[CONTRIBUTION FROM THE STARCH AND DEXTROSE DIVISION, NORTHERN REGIONAL RESEARCH LABORATORY[1]]

D-Galactosan<1,4>α<1,6>: Its Structure and Resistance to Periodate Oxidation[2]

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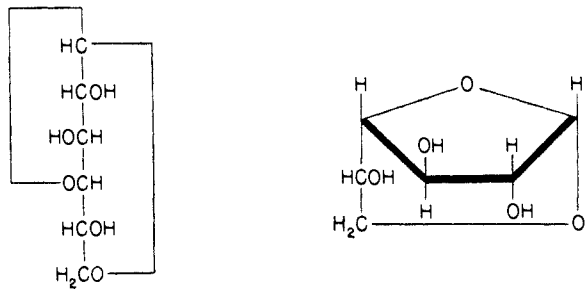
The unexpected resistance of the adjacent hydroxyl groups in D-glucosan<1,4>β<1,6> to periodate oxidation prompted a further search for compounds in the carbohydrate field showing similar anomalous behavior. The periodate resistant, dextrorotatory D-galactosan first obtained by Hann and Hudson now has been shown through methylation studies to have the structure D-galactosan<1,4>α<1,6>. The adjacent hydroxyl groups at C_2 and C_3 are resistant to oxidation by lead tetraacetate as well as by periodic acid and sodium periodate. This further example of the failure of periodate to cleave 1,2-diols emphasizes that resistance of a compound to periodate oxidation is not in itself proof of the absence of vicinal hydroxyl groups.

The ability of periodic acid to cleave the carbon–carbon bond in 1,2-glycols by oxidation has been utilized extensively in the study of carbohydrate structure.[3] Failure of periodic acid to react with polyhydroxy compounds under the usual analytical conditions was accepted as proof of the absence of vicinal hydroxyl groups.

In 1946, however, Dimler and co-workers[4] discovered a new sugar anhydride D-glucosan<1,4>β<1,6>, containing adjacent hydroxyl groups which resisted oxidation by periodic acid. A similar anomalous behavior was exhibited by D-glucosaccharo-1,4-lactone,[5] although in this particular case it is to be noted that the test was a qualitative one and was of only one hour duration. In both of these compounds the adjacent hydroxyl groups are *trans* as shown in the Fischer and Haworth structural formulas and in the molecular models.

A third example of this irregular behavior of *trans*-glycols now has been found in the dextrorotatory D-galactosan isolated by Hann and Hudson[6] in 1941. The ring system of this compound had been inferred to be <1,5>β<1,3> because of its resistance to oxidation by periodate. The similarity, however, between the D-galactosan and D-glucosan <1,4>β<1,6> with respect to chemical properties and method of preparation led us to consider the possibility of the galactosan being similar also in structure. Through arrangement with Prof. C. S. Hudson we undertook, therefore, the further examination of its structure and properties.

The structure of the D-galactosan proved to be D-galactosan<1,4>α<1,6>, (I), containing a *trans* adjacent hydroxyl grouping. Evidence for this configuration was obtained by preparation of the trimethyl-D-galactosan which, on hydrolysis, yielded the known sirupy 2,3,5-trimethyl-D-galactose. The latter was characterized by its conversion to the crystalline 2,3,5-trimethyl-D-galactono-γ-lactone[7] which in turn was converted to the crystalline amide of 2,3,5-trimethyl-D-galactonic acid.



(I) D-Galactosan<1,4>α<1,6>

The physical constants of 2,3,5-trimethyl-D-galactose and its derivatives distinguish it unmistakably from the 2,4,6-trimethyl-D-galactose which would have been obtained if the D-galactosan ring system had been <1,5>β<1,3>. The α-configuration of the <1,6> ring, in which the glycosidic oxygen has the same configuration as the hydroxyl on C_5, is assumed because only this structure can be built with molecular models.

The hydroxyl groups on carbon atoms 2 and 3 of D-galactosan<1,4>α<1,6> are resistant both to lead tetraacetate and to periodate under the conditions usually prescribed for the detection and determination of adjacent hydroxyl groups. In this case, as with the analogous D-glucosan<1,4>β<1,6>, the failure of oxidation can be considered to result from the relative *trans* arrangement of the adjacent hydroxyl groups together with the structural rigidity resulting from the introduction of a double ring system into the molecule. The hydroxyl groups thus are prevented from shifting to the relative positions necessary for combination with the oxidant in some such intermediate as a chelate complex. In view of this apparent effect of the spatial arrangement of adjacent hydroxyl groups, caution should be exercised in putting full reliance on negative periodate oxidation as evidence of the absence of 1,2-glycol groupings.

The possibility exists, of course, that the <1,4><1,6> double lactol ring system is the sole cause of the failure of periodate oxidation for the galactosan and glucosan. In that case, the corresponding mannosan, D-mannosan<1,4>β<1,6>, should also be resistant to periodate oxidation, in spite of the relatively *cis* arrangement of the adjacent hydroxyl groups. The preparation of this unreported mannosan and studies on its behavior toward oxidation would provide more specific evidence of the influence of ring systems and *cis–trans* arrangement of the hydroxyl groups on the periodate oxidation of 1,2-glycols.

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

(2) This paper is based upon the thesis submitted by Benjamin H. Alexander in June, 1950, to the Graduate School of Bradley University, in partial fulfillment of the requirements for the degree of Master of Science.

(3) E. L. Jackson (Adams, Editor-in-chief), "Organic Reactions," Vol. II, John Wiley and Sons, Inc., New York, N. Y., 1944, pp. 341–375.

(4) R. J. Dimler, H. A. Davis and G. E. Hilbert, THIS JOURNAL, **68**, 1377 (1946).

(5) F. Smith, *J. Chem. Soc.*, 633 (1944).

(6) R. M. Hann and C. S. Hudson, THIS JOURNAL, **63**, 2241 (1941).

(7) S. Luckett and F. Smith, *J. Chem. Soc.*, 1106 (1940).

Experimental

Pyrolysis of α-D-Galactose.—The pyrolysis[8] of 85 g. of α-D-galactose, melting at 165–168°, was carried out in a 1-liter round-bottom flask with a side arm. The flask was enclosed in an electrically heated jacket. The neck of the flask was fitted with a one-hole rubber stopper bearing a tube leading to a manometer. A 250-ml. filter flask served as the receiver, and was connected through a Dry Ice trap to a vacuum pump. The system was evacuated to 2 mm. of mercury and the heating carried out at such a rate that the electric heater reached 264° in 25 minutes. When the distillate began to collect in the receiver, the latter was placed in a water-bath at 50°. The temperature of the heater was slowly raised to 378° and maintained there until the distillation was complete (one hour). The yield of brown, viscous pyrolysate was usually 19–22 g. [54 g. of pyrolysate will give about 3 g. of pure D-galactosan<1,4>α<1,6>].

Isolation of the Crude D-Galactosan<1,4>α<1,6>.—Fifty-four grams of pyrolysate, collected from several runs, was acetonated by the addition of 450 ml. of anhydrous acetone and 45 g. of anhydrous copper sulfate. The reaction flask was shaken for 15 hours at room temperature. During this period the pyrolysate, initially incompletely soluble in the acetone, went completely into solution. The copper sulfate was filtered from the reaction mixture and the combined filtrate and washings were shaken with 40 g. of anhydrous copper sulfate for 35 hours. The reaction mixture was filtered and the filtrate concentrated *in vacuo* to a thin sirup.

Chloroform, 300 ml., was added to the thin sirup to dissolve the undesired 3,4-isopropylidene-D-galactosan. The mixture was cooled to 5° and the residue remaining after decantation of the chloroform was dissolved in cold water and thoroughly extracted with chloroform. The combined water layers, after filtering, were concentrated *in vacuo* to a light brown sirup which contained the unsubstituted D-galactosan<1,4>α<1,6>.

2,3,5-Triacetyl-D-galactosan<1,4>α<1,6>.—The sirup was dried by the repeated concentration *in vacuo* of its solution in pyridine. Acetic anhyride, 120 ml., was added to a solution of the dry sirup in pyridine at 0°. After 24 hours at room temperature the acetylation mixture was concentrated *in vacuo*. The nearly black sirup (31.5 g.), was distilled at or below 0.1 mm. pressure. The distillate was collected in three fractions, 0.6 g., 2.3 g. and 14.7 g., over the temperature ranges 70–90°, 90–132° and 133–140°, respectively. The latter two fractions, which crystallized on standing, were recrystallized from three parts isopropyl alcohol. The product, 6.5 g., was 2,3,5-triacetyl-D-galactosan<1,4>α<1,6>.

Anal. Calcd. for $C_{12}H_{16}O_8$: acetyl, 44.8. Found: acetyl, 44.8.

The pure triacetate melts at 79–80°; $[\alpha]^{25}D$ + 144.9° (*c* 1, $CHCl_3$). The melting point of the triacetate was unchanged by admixture with a sample of Hann and Hudson's triacetate (m.p. 79–80°).

D-Galactosan<1,4>α<1,6>.—Catalytic deacetylation of 4 g. of 2,3,5-triacetyl-D-galactosan<1,4>α<1,6> with barium methylate gave 2 g. of D-galactosan (87% of theory). Approximately 0.5 g. of the first crop of crystals, melting at 183–184°, was recrystallized from 10 parts absolute ethanol to obtain D-galactosan<1,4>α<1,6>; m.p. 183–184°, $[\alpha]^{25}D$ +56° (*c* 1, H_2O).

Anal. Calcd. for $C_6H_{10}O_5$: C, 44.4; H, 6.2. Found: C, 44.4; H, 6.0.

This D-galactosan preparation melts higher than the D-galactosan reported by Hann and Hudson, apparently because of more complete purification when prepared *via* the triacetate. The melting point of a mixture of this D-galactosan with a sample of D-galactosan (m.p. 174–177°) furnished by Prof. Hudson was between the values obtained for the individual substances.

2,3,5-Trimethyl-D-galactosan.—A 2-g. sample of D-galactosan<1,4>α<1,6> was partially methylated by using dimethyl sulfate and sodium hydroxide, then completely methylated by two treatments with methyl iodide and silver oxide.[9] The resulting thin sirup (2.3 g., theory 2.5 g.) was distilled at 0.5 mm. pressure. Two fractions were collected, 0.6 g. and 1.4 g., distilling at 72–83° and 84–90°, respectively.

Anal. Calcd. for $C_9H_{16}O_5$: OCH_3, 45.6. Found: first fraction 45.7, second fraction 44.9.

A similarly prepared 2,3,5-trimethyl-D-galactosan<1,4>α<1,6> had $[\alpha]^{25}D$ +73.6° (*c* 3, CH_3OH) and $n^{25}D$ 1.4579.

The D-galactosan<1,4>α<1,6> was unusually difficult to methylate with methyl iodide and silver oxide. Later experiments, however, showed that it was completely methylated after two treatments with sodium hydroxide and dimethyl sulfate.

2,3,5-Trimethyl-D-galactonolactone.—One gram of trimethyl-D-galactosan was hydrolyzed by heating for six hours at 98° in 0.5 *N* hydrochloric acid. The specific rotation of the resulting trimethyl galactose was calculated to be −8° on the basis of the optical activity of the hydrolysis solution. Luckett and Smith[7] reported an equilibrium $[\alpha]^{15}D$ −5° for the sirupy 2,3,5-trimethyl-D-galactose in water.

The trimethyl-D-galactose was converted to the trimethyl-D-galactonolactone (0.53 g.) by bromine oxidation and sublimation. The crystalline 2,3,5-trimethyl-D-galactonolactone, after recrystallization from a mixture of ethanol and petroleum ether (b.p. 63–70°), melted at 91–92°; $[\alpha]^{25}D$ −35° (after one-half hour; *c* 0.6, H_2O).

Anal. Calcd. for $C_9H_{16}O_6$: OCH_3, 42.3. Found: OCH_3, 42.1.

Luckett and Smith[7] reported m.p. 90° and $[\alpha]^{16}D$ −37° (initial) for 2,3,5-trimethyl-D-galactonolactone. A mixture of a sample of their compound with our product melted at 90°.

2,3,5-Trimethyl-D-galactonamide.—Treatment of the crystalline lactone (0.12 g.) with methanolic ammonia for 24 hours at 5° readily gave the 2,3,5-trimethyl-D-galactonamide, which crystallized on removal of the solvent; m.p. 156–157.5°. After recrystallization from absolute ethanol, the amide melted at 162–163°; $[\alpha]^{25}D$ +5° (*c* 0.5, H_2O).

Anal. Calcd. for $C_9H_{19}O_6N$: C, 45.6; H, 8.0; N, 5.9; OCH_3, 39.2. Found: C, 45.4; H, 8.0; N, 6.0; OCH_3, 39.1.

This melting point is higher than that (152°) reported by Luckett and Smith[7] for the amide; however, the optical rotations are essentially the same, *ca.* +3° compared to +5° for our amide. A mixture of their amide and our product had an intermediate melting point of 156–158°.

Resistance of D-Galactosan<1,4>α<1,6> to Periodic Acid and Lead Tetraacetate Oxidation.—Samples of D-galactosan<1,4>α<1,6> were treated with periodic acid, sodium periodate and lead tetraacetate essentially as described by Dimler, *et al.*[4] Analysis of aliquots of the oxidation mixtures showed that no reaction occurred over a period of seven days.

Acknowledgment.—Grateful acknowledgment is made to Prof. C. S. Hudson for his encouragement and interest in the planning and progress of this work, and to Dr. F. Smith for samples of derivatives of 2,3,5-trimethyl-D-galactonic acid. Thanks are also expressed to Mr. C. H. Van Etten and Mrs. Mary Wiele for the microanalyses.

PEORIA, ILL. RECEIVED MAY 19, 1951

(8) The pyrolysis was carried out according to the directions in an unpublished report "Levoglucosan" by I. A. Wolff and D. W. Olds at this Laboratory.

(9) F. J. Bates and Associates, "Polarimetry, Saccharimetry and the Sugars," U. S. Government Printing Office, Washington, 1942, pp. 506–507.