Unusual sugars of the GPL-type antigen of *Mycobacterium avium* serovar 19. Stereoselective synthesis of methyl 6-deoxy-3-*C*-methyl-2,4-di-*O*-methyl-α-L-mannopyranoside and its C-4 epimer

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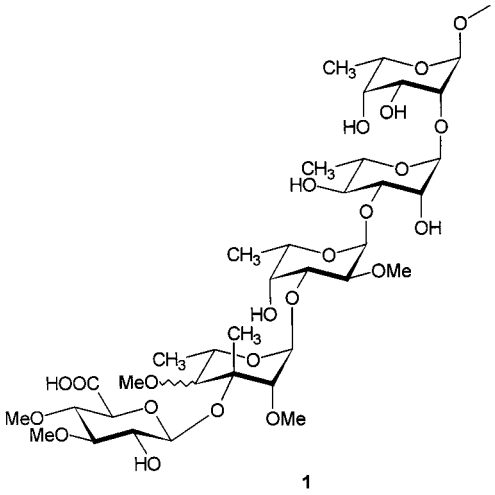
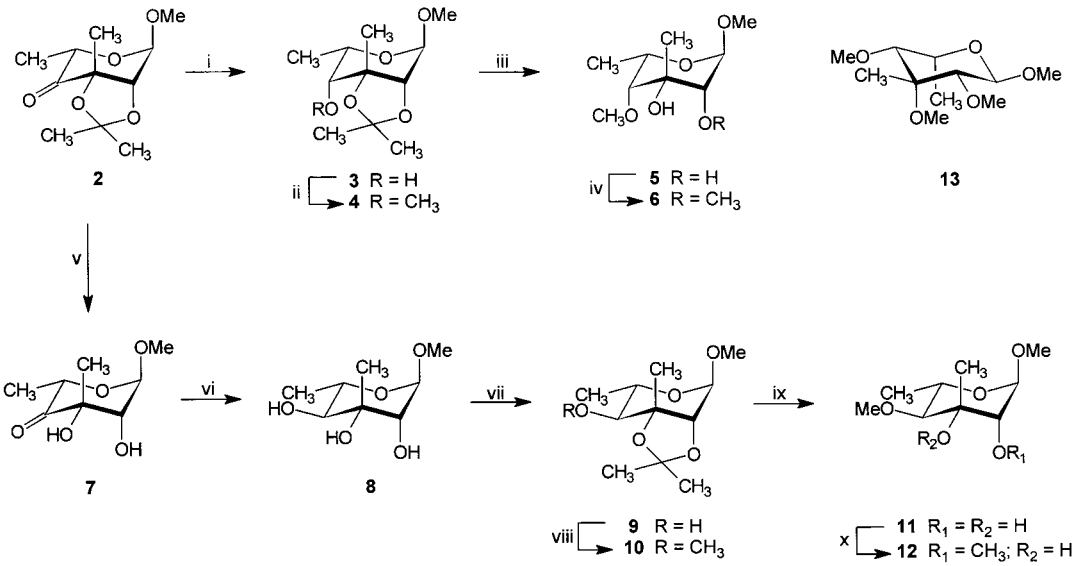
Abstract: Completely reversed stereoselectivity of reduction of methyl 6-deoxy-2,3-*O*-isopropylidene-3-*C*-methyl-α-L-mannopyranoside (**2**) and its deisopropylidenated derivative (**7**) was observed. Compound **2** gave exclusively the L-*talo*-isomer (**3**) with $NaBH_4$ in MeOH, but the reduction of **7** with $NaBH_4$ in acetic acid resulted in the L-*manno*-derivative **8**. It is assumed that in the first case the stereoselectivity is determined by the steric accessibility of the carbonyl group, while in the second case free OH-groups direct the selectivity of the reduction by complexation or ligand exchange.

Members of the *Mycobacterium avium* serocomplex[1,2] are opportunistic pathogens and can cause serious infections. Structural analysis of the cell-surface glycopeptidolipid-type (GPL) antigen of serovar 19 showed that it contains the *O*-linked pentasaccharide **1**[3]. While most of the structural features of the penultimate residue (Fig.) have been determined, ambiguity persists regarding the stereochemistry at C-4 of this unit.



Figure

Here we report stereoselective syntheses of both epimers (**6, 12**) of the unidentified unit as their methyl glycosides in the proposed, α anomeric forms[3]. Precursor to both targets was methyl 6-deoxy-2,3-*O*-isopropylidene-3-*C*-methyl-α-L-*lyxo*-hexopyranosid-4-ulose (**2**) that was prepared as described by Klemer from L-rhamnose in four steps[4]. Reduction of compound **2** either with $NaBH_4$ or with $LiAlH_4$ (Scheme) resulted in the 6-deoxy-L-*talo* isomer **3** exclusively, as confirmed by the low value (1 Hz) of the $^3J_{4,5}$ coupling constant. The complete stereoselectivity of this reduction can be explained by the extremely crowded β-(L) side of compound **2**: the hydride anion can approach the C-4 carbon atom only from the α-(L) face. Methylation of compound **3** resulted in **4** from which the isopropylidene group was removed by acidic hydrolysis to obtain **5**. Methylation of the axial OH-2 of **5** required rather drastic conditions to give **6**, and formation of the fully methylated product (**13**) was also observed (15 %). Compound **13** exists exclusively in $^4C_1$ (L) conformation.



Reagents and conditions: i. $NaBH_4$, MeOH, 90 %; ii. MeI, NaH, DMF, 83 %; iii. AcOH, H_2O, 86 %; iv. MeI, NaH, DMF, 59 %; v. AcOH, H_2O, TFA, 65 %; vi. $NaBH_4$, AcOH, 99 %; vii. DMP, pTsA, DMF, 98 %; viii. MeI, NaH, DMF, 68 %; ix. AcOH, H_2O, TFA, 80 %; x. MeI, NaOH, Bu_4NBr, CH_2Cl_2, 67 %

Scheme

To prepare the *rhamno*-isomer (**12**) from compound **2**, its isopropylidene group was hydrolyzed, then the free ulose-derivative (**7**) was treated with $NaBH(OAc)_3$ (Scheme 1)[5,6]. Methyl α-L-evalopyranoside was isolated in nearly quantitative yield. In its ^{1}H-NMR spectrum H-4 gave a doublet at 3.39 ppm having $^3J_{4,5}$ 10 Hz, that confirms the *trans*-diaxial relationship of the H-4 and H-5 protons. The completely reversed stereoselectivity compared to the reduction of **2** could only be achieved in the presence of the free OH groups suggesting that these play a role in the complexation of the hydride donor thus governing the direction of the attack of the hydride anion. Isopropylidenation of **8** gave compound **9** and after methylation (→**10**) and deprotection (→**11**) the resulting diol was selectively methylated at OH-2 at 0 °C under phase-transfer conditions. The isolated yield of compound **12** was 67 % after chromatographic purification.

The $[\alpha]_D$ value and the ^{1}H-NMR data of **12** were in good agreement with the data of one of the intermediates of L-nogalose synthesis that was obtained from methyl 3,6-dideoxy-2,4-di-*O*-methyl-3-*C*-methylene-α-L-*arabino*-hexopyranoside by epoxidation to provide a 1 : 1 ratio of two diastereoisomers followed by $LiAlH_4$ reduction of one of the epoxides[7].

In summary, we developed synthetic routes to C-4 epimers of a branched monosaccharide residue of the glycopeptidolipid antigen of *Mycobacterium avium* serovar 19 based on common precursor **2**.

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8. Physical and spectroscopic data of the synthesized compounds: **3** $[\alpha]_D$ -42.13 (c 1.08); ^{1}*H NMR:* δ 4.93 (s, 1H, H-1), 3.88 (dd, 1H, $J_{4,5}$ 1 Hz, $J_{5,Me(6)}$ 6.5 Hz, H-5), 3.75 (s, 1H, H-2), 3.40 (s, 3H, OCH_3), 3.17 (dd, 1H, $J_{4,5}$ 1 Hz, $J_{4,OH}$ 5 Hz, H-4), 2.48 (d, 1H, $J_{4,OH}$ 5 Hz, OH), 1.57 and 1.38 (s, 3-3H, Ip CH_3), 1.42 (s, 3H, CH_3(3)), 1.34 (d, 3H, $J_{5,Me(6)}$ 6 Hz, CH_3(6));
4 $[\alpha]_D$ -37.8 (c 0.35); ^{1}*H NMR* : δ 4.92 (s,1H, H-1), 3.83 (dd, 1H, $J_{5,6}$ 6 Hz, $J_{4,5}$ 1 Hz, H-5), 3.69 (s, 1H, H-2), 3.52 (s, 3H, OCH_3(4)), 3.37 (s, 3H, OCH_3(1)), 2.69 (d, 1H, $J_{4,5}$ 1 Hz, H-4), 1.56 and 1.4 (2s, 3-3H, Ip CH_3), 1.37 (s, 3H, CH_3(3)), 1.31 (d, 3H, $J_{5,6}$ 6 Hz, CH_3(6)); ^{13}*C NMR* : δ 109.03 (Ip), 98.4 (C-1), 89.2 (C-2), 78.69 (C-3) 78.6 and 64.4 (C-4 and C-5), 62.11 (OCH_3(4)), 55.05 (OCH_3(1)), 26.78 and 25.69 (2 x Ip CH_3), 24.84 and 16.75 (CH_3(3) and CH_3(6));
5 $[\alpha]_D$ -103.7 (c 1.04); ^{1}*H NMR* : δ 4.78 (s,1H, H-1), 3.94 (d, 1H, $J_{5,6}$ 6.5 Hz, H-5), 3.59 (s, 3H, OCH_3(4)), 3.37 (s, 3H, OCH_3(1)), 3.36 (bs, 2H, 2 x OH), 3.28 (s, 1H, H-2), 2.89 (s, 1H, H-4), 1.32 (s, 3H, CH_3(3)), 1.3 (d, 3H, $J_{5,6}$ 6.5 Hz, CH_3(6)); ^{13}*C NMR* : δ 102.49 (C-1), 87.27 (C-2), 73.80 and 65.13 (C-4 and C-5), 69.48 (C-3), 62.84 (OCH_3(4)), 55.27 (OCH_3(1)), 22.42 and 17.00 (CH_3(3) and CH_3(6));
6 $[\alpha]_D$ -56.33 (c 1.04); ^{1}*H NMR:* δ 4.78 (s, 1H, H-1), 3.91 (d, 1H, $J_{5,6}$ 6 Hz, H-5), 3.56 (s, 3H, OCH_3(4)), 3.47 (s, 1H, OCH_3(2)), 3.37 (s, 1H, OCH_3(1)), 2.84 and 2.76 (2s, 1-1H, H-2 and H-4), 1.33 (s, 3H, CH_3(3)), 1.32 (d, 1H, $J_{5,6}$ 6 Hz, CH_3(6)); ^{13}*C NMR* : δ 98.75 (C-1), 85.9 (C-2), 82.87 and 64.76 (C-4 and C-5), 69.1 (C-3), 55.73 (OCH_3), 23.86 and 16.77 (CH_3(3) and CH_3(6));
7 $[\alpha]_D$ -101.5 (c 1.26); ^{1}*H NMR* : δ 4.85 (s, 1H, H-1), 4.52 (d, 1H, $J_{5,6}$ 6.5 Hz, H-5), 4.22 and 3.97 (2s, 1-1H, 2 x OH), 3.52 (s, 3H, OCH_3), 3.43 (s, 1H, H-2), 1.54 (s, 3H, CH_3(3)), 1.34 (d, 3H, $J_{5,6}$ 6.5 Hz, CH_3(6)); ^{13}*C NMR* : δ 204.04 (C-4), 100.47 (C-1), 86.58 (C-3), 78.23 and 66.78 (C-2 and C-5), 55.98 (OCH_3), 23.88 (CH_3(3)), 13.81 (CH_3(6));
8 $[\alpha]_D$ -83.3 (c 0.85, MeOH); ^{1}*H NMR:* δ 4.58 (s, 1H, H-1), 3.56 (dd, 1H, $J_{4,5}$ 10 Hz, $J_{5,Me(6)}$ 6 Hz, H-5), 3.47 (s, 1H , H-2), 3.39 (d, 1H, $J_{4,5}$ 10 Hz, H-4) 3.34 (s, 3H, OCH_3), 1.26 (d, 3H, $J_{5,Me\,(6)}$ 6 Hz, CH_3(6)), 1.24 (s, 3H, CH_3(3)); ^{13}*C NMR :* δ 103.28 (C-1), 76.39 (C-2), 76.12 and 68.56 (C-4 and C-5), 73.58 (C-3) 55.3 (OCH_3), 19.18 and 18.4 (CH_3(3) and CH_3(6));
9 $[\alpha]_D$ -53.7 (c 1.10); ^{1}*H NMR:* δ 4.85 (s, 1H, H-1), 3.87 (s, 1H, H-2), 3.61 (dd, 1H, $J_{4,5}$ 9 Hz, $J_{5,Me(6)}$ 6 Hz, H-5), 3.54 (dd, 1H, $J_{4,5}$ 9 Hz, $J_{4,OH}$ 3.5 Hz, H-4), 3.38 (s, 3H, OCH_3), 3.02 (d, 1H, $J_{4,OH}$ 3.5 Hz, OH), 1.53 and 1.38 (2s, 3-3H, 2 x Ip CH_3), 1.36 (s, 3H, CH_3(3)), 1.3 (d, 3H, $J_{5,Me(6)}$ 6 Hz, CH_3(6)); ^{13}*C NMR*: δ 108.76 (Ip kvat.), 98.04 (C-1), 81.56 (C-3), 80.74 (C-2), 76.65 and 65.52 (C-4 and C-5), 54.95 (OCH_3), 28.2 and 26.4 (2 x Ip CH_3), 17.51 and 17.45 (CH_3(3) and CH_3(6));
10 $[\alpha]_D$ -72.7 (c 1.07); ^{1}*H NMR:* δ 4.83 (d, 1H, $J_{1,2}$ 1 Hz, H-1), 3.8 (d, 1H, $J_{1,2}$ 1 Hz, H-2), 3.56 (dd, 1H, $J_{4,5}$ 10 Hz, $J_{5,Me(6)}$ 6.5 Hz, H-5), 3.54 (s, 3H, OCH_3(4)), 3.37 (s, 3H, OCH_3(1)), 3.12 (d, 1H, $J_{4,5}$ 10 Hz, H-4), 1.55 and 1.37 (2s, 3-3H, 2 x Ip CH_3), 1.33 (s, 3H, CH_3(3)), 1.27 (d, 3H, $J_{5,Me(6)}$ 6,5 Hz, CH_3(6)); ^{13}*C NMR*: δ 108.54 (Ip kvat.), 98.01 (C-1), 85.74 (C-2), 81.90 (C-3), 81.06 and 65.2 (C-4 and C-5), 60.49 (OCH_3(4)), 54.99 (OCH_3(1)), 28.28 and 27.09 (2 x Ip CH_3) 17.87 and 17.73 (CH_3(3) and CH_3(6));
11 $[\alpha]_D$ -99.4 (c 0.81); ^{1}*H NMR:* δ 4.67 (d,1H, $J_{1,2}$ 1,5 Hz, H-1), 3.59 (dd, 1H, $J_{4,5}$ 10 Hz, $J_{5,6}$ 6 Hz, H-5), 3.57 (s, 3H, OCH_3(4)), 3.55 (d, 1H, $J_{1,2}$ 1,5 Hz, H-2), 3.36 (s, 3H, OCH_3(1)), 3.05 (d, 1H, $J_{4,5}$ 10 Hz, H-4), 2.68 (bs, 2H, 2 x OH), 1.32 (s, 3H, CH_3(3)), 1.31 (d, 3H, $J_{5,6}$ 6 Hz, CH_3(6)); ^{13}*C NMR*: δ 100.94 (C-1), 85.39 (C-2), 73.84 (C-3), 75.29 and 66.77 (C-4 and C-5), 61.72 (OCH_3(4)), 55.11 (OCH_3(1)), 19.41 and 18.08 (CH_3(3) and CH_3(6));
12 $[\alpha]_D$ -57.67 (c 0.75); ^{1}*H NMR:* δ 4.71 (d, 1H, $J_{1,2}$ 1 Hz, H-1), 3.58 (s, 3H, OCH_3(4)), 3.53 (dd, 1H, $J_{4,5}$ 10 Hz, $J_{5,6}$ 6 Hz, H-5), 3.49 (s, 3H, OCH_3(2)), 3.36 (s, 3H, OCH_3(1)), 3.07 (d, 1H, $J_{1,2}$ 1 Hz, H-2), 3.01 (s, 1H, OH), 2.9 (d,1H, $J_{4,5}$ 10 Hz, H-4), 1.3 (s, 3H, CH_3(3)), 1.28 (d, 3H, $J_{5,6}$ 6 Hz, CH_3(6)); ^{13}*C NMR*: δ 97.78 (C-1), 86.2 (C-2), 85.08 and 66.69 (C-4 and C-5), 73.25 (C-3), 61.6 and 59.07 (OCH_3(2) and OCH_3(4)), 54.94 (OCH_3(1)), 18.54 and 18.01 (CH_3(3) and CH_3(6));
13 ^{1}*H NMR:* δ 4.79 (d, 1H, $J_{1,2}$ 4 Hz, H-1), 4.10 (m, 1H, $J_{4,5}$ 3.5 Hz, $J_{5,6}$ 7 Hz, H-5), 3.51, 3.50, 3.43, 3.35 (s, 3-3H, OMe), 2.98 (d, 1H, H-4), 2.89 (d, 1H, H-2), 1.36 (d, 3H, CH_3(6)), 1.35 (s, 3H, CH_3(3)) ; ^{13}*C NMR*: δ 98.34 (C-1), 83.79 (C-2), 82.92, 67.45 (C-4 and C-5), 60.76, 60.33, 55.77, 50.82 (OCH_3), 18.54 (CH_3(3)), 15.01 (CH_3(6)).