The C-Disaccharide α-C(1→3)-Mannopyranoside of *N*-Acetylgalactosamine Is an Inhibitor of Glycohydrolases and of Human α-1,3-Fucosyltransferase VI. Its Epimer α-(1→3)-Mannopyranoside of *N*-Acetyltalosamine Is Not

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The radical C-glycosidation of (−)-(1*S*,4*R*,5*R*,6*R*)-6-*endo*-chloro-3-methylidene-5-*exo*-(phenylseleno)-7-oxabicyclo[2.2.1]heptan-2-one ((−)-**4**) with 2,3,4,6-tetra-*O*-acetyl-α-D-mannopyranosyl bromide gave (+)-(1*S*,3*R*,4*R*,5*R*,6*R*)-6-*endo*-chloro-5-*exo*-(phenylseleno)-3-*endo*-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-glycero-D-*manno*-heptitol-7′-C-yl)-7-oxabicyclo[2.2.1]hept-2-one ((+)-**5**) that was converted into (+)-(1*R*,2*S*,5*R*,6*R*)-5-acetamido-3-chloro-2-hydroxy-6-(1′,3′,4′,5′-tetra-*O*-acetyl)-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-C-yl)cyclohex-3-en-1-yl acetate ((+)-**10**) and into (+)-(1*R*,2*S*,5*R*,6*S*)-5-bromo-3-chloro-2-hydroxy-6-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-C-yl)cyclohex-3-en-1-yl acetate ((+)-**19**). Ozonolysis of (+)-**10** and further transformations provided 2-acetamido-2,3-dideoxy-3-C−(2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-C-yl)-D-galactose (α-C(1→3)-D-mannopyranoside of *N*-acetylgalactosamine (α-D-Man*p*-(1→3)CH_2-D-GalNAc): **1**). Displacement of the bromide (+)-**19** with NaN_3 in DMF provided the corresponding azide ((−)-**20**) following a S_N2 mechanism. Ozonolysis of (−)-**20** and further transformations led to 2-acetamido-2,3-dideoxy-3-C−(2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-C-yl)-D-talose (α-C(1→3)-D-mannopyranoside of *N*-acetyl D-talosamine (α-D-Man*p*-(1→3)-CH_2-D-TalNAc): **2**). The neutral C-disaccharide **1** inhibits several glycosidases (e.g., β-galactosidase from jack bean with $K_i = 7.5\ \mu$M, α-L-fucosidase from human placenta with $K_i = 28\ \mu$M, β-glucosidase from *Caldocellum saccharolyticum* with $K_i = 18\ \mu$M) and human α-1,3-fucosyltransferase VI (Fuc-TVI) with $K_i = 120\ \mu$M whereas it 2-epimer **2** does not. Double reciprocal analysis showed that the inhibition of Fuc-TVI by **1** displays a mixed pattern with respect to both the donor sugar GDP-fucose and the acceptor LacNAc with K_i of 123 and 128 μM, respectively.

Introduction

The interaction between cell surface carbohydrates and their protein receptors is implicated in several important biological events.[1] Carbohydrate mimetics are potentially useful tools to study cellular interactions, the biosynthesis of glycoproteins, the catabolism of glycoconjugates,[2] and the mechanisms of digestion.[3] Inhibitors of enzymes involved in these processes such as the glycosidases and the glycosyltransferases are potential anticancer, antiviral, and antidiabetic agents; they can also have insect antifeedant effects.[4] Disaccharide mimetics such as the C-disaccharides[5] are potential glycosidase and glycosyltransferase inhibitors and may represent nonhydrolyzable epitopes.[6] We present the syntheses of two new

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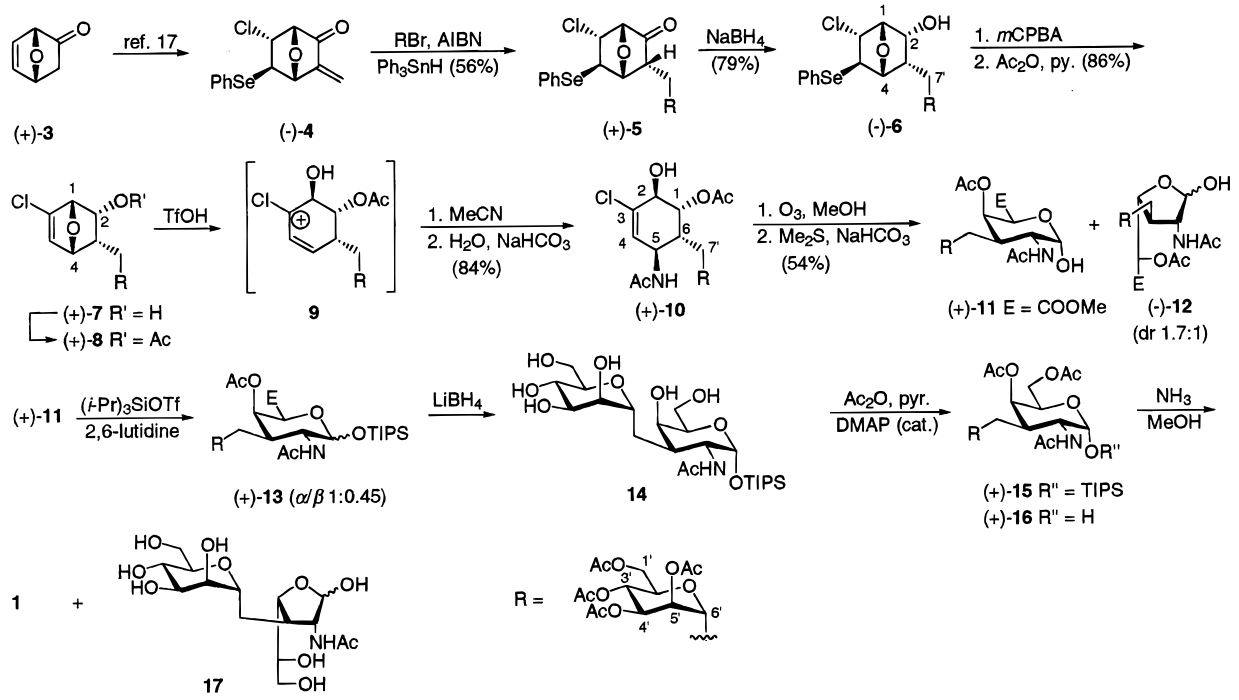
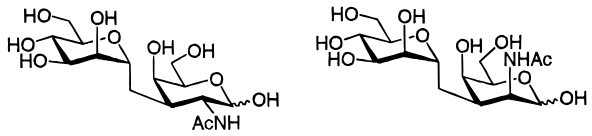
Scheme 1



C-disaccharides, i.e.: α-D-Man*p*(1→3)CH_2-D-GalNAc (**1**)[7] and its epimer α-D-Man*p*(1→3)CH_2-D-TalNAc (**2**) and show that **1** inhibits several glycosidase and human α-1,3-fucosyltransferase VI (Fuc-TVI) whereas **2** does not.



Fucosylated glycoconjugates play important roles in physiological and pathological processes such as fertilization, embryogenesis, lymphocyte trafficking, immune response, and cancer metastasis.[8] Among the eight Fuc-Ts cloned to date,[9] five of them catalyze the transfer from GDP-Fucose to *N*-acetylglucosamine in an α-1,3 linkage. These Fuc-Ts are designated Fuc-TIII to Fuc-TVII and differ in substrate specificity, cation requirement, sensitivity to inhibitors, and tissue distribution.[10] Cell surface α-1,3-fucosylated structures including the trisaccharide Lewis x (Le^x) and the tetrasaccharide sialyl-Lewis x (sLe^x) are involved in a variety of cell−cell interactions such as inflammation[11] and tumor metastasis.[12] Indeed, α-1,3-fucosylated glycoconjugates have been identified as ligands for E- and P-selectins, cell-adhesion molecules involved in recruitment of leukocytes into the site of inflammation.[13] Furthermore, many studies have shown that invasiveness of tumor cells is in correlation with an increase of serum Fuc-T activity or in fucose incorporation into certain surface glycoproteins.[14]

Synthesis of the C-Disaccharides. Giese's[15] radical C-glucosidation[16] and C-galactosidation[17] of enone (−)-**4** derived from (+)-(1*R*,4*R*)-7-oxabicyclo[2.2.1]hept-5-en-2-one ((+)-**3**: a "naked sugar" of the first generation)[18] has been shown to be highly diastereoselective, giving C-glycosides that have been converted into C-disaccharides and C-glycosides of carbapentopyranoses[19] and analogues.[20] Under modified conditions, the radical C-mannosidation between 2,3,4,6-tetra-*O*-acetyl-α-D-mannopy-

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ranosyl bromide and (−)-**4** is possible in benzene using Ph_3SnH as hydrogen donor, giving (+)-**5** in 56% yield (Scheme 1). The *endo* relative configuration of (+)-**5** was given by the coupling constant ^3J(H-3*exo*,H-4) = 5.6 Hz.[21] No trace of the isomer *exo* C-mannoside could be detected in the 400 MHz ^{1}H NMR spectrum of the crude reaction mixture.[16,17] The α-configuration of the α-C-mannoside (C-6′) was confirmed by the observation of ^3J(H-2′,H-3′) = 2.7 Hz, ^3J(H-5′,H-6′) = 8.5 Hz, and of a NOE between the signals of (+)-**5** assigned to H-6′ (δ_H 3.98 ppm) and H-1′ (δ_H 4.56 ppm).

As expected, reduction of the ketone (+)-**5** with $NaBH_4$ was *exo* face selective giving alcohol (−)-**6** (79%). Oxidative elimination of the benzeneselenyl group with *m*-CPBA gave (+)-**7**, and subsequent acetylation provided (+)-**8** (86%). Acid-promoted (CF_3SO_3H) oxa-ring opening of the 7-oxanorbornene (+)-**8** in anhydrous acetonitrile produced, after aqueous workup ($NaHCO_3$), the aminoconduritol derivative (+)-**10** (84%). Ozonolysis of chloroalkene (+)-**10** generated an acyl chloride−aldehyde intermediate that reacted with MeOH[22] to produce a 1.4:1 mixture of the methyl uronates (+)-**11** and (−)-**12** separated by flash chromatography on silica gel. The *galacto* configuration of the pyranose moiety of (+)-**11** was given by the vicinal coupling constant ^3J(H-2,H-3) = 9.9 Hz; ^3J(H-1,H-2) = 3.0 Hz confirmed the α-anomer and δ(C-1) = 91.5 ppm the pyranose system (δ(C-1) = 95.5 for the furanose system).[23] The high regioselectivity of the acid-induced isomerization (+)-**8** → (+)-**10** can be interpreted in terms of the preferred cleavage of C(4)−O(7) bond leading to the 1-chloroallyl cation intermediate **9**. Cleavage of the C(1)−O(7) bond would generate a 2-chloroallyl cation intermediate less stable than **9**. MeCN reacts on the less sterically hindered face of **9** giving an imminium ion intermediate that reacts with H_2O to afford (+)-**10** (Ritter reaction[24]).

Silylation of (+)-**11** with (i-Pr)$_3$SiOSO$_2$CF$_3$ and 2,6-lutidine gave (+)-**13** (1:0.45 mixture of α- and β-pyranoside). Distinction between the α- (major) and β-pyranoside (minor) was based on the vicinal coupling constants ^3J(H-1,H-2) = 3.3 Hz (**13α**) and 7.8 Hz (**13*β***). The β-D-pyranoside structure of **13*β*** was confirmed by the observation of NOE's between signals assigned to H-1 (4.89 ppm), H-3 (2.43 ppm), H-5 (4.21 ppm). Uronic ester (+)-**13α** was reduced with $LiBH_4$ affording **14** that was acetylated to give (+)-**15** (46% based on (+)-**11**). Desilylation with Bu_4NF of (+)-**15** furnished (+)-**16** (74%); then ammonolysis in MeOH, followed by chromatographic separation (Sep-Pak RP18, H_2O), afforded **1** (mixture of α- and β-pyranose) and **17** (mixture of α- and β-furanose) that was tested as such for its inhibitory activities toward glycosidases and glycosyltransferases (see below).

Treatment of the 7-oxanorbornene (+)-**8** with BBr_3 in CH_2Cl_2 at 0 °C gave (+)-**19** in 72% yield. Double irradiations in the ^{1}H NMR spectrum of (+)-**19** demonstrated that the hydroxy group resides at C-2 in the cyclohexenol moiety. Coupling constants between vicinal protons suggest the half-chair conformation shown in Figure 1. Product (+)-**19** results probably from the cationic intermediate **9** (Scheme 1) that is quenched by Br^- onto its less sterically hindered face. Alternatively, (+)-**19** could arise from an intramolecular transfer of Br^- in the zwitterion intermediate **18** (Scheme 2).[25] Displacement of the allylic bromide (+)-**19** with NaN_3/DMF (60 °C) occurred with complete inversion at C-5 and without allylic rearrangement giving (−)-**20** in 98% yield. Ozonolysis of the chloroalkene moiety of (−)-**20**, followed by workup with Me_2S in MeOH, furnished the methyl taluronate (−)-**21** (69% yield). This pyranoside exists predominantly as the α-anomer as confirmed by its ^{13}C NMR spectrum showing δ(C-1) = 92.8 ppm (^1J(C,H) = 172 Hz).[23] Silylation of (−)-**21** with (i-Pr)$_3$SiOSO$_2$CF$_3$ and 2,6-lutidine provided (−)-**22** (100%), the β-pyranoside structure being established by the 2D NOESY ^{1}H NMR spectrum of **25** (see below, Figure 3). The reduction of azide (−)-**22** (H_2, PtO_2) led to **23** that was acetylated into (−)-**24** (85%, 2 steps). Reduction of the uronic ester (−)-**24** with $LiBH_4$ in THF at 0 °C generated **25** that was acetylated to afford (−)-**26** (76%, two steps). NOE's observed between signals assigned to H-1 (4.87 ppm), H-3 (2.15−2.02 ppm), and H-5 (3.83 ppm) in the 2D NOESY ^{1}H NMR spectrum of (−)-**26** confirmed the β-D-*talo*pyranoside structure. Desilylation of (−)-**26** gave (+)-**27**, the α-D-pyranose configuration being given by δ(C-1) = 93.4 ppm (^1J(C,H) = 171 Hz).[23] Ammonolysis of (+)-**27** in MeOH delivered the unprotected C-disaccharide as a mixture of **2** and furanoses **28** that was used as such in the enzymatic assays (see below). With the hope of increasing the overall yield in **2** + **28**, we explored the possibility to use the (*tert*-butyl)dimethylsilyl β-pyranoside (−)-**29** (instead of (−)-**22**) that was obtained on treating (−)-**21** with (*t*-Bu)Me$_2$SiOSO$_2$CF$_3$ and 2,6-lutidine (75% yield). The β-D-*talo*-pyranoside structure of (−)-**29** was given by NOE's observed between the signals assigned to H-1 (4.94 ppm) and H-5 (4.20−4.10 ppm) in its 2D NOESY ^{1}H NMR spectrum. Reduction of azide (−)-**29** (H_2/PtO_2 (cat.), EtOAc), followed by acetylation gave (−)-**30** (89%). Unfortunately, our attempts to convert the uronic methyl ester into the corresponding talose derivative all failed, perhaps because of the lesser stability of the (*tert*-butyl)dimethylsilyl β-pyranoside (−)-**29** compared with that of (−)-**22**.

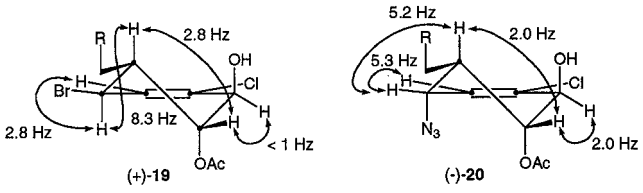

Figure 1. Average conformations of (+)-**19** and (−)-**20** as given by the ^3J(H,H) coupling constants.

Conformation of the C(1→3)-Disaccharides. Because the reducing sugar **1** and **2** are mixtures of α/β-pyranoses and α/β-furanoses their conformational study

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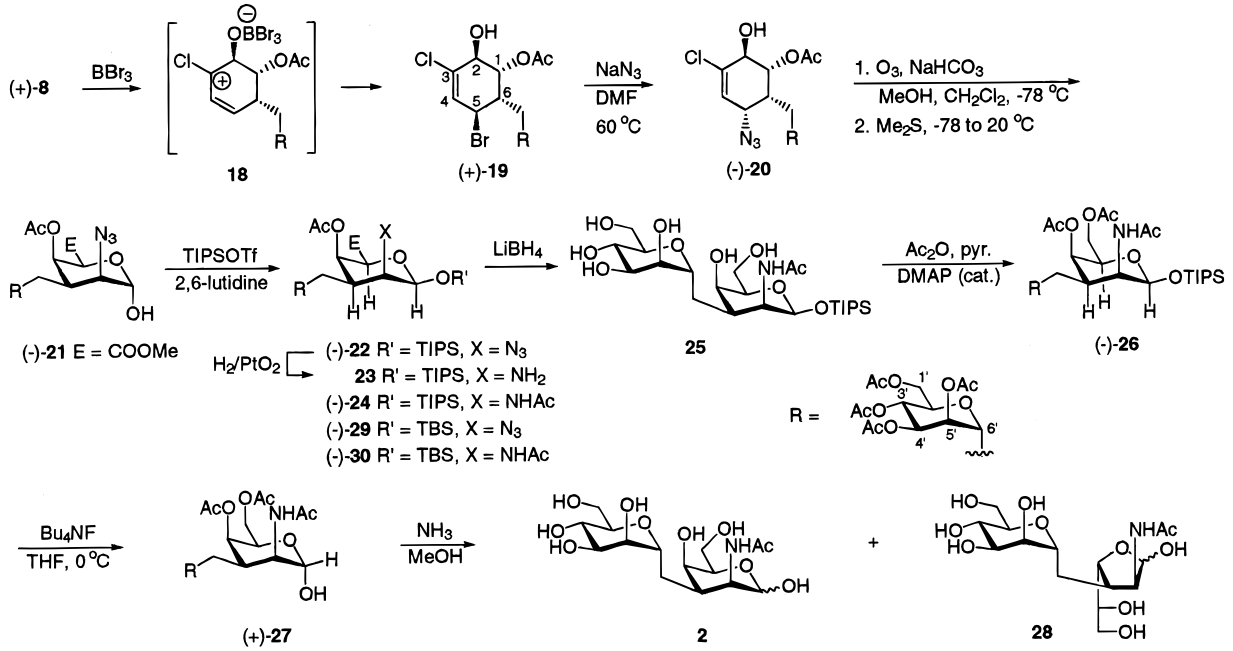
Scheme 2



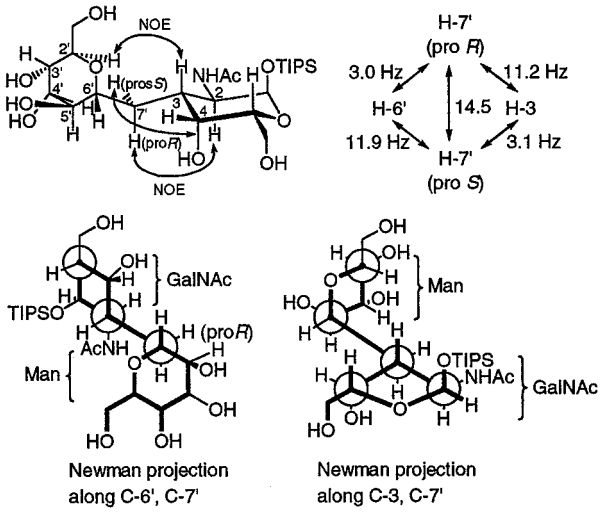


Figure 2. Preferred conformation of **14** (σ(C-5′,C-6′) and σ(C-7′,C-3) are antiperiplanar; σ(C-6′,C-7′) and σ(C-3,C-2) are antiperiplanar).

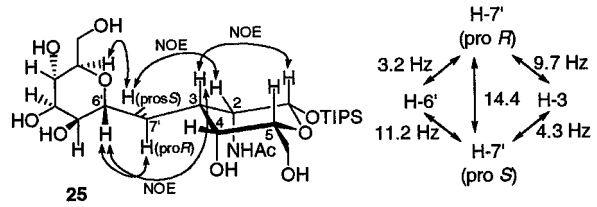


Figure 3. Preferred conformation of **25** (σ(C-5′,C-6′) and σ(C-7′,C-3) are antiperiplanar; σ(C-6′,C-7′) and σ(C-3,C-2) are antiperiplanar) with indication of the most significant NOE's observed in the 2D NOESY 600 MHz ^{1}H NMR spectrum.

by ^{1}H NMR could not be carried out. The coupling constants for the vicinal proton pairs (see Experimental Section) of ^{1}H NMR spectrum (600 MHz, CD_3OD) of the tris(isopropyl)silyl α-galactopyranoside **14** and NOE's (2D NOESY ^{1}H NMR and selective irradiation experiments) confirmed that both the C-mannopyranoside and the galactopyranoside units adopt chair conformations. The methano linker displayed one signal at 1.61 ppm that couples with a large constant (11.2 Hz) with H-3 and a small coupling constant with H-6′ (3.0 Hz), and the second signal at 1.84 ppm that couples with a large coupling constant with H-6′ (11.9 Hz) and a small coupling constant with H-3 (3.1 Hz). Together with the 2D NOESY ^{1}H NMR spectrum of **14** (NOE's between H-2 (4.18 ppm) and both protons H-7′ (1.84, 1.61 ppm), between H-2 (3.59 ppm) and H-3 (2.19 ppm)), these data are consistent with the conformation represented in Figure 2. The 600 MHz ^{1}H NMR spectrum **14** did not change between −40 and +60 °C, suggesting that the conformer of Figure 2 is significantly more stable than other ones.

The coupling constants for the vicinal proton pairs (see Experimental Section) in the ^{1}H NMR spectrum (600 MHz, C_5D_5N) of **25** and NOE's (2D NOESY ^{1}H NMR, and single irradiation experiments) confirmed that both the α-C-mannopyranoside and the β-talopyranoside units adopt chair conformations. The methano linker CH_2 displayed one signal at δ_H 2.43 ppm assigned to pro*S* H-7′, that couples with H-3 (^3J = 4.3 Hz) and with H-6′ (^3J = 11.2 Hz), and a signal at δ_H 2.54 ppm assigned to pro*R* H-7′ that couples with H-3 (9.7 Hz) and with H-6′ (3.2 Hz). This pattern is very similar to that observed in the ^{1}H NMR spectrum of **14**, suggesting therefore similar conformations for **14** (Figure 2) and **25** (Figure 3). This was confirmed by the 2D NOESY ^{1}H NMR spectrum of **25** that showed cross-peaks for proton pairs H-1 (5.10 ppm) and H-3 (2.63−2.59 ppm), H-2 (4.70 ppm) and pro*S* H-7′ (2.43 ppm), H-3 (2.63−2.59 ppm) and H-6′ (4.85 ppm), pro*R* H-7′ (2.54 ppm) and H-6′ (4.85 ppm), H-2′ (4.28 ppm) and pro*S* H-7′ (2.43 ppm).

The ^{1}H NMR spectrum of **25** (C_5D_5N, 600 MHz) did not vary between 20 and 105 °C, suggesting that the conformation shown in Figure 3 is more stable than alternative ones. As for a large number of C-linked disaccharides,[17,26] the α-C-mannopyranosides **14** and **25** adopt conformations in which the *exo*-C−C glycosidic bond σ(C-3,C-7′) is antiperiplanar with the σ(C-5′,C-6′) bond, not with the σ(O−C-6′) bond. The preferred orien-

tation of the GalNAc and TalNAc moieties in which the σ(C-2,C-3) and σ(C-6′,C-7′) bonds are antiperiplanar is not the same than that found for α-D-Gal*p*-(1→3)CH_2-α-D-Man*p*-OMe for which the rotamer with σ(C-3,C-4) and σ(C-6′,C-7′) antiperiplanar is preferred.[17]

Glycosidase Inhibition. The C-disaccharides **1** and **2** were tested for their inhibitory activity toward two α-L-fucosidases (from bovine epididymis, human placenta), three α-galactosidases (from coffee bean, *Aspergillus niger*, *Escherichia coli*), five *β*-galactosidases (bovine liver, *E. coli*, *A. niger*, *A. orizae*, jack bean), three α-glucosidases (maltases from yeast, from rice; isomaltase from baker yeast), two amyloglucosidases (from *A. niger*, *Rhizopus* mold), two *β*-glucosidases (from almond, *Caldocellum saccharolyticum*), two α-mannosidases (from jack bean, almond), one *β*-mannosidase (from *Helix pomatia*), one *β*-xylosidase (from *A. niger*), one α-*N*-acetylgalactosaminidase (from chicken liver), three *β*-*N*-acetyl-glucosaminidases (from jack bean, bovine epididymis A and B). At 1 mM concentration of **1** and **2** and under optimal pH conditions,[27] no inhibition was observed for **2** whereas **1** inhibited some of the enzymes tested as shown in Table 1. *N*-Acetylgalactosamine did not inhibit the enzymes tested. We verified that CH_3CONH_2, byproduct of the ammonolysis, did not inhibit the enzymes tested as well.

The fact that the two α-mannosidases tested ignore both **1** and **2** indicates that these enzymes recognize different disaccharides than α-D-Man*p* (1→3)-pyranosides. The *N*-acetylgalactosamine moiety of **1** allows this C-disaccharide to be recognized by the three α-galactosidases and by one of the five *β*-galactosidases tested. The highest inhibitory activity has been observed for the *β*-galactosidase from jack bean (K_i = 7.5 μM) for which the Lineweaver and Burk plots showed a mixed type (competitive, noncompetitive) of inhibition. It is probably for the same reason that two out of the three *β*-*N*-acetylglucosaminidases tested are inhibited moderately by **1**. The inhibition of the α-glucosidases from yeast and from bakers' yeasts are not explained readily. It appears that these enzymes are not very restrictive in the structural requirements of their sugar substrates. The rather good inhibitory activity of **1** observed for the α-L-fucosidases from bovine epididymis and human placenta is a surprise. These enzymes are known to be inhibited by 1-deoxymannonojirimycin with IC_{50} = 22 μM[28] and K_i = 6.6 μM,[29] respectively. This suggests that mimetics of D-mannopyranose are recognized by them. It is thus tempting to assign the inhibitory activity of **1** toward these enzymes to its α-C-mannopyranoside moiety. If this should be the explanation, we do not understand then why **2**, which possesses also a α-C-mannopyranoside unit, is not an inhibitor of the same enzymes!

Table 1. Inhibition Rate (in %) in the Presence of 1 mM of 1 for the Glycosidase-Catalyzed Hydrolysis of the Corresponding *p*-NO_2-C_6H_4-glycoside at 37 °C, IC_{50} and K_i in μM Values

enzyme	buffers	inhibn rate	IC_{50}	K_i
α-L-fucosidase EC 3.2.1.51				
bovine epididymis	*a*) pH 6	95	19	25
human placenta	*a*) pH 6	94	70	28
α-galactosidase EC 3.2.1.22				
coffee beans	*a*) pH 6	87	195	66
Aspergillus niger	*b*) pH 4	98	162	76
Escherichia coli	a) pH 7	99	68	39
***β*-galactosidase EC 3.2.1.23**				
Escherichia coli	*a*) pH 7	86	500	–
bovine liver	*a*) pH 7	34	–	–
Aspergillus niger	*b*) pH 4	–	–	–
Aspergillus orizae	*b*) pH 4	–	–	–
jack beans	*b*) pH 4	92	9.4	7.5
α-glucosidase EC 3.2.1.20				
yeast	*a*) pH 7	97	38	40
rice	*b*) pH 4	–	–	–
α-glucosidase EC 3.2.1.10				
bakers' yeasts	*a*) pH 7	98	34	38
amyloglucosidase EC 3.2.1.3				
Aspergillus niger	*a*) pH 5	–	–	–
Rhizopus mold	*a*) pH 5	–	–	–
***β*-glucosidase EC 3.2.1.21**				
almonds	*a*) pH 5	–	–	–
caldocellum saccharolyticum	*a*) pH 5	72[c])	45	18
α-mannosidase EC 3.2.1.24				
jack beans	*a*) pH 5	–	–	–
almonds	*a*) pH 5	–	–	–
***β*-mannosidase EC 3.2.1.25**				
Helix pomatia	*b*) pH 4	–	–	–
***β*-xylosidase EC 3.2.1.37**				
Aspergillus niger	*a*) pH 5	–	–	–
α-*N*-acetylgalactosaminidase EC 3.2.1.49				
chicken liver	*b*) pH 4	–	–	–
***β*-*N*-acetylglucosaminidase EC 3.2.1.30**				
jack bean	*a*) pH 5	–	–	–
bovine epididymis A	*b*) pH 4	99	130	135
bovine epididymis B	*b*) pH 4	99	55	102

[a] 0.1 M Na phosphate/citrate. [b] 0.1 M Na citrate. [c] At 0.5 mM.

Glycosyltransferase Inhibition Assays. Many glycosidase inhibitors can behave as inhibitors of glycosyltransferases. This was shown for fucosyltransferases which are inhibited moderately by some fucosidase inhibitors such as 1-deoxy-L-fuconojirimycin and homofuconojirimycin.[30−32] Some inhibitors of glycosyltransferases based on acceptor substrates have been developed, but their potency is generally low with K_i in the millimolar range.[30,31] Fucosyltransferase inhibitors prepared by Jefferies and Bowen[32] require GDP to be fully active. They are composed of 1-deoxy-L-fucononojirimycin linked to D-galactose, a disaccharide mimetic related to the transition state of the reaction. The group of Hindsgaul[33] has prepared a bisubstrate analogue composed of a phenyl *β*-D-galactoside linked through a flexible ethylene bridge to GDP, an inhibitor of α-1,2-fucosyltransferase. Van Boom and co-workers[34] have designed trisub-

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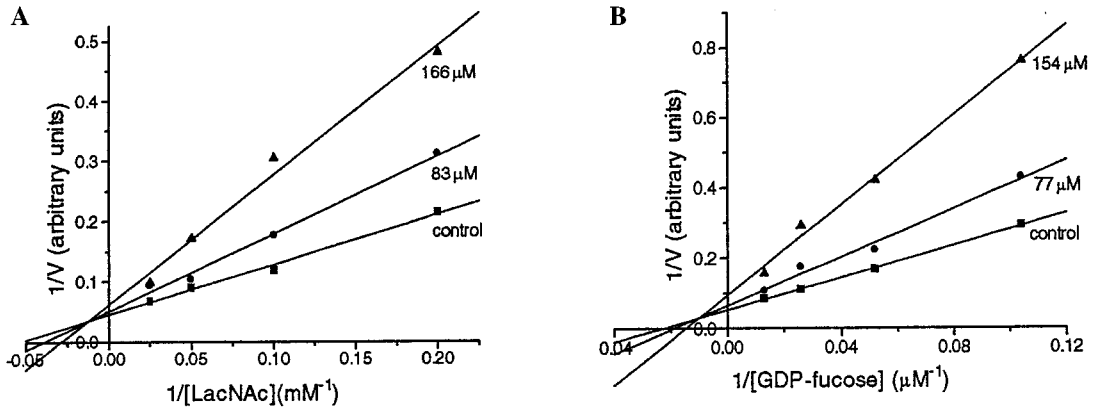

Figure 4. (A) Effect of LacNac concentration on Fuc-TVI inhibition at various concentrations of α-D-Man*p*(1→3)CH_2-D-GalNAc (**1**). Measurments were performed with 100 μM GDP-fuc at 37 °C, pH 6.2; (■) control without **1**, (●) 83 μM of **1**, (▲) 166 μM of **1**. (B) Effect of GDP-fucose concentration on Fuc-TVI inhibition at various concentrations of **1**. Measurements were performed with 20 mM LacNac at 37 °C, pH 6.2; (■) control without **1**, (●) 77 μM of **1**, (▲) 154 μM of **1**.

strate analogues containing D-glucose or D-*N*-acetylglucosamine linked to a GDP-fucose derivative. The most effective inhibitors of glycosyltransferases are often donor substrate analogues such as fluorinated sugar nucleotides.[35] These inhibitors are not expected to be specific because they lack information of the substrate acceptor (type of carbohydrate, position at which glycosylation occurs). We have found that **1** inhibits human α-1,3-fucosyltransferase VI but it is ignored by human α-2,6-sialyltransferase and by β-1,4-galactosyltransferase from human milk. We verified that acetamide, a possible impurity contaminating **1**, did not inhibit human α-1,3-fucosyltransferase VI. Epimer **2** had no inhibitory activity toward these enzymes at 2 mM concentration (see Experimental Section).

With human α-1,3-fucosyltransferase VI (Fuc-TVI) **1** was found to be an effective inhibitor with IC_{50} of 71 μM. Double reciprocal analysis showed that the inhibition displayed a mixed pattern with respect to both the donor sugar GDP-fucose and the acceptor substrate LacNAc with K_i of 123 μM and 128 μM, respectively (see Figure 4).

As the affinity of Fuc-TVI for its substrate LacNAc is low (K_m = 32 mM), α-D-Man*p*-(1→3)CH_2-D-GalNAc (**1**) represents one of the most powerful inhibitors of Fuc-TVI which is not an analogue of the donor substrate. As reported for other fucosyl transferases,[30] the product GDP is a competitive inhibitor of the reaction with a K_i of 120 μM. A synergistic inhibition of fucosyltransferases can be observed with iminosugar inhibitors in the presence of GDP, and this synergy increases considerably the power of the inhibition.[30,36] With **1** we did not observe such a synergistic inhibition of Fuc-TVI with GDP but, on the contrary, preincubation of the enzyme with the sugar nucleotide decreased the power of inhibition. This suggests that the two inhibitors compete for binding to the site of the enzyme.

No inhibition of Fuc-TVI was detected for *N*-acetylgalactosamine and for D-mannose at 5 mM concentrations. This suggests that both sugar moieties of the C-disaccharide **1** are necessary for its inhibitory activity of Fuc-TVI. As **1** is a mixture of α- and β-furanoses, we do not know which form is selected by the enzyme. No effect on the inhibition rate was detected on changing the preincubation time. Among the several mechanisms responsible of this inhibition, one is to invoke that **1** mimics the α-L-Fuc-(1→3)-D-GlcNAc portion of the Lewis x trisaccharide, the D-mannose replacing the fucose residue on one hand, and D-GalNAc, the D-GlcNAc moiety, on the other hand. The fact that GDP-mannose inhibits fucosyltransferases[36] and that 1-deoxymannonojirimycin can inhibit α-L-fucopyranoside[28,37] suggests that the C-α-D-mannopyranoside moiety of **1** can fit into the L-fucopyranoside binding site of the enzyme. The D-GalNAc moiety of **1** would thus take the place reserved for the D-GlcNAc portion of Lewis x trisaccharide in the Fuc-TVI active site, the enzyme permitting one "stereochemical mistake" for that sugar (D-GalNAc is the 4-epimer of D-GlcNAc). This hypothesis is consistent with the observed mixed pattern inhibition. Compound **1** is the first example of C-disaccharide to have glycosyltransferase inhibitory activity.

Conclusion

Efficient and stereoselective syntheses of two new C-disaccharides, α-D-Man*p*-(1→3)CH_2-D-GalNAc (**1**) and α-D-Man*p*-(1→3)CH_2-D-TalNAc (**2**), have been achieved applying the "naked sugar" methodology[16−18] and the Giese's radical C-glycosidation.[15] Whereas **2** did not inhibit any of the 25 glycosidases and 3 glycosyltransferases tested, **1** is a good inhibitor of β-galactosidase from jack beans, (K_i = 7.5 μM), of α-fucosidases from bovine epididymis (K_i = 25 μM) and from human placenta (K_i = 28 μM) and of human α-1,3-fucosyltransferase VI.[38] Given the low affinity of Fuc-TVI for its substrate (K_m = 32 mM) and the fact that it does not require nucleotide sugar GDP to inhibit significantly the enzyme, the neutral C-disaccharide **1** represents a compound of great

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interest. Further work aimed at assaying **1** on recombinant Fuc-TVII, an enzyme of crucial importance in the biosynthesis of selective ligands involved in early inflammatory response,[39] will be undertaken. Analogues of **1** and **2** deserve to be prepared and tested toward glycosidases and glycosyltransferases.[40]

Experimental Section

General Remarks. See ref 41. None of the procedures were optimized. Flash column chromatography (FC) was performed on Merck silica gel (230−400 mesh).[42] Thin-layer chromatography (TLC) was carried out on silica gel (Merck aluminum foils). ^{1}H NMR signals assignments were confirmed by double irradiation experiments and, when required, by 2D NOESY and COSY spectra. *J* values are given in hertz. 600 MHz ^{1}H NMR spectra were recorded on a Bruker AMX-600 FR spectrometer.

2-Acetamido-2,3-dideoxy-3-*C*-(2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-D-galactose (α-C(1→3)-D-mannopyranoside of *N*-Acetyl D-galactosamine: 1 + 17). A mixture of (+)-**16** (29 mg, 0.05 mmol) and MeOH saturated with gaseous NH_3 (3 mL) was stirred at 20 °C for 9.5 h. The solvent was evaporated to dryness in vacuo and the residue purified by column chromatography on Sep-Pak RP18-2gr H_2O as eluent. Yield: 18 mg (76%) of **1**, yellowish oil, contains a little of acetamide. IR (KBr) ν 3385, 1660, 1385, 1065. ^{1}H NMR (400 MHz, CD_3OD: mixture of α,β-D-pyranose and α,β-D-furanose) δ 5.18 (d, ^3J = 4.4), 5.17 (d, ^3J = 2.1), 5.06 (d, ^3J = 3.6), 4.57 (d, ^3J = 8.3, 4H-C(1)); 3.40−4.25 (m, 48H), 2.49−2.58 (m), 3.29−2.37 (m), 2.26−2.05 (m), 1.90−1.77 (m), 1.67−1.54 (m), 2.05−1.97 (5s, NHAc + NH_2Ac). ^{13}C NMR (101 MHz, CD_3OD) δ 176.4, 173.8, 173.5, 172.8 (4s, 4C═O NHAc + C═O NH_2Ac), 102.9, 98.9, 95.1, 92.2 (4d, 4C(1)), 83.9, 83.7, 79.9, 76.4, 76.3, 75.8, 75.7, 74.6, 74.5, 73.5, 73.1, 73.0 (*2), 72.8, 72.6, 72.3, 69.5, 69.3, 66.2, 65.6 (all *d*), 64.7, 63.3, 63.2 (*2), 63.0 (all *t*, 4C(1′) + 4 C(6)), 59.2, 53.1, 49.8 (*2), 43.1, 41.6, 37.6, 36.0 (all *d*), 32.4, 31.5, 26.3, 26.2 (all *t*, 4 C(7′)), 22.9, 22.8, 22.7, 22.6, 22.1 (q, 4CH_3 + CH_3CONH_2). ESI + QIMS: $[M + 1]^+$= 382.3.

2-Acetamido-2,3-dideoxy-3-*C*-(2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-D-talose (α-C(1→3)-D-mannopyranoside of *N*-Acetyl D-talosamine: 2 + 28). A mixture of (+)-**27** (25 mg, 0.04 mmol) and MeOH saturated with gaseous NH_3 (3 mL) was stirred at 20 °C for 7.5 h. The solvent was evaporated and the residue purified by column chromatography on Sep-Pak RP18-2gr, with H_2O as eluent, yielding 17 mg (92%) of a mixture of pyranose **2** and furanose **28**, contaminated with less than 20% of acetamide, yellowish oil. IR (KBr) ν 3355, 1660, 1400, 1065. ^{1}H NMR (400 MHz, CD_3OD) δ 5.36 (d, ^3J = 4.6), 5.05 (br s), 5.01 (br s), 4.90−4.80 (br s, 4H-C(1) + H_2O)), 4.57 (d, ^3J = 8.2), 4.47 (dd, ^3J = 9.1, 4.6), 4.36 (d, ^3J = 6.0), 4.20−4.05 (m), 3.94−3.56 (m), 3.50−3.43 (m, 48H), 2.08, 2.05, 2.03, 2.01 (4s, NHAc), 1.98 (s, NH_2-Ac), 2.88−2.80 (m), 1.87−1.70 (m), 1.67−1.58 (m). ^{13}C NMR (101 MHz, CD_3OD) δ 176.4, 173.8, 173.5, 172.7 (4s, 4C═O NHAc + C═O NH_2Ac), 101.8, 97.6, 97.1, 94.4 (4d, 4C(1)), 84.2, 83.9, 83.6, 80.6, 76.9, 76.6, 76.2, 76.0, 75.8, 75.3, 75.0, 74.2, 73.3, 72.9, 72.8, 72.6, 72.5, 72.3, 69.6, 69.5, 69.4, 67.2, 65.9 (all *d*), 65.9, 65.0, 64.5, 63.5, 63.2, 63.1, 62.9 (all t, 4C(1′) + 4C(6)), 60.0, 59.2, 55.4, 53.4, 52.8, 38.5, 36.8, 36.4, 32.7 (all *d*), 29.4, 28.0, 27.9, 27.2 (all *t*, 4C(7′)), 23.3, 23.2, 22.5, 22.0 (q, 4CH_3 + CH_3CONH_2). ESI + QIMS: $[M + 1]^+$= 382.3.

(+)-(1*S*,3*R*,4*R*,5*R*,6*R*)-6-*endo*-Chloro-5-*exo*-(phenylseleno)-3-*endo*-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-7-oxabicyclo-[2.2.1]heptan-2-one ((+)-5). A mixture of (−)-**4**[16,17] (200 mg, 0.64 mmol), acetobromomannose (2,3,4,6-tetra-*O*-acetyl-α-D-manno pyranosyl bromide 341 mg, 0.82 mmol), and AIBN (10.5 mg, 0.1 mmol) in anhydrous benzene (4 mL) was heated under reflux for 20 min. A solution of Ph_3SnH (225 mg, 0.64 mmol) and AIBN (10.5 mg, 0.1 mmol) in anhydrous benzene (2 mL) was added via a syringe in about 30 min. The mixture was then heated under reflux for 60 min and allowed to cool to 20 °C. KF (270 mg) was added, and the mixture was stirred at 20 °C for 14 h. After filtration through Celite (EtOAc as eluent) and solvent evaporation in vacuo, the residue was purified by FC (EtOAc/light petroleum ether 2:3), yielding 231 mg (56%) of (+)-**5**, white foam. $[\alpha]^{25}_D$ = 5.8 (*c* = 1.0, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.64−7.61 (m), 7.32−7.28 (m), 5.23 (dd, ^3J = 4.8, 3.2), 4.95 (d, ^3J = 5.6), 4.91 (dd, ^3J = 8.5, 3.2), 4.85 (dd, ^3J = 4.8, 2.7), 4.56 (dd, ^2J = 12.2, ^3J = 9.1), 4.48 (d, ^3J = 5.9), 4.32 (dd, ^3J = 5.9, 3.1), 4.05 (dd, ^2J = 12.2, ^3J = 3.9), 3.98 (ddd, ^3J = 10.3, 8.5, 2.0), 3.69 (d, ^3J = 3.1), 3.61 (ddd, ^3J = 9.1, 3.9, 2.7), 2.68 (ddd, ^3J = 10.5, 5.6, 5.4), 2.16−2.01 (4s + m), 1.47 (ddd, ^2J = 14.8, ^3J = 10.5, 10.3).

(−)-(1*S*,2*R*,3*S*,5*R*,6*R*)-6-*endo*-Chloro-5-*exo*-(phenylseleno)-3-*endo*-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-7-oxabicyclo[2.2.1]-heptan-2-*endo*-ol ((−)-6). $NaBH_4$ (156 mg, 4.13 mmol) was added to a solution of (+)-**5** (940 mg, 1.45 mmol) in THF/MeOH 1:1 (40 mL) cooled to 0 °C. After stirring at 0 °C for 5 min, the mixture was neutralized with 1 N HCl (if the pH is < 7, saturated aqueous $NaHCO_3$ solution is added until neutral pH). Then a part of methanol was evaporated under *vacuum*, and the aqueous phase was extracted with CH_2Cl_2. The organic extract was washed with brine (twice) and dried ($MgSO_4$), and the solvent was evaporated in vacuo. Residue was purified by FC (Et_2O/light petroleum ether 9:1) yielding 742 mg (79%) of (−)-**6**, white foam. $[\alpha]^{25}_D$ = −7.0 (*c* = 1.0, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.65−7.63 (m), 7.34−7.27 (m), 5.25 (dd, ^3J = 7.1, 3.3), 5.09 (dd, ^3J = 5.3, 3.3), 5.05 (dd, ^3J = 7.1, 5.9), 4.61 (d, ^3J = 5.5), 4.50−4.43 (m), 4.33−4.30 (m), 4.10 (dd, ^2J = 12.1, ^3J = 3.4), 4.00 (ddd, ^3J = 9.3, 5.3, 5.0), 3.77 (ddd, ^3J = 7.2, 5.9, 3.4), 3.65 (d, ^3J = 4.9), 2.45−2.36 (m), 2.10−2.00 (4s + m), 1.47 (ddd, ^2J = 14.4,^3J = 9.3 6.7).

(+)-(1*S*,2*R*,3*S*,4*R*)-6-Chloro-3-*endo*-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-7-oxabicyclo[2.2.1]hept-5-en-2-*endo*-ol ((+)-7). A solution of *m*-CPBA (70% 3-$ClC_6H_4CO_3H$ in 3-$ClC_6H_4CO_2H$, 167 mg, 0.68 mmol) in anhydrous CH_2Cl_2 (3.3 mL) was added dropwise to a stirred solution of (−)-**6** (400 mg, 0.62 mmol) in anhydrous CH_2Cl_2 (10.3 mL) cooled to −78 °C. The mixture was stirred at −78 °C for 3 h, allowed to warm to 20 °C, and stirred for 12 h. CH_2Cl_2 (20 mL) was added, and the solution was washed with saturated aqueous solution of $NaHCO_3$ (20 mL, twice). The aqueous phase was extracted with CH_2Cl_2 (20 mL, four times). The combined organic phases were washed with brine (20 mL, twice) and dried ($MgSO_4$). After solvent evaporation in vacuo, the residue was purified by FC (Et_2O/light petroleum ether 9:1), yielding 314 mg (94%) of (+)-**7**, white foam. $[\alpha]^{25}_D$ = 5.4 (*c* = 1.0, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 6.34 (d, ^3J = 1.9), 5.25−5.15 (m), 4.90 (dd, ^3J = 4.4, 1.9), 4.73 (d, ^3J = 4.4), 4.63 (ddd, ^3J = 7.7, 6.0, 4.4), 4.42 (dd, ^2J = 12.1, ^3J = 6.7), 4.11−4.03 (m), 3.92 (ddd, ^3J = 7.1, 6.7, 2.8), 2.78 (d, ^3J = 6.0), 2.30−2.23 (m), 2.13−2.05 (4s), 1.83 (ddd, ^2J = 14.4, ^3J = 7.1, 6.9), 1.39 (ddd, ^2J = 14.4, ^3J = 7.7, 5.2).

(+)-(1*S*,2*R*,3*S*,4*R*)-6-Chloro-3-*endo*-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-7-oxabicyclo[2.2.1]hept-5-en-2-*endo*-yl Acetate ((+)-8). A mixture of (+)-**7** (500 mg, 1.02 mmol), anhydrous pyridine (5.4 mL), acetic anhydride (482 μL), and a catalytic amount of 4-(dimethylamino)pyridine (8 mg) was stirred at 20 °C for 7 h. The solvent was evaporated in vacuo. Purification by FC (EtOAc/light petroleum ether 3:2) gave 500 mg (92%) of (+)-**8**, white foam. $[\alpha]^{25}_D$ = 24 (*c* = 1.0, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 6.35 (d, ^3J = 2.0), 5.22 (dd, ^3J = 8.2, 4.4), 5.22 (dd, ^3J = 7.3, 3.2), 5.12 (dd, ^3J = 7.4, 7.3), 5.05 (dd, ^3J = 4.5, 3.2),

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5.01 (ddd, ^3J = 4.3, 2.0, ^4J = 0.8), 4.99 (br d, ^3J = 4.4), 4.45 (dd, ^2J = 12.1, ^3J = 7.6), 4.14 (dd, ^2J = 12.1, ^3J = 3.0), 4.02 (ddd, ^3J = 10.5, 4.5, 3.1), 3.91 (ddd, ^3J = 7.6, 7.4, 3.0), 2.53−2.46 (m), 2.11−2.04 (4s), 1.56−1.41 (2m).

(+)-(1*R*,2*S*,5*R*,6*R*)-5-Acetamido-3-chloro-2-hydroxy-6-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)cyclohex-3-en-1-yl Acetate ((+)-10). CF_3SO_3H (90 μL, 1.03 mmol) was added to a solution of (+)-**8** (100 mg, 0.19 mmol) in dry MeCN (4.4 mL) cooled to 0 °C. After stirring at 0 °C for 5 min, the reaction was allowed to warm to 20 °C and stirred for an additional 40 min. The reaction was quenched with ice-cold saturated solution of $NaHCO_3$ and extracted with CH_2Cl_2 (20 mL, four times). The combined organic extracts were dried ($MgSO_4$), and after solvent evaporation in vacuo the residue was purified by FC (EtOAc/light petroleum ether 4:1) gaving 94 mg (84%) of (+)-**10**, white foam. $[\alpha]^{25}_D$ = 16 (*c* = 1.0, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.93 (d, ^3J = 9.5), 5.91 (d, ^3J = 2.5), 5.23 (dd, ^3J = 7.4, 3.1), 5.20 (dd, ^3J = 3.1, 2.2), 5.07−5.03 (m), 4.56 (ddd, ^3J = 11.0, 9.5, 2.5), 4.53 (dd, ^2J = 12.0, ^3J = 7.8), 4.10−4.05 (m), 4.02−3.98 (m), 3.50−3.40 (br d), 2.12−2.04 (5s + m), 1.82 (ddd, ^2J = 14.6, ^3J = 11.4, 2.9), 1.52 (ddd, ^2J = 14.6, ^3J = 10.6, 3.0).

Methyl 2-Acetamido-4-*O*-acetyl-2,3-dideoxy-3-*C*-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-α-D-galactopyranuronate ((+)-11) and Mixture of Methyl 2-Acetamido-5-*O*-acetyl-2,3-dideoxy-3-*C*-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-α-D- and β-D-Galactofuranuronate ((−)-12). $NaHCO_3$ (66 mg, 0.78 mmol) was added to a solution of (+)-**10** (80 mg, 0.14 mmol) in MeOH/CH_2Cl_2 1:1 (4.8 mL). The solution was cooled to −78 °C, and ozone was bubbled through it until starting material has disappeared by TLC (10−20 min). A stream of N_2 was bubbled for 10 min, and a few drops of Me_2S were added. After 10 min the mixture was allowed to warm to 20 °C and was stirred for 6 h. H_2O (10 mL) was added, and the aqueous phase was extracted with CH_2Cl_2 (15 mL, four times). The combined organic extracts were dried ($MgSO_4$), and after solvent evaporation in vacuo a 1.4:1 mixture of (+)-**11** and (−)-**12** was obtained. They were separated by FC (MeOH/AcOEt/CH_2Cl_2: 30:485:485), yielding 20 mg (23%) of (−)-**12** (a 1.7:1 mixture of anomers) and 27 mg (31%) of (+)-**11**.

Data for (+)-11: white foam. $[\alpha]^{25}_D$ = 50 (*c* = 0.5, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.81 (d, ^3J = 9.8), 5.57 (br s), 5.36 (d, ^3J = 3.0), 5.22 (dd, ^3J = 7.2, 3.3), 5.04 (dd, ^3J = 7.2, 7.1), 5.00 (dd, ^3J = 4.9, 3.3), 4.75 (br s), 4.48 (dd, ^2J = 12.0, ^3J = 8.3), 4.32 (ddd, ^3J = 9.9, 9.8, 3.0), 4.17 (dd, ^3J = 12.0, 4.9), 4.14−4.10 (m), 4.01−3.96 (m), 3.74 (s), 3.70−3.60 (br s), 2.37−2.29 (dd, ^3J = 9.9, 9.7), 2.14−2.04 (6s), 1.80−1.60 (m), 1.40−1.25 (m).

Data for (−)-12: white foam. $[\alpha]^{25}_D$ = −30 (*c* = 0.5, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ (1.7:1 mixture of anomers) 6.16, 6.07 (2s), 5.89 (d, ^3J = 7.1), 5.85 (d, ^3J = 7.8), 5.25−5.04 (2m), 4.35−4.03 (m), 3.84 (ddd, ^3J = 7.5, 7.4, 3.0), 3.74−3.69 (m), 3.48 (s), 3.44 (s), 2.75−2.67 (m), 2.20−2.05 (m), 1.89 (ddd, ^2J = 14.8, ^3J = 14.7, 4.3), 1.82−1.60 (m).

Methyl 2-Acetamido-4-*O*-acetyl-2,3-dideoxy-3-*C*-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-1-*O*-tris(isopropyl)silyl-α-D- and β-D-Galactopyranuronate ((+)-13, α/β 1:0.45). 2,6-Lutidine (38 μL, 0.33 mmol) and TIPSOTf (59 μL, 0.22 mmol) were added to a solution of (+)-**11** (68 mg, 0.11 mmol) in CH_2Cl_2 (5.4 mL) cooled at 0 °C under N_2. After stirring at 0 °C for 5 min, the reaction was allowed to warm to 20 °C and stirred for 1.5 h; by TLC there was still starting material so the reaction was cooled again to 0 °C, and 2,6-lutidine (38 μL, 0.33 mmol) and TIPSOTf (59 μL, 0.22 mmol) were added. The reaction was allowed to warm to 20 °C and stirred for 2 h more. Brine (15 mL) was added, and the aqueous phases were extracted with CH_2Cl_2 (20 mL, four times). After drying ($MgSO_4$) and solvent evaporation in vacuo, the residue was purified by FC (EtOAc/light petroleum ether 3:2), yielding 16 mg of β-**13** and 40 mg (59%) of α-**13**. Pure β-**13** was obtained by FC (Et_2O).

Data for (+)-α-13: white solid, mp 152−153 °C. $[\alpha]^{25}_D$ = 43 (*c* = 0.5, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.55 (br s), 5.52 (d, ^3J = 10.0), 5.35 (d, ^3J = 3.3), 5.25 (dd, ^3J = 5.9, 3.1), 5.02 (dd, ^3J = 5.9, 4.3), 4.92 (dd, ^3J = 6.9, 3.1), 4.66 (br s), 4.61 (dd, ^2J = 12.1, ^3J = 7.7), 4.34 (ddd, ^3J = 12.0, ^3J = 10.0, 3.3), 4.18 (dd, ^2J = 12.1, ^3J = 4.4), 4.10 (ddd, ^3J = 10.8, 6.9, 2.4), 3.98 (ddd, ^3J = 7.7, 4.4, 4.3), 3.72 (s), 2.32 (dddd, ^3J = 12.0, 11.4, 3.0, 2.9), 2.11−1.99 (6s), 1.65 (ddd, ^2J = 14.2, ^3J = 10.8, 3.0), 1.45 (ddd, ^2J = 14.2, ^3J = 11.4, 2.4), 1.17−1.06 (m).

Data for β-13: colorless, viscous oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.49 (br s), 5.47 (d, ^3J = 9.0), 5.20 (dd, ^3J = 7.5, 3.4), 5.09 (dd, ^3J = 7.5, 6.7), 5.01 (dd, ^3J = 4.1, 3.4), 4.89 (*d*, ^3J = 7.8), 4.46 (dd, ^2J = 12.1, ^3J = 6.6), 4.21 (d, ^3J = 1.2), 4.16 (dd, ^2J = 12.1, ^3J = 4.0), 4.11 (ddd, ^3J = 12.3, 4.1, 3.0), 3.89 (ddd, ^3J = 6.7, 6.6, 4.0), 3.70 (s), 3.64 (ddd, ^3J = 12.1, 9.0, 7.8), 2.43 (dddd, ^3J = 12.1, 11.3, 2.6, 2.5), 2.13−1.99 (6s), 1.85 (ddd, ^2J = 14.5, ^3J = 12.3, 2.6), 1.36 (ddd, ^2J = 14.5, ^3J = 11.3, 3.0), 1.29−1.07 (m).

Tris(isopropyl)silyl 2-Acetamido-2,3-dideoxy-3-*C*-(2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-α-D-galactopyranoside (14). A 2 M solution of $LiBH_4$ in anhydrous THF (135 μL, 0.27 mmol) was added slowly to a stirred solution of (+)-α-**13** (28 mg, 36 μmol) in anhydrous THF (3.8 mL) cooled to 0 °C. After stirring at 0 °C for 5 min, the mixture was allowed to warm to 20 °C and stirred for 23 h. H_2O (3 mL) was added under vigorous stirring. The solvent was evaporated in vacuo, and the residue was purified by chromatography on a short column (*h* = 3 cm, ∅ = 1 cm) of Dowex 50WX8 (100−200 mesh, 3.7% aq HCl, then H_2O, then MeOH). Solvent evaporation to dryness yielded 14 mg (72%) of **14** as a viscous oil. ^{1}H NMR (600 MHz, CD_3OD) δ 5.28 (d, ^3J = 3.3, H-C(1)), 4.18 (dd, ^3J = 12.2, 3.3, H-C(2)), 4.09 (ddd, ^3J(Hpro*S*-C(7′)-H-C(6′)) = 11.9, ^3J(Hpro*R*-C(7′)-H-C(6′)) = 3.0, ^3J(H-C(5′)-H-C(6′))= 2.8, H-C(6′)), 3.84 (dd, ^2J = 11.5, ^3J(H-C(1′)-H-C(2′)) = 2.9, H-C(1′)), 4.06−4.03 (m, 2H), 3.81−3.76 (m, 2H), 3.74−3.71 (m, 2H), 3.69−3.64 (m, 2H, H-C(1′), H-C(3′), H-C(4′), H-C(5′), H-C(4), H-C(5), 2H-C(6)), 3.59 (ddd, ^3J = 7.9, 6.7, 2.9, H-C(2′)), 2.19 (dddd, ^3J(H-C(2)−H-C(3)) = 12.2, ^3J(Hpro*R*-C(7′)-H-C(3)) = 11.2, ^3J(Hpro*S*-C(7′)-H-C(3)) = 3.1, ^3J(H-C(4)−H-C(3)) = 2.8, H-C(3)), 2.03 (*s*, NHAc), 1.84 (ddd, ^2J = 14.5, ^3J(Hpro*S*-C(7′)-H-C(6′)) = 11.9, ^3J(Hpro*S*-C(7′)-H-C(3)) = 3.1, Hpro*S*-C(7′)), 1.61 (ddd, ^2J = 14.5, ^3J(Hpro*R*-C(7′)-H-C(3)) = 11.2, ^3J(Hpro*R*-C(7′)-H-C(6′)) = 3.0, Hpro*R*-C(7′)), 1.24−1.15 (*m*, 21H, TIPS). ^{13}C NMR (101 MHz, CD_3OD) δ 173.3 (s, C=O), 97.6 (d, ^1J(C,H) = 168, C(1)), 76.2 (d, ^1J(C,H) = 139), 74.1 (d, ^1J(C,H) = 148), 73.2 (d, ^1J(C,H) = 134), 72.9 (d, ^1J(C,H) = 142), 72.3 (d, ^1J(C,H) = 145), 69.5 (d, ^1J(C,H) = 145), 65.8 (d, ^1J(C,H) = 146), 51.0 (d, ^1J(C,H) = 141), 35.5 (d, ^1J(C,H) = 128), C(2), C(3), C(4), C(5), C(2′), C(3′), C(4′), C(5′), C(6′), 62.9 (t, ^1J(C,H) = 141), 62.4 (t, ^1J(C,H) = 142), C(1′), C(6), 26.9 (t, ^1J(C,H) = 126, C(7′)), 22.9 (q, ^1J(C,H) = 128, NHAc), 18.4, 18.3 (q, ^1J(C,H) = 120, TIPS), 13.5 (d, ^1J(C,H) = 118, TIPS).

Tris(isopropyl)silyl 2-Acetamido-4,6-di-*O*-acetyl-2,3-dideoxy-3-*C*-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-α-D-galactopyranoside ((+)-15). A mixture of **14** (14 mg, 26 μmol), anhydrous pyridine (3.5 mL), Ac_2O (1 mL), and 4-(dimethylamino)pyridine (1 mg) was stirred at 20 °C for 12 h. The solvent was evaporated in vacuo. The residue was purified by FC (EtOAc/light petroleum ether 3:2), yielding 21 mg (96%) of (+)-**15**, colorless oil. $[\alpha]^{25}_D$ = 43 (*c* = 0.5, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.52 (d, ^3J = 10.0), 5.34 (br s), 5.28−5.24 (m), 5.02 (dd, ^3J = 6.9, 4.5), 4.91 (dd, ^3J = 7.0, 3.1), 4.65 (dd, ^2J = 12.1, ^3J = 7.6), 4.30−4.20 (m), 4.20−4.10 (m), 4.00−3.90 (m), 2.26 (dddd, ^3J = 11.6, 11.5, 2.8, 2.7), 2.12−1.99 (7s), 1.65−1.57 (m), 1.40 (dddd, ^2J = 11.7, ^3J = 11.6, 2.4, 2.3), 1.20−1.07 (m).

2-Acetamido-4,6-di-*O*-acetyl-2,3-dideoxy-3-*C*-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-α-D-galactopyranose ((+)-16). A mixture of (+)-**15** (58 mg, 0.07 mmol), THF (3.5 mL), and tetrabutylammonium fluoride (0.11 mL, 0.11 mmol) was stirred at 0 °C under N_2 atmosphere for 5 min. H_2O (6 mL) was added, and the aqueous layer was extracted with Et_2O (6 mL) and then

with CH_2Cl_2 (6 mL, four times). The combined organic extracts were dried ($MgSO_4$), and the solvent was evaporated. The residue was purified by FC (CH_2Cl_2/MeOH 96:4), yielding 34 mg (74%) of (+)-**16**, white foam. $[\alpha]^{25}_D = 37$ ($c = 0.5$, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.82 (d, $^3J = 9.8$), 5.29 (br s), 5.24−5.19 (m), 5.02 (dd, $^3J = 6.3, 6.2$), 4.98 (dd, $^3J = 5.2, 3.3$), 4.48 (dd, $^2J = 12.0$, $^3J = 8.4$), 4.32 (dd, $^3J = 6.5, 6.4$), 4.24 (ddd, $^3J = 12.7, 9.8, 3.5$), 4.19−4.08 (m), 4.02−3.93 (m), 2.24 (dddd, $^2J = 12.7$, $^3J = 12.0, 2.8, 2.2$), 2.14−2.05 (7s), 1.70 (ddd, $^2J = 14.0$, $^3J = 12.0, 2.2$), 1.33 (ddd, $^2J = 14.0$, $^3J = 10.7, 2.2$).

(+)-(1*R*,2*S*,5*R*,6*S*)-5-Bromo-3-chloro-2-hydroxy-6-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)cyclohex-3-en-1-yl Acetate ((+)-19). A 1 M solution of BBr_3 in anhydrous CH_2Cl_2 (1.14 mL, 1.14 mmol) was added to a solution of (+)-**8** (402 mg, 0.76 mmol) in anhydrous CH_2Cl_2 (20 mL) cooled to 0 °C under N_2 atmosphere. After stirring at 0 °C for 15 min, a saturated aqueous solution of $NaHCO_3$ (25 mL) was added and the mixture was extracted with CH_2Cl_2 (25 mL, four times). The combined organic extracts were dried ($MgSO_4$), and the solvent was evaporated in vacuo. The residue was purified by FC (Et_2O/light petroleum ether 7:3) yielding 338 mg (72%) of (+)-**19**, white foam. $[\alpha]^{25}_D = 36$ ($c = 1.0$, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 6.24 (d, $^3J = 2.8$), 5.24 (dd, $^3J = 7.9, 3.3$), 5.24−5.22 (m), 5.16 (dd, $^3J = 7.9, 7.5$), 5.13 (dd, $^3J = 3.4, 3.3$), 4.56 (dd, $^3J = 8.3, 2.8$), 4.44 (dd, $^2J = 12.2$, $^3J = 7.2$), 4.13−4.07 (m), 4.08 (dd, $^2J = 12.2$, $^3J = 2.4$), 3.96 (ddd, $^3J = 7.5, 7.2, 2.4$), 2.95−2.80 (br s), 2.58 (dddd, $^3J = 10.9, 8.3, 2.9, 2.9$), 2.32−2.24 (m), 2.13−2.05 (5s), 1.51 (ddd, $^2J = 14.4$, $^3J = 10.9, 2.8$).

(1*R*,2*S*,5*S*,6*R*)-5-Azido-3-chloro-2-hydroxy-6-(1′,3′,4′,5′-tetra-*O*-acetyl-2,6-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)cyclohex-3-en-1-yl Acetate ((−)-20). A mixture of (+)-**19** (340 mg, 0.55 mmol), DMF (18.3 mL), and NaN_3 (70 mg, 1.1 mmol) was heated in the dark to 60 °C for 15 min. After cooling to 20 °C, H_2O (20 mL) was added. The aqueous phase was extracted with Et_2O (20 mL, 4 times). The combined organic extracts were washed with H_2O (20 mL, twice) and dried ($MgSO_4$). After solvent evaporation, the residue was purified by FC (Et_2O/light petroleum ether 7:3), yielding 310 mg (98%) of (−)-**20**, white foam. $[\alpha]^{25}_D = -173$ ($c = 1.0$, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 6.19 (d, $^3J = 5.3$), 5.26 (dd, $^3J = 7.9, 3.3$), 5.17 (dd, $^3J = 2.2, 2.1$), 5.14 (dd, $^3J = 7.9, 7.0$), 5.09 (dd, $^3J = 4.0, 3.3$), 4.50 (dd, $^2J = 12.2$, $^3J = 6.8$), 4.15−4.08 (m), 4.04 (dd, $^2J = 12.2$, $^3J = 2.6$), 3.97 (ddd, $^3J = 7.0, 6.8, 2.6$), 3.90 (dd, $^3J = 5.3, 5.2$), 2.44−2.39 (m), 2.17−2.07 (5s), 2.05−1.94 (m), 1.69−1.63 (m).

Methyl (4-*O*-Acetyl-2-azido-2,3-dideoxy-3-*C*-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-α-D-talopyranuronate ((−)-21). $NaHCO_3$ (11 mg, 0.13 mmol) was added to a solution of (−)-**20** (120 mg, 0.21 mmol) in MeOH/CH_2Cl_2 1:1 (8 mL). The solution was cooled to −78 °C, and ozone was bubbled through it for 3 h. A stream of N_2 was bubbled for 10 min, and a few drops of Me_2S were added. The mixture was allowed to warm to 20 °C and stirred for 12 h. H_2O (10 mL) was added, and the aqueous phase was extracted with CH_2Cl_2 (10 mL, four times). The combined organic extracts were dried ($MgSO_4$), and after solvent evaporation in vacuo the residue was purified by FC (EtOAc/light petroleum ether 3:2), yielding 88 mg (69%) of (−)-**21**, white foam. $[\alpha]^{25}_D = -13$ ($c = 1.0$, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.62 (d, $^3J = 3.1$), 5.49 (br s), 5.26 (dd, $^3J = 7.0, 3.2$), 5.10 (dd, $^3J = 7.1, 7.0$), 5.05 (dd, $^3J = 4.6, 3.2$), 4.82 (d, $^3J = 1.5$), 4.49 (dd, $^2J = 12.1$, $^3J = 7.2$), 4.19 (ddd, $^3J = 12.1, 4.6, 4.5$), 4.12 (dd, $^2J = 12.1$, $^3J = 2.9$), 3.93 (ddd, $^3J = 7.2, 7.1, 2.9$), 3.74 (s), 3.42 (br d, $^3J \cong 2.9$), 2.60−2.52 (m), 2.12−2.08 (5s), 1.80−1.66 (m).

Methyl 4-*O*-Acetyl-2-azido-2,3-dideoxy-3-*C*-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-1-*O*-(tris(isopropyl)silyl)-β-D-talopyranuronate ((−)-22). A mixture of (−)-**21** (71 mg, 0.12 mmol), CH_2Cl_2 (6 mL), 2,6-lutidine (41 μL, 0.36 mmol), and triisopropylsilyl trifluoromethanesulfonate (TIPSOTf, 65 μL, 0.24 mmol) was stirred at 0 °C under N_2 atmosphere for 5 min and then at 20 °C for 5 h. More 2,6-lutidine (41 μL, 0.36 mmol) and TIPSOTf (65 μL, 0.24 mmol) were added if the reaction was incomplete (control by TLC), and the mixture was stirred at 20 °C for 4 more days. Brine (10 mL) was added, and the aqueous phase was extracted with CH_2Cl_2 (10 mL, 4 times). The combined organic extracts were dried ($MgSO_4$), and the solvent was evaporated. The residue was purified by FC (EtOAc/light petroleum ether 2:3), yielding 95 mg (100%) of (−)-**22**, yellowish oil. $[\alpha]^{25}_D = -51$ ($c = 1.0$, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.37 (br t, $^3J \cong 1.5$), 5.24 (dd, $^3J = 6.5, 3.3$), 5.10 (dd, $^3J = 6.6, 6.5$), 5.01 (dd, $^3J = 5.0, 3.3$), 4.98 (d, $^3J = 1.4$), 4.49 (dd, $^2J = 12.2$, $^3J = 6.5$), 4.18−4.12 (m), 3.92 (ddd, $^3J = 6.6, 6.5, 3.4$), 3.41 (br d, $^3J \cong 2.0$), 2.16−2.08 (m + 5s), 1.76−1.70 (m), 1.21−1.09 (m).

Methyl 2-Acetamido-4-*O*-acetyl-2,3-dideoxy-3-C-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl]-1-*O*-(tris(isopropyl)silyl)-β-D-talopyranuronate ((−)-24). A mixture of (−)-**22** (38 mg, 0.05 mmol), EtOAc (3.3 mL), and PtO_2 (80 mg) was degassed and pressurized with H_2 (1 atm.). After shaking for 1 h at 20 °C, the catalyst was filtered off through Celite and the solvent evaporated in vacuo. The residue (compound **23**) was taken up in Ac_2O (1.6 mL) and pyridine (42 μL). After stirring at 20 °C for 2 h, the solvent was evaporated in vacuo. The residue was purified by FC (EtOAc/light petroleum ether 1:1), yielding 33 mg (85%) of (−)-**24**, white foam. $[\alpha]^{25}_D = -0.6$ ($c = 1.0$, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.95 (d, $^3J = 10.4$), 5.43 (br s), 5.20−5.10 (br d), 5.05 (dd, $^3J = 2.7, 2.6$), 4.89 (d, $^3J = 1.7$), 4.41 (dd, $^2J = 12.2$, $^3J = 6.0$), 4.28−4.22 (br d, $^3J \cong 10.0$), 4.19 (d, $^3J = 1.3$), 4.10 (dd, $^2J = 12.2$, $^3J = 3.1$), 4.14−4.08 (m), 3.90−3.85 (m), 3.71 (s), 2.25−2.17 (m), 2.09−2.00 (6s), 1.80 (ddd, $^2J = 11.1$, $^3J = 11.2, 3.1$), 1.51 (ddd, $^2J = 11.1$, $^3J = 11.0, 2.9$), 1.10−1.00 (m).

Tris(isopropyl)silyl 2-Acetamido-2,3-dideoxy-3-*C*-(2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-β-D-talopyranoside (25). A 2 M solution of $LiBH_4$ in anhydrous THF (0.12 mL, 0.24 mmol) was added to a stirred solution of (−)-**24** (24 mg, 0.03 mmol) in anhydrous THF (3.4 mL) cooled to 0 °C. After stirring at 0 °C for 2 h, the mixture was stirred at 20 °C for 22 h. H_2O (2 mL) was added, and the solvent was evaporated in vacuo. The residue was purified by filtration through a column (∅ = 1 cm, $h = 3$ cm) of Dowex 50WX8 and rinsing with MeOH. (The Dowex was prewashed with 3.7% aqueous HCl first and then with H_2O and finally with MeOH). Solvent evaporation yielded 13 mg (78%) of **25**, yellowish oil. ^{1}H NMR (600 MHz, C_5D_5N) δ 7.34 (d, $^3J = 10.0$, NH), 5.10 (d, $^3J = 1.6$, H-C(1)), 4.85 (ddd, ^3J(HproS-C(7′)-H-C(6′)) = 11.2, ^3J(HproR-C(7′)-H-C(6′)) = 3.2, ^3J(H-C(5′)-H-C(6′)) = 3.1, H-C(6′)), 4.70 (br d, $^3J \cong 10.0$, H-C(2)), 4.58−4.55 (m, H-C(4), H-C(1′)), 4.49 (dd, $^3J = 7.9, 3.3$, H-C(4′)), 4.46 (dd, $^2J = 11.0$, $^3J = 3.3$, H-C(1′)), 4.39−4.37 (m, H-C(3′), H-C(5′)), 4.35 (dd, $^2J = 10.8$, $^3J = 6.7$, H-C(6)), 4.28 (ddd, $^3J = 7.5, 7.5, 3.3$, H-C(2′)), 4.23 (dd, $^2J = 10.8$, $^3J = 5.4$, H-C(6)), 3.85 (br t, $^3J \cong 10.0$, H-C(5)), 2.63−2.59 (*m*, H-C(3)), 2.54 (ddd, $^2J = 14.4$, ^3J(HproR-C(7′)-H-C(3)) = 9.7, ^3J(HproR-C(7′)-H-C(6′)) = 3.2, HproR-C(7′)), 2.43 (ddd, $^2J = 14.4$, ^3J(HproS-C(7′)-H-C(6′)) = 11.2, ^3J(HproS-C(7′)-H-C(3)) = 4.3, HproS−C(7′)), 1.93 (s, NHAc), 1.20−1.00 (m, 21H, TIPS).

Tris(isopropyl)silyl 2-Acetamido-4,6-di-*O*-acetyl-2,3-dideoxy-3-*C*-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-β-D-talopyranoside ((−)-26). A mixture of **25** (13 mg, 24 μmol), pyridine (2.8 mL), Ac_2O (0.9 mL), and 4-(dimethylamino)pyridine (2 mg) was stirred at 20 °C for 12 h. The solvent was evaporated in vacuo and the residue purified by FC (EtOAc/light petroleum ether 1:1), yielding 18 mg (93%) of (−)-**26**, yellowish oil. $[\alpha]^{25}_D = -5.0$ ($c = 0.25$, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.87 (d, $^3J = 10.4$), 5.20−5.10 (m), 5.06 (dd, $^3J = 2.8, 2.7$), 4.87 (d, $^3J = 1.7$), 4.42 (dd, $^2J = 12.2$, $^3J = 5.9$), 4.26−4.22 (br d, $^3J \cong 9.0$), 4.15−4.00 (m), 3.90−3.86 (m), 3.83 (br t, $^3J \cong 6.8$), 2.15−2.02 (7s + m), 1.80 (ddd, $^2J = 14.4$, $^3J = 14.5, 3.2$), 1.50 (ddd, $^2J = 14.4$, $^3J = 14.3, 2.8$), 1.12−1.04 (m).

2-Acetamido-4,6-di-*O*-acetyl-2,3-dideoxy-3-*C*-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-α-D-talopyranose ((+)-27). A mixture of (−)-**26** (70 mg, 0.09 mmol), THF (4.2 mL), and tetrabutylammonium fluoride (130 μL, 0.13 mmol) was stirred at 0 °C under

N_2 atmosphere for 5 min. After filtration through a column of Florisil (∅ = 2 cm, *h* = 3 cm) and rinsing with EtOAc (50 mL), the solvent was evaporated in vacuo. The residue was purified by FC on Florisil (EtOAc), yielding 42 mg (74%) of (+)-**27**, yellow oil. $[\alpha]^{25}_D = 14$ (*c* = 1.0, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 6.29 (d, ^3J = 9.9), 5.24 (br s), 5.21−5.11 (m), 5.09 (d, ^3J = 3.7), 5.07 (dd, ^3J = 3.3, 3.2), 4.43−4.36 (m), 4.15−3.98 (m), 3.85 (ddd, ^3J = 7.2, 7.1, 3.0), 2.60 (ddd, ^3J = 10.2, 6.4, 3.4), 2.17−2.02 (7s), 1.68 (ddd, ^2J = 14.5, ^3J = 12.0, 3.4), 1.44 (ddd, ^2J = 14.5, ^3J = 10.7, 3.0).

Methyl (4-*O*-Acetyl-2-azido-1-*O*-(*tert*-butyldimethylsilyl)-2,3-dideoxy-3-*C*-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-β-D-talopyranuronate ((−)-29). (−)-**21** (631 mg, 1.04 mmol) was dissolved in CH_2Cl_2 (19 mL) at 0 °C under N_2, and then 2,6-lutidine (334 mg, 3.12 mmol) and (*tert*-butyl)dimethylsilyl trifluoromethanesulfonate (TBDMSOTf, 550 mg, 2.08 mmol) were added. After stirring at 0 °C for 20 min, the mixture was allowed to warm to 20 °C and stirred for 2.5 h; by TLC there was still unreacted (−)-**21**. The mixture was thus cooled again to 0 °C, and 2,6-lutidine (167 mg, 1.56 mmol) and TBDMSOTf (275 mg, 1.04 mmol) were added. The mixture was allowed to warm to 20 °C and stirred for 1.5 h. Brine (20 mL) was added, and the aqueous phase was extracted with CH_2Cl_2 and (20 mL, four times). The combined extracts were dried ($MgSO_4$). After solvent evaporation in vacuo, the residue was purified by FC (EtOAc/light petroleum ether 2:3), yielding 560 mg (75%) of (−)-**29** contaminated by 10−20% of the α-pyranoside. White foam. $[\alpha]^{25}_D = -56$ (*c* = 1.0, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.38 (dd, ^3J = 3.0, 2.0), 5.25 (dd, ^3J = 7.4, 3.2), 5.12 (dd, ^3J = 7.4, 6.5), 5.03 (dd, ^3J = 5.0, 3.2), 4.94 (d, ^3J = 1.7), 4.49 (dd, ^2J = 12.2, ^3J = 6.8), 4.20−4.10 (m), 3.95 (ddd, ^3J = 6.8, 6.5, 3.3), 3.74 (s), 3.44 (dd, ^3J = 3.6, 1.7), 2.20−2.00 (5s + m), 1.83−1.67 (m), 0.96 (s), 0.24 (s), 0.17 (s).

Methyl (2-Acetamido-4-*O*-acetyl-1-*O*-(*tert*-butyldimethylsilyl)-2,3-dideoxy-3-*C*-(1′,3′,4′,5′-tetra-*O*-acetyl-2′,6′-anhydro-7′-deoxy-D-*glycero*-D-*manno*-heptitol-7′-*C*-yl)-β-D-talopyranuronate ((−)-30). After degassing, a mixture of (−)-**29** (508 mg, 0.71 mmol), EtOAc (47 mL) and PtO_2 (250 mg) was pressurized with H_2 (1 atm) and shaken at 20 °C for 18 h. The catalyst was filtered off and the solvent evaporated. The residue was taken up in Ac_2O (14 mL). After the addition of pyridine (0.35 mL), the mixture was stirred at 20 °C for 2 h. The solvent was evaporated in vacuo and the residue purified by FC (EtOAc/light petroleum ether 7:3), yielding 465 mg (89%) of (−)-**30**, white foam. $[\alpha]^{25}_D = -4.3$ (*c* = 1.0, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ 5.94 (d, ^3J = 10.4), 5.43 (br s), 5.19−5.17 (m), 5.06 (dd, ^3J = 3.0, 2.9), 4.84 (d, ^3J = 1.8), 4.42 (dd, ^2J = 12.2, ^3J = 6.0), 4.23−4.18 (m), 4.15−4.09 (m), 3.91−3.85 (m), 3.73 (s), 2.30−2.03 (5s + m), 1.80 (ddd, ^2J = 14.5, ^3J = 12.0, 3.1), 1.52 (ddd, ^2J = 14.5, ^3J = 10.7, 3.1), 0.90 (s), 0.17 (s), 0.11 (s).

Glycosidase Inhibition Assays. See ref 27.

Glycosyltransferase Inhibition Assays. The β-1,4-galactosyltransferase from human milk was assayed following the protocol of Berger[43] using the following conditions: 10 mM $MnCl_2$, 10 mM GlcNAc, 400 μM UDP-Gal, 0.15 μM ^{14}C-UDP, and enzyme in Tris-HCl (100 mM, pH 7.4), final volume of 50 μL. The α-2,6-sialtransferase, a soluble form of which that had been expressed in *Pichia pastoris* has been tested as described previously[44] using the following conditions: 10 mM LacNAc, 500 μM CMP-Nana (*N*-acetylneuraminic acid), 0.035 μmol ^{14}C-CPM-Nana, and enzyme cocodylate buffer (100 μM, pH 6.8), final volume of 50 μL. The α-1,3-fucosyltransferase VI (Fuc-TVI), a soluble form of which that has been expressed in *Pichia pastoris*, was assayed as described previously[44] with the following modifications:[45] 5 mM ATP, 10 mM $MnCl_2$, 100 μM GDP-fucose, 5 mL LacNAc (unless specified), 1.6 μM ^{14}C-GDP-fucose, and enzyme in cacodylate buffer (1 M, pH 6.2), final volume of 50 μL. The C-disaccharides **1** and **2**, and test compounds such as D-mannose, *N*-acetylgalactosamine, and CH_3CONH_2, were soluble in H_2O. For inhibition measurements the enzymes were preincubated for 5 min at 20 °C with the inhibitor of various concentrations. The reactions were initiated on adding the substrate and were run for 20 min at 37 °C for Fuc-TVI[46,47] and galactosyltransferase, and for 2 h at 37 °C for sialyltransferase.

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Supporting Information Available: Detailed ^{1}H and ^{13}C NMR spectra and signal assignments, further optical rotation data, and UV, IR, and MS spectra. This material is available free of charge via the Internet at http://pubs.acs.org.

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