

Supporting Information (SI)

O-acetylated sugars in the gas phase: stability, migration, positional isomers and conformation

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Proton nuclear magnetic resonances (¹H NMR) and carbon nuclear magnetic resonances (¹³C NMR) were recorded in deuterated solvents on Bruker AC spectrometers (400 MHz). Chemical shifts are reported in parts per million (ppm, δ) relative to the deuterated solvent (CD₃OD, δ 3.31 & 4.87 ppm, indicated by an asterisk on the spectra). Traces of residual solvents are indicated by circles. ¹H NMR splitting patterns are designated as singlet (s), doublet (d), triplet (t), doublet of doublets (dd), doublet of doublets of doublets (ddd), or multiplet (m) if it is not interpreted correctly. Coupling constants are reported in Hertz (Hz). The assignment of resonances for all compounds was made by two-dimensional homonuclear and heteronuclear chemical shift correlation experiments. Analytical thin layer chromatography (TLC) was carried out on Silica Gel plates (Merck 60 F254). TLC plates were analysed by heating plates that were dipped in a solution of 10% H₂SO₄ in EtOH (v/v). Solvent mixtures used for TLC and column chromatography are reported in v/v ratios. All commercially available reagents and solvents used without further purification, and all reactions were carried out in oven-dried glassware and under argon atmosphere.

Methyl 3-O-acetyl-2-acetamido-2-deoxy- β -D-glucopyranoside (1)

To a solution of methyl 2-acetamido-2-deoxy- β -D-glucopyranoside (200 mg, 0.86 mmol) in anhydrous pyridine (5 mL), 4,4'-dimethoxytrityl chloride (1.2 equiv., 1.03 mmol) was added. The resulting solution was stirred at room temperature for 10 hours. After control that all reactant were converted by TLC, acetyl chloride (3.6 equiv., 3.09 mmol) was added stepwise 3 x 1.2 equivalents at 0°C. Methanol (3.6 equiv., 3.10 mmol) was added and solvents were removed *in vacuo*. The orange solid as obtained was dissolved in 2 mL of a solution of 2% dichloroacetic acid in dichloromethane. After 2 minutes, the orange solution was directly purified by SiO₂ column chromatography (DCM-MeOH, 95:5 and after 9:1) to give the monoester **1**, a white solid (146 mg, 62%). ¹H NMR (400 MHz, MeOD): δ **4.96** (dd, J = 10.6, 9.1 Hz, 1H, H₃), **4.43** (d, J = 8.5 Hz, 1H, H₁), **3.89** (dd, J = 12.0, 2.3 Hz, 1H, H_{6a}), **3.78** (dd, J = 10.6, 8.5 Hz, 1H, H₄), **3.71** (dd, J = 12.0, 5.5 Hz, 1H, H_{6b}), **3.52** (dd, J = 9.0, 7.3, 1H, H₂), **3.48** (s, 3H, OMe), **3.35** (ddd, J = 9.8, 5.5, 2.3 Hz, 1H, H₅), **2.04** (s, 3H, OAc), **1.89** (s, 3H, NHAc); ¹³C NMR (101 MHz, MeOD): δ **173.39** (O-C=O), **172.47** (HN-C=O), **103.08** (C₁), **77.82** (C₅), **77.38** (C₃), **69.64** (C₂), **62.39** (C₆), **57.10** (OMe), **55.36** (C₄), **22.73**, (AcN), **20.86** (AcO); HRMS m/z calcd. for C₁₁H₁₉NNaO₇ (M+Na⁺) 300.1059, found 300.1054.

Methyl 6-O-acetyl-2-acetamido-2-deoxy- β -D-glucopyranoside (2)

Methyl 2-acetamido-2-deoxy- β -D-glucopyranoside (50 mg, 21.5 mmol) and triphenylphosphine (112.8 mg, 43 mmol) were suspended in anhydrous toluene (15 mL). To remove the water content, 10 mL of solvent were evaporated off and the remaining suspension was placed under argon before dimethylformamide (1 mL) and acetic acid (24.6 μ L, 43 mmol) were added. The reaction was placed at 0°C and diisopropyl azodicarboxylate (105.7 μ L, 3.75 mmol) was introduced. The reaction mixture was left stirring for 3 h and then directly subjected to a silica gel column chromatography using dichloromethane/methanol (85/15) as eluent. After evaporation, compound **2** was obtained as a white solid (18 mg, 39%).

¹H NMR (400 MHz, MeOD) δ **4.41** (dd, J = 11.9, 2.1 Hz, 1H, H_{6a}), **4.31** (d, J = 8.4 Hz, 1H, H₁), **4.22** (dd, J = 11.9, 5.8 Hz, 1H, H_{6b}), **3.63** (dd, J = 10.2, 8.4 Hz, 1H, H₃), **3.48 – 3.44** (m, 2H, H₂ and H₅), **3.43** (s, 3H, OMe), **3.37 – 3.33** (m, 1H, H₄), **2.07** (s, 3H, OAc), **1.97** (s, 3H, NHAc); ¹³C NMR (101 MHz, MeOD) δ **173.78** (HN-C=O), **172.74** (O-C=O), **103.51** (C₁),

75.96 (C₂), 75.27 (C₅), 72.02 (C₄), 64.70 (C₆), 57.16 (C₃), 56.91 (OMe), 22.92 (AcN), 20.68 (AcO); HRMS m/z calcd. for C₁₁H₁₉NNaO₇ (M+Na⁺) 300.1059, found 300.1054.

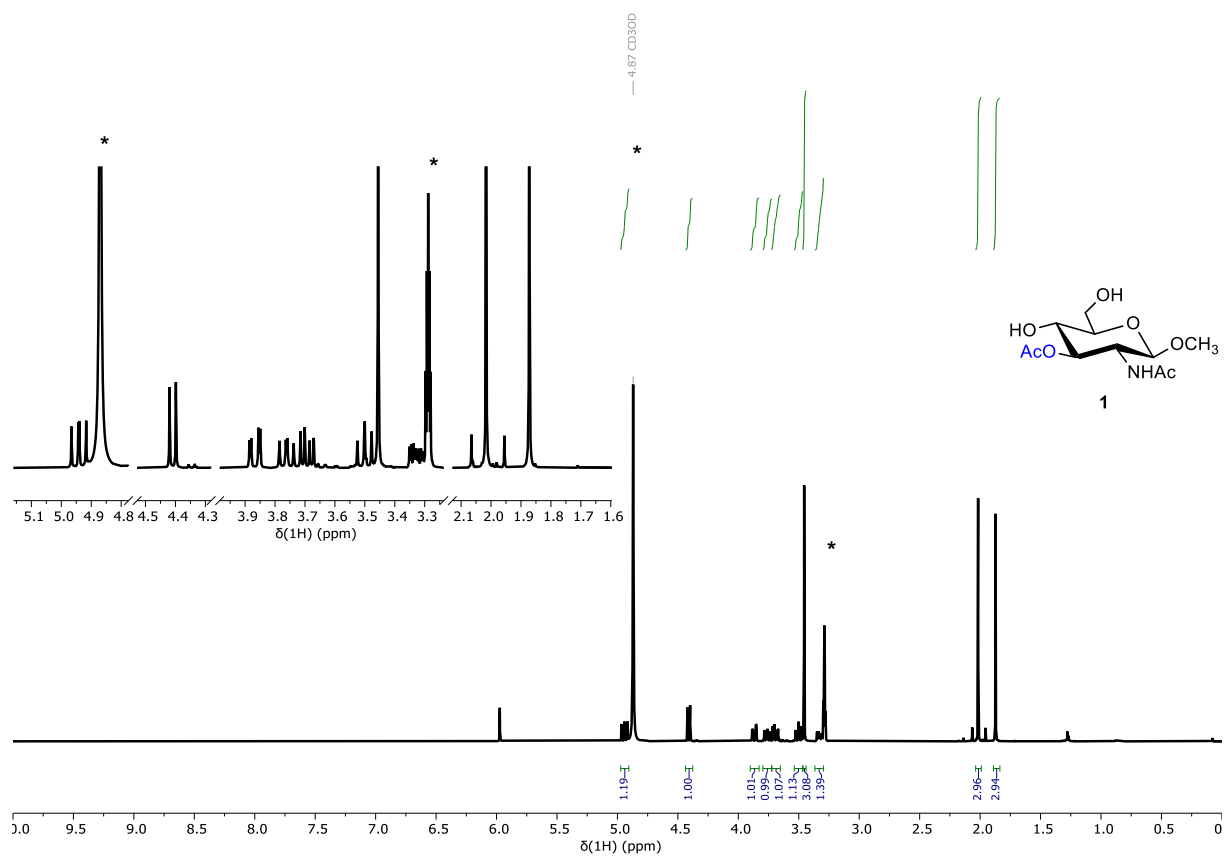


Fig S1. ¹H-NMR spectrum of β-GlcNAcOMe3OAc (**1**)

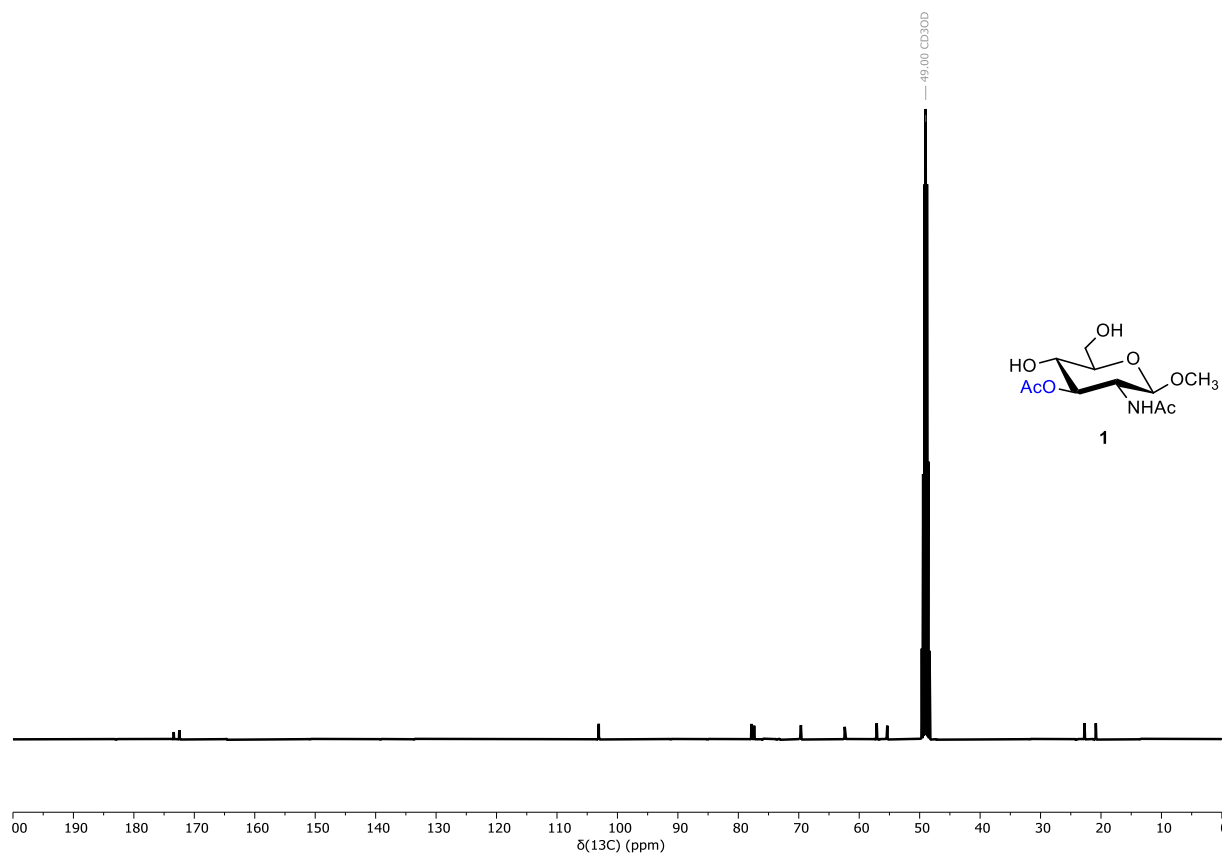


Fig S2. ^{13}C -NMR spectrum of β -GlcNAcOMe3OAc (**1**)

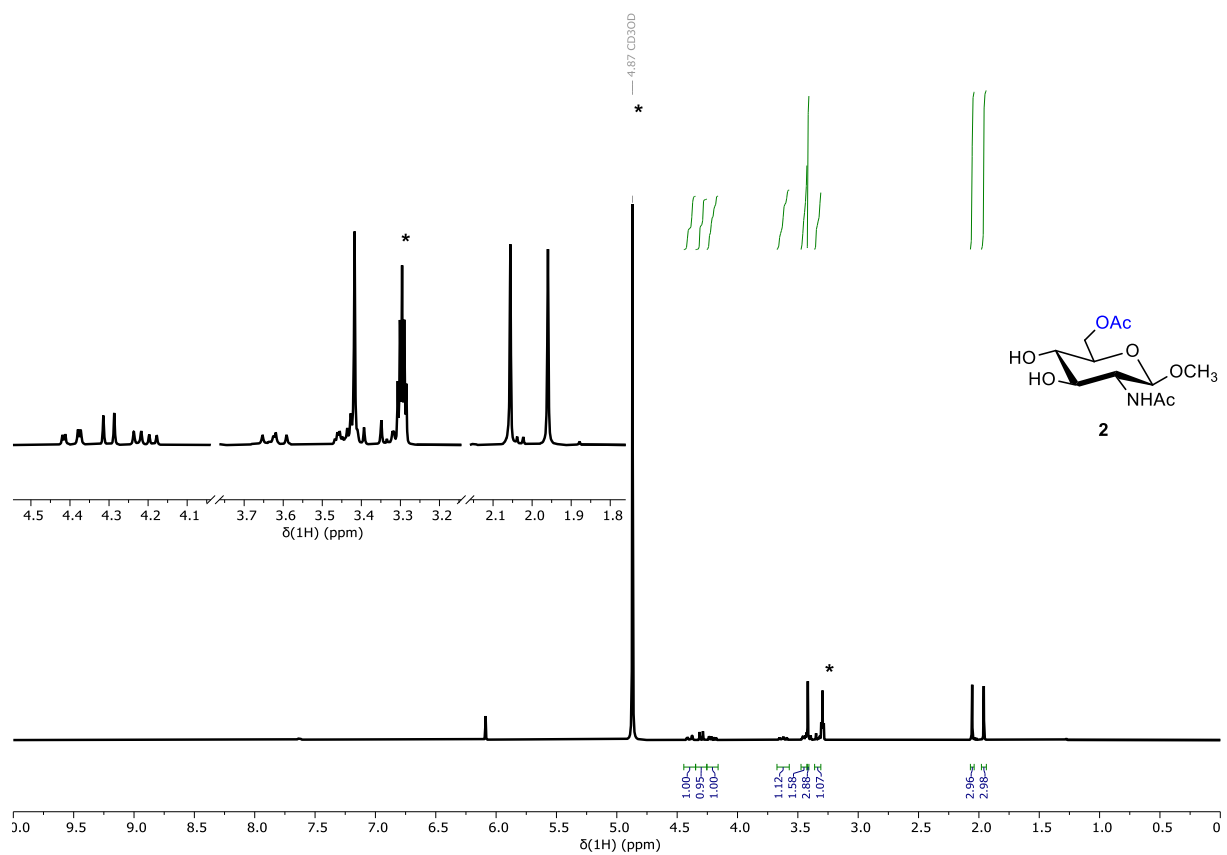


Fig S3. ^1H -NMR spectrum of β -GlcNAcOMe6OAc (**2**)

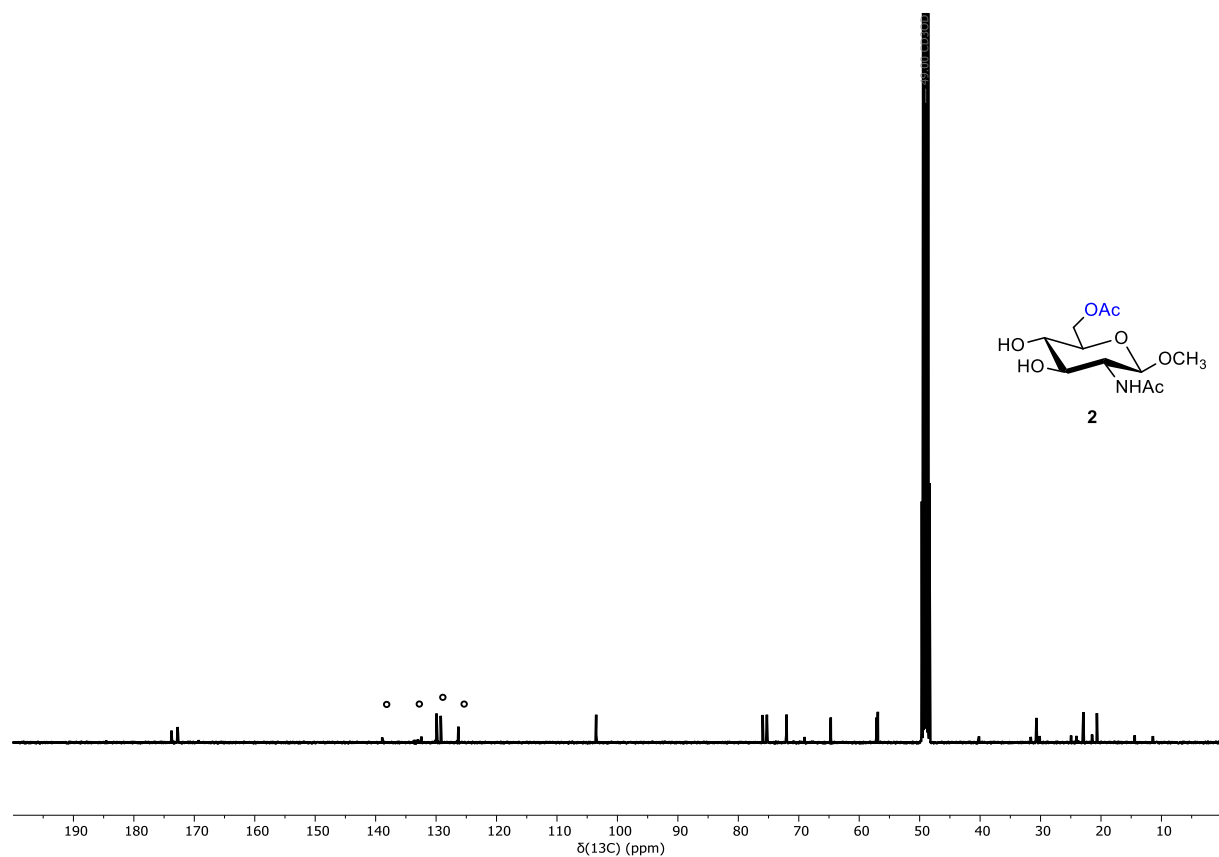


Fig S4. ¹³C-NMR spectrum of β -GlcNAcOMe6OAc (2)

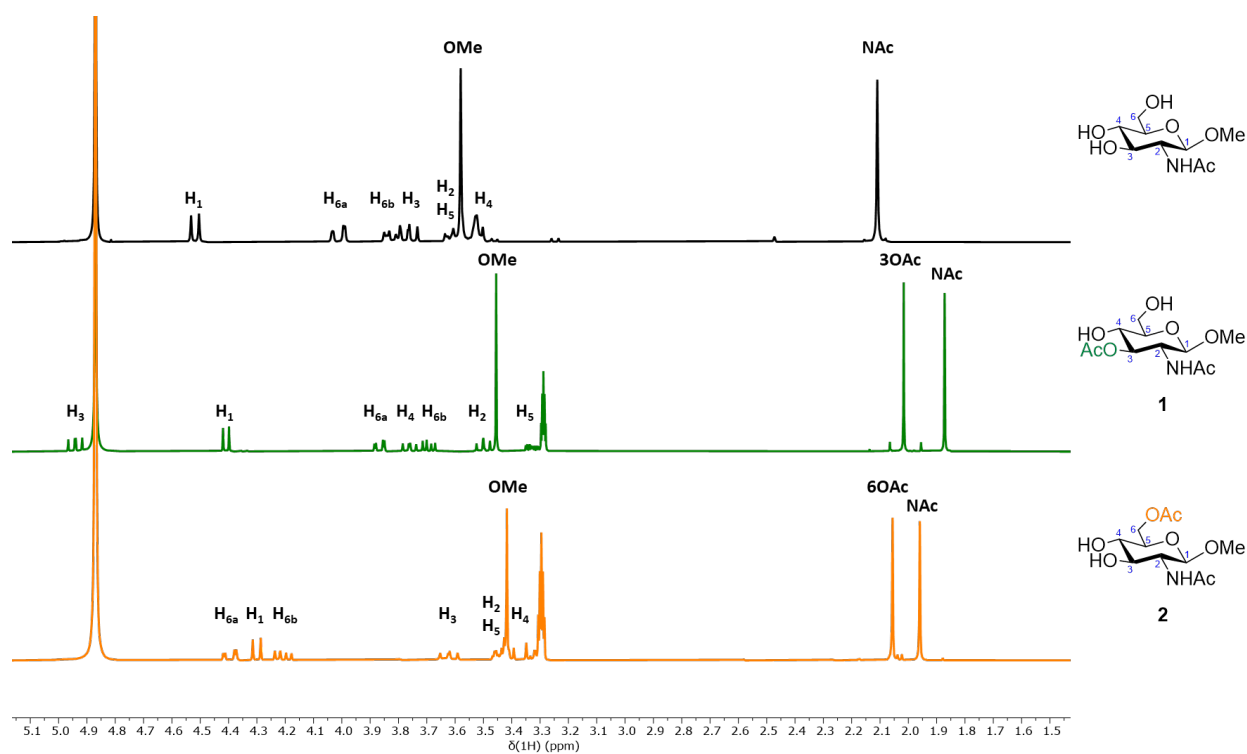


Fig S5. Assignment of the different protons of the ¹H-NMR spectra of β -GlcNAcOMe (black), compound 1 (green) and compound 2 (orange).

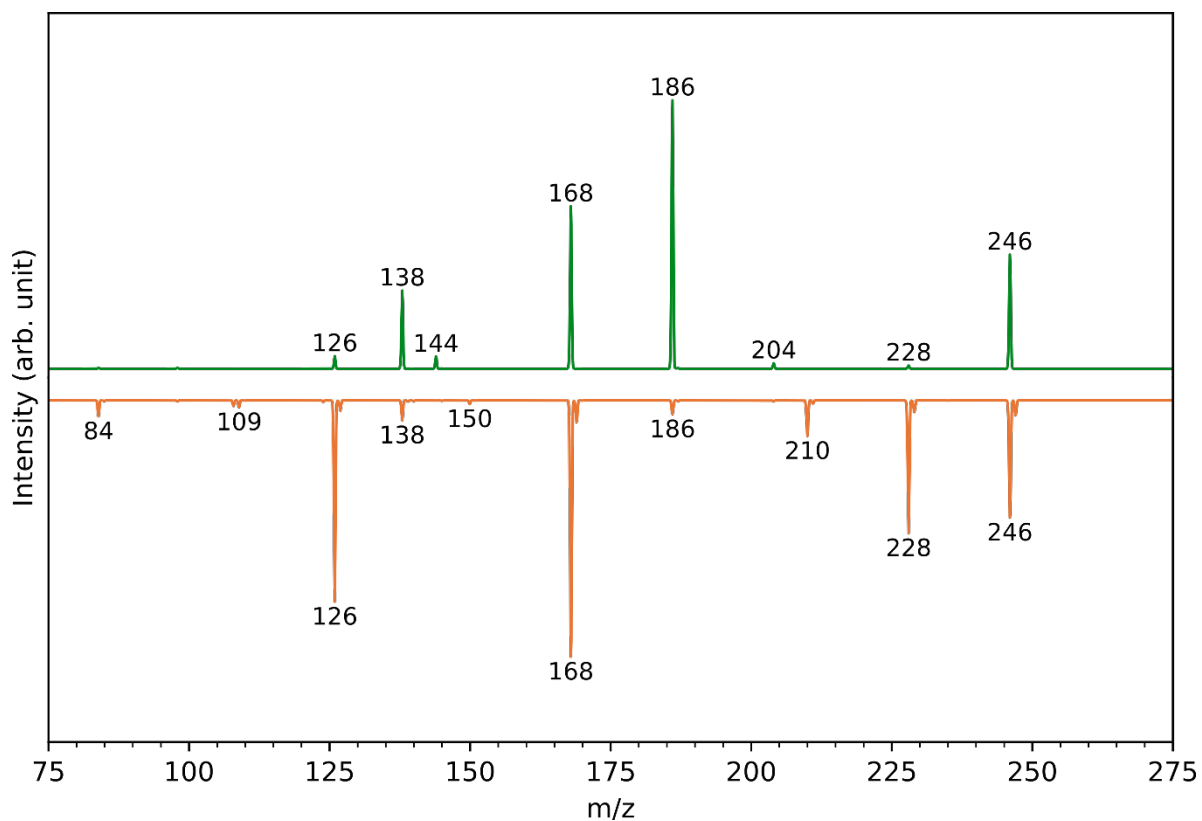
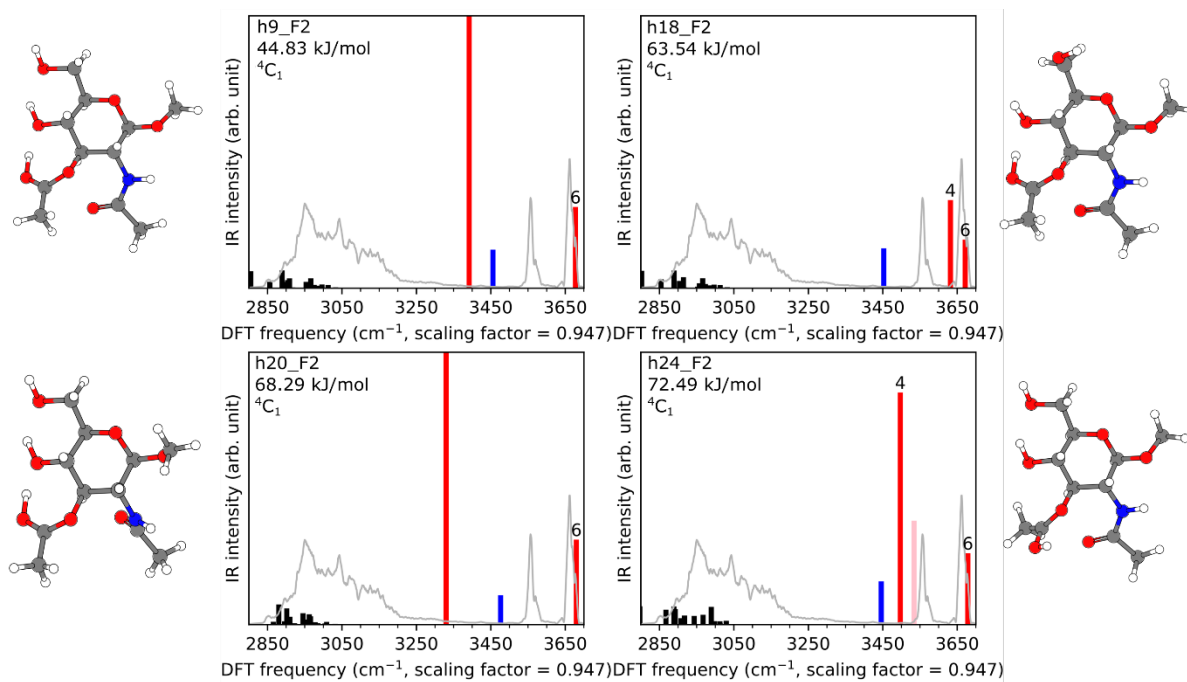


Fig S6. MS³ spectra of β GlcNAcOME acetylated on C3 (green) or C6 (orange)

a.



b.

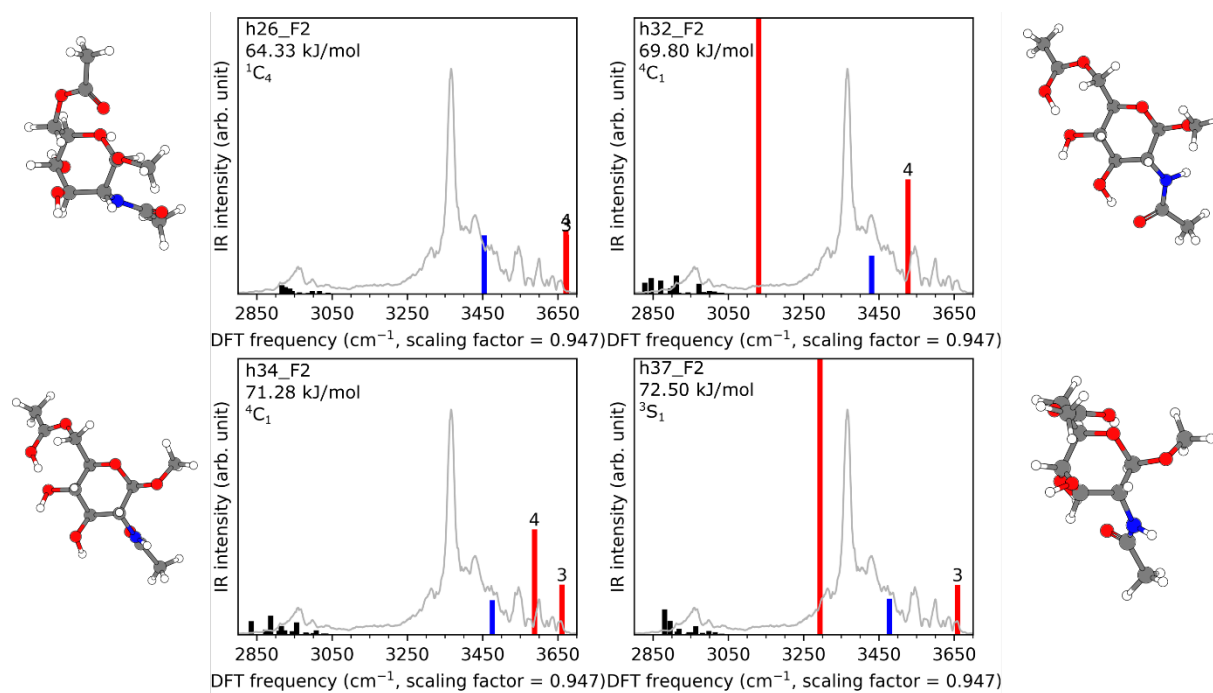


Fig S7. Experimental spectrum (grey line) compared with IR spectra of conformers with protonation on OAc of β GlcNAcOMe3OAc (a) and β GlcNAcOMe6OAc (b). OH stretches are colored in red (with label indicating its ring position), NH in blue, CH in black and acetyl OH in pink. The relative energies are given in reference to the most stable forms: the NAc protomers (4C_1 -3OAc and 4C_1 -6OAc, see figure 2)

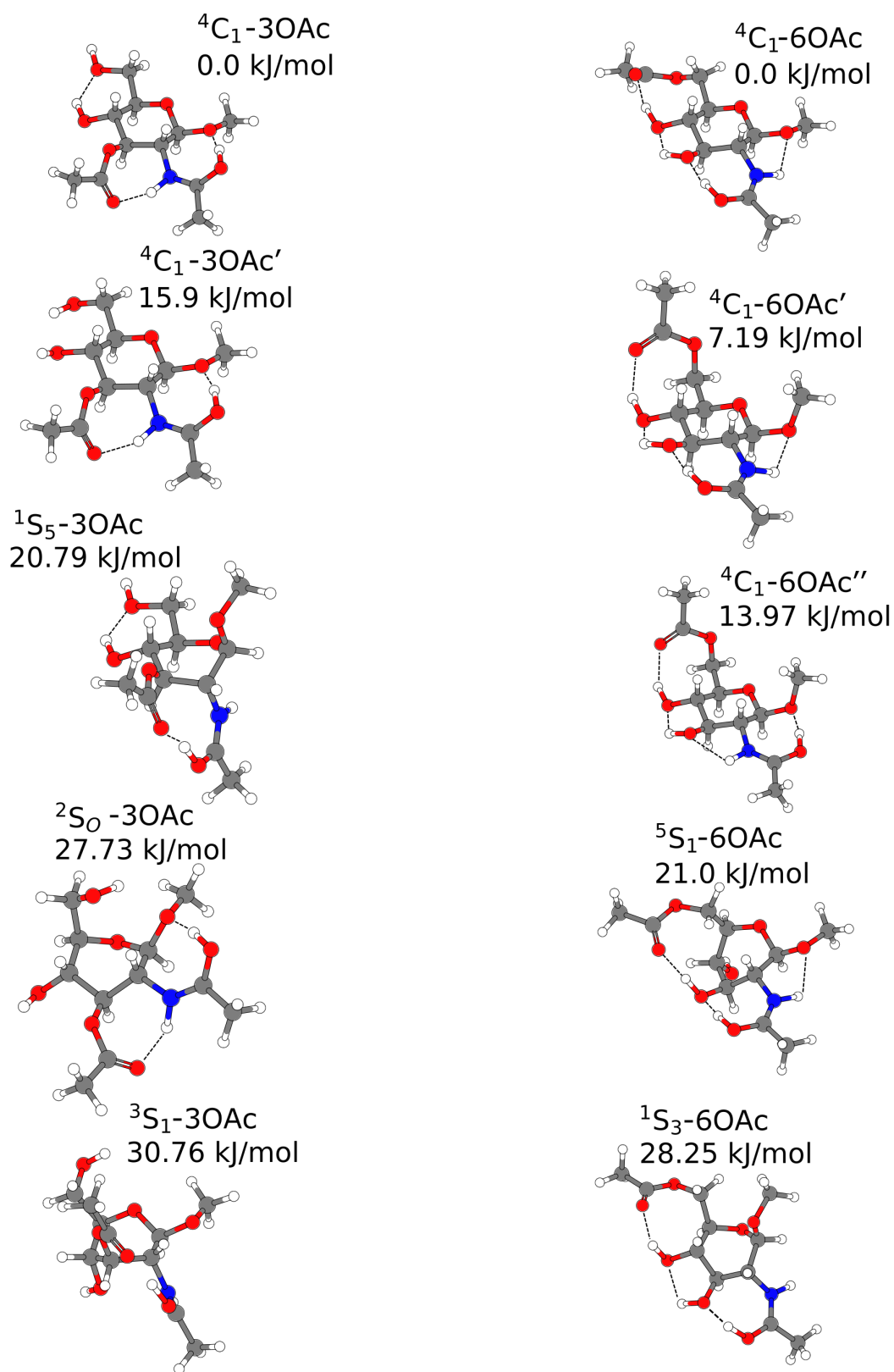


Fig S8. Molecular drawings of β GlcNAcOMe3OAc (left) and β GlcNAcOMe6OAc (right). Hydrogen bonds are represented by dotted lines.