH3
O
O
N
S
O
N
BocHN
CH3
NH
O
S12
CD_2Cl_2
H_2O
f1 (ppm)

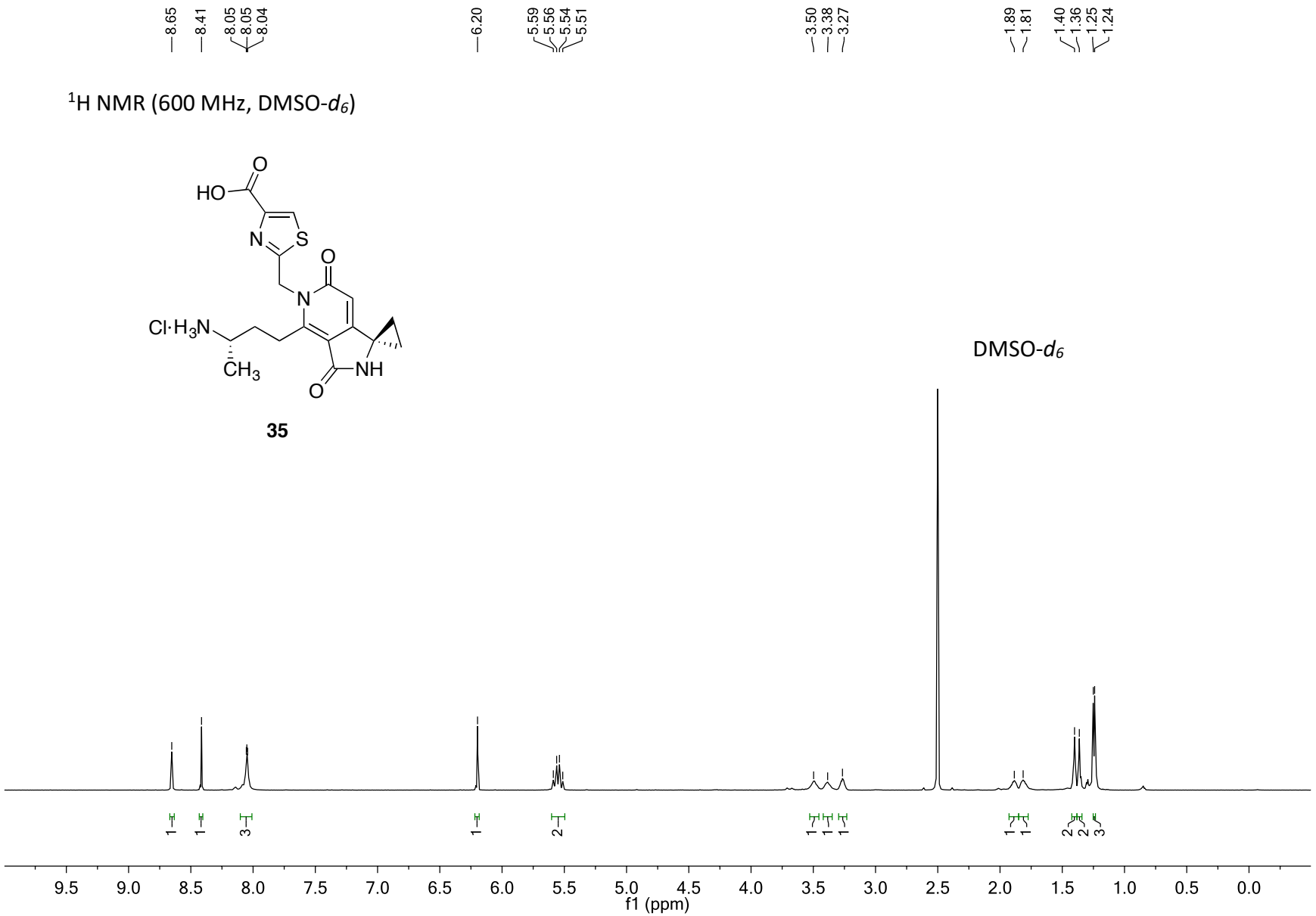

168.1
165.6
163.2
162.0
160.4
156.2
154.5
146.6
130.2
110.6
110.3
103.8
52.7
47.1
45.4
40.5
36.2
28.8
25.5
21.7
16.3
16.2
CD2Cl2
13C NMR (126 MHz, CD2Cl2)
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

^{1}H NMR (400 MHz, DMSO-d_6)



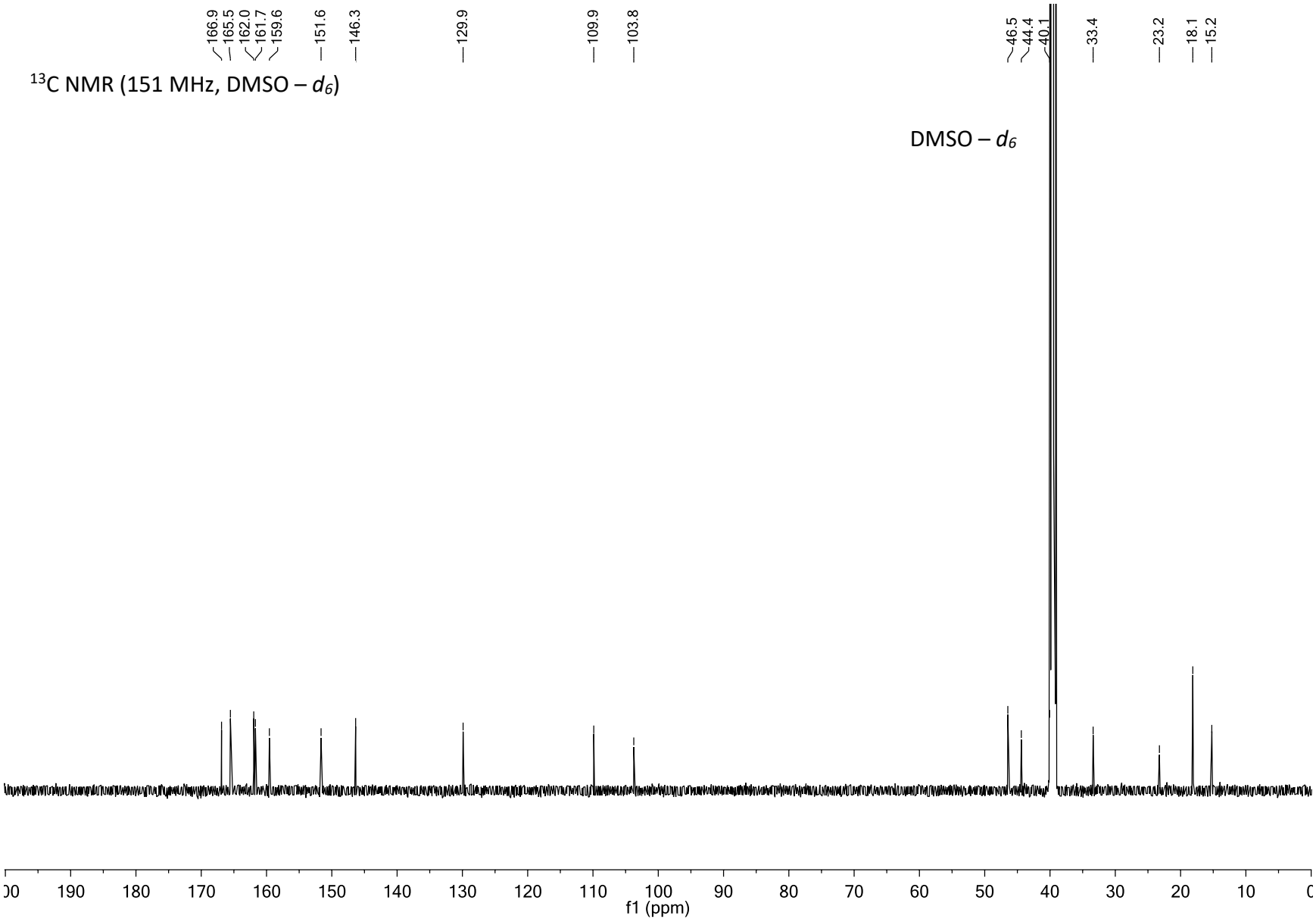
S13


166.7
165.7
161.8
161.7
159.8
155.1
152.9
146.6
129.4
109.6
103.2
77.4
54.9
46.0
44.2
35.5
29.0
28.2
24.3
21.0
20.8
15.2
^{13}C NMR (101 MHz, DMSO-d_6)
DMSO-d_6
190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0
f1 (ppm)



^{1}H NMR (600 MHz, DMSO-d_6)

35


166.9
165.5
162.0
161.7
159.6
151.6
146.3
129.9
109.9
103.8
46.5
44.4
40.1
33.4
23.2
18.1
15.2
^{13}C NMR (151 MHz, DMSO – d_6)
DMSO – d_6
f1 (ppm)

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