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An efficient and stereoselective synthesis of β-D-Ara*p*-(1→2)-β-D-Gal*p*-(1→3)-β-D-Gal*p*-(1→4)-α-D-Man*p*, a tetrasaccharide fragment of *Leishmania major* lipophosphoglycan

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Abstract

A tetrasaccharide fragment of *Leishmania major* lipophosphoglycan (which seems to be involved in a biological mechanism for the parasite transmission) has been synthesised using the thioglycoside, trichloroacetimidate and halide-exchange glycosylation procedures and step-wise chain elongation strategy. © 2001 Elsevier Science Ltd. All rights reserved.

Keywords: Carbohydrates; Oligomers; Glycosylation; Parasites

The surface lipophosphoglycan (LPG) produced by the insect dwelling infectious promastigote stage of all species of the *Leishmania* parasite contains a polymeric section consisting of β-D-Gal*p*-(1→4)-α-D-Man*p* phosphate repeat units. In LPG of *Leishmania major*,[1] the D-galactose units are, in the main, randomly substituted at O-3 with β-D-Gal*p*, β-D-Gal*p*-(1→3)-β-D-Gal*p* and β-D-Ara*p*-(1→2)-β-D-Gal*p*. The composition of the side-chains changes during the differentiation of the parasite, presenting various biological properties on the LPG.[2] The increase of the proportion of β-D-Ara*p*-terminating side-chains (from 9 to 45%) seems to be a biological mechanism for the detachment of the infectious metacyclic promastigotes from the mid-gut epithelial cells of the sand-fly vector[3] prior to their transmission to a mammalian host.

Recent reports from this laboratory disclosed the synthesis and biological evaluation of phosphosaccharide fragments of the LPG of *L. donovani*,[4] *L. mexicana*[5] and *L. major*[6] (the latter contained β-D-Gal*p* side-chains only). Here we report the synthesis of a tetrasaccharide β-D-Ara*p*-(1→2)-β-D-Gal*p*-(1→3)-β-D-Gal*p*-(1→4)-α-D-Man*p* (**1**), which is due to be incorporated further in the preparation of the phosphosaccharides **2** and **3** (Scheme 1) using the tetrasaccharide H-phosphonate derivative **4** and H-phosphonate methodol-

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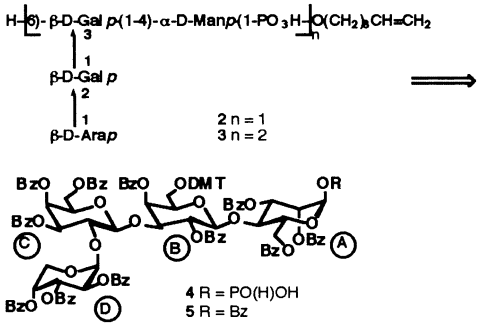
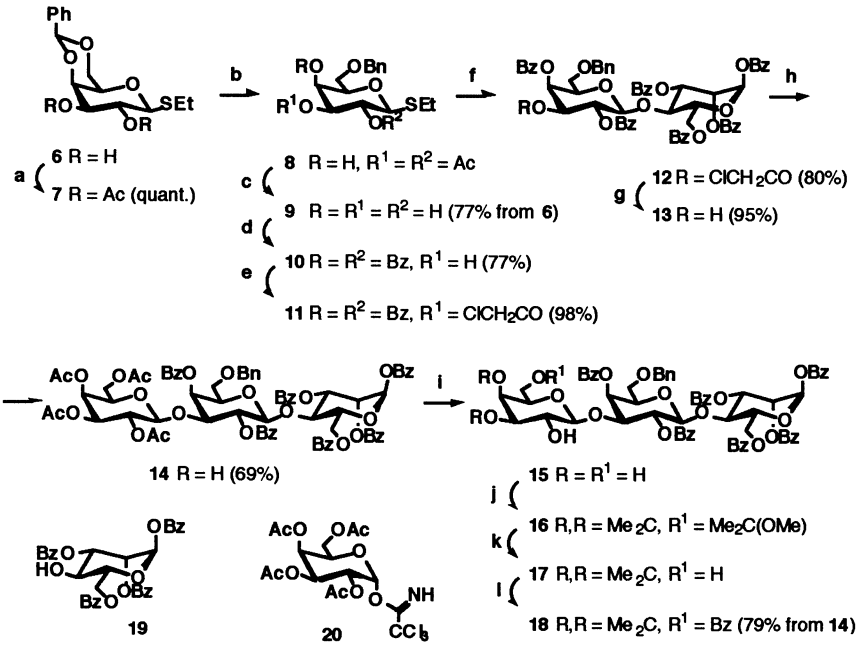


Scheme 1.

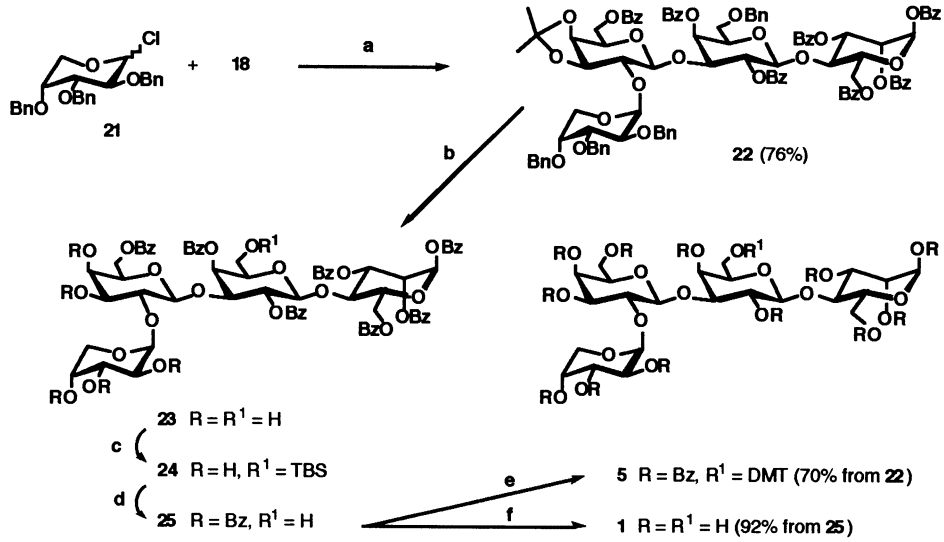
ogy.[4–6] The phosphosaccharides **2** and **3** will be used for the preparation of artificial antigens.

The synthetic plan leading to compound **5** (a direct precursor of the H-phosphonate **4**) was designed regarding that (1) the β-glycoside linkage between units **C** and **B** could be prepared with participating group at C-2 in the unit **C** and (2) the preparation of the **A**–**B** block should precede the rest of the synthesis.[7a] Thus, linear step-wise sequence **A** + **B** → **A**–**B** + **C** → **A**–**B**–**C** + **D** was chosen to ensure selective and efficient synthesis of the target tetrasaccharide molecule.[7b]

The required unit **B** (in a form of the ethylthio β-D-galactoside **11** containing temporary protections at O-3 and -6; Scheme 2) was prepared starting from the diol **6**.[8] The key step of the transformation was reductive benzylidene ring cleavage in the diacetate derivative **7**[9] with $NaBH_3CN$–$MeSO_3H$ system (it was the only one from a big variety of synthetic procedures of this kind,[10] which did work efficiently to produce compound **8**[9]). Successive deacetylation (→**9**;[9] 77% from **6**) and 2,4-protection via 3,4-*O*-orthobenzoate formation, 2-*O*-benzoylation and regioselective acidic cleavage of the orthobenzoate[11] gave the 3-OH derivative **10**[9] (77%). Standard chloroacetylation resulted in selectively protected compound **11**. Glycosylation of the D-mannose derivative **19**[4a] (unit **A**) with the thioglycoside **11** in the presence of MeOTf and MS 4Å provided stereoselectively the β-linked disaccharide **12**[9] ($J_{1',2'}$ 7.8 Hz) in 80% yield. It was further dechloroacetylated[5] with thiourea (→**13**,[9] 95%; the **A**–**B** block) followed by the reaction with the D-galactosyl trichloroacetimidate **20** (unit **C**) in the presence of TMSOTf[12] to produce the β,β-linked trisaccharide **14**[9] (69%; $J_{1'',2''}$ 7.8 Hz). A routine sequence of protecting group manipulation including deacetylation[13] (→**15**) with HCl in MeOH, acetalisation[14] (→**16**) with 2,2-



Scheme 2. Reagents and conditions: (a) Ac_2O, pyridine; (b) $NaBH_3CN$, $MeSO_3H$, THF; (c) MeOH, MeONa ca.; (d) (1) $PhC(OEt)_3$, $TsOH{\cdot}H_2O$, DCM; (2) BzCl, pyridine; (3) 1:0.1:1000 TFA–H_2O–DCM; (e) $ClCH_2COCl$, pyridine, DCM; (f) **19**, MeOTf, MS 4Å, DCM; (g) $(NH_2)_2CS$, 2,4,6-collidine, MeOH, DCM, Δ; (h) **20**, TMSOTf ca., DCM; (i) HCl, MeOH, DCM; (j) $Me_2C(OMe)_2$, $TsOH{\cdot}H_2O$; (k) 3:3:1000 TFA–H_2O–DCM; (l) BzCN, Et_3N, MeCN.



Scheme 3. Reagents and conditions: (a) *n*-Bu_4NBr, MS 4Å, DCE, Δ; (b) (1) 100:1:1000 TFA–H_2O–DCM; (2) H_2, $Pd(OH)_2$–C, MeOH; (c) TBSCl, DMAP, DCE; (d) (1) BzCl, pyridine; (2) HBF_4, MeCN; (e) DMTCl, DMAP, pyridine; (f) MeOH, MeONa ca.

dimethoxypropane–TsOH, mild-acid promoted cleavage of the mixed acetal (→**17**) and selective 6-*O*-benzoylation[15] with benzoyl cyanide afforded the monohydroxyl trisaccharide derivative **18**[9] (79% from **14**) ready to be used as the **A**–**B**–**C** glycosyl acceptor block.

Glycosylation of the trisaccharide **18** with the D-arabinopyranosyl chloride **21**[16] (unit **D**) in the presence of *n*-Bu_4NBr and MS 4Å (the halide-exchange procedure[17]) provided the β,β,β-linked tetrasaccharide **22**[9] (Scheme 3; 76%; $J_{1''',2'''}$ 3.4 Hz) as a single product of the reaction. Successive removal of isopropylidene and benzyl groups produced the hexaol **23** in 97% yield. Unexpectedly, direct 6′-*O*-tritylation of this polyol with DMTCl–DMAP–pyridine system failed due to, probably, partial migration of the 6″-*O*-benzoyl group to the 4″-*O*-position followed by bis-tritylation. Alternatively, selective silylation of the hexaol **23** with TBSCl–DMAP in 1,2-dichloroethane led smoothly to the 6′-*O*-TBS derivative **24**, which was then perbenzoylated followed by desilylation with HBF_4 (→**25**) and tritylation with DMTCl–DMAP to give the desired tetrasaccharide derivative **5**[9] in 70% overall yield. Debenzoylation of compound **25** with catalytic MeONa in MeOH produced the unprotected tetrasaccharide **1**[9] in 92% yield.

To summarise, we have successfully prepared a unique tetrasaccharide related to β-D-Ara*p* enriched LPGs from *L. major*.

Acknowledgements

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Appendix A. Analytical and spectroscopic data

Ethyl 2,3-di-O-*acetyl-4,6*-O-*benzylidene-1-thio-β*-D-*galactopyranoside* (**7**).—Solid; mp 112–113 °C (EtOH); $[\alpha]_D^{20}$ +35° (*c* 1, $CHCl_3$); R_f 0.88 (solvent A, 1:9 MeOH–CH_2Cl_2); (Anal. Found: C, 57.76; H, 6.12. $C_{19}H_{24}O_7S$ requires C, 57.56; H, 6.10); δ_H (300 MHz; the same for the all ^1H NMR data) 1.32 (3 H, t, *J* 7.4, SCH_2CH_3), 2.09 and 2.10 (2 × 3 H, 2 × s, Ac), 2.70–3.00 (2 H, m, SCH_2CH_3), 3.59 (1 H, br s, 5-H), 4.04 (1 H, dd, $J_{5,6a}$ 1.6, $J_{6a,6b}$ 12.5, 6-H[a]), 4.37 (1 H, dd, $J_{5,6b}$ 1.4, 6-H[b]), 4.44 (1 H, d, $J_{3,4}$ 3.5, 4-H), 4.48 (1 H, d, $J_{1,2}$ 10.0, 1-H), 5.01 (1 H, dd, $J_{2,3}$ 10.0, 3-H), 5.50 (1 H, t, 2-H), 5.52 (1 H, s, PhC*H*) and 7.40–7.60 (5 H, m, Ph).

Ethyl 2,3-di-O-*acetyl-6*-O-*benzyl-1-thio-β*-D-*galactopyranoside* (**8**).—Amorphous solid; $[\alpha]_D^{20}$ 0° (*c* 1, $CHCl_3$); R_f 0.28 (solvent B, 3:7 EtOAc–$PhCH_3$); (Anal. Found: C, 57.64; H,

6.79. $C_{19}H_{26}O_7S$ requires C, 57.27; H, 6.58); δ_H 1.19 (3 H, t, J 7.5, SCH_2CH_3), 1.98 and 2.62 (2 × 3 H, 2 × s, Ac), 2.58–2.77 (2 H, m, SCH_2CH_3), 2.87 (1 H, br s, OH), 3.60–3.75 (3 H, m, 5-H, 6-H[a] and 6-H[b]), 4.13 (1 H, br d, $J_{3,4}$ 3.0, 4-H), 4.37 (1 H, d, $J_{1,2}$ 9.9, 1-H), 4.46 and 4.51 (AB-system, J_{gem} 11.9, $PhCH_2$), 4.89 (1 H, dd, $J_{2,3}$ 9.8, 3-H), 5.28 (1 H, t, 2-H) and 7.23 (5 H, m, Ph).

*Ethyl 6-*O*-benzyl-1-thio-β-*D*-galactopyranoside* (**9**).—Amorphous solid; $[\alpha]_D^{20}$ +21° (*c* 1, $CHCl_3$); R_f 0.44 (solvent A); (Anal. Found: C, 57.04; H, 6.92. $C_{15}H_{22}O_5S$ requires C, 57.30; H, 7.05).

*Ethyl 2,4-di-*O*-benzoyl-6-*O*-benzyl-1-thio-β-*D*-galactopyranoside* (**10**).—Solid; mp 107–108 °C (4:1 hexane–EtOAc); $[\alpha]_D^{20}$ +9° (*c* 1, $CHCl_3$); R_f 0.56 (solvent B); (Anal. Found: C, 66.69; H, 5.79. $C_{29}H_{30}O_7S$ requires C, 66.65; H, 5.79); δ_H 1.33 (3 H, t, J 7.4, SCH_2CH_3), 2.72–2.92 (3 H, m, SCH_2CH_3 and OH), 3.67 (2 H, m, 6-H[a] and 6-H[b]), 4.01 (1 H, br t, $J_{5,6a} = J_{5,6b}$ 6.1, 5-H), 4.15 (1 H, m, 3-H), 4.45 and 4.55 (AB-system, J_{gem} 11.7, $PhCH_2$), 4.72 (1 H, d, $J_{1,2}$ 9.9, 1-H), 5.42 (1 H, t, $J_{2,3}$ 9.9, 2-H), 5.78 (1 H, d, $J_{3,4}$ 2.8, 4-H) and 7.20–8.30 (15 H, m, 3 × Ph).

*Ethyl 2,4-di-*O*-benzoyl-6-*O*-benzyl-3-*O*-chloroacetyl-1-thio-β-*D*-galactopyranoside* (**11**).—Solid; mp 105–106 °C; $[\alpha]_D^{20}$ +60° (*c* 1, $CHCl_3$); R_f 0.59 (solvent C, 1:9 EtOAc–$PhCH_3$); (Anal. Found: C, 62.23; H, 5.21. $C_{31}H_{31}ClO_8S$ requires C, 62.15; H, 5.22); δ_H 1.32 (3 H, t, J 7.4, SCH_2CH_3), 2.82 (2 H, m, SCH_2CH_3), 3.60 (1 H, dd, $J_{5,6a}$ 7.3, $J_{6a,6b}$ 9.4, 6-H[a]), 3.71 (1 H, dd, $J_{5,6b}$ 5.9, 6-H[b]), 3.88 and 3.95 (AB-system, J_{gem} 15.2, $ClCH_2$), 4.12 (1 H, br t, 5-H), 4.42 and 4.54 (AB-system, J_{gem} 11.8, $PhCH_2$), 4.75 (1 H, d, $J_{1,2}$ 9.9, 1-H), 5.45 (1 H, dd, $J_{3,4}$ 3.4, $J_{2,3}$ 9.9, 3-H), 5.63 (1 H, t, 2-H), 5.87 (1 H, d, 4-H) and 7.20–8.30 (15 H, m, 3 × Ph).

*2,4-Di-*O*-benzoyl-6-*O*-benzyl-3-*O*-chloroacetyl-β-*D*-galactopyranosyl-(1→4)-1,2,3,6-tetra-*O*-benzoyl-α-*D*-mannopyranose* (**12**).—Amorphous solid; $[\alpha]_D^{20}$ +37° (*c* 1, $CHCl_3$); R_f 0.44 (solvent C); (Anal. Found: C, 66.98; H, 4.77. $C_{63}H_{53}ClO_{18}$ requires C, 66.75; H, 4.71); δ_H 2.91 (1 H, dd, $J_{5',6'a}$ 5.4, $J_{6'a,6'b}$ 8.2, H′-6[a]), 3.00 (1 H, t, $J_{5',6'b}$ 8.9, 6′-H[b]), 3.55 (1 H, br dd, H′-5), 3.68 and 3.77 (AB-system, J_{gem} 15.3, $ClCH_2$), 3.91 and 4.08 (AB-system, J_{gem} 12.1, $PhCH_2$), 4.14 (1 H, br d, $J_{4,5}$ 9.8, 5-H), 4.39 (1 H, dd, $J_{5,6a}$ 2.4, $J_{6a,6b}$ 12.4, 6-H[a]), 4.54 (1 H, br d, 6-H[b]), 4.57 (1 H, t, $J_{3,4}$ 9.8, 4-H), 4.83 (1 H, d, $J_{1',2'}$ 7.8, 1′-H), 5.18 (1 H, dd, $J_{3',4'}$ 3.3, $J_{2',3'}$ 10.4, 3′-H), 5.45 (1 H, dd, 2′-H), 5.54 (1 H, d, 4′-H), 5.76 (1 H, dd, $J_{1,2}$ 1.9, $J_{2,3}$ 3.3, 2-H), 5.87 (1 H, dd, 3-H), 6.39 (1 H, d, 1-H) and 6.90–8.20 (35 H, 7 × Ph).

*2,4-Di-*O*-benzoyl-6-*O*-benzyl-β-*D*-galactopyranosyl-(1→4)-1,2,3,6-tetra-*O*-benzoyl-α-*D*-mannopyranose* (**13**).—Amorphous solid; $[\alpha]_D^{20}$ +10° (*c* 1, $CHCl_3$); R_f 0.38 (solvent D, 1:4 EtOAc–$PhCH_3$); (Anal. Found: C, 69.37; H, 5.07. $C_{61}H_{52}O_{17}$ requires C, 69.31; H, 4.96); δ_H 2.53 (1 H, br s, OH), 2.92 (2 H, d, $J_{5'6'}$ 6.9, H′-6[a,b]), 3.51 (1 H, br t, H′-5), 3.91 (1 H, dd, $J_{3',4'}$ 3.3, $J_{2',3'}$ 10.0, 3′-H), 3.97 and 4.06 (AB-system, J_{gem} 12.0, $PhCH_2$), 4.16 (1 H, br d, $J_{4,5}$ 9.7, 5-H), 4.54 (2 H, br s, 6-H[a,b]), 4.57 (1 H, t, $J_{3,4}$ 9.7, 4-H), 4.75 (1 H, d, $J_{1',2'}$ 7.8, 1′-H), 5.24 (1 H, dd, 2′-H), 5.46 (1 H, d, 4′-H), 5.75 (1 H, dd, $J_{1,2}$ 1.8, $J_{2,3}$ 3.2, 2-H), 5.87 (1 H, dd, 3-H), 6.41 (1 H, d, 1-H) and 6.95–8.20 (35 H, m, 7 × Ph).

*2,3,4,6-Tetra-*O*-acetyl-β-*D*-galactopyranosyl-(1→3)-2,4-di-*O*-benzoyl-6-*O*-benzyl-β-*D*-galactopyranosyl-(1→4)-1,2,3,6-tetra-*O*-benzoyl-α-*D*-mannopyranose* (**14**).—Amorphous solid; $[\alpha]_D^{20}$ +23° (*c* 1, $CHCl_3$); R_f 0.16 (solvent D); (Anal. Found: C, 64.71; H, 5.55. $C_{75}H_{70}O_{26}$ requires C, 64.93; H, 5.09); δ_H 1.36, 1.73, 1.88 and 1.90 (12 H, 4 × s, 4 × Ac), 2.95 (2 H, d, $J_{5',6'}$ 6.3, 6′-H[a,b]), 3.46 (1 H, br t, 5′-H), 3.64 (1 H, br t, $J_{5'',6''a} = J_{5'',6''b}$ 6.6, 5″-H), 3.85 (1 H, dd, $J_{6''a,6''b}$ 11.2, 6″-H[a]), 3.91 (1 H, dd, $J_{3',4'}$ 3.3, $J_{2',3'}$ 10.0, 3′-H), 4.04 (1 H, dd, 6″-H[b]), 4.06 (2 H, m, $PhCH_2$), 4.10 (1 H, m, 5-H), 4.41 (1 H, $J_{5,6a}$ 2.8, $J_{6a,6b}$ 11.2, 6-H[a]), 4.41 (1 H, d, $J_{1'',2''}$ 7.8, 1″-H), 4.51 (1 H, br d, 6-H[b]), 4.54 (1 H, dd, $J_{3'',4''}$ 3.4, $J_{2'',3''}$ 10.4, 3″-H), 4.60 (1 H, t, $J_{3,4} = J_{4,5} = 9.7$, 4-H), 4.73 (1 H, d, $J_{1',2'}$ 7.9, 1′-H), 4.82 (1 H, dd, 2″-H), 5.10 (1 H, d, 4″-H), 5.51 (1 H, br s, 4′-H), 5.51 (1 H, dd, 2′-H), 5.75 (1 H, dd, $J_{1,2}$ 1.7, $J_{2,3}$ 3.3, 2-H), 5.84 (1 H, dd, 3-H), 6.39 (1 H, d, 1-H) and 6.85–8.10 (35 H, 7 × Ph); δ_C (75 MHz ; the same for the all ^{13}C NMR data) 19.6, 20.3, 20.4 and 20.5 (4 × Ac), 60.7 (C-6″), 61.9 (C-6), 66.3 (C-4″), 67.4 (C-6′), 68.2 (C-2″), 69.4 (C-2), 69.6 (C-4′), 71.8 (C-2′), 70.0 (C-3), 70.3

(C-5″), 70.7 (C-3″), 71.6 (C-5), 72.3 (C-4), 72.9 (C-5′),73.2 (*C*H_2Ph), 77.0 (C-3′), 91.0 (C-1), 101.0 (C-1′and C-1″), 127.6–(Ph) and 164.7–170.2 (C=O).

6-O-Benzoyl-3,4-O-isopropylidene-β-D-galactopyranosyl-(1→3)-2,4-di-O-benzoyl-6-O-benzyl-β-D-galactopyranosyl-(1→4)-1,2,3,6-tetra-O-benzoyl-α-D-mannopyranose (**18**).—Amorphous solid; $[\alpha]_D^{20}$ +33° (*c* 1, $CHCl_3$); R_f 0.41 (solvent B); (Anal. Found: C, 68.13; H, 5.46. $C_{77}H_{70}O_{23}$ requires C, 67.83; H, 5.18); δ_H 1.14 and 1.27 (6 H, 2 × s, Me_2C), 2.36 (1 H, br s, OH), 2.92 (2 H, d, $J_{5',6'}$ 5.6, 6′-H^{a,b}), 3.21 (1 H, t, $J_{1'',2''} = J_{2'',3''}$ 7.5, 2″-H), 3.49 (1 H, br t, 5′-H), 3.65 (1 H, dd, $J_{3'',4''}$ 5.6, 3″-H), 3.84 (1 H, m, 5″-H), 3.88 (1 H, dd, $J_{3',4'}$ 3.2, $J_{2',3'}$ 10.0, 3′-H), 3.95 (1 H, dd, $J_{4'',5''}$ 1.9, 4″-H), 4.01 (1 H, d, 1″-H), 4.02 (2 H, m, PhCH_2), 4.15 (1 H, br d, $J_{4,5}$ 9.6, 5-H), 4.41 (2 H, m, 6″-H^{a,b}), 4.47 (1 H, dd, $J_{5,6a}$ 2.3, $J_{6a,6b}$ 12.1, 6-H^a), 4.58 (1 H, br d, 6-H^b), 4.63 (1 H, t, $J_{3,4} = J_{4,5} = 9.8$, 4-H), 4.78 (1 H, d, $J_{1',2'}$ 7.9, 1′-H), 5.48 (1 H, dd, 2′-H), 5.58 (1 H, d, 4′-H), 5.78 (1 H, dd, $J_{1,2}$ 1.5, $J_{2,3}$ 3.3, 2-H), 5.85 (1 H, dd, 3-H), 6.40 (1 H, d, 1-H) and 6.80–8.10 (40 H, 8 × Ph); δ_C 26.1 and 27.8 (*Me*$_2$C), 61.9 (C-6), 63.1 (C-6″), 67.4 (C-6′), 69.5 (C-2), 69.9 (C-3), 70.0 (C-4′), (C-70.9 (C-5″), 71.8 (C-5), 72.1 (C-2′), 72.3 (C-4), 72.5 (C-2″), 72.8 (C-4″), 73.0 (C-5′), 73.2 (*C*H_2Ph), 77.8 (C-3′), 78.0 (C-3″), 91.1 (C-1), 100.8 (C-1′), 103.2 (C-1″), 110.1 (Me$_2$*C*) 125.0–137.4 (Ph) and 163.9–166.0 (C=O).

2,3,4-Tri-O-benzyl-β-D-arabinopyranosyl-(1→2)-6-O-benzoyl-3,4-O-isopropylidene-β-D-galactopyranosyl-(1→3)-2,4-di-O-benzoyl-6-O-benzyl-β-D-galactopyranosyl-(1→4)-1,2,3,6-tetra-O-benzoyl-α-D-mannopyranose (**22**).—Amorphous solid; $[\alpha]_D^{20}$ −5° (*c* 1, $CHCl_3$); R_f 0.59 (solvent D); (Anal. Found: C, 69.97; H, 5.54. $C_{103}H_{96}O_{27}$ requires C, 70.06; H, 5.48); δ_H 1.18 and 1.32 (6 H, 2 × s, Me_2C), 2.60 (1 H, br d, $J_{5''',5'''b}$ 11.5, 5‴-H^a), 3.04 (2 H, br d, $J_{5',6'}$ 6.1, 6′-H^{a,b}), 3.43–3.58 (4 H, m, 4‴-H, 5′-H, 2″-H, 5‴-H^b), 3.64 (1 H, dd, $J_{2''',3'''}$ 10.2, $J_{3''',4'''}$ 2.8, 3‴-H), 3.73 (1 H, dd, $J_{1''',2'''}$ 3.4, 2‴-H), 3.80–3.90 (2 H, m, 3″-H and 5″-H), 3.96 (1 H, dd, $J_{3'',4''}$ 5.4, $J_{4'',5''}$ 1.9, 4″-H), 3.97–4.12 (4 H, m, 3′-H, 5-H and PhCH_2), 4.23–4.37 (4 H, m, 1″-H, 6-H^a and PhCH_2), 4.45–4.65 (7 H, m, 6-H^b, 6″-H^{ab} and 2 × PhCH_2), 4.60 (1 H, t, $J_{3,4} = J_{4,5}$ 9.6, 4-H), 4.70 (1 H, d, $J_{1',2'}$ 7.9, 1′-H), 5.22 (1 H, d, 1‴-H), 5.46 (1 H, dd, $J_{2',3'}$ 9.9, 2′-H), 5.67 (1 H, d, $J_{3',4'}$ 2.8, 4′-H), 5.77 (1 H, dd, $J_{1,2}$ 1.9, $J_{2,3}$ 3.2, 2-H), 5.81 (1 H, dd, 3-H), 6.36 (1 H, d, 1-H) and 6.80–8.00 (55 H, 11 × Ph); δ_C 26.5 and 27.7 (*Me*$_2$C), 59.6 (C-5‴), 61.9 (C-6), 63.3 (C-6″), 67.5 (C-6′), 69.4 (C-2), 70.1 (C-3), 70.5 (C-4′), 70.9 (C-5″), 71.1 (*C*H_2Ph), 71.8 (C-5), 72.2 (C-2′), 72.3(3) (C-4 and 2 × *C*H_2Ph), 73.1 (C-5′), 73.2 (*C*H_2Ph), 73.3 (C-4″), 74.5 (C-4‴), 74.8 (C-3′), 74.9 (C-2″), 76.0 (C-2‴), 77.2 (C-3‴), 79.7 (C-3″), 91.1 (C-1), 96.1 (C-1‴), 100.8 (C-1″), 101.2 (C-1′), 110.3 (Me$_2$*C*), 127.1–138.8 (Ph) and 164.0–166.1 (C=O).

2,3,4-Tri-O-benzoyl-β-D-arabinopyranosyl-(1→2)-3,4,6-tri-O-benzoyl-β-D-galactopyranosyl-(1→3)-2,4-di-O-benzoyl-6-O-(p,p′-dimethoxytrityl)-β-D-galactopyranosyl-(1→4)-1,2,3,6-tetra-O-benzoyl-α-D-mannopyranose (**5**).—Amorphous solid; $[\alpha]_D^{20}$ +22° (*c* 1, $CHCl_3$); R_f 0.66 (solvent D); (Anal. Found: C, 69.96; H, 4.92. $C_{128}H_{106}O_{34}$ requires C, 70.26; H, 4.88); δ_H 2.50 (1 H, br d, $J_{5'''a,5'''b}$ 12.2, 5‴-H^a),3.17 (1 H, br t, $J_{5',6'} = J_{6'a,6'b}$ 8.9, 6′-H^a), 3.28 (1 H, dd, $J_{5',6'b}$ 5.4, 6′-H^b), 3.51 and 3.57 (2 × 3 H, 2 × s, 2 × OMe), 3.81 (1 H, dd, 5′-H), 4.14–4.23 (3 H, m, 2″-H, 5-H, 5″-H), 4.36 (1 H, br d, 5‴-H^b), 4.38 (1 H m, 6-H^a), 4.39 (1 H, dd, $J_{2',3'}$ 9.8, $J_{3',4'}$ 3.4, 3′-H), 4.64 (2 H, m, 6″-H^{a,b}), 4.68 (1 H, t, $J_{3,4} = J_{4,5}$ 10.0, 4-H), 4.75 (1 H, d, $J_{1'',2''}$ 7.4, 1″-H), 4.84 (1 H, dd, $J_{5,6}$ 5.7, $J_{6a,6b}$ 10.9, 6-H^b), 4.89 (1 H, d, $J_{1',2'}$ 7.9, 1′-H), 5.05 (1 H, dd, $J_{3'',4''}$ 2.9, $J_{2'',3''}$ 10.4, 3″-H), 5.26 (1 H, d, $J_{1''',2'''}$ 4.0, 1‴-H), 5.42 (1 H, dd, $J_{2''',3'''}$ 10.6, 2‴-H), 5.57–5.65 (2 H, m, 2′-H, 4‴-H), 5.70 (1 H, dd, $J_{3''',4'''}$ 3.5, 3‴-H), 5.78 (1 H, d, 4″-H), 5.82–5.87 (2 H, m, 2-H, 3-H), 6.18 (1 H, d, 4′-H), 6.50 (1 H, d, $J_{1,2}$ 1.6, 1-H) and 6.52–8.40 (73 H, 12 × Bz and DMT); δ_C 54.9 and 55.0 (2 × *Me*), 59.3 (C-6′), 60.6 (C-5‴), 61.1 (C-6), 62.2 (C-6″), 67.4 (C-4″), 67.7 (C-3‴), 68.1 (C-2‴), 69.7 (C-2), 69.9(2) (C-3 and C-4′), 70.2 (C-4‴), 70.7 (C-5″), 71.9 (C-2′), 72.1(2) (C-4 and C-5), 72.6 (C-5′), 73.0 (C-2″), 74.2 (C-3″), 74.7 (C-3′), 86.1 (Ph$_3^*$*C*), 91.0 (C-1), 97.3 (C-1‴), 100.8 (C-1″), 101.3 (C-1′), 112.9–136.0 (Ph) and 164.0–166.0 (C=O).

β-D-Arabinopyranosyl-(1→2)-β-D-galactopyranosyl-(1→3)-β-D-galactopyranosyl-(1→4)-α-D-mannopyranose (**1**).—Amorphous solid; $[\alpha]_D^{20}$ −26° (*c* 1, water); δ_H (D_2O) (inter alia) 3.52 (1 H, br d, $J_{5'''a,5'''b}$ 12.7, 5‴-H[a]), 4.09 (1 H, d, $J_{3',4'}$ 3.0, 4′-H), 4.28 (1 H, br d, 5‴-H[b]), 4.38 (1 H, d, $J_{1',2'}$ 7.7, 1′-H), 4.65 (1 H, d, $J_{1',2'}$ 7.7, 1″-H), 5.09 (1 H, s, 1-H), 5.26 (1 H, d, $J_{1''',2'''}$ 3.2, 1‴-H); δ_C (D_2O) 61.41 (C-6), 61.96 (C-6″), 62.17 (C-6′), 64.38 (C-5″), 69.67, 69.76, 69.88, 69.94 and 70.02 (C-2‴, C-3, C-3‴, C-4′and C-4″), 70.02 (C-4‴), 71.25 (C-2′), 71.45 (C-2), 72.13 (C-5), 74.52 (C-3″), 75.96 (C-5″), 76.07 (C-5′), 77.14 (C-4), 77.91 (C-2″), 81.94 (C-3′), 94.86 (C-1), 100.76 (C-1‴), 103.79 (C-1″) and 103.90 (C-1′); β anomer components: 71.70 (C-2), 72.82 (C-3), 76.74 (C-5) and 94.68 (C-1); ES-MS(+): *m*/*z* 242.62 (100%, $[M + 4Na - H]^{3+}$ (expected *m*/*z*, 242.40. $C_{23}H_{40}O_{20}$ requires *M*, 636.21).

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   (b) All attempts to realise the (2 + 2) (i.e., **C**–**D** + **A**–**B**) synthetic scheme using the condensation of 2,3,4-tri-*O*-benzoyl-β-D-arabinopyranosyl-(1→2)-3,4,6-tri-*O*-benzoyl-α-D-galactopyranosyl trichloroacetimidate (as the **C**–**D** unit) and 2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-galactopyranosyl-(1→4)-1,2,3,6-tetra-*O*-benzoyl-α-D-mannopyranose (as the **A**–**B** unit) in propionitrile in the presence of TMSOTf failed providing stereospecifically the corresponding β,α,β-linked tetrasaccharide (64%; $J_{1'',2''}$ 3.4 Hz).
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