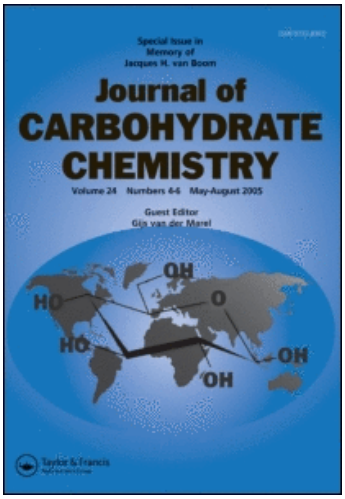

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Synthesis of Methyl 4-*O*-(β-D-Galactopyranosyl)-3-*O*(β-D-Glucopyranosyluronic Acid)-α-*L*-Rhamnopyranoside

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SYNTHESIS OF METHYL 4-O-(β-D-GALACTOPYRANOSYL)-3-O-(β-D-GLUCOPYRANOSYLURONIC ACID)-α-L-RHAMNOPYRANOSIDE

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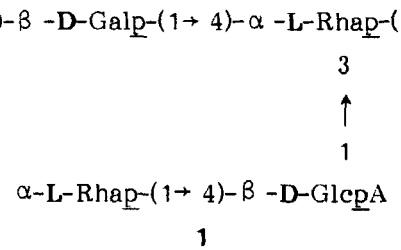
ABSTRACT

Methyl 4-O-(2,3,4,6-tetra-O-acetyl-β-D-galactopyranosyl)-2,3-O-isopropylidene-α-L-rhamnopyranoside was obtained by condensing methyl 2,3-O-isopropylidene-α-L-rhamnopyranoside with acetobromogalactose. This compound was benzylated, the isopropylidene group was removed and the product was then partially benzylated to give methyl 2-O-benzyl-4-O-(2,3,4,6-tetra-O-benzyl-β-D-galactopyranosyl)-α-L-rhamnopyranoside. Koenigs Knorr condensation of this compound with methyl (2,3,4-tri-O-acetyl-α-D-glucopyranosyl bromide) uronate gave a trisaccharide derivative which after deprotection gave the title trisaccharide.

INTRODUCTION

The structure of the repeating unit (1) of capsular polysaccharide from Klebsiella Type K-47 was established by Lindberg and his co-workers.[1] Recently Fugedi[2] has synthesized 4-O-(α-L-rhamnopyranosyl)-D-glucopyranosyluronic acid, which is a part of the

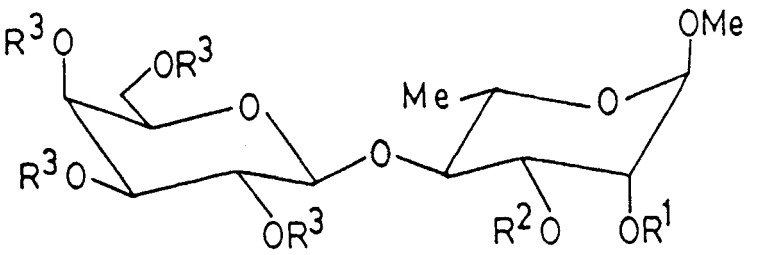
repeating unit of K-47 antigen. As a part of our programme to synthesize suspected immunodominant fragments related to different bacterial antigens, we wish to report the synthesis of 4-O-(β-D-galactopyranosyl)-3-O-(β-D-glucopyranosyluronic acid)-α-L-rhamnopyranoside (**10**), another part of the repeating unit of this same antigen.



RESULTS AND DISCUSSION

Methyl α-L-rhamnopyranoside (**2**), obtained by Fischer glycosidation[3] of L-rhamnose, was treated with 2,2-dimethoxypropane and a catalytic amount of toluene-p-sulfonic acid in dry N,N-dimethyl formamide giving 2,3-O-isopropylidene-α-L-rhamnopyranoside[4] (**3**). Condensation of **3** with 2,3,4,6-tetra-O-acetyl-α-D-galactopyranosyl bromide in the presence of mercury (II) cyanide in acetonitrile[5] gave methyl 4-O-(2,3,4,6-tetra-O-acetyl-β-D-galactopyranosyl)-2,3-O-isopro pylidene-α-L-rhamnopyranoside (**4**) in 76% yield, as fine crystals. Removal of the O-isopropylidene group with 85%acetic acid followed by de-O-acetylation with sodium methoxide gave methyl 4-O-(β-D-galactopyranosyl)-α-L-rhamnopyranoside (**5**) which was characterised in the usual way.

Compound **4** was benzylated[6] to give methyl 4-O-(2,3,4,6-tetra-O-benzyl-β-D-galactopyranosyl)-2,3-O-isopropylidene-α-L-rhamnopyranoside (**6**). The isopropylidene group of **6** was removed, and the resulting methyl 4-O-(2,3,4,6-tetra-O-benzyl-β-D-galactopyranosyl)-α-L-rhamnopyranoside (**7**) was partially benzylated by the phase transfer method[7] to give methyl 2-O-benzyl-4-O-(2,3,4,6-tetra-



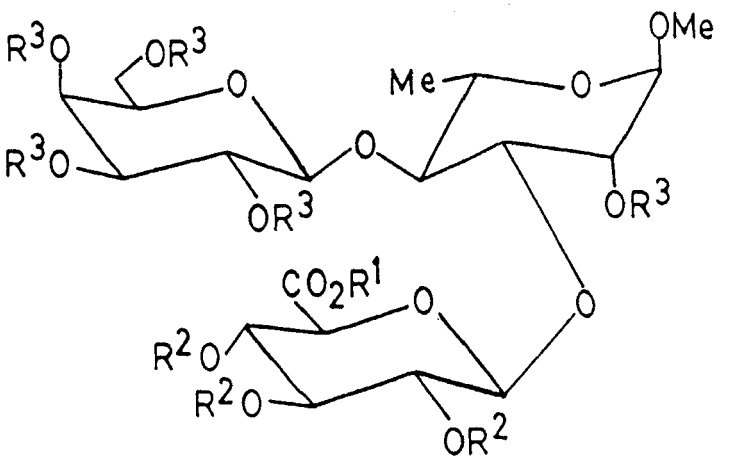
4 $R^1 R^2 = Me_2C$, $R^3 = Ac$

5 $R^1 = R^2 = R^3 = H$

6 $R^1 R^2 = Me_2C$, $R^3 = Bn$

7 $R^1 = R^2 = H$, $R^3 = Bn$

8 $R^1 = R^3 = Bn$, $R^2 = H$



9 $R^1 = Me$, $R^2 = Ac$, $R^3 = Bn$

10 $R^1 = R^2 = R^3 = H$

O-benzyl- β -D-galactopyranosyl)- α -L-rhamnopyranoside (**8**) in 68% yield. It has been reported[7] that during partial benzylation of carbohydrate derivatives by the phase transfer method, the more acidic hydroxyl group is substituted in major amount. Compound **8** was also characterized by methylation analysis[9] when rhamnose showed up as 3-O-methyl derivative.

The hydroxy compound (**8**) was allowed to react with methyl (2,3,4-tri-O-acetyl- α -D-glucopyranosyl bromide) uronate in the presence of silver triflate[5,8] and tetra-N-methyl urea in dichloromethane when methyl 2-O-benzyl- β -D-galactopyranosyl)-3-O-(methyl [2,3,4-tri-O-acetyl- β -D-glucopyranosyl] uronate)- α -L-rhamnopyranoside (**9**) was obtained as crystals (52% yield). Hydrogenolysis of **9** with 10% Pd/C followed by deacetylation and de-esterification of the product gave methyl 4-O-(β -D-galactopyranosyl)-3-O-(β -D-glucopyranosyluronic acid)- α -L-rhamnopyranoside (**10**). Compound **10** was characterised by acid hydrolysis and methylation analysis.

EXPERIMENTAL

General methods. All reactions were monitored by TLC on Silica Gel G (Merck). Column chromatography was performed on Silica Gel 60 (Merck). GLC was carried out by using a Hewlett-Packard Model 5730A instrument fitted with a flame ionization detector and a glass column (1.83m x 6mm) packed with 3% ECNSS-M on Gas Chrom Q(100-120 mesh). The chromatography was performed at 180 °C for neutral sugars and at 170 °C for methylated sugars by converting the sugars into their alditol acetates.[9]

Melting points were determined on a Fisher-Johns apparatus and are uncorrected. Optical rotations were measured with a Perkin-Elmer Model 241MC Spectropolarimeter. NMR spectra were taken with a Varian XL-200 spectrometer using TMS as an internal standard and deuterochloroform as the solvent.

Methyl 4-O-(2,3,4,6-tetra-O-acetyl-β -D-galactopyranosyl)-2,3-O-isopropylidene-α -L-rhamnopyranoside (4). Methyl 2,3,-O-isopropylidene-α -L-rhamnopyranoside (**3**) (5g; 22.9 mmol), prepared from methyl α-L-rhamnopyranoside[3] (**2**), in dry acetonitrile was stirred with acetobromogalactose[10] (10.2 g; 25 mmol), mercury (II) cyanide (6.6 g; 26 mmol), and molecular sieve (3Å) for 24 h at r.t. The mixture was diluted with $CHCl_3$(500 mL) and filtered through a bed of Celite. The filtrate was washed successively with 5% potassium iodide solution, saturated sodium bicarbonate solution, and water. The organic layer was dried and concentrated to dryness. The compound was purified by column chromatography using benzene-ether (3:1) as solvent. Crystallisation from ethanol gave pure **4** (9.6 g; 76%) having $[\alpha]_D^{24}$ -23.5° (c 3.2, $CHCl_3$) and mp 182-184 °C. ^1H NMR data: δ 1.19 (d, 3H, C_5-CH_3, J 6Hz), 1.35 and 1.54 [2s, 6H, $(CH_3)_2$ C], 2.00, 2.05, 2.09 and 2.16 (4s, 12H, 4OAc), 3.38 (s, 3H, OCH_3), 4.92 (d, 1H, J 7.5 Hz, H-1′), 5.10 (d, 1H, J 3.6 Hz, H-1).

Anal. Calcd for $C_{24}H_{36}O_{14}$: C, 52.55; H, 6.62. Found : C, 52.17; H, 6.78.

Methyl 2-O-benzyl-4-O-(2,3,4,6-tetra-O-benzyl-D-galactopyranosyl)-α -L-rhamnopyranoside (8). Methyl 4-O-(2,3,4,6-tetra-O-benzyl-β -D-galactopyranosyl)-α -L-rhamnopyranoside (**7**) (3 g; 4.28 mmol) obtained from **4**(6 g) by benzylation[6] followed by treatment with 85% acetic acid, was dissolved in dichloromethane (50 mL). Benzyl bromide (0.75 mL; 5.25 mmol), tetrabutylammonium bromide (0.35 g; 1.8 mmol), and 10% aqueous sodium hydroxide solution[7] (6 mL) were then added to it and the mixture was stirred vigorously for 2 days. The organic layer was washed with water, dried (Na_2SO_4), and concentrated to a syrup. Column chromatography (benzene-ether 5:1) of the product gave **8** (2.3 g; 68%) as syrup together with the 3-O-benzyl derivative (15%) and 2,3-di-O-benzyl derivative (5%). Compound **8** had $[\alpha]_D^{24}$ - 12.8°(c 1.8, $CHCl_3$). ^1H NMR data : δ 1.36 (d, 3H, J 6Hz, C_5-CH_3), 3.33 (s, 3H, OCH_3), 4.62 (d, 1H, J 6Hz, H-1′), 4.46-4.9 (2s, 10H, 5 $PhCH_2$), 4.96 (d, 1H, H-1), 7.32-7.36 (m, 25H, 5Ph).

Anal. Calcd for $C_{48}H_{54}O_{10}$: C, 72.89; H, 6.88. Found : C, 72.36; H, 6.98.

Methyl 2-O-benzyl-4-O-(2,3,4,6-tetra-O-benzyl- β -D-galacto-pyranosyl)-3-O-(methyl[2,3,4-tri-O-acetyl-β -D-glucopyranosyl] uronate) - α -L-rhamnopyranoside (9). A mixture of **8** (1.5 g; 1.89 mmol), dichloromethane (25 mL), 4Å molecular sieve (2 g) and tetra-N-methyl urea[8] (0.3 mL) was stirred at room temperature for 30 min. Methyl (2,3,4-tri-O-acetyl- α-D-glucopyranosyl bromide) uronate (0.8 g; 2 mmol) was added and the mixture was cooled to -10 °C in an alcohol bath. Solid silver triflate (0.52 g, 2 mmol) was then added. Stirring was continued for 20 h at -10 °C in the dark. The mixture was then filtered through a bed of Celite and the filtrate was washed with water, saturated $NaHCO_3$ solution and, again, water. The organic layer was dried (Na_2SO_4) and concentrated to a syrup. The product was purified by column chromatography using benzene-ether (6:1) as the eluent. Crystallisation from ethanol-ethyl acetate gave pure **9** (1.1 g, 52%) $[\alpha]_D^{24}$ - 36.6° (c 1.1, $CHCl_3$), mp 180-182 °C. ^1H NMR data : δ 1.26 (d, 3H, 6Hz, C_5-CH_3), 1.76-1.96 (3s, 9H, 3 OAc), 3.28 (s, 3H, OCH_3), 3.61 (s, 3H, $COOCH_3$), 4.4-4.7 (2s, 10H, 5PhCH_2), 4.46 (d, 1H, J 8Hz, H-1″), 4.90 (d, 1H, J 7.5Hz, H-1′), 4.95 (d, 1H, H-1), 7.24-7.32 (m, 25H, 5Ph).

Anal. Calcd for $C_{61}H_{70}O_{19}$: C, 66.17; H, 6.37. Found : C, 65.96; H, 6.51.

Methyl 4-O-(β -D-galactopyranosyl)-3-O-(β -D-glucopyranosyl-uronic acid)- α -L-rhamnopyranoside (10). A mixture of **9** (300 mg) and Pd/C (200 mg) in ethyl acetate (10 mL) was stirred under hydrogen at room temperature for 24 h. The mixture was filtered through a bed of Celite and the filtrate was concentrated to a syrup. The syrup was stirred with 0.1 M sodium methoxide in methanol (10 mL) for 3 h at r.t. A few drops of water were then added. The solution was allowed to stand for 1 h, neutralized with Amberlite IR-120 (H^+) ion-exchange resin and concentrated to dryness to give **10** (132 mg; 94%) $[\alpha]_D^{24}$ - 16.8° (c 1.2, water).

Anal. Calcd for $C_{20}H_{35}O_{16}$: C, 46.06; H, 6.77. Found : C, 45.91; H, 6.85.

Methylation analysis of Compound 8. To a solution of compound **8** (100 mg) in N,N-dimethylformamide (5 mL) were added silver oxide (1 g) and Drierite (1 g). The mixture was stirred for 30 min; methyl iodide (0.7 mL) was then added and stirring was continued for 24 h. Chloroform (25 mL) was added while the mixture was vigorously stirred. The precipitates formed were filtered off through a Celite bed, and the filtrate was concentrated to a syrup. The product was purified by column chromatography, using 9:1 benzene-ether as the solvent. The syrupy material was taken up in ethyl acetate and stirred with 10% Pd/C under hydrogen at room temperature for 24 h as described above. Alditol acetates were prepared[9] from the resulting monomethyl compound. Analysis by GLC showed peaks for galactose and 3-O-methyl rhamnose in the ratio 1:0.9 as identified by comparing with authentic samples.

Acid hydrolysis and methylation analysis of compounds 5 and 10. Compound **10** (5 mg) was hydrolysed with 2 M trifluoroacetic acid (1 mL) at 100 °C for 20 h. The acid was removed in vacuo by codistillation with water. Paper chromatography of the hydrolysate on Whatman DE81 paper with 3:1:1 ethyl acetate-acetic acid-water showed spots of rhamnose, galactose and glucuronic acid together with a slower moving spot of aldobiouronic acid.

Compounds **5** and **10** were methylated and alditol acetates were prepared as described above. In case of compound **5** analysis by GLC showed peaks of 2,3,4,6-tetra-O-methylgalactose and 2.3-di-O-methylrhamnose in the ratio of 1:1. Permethylated **10** on similar analysis gave 2,3,4,6-tetra-O-methylgalactose and 2-O-methylrhamnose in the ratio of 1:0.7.

REFERENCES

1. H. Bjorndal, B. Lindberg, J. Lonngren, K. -G. Rosell, and W. Nimmich, Carbohydr. Res., **27**, 373 (1973).

2. P. Fügedi, J. Carbohydr. Chem., **6**, 377 (1987).

3. E. Fischer, Ber., **28**, 1145 (1895); G. M. Bebault and G. G. S. Dutton, Can. J. Chem., **50**, 3373 (1972).

4. A. Hasegawa and H. G. Fletcher, Jr., Carbohydr. Res., **29**, 209 (1973); M. E. Evans and F. W. Parrish, Tet. Letts.,3805 (1966)

5. H. Paulsen, Angew. Chem. Int. Ed. Engl., **21**, 155 (1982) and references therein.

6. C. P. J. Glaudemans and H. G. Fletcher, Jr., J. Am. Chem. Soc., **87**, 4636 (1965).

7. P. J. Garegg, Acta. Chem. Scand., **17**, 1343 (1963); P. J. Garegg and T. Iversen, Carbohydr. Res., **50**, C12 (1976).

8. S. S. Rana, J. J. Barlow and K. L. Matta, Carbohydr. Res., **84**, 353 (1980).

9. A. K. Ray, A. Roy and N. Roy, Carbohydr. Res., **165**, 77 (1987).

10. P. G. Scheurer and F. Smith, J. Am. Chem. Soc., **76**, 3224 (1954).

11. H. G. Walker, Jr., M. Gee and R. M. McCready, J. Org. Chem., **27**, 2100 (1962).