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Note

Proton and carbon-13 n.m.r. spectra of six pseudo-2-acetamido-2-deoxy-DL-hexopyranoses

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Pseudo-(amino sugars), carbocyclic analogs of aminodeoxyhexopyranoses, have been found in such biologically active substances as antibiotics and enzyme inhibitors, as described by Ogawa *et al.*[1,2]. These authors described the synthesis of the fully *O*-acetylated derivatives of the pseudo-2-acetamido-2-deoxy-DL-hexopyranoses having the α- and β-DL-*galacto* (**1,2**), α- and β-DL-*gluco* (**3,4**), and α- and β-DL-*manno* (**5,6**) configurations[2].

We now report the preparation of the unprotected pseudo-2-acetamido-2-deoxy-DL-hexopyranoses **1–6**, together with a complete analysis of their ^{1}H- and ^{13}C-n.m.r.-spectral data for solutions in D_2O, in order to complement our previous publication on the n.m.r. data for the corresponding pseudo-hexopyranoses[3].

Compounds **1** and **3–6** were prepared by *O*-deacetylation of the corresponding *N*-acetyltetra-*O*-acetyl derivatives, synthesized previously[1,2,4,5], whereas compound **2** was prepared from the 4,7-*O*-isopropylidene derivative[4,5] by treatment with aqueous acetic acid (80%).

The ^{1}H- and ^{13}C-n.m.r. data were measured at 500 and 125.7 MHz, respectively, which allowed a complete analysis of all the ^{1}H-n.m.r. chemical shifts and coupling constants, and the ^{13}C-n.m.r. chemical shifts.

The ^{1}H-n.m.r. data for **1–6** (as 0.1M solutions in D_2O) at 300 K, are shown in Table I. The assignments are based on homonuclear COSY experiments[6]. The coupling constants were determined on a first-order basis from the one-dimensional spectra. The proton-decoupled ^{13}C-n.m.r. spectra were recorded likewise, and the results are given in Table II, together with the data for the corresponding *N*-acetylhexosamines. The assignments of the signals for pseudo-hexoses were based on heteronuclear shift correlation experiments (CHORTLE)[7].

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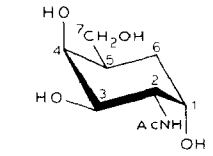

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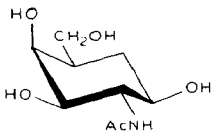

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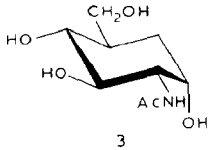

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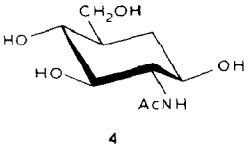

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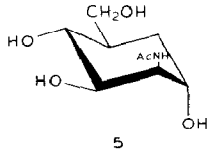

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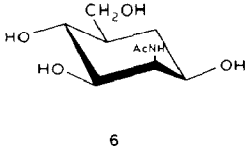

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The observed vicinal coupling constants (see Table I) for the ring protons of **1** to **6** all suggest that no significant distortion of the $^4C_1$ conformations take place in the pseudo-*N*-acetyl-sugars, in analogy with the results from the corresponding pseudo-sugars[3]. This conclusion is based on the dependence of the coupling constants on torsion angles, for neighboring protons (H-2 to H-4) on carbon atoms which both carry an oxygen substituent (calculated using the CAGPLUS program[8,9]).

A comparison of the proton n.m.r. chemical shifts with those published[10] for the methyl glycosides of the monosaccharides corresponding to **1, 2, 3** and **4** show, in addition to the expected shifts for H-1 and H-5 of ~0.8 and ~1.8 p.p.m., respectively, that the H-2 atoms in a compounds **1** and **3** are shifted upfield by 0.2 p.p.m. relative to the corresponding methyl glycosides, whereas this shift difference is much smaller (0.03 p.p.m.) for the corresponding β compounds **2** and **4**. On the other hand, the H-4 atoms of the pseudo-2-acetamido-2-deoxy-DL-galacto-pyranoses (**1** and **2**) are shifted ~0.12 p.p.m. downfield relative to the corresponding methyl glycosides, whereas the H-4 atoms in the *gluco* derivatives **3** and **4** are shifted ~0.1 p.p.m. upfield relative to the corresponding methyl glycosides.

Furthermore, the chemical shifts for H-6 and H-6′ (sugar numbering — see chart) and the coupling constants $J_{5,6}$, are somewhat different from those published[11] for the 2-acetamido-2-deoxy-α- (and β-)D-glucopyranosides, assuming that the assignment for H-6 is H-6*S* and for H-6′ is H-6*R*, in analogy with the data presented by Nishida *et al.*[11]. These results are comparable with the observation made for the pseudo-hexoses[2], most likely originating from a different rotameric distribution of the 5-*C*-(hydroxymethyl) group. However the coupling constants for the *gluco* and *manno* derivatives **3, 4, 5**, and **6** indicate that the *gg* and *gt* rotaconformers are the

TABLE I

^{1}H-N.M.R. DATA FOR *N*-ACETYL-PSEUDO-HEXOSAMINES **1–6**

Configuration of pseudo-sugar	*Compound number*	*Chemical shifts*[a]									
		H-1	*H-2*	*H-3*	*H-4*	*H-5*	*H-6*	*H-6'*	*H-7*	*H-7'*	*N-Ac*
α-GalNAc	1	4.10	3.98	3.80	4.12	2.04	3.67	3.54	1.65	1.60	2.0
β-GalNAc	2	3.57	3.91	3.50	4.06	1.78	3.69	3.57	1.82	1.44	2.0
α-GlcNAc	3	4.08	3.70	3.63	3.37	1.89	3.74	3.69	1.91	1.53	2.0
β-GlcNAc	4	3.60	3.63	3.31	3.38	1.62	3.78	3.66	2.07	1.35	2.0
α-ManNAc	5	3.99	4.28	3.94	3.59	1.91	3.74	3.73	1.81	1.67	2.0
β-ManNAc	6	3.97	4.57	3.65	3.46	1.61	3.78	3.70	1.89	1.47	2.1
		Coupling constants (Hz)[b]									
		$J_{1,2}$ $J_{1,7}$ $J_{1,7'}$	$J_{2,3}$ $J_{2,7}$	$J_{3,4}$	$J_{4,5}$ $J_{4,7}$	$J_{5,6}$ $J_{5,6'}$ $J_{5,7}$ $J_{5,7'}$	$J_{6,6'}$	$J_{7,7'}$			
α-GalNAc	1	3.0 3.0 3.0	11.1	3.0	2.4 0.8	7.9 6.3 4.0 13.0	11.0	14.0			
β-GalNAc	2	10.5 4.5 11.0	10.4	2.9	2.4 0.8	7.7 6.5 4.4 12.0	11.0	13.0			
α-GlcNAc	3	2.8 2.8 2.8	10.5	9.0	10.2	3.8 5.6 13.0	11.0	15.0			
β-GlcNAc	4	10.5 4.0 10.5	9.5	9.5	9.5	3.6 6.2 4.0 12.0	11.0	13.0			
α-ManNAc	5	3.5 4.0 2.9	4.7 1.5	9.6	9.6	4.6 5.8 4.0 12.1	11.8	15.0			
β-ManNAc	6	4.4 4.3 11.9	4.3 1.3	10.0	10.1	3.5 5.9 4.5 12.5	11.2	13.2			

[a]P.p.m. from Me_4Si, measured from acetone as the internal reference (δ 2.22). [b]Observed first-order coupling-constants (±0.3 Hz).

favored conformations observed and almost equally populated, using the method described recently[12]. In Figs. 1A and 1B are shown the angular dependence of the H-5 to H-6*R* and H-5 to H-6*S* coupling constants for pseudo-hexoses and hexoses, respectively, using the CAGPLUS program[8]. Based on the limiting values for the staggered conformations (*gt, gg, tg*) as discussed in ref. 13, it can be estimated that

TABLE II

^{13}C-N.M.R. DATA FOR *N*-ACETYL-PSEUDO-HEXOSAMINES **1–6** AND CORRESPONDING TRUE SUGARS

Configuration	*Compound number*	*Chemical shifts*[a]							
		C-1	*C-2*	*C-3*	*C-4*	*C-5*	*C-6*	*C-7*	*N-Ac*
		Pseudo-hexosamines							
α-GalNAc	**1**	68.4	53.2	70.2	70.3	36.9	63.4	28.6	22.8
β-GalNAc	**2**	71.1	56.6	73.5	69.7	39.1	63.1	30.1	23.1
α-GlcNAc	**3**	68.3	56.7	73.2	74.6	38.8	63.1	32.0	22.8
β-GlcNAc	**4**	70.5	59.5	75.9	74.0	40.8	63.0	33.7	23.1
α-ManNAc	**5**	68.3	55.3	71.3	71.2	39.6	63.0	29.5	22.8
β-ManNAc	**6**	68.1	55.5	73.6	70.7	41.2	62.9	30.4	23.0
		True hexosamines[b]							
α-GalNAc		92.2	51.4	68.6	69.7	71.6	62.4		
β-GalNAc		96.5	54.9	72.3	69.0	76.3	62.2		
α-GlcNAc		92.1	55.3	72.0	71.4	72.8	61.9		
β-GlcNAc		96.2	58.0	75.2	71.2	77.2	62.0		
α-ManNAc		94.3	54.4	70.1	68.0	73.2	61.7		
β-ManNAc		94.3	55.3	73.2	67.8	77.5	61.7		

[a]P.p.m. from Me_4Si, measured from 1,4-dioxane as the internal reference (δ 67.4). [b]Data taken from ref. 11.

the population of the *tg* conformer for compounds **3** to **6** is 10%, and that the *gg* and *gt* conformers are almost equally populated. For compounds **1** and **2**, however, the *gg* conformer is populated ~10%, and the *gt* conformer preponderates over the *tg* conformer, but the ratio is quite different from the results observed for the corresponding hexopyranosides[14].

A comparison of the ^{13}C-n.m.r. chemical shifts (see Table II) for **1–6** with the data published[15] for the corresponding *N*-acetylhexosamines shows significant changes only for C-1 and C-5 (23–26 and 33–37 p.p.m., respectively). All other chemical-shift differences are in the range 0–3 p.p.m. and do not suggest any major conformational differences between the two classes of compounds.

EXPERIMENTAL

General methods. — Melting points were determined with a Mel-Temp capillary melting-point apparatus and are uncorrected. T.l.c. was performed on Wakogel B-10 (Wako Co., Osaka, Japan), with detection by charring with 10% sulfuric acid. Organic solutions were dried (anhydrous Na_2SO_4), and evaporated at <50° under diminished pressure.

N.m.r. spectra were recorded with a Bruker AM 500 spectrometer operated at 500 MHz for ^{1}H spectra. Solutions (0.1M) in D_2O were measured at 300 K (internal acetone, 2.22 p.p.m.; DOH signal at 4.75 p.p.m.). A spectral width of 5 kHz, using 32 kbytes of computer memory (giving a digital resolution of 0.3 Hz/pt.)

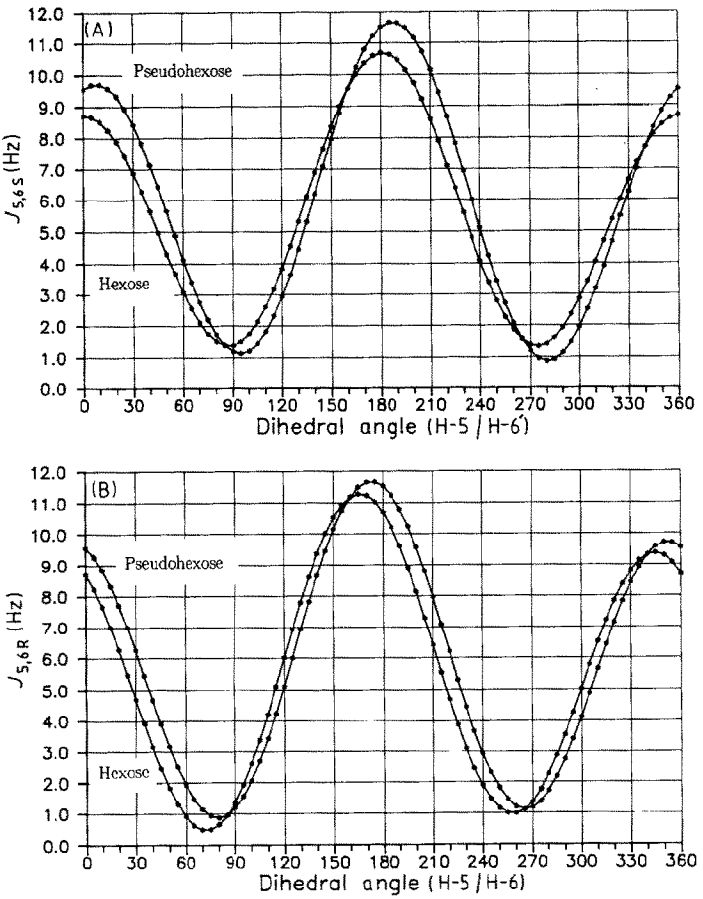

Fig. 1. (A) Angular dependence of the vicinal coupling constants $J_{5,6S}$ based on the Altona and Haasnoot approach[8] for hexoses and pseudohexoses, respectively. (B) Similar plot for the coupling constants $J_{5,6R}$.

was used, together with pulse angles of 90° (10 μs). COSY-90 experiments[6] were performed by using Bruker standard software. The ^{13}C-n.m.r. spectra were obtained on the same spectrometer operated at 125.7 MHz at 300 K (internal 1,4-dioxane, 67.4 p.p.m.). A spectral width of 25 kHz, using 64 kbytes of computer memory (giving a digital resolution of 0.8 Hz/pt.) was used, together with a pulse angle of 53° (90° = 8.5 μs). ^{13}C–^{1}H correlation experiments were made by using CHORTLE experiments[7].

DL-*(1,2/3,4,5)-2-Acetamido-5-(hydroxymethyl)-1,3,4-cyclohexanetriol (pseudo-2-acetamido-2-deoxy-α-*DL-*galactopyranose)* (**1**). — The *N*-acetyltetra-*O*-acetyl derivative[1,2] (120 mg, 0.31 mmol) was treated with M methanolic sodium methoxide for 1 h at room temperature, and then the solution was made neutral with Amber-

lite IR 120B (H^+) resin and evaporated. The residue (65 mg) crystallized from ethanol, to give **1** (37 mg, 54%) as prisms, m.p. 180–181°.

Anal. Calc. for $C_9H_{17}NO_5$: C, 49.31; H, 7.82; N, 6.39. Found: C, 49.03; H, 7.69; N, 6.17.

DL-*(1,3,4,5/2)-2-Acetamido-5-(hydroxymethyl)-1,3,4-cyclohexanetriol (pseudo-2-acetamido-2-deoxy-β-*DL-*galactopyranose)* (**2**). — The 4,7-*O*-isopropylidene derivative[2,*] (65 mg, 0.25 mmol) of **2** was treated with aqueous 80% acetic acid (3 mL) for 4.5 h at 95°, and the solution then cooled and evaporated. The resulting crystals (55 mg) were recrystallized from ethanol, to give **2** (31 mg, 56%) as hygroscopic crystals, m.p. 105–113°.

Anal. Calc. for $C_9H_{17}NO_5 \cdot 0.75\ H_2O$: C, 46.44; H, 8.01; N, 6.02. Found: C, 46.80; H, 7.58; N, 6.01.

DL-*(1,2,4/3,5)-2-Acetamido-5-(hydroxymethyl)-1,3,4-cyclohexanetriol (pseudo-2-acetamido-2-deoxy-α-*DL-*glucopyranose)* (**3**). — The *N*-acetyltetra-*O*-acetyl derivative[2,4] (0.31 g, 0.80 mmol) was *O*-deacetylated as described for **1**, and the product (168 mg) crystallized from chloroform–methanol, to give **3** (0.12 g, 67%) as crystals, m.p. 200–202°.

Anal. Calc. for $C_9H_{17}NO_5$: C, 49.31; H, 7.82; N, 6.39. Found: C, 49.09; H, 7.61; N, 6.54.

DL-*(1,3,5/2,4)-2-Acetamido-5-(hydroxymethyl)-1,3,4-cyclohexanetriol (pseudo-2-acetamido-2-deoxy-β-*DL-*glucopyranose)* (**4**). — The *N*-acetyltetra-*O*-acetyl derivative[2] (250 mg, 0.65 mmol) of **4** was *O*-deacetylated with methanolic sodium methoxide to give a crystalline product (131 mg, 93%). Recrystallization from ethanol afforded **4** (45 mg, 32%) as prisms, m.p. 226–227.5°.

Anal. Calc. for $C_9H_{17}NO_5$: C, 49.31; H, 7.82; N, 6.39. Found: C, 49.05; H, 7.42; N, 6.21.

DL-*(1,4/2,3,5)-2-Acetamido-5-(hydroxymethyl)-1,3,4-cyclohexanetriol (pseudo-2-acetamido-2-deoxy-α-*DL-*mannopyranose)* (**5**). — The *N*-acetyltetra-*O*-acetyl derivative[2] (149 mg, 0.39 mmol) of **5** was *O*-deacetylated with methanolic sodium methoxide and the product (76 mg) was purified by chromatography on a column of silica gel with 8:1 chloroform–methanol, to give **5** (50 mg, 59%) as a syrup.

Anal. Calc. for $C_9H_{17}NO_5 \cdot H_2O$: C, 45.56; H, 8.07; N, 5.90. Found: C, 45.20; H, 7.72; N, 5.60.

DL-*(1,2,3,5/4)-2-Acetamido-5-(hydroxymethyl)-1,3,4-cyclohexanetriol (pseudo-2-acetamido-2-deoxy-β-*DL-*mannopyranose)* (**6**). — The *N*-acetyltetra-*O*-acetyl derivative[5] (70 mg, 0.18 mmol) of **6** was *O*-deacetylated with methanolic sodium methoxide, and the syrupy product was purified by chromatography on a column of silica gel with 3:1 chloroform–methanol, to give **6** (31 mg, 78%) as a syrup.

Anal. Calc. for $C_9H_{17}NO_5 \cdot 1.5\ H_2O$: C, 43.90; H, 8.19; N, 5.69. Found: C, 43.77; H, 7.90; N, 5.47.

*Cyclitol numbering. Side chain is C-7.

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