

Pyrolytic Reactions of Carbohydrates. Part I. Mutarotation of Molten D-Glucose

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The mutarotation of α - and β -D-glucose near their melting points has been studied in the neat materials as well as in the presence of water, and at low temperature in the presence of H_3BO_3 and $NaHCO_3$. The process of mutarotation was found to become very fast as soon as the bulk of the material melted.

THE investigation of chemical changes in solvent-free organic systems has begun to attract attention recently.^{1,2} The study of the thermal behaviour of neat sugars and sugar derivatives is important for a variety of practical applications and especially for the problem of decomposition of cellulose and similar compounds in wood, which is probably of vital importance in the spreading of forest fires.³

Glucose, both by itself and as a basic unit of polysaccharides, is probably the most widely occurring sugar in Nature⁴ and there is some evidence⁵ that several decomposition products of D-glucose and of some oligo- and poly-saccharides derived from it are similar or even identical in nature, although differing in relative amounts. Hence, the choice of D-glucose as a model substance seems to be justified.

D-Glucose, when heated at 130° for long periods⁶ or above its melting point for shorter periods, yields "anhydro-sugars" and higher polysaccharides.⁶⁻⁹ Among the identified products are levoglucosan, 5-hydroxymethylfurfural, and di- and tri-saccharides.¹⁰ At higher temperatures, carbon-carbon cleavage occurs and gaseous products are formed.⁶ Reports relating to α - and β -D-glucose seem to indicate (though not clearly) similar or slightly differing^{8,9,11,12} reactivities in the pyrolytic behaviour of the two anomers. As the first stage of a systematic study of the thermal reactions of D-glucose we investigated its mutarotation in neat samples, and in the presence of water and of two catalysts, under conditions which gave hardly any intrusion of other reactions.

EXPERIMENTAL

Materials.— α -D-Glucose (B.D.H., AnalaR), m. p. 146° [α]_D²⁰ (in H₂O) +110.9° $\rightarrow$ +52.5° and β -D-glucose (Nutritional Biochemical Co.), m. p. 150° [α]_D²⁰ (in H₂O) +20.3° $\rightarrow$ +52.4° were used. The amount of water (by drying at 110° to constant weight^{13a}) was found to be

0.17% in the α -form and in the β -form about 0.15%. Literature values of [α]_D for the α -anomer range from +109° to +113° and for the β -anomer from +17.5° to +20.5°^{13b,14-17} though all these authors seem to have worked with quite pure materials, as can be judged from the excellent uniformity of the final equilibrium value in water ([α]_{D, equil} +52.6° $\pm$ 0.1°) given in all these sources.

General Procedure. Samples of 2.0 g. each of α - or β -D-glucose in small open Pyrex tubes were pre-heated to 135–140° in an oven for 15 min., showing no change in this period. The samples were then heated in an oil-bath for predetermined times and, after cooling, weighed to ± 1 mg. Aliquot portions were taken for the measurements described below. Runs were made at various temperatures around the melting point of glucose; some experiments were also made with samples of α -D-glucose containing water, sodium hydrogen carbonate, or boric acid.

Determination of the Specific Optical Rotation of Glucose Melts.—The glucose was dissolved in water to give solutions of 0.04–0.06 g./ml. Zero time was taken at the middle of the period needed for complete dissolution of the sample. The specific optical rotation of the neat sample, [α]_{D, neat} was obtained by plotting $\log(\alpha_t - \alpha_\infty)$ versus time (where α_t is the observed optical rotation at time t , and α_∞ is the final observed optical rotation) and extrapolating the straight line so obtained to zero time. The value of [α]_{D, equil}, i.e., the final specific optical rotation of each sample in aqueous solution, was determined after 24 hr. Simultaneously with the mutarotation, higher sugars (mainly disaccharides) are formed.^{8,10} Since, nevertheless, the $\log(\alpha_t - \alpha_\infty)$ vs. time plots were good straight lines it can be assumed that new substances are formed in the melt in a state close to optical equilibrium.

Analyses.—We prepared the trimethylsilyl derivatives of the anomers for gas-chromatographic analysis.¹⁸ An Aerograph A 700 gas chromatograph with a katarometer detector was used, with a copper column (2.7 m. $\times$ 6.35 mm. i.d.). The stationary phase at 225° was 20% w/w silicon DC 11 on Chromosorb W, and the carrier gas was helium (170 ml./min.), 40 μ l. of the solution of trimethylsilyl derivatives was injected. The amount of α - or β -D-glucose

¹⁰ H. Sugisawa and H. Edo, *Chem. and Ind.*, 1964, 892.

¹¹ H. Morita, *Analyt. Chem.*, 1956, **28**, 64.

¹² H. Morita and H. M. Rice, *Analyt. Chem.*, 1955, **27**, 336.

¹³ C. A. Browne and F. W. Zerban, "Physical and Chemical Methods of Sugar Analysis," Wiley, New York, 3rd edn., 1941, (a) p. 40; (b) p. 263ff.

¹⁴ "The Merck Index of Chemicals and Drugs," 7th edn., Merck and Co. Inc., 1960, p. 484.

¹⁵ "Handbook of Biological Data," ed. W. S. Spector, National Academy of Sciences, National Research Council, W. B. Saunders Co., 1956.

¹⁶ C. S. Hudson and E. Yanovsky, *J. Amer. Chem. Soc.*, 1917, **39**, 1035.

¹⁷ I. M. Heilbron and H. M. Bunbury, "Dictionary of Organic Compounds," Eyre and Spottiswoode, London, 1943, vol. II.

¹⁸ C. C. Sweeley, R. Bently, M. Makita, and W. W. Wells, *J. Amer. Chem. Soc.*, 1963, **85**, 2497.

¹ "Physics and Chemistry of the Organic Solid State," eds. D. Fox, H. H. Labes, and A. Weissberger, Interscience, New York and London, 1963, vol. I, chs. 4 and 5.

² Much material can be found in the Proceedings of International Symposia on the Reactivity of Solids, held usually at four-yearly intervals, sponsored by I.U.P.A.P. and I.U.P.A.C.

³ A. Broido and S. B. Martin, *Fire Research Abs. Reviews*, 1961, vol. 3, No. 3, 193.

⁴ "The Carbohydrates," ed. W. Pigman, Academic Press, 1957, p. 77.

⁵ D. J. Bryce and C. T. Greenwood, *Stärke*, 1963, 359.

⁶ I. E. Puddington, *Canad. J. Res.*, 1948, **B**, 26, 413.

⁷ A. Pictet and P. Castan, *Helv. Chim. Acta*, 1920, **3**, 645.

⁸ C. D. Hurd and O. E. Edwards, *J. Org. Chem.*, 1949, **14**, 680.

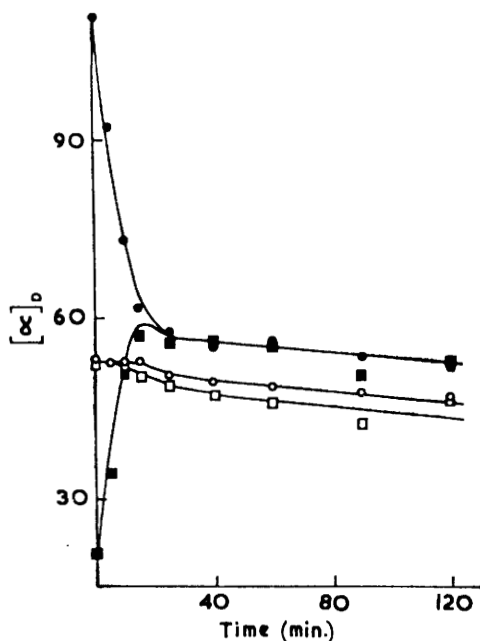
⁹ J. W. Liskowitz, *Diss. Abs.*, 1962, **23**, 1945.

was determined from the peak heights, which were found under these conditions to be linearly proportional to the amount of the anomers in the injected sample.

Infrared spectra were determined in Nujol using a Perkin Elmer model 337 spectrophotometer.

RESULTS

Mutarotation of Pure α - or β -Glucose.—(a) α -D-Glucose ($[\alpha]_{D, \text{neat}} +110.9^\circ$) was heated at $140^\circ \pm 2^\circ$ and showed no change after 30 min., but the specific optical rotation fell to $[\alpha]_{D, \text{neat}} +91.9^\circ$ after 6 hr., while the $[\alpha]_{D, \text{equil}}$ of the sample was $+51.6^\circ$, i.e., showing only a slight change, hardly above the experimental error, although the sample became pale yellow and was partially sintered. Total weight loss was less than 0.5%. Gas-liquid chromatography showed the ratio of α - to β -D-glucose in the tested



Plot of $[\alpha]_D$ values vs. time of heating at 151° : ● — $[\alpha]_{D, \text{neat}}$ starting from α -D-glucose. ○ — $[\alpha]_{D, \text{equil}}$ starting from α -D-glucose. ■ — $[\alpha]_{D, \text{neat}}$ starting from β -D-glucose. □ — $[\alpha]_{D, \text{equil}}$ starting from β -D-glucose

sample to be 4.17, in good agreement with the ratio corresponding to the experimental $[\alpha]_{D, \text{neat}}$ value.

(b) At 151° samples melted after 2 min. and reached the oil-bath temperature after 10 min. The samples turned pale yellow after about 15 min. (weight loss $\sim 0.25\%$) and became brown in about 2 hr. (weight loss $\sim 0.5\%$). The Figure shows the values of $[\alpha]_{D, \text{neat}}$ and $[\alpha]_{D, \text{equil}}$ and Table 1 shows the ratios of α/β after various reaction times for both α - and β -D-glucose, and the sums ($\alpha + \beta$).

(c) At 165° the α -D-glucose sample melted in about 1 min. and after 10 min. mutarotation was practically complete. Some decomposition was shown by the determined $[\alpha]_{D, \text{equil}}$ values (e.g., $[\alpha]_{D, \text{equil}}$ of 52.5° , 52.5° , 51.8° , and 47.5° at 0, 10, 15, and 30 min., respectively). The sample became yellow after 10 min. and yellowish-brown after 30 min. (weight loss, $\sim 0.4\%$).

Equilibrium of Anomers in the Molten State.—Table 1 shows the ratio of α - to β -D-glucose in the melt at 151° at various times, determined by gas-liquid chromatographic

analysis of the trimethylsilyl derivatives. The experimental error in the estimation of the sum $\alpha + \beta$ (i.e., the total amount of glucose anomers in the melt) was about $\pm 8\%$. This inaccuracy does not effect the determination of the α/β ratio in a single analysed sample, (given by the ratio of peak heights), which was better than $\pm 2\%$. Reaching the constant value of α/β after 15 min. can be taken as reliable proof that at this time equilibrium between the anomers has been reached. The corresponding values of $\alpha + \beta$, although much less reliable, leave no doubt that the bulk of the material at this stage is still glucose.

TABLE 1

Ratios and total amounts of α - and β -D-glucose after various heating times

Time (min.)	Starting from α		Starting from β	
	α/β	$\alpha + \beta$ (%)	α/β	$\alpha + \beta$ (%)
5	3.879	92	0.234	105
10	1.204	103	0.536	107
15	0.792	99	0.792	108
25	0.784	92	0.801	89
40	0.779	88	0.781	80
60	0.775	88	0.782	80
90	0.801	78	0.804	71
120	0.802	68	0.803	77

Both at 151° and at 165° , even when considerable decomposition of the glucose occurs, the α/β equilibrium values still remain constant, corresponding to 44% of α - and 56% of β -D-glucose in the melt. The specific optical rotation of such an anomer mixture should be $[\alpha]_{D, \text{neat}} + 60.2^\circ$, in excellent agreement with the value of $[\alpha]_{D, \text{neat}} + 60^\circ \pm 0.5^\circ$, obtained from the Figure by continuing the experimental line of the $[\alpha]_{D, \text{neat}}$ values to zero time (so that the $[\alpha]_{D, \text{neat}}$ line should remain parallel with the $[\alpha]_{D, \text{equil}}$ line). This value seems to be unaffected by the temperature of the melt. Tollens¹⁹ has shown that the equilibrium specific optical rotation in water of a mixture of D-glucose anomers depends only on their concentration and not on the temperature, and follows the equation: $[\alpha]_D = +52.5 + 0.018796p + 0.00051683p^2$, where p is the w/w concentration. Tollens tested his equation up to 90% concentration of glucose and his value for $p = 100$ would be $+59.5^\circ$, in good agreement with our experimental value.

Infrared Spectra.—Earlier results²⁰ were reproduced in reasonable agreement for α and β -D-glucose. The spectra of the molten equilibrium mixtures obtained by heating either α - or β -D-glucose were practically identical with the spectrum of a mixture prepared from the two pure anomers in the appropriate proportion.

Attempts were made to use the data in Table 2 for proof of the presence or absence of some probable reaction products. 5% of 1,6-anhydro- β -D-glucopyranose (levoglucosan) was easily recognisable by peaks at 862, 932, and 950 cm^{-1} when added to an equilibrium mixture of the two glucose anomers. On the other hand, admixture of 10% of maltose or of cellobiose did not change the infrared spectra of the equilibrium mixture perceptibly. Hence, the data in Table 2 give additional evidence for the equilibrium process, but do not exclude the presence of small

¹⁹ B. Tollens, *Ber.*, 1884, **17**, 2238.

²⁰ S. A. Barker, E. J. Bourne, R. Stephens, and D. H. Whiffen, *J. Chem. Soc.*, 1954, 3468.

amounts of 1,6-anhydro- β -D-glucopyranose or of disaccharides.

Experiments with Initially Added Water.—The mutarotation of α -D-glucose in the presence of 10% w/w of initially added water was determined at 149°. The sample melted in less than 1 min., and the equilibrium value was reached in about 10 min.

TABLE 2

Peaks in the region 750–1000 cm^{-1} at which anomeric configuration can be distinguished. Data in brackets are from ref. 20

D-Glucose		(cm. ⁻¹)			
α	917s (914)s			840s (837)s	777s (774)s
β	—	914m (909)m	902s (896)s	—	—
Mixture (40% α + 60% β)	918m	—	903s	841s	778s
α (Heated at 150° for 1 hr.)	918m	—	903s	841s	778s
β (Heated at 150° for 1 hr.)	917m	—	899s	838s	773s

TABLE 3

$[\alpha]_D$ values of glucose samples in the presence of initially added 10% w/w of water after various heating times at 149°

Time (min.)	0	10	30	120
$[\alpha]_D$, neat	110.9	60.9	57.4	54.2
$[\alpha]_D$, equil	52.5	52.5	48.6	47.2

Weight loss after 2 hr. was about 5.5%, showing that at least 4.5% w/w of water remained in the material.

Experiments with Sodium Hydrogen Carbonate and with Boric Acid.—Samples of α -D-glucose were heated at 130° for 15 min. in the presence of different amounts of sodium hydrogen carbonate and boric acid.

Checks on unheated samples containing 0.5% of either sodium hydrogen carbonate or boric acid showed that the

TABLE 4

Values of $[\alpha]_D$ after 15 min. heating at 130°, in the presence of various amounts of additives

Additive (%) w/w)	NaHCO ₃				H ₃ BO ₃		
	None	0.1	0.25	0.50	0.1	0.25	0.50
$[\alpha]_D$, neat	110.9	101.1	93.4	84.8	110.1	105.5	97.1
$[\alpha]_D$, equil	52.5	49.9	47.4	43.7	52.5	52.4	52.6

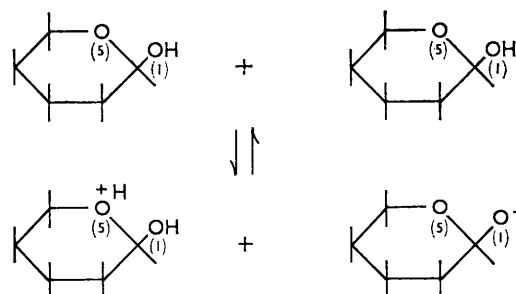
presence of these additives in the aqueous solution did not influence the equilibrium specific optical rotation values. The results in Table 4 indicate that both substances strongly catalyse the mutarotation, but boric acid does not catalyse side-reactions while the sodium salt does so strongly. In the presence of the acid the sample remained completely colourless, while in the presence of the salt fast browning occurred.

DISCUSSION

The concerted evidence presented by the Figure, by Table 1, by the infrared spectra, and by the small weight

loss show clearly that, in the conditions employed, mutarotation is practically complete before other reactions take place to any great extent. The α/β equilibrium is reached in about 15 minutes at 151° and in about 10 minutes at 165°, while the bulk of the sample is still glucose. Hence, the use of either of the anomers or any of their mixtures is probably permissible for the study of thermal reactions of glucose at higher temperatures and the same is possibly true also for anomer pairs of other sugars. The results show that there is no need for the presence of a solvent or of any foreign substance. The only other material present in the dried and preheated samples in the first stages of the reaction is a very small amount of water (<0.1%), although in the latter stages, more water is formed through side-reactions. Most probably the presence of water (Table 3) enhances the mutarotation rates, not by any specific catalytic effect but simply by hastening the process of melting. Hence, it has to be assumed, that the mutarotation may occur as a spontaneous reaction either unimolecularly, or more probably, catalysed by neighbouring like molecules.

Proton transfer between two glucose molecules may be most easily envisaged by assuming loss of the proton of the anomeric hydroxyl group (which is the most acidic site of the molecule), and its attachment to the ether oxygen (O-5, the most basic site) in a second glucose molecule. The enhanced ease of anomerisation of the O-5-protonated and the O-1 deprotonated intermediate is well documented, with a variety of detailed mechanisms having been proposed for either case.²¹ This Scheme is



not intended to exclude other protonated forms, but only the O-5-protonated form shown may undergo faster ring-opening than glucopyranose itself.

The mutarotation rates do not change drastically on addition of an equimolecular amount (10% w/w) of water, and we estimate from our results that the ratio of the half-times in the presence or absence of water, is probably about 2:1 or 3:1. Therefore, we favour the conclusion that the protonating ability of molten glucose does not differ very considerably from that of water. This conclusion is supported by the various estimates of the acidity of glucose.²²⁻²⁴

²¹ Y. Pocker, *Chem. and Ind.*, 1960, 968. C. G. Swain and J. F. Brown, *J. Amer. Chem. Soc.*, 1952, **74**, 2534; B. C. Challis, F. A. Long, and Y. Pocker, *J. Chem. Soc.*, 1957, 4679.

²² H. Euler, *Ber.*, 1906, **39**, 350.

²³ L. Michaelis and P. Rona, *Biochem. Z.*, 1913, **49**, 247.

²⁴ N. A. Ramaiah and S. S. Katiyar, *Proc. Ann. Conv. Sugar Technol. Assoc. India*, 1961, **29**, [2], 77; (*Chem. Abs.*, 1963, **59**, 14641).

The question arises as to whether mutarotation may also take place in the solid state. At all temperatures at which mutarotation could be experimentally observed, at least partial sintering of the material occurred, so that the reaction could have taken place in the molten surface layer. Nevertheless, we believe that, at least in part, the reaction can occur also in the solid state. This belief is founded on the results in the presence of boric acid at 130°, in which case the sintering was quite superficial, but nevertheless 4% inversion of α -glucose

occurred in the presence of 0.25% and 10% inversion in the presence of 0.50% of boric acid, while at the same time no observable side-reaction took place.

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