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## Note

# $^{13}$C-N.m.r. studies of peracetylated derivatives of *O*-α- and *O*-β-D-galactopyranosyl-(1→3)- and -(1→4)-α-D-galactopyranose

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The $^{13}$C-n.m.r. spectra of galactose-containing oligosaccharides have been extensively studied[1], but few data on peracetylated derivatives have been reported[2–5]. Such derivatives are frequently prepared and reliable $^{13}$C-chemical shift data for these compounds are of value.

We now report the complete analysis by 2D-n.m.r. heteronuclear correlation shift experiments of the $^{13}$C-n.m.r. spectra of 1,2,4,6-tetra-*O*-acetyl-3-*O*-(2,3,4,6-tetra-*O*-acetyl-α-D-galactopyranosyl)-α-D-galactopyranose (**1**), 1,2,4,6-tetra-*O*-acetyl-3-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranosyl)-α-D-galactopyranose (**2**), 1,2,3,6-tetra-*O*-acetyl-4-*O*-(2,3,4,6-tetra-*O*-acetyl-α-D-galactopyranosyl)-α-D-galactopyranose (**3**), 1,2,3,6-tetra-*O*-acetyl-4-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranosyl)-α-D-galactopyranose (**4**), methyl 2,3,4,6-tetra-*O*-acetyl-α-D-galactopyranoside (**5**), methyl 2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranoside (**6**), 1,2,4,6-tetra-*O*-acetyl-3-*O*-methyl-α-D-galactopyranose (**7**), and 1,2,3,6-tetra-*O*-acetyl-4-*O*-methyl-α-D-galactopyranose (**8**). The methylation- and glycosylation-induced shifts, calculated from the spectra of compounds **1–8** and from those of some monosaccharide[6–10] (**9–14**) and disaccharide[2,3] (**15–22**) derivatives reported in the literature, are compared and discussed.

The $^{13}$C-n.m.r. chemical shifts of the peracetylated disaccharide (**1–4**) and monosaccharide (**5–8**) derivatives are shown in Table I. The $^{13}$C-n.m.r. spectra of compounds **5** and **6** have been assigned[6–8,11]. Our assignments accord with those reported by Kalinovskii and Evtushenko[11], but differ from those given by other authors[7,8]. The spectra of compounds **1–8** have been analysed by 2D-n.m.r. heteronuclear correlation shift experiments which allowed the assignment of most resonances of the sugar rings. In a few cases in which resolution (0.09 p.p.m. in the $^{13}$C axis) was not enough to resolve the signals, the assignments were confirmed by measuring the $^{13}$C–$^{1}$H residual couplings in a series of SFORD experiments with low-power coherent irradiation and different irradiation offsets, and calculating the $^{1}$H-chemical shift of the proton directly bonded to the observed carbon atom by

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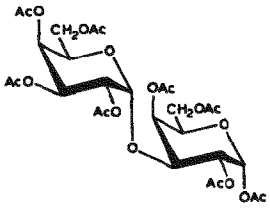

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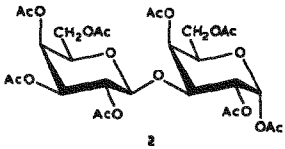

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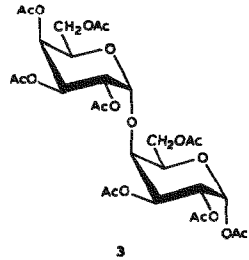

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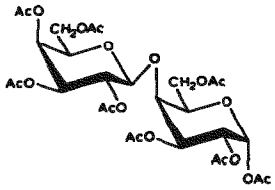

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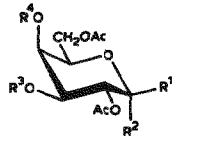
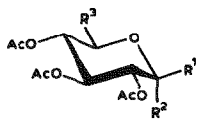
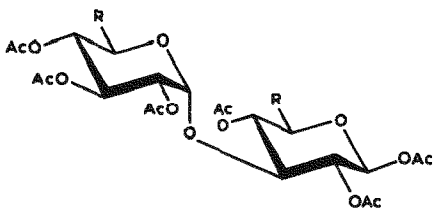


| | $R^1$ | $R^2$ | $R^3$ | $R^4$ |
|---|---|---|---|---|
| 5 | H | OMe | Ac | Ac |
| 6 | OMe | H | Ac | Ac |
| 7 | H | OAc | Me | Ac |
| 8 | H | OAc | Ac | Me |
| 9 | H | OAc | Ac | Ac |
| 10 | OAc | H | Ac | Ac |

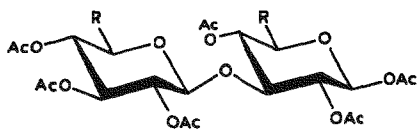


| | $R^1$ | $R^2$ | $R^3$ |
|---|---|---|---|
| 11 | H | OMe | $CH_2OAc$ |
| 12 | OMe | H | $CH_2OAc$ |
| 13 | H | OMe | H |
| 14 | OMe | H | H |



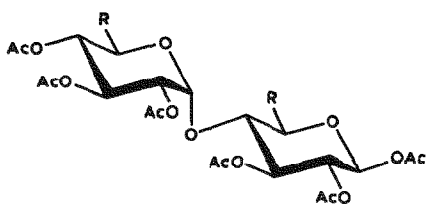
15 R = $CH_2OAc$

19 R = H



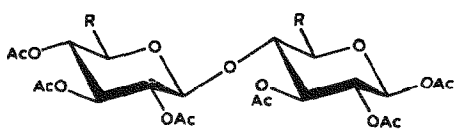
16 R = $CH_2OAc$

20 R = H



17 R = $CH_2OAc$

21 R = H



18 R = $CH_2OAc$

22 R = H

TABLE I

$^{13}$C-N.M.R. CHEMICAL SHIFTS[a] FOR COMPOUNDS **1–8**

| *Compound* | *C-1* | *C-2* | *C-3* | *C-4* | *C-5* | *C-6* | *C-1'* | *C-2'* | *C-3'* | *C-4'* | *C-5'* | *C-6'* | *OMe* |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **1** | 89.63 | 66.56 | 68.78 | 65.51 | 68.95 | 61.35 | 93.21 | 67.77 | 67.09 | 67.63 | 66.46 | 61.00 | |
| **2** | 89.85 | 68.92 | 71.86 | 68.99 | 69.53 | 62.12 | 100.90 | 68.92 | 70.69 | 66.67 | 70.69 | 60.90 | |
| **3** | 89.78 | 66.18 | 69.23 | 77.04 | 70.35 | 61.74 | 99.15 | 68.18 | 67.26 | 67.77 | 67.09 | 60.62 | |
| **4** | 89.96 | 66.17 | 70.04 | 74.52 | 70.31 | 63.47 | 101.88 | 68.65 | 70.76 | 66.81 | 70.69 | 61.29 | |
| **5** | 97.17 | 68.10 | 67.52 | 68.14 | 66.17 | 61.79 | | | | | | | 55.41 |
| **6** | 101.83 | 68.86 | 70.97 | 67.36 | 70.59 | 61.44 | | | | | | | 56.65 |
| **7** | 89.89 | 68.16 | 75.19 | 65.91 | 69.09 | 61.88 | | | | | | | 57.82 |
| **8** | 89.68 | 66.88 | 70.49 | 76.73 | 70.41 | 62.26 | | | | | | | 61.56 |

[a]In p.p.m. from internal $Me_4Si$ measured in $CDCl_3$.

TABLE II

METHYLATION- AND GLYCOSIDATION-INDUCED SHIFT EFFECTS[a] FOR COMPOUNDS **1–22**

| *Compound* | *C-1* | *C-2* | *C-3* | *C-4* | *C-5* | *C-6* | *C-1'* | *C-2'* | *C-3'* | *C-4'* | *C-5'* | *C-6'* |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **1** | 0.1 | −0.6 | 1.6 | −0.7 | 0.4 | 0.3 | 3.7 | 0.6 | −0.1 | 1.4 | −2.0 | 0.0 |
| **2** | 0.3 | 1.7 | 4.7 | 2.8 | 1.0 | 1.1 | 9.1 | 1.1 | 0.1 | −0.1 | −0.8 | −0.1 |
| **3** | 0.3 | −1.0 | 2.0 | 10.8 | 1.8 | 0.7 | 9.7 | 1.0 | 0.1 | 1.6 | −1.4 | −0.4 |
| **4** | 0.5 | −1.0 | 2.8 | 8.3 | 1.8 | 2.5 | 10.1 | 0.8 | 0.2 | 0.0 | −0.8 | 0.3 |
| **15** | 0.1 | 0.7 | 4.8 | 0.8 | 0.0 | 0.0 | 7.1 | 2.0 | −0.5 | 0.0 | −1.4 | −0.3 |
| **16** | 0.0 | 1.3 | 6.0 | −0.4 | −0.1 | 0.2 | 9.2 | 0.7 | 0.1 | 0.1 | −1.0 | 0.2 |
| **17** | −0.4 | 0.6 | 2.2 | 4.9 | 0.2 | 1.1 | 6.7 | 0.9 | −0.5 | 0.2 | −1.2 | 0.1 |
| **18** | −0.2 | 1.9 | 2.4 | 7.9 | 0.7 | 0.1 | 8.9 | 1.1 | 0.0 | −0.1 | −0.9 | 0.1 |
| **19** | 0.1 | 0.2 | 2.9 | 0.9 | −0.9 | — | 7.1 | 1.9 | 0.0 | 0.2 | −1.5 | — |
| **20** | 0.4 | 0.8 | 6.1 | 0.5 | −0.1 | — | 9.3 | 1.0 | 0.0 | 0.5 | −0.7 | — |
| **21** | 0.3 | 0.9 | 2.4 | 4.9 | 1.8 | — | 7.7 | 1.7 | −0.1 | 0.2 | −1.7 | — |
| **22** | 0.7 | 0.8 | 1.4 | 6.2 | 1.0 | — | 8.0 | 1.4 | 0.0 | 0.5 | −0.7 | — |
| **5** | 7.7 | 0.9 | 0.3 | 1.9 | −2.3 | 0.8 | | | | | | |
| **6** | 10.0 | 1.1 | 0.4 | 0.6 | −0.9 | 0.5 | | | | | | |
| **7** | 0.4 | 1.0 | 8.0 | −0.3 | 0.6 | 0.9 | | | | | | |
| **8** | 0.5 | −0.3 | 3.3 | 10.5 | 1.9 | 1.3 | | | | | | |
| **11** | 7.1 | 1.1 | −0.2 | 0.2 | −3.1 | −0.1 | | | | | | |
| **12** | 9.8 | 0.7 | 0.0 | 0.5 | −1.2 | 0.3 | | | | | | |
| **13** | 7.5 | 1.3 | −0.1 | 0.0 | −2.8 | — | | | | | | |
| **14** | 9.3 | 0.9 | 0.2 | 0.2 | −1.2 | — | | | | | | |

[a]Calculated (in p.p.m.) as the differences between the chemical shifts of the carbons of the compound and those of the corresponding carbons in the peracetylated pyranose; downfield shifts are indicated by positive values.

using the equation of Anderson[12,13]. This method allowed the assignment of $^{13}C$ signals whose chemical shifts were very similar, provided that the chemical shifts of the protons directly bonded to those carbon atoms differed by >10 Hz.

*O*-Methyl derivatives of monosaccharides have been used as model compounds in the analysis of the $^{13}C$-n.m.r. spectra of oligo- and poly-saccharides[14]. We have now extended these studies to acetylated derivatives of D-galactopyranose. Induced shift effects due to methylation have been calculated as the differences between the chemical shifts of the signals assigned to the carbon atoms of the methylated compounds and those of the peracetylated compounds. Comparison (Table II) of the chemical shifts of the anomeric carbon of peracetylated methyl D-galactopyranosides (**5** and **6**) with those of peracetylated D-galactopyranoses (**9** and **10**) shows a downfield $\alpha$-effect upon methylation of ~7.4 p.p.m. for the $\alpha$ anomer and ~9.7 p.p.m. for the $\beta$ anomer. This effect does not seem to depend on the configuration at C-4 or the substitution at C-5, since D-glucopyranose (**11** and **12**) and D-xylopyranose (**13** and **14**) derivatives display almost identical downfield shifts[8–10]. The interaction of the methyl group with H-5 in the peracetylated methyl $\alpha$-D-glycopyranosides (**5**, **11**, and **13**) caused an upfield shift of the signal assigned to C-5 of ~2.7 p.p.m. The $\alpha$-effects caused by methylation at C-3 and C-4 (compounds **7** and **8**) are of the same order as that at C-1. No rule can be stated for methylation-induced $\beta$-effects with the data available at present.

The shifts induced by glycosylation have been calculated as for those caused by methylation. Comparison of the $^{13}C$-chemical shifts for the peracetylated disaccharides with those for the structurally related peracetylated monosaccharides (Table II) indicated that the glycosylation shifts at C-1′, C-3, and C-4 ($\alpha$-effect) follow the same trends as for methylation at C-1. As a rule, these $\alpha$-effects are smaller when the configuration at C-1 is $\alpha$, the lowest values being for $\alpha$-(1→3)-linked disaccharides especially for **1** (3.7 p.p.m. for C-1′ and 1.6 p.p.m. for C-3). These anomalous values may be explained by taking into consideration the fact that, in the major conformation, the distance between H-1′ and H-3′ becomes larger, thereby decreasing the downfield shift of the signals assigned to both C-1′ and C-3[15,16]. This conformation would also explain the anomalous upfield induced shift at C-4 as a $\gamma$ interaction between H-1′ and H-4. Finally, there is a shielding effect of −1.5 p.p.m. (mean value) for C-5′ in peracetylated $\alpha$-linked disaccharides (**1, 3, 15, 18,** and **21**) *versus* −0.8 p.p.m. for the corresponding $\beta$ anomers.

## EXPERIMENTAL

*Materials.* — Compounds **1–8** were synthesised as previously reported[17,18].

*N.m.r. spectroscopy.* — $^1H$-Decoupled $^{13}C$-n.m.r. spectra of compounds **1–8** were recorded with a Varian XL-300 spectrometer, operating at 75 MHz, and 0.2M solutions in $CDCl_3$ (internal $Me_4Si$) at 293 K. The acquisition parameters were: spectral width, 16 kHz; computer data points, 32k; acquisition time, 1 s; resolution, 0.003 p.p.m. per point; pulse width, 5.5 $\mu$s (42°). SFORD $^{13}C$-n.m.r. spectra were obtained under the above conditions with low-power, selective, proton decoupling at different irradiation offsets with an irradiation power of 300 Hz. $^1H$-N.m.r. spectra were recorded with the same spectrometer operating at 300 MHz. 2D-Heteronuclear correlation experiments were carried out using the Varian software[19]. Typical parameters were: spectral widths, 1000 Hz ($^1H$ axis) and 3500 Hz ($^{13}C$ axis); relaxation delay, 2 s; number of increments, 256; 90° pulse width for $^{13}C$ and $^1H$, 11.7 $\mu$s respectively; and 1024 × 512 transformed data points; $^{13}C$-$^1H$ direct couplings were assumed to be 150 Hz.

## ACKNOWLEDGMENTS

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