Carbohydrate Research
Elsevier Publishing Company, Amsterdam
Printed in Belgium

EXTENSION OF SUGAR CHAINS THROUGH ACETYLENIC INTERMEDIATES

PART II[1]. DERIVATIVES OF 1-HEPTYNE-L-*gluco*- AND L-*manno*-3,4,5,6,7-PENTOL

D. HORTON AND J. M. J. TRONCHET
Department of Chemistry, The Ohio State University, Columbus, Ohio 43210 (U.S.A.)

(Received March 11th, 1966)

The acetylene function is a versatile group which can be employed in a wide range of synthetic reactions[2]. It has been utilized in the carbohydrate field for the total syntheses of meso and DL alditols[3,4], and related derivatives[5,6], from non-carbohydrate precursors. Hydroxylated, acetylenic derivatives are of interest as potential tranquilizers[7]. The present report* describes the synthesis of optically active, acetylenic sugar derivatives by the ethynylation of an aldehydo sugar derivative. Also described are methods for separation of the epimers formed in the reaction, procedures for configurational assignment at the newly formed asymmetric center, and applications of n.m.r. techniques for structural assignment.

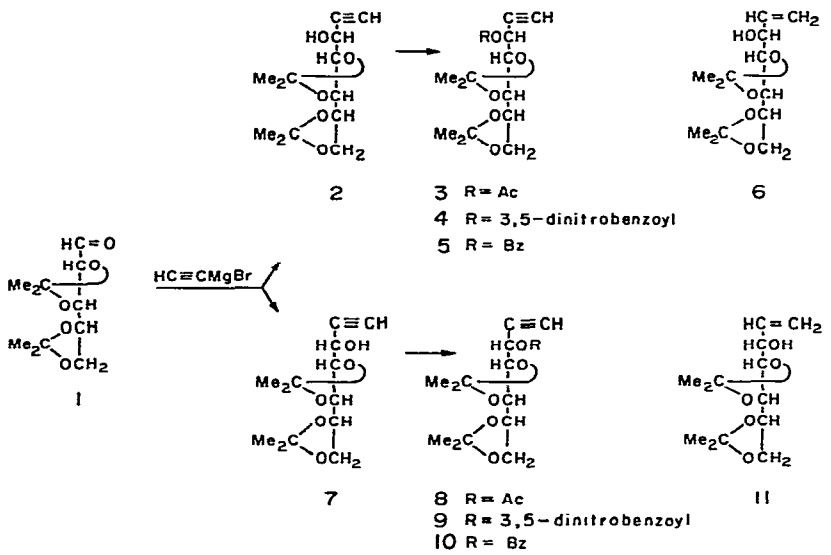
2,3:4,5-Di-*O*-isopropylidene-*aldehydo*-L-arabinose (**1**), prepared through L-arabinose diethyl dithioacetal by a standard procedure[8], underwent reaction with ethynylmagnesium bromide in tetrahydrofuran at room temperature to give the mixed 3-epimeric heptyne derivatives (**2** and **7**) in almost quantitative yield. It was essential to maintain a large excess of acetylene in the system during the reaction, as otherwise the yield of **2** and **7** was diminished by the condensation of two molecules of sugar derivative with one of acetylene, to give a high-boiling side-product. The product (**2** and **7**) showed the anticipated acetylenic hydrogen and C≡C stretching frequencies in its infrared spectrum, at 3.06 μ and 4.74 μ, respectively. The acetylenic hydrogen signal at $\tau \sim 7.50$ in the n.m.r. spectrum was very characteristic. The two epimers were resolved on thin-layer chromatograms, and the evidence of t.l.c. and integrated n.m.r. spectra provided the most reliable criteria of purity of the product.

The two epimers were effectively separated by preparative, gas–liquid chromatography (g.l.c.). The first component to be eluted, approximately 60% of the mixture, was obtained crystalline, m.p. 67–69°, $[\alpha]_D$ +6° (chloroform), and subsequent degradation studies showed it to be the L-*gluco* epimer (**2**). The slower-moving component, about 40% of the mixture, was obtained as a liquid, $[\alpha]_D$ −24° (chloroform), and this was subsequently shown to be the L-*manno* epimer (**7**).

A procedure was sought that would provide a convenient separation of the epimers on a larger scale. Acetylation of the epimers **2** and **7** gave the corresponding 3-acetates (**3** and **8**), which were characterized by physical methods, but they proved

*For a preliminary communication, see Ref. 1.

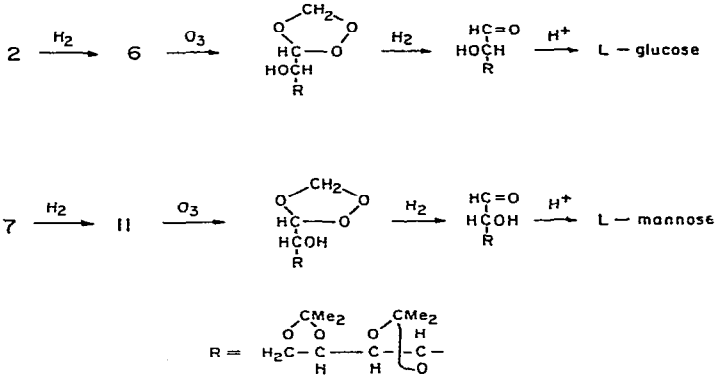
difficult to separate in admixture on a preparative scale. The corresponding 3-benzoates (**5** and **10**) were also prepared, and were characterized by physical methods, but a suitable method for separation in admixture was not found.



Acylation of the mixed epimers (**2** and **7**) with 3,5-dinitrobenzoyl chloride gave the crystalline, mixed, 3-epimeric 3,5-dinitrobenzoates **4** and **9** in essentially quantitative yield. Recrystallization of this mixture several times from ethyl alcohol gave the pure L-*manno* epimer (**9**) in 36% yield (32% overall from **1**); its melting point (167.0–167.5°) and specific rotation (+2.9°) did not change on further recrystallization, and it was identical with a sample of **9** which had been prepared by 3,5-dinitrobenzoylation of the pure epimer **7** obtained by g.l.c. of the mixture of **2** and **7**. The mother liquors from the crystallization of **9** contained the L-*gluco* epimer (**4**) almost exclusively (50% overall yield from **1**), and recrystallization gave pure **4**, m.p. 134.5–135.5°, $[\alpha]_D$ −19.5°.

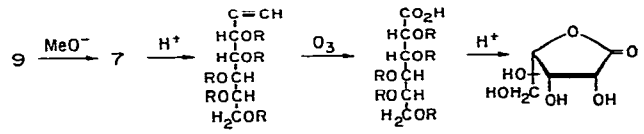
Proof of configuration of **2** and its derivatives (**3**, **4**, and **5**), and of **7** and its derivatives (**8**, **9**, and **10**), was achieved by two independent degradative routes. In the first route, the heptyne derivatives were degraded by C-1–C-2 cleavage to the corresponding hexoses. Conditions were established with the mixed heptyne derivatives (**2** and **7**) for hemihydrogenation to the corresponding heptenes (**6** and **11**), over a partially poisoned palladium catalyst (Lindlar[9] catalyst). N.m.r. spectroscopy provided a very sensitive index of the progress of the reaction; the sharp one-proton doublet at τ 7.50 for the acetylenic hydrogen disappeared, and was replaced by a three-proton multiplet in the vinyl region (τ 3.68–4.95). Over-reduction was indicated by the appearance of a triplet at τ 8.82 and a quartet at τ 6.38, due to an ethyl group, and a diminution in the intensity of the vinyl signals. Different batches of catalyst required different lengths of time for hemihydrogenation, but, in each instance, it was

possible to interrupt the hydrogenation when all of the acetylene had been reduced (but no reduction of the alkene had occurred). Each batch of catalyst was evaluated by reduction of the model alkynols 3-methyl-1-butyn-3-ol and 3-methyl-1-pentyn-3-ol to the corresponding alkenes; n.m.r. data for the model compounds and their reduction products are recorded in the Experimental section. The heptyne derivatives (**2** and **7**) were separately reduced to the heptenes (**6** and **11**), and each product was subjected to ozonolysis. The "ozonides" were cleaved by hydrogenolysis, and the resultant 3,4:5,6-di-*O*-isopropylidene-*aldehydo*-L-hexoses were hydrolyzed to the free hexoses. The hexose obtained from **2** was chromatographically indistinguishable from a glucose, and that from **7** was indistinguishable from a mannose, indicating that **2** and its derivatives have the L-*gluco* configuration, and **7** and its derivatives have the L-*manno* configuration. Some arabinose was present in each hydrolysis product, and it was probably formed by additional cleavage during the ozonolysis step. When the degradation was repeated with the corresponding 3-acetates **3** and **8**, the extent of degradation to arabinose was much diminished, and the hexoses formed were again glucose and mannose, respectively. The overall sequence of conversions from **1** constitutes a method for chain ascent in the sugars.

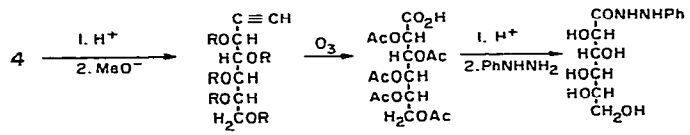


The second sequence for proof of the configuration of **2** and **7** sets out from the crystalline 3,5-dinitrobenzoates **4** and **9**, and the final products were characterized on a crystalline basis. The less-soluble isomer (**9**) of the two 3,5-dinitrobenzoates was saponified to give **7**, and the isopropylidene groups were removed by hydrolysis with 60% acetic acid to give the heptynepentol **12**, which was converted into the pentaacetate **13**. Ozonolysis of **13** for an extended period of time gave penta-*O*-acetyl-L-mannonic acid (**14**), in 41% overall yield from **9**. The latter reaction[10] was first applied in the carbohydrate field by Raphael, for the conversion of 1-pentyne-DL-*erythro*-3,4,5-triol into DL-erythronic acid[4]. Saponification of the pentaacetate **14** gave L-mannonic acid (**15**), which was converted into L-mannono-1,4-lactone (**16**). The latter was identified by chromatography, by its sign of rotation, and by the fact that the X-ray powder diffraction pattern of the crystalline material was identical with that of a sample of the authentic D enantiomorph. Similarly, the

3,5-dinitrobenzoate **4** was deacetonated, and the product was converted, by way of the heptynepentol **17** into the pentaacetate **18**. Ozonolysis of the latter gave penta-*O*-acetyl-L-gluconic acid (**19**), which, on hydrolysis followed by treatment with phenylhydrazine, gave crystalline L-gluconic acid phenylhydrazide (**20**). The X-ray powder diffraction pattern of **20** was identical with that of an authentic sample of the D enantiomorph. An alternative conversion of **4** into **20** was effected by ozonolysis of **4**, followed by hydrolysis, and conversion of the resultant D-glucono-1,4(and 1,5)-lactones into **20**.



12 R = H
13 R = Ac
14 R = Ac
15 R = H
16



17 R = H
18 R = Ac
19
20

It is possible to correlate the appearance of the n.m.r. signal for H-3 with the configuration at C-3 in the heptyne derivatives. The spectra of **2** and **7** are not suitable for exact analysis, owing to coupling of the hydroxyl proton with H-3, and because the H-3 signal occurs close to the multiplet "envelope" of signals for hydrogens on the rest of the chain. The H-3 signal is, however, shifted to low field in the acylated derivatives, and coupling of this proton to H-1 and H-4 can be observed. All of the derivatives showed coupling of H-1 with H-3, $J_{1,3}$ 2.2–2.3 c.p.s. The L-*gluco* derivatives **3**, **4**, and **5** all showed larger coupling of H-3 with H-4 ($J_{3,4}$ 3.6–4.0 c.p.s.) than the corresponding L-*manno* derivatives **8**, **9**, and **10** ($J_{3,4}$ 2.4–2.9 c.p.s.). This observation may be of general utility for assignment of configuration at the newly formed asymmetric center in derivatives of this type[11]. The data indicate that the stereoelectronic requirements of the various ester functions (OR') exceed those of

A B

the ethynyl group, so that the favored rotamer state of the L-*gluco* derivatives **3**, **4**, and **5** is that indicated in A, wherein the OR' group is antiparallel to C-5 and the

rest of the sugar chain (R), and the favored rotamer state in the L-*manno* derivatives **8**, **9**, and **10** is B, having a similar disposition of the R and OR' groups. The observed $J_{3,4}$ couplings, which will reflect a population-weighted time-average of the three rotamer states, are greater for **3**, **4**, and **5** (large contribution from rotamer A, with antiparallel H-3 and H-4) than for **8**, **9**, and **10** (large contribution from rotamer B, with gauche H-3 and H-4).

EXPERIMENTAL

General methods

Solutions were concentrated below 40° under diminished pressure. Melting points were determined with a Thomas–Hoover melting-point apparatus (Arthur H. Thomas Co., Philadelphia, Pennsylvania) and are uncorrected. Specific rotations were determined in a 2-dm polarimeter tube. Infrared spectra were measured with a Perkin-Elmer Model 137 "Infracord" infrared spectrometer. Nuclear magnetic resonance spectra were measured at 60 Mc.p.s. with a Varian A-60 n.m.r. spectrometer. Chemical shifts are given on the τ scale, and spectra were measured, unless otherwise stated, at *ca* 30° with solutions (10–20%) in deuteriochloroform, with tetramethylsilane (τ 10.00) as the internal standard. Deuteration was effected by shaking the prepared sample with one drop of deuterium oxide. Spectra were analyzed on a first-order basis. Microanalyses were determined by W. N. Rond. X-Ray powder diffraction data give interplanar spacings, Å, for CuK_α radiation. Relative intensities were estimated visually: s, strong; m, moderate; w, weak; v, very. The strongest lines are numbered (1, strongest); double numbers indicate approximately equal intensities. Thin-layer chromatography was performed with Desaga equipment, with silica gel G (E. Merck, Darmstadt, Germany) activated at 110° as the adsorbent. Indication was effected with sulfuric acid.

4,5:6,7-Di-O-*isopropylidene*-1-*heptyne*-L-gluco(*and* L-manno)-*3,4,5,6,7-pentol* (**2** *and* **7**)

A solution of ethylmagnesium bromide, prepared in dry tetrahydrofuran (300 ml) from magnesium (4.00 g) and ethyl bromide (20 ml), was added dropwise to tetrahydrofuran (600 ml) saturated with acetylene at room temperature. A stream of acetylene was passed through the solution throughout the addition, and for 1 h afterwards. To the resultant solution of ethynylmagnesium bromide was added slowly, with stirring, a solution of freshly prepared, freshly distilled 2,3:4,5-di-*O*-isopropylidene-*aldehydo*-L-arabinose[8] (**1**, 22.0 g) in tetrahydrofuran (100 ml). A slow stream of acetylene was passed through the solution throughout the addition, and subsequently for a further 3 h. The solution was concentrated to 300 ml, ether (300 ml) was added, and the solution was washed at 0° with three 400-ml portions of 10% aqueous ammonium chloride, three 400-ml portions of water, and dried (magnesium sulfate). Evaporation of the solution gave the product as a syrup which crystallized partially upon refrigeration, yield 22.0 g (90%). This product contained none of the starting material, and was composed almost exclusively of the two 3-epimeric heptyne

derivatives, as revealed by t.l.c. (4:1 benzene–methanol) and g.l.c. (see below). A sample distilled at 0.1 mm Hg furnished the analytical sample.

Anal. Calc. for $C_{13}H_{20}O_5$: C, 60.92; H, 7.86. Found: C, 61.02; H, 7.88.

The n.m.r. spectrum of this product gave only those signals anticipated for a mixture of **2** and **7**. In five such experiments, the yields were in the range 85–92%.

Separation of the 3-epimers

The product from the preceding preparation was resolved by preparative g.l.c. on a Beckman GC2A gas chromatograph (Beckman Instruments, Inc., Fullerton, California) equipped with a thermal conductivity detector and a 10 ft × 5/8 in stainless-steel column packed with Carbowax* 20M (15%) on Chromosorb* W (60–100 mesh). The carrier gas was helium at an inlet pressure of 30 lb. in^{-2}, and samples (0.3–0.6 g in 2–3 ml of chloroform) were injected manually, with the column temperature initially at 200°, rising to 240° during 80 min. Two components were eluted, having retention times 122 and 136 min, in the weight ratio of 3:2. The components were completely separated from one another, and no other components were detected in the samples. The component eluted first, identified (by subsequent conversions) as 4,5:6,7-di-*O*-isopropylidene-1-heptyne-L-*gluco*-3,4,5,6,7-pentol (**2**), crystallized spontaneously; it had m.p. 67–69°; $[\alpha]_D^{22}$ +6 ±1° (*c* 3, chloroform)**; R_F 0.80 (4:1 benzene–methanol); λ_{max}^{KBr} 2.91 (OH), 3.11 (C≡CH), 4.74 (C≡C), 7.25, 7.31 μ (CMe_2); n.m.r. data (deuteriochloroform): τ 8.52, 8.57, 8.64 (3-, 6-, and 3-proton singlets, CMe_2), τ 7.50 (1-proton doublet, shifts to τ 7.68 at lower concentration, $J_{1,3}$ 2.3 c.p.s., H-1), τ 6.53 (1-proton broad doublet, shifts to higher field at lower concentration, OH), τ 5.80–6.10, 6.27–6.39 (multiplets, 5 protons, H-4,5,6,7,7'), τ 5.41 (1-proton multiplet, H-3); X-ray powder diffraction data: 10.22 s (2,2), 7.90 s (2,2), 5.73 w, 5.54 vs (1,1), 4.95 vs (1,1), 4.64 m (3,3), 3.97 m (4), 3.75 m (3,3), 3.24 w, 3.09 vw, 2.97 vw, 2.48 vw, 2.40 vw, 1.99 w.

Anal. Calc. for $C_{13}H_{20}O_5$: C, 60.92; H, 7.86. Found: C, 60.94; H, 7.94.

The product having retention time of 136 min, 4,5:6,7-di-*O*-isopropylidene-1-heptyne-L-*manno*-3,4,5,6,7-pentol (**7**) was obtained as a liquid, $[\alpha]_D^{22}$ −24 ±1° (*c* 2.3, chloroform)**; R_F 0.66 (4:1 benzene–methanol); λ_{max}^{KBr} 2.93 (OH), 3.06 (C≡CH), 4.72 (C≡C), 7.26, 7.32 μ (CMe_2); n.m.r. data (deuteriochloroform): τ 8.56, 8.65, 8.75 (singlets, 12 protons CMe_2), τ 7.50 (1-proton doublet, shifts to τ 7.67 at lower concentration, $J_{1,3}$ 2.3 c.p.s., H-1), τ 7.02 (1-proton broad singlet, shifts to higher field at lower concentration, OH), τ 5.76–6.11, 6.25–6.36 (multiplets, 5 protons, H-4,5,6,7,7'), τ 5.48 (1-proton multiplet, H-3).

Both products were homogeneous by t.l.c.

*3-O-Acetyl-4,5:6,7-di-O-isopropylidene-1-heptyne-*L-gluco(*and* L-manno)*-3,4,5,6,7-pentol* (**3** *and* **8**)

A solution of the mixed epimers (**2** and **7**, 9.1 g) in pyridine (20 ml) was treated

*Analabs, Inc., Hamden, Connecticut.

The signs of rotation of **2 and **7** were inadvertently reversed in Ref. 1.

with acetic anhydride (20 ml). After 18 h at room temperature, the mixture was poured into water, stirred for 30 min, and the product was extracted with dichloromethane. The extract was washed at 0° with N hydrochloric acid, water, dried (magnesium sulfate), evaporated, and the product distilled at 0.15 mm Hg to give the mixed epimeric 3-acetates (**3** and **8**), yield 8.14 g (76%); b.p./0.15 mm 108–109°; R_F 0.96 (4:1 benzene–methanol); λ_{max}^{film} 3.09 (C≡CH), 4.72 (C≡C), 5.72 μ (OAc).

Anal. Calc. for $C_{15}H_{22}O_6$: C, 60.39; H, 7.43. Found: C, 59.92; H, 7.74.

Acetylation of **2** under the above conditions gave the L-*gluco* epimer (**3**), R_F 0.96 (4:1 benzene–methanol); n.m.r. data (deuteriochloroform): τ 8.54, 8.58, 8.62 (6-, 3-, and 3-proton singlets, CMe_2), τ 7.87 (3-proton singlet, OAc), τ 7.49 (1-proton doublet, $J_{1,3}$ 2.2 c.p.s., H-1), τ 5.50–6.12 (5-proton multiplet, H-4,5,6,7,7'), τ 4.46 (1-proton quartet, $J_{3,4}$ 3.8 c.p.s., H-3). Acetylation of **7** gave the L-*manno* epimer (**8**), R_F 0.96 (4:1 benzene–methanol); n.m.r. data (deuteriochloroform): τ 8.53, 8.63, 8.72 (6-, 3-, and 3-proton singlets, CMe_2), τ 7.85 (3-proton singlet, OAc), τ 7.51 (1-proton doublet, $J_{1,3}$ 2.2 c.p.s., H-1), τ 5.45–6.15 (5-proton multiplet, H-4,5,6,7,7'), τ 4.36 (1-proton triplet, $J_{3,4}$ 2.4 c.p.s., H-3).

The mixture of **3** and **8** gave an n.m.r. spectrum corresponding to a 3:2 mixture of the two isomers.

*3-O-(3,5-Dinitrobenzoyl)-4,5:6,7-di-O-isopropylidene-1-heptyne-*L-manno*-3,4,5,6,7-pentol* (**9**)

To 3,5-dinitrobenzoyl chloride (20.5 g) in benzene (1 l) was added dropwise, with stirring at 0°, a solution of the mixed epimers **2** and **7** (15.3 g) in pyridine (50 ml). The mixture was kept 30 min at 0°, and 18 h at room temperature, 5 ml of water was added, and, after 1 h, ether (300 ml) was added. The solution was washed at 0° with water, N hydrochloric acid, 0.5N sodium hydroxide, and water, dried (magnesium sulfate), and evaporated, to give the mixed 3-epimeric 3-(3,5-dinitrobenzoates) (**4** and **9**) as a solid, yield 25 g (93.5%); m.p. 97–151°; R_F 0.82 (1:1 benzene–ether). Five recrystallizations from ethanol gave the pure L-*manno* epimer (**9**) as fine needles, yield 9.6 g (36%), m.p. 167.0–167.5°; $[\alpha]_D^{22}$ +2.9 ±0.8° (*c* 2, chloroform); R_F 0.82 (1:1 benzene–ether); λ_{max}^{KBr} 3.08 (C≡CH), 4.73 (C≡C), 5.76 (OAc), 6.14 (aryl), 6.46 (NO_2), 7.24, 7.28 (CMe_2), 7.44 μ (NO_2); n.m.r. data: τ 8.46, 8.50, 8.53, 8.62 (3-proton singlets, CMe_2), τ 7.28 (1-proton doublet, $J_{1,3}$ 2.3 c.p.s., H-1), τ 5.50–6.18 (5-proton multiplet, H-4,5,6,7,7'), τ 3.98 (1-proton triplet, $J_{3,4}$ 2.7 c.p.s., H-3), τ 0.60 (3 protons, aryl); X-ray powder diffraction data: 9.61 vw, 8.84 w, 8.12 w, 7.31 m, 6.37 s (6), 5.13 vs (2), 5.04 vs (1), 4.67 m, 4.52 vw, 4.21 s (5), 4.08 vw, 3.82 s (3,3) 3.63 s (3,3), 3.47 s (4), 3.38 vw, 3.18 vw, 3.01 w, 2.67 m, 2.55 w, 2.43 w, 2.35 vw.

Anal. Calc. for $C_{20}H_{22}N_2O_{10}$: C, 53.33; H, 4.92; N, 6.22. Found: C, 53.29; H, 5.03; N, 6.33.

A sample of **7**, which had been isolated by g.l.c. of the mixture of epimers, was acylated with 3,5-dinitrobenzoyl chloride by the foregoing procedure, to give **9**, m.p. 166.5–167.5°, identical with the product obtained from the mixed epimers, by mixed m.p., X-ray powder diffraction pattern, and i.r. and n.m.r. spectra.

3-O-(3,5-Dinitrobenzoyl)-4,5:6,7-di-O-isopropylidene-1-heptyne-L-gluco-*3,4,5,6,7-pentol* (**4**)

The mother liquors from the foregoing preparation from the mixed epimers (**2** and **7**) contained almost exclusively the L-*gluco* derivative (**4**), crude yield 15 g (56%), and it was obtained pure after several recrystallizations (from ethanol) to constant specific rotation; yield 4.16 g (15.5%); m.p. 134.5–135.5°; $[\alpha]_D^{22}$ −19.5 ±0.8° (*c* 2.1, chloroform); R_F 0.82 (1:1 benzene–ether); λ_{max}^{KBr} 3.11 (C≡CH), 4.71 (C≡C), 5.77 (C=O), 6.12 (aryl), 6.40 (NO_2), 7.23, 7.30 (CMe_2), 7.44 μ (NO_2); n.m.r. data: τ 8.46, 8.53, 8.62, 8.65 (3-proton singlets, CMe_2), τ 7.30 (1-proton doublet, $J_{1,3}$ 2.3 c.p.s., H-1), τ 5.48–6.19 (5-proton multiplet, H-4,5,6,7,7′), τ 4.10 (1-proton quartet, $J_{3,4}$ 4.0 c.p.s., H-3), τ 0.76 (3 protons, aryl); X-ray powder diffraction data: 10.69 m, 9.40 w, 8.08 m, 6.76 m, 5.59 vs (1), 5.04 w, 4.67 s (3,3), 4.35 s (3,3), 4.00 s (2), 3.76 vw, 3.65 vw, 3.44 w, 3.26 w, 3.12 vw, 2.91 vw, 2.83 vw.

Anal. Calc. for $C_{20}H_{22}N_2O_{10}$: C, 53.33; H, 4.92; N, 6.22. Found: C, 53.36; H, 5.06; N, 6.38.

A mixture of **4** and **9** melted unsharply over the range 114–125°.

3-O-Benzoyl-4,5:6,7-di-O-isopropylidene-1-heptyne-L-gluco (*and* L-manno)-*3,4,5,6,7-pentol* (**5** *and* **10**)

The heptyne derivatives **2** and **7**, which had been separated by g.l.c., were separately benzoylated. Each derivative (500 mg) in pyridine (10 ml) was treated with benzoyl chloride (0.5 ml), and the reaction mixture was processed by the procedure used for the 3,5-dinitrobenzoates, to give the 3-benzoates **5** and **10** as oils in essentially quantitative yield. The L-*gluco* isomer **5** had R_F 0.93 (4:1 benzene–methanol); n.m.r. data: τ 8.41, 8.49, 8.65, 8.68 (3-proton singlets, CMe_2), τ 7.27 (1-proton doublet, $J_{1,3}$ 2.3 c.p.s., H-1), τ 5.50–6.16 (5-proton multiplet, H-4,5,6,7,7′), τ 4.11 (1-proton quartet, $J_{3,4}$ 3.6 c.p.s., H-3), τ 1.91, 2.54 (multiplets, 5 protons, Ph). The L-*manno* isomer **10** had R_F 0.93 (4:1 benzene–methanol), λ_{max}^{film} 3,09 (C≡CH), 4.71 (C≡C), 5.80 (OBz), 7.25, 7.32 (CMe_2), 6.23 μ (aryl); n.m.r. data (deuteriochloroform): τ 8.52, 8.58, 8.68 (3-, 6-, and 3-proton singlets, CMe_2), τ 7.30 (1-proton doublet, $J_{1,3}$ 2.3 c.p.s., H-1), τ 5.58-6.06 (5-proton multiplet, H-4,5,6,7,7′), τ 4.10 (1-proton triplet, $J_{3,4}$ 2.9 c.p.s., H-3) τ 1.92, 2.56 (multiplets, 5 protons, Ph).

Anal. (for **10**): Calc. for $C_{20}H_{24}O_6$: C, 66.65; H, 6.71. Found: C, 66.61, H, 6.79.

4,5:6,7-Di-O-isopropylidene-1-heptene-L-gluco(*and* L-manno)-*3,4,5,6,7-pentol* (**6** *and* **11**)

To a solution of the mixed epimeric heptynes (**2** and **7**, 4.0 g) in cyclohexane (100 ml) and ethyl acetate (100 ml) was added freshly prepared Lindlar catalyst[9] (2 g) and quinoline (1 g). The mixture was shaken in an atmosphere of hydrogen at a pressure of 15 lb.in^{-2} until 1 mole/mole of hydrogen had been absorbed (35–40 min). The catalyst was filtered off, the filtrate was washed at 0° with N hydrochloric acid, and water, and the dried (magnesium sulfate) organic layer was evaporated to give the mixed 3-epimeric heptenes (**6** and **11**) as an oil; yield, essentially quantitative;

λ_{max}^{film} 2.91 (OH), 6.07 (CH_2=CH), 7.26, 7.32 μ (CMe_2); n.m.r. data: τ 8.60, 8.63, 8.68 (singlets, 12 protons, CMe_2), τ 5.67–6.25 (7-proton multiplet, H-3,4,5,6,7,7′, OH), τ 3.68–4.95 (3-proton multiplet, H-1,1′,2).

Anal. Calc. for $C_{13}H_{22}O_5$: C, 60.44; H, 8.58. Found: C, 60.39; H, 8.59.

When the time of reduction was increased, the n.m.r. spectrum showed appearance of a triplet, τ 8.82, and a quartet, τ 6.38, having coupling J 7 c.p.s. and intensity ratios characteristic of the ethyl group. Different batches of catalyst required different reaction times for reduction to the alkene without further reduction to the saturated derivative.

Reduction of 3-methyl-1-butyn-3-ol and 3-methyl-1-pentyn-3-ol to the corresponding alkenes

The alkyne derivatives were reduced over Lindlar catalyst, essentially by the procedure of the preceding experiment, to establish the optimum time of reduction for each batch of catalyst, and the extent of conversion was determined by n.m.r. spectroscopy. The butyne derivative, n.m.r. data (neat liquid): τ 8.48 (6-proton singlet, Me), τ 7.43 (1-proton singlet, H-1), τ 5.63 (1-proton singlet, OH), was converted into 3-methyl-1-buten-3-ol, n.m.r. data (neat liquid): τ 8.78 (6-proton singlet, Me), τ 6.05 (1-proton singlet, OH), τ 5.10 (1 proton, $J_{1,1'}$ 1.7 c.p.s., $J_{1,2}$ 10.0 c.p.s., H-1), τ 4.87 (1 proton $J_{1',2}$ 18.0 c.p.s., H-1′), τ 4.02 (1 proton, H-2). Similarly, the pentyne derivative, n.m.r. data (neat liquid): τ 8.98 (3-proton triplet, $J_{4,5}$ 7 c.p.s., H-5), τ 8.53 (3-proton singlet, 3-Me), τ 8.30 (2-proton multiplet, H-4), τ 7.48 (1-proton singlet, H-1), τ 6.03 (1-proton singlet, OH), was converted into 3-methyl-1-penten-3-ol, n.m.r. data (deuteriochloroform): τ 8.85 (3-proton triplet, $J_{4,5}$ 7 c.p.s., H-5), τ 8.48 (3-proton singlet, 3-Me), τ 8.01–8.61 (3 protons, H-4, OH), τ 4.55 (1 proton, $J_{1,1'}$ 1.7 c.p.s., $J_{1,2}$ 10 c.p.s., H-1), τ 4.51 (1 proton, $J_{1,2}$ 17.5 c.p.s., H-1′), τ 3.77 (1 proton, H-2).

Conversion of **2** *(or* **3***) into a glucose and* **7** *(or* **8***) into a mannose*

The crystalline heptyne derivative **2**, m.p. 67–69°, $[\alpha]_D$ +6° (40 mg) in cyclohexane (50 ml), ethyl acetate (50 ml), and quinoline (1 drop) was hydrogenated over Lindlar[9] catalyst (0.1 g) for 1 h under 15 lb.in^{-2} pressure. The mixture was filtered, and a stream of ozonized oxygen was passed through the filtrate for 1 h at room temperature. Adams catalyst (platinum oxide) (0.1 g) was added, the mixture was hydrogenated for 1 h under 15 lb.in^{-2} pressure, and filtered, and the filtrate was washed at 0° with N hydrochloric acid, and water, dried (magnesium sulfate), and evaporated. The product, which gave a positive Schiff test, was heated in 1% aqueous sulfuric acid (50 ml) for 3 h at 95°, the solution was neutralized with barium carbonate, filtered, and the filtrate was treated with a 1:1 mixture (20 ml) of Dowex-50 (H^+) and Dowex-1 (OH^-) ion-exchange resins. The solution was evaporated, and the residue was compared by paper chromatography with reference samples of D-glucose, D-mannose, and L-arabinose. The product contained a major component and a minor component, as revealed by aniline hydrogen phthalate[12] or silver nitrate–sodium

hydroxide[13]. The major component was indistinguishable from a glucose, the minor component was indistinguishable from an arabinose, and no component corresponding to a mannose was observed, in each of the following solvent systems: 4:1:5 butyl alcohol–ethanol–water (upper phase), 45:5:49:1 butyl alcohol–ethanol–water–ammonium hydroxide (upper phase), 6:4:3 butyl alcohol–pyridine–water, and 5:7.2:2:2 propyl alcohol–benzyl alcohol–85% formic acid–water.

When the procedure was repeated with the 3-acetate **3**, an essentially similar result was obtained, except that the proportion of glucose to arabinose in the final product was greater.

The same degradation was applied to the noncrystalline heptyne derivative **7**, $[\alpha]_D$ −24°. Paper chromatography, with the same systems as previously used, revealed that the product contained a component indistinguishable from a mannose, and a component corresponding to an arabinose. No component corresponding to a glucose was observed. Degradation of the corresponding 3-acetate (**8**) gave an essentially similar result, except that a greater proportion of mannose to arabinose was observed.

Conversion of **9** *into* L-*mannono-1,4-lactone* (**16**)

A solution of the 3,5-dinitrobenzoate **9**, m.p. 166.5–167.5°, (1.00 g) in methanol (10 ml) was treated with a catalytic amount of sodium methoxide for 2 h at room temperature, the precipitated methyl 3,5-dinitrobenzoate was removed, the filtrate was deionized by treatment with Dowex-50 (H^+) ion-exchange resin, and the solution was evaporated. The residue was heated with 60% aqueous acetic acid (50 ml) for 1 h at 95°, the solution was concentrated to 20 ml, and water was added to precipitate residual methyl 3,5-dinitrobenzoate. The mixture was filtered, and the filtrate was evaporated to give 1-heptyne-L-*manno*-3,4,5,6,7-pentol (**12**) as a white solid. The latter was dissolved in pyridine (10 ml), acetic anhydride (10 ml) was added, the mixture was kept overnight at room temperature, poured into ice and water, and the mixture was extracted with chloroform. The extract was washed successively at 0° with water, N hydrochloric acid, and aqueous sodium bicarbonate, and dried (magnesium sulfate). Evaporation of the solution gave L-*manno*-3,4,5,6,7-penta-acetoxy-1-heptyne (**13**) as a syrup; λ_{max}^{film} 3.09 (C≡CH), 4.72 (C≡C), 5.75 μ (OAc).

A stream of ozonized oxygen was passed through a solution of the foregoing product (**13**), in carbon tetrachloride (100 ml), for 6 h at 0°. Water (100 ml) was added, the mixture was evaporated until the carbon tetrachloride had been removed, and the remaining aqueous solution was maintained for 15 min at 60–70°. The strongly acidic solution was neutralized with sodium hydrogen carbonate, and washed with dichloromethane (200 ml), and the aqueous layer was acidified with N hydrochloric acid, and extracted with dichloromethane (200 ml). The organic extract was washed with water, dried (magnesium sulfate), and evaporated, to give penta-*O*-acetyl-L-mannonic acid (**14**), yield 372 mg (41% from **9**); λ_{max}^{film} 3.42, 3.92 (CO_2H), 5.77 μ (OAc, CO_2H).

A solution of **14** (185 mg) in methanol (10 ml) was treated with 3 molar equiva-

lents of sodium methoxide; a white precipitate formed. After 2 h at room temperature, the mixture was decationized by shaking with an excess of Dowex-50 (H^+) ion-exchange resin, whereupon the precipitate redissolved. Evaporation of the methanolic solution gave L-mannonic acid (**15**) as a partially crystallized syrup, yield 51 mg (~60%). This product (10 mg) in N hydrochloric acid (0.5 ml) was heated for 30 min at 95°, and the solution was evaporated, and examined by paper chromatography (3:1:3 ethyl acetate–acetic acid–water) and by t.l.c. with "Avicel-Technical Grade" microcrystalline cellulose[14] (4:1:5 butyl alcohol–ethanol–water, upper phase), with hydroxylamine–ferric chloride[15] as the detecting reagent. Reference samples of D-mannonic acid and D-gluconic acid were submitted to a similar acid treatment. The product gave a single component (on the chromatograms) which was indistinguishable from a mannono-1,4-lactone (**16**). The remainder of the L-mannonic acid (40 mg) was evaporated four times with propyl alcohol containing a trace of hydrochloric acid, and the crystalline L-mannono-1,4-lactone (**16**) was washed with propyl alcohol; yield 30 mg (37%); $[\alpha]_D^{22}$ *ca* −40° (30 min, water) [lit.[16] $[\alpha]_D$ −51.8° (water)]; X-ray powder diffraction data: 15.06 w, 8.96 vw, 6.97 vw, 5.88 m, 5.11 s (3), 4.57 s (4), 4.20 vs (1), 3.98 vw, 3.75 m (6), 3.54 m, 3.21 s (2), 2.87 s (5), 2.71 w, 2.56 m, 2.43 vw, 2.38 vw, 2.24 w, 2.14 w, 2.07 vw. A sample of D-mannono-1,4-lactone, m.p. 151°, gave an identical X-ray powder diffraction pattern.

Conversion of **4** *into* L-*gluconic acid phenylhydrazide* (**20**)

A solution of the 3,5-dinitrobenzoate **4**, m.p. 134.5–135.5°, (0.85 g) in 60% aqueous acetic acid (50 ml) was heated for 1 h at 95°, and then evaporated. The residue was dissolved in methanol (10 ml), a trace of sodium methoxide was added, and, after 2 h at 22°, the solution was filtered, decationized by shaking with Dowex-50 (H^+) resin, and evaporated, to give syrupy 1-heptyne-L-*gluco*-3,4,5,6,7-pentol (**17**). The latter was acetylated, the resultant pentaacetate (**18**) was ozonized, and the product was purified, essentially by the procedure used for conversion of **12** into **14**, to give 2,3,4,5,6-penta-*O*-acetyl-L-gluconic acid (**19**) as a partially crystallized syrup, yield 200 mg. A solution of **19** in N hydrochloric acid (10 ml) was heated for 2 h at 96°, and then evaporated. Chromatographic examination of the product, with the systems used for characterization of the lactone **16**, revealed two components, whose chromatographic properties were indistinguishable from those of D-glucono-1,4-lactone and D-glucono-1,5-lactone. Subjection of calcium D-gluconate to the hydrolytic conditions used with **19** gave a mixture of the two D-gluconolactones, chromatographically indistinguishable from the hydrolyzate from **19**. The mixture of L-gluconolactones from **19** (ca 50 mg) was heated with acetic acid (0.05 ml) and phenylhydrazine (0.05 ml) for 2 h at 96°, and the product was purified by chromatography on microcrystalline cellulose[14], to give crystalline L-gluconic acid phenylhydrazide (**20**), R_F 0.47 (4:1:5 butyl alcohol–ethanol–water, upper phase); X-ray powder diffraction data: 13.10 vs (1,1,1), 7.56 m, 5.41 vw, 5.04 vs (1,1,1), 4.55 vs (1,1,1), 4.32 m (3), 4.00 m (2), 3.77 vw, 3.55 vw, 3.39 m (4), 3.20 w, 3.09 vw, 2.70 w, 2.59 w, 2.25 w.

A sample of D-gluconic acid phenylhydrazide[17] had the same R_F value, and gave identical X-ray powder diffraction data.

In a second experiment, a stream of ozonized oxygen was passed for 6 h at 0° through a solution of the 3,5-dinitrobenzoate **4** (0.76 g) in carbon tetrachloride (100 ml). Water (100 ml) was added, the carbon tetrachloride was removed by evaporation, and the remaining aqueous solution was kept for 15 min at 60–70°. The solution was neutralized with sodium hydrogen carbonate, and washed with ether (200 ml), and the aqueous layer was acidified with N hydrochloric acid, and the product extracted with ether (2 × 200 ml). The extract was washed with water, dried (magnesium sulfate), and evaporated, and the residue was heated for 2 h at 95° with N hydrochloric acid (10 ml). The mixture was filtered, and the filtrate was evaporated to give a syrup, yield 250 mg (83%), which, by chromatography with the systems used for the lactone **14**, was shown to consist, almost exclusively, of the glucono-1,4(and 1,5)-lactones. The product could be converted into crystalline L-gluconic acid phenylhydrazide (**20**), identical in all respects with the product prepared by the preceding route.

ACKNOWLEDGMENTS

This work was supported by funds from the National Institutes of Health, Public Health Service, Department of Health, Education, and Welfare, Bethesda 14, Maryland; Grant No. GM-11976-01 (The Ohio State University Research Foundation Project 1820). Funds for purchase of the n.m.r. spectrometer were provided by the National Science Foundation, Washington, D.C. The authors thank Mr. J. B. Hughes and Mr. J. S. Jewell for the n.m.r. measurements.

SUMMARY

Ethynylation of 2,3:4,5-di-*O*-isopropylidene-*aldehydo*-L-arabinose (**1**) gave a high yield of the 3-epimeric heptyne derivatives **2** and **7**, separable by preparative g.l.c. or by fractional recrystallization of the derived 3,5-dinitrobenzoates **4** and **9**. Proof of configuration of **2** and **7** was effected by ozonolysis of the derived alkenes **6** and **11** to give derivatives of L-glucose and L-mannose, respectively. A second proof was afforded by conversion of **2** and **7** into the corresponding pentaacetoxyheptynes **18** and **13**, followed by ozonolysis to give derivatives of L-gluconic and L-mannonic acids, respectively. Differences in the n.m.r. spectra of the two series of heptyne derivatives were correlated with the difference in configuration at C-3.

REFERENCES

1 D. HORTON, J. B. HUGHES, AND J. M. J. TRONCHET, *Chem. Commun.* (London), (1965) 481.
2 W. REPPE, *Chemie des Acetylens und Kohlenoxyds*, Springer Verlag, Berlin, 1949; R. A. RAPHAEL, *Acetylenic Compounds in Organic Synthesis*, Academic Press Inc., New York, 1955.
3 R. LESPIEAU, *Advan. Carbohydrate Chem.*, 2 (1946) 107.

4 R. A. RAPHAEL, *J. Chem. Soc.*, (1949) S44.
5 R. A. RAPHAEL, *J. Chem. Soc.*, (1952) 401; R. A. RAPHAEL AND C. M. ROXBURGH, *J. Chem. Soc.*, (1952) 3875; M. M. FRASER AND R. A. RAPHAEL, *J. Chem. Soc.*, (1955) 4280.
6 I. IWAI AND K. TOMITA, *Chem. Pharm. Bull.* (Tokyo), 9 (1961) 976; 11 (1963) 184; I. IWAI, T. IWASHIGE, M. ASAI, K. TOMITA, T. HIROAKA, AND J. IDE, *Chem. Pharm. Bull.* (Tokyo), 11 (1963) 188; I. IWAI, *Kagaku No Ryoiki*, 18 (1964) 307.
7 D. PAPA, F. J. VILLANI, AND H. F. GINSBERG, *Arch. Biochem. Biophys.*, 33 (1951) 482; R. G. BUSNEL, A. LEHMANN, AND M. C. BUSNEL, *Pathol. Biol., Semaine Hop.*, 6 (1958) 749.
8 J. ENGLISH AND P. M. GRISWOLD, *J. Am. Chem. Soc.*, 67 (1945) 2039; H. ZINNER, E. WITTENBURG, AND G. REMBARZ, *Chem. Ber.*, 92 (1959) 1614.
9 H. LINDLAR, *Helv. Chim. Acta*, 35 (1952) 446.
10 C. D. HURD AND R. E. CHRIST, *J. Org. Chem.*, 1 (1936) 141.
11 D. HORTON AND J. B. HUGHES, to be published; *cf.*, D. HORTON AND MARTHA J. MILLER, *J. Org. Chem.*, 30 (1965) 2457.
12 S. M. PARTRIDGE, *Nature*, 164 (1949) 443.
13 W. E. TREVELYAN, D. P. PROCTER, AND J. S. HARRISON, *Nature*, 166 (1950) 444.
14 M. L. WOLFROM, D. L. PATIN, AND ROSA M. DE LEDERKREMER, *J. Chromatog.*, 17 (1965) 488.
15 M. ABDEL AKHER AND F. SMITH, *J. Am. Chem. Soc.*, 73 (1951) 5859.
16 F. W. UPSON, L. SANDS, AND C. H. WHITNAH, *J. Am. Chem. Soc.*, 50 (1928) 519.
17 E. FISCHER AND F. PASSMORE, *Ber.*, 22 (1889) 2728.