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Note

Conformation of 3-*O*-β-D-galactopyranosyl-L-arabinose and a comparison with its α-linked isomer*

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We have reported[1] the complete assignment of the ^{1}H- and ^{13}C-n.m.r. spectra of 3-*O*-α-D-galactopyranosyl-L-arabinose (**1**), isolated from the corm-sac polysaccharide obtained from *Watsonia pyramidata*[2]. This disaccharide moiety is a constituent of gum exudates from numerous plant sources[3,4], and the corresponding β-isomer (**2**) links acidic arabinogalactan assemblies to D-mannose residues in polysaccharides of the mannoglucuronoglycan type[3]. The 3-*O*-β-D-galactopyranosyl-α,β-L-arabinose (**2**) used in the present n.m.r. study was isolated after partial hydrolysis of the exudate from *Encephalartos longifolius* cones[5,6].

The assignment of the ^{1}H- and ^{13}C-n.m.r. spectra of **2** were made for solutions in D_2O, using 2D COSY and HETCOR spectra as described[7]. In addition, the *J*-resolved ^{1}H-n.m.r. spectra were recorded in order to aid the assignment of ^{1}H signals. The assignments are given in Table I; the resonances for C-1′ and C-5′ (Gal moiety) were twinned[1] because of the presence of α and β forms of the L-arabinose moiety. That the Gal residue is β-D follows from the ^{1}H and ^{13}C chemical shifts. Table II lists the $\triangle\delta$ values for the ^{13}C resonances of the monosaccharide units in **2** relative to the corresponding resonances of 3-*O*-β-D-galactopyranosyl-D-galactose and of L-arabinose. The C-3 (Ara moiety) was deshielded by 9.2 and 9.8 p.p.m. for the α and β anomers, respectively, providing thereby the presence of a (1→3) linkage, whereas C-2 was shielded by 1.2 and 1.3 p.p.m., respectively. The latter shielding may be caused[8] by hydrogen bonding between the Ara hydroxyl groups and the Gal ring oxygen. On comparing the chemical shift data for the Ara moiety in **2** with those for **1**, C-2 in **1** experienced greater (2.0 p.p.m.) shielding and C-3 lesser (4.2 and 5.2 p.p.m.) deshielding for both the α and β forms. Additionally, there was a shielding effect of 4 p.p.m. at C-4 in **1** which was not observed for **2**. However, on comparing the chemical shifts of the resonances of C-2,3,4 (Gal moiety) of α-Glc-(1→3)-Gal and β-Gal-(1→3)-Gal[9] with those for **1** and **2**, it is apparent that the differences can be reconciled simply on the

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TABLE I

^{1}H- and ^{13}C-n.m.r. assignments for 3-*O*-β-D-galactopyranosyl-α,β-L-arabinose (δ in p.p.m.)

Gal Moiety			*Ara Moiety*		
^{13}C		^1H	^{13}C		^1H
104.95	1′α	4.60[a]	97.24	1α	4.55[b]
104.85	1′β		93.26	1β	5.24[c]
71.83	2′	3.52–3.60	71.74	2α	3.55–3.63
		3.57[d]			
			68.24	2β	3.92–3.96
73.29	3′	3.58–3.67	82.69	3α	3.76–3.80
			79.26	3β	3.98
69.32	4′	3.85–3.94	69.05	4α	4.15–4.20
		3.91[d]			4.17[d]
			69.21	4β	4.20–4.25
					4.22[d]
75.77	5′α		66.56	5α	3.82–3.86
		3.62–3.68			3.84[d]
75.74	5′β		62.74	5β	–[e]
61.68	6′	3.70–3.73			
		3.72[d]			

[a] $J_{1,2}$ 6.77 Hz. [b] $J_{1,2}$ 7.75 Hz, [c] $J_{1,2}$ 2.50 Hz. [d] From *J*-resolved experiment. [e] There was no visible correlation in the HETCOR experiment.

TABLE II

Δδ Values[a] for the ^{13}C resonances of **1** and **2**

Resonance	*β-Gal-(1→3)-Ara* (**2**)			*α-Gal-(1→3)-Ara* (**1**)	
	β-Gal[b]	*α-Ara*[c]	*β-Ara*[c]	*α-Ara*[c]	*β-Ara*[c]
C-1	−0.2	−0.8	−0.1		
C-2	−0.2	−1.2	−1.3	−2.0	−2.0
C-3	−0.1	+9.2	+9.8	+4.2	+5.2
C-4	−0.1	−0.5	−0.3	~ −4	~ −4
C-5	−0.1	−0.6	−0.7		
C-6	−0.1				

[a] Negative values represent shielding. [b] Reference: nonreducing moiety in β-Gal-(1→3)-Gal. [c] Reference: Ara.

basis[10] of the glycosyl substituent being α or β (Table III). It is assumed that the influences of the Galα and Glcα substituents are similar.

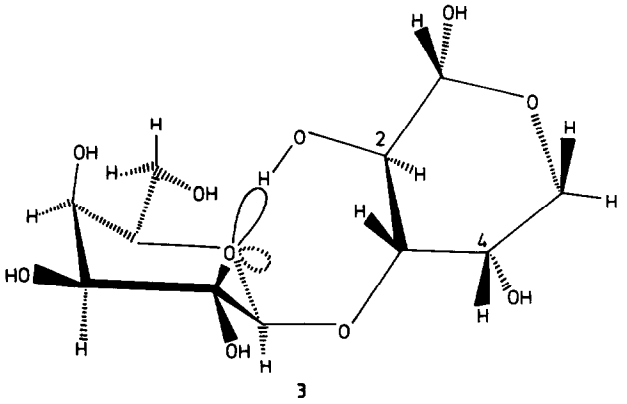
Glycosylation at C-3 of Ara in **2** appears to anchor the Ara moiety in the $^4C_1$ conformation as in **1**[1]. In the conformation shown in **3**, there would be favourable intramolecular hydrogen bonding between the ring oxygen (O-5) of Gal and the equatorial HO-2 of Ara. This inference conforms with the upfield shift of the C-2

TABLE III

Comparison of the ^{13}C chemical shift data for β-Gal-(1→3)-α-Ara with data in the literature[1,9]

Reducing moiety	*α-Glc-(1→3)-β-Gal*	*β-Gal-(1→3)-β-Gal*	*β-Gal-(1→3)-α-Ara*	*β-Gal-(1→3)-α-Ara*
			(α-1)	(α-2)
C-1	97.7	97.0	97.19	97.24
C-2	71.5	71.8	70.94	71.74
C-3	78.8	83.3	77.71	82.69
C-4	66.3	69.4	65.44	69.05
C-5	76.1	75.6	66.62	66.56
C-6	62.2	61.8	–	–

resonance of the Ara moiety[8]. Glycosylation of the equatorial HO-3 should cause an upfield shift in the resonance of the neighbouring skeletal C atom if it carried an equatorial H, due to interaction of H-4 of Ara and H-1′ of Gal when in the *γ-gauche* conformation[11]. However, there was no change (see Table II) in chemical shift of the Ara C-4 resonance relative to that of C-4 in unsubstituted Ara. Therefore, the conformation of **2** shown in **3** accords with the results obtained.



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For **1**, there is the possibility of hydrogen bonding of both HO-4 and HO-2 with the equatorial lobe of the ring oxygen of the Gal residue. For **2**, on the other hand, the only possibility of hydrogen bonding is of HO-2 with the axial lobe of the ring oxygen, which gives the "chair/chair/chair" conformation shown in **3**.

EXPERIMENTAL

Gum exudate from *Encephalartos longifolius* cones was heated at pH 2 for 13 h on a boiling water bath, and the degraded polysaccharide was then further hydrolysed in 0.25M H_2SO_4 for 1 h at 100°. The neutralised ($BaCO_3$) product was dialysed, and the

dialysate concentrated to a syrup. The syrup was extracted with boiling EtOH, and the soluble mixture of neutral sugars and oligosaccharides separated into fractions on charcoal–Celite[12] by elution with increasing concentrations of EtOH in water. After removal of monosaccharides, elution with 5% EtOH afforded 3-*O*-*β*-D-galactopyranosyl-L-arabinose (**2**), which had $[\alpha]_D$ +42° (*c* 0.3) and mobilities in p.c. (acidic, neutral, and basic solvent mixtures) similar to, but not identical with, those of **1**. Hydrolysis of **2** and characterisation of the resulting sugars by g.l.c. of their alditol acetates revealed equal molar proportions of Gal and Ara. Borohydride reduction of **2** and then hydrolysis gave Gal as the only reducing sugar.

A solution of **2** in D_2O was freeze-dried (three times) and the ^1H- and ^{13}C-n.m.r. spectra were recorded for solutions in D_2O (internal acetone, δ 2.21 for ^1H and δ 31.0 for ^{13}C), with a Varian VXR-200 spectrometer[7].

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