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Studies of the molecular recognition of synthetic methyl β-lactoside analogues by *Ricinus communis* agglutinin

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ABSTRACT

The 2-, 3-, 6-, 2′-, 3′-, 4′-, and 6′-deoxy derivatives and the 3-*O*-methyl derivative of methyl β-lactoside have been synthesised and their binding to the galactose-specific agglutinin from *Ricinus communis* (RCA-120) has been investigated. The results indicate that HO-3,4,6 of the β-D-galactopyranose moiety are the key polar groups. The main difference from the closely related ricin lectin RCA-60 involves HO-6 of the D-glucopyranose moiety, which seems to contribute to the binding of the carbohydrate to RCA-60 but not to RCA-120.

INTRODUCTION

The specific interaction of carbohydrates with lectins and antibodies involves polar and short-range multipoint van der Waals interactions [1–4]. We have reported on the binding of methyl β-lactoside (**1**) and some analogues to ricin (RCA-60), the cytotoxic lectin isolated from the seeds of *Ricinus communis* [5]. The distributions of low-energy conformers of **1** and the 2- (**2**), 3- (**3**), 6- (**6**), 2′- (**7**), 3′- (**8**), 4′- (**9**), and 6′-deoxy (**10**) derivatives, the 3-*O*-methyl derivative (**4**), methyl 4-*O*-β-D-galactopyranosyl-β-D-xylopyranoside (**5**), 1,6-anhydrolactose (**11**), and lactal (**12**) were determined using molecular mechanics calculations and NMR spectroscopy [5]. The affinity of the lectin for disaccharide molecules **1**–**12** was also determined [5].

The seeds of *R. communis* also contain a galactopyranosyl-specific agglutinin (RCA-120) which has a molecular weight of 120 000 and consists of two A chains and two B chains with molecular weights of 29 500 and 37 000, respectively [6]. The polypeptide chains of RCA-60 and RCA-120 are closely related as demonstrated

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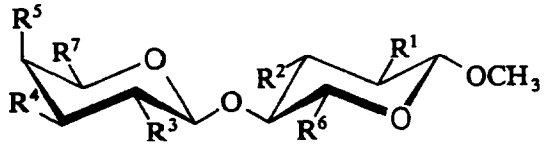
by the extensive cross-reactivity of antibodies directed against either the A or B chains [6], the similarity of the overall amino acid composition [6], and the homology of the first nineteen amino acids of the A chains and seventeen of the first eighteen amino acids of the B chains [7]. Both the A and B subunits of RCA-60 and RCA-120 are derived from a single polypeptide chain [8]. However, the lectins have different specificities for binding monosaccharide derivatives [6,9], oligosaccharides [10], and glycopeptides [11,12].

We now report the synthesis of the compounds **2–10**, previously used [5] to probe the combining site of RCA-60. The binding of these compounds to RCA-120 is also reported and the results are compared to those obtained for RCA-60.

RESULTS AND DISCUSSION

Methyl 2-deoxy-4-*O*-β-D-galactopyranosyl-β-D-*arabino*-hexopyranoside (methyl 2-deoxy-β-lactoside, **2**) was prepared from methyl 3′,4′-*O*-isopropylidene-β-lactoside (**11**) by selective allylation *via* the corresponding 2,3-*O*-dibutylstannylene derivative to give [13] methyl 2-*O*-allyl-3′,4′-*O*-isopropylidene-β-lactoside (**12**, 73%), characterised as the tetra-acetate **38**. Benzylation of **12** gave the 3,6,2′,6′-tetra-*O*-benzyl derivative (**13**, 78%), which was deallylated to afford methyl 3,6,2′,6′-tetra-*O*-benzyl-3′,4′-*O*-isopropylidene-β-lactoside (**14**, 57%). Treatment of crude **14** with sodium hydride and carbon disulfide, then with methyl iodide to give the corresponding xanthate, and subsequently with tributyltin hydride [14] yielded methyl 3,6,2′,6′-tetra-*O*-benzyl-2-deoxy-3′,4′-*O*-isopropylidene-β-lactoside (**15**), which, after hydrogenolysis and acid hydrolysis, gave **2** (73%).



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4 $R^1 = R^3 = R^4 = R^5 = OH$, $R^2 = CH_3$, $R^6 = R^7 = CH_2OH$
5 $R^1 = R^2 = R^3 = R^4 = R^5 = OH$, $R^6 = H$, $R^7 = CH_2OH$
6 $R^1 = R^2 = R^3 = R^4 = R^5 = OH$, $R^6 = CH_3$, $R^7 = CH_2OH$
7 $R^1 = R^2 = R^4 = R^5 = OH$, $R^3 = H$, $R^6 = R^7 = CH_2OH$
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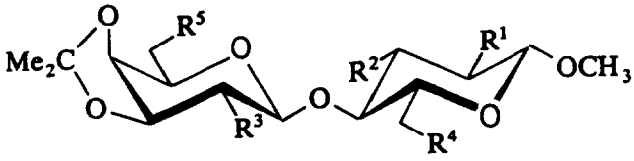
10 $R^1 = R^2 = R^3 = R^4 = R^5 = OH$, $R^6 = CH_2OH$, $R^7 = CH_3$
24 $R^1 = R^2 = R^3 = R^4 = R^5 = OAc$, $R^6 = H$, $R^7 = CH_2OAc$
29 $R^1 = R^2 = R^3 = R^5 = OH$, $R^4 = OAllyl$, $R^6 = R^7 = CH_2OH$
33 $R^1 = R^2 = R^3 = OH$, $R^4, R^5 = O\text{-}CS\text{-}O$, $R^6 = R^7 = CH_2OH$
41 $R^1 = R^2 = R^3 = R^5 = OAc$, $R^4 = OAllyl$, $R^6 = R^7 = CH_2OAc$



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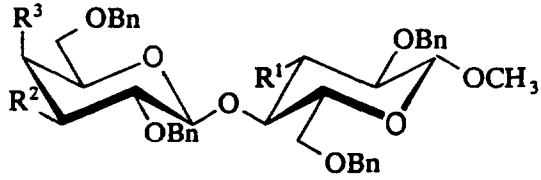
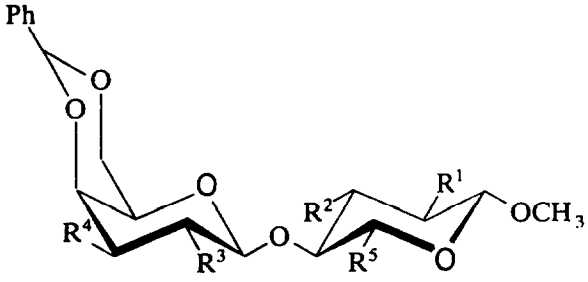
14 $R^2 = R^3 = R^4 = R^5 = OBn$, $R^1 = OH$
15 $R^2 = R^3 = R^4 = R^5 = OBn$, $R^1 = H$
25 $R^1 = R^4 = R^5 = OBn$, $R^2 = R^3 = OH$
26 $R^1 = R^4 = R^5 = OBn$, $R^2 = OH$, $R^3 = OC(OPh)S$
27 $R^1 = R^4 = R^5 = OBn$, $R^2 = OH$, $R^3 = H$
28 $R^1 = R^4 = R^5 = OBn$, $R^2 = H$, $R^3 = OH$
38 $R^2 = R^3 = R^4 = R^5 = OAc$, $R^1 = OAllyl$
39 $R^2 = R^3 = R^4 = R^5 = OBn$, $R^1 = OAc$



16 $R^1 = OH$, $R^2 = R^3 = OBn$
17 $R^1 = H$, $R^2 = R^3 = OBn$
18 $R^1 = OMe$, $R^2 = R^3 = OBn$
30 $R^1 = R^3 = OBn$, $R^2 = OAllyl$
31 $R^1 = R^3 = OBn$, $R^2 = OH$
32 $R^1 = R^3 = OBn$, $R^2 = H$
36 $R^1 = R^2 = OBn$, $R^3 = OH$
37 $R^1 = R^2 = OBn$, $R^3 = H$
40 $R^1 = OAc$, $R^2 = R^3 = OBn$
42 $R^1 = R^2 = OBn$, $R^3 = OAc$

Methyl 3-deoxy-4-*O*-β-D-galactopyranosyl-β-D-*ribo*-hexopyranoside (methyl 3-deoxy-β-lactoside, **3**) was prepared by partial benzylation [15] of methyl β-lactoside (**1**) to give the 2,6,2′,3′,4′,6′-hexa-*O*-benzyl derivative (**16**, 43%) followed by deoxygenation, as described above for **14**, to yield the 3-deoxy derivative (**17**, 79%), hydrogenolysis of which gave **3** (87%).

Methyl 3-*O*-methyl-β-lactoside (**4**, 83%) was obtained by conventional methylation of **16** to afford methyl 2,6,2′,3′,4′,6′-hexa-*O*-benzyl-3-*O*-methyl-β-lactoside (**18**, 88%) followed by hydrogenolysis.



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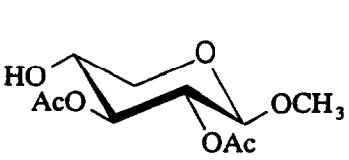
21 $R^1 = R^2 = R^3 = R^4 = OH$, $R^5 = CH_3$
34 $R^1 = R^2 = R^3 = OH$, $R^4 = OC(OPh)S$, $R^5 = CH_2OH$
35 $R^1 = R^2 = R^3 = R^4 = OBn$, $R^5 = CH_2OBn$



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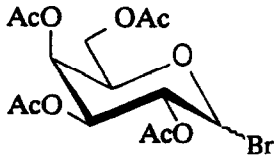

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Methyl 6-deoxy-β-lactoside (**6**, 82% from **20**) was synthesised from methyl 4′,6′-*O*-benzylidene-β-lactoside [16] (**19**) by iodination [17] to give the 6-deoxy-6-iodo derivative (**20**, 81%) followed by hydrogenolysis, to yield methyl 4′,6′-*O*-benzylidene-6-deoxy-β-lactoside (**21**), and acid hydrolysis.

Methyl 4-*O*-β-D-galactopyranosyl-β-D-xylopyranoside (**5**, 95%) was synthesised from methyl 2,3-di-*O*-acetyl-β-D-xylopyranoside [18] (**22**) by glycosylation with 2,3,4,6-tetra-*O*-acetyl-α-D-galactopyranosyl bromide (**23**) in the presence of mercuric cyanide and mercuric bromide, to give **24** (55%), followed by deacetylation.

Methyl 4-*O*-(2-deoxy-β-D-*lyxo*-hexopyranosyl)-β-D-glucopyranoside (methyl 2′-deoxy-β-lactoside, **7**) was prepared by tributyltin ether-mediated partial benzylation of **11**, which gave methyl 2,6,6′-tri-*O*-benzyl-3′,4′-*O*-isopropylidene-β-lactoside [15] (**25**, 49%). Treatment of **25** with phenoxythiocarbonyl chloride afforded the 2′-*O*-phenoxythiocarbonyl derivative (**26**, 62%) which reacted with tributyltin hydride to give the 2′-deoxy derivative (**27**, 84%). In some experiments, however, a mixture of **27** (45%) and the 3-deoxy derivative (**28**, 44%) was obtained probably due to 2 → 3 migration of the phenoxythiocarbonyl group. Hydrogenolysis of **27** followed by acid hydrolysis gave **7** (72%). Compound **7** has been synthesised [19] from 2,3:5,6:3′,4′-tri-*O*-isopropylidene-6′-*O*-triphenylmethyl-lactose dimethyl acetal.

Methyl 4-*O*-(3-deoxy-β-D-*xylo*-hexopyranosyl)-β-D-glucopyranoside (methyl 3′-deoxy-β-lactoside, **8**) was prepared from **1** by regioselective allylation [20–22], via the corresponding 3′,4′-*O*-stannylidene derivative, to give methyl 3′-*O*-allyl-β-lactoside

(**29**, 78%). Benzylation then gave the 2,3,6,2′,4′,6′-hexa-*O*-benzyl derivative (**30**, 82%), which was then deallylated to afford methyl 2,3,6,2′,4′,6′-hexa-*O*-benzyl-β-lactoside (**31**, 69%). Deoxygenation of **31**, as described for **14**, gave the 3′-deoxy derivative (**32**, 77%), hydrogenolysis of which gave **8** (82%). A shorter route involving regioselective thioacylation of HO-3′ of **1** by treatment with dibutyltin oxide and phenoxythiocarbonyl chloride [23] failed. No 3′-*O*-phenoxythiocarbonyl derivative could be detected and the cyclic thiocarbonate **33** was isolated. However, reaction of **19** with dibutyltin oxide and phenoxythiocarbonyl chloride gave methyl 4′,6′-*O*-benzylidene-3′-*O*-phenoxythiocarbonyl-β-lactoside (**34**, 82%), treatment of which with tributyltin hydride and subsequent removal of the 4′,6′-*O*-benzylidene group afforded **8** (68%). Compound **8** has been synthesised [19] from 3-deoxy-D-*xylo*-hexose.

Methyl 4-*O*-(4-deoxy-β-D-*xylo*-hexopyranosyl)-β-D-glucopyranoside (methyl 4′-deoxy-β-lactoside, **9**) was prepared by benzylation of **19** to give methyl 2,3,6,2′,3′-penta-*O*-benzyl-4′,6′-*O*-benzylidene-β-lactoside (**35**, 88%) which, after treatment with sodium cyanoborohydride [24], yielded methyl 2,3,6,2′,3′,6′-hexa-*O*-benzyl-β-lactoside (**36**, 86%), characterised as the 4′-acetate **42**. Reaction of **36** with carbon disulfide and methyl iodide and subsequently with tributyltin hydride gave the 4′-deoxy derivative (**37**, 85%), hydrogenolysis of which afforded **9** (80%). Compound **9** has also been synthesised by a different route [19].

Methyl 6′-deoxy-β-lactoside (**10**) was synthesised from **18** according to Bock and Adelhorst [19].

The distributions of the low-energy conformers in solutions of **1–10** are similar [5] and consist of a major form (ϕ_H 49°, ψ_H 5°) with smaller proportions of three other conformers (ϕ_H/ψ_H 24°/−59°, 22°/−32°, and 6°/−44°). The apparent dissociation constants for the binding of these derivatives to RCA-120 were determined as reported [5]. Each of the plots of the reciprocal of the lectin fraction bound to Sepharose 6B in the presence of the disaccharide derivative versus the concentration of the latter could be adjusted to a straight line with a mean correlation coefficient of 0.990. The existence of two binding sites in the B chains of RCA-120 has been demonstrated [25]. The linearity of the binding data now reported suggests similar affinities of each site for β-galactosides. However, the possibility that only one kind of site is operative for binding to the Sepharose 6B matrix cannot be excluded.

The apparent dissociation constants are given in Table I. The values for galactose, lactose, lactal, and 1,6-anhydrolactose have also been included and similar data [5] for RCA-60 are included for comparative purposes.

As expected, deoxygenation in the D-galactopyranose moiety strongly influences the binding. As found [5] for RCA-60, HO-4′, HO-3′, and HO-6′ are key polar groups in the interaction with RCA-120. These results are not in complete agreement with data on deoxyfluoro derivatives, which suggested [26] that HO-3′ and HO-4′ act as hydrogen donors to charged residues of the lectin, but that HO-6′ is not involved in binding to the lectin.

TABLE I

Apparent dissociation constants for the binding of methyl β-lactoside derivatives to agglutinin and ricin[5]

Compound	K_d (μM)	
	RCA-60	RCA-120
Galactose	232±20	227±21
Lactose	70± 6	56±11
1	56± 5	45± 9
2	59± 9	56± 3
3	27± 9	33± 8
4	311± 3	308±16
5	114± 4	50±10
6	109± 4	48± 7
7	318±13	423±15
8	1400±45	1100±30
9	3040±63	1600±17
10	1600±18	1200±62
1,6-Anhydrolactose	122± 5	88±14
Lactal	43±12	40± 7

There is also a smaller polar interaction involving HO-2′, as indicated by the values for **7**. It has been suggested [26] that HO-2′ is involved in two hydrogen bonds, but they were not specified. RCA-60, but not RCA-120, is inhibited by 2-acetamido-2-deoxy-D-galactose [11], whereas the 2′-deoxy derivative **7** has a similar effect on each lectin. These facts suggest the presence in RCA 60 of a locus for the acetamido group of the sugar, which is absent from RCA-120.

The D-glucopyranose unit is also involved in the binding. Comparison of the values (Table I) for D-galactose, 1,6-anhydrolactose, lactose, and methyl β-lactoside (**1**) indicates that the $^4C_1$ conformation of this unit is important for recognition and binding. As for RCA-60 [5], an enhancement of the binding is observed for the 3-deoxy derivative (**3**), which may indicate a non-polar interaction of this region of the molecule with the protein [27–32] or reduced steric hindrance with concomitant strengthening of other hydrogen bonds. The presence of a methyl group at position 3 (**4**), which precludes hydrogen bonding with O-5′, decreases the strength of the binding.

The specificities of the two lectins towards the disaccharide structures studied are similar. Only a different influence of the hydroxymethyl group at C-5 of the disaccharide derivatives is observed. Thus, the values for the binding of **5** and **6** to RCA-120 are similar to that of methyl β-lactoside (**1**), whereas the affinity of RCA-60 for these compounds decreases significantly. These facts suggest that the hydrophilic substituent at C-5 contributes to the binding to RCA-60 but not to RCA-120.

EXPERIMENTAL

General.—Melting points were determined on a Kofler hot-stage apparatus and are uncorrected. TLC was performed on Silica Gel GF_{254} (Merck) with detection by charring with H_2SO_4. Column chromatography was performed on silica gel (Merck, 70–230 mesh). ^{1}H-NMR spectra were recorded with a Varian XL-300 (300 MHz) spectrometer, and ^{13}C-NMR spectra with a Bruker AM-200 (50 MHz) spectrometer. Optical rotations were determined with a Perkin–Elmer 141 polarimeter.

RCA-120 was obtained from *Ricinus communis* seeds (Jardín Botánico, C.S.I.C., Madrid) as reported [6]. The lectin was separated from RCA-60 by gel filtration on Sephadex G-100 and enzymically labelled with ^{125}I in the presence of 0.1 M lactose, using the lactoperoxidase method [33]. ^{125}I-RCA-120 was indistinguishable from the unlabelled protein as determined by sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) and autoradiography. The concentration of the lectin was determined spectrophotometrically using a value [34] of $A^{1\%}_{280}$ of 14.

The affinity of RCA-120 for the sugar derivatives was determined from the amounts of ^{125}I-RCA-120 bound to Sepharose 6B in the presence of different concentrations of the carbohydrate derivative. Aliquots (200 μL) of sedimented Sepharose 6B (Pharmacia), previously washed with 5 mM phosphate buffer (pH 7.2) containing 0.2 M NaCl (PBS), were incubated with 10 μL of ^{125}I-RCA-120 solution (40.000 cpm, 0.7 μM final concentration) and 0.3 mL of the carbohydrate solution in PBS (from 50 μM to 3 mM, final concentration). After storage for 1 h at room temperature with occasional shaking, each sample was centrifuged and the radioactivity of the supernatant solution was counted in an LKB Mini-gamma counter. A time course of the binding of ^{125}I-lectin to Sepharose 6B showed that the equilibrium of the binding had been reached. Under the experimental conditions used, $> 80\%$ of RCA-120 was bound to Sepharose in the absence of sugar, and $< 3\%$ of the radioactivity applied was trapped on the gel in the presence of 0.1 M lactose (final concentration).

The apparent dissociation constants were calculated taking into account that:

$$M_t = [M] + [ML_1] + [ML_2]$$

where M_t is the total amount of RCA-120 in the incubation mixture, and L_1 and L_2 refer to Sepharose 6B and sugar, respectively. The above expression can be substituted in terms of the dissociation constants of the complexes of RCA-120 with Sepharose (K_1) and with sugar (K_2); referred to $[ML_1]$ and rearranged to:

$$\frac{M_t}{[ML_1]} = 1 + \frac{K_1}{[L_1]} + \frac{K_1[L_2]}{[L_1]K_2}$$

The concentration of free ligands $[L_1]$ and $[L_2]$ can be considered equal to the total amount present in the incubation mixture since their concentrations are much larger than the concentration of RCA-120. Therefore, a plot of the reciprocal of the RCA-120 fraction bound to Sepharose 6B (M_t/ML_1) versus the sugar concen-

tration $[L_2]$ should give a straight line since $[L_1]$ is a fixed value, being $K_2 =$ (intercept − 1)/slope.

Methyl 2-deoxy-β-lactoside (methyl 2-deoxy-4-O-β-D-galactopyranosyl-β-D-arabino-hexopyranoside, 2).—A mixture of methyl β-lactoside [35] (**1**; 1.45 g, 4.1 mmol), *N*,*N*-dimethylformamide (2.5 mL), acetone (80 mL), 2,2-dimethoxypropane (2.5 mL), and concd H_2SO_4 (2.5 μL) was boiled under reflux until dissolution was complete (45 min). The mixture was cooled to room temperature, and the white solid was collected, and washed with cold acetone to give methyl 3′,4′-*O*-isopropylidene-β-lactoside (**11**; 1.1 g, 68%), mp 220–224°, $[\alpha]_D$ +17° (*c* 1.16, H_2O). ^{1}H-NMR data ($CDCl_3$): δ 4.48 (d, 1 H, $J_{1',2'}$ 8.3 Hz, H-1′), 4.39 (d, 1 H, $J_{1,2}$ 7.9, H-1), 4.35 (dd, 1 H, $J_{3',4'}$ 5.4, $J_{4',5'}$ 1.8 Hz, H-4′), 4.20 (dd, 1 H, $J_{2',3'}$ 7.6 Hz, H-3′), 4.08 (ddd, 1 H, $J_{5',6'a}$ 4.3, $J_{5',6'b}$ 7.7 Hz, H-5′), 3.96 (dd, 1 H, $J_{5,6a}$ 1.8, $J_{6a,6b}$ 12.1 Hz, H-6a), 3.89–3.75 (m, 3 H, H-6b,6′a,6′b), 3.67–3.56 (m, 3 H, H-3,4,5), 3.56 (s, 3 H, OMe), 3.50 (t, 1 H, H-2′), 3.30 (m, 1 H, H-2), 1.53 and 1.38 (2 s, each 3 H, CMe_2).

Anal. Calcd for $C_{16}H_{28}O_{11}$: C, 48.48; H, 7.08. Found: C, 48.50; H, 7.08.

A mixture of **11** (1.50 g, 3.8 mmol), dibutyltin oxide (1.04 g, 4.18 mmol), powdered 4A molecular sieves (6 g), and acetonitrile (70 mL) was boiled under reflux overnight under Ar. *N*-Methylimidazole (0.36 mL, 4.52 mmol) and allyl bromide (35 mL) were then added and boiling was continued for 48 h. The mixture was cooled, filtered through Celite, and concentrated. Column chromatography (12:1 EtOAc–MeOH) of the residue gave methyl 2-*O*-allyl-3′,4′-*O*-isopropylidene-β-lactoside (**12**), isolated as a syrup which was treated conventionally with acetic anhydride–pyridine to give the syrupy 2,6,2′,6′-tetra-acetate **38**, $[\alpha]_D^{20}$ +21° (*c* 0.5, $CHCl_3$). ^{1}H-NMR data (C_6D_6): δ 5.82 (m, 1 H, CH_2=C*H*CH_2), 5.44 (t, 1 H, $J_{2,3} = J_{3,4} = 9.6$ Hz, H-3), 5.26 (m, 2 H, CH_2=CH and H-2′), 5.02 (m, 1 H, CH_2=CH), 4.49 (dd, 1 H, $J_{5,6a}$ 1.8, $J_{6a,6b}$ 11.9 Hz, H-6a), 4.49–4.32 (m, 3 H, H-6′a,6′b and CH_2=CHCH_2), 4.39 (d, 1 H, $J_{1',2'}$ 8.2 Hz, H-1′), 4.28 (dd, 1 H, $J_{5,6b}$ 5.0 Hz, H-6b), 4.05 (d, 1 H, $J_{1,2}$ 7.6 Hz, H-1), 4.04 (m, 1 H, CH_2=CHCH_2), 3.92 (dd, 1 H, $J_{2',3'}$ 6.5, $J_{3',4'}$ 5.4 Hz, H-3′), 3.78 (t, 1 H, $J_{3,4} = J_{4,5} = 9.6$ Hz, H-4), 3.72–3.64 (m, 2 H, H-4′,5′), 3.37 (dd, 1 H, H-2), 3.30 (ddd, 1 H, H-5), 3.24 (s, 3 H, OMe), 2.00, 1.86, 1.74, and 1.68 (4 s, each 3 H, 4 Ac), 1.50 and 1.14 (2 s, each 3 H, CMe_2).

A solution of **12** (0.90 g, 2.06 mmol) in *N*,*N*-dimethylformamide (40 mL) was treated with NaH (0.30 g) and then with benzyl bromide (2 mL) overnight at room temperature. Methanol (15 mL) and then water (20 mL) were added, the mixture was extracted with $CHCl_3$, and the extract was dried (Na_2SO_4) and concentrated. Column chromatography (6:1 hexane–EtOAc) of the residue gave methyl 2-*O*-allyl-3,6,2′,6′-tetra-*O*-benzyl-3′,4′-*O*-isopropylidene-β-lactoside (**13**, 1.28 g, 78%), isolated as a syrup, $[\alpha]_D^{20}$ +6° (*c* 1.48, $CHCl_3$). ^{1}H-NMR data ($CDCl_3$): δ 7.42–7.22 (m, 20 H, 4 Ph), 5.93 (m, 1 H, CH_2=C*H*), 5.26 (m, 1 H, CH_2=CH), 5.13 (m, 1 H, CH_2=CH), 4.91 and 4.74 (ABq, 2 H, *J* 10.7 Hz, CH_2Ph), 4.79 and 4.66 (ABq, 2 H, *J* 11.8 Hz, CH_2Ph), 4.56 and 4.41 (ABq, 2 H, *J* 12.3 Hz, CH_2Ph), 4.50 and 4.31

(ABq, 2 H, J 12.2 Hz, CH_2Ph), 4.40 (d, 1 H, $J_{1',2'}$ 8.0 Hz, H-1'), 4.34 (m, 1 H, CH_2=CHCH_2), 4.23 (d, 1 H, $J_{1,2}$ 7.8 Hz, H-1), 4.18 (m, 1 H, CH_2=CHCH_2), 4.10 (dd, 1 H, $J_{3',4'}$ 5.6, $J_{4',5'}$ 1.7 Hz, H-4'), 4.02 (t, 1 H, H-3'), 3.91 (t, 1 H, $J_{3,4} = J_{4,5} = 9.2$ Hz, H-4), 3.76 (dd, 1 H, $J_{5,6a}$ 4.2, $J_{6a,6b}$ 11.3 Hz, H-6a), 3.73 (dd, $J_{5,6b}$ 1.8 Hz, H-6b), 3.71–3.63 (m, 2 H, H-6'a,6'b), 3.53 (m, 1 H, H-5'), 3.52 (s, 3 H, OMe), 3.51 (t, 1 H, $J_{2,3} = J_{3,4} = 9.2$ Hz, H-3), 3.39 (ddd, 1 H, H-5), 3.34 (dd, 1 H, $J_{2',3'}$ 6.7 Hz, H-2'), 3.24 (dd, 1 H, H-2), 1.41 and 1.35 (2 s, each 3 H, CMe_2).

A solution of **13** (0.80 g, 1 mmol) in Me_2SO (10 mL) was stirred with potassium *tert*-butoxide (0.36 g, 3.2 mmol) at 95° for 2 h, then cooled, water (150 mL) was added, and the mixture was extracted immediately with ether. The extract was dried ($MgSO_4$) and concentrated. A solution of the residue in 10:1 acetone–water (40 mL) was treated with yellow mercuric oxide (0.36 g). A solution (10 mL) of mercuric chloride (0.36 g) in 10:1 acetone–water was then added during 40 min. After 1 h, the mixture was filtered through Celite and concentrated, and the residue was partitioned between water and ether. The ether phase was washed with aq 10% KI, dried ($MgSO_4$), and concentrated. Column chromatography (2:1 hexane–EtOAc) of the residue gave methyl 3,6,2',6'-tetra-*O*-benzyl-3',4'-*O*-isopropylidene-β-lactoside (**14**; 0.43 g, 57%), $[\alpha]_D^{20}$ −5° (*c* 1.0, $CHCl_3$). A small portion of **14** was treated conventionally with acetic anhydride–pyridine to give the syrupy 2-acetate **39**. ^{1}H-NMR data ($CDCl_3$): δ 7.22–7.35 (m, 30 H, 6 Ph), 4.97 (dd, 1 H, $J_{1,2}$ 8.0, $J_{2,3}$ 9.4 Hz, H-2), 4.90 and 4.60 (ABq, 2 H, J 11.4 Hz, CH_2Ph), 4.80 and 4.67 (ABq, 2 H, J 11.8 Hz, CH_2Ph), 4.57 and 4.42 (ABq, 2 H, J 12.2 Hz, CH_2PH), 4.47 and 4.31 (ABq, 2 H, J 12.0 Hz, CH_2Ph), 4.41 (d, 1 H, $J_{1',2'}$ 8.0 Hz, H-1'), 4.30 (d, 1 H, H-1), 4.09 (dd, 1 H, $J_{3',4'}$ 5.6, $J_{4',5'}$ 1.8 Hz, H-4'), 4.03 (bt, 1 H, H-3'), 4.01 (t, 1 H, $J_{3,4} = J_{4,5} = 9.3$ Hz, H-4), 3.83 (dd, 1 H, $J_{5,6a}$ 4.2, $J_{6a,6b}$ 11.0 Hz, H-6a), 3.74 (dd, 1 H, $J_{5,6b}$ 1.9 Hz, H-6b), 3.70–3.49 (m, 3 H, H-5',6'a,6'b), 3.60 (t, 1 H, $J_{2,3} = J_{3,4} = 9.4$ Hz, H-3), 3.46 (s, 3 H, OMe), 3.45 (ddd, 1 H, H-5), 3.35 (dd, 1 H, $J_{2',3'}$ 6.6 Hz, H-2'), 1.97 (s, 3 H, Ac), 1.41 and 1.34 (2 s, each 3 H, CMe_2).

A solution of **14** (0.40 g, 0.53 mmol) in tetrahydrofuran (25 mL) was treated with NaH (30 mL) and then CS_2 (45 μL, 0.58 mmol) under Ar with stirring. When the mixture became orange in colour (after ~ 90 min), MeI (1 mL) was added, and the mixture was left overnight at room temperature. Water (25 mL) was added, the mixture was repeatedly extracted with ether, and the combined extracts were dried ($MgSO_4$) and concentrated. A solution of the residue in toluene (40 mL) was boiled under reflux and treated with α,α'-azobisisobutyronitrile (10 mg). A solution of tributyltin hydride (0.2 mL, 0.58 mmol) in toluene (2 mL) was added during 45 min, and the mixture was stirred overnight, then concentrated to dryness to give crude methyl 3,6,2',6'-tetra-*O*-benzyl-2-deoxy-3',4'-*O*-isopropylidene-β-lactoside (**15**).

A solution of **15** (0.20 g, 0.27 mmol) in 1:5 EtOAc–EtOH was treated overnight with H_2 in the presence of 10% Pd–C (60 mg), then filtered through Celite, and concentrated. The residue was treated with aq 50% acetic acid at 65° for 1 h, then concentrated to give **2** (67 mg, 73%). Crystallisation from aq EtOH gave **2**, mp

148–149°, $[\alpha]_D^{20}$ −4.8° (*c* 1.0, H_2O). NMR data (D_2O): ^1H, δ 4.65 (dd, 1 H, $J_{1,2eq}$ 2.0, $J_{1,2ax}$ 9.9 Hz, H-1), 4.44 (d, 1 H, $J_{1',2'}$ 7.7 Hz, H-1'), 3.99 (dd, 1 H, $J_{5,6a}$ 1.5, $J_{6a,6b}$ 12.2 Hz, H-6a), 3.92 (bd, 1 H, $J_{3',4'}$ 3.4, $J_{4',5'} < 0.5$ Hz, H-4'), 3.85 (m, 1 H, H-3), 3.81 (dd, 1 H, $J_{5,6b}$ 4.9 Hz, H-6b), 3.80–3.72 (m, 2 H, H-6'a,6'b), 3.71 (bdd, 1 H, $J_{5',6'a}$ 8.3, $J_{5',6'b}$ 3.8 Hz, H-5'), 3.66 (dd, 1 H, $J_{2',3'}$ 9.9 Hz, H-3'), 3.54 (dd, 1 H, H-2'), 3.52 (s, 3 H, OMe), 3.51 (m, 2 H, H-4,5), 2.28 (ddd, 1 H, $J_{2eq,3}$ 5.2, $J_{2eq,2ax}$ 12.4 Hz, H-2*eq*), 1.48 (dt, 1 H, $J_{2ax,3} = J_{2ax,2eq} = 12.1$ Hz, H-2*ax*); ^{13}C, δ 104.4 (C-1'), 101.8 (C-1), 81.6 (C-4), 76.7 (C-5'), 76.1 (C-5), 73.9 (C-3'), 72.3 (C-2'), 70.5 (C-3), 69.8 (C-4'), 62.2 (C-6'), 61.7 (C-6), 57.8 (OCH_3), 38.5 (C-2).

Anal. Calcd for $C_{13}H_{24}O_{10}$: C, 45.88; H, 7.06. Found: C, 45.65; H, 7.04.

Methyl 3-deoxy-β-lactoside (methyl 3-deoxy-4-O-β-D-galactopyranosyl-β-D-ribo-hexopyranoside, **3***).*—Methyl β-lactoside (**1**; 1 g, 2.8 mmol) was stirred with powdered KOH (1.5 g) and benzyl chloride (6 mL) at 100° for 45 min. The mixture was cooled to room temperature, $CHCl_3$ (60 mL) was added, and the solution was washed with water, 0.5 M H_2SO_4, and water, dried (Na_2SO_4), and concentrated. Column chromatography (6:1 hexane–EtOAc) of the residue gave methyl 2,6,2',3',4',6'-hexa-*O*-benzyl-β-lactoside (**16**; 1.08 g, 43%), isolated as a syrup, $[\alpha]_D^{20}$ +3.8° (*c* 1.0, $CHCl_3$). NMR data ($CDCl_3$): ^1H, δ 7.38–7.12 (m, 30 H, 5 Ph), 4.84 and 4.48 (ABq, 2 H, *J* 11.7 Hz, CH_2Ph), 4.78 and 4.73 (ABq, 2 H, *J* 11.2 Hz, CH_2Ph), 4.77 (s, 2 H, CH_2Ph), 4.64 (s, 2 H, CH_2Ph), 4.40 and 4.33 (ABq, 2 H, *J* 12.0 Hz, CH_2Ph), 4.28 and 4.21 (ABq, 2 H, *J* 11.9 Hz, CH_2Ph), 4.23 (d, 1 H, $J_{1',2'}$ 7.7 Hz, H-1'), 4.22 (d, 1 H, $J_{1,2}$ 7.7 Hz, H-1), 3.76 (bd, 1 H, $J_{3',4'}$ 2.8, $J_{4',5'} < 0.5$ Hz, H-4'), 3.71 (dd, 1 H, $J_{2',3'}$ 9.7 Hz, H-2'), 3.69 (m, 1 H, $J_{2,3}$ 8.8 Hz, H-3), 3.46 (s, 3 H, OMe), 3.39 (dd, 1 H, H-3'), 3.24 (dd, 1 H, H-2), 1.51 (s, 1 H, OH); ^{13}C, δ 138.8, 138.3, 138.2, 138.1, 138.0, 137.4 (6 C_{ipso} Ph), 128.3–127.1 (Ph), 103.8 and 103.6 (C-1,1'), 82.2, 81.4, 81.3, 78.8, 75.0, 74.1, 73.5 and 73.1 (C-2/5 and C-2'/5'), 75.3, 74.5 (2 C), 73.5, 72.9, and 72.8 (O*C*H_2Ph), 68.6 and 68.5 (C-6,6'), 56.9 (OCH_3).

Anal. Calcd for $C_{55}H_{60}O_{11}$: C, 73.66; H, 6.70. Found: C, 73.42; H, 6.79.

Conventional treatment of **16** with acetic anhydride–pyridine gave the syrupy 3-acetate **40**. ^1H-NMR data ($CDCl_3$): δ 7.15–7.31 (m, 30 H, 5 Ph), 5.06 (t, 1 H, $J_{2,3} = J_{3,4} = 9.6$ Hz, H-3), 4.86 and 4.44 (ABq, 2 H, *J* 11.5 Hz, CH_2PH), 4.75 and 4.52 (ABq, 2 H, *J* 12.0 Hz, CH_2Ph), 4.67 (s, 2 H, CH_2Ph), 4.61 (s, 2 H, CH_2Ph), 4.45 and 4.29 (ABq, 2 H, *J* 12.1 Hz, CH_2Ph), 4.36 and 4.31 (ABq, 2 H, *J* 11.6 Hz, CH_2Ph), 4.26 (d, 1 H, $J_{1,2}$ 7.6 Hz, H-1), 4.16 (d, 1 H, $J_{1',2'}$ 7.7 Hz, H-1'), 3.77 (bd, 1 H, $J_{3',4'}$ 2.4, $J_{4',5'} < 0.5$ Hz, H-4'), 3.74 (t, 1 H, $J_{3,4} = J_{4,5} = 9.6$ Hz, H-4), 3.67 (dd, 1 H, $J_{5,6a}$ 3.9, $J_{6a,6b}$ 10.8 Hz, H-6a), 3.59 (dd, 1 H, $J_{5,6b}$ 1.4 Hz, H-6b), 3.57 (dd, 1 H, $J_{2',3'}$ 9.5 Hz, H-2'), 3.47 (s, 3 H, OMe), 3.46 (m, 2 H, H-6'a,6'b), 3.36 (ddd, 1 H, H-5), 3.28 (m, 1 H, H-5'), 3.27 (dd, 1 H, H-3'), 3.25 (dd, 1 H, H-2), 1.77 (s, 3 H, Ac).

A solution of **16** (90 mg, 0.1 mmol) in tetrahydrofuran (5 mL) was stirred with NaH (6 mg, 0.2 mmol) under Ar at room temperature for 15 min. Carbon disulfide (0.05 mL) was added, followed by MeI (0.5 mL), α,α'-azobisisobutyronitrile (10 mg), and tributyltin hydride (0.03 mL, 0.1 mmol), as described for the conversion

14 → **15**. Column chromatography (6 : 1 hexane–EtOAc) of the product gave methyl 2,6,2′,3′,4′,6′-hexa-*O*-benzyl-3-deoxy-β-lactoside (**17**; 63 mg, 71%), isolated as a syrup, $[\alpha]_D^{20}$ −7.6° (*c* 0.1, $CHCl_3$). ^{1}H-NMR data ($CDCl_3$): δ 7.32–7.23 (m, 30 H, 6 Ph), 4.91 and 4.60 (ABq, 2 H, *J* 11.7 Hz, C*H*$_2$Ph), 4.78 (s, 2 H, C*H*$_2$Ph), 4.72 and 4.57 (ABq, 2 H, *J* 12.0 Hz, C*H*$_2$Ph), 4.69 (s, 2 H, C*H*$_2$Ph), 4.43 and 4.35 (ABq, 2 H, *J* 12.1 Hz, C*H*$_2$Ph), 4.42 (s, 2 H, C*H*$_2$Ph), 4.31 (d, 1 H, $J_{1',2'}$ 7.8 Hz, H-1′), 4.26 (d, 1 H, $J_{1,2}$ 7.6 Hz, H-1), 3.86 (bd, 1 H, $J_{3',4'}$ 2.8, $J_{4',5'}$ < 0.8 Hz, H-4′), 3.78–3.54 (m, 6 H, H-4,6a,6b,2′,6′a,6′b), 3.54 (s, 3 H, OMe), 3.53–3.40 (m, 2 H, H-5,5′), 3.42 (dd, 1 H, $J_{2',3'}$ 9.8 Hz, H-3′), 3.27 (ddd, 1 H, $J_{2,3ax}$ 12.0, $J_{2,3eq}$ 4.9 Hz, H-2), 2.55 (dt, 1 H, $J_{2,3eq} = J_{3eq,4} = 5.1$, $J_{3eq,3ax}$ 12.1 Hz, H-3*eq*), 1.67 (dt, 1 H, $J_{2,3ax} = J_{3ax,4} =$ 12.1 Hz, H-3*ax*).

Anal. Calcd for $C_{55}H_{60}O_{10}$: C, 75.00; H, 6.82. Found: C, 75.27; H, 6.88.

A solution of **17** (60 mg, 0.07 mmol) in 5 : 1 EtOH–EtOAc (6 mL) was treated with H_2 in the presence of 10% Pd–C (30 mg) for 12 h. Crystallisation of the product from aq EtOH gave **3**, mp 184–185°, $[\alpha]_D^{20}$ −1.4° (*c* 0.5, H_2O). NMR data (D_2O): ^{1}H, δ 4.44 (d, 1 H, $J_{1',2'}$ 8.0 Hz, H-1′), 4.32 (d, 1 H, $J_{1,2}$ 7.8 Hz, H-1), 3.95 (dd, 1 H, $J_{5,6a}$ 2.3, $J_{6a,6b}$ 12.2 Hz, H-6a), 3.91 (dd, 1 H, $J_{3',4'}$ 3.5, $J_{4',5'}$ 0.8 Hz, H-4′), 3.77 (dd, 1 H, $J_{5',6'a}$ 7.4, $J_{6'a,6'b}$ 11.5 Hz, H-6′a), 3.75 (ddd, 1 H, $J_{3eq,4}$ 11.7, $J_{3ax,4}$ 5.0, $J_{4,5}$ 9.9 Hz, H-4), 3.74 (dd, 1 H, $J_{5,6b}$ 5.8 Hz, H-6b), 3.73 (dd, 1 H, $J_{5',6'b}$ 4.4 Hz, H-6′b), 3.67 (ddd, 1 H, H-5′), 3.62 (dd, 1 H, $J_{2',3'}$ 10.0 Hz, H-3′), 3.56 (ddd, 1 H, H-5), 3.55 (s, 3 H, OMe), 3.48 (dd, 1 H, H-2′), 3.46 (ddd, 1 H, $J_{2,3eq}$ 11.6, $J_{2,3ax}$ 4.9 Hz, H-2), 2.58 (dt, 1 H, $J_{3eq,3ax}$ 11.8 Hz, H-3*eq*), 1.65 (dt, 1 H, H-3*ax*); ^{13}C, δ 105.6 (C-1), 104.1 (C-1′), 78.7 (C-4), 75.4 (C-5′), 74.2 (C-5), 73.1 (C-3′), 71.2 (C-2′), 68.9 (C-4′), 67.9 (C-2), 61.2 (C-6′), 60.9 (C-6), 57.1 (OCH_3), 37.8 (C-3).

Anal. Calcd for $C_{13}H_{24}O_{10}$: C, 45.88; H, 7.06. Found: C, 45.50; H, 7.41.

Methyl 3-O-methyl-β-lactoside (**4**).—A solution of **16** (0.30 g, 0.33 mmol) in tetrahydrofuran (3 mL) was treated with NaH (65 mg, 2.17 mmol) at 50°. After 30 min, MeI (1.4 mL) was added followed, after 1 h, by water (10 mL). The mixture was extracted with EtOAc (2 × 30 mL), and the combined extracts were dried (Na_2SO_4), treated with solid sodium sulfite, and concentrated to give methyl 2,6,2′,3′,4′,6′-hexa-*O*-benzyl-3-*O*-methyl-β-lactoside (**18**; 0.27 g, 88%) as a syrup. NMR data ($CDCl_3$): ^{1}H, δ 7.12–7.34 (m, 30 H, 6 Ph), 4.86 and 4.51 (ABq, 2 H, *J* 11.7 Hz, C*H*$_2$Ph), 4.77 and 4.63 (ABq, 2 H, *J* 11.2 Hz, C*H*$_2$Ph), 4.75 and 4.65 (ABq, 2 H, *J* 11.1 Hz, C*H*$_2$Ph), 4.63 and 4.58 (ABq, 2 H, *J* 11.2 Hz, C*H*$_2$Ph), 4.42 and 4.32 (ABq, 2 H, *J* 11.5 Hz, C*H*$_2$Ph), 4.38 and 4.29 (ABq, 2 H, *J* 12.3 Hz, C*H*$_2$Ph), 4.38 (d, 1 H, $J_{1',2'}$ 7.7 Hz, H-1′), 4.18 (d, 1 H, $J_{1,2}$ 7.3 Hz, H-1), 3.84 (bd, 1 H, $J_{3',4'}$ 3.29, $J_{4',5'}$ < 0.5 Hz, H-4′), 3.70–3.44 (m, 6 H, H-4,6a,6b,2′,6′a,6′b), 3.50 (s, 3 H, OMe), 3.45 (s, 3 H, OMe), 3.39 (m, 1 H, H-5′), 3.35 (dd, 1 H, $J_{2',3'}$ 9.7, $J_{3',4'}$ 3.0 Hz, H-3′), 3.32 (m, 1 H, H-5), 3.27 (t, 1 H, $J_{2,3} = J_{3,4} = 8.3$ Hz), 3.20 (dd, 1 H, H-2); ^{13}C, δ 138.5, 138.3, 138.0, 137.6 (C_{ipso} Ph), 126.9–127.9 (Ph), 103.8 and 102.6 (C-1,1′), 84.3, 82.1, 81.3, 79.3, 77.1, 74.4, 73.0, and 72.6 (C-2/5 and C-2′/5′), 74.6, 74.2, 74.0, 73.0, 72.5, and 72.0 (*C*H$_2$Ph), 68.1 and 68.0 (C-6,6′), 60.4 and 56.4 (2 OCH_3).

A solution of **18** (0.3 g, 0.33 mmol) in 10:1 EtOH–EtOAc (30 mL) was treated with H_2 in the presence of 10% Pd–C (200 mg) for 20 h to give **4** (0.10 g, 83%), mp 209–211° (from EtOH), $[\alpha]_D^{20}$ +0.5° (*c* 1.0, H_2O). NMR data (D_2O): ^1H, δ 4.46 (d, 1 H, $J_{1',2'}$ 7.6 Hz, H-1'), 4.39 (d, 1 H, $J_{1,2}$ 8.0 Hz, H-1), 4.01 (dd, 1 H, $J_{5,6a}$ 2.3, $J_{6a,6b}$ 12.4 Hz, H-6a), 3.93 (dd, 1 H, $J_{3',4'}$ 4.1, $J_{4',5'}$ 1.0 Hz, H-4'), 3.83 (dd, 1 H, $J_{5,6b}$ 5.27 Hz, H-6b), 3.81 (dd, 1 H, $J_{5',6'a}$ 7.6, $J_{6'a,6'b}$ 11.6 Hz, H-6'a), 3.80 (t, 1 H, $J_{3,4} = J_{4,5} = 9.5$ Hz, H-4), 3.75 (dd, 1 H, $J_{5',6'b}$ 4.5 Hz, H-6'b), 3.66 (ddd, 1 H, H-5'), 3.65 (dd, 1 H, $J_{2',3'}$ 10.6 Hz, H-3'), 3.60 (s, 3 H, OMe), 3.57 (ddd, 1 H, H-5), 3.56 (s, 3 H, OMe), 3.52 (dd, 1 H, H-2'), 3.46 (t, 1 H, $J_{2,3}$ 9.4 Hz, H-3), 3.37 (dd, 1 H, H-2); ^{13}C, δ 103.5 (C-1'), 103.2 (C-1), 84.0 (C-4), 75.8 (C-5'), 75.5 (C-5), 75.5 (C-3), 73.1 (C-3'), 72.5 (C-2), 71.7 (C-2'), 69.0 (C-4'), 61.4 (C-6'), 60.3 (C-6), 59.4 (OCH_3-3), 57.4 (OCH_3-1).

Anal. Calcd for $C_{14}H_{26}O_{11}$: C, 45.41; H, 7.03. Found: C, 45.60; H, 6.92.

Methyl 6-deoxy-β-lactoside (**6**).—A solution of methyl 4',6'-*O*-benzylidene-β-lactoside [16] (**19**; 0.36 g, 0.82 mmol) in 3:1 toluene–acetonitrile (40 mL) was stirred at 80° under Ar with triphenylphosphine (0.32 g, 1.22 mmol), imidazole (0.16 g, 2.35 mmol), and iodine (0.30 g, 1.18 mmol) for 4 h, then cooled to room temperature. Methanol (25 mL) was added and the mixture was concentrated. Column chromatography (12:1 $CHCl_3$–MeOH) of the residue gave methyl 4',6'-*O*-benzylidene-6-deoxy-6-iodo-β-lactoside (**20**; 0.37 g, 81%), isolated as a syrup, $[\alpha]_D^{20}$ −4° (*c* 1.0, H_2O). NMR data (D_2O): ^1H, δ 7.56–7.42 (m, 5 H, Ph), 5.74 (s, 1 H, C*H*Ph), 4.66 (d, 1 H, $J_{1',2'}$ 7.8 Hz, H-1'), 4.47 (d, 1 H, $J_{1,2}$ 8.1 Hz, H-1), 4.37 (bd, 1 H, $J_{3',4'}$ 3.6, $J_{4',5'} < 0.5$ Hz, H-4'), 4.25 (bs, 2 H, H-6'a,6'b), 3.84 (dd, 1 H, $J_{2',3'}$ 10.0, H-3'), 3.80 (bs, 1 H, H-5'), 3.76 (dd, 1 H, $J_{5,6a}$ 2.5, $J_{6a,6b}$ 10.7 Hz, H-6a), 3.70 (bt, 2 H, H-3,2'), 3.58 (s, 3 H, OMe), 3.55 (t, 1 H, $J_{3,4} = J_{4,5} = 9.0$ Hz, H-4), 3.51 (dd, 1 H, $J_{5,6b}$ 5.9 Hz, H-6b), 3.42 (ddd, 1 H, H-5), 3.70 (dd, 1 H, $J_{2,3}$ 9.4 Hz, H-2); ^{13}C, δ 137.9 (C_{ipso}, Ph), 131.0, 129.8, 127.4 (Ph), 104.3, 104.2, 102.3 (C-1,1' and *C*HPh), 83.5, 76.9, 74.9, 74.2, 73.9, 72.3, 71.7, 70.2, 67.8 (C-2/5 and C-2'/6'), 58.7 (OCH_3), 7.0 (C-6).

Anal. Calcd for $C_{20}H_{27}IO_{10}$: C, 43.32; H, 4.87. Found: C, 43.04; H, 5.10.

A solution of **20** (0.35 g, 0.63 mmol) in 5:1 EtOH–EtOAc (30 mL) and triethylamine (0.1 mL) was treated with H_2 in the presence of 10% Pd–C (0.20 g) to give methyl 4',6'-*O*-benzylidene-6-deoxy-β-lactoside (**21**; 0.25 g, 91%), as a syrup, $[\alpha]_D^{20}$ −34° (*c* 0.5, $CHCl_3$). ^1H-NMR data (D_2O): δ 7.51–7.37 (m, 5 H, Ph), 5.68 (s, 1 H, C*H*Ph), 4.53 (d, 1 H, $J_{1',2'}$ 7.7 Hz, H-1'), 4.31 (d, 1 H, $J_{1,2}$ 8.1 Hz, H-1), 4.30 (bd, 1 H, $J_{3',4'}$ 3.7, $J_{4',5'} < 0.5$ Hz, H-4'), 4.19 (bs, 2 H, H-6'a,6'b), 3.76 (dd, 1 H, $J_{2',3'}$ 10.0 Hz, H-3'), 3.71 (bs, 1 H, H-5'), 3.64 (dd, 1 H, H-2'), 3.58 (dd, 1 H, $J_{4,5}$ 9.3, $J_{5,6}$ 6.1 Hz, H-5), 3.54 (t, 1 H, $J_{2,3} = J_{3,4} = 9.3$ Hz, H-3), 3.48 (s, 3 H, OMe), 3.29 (t, 1 H, $J_{3,4} = J_{4,5} = 9.3$ Hz, H-4), 3.23 (dd, 1 H, H-2), 1.32 (d, 3 H, H-6,6,6); ^{13}C, δ 131.0, 129.9, and 127.5 (Ph), 104.4 (*C*HPh), 104.3 and 102.4 (C-1,1'), 85.5, 77.0, 75.4, 74.2, 72.5, 72.2, 71.9, 70.2, and 97.8 (C-2/5 and C-2'/6'), 58.5 (OCH_3), 17.8 (CH_3).

A solution of **21** (0.25 g) was treated with acetic acid (15 mL) at 100°. Water (8

mL) was added slowly and the reaction was continued for 2 h. The solvent was evaporated, and a solution of the residue in water was co-concentrated several times with toluene to leave **6** (0.17 g, 89%). Crystallisation from aq EtOH gave **6**, mp 243–245°, $[\alpha]_D^{20}$ −3.3° (*c* 0.3, H_2O). NMR data (D_2O): ^1H, δ 4.50 (d, 1 H, $J_{1',2'}$ 7.7 Hz, H-1'), 4.38 (d, 1 H, $J_{1,2}$ 8.0, H-1), 3.93 (bd, 1 H, $J_{3',4'}$ 3.3, $J_{4',5'} < 0.5$ Hz, H-4'), 3.80 (dd, 1 H, $J_{5',6'a}$ 8.3, $J_{6'a,6'b}$ 11.5 Hz, H-6'), 3.74 (dd, 1 H, $J_{5',6'b}$ 2.8 Hz, H-6'b), 3.71 (bdd, 1 H, H-5'), 3.66 (dd, 1 H, $J_{2',3'}$ 9.9 Hz, H-3'), 3.63 (dd, 1 H, $J_{4,5}$ 9.4, $J_{5,6}$ 6.5 Hz, H-5), 3.57 (t, 1 H, $J_{2,3} = J_{3,4} = 9.2$ Hz, H-3), 3.55 (s, 3 H, OMe), 3.54 (dd, 1 H, H-2), 3.39 (t, 1 H, H-4), 3.30 (dd, 1 H, H-2), 1.37 (d, 3 H, H-6,6,6); ^{13}C, δ 102.8 (C-1'), 102.6 (C-1), 83.4 (C-4), 74.9 (C-5'), 73.9 (C-3), 72.6 (C-2), 72.2 (C-3'), 70.6 (C-2'), 70.4 (C-5), 68.2 (C-4'), 60.6 (C-6'), 56.7 (OCH_3), 16.2 (C-6).

Anal. Calcd for $C_{13}H_{24}O_{10}$: C, 45.88; H, 7.06. Found: C, 46.03; H, 7.17.

Methyl 4-O-β-D-galactopyranosyl-β-D-xylopyranoside (**5**).—A solution of methyl 2,3-di-*O*-acetyl-β-D-xylopyranoside [18] (**22**; 0.11 g, 0.44 mmol) in acetonitrile (1 mL) was stirred at room temperature for 10 min with mercuric cyanide (0.10 g) and mercuric bromide (15 mg) under Ar in the dark. A solution of 2,3,4,6-tetra-*O*-acetyl-α-D-galactopyranosyl bromide (**23**; 0.18 g, 0.45 mmol) in acetonitrile (0.25 mL) was added and the reaction continued under the same conditions for 90 min. More **23** (0.18 g, 0.45 mmol) in acetonitrile (0.25 mL) was added and the reaction was continued overnight. The mixture was diluted with $CHCl_3$ (20 mL), washed with aq 10% KI and water, the aqueous phases were extracted with $CHCl_3$, and the combined organic phases were concentrated. The residue was dissolved in benzene and treated conventionally with acetic anhydride–pyridine, and the solution was concentrated. Column chromatography (2.5 : 1 → 1.5 : 1 hexane–EtOAc) of the residue gave 1,2,3,4,6-penta-*O*-acetyl-α,β-D-galactopyranose, then methyl 2,3-di-*O*-acetyl-4-*O*-(2,3,4,6-tetra-*O*-acetyl-α-D-galactopyranosyl)-β-D-xylopyranoside (70 mg, 28%), and finally methyl 2,3,2',3',4',6'-hexa-*O*-acetyl-6-deoxy-β-lactoside (**24**; 0.145 g, 58%), isolated as a syrup, $[\alpha]_D^{20}$ +26° (*c* 1.1, $CHCl_3$). ^1H-NMR data ($CDCl_3$): δ 5.38 (dd, 1 H, $J_{3',4'}$ 3.4, $J_{4',5'}$ 1.1 Hz, H-4'), 5.15 (t, 1 H, $J_{2,3} = J_{3,4} = 8.4$ Hz, H-3), 5.14 (dd, 1 H, $J_{1',2'}$ 7.8, $J_{2',3'}$ 10.4 Hz, H-2'), 5.01 (dd, 1 H, H-3'), 4.85 (dd, 1 H, $J_{1,2}$ 6.8, $J_{2,3}$ 8.6 Hz, H-2), 4.53 (d, 1 H, H-1'), 4.38 (d, 1 H, H-1), 4.13 (m, 2 H, H-6'a,6'b), 4.00 (dd, 1 H, $J_{4,5eq}$ 5.1, $J_{5eq,5ax}$ 11.8 Hz, H-5*eq*), 3.93 (td, 1 H, $J_{5',6'a} = J_{5',6'b} = 6.6$ Hz, H-5'), 3.84 (ddd, 1 H, $J_{4,5ax}$ 9.0, H-4), 3.49 (s, 3 H, OMe), 3.36 (dd, 1 H, H-5*ax*), 2.18, 2.08, 2.08, 2.07, 2.07, 2.00 (6 s, each 3 H, 6 Ac).

A solution of **24** (0.145 g, 0.26 mmol) in MeOH (5 mL) was treated with methanolic 0.1 M sodium methoxide for 2 h. The mixture was neutralised with Amberlite IR-120 (H^+) resin, filtered, and concentrated to give **5** (74 mg, 95%), mp 248–250° (from aq MeOH), $[\alpha]_D^{20}$ −8.7° (*c* 1.0, H_2O). NMR data: ^1H ($CDCl_3$), δ 4.46 (d, 1 H, $J_{1',2'}$ 7.7 Hz, H-1'), 4.34 (d, 1 H, $J_{1,2}$ 7.8 Hz, H-1), 4.10 (dd, 1 H, $J_{4,5eq}$ 5.3, $J_{5eq,5ax}$ 11.6 Hz, H-5*eq*), 3.91 (dd, 1 H, $J_{3',4'}$ 3.5, $J_{4',5'}$ 0.8 Hz, H-4'), 3.83 (ddd, 1 H, $J_{3,4}$ 9.0, $J_{4,5ax}$ 10.4 Hz, H-4), 3.66 (dd, 1 H, $J_{2',3'}$ 10.0 Hz, H-3'), 3.65–3.58 (m, 3 H, H-5',6'a,6'b), 3.62 (dd, 1 H, $J_{2',3'}$ 9.5 Hz, H-3), 3.50 (dd, 1 H,

H-2′), 3.54 (s, 3 H, OMe), 3.40 (dd, 1 H, H-5*ax*), 3.29 (dd, 1 H, H-2); ^{13}C (D_2O), δ 105.1 (C-1′), 103.0 (C-1), 77.9 (C-4), 76.6 (C-5′), 75.2 (C-3), 74.0 (C-2), 73.9 (C-3′), 71.9 (C-2′), 69.9 (C-4′), 64.2 (C-5), 62.4 (C-6′), 58.4 (OCH_3).

Anal. Calcd for $C_{12}H_{22}O_{10}$: C, 50.35; H, 7.69. Found: C, 50.51; H, 7.78.

*Methyl 2′-deoxy-β-lactoside [methyl 4-O-(2-deoxy-β-*D*-lyxo-hexopyranosyl-β-*D*-glucopyranoside, 7].*—A mixture of **11** (1 g, 2.53 mmol), tributyltin oxide (3.25 mL, 6.40 mmol), powdered 3A molecular sieves (7.5 g), and toluene (70 mL) was stirred at 120° under Ar overnight. The mixture was cooled to 95°, *N*-methylimidazole (0.51 mL, 6.40 mmol) and benzyl bromide (10 mL) were added, and the reaction was continued under the same conditions for 4 days. The mixture was filtered through Celite and washed with water, the aqueous fraction was extracted with ether, and the combined organic phases were concentrated. Column chromatography (2:1 hexane–EtOAc) of the residue gave methyl 2,6,6′-tri-*O*-benzyl-3′,4′-*O*-isopropylidene-β-lactoside (**25**; 0.83 g, 49%), isolated as a syrup. NMR data ($CDCl_3$): ^{1}H, δ 7.42–7.21 (m, 15 H, 3 Ph), 4.84 (s, 2 H, CH_2Ph), 4.65 and 4.53 (ABq, 2 H, *J* 12.0 Hz, CH_2Ph), 4.63 and 4.55 (ABq, 2 H, *J* 12.2 Hz, CH_2Ph), 4.48 (d, 1 H, $J_{3,HO-3}$ 1.6 Hz, HO-3), 4.30 (d, 1 H, $J_{1,2}$ 7.8 Hz, H-1), 4.21 (d, 1 H, $J_{1',2'}$ 8.3 Hz, H-1′), 4.09 (dd, 1 H, $J_{3',4'}$ 5.5, $J_{4',5'}$ 2.1 Hz, H-4′), 4.04–3.99 (m, 2 H, H-3′,5′), 3.79 (dd, 1 H, $J_{5',6'a}$ 7.8, $J_{6'a,6'b}$ 10.0 Hz, H-6′a), 3.80–3.72 (m, 3 H, H-3,6a,6b), 3.71 (dd, 1 H, $J_{5',6'b}$ 4.2 Hz, H-6′b), 3.55–3.49 (m, 3 H, H-4,5,2′), 3.53 (s, 3 H, OMe), 3.31 (dd, 1 H, $J_{2,3}$ 9.0 Hz, H-2), 3.06 (d, 1 H, $J_{2,HO-2}$ 2.4 Hz, HO-2), 1.50 and 1.32 (2 s, each 3 H, CMe_2); ^{13}C, δ 138.9, 137.7, 137.6 (C_{ipso}, Ph), 128.9–126.0 (Ph), 110.4 (*C*Me_2), 104.2, 103.4 (C-1,1′), 83.7, 81.3, 79.0, 77.2, 75.5, 73.5, 72.8, 72.7 (C-2/5 and C-2′/5′), 74.8, 73.7, 73.6 (3 *C*H_2Ph), 70.1, 69.2 (C-6,6′), 28.1 and 6.3 [(*C*H_3)$_2$C].

Anal. Calcd for $C_{36}H_{46}O_{11}$: C, 66.67; H, 6.91. Found: C, 66.88; H, 7.05.

A solution of **25** (1.3 g, 1.95 mmol) and 4-dimethylaminopyridine (0.54 g, 4.43 mmol) in toluene (130 mL) at 100° under Ar was treated with phenoxythiocarbonyl chloride (330 μL, 2.4 mmol) for 72 h, then concentrated. Column chromatography (2:1 hexane–EtOAc) of the residue gave methyl 2,6,6′-tri-*O*-benzyl-3′,4′-*O*-isopropylidene-2′-phenoxythiocarbonyl-β-lactoside (**26**; 0.96 g, 62%), isolated as a syrup. ^{1}H-NMR data ($CDCl_3$): δ 7.42–7.21 and 7.12–7.08 (2 m, 18 and 2 H, 4 Ph), 5.59 (t, 1 H, H-2′), 4.85 (s, 2 H, CH_2Ph), 4.65 and 4.59 (ABq, 2 H, *J* 11.9 Hz, CH_2Ph), 4.65 and 4.55 (ABq, 2 H, *J* 12.0 Hz, CH_2Ph), 4.49 (d, 1 H, $J_{1',2'}$ 7.9 Hz, H-1′), 4.35 (d, 1 H, $J_{3,HO-3}$ 1.8 Hz, HO-3), 4.30 (d, 1 H, $J_{1,2}$ 7.9 Hz, H-1), 4.27 (dd, 1 H, $J_{2',3'}$ 7.2, $J_{3',4'}$ 5.5 Hz, H-3′), 4.17 (dd, 1 H, $J_{4',5'}$ 2.1 Hz, H-4′), 4.02 (ddd, 1 H, $J_{5',6'a}$ 4.4, $J_{5',6'b}$ 7.7 Hz, H-5′), 3.88 (dd, 1 H, $J_{5,6a}$ 1.7, $J_{6a,6b}$ 10.5 Hz, H-6a), 3.84–3.77 (m, 3 H, H-3, 6′a,b), 3.73 (dd, 1 H, $J_{5,6b}$ 4.4 Hz, H-6b), 3.63 (dd, 1 H, $J_{3,4}$ 8.4, $J_{4,5}$ 9.7 Hz, H-4), 3.55 (s, 3 H, OMe), 3.51 (ddd, 1 H, H-5), 3.35 (dd, 1 H, $J_{2,3}$ 8.9 Hz, H-2), 1.58 and 1.34 (2 s, each 3 H, CMe_2).

Anal. Calcd for $C_{44}H_{50}O_{12}S$: C, 65.84; H, 6.23; S, 3.99. Found: C, 66.01; H, 6.02; S, 4.21.

A mixture of **26** (63 mg, 0.08 mmol), α,α′-azobisisobutyronitrile (2 mg), and

toluene (20 mL) was boiled under reflux under Ar. A solution of tributyltin hydride (30 μL, 0.10 mmol) in toluene (3 mL) was added dropwise, the reaction was continued for 30 h under the same conditions, and the solvent was then evaporated. Column chromatography (2:1 hexane–EtOAc) of the residue gave methyl 2,6-di-*O*-benzyl-4-*O*-(6-*O*-benzyl-2-deoxy-3,4-*O*-isopropylidene-β-D-*lyxo*-hexopyranosyl)-β-D-glucopyranoside (**27**; 43 mg, 84%), isolated as a syrup, $[\alpha]_D^{20}$ +0.1° (*c* 1.3, $CHCl_3$). ^{1}H-NMR data ($CDCl_3$): δ 7.42–7.21 (m, 15 H, 3 Ph), 4.88 and 4.83 (ABq, 2 H, *J* 11.4 Hz, CH_2Ph), 4.70 and 4.44 (ABq, 2 H, *J* 12.2 Hz, CH_2Ph), 4.65 (d, 1 H, $J_{3,HO-3}$ 1.2 Hz, HO-3), 4.64 and 4.53 (ABq, 2 H, *J* 12.0 Hz, CH_2Ph), 4.30 (dd, 1 H, $J_{1',2'eq}$ 2.4, $J_{1',2'ax}$ 8.9 Hz, H-1′), 4.29 (d, 1 H, $J_{1,2}$ 7.8 Hz, H-1), 4.14 (ddd, 1 H, $J_{2'eq,3'}$ 6.7, $J_{2'ax,3'}$ 9.1, $J_{3',4'}$ 5.4 Hz, H-3′), 3.94 (dd, 1 H, $J_{4',5'}$ 2.0 Hz, H-4′), 3.88 (ddd, 1 H, $J_{5',6'a}$ 7.1, $J_{5',6'b}$ 4.8 Hz, H-5′), 3.78 (dd, 1 H, $J_{6'a,6'b}$ 9.9 Hz, H-6′a), 3.74 (dt, 1 H, $J_{2,3} = J_{3,4} = 8.7$ Hz, H-3), 3.72 (dd, 1 H, H-6′b), 3.67 (d, 2 H, H-6a,6b), 3.59 (dd, 1 H, $J_{3,4}$ 8.4, $J_{4,5}$ 9.8 Hz, H-4), 3.54 (s, 3 H, OMe), 3.42 (dt, 1 H, $J_{5,6a} = J_{5,6b} = 2.9$ Hz, H-5), 3.34 (dd, 1 H, $J_{2,3}$ 8.8 Hz, H-2), 1.76 (ddd, 1 H, $J_{2'eq,2'ax}$ 13.2 Hz, H-2′*eq*), 1.58 (dt, 1 H, $J_{1',2'ax} = J_{2'ax,3'} = 9.0$ Hz, H-2′*ax*), 1.47 and 1.29 (2 s, each 3 H, CMe_2).

Anal. Calcd for $C_{37}H_{46}O_{10}$: C, 68.31; H, 7.08. Found: C, 68.60; H, 6.89.

A solution of **27** (0.20 g, 0.31 mmol) in 2:1 CH_2Cl_2–EtOH (10 mL) was treated overnight with H_2 in the presence of 10% Pd–C (60 mg) to give a syrupy product, a solution of which in aq 50% acetic acid (10 mL) was kept at 65° for 1 h, then concentrated to give **7** (75 mg, 72%), mp 222–225° (from aq EtOH), $[\alpha]_D^{20}$ −7.2° (*c* 1.0, H_2O). NMR data (D_2O): ^{1}H, δ 4.69 (dd, 1 H, $J_{1',2'eq}$ 2.3, $J_{1',2'ax}$ 9.9 Hz, H-1′), 4.39 (d, 1 H, $J_{1,2}$ 7.9 Hz, H-1), 3.90 (dd, 1 H, $J_{5,6a}$ 2.3, $J_{6a,6b}$ 12.3 Hz, H-6a), 3.88 (ddd, 1 H, $J_{2'ax,3'}$ 12.1, $J_{3',4'}$ 3.2 Hz, H-3′), 3.80 (dd, 1 H, $J_{5',6'a}$ 7.9, $J_{6'a,6'b}$ 11.9 Hz, H-6′a), 3.77 (bd, 1 H, H-4′), 3.74 (dd, 1 H, $J_{5',6'b}$ 4.8 Hz, H-6′b), 3.73 (dd, 1 H, $J_{5,6b}$ 5.2 Hz, H-6b), 3.67 (t, 1 H, $J_{3,4} = J_{4,5} = 8.9$ Hz, H-4), 3.61 (t, 1 H, $J_{2,3} = J_{3,4} = 8.7$ Hz, H-3), 3.60 (ddd, 1 H, $J_{4',5'}$ 0.8 Hz, H-5′), 3.56 (s, 3 H, OMe), 3.53 (ddd, 1 H, H-5), 3.29 (dd, 1 H, H-2), 2.07 (ddd, 1 H, $J_{2'eq,2'ax}$ 12.3 Hz, H-2′*eq*), 1.69 (dt, 1 H, H-2′*ax*); ^{13}C, δ 104.3 (C-1), 101.5 (C-1′), 79.5 (C-4), 76.8 (C-5′), 75.8 (C-5), 75.5 (C-3), 74.0 (C-2), 68.8 (C-3′), 67.8 (C-4′), 62.6 (C-6′), 61.4 (C-6), 58.4 (OCH_3), 34.6 (C-2′).

Anal. Calcd for $C_{13}H_{24}O_{10}$: C, 45.88; H, 7.06. Found: C, 45.90; H, 7.06.

Methyl 3′-deoxy-β-lactoside [methyl 4-O-(3-deoxy-β-D-xylo-hexopyranosyl)-β-D-glucopyranoside, **8***].—(a) From* **1**. A mixture of **1** (1 g, 2.81 mmol), dibutyltin oxide (0.75 g, 3.01 mmol), 3A molecular sieves, and acetonitrile (50 mL) was boiled under reflux overnight under Ar. Tetrabutylammonium bromide (0.40 g) and allyl bromide (7 mL) were added and the reaction was continued under the same conditions for 48 h. The mixture was cooled, filtered through Celite, and concentrated. Column chromatography (5:1 $CHCl_3$–MeOH) of the residue gave methyl 3′-*O*-allyl-β-lactoside (**29**; 0.87 g, 78%), isolated as a syrup, $[\alpha]_D^{20}$ +3.2° (*c* 1.0, MeOH). ^{1}H-NMR data ($CDCl_3$): δ 5.75 (ddt, 1 H, J_{cis} 10.5, J_{trans} 17.1, $J_{H,CHa} = J_{H,CHb} = 6.6$ Hz, CH_2=C*H*CH_2), 5.15 (ddt, 1 H, $J_{gem} = J_{allyl\text{-}cis} = J_{allyl\text{-}trans} = 1.5$,

J_{trans} 17.1 Hz, CH_2=CHCH$_2$), 5.06 (ddt, 1 H, $J_{allyl\text{-}cis} = J_{allyl\text{-}trans} = 1.5$, $J_{cis} = 10.2$ Hz, CH_2=CHCH$_2$), 4.24 (d, 1 H, $J_{1',2'}$ 7.3 Hz, H-1'), 4.18 (d, 1 H, $J_{1,2}$ 8.1 Hz, H-1), 4.01 (ddt, 1 H, J_{gem} 12.5 Hz, CH$_2$=CHC*H*a), 3.93 (dd, 1 H, $J_{3',4'}$ 2.8, $J_{4',5'} < 0.5$ Hz, H-4'), 3.88 (ddt, 1 H, CH$_2$=CHC*H*b), 3.77 (dd, 1 H, $J_{5,6a}$ 2.0, $J_{6a,6b}$ 12.2 Hz, H-6a), 3.59 (dd, 1 H, $J_{5,6b}$ 5.2 Hz, H-6b), 3.35 (s, 3 H, OMe), 3.30 (dd, 1 H, $J_{2',3'}$ 9.9 Hz, H-3'), 3.08 (m, 1 H, H-2).

Anal. Calcd for $C_{16}H_{28}O_{11}$: C, 48.48; N, 7.07. Found: C, 48.65; H, 6.89.

Conventional acetylation of **29** gave the 2,3,6,2',4',6'-hexa-acetate **41**. ^{1}H-NMR data ($CDCl_3$): δ 5.85–5.66 (dddd, 1 H, J_{trans} 17.2, J_{cis} 10.3, $J_{H,CHa}$ 5.0, $J_{H,CHb}$ 6.0 Hz, CH$_2$=C*H*CH$_2$), 5.38 (dd, 1 H, $J_{3',4'}$ 3.6, $J_{4',5'}$ 1.2 Hz, H-4'), 5.27–5.13 (m, 2 H, CH_2=CH), 5.19 (dd, 1 H, $J_{2,3}$ 9.5, $J_{3,4}$ 8.7 Hz, H-3), 4.99 (dd, 1 H, $J_{1',2'}$ 8.0, $J_{2',3'}$ 10.1 Hz, H-2'), 4.88 (dd, 1 H, $J_{1,2}$ 7.8 Hz, H-2), 4.48 (dd, 1 H, $J_{5,6a}$ 2.1, $J_{6a,6b}$ 11.9 Hz, H-6a), 4.41 (d, 1 H, H-1'), 4.40 (d, 1 H, H-1), 4.17–4.06 (m, 3 H, H-6'a,6'b and =CHC*H*a), 4.15 (dd, 1 H, $J_{5,6b}$ 5.12 Hz, H-6b), 3.86 (ddt, 1 H, J_{gem} 13.0, $J_{H,CH_2=CH}$ 6.0, $J_{allyl\text{-}cis} = J_{allyl\text{-}trans} = 1.3$ Hz, CH$_2$=CHC*H*b), 3.78 (m, 1 H, H-5'), 3.75 (dd, 1 H, $J_{4,5}$ 9.9 Hz, H-4), 3.62 (ddd, 1 H, H-5), 3.48 (s, 3 H, OMe), 3.46 (dd, 1 H, $J_{2',3'}$ 10.1, $J_{3',4'}$ 3.5 Hz, H-3'), 2.14 (s, 3 H, OMe), 2.12, 2.09, 2.07, 2.05, and 2.04 (6 s, each 3 H, 6 OMe).

A solution of **29** (0.75 g, 1.9 mmol) in *N*,*N*-dimethylformamide (30 mL) was stirred with NaH (1.0 g) and benzyl bromide (5 mL) overnight. Methanol (15 mL) and then water (20 mL) were added, the mixture was extracted with $CHCl_3$, and the extract was dried (Na_2SO_4) and concentrated. Column chromatography (hexane → 5 : 1 hexane–EtOAc) of the residue gave methyl 3'-*O*-allyl-2,3,6,2',4',6'-hexa-*O*-benzyl-β-lactoside (**30**; 1.45 g, 82%), isolated as a syrup. NMR data ($CDCl_3$): ^{1}H, δ 7.38–7.12 (m, 30 H, 6 Ph), 5.92 (ddt, 1 H, J_{trans} 17.2, J_{cis} 10.4, $J_{H,CHa} = J_{H,CHb} = 5.4$ Hz, CH$_2$=C*H*CH$_2$), 5.31 (ddt, 1 H, $J_{gem} = J_{allyl\text{-}cis} = J_{allyl\text{-}trans} = 1.5$, J_{trans} 17.2 Hz, CH_2=CH), 5.17 (ddt, 1 H, $J_{gem} = J_{allyl\text{-}cis} = J_{allyl\text{-}trans} = 1.3$, J_{cis} 10.4 Hz, CH_2=CH), 5.02 and 4.71 (ABq, 2 H, J 10.9 Hz, CH_2Ph), 4.95 and 4.53 (ABq, 2 H, J 11.4 Hz, CH_2Ph), 4.86 and 4.72 (ABq, 2 H, J 11.0 Hz, CH_2Ph), 4.80 and 4.73 (ABq, 2 H, J 11.0 Hz, CH_2Ph), 4.53 and 4.40 (ABq, 2 H, J 12.3 Hz, CH_2Ph), 4.43 (d, 1 H, $J_{1',2'}$ 7.3 Hz, H-1'), 4.33 and 4.23 (ABq, 2 H, J 12.3 Hz, CH_2Ph), 4.29 (d, 1 H, $J_{1,2}$ 7.6 Hz, H-1), 4.15 (bd, 2 H, CH$_2$=CHCH_2), 3.92 (t, 1 H, $J_{3,4} = J_{4,5} = 9.3$ Hz, H-4), 3.85 (bd, 1 H, $J_{3',4'}$ 2.6, $J_{4',5'}$ 0.5 Hz, H-4'), 3.80 (dd, 1 H, $J_{5,6a}$ 4.3, $J_{6a,6b}$ 10.8 Hz, H-6a), 3.74 (dd, 1 H, $J_{5,6b}$ 1.5 Hz, H-6b), 3.69 (dd, 1 H, $J_{2',3'}$ 9.8 Hz, H-2'), 3.58–3.51 (m, 2 H, H-3,5'), 3.55 (s, 3 H, OMe), 3.40–3.31 (m, 3 H, H-5,6'a,6'b), 3.38 (dd, 1 H, $J_{2,3}$ 8.8 Hz, H-2), 3.30 (dd, 1 H, H-3'); ^{13}C, δ 139.1, 139.0, 138.8, 138.7, 138.3, 138.0 (C_{ipso}, Ph), 134.9 (CH$_2$=*C*H), 128.5–126.9 (Ph), 116.4 (*C*H$_2$=CH), 104 and 102.7 (C-1,1'), 82.8, 82.3, 81.7, 79.8, 76.7, 75.3, 75.2, 75.1, 74.8, 74.5, 73.3, 73.0, 72.8, 71.4, 68.3, 68.0 (6 *C*H$_2$–Ph, C-2/6, and C-2'/6'), 65.1 (=CH*C*H$_2$), 60.0 (OCH$_3$).

A solution of **30** (0.85 g, 0.91 mmol) in Me_2SO (10 mL) was stirred with potassium *tert*-butoxide (0.30 g, 2.6 mmol) at 95° for 2 h, then cooled, poured into water (150 mL), and extracted with ether. The extract was dried ($MgSO_4$) and

concentrated. A solution of the residue in 10:1 water–acetone (40 mL) was treated with yellow mercuric oxide (0.30 g) and, after 30 min, with a solution of mercuric chloride (0.30 g) in 10:1 acetone–water (10 mL). After 40 min, the solution was filtered and concentrated, and the residue was suspended in water and extracted twice with ether. The combined extracts were washed with aq 10% KI, dried ($MgSO_4$), and concentrated. Column chromatography (2:1 hexane–EtOAc) of the residue gave methyl 2,3,6,2′,4′,6′-hexa-*O*-benzyl-β-lactoside (**31**; 0.59 g, 69%), isolated as a syrup, $[\alpha]_D^{20}$ +6.7° (*c* 1.0, $CHCl_3$). A solution of **31** in tetrahydrofuran (25 mL) was treated with NaH (50 mg) under Ar. After 30 min, CS_2 (0.3 mL) was added, followed by MeI (1 mL), α,α′-azobisisobutyronitrile (10 mg), and tributyltin hydride (0.2 mL, 0.58 mmol), as described for the conversion **14** → **15** to give methyl 2,3,6-tri-*O*-benzyl-4-*O*-(2,4,6-tri-*O*-benzyl-3-deoxy-β-D-*xylo*-hexopyranosyl)-β-D-glucopyranoside (**32**; 0.38 g, 77%), isolated as a syrup. A solution of **32** in 5:1 EtOH–EtOAc (10 mL) was hydrogenated in the presence of 10% Pd–C (60 mg) overnight at room temperature to give **8** (67 mg, 82%), isolated as a syrup, $[\alpha]_D^{20}$ −23° (*c* 1.0, H_2O). NMR data (D_2O): ^1H, δ 4.42 (d, 1 H, $J_{1',2'}$ 7.9 Hz, H-1′), 4.39 (d, 1 H, $J_{1,2}$ 8.0 Hz, H-1), 3.97 (bd, 1 H, $J_{4',5'} < 0.8$ Hz, H-4′), 3.96 (dd, 1 H, $J_{5,6a}$ 1.6, $J_{6a,6b}$ 12.4 Hz, H-6a), 3.78 (dd, 1 H, $J_{5,6b}$ 4.7 Hz, H-6b), 3.76–3.68 (m, 4 H, H-2′,5′,6′a,6′b), 3.65–3.53 (m, 3 H, H-3,4,5), 3.55 (s, 3 H, OMe), 3.29 (m, 1 H, H-2), 2.20 (ddd, 1 H, $J_{2',3'eq}$ 5.2, $J_{3'eq,4'}$ 3.1, $J_{3'ax,3'eq}$ 13.9 Hz, H-3′*eq*), 1.72 (ddd, 1 H, $J_{2',3'ax}$ 12.1, $J_{3'ax,4'}$ 2.9 Hz, H-3′*ax*); ^{13}C, δ 106.1 (C-1′), 104.3 (C-1), 79.8 and 79.6 (C-4,5′), 76.6 (C-5), 75.6 (C-3), 74.0 (C-2), 66.8 (C-2′), 66.7 (C-4′), 62.4 (C-6′), 61.3 (C-6), 58.5 (OCH_3), 38.2 (C-3′).

Anal. Calcd for $C_{13}H_{24}O_{10}$: C, 45.88; H, 7.06. Found: C, 45.63; H, 7.21.

(b) From **19**.—A mixture of **19** (0.10 g, 0.23 mmol), dibutyltin oxide (72 mg, 0.29 mmol), and MeOH (4 mL) was boiled under Ar until dissolution was complete (2 h). The solvent was then evaporated, and to a solution of the residue in 1,4-dioxane (4 mL) was added phenoxythiocarbonyl chloride (43.2 mg, 0.25 mmol). The mixture was stirred at room temperature for 40 h then concentrated. Preparative TLC (4:1 $CHCl_3$–MeOH) of the residue gave methyl 3′-*O*-phenoxythiocarbonyl-β-lactoside (**34**; 107 mg, 82%), isolated as a syrup. A solution of **34** (50 mg, 0.1 mmol) in toluene (20 mL) was boiled under reflux under Ar. A solution of tributyltin hydride (0.03 mL, 0.1 mM) in toluene 2 mL) was added, and the mixture was stirred for 10 h, then concentrated. A solution of the residue in acetic acid (5 mL) was heated to 100°, water (2 mL) was added slowly, and the reaction was continued for 2 h at 100°. The mixture was cooled, then concentrated, and a solution of the residue in water was co-concentrated several times with toluene to give syrupy **8** (20 mg, 68%).

*Methyl 4′-deoxy-β-lactoside (methyl 4-*O*-(4-deoxy-β-*D*-xylo-hexopyranosyl)-β-D-glucopyranoside,* **9**).—A stirred solution **19** (0.44 g, 1 mmol) in *N*,*N*-dimethylformamide (30 mL) was treated with NaH (0.60 g) and then benzyl bromide (10 mL) overnight. Methanol (15 mL) and then water (20 mL) were added, the mixture was extracted with $CHCl_3$, and the combined extracts were dried (Na_2SO_4) and

concentrated. Column chromatography (hexane → 5:1 hexane–EtOAc) of the residue gave methyl 2,3,6,2′,3′-penta-*O*-benzyl-4′,6′-*O*-benzylidene-β-lactoside (**35**; 0.78 g, 88%), $[\alpha]_D^{20}$ +17.5° (*c* 1.1, $CHCl_3$). ^{1}H-NMR data ($CDCl_3$): δ 7.29–7.11 (m, 30 H, 6 Ph), 5.40 (s, 1 H, C*H*Ph), 5.13 and 4.72 (ABq, 2 H, *J* 10.7 Hz, CH_2Ph), 4.83 and 4.70 (ABq, 2 H, *J* 11.1 Hz, CH_2Ph), 4.79 and 4.68 (ABq, 2 H, *J* 11.1 Hz, CH_2Ph), 4.67 (s, 2 H, CH_2Ph), 4.51 and 4.29 (ABq, 2 H, *J* 12.1 Hz, CH_2Ph), 4.41 (d, 1 H, $J_{1',2'}$ 7.8 Hz, H-1′), 4.25 (d, 1 H, $J_{1,2}$ 7.8 Hz, H-1), 4.16 (dd, 1 H, $J_{5',6'a}$ 1.3, $J_{6'a,6'b}$ 12.2 Hz, H-6′a), 3.97 (bd, 1 H, $J_{3',4'}$ 3.6, $J_{4',5'} < 0.5$ Hz, H-4′), 3.93 (dd, 1 H, $J_{3,4}$ 9.0, $J_{4,5}$ 9.7 Hz, H-4), 3.84 (dd, 1 H, $J_{5,6a}$ 4.1, $J_{6a,6b}$ 10.9 Hz, H-6a), 3.78 (dd, 1 H, $J_{5',6'b}$ 1.8 Hz, H-6′b), 3.71 (dd, 1 H, $J_{2',3'}$ 9.6 Hz, H-2′), 3.67 (dd, 1 H, $J_{5,6b}$ 1.7 Hz, H-6b), 3.57 (t, 1 H, $J_{2,3} = J_{3,4} = 9.0$ Hz, H-3), 3.51 (s, 3 H, OMe), 3.37 (dd, 1 H, H-2), 3.33 (dd, 1 H, H-3′), 3.32 (ddd, 1 H, H-5), 2.88 (bs, 1 H, H-5′).

Anal. Calcd for $C_{55}H_{58}O_{11}$: C, 73.83; H, 6.89. Found: C, 74.89; H, 7.08.

A solution of **35** (1.5 g, 1.68 mmol) and sodium cyanoborohydride (1.5 g) in tetrahydrofuran (100 mL) was stirred with 3A molecular sieves (1.5 g). A saturated solution of HCl in ether was added dropwise until the evolution of gas ceased and stirring was then continued for 30 min. Dichloromethane (100 mL) was added, and the mixture was filtered, washed with water and aq $NaHCO_3$, dried (Na_2SO_4), and concentrated. Column chromatography (2:1 hexane–EtOAc) of the residue gave methyl 2,3,6,2′,3′,6′-hexa-*O*-benzyl-β-lactoside (**36**; 1.28 g, 86%), isolated as a syrup, $[\alpha]_D^{20}$ +20° (*c* 1.0, $CHCl_3$). NMR data ($CDCl_3$): ^{1}H, δ 7.41–7.20 (m, 30 H, 6 Ph), 4.98 and 4.76 (ABq, 2 H, *J* 10.9 Hz, CH_2Ph), 4.86 and 4.71 (ABq, 2 H, *J* 11.1 Hz, CH_2Ph), 4.77 (s, 2 H, CH_2Ph), 4.72 and 4.66 (ABq, 2 H, *J* 11.5 Hz, CH_2Ph), 4.57 and 4.40 (ABq, 2 H, *J* 12.1 Hz, CH_2Ph), 4.46 and 4.39 (ABq, 2 H, *J* 12.0 Hz, CH_2Ph), 4.43 (d, 1 H, $J_{1',2'}$ 7.8 Hz, H-1′), 4.29 (d, 1 H, $J_{1,2}$ 7.7 Hz, H-1), 4.01 (m, 1 H, H-4′), 3.97 (t, 1 H, $J_{2,3} = J_{3,4} = 7.6$ Hz, H-3), 3.82 (dd, 1 H, $J_{5,6a}$ 4.2, $J_{6a,6b}$ 10.9 Hz, H-6a), 3.73 (dd, 1 H, $J_{5,6b}$ 1.9 Hz, H-6b), 3.66 (dd, 1 H, $J_{5',6'a}$ 7.1, $J_{6'a,6'b}$ 9.7 Hz, H-6′a), 3.59 (dd, 1 H, $J_{2',3'}$ 9.4 Hz, H-2′), 3.57 (t, 1 H, $J_{3,4} = J_{4,5} = 9.0$ Hz, H-4), 3.56 (s, 3 H, OMe), 3.48 (dd, 1 H, $J_{5',6'b}$ 5.3 Hz, H-6′b), 3.39 (dd, 1 H, H-2), 3.39 (ddd, 1 H, H-5), 3.37 (dd, 1 H, $J_{3',4'}$ 3.4 Hz, H-3′), 3.32 (bt, 1 H, H-5′); ^{13}C, δ 138.6, 138.2, 138.1, 137.7, 137.7, 137.4 (C_{ipso}, Ph), 127.8–126.6 (Ph), 104.1, 101.9 (C-1,1′), 82.2, 81.2, 80.5, 78.8, 75.9, 74.5, 72.3, 65.5, (C-2/5 and C-2′/5′), 74.7, 74.6, 74.2, 72.9, 72.5, 71.3, 67.9, 67.6 (6 CH_2Ph and C-6,6′), 56.3 (OCH_3).

Anal. Calcd for: $C_{55}H_{60}O_{11}$: C, 73.66; H, 6.70. Found: C, 73.20; H, 6.60.

Conventional acetylation of **36** gave the 4′-acetate **42**. ^{1}H-NMR data ($CDCl_3$): δ 7.28–7.15 (m, 30 H, 6 Ph), 5.54 (dd, 1 H, $J_{3',4'}$ 2.7, $J_{4',5'}$ 0.5 Hz, H-4′), 4.97 and 4.74 (ABq, 2 H, *J* 10.6 Hz, CH_2Ph), 4.86 and 4.70 (ABq, 2 H, *J* 11.1 Hz, CH_2Ph), 4.78 and 4.70 (ABq, 2 H, *J* 11.3 Hz, CH_2Ph), 4.76 and 4.47 (ABq, 2 H, *J* 11.3 Hz, CH_2Ph), 4.55 and 4.36 (ABq, 2 H, *J* 12.2 Hz, CH_2Ph), 4.47 and 4.26 (ABq, 2 H, *J* 12.0 Hz, CH_2Ph), 4.45 (d, 1 H, $J_{1',2'}$ 7.3 Hz, H-1′), 4.28 (d, 1 H, $J_{1,2}$ 7.7 Hz, H-1), 3.96 (t, 1 H, $J_{2,3} = J_{3,4} = 9.3$ Hz, H-3), 3.77 (dd, 1 H, $J_{5,6a}$ 6.7, $J_{6a,6b}$ 10.8 Hz, H-6a), 3.70 (dd, 1 H, $J_{5,6b}$ 1.8 Hz, H-6b), 3.55 (s, 3 H, OMe), 3.54 (t, 1 H, $J_{3,4} = J_{4,5} = 9.0$ Hz, H-4), 3.48 (dd, 1 H, $J_{2',3'}$ 9.6 Hz, H-2′), 3.42 (m, 1 H, H-5), 3.41 (dd, 1 H, $J_{2',3'}$

9.6, $J_{3',4'}$ 3.4 Hz, H-3'), 3.38 (dd, 1 H, $J_{2,3}$ 9.2 Hz, H-2), 3.36 (m, 1 H, H-5), 3.33 (m, 2 H, H-6a,6b), 2.03 (s, 3 H, Ac).

A solution of **36** (1.5 g, 1.67 mmol) in tetrahydrofuran (60 mL) under Ar, was treated with NaH (100 mg), then with CS_2 (0.3 mL), MeI (1 mL), α,α'-azobisisobutyronitrile (10 mg), and tributyltin hydride (0.6 mL, 1.7 mmol), as described for the conversion **14** → **15**, to give methyl 2,3,6-tri-*O*-benzyl-4-*O*-(2,3,6-tri-*O*-benzyl-4-deoxy-β-D-*xylo*-hexopyranosyl)-β-D-glucopyranoside (**37**; 1.25 g, 85%). $[\alpha]_D^{20}$ +7.5° (*c* 0.3, $CHCl_3$). ^{1}H-NMR data ($CDCl_3$): δ 7.37–7.19 (m, 30 H, 6 Ph), 4.98 and 4.75 (ABq, 2 H, *J* 11.0 Hz, C*H*$_2$Ph), 4.85 and 4.69 (ABq, 2 H, *J* 11.0 Hz, C*H*$_2$Ph), 4.81 and 4.77 (ABq, 2 H, *J* 10.1 Hz, C*H*$_2$Ph), 4.66 and 4.61 (ABq, 2 H, *J* 11.8 Hz, C*H*$_2$Ph), 4.55 and 4.41 (ABq, 2 H, *J* 12.3 Hz, C*H*$_2$Ph), 4.46 and 4.33 (ABq, 2 H, *J* 12.0 Hz, C*H*$_2$Ph), 4.42 (d, 1 H, $J_{1',2'}$ 7.7 Hz, H-1'), 4.28 (d, 1 H, $J_{1,2}$ 7.8 Hz, H-1), 3.95 (t, 1 H, $J_{2,3} = J_{3,4} = 9.3$ Hz, H-3), 3.81 (dd, 1 H, $J_{5,6a}$ 4.4, $J_{6a,6b}$ 10.9 Hz, H-6a), 3.73 (dd, 1 H, $J_{5,6b}$ 2.0 Hz, H-6b), 3.56 (t, 1 H, $J_{3,4} = J_{4,5} = 9.0$ Hz, H-4), 3.55 (s, 3 H, OMe), 3.48 (ddd, 1 H, $J_{2',3'}$ 8.3, $J_{3',4'a}$ 4.9, $J_{3',4'b}$ 2.0 Hz, H-3'), 3.44–3.32 (m, 3 H, H-5,5',6'a), 3.38 (dd, 1 H, $J_{2,3}$ 9.1 Hz, H-2), 3.30 (dd, 1 H, $J_{5',6'b}$ 6.2, $J_{6'a,6'b}$ 11.4 Hz, H-6'b), 3.24 (dd, 1 H, H-2'), 2.17 (ddd, 1 H, $J_{3',4'a}$ 4.9, $J_{4'a,5'}$ 0.8, $J_{4'a,4'b}$ 12.5 Hz, H-4'a), 1.64 (dt, $J_{3',4'b} = J_{4'b,5'} = 2.0$ Hz, H-4'b).

A solution of **37** (1.30 g, 1.48 mmol) in 2:1 CH_2Cl_2–MeOH (40 mL) was treated with H_2 in the presence of 10% Pd–C (260 mg). The product (0.40 g, 80%) was crystallised from EtOH–water, to give **9**, mp 189–191°, $[\alpha]_D^{20}$ −17° (*c* 1.0, H_2O). NMR data (D_2O): ^{1}H, δ 4.40 (d, 1 H, $J_{1',2'}$ 7.8 Hz, H-1'), 4.37 (d, 1 H, $J_{1,2}$ 7.9 Hz, H-1), 3.96 (bd, 1 H, $J_{5,6a} < 0.5$, $J_{6a,6b}$ 12.0 Hz, H-6a), 3.78 (dd, 1 H, $J_{5,6b}$ 4.4 Hz, H-6b), 3.80–3.66 (m, 3 H, H-4,3',5'), 3.62–3.57 (m, 4 H, H-3,5,6'a,6'b), 3.55 (s, 3 H, OMe), 3.27 (m, 1 H, H-2), 3.18 (dd, 1 H, $J_{2',3'}$ 9.2 Hz, H-2'), 1.94 (ddd, 1 H $J_{3',4'eq}$ 1.8, $J_{4'ax,4'eq}$ 12.6, $J_{5',4'eq}$ 5.1 Hz, H-4'*eq*), 1.41 (dt, $J_{3',4'ax} = J_{4'ax,5'} = 11.9$ Hz, H-4'*ax*); ^{13}C, δ 104.2 (C-1), 104.2 (C-1'), 80.2 (C-4), 76.1 (C-5'), 75.9 (C-5), 75.6 (C-3), 74.0 (C-2), 74.0 and 71.3 (C-2',3'), 64.6 (C-6'), 61.3 (C-6), 58.3 (OCH_3), 35.1 (C-4').

Anal. Calcd for $C_{13}H_{24}O_{10}$: C, 45.88; H, 7.06. Found: C, 46.01; H, 6.98.

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