Electronic Supporting Information (ESI)

Diversion of a thioglycoligase for the synthesis of 1-*O*-acyl arabinofuranoses

Q. Pavic, S. Tranchimand, L. Lemiègre and L. Legentil*

*Email : laurent.legentil@ensc-rennes.fr

Table of content

General Experimental Details

All reagents were purchased from commercial sources and were used without further purification unless noted. 2'-benzimidazolyl-1-thio-α-*L*-arabinofuranoside **1** was synthesized according to literature procedure.[1] Unless otherwise stated, all reactions were monitored by TLC on Silica Gel 60 F_{254}. TLC spots were detected under 254 nm UV-light or by staining with cerium ammonium molybdate solution. Column chromatography was performed on Silica Gel (50 µm). Melting points were measured on a Stuart™ melting point apparatus SMP10. Optical rotations were measured at 20 °C on a Perkin-Elmer 341 polarimeter. NMR spectra were recorded at 400 MHz for ^{1}H and 100 MHz for ^{13}C. Chemical shifts are given in δ units (ppm) and referenced to DMSO-d_6. Coupling constants *J* were calculated in Hertz (Hz). Proton and carbon NMR peaks were unambiguously assigned by J-Mod (J-Modulated spin echo), COSY (double quantum filtered with gradient pulse for selection), HSQC (gradient echo-anti echo selection and shape pulse) and HMBC (echo-anti echo gradient selection, magnitude mode) correlation experiments. High Resolution Masses were recorded in positive mode using direct Electrospray ionization on a Waters Q-Tof 2 spectrometer.

Experimental procedures

Expression and purification of *Ct*Ara*f*51 E173A

Starting from a glycerol stock, *Escherichia coli* BL21 DE3 cells expressing *Ct*Ara*f*51 E173A[2] were cultured in 10 mL LB, supplemented with kanamycin (50 µg/mL), at 300 rpm, 37°C, overnight. 1 mL of the resulting saturated culture was subsequently used to prepare 1 L of autoinduction media ZYP-5052 containing 400 µg/mL of kanamycin. Cells were incubated for 72h at 250 rpm, 25°C. After centrifugation, 30 min at 4°C, pellet was resuspended in 100 mL of 50 mM phosphate buffer pH 7. Recombinant proteins were extracted from cell by french press and purified by 20 min heat denaturation step at 70°C, centrifugation 30 min at 4°C followed by purification on Ni affinity column.

Enzymatic synthesis of glycosyl esters:

General procedure:

A mixture of 4 equivalent of carboxylic acid acceptor in phosphate buffer (20 mL, 50 mM, pH = 6) was prepared and pH was adjusted to 6 with NaOH 2N. The mutant enzyme Ara*f*51 E173A (≈ 5 mg, 0.00025 eq) and 2'-benzimidazolyl-1-thio-α-*L*-arabinofuranoside **1** (≈ 100 mg, 0.35 mmol, 1 eq) were added. The mixture was stirred at RT for 16 hours. After freeze-drying, the mixture was purified by column chromatography on silica gel (dichloromethane/methanol 99:1 → 90:10) to yield the corresponding 1-*O*-acyl arabinofuranose.

Synthesis of acyl arabinofuranoses **2**, **17-26** and **30**:

1-(4'-methoxybenzoyl)-α-L-arabinofuranose (**2**): Product **2** was synthetized according to the general procedure using 2'-benzimidazolyl-1-thio-α-*L*-arabinofuranoside **1** (100 mg, 0.35 mmol) and 4-methoxybenzoic acid (216 mg, 1.42 mmol). Column chromatography gave **2** (77 mg, 78%) as a white solid. mp 125 °C. $[\alpha]_D$ = -15 (c 0.73 in MeOH). δH (400 MHz; DMSO-d_6) 3.47 (dt, 1 H, $J_{5,5'}$ = 11.8 Hz, $J_{5',8}$ = $J_{5',4}$ = 5.8 Hz, H-5'), 3.57 (ddd, 1 H, $J_{5,5'}$ = 11.8 Hz, $J_{5,8}$ = 5.5 Hz, $J_{5,4}$ = 3.9 Hz, H-5), 3.81 (m, 1 H, H-3), 3.84 (s, 3 H, H-14), 3.98 (dt, 1 H, $J_{4,3}$ = $J_{4\text{-}5'}$= 5.8 Hz, $J_{4,5}$ = 3.9 Hz, H-4), 4.10 (ddd, 1 H, $J_{2,6}$ = 4.8 Hz, $J_{2,3}$ =

3.2 Hz, $J_{2,1}$ = 1.4 Hz, H-2), 4.88 (t, 1 H $J_{8,5}$ = 5.6 Hz, OH-8), 5.37 (d, 1 H, $J_{7,3}$ = 4.7 Hz, OH-7), 5.56 (d, 1 H, $J_{6,2}$ = 5.0 Hz OH-6), 6.05 (d, 1 H, $J_{1,2}$ = 1.1 Hz, H-1), 7.06 (d, 2 H, $J_{12\text{-}11}$ = 8.9 Hz, H-12), 7.94 (m, 2 H, $J_{11\text{-}12}$ = 8.9 Hz, H-11). δC (100 MHz, DMSO) 55.6 (C-14), 61.4 (C-5), 76.8 (C-3), 81.2 (C-2), 86.7 (C-4), 102.3 (C-1), 114.1 (C-12), 121.8 (C-10), 131.5 (C-11), 163.4 (C-13), 164.6 (C-9). HRMS: *m/z* = 307.0791 $[M + Na]^+$ (expected for $C_{13}H_{16}O_7Na^+$: *m/z* = 307.0788).

1-O-benzoyl-α-L-arabinofuranose (**17**): Product **17** was synthetized according to the general procedure using 2'-benzimidazolyl-1-thio-α-*L*-arabinofuranoside **1** (100 mg, 0.35 mmol) and benzoic acid (173 mg, 1.42 mmol). Column chromatography gave **17** (51 mg, 57%) as a white solid. mp 119 °C. $[\alpha]_D$ = -16 (c 0.76 in MeOH). δH (400 MHz; DMSO-d_6) 3.48 (m, 1 H, H-5'), 3.57 (ddd, 1 H, $J_{5,5'}$ = 11.7 Hz, $J_{5,8}$ = 5.4 Hz, $J_{5,4}$ = 3.9 Hz, H-5), 3.83 (m, 1 H, H-3), 4.00 (ddd, 1 H, $J_{4,3}$ = $J_{4,5'}$ = 5.7 Hz, $J_{4,5}$ = 3.9 Hz, H-4), 4.12 (ddd, 1 H, $J_{2,6}$ = 4.8 Hz, $J_{2,3}$ = 3.3 Hz, $J_{2,1}$ = 1.3 Hz, H-2), 4.90 (t, 1 H, $J_{OH\ 8,5}$ = 5.4 Hz, OH-8), 5.39 (d, 1 H, $J_{OH\ 7,3}$ = 4.6 Hz, OH-7), 5.60 (d, 1 H, $J_{OH\ 6,2}$ = 4.8 Hz, OH-6), 6.09 (d, 1 H, $J_{1,2}$ = 1.3 Hz, H-1), 7.55 (m, 2 H, H-12), 7.68 (m, 1 H, H-13), 7.99 (m, 2 H, H-11). δC (100 MHz, DMSO) 61.4 (C-5), 76.7 (C-3), 81.2 (C-2), 87.1 (C-4), 102.7 (C-1), 128.8 (C-12), 129.3 (C-11), 129.6 (C-10), 133.7 (C-13), 165.0 (C-9). HRMS: *m/z* = 277.0684 $[M + Na]^+$ (expected for $C_{12}H_{14}O_6Na^+$: *m/z* = 277.0683).

1-O-(4'-bromobenzoyl)-α-L-arabinofuranose (**18**): Product **18** was synthetized according to the general procedure using 2'-benzimidazolyl-1-thio-α-*L*-arabinofuranoside **1** (100 mg, 0.35 mmol) and 4-bromobenzoic acid (281 mg, 1.42 mmol). Column chromatography gave **18** (55 mg, 47%) as a colourless oil. $[\alpha]_D$ = -17 (c 0.85 in MeOH). δH (400 MHz; DMSO-d_6) 3.47 (dt, 1 H, $J_{5,5'}$ = 11.1 Hz, $J_{5,4}$ = 4.99 Hz H-5'), 3.57 (dt, 1 H, $J_{5,5'}$ = 11.1 Hz, $J_{5,4}$ = 3.6 Hz, H-5), 3.83 (m, 1 H, H-3), 4.00 (m, 1 H, H-4), 4.12 (m, 1 H, H-2), 4.91 (bs, 1 H, OH-8), 5.39 (bs, 1 H, OH-7), 5.60 (bs, 1 H, OH-6), 6.08 (d, 1 H, $J_{1,2}$ = 1.2 Hz, H-1), 7.77 (d, 2 H, $J_{11,12}$ = 8.6 Hz, H-11), 7.90 (d, 2H, $J_{12,11}$ = 8.6 Hz, H-12). δC (100 MHz, DMSO) 61.4 (C-5), 76.7 (C-3), 81.1 (C-2), 87.2 (C-4), 103.0 (C-1), 127.8 (C-13), 128.8 (C-10), 131.3 (C-12), 132.0 (C-11), 164.3 (C-9). HRMS: *m/z* = 354.9787 $[M + Na]^+$ (expected for $C_{12}H_{13}BrO_6Na^+$: *m/z* = 354.9788).

1-(4'-trifluoromethylbenzoyl)-α-L-arabinofuranose (**19**): Product **19** was synthetized according to the general procedure using 2'-benzimidazolyl-1-thio-α-*L*-arabinofuranoside **1** (100 mg, 0.35 mmol) and 4-trifluoromethylbenzoic acid (270 mg, 1.42 mmol). Column chromatography gave **19** (64 mg, 57%) as a white solid. mp degradation above 110 °C. $[\alpha]_D$ = -3 (c 0.58 in MeOH). δH (400 MHz; DMSO-d_6) 3.48 (ddd, 1 H, $J_{5,5'}$ = 11.8 Hz, $J_{5',8}$ = $J_{5',4}$ = 5.5 Hz, H-5'), 3.57 (ddd, 1 H, $J_{5,5'}$ = 11.8 Hz, $J_{5,8}$ = 5.5 Hz, $J_{5,4}$ = 4.1 Hz, H-5), 3.85 (m, 1 H, H-3), 4.03 (m, 1 H, H-4), 4.15 (ddd, 1 H, $J_{2,6}$ = 4.8 Hz, $J_{2,3}$ = 3.0 Hz, $J_{2,1}$ = 1.2 Hz, H-2), 4.92 (t, 1 H $J_{8,5}$ = 5.6 Hz, OH-8), 5.41 (d, 1 H, $J_{7,3}$ = 4.8 Hz, OH-7), 5.63 (d, 1 H, $J_{6,2}$ = 4.8 Hz OH-6), 6.11 (d, 1 H, $J_{1,2}$ = 1.2 Hz, H-1), 7.94 (d, 2 H, $J_{12\text{-}11}$ = 8.1 Hz, H-12), 8.18 (d, 2 H, $J_{11\text{-}12}$ = 8.1 Hz, H-11). δC (100 MHz, DMSO) 61.3 (C-5), 76.7 (C-3), 81.1 (C-2), 87.4 (C-4), 103.4 (C-1), 123.7 (q, $J_{C\text{-}F}$ = 272.9 Hz, C-14), 125.9 (q, $J_{C\text{-}F}$ = 3.8 Hz, C12), 130.2 (C-11), 132.9 5 (q, $J_{C\text{-}F}$ = 32.0 Hz, C-13), 133.4 (d, $J_{C\text{-}F}$ = 1.8 Hz, C-10), 163.9 (C-9).HRMS: *m/z* = 345.0563 $[M + Na]^+$ (expected for $C_{13}H_{16}O_7Na^+$: *m/z* = 345.0556).

1-(4'-nitrobenzoyl)-α-L-arabinofuranose (**20**): Product **20** was synthetized according to the general procedure using 2'-benzimidazolyl-1-thio-α-*L*-arabinofuranoside **1** (100 mg, 0.35 mmol) and 4-nitrobenzoic acid (234 mg, 1.42 mmol). Column chromatography gave **6** (NMR conversion 18%, in mixture with 4-nitrobenzoic acid). δH (400 MHz; DMSO-d_6) 3.48 (dd, 1 H, $J_{5',5}$ = 11.8 Hz, $J_{5',4}$ = 5.6 Hz, H-5'); 3.58 (dd, 1 H, $J_{5,5'}$ = 11.8 Hz, $J_{5,4}$ = 4.1 Hz, H-5), 3.85 (dd, 1 H, $J_{3,4}$ = 5.3 Hz, $J_{3,2}$ = 3.0 Hz, H-3), 4.04 (ddd, 1 H, $J_{4,3}$ = $J_{4,5'}$ = 5.6 Hz, $J_{4,5}$ = 4.1 Hz, H-4), 4.16 (dd, 1 H, $J_{2,3}$ = 3.0 Hz, $J_{2,1}$ = 1.3 Hz, H-2), 4.93 (bs, 1 H, OH-8), 5.43 (bs, 1 H, OH-7), 5.65 (bs, 1 H, OH-6), 6.12 (d, 1 H, $J_{1,2}$ = 1.1 Hz, H-1), 8.21 (d, 2H, $J_{12,11}$ = 9.0 Hz, H-11), 8.38 (d, 2H, $J_{12,11}$ = 9.0 Hz, H-12). δC (100 MHz, DMSO) 61.3 (C-5), 76.6 (C-3), 81.1 (C-2), 87.5 (C-4), 103.6 (C-1), 124.0 (C-12), 130.8 (C-11), 135.0 (C-10), 150.5 (C-13), 163.5 (C-9). HRMS: *m/z* = 322.0533 $[M + Na]^+$ (expected for $C_{12}H_{13}NO_8Na^+$: *m/z* = 322.0533).

1-(4'-hydroxybenzoyl)-α-L-arabinofuranose (**21**): Product **21** was synthetized according to the general procedure using 2'-benzimidazolyl-1-thio-α-*L*-arabinofuranoside **1** (100 mg, 0.35 mmol) and 4-hydroxybenzoic acid (196 mg, 1.42 mmol). Column chromatography gave **21** (74 mg, 78%) as a white solid. mp 120 °C. $[\alpha]_D$ = -73 (c 1.19 in MeOH). δH (400 MHz; DMSO-d_6) 3.58-3.45 (m, 2 H, H-5), 3.80 (dt, 1 H, $J_{3,4} = J_{3,7}$ = 4.8 Hz, $J_{3,2}$ = 3.3 Hz H-3), 3.96 (dt, 1 H, $J_{4,3} = J_{4,5'}$ = 5.8 Hz, $J_{4,5}$ = 3.8 Hz, H-4), 4.08 (m, 1 H, H-2), 4.90 (t, 1 H $J_{8,5}$ = 5.6 Hz, OH-8), 5.38 (d, 1 H, $J_{7,3}$ = 4.8 Hz, OH-7), 5.57 (d, 1 H, $J_{6,2}$ = 5.0 Hz OH-6), 6.03 (d, 1 H, $J_{1,2}$ = 1.3 Hz, H-1), 6.86 (d, 2 H, $J_{12,11}$ = 8.7 Hz, H-12), 7.83 (d, 2 H, $J_{11,12}$ = 8.7 Hz, H-11). δC (100 MHz, DMSO) 61.4 (C-5), 76.8 (C-3), 81.3 (C-2), 86.8 (C-4), 102.2 (C-1), 115.4 (C-12), 120.24 (C-10), 131.8 (C-11), 162.2 (C-13), 164.8 (C-9). HRMS: *m/z* = 293.0633 $[M + Na]^+$ (expected for $C_{12}H_{14}O_7Na^+$: *m/z* = 293.0632).

1-O-phenylacetoyl-α-L-arabinofuranose (**22**): Product **22** was synthetized according to the general procedure using 2'-benzimidazolyl-1-thio-α-*L*-arabinofuranoside **1** (100 mg, 0.35 mmol) and phenylacetic acid (193 mg, 1.42 mmol). Column chromatography gave **22** (63 mg, 66%) as a colourless oil. $[\alpha]_D$ = -18 (c 0.63 in MeOH). δH (400 MHz; DMSO-d_6) 3.42 (dd, 1 H, $J_{5',5}$ = 12.0 Hz, $J_{5',4}$ = 5.8 Hz ,H-5'), 3.54 (dd, 1 H, $J_{5,5'}$ = 12.1 Hz, $J_{5,4}$ = 3.4 Hz, H-5), 3.69 (s, 2 H, H-10), 3.75 (dd, 1 H, $J_{3,4}$ = 6.0 Hz, $J_{3,2}$ = 3.6 Hz, H-3), 3.88 (ddd, 1 H, $J_{4,3} = J_{4,5'}$ = 5.9 Hz, $J_{4,5}$ = 3.7 Hz, H-4), 3.96 (dd, 1 H, $J_{2,3}$ = 3.6 Hz, $J_{2,1}$ = 1.6 Hz, H-2), 4.86 (br s, 1 H, OH-8), 5.33 (br s, 1 H, OH-7), 5.53 (br s, 1 H, OH-6), 5.84 (d, 1 H, $J_{1,2}$ = 1.6 Hz, H-1), 7.35-7.24 (m, 5 H, H-Ar). δC (100 MHz, DMSO) 40.4 (C-10), 61.2 (C-5), 76.6 (C-3), 81.3 (C-2), 86.5 (C-4), 102.4 (C-1), 126.9 (C-14), 128.3 (C-13), 129.4 (C-12), 134.1 (C-11), 170.6 (C-9). HRMS: *m/z* = 291.0837 $[M + Na]^+$ (expected for $C_{13}H_{16}O_6Na^+$: *m/z* = 291.0839).

1-hexanoyl-α-L-arabinofuranose (**23**): Product **23** was synthetized according to the general procedure using 2'-benzimidazolyl-1-thio-α-*L*-arabinofuranoside **1** (100 mg, 0.35 mmol) and hexanoic acid (163 mg, 1.42 mmol). Column chromatography gave **23** (67 mg, 77%) as a viscous liquid. $[\alpha]_D$ = -75 (c 0.55 in MeOH). δH (400 MHz; DMSO-d_6) 0.87 (t, 3 H, $J_{14,13}$ = 6.9 Hz, H-14), 1.29-1.23 (m, 4 H, H-12, H-13), 1.52 (m, 2 H, H-11), 2.29 (t, 2 H, $J_{10,11}$ = 7.4 Hz, H-10), 3.42 (ddd, 1 H, $J_{5',5}$ = 11.8 Hz, $J_{5',4}$ = 5.8 Hz, $J_{5',8\ OH}$ = 5.6 Hz, H-5'), 3.53 (ddd, 1 H, $J_{5,5'}$ = 11.8 Hz, $J_{5,8\ OH}$ = 5.6 Hz, $J_{5,4}$ = 3.7 Hz, H-5), 3.73 (ddd, 1 H, $J_{3,7\ OH}$ = 4.8 Hz, $J_{1,2}$ = 1.3 Hz, $J_{3,2}$ = 3.6 Hz, H-3), 3.85 (dt, 1 H, $J_{4,3}$ = 5.8 Hz, $J_{4,5}$ = 3.7 Hz, H-4), 3.92 (ddd, 1 H, $J_{2,6\ OH}$ = 5.2 Hz, $J_{2,3}$ = 3.6 Hz, $J_{2,1}$ = 1.6 Hz H-2), 4.84 (t, 1 H $J_{8\ OH,5}$ = 5.6 Hz, OH-8), 5.28 (d, 1 H, $J_{7\ OH,3}$ = 4.8 Hz OH-7), 5.49 (d, 1 H, $J_{6\ OH,2}$ = 5.2 Hz OH-6), 5.82 (d, 1 H, $J_{1,2}$ = 1.6 Hz, H-1). δC (100 MHz, DMSO) 14.3 (C-14), 22.3-31.0 (C-12,13), 24.4 (C-11), 34.1 (C-10), 61.7 (C-5), 77.1 (C-3), 81.8 (C-2), 86.8 (C-4), 102.3 (C-1), 172.9 (C-9). HRMS: *m/z* = 271.1153 $[M + Na]^+$ (expected for $C_{11}H_{20}O_6Na^+$: *m/z* = 271.1152).

1-(2',2'-dimethylpropanoyl)-α-L-arabinofuranose (**24**): Product **24** was synthetized according to the general procedure using 2'-benzimidazolyl-1-thio-α-*L*-arabinofuranoside (100 mg, 0.35 mmol) and pivaloic acid (143 mg, 1.42 mmol). Column chromatography gave **24** (18 mg, 25%) as colourless oil. $[\alpha]_D$ = -78 (c 0.86 in MeOH). δH (400 MHz; DMSO-d_6) 1.13 (s, 9 H, H-11), 3.51 (m, 2 H, H-5), 3.73 (m, 1 H, H-3), 3.88 (dt, 1 H, $J_{4,3} = J_{4,5'}$ = 5.8 Hz, $J_{4,5}$ = 3.8 Hz, H-4), 3.46 (m, 1 H, H-2), 4.87 (t, 1 H $J_{8,5}$ = 5.6 Hz, OH-8), 5.29 (d, 1 H, $J_{7,3}$ = 4.6 Hz, OH-7), 5.49 (d, 1 H, $J_{6,2}$ = 4.9 Hz OH-6), 5.79 (d, 1 H, $J_{1,2}$ = 1.6 Hz, H-1). δC (100 MHz, DMSO) 26.7 (C-11), 38.4 (C-10), 61.4 (C-5), 76.9 (C-3), 81.3 (C-2), 86.7 (C-4), 102.1 (C-1), 176.79 (C-9). HRMS: *m/z* = 257.0995 $[M + Na]^+$ (expected for $C_{10}H_{18}O_6Na^+$: *m/z* = 257.0995).

(S)-Ibuprofen-α-L-arabinofuranose (**25**): Product **25** was synthetized according to the general procedure using 2'-benzimidazolyl-1-thio-α-*L*-arabinofuranoside **1** (100 mg, 0.35 mmol) and (*S*)-Ibuprofen (292 mg, 1.42 mmol). Column chromatography gave **25** (99 mg, 83%) as colourless oil. $[\alpha]_D$ = -6 (c 0.56 in MeOH). δH (400 MHz; DMSO-d_6) 0.85 (d, 6 H, $J_{18,17}$ = 6.7 Hz, H-18), 1.37 (d, 3 H, $J_{11,10'}$ = 7.2 Hz, H-11), 1.80 (dh, 1 H, $J_{17,16}$ = 7.2 Hz, $J_{17\text{-}18}$ = 6.7 Hz, H-17), 2.41 (d, 2 H, $J_{16,17}$ = 7.2 Hz, H-16), 3.39 (d, 1 H, $J_{5',5}$ = 11.9 Hz, H-5'), 3.48 (m, 1 H, H-5), 3.77-3.71 (m, 3 H, H-3, H-4, H-10), 3.93 (m, 1 H, H-2), 4.81 (bs, 1 H, H-8), 4.93 (bs, 1 H, H-6), 5.26 (bs, 1 H, H-7), 5.82 (d, 1 H, $J_{1,2}$ = 1.5 Hz, H-1), 7.10 (d, 2 H,

$J_{14,13}$ = 8.1 Hz, H-14), 7.20 (d, 2 H, $J_{13,14}$ = 8.1 Hz, H-13). δC (100 MHz, DMSO) 18.4 (C-11), 22.2 (C-18), 29.6 (C-17), 44.21 (C-10), 44.23 (C-16), 61.0 (C-5), 76.6 (C-3), 81.3 (C-2), 86.4 (C-4), 102.4 (C-1), 127.1-129.0 (C-13,14), 137.5-139.8 (C-12,15), 173.2 (C-9). HRMS: *m/z* = 361.1622 $[M + Na]^+$ (expected for $C_{18}H_{26}O_6Na^+$: *m/z* = 361.1622).

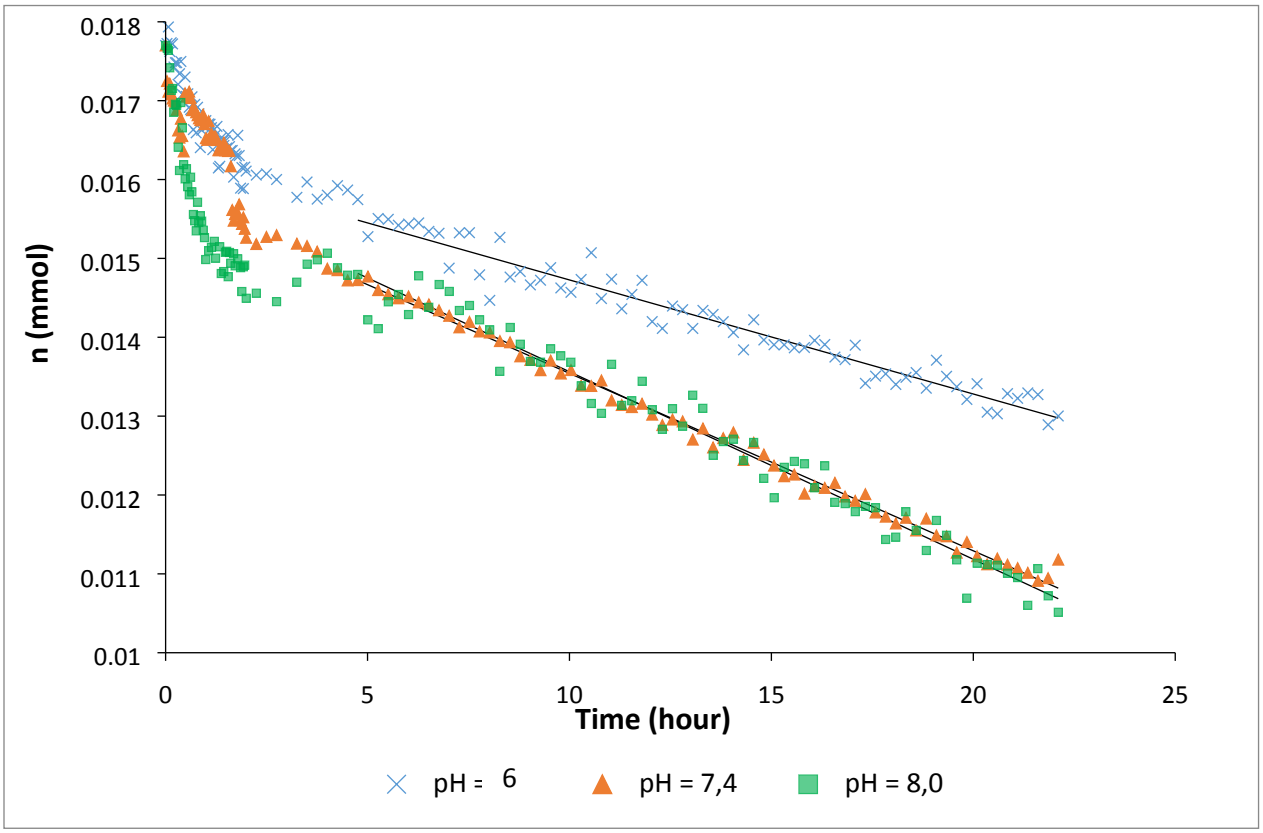
1-(6'-hydroxyhexanoyl)-α-L-arabinofuranose (**26**): Product **26** was synthetized according to the general procedure using 2'-benzimidazolyl-1-thio-α-*L*-arabinofuranoside **1** (100 mg, 0.35 mmol) and 6-hydroxyhexanoic acid (188 mg, 1.42 mmol). Column chromatography gave **26** (49 mg, 53%) as a colourless oil. $[\alpha]_D$ = -73 (c 1.35 in MeOH). δH (400 MHz; DMSO-d_6) 1.29 (m, 2H, H-12), 1.40 (m, 2H, H-13), 1.52 (p, 2H, $J_{11,10} = J_{11,12}$ = 7.4 Hz, H-11), 2.29 (t, 2H, $J_{10,11}$ = 7.4 Hz, H-10), 3.36 (m, 3H, H-5', H-14), 3.53 (ddd, 1 H, $J_{5,5}$ = 11.6 Hz, $J_{5,8}$ = 5.3 Hz, $J_{5,4}$ = 3.6 Hz, H-5), 3.73 (dt, 1 H, $J_{3,4} = J_{3,7}$ = 5.3 Hz, $J_{3,2}$ = 3.5 Hz H-3), 3.85 (dt, 1 H, $J_{4,3} = J_{4,5'}$ = 5.8 Hz, $J_{4,5}$ = 3.7 Hz, H-4), 3.92 (ddd, 1 H, $J_{2,6}$= 5.2 Hz, $J_{2,3}$ = 3.5 Hz, $J_{2,1}$ = 1.6 Hz, H-2), 4.69 (t, 1H, $J_{15,14}$ = 5.2 Hz, OH-15), 4.87 (t, 1 H $J_{8,5}$ = 5.6 Hz, OH-8), 5.31 (d, 1 H, $J_{7,3}$ = 4.9 Hz, OH-7), 5.52 (d, 1 H, $J_{6,2}$ = 5.2 Hz OH-6), 5.82 (d, 1 H, $J_{1,2}$ = 1.6 Hz, H-1). δC (100 MHz, DMSO) 24.2, 25.0 (C-11, C-12), 32.2, 33.8 (C-10, C-13),60.6 (C-14), 61.3 (C-5), 76.7 (C-3), 81.3 (C-2), 86.4 (C-4), 101.9 (C-1), 172.5 (C-9). HRMS: *m/z* = 401.1783 $[M + Na]^+$ (expected for $C_{11}H_{20}O_7Na^+$: *m/z* =401.1782).

N-Boc-phenylalanine-α-L-arabinofuranose (**30**): Product **30** was synthetized according to the general procedure using 2'-benzimidazolyl-1-thio-α-*L*-arabinofuranoside **1** (100 mg, 0.35 mmol) and N-Boc-phenylalanine **16** (376 mg, 1.42 mmol). Column chromatography gave **30** (53 mg, 38%) as white solid. mp 95-100 °C. $[\alpha]_D$ = +4 (c 1.11 in MeOH). δH (400 MHz; DMSO-d_6) 1.32 (s, 9 H, H-19), 2.85 (dd, 1 H, , $J_{11',11}$ = 13.6 Hz, $J_{11',10}$ = 10.1 Hz, H-11'), 3.02 (dd, 1 H, , $J_{11,11'}$ = 13.6 Hz, $J_{11,10}$ = 4.9 Hz, H-11), 3.43 (dd, 1 H, $J_{5',5}$ = 11.9 Hz, $J_{5',4}$ = 5.8 Hz, H-5'), 3.54 (dd, 1 H, $J_{5,5'}$ = 12.0 Hz, $J_{5,4}$ = 3.6 Hz, H-5), 3.77 (dd, 1 H, $J_{3,4}$ = 5.8 Hz, $J_{3,2}$ = 3.5 Hz, H-3), 3.84 (dt, 1 H, $J_{4,3}$ = 5.8 Hz, $J_{4,5}$ = 3.6 Hz, H-4), 3.99 (dd, 1 H, $J_{2,3}$ = 3.5 Hz, $J_{2,1}$ = 1.5 Hz, H-2), 4.17 (ddd, 1 H, $J_{10,11}$ = 10.0 Hz, $J_{10,NH}$ = 8.13 Hz, $J_{10,11'}$ = 4.6 Hz, H-10), 5.84 (d, 1 H, $J_{1,2}$ = 1.4 Hz, H-1), 7.29-7.18 (m, 6 H, H-Ar, NH). δC (100 MHz, DMSO) 28.2 (C-19), 36.2 (C-11), 55.2 (C-10), 61.0 (C-5), 76.7 (C-3), 78.4 (C-18), 81.3 (C-2), 86.4 (C-4), 102.6 (C-1), 126.4 (C-15), 128.2 (C-Ar), 129.1 (C-Ar), 137.5 (C-12), 155.4 (C-17), 171.4 (C-9). HRMS: *m/z* = 420.1629 $[M + Na]^+$ (expected for $C_{19}H_{27}NO_8Na^+$: *m/z* = 420.1629).

References

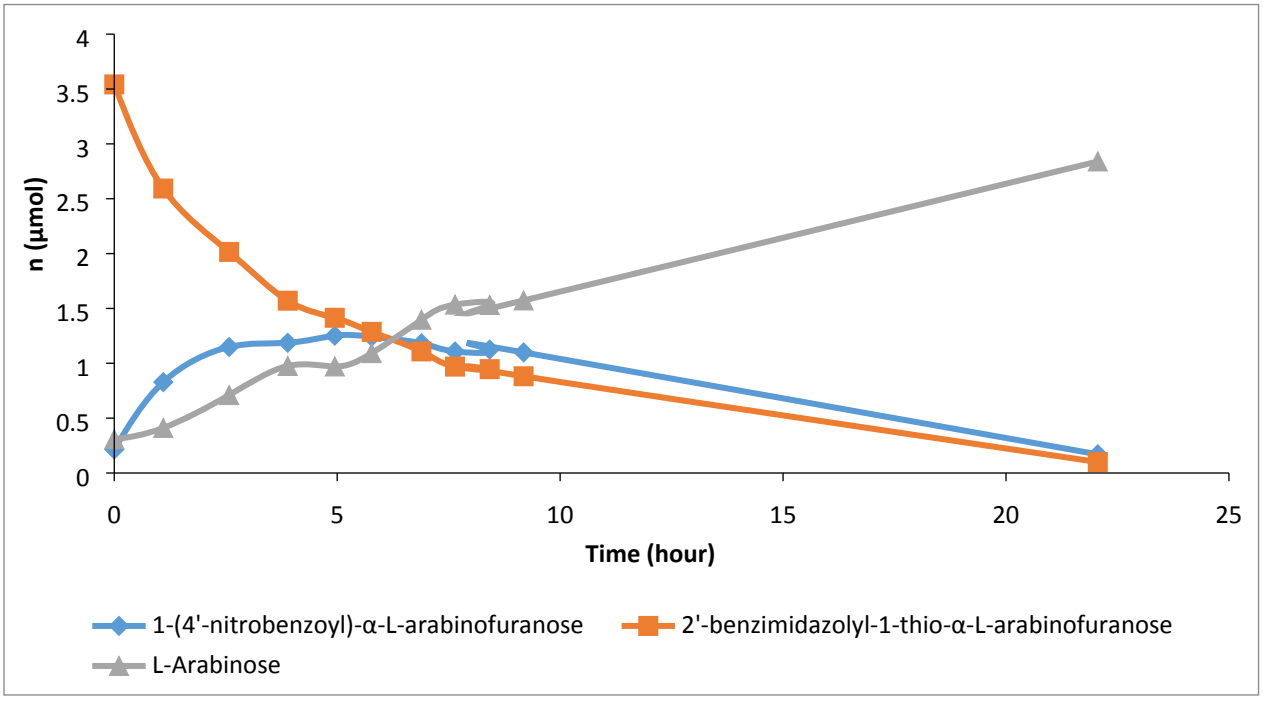
1. R. Euzen, V. Ferrières and D. Plusquellec, *J. Org. Chem.*, 2005, **70**, 847-855.
2. Edward J. Taylor, Nicola L. Smith, Johan P. Turkenburg, S. D'Souza, Harry J. Gilbert and Gideon J. Davies, *Biochem. J.*, 2006, **395**, 31-37.

Kinetics of hydrolysis at different pH of 2 followed by ^{1}H NMR

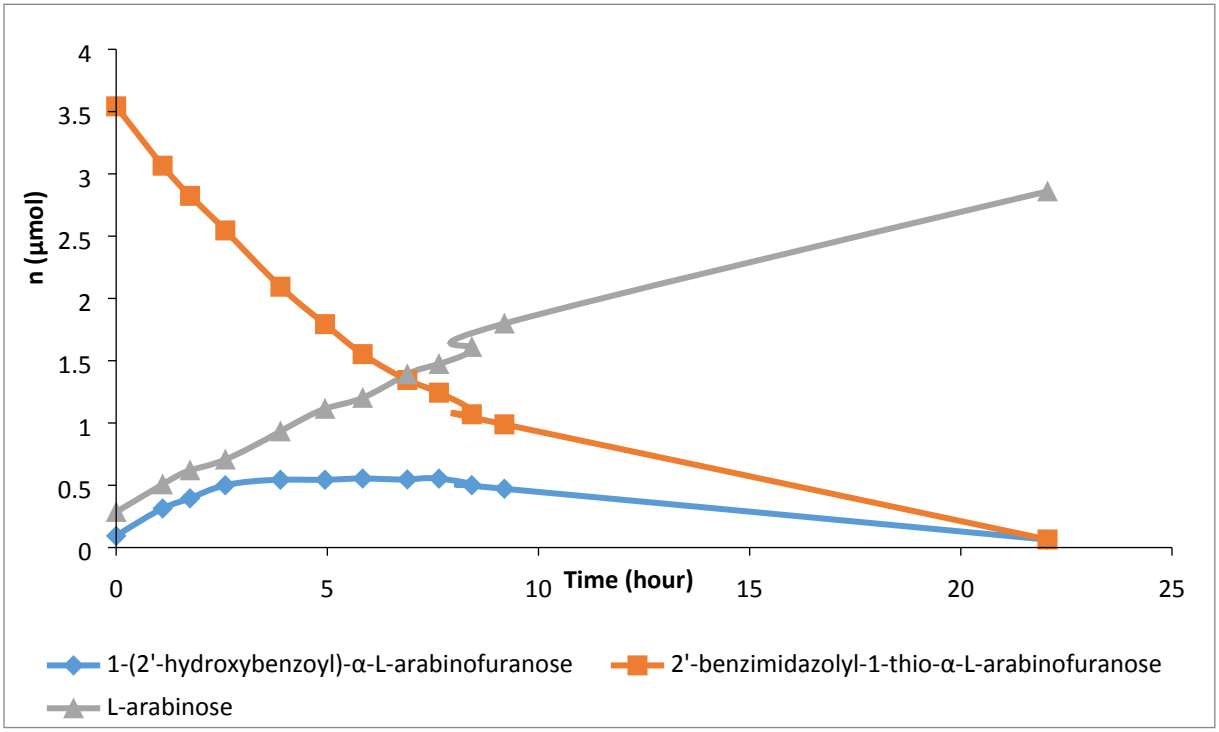


Reaction monitoring of acylation

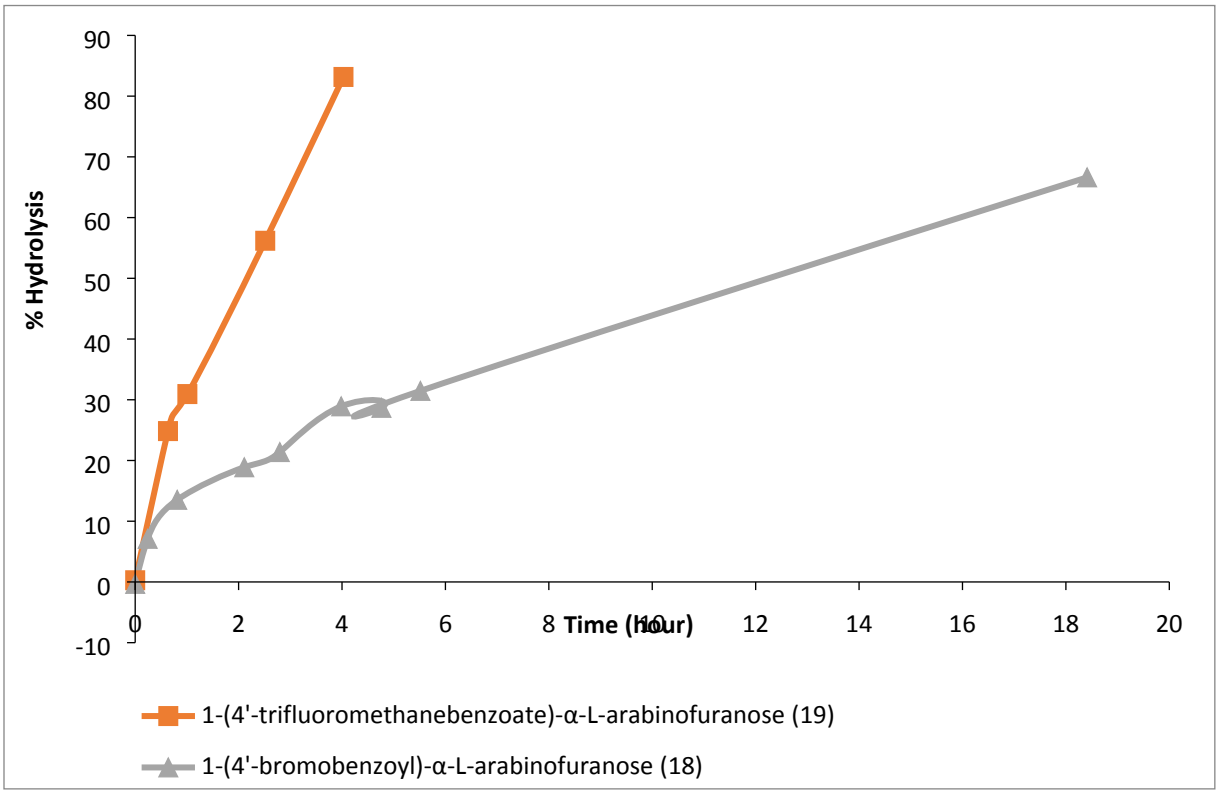
- with acceptor 4-nitrobenzoic acid



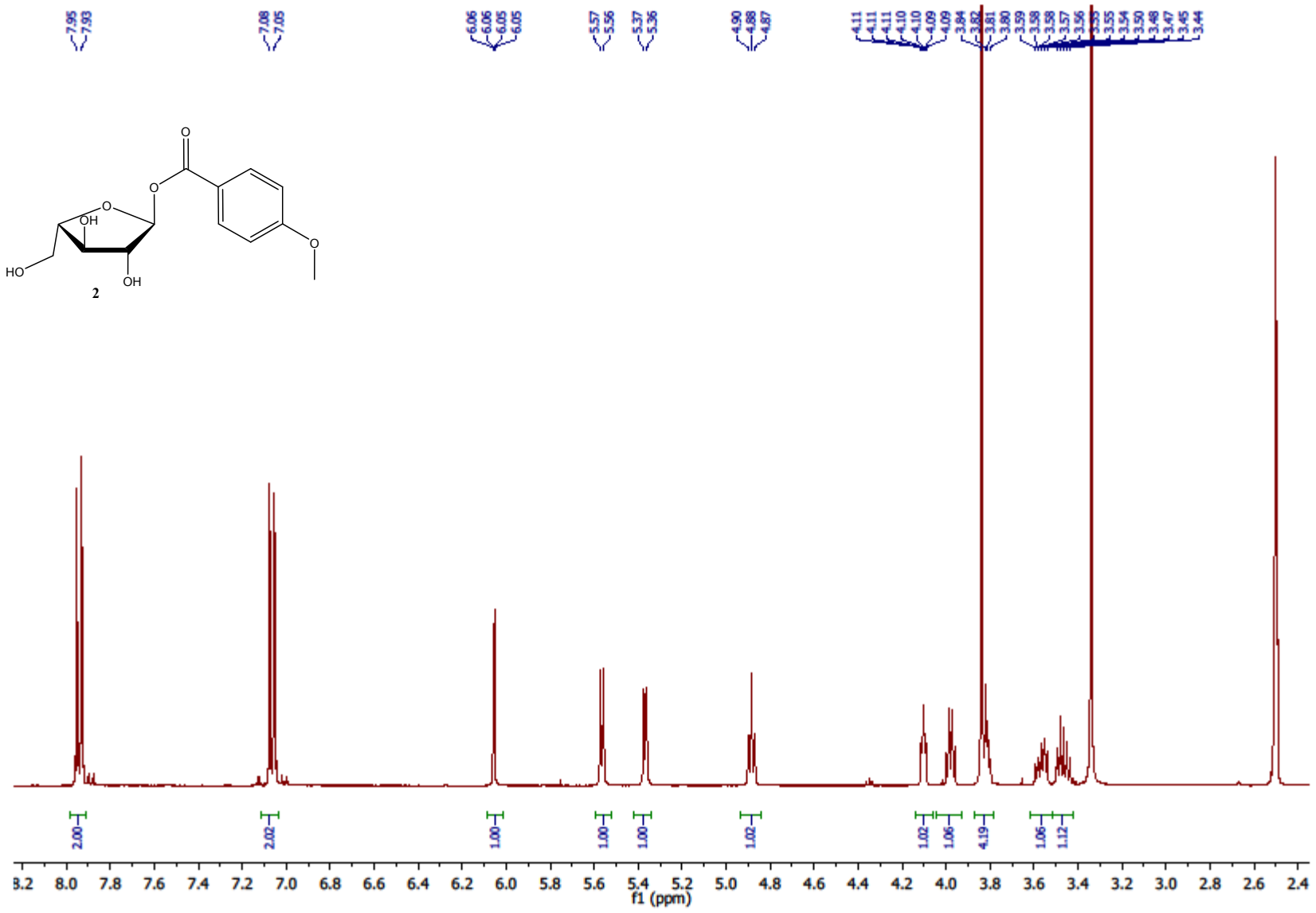
- with acceptor 2-hydroxybenzoic acid

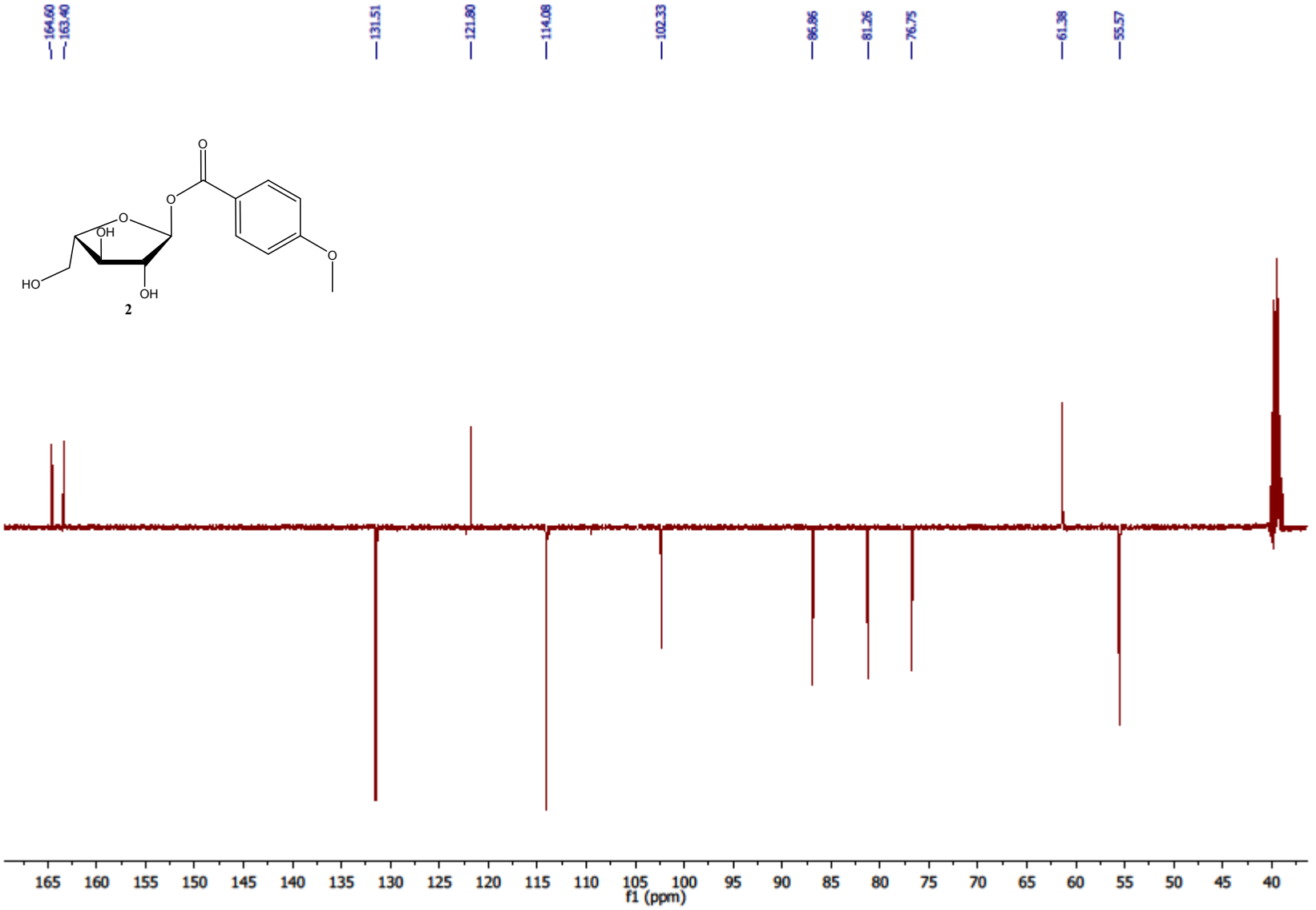


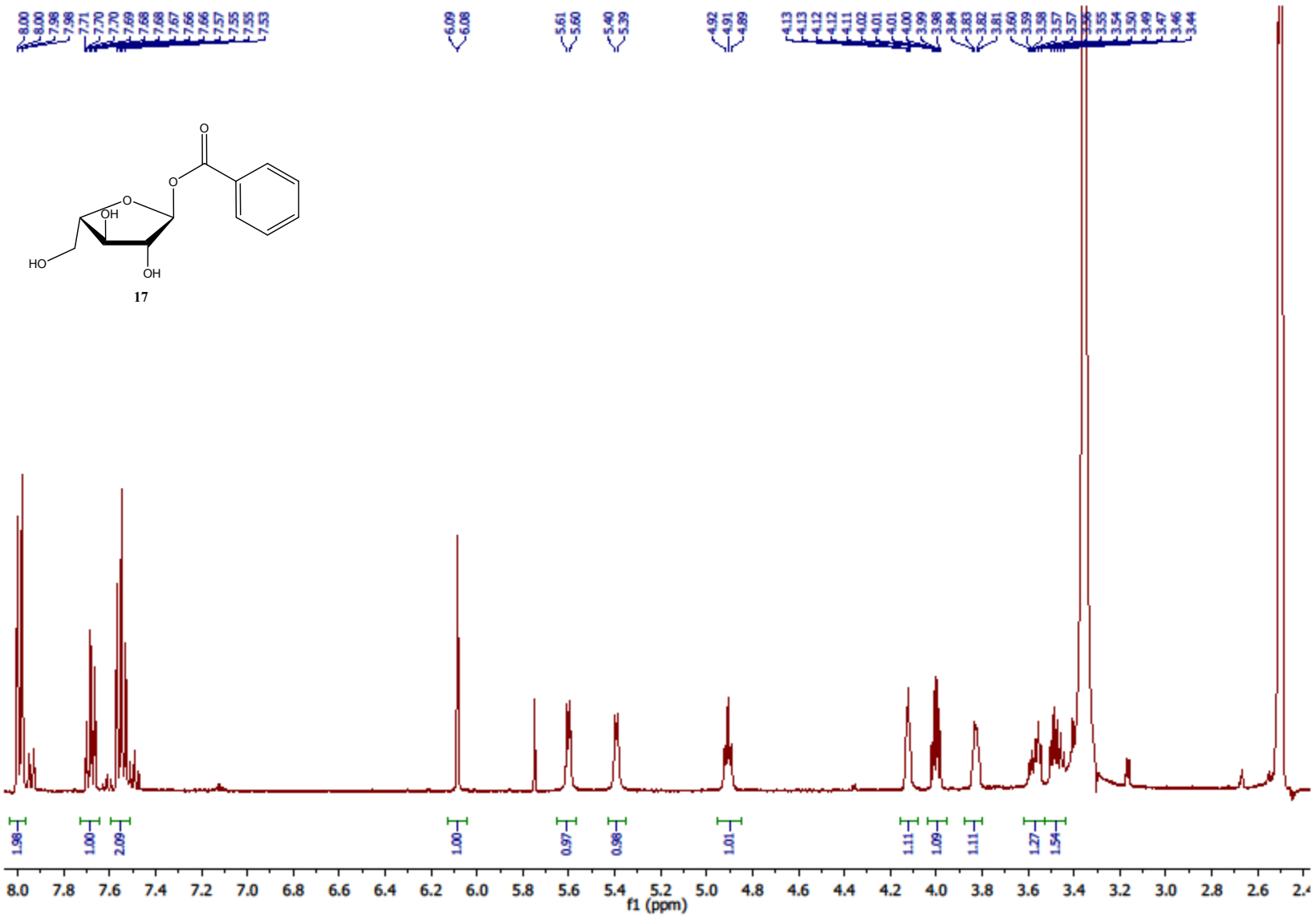
Stability of acyl-α-L-arabinofuranoses 18 and 19 with enzyme *Ct*Araf51 E173A

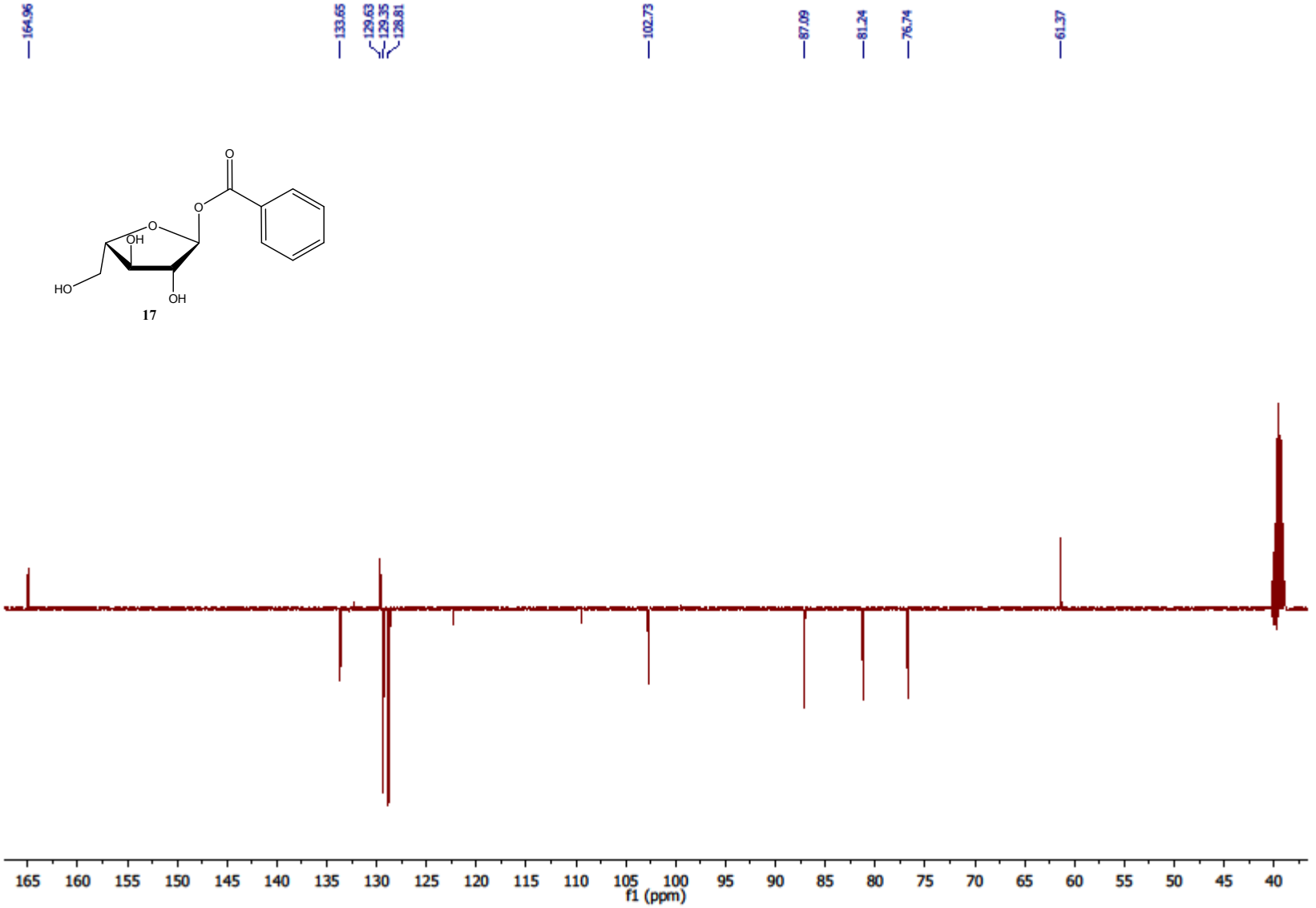


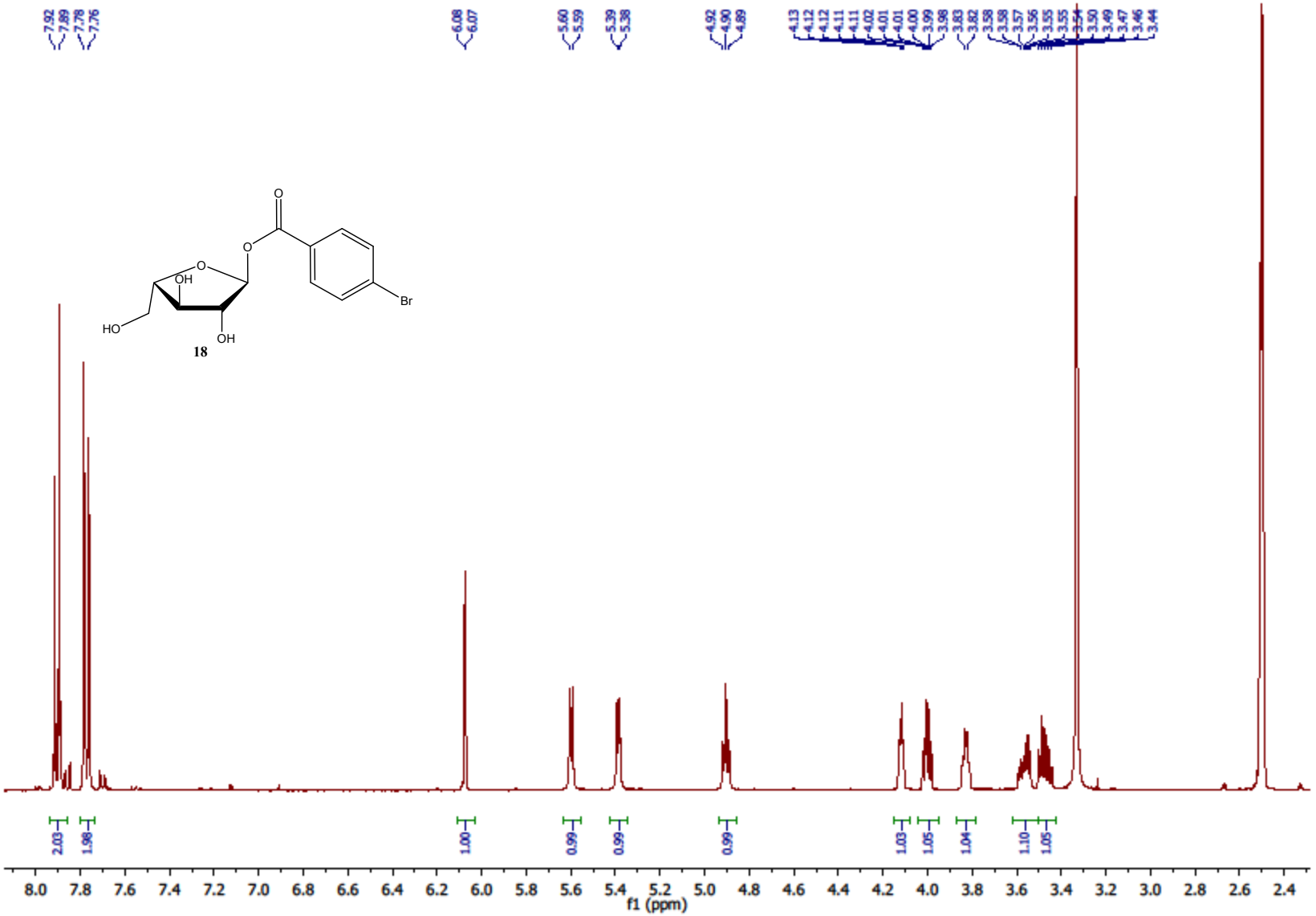
^{1}H and ^{13}C NMR spectrum of isolated acyl arabinofuranoses **2**, **17-27** and **30**

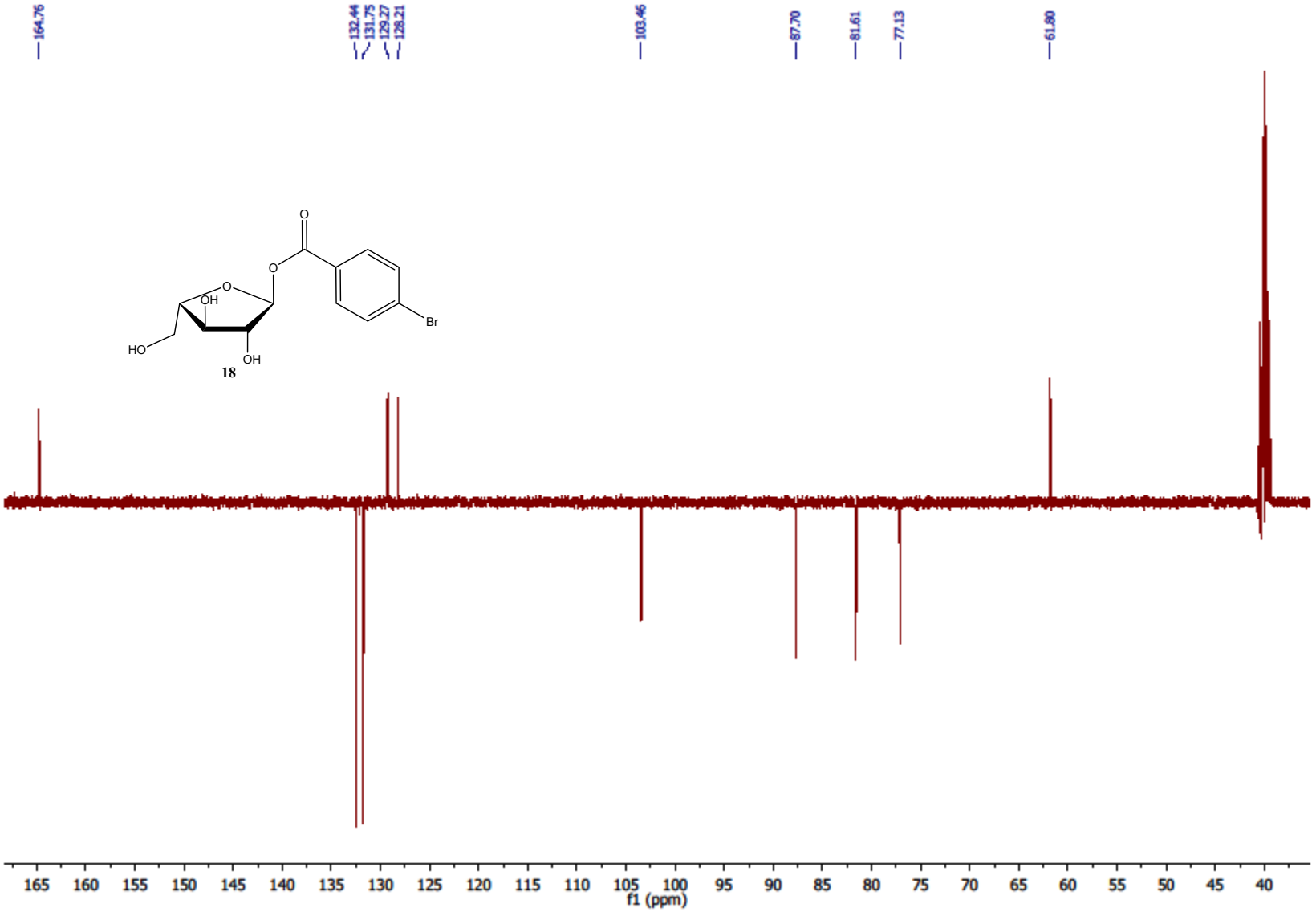

7.95
7.93
7.08
7.05
6.06
6.06
6.05
6.05
5.57
5.56
5.37
5.36
4.90
4.88
4.87
4.11
4.11
4.11
4.10
4.10
4.09
4.09
3.84
3.82
3.81
3.80
3.59
3.58
3.58
3.57
3.56
3.55
3.54
3.50
3.48
3.47
3.45
3.44
O
O
O
OH
HO
OH
O
2
2.00
2.02
1.00
1.00
1.00
1.02
1.02
1.06
4.19
1.06
1.12
8.2
8.0
7.8
7.6
7.4
7.2
7.0
6.8
6.6
6.4
6.2
6.0
5.8
5.6
5.4
5.2
5.0
4.8
4.6
4.4
4.2
4.0
3.8
3.6
3.4
3.2
3.0
2.8
2.6
2.4
f1 (ppm)

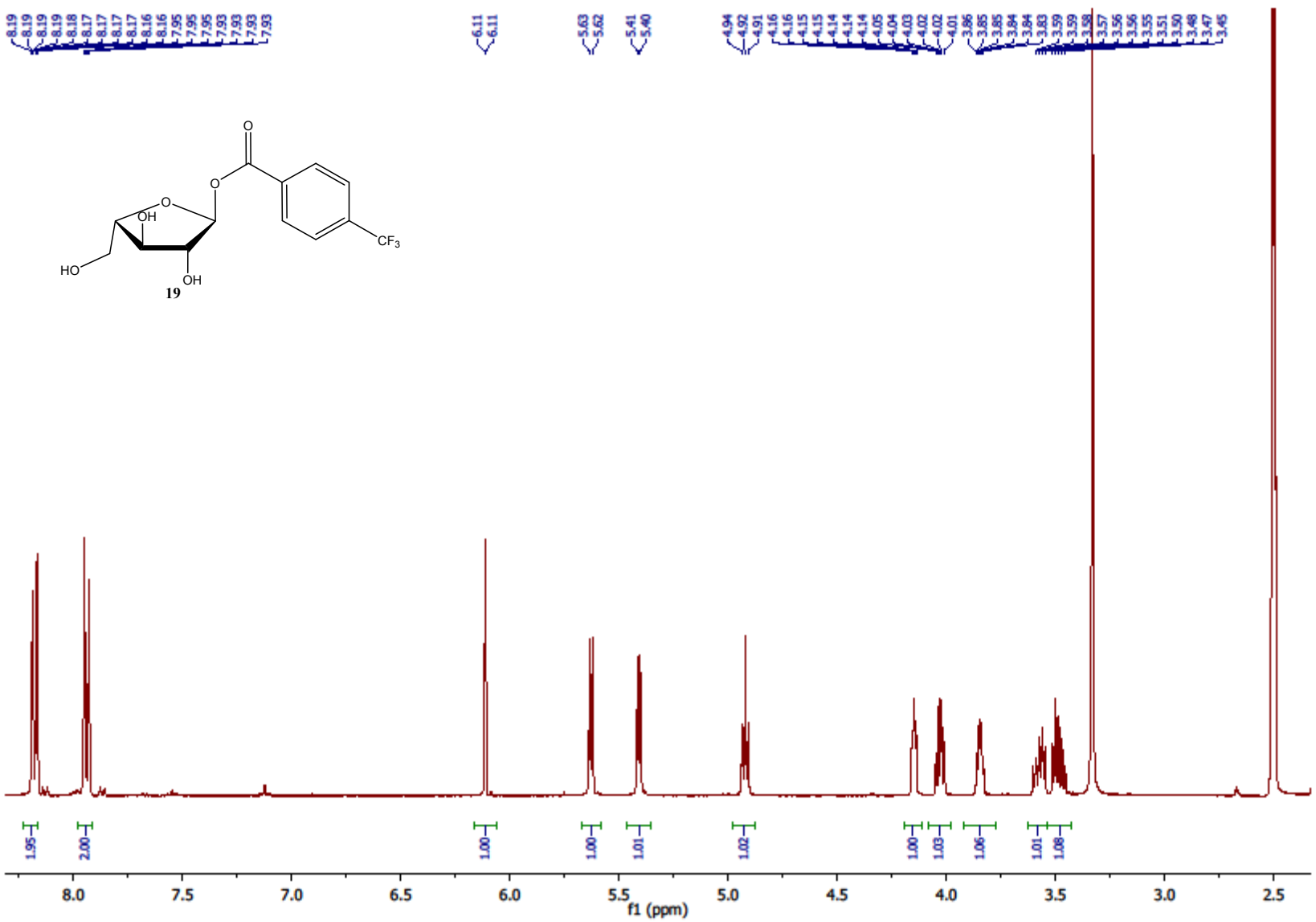

164.60
163.40
131.51
121.80
114.08
102.33
86.86
81.26
76.75
61.38
55.57
O
O
O
OH
HO
OH
O
2
165 160 155 150 145 140 135 130 125 120 115 110 105 100 95 90 85 80 75 70 65 60 55 50 45 40
f1 (ppm)

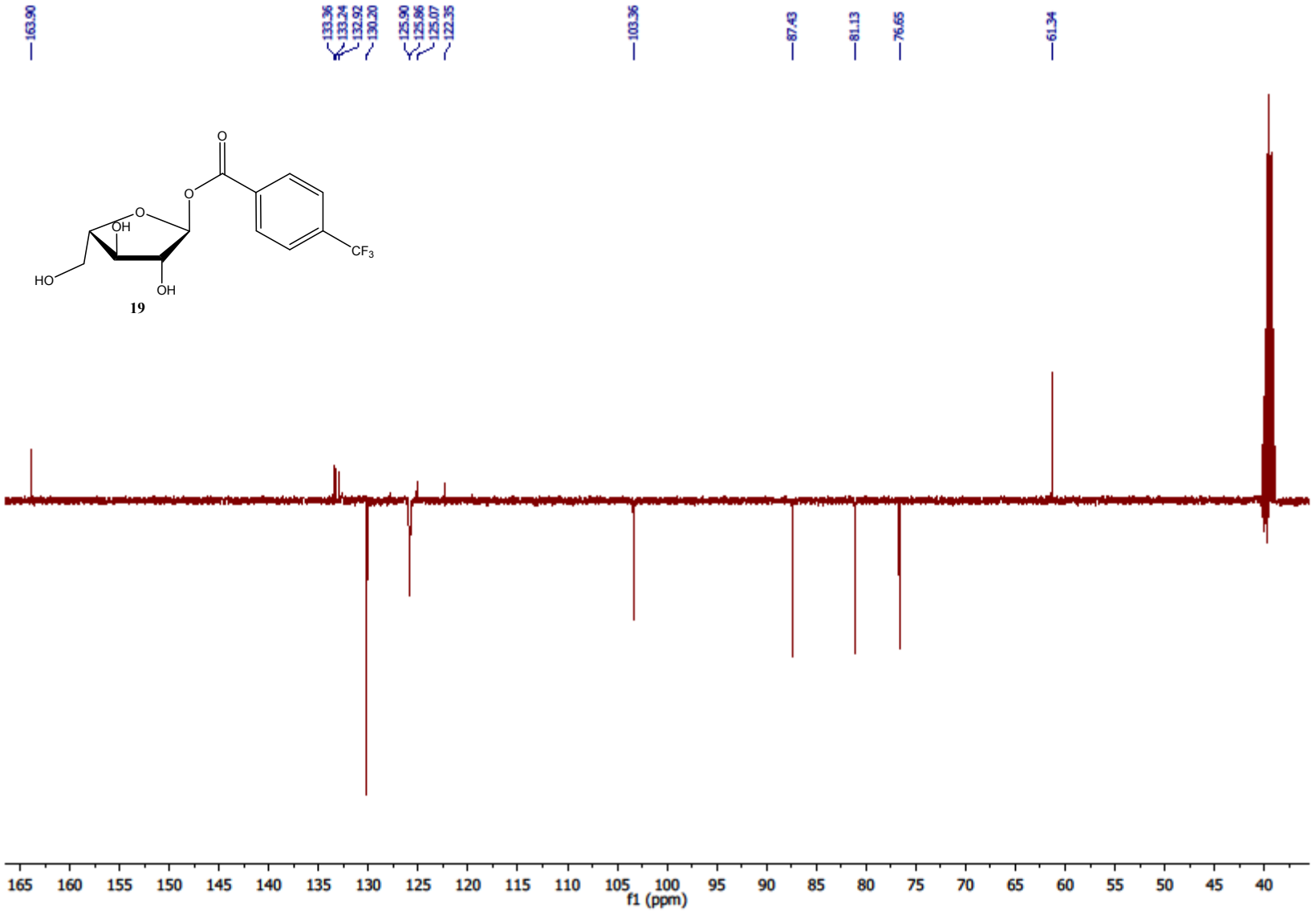

8.00 8.00 7.98 7.98 7.71 7.70 7.70 7.69 7.68 7.68 7.67 7.66 7.66 7.57 7.55 7.55 7.53
6.09 6.08
5.61 5.60
5.40 5.39
4.92 4.91 4.89
4.13 4.13 4.12 4.12 4.11 4.02 4.01 4.01 4.00 3.99 3.98 3.84 3.83 3.82 3.81 3.60 3.59 3.58 3.57 3.57 3.56 3.55 3.54 3.50 3.49 3.47 3.46 3.44
O
O
O
OH
HO
OH
17
1.98 1.00 2.09 1.00 0.97 0.98 1.01 1.11 1.09 1.11 1.27 1.54
8.0 7.8 7.6 7.4 7.2 7.0 6.8 6.6 6.4 6.2 6.0 5.8 5.6 5.4 5.2 5.0 4.8 4.6 4.4 4.2 4.0 3.8 3.6 3.4 3.2 3.0 2.8 2.6
f1 (ppm)

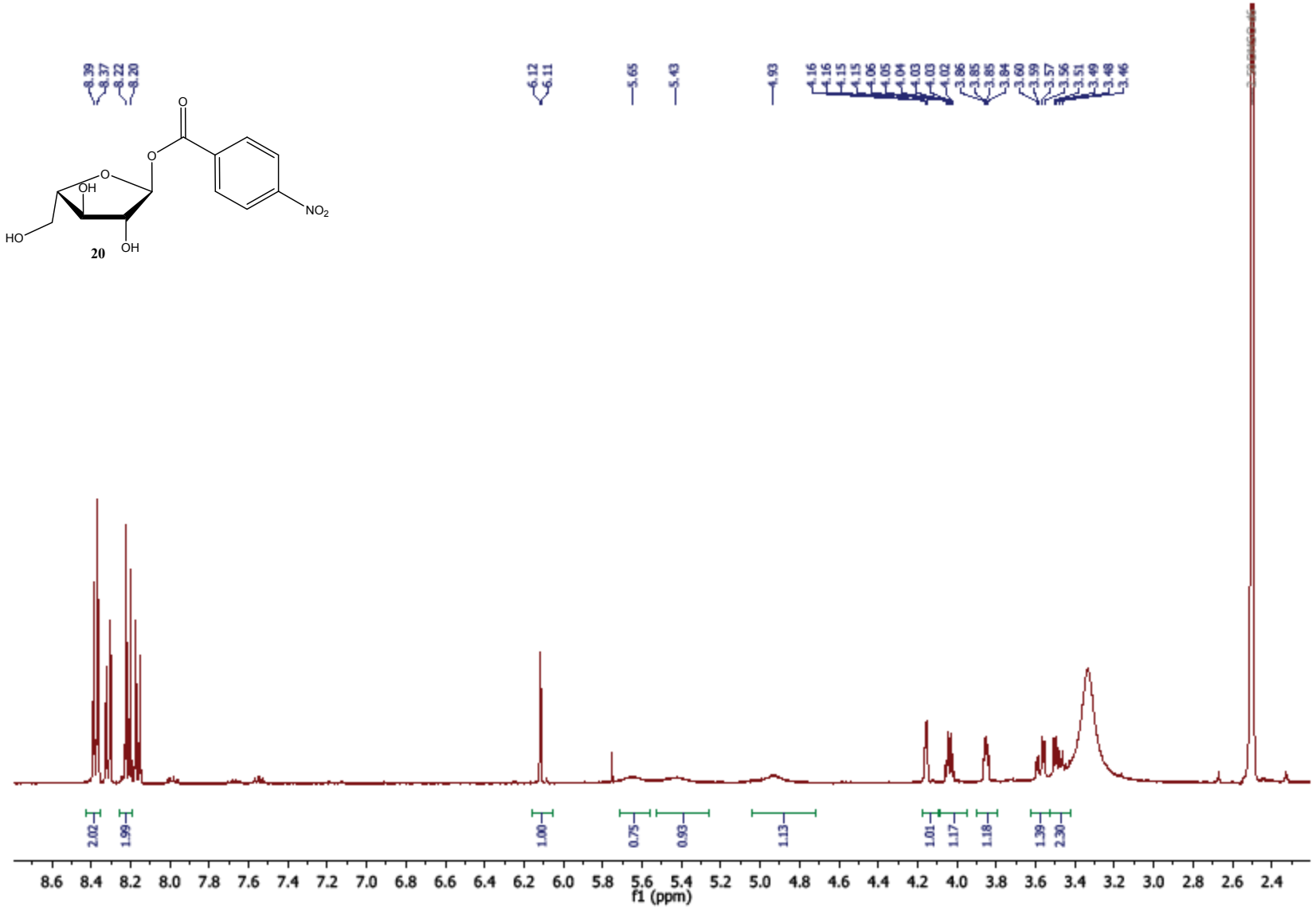

164.96
133.65
129.63
129.35
128.81
102.73
87.09
81.24
76.74
61.37
O
O
O
OH
HO
OH
17
165 160 155 150 145 140 135 130 125 120 115 110 105 100 95 90 85 80 75 70 65 60 55 50 45 40
f1 (ppm)

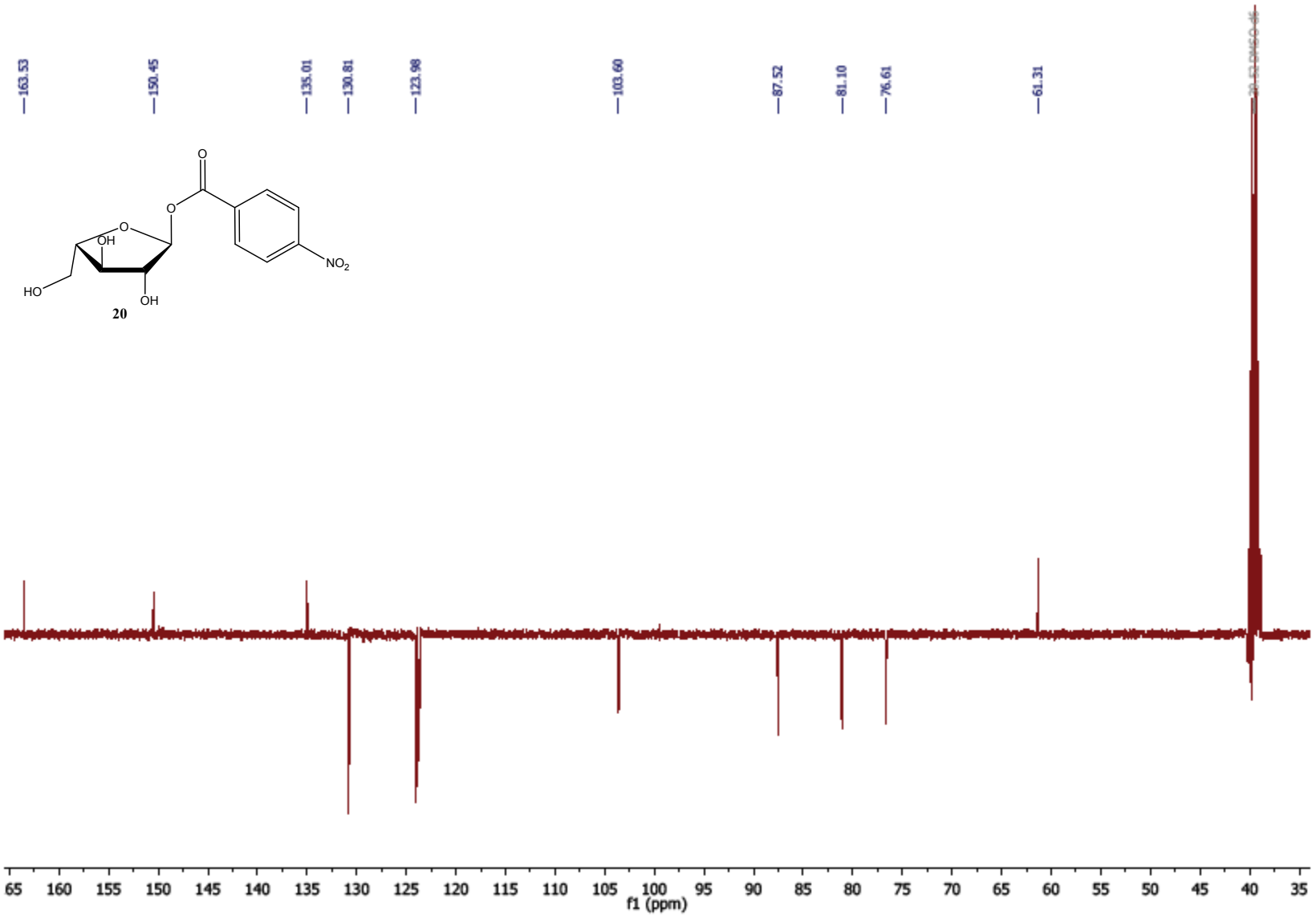

7.92
7.89
7.78
7.76
6.08
6.07
5.60
5.59
5.39
5.38
4.92
4.90
4.89
4.13
4.12
4.12
4.11
4.11
4.02
4.01
4.01
4.00
3.99
3.98
3.83
3.82
3.58
3.58
3.57
3.56
3.55
3.55
3.54
3.50
3.49
3.47
3.46
3.44
O
O
O
OH
HO
OH
Br
18
2.03
1.98
1.00
0.99
0.99
0.99
1.03
1.05
1.04
1.10
1.05
8.0
7.8
7.6
7.4
7.2
7.0
6.8
6.6
6.4
6.2
6.0
5.8
5.6
5.4
5.2
5.0
4.8
4.6
4.4
4.2
4.0
3.8
3.6
3.4
3.2
3.0
2.8
2.6
2.4
f1 (ppm)

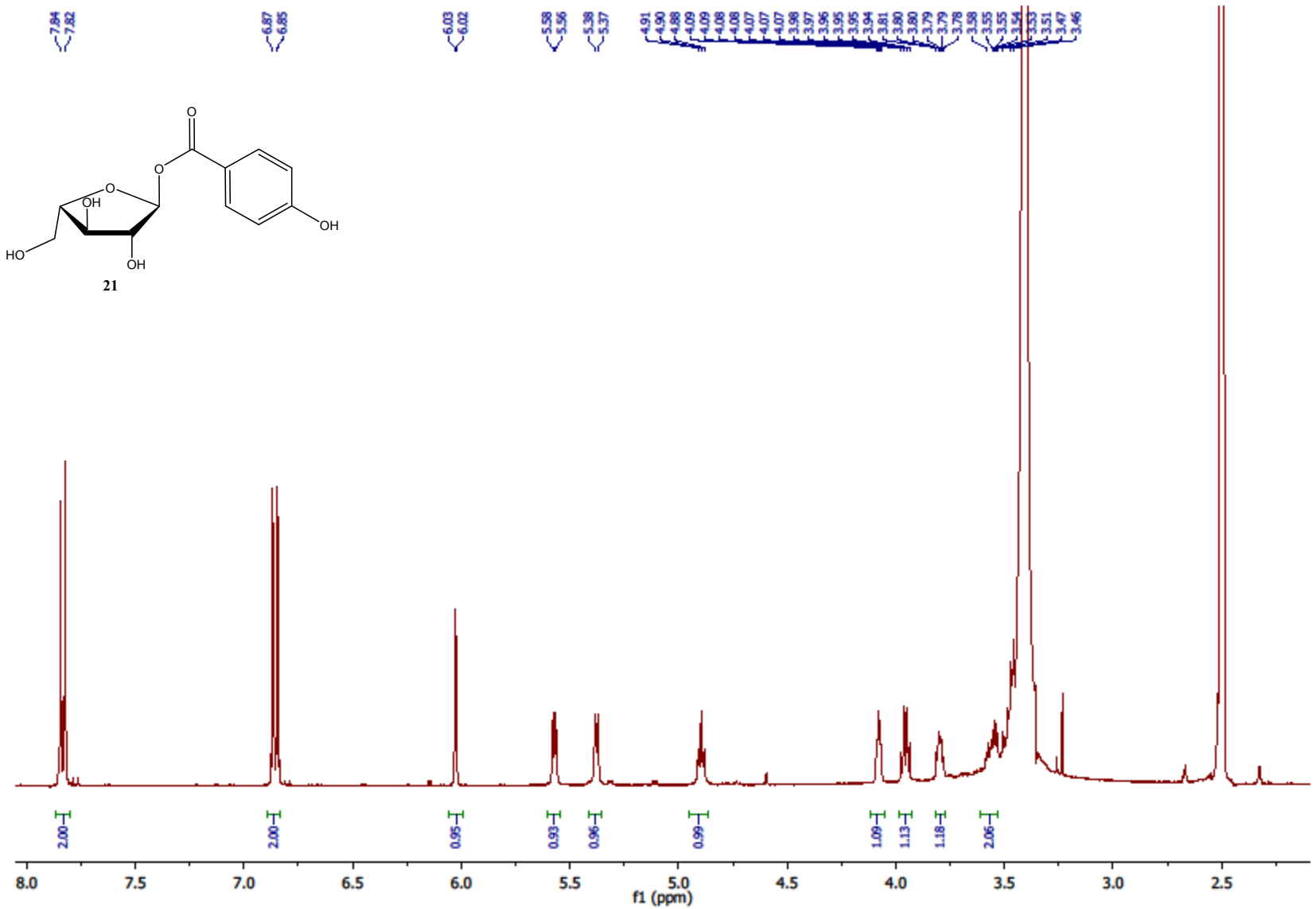

164.76
132.44
131.75
129.27
128.21
103.46
87.70
81.61
77.13
61.80
O
O
O
OH
HO
OH
Br
18
165 160 155 150 145 140 135 130 125 120 115 110 105 100 95 90 85 80 75 70 65 60 55 50 45 40
f1 (ppm)

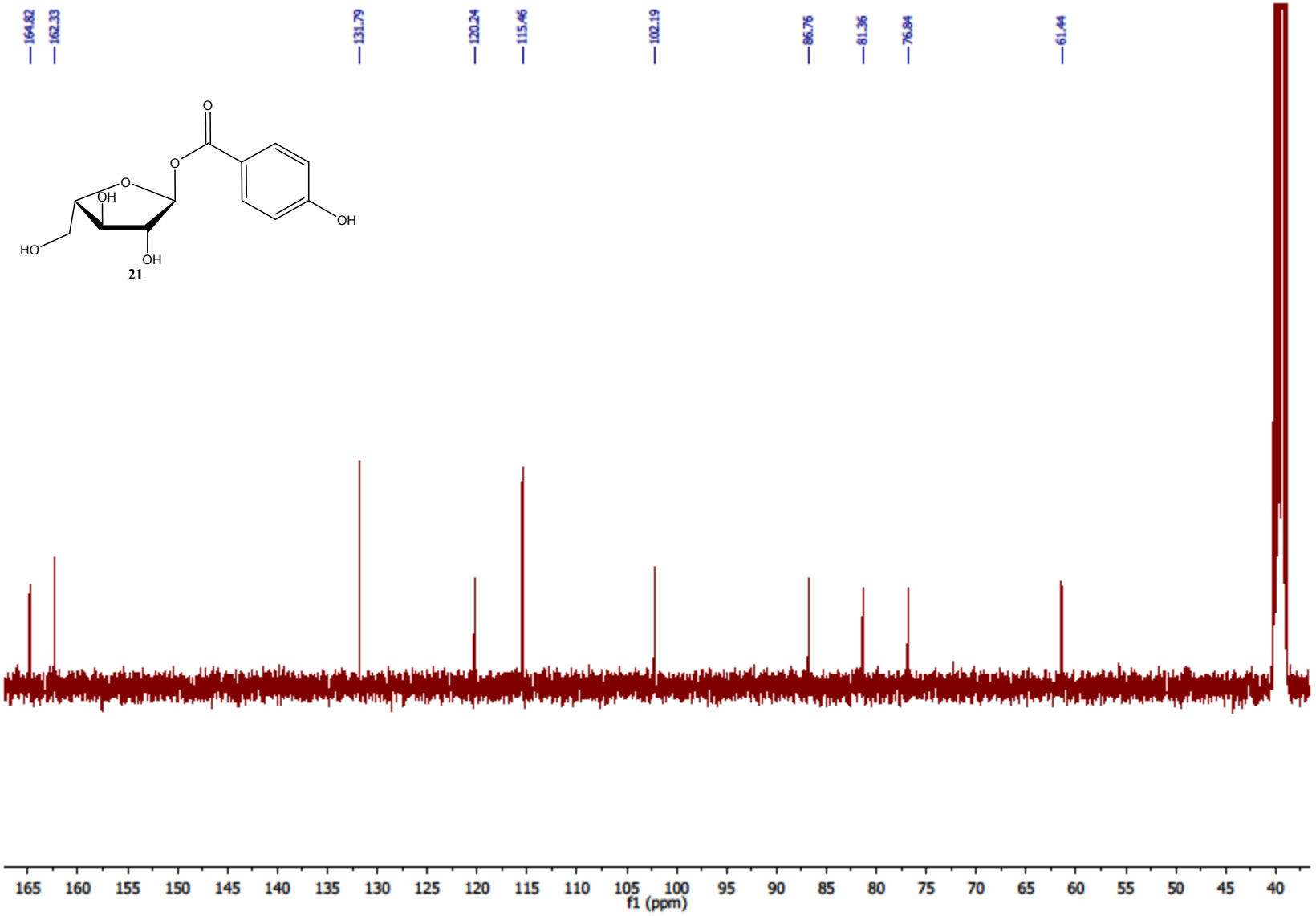

8.19 8.19 8.19 8.19 8.18 8.17 8.17 8.17 8.17 8.16 8.16 7.95 7.95 7.95 7.93 7.93 7.93 7.93
6.11 6.11
5.63 5.62
5.41 5.40
4.94 4.92 4.91 4.16 4.16 4.15 4.15 4.14 4.14 4.14 4.05 4.04 4.03 4.02 4.02 4.01
3.86 3.85 3.85 3.84 3.84 3.83 3.59 3.59 3.58 3.57 3.56 3.56 3.55 3.51 3.50 3.48 3.47 3.45
O
O
O
OH
HO
OH
CF3
19
1.95
2.00
1.00
1.00
1.01
1.02
1.00
1.03
1.06
1.01
1.08
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
f1 (ppm)

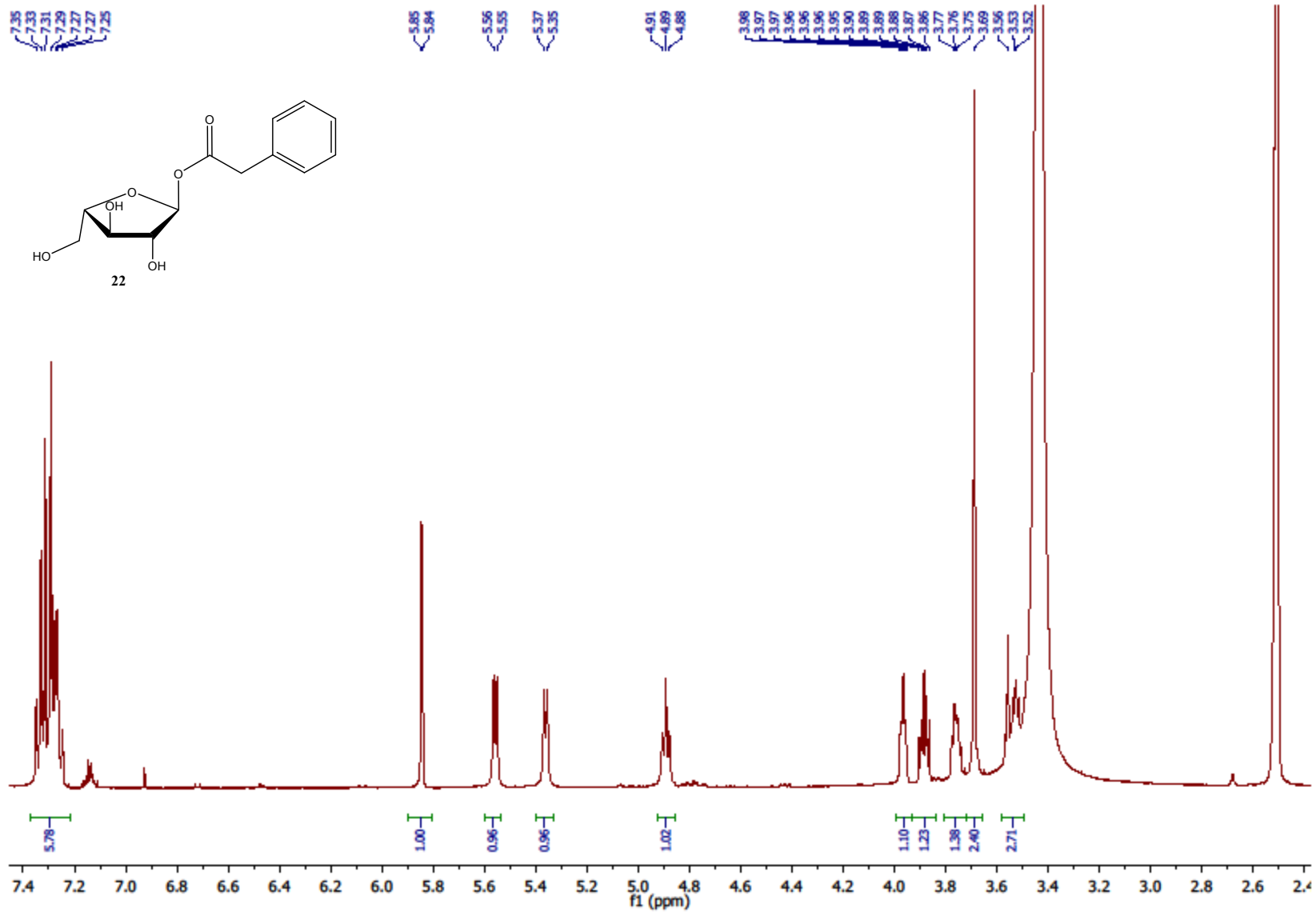

163.90
133.36
133.24
132.92
130.20
125.90
125.86
125.07
122.35
103.36
87.43
81.13
76.65
61.34
O
O
O
OH
HO
OH
CF3
19
165 160 155 150 145 140 135 130 125 120 115 110 105 100 95 90 85 80 75 70 65 60 55 50 45 40
f1 (ppm)

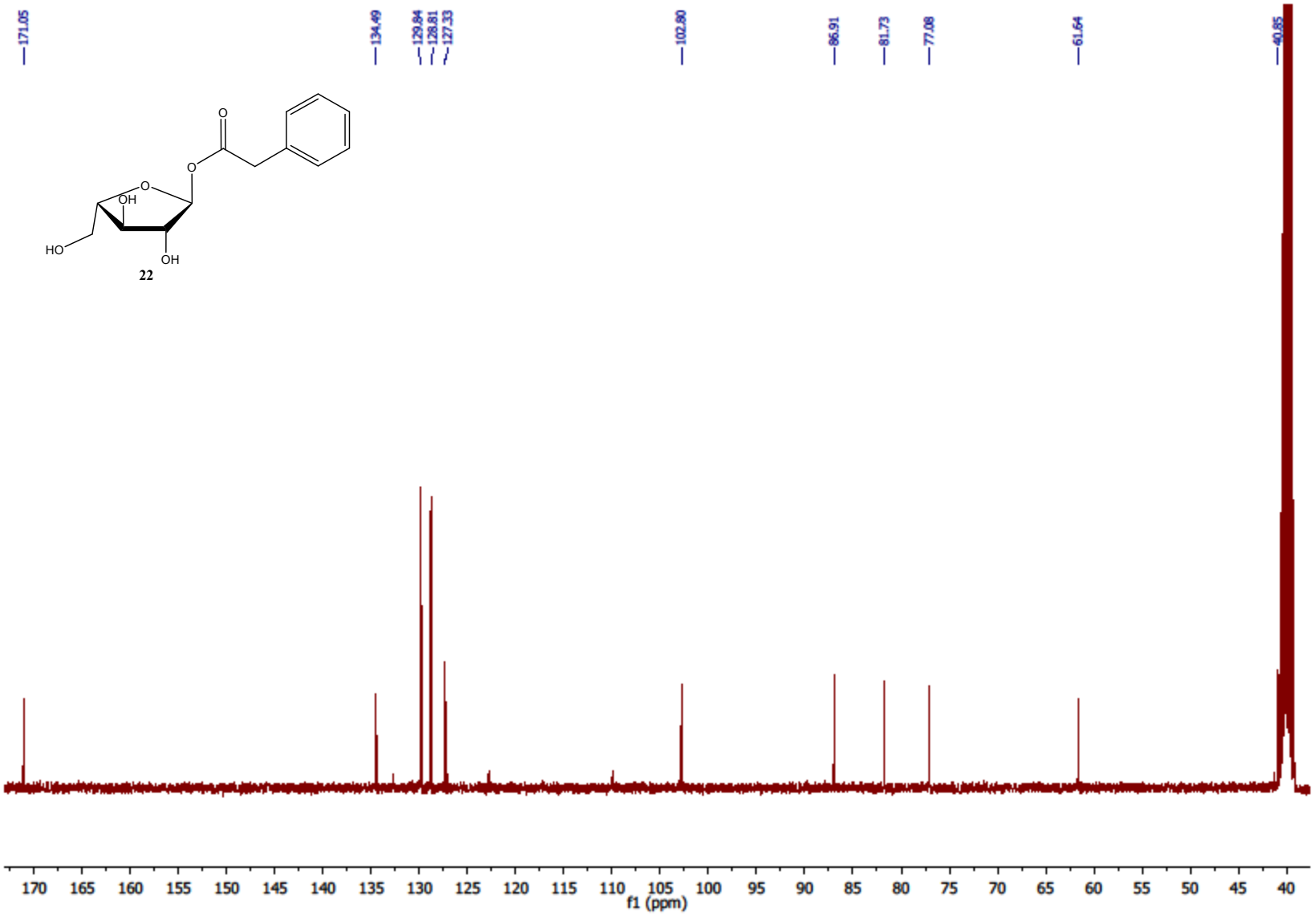

8.39
8.37
8.22
8.20
6.12
6.11
5.65
5.43
4.93
4.16
4.16
4.15
4.15
4.06
4.05
4.04
4.03
4.03
4.02
3.86
3.85
3.84
3.60
3.59
3.57
3.56
3.51
3.49
3.48
3.46
O
O
O
OH
HO
OH
NO2
20
2.02
1.99
1.00
0.75
0.93
1.13
1.01
1.17
1.18
1.39
2.30
8.6 8.4 8.2 8.0 7.8 7.6 7.4 7.2 7.0 6.8 6.6 6.4 6.2 6.0 5.8 5.6 5.4 5.2 5.0 4.8 4.6 4.4 4.2 4.0 3.8 3.6 3.4 3.2 3.0 2.8 2.6 2.4
f1 (ppm)

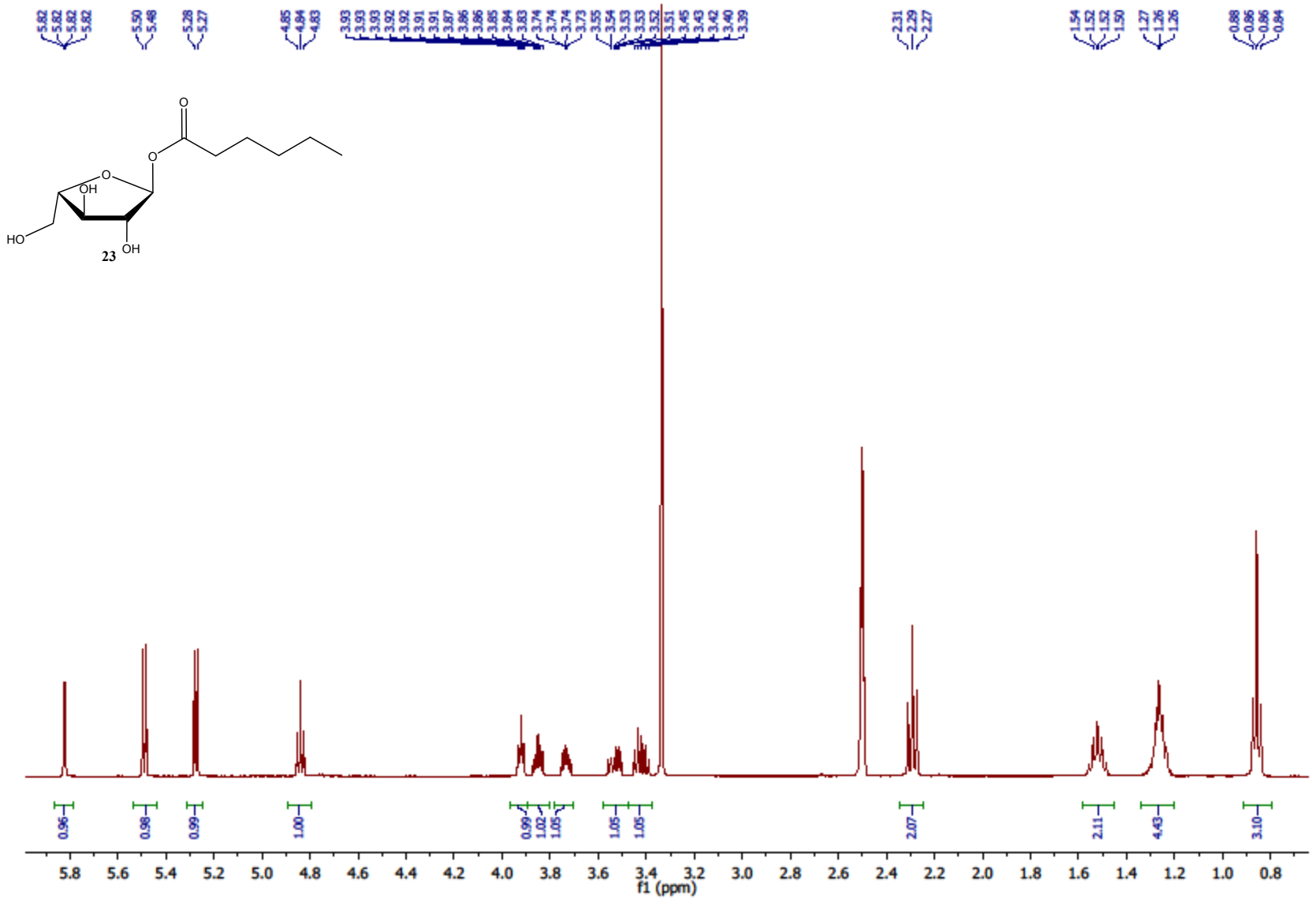

163.53
150.45
135.01
130.81
123.98
103.60
87.52
81.10
76.61
61.31
O
O
O
OH
HO
OH
NO2
20
65 160 155 150 145 140 135 130 125 120 115 110 105 100 95 90 85 80 75 70 65 60 55 50 45 40 35
f1 (ppm)

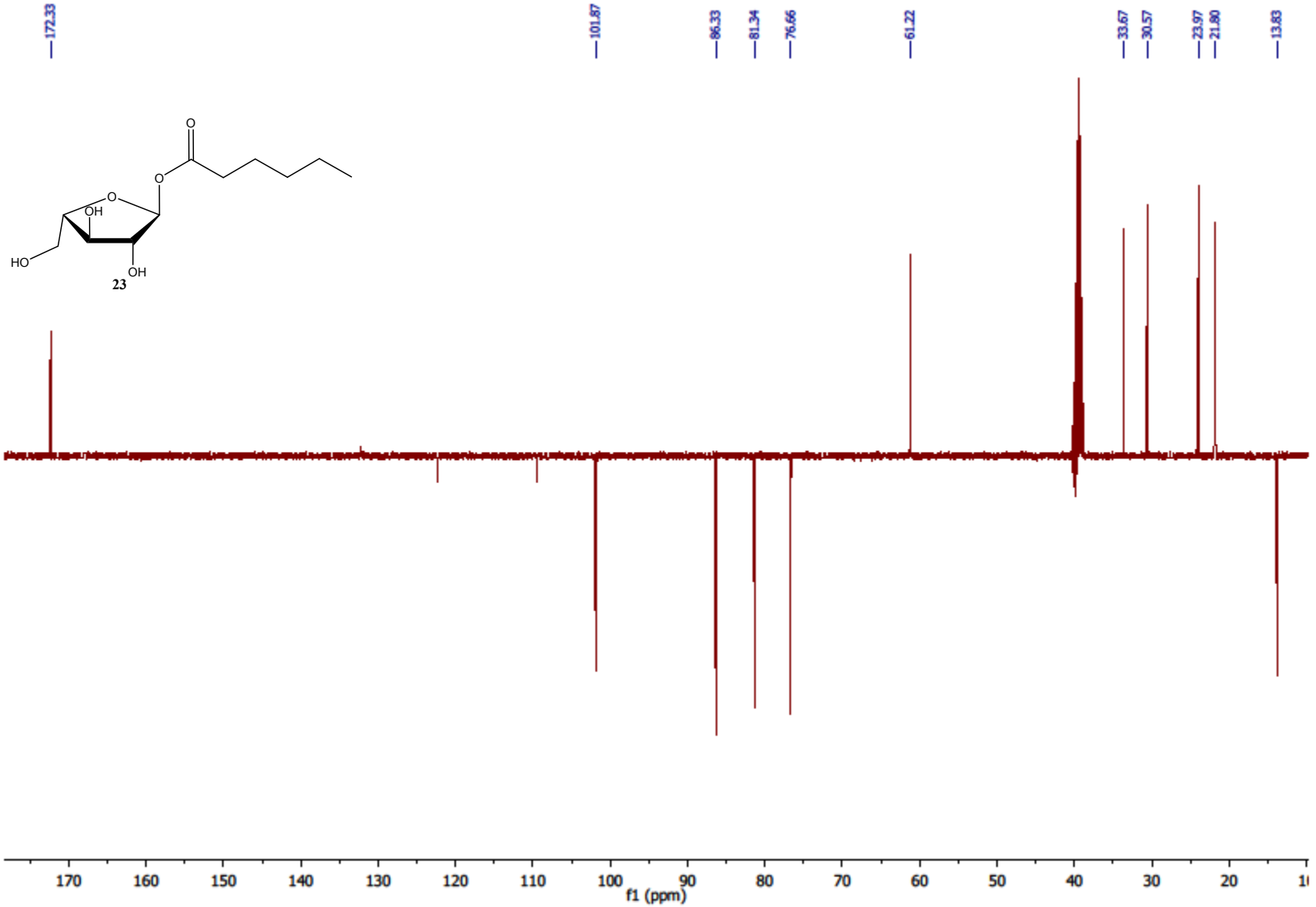

7.84
7.82
6.87
6.85
6.03
6.02
5.58
5.56
5.38
5.37
4.91
4.90
4.88
4.09
4.09
4.08
4.08
4.07
4.07
4.07
3.98
3.97
3.96
3.95
3.95
3.94
3.81
3.80
3.80
3.79
3.79
3.78
3.58
3.55
3.55
3.54
3.53
3.51
3.47
3.46
O
O
O
OH
HO
OH
OH
21
2.00
2.00
0.95
0.93
0.96
0.99
1.09
1.13
1.18
2.06
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
f1 (ppm)

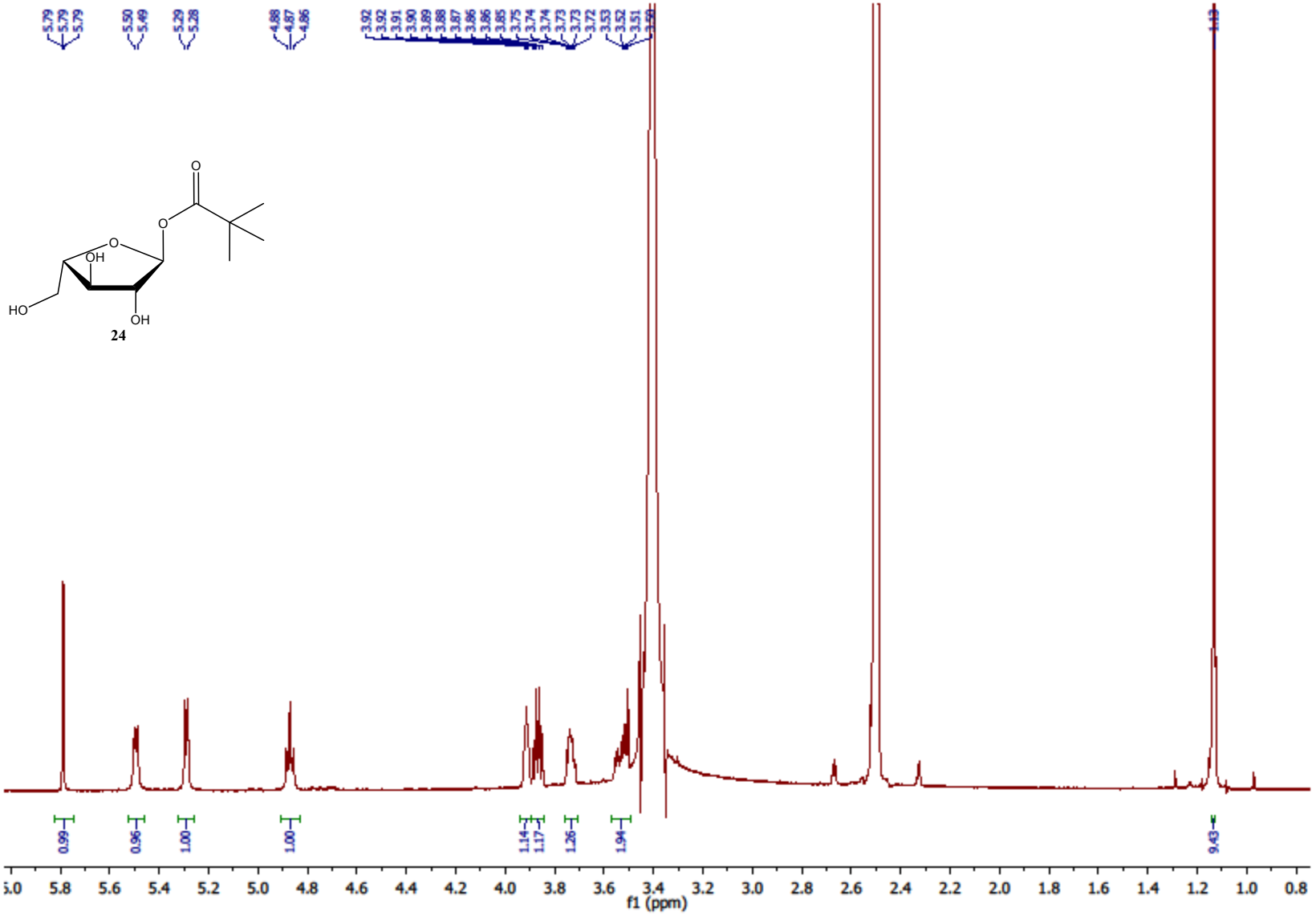

164.82
162.33
131.79
120.24
115.46
102.19
86.76
81.36
76.84
61.44
O
O
O
OH
OH
HO
OH
21
165 160 155 150 145 140 135 130 125 120 115 110 105 100 95 90 85 80 75 70 65 60 55 50 45 40
f1 (ppm)

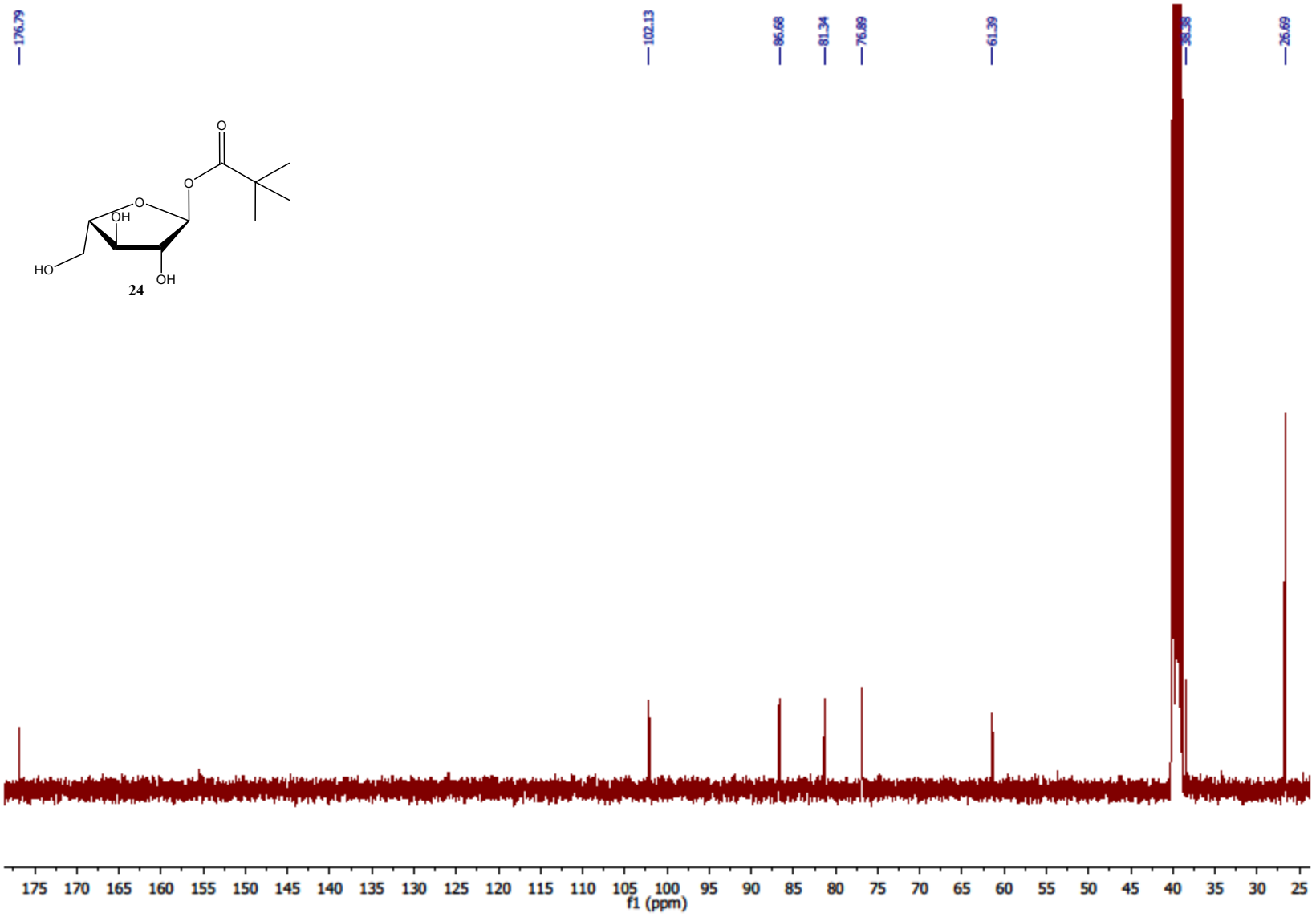

7.35
7.33
7.31
7.29
7.27
7.27
7.25
5.85
5.84
5.56
5.55
5.37
5.35
4.91
4.89
4.88
3.98
3.97
3.97
3.96
3.96
3.96
3.95
3.90
3.89
3.89
3.88
3.87
3.86
3.77
3.76
3.75
3.69
3.56
3.53
3.52
O
O
O
OH
HO
OH
22
5.78
1.00
0.96
0.96
1.02
1.10
1.23
1.38
2.40
2.71
7.4
7.2
7.0
6.8
6.6
6.4
6.2
6.0
5.8
5.6
5.4
5.2
5.0
4.8
4.6
4.4
4.2
4.0
3.8
3.6
3.4
3.2
3.0
2.8
2.6
f1 (ppm)

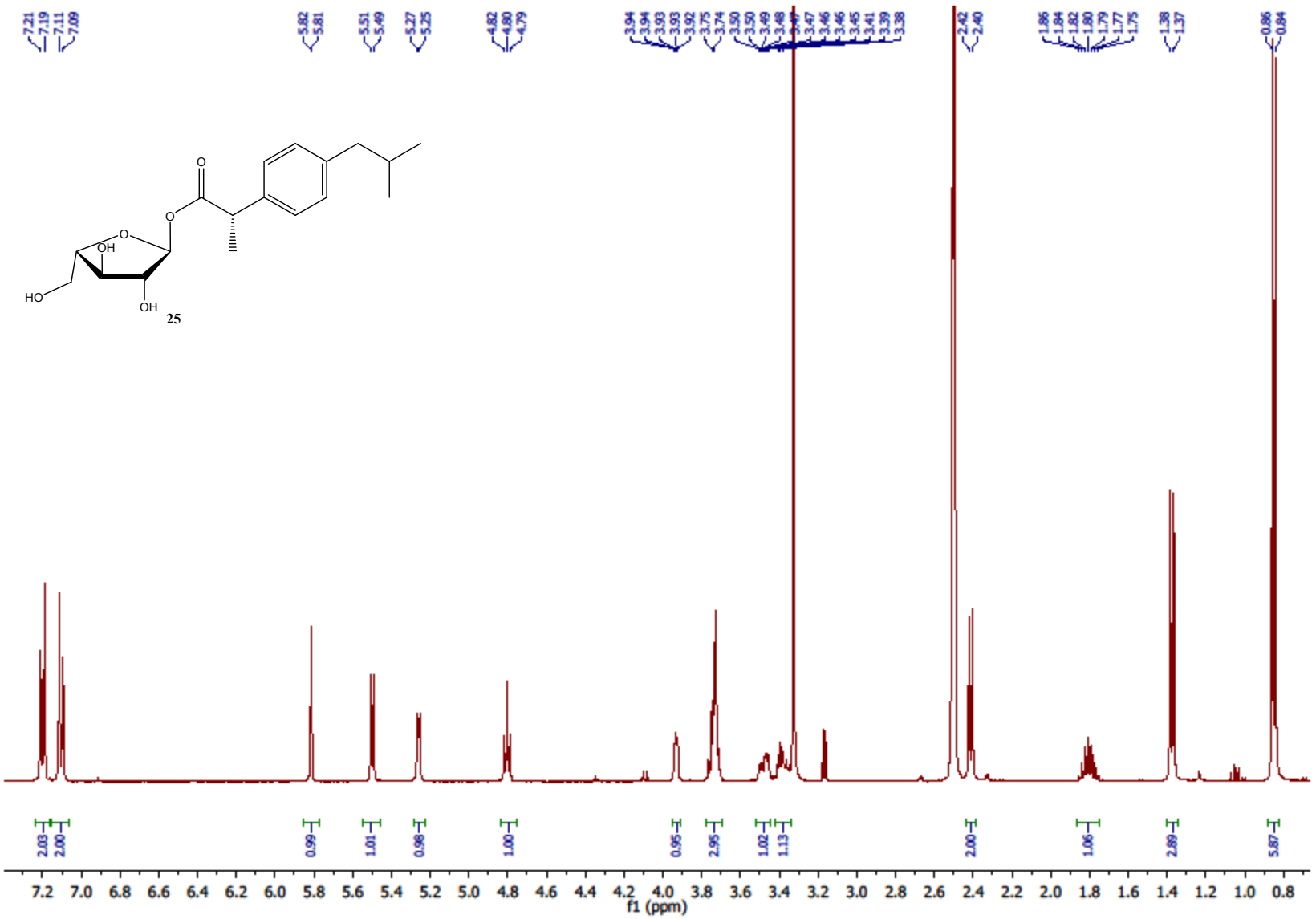

171.05
134.49
129.84
128.81
127.33
102.80
86.91
81.73
77.08
61.64
40.85
O
O
O
OH
HO
OH
22
170 165 160 155 150 145 140 135 130 125 120 115 110 105 100 95 90 85 80 75 70 65 60 55 50 45 40
f1 (ppm)

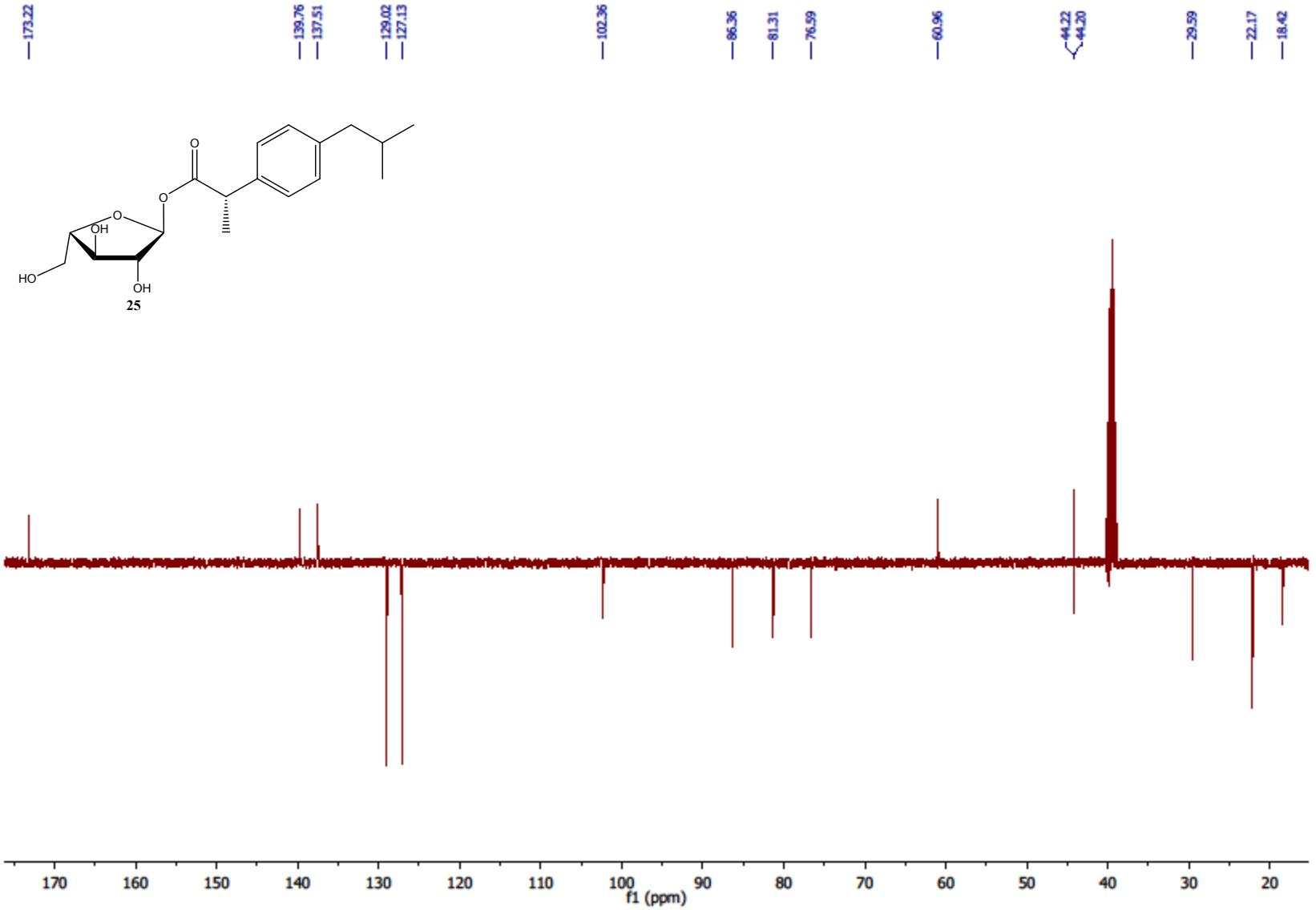

HO
OH
OH
O
O
O
23
f1 (ppm)

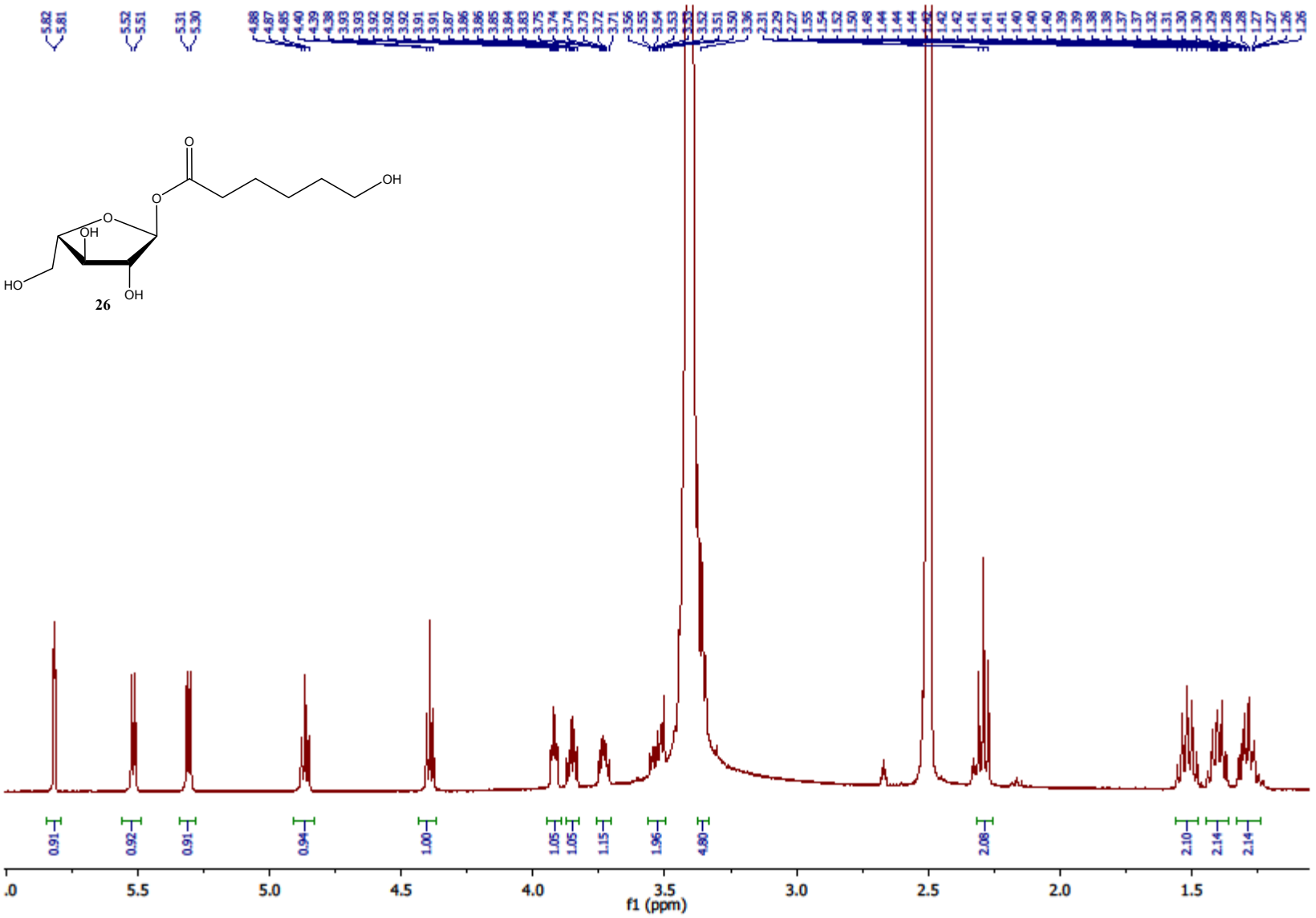

172.33
101.87
86.33
81.34
76.66
61.22
33.67
30.57
23.97
21.80
13.83
O
O
O
OH
HO
OH
23
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
f1 (ppm)

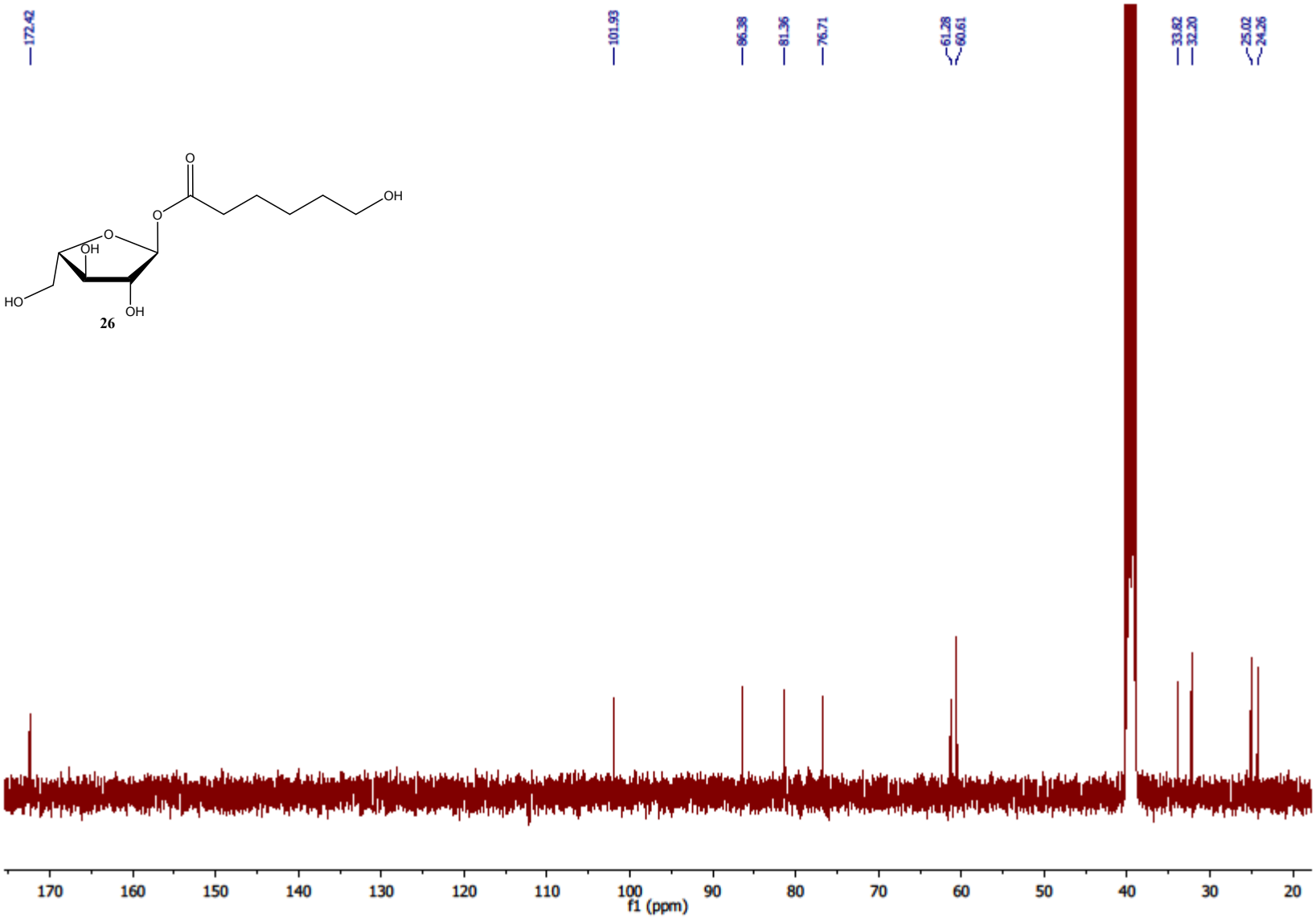

O
O
O
OH
HO
OH
24
f1 (ppm)

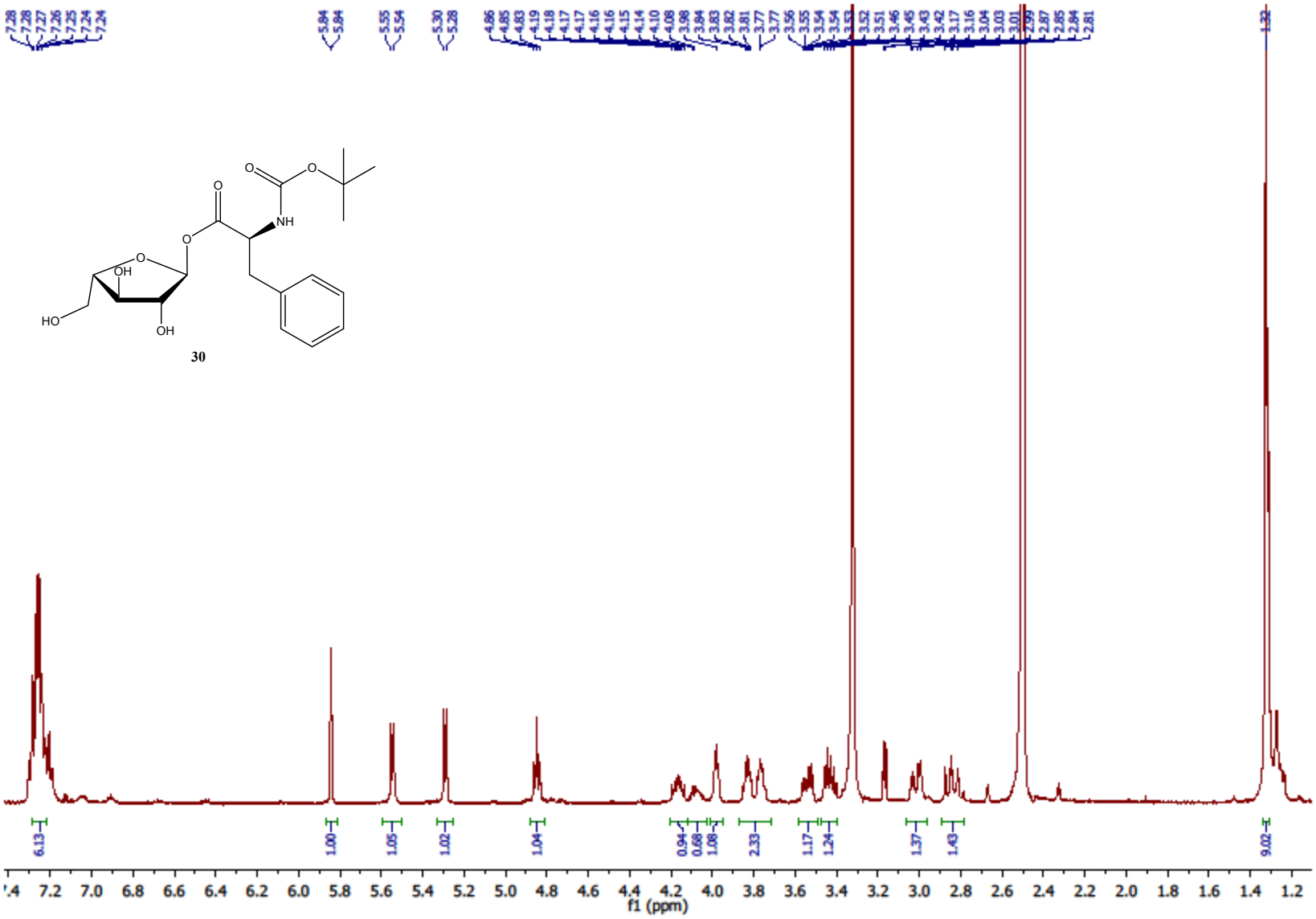

176.79
102.13
86.68
81.34
76.89
61.39
38.38
26.69
O
O
OH
HO
OH
24
175 170 165 160 155 150 145 140 135 130 125 120 115 110 105 100 95 90 85 80 75 70 65 60 55 50 45 40 35 30 25
f1 (ppm)

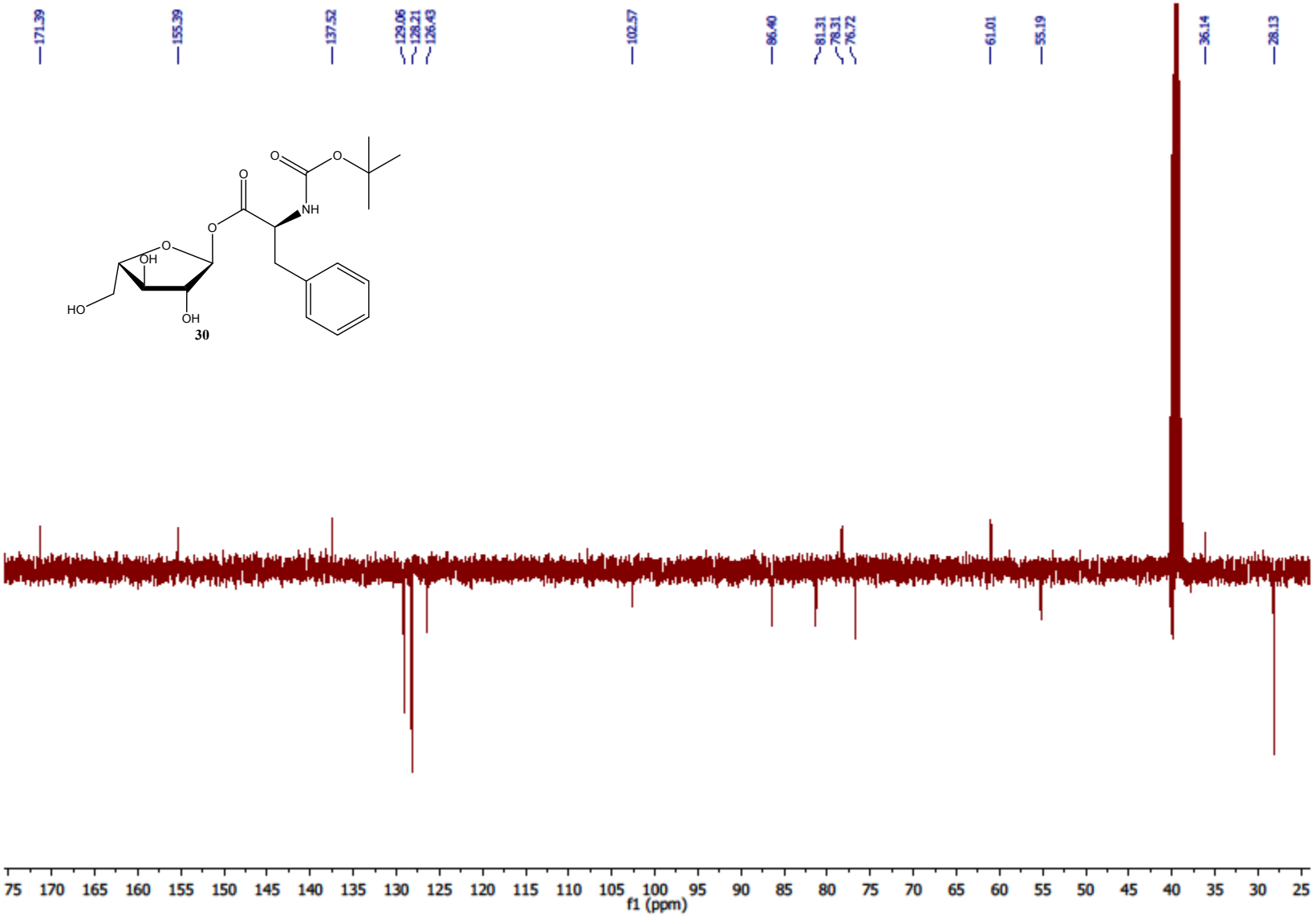

7.21
7.19
7.11
7.09
5.82
5.81
5.51
5.49
5.27
5.25
4.82
4.80
4.79
3.94
3.94
3.93
3.93
3.92
3.75
3.74
3.50
3.50
3.49
3.48
3.47
3.47
3.46
3.46
3.45
3.41
3.39
3.38
2.42
2.40
1.86
1.84
1.82
1.80
1.79
1.77
1.75
1.38
1.37
0.86
0.84
O
O
O
OH
HO
OH
25
2.03
2.00
0.99
1.01
0.98
1.00
0.95
2.95
1.02
1.13
2.00
1.06
2.89
5.87
7.2 7.0 6.8 6.6 6.4 6.2 6.0 5.8 5.6 5.4 5.2 5.0 4.8 4.6 4.4 4.2 4.0 3.8 3.6 3.4 3.2 3.0 2.8 2.6 2.4 2.2 2.0 1.8 1.6 1.4 1.2 1.0 0.8
f1 (ppm)


173.22
139.76
137.51
129.02
127.13
102.36
86.36
81.31
76.59
60.96
44.22
44.20
29.59
22.17
18.42
O
O
O
OH
HO
OH
25
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
f1 (ppm)


HO
OH
O
O
OH
O
OH
26
f1 (ppm)


172.42
101.93
86.38
81.36
76.71
61.28
60.61
33.82
32.20
25.02
24.26
O
O
OH
O
OH
HO
OH
26
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
f1 (ppm)


7.28
7.28
7.27
7.26
7.25
7.24
7.24
5.84
5.84
5.55
5.54
5.30
5.28
4.86
4.85
4.83
4.19
4.18
4.17
4.17
4.16
4.16
4.15
4.14
4.10
4.08
3.98
3.84
3.83
3.82
3.81
3.77
3.77
3.56
3.55
3.54
3.54
3.53
3.52
3.51
3.46
3.45
3.43
3.42
3.17
3.16
3.04
3.03
3.01
2.99
2.87
2.85
2.84
2.81
1.32
O
O
O
NH
O
O
OH
HO
OH
30
6.13
1.00
1.05
1.02
1.04
0.94
0.68
1.08
2.33
1.17
1.24
1.37
1.43
9.02
f1 (ppm)


171.39
155.39
137.52
129.06
128.21
126.43
102.57
86.40
81.31
78.31
76.72
61.01
55.19
36.14
28.13
O
O
O
NH
O
O
OH
HO
OH
30
75 170 165 160 155 150 145 140 135 130 125 120 115 110 105 100 95 90 85 80 75 70 65 60 55 50 45 40 35 30 25
f1 (ppm)

^{1}H NMR spectrum of (R,S)-Ibuprofen-α-L-arabinofuranose