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Preliminary communication

Synthesis of 8-(methoxycarbonyl)octyl glycosides of *O*-α-L-rhamnopyranosyl-(1→3)-*O*-α-L-rhamnopyranosyl-(1→3)-2-acetamido-2,6-dideoxy-D-glucopyranose; models for the antigen of *Pseudomonas aeruginosa* Fisher immunotype 5

DEREK HORTON and SOTH SAMRETH
Department of Chemistry, The Ohio State University, Columbus, Ohio 43210 (U.S.A.)

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The cell-surface lipopolysaccharide antigens of the seven Fisher immunotypes of *Pseudomonas aeruginosa* show major, qualitative differences in their polysaccharide chain-components[1]; trisaccharide repeating-units in this component have been established[2] for Types 1, 2, and 5. Use of these whole antigens as human vaccines[3] is complicated by the fact that their lipid A component is strongly immunogenic. The isolated polysaccharide chain-components have molecular weights[4] too low[5] for them to serve as effective antigens unless conjugated to a carrier of high molecular weight[6]. The present work was undertaken to provide a single repeat of the Type 5 sequence in the form of a long-chain glycoside in which the aglycon is presumed[7] to be non-immunogenic. The terminal methoxycarbonyl functionality should permit attachment of the trisaccharide hapten, by way of the derived[7] acyl azide, to such protein acceptors as human, or other mammalian, serum proteins, in order to furnish artificial antigens, and to suitable, functionalized solid supports that may be effective affinity-adsorbents for *P. aeruginosa* Type 5 antibodies.

8-(Methoxycarbonyl)octanol reacted with 2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy-α-D-glucopyranosyl chloride[8] in benzene containing $Hg(CN)_2$ at 25° to give 90% of the acetylated β-glycoside **1**, m.p. 102°, $[\alpha]_D$ −10° ($CHCl_3$), convertible[9] by $ZnCl_2$ and an excess of 8-(methoxycarbonyl)octanol at 130° (vacuum) into 60% of syrupy α-glycoside **3**, $[\alpha]_D^{25}$ +78° (MeOH). *O*-Deacetylation (Zemplén) of **1** gave ~100% of **9**, m.p. 156°, $[\alpha]_D$ −21° (MeOH), and similarly, **3** gave 80% of its *O*-deacetylated analog **4**, m.p. 139°, $[\alpha]_D$ +112° (MeOH). Benzylidenation of **2** with α,α-dimethoxytoluene at 25° (TsOH) gave 87% of the 4,6-benzylidene acetal[7] **5**, m.p. 222°, $[\alpha]_D$ −51° ($HCONMe_2$), and similar treatment of **4** gave 90% of the 4,6-benzylidene acetal **6**, m.p. 138°, $[\alpha]_D$ +61° (MeOH).

Methyl α-L-rhamnopyranoside underwent selective[10] glycosylation at O-3 by tri-*O*-acetyl-α-L-rhamnopyranosyl bromide, and the product obtained by acetylation and

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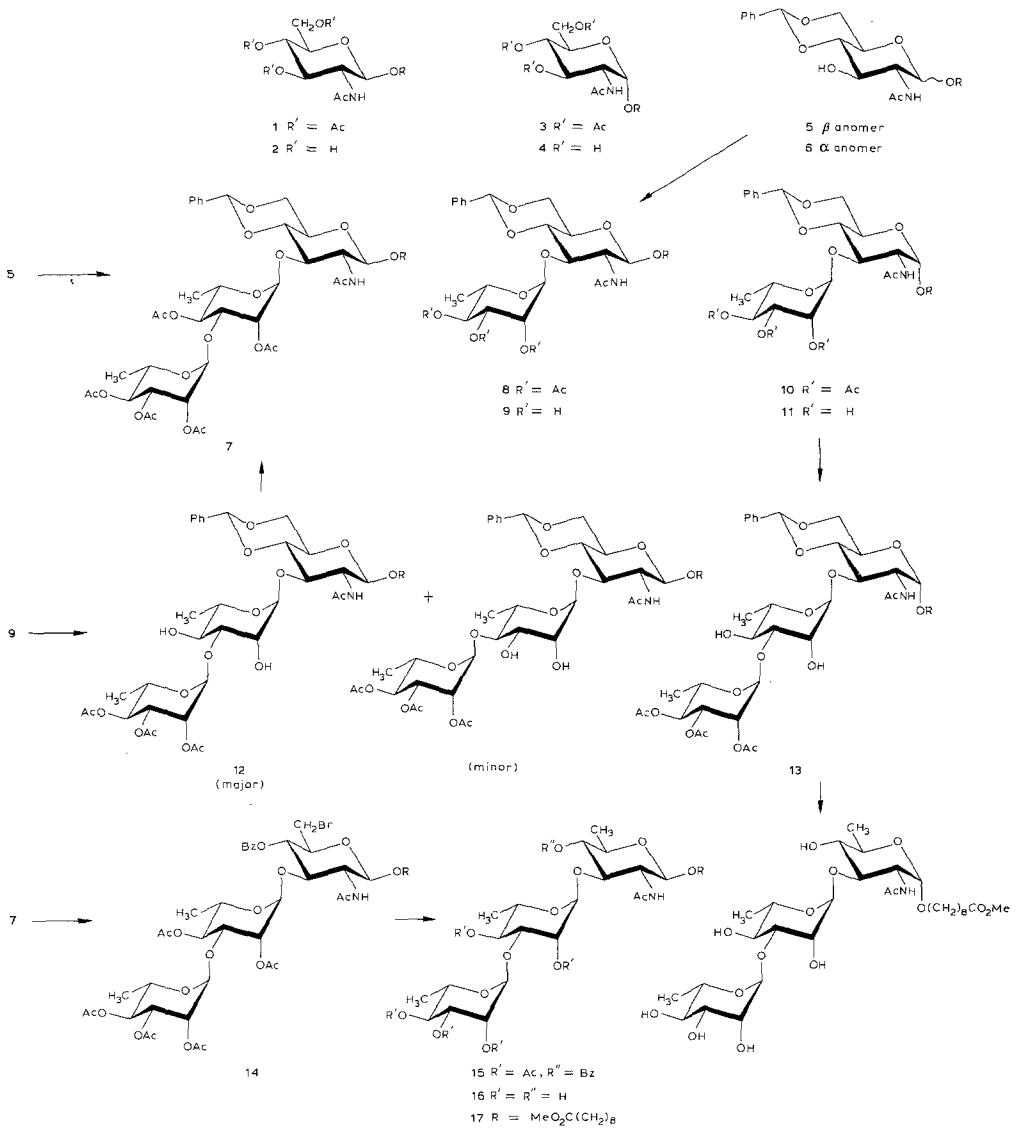

subsequent acetolysis with Ac_2O containing 1% of H_2SO_4 was treated with HBr–HOAc, to give 2,4-di-*O*-acetyl-3-*O*-(2,3,4-tri-*O*-acetyl-α-L-rhamnopyranosyl)-α-L-rhamnopyranosyl bromide[11]. The latter underwent reaction with **5** in benzene $[Hg(CN)_2]$ at ~25°, to give 8-(methoxycarbonyl)octyl 2-acetamido-4,6-*O*-benzylidene-2-deoxy-3-*O*-[2,4-di-*O*-acetyl-3-*O*-(2,3,4-tri-*O*-acetyl-α-L-rhamnopyranosyl)-α-L-rhamnopyranosyl]-β-D-glucopyranoside (**7**), isolated chromatographically homogeneous, but in rather low yield. In an alternative approach, glycoside **5** was condensed at 50° with tri-*O*-acetyl-α-L-rhamnopyranosyl bromide in 1:1 benzene–acetonitrile $[Hg(CN)_2]$ to give 90% of the disaccharide derivative[12] **8**, m.p. 142°, $[\alpha]_D$ -104° (MeOH), and the α precursor **6** likewise gave 90–100% of the syrupy disaccharide analog **10**, $[\alpha]_D$ +7° (MeOH).

Zemplén *O*-deacetylation of **8** gave 80% of 8-(methoxycarbonyl)octyl 2-acetamido-4,6-*O*-benzylidene-2-deoxy-3-*O*-α-L-rhamnopyranosyl-β-D-glucopyranoside (**9**), m.p. 192°, $[\alpha]_D$ −86° (MeOH), which underwent reaction with tri-*O*-acetyl-α-L-rhamnopyranosyl bromide in MeCN–$MeNO_2$ [$Hg(CN)_2$], to give 25% of the 3-*O*-substituted product (**12**), m.p. 123–124°, $[\alpha]_D$ −80° (MeOH) and 10% of the 4-*O*-substituted isomer, m.p. 119°, $[\alpha]_D$ −92° (MeOH). The latter reacted with periodate, and was converted by acetone and a trace of acid into an isopropylidene acetal (n.m.r.), whereas **12** did not react with periodate or acetone–H^+, and was converted by acetylation (100%) into trisaccharide derivative **7**, m.p. 119–120°, $[\alpha]_D$ −64° (MeOH), identical (by ^{1}H-n.m.r.) with that prepared previously.

O-Deacylation of **10** gave 96% of the corresponding triol **11**, m.p. 194°, $[\alpha]_D$ +4° (MeOH), which, on treatment with tri-*O*-acetyl-α-L-rhamnopyranosyl bromide in benzene–MeCN [$Hg(CN)_2$] at 30°, gave 63% of the 3-*O*-glycosylated product (**13**) as a foam, m.p. 102°, $[\alpha]_D$ −16° (MeOH), accompanied by minor proportions of the 4-*O*-substituted product (14%) and a tetrasaccharide derivative.

The fully acetylated β-trisaccharide derivative **7** reacted with 1.1 mol of *N*-bromosuccinimide in boiling, 1:9 $(CHCl_2)_2$–CCl_4 (tungsten light) during 5 min to open the benzylidene acetal ring and give 82% of the corresponding 6-bromide 4-benzoate **14**, m.p. 163–164°, $[\alpha]_D$ −49° (MeOH), and reduction of the latter with hydrogen (50 lb. $in.^{-2}$)–Raney nickel gave 80% of the 6-deoxy analog **15**. m.p. 176–177°, $[\alpha]_D$ −55° (MeOH). Final Zemplén deprotection of **15** afforded 100% of the target β-glycoside, namely 8-(methoxycarbonyl)octyl 2-acetamido-2,6-dideoxy-3-*O*-[3-*O*-(α-L-rhamnopyranosyl)-α-L-rhamnopyranosyl]-β-D-glucopyranoside (**16**), m.p. 98–100° (dec.), $[\alpha]_D$ −66° (MeOH). Similarly, acetylation of the α-trisaccharide derivative **13**, followed by treatment of the acetate ($[\alpha]_D$ +7° in MeOH) with 1.1 mol of *N*-bromosuccinimide in boiling CCl_4 for 5 min (tungsten light) gave 88% of the syrupy 6-bromide 4-benzoate α-analog ($[\alpha]_D$ ~0° in MeOH) of **14**, which, on hydrogenolysis (86%; product $[\alpha]_D$ −3° in MeOH), followed by *O*-deacylation (83%), gave the α anomer (**17**) of **16**.

The foregoing syntheses gave the target trisaccharide glycosides in acceptable net yields at the 0.5-g level, and all products were adequately identified by ^{1}H- and ^{13}C-n.m.r. spectroscopy, and by other analytical data. The practical feasibility of the 6-deoxygenation step[13] at the trisaccharide level is noteworthy, and the consequent availability of the 4,6-protecting group on the amino sugar considerably simplified the synthetic strategy.

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