

Supporting Information

Chloramphenicol Derivatives with Antibacterial Activity Identified by Functional Metagenomics

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Experimental procedure for the synthesis of compounds 15–18

To a solution of **1** (3 g) in dry DCM (10 mL) was added imidazole (0.756 g, 1.2 eq.) in three portions. After 5 min TBSCl (1.675 g, 1.1 eq.) was added and stirring continued for 2 h. The reaction mixture was diluted with H₂O (10 mL) and extracted with DCM (200 mL × 2). Removal of the solvent by evaporation gave a crude mixture, which was chromatographed on silica gel using 10% EtOAc in hexanes to afford compound **13** (4.02 g, 99%) as a colorless liquid; ¹H NMR (CDCl₃, 400 MHz) δ 8.20 (2H, d, *J* = 8.6 Hz), 7.56 (2H, d, *J* = 8.6 Hz), 7.20 (1H, d, *J* = 9.4 Hz, NH), 5.80 (1H, s, CHCl₂), 5.30 (1H, brs, H-1), 4.14 (1H, m), 4.01 (2H, m, H-2, 3a), 3.94 (1H, m, H-3b), 0.94 [9H, s, (CH₃)₃C–Si], 0.14 [6H, s, (CH₃)₂–Si]; ¹³C NMR (CDCl₃, 100 MHz) δ 164.2, 147.7, 147.6, 126.8, 123.7, 74.0, 66.3, 65.2, 55.1, 25.9, 18.2, -5.4, -5.5.

To a solution of **13** (50 mg) in dry DCM (3 mL) was added pyridine (0.02 mL, 1.2 eq.) and an acyl reagent (acetic anhydride, propanoic anhydride, *p*-nitrobenzoyl chloride, and cinnamoyl chloride) (1.2 eq.). After stirring at room temperature for 4 h, the solvents were removed from the reaction mixture by evaporation. The resultant residue was chromatographed on silica gel using 10% EtOAc in hexanes to afford compounds **14a–14d** in a yield of approximately 95%.

1-Acetyl-3-tert-butyldimethylsilylchloramphenicol (14a): colorless liquid; ¹H NMR (CDCl₃, 400 MHz) δ 8.24 (2H, d, *J* = 8.6 Hz), 7.58 (2H, d, *J* = 8.6 Hz), 6.97 (1H, d, *J* = 9.4 Hz, NH), 6.12 (1H, d, *J* = 7.8 Hz, H-1), 5.91 (1H, s, CHCl₂), 4.34 (1H, m, H-2), 3.62 (1H, dd, *J* = 10.9, 4.7 Hz, H-3a), 3.42 (1H, dd, *J* = 10.9, 2.3 Hz, H-3b), 2.12 (3H, s), 0.93 [9H, s, (CH₃)₃C–Si], 0.06 (3H, s, CH₃–Si), 0.05 (3H, s, CH₃–Si); ¹³C NMR (CDCl₃, 100 MHz) δ 170.1, 164.1, 148.2, 144.3, 128.3, 124.1, 73.6, 66.4, 61.5, 55.1, 25.9, 21.1, 18.3, -5.4, -5.5.

1-Propanoyl-3-tert-butyldimethylsilylchloramphenicol (14b): colorless liquid; ¹H NMR (CDCl₃, 400 MHz) δ 8.19 (2H, d, *J* = Hz), 7.55 (2H, d, *J* = Hz), 6.99 (1H, d, *J* = 9.4 Hz, NH), 6.10 (1H, d, *J* =

7.8 Hz, H-1), 5.90 (1H, s, CHCl_2), 4.30 (1H, m, H-2), 3.59 (1H, dd, $J = 10.9, 4.7$ Hz, H-3a), 3.39 (1H, d, $J = 10.9, 3.1$ Hz, H-3b), 2.37 (2H, m), 1.08 (3H, t, $J = 7.8$ Hz), 0.89 [9H, s, $(\text{CH}_3)_3\text{C-Si}$], 0.02 (3H, s, $\text{CH}_3\text{-Si}$), 0.01 (3H, s, $\text{CH}_3\text{-Si}$); ^{13}C NMR (CDCl_3 , 100 MHz) δ 173.5, 164.0, 148.0, 144.4, 128.1, 123.9, 73.3, 66.3, 61.4, 55.1, 27.6, 25.8, 18.2, 8.9, -5.5, -5.6.

1-Cinnamoyl-3-tert-butyldimethylsilylchloramphenicol (14c): colorless liquid; ^1H NMR (CDCl_3 , 400 MHz) δ 8.26 (2H, d, $J = 8.6$ Hz), 7.73 (1H, d, $J = 15.6$ Hz), 7.64 (2H, d, $J = 9.4$ Hz), 7.54 (2H, m), 7.40 (3H, m), 7.07 (1H, d, $J = 9.4$ Hz, NH), 6.46 (1H, d, $J = 15.6$ Hz), 6.21 (1H, d, $J = 7.8$ Hz, H-1), 5.91 (1H, s, CHCl_2), 4.43 (1H, m, H-2), 3.67 (1H, dd, $J = 10.9, 3.9$ Hz, H-3a), 3.46 (1H, dd, $J = 10.9, 2.3$ Hz, H-3b), 0.94 [9H, s, $(\text{CH}_3)_3\text{C-Si}$], 0.07 (3H, s, $\text{CH}_3\text{-Si}$), 0.06 (3H, s, $\text{CH}_3\text{-Si}$); ^{13}C NMR (CDCl_3 , 100 MHz) δ 165.9, 163.9, 148.1, 146.8, 144.4, 133.9, 130.8, 128.9, 128.3, 128.1, 123.9, 116.6, 73.7, 66.4, 61.4, 55.0, 25.9, 18.3, -5.4, -5.5.

1-p-Nitrobenzoyl-3-tert-butyldimethylsilylchloramphenicol (14d): colorless liquid; ^1H NMR (CDCl_3 , 400 MHz) δ 8.29 (4H, d, $J = 7.8$ Hz), 8.21 (2H, d, $J = 8.6$ Hz), 7.71 (2H, d, $J = 8.6$ Hz), 7.18 (1H, d, $J = 9.4$ Hz, NH), 6.25 (1H, d, $J = 9.4$ Hz, H-1), 5.89 (1H, s, CHCl_2), 4.56 (1H, m, H-2), 3.68 (1H, dd, $J = 10.9, 3.1$ Hz, H-3a), 3.44 (1H, br d, $J = 10.9$ Hz, H-3b), 0.96 [9H, s, $(\text{CH}_3)_3\text{C-Si}$], 0.08 [6H, br s, $(\text{CH}_3)_2\text{-Si}$]; ^{13}C NMR (CDCl_3 , 100 MHz) δ 164.1, 163.9, 150.9, 148.4, 143.4, 134.4, 131.1, 128.4, 124.2, 123.7, 75.9, 66.2, 61.3, 54.6, 25.8, -5.5, -5.7; HRESIMS m/z 586.1170/588.1148 (calcd for $[\text{C}_{24}\text{H}_{29}\text{N}_3\text{Cl}_2\text{O}_8\text{Si} + \text{H}]^+$, 586.1174/588.1144).

To a solution of a compound in **14a**, **14b**, or **14c** (30 mg) in H_2O -THF (1:4, 3 mL) was added PTSA (0.1 eq) in two portions. After stirring at room temperature for overnight, the reaction mixture was diluted with H_2O (5 mL) and extracted with EtOAc (20 mL $\times$ 2). The organic layer was concentrated to dryness to give a residue, which was chromatographed on silica gel using 30% EtOAc in hexanes to give the target compound.

1-Acetylchloramphenicol (15): yield, 60% from **14a**, white solid; $[\alpha]_D^{25} +30.7$ (c 0.35, EtOH); ^1H and ^{13}C NMR data matching the reported values in literature (Gross, F.; Lewis, E. A.; Pirae, M.; van Pee, K. H.; Vining, L. C.; White, R. L. *Bioorg. Med. Chem. Lett.* **2002**, 12, 283-286); HRESIMS m/z 387.0125/389.0096 $[\text{M} + \text{Na}]^+$ (calcd for $[\text{C}_{13}\text{H}_{14}\text{N}_2\text{Cl}_2\text{O}_6 + \text{Na}]^+$, 387.0121/389.0092).

1-Propanoylchloramphenicol (16): yield, 60% from **14b**, white solid; $[\alpha]_D^{25} -5.6$ (c 0.6, DCM); ^1H and ^{13}C NMR data matching reported values in literature (Gross, F.; Lewis, E. A.; Pirae, M.; van Pee, K. H.; Vining, L. C.; White, R. L. *Bioorg. Med. Chem. Lett.* **2002**, 12, 283-286); HRESIMS m/z 401.0287/403.0256 $[\text{M} + \text{Na}]^+$ (calcd for $[\text{C}_{14}\text{H}_{16}\text{N}_2\text{Cl}_2\text{O}_6 + \text{Na}]^+$, 401.0278.0458/403.0248).

1-Cinnamoylchloramphenicol (17): yield, 85% from **14c**, white solid; $[\alpha]_D^{25} + 31.2$ (c 0.16, EtOH); ^1H NMR (CDCl_3 , 400 MHz) δ 8.23 (2H, d, $J = 8.6$ Hz), 7.76 (1H, d, $J = 15.6$ Hz), 7.65 (2H, d, $J = 8.6$ Hz), 7.53 (2H, m), 7.40 (3H, m), 7.09 (1H, d, $J = 8.6$ Hz, NH), 6.50 (1H, d, $J = 15.6$ Hz), 6.33 (1H, d, $J = 6.3$ Hz, C-1), 5.89 (1H, s, CHCl₂), 4.47 (1H, m, H-2), 3.74 (1H, dd, $J = 10.9, 4.7$ Hz, H-3a), 3.59 (1H, dd, $J = 10.9, 3.1$ Hz, H-3b); ^{13}C NMR (CDCl_3 , 100 MHz) δ 166.2, 164.5, 148.2, 147.4, 144.3, 133.9, 131.1, 129.2, 128.5, 128.0, 124.1, 116.5, 73.3, 66.4, 61.2, 55.6; HRESIMS m/z 451.0477/453.0450 (calcd for $[\text{C}_{20}\text{H}_{18}\text{N}_2\text{Cl}_2\text{O}_6 - \text{H}]^-$, 451.0469/453.0440).

1-p-Nitrobenzoylchloramphenicol (18): yield, 82% from **14d**, white solid; $[\alpha]_D^{25} + 51.5$ (c 0.16, EtOH); ^1H NMR ($\text{MeOH-}d_4/\text{CDCl}_3$, 400 MHz) δ 8.27 (6H, m), 7.75 (2H, d, $J = 8.6$ Hz), 6.31 (1H, d, $J = 6.3$ Hz, H-1), 6.03 (1H, s, CHCl₂), 4.55 (1H, m, H-2), 3.68 (1H, dd, $J = 10.9, 5.4$ Hz, H-3a), 3.47 (1H, dd, $J = 10.9, 3.2$ Hz, H-3b); ^{13}C NMR (acetone- d_6 , 100 MHz) δ 164.9, 164.5, 151.9, 148.9, 145.8, 135.8, 132.1, 129.1, 124.5, 76.2, 67.5, 61.1, 56.3; HRESIMS m/z 472.0307/474.0276 (calcd for $[\text{C}_{18}\text{H}_{15}\text{N}_3\text{Cl}_2\text{O}_8 + \text{H}]^+$, 472.0309/474.0279).

Table S1. Characterization of metagenomic clone-derived bioactivity inhibiting MRSA growth. MRSA strain EAMC30 was used as the tester strain in all bioassays. Clones that were observed to produce Cm derivatives are in bold. ^aPercent inhibition values were calculated in comparison with the corresponding empty vector negative control by measuring the fluorescence of reduced resazurin.

Clone ID	Predicted phylum of origin	^a % MRSA Inhibition	% Inhibition in the cell lysate	% Inhibition in the supernatant	% Inhibition after 10 min at 100 °C	% Inhibition in < 3 kDa fraction
P2P12	Bacteroidetes	52 ± 4.5	34.6 ± 8.0	48 ± 4.5	56.7 ± 1.9	59.6 ± 1.6
P3A13	Acidobacteria	8.1 ± 0.2	89 ± 0.5	91.9 ± 0.2	86.4 ± 0.5	38.7 ± 9.8
P5A4	Proteobacteria	63.7 ± 6.1	3.6 ± 0.1	36.3 ± 6.1	24.7 ± 3.7	52.2 ± 0.5
P5C24	Acidobacteria	17.8 ± 1.6	84.2 ± 1.4	82.2 ± 1.6	68.7 ± 3.3	65 ± 6.3
P6B5	Proteobacteria	43.1 ± 0.5	56.9 ± 2.1	19.3 ± 0.5	92.2 ± 0.6	76.5 ± 3.4
P6L4	Unknown	7.5 ± 0.4	88.3 ± 0.1	92.5 ± 0.4	89.1 ± 0.1	85.4 ± 1.5
P6L5	Proteobacteria	25.7 ± 1.8	6.8 ± 4.2	74.3 ± 1.8	14.9 ± 6.0	54.4 ± 3.8
P9L21	Unknown	7.8 ± 0.3	5.7 ± 1.7	92.2 ± 0.3	58.4 ± 1.3	71.8 ± 2.8
P14O1	Planctomycetes	75.2 ± 6.1	12.6 ± 3.7	24.8 ± 6.1	-2.1 ± 3.0	36.7 ± 0.9
P18N22	Proteobacteria	10.8 ± 0.3	5.9 ± 4.9	89.2 ± 0.3	60.8 ± 4.8	72.3 ± 0.0
P20I6	Acidobacteria	11.4 ± 0.6	55.1 ± 3.0	88.6 ± 0.6	78.5 ± 1.7	22.9 ± 15.2
P22C4	Acidobacteria	60.5 ± 1.9	24 ± 0.8	39.5 ± 1.9	-3.1 ± 7.2	39.1 ± 3.9
P22E10	Proteobacteria	53.4 ± 0.5	7.6 ± 4.9	46.6 ± 0.5	71.7 ± 3.5	54.9 ± 2.3
P23K15	Proteobacteria	7.7 ± 0.3	86.7 ± 1.8	92.3 ± 0.3	86.5 ± 0.7	81.0 ± 3.5
P27K16	Gemmatimonadetes	11.6 ± 0.1	15.2 ± 4.9	88.4 ± 0.1	8.3 ± 5.4	52.8 ± 5.4
P27M10	Acidobacteria	7.7 ± 0.0	87 ± 0.4	92.3 ± 0.0	88.3 ± 0.4	82.7 ± 0.5
P28H1	Firmicutes	8.6 ± 0.1	86.8 ± 0.6	91.4 ± 0.1	80.8 ± 0.6	76.5 ± 1.0
P28I7	Acidobacteria	8.2 ± 0.1	72.8 ± 1.4	91.8 ± 0.1	84.4 ± 0.4	81.3 ± 0.9
P28L21	Acidobacteria	8.4 ± 0.1	70.7 ± 0.9	91.6 ± 0.1	75.6 ± 3.4	78.7 ± 0.6
P31G24	Acidobacteria	8.3 ± 0.1	87.9 ± 0.1	91.7 ± 0.1	86.3 ± 0.4	73.3 ± 7.8
P35B14	Proteobacteria	39.6 ± 2.1	34.9 ± 5.3	60.4 ± 2.1	-6.9 ± 12.6	48.2 ± 4.1
P36M1	Acidobacteria	56 ± 2.0	5.5 ± 0.8	44.0 ± 2.0	8.9 ± 14.2	55.8 ± 3.7
P37A11	Planctomycetes	59.5 ± 1.1	15.6 ± 3.9	40.5 ± 1.1	7.0 ± 0.5	53.7 ± 2.2
P37O10	Proteobacteria	41.2 ± 6.4	58.8 ± 1.8	29.1 ± 6.4	90.0 ± 1.1	16.0 ± 3.7
P43A3	Unknown	66.5 ± 1.2	3.8 ± 3.1	33.5 ± 1.2	28.5 ± 7.8	46.2 ± 0.5
P46O24	Bacteroidetes	8.6 ± 0.2	50.1 ± 5.2	91.4 ± 0.2	55.0 ± 6.7	65.7 ± 1.8
P49M4	Proteobacteria	10.0 ± 0.6	29.7 ± 7.6	90.0 ± 0.6	7.4 ± 7.2	55.7 ± 2.4

Table S2. Antibacterial Activity of Compounds **1–7**, **10–12**, and **15–18**.

Compound	IC ₅₀ ^a /MIC ^b (µg/mL)	
	<i>M. intracellulare</i>	<i>M. tuberculosis</i>
	ATCC 23068	ATCC 25177
1 (chloramphenicol)	17.2 ± 0.02 / 25.0 ± 0	67.6 ± 16.7 / – ^c
2	18.9 ± 1.2 / 25.0 ± 0	–/–
3	32.3 ± 9.1 / 75.0 ± 35.3	–/–
4	12.6 ± 0.5 / 25.0 ± 0	–/–
5	–/–	–/–
6	23.8 ± 2.5 / 25.0 ± 0	–/–
7	–/–	–/–
10	20.2 ± 4.2 / 37.5 ± 17.7	29.9 ± 2.5 / 50.0 ± 0
11	45.3 ± 0.1 / 50.0 ± 0	–/–
12	40.7 ± 11.7 / 50.0 ± 0	57.0 ± 7.0 / 100.0 ± 0
15	15.4 ± 1.4 / 25.0 ± 0	–/–
16	32.2 ± 2.0 / 50.0 ± 0	–/–
17	27.5 ± 2.0 / 50.0 ± 0	87.7 ± 2.4 / –
18	6.8 ± 0.2 / 12.5 ± 0	27.4 ± 6.2 / 50.0 ± 0
ciprofloxacin	0.8 ± 0.2 / 1.0 ± 0	0.9 ± 0.04 / 2.5 ± 0
streptomycin	1.8 ± 0.06 / 3.1 ± 0	3.3 ± 0.8 / 6.3 ± 0
rifampin	–/– ^c	–/– ^c

^a50% inhibition of bacterial growth. ^bMinimum inhibitory concentration (the lowest concentration that allows no detectable growth). ^cNot active at the highest test concentration 100 µg/mL.

Table S3. List of plasmids used in this study.

Plasmid name	Relevant characteristics ^a	Source or reference
pSmartBAC-S	Cm ^R , conjugative, integrative, inducible-copy	This study
pSMART HC Amp	Amp ^R , high copy number plamid	Lucigen Corp, WI
pSMART HC Amp: <i>trfA</i>	<i>trfA</i> cloned into pSMART HC Amp	This study
pRham N-His	Vector for expressing recombinant proteins	Lucigen Corp, WI
pRham-esterase	Esterase gene cloned into pRham vector	This study
pRham-carboxyesterase	Carboxylesterase gene cloned into pRham vector	This study

^a Abbreviations: Cm^R, chloramphenicol resistant;

Table S4. List of oligonucleotides used in this study.

Primer ID	Sequences
SR4F	5'-GGAGAAGTACCGCAAGCTGTCTG-3'
SR5R	5'-CGTCATAGTTCCTCGCGTGTCTG-3'
SR4R	5'-TCTTGGTCGTCATAGTTCCTCTG-3'
SL2F	5'-AAGATCGAGCGCGACAGCGT-3'
SR3F(R)	5'-AATCCGATCCGCACATGAGG-3'
SL1	5'-CAGTCCAGTTACGCTGGAGTC-3'
SR2	5'-GGTCAGGTATGATTAAATGGTCAGT-3'
9_SR6F	5'-GGTGATCTTCACGTCCTTGTTG-3'
9_SR6R	5'-CTTGTTTCGATATTGCGCCGTGG-3'
TRA-IntF	5'-GGAACATATGACGACCAAGAAGC-3'
TRA- IntR	5'- GCTCGATCTTGGCCGTAGCTT-3'
PE-F	5'- GCG CGATGGTCTTCTTTTAGT-3'
PE-R	5'-TTAAGCGAAAGCGTCGCCGGG-3'
ME-F	5'-ATCTATGGCGTCAAAAAGGTA-3'
ME-R	5'- CGGTGCCGCCCCGCAGCGTAAT-3'
PCE-F	5'- CCTTTGCTGCATCAGTTCTAC-3'
PCE-R	5'- TTCATGGTGCAGCCCTCGGAA-3'

Figure S1. Restriction fragment length polymorphism pattern of BAC DNAs isolated from representative metagenomic clones that inhibited MRSA growth.



Figure S2. Effect of arabinose induction on the antimicrobial activity against respective tester strains. Shown here is comparison of representative cultures grown in the presence and absence of arabinose indicating the % growth inhibition of the MRSA culture by the metagenomic clones and relative to the empty vector negative control, considered to have no inhibitory effect.

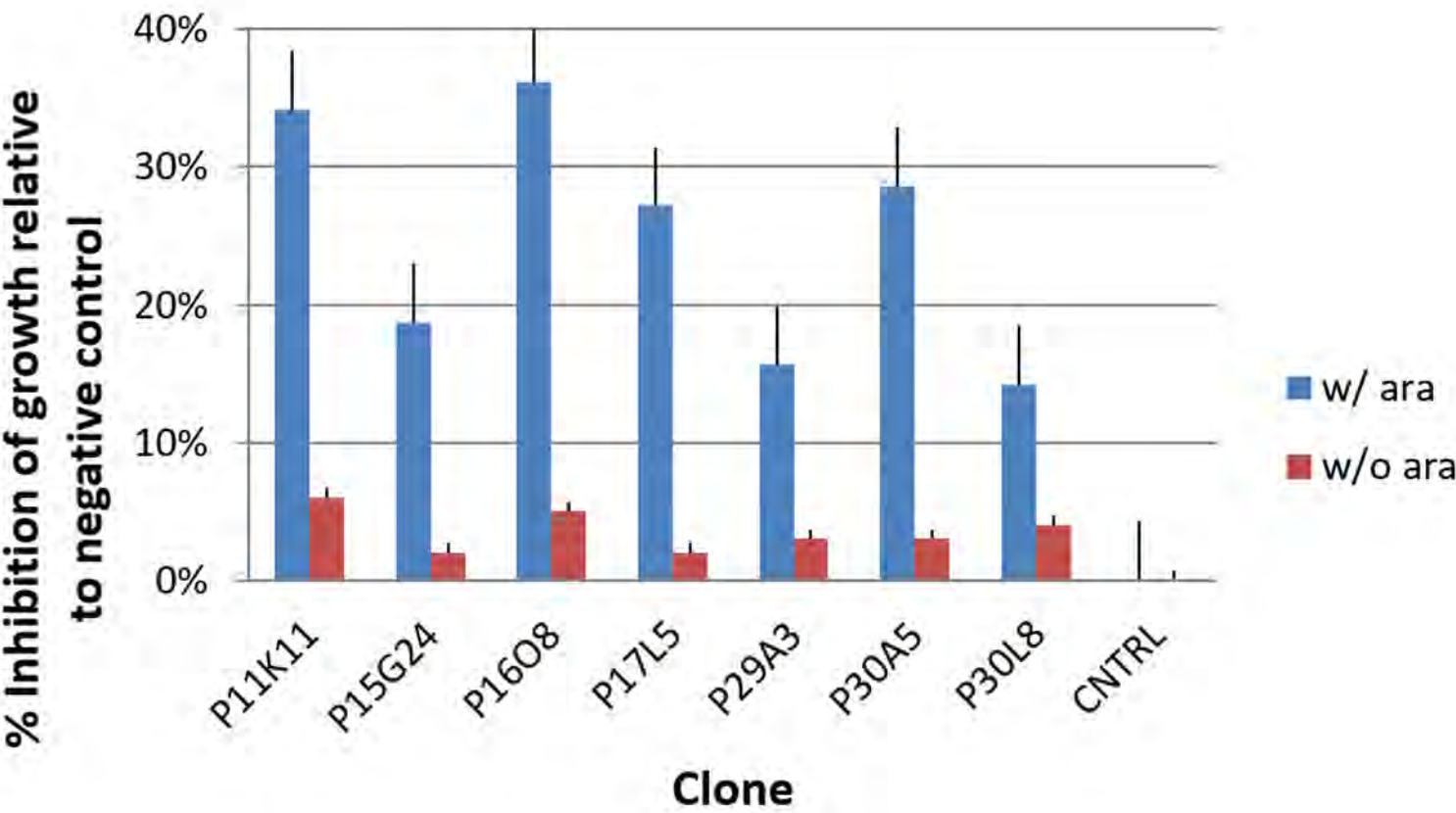
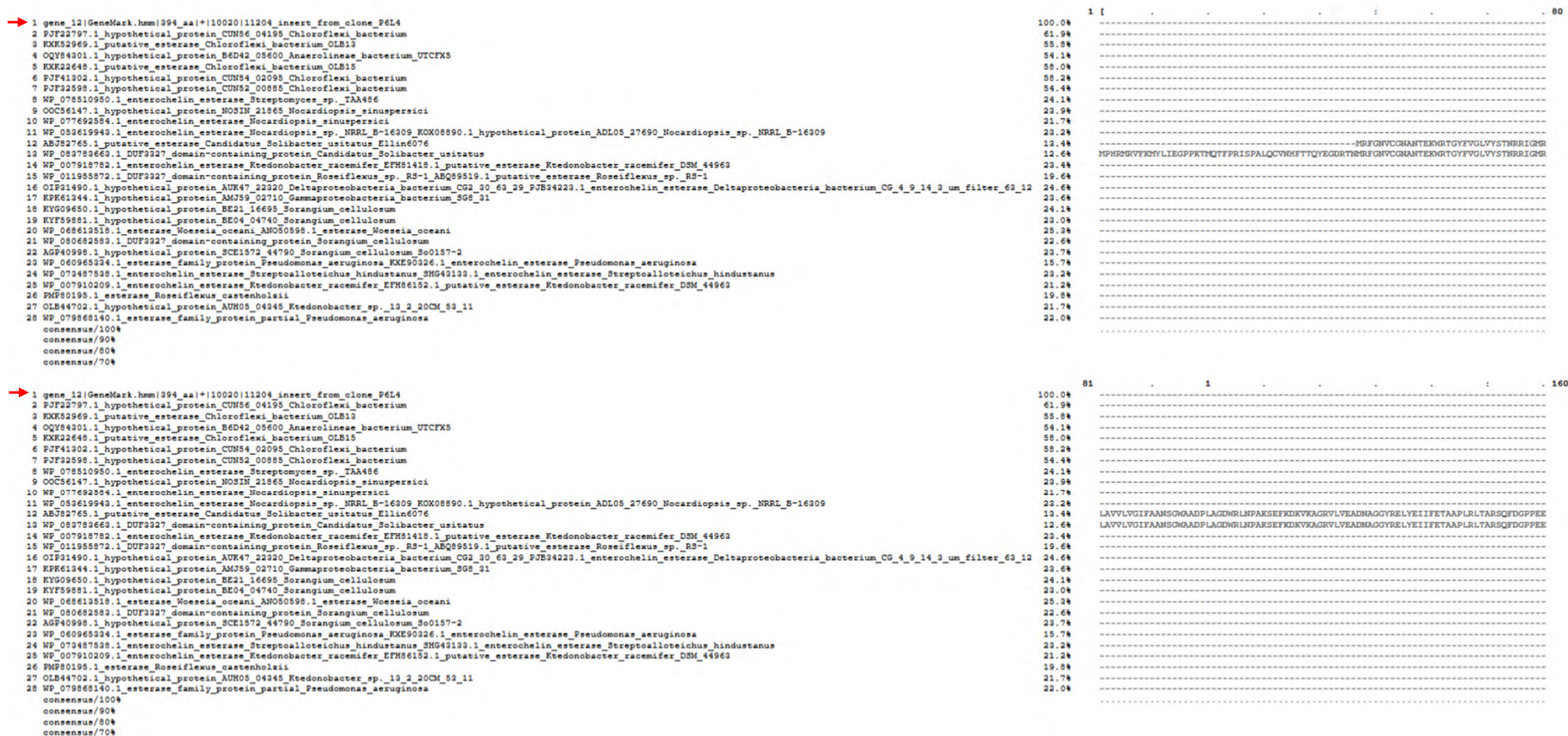


Figure S3. Multiple alignment of a putative esterase (clone P6L4; ORF12) compared to esterase amino acid sequences obtained from GenBank. The multiple alignment was conducted using MUSCLE and visualized using MVIEW. The sequence derived from metagenomic clone P6L4 is marked by a red arrow.



→ 1	gene_12 GeneMark.hmm 394_aa 10020 11204_insert_from_clone_F6L4	100.0%	161	2	240
2	PJT22797.1_hypothetical_protein_CUN56_04195_Chloroflexi_bacterium	61.9%			
3	KXR52969.1_putative_esterase_Chloroflexi_bacterium_OLB13	55.8%			
4	OQY84301.1_hypothetical_protein_B6D42_05600_Anaerolineae_bacterium_UTCFX5	54.1%			
5	KXR22648.1_putative_esterase_Chloroflexi_bacterium_OLB15	58.0%			
6	PJF41302.1_hypothetical_protein_CUN54_02095_Chloroflexi_bacterium	58.2%			
7	PJF32598.1_hypothetical_protein_CUN52_00885_Chloroflexi_bacterium	54.4%			
8	WP_078510950.1_enterochelin_esterase_Streptomyces_sp._TAA486	24.1%			
9	OOC56147.1_hypothetical_protein_NOSIN_21865_Nocardiopsis_sinuspersici	29.9%			
10	WP_077692584.1_enterochelin_esterase_Nocardiopsis_sinuspersici	21.7%			
11	WP_053619943.1_enterochelin_esterase_Nocardiopsis_sp._NRRL_B-16309_KOX0890.1_hypothetical_protein_ADL05_27690_Nocardiopsis_sp._NRRL_B-16309	29.2%			
12	ABJ82765.1_putative_esterase_Candidatus_Solibacter_usitatus_Ellin6076	12.4%			TSLDQGVRYTSTRIDANAFETTYDRGTERVSRTRFTAEPRDRMLTVLTGSKAVFEKLEGP.LLEAGKSVEQTFGAG
13	WP_082783663.1_DUF3327_domain-containing_protein_Candidatus_Solibacter_usitatus	12.6%			TSLDQGVRYTSTRIDANAFETTYDRGTERVSRTRFTAEPRDRMLTVLTGSKAVFEKLEGP.LLEAGKSVEQTFGAG
14	WP_007918782.1_enterochelin_esterase_Ktedonobacter_racemifer_EFM81418.1_putative_esterase_Ktedonobacter_racemifer_DSM_44963	29.4%			
15	WP_011955872.1_DUF3327_domain-containing_protein_Roseiflexus_sp._RS-1_ABQ89519.1_putative_esterase_Roseiflexus_sp._RS-1	19.6%			
16	OIP91490.1_hypothetical_protein_AUK47_22320_Deltaproteobacteria_bacterium_CG2_30_69_29_PJB34223.1_enterochelin_esterase_Deltaproteobacteria_bacterium_CG_4_9_14_3_um_filter_63_12	24.6%			
17	KPK61344.1_hypothetical_protein_AMJ59_02710_Gammaproteobacteria_bacterium_368_31	23.6%			
18	KYG09650.1_hypothetical_protein_BE21_16695_Sorangium_cellulosum	24.1%			
19	KYFS9881.1_hypothetical_protein_BE04_04740_Sorangium_cellulosum	29.0%			
20	WP_068613518.1_esterase_Woeseia_oceani_ANO50598.1_esterase_Woeseia_oceani	25.3%			
21	WP_080682583.1_DUF3327_domain-containing_protein_Sorangium_cellulosum	22.6%			
22	AGF40998.1_hypothetical_protein_SCE1572_44790_Sorangium_cellulosum_So0157-2	29.7%			
23	WP_060965334.1_esterase_family_protein_Pseudomonas_aeruginosa_KXE90326.1_enterochelin_esterase_Pseudomonas_aeruginosa	15.7%			MSAVSTSLLLFLMLLTPQAAGPPPLADGRLEIAAGESPQRY--
24	WP_073487538.1_enterochelin_esterase_Streptoalloteichus_hindustanus_SHG49133.1_enterochelin_esterase_Streptoalloteichus_hindustanus	29.2%			
25	WP_007910209.1_enterochelin_esterase_Ktedonobacter_racemifer_EFM8152.1_putative_esterase_Ktedonobacter_racemifer_DSM_44963	21.2%			
26	KPF80195.1_esterase_Roseiflexus_castenholzii	19.8%			
27	OLB44702.1_hypothetical_protein_AUH05_04345_Ktedonobacter_sp._13_2_20CM_53_11	21.7%			
28	WP_079868140.1_esterase_family_protein_partial_Pseudomonas_aeruginosa	22.0%			
	consensus/100%				
	consensus/90%				
	consensus/80%				
	consensus/70%				
→ 1	gene_12 GeneMark.hmm 394_aa 10020 11204_insert_from_clone_F6L4	100.0%	241	3	320
2	PJT22797.1_hypothetical_protein_CUN56_04195_Chloroflexi_bacterium	61.9%			
3	KXR52969.1_putative_esterase_Chloroflexi_bacterium_OLB13	55.8%			
4	OQY84301.1_hypothetical_protein_B6D42_05600_Anaerolineae_bacterium_UTCFX5	54.1%			
5	KXR22648.1_putative_esterase_Chloroflexi_bacterium_OLB15	58.0%			
6	PJF41302.1_hypothetical_protein_CUN54_02095_Chloroflexi_bacterium	58.2%			
7	PJF32598.1_hypothetical_protein_CUN52_00885_Chloroflexi_bacterium	54.4%			
8	WP_078510950.1_enterochelin_esterase_Streptomyces_sp._TAA486	24.1%			
9	OOC56147.1_hypothetical_protein_NOSIN_21865_Nocardiopsis_sinuspersici	29.9%			
10	WP_077692584.1_enterochelin_esterase_Nocardiopsis_sinuspersici	21.7%			
11	WP_053619943.1_enterochelin_esterase_Nocardiopsis_sp._NRRL_B-16309_KOX0890.1_hypothetical_protein_ADL05_27690_Nocardiopsis_sp._NRRL_B-16309	29.2%			MSSTTVPTTAKAGRTLRSTCTCSDRHLCRP
12	ABJ82765.1_putative_esterase_Candidatus_Solibacter_usitatus_Ellin6076	13.4%			AVVEYRIRLKAGEYCAGRIEPEKGGVNGAFYGPQGERLYNLGGPAAENRPFGLAEPVAGIYKALRSATPAKSFYTKVD
13	WP_082783663.1_DUF3327_domain-containing_protein_Candidatus_Solibacter_usitatus	12.6%			AVVEYRIRLKAGEYCAGRIEPEKGGVNGAFYGPQGERLYNLGGPAAENRPFGLAEPVAGIYKALRSATPAKSFYTKVD
14	WP_007918782.1_enterochelin_esterase_Ktedonobacter_racemifer_EFM81418.1_putative_esterase_Ktedonobacter_racemifer_DSM_44963	29.4%			
15	WP_011955872.1_DUF3327_domain-containing_protein_Roseiflexus_sp._RS-1_ABQ89519.1_putative_esterase_Roseiflexus_sp._RS-1	19.6%			
16	OIP91490.1_hypothetical_protein_AUK47_22320_Deltaproteobacteria_bacterium_CG2_30_69_29_PJB34223.1_enterochelin_esterase_Deltaproteobacteria_bacterium_CG_4_9_14_3_um_filter_63_12	24.6%			
17	KPK61344.1_hypothetical_protein_AMJ59_02710_Gammaproteobacteria_bacterium_368_31	23.6%			
18	KYG09650.1_hypothetical_protein_BE21_16695_Sorangium_cellulosum	24.1%			
19	KYFS9881.1_hypothetical_protein_BE04_04740_Sorangium_cellulosum	29.0%			
20	WP_068613518.1_esterase_Woeseia_oceani_ANO50598.1_esterase_Woeseia_oceani	25.3%			
21	WP_080682583.1_DUF3327_domain-containing_protein_Sorangium_cellulosum	22.6%			
22	AGF40998.1_hypothetical_protein_SCE1572_44790_Sorangium_cellulosum_So0157-2	29.7%			
23	WP_060965334.1_esterase_family_protein_Pseudomonas_aeruginosa_KXE90326.1_enterochelin_esterase_Pseudomonas_aeruginosa	15.7%			LDLAPGELQLLDLEVDGRVRLRATTFDQKALR-----ATELAGRQSLGLQAGERQGLRLTFSADEDTA
24	WP_073487538.1_enterochelin_esterase_Streptoalloteichus_hindustanus_SHG49133.1_enterochelin_esterase_Streptoalloteichus_hindustanus	29.2%			
25	WP_007910209.1_enterochelin_esterase_Ktedonobacter_racemifer_EFM8152.1_putative_esterase_Ktedonobacter_racemifer_DSM_44963	21.2%			
26	KPF80195.1_esterase_Roseiflexus_castenholzii	19.8%			
27	OLB44702.1_hypothetical_protein_AUH05_04345_Ktedonobacter_sp._13_2_20CM_53_11	21.7%			
28	WP_079868140.1_esterase_family_protein_partial_Pseudomonas_aeruginosa	22.0%			
	consensus/100%				
	consensus/90%				
	consensus/80%				
	consensus/70%				

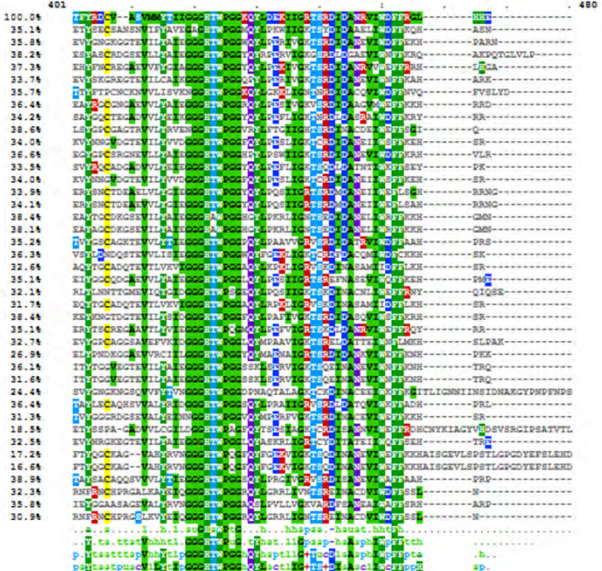
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3 OG233592.1_hypothetical_protein_A3154_03130 Omnitopoda_WOR_1_bacterium_R1CF3PLOWO2_02_FUTL_50_19
4 OG233592.1_hypothetical_protein_A3154_04010 Candidatus Hypotrichobacteria_bacterium_R83_16_44_11
5 OG233592.1_hypothetical_protein_A3154_04020 Thermomococcus radiotolerans_XJ334325.1_hypothetical_protein_A3Y94_03255 Thermomococcus radiotolerans
6 OJ197363.1_hypothetical_protein_A2V65_03603 Candidatus Desantiibacteria_bacterium_R83_02_38_46
7 OJ197460.1_hypothetical_protein_A2W81_05075 Bacteroidetes_bacterium_OJ272_38_235_OJ779197.1_hypothetical_protein_A2281_10345 Bacteroidetes
8 OG500575.1_hypothetical_protein_A2V78_11250 Nitrospirae_bacterium_R83_16_44_26
9 WP_23824279.1_hypothetical_protein_Delta_Proteobacteria_PSC02_2495
10 OJ065712.1_hypothetical_protein_A2V16_13810 Spirochaetes_bacterium_R83_13_31_14
11 OJ170152.1_hypothetical_protein_A2V71_02805 Bacteroidetes_bacterium_R83_13_42_15
12 OJ170152.1_hypothetical_protein_A2V78_09945 Deltaaproteobacteria_bacterium_R83_16_45_23
13 OJ480885.1_hypothetical_protein_Delta_Proteobacteria_PSC02_2342
14 OJ177857.1_hypothetical_protein_A2V46_04800 Bacteroidetes_bacterium_R83_19F7_COMBO_42_7
15 WP_088861017.1_phospholipase_Thermomococcus radiotolerans_AJ313902.1_hypothetical_protein_AJ110_01660 Thermomococcus radiotolerans
16 WP_088165030.1_phospholipase_Thermomococcus sp_3_4_A3A77645.1_phospholipase_Thermomococcus sp_3_4
17 OJ177857.1_hypothetical_protein_A2V20_12475 Betaproteobacteria_bacterium_R83_16_48_11
18 OJ197460.1_hypothetical_protein_A2V44_10360 Betaproteobacteria_bacterium_R83_19F7_COMBO_88_11
19 KMT71920.1_esterase_polyhydroxybutyrate_depolymerase_Arammatonadetes_bacterium_CPF1-3
20 WP_0931237.1_phospholipase_bacterium_R83-PSC02-bacteria-21
21 WP_09313371.1_esterase_Acidiphilobacteria_COMOE_5410
22 OJ293287.1_hypothetical_protein_A2V75_05310 Chloroflexi_bacterium_R83_13_30_10
23 WP_000840001.1_phospholipase_Acidiphilobacterium_boonae_IDJ33505.1_phospholipase/carboxylesterase_family_Acidiphilopfundum_boonae_T469_A0001
24 WP_012164715.1_esterase_Acidiphilobacteria_marina_OJ197397.1_esterase_putative_Acidiphilobacteria_marina_R1HC11017
25 OJ197397.1_hypothetical_protein_A2V67_03603 Candidatus Desantiibacteria_bacterium_R83_13_44_24
26 KPF13252.1_hypothetical_protein_A2V67_17350 Betaproteobacteria_bacterium_R83_16_48_11
27 OJ293280.1_hypothetical_protein_A2V78_00935 Elusimicrobia_bacterium_R1FOKYX2_FUTL_62_6
28 OJ292137.1_hypothetical_protein_A2V98_13175 Spirochaetes_bacterium_OJ71_41_1
29 OJ161805.1_hypothetical_protein_VJ13_04100 Mesotoga sp. N07_pap.5.1
30 WP_099771784.1_phospholipase_Mesotoga sp. N07_pap.5.3
31 OJ197460.1_hypothetical_protein_A2V21_09955 Candidatus Fiskebäcksbacteria_bacterium_R1FOXTA2_FUTL_40_8
32 OJ293280.1_hypothetical_protein_A2V75_05310 Armatimonadetes_bacterium_R83_13_30CM_10
33 OJ500575.1_hypothetical_protein_A2V44_15455 Candidatus Fiskebäcksbacteria_bacterium_R1FOXTA2_FUTL_43_8
34 OJ294968.1_hypothetical_protein_A2V34_00165 Elusimicrobia_bacterium_R1FOKYX2_FUTL_50_12
35 KFY35392.1_hypothetical_protein_AYX15_02040 Thermomococcus radiotolerans_OJ70-1
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37 OJ197397.1_hypothetical_protein_A3154_03130 Candidatus Desantiibacteria_bacterium_R1CF3PLOWO2_01_FUTL_43_24
38 OJ174798.1_hypothetical_protein_A2V58_04025 Armatimonadetes_bacterium_R83_19F7_COMBO_69_19
39 WP_251891812.1_MUT278P8C125_hypothetical_protein_Avenibacteres
40 KPF17458.1_hypothetical_protein_A2V42_17150 Methylthiobacillus sp. R83
41 WP_093287809.1_hypothetical_protein_Avenibacteres_algicola
42 consensus/1004
43 consensus/806
44 consensus/808
45 consensus/708

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 4 GGI44482.1_hypothetical_protein_A2V62_02040_Candidatus_Fischerbacteria_bacterium_RBG_16_64_11
 5 WP_024803760.1_hypothetical_protein_Thermococcus_californiensis_KUN34526.1_hypothetical_protein_APY94_01925_Thermococcus_californiensis
 6 OI897363.1_hypothetical_protein_A2V66_03605_Candidatus_Desantisbacteria_bacterium_CGI_02_38_46
 7 OI796160.1_hypothetical_protein_A2V91_05075_Bacteroidetes_bacterium_GW2_38_335_OFY79797.1_hypothetical_protein_A2281_10345_Bacteroidetes_bacterium_RIFOXYA12_FULL_38_20
 8 GGG60575.1_hypothetical_protein_A2V91_10230_Mitosepias_bacterium_RBG_16_64_22
 9 WP_024803760.1_hypothetical_protein_delta_protobacterium_PSC02_5451
 10 OMD65713.1_hypothetical_protein_A2V76_13850_Spirochaetes_bacterium_RBG_13_51_14
 11 OFY71052.1_hypothetical_protein_A2V71_02805_Bacteroidetes_bacterium_RBG_13_42_18
 12 GGF77132.1_hypothetical_protein_A2V66_09965_Deltaaproteobacteria_bacterium_RBG_16_49_23
 13 WP_024803760.1_hypothetical_protein_delta_protobacterium_PSC02_5451
 14 OFY77857.1_hypothetical_protein_A2V46_04900_Bacteroidetes_bacterium_RBG_18FT_COMBO_42_7
 15 WP_024803760.1_phospholipase_Thermococcus_radiotolerans_A3713902.1_hypothetical_protein_A3710_01660_Thermococcus_radiotolerans
 16 WP_024803760.1_phospholipase_Thermococcus_sp._5-4_A3A77645.1_phospholipase_Thermococcus_sp._5-4
 17 OFY78005.1_hypothetical_protein_A2201_10470_Betaproteobacteria_bacterium_RBG_16_58_11
 18 OFY87407.1_hypothetical_protein_A2244_10360_Betaproteobacteria_bacterium_RBG_18FT_COMBO_58_11
 19 KX277820.1_esterase_polyhydroxybutyrate_depolymerase_Amatmonnabacteria_bacterium_CSP1-3
 20 KXP21237.1_phospholipase_Bacteroidetes_bacterium_RBG-Bacteroidetes-21
 21 WP_024803760.1_esterase_Acaryochloris_sp._CCEM_5410
 22 GGG33592.1_hypothetical_protein_A2V62_02040_Candidatus_Fischerbacteria_bacterium_RBG_13_37_8
 23 WP_024803760.1_phospholipase_Aciduliprofundum_boonae_EDV35805.1_phospholipase/carboxylesterase_family_Aciduliprofundum_boonae_T469_ADO09207.1_phospholipase/Carboxylesterase_Aciduliprofundum_boonae_T469
 24 WP_012164719.1_esterase_Acaryochloris_marina_ABN29397.1_esterase_putative_Acaryochloris_marina_MBC11017
 25 OFY77597.1_hypothetical_protein_A2V69_01170_Bacteroidetes_bacterium_RBG_13_44_24
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 28 OMD62137.1_hypothetical_protein_A2V96_13175_Spirochaetes_bacterium_GW1_41_5
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 38 OFY4758.1_hypothetical_protein_A2V59_02425_Amatmonnabacteria_bacterium_RBG_18FT_COMBO_69_19
 39 WP_024803760.1_MULTISPECIES_hypothetical_protein_Avenibacter
 40 KX217145.1_hypothetical_protein_A2V42_17130_Methylobium_sp._M2
 41 WP_024803760.1_hypothetical_protein_Avenibacter_algicola
 consensus/100
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 consensus/70

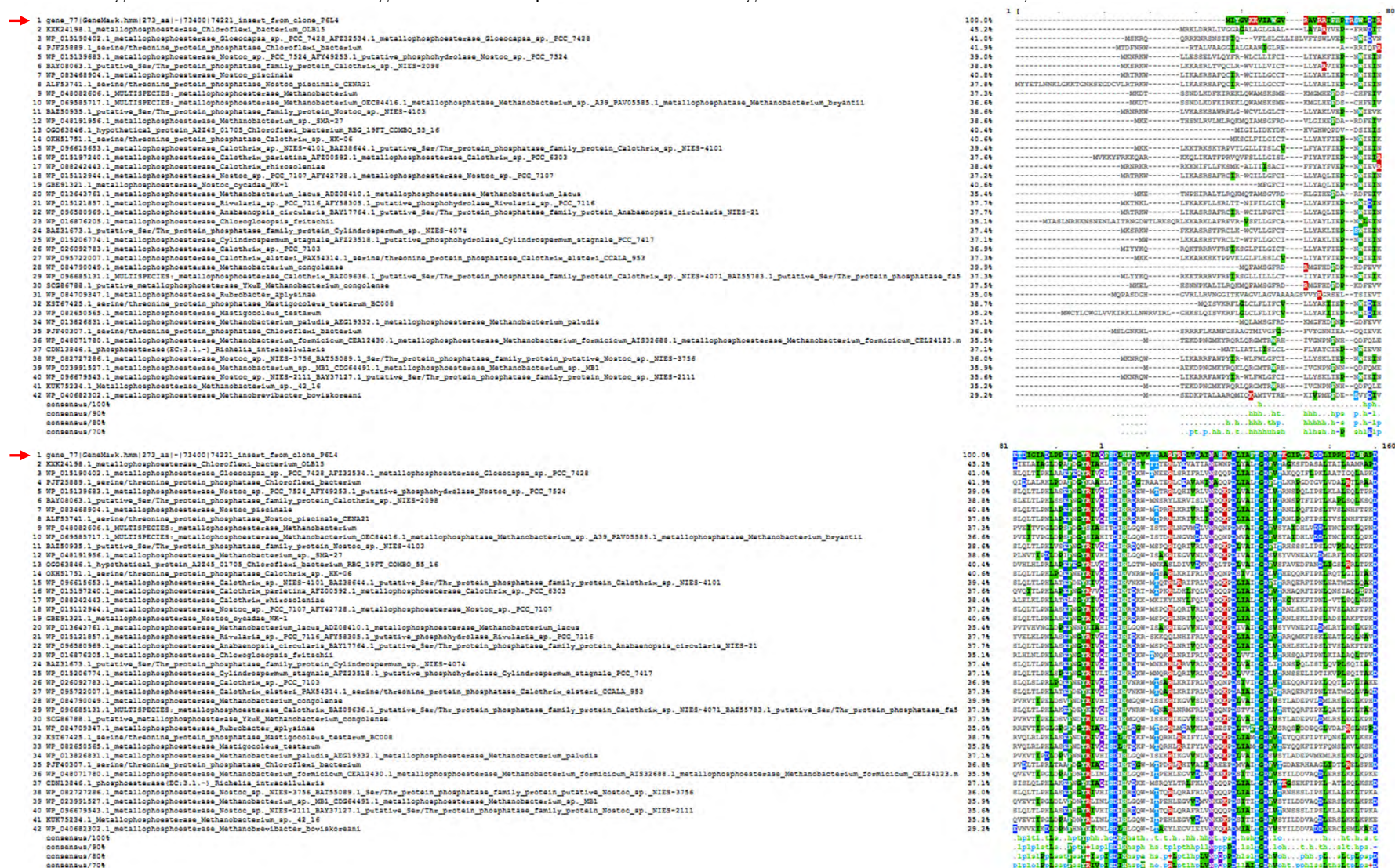


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 5 WP_024803760.1_hypothetical_protein_Thermococcus_californiensis_KUN34526.1_hypothetical_protein_APY94_01925_Thermococcus_californiensis
 6 OI897363.1_hypothetical_protein_A2V66_03605_Candidatus_Desantisbacteria_bacterium_CGI_02_38_46
 7 OI796160.1_hypothetical_protein_A2V91_05075_Bacteroidetes_bacterium_GW2_38_335_OFY79797.1_hypothetical_protein_A2281_10345_Bacteroidetes_bacterium_RIFOXYA12_FULL_38_20
 8 GGG60575.1_hypothetical_protein_A2V91_10230_Mitosepias_bacterium_RBG_16_64_22
 9 WP_024803760.1_hypothetical_protein_delta_protobacterium_PSC02_5451
 10 OMD65713.1_hypothetical_protein_A2V76_13850_Spirochaetes_bacterium_RBG_13_51_14
 11 OFY71052.1_hypothetical_protein_A2V71_02805_Bacteroidetes_bacterium_RBG_13_42_18
 12 GGF77132.1_hypothetical_protein_A2V66_09965_Deltaaproteobacteria_bacterium_RBG_16_49_23
 13 WP_024803760.1_hypothetical_protein_delta_protobacterium_PSC02_5451
 14 OFY77857.1_hypothetical_protein_A2V46_04900_Bacteroidetes_bacterium_RBG_18FT_COMBO_42_7
 15 WP_024803760.1_phospholipase_Thermococcus_radiotolerans_A3713902.1_hypothetical_protein_A3710_01660_Thermococcus_radiotolerans
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 17 OFY78005.1_hypothetical_protein_A2201_10470_Betaproteobacteria_bacterium_RBG_16_58_11
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 consensus/100
 consensus/50
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 consensus/70



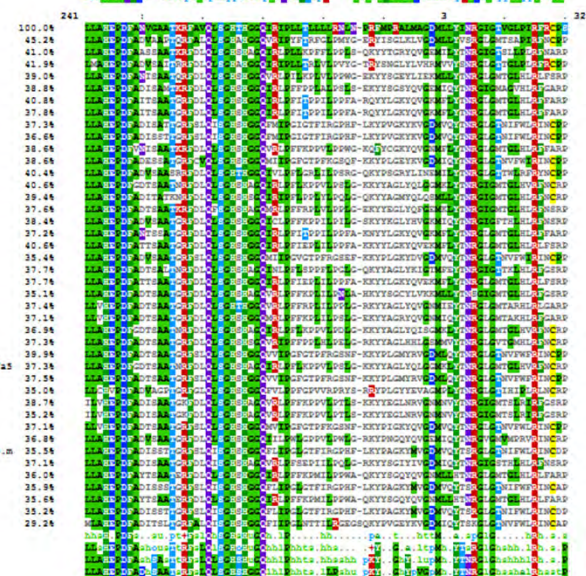
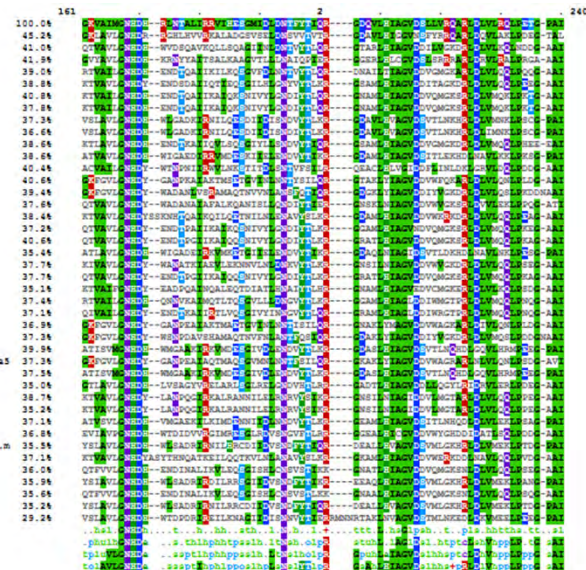
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17	OFY71055.1_hypothetical_protein_A2E01_10470_Betaproteobacteria_bacterium_RBG_16_58_11	38.46			
18	OFZ97407.1_hypothetical_protein_A2I44_10360_Betaproteobacteria_bacterium_RBG_19FT_COMBO_58_11	38.16			
19	KMT77820.1_esterase_polyhydroxybutyrate_depolymerase_Amatimonadetes_bacterium_CSP1-3	35.26			
20	FKP21237.1_phospholipase_Bacteroidetes_bacterium_RGW-Bacteroidetes-21	36.36			
21	WP_029319312.1_esterase_Acaryochloris_sp._COMEX_5410	32.66			
22	OGN93287.1_hypothetical_protein_A2I75_03310_Chloroflexi_bacterium_RBG_13_50_10	35.16			
23	WP_008084028.1_phospholipase_Aciduliprofundum_boonae_EDY35805.1_phospholipase/Caeobacteriaceae_Aciduliprofundum_boonae_T469_A0009207.1_phospholipase/Caeobacteriaceae_Aciduliprofundum_boonae_T469	32.16			
24	WP_012164719.1_esterase_Acaryochloris_marina_A3W29397.1_esterase_putative_Acaryochloris_marina_M8IC11017	31.76			
25	OFY37597.1_hypothetical_protein_A2I69_01170_Bacteroidetes_bacterium_RBG_13_44_24	38.46			
26	KPK13523.1_hypothetical_protein_A3T67_11350_Betaproteobacteria_bacterium_RBG_41	35.16			
27	OGS51054.1_hypothetical_protein_A3T78_00935_Elusimicrobia_bacterium_RIFOKYB2_FULL_62_6	32.76			
28	ORW62137.1_hypothetical_protein_A2I96_13175_Spirochaetes_bacterium_GWF1_41_5	26.96			
29	FIJ61805.1_hypothetical_protein_V513_04100_Mesotoga_sp._R07.pap.5.3	36.16			
30	WP_089771798.1_phospholipase_Mesotoga_sp._R07.pap.5.3	31.66			
31	OGF46285.1_hypothetical_protein_A2I231_08565_Candidatus_Firestonebacteria_bacterium_RIFOKYA2_FULL_40_8	24.46			
32	OGK33592.1_hypothetical_protein_A2143_01350_Omnitrophica_MOR_2_bacterium_RIFCSPLOW02_01_FULL_50_19	35.08			
33	OGF50273.1_hypothetical_protein_A2Q44_05455_Candidatus_Firestonebacteria_bacterium_GWA2_43_8	31.36			
34	OGS24968.1_hypothetical_protein_A2314_00165_Elusimicrobia_bacterium_RIFOKYB2_FULL_50_12	18.56			
35	KPK33592.1_hypothetical_protein_AYK19_02040_Thionarchaea_mechanoe_DG-70-1	32.56			
36	OGF45857.1_hypothetical_protein_A2I69_01140_partial_Candidatus_Glovannobacteria_bacterium_RIFCSPH2G02_01_FULL_45_24	17.26			
37	OGF71057.1_hypothetical_protein_A3B19_03735_Candidatus_Glovannobacteria_bacterium_RIFCSPH2G02_01_FULL_46_32	36.46			
38	OFK14798.1_hypothetical_protein_A2V59_00425_Amatimonadetes_bacterium_RBG_19FT_COMBO_69_19	38.96			
39	WP_051891812.1_MULTISPECIES_hypothetical_protein_Arenibacter	32.36			
40	KNE31745.1_hypothetical_protein_AD742_17130_Methylobium_sp._M2G	35.86			
41	WP_059378198.1_hypothetical_protein_Arenibacter_ajigicola	30.96			
	consensus/100%				
	consensus/90%				
	consensus/80%				
	consensus/70%				
→ 1	gene_35 GeneMark.hmm 278_aa -135192 36028_insert_from_clone_PEL4	100.00	561	6	640
2	OGF45857.1_hypothetical_protein_A21491_02040_Candidatus_Flacheobacteria_bacterium_RBG_13_37_8	35.16			
3	OGK33592.1_hypothetical_protein_A2143_01350_Omnitrophica_MOR_2_bacterium_RIFCSPLOW02_01_FULL_50_19	35.08			
4	OG244382.1_hypothetical_protein_A2150_06050_Candidatus_Mycotheobacteria_bacterium_RBG_16_64_11	38.26			
5	WP_058937961.1_hypothetical_protein_Thermococcus_celerisreascens_KUN34526.1_hypothetical_protein_APY94_01525_Thermococcus_celerisreascens	37.36			
6	OGN97363.1_hypothetical_protein_AU766_03605_Candidatus_Dasantisbacteria_bacterium_CGI_02_38_46	33.76			
7	OFY34160.1_hypothetical_protein_A2991_05075_Bacteroidetes_bacterium_GWF2_38_335_OFY79797.1_hypothetical_protein_A2281_10345_Bacteroidetes_bacterium_RIFOKYA12_FULL_38_20	35.76			
8	OGW60075.1_hypothetical_protein_A2983_12120_Mitocapsella_bacterium_RBG_16_64_22	36.48			
9	WP_038340759.1_hypothetical_protein_delta_protobacterium_PSCOC_5451	34.26			
10	ORW65713.1_hypothetical_protein_A2176_13850_Spirochaetes_bacterium_RBG_13_81_14	38.66			
11	OFY71052.1_hypothetical_protein_A2Y71_02805_Bacteroidetes_bacterium_RBG_13_42_19	34.06			
12	OGF71132.1_hypothetical_protein_A2V86_09965_Deltaaproteobacteria_bacterium_RBG_16_49_23	36.66			
13	WP_048908850.1_hypothetical_protein_delta_protobacterium_PSCOC_5342	33.56			
14	OFY77857.1_hypothetical_protein_A2V46_04900_Bacteroidetes_bacterium_RBG_19FT_COMBO_42_7	34.06			
15	WP_088666107.1_phospholipase_Thermococcus_radiotolerans_A3713902.1_hypothetical_protein_A3L10_01660_Thermococcus_radiotolerans	33.96			
16	WP_088180530.1_phospholipase_Thermococcus_sp._5-4_A3A77645.1_phospholipase_Thermococcus_sp._5-4	34.16			
17	OFY71055.1_hypothetical_protein_A2E01_10470_Betaproteobacteria_bacterium_RBG_16_58_11	38.46			
18	OFZ97407.1_hypothetical_protein_A2I44_10360_Betaproteobacteria_bacterium_RBG_19FT_COMBO_58_11	38.16			
19	KMT77820.1_esterase_polyhydroxybutyrate_depolymerase_Amatimonadetes_bacterium_CSP1-3	35.26			
20	FKP21237.1_phospholipase_Bacteroidetes_bacterium_RGW-Bacteroidetes-21	36.36			
21	WP_029319312.1_esterase_Acaryochloris_sp._COMEX_5410	32.66			
22	OGN93287.1_hypothetical_protein_A2I75_03310_Chloroflexi_bacterium_RBG_13_50_10	35.16			
23	WP_008084028.1_phospholipase_Aciduliprofundum_boonae_EDY35805.1_phospholipase/Caeobacteriaceae_Aciduliprofundum_boonae_T469_A0009207.1_phospholipase/Caeobacteriaceae_Aciduliprofundum_boonae_T469	32.16			
24	WP_012164719.1_esterase_Acaryochloris_marina_A3W29397.1_esterase_putative_Acaryochloris_marina_M8IC11017	31.76			
25	OFY37597.1_hypothetical_protein_A2I69_01170_Bacteroidetes_bacterium_RBG_13_44_24	38.46			
26	KPK13523.1_hypothetical_protein_A3T67_11350_Betaproteobacteria_bacterium_RBG_41	35.16			
27	OGS51054.1_hypothetical_protein_A3T78_00935_Elusimicrobia_bacterium_RIFOKYB2_FULL_62_6	32.76			
28	ORW62137.1_hypothetical_protein_A2I96_13175_Spirochaetes_bacterium_GWF1_41_5	26.96			
29	FIJ61805.1_hypothetical_protein_V513_04100_Mesotoga_sp._R07.pap.5.3	36.16			
30	WP_089771798.1_phospholipase_Mesotoga_sp._R07.pap.5.3	31.66			
31	OGF46285.1_hypothetical_protein_A2I231_08565_Candidatus_Firestonebacteria_bacterium_RIFOKYA2_FULL_40_8	24.46			
32	OGK33592.1_hypothetical_protein_A2143_01350_Omnitrophica_MOR_2_bacterium_RIFCSPLOW02_01_FULL_50_19	35.08			
33	OGF50273.1_hypothetical_protein_A2Q44_05455_Candidatus_Firestonebacteria_bacterium_GWA2_43_8	31.36			
34	OGS24968.1_hypothetical_protein_A2314_00165_Elusimicrobia_bacterium_RIFOKYB2_FULL_50_12	18.56			
35	KPK33592.1_hypothetical_protein_AYK19_02040_Thionarchaea_mechanoe_DG-70-1	32.56			
36	OGF45857.1_hypothetical_protein_A2I69_01140_partial_Candidatus_Glovannobacteria_bacterium_RIFCSPH2G02_01_FULL_45_24	17.26			
37	OGF71057.1_hypothetical_protein_A3B19_03735_Candidatus_Glovannobacteria_bacterium_RIFCSPH2G02_01_FULL_46_32	36.46			
38	OFK14798.1_hypothetical_protein_A2V59_00425_Amatimonadetes_bacterium_RBG_19FT_COMBO_69_19	38.96			
39	WP_051891812.1_MULTISPECIES_hypothetical_protein_Arenibacter	32.36			
40	KNE31745.1_hypothetical_protein_AD742_17130_Methylobium_sp._M2G	35.86			
41	WP_059378198.1_hypothetical_protein_Arenibacter_ajigicola	30.96			
	consensus/100%				
	consensus/90%				
	consensus/80%				
	consensus/70%				

Figure S5. Multiple alignment of a putative metallophosphoesterase (clone P6L4; ORF35) compared to esterase amino acid sequences obtained from GenBank. The multiple alignment was conducted using MUSCLE and visualized using MVIEW. The sequence derived from metagenomic clone P6L4 is marked by a red arrow.



→ 1 gene_77(GeneMark.hmm/273_aa)~(73400/74221_insert_from_clone_F614
2 K0XK24193.1_metallophosphatase_Chalcofilax_bacterium_C01B15
3 WP_015190402.1_metallophosphatase_Gloeocephala_sp._PCC_7428
4 P0721888.1_serine/threonine_protein_phosphatase_Chalcofilax_bacterium
5 WP_015139683.1_metallophosphatase_Mostoc_sp._PCC_7524_AFY49253.1_putative_phosphohydrolase_Mostoc_sp._PCC_7524
6 BAY00603.1_putative_ser/Thr_protein_phosphatase_family_protein_Calothrix_sp._NIES-2098
7 WP_08348904.1_metallophosphatase_Mostoc_gischnalis
8 ALP57471.1_serine/threonine_protein_phosphatase_Mostoc_gischnalis_CEN421
9 WP_048082626.1_MULTISPECIES_metallophosphatase_Methanobacterium
10 WP_069585717.1_MULTISPECIES_metallophosphatase_Methanobacterium_OEC4416.1_metallophosphatase_Methanobacterium_bryantii
11 BA550935.1_putative_ser/Thr_protein_phosphatase_family_protein_Mostoc_sp._NIES-4103
12 WP_048191856.1_metallophosphatase_Methanobacterium_sp._R80-27
13 OQ06846.1_hypothetical_protein_A2245_01705_Chloroflexus_bacterium_RBG_19PT_COMBO_55_16
14 OXK51751.1_serine/threonine_protein_phosphatase_Calothrix_sp._RK-06
15 WP_066156593.1_metallophosphatase_Calothrix_sp._NIES-4101_BA238644.1_putative_ser/Thr_protein_phosphatase_family_protein_Calothrix_sp._NIES-4101
16 WP_015197240.1_metallophosphatase_Calothrix_piscicaria_AFY00592.1_metallophosphatase_Calothrix_sp._PCC_6303
17 WP_088242443.1_metallophosphatase_Calothrix_piscicaria
18 WP_01512944.1_metallophosphatase_Mostoc_sp._PCC_7107_AFY42728.1_metallophosphatase_Mostoc_sp._PCC_7107
19 CB891321.1_metallophosphatase_Mostoc_cycladae_RK-1
20 WP_013643761.1_metallophosphatase_Methanobacterium_lacus_A2020410.1_metallophosphatase_Methanobacterium_lacus
21 WP_015121857.1_metallophosphatase_Rivularia_sp._PCC_7116_AFY38305.1_putative_phosphohydrolase_Rivularia_sp._PCC_7116
22 WP_069580869.1_metallophosphatase_Anabaenopsis_circulans_BAY17764.1_putative_ser/Thr_protein_phosphatase_family_protein_Anabaenopsis_circulans_NIES-21
23 WP_016876205.1_metallophosphatase_Chloroclospira_fritschii
24 BA231670.1_putative_ser/Thr_protein_phosphatase_family_protein_Cylindrocapsa_sp._NIES-4074
25 WP_015206774.1_metallophosphatase_Cylindrocapsa_stagnalis_AFY223518.1_putative_phosphohydrolase_Cylindrocapsa_stagnalis_PCC_7417
26 WP_026092783.1_metallophosphatase_Calothrix_sp._PCC_7103
27 WP_095722007.1_metallophosphatase_Calothrix_elstera_PAK54314.1_serine/threonine_protein_phosphatase_Calothrix_elstera_CCALA_953
28 WP_04790049.1_metallophosphatase_Methanobacterium_congolense
29 WP_06685131.1_MULTISPECIES_metallophosphatase_Calothrix_BA238636.1_putative_ser/Thr_protein_phosphatase_family_protein_Calothrix_sp._NIES-4071_BA238783.1_putative_ser/Thr_protein_phosphatase_fa5
30 SC026788.1_putative_metallophosphatase_Vulva_Methanobacterium_congolense
31 WP_084709347.1_metallophosphatase_Rubrobacter_aplysiaceae
32 KX57425.1_serine/threonine_protein_phosphatase_Mastigococcus_testaceum_BCC06
33 WP_062650565.1_metallophosphatase_Mastigococcus_testaceum
34 WP_013826831.1_metallophosphatase_Methanobacterium_paludis_AFG19332.1_metallophosphatase_Methanobacterium_paludis
35 PTF40307.1_serine/threonine_protein_phosphatase_Chalcofilax_bacterium
36 WP_048071705.1_metallophosphatase_Methanobacterium_fomicium_CEA12430.1_metallophosphatase_Methanobacterium_fomicium_A1932688.1_metallophosphatase_Methanobacterium_fomicium_CEA124123.m
37 CDM13846.1_phosphatase(EC3.1.1)_Richelia_intercellulalis
38 WP_08272286.1_metallophosphatase_Mostoc_sp._NIES-3756_BA235089.1_ser/Thr_protein_phosphatase_family_protein_putative_Mostoc_sp._NIES-3756
39 WP_023991927.1_metallophosphatase_Methanobacterium_sp._MB1_CD064491.1_metallophosphatase_Methanobacterium_sp._MB1
40 WP_066879543.1_metallophosphatase_Mostoc_sp._NIES-2111_BA237117.1_putative_ser/Thr_protein_phosphatase_family_protein_Mostoc_sp._NIES-2111
41 KX573234.1_metallophosphatase_Methanobacterium_sp._42_16
42 WP_040682032.1_metallophosphatase_Methanobrevibacter_dovskorensis
consensus/100%
consensus/90%
consensus/ECN
consensus/70%

→ 1 gene_77(GeneMark.hmm/273_aa)~(73400/74221_insert_from_clone_F614
2 K0XK24193.1_metallophosphatase_Chalcofilax_bacterium_C01B15
3 WP_015190402.1_metallophosphatase_Gloeocephala_sp._PCC_7428
4 P0721888.1_serine/threonine_protein_phosphatase_Chalcofilax_bacterium
5 WP_015139683.1_metallophosphatase_Mostoc_sp._PCC_7524_AFY49253.1_putative_phosphohydrolase_Mostoc_sp._PCC_7524
6 BAY00603.1_putative_ser/Thr_protein_phosphatase_family_protein_Calothrix_sp._NIES-2098
7 WP_08348904.1_metallophosphatase_Mostoc_gischnalis
8 ALP57471.1_serine/threonine_protein_phosphatase_Mostoc_gischnalis_CEN421
9 WP_048082626.1_MULTISPECIES_metallophosphatase_Methanobacterium
10 WP_069585717.1_MULTISPECIES_metallophosphatase_Methanobacterium_OEC4416.1_metallophosphatase_Methanobacterium_bryantii
11 BA550935.1_putative_ser/Thr_protein_phosphatase_family_protein_Mostoc_sp._NIES-4103
12 WP_048191856.1_metallophosphatase_Methanobacterium_sp._R80-27
13 OQ06846.1_hypothetical_protein_A2245_01705_Chloroflexus_bacterium_RBG_19PT_COMBO_55_16
14 OXK51751.1_serine/threonine_protein_phosphatase_Calothrix_sp._RK-06
15 WP_066156593.1_metallophosphatase_Calothrix_sp._NIES-4101_BA238644.1_putative_ser/Thr_protein_phosphatase_family_protein_Calothrix_sp._NIES-4101
16 WP_015197240.1_metallophosphatase_Calothrix_piscicaria_AFY00592.1_metallophosphatase_Calothrix_sp._PCC_6303
17 WP_088242443.1_metallophosphatase_Calothrix_piscicaria
18 WP_01512944.1_metallophosphatase_Mostoc_sp._PCC_7107_AFY42728.1_metallophosphatase_Mostoc_sp._PCC_7107
19 CB891321.1_metallophosphatase_Mostoc_cycladae_RK-1
20 WP_013643761.1_metallophosphatase_Methanobacterium_lacus_A2020410.1_metallophosphatase_Methanobacterium_lacus
21 WP_015121857.1_metallophosphatase_Rivularia_sp._PCC_7116_AFY38305.1_putative_phosphohydrolase_Rivularia_sp._PCC_7116
22 WP_069580869.1_metallophosphatase_Anabaenopsis_circulans_BAY17764.1_putative_ser/Thr_protein_phosphatase_family_protein_Anabaenopsis_circulans_NIES-21
23 WP_016876205.1_metallophosphatase_Chloroclospira_fritschii
24 BA231670.1_putative_ser/Thr_protein_phosphatase_family_protein_Cylindrocapsa_sp._NIES-4074
25 WP_015206774.1_metallophosphatase_Cylindrocapsa_stagnalis_AFY223518.1_putative_phosphohydrolase_Cylindrocapsa_stagnalis_PCC_7417
26 WP_026092783.1_metallophosphatase_Calothrix_sp._PCC_7103
27 WP_095722007.1_metallophosphatase_Calothrix_elstera_PAK54314.1_serine/threonine_protein_phosphatase_Calothrix_elstera_CCALA_953
28 WP_04790049.1_metallophosphatase_Methanobacterium_congolense
29 WP_06685131.1_MULTISPECIES_metallophosphatase_Calothrix_BA238636.1_putative_ser/Thr_protein_phosphatase_family_protein_Calothrix_sp._NIES-4071_BA238783.1_putative_ser/Thr_protein_phosphatase_fa5
30 SC026788.1_putative_metallophosphatase_Vulva_Methanobacterium_congolense
31 WP_084709347.1_metallophosphatase_Rubrobacter_aplysiaceae
32 KX57425.1_serine/threonine_protein_phosphatase_Mastigococcus_testaceum_BCC06
33 WP_062650565.1_metallophosphatase_Mastigococcus_testaceum
34 WP_013826831.1_metallophosphatase_Methanobacterium_paludis_AFG19332.1_metallophosphatase_Methanobacterium_paludis
35 PTF40307.1_serine/threonine_protein_phosphatase_Chalcofilax_bacterium
36 WP_048071705.1_metallophosphatase_Methanobacterium_fomicium_CEA12430.1_metallophosphatase_Methanobacterium_fomicium_A1932688.1_metallophosphatase_Methanobacterium_fomicium_CEA124123.m
37 CDM13846.1_phosphatase(EC3.1.1)_Richelia_intercellulalis
38 WP_08272286.1_metallophosphatase_Mostoc_sp._NIES-3756_BA235089.1_ser/Thr_protein_phosphatase_family_protein_putative_Mostoc_sp._NIES-3756
39 WP_023991927.1_metallophosphatase_Methanobacterium_sp._MB1_CD064491.1_metallophosphatase_Methanobacterium_sp._MB1
40 WP_066879543.1_metallophosphatase_Mostoc_sp._NIES-2111_BA237117.1_putative_ser/Thr_protein_phosphatase_family_protein_Mostoc_sp._NIES-2111
41 KX573234.1_metallophosphatase_Methanobacterium_sp._42_16
42 WP_040682032.1_metallophosphatase_Methanobrevibacter_dovskorensis
consensus/100%
consensus/90%
consensus/ECN
consensus/70%



1 gene_771|GeneMark.hmm|273_aa|~173400|74921_inser_from_clone_P8L4
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 3 WP_015190402.1_metallophosphoesterase_Gloeoscapsa_sp._PCC_7428_AFE232534.1_metallophosphoesterase_Gloeoscapsa_sp._PCC_7428
 4 P3F25889.1_serine/threonine_protein_phosphatase_Chloroflexi_bacterium
 5 WP_015139883.1_metallophosphoesterase_Mostoc_sp._PCC_7524_AFY49253.1_putative_phosphohydrolase_Mostoc_sp._PCC_7524
 6 BAY08063.1_putative_Ser/Thr_protein_phosphatase_family_protein_Calothrix_sp._NIES-7098
 7 WP_083468904.1_metallophosphoesterase_Mostoc_piscinale
 8 ALF53741.1_serine/threonine_protein_phosphatase_Mostoc_piscinale_CEN421
 9 WP_048082606.1_MULTISPECIES_metallophosphoesterase_Methanobacterium
 10 WP_049585717.1_MULTISPECIES_metallophosphoesterase_Methanobacterium_C8C84416.1_metallophosphatase_Methanobacterium_sp._A39_BAV05585.1_metallophosphatase_Methanobacterium_bryantii
 11 BAI50935.1_putative_Ser/Thr_protein_phosphatase_family_protein_Mostoc_sp._NIES-4103
 12 WP_048191956.1_metallophosphoesterase_Methanobacterium_sp._SMA-27
 13 OGG63846.1_hypothetical_protein_A2145_01705_Chloroflexi_bacterium_XBG_19PT_COMBO_55_16
 14 OGG51751.1_serine/threonine_protein_phosphatase_Calothrix_sp._NR-56
 15 WP_084616653.1_metallophosphoesterase_Calothrix_sp._NIES-4101_BAI38644.1_putative_Ser/Thr_protein_phosphatase_family_protein_Calothrix_sp._NIES-4101
 16 WP_015197240.1_metallophosphoesterase_Calothrix_pavlovii_AFI00592.1_metallophosphoesterase_Calothrix_sp._PCC_6303
 17 WP_089242443.1_metallophosphoesterase_Calothrix_rhizosoleniae
 18 WP_015112944.1_metallophosphoesterase_Mostoc_sp._PCC_7107_AFY42728.1_metallophosphoesterase_Mostoc_sp._PCC_7107
 19 CBB51321.1_metallophosphoesterase_Mostoc_cycladae_NR-1
 20 WP_013643761.1_metallophosphoesterase_Methanobacterium_lacus_A208410.1_metallophosphoesterase_Methanobacterium_lacus
 21 WP_015121857.1_metallophosphoesterase_Rivularia_sp._PCC_7116_AFY38305.1_putative_phosphohydrolase_Rivularia_sp._PCC_7116
 22 WP_095850899.1_metallophosphoesterase_Anabaenopsis_circularis_BAY17764.1_putative_Ser/Thr_protein_phosphatase_family_protein_Anabaenopsis_circularis_NIES-21
 23 WP_016876205.1_metallophosphoesterase_Chlorogloeopsis_ferrireducens
 24 BAI31673.1_putative_Ser/Thr_protein_phosphatase_family_protein_Cylindrocapsa_sp._NIES-4074
 25 WP_015206774.1_metallophosphoesterase_Cylindrocapsa_sp._NIES-4074_AFE232510.1_putative_phosphohydrolase_Cylindrocapsa_sp._NIES-4074
 26 WP_026092783.1_metallophosphoesterase_Calothrix_sp._PCC_7103
 27 WP_095722007.1_metallophosphoesterase_Calothrix_elsteri_PAX4314.1_serine/threonine_protein_phosphatase_Calothrix_elsteri_CCALA_953
 28 WP_084795049.1_metallophosphoesterase_Methanobacterium_congolense
 29 WP_096685131.1_MULTISPECIES_metallophosphoesterase_Calothrix_BAI09636.1_putative_Ser/Thr_protein_phosphatase_family_protein_Calothrix_sp._NIES-4071_BAI55783.1_putative_Ser/Thr_protein_phosphatase_fas
 30 SCG67688.1_putative_metallophosphoesterase_Yku_Methanobacterium_congolense
 31 WP_084709347.1_metallophosphoesterase_Rubrobacter_aplysiinae
 32 KST67425.1_serine/threonine_protein_phosphatase_Mastigocoleus_testaceus_BCC008
 33 WP_082650565.1_metallophosphoesterase_Mastigocoleus_testaceus
 34 WP_013826931.1_metallophosphoesterase_Methanobacterium_paludis_ARG19332.1_metallophosphoesterase_Methanobacterium_paludis
 35 P3F40307.1_serine/threonine_protein_phosphatase_Chloroflexi_bacterium
 36 WP_048071780.1_metallophosphoesterase_Methanobacterium_fomicium_CEL12430.1_metallophosphoesterase_Methanobacterium_fomicium_CEL12430.M
 37 CDB13846.1_phosphoesterase(EC:3.1.-)_Methanobacterium_fomicium
 38 WP_082727286.1_metallophosphoesterase_Mostoc_sp._NIES-3756_BAT55089.1_Ser/Thr_protein_phosphatase_family_protein_putative_Mostoc_sp._NIES-3756
 39 WP_023991927.1_metallophosphoesterase_Methanobacterium_sp._MB1_CDB44491.1_metallophosphoesterase_Methanobacterium_sp._MB1
 40 WP_086678543.1_metallophosphoesterase_Mostoc_sp._NIES-2111_BAY77127.1_putative_Ser/Thr_protein_phosphatase_family_protein_Mostoc_sp._NIES-2111
 41 KTX75234.1_Metallophosphoesterase_Methanobacterium_sp._42_16
 42 WP_040682302.1_metallophosphoesterase_Methanobacterium_sp._42_16

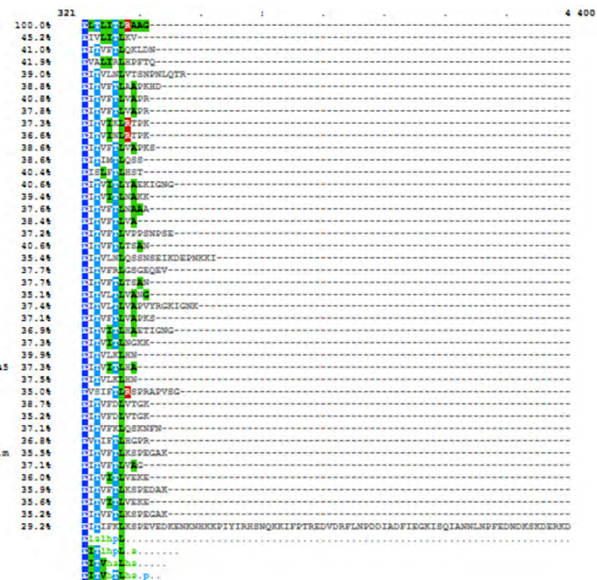


Figure S6. Maximum likelihood analysis of predicted esterase sequences. The tree with the highest log likelihood (-9571.0445) is shown. The percentage of trees in which the associated taxa clustered together is shown next to the branches. Initial tree(s) for the heuristic search were obtained automatically by applying Neighbor-Join and BioNJ algorithms to a matrix of pairwise distances estimated using a JTT model, and then selecting the topology with superior log likelihood value. The tree is drawn to scale, with branch lengths measured in the number of substitutions per site. The analysis involved 28 amino acid sequences. All positions containing gaps and missing data were eliminated. There were a total of 271 positions in the final dataset. Evolutionary analyses were conducted in MEGA7. The predicted esterase derived from metagenomic clone P6L4 is marked by a red arrow.

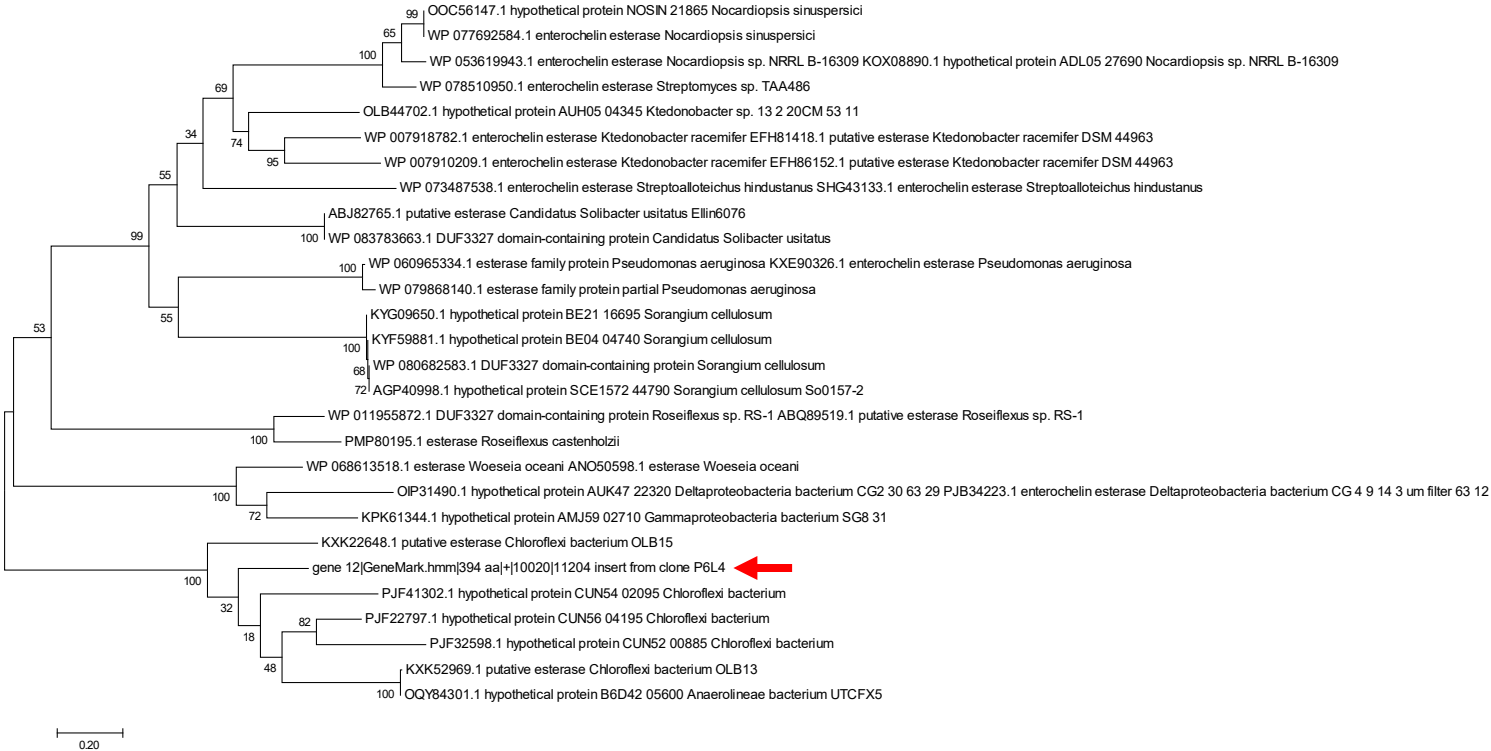


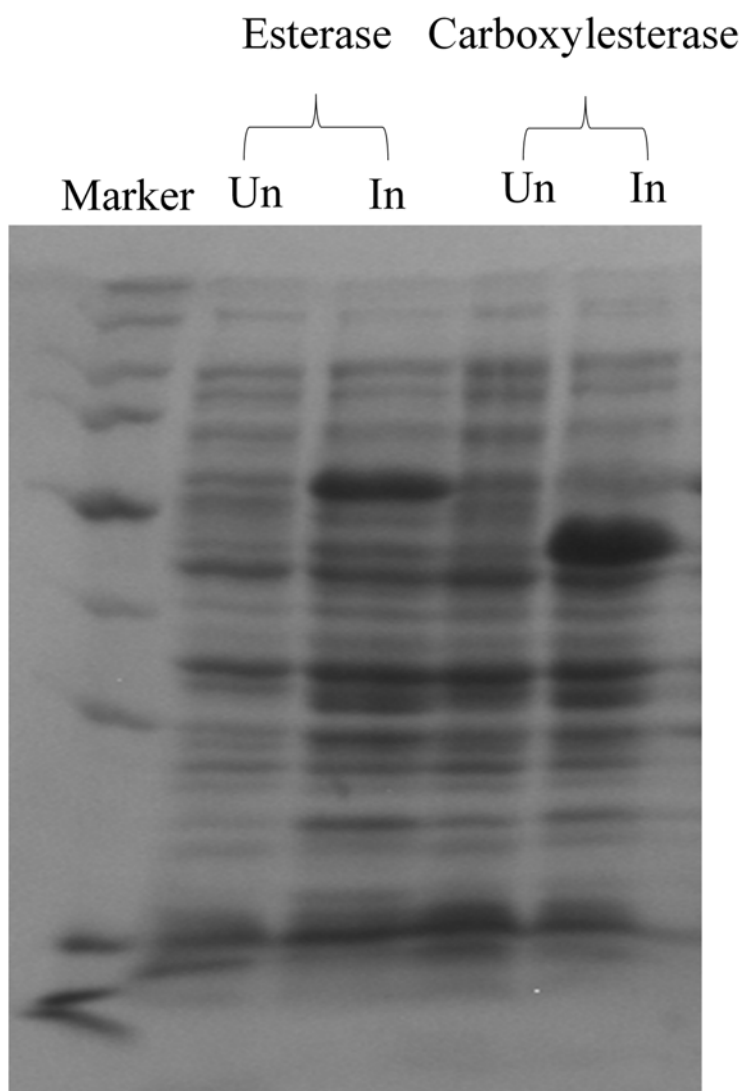
Figure S7. Maximum likelihood analysis of predicted carboxylesterase sequences. The tree with the highest log likelihood (-12477.0427) is shown. The percentage of trees in which the associated taxa clustered together is shown next to the branches. Initial tree(s) for the heuristic search were obtained automatically by applying Neighbor-Join and BioNJ algorithms to a matrix of pairwise distances estimated using a JTT model, and then selecting the topology with superior log likelihood value. The tree is drawn to scale, with branch lengths measured in the number of substitutions per site. The analysis involved 41 amino acid sequences. All positions containing gaps and missing data were eliminated. There were a total of 240 positions in the final dataset. Evolutionary analyses were conducted in MEGA7. The predicted esterase derived from metagenomic clone P6L4 is marked by a red arrow.



Figure S8. Maximum likelihood analysis of predicted metallophosphoesterase sequences. The tree with the highest log likelihood (-10240.6863) is shown. The percentage of trees in which the associated taxa clustered together is shown next to the branches. Initial tree(s) for the heuristic search were obtained automatically by applying Neighbor-Join and BioNJ algorithms to a matrix of pairwise distances estimated using a JTT model, and then selecting the topology with superior log likelihood value. The tree is drawn to scale, with branch lengths measured in the number of substitutions per site. The analysis involved 42 amino acid sequences. All positions containing gaps and missing data were eliminated. There were a total of 252 positions in the final dataset. Evolutionary analyses were conducted in MEGA7. The predicted esterase derived from metagenomic clone P6L4 is marked by a red arrow.



Figure S9. Induced expression of esterase genes subcloned from clone P6L4 using the Expresso Rhamnose SUMO system. The SDS-PAGE gel indicates protein bands observed in the *E. coli* cultures of the subcloned esterase or carboxylesterase in the absence of rhamnose (uninduced; Un) or in the presence of rhamnose to induce expression (induced; In). Bands corresponding to the respective recombinant proteins can be observed after induction.



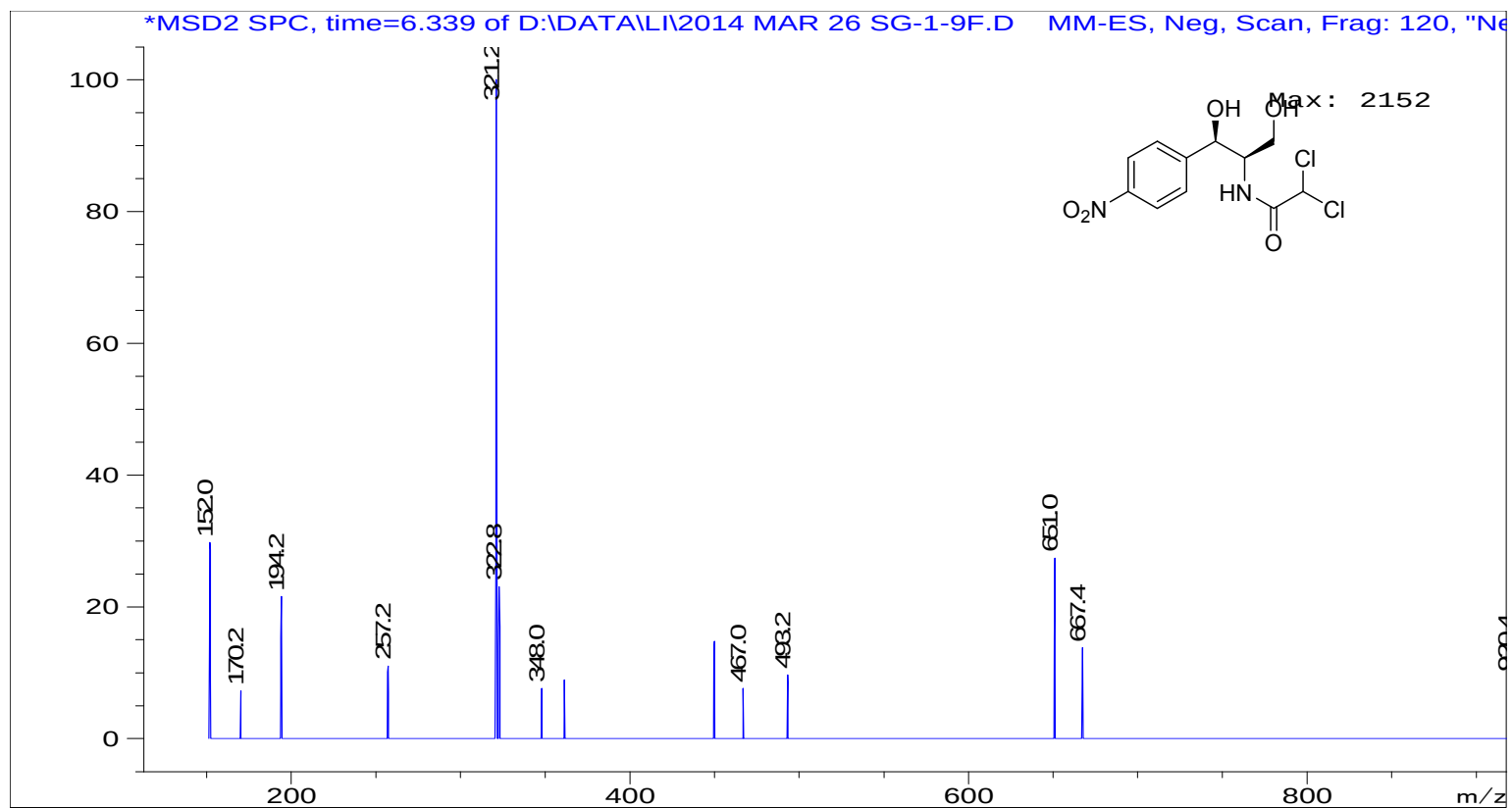


Figure S10. ESI-MS Spectrum of Compound 1

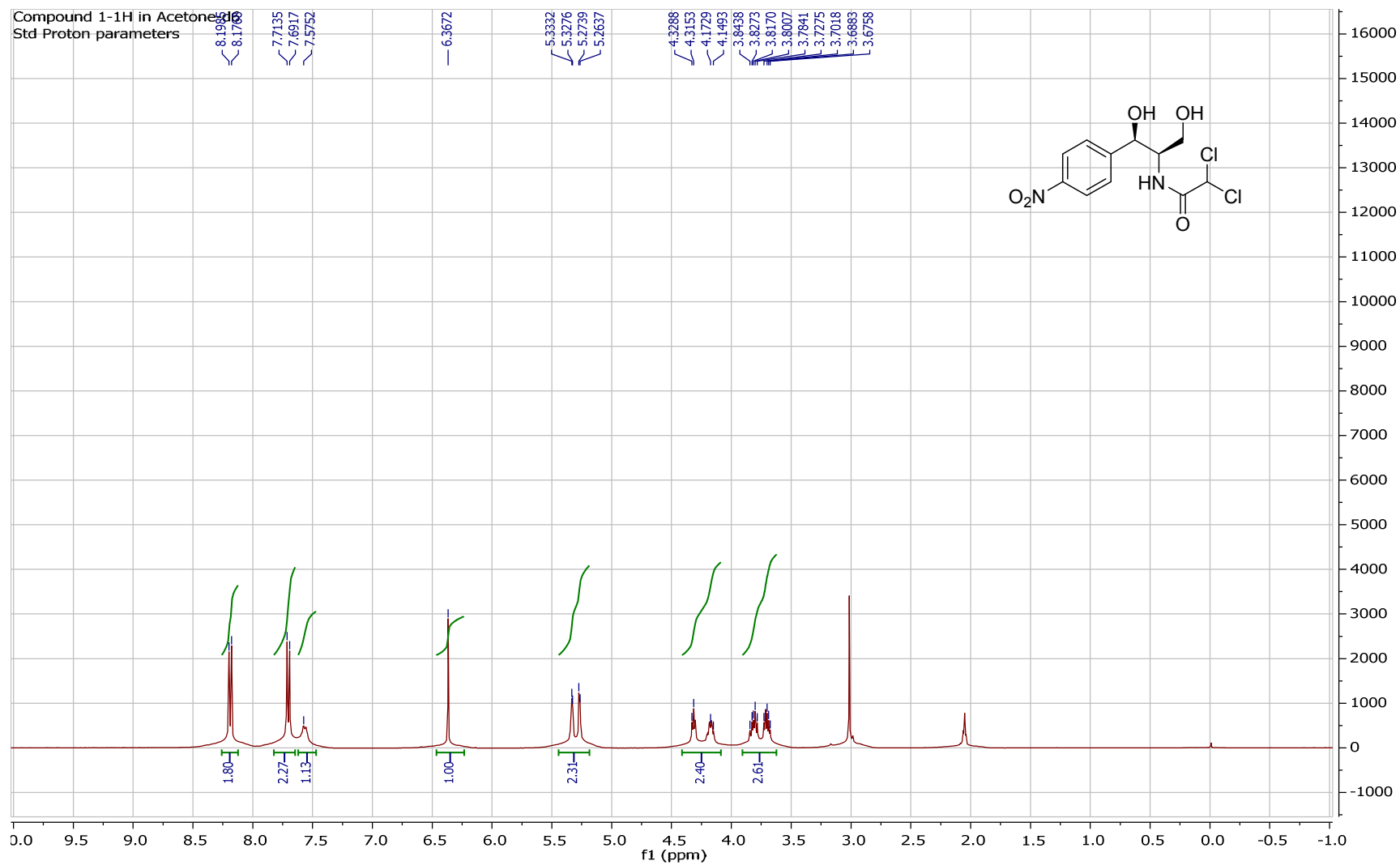
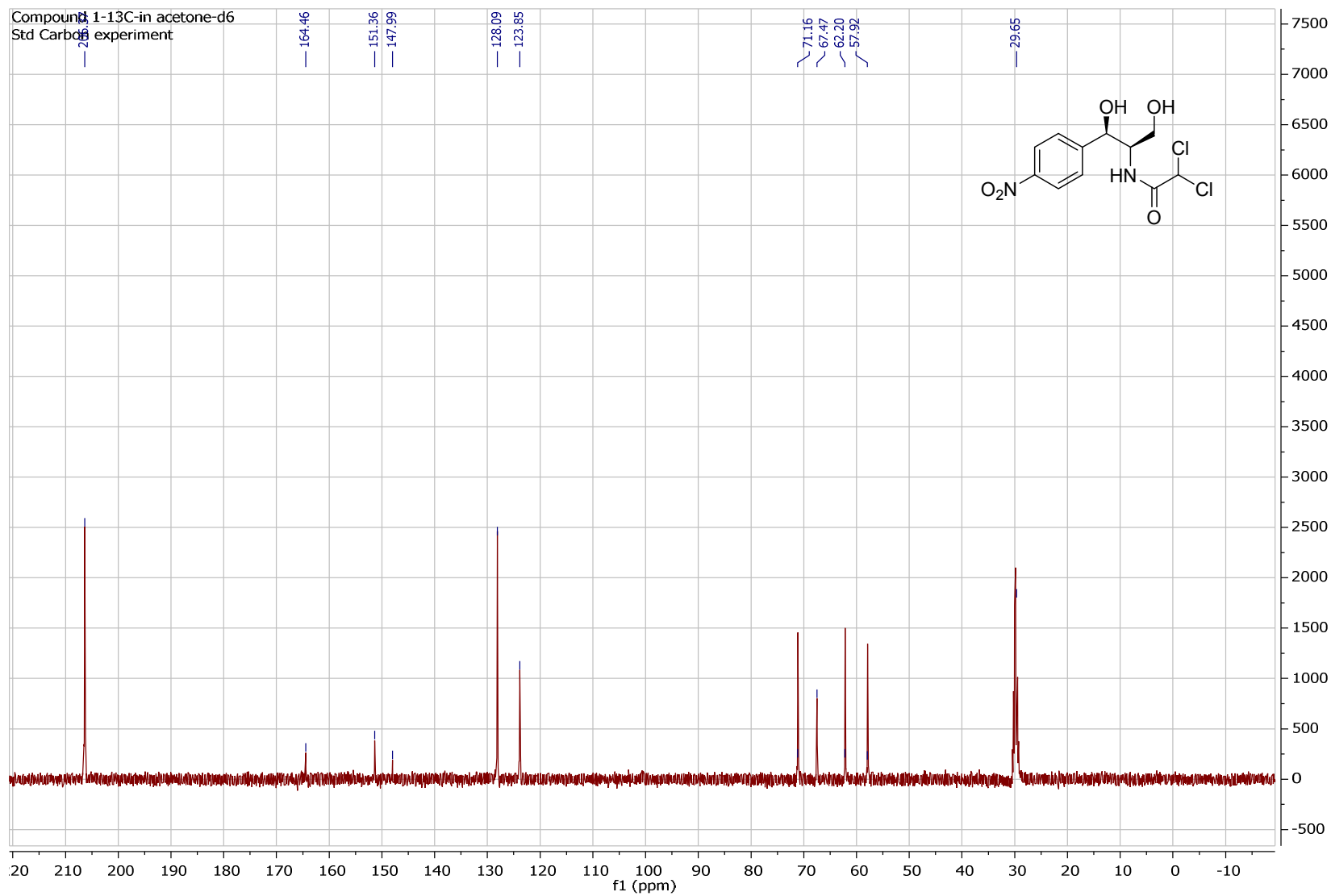


Figure S11. ^1H NMR Spectrum of Compound 1 in Acetone- d_6



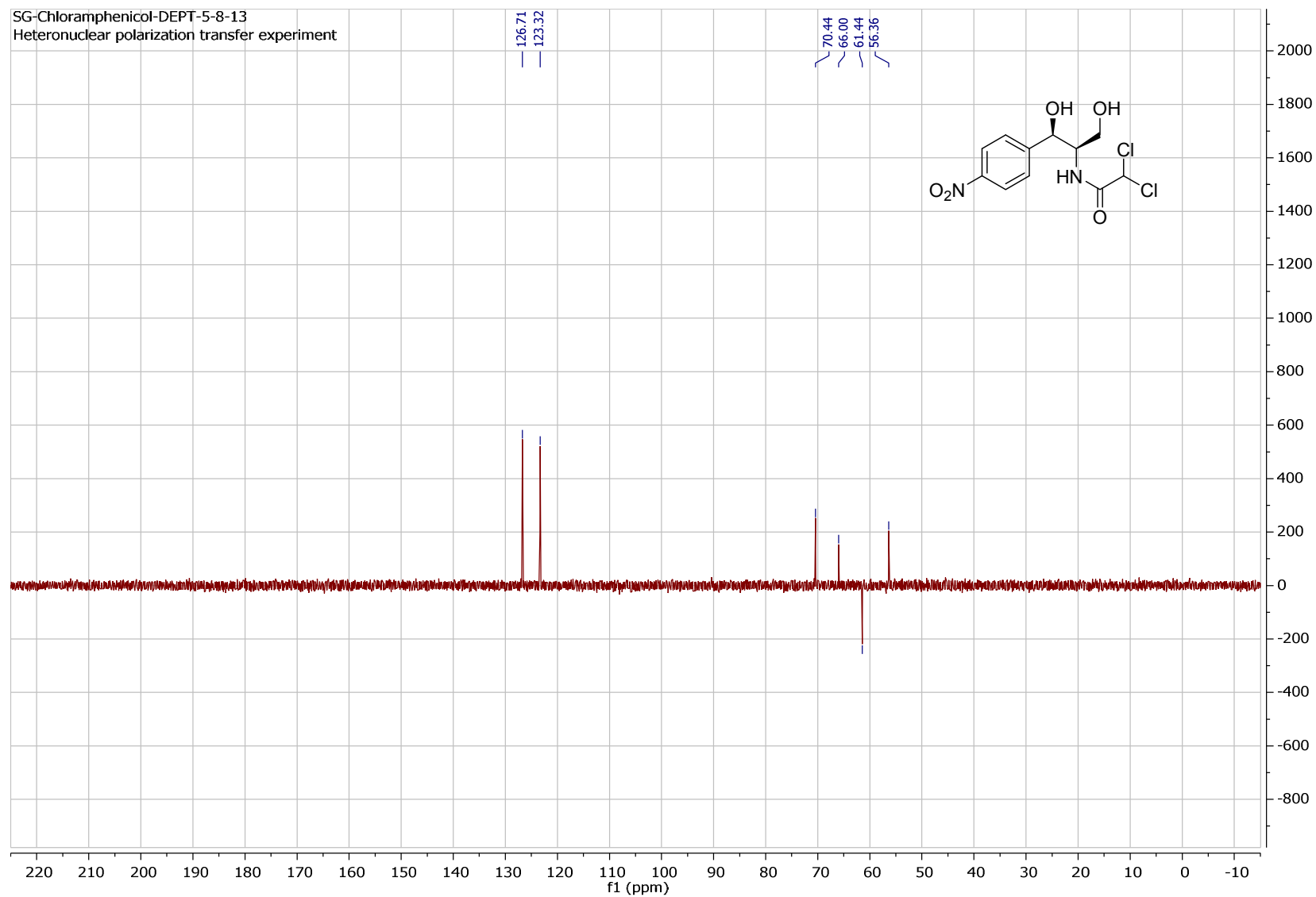


Figure S13. DEPT-135 Spectrum of Compound 1 in Acetone-*d*₆

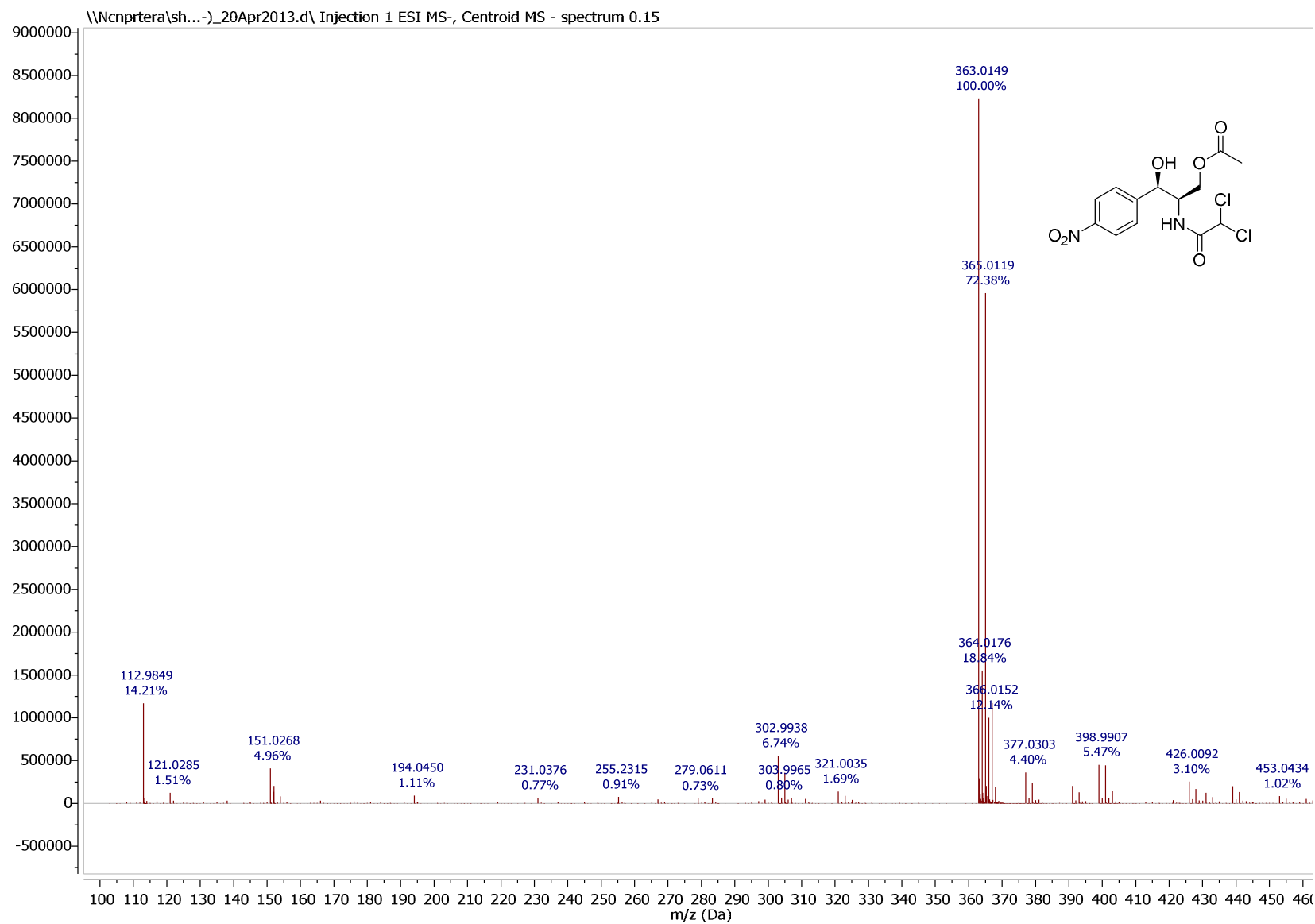


Figure S14. HRESI-MS Spectrum of Compound 2

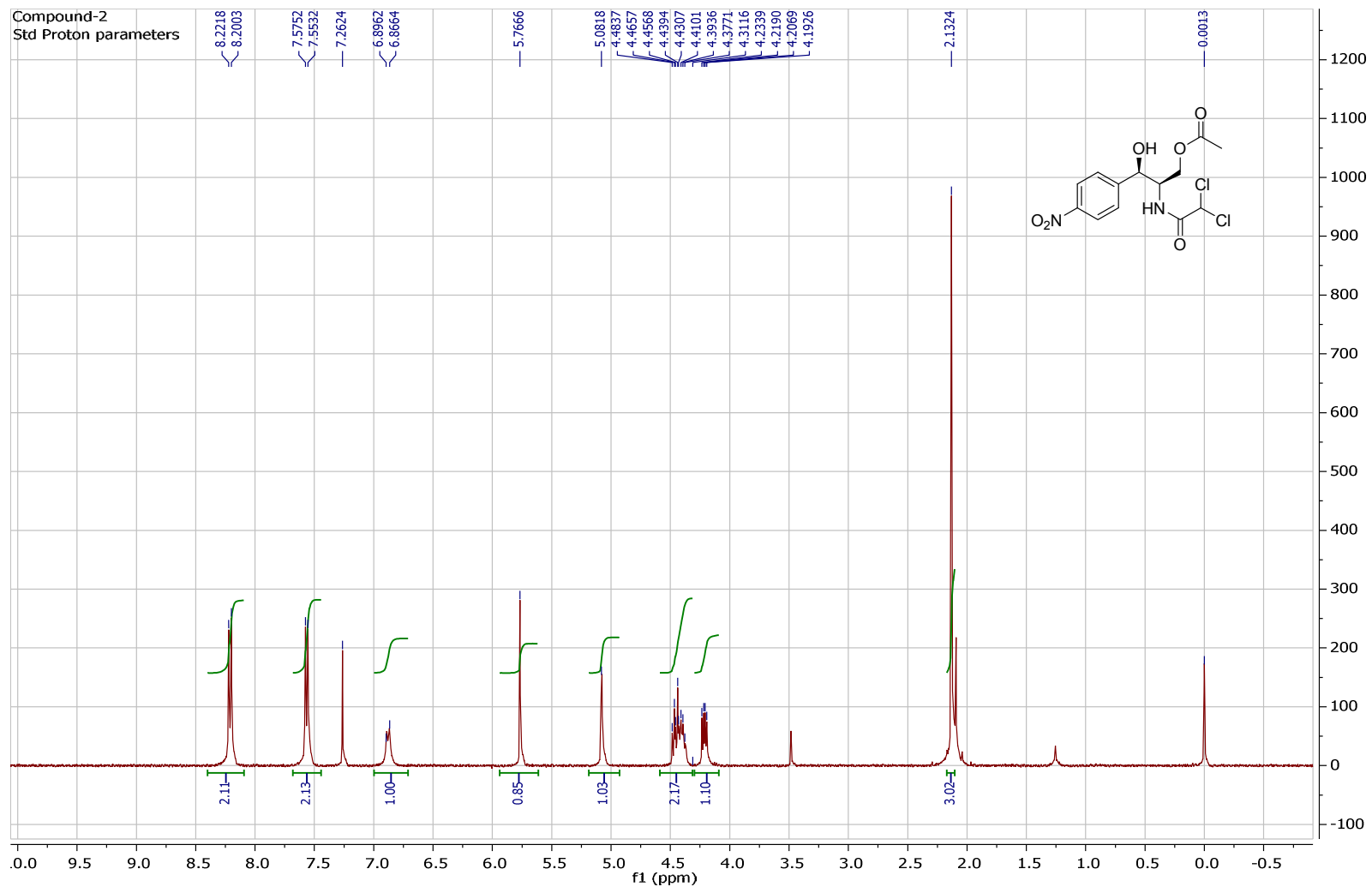


Figure S15. ^1H NMR Spectrum of Compound 2 in CDCl_3

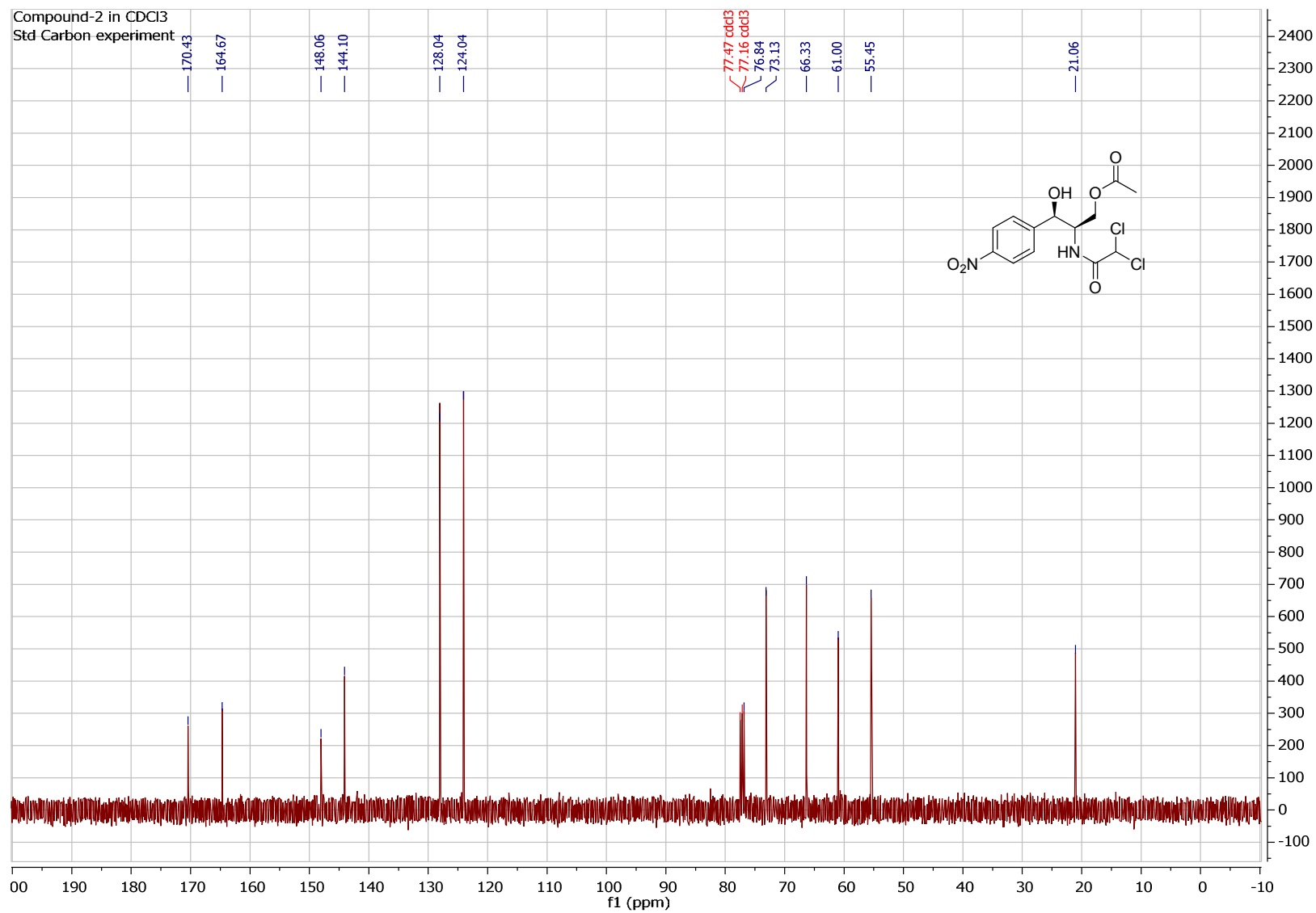


Figure S16. ¹³C NMR spectrum of Compound 2 in CDCl₃

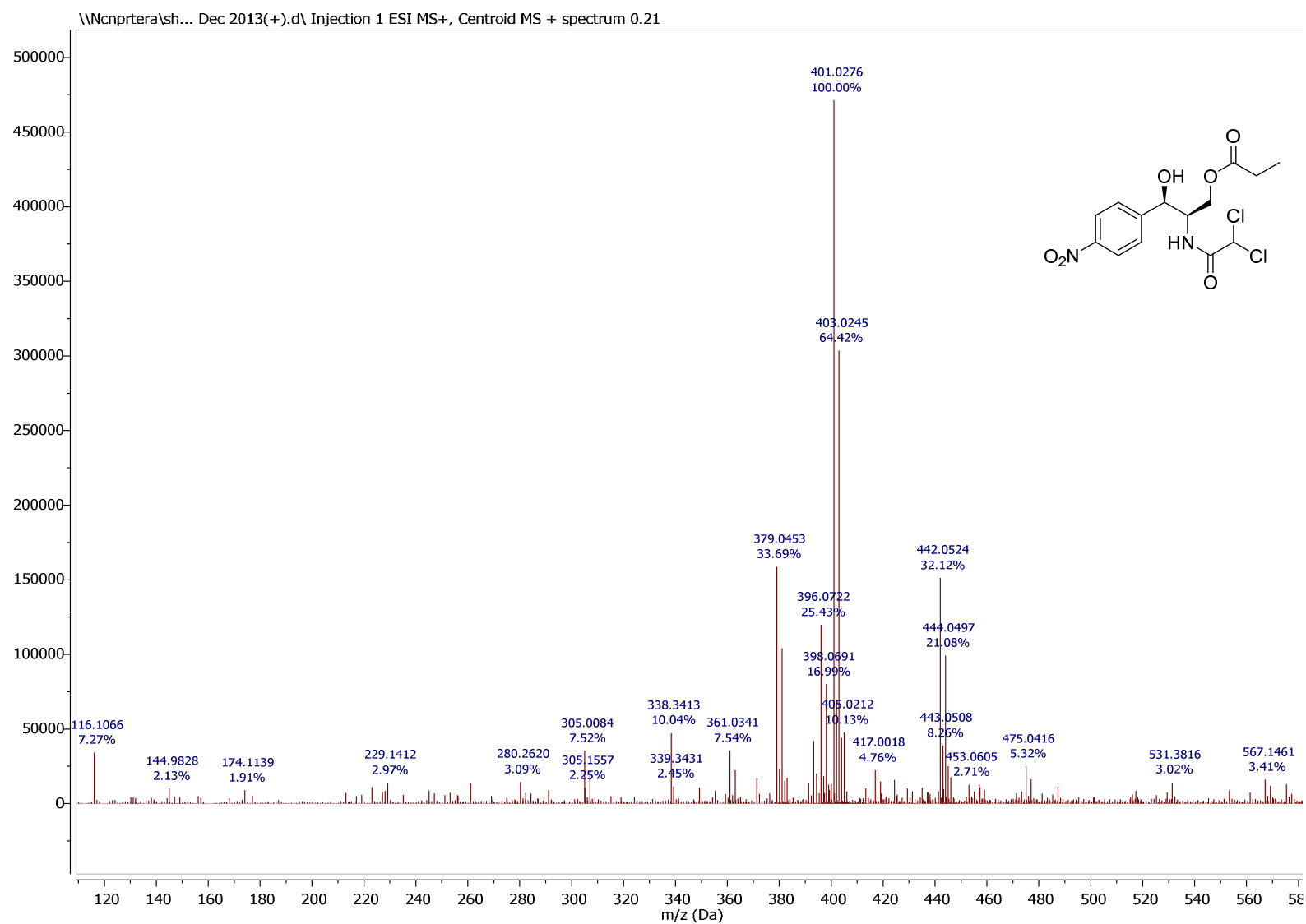


Figure S17. HRESI-MS Spectrum of Compound 3

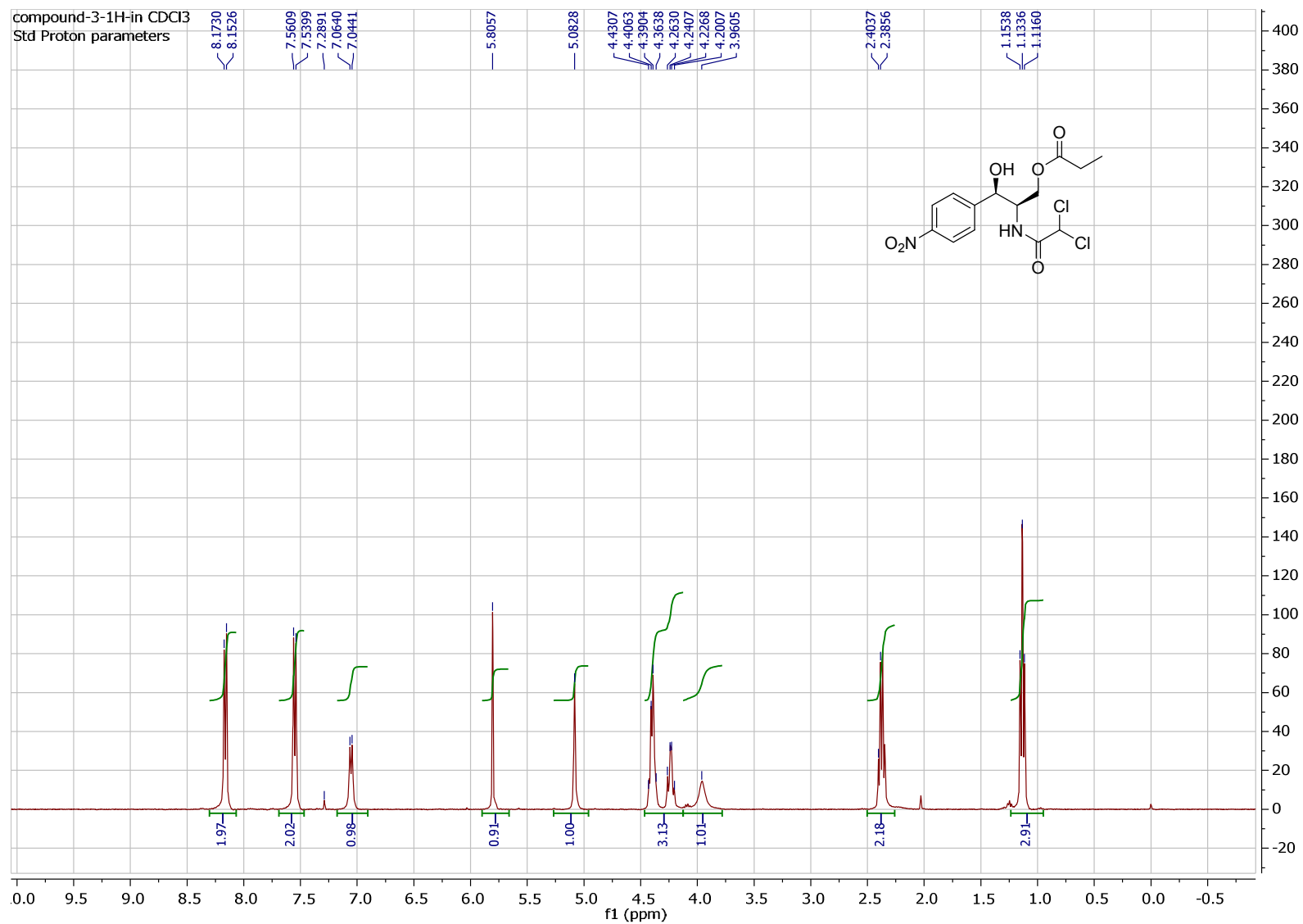


Figure S18. ¹H NMR Spectrum of Compound 3 in CDCl₃

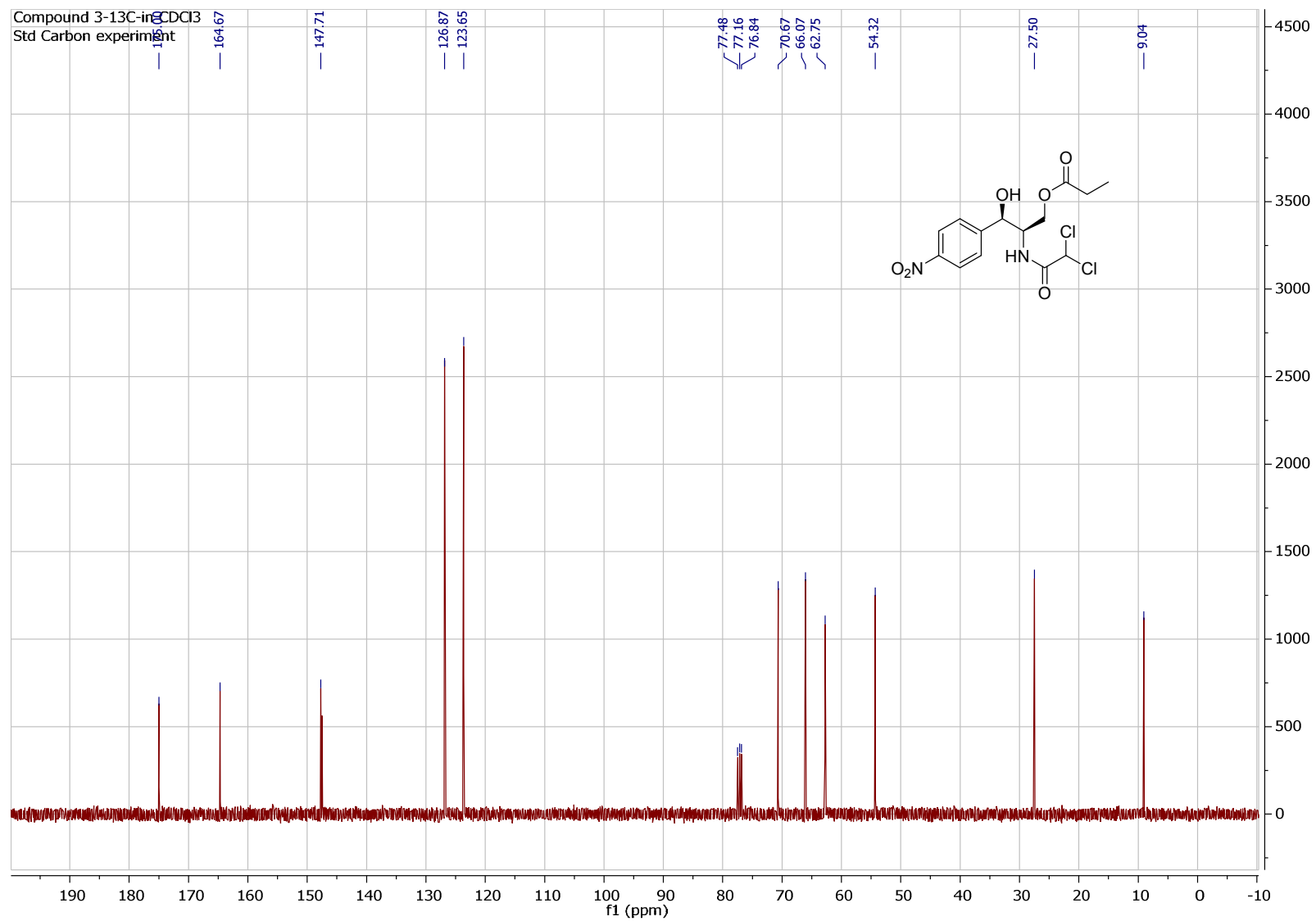


Figure S19. ¹³C NMR Spectrum of Compound 3 in CDCl₃

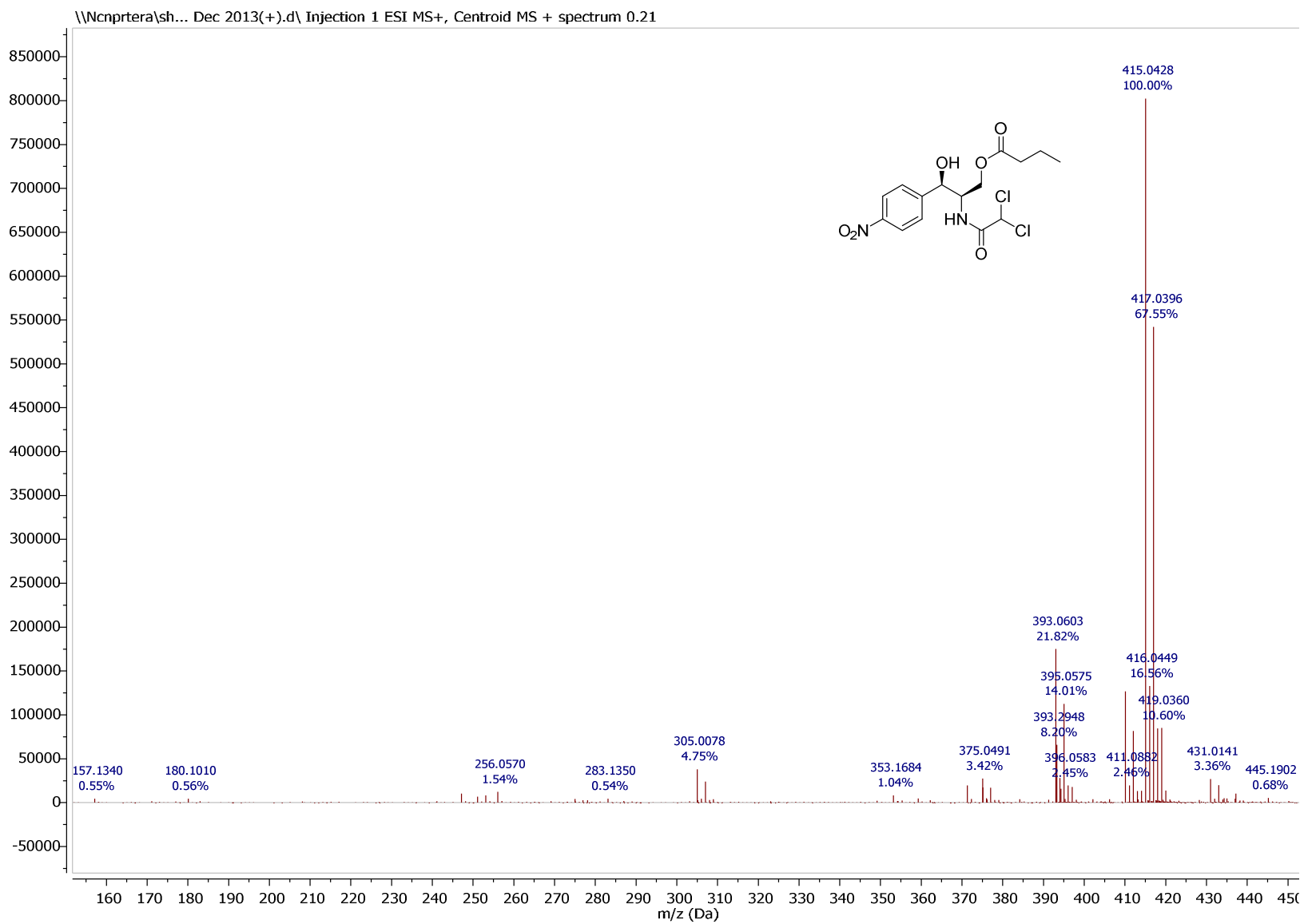


Figure S20. HRESI-MS Spectrum of Compound 4

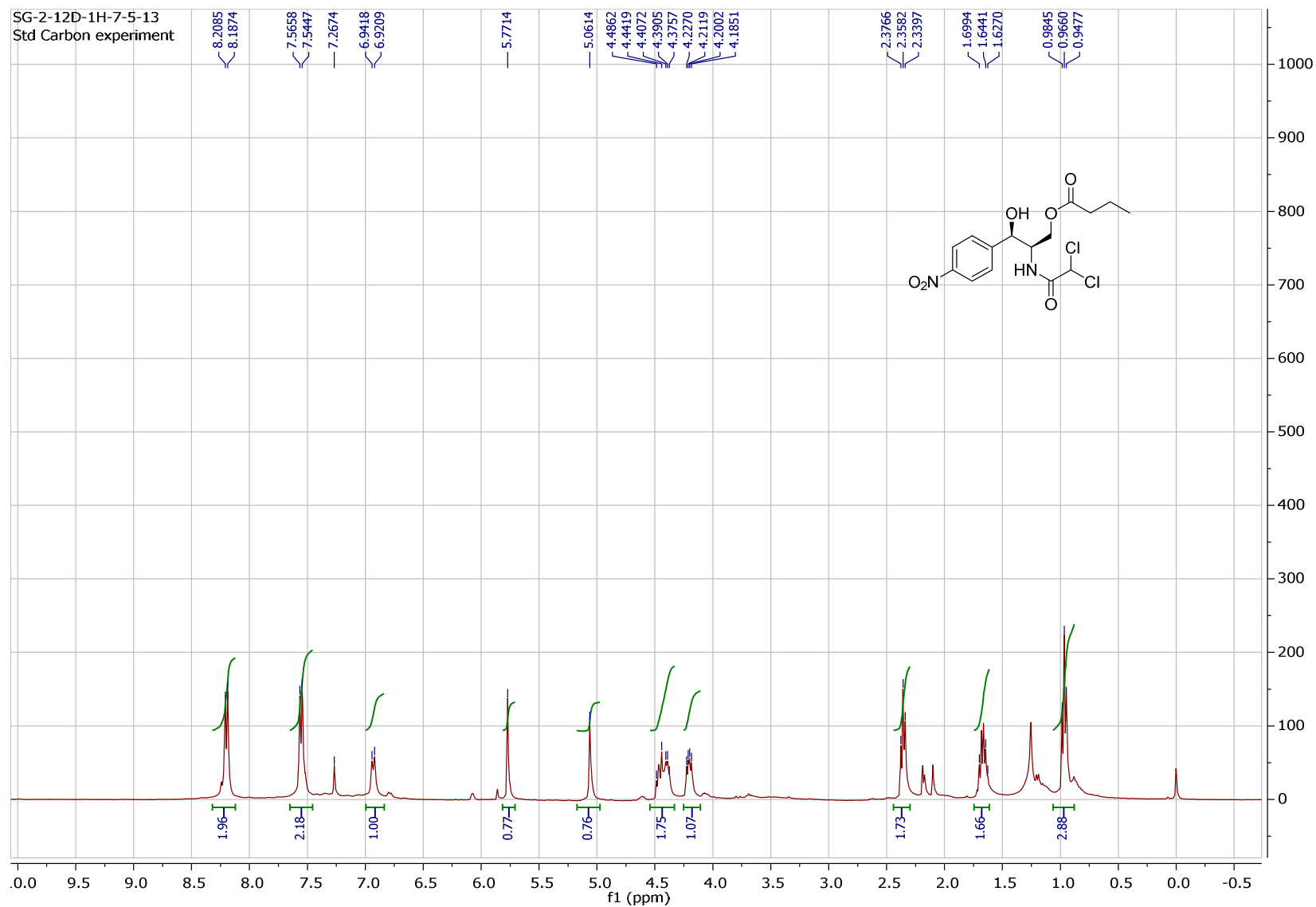


Figure S21. ^1H NMR Spectrum of Compound 4 in CDCl_3

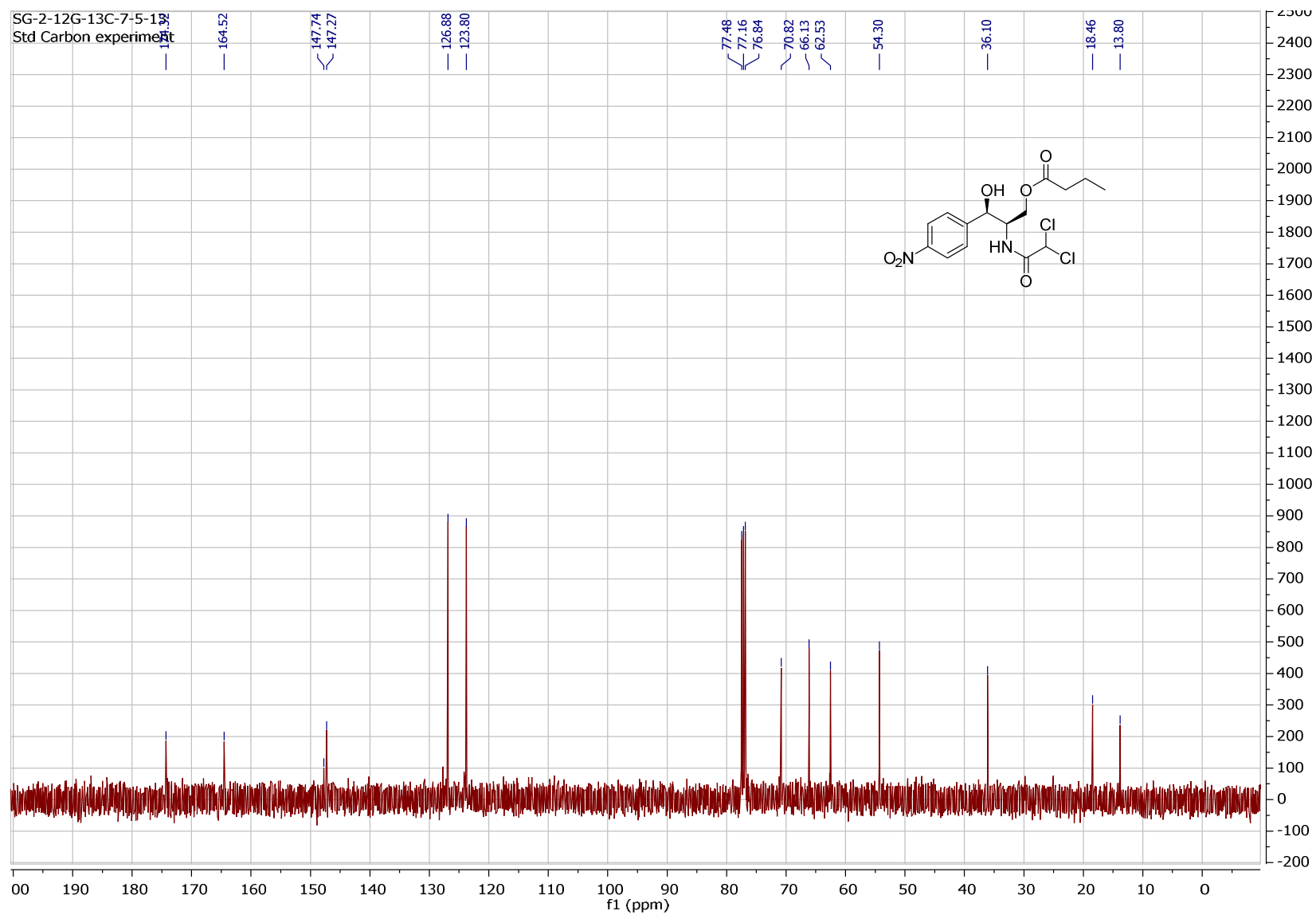


Figure S22. ^{13}C NMR Spectrum of Compound 4 in CDCl_3

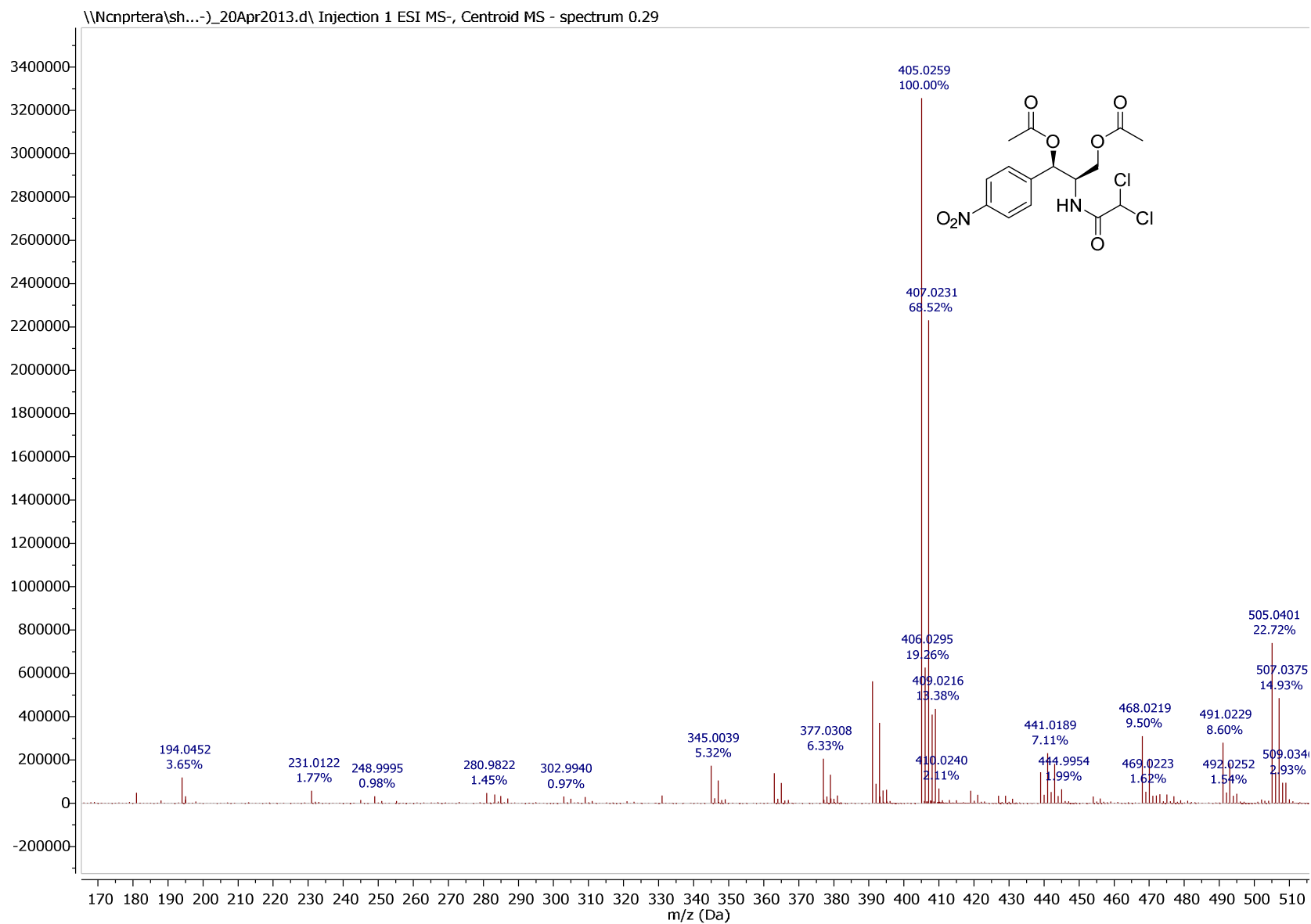


Figure S23. HRESI-MS Spectrum of Compound 5

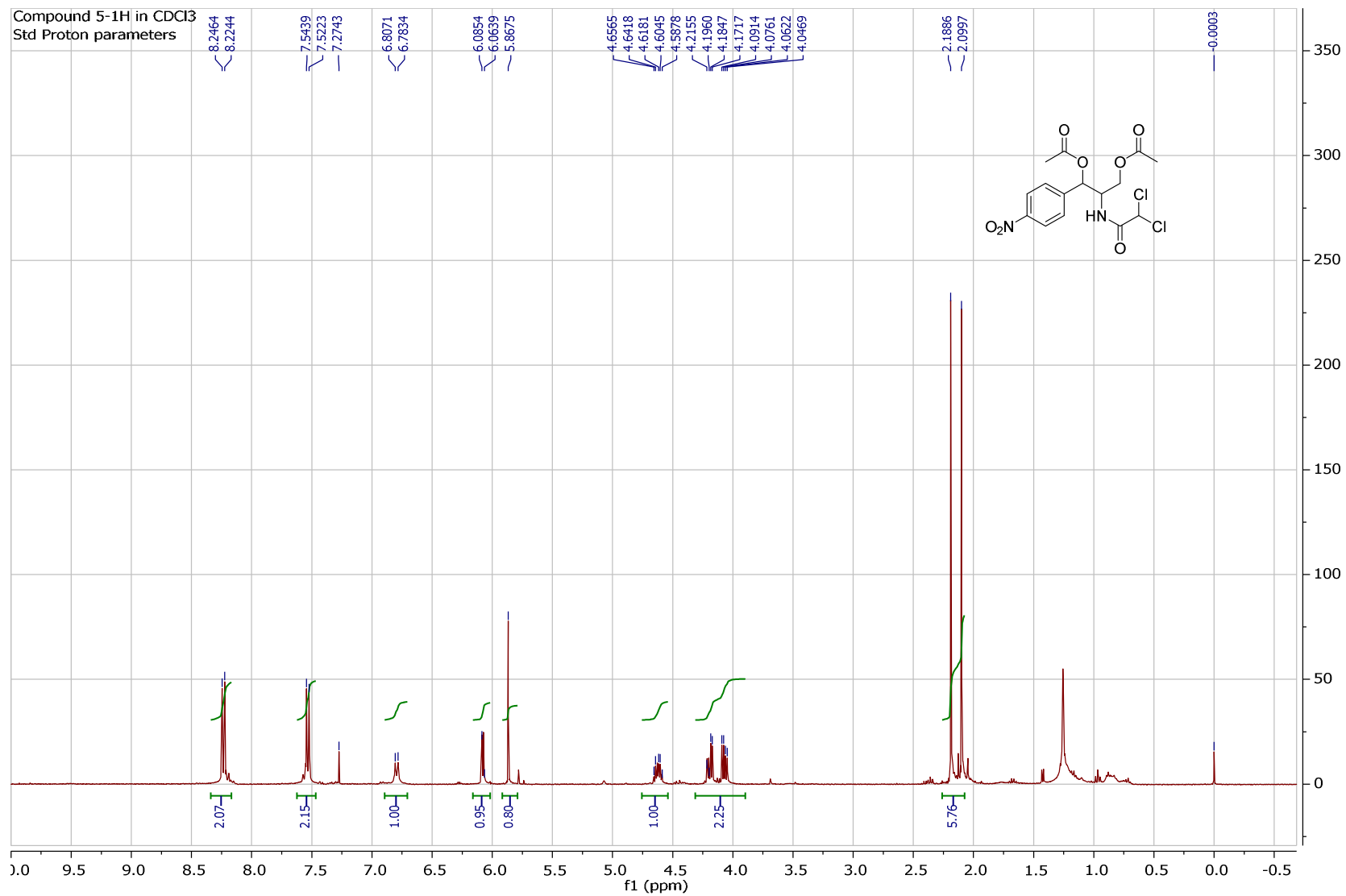


Figure S24. ¹H NMR Spectrum of Compound 5 in CDCl₃

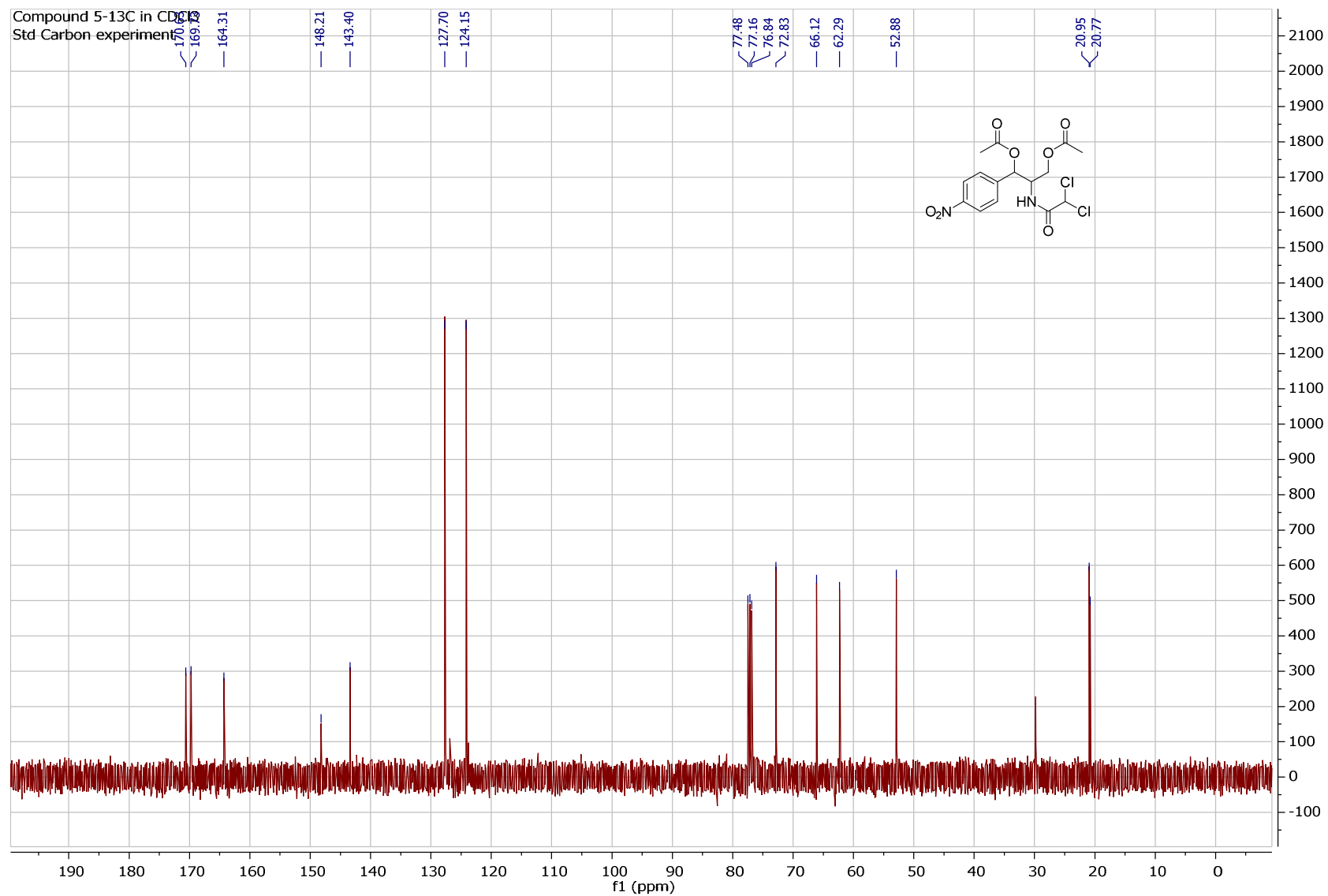


Figure S25. ¹³C NMR Spectrum of Compound 5 in CDCl₃

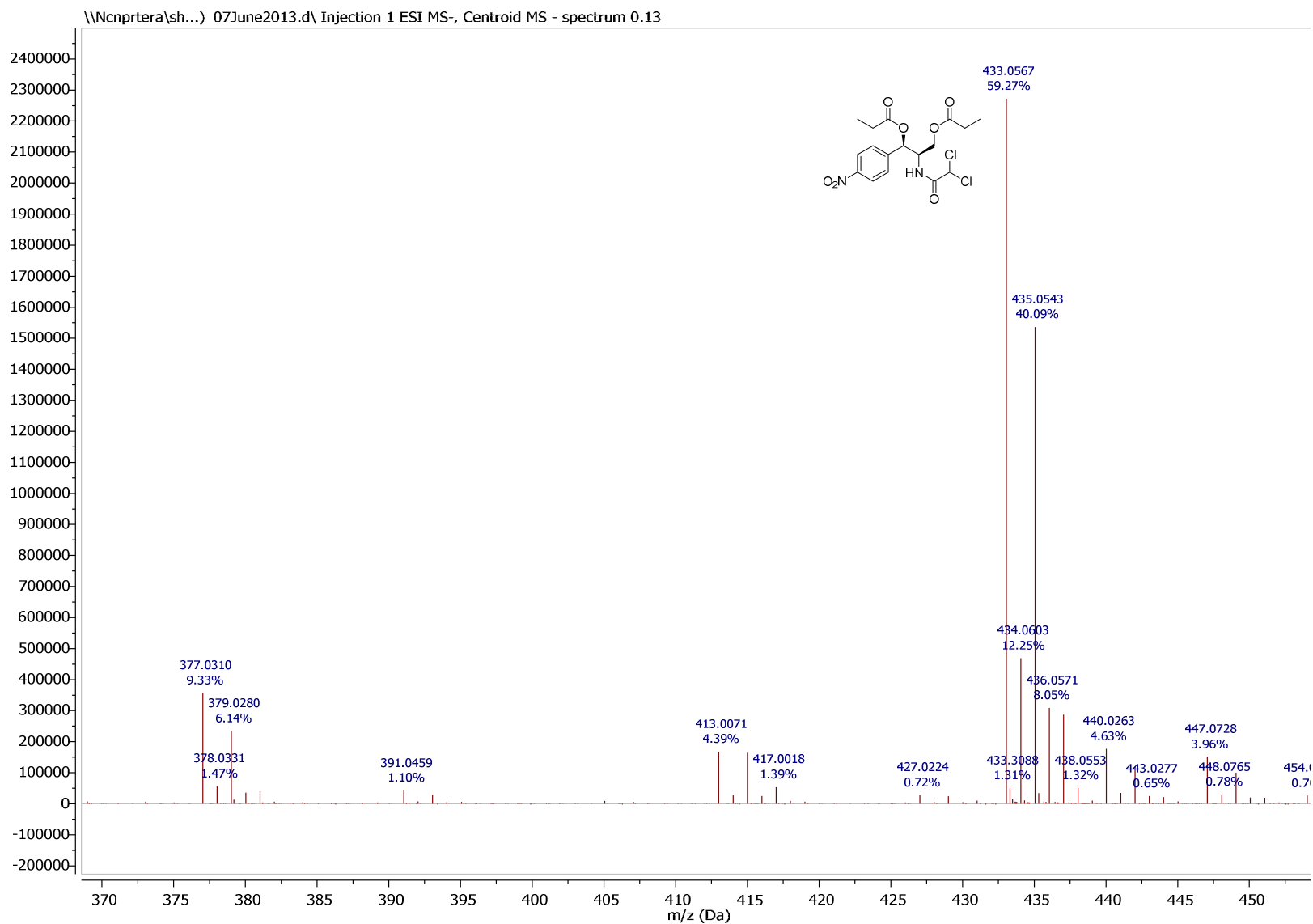


Figure S26. HRESI-MS Spectrum of Compound 6

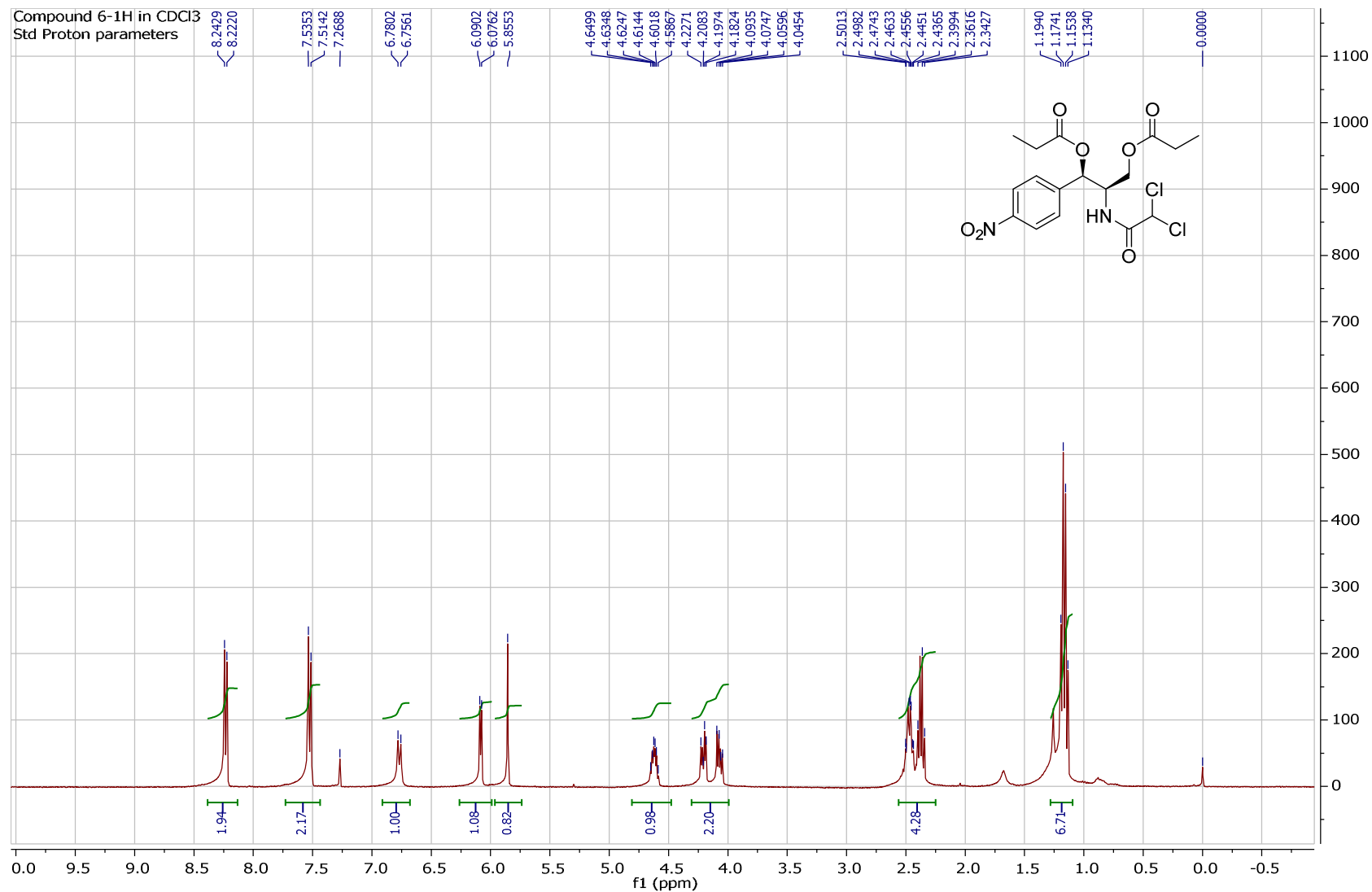


Figure S27. ¹H NMR Spectrum of Compound 6 in CDCl₃

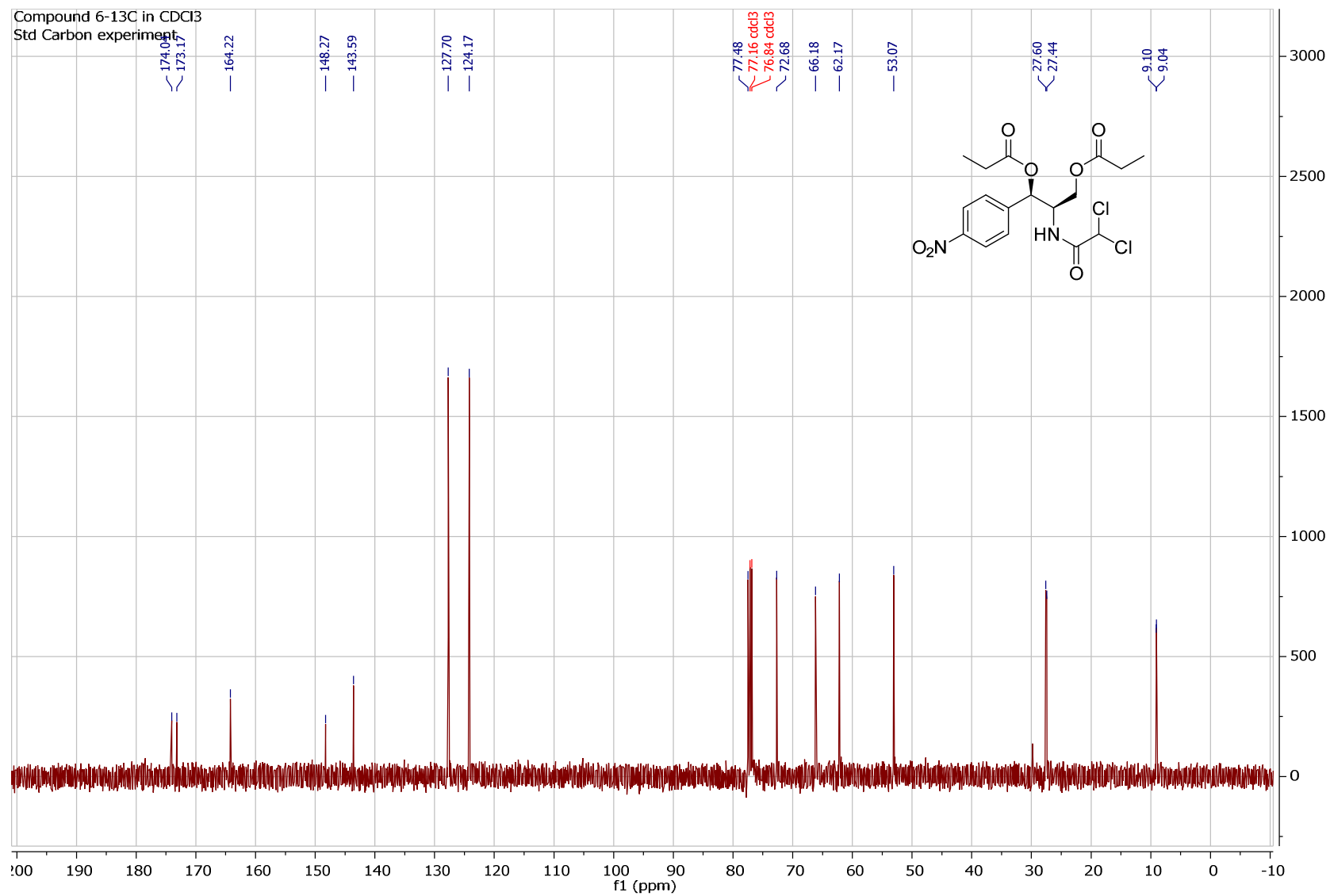


Figure S28. ¹³C NMR Spectrum of Compound 6 in CDCl₃

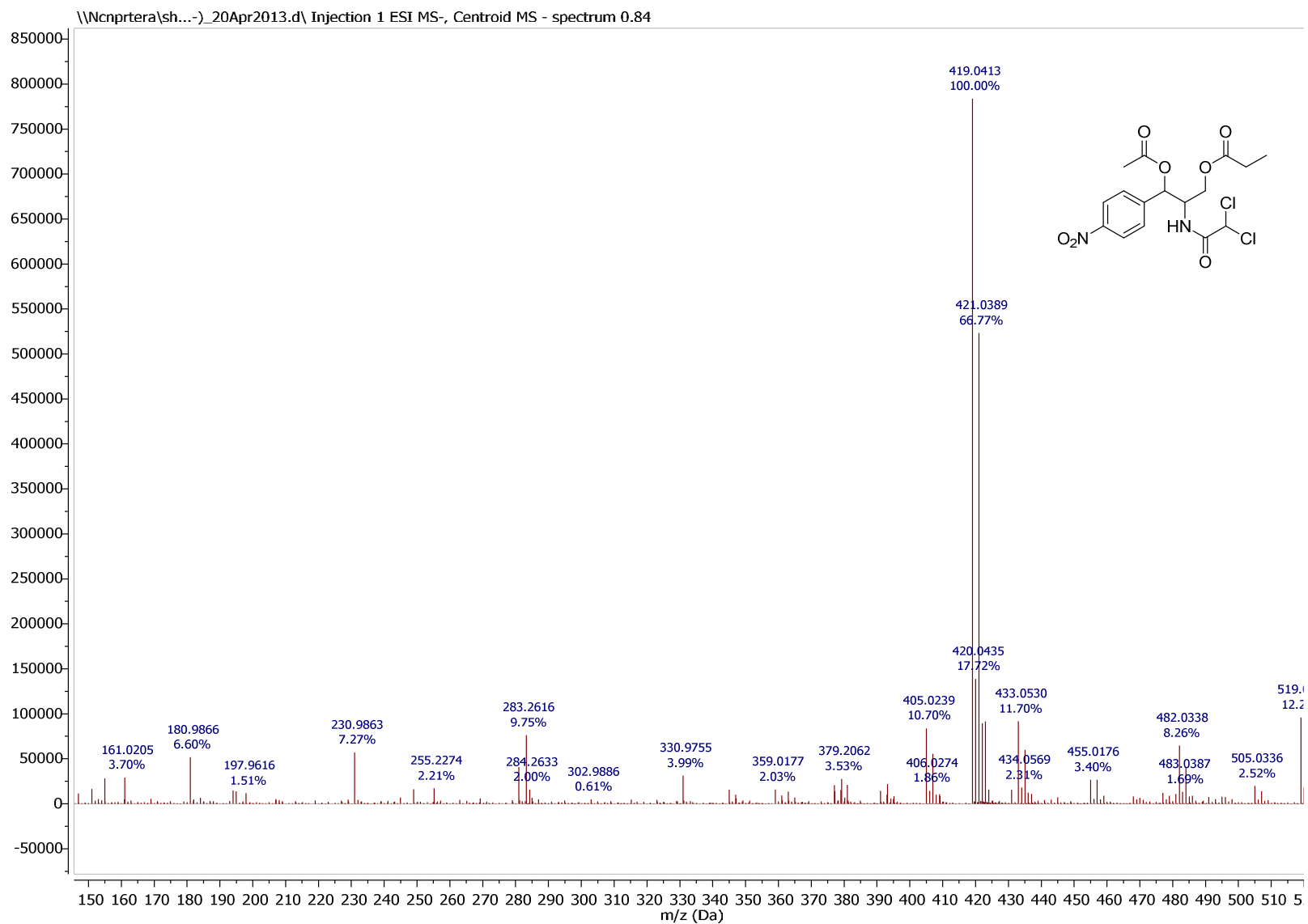


Figure S29. HRESI-MS Spectrum of Compound 7

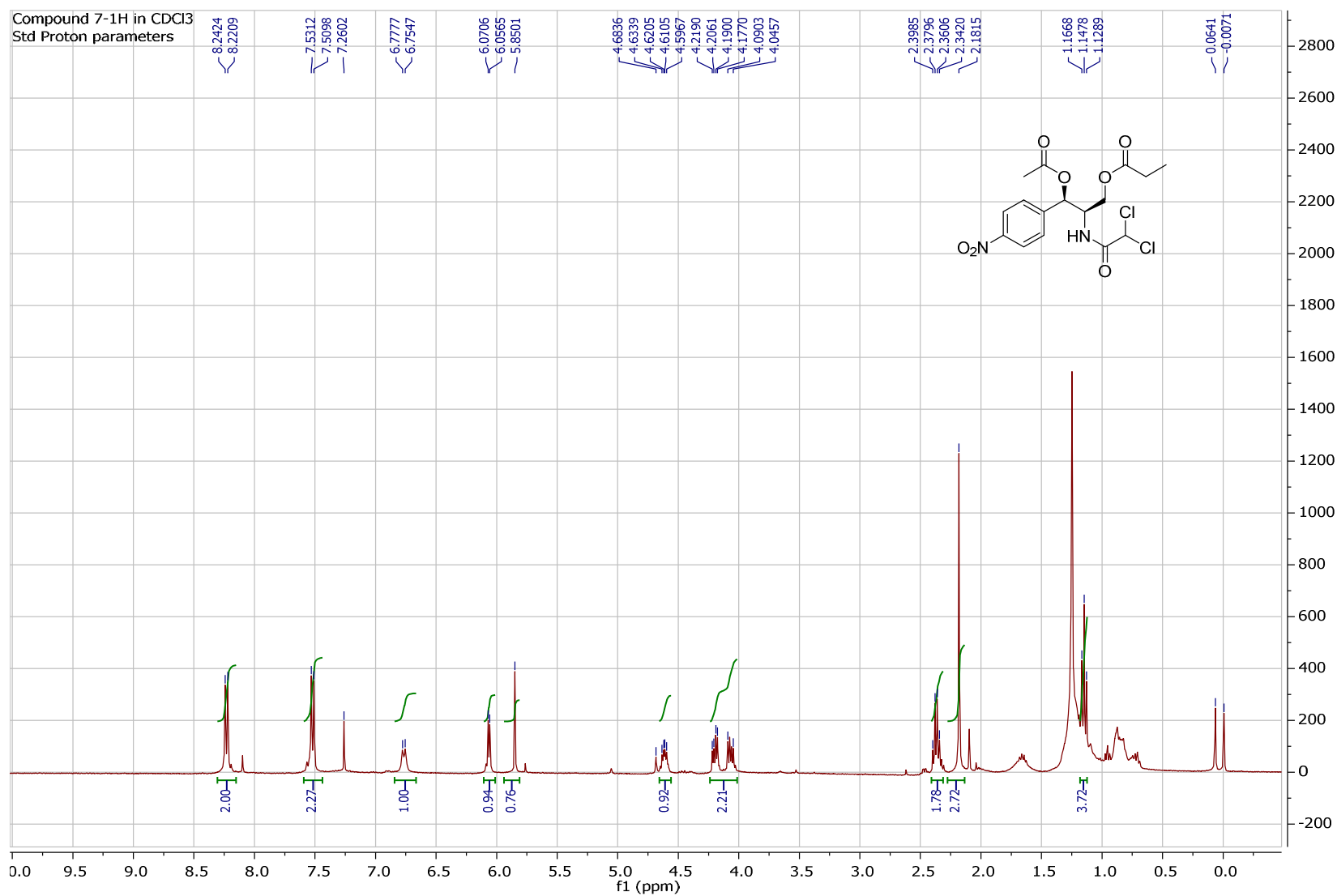


Figure S30. ¹H NMR Spectrum of Compound 7 in CDCl₃

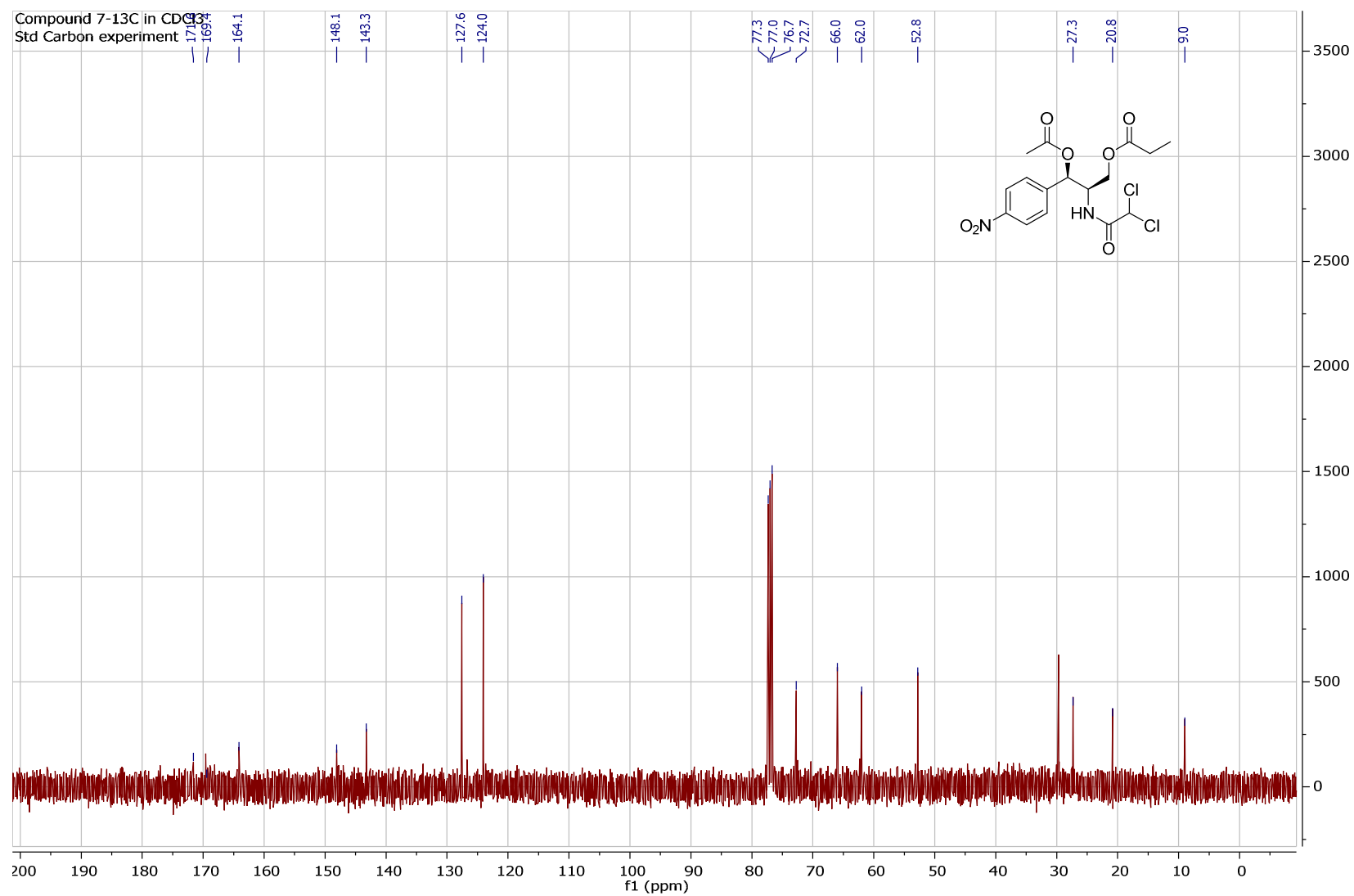


Figure S31. ¹³C NMR Spectrum of Compound 7 in CDCl₃

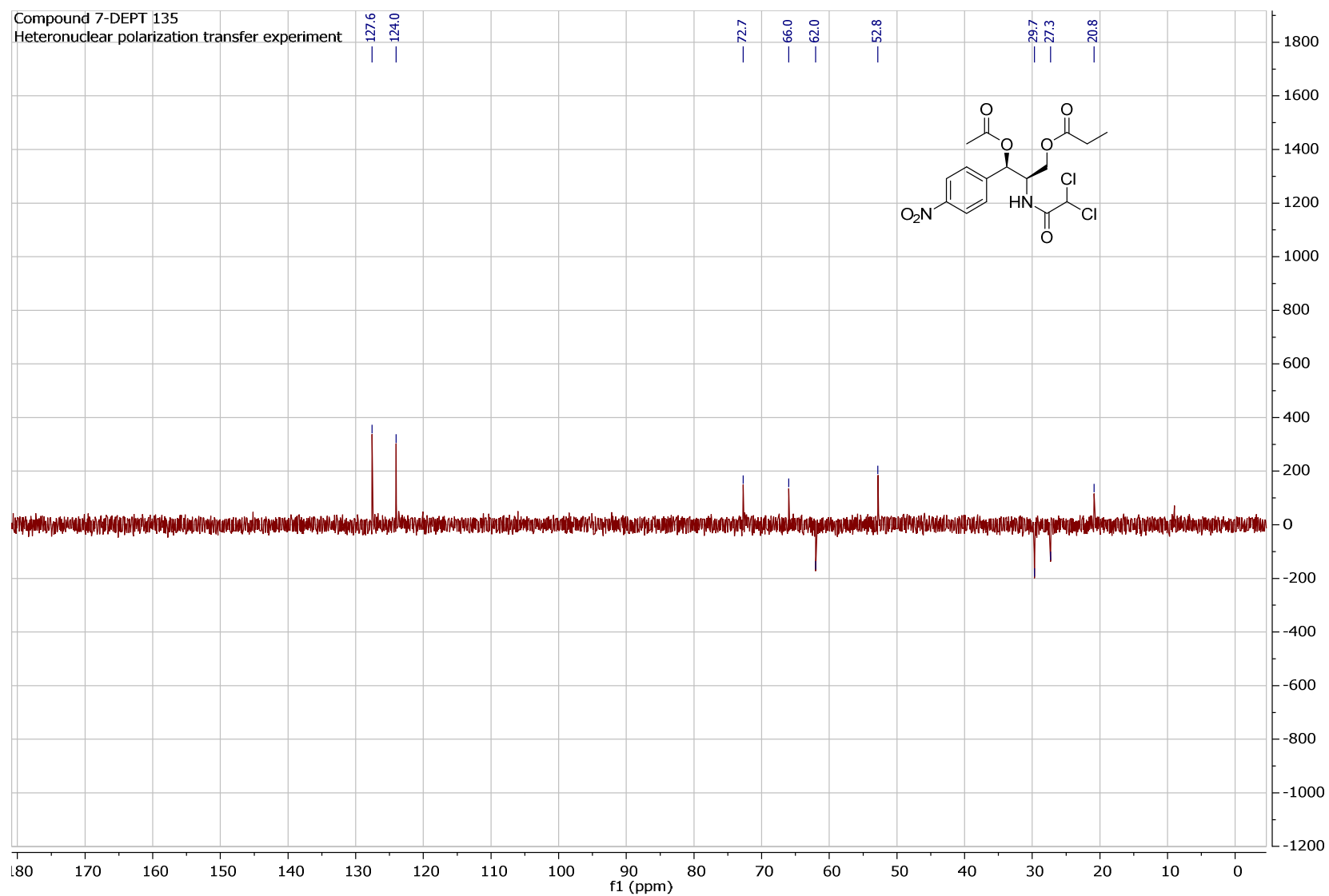


Figure S32. DEPT 135° Spectrum of Compound 7

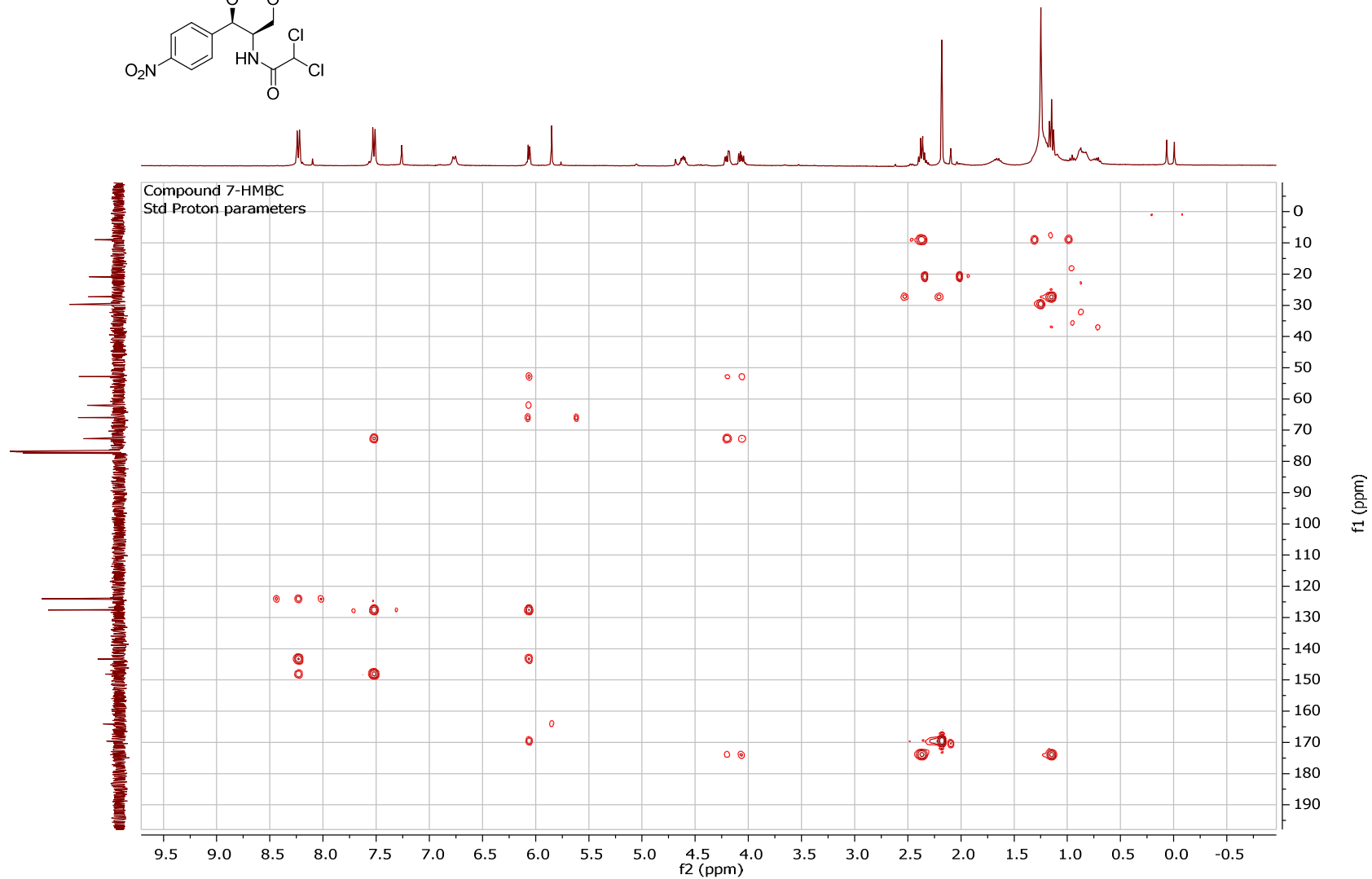
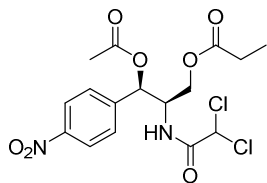


Figure S33. HMBC Spectrum of Compound 7 in CDCl_3

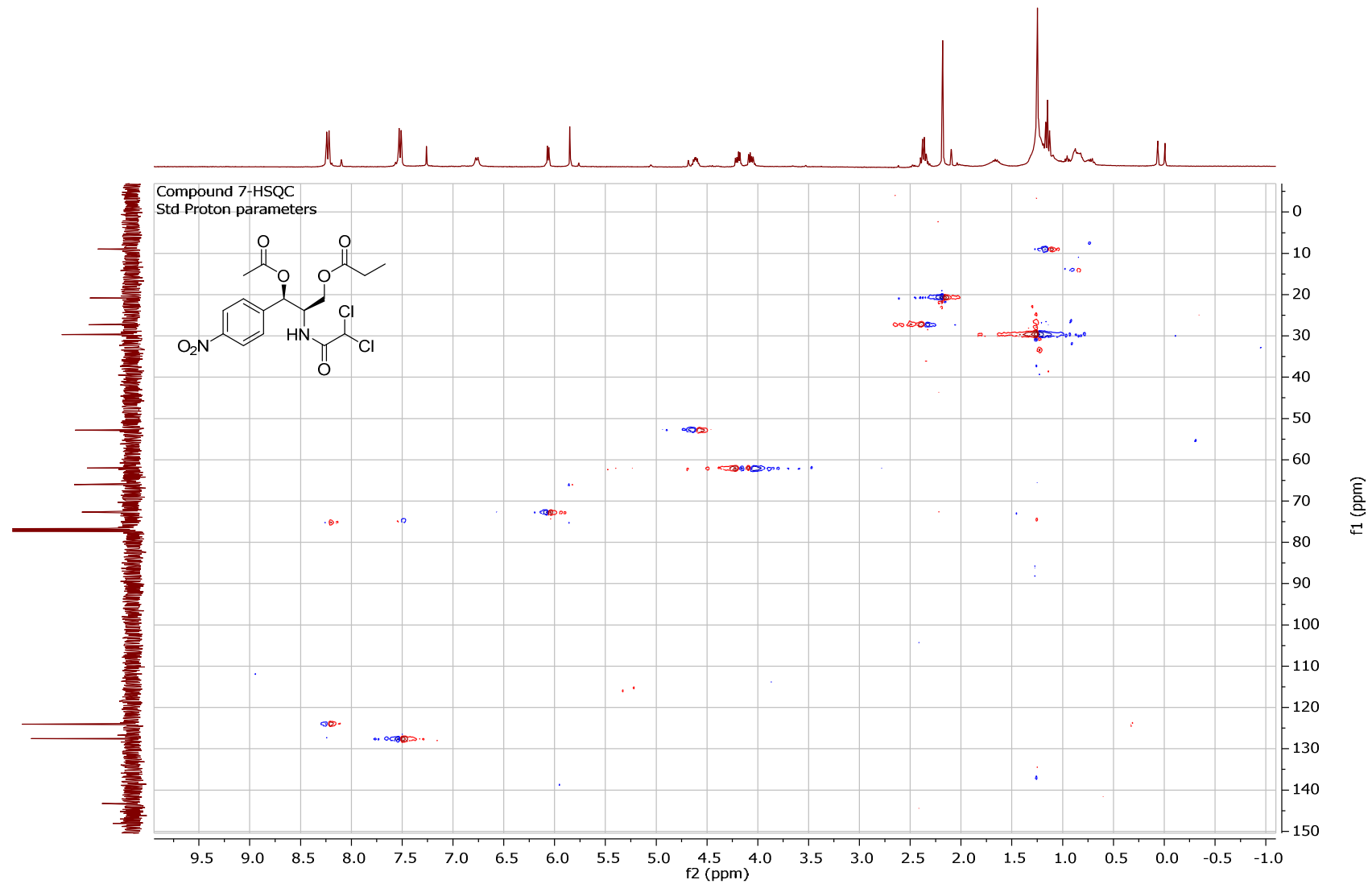


Figure S34. HSQC Spectrum of Compound 7 in CDCl_3

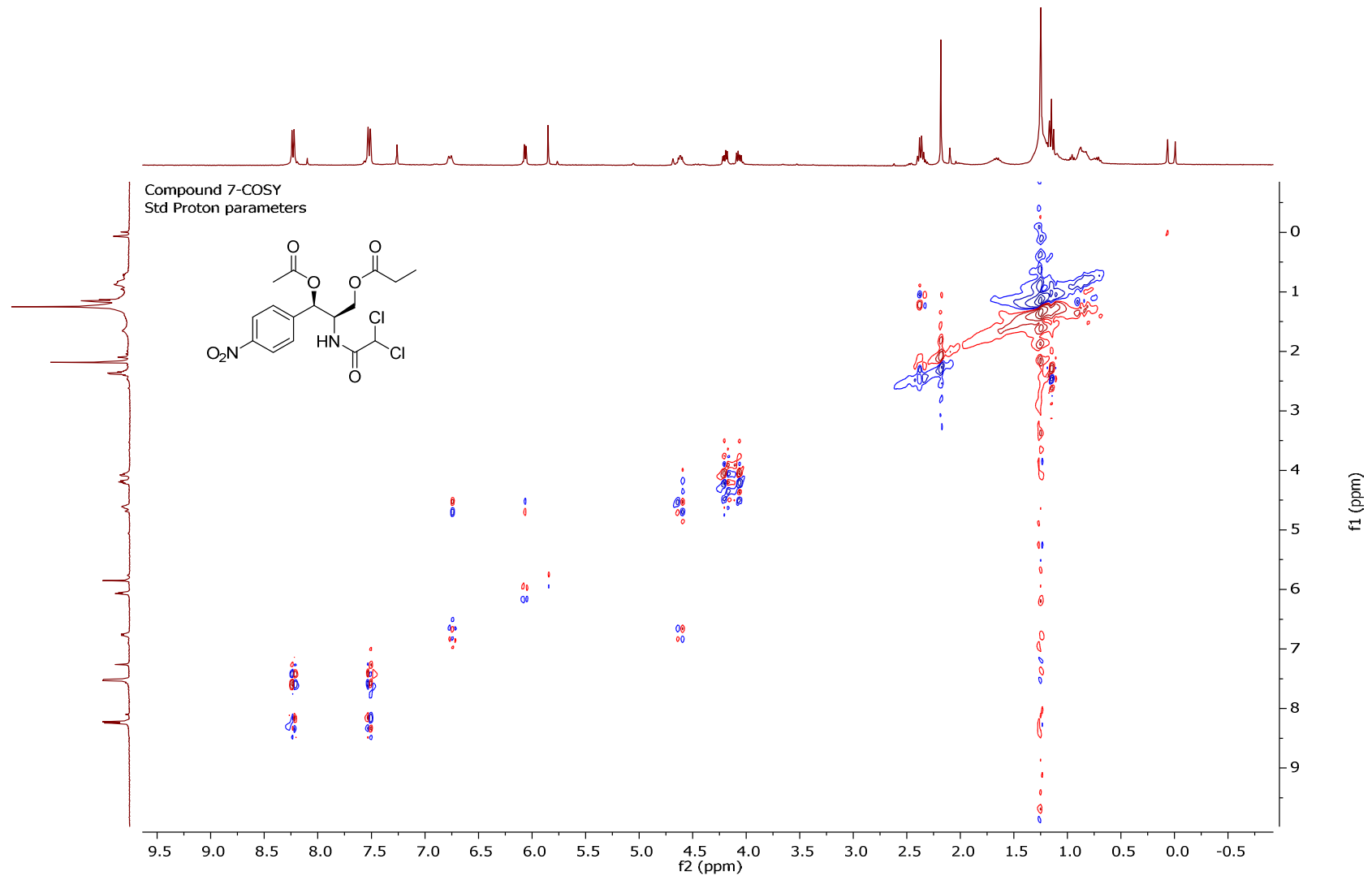


Figure S35. ^1H - ^1H COSY of Compound 7

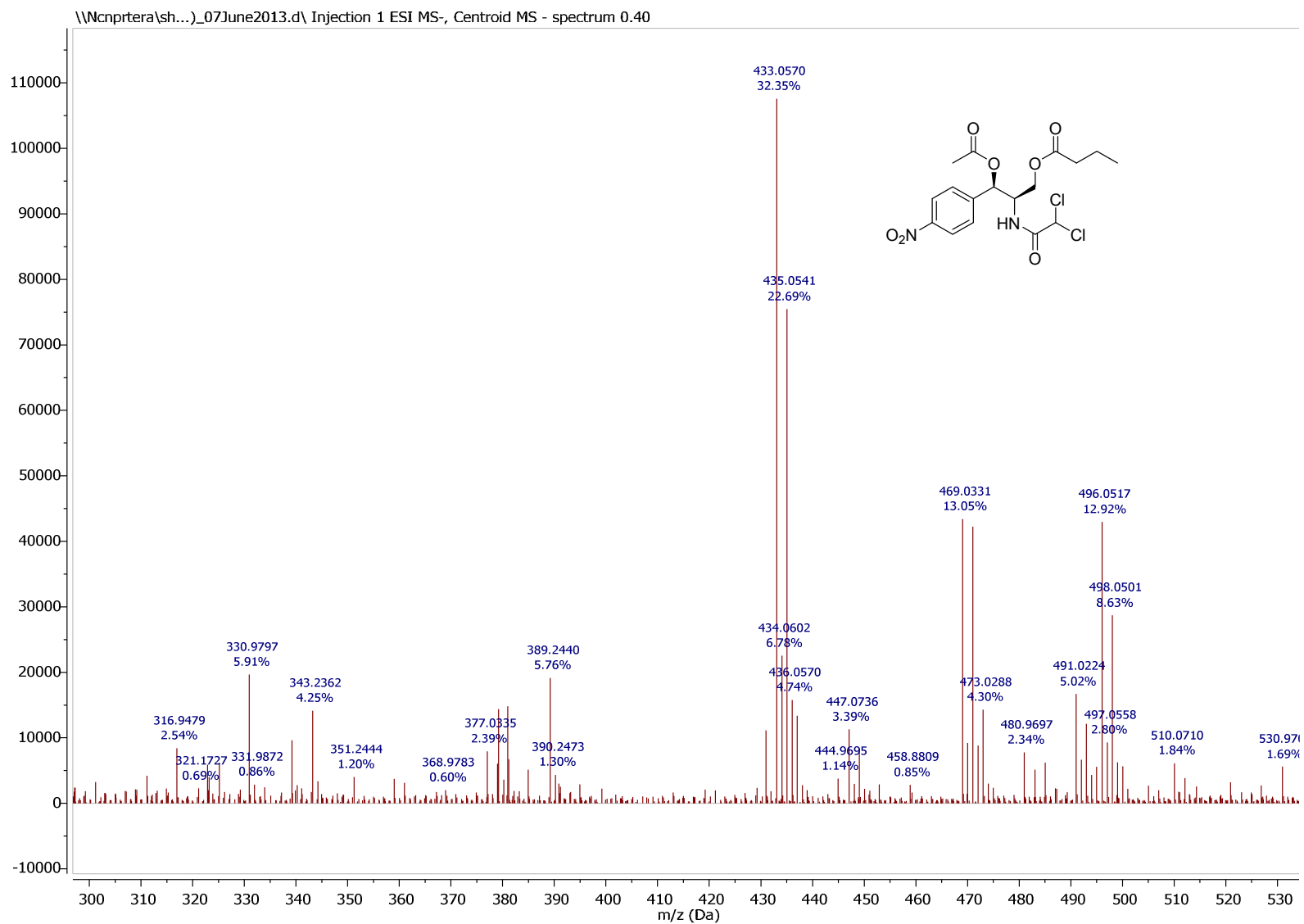


Figure S36. HRESI-MS Spectrum of Compound 8

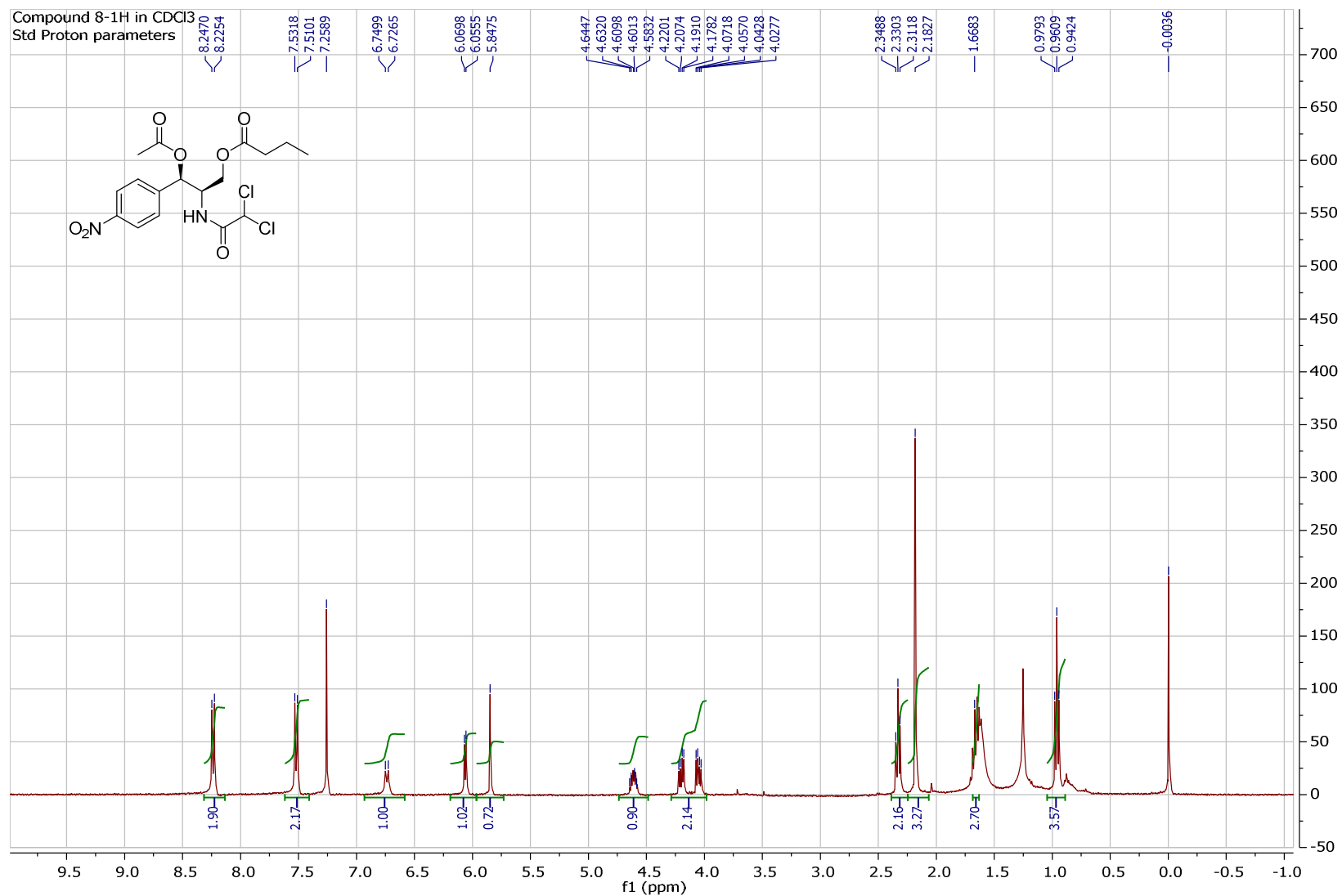


Figure S37. ¹H NMR Spectrum of Compound 8 in CDCl₃

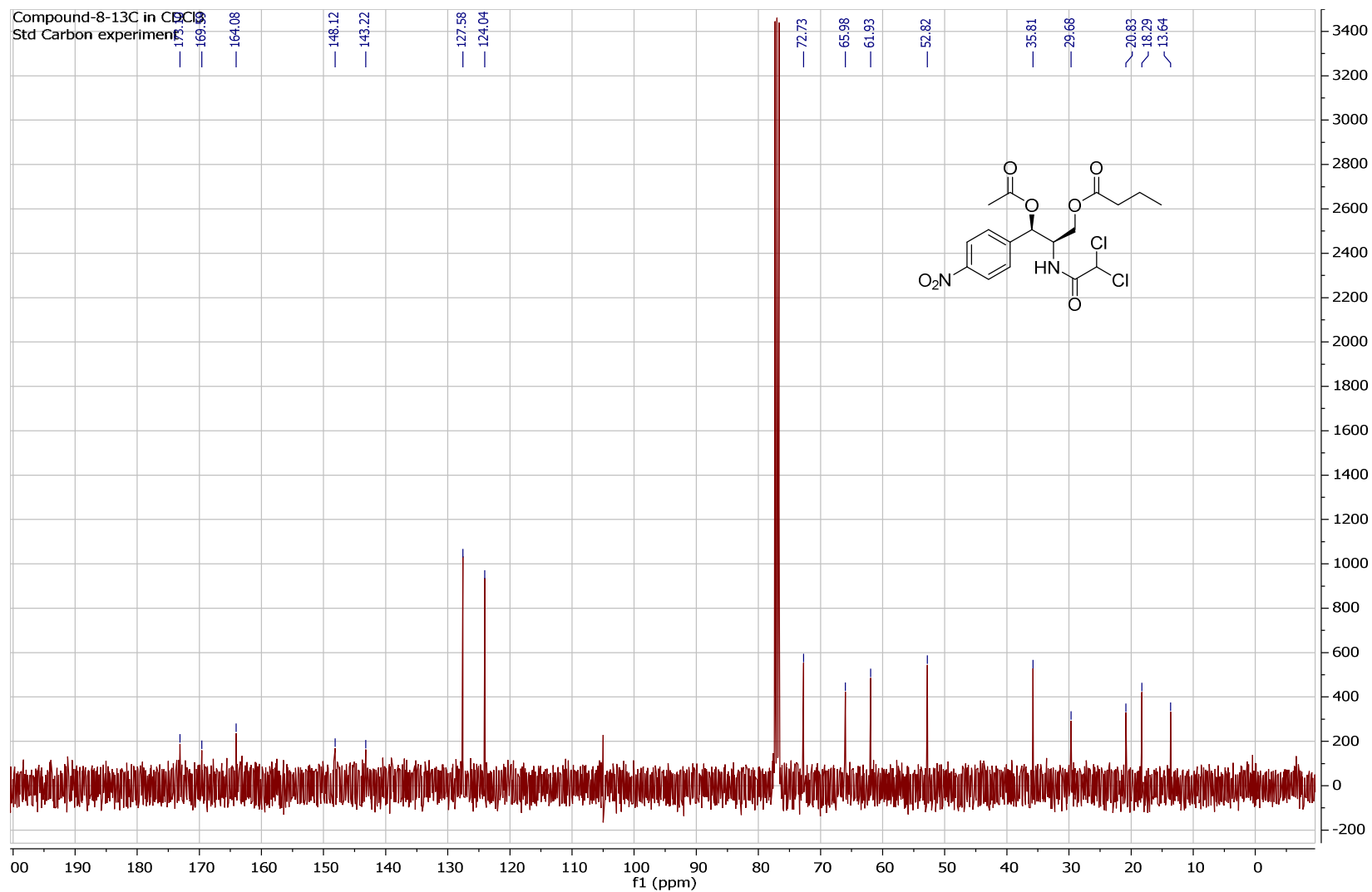


Figure S38. ¹³C NMR Spectrum of Compound 8 in CDCl₃

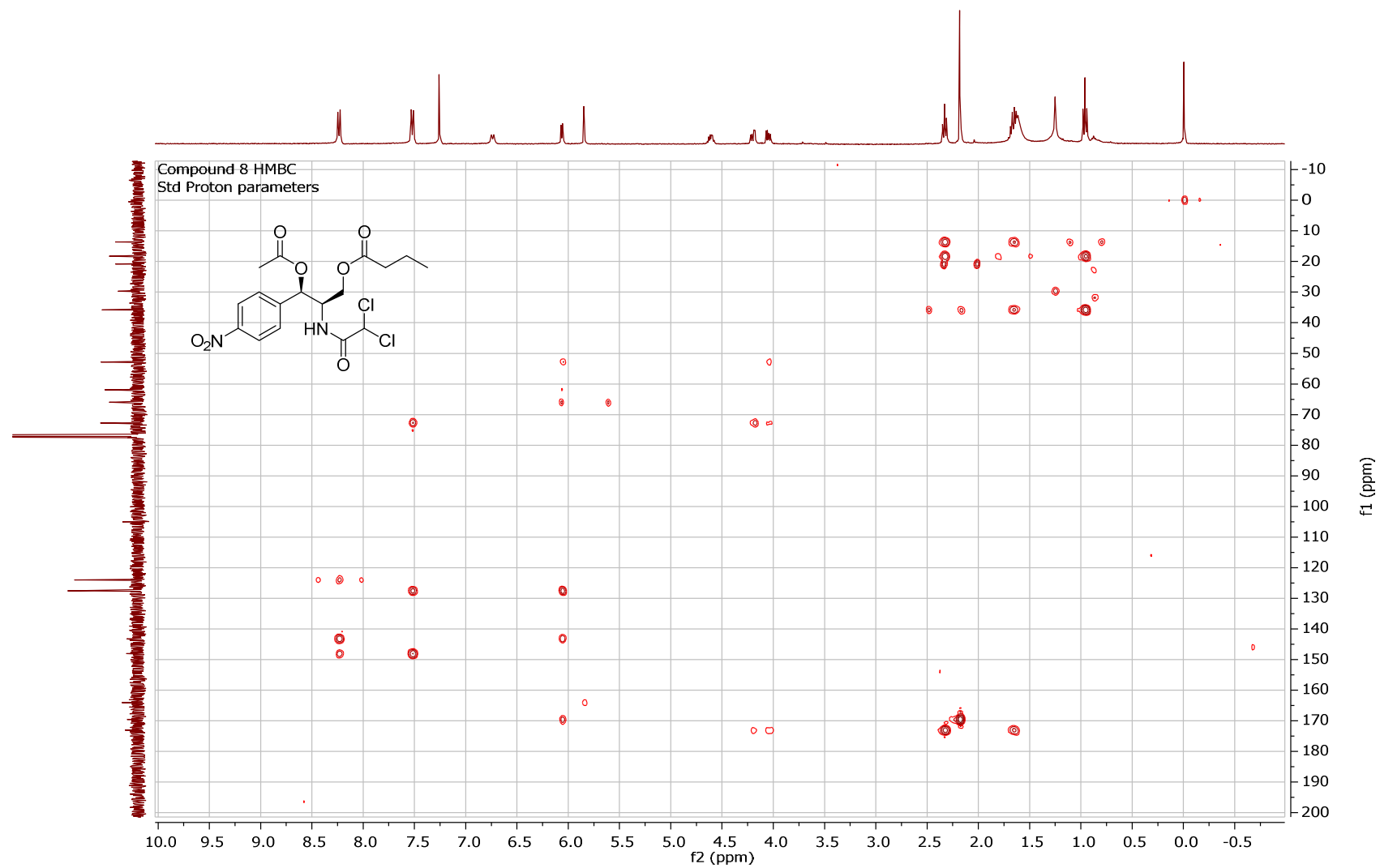


Figure S39. HMBC Spectrum of Compound 8 in CDCl_3

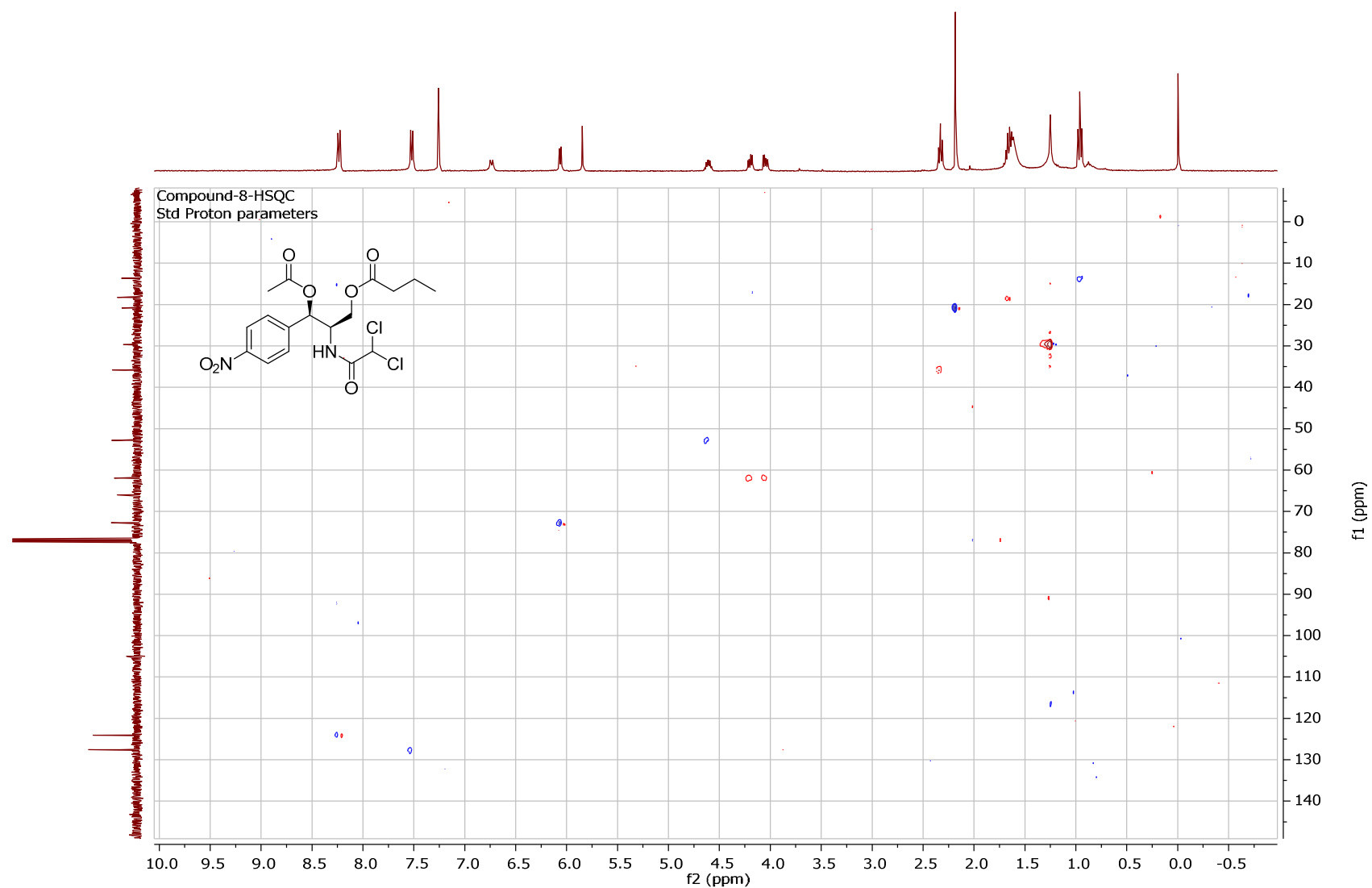


Figure S40. HSQC Spectrum of Compound 8 in CDCl₃

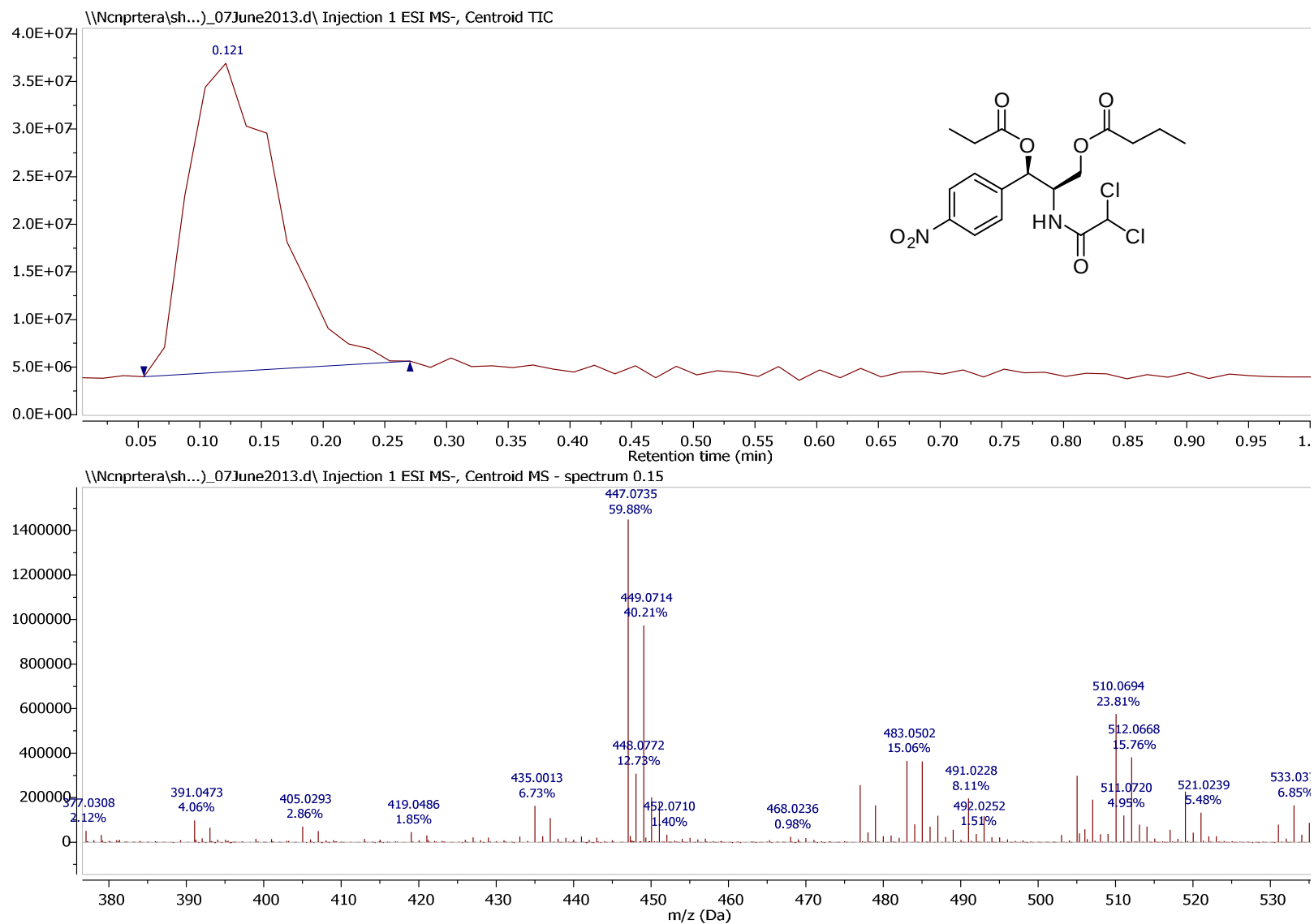


Figure S41. HRESI-MS Spectrum of Compound 9

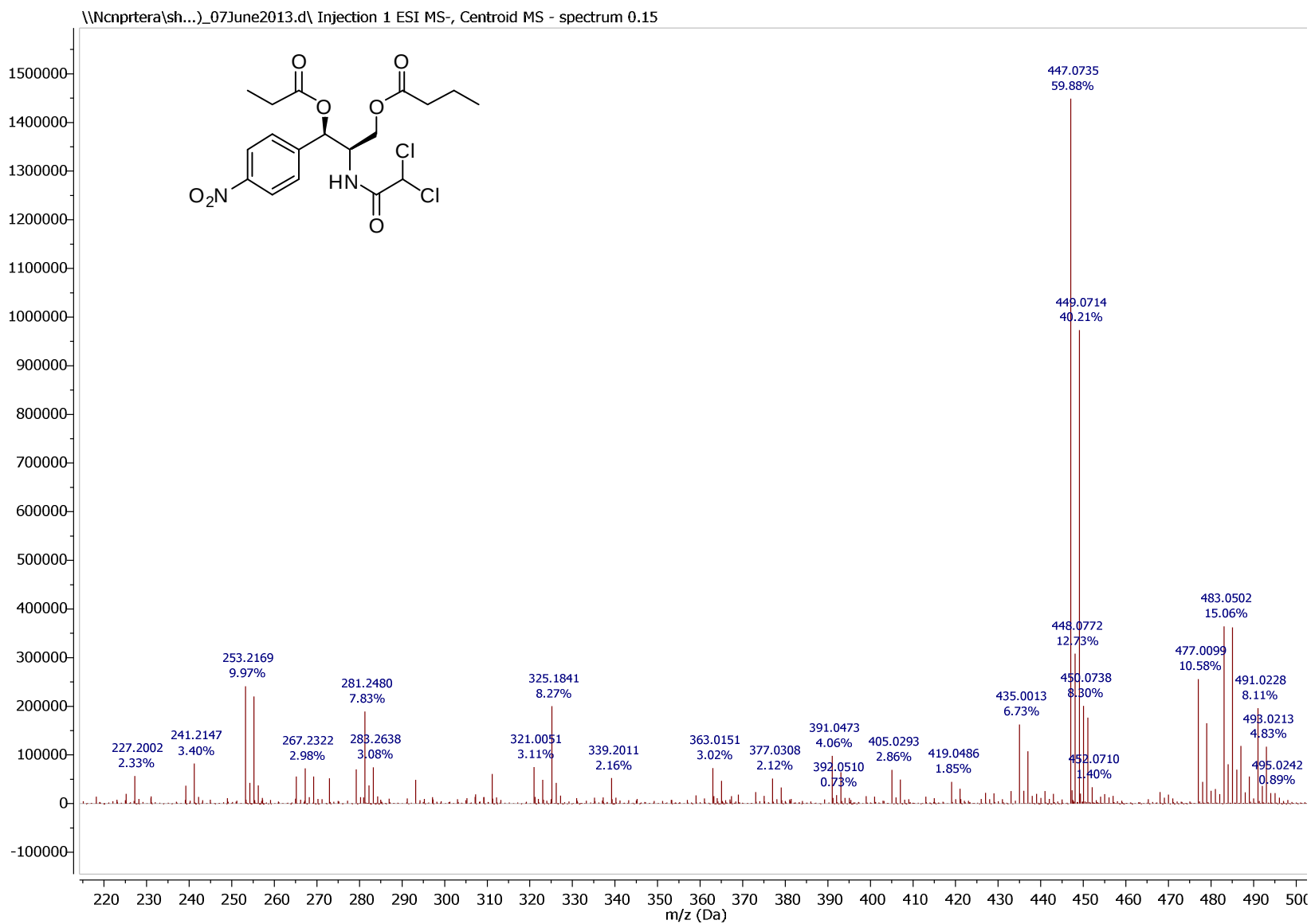


Figure S42. HRESI-MS Spectrum of Compound 9

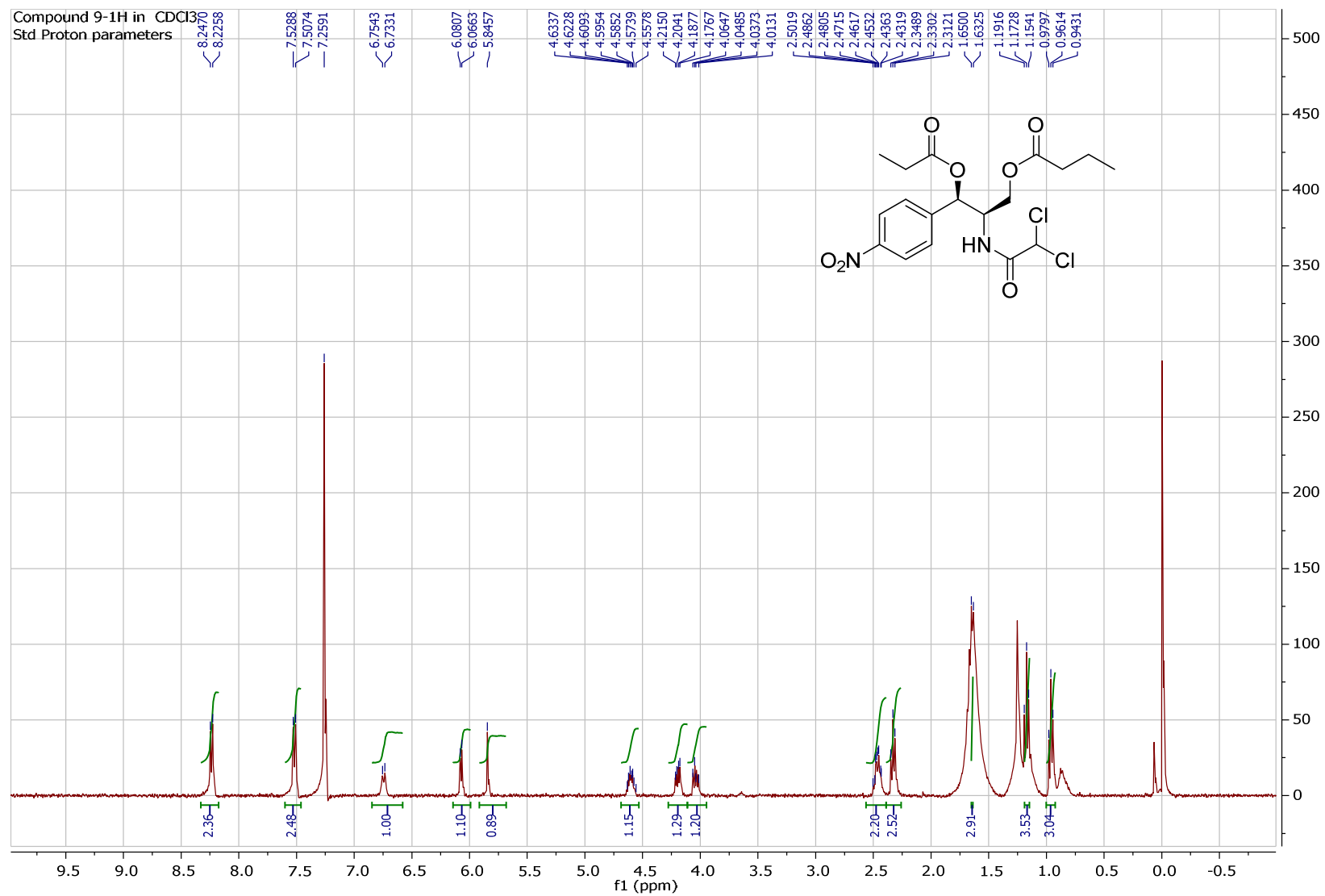


Figure S43. ¹H NMR Spectrum of Compound 9 in CDCl₃

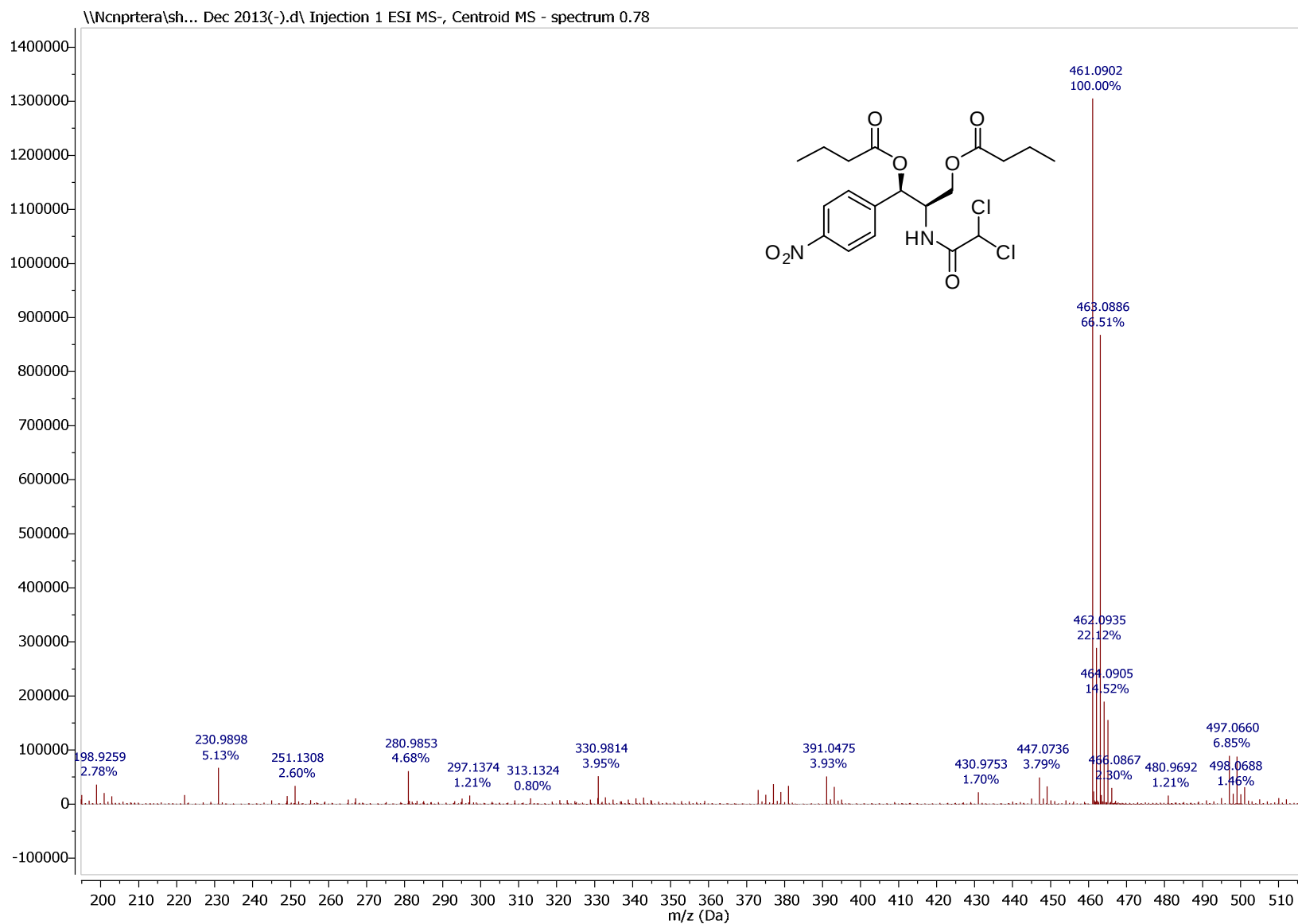


Figure S44. Negative HRESI-MS Spectrum of Compound 10

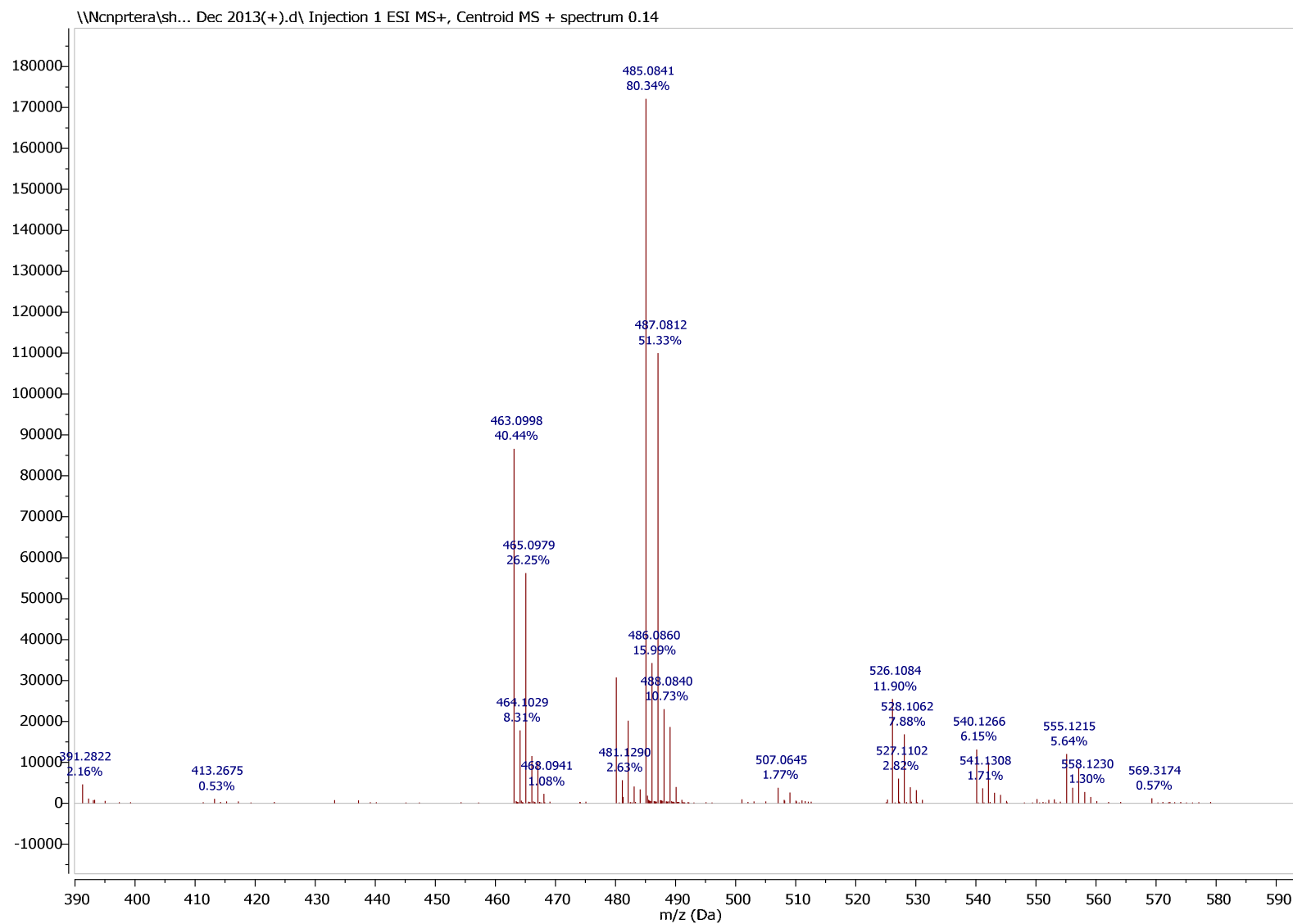


Figure S45. Positive HRESI-MS Spectrum of Compound 10

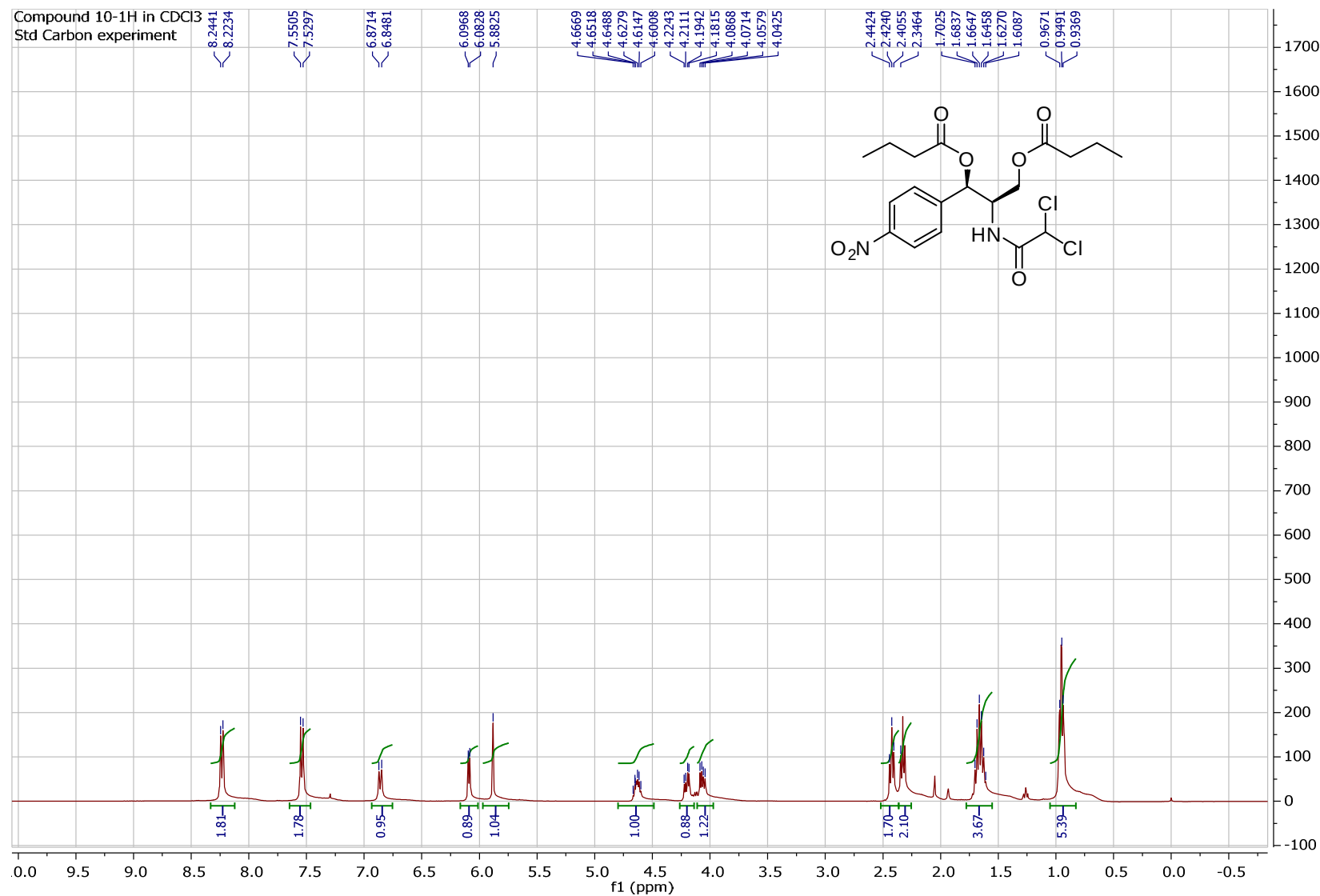


Figure S46. ¹H NMR Spectrum of Compound 10 in CDCl₃

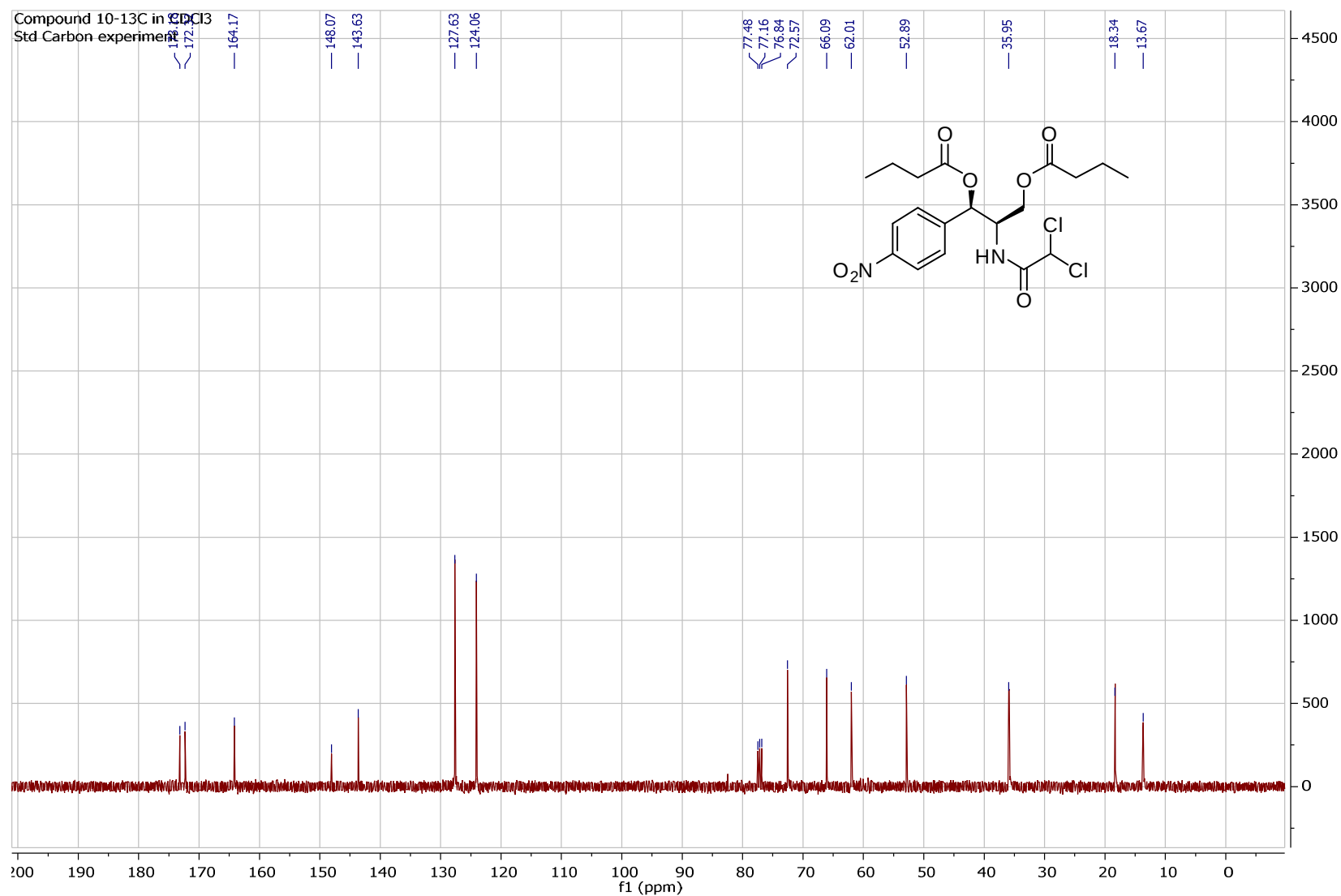


Figure S47. ¹³C NMR Spectrum of Compound 10 in CDCl₃

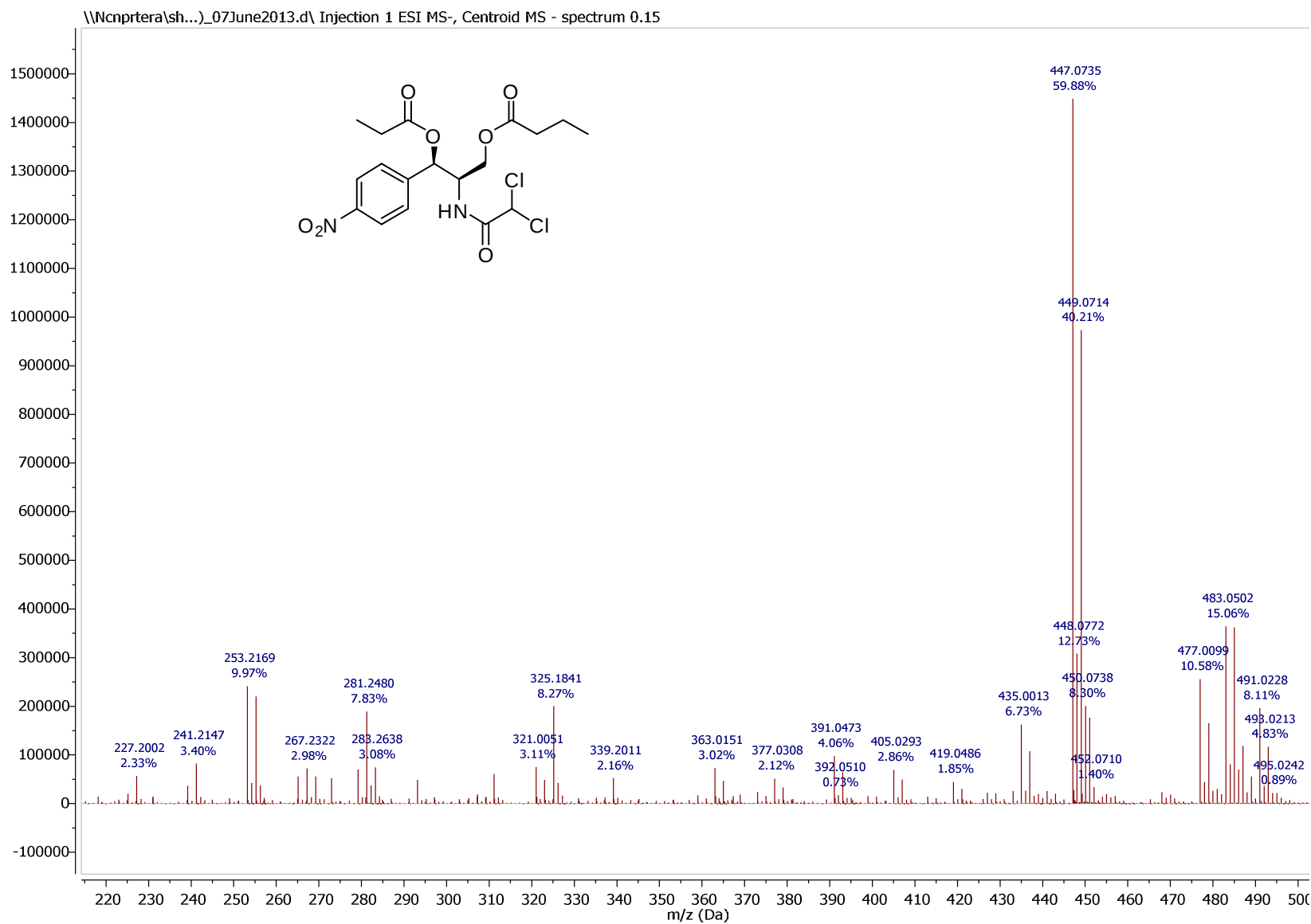


Figure S48. HRESI-MS Spectrum of Compound 11

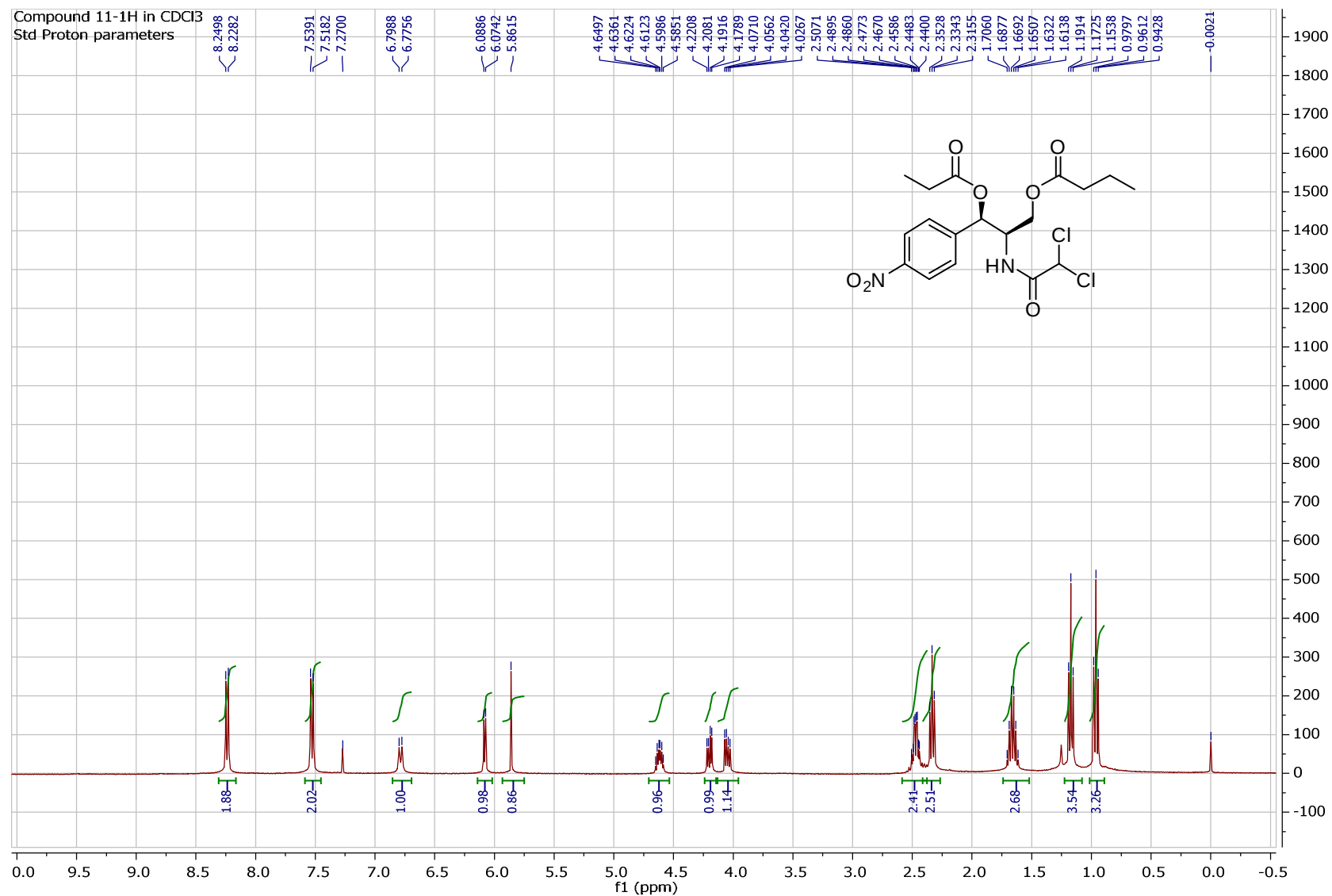


Figure S49. ¹H NMR Spectrum of Compound 11 in CDCl₃

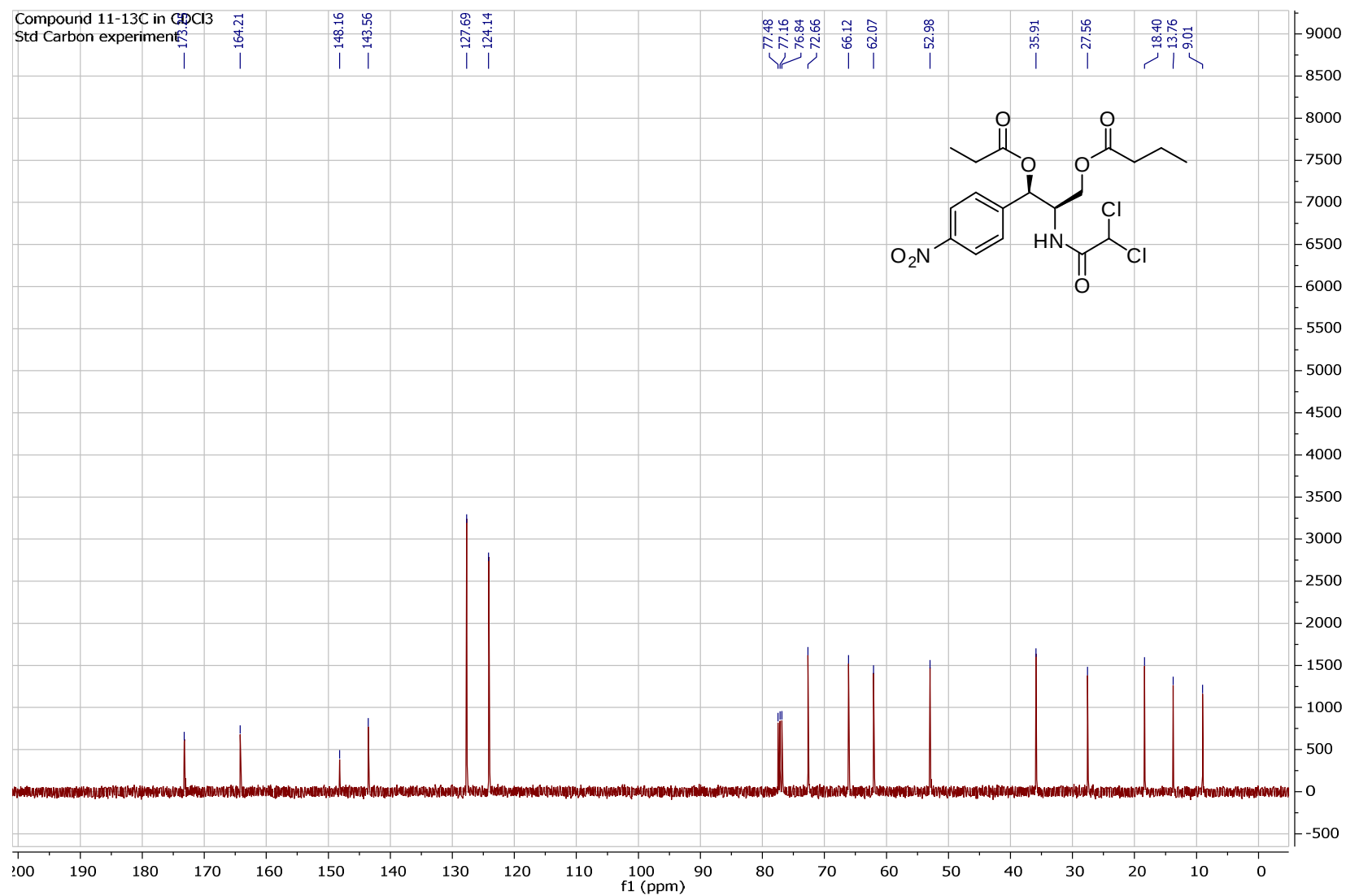


Figure S50. ¹³C NMR Spectrum of Compound 11 in CDCl₃

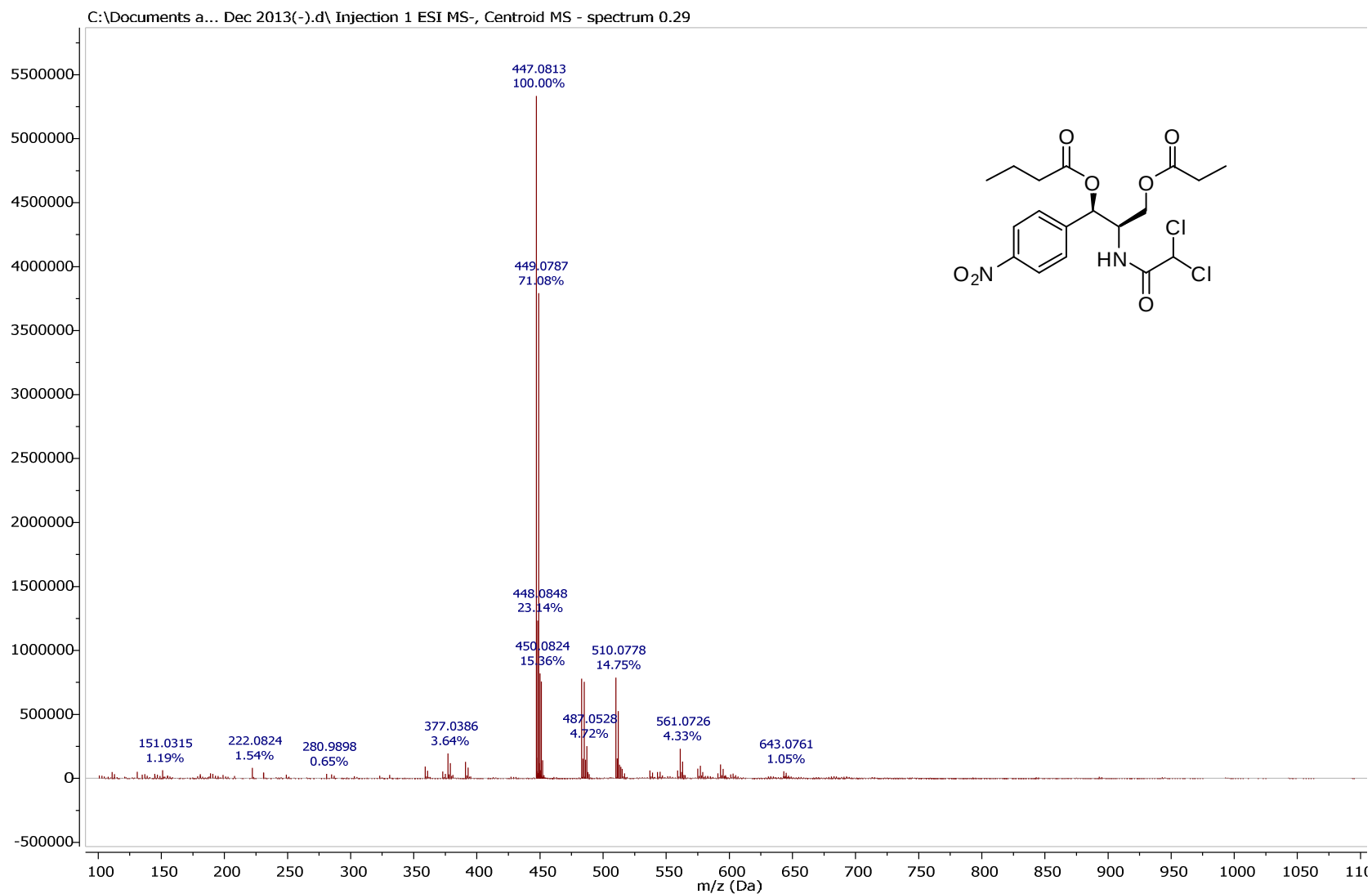


Figure S51. HRESI-MS Spectrum of Compound 12

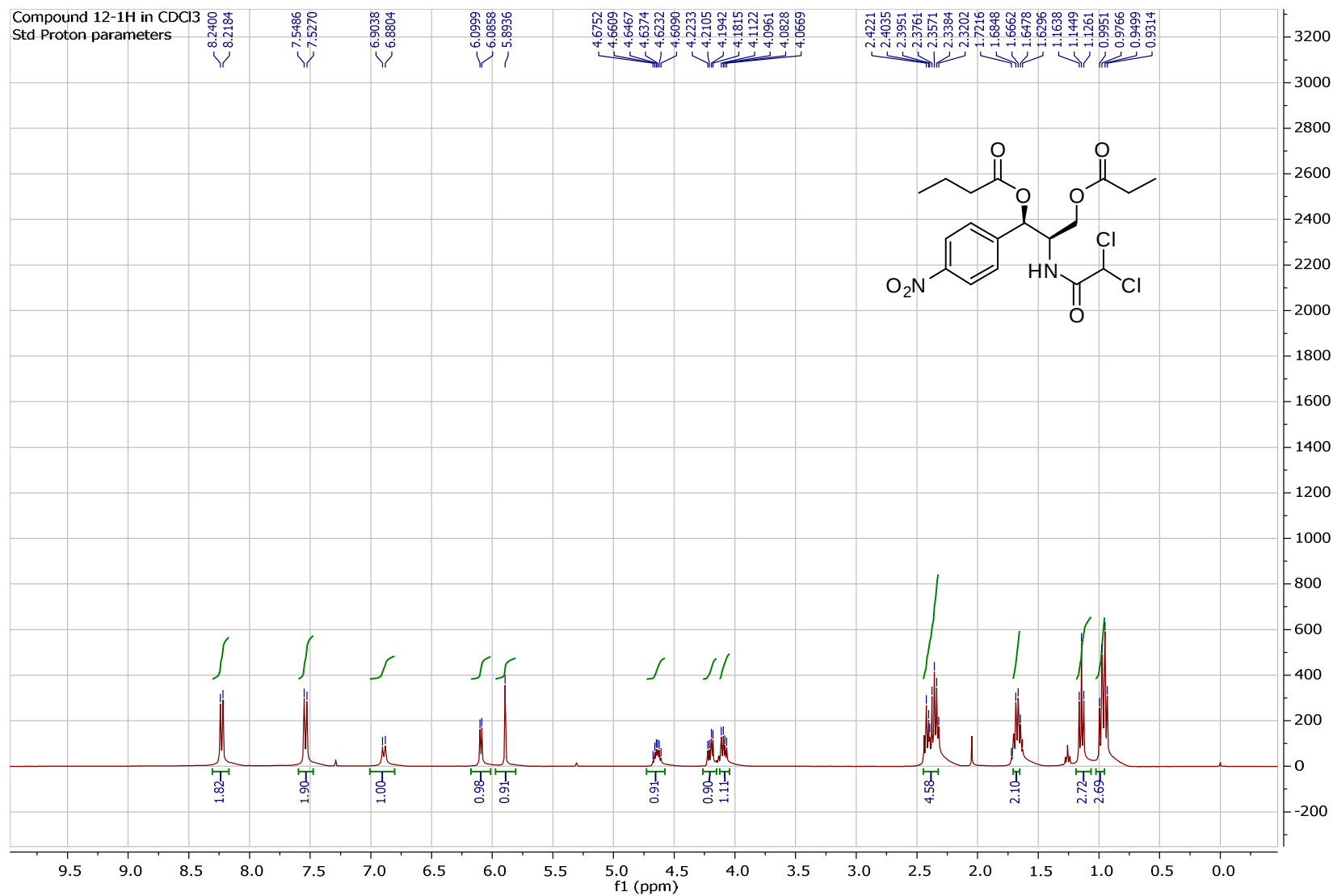


Figure S52. ¹H NMR Spectrum of Compound 12 in CDCl₃

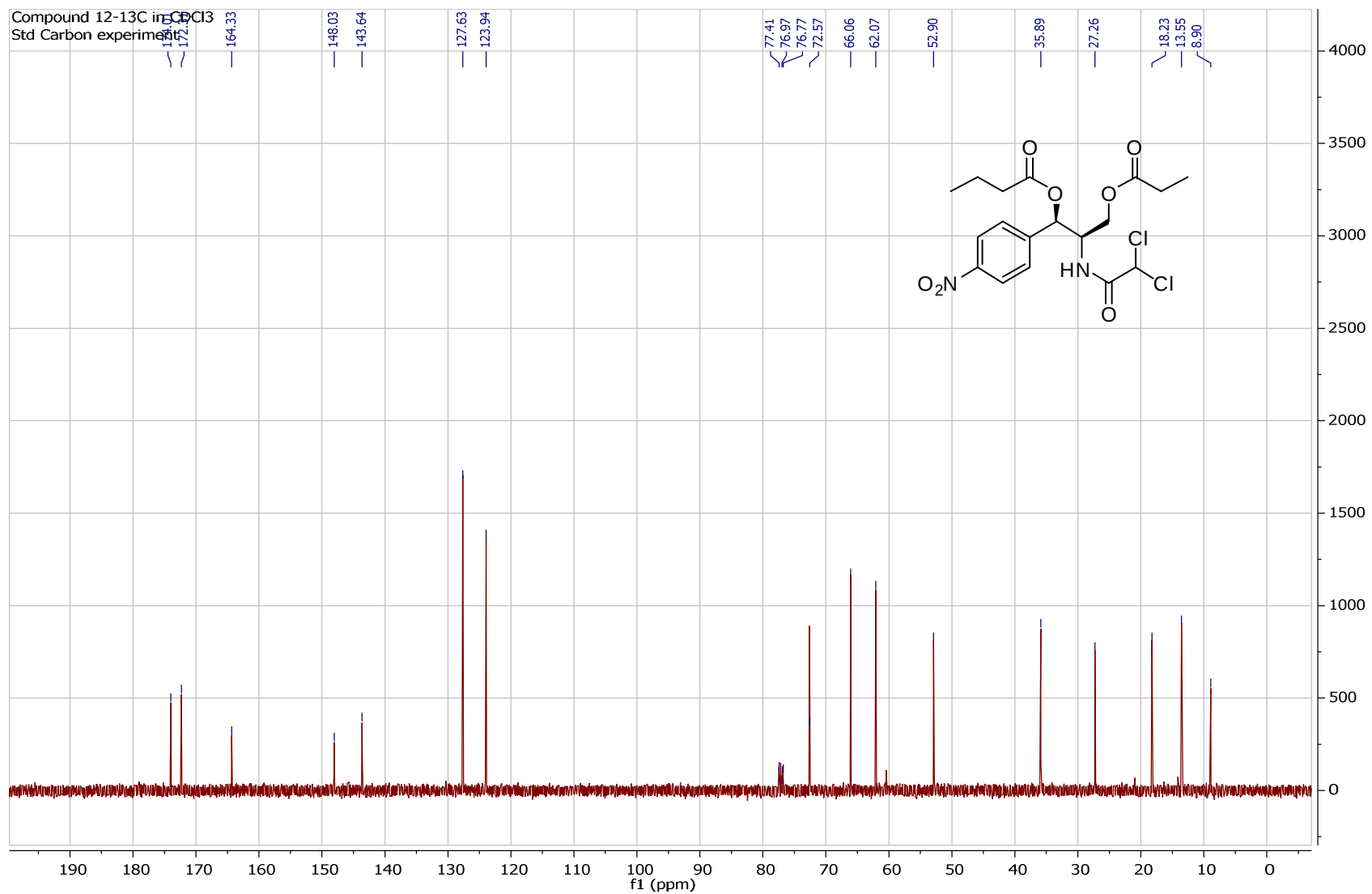


Figure S53. ¹³C NMR Spectrum of Compound 12 in CDCl₃

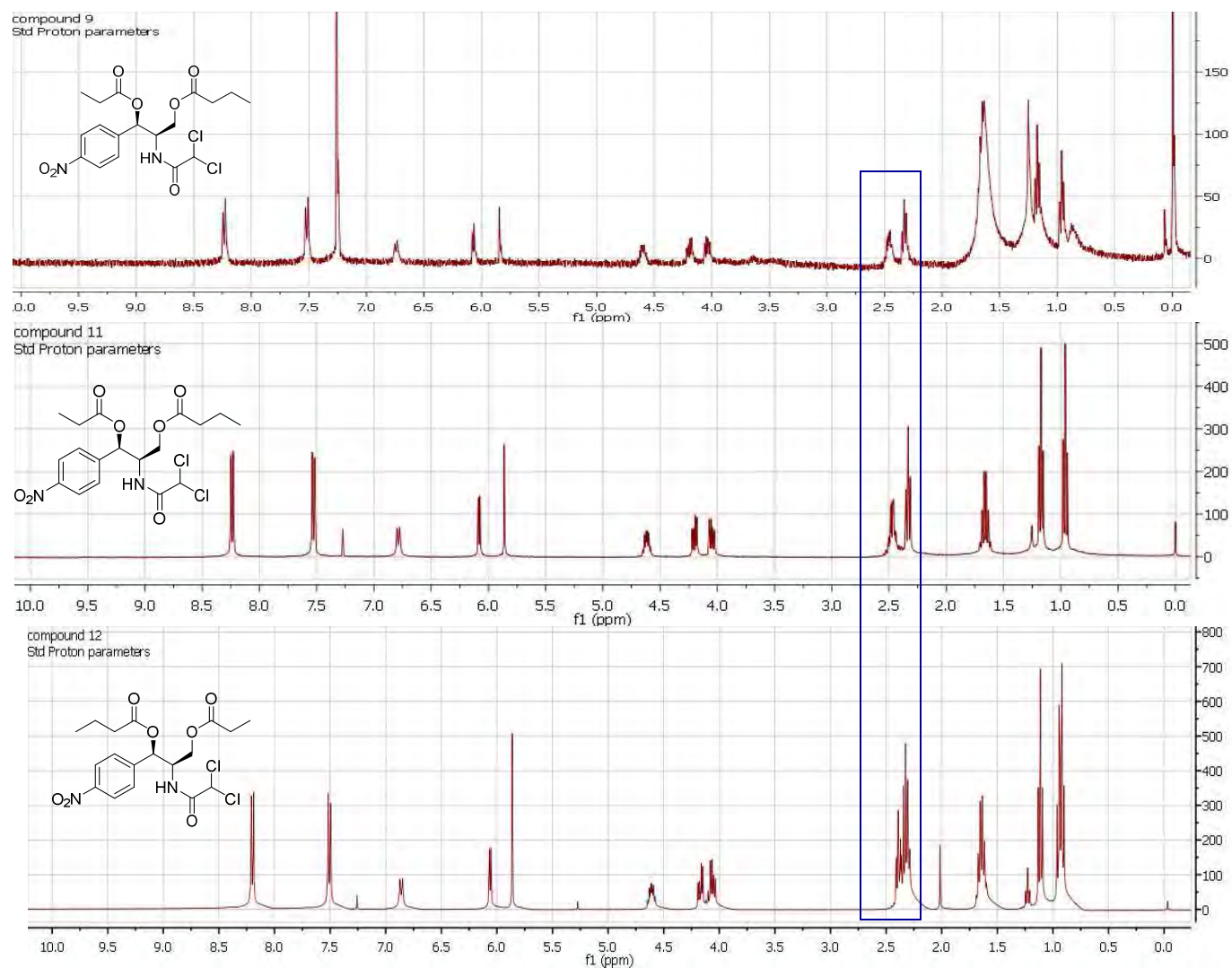
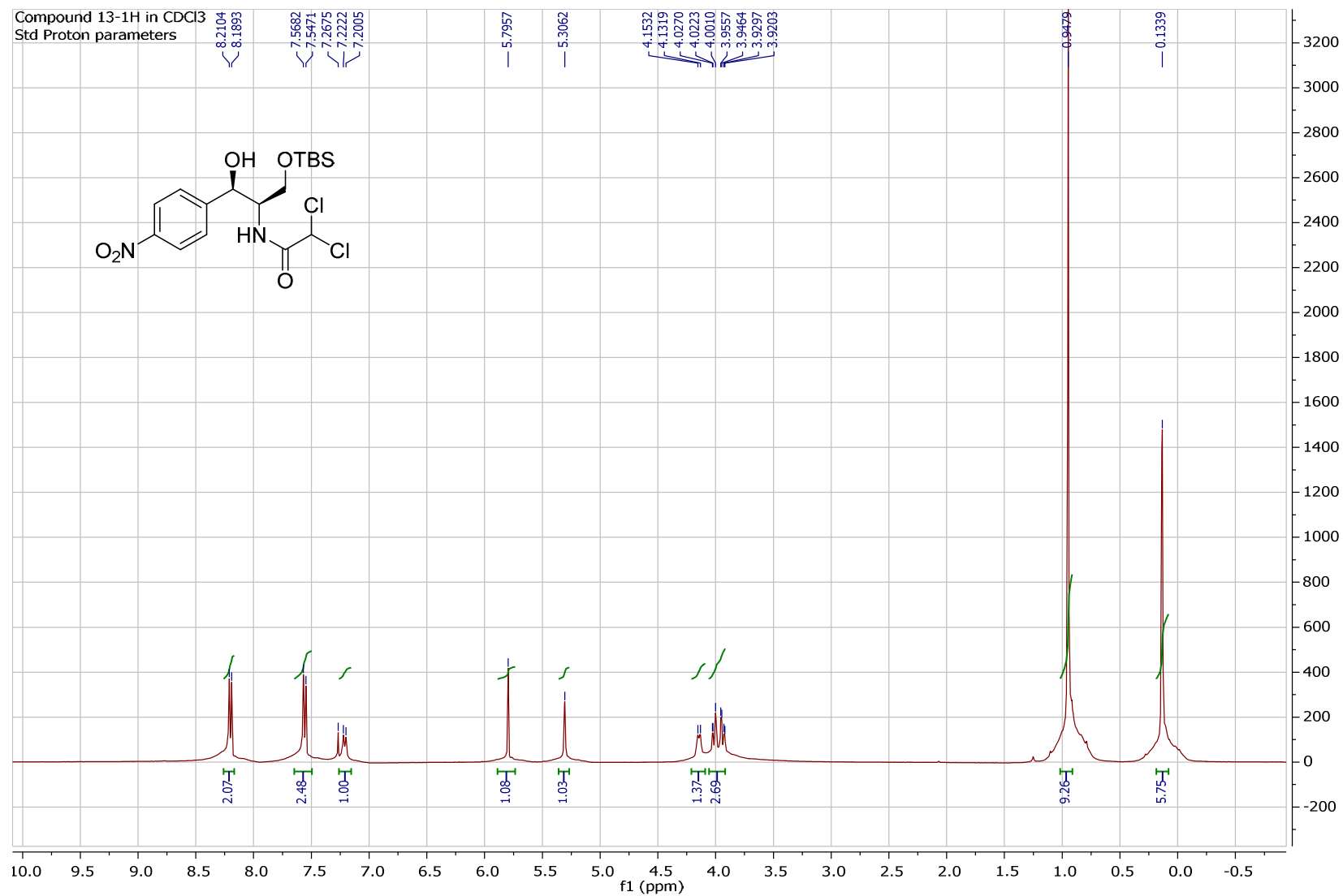


Figure S54. Comparison ^1H NMR (in CDCl_3) spectra of natural and synthetic compounds 9, 11 and 12



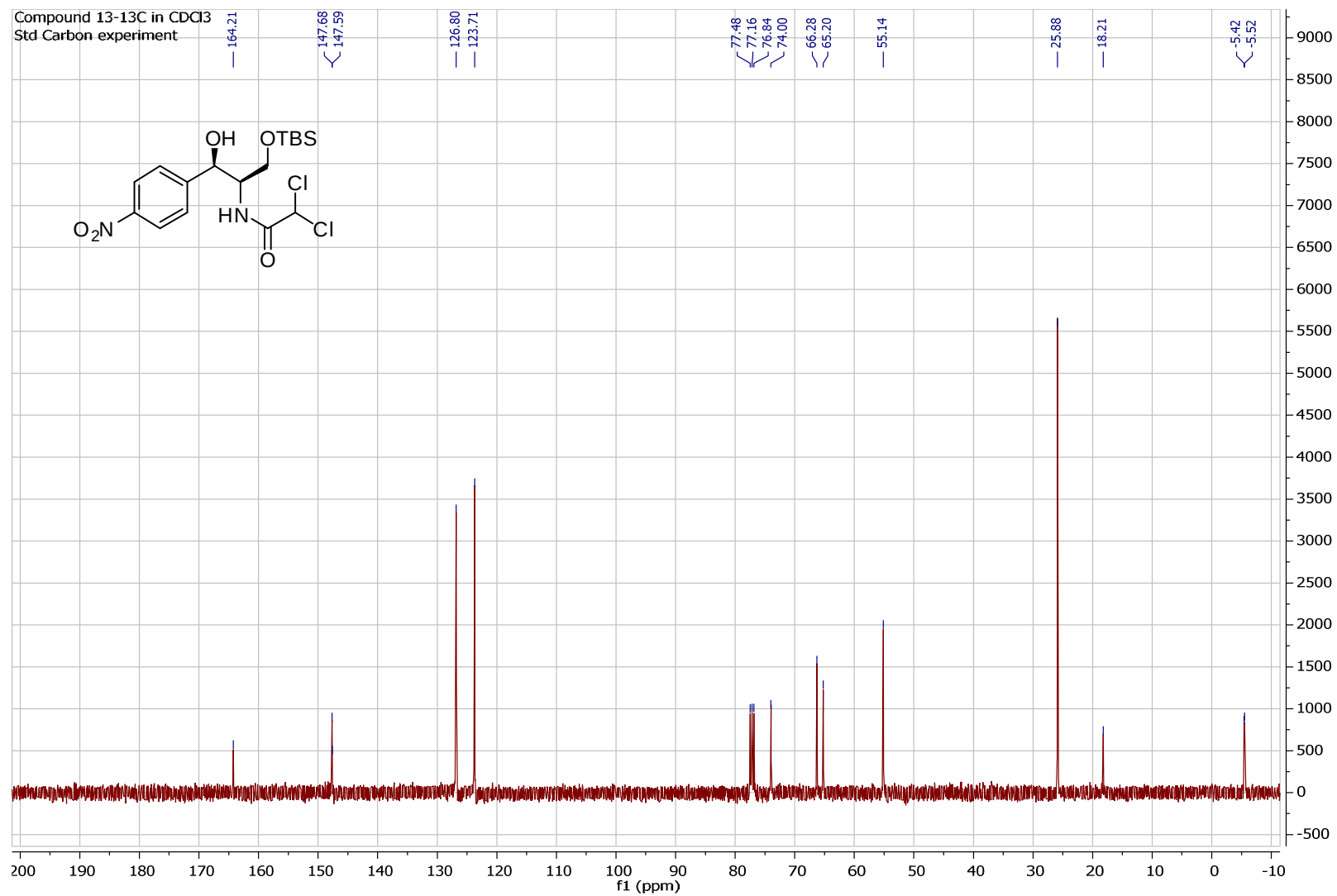


Figure S56. ¹³C NMR spectrum of Compound 13 in CDCl₃

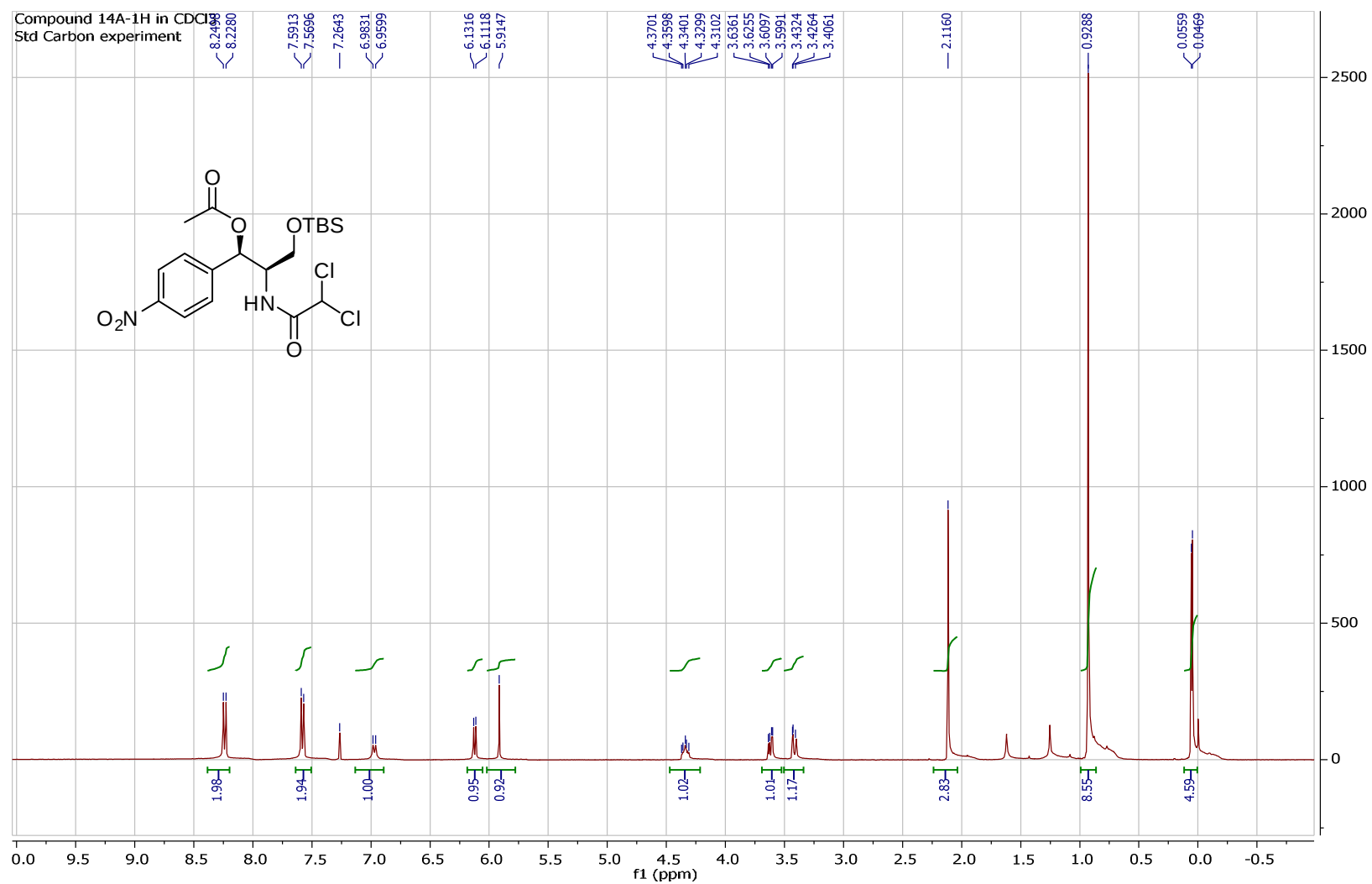


Figure S57. ¹H NMR Spectrum of Compound 14a in CDCl₃

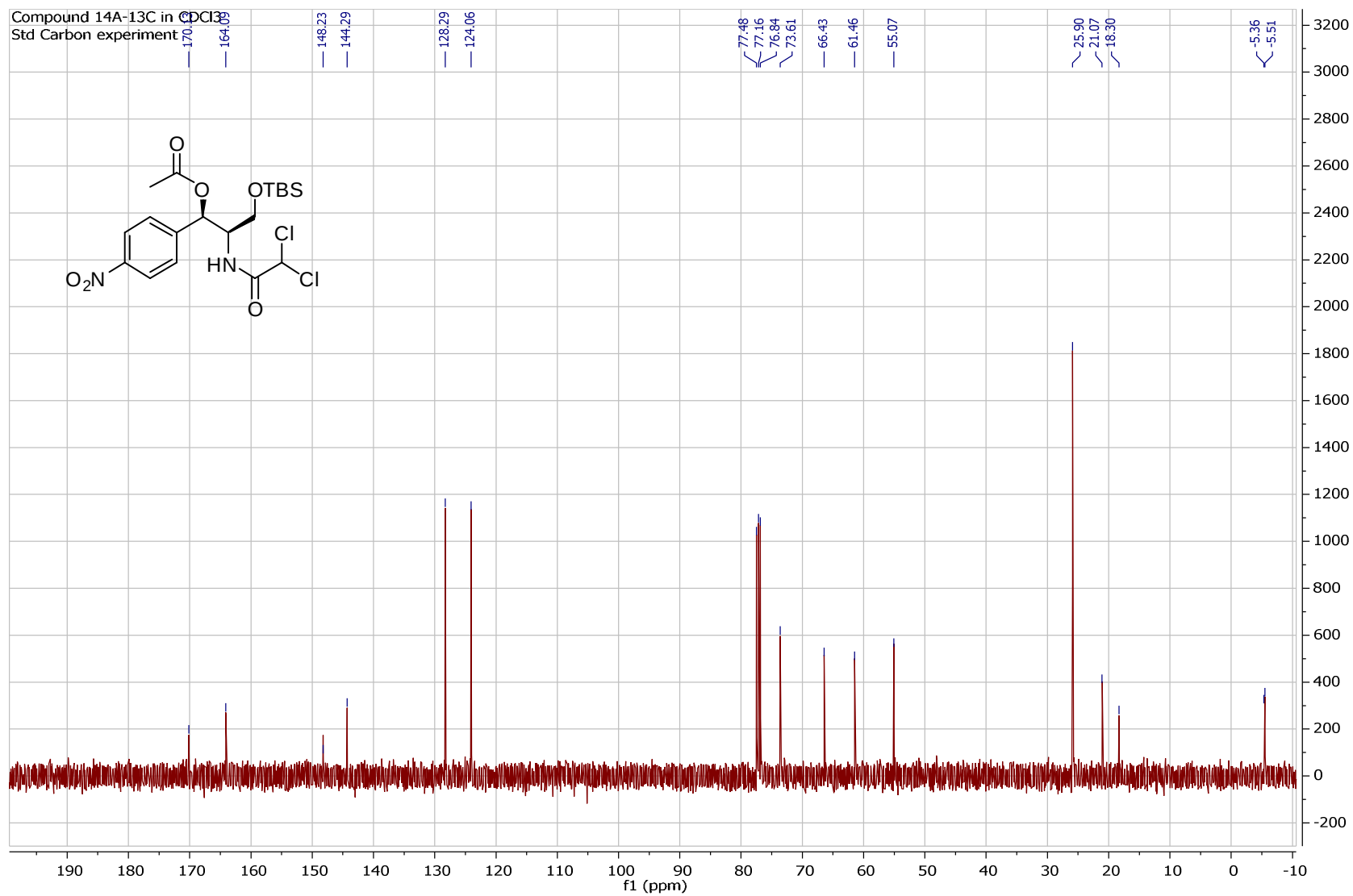


Figure S58. ¹³C NMR spectrum of Compound 14a in CDCl₃

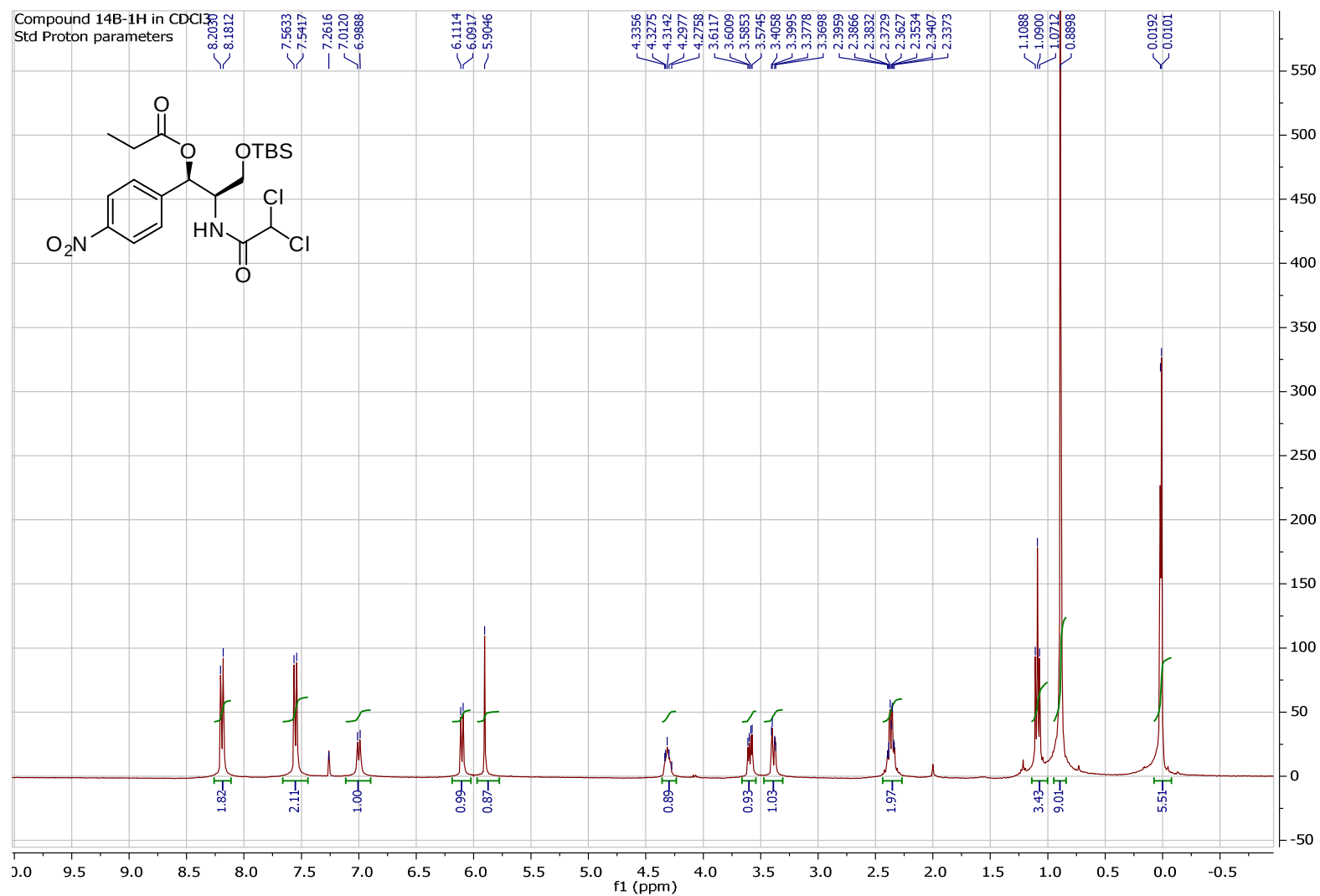


Figure S59. ¹H NMR Spectrum of Compound 14b in CDCl₃

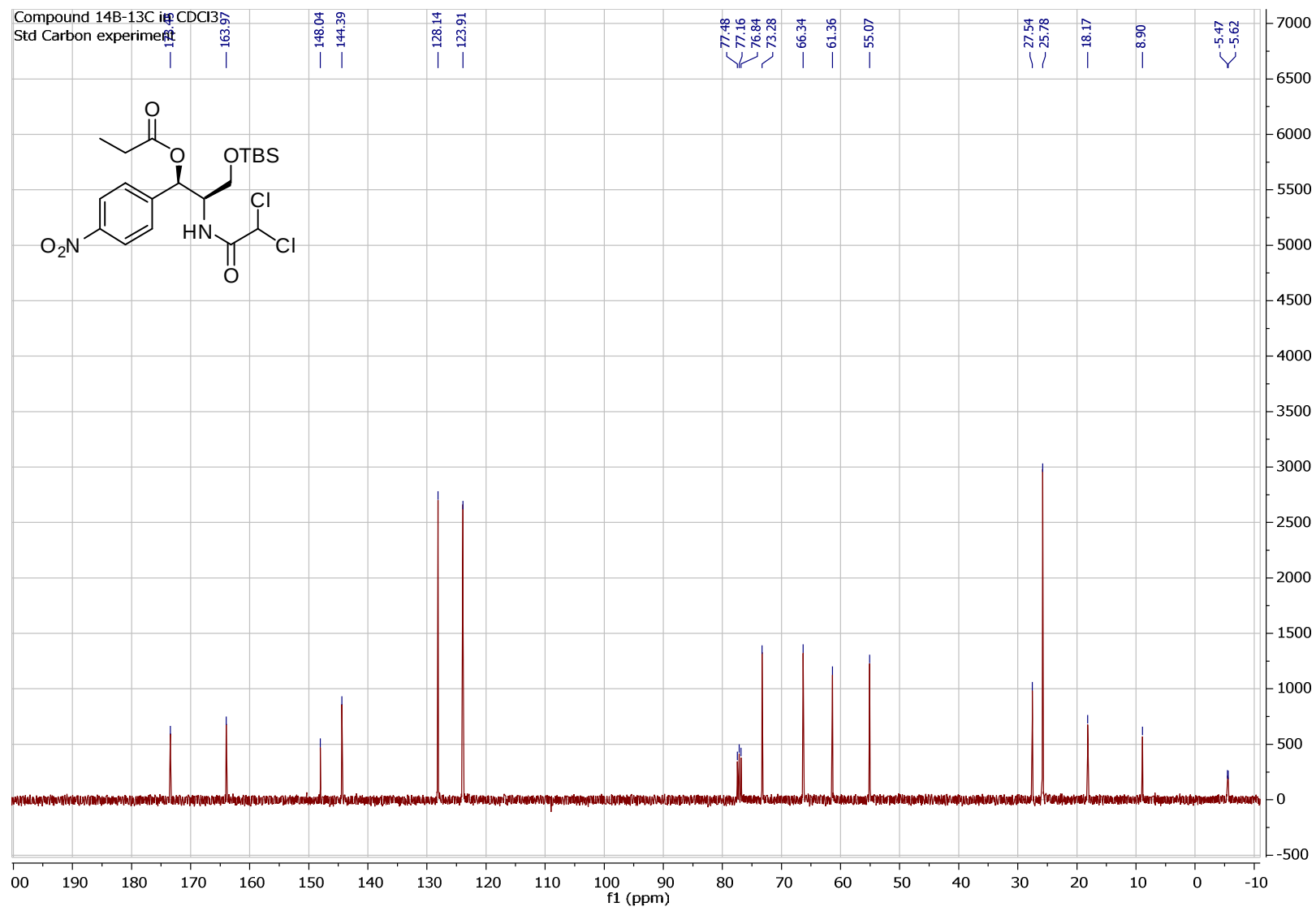


Figure S60. ¹³C NMR Spectrum of Compound 14b in CDCl₃

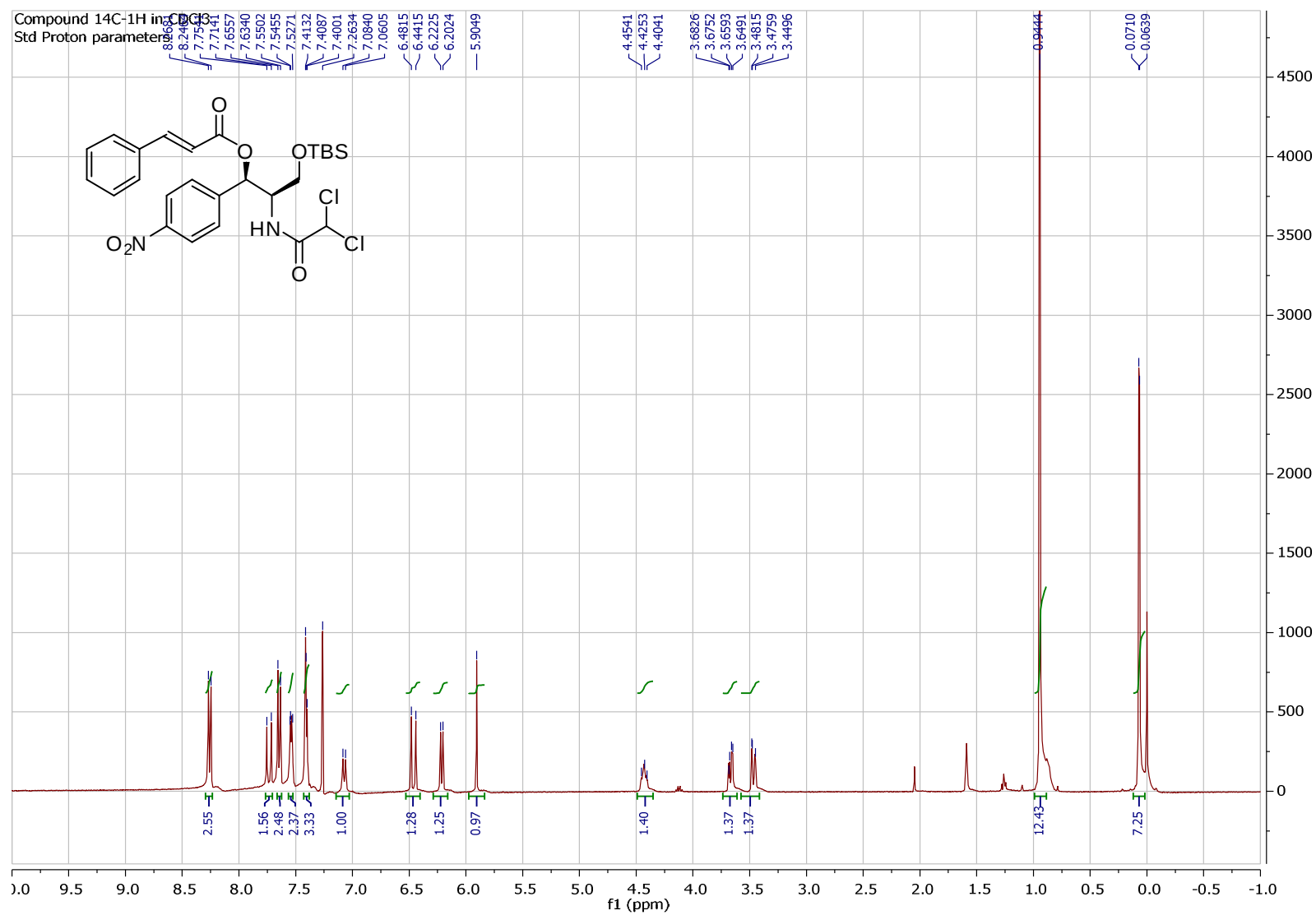


Figure S61. ¹H NMR Spectrum of Compound 14c in CDCl₃

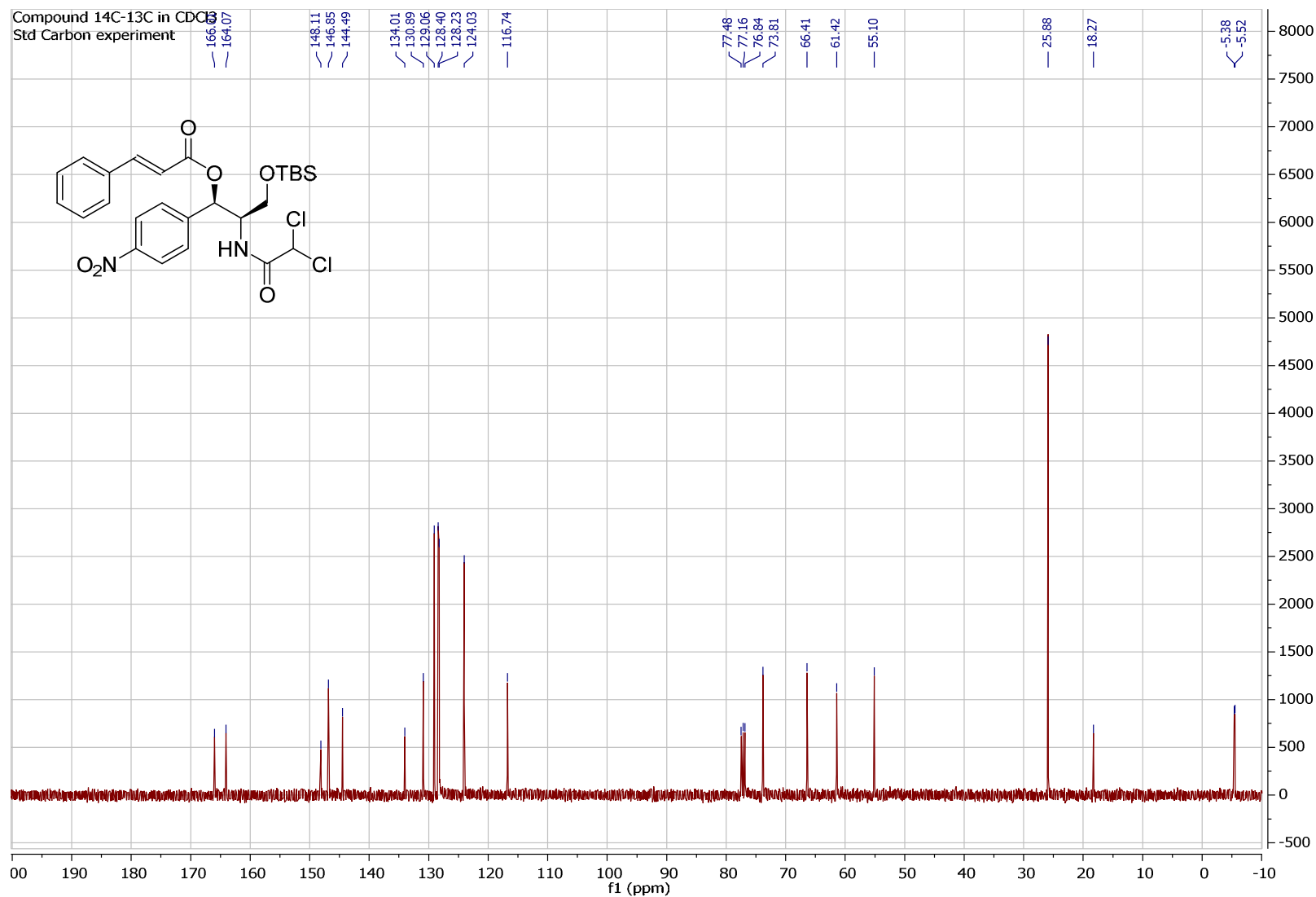


Figure S62. ¹³C NMR Spectrum of Compound 14c in CDCl₃

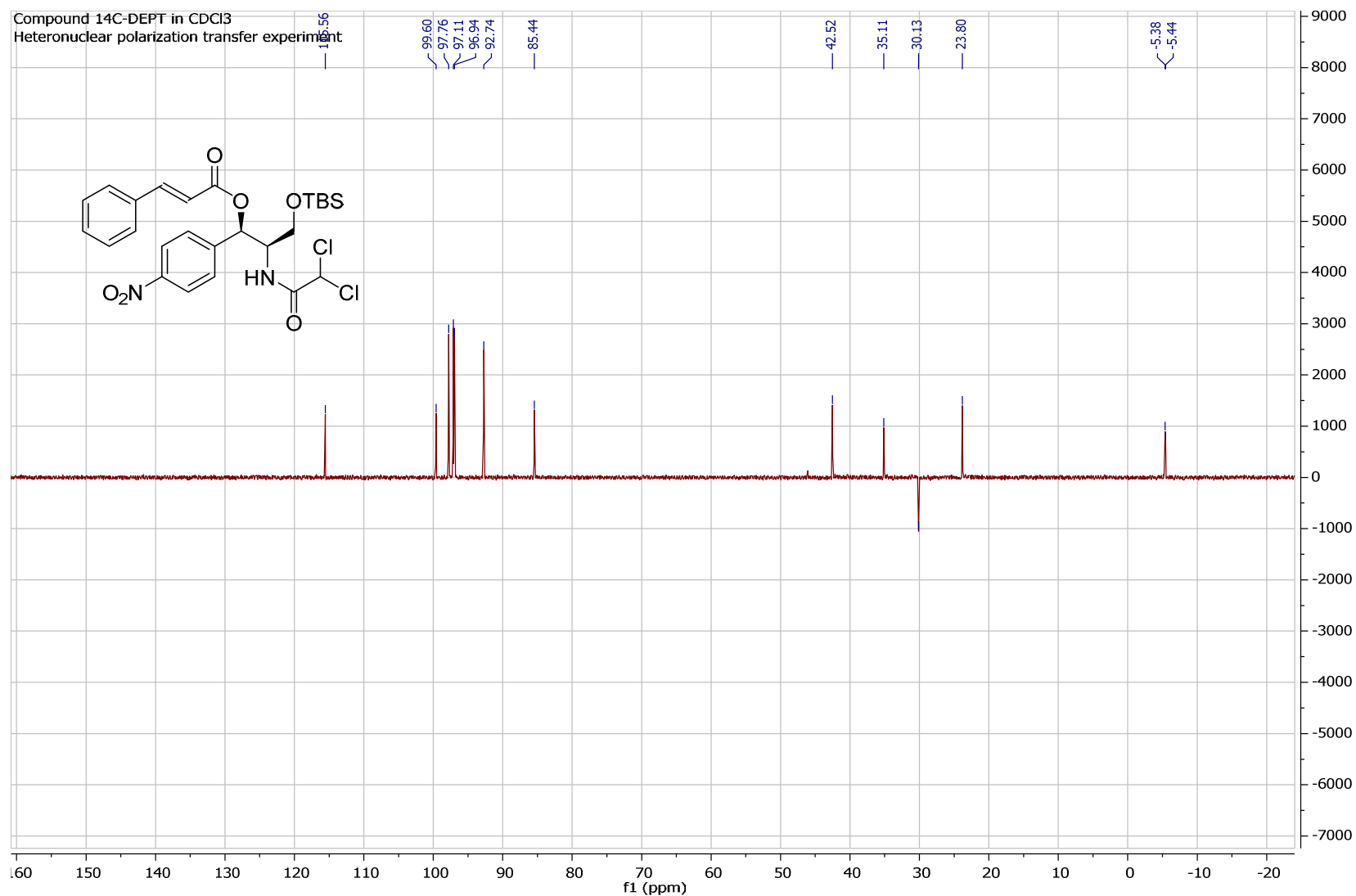


Figure S63. DEPT 135° Spectrum of Compound 14c in CDCl₃

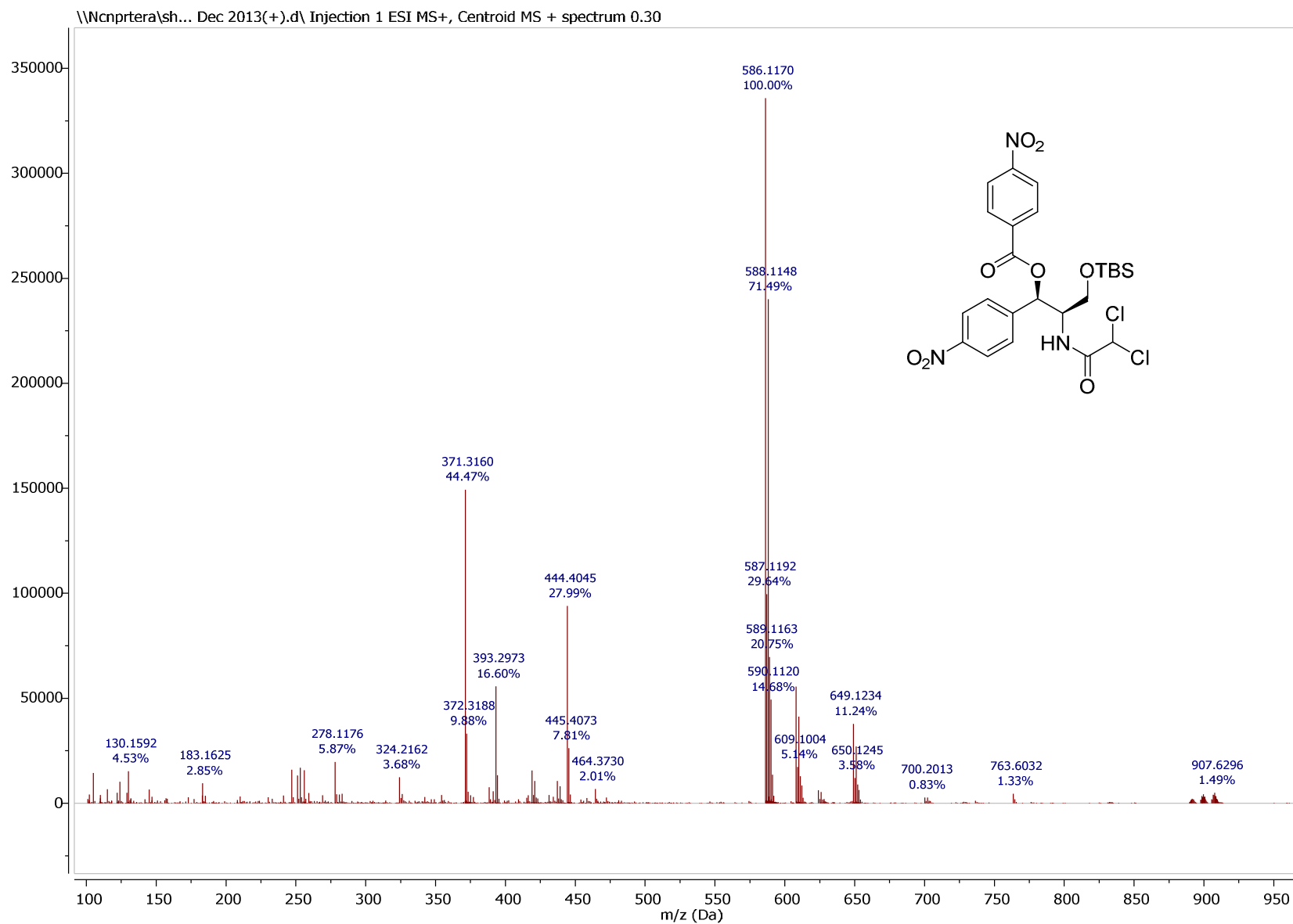


Figure S64. HRESI-MS Spectrum of Compound 14d

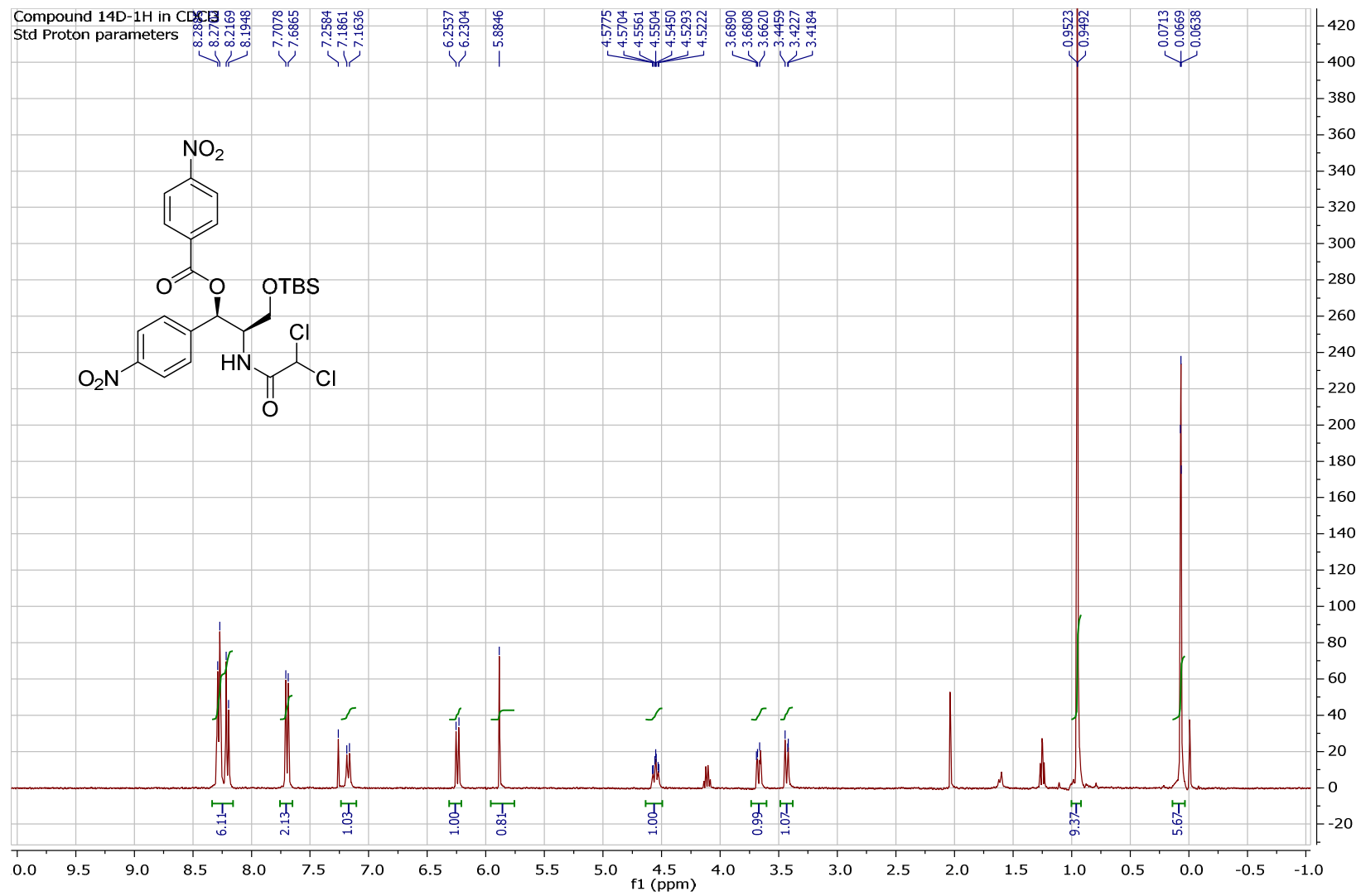


Figure S65. ¹H NMR Spectrum of Compound 14d in CDCl₃

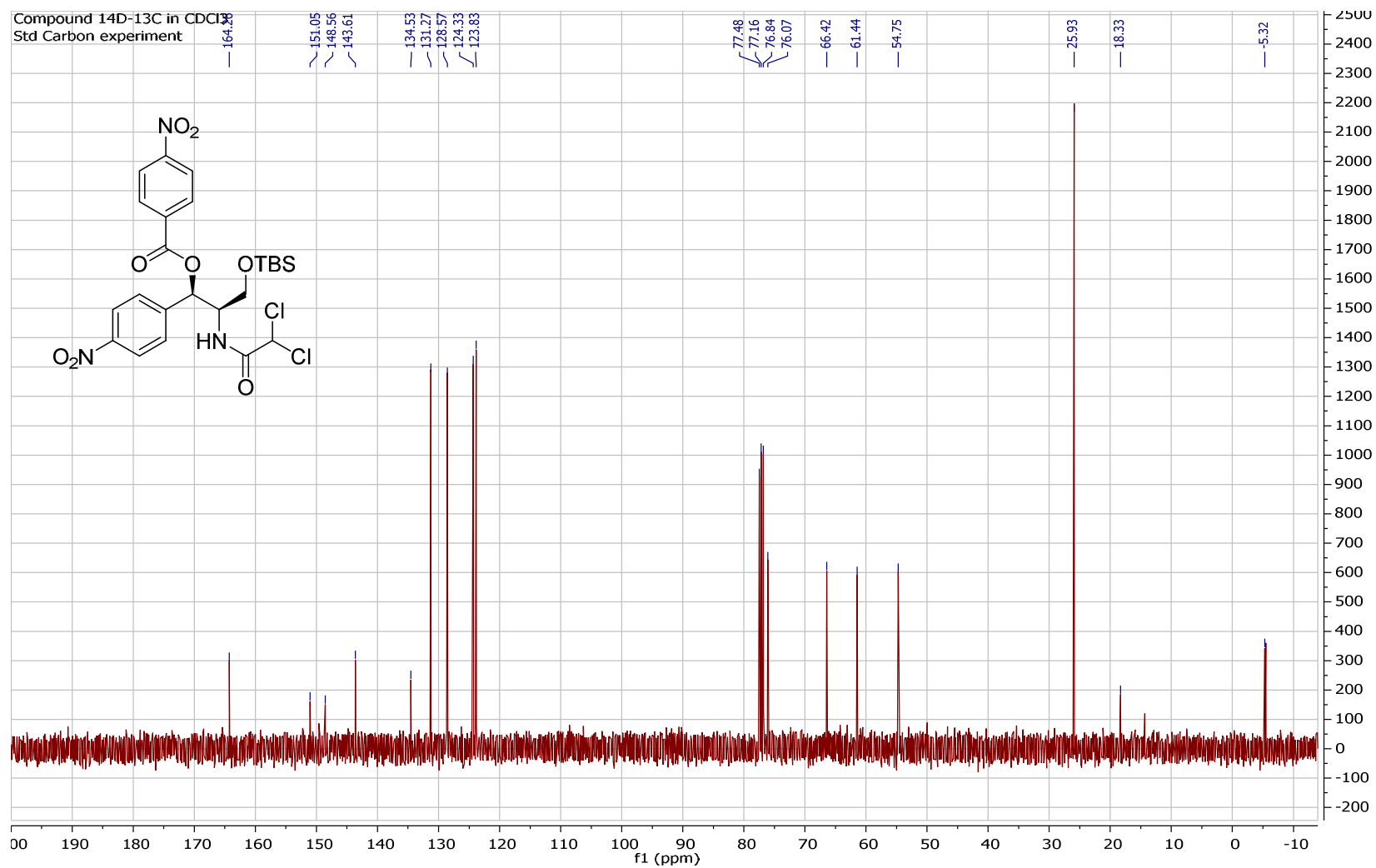


Figure S66. ¹³C NMR Spectrum of Compound 14d in CDCl₃

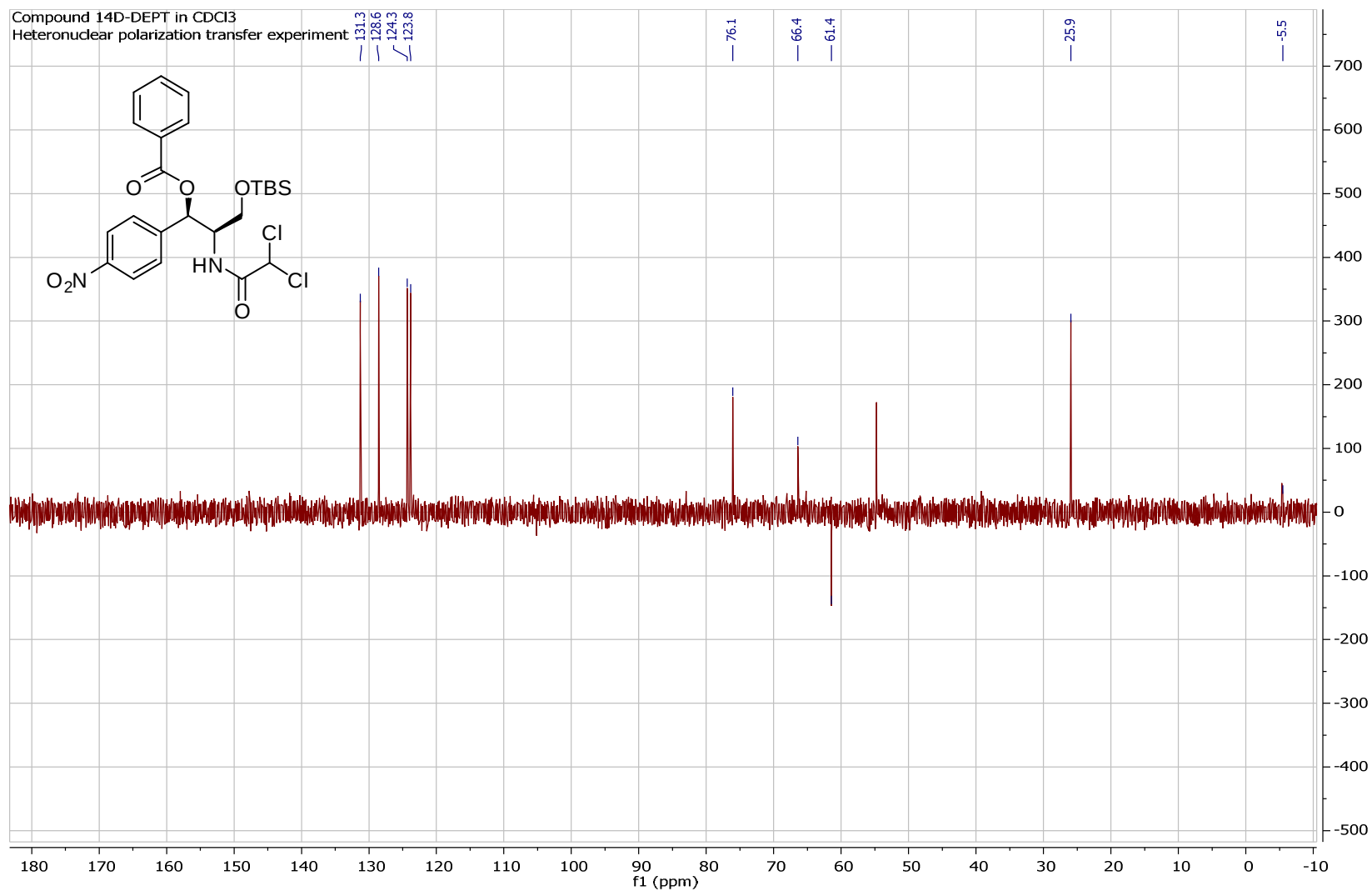


Figure S67. DEPT 135° Spectrum of Compound 14d

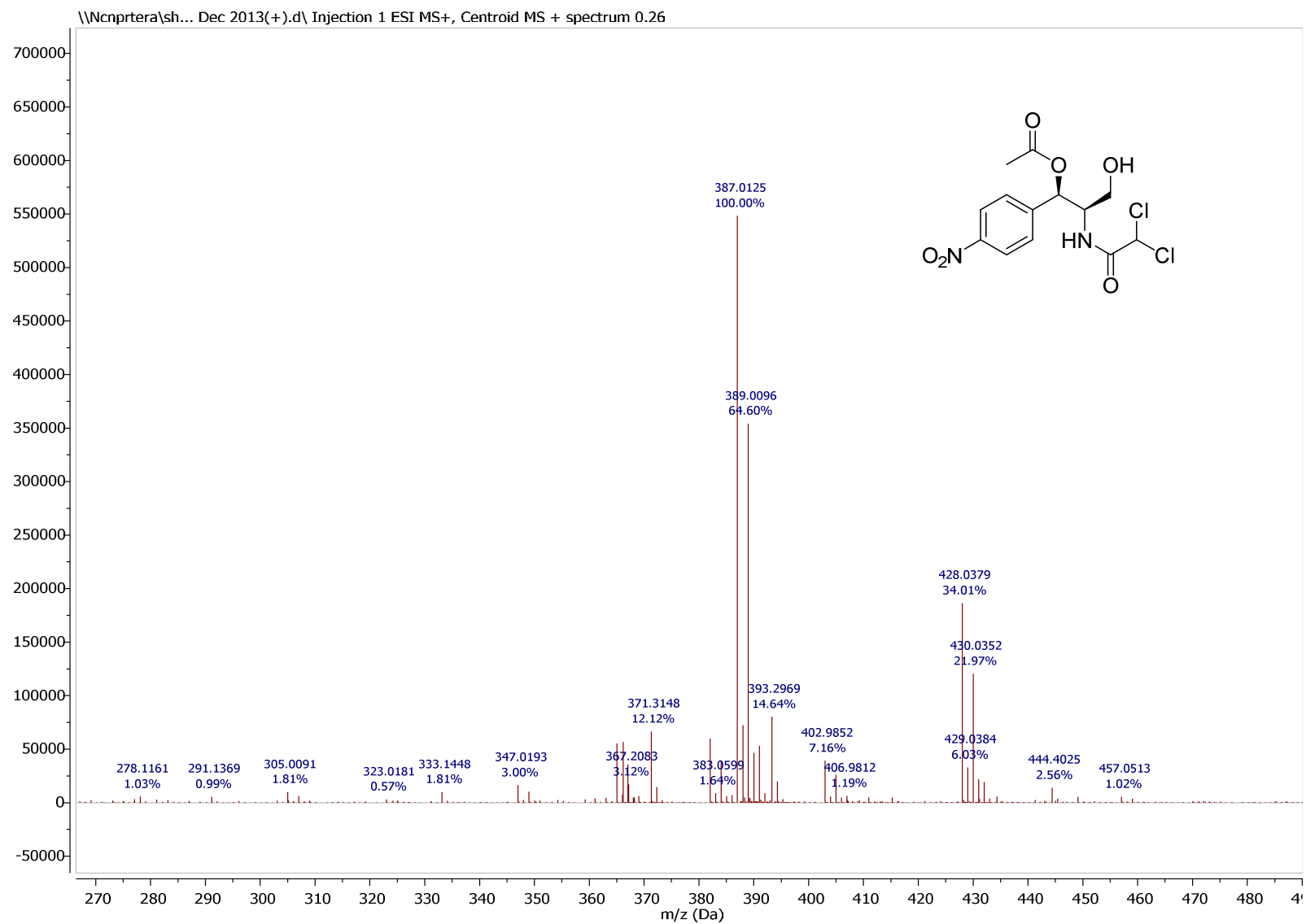


Figure S68. HRESI-MS Spectrum of Compound 15

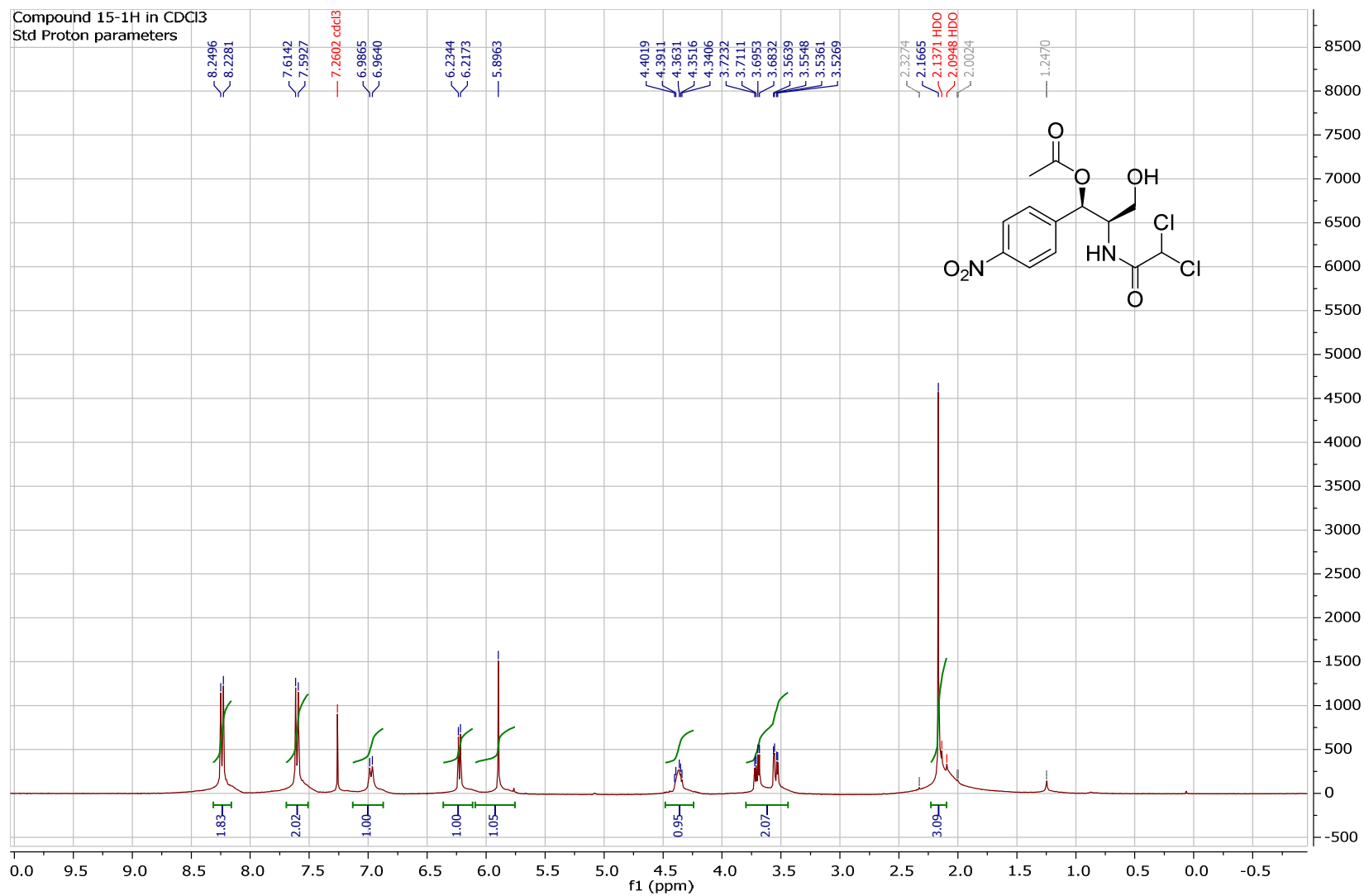


Figure S69. ¹H NMR Spectrum of Compound 15 in CDCl₃

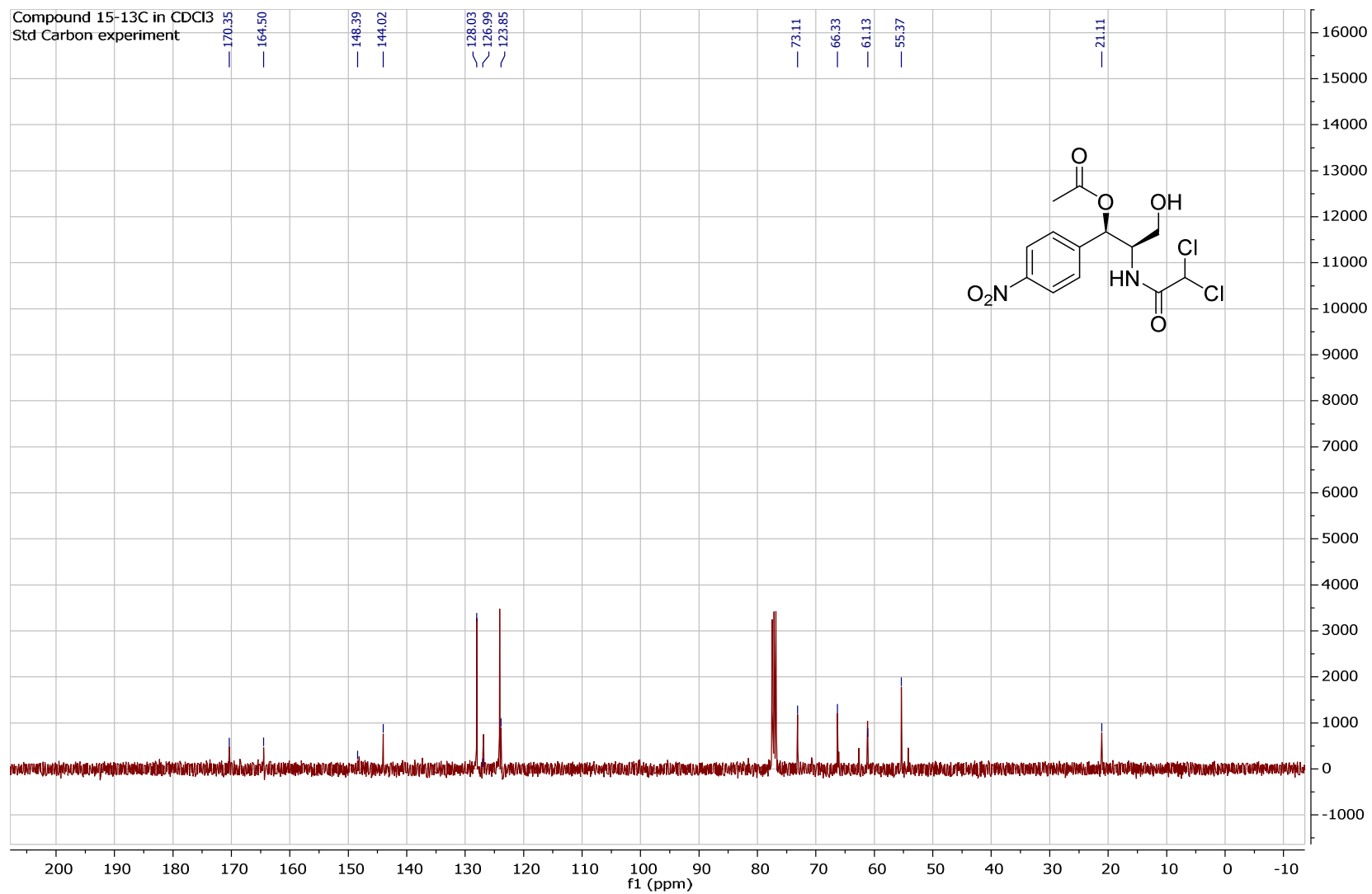


Figure S70. ¹³C NMR Spectrum of Compound 15 in CDCl₃

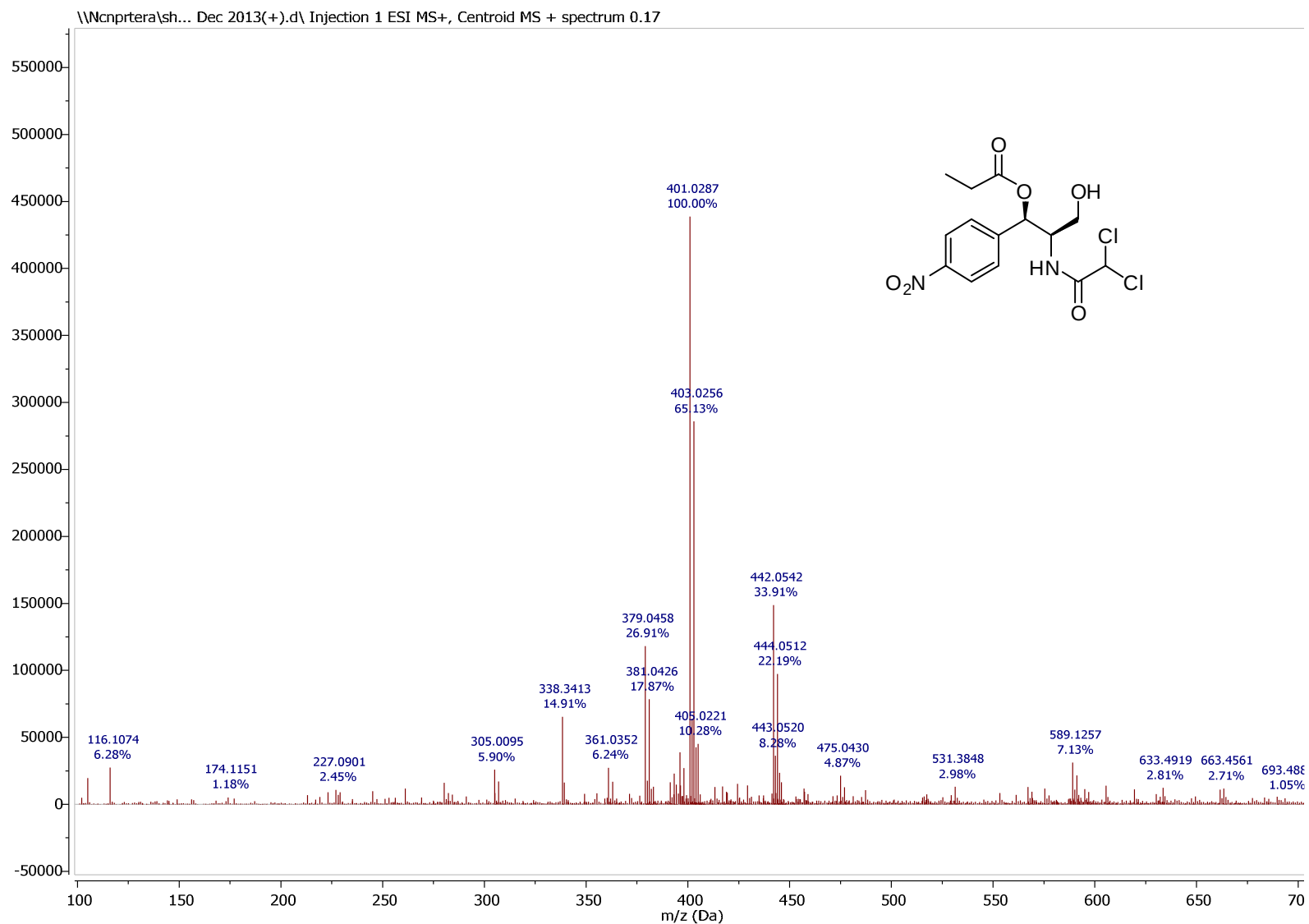


Figure S71. HRESI-MS spectrum of Compound 16

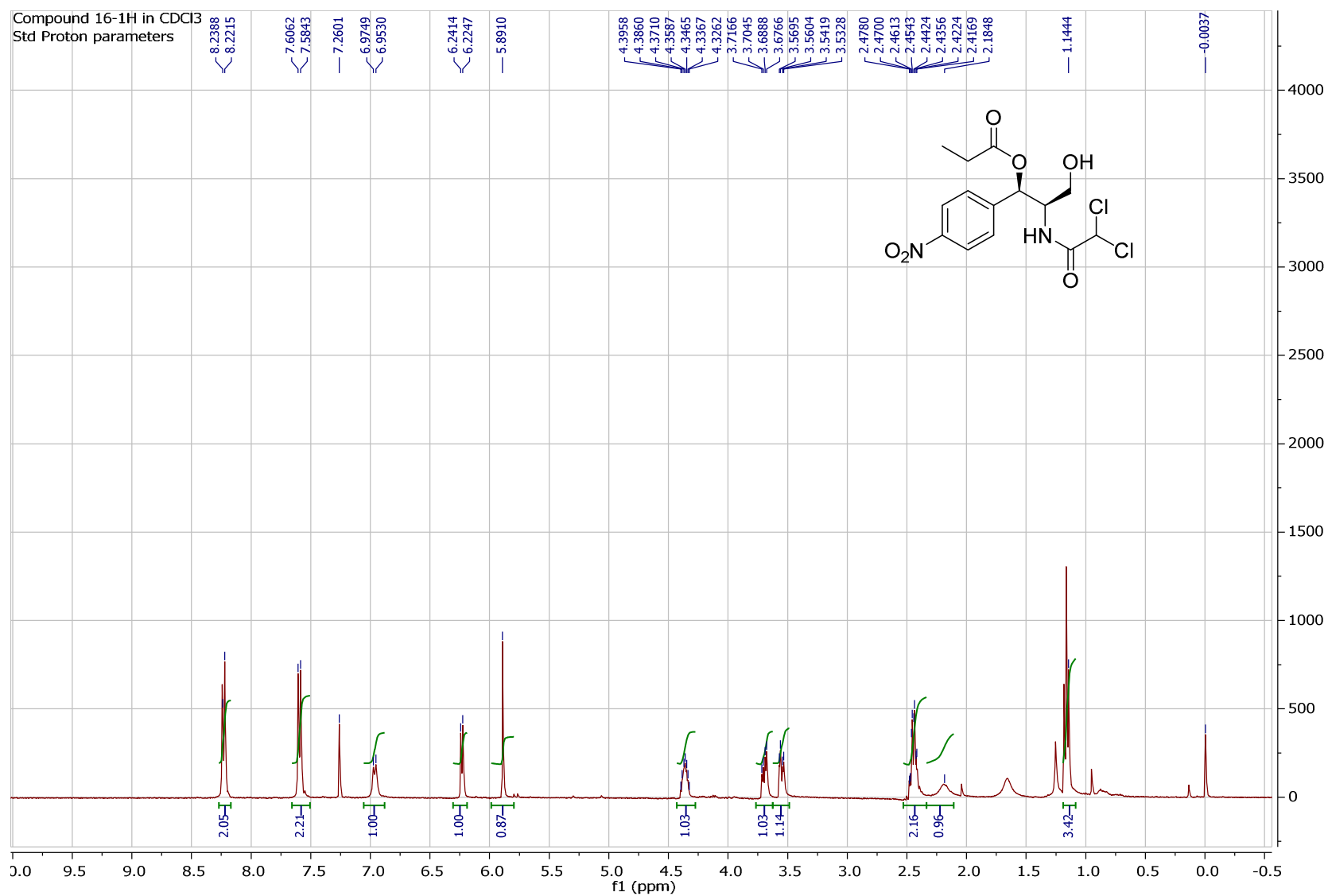


Figure S72. ¹H NMR Spectrum of Compound 16 in CDCl₃

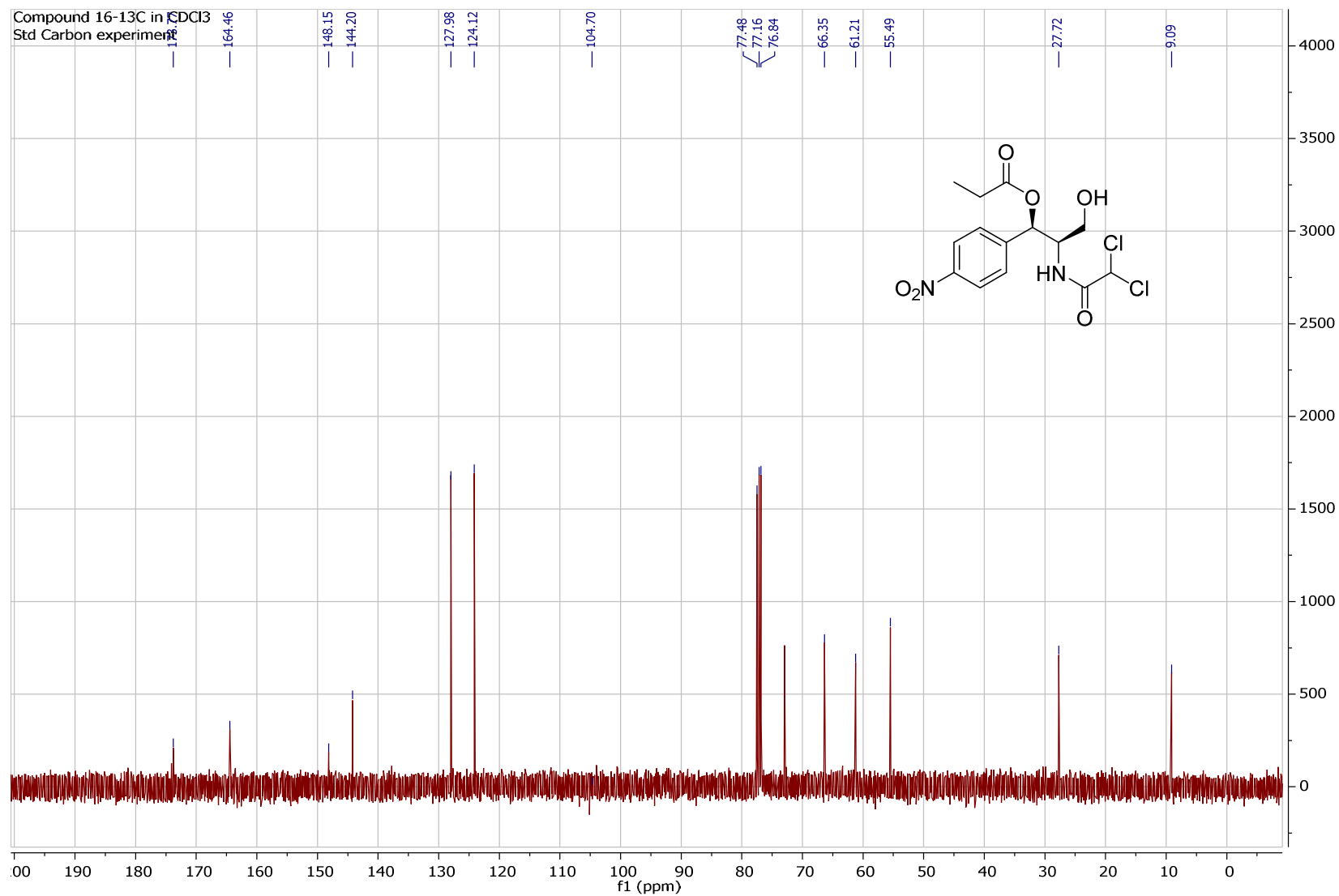


Figure S73. ¹³C NMR Spectrum of Compound 16 in CDCl₃

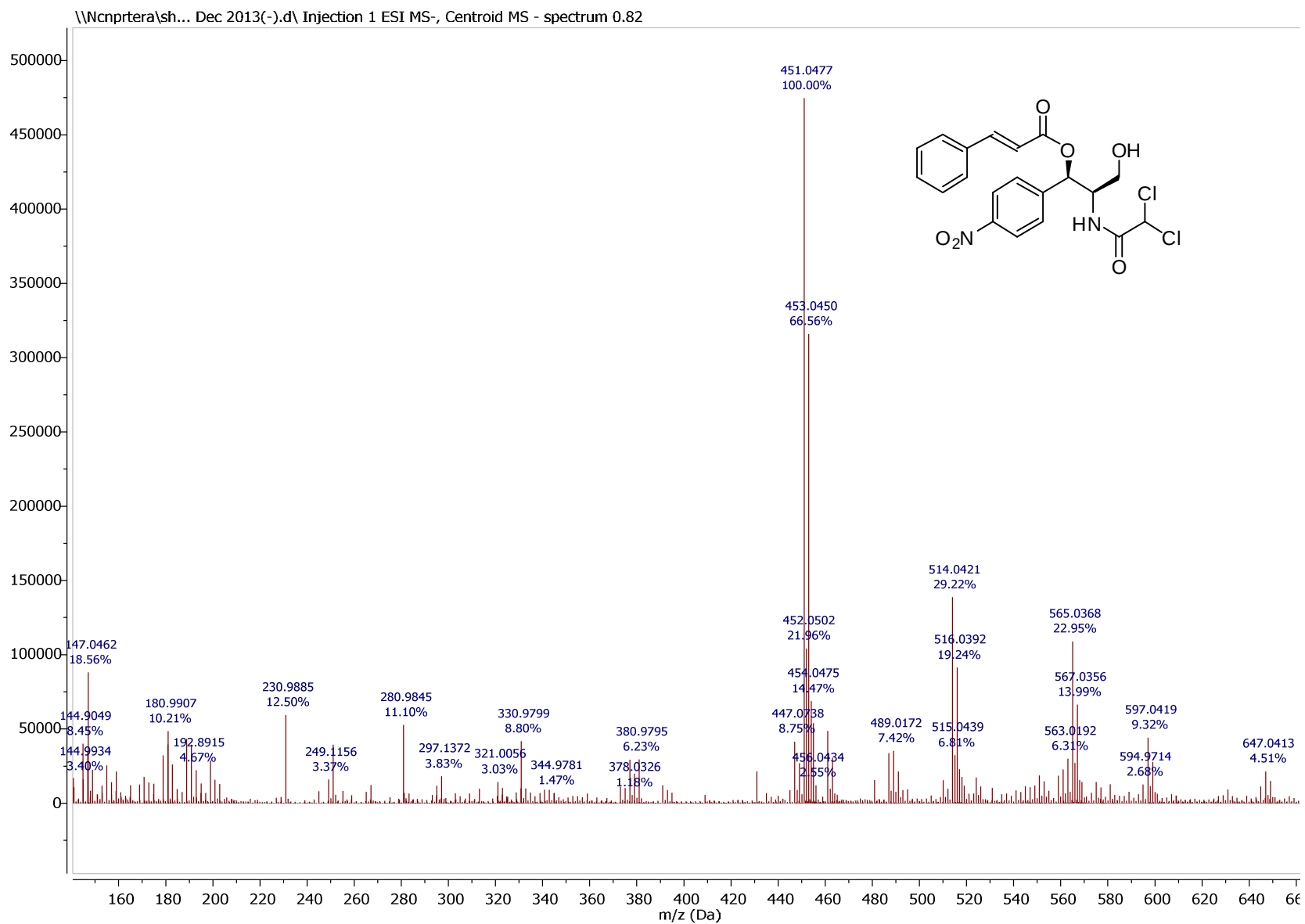


Figure S74. HRESI-MS Spectrum of Compound 17

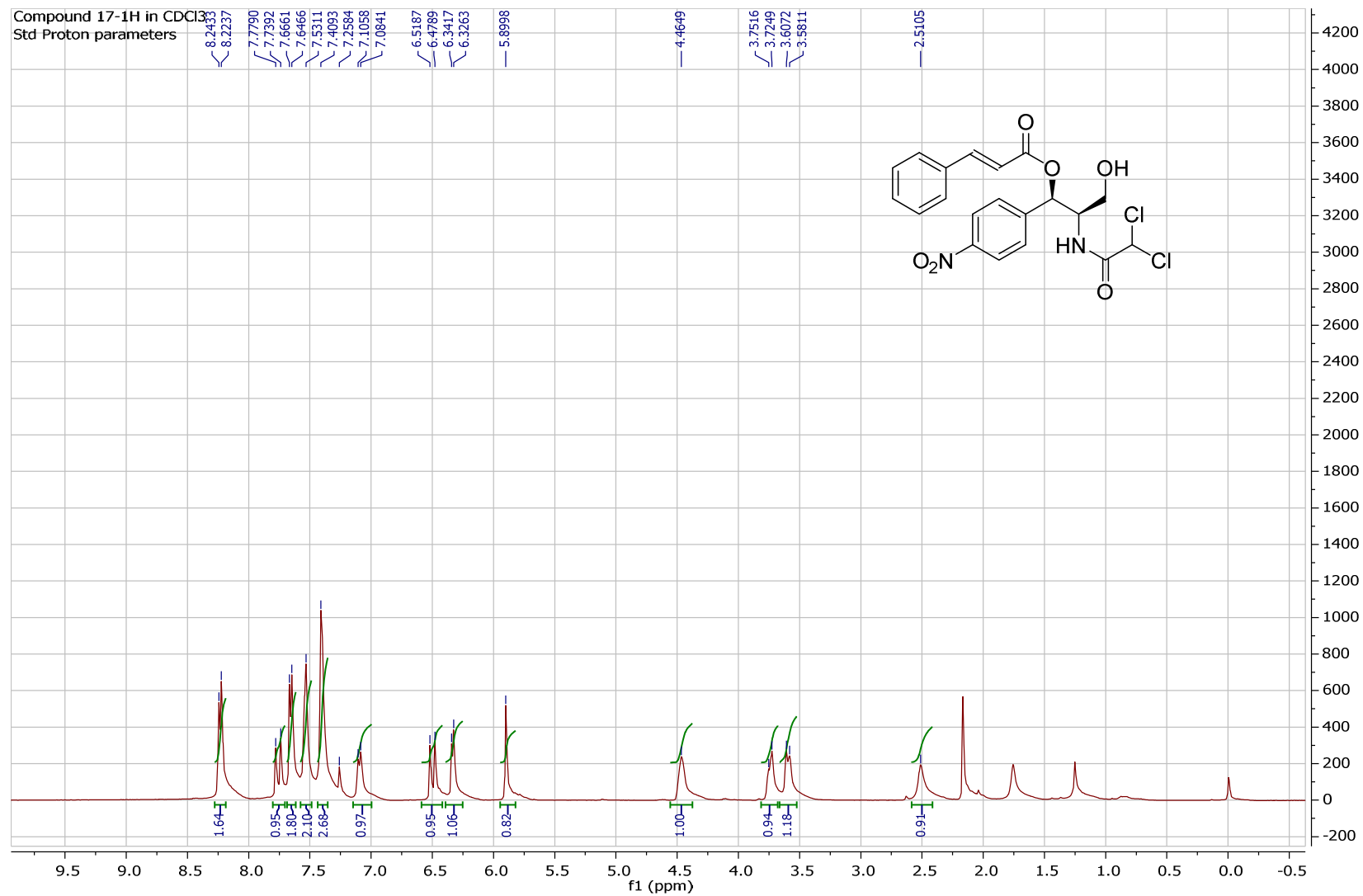


Figure S75. ¹H NMR Spectrum of Compound 17 in CDCl₃

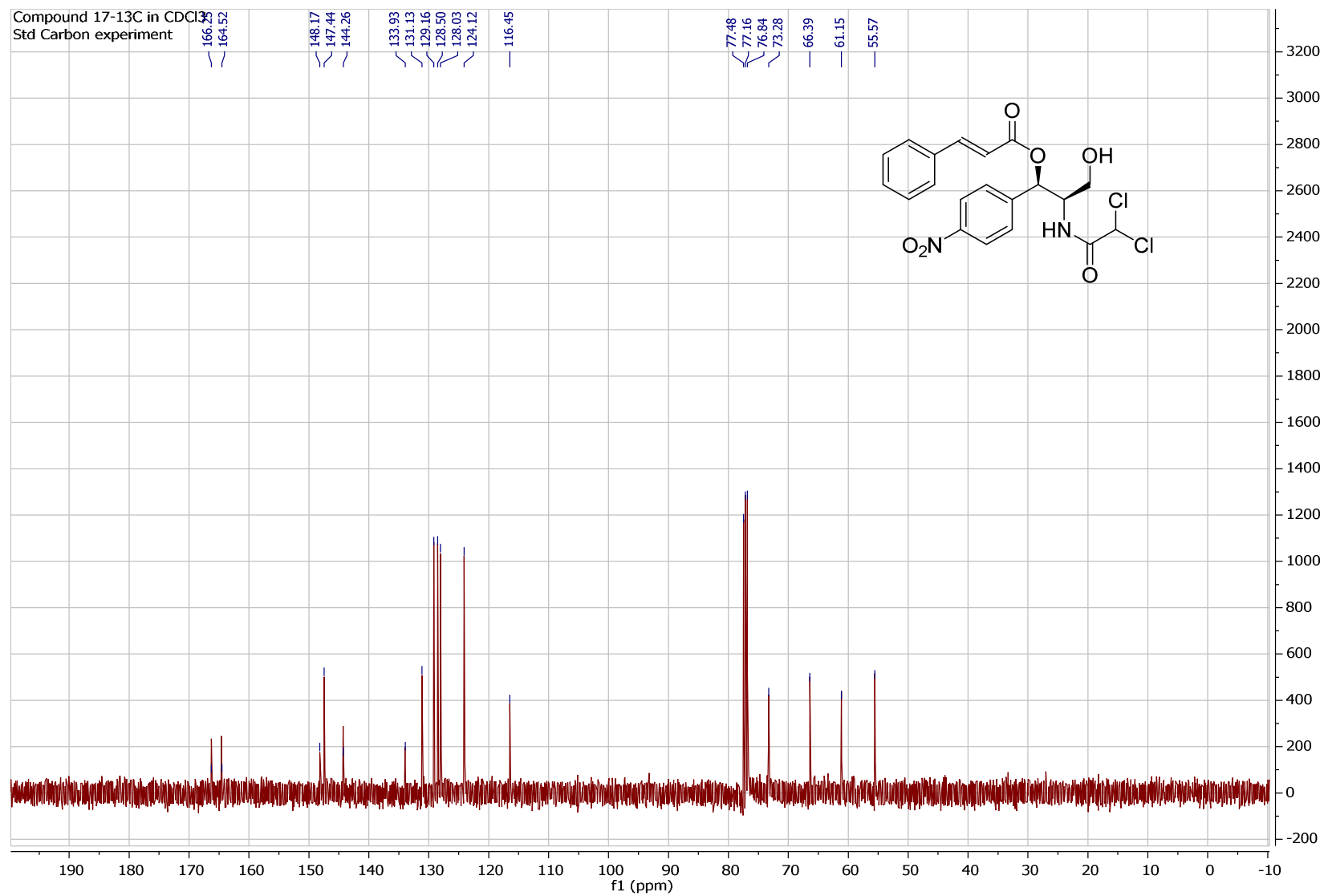


Figure S76. ¹³C NMR Spectrum of Compound 17 in CDCl₃

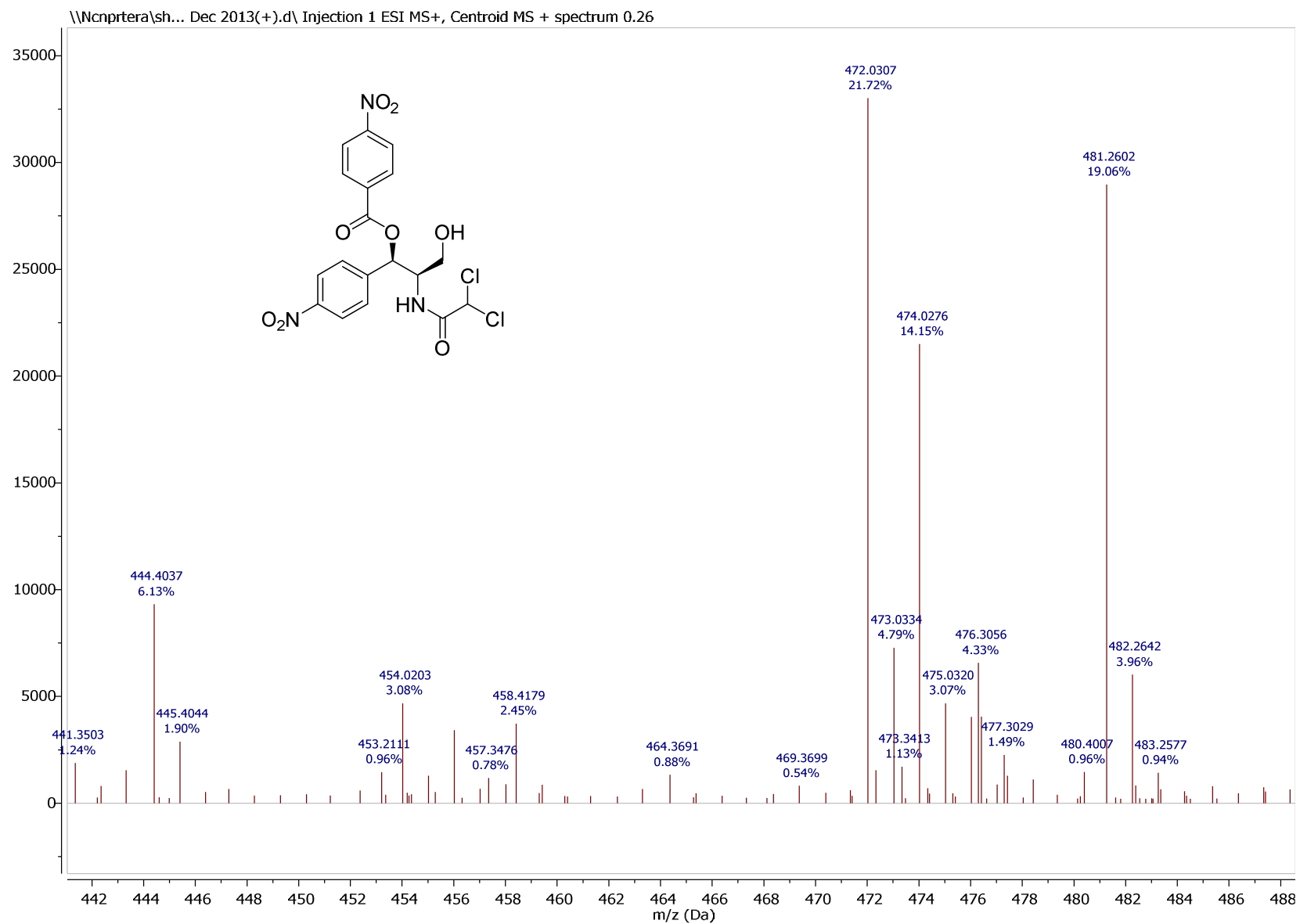
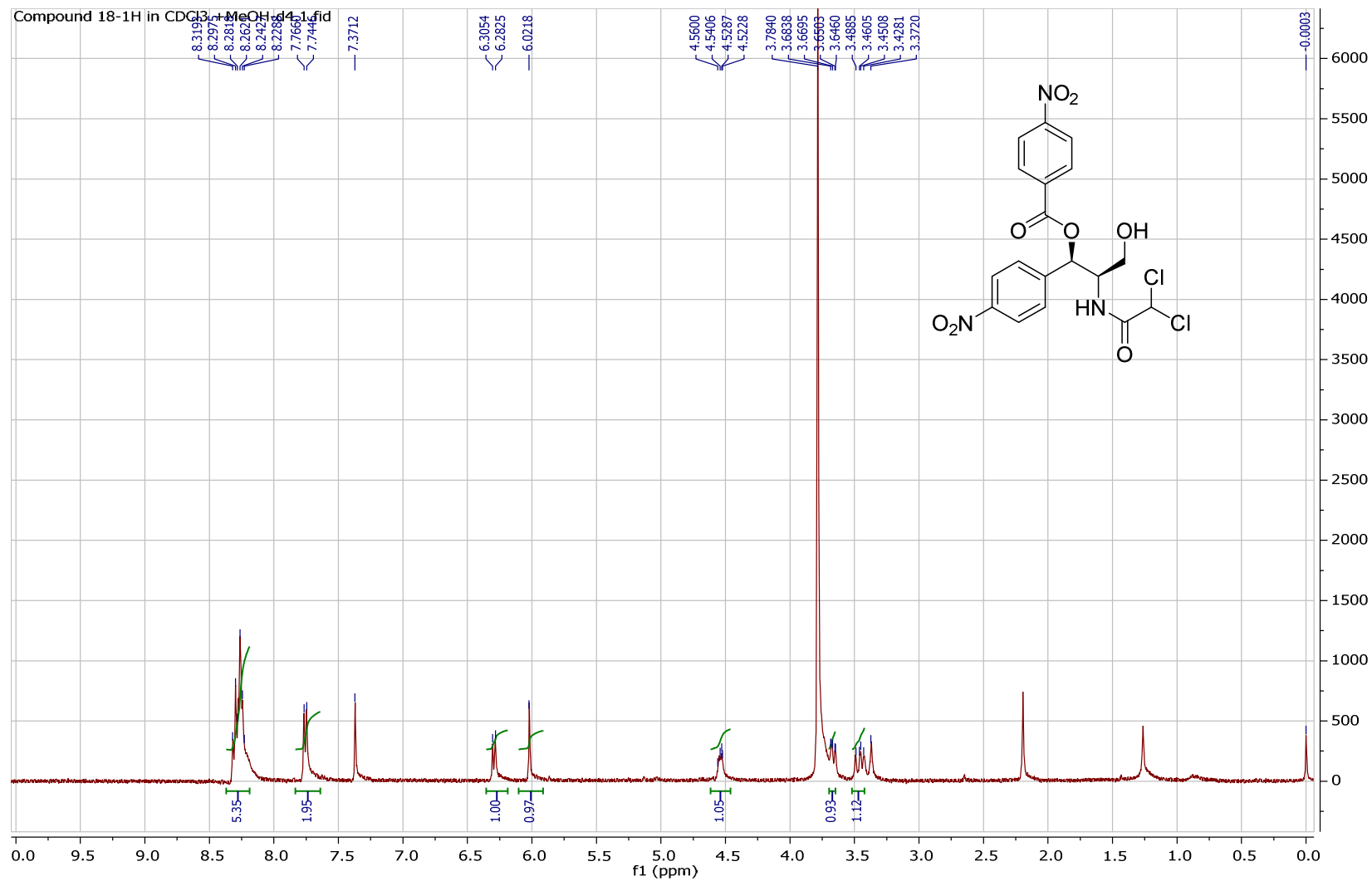


Figure S77. HRESI-MS Spectrum of Compound 18



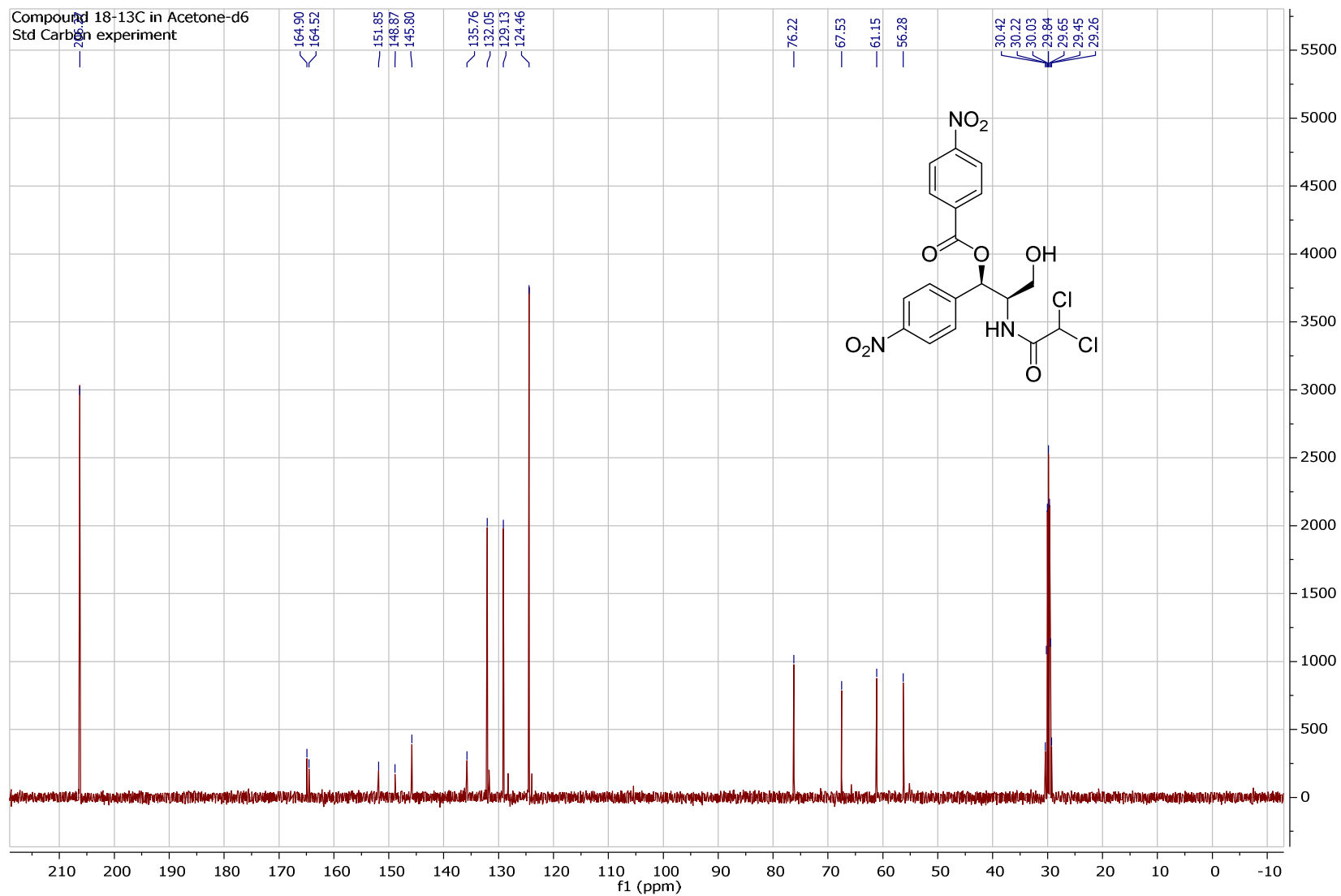


Figure S79. ¹³C NMR Spectrum of Compound 18 in Acetone-*d*₆