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Note

Studies on oligosaccharide chemistry. Part I. The synthesis of benzyl 2,4-di-*O*-benzyl-β-D-galactopyranoside*

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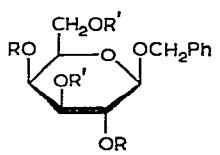
Studies of the structure of antigenic determinants of soluble blood groups have led to the isolation of many oligosaccharides from glycoproteins of blood-group substances[1,2]. A proposed structure for the carbohydrate moiety of these glycoproteins locates a β-D-galactose residue at the main branch-point[3,4]. A partial basis for this proposal was the isolation[5] of the trisaccharide *O*-(2-acetamido-2-deoxy-β-D-glucopyranosyl)-(1→3)-[*O*-(2-acetamido-2-deoxy-β-D-glucopyranosyl)-(1→6)]-D-galactose from blood-group glycoproteins isolated from hog-gastric mucin and of the more complex oligosaccharide *O*-β-D-galactopyranosyl-(1→3)-*O*-(2-acetamido-2-deoxy-β-D-glucopyranosyl)-(1→3)-[*O*-β-D-galactopyranosyl-(1→4)-*O*-(2-acetamido-2-deoxy-β-D-glucopyranosyl)-(1→6)]-D-galactose from human, ovarian-cyst material[3]. Although these two saccharides have been characterized chemically, they have not been synthesized.

As a possible starting material for the preparation of the trisaccharide, we synthesized benzyl 2,4-di-*O*-benzyl-β-D-galactopyranoside (**4**), properly substituted for condensation at positions 3 and 6.



1 R = R' = H
2 R = H, R' = Ms
3 R = CH_2Ph, R' = Ms
4 R = CH_2Ph, R' = H

A compound analogous to **4**, methyl 2,4-di-*O*-methyl-β-D-galactopyranoside, has been prepared by Jeanloz[6] and by Westwood *et al.*[7]. For the synthesis of an unsubstituted trisaccharide, it is necessary, however, to use intermediates having easily removed blocking groups. Therefore, we selected the benzyl derivative **4**, since both benzyl ethers and glycosides are easily reduced under mild conditions[8].

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Our procedures for the synthesis of **4** followed, in general, the procedures used by Westwood *et al.*[7] for the methyl derivative. Benzyl β-D-galactopyranoside[9] (**1**) was mesylated at −40° to give a mixture of products. The main product was isolated by column chromatography and shown to be benzyl 3,6-di-*O*-mesyl-β-D-galactopyranoside (**2**); it was not oxidized by periodic acid, as expected for a vicinal diol[10], under conditions which led to the oxidation of D-galactose and to the gradual oxidation of β-D-galactose pentaacetate. The i.r. spectrum showed aromatic and mesyl absorptions and the n.m.r. spectrum gave the expected ratio of methyl to aromatic protons.

The benzylation procedure described by Ballou and Pizer[11] was used for the conversion of **2** into benzyl 2,4-di-*O*-benzyl-3,6-di-*O*-mesyl-β-D-galactopyranoside (**3**), except that the reaction mixture was protected from light and the product was purified by chromatography on a silica gel column. The i.r. spectrum showed aromatic mesyl absorptions, and the correct ratio of methyl to aromatic protons was confirmed by the n.m.r. spectrum.

Lithium aluminium hydride reduction of **3** in tetrahydrofuran gave a mixture of products from which benzyl 2,4-di-*O*-benzyl-β-D-galactopyranoside (**4**) was separated by crystallization from benzene. The hydroxyl protons show an i.r. absorption at 3535 and 3440–3240 cm^{-1}, and the n.m.r. absorption corresponds to two hydroxyl protons which disappeared upon addition of deuterium oxide.

EXPERIMENTAL

General methods. — Solvents were evaporated under diminished pressure with a bath temperature below 50°. Melting points were determined with a Büchi melting-point apparatus and are uncorrected. Specific rotations were measured with a Roussel and Jouan electronic, digital, micropolarimeter. I.r. spectra were measured on solid samples, dispersed in KBr, with a Perkin–Elmer model 257 spectrometer. N.m.r. spectra were recorded with a Jeol-C-60-H n.m.r. spectrometer at 60 MHz, with chloroform-*d* as solvent and tetramethylsilane as internal standard. Microanalyses were performed by the Laboratoire Central de Micro-Analyse du C.N.R.S.. Each compound was homogeneous when examined by t.l.c. on precoated plates of silica gel (E. Merck, Darmstadt, Germany) 0.25-mm of thickness. Silica gel Merck (70–325 mesh; E. Merck, Darmstadt, Germany) was used for column chromatography.

Benzyl 3,6-di-O-mesyl-β-D-galactopyranoside (**2**). — The selective mesylation procedures of Chalk *et al.*[12] for mesylation of methyl glycosides was used for the preparation of this compound. A solution of benzyl β-D-galactopyranoside[9] (4.72 g) in dry pyridine (35 ml) was cooled in a dry ice–acetone bath to −40°. Mesyl chloride (2.76 ml) was added dropwise during 1.5 h while the bath was maintained at −40°. The solution was stored for 24 h at −20° and then pyridine hydrochloride was removed by filtration. The solution was kept for 24 h at room temperature, and most of the pyridine was removed at 30° under reduced pressure. The resulting, thick syrup was fractionated on a column of silica gel (350 g) with 9:1 chloroform–methanol to

give a syrup (4.33 g, 57%), $[\alpha]_D^{20}$ −28° (*c* 1, chloroform); i.r. data: ν_{max}^{KBr} 1353 (–SO_2–), 750, 703 cm^{-1} (Ph); n.m.r. data (chloroform-*d*): δ 3.05 (2 Me) and 7.32 (Ph); t.l.c. in 9:1 chloroform–methanol: R_F 0.42.

Anal. Calc. for $C_{15}H_{22}O_{10}S_2$: C, 42.30; H, 5.20; S, 15.03. Found: C, 41.59; H, 5.07; S, 14.77.

Compound **2** was identified as the 3,6-di-*O*-mesyl derivative by its resistance to oxidation with periodic acid under the conditions described by Vogel[10].

Under the same conditions, D-galactose was rapidly and β-D-galactose pentaacetate gradually oxidized.

*Benzyl 2,4-di-*O*-benzyl-3,6-di-*O*-mesyl-β-*D*-galactopyranoside* (**3**). — Benzylation was performed with a modification of the procedure described by Ballou and Pizer[11]. A solution of **2** (4.12 g) in dry *N,N*-dimethylformamide (60 ml) was cooled in an ice bath and benzyl bromide (30 ml) was added. The solution was protected from light and silver oxide (20 g) was added during 1 h. The mixture was stirred for 20 h at room temperature. The salts were filtered off, and the filtrate was concentrated in a high vacuum to a thick syrup. The syrup was dissolved in 39:1 chloroform–methanol and filtered on silica gel (100 g) to remove remaining salts. The filtrate was evaporated and the residue was chromatographed on a column of silica gel with 39:1 chloroform–methanol to give a pure fraction. Crystallization from methanol gave 1.51 g (26%), m.p. 104–106°; $[\alpha]_D^{20}$ −33° (*c* 1, chloroform); i.r. data: ν_{max}^{KBr} 1350 (–SO_2–), 738 and 699 cm^{-1} (Ph); n.m.r. data (chloroform-*d*): δ 2.91 (2 Me) and 7.46 (3 Ph); t.l.c. in 39:1 chloroform–methanol: R_F 0.95.

Anal. Calc. for $C_{29}H_{34}O_{10}S_2$: C, 57.40; H, 5.56. Found: C, 57.30; H, 5.60.

*Benzyl-2,4-di-*O*-benzyl-β-*D*-galactopyranoside* (**4**). — The mesyl groups were reduced according to the procedures described by Westwood *et al.*[7]. A solution of **3** (1.36 g) in dry tetrahydrofuran (50 ml) was stirred with lithium aluminum hydride (0.60 g) for 24 h at 50°. The solution was cooled in an ice bath, and the excess hydride was destroyed by addition of aqueous tetrahydrofuran. After filtration, the residue was washed with tetrahydrofuran and the filtrate and washings were combined and concentrated to give a partially crystalline residue having an odor of sulfenic acid. Crystallization from benzene gave 0.54 g (53%), m.p. 111–113°; $[\alpha]_D^{20}$ −22° (*c* 1, chloroform); i.r. data: ν_{max}^{KBr} 3535, 3440–3240 (OH), 734, 697 cm^{-1} (Ph); n.m.r. data (chloroform-*d*): δ 1.80 and 2.45 (2 OH); t.l.c. in 19:1 chloroform–methanol: R_F 0.70.

Anal. Calc. for $C_{21}H_{30}O_6$: C, 71.98; H, 6.71; O, 21.31. Found: C, 71.83; H, 6.74; O, 21.61.

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