TABLE I

EFFECT OF *p*H ON THE RATE OF THE EXCHANGE REACTION AT 61°

*p*H	Buffer system	$t_{1/2}$, hr.
1.5	Oxalic acid	0.67
2.15	HCl-KCl	1.84
2.80	Phthalate	7.7
3.40	Acetate	10.0
5.0	Acetate	10.2
6.0	Phosphate	4.3
6.5	Phosphate	2.2
7.0	Phosphate	1.9
8.15	Barbiturate	0.83

Discussion

The effect of *p*H on the rate of the exchange reaction suggests that there are two independent reactions: one base catalyzed, the other acid. The rate of exchange is not linear with the hydrogen ion concentration in the acid range nor with the hydroxyl ion concentration in the alkaline region. The rate-limiting step for the exchange reaction is not the same as for mutarotation since it is less than one-thirtieth that calculated for the rate of mutarotation at 61°. At *p*H 7 the energy of activation of the exchange reaction calculated from the slope of the curve in Fig. 3 is 23,400 cal. while that for mutarotation is but 17,200 cal.[7] The kinetics as well as the different shapes of the rate *vs.* *p*H curves suggest that the rate-limiting steps of the mutarotation and the exchange are different. Our data are consistent with the hypothesis that the exchange takes place with the free aldehyde form of glucose.

Acknowledgment.—We are indebted to Dr. Israel Dostrovsky of the Weizmann Institute, Rehovoth, Israel, who supplied us with the O^{18}-labeled water and to Miss Laura Ponticorvo for her highly skilled assistance.

(7) Reference 6, p. 448.

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[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY AND CHEMICAL ENGINEERING, STANFORD UNIVERSITY]

Isomers of Tetra-*O*-acetyl-D-mannopyranose

BY WILLIAM A. BONNER

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When water and silver carbonate acted upon crystalline tetra-*O*-acetyl-α-D-mannopyranosyl chloride a levorotatory sirup resulted, from which two isomeric tetra-*O*-acetyl-D-mannopyranose derivatives could be crystallized: A, m.p. 124°, and B, m.p. 95°, respectively. When sirupy tetra-*O*-acetyl-α-D-mannopyranosyl bromide was similarly allowed to react isomers A and B were again obtained, accompanied by a third isomeric tetra-*O*-acetyl-D-mannose, C, m.p. 164°. Acetylation of isomer A with radioactive acetic anhydride and pyridine at −5° afforded an acetyl-labeled penta-*O*-acetyl-β-D-mannopyranose bearing 85% of its label at C-1, as indicated by its physical properties and the fact that it exchanged 85% of its radioactive acetyl groups under anomerizing conditions. Isomer A dextromutarotated rapidly in aqueous acetone, slowly in pyridine and rapidly in a pyridine–phenol mixture. Acetylation of isomer A with acetic anhydride in pyridine was markedly more rapid than its mutarotation in pyridine. Methylation of isomer A by several techniques afforded crystalline methyl tetra-*O*-acetyl-β-D-mannopyranoside. A study of the carbonyl absorption intensity of isomer A indicated four normal acetyl groups and no ortho-acid structure. These data are all explicable in terms of 2,3,4,6-tetra-*O*-acetyl-β-D-mannopyranose as the structure of isomer A. Acetylation of isomer B with radioactive acetic anhydride and pyridine produced penta-*O*-acetyl-α-D-mannopyranose acetyl-labeled at C-1, characterized again by its physical properties, its carbonyl absorption intensity and its loss of all of its label under anomerizing conditions. The structure 2,3,4,6-tetra-*O*-acetyl-α-D-mannopyranose is thus indicated for isomer B. Acetylation of isomer C with radioactive acetic anhydride yielded a labeled penta-*O*-acetyl-β-D-mannopyranose which *failed* to exchange its label under anomerizing conditions, a fact indicating that some hydroxyl group other than that at C-1 was free in this isomer. In aqueous acetone isomer C mutarotated to produce isomer B, a reaction clearly involving acetyl migration. Consideration of the known tetra-*O*-acetyl-D-mannopyranose derivatives in the literature, and of molecular models indicates that the structure of isomer C is best represented either by 1,3,4,6- or 1,2,4,6-tetra-*O*-acetyl-β-D-mannopyranose.

Introduction

Four tetraacetyl derivatives of mannose are described in the literature. Of these, only 1,2,3,4-tetra-*O*-acetyl-β-D-mannopyranose has been particularly well characterized.[1,2] It is obtained by the usual detritylation of its 6-trityl precursor with a cold hydrogen bromide–acetic acid mixture. Having m.p. 135.5–136.5° and $[\alpha]^{20}D$ −22.5° ($CHCl_3$), its structure rests on: (a) its method of preparation, (b) its conversion[1] to penta-*O*-acetyl-β-D-mannopyranose with acetic anhydride, (c) its conversion to the corresponding 6-chlorohydrin with phosphoryl chloride,[1] (d) its lack of mutarotation[1] and (e) its conversion[2] to 1,6-linked di- and trisaccharide derivatives on reaction with poly-*O*-acetylglycopyranosyl halides.

A second tetra-*O*-acetyl-D-mannose has been described[3] by Micheel and Micheel. This material, obtained in very low yield on reaction of trimethylamine with tetra-*O*-acetyl-α-D-mannopyranosyl bromide, had m.p. 159–160° and $[\alpha]^{19}D$ −24.2° ($CHCl_3$). No further physical or chemical properties of this substance have been described, although the authors suggest 2,3,4,6-tetra-*O*-acetyl-D-mannopyranose as its possible structure.

In an investigation concerned with the ring structure of mannose acetates, Levene and Tipson report[4] a third tetra-*O*-acetyl-D-mannose. This substance, m.p. 93° and $[\alpha]^{27}D$ +26.3° ($CHCl_3$) resulted by action of excess silver carbonate and slightly over the calculated quantity of water on crystalline tetra-*O*-acetyl-α-D-mannopyranosyl bro-

(1) B. Helferich and J. F. Leete, *Ber.*, **62**, 1549 (1929).
(2) D. D. Reynolds and W. L. Evans, THIS JOURNAL, **62**, 66 (1940).
(3) F. Micheel and H. Micheel, *Ber.*, **63**, 386 (1930).
(4) P. A. Levene and R. S. Tipson, *J. Biol. Chem.*, **90**, 89 (1931).

mide. On acetylation with "cold" acetic anhydride in pyridine, Levene and Tipson converted their tetraacetate into penta-O-acetyl-β-D-mannopyranose, m.p. 117°, $[\alpha]^{27}D$ −24.1° ($CHCl_3$). More recently Gakhokidze and Kutidze have described[5] this same tetraacetyl derivative, m.p. 94°, $[\alpha]^{20}$ D +25.5° ($CHCl_3$), as obtainable in 72% yield from a similar reaction with tetra-O-acetyl-α-D-mannopyranosyl bromide. The latter investigators also report a 1,3,4,6-tetra-O-acetyl-D-mannose, m.p. 131°, from the action of silver acetate on 3,4,6-tri-O-acetyl-D-mannopyranosyl chloride. The structure of this isomer rests on its conversion to a 2-O-(2,3,4,6-tetra-O-acetyl-D-mannosyl)-1,3,4,-6-tetra-O-acetyl-D-mannose derivative, whose structure is in turn supported by appropriate degradative reactions. Its anomeric configuration is, however, not established.

Recently a need for the anomeric penta-O-acetyl-D-mannopyranoses labeled in the C-1 acetoxy group with carbon-14 has prompted us to undertake the preparation and acetylation of several tetra-O-acetyl derivatives of D-mannose. The isolation and structure proof of these is the subject of the present communication.

Results and Discussion

Unable to obtain crystalline tetra-O-acetyl-α-D-mannopyranosyl bromide by action of hydrogen bromide in acetic acid on a sirupy mixture of the anomeric penta-O-acetyl-D-mannopyranoses, we found that the latter mixture readily yielded crystalline tetra-O-acetyl-α-D-mannopyranosyl chloride on treatment with titanium tetrachloride in chloroform.[6] When this chloride reacted with one equivalent of water in the presence of silver carbonate in anhydrous ether, the resulting crude product was a levorotatory sirup. From an ether solution of this sirup was readily obtained a tetra-O-acetyl-D-mannose (isomer A) having m.p. 124–124.5°. Evaporation of the mother liquors from isomer A yielded a sirup which gradually crystallized as huge prisms. This second tetra-O-acetyl-D-mannose had m.p. 95–96° and is discussed below as isomer B.

Since isomer A has not been described before, and since B proved (*vide infra*) to show unexpected behavior during acetylation, we attempted to repeat the above preparations starting with tetra-O-acetyl-α-D-mannopyranosyl bromide. Failing to obtain a crystalline sample, we subjected the crude sirupy bromide to reaction conditions identical to those used with the chloride above. The crude reaction product was a slightly dextrorotatory sirup. An ether solution of this sirup yielded initially a very small quantity of a third isomeric tetra-O-acetyl-D-mannose, isomer C, m.p. 164–165°. From the mother liquors of the above substance A and B were readily recoverable. The physical properties of the three products obtained are summarized in Table I. Chemical properties of each isomer, leading to conclusions regarding the structure of each, are discussed individually below.

Isomer A.—Acetylation of isomer A with chilled (−5°) acetic anhydride and pyridine, conditions generally assumed[7] to engender no significant anomerization or structural rearrangement in the carbohydrate series, led to almost optically pure penta-O-acetyl-β-D-mannopyranose, pure after one recrystallization. This fact clearly indicates that isomer A is a tetra-O-acetyl-β-D-mannopyranose, but is not conclusive regarding the location of its unacetylated hydroxyl group. Evidence on this question was secured by similar acetylation with radioactive acetic anhydride, yielding a penta-O-acetyl-β-D-mannopyranose labeled with carbon-14 in the acetyl group corresponding to the free -OH position. When placed in a 1:1 mixture of acetic anhydride and acetic acid, 0.5 *M* in sulfuric acid catalyst, conditions which result in anomerization of polyacetylglycose derivatives, this radioactive penta-O-acetyl-β-D-mannopyranose not only anomerized but lost its radioactivity according to the first-order kinetics law, with $k_{exchange} = 0.0333 \pm 0.0018$ min.$^{-1}$. We have shown previously by product analysis experiments and by infrared spectrophotometry,[8] and Lemieux has more recently confirmed by product analysis[9] and radiochemical isotope dilution techniques,[10] that under such anomerizing conditions acetyl exchange occurs specifically at the anomeric center, and at no other carbon atom in the acetylated aldopyranose molecule. Thus the occurrence of radioactive acetyl exchange under anomerizing conditions in the present labeled penta-O-acetyl-β-D-mannopyranose indicates clearly that the C-1 position thereof possessed the labeled acetyl group, and suggests strongly that the C-1 hydroxyl group must have been free in its tetraacetate precursor. The tentative conclusion follows that isomer A possesses the structure 2,3,-4,6-tetra-O-acetyl-β-D-mannose (I).

TABLE I

PHYSICAL PROPERTIES OF ISOMERIC TETRA-O-ACETYL-D-MANNOSE DERIVATIVES

Isomer	M.p. °C.	$[\alpha]_D^{25}$ ($CHCl_3$)	Concn., g./100 ml.
A	124–124.5	−13.6°	3.1
B	95–96	+23.1	4.6
C	164–165	−25.2	0.5

The validity of this conclusion rests, of course, on the correctness of the assumption that little or no acetyl migration attended the acetylation of isomer A with radioactive acetic anhydride in pyridine at −5°. If significant acetyl migration occurred, the above conclusion would be invalid. Thus, for example, II, with its C-2 hydroxyl free could yield C-1 acetyl labeled penta-O-acetyl-β-D-mannopyranose if the unlabeled acetyl at C-1 migrated totally to C-2 during acetylation in the cold. We have attempted to assess this possibility by investigating the extent of loss of labeled acetyl attending complete anomerization.

When the above labeled penta-O-acetyl-β-D-mannopyranose was allowed to anomerize completely to its equilibrium anomeric composition, the

(5) A. M. Gakhokidze and N. D. Kutidze, *J. Gen. Chem. U.S.S.R.*, **22**, 247 (1952); *C. A.*, **46**, 11117 (1952).

(6) E. Pacsu, *Ber.*, **61**, 1508 (1928).

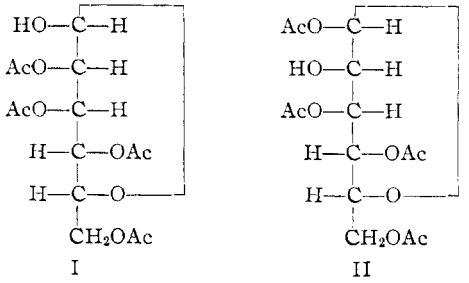
(7) W. W. Pigman and R. M. Goepp, Jr., "Chemistry of the Carbohydrates," Academic Press, Inc., New York, N. Y., 1948, p. 151; F. J. Bates *et al.*, "Polarimetry, Saccharimetry and the Sugars," Circ. C440, Nat. Bur. Standards, 1942, p. 486.

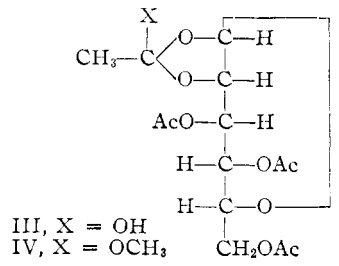
(8) W. A. Bonner, THIS JOURNAL, **73**, 2659 (1951).

(9) R. U. Lemieux and Carol Brice, *Can. J. Chem.*, **30**, 295 (1952).

(10) R. U. Lemieux, Carol Brice and G. Huber, *ibid.*, **33**, 134 (1955).

α-anomer isolated from the equilibrated mixture was found to possess 15% of the original radioactivity. Thus 15% of the labeled acetyl in the above radioactive pentaacetate derived from isomer A was located at some carbon other than C-1, indicating that a certain amount of acetyl migration had in fact occurred during acetylation of isomer A. We are thus confronted with two possibilities: (1) isomer A possesses structure I, and 15% of unlabeled acetyl migrated *toward* the C-1 OH during its acetylation with labeled acetic anhydride or (2) isomer A possesses some other structure, *e.g.*, II, and 85% of unlabeled acetyl migrated *away* from C-1 during acetylation.





A decision between structures I and II (or its equivalent) for isomer A can be made on the basis of comparative rates of acetylation and isomerization. In aqueous acetone isomer A underwent rapid, first-order dextromutarotation (k_A 0.144 min.$^{-1}$), reaching equilibrium in about 30 minutes. From the equilibrium mixture isomer B could be isolated readily. In pyridine isomer A showed gradual dextromutarotation (k'_A 0.0110 min.$^{-1}$), while in a 5:1 pyridine–phenol mixture the rate of this mutarotation was increased twenty-fold (k''_A = 0.218 min.$^{-1}$). These data are consistent with structure I in that it is well known[11,12] that 2,3,4,6-tetra-*O*-methyl-D-glucose, for example, anomerizes very slowly in pyridine but rapidly in pyridine–phenol mixtures, in accord with the need for general acid–base catalysis.[12] In a 1:1 mixture of pyridine and acetic anhydride at 25°, acetylation of isomer A was found to be substantially complete in 25 minutes, affording again almost optically pure penta-*O*-acetyl-β-D-mannopyranose. In contrast the above mutarotation of isomer A is only about 25% complete during 25 minutes in pyridine *lacking* acetic anhydride. Thus the acetylation of isomer A occurs considerably more rapidly than does its isomerization. Similarly, mutarotation of isomer A in pyridine is still only 80% complete after two days at −5°.

(11) T. M. Lowry, *J. Chem. Soc.*, **127**, 1383, 2883 (1925).

(12) C. G. Swain, *Record Chem. Progr.*, **12**, 24 (1951); THIS JOURNAL, **74**, 2534 (1952).

Incapable of simple anomerization under these conditions (penta-*O*-acetyl-β-D-mannopyranose, for example, is optically stable in both pyridine and aqueous acetone) any mutarotation by structure II must be due initially to acetyl migration. Furthermore, to permit the introduction of labeled acetyl at C-1 on acetylation of II, such migration would have to involve the transformation II → I. If isomer A had structure II, its acetylation under these conditions with radioactive acetic anhydride could therefore not afford a penta-*O*-acetyl-β-D-mannopyranose extensively labeled at C-1, since the mutarotation of isomer A (initially due under this hypothesis to its isomerization into I) is *slower* than its acetylation. Thus the 85% acetyl migration from C-1 to C-2 *prior to* acetylation, demanded of II by our above radiochemical data, is impossible and II (or its equivalent) is excluded as a structural possibility for isomer A. It should also be pointed out that any conceivable concerted process whereby C-1 to C-2 acetyl migration in II occurred synchronously with the introduction of a labeled C-1 acetoxy group would involve formation of penta-*O*-acetyl-α-D-mannopyranose, all of whose labeled acetyl should be exchangeable under anomerizing conditions, rather than the observed β-anomer.

A third alternative[13] for the structure of isomer A is III, possessing an acidic ortho-ester grouping. While structures such as III have been postulated[14,15] as intermediates during the alkali-catalyzed migrations of acetyl groups in partially acetylated aldoses, and have been reported several times as distinct substances,[14] the evidence for their discrete existence is by no means unambiguous, since the chemical behavior of supposed cyclic ortho-acids such as III can be equally well rationalized[16,17] in terms of structures such as I or II. Structure III, however, could explain the above results on acetylation of isomer A with radioactive acetic anhydride if the ortho-acid structure opened up in such a way as to permit 85% of the incoming acetyl groups to attach to the oxygen at C-1 and 15% to attach to the oxygen at C-2. This alternative, however, is definitely excluded by the following lines of evidence.

Table II indicates both the ratios of the "integrated areas" and of the molar extinction coefficients for the 5.75 μ ester carbonyl stretching band shown by isomer A, isomer B and several other D-mannose derivatives of interest for comparison purposes. Both the integrated areas under carbonyl absorption bands[18] and the molar extinction coefficients of such bands[16] have been used in past investigations to establish the number of ester groupings in molecules. Whereas there are no suitable chemical methods for distinguishing between partially acetylated carbohydrate esters such

(13) J. K. Dale, *ibid.*, **46**, 1046 (1924).

(14) W. W. Pigman and R. M. Goepp, Jr., "Chemistry of the Carbohydrates," Academic Press, Inc., New York, N. Y., 1948, p. 159.

(15) E. Pacsu in W. W. Pigman and M. L. Wolfrom, "Advances in Carbohydrate Chemistry," Vol. I, Academic Press, Inc., New York, N. Y., 1945, pp. 108 ff.

(16) R. K. Ness and H. G. Fletcher, Jr., THIS JOURNAL, **78**, 4710 (1956).

(17) H. B. Wood, Jr., and H. G. Fletcher, Jr., *ibid.*, **78**, 2849 (1956).

(18) R. N. Jones and co-workers, *ibid.*, **74**, 80 (1952); **72**, 956 (1950).

as I or II and ortho-acid esters such as III,[16,17,19] these infrared absorption techniques are admirably adapted. In Table II, no. 1 possesses three, no. 2 four and no. 5 five acetyl groups. With respect to both molecular extinction coefficient and integrated area under the carbonyl absorption band, it is clear that the theoretical ratios of 3/3, 4/3 and 5/3, respectively, are very closely in fact observed. Both our isomer A and isomer B show corresponding ratios close to 4/3, indicating conclusively that each possesses four normal acetyl groups, and eliminating III, having three acetyl groups, as a structural possibility for either. Also in accord with this conclusion regarding isomer A is the fact that this substance yields the normal methyl 2,3,4,6-tetra-O-acetyl-β-D-mannoside on methylation with either methyl iodide and silver oxide or with ethereal diazomethane. In the latter case, at least, the ortho-ester IV might have been anticipated[20] from structure III. The combined data above thus argue for I as the only reasonable structure for Isomer A.

TABLE II

RATIOS OF "INTEGRATED AREAS" AND MOLAR EXTINCTION COEFFICIENTS FOR 5.72 μ INFRARED ABSORPTION BAND IN SEVERAL D-MANNOPYRANOSE DERIVATIVES

Substance[a]	Ratio of molar extinction coeff. to extinction coeff. of no. 1	Ratio of integrated area to integrated area of no. 1	Theoretical ratio to no. 1
1	1.00	1.00	1.00
2	1.29	1.24	1.33
3	1.32	1.31	1.33
4	1.32	1.31	1.33
5	1.64	1.57	1.67

[a] 1 = 3,4,6-tri-O-acetyl-β-D-mannose 1,2-(methyl orthoacetate) (IV), 2 = methyl 2,3,4,6-tetra-O-acetyl-β-D-mannoside, 3 = isomer A, 4 = isomer B, 5 = 1,2,3,4,6-penta-O-acetyl-α-D-mannose.

Isomer B.—That this substance is a normal tetra-O-acetyl-D-mannose derivative is indicated by combustion analysis and the infrared data in Table II. Acetylation with chilled acetic anhydride in pyridine afforded essentially optically pure penta-O-acetyl-α-D-mannopyranose, a result also observed when acetylation was conducted at room temperature. That the C-1 hydroxyl group in isomer B was free was similarly shown by acetylation with radioactive acetic anhydride. The resulting acetyl-labeled penta-O-acetyl-α-D-mannopyranose was subjected to anomerizing conditions as before, and found to lose radioactivity at a first-order rate of 0.0189 ± 0.0004 min.$^{-1}$, an observation possible only if the C-1 acetoxy group in the present α-pentaacetate bore the carbon-14 label. When allowed to anomerize to its equilibrium mixture, the above radioactive pentaacetate produced a sample of reisolated penta-O-acetyl-α-D-mannopyranose containing no detectable residual radioactivity, thus indicating that the radioactive precursor contained *all* of its label at the anomeric acetyl group and eliminating any reasonable possibility that acetyl migration might have attended the above acetylation. Isomer B failed to show appreciable or rapid mutarotation in aqueous acetone, pyridine or a pyridine–phenol mixture, and was recoverable unchanged from the aqueous acetone experiment. The above data are consistent only with the structure 2,3,4,6-tetra-O-acetyl-α-D-mannose, the anomer of I, for that of isomer B.

As seen in Table I, isomer B has physical properties essentially identical with those, m.p. 93°, $[\alpha]^{27}D$ +26.3° ($CHCl_3$), reported by Levene and Tipson[4] for the tetra-O-acetyl-D-mannose which they obtained by action of a moist ethereal suspension of silver carbonate on crystalline tetra-O-acetyl-D-mannopyranosyl bromide. Their tetra-O-acetyl derivative, however, is reported to yield penta-O-acetyl-β-D-mannopyranose on acetylation with "cold" acetic anhydride and pyridine, whereas our isomer B has repeatedly afforded only an almost optically pure penta-O-acetyl-α-D-mannopyranose by acetylation either at −5° or at room temperature. We can rationalize this discrepancy only by the alternatives that either (a) Levene and Tipson's report of their acetylation unintentionally omits the mention of some obscure but critical detail of technique, or (b) isomer B and Levene and Tipson's tetraacetate are not identical.[21] In this connection it is also of interest that isomer B yielded chiefly an α-penta-O-acetate when acetylated with acetic anhydride and sodium acetate at 100°.

Isomer C.—The physical properties of this substance (Table I) bear a striking resemblance to those reported[3] by Micheel, m.p. 159–160°, $[\alpha]^{19}D$ −24.2° ($CHCl_3$), for the tetraacetate obtained on reaction of trimethylamine with tetra-O-acetyl-α-D-mannopyranosyl bromide. Unfortunately both our product and Micheel's product were obtained in very low yield, and there has been no convenient method of checking their apparent identity. The small amount of material on hand has allowed only a preliminary characterization of isomer C.

On acetylation with a chilled mixture of radioactive acetic anhydride and pyridine, isomer C gave a quantitative yield of almost optically pure penta-O-acetyl-β-D-mannopyranose. This pentaacetate was subjected to similar anomerizing conditions as before for a period of one exchange half-life for the C-1 β-acetyl group. The crude product obtained had a radioactivity assay equal within experimental error to that of its non-anomerized precursor. The failure to detect acetyl exchange under these conditions clearly indicates that the labeled acetyl group in the penta-O-acetyl-β-D-mannopyranose in question *did not* reside at C-1, and therefore, barring the unlikelihood of *complete* acetyl migration *toward* C-1 during acetylation, that an hydroxyl group on some carbon *other than* C-1 was unacetylated in Isomer C. This observation also eliminates the ortho-acid structure III, since III would certainly give some C-1 acetyl-labeled penta-O-acetyl-β-D-mannopyranose if it rearranged to a

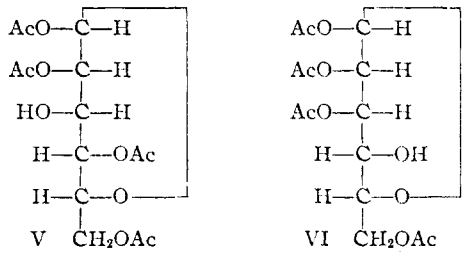
(19) W. W. Pigman and H. S. Isbell, *J. Research Nat. Bur. Standards*, **19**, 203 (1937).

(20) H. Meerwein and co-workers, *Ann.*, **484**, 1 (1930); *Ber.*, **64**, 2375 (1931); *J. prakt. Chem.*, **245**, 295 (1933).

(21) After very helpful correspondence from Dr. R. S. Tipson regarding the above matter, we contacted Dr. Lyman C. Craig, custodian of the P. A. Levene Memorial Collection at the Rockefeller Institute for Medical Research in an effort to secure a sample of Levene and Tipson's tetraacetate for chemical and physical comparison with isomer B. We were informed that the desired sample was unfortunately not on the list of chemicals in the Levene Collection.

"normal" tetraacetate during acetylation with labeled acetic anhydride.

Since isomer C cannot have its free hydroxyl group at C-6 (m.p. 135.5–136.5°, $[\alpha]^{20}D$ −22.5° ($CHCl_3$)[1,2]), and since both a C-1 hydroxyl structure and the ortho-acid structure III are excluded by the radiochemical observations above, only 1,3,4,6- (II), 1,2,4,6- (V) or 1,2,3,6-tetra-*O*-acetyl-β-D-mannopyranose (VI) remain as structural possibilities for isomer C. Furanose structures and α-anomer structures are excluded[7] by the above acetylation producing only penta-*O*-acetyl-β-D-mannopyranose.



Although optically stable in pyridine solution, isomer C mutarotated slowly with decomposition (darkening) in pyridine–phenol mixtures, and mutarotated fairly rapidly (k_C 0.0374 min.$^{-1}$) in aqueous acetone to an equilibrium rotation identical with that noted during similar mutarotation of isomer A. From the equilibrated solution of isomer C, isomer B could again be isolated. Since the anomeric center of isomer C is acetylated and thus incapable of simple anomerization, the present mutarotation must arise from an acetyl migration, obviously from C-1 to C-2, C-3 or C-4. Of the apparently possible structures, II, V and VI, for isomer C, structures II and V are clearly the only ones which would permit such acetyl migration from C-1, since the involved groups at C-1 and C-2 (II) or C-3 (V) are in a *cis*-relationship to one another. Fisher–Hirschfelder models show that, in the proper ring conformation at least, acetyl could readily migrate from C-1 to C-2 or C-3 *via* the generally postulated[15] ortho-acid intermediate (*e.g.*, III). Models show also that such an intermediate arising from VI is impossible, due to the *trans* relationships of the groups involved at C-1 and C-4. For these reasons we believe the structure of isomer C is best represented by II or V.

Since isomer C was not obtained starting with pure, crystalline tetra-*O*-acetyl-α-D-mannopyranosyl chloride, but resulted only in very small yield starting with the corresponding crude, sirupy bromide, the possibility is not excluded that isomer C was itself not formed during reaction of the latter bromide, but was rather already present either therein, or in its sirupy penta-*O*-acetyl-D-mannose precursor. The precise source of isomer C, however, has no bearing on the above structural arguments.

Experimental

Tetra-*O*-acetyl-α-D-mannopyranosyl Chloride.—Commercial D-mannose (20 g.) was treated with pyridine (100 ml.) and acetic anhydride (70 ml.) stirring until dissolved. After 15 hours the mixture was thrown into ice-water (500 ml.). The solution was stirred for 90 minutes, then extracted with chloroform. The extract was washed successively with dilute hydrochloric acid, water and sodium bicarbonate solution, then dried, filtered, and freed of solvent *in vacuo* on the steam-bath. There resulted 39.4 g. (90.5%) of crude acetylated mannose, which was used directly for reaction with titanium tetrachloride after the method of Pacsu.[6]

The above sirup (31.7 g.) in dry chloroform (105 ml.) was treated with titanium tetrachloride (9 ml.) in dry chloroform (96 ml.). After refluxing for four hours, the mixture was cooled, washed twice with ice-water, then with sodium bicarbonate solution, dried over anhydrous sodium sulfate, and freed of solvent *in vacuo* at a temperature of *ca.* 50°. The residue was dissolved in ether and the solution was decolorized by filtration through a Norit bed. Ligroin was added to the filtrate almost to incipient turbidity, and the solution was seeded with a product sample which crystallized spontaneously in a previous preparation. At 0° a 15.3-g. first crop was obtained, m.p. 77°, $[\alpha]^{25}D$ +89.4° (*c* 2.5, $CHCl_3$), in essential agreement with the recorded values[22] of m.p. 81°, $[\alpha]^{20}D$ +90.13° ($CHCl_3$). A second crop, 5.81 g., m.p. 74.5–75°, was obtained by concentrating the mother liquors to 40 ml., adding a little ligroin, and chilling to 0°. The total yield of crystalline material (71%) was used below without further purification.

The Anomers of 2,3,4,6-Tetra-*O*-acetyl-D-mannose.—Tetra-*O*-acetyl-α-D-mannopyranosyl chloride (29.2 g.), m.p. 77°, was dissolved in anhydrous ether (200 ml.). The solution was treated with silver carbonate (19.5 g.) and glass beads, after which water (1.46 ml.) was added with vigorous stirring over a 5-minute period. The mixture was stirred for 45 minutes, then filtered (Celite). The cake was washed by resuspension in hot anhydrous acetone (90 ml.), then refiltered. Solvent evaporation at 40–45° *in vacuo* in a rotary evaporator yielded 28.5 g. (103%) of white sirup, $[\alpha]^{25}D$ −5.7° (*c* 3.3, $CHCl_3$). This was dissolved in anhydrous ether (100 ml.), whereupon crystallization proceeded rapidly. After one hour at 0° 18.9 g. of isomer A, 2,3,4,6-tetra-*O*-acetyl-β-D-mannose, was collected, m.p. 123–123.5°, $[\alpha]^{25}D$ −15.75° (*c* 2.0, $CHCl_3$). The mother liquors were treated as described below. For purification the above solid (2.0 g.) was dissolved in anhydrous chloroform (3 ml.) and the solution was treated with anhydrous ether (10 ml.), seeded and chilled. Two such recrystallizations produced a sample having m.p. 124–124.5°, $[\alpha]^{25}D$ −13.62° → −12.0° (in 2 hours) (*c* 3.1, $CHCl_3$).

Anal. Calcd. for $C_{14}H_{20}O_{10}$: C, 48.27; H, 5.79. Found: C, 48.03, 48.05; H, 5.83, 5.85.

The mother liquors from an experiment similar to that above were placed in an open beaker and allowed to evaporate to a thick sirup, $[\alpha]^{25}D$ +19.3° (*c* 2.6, $CHCl_3$). On standing several days this sirup partially crystallized as huge prisms, which were slurried with ether and filtered, m.p. 93–94°, $[\alpha]^{25}D$ +22.8° (*c* 1.0, $CHCl_3$). These were used as seed for the mother liquor sirup obtained in the present experiment. On standing, 3.3 g. of sturdy prisms resulted. These were dissolved in chloroform (3 ml.) and treated with ether (12 ml.) and ligroin (10 ml.). Seeding and chilling yielded 2.3 g. of essentially pure isomer B, 2,3,4,6-tetra-*O*-acetyl-α-D-mannose, m.p. 95–96°, $[\alpha]^{25}D$ +23.1° (*c* 4.6, $CHCl_3$). Further crops were obtained on allowing the mother liquors to stand.

Anal. Calcd. for $C_{14}H_{20}O_{10}$: C, 48.27; H, 5.79. Found: C, 48.33, 48.18; H, 5.92, 5.89.

In earlier experiments where less careful attention was paid to the use of strictly anhydrous solvents, the two above products were isolated similarly. The originally obtained sirup, however, was higher rotating ($[\alpha]^{25}D$ + 4.1 to 4.8° ($CHCl_3$)) and produced a lower yield of the low-rotating β-anomer and a higher yield of the high rotating α-anomer. Similarly, on recrystallization of the β-anomer, strictly anhydrous solvents proved necessary; otherwise partial conversion to the α-anomer attended recrystallization. Presumably in the presence of excess water some conversion of the β-anomer to the α-anomer takes place (*cf.* below the mutarotation of isomer A in aqueous acetone).

Acetylation of 2,3,4,6-Tetra-*O*-acetyl-β-D-mannose.—The above isomer A (1.00 g.) was dissolved in a 1:1 mixture (14 ml.) of acetic anhydride and pyridine which had been chilled to −5°. The solution was allowed to stand at −5° for three days, then thrown into water. After 45 minutes the solution was extracted twice with chloroform. The extract was washed successively with dilute hydrochloric acid, water

(22) D. H. Brauns, *J. Research Nat. Bur. Standards*, **7**, 581 (1931).

and sodium bicarbonate solution, then dried over anhydrous sodium sulfate, filtered and freed of solvent at 100° *in vacuo*. There resulted an 85–95% yield of crude sirup which crystallized on scratching, m.p. 111–113°, $[\alpha]^{25}D$ −23.8° (*c* 1.2, $CHCl_3$) (corresponding to an anomeric composition of 97% β and 3% α). This was recrystallized from a mixture of 2-propanol (3 ml.) and water (2 ml.), to produce 0.64 g. of pure penta-*O*-acetyl-β-D-mannopyranose, m.p. 115.5–116°, mixed m.p. with an authentic sample 115.5–116.5°, $[\alpha]^{25}D$ −25.4° (*c* 2.6, $CHCl_3$).

The above acetylation of isomer A was repeated using acetic anhydride methyl-labeled with C^{14}. A quantitative yield of crude acetyl-labeled penta-*O*-acetyl-β-D-mannopyranose was obtained, which had m.p. 115–115.5° after three recrystallizations as above. This was used in the acetyl exchange experiments described below.

In another experiment isomer A (0.374 g.) was dissolved in 10 ml. of a 1:1 pyridine–acetic anhydride mixture. Placed in a 2-dm. polarimeter tube at 25°, the following specific rotations (°) were observed at the indicated times (minutes): −38.4, 2; −36.4, 5; −35.6, 10; −34.2, 25; −33.7, 60; −33.7, 120. Thus polarimetrically the acetylation appeared substantially complete in about 30 minutes. The solution was poured into water after two hours, and the product was isolated as before, 0.41 g. (98%) of clear sirup which rapidly crystallized. Its rotation $[\alpha]^{25}D$ −20.5° (*c* 2.44, $CHCl_3$) corresponded to an anomeric composition of 93.5% β and 6.5% α. Recrystallized from 2-propanol (1.5 ml.) the crude sample afforded 0.31 g. of pure penta-*O*-acetyl-β-D-mannopyranose, m.p. 115–115.5°. Acetylation of isomer A thus proceeded considerably more rapidly than did its mutarotation (*cf.* below) in pyridine.

Acetylation of 2,3,4,6-Tetra-*O*-acetyl-α-D-mannose.—A one-gram sample of the above isomer B was acetylated at −5° in the manner described above. A quantitative yield of crude sirupy penta-*O*-acetyl-α-D-mannopyranose, $[\alpha]^{25}D$ +51.5° (*c* 3.6, $CHCl_3$), resulted. This was recrystallized from a mixture of 2-propanol (3 ml.) and water (3 ml.) at 0°, seeding[23] with crystallized product, to produce 0.66 g. of α-D-mannopyranose pentaacetate, m.p. 74–75°, mixed m.p. with an authentic sample[23] undepressed, $[\alpha]^{25}D$ +56.8° (*c* 1.8, $CHCl_3$).

This acetylation was repeated using methyl-labeled acetic anhydride, to provide a sample of penta-*O*-acetyl-α-D-mannopyranose, m.p. 74–75°, $[\alpha]^{25}D$ +56.7° (*c* 1.5, $CHCl_3$), labeled in the C-1 acetyl group for the acetyl exchange experiments described below.

When 0.50 g. of isomer B was acetylated with acetic anhydride (3 ml.) and pyridine (3 ml.) for 42 hours at 25°, similar processing yielded 0.58 g. (103%) of crude sirup whose rotation, $[\alpha]^{25}D$ +45.0° (*c* 2.1, $CHCl_3$), corresponded to that of a mixture of 88% penta-*O*-acetyl-α-D-mannose and 12% β-anomer. The pure α-anomer was again obtained by one crystallization. The rotation of the crude acetylated product from the −5° acetylation above corresponded to 94% α- and 6% β-anomer.

On acetylation of 0.50 g. of isomer B with acetic anhydride (5 ml.) and anhydrous sodium acetate (1.0 g.) for one hour at 100°, 0.59 g. (105%) of crude sirup resulted, $[\alpha]^{25}D$ +36.9° (*c* 2.9, $CHCl_3$). This rotation corresponds to an anomeric composition of 76% α- and 24% β-penta-*O*-acetate.

Methylation of 2,3,4,6-Tetra-*O*-acetyl-β-D-mannose, with Methyl Iodide.—A mixture of the above isomer A (5.0 g.), silver carbonate (5.0 g.), Drierite (5.0 g.), glass beads and methyl iodide (25 ml.) in anhydrous ether (50 ml.) was stirred at room temperature under $CaCl_2$-tube protection during 19 hours. The mixture was filtered and the cake was rinsed twice with acetone. The filtrate and rinsings were evaporated *in vacuo* to yield 5.0 g. of clear sirup which partially crystallized on scratching with ethanol, $[\alpha]^{25}D$ + 4.03° (*c* 3.5, $CHCl_3$). This was dissolved in ethanol (10 ml.), seeded and chilled to 0°, depositing 0.95 g. of crude methyl 2,3,4,6-tetra-*O*-acetyl-β-D-mannoside, m.p. 151–153°. Two further recrystallizations from 2-propanol (3 ml.) afforded a sample having m.p. 158–158.5°, $[\alpha]^{25}D$ −45.8° (*c* 1.5, $CHCl_3$), properties in substantial agreement with those described[13] in the literature, m.p. 161°, $[\alpha]^{20}D$ −47° ($CHCl_3$). No further crystalline material was isolated from the mother liquors.

(23) We are indebted to Dr. H. S. Isbell for kindly furnishing seed crystals of penta-*O*-acetyl-α-D-mannopyranose.

With Diazomethane.—Diazomethane was prepared in benzene solution (50 ml.) according to the procedure of Arndt.[24] The solution was dried over potassium hydroxide and decanted prior to use. Two grams of the above isomer A was dissolved in chloroform (15 ml.), and the diazomethane solution (15 ml., *ca.* 200% excess) was added. Gradual nitrogen evolution was noted at the outset. After three days at room temperature the solution was treated with acetic acid (0.3 ml.) then freed of its solvents *in vacuo* in a rotary evaporator. There resulted 2.14 g. of a semicrystalline tan paste, $[\alpha]^{25}D$ +6.6° (*c* 2.4, $CHCl_3$). This was dissolved in hot chloroform (5 ml.) and treated with ether (5 ml.). On standing, rapid crystallization proceeded, yielding 0.40 g. of tan platelets, m.p. 155–156°. These were recrystallized from 2-propanol (3 ml.) to give 0.30 g. of white platelets, m.p. 159–160°, $[\alpha]^{25}D$ −47.7° (*c* 1.2, $CHCl_3$). This material showed no melting point depression (158.5–159.5°) on admixture with the above methyl 2,3,4,6-tetra-*O*-acetyl-β-D-mannoside.

Action of Moist Silver Carbonate on Crude Tetra-*O*-acetyl-α-D-mannopyranosyl Bromide.—The sirupy penta-*O*-acetyl-D-mannose (20.8 g.), described in the first experiment above, was treated with a cold 43% solution of hydrogen bromide in acetic acid (44 ml.) and the mixture was shaken for 2.5 hours at room temperature. It was then diluted with chloroform and washed three times with ice-water and once with iced sodium bicarbonate solution. After drying over sodium sulfate the chloroform was evaporated *in vacuo* at room temperature in a rotary evaporator, yielding 23.3 g. of amber sirup. This failed to crystallize on standing *in vacuo* at 0° for one week over P_2O_5 and NaOH. It was accordingly dissolved in ether, decolorized with Norit, and the solution was treated with ligroin. Three days standing at 0° produced only an oil. This was redissolved, decolorized again, and freed of solvent as before in a rotary evaporator. There resulted 18.9 g. of amber sirup, $[\alpha]^{25}D$ +111.8° (*c* 4.8, $CHCl_3$), which was used directly below. The physical properties for pure tetra-*O*-acetyl-α-D-mannopyranosyl bromide are given[4] by Levene and Tipson as m.p. 53–54° $[\alpha]^{26}D$ +123.2° ($CHCl_3$).

The above sirup (18.8 g.), silver carbonate (9.4 g.) and glass beads in anhydrous ether (94 ml.) were treated on stirring with 0.94 ml. of water. The mixture was stirred for two hours, filtered, and the cake was rinsed with acetone. Rotary evaporation of the solvent left 15.2 g. of clear sirup, $[\alpha]^{25}D$ +6.75° (*c* 3.7, $CHCl_3$). This was dissolved in ether (40 ml.); scratching produced 0.46 g. of isomer C, m.p. 162–163°, which was investigated below. The filtrate was placed at 0° after seeding, yielding 1.7 g. of isomer A, m.p. 123–124°, mixed m.p. with the previously obtained sample of isomer A undepressed, infrared spectrum in chloroform solution identical with that of previously obtained sample, $[\alpha]^{25}D$ −15.8° (*c* 3.0, $CHCl_3$).

The mother liquors were allowed to evaporate slowly, depositing 4.1 g. of isomer B, m.p. 95.5–96°, mixed m.p. with previous sample of isomer B, undepressed, infrared spectrum in chloroform identical with that of previous sample, $[\alpha]^{25}D$ +24.8° (*c* 3.2, $CHCl_3$). This material was acetylated at −5° with acetic anhydride in pyridine as before, producing similarly a quantitative yield of almost pure penta-*O*-acetyl-α-D-mannopyranose, $[\alpha]^{25}D$ +51.5° (*c* 3.6, $CHCl_3$). After one crystallization from dilute 2-propanol the product had m.p. 74–75°, $[\alpha]^{25}D$ +56.8° (*c* 1.8, $CHCl_3$).

Purification and Acetylation of Isomer C.—A sample (0.31 g.) of isomer C, m.p. 162–163°, obtained in the preceding experiment, was dissolved in chloroform (1 ml.). The solution was filtered through Celite, rinsing with additional chloroform. The filtrate was concentrated in an air-stream to *ca.* 0.3 ml., then treated with ether (3 ml.). Cooling to 0° produced 0.21 g. of pure product, m.p. 164–165°, $[\alpha]^{25}D$ −25.2° (*c* 0.52, $CHCl_3$).

Anal. Calcd. for $C_{14}H_{20}O_{10}$: C, 48.27; H, 5.79. Found: C, 48.48, 48.35; H, 5.88, 5.81.

Another sample (0.15 g.) of the above 162–163° product was dissolved in 2 ml. of a 1:1 mixture of pyridine and methyl-labeled acetic anhydride chilled to −5°. The solution stood at −5° for three days, was thrown into water, and the product was isolated as before. There was obtained 0.18 g. (106%) of crude sirup, $[\alpha]^{25}D$ −22.0° (*c* 1.7, $CHCl_3$). This material was crystallized from a mixture of

(24) F. Arndt, "Organic Syntheses," Coll. Vol. II, John Wiley and Sons, Inc., New York, N. Y., p. 166.

2-propanol (0.75 ml.) and water (0.60 ml.) at −5°, seeding with penta-*O*-acetyl-β-D-mannopyranose, yielding 0.137 g. of fairly pure product, m.p. 114.5–115°, $[\alpha]^{25}D$ −25.4° (*c* 1.8, $CHCl_3$), mixed m.p. with authentic sample 115.5–116.5°. This entire product was used in the acetyl exchange experiment described below.

Radioactive Acetyl Exchange Experiments.—In general the following procedure was employed to detect the loss of labeled acetyl under anomerizing conditions from the three labeled penta-*O*-acetyl-D-mannopyranose samples in question. The indicated weight of the pentaacetate was dissolved in the indicated volume of a 1:1 acetic acid–acetic anhydride mixture. At time zero an equal volume of a 1 *M* solution of sulfuric acid in 1:1 acetic acid–acetic anhydride was added, and the stopwatch was started. Both solutions were thermostated at 25° prior to mixing, and kept at this temperature during the run. Each final solution in question was 0.1 *M* in penta-*O*-acetyl-D-mannose and 0.5 *M* in sulfuric acid. At the indicated time intervals a 2-ml. aliquot of the reaction mixture was removed and quenched in water (20 ml.). After five minutes the product from each aliquot was extracted into chloroform (10 ml.). The extract was washed with sodium bicarbonate solution, dried over anhydrous sodium sulfate, decanted, and freed of solvent (10 ml. beaker) in an air-stream on the steam-bath. The residue from each aliquot was dried *in vacuo* over P_2O_5 and NaOH for several days, then assayed for radioactivity by wet-combustion,[25] followed by counting[26] in an ionization chamber with the aid of a Cary model 31 vibrating reed electrometer. The first-order rate constants for the loss of radioactive acetoxy were calculated by means of the usual equation[27]

$$k_{exch} = \frac{2.3}{t} \log \frac{a}{a - x}$$

where a is the radioactivity assay at zero time and $a - x$ the assay at time t. The data for such experiments on each of the mannose pentaacetates in question are given individually below.

Penta-*O*-acetyl-β-D-mannopyranose from Isomer A

Sample size, 0.3902 g.; solvent, 5.0 ml.; 1.0 *M* sulfuric acid solution, 5.0 ml.

t, min.	Assay, mc./mole	$k_{exch.}$, min.$^{-1}$
0	0.307	....
7	.242	0.0339
14	.185	.0363
21	.153	.0332
30	.125	.0299

Average 0.0333 ± 0.0018

Penta-*O*-acetyl-α-D-Mannopyranose from Isomer B

Sample size, 0.3902 g.; solvent, 5.0 ml.; 1.0 *M* sulfuric acid solution, 5.0 ml.

t, min.	Assay, mc./mole	$k_{exch.}$, min.$^{-1}$
0	0.0773	....
25	.0480	0.0190
50	.0310	.0182
75	.0178	.0195
100	.0117	.0188

Average 0.0189 ± 0.0004

Penta-*O*-acetyl-β-D-mannopyranose from Isomer C: — sample size, 0.1369 g.; solution, 3.51 ml. of 1:1 acetic acid acetic anhydride, 0.5 *M* in sulfuric acid; assay at time zero, 0.0762 mc./mole; assay after 21 minutes, 0.0776 mc./mole. Thus no exchange was evident after one half-life for the exchange of the C-1 acetyl group of penta-*O*-acetyl-β-D-mannopyranose.

Acetyl Exchange on Complete Anomerization. Penta-*O*-acetyl-β-D-mannopyranose from Isomer A.—Another sample of penta-*O*-acetyl-β-D-mannopyranose, m.p. 116–116.5°, $[\alpha]^{25}D$ −25.5° (*c* 2.7, $CHCl_3$), obtained by acetylation of isomer A as before with radioactive acetic anhydride and pyridine at −5°, had a specific radioactivity of 0.0806 mc./mole; 300 mg. of this sample was dissolved in 7.5 ml. of a 1:1 mixture of acetic anhydride and acetic acid which was 1.0 *M* in sulfuric acid. After 4.5 hours at room temperature the reaction was quenched in water and the product was isolated as before; 0.29 g. (97%) of crude α-anomer. This was recrystallized twice from dilute 2-propanol to yield 0.12 g. of pure penta-*O*-acetyl-α-D-mannopyranose, m.p. 74°. This latter sample had a radioactivity assay of 0.0124 mc./mole, indicating that 15.4% of the original radioactive acetyl was still in the sample. In an earlier experiment where somewhat less pure reactants were employed, 16% of the original label was found in the anomerized product. Anomerization, as measured polarimetrically, is complete under the above conditions in about 1.5 hours.

Penta-*O*-acetyl-α-D-mannopyranose from Isomer β.—Two hundred milligrams of the above penta-*O*-acetyl-α-D-mannopyranose from isomer B having a specific activity of 0.0773 mc./mole was placed in 5 ml. of 1:1 acetic anhydride acetic acid 1.0 *M* in sulfuric acid. After 5.25 hours the α-acetate was re-isolated as usual and found to have a radioactivity level indistinguishable from the background level under the conditions of assay.

Mutarotation Experiments.—Isomers A, B and C were each placed in three different solvents to observe their mutarotations. In each of the experiments below are given (a) solute concentration; (b) time in minutes and corresponding specific rotation, $[\alpha]^{25}D$; (c) $[\alpha]^{25}D$ at zero time, α_0, obtained by graphical extrapolation of the plotted mutarotation curves; and (d) the average of all first-order isomerization rates for pertinent cases, calculated according to the equation[28]

$$k_{isom} = \frac{2.3}{t} \log \frac{\alpha_0 - \alpha_e}{\alpha_t - \alpha_e}$$

Aqueous Acetone.—The solute was dissolved in 6 volumes of acetone. The solution was treated with 4 volumes of water at time zero then placed immediately in the polarimeter.

Isomer A: (a) concn., 3.00 g./100 ml.; (b) *t*, $[\alpha]^{25}D$: 1, −14.0°; 3, −6.2°; 5, −0.8°; 7, +3.3°; 10, +6.8°; 15, +15.2°; 20, +17.8°; 30, +19.4°; final, +19.6°; (c) α_0 approximately −20.0°; (d) k_A, 0.144 min.$^{-1}$. An equilibrated mixture was evaporated to dryness in an air-stream yielded 0.30 g. of sirup, $[\alpha]^{25}D$ +18.1° (*c* 1.2; $CHCl_3$; this was dissolved in ether and seeded. Slow evaporation produced 0.11 g. of 2,3,4,6-tetra-*O*-acetyl-α-D-mannose, m.p. 95–95.5°, $[\alpha]^{25}D$ +24.1° (*c* 1.5, $CHCl_3$), mixed m.p. with sample above undepressed).

Isomer B: concn., 3.00 g./100 ml.; (b) *t*, $[\alpha]^{25}D$: 2, +22.7°; 45, 21.0°; 125, 22.0°; 300, 22.3°; 1440, 23.0°. (Similar isolation led to 0.29 g. of sirup which again yielded from ether crystalline 2,3,4,6-tetra-*O*-acetyl-α-D-mannose, m.p. 95–95.5°.)

Isomer C: (a) concn., 0.934 g./100 ml.; (b) *t*, $[\alpha]^{25}D$: 1, −26.3°; 13, −7.0°; 21, +2.1°; 47, +12.3°; 139, +19.3°; final, +19.8°; (c) α_0 approximately −28.0°; (d) k_c 0.0374 min.$^{-1}$. An equilibrated mixture was evaporated to dryness in an air-stream; the residue was dissolved in a little ether. The solution was filtered and the filtrate treated with ligroin, seeded and chilled. There resulted 0.06 g. of solid, m.p. 89–90°, $[\alpha]^{25}D$ +24.7° (*c* 1.6, $CHCl_3$), mixed m.p. with isomer B, 89–90°, whose infrared spectrum in chloroform solution was identical in all respects to that of isomer B in chloroform.

Pyridine. Isomer A: (a) concn., 1.995 g./100 ml.; (b) *t*, $[\alpha]^{25}D$:1, −52.5°; 5, −50.0°; 30, −35.6°; 65, −20.8°; 107, −9.0°; 200, +3.8°; 285, +7.8°; 360, +10.3°; 405, +10.5°; final, +11.0°; (c) α_0 approximately −53.0°; (d) k'_A, 0.0110 min.$^{-1}$.

When 0.334 g. of isomer A was dissolved in 10 ml. of chilled (−5°) pyridine and placed at −5° for 47 hours, then allowed to warm to room temperature, $[\alpha]^{23}D$ was −4.3°. This rotation corresponds to a mixture of 20% isomer A and 80% equilibrated product.

Isomer B: (a) concn., 1.179 g./100 ml.; (b) *t*, $[\alpha]^{25}D$: 4, +29.2°; 44, +30.9°; 1004, +27.6°; 1320, +25.4°; 1382, +25.4°.

Isomer C: (a) concn., 0.657 g./100 ml.; (b) *t*, $[\alpha]^{25}D$: 10, −68.5°, unchanged over five observations to 1345, −68.5°.

Pyridine–Phenol.—The solute was dissolved in 2 volumes of pyridine, then treated immediately with 1 volume of a 1:1

(25) O. K. Neville, THIS JOURNAL, **70**, 3501 (1948).

(26) V. A. Raaen and G. A. Ropp, *Anal. Chem.*, **25**, 174 (1953).

(27) W. A. Bonner and C. J. Collins, THIS JOURNAL, **77**, 102 (1955).

(28) C. S. Hudson, *Z. physik. Chem.*, **44**, 487 (1903).

mixture of pyridine and phenol at time zero, giving a 5:1 pyridine–phenol solvent. In the case of B, a 1:1 pyridine–phenol solvent was employed.

Isomer A: (a) concn., 1.683 g./100 ml.; (b) *t*, $[\alpha]^{25}D$: 1, −40.0°; 3, −22.2°; 4, −15.1°; 6, −5.9°; 10, +2.4°; 15, +8.3°; 20, +9.8°; final, +10.7°; (c) α_0 approximately −53.0° (from corresponding mutarotation in pyridine above); (d) k''_A, 0.218 min.$^{-1}$.

Isomer B: (a) concn., 0.737 g./100 ml.; (b) *t*, $[\alpha]^{25}D$: 2, −80°; 60, −67.8°; 130, −57.0°; 200, −47.4°; 555, +10.8° (at this point the solution had darkened extensively, and further polarimetric observations were impossible).

3,4,6-Tri-*O*-acetyl-β-D-mannose 1,2-(methylorthoacetate) (IV) was prepared from sirupy tetra-*O*-acetyl-α-D-mannopyranosyl bromide by treatment with a suspension of silver carbonate in methanol after the procedure of Dale.[13] After recrystallization from a 1:6 chloroform–ether mixture the product had m.p. 101–102°, $[\alpha]^{25}D$ −29.0° (*c* 2.4, $CHCl_3$), in substantial agreement with the m.p. 105°, $[\alpha]^{20}D$ −26.6° ($CHCl_3$) reported.[13]

Infrared Absorption Studies.—Chloroform solutions of the substances listed in Table II were made up at a concentration of 0.025 *M* each. These solutions were in turn placed in a 0.105-mm. sodium chloride cell, and the infrared spectrum of each was scanned in the region 5.0–6.2 μ, using a Perkin–Elmer model 21 double-beam infrared spectrophotometer. Chloroform solvent was used in a 0.107-mm. cell in the I_o beam, and a slit-width of 57 μ was employed. Since the carbonyl absorption band in each of these compounds proved almost completely symmetrical, the areas under each band were approximated by multiplying the peak height by the half-peak height width. The areas so measured were 5.25, 6.53, 6.90, 6.90 and 8.26 cm.^{2}, respectively, for compounds 1 through 5 in Table II. The molar extinction coefficients, calculated in the usual way from the peak heights and the relationship $\epsilon = (\log I_0/I_t) \times (1/lc)$ were 903, 1165, 1195, 1195 and 1476, respectively, for compounds 1 through 5 in Table II.

STANFORD, CALIFORNIA

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, THE OHIO STATE UNIVERSITY, AND THE DEPARTMENT OF BIOCHEMISTRY, UNIVERSITY OF CALIFORNIA, BERKELEY]

The Synthesis of D-*erythro*-Pentulose Tetrabenzoate[1]

BY D. L. MACDONALD,[2a] J. D. CRUM[2b] AND R. BARKER[2a]

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The synthesis of 1,3,4,5-tetra-*O*-benzoyl-*keto*-D-*erythro*-pentulose is described. The fully benzoylated aldotetronic acid was converted to the acyl chloride and this with diazomethane yielded the diazomethyl ketone. The latter was transformed directly into the ketopentose tetrabenzoate and the identity of the keto ester was demonstrated by reduction and isolation of known arabinitol and ribitol derivatives.

In continuation of our work on the general method for the preparation of ketoses from the lactones of the sugar acids with one less carbon atom,[3] we wish to describe the application of this method to the synthesis of D-*erythro*-pentulose ("ribulose") as the crystalline tetrabenzoate. The reaction sequence employed was

$$RCO_2H \xrightarrow{PCl_5} RCOCl \xrightarrow{CH_2N_2} RCOCHN_2 \xrightarrow[Cu]{C_6H_5COOH} RCOCH_2OCOC_6H_5$$

R = $CH_2OCOC_6H_5(CHOCOC_6H_5)_2$–

The starting point for our synthesis was D-erythrono-1,4-lactone prepared by the selective degradation of D-glucose by lead tetraacetate[4] and subsequent bromine oxidation of the D-erythrose to D-erythronolactone or, more conveniently for larger quantities, by the oxidative scission of D-*erythro*-2-hexulosono-1,4-lactone 2,3-*cis*-enediol ("D-araboascorbic acid") with *p*-toluenediazonium sulfate.[5] The lactone was converted to the amide with liquid ammonia[6] and subsequent benzoylation in anhydrous pyridine with benzoyl chloride gave 2,3,4-tri-*O*-benzoyl-D-erythronamide.[7]

The benzoylated amide was converted to 2,3,4-tri-*O*-benzoyl-D-erythronic acid with nitrosyl chloride in dioxane, and then to 2,3,4-tri-*O*-benzoyl-D-erythronyl chloride with phosphorus pentachloride. Reaction with diazomethane and silicate column chromatography of the reaction product gave 1-deoxy-1-diazo-*keto*-D-*erythro*-pentulose tribenzoate. Treatment of the diazomethyl ketone with benzoic acid and copper bronze yielded 1,3,4,5-tetra-*O*-benzoyl-D-*erythro*-pentulose.

The free keto structure of this tetrabenzoate was demonstrated by reduction of the carbonyl group with sodium borohydride to the corresponding diastereoisomeric pentitol tetrabenzoates which, after saponification with aqueous sodium hydroxide and treatment with hydrochloric acid,[8a] were separated by paper chromatography and identified as D-arabinitol (and D-arabinitol pentaacetate) and 1,4-anhydro-DL-ribitol (and 2,3,5-tri-*O*-benzoyl-1,4-anhydro-DL-ribitol). The treatment of D-arabinitol and ribitol with hydrochloric acid under like conditions has been shown[8a] previously by paper chromatography to yield unchanged D-arabinitol and 1,4-anhydro-DL-ribitol, respectively. The separation of these compounds and the synthesis of the latter are herein described on a preparative basis.[8b] The isolation of the two 2-epimeric pentitols thus establishes the free keto nature of the second carbon atom in the crystalline ketopentose tetrabenzoate.

D-*erythro*-Pentulose ("ribulose") was first synthesized by Glatthaar and Reichstein[9] by the pyridine interconversion of D-arabinose and isolated as

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