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Supporting Information

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Two-Step Functionalization of Oligosaccharides Using Glycosyl Iodide and Trimethylene Oxide and Its Applications to Multivalent Glycoconjugates

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Supporting Information

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General Information

All reactions were conducted under a dried argon atmosphere. The anhydrous solvents (dichloromethane (DCM) 99.8%, benzene (PhH) 99.8%, Methanol (MeOH) 99.8%, *N,N*-dimethylformamide (DMF) 99.8% and pyridine (pyr.) 99.8%) were purchased from commercial sources without further purification. In order to maintain water content of the solvents under 15 ppm, the solvents were dried and stored under 4 Å molecular sieves according to literature procedure.¹ Trimethylsilyl iodide (TMSI, stabilized with copper) was stored at -20 °C under a desiccated Ar atmosphere. TMSI in good condition should be a colorless transparent liquid. All other solvents and reagents were purchased from commercial sources and used without further purification. All glassware utilized was oven-dried or flame-dried before use. Glass-backed TLC plates (Silica Gel 60 with a 254 nm fluorescent indicator) were used without further manipulation and stored with desiccant. TLC plates were visualized using a short-wave UV lamp, stained with an I₂-SiO₂ mixture, and/or by heating TLC plates that were dipped in a solution of ammonium molybdate/cerium (IV) sulfate or anisaldehyde/H₂SO₄/AcOH/EtOH. Flash column chromatography (FCC) was performed using a silica gel (32-63 µm) stationary phase with a variable mobile phase correlated with TLC mobility. NMR experiments were conducted on 400, 800 or 600 MHz instruments using CDCl₃ (99.9% D), D₂O (99.9% D) and methanol-d₄ (99.8% D) as the solvent. Chemical shifts were referenced to the appropriate deuterated solvent peak (7.26 ppm for CDCl₃; 4.79 ppm for D₂O; 3.31 ppm for methanol-d₄) and were reported in parts per million (ppm). Coupling constants of the coupled protons were averaged to match with each other. High resolution mass spectra were recorded using ESI-Orbitrap LC-MS with internal calibration. The microwave-assisted regioselective silyl exchange

technology (ReSET) reactions were conducted in sealed 10 mL microwave vessels in a commercial microwave reactor (CEM DiscoverTM) which was operated by the SynergyTM software. The reaction temperatures were monitored by the reactor's built-in infrared (IR) detector.

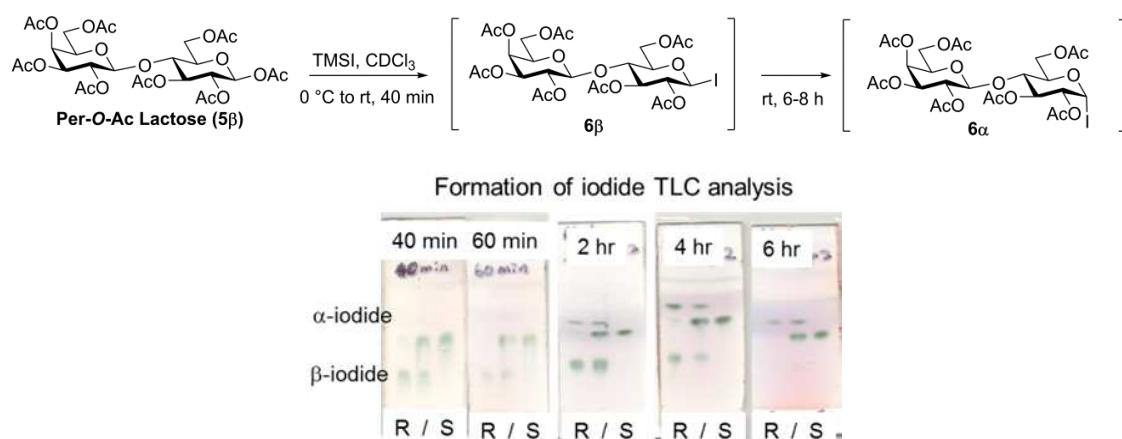


Figure S1. Per-*O*-Ac lactosyl iodide formation was stable enough to monitor by TLC

Synthesis, Purification and Characterization of Compound (25):

Table S1. Reaction mixture composition for synthesis of compound **25**

Compound	MW	Mass (mg)	mmol	Equiv.	Volume (mL)	Stock conc. (mM)	Final conc. (mM)
Trialkyne-PEG ₂ -PADRE (24)	1827.17	1.4	0.0008	1.0	0.5	1.6	0.33
Globotriaose azide (20)	587.53	4.2	0.0072	9.0	1.5	4.8	2.92
CuSO ₄ pentahydrate	249.53	0.1	0.0004	0.5	0.02	20	0.16
THPTA ligand	434.50	0.9	0.0021	2.6	0.04	50	0.85
Amino-guanidine	110.55	2.3	0.0208	26.0	0.2	100	8.5
Sodium Ascorbate	198.11	4.1	0.0208	26.0	0.2	100	8.5

HPLC condition: HPLC was carried out at 25 °C with semi-prep/analytic HPLC system and set detection at 280 nm. Product **25** eluted from column using mobile phase A:

MeCN and B: 0.1% TFA in H₂O. Analytic set-up involves: C₁₈-RP column (250 × 4.6 mm) with a flow rate of 1 mL/min.

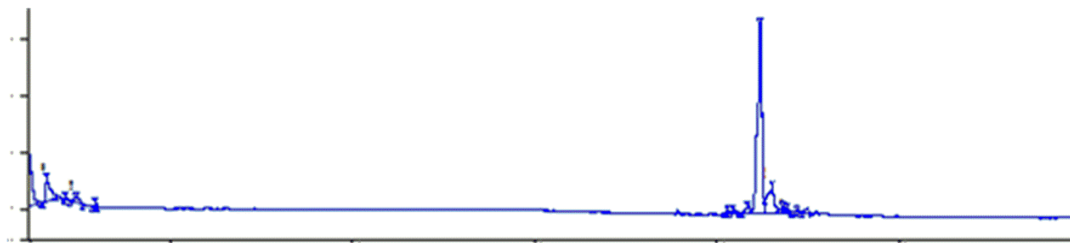


Figure S2. Analytical HPLC profile for compound **25**. $T_R = 24.9$ min ($\lambda = 280$ nm, with gradient of 5% A solution for 10 min, 5-40% A solution for 10 min, 40% A solution for 10 min, and 40-95% A solution for 5 min). A solution: MeCN, B solution: H₂O + 0.1% trifluoroacetic acid.

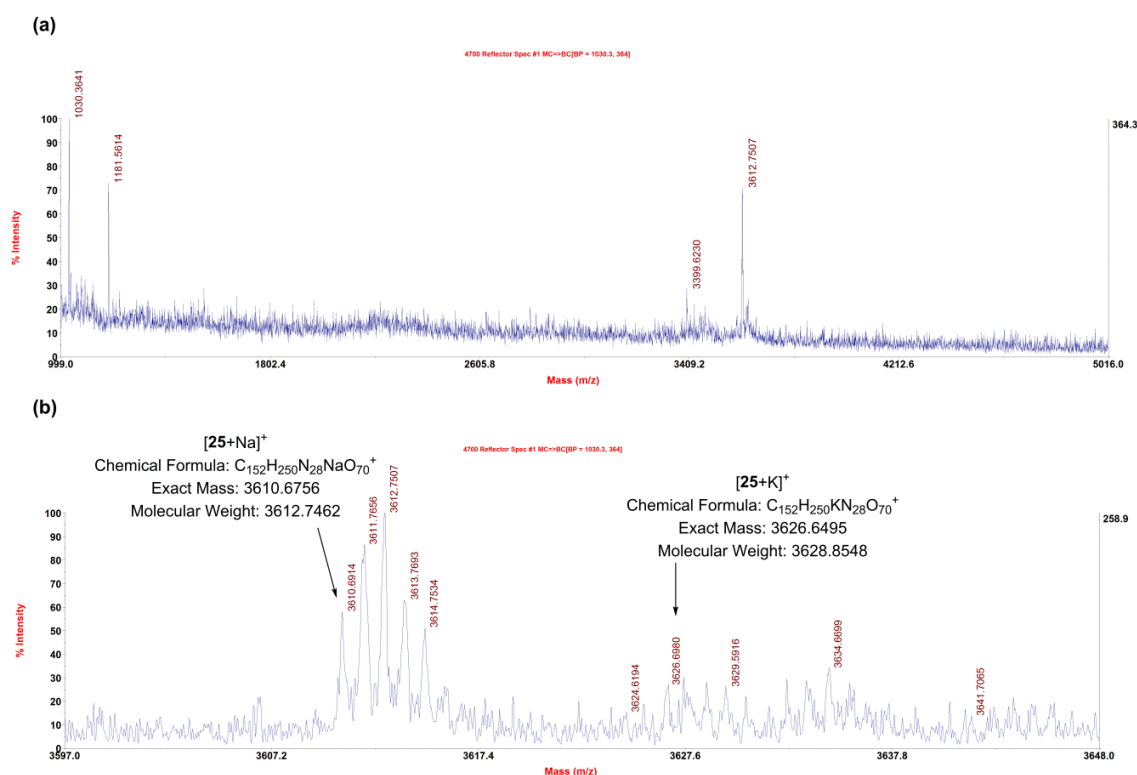
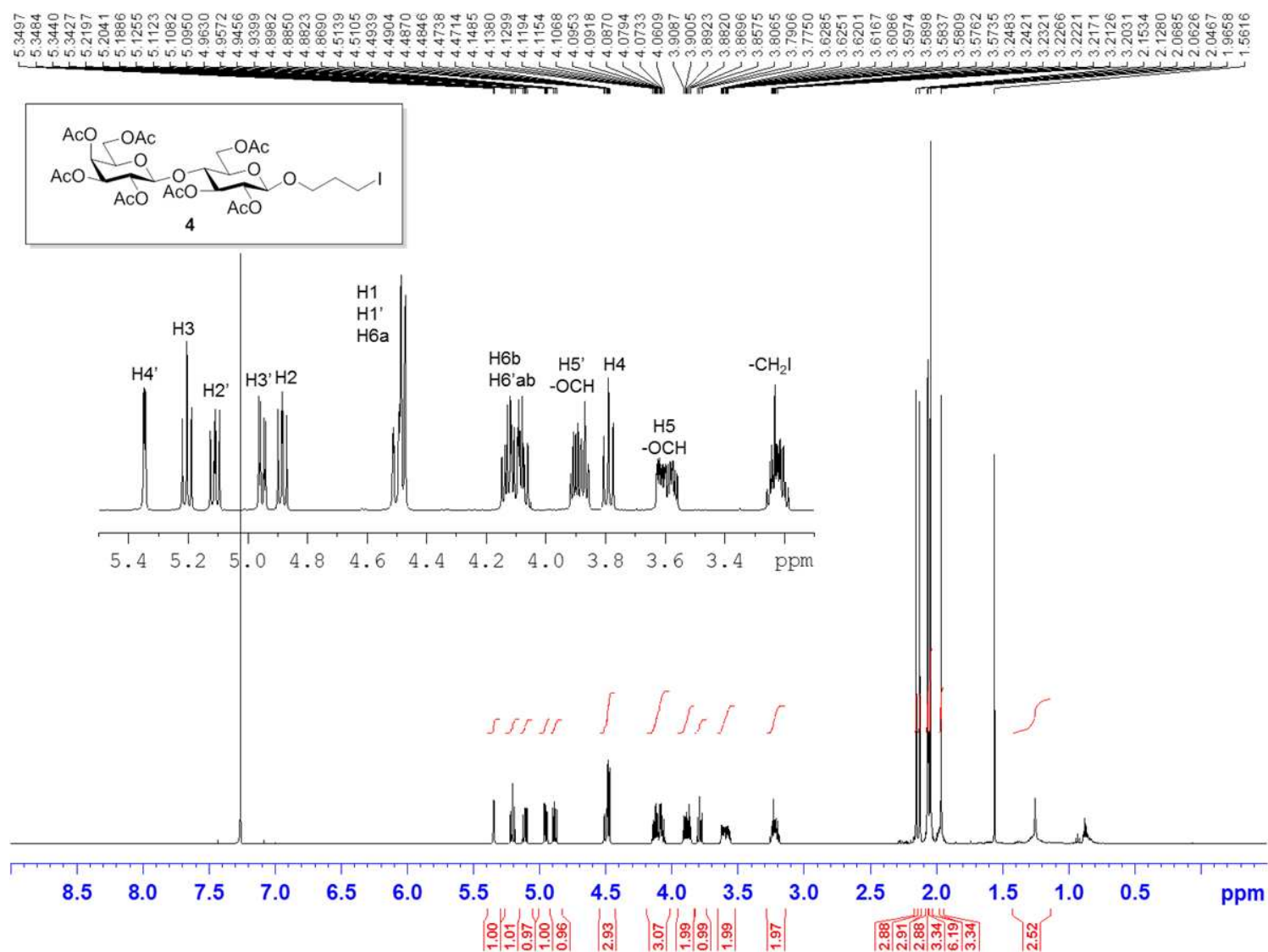
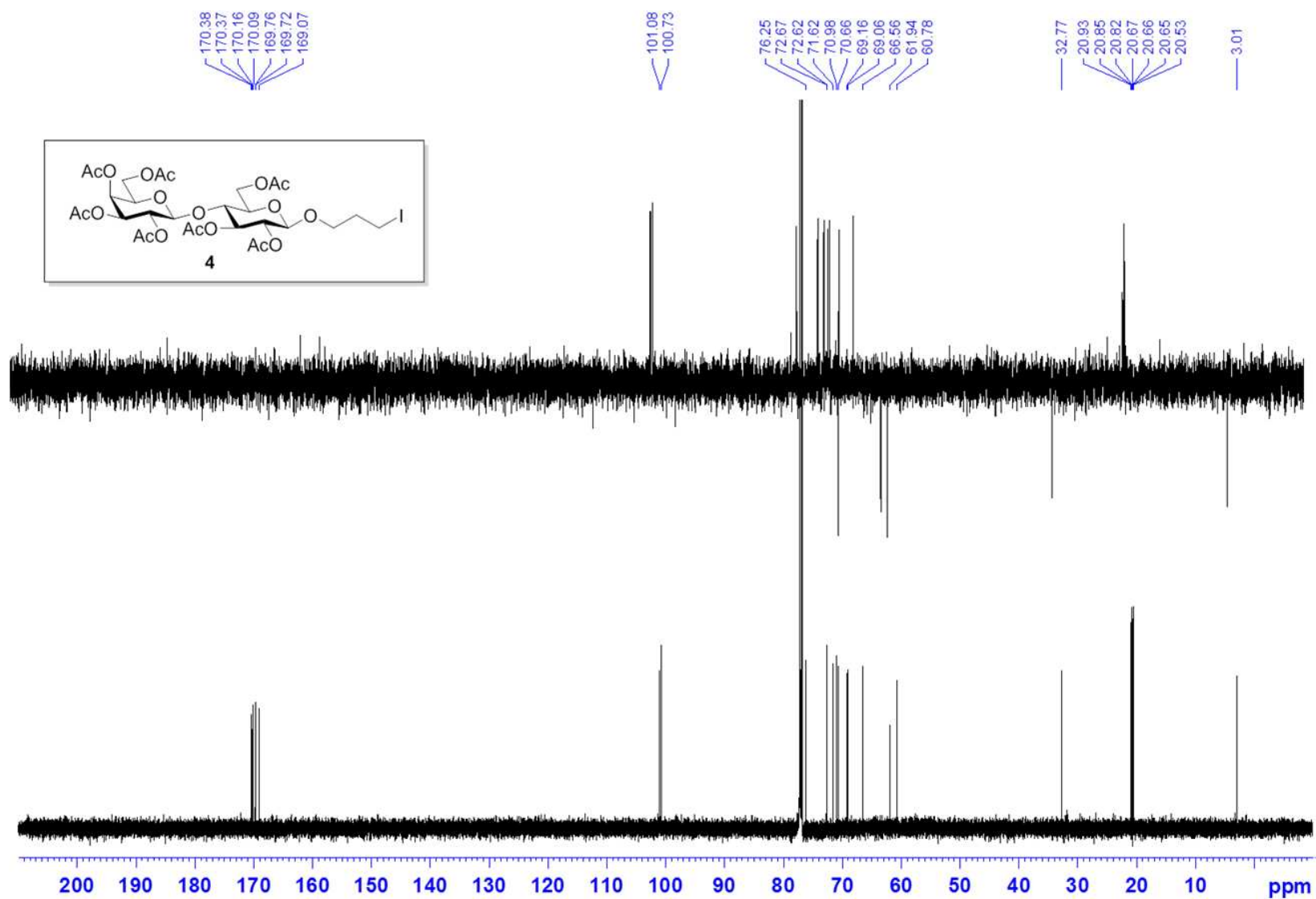


Figure S3. MALDI-TOF HRMS of compound **25**.

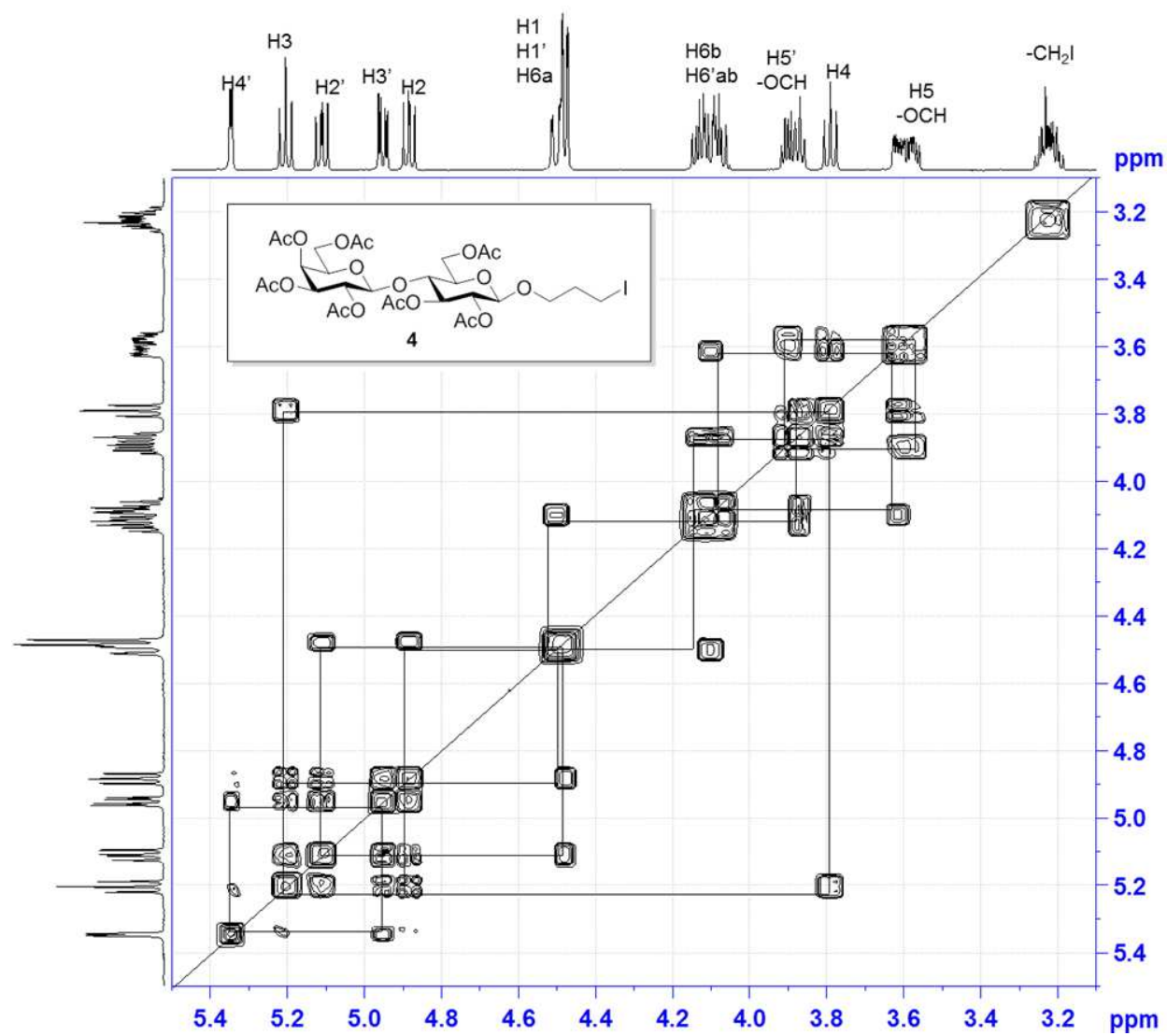
Reference (1) Williams, D. B. G.; Lawton, M. J. *Org. Chem.* **2010**, 75, 8351-8354.



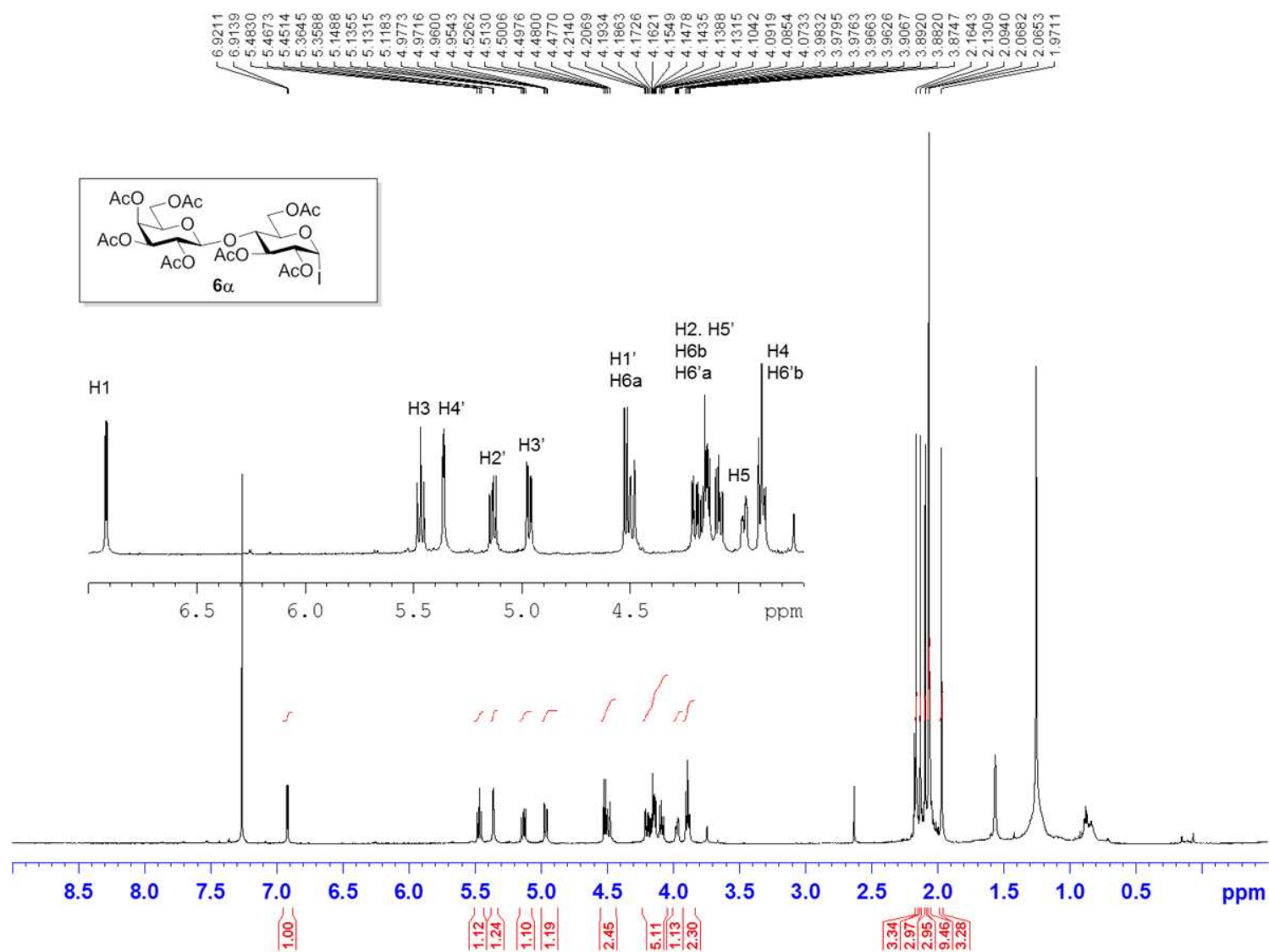
¹H NMR spectrum of compound **4** (CDCl₃, 600 MHz)



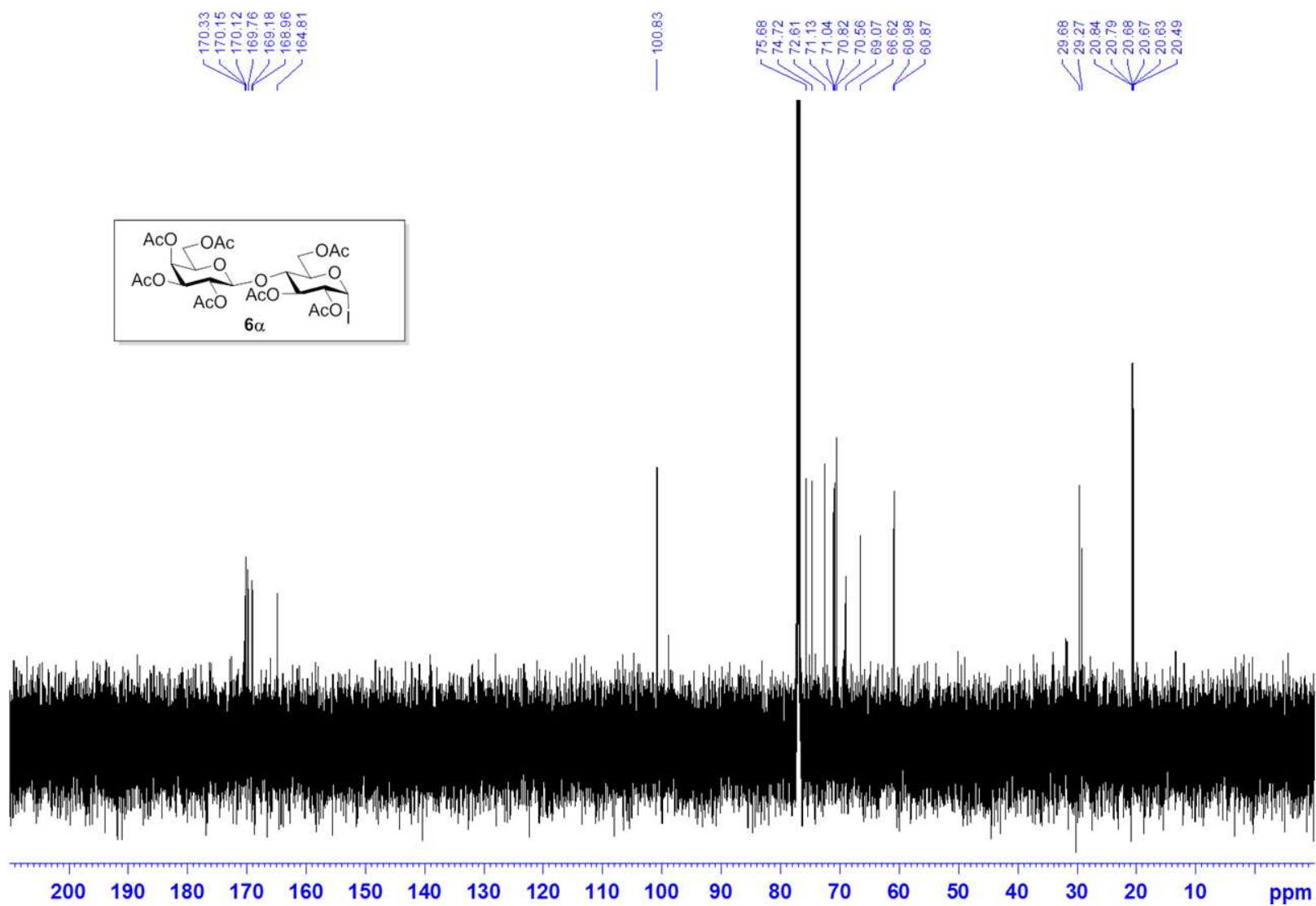
^{13}C and DEPT135 NMR spectrum of compound **4** (CDCl_3 , 150 MHz)

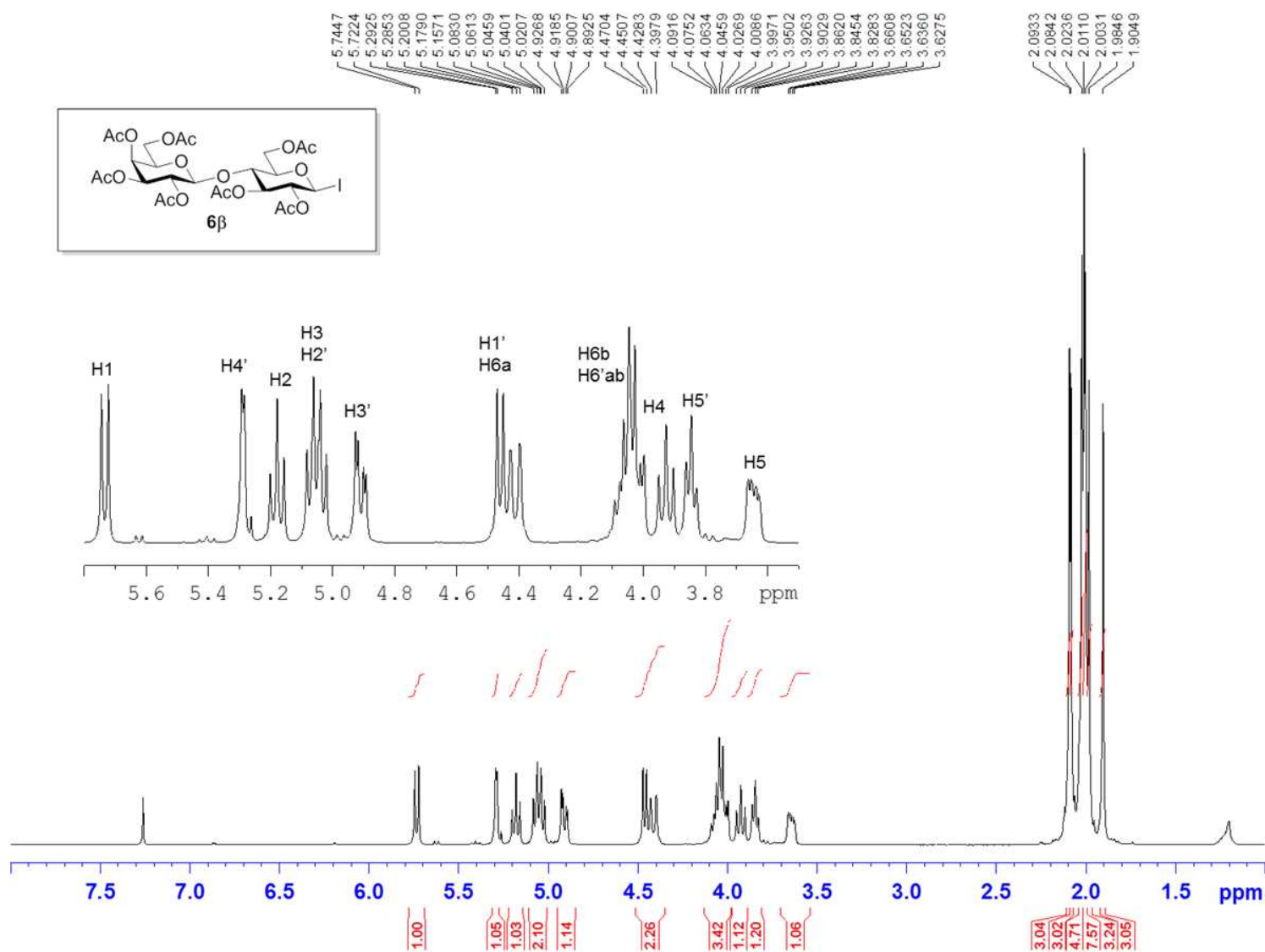


^1H - ^1H COSY spectrum of compound **4** (CDCl_3 , 600 MHz)

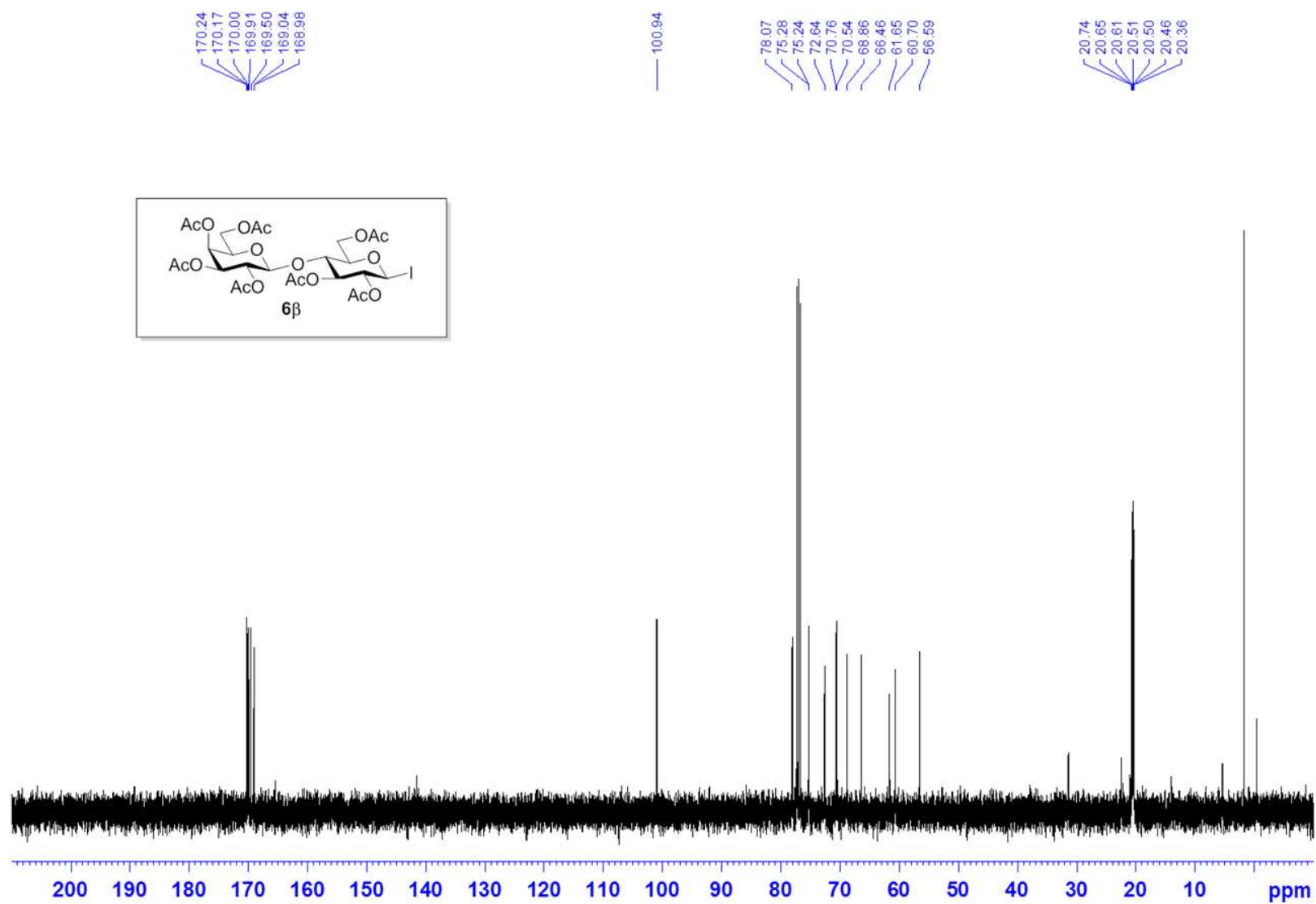


in situ ^1H NMR spectrum of compound **6α** (CDCl_3 , 600 MHz)

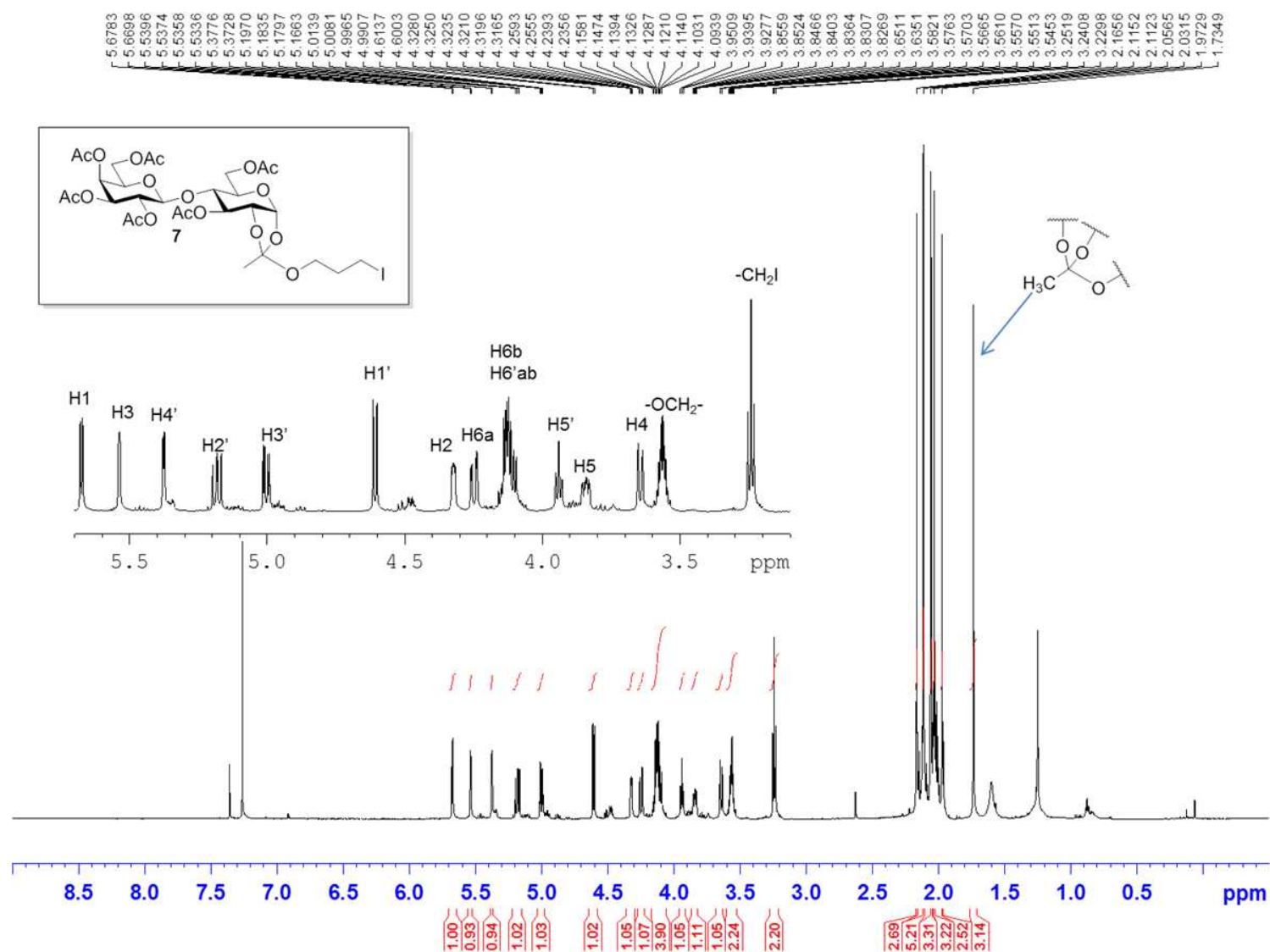




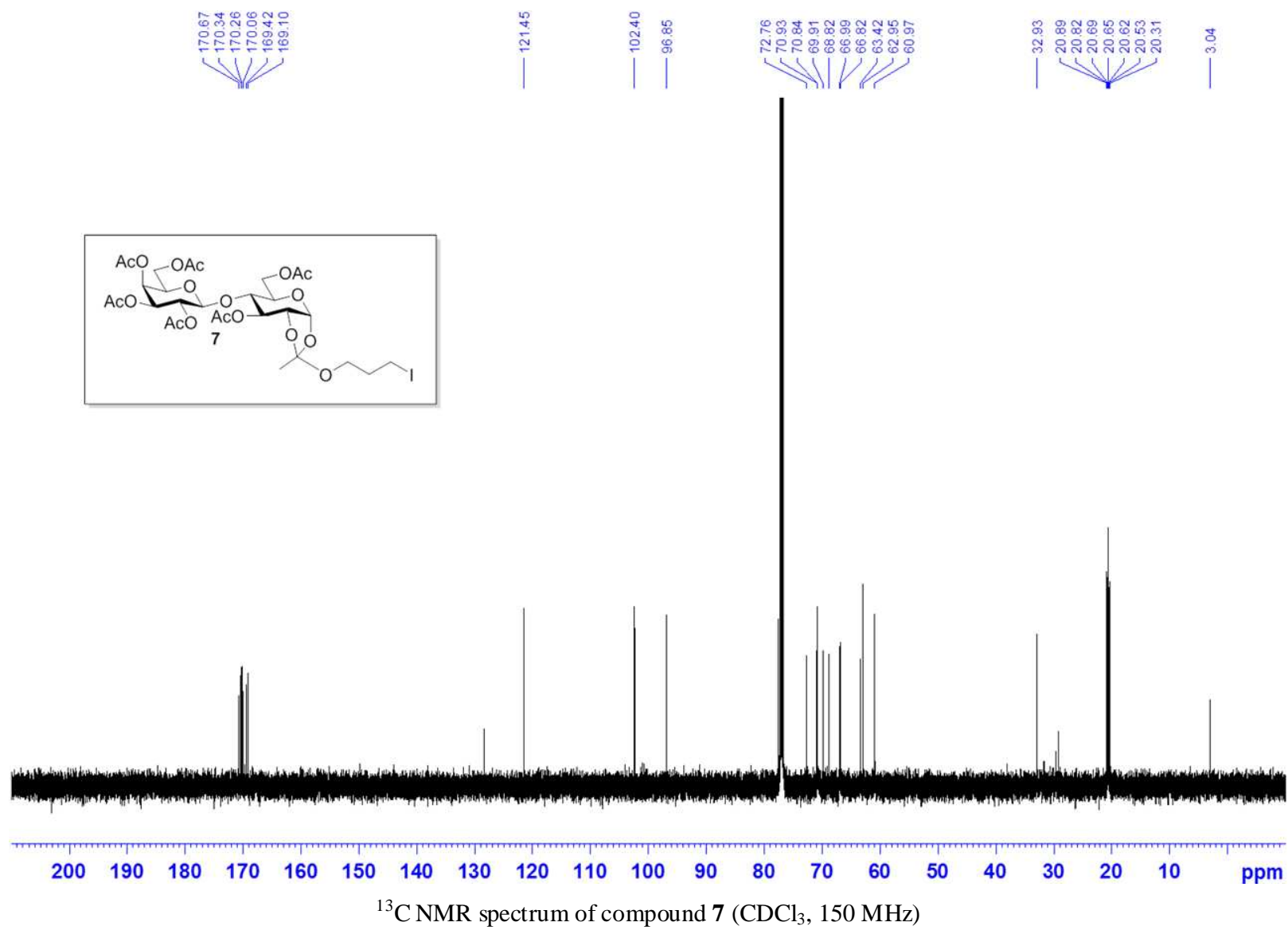
in situ ^1H NMR spectrum of compound **6 β** (CDCl_3 , 400 MHz)

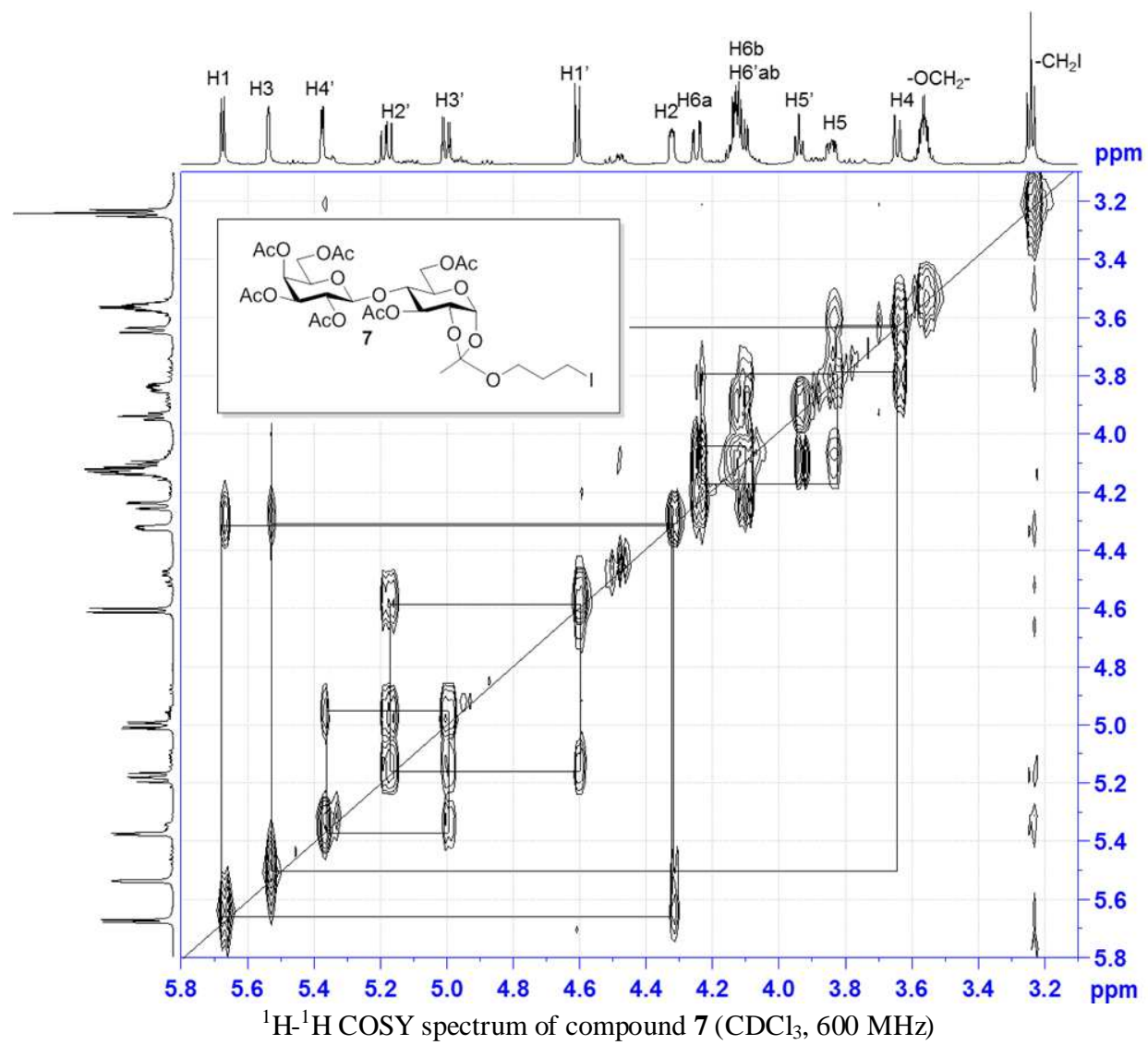


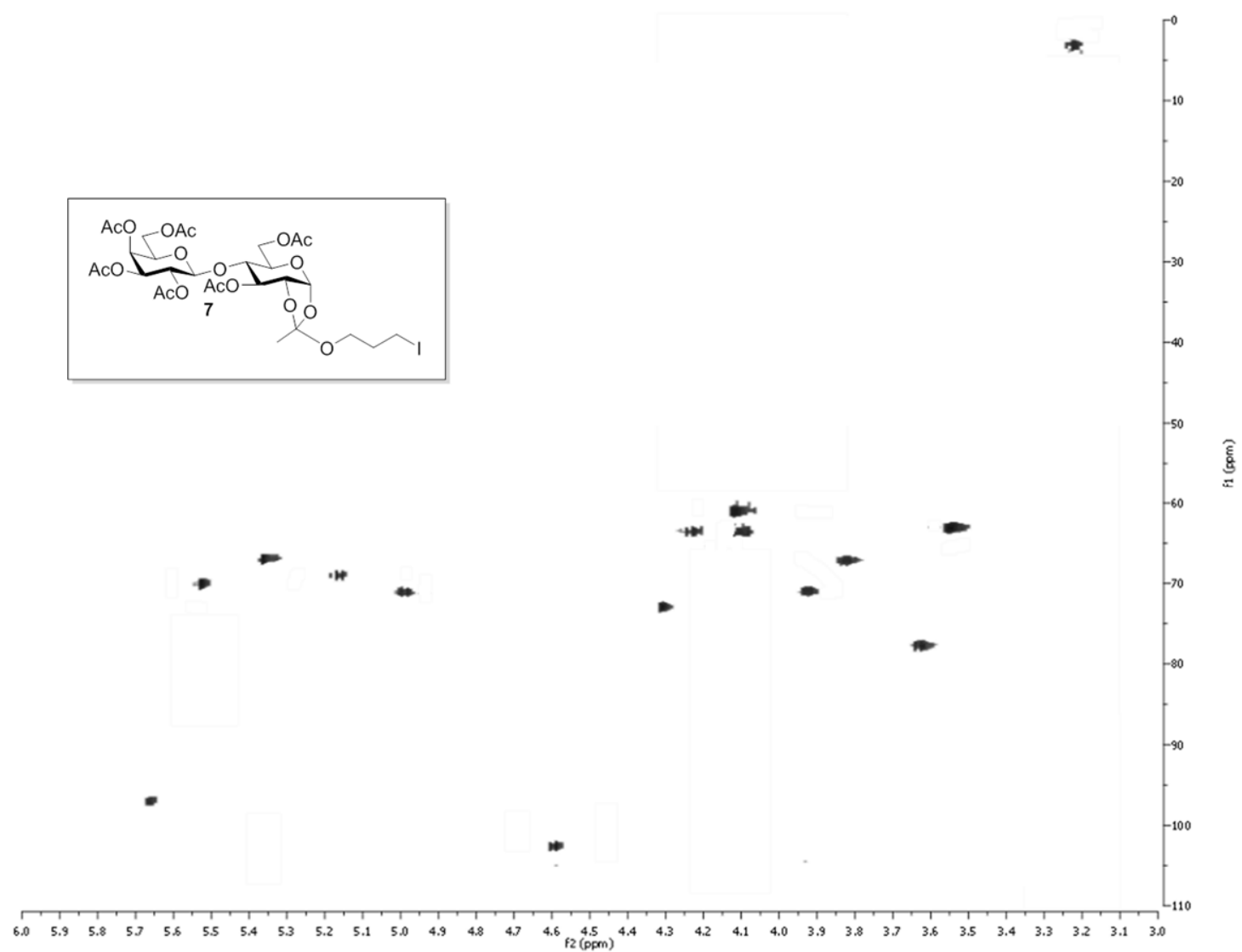
in situ ¹³C NMR spectrum of compound **6β** (CDCl₃, 100 MHz)



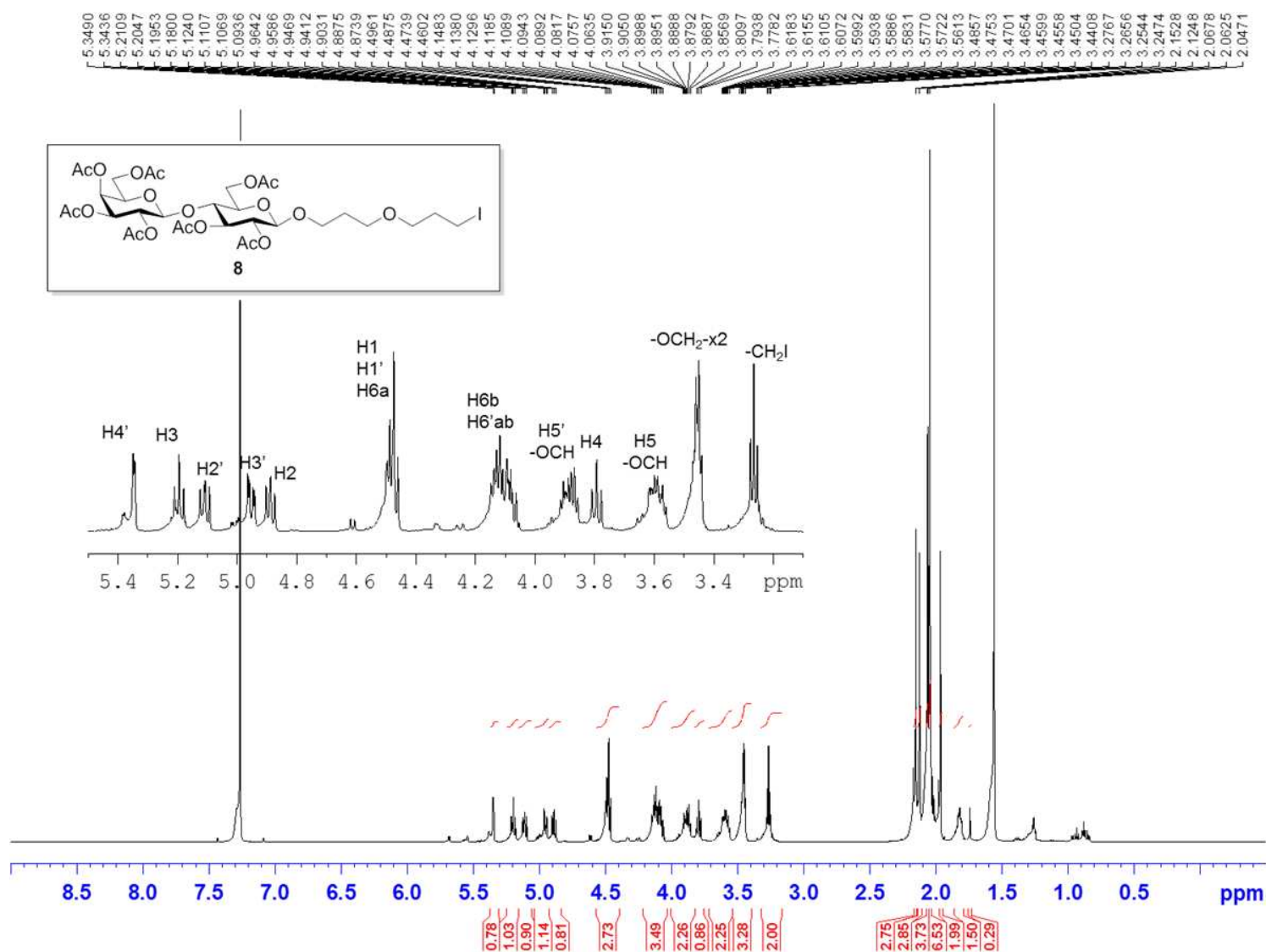
¹H NMR spectrum of compound **7** (CDCl₃, 600 MHz)



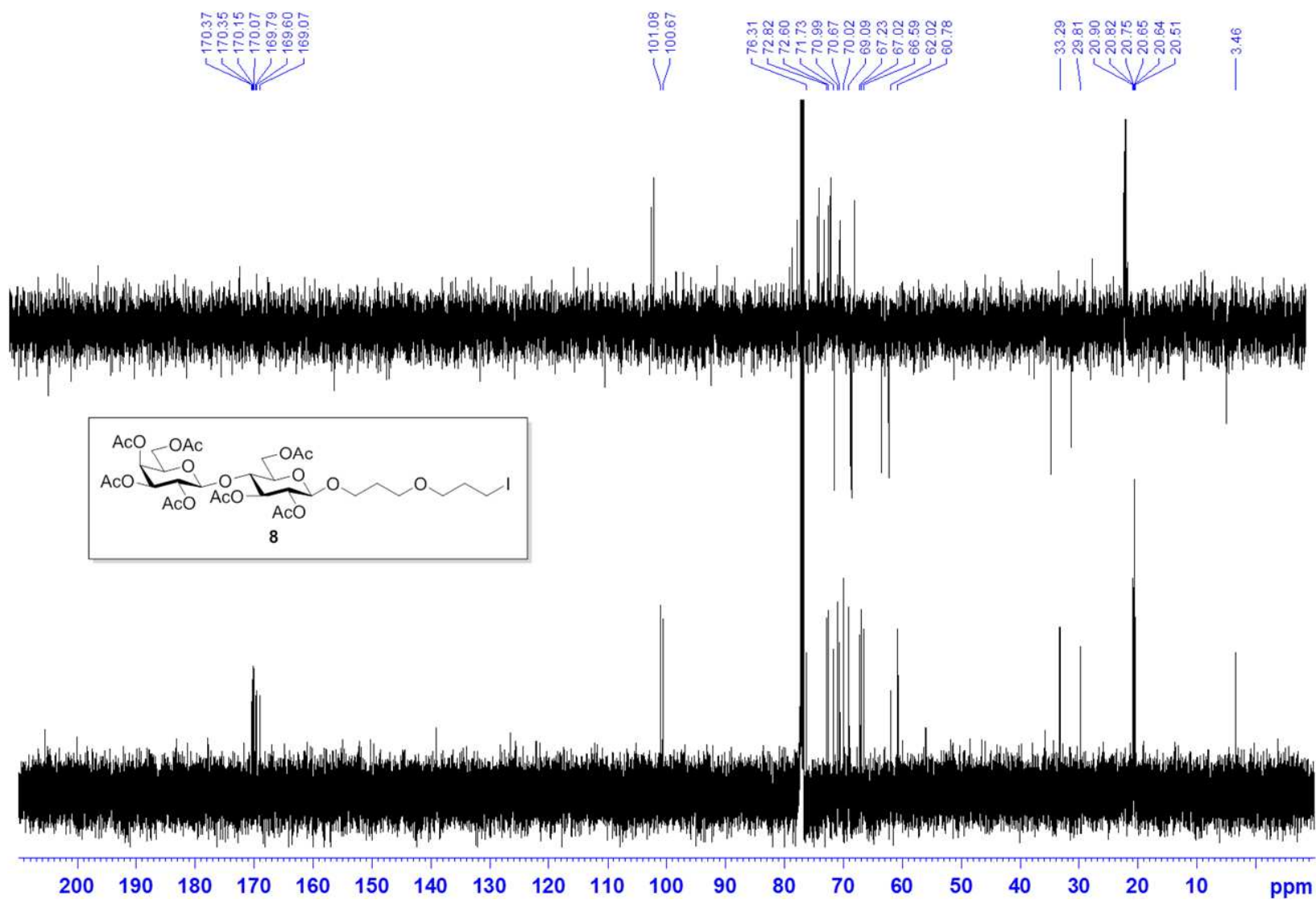




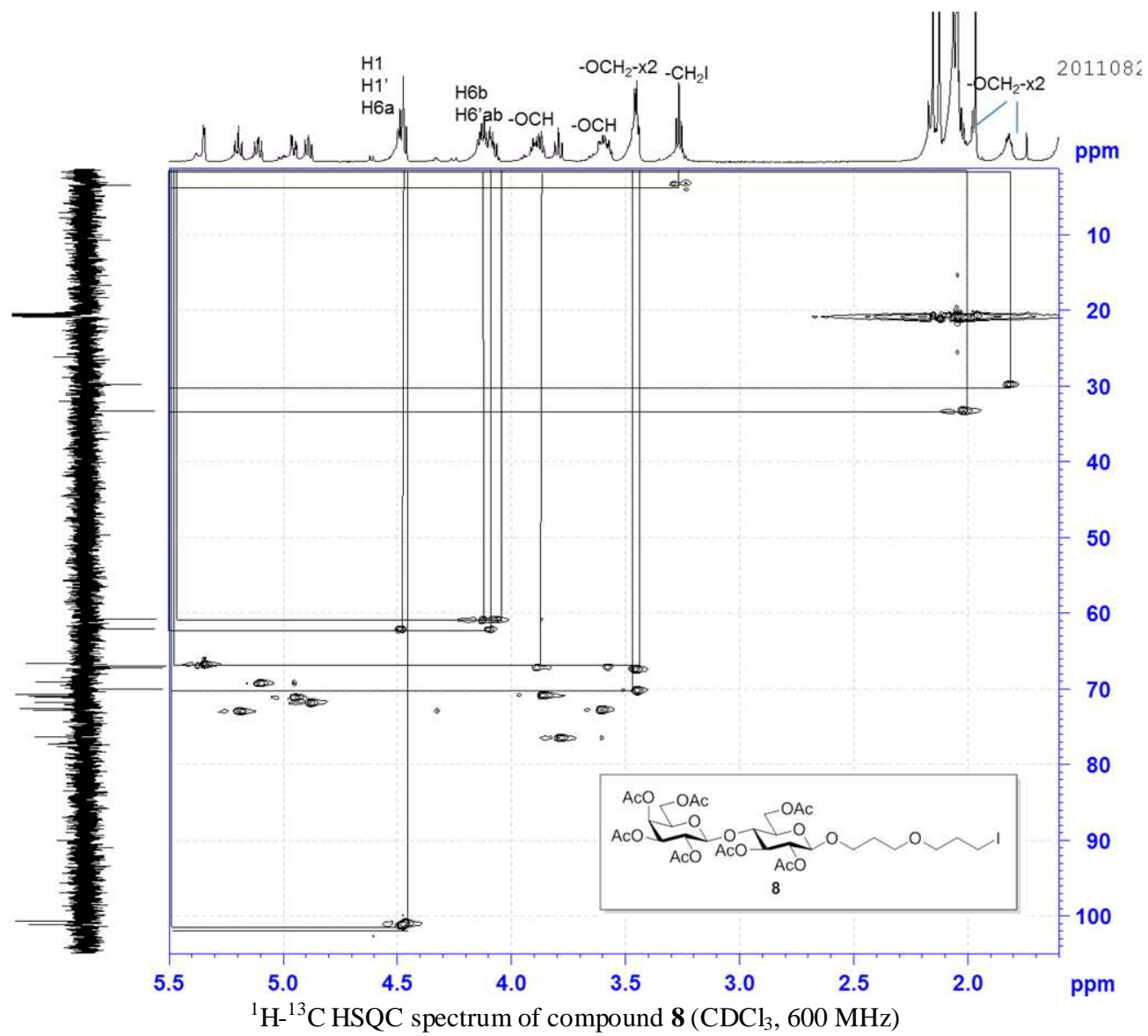
^1H - ^{13}C HSQC spectrum of compound **7** (CDCl_3 , 600 MHz)

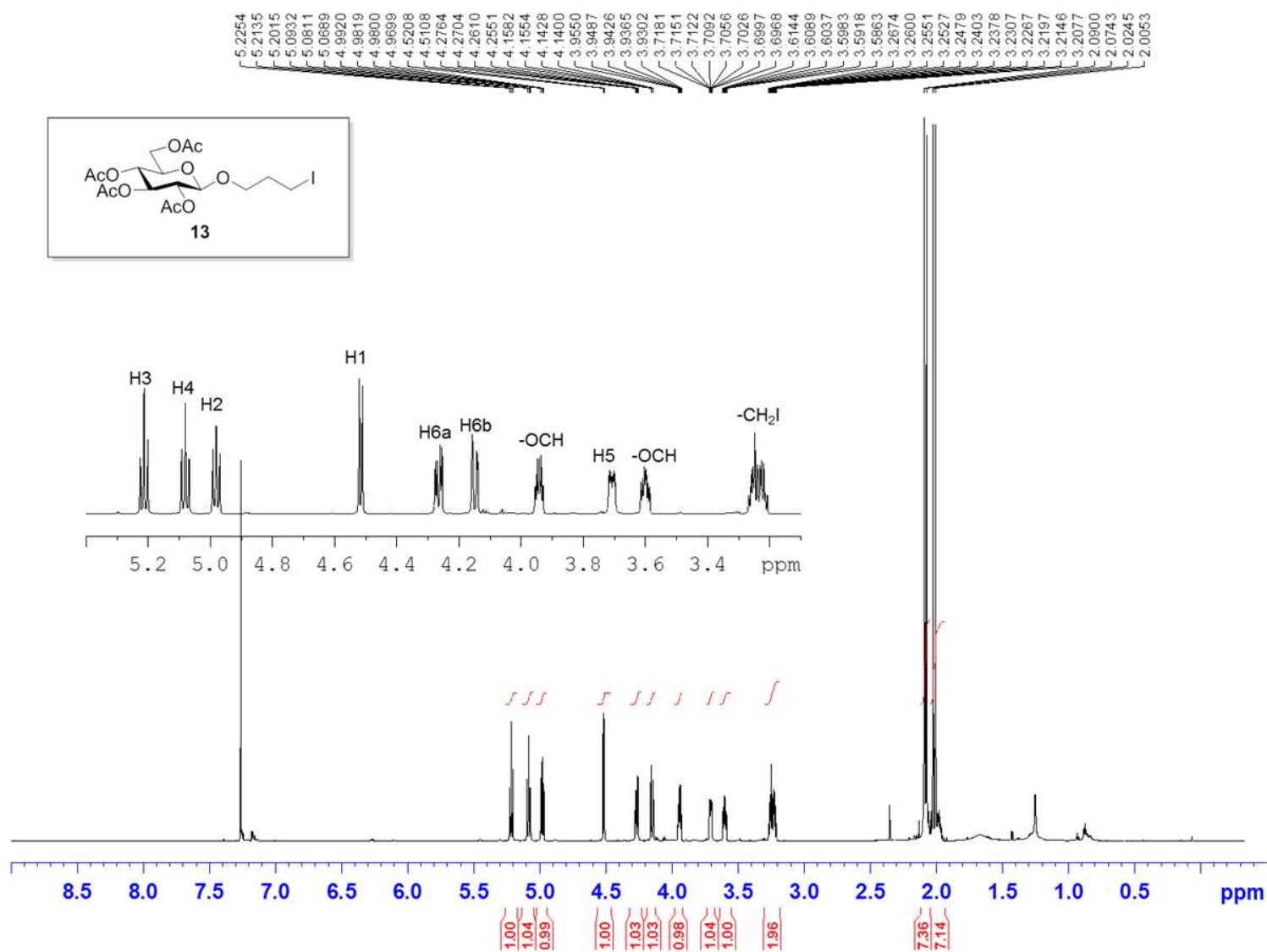


¹H NMR spectrum of compound **8** (CDCl₃, 600 MHz)

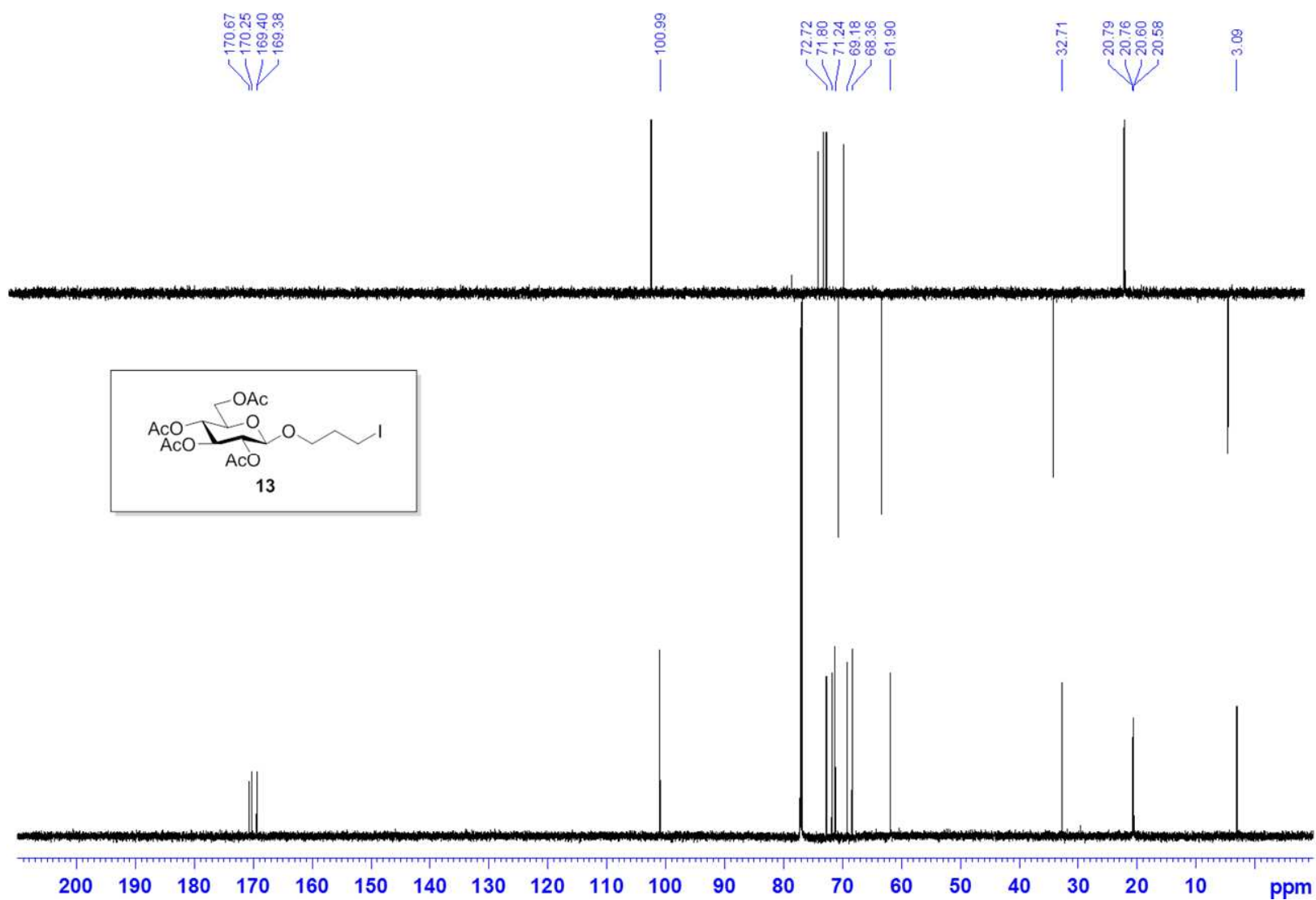


¹³C and DEPT135 NMR spectrum of compound **8** (CDCl₃, 150 MHz)

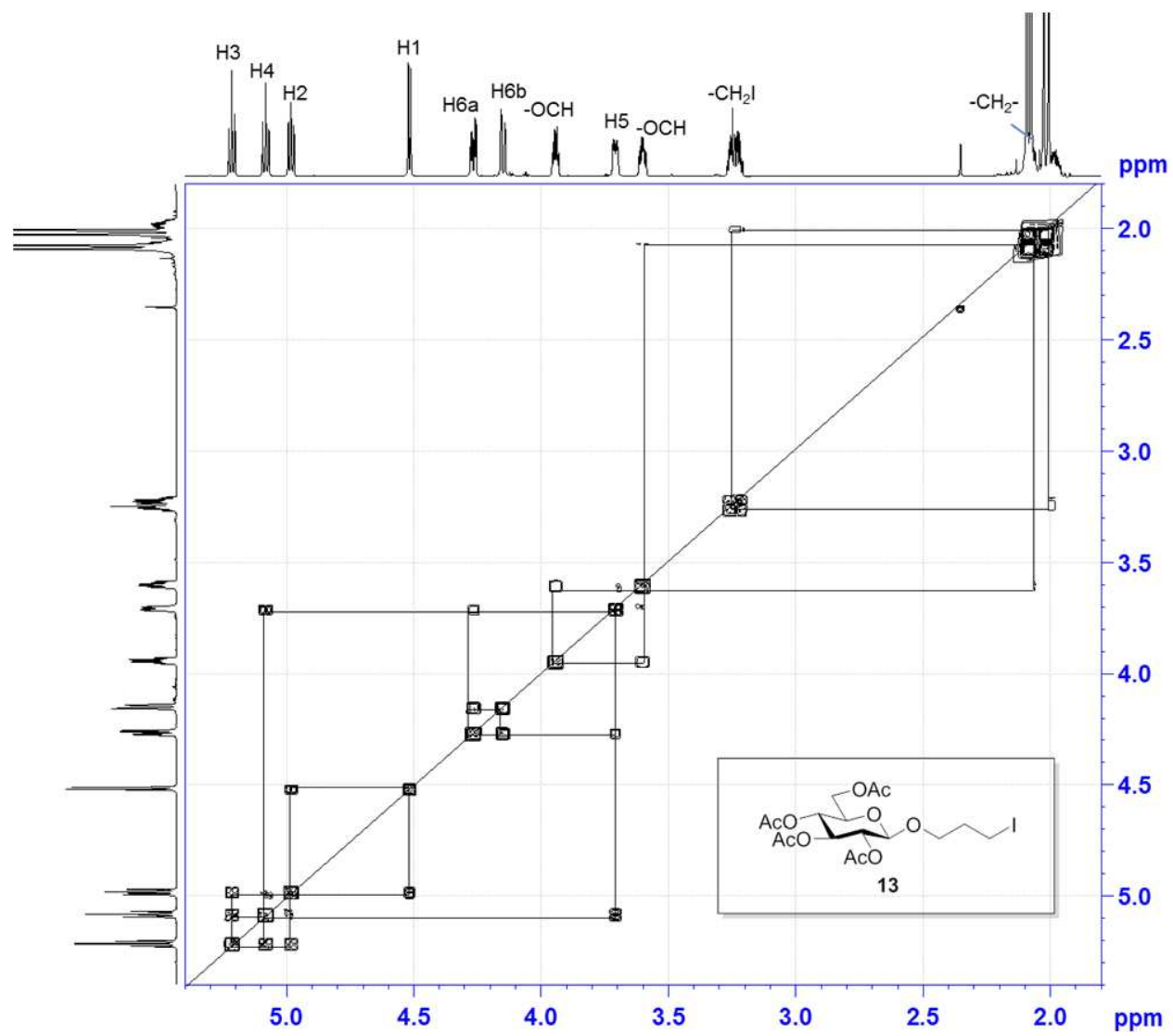




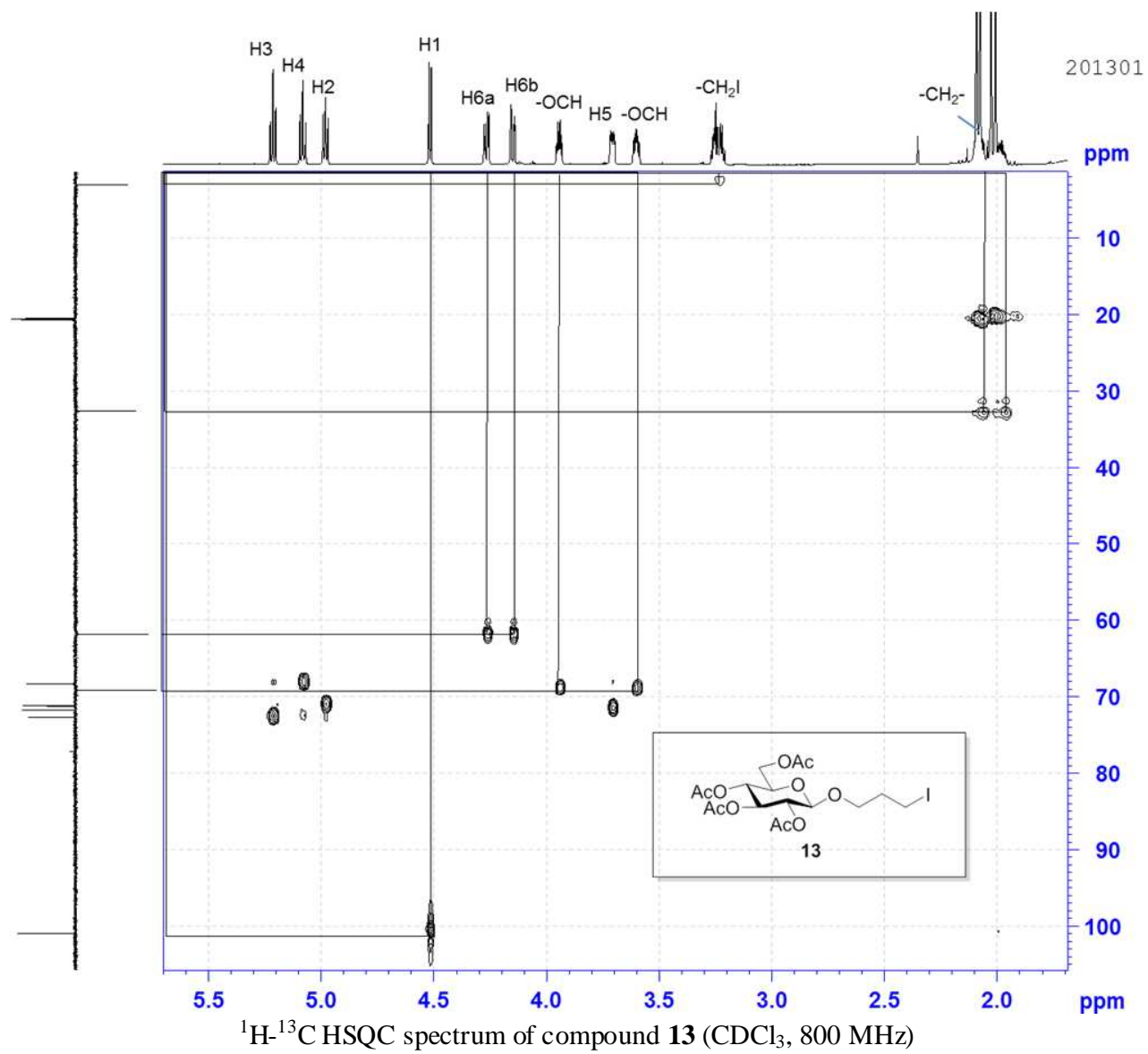
¹H NMR spectrum of compound **13** (CDCl₃, 800 MHz)

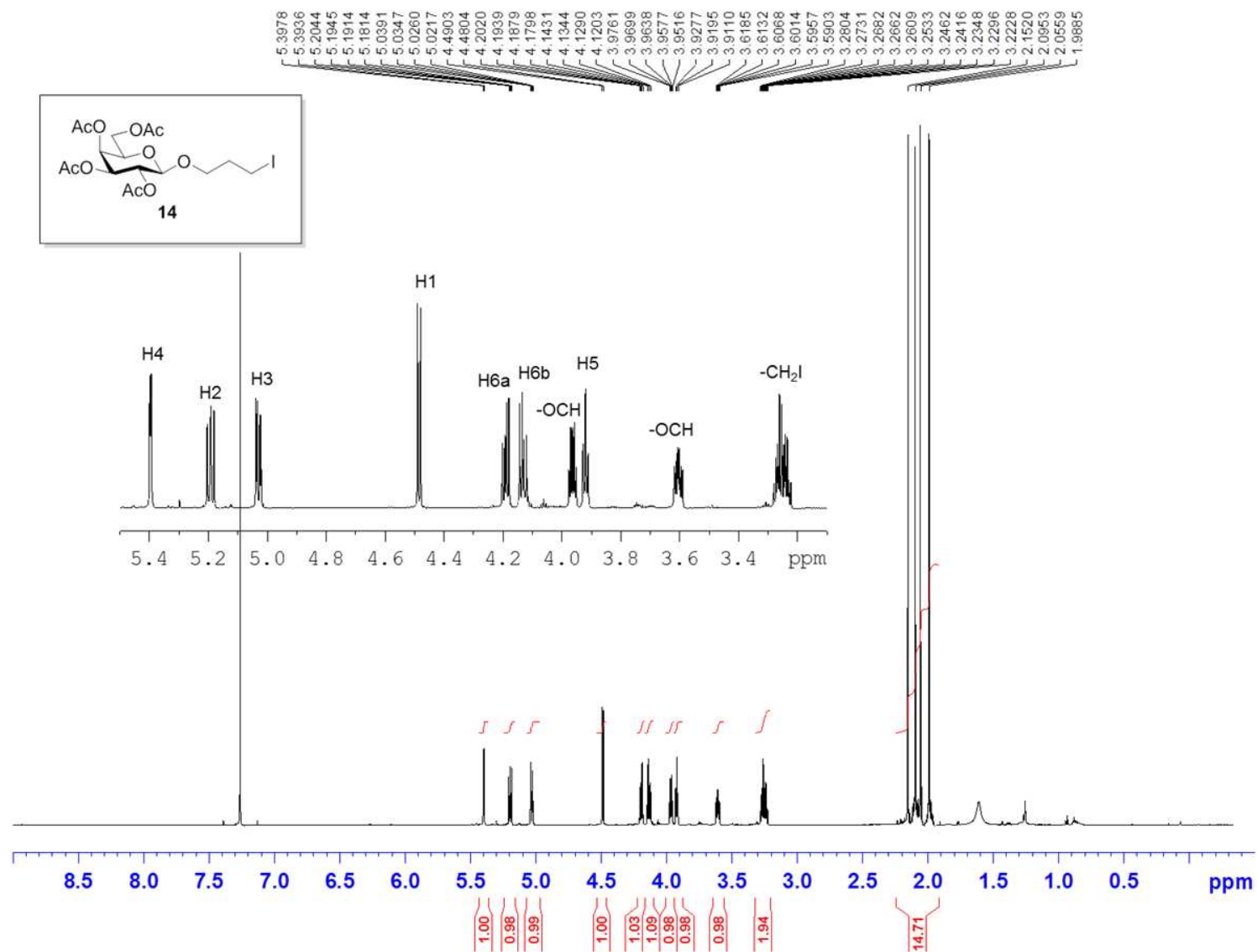


^{13}C and DEPT135 NMR spectrum of compound **13** (CDCl_3 , 200 MHz)

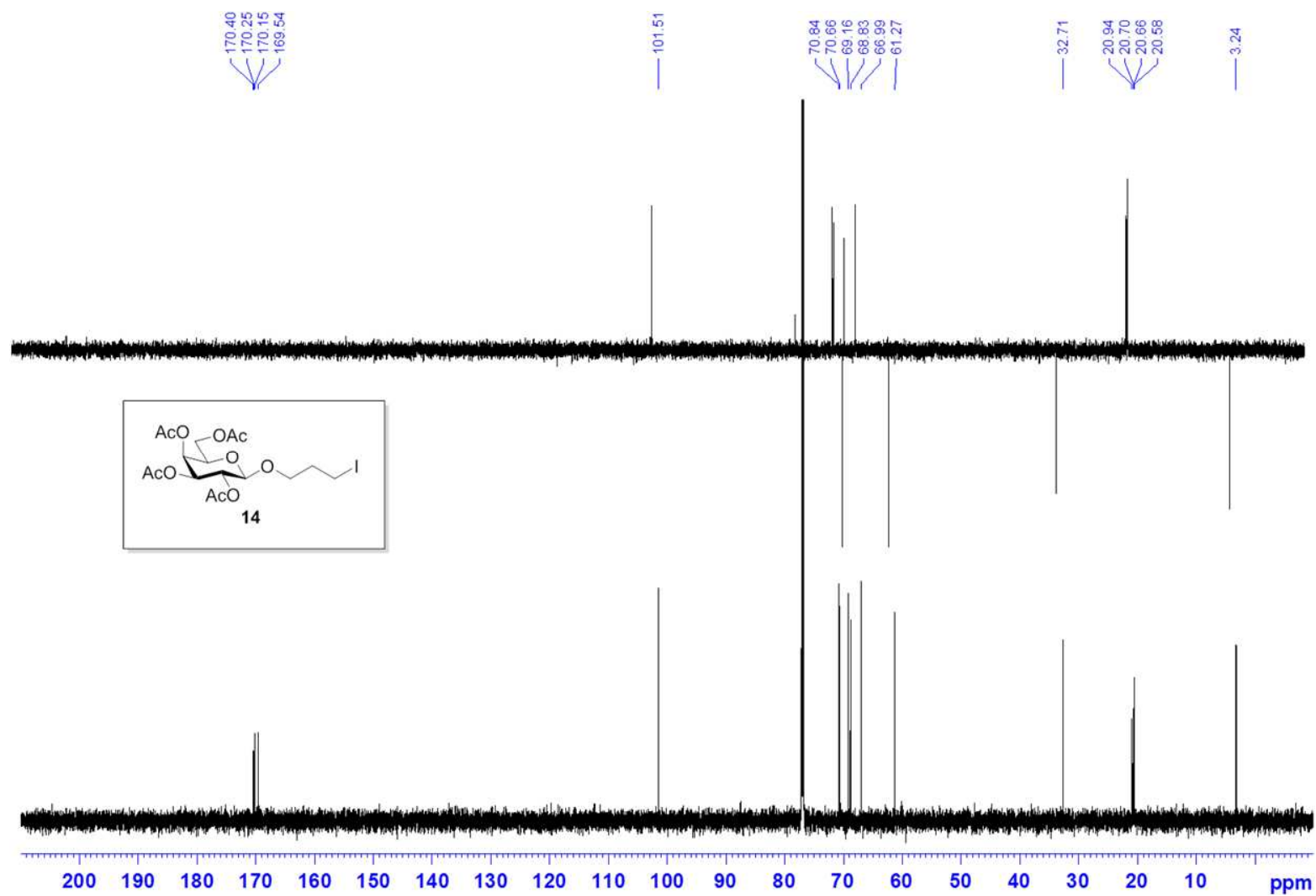


^1H - ^1H COSY spectrum of compound **13** (CDCl_3 , 800 MHz)

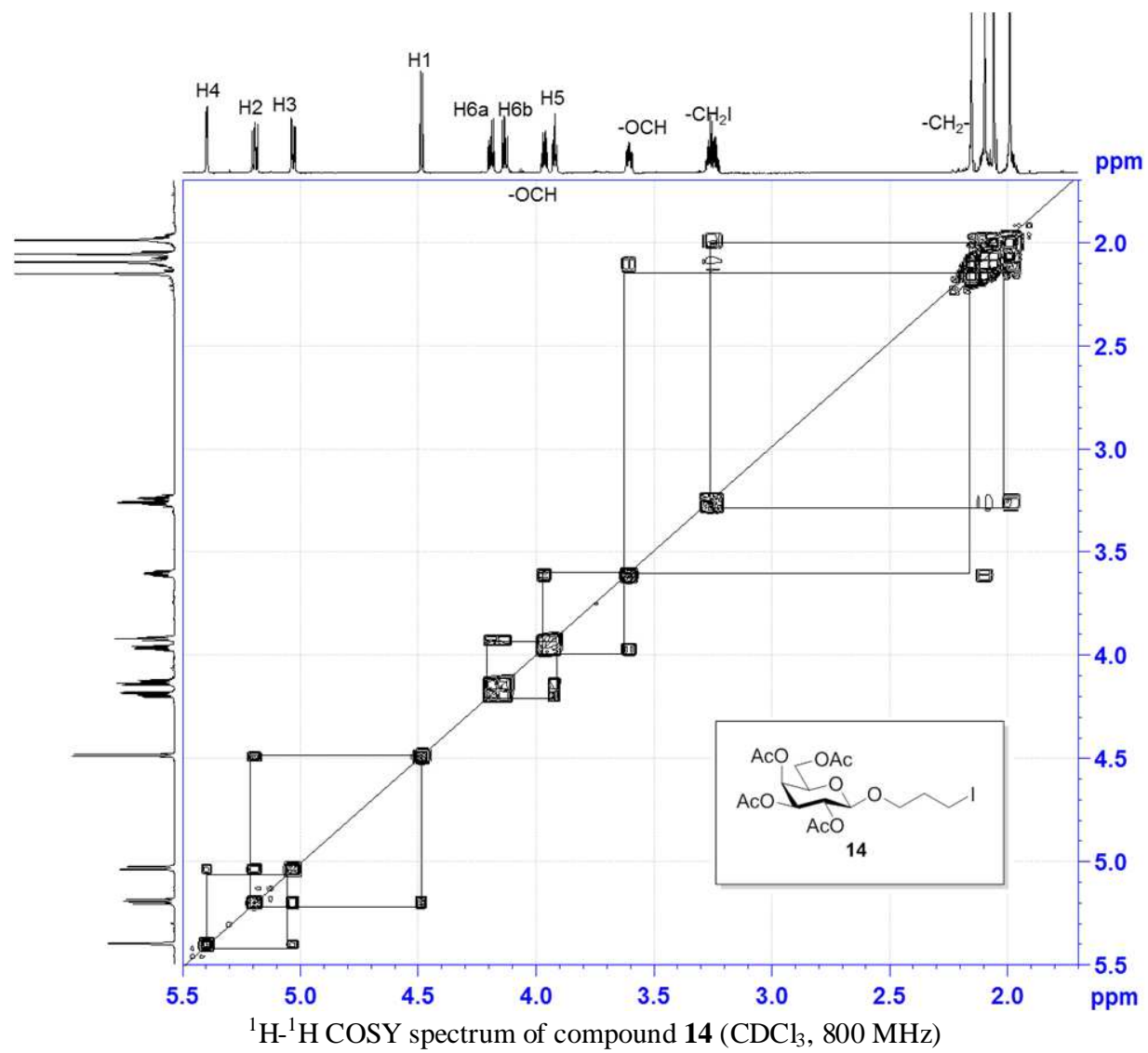


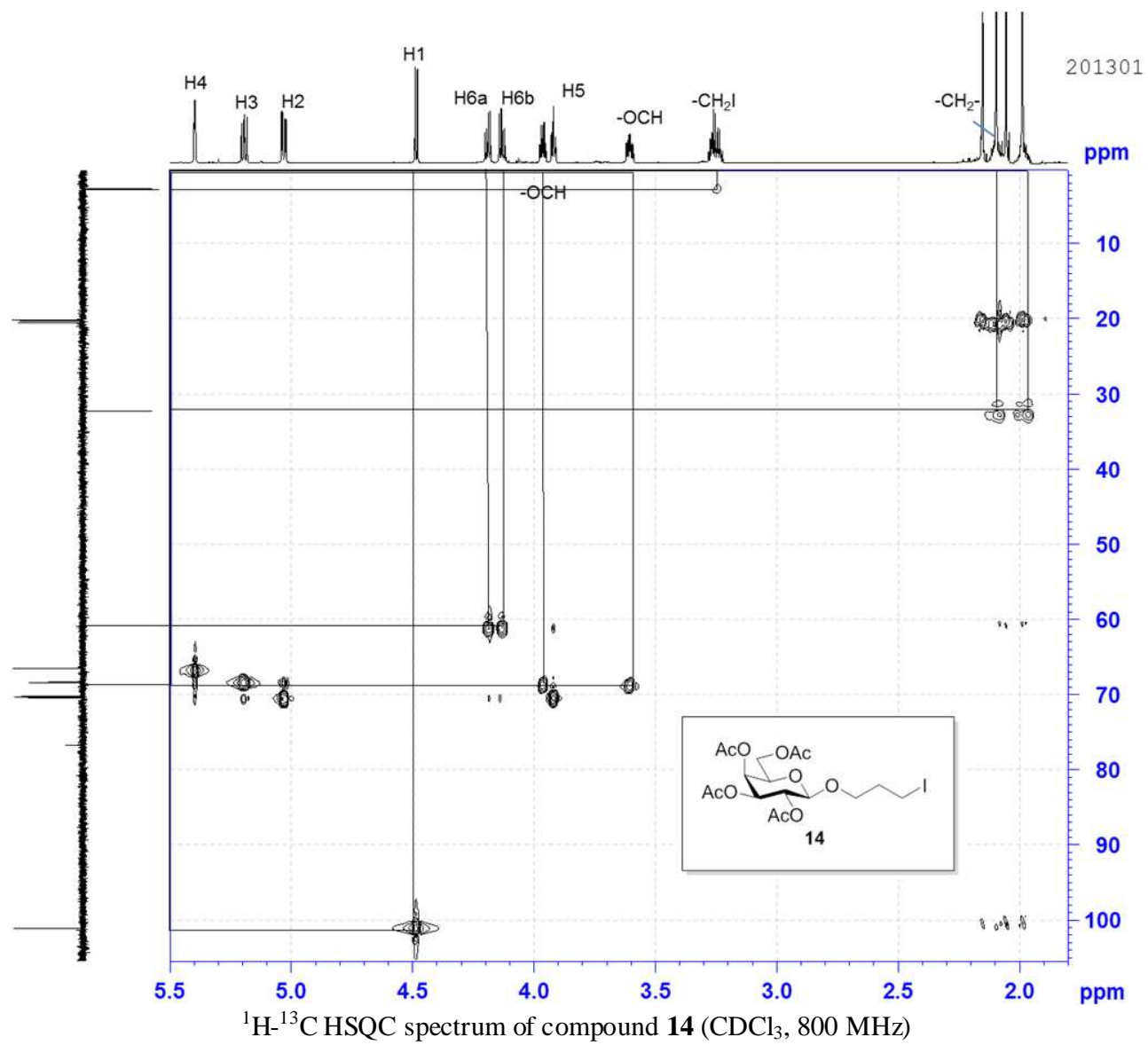


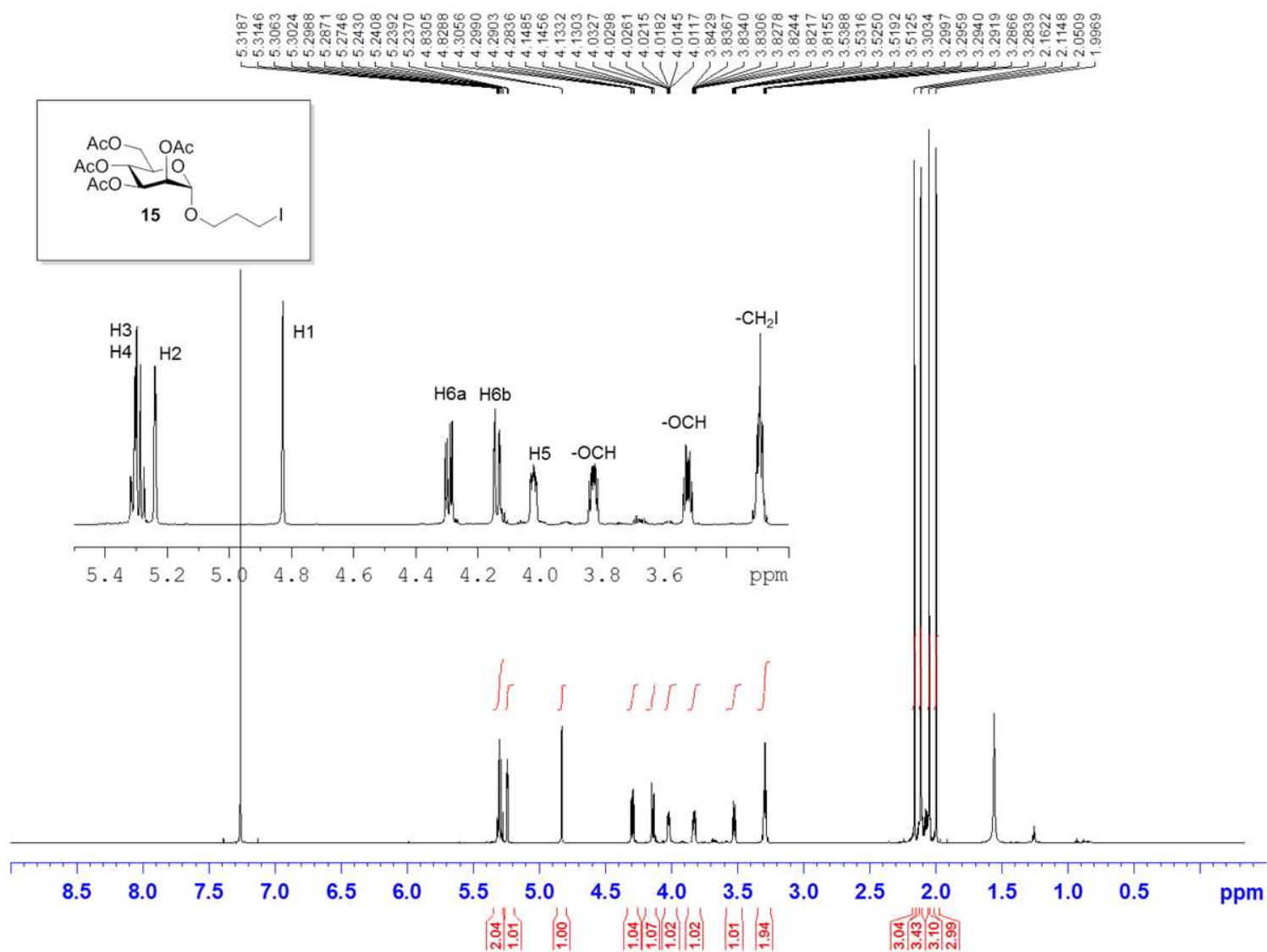
¹H NMR spectrum of compound **14** (CDCl₃, 800 MHz)



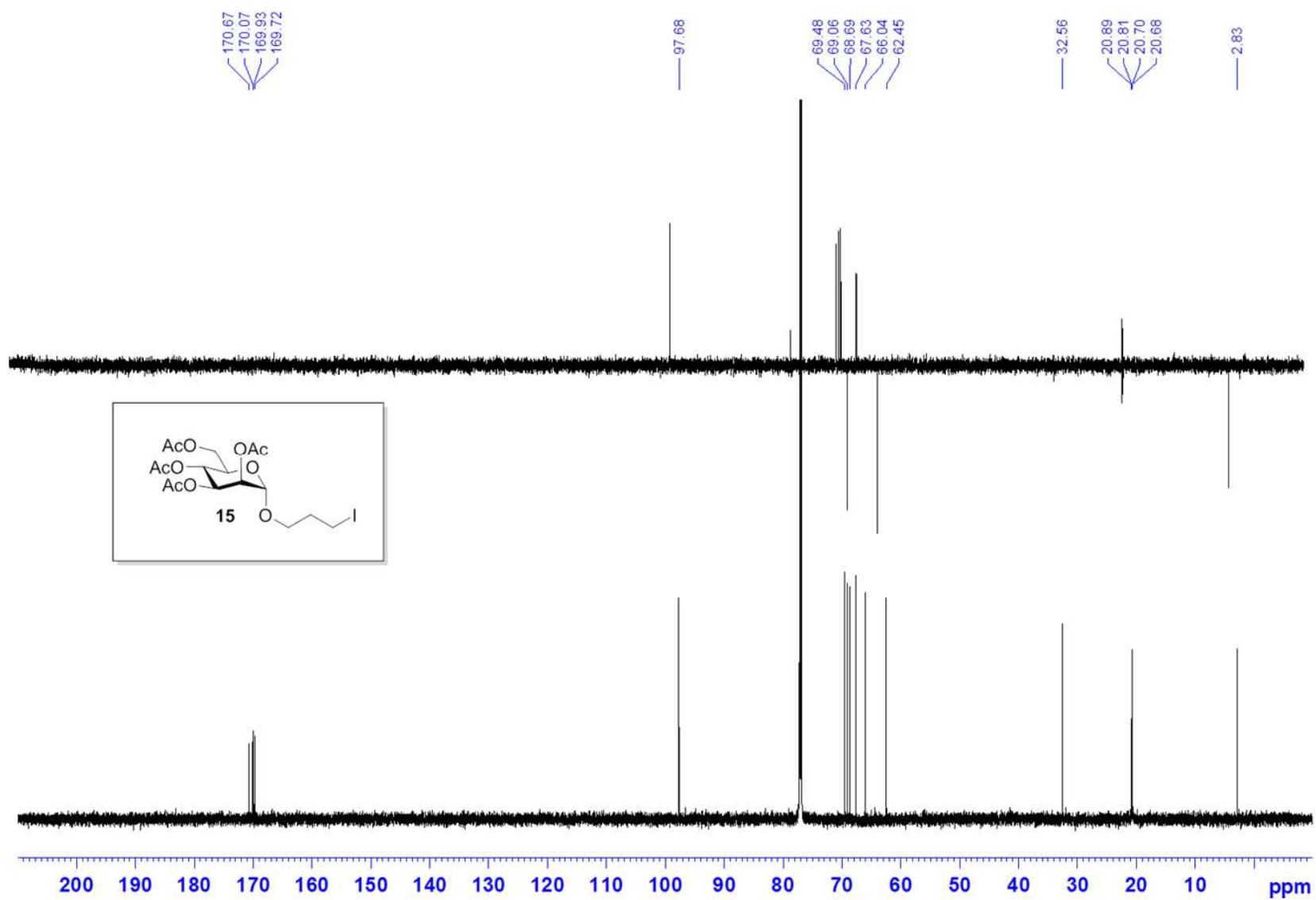
¹³C and DEPT135 NMR spectrum of compound **14** (CDCl₃, 200 MHz)



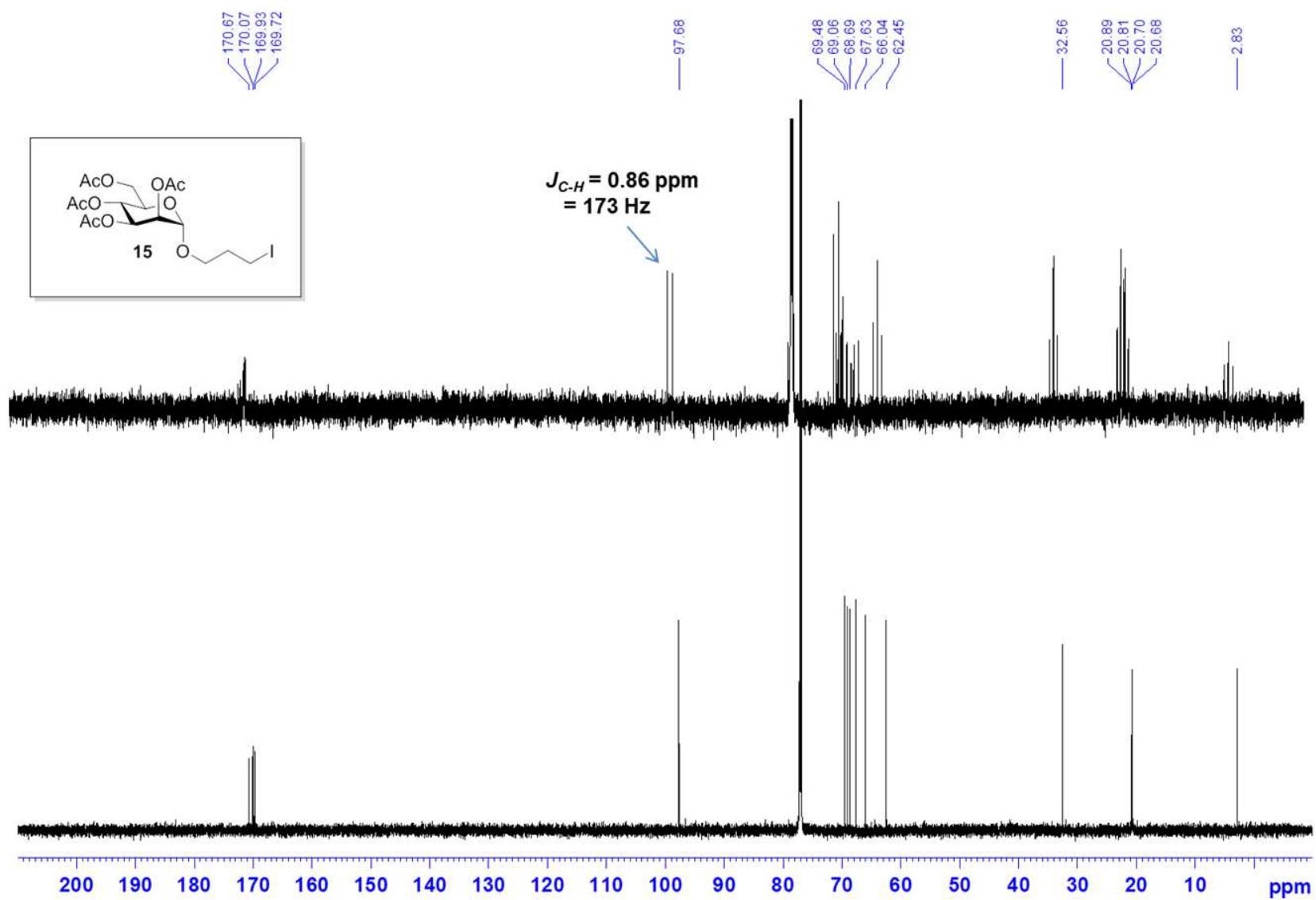


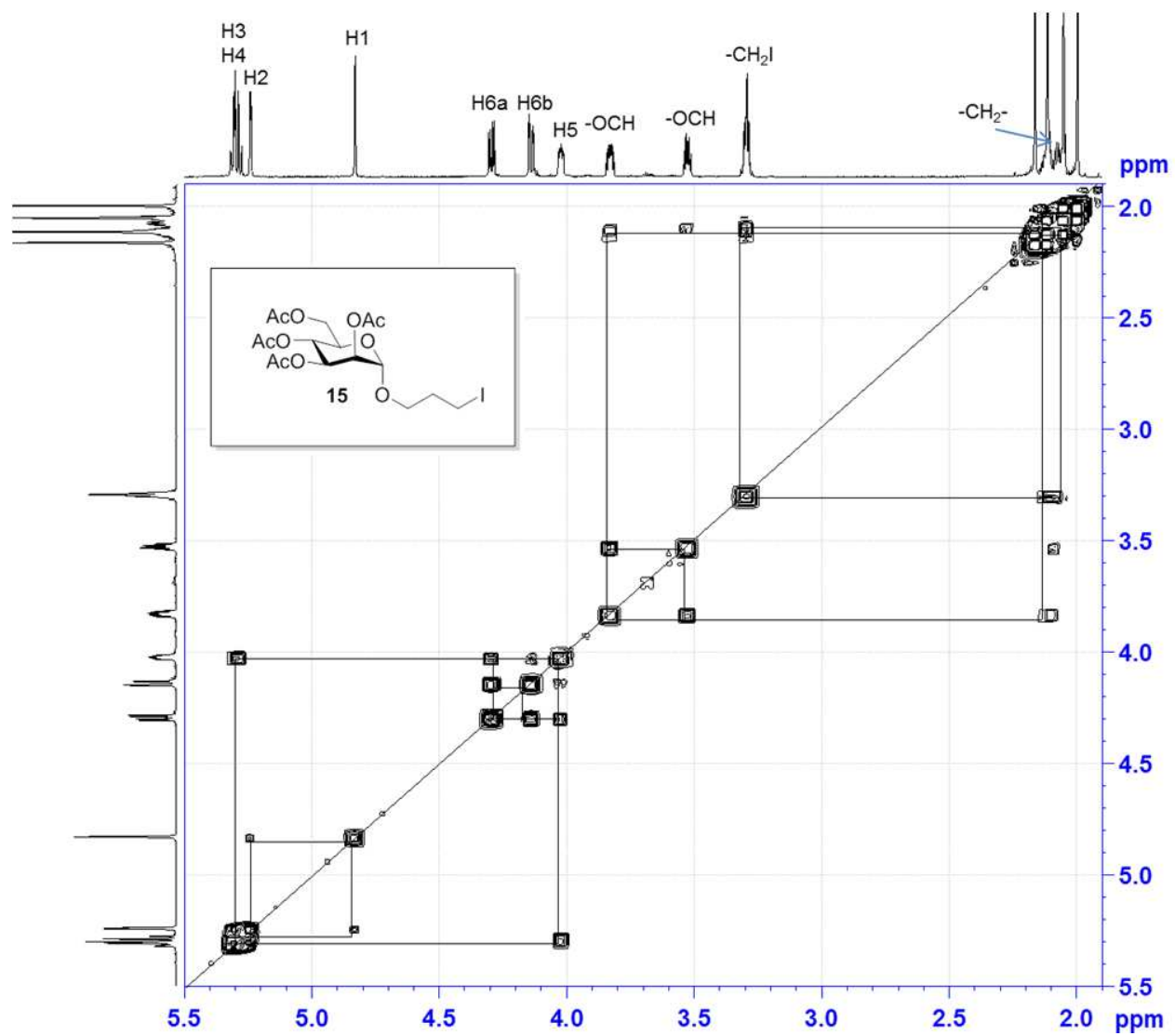


¹H NMR spectrum of compound **15** (CDCl₃, 800 MHz)

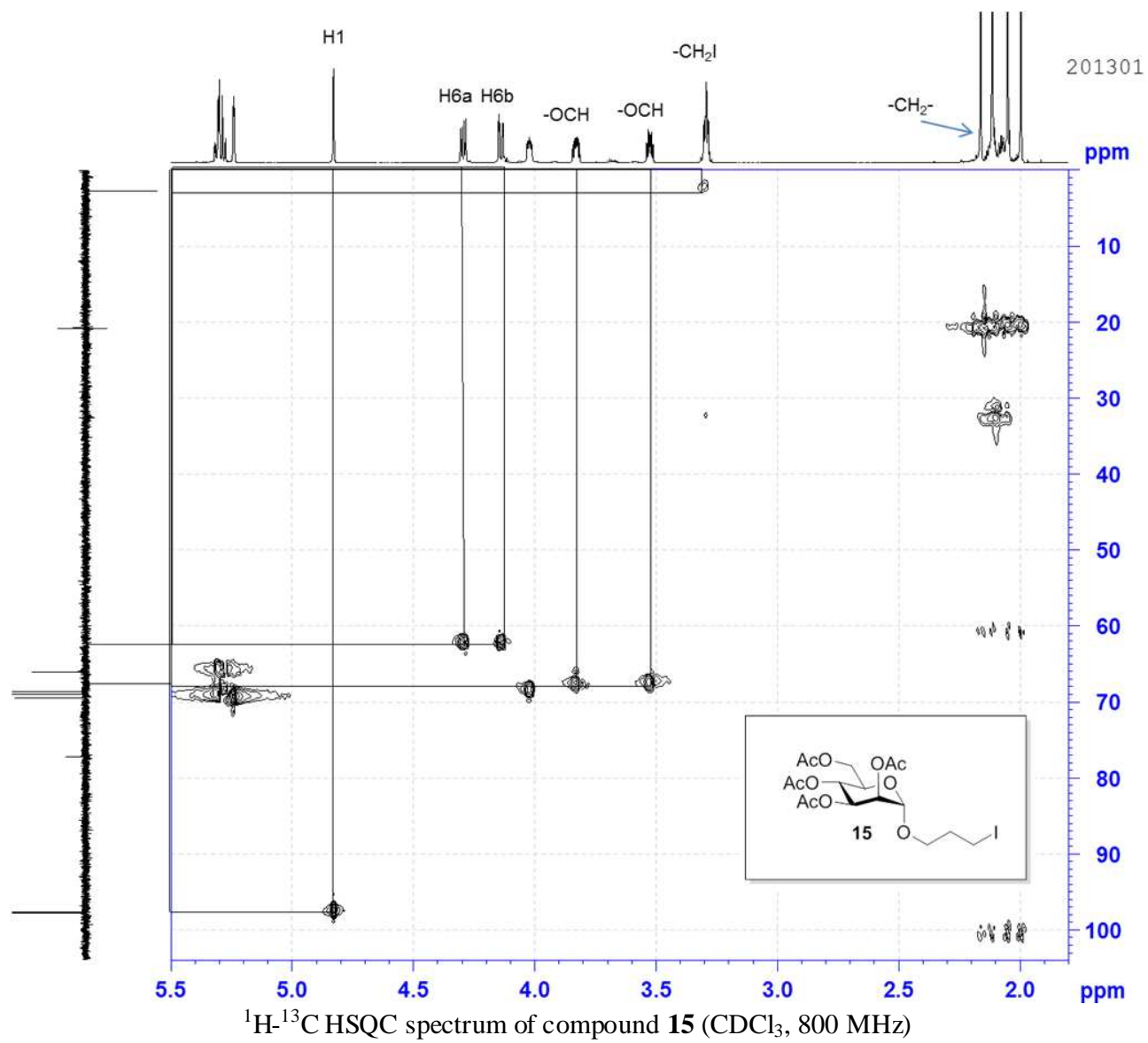


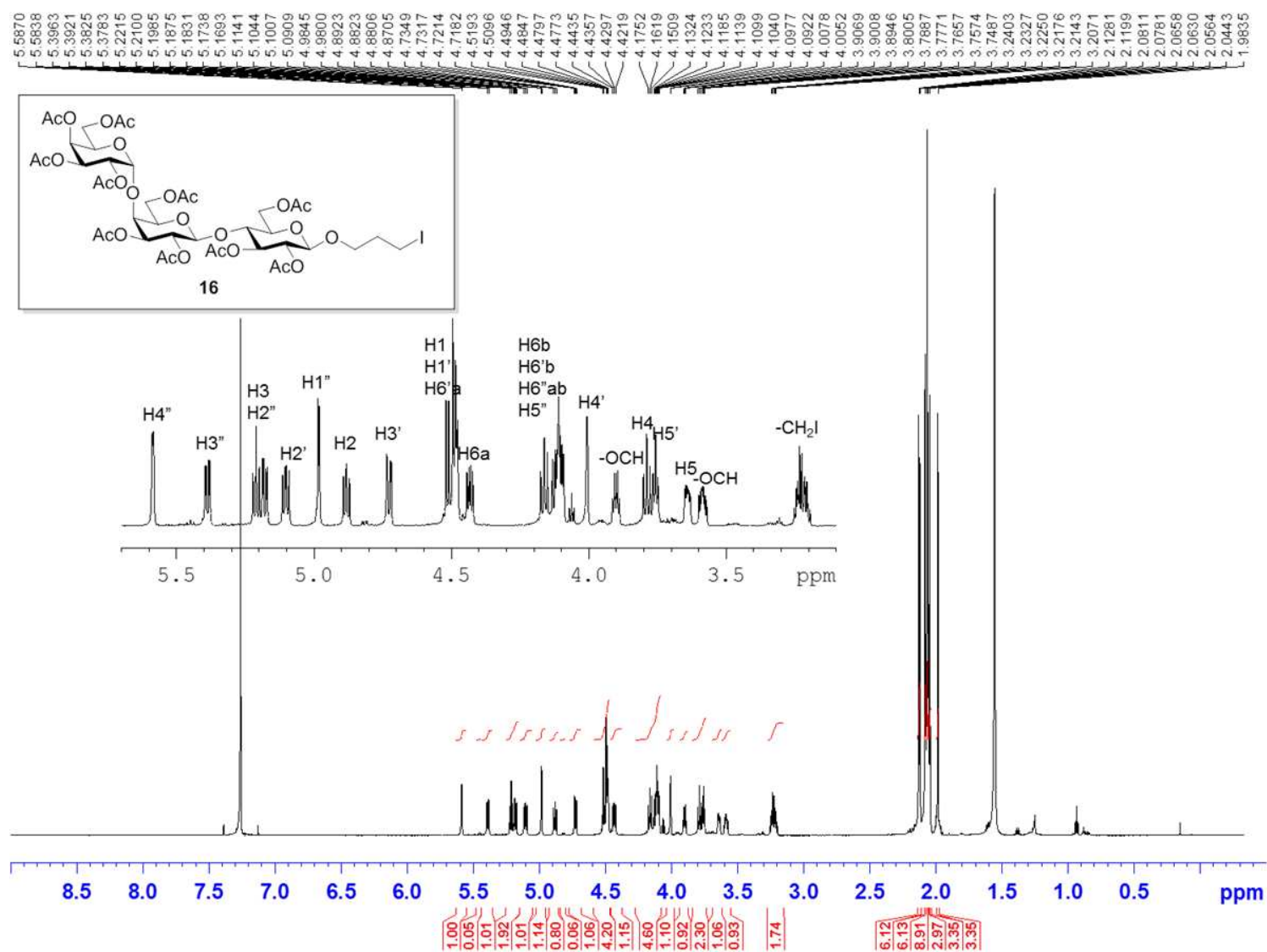
¹³C and DEPT135 NMR spectrum of compound **15** (CDCl₃, 200 MHz)



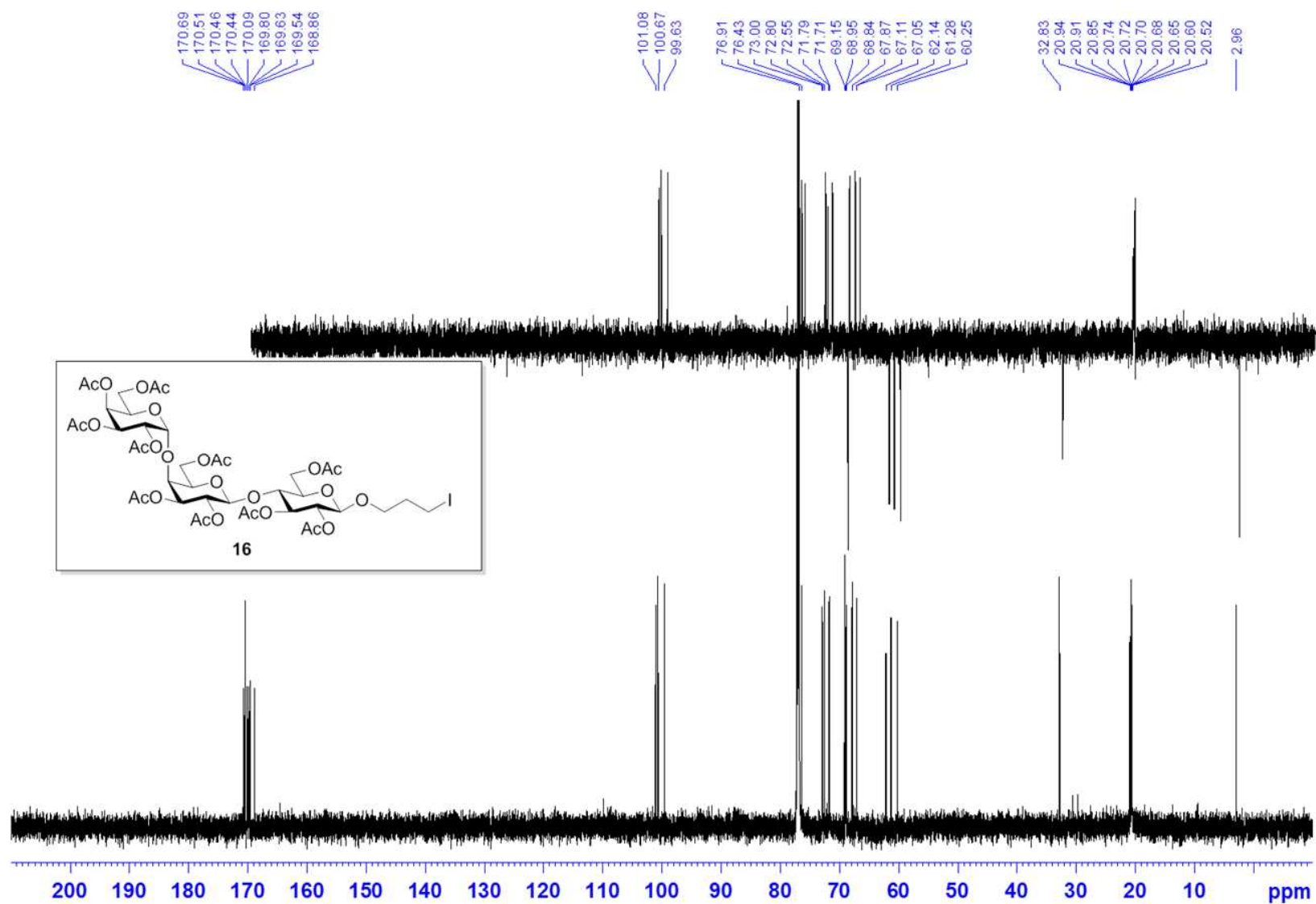


^1H - ^1H COSY spectrum of compound **15** (CDCl_3 , 800 MHz)

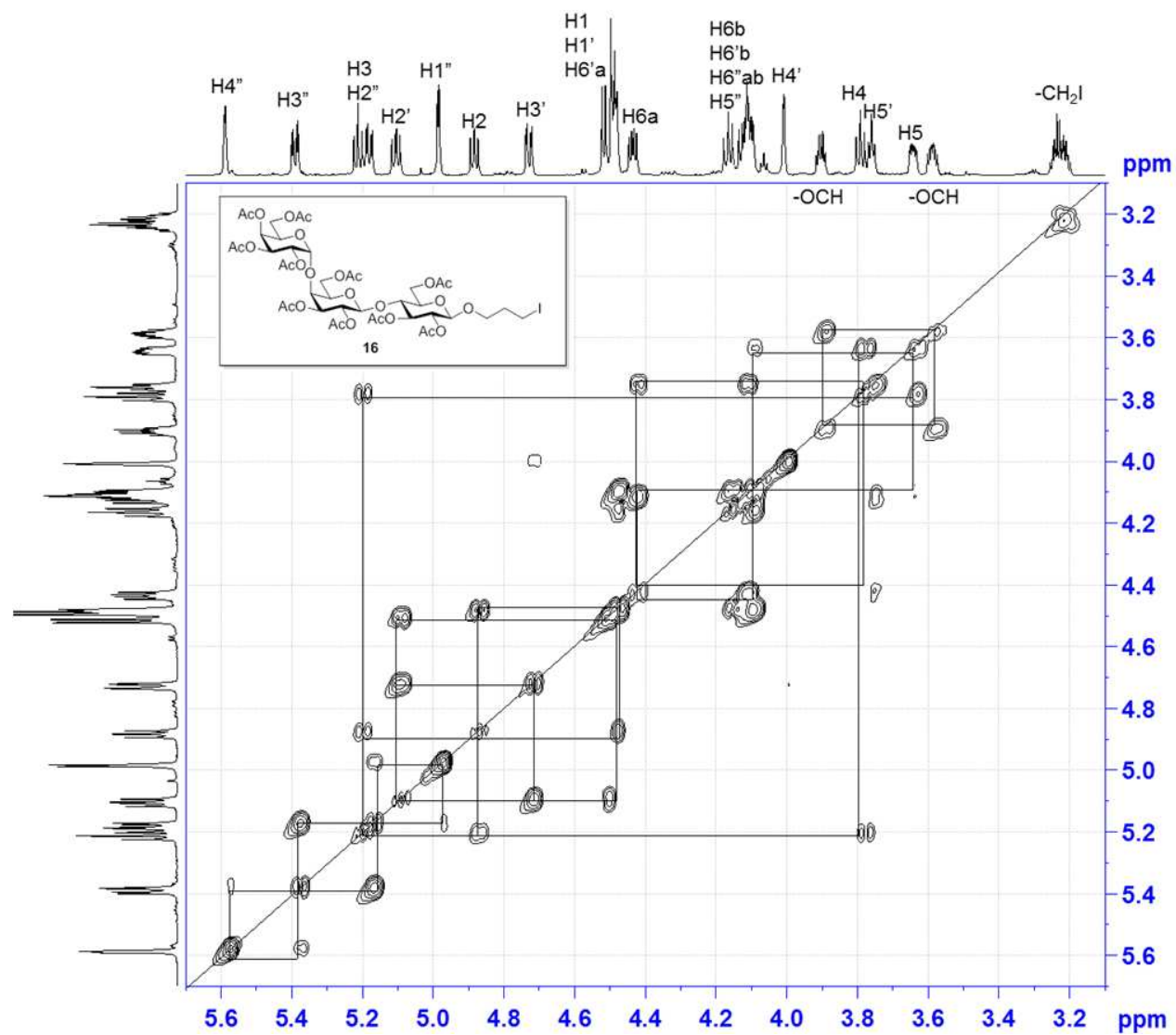




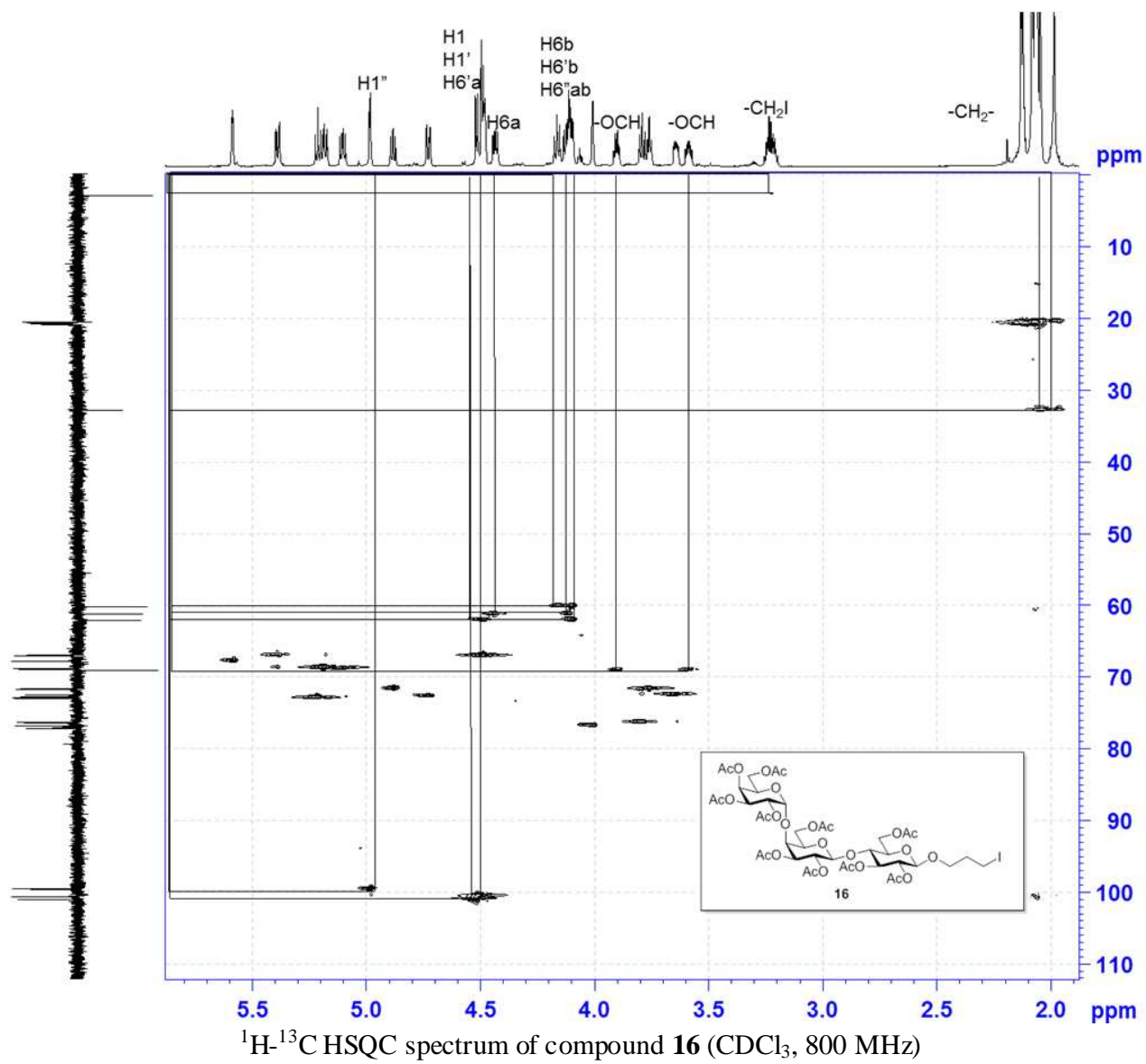
^1H NMR spectrum of compound **16** (CDCl_3 , 800 MHz)

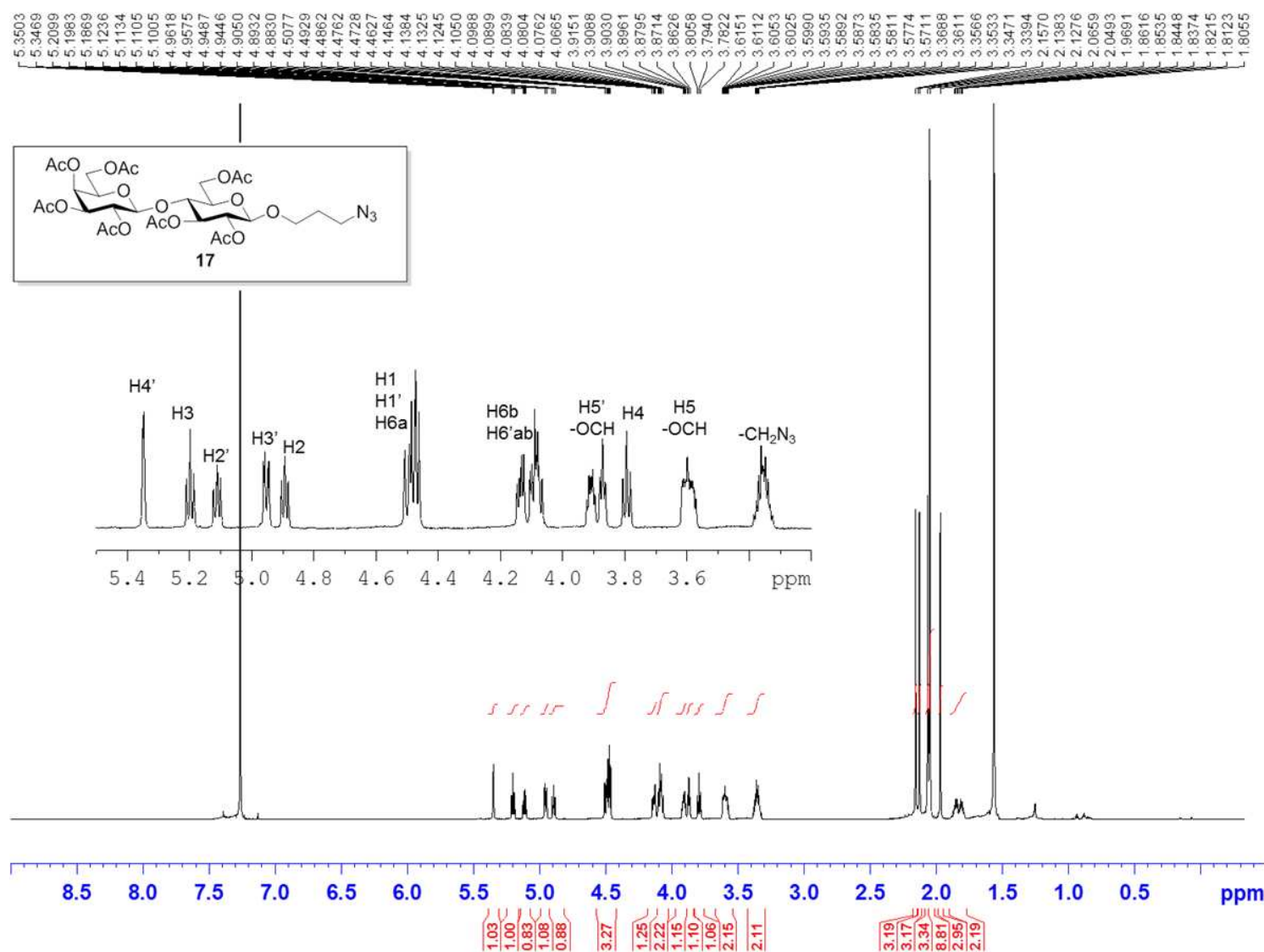


^{13}C and DEPT135 NMR spectrum of compound **16** (CDCl_3 , 200 MHz)

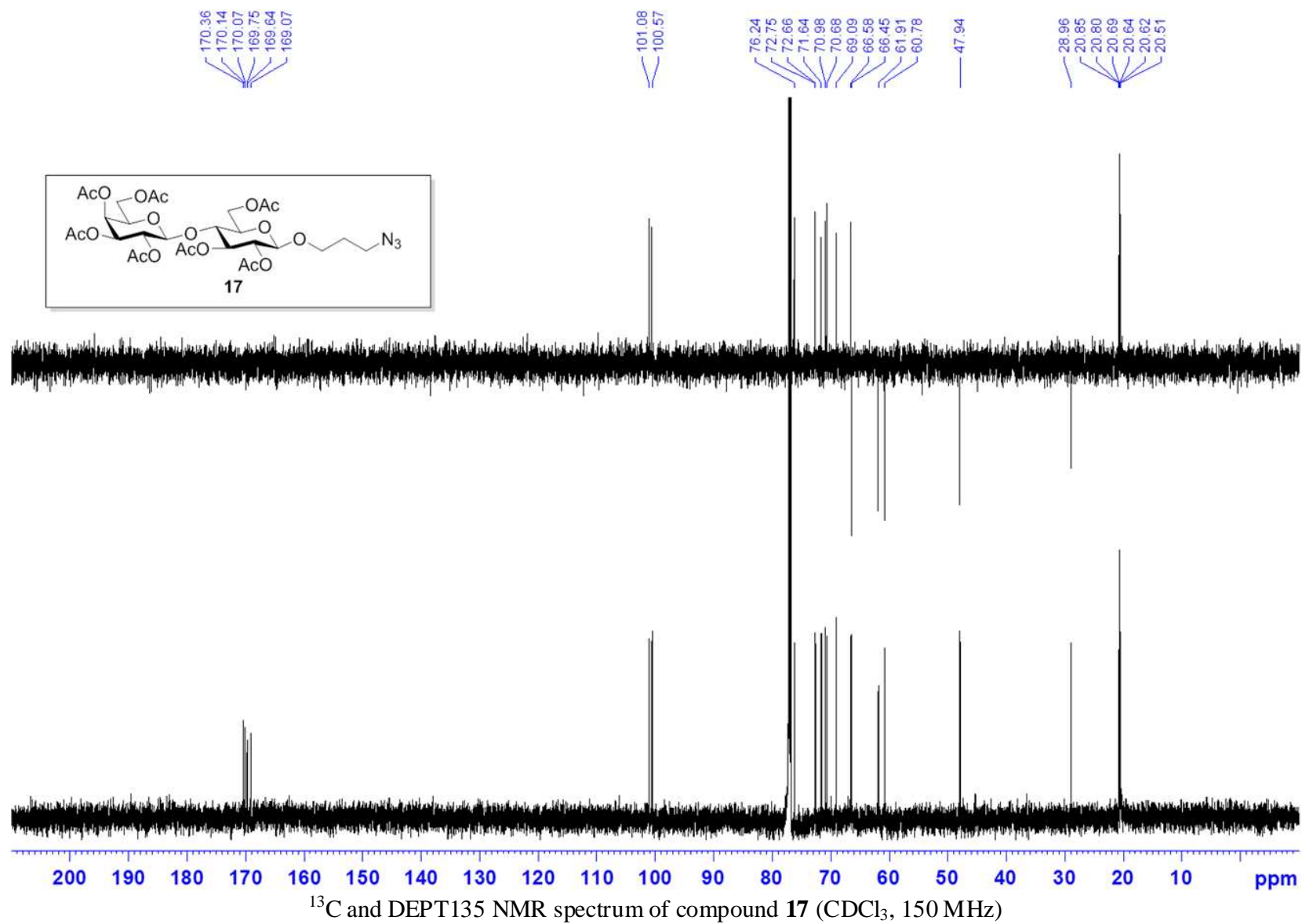


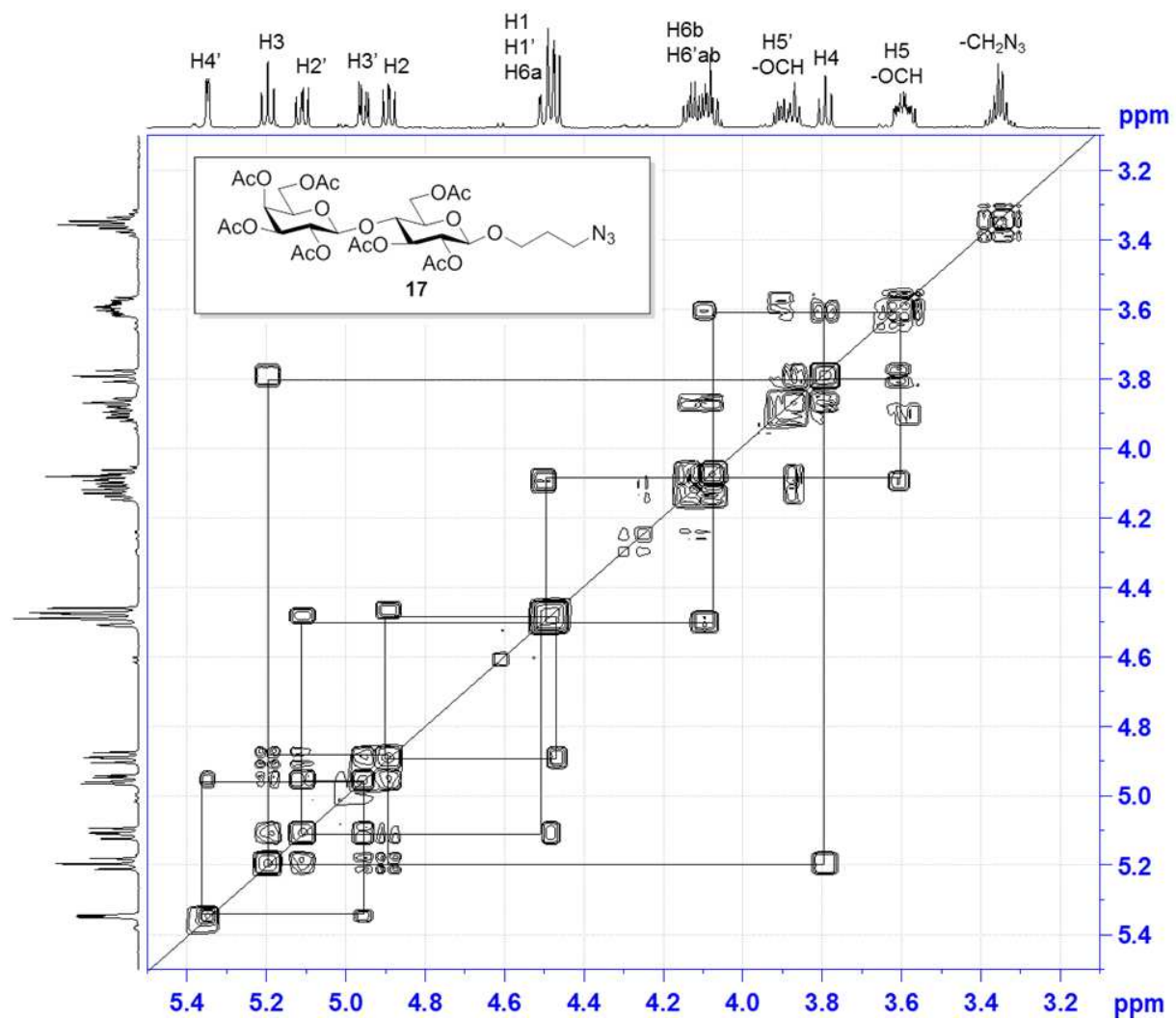
¹H-¹H COSY spectrum of compound **16** (CDCl₃, 800 MHz)



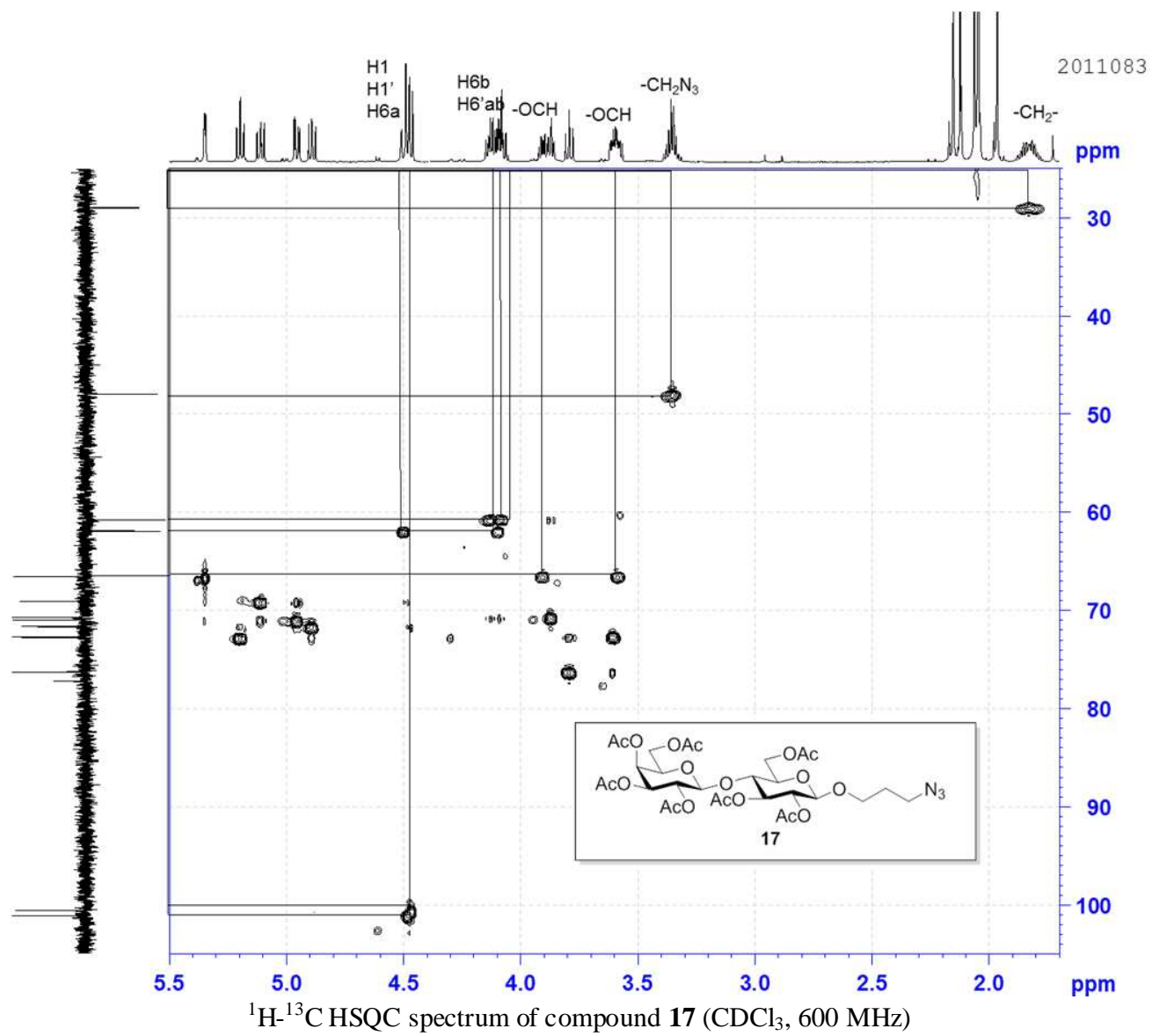


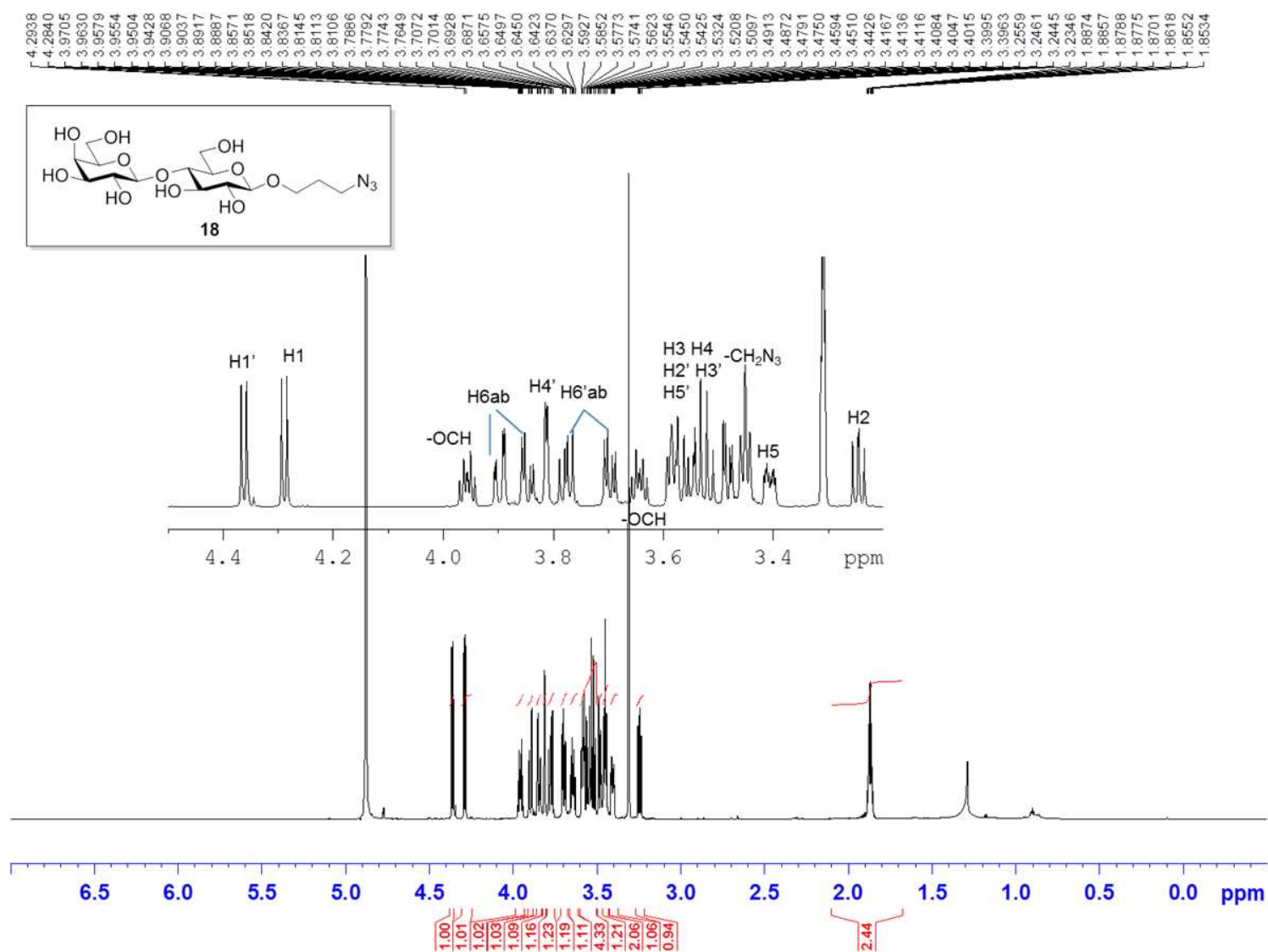
^1H NMR spectrum of compound **17** (CDCl_3 , 600 MHz)



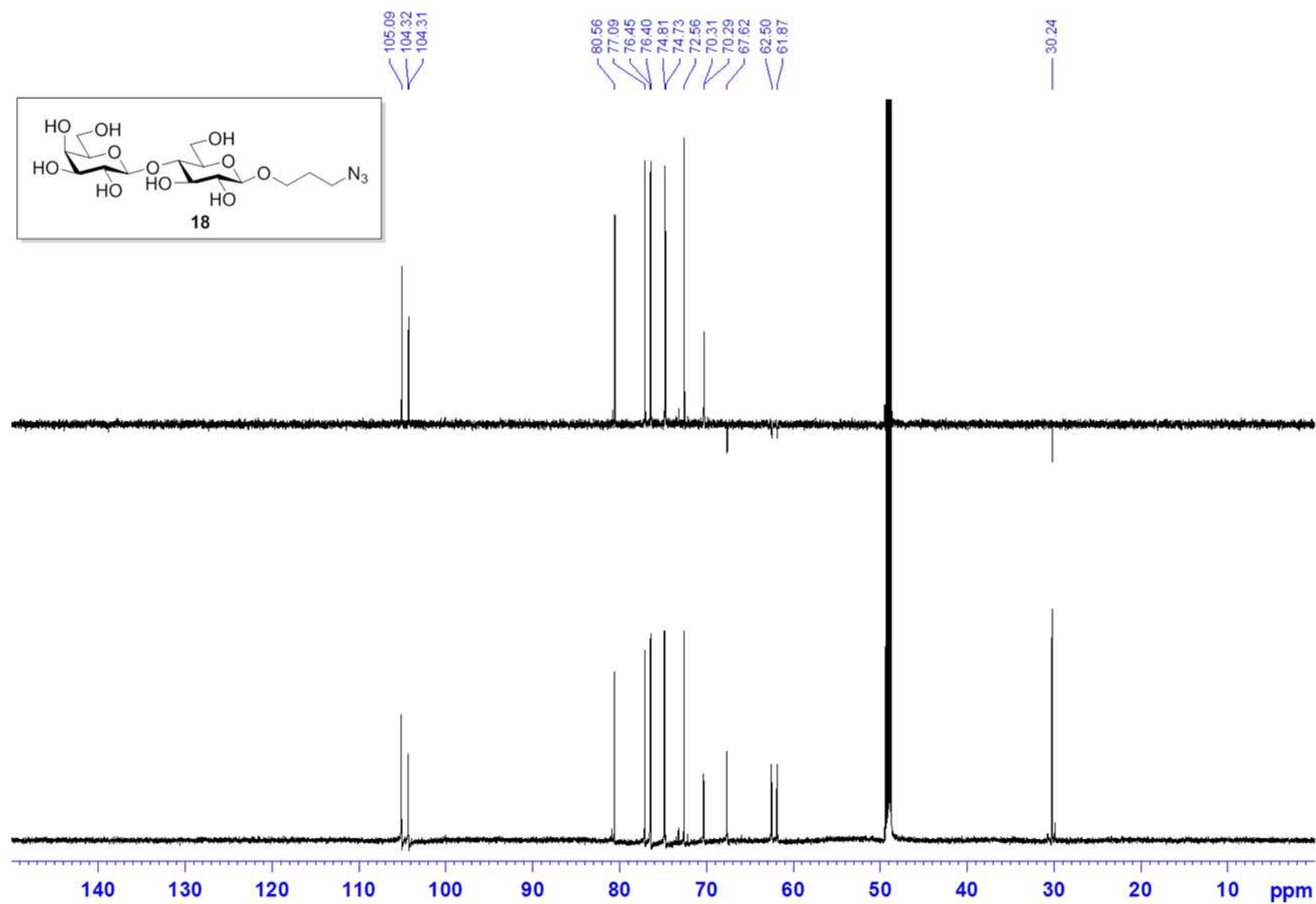


^1H - ^1H COSY spectrum of compound **17** (CDCl_3 , 600 MHz)

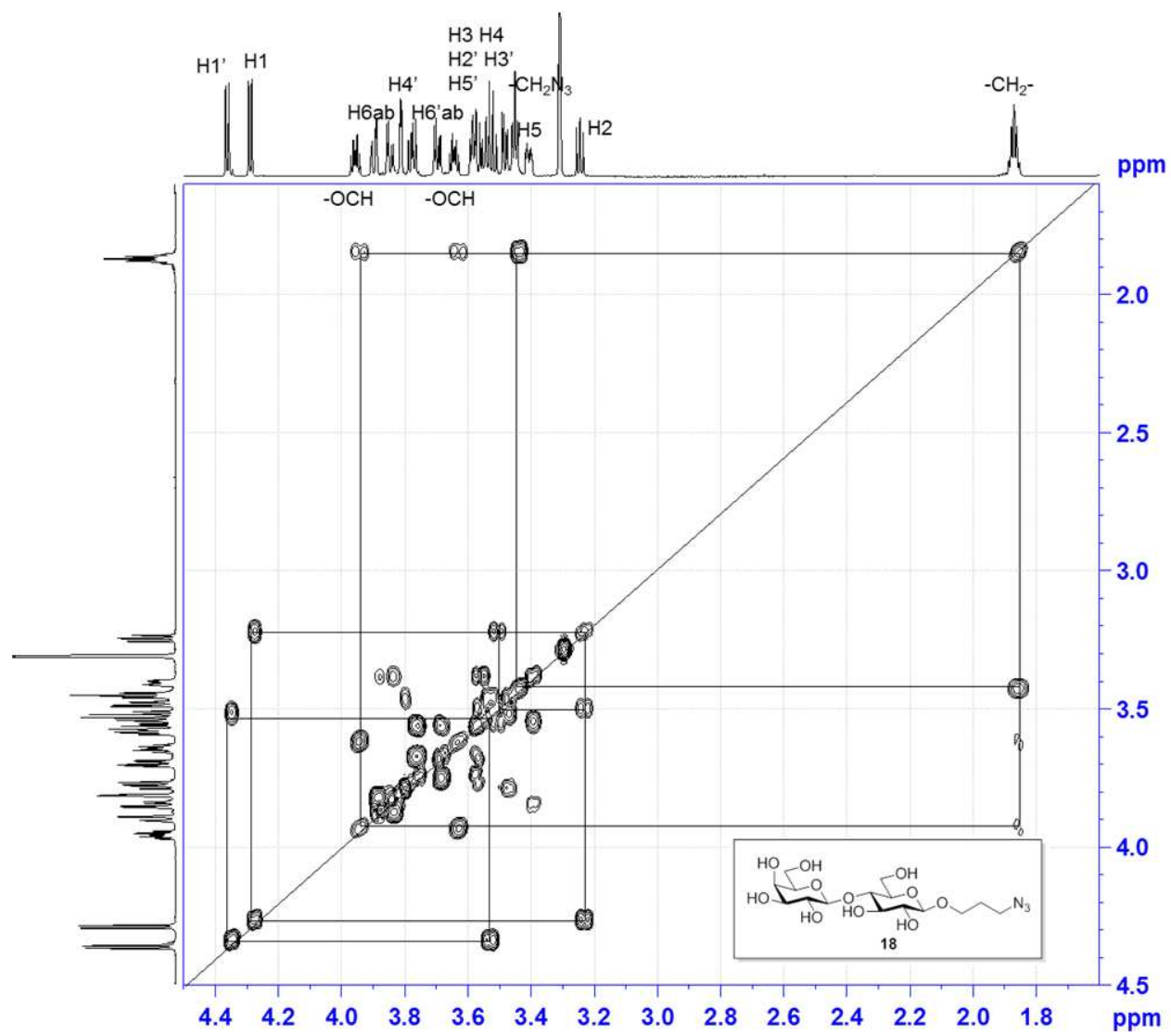




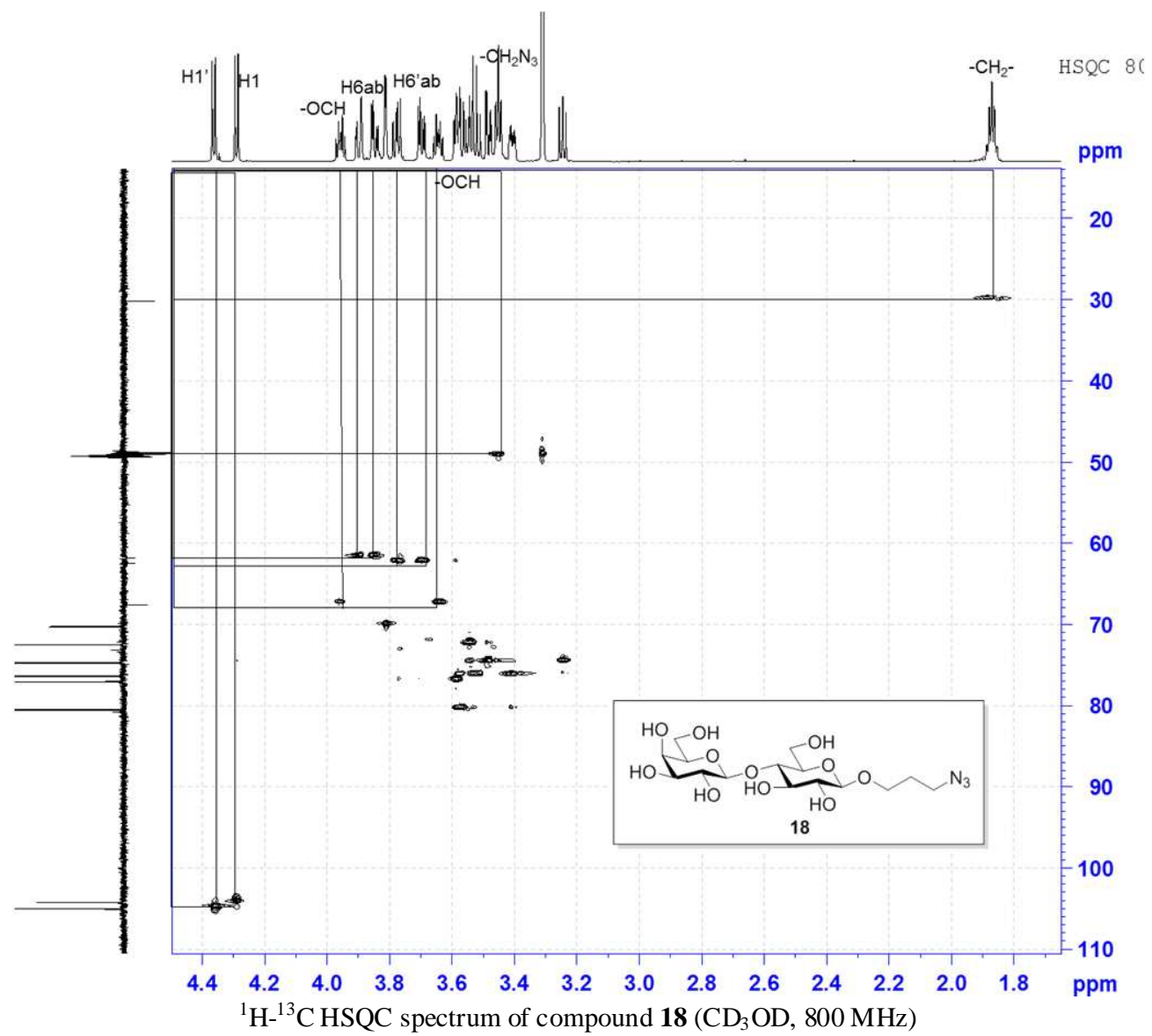
¹H NMR spectrum of compound **18** (CD₃OD, 800 MHz)

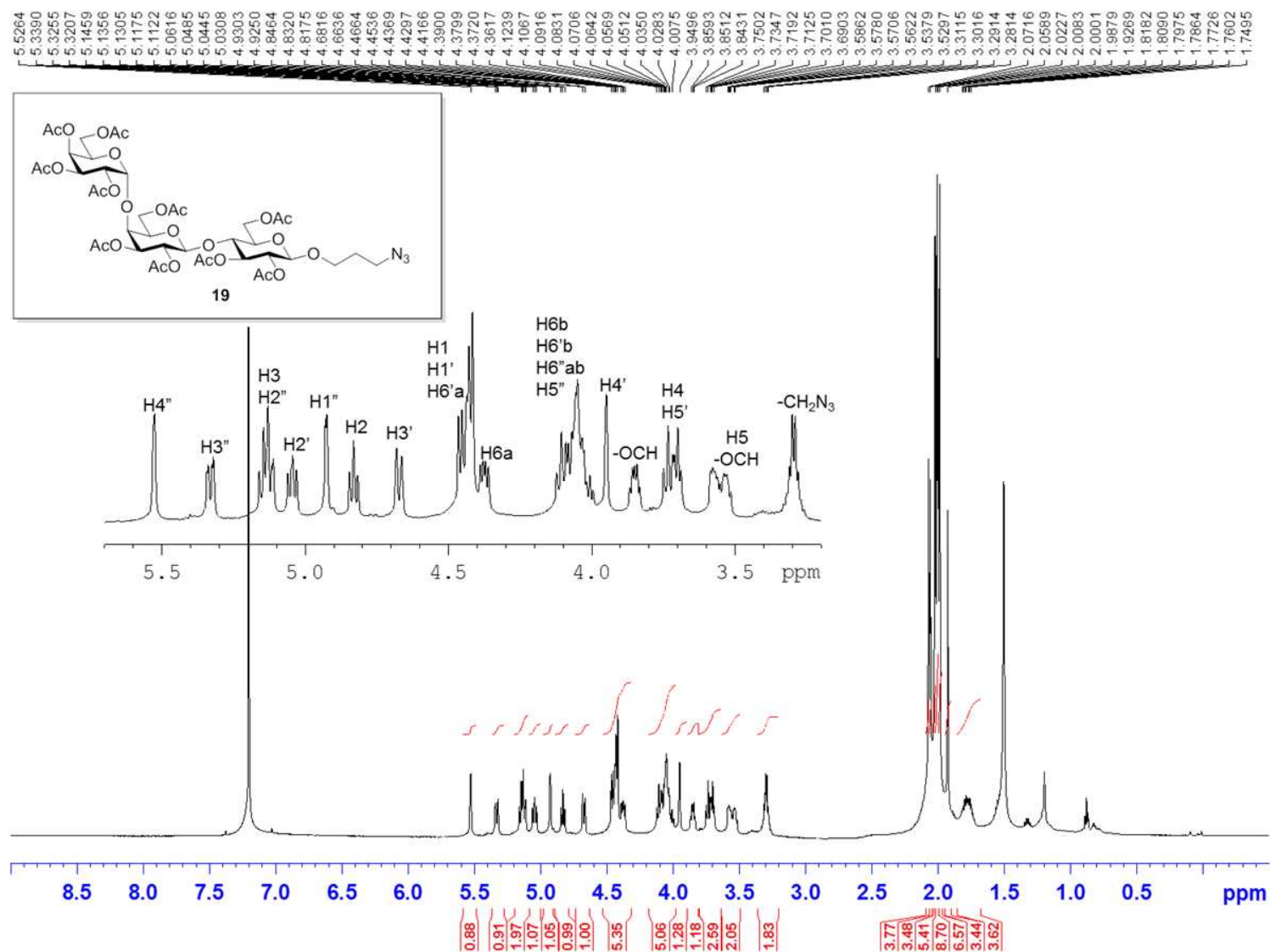


¹³C and DEPT135 NMR spectrum of compound **18** (CD₃OD, 200 MHz)

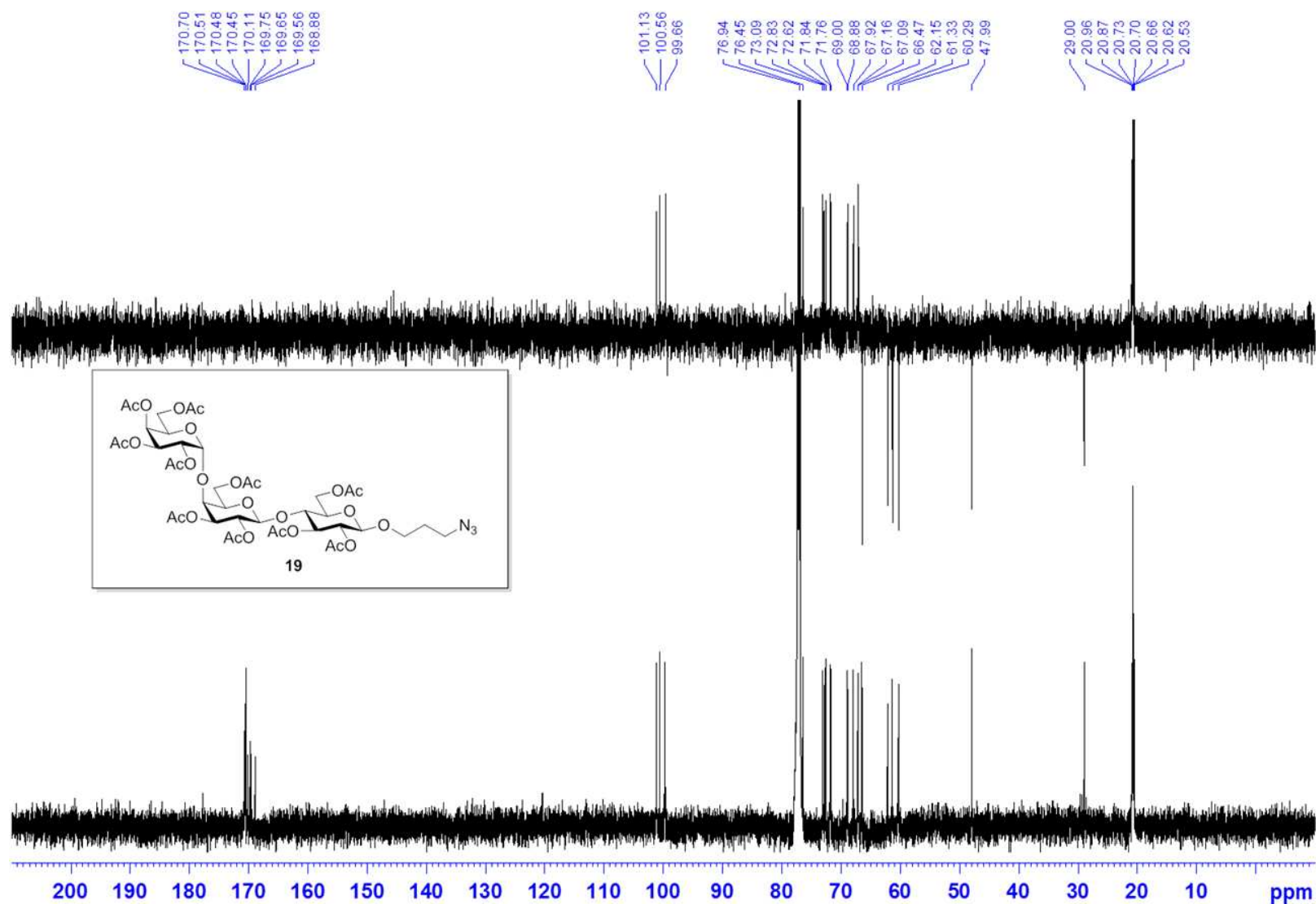


^1H - ^1H COSY spectrum of compound **18** (CD_3OD , 800 MHz)

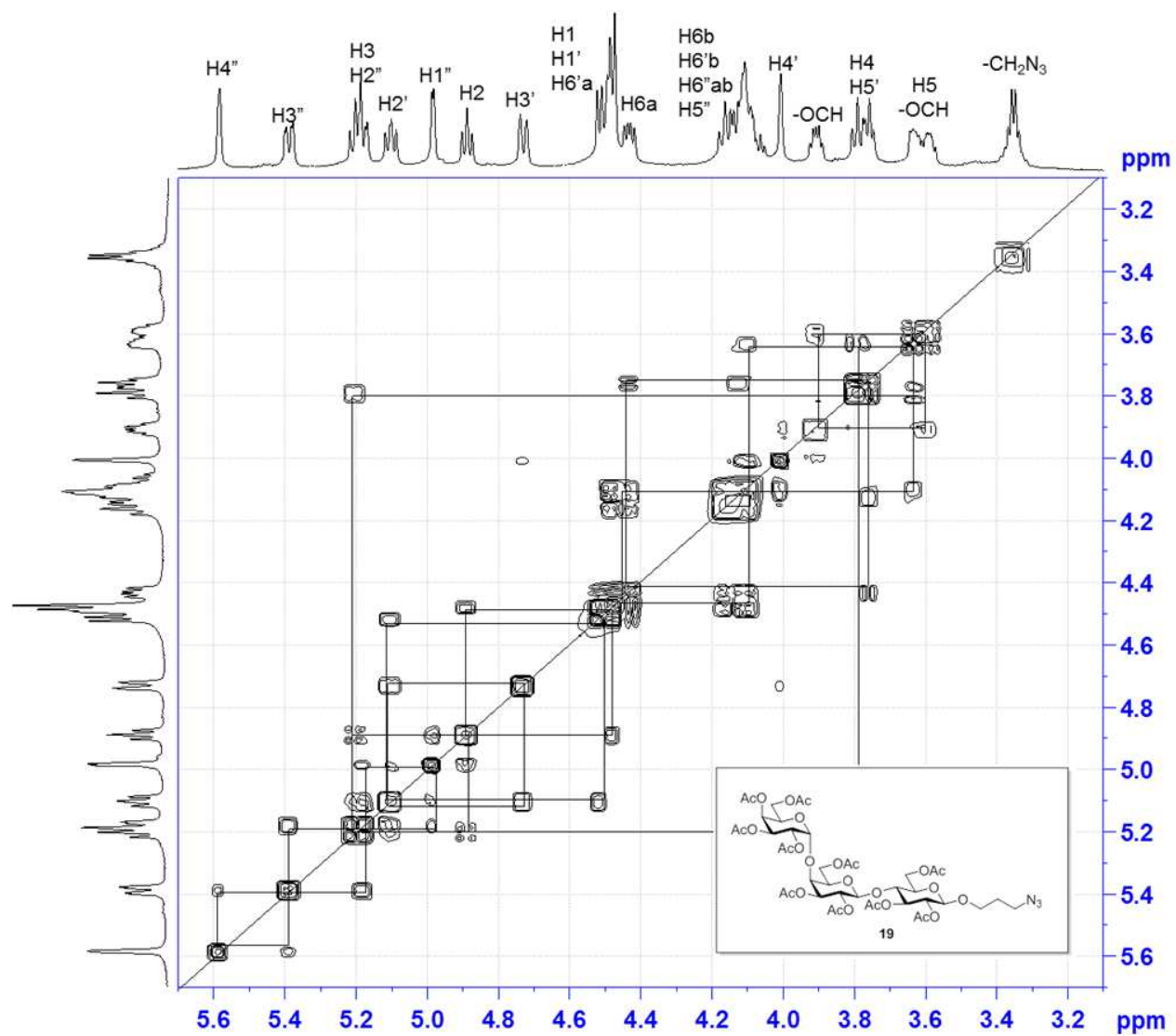




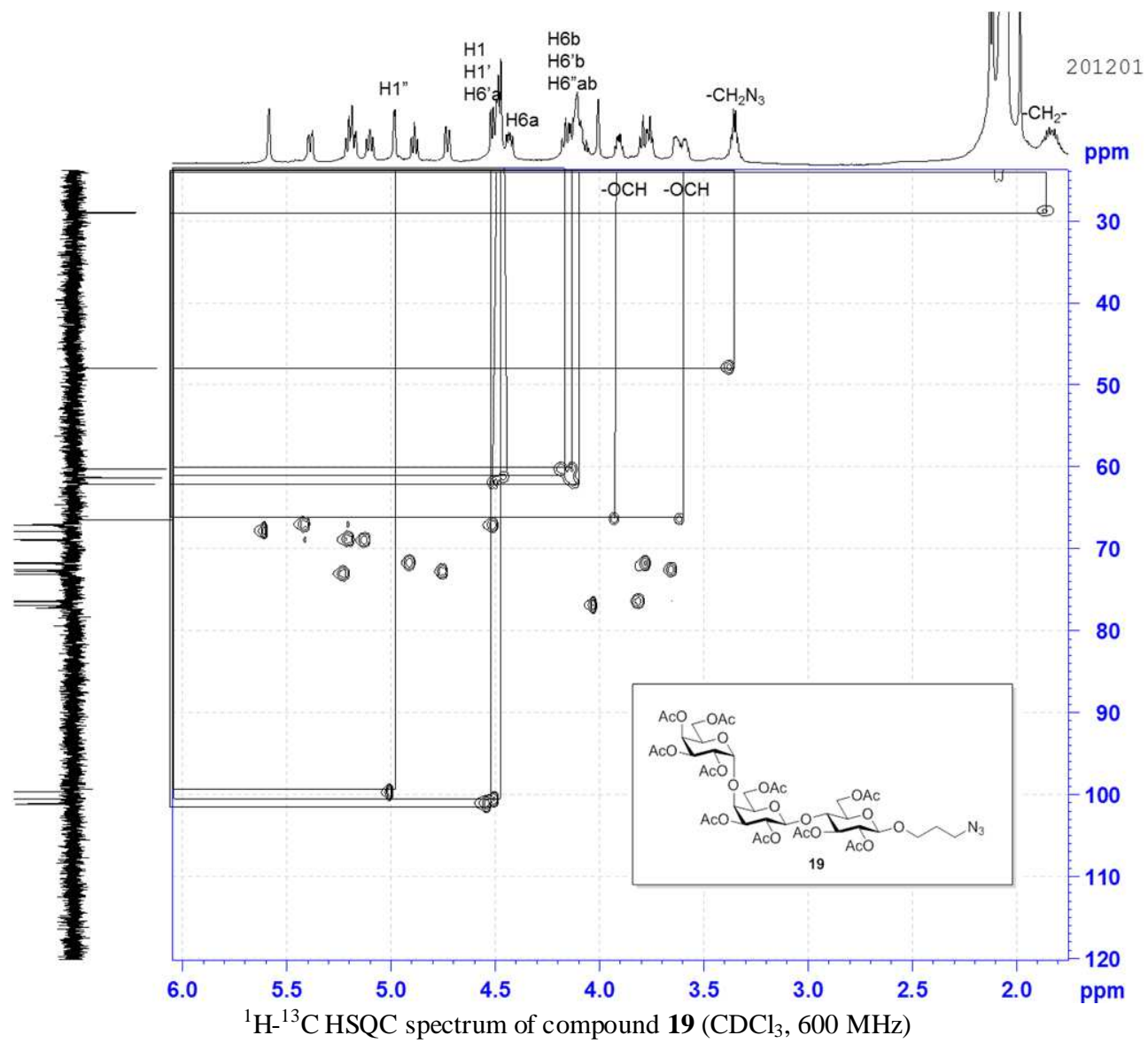
¹H NMR spectrum of compound **19** (CDCl₃, 600 MHz)

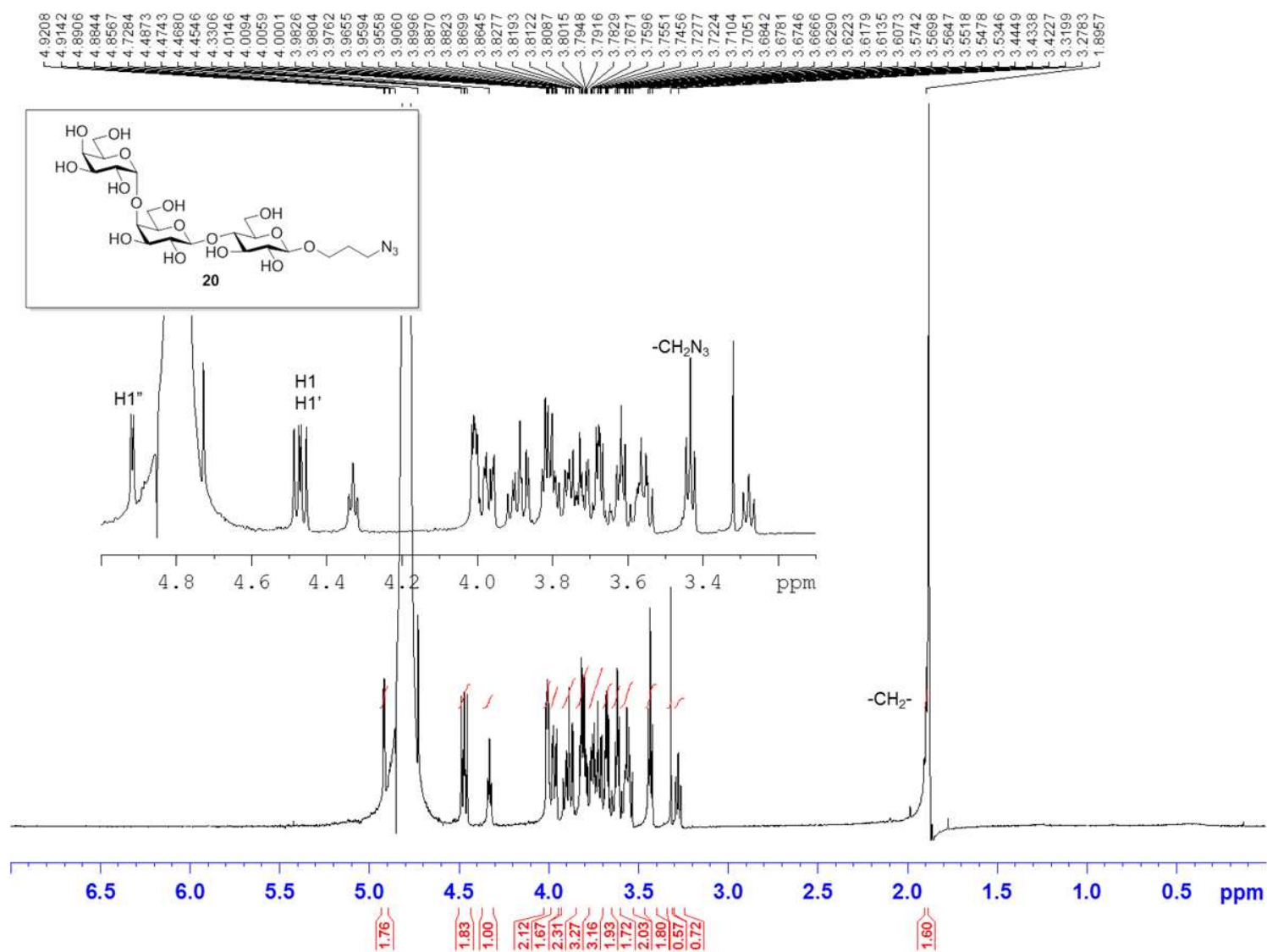


¹³C and DEPT135 NMR spectrum of compound **19** (CDCl₃, 150 MHz)

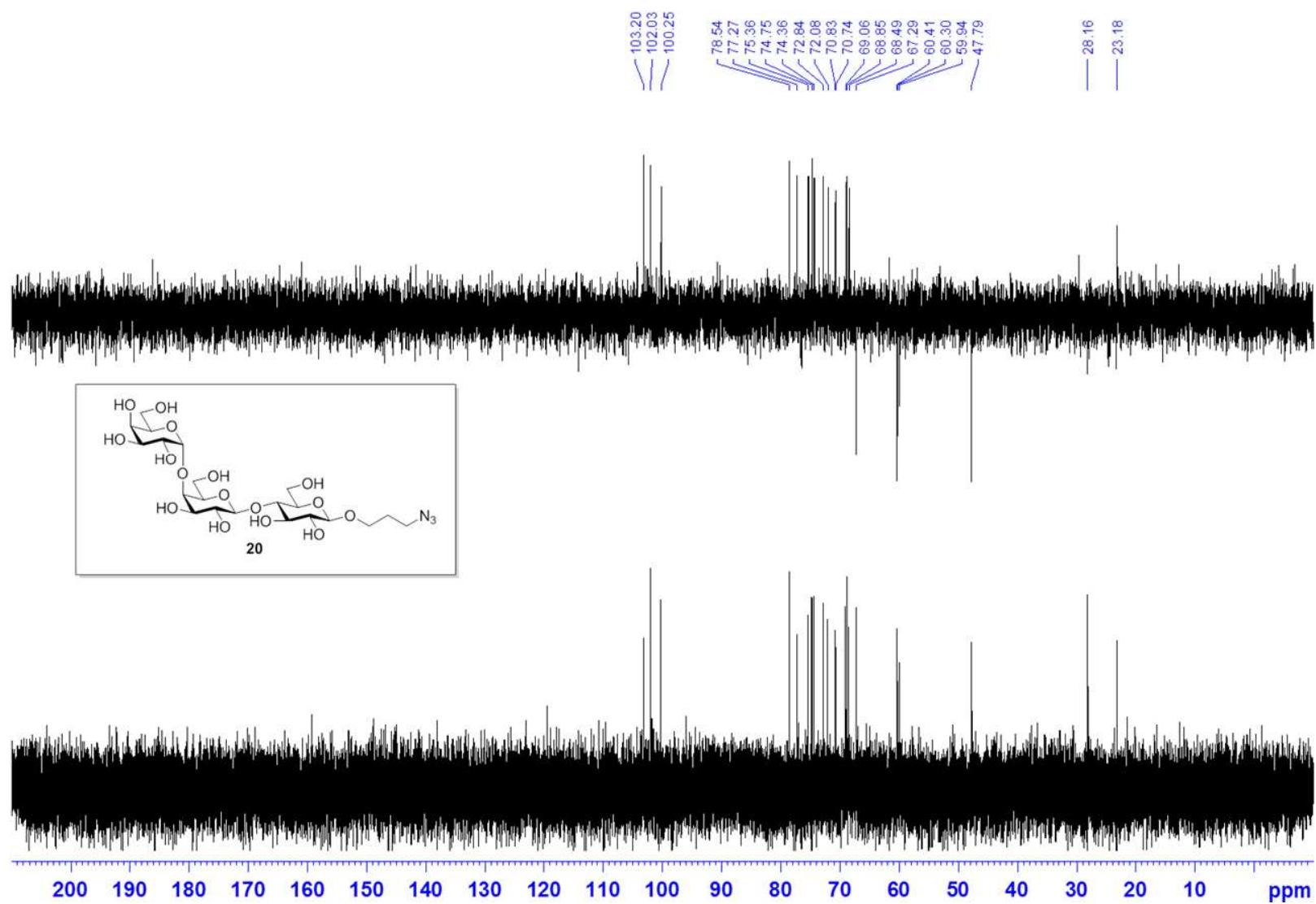


^1H - ^1H COSY spectrum of compound **19** (CDCl_3 , 600 MHz)

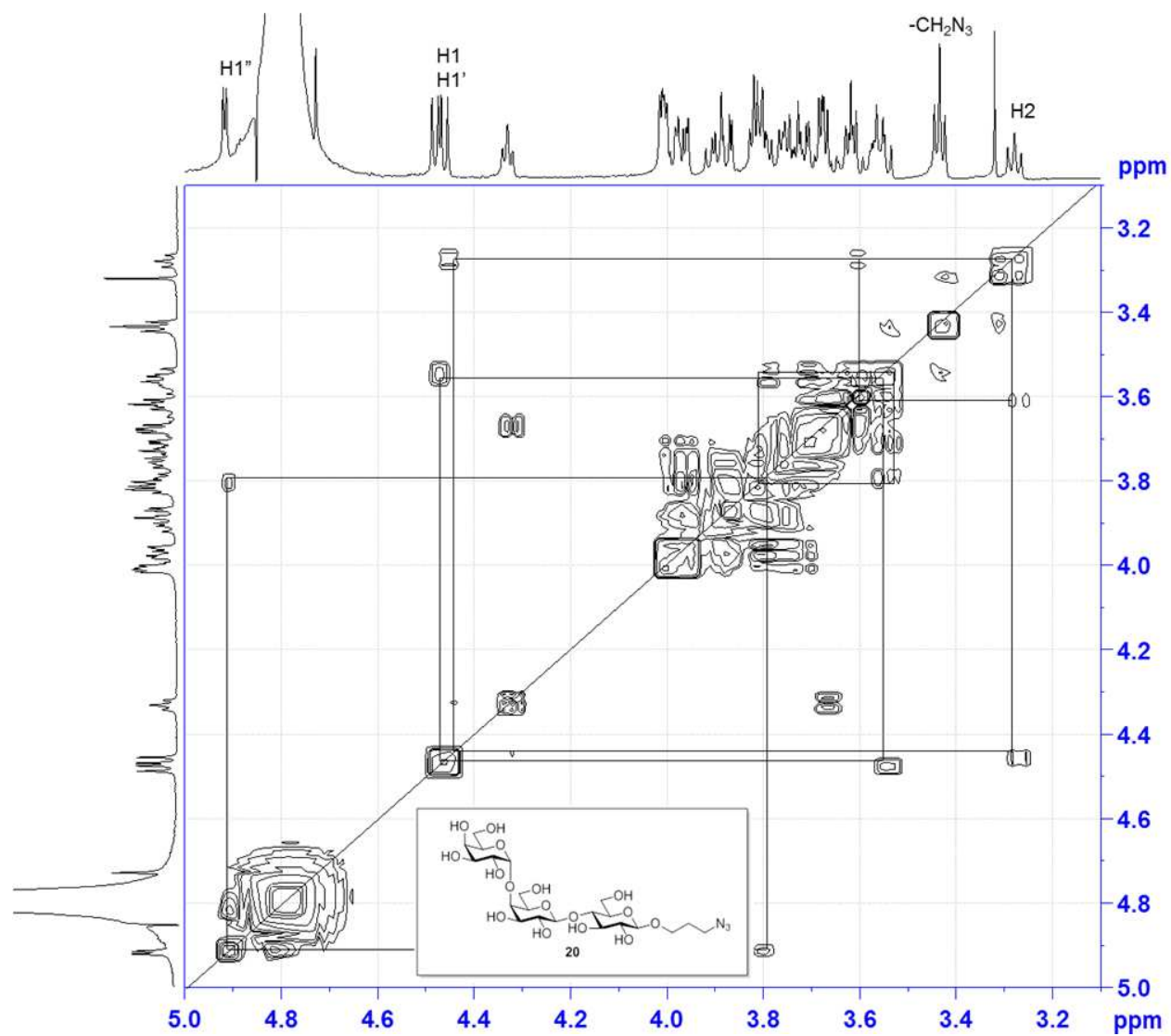




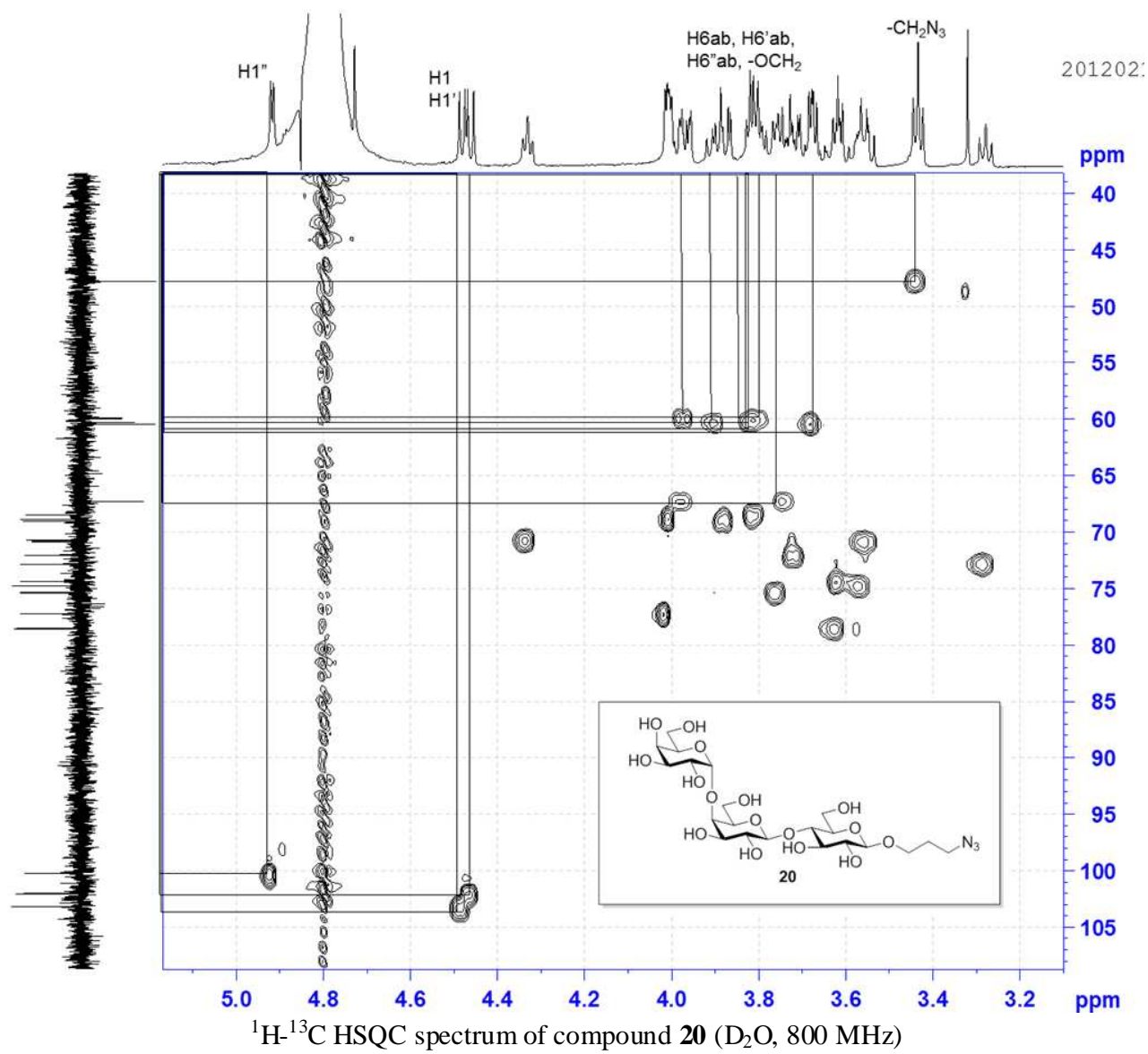
^1H NMR spectrum of compound **20** (D_2O , 600 MHz)

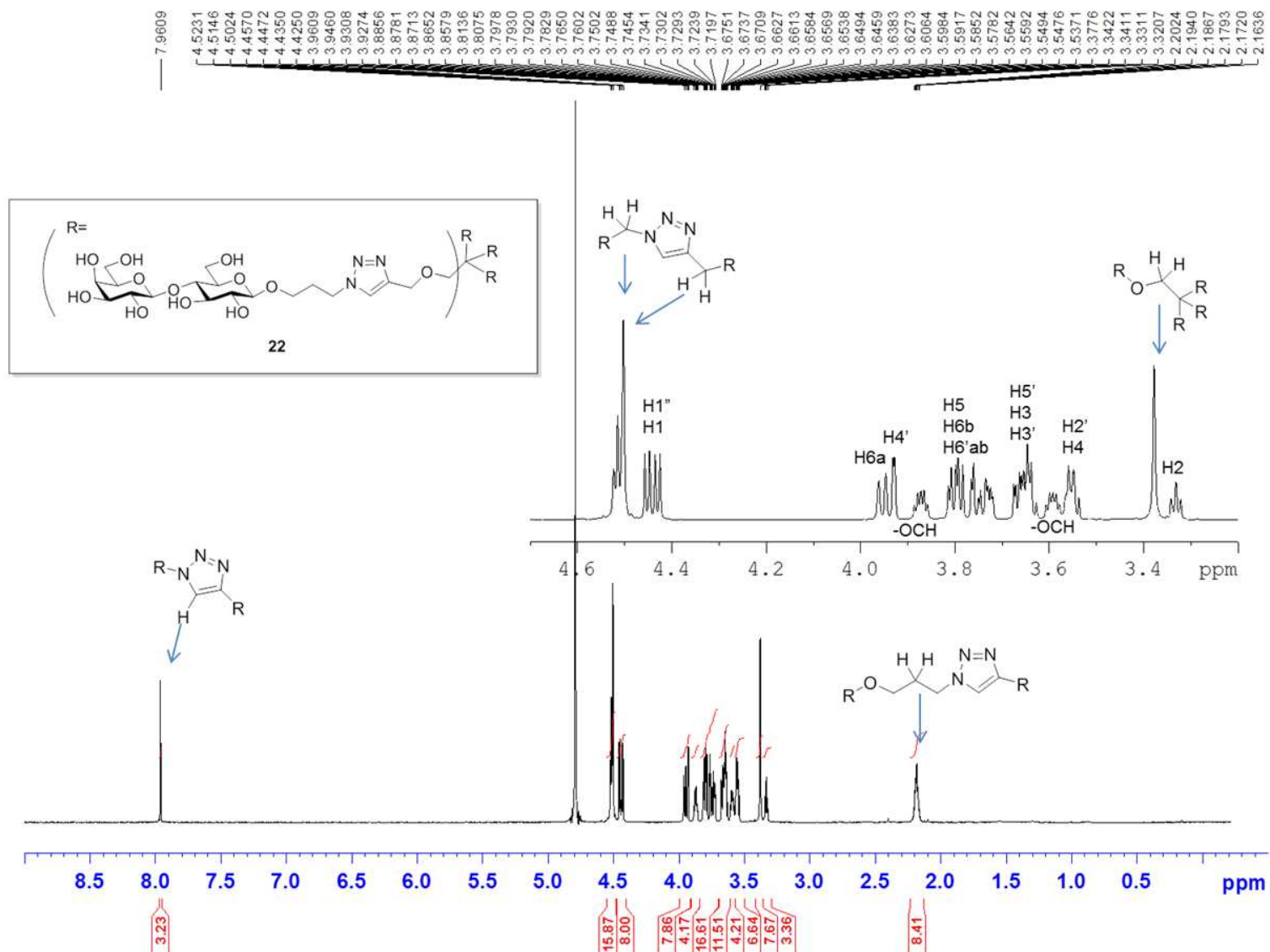


^{13}C and DEPT135 NMR spectrum of compound **20** (D_2O , 200 MHz)

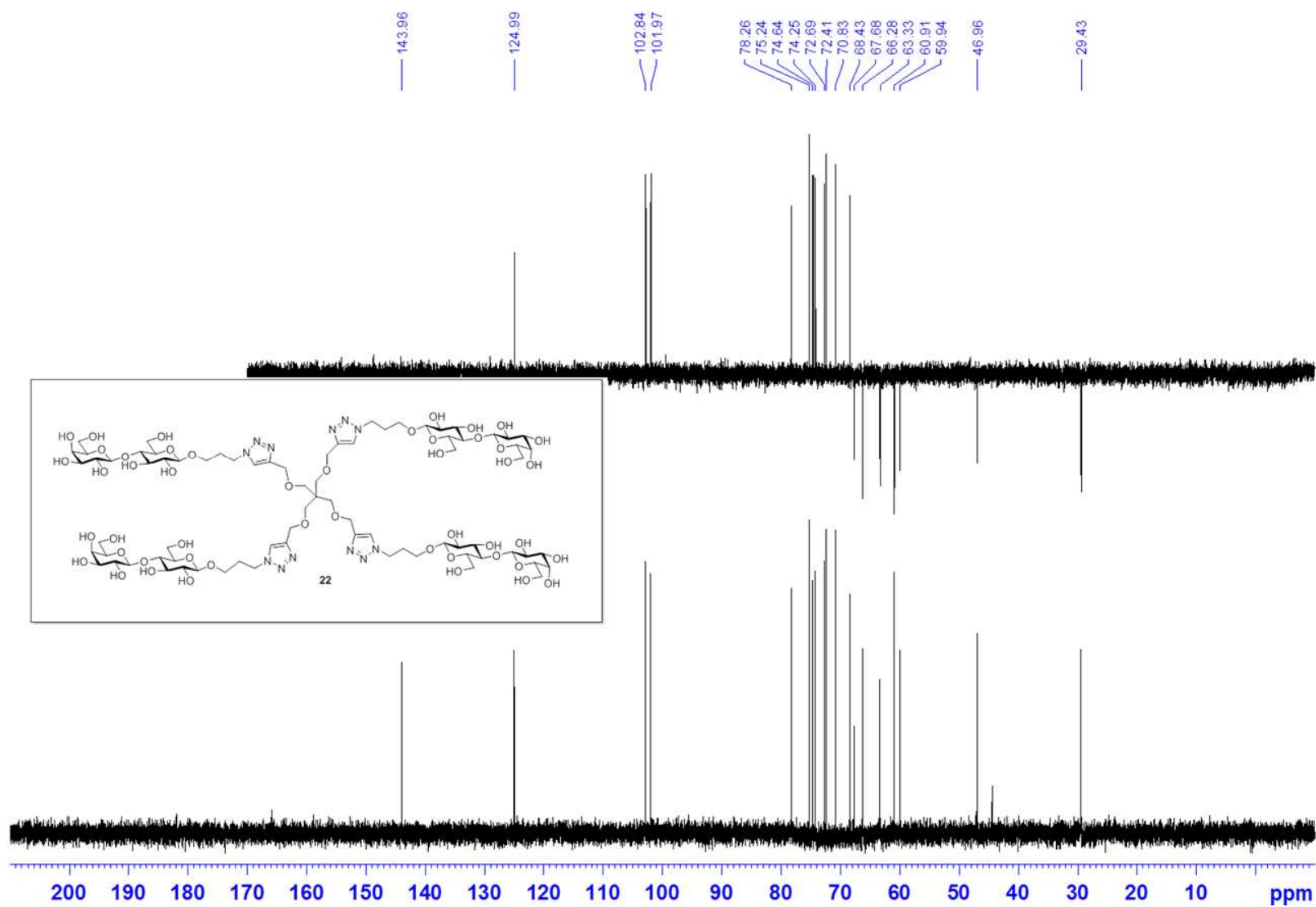


^1H - ^1H COSY spectrum of compound **20** (D_2O , 800 MHz)

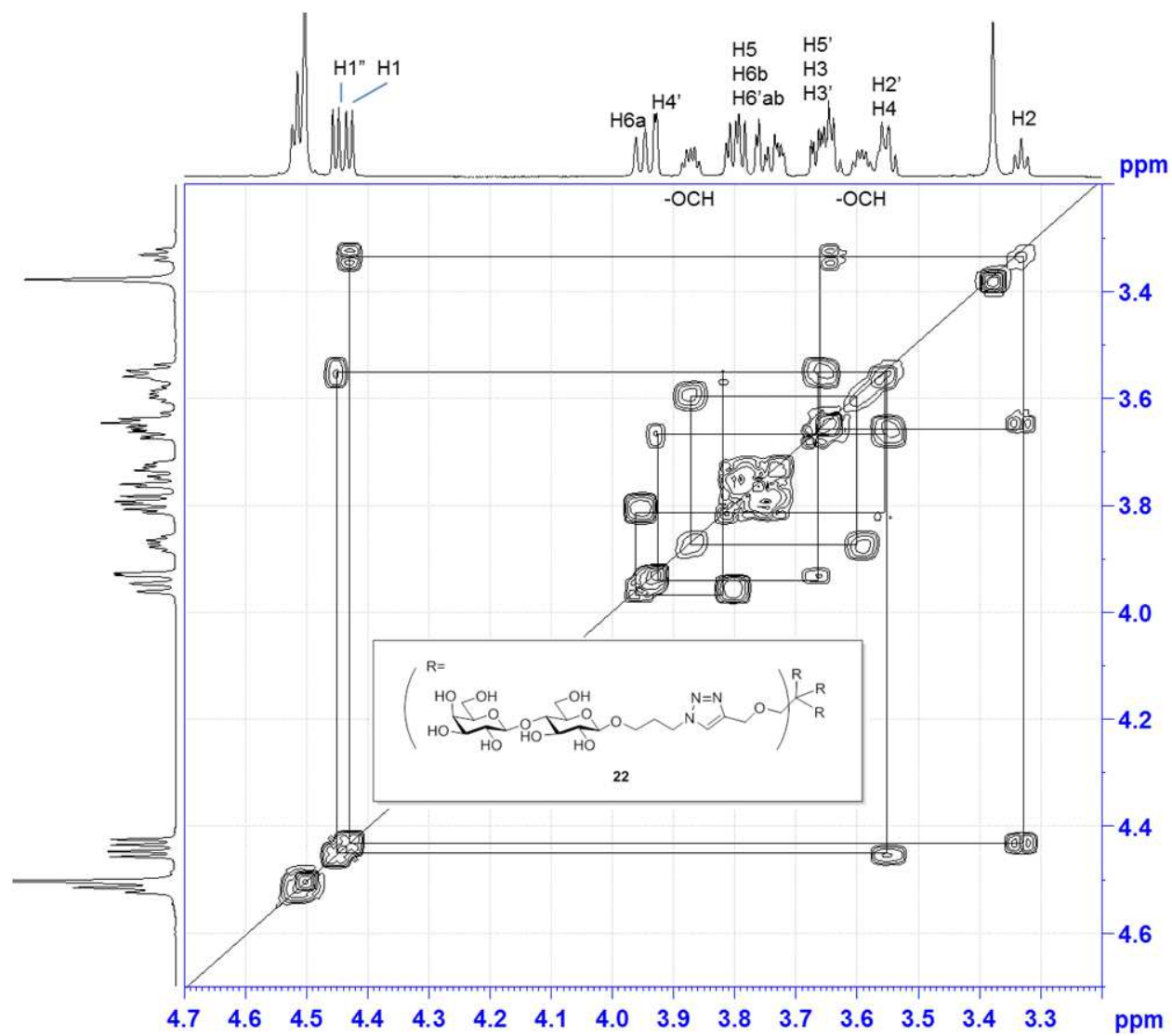




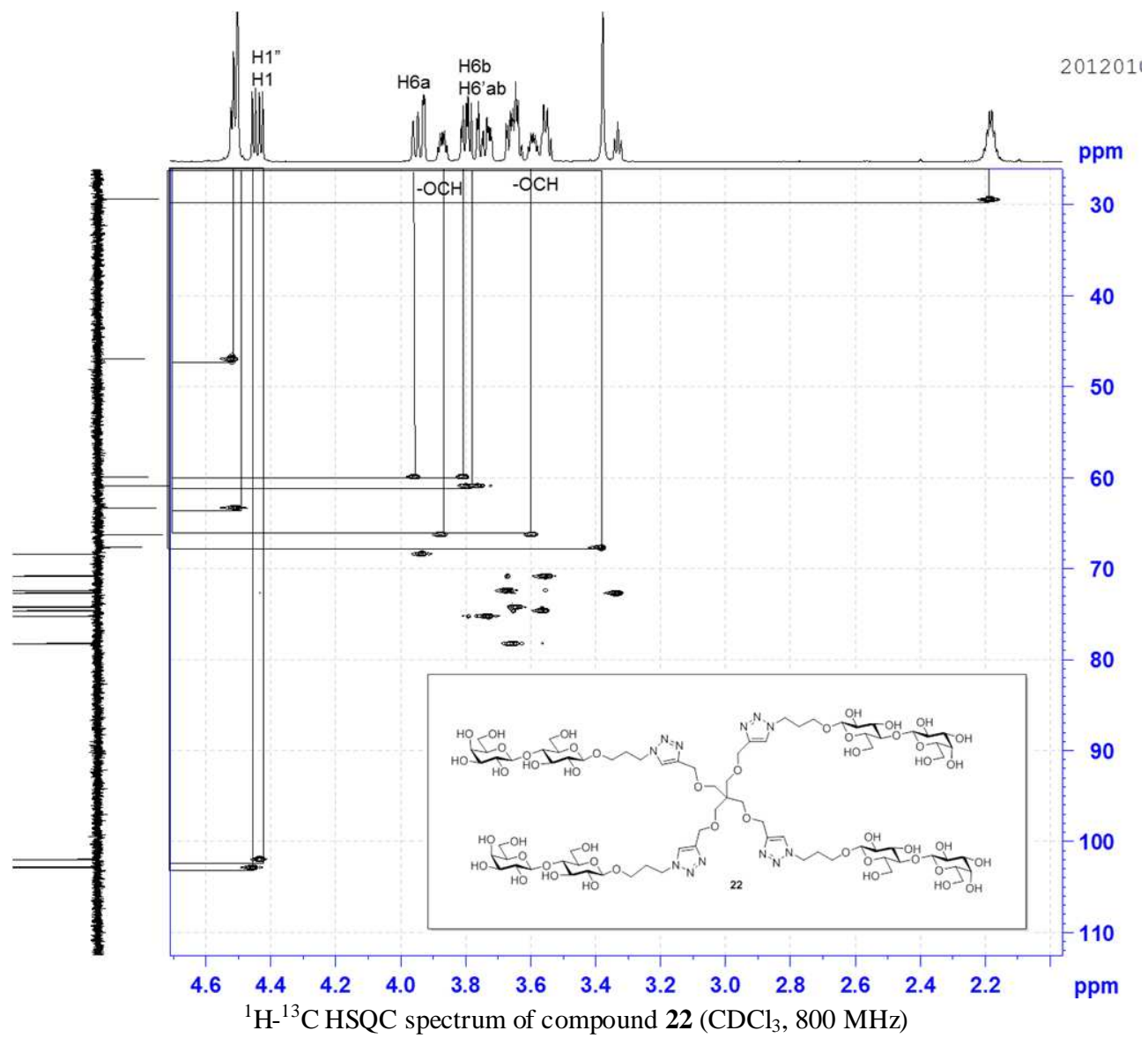
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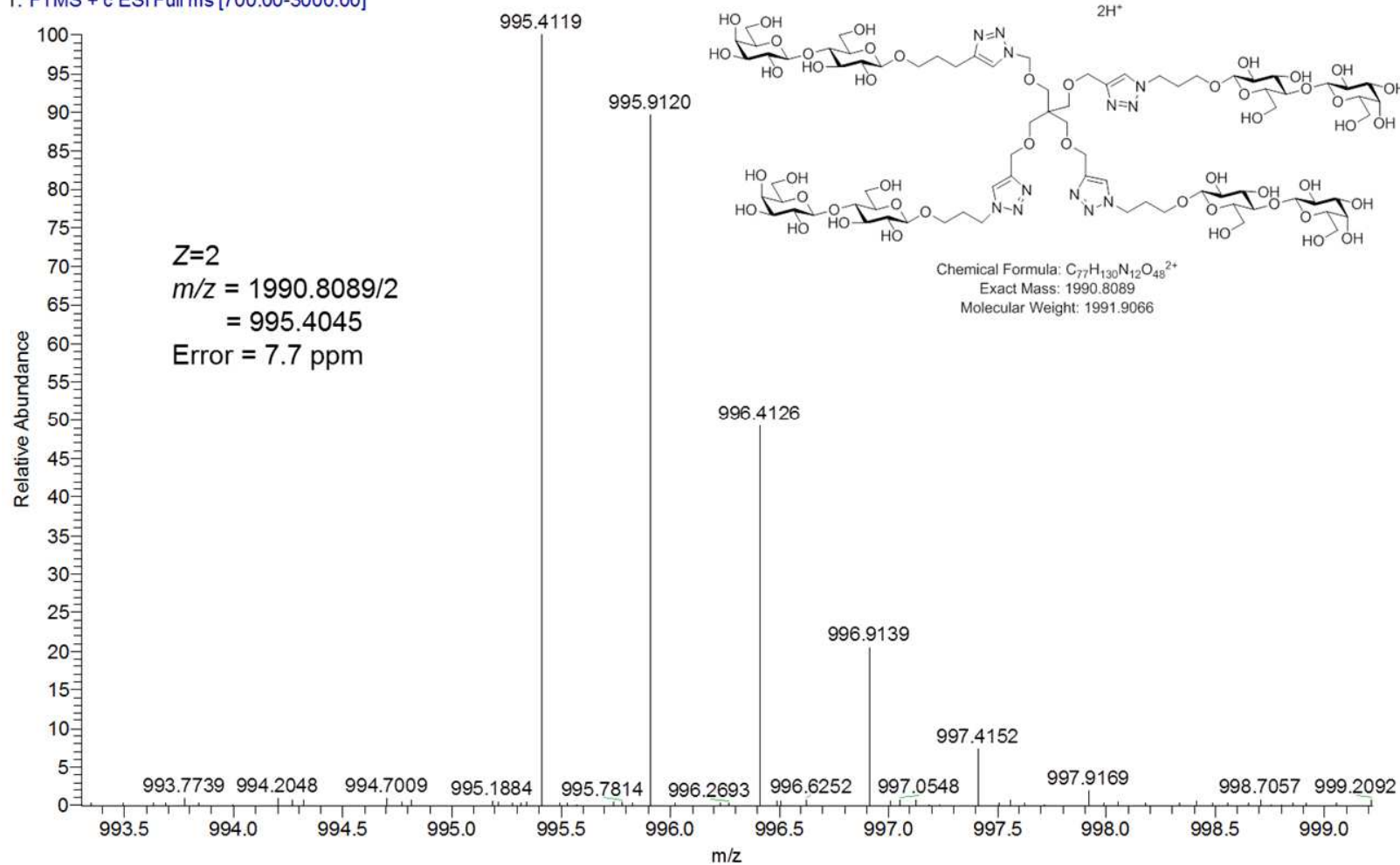
^{13}C and DEPT135 NMR spectrum of compound **22** (CDCl₃, 200 MHz)



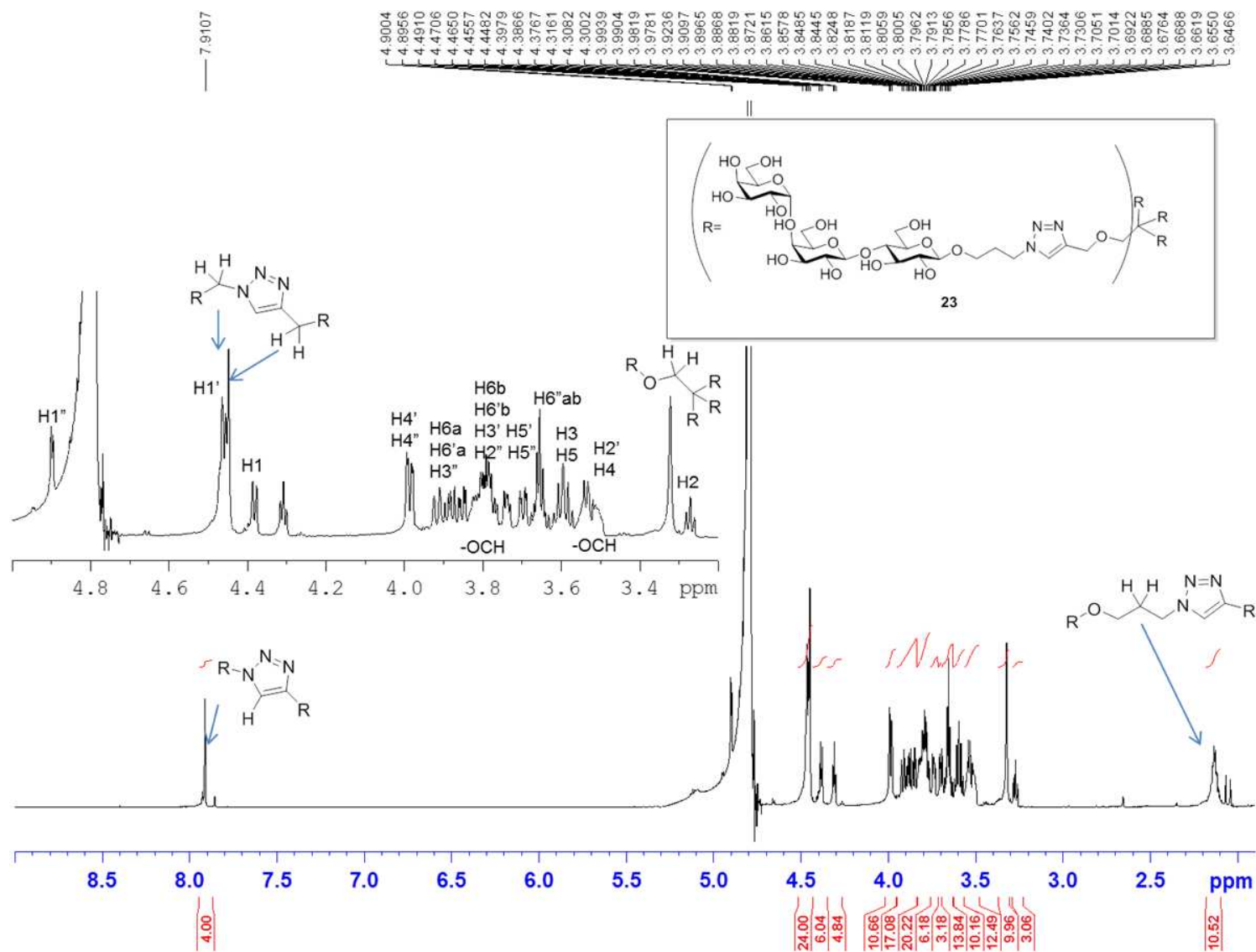
^1H - ^1H COSY spectrum of compound **22** (CDCl_3 , 800 MHz)



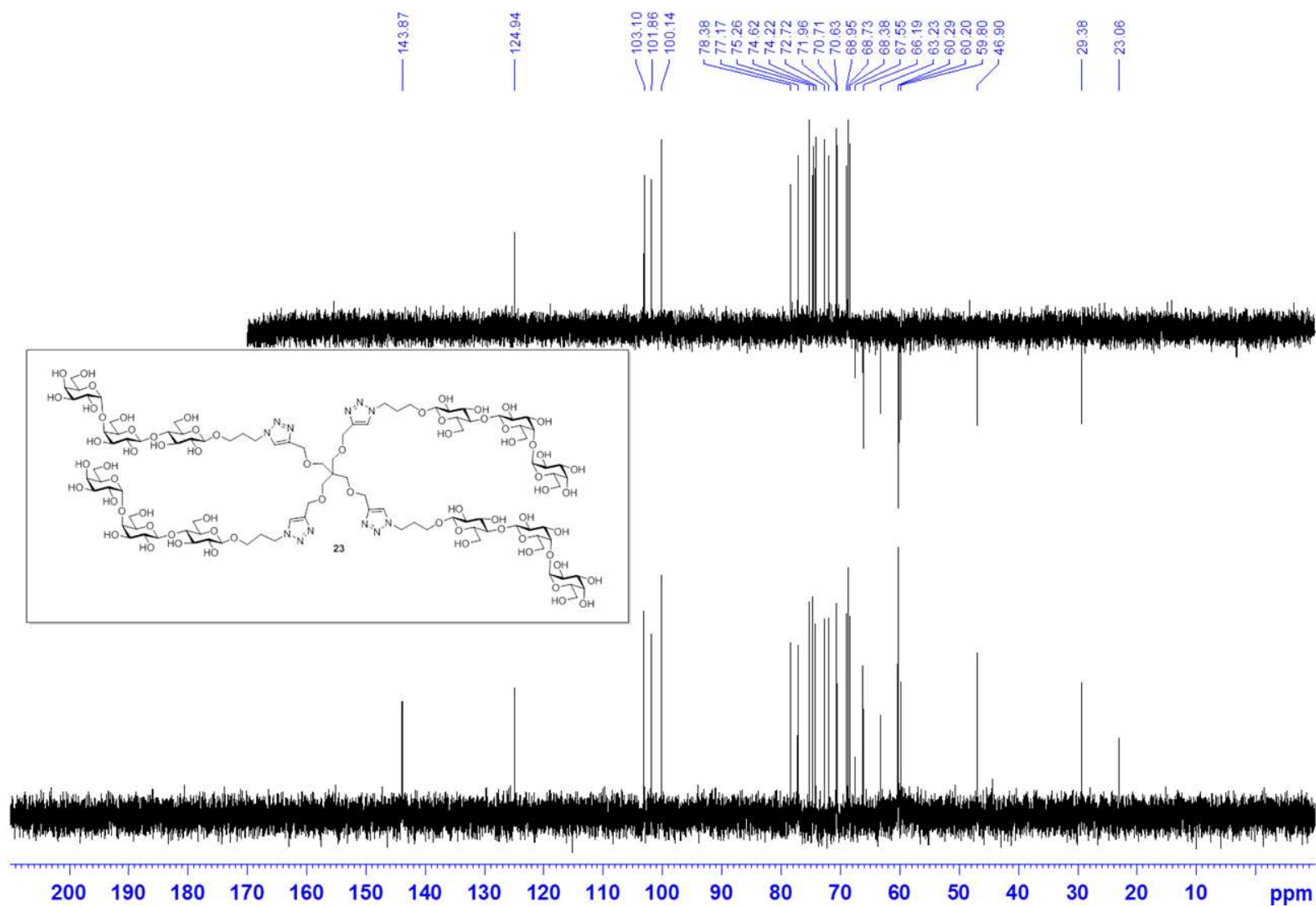
HW105 lac-tetramer_120416140401 #130 RT: 1.58 AV: 1 NL: 3.10E6
T: FTMS + c ESI Full ms [700.00-3000.00]



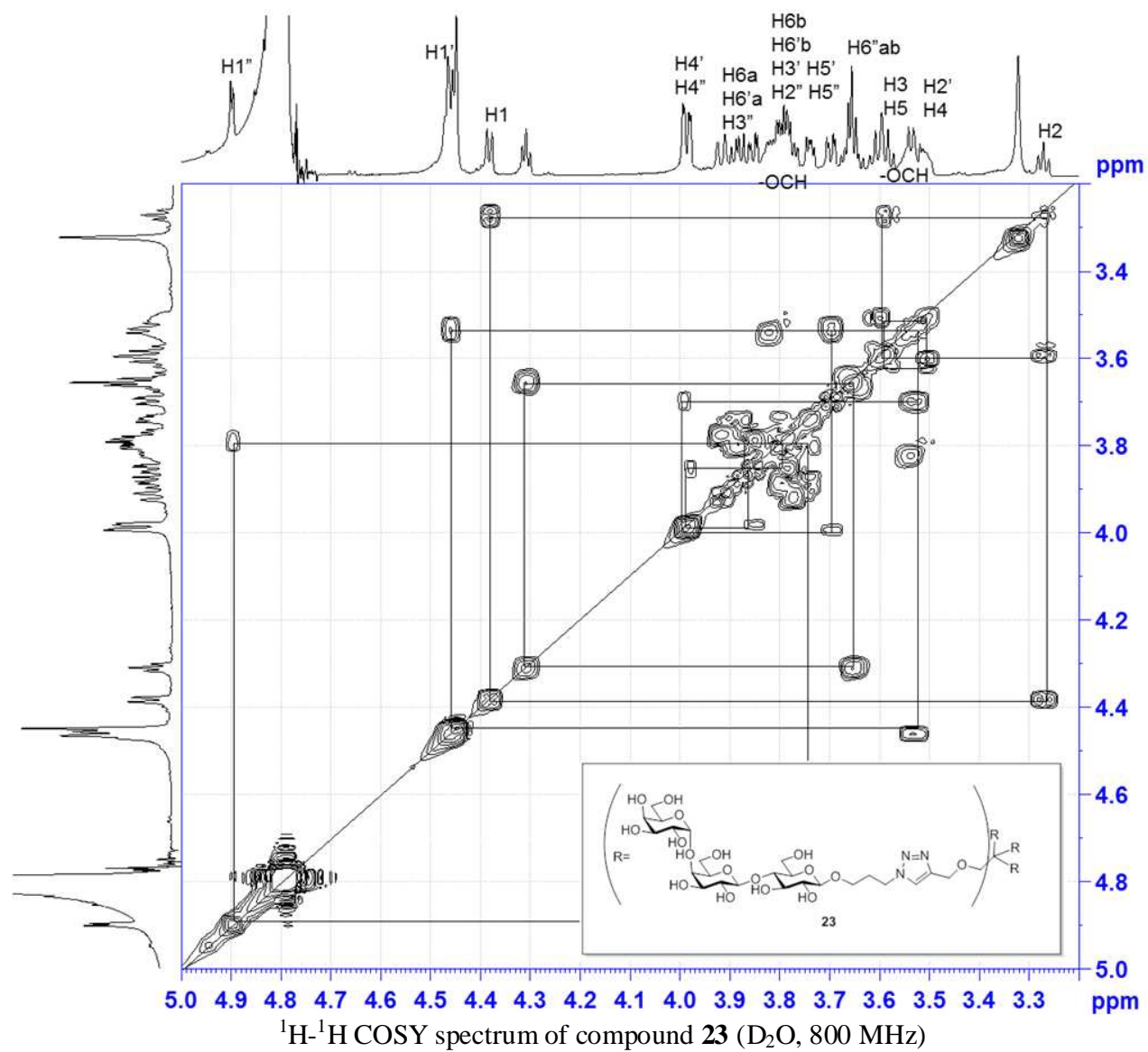
ESI-HRMS of compound **22**

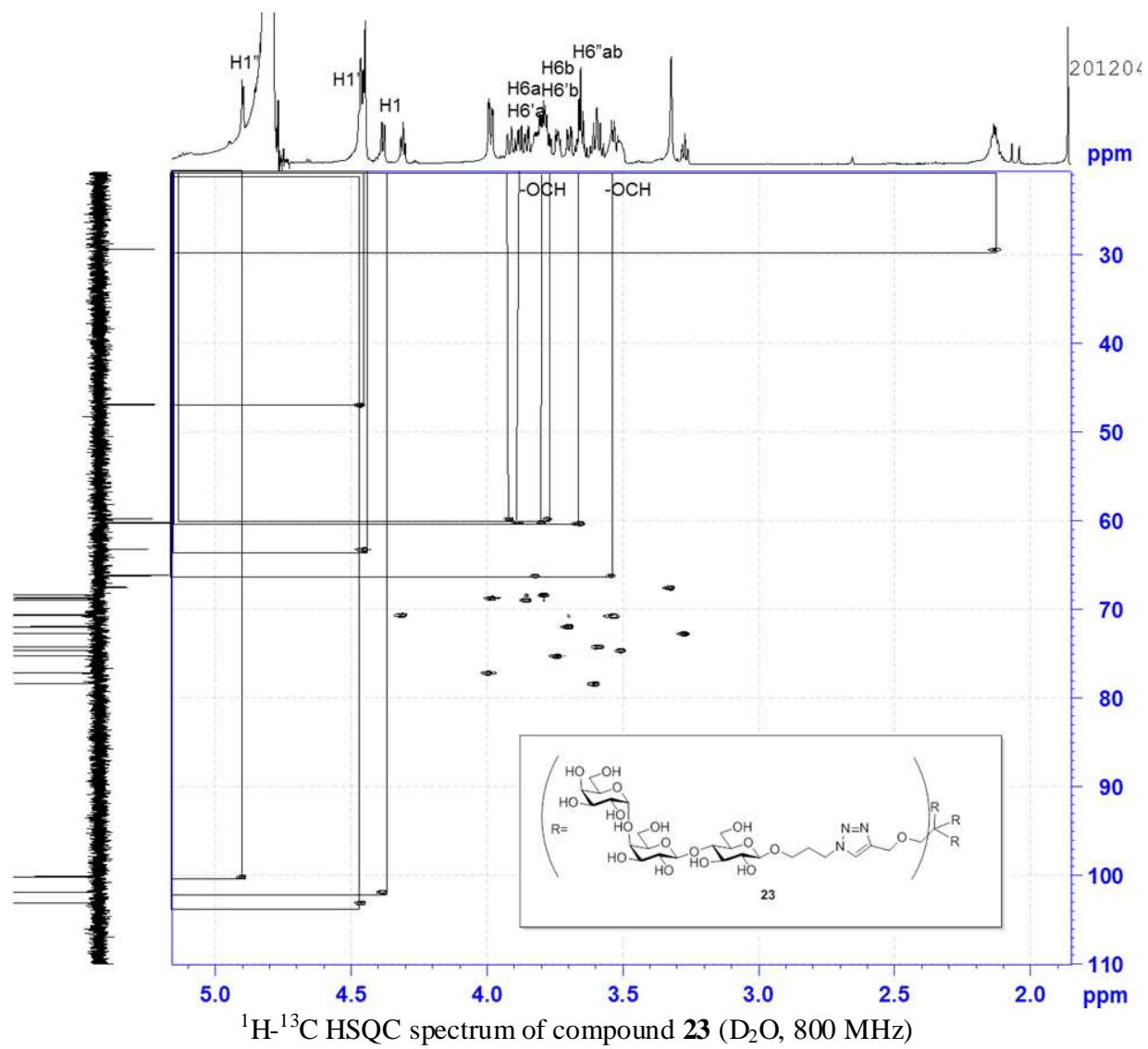


^1H NMR spectrum of compound **23** (D_2O , 800 MHz)

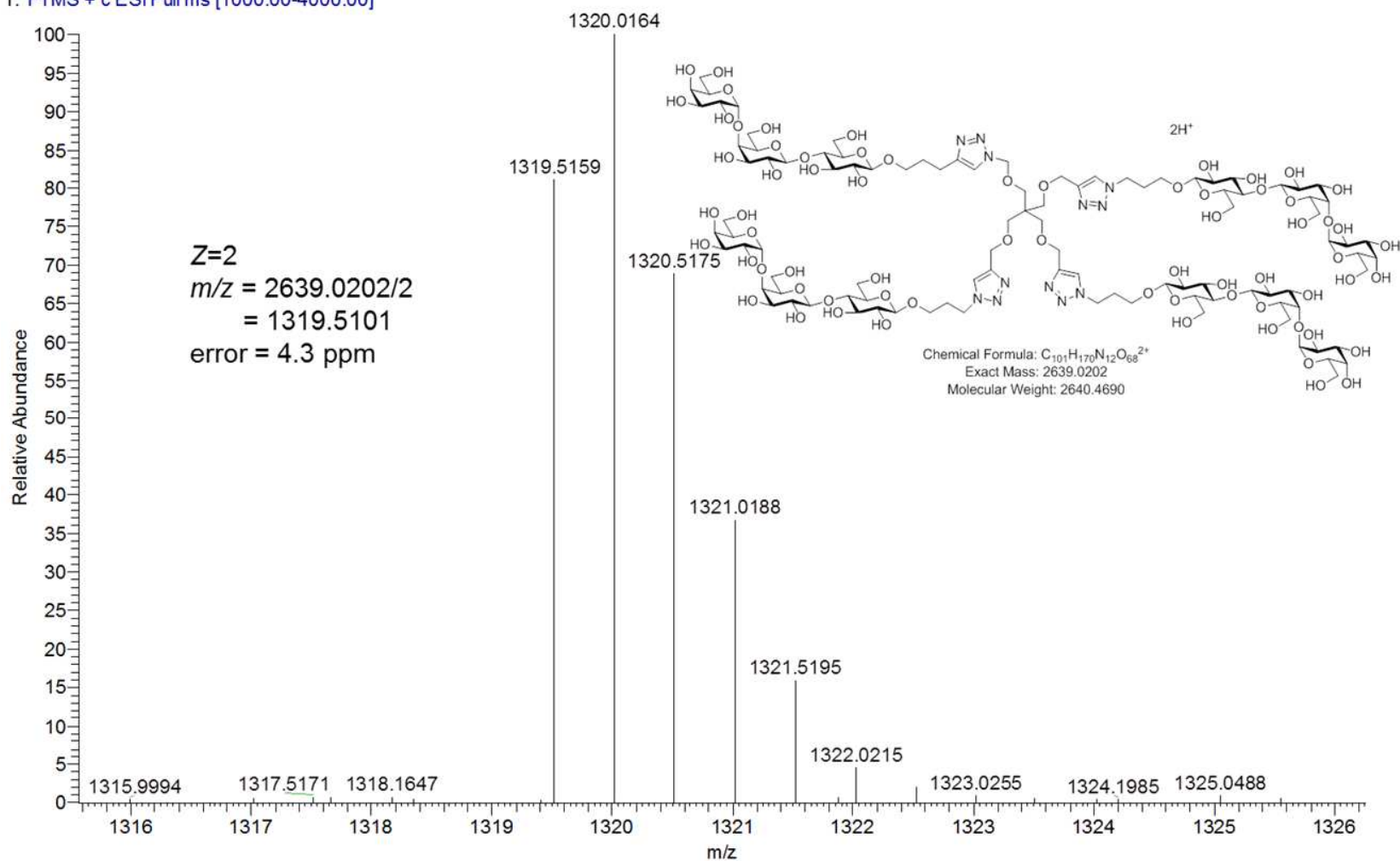


^{13}C and DEPT135 NMR spectrum of compound **23** (D₂O, 200 MHz)





HW127-Gb3tetramer #195 RT: 2.35 AV: 1 NL: 9.36E5
T: FTMS + c ESI Full ms [1000.00-4000.00]



ESI-HRMS spectrum of compound **23**