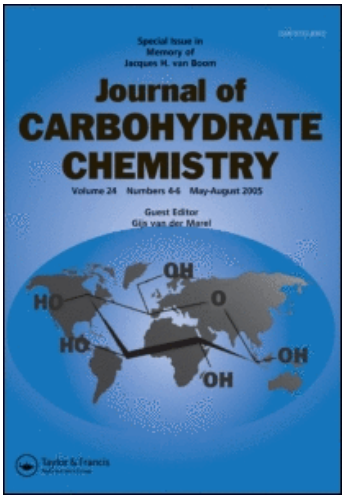

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Synthetic α,β-(1→4)-Glucan Oligosaccharides as Models for Heparan Sulfate. Part II.

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SYNTHETIC α,β-(1→4)-GLUCAN OLIGOSACCHARIDES AS MODELS FOR HEPARAN SULFATE. PART II.

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ABSTRACT

α,β-(1→4)-Glucan oligosaccharides were prepared as models for heparan sulfate with the simplifying assumptions that carboxyl-reduction and sulfation of heparan sulfate does not decrease the SMC antiproliferative activity and that *N*-sulfates in glucosamines can be replaced by *O*-sulfates. The target saccharides were synthesized using maltosyl building blocks. Glycosylation of methyl 2,3,6-tri-*O*-benzyl-α-D-glucopyranoside (**1**) with hepta-*O*-acetyl-α-maltosyl bromide (**2**) furnished trisaccharide **3** which was deprotected to α-D-Glc-(1→4)-β-D-Glc-(1→4)-α-D-Glc(1→OCH_3) (**5**) or, alternatively, converted to the trisaccharide glycosyl acceptor (**8**) with one free hydroxyl function (4''-OH). Further silver triflate mediated glycosylation with glucosyl or maltosyl bromide followed by deblocking gave the tetrasaccharide β-D-Glc-(1→4)-α-D-Glc-(1→4)-β-D-Glc-(1→4)-α-D-Glc-(1→OCH_3) (**11**) and the pentasaccharide [α-D-Glc-(1→4)-β-D-Glc-(1→4)]$_2$-α-D-Glc-(1→OCH_3) (**14**). The trisaccharides **3**, **4**, **6**, and **8** as well as pentasaccharide **12** were fully characterized by ^1H, **3**, **8** and **12** also by ^{13}C NMR spectroscopy. Assignments were possible using 1D TOCSY, in some cases supplemented by 2D T-ROESY, $^1H,^1H$ 2D COSY, and ^1H-detected one-bond and multiple-bond $^1H,^{13}C$ 2D COSY experiments.

$[\rightarrow 4)$-β-D-GlcA-$(1\rightarrow 4)$-α-D-GlcN$]_n$ Heparan sulfate, constitutive backbone

i) ⇓

$[\rightarrow 4)$-β-D-Glc-$(1\rightarrow 4)$-α-D-GlcN$]_n$ CRS-Heparan sulfate

ii) ⇓

$[\rightarrow 4)$-β-D-Glc-$(1\rightarrow 4)$-α-D-Glc$]_n$ α,β-$(1\rightarrow 4)$-Glucan

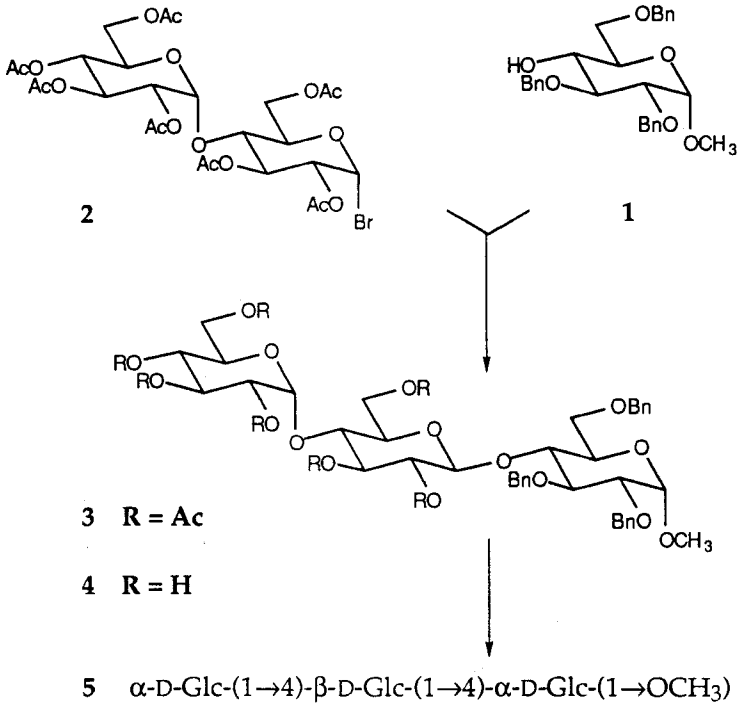
Scheme 1

INTRODUCTION

Heparan sulfates[1,2] isolated from different cells displayed high smooth muscle cell (SMC) antiproliferative activity, an important process in the development of arteriosclerotic lesions,[3,4] and are thus believed to be endogenous regulators of SMC growth. Therefore, the synthesis of substructures is of great interest since there are indications[5,6] that heparan sulfate oligosaccharides distinctly smaller than dodecasaccharides may be biologically active. In a preceding paper[7] we have delineated a strategy towards simplified heparan sulfate structures. With the simplifying assumptions that carboxyl-reduction and sulfation[8] of heparan sulfate does not decrease the SMC antiproliferative activity and that *N*-sulfates in glucosamines can be replaced by *O*-sulfates[9] the relatively complex heparan sulfate structure would be reduced to a regular α,β-$(1\rightarrow 4)$-glucan (Scheme 1). The preparation of tetra- to hexasaccharide structures α-D-Glc-$(1\rightarrow 4)$-β-D-Glc-$(1\rightarrow 4)$-α-D-Glc-$(1\rightarrow 4)$-β-D-Glc-$(1\rightarrow OCH_3)$, $[$β-D-Glc-$(1\rightarrow 4)$-α-D-Glc-$(1\rightarrow 4)]_2$-β-D-Glc-$(1\rightarrow OCH_3)$, and $[$α-D-Glc-$(1\rightarrow 4)$-β-D-Glc-$(1\rightarrow 4)]_2$-α-D-Glc-$(1\rightarrow 4)$-β-D-Glc-$(1\rightarrow OCH_3)$ has been reported.[7] Here we describe the synthesis of the "frame-shifted" oligosaccharides starting with an α-D-glucopyranoside unit.

RESULTS AND DISCUSSION

For a synthetic approach to α,β-$(1\rightarrow 4)$-glucans in a blockwise fashion as chosen before,[7] there would be the possibilities to α-D-link cellobiosyl (β-D-Glc-$(1\rightarrow 4)$-D-Glc) units or to β-D-link maltosyl (α-D-Glc-$(1\rightarrow 4)$-D-Glc) building blocks. While both disaccharides are readily available the construction of β-D-



Scheme 2

linkages is clearly to be preferred over α-D-glucosylation because β-D-glucosides can be prepared with better control of stereochemistry.

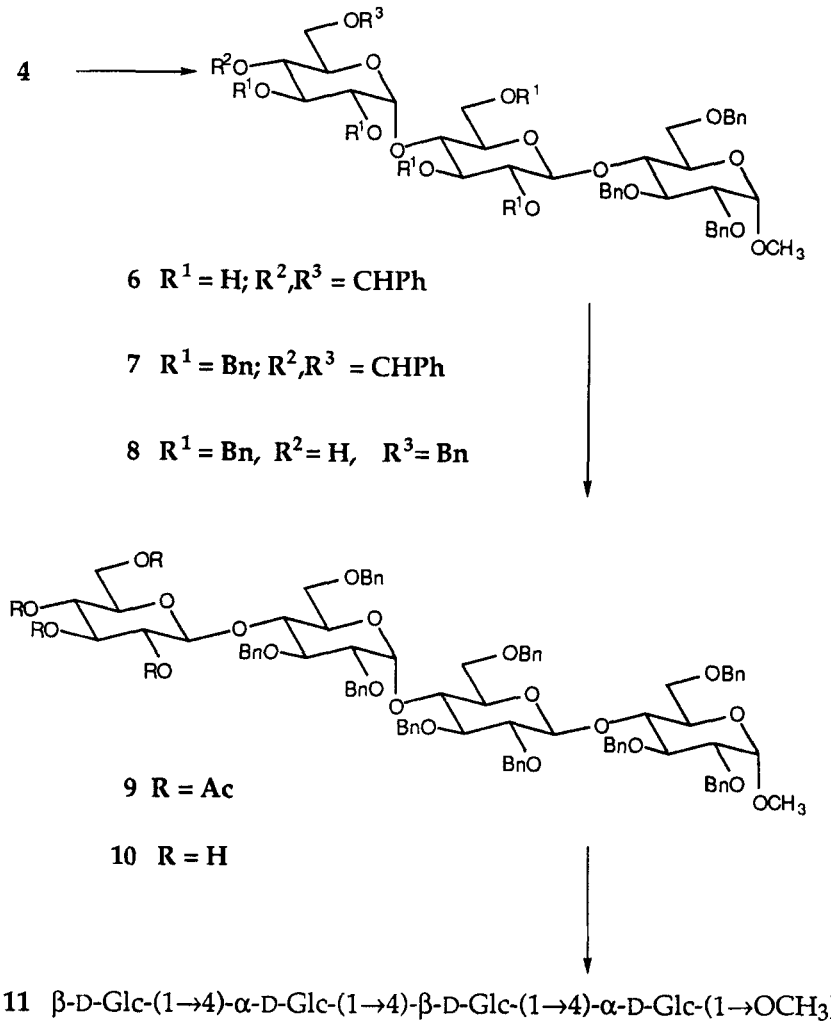
Thus we started with a methyl α-D-monosaccharide at the reducing end to build up oligosaccharides by β-D-glucosylation. The glucoside of choice was methyl 2,3,6-tri-*O*-benzyl-β-D-glucopyranoside (**1**),[10] which has often been used as a model glycosyl acceptor.[11] Silver triflate mediated glycosylation[12] of **1** with hepta-*O*-acetyl-α-maltosyl bromide[13] (**2**) furnished the trisaccharide **3** in 95 % yield. Standard deacetylation of **3** with methanolate gave **4** (79 %), which was further deblocked by hydrogenolysis to give the free tetrasaccharide **5** in 93 % yield.

The protected trisaccharide **3** was fully analyzed by NMR spectroscopy. The proton signals of the three anomeric protons were sufficiently separated so that practically all protons could be readily assigned by a series of 1D TOCSY experiments as described before.[7] The assignment of the sub-spectra to the in-

dividual pyranose rings was straightforward because the added maltosyl moiety is acetylated so that their proton signals are subjected to a characteristic downfield shift. These assignments were supported by the appearance of typical inter-ring cross peaks (C-1''/H-4' and C-1'/H-4) in the multiple-bond $^{1}H,^{13}C$ 2D COSY spectra. These spectra confirmed also the assignments of ring protons and allowed the assignment of the benzylic methylene protons. The typical coupling constant of $J_{1',2'} = 8.1$ Hz proved the diaxial orientation of H-1'/H-2' and thus the β-D-linkage of the newly formed glycosidic bond. The assignment of the ^{13}C signals was derived from $^{1}H,^{13}C$ 2D COSY experiments; in some cases, where protons had very similar chemical shifts (e.g. H-4', H-5'') additional information from the multiple-bond $^{1}H,^{13}C$ 2D COSY spectra was required. The interpretation of the ^{1}H NMR spectrum of the deacetylated trisaccharide **4** in Me_2SO-d_6 was straightforward using 1D TOCSY. Selective inversions of the anomeric protons identified all ring protons plus the hydroxyl protons. The assignments of the hydroxyl protons to the individual rings plus a H-1/OCH_3 ROE led to the unequivocal assignment of the sub-spectra to the individual pyranose rings.

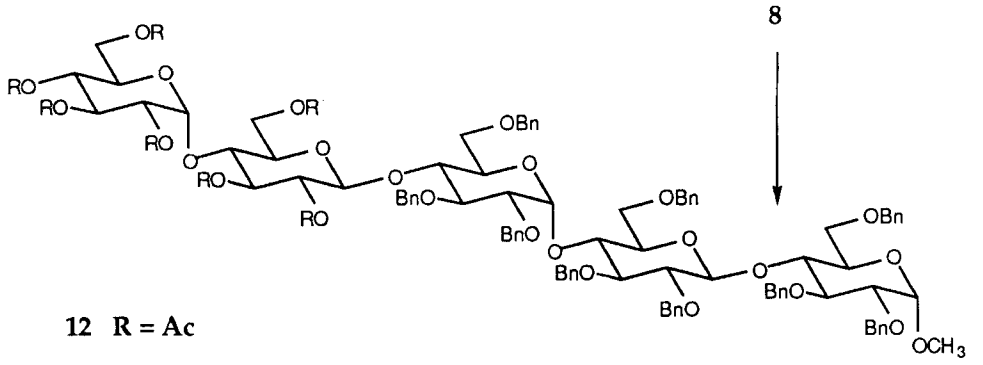
In the build-up of higher glycosides we employed the strategy that was used in the construction of glycosyl acceptor **1**. Thus, trisaccharide **4** was selectively protected in the 4'',6''-positions by benzylidenation with benzaldehyde and zinc chloride to give **6** in a yield of 73 %. The ^{1}H NMR spectrum of this compound was fully interpreted using 1D TOCSY. The remaining hydroxyl groups of **6** were benzylated with sodium hydride and benzyl chloride in Me_2SO to furnish **7** in very good yield (91 %). The benzylidene acetal of **7** was opened regioselectively with sodium cyanoborohydride in the presence of hydrochloric acid in diethyl ether as described by Garegg et al.[19] to produce **8** in good yield (88 %).

Trisaccharide **8** was completely analyzed by NMR spectroscopy. H-1'' was readily identified by its characteristic downfield shift. The other anomeric protons were, however, not unambiguously identified in the ^{1}H NMR due to severe overlap with benzylic protons, and there were also no other well isolated protons to be used in 1D TOCSY experiments. Thus, the approximate chemical shifts of H-1 and H-1' were extracted from a one-bond $^{1}H,^{13}C$ 2D correlation experiment. In the ^{13}C NMR spectrum, the signals of the anomeric carbons absorb between 96.77 and 102.15 ppm, well separated from all other signals. The ^{1}H shifts of their directly attached protons were then readily accessible from the position of the cross peaks in the 2D



Scheme 3

spectrum. This considerably simplified their identification in the ^1H spectrum. Starting from the anomeric protons, the subsequent 1D TOCSY experiments performed with increasing mixing times generated three sets of sub-spectra of the different rings leading to the identification of the proton signals, except H-6b. The assignment of the sub-spectra to the individual pyranose rings was deduced from inter-ring cross peaks (H-1"/H-4', H-1'/H-4, H-1/OCH_3) in the 2D T-ROESY experiment. These results were furthermore confirmed through corresponding cross peaks (C-1/OCH_3, OCH_3/H-1, C-4/H-



12 R = Ac

13 R = H

14 [α-D-Glc-(1→4)-β-D-Glc-(1→4)]$_2$-α-D-Glc-(1→OCH_3)

Scheme 4

1', C-1'/H-4, C-4'/H-1'', C-1''/H-4') in the multiple-bond $^1H,^{13}C$ 2D COSY experiment caused by three-bond $^1H,^{13}C$ couplings. This experiment also allowed the assignment of the benzylic methylene protons as well as the remaining ring proton. Further confirmation of the assignment of some protons was provided by a $^1H,^1H$ 2D COSY experiment.

The trisaccharide **8** has a single free hydroxyl function (4''-OH) ready for glycosylation and was used as a glycosyl acceptor in the following reactions. Koenigs-Knorr glucosylation of **8** with tetra-*O*-acetyl-α-D-glucosyl bromide and silver triflate[12] as promoter furnished tetrasaccharide **9** in a good yield (83 %). The structure of this compound was supported by a FAB mass spectrum and the acetyl peaks in the ^1H NMR spectrum. Standard deprotection of **9** with methanolate to give **10** (91 % yield) followed by hydrogenation furnished the free tetrasaccharide **11** in very good yield (93 %). The two protons with diaxial couplings (J = 7.9 Hz) confirmed the β-D-linkage of the newly added glucoside.

Analogous silver triflate mediated glycosylation of trisaccharide **8** with maltosyl bromide **2** furnished pentasaccharide **12**, again in good yield (89 %).

An FAB mass spectrum supported the structure and, moreover, the compound was extensively analyzed by NMR spectroscopy. While H-1'' was identified by virtue of its characteristic downfield shift and also H-1'''' was well isolated, the location of the remaining anomeric protons was extracted from a one-bond $^{1}H,^{13}C$ 2D correlation experiment as described for trisaccharide **8**. The axial protons H-1' and H-1''' were identified by the width of the cross peaks and the typical $^{1}J_{C,H}$ one-bond coupling constants. Now, all ring protons could be identified by 1D TOCSY. Cross peaks in the 2D T-ROESY experiment and in the multiple-bond $^{1}H,^{13}C$ correlation revealed the assignment of the five sub-spectra to the individual pyranose rings. Furthermore, the assignments of benzylic methylene protons were taken from the multiple-bond $^{1}H,^{13}C$ correlation experiment.

Deprotection of **12** as described above for **9** led to the partially and then fully deblocked pentasaccharides **13** and **14**, respectively in a yield of 91 %. The correct β-D-stereochemistry at the newly formed anomeric center C-1''' was evident from the ^{1}H NMR spectrum displaying the presence of two anomeric protons (H-1', H-1''') with the typical coupling constants for diaxial protons.

In conclusion, three α,β-(1→4)-glucan oligosaccharides, namely trisaccharide **5**, tetrasaccharide **11**, and pentasaccharide **14**, were effectively prepared in a block synthesis approach.

EXPERIMENTAL

General Procedures. Solvents and reagents were bought from Fluka. Evaporation: *in vacuo*, conducted with Büchi rotary evaporator. TLC: precoated silica gel 60F-254 plates (Merck), detection by UV light (254 nm) and spraying with a 10% solution of concentrated sulfuric acid in methanol followed by heating. Specific rotations: Perkin-Elmer Polarimeter 241, measured at 20 °C. ^{1}H NMR: Bruker AC 250 (250 MHz), AM-400 (400 MHz) with Aspect 3000, ARX-400 (400 MHz) with ASPECT station 1 and z-gradient accessory kit with 10 Amps power amplifier for pulsed field z-gradient (PFG) experiments, AMX2-600 (600 MHz) with ASPECT station 1; chemical shifts in ppm relative to tetramethylsilane or sodium 2,2,3,3-tetradeutero-3-(trimethylsilyl)propionate as internal standard. Standard Bruker pulse programs were applied for the PFG $^{1}H,^{1}H$ 2D COSY and the ^{1}H-detected multiple-bond $^{1}H,^{13}C$ HMQC correlation experiments (pulse sequences

COSYGS and INV4GSLRLP). The one-bond PFG $^{1}H,^{13}C$ HSQC experiment was as described recently.[14] Pulse sequences and experimental conditions for the 1D TOCSY experiments with selective excitation by a sequence of DANTE pulses and for the 2D T-ROESY experiments with $(180°_{x} - 180°_{-x})_{n}$ spin-lock of 0.6 s duration (n = 2400) were essentially as described before.[15,16]

Methyl *O*-(2,3,4,6-Tetra-*O*-acetyl-α-D-glucopyranosyl)-(1→4)-*O*-(2,3,6-tri-*O*-acetyl-β-D-glucopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-α-D-glucopyranoside (3). To a soln of glycosyl acceptor **1**[10] (26.0 g, 59.0 mmol) and acetobromomaltose **2**[13] (58.8 g, 84 mmol) in abs dichloromethane (360 mL) was added tetramethylurea (14.6 g, 126 mmol) and silver triflate (21.6 g, 84 mmol) at -10 °C. The reaction mixture was stirred at rt for 1 h and at 30-32 °C for 3 h. A second addition of **2** (5.88 g, 8.4 mmol) and silver triflate (2.16 g, 8.4 mmol) was made and stirring was continued for 1 h. Then the reaction mixture was cooled to 20 °C and filtered through a pad of filter aid. The filtrate and dichloromethane washings were combined and washed twice with aq sodium bicarbonate soln. The organic phases were dried over magnesium sulfate and concentrated. The residue was chromatographed on silica gel using ethyl acetate/ hexane 1:2 and 1:1 as eluents to furnish pure **3** (71.0 g, 95 %) as a colourless foam: $[\alpha]_{D}^{20}$ +44.0 ° (*c* 0.5, chloroform); MS (ionspray) *m/z* 1100.7 (96%, $[M + NH_4]^+$); ^{1}H NMR ($CDCl_3$, 400 MHz; 1D TOCSY, $^{1}H,^{13}C$ 2D COSY) δ 7.48 - 7.26 (m, 15H, aromat), 5.39 (dd ~ t, 1H, $J_{3'',4''}$ = 9.5 Hz, H-3''), 5.33 (d, 1H, $J_{1'',2''}$ = 3.9 Hz, H-1''), 5.06 (dd ~ t, 1H, $J_{4'',5''}$ = 10.3 Hz, H-4''), 5.03 (dd ~ t, 1H, $J_{2',3'}$ = 9.3 Hz, H-3'), 4.96, 4.71 (2d, 2H, J_{gem} = 11.6 Hz, C-3-CH_2Ph), 4.86 (dd, 1H, $J_{2'',3''}$ = 10.6 Hz, H-2''), 4.78, 4.42 (2d, 2H, J_{gem} = 12.0 Hz, C-6-CH_2Ph), 4.72 (~dd, 1H, H-2'), 4.71, 4.57 (2d, 2H, J_{gem} = 12.0 Hz, C-2-CH_2Ph), 4.57 (d, 1H, $J_{1,2}$ = 3.7 Hz, H-1), 4.46 (d, 1H, $J_{1',2'}$ = 8.1 Hz, H-1'), 4.22 (dd, 1H, $J_{5'',6a''}$ = 3.8 Hz, $J_{6a'',6b''}$ = 12.6 Hz, H-6a''), 4.14 (dd, 1H, $J_{5',6a'}$ = 2.8 Hz, $J_{6a',6b'}$ = 12.0 Hz, H-6a'), 4.06 (dd, 1H, $J_{5',6b'}$ = 3.9 Hz, H-6b'), 3.98 (dd, 1H, $J_{5'',6b''}$ = 2.2 Hz, H-6b''), 3.88 (dd ~ t, 1H, $J_{3',4'}$ = 8.8 Hz, H-4'), 3.88 (~ddd, 1H, H-5''), 3.86 (dd ~ t, 1H, $J_{4,5} \approx$ 9 Hz, H-4), 3.80 (dd ~ t, 1H, $J_{3,4} \approx$ 8.9 Hz, H-3), 3.79 (dd, 1H, $J_{5,6a}$ = 2.8 Hz, $J_{6a,6b}$ = 10.5 Hz, H-6a), 3.62 (dd, 1H, $J_{5,6b}$ = 1.5 Hz, H-6b), 3.60 (ddd ~ dt, 1H, H-5), 3.46 (dd, 1H, $J_{2,3}$ = 9.1 Hz, H-2), 3.36 (s, 3H, OCH_3), 3.15 (ddd ~ dt, 1H, $J_{4',5'}$ = 9.6 Hz, H-5'), 2.10, 2.08, 2.04, 2.02, 1.97 (5s, 15H, OAc), 1.95 (s, 6H, OAc); ^{13}C NMR ($CDCl_3$, 100 MHz; $^{1}H,^{13}C$ 2D COSY) δ 170.51 (3C, C-2''-, C-6'-, C-6''-C=O), 170.27 (C-3'-C=O), 169.95 (C-3''-C=O), 169.53 (C-2'-C=O), 169.41 (C-4''-C=O), 139.50, 138.22, 137.70 (3C, quat. C arom.), 128.79 - 127.16 (aromatic CH), 99.44 (C-1'), 98.44 (C-1), 95.66 (C-1''), 79.94 (C-3), 78.72 (C-2), 77.03 (C-4), 75.93 (C-3'), 74.98

(C-3-CH_2Ph), 73.76 (C-6-CH_2Ph), 73.52 (C-2-CH_2Ph), 72.92 (C-4'), 72.71 (C-2'), 71.58 (C-5'), 70.04 (C-2''), 69.68 (C-5), 69.26 (C-3''), 68.50 (C-5''), 67.90 (C-4''), 67.70 (C-6), 63.05 (C-6'), 61.35 (C-6''), 55.41 (OCH_3), 20.93, 20.74, 20.69 (3C, Ac), 20.63 (4C, Ac).

Anal. Calcd for $C_{54}H_{66}O_{23}$: C, 59.88; H, 6.14. Found: C, 59.77; H, 6.08.

Methyl *O*-(α-D-Glucopyranosyl)-(1→4)-*O*-(β-D-glucopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-α-D-glucopyranoside (**4**). To a soln of **3** (60.5 g, 55 mmol) in diethyl ether (120 mL) and methanol (500 mL) was a added a soln of sodium methanolate (30 mL of 2 g Na/ 100 mL methanol) at rt. The reaction mixture was kept for 4 h at rt, neutralized with Amberlite IR 120 (H^+) and filtered. After addition of a few drops of triethylamine, the filtrate and methanol washings were concentrated. The residue was chromatographed on silica gel using ethyl acetate/ methanol/ water 94:4:2 and 93:5:3 as eluents to obtain pure **4** (34.9 g, 79 %) as a colourless amorphous powder: $[\alpha]_D^{20}$ +97.4 ° (*c* 0.4, methanol); MS (ionspray) *m/z* 806.6 (76%, $[M + NH_4]^+$); ^1H NMR (DMSO-d_6, 400 MHz; 1D TOCSY) δ 7.43 - 7.21 (m, 15H, aromat), 5.50 (d, 1H, $J_{H\text{-}3',3'\text{-}OH}$ = 2.3 Hz, 3'-OH), 5.44 (d, 1H, $J_{H\text{-}2'',2''\text{-}OH}$ = 6.0 Hz, 2''-OH), 5.30 (d, 1H, $J_{H\text{-}2',2'\text{-}OH}$ = 4.6 Hz, 2'-OH), 5.03 (d, 1H, $J_{1'',2''}$ = 3.8 Hz, H-1''), 4.94, 4.65 (2d, 2H, J_{gem} = 10.8 Hz, CH_2Ph), 4.92, 4.91 (2d, 2H, 3''-OH, 4''-OH), 4.78 (d, 1H, $J_{1,2}$ = 3.4 Hz, H-1), 4.59 (s, 2H, CH_2Ph), 4.57 (dd ~ t, 1H, $J_{H\text{-}6'',6''\text{-}OH}$ = 5.6 Hz, 6''-OH), 4.54, 4.49 (2d, 2H, J_{gem} = 12.0 Hz, CH_2Ph), 4.33 (d, 1H, $J_{1',2'}$ = 7.9 Hz, H-1'), 4.18 (dd ~ t, 1H, $J_{H\text{-}6',6'\text{-}OH}$ = 5.4 Hz, 6'-OH), 3.93 (dd, 1H, $J_{5,6a}$ = 3.9 Hz, $J_{6a,6b}$ = 10.8 Hz, H-6a), 3.79 (dd ~ t, 1H, $J_{4,5} \approx 9.6$ Hz, H-4), 3.72 (dd ~ br d, 1H, $J_{5,6b} \leq 1.5$ Hz, H-6b), 3.69 (dd ~ t, 1H, $J_{3,4} \approx 8.9$ Hz, H-3), 3.67 (~ddd, 1H, H-6a'), 3.65 (~ ddd, 1H, H-6a''), 3.55 (ddd ~ dt, 1H, $J_{5',6b'}$ = 5.1 Hz, $J_{6a',6b'}$ = 11.5 Hz, H-6b'), 3.49, 3.45 (2 m_c, 2H, H-5'', H-6b''), 3.41 (dd, 1H, $J_{2,3}$ = 9.2 Hz, H-2), 3.38 (ddd, 1H, H-3''), 3.36 (m_c, 2H, H-3', H-4'), 3.28 (s, 3H, OCH_3), 3.25 (ddd, 1H, $J_{2'',3''}$ = 9.2 Hz, H-2''), ~3.08 (1H, H-5'), 3.07 (ddd ~ dt, 1H, H-4''), 3.04 (ddd ~ dt, 1H, $J_{2',3'}$ = 9.1 Hz, H-2').

Anal. Calcd for $C_{40}H_{52}O_{16}$: C, 60.90; H, 6.64. Found: C, 60.82; H, 6.43.

Methyl *O*-(α-D-Glucopyranosyl)-(1→4)-*O*-(β-D-glucopyranosyl)-(1→4)-α-D-glucopyranoside (**5**). A soln of **4** (2.95 g, 3.7 mmol) in ethanol (80 mL) and water (20 mL) was hydrogenated in the presence of 10 % palladium on charcoal (1.0 g) at 1.1 bar for 4 h. The reaction mixture was filtered through a pad of filter aid and washed with ethanol/ water 1:1. After addition of a few drops of triethylamine the filtrate was concentrated. The aqueous residue was crystallized from ethanol/ ethyl acetate to obtain pure **5** (1.78 g, 93 %) as colourless crystals: mp 259 °C (decomp); $[\alpha]_D^{20}$ +143.8 ° (*c* 0.5, water); MS

(ionspray) *m/z* 541.4 (100%, $[M + Na]^+$); ^1H NMR (D_2O, 400 MHz) δ 5.42 (d, 1H, $J_{1'',2''}$ = 3.2 Hz, H-1''), ~4.81 (under HOD signal, H-1), 4.53 (d, 1H, $J_{1',2'}$ = 7.9 Hz, H-1'), 3.43 (s, 3H, OCH_3).

Anal. Calcd for $C_{19}H_{34}O_{16}$: C, 44.02; H, 6.61. Found: C, 43.88; H, 6.71.

Methyl *O*-[4,6-*O*-(*R*)-Benzylidene-α-D-glucopyranosyl]-(1→4)-*O*-(β-D-glucopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-α-D-glucopyranoside (6). A suspension of **4** (31.5 g, 40.0 mmol) in benzaldehyde (530 mL) was stirred in the presence of zinc chloride (126 g) for 20 h at rt and then poured into a stirred mixture of hexane (3 L) and ice water (700 mL). The organic phase was decanted. The aqueous phase and the syrupy residue were extracted twice with ethyl acetate. The extracts were washed with brine, dried over magnesium sulfate, and concentrated. The residue was chromatographed on silica gel using ethyl acetate as eluent. Product fractions were crystallized from ethyl acetate to furnish pure **6** (25.4 g, 73 %) as colourless crystals: mp 184-185 °C; $[\alpha]_D^{20}$ +98.4 ° (*c* 0.5, methanol); MS (ionspray) *m/z* 894.6 (100%, $[M + NH_4]^+$); ^1H NMR (DMSO-d_6, 400 MHz; 1D TOCSY in DMSO-d_6, trace D_2O) δ 7.47-7.24 (m, 15H, aromat), 5.65 (d, 1H, $J_{H\text{-}2'',2''\text{-}OH}$ = 6.5 Hz, 2''-OH), 5.59 (s, 1H, CHPh), 5.51 (d, 1H, $J_{H\text{-}3',3'\text{-}OH}$ = 2.9 Hz, 3'-OH), 5.33 (d, 1H, $J_{H\text{-}3'',3''\text{-}OH}$ = 5.0 Hz, 3''-OH), 5.32 (d, 1H, $J_{H\text{-}2',2'\text{-}OH}$ = 5.0 Hz, 2'-OH), 5.13 (d, 1H, $J_{1'',2''}$ = 3.9 Hz, H-1''), 4.94, 4.65 (2d, 2H, J_{gem} = 11.0 Hz, CH_2Ph), 4.79 (d, 1H, $J_{1,2}$ = 3.4 Hz, H-1), 4.60 (s, 2H, CH_2Ph), 4.55, 4.49 (2d, 2H, J_{gem} = 11.5 Hz, CH_2Ph), 4.38 (dd ~ t, 1H, $J_{H\text{-}6',6'\text{-}OH}$ = 5.1 Hz, 6'-OH), 4.35 (d, 1H, $J_{1',2'}$ = 7.8 Hz, H-1'), 4.15 (dd, 1H, $J_{5'',6a''}$ = 3.8 Hz, $J_{6a'',6b''}$ = 9.0 Hz, H-6a''), 3.94 (dd, 1H, $J_{5,6a}$ = 4.0 Hz, $J_{6a,6b}$ = 11.0 Hz, H-6a), 3.81 (dd ~ t, 1H, $J_{4,5}$ = 9.0 Hz, H-4), 3.74 (ddd ~ dt, 1H, H-5''), 3.72 (dd ~ br d, 1H, H-6b), 3.69 (2H, dd ~ t, H-3, H-6b''), 3.68 (ddd, 1H, H-6a'), 3.65 (ddd ~ br dt, 1H, H-5), 3.58 (ddd ~ dt, 1H, $J_{3'',4''} \approx 9.5$ Hz, $J_{H\text{-}3'',3''\text{-}OH}$ = 5.0 Hz, H-3''), 3.49 (ddd ~ dt, 1H, $J_{5',6b'}$ = 5.4 Hz, $J_{6a',6b'}$ = 11.0 Hz, H-6b'), 3.42 (dd, 1H, $J_{2,3}$ = 9.8 Hz, H-2), 3.29 (2H, ddd, $J_{2'',3''} \approx 9$ Hz, H-2''; dd ~ t, H-4''), 3.38 (m_C, 2H, H-3', H-4'), 3.29 (s, 3H, OCH_3), 3.08 (br ddd, 1H, H-5'), 3.05 (m_C, 1H, H-2').

Anal. Calcd for $C_{47}H_{56}O_{16}$: C, 64.37; H, 6.44. Found: C, 64.30; H, 6.55.

Methyl *O*-[2,3-Di-*O*-benzyl-4,6-*O*-(*R*)-benzylidene-α-D-glucopyranosyl]-(1→4)-*O*-(2,3,6-tri-*O*-benzyl-β-D-glucopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-α-D-glucopyranoside (7). A soln of **6** (21.9 g, 25.0 mmol) in Me_2SO (110 mL) was added dropwise within 45 min to a slurry of sodium hydride (7.5 g, 187.5 mmol, 60 % in mineral oil, washed with hexane) in Me_2SO (225 mL). Hydrogen evolved while stirring for 2.5 h at 20 25 °C. Benzyl chloride (25.3 g,

200 mmol) in Me_2SO (50 mL) was added dropwise within 30 min at rt and stirring was continued for 2 h. The reaction mixture was then poured on ice/water and extracted twice with ethyl acetate. The organic phases were washed with cold water and brine, dried over magnesium sulfate, and concentrated. The residue was chromatographed on silica gel using ethyl acetate/ hexane 2:1, 3:1, and 1:3 as eluents to give **7** (30.3 g, 91 %) as a foam; $[\alpha]_D^{20}$ +18.6 ° (*c* 0.5, chloroform); MS (FAB) *m/z* 1365.8 (100%, $[M + K]^+$), 1349.8 (85%, $[M + Na]^+$); ^1H NMR ($CDCl_3$, 400 MHz) δ 7.51-7.48 (m, 2H, aromat), 7.40-7.06 (m, 43H, aromat), 5.66 (d, 1H, $J_{1'',2''}$ = 3.7 Hz, H-1''), 5.53 (s, 1H, CHPh), 4.57 (d, 1H, $J_{1,2}$ = 3.6 Hz, H-1), 4.34 (d, 1H, $J_{1',2'}$ = 7.8 Hz, H-1'), 3.35 (s, 3H, OCH_3).

Anal. Calcd for $C_{82}H_{86}O_{16}$: C, 74.19; H, 6.53. Found: C, 74.26; H, 6.54.

Methyl *O*-(2,3,6-Tri-*O*-benzyl-α-D-glucopyranosyl)-(1→4)-*O*-(2,3,6-tri-*O*-benzyl-β-D-glucopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-α-D-glucopyranoside (8). To a soln of **7** (30.0 g, 22.6 mmol) in abs tetrahydrofuran (250 mL) were added 3Å pulverized molecular sieves (10 g) at 0 °C followed by sodium cyanoborohydride (8.9 g, 120 mmol) and a few crystals of methyl orange. Hydrogen chloride in diethyl ether (150 mL of 1.2 m soln, 180 mmol) was added dropwise to the milky reaction mixture within 30 min at rt. After stirring for 1 h at 15 - 20 °C the orange-red reaction mixture was poured into sodium bicarbonate soln, and tetrahydrofuran was evaporated under reduced pressure. The aqueous residue was extracted with ethyl acetate. The organic phases were washed with brine, dried over magnesium sulfate, and concentrated. The residue was chromatographed on silica gel using ethyl acetate/ hexane 1:3 and 1:2 as eluents to give **8** (26.4 g, 88 %) as a colourless syrup; $[\alpha]_D^{20}$ +25.2 ° (*c* 0.5, chloroform); MS (ionspray) *m/z* 1347.8 (4%, $[M + NH_4]^+$); ^1H NMR ($CDCl_3$, 400 MHz; 1D TOCSY, 2D T-ROESY, $^1H,^1H$-COSY, $^1H,^{13}C$ 2D COSY) δ 7.39-7.37 (m, 2H, aromat), 7.32-7.15 (m, 43H, aromat), 5.64 (d, 1H, $J_{1'',2''}$ = 3.6 Hz, H-1''), 5.05, 4.76 (2 d, 2H, J_{gem} = 11.5 Hz, C-3-CH_2Ph), 4.90, 4.73 (2 d, 2H, J_{gem} = 11.2 Hz, C-3''-CH_2Ph), 4.87, 4.77 (2 d, 2H, J_{gem} = 11.5 Hz, C-3'-CH_2Ph), 4.77, 4.60 (2 d, 2H, J_{gem} = 12.0 Hz, C-2''-CH_2Ph), 4.74, 4.61 (2 d, 2H, J_{gem} = 11.0 Hz, C-2'-CH_2Ph), 4.61, 4.39 (2 d, 2H, J_{gem} ≈ 12 Hz, C-6-CH_2Ph), 4.59, 4.53 (2 d, 2H, J_{gem} = 11.8 Hz, C-2-CH_2Ph), 4.57 (d, 1H, H-1), 4.48, 4.31 (2 d, 2H, J_{gem} = 12.0 Hz, C-6'-CH_2Ph), 4.47, 4.38 (2 d, 2H, J_{gem} = 12.0 Hz, C-6''-CH_2Ph), 4.37 (d, 1H, H-1'), 4.01 (dd ~ t, 1H, $J_{3',4'}$ = 8.0 Hz, H-4'), 3.97 (dd ~ t, 1H, $J_{4,5}$ = 10.3 Hz, H-4), 3.83 (dd ~ t, 1H, $J_{3,4}$ = 8.2 Hz, H-3), 3.80 (dd, 1H, $J_{5,6a}$ ≈ 2.4 Hz, $J_{6a,6b}$ = 10.8 Hz, H-6a), 3.77 (dd ~ t, 1H, $J_{3'',4''}$ = 8.4 Hz, H-3''), 3.75 (2H, ~ddd, 1H, H-5''; dd, $J_{5',6a'}$ = 1.8 Hz, H-6a'), 3.67 (dd, 1H, $J_{5',6b'}$ = 4.2 Hz, $J_{6a',6b'}$ =

11.8 Hz, H-6b'), 3.64 (dd ~ t, 1H, $J_{4'',5''} \approx 9.6$ Hz, H-4''), 3.56 (2H, dd ~ t, $J_{2',3'} \approx$ 9.0 Hz, H-3'; ddd ~ br d, H-5), 3.55 (dd, 1H, $J_{5'',6a''} = 3.6$ Hz, $J_{6a'',6b''} = 10.4$ Hz, H-6a''), 3.47 (3H, dd, H-6b; dd, $J_{5'',6b''} = 4.0$ Hz, H-6b''; dd, $J_{1,2} = 3.5$ Hz, $J_{2,3} =$ 9.8 Hz, H-2), 3.44 (dd, 1H, $J_{2'',3''} = 9.7$ Hz, H-2''), 3.39 (dd, 1H, $J_{1',2'} \approx 7.8$ Hz, H-2'), 3.35 (s, 3H, OCH_3), 3.32 (ddd, 1H, $J_{4',5'} = 9.5$ Hz, H-5'), 2.44 (br s, 1H, 4''-OH); ^{13}C NMR ($CDCl_3$, 100 MHz; DEPT, ^1H,^{13}C 2D COSY) δ 139.50, 138.81 (2C, quat. C arom.), 138.78 (2C, quat. C arom.), 138.40 (2C, quat. C arom.), 138.04, 137.97, 137.80 (3C, quat. C arom.), 128.53 - 126.67 (aromatic CH), 102.15 (C-1'), 98.44 (C-1), 96.77 (C-1''), 84.85 (C-3'), 82.67 (C-2'), 81.25 (C-3''), 80.25 (C-3), 79.01 (C-2''), 78.92 (C-2), 76.20 (C-4), 75.34 (C-3-CH_2Ph), 75.28 (C-4-CH_2Ph), 74.90 (C-5'), 74.76 (CH_2Ph), 73.95 (C-3'-CH_2Ph), 73.62, 73.57, 73.46, 73.19, 73.16 (5C, 5 CH_2Ph), 73.15 (C-4'), 71.54 (C-4''), 70.58 (C-5''), 69.99 (C-5), 69.72 (C-6''), 69.31 (C-6'), 67.84 (C-6), 55.31 (OCH_3).

Anal. Calcd for $C_{82}H_{88}O_{16}$: C, 74.07; H, 6.67. Found: C, 73.95; H, 6.58.

Methyl *O*-(2,3,4,6-Tetra-*O*-acetyl-β-D-glucopyranosyl)-(1→4)-*O*-(2,3,6-tri-*O*-benzyl-α-D-glucopyranosyl)-(1→4)-*O*-(2,3,6-tri-*O*-benzyl-β-D-glucopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-α-D-glucopyranoside (9). To a soln of glycosyl acceptor **8** (7.45 g, 5.6 mmol) and acetobromoglucose (3.7 g, 9.0 mmol) in abs dichloromethane (40 mL) was added tetramethylurea (1.85 g, 16 mmol) and silver triflate (2.32 g, 9.0 mmol) at -10 °C. The reaction mixture was stirred for 1.5 h at rt and for 5 h at 30 - 35 °C and was then filtered through a pad of filter aid. The filtrate and dichloromethane washings were combined and washed twice with aq sodium bicarbonate soln. The organic phases were dried over magnesium sulfate and concentrated. The residue was chromatographed on silica gel using ethyl acetate/ hexane 1:2 and 1:1 as eluents to furnish **9** (7.7 g, 83 %) as a colourless foam: $[\alpha]_D^{20}$ +17.4 ° (*c* 0.5, chloroform); MS (FAB) *m/z* 1682.7 (90%, $[M + Na]^+$); ^1H NMR ($CDCl_3$, 400 MHz) δ 7.39-7.09 (m, 45H, aromat), 5.66 (d, 1H, $J_{1'',2''} = 3.8$ Hz, H-1''), 3.34 (s, 3H, OCH_3), 2.00, 1.97, 1.90, 1.82 (4 s, 12H, Ac).

Anal. Calcd for $C_{96}H_{106}O_{25}$: C, 69.47; H, 6.44. Found: C, 69.30; H, 6.37.

Methyl *O*-(β-D-Glucopyranosyl)-(1→4)-*O*-(2,3,6-tri-*O*-benzyl-α-D-glucopyranosyl)-(1→4)-*O*-(2,3,6-tri-*O*-benzyl-β-D-glucopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-α-D-glucopyranoside (10). To a soln of **9** (7.62 g, 4.6 mmol) in dimethoxyethane (18 mL) and methanol (90 mL) was added a soln of sodium methanolate (4.0 mL of 2.0 g Na/ 100 mL methanol) at rt. The reaction mixture was kept for 16 h at rt, neutralized with Amberlite IR 120 (H^+) and filtered. After addition of a few drops of triethylamine, the filtrate and

methanol washings were concentrated. The residue was chromatographed on silica gel using ethyl acetate/ hexane 2:1 and ethyl acetate as eluent to obtain pure **10** (6.3 g, 91 %) as a colourless foam: $[\alpha]_D^{20}$ +44.6 ° (*c* 0.5, chloroform); MS (FAB) *m/z* 1513.6 (70%, [M + Na]$^+$); ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.38-7.12 (m, 45H, aromat), 5.66 (d, 1H, $J_{1'',2''}$ = 3.5 Hz, H-1''), 3.45 (s, 3H, OCH_3).

Anal. Calcd for $C_{88}H_{98}O_{21}$: C, 70.86; H, 6.62. Found: C, 71.00; H, 6.67.

Methyl *O*-(β-D-Glucopyranosyl)-(1→4)-*O*-(α-D-glucopyranosyl)-(1→4)-*O*-(β-D-glucopyranosyl-(1→4)-α-D-glucopyranoside (11). A soln of **10** (2.53 g, 1.7 mmol) in ethanol (75 mL) and water (15 mL) was hydrogenated in the presence of 10 % palladium on charcoal (800 mg) at 1.1 bar for 6 h. The reaction mixture was filtered through a pad of filter aid and washed with ethanol water 1:1. After addition of a few drops of triethylamine the filtrate was concentrated. The aqueous residue (ca 10 mL) was lyophilized to obtain pure **11** (1.08 g, 93 %) as an amorphous colourless powder; $[\alpha]_D^{20}$ +113.4 ° (*c* 0.5, water); MS (FAB) *m/z* 703.3 (95%, [M + Na]$^+$), 681.3 (40%, [M + H]$^+$); ^{1}H NMR (D_2O, 400 MHz) δ 5.42 (d, 1H, $J_{1'',2''}$ = 3.9 Hz, H-1''), ~4.82 (under HOD signal, H-1), 4.53, 4.52 (2d, 2H, $J_{1',2'} = J_{1''',2'''}$ = 7.9 Hz, H-1', H-1'''), 3.43 (s, 3H, OCH_3).

Anal. Calcd for $C_{25}H_{44}O_{21}$: C, 44.12; H, 6.52. Found: C, 41.87; H, 6.63.

Methyl *O*-(2,3,4,6-Tetra-*O*-acetyl-α-D-glucopyranosyl)-(1→4)-*O*-(2,3,6-tri-*O*-acetyl-β-D-glucopyranosyl)-(1→4)-*O*-(2,3,6-tri-*O*-benzyl-α-D-glucopyranosyl)-(1→4)-*O*-(2,3,6-tri-*O*-benzyl-β-D-glucopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-α-D-glucopyranoside (12). To a soln of glycosyl acceptor **8** (7.45 g, 5.6 mmol) and acetobromomaltose[13] (6.30 g, 9.0 mmol) in abs dichloromethane (40 mL) was added tetramethylurea (1.85 g, 16 mmol) and silver triflate (2.32 g, 9.0 mmol) at -10 °C. The reaction mixture was stirred at rt for 1 h and at 30-35 °C for 5 h, and then filtered through a pad of filter aid. The filtrate and dichloromethane washings were combined and washed twice with aq sodium bicarbonate soln. The organic phases were dried over magnesium sulfate and concentrated. The residue was chromatographed on silica gel using ethyl acetate/ hexane 1:2 and 1:1 as eluents to furnish **12** (9.7 g, 89 %) as a colourless foam: $[\alpha]_D^{20}$ +47.4 ° (*c* 0.5, chloroform); MS (FAB) *m/z* 1970.7 (90%, [M + Na]$^+$); ^{1}H NMR ($CDCl_3$, 400 MHz; 1D TOCSY, ^{1}H,^{13}C 2D COSY, 2D T-ROESY, ^{1}H,^{1}H 2D COSY) δ 7.43-7.02 (m, 45H, aromat), 5.64 (d, 1H, $J_{1'',2''}$ = 3.8 Hz, H-1''), 5.39 (dd ~t, 1H, $J_{3'''',4''''}$ = 9.6 Hz, H-3''''), 5.30 (d, 1H, $J_{1'''',2''''}$ = 3.9 Hz, H-1''''), 5.05 (dd ~ t, 1H, $J_{4'''',5''''}$ = 10.0 Hz, H-4''''), 5.01, 4.74 (2d, 2H, J_{gem} = 11.5 Hz, C-3-

CH_2Ph), 4.98, 4.62 (2d, 2H, J_{gem} ≈ 11.0 Hz, C-3''-CH_2Ph), 4.96 (dd ~ t, 1H, $J_{3''',4'''}$ = 8.2 Hz, H-3'''), 4.86 (dd, 1H, $J_{2'''',3''''}$ = 10.2 Hz, H-2''''), 4.81, 4.76 (2d, 2H, J_{gem} = 11.8 Hz, C-3'-CH_2Ph), 4.76, 4.60 (2d, 2H, J_{gem} = 12.0 Hz, C-2-CH_2Ph), 4.71, 4.60 (2d, 2H, J_{gem} = 11.0 Hz, C-2'-CH_2Ph), 4.70, 4.20 (2d, 2H, J_{gem} = 11.8 Hz, C-6''-CH_2Ph), 4.69 (dd ~ t, 1H, $J_{2''',3'''}$ = 9.6 Hz, H-2'''), 4.62, 4.36 (2d, 2H, J_{gem} ≈ 12 Hz, C-6-CH_2Ph), 4.59, 4.43 (2d, 2H, J_{gem} = 12.0 Hz, C-2''-CH_2Ph), 4.57 (d, 1H, H-1), 4.38, 4.31 (2d, 2H, J_{gem} = 11.5 Hz, C-6'-CH_2Ph), 4.37 (d, 1H, $J_{1''',2'''}$ = 8.0 Hz, H-1'''), 4.30 (d, 1H, $J_{1',2'}$ = 8.0 Hz, H-1'), 4.21 (dd, 1H, $J_{5'''',6a''''}$ = 3.5 Hz, $J_{6a'''',6b''''}$ = 12.0 Hz, H-6a''''), 4.12 (dd, 1H, $J_{5''',6a'''}$ = 2.2 Hz, $J_{6a''',6b'''}$ = 12.0 Hz, H-6a'''), 4.03 (dd, 1H, $J_{5''',6b'''}$ = 4.2 Hz, H-6b'''), 3.99 (dd ~ t, 1H, H-4'), 3.98 (dd, 1H, $J_{5'''',6b''''}$ ≈ 2 Hz, H-6b''''), 3.95 (dd ~ t, 1H, H-4), 3.88 (ddd ~ dt, 1H, H-5''''), 3.87 (dd ~ t, 1H, $J_{4'',5''}$ ≈ 10 Hz, H-4''), 3.83 (dd ~ t, 1H, H-4'''), 3.81 (dd ~ t, 1H, $J_{3,4}$ = 9.4 Hz, H-3), 3.79 (dd, 1H, $J_{5,6a}$ ≈ 2.5 Hz, H-6a), 3.77 (dd ~ t, 1H, $J_{3'',4''}$ ≈ 9.4 Hz, H-3''), 3.68 (dd, 1H, $J_{5',6a'}$ = 2.0 Hz, $J_{6a',6b'}$ ≈ 12.0 Hz, H-6a'), 3.64 (2H, dd, H-6b', ddd ~ br d, H-5''), 3.61 (dd, 1H, $J_{5'',6a''}$ ≈ 1.5 Hz, $J_{6a'',6b''}$ = 10.8 Hz, H-6a''), 3.53 (ddd ~ br dt, 1H, $J_{4,5}$ = 9.8 Hz, H-5), 3.50 (d, 1H, $J_{3',4'}$ = 8.8 Hz, H-3'), 3.48 (dd, 1H, $J_{1,2}$ = 3.9 Hz, $J_{2,3}$ = 9.4 Hz, H-2), 3.45 (dd, 1H, $J_{5,6b}$ ≤ 1.5 Hz, $J_{6a,6b}$ ≈ 9.5 Hz, H-6b), 3.43 (dd, 1H, $J_{2'',3''}$ = 9.6 Hz, H-2''), 3.38 (dd, 1H, H-6b''), 3.36 (dd ~ t, 1H, $J_{2',3'}$ = 9.0 Hz, H-2'), 3.34 (s, 3H, OCH_3), 3.24 (ddd ~ br d, 1H, $J_{4',5'}$ = 9.8 Hz, $J_{5',6b'}$ ≈ 2.5 Hz, H-5'), 3.13 (ddd ~ dt, 1H, $J_{4''',5'''}$ = 9.9 Hz, H-5'''), 2.11, 2.06, 2.03, 2.01, 1.96, 1.89, 1.83 (7 s, 21H, Ac); ^{13}C NMR ($CDCl_3$, 100 MHz; DEPT, ^1H,^{13}C 2D COSY) δ 170.51 (3C, C-2''''-, C-6''''-, C-6'''-C=O), 170.24 (C-3'''-C=O), 169.95 (C-3''''-C=O), 169.56 (C-2'''-C=O), 169.42 (C-4''''-C=O), 139.50, 139.47, 138.75, 138.69, 138.43, 138,37, 138.11, 137.74, 137.65 (9C, quat. C arom.), 128.79 - 126.81 (aromatic CH), 102.10 ($^1J_{C,H}$ ≈ 160 Hz, C-1'), 99.37 ($^1J_{C,H}$ ≈ 163 Hz, C-1'''), 98.40 (C-1), 96.92 ($^1J_{C,H}$ ≈ 174 Hz, C-1''), 95.71 ($^1J_{C,H}$ ≈ 175 Hz, C-1''''), 84.76 (C-3'), 83.00 (C-2'), 80.17 (C-3), 80.00 (C-3''), 78.94 (C-2), 78.30 (C-2''), 77.00 (C-4''), 76.00 (C-4), 75.94 (C-3'''), 75.23 (CH_2Ph), 75.15 (C-5'), 74.82 (CH_2Ph), 74.79 (CH_2Ph), 74.19 (CH_2Ph), 73.72 (2C, CH_2Ph), 73.56 (2C, CH_2Ph), 73.53 (CH_2Ph), 73.15 (C-4'''), 73.02 (C-4'), 72.70 (C-2'''), 71.56 (C-5'''), 70.50 (C-5''), 70.06 (C-2''''), 69.95 (C-5), 69.28 (C-3''''), 68.94 (C-6'), 68.52 (C-5''''), 67.95 (C-4''''), 67.74 (C-6), 67.52 (C-6''), 63.12 (C-6'''), 61.39 (C-6''''), 55.31 (OCH_3), 20.94 (OAc), 20.67 (2C, Ac), 20.60 (4C, Ac).

Anal. Calcd for $C_{108}H_{122}O_{33}$: C, 66.59; H, 6.31. Found: C, 66.78; H, 6.24.

Methyl *O*-(α-D-Glucopyranosyl)-(1→4)-*O*-(β-D-glucopyranosyl)-(1→4)-(2,3,6-tri-*O*-benzyl-α-D-glucopyranosyl)-(1→4)-*O*-(2,3,6-tri-*O*-benzyl-β-D-glucopyranosyl)-(1→4)-2,3,6-tri-*O*-benzyl-α-D-glucopyranoside (13). To a soln of **12**

(9.53 g, 4.9 mmol) in dimethoxyethane (20 mL) and methanol (100 mL) was added a soln of sodium methanolate (5 mL of 2.0 g Na/ 100 mL methanol) at rt. The reaction mixture was kept for 16 h at rt, neutralized with Amberlite IR 120 (H^+) and filtered. After addition of a few drops of triethylamine, the filtrate and methanol washings were concentrated. The residue was chromatographed on silica gel using ethyl acetate/ methanol/ water 98 : 1 : 1 as eluent to obtain pure **13** (7.39 g, 91 %) as a colourless foam: $[\alpha]_D^{20}$ +56.8 ° (*c* 0.5, chloroform); MS (FAB) *m/z* 1676.7 (40 %, $[M + Na]^+$); ^1H NMR ($CDCl_3$, 400 MHz) δ 7.37-7.35 (m, 2H, aromat), 7.27-7.12 (m, 43H, aromat), 5.63 (d, 1H, $J_{1'',2''}$ = 3.2 Hz, H-1'), 5.01 (br d, 1H, H-1'''), 3.34 (s, 3H, OCH_3).

Anal. Calcd for $C_{94}H_{108}O_{26}$: C, 68.27; H, 6.58. Found: C, 68.00; H, 6.52.

Methyl *O*-(α-D-Glucopyranosyl)-(1→4)-*O*-(β-D-glucopyranosyl)-(1→4)-(α-D-glucopyranosyl)-(1→4)-*O*-(β-D-glucopyranosyl)-(1→4)-α-D-glucopyranoside (14). A soln of **13** (2.81 g, 1.7 mmol) in ethanol (75 mL) and water (15 mL) was hydrogenated in the presence of 10 % palladium on charcoal (900 mg) at 1.1 bar for 6 h. The reaction mixture was filtered through a pad of filter aid and washed with ethanol water 1:1. After addition of a few drops of triethylamine the filtrate was concentrated. The aqueous residue (ca 10 mL) was lyophilized to obtain pure **14** (210 mg) as an amorphous colourless powder in quantitative yield; $[\alpha]_D^{20}$ +135.8 ° (*c* 0.5, water); MS (FAB) *m/z* 843.3 (35 %, $[M + H]^+$), 843.4 (15%, $[M + H]^+$); ^1H NMR (D_2O, 400 MHz) δ 5.42 (d, 2H, J = 3.9 Hz, H-1'', H-1''''), 4.81 (under HOD signal, H-1), 4.54, 4.53 (2d, 2H, $J_{1',2'} = J_{1''',2'''}$ = 7.9 Hz, H-1',H-1'''), 3.43 (s, 3H, OCH_3).

Anal. Calcd for $C_{31}H_{54}O_{26}$: C, 44.18; H, 6.46. Found: C, 43.98; H, 6.56.

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