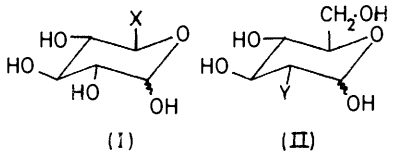
Kinetics and Mechanism of the Mutarotation of Aldoses

By **Brian Capon** * and **Robert B. Walker,** Chemistry Department, Glasgow University, Glasgow G12 8QQ

The kinetics of the mutarotation of xylose, and series of 6-substituted 6-deoxy-glucoses, and a series of 2-substituted 2-deoxy-glucoses catalysed by oxonium ion, water, 4-substituted pyridines, 2,6-lutidine, Tris, 2,2'-iminodiethanol, and morpholine have been measured. Electron-withdrawing substituents at the 6-position decreased the rate of mutarotation catalysed by oxonium ion and by water but increased that of the mutarotation catalysed by bases. Electron-withdrawing substituents at the 2-position decreased the rate of mutarotation with all the catalysts studied except in the case of the 2,6-lutidine-catalysed reaction of 2-amino-2-deoxy-D-glucose hydrochloride, which occurred slightly faster than that of 2-deoxy-D-glucose. The mechanisms of these mutarotations are discussed in the light of these substituent effects, the solvent-isotope effect, and the diffusion-controlled limit of the rate of bimolecular reactions. Steric-hindrance factors were calculated for the 2,6-lutidine-catalysed reactions but there appears to be no correlation between these and the size of the 2- or the 6-substituents.

Intramolecular catalysis was found for the mutarotation of 6-deoxy-D-*gluco*-hepturonic acid and 6-*O*-(*o*-hydroxyphenyl)-D-glucose.

THE rate-limiting step in the mutarotation of glucose is ring opening to give the aldehyde form. This involves removal of a proton from the 1-hydroxy-group, breaking of the ring C(1)–O bond, and transfer of a proton to the ring oxygen atom. The timing of these processes has been discussed frequently but no clear decision on the order in which they occur has been possible.[1] In an attempt to obtain more information we have studied the kinetics of the mutarotation of a series of 6- and 2-substituted glucoses [(I) and (II)].



(I) (II)

Kinetic Analysis.—The simplest way to analyse the

[1] See B. Capon, *Chem. Rev.*, 1969, **69**, 454.

kinetics of the mutarotations studied in this investigation is to regard them as reversible first-order reactions [equation (1)]. The rate of formation of the β-anomer

$$\alpha \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} \beta \qquad (1)$$

is then given by equation (2) and the experimentally

$$d[\beta]/dt = k_1[\alpha] - k_{-1}[\beta] \qquad (2)$$

determined first-order rate constant, k, by equation (3).

$$k = k_1 + k_{-1} \qquad (3)$$

The equilibrium constant, $K = [\beta]_{eq}/[\alpha]_{eq}$, is equal to k_1/k_{-1}, and hence if this is known k can be subdivided into k_1 and k_{-1}. K Values for the 6-substituted sugars (I) are constant within experimental error (Table 1)

TABLE 1

Equilibrium proportions of the sugars studied (±5%)

	α	β
6-Deoxy-D-glucose	31	69
D-Xylose	37 (33) [a]	63 (67) [a]
D-Glucose	37 (36) [a]	63 (64) [a]
6-*O*-Methyl-D-glucose	35	65
6-Acetamido-6-deoxy-D-glucose	32	68
Gentiobiose	31	69
6-*O*-Phenyl-D-glucose	32	68
6-Chloro-6-deoxy-D-glucose	32	67
6-Deoxy-D-*gluco*-hepturonic acid	36	64
6-Cyano-6-deoxy-D-glucose	39	61
2-Deoxy-D-glucose	55	45
2-Amino-2-deoxy-D-glucose HCl	61 (63) [b]	39 (37) [b]
2-Acetamido-2-deoxy-D-glucose	(66) [b]	(34) [b]
2-*O*-Methylglucose	50	50

[a] S. J. Angyal, *Angew. Chem. Internat. Edn.*, 1969, **8**, 157. [b] A. Neuberger and A. P. Fletcher, *Carbohydrate Res.*, 1971, **17**, 79.

and hence any variation in k with structure is proportional to the variations in k_1 and k_{-1}. Our discussion of the results for these compounds is therefore in terms of k or the corresponding catalytic constants. The equilibrium constants, K, for the 2-substituted sugars (II) vary with structure, however, and the values of k have been subdivided into k_1 and k_{-1}.

Actually k_1 and k_{-1} are themselves complex constants since the reaction is more complicated than equation (1) and the *aldehydro*-form is an intermediate which is only ever present at a low concentration as shown in equation (4). If the steady state hypothesis is applied the

$$\alpha \underset{k_{-\alpha}}{\overset{k_\alpha}{\rightleftharpoons}} A \underset{k_\beta}{\overset{k_{-\beta}}{\rightleftharpoons}} \beta \qquad (4)$$

concentration of the *aldehydo*-form is given by equation (5) and the rate of formation of the β-anomer by

$$[A] = (k_\alpha[\alpha] + k_\beta[\beta])/(k_{-\alpha} + k_{-\beta}) \qquad (5)$$

equations (6) and (7), where $p = k_{-\alpha}/k_{-\beta}$, the partitioning

$$\begin{aligned} d[\beta]/dt &= k_{-\beta}[A] - k_\beta[\beta] \\ &= k_{-\beta}\left\{\frac{k_\alpha[\alpha] + k_\beta[\beta]}{k_{-\alpha} + k_{-\beta}}\right\} - k_\beta[\beta] \qquad (6) \\ &= (k_\alpha[\alpha] + k_\beta[\beta])/(1 + p) - k_\beta[\beta] \end{aligned}$$

$$d[\beta]/dt = k_\alpha[\alpha]/(1 + p) - pk_\beta[\beta]/(1 + p) \qquad (7)$$

ratio of the *aldehydo*-form. If equations (2) and (7) are compared at zero time for an experiment in which the mutarotation of a pure α-anomer is being studied, one obtains equation (8). This leads to equation (9), and equation (10) is obtained similarly.

$$(d[\beta]/dt)_0 = k_1[\alpha]_0 = k_\alpha[\alpha]_0/(1 + p) \qquad (8)$$

$$k_1 = k_\alpha/(1 + p) \qquad (9)$$

$$k_{-1} = k_\beta/(1 + 1/p) \qquad (10)$$

Any discussion of the mechanism of mutarotation is necessarily in terms of ring-opening of the cyclic forms. The rate constants for these processes are k_α and k_β. However the only measurable rate constants are k_1 and k_{-1} and any variation of these with structure depends not only on the variation of k_α and k_β but also on the variation of p, which is not known. Thus one possible cause of failure to obtain good correlations when variations of k_1 and k_{-1} or k with structure are studied is that p is not a constant and the *aldehydo*-form of the different 6- and 2-substituted sugars partitions differently between the α- and β-forms.

Oxonium-ion-catalysed Reaction.—The values of $k(H_3O^+)$ of the substituted aldoses (I) increase as X becomes more electron-releasing (Table 2). The plot of log $k(H_3O^+)$ against σ_I [2] for all the compounds except xylose (I; X = H) and gentiobiose yields a ρ value of

TABLE 2

Catalytic constants (l mol^{-1} s^{-1}) for the mutarotation of aldoses (I) catalysed by H_3O^+, D_3O^+, H_2O, and D_2O at 25·0° [a]

X:	Me	H	CH_2·OH	CH_2·OMe	CH_2·NHAc	CH_2·OPh	CH_2Cl	CH_2·CO_2H	CH_2·CN	CH_2·$OC_6H_{11}O_5$ [b]
σ_I [c]	−0·05	0·00	0·05	0·07	0·07	0·11	0·15	0·17	0·18	
$10^3k(H_3O^+)$	18·1	34·6	11·1	8·66	7·72	7·94	6·07	5·16	2·98	9·82
$10^3k(D_3O^+)$	16·8	30·5	9·63	7·38	6·73	6·80	5·30	4·78	2·67	8·81
$k(H_3O^+)/k(D_3O^+)$	1·08	1·14	1·15	1·17	1·15	1·17	1·15	1·08	1·11	1·11
$10^6k(H_2O)$ [d]	16·4	24·0	7·21	6·20	4·53	5·60	4·48	4·10	2·41	6·63
$10^6k(D_2O)$ [e]	4·58	7·20	2·13	1·79	1·36	1·50	1·16	1·11	0·60	1·61
$k(H_2O)/k(D_2O)$	3·58	3·34	3·38	3·46	3·33	3·73	3·86	3·69	4·00	4·11

[a] Rate constants were measured in hydrochloric acid (0·01—0·10M) with ionic strength made up to 0·10M with KCl. [b] Gentiobiose. [c] M. Charton, *J. Org. Chem.*, 1964, **29**, 1222. [d] Intercept of plot of k_{obs} against $[H_3O^+]$ divided by 55·5 mol l^{-1}. [e] Intercept of plot of k_{obs} against $[D_3O^+]$ divided by 55·25 mol l^{-1}.

−2·87 with $r = 0·944$ and a significant level [3] of less than 0·1%. The point for xylose lies 0·396 log units above the correlation line. Similar behaviour is found with all the catalysts studied and must arise from a difference in the steric effect of the group X when it

[2] M. Charton, *J. Org. Chem.*, 1964, **29**, 1222.

[3] See J. Shorter, 'Correlation Analysis in Organic Chemistry,' Clarendon Press, Oxford, 1973, p. 106.

is H and when it is a substituted methylene. It was not possible to use the result for gentiobiose as the σ_I value for the glucosyl residue is unknown.

The ρ value of −2·87 is consistent with a mechanism in which proton transfer to the ring oxygen atom is further advanced than breaking of the ring C(1)–O bond. It does not really shed any light on whether the proton is transferred in a rapid equilibrium step or in a slow step concerted with C–O bond breaking. The isotope effect $k(H_3O^+)/k(D_3O^+)$ varies only slightly with structure and for the compounds studied lies between 1·08 and 1·17 (Table 2). This would seem to be small if the mechanism involved a slow proton transfer to the ring oxygen atom concerted with proton removal from O-1. A mechanism involving a slow proton transfer to the ring oxygen atom without any proton transfer from O-1 seems unlikely, since if this mechanism were

TABLE 3

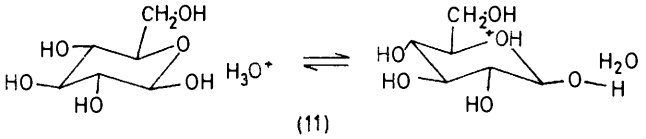
Catalytic constants (l mol^{-1} s^{-1}) for the oxonium ion- and water-catalysed mutarotation of aldoses (II) at 25·0° in hydrochloric acid [a]

Y:	H	OH	OMe	NHAc	NH_3^+
σ_I [b]	0·00	+0·25	+0·28	+0·28	+0·60
$10^3k(H_3O^+)$	>1000	11·1	9·49	6·27	0·36
$10^6k(H_2O)$ [c]	*ca.* 25	7·21	8·97	8·67	8·40

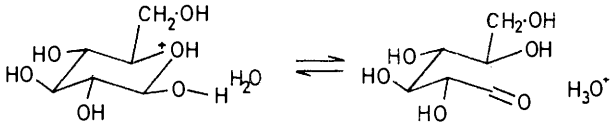
[a] Rate constants were measured in hydrochloric acid (0·01—0·10M) with ionic strength made up to 0·10M with KCl. The values for $k_1 + k_{-1}$ are given. [b] M. Charton, *J. Org. Chem.*, 1964, **29**, 1222. [c] Intercept of k_{obs} against $[H_3O^+]$ divided by 55·5 mol l^{-1}.

followed the methyl glucosides should react just as rapidly, and they do not. The most reasonable mechanism therefore seems to be one which involves a rapid and reversible proton transfer to the ring oxygen atom followed by a slow removal of the proton from O-1 concerted with ring opening as shown in equations (11) and (12). As pointed out by Challis, Long, and Pocker,[4] the first step should lead to an isotope effect $[k(H)/k(D)]$ less than one and the second step to an effect greater than one, and these would tend to cancel each other. This mechanism is also consistent with a consideration of the diffusion-controlled limit as discussed below, with a Bunnett *w*-value of 5·87 at 25° for the mutarotation of 2-acetamido-2-deoxy-D-glucose,[5] and with the effect of solvent composition on the rate in mixtures of water and dimethyl sulphoxide.[6]

The series of 2-substituted glucoses (II) studied was less extensive (Table 3). The most notable results are the high rate of mutarotation of 2-deoxyglucose and the low rate for the conjugate acid of 2-amino-2-deoxyglucose. These results are readily explicable in terms of the mechanism of equations (11) and (12), since the equilibrium constant for formation of the conjugate acid should be increased when the 2-hydroxy-group is replaced by a hydrogen atom and decreased when it is replaced by NH_3^+.



(11)



(12)

Spontaneous or Water-catalysed Reaction.—The variation of $k(H_2O)$ with structure for the sugars (I) is similar to that of $k(H_3O^+)$ (Table 2). The plot of $k(H_2O)$ against σ_I for all the compounds except xylose and gentiobiose yields a ρ value of −2·92 with $r = 0·92$ (significance level [3] <0·1%). The similarity of the ρ value to that for the oxonium-ion catalysed reaction suggests at first sight that the structure of the transition state is similar. Nevertheless, as discussed below, a consideration of the mechanism with reference to the diffusion-controlled limit suggests that there are important differences. Also although replacement of the 2-hydroxy-group by a hydrogen atom again leads to an increase in the rate, replacement by NH_3^+ does not result in a rate decrease as found with the oxonium-ion catalysed reaction. This is a further indication that the transition state for the water-catalysed reaction differs from that of the oxonium-ion-catalysed one (see also ref. 5 for a discussion of this point).

TABLE 4

Catalytic constants [10^2 × *slope* (i) in l mol^{-1} s^{-1}] for the Tris-catalysed mutarotation of aldoses (I) at 25·0° (*I* 0·100M)

X:	Me	H	CH_2·OH	CH_2·OMe	CH_2·NHAc	CH_2·OPh	CH_2Cl	CH_2·CN	CH_2·$OC_6H_{11}O_5$ [a]
pH									
8·10	1·07	8·01	1·43	1·56		3·11	4·85	7·92	3·26
8·33	1·07	8·29	1·48	1·59	1·60	3·35	5·02	8·56	3·39
8·56	1·10	8·04	1·44	1·59	1·72	3·38	5·55	9·39	3·47
8·87	1·12	8·56	1·48	1·57	1·75	3·50	5·30	12·31	3·54
9·16	1·14	8·69	1·69	1·62	1·84	3·50	6·05		3·33
Mean [b]	1·08	8·11	1·45	1·58	1·66	3·28	5·14	7·92 [c]	3·37

[a] Gentiobiose. [b] Mean of the values at pH 8·10, 8·33, and 8·56. [c] Value at pH 8·10.

Catalysis by Tris and Hydroxide Ion.—The catalytic coefficients for Tris and hydroxide ion were obtained from the values of k_{obs} in Tris buffers at five different pH values (Table 4). Under these conditions the following equation applies, where G^- represents the

[4] B. C. Challis, F. A. Long, and Y. Pocker, *J. Chem. Soc.*, 1957, 4679.

[5] D. M. L. Morgan and A. Neuberger, *Proc. Roy. Soc.*, *A*, 1974, **337**, 317.

[6] N. M. Ballash and E. B. Robertson, *Canad. J. Chem.*, 1973, **51**, 556.

glucose anion, which is also catalytically active, and $TrisH^+$ is the conjugate acid of Tris:

$$k_{obs} = k(H_2O)[H_2O] + k_{Tris}[Tris] + k_{TrisH}[TrisH^+] + k(HO^-)[HO^-] + k_G[G^-]$$

Under the conditions used, pH $\leqslant$9·16, $[G^-]$ is a small fraction of the total concentration of glucose and hence can be written:

$$[G^-] = [GH]a_{OH}K_G f_{GH}/K_w f_G$$

and also

$$[TrisH^+] = [Tris]a_H f_{Tris}/(f_{TrisH} \times K_{Tris}).$$

Therefore

$$k_{obs} = k(H_2O)[H_2O] + k_{Tris}[Tris] + k_{TrisH}[Tris]a_H f_{Tris}/f_{TrisH} + k(HO^-)[HO^-] + k_G[GH]a_{OH}K_G f_{GH}/K_w f_G.$$

Therefore the plot of k_{obs} against [Tris], the concentration of free Tris, at constant pH yields

$$\text{slope (i)} = k_{Tris} + k_{TrisH}a_H f_{Tris}/(f_{TrisH} \times K_{Tris})$$

$$\text{intercept (i)} = k(H_2O)[H_2O] + k(HO^-)[HO^-] + k_G[GH]a_{OH}K_G f_{GH}/K_w f_G.$$

The plot of intercept (i) against a_{OH} from the results for five different buffer ratios yield a straight line with

$$\text{slope (ii)} = k(HO^-) + k_G[GH]K_G f_{GH}/K_w f_G$$

$$\text{intercept (ii)} = k(H_2O)[H_2O].$$

The importance of the term $k_G[GH]a_{OH}f_{GH}/K_w f_G$ to intercept (i) was evaluated for glucose by making use of the results of Smith [7] and of Kilde and Wynne-Jones,[8] who showed that k_G is about $k(HO^-)/200$. The concentration of glucose used in our work is 0·02 mol l^{-1}; at 25° $K_G = 10^{-14}$ and $K_G = 4{\cdot}6 \times 10^{-13}$. Therefore

$$k(HO^-)/(k_G[GH]K_G/K_w) = 1/4{\cdot}6 \times 10^{-3}.$$

The contribution of catalysis by glucosate ion to slope (ii) is therefore less than 0·5% unless the activity coefficient ratio, f_{GH}/f_G, is very small and slope (ii) was therefore taken to be equal to $k(HO^-)$ (Table 5). The reason that catalysis by glucose anion is negligible in our work is that the concentration of glucose (0·02 mol l^{-1}) is much less than that used by Smith [7] and by Kilde and Wynne-Jones.[8]

The value of slope (i) should increase with increasing a_H (*i.e.* decreasing pH) if there were a significant contribution from catalysis by the conjugate acid of Tris. In fact there is a slight trend in the opposite sense (Table 4) but we are not sure of the reason for this. It is most noticeable with 6-chloro-6-deoxy- and 6-cyano-6-deoxy-glucose. To minimise the effect of this the catalytic coefficient has been taken as the average of slope (i) at the three lowest pH values. As the increase was most significant with 6-cyano-6-deoxy-glucose it was thought possible that the change in rotation in the more alkaline buffers might arise from hydrolysis of the nitrile group as well as anomerisation. Therefore 6-cyano-6-deoxyglucose was heated at 60° in the most alkaline buffer studied (pH 9·16) for 3 h. At the end of this time t.l.c. showed one spot with R_F value identical with that of the starting material, and the i.r. spectrum showed an unchanged C≡N stretching absorption but no carbonyl absorption, which would result if the nitrile group were hydrolysed.

The values of both k_{Tris} and $k(HO^-)$ increase as the electron withdrawing power of X increases (Table 4) but the correlations with σ_I are not as good as those found for catalysis by H_2O and H_3O^+. The ρ values are +3·97 and +6·22, respectively, with $r = 0{\cdot}924$ and 0·848 and significance levels [3] lying between 1 and 0·1% and 5 and 1%, respectively. Possibly the methods used to extract these constants have led to an accumulation of errors, or possibly variations of f with structure are important as discussed above.

It is interesting that the effect of the substituents X of (I) on the rate of mutarotation are in opposite senses for the water- and hydroxide-ion-catalysed reactions, since it has frequently been suggested [9] that water acts as a base catalyst. In fact the effect of substituents on the rate of the water-catalysed mutarotation is in the opposite sense to that found with all the base catalysts studied in this investigation. This suggests that there is an important difference between water catalysis and base catalysis.

Catalysis by Pyridines.—Generally the rate of the pyridine-catalysed mutarotation of aldoses (I) increases as the electron-withdrawing power of the substituent X increases (Tables 6 and 7). However when log k_{cat} is plotted against σ_I for all the sugars except xylose and gentiobiose the correlations are poor (significance levels >5%). Throughout, the catalytic constants for 6-*O*-phenylglucose appear to be 2—3 times greater than expected from those for the other 6-substituted glucoses. Possibly there is a weak hydrophobic or charge-transfer

TABLE 5

Catalytic constants (l mol^{-1} s^{-1}) for the hydroxide ion- and water-catalysed mutarotation of aldoses (I) at 25·0° (*I* 0·100M)

X:	Me	H	CH_2·OH	CH_2·OMe	CH_2·OPh	CH_2Cl	CH_2·CN	CH_2·$OC_6H_{11}O_5$ [a]
$10^2k(HO^-)$ [b]	0·33	4·61	0·80	0·50	0·84	3·21	12·2	1·96
$10^6k(H_2O)$ [c]	16·0	26·7	7·19	6·01	5·27	5·18	8·33 [d]	6·80

[a] Gentiobiose. [b] Slope (ii). [c] Intercept (ii)/55·5 mol l^{-1}. [d] This value is not very accurate and k_{H_3O} only makes a small contribution to the rate in alkaline buffers. The values given in Tables 2 or 6 are more accurate.

[7] G. F. Smith, *J. Chem. Soc.*, 1936, 1824; G. F. Smith and M. C. Smith, *ibid.*, 1937, 1413.

[8] G. Kilde and W. F. K. Wynne-Jones, *Trans. Faraday Soc.*, 1953, **49**, 243.

[9] *Cf.* H. S. Isbell and W. Pigman, *Adv. Carbohydrate Chem.*, 1969, **24**, 24.

interaction between the pyridines and 6-*O*-phenylglucose. A similar explanation has been proposed to explain the enhanced catalytic effect of pivalate in the hydrolysis of vinyl ethers [10] and of aromatic amines in the hydrolysis of *p*-nitrophenyl acetate.[11] The rate constant for the mutarotation of 6-*O*-phenylglucose catalysed by pyridine increases slightly with decreasing pH. This variation is not large enough to account for the anomaly mentioned above and may be caused by catalysis by pyridinium ion. The catalytic constants for all the other sugars studied are independent of pH within experimental error.

When the results for 6-*O*-phenylglucose were omitted much better correlations with σ_I were obtained (significance level <1%) with $\rho = 2{\cdot}4$, 2·1, and 1·2 for catalysis by 4-ethoxypyridine, 4-methylpyridine, and pyridine, respectively. The Brønsted β values obtained from the results with these three bases increase with electron-withdrawing power of the substituent at position 6. If, as discussed below, the mechanism of equation (22) is correct then the α value (= 1 − β) for the variation of k_S with structure becomes greater as X becomes more electron-releasing. Therefore if the α value can be taken as a measure of the degree of proton transfer in the transition state,[12] the less easily the ring C(1)–O bond breaks (assuming electron-withdrawing groups in X facilitate this) the greater the degree of proton transfer in the transition state, which is reasonable. The β values for the mutarotation of 2-substituted aldoses (II) are almost constant. Again 2-deoxyglucose mutarotates faster than the other compounds but the β value is unchanged (Table 8).

It has been shown that the 2,6-lutidine-catalysed mutarotation of glucose and tetramethylglucose is subject to steric hindrance since 2,6-lutidine is a poorer

TABLE 6

Catalytic constants (l mol^{-1} s^{-1}) for the pyridine- and water-catalysed mutarotation of aldoses (I) at 25·0° (*I* 0·100M)

X:	Me	H	CH_2·OH	CH_2·OMe	CH_2·NHAc	CH_2·OPh	CH_2Cl	CH_2·CN	CH_2·$OC_6H_{11}O_5$
					10^3k_{pyr}				
pH									
5·93	5·84	14·88		6·78	7·21		10·87	10·00	10·22
5·72	5·50	15·04	5·91	7·48		15·23	10·53	9·84	9·72
5·35	5·73	15·95	6·00	7·84		16·68	10·27	9·48	10·21
5·00	6·07	15·15	6·02	7·35		17·85	10·14	9·12	9·75
Average	5·79	15·27	5·98	7·36	7·21	16·59	10·44	9·60	9·99
					$10^6k(H_2O)$				
5·93	15·77	24·09		6·31	4·38		4·59	2·83	6·94
5·72	16·14	24·11	7·46	6·16		6·27	4·63	2·53	7·22
5·35	15·84	23·39	7·42	6·23		5·28	4·72	2·63	6·81
5·00	15·84	24·42	7·41	6·42		5·78	4·64	2·50	6·55
Average	15·90	23·75	7·33	6·31	4·38	5·77	4·65	2·65	6·88

TABLE 7

Catalytic constants (l mol^{-1} s^{-1}) for the mutarotation of aldoses (I) catalysed by unsubstituted pyridines at 25·0° (*I* 0·100M)

	pK_a	X:	Me	H	CH_2·OH	CH_2·OMe	CH_2·NHAc	CH_2·OPh	CH_2Cl	CH_2·CN	CH_2·$OC_6H_{11}O_5$
4-Ethoxy	6·67	10^2k_{cat}	1·93	5·33	2·20	2·67	3·59	7·94	5·55	6·35	3·93
4-Methyl	5·98	10^2k_{cat}	1·24	3·43	1·42	1·80	2·09	5·17	3·24	3·32	2·59
Unsubst.	5·22	10^3k_{cat}	5·79	15·27	5·98	7·36	7·21	16·59	10·44	9·60	9·99
		β	0·36	0·37	0·39	0·39	0·48	0·47	0·50	0·57	0·41

TABLE 8

Catalytic constants (l mol^{-1} s^{-1}) for the mutarotation of aldoses (II) catalysed by substituted pyridines at 25·0° (*I* 0·100M) [a]

	pK_a	Y:	H	OH	OMe	NHAc	NH_3^+
4-Ethoxy	6·67	10^2k_{cat}	14·0 (6·28)	2·20 (1·41)	3·58 (1·79)	1·79 (0·57)	1·94 (0·72)
4-Methyl	5·98	10^2k_{cat}	8·84 (3·98)	1·42 (0·91)	2·17 (1·09)	1·27 (0·41)	1·37 (0·51)
Unsubst.	5·22	10^3k_{cat}	32·2 (15·0)	5·98 (3·83)	8·67 (4·34)	4·88 (1·56)	5·30 (1·96)
		β	0·43	0·39	0·43	0·39	0·39

[a] The values outside the parentheses are $k_1 + k_{-1}$ and the values inside are k_1, the rate constants for the conversion of the α- into the β-anomers.

TABLE 9

Catalytic constants and steric hindrance factors (S.H.F.) for the mutarotation of aldoses (I) catalysed by 2,6-lutidine at 25·0° (*I* 0·100M)

X:	Me	H	CH_2·OH	CH_2·OMe	CH_2·NHAc	CH_2·OPh	CH_2Cl	CH_2·CN	CH_2·$OC_6H_{11}O_5$
$10k_{cat}$ (obs.)	2·32	7·40	1·80	2·36	5·27	12·55	5·65	9·66	2·81
10^3k_{cat} (calc.)	21·1	58·7	24·5	30·1	41·0	93·6	64·3	74·0	44·7
S.H.F.	9·1	7·9	13·6	12·8	7·8	7·5	11·4	7·7	15·9

[10] A. J. Kresge, H. L. Chen, Y. Chiang, E. Murrill, M. A. Payne, and D. S. Sagatys, *J. Amer. Chem. Soc.*, 1971, **93**, 413.

[11] F. Schneider and H. Wenck, *Z. physiol. Chem.*, 1967, **348**, 122.

[12] *Cf.* A. J. Kresge, *Chem. Soc. Rev.*, 1973, **2**, 475.

catalyst than predicted from the Brønsted relationship for catalysis by 4-substituted pyridines and the pK_a of 2,6-lutidine.[13,14] Steric hindrance factors (S.H.F.), defined as the ratio of the experimental catalytic constant for the 2,6-lutidine-catalysed mutarotation to that calculated from the Brønsted relationship, obtained in this investigation are given in Tables 9 and 10. The largest S.H.F. is found with gentiobiose, which probably has the largest substituent, but this is only slightly larger than that found with 6-*O*-methylglucose. If the steric effect results from a direct interaction between the methyl groups on the lutidine and the sugar the transition state must therefore be one in which the substituents at C-2 and C-5 are not directly involved.

TABLE 10

Catalytic constants and steric hindrance factors (S.H.F.) for the mutarotation of aldoses (II) catalysed by 2,6-lutidine at 25·0° (*I* 0·100M) [a]

Y:	H	OH	OMe	NHAc	NH_3^+
10^3k_{cat} (obs.)	5·65 (2·54)	1·80 (1·13)	2·94 (1·47)	1·34 (0·46)	6·47 (2·52)
10^3k_{cat} (calc.)	(71·4)	(15·7)	(20·0)	(6·59)	(8·29)
S.H.F.	28·1	13·9	13·0	14·3	3·29

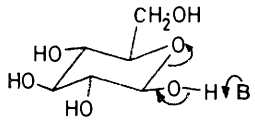
[a] The values outside the parentheses are $k_1 + k_{-1}$ and the values inside are k_1, the rate constants for the conversion of the α- into the β-anomers.

TABLE 11

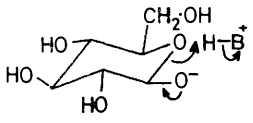
Catalytic constants (l mol^{-1} s^{-1}) for the morpholine- and 2,2′-iminodiethanol-catalysed mutarotation of aldoses (I) at 25·0° (*I* 0·100M)

X:	Me	H	CH_2·OH	CH_2·OMe	CH_2·NHAc	CH_2·OPh	CH_2Cl	CH_2·CN	CH_2·$OC_6H_{11}O_5$
10^2k(morpholine)	9·27	46·8	11·0	13·3	9·59	34·2	42·9	51·4	23·7
10^2k(2,2′-iminodiethanol)	3·76	26·2	6·07	7·40	7·19	16·02	22·8	34·7	11·8

variation of the S.H.F. values with structure is fairly small and appears to bear no relationship to the size of the substituents. The slowness of the 2,6-lutidine-catalysed mutarotation implies that there is an unfavourable steric interaction in the transition state for mutarotation which is absent or is smaller in the equilibrium transfer of a proton to 2,6-lutidine. This could arise from non-bonding interactions between the methyl groups of the lutidine and the sugar or from a steric interaction with the solvent that is present when the proton is partly transferred to or from the 2,6-lutidine but is not present when proton transfer is complete. Two types of transition state for the 2,6-lutidine-catalysed mutarotation of aldoses are possible, (III) and (IV), depending on whether the lutidine acts by abstracting a proton from the 1-hydroxy-group or by its

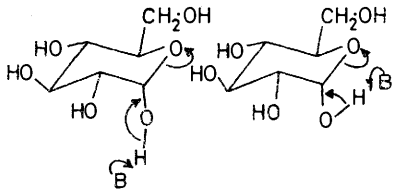


(III)



(IV)

conjugate acid donating a proton to the ring oxygen atom. Whichever of these is correct it seems unlikely that there is a steric interaction between the lutidine and the substituent at C-2 or C-5 since the variation in steric hindrance factors is small and does not appear to correlate with the size of these substituents. Thus with 2-substituted aldoses (II) the largest S.H.F. is found with 2-deoxyglucose (Y = H). With the aldoses (I) *anti*- (V), *syn*- (VI), and perpendicular (VII) stereochemistries are possible if the lutidine acts by abstracting



(V) (VI) (VII)

a proton from the 1-hydroxy-group. In the transition state with *anti*-stereochemistry (V) there would be an unfavourable steric interaction between the lutidine and the substituent at C-2. The results in Table 10 therefore exclude this possibility. As discussed by Kerschner and Schowen [15] a perpendicular stereochemistry is most likely in elimination reactions which involve proton transfer from oxygen as this allows the oxygen lone pair to interact with the developing double bond. However as discussed below a transition state of type (IV) seems more likely for base-catalysed mutarotation.

Steric hindrance is also found in the 2,2′-iminodiethanol-catalysed mutarotation of aldoses (I), which is slower than the morpholine-catalysed mutarotation (Table 11) despite iminodiethanol being the stronger base [pK_a(iminodiethanol) = 9·18, pK_a(morpholine) = 8·6].

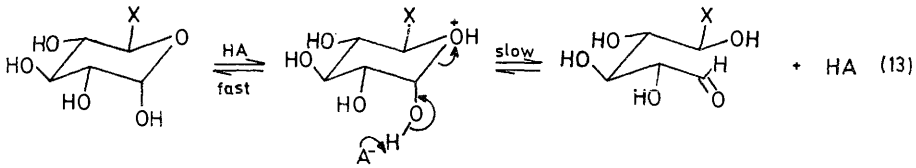
Diffusion-controlled Limit and Mechanism of Mutarotation.—A way which is sometimes used to test if a mechanism is a valid one is to calculate whether a thermodynamically unstable intermediate would have

[13] F. Covitz and F. H. Westheimer, *J. Amer. Chem. Soc.*, 1963, **85**, 1773.

[14] H. H. Huang, A. N. H. Yeo, and L. H. L. Chia, *J. Chem. Soc.* (*B*), 1969, 836.

[15] L. D. Kershner and R. L. Schowen, *J. Amer. Chem. Soc.*, 1971, **93**, 2014; R. L. Schowen, *Progr. Phys. Org. Chem.*, 1971, **9**, 275.

to react with another reactant at a rate greater than the diffusion-controlled limit. Eigen showed that a mechanism analogous to (13) for the water-catalysed dehydration of acetaldehyde hydrate would require the rate-determining step to be faster than the rate of diffusion.[16] If a



similar treatment is applied to the mutarotation reaction, the rate is given by equation (14) where S is the aldose, SH^+ its conjugate acid, and K_{SH} its ionisation constant.

$$\text{Rate} = \frac{k_S[S][H^+]}{K_{SH}} \times K_{HA}\frac{[HA]}{[H^+]}$$

$$= k_S K_{HA}[S][HA]/K_{SH} \quad (14)$$

The experimental rate constant for acid catalysis, k_{HA}, is related to the rate constant for the rate-limiting step, k_S, by equations (15) and (16). The value to be given

$$k_{HA} = k_S K_{HA}/K_{SH} \quad (15)$$

$$k_S = k_{HA} K_{SH}/K_{HA} \quad (16)$$

to K_{SH} is uncertain but the ring oxygen atom of a sugar should be less basic than the oxygen atom of diethyl ether for which $pK_a = -3{\cdot}6$.[17] Therefore K_{SH} is probably in the range 10^4—10^6 and k_S can now be evaluated for certain catalysts. For α-D-glucose at 25° $k(H_3O^+) = 6{\cdot}97 \times 10^{-3}$ l mol^{-1} s^{-1}. Therefore

$$k_S = 6{\cdot}97 \times 10^{-3} \times K_{SH}/55{\cdot}5$$

$$= 1{\cdot}26 \times 10^{-4} \times K_{SH} \text{ l mol}^{-1} \text{ s}^{-1},$$

and the rate constant for the rate-limiting step should be well below the diffusion-controlled limit. For catalysis by acetic acid,

$$k_{AcOH} = 7{\cdot}75 \times 10^{-4} \text{ l mol}^{-1} \text{ s}^{-1} \text{ at } 25°,^{18}$$

and

$$k_S = 7{\cdot}75 \times 10^{-4} \times K_{SH}/1{\cdot}7 \times 10^{-5}$$

$$= 4{\cdot}56 \times K_{SH} \text{ l mol}^{-1} \text{ s}^{-1}$$

Again k_S should be well below the diffusion-controlled limit for the expected range of values of K_{SH}. For catalysis by water

$$k(H_2O) = 4{\cdot}56 \times 10^{-6} \text{ l mol}^{-1} \text{ s}^{-1}$$

and

$$k_S = 4{\cdot}54 \times 10^{-6} \times K_{SH}/10^{-14}$$

$$= 4{\cdot}54 \times 10^{8} \times K_{SH} \text{ l mol}^{-1} \text{ s}^{-1}$$

Now k_S would be above the rate constant for diffusion. Hence equation (13) appears to be a valid mechanism for the oxonium-ion- and acetic-acid-catalysed mutarotation but not for the water-catalysed reaction.

Jencks has shown that the mechanism analogous to that of equation (17) for the dehydration of acetaldehyde hydrate is invalid by calculating that the reverse reaction of the rate-determining step (r.d.s.) must be faster than the diffusion-controlled limit.[19] The calculation for the mutarotation reaction is as follows:

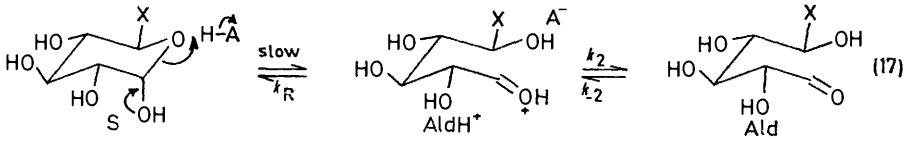
Rate of reverse of r.d.s.

$$= k_R[\text{AldH}^+][A^-]$$

$$= k_R[\text{Ald}][H^+]K_{HA}[HA]/K_{AldH}[H^+]$$

$$= k_R K_{HA}[\text{Ald}][HA]/K_{AldH}$$

where K_{AldH} is the dissociation constant of the conjugate acid of the acyclic aldehyde. The concentration of the acyclic aldehyde form of glucose is not known definitely



but one estimate is 0·0002%,[20] so that [Ald]/[S] = 2×10^{-5} and

Rate of reverse of r.d.s.

$$= k_R K_{HA}[S][HA] \times 2 \times 10^{-5}/K_{AldH}$$

The experimental rate constant for acid catalysis, k_{HA}, is not simply related to the rate constant for the reverse of the rate-determining step, k_R. If the steady-state hypothesis is applied to $AldH^+$ in equation (17) then equation (18) is obtained, which leads to equation (19).

$$k_{HA}[S][HA] + k_{-2}[\text{Ald}][HA] = k_R K_{HA} \times 2 \times 10^{-5} \times [S][HA]/K_{AldH} + k_2[\text{AldH}^+][A^-] \quad (18)$$

$$k_R = 5 \times 10^4 k_{HA} K_{AldH}/K_{HA} + k_{-2} K_{AldH}/K_{HA} - k_2 \quad (19)$$

The dissociation constant for the conjugate acid of the *aldehydo*-form of glucose is not known but it should be a stronger acid (*i.e.* the aldehyde group should be less basic) than that of acetaldehyde for which the pK_a is −10·2. By using a value of 10^{11} for K_{AldH} the first term on the right hand side of equation (19) can be

[16] M. Eigen, *Discuss. Faraday Soc.*, 1965, **39**, 7.
[17] E. M. Arnett, *Progr. Phys. Org. Chem.*, 1963, **1**, 223.
[18] H. Schmid and G. Bauer, *Monatsh.*, 1966, **97**, 168.
[19] W. P. Jencks, 'Catalysis in Chemistry and Enzymology,' McGraw-Hill, New York, 1969, pp. 211—217.
[20] J. M. Los, L. B. Simpson, and K. Wiesner, *J. Amer. Chem. Soc.*, 1956, **78**, 1564.

evaluated for various catalysts. For α-D-glucose, $k(H_3O^+) = 6{\cdot}97 \times 10^{-3}$ l mol^{-1} s^{-1}, and

$$k_R = \frac{5 \times 10^4 \times 6{\cdot}97 \times 10^{-3} \times 10^{11}}{55{\cdot}5} + k_{-2} \times 10^{10}/55{\cdot}5 - k_2$$
$$= 6{\cdot}28 \times 10^{11} + 1{\cdot}8 \times 10^8 \times k_{-2} - k_2 \text{ l mol}^{-1} \text{ s}^{-1}.$$

The value of k_2 must be close to the diffusion-controlled limit (10^9—10^{10} l mol^{-1} s^{-1}) but k_{-2} must be much smaller. Therefore although we cannot evaluate k_{-2} its value will not affect the conclusion that for the oxonium-ion-catalysed mutarotation of glucose equation (17) requires that k_R takes a value of at least 10^{11}—10^{12} l mol^{-1} s^{-1}. Similar calculations for catalysis by acetic acid and by water using the same values of the catalytic coefficients as were used before lead to expressions for k_R:

For acetic acid catalysis

$$k_R = 2{\cdot}3 \times 10^{15} - k_2 + 6 \times 10^{14} \times k_{-2} \text{ l mol}^{-1} \text{ s}^{-1}$$

For water catalysis

$$k_R = 2{\cdot}3 \times 10^{23} - k_2 + 10^{24} k_{-2} \text{ l mol}^{-1} \text{ s}^{-1}$$

Clearly k_R will be much greater than the diffusion-controlled limit for all reasonable values of k_{-2}. Therefore Jencks' objection to the mechanism of equation (17) is valid not only for water catalysis but also for catalysis by acetic acid and by oxonium ion. Therefore the mechanism of equation (13) is the preferred one for acid catalysis but it is not possible for the spontaneous or water-catalysed reaction. This mechanism is analogous to that preferred by Bell and his co-workers for the dehydration of aldehyde hydrates.[21,22]

The same treatment can be applied to catalysis by bases. One possible mechanism is that shown in equation (20), which leads to equation (21) for k_R, the

$$k_R = 5 \times 10^4 k_B k_{BH^+}/K_{Ald} + k_{-2} \times K_{BH^+}/K_{Ald} - k_2 \quad (21)$$

rate constant for the reverse of the rate-determining step where K_{Ald} is the dissociation constant for the 5-hydroxy-group of the *aldehydo*-form (*ca.* 10^{-13} mol l^{-1}) and K_{BH^+} is the dissociation constant for the conjugate acid of the catalyst. If the values of k_B for the mutarotation of α-D-glucose at 25° are substituted into equation (21) the following expressions for k_R are obtained:

For pyridine catalysis

$$k_R = 1{\cdot}14 \times 10^{10} + 6{\cdot}03 \times 10^7 \times k_{-2} - k_2 \text{ l mol}^{-1} \text{ s}^{-1}$$

For morpholine catalysis

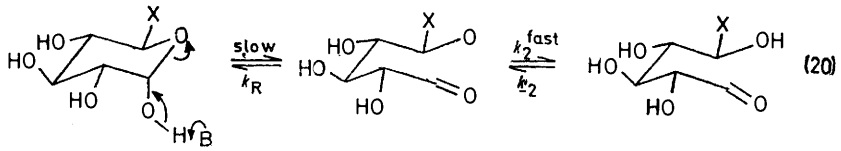
$$k_R = 1{\cdot}63 \times 10^8 + 4{\cdot}7 \times 10^4 \times k_{-2} - k_2 \text{ l mol}^{-1} \text{ s}^{-1}$$

Since k_2 is approximately 10^{10} l mol^{-1} s^{-1}, k_R will be close to or over the diffusion-controlled limit for catalysis by pyridine but for stronger bases such as morpholine the value of k_R is dependent on the value of k_2. Since k_R is greater than the diffusion-controlled limit for some bases the mechanism of equation (20) is probably incorrect.

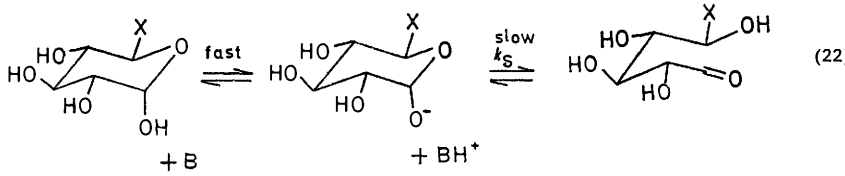
The mechanism given in equation (22) is also possible. Now $k_S = k_B K_{BH^+}/K_S$, where K_S is the dissociation constant of the aldose (for α-D-glucose, $4{\cdot}6 \times 10^{-13}$ at 25° [8]) and K_{BH^+} is the dissociation constant of the conjugate acid of the catalyst. For α-D-glucose at 25°,

$k_S = 4{\cdot}9 \times 10^4$ l mol^{-1} s^{-1} for catalysis by pyridine

$k_S = 7{\cdot}1 \times 10^2$ l mol^{-1} s^{-1} for catalysis by morpholine.



These values are well within the diffusion-controlled limit. We therefore favour the mechanism of equation (22) for base catalysis. Again this is analogous to the mechanism favoured by Bell and his co-workers [21,22] and by Bender [23] for the dehydration of aldehyde hydrates and also to that proposed by Schaleger and his co-workers for the decomposition of hemiacetals.[24] In contrast Pocker and Dickerson have concluded that this mechanism is unlikely for the hydration of aldehydes and the dehydration of their hydrates.[25] The rate constant k^* (analogous to k_S) would take a reasonable value for catalysis by most bases but not for catalysis by water, and since the point for water fitted on the same Brønsted plot as those for other bases it was concluded that this mechanism could not be correct. It does appear however (ref. 25, footnote 28) that water is eight times as effective a catalyst as would be expected



[21] R. P. Bell and W. C. E. Higginson, *Proc. Roy. Soc.*, 1949, *A*, **197**, 141.
[22] R. P. Bell and B. de B. Darwent, *Trans. Faraday Soc.*, 1950, **46**, 34.
[23] M. L. Bender, ' Mechanisms of Homogeneous Catalysis from Protons to Proteins,' Wiley-Interscience, New York, 1971, p. 130.
[24] A. L. Mori, M. A. Porzio, and L. L. Schaleger, *J. Amer. Chem. Soc.*, 1972, **94**, 5034.
[25] Y. Pocker and D. G. Dickerson, *J. Phys. Chem.*, 1969, **73**, 4005.

from the Brønsted plot for monofunctional bases and it is possible that the mechanisms of catalysis by mono- and bi-functional bases differ. Also the difference in the substituent effects between the water- and base-catalysed mutarotation of the 6-substituted aldose (I) makes it unlikely that these reactions follow the same mechanism.

The kinetic results for the water-catalysed mutarotation are inconsistent with all the stepwise mechanisms considered and therefore a concerted mechanism seems most reasonable, but in view of the large positive ρ-value for the reactions of compounds (I) this must be one in which the ring oxygen atom carries substantial positive charge in the transition state. A concerted mechanism for the water-catalysed reaction is also supported by the effect of solvent composition on the rate in mixtures of water with organic solvents.[6,26] It is also interesting that the rate of the spontaneous hydration of substituted trifluoroacetophenones in water–tetramethylene sulphone mixtures has a much greater dependence on water content than does the acid-catalysed reaction.[27]

Intramolecular Catalysis.[28]—Bailey, Fishman, and Pentchev have reported that the mutarotation of D-glucose 6-phosphate is 240 times faster than that of D-glucose, and attributed this rate enhancement to intramolecular catalysis.[29] To test whether other 6-substituted glucoses behaved similarly, 6-deoxy-D-*gluco*-hepturonic acid (VIII) and 6-*O*-(*o*-hydroxyphenyl)-D-glucose (IX) were synthesised and their mutarotations studied. The pH–rate profile for the spontaneous mutarotation of 6-deoxy-D-*gluco*-hepturonic acid (VIII) after extrapolation to zero buffer concentration is sigmoid, with $k_0 = (k_{SH} \times 10^{-pH}/K_a + k_S)/(1 + 10^{-pH}/K_a)$ (Table 12); k_{SH} and k_S, the rate constants for the

TABLE 12

pH–Rate profile for the spontaneous mutarotation of 6-deoxy-α-D-*gluco*-hepturonic acid at 25·0° (*I* 1·0M)

pH	Buffer	10^4k_0/s^{-1}	10^3k_{calc}/s^{-1} [a]
2·00	HCl	2·28 [b]	2·69
2·75	Formate	3·98	3·66
3·13	Formate	5·80	5·20
3·85	Acetate	13·8	13·7
4·27	Acetate	23·6	23·1
4·61	Acetate	30·9	31·1
4·91	Acetate	35·2	36·7
5·30	Acetate	39·9	41·1
5·92	Pyridine	44·3	43·9

[a] Calculated from the expression $k_{calc} = (k_{SH} \times 10^{-pH}/K_a + k_S)/(1 + 10^{-pH}/K_a)$ with $k_{SH} = 2{\cdot}48 \times 10^{-4}$ s^{-1}, $k_S = 4{\cdot}43 \times 10^{-3}$ s^{-1}, and $K_a = 5{\cdot}08 \times 10^{-5}$ mol l^{-1}. Experimental K_a is $4{\cdot}0 \times 10^{-5}$ mol l^{-1}. [b] k_{obs}.

mutarotation of the un-ionised and ionised forms respectively, have values $2{\cdot}48 \times 10^{-4}$ and $4{\cdot}49 \times 10^{-3}$ s^{-1} at 25°, and K_a, the apparent dissociation constant, is $5{\cdot}08 \times 10^{-5}$ mol l^{-1}. The value of k_S is about 8·3 times greater than that estimated from the linear free energy plot of the rate constants for the spontaneous mutarotation of the 6-substituted aldoses (I) given in Table 2. This rate enhancement may arise from intramolecular catalysis. The second-order constant for the mutarotation of D-glucose catalysed by a carboxylate ion of pK_a 4·3 was estimated from the results of Schmid and Bauer [30] for the formate- and acetate-catalysed reactions to be $1{\cdot}2 \times 10^{-3}$ l mol^{-1} s^{-1} at 25°. The effective concentration [31] of the internal carboxylate group in the mutarotation of the anion, if this reaction is intramolecularly catalysed, is therefore 3·5 mol l^{-1}. The analogous factor reported by Bailey, Fishman, and Pentchev for the mutarotation of D-glucose 6-phosphate is 2·2 mol l^{-1}.[29]

A more striking rate enhancement is found in the mutarotation of 6-*O*-(*o*-hydroxyphenyl)-D-glucose (IX). In the pH range 2·0—6·89 the pH–rate profile is of the form: $k_0 = k_{spon} + k_{OH} \times 10^{(pH-pK_w)}$ (Table 13) with

TABLE 13

Rate constants for the spontaneous mutarotation of 6-*O*-(*o*-hydroxyphenyl)-D-glucose and 6-*O*-phenyl-D-glucose at 25·0°

pH	*a*	*b*	*c*
2·00	3·11	3·23	3·23
5·00	3·20	5·75	5·51
5·92		22·3	22·2
6·14	2·80	33·8	34·7
6·90	3·07	>120	184

[a] Experimental values of 10^4k_0/s^{-1} for the mutarotation of 6-*O*-phenyl-D-glucose. [b] Experimental values of 10^4k_0/s^{-1} for the mutarotation of 6-*O*-(*o*-hydroxyphenyl)-D-glucose. [c] Calculated values of 10^4k_0/s^{-1} for the mutarotation of 6-*O*-(*o*-hydroxyphenyl)-D-glucose from the equation $k_0 = k_{spon} + k_{OH} \times 10^{(pH-pK_w)}$ with $k_{spon} = 3{\cdot}23 \times 10^{-4}$ s^{-1} and $k_{OH} = 2{\cdot}28 \times 10^5$ l mol^{-1} s^{-1}.

TABLE 14

The phenolate ion-catalysed mutarotation of 6-*O*-phenyl-α-D-glucose at 25·0° (*I* 0·10M,[a] pH 9·13)

[phenolate]	10^3k_{obs}	10^3k_{calc}
0·006	8·78	8·88
0·004	6·43	6·41
0·003	5·26	5·18
0·002	3·92	3·94

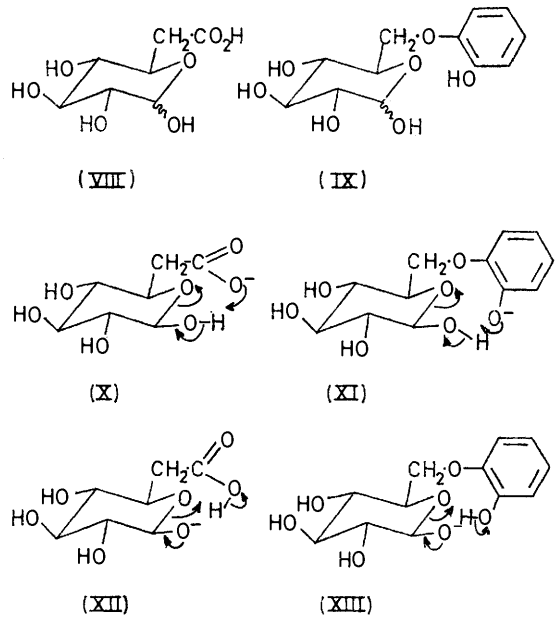
[a] Ionic strength maintained constant with potassium chloride. [b] Calculated from the expression $k_{calc} = k_{PhO}[PhO^-] + k_{int}$ with $k_{PhO} = 1{\cdot}24$ l mol^{-1} s^{-1} and $k_{int} = 1{\cdot}47 \times 10^{-3}$ s^{-1}.

$k_{spon} = 3{\cdot}23 \times 10^{-4}$ s^{-1} and $k_{OH} = 2{\cdot}28 \times 10^5$ l mol^{-1} s^{-1} and at pH 6·89 k_0 is more than 40 times greater than k_0 for the mutarotation of 6-*O*-phenyl-D-glucose; k_{OH} is 2700 times greater than the rate constant for the hydroxide-ion-catalysed mutarotation of 6-*O*-phenyl-D-glucose. This rate enhancement presumably arises from the rapidity of the mutarotation of the ionised form of 6-*O*-(*o*-hydroxyphenyl)-D-glucose and the rate constant is 14·0 s^{-1} (the pK_a value is 9·78). The second-order constant for the mutarotation of 6-*O*-phenyl-D-glucose catalysed by phenolate ion (pK_a = 9·98) is 1·24 l mol^{-1} s^{-1} (Table 14), and that for a base of pK_a 9·78 was estimated

[26] F. Gram, J. A. Hveding, and A. Reine, *Acta Chem. Scand.*, 1973, **27**, 3616.
[27] R. Stewart and J. D. Van Dyke, *Canad. J. Chem.*, 1972, **50**, 1992.
[28] Preliminary communication, B. Capon and R. B. Walker, *Chem. Comm.*, 1971, 1323.
[29] J. M. Bailey, P. H. Fishman, and P. G. Pentchev, *J. Biol. Chem.*, 1968, 243, 4827; *Biochemistry*, 1970, **9**, 1189.
[30] H. Schmid and G. Bauer, *Monatsh.*, 1965, **96**, 1503.
[31] B. Capon, *Essays in Chemistry*, 1972, **3**, 149.

to be 1·0 l mol^{-1} s^{-1}. The effective concentration of the internal phenolate ion in the mutarotation of 6-*O*-(*o*-hydroxyphenyl)-D-glucose is therefore *ca.* 14 mol l^{-1}.

The intramolecular catalysis could be general-base catalysis [(X) and (XI)] or general-acid catalysis [(XII) and (XIII)]. The latter mechanisms are analogous to the mechanism of equation (22) which was favoured for intermolecular catalysis. They are also stereochemically more favourable and (XI) would not be possible without the intervention of a chain of water molecules. On these grounds we favour (XII) and (XIII) for the mechanisms of the intramolecularly catalysed reactions.



(VIII) (IX)

(X) (XI)

(XII) (XIII)

EXPERIMENTAL

6-*Acetamido*-6-*deoxy*-β-D-*glucose*.— 6-Acetamido-6-deoxy-1,2-*O*-isopropylidene-α-D-glucofuranose [32] (2·5 g) was dissolved in 90% (v/v) trifluoroacetic acid–water (25 ml) and left for 15 min at room temperature; the solution was then evaporated *in vacuo* below 45°. Traces of water and trifluoroacetic acid were entrained out by several co-evaporations with absolute ethanol. The resulting syrup was dissolved in absolute ethanol and ether was added until the solution became turbid. This solution was left at 0° overnight, and crystalline 6-*acetamido*-6-*deoxy*-D-*glucose* (1·6 g) was obtained; m.p. 200—202° (from ethanol). Unlike the compound (m.p. 182—183°) previously reported this appears from the n.m.r. spectrum to be the β-anomer (Found: C, 43·2; H, 6·8; N, 6·3. $C_8H_{15}NO_6$ requires C, 43·4; H, 6·8; N, 6·3%); δ [$(CD_3)_2SO$] 1·66 (3H, s), 2·76—3·43 [6H, m, OH, and C(6)H_2], 4·23—4·79 (4H, m), 6·49 (1H, d, splitting 6 Hz), and 7·72br (1H, s); $\nu_{max.}$ (Nujol) 3500—3100, 1630, 1580, 1170, 1075, and 1035 cm^{-1}.

6-*Cyano*-6-*deoxy*-β-D-*glucose*.—The isopropylidene group of 6-cyano-6-deoxy-1,2-*O*-isopropylidene-α-D-glucofuranose [33] was removed by treatment with aqueous 90% trifluoroacetic acid as described above. The residue obtained on evaporation of the solvent was entrained with absolute ethanol to yield white crystalline material, m.p. 148—149° (from absolute ethanol) (lit.,[33] 147°) (Found: C, 44·4; H, 5·8; N, 7·4. Calc. for $C_7H_{11}NO_5$: C, 44·4; H, 5·9; N, 7·4%); δ (C_5D_5N) 3·22br (2H, s), 3·25—3·75 (4H, m), 5·30 (1H, d, splitting 6 Hz), and 7·25br (4H, s); $\nu_{max.}$ (Nujol) 3440, 3300, 2250, 1310, 1155, 1110, 1085, 1055, 1030, 1015, and 880 cm^{-1}.

6-*Deoxy*-α-D-gluco-*hepturonic Acid*.—The nitrile group of 6-cyano-6-deoxy-1,2-*O*-isopropylidene-α-D-glucofuranose was converted into a carboxylate group by heating with barium hydroxide at 90° for 4 h. The isopropylidene group of the resulting barium salt was hydrolysed with sulphuric acid to yield 6-deoxy-D-*gluco*-hepturonic acid, m.p. 175—177° (from absolute methanol) (lit.,[33] 175—176°) (Found: C, 40·2; H, 5·9. Calc. for $C_7H_{12}O_7$: C, 40·4; H, 5·8%), δ (C_5D_5N) 2·79—5·47 (6H, complex series of multiplets), 4·81 (1H, d, splitting 3·5 Hz), and 8·33 (5H, s); $\nu_{max.}$ (Nujol) 3320, 3200, 3000—2500, 1715, 1155, 1110, 1055, and 1015 cm^{-1}.

6-*Chloro*-6-*deoxy*-α-D-*glucose*.—Methyl 6-chloro-6-deoxy-α-D-glucoside [34] was hydrolysed by heating with hydrochloric acid [35] and the product recrystallised from acetone; m.p. 138° (lit.,[35] 135—136°).

6-O-*Methyl*-α-D-*glucose*.— 1,2-*O*-Isopropylidene-6-*O*-*p*-tolylsulphonyl-α-D-glucofuranose (7·48 g) was dissolved in absolute methanol and sodium methoxide (3·24 g) in absolute methanol (25 ml) was added. The mixture was left overnight at room temperature; t.l.c. (eluant ethyl acetate) then showed that the starting material (R_F 0·85) had been converted quantitatively into a main product (R_F 0·5) with two minor contaminants. The methanol was evaporated off *in vacuo* and the residue was extracted with dry acetone. The insoluble salts were filtered off and the filtrate was neutralised with hydrochloric acid. The precipitated salts were again filtered off and the solvent was removed *in vacuo*. The residue was chromatographed on silica (eluant ethyl acetate) to give a compound which showed only one spot (R_F 0·5) on t.l.c. with ethyl acetate. This could not be crystallised and so was hydrolysed (9 : 1 trifluoroacetic acid–water). The crude product (1·8 g, 46%) afforded 6-*O*-methyl-α-D-glucose (1·5 g), m.p. 141—142° (from ethanol) (lit.,[36] 142—143°) (Found: C, 43·2; H, 7·1. Calc. for $C_7H_{14}O_6$: C, 43·3; H, 7·3%); δ (C_5D_5N) 3·43 (3H, s), 4·1 (4H, m), 4·4 (2H, m), 5·88 (1H, d, splitting 3 Hz), and 6·8br (4H, s).

6-O-*Phenyl*-α-D-*glucose*.—Phenol (3·76 g) dissolved in dry dimethylformamide (DMF) was added dropwise to a cooled, stirred suspension of sodium hydride (0·96 g) in dry DMF (20 ml) in a flask equipped with an efficient reflux condenser. After evolution of hydrogen had ceased, 1,2-*O*-isopropylidene-6-*O*-*p*-tolylsulphonyl-D-glucofuranose (7·5 g) in dry DMF (10 ml) was added dropwise. The solution was allowed to come to room temperature and then heated at 45° for 4 h, diluted with water (200 ml), and thoroughly extracted with ethyl acetate (6 × 100 ml). The combined extracts were washed with 0·5M-sodium hydroxide and water (2 × 100 ml), dried (Na_2SO_4), and evaporated to yield a pale yellow oil (3·8 g). This could not be crystallised and the isopropylidene group was removed as before to yield 6-*O*-phenyl-α-D-glucose (3·0 g), m.p. 161—162° (from absolute ethanol) (lit.,[37] 180°) (Found: C, 56·4; H,

[32] F. Cramer, *Methods Carbohydrate Chem.*, 1962, **1**, 244.
[33] V. Prey and O. Szabolcs, *Monatsh.*, 1958, **89**, 350.
[34] M. E. Evans, L. Long, and F. W. Parrish, *J. Org. Chem.*, 1968, **33**, 1074.
[35] B. Helferich and H. Bredereck, *Ber.*, 1927, **60**, 1995.
[36] H. B. Wood, H. W. Diehl, and H. G. Fletcher, *J. Amer. Chem. Soc.*, 1957, **79**, 3862.
[37] H. Ohle, E. Euler, and R. Voullième, *Ber.*, 1938, **71**, 2250.

6·3. Calc. for $C_{12}H_{16}O_6$: C, 56·25; H, 6·3%); δ (C_5D_5N) 4·2—4·75 (6H, m), 5·85 (1H, d, splitting 3·5 Hz), and 6·40—7·40 (5H, m).

6-O-(o-*Hydroxyphenyl*)-β-D-*glucose*.—The method was similar to that used for the preparation of 6-*O*-phenyl-α-D-glucose except that instead of the excess of catechol being extracted with alkali it was separated by chromatography on silica (eluant 50% chloroform–ethyl acetate). 1,2-*O*-Isopropylidene-6-*O*-(*o*-hydroxyphenyl)-α-D-glucofuranose was obtained as a gum and the isopropylidene group was removed as before to yield 6-O-(o-*hydroxyphenyl*)-β-D-*glucose*, m.p. 153—154° (from ethanol–diethyl ether) (Found: C, 53·0; H, 5·9. $C_{12}H_{16}O_7$ requires C, 52·9; H, 5·9%), δ (C_5D_5N) 4·1br (s), 4·7 (3H, m), 5·15 (1H, d, splitting 6·5 Hz), and 6·55—7·35 (4H, m); $\nu_{max.}$ 3500, 3430, 3325, 1609, 1597, 1503, 1409, 1269, 1204, 1142, 1115, 1070, 1018, 927, and 734 cm^{-1}.

2-O-*Methyl*-β-D-*glucose*.—This compound was prepared as described by Hodge and Rist;[38] m.p. 159—160° (lit.,[38] 160°) (Found: C, 43·1; H, 7·3. Calc. for $C_7H_{14}O_6$: C, 43·3; H, 7·3%). The n.m.r. spectrum showed that a mixture of anomers was present (α : β *ca.* 1 : 2). The signals of the anomeric protons appeared at δ 5·23 (d, splitting 7·5 Hz) and 5·90 (d, splitting 3·5 Hz).

Other Aldoses.—α-D-Glucose, α-D-xylose, 6-deoxy-α-D-glucose, 2-acetamido-2-deoxy-α-D-glucose, 2-amino-2-deoxy-α-D-glucose hydrochloride, and β-gentiobiose were commercial materials which were recrystallised from aqueous ethanol.

Other Materials.—Pyridine, 4-methylpyridine, 2,6-lutidine, morpholine, and 2,2′-iminodiethanol were purified by refluxing over potassium hydroxide and fractionally distilling through a column packed with Fenske helices. 4-Ethoxypyridine was prepared by refluxing 4-chloropyridine hydrochloride with sodium ethoxide (3 equiv.) in ethanol and purified by fractional distillation. The purity of these amines was checked by g.l.c. Analytical grade Tris was used without further purification. The isotopic purity of D_2O and 20% DCl (Koch–Light) was checked by adding dioxan as a standard and measuring the quantity of water by n.m.r. spectroscopy. The concentration of protons was always less than 0·3 atom %.

Buffer Solutions.—These were prepared with degassed distilled water and potassium chloride was used to maintain the ionic strength constant. The pH values were measured at the temperature of the kinetic experiment with a Radiometer model 26 pH meter (G202C glass electrode and K401 calomel electrode).

Kinetic Measurements.—These were performed with a Perkin-Elmer 141 polarimeter fitted with a transmitting potentiometer which fed a voltage proportional to optical rotation to a strip-chart recorder and to a Solatron Compact Data Logger. The latter digitised the analogue voltages and punched their values at convenient time intervals with a Creed tape punch. Normally between 100 and 700 values were taken and fitted to the first-order rate equation using a generalised least-squares method.[39] Calculations were carried out with Glasgow University's KDF9 computer. The slopes and intercepts of the plots of k_{obs} against buffer concentration and the parameters of pH–rate profiles were also obtained by generalised least-squares methods. Each catalytic constant was evaluated from k_{obs} values determined at 5 or 6 concentrations of catalyst.

The cell of the polarimeter was maintained at 25° by circulating water from a Lauda thermostat bath. The temperature difference between the cell and bath was measured by a thermocouple and was always less than 0·1°. The temperature of the bath was measured by an N.P.L.-calibrated thermometer.

Determination of the Anomeric Compositions of the Aldoses at Equilibrium.—This was carried out for solutions in deuterium oxide by integration of the n.m.r. signals of the anomeric protons (Varian HA-100 spectrometer). The solutions were prepared from material which had undergone two pre-evaporations with D_2O to exchange the oxygen-bound protons. The results are given in Table 1.

We thank the S.R.C. for financial support, and Professor A. Neuberger and Mr. D. M. L. Morgan for sending us a copy of ref. 5 prior to publication and for a discussion.

[4/806 *Received*, 22*nd April*, 1974]

[38] J. Hodge and C. E. Rist, *J. Amer. Chem. Soc.*, 1952, **74**, 1498.
[39] W. Deming, ' Statistical Adjustment of Data,' Dover, New York, 1964; W. E. Wentworth, *J. Chem. Educ.*, 1965, **42**, 96, 162.