Anal. Calcd. for $C_{18}H_{14}N_2O_9$: C, 53.7; H, 3.5; N, 7.0. Found: C, 53.8; H, 3.6; N, 7.0.

B. Synthesis of Erythritan (IV).—A solution of erythritol (3 g.) in a mixture of sulfuric acid (3 g.) and water (3 g.) was boiled gently under reflux for 15 hours. The erythritan, isolated in the same manner as L-threitan, was a colorless, hygroscopic liquid, b.p. (bath temperature) 160–165° (0.17 mm.), $n^{24}D$ 1.4370 (yield 1.3 g.).[15]

Anal. Calcd. for $C_4H_8O_3$: C, 46.2; H, 7.8. Found: C, 46.0; H, 7.8.

Erythritan di-*p*-nitrobenzoate, prepared in the usual way, separated from acetone in the form of needles, m.p. 173–174°.

Anal. Calcd. for $C_{18}H_{14}N_2O_9$: C, 53.7; H, 3.5; N, 7.0. Found: C, 53.9; H, 3.6; N, 7.1.

C. Oxidation with Periodate.—The compounds were oxidized with sodium periodate and with periodic acid in aqueous solution under the conditions previously used.[12] The results of the oxidations with 0.1 *N* sodium periodate are given in Table I. Oxidation with 0.1 *N* periodic acid, which proceeded according to the results recorded in Table II and represented graphically in Fig. 1, illustrates clearly the marked effect of the stereochemical arrangement of adjacent hydroxyl groups on the rate of oxidation by periodate.

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[CONTRIBUTION FROM THE NORTHERN REGIONAL RESEARCH LABORATORY[1]]

Dextran Triacetates

BY ALLENE JEANES AND C. A. WILHAM

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A simple, non-degradative method is described for preparing dextran triacetates from dextrans in the form of either the hydrated gum or the dry powder. This method has been applied to the acetylation of nine bacterial dextrans differing widely in chemical and physical characteristics. Intrinsic viscosities and specific rotations of the dextran triacetates show close relationship to the corresponding data for the respective dextrans as well as to the periodate oxidation data for these dextrans. This proves that differences among the dextrans are due to fundamental structural characteristics which are carried over into the triacetates. The degradation temperatures of these dextran triacetates and the film-forming ability of one of them are shown to correlate with their chemical structures.

This report describes the preparation and some of the properties of triacetates of nine bacterial dextrans. The particular dextrans were chosen for this purpose because of their widely different characteristics. These triacetates are being employed at this Laboratory in a program on chemical and physical characterization of many dextrans and in comparative studies on the α-1,6-glucopyranosidic linkage in starch and in dextrans.[2] The rapidly expanding interest in the structure,[3–5] properties[2,6] and application in medicine[7] of dextrans makes it desirable to confirm and extend, through study of a derivative such as the triacetate, differences previously established only in the polysaccharides. The observations reported here also augment the already recognized industrial potentialities of dextran acetates[8] by making available for the first time undegraded products of favorable solubility characteristics.

Previously reported methods for preparation of dextran acetates resulted in incomplete acetylation[9,10] or in products which appeared to be fractionated[10,11] or degraded.[11]

(1) One of the laboratories of the Bureau of Agricultural and Industrial Chemistry, Agricultural Research Administration, U. S. Department of Agriculture. Article not copyrighted.

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Experimental

Materials and Analytical Methods.—All the dextrans used in this study were prepared in a manner similar to that already described for dextran from *Leuconostoc mesenteroides* NRRL B-512.[2] They contained no more than about 0.05% ash, 0.01% nitrogen, 0.001% phosphorus and no detectable fructose.

Formamide was a neutral fraction obtained from commercial material by distillation *in vacuo*. *sym*-Tetrachloroethane was washed free of acid, dried and distilled before use.

Acetyl determinations were made on 150-mg. samples (80 mesh, unless the preparation had been freeze-dried) by a modified Eberstadt method as described by Murray, Staud and Gray.[12] The values are reproducible to ±0.2% acetyl.

Viscosity measurements were made in Ostwald-Cannon-Fenske No. 100 tubes.

All samples for analytical measurements were equilibrated with atmospheric moisture in the room in which the samples were weighed and where a constant relative humidity of 61% at 21° was maintained. The dextran triacetates were found to have moisture contents of 3 to 4% by drying samples *in vacuo* over phosphorus pentoxide for 4 hours at 100°. All calculations were made on a dry basis.

Acetylation in Formamide.—An adaptation of the method of Carson and Maclay was employed,[13] under conditions so mild as to preclude the likelihood of degradation. Air-dry dextran in a fluffy, homogeneously reactive state[2] was used, or gum dextran which had been precipitated from aqueous solution by addition of ethanol to 50% by volume and from which excess aqueous ethanol had been expressed. Five grams (dry basis) of the dextran was mixed with 75 ml. of ice-cold formamide and solution was completed at room temperature. To this stirred solution was added slowly 75 ml. of pyridine and then 65 ml. of acetic anhydride was added over a period of about 1.5 to 3 hours. The reaction mixture was maintained at room temperature by cooling, if necessary. In most cases addition of acetic anhydride caused a finely divided precipitate to form. However, acetylation mixtures of dextrans from B-1299 and B-1355 and of fraction C from B-742, remained homogeneous, and that from B-523 was heterogeneous throughout the acetylation procedure. The reaction mixture was stirred for 4 hours at room temperature and allowed to stand 20 hours.

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TABLE I

PROPERTIES OF DEXTRANS AND OF THEIR TRIACETATES

Strain	Dextran						Dextran triacetate		
	Types of linkages calcd. from periodate oxidation, %[4,5b]								
NRRL strain no. of organism[a]	1,6-	1,4-like	1,3-like	$[\eta]$ water, 25°	$[\alpha]^{25}D$ (c 1, formamide), ±2[d]	Acetyl content, %	$[\eta]$, *sym*-tetrachloroethane, 25°	$[\alpha]^{25}D$ (c 1, *sym*-tetrachloroethane), ±1°	Decomposition range, °C. (cor.)[b]
B-1299	50	50		0.47	+216°	44.8	0.53	+213°	203–225
B-1355	62	7	31	.37	234	44.7	.40	199	230–240
B-742 (C)[c]	67	24	9	.24	224	44.6	.25	200	220–230
B-742 (L)[c]	79	20	[d]	.14[e]	216	44.5	.18	212	220–230
B-523[f]	81	19				45.0			295–305
B-1254	90	8		.56	218	44.5	.50	214	290–300
B-512	95	5		1.03	215	44.9	1.03	218	295–305
B-1064	96	2		0.89	214	44.5	0.85	218	295–300
B-1146	97	3		1.21	214	44.7	1.15	219	290–300

[a] These organisms, numbered as indicated in the NRRL Culture Collection, are all *Leuconostoc mesenteroides* except B-1254, which is *Streptobacterium dextranicum*, and B-1146, which is *Leuconostoc dextranicum*. [b] Fusion occurred over a 10–20° range before decomposition began. [c] Fractions C and L came from the same dextran preparation. Fraction L, insoluble in 35% ethanol, was obtained in a yield of 17% of the theoretical; Fraction C, soluble in 35% ethanol, was obtained in 36% yield. [d] Calculated percentages of 2% or less are of doubtful significance and are not recorded. [e] Although this dextran had a very low viscosity, like all the other dextrans its particle weight was of the order of many millions. [f] Both the dextran and its triacetate were insoluble in the solvents indicated.

It was added to ice and water and the gelatinous precipitate was washed with water and ethanol until free of acid. The gummy product was dried in a vacuum desiccator over anhydrous calcium chloride or from the frozen state of an aqueous suspension. The yield was quantitative, usually about 97% being recovered. The acetyl contents indicated complete acetylation to the triacetate (Table I).

Unless otherwise stated, all dextran triacetates referred to subsequently in this paper were prepared by the foregoing method.

A second acetylation treatment, such as is reported to be necessary for complete acetylation of pectic and amylaceous substances by this method,[13,14] made no detectable change in these dextran triacetates.

Acetylation by Other Methods.—Dry B-512 dextran was acetylated to the triacetate in heterogeneous mixture by treatment with acetic anhydride and pyridine at 100° for 16 hours, or with fused sodium acetate and acetic anhydride at 100° for 24 hours.

Acetylation of dextran catalyzed by sulfuric acid in heated solutions has been reported.[8] However, we found that even when the acetate was prepared at room temperature or lower with a minimum of sulfuric acid as catalyst, the odor of acetic acid became apparent in samples of the purified, dry acetate which had been bottled several weeks.

Properties of Dextran Triacetates.—Dextran triacetates prepared by the formamide method did not contain residual formamide or combined amido or formyl groups. This was indicated by the nitrogen contents being less than 0.01%, and the negative tests for formic acid[15,16] being obtained on distillates containing the volatile acids liberated from the triacetates by saponification and subsequent acidification.

The triacetate from the water-insoluble dextran from B-523 was insoluble in all solvents tested. The only triacetate soluble in acetone was that from B-1299. Triacetates from B-1299 and from the B-742 dextran fractions were unique in being soluble in chloroform as well as in *sym*-tetrachloroethane. The only solvent we found for all the other dextran triacetates at 25° is *sym*-tetrachloroethane. Solutions of 10% concentration have been obtained at 25° and of 20% by heating. Vacuum drying of the dextran triacetates from their frozen water suspension did not change their solubilities, but did increase the ease of solution. Dextran triacetates prepared by the pyridine or sodium acetate methods at 100° were less readily soluble than those prepared by the formamide procedure.

Observations reported here as well as previously[17] show that dextran from B-512 has characteristics of linear rather than of much-branched molecules. As expected, a preliminary test showed that the triacetate of this dextran had film-forming ability. Although this film had low mechanical strength, it was superior to films from a branched material such as amylopectin triacetate.

Results and Discussion

Examination of the data shown in Table I discloses close relationship between the properties of the dextrans and their triacetates. The intrinsic viscosities of the triacetates followed the same sequence as those of their dextrans and, in general, increased with the apparent degree of linearity of the dextrans as indicated by periodate oxidation.[4] It is an exceptional coincidence that the intrinsic viscosities of the dextran triacetates were the same or very nearly the same as those of their dextrans, albeit acetylation to the triacetate resulted in an increase of 1.7 times in particle weight and the two solvents for viscosity measurements were very different. The slopes of plots of η_{sp}/c *vs.* c ranged from 1.8 for the B-1146 dextran triacetate to near zero for the B-742, fraction L, dextran triacetate.

The specific rotations of the dextrans varied from +234 to +214°, and of their corresponding triacetates from +199 to +219° (Table I). To facilitate consideration of the relationship among these specific rotations and the types and proportions of linkages found in these dextrans by periodate oxidation, the dextrans have been grouped into three classes. All the factors which influence these rotations may not yet be known.

The dextran from B-1355 and fraction C from B-742 have outstandingly high specific rotations which our data indicate to be approximately proportional to the relatively high percentages of 1,3-like linkages present. Specific rotations of the corresponding triacetates were outstandingly low, but were not strictly proportional to those of their dextrans. Infrared analysis of these dextrans has revealed a type of absorption not shown by dextrans containing only 1,6- and 1,4-like linkages[18] and which is quantitatively in agreement with the relative proportions of 1,3-like linkages indicated by periodate oxidation.

Dextrans such as those from B-512, B-1064

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and B-1146 showed the lowest specific rotations of all these dextrans, +214 to +215° and their triacetates showed the highest rotations, +218 to +219°. Periodate oxidation indicates that these dextrans have 5% or less of non-1,6-linkages and that, within the limitations of the method, these are of the 1,4-like type.

The dextrans from B-1254 and B-1299 and fraction L from B-742 constitute the third class of dextrans. Their specific rotations were +216 to +218° and those of their triacetates were +212 to +214°. Although these dextrans had 10% to 50% of non-1,6-linkages, they showed no detectable rotational dependence upon the content of these linkages. Periodate oxidation indicates these linkages to be 1,4-like, but this does not exclude the possibility of other types in small proportions. In fact, the rotational values of these three dextrans and of their triacetates, which were intermediate between those for the other two classes, suggest that small proportions of linkages, similar in influence to the 1,3-like, might be present.

The triacetates of dextrans having 50–80% 1,6-linkages decomposed at much lower temperatures than those from dextrans having higher proportions of these linkages. The behavior of the triacetate from the water-insoluble dextran B-523 was anomalous. These decomposition temperatures are believed to be indicative of the activation energies required to disrupt the forces of molecular aggregation.[19]

The observations reported here on the triacetates of nine bacterial dextrans of widely varying properties have established that differences in the dextrans manifested in values for periodate oxidation, specific rotation and intrinsic viscosity are due to fundamental structural characteristics which are carried through into the dextran triacetates.

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PEORIA, ILLINOIS

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Derivatives of L-Xylose

BY ROBERT K. NESS AND HEWITT G. FLETCHER, JR.

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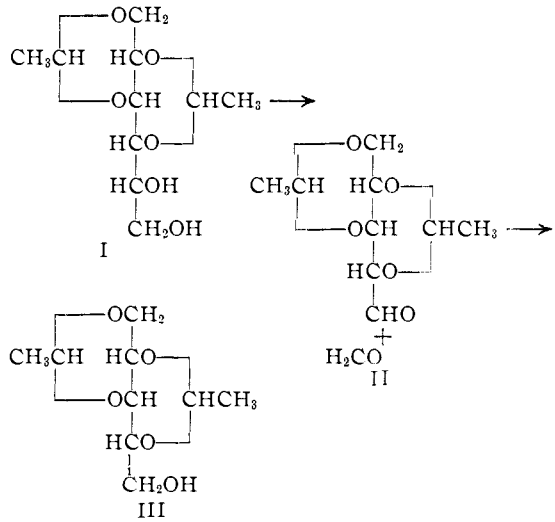
Improvements in the preparation of 1,3:2,4-diethylidene-D-glucitol and 2,4:3,5-diethylidene-L-xylose are described as well as various new derivatives of the latter. Crystalline 1,3:2,4-diethylidene-D-xylitol (= 2,4:3,5-diethylidene-L-xylitol) and some of its derivatives have also been prepared.

In the course of recent work, a quantity of 1,3:2,4-diethylidene-D-xylitol (III, synonym: 2,4:3,5-diethylidene-L-xylitol) was required as an intermediate. Since Hockett and Schaefer[1] have described the synthesis of 2,4:3,5-diethylidene-L-xylose (II) through 1,3:2,4-diethylidene-D-glucitol (I), this path of synthesis (I to III) was chosen.[1a]

The preparation of 1,3:2,4-diethylidene-D-glucitol, an important intermediate in the synthesis of L-xylose,[1] through the direct ethylidenation of D-glucitol has been studied by numerous authors.[1,2] By appropriate modification of the conditions of the reaction and through the use of ion-exchange resins we have raised the yield of crystalline 1,3:2,4-diethylidene-D-glucitol to 46%.

The oxidation of 1,3:2,4-diethylidene-D-glucitol (I) to 2,4:3,5-diethylidene-L-xylose (II) has been carried out with lead tetraacetate in acetic acid–benzene[2a] and in aqueous acetic acid.[1] While the work of Hockett and Schaefer[1] in this respect was fully confirmed, the product appeared to contain substances poisoning the platinum catalyst used in the succeeding hydrogenation. As an alternative, sodium metaperiodate in aqueous solution was employed as an oxidant and this proved to be preferable to lead tetraacetate for this purpose. Not only is the weight of sodium metaperiodate required less than half that of the lead tetraacetate but the



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(2) *Cf.* (a) H. Appel, *ibid.*, 425 (1935); (b) W. R. Sullivan, THIS JOURNAL, **67**, 837 (1945); (c) P. Bladon and L. N. Owen, *J. Chem. Soc.*, 591 (1950).