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Hydrolysis of 2-(*p*-nitrophenoxy)tetrahydropyran: solvent and α-deuterium secondary kinetic isotope effects and relationships with the solvolysis of simple secondary alkyl arenesulfonates and the enzyme-catalyzed hydrolysis of glycosides†

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ABSTRACT: The effect of solvent composition in aqueous ethanol, trifluoroethanol and hexafluoropropan-2-ol on the rate constant and activation parameters for the uncatalysed hydrolysis of 2-(*p*-nitrophenoxy)tetrahydropyran (**1**) was investigated, and the $m(Y_{OTs})$ value is 0.60. This appreciable but less than maximal value is in accordance with an S_N1 mechanism with rate-limiting ionization. The α-deuterium secondary kinetic isotope effect (α-kie) for the uncatalysed hydrolysis of **1** is 1.17 in water (46 °C), 1.15 in aqueous trifluoroethanol (50% mole fraction, 70.6 °C) and 1.13 in aqueous ethanol (50% mole fraction, 70.6 °C). These values correspond to about 1.19 at 25 °C, which is characteristic of rate-limiting ionization in an S_N1 reaction and appreciably higher than values for enzyme-catalysed glycolysis. The α-kie is smaller under aqueous acidic conditions (1.07, 0.1 mol dm^{-3} hydrochloric acid, 20.2 °C) when **1** hydrolyses with acid catalysis. The previously reported α-kie for the hydrolysis of **1** in buffered aqueous dioxan (1.063, 25 °C) is now seen to correspond to acid-catalysed hydrolysis. These new results for **1** indicate that transition structures in enzyme-catalysed glycolyses with α-kie values of less than about 1.15 at 25 °C involve a lower degree of carbenium ion character than has hitherto been assumed. Copyright © 2004 John Wiley & Sons, Ltd.

KEYWORDS: solvolysis; carbenium ion; oxocarbenium ion; S_N1; α-deuterium kinetic isotope effect; solvent effect; Winstein–Grunwald equation; acetal hydrolysis; glycoside hydrolysis

INTRODUCTION

It is well known that simple hemiacetals are unstable in aqueous solution, whereas many furanose and pyranose hemiacetals formed by intramolecular cyclisation of carbohydrates are isolable compounds. Correspondingly, simple acetals, although stable in neutral or basic aqueous solution, are unstable in aqueous acids, whereas acetals derived from carbohydrates, i.e. glycosides, are generally relatively unreactive towards hydrolysis under all pH conditions, and are easily hydrolysed only with enzymic catalysis, which may occur with retention or inversion of configuration at the anomeric carbon. Over the years, there have been many investigations using naturally occurring carbohydrate and model substrates into how enzymes bring about this dramatic catalysis.[1] Central amongst model substrates which undergo uncatalysed hydrolysis is 2-(*p*-nitrophenoxy)tetrahydropyran (**1**) with its structural simplicity and a pH-independent reaction over a wide pH range; it has a good nucleofuge and none of the severely deactivating hydroxy groups in the electrofuge so characteristic of carbohydrate-derived substrates.[2,3] In non-acidic aqueous solution, **1** is generally believed to undergo initial heterolysis with separation of the 4-nitrophenoxide anion via transition structure **2** to give the oxocarbenium ion intermediate **3**, which undergoes subsequent nucleophilic capture by water. Under acidic conditions, **1** shows general acid catalysis, in contrast to the specific acid catalysis which is normally observed for acetals and ketals.[2–4] Substituent effects in the nucleofuge,[5] medium effects,[6] solute effects[7] and electronic and steric effects in the electrofuge of **1** and related acetals have been reported.[8] Craze and Kirby demonstrated that the uncatalysed hydrolysis of acetals is characterized by high sensitivity to the base strength of the nucleofuge, and proposed that C—O cleavage is more likely to be rate limiting than diffusional separation

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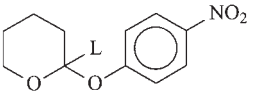
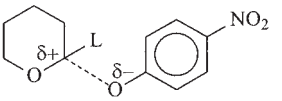
of the ion pair on the grounds that relatively stable carbenium ions do not usually undergo internal return.[9] Organic co-solvents, in particular acetonitrile, dimethyl sulfoxide and 1,4-dioxane, have a rate retarding effect on the hydrolysis of **1**,[6] in line with expectation for an S_N1 reaction in water, and the aqueous solvent deuterium kinetic isotope effect has been interpreted as indicating a degree of solvent participation in the transition state.[2] However, whilst solvent participation in nucleophilic solvolytic substitution reactions is usually nucleophilic, Fife and Jao[2] proposed that water facilitates departure of the nucleofuge from **1** by hydrogen bonding, i.e. electrophilic participation. Also, although it is generally agreed that **1** undergoes hydrolysis by an S_N1 mechanism, few authors have considered it within the extended ion pair range of S_N1 possibilities recognized since the work of Winstein, subsequently refined by Shiner and others.[10,11]

The α-deuterium secondary kinetic isotope effect (α-kie) has been widely used as a criterion of mechanism,[12] especially in attempts to characterize solvolytic reactions of secondary alkyl arenesulfonates.[13,14] The technique has also been used to investigate mechanisms of hydrolysis of acetals catalysed by enzymes such as lysozyme.[1,15] When the α-kie of an S_N1 reaction of a simple secondary alkyl substrate with a nucleofuge bonded through oxygen was believed to be in the region of 1.15 at 25 °C, values in the range ca 1.05–1.1 for enzyme-catalysed hydrolyses of glycosides were interpreted as indicating an appreciable degree of carbocation character in the transition state for formation of an oxocarbenium ion intermediate in the enzyme-catalysed reaction.[16] However, α-kie values of up to ca 1.19 (25 °C) are now known for S_N1 reactions that involve rate-limiting ionization of an oxygen bonded nucleofuge from a simple secondary alkyl residue.[17] Even higher values of ca 1.23 (25 °C) are observed in cases of rate-limiting separation of the initially formed intimate ion pair into a solvent-separated ion pair, for example when using highly ionizing, weakly nucleophilic solvents.[14,17] In this latter type of S_N1 mechanism, the trigonal carbocation is fully formed in the transition state and the α-kie attains its maximal value,[12–14] whatever that might be [an exceptionally high α-kie of 1.33 has recently been reported for solvolysis of (*Z*)-5-trimethylstannyl-2-adamantyl brosylate in 97% (w/w) aqueous trifluoroethanol at 25 °C,[18] which involves an unusally substituted nonplanar carbenium ion intermediate]. On the other hand, if ionization is rate limiting, vestigial bonding between the α-carbon and the oxygen of the departing nucleofuge remains in the transition structure, and a lower α-kie value is obtained according to the position of the transition structure within the reaction coordinate for bond heterolysis. Neighbouring group participation, either by lone pair donation from a heteroatom[19] or non-classical delocalization of σ-C—C electron density in the transition state,[20] also reduces the α-kie. When the substitution is bimolecular, however, the α-kie values are much closer to unity or even slightly less.[12] It follows that a value as low as ca 1.15 in a solvolytic reaction of a secondary alkyl substrate with an oxygen-bonded nucleofuge corresponds to an exceptionally early transition state in a rate-determining ionization, or appreciable nucleophilic assistance to ionization, or parallel unimolecular and bimolecular mechanisms. Consequently, if the solvolysis of simple secondary alkyl arenesulfonates is a proper model for the enzyme-catalysed hydrolysis of glycosides, results currently regarded as the upper limit for an S_N1 mechanism (ca 1.23 with full development of the carbenium ion centre in the transition state) require a reassessment of what now must be seen as rather low α-kie values for enzyme-catalysed hydrolysis of glycosides and related reactions.

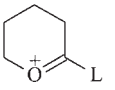
There have been relatively few α-kie results for the non-enzyme-catalysed hydrolysis of simple acetals which could act as proper models for enzyme catalysed reactions,[21,22] and some appreciable values have been reported for unambiguously bimolecular substitution reactions on acetals.[23,24] These have been ascribed to S_N2 mechanisms with only weakly bonded nucleophile and nucleofuge in the transition structure, i.e. the central carbon bears an appreciable positive charge, or to reactions with polarizable groups which bond through elements other than oxygen, e.g. when using iodide as a nucleophile. An early result by Bull *et al.* of $k^H/k^D = 1.063$ (buffered aqueous dioxane, 25 °C) for **1** was surprisingly low in view of even the early results mentioned above for secondary alkyl arenesulfonates.[22] This could have been evidence that the latter are not in fact good models for acetals, and that the endocyclic oxygen in an acetal somehow leads to an attenuated α-kie. For example, the lone pair donated by the endocyclic oxygen of the electrofuge in the ionization of **1** could exert the same sort of depressing effect on the α-kie as neighbouring group participation does in the solvolysis of (non-simple) secondary alkyl arenesulfonates.[19,20] If this is so, the basis for earlier interpretations of results for enzyme-catalysed glycolyses is totally undermined. Clearly, the result for **1** by Bull *et al.*[22] needed corroboration and the upper limit for the α-kie of an acetal undergoing hydrolysis by an S_N1 mechanism with rate-limiting ionization is needed. Furthermore, the possibility that acetals with



1a, L = H
1b, L = D



2, L = H, D



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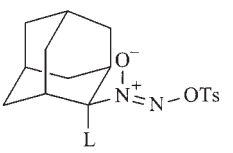

4, L = H, D

particular structural features may undergo S_N1 hydrolysis by rate-limiting ion pair separation, as is the case for some secondary alkyl arenesulfonates in highly ionizing media,[14] needs to be explored.

RESULTS

A preliminary account of some of this work has already appeared.[25]

Compound **1a** was prepared by a modification of a literature method;[2,26] it was established that pyridinium *p*-toluenesulfonate (tosylate) is a more effective catalyst than either 2,4-dinitrophenol or benzilic acid, and leads to more reproducible results than *p*-toluenesulfonic acid; dichloromethane proved to be the most effective solvent. Its rate of hydrolysis in water, aqueous ethanol, aqueous trifluoroethanol and aqueous hexafluoropropan-2-ol, with and without added hydroxide, was investigated UV spectrophotometrically by an established method,[17,27] and it was confirmed that the reaction is not base catalysed. Rate constants and activation parameters for reactions under various conditions are shown in Tables 1 and 2 and are in accord with results of previous investigations using narrower ranges of non-hydroxylic solvents.[6]

The isotopically labelled material (**1b**) was prepared from deuterated dihydropyran, which had been made by a literature method[28] and shown to be virtually fully deuterated at position 2 by ^{1}H NMR analysis (no signal at $\delta_H \sim 6.4$). In respects other than ^{1}H NMR spectroscopy (there was a barely detectable signal at δ_H 5.7–5.8), the deuterated product (**1b**) was identical with the unlabelled material. Integration of the NMR spectrum obtained by repeated scans indicated deuterium incorporation to be $>99\%$ at C-2. The α-kie was measured in slightly alkaline aqueous solution and as a function of the pH in buffered and unbuffered water with no significant concentration of any organic co-solvent, and the results are shown in Table 3. Clearly, the α-kie is ~ 1.17 at 46 °C in water under all conditions that do not allow acid catalysis, but in solutions of pH $<$ ca 3, the α-kie decreases and corresponds to the earlier value reported by Bull *et al.*[22] Results for aqueous ethanol and aqueous trifluoroethanol are also included in Table 3 and are seen to be very similar to those in non-acidic water once the temperature and lower precision are taken into account.

DISCUSSION

Solvent effect

As expected,[29] increasing proportions of co-solvents retard the rate of hydrolysis of **1** in water, and effects are illustrated in Figs 1 and 2. The effect of ethanol in Fig. 1 is similar to the effects of 1,4-dioxane, acetonitrile

Table 1. Rate constants at 25 °C and activation parameters for the hydrolysis of 2-(*p*-nitrophenoxy)tetrahydropyran (**1**) in aqueous ethanol[a]

EtOH (vol. %)	Mole fraction EtOH	Temperature range (°C)	$10^7\ k_{25}$ (s^{-1})	$\Delta H^{\ddagger}$ (kJ mol^{-1})	$\Delta S^{\ddagger}$ (J K^{-1} mol^{-1})
0	0.00	35–64	403	100	6
5	0.016	27–61	376	99	2
40	0.17	52–74	34.2	101	−9
52.0 (wt%)	0.298	44–75	9.27	99	−28
71.9 (wt%)	0.50	65–81	2.42	99	−39
82.2 (wt%)	0.643	60–82	0.950	100	−43
93	0.803	64–83	0.64[b]	97	−58
96.6 (wt%)	0.92	71–84	0.09[c]	—	—

[a] Reactions were usually 5×10^{-3} mol dm^{-3} in NaOH to allow UV monitoring of the formation of 4-nitrophenoxide at ca 405 nm. Estimated errors: ca 20% in k_{25}, ± 5 kJ mol^{-1} in $\Delta H^{\ddagger}$, ± 8 J K^{-1} mol^{-1} in $\Delta S^{\ddagger}$.
[b] Estimated errors: ca 100% in k_{25}, ± 8 kJ mol^{-1} in $\Delta H^{\ddagger}$, ± 15 J K^{-1} mol^{-1} in $\Delta S^{\ddagger}$.
[c] Approximate result.

Table 2. Rate constants at 25 °C and activation parameters for the hydrolysis of 2-(*p*-nitrophenoxy)tetrahydropyran (**1**) in aqueous trifluoroethanol[a]

TFE (vol. %)	Mole fraction TFE	Temperature range (°C)	$10^7\ k_{25}$ (s^{-1})	$\Delta H^{\ddagger}$ (kJ mol^{-1})	$\Delta S^{\ddagger}$ (J K^{-1} mol^{-1})
0.00	0.000	35–64	403	100	6
15.9 (wt%)	0.033	28–61	214	97	−12
50	0.200	46–74	12.2	102	−15
84.7 (wt%)	0.500	49–79	6.14	100	−29
97 (wt%)	0.853	51–81	4.84	98	−38
40% HFIP[b]	0.102	36–75	18.7	98	−26

[a] Reactions were usually 5×10^{-3} mol dm^{-3} in NaOH to allow UV monitoring of the formation of 4-nitrophenoxide at ca 405 nm. Estimated errors: ca 20% in k_{25}, ± 5 kJ mol^{-1} in $\Delta H^{\ddagger}$, ± 8 J K^{-1} mol^{-1} in $\Delta S^{\ddagger}$.
[b] Results for aqueous hexafluoropropan-2-ol; estimated errors: ca 50% in k_{25}, ± 10 kJ mol^{-1} in $\Delta H^{\ddagger}$, ± 15 J K^{-1} mol^{-1} in $\Delta S^{\ddagger}$.

Table 3. α-Deuterium secondary kinetic isotope effects on hydrolysis of 2-(*p*-nitrophenoxy)tetrahydropyran (**1**)[a]

pH	Medium	Temperature (°C)	10^4 k_H (s^{-1})	k_H/k_D
13.2	1 M NaOH	46.1	6.32	1.17
12.7	0.1 M NaOH	46.2	5.61	1.16
10.7	Unbuffered NaOH[b]	46.2	5.39	1.18
7.2	0.1 M Tris buffer	46.1	5.35	1.17
4.5	0.1 M acetate buffer	46.1	6.73	1.17
3.0	0.1 M chloroacetate buffer	46.1	14.2	1.11
1.0	0.1 M HCl	20.2[c]	6.50	1.07
—	EtOH–H_2O, mole fraction 1:1[b,d]	70.6	0.545	1.13
—	TFE–H_2O, mole fraction 1:1[b,d]	70.6	1.20	1.15

[a] Rates of reactions of protium and deuterium compounds were measured simultaneously in different cells of the same thermostatted cell block of a UV spectrophotometer, and all results shown are averages of at least three determinations. Estimated errors: ca 5% in k_H and ± 0.01 in the k_H/k_D rate ratios. A small increase in absorbance at 347 nm was monitored for the reactions at pH 1.0, 3.0 and 4.5; for all the other reactions, a much larger increase in absorbance at 405 nm due to 4-nitrophenolate was monitored.

[b] 10 µl of 1 mol dm^{-3} aqueous NaOH were added to ca 2.5 cm^3 of the reaction mixture in the UV cell.

[c] This reaction was too fast to follow by our technique at 46 °C.

[d] Estimated errors: ca 20% in k_H and ± 0.02 in the k_H/k_D rate ratios.

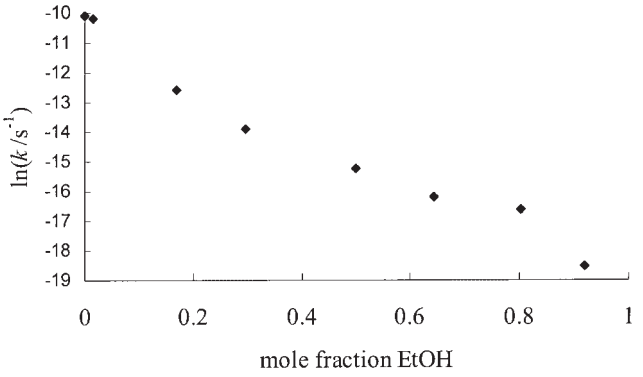

Figure 1. Graph of ln [k (s^{-1})] against mole fraction of ethanol for the hydrolysis of 2-(*p*-nitrophenoxy)tetrahydropyran (**1**) in aqueous ethanol, 25 °C

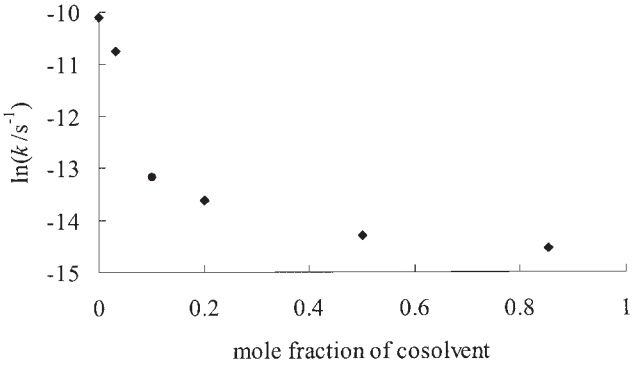

Figure 2. Graph of ln [k (s^{-1})] against mole fraction of cosolvent for the hydrolysis of 2-(*p*-nitrophenoxy)tetrahydropyran (**1**) in aqueous trifluoroethanol (◆) and hexafluoropropan-2-ol (●), 25 °C

and dimethyl sulfoxide reported earlier by Haak and Engberts,[6] except that the wider range of solvent composition now shows the plot of ln [k_{25} (s^{-1})] against mole fraction of ethanol to be slightly curved. This is more evident with the more acidic trifluoroethanol (TFE) and hexafluoropropan-2-ol (HFIP) as seen in Fig. 2. Interestingly, the limiting value at ever higher concentrations of these more acidic, less nucleophilic solvents corresponds to a faster reaction than in ethanol. This is as expected if the solvent effect is mainly due to its dielectric properties or to electrophilic rather than nucleophilic participation in the transition structure. However, a small measure of nucleophilic assistance in the transition state may be indicated by the ratio of ~6 in the rate constants in 40% aqueous ethanol and 85% aqueous trifluoroethanol (approximately the same Y_{OTs} values,[29] but very different nucleophilicities).

The effect of the polarity or ionizing power of a medium on the rate of solvolysis of a compound may be quantified using the Winstein–Grunwald equation[30] (or an extended[31] or modified[32] version). The gradient of a log($k_{compound}$) versus log($k_{standard}$) correlation close to unity means that the solvent effect for the solvolysis of the compound in question is the same as for the standard compound, and this is usually interpreted as evidence for closely similar mechanisms. The original standard compound, *tert*-butyl chloride, led to Y values of solvents being measures of their polarity or ionizing power, but 2-adamantyl tosylate is now regarded as more appropriate for substrates with oxygen-bonded nucleofuges, and Y_{OTs} values for solvents are now in more common use.[33] An m-value, the gradient of the plot of log[k (s^{-1})] against Y_{OTs}, approaching unity indicates an S_N1 mechanism closely similar to that of solvolysis of 2-adamantyl tosylate; an m-value lower than unity was initially interpreted as indicating that the reaction involves a low degree of charge separation in the transition structure (assuming there are other reasons for believing that the reaction is still S_N1), or that the mechanism is not S_N1 at all. More recent results, however, have shown that unambiguously S_N1 reactions may, for particular reasons, have relatively low m-values. The m-value for 2-adamantyl azoxytosylate (**4**), for example, which unquestionably undergoes S_N1 solvolysis with rate-limiting concerted

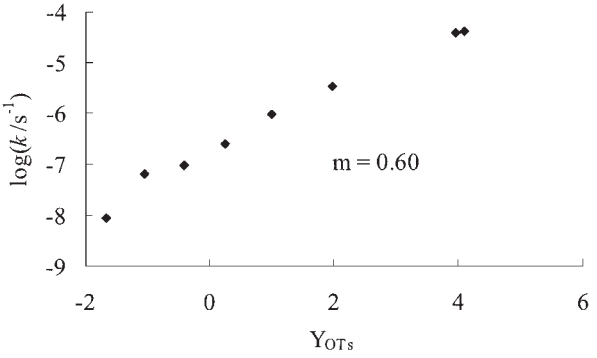

Figure 3. Graph of log [k (s^{-1})] against Y_{OTs} for the hydrolysis of 2-(*p*-nitrophenoxy)tetrahydropyran (**1**) in aqueous ethanol, 25 °C

heterolytic fragmentation,[34] is only 0.46.[35] Figure 3 shows the slightly curved plot for reactions of **1** in aqueous ethanol with an average *m*-value of 0.60 ($R > 0.99$). This less than maximal value is very similar to the result for S_N1 solvolysis of 1-adamantyl picrate ($m = 0.57$ in aqueous ethanol mixtures), a compound with a nucleofuge more akin to that in **1** than arenesulfonates are;[36] it is within the range expected for an S_N1 reaction of **1** with ionization rather than ion pair separation being rate limiting.

Activation parameters

Enthalpies of activation (Tables 1 and 2) are uniformly high, and the decrease in reactivity with increasing proportions of organic co-solvent is entirely due to changes in the entropy of activation. In pure water, formation of a more polar transition structure from a neutral substrate disrupts the structure of water itself, and the favourable entropy change associated with this and the bond-loosening in the substrate itself are not completely cancelled out by the loss of entropy due to enhanced solvation of the transition structure.[37] Consequently, S_N1 reactions of neutral substrates in pure water usually have modestly positive $\Delta S^{\ddagger}$ values, and **1** is no exception. Commonly, the same reactions in non-aqueous or mixed solvents have $\Delta S^{\ddagger}$ values closer to zero as the opposing contributions to $\Delta S^{\ddagger}$ more closely cancel each other out; the overall $\Delta S^{\ddagger}$ may even be slightly negative. In the present results, $\Delta S^{\ddagger}$ becomes increasingly negative as the proportion of co-solvent increases and the reaction becomes slower. This is wholly in accord with a reaction involving rate-limiting ionization and, as the reaction becomes slower, the transition structure (**2**) becomes increasingly like that of the incipient intermediate (the ion pair), i.e. more polar and hence more strongly solvated. Clearly, the increasing enthalpy cost of substrate ionization, as the proportion of co-solvent increases, remains balanced by the increasingly favourable enthalpy of transition state solvation.

α-Deuterium secondary kinetic isotope effect

The α-kie of 1.17 at 46 °C for the uncatalyzed hydrolysis of **1**, which corresponds to ca 1.19 at 25 °C, is virtually the same as values found for solvolysis of secondary alkyl arenesulfonates which react by rate-limiting ionization.[17] It appears, therefore, that the mechanism by which the adjacent endocyclic oxygen of **1** facilitates the departure of the nucleofuge and stabilizes the carbenium ion intermediate does not noticeably depress the α-kie. Even taking into account the slightly different conditions, this α-kie result for **1** is significantly larger than the value reported earlier by Bull *et al.* (1.063, aqueous dioxane, 25 °C).[22] Although the earlier reaction conditions were not explicitly described, the buffered reaction will have been mildly acidic, and it is known that **1** is susceptible to acid catalysis at low pH. The present results in Table 3 now show that the acid-catalysed reaction of **1** has an appreciably lower α-kie than the uncatalysed reaction, which almost certainly accounts for the earlier low result. We ascribe this depressed α-kie to an appreciably earlier transition state in the rate-limiting heterolysis due to the much better nucleofuge (4-nitrophenolate hydrogen-bonded to a hydronium ion) rather than to nucleophilic assistance. This corresponds to an unusual sensitivity of the α-kie to the effectiveness of the leaving group bonded through a common element, but is in accord with the previously reported sensitivity of the reaction to the base strength of the nucleofuge.[9] Moreover, if the similarity in the isotope effects for uncatalysed hydrolysis of **1** and solvolysis of simple secondary alkyl arenesulfonates indicates a similar extent of C—O bond cleavage in the respective transition structures, the facilitation provided by participation of a lone pair on the endocyclic oxygen of the acetal must be compensating for the very considerable difference in leaving group abilities of arenesulfonate and 4-nitrophenoxide (as reflected by the pK_a values of their conjugate acids).

Conclusions and relevance to enzyme-catalysed glycolysis

A value approaching ca 1.15 for an enzyme-catalysed glycolysis was previously taken to imply a late transition state on the way to an sp^2-hybridized oxocarbenium ion intermediate.[16] The principal basis for this inference was that solvolyses of simple secondary alkyl arenesulfonates had α-kie values of ca 1.15 and were believed to involve rate-determining formation of sp^2-hybridized carbenium ion intermediates in S_N1 reactions.[12,13] Subsequent considerations by Jencks and others of the putative oxocarbenium ion intermediates in glycolysis reactions indicated lifetimes in aqueous solution short compared with times of molecular vibrations,[1,24,38] i.e. oxocarbenium ions derived from typical carbohydrates are probably too unstable to exist in aqueous media. Hence,

although the evidence for intermediates in enzyme-catalysed glycolyses appears convincing,[1,39] these are clearly not free oxocarbenium ions. However, arguments against oxocarbenium ion intermediates based on an *overall* α-kie being normal rather than inverse are invalid.[40] It never can be *appreciably* inverse when the reaction centre in the initial state to which the kinetics measurements relate is saturated (sp^3), and could be only *slightly* inverse in the event of a rate-determining S_N2 step with a tight transition structure,[12] i.e. asynchronous in the associative sense. The current consensus is that the intermediates are covalently bonded glycosyl enzymes,[1] i.e. reactant, product *and* intermediate in enzyme-catalysed glycolysis have fully covalent sp^3-hybridized carbon at the anomeric centre, and either their formation or subsequent hydrolysis may be rate limiting. In the light of these considerations, and α-kie values greater than unity for bimolecular substitution reactions on simple acetals,[23,24] enzymatic glycolysis α-kie results were re-interpreted in terms of a bimolecular rate-limiting trigonal-bipyramidal transition structure with the central anomeric carbon only weakly bonded apically to *both* nucleofuge *and* nucleophile, and bearing an appreciable degree of positive charge. This was sometimes referred to as an S_N1-like S_N2 reaction with an open or exploded transition structure, i.e. a concerted mechanism which is appreciably asynchronous in the dissociative sense, regardless of whether it is the step for the intermediate's formation or its hydrolysis. It follows that the *overall* α-kie value for glycolysis sheds little light on whether the formation of the intermediate or its subsequent hydrolysis is rate limiting.

However, subsequent results have established that the α-kie associated with rate-limiting ionization of a simple secondary alkyl arenesulfonate can be as high as 1.19 (25 °C).[17] Moreover, the present results establish beyond doubt the close numerical similarity between the α-kie for solvolysis of simple secondary alkyl arenesulfonates and the uncatalysed hydrolysis of **1** (1.17 in water at 46 °C, corresponding to ca 1.19 at 25 °C) when initial heterolysis is rate limiting in both cases. The latter is obviously the more appropriate comparator for the hydrolysis of acetals. Values for enzyme-catalysed glycolyses (up to $\sim$1.15), therefore, are a smaller proportion of the maximum for acetal hydrolysis (at least 1.19) and consequently the degree of positive charge on the anomeric carbon in the transition structure must be less than was earlier believed. It remains to identify conditions under which **1** or an analogue reacts with rate-limiting ion pair separation and to determine the α-kie for that process. A reasonable interim assumption is that it will be very similar to that for the corresponding solvolytic mechanism of simple secondary alkyl arenesulfonates, ca 1.23 at 25 °C. This reduces further the estimated degree of carbenium ion character at the anomeric carbon in the rate-limiting transition structure of an enzyme-catalysed glycolysis, and hence the degree to which the mechanism of that step may be described as S_N1-like.

EXPERIMENTAL

The preparation of 2-(*p*-nitrophenoxy)tetrahydropyran (**1a**) was based on literature methods[2,26] but using pyridinium tosylate as catalyst and dry dichloromethane as solvent. The reaction was worked up in the usual way to give a quantitative yield of an oil which subsequently crystallized; m.p. (recryst. petroleum spirit, b.p. 30–40 °C) 57–58 °C (lit.[2] 59–60 °C); δ_H 8.20 (d) and 7.15 (d) (4H, arom), 5.75 (m, 1H), 3.6–3.9 (m, 2H), 1.6–1.8 (m, 4H), 1.8–2.1 (m, 2H); δ_C: 162.2 (C-1′), 142.0 (C-4′), 125.8 (C-3′,5′), 116.3 (C-2′,6′), 96.4 (C-2), 62.1 (C-6), 30.0 (C-3), 25.0 (C-4), 18.3 (C-5).

α-Deuteration of 2,3-dihydropyran. tert-Butyllithium (12 cm^3, 1.1 mol dm^{-3} in pentane, 15 mmol) was added dropwise to a mixture of THF (0.8 cm^3) and 2,3-dihydropyran (1 cm^3, 11 mmol) at −78 °C. The reaction mixture was allowed to warm to room temperature and then cooled again to −78 °C, whereupon deuterium oxide (99.9% ^2H_2O, 1 cm^3) was added. The reaction mixture was again warmed to room temperature; absence of a signal in the region δ_H 6.3–6.5 in the ^1H NMR spectrum of the organic phase indicated complete deuteration.

*2-Deutero-2-(p-nitrophenoxy)tetrahydropyran (**1b**).* In another preparation of α-deuterated 2,3-dihydropyran, anhydrous calcium chloride was added to the ^2H_2O-quenched reaction mixture to aid phase separation, the organic phase was removed by Pasteur pipette and the deuterated aqueous phase was extracted with dry dichloromethane (2×4 cm^3). The combined organic phase was dried by percolation through a plug of $MgSO_4$ plus Na_2CO_3 contained by cotton-wool in a Pasteur pipette. 4-Nitrophenol (1.53 g, 11 mmol) and a few crystals of pyridinium tosylate were added to the dried organic solution and the reaction mixture was stirred for 9 days at room temperature with monitoring by TLC. After the usual workup, the title compound was obtained in 15% yield; ^1H NMR analysis showed only an extremely weak signal in the region δ_H 5.5–5.8, indicating virtually complete deuteriation.

Kinetics. Water used for solvolytic studies was glass distilled and ethanol was dried by fractional distillation from magnesium ethoxide; solvents for kinetics were prepared by mixing accurately measured volumes or weights of components. Reactions were initiated by addition of up to 10 µl of a stock solution of the substrate in a non-reactive solvent, e.g. diethyl ether or ethanol. Our UV-based kinetic method has already been described,[27] and reactions were normally monitored for at least 2–3 half-lives. Under non-acidic unbuffered conditions, 10 µl of 1 mol dm^{-3} sodium hydroxide were normally added to ensure formation of 4-nitrophenoxide anion and hence a substantial increase in absorbance; this was not effective in HFIP. All reactions were cleanly

first order and standard deviations on individual rate constants were typically less than 1%.

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