Synthesis of Methyl (Ethyl 2-*O*-acyl-3,4-di-*O*-benzyl-1-thio-β-D-glucopyranosid)uronates and Evaluation of Their Use as Reactive β-Selective Glucuronic Acid Donors

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The synthesis of derivatives of methyl (ethyl 2-*O*-acyl-3,4-di-*O*-benzyl-1-thio-β-D-glucopyranosid)-uronate with different ester groups (acetyl, benzoyl, pivaloyl, and anisoyl) at O-2 is described. The synthesis proceeds *via* a two-step oxidation (DMSO/DCC followed by PDC/MeOH) of C-6 on a suitably protected glucose derivative to give directly the methyl glucuronic ester without affecting the thioglycoside. The thioglucuronides were tested as donors in coupling reactions with unreactive carbohydrate alcohols using dimethyl(methylthio)sulfonium triflate (DMTST) as promoter. Due to the activating benzyl protecting groups at O-3 and -4, the sluggishness of fully acylated glucuronic acid donors could be overcome and glucuronide disaccharides were produced in fair to good yields. The glucuronides were obtained with the desired β-configuration because of the participating group at O-2. Among the different ester groups tried, the benzoyl group was found to give the highest yield in the couplings.

Introduction

In the course of making biologically active bacterial oligosaccharides, the synthesis of a number of D-glucuronic acid containing oligosaccharide structures, *inter alia*, from *Streptococcus pneumoniae* and *Cryptococcus neoformans*, became of interest. These residues are all β-linked to rather unreactive hydroxyl groups, which causes problems in the syntheses since commonly used glucuronic acid donors are often stable and unreactive toward hindered alcohols; *e.g.*, methyl (2,3,4-tri-*O*-acetyl-α-D-glucopyranosyl bromide)uronate is crystallized from ethanol.[1] This influence of the carboxylic group on the anomeric center is also apparent in the slow rate of hydrolysis of the glycosidic linkage in polysaccharide glucuronides. The problem of synthesizing glucuronides with hindered aglycons can be circumvented by performing the glycosidation with a neutral hexose donor and oxidizing to the glucuronide afterwards, but this approach often requires extensive protective group manipulations, and the oxidation at the oligosaccharide level is not always straightforward. Therefore, we needed to find a glucuronic acid donor reactive enough to give high yields even with hindered aglycons. We also wished to investigate the use of thioglycosides as donors in glucuronide formation. Our results obtained with thioglycosides of neutral hexoses as donors indicate[2] that these also should be of value as glucuronic acid donors. Only few examples of glycosylations using thioglycosides of glucuronic acid as donors have been reported.[3-5]

Results and Discussion

Methyl 3-*O*-benzyl-4,6-*O*-benzylidene-α-D-mannopyranoside[6] (**12**) and 2,3-di-*O*-benzoyl-1,6-anhydro-β-D-glucopyranose[7] (**13**) were chosen as model aglycons, since glucuronic acid linked to the 2-position in mannose and to the 4-position of glucose was found in two of the biologically active oligosaccharides of interest (*Cryptococcus neoformans* serogroup A-D[8] and *Streptococcus pneumoniae* type 3,[9] respectively). Furthermore, they are good examples of rather unreactive aglycons suitable for testing the efficiency of various glucuronic acid donors. As expected, couplings with the usual donor methyl (2,3,4-tri-*O*-acetyl-α-D-glucopyranosyl bromide)uronate,[1] using either silver trifluoromethanesulfonate[10] or mercury salts[11] as promotor, gave low yields of the disaccharides. The glycosidations using the corresponding ethyl β-thioglucuronide as donor and dimethyl(methylthio)-sulfonium triflate (DMTST)[12] as promoter gave similar results. To improve the reactivity of the donor, derivatives with less electron-withdrawing benzyl protecting groups instead of acetyl groups[13] were synthesized. Thus, thioglycoside **3** was tested as donor in couplings with the two acceptors **12** and **13** using DMTST as promoter and was found to give good yields (76 and 69%) of coupling products **14** and **19**, but low stereoselectivity (α/β 2/1 and 1/1, respectively) (see Table 1). The corresponding trichloroacetimidate derivate, activated with BF_3-etherate, which has earlier been synthesized and used by Schmidt *et al.* with good yields and β-selectivity,[14] was also tried, but this gave lower yields (40 and 60%) and a somewhat better but still unsatisfactory β-stereoselectivity (α/β 1/1

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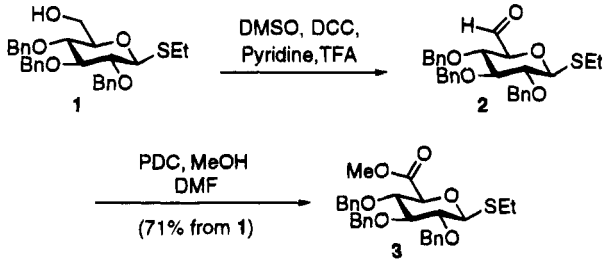
Table 1. Results from Couplings between Acceptors 12 and 13 and Different Thioglucuronide Donors

acceptor	donor with 2-O-group	yield of disaccharide (%)	α/β	molar equiv of donor
12	3, Bn-	76 (14)	2/1	2.5
12	7, Ac-	25 (15)	β	1.5
12	9, Bz-	68 (16)	β	1.5
12	10, MBz-	65 (17)	β	1.5
12	11, Piv-	31 (18)[a]	β	1.5
13	3, Bn-	69 (19)	1/1	1.9
13	7, Ac-	33 (20)	β	1.2
13	9, Bz-	69 (21)	β	1.2
13	9, Bz-	85 (21)	β	1.9
13	10, MBz-	60 (22)	β	1.2
13	11, Piv-	53 (23)	β	1.2

[a] Yield of debenzylidenated product (see Experimental Section).

Scheme 1. Synthesis of Fully Benzylated Thioglucuronide Donor



and 1/2). Derivatives **7**, **9**, **10**, and **11** were therefore synthesized in the hope that the benzyl groups in the 3- and 4-positions would render enough reactivity to the donor, whereas the 2-*O*-acyl group would give β-selectivity by neighboring group participation in the coupling reaction.

The syntheses of the different thioglucuronides needed some attention. Attempts to use standard conditions (BF_3·etherate or $ZnCl_2$ and a thiol), which give high yields of 1,2-*trans* thioglycoside from peracetylated hexoses, gave with peracetylated methyl β-D-glucopyranosiduronate an α,β-mixture of the thioglycoside in moderate yield only (50%). Among the methods earlier published for the synthesis of pure β-linked acetylated ethyl thioglucuronide methyl ester,[15,16] the method described by Sakata *et al.*,[16] a three-step synthesis from the peracetate *via* an anomeric ethyl xanthate, proved to be the best route. To make benzylated derivatives, the acetyl groups were removed from this derivative, and different benzylation methods, under basic as well as neutral conditions, were tried on the ethyl thioglucuronide methyl ester without success. Alkylation can be performed on the glucuronic acid,[17,18] but we decided instead to introduce the benzyl groups before the carboxyl group (Scheme 1) as earlier described by *inter alia* Keglevic *et al.*[19] However, special care has to be taken during the oxidation so as not to oxidize the sulfur atom as well. This was accomplished by a two-step procedure: first a Pfitzner–Moffat oxidation[20] of derivative **1** gave the aldehyde **2**, which then was oxidized by PDC in the presence of methanol to give, directly, the methyl ester **3**, as described by O'Connor and Just.[21] The compounds with both acyl and benzyl protecting groups were synthesized similarly (Scheme 2). Acetylation at O-2 of ethyl 3-*O*-benzyl-4,6-*O*-benzylidene-1-thio-β-D-glucopyranoside (**4**),[22] followed by reductive opening of the benzylidene acetal of **5** using borane·trimethylamine and aluminum chloride in dichloromethane/diethyl ether,[23] gave the 6-OH derivative **6**. This was oxidized in two steps as described above to give the methyl ester glucuronide **7**. Deacetylation, followed by acylation with various acyl chlorides, then gave the various other donors **9**–**11**. The carboxyl group clearly influences also the reactivity of the 2-position as well as the anomeric position as discussed above. Thus, both the deacetylation of **7** and the acylations of **8** were found to be difficult and required both prolonged reaction times and unusually harsh conditions (see Experimental Section). This low reactivity of the 2-position in glucuronic acid derivatives has also been noticed in this laboratory during attempts to perform nucleophilic displacement reactions.

Results from the couplings with these glycosyl donors, **12** and **13** as acceptors, and DMTST as promoter are summarized in Table 1. Due to the participating group at O-2 in the donors, all coupling products (**15**–**18** and **20**–**23**) were found to be β-linked. As is often found, benzoates[24] gave better yields than acetates, the pivaloyl group, which sometimes is used to minimize orthoester formation,[25] also gave lower yields, and the anisoyl group did not improve the yields. So, in summary, the donor methyl (ethyl 2-*O*-benzoyl-3,4-di-*O*-benzyl-1-thio-β-D-glucopyranosid)uronate (**9**) was found to be an efficient donor for the introduction of β-linked glucuronic acid residues at unreactive positions, even when used in low excess. The yields can even be further improved by using a larger excess of the donor. Thus, coupling between **9** and **13** using 1.9 equiv of the donor instead of 1.2 equiv raised the yield from 69 to 85%, based on the aglycon (Table 1). The different protecting groups in these donor derivatives also make further manipulations possible. Thus, in disaccharides **15**–**18**, the acyl group can be removed selectively to allow further elongation or transformations in the 2′-position, whereas in disaccharide **20**–**23** the benzyl groups can be removed to allow reactions at O-3′ and -4′.

Experimental Section

General Remarks. Melting points are corrected. Organic solutions were dried over $MgSO_4$ before concentrating, which was performed under reduced pressure at <40 °C (bath temperature). NMR spectra were recorded at 25 °C at 270 MHz (^1H) or 67.5 MHz (^{13}C) in $CDCl_3$ with Me_4Si as internal standard ($\delta = 0$). TLC was performed on silica gel F_{254} (E. Merck) with detection by UV light and/or by charring with 8% sulfuric acid. Silica gel (0.040–0.063 mm, Amicon) was used for column chromatography.

Methyl (Ethyl 2,3,4-tri-*O*-benzyl-1-thio-β-D-glucopyranosid)uronate (3). Ethyl 2,3,4-tri-*O*-benzyl-1-thio-β-D-glucopyranoside (**1**) (1.07 g, 2.17 mmol) was dissolved in DMSO (30 mL) at rt. To the solution was added pyridine (175 μL, 2.17 mmol), trifluoroacetic acid (86 μL, 1.12 mmol), and DCC (1.55 g, 7.52 mmol), and the mixture stirred overnight. Oxalic acid (1 g), dissolved in MeOH (25 mL), was added to the

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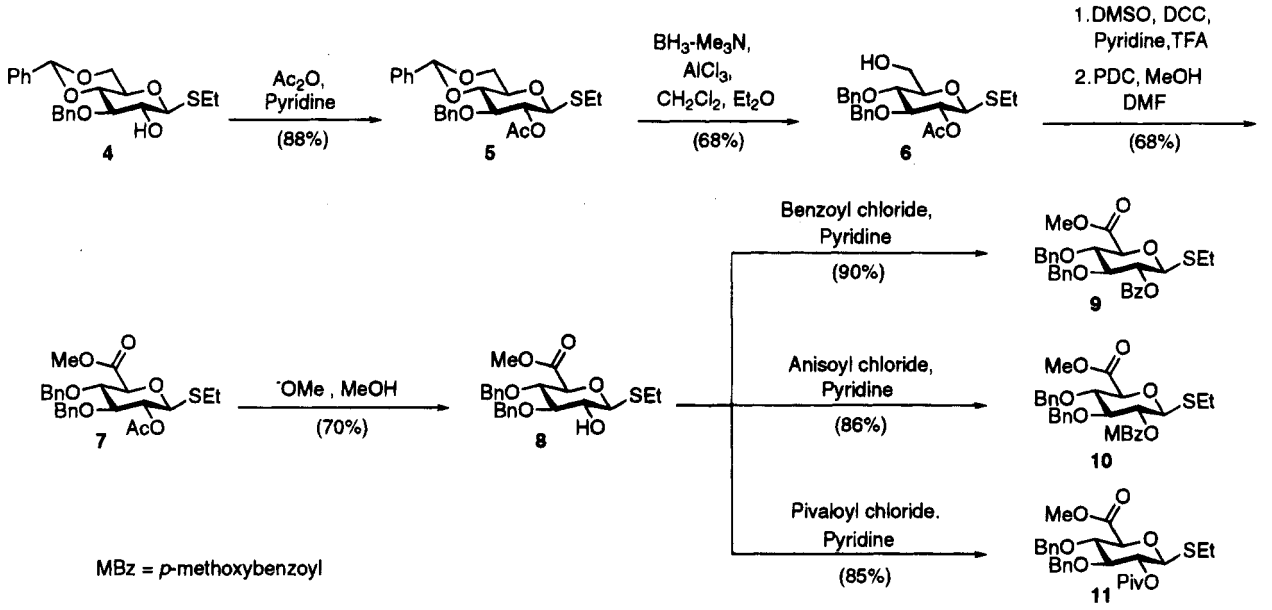
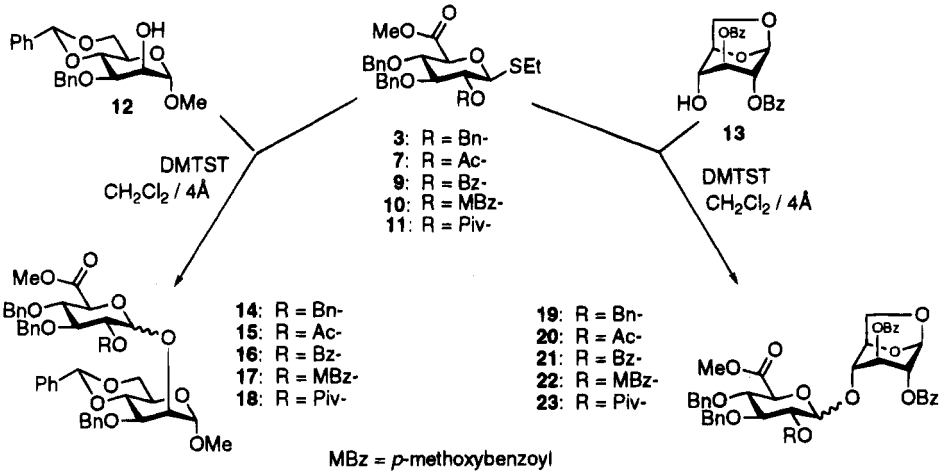
Scheme 2. Synthesis of 2-O-Acylated Thioglucuronide Donors



Scheme 3. Couplings between Different Thioglucuronide Donors and Acceptors 12 or 13



reaction mixture in order to precipitate formed *N,N*-dicyclohexylurea, and after 30 min the mixture was filtered. Toluene and aqueous (aq) saturated NaCl were added to the filtrate, and the phases were separated. The organic phase was further washed with water, aq $NaHCO_3$, and water. After drying and evaporation of the organic phase, the crude aldehyde **2** was dried *in vacuo* and used for the next oxidation without further purification. NMR showed that the aldehyde still contained *N,N*-dicyclohexylurea, but silica gel chromatography at this stage did not improve the overall yield.

Aldehyde **2** was dissolved in dry DMF (25 mL) containing MeOH (527 μL, 13 mmol) and cooled on ice for 30 min in a reaction vessel covered to exclude light. PDC (3.81 g, 13 mmol) was added in one portion. The mixture was allowed to attain rt and react overnight and was then transferred to a short silica gel column with a top layer of EtOAc where chromic salts precipitate. After 30 min the column was eluted with EtOAc, and all carbohydrate-containing fractions were pooled. The solvent was evaporated, and further purification was accomplished with flash chromatography (isooctane–EtOAc 6:1) which gave crystalline **3** in 71% yield (803 mg, 1.53 mmol). Compound **3** was recrystallized from EtOAc–hexane: mp 128–30 °C; $[\alpha]_D$-11° (*c* 1.1, chloroform); ^{13}C NMR δ 15.0, 25.1, 52.5, 75.1, 75.6, 75.8, 78.1, 79.3, 81.2, 85.8 (two C), 127.8, 127.9, 128.0, 128.3, 128.4, 137.8, 138.2, 168.7. Anal. Calcd for $C_{30}H_{34}O_6S$: C, 68.94; H, 6.56. Found: C, 68.95; H, 6.67.

Ethyl 2-*O*-Acetyl-3-*O*-benzyl-4,6-*O*-benzylidene-1-thio-β-D-glucopyranoside (5). Compound **4** (4.61 g, 11.5 mmol) was dissolved in pyridine–acetic acid anhydride (2:1, 120 mL). After 2 h the mixture was evaporated and then coevaporated twice with toluene. The residue was crystallized from EtOAc–hexane and gave 4.48 g of **5** (10.8 mmol, 88%): mp 107–109 °C; $[\alpha]_D$-22° (*c* 0.7, chloroform); ^{13}C NMR δ 14.8, 20.9, 23.9, 68.6, 70.6, 71.2, 74.3, 79.7, 81.5, 84.2, 101.2, 126.0, 127.7, 127.8, 128.3, 129.0, 137.1, 138.1, 169.2. Anal. Calcd for $C_{24}H_{28}O_6S$: C, 64.84; H, 6.35. Found: C, 64.72; H, 6.24.

Ethyl 2-*O*-Acetyl-3,4-di-*O*-benzyl-1-thio-β-D-glucopyranoside (6). A solution of aluminum chloride (1.47 g, 11.0 mmol) in diethyl ether (15 mL) was added during 10 min to a stirred mixture of **5** (1.23 g, 2.76 mmol), borane-trimethylamine complex (8.6 g, 110 mmol), and 4 Å molecular sieves in dichloromethane (50 mL) and diethyl ether (10 mL) at 0 °C. After 1 h the mixture was filtered through Celite, and 1 M H_2SO_4 was added to the filtrate, which then was stirred for 30 min. The phases were separated, and the organic layer was washed with aq $NaHCO_3$ (twice) and water. After drying, the solvents were evaporated. Flash chromatography (toluene–EtOAc 8:1) repeated twice gave 68% (842 mg, 1.89 mmol) of **6** which easily crystallized from EtOAc–isooctane: mp 115–116 °C; $[\alpha]_D$-8° (*c* 1.0, chloroform); ^{13}C NMR ^{13}C, δ 14.9, 20.9, 24.0, 61.9, 71.8, 75.1, 75.2, 77.6, 79.6, 83.6, 84.2, 127.8, 128.0, 128.1, 128.5, 129.0, 137.8, 138.0, 169.6. Anal. Calcd for $C_{24}H_{30}O_6S$: C, 64.55; H, 6.77. Found: C, 64.63; H, 6.85.

In addition, 5% (52 mg, 0.12 mmol) of the regioisomer ethyl 2-*O*-acetyl-3,6-di-*O*-benzyl-1-thio-β-D-glucopyranoside was also isolated. ^{13}C NMR δ 14.9, 21.0, 23.9, 70.4, 71.3, 72.1, 73.7,

Table 2. Glucuronic Acid Disaccharides, ^{13}C NMR Data

compd	NMR data, δ
14α	52.5 (OCH_3 uronate), 54.9 (OCH_3 Man), 64.3, 68.7, 70.7, 71.6, 74.0, 74.6, 75.3, 75.7, 76.7, 79.2, 79.4, 80.6 (C-2-6,2′-5′,OCH_2Ar), 97.6 ($J_{C\text{-}1,H\text{-}1}$ = 173 Hz, C-1′), 101.0 ($J_{C\text{-}1,H\text{-}1}$ = 174 Hz, C-1), 101.3 ($J_{C,H}$ = 163 Hz, OCHAr), 126.0, 127.8–128.9, 137.8, 137.9, 138.3, 138.8 (aromatic C), 170.0 (C=O uronate)
14β	52.3 (OCH_3 uronate), 54.9 (OCH_3 Man), 64.0, 69.0, 71.4, 74.0, 74.7, 75.2, 75.3, 75.8, 76.0, 78.4, 78.9, 81.3, 84.0 (C-2-6,2′-5-,OCH_2Ar), 99.7 ($J_{C\text{-}1,H\text{-}1}$ = 169 Hz, C-1), 101.6 ($J_{C,H}$ = 165 Hz, OCHAr), 103.3 ($J_{C\text{-}1,H\text{-}1}$ = 158 Hz, C-1′), 126.1, 127.4–128.8, 137.6, 137.9, 138.0, 138.4, 138.5 (aromatic C), 168.4 (C=O uronate)
15	20.9 (CH_3 acetyl), 52.4 (OCH_3 uronate), 54.9 (OCH_3 Man), 64.1, 68.7, 71.7, 72.4, 74.0, 74.8, 74.9, 75.2, 76.1, 78.0, 79.0, 81.9 (C-2-6, 2′-5′, OCH_2Ar), 99.4 ($J_{C\text{-}1,H\text{-}1}$ = 169 Hz, C-1), 100.7 ($J_{C\text{-}1,H\text{-}1}$ = 160 Hz, C-1′), 101.5 ($J_{C,H}$ = 163 Hz, OCHAr), 126.1, 127.3–128.7, 137.6, 137.7, 138.0, 138.6 (aromatic C), 168.1 (C=O uronate), 169.1 (C=O acetyl)
16	52.5 (OCH_3 uronate), 54.7 (OCH_3 Man), 63.9, 68.6, 71.8, 73.3, 74.2, 74.8, 74.9, 75.2, 76.3, 78.2, 79.0, 81.6 (C-2-6, 2′-5′, OCH_2Ar), 99.1 ($J_{C\text{-}1,H\text{-}1}$ = 169 Hz, C-1), 100.8 ($J_{C\text{-}1,H\text{-}1}$ = 160 Hz, C-1′), 101.3 ($J_{C,H}$ = 166 Hz, OCHAr), 126.0, 127.2–130.0, 133.0, 137.6, 137.7, 138.8 (aromatic C), 164.8 (C=O benzoyl), 168.3 (C=O uronate)
17	52.4 (OCH_3 uronate), 54.8 (OCH_3 Man), 55.5 ($ArOCH_3$), 63.9, 68.6, 70.6, 71.8, 73.1, 74.3, 74.8, 74.9, 75.1, 76.3, 78.2, 79.0, 81.7 (C-2-6, 2′-5′, OCH_2Ar), 99.3 ($J_{C\text{-}1,H\text{-}1}$ = 169 Hz, C-1), 100.8 ($J_{C\text{-}1,H\text{-}1}$ = 159 Hz, C-1′), 101.3 ($J_{C,H}$ = 163 Hz, OCHAr), 113.5, 122.4, 126.0, 127.2–128.7, 131.7, 137.7, 138.8 (aromatic C), 163.5, 164.5 (aromatic C and C=O *p*-methoxybenzoyl), 168.3 (C=O uronate)
18[a]	99.2 ($J_{C\text{-}1,H\text{-}1}$ = 168 Hz, C-1), 100.5 ($J_{C\text{-}1,H\text{-}1}$ = 159 Hz, C-1′), 101.4 ($J_{C,H}$ = 165 Hz, OCHAr)
19[b]	52.3, 52.5 (OCH_3 uronate), 65.0, 65.2, 68.7, 69.1, 70.1, 70.4, 71.1, 73.0, 73.9, 74.5, 74.8, 75.0, 75.3, 75.8, 75.8, 76.4, 78.8, 79.0, 79.4, 81.1, 82.0, 84.0 (C-2-6, 2′-5′, OCH_2Ar), 98.6 ($J_{C\text{-}1,H\text{-}1}$ = 174 Hz, C-1′α), 99.1 ($J_{C\text{-}1,H\text{-}1}$ = 180 Hz, C-1), 103.3 ($J_{C\text{-}1,H\text{-}1}$ = 161 Hz, C-1′β), 127.6–130.2, 133.2, 133.3, 133.5, 133.7, 137.9, 138.0, 138.2, 138.3, 138.6 (aromatic C), 164.7, 164.9, 165.4, 165.4 (C=O benzoyl), 168.4, 170.0 (C=O uronate)
20	20.8 (CH_3 acetyl), 52.4 (OCH_3 uronate), 65.0, 68.6, 70.3, 72.6, 73.7, 74.6, 75.0, 75.0, 75.8, 79.1, 82.1 (C-2-6, 2′-5-, OCH_2Ar), 99.0 ($J_{C\text{-}1,H\text{-}1}$ = 178 Hz, C-1), 100.5 ($J_{C\text{-}1,H\text{-}1}$ = 158 Hz, C-1′), 127.3–130.3, 133.4, 133.6, 137.7, 137.9 (aromatic C), 164.7, 165.4 (C=O benzoyl), 168.2 (C=O uronate), 169.2 (C=O acetyl)
21	52.3 (OCH_3 uronate), 65.0, 68.7, 70.0, 73.5, 73.9, 74.6, 74.9, 75.0, 76.6, 79.0, 81.8 (C-2-6, 2′-5′, OCH_2Ar), 98.9 ($J_{C\text{-}1,H\text{-}1}$ = 182 Hz, C-1), 101.2 ($J_{C\text{-}1,H\text{-}1}$ = 158 Hz, C-1′), 127.7–130.2, 133.2, 133.3, 133.6, 137.5, 137.8 (aromatic C), 164.9, 164.9, 165.5 (C=O benzoyl), 168.3 (C=O uronate)
22	52.3 (OCH_3 uronate), 55.4 ($ArOCH_3$), 65.0, 68.8, 70.0, 73.4, 74.0, 74.6, 74.8, 75.0, 76.6, 78.9, 81.9 (C-2-6, 2′-5′, OCH_2Ar), 98.9 ($J_{C\text{-}1,H\text{-}1}$ = 181 Hz, C-1), 101.3 ($J_{C\text{-}1,H\text{-}1}$ = 159 Hz, C-1′), 113.7, 122.0, 127.7–130.3, 131.8, 133.3, 133.6, 137.6, 137.9 (aromatic C), 163.6, 164.6, 164.9, 165.5 (aromatic C and C=O *p*-methoxybenzoyl, C=O benzoyl), 168.3 (C=O uronate)
23	27.0, 27.2 (CH_3 pivaloyl), 38.8 (C pivaloyl), 52.4 (OCH_3 uronate), 65.0, 68.5, 70.2, 72.5, 73.8, 74.6, 74.9, 74.9, 75.9, 78.8, 82.5 (C-2-6, 2′-5′, OCH_2Ar), 99.0 ($J_{C\text{-}1,H\text{-}1}$ = 178 Hz, C-1), 100.9 ($J_{C\text{-}1,H\text{-}1}$ = 158 Hz, C-1′), 127.5–130.3, 133.3, 133.6, 137.8, 137.8 (aromatic C), 164.8, 165.4 (C=O benzoyl), 168.3 (C=O uronate), 176.4 (C=O pivaloyl)

[a] Complete data not available, see Experimental Section. [b] Spectra recorded on an α/β-mixture of the compound.

74.6, 78.1, 83.5, 83.6, 127.5, 127.7, 127.8, 128.0, 128.1, 128.3, 128.5, 137.7, 138.3, 169.6.

The ^{13}C NMR signal at 61.9 ppm in **6** and the absence of that signal in the regioisomer show the positions of the OH groups.

Methyl (Ethyl 2-*O*-acetyl-3,4-di-*O*-benzyl-1-thio-β-glucopyranosid)uronate (7). Compound **6** (3.23 g, 7.25 mmol) was oxidized as described for compound **3** except for flash chromatography (toluene–EtOAc 14:1) and recrystallization (EtOAc–hexane), and 2.35 g of **7** was isolated (68%, 4.93 mmol): mp 95 °C; $[\alpha]_D$-13° (*c* 1.0, chloroform); ^{13}C NMR δ 14.7, 20.9, 23.9, 52.5, 71.1, 75.0, 75.2, 78.2, 79.3, 83.4, 84.1, 127.8, 127.9, 128.0, 128.2, 128.4, 137.6, 137.9, 168.3, 169.4. Anal. Calcd for $C_{25}H_{30}O_7S$: C, 63.26; H, 6.38. Found: C, 63.24; H, 6.30.

Methyl (Ethyl 3,4-di-*O*-benzyl-1-thio-β-D-glucopyranosid)uronate (8). Sodium methoxide in MeOH (0.2 mL, 1 M) was added to a solution of **7** (1.85 g, 3.88 mmol) in MeOH (50 mL). After 20 h the mixture was neutralized with Dowex 50 (H^+) resin, filtered, and evaporated. The residue was purified by column chromatography (toluene–EtOAc 13:1) to yield **8**. Fractions still containing **7** were treated as above for 4 h, and a total of 1.17 g of **8** (2.69 mmol, 70%) was isolated. Recrystallization from EtOAc–hexane gave needles: mp 74–75 °C; $[\alpha]_D$-31° (*c* 1.2, chloroform); ^{13}C NMR δ 15.1, 24.4, 52.5, 72.8, 75.0, 75.2, 78.3, 78.7, 85.0, 86.7, 127.8, 127.9, 128.3, 128.5, 137.8, 138.3, 168.6. Anal. Calcd for $C_{23}H_{28}O_6S$: C, 63.87; H, 6.53. Found: C, 64.01; H, 6.51.

Methyl (Ethyl 2-*O*-benzoyl-3,4-di-*O*-benzyl-1-thio-β-D-glucopyranosid)uronate (9). Benzoyl chloride (280 μL, 2.42 mmol) was added to a solution of **8** (350 mg, 0.80 mmol) in pyridine (25 mL) at 65 °C. After 66 h the reaction mixture was allowed to attain rt, and then it was partitioned between toluene and 1 M HCl. The organic layer was washed with 1 M HCl, aq $NaHCO_3$ (twice), and water (twice), dried, and evaporated. Flash chromatography (toluene–EtOAc 10:1) and recrystallization (EtOAc–hexane) of the residue yielded 90% of **9** (391 mg, 0.72 mmol): mp 105–106 °C; $[\alpha]_D$ +26° (*c* 1.0, chloroform); ^{13}C NMR δ 14.7, 23.9, 52.5, 71.8, 75.1, 75.2, 78.3, 79.3, 83.3, 84.1, 127.7, 128.0, 128.3, 128.4, 129.8, 133.2, 137.5, 137.6, 165.1, 168.3. Anal. Calcd for $C_{30}H_{32}O_7S$: C, 67.13; H, 6.02. Found: C, 67.04; H, 5.97.

Methyl (Ethyl 3,4-di-*O*-benzyl-2-*O*-(*p*-methoxybenzoyl)-1-thio-β-D-glucopyranosid)uronate (10). Anisoyl chloride (287 μL, 1.93 mmol) was added to a solution of **8** (279 mg, 0.64 mmol) in pyridine (25 mL) at 65 °C. After 20 h the reaction mixture was allowed to attain rt, and then it was partitioned between toluene and 1 M HCl. The organic layer was washed with 1 M HCl, aq $NaHCO_3$ (twice), and water (twice), dried, and evaporated. Column chromatography (toluene–EtOAc 14:1) and recrystallization (EtOAc–hexane) of the residue yielded 86% of **10** (311 mg, 0.55 mmol): mp 128–130 °C; $[\alpha]_D$ +30° (*c* 0.9, chloform); ^{13}C NMR δ 14.8, 24.0, 52.6, 55.6, 71.7, 75.3, 75.4, 78.5, 79.3, 83.5, 84.4, 113.8, 122.1, 127.8, 128.1, 128.2, 128.4, 128.5, 132.0, 137.7, 137.9, 163.8, 164.9, 168.6. Anal. Calcd for $C_{31}H_{34}O_8S$: C, 65.70; H, 6.05. Found: C, 65.71; H, 5.97.

Methyl (Ethyl 3,4-di-*O*-benzyl-2-*O*-pivaloyl-1-thio-β-D-glucopyranosid)uronate (11). Pivaloyl chloride (302 μL, 2.42 mmol) was added to a solution of **8** (350 mg, 0.80 mmol)

in pyridine (25 mL) at 65 °C. Further addition of pivaloyl chloride was done after 3, 4, and 5 days (302 μL, 2.42 mmol, each time). After 6 days the reaction mixture was allowed to attain rt, and then it was partitioned between toluene and 1 M HCl. The organic layer was washed with 1 M HCl, aq $NaHCO_3$ (twice), and water (twice), dried, and evaporated. Flash chromatography (toluene–EtOAc 14:1) and recrystallization (EtOAc–hexane) of the residue yielded 85% of **11** (363 mg, 0.70 mmol): mp 116 °C; $[\alpha]_D$-33° (*c* 1.0, chloroform); ^{13}C NMR δ 14.7, 23.8, 27.1, 38.7, 52.5, 70.9, 75.1, 75.2, 78.3, 79.1, 83.8, 84.2, 127.4, 127.7, 127.9, 128.0, 128.4, 137.7, 137.9, 168.4, 176.7. Anal. Calcd for $C_{28}H_{36}O_7S$: C, 65.09; H, 7.02. Found: C, 65.02; H, 7.00.

Procedure for Preparation of Disaccharides 14-23. Acceptor (**12** or **13**, 40–75 mg, 0.11–0.20 mmol) and donor (**3**, **7**, **9**, **10**, or **11**) were dissolved in 5–8 mL of distilled CH_2Cl_2 at rt together with 0.5 g of 4 Å molecular sieves. DMTST (4 equiv to acceptor) was added to the stirred solution after 30 min. The reaction was monitored by TLC (toluene-EtOAc 4:1) and quenched with triethylamine (1 mL) after an appropriate time (1–6 h, depending on donor–acceptor pair). The whole reaction mixture was then directly transferred to the top of a silica gel column and eluted (flash cromatography, toluene–EtOAc 20:1–14:1) to yield disaccharides **14–23**. In some cases additional purification was needed, and this was accomplished by column chromatography (toluene–EtOAc 14:1–10:1). **Products.** Details about yields, α/β-ratio, and equivalents of donor used are found in Table 1, and NMR data are in Table 2. The α/β-ratio of **14** was determined by separation of the disaccharides and NMR studies. For **19**, only NMR spectroscopy was used, because separation of the two forms was not possible. The yield of **18** is the yield after debenzylidenation (70% AcOH (aq) at 60 °C for 2 h, followed by evaporation and silica gel chromatography), since **18** could not be purified from byproducts (hydrolysis of **11**).

Methyl 3-*O*-benzyl-4,6-*O*-benzylidene-2-*O*-(methyl 2,3,4-tri-*O*-benzyl-α-D-glucopyranosyluronate)-α-D-mannopyranoside (14α): mp 128–30 °C (from EtOAc–isooctane). Anal. Calcd for $C_{49}H_{52}O_{12}$: C, 70.66; H, 6.29. Found: C, 69.69; H, 6.45.

Methyl 3-*O*-benzyl-4,6-*O*-benzylidene-2-*O*-(methyl 2,3,4-tri-*O*-benzyl-β-D-glucopyranosyluronate)-α-D-mannopyranoside (14β): syrup. Anal. Calcd for $C_{49}H_{52}O_{12}$: C, 70.66; H, 6.29. Found: C, 70.71; H, 6.35.

Methyl 3-*O*-benzyl-4,6-*O*-benzylidene-2-*O*-(methyl 2-*O*-acetyl-3,4-di-*O*-benzyl-β-D-glucopyranosyluronate)-α-D-mannopyranoside (15): syrup. Anal. Calcd for $C_{44}H_{48}O_{13}$: C, 67.34; H, 6.16. Found: C, 67.49; H, 6.32.

Methyl 3-*O*-benzyl-4,6-*O*-benzylidene-2-*O*-(methyl 2-*O*-benzoyl-3,4-di-*O*-benzyl-β-D-glucopyranosyluronate)-α-D-mannopyranoside (16): mp 121–22 °C (from EtOAc–hexane). Anal. Calcd for $C_{49}H_{49}O_{12}$: C, 69.49; H, 5.83. Found: C, 69.25; H, 5.96.

Methyl 3-*O*-benzyl-4,6-*O*-benzylidene-2-*O*-(methyl 3,4-di-*O*-benzyl-2-*O*-(*p*-methoxybenzoyl)-β-D-glucopyranosyluronate)-α-D-mannopyranoside (17): mp 146–47 °C (from EtOAc–hexane). Anal. Calcd for $C_{50}H_{51}O_{14}$: C, 68.48; H, 5.86. Found: C, 68.28; H, 5.99.

2,3-Di-*O*-benzoyl-4-*O*-(methyl 2,3,4-tri-*O*-benzyl-α,β-D-glucopyranosyluronate)-1,6-anhydro-β-D-glucopyranose (19): syrup. Anal. Calcd for $C_{48}H_{46}O_{13}$: C, 69.39; H, 5.58. Found: C, 68.57; H, 5.72.

2,3-Di-*O*-benzoyl-4-*O*-(methyl 2-*O*-acetyl-3,4-di-*O*-benzyl-β-D-glucopyranosyluronate)-1,6-anhydro-β-D-glucopyranose (20): syrup. Anal. Calcd for $C_{43}H_{42}O_{14}$: C, 65.98; H, 5.41. Found: C, 65.83; H, 5.58.

2,3-Di-*O*-benzoyl-4-*O*-(methyl 2-*O*-benzoyl-3,4-di-*O*-benzyl-β-D-glucopyranosyluronate)-1,6-anhydro-β-D-glucopyranose (21): mp 198–200 °C (from EtOAc–hexane). Anal. Calcd for $C_{48}H_{43}O_{14}$: C, 68.24; H, 5.13. Found: C, 68.00; H, 5.26.

2,3-Di-*O*-benzoyl-4-*O*-(methyl 3,4-di-*O*-benzyl-2-*O*-(*p*-methoxybenzoyl)-β-D-glucopyranosyluronate)-1,6-anhydro-β-D-glucopyranose (22): mp 195–197 °C (from EtOAc–hexane). Anal. Calcd for $C_{49}H_{45}O_{15}$: C, 67.27; H, 5.18. Found: C, 67.17; H, 5.31.

2,3-Di-*O*-benzoyl-4-*O*-(methyl 3,4-di-*O*-benzyl-2-*O*-pivaloyl-β-D-glucopyranosyluronate)-1,6-anhydro-β-D-glucopyranose (23): mp 165–66 °C (from EtOAc–hexane). Anal. Calcd for $C_{46}H_{48}O_{14}$: C, 66.98; H, 5.87. Found: C, 66.04; H, 5.98.

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Supplementary Material Available: 270 MHz ^1H NMR spectra of **14-17** and **19-23** (10 pages). This material is contained in libraries on microfiche, immediately follows this article in the microfilm version of the journal, and can be ordered from the ACS; see any current masthead page for ordering information.

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