

Supplementary Information

For

Enzymatic Route to Preparative-scale Synthesis of UDP-GlcNAc/GalNAc, their analogues and GDP-fucose

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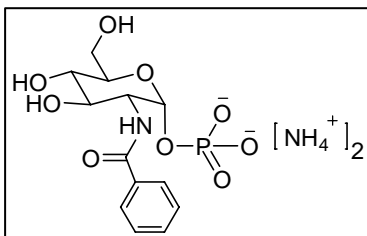
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c College of Pharmacy, Nankai University, Tianjin 300071, China

Correspondence should be addressed to P.G.W. (E-mail:wang.892@osu.edu)

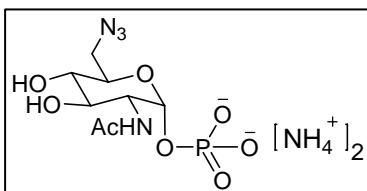
NMR, MS data and spectra for all new compounds



2-benzamido-2-deoxy- α -D-glucosyl diammonium

Phosphate: ^1H NMR (400 MHz, D_2O) δ 7.88-7.90 (m, 2H), 7.56-7.69 (m, 3H), 5.58 (dd, $J = 7.4, 3.3$ Hz, 1H), 4.23-4.26 (m, 1H), 3.94-4.06 (m, 3H), 3.86 (dd, $J = 12.3, 4.7$ Hz, 1H), 3.63 (app t, $J = 9.6$ Hz, 1H); ^{13}C NMR (100 MHz, D_2O) δ 171.6, 133.6, 132.2, 128.7, 127.5, 93.2 (d), 72.5, 71.1, 70.0, 60.6, 54.6 (d); ^{31}P NMR (162 MHz, D_2O)

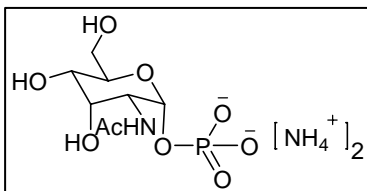
δ 0.71; HRMS (ESI) calcd for $\text{C}_{13}\text{H}_{17}\text{NO}_9\text{P}$ (M-H) $^-$ 362.0646, found 362.0643 m/z .



2-Acetamido-6-azido-2,6-dideoxy- α -D-glucosyl diammonium

Phosphate: ^1H NMR (400 MHz, D_2O) δ 5.42 (dd, $J = 7.2, 3.3$ Hz, 1H), 4.05-4.09 (m, 1H), 3.97-4.01 (m, 1H), 3.76-3.85 (m, 2H), 3.67 (dd, $J = 13.6, 4.2$ Hz, 1H), 3.59 (app t, $J = 9.3$ Hz, 1H), 2.10 (s, 3H); ^{13}C NMR (100 MHz, D_2O) δ 174.7, 93.1 (d), 71.0, 70.9, 70.4, 54.0

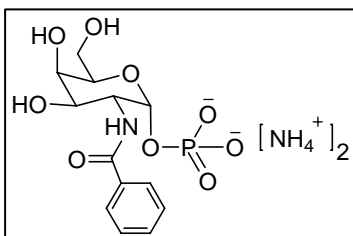
(d), 50.8, 22.0; ^{31}P NMR (162 MHz, D_2O) δ 0.60; HRMS (ESI) calcd for $\text{C}_8\text{H}_{14}\text{N}_4\text{O}_8\text{P}$ (M-H) $^-$ 325.0555, found 325.0559 m/z .



2-Acetamido-2-deoxy- α -D-allosyl diammonium

Phosphate: ^1H NMR (400 MHz, D_2O) δ 5.39 (dd, $J = 7.7, 3.6$ Hz, 1H), 4.02-4.13 (m, 3H), 3.94 (dd, $J = 12.3, 2.3$ Hz, 1H), 3.79 (dd, $J = 12.3, 5.3$ Hz, 1H), 3.70 (dd, $J = 10.5, 3.4$ Hz, 1H), 2.09 (s, 3H); ^{13}C NMR (100 MHz, D_2O) δ 174.1, 92.3 (d), 69.9, 67.3, 66.1, 60.9, 50.3 (d), 22.0; ^{31}P NMR

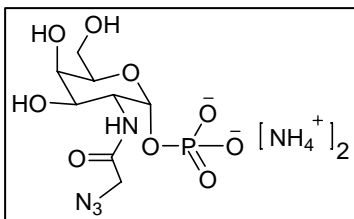
(162 MHz, D_2O) δ 2.17; HRMS (ESI) calcd for $\text{C}_8\text{H}_{15}\text{NO}_9\text{P}$ (M-H) $^-$ 300.0490, found 300.0481 m/z .



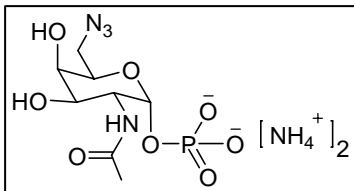
2-benzamido-2-deoxy- α -D-galactopyranosyl diammonium

Phosphate: ^1H NMR (400 MHz, D_2O): δ = 7.83-7.86 (m, 2H), 7.61-7.66 (m, 1H), 7.52-7.56 (m, 2H), 5.59 (dd, $J = 7.5, 3.5$ Hz, 1H), 4.45-4.49 (m, 1H), 4.23-4.26 (m, 1H), 4.09-4.15 (m, 2H), 3.77-3.83 (m, 2H); ^{13}C NMR (125 MHz, D_2O): δ = 171.8, 133.6, 132.2, 128.7, 127.5, 93.6

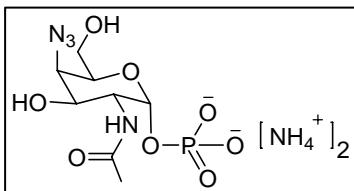
(d), 71.7, 68.7, 67.7, 61.3, 50.7 (d); ^{31}P NMR (162 MHz, D_2O): δ = -0.10; HRMS (ESI) calcd for $\text{C}_{13}\text{H}_{17}\text{NO}_9\text{P}$ (M-H) $^-$ 362.0646, found 362.0660 m/z .



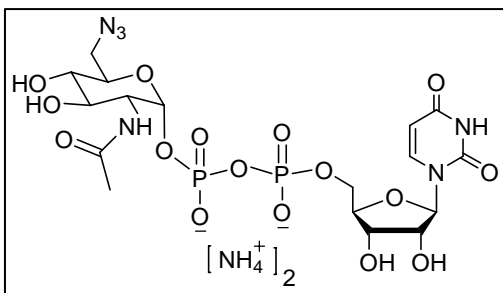
2-Azidoacetamido-2-deoxy- α -D-galactopyranosyl diammonium Phosphate: ^1H NMR (400 MHz, D_2O): $\delta = 5.30$ (dd, $J = 7.3, 3.3$ Hz, 1H), 4.06-4.10 (m, 1H), 3.86-4.00 (m, 4H), 3.81 (dd, $J = 11.1, 3.0$ Hz, 1H), 3.53-3.61 (m, 2H); ^{13}C NMR (100 MHz, D_2O): $\delta = 171.1, 93.4$ (d), 71.7, 68.4, 67.3, 61.1, 51.7, 50.1 (d); ^{31}P NMR (162 MHz, D_2O): $\delta = -0.74$; HRMS (ESI) calcd for $\text{C}_8\text{H}_{14}\text{N}_4\text{O}_9\text{P}$ (M-H) $^-$ 341.0504, found 341.0498 m/z .



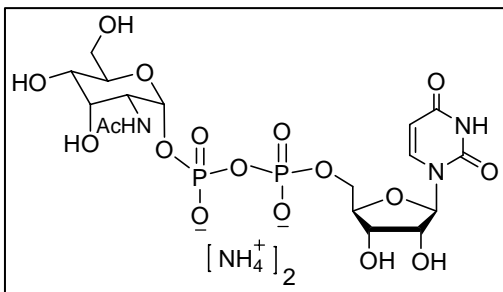
2-Acetamido-6-azido-2,6-dideoxy- α -D-galactopyranosyl diammonium Phosphate: ^1H NMR (400 MHz, D_2O): $\delta = 5.42$ (dd, $J = 7.3, 3.2$ Hz, 1H), 4.25 (app t, $J = 6.6$, 1H), 4.18-4.21 (m, 1H), 3.95-4.03 (m, 2H), 3.58-3.63 (m, 1H), 3.50-3.54 (m, 1H), 2.07 (s, 3H); ^{13}C NMR (100 MHz, D_2O): $\delta = 174.9, 93.3$ (d), 69.5, 68.6, 67.8, 50.5, 50.0 (d), 22.1; ^{31}P NMR (162 MHz, D_2O): $\delta = 0.58$; HRMS (ESI) calcd for $\text{C}_8\text{H}_{14}\text{N}_4\text{O}_8\text{P}$ (M-H) $^-$ 325.0555, found 325.0566 m/z .



2-Acetamido-4-azido-2,4-dideoxy- α -D-galactopyranosyl diammonium Phosphate: ^1H NMR (400 MHz, D_2O): $\delta = 5.43$ (dd, $J = 7.4, 3.0$ Hz, 1H), 4.22-4.27 (m, 3H), 4.15-4.16 (m, 1H), 3.75-3.78 (m, 2H), 2.06 (s, 3H); ^{13}C NMR (100 MHz, D_2O): $\delta = 174.8, 93.4$ (d), 70.1, 67.8, 62.8, 61.1, 50.2 (d), 22.0; ^{31}P NMR (162 MHz, D_2O): $\delta = -0.95$; HRMS (ESI) calcd for $\text{C}_8\text{H}_{14}\text{N}_4\text{O}_8\text{P}$ (M-H) $^-$ 325.0555, found 325.0542 m/z .

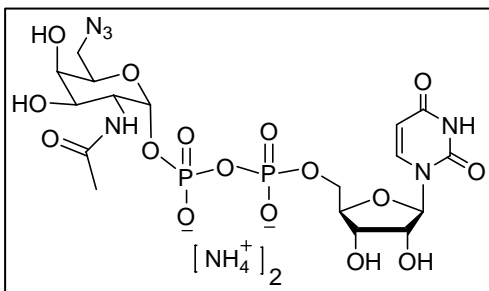


Uridine 5'-diphospho-2-acetamido-6-azido-2,6-dideoxy- α -D-glucopyranose diammonium salt: ^1H NMR (400 MHz, D_2O): $\delta = 7.98$ (d, $J = 8.1$ Hz, 1H), 5.97-5.99 (m, 2H), 5.51 (dd, $J = 7.2, 3.3$ Hz, 1H), 4.36-4.40 (m, 2H), 4.20-4.30 (m, 3H), 4.00-4.09 (m, 2H), 3.81 (t, $J = 9.2$ Hz, 1H), 3.74 (dd, $J = 13.5, 2.5$ Hz, 1H), 3.57-3.65 (m, 2H), 2.08 (s, 3H); ^{31}P NMR (162 MHz, D_2O): $\delta = -11.3$ (d, $J = 21.4$ Hz), -13.2 (d, $J = 21.4$ Hz); HRMS (ESI) calcd for $\text{C}_{17}\text{H}_{25}\text{N}_6\text{O}_{16}\text{P}_2$ (M-H) $^-$ 631.0808, found 631.0806 m/z .



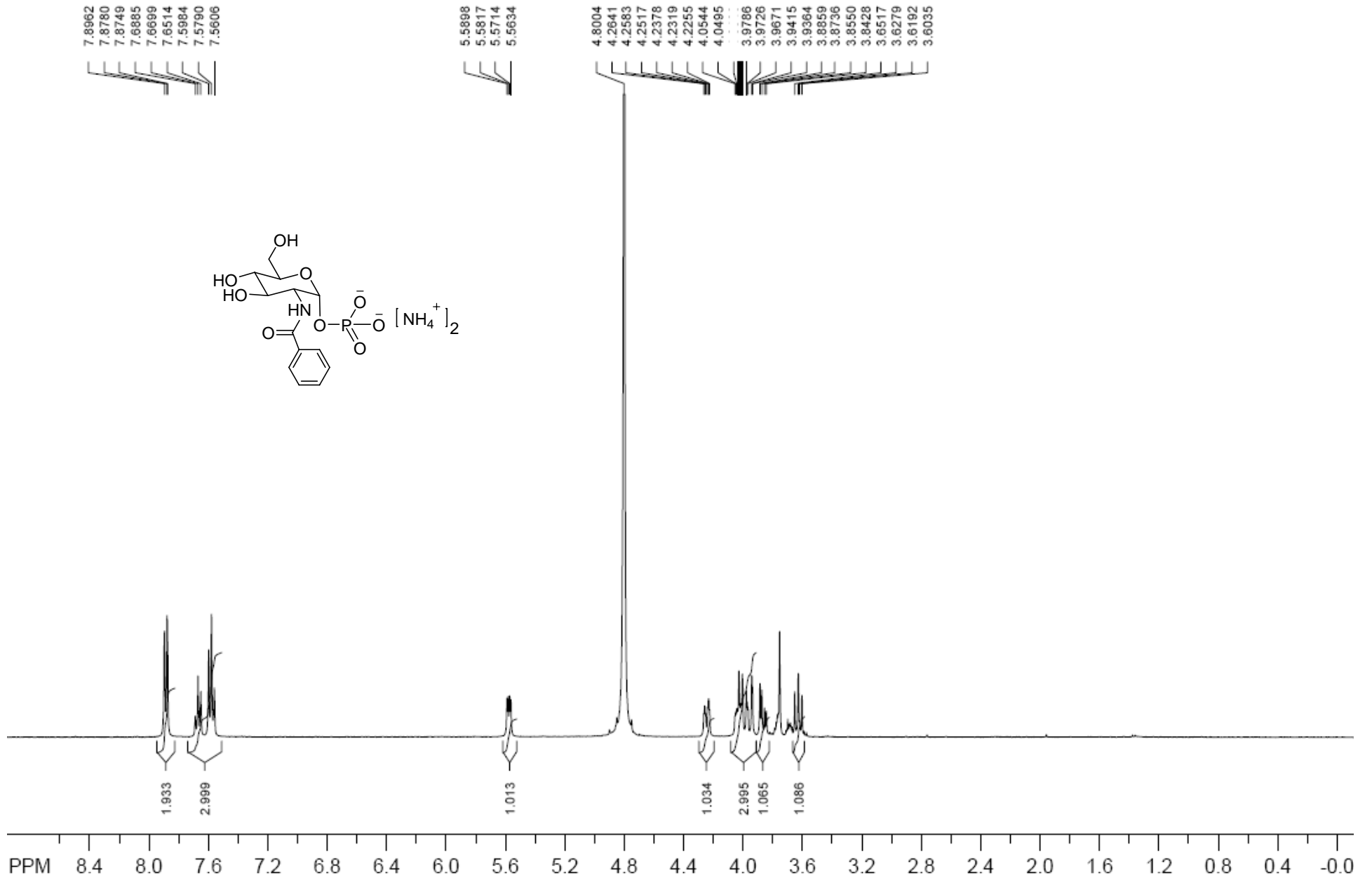
Uridine 5'-diphospho-2-acetamido-2-deoxy- α -D-allopyranose diammonium salt: ^1H NMR (400 MHz, D_2O): $\delta = 7.96$ (d, $J = 8.1$ Hz, 1H), 5.96-6.00 (m, 2H), 5.53 (dd, $J = 7.1, 3.8$ Hz, 1H), 4.36-4.40 (m, 2H), 4.21-4.30 (m, 3H), 4.08-4.15 (m, 3H), 3.92 (dd, $J = 12.6, 2.3$ Hz, 1H), 3.82 (dd, $J = 12.6, 4.5$ Hz, 1H), 3.77 (dd, $J = 10.5, 3.2$ Hz, 1H), 2.10 (s, 3H); ^{31}P NMR (162

MHz, D_2O): $\delta = -11.0$ (d, $J = 19.9$ Hz), -13.0 (d, $J = 21.7$ Hz); HRMS (ESI) calcd for $\text{C}_{17}\text{H}_{26}\text{N}_3\text{O}_{17}\text{P}_2$ (M-H) $^-$ 606.0743, found 606.0730 m/z .

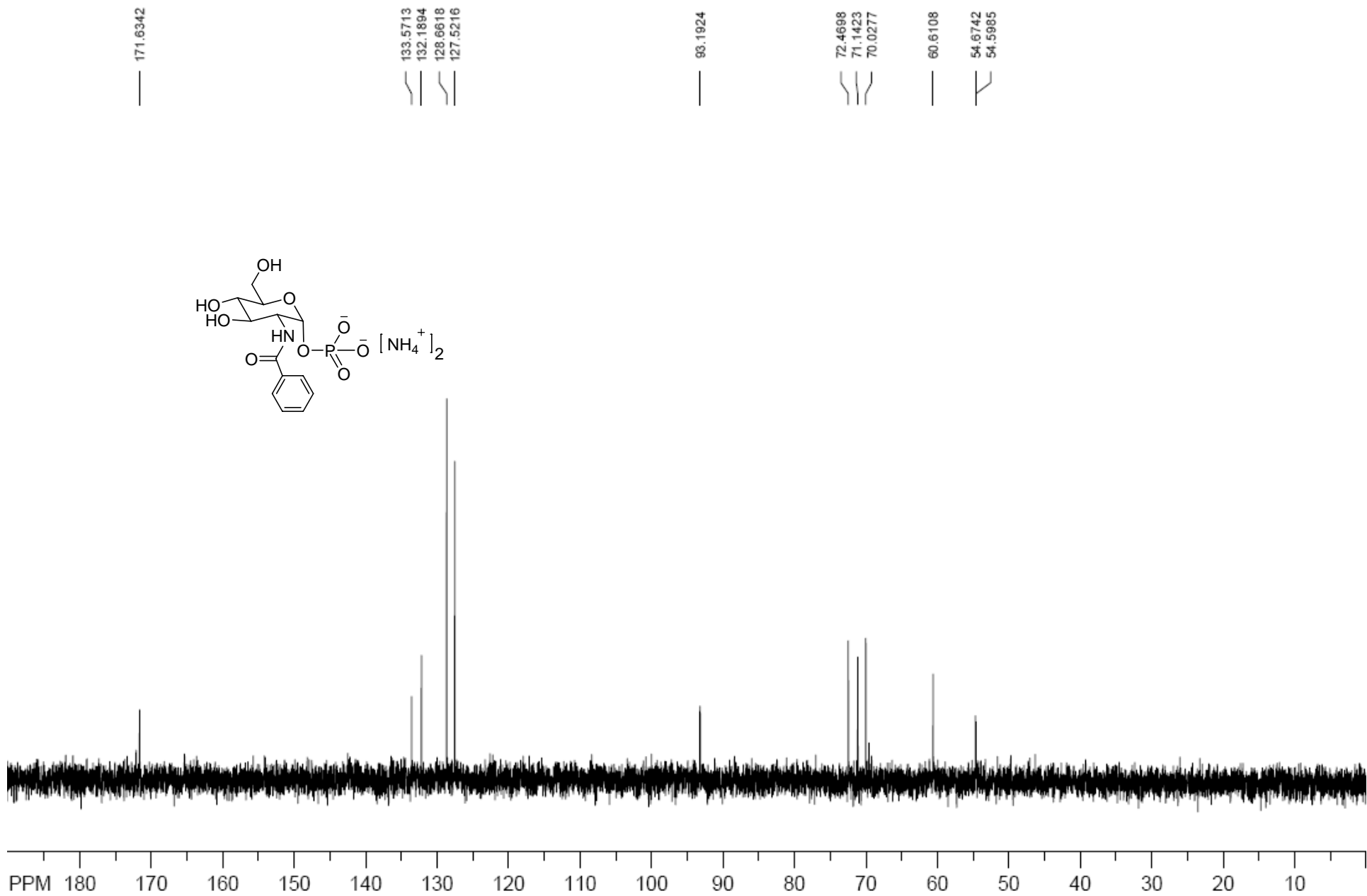
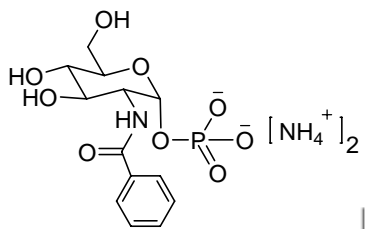


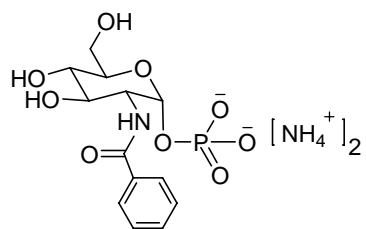
Uridine 5'-diphospho-2-acetamido-6-azido-2,6-dideoxy- α -D-galactopyranose diammonium salt: ^1H NMR (400 MHz, D_2O): $\delta = 7.98$ (d, $J = 8.2$ Hz, 1H), 5.97-5.99 (m, 2H), 5.54 (dd, $J = 7.2, 3.4$ Hz, 1H), 4.35-4.39 (m, 2H), 4.19-4.30 (m, 5H), 4.02 (m, 1H), 3.98 (dd, $J = 10.8, 3.2$ Hz, 1H), 3.60 (dd, $J = 12.8, 7.2$ Hz, 1H), 3.50 (dd, $J = 12.8, 6.0$ Hz, 1H), 2.09 (s, 3H); ^{31}P NMR (162

MHz, D_2O): $\delta = -11.2$ (d, $J = 21.5$ Hz), -13.0 (d, $J = 21.5$ Hz); HRMS (ESI) calcd for $\text{C}_{17}\text{H}_{25}\text{N}_6\text{O}_{16}\text{P}_2$ (M-H) $^-$ 631.0808, found 631.0801 m/z .



S5



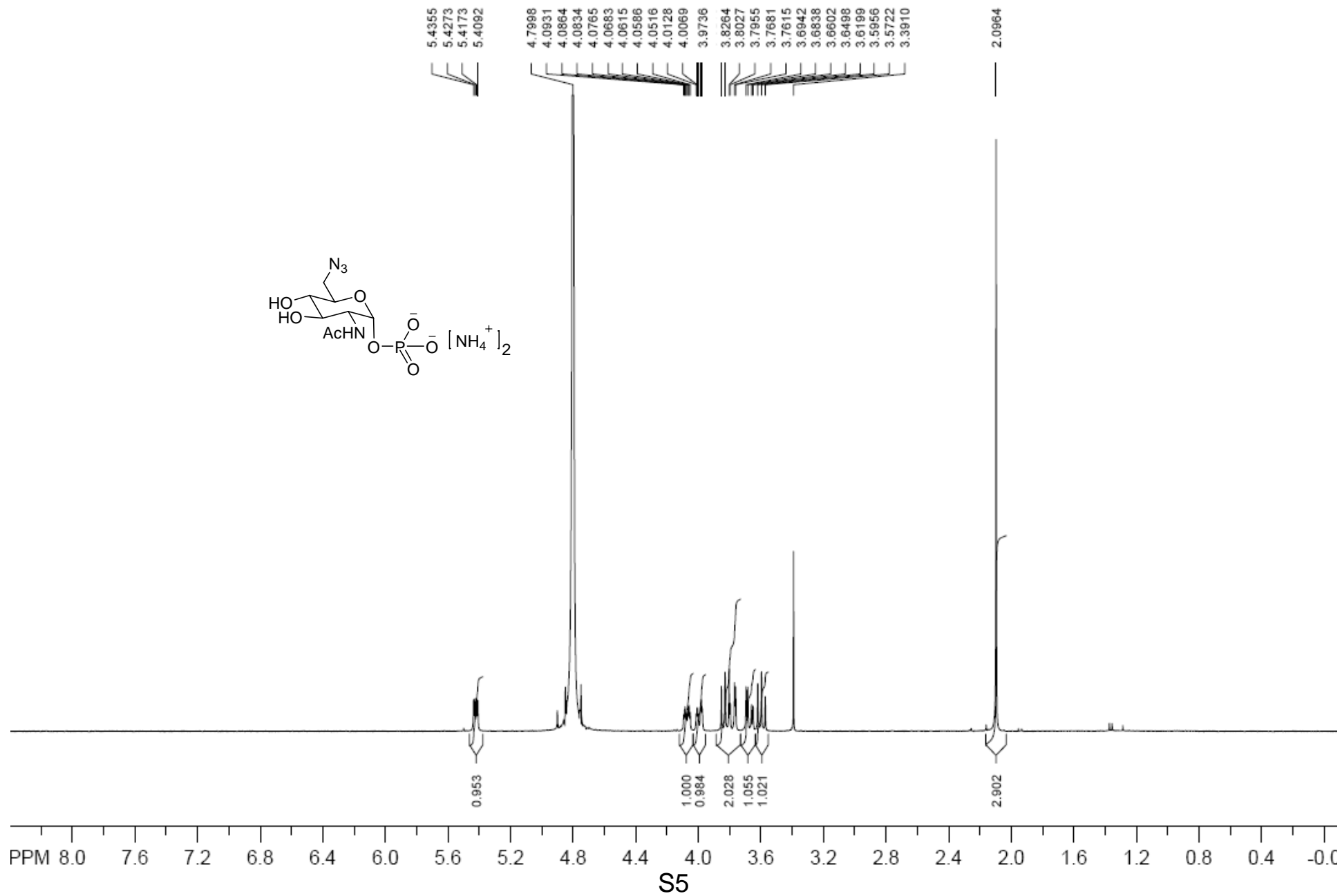
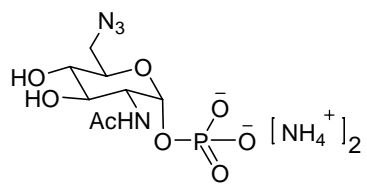


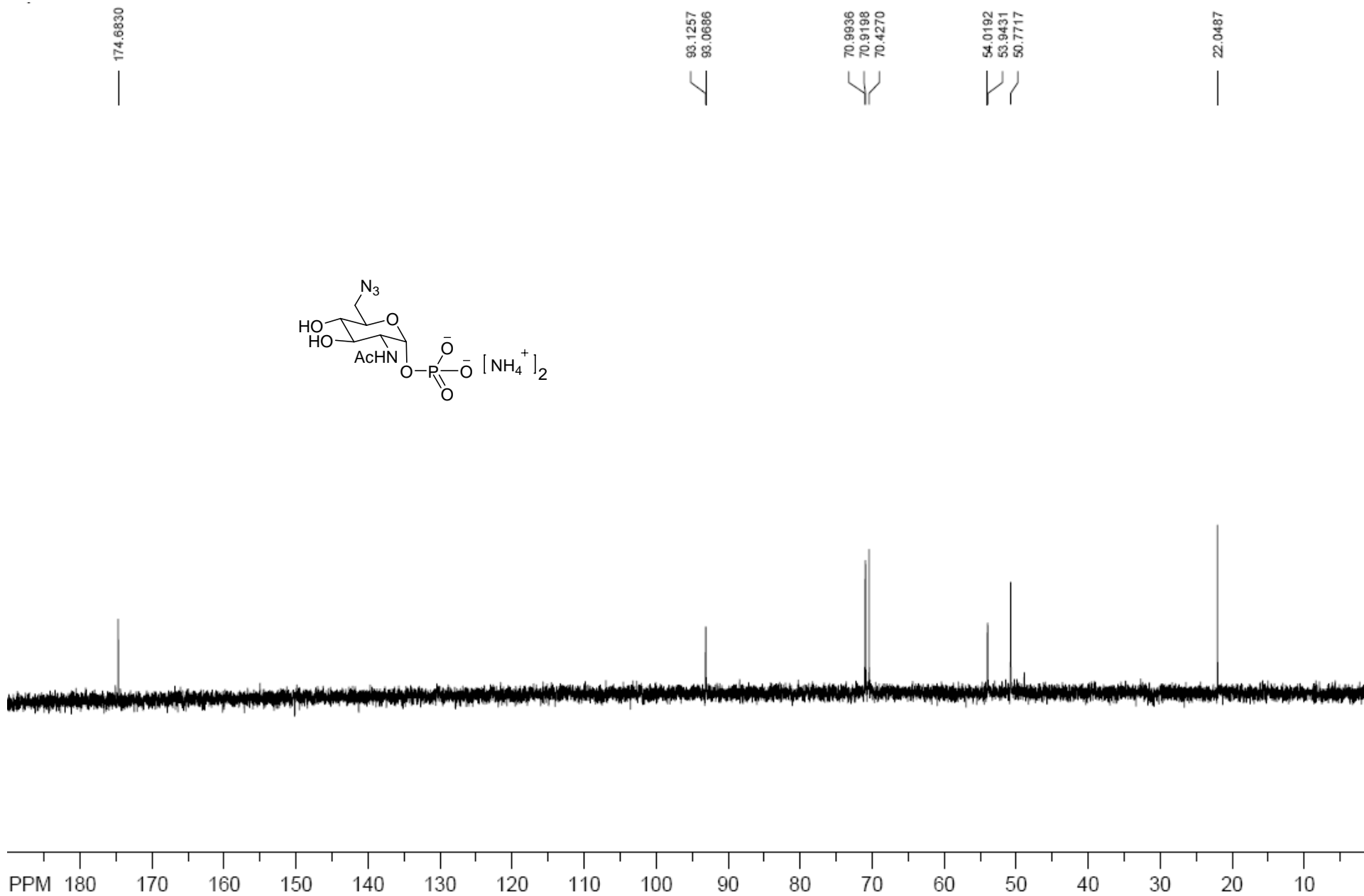
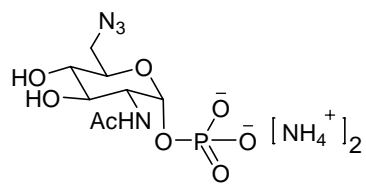
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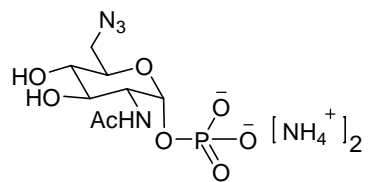
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S5

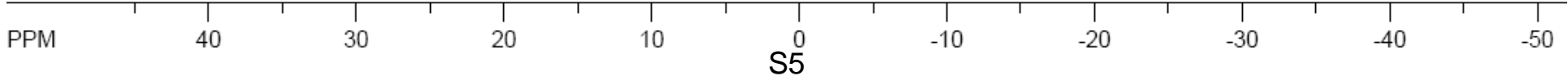


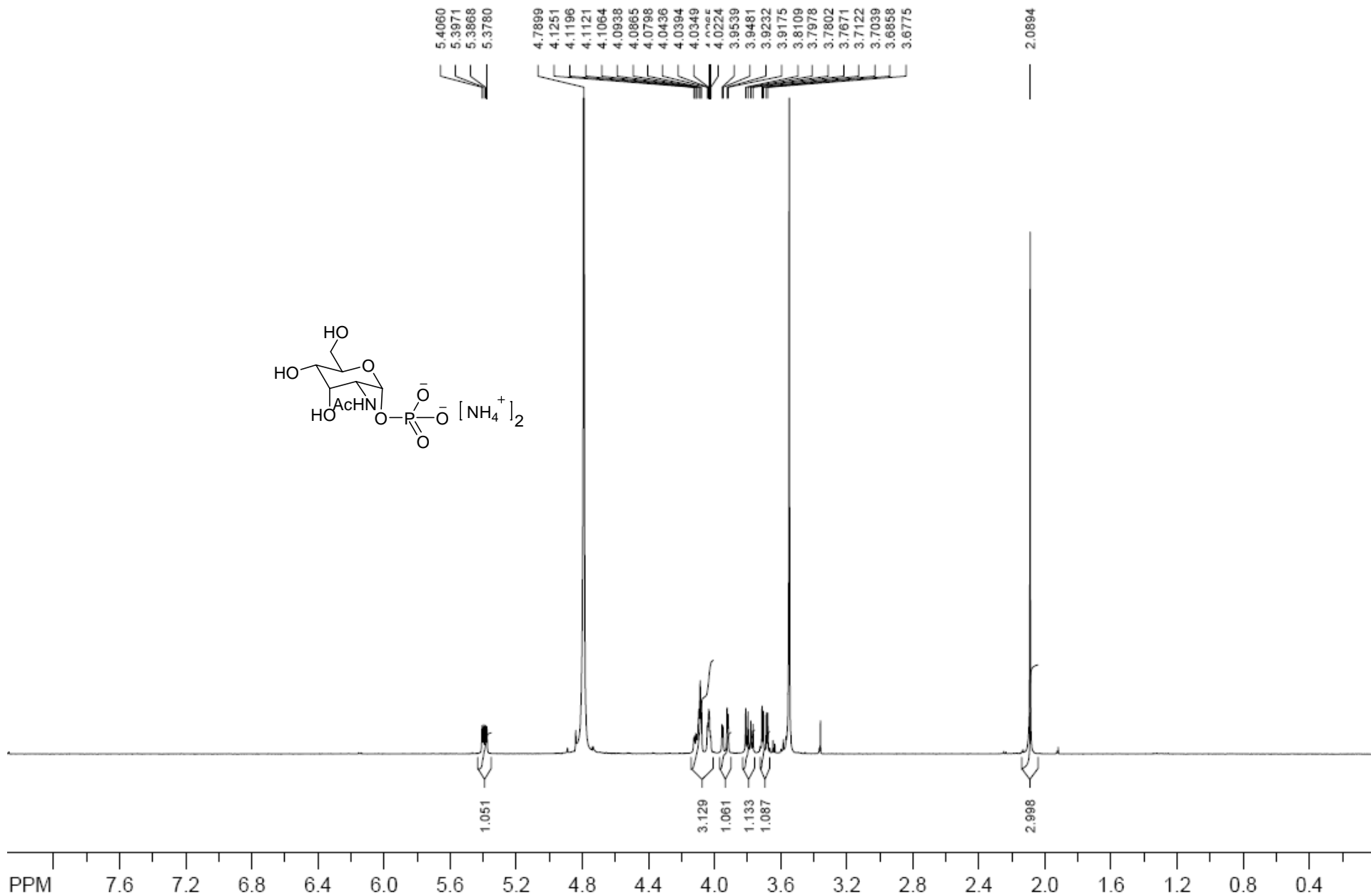


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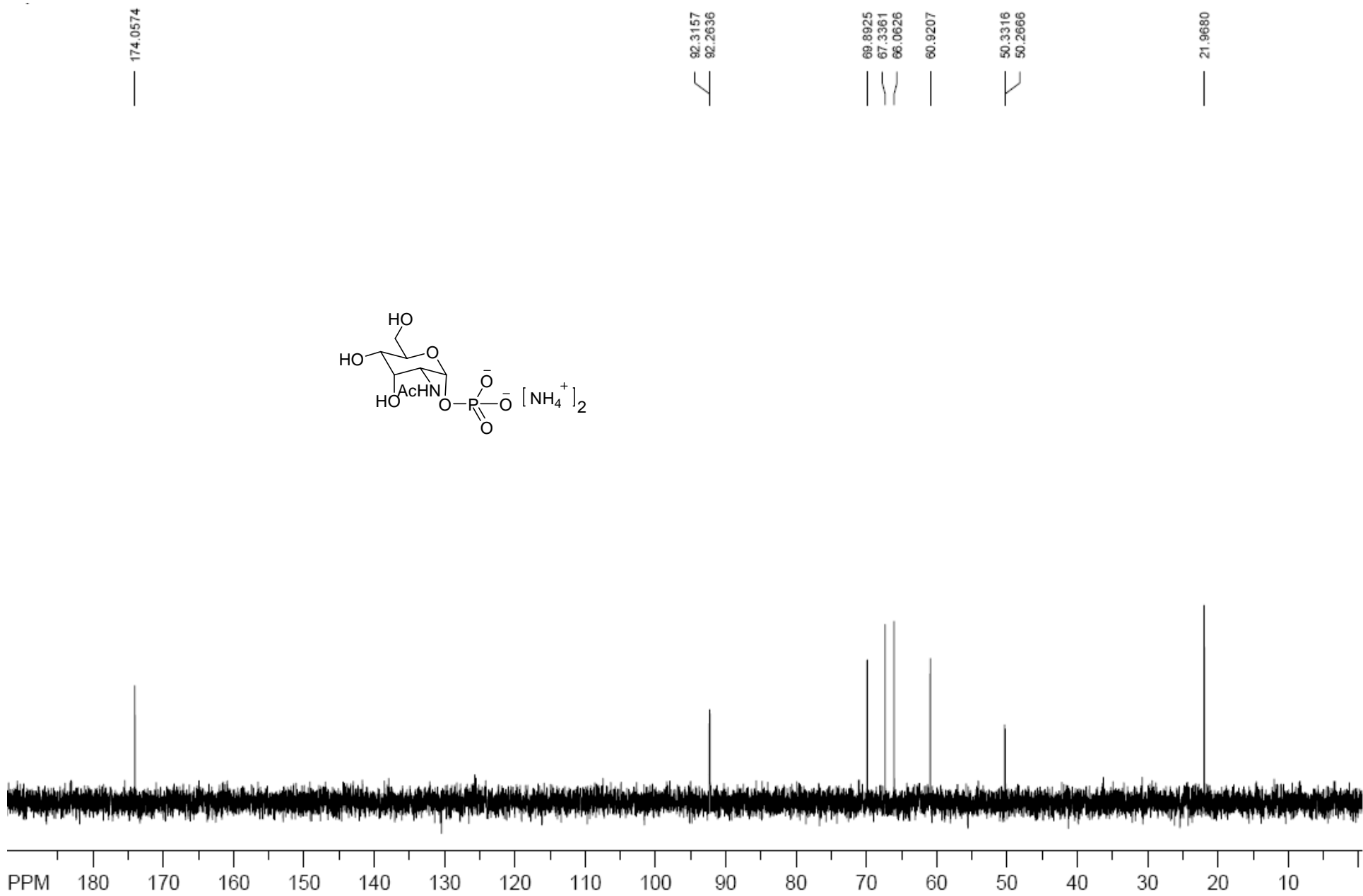


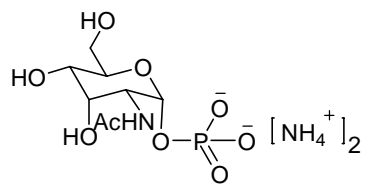
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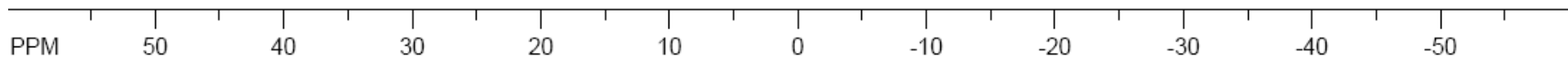


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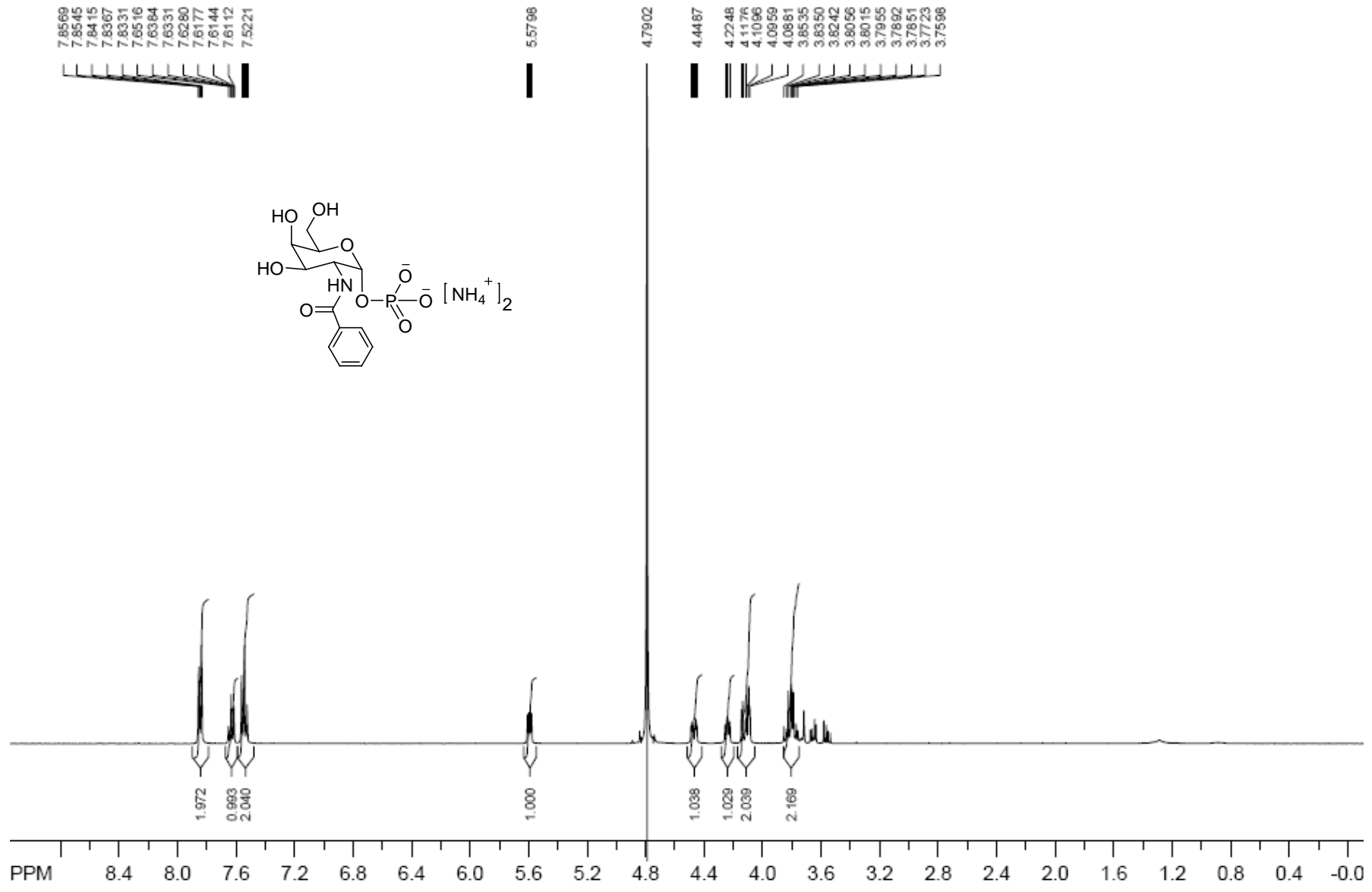


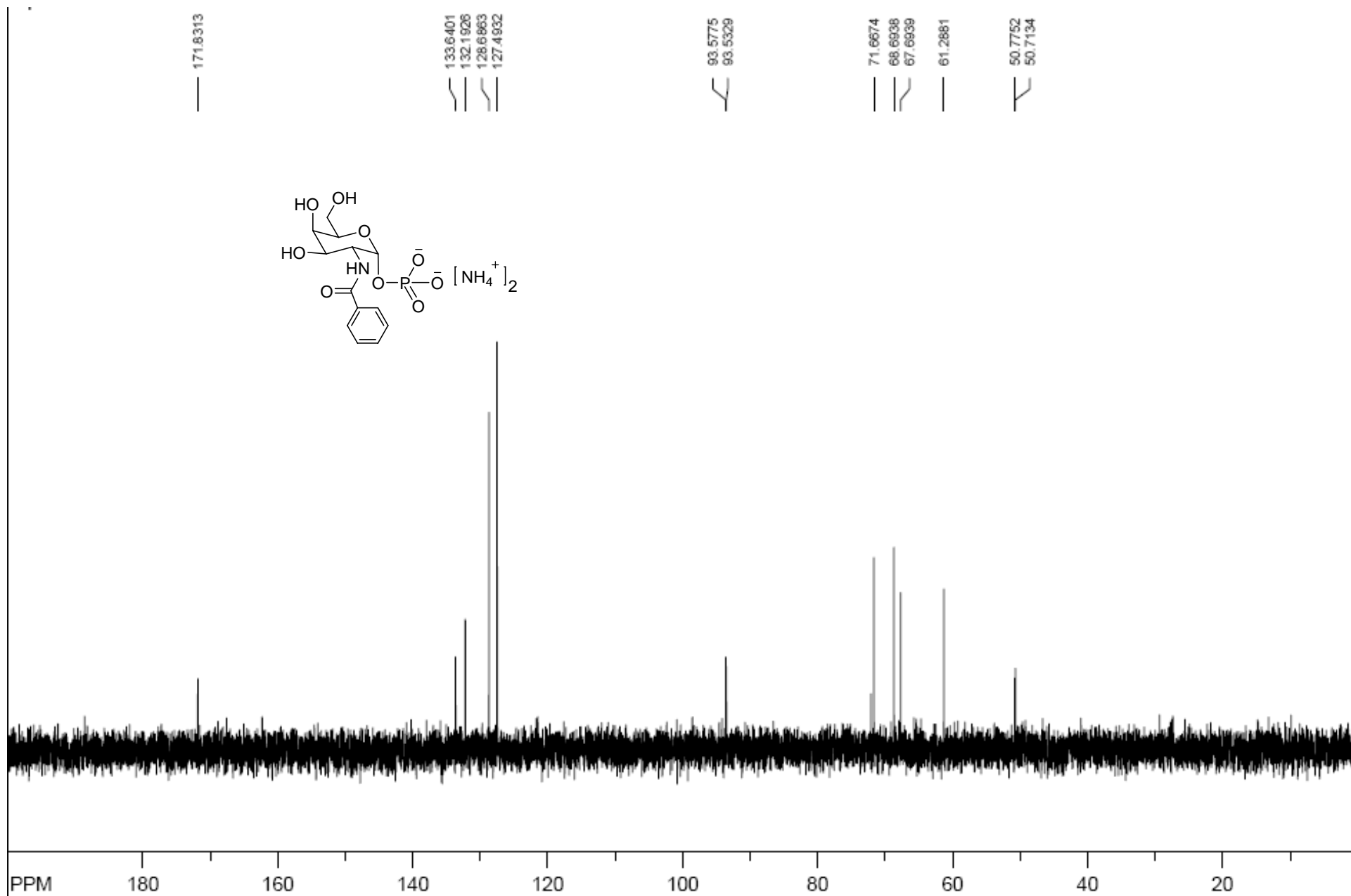


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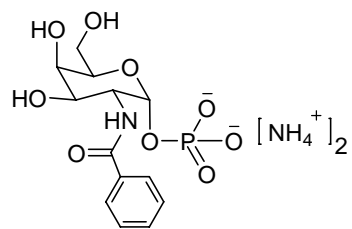


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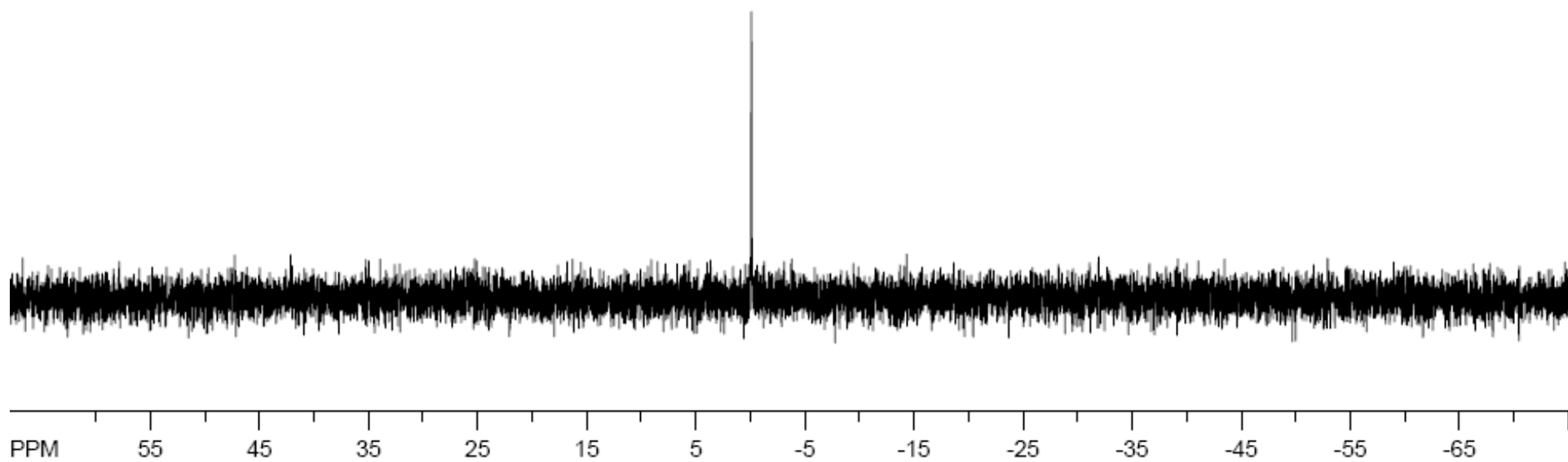




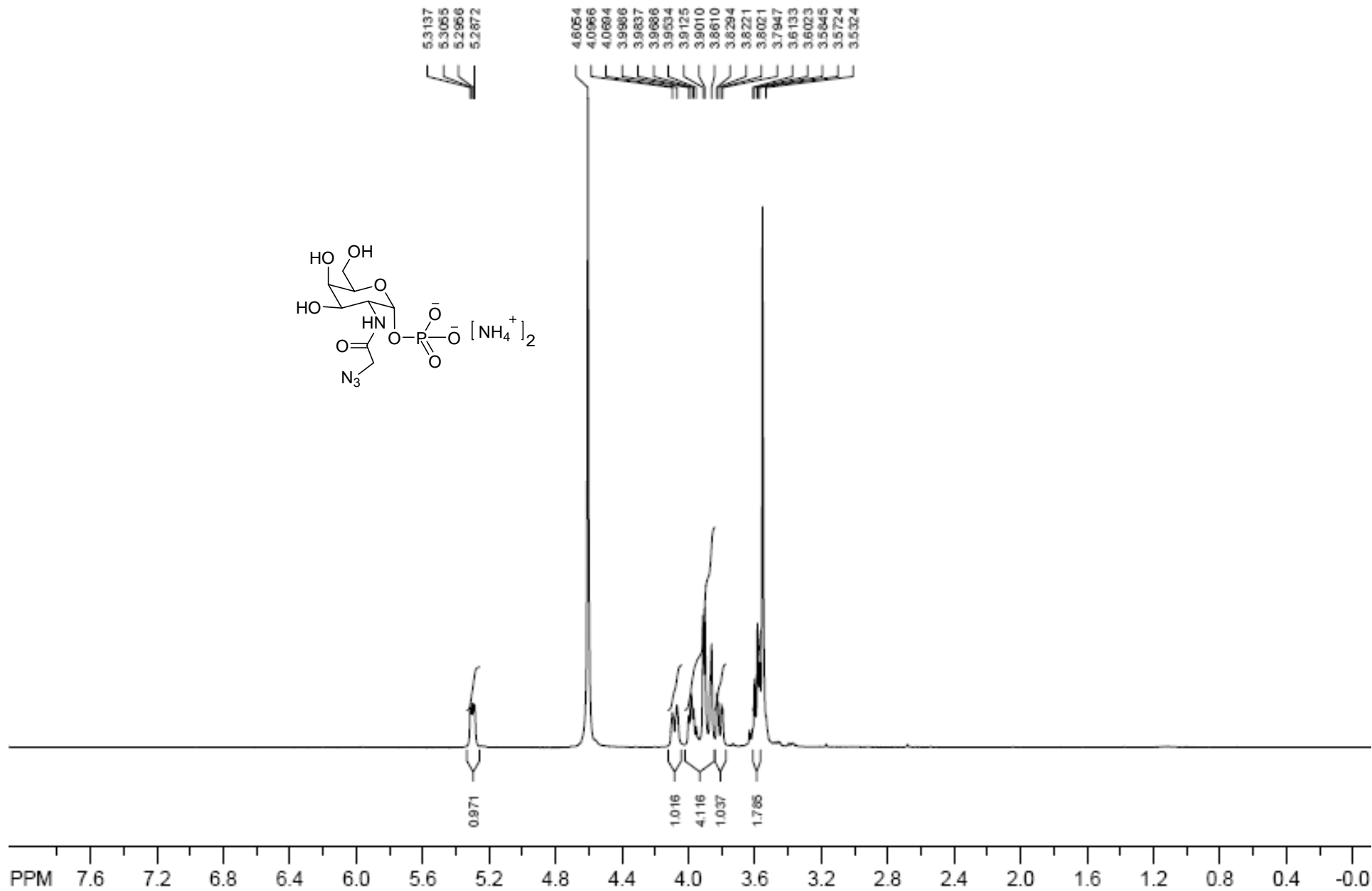
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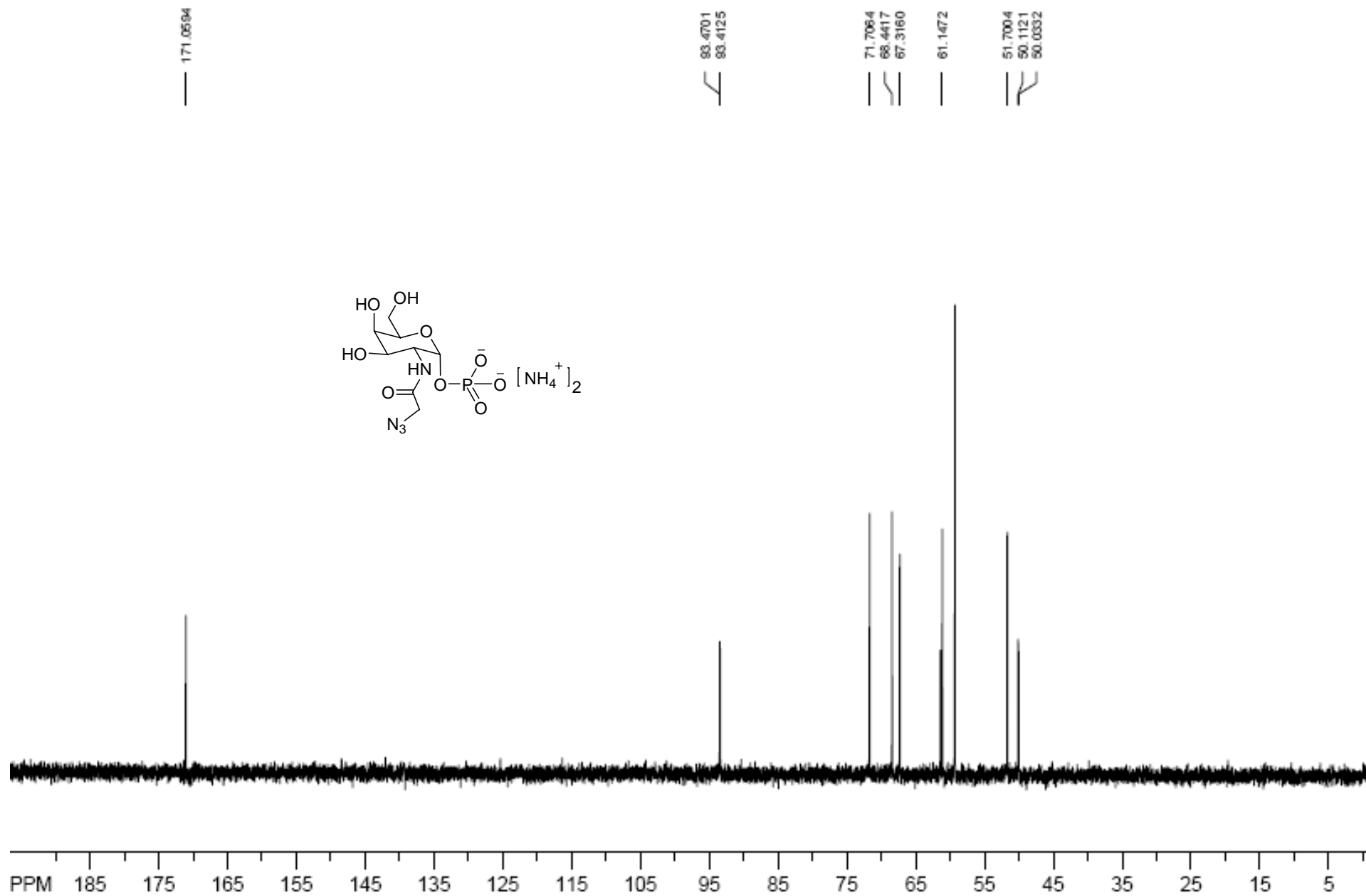
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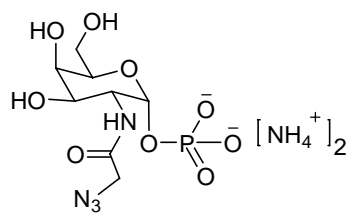
S5



S5



S5

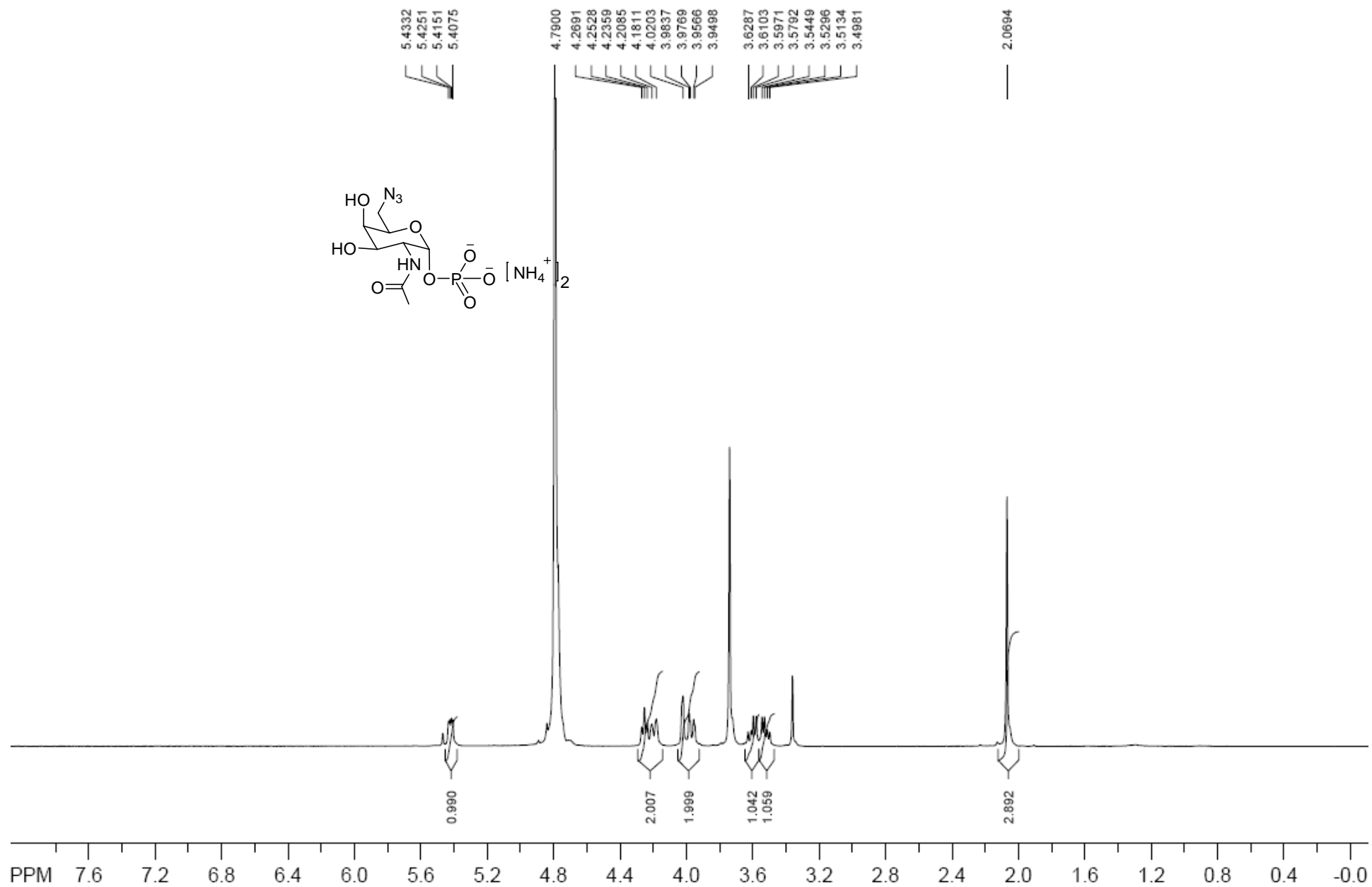


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S5



S5

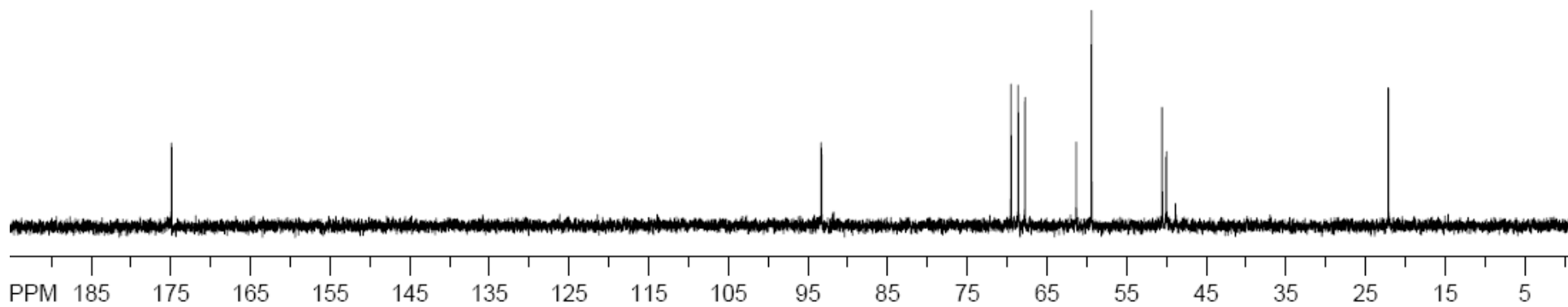
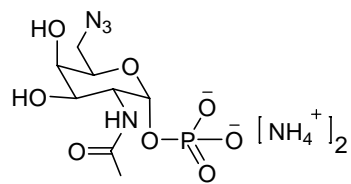
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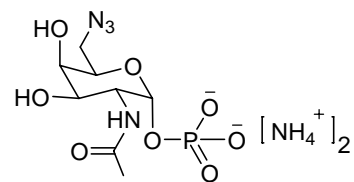
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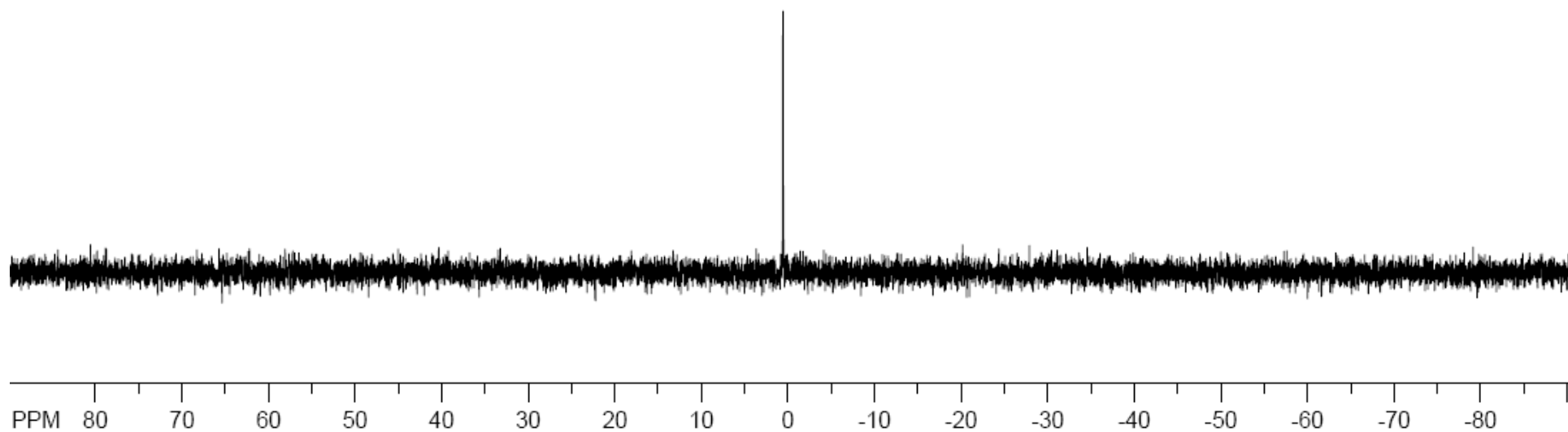
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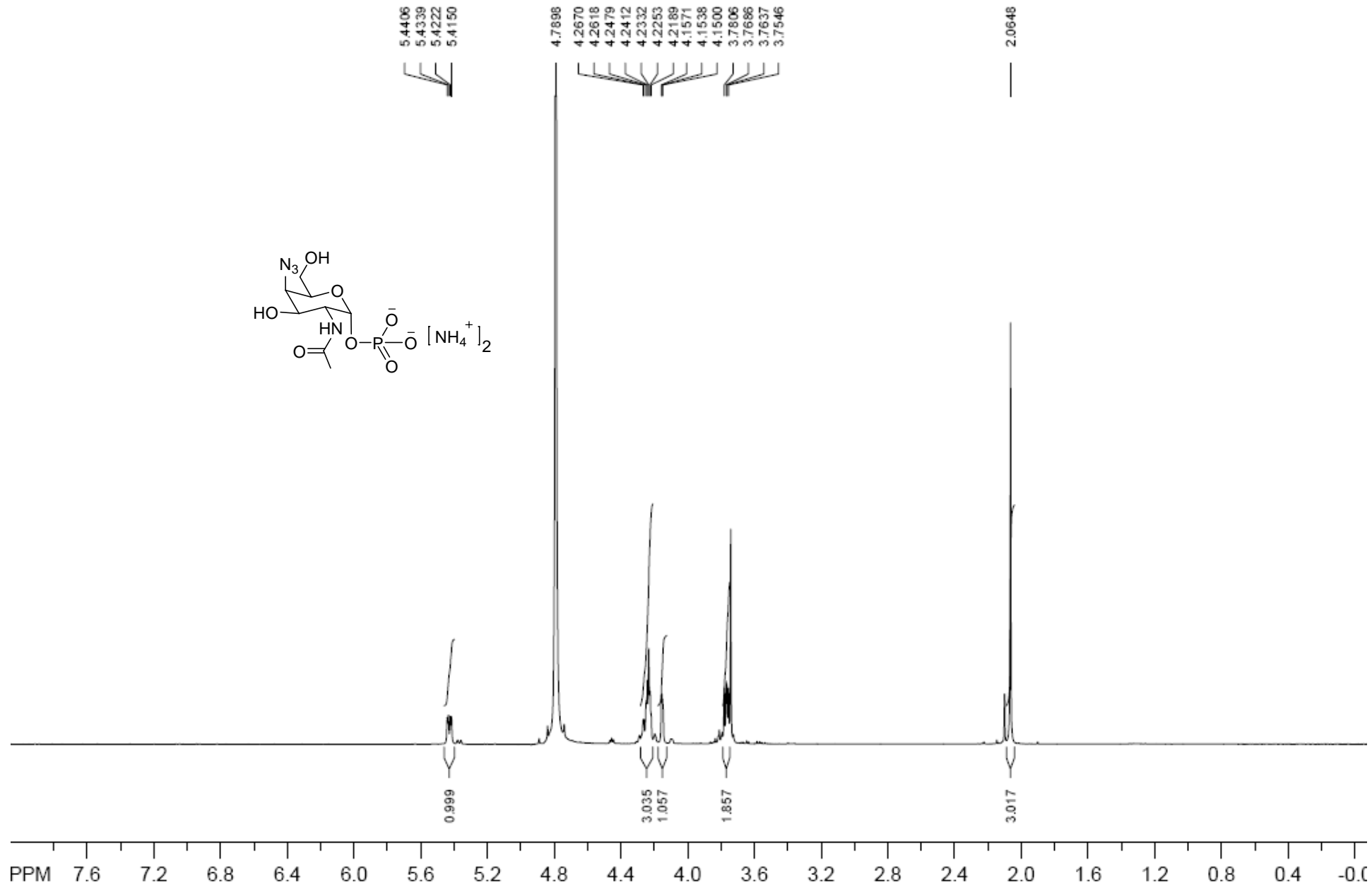


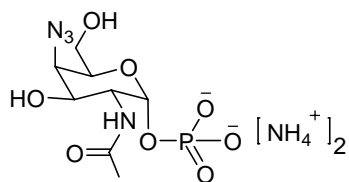
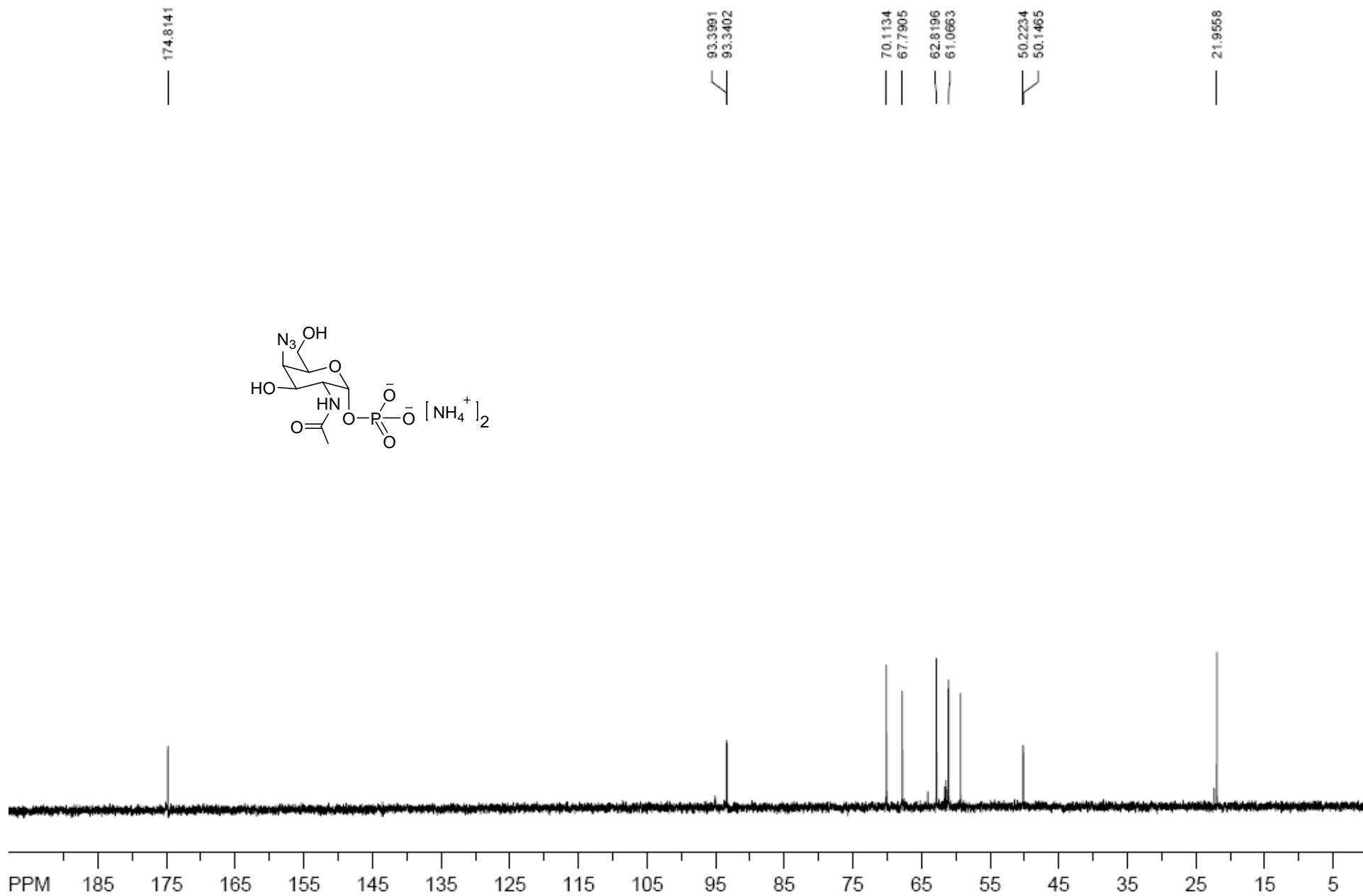


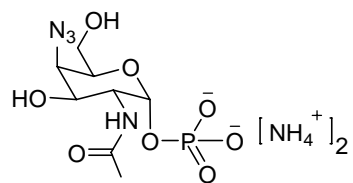
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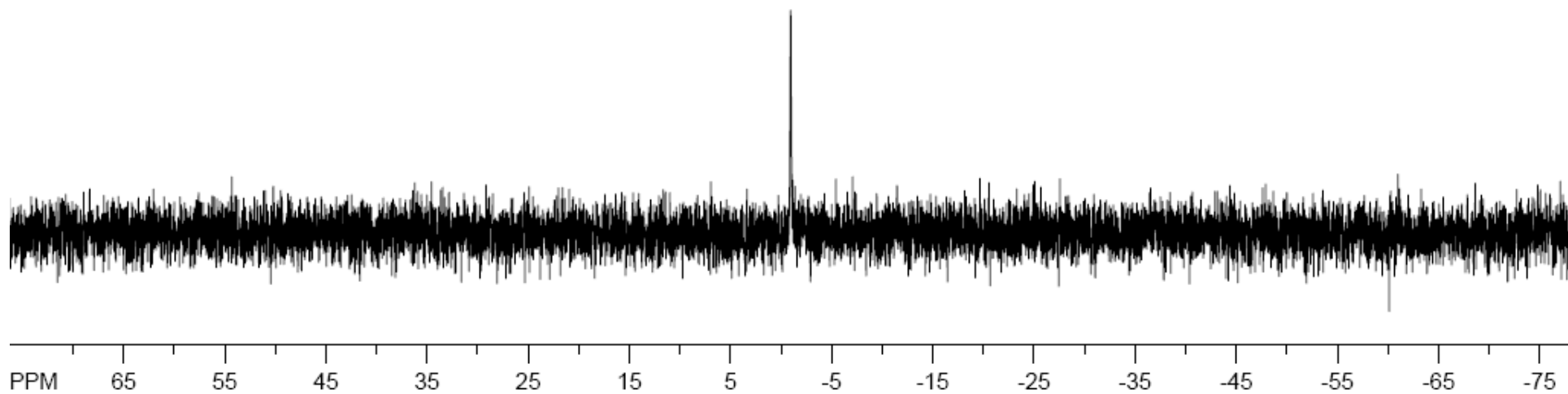
S5



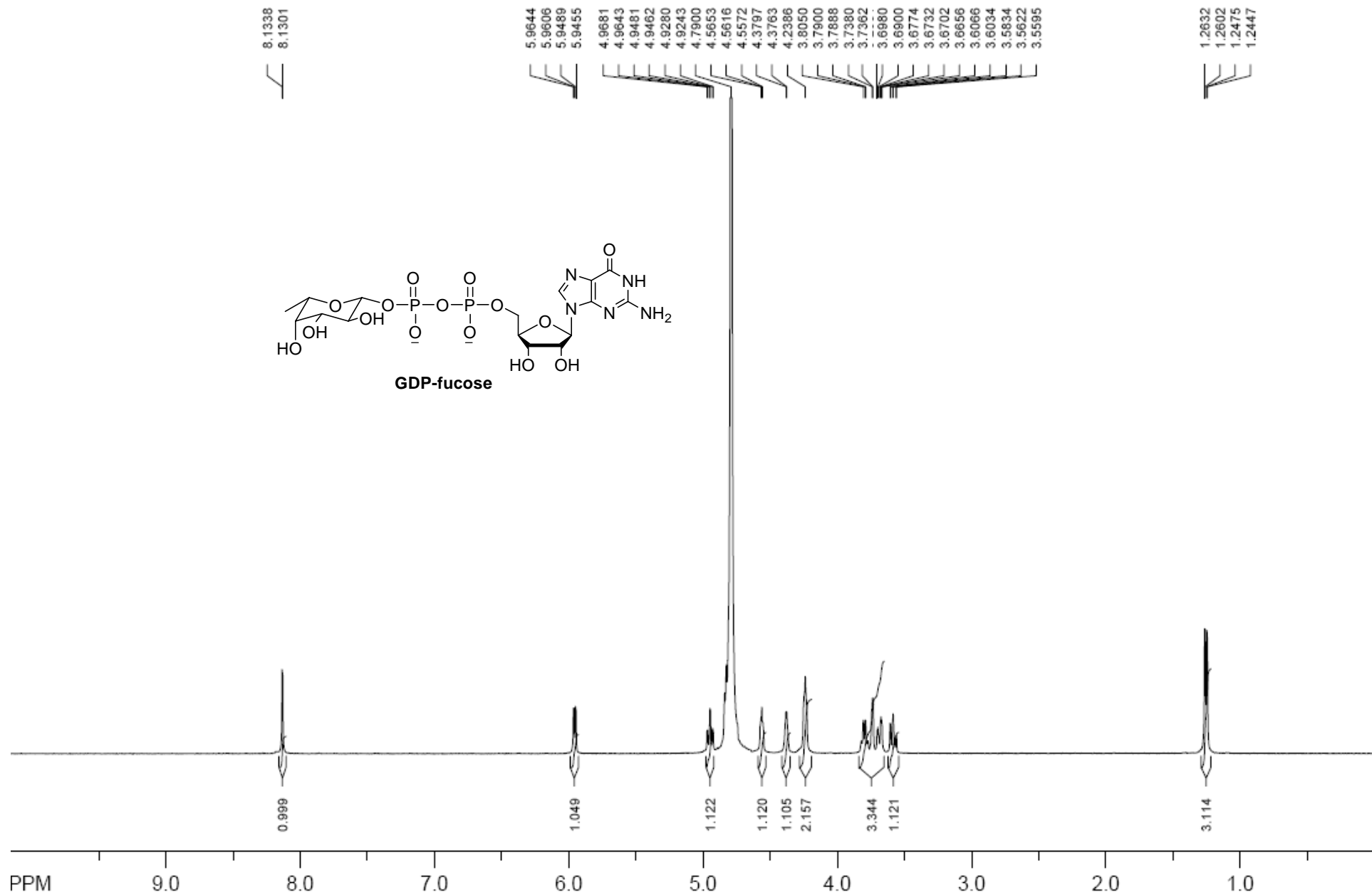
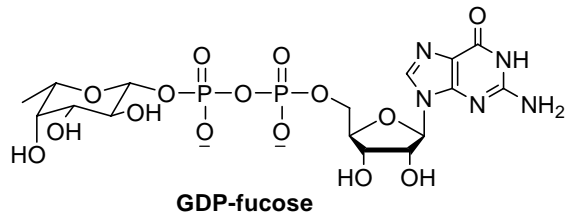


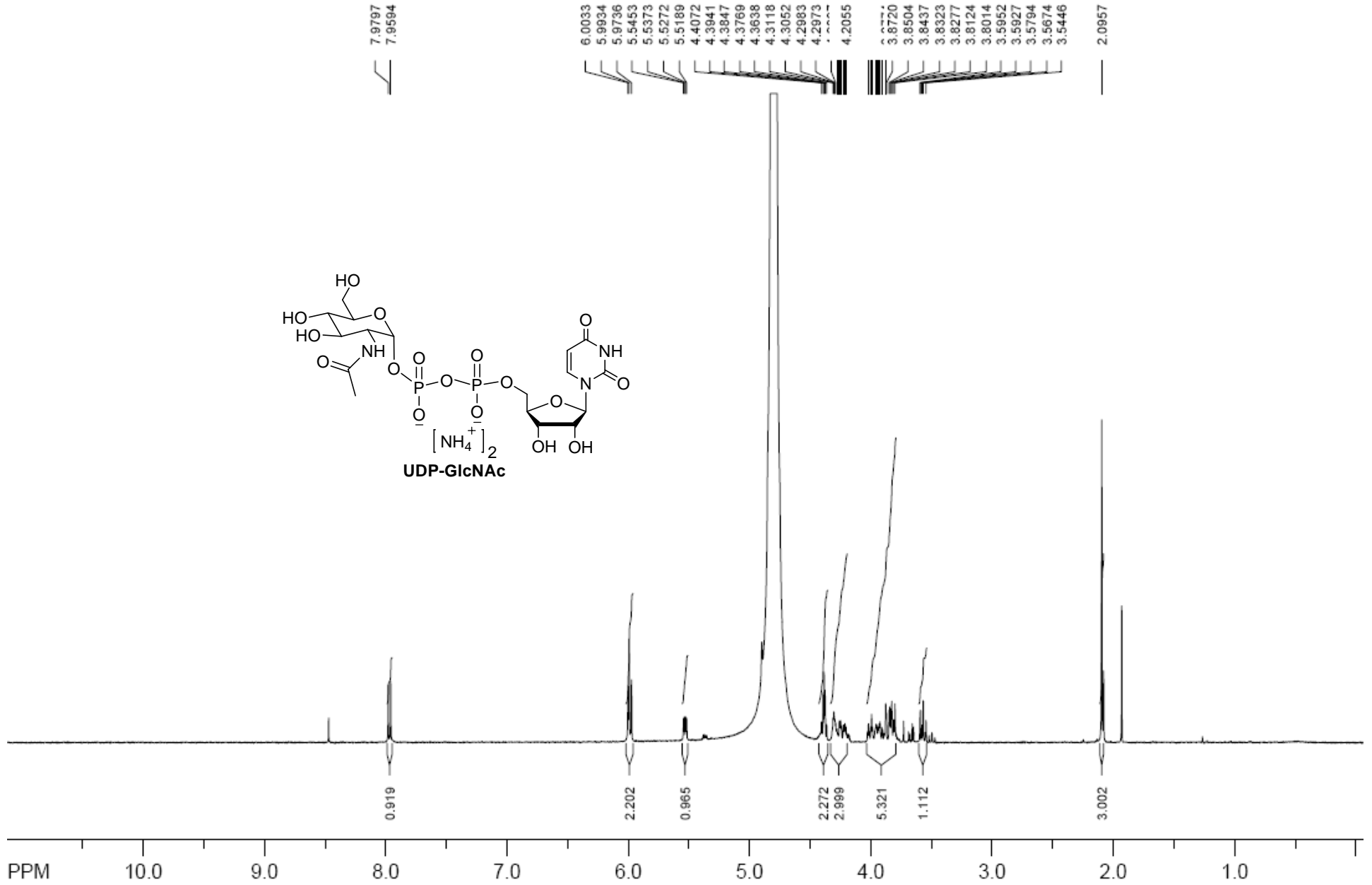


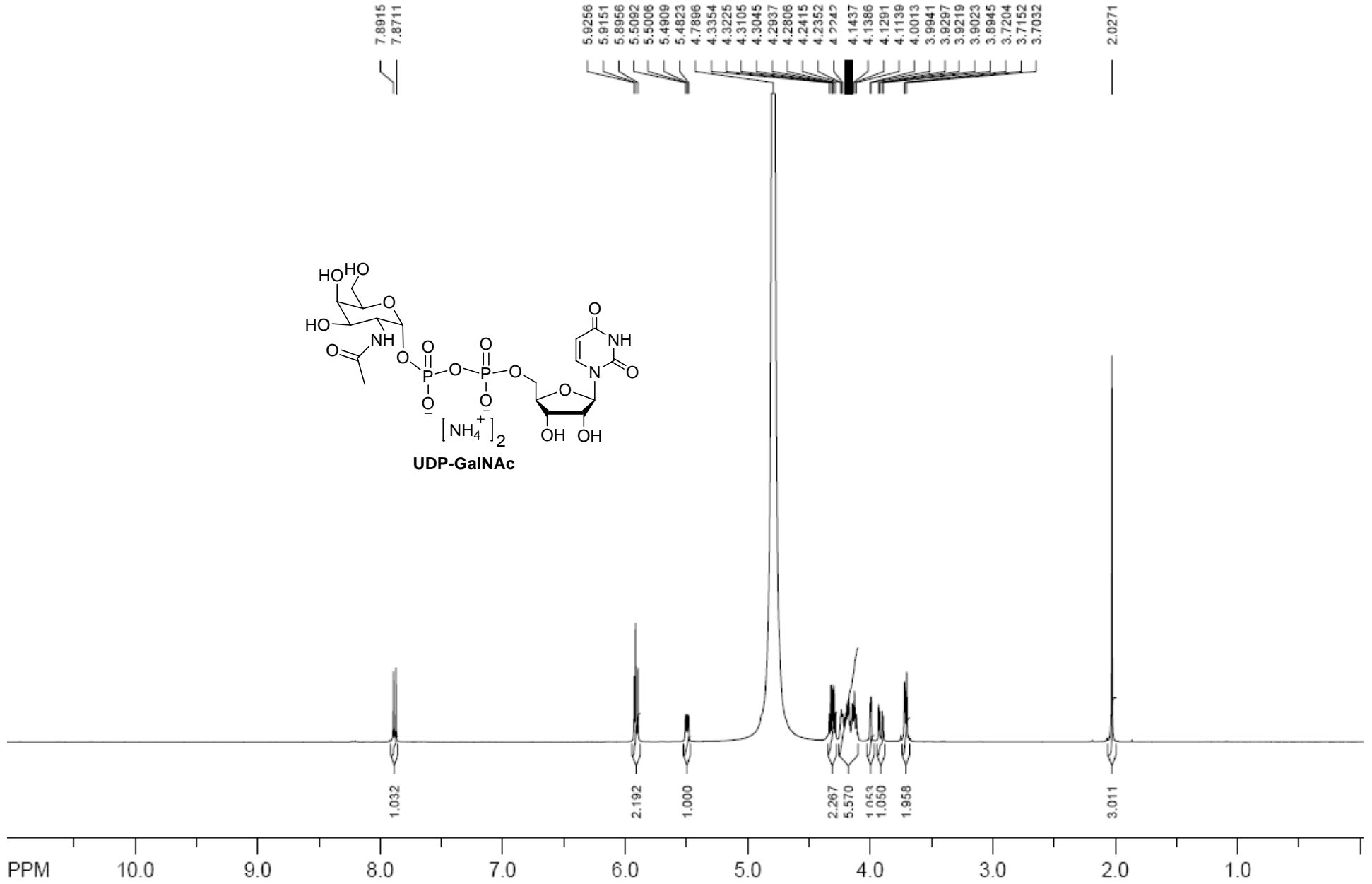
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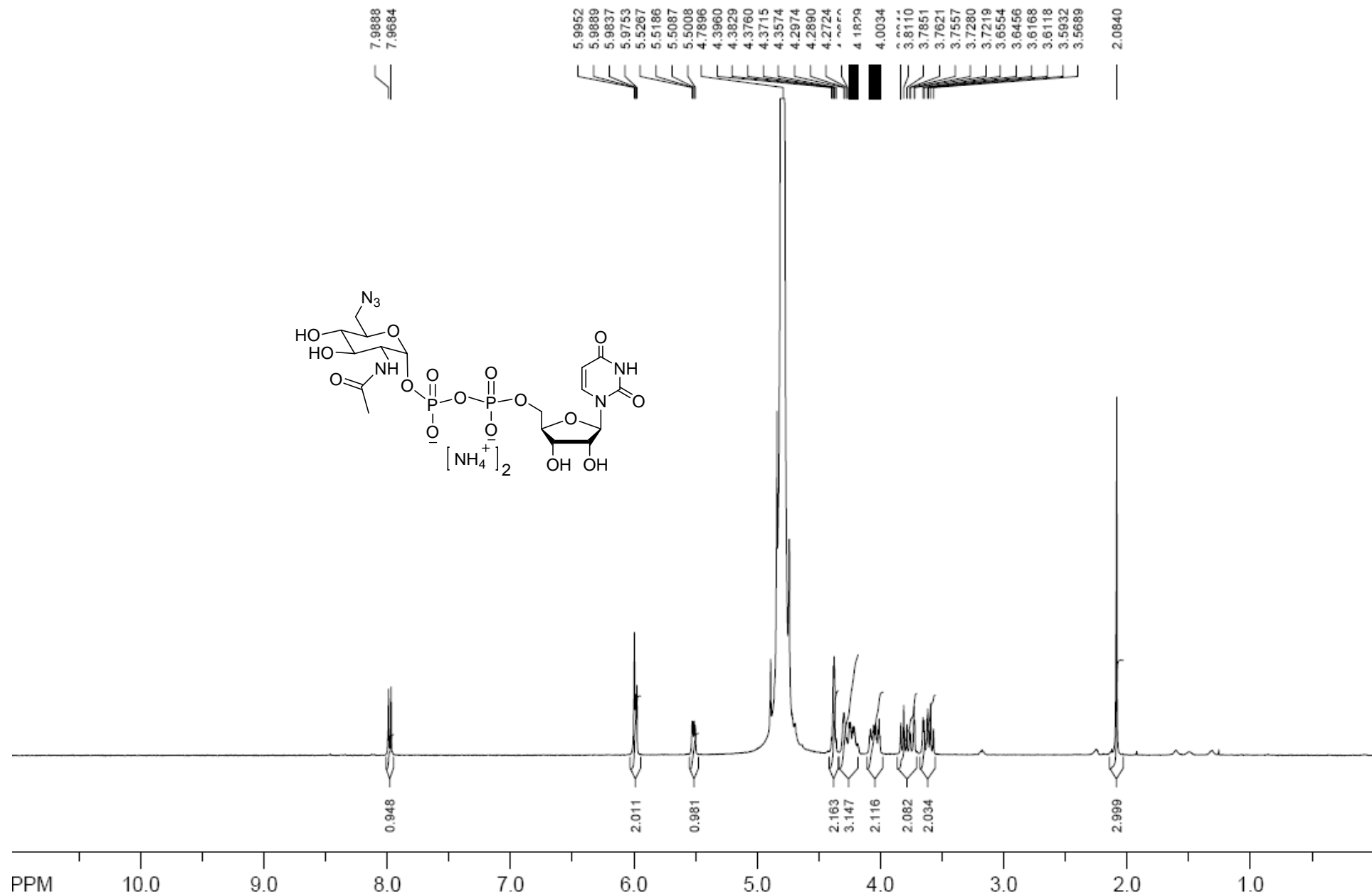


S5

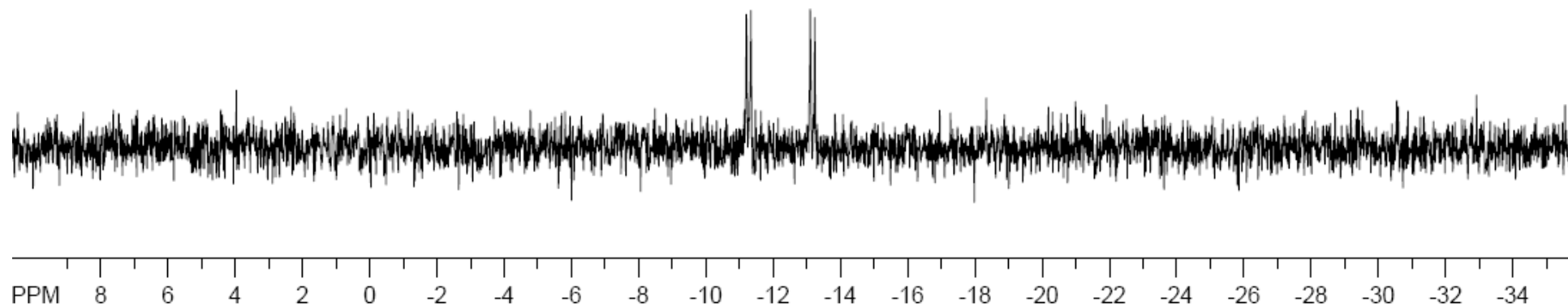
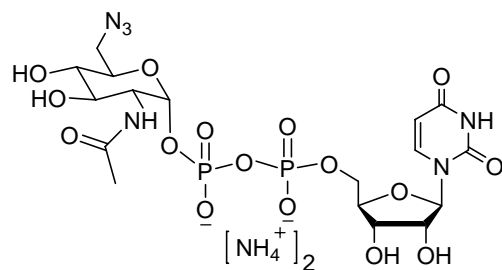




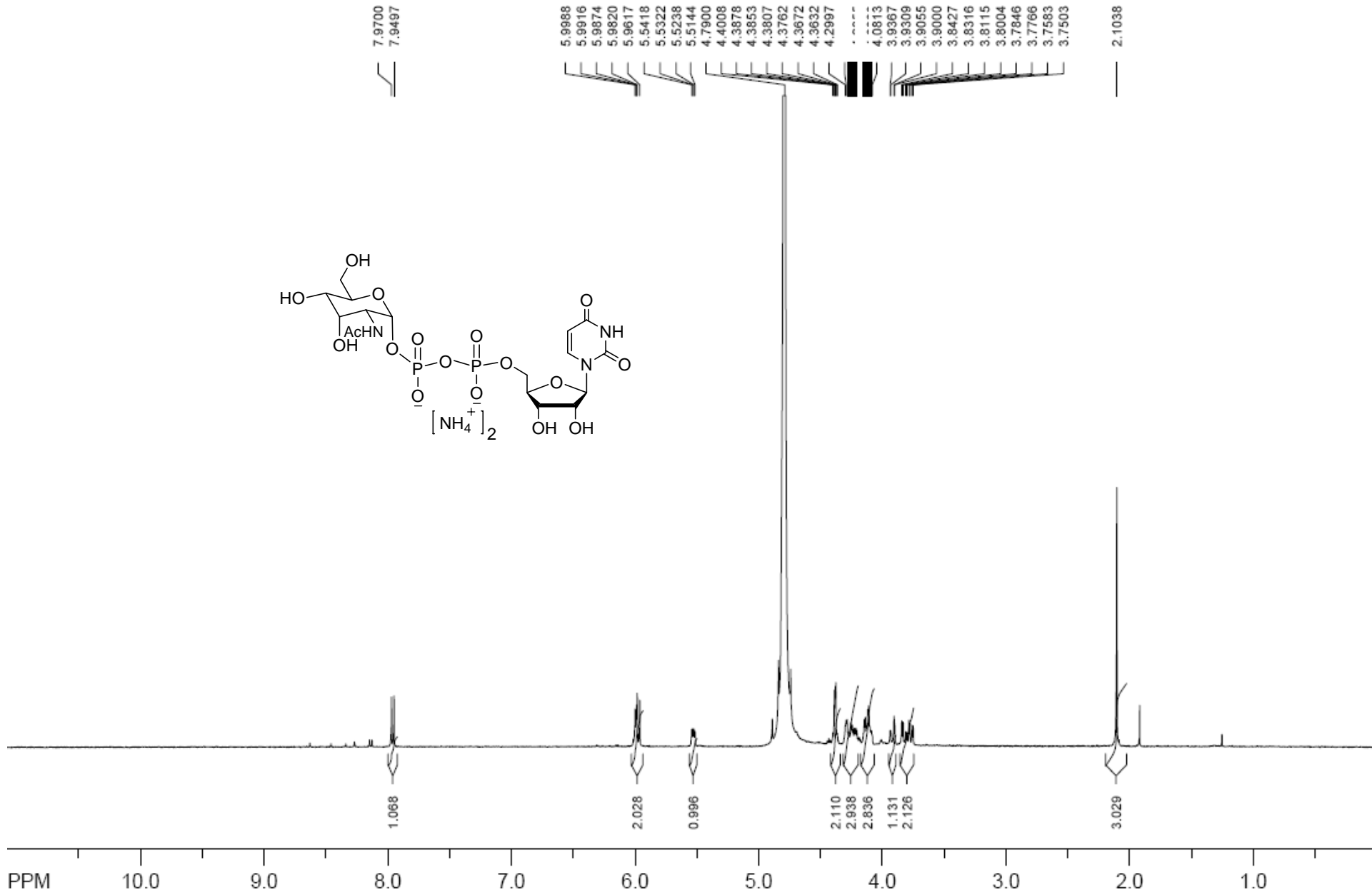


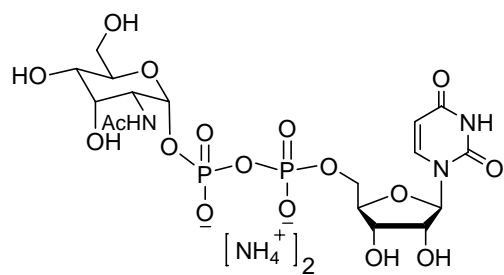


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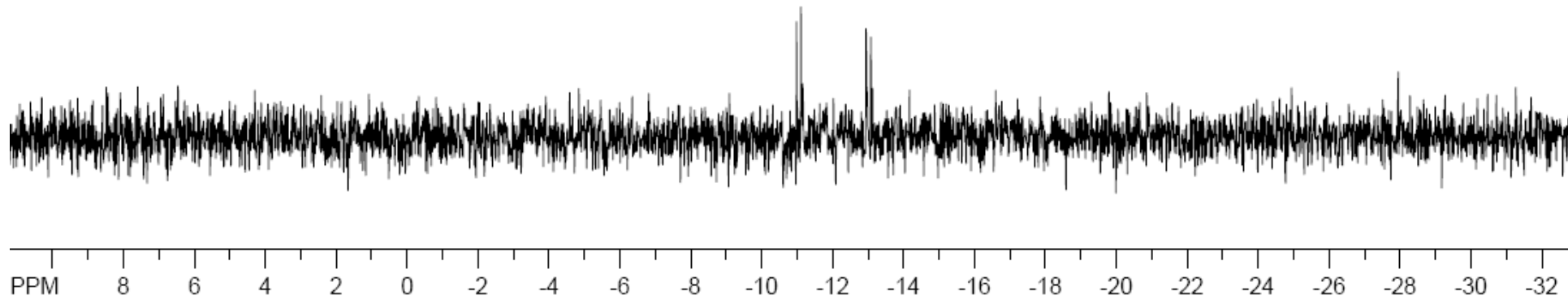
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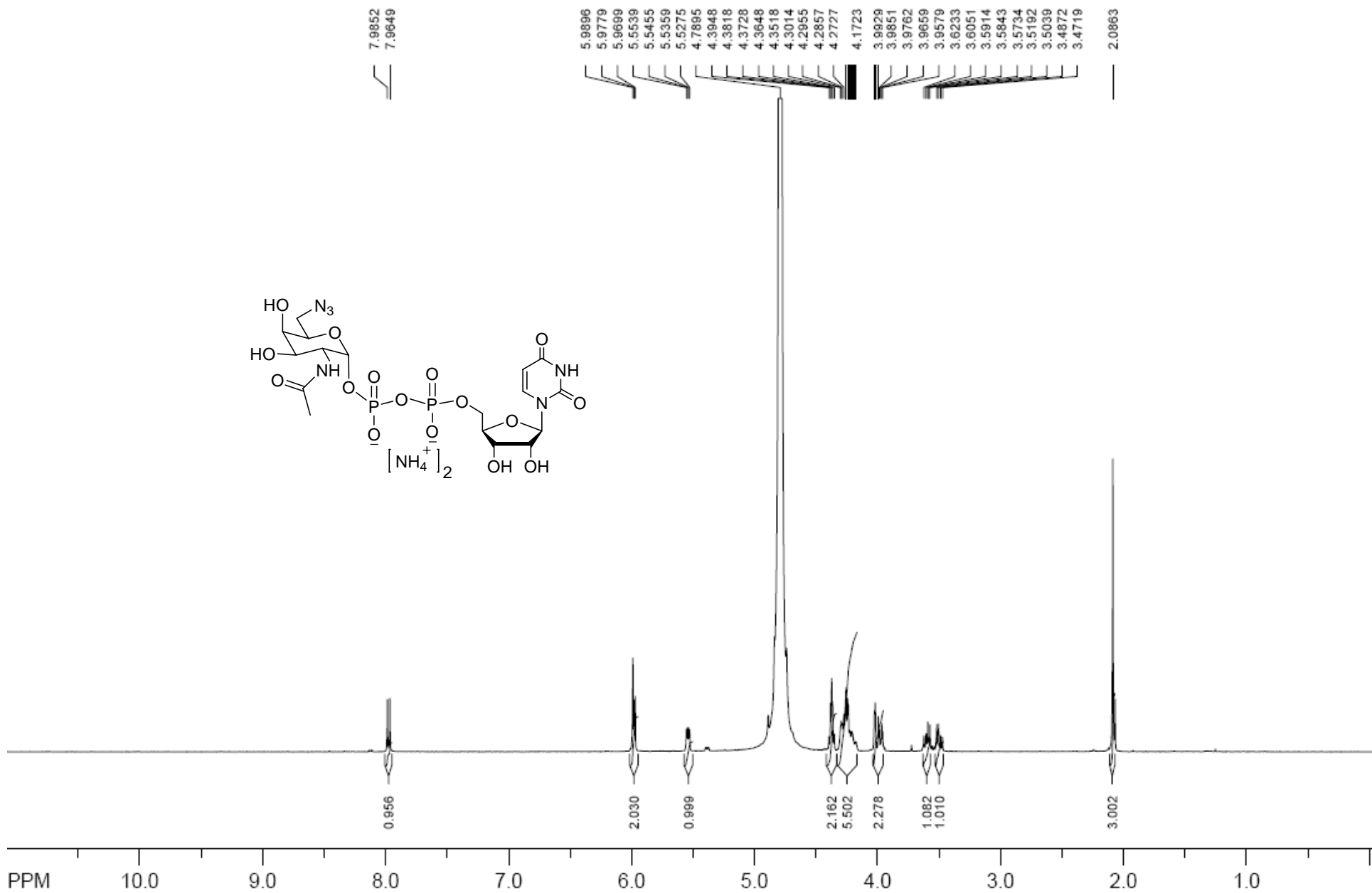


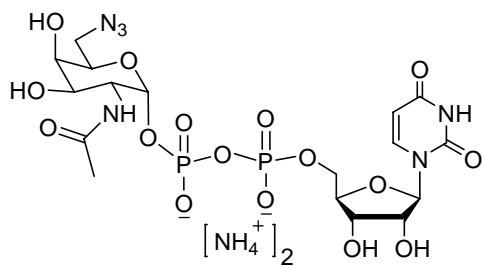
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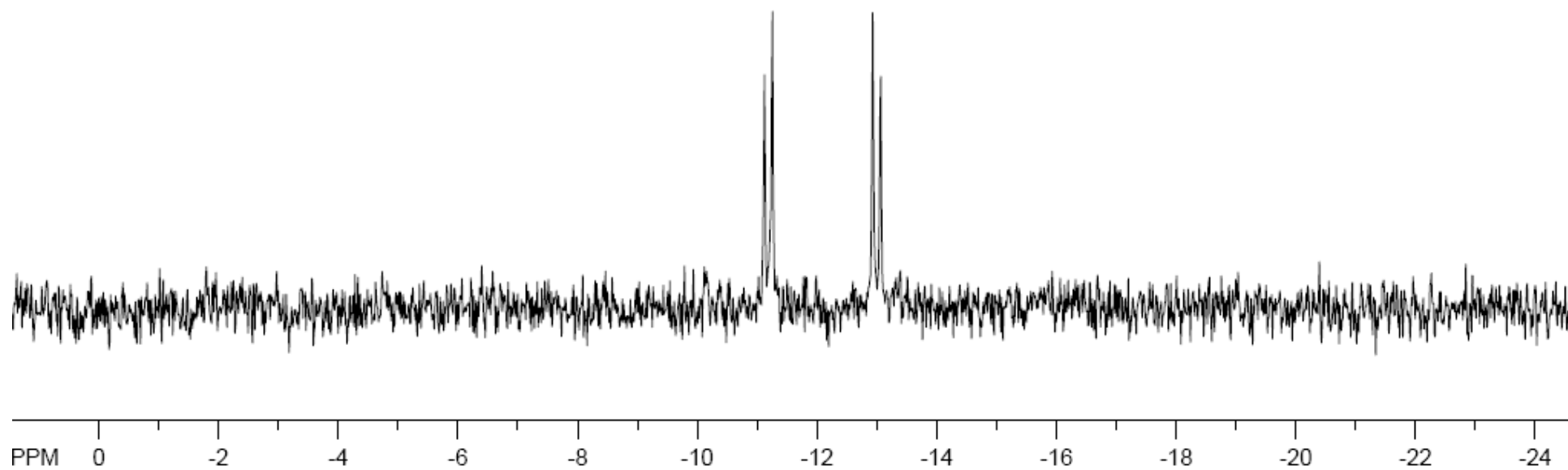
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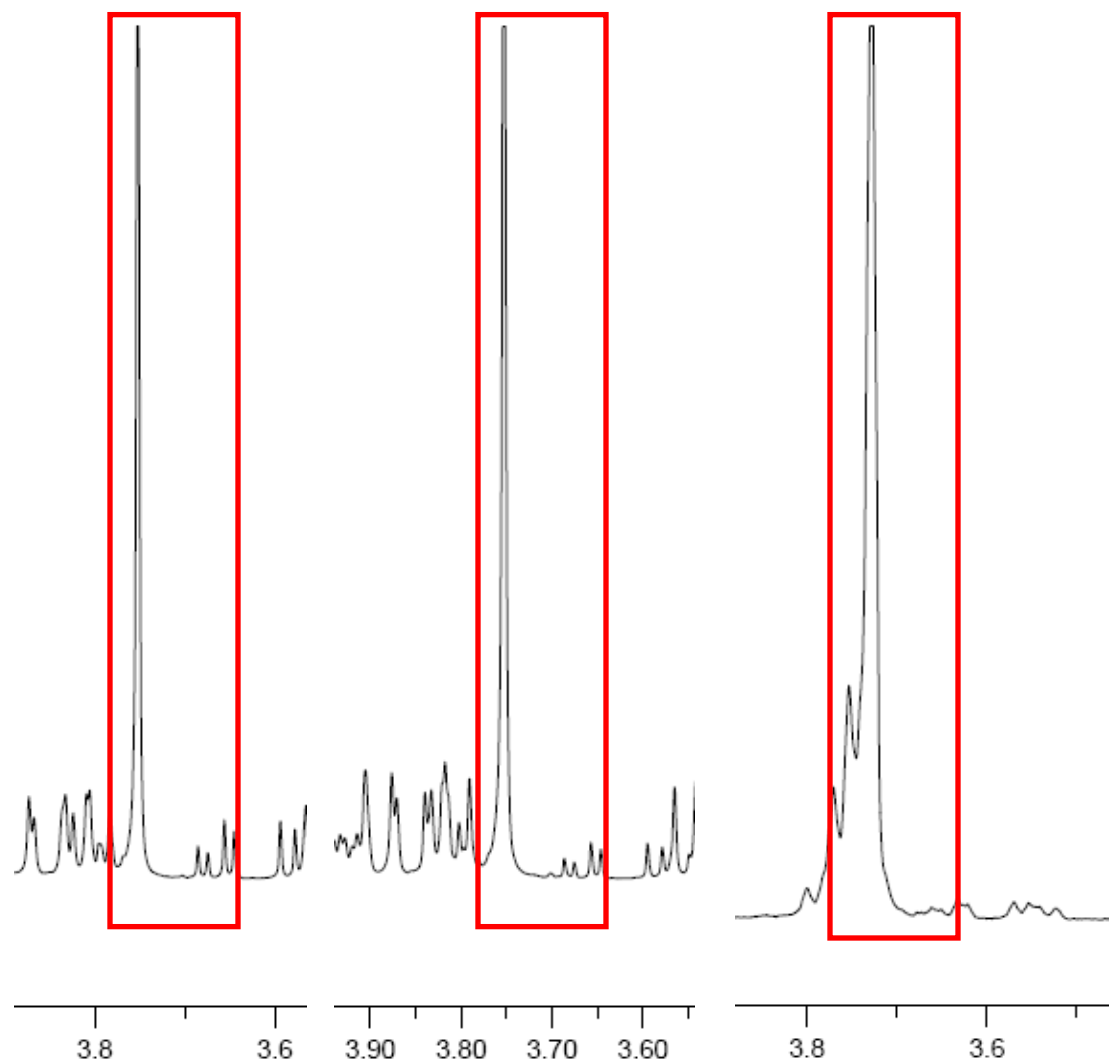


-11.1203
-11.2519

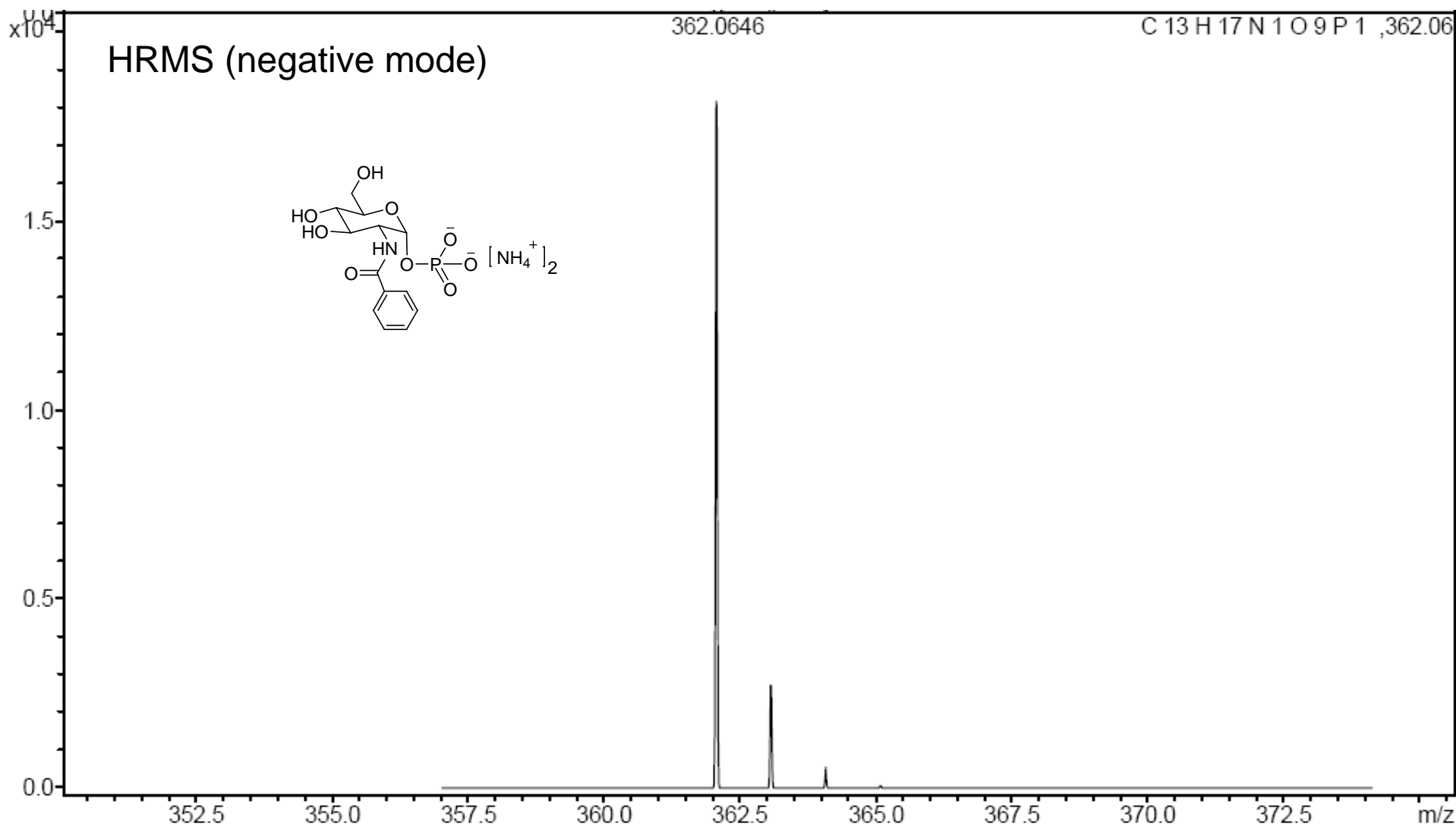
-12.9302
-13.0615



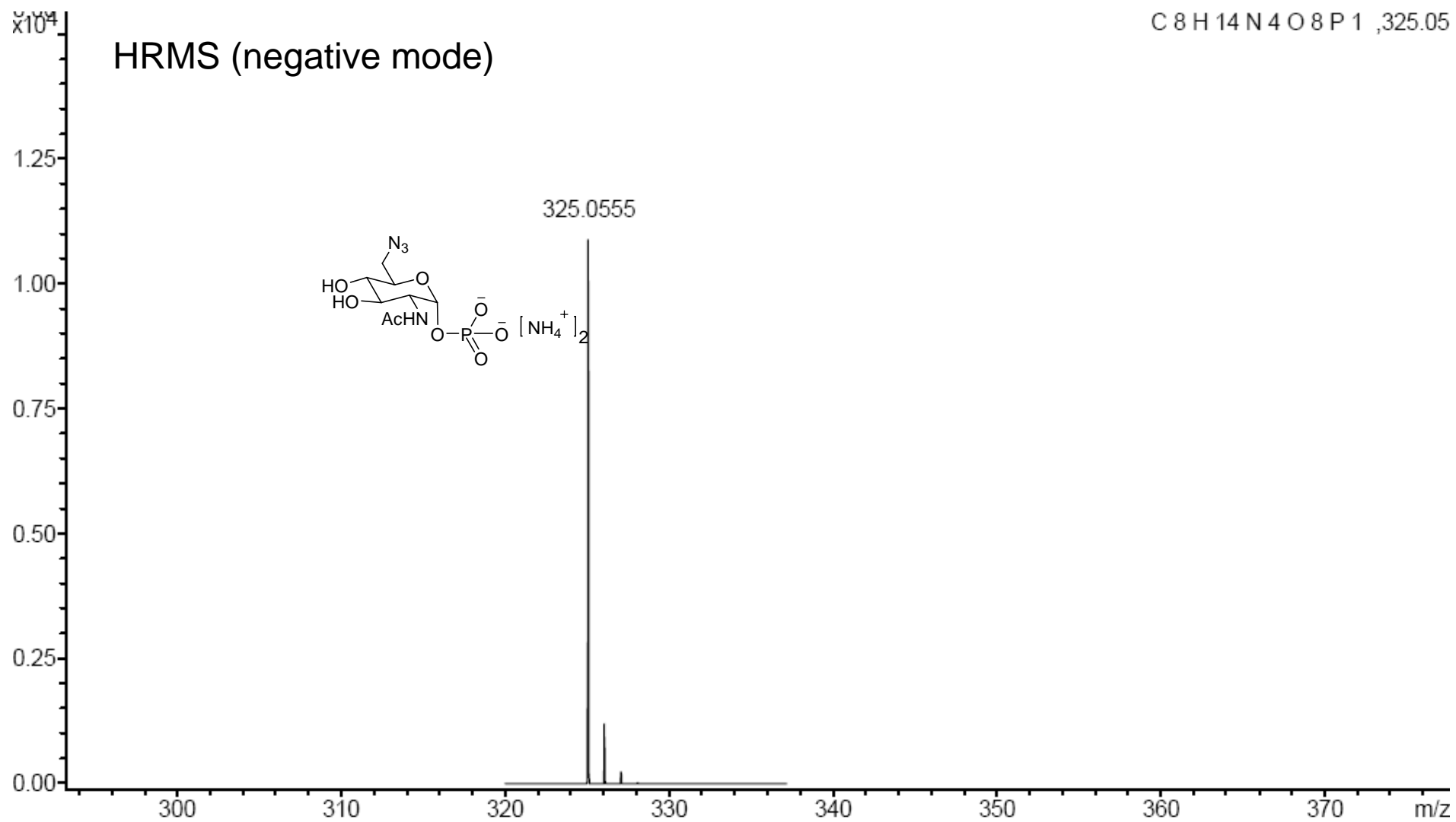
S5



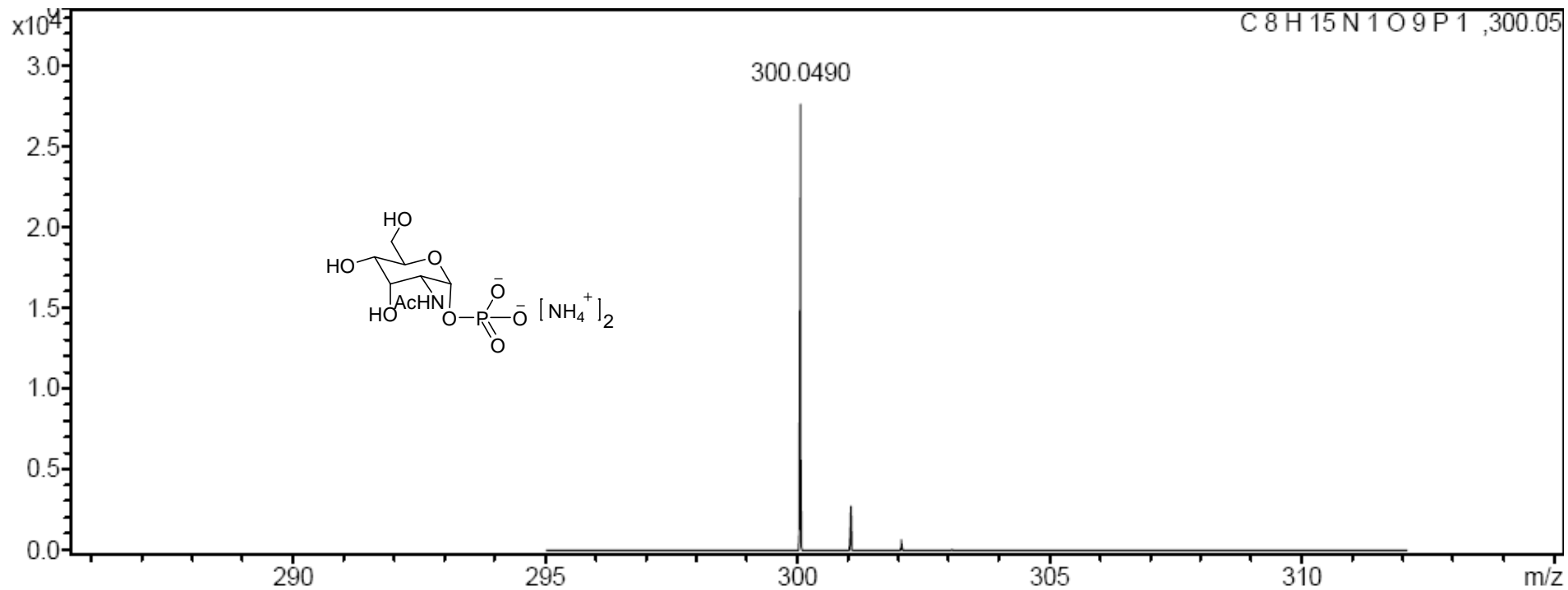
Note: NMR regions (red box) showing typical common impurities after the first time silica gel purification of sugar-1-P, second silica gel chromatography is needed to remove the impurities for NMR purposes.

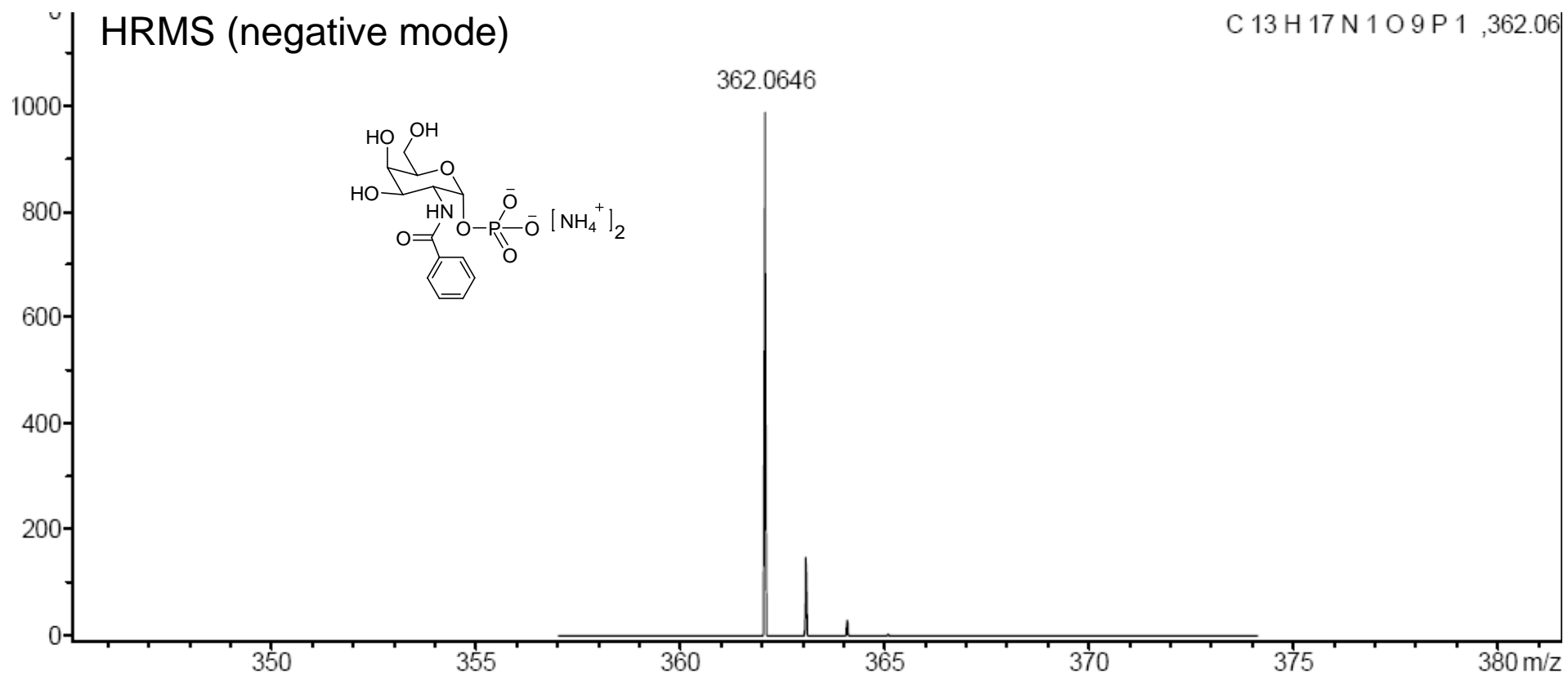


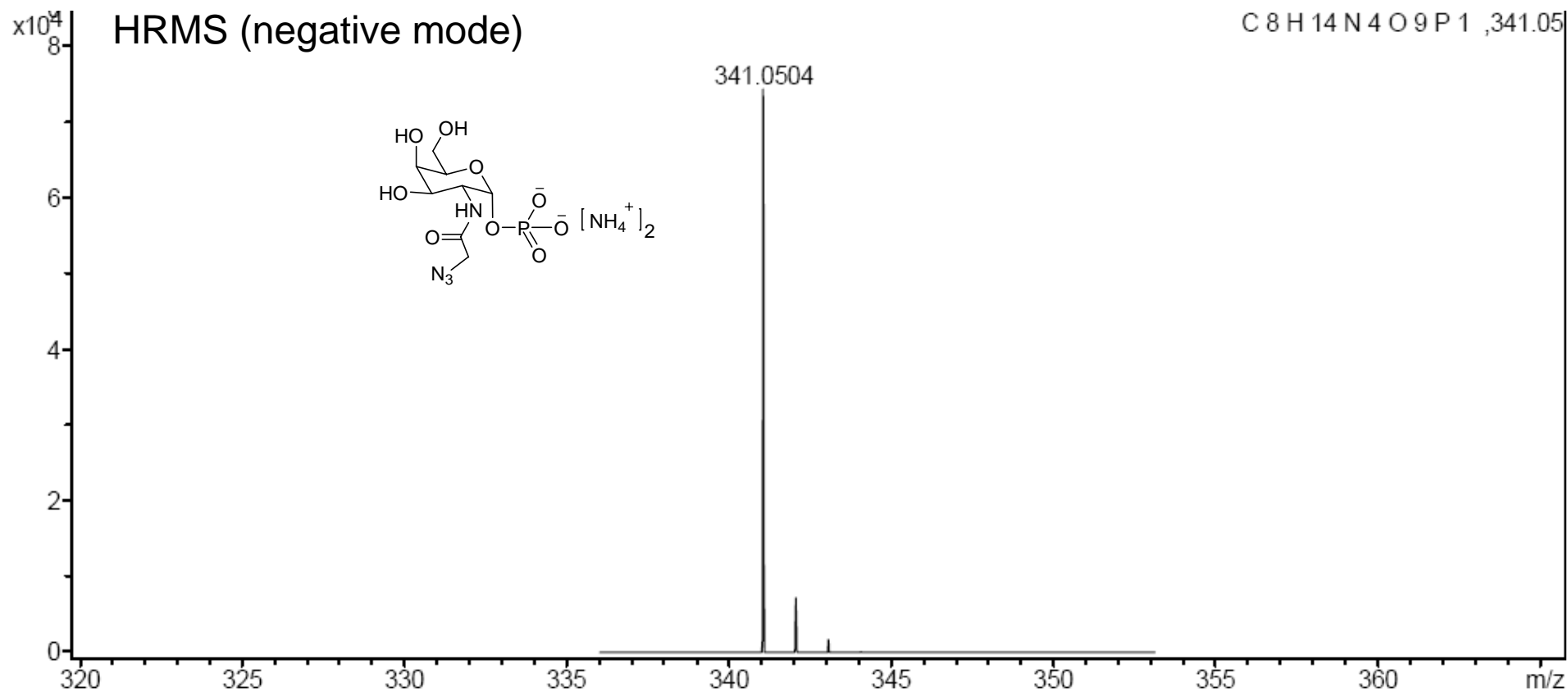
C₈H₁₄N₄O₈P₁, 325.05

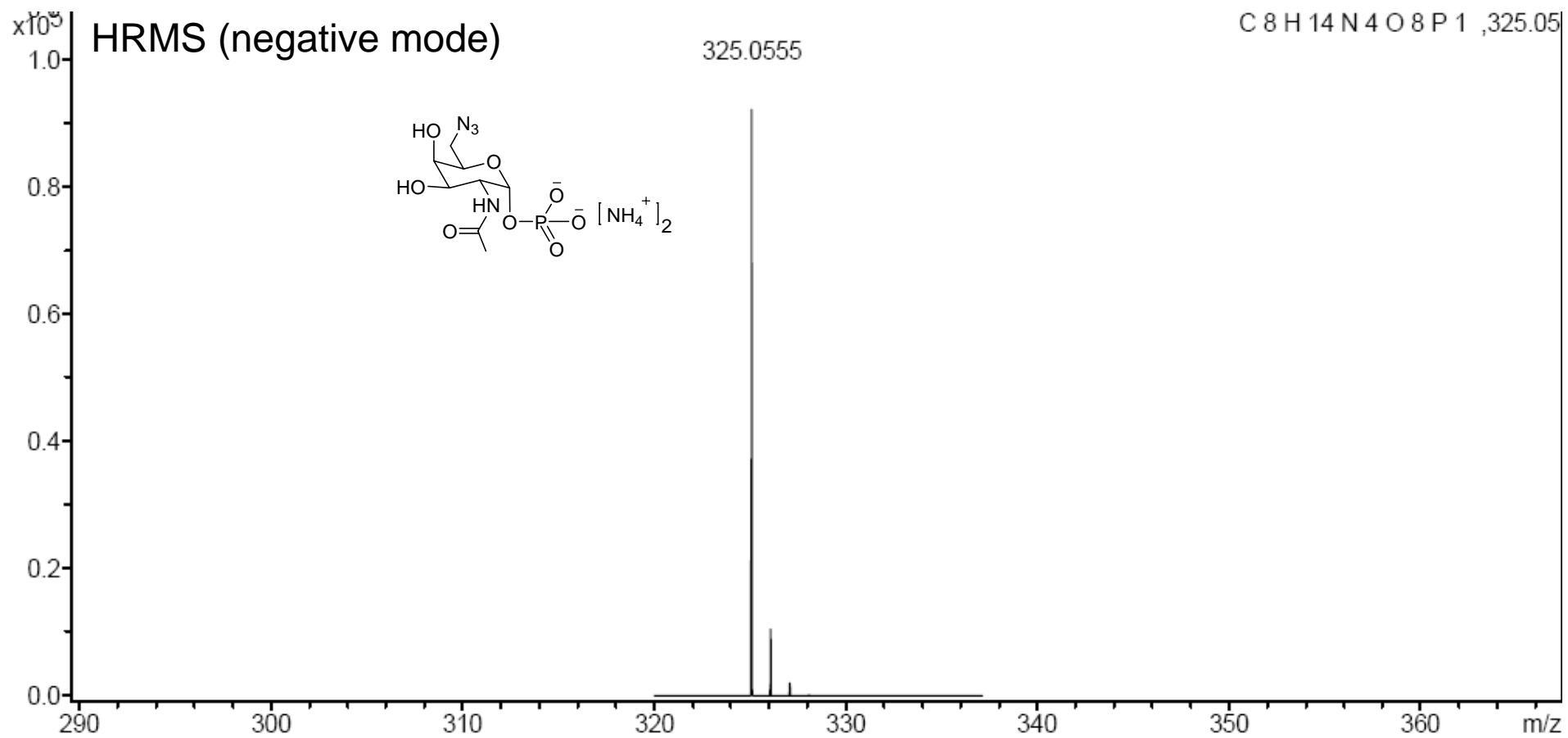


HRMS (negative mode)

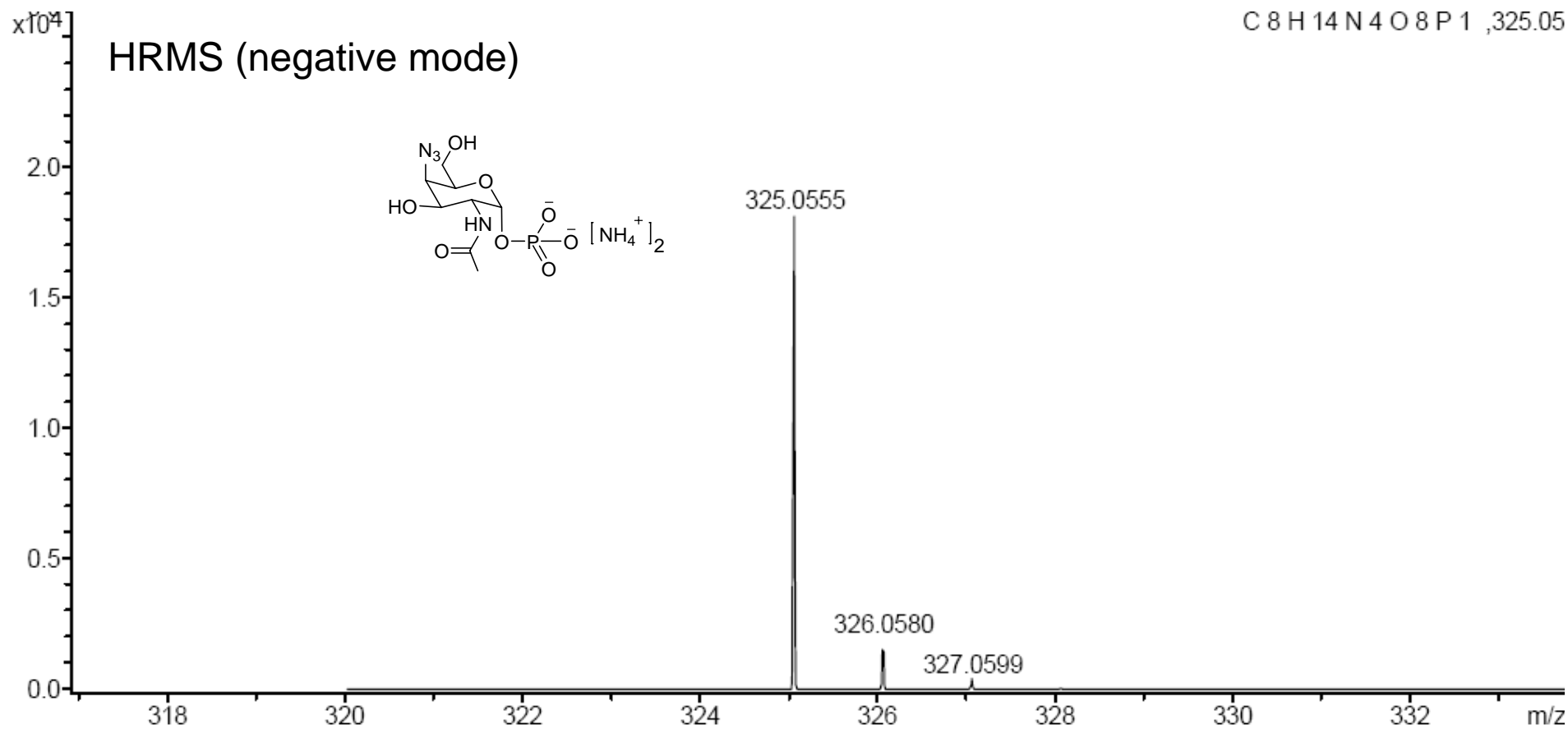


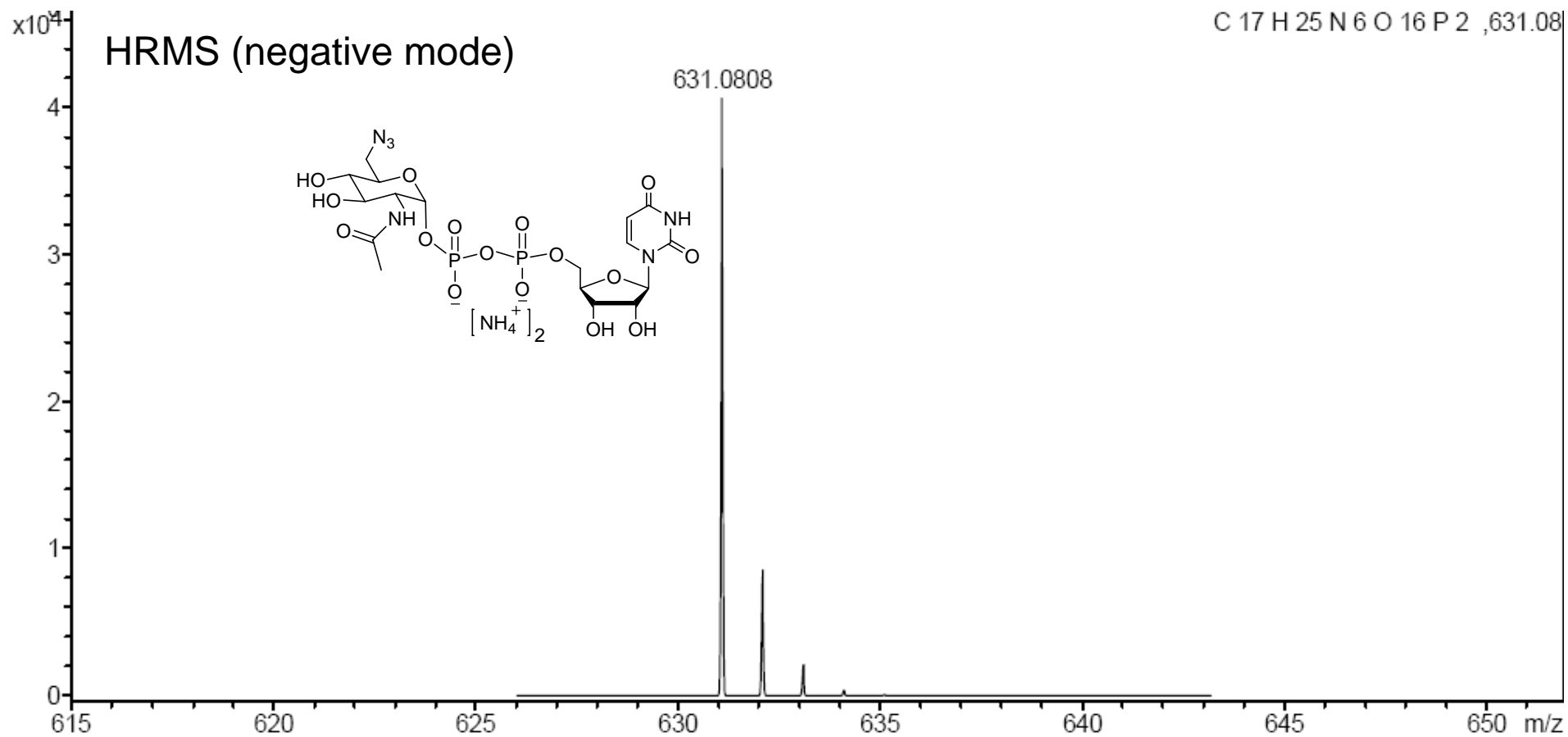






S5





S5

