618. *Reactions at Position* 1 *of Carbohydrates. Part VI.* The Oxidation of* α- *and* β-D-*Glucose with Bromine.*

By I. R. L. Barker, W. G. Overend, and C. W. Rees.

The oxidation of α-D-glucose with bromine is shown to proceed mainly by way of anomerisation to β-D-glucose. Evidence is presented to suggest that the α-anomers of a range of pentoses and hexoses are also oxidised mainly by way of a rate-determining anomerisation. The oxidation of β-D-glucose with bromine involves a complexity hitherto unreported. A larger rate ratio (*ca.* 250) than that previously reported is obtained for the direct oxidation of α- and β-D-glucose.

The oxidations of acetaldehyde and D-glucose with bromine at low pH have been investigated, and the results are compared. The mechanisms of these oxidations are discussed.

The kinetics of the oxidation of D-glucose with bromine water were first investigated in detail by Bunzel and Mathews;[1] in all their experiments a large excess of D-glucose was used and bromine was analysed titrimetrically. They concluded that free bromine rather than hypobromous acid was the oxidant, that the tribromide ion was not an oxidant, and that the rate of oxidation was inversely proportional to the acidity. Isbell and his co-workers showed that δ-lactones were the primary oxidation products in buffered, slightly acid solution for the α- and β-anomers of several sugars.[2,3] The rates of oxidation of aldopentoses, aldohexoses, and disaccharides were measured at 0° in a buffer, pH 5—6, consisting of barium carbonate suspended in the aqueous reaction mixture, through which carbon dioxide saturated with bromine was passed.[4-6] In general, an excess of bromine was used, and the reaction rate was obtained by estimation of the sugar.

Isbell and his co-workers realised that study of the rate of oxidation was complicated by the interconversion of the various modifications of the sugars which exist in aqueous

* Part V, preceding Paper.

[1] Bunzel and Mathews, *J. Amer. Chem. Soc.*, 1909, **31**, 464.
[2] Isbell, *J. Res. Nat. Bur. Stand.*, 1932, **8**, 615.
[3] Isbell and Hudson, *J. Res. Nat. Bur. Stand.*, 1932, **8**, 327.
[4] Isbell and Pigman, *J. Res. Nat. Bur. Stand.*, 1933, **10**, 337.
[5] Isbell and Pigman, *J. Res. Nat. Bur. Stand.*, 1937, **18**, 141.
[6] Isbell, *J. Res. Nat. Bur. Stand.*, 1937, **18**, 505.

solution. Generally, the oxidation of the β-anomers was too fast for concurrent transformation into α-anomers to be significant, but this was not so with the α-anomers. Hence, the velocity constants for the mutarotation of a number of sugars were measured polarimetrically for comparison with the velocity constants for oxidation. The results were tabulated,[5] and the assessment of the importance of anomerisation in the oxidations was left to the reader. It has not been generally appreciated that, whereas the rates of oxidation presented were measured in the above buffer, the rates of mutarotation were measured in pure water; some authors [7,8] have even omitted to mention the complication introduced by anomerisation. Mutarotation is general-acid- and -base-catalysed; [9] it is difficult to assess the magnitude of the increase in rate of mutarotation which the heterogeneous buffer would bring about in comparison with water, since polarimetric measurement is scarcely possible. Isbell and Pigman's results did show [5] that, for thirteen sugars, the β-anomer was always oxidised faster than the α-form. The work now reported was undertaken to re-evaluate the importance of anomerisation in these reactions, both oxidation and anomerisation rates being measured in the same homogeneous aqueous buffered solutions under identical conditions. The behaviour of bromine with D-glucose in solutions of low pH prompted some preliminary studies of the bromine oxidation of acetaldehyde.

Some of these results have been the subject of preliminary communications.[10]

Experimental

Materials.—α- and β-D-glucose were prepared by Hudson and Dale's method.[11] β-D-Glucose had m. p. 148° and initial specific rotation $[\alpha]_D^{20}$ +19·7° (*c* 2·0 in H_2O) (lit.,[12] m. p. 148—150°, $[\alpha]_D^{20}$ +19·7°); its infrared spectrum (KBr disc) showed a well-defined peak at 891 $cm.^{-1}$, characteristic of β-anomers, and none of the absorption at 844 $cm.^{-1}$ characteristic of α-anomers.[13] α-D-Glucose had m. p. 146° and initial specific rotation $[\alpha]_D^{20}$ +111° (*c* 2·0 in H_2O) (lit.,[12] m. p. 146°, $[\alpha]_D^{20}$ +113·4°); its infrared spectrum showed strong absorption at 844 $cm.^{-1}$.

Acetaldehyde was analytical-reagent grade, shown by gas chromatography to be about 99% pure. AnalaR bromine was used without further purification. Hypobromous acid,[14] was stored in a dark bottle at 0° and was used within 24 hr. of being distilled in an all-glass apparatus coated with black paint.

Reaction media consisted of aqueous sulphuric acid of pH 0 and 1, and aqueous buffers, as follows: pH 2, 0·2M-sodium acetate and sulphuric acid; pH 3, 0·3M-potassium dihydrogen phosphate and phosphoric acid; pH 4 and 5, 0·2M-sodium acetate and acetic acid; pH 6 and 7, 0·3M-potassium dihydrogen phosphate and sodium hydroxide. These were prepared in 2 l. portions, using glass or calomel electrodes standardised with 0·05M-potassium hydrogen phthalate of pH 4, and stored at 0°. Negligible changes in these pH values resulted from addition of the amounts of hydrobromic acid formed in the present reactions.

Kinetic Measurements.—In most experiments a large excess of bromine was used, to minimise the influence of tribromide ions and the relative importance of anomerisation. Reactions were quenched by addition to aqueous sodium arsenite, and the excess of arsenite was determined iodometrically. D-Glucose was analysed spectrophotometrically with anthrone.[15] The intensity of the colour developed was measured at 625 mμ (Unicam S.P. 600) against blanks containing appropriate quantities of sodium arsenite, arsenate, and bromide. Calibration curves were constructed from standard solutions of D-glucose containing sodium arsenite, arsenate, and bromide, and were linear over the range 2·5—10 × 10^{-5} g. $ml.^{-1}$ of D-glucose.

[7] Green in "The Carbohydrates," ed. Pigman, 2nd edn., Academic Press, New York, 1957, p. 336.
[8] Bentley, *J. Amer. Chem. Soc.*, 1957, **79**, 1720.
[9] Brönsted and Guggenheim, *J. Amer. Chem. Soc.*, 1927, **49**, 2554.
[10] Barker, Overend, and Rees, *Chem. and Ind.*, 1960, 1297, 1298.
[11] Hudson and Dale, *J. Amer. Chem. Soc.*, 1917, **39**, 320.
[12] "International Critical Tables" McGraw-Hill, New York, 1927, Vol. 2, p. 347 *et seq.*
[13] Barker, Bourne, Stacey, and Whiffen, *J.*, 1954, 171.
[14] Branch and Jones, *J.*, 1954, 2317.
[15] Trevelyan and Harrison, *Biochem. J.*, 1952, **50**, 298.

Anthrone analyses were performed in duplicate or triplicate. The reaction product, D-glucono-δ-lactone, gave no colour in this analysis, as noted previously.[16] Rates of mutarotation were measured polarimetrically with a Hilger standard polarimeter using a 2 dm. tube with a jacket.

TABLE 1.

Rates of mutarotation of α- and β-D-glucose in the pH 5 buffer.

Anomer	Glucose (M)	Temp.	$10^5(k_1 + k_{-1})$ (sec.$^{-1}$)	10^5k_1 (sec.$^{-1}$)
β	0·11	0°	5·64	2·01
α	0·11	0	5·49	3·50
α	0·02	0	5·72	3·71
α	0·02	10	17·0	11·1
α	0·02	20	49·5	32·3
α	0·02	25	80·7	52·4

TABLE 2.

Rates of mutarotation of α-D-glucose at 20°.

pH	Buffer	$10^4(k_1 + k_{-1})$ (sec.$^{-1}$)	10^4k_1 (sec.$^{-1}$)
4	0·2M-Sodium acetate and acetic acid	5·80	3·79
5	,, ,, ,,	4·95	3·23
6	,, ,, ,,	4·95	3·23
6	0·3M-Potassium dihydrogen phosphate and sodium hydroxide	24·6	16·0

TABLE 3.

The corrected second-order rate constant for the oxidation of α-D-glucose (0·005M) with bromine at 0° (see Fig. 1).

10^2[Bromine] (M)	10^5k_1 (sec.$^{-1}$)	$10^5k_{corr.}$ (sec.$^{-1}$)	$10^5k_2 = 10^5k_{corr.}/[Br_2]$ (l. mole^{-1} sec.$^{-1}$)
2·5	3·88	0·38	15·2
5·0	4·28	0·78	15·6
10·0	5·06	1·56	15·6
15·0	5·84	2·34	15·6

TABLE 4.

Oxidation of α-D-glucose (0·005M) with bromine (0·05M) at pH 5.

Temp.	0°	10	20	25
10^5k_1 (sec.$^{-1}$) ...	4·3	13·2	39·0	63·7

TABLE 5.

Oxidation of α-D-glucose (0·005M) with bromine (*ca.* 0·10M) at 20°; buffer solutions as in Table 2.

pH	4	5	6 (acetate)	6 (phosphate)
10^4k_1 (sec.$^{-1}$) ...	2·54	4·11	6·91	16·41

TABLE 6.

Second-order rate coefficients for the reaction between β-D-glucose and bromine in buffer of pH 5, at 0°.

10^2[Glucose] (M)	0·5	0·5	0·5	1·0	1·0	1·0	5·0
10^2[Bromine] (M)	1·03	2·3	5·1	1·03	1·43	5·0	0·55
10^2k_2 (l. mole^{-1} sec.$^{-1}$)	2·6	2·5	1·8	3·9	3·6	2·1	4·2

TABLE 7.

The reaction between acetaldehyde (0·05M) and bromine (0·005M) in the aqueous buffers at 20°.

pH	0	1	2	3	4	5	6	1 [a]	1 [b]
10^4k_1 (sec.$^{-1}$)	1·9 [c]	6·24	6·48′	6·91	7·89	9·21 [d]	1·44 [e]	0·887	1·95

[a] 0·05M-Potassium sulphate added. [b] 0·10M-Potassium bromide added. [c] k_1 increased 25% after 60% reaction. [d] k_1 had fallen 10% after 80% reaction. [e] k_1 had fallen 60% after 50% reaction.

Kinetic Results.—These are summarised in Tables 1—7. The rates of mutarotation of α- and β-D-glucose in the buffers used are given in Tables 1 and 2; the Arrhenius activation energy for the pH 5 buffer, obtained graphically, was 17·3 kcal. mole.$^{-1}$ (cf. ref. 17).

[16] Lichtin and Saxe, *J. Amer. Chem. Soc.*, 1955, **77**, 1875.
[17] Isbell and Pigman, *J. Org. Chem.*, 1937, **1**, 505.

The effect of changing the initial bromine concentration upon the first-order rate constant for the oxidation of α-D-glucose is shown in Fig. 1. The order with respect to bromine is considerably less than unity; extrapolation to zero [Br_2] gives a rate constant, $k_1 = 3{\cdot}5 \times 10^{-5}$ sec.$^{-1}$, for " oxidation," almost identical with that obtained polarimetrically for the conversion of α- into β-D-glucose under the same conditions (Table 1). The dependence of the increment in rate constant, after correction for the bromine-independent reaction (*i.e.*, anomerisation), upon the initial bromine concentration is illustrated in Table 3. Measurement of the overall rate of oxidation of α-D-glucose at four temperatures (Table 4) gave an activation energy, obtained graphically, of 17·8 kcal. mole.$^{-1}$. The effect of pH and buffer composition on the oxidation of the α-anomer is shown in Table 5.

Even when bromine was in large excess, the kinetics of the oxidation of β-D-glucose were complex. Under all conditions the rate coefficients diminished markedly during the course of the reaction, *e.g.*, for β-D-glucose (0·005M) and bromine (0·05M), k_1 decreased by 50% during

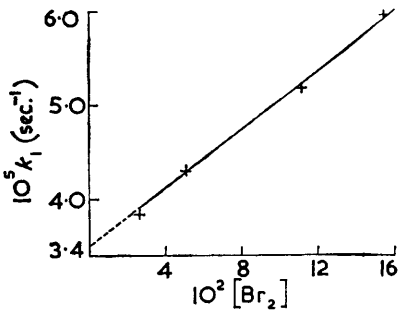


FIG. 1. First-order rate constants for the oxidation of α-D-glucose (0·005M) as a function of the initial bromine concentration.

80% reaction. Attempts to calculate, by standard methods, the kinetic order in both bromine and β-D-glucose, with respect to time and to concentration, further indicated the kinetic complexities, and the rate coefficients given in Table 6 were obtained from initial slopes on time–concentration graphs.

The rate of reaction of β-D-glucose (0·05M) with hypobromous acid (0·0055M) was determined by estimation of the latter with sodium arsenite and iodine. The reaction was initially very slow but gradually accelerated, the onset of the rapid reaction coinciding with the appearance of bromine in the mixture. The second-order rate constant for the initial reaction, calculated from initial slopes, was $k_2 = 3{\cdot}9 \times 10^{-4}$ l. mole^{-1} sec.$^{-1}$; this is about 100 times smaller than the oxidation with bromine under similar conditions. (This factor was erroneously reported as 1000 times smaller in a preliminary communication.[10])

The rate of oxidation of anomerically equilibrated D-glucose (0·05M) with bromine (0·005M) in aqueous sulphuric acid of pH 0 at 30° was also measured, in the same way. The result was unusual in that the reaction was first-order in total bromine ($Br_2 + Br_3^-$); the rate constant, obtained graphically, was $k_2 = 8{\cdot}9 \times 10^{-4}$ l. mole^{-1} sec.$^{-1}$.

The rates of the reaction between bromine (0·005M) and acetaldehyde (0·05M) in the standard buffer solutions at 20° were measured titrimetrically; first-order rate constants are given in Table 7. These reactions were also first-order in total bromine, except where noted.

DISCUSSION

Present results show that the importance of anomerisation in the overall rate of oxidation of glucose in the α-form is considerably greater than was formerly supposed. Isbell and Pigman [4] state that α-D-glucose is oxidised about five times faster than it is converted into the β-form. We find that, even with a ten-fold excess of bromine, " direct " oxidation (see below) of the α-form is only a minor reaction, most of the oxidation proceeding through anomerisation. A 45-fold molar excess of bromine would be required (Fig. 1) to make the rate of direct oxidation equal to that of anomerisation, in the aqueous acetate buffer of pH 5. Increasing the initial concentration of bromine increases the proportion

of direct oxidation at the expense of anomerisation. The former estimate [4] of the rate of direct oxidation of α-D-glucose, although corrected for mutarotation, is considered unreliable since the anomerisation and oxidation rates were obtained in different media, and, although the total bromine ($Br_2 + Br_3^-$) content was often in sufficient excess for first-order rate constants to be calculated, the free bromine was not. In many experiments the concentration of free bromine was less than that of the sugar (see, *e.g.*, ref. 4, pp. 348 and 350). Such a deficiency could only increase the importance of anomerisation. The value obtained [4] for the rate constant at 0° for direct oxidation of α-D-glucose ($k_2 = 9{\cdot}98 \times 10^{-4}$ l. mole^{-1} sec.$^{-1}$) was about six times greater than that found in the present work ($k_2 = 1{\cdot}56 \times 10^{-4}$ l. mole^{-1} sec.$^{-1}$). We conclude that the measured rate of oxidation of α-D-glucose is very largely that of its conversion into the β-anomer, and this is supported by very similar values (17·8 and 17·3 kcal. mole^{-1}) obtained for the activation energies of the " oxidation " and anomerisation reactions.

Measurement of the direct oxidation of the α-form by bromine in aqueous solution under conditions where anomerisation is unimportant appears to be very difficult, since a 45-fold excess of bromine is required merely to equalise the first-order rate constants for the two processes in our system, where the rate of anomerisation ($k_1 = 3{\cdot}5 \times 10^{-5}$ sec.$^{-1}$ at 0°) is only twice that in pure water ($k_1 = 1{\cdot}99 \times 10^{-5}$ sec.$^{-1}$ at 1·5°).[4]

The anomerisation of α-D-glucose is accompanied by a much slower reaction involving bromine, which is first-order in each reactant. Since this forms such a small part of the total oxidation reaction a study of its mechanism is difficult. Although referred to above as " direct " oxidation, in contrast to oxidation following anomerisation, this reaction could possibly be a bromine-catalysed anomerisation. To settle this point the rates of oxidation of α-D-glucose in buffers of pH 4—6 were measured. It is known that the rate of oxidation [1,18] of β-D-glucose is strongly dependent upon pH whereas the rate of mutarotation [19] is not, over the pH range *ca.* 3—6. If we assume that the direct oxidations of α- and β-D-glucose with bromine follow a similar mechanism, then the rate of direct oxidation of α-D-glucose should be inversely proportional to the acidity. Although changes in the oxidation rate with pH are in the right direction they are smaller than expected. The rate increase on passing from pH 5 to 6 is only about 40% of the expected value; at pH 4 the first-order rate constant for total oxidation is less than that for mutarotation. Comparison of the rates of oxidation and mutarotation in the two different buffers of pH 6 (Tables 2 and 5) demonstrates once more the importance of anomerisation in the oxidation of α-D-glucose.

As the measured oxidation rate of α-D-glucose is very largely that of its conversion into the β-anomer, it is possible that the literature [5] oxidation rates for the α-anomers of other hexoses and pentoses are largely their rates of anomerisation. In this connection it is interesting to compare the rate constants of Isbell and Pigman for the bromine oxidations in barium carbonate buffer with their rates for anomerisation in water. The latter may be calculated, for sugars which undergo simple mutarotation, from the principal mutarotation constants [5] and the concentration of the α- and β-forms at equilibrium. When the oxidation rate constants, k_2, are plotted against the anomerisation (β ⟶ α) rate constants, k_1, for the β-anomers, a random scatter of points is obtained (Fig. 2a) as is to be expected if the oxidation and anomerisation reactions are unrelated.

The corresponding plot (Fig. 2b) for the α-anomers gives a fairly good straight line with few deviations. The one marked exception is α-D-lyxose, where the rate of conversion of the α- into the β-form is nearly as great as the rate of oxidation of the β-form, and hence the simple picture of a single rate-determining process no longer applies. It seems probable, therefore, that the α-anomers of all these sugars are oxidised predominantly after rate-determining anomerisation.

[18] Perlmutter-Hayman and Persky, *J. Amer. Chem. Soc.*, 1960, **82**, 276
[19] Nelson and Beegle, *J. Amer. Chem. Soc.*, 1919, **41**, 559.

Isbell and Pigman [4] reported the bromine oxidation of β-D-glucose, as distinct from anomerically equilibrated D-glucose, to be first-order in free bromine and in β-D-glucose, and neither hypobromous acid nor the tribromide ion was a significant oxidant. Oxidation was fast in comparison with concurrent anomerisation of β- into α-D-glucose, so the complication noted for the latter is unimportant in the presence of an excess of bromine. The inverse dependence of rate of oxidation upon acidity was noted by Bunzel and Mathews.[1] The relationship for β-D-glucose was established by Perlmutter-Hayman and Persky,[18,20] although they used equilibrated αβ-D-glucose and calculated rate constants of the oxidation of the β-anomer for the two assumed cases where mutarotation was either very fast or very slow compared with oxidation.

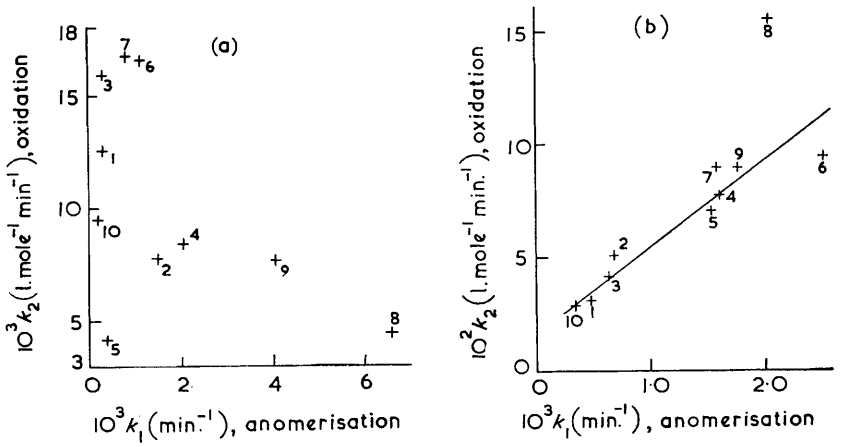


FIG. 2. Variation of oxidation rate constants with anomerisation rate constants for a series of (a) β-anomers and (b) α-anomers, based on measurements of Isbell and his co-workers.[5]

1, Glucose; 2, mannose; 3, galactose; 4, talose; 5, gulose; 6, arabinose; 7, xylose; 8, lyxose; 9, rhamnose; 10, lactose.

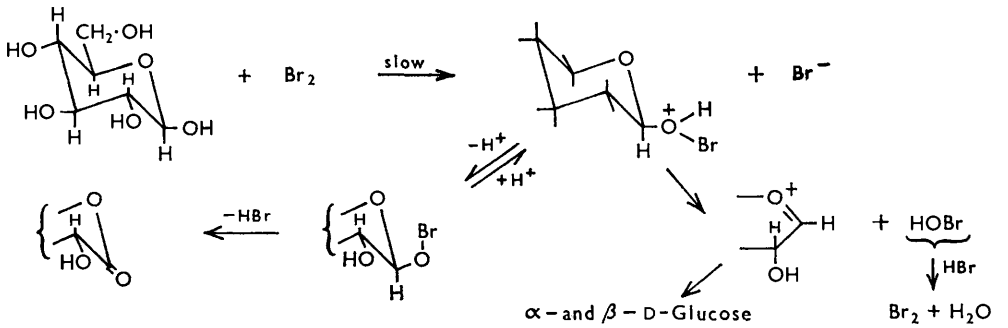
In agreement with other workers, we find that free bromine is the active oxidant and that neither hypobromous acid nor tribromide ions are significant oxidants under the conditions used. In contrast to all other reports, however, we find that the oxidation is not first-order in β-D-glucose. Even in the presence of a large excess of bromine the rate coefficients fall sharply as the reaction proceeds. This trend is not caused by the conversion of free bromine into tribromide ions because, although this does occur, its effect upon the rate is too small, as corresponding results for cyclohexanol show (see following Paper). With this alcohol there was no change in rate coefficients when a ten-fold excess of bromine was used. Nor was the retardation due to the conversion of β- into the much less readily oxidised α-D-glucose, since our results show that β-D-glucose is oxidised about 45 times faster by a ten-fold excess of bromine than it anomerises in the acetate buffer, pH 5.

The value of the rate constant for the oxidation of β-D-glucose at 0° reported by Isbell and Pigman [4] ($k_2 = 4{\cdot}8 \times 10^{-2}$ l. mole^{-1} sec.$^{-1}$) and that calculated from the data of Perlmutter-Hayman and Persky [18] for pH 5 ($k_2 = 2{\cdot}2 \times 10^{-2}$ l. mole^{-1} sec.$^{-1}$) are in broad agreement with our results (Table 6). However, Isbell and Pigman [4] calculate that β-D-glucose is oxidised by bromine about 53 times faster than α-D-glucose, after correction for mutarotation. If we assume that the slower reaction of bromine with α-D-glucose is direct oxidation, we obtain a value of not less than 250 for this β : α rate ratio, in acetate

[20] Perlmutter-Hayman and Persky, *J. Amer. Chem. Soc.*, 1960, **82**, 3809.

buffer, pH 5. Thus, bromine appears to be even more selective towards α- and β-D-glucose than the enzyme glucose oxidase, for which $k_\alpha/k_\beta = 156$.[21]

Mechanism of the Oxidation of β-D-*Glucose.*—The mechanism must explain the much greater reactivity of the β- than the α-anomer, the inverse dependence of the rate upon acid concentration,[18] and the puzzling feature that, whereas the rate is inversely proportional to the acidity, it appears to be inversely proportional to the concentration of bromide ions only to the extent that they convert free bromine into the inactive tribromide ions. The acidity-dependence of the oxidation rate may result from base catalysis by hydroxyl ions, or acidity may control the concentration of a reaction intermediate. Perlmutter-Hayman and Persky [18] discount general-base catalysis as they report that there is no influence of the buffer ion upon the oxidation rate under their conditions. A hypobromite inter-



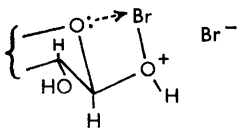
mediate, formed reversibly from glucose and bromine, is unlikely since, in such a reaction, the concentration of hypobromite would depend inversely on the concentration of hydrogen ions and bromide ions.[22,23] However, the conjugate acid of the hypobromite ester may be formed in a slow, irreversible step from β-D-glucose and bromine; this may reversibly lose a proton to form the hypobromite, or it may irreversibly lose hypobromous acid to form the oxonium ion, as shown. The oxonium ion will be rapidly solvolysed to α- and β-D-glucose, with the former, the product of inversion, most probably predominating. The α-anomer reacts so much more slowly than the β-form that it is effectively inert and the reaction rate will fall off; thus, the complexity of diminishing rate coefficients is explained. The free hypobromite, however, may readily undergo elimination of hydrogen bromide to yield the observed product, δ-gluconolactone. This mechanism explains the large difference in reactivity of the α- and β-anomers very satisfactorily, for, not only does the bromine molecule attack the more accessible equatorial hydroxyl group in β-D-glucose, but 1,2-elimination of hydrogen bromide is greatly facilitated since the hypobromite group can adopt the configuration required for *trans*-elimination; with the α-isomer the corresponding configuration, with the hypobromite axial, is very highly strained because of non-bonded interactions between the bromine and the two axial hydrogen atoms at C-3 and C-5. Elimination will be assisted by electrostatic repulsion between the incipient bromide ion and the 2-hydroxyl and ring oxygen atoms.

An alternative explanation for the acidity-dependence of this oxidation has been suggested.[1,18] Perlmutter-Hayman and Persky [18] suggest that, over the pH range 3—7, the oxidation of the anion of glucose predominates. They find an analogy in the bromine oxidation of formic acid, which involves reaction between free bromine and formate anions,[24,25] but the enormous difference in the acidity of formic acid (pK 4) and β-D-glucose

[21] Keilin and Hartree, *Biochem. J.*, 1952, **50**, 331.
[22] Perlmutter-Hayman and Weissmann, *J. Amer. Chem. Soc.*, 1962, **84**, 2323.
[23] Barker, Ph.D. Thesis, London 1962.
[24] Bognar, *Z. phys. Chem.*, 1910, **71**, 529.
[25] Hammick, Hutchinson, and Snell, *J.*, 1925, 2715.

(pK 13) must weaken this analogy; their mechanism would require a rate ratio of about 10^{11} for the oxidation of the anion and the neutral molecule of β-D-glucose. No explanation has yet been offered, on the basis of this mechanism, for the large difference in the rates of oxidation of the α- and β-anomers. Furthermore, if it is the conjugate base of the sugar which is being oxidised, oxidation rates might be expected to increase with acid-strength for a series of sugars. The reverse is found; the pK_a's of the following sugars [26] increase in the order D-mannose < D-glucose < D-galactose < D-arabinose, whereas the oxidation rates [5] diminish in the order β-L-arabinose > β-D-galactose > β-D-glucose > β-D-mannose.

The hypobromite mechanism proposed above also provides a simple explanation for the fact that β-D-glucose and cyclohexanol are oxidised by bromine by different mechanisms. (See following Paper.) Electrophilic attack by bromine on the glycosidic oxygen atom of the former to form the hypobromite conjugate acid could be greatly facilitated by neighbouring-group participation by the ring oxygen atom, forming an electrostatically bonded bromine bridge structure. Only in β-D-glucose is the positively polarised bromine orientated suitably to interact with the ring-oxygen lone-pair electrons. Similar electronic interactions have been considered in other reactions.[27,28]



Oxidation of Anomerically Equilibrated D-*Glucose and of Acetaldehyde.*—Acetaldehyde is oxidised with bromine in dilute aqueous solution to acetic acid and in 76% ethanol–water to ethyl acetate.[29,30] Oxidations in dilute aqueous solution containing methanol yield acetic acid and methyl acetate simultaneously;[30] the ester is not formed from the acid. As a consequence it has been suggested that, in dilute aqueous solution, acetaldehyde is oxidised by bromine in the form of its hydrate.[29–31] Hypobromous acid is not a significant oxidant under these conditions.[32] The kinetics of the hydration of acetaldehyde in aqueous solutions of acids and bases have been reported;[33] the half-lives range from 0·3 to 60 sec. at 25°. The hydration exhibits general-acid and -base catalysis and is said to be very similar in kinetics and mechanism to the mutarotation of glucose,[33] and it was this which made acetaldehyde of interest in the present work.

We find that the bromine oxidation of acetaldehyde in aqueous buffers over the pH range 1—5 is first-order in total bromine, no decrease in rate constant caused by formation of inactive tribromide ions being detected, even though acetaldehyde was in a large excess. We have shown (see following Paper) that under entirely similar conditions the bromine oxidation of cyclohexanol is first-order in free bromine, but not in total bromide ion. The rate of the acetaldehyde oxidation appears to be inversely proportional to acid concentration over the pH range 0—5 but the precise relationship is difficult to ascertain from the present results because of the presence of a negative salt effect (Table 7). Perlmutter-Hayman and Weissmann [22] also find the rate to be inversely proportional to acid concentration but, in contrast, report the absence of a salt effect at high concentrations (0·3—0·7M) of buffer salt and potassium bromide.

The bromine oxidation of acetaldehyde in dilute aqueous solution over the pH range 1—5 is thus unusual in that the rate appears to be unaffected by the conversion of bromine by bromide ions into tribromide ions. This is further indicated by the addition of potassium bromide at pH 1, in which case the rate is not depressed by a factor equivalent to the conversion of bromine into tribromide ions. Therefore, either Br_3^- is an oxidant

[26] Capon and Overend, *Adv. Carbohydrate Chem.*, 1960, **15**, 11.
[27] Edward, *Chem. and Ind.*, 1955, 1102.
[28] Overend, Rees, and Sequeira, *J.*, 1962, 3429.
[29] Bugarszky, *Z. phys. Chem.*, 1904, **48**, 63.
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of similar power to Br_2 for acetaldehyde hydrate, or a number of rates and equilibria are involved in this oxidation, the net result being that the oxidation rate is proportional to the total bromine concentration. Although the rate of hydration of acetaldehyde is much greater than its rate of oxidation, the various possible ionic intermediates in the hydration may be oxidised, together with acetaldehyde hydrate, at a much higher specific rate. This scheme would then be analogous to the halogenation of certain carbonyl compounds. Bell and his co-workers [34,35] have shown, for example, that in the bromination of ethyl malonate both Br_2 and Br_3^- react. At pH greater than 3 most of the bromination takes place through the anion of ethyl malonate, whilst at pH less than 3 the enol is more important. For the anion, Br_3^- has about 0·6 of the activity of Br_2, whilst for the enol the factor is only 0·17. Both Br_2 and Br_3^- react much more rapidly with the anion than with the enol; all rates are very fast.

As noted above, the bromine oxidation of anomerically equilibrated D-glucose in aqueous sulphuric acid of pH 0 is first-order in total bromine; in this it resembles acetaldehyde and is in marked contrast to the results at higher pH. It is possible, therefore, that oxidation of D-glucose at pH 0 may involve similar equilibria and reactions to those of acetaldehyde. It is suggested that, under these conditions, glucose is oxidised largely in the form of the hydrate of the open-chain aldehyde. This explains an apparent difficulty in the results of Perlmutter-Hayman and Persky,[18] who found that the linear relationship between pH and the logarithm of the rate constant for oxidation of β-D-glucose held for the pH range 3—7. In the range 0—3 the rate of oxidation reached a minimum and was only slightly dependent upon pH. These authors explained this by suggesting that, in the pH range 0—3, it is predominantly the neutral molecule which is oxidised, whereas from pH 3 to 7 oxidation of the anion is important. We note that the linear dependence of rate upon acidity changes just where mutarotation becomes faster than oxidation, and propose that when pH less than 3 open-chain and cationic species of D-glucose, present as intermediates in the mutarotation, oxidise more readily than β-D-glucose, for which the rate is strongly retarded by acid. The rate of oxidation of D-glucose is smaller than that for acetaldehyde in aqueous sulphuric acid at 0°, and this is presumably due, at least in part, to the much smaller concentration of aldehyde hydrate in the former than the latter case. Whereas acetaldehyde is hydrated to the extent of about 60% in water at room temperature,[33] the proportion of D-glucose present as the open-chain aldehyde has been shown [36] polarographically to be only 0·0026% at pH 6·9.

The kinetics of the oxidation of anomerically equilibrated D-glucose with chlorine in buffered aqueous solution at pH 2·2 and 3 have been studied.[16] The rate of mutarotation of D-glucose in these buffers was 10—100 times as fast as the consumption of chlorine. The rate data were for the oxidation to D-gluconic acid, although oxidation proceeded beyond this stage. The principal oxidant was molecular chlorine, and the reaction was first-order in this and in D-glucose. The rate of oxidation increased with acidity and with chloride ion concentration. This was interpreted as being due to a shift in the hydrolytic equilibrium of chlorine with added common ion. Lichtin and Saxe [16] state that the oxidations of D-glucose with chlorine and bromine are similar; we consider that the differences in the experimental conditions and the results are such as to invalidate comparison with the bromine oxidations.

Department of Chemistry, Birkbeck College, London W.C.1.
King's College, Strand, London W.C.2. [*Received, November* 1*st*, 1963.]

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