An enol glucoside of acetoacetic ester has been proposed as an intermediate in the formation of the product. *trans*-O-(β-D-Glucopyranosyl) methyl acetoacetate (III) resisted rearrangement on refluxing with zinc chloride in methanol, a fact which indicates that a derivative of this type is probably not an intermediate in the formation of XXX.

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[CONTRIBUTION FROM THE DEPARTMENT OF BIOCHEMISTRY, UNIVERSITY OF WISCONSIN]

Reductive Cleavage of Benzyl Glycosides for Relating Anomeric Configurations. Preparation of Some New Benzyl Pentosides[1]

BY CLINTON E. BALLOU, SAUL ROSEMAN[2] AND KARL PAUL LINK

Conditions for the reductive cleavage of acetylated benzyl glycosides which permit isolation of the unmutarotated 1-hydroxy acetylated aldoses are given. The reduction is effected with palladium and hydrogen in a neutral inert solvent (ethyl ether). Cleavages of benzyl α-D-xylopyranoside triacetate to 2,3,4-triacetyl-α-D-xylopyranose, and of benzyl β-D-xylopyranoside triacetate to 2,3,4-triacetyl-β-D-xylopyranose were accomplished with isolation of the products in a pure, crystalline form. Similar conversions were made with the D-glucopyranosides as well as with D- and L-arabinopyranosides. The products when crystalline were obtained in a yield of 70 to 90%, and in a high state of purity.

These transformations have a special significance with respect to the classical enzymatic experiments of Armstrong used in relating anomeric configurations of methyl D-glucosides and D-glucose. The possibility of inversion of configuration during enzymatic hydrolysis has never been disproved. However, the reductive cleavage of benzyl glycosides offers no opportunity for inversion, and relates anomeric configurations of the acetylated benzyl glycosides and the 1-hydroxy acetylated aldoses in a conclusive manner. This chemical conversion affords a complete substantiation of the classification of anomeric forms of the acetylated glycosides and the corresponding polyacetyl 1-hydroxy aldoses proposed by Hudson.

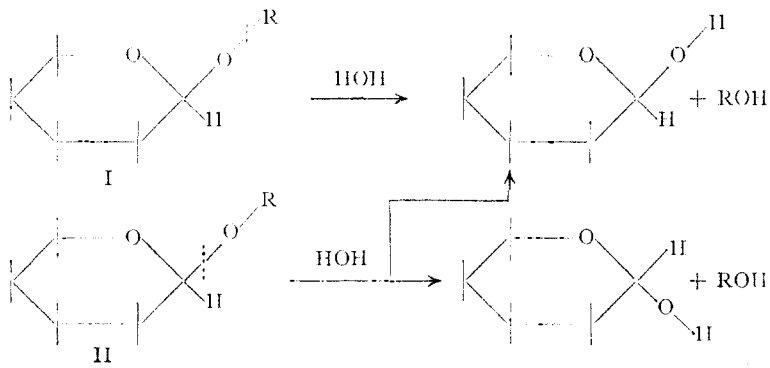
For the purpose of this investigation the following new pentosides were made: benzyl α- and β-D-xylopyranoside; benzyl α- and β-D-xylopyranoside triacetate; benzyl α- and β-D-arabinopyranoside; benzyl α- and β-D-arabinopyranoside triacetate; and benzyl α-L-arabinopyranoside, benzyl α-L-arabinopyranoside triacetate, and benzyl β-L-arabinopyranoside triacetate.

The classical experiments of Armstrong[3] on the enzymatic cleavage of methyl glucosides form an experimental cornerstone on which the anomeric configurations of the glucosides and free glucose are related. Armstrong presented evidence that methyl α-D-glucopyranoside and methyl β-D-glucopyranoside have the anomeric configurations, respectively, of α- and β-D-glucose.

The periodate oxidation studies of Jackson and Hudson[4] in 1937 extended this relationship to include other methyl glycosides. They established the anomeric configurations of several methyl aldohexopyranosides with respect to the two methyl D-glucopyranosides. Thus, if the evidence presented by Armstrong[3] be accepted without reserve, the anomeric configurations of the methyl hexopyranosides (and probably the glycosides in general) have been satisfactorily related to the α- and β-forms of D-glucose.

However, there is cause to reconsider the data presented by Armstrong.[3] Briefly described, he treated the two methyl D-glucopyranosides with the appropriate enzymes, and followed the hydrolyses polarimetrically. He remarked, "As a glucose of high initial rotatory power was obtained from α-methyl glucose, and one of low initial rotatory power from the β-glucoside, it is clear that α- and β-glucose correspond, respectively, to the α- and β-glucoside."

This conclusion is valid if one assumes that enzymatic cleavage occurs without inversion of anomeric configuration. The mechanism of enzymatic hydrolysis of the glycosidic linkage has not been established, but it is apparent that the possibility of inversion will depend on which bond is cleaved. Thus, inversion could occur only if the hydrolysis proceeds by reaction II. Recent studies



in this Laboratory on the alkaline methanolysis of certain alkali-sensitive glucosides[5,6,7] have revealed that methanolysis may occur on either side of the glucosidic oxygen, depending on the nature of the aglucon. The point of enzymatic cleavage might likewise vary depending on some similar but unknown factors, and the possibility of inversion might exist. Cohn[8] has demonstrated cleavages of both reaction types I and II on D-glucose-1-phosphate by phosphatases and phosphorylases. These occurred without apparent

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(2) Bobs Roberts Memorial Hospital for Children, The University of Chicago, Chicago, Illinois.

(3) Armstrong, *J. Chem. Soc.*, **83**, 1305 (1903).

(4) Jackson and Hudson, THIS JOURNAL, **59**, 994 (1937).

(5) Huebner, Karjala, Sullivan and Link, *ibid.*, **66**, 906 (1944).

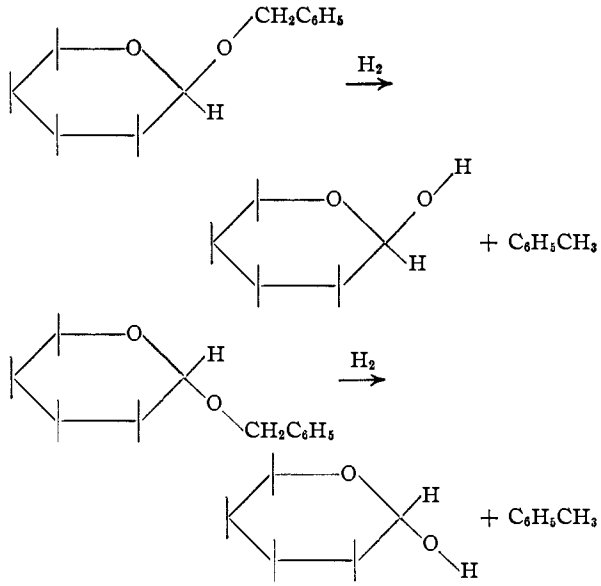
(6) Spero, Ballou and Link, *ibid.*, **71**, 3740 (1949).

(7) Ballou and Link, *ibid.*, **71**, 3743 (1949).

(8) Cohn, *J. Biol. Chem.*, **180**, 771 (1949).

inversion. The studies of Kuhn,[9] later corroborated by Freeman and Hopkins,[10] indicate that amylolysis of starch proceeds with the formation of α-maltose or β-maltose, respectively, depending on the enzyme used. Thus, inversion apparently occurs during cleavage by the β-amylases. In light of the above discussion, the need for more absolute methods for relating configurations is evident.

The object of this investigation was to adapt a well defined chemical reaction, the reductive cleavage of benzyl glycosides, to the purpose of relating anomeric configurations between the glycosides and the free sugars. This method of cleavage involves addition of hydrogen in such a manner that inversion of configuration of the anomeric carbon atom becomes unlikely. The products of cleavage would be toluene and the sugar of anomeric configuration identical with the starting benzyl glycoside. The sugar resulting from such a reaction



could be isolated, crystallized and characterized by chemical and physical means.

The experiments to be described offer independent evidence in complete accord with the relationships concluded by Armstrong.[3] The chemical interrelation of anomeric configurations was demonstrated with the benzyl glucosides and the benzyl xylo- and arabinosides. Because of the techniques employed, comparisons of the acetylated rather than the free sugars were made. This was due to the solubility characteristics of carbohydrates in non-polar solvents. However, present knowledge of the contributory nature to the molecular rotation of the anomeric carbon atom and that of the other asymmetric centers permits a relative comparison to be extended to the unacetylated glycosides and the free sugars.[11]

The reductive cleavage of benzyl glycosides originally reported by Freudenberg, Toepffer and Andersen[12] was subsequently studed at length by Richtmyer.[13] The conditions recommended involve use of palladium black and hydrogen in a solvent such as ethanol or acetic acid. Obviously these conditions would lead to mutarotation of the sugar, and its identifying configuration would be destroyed. A suitable solvent was not found that would dissolve the unacetylated glycosides, and the study could not be pursued on these derivatives. However, it was found that acetylated benzyl glycosides will undergo reductive cleavage with palladium and hydrogen in a neutral, inert solvent (absolute ethyl ether), in which subsequent mutarotation of the product is minimized. The use of the acetylated derivatives has the added advantage that the 1-hydroxy polyacetyl aldoses have good crystallizing properties. A palladium black catalyst similar to that previously used in cleaving benzyl glycosides[13] was found to be unsatisfactory in dry neutral ether. A very good catalyst for this purpose was that prepared from palladium chloride according to Mozingo (Method C).[14] The freshly reduced catalyst, washed with methanol and ethyl ether to remove traces of acid, had a high activity.

For this investigation, the new benzyl pentosides listed in the table were prepared. They were made by standard methods, and representative syntheses are given in the experimental part.

When benzyl α-D-xylopyranoside triacetate, $[\alpha]^{25}D$ + 142.0° (chloroform), is subjected to reductive cleavage there is isolated 2,3,4-triacetyl-α-D-xylopyranose, $[\alpha]^{25}D$ + 71.6° (chloroform), in a yield of 70%. The cleavage of benzyl β-D-xylopyranoside triacetate, $[\alpha]^{25}D$ − 86.7° (chloroform), results in the formation of 2,3,4-triacetyl-β-D-xylopyranose, $[\alpha]^{25}D$ − 22.4° (chloroform), the yield being 87%. The products crystallized spontaneously without seeding. The 2,3,4-triacetyl-α-D-xylopyranose, first reported by Hudson and Dale,[15] was formed on hydrolysis of triacetyl-D-xylopyranosyl bromide. They reported a specific rotation in chloroform of +70.4° which slowly mutarotated to a final value of +40.8°. The β-form is reported here for the first time, and has a specific rotation of −22.4° in chloroform which changes to a final reading of +40.7°. Thus, both 2,3,4-triacetyl-D-xylopyranoses are now known, and they have been related chemically to the benzyl D-xylopyranoside triacetates. The results obtained in the reductive cleavage of these xylosides are consistent with the original observations of Armstrong[3] described above. That is, the levorotatory benzyl D-xylopyranoside triacetate gives the levorotatory triacetyl-D-xylopyranose, while the dextrorotatory benzyl D-xylopyranoside triacetate gives dextrorotatory triacetyl-β-xylopyranose.

Furthermore, the anomeric relationships concluded for the benzyl xylopyranosides and xylopyranoses in this study may be correlated with those assigned according to the isorotation rules of Hudson.[16] The complete agreement substantiates the classifications of anomeric forms of the

(9) Kuhn, *Ann.*, **443**, 1 (1925).
(10) Freeman and Hopkins, *Biochem. J.*, **30**, 451 (1936).
(11) Bates, "Polarimetry, Saccharimetry and the Sugars," Circular C440 of the Natl. Bur. of Standards, p. 432 (1942).
(12) Freudenberg, Toepffer and Andersen, *Ber.*, **61**, 1750 (1928).
(13) Richtmyer, THIS JOURNAL, **56**, 1633 (1934).
(14) Mozingo, *Org. Syntheses*, **26**, 77 (1946).
(15) Hudson and Dale, THIS JOURNAL, **40**, 997 (1918).
(16) Hudson, *ibid.*, **31**, 66 (1909).

acetylated glycosides and the corresponding polyacetyl 1-hydroxy aldoses which were proposed by Hudson in 1909[16] and in subsequent years.

Benzyl β-D-glucopyranoside tetraacetate was cleaved to 2,3,4,6-tetraacetyl-β-D-glucopyranose in a good yield. Benzyl α-D-glucopyranoside tetraacetate cleaved with difficulty, and the product was presumably a sirupy mixture of 2,3,4,6-tetraacetyl-α-D-glucopyranose and uncleaved glucoside. The great difference in the rate of cleavage of the α- and β-forms of the glucoside is noteworthy. A similar, though less exaggerated, difference was observed with the xylosides. The observation is reminiscent of the rate difference found in acid hydrolysis of glycosides that was correlated with the relative configurations of carbon atoms 1 and 3.[17] One might suspect group interaction in the acetylated α-forms, since the substituents of carbon atoms 1 and 2 are *cis*. As these compounds deacetylated normally, the association would have to be of an unstable type.

Cleavage of the benzyl arabinoside triacetates produced the same conclusive results. Benzyl α-D-arabinopyranoside triacetate gave the new 2,3,4-triacetyl-α-D-arabinopyranose in a yield of 80%, while benzyl β-D-arabinopyranoside triacetate was cleaved to a levorotatory sirup which mutarotated upward. It was not obtained crystalline. Benzyl α-L-arabinopyranoside triacetate underwent cleavage to 2,3,4-triacetyl-α-L-arabinopyranose. Though but one form of the triacetyl arabinoses is known in a crystalline state, it is safe to assign the α-configuration because the products arose from the α-glycosides in a manner free from possible inversion.

It was observed that the two anomeric forms of the benzyl arabinosides also differ in the rate of cleavage, although in these compounds the α-form cleaves most readily. Thus, the acetylated benzyl glucoside, xyloside and arabinoside in which the aglycon and 2-acetoxy groups are *trans* cleave much more easily than those with a *cis* relationship.

The excellent yields of pure products recommends the above described procedure as a method for the preparation of 1-hydroxy polyacetyl aldoses of defined anomeric configuration. The possibility of its extension to the ketoses, both as a method of relating configurations and for preparing the corresponding derivatives, would appear to be promising.

Experimental

Preparation of Benzyl Glycosides.—Benzyl β-D-xylopyranoside triacetate, benzyl α-D- and α-L-arabinopyranoside triacetate and benzyl β-D-glucopyranoside tetraacetate were prepared from the respective acetylated glycosyl bromides according to the directions of Fischer and Helferich.[18]

Benzyl α-D-glucopyranoside tetraacetate was prepared from tetraacetyl-D-glucopyranosyl iodide as directed by Helferich and Gootz[19] and also by the action of titanium tetrachloride on benzyl β-D-glucopyranoside tetraacetate.[20]

Benzyl α-D-xylopyranoside and benzyl β-D-arabinopyranoside were made by the classical Fischer and Beensch glycoside synthesis[21] through the action of benzyl alcohol and hydrogen chloride on the respective pentoses. The xyloside was acetylated with acetic anhydride and fused sodium acetate in the usual manner[22] to give benzyl α-D-xylopyranoside triacetate, a colorless sirup that did not crystallize. The arabinoside was acetylated with pyridine and acetic anhydride at 0°[23] to give crystalline benzyl β-D-arabinopyranoside triacetate.

In repeating the synthesis of benzyl β-L-arabinopyranoside by treating L-arabinose in benzyl alcohol with hydrogen chloride,[21] the reaction mixture was shaken for about 24 hours. By this time, the arabinoside crystallized spontaneously and formed a solid cake in the flask. It was collected and washed with ether, giving twice the yield reported. This product was acetylated by the procedure used for acetylation of benzyl β-D-arabinopyranoside.

Attempted acetylation of the L-arabinoside with hot acetic anhydride and zinc chloride produced hexaacetyl-L-arabinose, m.p. 90°, a transformation previously noted in acetylations of arabinosides employing acetic anhydride and sulfuric acid.[24]

Benzyl β-D-xylopyranoside, benzyl α-D-arabinopyranoside and benzyl α-L-arabinopyranoside were prepared by deacetylation of the respective acetylated derivatives by the catalytic barium methoxide method.[25] Analyses of the new benzyl pentosides are listed in Table I.

Preparation of Benzyl α-D-Xylopyranoside Triacetate.—A mixture of 50.0 g. of D-xylose in 250 ml. of benzyl alcohol was cooled in an ice–salt-bath and saturated with dry hydrogen chloride gas. The mixture was shaken for 24 hours at room temperature, after which a dark solution was obtained. This was poured into 500 ml. of water and solid barium carbonate was added to neutralize the acid. The solid material was removed by filtration, and the excess benzyl alcohol in the filtrate separated from the water layer by ether extraction. The water layer was concentrated to dryness *in vacuo* at 50° and the mixture of salt and sirup extracted three times with absolute ethanol. The alcohol layer was then concentrated to a thick sirup, which was dissolved in 40 ml. of absolute ethanol and left at 5° in the refrigerator. Crystals formed readily. The product was collected and washed with ether, yield 10 g. It was recrystallized by dissolving it in warm absolute ethanol and adding ether to turbidity. The needles had a m.p. 127–128.5° and $[\alpha]^{25}D$ +139.2° (*c*, 4, water). It analyzed correctly for benzyl α-D-xylopyranoside.

Acetylation of this product was carried out as follows. To a mixture of 100 ml. of acetic anhydride and 5 g. of fused sodium acetate was added 10 g. of benzyl α-D-xylopyranoside. The xyloside dissolved readily, and the mixture was warmed for 2 hours on a steam-bath. The solution was cooled and poured into 100 ml. of ice-water which was then stirred for 4 hours. The water was changed several times by decanting it from the heavy sirup. This colorless sirup was dissolved in 50 ml. of ethanol, filtered and poured slowly into 300 ml. of water with rapid stirring. The product, a thick sirup, was collected and dried in a desiccator over phosphorus pentoxide. It had $[\alpha]^{25}D$ +142° (*c*, 3, chloroform), and gave the correct acetyl analysis for benzyl α-D-xylopyranoside triacetate.

Preparation of Benzyl β-D-Xylopyranoside Triacetate.—To 600 ml. of dry ether in a liter bottle were added 50.0 g. of triacetyl-D-xylopyranosyl bromide, 240 g. of benzyl alcohol and 100 g. of drierite. The mixture was shaken for 15 minutes, and then 32 g. of silver oxide was added. The shaking was continued for 3 hours, when a negative bromide ion test was obtained. The mixture was filtered, the ether removed from the filtrate by distillation and the excess benzyl alcohol by steam distillation. A thin colorless sirup was obtained which was crystallized from 50% ethanol–water. The small hexagonal plates, yield 27 g., melted at 90–92°, and upon recrystallization from the same solvent had m.p. 91.0–92.5°, $[\alpha]^{25}D$ −86.7° (*c*, 1, chloroform).

Preparation of Benzyl β-D-Arabinopyranoside Triacetate.—Benzyl β-D-arabinopyranoside was prepared by the same method used in making the α-D-xyloside. From 30 g. of D-arabinose was obtained 14 g. of the desired product. After five recrystallizations from water and two from ab-

(17) Isbell and Frush, *J. Research Natl. Bur. Standards*, **24**, 125 (1940).

(18) Fischer and Helferich, *Ann.*, **383**, 68 (1911).

(19) Helferich and Gootz, *Ber.*, **62**, 2788 (1929).

(20) Piel and Purves, THIS JOURNAL, **61**, 2978 (1939).

(21) Fischer and Beensch, *Ber.*, **27**, 2478 (1894).

(22) Erwig and Koenigs, *ibid.*, **22**, 2207 (1889).

(23) Behrend and Roth, *Ann.*, **331**, 362 (1904).

(24) Montgomery, Hann and Hudson, THIS JOURNAL, **59**, 1124 (1937).

(25) Isbell, *J. Research Natl. Bur. Standards*, **5**, 1185 (1930).

TABLE I

ANALYSES AND CONSTANTS OF BENZYL PENTOSIDES

Benzyl compounds	M. p., °C.	$[\alpha]^{25}D$	Recryst. from	Analyses, % Calcd. C	Calcd. H	Found C	Found H
α-D-Xylopyranoside triacetate	Sirup	+142.0 (*c*, 3, $CHCl_3$)	.....	Acetyl, 36.7		Acetyl, 36.1	
β-D-Xylopyranoside triacetate	91–92.5	− 86.7 (*c*, 1, $CHCl_3$)	50% EtOH–H_2O	59.00	6.06	58.90	6.05
α-D-Arabinopyranoside triacetate	80–81	+ 25.7 (*c*, 3, $CHCl_3$)	50% EtOH–H_2O	59.00	6.06	58.80	5.95
β-D-Arabinopyranoside triacetate	98–100	−200.5 (*c*, 3, $CHCl_3$)	50% EtOH–H_2O	59.00	6.06	58.90	6.19
α-L-Arabinopyranoside triacetate	79.5–81	− 24.4 (*c*, 3, $CHCl_3$)	95% EtOH	59.00	6.06	58.80	6.32
β-L-Arabinopyranoside triacetate	98–100	+200.5 (*c*, 3, $CHCl_3$)	50% EtOH–H_2O	59.00	6.06	58.75	6.11
α-D-Xylopyranoside	127–128.5	+139.2 (*c*, 4, H_2O)	Abs. EtOH–ether	60.00	6.66	59.75	6.82
β-D-Xylopyranoside	113–115	− 72.1 (*c*, 3 ,EtOH)	EtOAc	60.00	6.66	59.50	6.85
α-D-Arabinopyranoside[a]	138–140	+ 49.8 (*c*, 1, EtOH)	MeOH	60.00	6.66	59.90	6.70
β-D-Arabinopyranoside[a]	169–171	−212 (*c*, 0.5, H_2O)	H_2O	60.00	6.66	60.10	6.85
α-L-Arabinopyranoside[a]	138–140	− 44.6 (*c*, 1, EtOH)	MeOH	60.00	6.66	59.90	6.69

[a] We have recently learned that these compounds were prepared independently by Drs. H. G. Fletcher, Jr., and C. S. Hudson, National Institutes of Health, Bethesda, Maryland; and by Drs. L. J. Heidt and B. F. Van Tassel, Department of Chemistry, Massachusetts Institute of Technology, Cambridge, Massachusetts.

solute ethanol it melted at 169–171° and showed $[\alpha]^{26}D$ −212° (*c*, 0.5, water).

The benzyl β-D-arabinopyranoside was acetylated by the low temperature pyridine method.[23] A solution of 35 ml. of dry pyridine and 23 ml. of acetic anhydride was cooled in an ice-bath and 5 g. of the arabinoside added. The mixture was left in the refrigerator for 5 days. A colorless solution resulted. It was poured into ice-water and the mixture was stirred for several hours, the water being changed three times. The thick sirup was crystallized from 50% ethanol–water. A yield of 6.2 g. of oblong plates crystallizing in rosettes was realized. Benzyl β-D-arabinopyranoside triacetate melts at 98–100° and shows $[\alpha]^{25}D$ −200.5° (*c*, 3, chloroform).

Reductive Cleavage of Benzyl β-D-Xylopyranoside Triacetate.—Two grams of the palladium catalyst (5% palladium chloride on activated carbon) suspended in 100 ml. of absolute methanol was reduced by shaking with hydrogen at atmospheric pressure in a U-shaped hydrogenation chamber, similar to one shown by Weygand.[26] One end terminated in a 24/40 standard taper inner joint to which could be fitted a coarse sintered glass funnel sealed to a 24/40 standard taper outer joint. The methanol was drawn off through the funnel. The catalyst was resuspended in a small amount of solvent and again the solvent was removed. In this manner, the catalyst was washed three times with absolute methanol and three times with dry ethyl ether. It was finally suspended in 200 ml. of dry ethyl ether, and the mixture was shaken with hydrogen until a constant volume was maintained.

The benzyl β-D-xylopyranoside triacetate (5 g.) was then introduced into the hydrogenation chamber. The hydrogen uptake started immediately and was complete in 25 minutes with the absorption of the theoretical amount (360 ml., 25°, 740 mm.). This reaction mixture had a specific rotation of −24.8° (chloroform) calculated on the basis of triacetylxylose. The catalyst was removed and the filtrate concentrated on a steam-bath until crystallization began. The mixture was set in the refrigerator at 5° for 12 hours, and the product was then collected. The yield was 3.3 g. (87%) and after recrystallization from ether the product melted at 136–137°.

Anal. Calcd. for $C_{11}H_{16}O_8$: C, 47.80; H, 5.84. Found: C, 47.60; H, 5.93.

This 2,3,4-triacetyl-β-D-xylopyranose exhibited no detectable mutarotation in pure, neutral chloroform. The rotation 5 minutes after solution was $[\alpha]^{25}D$ −22.4° (*c*, 1, chloroform). After addition of a trace of acid to catalyze mutarotation the value changed, and in a few hours became constant at +40.7°. Another 2,3,4-triacetyl-D-xylopyranose reported by Hudson and Dale[15] had m.p. 138–141° and $[\alpha]^{25}D$ +70.4° (*c*, 2, U.S.P. chloroform), which changed on standing to +40.8°.

Reductive Cleavage of Benzyl α-D-Xylopyranoside Triacetate.—One gram of the sirupy benzyl α-D-xylopyranoside triacetate was cleaved as described above. The reduction went at about one-fourth the rate of that of the β-form. The crude reaction product had a specific rotation of +72.5° (chloroform) calculated as the triacetyl xylose.

Upon concentration to dryness, the product crystallized spontaneously. It was recrystallized from dry ethyl ether, the yield being 0.55 g. (70%). The melting point was 135–138°, and changed to 137–140° upon several recrystallizations from dry ether.

The specific rotation at 25° of this 2,3,4-triacetyl-α-D-xylopyranose was +71.6° (*c*, 2, chloroform), and it exhibited no detectable mutarotation in pure neutral chloroform over a period of 24 hours. Upon addition of a trace of hydrochloric acid, the rotation changed in 1 hour to +40.6° (chloroform). These constants compare well with those reported by Hudson and Dale.[15]

Reductive Cleavage of Benzyl D-Glucopyranoside Tetraacetates.—The cleavage of benzyl β-D-glucopyranoside tetraacetate went rapidly under the conditions used above, and was complete in about 3 minutes. The specific rotation of the reaction mixture calculated as tetraacetyl glucose was +12° (chloroform), indicating about 10% mutarotation of the product. The yield of 2,3,4,6-tetraacetyl-β-D-glucopyranose, crystallized from ether, was 0.65 g. from 1.0 g. of the starting glucoside (80%). It melted at 122–124°, and had $[\alpha]^{25}D$ +3.3° (*c*, 5, ethanol). The constants reported for 2,3,4,6-tetraacetyl-β-D-glucopyranose are m.p. 120°, $[\alpha]^{20}D$ +2.2° (*c*, 4, ethanol) changing to +82.7°.[27]

Cleavage of benzyl α-D-glucopyranoside tetraacetate could not be carried out successfully with the isolation of a crystalline product. In a reaction analogous to the one just described, the theoretical uptake of hydrogen was complete only after 8 hours. The sirup obtained had a specific rotation of +129° (chloroform), calculated as tetraacetyl glucose, and mutarotated downward to +105°. It reduced hot Fehling solution readily. The fact of mutarotation and its direction indicates formation of some 2,3,4,6-tetraacetyl-α-D-glucopyranose, $[\alpha]^{20}D$ +139° (*c*, 1, chloroform), but the final value when the rotation became constant indicates a mixture of the starting glucoside and the desired product in the proportion 1:1. The slow cleavage probably results in partial reduction of the aromatic ring,[13] giving a side product which does not undergo reductive cleavage. When 5 g. of catalyst to 1 g. of glucoside was used, and the reduction carried out at 0° to further minimize mutarotation, the results were similar although the uptake of hydrogen proceeded at a faster rate.

Reductive Cleavage of Benzyl Arabinopyranoside Triacetates.—Benzyl α-D-arabinopyranoside triacetate (2 g.) cleaved readily, and was complete in 5 minutes. The sirup showed $[\alpha]^{26}D$ −48.1° (chloroform) calculated as triacetyl arabinose. It was crystallized from an ethyl ether–petroleum ether mixture, and recrystallized from ethyl ether. This 2,3,4-triacetyl-α-D-arabinopyranose obtained as rectangular plates in a yield of 1.2 g. (77%), melted at 105–108° and showed $[\alpha]^{26}D$ −47.1° (*c*, 2, chloroform). Upon addition of a trace of acid it mutarotated to a final value of −109°.

(26) Weygand, "Organic Preparations," Interscience Publishers, New York, N. Y., 1945, p. 11.

(27) Schlubach and Wolf, *Ber.*, **62**, 1507 (1929).

Anal. Calcd. for $C_{11}H_{16}O_8$: C, 47.80; H, 5.84. Found: C, 47.80; H, 5.87.

Benzyl β-D-arabinopyranoside triacetate cleaved at a slower rate than the α-form and was complete in 40 minutes. The sirup, impure 2,3,4-triacetyl-α-D-arabinopyranose, showed $[\alpha]^{25}D$ −123.5° (chloroform) calculated as triacetyl arabinose, and mutarotated to a constant value of −107.2°. It has not been crystallized.

Benzyl α-L-arabinopyranoside triacetate responded to reductive cleavage in a manner similar to the α-D form to give a 60% yield of 2,3,4-triacetyl-α-L-arabinopyranose melting at 105–108° and showing $[\alpha]^{25}D$ +50.1° (*c*, 2, chloroform).

Anal. Calcd. for $C_{11}H_{16}O_8$: C, 47.80; H, 5.84. Found: C, 47.95; H, 5.96.

A trace of acid causes mutarotation of this product in chloroform solution, and an equilibrium value $[\alpha]^{25}D$ +110.5° results.

MADISON 6, WISCONSIN RECEIVED JULY 5, 1950

[CONTRIBUTION FROM THE CHEMICAL RESEARCH DIVISION OF SCHERING CORPORATION]

Catalytic Hydrogenation of Cholesterol

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Strong acids have been shown to act as promoters for the low-pressure hydrogenation of cholesterol using Adams platinum oxide catalyst. When perchloric acid was used as the promoter, cholestanol was obtained in 87–90% yield. Cholestane, cholestanyl acetate and coprostanol were formed as by-products.

For many years the catalytic hydrogenation of cholesterol has been an outstanding example of a difficult and capricious hydrogenation. Despite the relative ease of reaction reported by W. F. Bruce,[1] in this Laboratory the authors found that the specified amount of platinum catalyst produced only a slight absorption of hydrogen. Even excessively large amounts of catalyst permitted only a slow and halting reaction. Neither careful crystallization of the commercial material to the highest recorded constants of melting point and optical rotation nor a pretreatment with Raney nickel[2,3] accelerated its rate of hydrogenation. This was in sharp contrast to the hydrogenation of sitosterol and stigmasterol which proceeded smoothly at room temperature in acetic acid solution and which indicated that the catalyst was not at fault. Even cholesterol resynthesized from 25-norcholestenolone[4] did not absorb hydrogen at an appreciable rate.

A pretreatment of the cholesterol dissolved in acetic acid and warmed on the steam-bath for 7 hours with a small amount of 30% hydrogen peroxide gave a product which would reduce reliably though slowly at room temperature with Adams catalyst, approximating the rate observed in the "Organic Syntheses" procedure.[1] Other oxidizing agents accomplished the same improvement though the process could still not be considered practical for the preparation of large amounts of cholestanol.

In the search for promoters for this reaction perchloric acid was found to have a powerful activating effect. So little of it was necessary to accelerate the rate of hydrogenation that it could not be considered to be an oxidizing agent. This interpretation was later substantiated by the observation that many other acids produced the same effect roughly parallel to their acid strength. Thus, sulfuric, maleic, oxalic, phosphoric, hydrochloric, *p*-toluenesulfonic and citric acids, all lower than *pK* 3, were effective promoters. Acetic acid (*pK* 4.76) and benzoic acid (*pK* 4.20) produced a slow and incomplete reaction while desoxycholic acid and catechol showed no promoter effect.

Using perchloric acid as an accelerator, it was no longer necessary to limit the hydrogenation to acetic acid as a solvent or to conduct the hydrogenation much above room temperature, though the heat evolved by the now rapid hydrogenation maintained the temperature of the resultant solution at 40–50°. Usually, the hydrogenation solution was warmed initially to 40–50° in order to dissolve and hydrogenate a greater amount of cholesterol in the hydrogenator at our disposal. The use of ethyl acetate, which is a good solvent for cholesterol, had the additional advantage that very little cholestanyl acetate was formed, whereas acetic acid in the presence of platinum catalyst caused considerable ester formation and it was then necessary to hydrolyze the entire product in order to obtain pure cholestanol.[1]

It is interesting to note that of the seven active promoters found, perchloric,[5] sulfuric,[6] phosphoric,[6] *p*-toluenesulfonic,[6] hydrochloric[7] and oxalic[8] acids are known to form addition products with cholesterol.

A procedure was developed in which a suspension of 1.25 kg. of cholesterol in 17 l. of ethyl acetate was hydrogenated completely in 30–45 min. with 25 g. of Adams platinum oxide catalyst using 1–2 ml. of 72% perchloric acid promoter. From 87 to 90% of the theoretical amount of cholestanol was obtained, and this product gave a negative Liebermann–Burchard reaction.

The best yield of cholestanol obtained in large scale hydrogenations using perchloric acid as a promoter was about 90% of the theoretical. A careful large scale chromatographic separation of the residues remaining after the crystallization of the cholestanol indicated the presence of small amounts of three other compounds: cholestanyl acetate, coprostanol and cholestane, all by-products

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(2) A. Wilds and C. Djerassi, THIS JOURNAL, **68**, 1712 (1946).

(3) J. R. Durland, Ph.D., Thesis, University of Wisconsin, 1939.

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(5) W. Lange, R. G. Folzenlogen and D. G. Kolp, *ibid.*, **71**, 1733 (1949).

(6) A. Ryer and W. Gebert, this Laboratory, unpublished work.

(7) U. S. Patent 2,322,906.

(8) L. Yoder, O. R. Sweeney and L. K. Arnold, *Ind. Eng. Chem.*, **37**, 374 (1945).