The α-Deuterium Secondary Kinetic Isotope Effect upon the Hydrolysis of 2-(*p*-Nitrophenoxy)tetrahydropyran and its Relationship to Values for the Solvolysis of Secondary Alkyl Arenesulfonates and the Enzyme-catalysed Hydrolysis of Acetals

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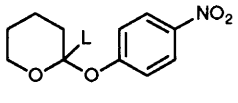
The α-deuterium secondary kinetic isotope effect for the uncatalysed hydrolysis of 2-(*p*-nitrophenoxy)tetrahydropyran is 1.17 in water (46 °C), a result which is very similar to values for the solvolysis of simple secondary alkyl arenesulfonates which proceed with rate-limiting ionization and appreciably higher than results for enzyme-catalysed hydrolysis of other acetals.

The α-deuterium secondary kinetic isotope effect (α-KIE) has been widely used as a criterion of mechanism, and has been particularly effective in characterizing solvolytic reactions of secondary alkyl arenesulfonates.[1] The general strategy has been to compare the result for the reaction under investigation with known results for other reactions whose mechanisms are believed to be understood, the comparison invariably being supported by semiquantitative theory propounded some years ago.[2] The technique was also claimed at an early stage as one that could help to elucidate some of the details of the mechanisms of hydrolysis of acetals catalysed by enzymes such as lysozyme.[3]

When the α-KIE of an S_N1 reaction of a secondary alkyl substrate with a nucleofuge bonded through oxygen was believed to be in the region of 1.15 at 25 °C, comparable values for the enzyme-catalysed hydrolyses of acetals were reasonably interpreted as indicating a high degree of carbocation character in the transition state of the enzyme catalysed reaction.[4] More recently, it has been recognised that an α-KIE of *ca.* 1.19 (25 °C) is expected for an S_N1 reaction that involves rate-limiting ionization of an oxygen-bonded nucleofuge from a simple secondary alkyl residue, and an even higher value of *ca.* 1.23 (25 °C) is observed in the case 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.[5] 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. On the other hand, if ionization is rate limiting, vestigial bonding remains between the α-carbon and the oxygen of the departing nucleofuge and lower α-KIE values are obtained according to the position of the transition state within the reaction coordinate for bond heterolysis. If the solvolysis of secondary alkyl arenesulfonates is a proper model for the hydrolysis of acetals, these results require a reassessment of early interpretations of the significantly lower α-KIE values reported for enzyme-catalysed hydrolysis of acetals and related reactions.

There have been only few α-KIE results for the hydrolysis of acetals themselves which could act as proper models for the enzyme-catalysed reactions,[6] and an early result by Bull *et al.* of k_H/k_D = 1.063 (aqueous dioxane, 25 °C) for the title compound **1** was surprisingly low in view of the results mentioned above for secondary alkyl arenesulfonates.[7] This could have been evidence that the latter are not in fact good models for acetals, and that the non-departing oxygen in an acetal somehow leads to an attenuated α-KIE. If this is so, the basis for the earlier interpretation of results for enzyme-catalysed reactions is totally undermined. Clearly, Bull *et al.*'s result needed corroboration, and the upper limit for the α-KIE of an acetal undergoing hydrolysis by an S_N1 mechanism was needed. Furthermore, the possibility that reactive acetals may, in the limit, undergo S_N1 hydrolysis by rate-limiting ion-pair separation, as is the case for secondary alkyl arenesulfonates, needed to be explored.



1a; L = H
1b; L = D

Compound **1a** was prepared by a modification of a literature method,[8] and its rate of hydrolysis in slightly alkaline water, aqueous ethanol, and aqueous trifluoroethanol was investigated by monitoring the increase in UV absorbance due to the formation of *p*-nitrophenolate by an established method;[5,9] it was confirmed that the reaction is not base catalysed. The very substantial solvent effects upon the reaction rate and activation parameters are entirely in accord with the results of other investigations,[10] and as expected for an S_N1 mechanism with rate-limiting initial ionization. The isotopically labelled material **1b** was prepared from deuteriated dihydropyran which had been made by a literature method[11] and shown to be virtually fully deuteriated at position 2 by ^1H NMR analysis. The α-KIE in unbuffered alkaline water was 1.17 (46 °C). Taking into account the temperature difference, this is significantly larger than the result reported earlier by Bull *et al.*, but entirely in line with values obtained previously for secondary alkyl arenesulfonates undergoing solvolysis with rate-limiting ionization. Whilst the reaction conditions employed by Bull *et al.* were not explicitly described, the reaction was buffered under mildly acidic conditions, and it is known that compound **1** is susceptible to acid catalysis at low pH. Consequently, we determined the α-KIE as a function of the pH in buffered aqueous media with no significant concentration of any organic cosolvent, and the results are shown in Table 1. Clearly, the α-KIE is about 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 report from Bull *et al.* It appears, therefore, that the uncatalysed reaction of **1** has an α-KIE entirely in line with values found for solvolysis of secondary alkyl arenesulfonates which react by rate-limiting ionization,

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

pH	System	*T*/°C	$k_H/10^4\ s^{-1}$	k_H/k_D
	1 mol dm^{-3} NaOH	46.1	6.32	1.17
	0.1 mol dm^{-3} NaOH	46.2	5.61	1.16
10.7	Unbuffered NaOH[b]	46.2	5.39	1.18
7.2	Tris buffer	46.1	5.35	1.17
4.5	$MeCO_2^-$ buffer	46.1	6.73	1.17
3.0	$ClCH_2CO_2^-$ buffer	46.1	14.2	1.11
	1 mol dm^{-3} HCl	20.2[c]	6.50	1.07

[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 in the rate constants are *ca.* 5% 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 4.5 and below; for all the other reactions, a much larger increase in absorbance at 405 nm due to *p*-nitrophenolate was monitored. [b] 10 μl of 1 mol dm^{-3} aqueous NaOH was added to *ca.* 2.5 cm^3 of the reaction mixture in a UV cell. [c] This reaction was too fast to follow by our technique at 46 °C.

but that acid catalysis leads to a significantly lower value. Presumably, this corresponds to an appreciably earlier transition state in the rate-limiting ionization due to the much better leaving group in the protonated substrate. This corresponds to a quite unusual sensitivity of the value of the α-KIE to the effectiveness of the leaving group bonded through a common element. It remains to identify conditions under which **1** reacts with rate-limiting ion pair separation and to determine the α-KIE for that process, whereupon the results of the enzymic reactions may be more confidently interpreted. It is clear from the present results, however, that the endocyclic oxygen of **1**, which facilitates the departure of the nucleofuge, does not significantly affect the α-KIE. 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 transition states, the nucleophilic assistance provided by the α-oxygen of the acetal must be compensating for the very considerable difference in leaving group abilities of arenesulfonate and *p*-nitrophenoxide as reflected by the pK_a values of their conjugate acids.

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