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Note

p-Benzoquinone as precursor for the synthesis of modified D- and L-hexoses: Preparation of 2-acetamido-2,4-dideoxy-D- and L-*xylo*-hexopyranose

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Six-membered carbocycles are useful starting materials for the preparation of modified hexoses. This was demonstrated for the first time by the conversion of *cis*-3,5-cyclo-hexadien-1,2-diol into *N*-acetyl D- and L-glucosamine[1]. *cis*-3,5-Cyclohexadien-1,2-diol prepared by microbial oxidation of benzene has the disadvantage of being extremely expensive [Aldrich Chemical Co., No. 36,506-8: *cis*-3,5-cyclohexadien-1,2-diol (20 wt% in ethyl acetate)]. We now resort to (±)-(3/4,5,6)-4-bromo-5,6-epoxy-3-hydroxycyclohexene[2] (**1**) (only one enantiomer is depicted) for the facile preparation of the title compound as pure enantiomers, destined for use in investigations of galactosyltransferase acceptor binding-sites with spacer-modified disaccharides[3].

2-Acetamido-2,4-dideoxy-D-*xylo*-hexopyranose, one of the enantiomers, has been prepared from 2-acetamido-D-glucose[4] as a precursor, from 1,6-anhydro-D-glucose[5], and from (*S*)-malic acid[6].

The epoxide **1**, which is readily available from *p*-benzoquinone in large quantities and in only three steps[2,7], reacts regio- and stereo-selectively with sodium azide[7] to yield (*3,6/4,5*)-6-azido-4-bromo-3,5-dihydroxycyclohexene (**2**). The double bond in **2** undergoes quantitative ozonolysis under mild conditions, whereby the dialdehyde (**3a**) is formed. The enantiomeric aldohexoses (**4**) are obtained by treating the mixture with sodium cyanoborohydride. Specific reduction of **3a** is based on the fact that it cyclises spontaneously with predominant formation of the six-membered cyclic hemiacetal (**3b**). This specific reduction may also be used with sodium [^{3}H]cyanoborohydride, to afford radioactively labelled material, which is needed for the aforementioned biochemical investigations. The azido group in the

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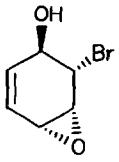
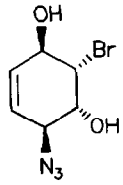


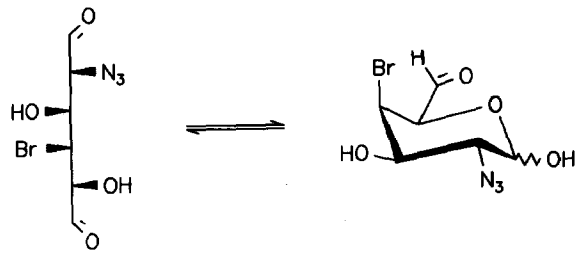
1



2

O-acetylated compound **5** is reduced by hydrogen using Pd/C to yield an amino hexose **6**, which is converted into the corresponding phthalimido hexose **7**. Bromine in **7** is readily replaced by hydrogen using tributylstannane to yield the enantiomeric 2-phthalimido-2,4-dideoxy-hexopyranoses (**8**), which are convertible into the allyl β-D,L-glycosides (**9**). Liberation of the amino group by removal of the phthalyl blocking group, followed by *N*-acetylation gives **10**, of which the D enantiomer is quantitatively hydrolysed by commercial hexosaminidase to give 2-acetamido-2,4-dideoxy-D-*xylo*-hexopyranose (**11**). The remaining allyl 2-acetamido-2,4-dideoxy-β-L-*xylo*-hexopyranoside (**12**) is then chemically hydrolysed. The separated, enantiomeric *N*-acetylhexosamines **11** and **13** have the same numerical values of optical rotation with opposite signs.

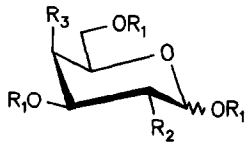
The ^{1}H NMR data (D_2O) of **11** and **13** were identical with those of an authentic sample of 2-acetamido-2,4-dideoxy-D-*xylo*-hexopyranose[6].



3a 3b

EXPERIMENTAL

General methods.—All reactions were monitored by TLC on Silica Gel 60 F_{254} (Merck). Flash-column chromatography[8] was performed on Silica 32–63, 60A (ICN Biomedicals). Size-exclusion chromatography was performed on Bio-Gel P-2 (−400 mesh, Bio-Rad). Optical rotations were obtained with a Schmidt & Haensch Polartronic I polarimeter. ^{1}H NMR spectra were recorded with a Bruker WM 250 spectrometer at 250 MHz for solutions in $CDCl_3$, MeOH-d_4 (internal Me_4Si) and D_2O (internal DSS). Melting points were measured with a Büchi apparatus and are uncorrected. Elemental analyses were obtained with a Perkin–Elmer 240



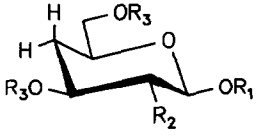
4 R_1 = H, R_2 = N_3, R_3 = Br

5 R_1 = Ac, R_2 = N_3, R_3 = Br

6 R_1 = Ac, R_2 = NH_2, R_3 = Br

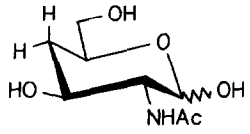
7 R_1 = Ac, R_2 = NPhth, R_3 = Br

8 R_1 = Ac, R_2 = NPhth, R_3 = H

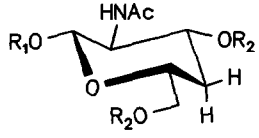


9 R_1 = Allyl, R_2 = NPhth, R_3 = Ac

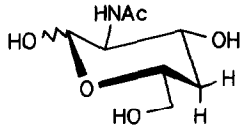
10 R_1 = Allyl, R_2 = NHAc, R_3 = H



11



12 R_1 = Allyl, R_2 = H



13

analyzer. IR spectra were recorded with a Perkin–Elmer 1320 spectrophotometer. Ozonolyses were carried out with a Fischer ozone generator 500 M.

Materials.—*N*-Acetyl-β-D-glucosaminidase (2-acetamido-2-deoxy-β-D-glucoside acetamido-deoxyglucohydrolase; EC 3.2.1.30) from jack beans [56 U/mg, 1.0 mg protein/mL, suspension in 2.5 M $(NH_4)_2SO_4$] was purchased from Sigma.

*1,3,6-Tri-*O*-acetyl 2-azido-4-bromo-2,4-dideoxy-*D,L*-galactopyranose* (**5**).—Ozone (30 L/h O_2, 10 mmol O_3/h) was bubbled through a solution of (*3,6 / 4,5*)-6-azido-4-bromo-3,5-dihydroxy-cyclohexene **2** (9.3 g, 39.7 mmol) in MeOH (100 mL) at −78°C. After the blue colour of the solution persisted for 10 min, the excess ozone was removed with a stream of oxygen. Dimethyl sulfide (6 mL) was added and the mixture was allowed to attain 25°C. After additional stirring for 2 h the solution was concentrated in vacuo and MeOH (3 × 50 mL) distilled from the residue to give crude **3a,b**, which was used without further purification. Product **3a,b** was dissolved in MeOH (100 mL), sodium cyanoborohydride (1.0 g, 15.9 mmol) was added, and the mixture was stirred at 25°C, while AcOH (6 mL) was added, to keep the pH at 3–4. After the reduction was complete, the solvent was removed

under diminished pressure and MeOH (2 × 50 mL) was distilled from the residue to give crude **4**, which was acetylated using 2:1 pyridine–Ac_2O (60 mL) at 25°C overnight. The mixture was concentrated in vacuo and toluene (2 × 100 mL) distilled from the residue, and then dissolved in CH_2Cl_2 (300 mL). The organic layer was neutralized with satd aq $NaHCO_3$ (2 × 100 mL), washed with water (100 mL), dried ($MgSO_4$), and concentrated in vacuo. Flash-column chromatography (1:3 EtOAc-cyclohexane) of the residue gave **5** (11 g, 70% overall yield from **2**), isolated as a colourless syrup which was precipitated in EtOAc–cyclohexane as an anomeric mixture; R_f α,β anomers 0.28, 0.25 (1:3 EtOAc–cyclohexane); ν^{film} 2100 cm^{-1} (N_3); ^1H NMR data ($CDCl_3$): α anomer: δ 6.31 (d, 1 H, $J_{1,2}$ 3.75 Hz, H-1), 5.10 (dd, 1 H, $J_{3,2}$ 10.5, $J_{3,4}$ 3.75 Hz, H-3), 4.73 (dd, 1 H, $J_{4,5}$ 1.5 Hz, H-4), 4.26 (dd, 1 H, $J_{6a,6b}$ 9, $J_{6a,5}$ 5.25 Hz, H-6a), 4.20 (m, 1 H, H-5), 4.13 (dd, 1 H, $J_{6b,5}$ 4.5 Hz, H-6b), 4.11 (dd, 1 H, H-2), 2.22 (s, 3 H, Ac), 2.17 (s, 3 H, Ac), and 2.08 (s, 3 H, Ac). ^1H NMR data ($CDCl_3$): β anomer: δ 5.54 (d, 1 H, $J_{1,2}$ 8.25 Hz, H-1), 4.72 (dd, 1 H, $J_{3,2}$ 10.5, $J_{3,4}$ 3.75 Hz, H-3), 4.58 (dd, 1 H, $J_{4,5}$ 1.5 Hz, H-4), 4.30 (dd, 1 H, $J_{6a,6b}$ 12, $J_{6a,5}$ 6.5 Hz, H-6a), 4.14 (dd, 1 H, H-6b), 3.98 (dd, 1 H, H-2), 3.89 (m, 1 H, H-5), and 2.21-2.07 (3 s, 9 H, 3 Ac). Anal. Calcd for $C_{12}H_{16}BrN_3O_7$: C, 36.56; H, 4.10; Br, 20.27; N, 10.66. Found: C, 36.80; H, 4.11; Br, 20.12; N, 10.57.

*1,3,6-Tri-O-acetyl-4-bromo-2,4-dideoxy-2-phthalimido-*D,L*-galactopyranose* (**7**).—To a solution of **5** (3 g, 7.7 mmol) in EtOAc (50 mL) Pd/C was added and treated with H_2 under atmospheric pressure at 25°C. After the reduction was complete, shown by TLC (R_f 0.63, 7:2:1 EtOAc–MeOH–H_2O), the catalyst was filtered off and the solvent was evaporated in vacuo. The crude amine **6** was dissolved in MeOH (50 mL) and phthalic anhydride (859 mg, 5.8 mmol) was added. The solution was stirred at 45°C and after 10 min Et_3N (1.61 mL, 11.6 mmol) and additional phthalic anhydride (859 mg, 5.8 mmol) were added. After stirring for 18 h at 45°C, the solvent was removed under diminished pressure and the residue was dissolved in 2:1 pyridine–Ac_2O (50 mL) and kept at 25°C overnight. The mixture was concentrated in vacuo and toluene (2 × 100 mL) distilled from the residue, then filtered through silica gel (EtOAc) to yield a yellow syrup which was purified by flash-column chromatography (1:2 EtOAc–cyclohexane) to give **7** as a colourless syrup. Compound **7** was precipitated by EtOH as an anomeric mixture (3.1 g, 81%), R_f 0.46 (1:1 EtOAc–cyclohexane); ^1H NMR data ($CDCl_3$): β anomer: δ 7.92–7.74 (m, 4 H, Phth), 6.42 (d, 1 H, $J_{1,2}$ 9 Hz, H-1), 5.77 (dd, 1 H, $J_{3,2}$ 10.5, $J_{3,4}$ 3.75 Hz, H-3), 4.47–4.10 (m, 5 H, H-2, H-4, H-5, 6a,b), 2.12 (s, 3 H, Ac), 2.03 (s, 3 H, Ac), and 2.00 (s, 3H, Ac); ^1H NMR data ($CDCl_3$): α anomer: δ 7.92–7.74 (m, 4H, Phth), 6.34 (dd, 1 H, $J_{3,2}$ 12, $J_{3,4}$ 3.75 Hz, H-3), 6.31 (d, 1 H, $J_{1,2}$ 3.75 Hz, H-1), 5.06 (dd, 1 H, H-2), 4.95 (dd, 1 H, $J_{4,5}$ 1.5 Hz, H-4), 4.85–4.76 (m, 3 H, H-5, 6a,b), 2.11 (s, 3 H, Ac), 2.06 (s, 3 H, Ac), and 2.03 (s, 3 H, Ac). Anal. Calcd for $C_{20}H_{20}BrNO_9$: C, 48.21; H, 4.05; Br, 16.04; N, 2.81. Found: C, 48.50; H, 4.28; Br, 15.82; N, 2.62.

*1,3,6-Tri-O-acetyl-2,4-dideoxy-2-phthalimido-*D,L*-xylo-hexopyranose* (**8**).—Tributylstannane (1.6 mL, 5.9 mmol) and 2,2-azo-bis-(2-methyl-propanonitrile) (20

mg, 0.12 mmol) was added under N_2 to a benzene solution (30 mL) of **7** (2.7 g, 5.3 mmol). The solution was boiled under reflux for 8 h. After cooling, the unreacted stannane was decomposed by addition of CCl_4 (10 mL), the solution was evaporated in vacuo, and the product purified by flash-column chromatography (1:1 EtOAc–cyclohexane) to give an anomeric mixture of **8** as a colourless syrup (1 g, 90%), from which the α anomer crystallised from MeOH; R_f 0.35 (1:1 EtOAc–cyclohexane); mp 153–154°C; ^{1}H NMR data ($CDCl_3$): δ 7.88–7.72 (m, 4 H, Phth), 6.40 (dt, 1 H, $J_{3,2}$ 11.25, $J_{3,4ax}$ 11.25, $J_{3,4eq}$ 4.5 Hz, H-3), 6.33 (d, 1 H, $J_{1,2}$ 3.75 Hz, H-1), 4.56 (dd, 1 H, H-2), 4.36 (m, 1 H, $J_{5,4ax}$ 12, $J_{5,6a}$ 4.5, $J_{5,6b}$ 2.25, $J_{5,4eq}$ 2.25 Hz, H-5), 4.24–4.10 (m, 2 H, $J_{6a,b}$ 12 Hz, H-6a,b), 2.50 (ddd, 1 H, $J_{4eq,4ax}$ 12 Hz, H-4*eq*), 2.13 (s, 3 H, Ac), 2.06 (s, 3 H, Ac), 1.94 (s, 3 H, Ac), 1.73–1.57 (m, 1 H, H-4*ax*). Anal. Calcd for $C_{20}H_{21}NO_9$: C, 57.28; H, 5.05; N, 3.34. Found: C, 57.31; H, 5.20; N, 3.31.

*Allyl 3,6-di-O-acetyl-2,4-dideoxy-2-phthalimido-β-*D,L*-xylo-hexopyranoside* (**9**).—To a solution of **8** (1 g, 2.4 mmol) in dry CH_2Cl_2 (20 mL) $SnCl_4$ (0.3 mL, 2.4 mmol) was added at 0°C under anhydrous conditions. The mixture was stirred for 15 min, allyl alcohol (0.25 mL, 3.65 mmol) was added, and stirring was continued for 4 h at 0°C. The mixture was poured into ice-cold satd aq $NaHCO_3$ (100 mL) and stirred for 10 min. The organic layer was separated and the aqueous layer extracted with $CHCl_3$ (4 × 50 mL). The combined extracts were made neutral with satd aq $NaHCO_3$ (50 mL), washed with water (50 mL), dried ($MgSO_4$), and concentrated. Flash-column chromatography (1:2 EtOAc–cyclohexane) of the residue gave **9** as a colourless syrup, which crystallised from EtOH (900 mg, 89%); R_f 0.17 (1:2 EtOAc–cyclohexane); mp 73–74°C, ^{1}H NMR data ($CDCl_3$): δ 7.90–7.72 (m, 4 H, Phth), 5.81–5.62 (m, 2 H, allyl), 5.31 (d, 1 H, $J_{1,2}$ 8.25 Hz, H-1), 5.18–5.08 (m, 1 H, allyl), 5.07–5.01 (m, 1 H, allyl), 4.33–4.14 (m, 4 H, H-2,3,6a,b), 4.10–4.00 (m, 1 H, allyl), 3.92 (m, 1 H, H-5), 2.26 (ddd, 1 H, $J_{4eq,4ax}$ 12.75, $J_{4eq,3}$ 5.25, $J_{4eq,5}$ 2.25 Hz, H-4*eq*), 2.13 (s, 3 H, Ac), 1.90 (s, 3 H, Ac), and 1.74–1.58 (m, 1 H, H-4*ax*). Anal. Calcd for $C_{21}H_{23}NO_8$: C, 60.43; H, 5.55; N, 3.36. Found: C, 59.82; H, 5.62; N, 3.08.

*Allyl 2-acetamido-2,4-dideoxy-β-*D,L*-xylo-hexopyranoside* (**10**).—Compound **9** (1 g, 2.4 mmol) was dissolved in a mixture of 1:1 EtOH–$BuNH_2$ (25 mL) and the solution was heated under reflux for 20 h. After cooling, the mixture was concentrated in vacuo and MeOH was distilled from the residue. The residue was dissolved in 6:1 MeOH–Ac_2O (35 mL) and the solution kept for 5 h at 25°C. The solution was concentrated in vacuo and toluene (3 × 30 mL) distilled from the residue. Flash-column chromatography (17:2:1 EtOAc–MeOH–H_2O) of the residue yielded **10** as a syrup, which crystallised from EtOH–ether (400 mg, 70%); R_f 0.22 (17:2:1 EtOAc–MeOH–H_2O); mp 170–171°C; ^{1}H NMR data (D_2O): δ 5.86–5.69 (m, 1 H, allyl), 5.22–5.10 (m, 2 H, allyl), 4.35 (d, 1 H, $J_{1,2}$ 8.25 Hz, H-1), 4.21 (m, 1 H, allyl), 4.02 (m, 1 H, allyl), 3.77–3.38 (m, 5 H, H-2,3,5,6a,b), 1.94–1.85 (ddd, 1 H, $J_{4eq,4ax}$ 12, $J_{4eq,3}$ 5.25, $J_{4eq,5}$ 1.5 Hz, H-4*eq*), and 1.39–1.23 (m, 1 H, H-4*ax*). Anal. Calcd for $C_{11}H_{19}NO_5$: C, 53.87; H, 7.81; N, 5.71. Found: C, 53.64; H, 7.73; N, 5.61.

*2-Acetamido-2,4-dideoxy-*D*-xylo-hexopyranose* (**11**) *and allyl 2-acetamido-2,4-dideoxy-β-*L*-xylo-hexopyranoside* (**12**).—To a solution of **10** (286 mg, 1.2 mmol) in sodium citrate–HCl buffer (8 mL, 50 μmol, pH 5.0) *N*-acetyl-β-D-glucosaminidase (20 units) was added. The mixture was kept at 25°C. The reaction was monitored by TLC (7:2:1 EtOAc–MeOH–H_2O). After 7 days, the mixture was concentrated in vacuo and MeOH was distilled from the residue. Compounds **11** and **12** were separated by flash-column chromatography (7:2:1 EtOAc–MeOH–H_2O), to yield **11** (113 mg, 0.55 mmol) as a colourless syrup. Compound **11** (75 mg, 0.37 mmol) was purified by size-exclusion chromatography with a column of Bio-Gel P-2 (2.5 × 140 cm, 40°C, 100 mL H_2O/h) and lyophilized; R_f α, β anomers 0.32, 0.21 (7:2:1 EtOAc–MeOH–H_2O); $[\alpha]_D^{23}$ + 63° (*c* 0.5, 25 h, H_2O); ref.[6] $[\alpha]_D^{22}$ + 74.7 (*c* 1.04, 15 min, H_2O), $[\alpha]_D^{22}$ + 68.0° (*c* 1.04, 25 h, H_2O); ref.[4] $[\alpha]_D^{22}$ + 78° (*c* 1.58, H_2O); ref.[5] $[\alpha]_D^{22}$ + 73° (*c* 1.05, 12 h, H_2O). The ^1H NMR data were identical with those of an authentic sample[7] of **11**. ^1H NMR data (D_2O): δ 5.24 (d, 1 H, $J_{1,2}$ 3.5 Hz, H-1α), 4.63 (d, 1 H, $J_{1,2}$ 8.5 Hz, H-1β), 4.11 (m, 1 H, H-5α/β), 3.99 (ddd, 1 H, H-3α/β), 3.85–3.47 (m, 8 H, H-2α, 6a,bα, 2β, 3α/β, 5α/β, 6a,bβ), 2.11–1.96 (m, 2 H, H-4*eq*α,β), 2.04 (s, 3 H, Ac), and 1.58–1.38 (m, 2 H, H-4*ax*α,β).

Compound **12** was crystallised from EtOH–ether (116 mg, 0.45 mmol); R_f 0.39 (7:2:1 EtOAc–MeOH–H_2O); mp 170–171°C, $[\alpha]_D^{23}$ + 7° (*c* 1, H_2O); ^1H NMR data (CD_3OD): δ 5.96–5.81 (m, 1 H, allyl), 5.32–5.21 (m, 1 H, allyl), 5.17–5.08 (m, 1 H, allyl), 4.36–4.28 (m, 1 H, allyl), 4.35 (d, 1 H, $J_{1,2}$ 8.25 Hz, H-1), 4.06 (m, 1 H, allyl), 3.74–3.46 (m, 5 H, H-2,3,5,6a,b), 2.03–1.93 (ddd, 1 H, $J_{4eq,4ax}$ 12.75, $J_{4eq,3}$ 4.5, $J_{4eq,5}$ 1.5 Hz, H-4*eq*), and 1.47–1.32 (dt, 1 H, H-4*ax*). Anal. Calcd for $C_{11}H_{19}NO_5$: C, 53.87; H, 7.81; N, 5.71. Found: C, 53.80; H, 7.77; N, 5.61.

*2-Acetamido-2,4-dideoxy-*L*-xylo-hexopyranose* (**13**).—To a solution of **12** (84 mg, 0.34 mmol) in EtOH (8 mL), ethyl diisopropylamine (0.5 mL), and tris(triphenylphosphine)rhodium(I) chloride (30 mg, 0.03 mmol) was added. The mixture was heated under reflux for 5 h. The solution was concentrated in vacuo and CCl_4 (5 mL) distilled from the residue. The residue was dissolved in 9:1 acetone–water (10 mL) and $HgCl_2$ (10 mg, 0.04 mmol) was added. The solution was stirred for 1 h at 25°C and concentrated in vacuo, MeOH was distilled from the residue, and filtered through silica gel (MeOH). The filtrate was concentrated under diminished pressure and the residue purified by flash-column chromatography (7:2:1 EtOAc–MeOH–H_2O) to yield **13** as a colourless syrup, which was dissolved in water and lyophilized (43 mg, 62%); R_f α,β anomers 0.32, 0.21 (7:2:1 EtOAc–MeOH–H_2O), $[\alpha]_D^{23}$ – 63° (*c* 0.8, 25 h, H_2O); ^1H NMR data (D_2O): identical with ^1H NMR data of **11**.

ACKNOWLEDGMENT

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