Supporting Information

3-Trifluoromethyl-3-Aryldiazirine Photolabels with Enhanced Ambient Light Stability

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Figure S1. Distribution of diazirine, linear diazo and carbene insertion product at different time intervals of UV activation. The distribution of the three species were determined by photoactivating a solution of the photolabel in d_4-methanol with UV light and measuring the compound distribution using ^{19}F NMR at different time intervals.

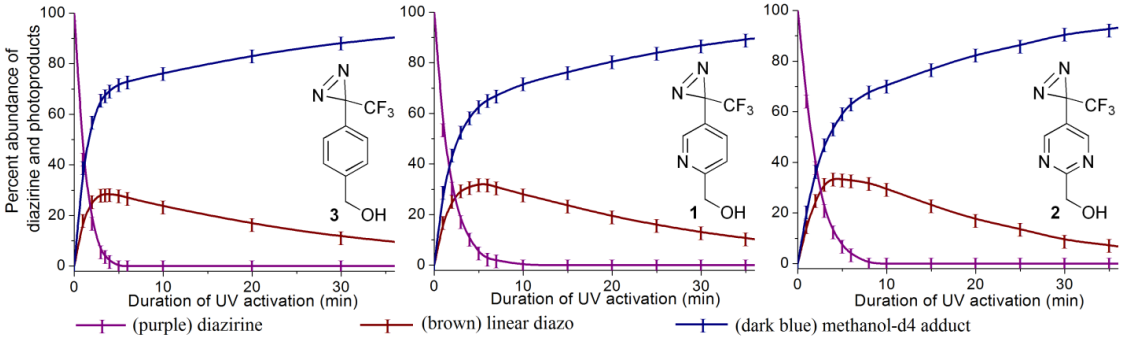


Table S1. Comparison of ambient light stability of modified trifluoromethylaryl diazirines **1** and **2** versus the conventional trifluoromethylphenyl diazirine. A solution of trifluoromethylaryl diazirines **1**, **2** or **3** in d4-methanol was exposed to light from two linear fluorescent lamps (28 W each) at room temperature and the photodecomposition of the diazirines were followed by ^{19}F NMR. Percentage of diazirine intact upon exposure to ambient light at various time intervals.

Duration of ambient light exposure (days)	Amounts of unreacted diazirine (%)		
	3	**1**	**2**
0	100	100	100
4	87.7	97.1	99
7	78.1	95.2	98
14	58.1	90.1	95.2
18	49	87.7	94.3
26	35	82.6	92.6
31	26.8	79.4	90.1

Table S2 Comparison of stability of modified trifluoromethylaryl diazirines **1** and **2** versus the conventional trifluoromethylphenyl diazirine **3** under exposure to incandescent light. Percentage of diazirine intact upon exposure to incandescent light at various time intervals.

Duration of incandescent lamp exposure (days)	Amounts of unreacted diazirine (%)		
	phenyl	pyridinyl	pyrimidinyl
0	100.0	100.0	100.0
1	94.3	97.1	98.0
3	85.5	93.5	96.2
5	74.1	87.7	91.7
15	22.5	54.9	67.1

The Table S3 shows that the conventional photolabel **3** and modified trifluoromethylaryl diazirine photolabels **1** and **2** are all stable as methanolic solutions at 21°C in the dark.

Table S3 Comparison of thermal stability of *d*4-methanolic solution of **1**, **2** and **3** in the dark at room temperature using ^{19}F NMR

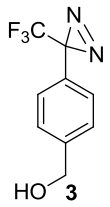
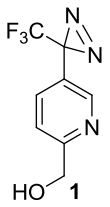
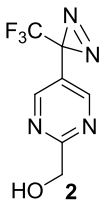
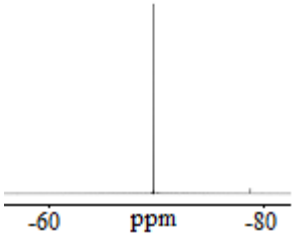
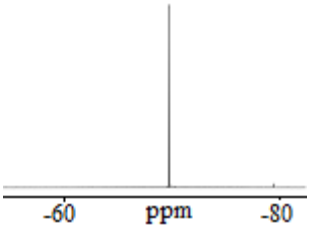
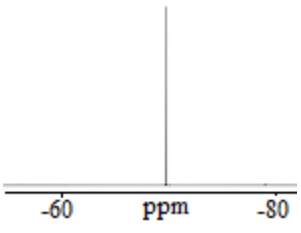
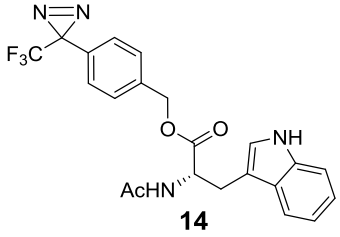
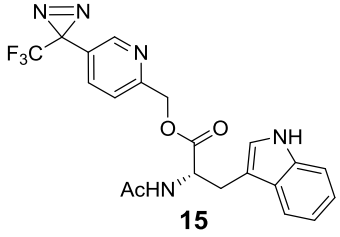
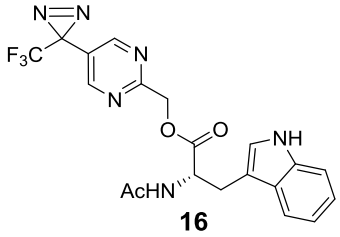
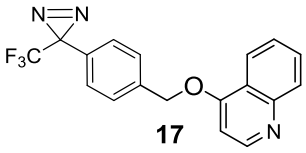
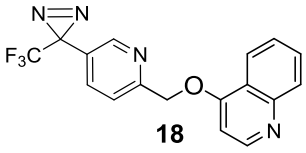
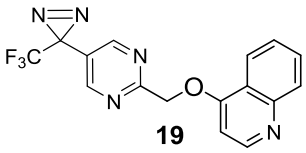
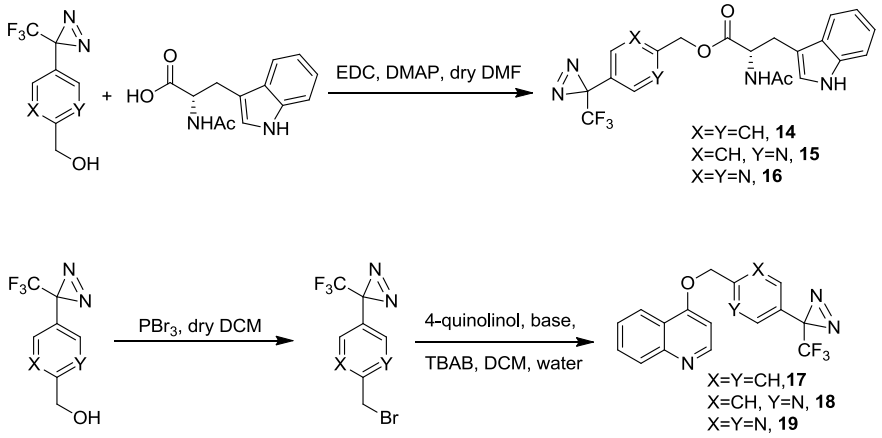
Duration of *d*4-methanolic solution in the dark	 **3**	 **1**	 **2**
0 days			
14 days			
31 days			

Table S4 Comparison of aqueous solubility of photoaffinity probes derivatized with modified photolabels **1** and **2** with conventional photolabel **3**. The aqueous solubility of the photolabels increased by several orders of magnitude when the phenyl derived diazirine was replaced with the pyridinyl or pyrimidinyl derived diazirine.

Compound	Aqueous Solubility (μM)	
	pH 7.4	pH 5.0
 **14**	< 0.02	< 0.02
 **15**	4.09 ± 0.19	4.29 ± 0.14
 **16**	133 ± 0.5	131 ± 1.7
 **17**	11.3 ± 0.55	14.1 ± 0.43
 **18**	374 ± 2.7	422 ± 4.9
 **19**	≥ 1,000	≥ 1,000

Synthesis of Photoaffinity Probes 14-19 and 20-22

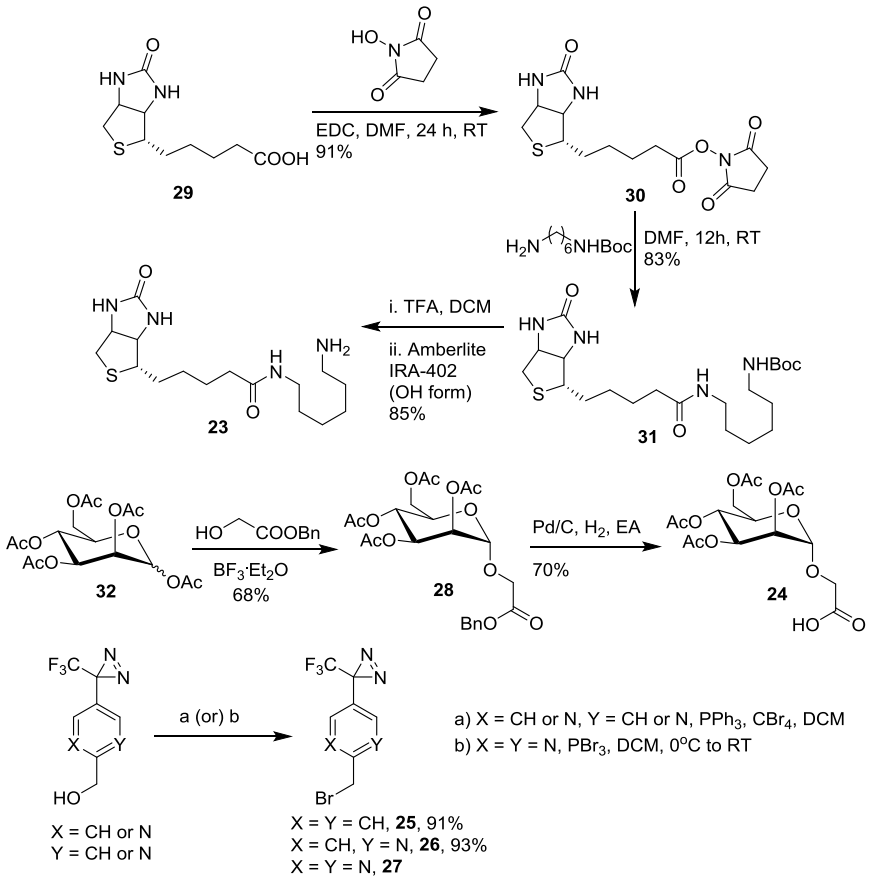
Synthesis of photoaffinity probes 14-19 for solubility studies. Photoaffinity probes **14-19** were synthesized to evaluate the aqueous solubility (scheme S1). The *N*-acetyl tryptophan derived photoaffinity probes **14**, **15**, and **16** were synthesized by a conventional esterification reaction using 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide (EDC) coupling (Scheme S1). The quinolone derivatives **17**, **18**, and **19** were prepared by a two-step process in which the alcohol function of the photolabel bearing moiety was converted to the bromide using phosphorus tribromide (Scheme S1). In the second step the resultant bromide was subjected to a nucleophilic substitution reaction with quinolinol under basic conditions. The use of sodium hydroxide as base was successful in the preparation of **17** and **18**, but resulted in undesired side products with pyrimidinyl derivative. Thus potassium carbonate was used to deprotonate the quinolinol for the substitution reaction with pyrimidinyl derived photo probe to obtain **19**.



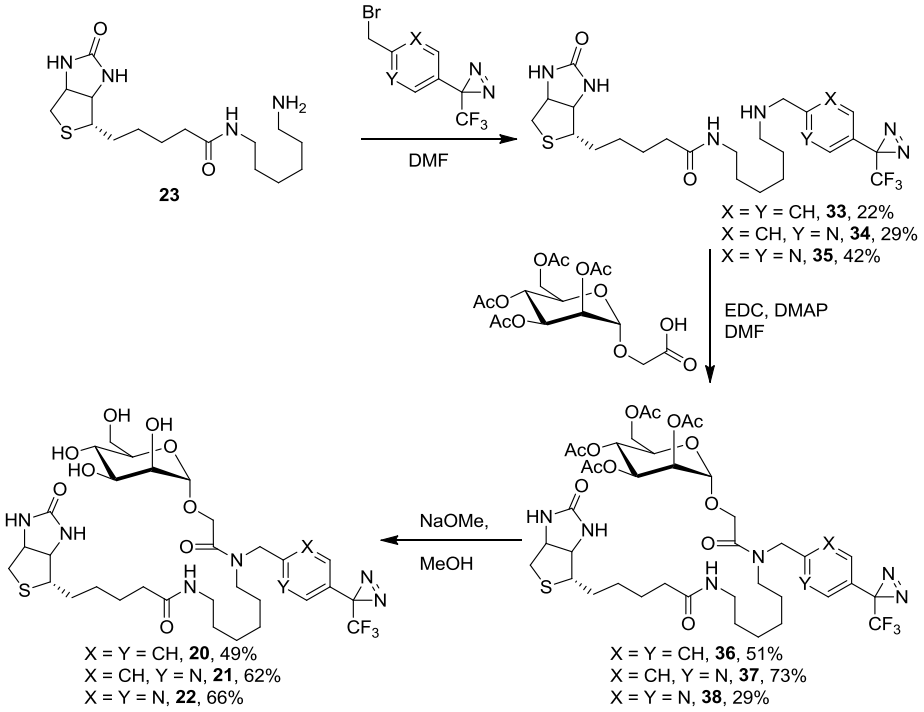
Scheme S1. Synthesis of photoaffinity probes **14** to **19** for solubility studies

Synthesis of photoaffinity labels based on conventional photolabel 3 and modified trifluoromethylaryl diazirine photolabels 1 and 2. The synthesis of photoaffinity labels **20**, **21**, and **22** was achieved by a convergent route, in which biotin amine **23**, carboxylic acid derived mannose **24**, and diazirines **25**, **26** and **27** were first synthesized separately as key intermediates (Scheme S2) and coupled together in the last steps of the synthesis. Biotin amine **23** was synthesized as reported earlier[1] and the free base was generated using the basic resin Amberlite IRA-402 (OH form). Benzyl ester **28** was synthesized by a boron trifluoride coupling of D-mannose pentaacetate with the corresponding alcohol in moderate yield. Benzyl ester **28** was exclusively α-mannoside as confirmed by the ^{1}H NMR coupling constants. The free carboxylic acid **24** was synthesized by benzyl deprotection of **28** using Pd/C and hydrogen in good yield. The bromides **25** and **26** were synthesized from the corresponding phenyl diazirine **3** and pyridinyl diazirine **1** *via* the Apple reaction. In contrast, the Apple reaction did not yield bromide **27** with pyrimidinyl diazirine **2**. However, the reaction of pyrimidinyl diazirine **2** with phosphorus tribromide yielded the desired bromide **27**.

In the convergent step of the synthesis, bromide **25**, **26**, or **27** was treated with an excess of primary amine **23** to obtain the desired secondary amine **33**, **34**, or **35** (Scheme S3). The corresponding secondary amine was subsequently subjected to an EDC coupling with carboxylic acid **24** to get the amide **36**, **37**, or **38**, which was deacetylated using a catalytic amount of sodium methoxide in methanol to furnish the photoaffinity label **20**, **21**, or **22**.



Scheme S2. Synthesis of intermediates **23**, **24**, **25**, **26** and **27**



Scheme S3. Synthesis of photoaffinity labels **20**, **21**, and **22**

Table S5 Identification of peptide sequence photolabeled with photoaffinity labels **20**, **21** and **22** using high resolution mass spectrometry.

Photo probe	Mass calculated			Mass found		Error in mass accuracy (ppm)
	Mass of peptide Val91 - Lys101 (VGLSASTGLYK)	Mass of (probe -N_2)	Mass of (peptide + probe - N_2)	Mass of doubly protonated peptide	Mass of neutral peptide	
20	1094.5972	732.3016	1826.8988	914.4585	1826.9024	1.97
21	1094.5972	733.2968	1827.8940	914.9558	1827.8970	1.64
22	1094.5972	734.2921	1828.8893	915.4543	1828.8940	2.57

When the photoaffinity probes are subjected to mass spectroscopic analysis under the conditions the peptide samples are analyzed, the major fragmentation was the loss of mannose residue in the mass spectrometer. In the same retention time that the above labeled peptide was found another peptide fragment was found (Figure S2) with the mass corresponding to same peptide sequence (Val91 - Lys101) plus the photoaffinity probe with the loss of mannose residue (Table S4).

Figure S2 Loss of mannose residue in the mass spectrometer by the peptides labeled with photolabels **20**, **21** and **22**

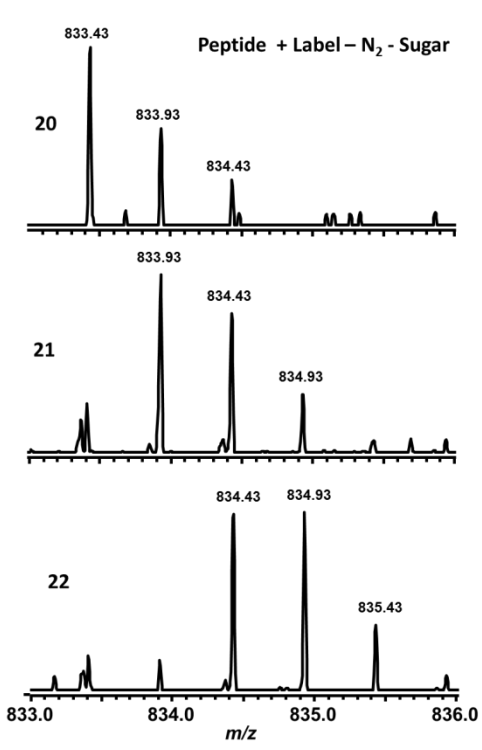


Table S6 Peptide fragments photolabeled with photoaffinity labels **20**, **21**, and **22** upon the loss of mannose residue in the mass spectrometer

Photo probe	Mass Calculated			Mass Found		Error in mass accuracy (ppm)
	Mass of peptide Val91 - Lys101 (VGLSASTGLYK)	Mass of (probe - N_2 - mannose)	Mass of (peptide + probe - N_2 - mannose)	Mass of doubly protonated peptide	Mass of neutral peptide	
20	1094.5972	570.2488	1664.8460	833.4328	1664.8510	3.00
21	1094.5972	571.2440	1665.8412	833.9284	1665.8422	0.60
22	1094.5972	572.2393	1666.8365	834.4272	1666.8398	1.98

Figure S3 MS/MS fragmentation pattern of peptide labeled with photoprobe **22**

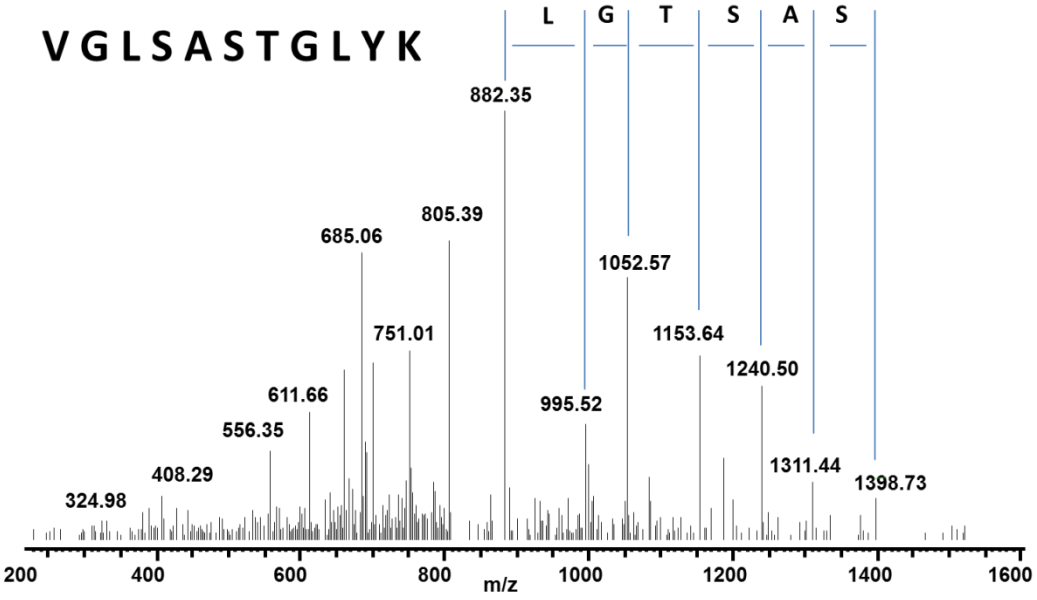
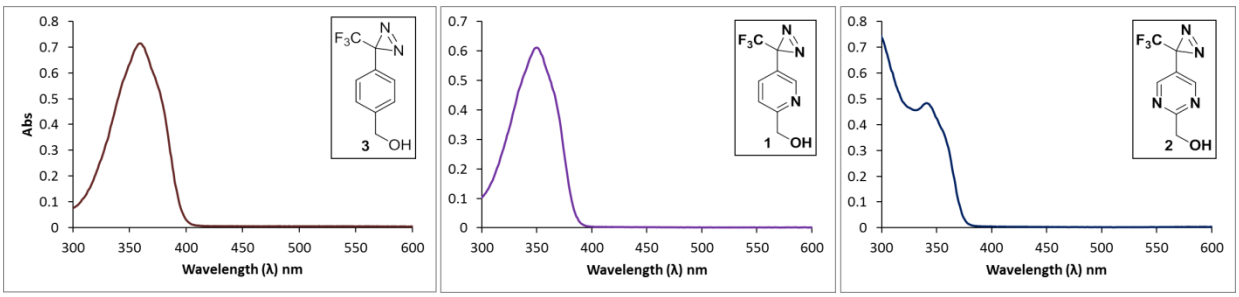


Figure S4. Near UV-visible spectra of photolabels **1, 2** and **3**, recorded using 2.5 mM solution in methanol



Experimental Procedures for Synthesis and Biochemical Experiments

General information. Commercially available reagents and solvents were used without further purification. All the reactions were done under anhydrous condition and argon atmosphere, unless specified otherwise. Room temperature (RT) experiments were done at 21°C and overnight experiments were done for 16 hours. Thin layer chromatography (TLC) was performed using EMD silica gel 60-F plates (it is individually specified in instances where neutral alumina TLC plate was used instead) and spots were visualized using UV light or phosphomolybdic acid (PMA) staining solution. Purification by flash chromatography was done using EMD silica gel (230 – 400 mesh) (it is individually specified in instances where neutral alumina was used instead). NMR experiments were done on a Bruker DPX-250 (^{1}H at 250 MHz and ^{13}C at 63 MHz), a Varian Inova 400 MHz spectrometer (^{1}H at 400 MHz, ^{13}C at 100 MHz, and ^{19}F at 376 MHz), an Inova 500 MHz spectrometer (^{1}H at 500 MHz and ^{13}C at 125 MHz) or an Inova 600 MHz spectrometer (^{1}H at 600 MHz and ^{13}C at 150 MHz) and the data was processed using MestReNova. Chemical shifts (δ) are reported in parts per million (ppm) with the solvent peak as the internal standard for ^{1}H and ^{13}C, while trifluorotoluene was used as external standard for ^{19}F NMR spectroscopy. Coupling constants are reported in Hz. Signals are quoted as s (singlet), d (doublet), t (triplet), q (quartet), m (multiplet), br (broad), dd (doublet of doublets), ddd (doublet of doublets of doublets) or dt (doublet of triplets). High resolution mass spectrometry was performed on Agilent 6540 Ultra-High-Definition (UHD) Q-TOF LC-MS with electrospray ionization. Preparative HPLC was conducted using Agilent Eclipse XDB-C18 PN 990967-202 column with gradient 10% to 50% of acetonitrile in water with 0.05% TFA over 15 min followed by 100% acetonitrile for 5 min (flow rate: 5 ml min^{-1}). Agilent 1100 series LC/MSD was used for the solubility measurement experiments. Orel Instruments housing with an Osram 150 W XBO xenon short-arc lamp, fitted with a Schott WG-320 filter to eliminate UV lights below 320 nm, was used for the photoactivation studies. For Western blot analysis anti-biotin−peroxidase antibody produced in goat purchased from Sigma (A4541) was used to detect the biotin labeled proteins. The photographic films (Thermo scientific # 34091) used for recording the chemiluminescence in Western blot analysis were developed using Konica Minolta medical film processor (Model SRX-101A). Photolabeled samples were analyzed using a linear ion trap Orbitrap XL (LTQ OrbiTrap XL, Thermo Fisher Corp., Bremen, Germany).

5-bromo-2-(((tert-butyldimethylsilyl)oxy)methyl)pyridine (5). To a solution of (5-bromopyridin-2-yl)methanol[2] (3.65 g, 19.4 mmol) in anhydrous dichloromethane (DCM) (50 mL), *tert*-butyldimethylsilyl chloride (3.22 g, 21.3 mmol) and imidazole (2.90 g, 42.6 mmol) were added and stirred overnight at room temperature (RT). The reaction mixture was quenched with saturated ammonium chloride and extracted with dichloromethane (3 times). The combined organic layer was dried with anhydrous sodium sulfate and concentrated. The resultant crude was subjected to flash chromatography with silica column and 10% ethyl acetate in hexanes as eluent to give **5** (5.29 g, 90%) as colorless oil. R_f 0.51 (10% ethyl acetate in hexanes). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.54 (d, J = 2.4 Hz, 1H), 7.80 (dd, J = 8.4 Hz, J = 2.3 Hz, 1H), 7.40 (d, J = 8.4 Hz, 1H), 4.76 (s, 2H), 0.94 (s, 9H), 0.10 (s, 6H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 160.1, 149.7, 139.3, 121.6, 118.6, 65.7, 26.0, 18.4, -5.3. HRMS (ESI^+) calcd for $C_{12}H_{21}BrNOSi^+$ $[M + H]^+$ 302.0570, found: 302.0577.

1-(6-(((tert-butyldimethylsilyl)oxy)methyl)pyridin-3-yl)-2,2,2-trifluoroethanone (6). To a solution of **5** (4.10 g, 13.6 mmol) in diethyl ether (60 mL) in an argon back flushed flask, *n*-butyllithium (6.5 mL of 2.5 M solution in hexane) was slowly added at -78°C and left to stir. After 30 minutes methyl trifluoroacetate (2.09 g, 16.3 mmol) was added and stirred at -78°C for 2 hours and warmed to room temperature. The reaction was quenched with saturated ammonium chloride and extracted with ethyl acetate (3 times). The combined organic layer was dried with anhydrous sodium sulfate and concentrated. The resultant crude was subjected to flash chromatography with neutral alumina column and 4% methanol in dichloromethane as eluent to give **6** (3.3 g, 76%) as pale yellow oil. R_f 0.45 (6% methanol in dichloromethane, neutral alumina TLC plate). ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.14 (s, 1H), 8.35 (d, J = 7.8

Hz, 1H), 7.74 (d, J = 7.9 Hz, 1H), 4.90 (s, 2H), 0.96 (s, 9H), 0.14 (s, 6H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 179.8 (q, J = 36.6 Hz), 169.0 (s), 150.5 (q, J = 2.7 Hz), 138.1 (q, J = 1.9 Hz), 124.4 (s), 120.2 (s), 116.5 (q, J = 291.0 Hz), 66.07 (s), 26.0 (s), 18.5 (s), -5.3 (s). ^{19}F NMR (376 MHz, $CDCl_3$) δ -72.30 (s). HRMS (ESI^+) calcd for $C_{14}H_{21}F_3NO_2Si^+$ $[M + H]^+$ 320.1288, found: 320.1297.

1-(6-(((tert-butyldimethylsilyl)oxy)methyl)pyridin-3-yl)-2,2,2-trifluoroethanone O-tosyl oxime (7). A suspension of hydroxylamine hydrochloride (0.36 g, 5.18 mmol) and sodium acetate trihydrate (0.93 g, 6.83 mmol) in ethanol (5 mL) was stirred for 10 minutes and allowed to settle. The supernatant of the above mixture was transferred to a solution of **6** (0.55 g, 1.72 mmol) in ethanol (2 mL) and refluxed for 16 hours. Upon cooling to room temperature ethanol was removed under reduced pressure and the resultant concentrate was extracted between water and dichloromethane (water layer was extracted with dichloromethane 3 times). The combined organic layer was dried with anhydrous sodium sulfate and concentrated. The concentrated crude was back flushed with argon and anhydrous dichloromethane (5 mL) was added followed by pyridine (273 mg, 3.45 mmol) and 4-dimethylaminopyridine (DMAP) (21.0 mg, 0.17 mmol). The solution was cooled to 0°C and *p*-toluenesulfonic anhydride (619 mg, 1.90 mmol) was added slowly and stirred at the same temperature for 30 minutes followed by 2 hours at room temperature. The reaction was quenched with water and extracted with dichloromethane (3 times). The combined organic layer was dried with anhydrous sodium sulfate and concentrated. The resultant crude was subjected to flash chromatography with silica column and 10% ethyl acetate in hexanes as eluent to give **7** (0.51 g, 61%) as colorless oil. R_f 0.37 (10% ethyl aceate in hexanes). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.51 (s, 1H), 7.86 (d, J = 8.2 Hz, 2H), 7.76 (m, 1H), 7.65 (m, 1H), 7.36 (d, J = 8.0 Hz, 2H), 4.84 (s, 2H), 2.44 (s, 3H), 0.94 (s, 9H), 0.12 (s, 6H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 165.3, 151.8 (q, J = 34.3 Hz), 147.9, 146.5, 136.9, 131.1, 130.1, 129.3, 119.8, 119.6 (q, J = 277 Hz), 119.4, 65.9, 25.9, 21.8, 18.4, -5.3. ^{19}F NMR (376 MHz, $CDCl_3$) δ -66.81 (s). HRMS (ESI^+) calcd for $C_{21}H_{28}F_3N_2O_4SSi^+$ $[M + H]^+$ 489.1486, found: 489.1497.

2-(((tert-butyldimethylsilyl)oxy)methyl)-5-(3-(trifluoromethyl)diaziridin-3-yl)pyridine (8). To a solution of **7** (1.28 g, 2.62 mmol) in anhydrous diethyl ether (15 mL) at -50°C, ammonia gas was bubbled and condensed till the volume increased by 15 mL. This solution was stirred vigorously at -50°C overnight and the ammonia was allowed to evaporate by removing the cold bath. After warming to room temperature the reaction mixture was extracted between water/brine (4:1) and diethyl ether (aqueous layer was extracted with ether 3 times) and the combined organic layer was dried with anhydrous sodium sulfate and concentrated. The concentrated crude was subjected to flash chromatography with silica column and 15% ethyl acetate in hexanes as eluent to give **8** (0.82 g, 94%) as colorless oil. R_f 0.33 (20% ethyl aceate in hexanes). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.65 (s, 1H), 7.89 (d, J = 8.2 Hz, 1H), 7.53 (d, J = 8.2 Hz, 1H), 4.79 (s, 2H), 2.89 (d, J = 8.7 Hz, 1H), 2.43 (d, J = 8.7 Hz, 1H), 0.92 (s, 9H), 0.09 (s, 6H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 163.6, 148.3, 136.7, 126.0, 123.4 (q, J = 278 Hz), 119.7, 65.8, 56.6 (q, J = 36.7 Hz), 25.9, 18.4, -5.38. ^{19}F NMR (376 MHz, $CDCl_3$) δ -75.73 (s). HRMS (ESI^+) calcd for $C_{14}H_{23}F_3N_3OSi^+$ $[M + H]^+$ 334.1557, found: 334.1568.

(5-(3-(trifluoromethyl)-3*H*-diazirin-3-yl)pyridin-2-yl)methanol (1). To a solution of **8** (0.13 g, 0.390 mmol) in anhydrous diethyl ether (4.0 mL), freshly prepared Ag_2O (prepared by dropwise addition of a 10% aqueous solution of sodium hydroxide to a 10% solution of silver nitrate under constant stirring and the resultant black residue was filtered, washed with deionized water and oven dried for 2 hours) (0.45 g, 1.94 mmol) was added and stirred overnight at room temperature. The reaction was filtered and the filtrate was concentrated, to it THF (2 mL) was added and redissolved. This solution was cooled to 0°C and a 1M solution of tetrabutylammonium fluoride (TBAF) in THF (0.47 mL, 0.470 mmol) was added drop wise and stirred till the completion of reaction as indicated by TLC (about 1 hour). The reaction was extracted between water/brine (1:1) and ethyl acetate (aqueous layer was extracted with ethyl acetate 3 times) and the combined organic layer was dried with anhydrous sodium sulfate and concentrated. The resultant crude was purified by flash column chromatography with silica column and 20% ethyl acetate in

hexanes as eluent to give **1** (64.4 mg, 76%) as colorless oil. R_f 0.46 (50% ethyl acetate in hexanes). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.43 (s, 1H), 7.56 (d, J = 8.2 Hz, 1H), 7.34 (d, J = 8.3 Hz, 1H), 4.79 (s, 2H), 3.63 (s, 1H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 161.5, 146.9, 135.2, 124.1, 121.8 (q, J = 275 Hz), 120.5, 64.2, 27.1 (q, J = 41.6 Hz). ^{19}F NMR (376 MHz, $CDCl_3$) δ -65.68 (s). HRMS (ESI^+) calcd for $C_8H_7F_3N_3O^+$ $[M + H]^+$ 218.0536, found: 218.0537.

5-bromo-2-(((tert-butyldiphenylsilyl)oxy)methyl)pyrimidine (10). To a solution of (5-bromopyrimidin-2-yl)methanol[3] (5.93 g, 31.4 mmol) in anhydrous dichloromethane (DCM) (90 mL), *tert*-butyldiphenylsilyl chloride (10.3 g, 37.6 mmol) and imidazole (5.13 g, 75.3 mmol) were added and stirred overnight at room temperature. The reaction mixture was quenched with saturated ammonium chloride and extracted with dichloromethane (the aqueous layer was extracted with dichloromethane 3 times). The combined organic layers were dried with anhydrous sodium sulfate and concentrated. The resultant crude was subjected to flash chromatography with silica column and 3% ethyl acetate in hexanes as eluent to give **10** (12.2 g, 91%) as colorless oil. R_f 0.60 (10% ethyl acetate in hexanes). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.74 (s, 2H), 7.76 – 7.69 (m, 4H), 7.45 – 7.32 (m, 6H), 4.89 (s, 2H), 1.10 (s, 9H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 167.0, 157.8, 135.8, 133.4, 129.9, 127.8, 118.7, 67.1, 26.9, 19.5. HRMS (ESI^+) calcd for $C_{15}H_{18}BrN_2OSi^+$ $[M - C_6H_5^-]^+$ 349.0366, found: 349.0374.

1-(2-(((tert-butyldiphenylsilyl)oxy)methyl)pyrimidin-5-yl)-2,2,2-trifluoroethanone (11). A solution of **10** (10.0 g, 23.4 mmol) and tetramethylethylenediamine (TMEDA) (3.53 g, 30.4 mmol) in anhydrous THF (195 mL) in an argon back flushed flask was cooled to -110°C (ethanol and liquid N_2 bath). To this cold solution *n*-butyllithium (9.9 mL of 2.5 M solution in hexanes) was added very slowly under constant stirring. After 3 minutes (any longer would result in undesired side products), methyl trifluoroacetate (5.99 g, 46.8 mmol) was added dropwise and stirred at -110°C for 30 minutes and slowly warmed to RT. The reaction was quenched with saturated aqueous ammonium chloride solution and extracted with ethyl acetate (the aqueous layer was extracted with ethyl acetate 3 times). The combined organic layer was dried with anhydrous sodium sulfate and concentrated. The resultant crude was subjected to flash chromatography with neutral alumina column and 5% methanol in dichloromethane as eluent to give **11** (7.81 g, 75%) as pale yellow liquid. R_f 0.49 (5% methanol in dichloromethane, neutral alumina TLC plate). HRMS (ESI^+) calcd for $C_{23}H_{26}F_3N_2O_3Si^+$ $[M + H_3O]^+$ (since **11** exists as geminal diol) 463.1659, found: 463.1664.

1-(2-(((tert-butyldiphenylsilyl)oxy)methyl)pyrimidin-5-yl)-2,2,2-trifluoroethanone O-tosyl oxime (12). A suspension of hydroxylamine hydrochloride (7.04 g, 101 mmol) and sodium acetate trihydrate (23.6 g, 173 mmol) in ethanol (128 mL) was stirred vigorously for 10 minutes and allowed to settle. The clear supernatant (100 mL) of the above mixture was transferred to a flask with **11** (6.44 g, 14.5 mmol) and refluxed for 40 hours. Upon cooling to room temperature ethanol was removed under reduced pressure and the resultant residue was extracted between water and dichloromethane (the aqueous layer was extracted with dichloromethane 3 times) and the combined organic layer was dried with anhydrous sodium sulfate and concentrated. The concentrated crude was back flushed with argon and anhydrous dichloromethane (48 mL) was added, followed by 4-dimethylaminopyridine (DMAP) (178 mg, 1.45 mmol). After cooling the solution to -50°C, *N*,*N*-Diisopropylethylamine (DIPEA) (2.06 g, 15.9 mmol) was added followed by part-wise addition of *p*-toluenesulfonyl chloride (3.03 g, 15.9 mmol) and the reaction temperature was increased slowly in such a way that it reached 0°C in 2 hours. The reaction was quenched with water and extracted with dichloromethane (the aqueous layer was extracted with dichloromethane 3 times). The combined organic layer was dried with anhydrous sodium sulfate and concentrated. The resultant crude was subjected to flash column chromatography with silica column and 10% ethyl acetate in hexanes as eluent to give **12** (3.73 g, 42%) as pale yellow oil. R_f 0.38 (10% ethyl aceate in hexanes). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.77 (s, 2H), 7.91 (d, J = 8.3 Hz, 2H), 7.74 (dd, J = 7.9, 1.4 Hz, 4H), 7.45 – 7.36 (m, 8H), 5.02 (s, 2H), 2.49 (s, 3H), 1.14 (s, 9H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 171.5, 156.4, 149.2 (q, J = 35.1 Hz), 146.8, 135.8, 133.2, 130.8, 130.2, 129.9, 129.5, 127.9,

119.4 (q, J = 277 Hz), 118.1, 67.2, 26.9, 21.9, 19.5. ^{19}F NMR (376 MHz, $CDCl_3$) δ -66.69 (s). HRMS (ESI^+) calcd for $C_{30}H_{30}F_3N_3NaO_4SSi^+$ $[M + Na]^+$ 636.1571, found: 636.1587.

2-(((tert-butyldiphenylsilyl)oxy)methyl)-5-(3-(trifluoromethyl)diaziridin-3-yl)pyrimidine (13). A solution of **12** (1.74 g, 2.83 mmol) in anhydrous diethyl ether (15 mL) was cooled to -50°C and ammonia gas was bubbled and condensed till the volume increased by 15mL. This solution was stirred vigorously at -50°C overnight and the ammonia was allowed to evaporate by removing the cold bath. After warming to RT the reaction mixture was extracted between water/brine (4:1) and diethyl ether (the aqueous layer was extracted with ether 3 times) and the combined organic layer was dried with anhydrous sodium sulfate and concentrated. The concentrated crude was subjected to flash chromatography with silica column and 15% ethyl acetate in hexanes as eluent to give **13** (881 mg, 68%) as colorless oil. R_f 0.31 (20% ethyl aceate in hexanes). ^{1}H NMR (600 MHz, $CDCl_3$) δ 8.92 (s, 2H), 7.74 (m, 4H), 7.43 – 7.39 (m, 2H), 7.38 – 7.34 (m, 4H), 4.99 (s, 2H), 2.92 (d, J = 8.8 Hz, 1H), 2.30 (d, J = 8.8 Hz, 1H), 1.12 (s, 9H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 170.4, 156.9, 135.8, 133.3, 129.9, 127.8, 124.2, 123.0 (q, J = 278 Hz), 67.3, 55.2 (q, J = 37.5 Hz), 26.9, 19.5. ^{19}F NMR (376 MHz, $CDCl_3$) δ -75.5 (s). HRMS (ESI^+) calcd for $C_{23}H_{26}F_3N_4OSi^+$ $[M + H]^+$ 459.1822, found: 459.1833.

(5-(3-(trifluoromethyl)-3*H*-diazirin-3-yl)pyrimidin-2-yl)methanol (2). To a solution of **13** (790 mg, 1.72 mmol) in anhydrous diethyl ether (20 mL), freshly prepared Ag_2O (prepared by dropwise addition of a 10% aqueous solution of sodium hydroxide to a 10% solution of silver nitrate under constant stirring and the resultant black residue was filtered, washed with deionized water and oven dried for 2 hours) (1.90 g, 8.20 mmol) was added and stirred overnight at room temperature. The reaction was filtered and the filtrate was concentrated under reduced pressure. To the resultant concentrate THF (7 mL) was added and redissolved. To it a 1M solution of tetrabutylammonium fluoride (TBAF) in THF (1.9 mL) was added drop wise at 0°C and stirred for 30 minutes. The reaction was extracted between brine and ethyl acetate (the aqueous layer was extracted with ethyl acetate 2 times) and the combined organic layer was dried with anhydrous sodium sulfate and concentrated. The resultant crude was purified by flash column chromatography with silica column and 25% ethyl acetate in hexanes as eluent to give **2** (317 mg, 85%) as colorless oil. R_f 0.48 (50% ethyl acetate in hexanes). ^{1}H NMR (400 MHz, CD_3OD) δ 8.76 (s, 2H), 4.80 (s, 2H). ^{13}C NMR (101 MHz, CD_3OD) δ 170.3, 155.8, 121.6, 121.5 (q, J = 274 Hz), 64.2, 25.5 (q, J = 42.5 Hz). ^{19}F NMR (376 MHz, CD_3OD) δ -67.8 (s). HRMS (ESI^+) calcd for $C_7H_6F_3N_4O^+$ $[M + H]^+$ 219.0488, found: 219.0495.

(S)-4-(3-(trifluoromethyl)-3*H*-diazirin-3-yl)benzyl 2-acetamido-3-(1*H*-indol-3-yl)propanoate (14). To a solution of (4-(3-(trifluoromethyl)-3*H*-diazirin-3-yl)phenyl)methanol (30.0 mg, 139 µmol) in anhydrous dimethylformamide (DMF) (0.9 mL), *N*-acetyl tryptophan (51.3 mg, 208 µmol), EDC (41.0 mg, 214 µmol) and DMAP (26.0 mg, 213 µmol) were added at room temperature and stirred for 2 hours. The reaction was quenched with the addition of water (3 mL) and extracted with diethyl ether (the aqueous layer was extracted with ether 3 times). The combined organic layer was dried with anhydrous sodium sulfate and concentrated. The resultant crude compound was purified by silica flash column chromatography with 50% ethyl acetate in hexanes as eluent to yield **14** (41.0 mg, 66%) as colorless oil. R_f 0.23 (50% ethyl acetate in hexanes). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.33 (s, 1H), 7.51 (d, J = 7.9 Hz, 1H), 7.35 (d, J = 8.1 Hz, 1H), 7.22 – 7.16 (m, 3H), 7.16 – 7.07 (m, 3H), 6.82 (d, J = 2.1 Hz, 1H), 6.09 (d, J = 7.8 Hz, 1H), 5.06 (s, 2H), 4.99 (dt, J = 7.8, 5.6 Hz, 1H), 3.30 (d, J = 5.6 Hz, 2H), 1.95 (s, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 171.9, 170.0, 137.0, 136.3, 129.2, 128.6, 127.7, 126.8, 122.9, 122.4, 122.2 (q, J = 275 Hz), 119.8, 118.5, 111.5, 109.8, 66.3, 53.3, 28.4 (q, J = 40.4 Hz), 27.8, 23.3. ^{19}F NMR (376 MHz, $CDCl_3$) δ -65.22 (s). HRMS (ESI^+) calcd for $C_{22}H_{20}F_3N_4O_3^+$ $[M + H]^+$ 445.1482, found: 445.1480.

(S)-(5-(3-(trifluoromethyl)-3H-diazirin-3-yl)pyridin-2-yl)methyl 2-acetamido-3-(1H-indol-3-yl)propanoate (15). To a solution of **1** (24.0 mg, 111 µmol) in anhydrous DMF (0.7 mL) *N*-acetyl tryptophan (41.0 mg, 166 µmol), EDC (30.0 mg, 156 µmol) and DMAP (19.0 mg, 156 µmol) were added at room temperature and stirred for 16 hours. The reaction was quenched with the addition of water (2

mL) and extracted with diethyl ether (the aqueous layer was extracted with ether 3 times). The combined organic layer was dried with anhydrous sodium sulfate and concentrated. The resultant crude compound was purified by silica flash column chromatography with 60% ethyl acetate in hexanes as eluent to yield **15** (38.2 mg, 77%) as white solid. R_f 0.37 (70% ethyl acetate in hexanes). mp 115-117°C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.49 (s, 1H), 8.39 (s, 1H), 7.53 (d, J = 7.9 Hz, 1H), 7.42 (dd, J = 8.3, 2.3 Hz, 1H), 7.33 (d, J = 8.1 Hz, 1H), 7.20 – 7.15 (m, 1H), 7.12 – 7.04 (m, 2H), 6.98 (d, J = 2.3 Hz, 1H), 6.19 (d, J = 7.6 Hz, 1H), 5.19 (q, J = 14.0 Hz, 2H), 5.03 (dt, J = 7.6, 5.9 Hz, 1H), 3.33 (d, J = 5.9 Hz, 2H), 1.96 (s, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 171.8, 170.2, 156.8, 147.2, 136.3, 135.4, 127.7, 124.7, 123.0, 122.4, 121.8 (q, J = 275 Hz), 121.2, 119.9, 118.5, 111.5, 109.8, 66.7, 53.4, 27.8, 27.1 (q, J = 41.8 Hz), 23.2. ^{19}F NMR (376 MHz, $CDCl_3$) δ -65.52 (s). HRMS (ESI^+) calcd for $C_{21}H_{19}F_3N_5O_3^+$ $[M + H]^+$ 446.1435, found: 446.1446.

(S)-(5-(3-(trifluoromethyl)-3H-diazirin-3-yl)pyrimidin-2-yl)methyl 2-acetamido-3-(1H-indol-3-yl)propanoate (16). To a solution of **2** (22.0 mg, 101 µmol) in anhydrous DMF (0.7 mL) *N*-acetyl tryptophan (38.0 mg, 154 µmol), EDC (30.0 mg, 156 µmol) and DMAP (19.0 mg, 156 µmol) were added at room temperature and stirred for 4 hours. The reaction was quenched with the addition of water (2 mL) and extracted with diethyl ether (the aqueous layer was extracted with ether 3 times). The combined organic layer was dried with anhydrous sodium sulfate and concentrated. The resultant crude compound was purified by silica flash column chromatography with 70% ethyl acetate in hexanes as eluent to yield **16** (28.0 mg, 62%) as white solid. R_f 0.26 (70% ethyl acetate in hexanes). mp 94-96°C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.53 (s, 2H), 8.46 (s, 1H), 7.55 (d, J = 7.9 Hz, 1H), 7.34 (d, J = 8.1 Hz, 1H), 7.17 (t, J = 7.6 Hz, 1H), 7.12 – 7.05 (m, 2H), 6.15 (d, J = 7.8 Hz, 1H), 5.41 – 5.30 (m, 2H), 5.12 (dd, J = 7.7, 5.5 Hz, 1H), 3.48-3.33 (m, 2H), 1.93 (s, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 171.8, 170.1, 165.6, 155.7, 136.2, 127.9, 123.2, 122.6, 122.3, 121.4 (q, J = 275 Hz), 119.8, 118.6, 111.4, 110.0, 66.0, 53.2, 27.5, 25.9 (q, J = 42.7 Hz), 23.3. ^{19}F NMR (376 MHz, $CDCl_3$) δ -65.67 (s). HRMS (ESI^+) calcd for $C_{20}H_{18}F_3N_6O_3^+$ $[M + H]^+$ 447.1387, found: 447.1383.

4-((4-(3-(trifluoromethyl)-3H-diazirin-3-yl)benzyl)oxy)quinoline (17). To a solution of (4-(3-(trifluoromethyl)-3*H*-diazirin-3-yl)phenyl)methanol (35.4 mg, 164 µmol) in anhydrous dichloromethane (2.5 mL), phosphorous tribromide (51.0 mg, 188 µmol) was added dropwise at 0°C and warmed to room temperature. After stirring for 8 hours the reaction was quenched with the addition of brine solution (0.5 ml) and followed by the dropwise addition of saturated sodium bicarbonate solution till the bubbling stops. The dichloromethane layer was separated and further extraction of the aqueous layer was done with fresh dichloromethane (2 more times). The combined dichloromethane layer was dried with anhydrous sodium sulfate and concentrated, high vacuum should be avoided during concentration due to the volatile nature of the product. To the resultant concentrate dichloromethane (2.5 mL), water (2.5 mL), 4-quinolinol (71.0 mg, 489 µmol), tetra-*n*-butylammonium bromide (53.0 mg, 164 µmol) and sodium hydroxide (20.0 mg, 500 µmol) were added at room temperature and stirred for 16 hours. The dichloromethane layer was isolated and the water layer was extracted 2 more times with fresh dichloromethane. The combined dichloromethane layer was dried with anhydrous sodium sulfate and concentrated. The resultant crude compound was purified by silica flash column chromatography with 80% ethyl acetate in hexanes as eluent to yield **17** as white solid (43.2 mg, 77%). mp 123-125°C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.44 (d, J = 7.9 Hz, 1H), 7.63 (d, J = 7.6 Hz, 1H), 7.52 (t, J = 7.6 Hz, 1H), 7.33 (t, J = 7.5 Hz, 1H), 7.22 – 7.11 (m, 5H), 6.33 (d, J = 7.7 Hz, 1H), 5.34 (s, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 178.3, 143.7, 140.0, 137.1, 132.5, 129.4, 127.5, 127.2, 126.6, 124.1, 120.7, 116.0, 110.7, 56.0. ^{19}F NMR (376 MHz, $CDCl_3$) δ -65.23 (s). HRMS (ESI^+) calcd for $C_{18}H_{13}F_3N_3O^+$ $[M + H]^+$ 344.1005, found: 344.1014.

4-((5-(3-(trifluoromethyl)-3H-diazirin-3-yl)pyridin-2-yl)methoxy)quinoline (18). To a solution of **1** (23.0 mg, 106 µmol) in anhydrous dichloromethane (1.5 mL), phosphorous tribromide (33.0 mg, 122 µmol) was added dropwise at 0°C and warmed to room temperature. After stirring for 10 hours the reaction was quenched with the addition of brine solution (0.5 ml) and followed by the dropwise addition

of saturated sodium bicarbonate solution till the bubbling stops. The dichloromethane layer was separated and further extraction of the aqueous layer was done with fresh dichloromethane (2 more times). The combined dichloromethane layer was dried with anhydrous sodium sulfate and concentrated, high vacuum should be avoided during concentration due to the volatile nature of the product. To the resultant concentrate dichloromethane (1.5 mL), water (1.5 mL), 4-quinolinol (46.0 mg, 317 µmol), tetra-*n*-butylammonium bromide (34.0 mg, 105 µmol) and sodium hydroxide (13.0 mg, 325 µmol) were added at room temperature and stirred for 3 hours. The dichloromethane layer was isolated and the water layer was extracted 2 more times with fresh dichloromethane. The combined dichloromethane layer was dried with anhydrous sodium sulfate and concentrated. The resultant crude compound was purified by silica flash column chromatography with 2% methanol in dichloromethane as eluent to yield **18** as white solid (18.1 mg, 50%). R_f 0.39 (5% methanol in dichloromethane). mp 123-125. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.46 – 8.40 (m, 2H), 7.71 (d, J = 7.7 Hz, 1H), 7.52 (ddd, J = 8.7, 7.1, 1.6 Hz, 1H), 7.44 (dd, J = 8.3, 2.3 Hz, 1H), 7.33 (t, J = 7.6 Hz, 1H), 7.22 (d, J = 8.7 Hz, 1H), 7.02 (d, J = 8.3 Hz, 1H), 6.37 (d, J = 7.7 Hz, 1H), 5.42 (s, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 178.2, 156.8, 148.2, 144.1, 140.0, 135.7, 132.7, 127.3, 125.2, 124.2, 121.7 (q, J = 275 Hz), 120.6, 115.9, 110.8, 57.9, 27.1 (q, J = 41.8 Hz). ^{19}F NMR (376 MHz, $CDCl_3$) δ -65.49 (s). HRMS (ESI^+) calcd for $C_{17}H_{12}F_3N_4O^+$ $[M + H]^+$ 345.0958, found: 345.0966.

4-((5-(3-(trifluoromethyl)-3H-diazirin-3-yl)pyrimidin-2-yl)methoxy)quinoline (19). To a solution of **2** (23.0 mg, 105 µmol) in anhydrous dichloromethane (1.5 mL), phosphorous tribromide (33.0 mg, 122 µmol) was added dropwise at 0°C and warmed to room temperature. After stirring for 10 hours the reaction was quenched with the addition of brine solution (0.5 ml) and followed by the dropwise addition of saturated sodium bicarbonate solution till the bubbling stops. The dichloromethane layer was separated and further extraction of the aqueous layer was done with fresh dichloromethane (2 more times). The combined dichloromethane layer was dried with anhydrous sodium sulfate and concentrated, high vacuum should be avoided during concentration due to the volatile nature of the product. A solution of 4-quinolinol (60.0 mg, 413 µmol) and potassium carbonate (36.0 mg, 261 µmol) in water (1.5 mL) was stirred for 10 minutes and filtered through 0.45 µm syringe filter. This aqueous solution was added to the crude concentrate of the bromide intermediate in dichloromethane (1.5 mL) and followed by the addition of tetra-*n*-butylammonium bromide (34.0 mg, 105 µmol) at room temperature and stirred for 2 hours. The dichloromethane layer was isolated and the water layer was extracted 2 more times with fresh dichloromethane. The combined dichloromethane layer was dried with anhydrous sodium sulfate and concentrated. The resultant crude compound was purified by silica flash column chromatography with 2% methanol in dichloromethane as eluent to yield **19** as white solid (24.5 mg, 68%). R_f 0.37 (5% methanol in dichloromethane). mp 115-117°C. ^{1}H NMR (400 MHz, $CDCl_3$ with 3% CD_3OD) δ 8.54 (s, 2H), 8.40 – 8.35 (m, 1H), 7.76 (d, J = 7.7 Hz, 1H), 7.53 (ddd, J = 8.7, 7.2, 1.5 Hz, 1H), 7.35 – 7.29 (m, 2H), 6.33 (d, J = 7.7 Hz, 1H), 5.48 (s, 2H). ^{13}C NMR (101 MHz, $CDCl_3$ with 3% CD_3OD) δ 178.9, 165.4, 156.1, 144.8, 140.2, 132.6, 127.1, 127.0, 124.1, 123.2, 121.3 (q, J = 275 Hz), 115.7, 110.3, 58.1, 25.8 (q, J = 42.8 Hz). ^{19}F NMR (376 MHz, $CDCl_3$ with 3% CD_3OD) δ -65.67 (s). HRMS (ESI^+) calcd for $C_{16}H_{11}F_3N_5O^+$ $[M + H]^+$ 346.0910, found: 346.0918.

N-(6-aminohexyl)-5-((4S)-2-oxohexahydro-1*H*-thieno[3,4-d]imidazol-4-yl)pentanamide (23). The trifluoroacetic acid salt of amine **23** was synthesized as outlined by Kottani and co-workers.[1] The free base of **23** was prepared by using the following procedure using Amberlite IRA-402 (OH^- form) (Amberlite IRA-402 was freshly activated by stirring the resin in 10% NaOH aqueous solution for 10 minutes, followed by filtration and washing the resin with deionized water). After the BOC deprotection the excess trifluroacetic acid (TFA) and dichloromethane were removed in the rotavap followed by high vacuum for 6 hours with slow stirring. The resulting TFA salt residue of **23** was redissolved in water (2 mL per 100 mg of residue) and to this solution freshly activated Amberlite IRA-402 (OH^- form) was added in small portions with vigorous stirring. The addition of Amberlite IRA-402 was continued till the pH of the solution was found to be slightly basic as indicated by pH paper. The resin was filtered off and the water layer was freeze dried to get **23** as a free base. ^{1}H NMR (400 MHz, CD_3OD) δ 4.49 (ddd, J =

7.9, 5.0, 0.9 Hz, 1H), 4.30 (dd, J = 7.9, 4.5 Hz, 1H), 3.28 – 3.07 (m, 4H), 2.98 – 2.83 (m, 1H), 2.76 – 2.54 (m, 3H), 2.19 (t, J = 7.4 Hz, 2H), 1.83 – 1.23 (m, 15H). ^{13}C NMR (101 MHz, CD_3OD) δ 176.0, 166.1, 63.4, 61.6, 57.0, 41.70, 41.0, 40.2, 36.8, 31.5, 30.3, 29.8, 29.5, 27.6, 27.4, 26.9. HRMS (ESI^+) calcd for $C_{16}H_{31}N_4O_2S^+$ $[M + H]^+$ 343.2162, found: 343.2164.

(2R,3R,4S,5S,6S)-2-(acetoxymethyl)-6-(2-(benzyloxy)-2-oxoethoxy)tetrahydro-2H-pyran-3,4,5-triyl triacetate (28). To a solution of mannose pentaacetate (0.50 g, 1.28 mmol) and benzyl 2-hydroxyacetate (425 mg, 2.56 mmol) in anhydrous dichloromethane (7 mL) at 0°C, boron trifluoride etherate (0.8 mL) was added slowly and let to stir overnight at room temperature. The reaction was quenched by the drop wise addition of aqueous saturated sodium bicarbonate solution and extracted between dichloromethane and aqueous saturated sodium bicarbonate solution (the aqueous layer was extracted with dichloromethane 3 times). The combine organic layer was dried with anhydrous sodium sulfate and concentrated under reduced pressure. The concentrated crude was subjected to flash chromatography with silica column and 30% ethyl acetate in hexanes as eluent to give **28** (0.433 g, 68%) as colorless gum. R_f 0.56 (50% ethyl aceate in hexanes). ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.31 – 7.23 (m, 5H), 5.32 – 5.27 (m, 2H), 5.26 – 5.20 (m, 1H), 5.11 (d, J = 3.0 Hz, 2H), 4.88 (d, J = 1.4 Hz, 1H), 4.24 (d, J = 16.5 Hz, 1H), 4.17 (dd, J = 12.3, 5.0 Hz, 1H), 4.13 (d, J = 16.4 Hz, 1H), 4.10 – 4.05 (m, 1H), 3.97 (dd, J = 12.3, 2.4 Hz, 1H), 2.07 (s, 3H), 2.00 (s, 3H), 1.96 (s, 3H), 1.91 (s, 3H). ^{13}C NMR (63 MHz, $CDCl_3$) δ 170.3, 169.5, 169.5, 168.7, 135.0, 128.4, 128.3, 128.2, 97.7, 68.9, 68.7, 66.6, 65.6, 64.4, 62.0, 20.5, 20.4, 20.4, 20.4. HRMS (ESI^+) calcd for $C_{23}H_{32}NO_{12}^+$ $[M + NH_4]^+$ 514.1919, found: 514.1930.

2-(((2S,3S,4S,5R,6R)-3,4,5-triacetoxy-6-(acetoxymethyl)tetrahydro-2*H*-pyran-2-yl)oxy)acetic acid (24). To a solution of **28** (1.22 g, 2.46 mmol) in ethyl acetate (8 mL), palladium (10%) on carbon (0.25 g) was added and subjected to hydrogenation at 60 psi for 18 hours in a hydrogenator. After the reaction was complete as indicated by LC-MS, it was filtered using a syringe filter and the resultant filtrate was concentrated and subjected to preparative HPLC using reverse phase separation to yield the desired **24** (0.70 g, 70%) as colorless gum. ^{1}H NMR (600 MHz, CD_3OD) δ 5.35 (dd, J = 3.4, 1.7 Hz, 1H), 5.32 (dd, J = 10.1, 3.4 Hz, 1H), 5.28 - 5.23 m, 1H), 4.95 (d, J = 1.7 Hz, 1H), 4.31 – 4.27 (m, 1H), 4.26 – 4.21 (m, 2H), 4.20 – 4.17 (m, 1H), 4.13 – 4.09 (m, 1H), 2.14 (s, 3H), 2.06 (s, 3H), 2.04 (s, 3H), 1.96 (s, 3H). ^{13}C NMR (63 MHz, CD_3OD) δ 172.8, 172.4, 171.6, 171.5, 99.2, 70.7, 70.5, 70.4, 67.1, 65.3, 63.5, 20.7, 20.64, 20.6. HRMS (ESI^+) calcd for $C_{16}H_{26}NO_{12}^+$ $[M + NH_4]^+$ 424.1450, found: 424.1450.

3-(4-(bromomethyl)phenyl)-3-(trifluoromethyl)-3*H*-diazirine (25). To a solution of (4-(3-(trifluoromethyl)-3*H*-diazirin-3-yl)phenyl)methanol (95.0 mg, 0.439 mmol) in anhydrous dichloromethane (1.5 mL), triphenylphosphine (134 mg, 0.51 mmol) and carbon tetrabromide (168 mg, 0.51 mmol) were added at room temperature and stirred overnight. The reaction was quenched with the addition of pentane and filtered. The filtrate was concentrated, the resultant crude was subjected to flash chromatography with silica column and 5% ether in pentane was used as eluent. The pure fractions as identified by TLC were combined and concentrated. Due to the volatile nature of the product only mild vacuum should be employed to remove the solvents to yield **25** (112 mg, 91%) as colorless liquid. R_f = 0.61 (10% ethyl aceate in hexanes). ^{1}H NMR (400 MHz, CD_2Cl_2) δ 7.45 (d, J = 8.3 Hz, 2H), 7.20 (d, J = 8.3 Hz, 2H), 4.50 (s, 2H). ^{13}C NMR (101 MHz, CD_2Cl_2) δ 139.7, 129.4, 129.0, 126.8 (q, *J* = 1.3 Hz), 122.0 (q, *J* = 275 Hz), 32.1, 28.2 (q, *J* = 40.4 Hz). ^{19}F NMR (376 MHz, CD_2Cl_2) δ -65.67 (s).

2-(bromomethyl)-5-(3-(trifluoromethyl)-3*H*-diazirin-3-yl)pyridine (26). To a solution of **1** (59.0 mg, 272 µmol) in anhydrous dichloromethane (0.8 mL), triphenylphosphine (79.0 mg, 300 µmol) and carbon tetrabromide (99.0 mg, 299 µmol) were added at room temperature and stirred overnight. The reaction was quenched with the addition of pentane and filtered. The filtrate was concentrated, the resultant crude was subjected to flash chromatography with silica column and 5% ether in pentane was used as eluent. The pure fractions as identified by TLC were combined and concentrated. Due to the volatile nature of the product only mild vacuum should be employed to remove the solvents to yield **26** (71.0 mg, 93%) as colorless liquid. R_f = 0.54 (10% ethyl ether in pentane). ^{1}H NMR (400 MHz, CD_2Cl_2) δ 8.44 (d, J = 2.4

Hz, 1H), 7.57 (dd, J = 8.3, 2.4 Hz, 1H), 7.49 (d, J = 8.3 Hz, 1H), 4.55 (s, 2H). ^{13}C NMR (101 MHz, CD_2Cl_2) δ 159.0, 148.2 (q, J = 1.6 Hz), 135.9 (q, J = 1.2 Hz), 125.1, 123.7, 122.3 (q, J = 274 Hz), 33.5, 27.7 (q, J = 41.8 Hz). ^{19}F NMR (376 MHz, CD_2Cl_2) δ -65.99 (s). HRMS (ESI^+) calcd for $C_8H_6BrF_3N_3^+$ $[M + H]^+$ 279.9692, found: 279.9688.

2-(bromomethyl)-5-(3-(trifluoromethyl)-3*H*-diazirin-3-yl)pyrimidine (27). To a solution of **2** (30.0 mg, 137 µmol) in anhydrous dichloromethane (1.5 mL) at 0°C, phosphorus tribromide (44.4 mg, 164 µmol) was added slowly. The reaction was slowly warmed to RT and stirred overnight. The reaction was quenched with dropwise addition of saturated aqueous sodium bicarbonate and extracted between dichloromethane and aqueous sodium bicarbonate (the aqueous layer was extracted with dichloromethane 3 times). The combined organic layer was dried with anhydrous sodium sulfate and concentrated. Due to the volatile nature of the product only mild vacuum should be employed to remove the solvents to yield **27**. The product obtained was used without further purification for the next step.

5-(2-oxohexahydro-1*H*-thieno[3,4-d]imidazol-4-yl)-*N*-(6-((4-(3-(trifluoromethyl)-3*H*-diazirin-3-yl)benzyl)amino)hexyl)pentanamide (33). To a solution of **23** (53.0 mg, 0.155 mmol) in anhydrous DMF (1.5 mL), a solution of **25** (20 mg, 71.7 µmol) in DMF (0.5 mL) was added slowly at room temperature and stirred for 3 hours. The reaction mixture was diluted with acetonitrile (10 mL) and subjected to reverse phase preparative HPLC purification. The fractions with the desired compound were collected and concentrated by removal of acetonitrile on a rotary evaporator and freeze drying the resultant aqueous solution to yield a white powder of **33** (10.1 mg, 22 %) as a TFA salt. This salt was used for the next step without further purification.

(2R,3S,4S,5S,6S)-2-(acetoxymethyl)-6-(2-oxo-2-((6-(5-(2-oxohexahydro-1*H*-thieno[3,4-d]imidazol-4-yl)pentanamido)hexyl)(4-(3-(trifluoromethyl)-3*H*-diazirin-3-yl)benzyl)amino)ethoxy)tetrahydro-2*H*-pyran-3,4,5-triyl triacetate (36). To a solution of TFA salt of **33** (10.0 mg, 15.7 µmol) in anhydrous DMF (1 mL), **24** (16.0 mg, 39.4 µmol), EDC (7.1 mg, 37.0 µmol) and DMAP (5.0 mg, 40.9 µmol) were added at room temperature and stirred overnight. The reaction mixture was diluted with acetonitrile (10 mL) and subjected to reverse phase preparative HPLC purification. The fractions with the desired compound were collected and concentrated by removal of acetonitrile on a rotary evaporator and freeze drying the resultant aqueous solution to yield **36** (7.5 mg, 51%) as white amorphous solid. The NMR analysis indicated the existence of **36** as rotamers. ^{1}H NMR (600 MHz, CD_3OD) δ 7.37 (d, J = 8.1 Hz, 2H), 7.26 (d, 8.1 Hz, 2H), 5.40 – 5.17 (m, 3H), 4.99 (d, J = 1.5 Hz, 1H), 4.68 - 4.60 (m, 2H), 4.52 – 4.46 (m, 2H), 4.43 – 4.21 (m, 3H), 4.21 – 4.09 (m, 2H), 4.04 – 3.97 (m, 1H), 3.36 – 3.32 (m, 1H), 3.29 – 3.25 (m, 1H), 3.23 – 3.17 (m, 1H), 3.17 – 3.10 (m, 2H), 2.92 (ddd, J = 12.7, 4.9, 3.7 Hz, 1H), 2.70 (d, J = 12.7 Hz, 1H), 2.21 – 2.16 (m, 2H), 2.16 – 2.11 (m, 3H), 2.08 – 1.98 (m, 6H), 1.98 – 1.94 (m, 3H), 1.77 – 1.52 (m, 6H), 1.51 – 1.39 (m, 4H), 1.36 - 1.26 (m, 4H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 170.9, 170.8, 170.3, 170.2, 169.9, 169.8, 138.8, 137.8, 129.1, 128.5, 128.4, 127.5, 127.0, 126.8, 122.1 (q, J = 275 Hz), 97.8, 69.3, 69.1, 65.9, 65.9, 65.4, 65.2, 62.4, 61.3, 61.0, 55.4, 49.9, 48.2, 46.9, 46.5, 40.4, 39.6, 39.5, 35.4, 29.8, 29.3, 29.0, 28.5, 27.7, 27.0, 26.5, 26.2, 25.4, 20.9, 20.9, 20.9, 20.8, 20.8. ^{19}F NMR (376 MHz, $CDCl_3$) δ -65.31 (s). HRMS (ESI^+) calcd for $C_{41}H_{55}F_3N_6NaO_{13}S^+$ $[M + Na]^+$ 951.3392, found: 951.3418.

5-(2-oxohexahydro-1*H*-thieno[3,4-d]imidazol-4-yl)-*N*-(6-(*N*-(4-(3-(trifluoromethyl)-3*H*-diazirin-3-yl)benzyl)-2-(((2S,3S,4S,5S,6R)-3,4,5-trihydroxy-6-(hydroxymethyl)tetrahydro-2*H*-pyran-2-yl)oxy)acetamido)hexyl)pentanamide (20). To a solution of **36** (6.2 mg, 6.67 µmol) in anhydrous methanol (0.5 mL), 25% sodium methoxide in methanol solution (40 µL) was added and let to stir for 3 hours at room temperature. The reaction was quenched with the addition of 0.1% TFA in methanol solution (5 mL) and subjected to reverse phase preparative HPLC purification. The fractions with the desired compound were collected and concentrated by removal of acetonitrile on a rotary evaporator and freeze drying the resultant aqueous solution to yield **20** (2.5 mg, 49%) as white amorphous solid. The NMR analysis indicated the existence of **20** as rotamers. ^{1}H NMR (600 MHz, CD_3OD) δ 7.34 (d, J = 7.2 Hz, 2H), 7.22 (d, J = 7.2 Hz, 2H), 4.85 (d, J = 1.6 Hz, 1H), 4.61 (d, J = 8.6 Hz, 2H), 4.49 – 4.38 (m, 2H),

4.32 – 4.25 (m, 1.5H), 3.93 (dd, J = 3.4, 1.7 Hz, 0.5H), 3.86 – 3.81 (m, 1H), 3.76 – 3.70 (m, 1H), 3.69 – 3.41 (m, 3H), 3.26 – 3.21 (m, 1H), 3.20 – 3.15 (m, 1H), 3.14 – 3.09 (m, 2H), 2.93- 2.88 (m, 1H), 2.67 (d, J = 8.7 Hz, 1H), 2.16 (t, J = 7.3 Hz, 2H), 1.74 – 1.37 (m, 10H), 1.32 – 1.24 (m, 5H). ^{13}C NMR (126 MHz, CD_3OD) δ 176.0, 171.4, 166.1, 141.2, 140.5, 129.5, 128.9, 128.6, 128.2, 127.8, 101.4, 75.4, 75.2, 72.4, 72.4, 71.7, 68.6, 68.4, 66.5, 65.6, 65.2, 63.4, 63.0, 62.8, 61.6, 57.0, 41.0, 40.1, 36.8, 30.3, 29.8, 29.5, 29.4, 28.1, 27.6, 27.5, 27.4, 26.9. ^{19}F NMR (376 MHz, CD_3OD) δ -65.42 (s), -65.48 (s). HRMS (ESI^+) calcd for $C_{33}H_{47}F_3N_6NaO_9S^+$ $[M + Na]^+$ 783.2970, found: 783.2973.

5-(2-oxohexahydro-1*H*-thieno[3,4-d]imidazol-4-yl)-*N*-(6-(((5-(3-(trifluoromethyl)-3*H*-diazirin-3-yl)pyridin-2-yl)methyl)amino)hexyl)pentanamide (34). To a solution of **23** (70.0 mg, 205 µmol) in anhydrous DMF (2 mL), a solution of **26** (20 mg, 71.4 µmol) in anhydrous DMF (0.5 mL) was added dropwise at room temperature and stirred for 3 hours. The reaction mixture was diluted with acetonitrile (10 mL) and subjected to reverse phase preparative HPLC purification. The fractions with the desired compound were collected and concentrated by removal of acetonitrile on a rotary evaporator and freeze drying the resultant aqueous solution to yield white powder of **34** (13.0 mg, 29 %) as a TFA salt. This salt was used for the next step without further purification.

(2R,3R,4S,5S,6S)-2-(acetoxymethyl)-6-(2-oxo-2-((6-(5-(2-oxohexahydro-1*H*-thieno[3,4-d]imidazol-4-yl)pentanamido)hexyl)((5-(3-(trifluoromethyl)-3*H*-diazirin-3-yl)pyridin-2-yl)methyl)amino)ethoxy)tetrahydro-2*H*-pyran-3,4,5-triyl triacetate (37). To a solution of the TFA salt of **34** (7.0 mg, 11.0 µmol) in DMF (1 mL), **24** (11.2 mg, 27.6 µmol), EDC (5.0 mg, 26.1 µmol) and DMAP (3.5 mg, 28.6 µmol) were added at room temperature and stirred overnight. The reaction mixture was extracted between brine and ethyl acetate (the aqueous layer was extracted with ethyl acetate 3 times) and the combined organic layer was dried with anhydrous sodium sulfate and concentrated. The resultant crude was subjected to reverse phase preparative HPLC purification. The fractions with the desired compound were collected and concentrated by removal of acetonitrile on a rotary evaporator and freeze drying the resultant aqueous solution to yield **37** (7.5 mg, 73%) as white amorphous solid. The NMR analysis indicated the existence of **37** as rotamers in 1:1 ratio. ^{1}H NMR (600 MHz, $CDCl_3$) δ 8.45 (s, 0.5H), 8.40 (s, 0.5H), 7.61 (d, J = 8.0 Hz, 0.5H), 7.57 (d, J = 8.0 Hz, 0.5H), 7.37 (d, J = 8.0 Hz, 0.5H), 7.27 (d, J = 8.0 Hz, 0.5H), 6.77 - 6.67 (m, 0.5H), 6.29 (s, 0.5H), 6.13 (s, 0.5H), 5.37 – 5.24 (m, 2.5H), 5.19 - 5.13 (m, 1H), 5.01 - 4.88 (m, 1H), 4.73 - 4.55 (m, 3H), 4.43 – 4.31 (m, 3H), 4.29 - 4.20 (m, 1H), 4.15 – 3.96 (m, 2H), 3.41 – 3.11 (m, 5H), 2.99 - 2.87 (m, 1H), 2.77 (t, J = 13.6 Hz, 1H), 2.32 – 2.17 (m, 2H), 2.14 (d, 3H), 2.10 - 2.02 (m, 6H), 1.98 (d, 3H), 1.77 - 1.56 (m, 5H), 1.54 – 1.39 (m, 5H), 1.35 - 1.21 (m, 4H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 174.0, 173.8, 170.9, 170.8, 170.2, 170.1, 169.8, 168.9, 168.4, 158.4, 157.8, 148.0, 146.9, 138.0, 125.1, 124.9, 124.7, 122.9, 122.7, 120.9, 120.7, 97.9, 97.8, 69.3, 69.2, 69.1, 66.0, 65.9, 65.1, 62.4, 60.9, 60.8, 55.5, 51.9, 50.3, 48.1, 46.8, 40.5, 40.4, 39.5, 39.4, 35.7, 35.5, 29.4, 29.2, 28.7, 27.9, 27.8, 27.3, 27.1, 27.0, 26.6, 26.5, 26.3, 25.5, 21.0, 20.9, 20.8, 20.7. HRMS (ESI^+) calcd for $C_{40}H_{55}F_3N_7O_{13}S^+$ $[M + H]^+$ 930.3525, found: 930.3532.

5-(2-oxohexahydro-1*H*-thieno[3,4-d]imidazol-4-yl)-*N*-(6-(*N*-((5-(3-(trifluoromethyl)-3*H*-diazirin-3-yl)pyridin-2-yl)methyl)-2-(((2S,3S,4S,5S,6R)-3,4,5-trihydroxy-6-(hydroxymethyl)tetrahydro-2*H*-pyran-2-yl)oxy)acetamido)hexyl)pentanamide (21). To a solution of **37** (7.5 mg, 8.06 µmol) in anhydrous methanol (0.9 mL), 25% sodium methoxide in methanol solution (10 µL) was added and let to stir for 3 hours at room temperature. The reaction was quenched with the addition of 0.1% TFA in methanol solution (3 mL) and subjected to reverse phase preparative HPLC purification. The fractions with the desired compound were collected and concentrated by removal of acetonitrile on a rotary evaporator and freeze drying the resultant aqueous solution to yield **21** (3.8 mg, 62%) as white amorphous solid. The NMR analysis indicated the existence of **21** as rotamers. ^{1}H NMR (500 MHz, CD_3OD) δ 8.49 (s, 0.5H), 8.43 (s, 0.5H), 7.78 – 7.68 (m, 1H), 7.49 – 7.40 (m, 1H), 4.86 (s, 4H), 4.74 - 4.70 (m, 2H), 4.53 – 4.36 (m, 3H), 4.34 - 4.28 (m, 1H), 3.98 – 3.82 (m, 2H), 3.80 – 3.52 (m, 5H), 3.42 - 3.33 (m, 2H), 3.25 - 3.12 (m, 3H), 2.97 - 2.89 (m, 1H), 2.74 - 2.68 (m, 1H), 2.25 - 2.16 (m, 2H), 1.79 – 1.56 (m, 5H), 1.56 –

1.39 (m, 5H), 1.38 - 1.24 (m, 4H). ^{13}C NMR (126 MHz, CD_3OD) δ 176.0, 175.9, 171.9, 171.5, 166.1, 160.7, 160.0, 148.9, 148.3, 137.0, 136.9, 125.4, 125.0, 123.3, 123.2 (q, J = 274 Hz), 123.0, 101.4, 75.4, 75.2, 72.4, 71.7, 68.6, 68.5, 65.7, 65.2, 63.4, 63.0, 62.9, 61.6, 57.0, 52.5, 51.8, 47.9, 41.0, 40.1, 36.8, 30.3, 30.2, 29.8, 29.5, 28.2, 27.6, 27.5, 27.4, 26.9. ^{19}F NMR (376 MHz, CD_3OD) δ -67.44 (s), -67.53 (s). HRMS (ESI^+) calcd for $C_{32}H_{46}F_3N_7NaO_9S^+$ $[M + Na]^+$ 784.2922, found: 784.2924.

5-(2-oxohexahydro-1*H*-thieno[3,4-d]imidazol-4-yl)-*N*-(6-(((5-(3-(trifluoromethyl)-3*H*-diazirin-3-yl)pyrimidin-2-yl)methyl)amino)hexyl)pentanamide (35). To a solution of **23** (70.0 mg, 205 μmol) in anhydrous DMF (1.5 mL), a solution of **27** (137 μmol) in anhydrous DMF (0.5 mL) was added dropwise slowly at room temperature and stirred for 6 hours. The reaction mixture was diluted with acetonitrile (10 mL) and subjected to reverse phase preparative HPLC purification. The fractions with the desired compound were collected and concentrated by removal of acetonitrile on a rotary evaporator and freeze drying the resultant aqueous solution to yield white powder of **35** (37.2 mg, 42%) as a TFA salt. This salt was used for the next step without further purification.

(2R,3S,4S,5S,6S)-2-(acetoxymethyl)-6-(2-oxo-2-((6-(5-(2-oxohexahydro-1*H*-thieno[3,4-d]imidazol-4-yl)pentanamido)hexyl)((5-(3-(trifluoromethyl)-3*H*-diazirin-3-yl)pyrimidin-2-yl)methyl)amino)ethoxy)tetrahydro-2*H*-pyran-3,4,5-triyl triacetate (38). To a solution of **24** (16.0 mg, 39.4 μmol) in anhydrous DMF (2 mL), EDC (8.50 mg, 44.3 μmol) and DMAP (5.70 mg, 46.7 μmol) were added at room temperature and stirred for 10 minutes. This solution was then added to a solution of TFA salt of **35** (15 mg, 23.4 μmol) in anhydrous DMF (0.5 mL) and stirred overnight. The reaction mixture was extracted between brine and ethyl acetate (the aqueous layer was extracted with ethyl acetate 3 times) and the combined organic layer was dried with anhydrous sodium sulfate and concentrated. The resultant crude was subjected to reverse phase preparative HPLC purification. The fractions with the desired compound were collected and concentrated by removal of acetonitrile on a rotary evaporator and freeze drying the resultant aqueous solution to yield **38** (15.6 mg, 71%) as white amorphous solid. The NMR analysis indicated the existence of **38** as rotamers. ^{1}H NMR (500 MHz, CD_3OD) δ 8.75 (s, 1H), 8.70 (s, 1H), 5.41 – 5.16 (m, 3H), 5.08 – 5.03 (m, 1H), 4.98 (d, J = 1.6 Hz, 1H), 4.85 – 4.75 (m, 2H), 4.53 – 4.47 (m, 2H), 4.44 (d, J = 2.3 Hz, 1H), 4.33 – 4.28 (m, 1H), 4.26 – 3.99 (m, 4H), 3.50 – 3.40 (m, 2H), 3.23 – 3.12 (m, 3H), 2.93 (ddd, J = 12.7, 4.9, 3.8 Hz, 1H), 2.71 (d, J = 12.7 Hz, 1H), 2.22 – 2.17 (m, 2H), 2.16 – 2.11 (m, 3H), 2.09 – 2.02 (m, 6H), 1.97 – 1.93 (m, 3H), 1.76 – 1.58 (m, 5H), 1.57 – 1.27 (m, 10H). ^{13}C NMR (126 MHz, CD_3OD) δ 175.9, 175.9, 172.3, 172.3, 171.5, 171.4, 171.4, 171.3, 171.3, 171.3, 171.0, 169.0, 168.7, 166.1, 157.7, 157.3, 123.5, 123.0 (q, J = 275 Hz), 122.9, 98.9, 98.9, 70.6, 70.5, 70.4, 70.3, 70.3, 70.1, 67.1, 66.9, 66.9, 66.9, 63.4, 63.3, 61.6, 57.0, 57.0, 53.6, 52.6, 41.0, 40.2, 40.2, 36.8, 30.3, 30.3, 29.8, 29.8, 29.7, 29.5, 29.5, 28.1, 27.7, 27.6, 27.5, 26.9, 20.7, 20.7, 20.6, 20.6, 20.6, 20.5. ^{19}F NMR (376 MHz, CD_3OD) δ -66.08, -66.22. HRMS (ESI^+) calcd for $C_{39}H_{53}F_3N_8NaO_{13}S^+$ $[M + Na]^+$ 953.3297, found: 953.3305.

5-(2-oxohexahydro-1*H*-thieno[3,4-d]imidazol-4-yl)-*N*-(6-(*N*-((5-(3-(trifluoromethyl)-3*H*-diazirin-3-yl)pyrimidin-2-yl)methyl)-2-(((2S,3S,4S,5S,6R)-3,4,5-trihydroxy-6-(hydroxymethyl)tetrahydro-2*H*-pyran-2-yl)oxy)acetamido)hexyl)pentanamide (22). To a solution of **38** (6.8 mg, 7.30 μmol) in anhydrous methanol (1 mL), 25% sodium methoxide in methanol solution (10 μL) was added and let to stir for 3 hours at room temperature. The reaction was quenched with the addition of 0.1% TFA in methanol solution (3 mL) and subjected to reverse phase preparative HPLC purification. The fractions with the desired compound were collected and concentrated by removal of acetonitrile on a rotary evaporator and freeze drying the resultant aqueous solution to yield **22** (3.7 mg, 66%) as white amorphous solid. The NMR analysis indicated the existence of **22** as rotamers. ^{1}H NMR (600 MHz, CD_3OD) δ 8.76 (s, 1H), 8.71 (s, 1H), 4.83 – 4.78 (m, 3H), 4.51 – 4.35 (m, 3H), 4.31 (dd, J = 7.7, 4.5 Hz, 1H), 3.94 – 3.81 (m, 1.5H), 3.78 – 3.53 (m, 4H), 3.49 – 3.36 (m, 2.5H), 3.23 – 3.12 (m, 3H), 2.93 (ddd, J = 12.7, 5.0, 3.1 Hz, 1H), 2.71 (dd, J = 12.7, 3.0 Hz, 1H), 2.19 (t, J = 7.4 Hz, 2H), 1.78 – 1.56 (m, 5H), 1.55 – 1.41 (m, 5H), 1.41 – 1.26 (m, 5H). ^{13}C NMR (126 MHz, CD_3OD) δ 176.0, 175.9, 172.1, 171.7, 169.1, 168.7,

166.1, 157.6, 157.3, 123.5, 123.0 (q, J = 274 Hz), 122.9, 101.3, 75.3, 75.2, 72.4, 72.3, 71.7, 71.7, 68.5, 68.5, 65.6, 65.1, 63.4, 63.0, 62.9, 61.6, 57.0, 57.0, 53.4, 52.7, 41.0, 40.2, 36.8, 30.3, 30.3, 29.8, 29.6, 29.5, 28.3, 27.7, 27.6, 27.5, 27.5, 26.9. ^{19}F NMR (376 MHz, CD_3OD) δ -66.17, -66.30. HRMS (ESI^+) calcd for $C_{31}H_{46}F_3N_8O_9S^+$ $[M + H]^+$ 763.3055, found: 763.3074.

General procedure for ambient light stability evaluation. Photolabel (**1**, **2,** or **3**) (1 mg) was dissolved in *d*4-methanol (500 µL) in a 5 mm NMR tube. The ^{19}F NMR of this solution was recorded before any exposure to ambient light as the zero day reading. The NMR tube was then placed directly under two linear fluorescent lamps (28 W each) at room temperature and the ^{19}F NMR of this solution was recorded periodically (4, 7, 14, 18, 26 and 31 days). The analysis was repeated in duplicate.

General procedure for thermal stability evaluation. Photolabel (**1**, **2,** or **3**) (1 mg) was dissolved in *d*4-methanol (500 µL) in a 5 mm NMR tube. The ^{19}F NMR of this solution was recorded before any exposure to ambient light as the zero day reading. The NMR tube was then placed in the dark at room temperature and the ^{19}F NMR of this solution was recorded periodically (8, 14, and 31 days). The analysis was repeated in duplicate.

Procedure for evaluation of stability under incandescent light. Photolabel (**1**, **2,** or **3**) (1 mg) was dissolved in *d*4-methanol (500 µL) in a 5 mm NMR tube. The ^{19}F NMR of this solution was recorded before any exposure to incandescent light as the zero day reading. The NMR tube was then placed directly under an incandescent lamp (65 W) at room temperature and the ^{19}F NMR of this solution was recorded periodically (1, 3, 5 and 15 days).

General procedure for aqueous solubility measurement. Aqueous solubility was measured using a HPLC-MS method.[4] A calibration curve was generated by plotting the area count (HPLC) against the known concentration of compound prepared by serial dilution (1000 µM to 0.017 µM) using DMSO as the solvent. For generating calibration curve in the low concentration range (0.017 – 1.00 µM) the area count obtained by LC-MS (selective ion mode) was used. For generating calibration curve in the high concentration range (5.00 - 1000 µM) the area count obtained by HPLC-DAD (UV absorbance) was used. A 100 mM solution of the compound (6 µL) was added to the 100 mM phosphate buffer solution (594 µL) (pH 7.4 or 5.0) to get a 100 fold dilution. This mixture was incubated at 21°C for 18 hours, filtered using a membrane filter (PVDF, 0.2 µm), and injected into the HPLC. The concentration in the aqueous solution was then determined by interpolating the sample's area count with the respective calibration curve. The analysis was performed in four replicates.

General procedure for photolabeling. To a solution of concanavalin A (Con A) (0.2 mg) in acetate buffer (10 mM, pH=5 with calcium chloride (1 mM), manganese (II) chloride (1 mM) and sodium chloride (200 mM)) (1 ml), the photoaffinity label (**20**, **21** or **22**) solution (10 mM) in methanol (3 µL) was added at 0°C. This solution was bubbled with a gentle stream of nitrogen gas, using a clean needle, for 5 minutes and incubated in the dark at 0°C for 30 minutes. The sample was then transferred to a disposable cuvette and photoactivated for 10 minutes using a 150 W XBO xenon short-arc UV lamp fitted with a filter that cuts off light with wavelengths less than 320 nm. The resultant sample can be stored at -80°C and used for SDS gel, Western blot analysis or mass spectroscopic analysis.

Photolabeling in the presence of mannose. For the competitive binding studies with mannose, to a solution of Con A (0.2 mg) in acetate buffer (10 mM, pH=5 with calcium chloride (1 mM), manganese (II) chloride (1 mM) and sodium chloride (200 mM)) (1 ml) a aqueous solution of mannose (30 µL of 100 mM solution for 100 fold [or] 30 µL of 1 M solution for 1000 fold) was added and mixed. To this the photoaffinity label (**20**, **21** or **22**) solution (10 mM) in methanol (3 µL) was added at 0°C. This solution was bubbled with a gentle stream of nitrogen gas, using a clean needle, for 5 minutes and incubated in the dark at 0°C for 30 minutes. The sample was then transferred to a disposable cuvette and photoactivated for 10 minutes using a 150 W XBO xenon short-arc UV lamp fitted with a filter that cuts off light with wavelengths less than 320 nm. The resultant sample can be stored at -80°C and used for SDS gel and Western blot analysis.

General procedure for Western blot analysis. Upon running the SDS gel, the PVDF membrane and gel was shaken in transfer buffer for 15 minutes. The holder cassette was placed opened in a shallow vessel such that the black panel was lying flat on the bottom of the vessel. Fiber pad presoaked with transfer buffer was then placed on the black panel of the holder cassette and a presoaked filter paper was placed over it. The equilibrated gel was carefully placed over the filter paper to avoid any air bubble getting trapped between the layers. Then the PVDF membrane was laid over the gel with care towards avoiding any air pocket between the layers. A presoaked filter paper was placed over the membrane followed by a filter pad. The resultant sandwich was firmly secured and the cassette was closed. The cassette holder was placed in the tank such that the black panel of the holder was on the black panel electrode. Insert the ice pack on the other side of the tank and place the tank on a magnetic stirrer. The tank was filled to the top row of circles in the cassette with transfer buffer. The magnetic stirrer was turned on and the lid was closed with black wire to black panel, red wire to red panel. The unit was connected to a power supply and ran at constant voltage of 70 V for 60 minutes. The membrane was carefully removed and the membrane was blocked with 5% (w/v) NFDM (non-fat dry milk) in tris saline Tween-20 buffer (TBS-T) for 1 hour at room temperature. The blocked membrane was then incubated overnight on a shaker at 4°C with anti-biotin−peroxidase antibody in TBS-T buffer containing 1% NFDM. The membrane was washed five times (10 minutes each time on a shaker) with TBS-T buffer. The membrane was incubated for 5 minutes at room temperature with SuperSignal West Pico chemiluminescence substrate as per manufacturer's protocol. The resultant chemiluminescence of the bands was recorded on a photographic film and developed using a film processor.

Experimental Procedures for Mass Spectrometry

Sample preparation. The sample was subjected to protein precipitation by adding cold (-20°C) acetone (1.2 mL) to the solution of photolabeled sample (0.3 mL) and vortexed. The resultant sample was incubated for 60 minutes at -20°C. The sample was placed in a centrifuge precooled in a cold room (4°C) and centrifuged at 13,000 x g for 10 minutes. The supernatant was carefully decanted without dislodging the protein pellet. The remaining acetone was allowed to evaporate by keeping the tube uncapped at room temperature for 30 minutes (the protein might not dissolve properly if the pellet was over dried). The pellet was redissolved in 50 mM ammonium bicarbonate buffer (50 µL) and 3 µg of sequencing grade modified trypsin (Promega catalog # V5111) in trypsin resuspension buffer (provided with the commercial trypsin) (10 µL) was added to it. The resultant solution was incubated at 37°C for 18 hours followed by the addition of 5% formic acid in acetonitrile (200 µL). The solution was speed vacuumed to dryness and another portion of 5% formic acid in acetonitrile (200 µL) was added and speed vacuumed till all the liquid was removed. The resultant residue was redissolved in 0.1% formic acid aqueous solution (50 µL) and sonicated for 5 minutes. The sample was subjected to centrifugation at 5,000 x g for 5 minutes and the supernatant was taken for mass spectroscopic analysis.

Mass spectroscopic analysis. Peptides resulting from the digestion were analyzed by liquid chromatography mass spectrometry (LC MS). Briefly, chromatography was performed using a Nano-LC Ultra 2D+ (Eksigent, Dublin, CA) equipped with a Proteopep 2 IntegraFrit trapping column (100 µm i.d. x 2.5 cm; C18, 5 µm, 300Å) and a Proteopep 2 IntegraFrit analytical column (75 µm i.d. x 10 cm; C18, 5 µm, 300Å, New Objective, Woburn, MA). Sample (5 µL for a total of 1.5 µg) was loaded onto the trap column at 3 µL/min (Solvent A) for 7 minutes, after which a valve was switched to include the analytical column. Peptides were then eluted with a gradient (300 nL/min) of 2% B to 40% B over 80 minutes (Solvent A: 0.5% formic acid in water, Solvent B: 0.5% formic acid in acetonitrile). Nano-LC effluent was analyzed on-line by positive-ion micro-electrospray with a linear ion trap Orbitrap XL (LTQ OrbiTrap XL, Thermo Fisher Corp., Bremen, Germany) operated in 'top-5 data-dependent' acquisition mode. Labeled peptides were found by subtractive analysis between non-labeled and labeled samples. MS/MS results were sequenced heuristically.

References

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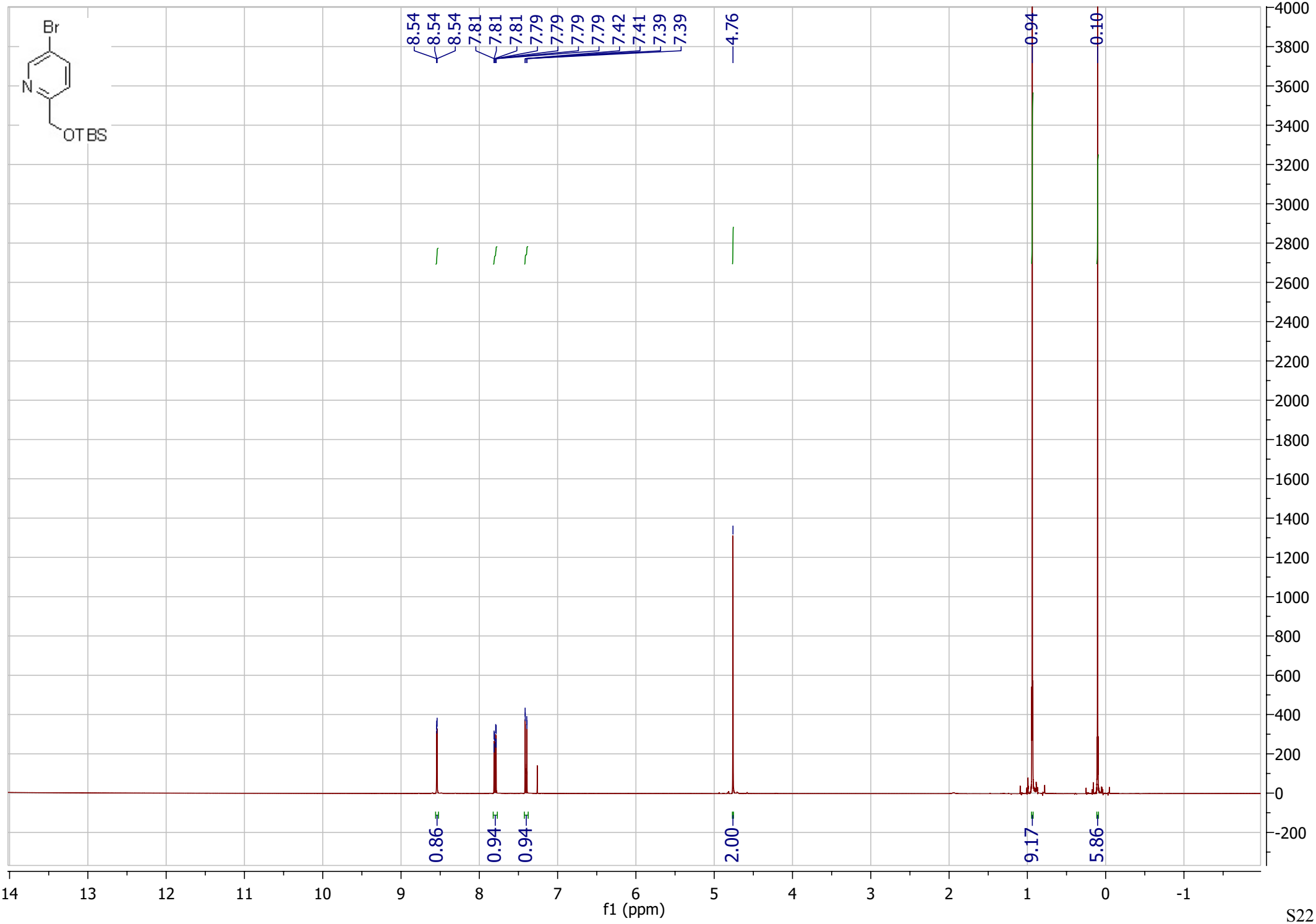
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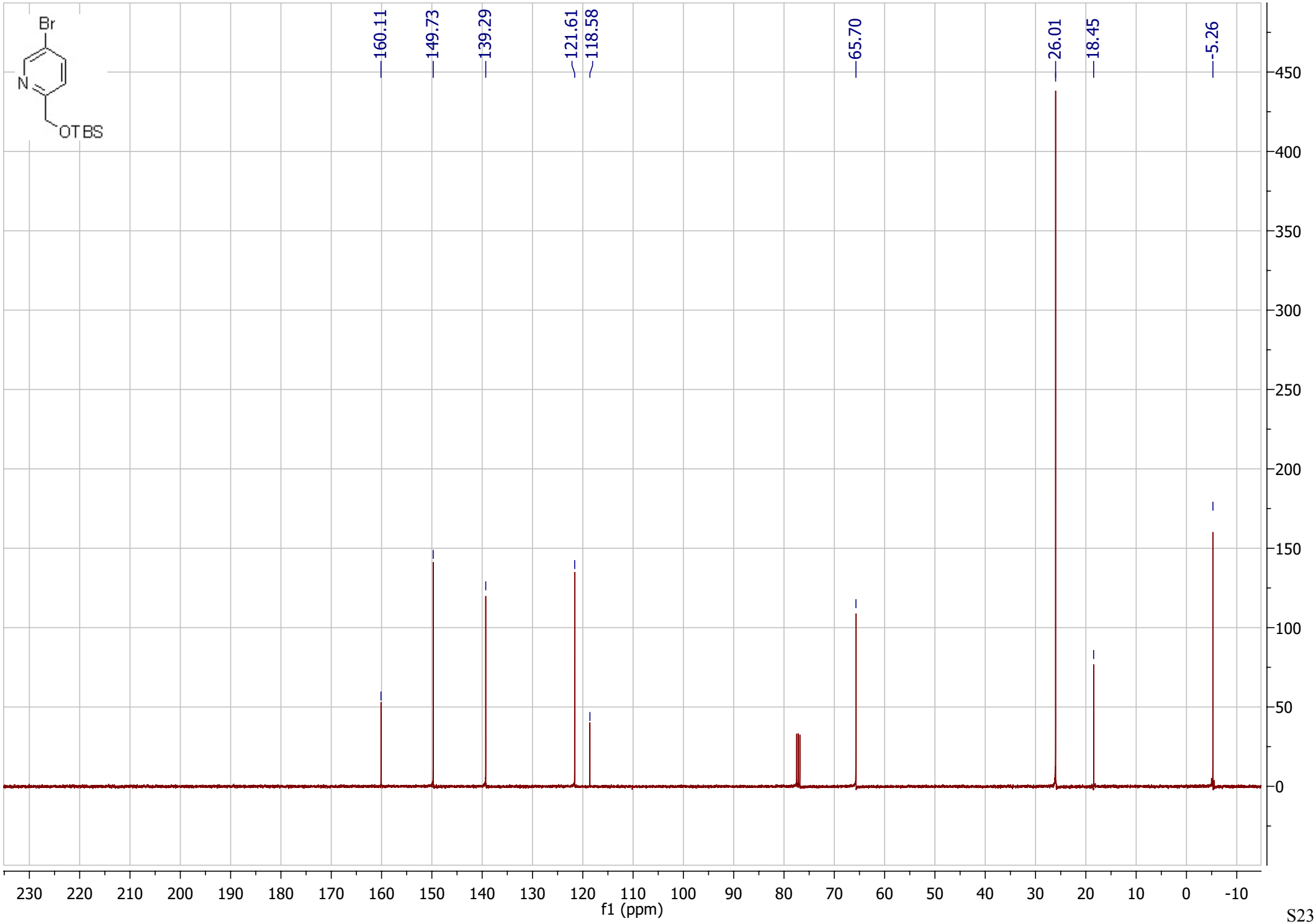
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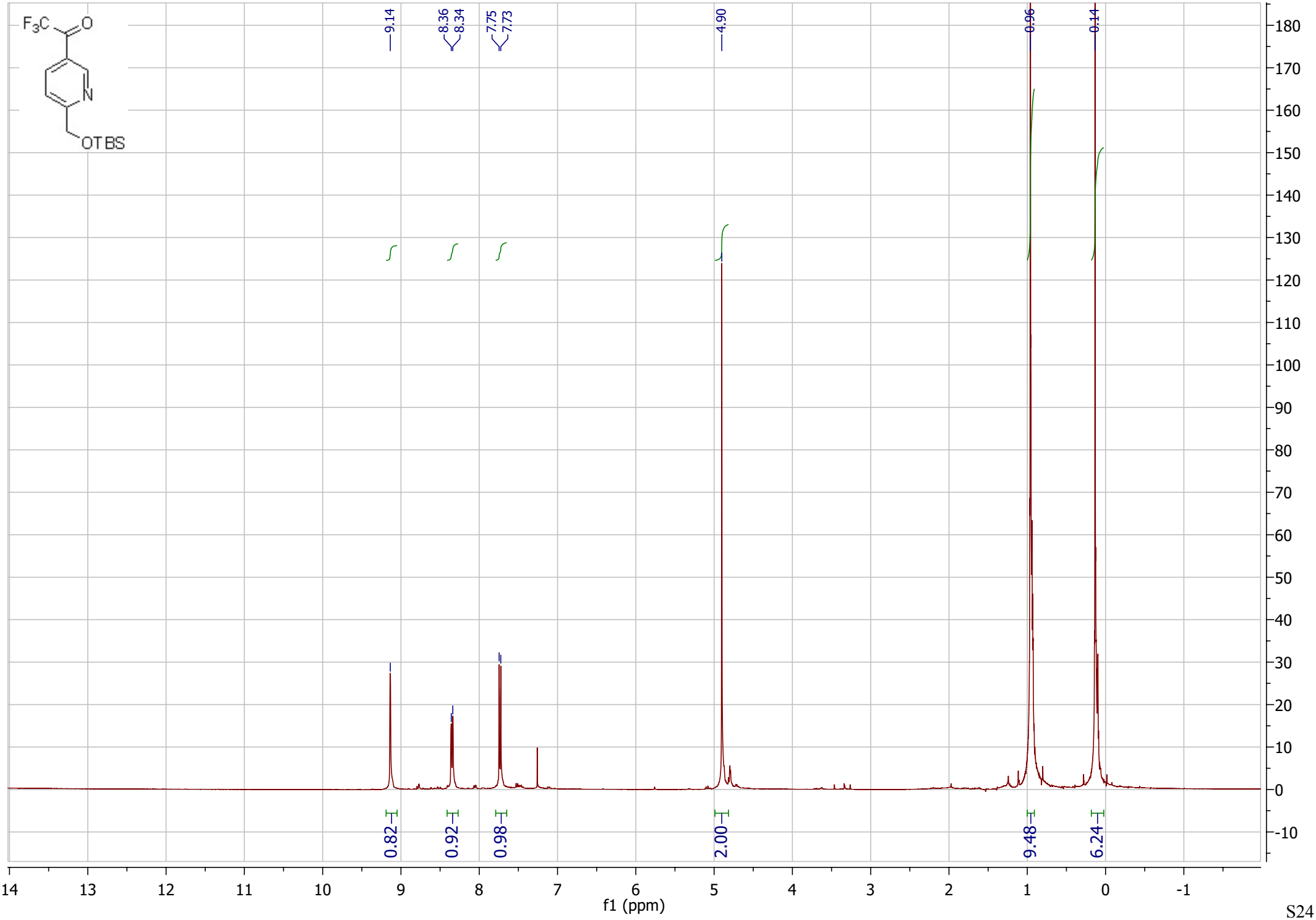
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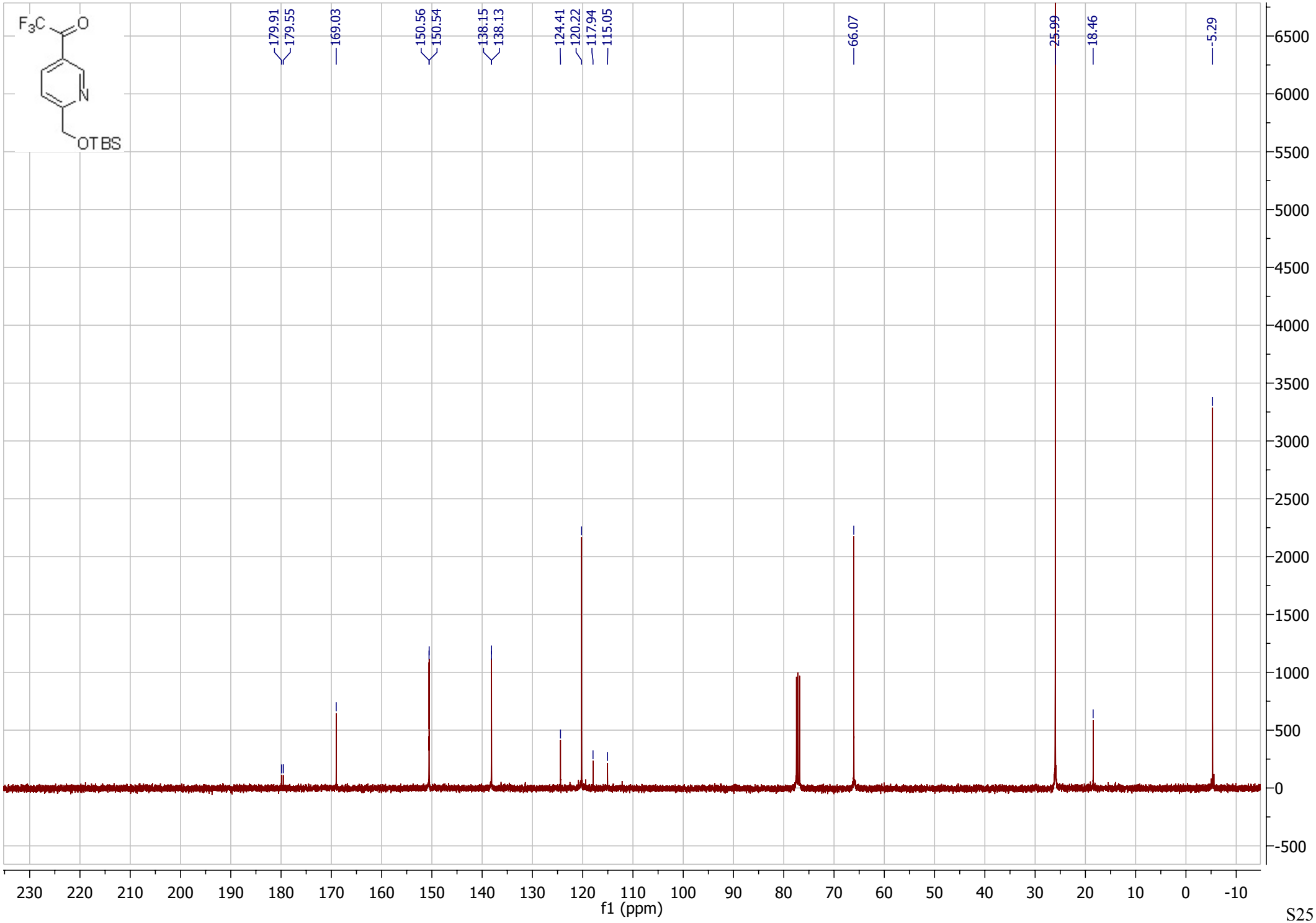
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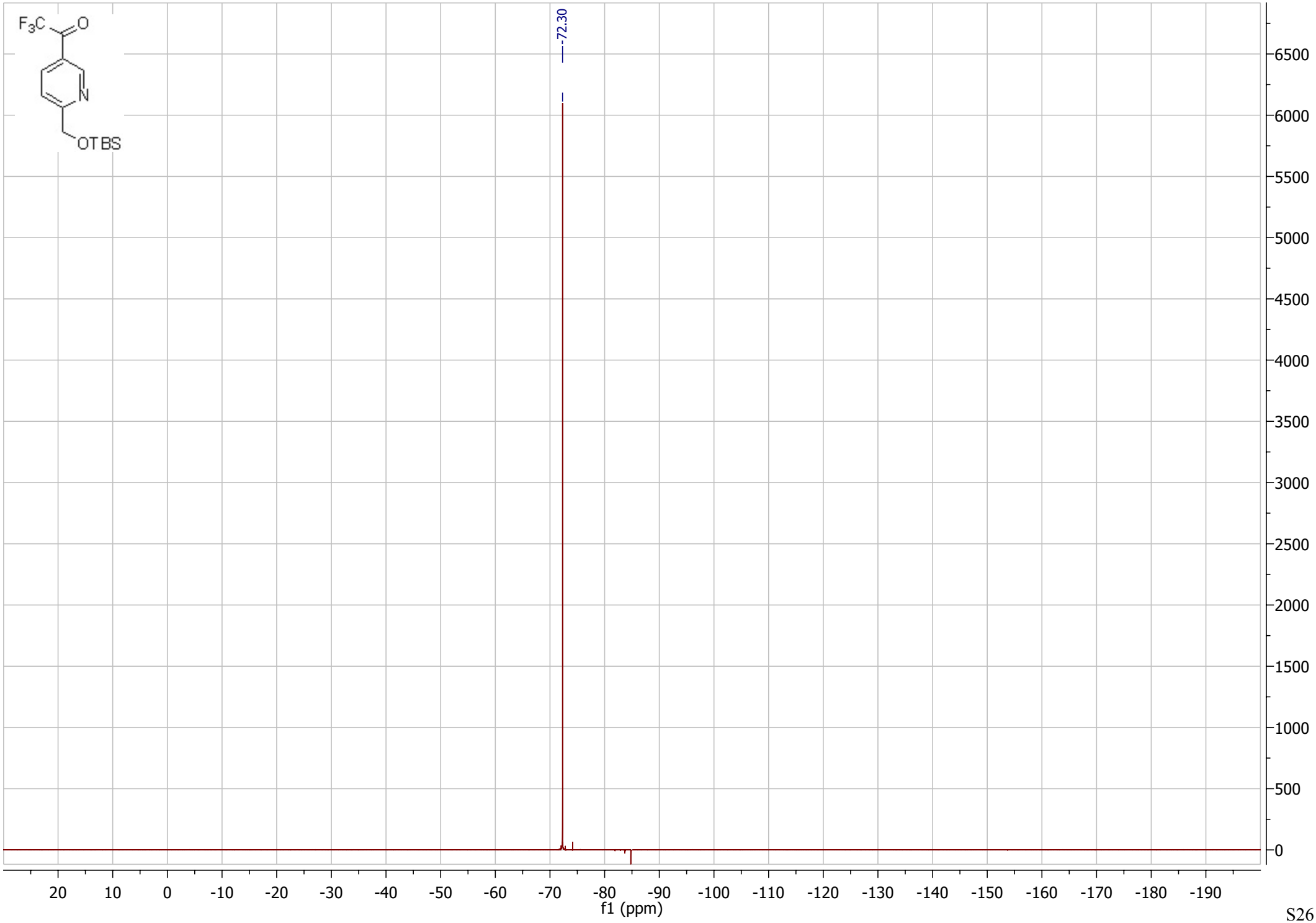
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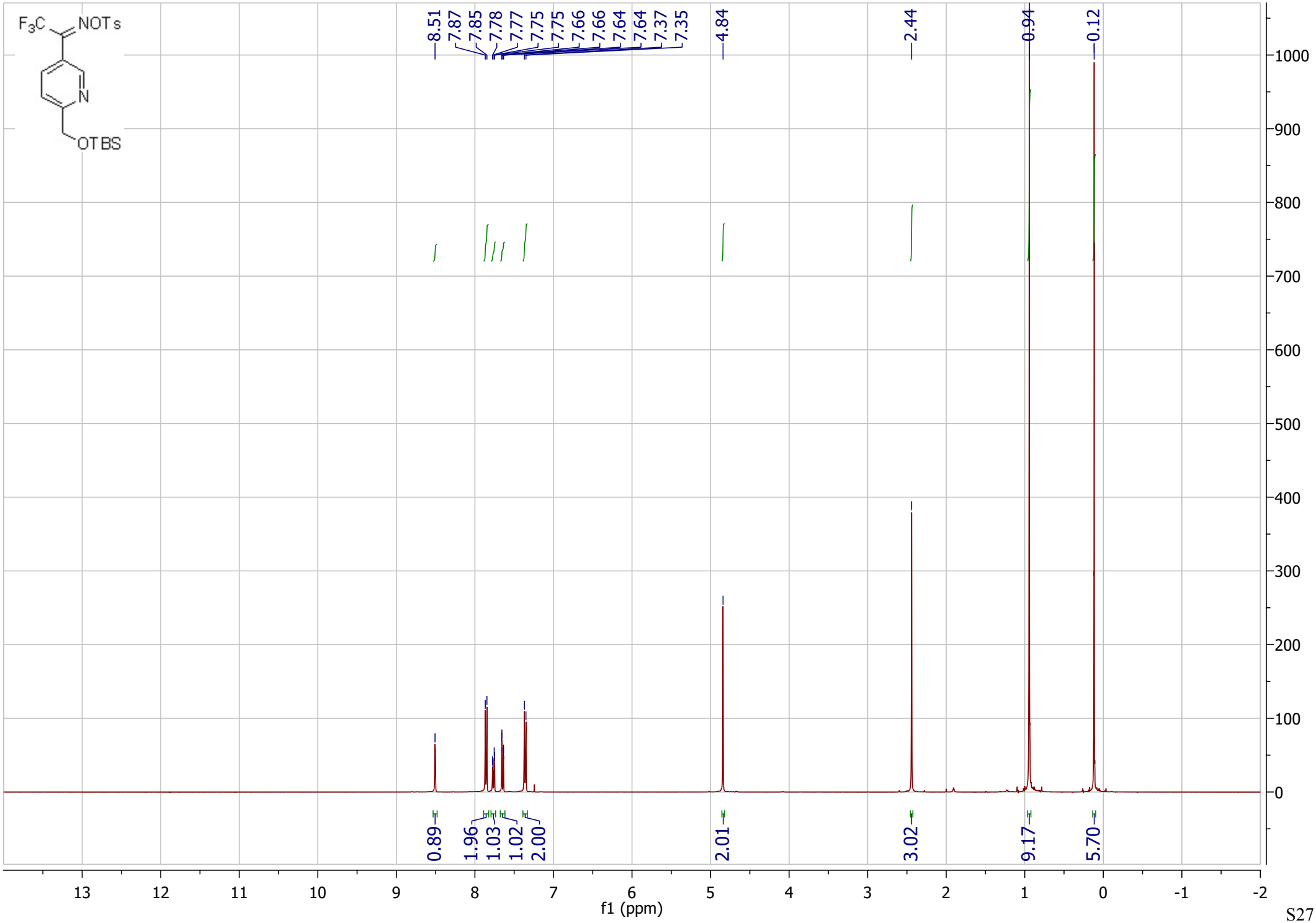
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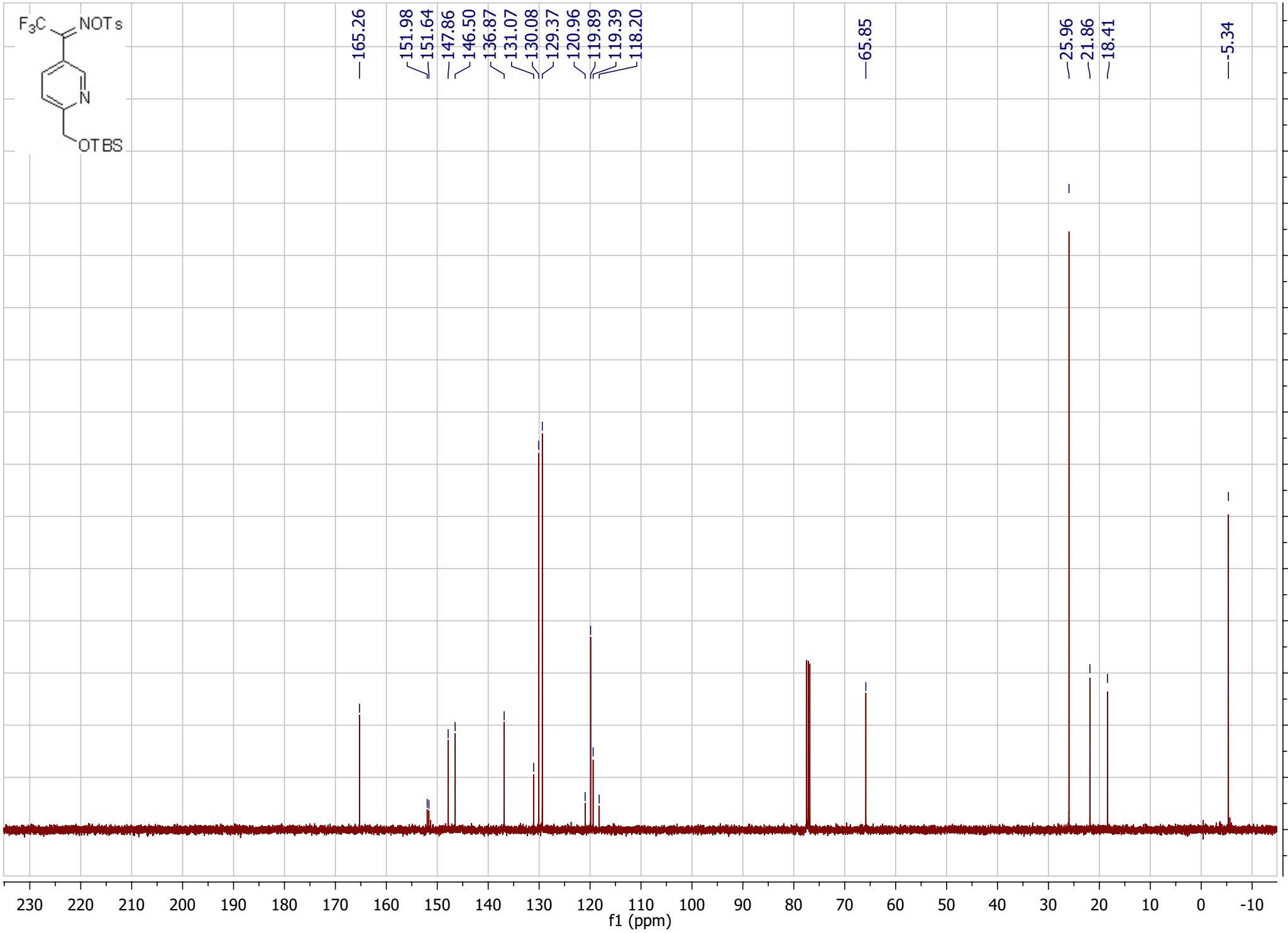
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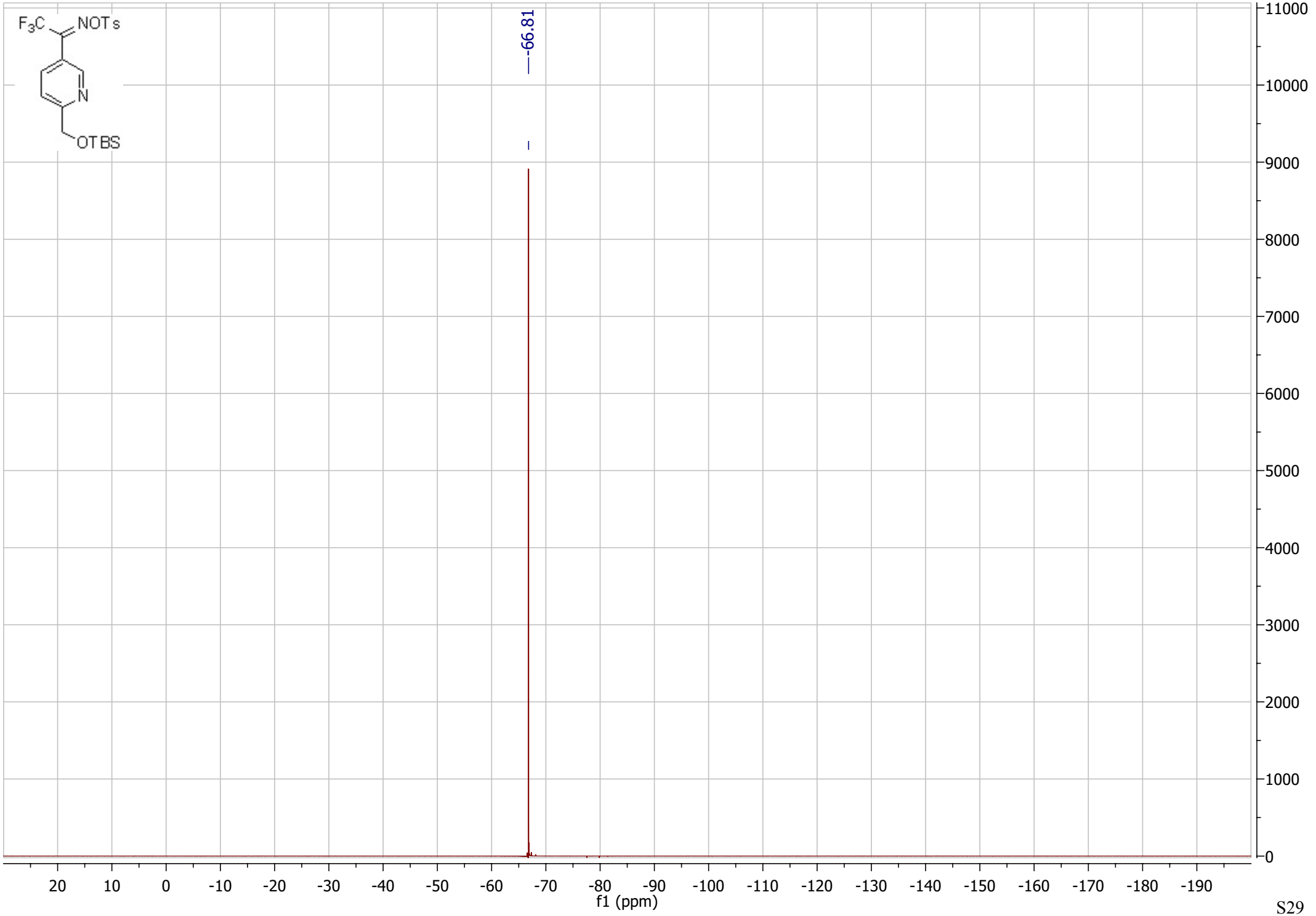
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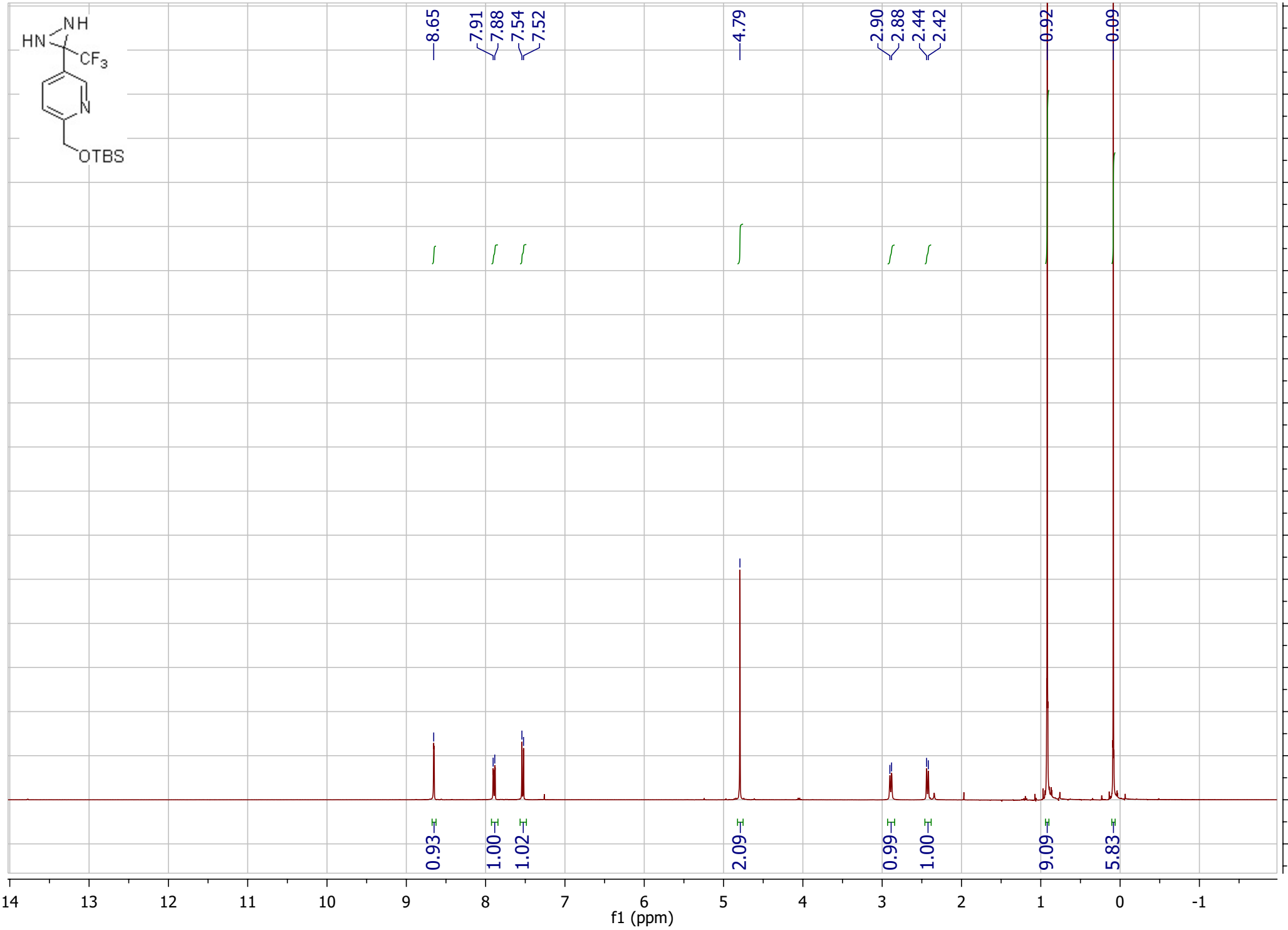
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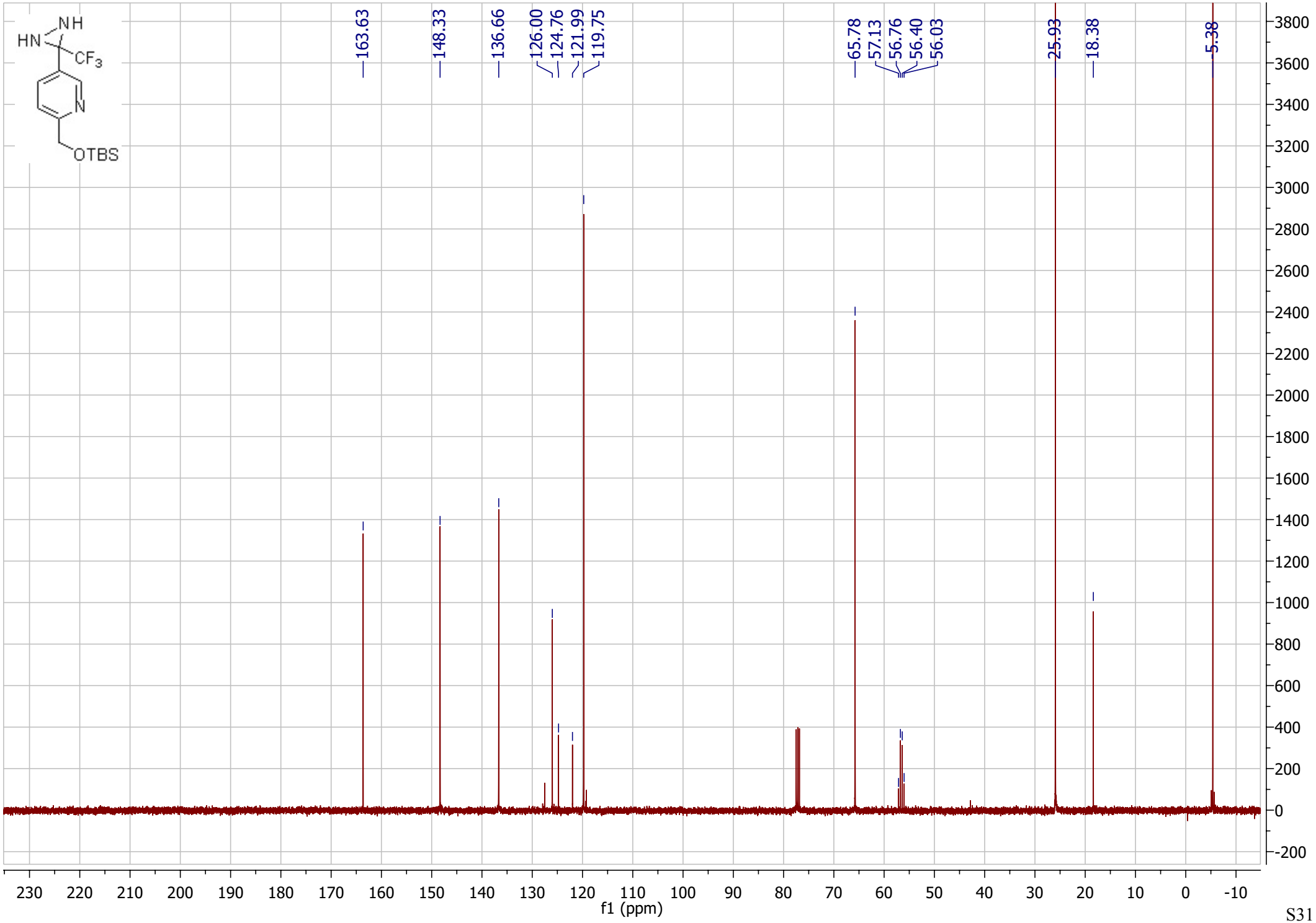
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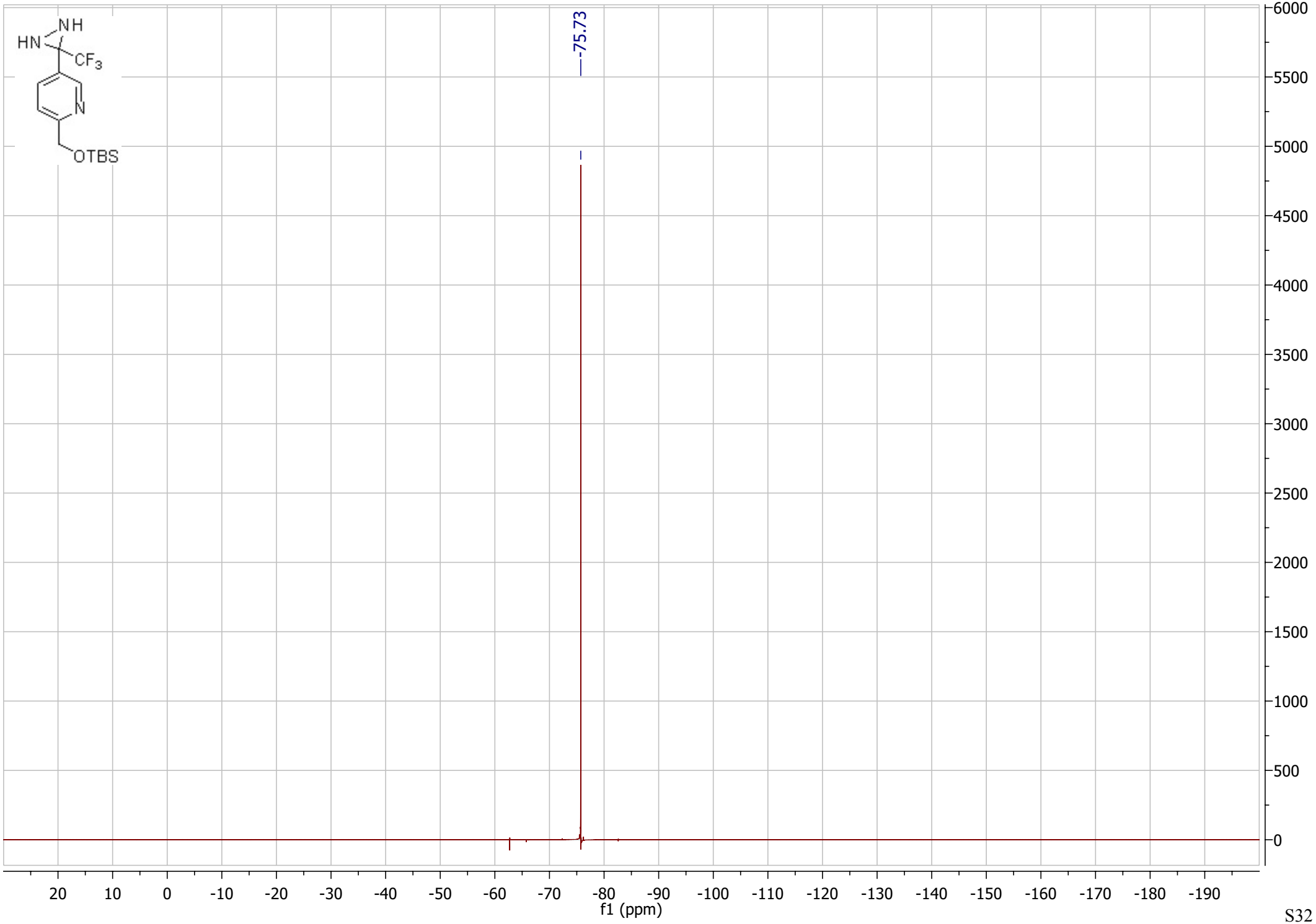
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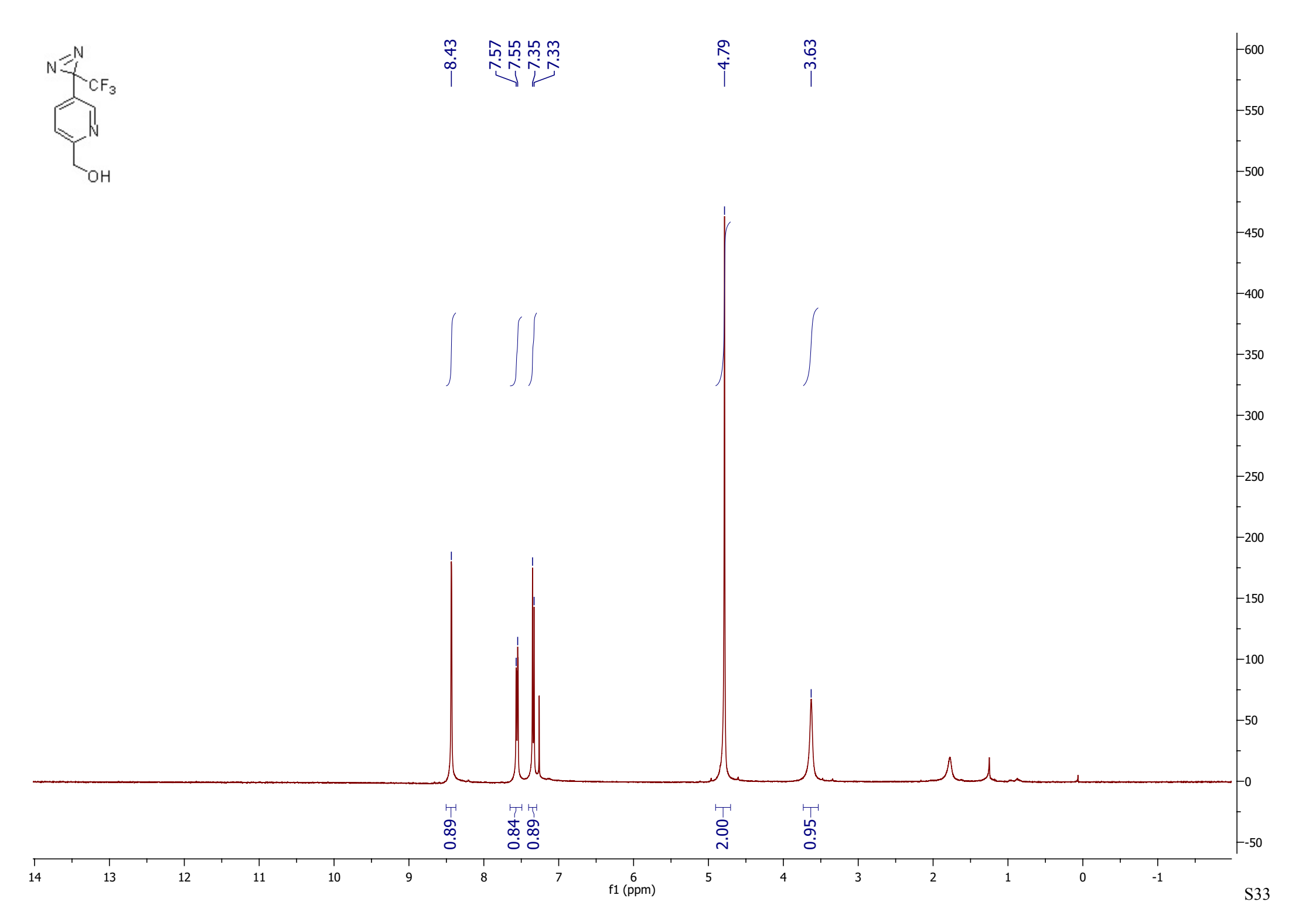
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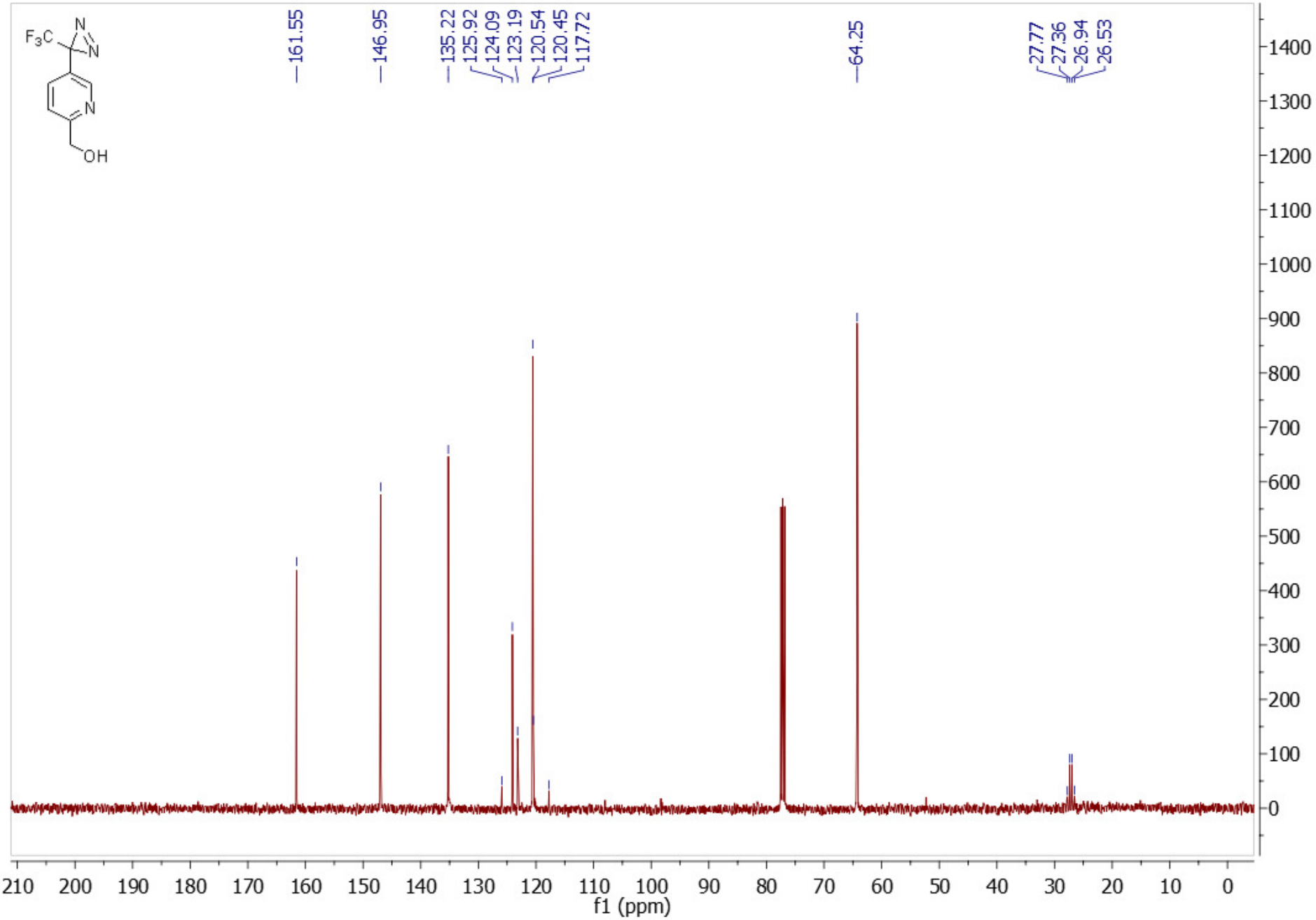
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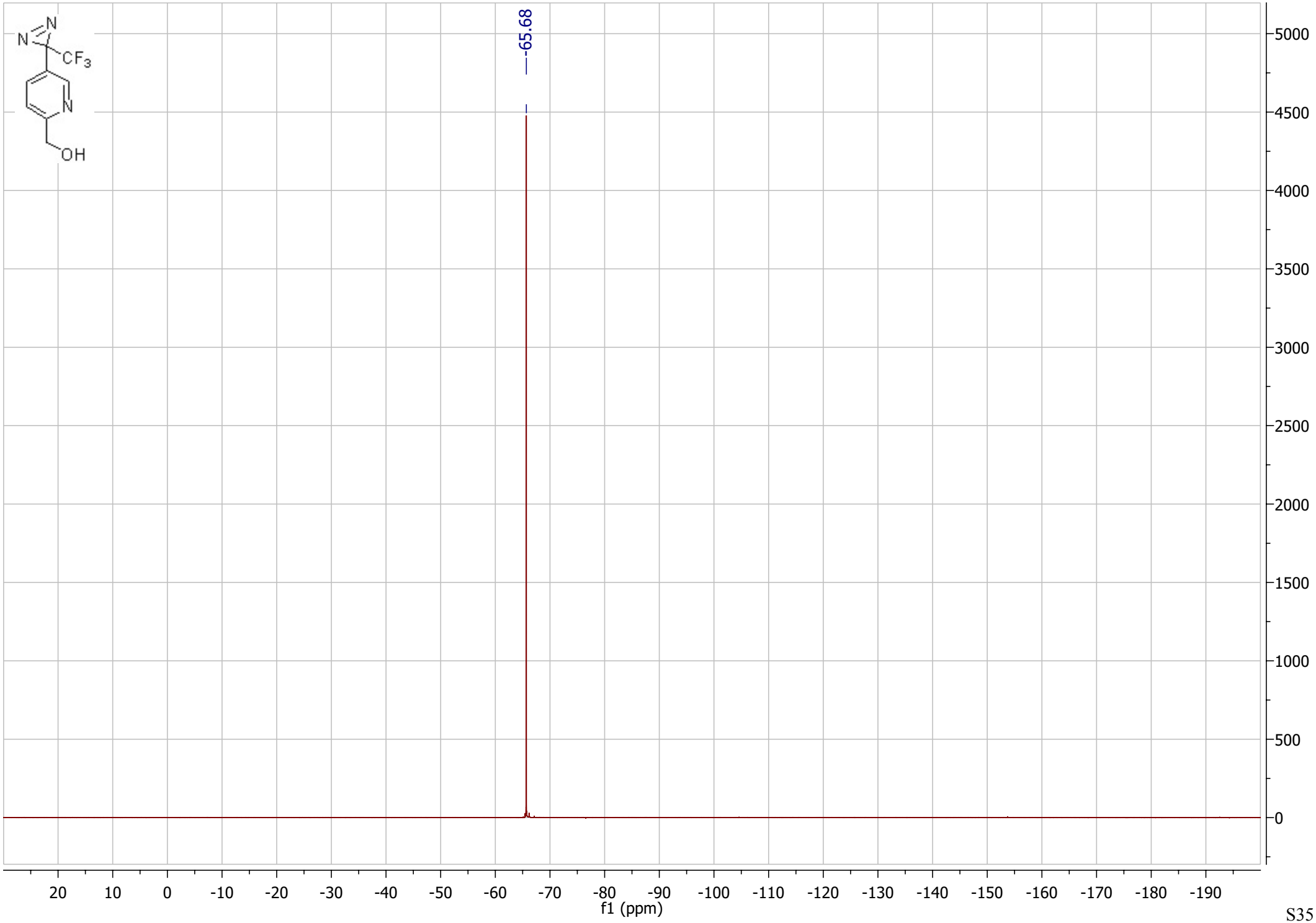
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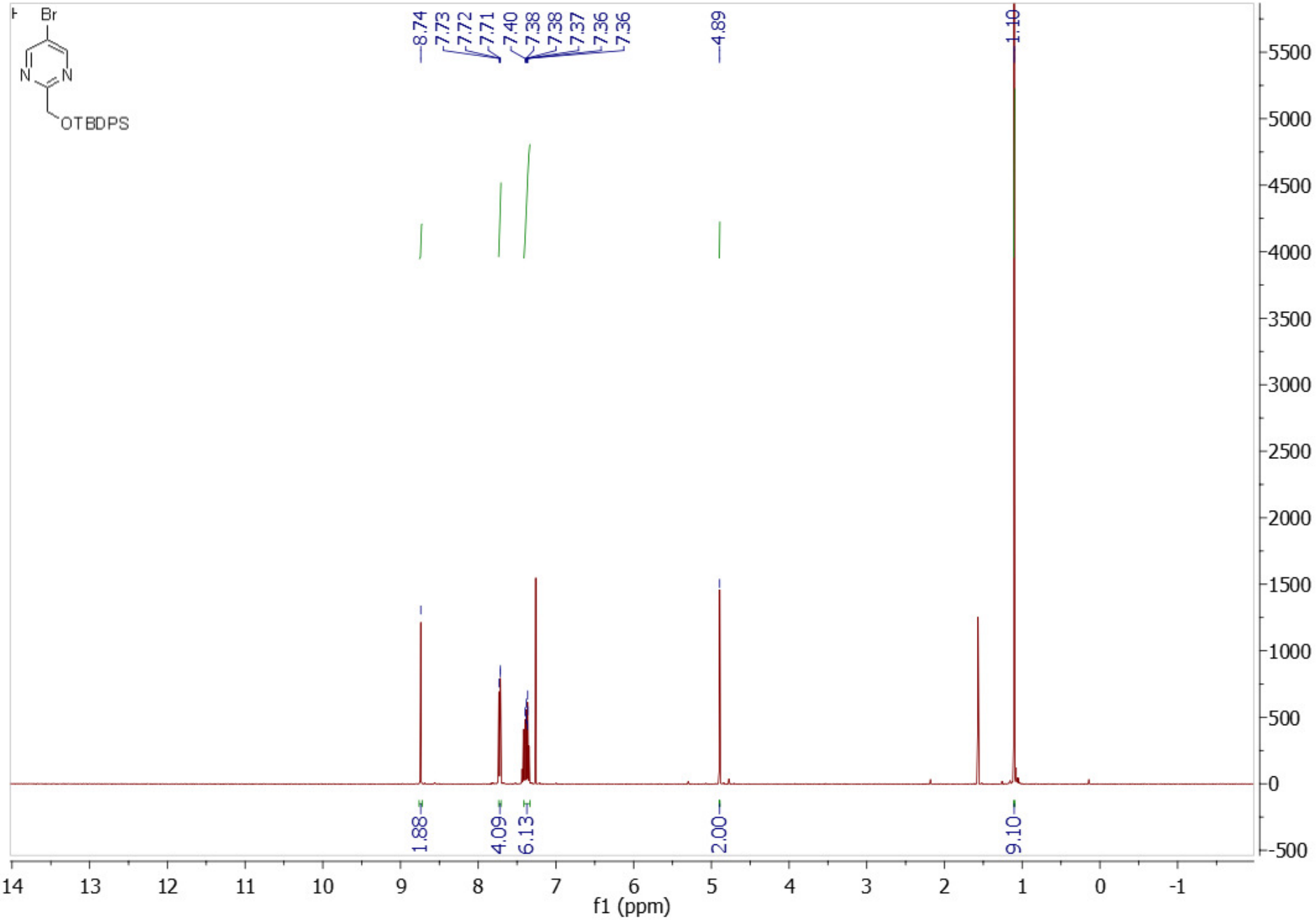
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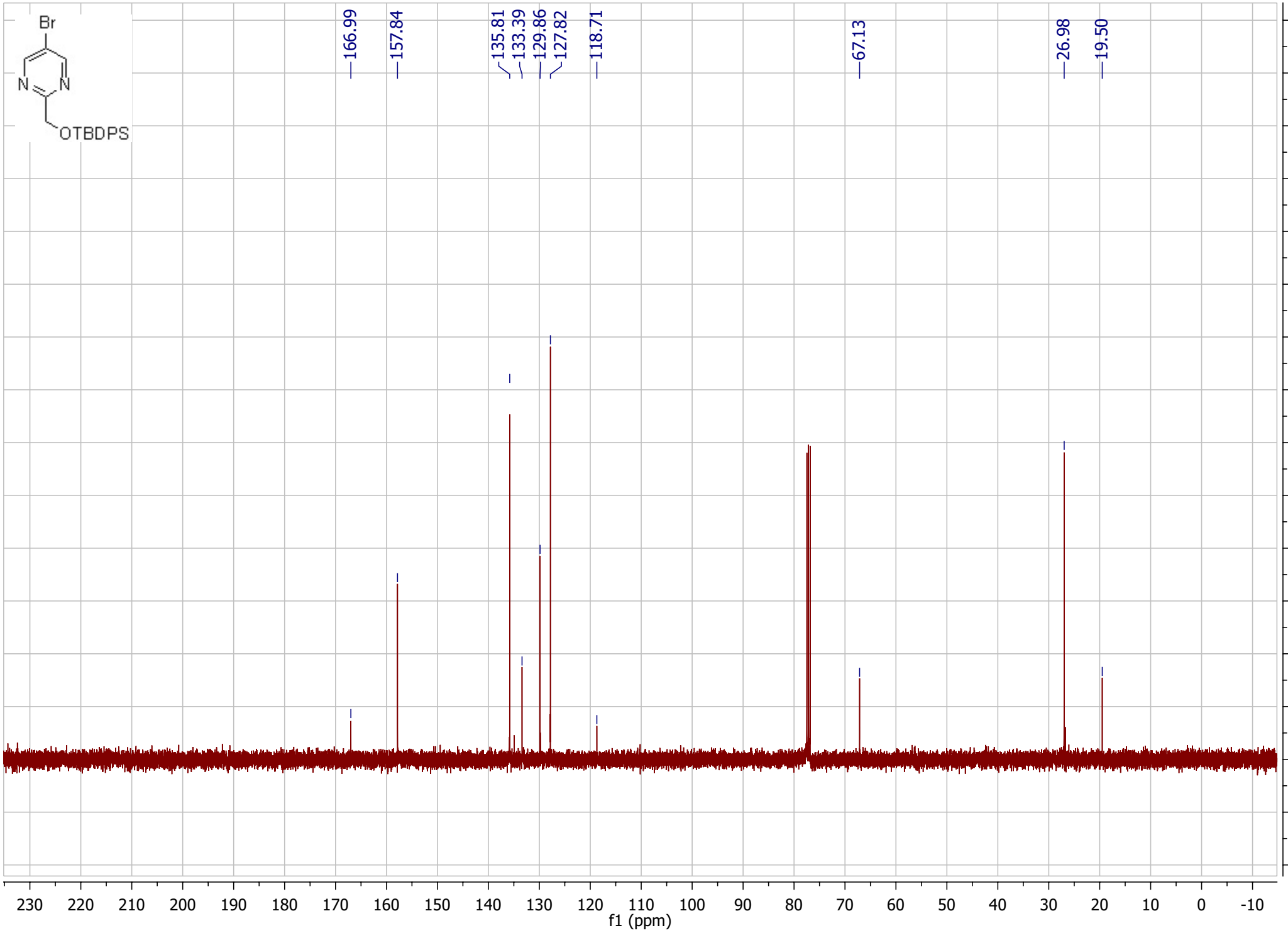
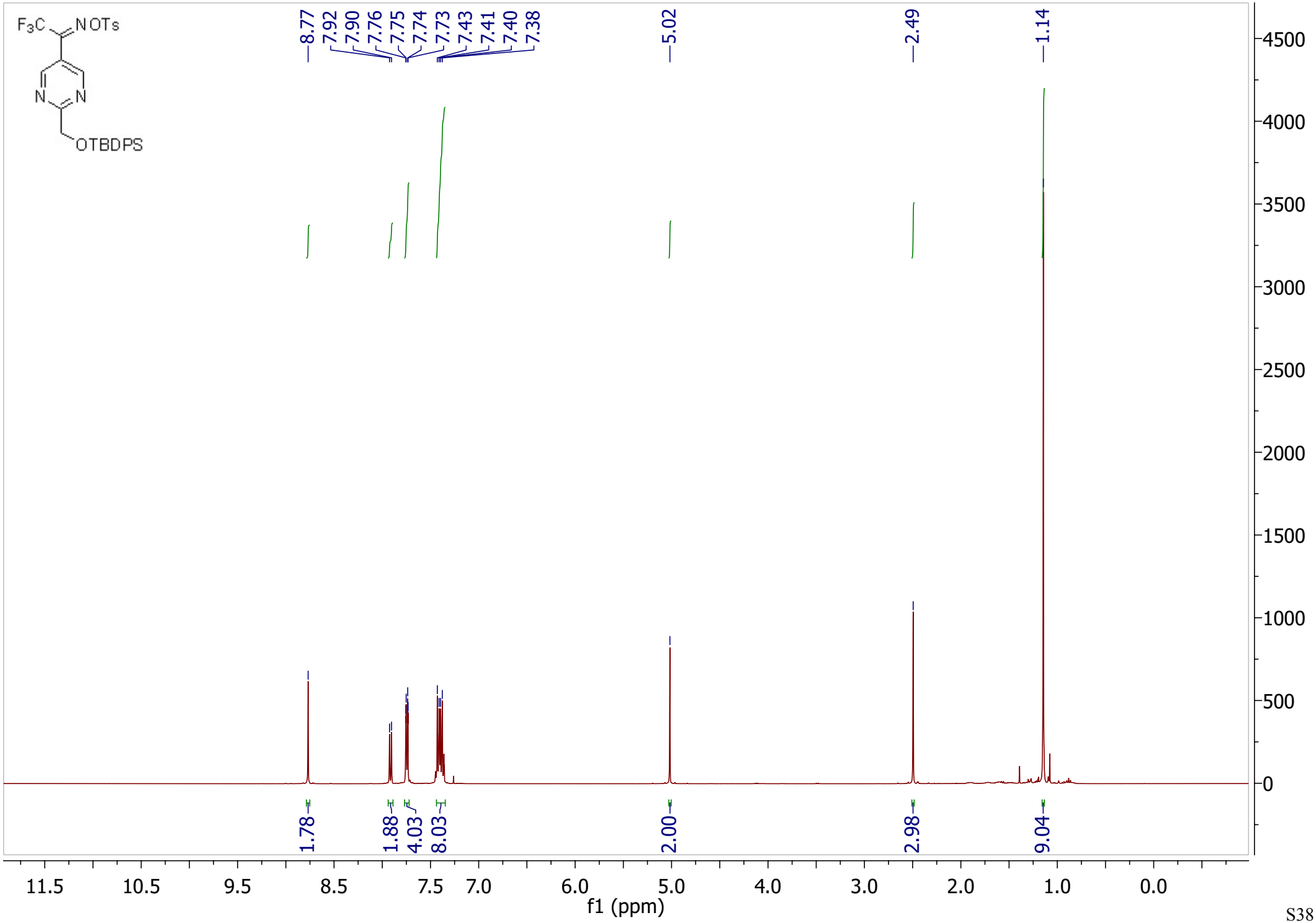
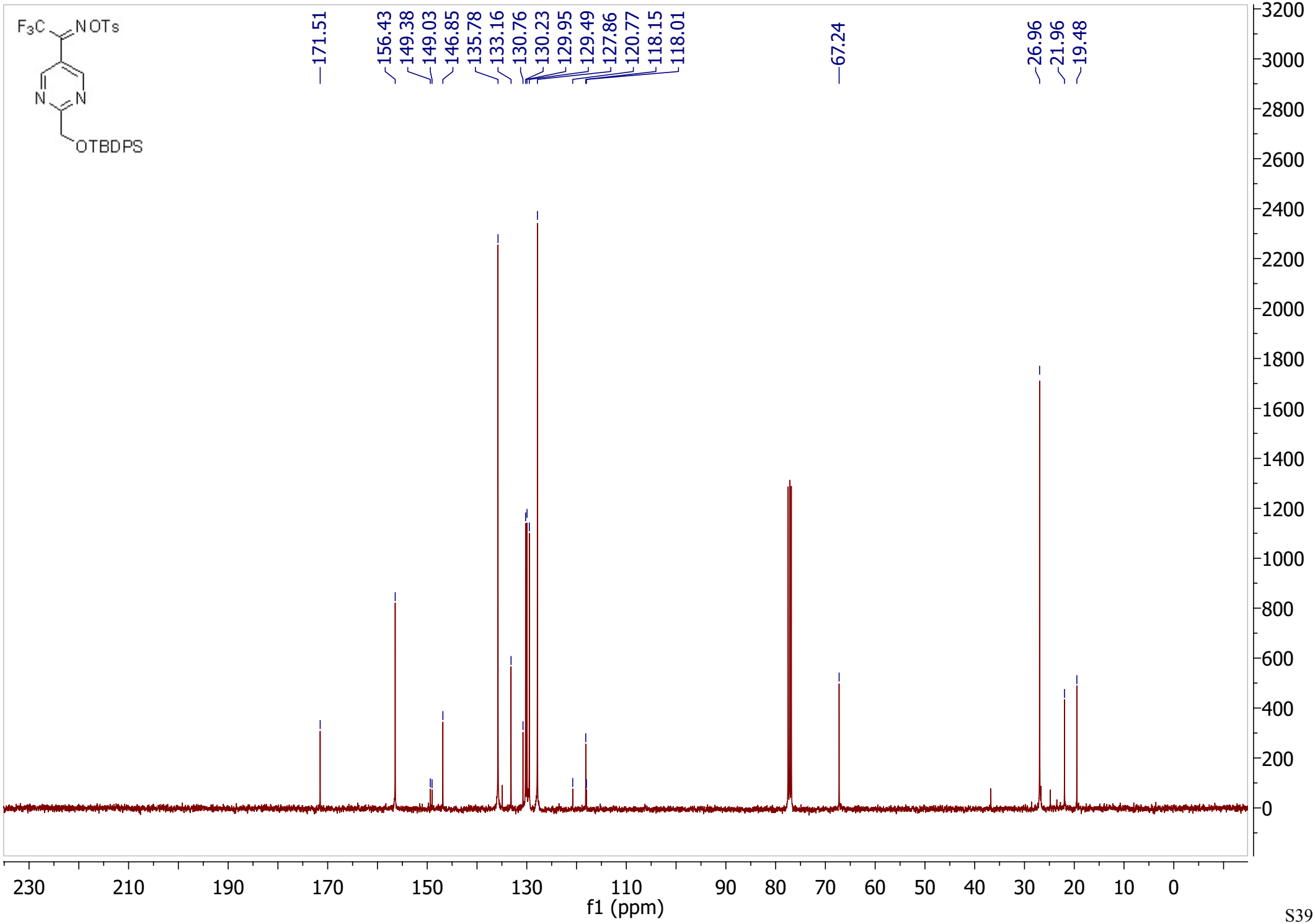
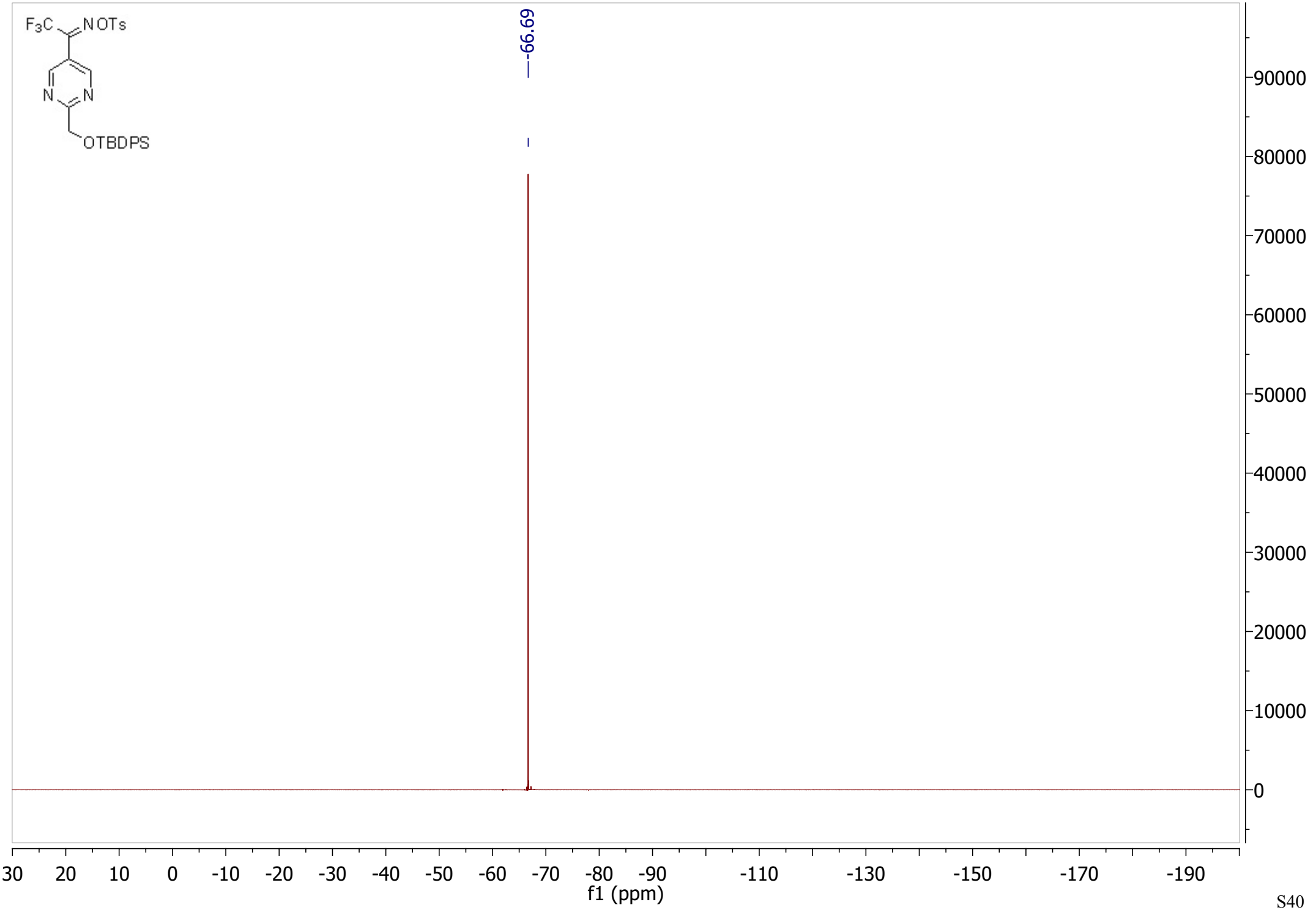
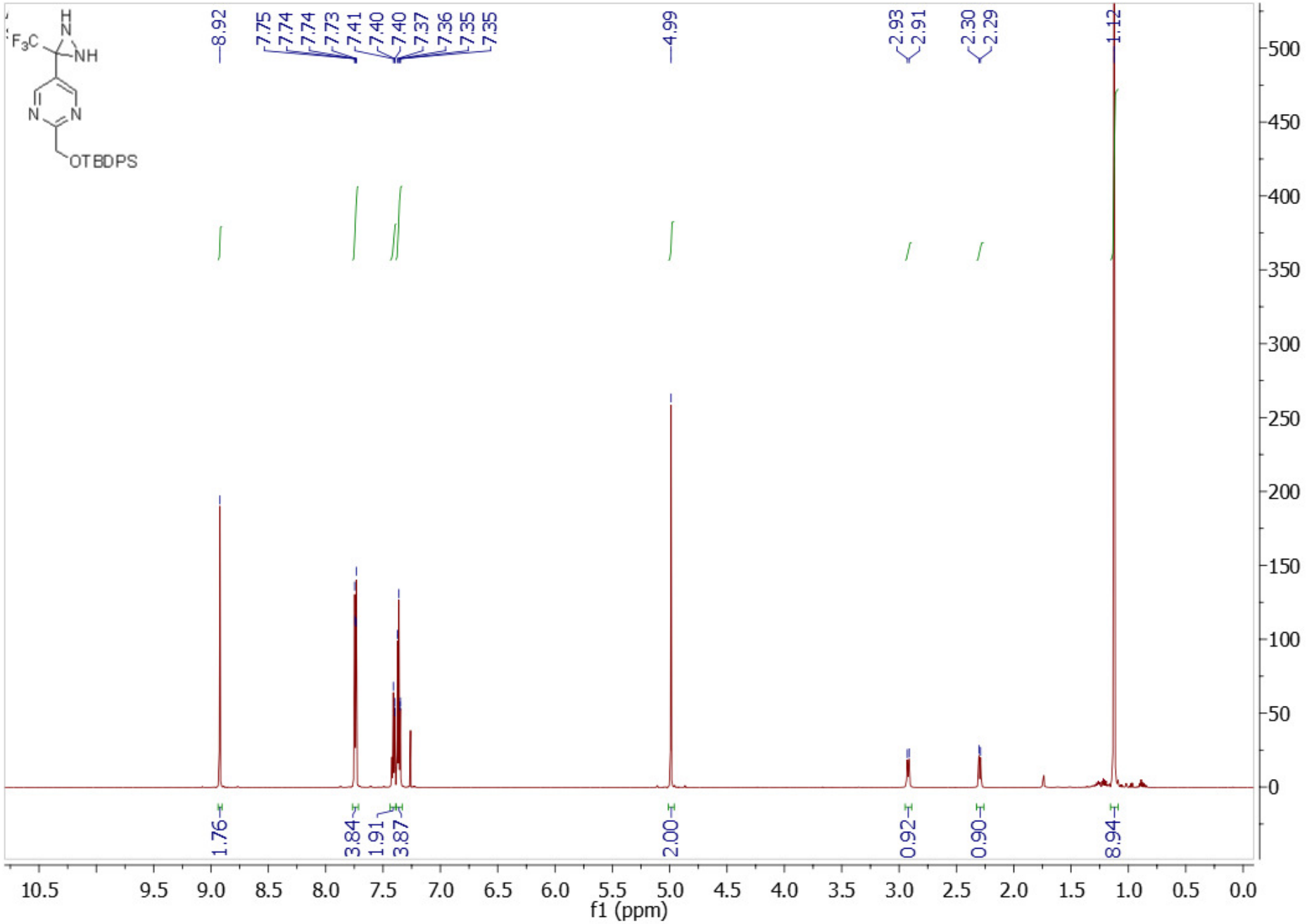
N
N
OTBDPS
8.74
7.73
7.72
7.71
7.40
7.38
7.38
7.37
7.36
7.36
4.89
1.10
1.88
4.09
6.13
2.00
9.10
f1 (ppm)

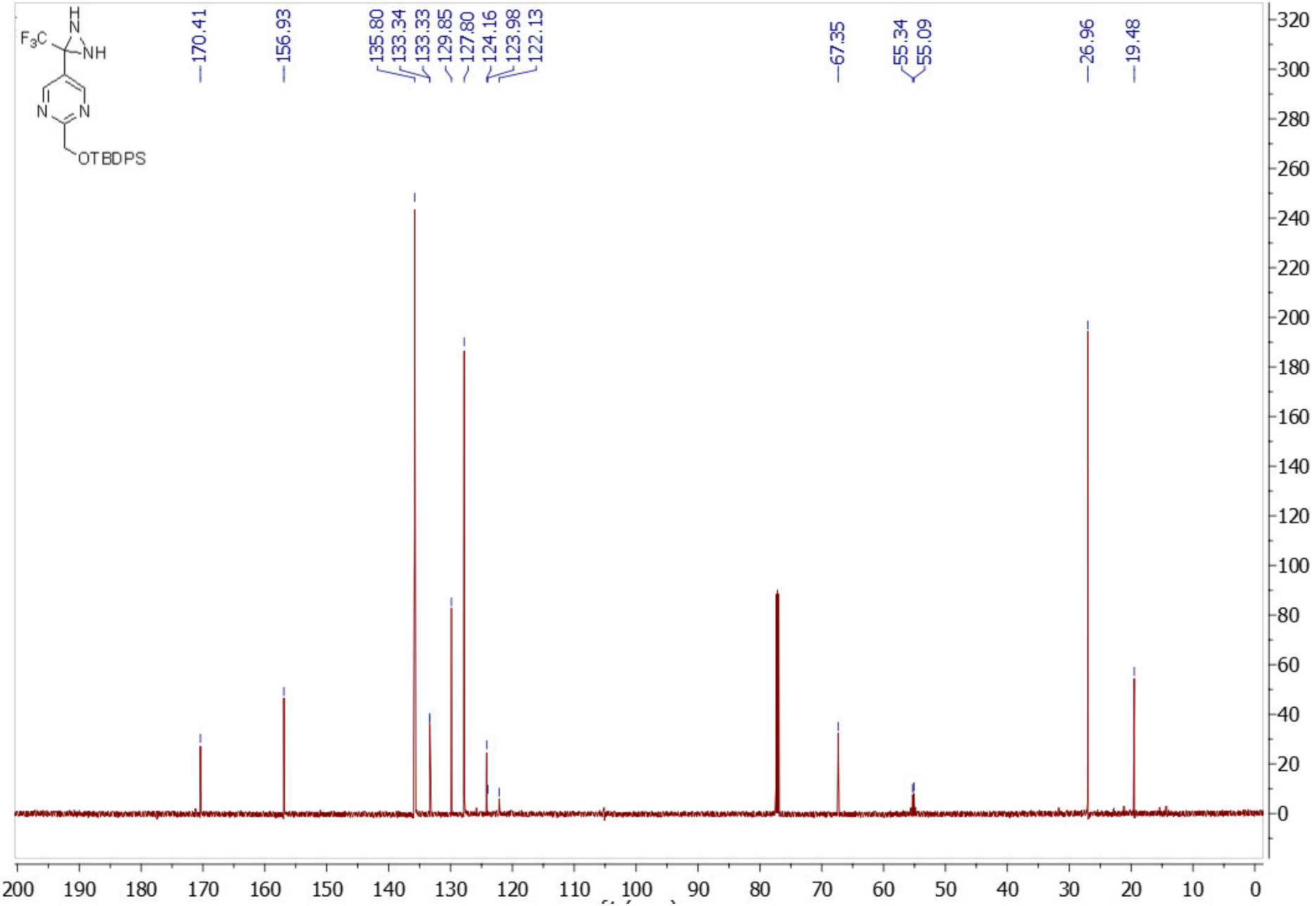

Br
N
N
OTBDPS
166.99
157.84
135.81
133.39
129.86
127.82
118.71
67.13
26.98
19.50
f1 (ppm)

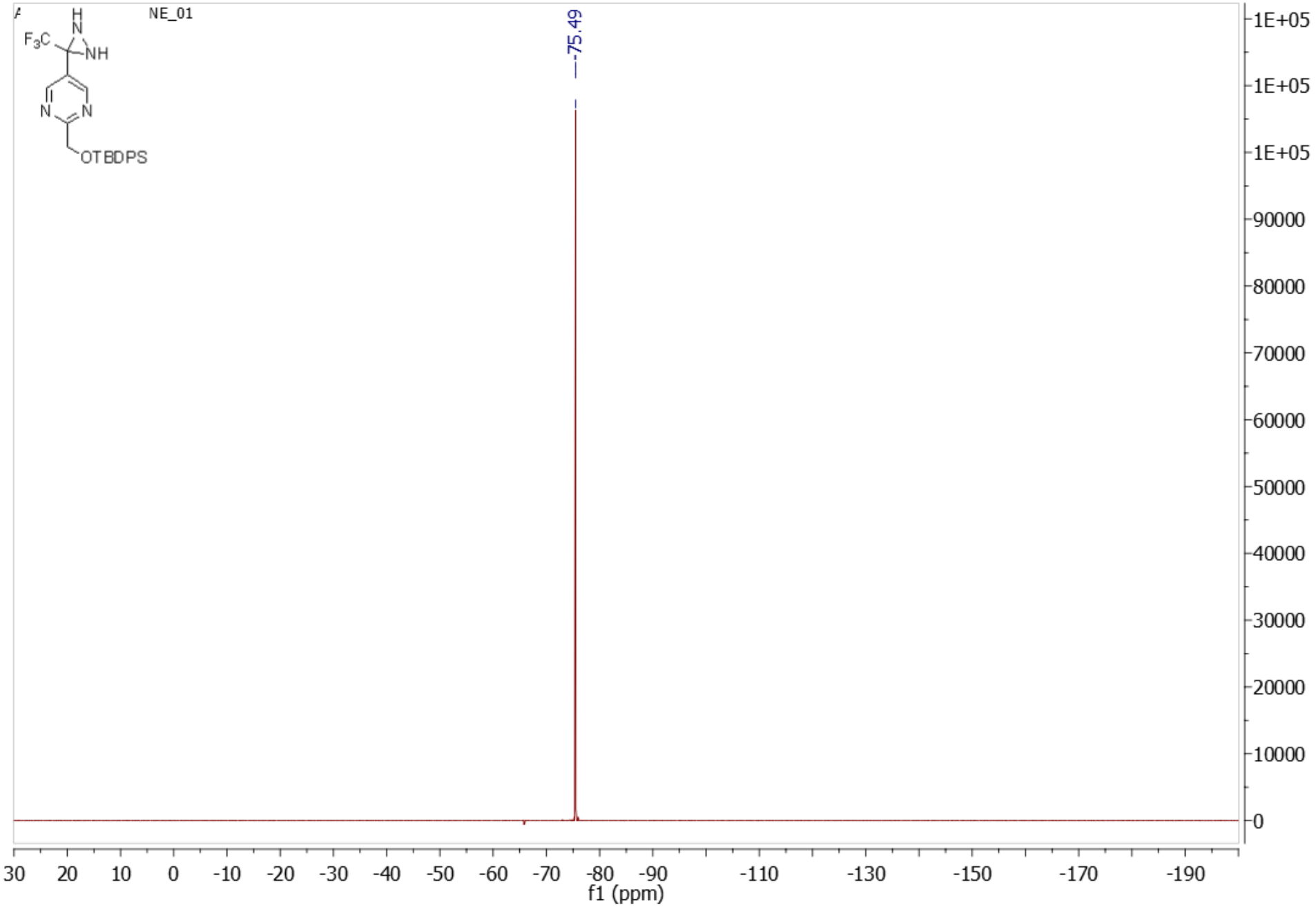

F3C
NOTs
N
N
OTBDPS
8.77
7.92
7.90
7.76
7.75
7.74
7.73
7.43
7.41
7.40
7.38
5.02
2.49
1.14
1.78
1.88
4.03
8.03
2.00
2.98
9.04
4500
4000
3500
3000
2500
2000
1500
1000
500
0
11.5
10.5
9.5
8.5
7.5
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
f1 (ppm)

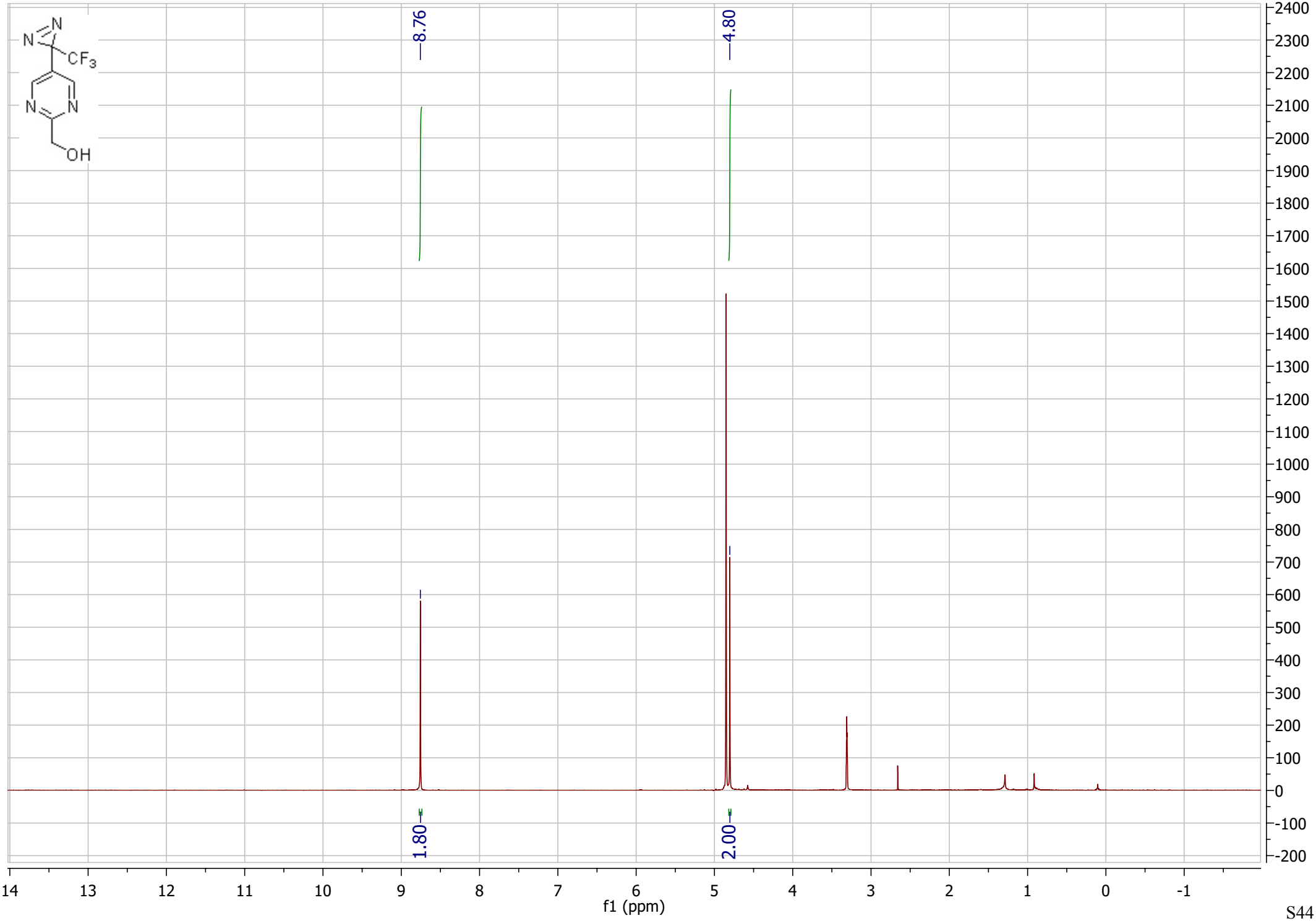

F3C
NOTs
N
N
OTBDPS
171.51
156.43
149.38
149.03
146.85
135.78
133.16
130.76
130.23
129.95
129.49
127.86
120.77
118.15
118.01
67.24
26.96
21.96
19.48
3200
3000
2800
2600
2400
2200
2000
1800
1600
1400
1200
1000
800
600
400
200
0
230
210
190
170
150
130
110
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

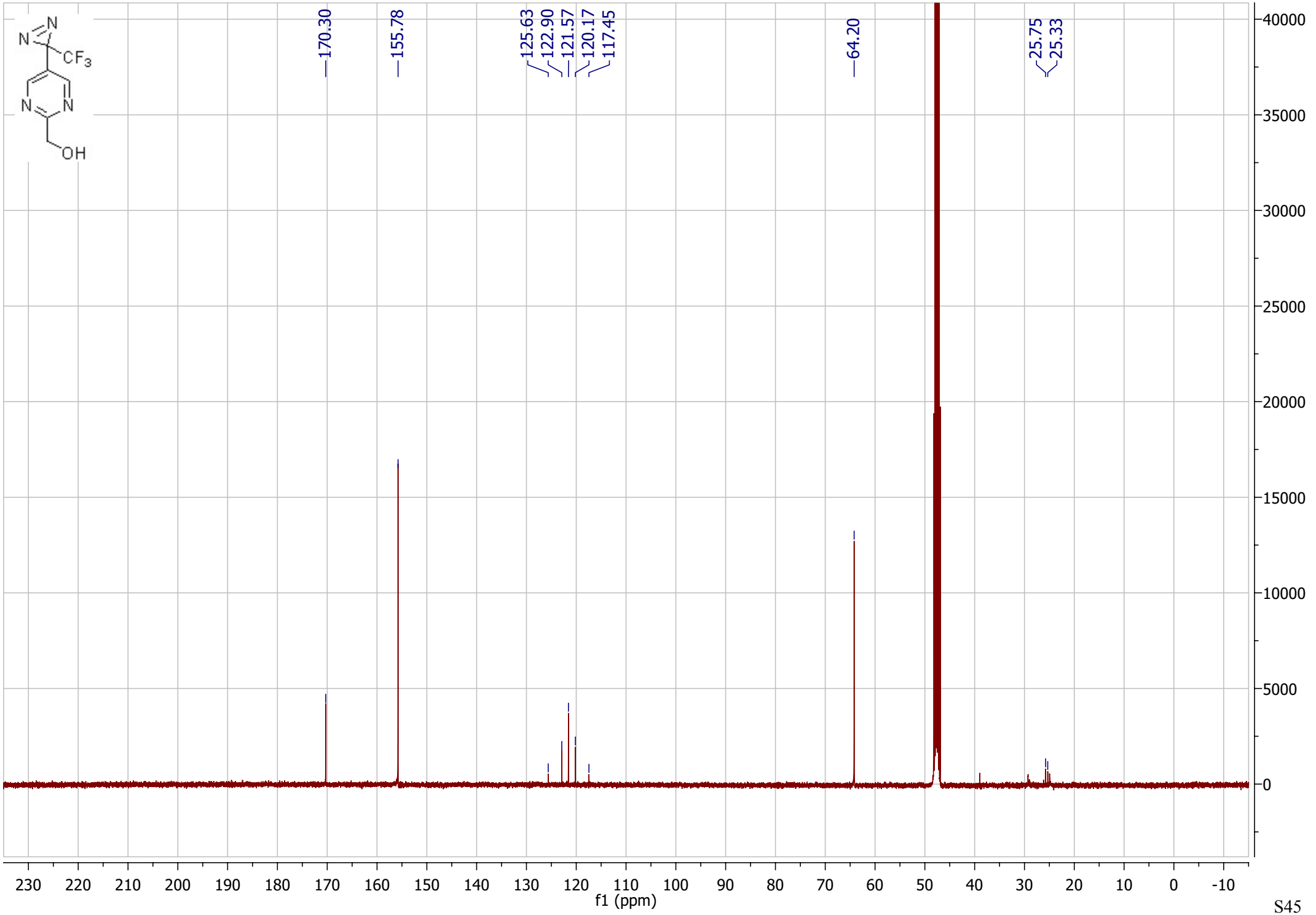

F3C
NOTs
N
N
OTBDPS
-66.69
90000
80000
70000
60000
50000
40000
30000
20000
10000
0
30
20
10
0
-10
-20
-30
-40
-50
-60
-70
-80
-90
-110
-130
-150
-170
-190
f1 (ppm)

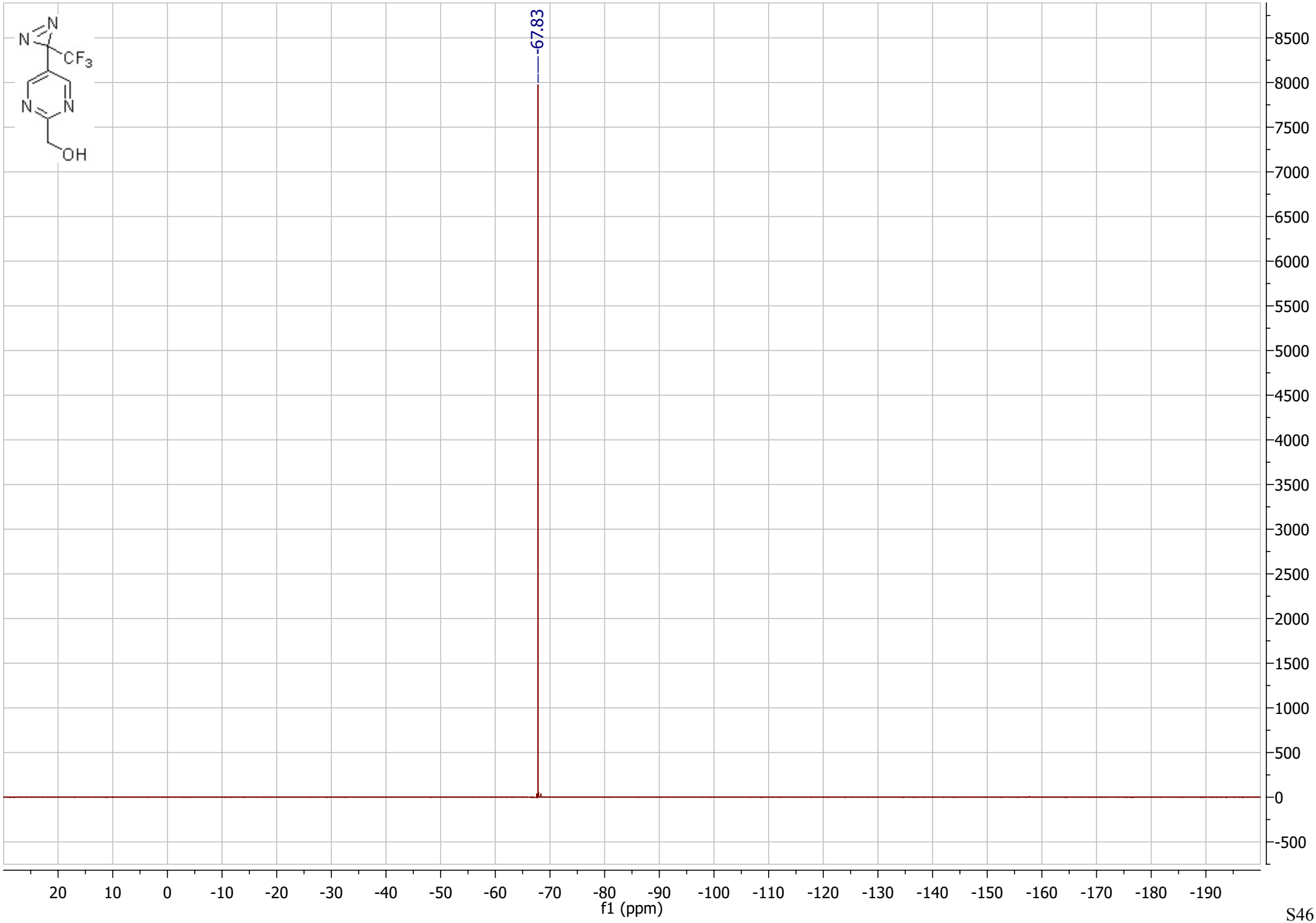

F3C
NH
N
N
OTBDPS
8.92
7.75
7.74
7.74
7.73
7.41
7.40
7.40
7.37
7.36
7.35
7.35
4.99
2.93
2.91
2.30
2.29
1.12
1.76
3.84
1.91
3.87
2.00
0.92
0.90
8.94
f1 (ppm)

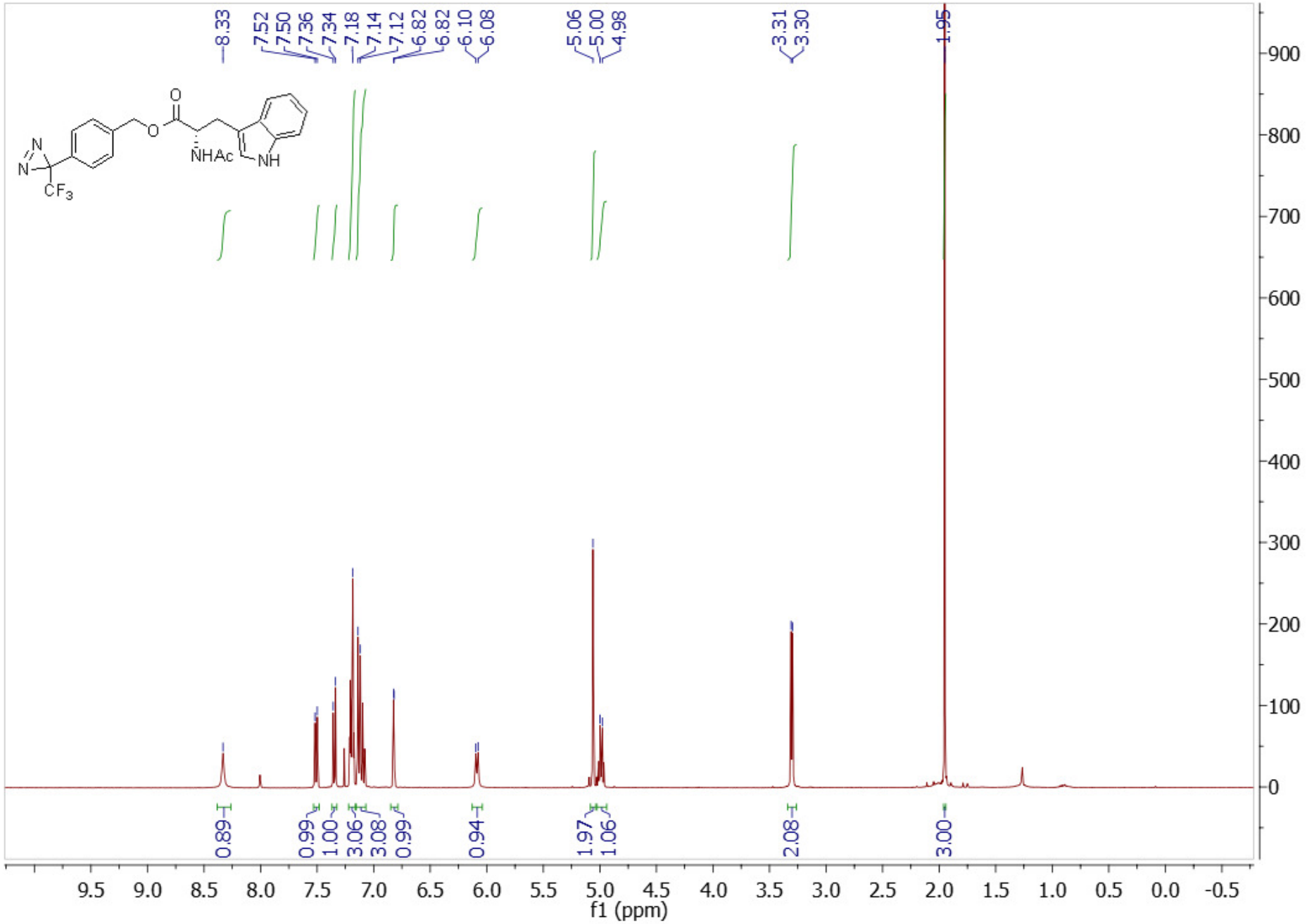

F3C
NH
N
N
OTBDPS
170.41
156.93
135.80
133.34
133.33
129.85
127.80
124.16
123.98
122.13
67.35
55.34
55.09
26.96
19.48
320
300
280
260
240
220
200
180
160
140
120
100
80
60
40
20
0
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0

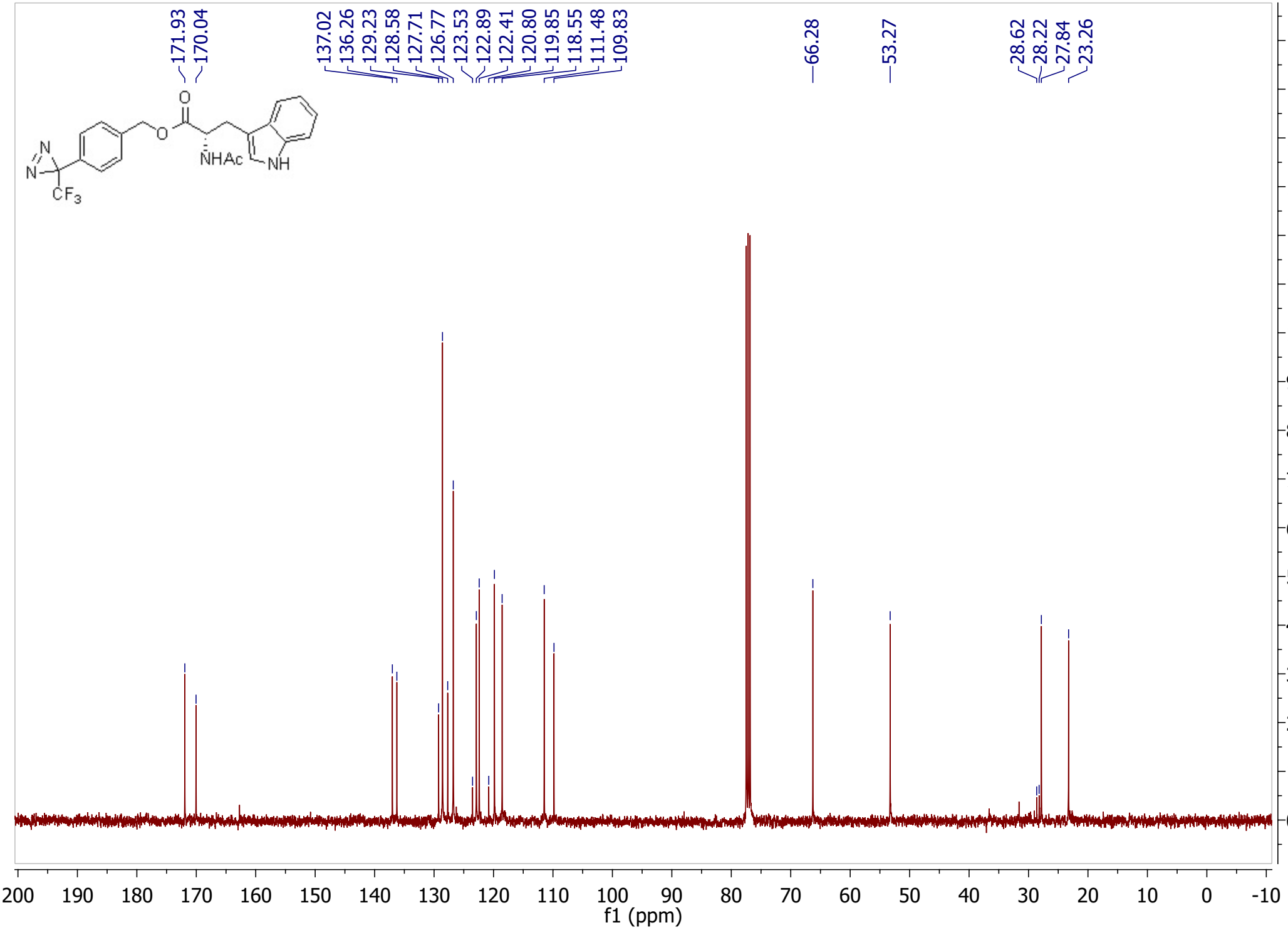

NE_01
-75.49
1E+05
1E+05
1E+05
90000
80000
70000
60000
50000
40000
30000
20000
10000
0
30 20 10 0 -10 -20 -30 -40 -50 -60 -70 -80 -90 -110 -130 -150 -170 -190
f1 (ppm)

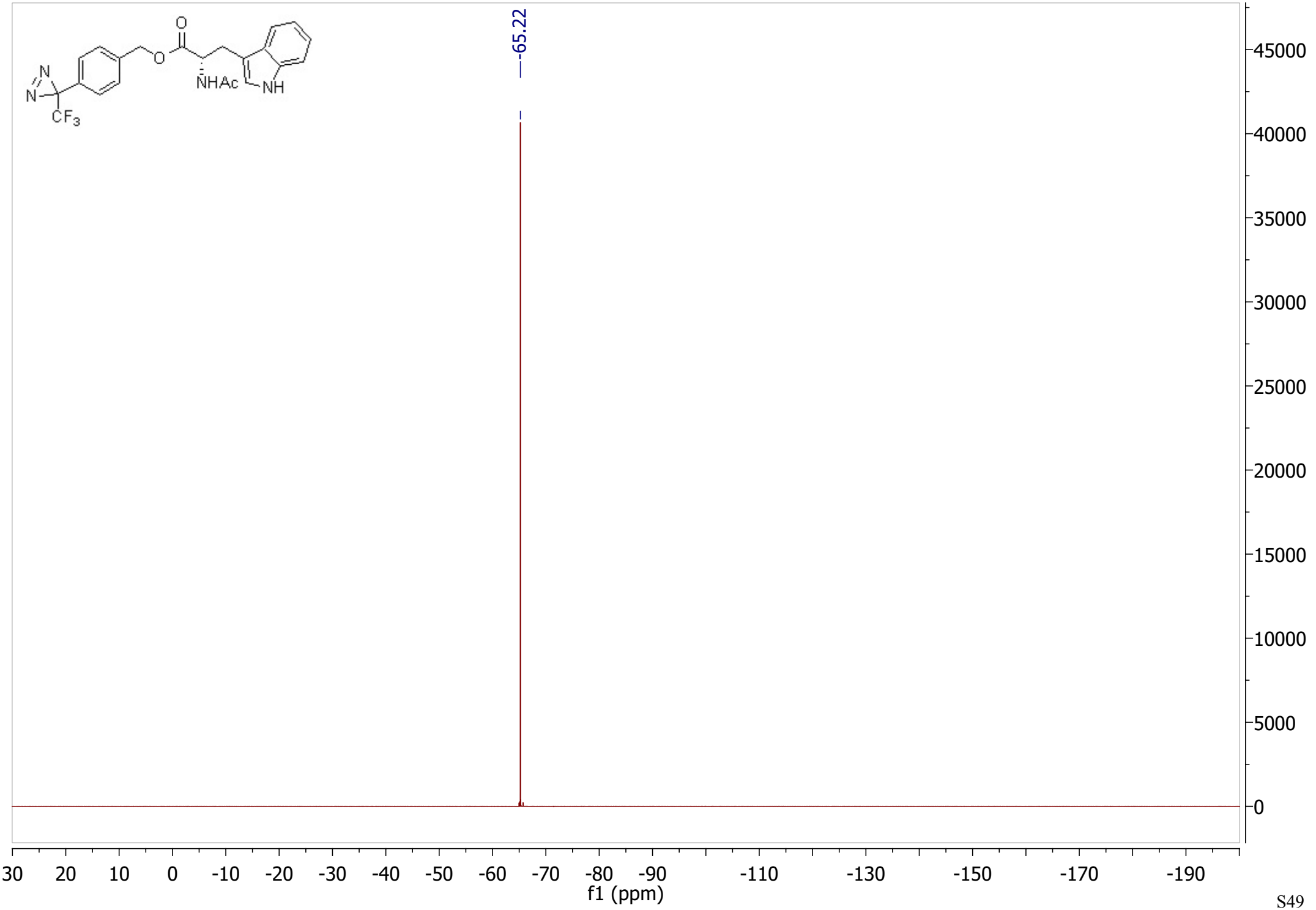

8.76
4.80
1.80
2.00
f1 (ppm)

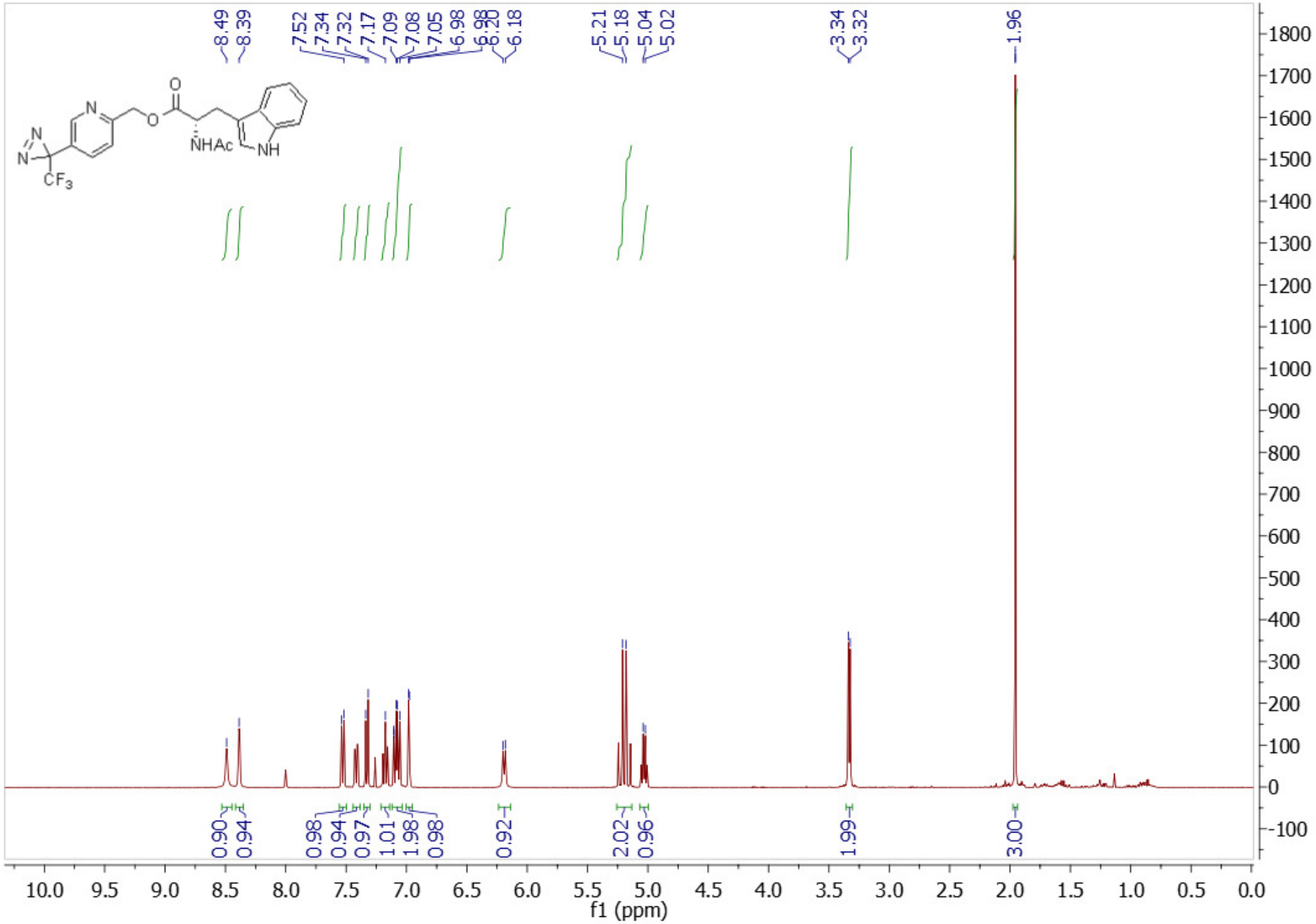

170.30
155.78
125.63
122.90
121.57
120.17
117.45
64.20
25.75
25.33
N
N
CF3
N
N
OH
40000
35000
30000
25000
20000
15000
10000
5000
0
230 220 210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10
f1 (ppm)

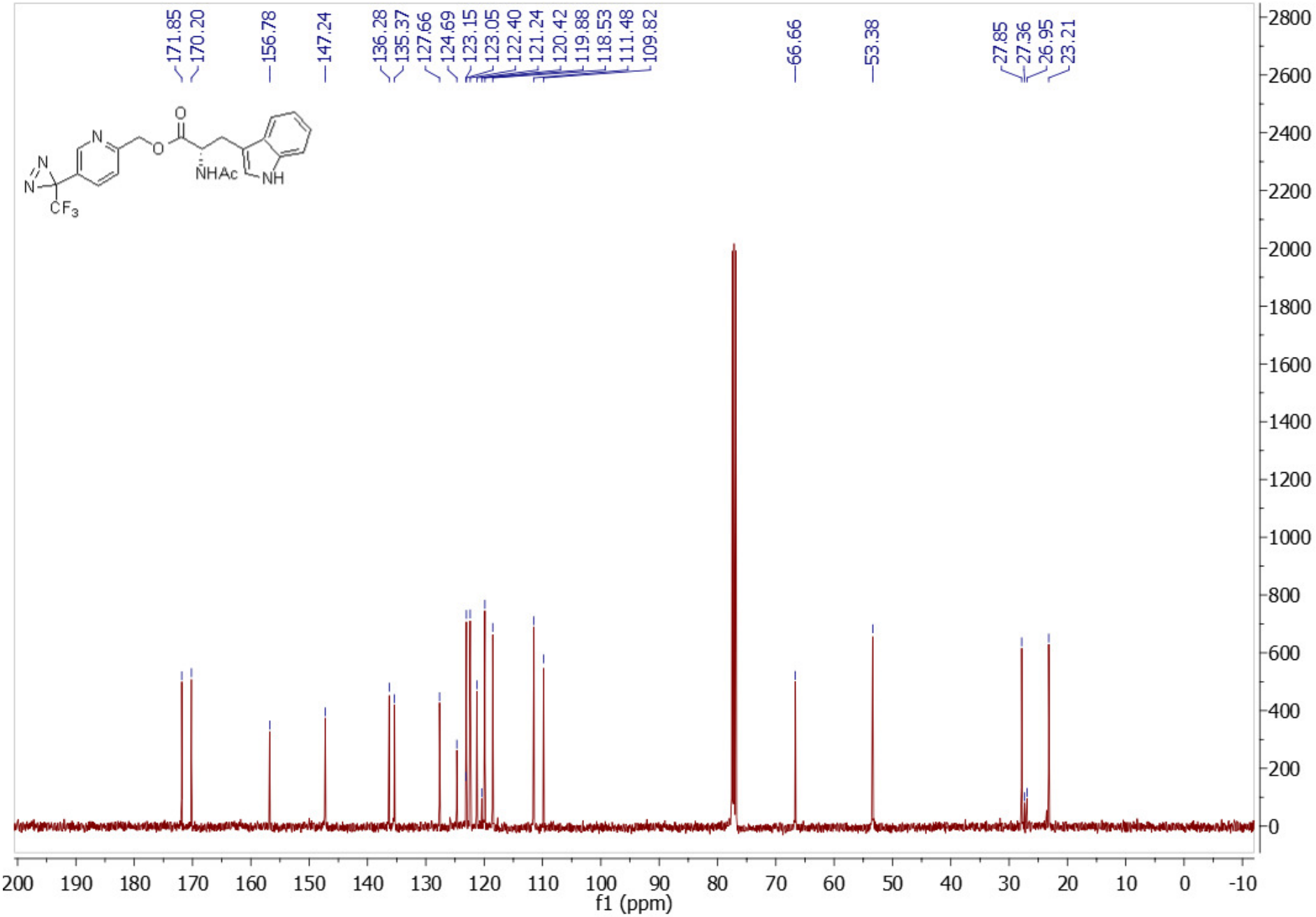

-67.83
N
N
CF3
N
N
OH
8500
8000
7500
7000
6500
6000
5500
5000
4500
4000
3500
3000
2500
2000
1500
1000
500
0
-500
20
10
0
-10
-20
-30
-40
-50
-60
-70
-80
-90
-100
-110
-120
-130
-140
-150
-160
-170
-180
-190
f1 (ppm)

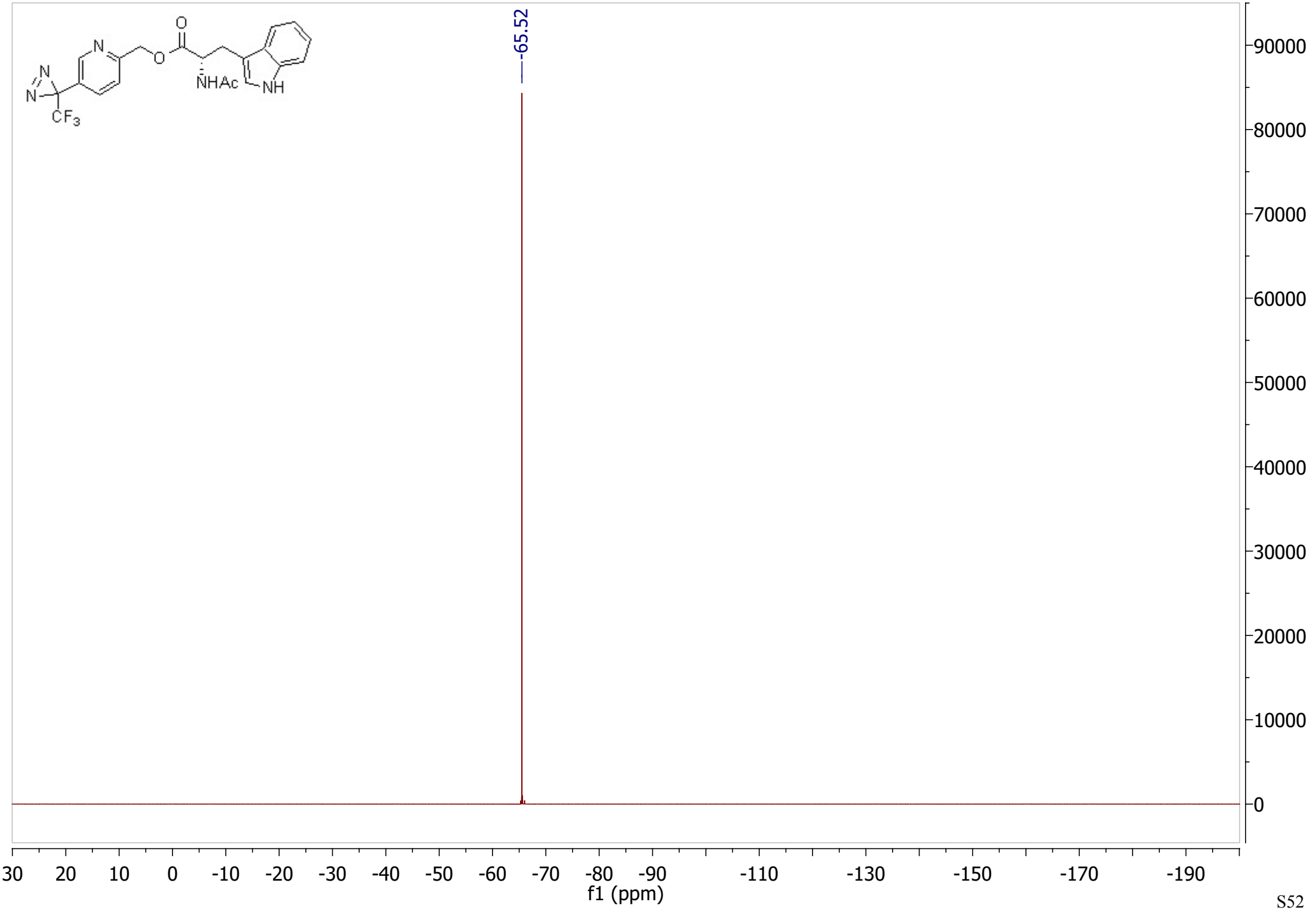

8.33
7.52
7.50
7.36
7.34
7.18
7.14
7.12
6.82
6.82
6.10
6.08
5.06
5.00
4.98
3.31
3.30
1.95
NHAc
NH
CF3
0.89
0.99
1.00
3.06
3.08
0.99
0.94
1.97
1.06
2.08
3.00
f1 (ppm)

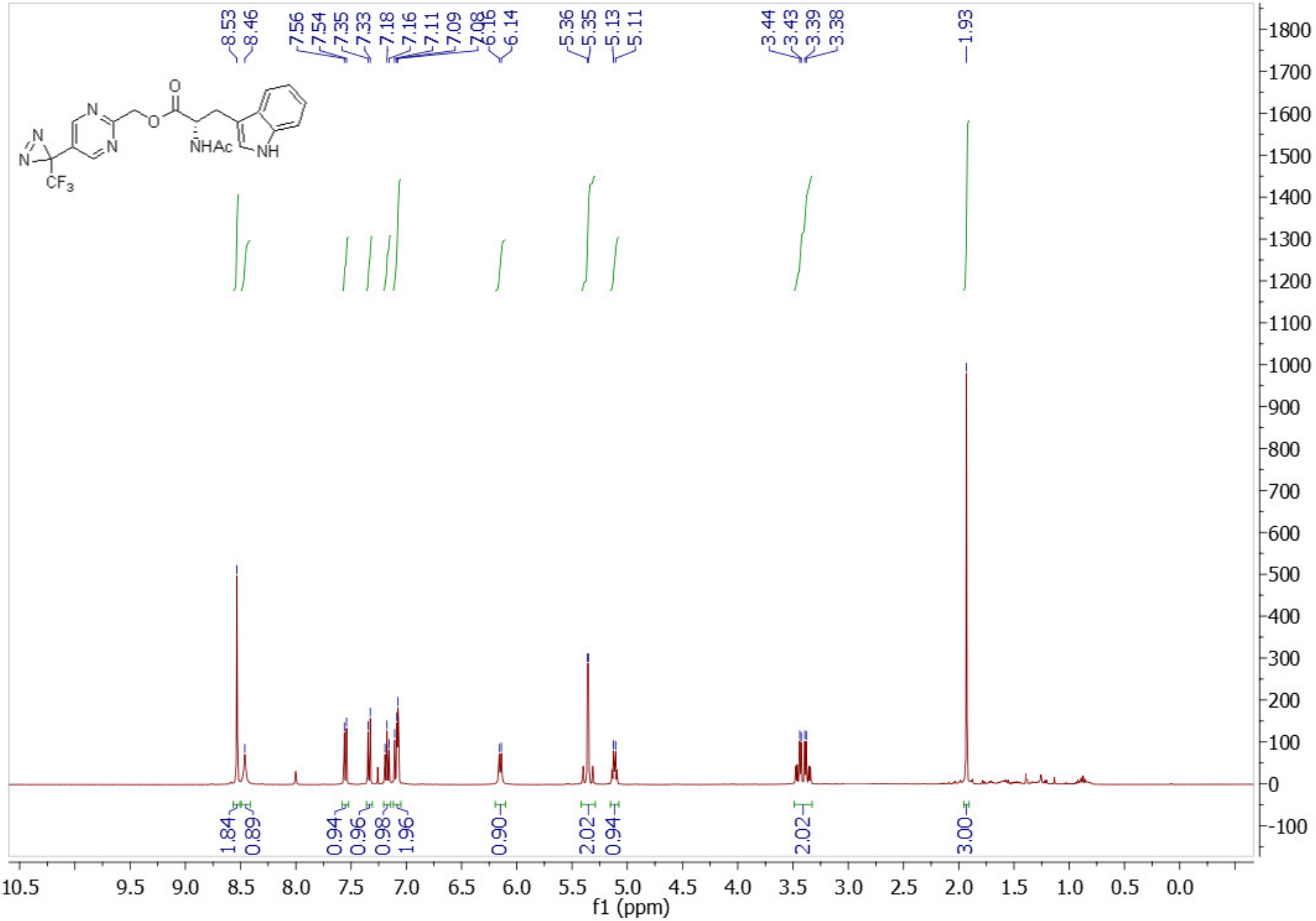

171.93
170.04
137.02
136.26
129.23
128.58
127.71
126.77
123.53
122.89
122.41
120.80
119.85
118.55
111.48
109.83
66.28
53.27
28.62
28.22
27.84
23.26
O
O
N
N
CF3
NHAc
NH
f1 (ppm)

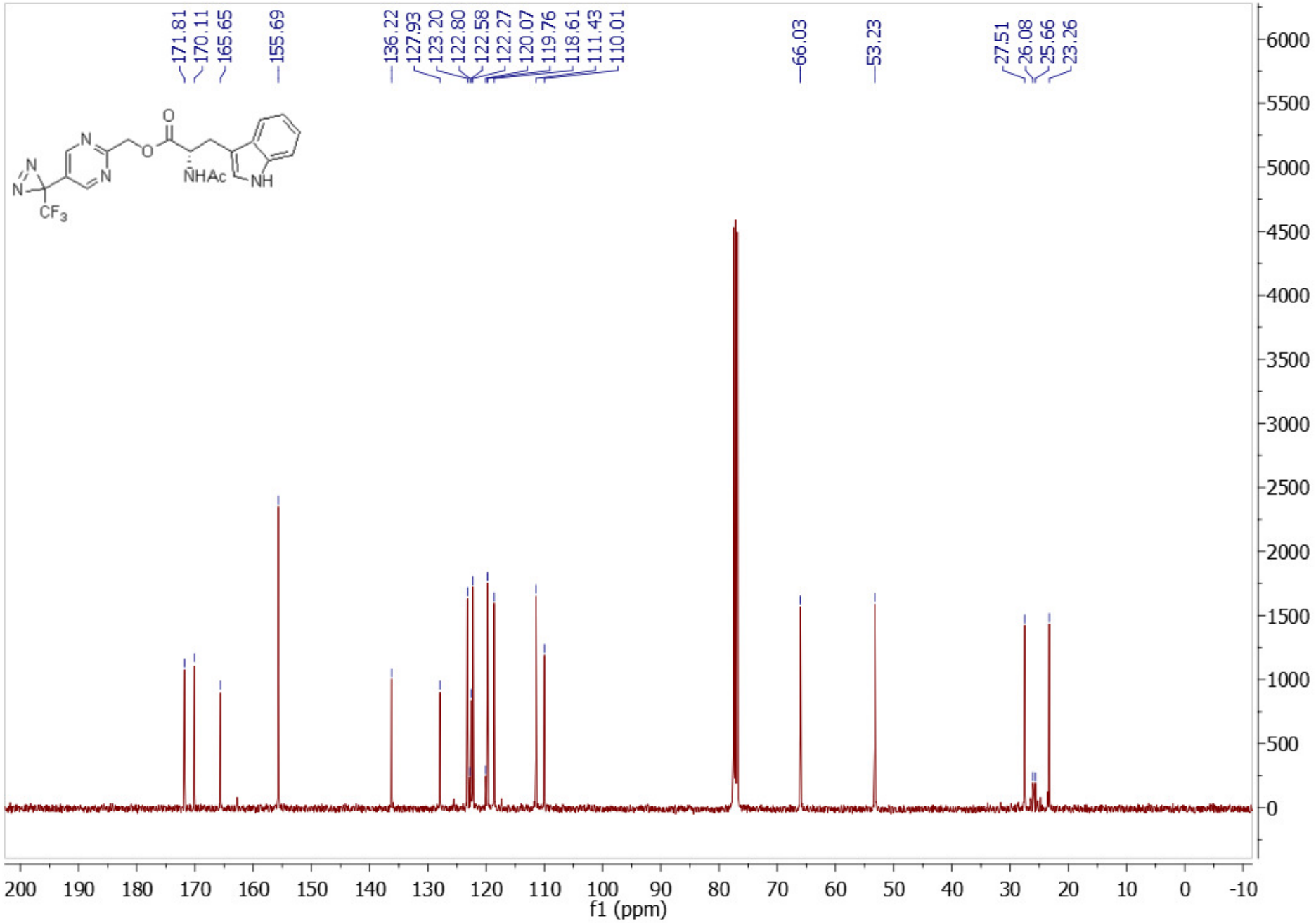

-65.22
45000
40000
35000
30000
25000
20000
15000
10000
5000
0
30 20 10 0 -10 -20 -30 -40 -50 -60 -70 -80 -90 -110 -130 -150 -170 -190
f1 (ppm)

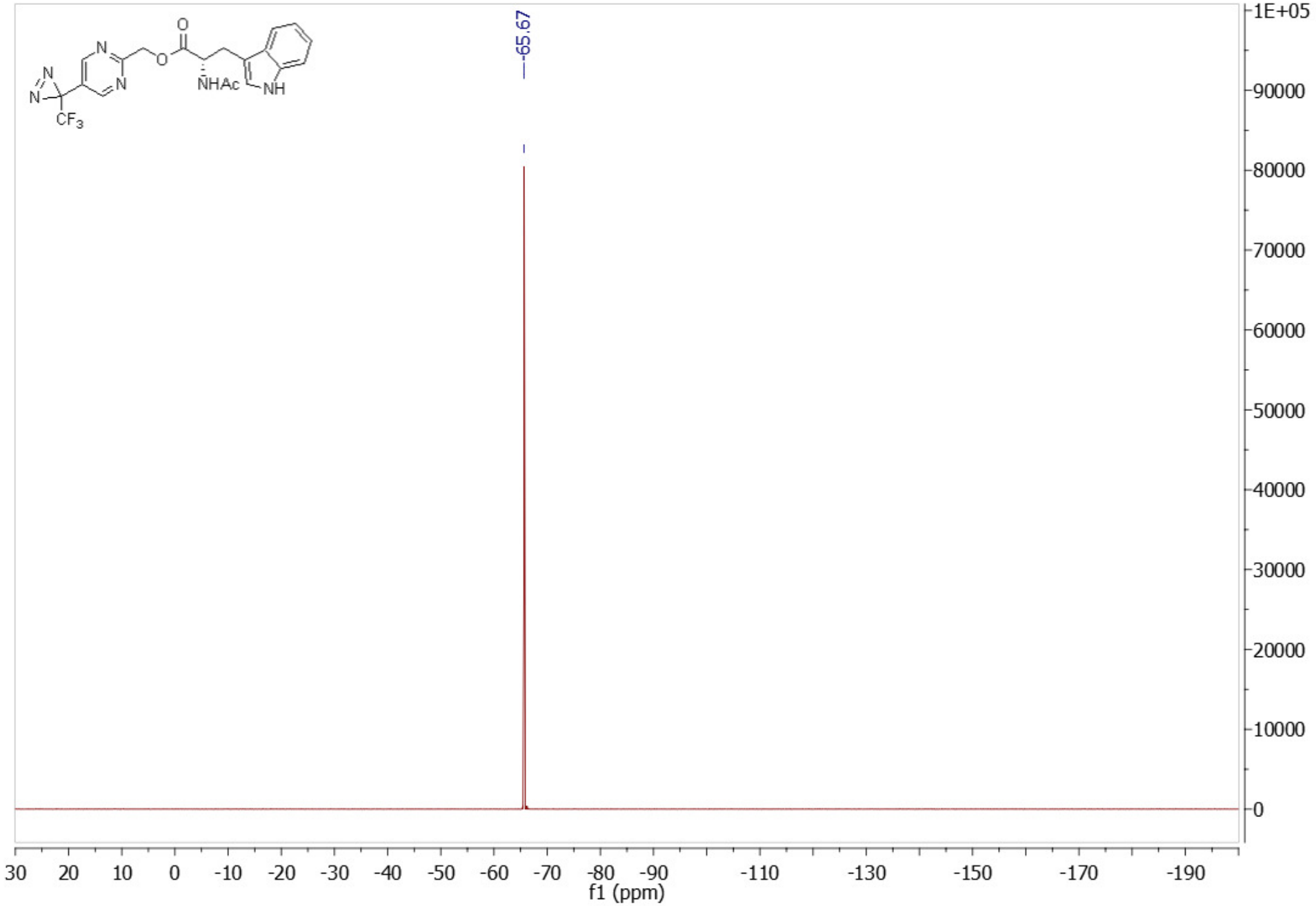

8.49
8.39
7.52
7.34
7.32
7.17
7.09
7.08
7.05
6.98
6.98
6.20
6.18
5.21
5.18
5.04
5.02
3.34
3.32
1.96
NHAc
NH
CF3
0.90
0.94
0.98
0.94
0.97
1.01
1.98
0.98
0.92
2.02
0.96
1.99
3.00
f1 (ppm)

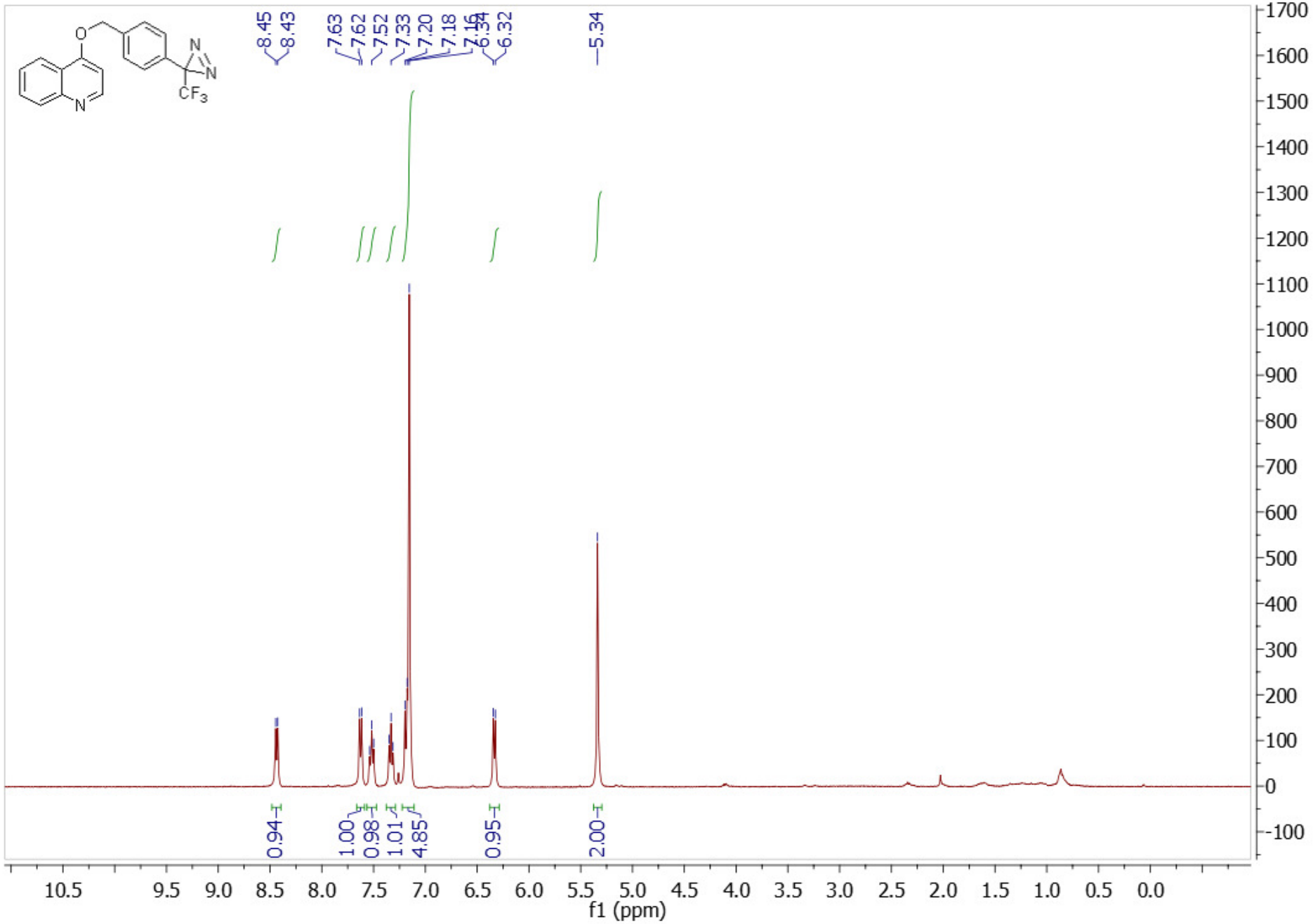

171.85
170.20
156.78
147.24
136.28
135.37
127.66
124.69
123.15
123.05
122.40
121.24
120.42
119.88
118.53
111.48
109.82
66.66
53.38
27.85
27.36
26.95
23.21
NHAc
NH
CF3
f1 (ppm)

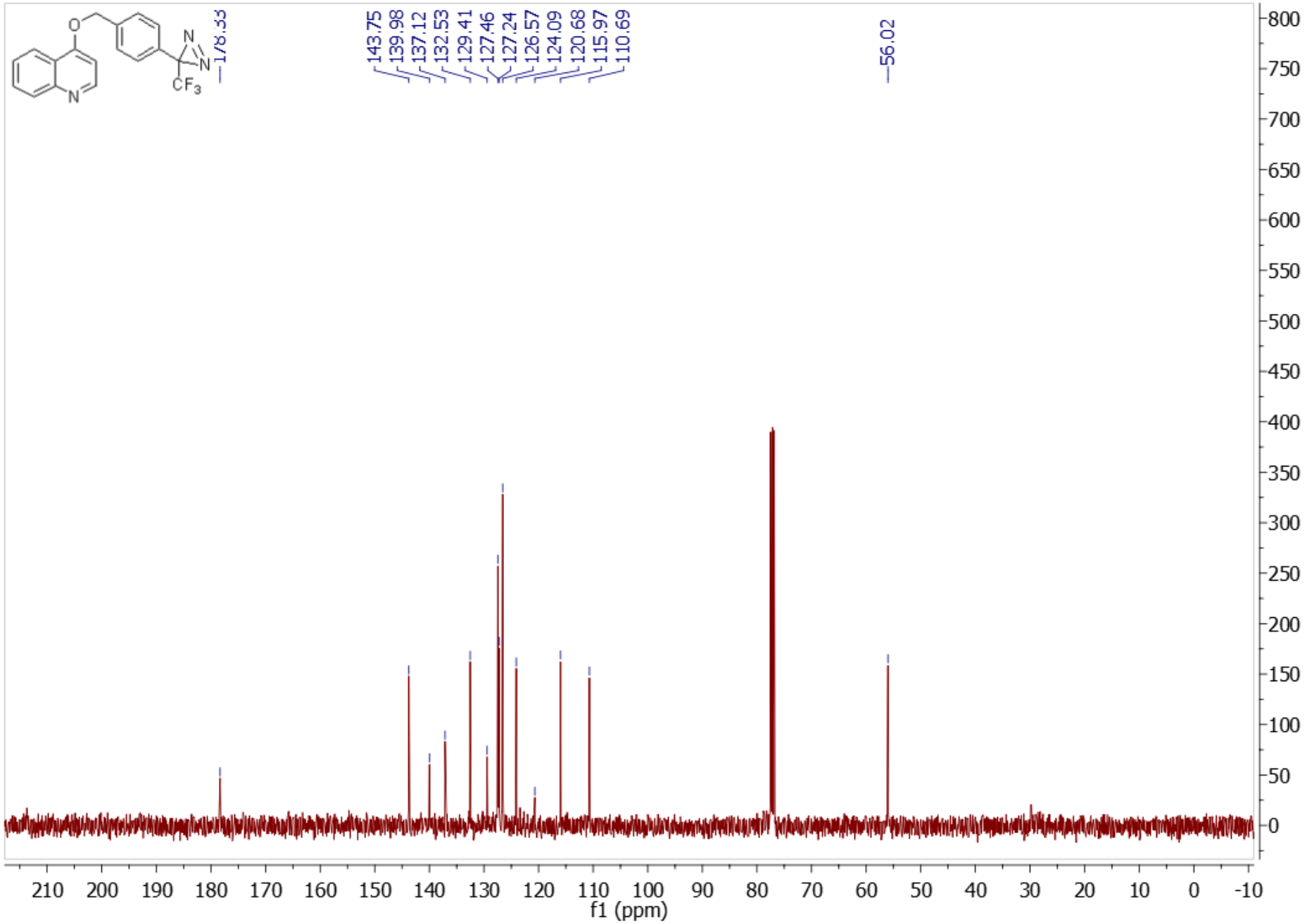

-65.52
90000
80000
70000
60000
50000
40000
30000
20000
10000
0
30 20 10 0 -10 -20 -30 -40 -50 -60 -70 -80 -90 -110 -130 -150 -170 -190
f1 (ppm)

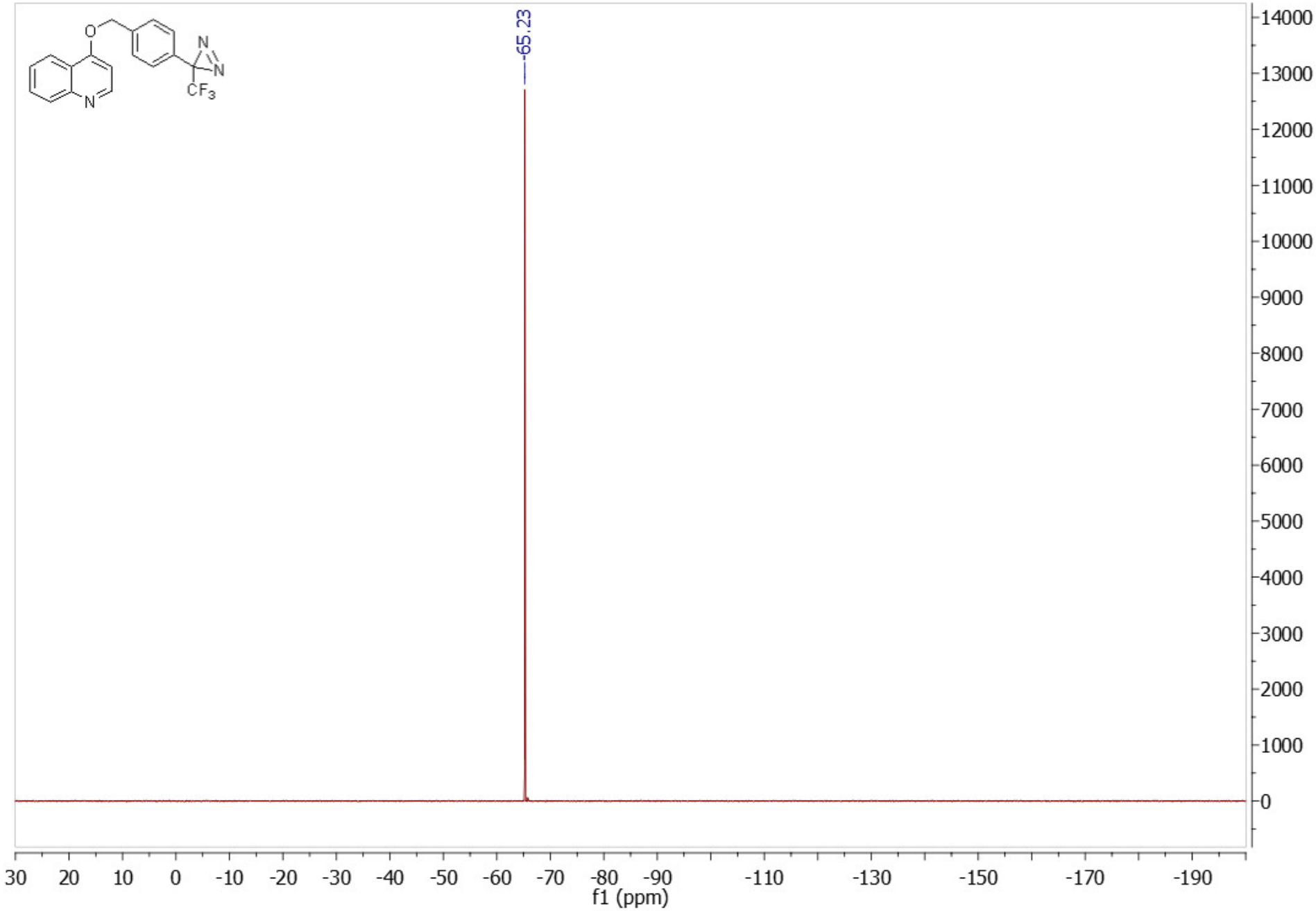

8.53
8.46
7.56
7.54
7.35
7.33
7.18
7.16
7.11
7.09
7.08
6.16
6.14
5.36
5.35
5.13
5.11
3.44
3.43
3.39
3.38
1.93
NHAc
NH
CF3
1.84
0.89
0.94
0.96
0.98
1.96
0.90
2.02
0.94
2.02
3.00
f1 (ppm)

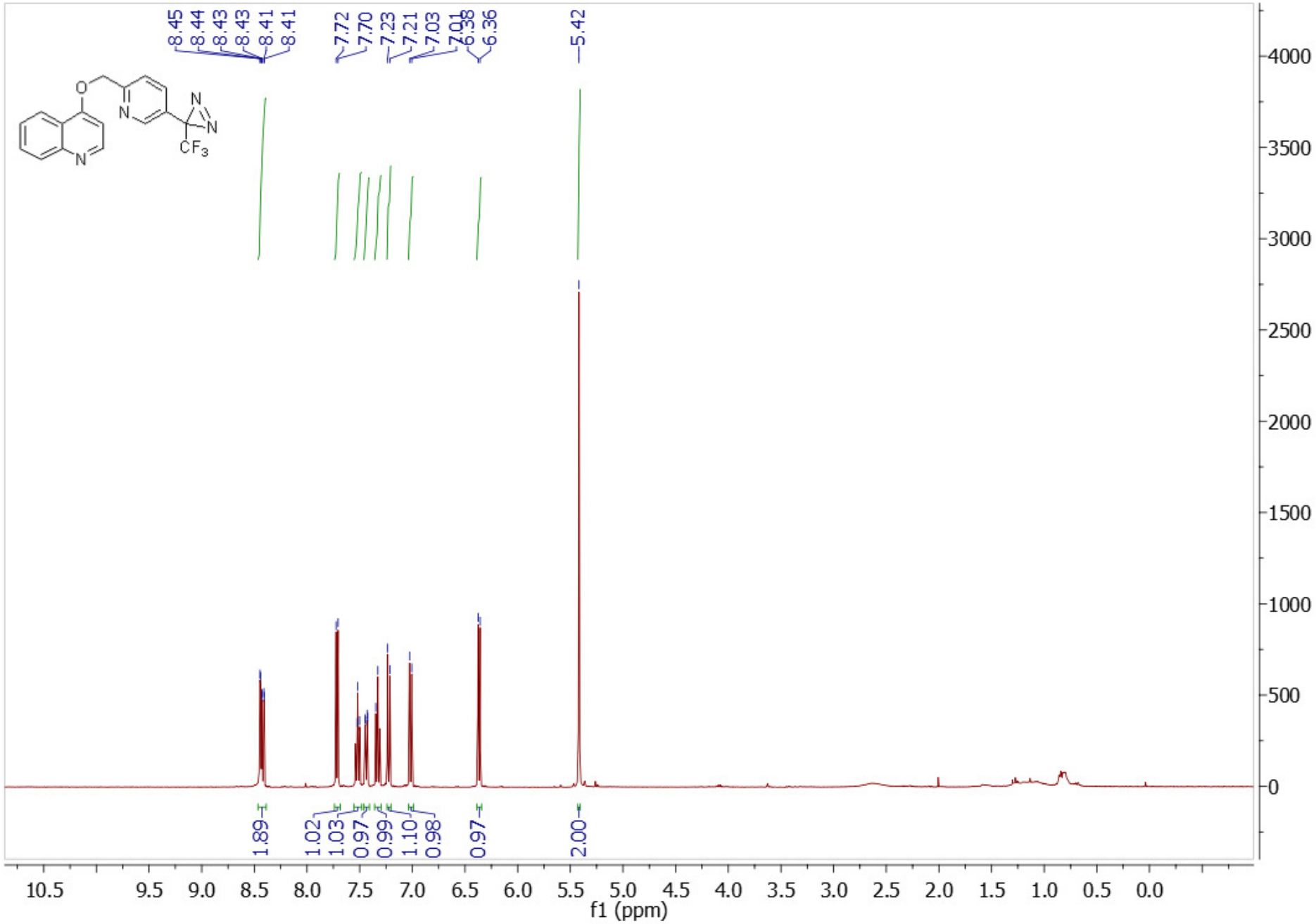

171.81
170.11
165.65
155.69
136.22
127.93
123.20
122.80
122.58
122.27
120.07
119.76
118.61
111.43
110.01
66.03
53.23
27.51
26.08
25.66
23.26
O
N
N
N
O
NHAc
NH
N
CF3
6000
5500
5000
4500
4000
3500
3000
2500
2000
1500
1000
500
0
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

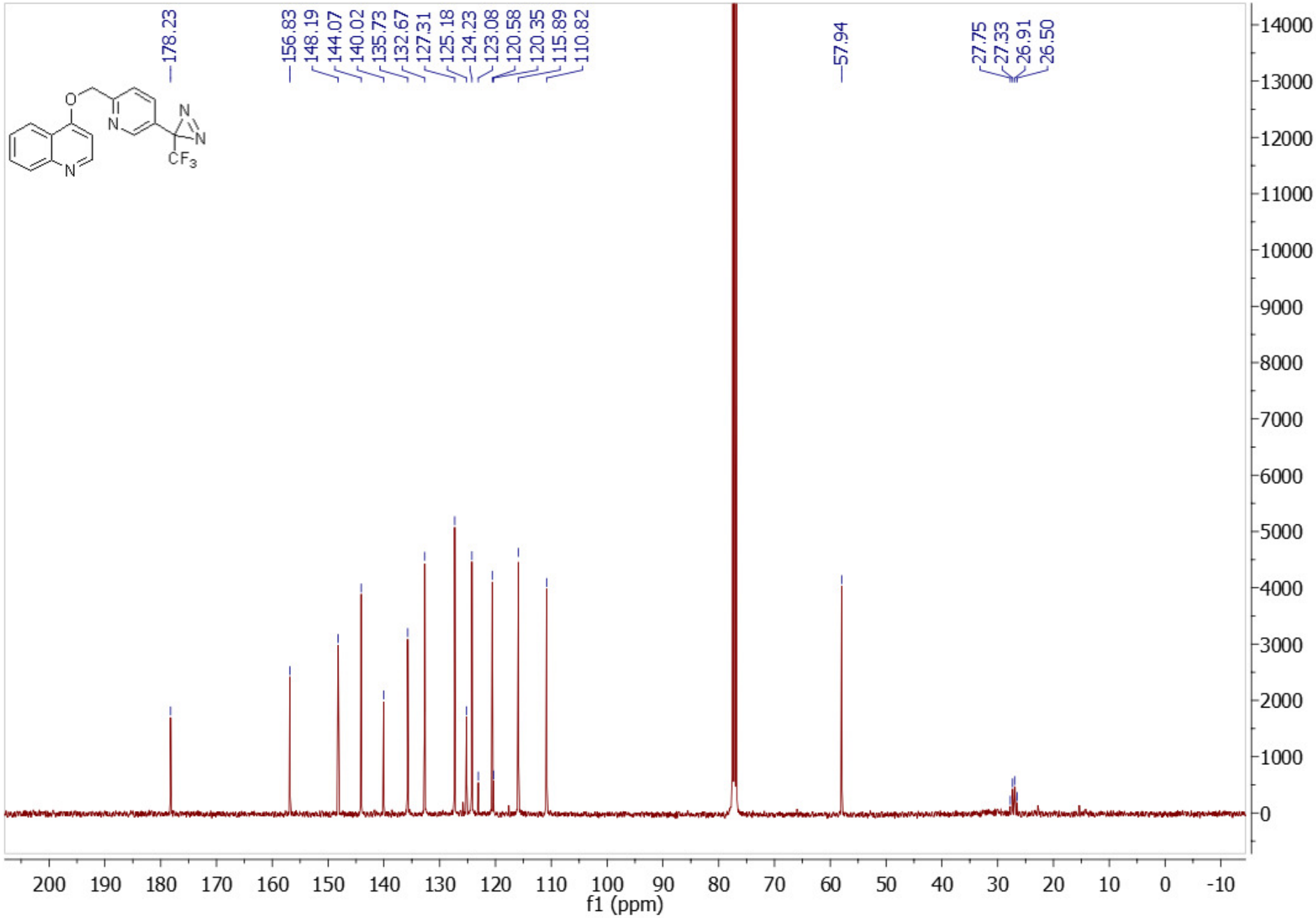

-65.67
1E+05
90000
80000
70000
60000
50000
40000
30000
20000
10000
0
30 20 10 0 -10 -20 -30 -40 -50 -60 -70 -80 -90 -110 -130 -150 -170 -190
f1 (ppm)
O
N
O
N
N
NHAc
NH
N
CF3

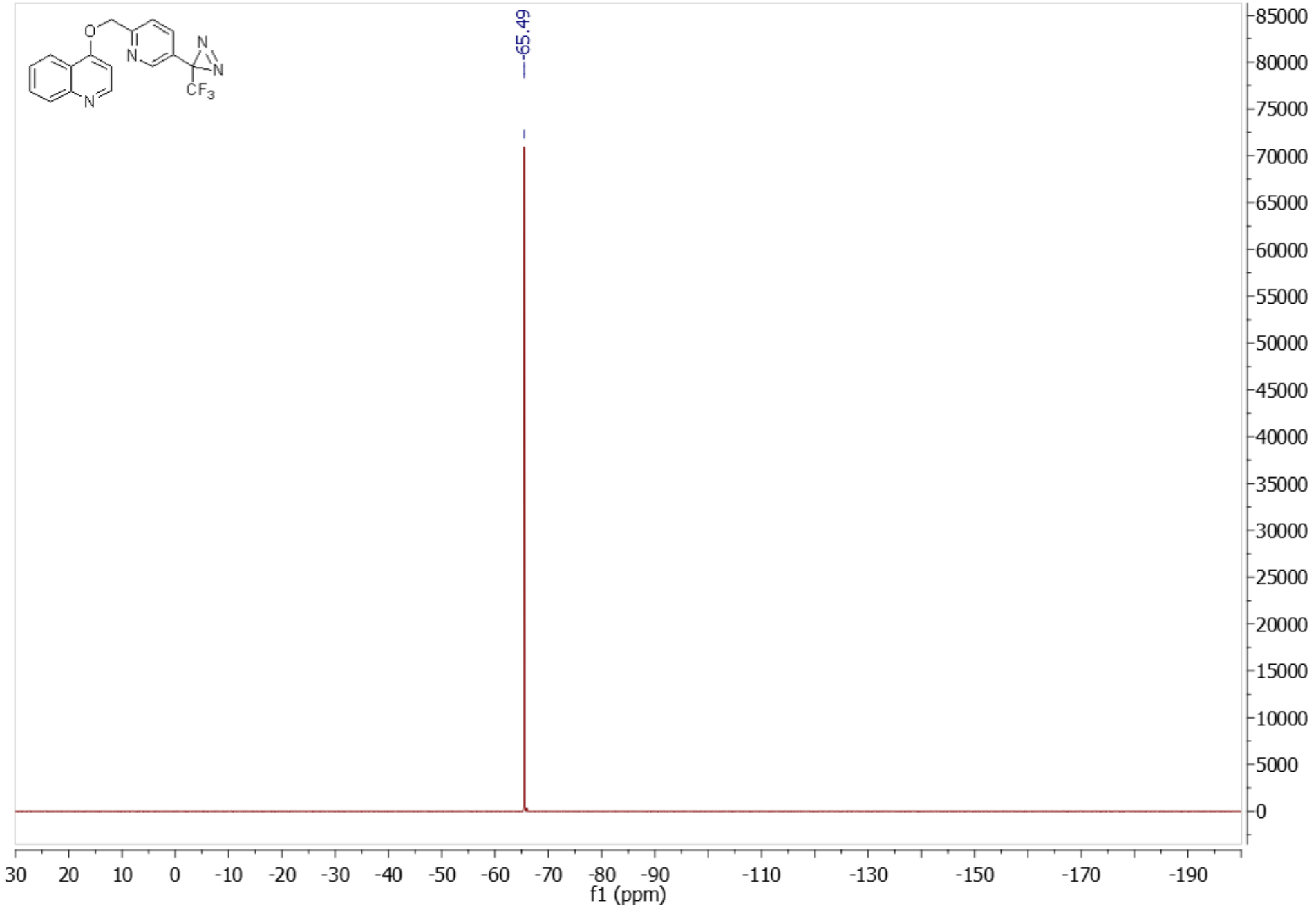

8.45
8.43
7.63
7.62
7.52
7.33
7.20
7.18
7.16
7.14
6.34
6.32
5.34
0.94
1.00
0.98
1.01
4.85
0.95
2.00
f1 (ppm)

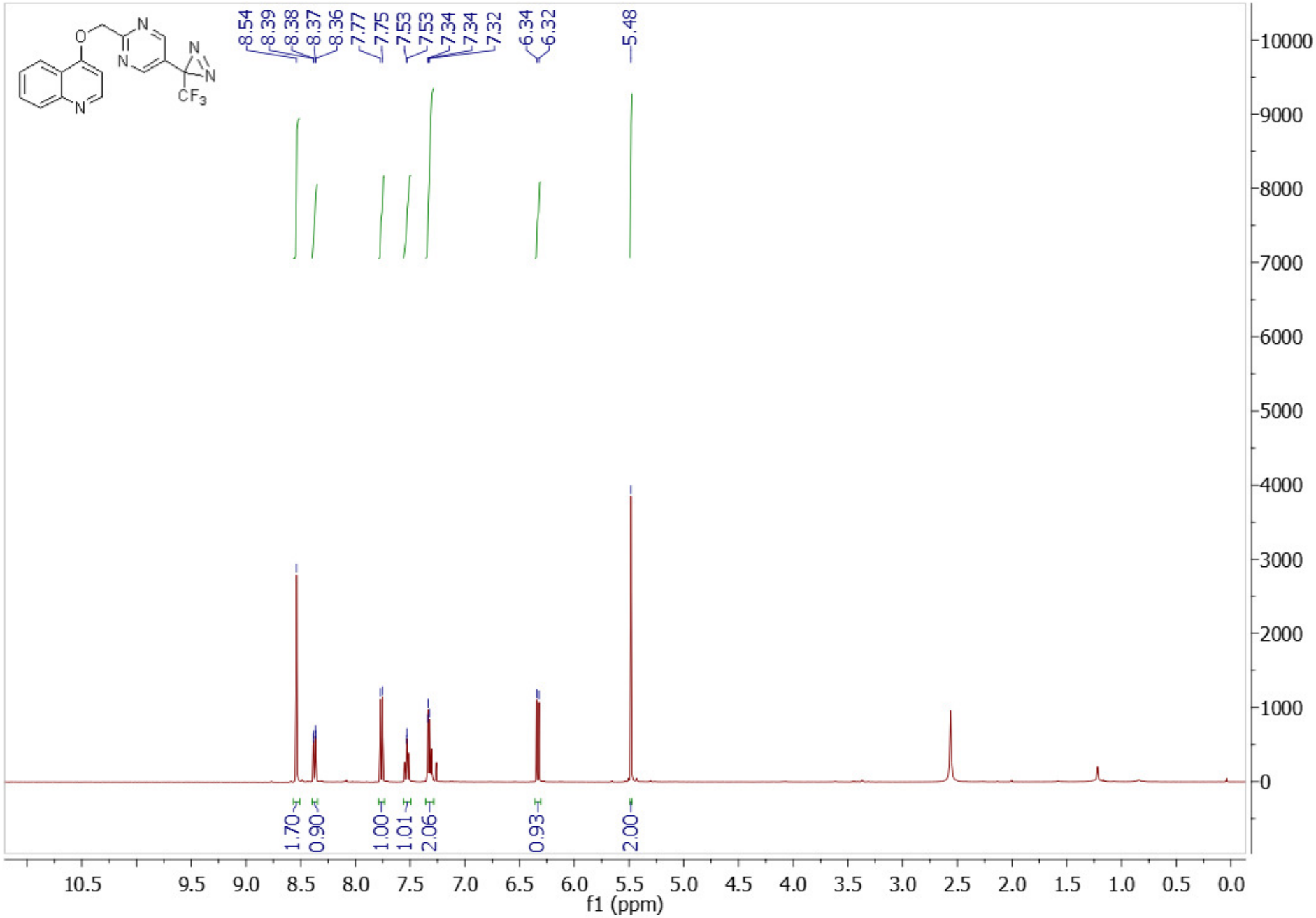

178.33
143.75
139.98
137.12
132.53
129.41
127.46
127.24
126.57
124.09
120.68
115.97
110.69
56.02
f1 (ppm)

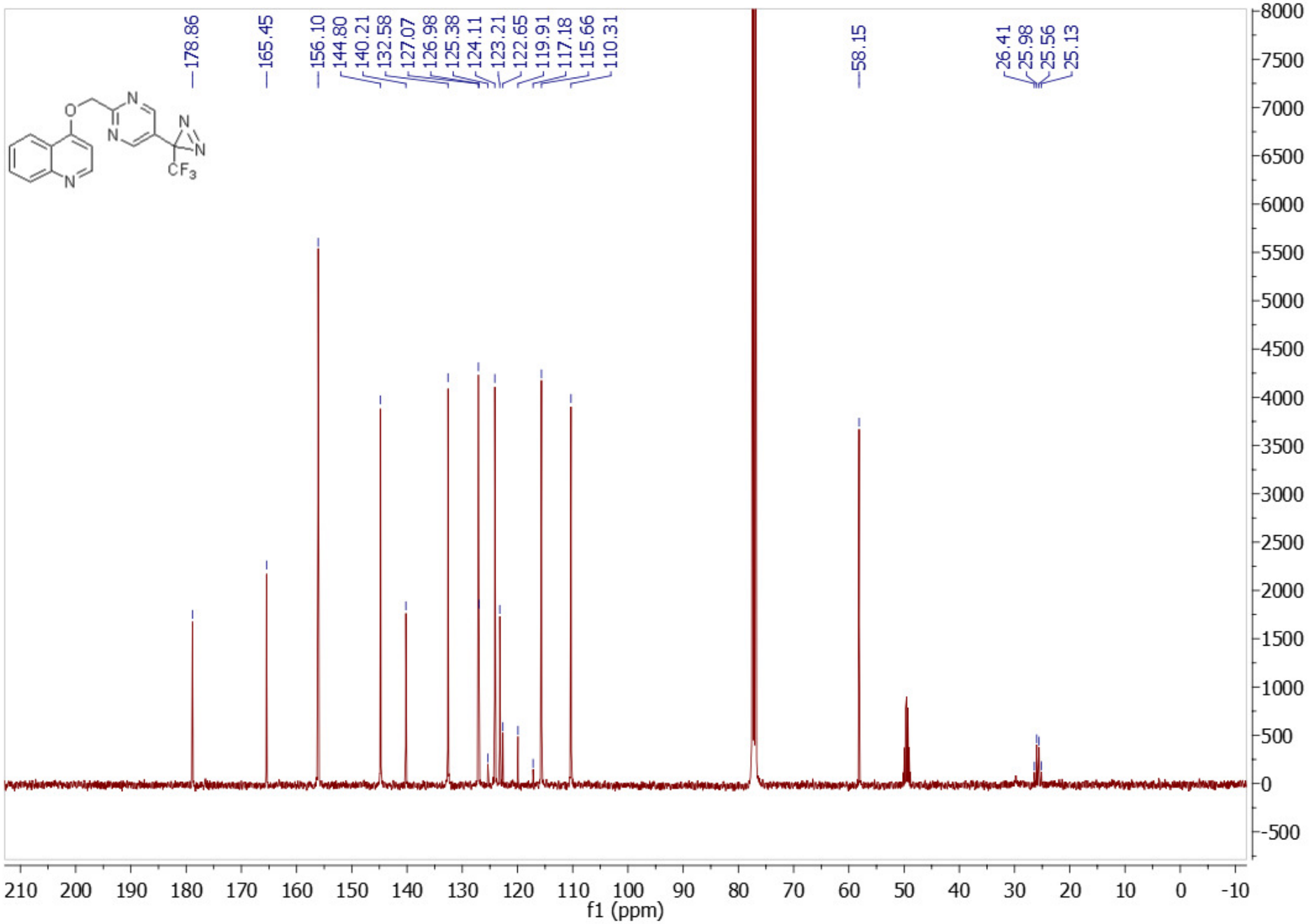

-65.23
14000
13000
12000
11000
10000
9000
8000
7000
6000
5000
4000
3000
2000
1000
0
30 20 10 0 -10 -20 -30 -40 -50 -60 -70 -80 -90 -110 -130 -150 -170 -190
f1 (ppm)
O
N
N
N
CF3

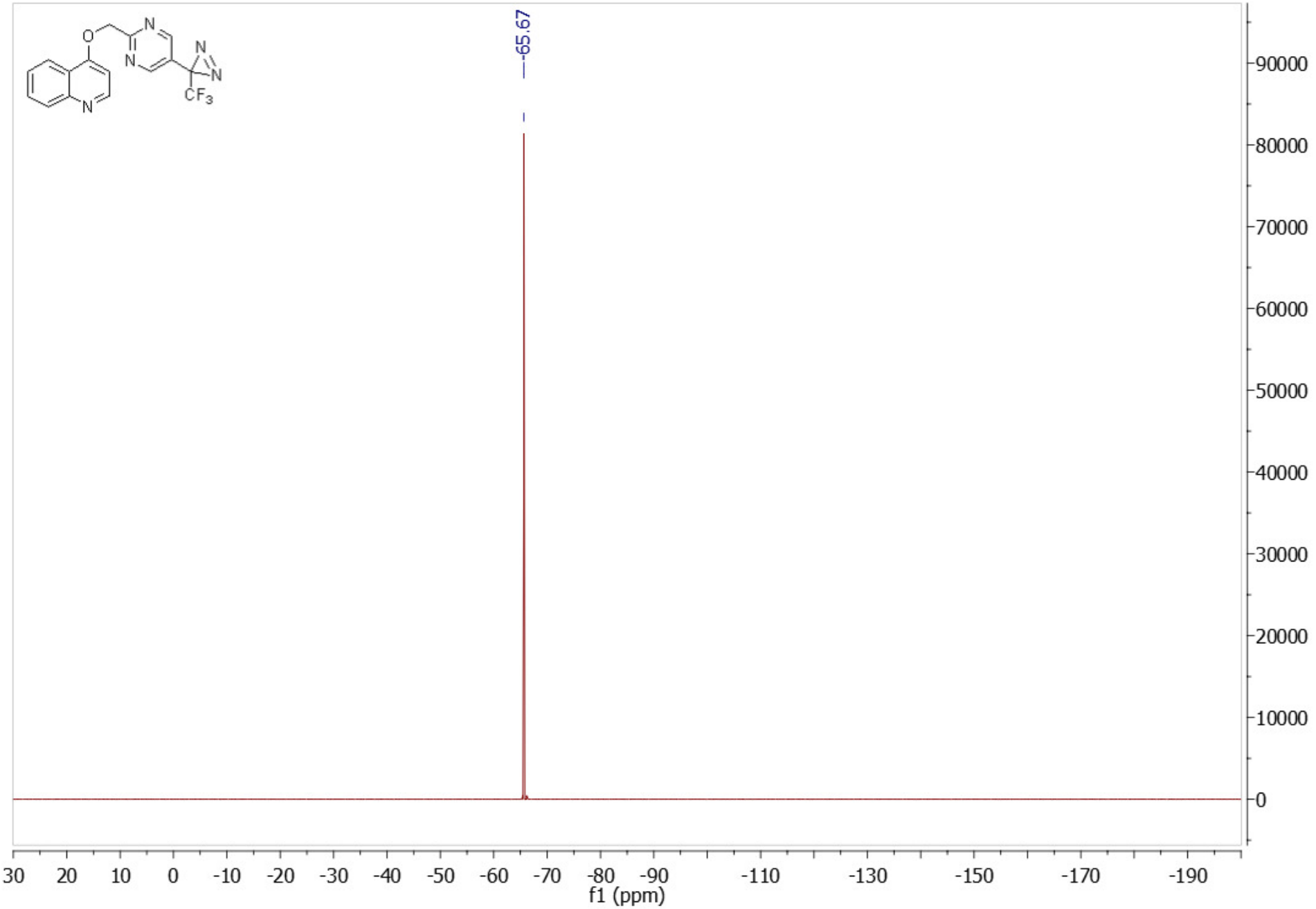

8.45
8.44
8.43
8.43
8.41
8.41
7.72
7.70
7.23
7.21
7.03
7.01
6.38
6.36
5.42
O
N
N
N
N
CF3
4000
3500
3000
2500
2000
1500
1000
500
0
1.89
1.02
1.03
0.97
0.99
1.10
0.98
0.97
2.00
10.5
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
f1 (ppm)

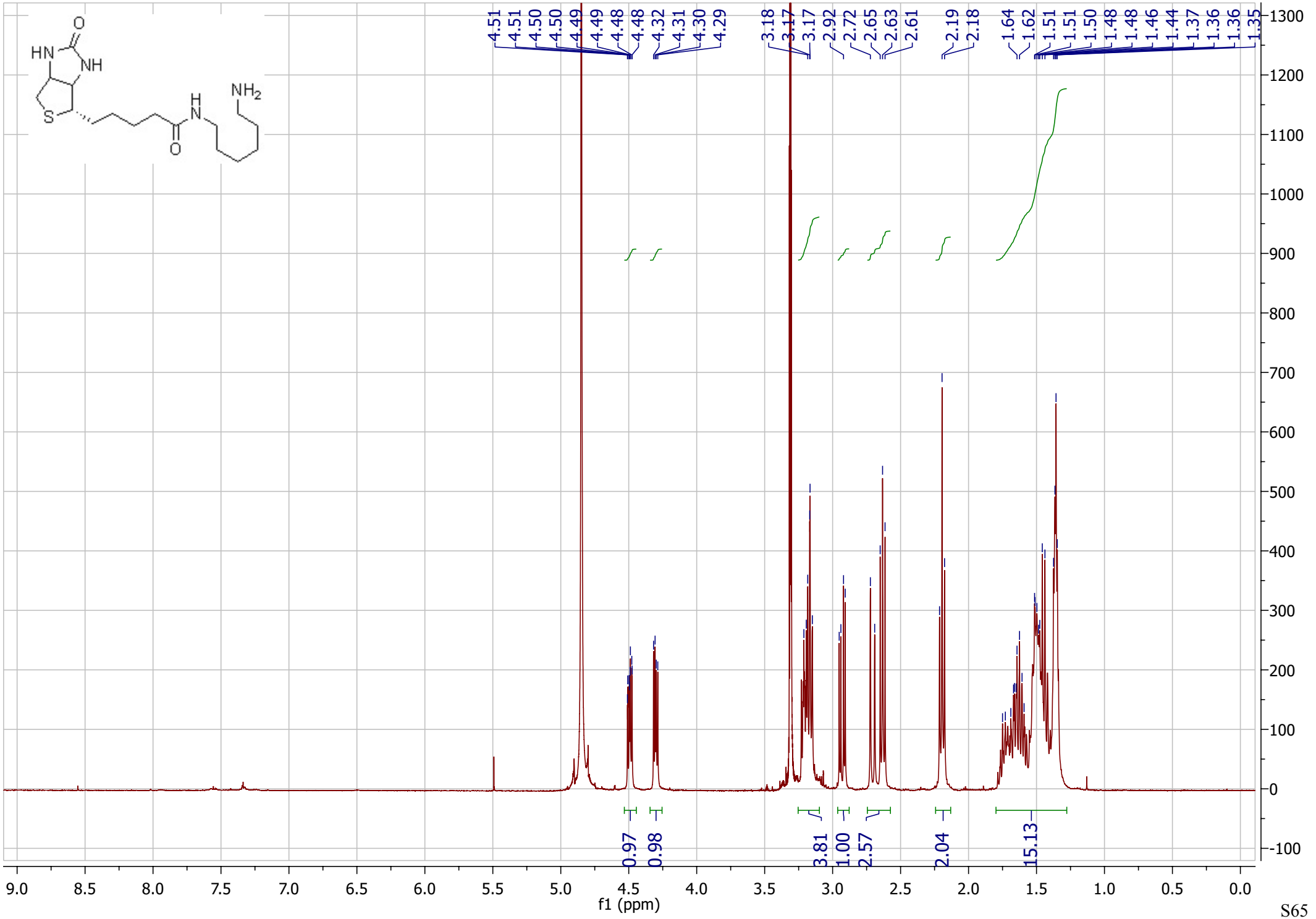

178.23
156.83
148.19
144.07
140.02
135.73
132.67
127.31
125.18
124.23
123.08
120.58
120.35
115.89
110.82
57.94
27.75
27.33
26.91
26.50
O
N
N
N
N
CF3
14000
13000
12000
11000
10000
9000
8000
7000
6000
5000
4000
3000
2000
1000
0
200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10
f1 (ppm)

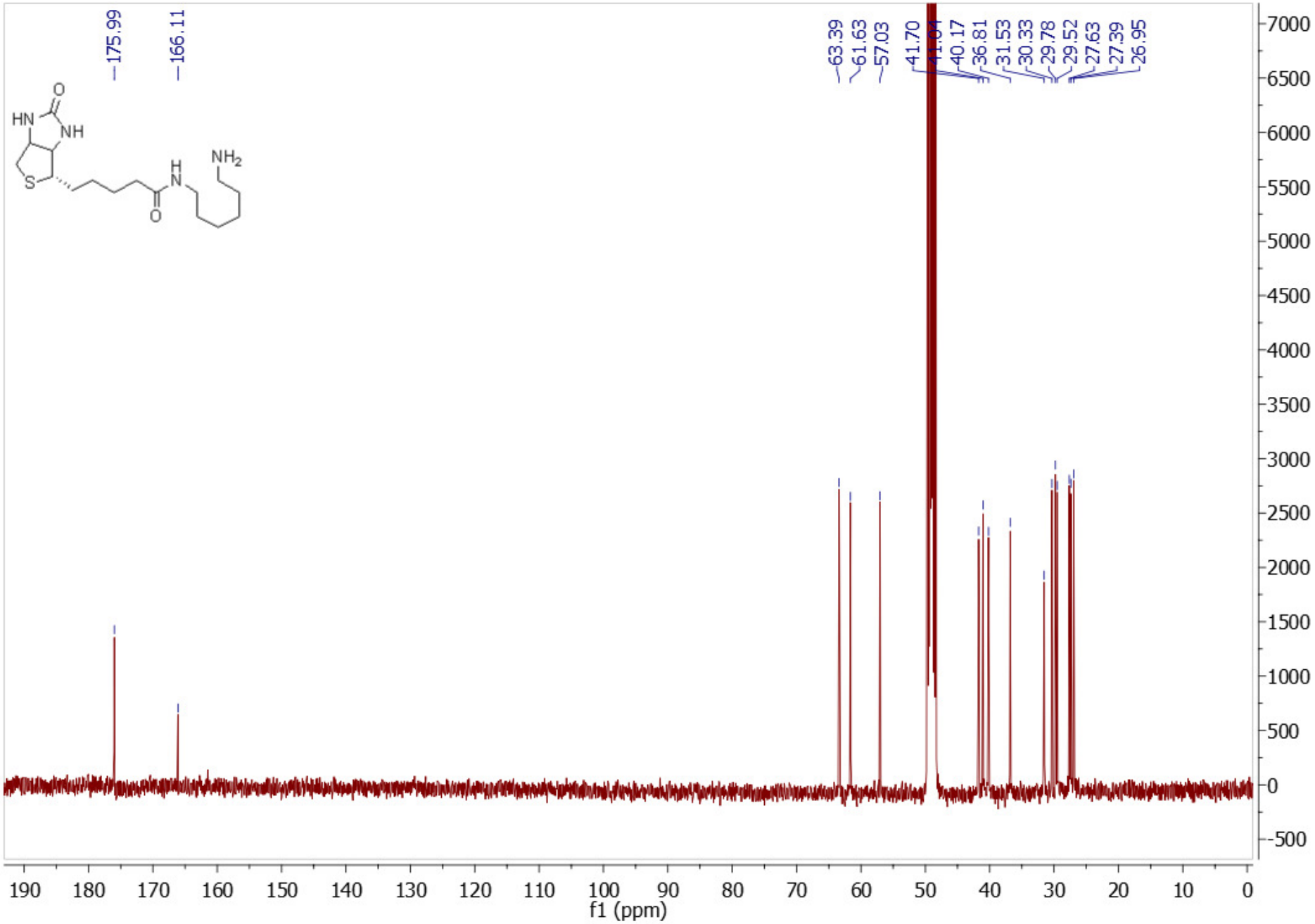

-65.49
85000
80000
75000
70000
65000
60000
55000
50000
45000
40000
35000
30000
25000
20000
15000
10000
5000
0
30 20 10 0 -10 -20 -30 -40 -50 -60 -70 -80 -90 -110 -130 -150 -170 -190
f1 (ppm)

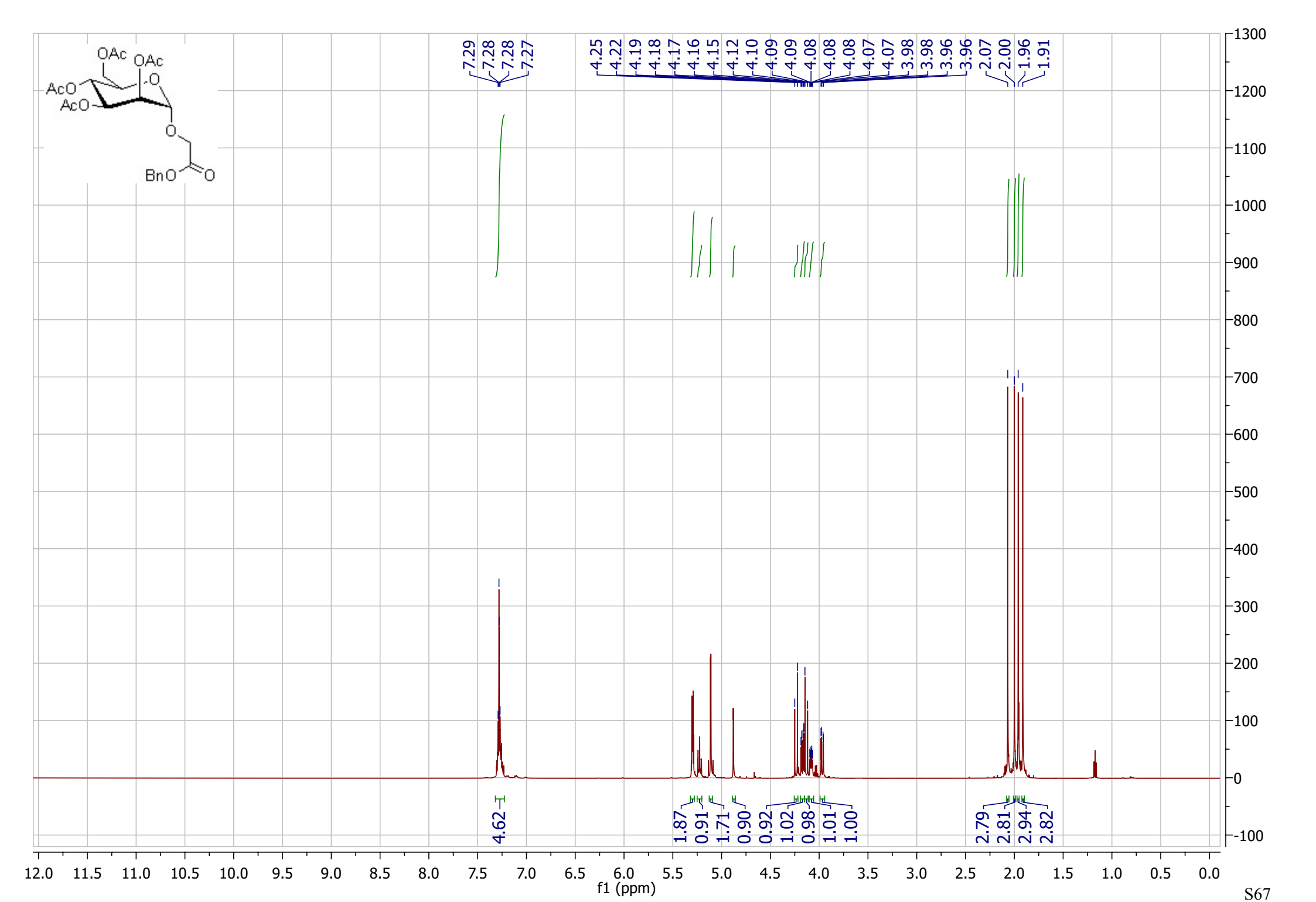

8.54
8.39
8.38
8.37
8.36
7.77
7.75
7.53
7.53
7.34
7.34
7.32
6.34
6.32
5.48
1.70
0.90
1.00
1.01
2.06
0.93
2.00
f1 (ppm)

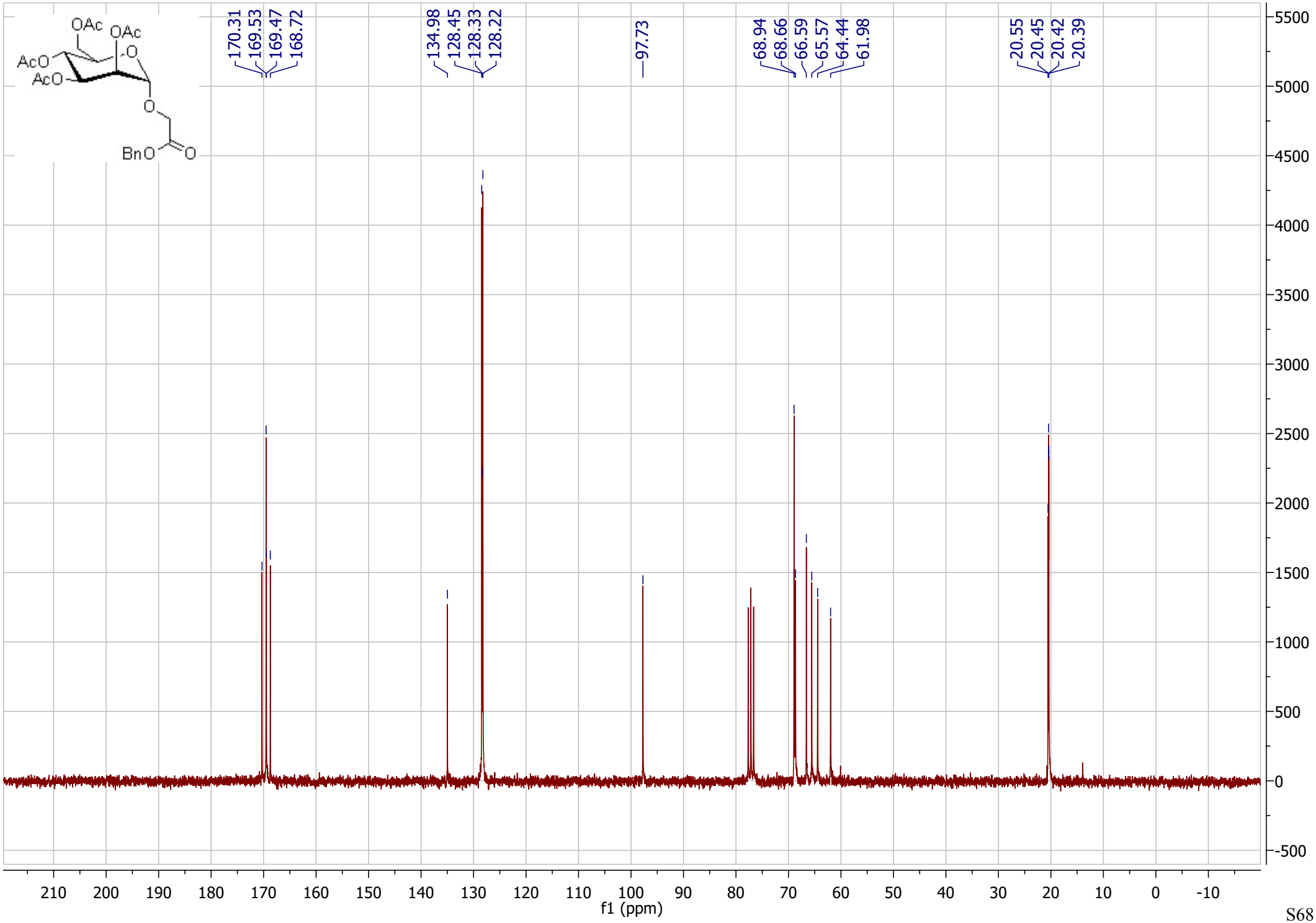

178.86
165.45
156.10
144.80
140.21
132.58
127.07
126.98
125.38
124.11
123.21
122.65
119.91
117.18
115.66
110.31
58.15
26.41
25.98
25.56
25.13
f1 (ppm)

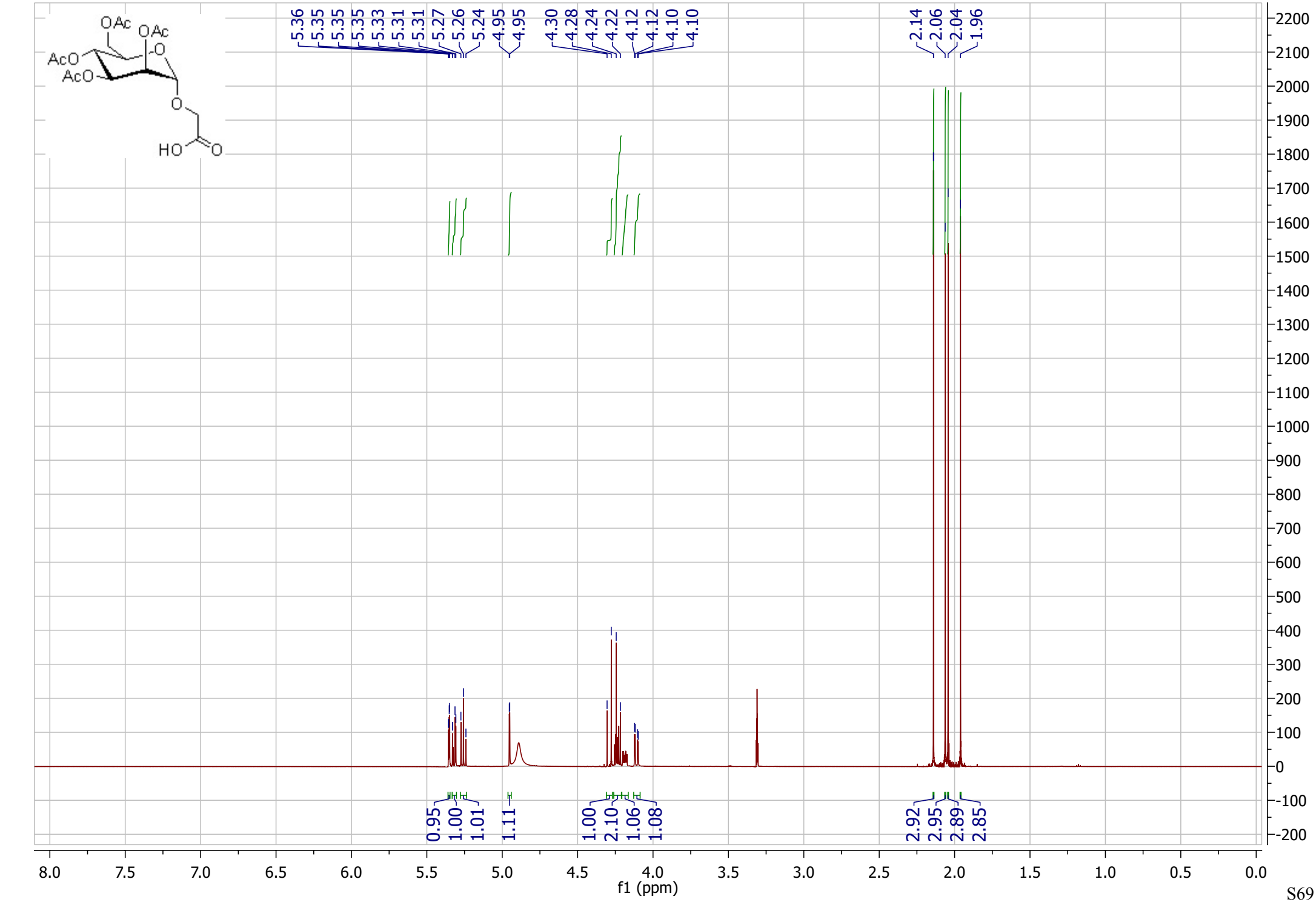

-65.67
90000
80000
70000
60000
50000
40000
30000
20000
10000
0
30 20 10 0 -10 -20 -30 -40 -50 -60 -70 -80 -90 -110 -130 -150 -170 -190
f1 (ppm)

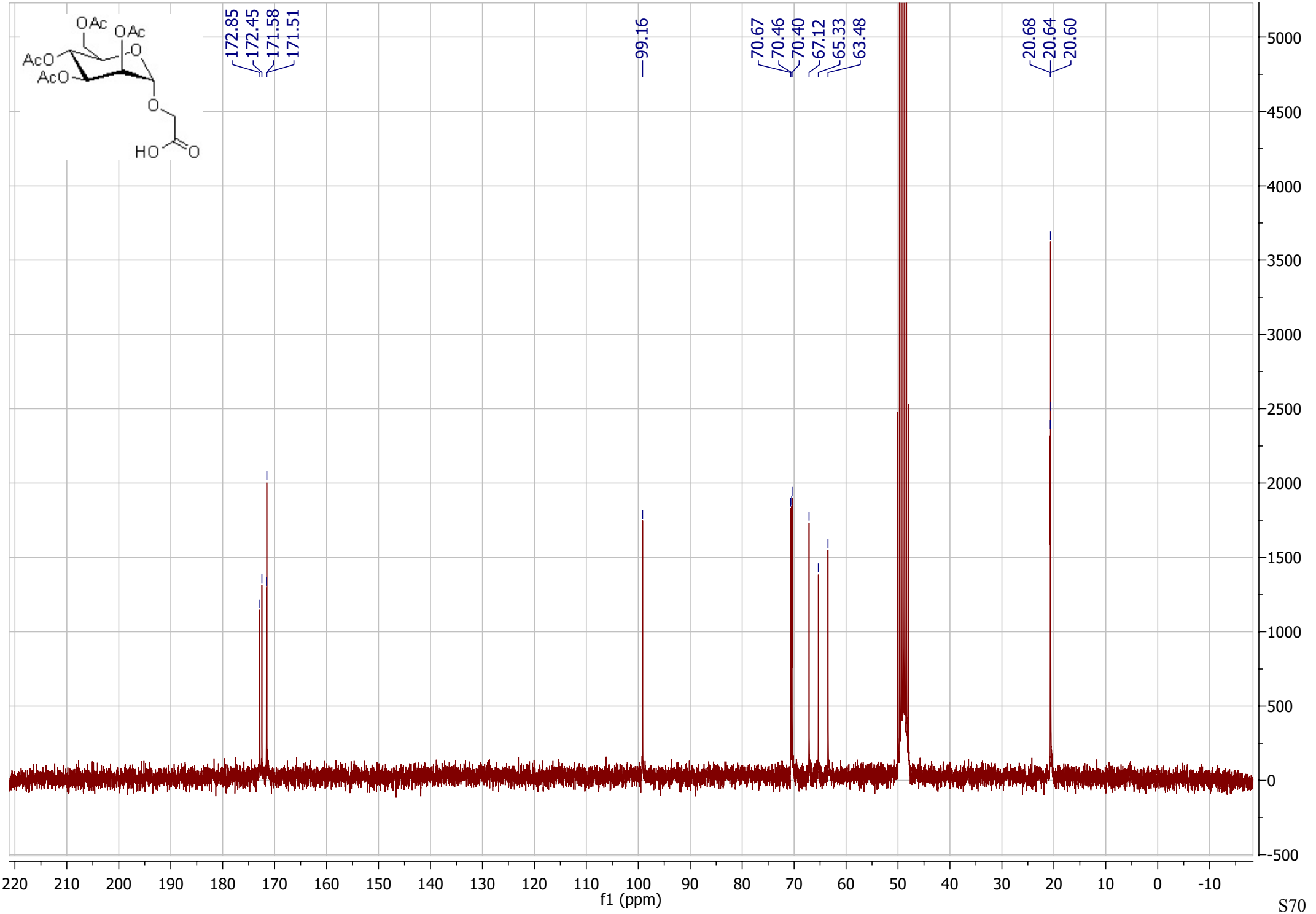

4.51
4.51
4.50
4.50
4.49
4.49
4.48
4.48
4.32
4.31
4.30
4.29
3.18
3.17
3.17
2.92
2.72
2.65
2.63
2.61
2.19
2.18
1.64
1.62
1.51
1.51
1.50
1.48
1.48
1.46
1.44
1.37
1.36
1.36
1.35
0.97
0.98
3.81
1.00
2.57
2.04
15.13
f1 (ppm)

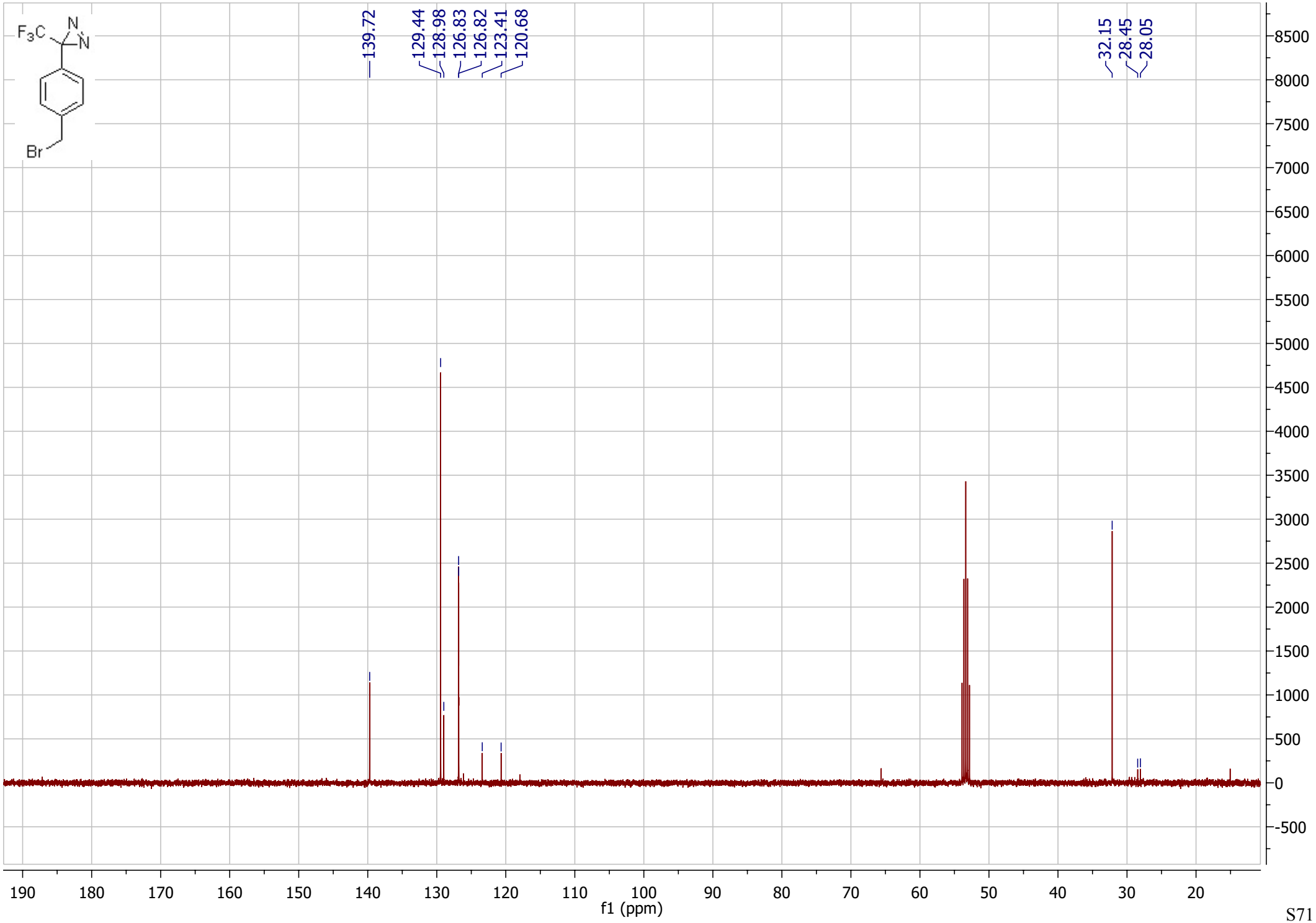

175.99
166.11
63.39
61.63
57.03
41.70
41.04
40.17
36.81
31.53
30.33
29.78
29.52
27.63
27.39
26.95
HN
NH
O
S
H
N
O
NH2
f1 (ppm)

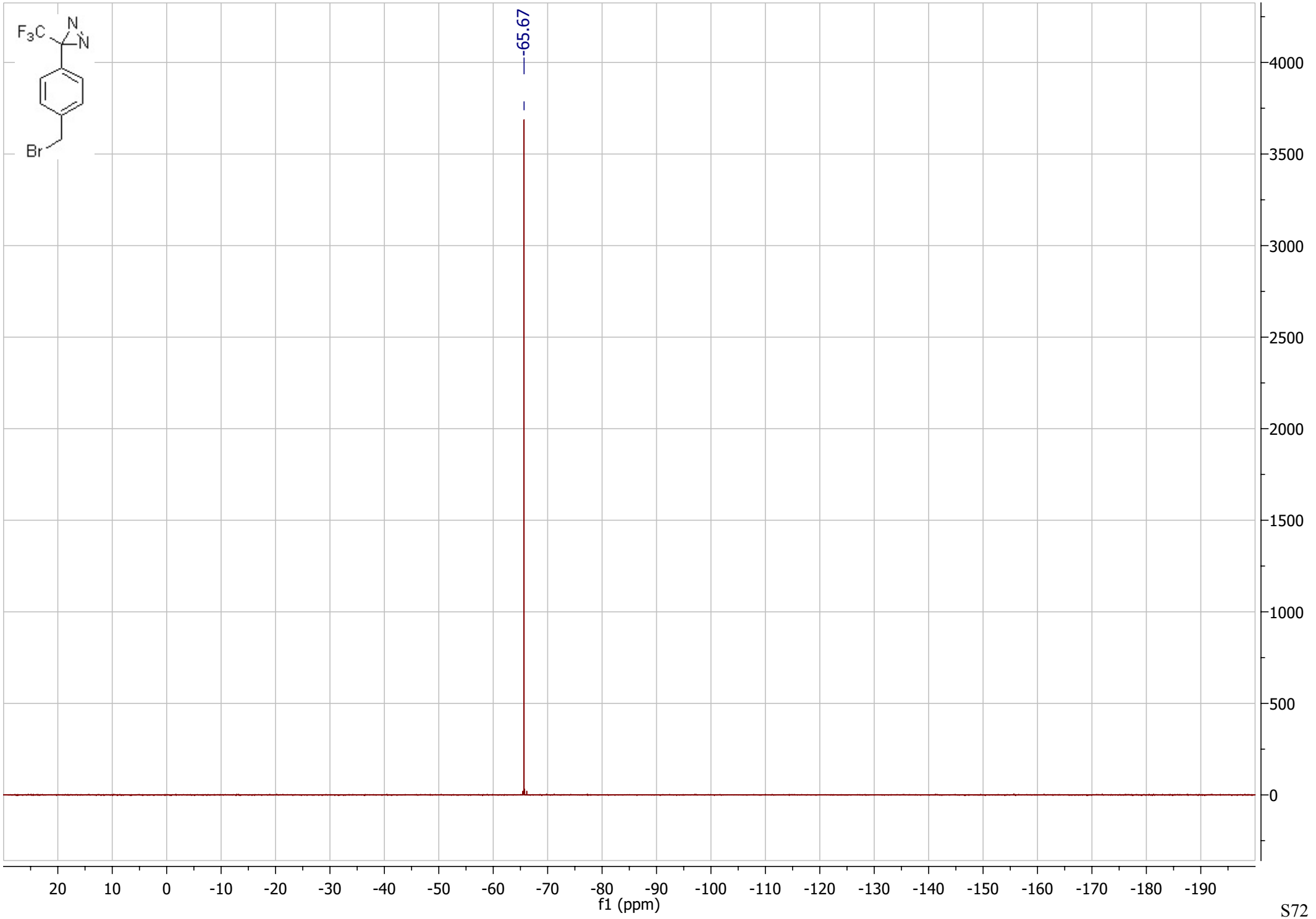

OAc
OAc
AcO
AcO
O
O
BnO
O
7.29
7.28
7.28
7.27
4.25
4.22
4.19
4.18
4.17
4.16
4.15
4.12
4.10
4.09
4.09
4.08
4.08
4.08
4.07
4.07
3.98
3.98
3.96
3.96
2.07
2.00
1.96
1.91
4.62
1.87
0.91
1.71
0.90
0.92
1.02
0.98
1.01
1.00
2.79
2.81
2.94
2.82
f1 (ppm)

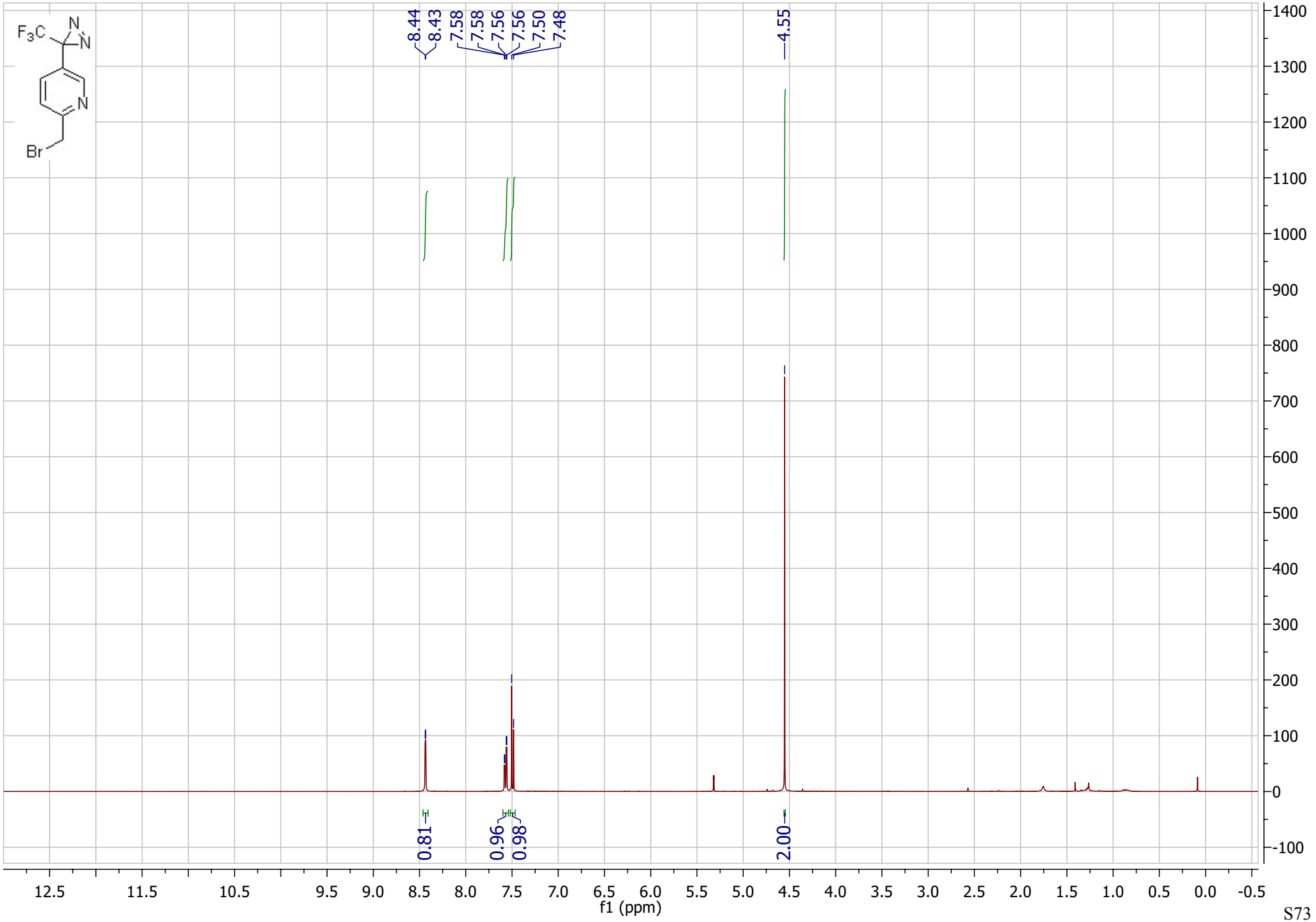

OAc
OAc
AcO
AcO
O
O
BnO
O
170.31
169.53
169.47
168.72
134.98
128.45
128.33
128.22
97.73
68.94
68.66
66.59
65.57
64.44
61.98
20.55
20.45
20.42
20.39
5500
5000
4500
4000
3500
3000
2500
2000
1500
1000
500
0
-500
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

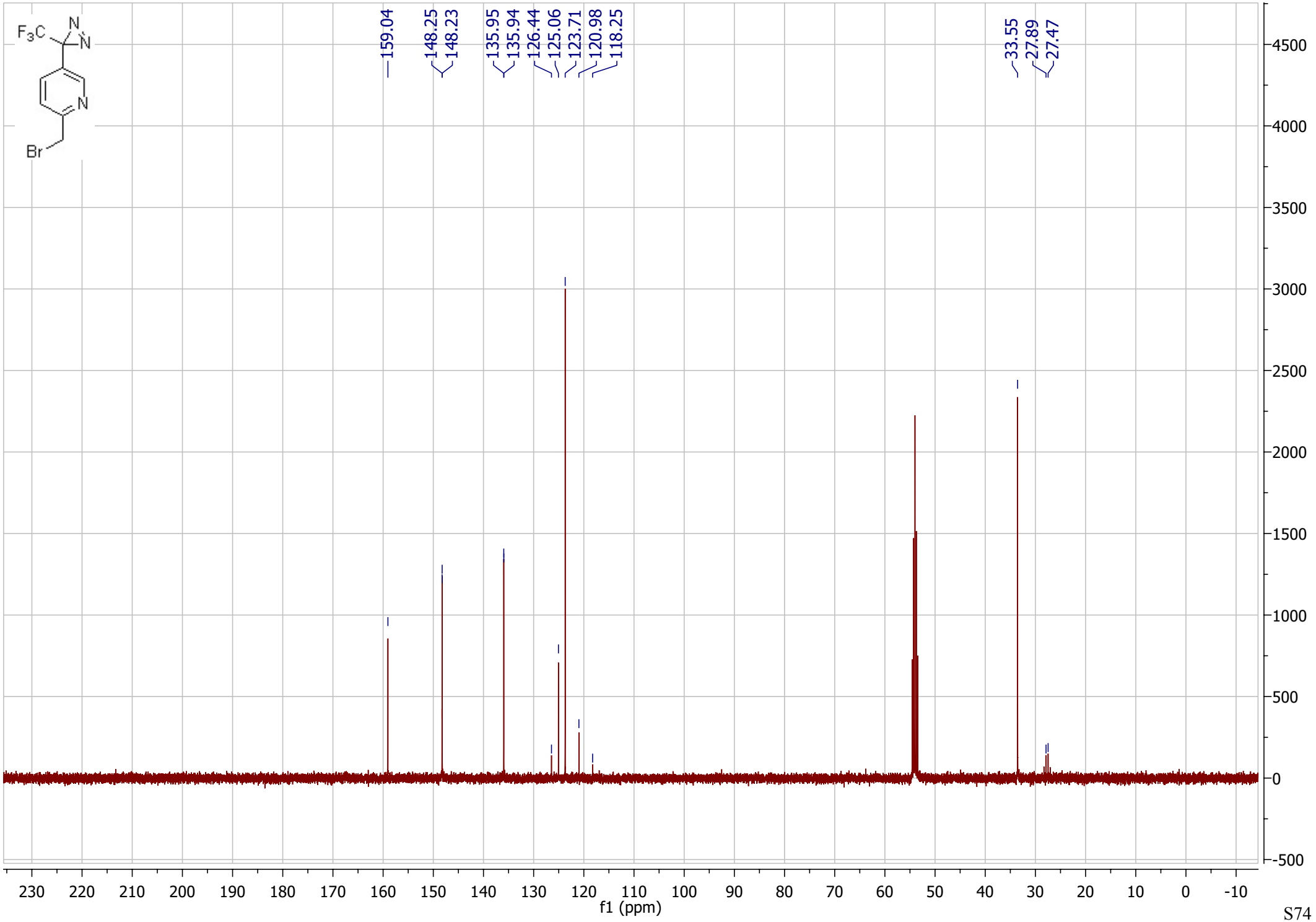

OAc
OAc
AcO
AcO
O
O
HO
O
5.36
5.35
5.35
5.35
5.33
5.31
5.31
5.27
5.26
5.24
4.95
4.95
4.30
4.28
4.24
4.22
4.12
4.12
4.10
4.10
2.14
2.06
2.04
1.96
0.95
1.00
1.01
1.11
1.00
2.10
1.06
1.08
2.92
2.95
2.89
2.85
f1 (ppm)

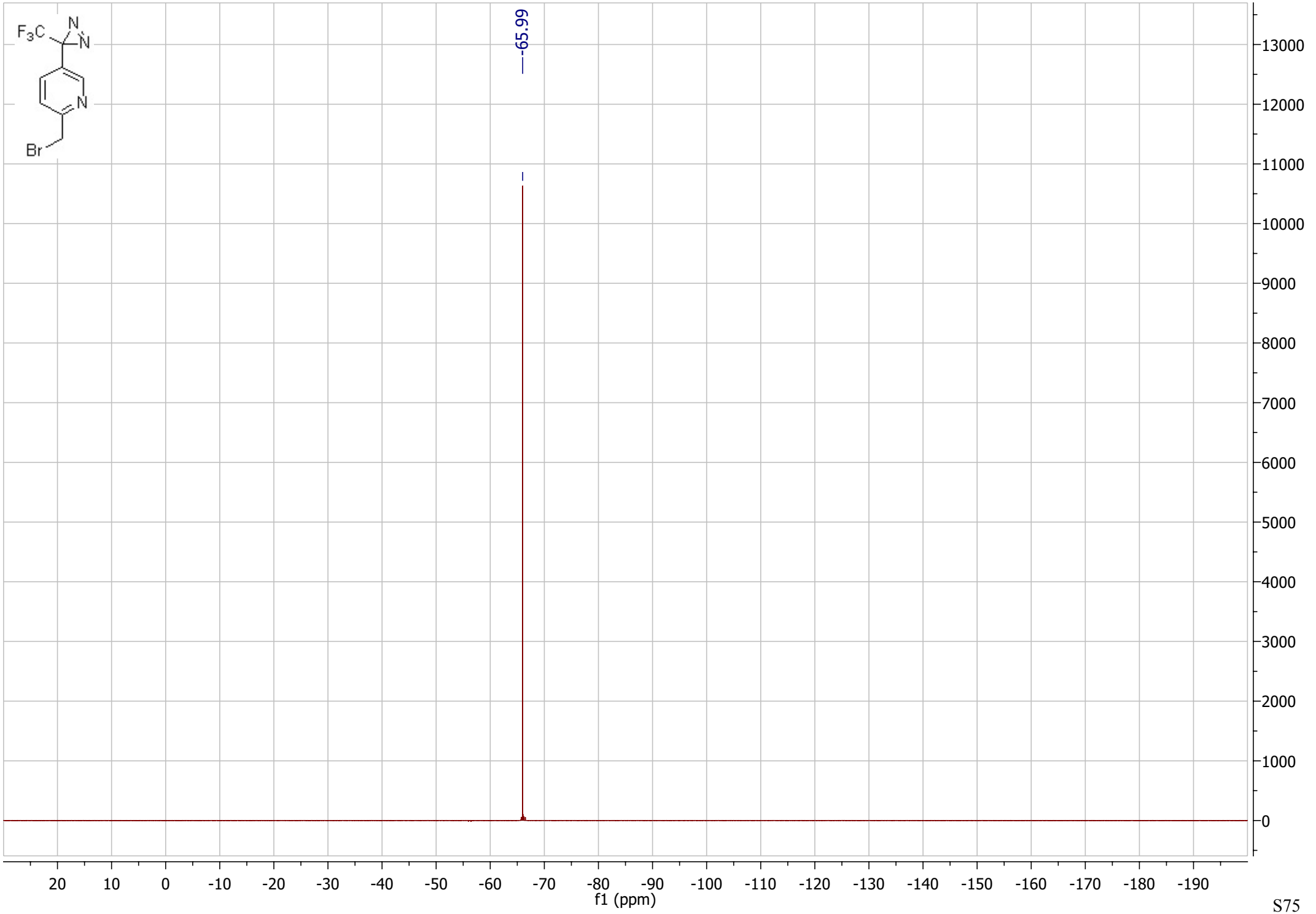

OAc
OAc
AcO
AcO
O
O
HO
O
172.85
172.45
171.58
171.51
99.16
70.67
70.46
70.40
67.12
65.33
63.48
20.68
20.64
20.60
5000
4500
4000
3500
3000
2500
2000
1500
1000
500
0
-500
220
210
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

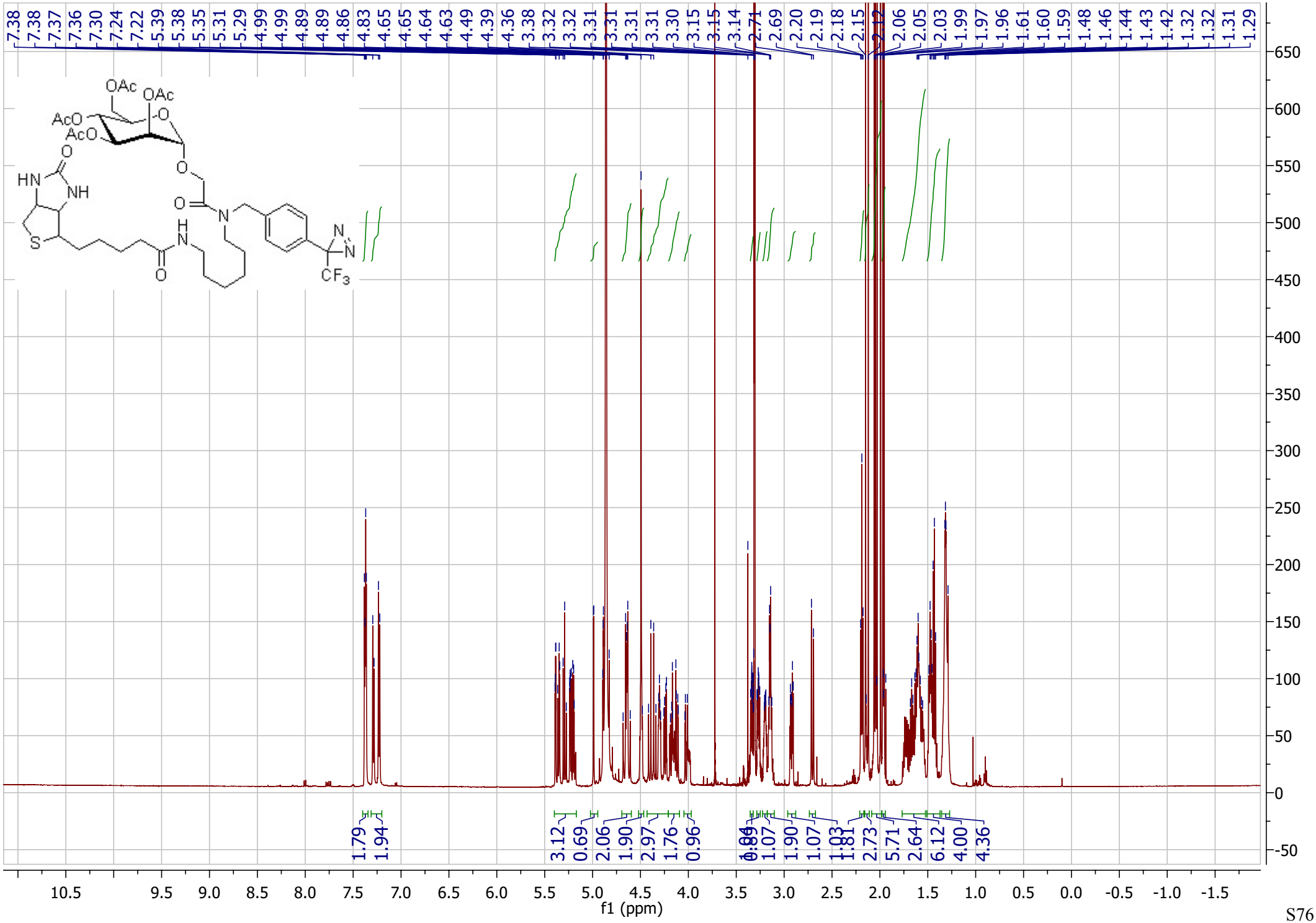

F3C
N
N
Br
139.72
129.44
128.98
126.83
126.82
123.41
120.68
32.15
28.45
28.05
8500
8000
7500
7000
6500
6000
5500
5000
4500
4000
3500
3000
2500
2000
1500
1000
500
0
-500
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
f1 (ppm)

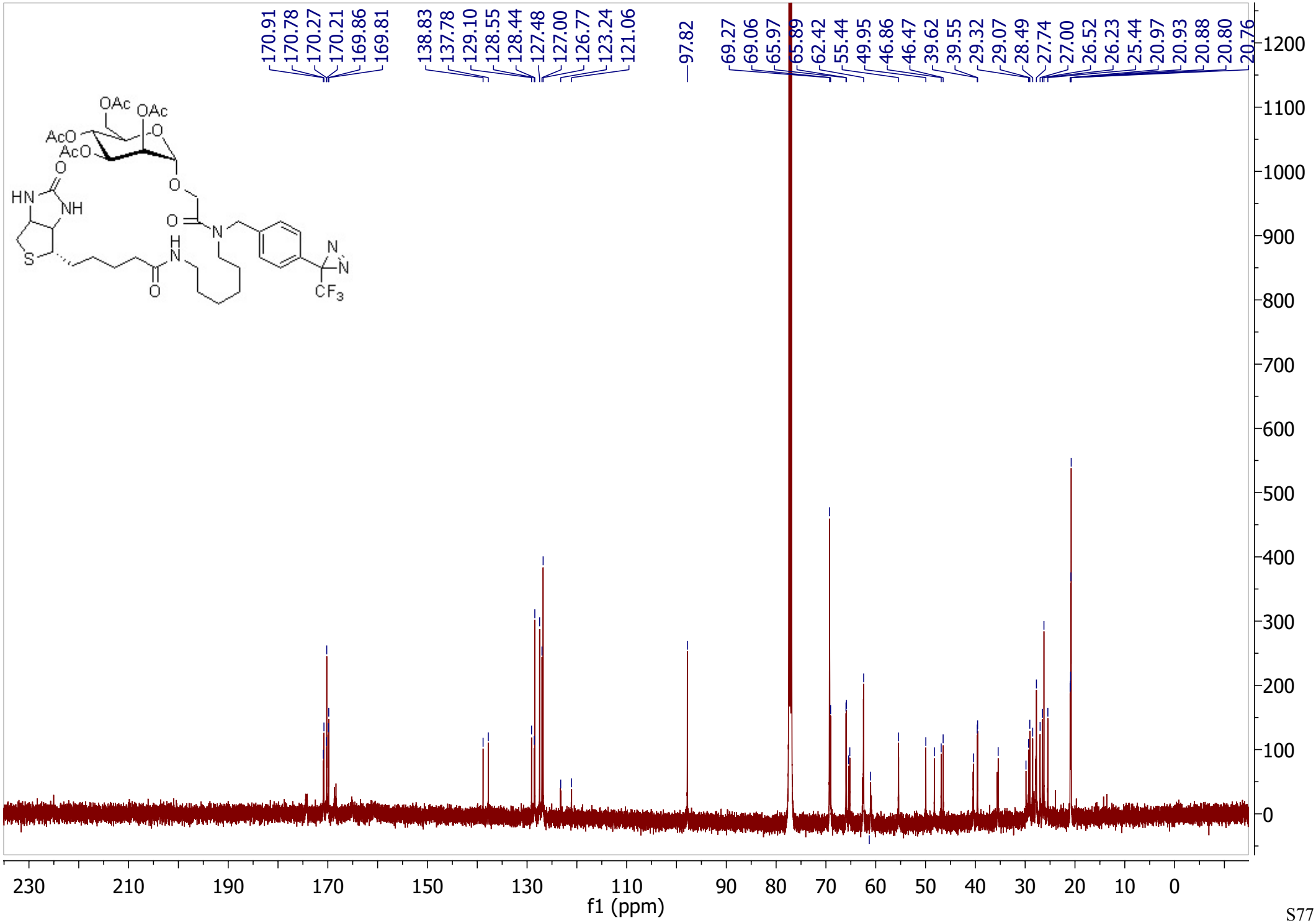

F3C
N
N
Br
-65.67
4000
3500
3000
2500
2000
1500
1000
500
0
20
10
0
-10
-20
-30
-40
-50
-60
-70
-80
-90
-100
-110
-120
-130
-140
-150
-160
-170
-180
-190
f1 (ppm)

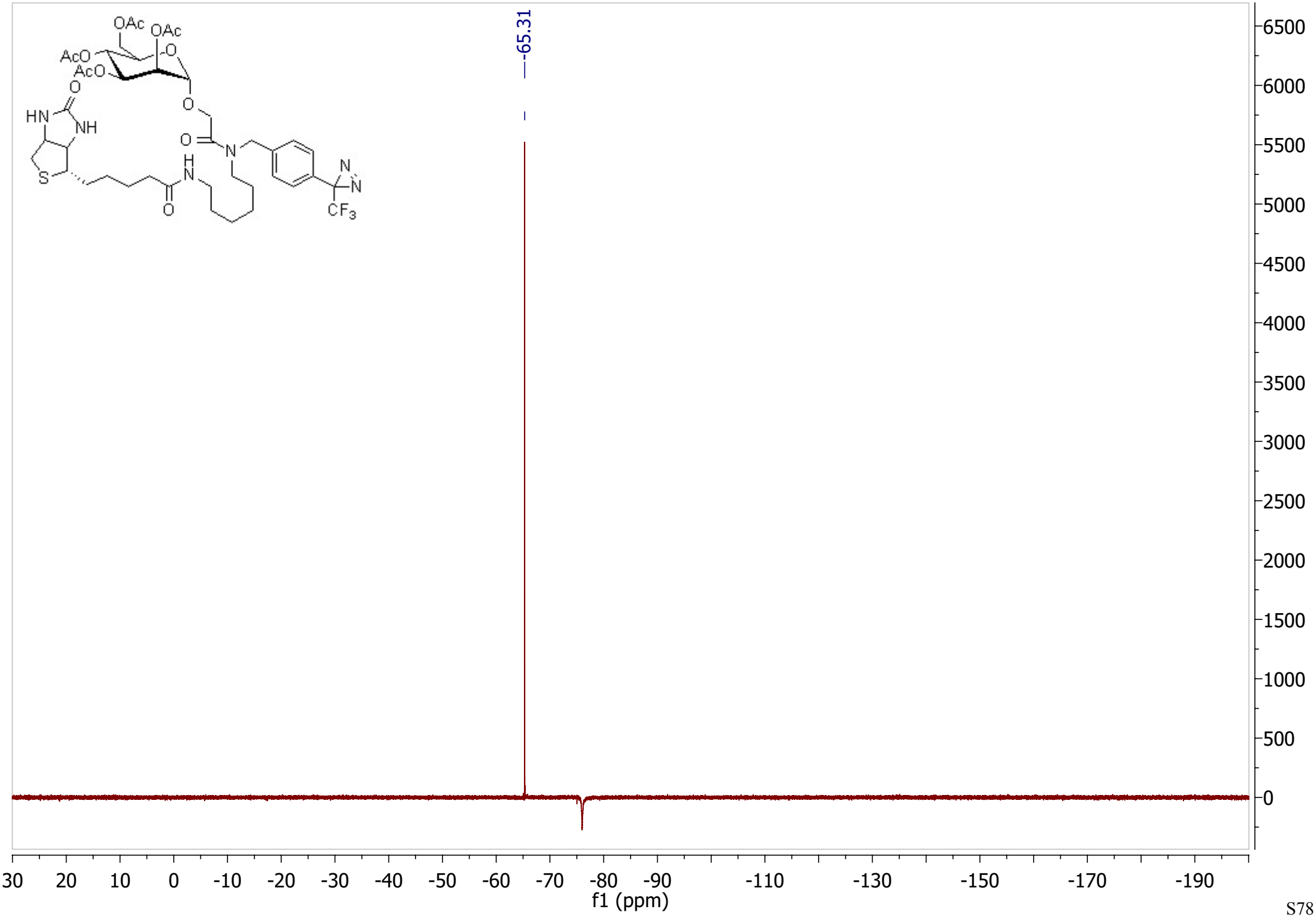

8.44
8.43
7.58
7.58
7.56
7.56
7.50
7.48
4.55
0.81
0.96
0.98
2.00
f1 (ppm)

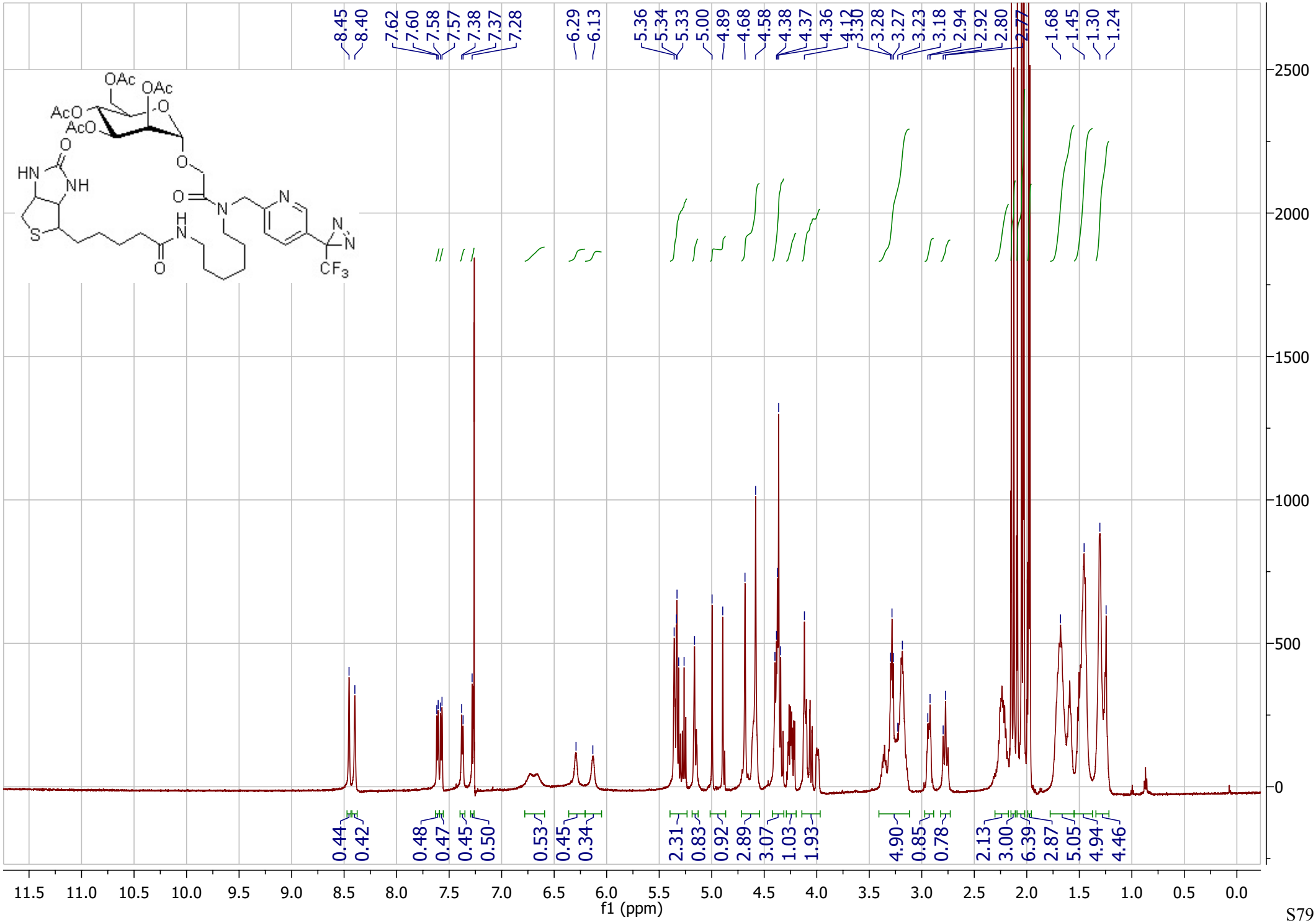

159.04
148.25
148.23
135.95
135.94
126.44
125.06
123.71
120.98
118.25
33.55
27.89
27.47
f1 (ppm)

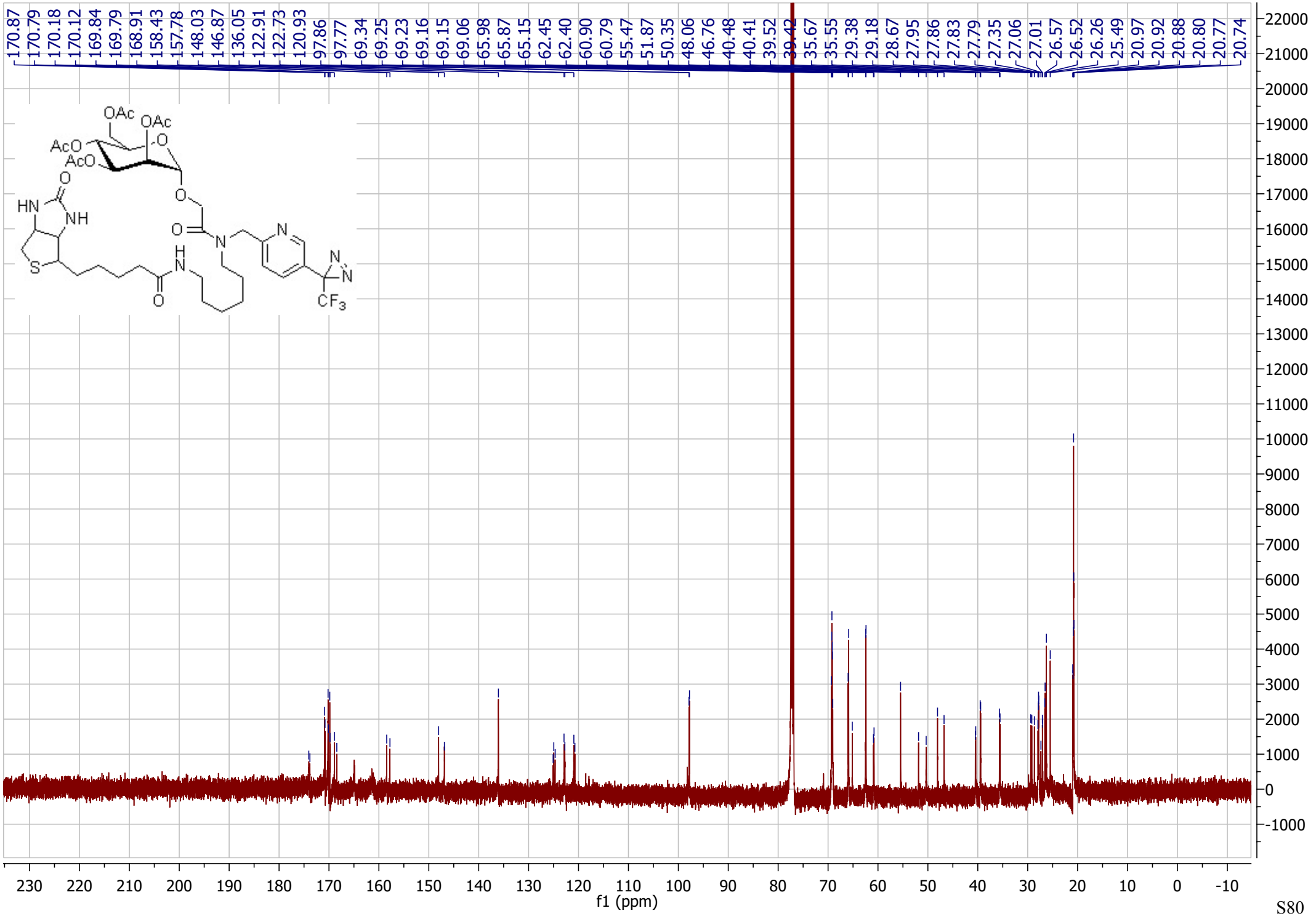

F3C
N
N
N
Br
-65.99
13000
12000
11000
10000
9000
8000
7000
6000
5000
4000
3000
2000
1000
0
20
10
0
-10
-20
-30
-40
-50
-60
-70
-80
-90
-100
-110
-120
-130
-140
-150
-160
-170
-180
-190
f1 (ppm)

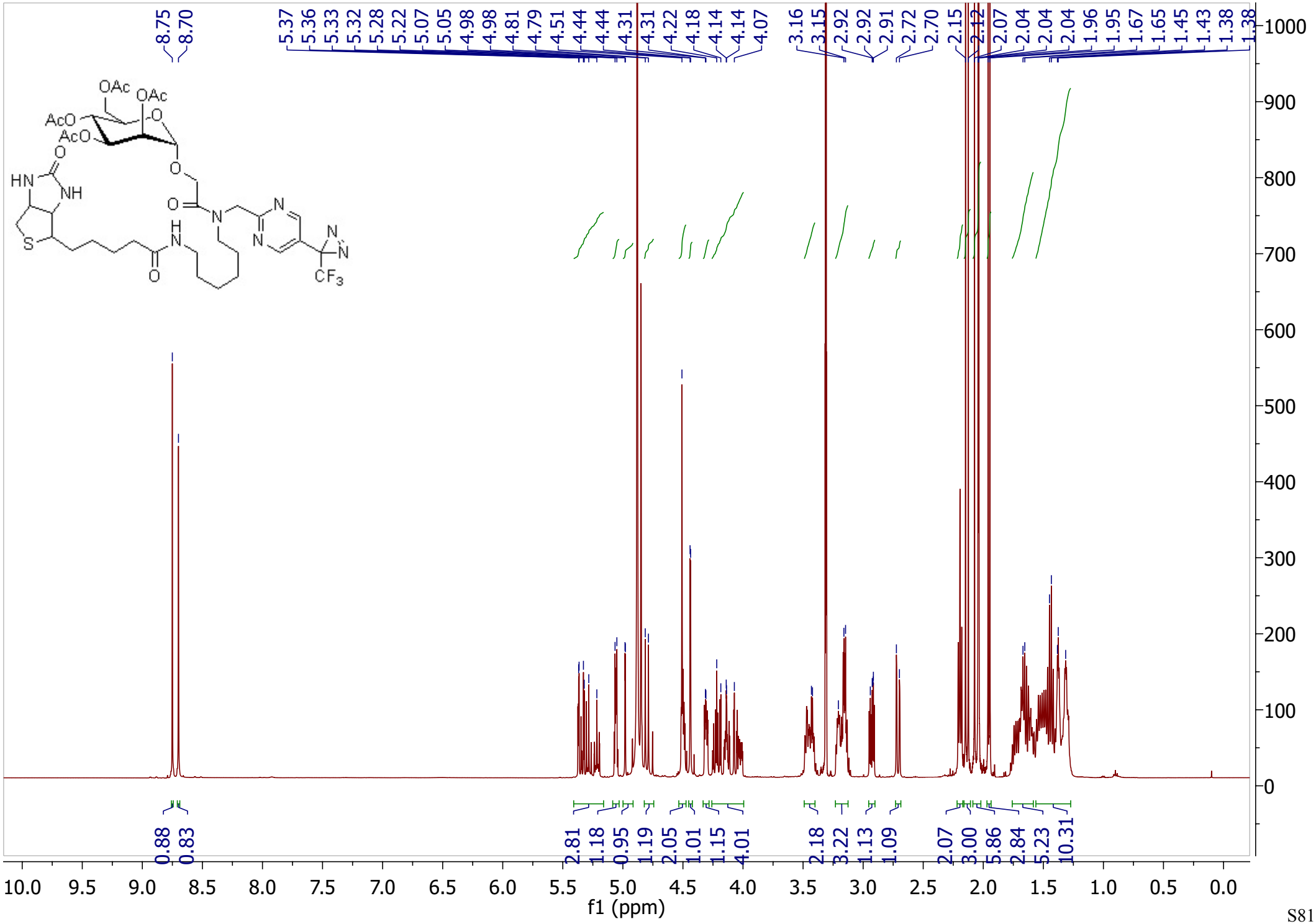

OAc
OAc
AcO
AcO
O
O
HN
NH
O
O
N
H
N
O
S
N
N
CF3
f1 (ppm)

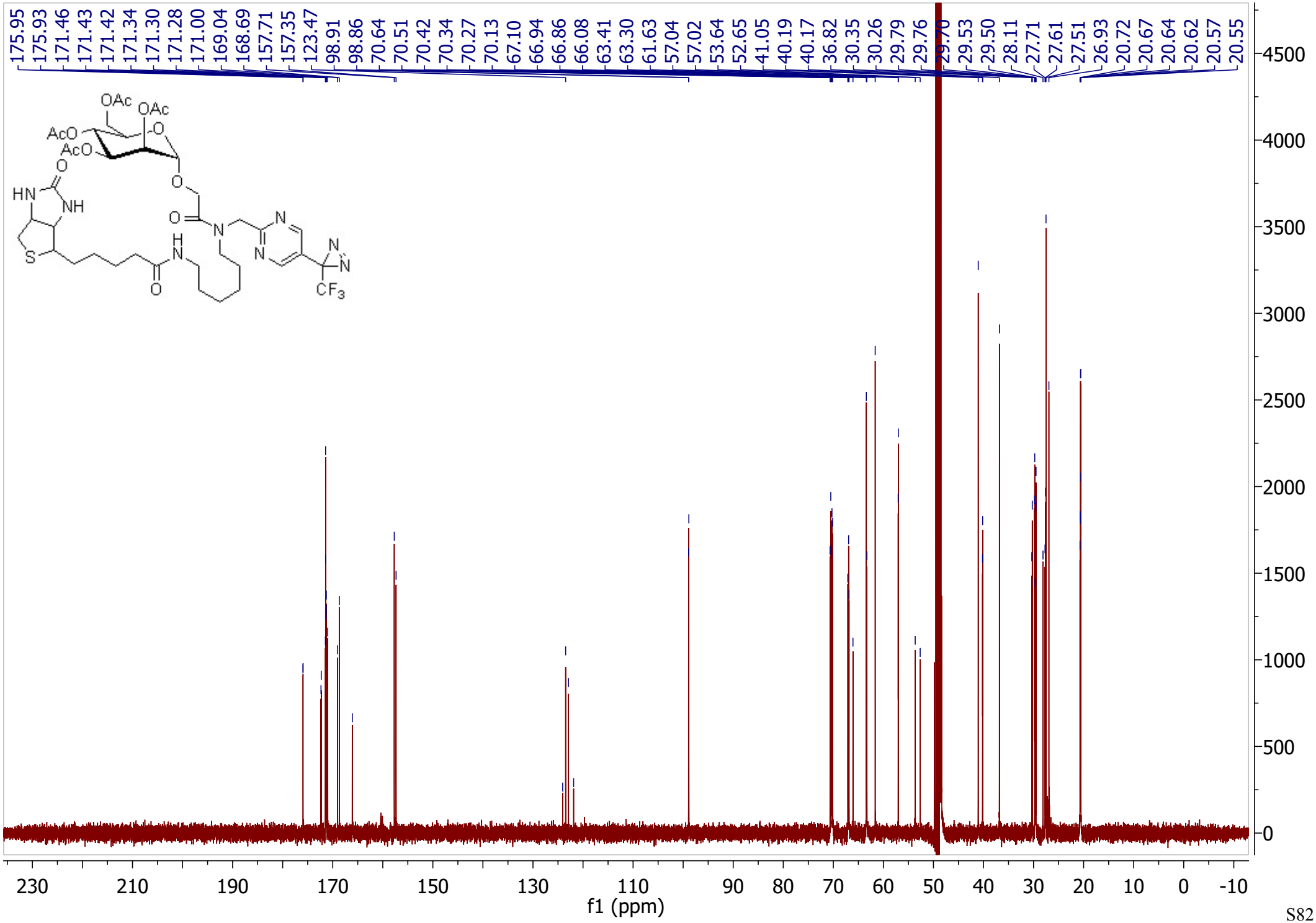

170.91
170.78
170.27
170.21
169.86
169.81
138.83
137.78
129.10
128.55
128.44
127.48
127.00
126.77
123.24
121.06
97.82
69.27
69.06
65.97
65.89
62.42
55.44
49.95
46.86
46.47
39.62
39.55
29.32
29.07
28.49
27.74
27.00
26.52
26.23
25.44
20.97
20.93
20.88
20.80
20.76
OAc
OAc
AcO
AcO
O
HN
NH
S
O
O
N
H
N
O
N
N
CF3
1200
1100
1000
900
800
700
600
500
400
300
200
100
0
230
210
190
170
150
130
110
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

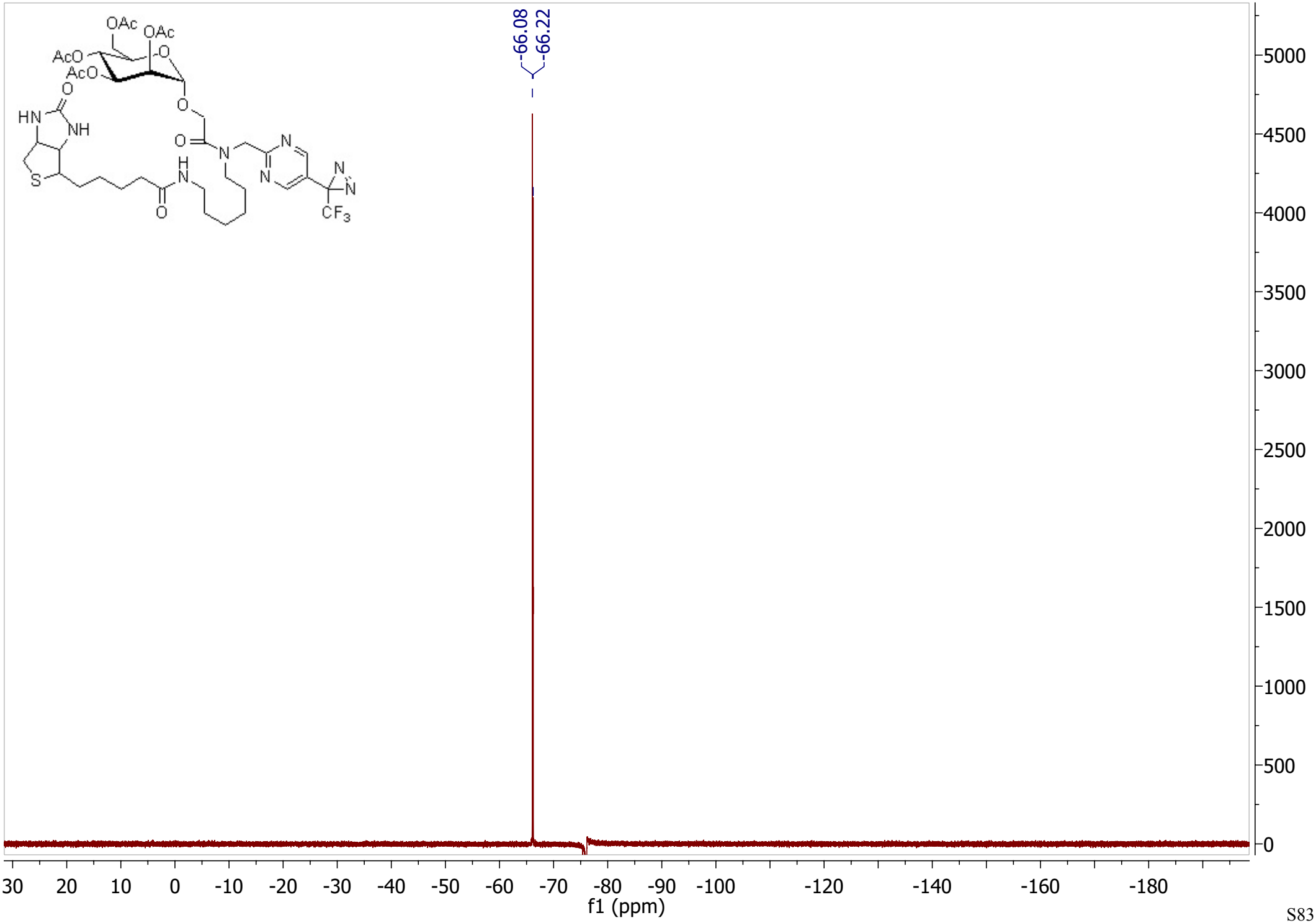

-65.31
OAc
OAc
AcO
AcO
O
HN
NH
O
S
O
O
N
H
N
O
N
N
CF3
6500
6000
5500
5000
4500
4000
3500
3000
2500
2000
1500
1000
500
0
30
20
10
0
-10
-20
-30
-40
-50
-60
-70
-80
-90
-110
-130
-150
-170
-190
f1 (ppm)

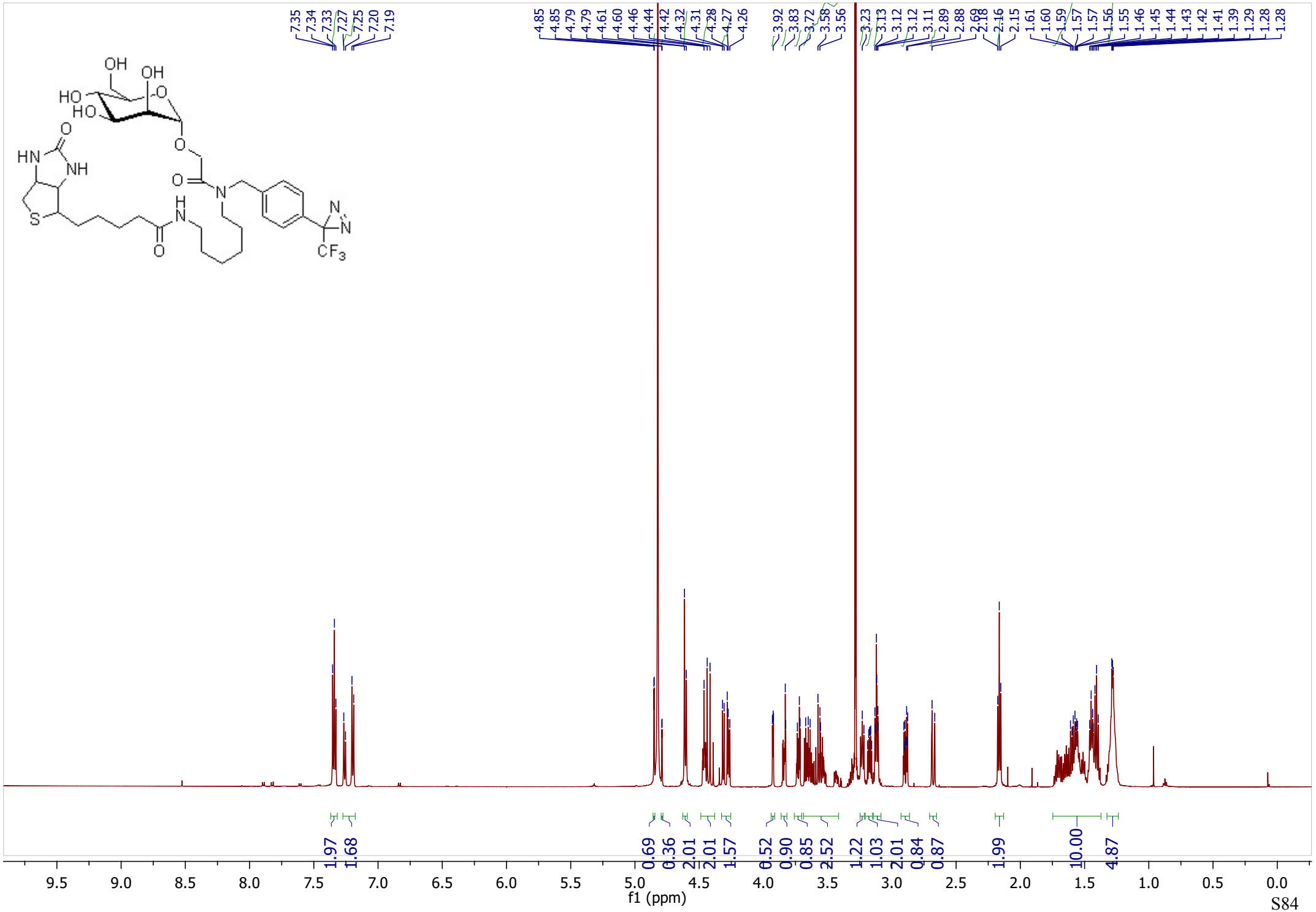

8.45
8.40
7.62
7.60
7.58
7.57
7.38
7.37
7.28
6.29
6.13
5.36
5.34
5.33
5.00
4.89
4.68
4.58
4.38
4.37
4.36
4.12
4.10
3.30
3.28
3.27
3.23
3.18
2.94
2.92
2.80
2.77
1.68
1.45
1.30
1.24
OAc
OAc
AcO
AcO
O
HN
NH
S
O
O
O
N
N
N
N
CF3
2500
2000
1500
1000
500
0
0.44
0.42
0.48
0.47
0.45
0.50
0.53
0.45
0.34
2.31
0.83
0.92
2.89
3.07
1.03
1.93
4.90
0.85
0.78
2.13
3.00
6.39
2.87
5.05
4.94
4.46
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
f1 (ppm)

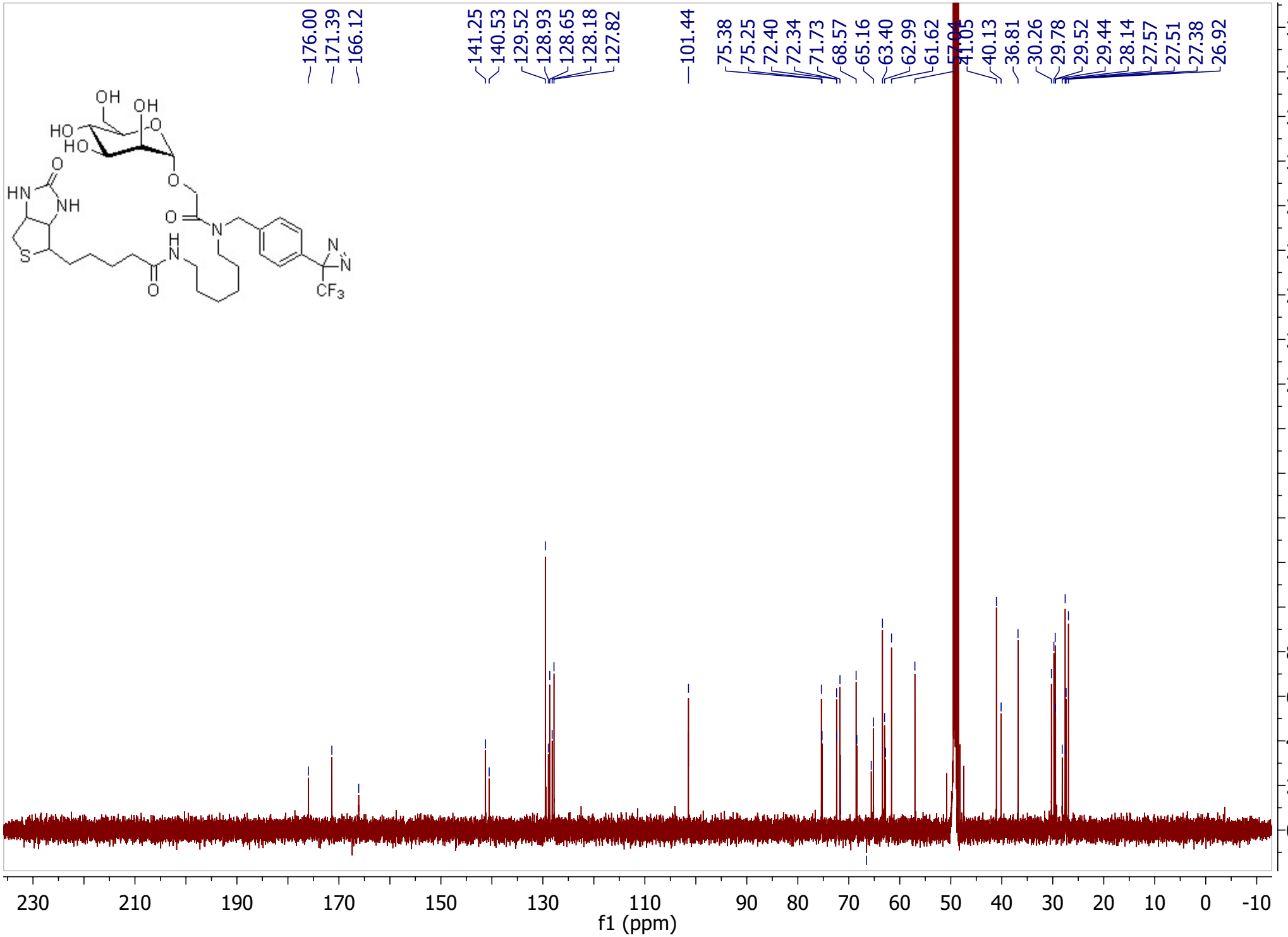

170.87
170.79
170.18
170.12
169.84
169.79
168.91
158.43
157.78
148.03
146.87
136.05
122.91
122.73
120.93
97.86
97.77
69.34
69.25
69.23
69.16
69.15
69.06
65.98
65.87
65.15
62.45
62.40
60.90
60.79
55.47
51.87
50.35
48.06
46.76
40.48
40.41
39.52
39.42
35.67
35.55
29.38
29.18
28.67
27.95
27.86
27.83
27.79
27.35
27.06
27.01
26.57
26.52
26.26
25.49
20.97
20.92
20.88
20.80
20.77
20.74
OAc
OAc
AcO
AcO
O
HN
NH
O
O
O
N
N
H
N
S
O
N
N
CF3
22000
21000
20000
19000
18000
17000
16000
15000
14000
13000
12000
11000
10000
9000
8000
7000
6000
5000
4000
3000
2000
1000
0
-1000
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)

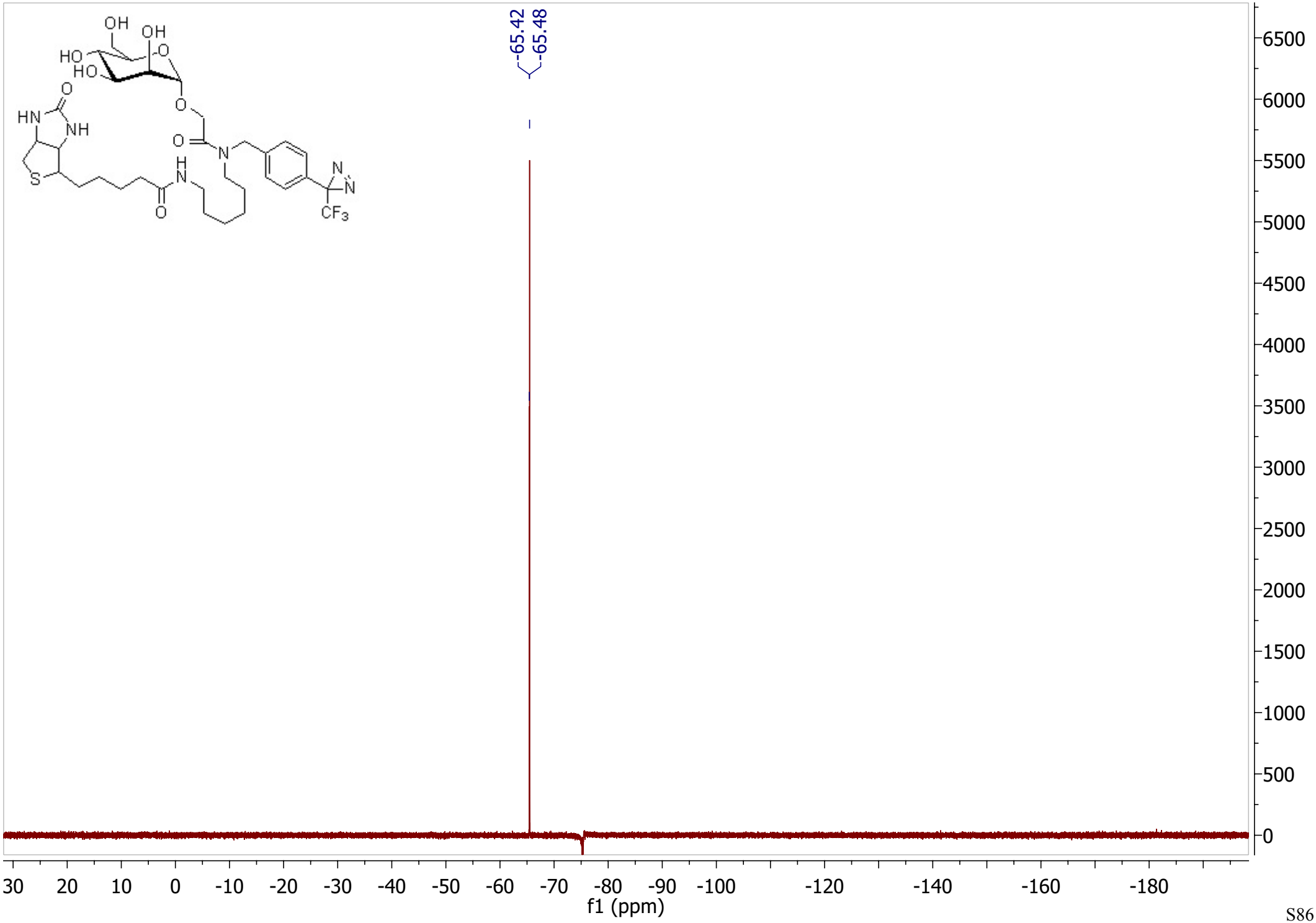

8.75
8.70
5.37
5.36
5.33
5.32
5.28
5.22
5.07
5.05
4.98
4.98
4.81
4.79
4.51
4.44
4.44
4.31
4.31
4.22
4.18
4.14
4.14
4.07
3.16
3.15
2.92
2.92
2.91
2.72
2.70
2.15
2.12
2.07
2.04
2.04
2.04
1.96
1.95
1.67
1.65
1.45
1.43
1.38
1.38
OAc
OAc
AcO
AcO
O
HN
NH
O
O
N
N
N
N
H
N
O
S
CF3
0.88
0.83
2.81
1.18
0.95
1.19
2.05
1.01
1.15
4.01
2.18
3.22
1.13
1.09
2.07
3.00
5.86
2.84
5.23
10.31
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
f1 (ppm)
1000
900
800
700
600
500
400
300
200
100
0

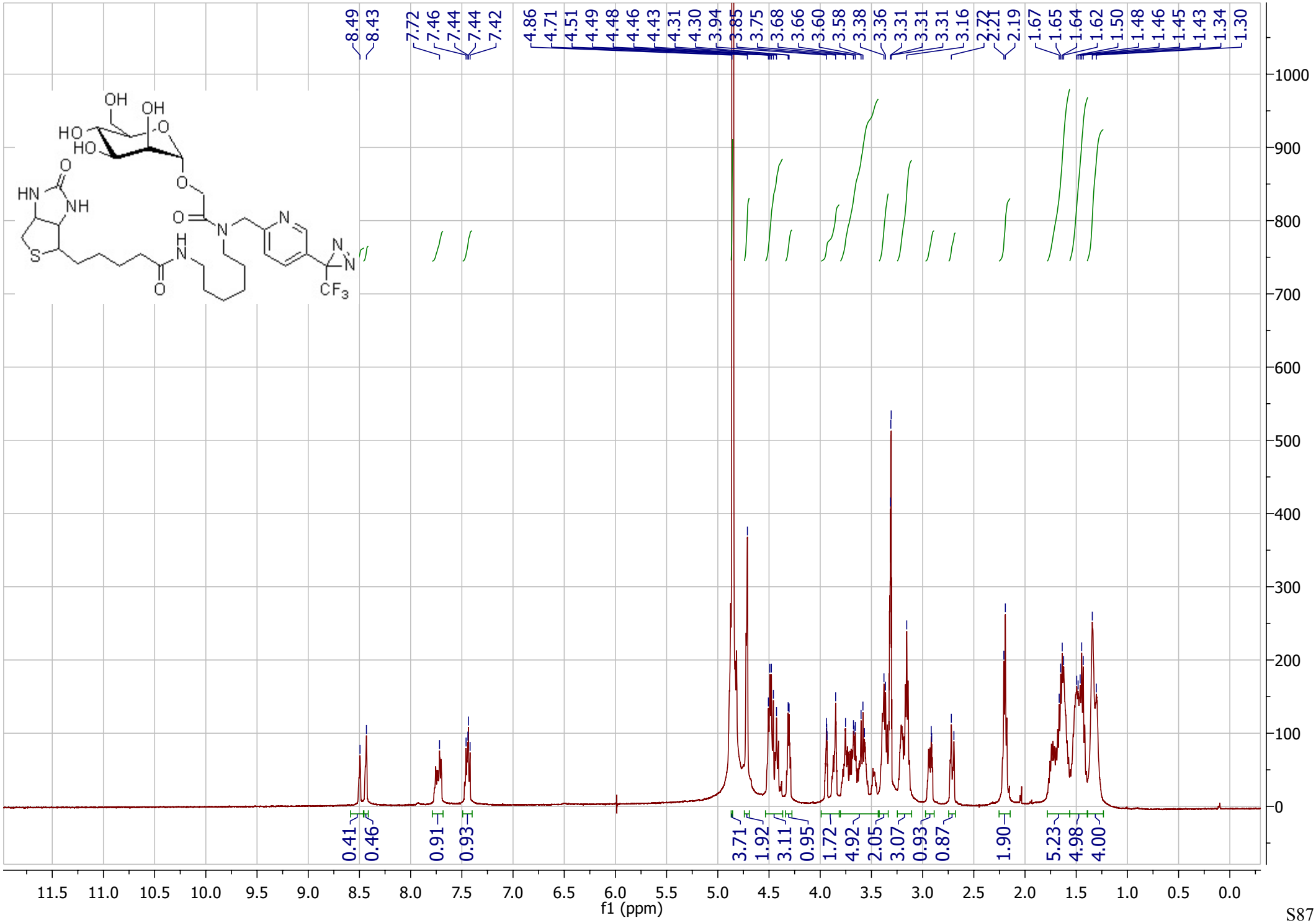

175.95
175.93
171.46
171.43
171.42
171.34
171.30
171.28
171.00
169.04
168.69
157.71
157.35
123.47
98.91
98.86
70.64
70.51
70.42
70.34
70.27
70.13
67.10
66.94
66.86
66.08
63.41
63.30
61.63
57.04
57.02
53.64
52.65
41.05
40.19
40.17
36.82
30.35
30.26
29.79
29.76
29.70
29.53
29.50
28.11
27.71
27.61
27.51
26.93
20.72
20.67
20.64
20.62
20.57
20.55
OAc
OAc
AcO
AcO
O
HN
NH
S
O
O
N
N
N
N
N
CF3
H
O
4500
4000
3500
3000
2500
2000
1500
1000
500
0
230
210
190
170
150
130
110
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)

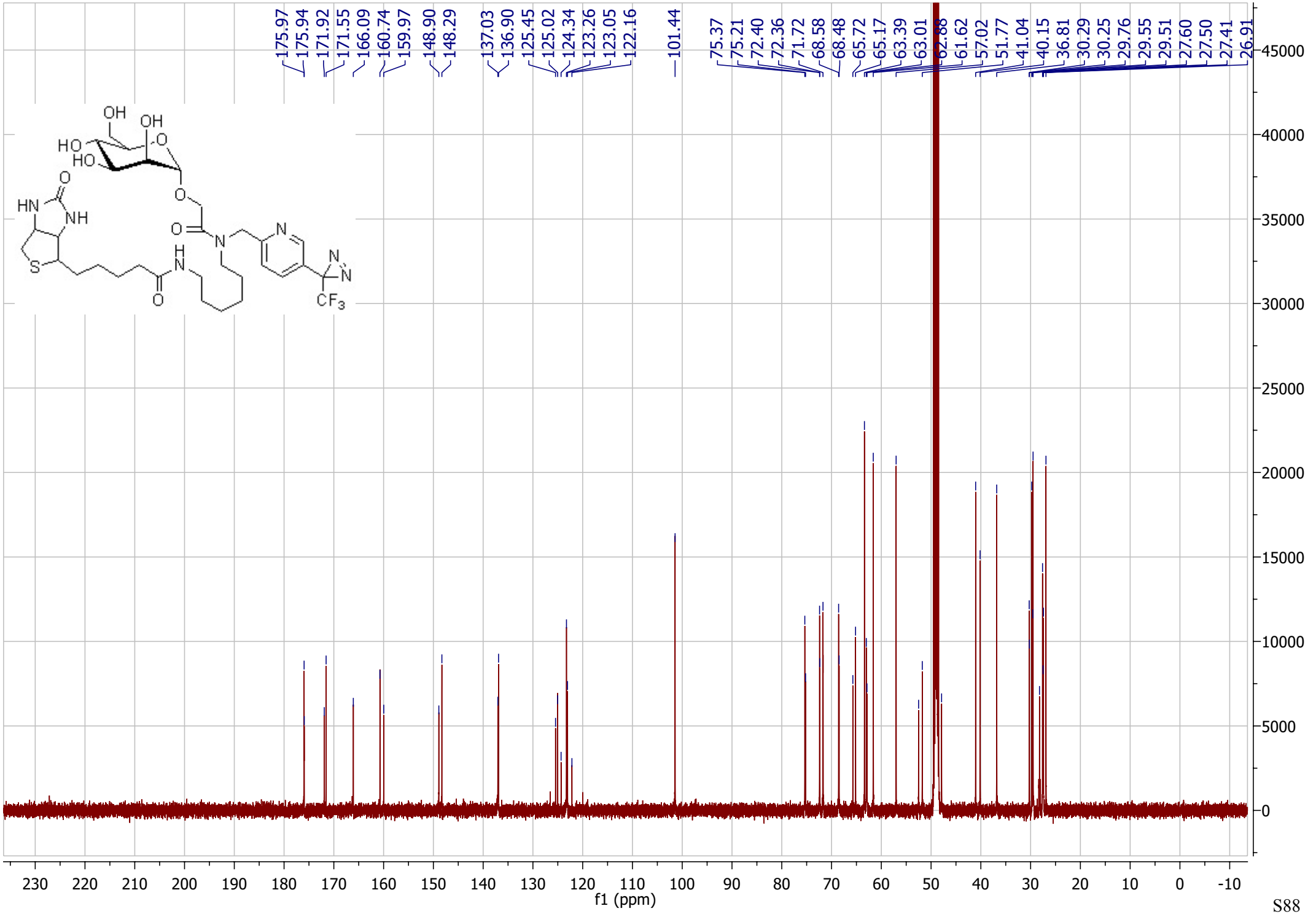

-66.08
-66.22
OAc
OAc
AcO
AcO
O
HN
NH
O
S
O
O
H
N
O
N
N
N
N
CF3
5000
4500
4000
3500
3000
2500
2000
1500
1000
500
0
30
20
10
0
-10
-20
-30
-40
-50
-60
-70
-80
-90
-100
-120
-140
-160
-180
f1 (ppm)


7.35 7.34 7.33 7.27 7.25 7.20 7.19
4.85 4.85 4.79 4.79 4.61 4.60 4.46 4.44 4.42 4.32 4.31 4.28 4.27 4.26
3.92 3.83 3.72 3.58 3.56
3.23 3.13 3.12 3.12 3.11 2.89 2.88 2.68 2.18 2.16 2.15
1.61 1.60 1.59 1.57 1.57 1.56 1.55 1.46 1.45 1.44 1.43 1.42 1.41 1.39 1.29 1.28 1.28
OH OH HO HO O O HN NH O O N H N S O N N CF3
1.97 1.68 0.69 0.36 2.01 2.01 1.57 0.52 0.90 0.85 2.52 1.22 1.03 2.01 0.84 0.87 1.99 10.00 4.87
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
f1 (ppm)


176.00
171.39
166.12
141.25
140.53
129.52
128.93
128.65
128.18
127.82
101.44
75.38
75.25
72.40
72.34
71.73
68.57
65.16
63.40
62.99
61.62
57.04
41.05
40.13
36.81
30.26
29.78
29.52
29.44
28.14
27.57
27.51
27.38
26.92
OH
OH
HO
HO
O
O
HN
NH
O
O
N
H
N
S
O
N
N
CF3
3600
3400
3200
3000
2800
2600
2400
2200
2000
1800
1600
1400
1200
1000
800
600
400
200
0
230
210
190
170
150
130
110
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)


-65.42
-65.48
6500
6000
5500
5000
4500
4000
3500
3000
2500
2000
1500
1000
500
0
30
20
10
0
-10
-20
-30
-40
-50
-60
-70
-80
-90
-100
-120
-140
-160
-180
f1 (ppm)


8.49
8.43
7.72
7.46
7.44
7.44
7.42
4.86
4.71
4.51
4.49
4.48
4.46
4.43
4.31
4.30
3.94
3.85
3.75
3.68
3.66
3.60
3.58
3.38
3.36
3.31
3.31
3.31
3.16
2.22
2.21
2.19
1.67
1.65
1.64
1.62
1.50
1.48
1.46
1.45
1.43
1.34
1.30
0.41
0.46
0.91
0.93
3.71
1.92
3.11
0.95
1.72
4.92
2.05
3.07
0.93
0.87
1.90
5.23
4.98
4.00
f1 (ppm)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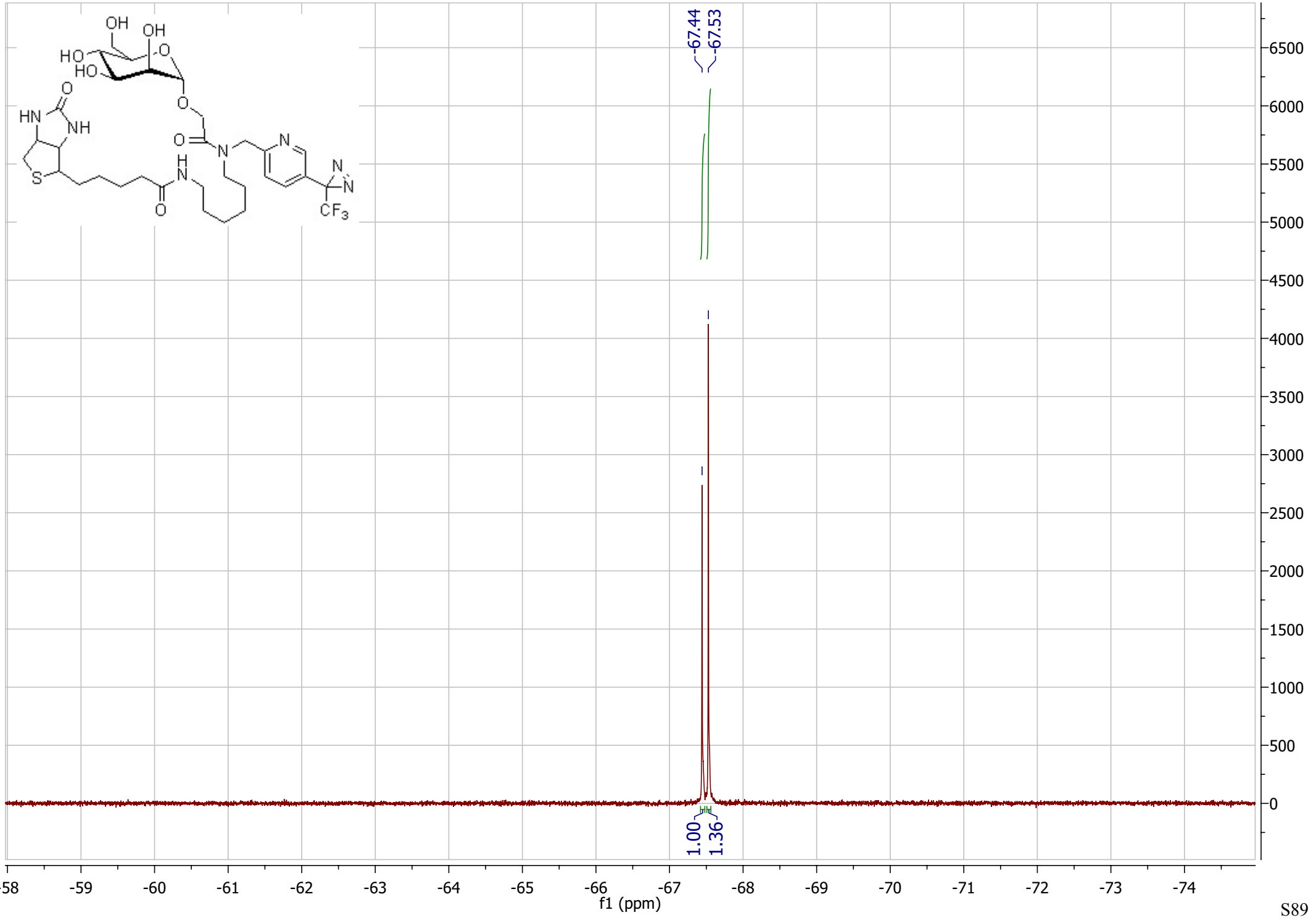
175.94
171.92
171.55
166.09
160.74
159.97
148.90
148.29
137.03
136.90
125.45
125.02
124.34
123.26
123.05
122.16
101.44
75.37
75.21
72.40
72.36
71.72
68.58
68.48
65.72
65.17
63.39
63.01
62.88
61.62
57.02
51.77
41.04
40.15
36.81
30.29
30.25
29.76
29.55
29.51
27.60
27.50
27.41
26.91
OH
OH
HO
HO
O
O
HN
NH
O
O
N
N
H
N
S
O
N
N
CF3
45000
40000
35000
30000
25000
20000
15000
10000
5000
0
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)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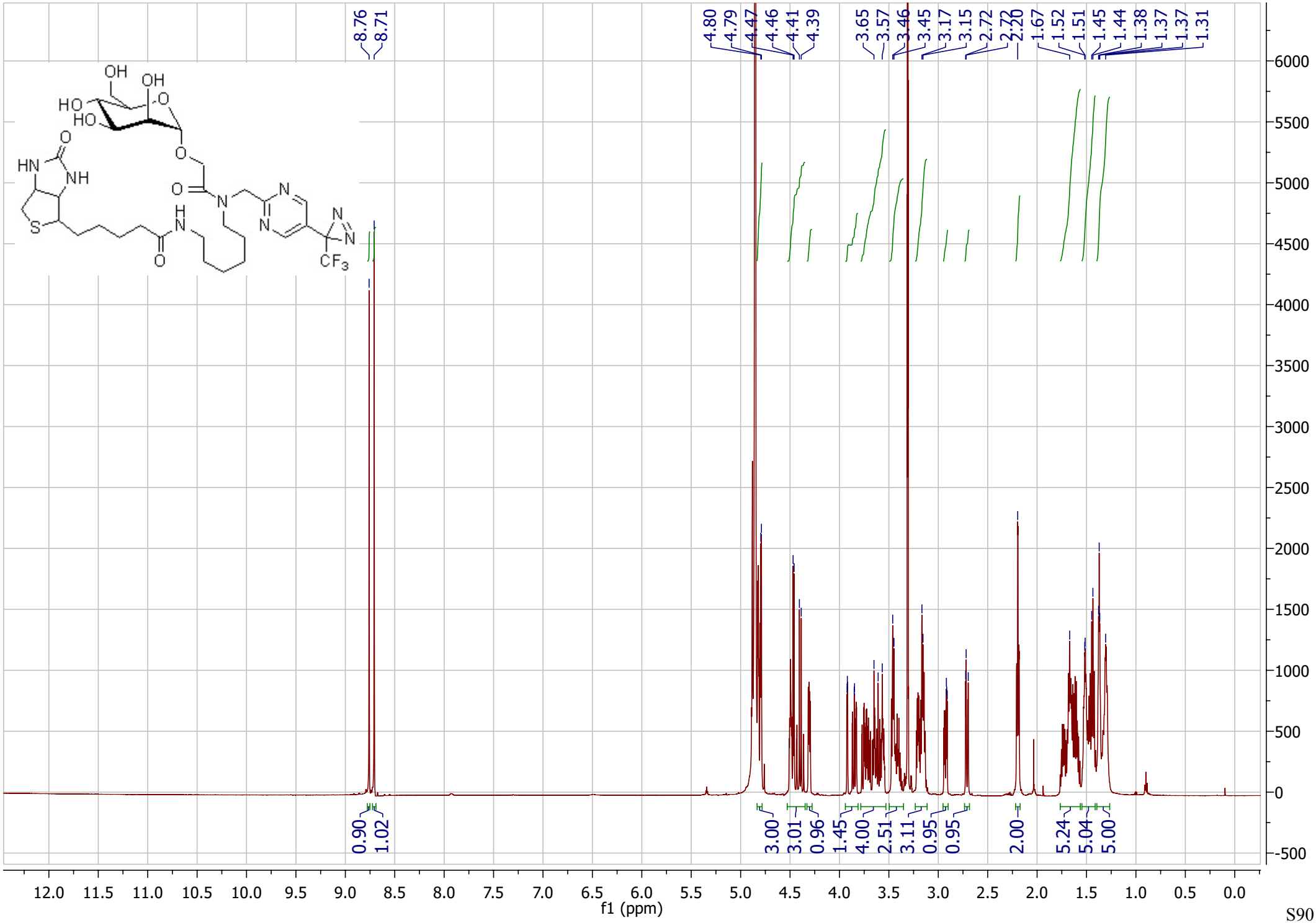
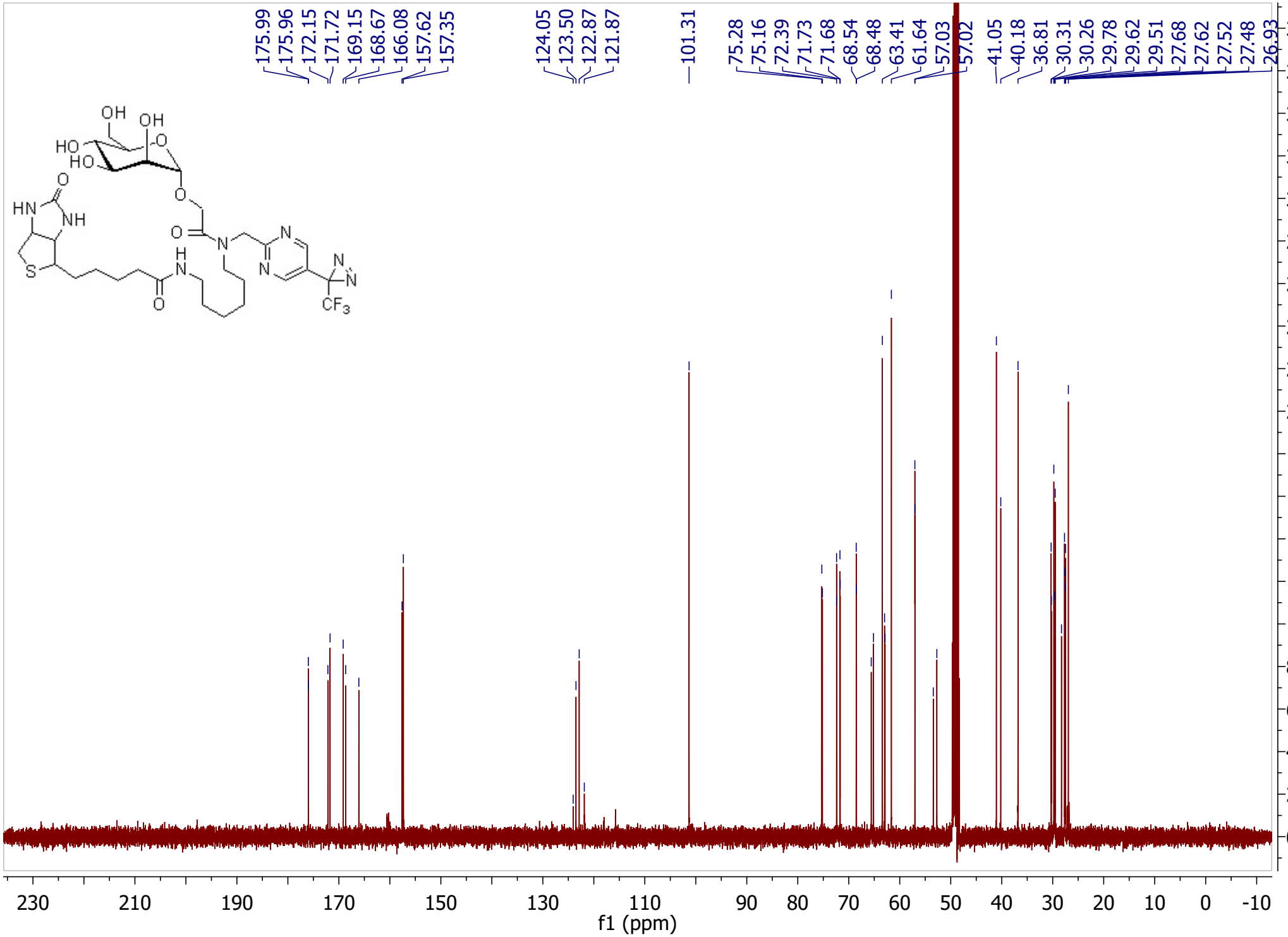
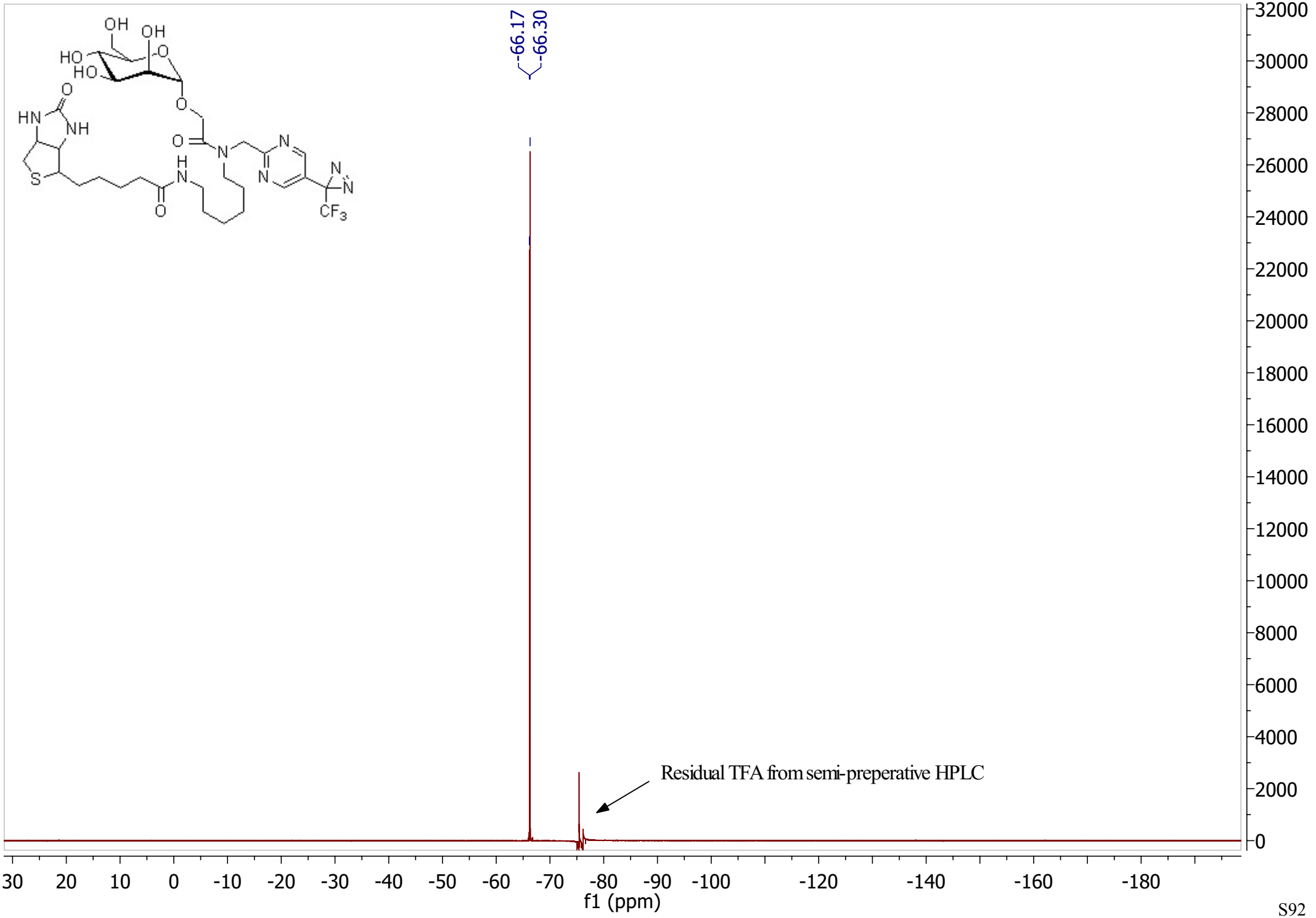
-67.53
1.00
1.36
f1 (ppm)


8.76
8.71
4.80
4.79
4.47
4.46
4.41
4.39
3.65
3.57
3.46
3.45
3.17
3.15
2.72
2.72
2.20
1.67
1.52
1.51
1.45
1.44
1.38
1.37
1.37
1.31
0.90
1.02
3.00
3.01
0.96
1.45
4.00
2.51
3.11
0.95
0.95
2.00
5.24
5.04
5.00
f1 (ppm)


175.99
175.96
172.15
171.72
169.15
168.67
166.08
157.62
157.35
124.05
123.50
122.87
121.87
101.31
75.28
75.16
72.39
71.73
71.68
68.54
68.48
63.41
61.64
57.03
57.02
41.05
40.18
36.81
30.31
30.26
29.78
29.62
29.51
27.68
27.62
27.52
27.48
26.93
OH
OH
HO
HO
O
O
HN
NH
O
O
N
N
N
H
N
S
O
N
N
CF3
3800
3600
3400
3200
3000
2800
2600
2400
2200
2000
1800
1600
1400
1200
1000
800
600
400
200
0
230
210
190
170
150
130
110
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)


-66.17
-66.30
Residual TFA from semi-preperative HPLC
f1 (ppm)