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Synthesis of *O*-α-D-glucopyranosyl-(1→4)-*O*-α-D-xylopyranosyl-(1→4)-*O*-α-D-xylopyranosyl-(1→4)-D-glucopyranose as a substrate analogue of alpha amylase

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ABSTRACT

The tetrasaccharide α-D-Glc*p*-(1→4)-α-D-Xyl*p*-(1→4)-α-D-Xyl*p*-(1→4)-D-Glc*p* (**1**) has been synthesized, as a substrate analogue of alpha amylase, by silver perchlorate-catalyzed glycosylation of benzyl 2,3,6-tri-*O*-benzyl-4-*O*-(2,3-di-*O*-benzyl-α-D-xylopyranosyl)-β-D-glucopyranoside (**30**) with 2,3-di-*O*-benzyl-4-*O*-(2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl)-α-D-xylopyranosyl chloride or by methyl triflate-promoted condensation of **30** with methyl 2,3-di-*O*-benzyl-4-*O*-(2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl)-1-thio-β-D-xylopyranoside, followed by removal of protecting groups of the resulting tetrasaccharide derivative **40**.

INTRODUCTION

A substrate analogue that binds to an enzyme in a manner similar to a productive binding-mode of the substrate, but is not hydrolyzed by the enzyme, is strongly in demand not only as a reagent in mechanistic studies of enzymes but also for providing a crystalline enzyme–inhibitor complex for X-ray analysis to determine the active site. We envisaged that the tetrasaccharide *O*-α-D-Glc*p*-(1→4)-*O*-α-D-Xyl*p*-(1→4)-*O*-α-D-Xyl*p*-(1→4)-D-Glc*p* (**1**) may serve in this capacity in studies of Taka-amylase A (alpha amylase from *Aspergillus oryzae*) and porcine pancreatic alpha amylase, whose three-dimensional structures are known from X-ray analysis[1,2]. The structure of **1** was designed on the basis of the subsite structure and the properties of the amylases as determined kinetically[3–5]: (*a*) maltotetraose is the smallest substrate of the amylases required for investigating the structures of the active sites[3,4]; (*b*) the two D-glucose residues at the reducing and terminal non-reducing ends in maltotetraose are especially important to the productive binding at each active site of the enzymes[3,4]; and (*c*) replacement by D-xylose residues of two consecutive D-glucose units at the middle positions in maltotetraose yields a product that may resist hydrolysis by the enzymes, as the hydroxymethyl group of the D-glucose residue in substrates at the subsites near the

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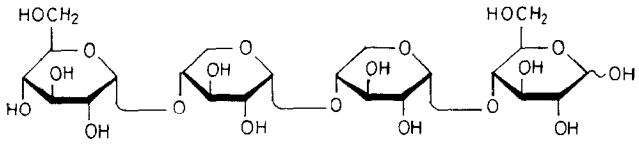
catalytic site is essential for hydrolysis of the D-glucosidic linkage by the enzymes[5]. We now report the synthesis of **1**.

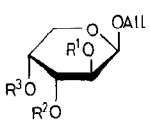


1

RESULTS AND DISCUSSION

The strategy for the synthesis of **1** involves (*a*) the synthesis of benzyl 2,3,6-tri-*O*-benzyl-4-*O*-(2,3-di-*O*-benzyl-*α*-D-xylopyranosyl)-*β*-D-glucopyranoside (**30**), allyl 2,3-di-*O*-benzyl-4-*O*-(2,3,4,6-tetra-*O*-benzyl-*α*-D-glucopyranosyl)-*β*-D-xylopyranoside (**34**), and methyl 2,3-di-*O*-benzyl-4-*O*-(2,3,4,6-tetra-*O*-benzyl-*α*-D-glucopyranosyl)-1-thio-*β*-D-xylopyranoside (**37**); (*b*) conversion of **34** into the corresponding disaccharide *α*-chloride **39**; and (*c*) block condensation of **30** with each of the glycosyl donors **37** and **39**, followed by removal of blocking groups.

The preparation of D-xylosyl acceptors and donors necessary for the synthesis of **30**, **34**, and **37** was first investigated. Allyl *β*-D-arabinopyranoside (**2**) was prepared (41%) by reaction of D-arabinose with allyl alcohol–hydrogen chloride and converted, *via* **3**, **4**, and **5**, into allyl 2,3-anhydro-*β*-D-ribopyranoside (**6**, 49% from **2**) according to the procedure analogues to that used for the synthesis of the corresponding methyl[6] and benzyl[7] glycosides. Alkylation of **6** with *p*-methoxybenzyl chloride and sodium hydride in *N,N*-dimethylformamide[8] gave **7**. Opening of the anhydro ring[7,9] in **7** by treatment with aqueous potassium hydroxide afforded 76% of methyl 4-*O*-(*p*-methoxybenzyl)-*β*-D-xylopyranoside (**8**) which, with benzyl bromide and sodium hydride in *N,N*-dimethylformamide[8], gave allyl 2,3-di-*O*-benzyl-4-*O*-(*p*-methoxybenzyl)-*β*-D-xylopyranoside (**9**). The allyl group in **9** was removed by rearrangement first to the propenyl ether with tris(triphenylphosphine)rhodium(I) chloride[10,11] in the presence of 1,4-diazabicyclo[2.2.2]octane[10], followed by hydrolysis with mercuric chloride and mercuric oxide[12], to afford 2,3-di-*O*-benzyl-4-*O*-(*p*-methoxybenzyl)-D-xylopyranose (**12**). However, attempts to convert **12** into the corresponding *α*-chloride **13** by treatment with (chloromethylene)dimethyliminium chloride[13] in dichloromethane or with thionyl chloride in dichloromethane in the presence of 2,4,6-trimethylpyridine[14] resulted in extensive cleavage of the *p*-methoxybenzyl group, so that the *p*-methoxybenzyl group in **9** was replaced by an acetyl group. Treatment of **9** with ceric ammonium nitrate in acetonitrile–water[15] selectively removed the *p*-methoxybenzyl group to give allyl 2,3-di-*O*-benzyl-*β*-D-xylopyranoside (**10**) which, on acetylation, afforded **11**. Isomerization of the allyl group in **11** to the propenyl ether with the rhodium complex[10,11], followed by hydrolysis with dilute acid[16], gave 4-*O*-acetyl-2,3-di-*O*-benzyl-D-xylopyranose (**14**) which, with oxalyl chloride–*N,N*-dimethylformamide in dichloromethane[17], was transformed into 4-*O*-acetyl-2,3-di-*O*-benzyl-*α*-D-xylopyranosyl chloride (**15**).

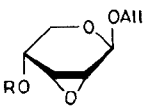


2 $R^1 = R^2 = R^3 = H$

3 $R^1 = H$, R^2, $R^3 = CMe_2$

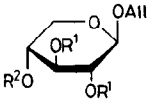
4 $R^1 = Ts$, R^2, $R^3 = CMe_2$

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



6 R = H

7 R = MBn

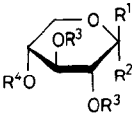


8 $R^1 = H$, $R^2 = MBn$

9 $R^1 = Bzl$, $R^2 = MBn$

10 $R^1 = Bzl$, $R^2 = H$

11 $R^1 = Bzl$, $R^2 = Ac$



12 R^1, $R^2 = H$, OH; $R^3 = Bzl$, $R^4 = MBn$

13 $R^1 = H$, $R^2 = Cl$, $R^3 = Bzl$, $R^4 = MBn$

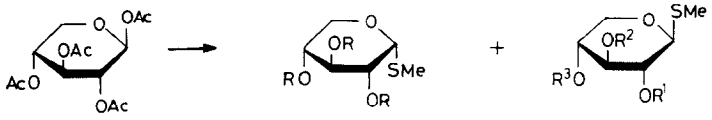
14 R^1, $R^2 = H$, OH; $R^3 = Bzl$, $R^4 = Ac$

15 $R^1 = H$, $R^2 = Cl$, $R^3 = Bzl$, $R^4 = Ac$

MBn = *p*-methoxybenzyl

Reaction of β-D-xylose tetraacetate (**16**) with methyl tributyltin sulfide in 1,2-dichloroethane in the presence of stannic chloride[18] gave, after column chromatography, 90% of a mixture of methyl 2,3,4-tri-*O*-acetyl-1-thio-α- (**17**) and β-D-xylopyranoside[19] (**20**) in the ratio of 1:1.7, as indicated by ^{13}C-n.m.r. spectroscopy, which showed the signals for C-1 at 83.1 and 83.3 p.p.m., respectively. Fractional crystallization of the mixture afforded 38% of **20**, which was *O*-deacetylated to give methyl 1-thio-β-D-xylopyranoside[19] (**21**). More **21** (8%) was isolated from a mixture of **18** and **21** obtained after *O*-deacetylation of the mother liquor of **20**. Compound **18** could not be obtained pure, but was characterized as the crystalline tribenzyl derivative **19**. Acetonation of **21** with 2-methoxypropene in *N,N*-dimethylformamide in the presence of a catalytic amount of methanolic hydrogen chloride[20] gave 70% of methyl 2,3-*O*-isopropylidene-1-thio-β-D-xylopyranoside (**22**) after column chromatography. Acetylation of **22** gave the 4-*O*-acetyl-2,3-*O*-isopropylidene derivative **23**, the ^1H-n.m.r. spectrum of which showed an octet for H-4 at δ 5.04, confirming the position of the acetal group in **22**. *p*-Methoxybenzylation of **22** afforded the 2,3-*O*-isopropylidene-4-*O*-(*p*-methoxybenzyl) derivative **24**. Selective cleavage of the acetal group in **24** with mild acid gave **25**, which was benzylated to afford methyl 2,3-di-*O*-benzyl-4-*O*-(*p*-methoxybenzyl)-1-thio-β-D-xylopyranoside (**26**).

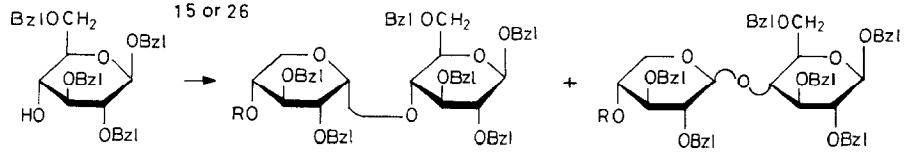
Glycosylation of benzyl 2,3,6-tri-*O*-benzyl-β-D-glucopyranoside[21] (**27**) with **15** in ether in the presence of silver perchlorate[22] and 2,4,6-trimethylpyridine gave the α- (**28**, 27%) and β-(1→4)-linked disaccharide derivative (**31**, 55%) after column chromatography. In the ^{13}C-n.m.r. spectra of **28** and **31**, the signals for C-1′ appeared at 96.95 and 102.8 p.p.m., indicating[23] the configurations at C-1′ to be α and β, respectively.



16

17 R = Ac
18 R = H
19 R = Bzl

20 $R^1 = R^2 = R^3$ = Ac
21 $R^1 = R^2 = R^3$ = H
22 R^1, R^2 = CMe_2; R^3 = H
23 R^1, R^2 = CMe_2; R^3 = Ac
24 R^1, R^2 = CMe_2; R^3 = MBn
25 $R^1 = R^2$ = H, R^3 = MBn
26 $R^1 = R^2$ = Bzl, R^3 = MBn



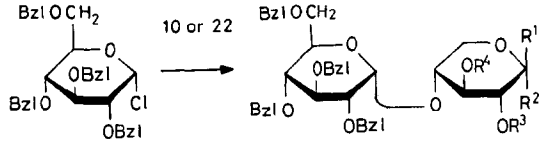
27

28 R = Ac
29 R = MBn
30 R = H

31 R = Ac
32 R = MBn

Condensation of **27** with **26** in ether in the presence of methyl triflate[24] and molecular sieve afforded **29** (19%) and **32** (52%) after column chromatography. The *α* and *β* configurations at C-1′ were apparent[23] from the ^{13}C-n.m.r. signals at 96.4 and 103.0 p.p.m., respectively. Reaction of **27** with **26** in 1,2-dichloroethane–*N*,*N*-dimethylformamide in the presence of cupric bromide, tetrabutylammonium bromide, and molecular sieve[25], followed by column chromatography of the product, afforded **29** (50%) and **32** (22%). *O*-Deacetylation of **28** provided the disaccharide glycosyl acceptor **30** having HO-4′ unsubstituted. The same compound was also obtained by selective cleavage of the *p*-methoxybenzyl group of **29** with ceric ammonium nitrate[15]. Condensation of **10** with 2,3,4,6-tetra-*O*-benzyl-*α*-D-glucopyranosyl chloride[26] (**33**), promoted by silver perchlorate[22] as before, after column chromatography, gave **34** (75%), the ^{13}C-n.m.r. spectrum of which showed the signal for C-1′ at 98.8 p.p.m. Under these conditions and with column chromatography of the product, reaction of **22** with **33** afforded **35** (71%), the ^{13}C-n.m.r. spectrum of which showed the signal for C-1′ at 95.9 p.p.m. Compound **35** was transformed into **37** in sequence by deisopropylidenation (→**36**) and benzylation.

Deallylation of **34**, as for **9**, gave the hemiacetal **38** which, with oxalyl chloride–*N*,*N*-dimethylformamide[17], was converted into the corresponding *α*-chloride **39**. Glycosylation of **30** with **39**, catalyzed by silver perchlorate[22] as before, provided 58% of the tetrasaccharide derivative **40** after column chromatography. In the ^{13}C-n.m.r. spectrum



33

34 R^1 = All, R^2 = H, R^3 = R^4 = Bzl

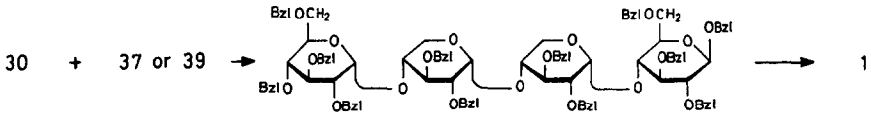
35 R^1 = SMe, R^2 = H, R^3 = R^4 = CMe_2

36 R^1 = SMe, R^2 = R^3 = R^4 = H

37 R^1 = SMe, R^2 = H, R^3 = R^4 = Bzl

38 R^1, R^2 = H, OH; R^3 = R^4 = Bzl

39 R^1 = H, R^2 = Cl, R^3 = R^4 = Bzl



40

of **40**, the presence of the signals for C-1′, C-1″, and C-1‴ at 98.65, 98.48, and 96.76 p.p.m. established[23] the *α* configuration at each anomeric center. Coupling of **30** with **37**, promoted by methyl triflate[24] as before, followed column chromatography of the product, afforded 43% of **40**. Condensation of **30** with **37**, catalyzed by cupric bromide–tetrabutylammonium bromide[25] as before, was very sluggish and a large proportion of **30** remained unreacted even after 3 days, giving 10% of **40** after column chromatography. Catalytic hydrogenolysis (Pd–C) of **40** and purification of the product by column chromatography furnished 84% of **1**, the structure and purity of which were determined by ^{1}H- and ^{13}C-n.m.r. spectroscopy, methylation analysis[27], and l.c.

Preliminary experiments indicated that **1** is not hydrolyzed by the alpha amylases described earlier and is for both enzymes an inhibitor having the same value of the inhibitor constant as that of the Michaelis constant of maltotetraose[3]. A detailed kinetic study is under way.

EXPERIMENTAL

General methods. — Melting points were determined with a Yanagimoto micro melting-point apparatus and are uncorrected. Optical rotations were determined with an Applied Electronic automatic polarimeter Model MP-1T. N.m.r. spectra (^{1}H 90 MHz; ^{13}C 22.6 MHz) were recorded with a Hitachi R-90H spectrometer for solutions in $CDCl_3$ (internal Me_4Si) or D_2O (internal sodium 4,4-dimethyl-4-silapentanoate-d_4). N.m.r. spectra (^{1}H 270 MHz; ^{13}C 67.8 MHz) of **1** and **40** were recorded with a Jeol JNM GX-270 spectrometer for solutions in $CDCl_3$ (internal Me_4Si) or D_2O (^{1}H, external Me_4Si; ^{13}C, internal MeOH, δ_{MeOH} *vs* δ_{Me4Si} 49.80). H.p.l.c. was performed with a Jasco

880-PU provided with a Shodex SE-61 r.i. detector and a column of Finepac Sil NH_2 (10 μm, 250 x 4.6 mm i.d., Jasco). G.l.c. was performed under the same conditions as described previously[28]. Elemental analyses were not obtained for syrupy products, but they were shown to be pure by chromatography and n.m.r. spectroscopy. Organic solutions were dried over anhydrous Na_2SO_4 or $MgSO_4$. Solutions were evaporated at a temperature $< 50°$ under diminished pressure. T.l.c. was performed on Silica Gel 60 (No. 7734, Merck) with detection by charring with H_2SO_4. Column chromatography was performed on Wako Gel C-300.

*Allyl β-*D*-arabinopyranoside* (**2**). — Acetyl chloride (50 mL) was added slowly with stirring at 0° to anhydrous allyl alcohol (200 mL), followed by finely powdered D-arabinose (50 g). The mixture was stirred overnight at room temperature, then cooled to 0°, and neutralized with solid $NaHCO_3$. Insoluble material was collected on a Celite pad and washed with EtOH, and the combined filtrate and washings were evaporated. Toluene was evaporated from the residue which was recrystallized twice from EtOH to give **2** (25.9 g, 41%), m.p. 121–123°, $[\alpha]_D^{21}$ −216° (*c* 1.1, H_2O); ^{13}C-n.m.r. (D_2O): δ 136.3 and 120.7 ($CH=CH_2$), 100.6 (C-1), 71.5 (2C) C-3,4), 71.3 ($CH_2=CH-CH_2$), 70.8 (C-2), and 65.4 (C-5).

Anal. Calc. for $C_8H_{14}O_5$: C, 50.52; H, 7.42. Found: C, 50.64; H, 7.48.

*Allyl 3,4-*O*-isopropylidene-β-*D*-arabinopyranoside* (**3**). — A mixture of **2** (25 g), 2,2-dimethoxypropane (90 mL), and $TsOH \cdot H_2O$ (0.2 g) was stirred for 2 h at room temperature. Triethylamine (1 mL) was added and the mixture was evaporated. The residue was recrystallized twice from light petroleum to afford **3** (27.7 g, 81%), m.p. 71–73°, $[\alpha]_D^{21}$ −191° (*c* 1.2, $CHCl_3$); n.m.r. ($CDCl_3$): ^{1}H, δ 6.10–5.73 (m, 1 H, –CH=), 2.74 (d, $J_{2,2\text{-OH}}$ 7.0 Hz, exchangeable with D_2O, HO-2), and 1.53 and 1.36 (2 s, each 3 H, CMe_2); ^{13}C, δ 133.6 and 117.6 ($CH=CH_2$), 109.0 (CMe_2), 96.9 (C-1), and 27.9 and 25.9 (CMe_2).

Anal. Calc. for $C_{11}H_{18}O_5$: C, 57.38; H, 7.88. Found: C, 57.29; H, 7.92.

*Allyl 3,4-*O*-isopropylidene-2-*O*-p-tolylsulfonyl-β-*D*-arabinopyranoside* (**4**). — A solution of **3** (25.2 g) in pyridine (130 mL) was cooled to 0° and treated with TsCl (30 g). The mixture was stirred for 5 h at room temperature and overnight at 40°, and then diluted with $CHCl_3$. The solution was washed successively with cold 5% HCl, aq. $NaHCO_3$, and water, dried, and evaporated. Crystallization of the residue from ether–hexane gave **4** (34.9 g, 83%), m.p. 77–78°, $[\alpha]_D^{21}$ −184° (*c* 1.2, $CHCl_3$); n.m.r. ($CDCl_3$): ^{1}H, δ 7.83 and 7.33 (2 d, each 2 H, *J* 7.9 Hz, aromatic H), 6.05–5.67 (m, 1 H, –CH=), 2.43 (s, 3 H, aryl CH_3), and 1.25 and 1.13 (2 s, each 3 H, CMe_2); ^{13}C, δ 144.7, 133.4, 129.5, and 128.2 (aromatic C), 133.1 and 117.7 ($CH=CH_2$), 109.1 (CMe_2), 95.6 (C-1), 27.5 and 26.1 (CMe_2), and 21.55 (aromatic CH_3).

Anal. Calc. for $C_{18}H_{24}O_7S$: C, 56.24; H, 6.29. Found: C, 56.32; H, 6.23.

*Allyl 2-*O*-p-tolylsulfonyl-β-*D*-arabinopyranoside* (**5**). — To a solution of **4** (32 g) in AcOH (120 mL) at 90° was added dropwise water (80 mL), and the mixture was stirred for 20 min, then cooled, and evaporated. The last traces of the solvents were removed by repeated evaporation of toluene from the residue, which was recrystallized from CH_2Cl_2–ether to afford **5** (24.7 g, 86%), m.p. 69–71°, $[\alpha]_D^{21}$ −125° (*c* 1.2, $CHCl_3$); n.m.r.

($CDCl_3$): ^1H, δ 7.83 and 7.33 (2 d, each 2 H, J 7.9 Hz, aromatic H), 5.97–5.60 (m, 1 H, –CH–), and 2.43 (s, 3 H, aryl CH_3); ^{13}C, δ 144.9, 133.4, 129.7, and 127.9 (aromatic C), 133.2 and 117.4 ($CH=CH_2$), 95.8 (C-1), and 21.6 (aromatic CH_3).

Anal. Calc. for $C_{15}H_{20}O_7S$: C, 52.32; H, 5.85. Found: C, 52.40; H, 5.90.

*Allyl 2,3-anhydro-β-*D*-ribopyranoside* (**6**). — Compound **5** (23.5 g) was dissolved in dry MeOH (300 mL) containing sodium (4.9 g). The solution was kept overnight at room temperature, then cooled, and neutralized with dil. H_2SO_4. The mixture was evaporated and the residue was extracted thoroughly with $CHCl_3$. The extracts were washed with water, dried, and evaporated. Crystallization of the residue from CH_2Cl_2–ether gave **6** (9.9 g, 84%), m.p. 59–61°, $[\alpha]_D^{21}$ −51° (*c* 1.2, $CHCl_3$); ^{13}C-n.m.r. data ($CDCl_3$): δ 133.6 and 117.6 ($CH=CH_2$), 93.7 (C-1), 69.0 ($CH_2=CH-CH_2$), 62.0 (C-4), 61.8 (C-5), 52.0 (C-3), and 51.9 (C-2).

Anal. Calc. for $C_8H_{12}O_4$: C, 55.81; H, 7.03. Found: C, 55.77; H, 7.09.

*Allyl 2,3-anhydro-4-*O*-(*p*-methoxybenzyl)-β-*D*-ribopyranoside* (**7**). — Sodium hydride (5.5 g; 50% mineral oil) was added at 0° to a solution of **6** (13.2 g) in *N,N*-dimethylformamide (70 mL), and the mixture was stirred for 30 min at room temperature and then cooled to 0°. *p*-Methoxybenzyl chloride (20 mL) was added dropwise, and the mixture was stirred for 2 h at room temperature, and then cooled. The excess of hydride was decomposed by cautious addition of ice chips and the mixture was poured into ice–water. The precipitate formed was filtered off, washed with cold water, and dissolved in $CHCl_3$. The solution was washed with water, dried, and evaporated. Crystallization of the residue from ether–light petroleum gave **7** (18.4 g, 82%), m.p. 39–42°, $[\alpha]_D^{21}$ +13° (*c* 1.4, $CHCl_3$); n.m.r. ($CDCl_3$): ^1H, δ 7.29 and 6.85 (2 d, each 2 H, J 8.8 Hz, aromatic H), 6.14–5.71 (m, 1 H, –CH=), and 4.61 (s, 3 H, OMe); ^{13}C, δ 159.2, 129.9, 129.3, and 113.8 (aromatic C, *p*-methoxybenzyl), 133.7 and 117.5 ($CH=CH_2$), 94.4 (C-1), 69.2 ($CH_2=CH-CH_2$), 68.6 (C-4), 59.3 (C-5), 55.2 (OMe), 52.1 C-3), and 50.15 (C-2).

Anal. Calc. for $C_{16}H_{20}O_5$: C, 65.74; H, 6.90. Found: C, 65.69; H, 6.85.

*Allyl 4-*O*-(*p*-methoxybenzyl)-β-*D*-xylopyranoside* (**8**). — A suspension of **7** (10 g) in 10% aq. KOH (500 mL) was stirred for 2 days at 100°. The solution was cooled, neutralized with dil. H_2SO_4, extracted thoroughly with $CHCl_3$, and the extracts were dried and evaporated. Column chromatography (4:1 benzene–EtOAc) of the residue afforded **8** (8.05 g, 76%), m.p. 53–54° (from ether–light petroleum), $[\alpha]_D^{21}$ −75° (*c* 1.3, $CHCl_3$); n.m.r. ($CDCl_3$): ^1H, δ 7.25 and 6.85 (2 d, each 2 H, J 8.6 Hz, aromatic H), 6.14–5.71 (m, 1 H, –CH=), and 3.78 (s, 3 H, OMe); ^{13}C, δ 159.3, 129.8, 129.4, and 113.5 (aromatic C, *p*-methoxybenzyl), 133.6 and 117.8 ($CH=CH_2$), 101.5 (C-1), 76.4 (C-4), 73.0 (C-3), 71.9 (C-2), 69.6 ($CH_2=CH-CH_2$), 61.8 (C-5), and 55.2 (OMe).

Anal. Calc. for $C_{16}H_{22}O_6$: C, 61.92; H, 7.15. Found: C, 62.00; H, 7.21.

*Allyl 2,3-di-*O*-benzyl-4-*O*-(*p*-methoxybenzyl)-β-*D*-xylopyranoside* (**9**). — A solution of **8** (7.6 g) in *N,N*-dimethylformamide (80 mL) was treated with NaH (3.5 g; 50% mineral oil) and then cooled to 0°. Benzyl bromide (8.1 mL) was added dropwise and the mixture was stirred overnight at room temperature. Methanol was then added to decompose the excess of hydride, the solvents were evaporated, and a solution of the

residue in CH_2Cl_2 was washed with water, dried, and evaporated. The residue was subjected to column chromatography (20:1 benzene–EtOAc) to give **9**, isolated as a syrup (10.6 g, 88%), $[\alpha]_D^{21}$ +4° (*c* 1.2, $CHCl_3$); ^{13}C-n.m.r. ($CDCl_3$): δ 159.2 and 113.75 (aromatic C, *p*-methoxybenzyl), 138.5 and 137.9 (aromatic C-1, benzyl), 133.9 and 117.1 ($CH=CH_2$), 103.2 (C-1), 83.6 (C-3), 81.9 (C-2), 63.9 (C-5), and 55.1 (OMe).

*Allyl 2,3-di-*O*-benzyl-β-*D*-xylopyranoside* (**10**). — A solution of **9** (6.8 g) in 9:1 MeCN–water (140 mL) was stirred with ceric ammonium nitrate (15.2 g) for 2 h at room temperature. The mixture was diluted with $CHCl_3$, washed successively with water, aq. $NaHCO_3$, and water, dried, and evaporated. Column chromatography (10:1 benzene–EtOAc) of the product gave **10** (4.3 g, 84%), m.p. 78–81° (from ether–hexane), $[\alpha]_D^{21}$ −40° (*c* 1.2, $CHCl_3$; ^{13}C-n.m.r. ($CDCl_3$): δ 138.4 and 138.05 (aromatic C-1), 133.9 and 117.2 ($CH=CH_2$), 102.4 (C-1), 82.3 (C-3), 80.4 (C-2), and 64.5 (C-5).

Anal. Calc. for $C_{22}H_{26}O_5$: C, 71.33; H, 7.07. Found: C, 71.35; H, 7.13.

*Allyl 4-*O*-acetyl-2,3-di-*O*-benzyl-β-*D*-xylopyranoside* (**11**). — Acetylation of **10** (3.9 g) in 1:1 Ac_2O–pyridine (30 mL) afforded **11** as a syrup (4.1 g, 95%), $[\alpha]_D^{21}$ −33.5° (*c* 1.3, $CHCl_3$); n.m.r. ($CDCl_3$): ^{1}H, δ 7.33–7.21 (m, 10 H, 2 Ph), 6.10–5.73 (m, 1 H, –CH=), and 1.94 (s, 3 H, OAc); ^{13}C, δ 169.7 (C=O), 138.4 and 138.2 (aromatic C-1), 133.8 and 117.2 ($CH=CH_2$), 102.7 (C-1), 81.3 (C-3), 80.6 (C-2), 62.5 (C-5), and 20.8 ($COCH_3$).

*2,3-Di-*O*-benzyl-4-*O*-(p-methoxybenzyl)-*D*-xylopyranose* (**12**). — A mixture of **9** (3.1 g), tris(triphenylphosphine)rhodium(I) chloride (0.25 g), and 1,4-diazabicyclo[2.2.2]octane (1 g) in 10:3:1 EtOH–PhMe–water (98 mL) was boiled under reflux for 8 h and then evaporated. The residue was extracted with CH_2Cl_2, and the extract was washed successively with water, cold M HCl, aq. $NaHCO_3$, and water, dried, and evaporated. To a solution of the residue in 9:1 acetone–water (20 mL) was added HgO (1.0 g), followed by a solution of $HgCl_2$ (1.0 g) in 9:1 acetone–water (20 mL). The suspension was stirred for 30 min at room temperature, the solids were removed by filtration, and the filtrate was evaporated. A solution of the residue in $CHCl_3$ was washed successively with water, aq. KI, and water, dried, and evaporated. Column chromatography (10:1 benzene–EtOAc) of the product gave **12** (2.3 g, 81%), m.p. 80–85° and 93–97° (from ether–light petroleum), $[\alpha]_D^{21}$ +13° (*c* 1.1, $CHCl_3$); ^{13}C-n.m.r. ($CDCl_3$): 159.1 and 113.7 (aromatic C, *p*-methoxybenzyl), 138.6–137.6 (aromatic C-1, benzyl), 97.7 (C-1β), 91.2 (C-1α), 64.0 (C-5β), 60.2 (C-5α), 55.2 (OMe).

Anal. Calc. for $C_{27}H_{30}O_6$: C, 71.98; H, 6.71. Found: C, 72.12; H, 6.77.

*4-*O*-Acetyl-2,3-di-*O*-benzyl-*D*-xylopyranose* (**14**). — A mixture of **11** (3.8 g), tris(triphenylphosphine)rhodium(I) chloride (0.3 g), and 1,4-diazabicyclo[2.2.2]octane (1.5 g) in 8:3:1 EtOH–PhMe–water (96 mL) was boiled under reflux for 6 h, and then processed as just described. A solution of the residue in 20:1 acetone–M HCl (42 mL) was boiled under reflux for 20 min, then cooled, neutralized with aq. $NaHCO_3$, and evaporated, and the residue was extracted with CH_2Cl_2. The extract was washed with water, dried, and evaporated. Column chromatography (10:1 benzene–EtOAc) of the product gave **14**, isolated as a syrup (2.85 g, 83%), $[\alpha]_D^{21}$ −5° (*c* 1.2, $CHCl_3$); n.m.r. ($CDCl_3$): ^{1}H, δ 7.38–7.22 (m, 10 H, 2 Ph) and 1.95 (s, 3 H, OAc); ^{13}C, δ 170.1 (C–O), 138.3 and 137.6 (aromatic C-1), 97.0 (C-1β), 91.3 (C-1α), 61.7 (C-5β), 59.6 (C-5α), and 20.9 ($COCH_3$).

*Methyl 2,3,4-tri-*O*-acetyl-1-thio-β-*D*-xylopyranoside* (**20**) *and methyl 1-thio-β-*D*-xylopyranoside* (**21**). — To a stirred solution of **16** (25 g, 78.5 mmol) and methyl tributyltin sulfide (29.1 g, 86.3 mmol) in 1,2-dichloroethane (300 mL) at 0° was added dropwise a solution of $SnCl_4$ (10.1 mL, 86.3 mmol) in 1,2-dichloroethane (100 mL). The mixture was stirred for 3 h at room temperature, poured into ice–aq. $NaHCO_3$, and filtered through a Celite pad which was washed with $CHCl_3$. The combined filtrate and washings were washed with water, dried, and evaporated. Column chromatography (9:1 benzene–EtOAc) of the residue gave a mixture of methyl 2,3,4-tri-*O*-acetyl-1-thio-α-D-xylopyranoside (**17**) and **20** (21.7 g, 90%) in the ratio 1:1.7 (from the ^{13}C-n.m.r spectrum). Crystallization of the mixture from EtOH afforded **20** (9.1 g, 38%), m.p. 90–91°, $[\alpha]_D^{21}$ −75° (*c* 1.25, $CHCl_3$); lit.[19] m.p. 87.5–90°, $[\alpha]_D$ −73.4° (*c* 0.49, $CHCl_3$); ^{13}C-n.m.r. ($CDCl_3$): δ 169.6, 169.5, and 169.2 (C=O), 83.3 (C-1), 72.6 (C-3), 69.1 (C-2), 68.8 (C-4), 65.9 (C-5), 20.6 ($COCH_3$), and 11.5 (SMe).

^{13}C-N.m.r. ($CDCl_3$) for **17**: δ 83.1 (C-1), 70.7 (C-2), 69.6 (C-3), 69.1 (C-4), 69.0 (C-5), and 12.6 (SMe).

A solution of **20** (8.5 g) in anhydrous MeOH (100 mL) containing methanolic NaOMe (3 mL) was kept for 1 h at room temperature, neutralized with Amberlite IR-120 (H^+) resin, filtered, and evaporated. The residue was recrystallized from EtOH to give **21** (4.6 g, 92%), m.p. 173–174°, $[\alpha]_D^{21}$ −69° (*c* 1.2, water); lit.[19] 168.5–170°, $[\alpha]_D$ −72° (*c* 0.5, water); n.m.r. (D_2O): ^1H, 4.39 (d, 1 H, $J_{1,2}$ 4.7 Hz, H-1) and 2.20 (s, 3 H, SMe); ^{13}C, δ 88.8 (C-1), 79.7 (C-3), 74.0 (C-2), 71.65 (C-4), 71.4 (C-5), and 14.1 (SMe).

The mother liquor obtained by removal of **20** from a mixture of **17** and **20** was deacetylated, as just described, to afford another **21** (1.1 g, 8%; total yield of **21**, 43%).

*Methyl 2,3,4,6-tetra-*O*-benzyl-1-thio-α-*D*-xylopyranoside* (**19**). — The mother liquor obtained after removal of **21** from a mixture of **18** and **21**, as just described, was evaporated. A portion of the syrupy residue (2.3 g) was treated with NaH (2.75 g; 50% mineral oil) in *N,N*-dimethylformamide (30 mL), followed by $PhCH_2Br$ (6.3 mL), and processed as described for the preparation of **9**. The residue was recrystallized three times from light petroleum to give **19** (3.1 g, 55%), m.p. 81–82°, $[\alpha]_D^{26}$ +110° (*c* 1.75, $CHCl_3$); n.m.r. ($CDCl_3$): ^1H, δ 7.42–7.15 (m, 15 H, 3 Ph), 4.61 (d, 1 H, $J_{1,2}$ 4.6 Hz, H-1), and 2.00 (SMe); ^{13}C, δ 138.7, 138.2, and 137.8 (aromatic C-1), 84.7 (C-1), 60.2 (C-5), and 12.3 (SMe).

Anal. Calc. for $C_{27}H_{30}O_4S$: C, 71.97; H, 6.71. Found: C, 72.05; H, 6.78.

*Methyl 2,3-*O*-isopropylidene-1-thio-β-*D*-xylopyranoside* (**22**). — 2-Methoxypropene (8.1 mL, 84.6 mmol) was added dropwise at 5° to a stirred solution of **21** (5.1 g, 28.3 mmol) in *N,N*-dimethylformamide (40 mL) containing 5M HCl in MeOH (0.02 mL). The mixture was stirred for 30 min at 5°, after which time t.l.c. (1:1 hexane–EtOAc) showed the disappearance of **21** and the presence of two major [R_F 0.44 (**22**) and 0.57] and one minor (R_F 0.54) components. The mixture was diluted with $CHCl_3$ and washed with water. The $CHCl_3$ solution was made slightly acidic (as determined with indicator paper) by addition of a few drops of 5M HCl in MeOH. After 10 min, t.l.c. (1:1 hexane–EtOAc) showed conversion of the component (R_F 0.57) into the major (R_F 0.44) and minor (R_F 0.54) components. The solution was washed successively with aq.

$NaHCO_3$, and water, dried, and evaporated. Column chromatography (2:1→1:1 hexane–EtOAc, stepwise) of the residue gave **22** (4.35 g, 70%), m.p. 94–96° (from ether), $[\alpha]_D^{20}$ −61° (*c* 1.1, $CHCl_3$); n.m.r. ($CDCl_3$): ^1H, δ 4.52 (d, 1 H, $J_{1,2}$ 9.0 Hz, H-1), 3.77 (d, 1 H, $J_{4,4\text{-OH}}$ 3.5 Hz, exchangeable with D_2O, HO-4), 3.52 (t, 1 H, $J_{3,4}$ 8.7 Hz, H-3), 3.28 (t, 1 H, $J_{2,3}$ 8.8 Hz, H-2), 2.23 (s, 3 H, SMe), and 1.47 (s, 6 H, CMe_2); ^{13}C, δ 111.0 (CMe_2), 83.75 (C-1), 82.8 and 75.3 (C-2, 3), 69.9 (C-4), 68.7 (C-5), 26.7 and 26.5 (CMe_2), and 12.3 (SMe).

Anal. Calc. for $C_9H_{16}O_4S$: C, 49.07; H, 7.32. Found: C, 48.95; H, 7.28.

*Methyl 4-*O*-acetyl-2,3-*O*-isopropylidene-1-thio-β-*D*-xylopyranoside* (**23**). — Acetylation of **22** (0.11 g) with 1:1 Ac_2O–pyridine (1 mL) gave **23** as a syrup (0.12 g, 92%), $[\alpha]_D^{20}$ −90° (*c* 1.0, $CHCl_3$); n.m.r. data ($CDCl_3$): ^1H, δ 5.04 (o, 1 H, $J_{4,5e}$ 5.3 Hz, $J_{4,5a}$ 8.1 Hz, H-4), 4.61 (d, 1 H, $J_{1,2}$ 9.0 Hz, H-1), 3.74 (t, 1 H, $J_{3,4}$ 9.2 Hz, H-3), 3.40 (t, 1 H, $J_{2,3}$ 9.2 Hz, H-2), 2.22 (s, 3 H, SMe), 2.09 (s, 3 H, OAc), and 1.48 (s, 6 H, CMe_2); ^{13}C, δ 169.0 (C=O), 111.5 (CMe_2), 83.8 (C-1), 79.2, 75.6, 70.6, and 66.7 (C-2,3,4,5), 26.7 and 26.5 (CMe_2), 20.8 ($COCH_3$), and 12.25 (SMe).

*Methyl 2,3-*O*-isopropylidene-4-*O*-(*p*-methoxybenzyl)-1-thio-β-*D*-xylopyranoside* (**24**). — A solution of **22** (2.55 g) in *N,N*-dimethylformamide (20 mL) was treated with NaH (0.78 g; 50% mineral oil), followed by *p*-methoxybenzyl chloride (2 mL), and processed as described for the preparation of **9**. Column chromatography (4:1 hexane–EtOAc) of the product afforded **24** (2.54 g, 90%), m.p. 62–64° (from EtOH), $[\alpha]_D^{20}$ −2° (*c* 1.3, $CHCl_3$); n.m.r. ($CDCl_3$): ^1H, δ 7.27 and 6.86 (2 d, *J* 8.6 Hz, aromatic H), 3.78 (s, 3 H, OMe), 2.20 (s, 3 H, SMe), and 1.48 (s, 3 H, CMe_2); ^{13}C, δ 159.2, 129.9, 129.3, and 113.8 (aromatic C, *p*-methoxybenzyl), 110.95 (*C*Me), 83.8 (C-1), 68.35 (C-5), 55.2 (OMe), 26.9 and 26.6 (CMe_2), and 12.4 (SMe).

Anal. Calc. for $C_{17}H_{24}O_5S$: C, 59.98; H, 7.11. Found: C, 59.94; H, 7.15.

*Methyl 4-*O*-(p-methoxybenzyl)-1-thio-β-*D*-xylopyranoside* (**25**). — To a solution of **24** (2.42 g) in acetone (50 mL) was added M HCl acid (1 mL). The mixture was stirred for 1 h at room temperature, neutralized with solid $NaHCO_3$, and filtered through a Celite pad. The filtrate was concentrated and a solution of the residue in CH_2Cl_2 was washed with water, dried, and evaporated. Crystallization of the residue from ether gave **25** (1.93 g, 90%), m.p. 87–89°, $[\alpha]_D^{20}$ −50° (*c* 1.2, $CHCl_3$); n.m.r. ($CDCl_3$): ^1H, δ 7.20 and 6.90 (2 d, each 2 H, *J* 8.6 Hz, aromatic H), 4.20 (d, 1 H, $J_{1,2}$ 8.6 Hz, H-1), 3.77 (s, 3 H, OMe), and 2.15 (s, 3 H, SMe); ^{13}C, δ 159.3, 130.0, 129.4, and 113.9 (aromatic C, *p*-methoxybenzyl), 86.25 (C-1), 67.3 (C-5), 55.2 (OMe), and 12.1 (SMe).

Anal. Calc. for $C_{14}H_{20}O_5S$: C, 55.98; H, 6.71. Found: C, 56.15; H, 6.80.

*Methyl 2,3-di-*O*-benzyl-4-*O*-(p-methoxybenzyl)-1-thio-β-*D*-xylopyranoside* (**26**). — A solution of **25** (3.1 g) in *N,N*-dimethylformamide (40 mL) was treated with NaH (1.5 g; 50% mineral oil), followed by $PhCH_2Br$ (3.4 mL), and processed as described for the preparation of **9**. Column chromatography (5:1 hexane–EtOAc) of the product gave **26** (4.51 g, 91%), m.p. 53–56° (from EtOH), $[\alpha]_D^{20}$ +3° (*c* 1.2, $CHCl_3$); ^{13}C-n.m.r. ($CDCl_3$): δ 159.2, 130.1, 129.3, and 113.75 (aromatic C, *p*-methoxybenzyl), 138.5 and 137.9 (aromatic C-1, benzyl), 86.1 (C-1), 67.6 (C-5), 55.1 (OMe), and 12.9 (SMe).

Anal. Calc. for $C_{28}H_{32}O_5S$: C, 69.97; H, 6.71. Found: C, 69.92; H, 6.67.

Benzyl 4-O-(4-O-acetyl-2,3-di-O-benzyl-α- and β-D-xylopyranosyl)-2,3,6-tri-O-benzyl-β-D-glucopyranoside (**28** and **31**). — A solution of oxalyl chloride (1.7 mL) in CH_2Cl_2 (10 mL) was added dropwise at 0° to a solution of **14** (2.52 g) in CH_2Cl_2 (20 mL) containing *N,N*-dimethylformamide (0.2 mL). The mixture was kept for 30 min at room temperature and then evaporated. A solution of the residue in 1:1 hexane–EtOAc (25 mL) was filtered through a layer of silica gel (6 g) and the layer was washed with 1:1 hexane–EtOAc (20 mL). The combined filtrate and washings were evaporated to give 4-*O*-acetyl-2,3-di-*O*-benzyl-α-D-xylopyranosyl chloride (**15**) as a syrup (2.45 g, 95%), $[\alpha]_D^{19}$ +67° (*c* 1.1, CH_2Cl_2), which was used in the glycosylation step without purification; N.m.r. ($CDCl_3$): ^1H, δ 7.40–7.22 (m, 10 H, 2 Ph), 6.00 (d, 1 H, $J_{1,2}$ 3.73 Hz, H-1), and 1.95 (s, 3 H, OAc); ^{13}C, δ 169.85 (C=O), 138.2 and 137.2 (aromatic C-1), 93.35 (C-1), and 20.7 ($COCH_3$).

A solution of **15** (2.19 g, 5.6 mmol) in ether (20 mL) was added dropwise at 0° to a stirred mixture of **27** (2.02 g, 3.7 mmol), $AgClO_4$ (1.50 g, 7.2 mmol), and 2,4,6-trimethylpyridine (0.95 mL, 7.2 mmol) in ether (90 mL) with exclusion of moisture and light. The mixture was allowed to reach room temperature and stirred for 5 h at room temperature. Insoluble material was collected on a layer of Celite and washed with ether, and the combined filtrate and washings were washed successively with cold dil. H_2SO_4, aq. $NaHCO_3$, and water, dried, and evaporated. The residue was subjected to column chromatography (20:1 benzene–EtOAc). Eluted first was **28**, isolated as a syrup (0.90 g, 27%), $[\alpha]_D^{20}$ +4° (*c* 0.9), $CHCl_3$); R_F 0.52 (t.l.c. in 10:1 benzene–EtOAc); n.m.r. ($CDCl_3$): ^1H, δ 7.33–7.18 (m, 30 H, 5 Ph), 5.46 (d, 1 H, $J_{1,2}$ 3.1 Hz, H-1′), and 1.93 (s, 3 H, OAc); ^{13}C, δ 102.1 (C-1), 96.65 (C-1′), 69.5 (C-6), 60.2 (C-5′), and 20.8 ($COCH_3$).

Eluted next was **31** (1.84 g, 55%), m.p. 89–91° (from MeOH), $[\alpha]_D^{20}$ −10° (*c* 1.2, $CHCl_3$); R_F 0.43 (t.l.c. in 10:1 benzene–EtOAc); ^{13}C-n.m.r. ($CDCl_3$): δ 169.6 (C=O), 102.8 (C-1′), 102.4 (C-1), 68.05 (C-6), 62.3 (C-5′), and 20.8 ($COCH_3$).

Anal. Calc. for $C_{55}H_{58}O_{11}$: C, 73.81; H, 6.53. Found: C, 73.97; H, 6.67.

Benzyl 2,3,6-tri-O-benzyl-4-O-[2,3-di-O-benzyl-4-O-(p-methoxybenzyl)- α- and β-D-xylopyranosyl]-β-D-glucopyranoside (**29** and **32**). — (*a*) A mixture of **26** (0.40 g, 832 μmol), **27** (0.35 g, 647 μmol), and powdered molecular sieve 4A (3 g) in ether (20 mL) was stirred under argon for 30 min at room temperature. Methyl triflate (0.47 mL, 4 mmol) was injected through a rubber septum and the mixture was stirred for 6 h at room temperature. Triethylamine (1.2 mL) was added and the mixture was filtered through a Celite pad which was washed with ether. The combined filtrate and washings were evaporated and the residue was subjected to column chromatography (30:1→15:1 benzene–EtOAc). The first fraction afforded **29**, isolated as a syrup (0.12 g, 19%), $[\alpha]_D^{20}$ +13° (*c* 0.8, chloroform); R_F 0.5 (t.l.c. in 15:1 benzene–EtOAc, stepwise); n.m.r. ($CDCl_3$): ^1H, δ 5.25 (d, 1 H, $J_{1,2}$ 3.7 Hz, H-1′), and 3.77 (s, 3 H, OMe); ^{13}C, δ 159.2 and 113.7 (aromatic C, *p*-methoxybenzyl), 102.2 (C-1), 96.4 (C-1′), 69.3 (C-6), 60.9 (C-5′), and 55.2 (OMe).

The second fraction gave **32** (0.33 g, 52%), m.p. 109–110° (from MeOH), $[\alpha]_D^{20}$ +8° (*c* 1.2, $CHCl_3$); R_F 0.34 (t.l.c. in 15:1 benzene–EtOAc); ^{13}C-n.m.r. ($CDCl_3$): δ 159.2

and 113.35 (aromatic C, *p*-methoxybenzyl), 103.0 (C-1'), 102.35 (C-1), 68.0 (C-6), 63.6 (C-5'), and 55.15 (OMe).

Anal. Calc. for $C_{61}H_{64}O_{11}$: C, 75.29; H, 6.63. Found: C, 75.40; H, 6.54.

(*b*) A mixture of $CuBr_2$ (1.90 g, 8.5 mmol), Bu_4NBr (0.37 g, 11.5 mmol), and powdered molecular sieve 4A (10 g) in 1,2-dichloroethane (25 mL) and *N,N*-dimethylformamide (8 mL) was stirred under argon for 1 h at room temperature. A solution of **26** (2.73 g, 5.7 mmol) and **27** (2.05 g, 3.8 mmol) in 1,2-dichloroethane (15 mL) was added and the mixture was stirred for 3 days at room temperature and then filtered through a Celite pad. The solids were washed with $CHCl_3$ and the combined filtrate and washings were washed successively with aq. $NaHCO_3$, and water, dried, and evaporated. Column chromatography of the product, as described in (*a*), gave **29** (1.85 g, 50%) and **32** (0.81 g, 22%).

*Benzyl 2,3,6-tri-*O*-benzyl-4-*O*-(2,3-di-*O*-benzyl-α-*D*-xylopyranosyl)-β-*D*-glucopyranoside* (**30**). — (*a*) *O*-Deacetylation of **28** (0.75 g), as described for the preparation of **21**, afforded **30** as a syrup (0.66 g, 93%), $[\alpha]_D^{20}$ +7° (*c* 1.1, $CHCl_3$); R_f 0.36 (t.l.c. in 10:1 benzene–EtOAc); ^{13}C-n.m.r. ($CDCl_3$): δ 102.1 (C-1') and 99.2 (C-1).

(*b*) Compound **29** (1.72 g) was treated with ceric ammonium nitrate (1.94 g) in 9:1 MeCN–water (40 mL), as described for the preparation of **10**, followed by column chromatography (10:1 benzene–EtOAc) of the product, gave **30** (1.22 g, 81%).

*Allyl 2,3-di-*O*-benzyl-4-*O*-(2,3,4,6-tetra-*O*-benzyl-α-*D*-glucopyranosyl)-β-*D*-xylopyranoside* (**34**). — A solution of 2,3,4,6-tetra-*O*-benzyl-D-glucopyranose (2.80 g) in CH_2Cl_2 (20 mL) and *N,N*-dimethylformamide (0.1 mL) was treated with a solution of oxalyl chloride (1.35 mL) in CH_2Cl_2 (10 mL), and processed as described for the preparation of **15**, to give **33** (2.80 g, 95%), $[\alpha]_D^{20}$ +96° (*c* 1.5, C_6H_6); lit.[26] $[\alpha]_D$ +95° (C_6H_6), which was used in the coupling reaction without purification. ^{1}H-n.m.r. ($CDCl_3$): δ 6.07 (d, 1 H, $J_{1,2}$ 3.5 Hz, H-1).

A mixture of **10** (1.15 g, 3.1 mmol), $AgClO_4$ (1.48 g, 7.1 mmol), and 2,4,6-trimethylpyridine (0.94 mL, 7.1 mmol) in ether (20 mL) was treated with a solution of **33** (2.60 g, 4.65 mmol) in ether (20 mL), and processed as described for the reaction of **15** with **27**. Column chromatography (20:1 benzene–EtOAc) of the product gave **34** (2.08 g, 75%), m.p. 90–93° (from EtOH), $[\alpha]_D^{20}$ +30.5° (*c* 1.1, $CHCl_3$); n.m.r. ($CDCl_3$): ^{1}H, δ 6.01–5.87 (m, 1 H, –CH–) and 5.11 (d, 1 H, $J_{1,2}$ 3.7 Hz, H-1); ^{13}C, δ 133.9 and 117.35 (CH=CH_2), 103.1 (C-1), 98.8 (C-1'), 68.3 (C-6'), and 64.6 (C-5).

Anal. Calc. for $C_{56}H_{60}O_{10}$: C, 75.31; H, 6.77. Found: C, 75.40; H, 6.74.

*Methyl 2,3-*O*-isopropylidene-4-*O*-(2,3,4,6-tetra-*O*-benzyl-α-*D*-glucopyranosyl)-1-thio-β-*D*-xylopyranoside* (**35**). — A mixture of **22** (1.04 g, 4.7 mmol), $AgClO_4$ (2.2 g, 10.6 mmol), and 2,4,6-trimethylpyridine (2.1 mmol, 15.9 mmol) in ether (100 mL) was treated with a solution of **33** (3.96 g, 7.1 mmol) in ether (30 mL). Processing of the mixture, as described for the reaction of **15** with **27**, followed by column chromatography (20:1 benzene–EtOAc) of the product afforded **35** as a syrup (2.49 g, 71%), $[\alpha]_D^{20}$ +41.5° (*c* 1.2, $CHCl_3$); R_f 0.38 (t.l.c. in 10:1 benzene–EtOAc); n.m.r. ($CDCl_3$): ^{1}H, δ 5.38 (d, 1 H, $J_{1,2}$ 3.5 Hz, H-1), 2.22 (s, 3 H, SMe), and 1.48 (s, 6 H, CMe_2); ^{13}C, δ 111.1 (CMe_2), 95.9 (C-1'), 83.7 (C-1), 26.9 and 26.6 (CMe_2), and 12.3 (SMe).

Methyl 4-O-(2,3,4,6-tetra-O-benzyl-α-D-glucopyranosyl)-1-thio-β-D-xylopyranoside (**36**). — A solution of **35** (2.15 g) in acetone (60 mL) was treated with M HCl (1 mL), and processed as described for the preparation of **25**. The residue was purified by column chromatography (4:1 benzene–EtOAc) to give **36** (1.77 g, 87%), m.p. 129–131° (from EtOH, $[\alpha]_D^{20}$ +33° (*c* 1.1, $CHCl_3$); ^{13}C-n.m.r. ($CDCl_3$): δ 99.4 (C-1′), 86.1 (C-1), and 11.95 (SMe).

Anal. Calc. for $C_{40}H_{46}O_9S$: C, 68.35; H, 6.60. Found: C, 68.27; H, 6.54.

Methyl 2,3-di-O-benzyl-4-O-(2,3,4,6-tetra-O-benzyl-α-D-glucopyranosyl)-1-thio-β-D-xylopyranoside (**37**). — Treatment of **36** (1.48 g) in *N,N*-dimethylformamide (20 mL) with NaH (0.32 g; 50% mineral oil) and benzyl bromide (0.74 mL), as described for the preparation of **9**, followed by column chromatography (15:1 benzene–EtOAc) of the product, afforded **37** (1.65 g, 89%), m.p. 86–89° (from EtOH–CH_2Cl_2), $[\alpha]_D^{20}$ +25.5° (*c* 1.3, $CHCl_3$); ^{13}C-n.m.r. ($CDCl_3$): δ 99.0 (C-1′), 86.1 (C-1), and 12.9 (SMe).

Anal. Calc. for $C_{54}H_{58}O_9S$: C, 73.44; H, 6.62. Found: C, 73.56; H, 6.71.

2,3-Di-O-benzyl-4-O-(2,3,4,6-tetra-O-benzyl-α-D-glucopyranosyl)-D-xylopyranose (**38**). — Compound **34** (1.75 g) was boiled with tris(triphenylphosphine)rhodium(I) chloride (0.2 g) and 1,4-diazabicyclo[2.2.2]octane (0.8 g) in 8:3:1 EtOH–PhMe–water (36 mL) under reflux for 8 h. The mixture was processed and the product was treated with HgO (0.5 g) and $HgCl_2$ (0.5 g) in 9:1 acetone–water (25 mL), as described for the preparation of **12**. Column chromatography (15:1 benzene–EtOAc) of the product gave **38** (1.34 g, 80%), m.p. 104–108° (from EtOH), $[\alpha]_D^{21}$ +32° (*c* 1.2, $CHCl_3$); ^{13}C-n.m.r. ($CDCl_3$): δ 97.7 (C-1β), 91.1 (C-1α), and 98.8 (C-1′).

Anal. Calc. for $C_{53}H_{56}O_{10}$: C, 74.63; H, 6.62. Found: C, 74.55; H, 6.60.

Benzyl O-(2,3,4,6-tetra-O-benzyl-α-D-glucopyranosyl)-(1→4)-O-(2,3-di-O-benzyl-α-D-xylopyranosyl)-(1→4)-O-(2,3-di-O-benzyl-α-D-xylopyranosyl)-(1→4)-2,3,6-tri-O-benzyl-β-D-glucopyranoside (**40**). — (*a*) Compound **38** (1.1 g) was treated in CH_2Cl_2 (15 mL) containing *N,N*-dimethylformamide (0.1 mL) with a solution of oxalyl chloride (0.33 mL) in CH_2Cl_2 (3 mL) and processed as described for the preparation of **15** to give 2,3-di-*O*-benzyl-4-*O*-(2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl)-α-D-xylopyranosyl chloride (**38**) as an amorphous powder (1.04 g, 92%), $[\alpha]_D^{21}$ +72° (*c* 0.9, CH_2Cl_2), which was used in the glycosylation without purification. N.m.r. ($CDCl_3$): ^{1}H, δ 5.97 (d, 1 H, $J_{1,2}$ 3.7 Hz, H-1); ^{13}C, δ 98.9 (C-1′) and 93.5 (C-1).

A mixture of **30** (0.51 g, 598 μmol), $AgClO_4$ (0.3 g, 1.4 mmol), and 2,4,6-trimethylpyridine (0.2 mL, 1.5 mmol) in ether (18 mL) was treated with a solution of **38** (0.38 g, 952 μmol) in ether (5 mL), and processed as described for the reaction of **15** with **27**. Column chromatography (30:1→20:1 benzene–EtOAc, stepwise) of the product gave **40** as a syrup (0.59 g, 58%), $[\alpha]_D^{20}$ +32° (*c* 1.2, $CHCl_3$); R_f 0.39 (t.l.c. in 15:1 benzene–EtOAc), n.m.r ($CDCl_3$): δ 5.42 (d, 1 H, $J_{1''',2'''}$ 3.66 Hz, H-1‴), 5.23 (d, 1 H, $J_{1'',2''}$ 3.67 Hz, H-1″), and 5.07 (d, 1 H, $J_{1',2'}$ 3.66 Hz, H-1′); ^{13}C, 102.25 (C-1), 98.65 (C-1‴), 98.48 and 96.76 (C-1′, 1″), 69.23 and 68.25 (C-6, 6‴), and 61.43 and 61.03 (C-5′, 5″).

(*b*) The product obtained by treatment of a mixture of **30** (0.49 g, 574 μmol), **37** (0.76 g, 861 μmol) and powdered molecular sieve 4A (3 g) in ether (15 mL) with methyl

triflate (0.49 mL, 4.3 mmol), as described for the reaction of **26** with **27**, was subjected to column chromatography as described in (*a*), to afford **40** (0.42 g, 43%).

(*c*) The product obtained by treatment of a mixture of **30** (0.33 g, 386 μmol), $CuBr_2$ (0.19 g, 851 μmol), Bu_4NBr (62 mg, 192 μmol) in 1,2-dichloroethane (7.5 mL) and *N,N*-dimethylformamide (1.5 mL) with **37** (0.51 g, 578 μmol), as described for the reaction of **26** with **27**, was subjected to column chromatography as described in (*a*), to give **40** (66 mg, 10%).

O-α-D-*Glucopyranosyl-(1→4)*-O-α-D-*xylopyranosyl-(1→4)*-O-α-D-*xylopyranosyl-(1→4)*-D-*glucopyranose* (**1**). — A solution of **40** (1.21 g) in 2-methoxycthanol (40 mL) was hydrogenated in the presence of 10% Pd–C (1.0 g) at normal pressure for 1 day at room temperature. Insoluble material was collected on a Celite pad and washed with MeOH, and the combined filtrate and washings were evaporated. The residue was purified by column chromatography (15:10:1→5:5:1 $CHCl_3$–MeOH–water, stepwise) to give **1** as a hygroscopic, amorphous powder (0.36 g, 84%), $[\alpha]_D^{25}$ + 157° (*c* 1.2, H_2O); R_F 0.26 (t.l.c. in 15:10:1 $CHCl_3$–MeOH–water); $R_{Maltotetraose}$ 0.91 (l.c.); n.m.r. (D_2O): ^1H, δ 5.32 (d, 1 H, $J_{1''',2'''}$ 3.36 Hz, H-1‴), 5.20 (d, 0.4 H, $J_{1,2}$ 3.66 Hz, H-1α), 5.12 (d, 1 H, $J_{1'',2''}$ 3.66 Hz, H-1″), 5.09 (d, 1 H, $J_{1',2'}$ 3.97 Hz, H-1′), and 4.63 (d, 0.6 H, $J_{1,2}$ 7.39 Hz, H-1β); ^{13}C, δ 100.90 (C-1‴), 100.74 (C-1″), 100.48 and 100.38 (C-1′), 96.69 (C-1β), 92.83 (C-1α), 78.90 (C-4′, 4″), 78.79 (C-4), 61.98 (C-6‴), 61.60 (C-6′β), and 61.51 (C-5′, 5″, 6α).

Methylation[27] of a portion of **1**, followed by successive hydrolysis, borohydride reduction, acetylation, and g.l.c.[28] of the resulting products gave peaks corresponding to the peracetates of 2,3,4,6-tetra-*O*-methyl-D-glucitol, 2,3-di-*O*-methyl-D-xylitol, and 2,3,6-tri-*O*-methyl-D-glucitol in the ratio ~1:2:1.

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