845. *The Acid and Alkaline Hydrolysis of Some Substituted Phenyl* α-D-*Glucosides.*

By A. N. Hall, Sheila Hollingshead, and H. N. Rydon.

The rates of hydrolysis, by acid and by alkali, of a number of substituted phenyl α-D-glucosides have been measured. As in the β-series, the rate of alkaline hydrolysis is markedly enhanced by the introduction of electron-withdrawing substituents. The rate of acid hydrolysis, however, is unaffected by substitution in the phenyl group and this difference from the β-series is ascribed to a difference in mechanism, that operating in the α-series involving a stereochemically controlled, diaxial elimination.

Nath and Rydon [1] studied the hydrolysis of a number of substituted phenyl β-D-glucosides by acid and alkali, and by the enzyme emulsin, and showed that acid and alkaline hydrolysis were oppositely affected by the nature of the substituents in the phenyl group, acid hydrolysis being facilitated by electron-releasing substituents and alkaline hydrolysis by electron-withdrawing substituents. There being no similar systematic study of the hydrolysis of the corresponding α-glucosides in the literature, we undertook such an investigation and now record our findings; parallel work on the enzymic hydrolysis of these glucosides will be submitted elsewhere.[2]

The substituted phenyl α-D-glucosides were prepared by catalytic de-acetylation [3] of the tetra-acetyl derivatives; these were obtained by heating the appropriate phenol and α-D-glucose penta-acetate with zinc chloride,[4] dissolved in a mixture of acetic acid and acetic anhydride,[5] under reduced pressure.[6] This condensation gives rise to a mixture of α- and β-anomers, the composition of which varies from experiment to experiment; this can only be separated into its components by an often very tedious process of fractional crystallisation. From *o*-chloro-, *o*-nitro-, and *o*-methoxy-phenol we were not able to obtain the α-anomers, although Montgomery *et al.*[5] report a successful preparation of *o*-nitrophenyl tetra-*O*-acetyl-α-D-glucoside by this method. In a recent paper, Helferich

[1] Nath and Rydon, *Biochem. J.*, 1954, **57**, 1.
[2] Hall, Hollingshead, and Rydon, *Biochem. J.*, in preparation.
[3] Zemplen and Pacsu, *Ber.*, 1929, **62**, 1613.
[4] Helferich and Hillebrecht, *Ber.*, 1933, **66**, 378.
[5] Montgomery, Richtmeyer, and Hudson, *J. Amer. Chem. Soc.*, 1942, **64**, 690.
[6] Sisido, *J. Soc. Chem. Ind., Japan*, 1936, **39**, 217; Kazutosi and Nisizawa, *Bull. Chem. Soc. Japan*, 1941, **16**, 155.

and Johannis,[7] repeating the work of Nisizawa,[8] likewise report inability to obtain crystalline *o*-methoxyphenyl tetra-*O*-acetyl-α-D-glucoside by this method; however, they succeeded in obtaining the free α-D-glucoside by de-acetylation of the amorphous α-tetra-acetate remaining after crystallisation of the β-anomer.

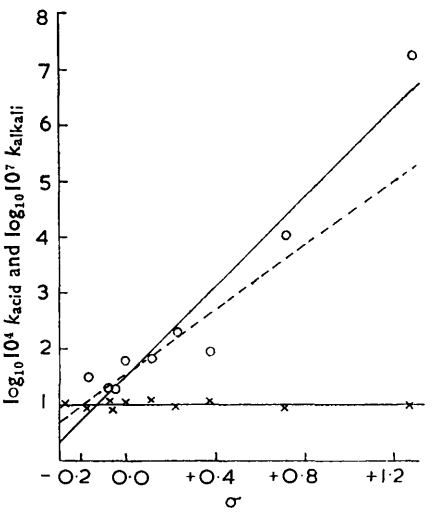
The hydrolysis of the glucosides, in acid solution (0·001M in 0·1N-hydrochloric acid at 70°) and in alkaline solution (0·002M in 3·9N-sodium hydroxide at 70°), was followed by colorimetric estimation of the liberated phenol. The results are collected in Table 1, in

TABLE 1.

Hydrolysis of substituted phenyl α-D-glucosides.

Subst.	σ	10^4k_{acid} (min.$^{-1}$)	$10^4k_{alk.}$ (min.$^{-1}$)	Subst.	σ	10^4k_{acid} (min.$^{-1}$)	$10^4k_{alk.}$ (min.$^{-1}$)
None	0·000	11·7	0·06	*p*-MeO	−0·268	10·7	—
o-Me	−0·054	7·7	0·02	*m*-Cl	+0·373	12·0	0·09
m-Me	−0·069	11·5	0·02	*p*-Cl	+0·227	9·7	0·20
p-Me	−0·170	9·2	0·03	*m*-NO_2	+0·710	9·1	10·7
m-MeO.....	+0·115	12·2	0·07	*p*-NO_2	+1·270	10·1	*ca.* 18,000

which they are expressed as first-order velocity constants, evaluated with respect to the glucosides; the substituent constants, σ, are those given by Hammett [9] for *meta*- and



Correlation of rates of hydrolysis of substituted phenyl α-D-glucosides with the nature of the substituent.
×, Acid hydrolysis. ○, Alkaline hydrolysis.

para-substituents and by Mamalis and Rydon [10] for *ortho*-substituents. The *p*-methoxy-compound was hydrolysed too slowly and the *p*-nitro-compound too rapidly under the standard alkaline conditions to enable $k_{alk.}$ to be determined for these compounds; the very approximate value given in the Table for the *p*-nitro-compound was obtained by extrapolation from experiments in which weaker alkali was used.

The logarithms of the velocity constants are plotted against the substituent constants in the Figure, the regression lines having been calculated by the method of least squares; two lines are shown for the alkaline hydrolysis, the full line having been calculated by including the uncertain value of $k_{alk.}$ for the *p*-nitro-compound and the broken line by omitting this value.

In agreement with the results of earlier, less extensive, studies [5,11] the alkaline

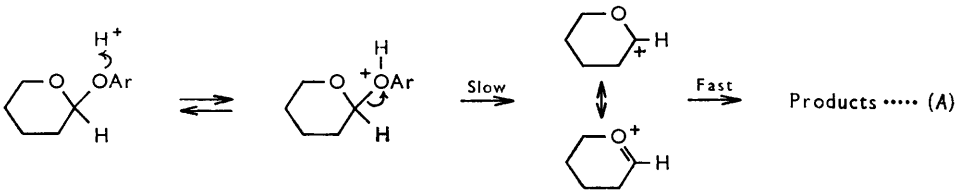
[7] Helferich and Johannis, *Z. physiol. Chem.*, 1960, **320**, 75.
[8] Nisizawa, *Bull. Chem. Soc. Japan*, 1941, **16**, 155.
[9] Hammett, " Physical Organic Chemistry," McGraw-Hill Book Co., New York, 1940, p. 186.
[10] Mamalis and Rydon, *J.*, 1955, 1049.
[11] McCloskey and Coleman, *J. Org. Chem.*, 1945, **10**, 184; Ballou, *Adv. Carbohydrate Chem.*, 1954, **9**, 59.

hydrolysis of the aryl α-D-glucosides resembles that of the β-anomers [1] in being strongly facilitated by electron-withdrawing substituents in the phenyl group, the effect being rather more marked in the α- (reaction constant [9] between +2·8 and +4·0) than in the β-series (reaction constant +2·48). This effect is to be expected for a reaction involving hydroxyl ions, and the high value of the reaction constant suggests that the attack is on an atom close to the substituents; we are not aware of any evidence which invalidates the suggestion [1] that the point of attack is $C_{(1)}$ of the benzene ring.

The rate of hydrolysis of the substituted phenyl α-D-glucosides by acid is, by contrast, almost unaffected by the nature of the substituents in the phenyl group; the Hammett reaction constant [9] is vanishingly small (−0·006) and the most divergent first-order velocity constant (that for the *o*-methyl compound) differs from the mean value ($10{\cdot}5 \times 10^{-4}$ min.$^{-1}$) for the ten glucosides studied by only 26%. In the β-series [1] the acid hydrolysis is facilitated by the presence of electron-releasing substituents in the phenyl group, the reaction constant being −0·66 and the first-order velocity constants for the fifteen β-glucosides studied varying between $0{\cdot}33 \times 10^{-4}$ min.$^{-1}$ and $8{\cdot}96 \times 10^{-4}$ min.$^{-1}$.

Bunton and Vernon and their co-workers [12] showed that the acid hydrolysis of α- and β-D-glucosides involved unimolecular decomposition of their conjugate acids, with fission of the glucosyl–oxygen bond, and put forward two alternative possible mechanisms; one of these (mechanism *A*, below) involved protonation of the glucosidic oxygen, followed by formation of a carbonium ion on $C_{(1)}$ of the glucose, and the other (mechanism *B*) protonation of the ring oxygen, followed by opening of the glucopyranose ring. Although Shafizadeh [13] has drawn attention to much circumstantial evidence in favour of a mechanism involving ring-opening, a forthcoming paper by Vernon and his co-workers [14] presents direct evidence (stereochemical inversion in acid-catalysed methanolysis and an oxygen-isotope effect in acid hydrolysis) which leads to the general conclusion that acid-catalysed solvolyses of α- and β-D-glucosides do not inolve ring-opening at any stage; we accept this conclusion as a basis for the discussion of our own results.

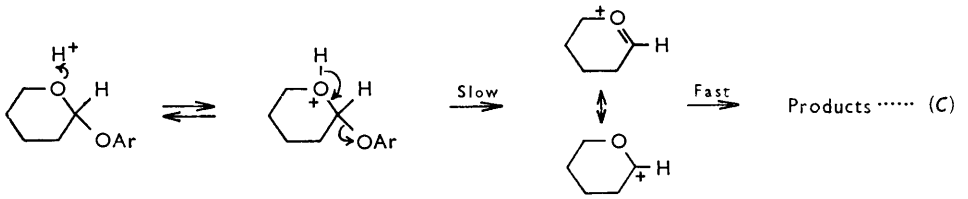
Mechanism *A* of Bunton and his co-workers [12] may be formulated as follows [15] for the acid hydrolysis of an aryl β-D-glucoside (here, and later, substituents on all but $C_{(1)}$ of the glucose are omitted for clarity):



Substituents in the aryl group will affect the observed rate of reaction by their influence on both the formation of the conjugate acid and its subsequent, " rate-determining," decomposition to the 'onium ion and will affect these two processes in opposing manners, electron-releasing substituents facilitating the former and hindering the latter. Such mutual partial cancellation of opposing effects will necessarily lead to a low value for the Hammett reaction constant and it is reasonable to suppose, with Bunton and his co-workers,[12] that the low value of the constant found for the acid hydrolysis of aryl β-D-glucosides [1] is, indeed, due to such partial cancellation. In the α-series, however, the effect of substitution in the aryl group is virtually nil and it is an unsatisfying invocation of coincidence to suppose this to be due to a chance exact cancellation of two opposing effects; although this explanation cannot be ruled out, we prefer to seek some other, more satisfying, alternative.

[12] Bunton, Lewis, Llewellyn, and Vernon, *J.*, 1955, 4419.
[13] Shafizadeh, *Adv. Carbohydrate Chem.*, 1958, **13**, 9.
[14] Banks, Meinwald, Rhind-Tutt, Sheft, and Vernon, *J.*, 1961, 3240.
[15] Cf. Edward, *Chem. and Ind.*, 1955, 1102.

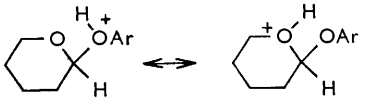
There is a third possible mechanism for the acid hydrolysis of glucosides, not considered by Bunton and his co-workers,[12] which leads from the conjugate acid protonated on the ring oxygen, to the same mesomeric 'onium ion as is involved in mechanism *A*. Unlike mechanism *B* of Bunton *et al.*,[12] which involves the same conjugate acid, this mechanism (*C*) does not involve ring opening and thus accommodates the findings of Vernon and his



co-workers.[14] This, we suggest, is the mechanism which operates in the acid hydrolysis of aryl α-D-glucosides. It is doubtful whether it can apply to alkyl α-D-glucosides; it is noteworthy in this connexion that, under comparable conditions, phenyl α-D-glucoside is hydrolysed by acid nearly 50 times more rapidly than methyl α-D-glucoside,[12] whereas the corresponding rate-ratio in the β-series is only about 8. Inspection of models shows that protonation of the ring oxygen in an α-D-glucoside is likely to take place in the axial position, the equatorial position being shielded by the aryloxy-group. The decomposition of the conjugate acid thus involves the *trans*-elimination of two axially placed groups (the 'onium hydrogen and the aryloxy-group), a type of reaction which is well known to occur very easily.[16] Such reactions are likely to be under steric, rather than electronic, control; if this is so, the vanishingly small effect of substitution in the aryl group, revealed by our experiments, is not surprising.

Mechanism *C* is not available for β-glucosides, since in these the aryloxy-group occupies an equatorial position and the ready diaxial elimination, which is the essential feature of this mechanism, is not possible. The aryl β-D-glucosides, therefore, undergo acid hydrolysis by mechanism *A* and it is noteworthy that they are well known to undergo acid hydrolysis more slowly than their α-anomers.

There is an apparent anomaly in that whereas, in mechanism *A*, the aryl β-D-glucosides are assumed to be protonated on the glucosidic oxygen, the aryl α-D-glucosides are assumed, in mechanism *C*, to be protonated on the ring oxygen; of these two atoms, the ring oxygen would be expected to be the more readily protonated, owing to the base-weakening effect of the aryl group on the glucosidic oxygen. Inspection of models, however, shows that in the β-glucosides the two oxygen atoms are so close together as to make it likely that the two conjugate acids are but two contributory forms of a single mesomeric cation:



Experimental

Preparation of Substituted Phenyl Tetra-O-acetyl-α-D-glucosides.—α-D-Glucose penta-acetate (30 g.) and the appropriate phenol (3—4 mol.) were fused together and the molten mixture treated with a solution of fused zinc chloride (7 g.) in a mixture of acetic acid and acetic anhydride (95 : 5 v/v; 20 ml.). The mixture was heated under reduced pressure at 120—130° for 1—2 hr. and the cooled residue dissolved in benzene or ethylene dichloride (300 ml.). The solution was thoroughly washed with 2N-sodium hydroxide and water, dried ($CaCl_2$), and evaporated to dryness under reduced pressure. After the resulting gum had crystallised, wholly or partly, a process which often required long storage in a vacuum-desiccator, it was fractionally crystallised until the α-anomer reached constant m. p. and optical rotation.

[16] Barton and Cookson, *Quart. Rev.*, 1956, **10**, 44; cf. Rhind-Tutt and Vernon, *J.*, 1960, 4637.

The following *tetra*-O-*acetyl*-α-D-*glucopyranosides*, so prepared, have not previously been described:

m-*Methoxyphenyl*, m. p. 78—80° (from methanol), $[\alpha]_D^{20}$ +117·0° (*c* 1·0 in $CHCl_3$) (Found: C, 55·5; H, 5·8. $C_{21}H_{26}O_{11}$ requires C, 55·5; H, 5·7%).

p-*Methoxyphenyl*, m. p. 81—82° (from methanol), $[\alpha]_D^{22}$ +158·9° (*c* 1·1 in $CHCl_3$) (Found: C, 55·1; H, 5·9%).

m-*Chlorophenyl*, m. p. 110° (from methanol), $[\alpha]_D^{22}$ +161·8° (*c* 0·6 in $CHCl_3$) (Found: C, 52·3; H, 5·1; Cl, 7·7. $C_{20}H_{23}ClO_{10}$ requires C, 52·3; H, 5·0; Cl, 7·7%).

p-*Chlorophenyl*, m. p. 100—101°, $[\alpha]_D^{20}$ +169·0° (*c* 1·0 in $CHCl_3$) (Found: C, 52·0; H, 5·3%). This compound was prepared by heating the free glucoside (250 mg.) with acetic anhydride (3 ml.) and pyridine (2 ml.) at 85° for 2 hr.; the product which separated when the mixture was poured into ice-water was recrystallised from methanol.

Preparation of Substituted Phenyl α-D-*Glucosides*.—The tetra-*O*-acetyl compound (0·01 mole) was dissolved in anhydrous methanol (100 ml.) and a freshly prepared solution of sodium methoxide (from sodium, 2 mg., and methanol, 10 ml.) added. The mixture was heated under reflux on a boiling-water bath for 5 min. and then kept at room temperature overnight. The methanol was then removed under reduced pressure and the residue recrystallised to constant m. p. and optical rotation.

The following α-D-*glucopyranosides* were prepared in this way (all rotations were determined for water solutions, *c* 0·4—1·0):

Phenyl, needles, m. p. 171—172°, from ethanol–light petroleum (b. p. 60—80°), $[\alpha]_D^{23}$ +177° (lit.,[17] m. p. 173—174°, $[\alpha]_D^{20}$ +180°).

o-Tolyl, m. p. 167° (from water), $[\alpha]_D^{21}$ +154° (lit.,[18] m. p. 170—172°, $[\alpha]_D^{20}$ +156°).

m-Tolyl, needles, m. p. 159—161°, from water, $[\alpha]_D^{21}$ +178° (Found: C, 57·8; H, 6·6. Calc. for $C_{13}H_{18}O_6$: C, 57·7; H, 6·7%) (lit.,[7] m. p. 165°, $[\alpha]_D^{25}$ +175°).

p-Tolyl, m. p. 189—190° (from aqueous ethanol), $[\alpha]_D^{23}$ +182° (Kazutosi and Nisizawa [6] record m. p. 190—191°, $[\alpha]_D^{18}$ +179°).

m-*Methxoyphenyl*, plates, m. p. 129—130°, from ethanol–light petroleum (b. p. 60—80°), $[\alpha]_D^{22}$ +166° (Found: C, 54·6; H, 6·3. $C_{13}H_{18}O_7$ requires C, 54·5; H, 6·3%).

p-*Methoxyphenyl*, m. p. 146—147° (from water), $[\alpha]_D^{18}$ +174° (Found: C, 55·1; H, 6·5%).

m-*Chlorophenyl*, needles, m. p. 163—164°, from water, $[\alpha]_D^{21}$ +176° (Found: C, 49·2; H, 5·2. $C_{12}H_{15}ClO_6$ requires C, 49·6; H, 5·2%).

p-*Chlorophenyl*, needles, m. p. 190—192°, from water or methanol, $[\alpha]_D^{20}$ +176° (Found: C, 49·5; H, 5·6%). In this case a pure product was obtained only after re-acetylation of the product obtained as usual and de-acetylation of the resulting tetra-acetate, with fractional crystallisation at each stage.

m-Nitrophenyl, m. p. 169° (from water), $[\alpha]_D^{21}$ +181° (lit.,[19] m. p. 172°, $[\alpha]_D^{20}$ +189°).

p-Nitrophenyl, prisms, m. p. 210°, from water, $[\alpha]_D^{20}$ +211° (lit.,[5] m. p. 216°, $[\alpha]_D^{20}$ +215°).

Kinetic Experiments.—The hydrolyses were carried out in 50 ml. flasks, fitted with cold-finger condensers,[20] immersed in a thermostat bath at 70° ± 0·05°. Samples were withdrawn from time to time for colorimetric determination of the liberated phenol, a Spekker photoelectric absorptiometer being used. For the nitrophenols the yellow colour developed in alkali [1] was measured with Ilford Spectrum Violet filters, No. 601; for the other phenols the method of Folin and Ciocalteu [21] was employed, the blue colours being measured by using Ilford Spectrum Red filters, No. 608.

The velocity constants were determined from the slopes of the usual log plots, which were satisfactorily linear.[22] The values recorded in Table 1 are the means of two independent determinations in each case. The approximate value of $k_{alk.}$ for the *p*-nitrophenyl compound was estimated by linear extrapolation from the following results at lower alkali concentrations:

[NaOH]	0·02N	0·1N	0·2N
$10^4 k_{alk.}$	667	1076	1503

[17] Fischer and Mechel, *Ber.*, 1916, **49**, 2814.
[18] Helferich, Lampert, and Sparmberg, *Ber.*, 1934, **67**, 1809.
[19] Pigman, *J. Res. Nat. Bur. Stand.*, 1944, **33**, 129.
[20] Braude, Jones, and Stern, *J.*, 1946, 401.
[21] Folin and Ciocalteu, *J. Biol. Chem.*, 1927, **73**, 627; cf. Hawk, Oser, and Summerson, " Practical Physiological Chemistry," Blakiston, Philadelphia, 1947, p. 879.
[22] For fuller experimental details see Hollingshead, Ph.D. Thesis, Manchester, 1958.

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MANCHESTER COLLEGE OF SCIENCE AND TECHNOLOGY,
MANCHESTER, 1.

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