Iminolactones. II. Catalytic Effects on the Nature of the Products of Hydrolysis

Bruce A. Cunningham[1] and Gaston L. Schmir

Contribution from the Department of Biochemistry, Yale University, New Haven, Connecticut 06510. *Received September 22, 1965*

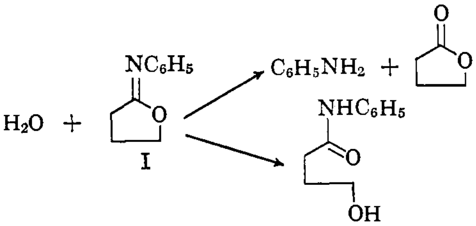
Abstract: The hydrolysis of the iminolactone N-phenyliminotetrahydrofuran (I) yields aniline (and butyrolactone) at acid pH and γ-hydroxybutyranilide in alkaline solution. The extent of conversion of iminolactone to aniline at constant pH in the region of pH 7–9 depends upon the nature and concentration of the buffer. Low concentrations of phosphate or bicarbonate buffers direct the breakdown of a tetrahedral intermediate in favor of aniline formation, at the expense of the yield of hydroxyanilide, *without affecting the rate of iminolactone disappearance.* A mechanism including cyclic proton transfers in the carbinolamine addition intermediate appears to account quantitatively for the observed buffer effects.

The central role ascribed[2] to iminolactones as intermediates in the selective, chemical cleavage of peptide bonds led us to investigate[3] the mechanism of hydrolysis of this hitherto little-studied class of substances. The effect of pH on the rate of hydrolysis and on the nature of the products provided evidence for the existence of an addition intermediate of significant stability, formed during the hydrolysis process. In this respect, the behavior of iminolactones parallels that of numerous other systems, where nucleophilic reaction at carbonyl, acyl, or imino (imidate, thioimidate) function leads to the appearance of transient, but kinetically demonstrable, addition intermediates.[4] To account for the unexpected observation that the composition of the product mixture varied with pH, we proposed[3] the occurrence of two pathways of product formation, one involving a neutral carbinolamine intermediate, the other its conjugate base.

Further scrutiny of the factors influencing the nature of the products of iminolactone hydrolysis has revealed the unusual response of the reaction course to the presence of low concentrations of certain buffers. In this report, we describe experiments which suggest that the breakdown of carbinolamine addition intermediates may be exceptionally sensitive to catalytic processes, particularly those of a concerted acid–base nature.

Results

The hydrolysis of N-phenyliminotetrahydrofuran (I) produces either aniline (and γ-butyrolactone) or γ-hydroxybutyranilide (eq. 1). While aniline is the predominant reaction product in the pH range 0–6, it is formed in decreasing amounts at pH 6–9 (imidazole and Tris buffers); above pH 9, γ-hydroxybutyranilide is essentially the sole product of hydrolysis.[3] As shown in Figure 1, the gradual transition from the appearance of aniline to that of hydroxyanilide is reasonably well correlated with the ionization pattern of a hypothetical univalent acid of pK' = 7.07 (Figure 1, solid curve). This behavior was interpreted[3] in terms of the ionization[5] of a tetrahedral addition intermediate (eq. 2); according to this formulation, decomposition of the neutral carbinolamine (or of its zwitterionic equivalent) leads to the formation of aniline (and butyrolactone); alternatively, breakdown of the anionic intermediate results in the appearance of hydroxyanilide.



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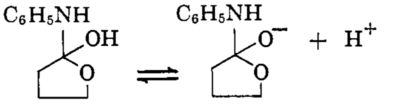

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In accord with the hypothesis that the aniline yield from iminolactone hydrolysis is solely a function of pH, substitution of Tris for imidazole buffers at fixed pH caused no significant alteration of product composition. However, the use of phosphate (0.03 *M*) or bicarbonate buffers instead of the customary amine buffers resulted in major increases in aniline yields (Figure 1). Control experiments indicated that the increased formation of aniline was not the result of hydrolysis of initially formed γ-hydroxybutyranilide. These observations initiated a systematic investigation of buffer effects on iminolactone hydrolysis, the results of which are detailed below.

In typical experiments, iminolactone hydrolysis was carried out at 30° in 0.03 *M* Tris buffer (whose presence ensured constancy of pH) containing increasing amounts of an additional buffer. Aniline determination was

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[1] This work is taken from a dissertation to be presented by B. A. C. in partial fulfillment of the requirements for the Ph.D. Degree, Yale University.

[2] B. Witkop, *Advan. Protein Chem.*, **16**, 221 (1961); L. A. Cohen and B. Witkop in "Molecular Rearrangements," Part 2, P. de Mayo, Ed., Interscience Publishers, Inc., New York, N. Y., 1964, p. 1004; B. Witkop and L. K. Ramachandran, *Metab. Clin. Exptl.*, **13**, 1016 (1964).

[3] G. L. Schmir and B. A. Cunningham, *J. Am. Chem. Soc.*, **87**, 5692 (1965).

[4] (a) W. P. Jencks, *Progr. Phys. Org. Chem.*, **2**, 63 (1964); (b) for a compilation of references to recent work in this area, see A. J. Kirby and W. P. Jencks, *J. Am. Chem. Soc.*, **87**, 3217 (1965), footnote 3; (c) G. L. Schmir, *ibid.*, **87**, 2743 (1965).

[5] It has been emphasized[3] that the *apparent* dissociation constant derived from the data of Figure 1 (pK' = 7.07) is not necessarily equivalent to the acid dissociation constant of the carbinolamine. The available information does not permit evaluation of the latter constant. Indeed, the reported[6] pK value of 9.07 (in H_2O) for a carbinolamine of related structure suggests that the ionization process of eq. 2 is characterized by a pK value appreciably higher than 7.

[6] H. Ott, A. J. Frey, and A. Hofmann, *Tetrahedron*, **19**, 1675 (1963); see also A. Hofmann, H. Ott, R. Griot, P. A. Stadler, and A. J. Frey, *Helv. Chim. Acta*, **46**, 2306 (1963).

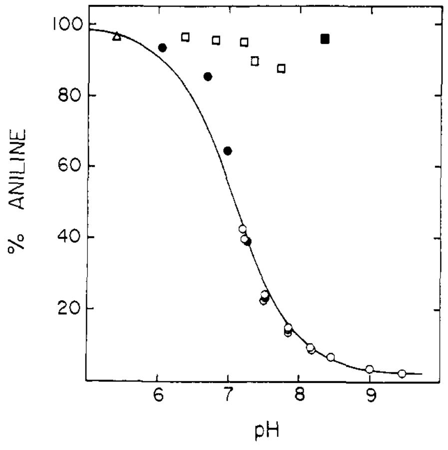

Figure 1. Effect of pH and of buffers on the nature of the products of iminolactone hydrolysis: △, acetate, 0.05 *M*; ●, imidazole, 0.01 *M* at pH 6, 0.0005 *M* at all other pH values; ○, Tris, 0.03 *M*; □, phosphate, 0.03 *M*; ■, sodium bicarbonate, 0.1 *M*. Solid line is the calculated titration curve of a monovalent acid of p*K* = 7.07.

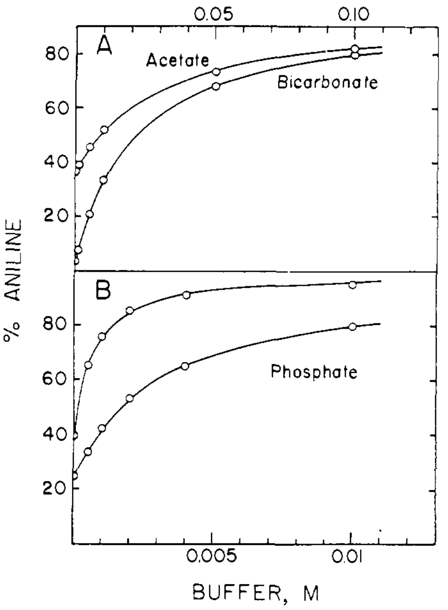

Figure 2. Effect of added buffers on yield of aniline resulting from iminolactone hydrolysis: (A) acetate, pH 7.26; bicarbonate, pH 9.02 (abscissa, upper scale); (B) phosphate, upper curve, pH 7.27; lower curve, pH 7.88 (abscissa, lower scale). All reaction mixtures contain 0.03 *M* Tris.

performed on completion of hydrolysis (7–10 half-lives). The striking ability of added phosphate, bicarbonate, or acetate buffers to increase the extent of conversion of iminolactone to aniline is illustrated in Figure 2. Additional data are presented in Table I. In contrast, variation of Tris or imidazole buffer concentration at fixed pH only slightly affects aniline yield (Table II). The following conclusions may be drawn from these results.

Table I. Effect of Phosphate, Bicarbonate, and Acetate Buffers on Aniline Yield from Iminolactone Hydrolysis[a]

Expt.	pH	Buffer,[b] *M*	Aniline, %	$K_{app} \times 10^3$, *M*
		(a) Phosphate		
1	7.25	...	39.5	0.69
	7.25	0.0005	65.0	
	7.25	0.002	85.2	
	7.21	0.01	95.2	
2	7.87	...	25.6	3.1
	7.88	0.0005	34.7	
	7.88	0.005	71.0	
	7.88	0.01	80.2	
3	8.28	...	14.3	6.9
	8.29	0.001	24.2	
	8.29	0.01	57.1	
	8.36	0.1	90.3	
4	8.99	...	5.8	59
	9.00	0.005	15.3	
	9.02	0.05	51.7	
5[c]	7.34	...	52.2	1.1
	7.33	0.0005	66.8	
	7.33	0.001	74.4	
	7.31	0.01	93.4	
6[d]	7.20	0.03	95.0	
	7.72	0.03	87.7	
		(b) Bicarbonate		
7	7.24	...	40.6	1.2
	7.25	0.0005	58.8	
	7.28	0.002	79.2	
8	9.02	...	3.5	21.5
	9.02	0.005	21.0	
	9.00	0.05	68.1	
	8.97	0.1	80.1	
9[e]	8.4	0.1	96.2	
		(c) Acetate		
10	7.85	...	13.3	395
	7.85	0.04	20.6	
	7.87	0.2	40.6	

[a] At 30° in 10% acetonitrile–water, μ = 0.5. [b] All reaction mixtures contain 0.03 *M* Tris buffer, except where stated. [c] Tris buffer replaced by 0.03 *M* imidazole buffer. [d] Phosphate buffer only. [e] Bicarbonate buffer only.

Table II. Effect of Tris and Imidazole Buffers on Aniline Yield from Iminolactone Hydrolysis[a]

Expt.	pH	Buffer, *M*: Tris	Buffer, *M*: Imidazole	Aniline, %
11	7.68	0.01	...	23.0
	7.68	0.03	...	23.7
	7.68	0.10	...	24.0
12	8.20	0.025	...	14.9
	8.20	0.05	...	16.3
	8.19	0.10	...	18.2
13	7.60	0.03	...	31.7
	7.62	0.03	0.001	32.7
	7.60	0.03	0.01	35.7
	7.64	...	0.01	37.2
14[b]	7.33	0.03	0.05	55.4
	7.35	0.03	0.20	69.6
	7.35	...	0.20	71.0

[a] At 30° in 10% acetonitrile–water, μ = 0.5. [b] Approximate value of K_{app} for imidazole at pH 7.35 is 0.17 *M*.

(a) Very low concentrations of phosphate buffer cause major increases in aniline yield, over that produced in Tris buffer alone (experiments 1–3). (b) The effect of phosphate buffer does not depend on the presence of Tris. Similar increases are produced by phosphate in imidazole buffer (experiment 5) or by phosphate buffer alone (compare experiments 1, 2, and 6; see also Figure 1). (c) The effectiveness of a given concentration of phosphate in increasing aniline yield falls off with increasing pH. (d) The influence of added bicarbonate buffer or of bicarbonate buffer alone is generally similar to that of phosphate (experiments 7–9). (e) Although acetate buffer increases aniline yield also, the required concentration of acetate is much higher than that of phosphate or bicarbonate at comparable pH (compare experiments 2 and 10; also acetate and phosphate in Figure 2). (f) Variation in Tris buffer concentration has nearly no effect on aniline yield (experiments 11 and 12). (g) High concentrations of imidazole buffers (with or without Tris) lead to moderate increases in aniline produced.

Other substances examined for their ability to alter the course of iminolactone hydrolysis were the dicarboxylic acid[7] 1,1-cyclopentanediacetic acid (3,3-tetramethyleneglutaric acid), arsenate, monophenylphosphate, and *p*-nitrophenol. All but the last increased aniline yields, arsenate being particularly effective (Table III).

Table III. Buffer Effects on Aniline Yield from Iminolactone Hydrolysis[a]

Expt.	Buffer[b]	M	pH	Aniline, %
	Tris	0.03	7.27	39.4
15	1,1-Cyclopentane-diacetic acid	0.005	7.27	61.3
		0.01	7.27	68.8
		0.05	7.27	85.4
16	Arsenate	0.0005	7.27	65.6
		0.002	7.27	84.6
		0.01	7.26	93.9
17	Monophenyl-phosphate	. . .	7.22	39.4
		0.001	7.21	55.9
		0.01	7.23	87.7

[a] At 30° in 10% acetonitrile–water, $\mu = 0.5$. [b] All reaction mixtures contain 0.03 M Tris buffer.

The following empirical treatment was employed to evaluate quantitatively the ability of a given buffer to increase aniline yield (at fixed pH) above that value obtained in Tris buffer alone. Assume that interaction occurs between buffer and iminolactone (S) (or some species derived from iminolactone) and that the increase in aniline yield (ΔA) is proportional to the extent of complex formation.

$$\text{buffer} + \text{S} \rightleftharpoons \text{complex}$$

$$K_{\text{app}} = \frac{[\text{S}][\text{buffer}]}{[\text{complex}]}$$

$$[\text{S}] + [\text{complex}] = [\text{S}_\text{T}]$$

$$\frac{[\text{complex}]}{[\text{S}_\text{T}]} = \frac{[\text{buffer}]}{[\text{buffer}] + K_{\text{app}}}$$

A_B is the aniline yield at a given buffer concentration, A_0 is the aniline yield in the absence of added buffer, $\Delta A = A_\text{B} - A_0$. $\Delta A_{\text{max}} = \Delta A$ at infinite buffer concentration. If the increase in aniline yield is in fact proportional to the extent of (hypothetical) complex formation, then

$$\frac{\Delta A}{\Delta A_{\text{max}}} = \frac{[\text{complex}]}{[\text{S}_\text{T}]} = \frac{[\text{buffer}]}{[\text{buffer}] + K_{\text{app}}} \tag{3}$$

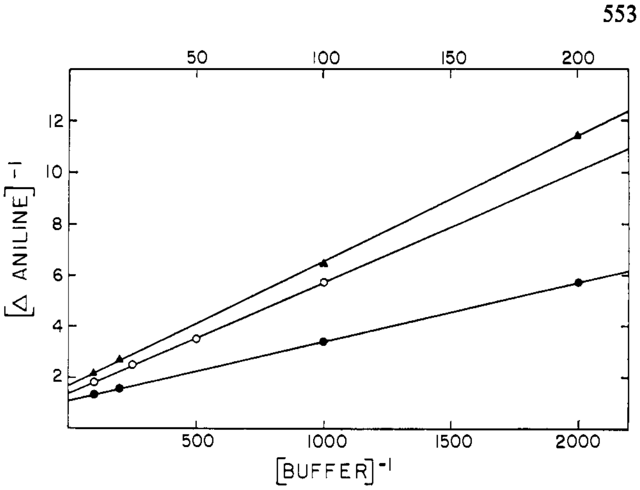

Figure 3. Double reciprocal plot of the increase in aniline yield (expressed as mole fraction) as a function of buffer concentration: ▲, acetate, pH 7.26 (abscissa, upper scale); ○, phosphate, pH 7.88 (abscissa, lower scale); ●, bicarbonate, pH 9.02 (abscissa, upper scale). All reaction mixtures contain 0.03 M Tris.

According to eq. 3, plots of $1/\Delta A$ *vs.* the reciprocal of buffer concentration should be linear, allowing evaluation of K_{app} from the slope and intercept (the intercept being equal to $1/\Delta A_{\text{max}}$). Excellent linear relationships were obtained from such treatment of data of the type shown in Figure 2. In all cases, it was found that, at constant pH and for a given buffer, $\Delta A_{\text{max}} + A_0 = 94\text{–}98\%$, indicating that essentially complete conversion of iminolactone to aniline would occur at all pH values studied (7–9.5) in the presence of sufficiently high buffer concentration.

Double reciprocal plots for three of the four experiments of Figure 2 are presented in Figure 3. The derived values of K_{app} are 0.003, 0.021, and 0.028 M, for phosphate, bicarbonate, and acetate, respectively. These values of K_{app} represent simply the concentration of the given buffer required to produce a half-maximal increase in aniline yield ($\Delta A_{\text{max}}/2$) at the given pH. It should be remembered that A_0, the aniline yield in Tris buffer alone, varies with pH (Figure 1), so that ΔA_{max} increases with increasing pH.

Table IV. Summary of Values of K_{app} Determined at pH 7.3

Buffer	pK	$K_{\text{app}} \times 10^3\ M$	K_{app} rel.
Acetate	4.76[a]	28	39
Monophenyl-phosphate[b]	5.88[c]	2.75	3.9
1,1-Cyclopentane-diacetic acid	6.70[a,d]	12	17
Phosphate	6.77[a,e]	0.69	1.0
Arsenate	6.98[e,f]	0.53	0.7
Imidazole	7.02[a]	*Ca.* 170	240
p-Nitrophenol	7.15[g]	>100	>145
Tris[h]	8.20[a]	>1000	>1450
Bicarbonate	10.13[a,e]	1.2	1.7

[a] This study. [b] K_{app} determined at pH 7.2. [c] pK_2 in H_2O at 26°, $\mu = 0.1$: J. D. Chanley and E. Feageson, *J. Am. Chem. Soc.*, **77**, 4002 (1955). [d] p$K_2 = 6.77$ in H_2O at 25°: G. Kortüm, W. Vogel, and K. Andrussow, "Dissociation Constants of Organic Acids in Aqueous Solution," Butterworth and Co. Ltd., London, 1961, p. 278. [e] pK_2. [f] S. Bruckenstein and I. M. Kolthoff, "Treatise on Analytical Chemistry," Part I, Vol. 1, I. M. Kolthoff and P. J. Elving, Ed., Interscience Publishers, Inc., New York, N. Y., 1959, p. 432. [g] In H_2O at 25°; *cf.* footnote *d*, p. 451. [h] Estimated lower limit for K_{app} at pH 7.7.

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(7) Preliminary experiments indicated that maleic acid at pH 7.3 was also reasonably effective.

Table V. Buffer Effects on Rates and Products of Iminolactone Hydrolysis[a]

pH	Buffer: Tris, M	Buffer: Other, M	$k_0 \times 10^3$,[b] min.$^{-1}$	Aniline, %
7.27	0.03	...	5.53 ± 0.18	39.5
7.22	0.03	Phosphate, 0.01	5.70	95.2
7.27	0.03	Arsenate, 0.01	5.54	93.9
7.28	0.03	Acetate, 0.10	5.59	82.6
8.48	0.03	...	1.90	11.4
8.56	0.03	Phosphate, 0.10	1.91	85.0
9.02	0.03	...	1.56	5.3
8.99	0.03	Bicarbonate, 0.10	1.57	83.0

[a] At 30° in 10% acetonitrile–water, $\mu = 0.5$. [b] Observed first-order rate constant for iminolactone disappearance.

The differing abilities of the several buffers of this study to increase aniline yield at pH 7.3 are indicated by the K_{app} values presented in Table IV. In this manner, the qualitative conclusions stated earlier find quantitative expression. Comment on mechanistic implications is deferred to a later section.

Detailed examination of the effect of pH on K_{app} was carried out for phosphate, bicarbonate, and acetate buffers (Figure 4). As noted above, the effectiveness of each buffer falls off rapidly with increasing pH, especially so in the case of acetate. While for phosphate and bicarbonate, the plot of log K_{app} *vs.* pH exhibits a slope of nearly 1, the corresponding slope for acetate is about 2.

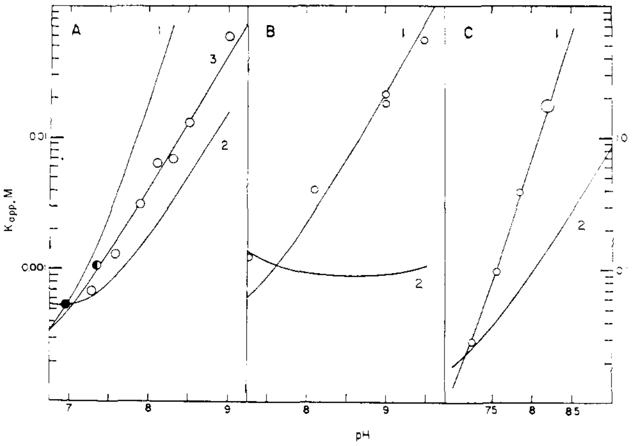

Figure 4. pH dependence of the effectiveness of buffers (given by K_{app}) in increasing the aniline yield from iminolactone hydrolysis. All reactions carried out in 0.03 M Tris, except where noted: (A) phosphate (left ordinate); ●, in 0.02 M imidazole; ◐, in 0.03 M imidazole; (B) bicarbonate (left ordinate); (C) acetate (right ordinate). Solid lines are theoretical curves computed as described in text: 1, reaction of conjugate acid of buffer with neutral carbinolamine; 2, reaction of conjugate base of buffer with neutral carbinolamine; 3, reaction of both buffer species with neutral carbinolamine.

It is known[3] that the hydrolysis of iminolactone I is subject to general base catalysis by phosphate dianion. The addition of 0.002 M phosphate buffer at pH 7.25 would be expected[8] to increase the rate of hydrolysis by approximately 1%. This concentration of phosphate buffer, however, is sufficient to increase aniline yield from 40 to 85% (Table I, experiment 1). It is shown in Table V that concentrations of phosphate, arsenate, acetate, or bicarbonate buffers capable of markedly increasing the extent of conversion of iminolactone to aniline have no measurable effect on the rate of disappearance of iminolactone. The effect of adding 0.10 M $NaHCO_3$ buffer to 0.03 M Tris buffer at pH 9.0 is particularly striking: while aniline yield changes from 5% (in Tris alone) to 83% (in Tris + $NaHCO_3$), the rates of iminolactone hydrolysis are identical within the limits of experimental error.

Attempts were made to demonstrate spectrally the accumulation of intermediate species during the transformation of iminolactone to a mixture of aniline (butyrolactone) and γ-hydroxybutyranilide. Hydrolysis was allowed to proceed at pH 7.3 in 0.03 M Tris buffer containing either phosphate (0.0007 M), 1,1-cyclopentanediacetic acid (0.012 M), or imidazole (0.17 M). The concentrations of the added buffers were those at which yields of 68–70% aniline were obtained in each case. The spectral course of the reaction is shown in Figure 5 for cyclopentanediacetic acid. The family of curves exhibits three well-defined isosbestic points at 222, 242, and 279 mμ. The spectrum of the reaction mixture at completion of hydrolysis ($t_{1/2}$ *ca.* 2 hr.) is essentially identical with that of a product mixture containing 70%

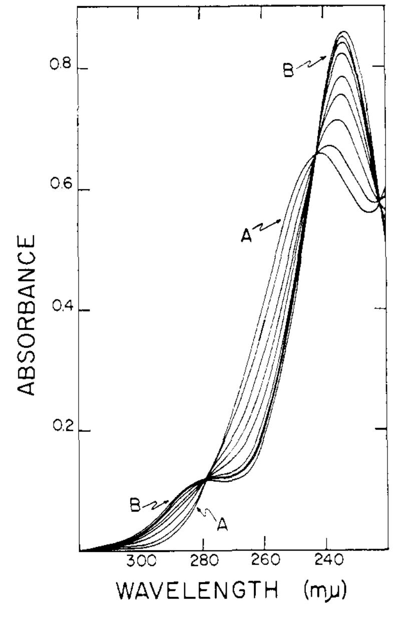

Figure 5. Hydrolysis of iminolactone (1.06 × 10^{-4} M) at pH 7.27 in 0.03 M Tris buffer containing 0.012 M 1,1-cyclopentanediacetic acid: (A) spectrum at zero time; (B) after 20 hr.

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[8] Calculated from the data shown in Table I and Figure 2 of ref. 3.

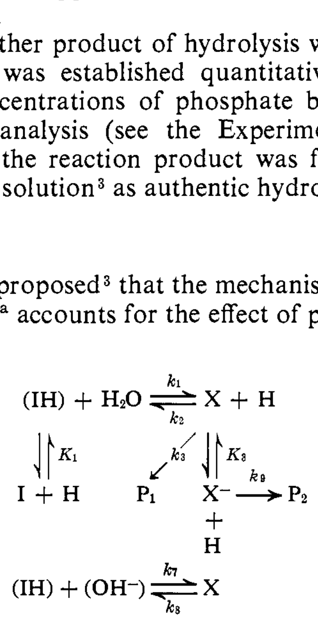
of aniline and 30% of γ-hydroxybutyranilide. Identical spectral families were obtained with phosphate or imidazole buffers.[9] Thus, no evidence could be found for the existence of intermediate species or of unexpected products; despite the considerable differences in buffer concentrations required, and however dissimilar the detailed reaction mechanisms may be, the over-all reaction course appears to be the same in the three cases studied.

That the other product of hydrolysis was γ-hydroxybutyranilide was established quantitatively at several different concentrations of phosphate buffer (pH 7.6) by spectral analysis (see the Experimental Section). In addition, the reaction product was found to be as labile in acid solution[3] as authentic hydroxyanilide.

Discussion

It has been proposed[3] that the mechanism summarized in Scheme I[10a] accounts for the effect of pH on rate and on product variation in the hydrolysis of N-phenyliminotetrahydrofuran. Some aspects of this formulation are as follows. Rate-determining addition of water (in weakly acidic solution) or of hydroxide ion (at pH >8) to protonated iminolactone leads to a neutral tetrahedral intermediate (X). Breakdown of this neutral carbinolamine yields predominantly aniline (and butyrolactone) while breakdown of its conjugate base (X^-) (see eq. 2) affords mainly γ-hydroxybutyranilide. At values of pH below 2, the rate-limiting step is decomposition of the tetrahedral intermediate rather than hydration of the iminolactone.[10b]

Scheme I



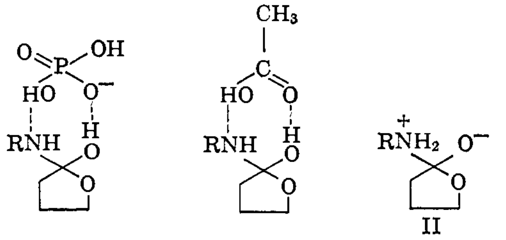
The additional facts which must be accomodated now by any comprehensive mechanism for iminolactone hydrolysis are as follows. (a) The nature of the products not only varies with pH, but also depends on the identity and concentration of the buffer at fixed pH. (b) Buffers of similar pK have widely differing abilities to alter the course of the reaction. (c) Under conditions where the addition of a given buffer results in a marked increase in aniline yield, the rate of iminolactone hydrolysis is unchanged.

The absence of a detectable rate alteration suggests that the reactive buffers affect the pathways of decomposition of one or more ionic species of the tetrahedral intermediate, rather than the rate-determining formation of the intermediates.

Increasing pH rapidly reduces the ability of buffers to increase aniline yield (Figure 4), suggesting the possibility that the buffer effect represents general acid–base catalysis. The fact that phosphate buffer (pK_2 = 6.77) is 240 times more effective than imidazole buffer (pK_2 = 7.02) appears to rule out classical general acid catalysis. The ratio of reactivity of imidazole and phosphate in nucleophilic or general acid–base catalysis of reactions at the acyl group is variable: imidazole is about 10^3 times as reactive as phosphate dianion in nucleophilic attack on p-nitrophenyl acetate[11] or acetyl phenyl phosphate,[12] but the two substances are of equal reactivity with Δ-thiolvalerolactone[13]; nearly identical reactivity is found for imidazole and phosphate dianion in the general base catalyzed hydrolysis of ethyl dichloroacetate[14] and for imidazolium ion and phosphate monoanion in general acid catalyzed semicarbazone formation.[15] In the present work, phosphate dianion is about twice as effective as imidazole in increasing the rate of hydration of iminolactone (presumably by general base catalysis[3]), but phosphate buffer is 240 times more effective than imidazole buffer in promoting the conversion of iminolactone to aniline. The significantly greater reactivity of phosphate observed in the latter process is unusual. Thus, both nucleophilic and classical acid–base catalysis seem unlikely on kinetic grounds.

A common feature of the buffers which exhibit the ability to increase aniline yield at very low buffer concentration is the presence of (potential) acidic and basic groups attached to a central atom. To account for the rather narrow structural specificity and the pH dependence of the buffer effect, it is proposed that effective catalysts are those substances which are able to promote a (concerted) cyclic proton shift in the neutral tetrahedral intermediate. The interactions envisaged are pictured in Scheme II for phosphate monoanion and acetic acid. Analogous structures may be written for bicarbonate, phosphate dianion, arsenate, and monophenyl phosphate. The essential result of such proton transfer is equivalent to a shift in the equilibrium between the uncharged neutral carbinolamine and its zwitterionic form II, required for cleavage of C–N bond and aniline formation. Equations 4 and 5 describe the interaction of *neutral* carbinolamine (X) with

Scheme II



II

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[9] The high end absorption of imidazole obscured the isosbestic point at 222 mμ.

[10] (a) I = iminolactone in free base form; (IH) = protonated iminolactone; X = neutral carbinolamine; (X^-) = anionic carbinolamine; P_1 = aniline + butyrolactone; P_2 = γ-hydroxybutyranilide. (b) As discussed earlier,[3] it is not possible to rule out conclusively an alternate mechanism, according to which iminolactone hydrolysis proceeds *via* the decomposition of a cationic carbinolamine in acid solution. Product variation would then be ascribed to the fact that the cationic intermediate yields aniline and the neutral intermediate is converted to hydroxyanilide. The quantitative arguments developed in the present study to account for the dependence of K_{app} on pH are in no way affected if cationic and neutral intermediates replace, respectively, neutral and anionic intermediates. Scheme II (this paper) would naturally no longer be applicable. Since it is difficult to envisage structurally how interaction of acetic acid or phosphate monoanion with cationic carbinolamine might enhance aniline formation, the favored mechanism is that advanced in Schemes I and II, and eq. 4 and 5.

[11] (a) T. C. Bruice and R. Lapinski, *J. Am. Chem. Soc.*, **80**, 2265 (1958); (b) W. P. Jencks and J. Carriuolo, *ibid.*, **82**, 1778 (1960).

[12] G. DiSabato and W. P. Jencks, *ibid.*, **83**, 4393 (1961).

[13] T. C. Bruice, J. J. Bruno, and W.-S. Chou, *ibid.*, **85**, 1659 (1963).

[14] W. P. Jencks and J. Carriuolo, *ibid.*, **83**, 1743 (1961).

[15] E. H. Cordes and W. P. Jencks, *ibid.*, **84**, 4319 (1962).

the *acidic* form (BH) of a buffer (acid dissociation constant K_2) to form a complex (X′) whose decomposition yields aniline (and butyrolactone). The dissociation constant of the complex is denoted by K_4.

$$\mathrm{X} + \mathrm{BH} \overset{K_4}{\rightleftharpoons} \mathrm{X'} \tag{4}$$

$$\mathrm{X'} \xrightarrow{k_{13}} \mathrm{P_1} \tag{5}$$

Assumption of the mechanism summarized in Scheme I and eq. 4 and 5 leads to the following conclusions (see the Appendix). (a) At fixed pH, the plot of $1/\Delta A$ *vs.* $1/B_0$ is linear, in agreement with experiment. (b) The dependence of K_{app} on pH is given by eq. 6, where

$$K_{\mathrm{app}} = \frac{(K_4k_3/k_{13})([\mathrm{H}] + K')([\mathrm{H}] + K_2)}{[\mathrm{H}]^2} \tag{6}$$

$K' = K_3k_9/k_3$; since K_2 is known and K' is the apparent dissociation constant defined[3] by the product variation with pH (Figure 1), only one numerical assumption (K_4k_3/k_{13}) need be made to calculate the variation of K_{app} with pH for a given buffer. Curves 1A, 1B, and 1C (Figure 4) are calculated from eq. 6 for phosphate monoanion, bicarbonate ion, and acetic acid, using values for K_4k_3/k_{13} of 1.14×10^{-4}, 2.39×10^{-4}, and 4.08×10^{-5} M, respectively, and $K' = 8.5 \times 10^{-8}$ M ($pK' = 7.07$).

Assumption that the buffer effect results from interaction of the *basic* buffer species with *neutral* carbinolamine (or the kinetically equivalent interaction of *acidic* buffer species with *anionic* carbinolamine) leads to eq. 7 for the dependence of K_{app} on pH (see Appendix).

$$K_{\mathrm{app}} = \frac{(K_4k_3/k_{13})([\mathrm{H}] + K')([\mathrm{H}] + K_2)}{[\mathrm{H}]K_2} \tag{7}$$

Curves 2A, 2B, and 2C (Figure 4) are calculated from eq. 7 using values for K_4k_3/k_{13} of 1.82×10^{-4}, 7.36×10^{-7}, and 0.01 M, respectively.

With acetate buffer, excellent agreement is obtained between observation and theory for the mechanism involving interaction of neutral carbinolamine with the buffer conjugate acid. The results shown for bicarbonate–carbonate buffer leave little doubt that the same mechanism is operative, although agreement is less satisfactory, possibly owing to dehydration of bicarbonate at the lower pH values. With phosphate buffer, acceptable correlation of data with theory could be obtained only by assuming that *both* phosphate monoanion and dianion were effective, with the ratio of reactivity of dianion to monoanion (e) being 0.3. Curve 3A is calculated from eq. 11, with $K_4k_3/k_{13} = 1.5 \times 10^{-4}$ M and $e = 0.3$. Since both ionic species of phosphate contain the requisite acidic and basic functions for concerted proton transfer, this conclusion might have been anticipated.

The rate constant k_3 being (presumably) unaffected by buffer effects, the relative values of the reciprocals of the term K_4k_3/k_{13} are an index of the effectiveness of the buffer catalysis. Relative to $H_2PO_4^-$ (1.0), these values are 0.3 for HPO_4^{2-}, 0.6 for HCO_3^-, and 3.7 for CH_3COOH. Thus, acetic acid is intrinsically the best catalyst encountered in this study, its moderate observed effect being due to the very low concentrations of acetic acid present above pH 7.

Analogous mechanisms have been advanced to explain carboxylic acid catalysis of the cyclization of glutamic acid esters to derivatives of 2-pyrrolidone-5-carboxylic acid[16] and the specific catalytic effects of carboxylic, sulfuric, and phosphoric acids on the rearrangement of N,N′-diacylhydrazines.[17] Unlike the present work, however, these reactions were carried out in nonaqueous media[18] and effective catalyst concentrations were in the range of 0.1–2 M. Unexpected catalytic effects of phosphate monoanion and bicarbonate ion on the reaction of urea with formaldehyde in water have been reported[19] in a preliminary form. It is well established[20] that the conversion of glutamine to pyrrolidonecarboxylic acid in water is accelerated by high concentrations of phosphate, arsenate, and bicarbonate ions. Insufficient information on this system is available to permit assignment of mechanism; plausible hypotheses include concerted proton transfer or, as previously suggested,[21] simple general catalytic effects.

The effect of high concentrations of imidazole on iminolactone hydrolysis has not been discussed. It is possible that the breakdown of the carbinolamine intermediates is also influenced by classical general acid–base catalysis. Since the rate of iminolactone disappearance is increased[3] at the imidazole concentrations employed, the imidazole effect on aniline yield is not easily interpreted.

Although cyclic proton transfer mechanisms have been proposed frequently, they have seldom been supported by compelling evidence, especially for reactions in aqueous solution.[22] Clearly, the results described in this paper do not rigorously exclude other mechanistic formulations; at this time, however, we are unable to account satisfactorily for the present data in terms of alternative hypotheses. Whatever the eventual detailed interpretation of these observations may be, it is obviously of interest to define the extent to which other reactions of acyl derivatives are subject to similar catalytic influences.

Experimental Section

Materials. 2-Phenyliminotetrahydrofuran was prepared in 67% yield as previously described.[3] Imidazole (Eastman Kodak Co.) was recrystallized from acetone–petroleum ether (b.p. 30–60°). 1,1-Cyclopentanediacetic acid (Aldrich Chemical Co.) had m.p. 175–177°. Monophenylphosphate, prepared by alkaline hydrolysis of phenyl phosphorodichloridate, melted at 98–99° (lit.[23] 99.5°) after recrystallization from benzene. Analysis[24] for inorganic phosphate indicated the presence of <0.2% (w./w.) of phosphoric acid. Buffers and inorganic salts were of reagent grade and were used without further purification. Acetonitrile was treated with

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(16) A. J. Hubert, R. Buyle, and B. Hargitay, *Helv. Chim. Acta*, **46**, 1429 (1963).

(17) M. Brenner and W. Hofer, *ibid.*, **44**, 1794 (1961); M. Brenner in "Peptides. Proceedings of the Fifth European Symposium, Oxford, September, 1962," G. T. Young, Ed., The McMillan Co., New York, N. Y., 1963, p. 126; W. Hofer and M. Brenner, *Helv. Chim. Acta*, **47**, 1625 (1964).

(18) Weak catalysis of the diacylhydrazine rearrangement by aqueous carbonate or bicarbonate solution has also been described.[17]

(19) B. Glutz and H. Zollinger, *Angew. Chem. Intern. Ed. Engl.*, **4**, 440 (1965).

(20) P. B. Hamilton, *J. Biol. Chem.*, **158**, 375 (1945); J. B. Gilbert, V. E. Price, and J. P. Greenstein, *ibid.*, **180**, 209 (1949); A. Meister, *ibid.*, **210**, 17 (1954).

(21) R. B. Martin, A. Parcell, and R. I. Hedrick, *J. Am. Chem. Soc.*, **86**, 2406 (1964).

(22) (a) *Cf.* ref. 4a, p. 95; (b) W. P. Jencks, *Ann. Rev. Biochem.*, **32**, 639 (1963).

(23) J. M. A. Hoeflake, *Rec. Trav. Chim.*, **36**, 24 (1916).

(24) C. H. Fiske and Y. Subbarow, *J. Biol. Chem.*, **66**, 375 (1925).

calcium hydride and distilled from P_2O_5, according to method D of Coetzee, *et al.*[25]

Product Analysis. All hydrolysis reactions were carried out at 30° in 10% acetonitrile-water (v./v.), at ionic strength 0.50, maintained with added KCl. Once the influence of bicarbonate ions on the products of iminolactone hydrolysis was recognized, care was taken to use boiled distilled water and to protect reagent solutions and reaction mixtures from atmospheric CO_2. Scatter in the experimental data was significantly reduced by these precautions.

A. **Aniline** was estimated colorimetrically by means of a modification[3] of the Bratton-Marshall procedure. Color development reached 95% of completion in 1 hr.; generally, absorbance measurements were made after 3 hr., the color being stable for at least 24 hr. No interference with the aniline assay was caused by the presence of 0.2 *M* imidazole or 0.05 *M* *p*-nitrophenol in the reaction mixture.

The following procedure was employed for the determination of buffer effect on aniline yield (K_{app}) at fixed pH. The hydrolysis of the iminolactone (*ca.* 10^{-4} *M*) was carried out in five or six parallel reaction mixtures containing increasing concentrations of the buffer under scrutiny (*e.g.*, phosphate), as well as (usually) 0.03 *M* Tris buffer. The extent of conversion of iminolactone to aniline was measured after 7–10 half-lives of hydrolysis. K_{app} was calculated from double reciprocal plots of the increase in aniline yield (ΔA) *vs.* buffer concentration (see Results). In the case of monophenylphosphate, assay[24] for inorganic phosphate indicated that no detectable hydrolysis of the phosphate ester had occurred during the time required for iminolactone hydrolysis.

B. **Spectral Analysis.** Iminolactone hydrolysis was carried out in the presence of 0.03 *M* Tris buffer containing either phosphate (0.0007 *M*), 1,1-cyclopentanediacetic acid (0.012 *M*), or imidazole (0.17 *M*) buffer (final pH 7.3 in all cases). Reaction was allowed to proceed in a 4-ml. quartz cuvette inserted in the water-jacketed (30°) cell holder of a Bausch and Lomb Spectronic 505 spectrophotometer, and the spectral region of 220–300 mμ was scanned periodically during 20 hr. On completion of reaction, aliquots of each solution were analyzed for aniline.

Quantitative identification of γ-hydroxybutyranilide as the alternate product of hydrolysis was accomplished as follows. Hydrolysis of iminolactone was carried out at pH 7.6 in reaction mixtures containing 0.03 *M* Tris and, respectively, 0, 0.0005, 0.002, and 0.006 *M* phosphate buffer. Aniline assay performed after 42 hr. revealed the presence of 22.6, 43.1, 67.5, and 83.1% aniline, respectively. When each reaction mixture was acidified to pH 1.7 with 2 *M* phosphate buffer, complete ultraviolet spectra were identical with that of γ-hydroxybutyranilide (anilinium ion exhibiting negligible absorption) and the absorbance values at 239 mμ (ε 10,550) indicated the formation of hydroxyanilide in yields of 77.4, 59.3, 34, and 12.1%, respectively. The identity of the hydroxyanilide was further verified by the following experiment. An aliquot of the reaction mixture containing 0.0005 *M* phosphate buffer was acidified to a final concentration of 1.2 *N* HCl and the rate of disappearance of the product anilide was measured at 30° by the absorbance decrease at 239 mμ. The resulting rate constant (4.33 × 10^{-3} min.$^{-1}$) was identical with that for the hydrolysis of authentic anilide under the same conditions.

Kinetic measurements of iminolactone hydrolysis were performed using the apparatus described.[3] Iminolactone concentration was 1-2 × 10^{-4} *M* and wavelengths employed were 250 or 260 mμ (for reactions producing high aniline yields) and 236–243 mμ (for reactions producing low aniline yields).

p*K* Determinations. pK_a values at 30° in 10% acetonitrile-water for several of the buffers of this study were measured potentiometrically as described[3] earlier and are listed in Table IV.

Appendix

Dependence of Product Composition on Buffer Concentration. Variation of K_{app} with pH. Derivation of Eq. 6 and 7. In Tris (or other unreactive buffer), the fraction of aniline in the total product is given[3] by

$$A_0 = \frac{[H]}{[H] + (K_3k_9)/k_3} = \frac{[H]}{[H] + K'}$$

Consider an additional reaction path involving interaction of X with a reactive buffer B_0 (reactive ionic species unspecified)

$$X + B_0 \overset{K_4}{\rightleftharpoons} X' \qquad (7a)$$

$$X' \xrightarrow{k_{13}} P_1 \qquad (7b)$$

According to Scheme I, eq. 7a and 7b, and the relationship $[X][B_0]/[X'] = K_4$, the rate of formation of aniline is given by

$$\frac{d[P_1]}{dt} = k_3[X] + k_{13}[X'] = [X](k_3 + (k_{13}[B_0])/K_4) = k_3'''[X] \qquad (8)$$

Following the method previously[3] outlined, the fraction of aniline in the total product is given by

$$A_B = \frac{[H]}{[H] + (K_3k_9)/k_3'''}$$

$$\Delta A = A_B - A_0 = \left(\frac{[H]}{[H] + (K_3k_9)/k_3'''}\right) - \left(\frac{[H]}{[H] + (K_3k_9)/k_3}\right) \qquad (9)$$

From eq. 9, the definition of k_3''' (eq. 8) and the expression $K' = K_3k_9/k_3$

$$\frac{1}{\Delta A} = \frac{1}{[B_0]}\left(\frac{(K_4k_3/k_{13})([H] + K')^2}{[H]K'}\right) + \left(\frac{[H] + K'}{K'}\right)$$

From the linear plot of $1/\Delta A$ *vs.* $1/[B_0]$

$$\frac{\text{slope}}{\text{intercept}} = K_{app} = \frac{(K_4k_3/k_{13})([H] + K')}{[H]} \qquad (10)$$

A. Reaction with Conjugate Acid of Buffer. By means of the relationships $[B][H]/[BH] = K_2$ and $[B] + [BH] = [B_0]$, and substituting [BH] for $[B_0]$ in eq. 8, it may be shown that for interaction of [X] with [BH] only (Scheme I and eq. 4 and 5)

$$\frac{1}{\Delta A} = \frac{1}{[B_0]}\left(\frac{(K_4k_3/k_{13})([H] + K')^2([H] + K_2)}{[H]^2K'}\right) + \left(\frac{[H] + K'}{K'}\right)$$

$$\frac{\text{slope}}{\text{intercept}} = K_{app} = \frac{(K_4k_3/k_{13})([H] + K')([H] + K_2)}{[H]^2} \qquad (6)$$

B. Reaction with Conjugate Base of Buffer. For interaction of X with species B only, the dependence of ΔA on B_0 is given by

$$\frac{1}{\Delta A} = \frac{1}{B_0}\left(\frac{(K_4k_3/k_{13})([H] + K')^2([H] + K_2)}{[H]K'K_2}\right) + \left(\frac{[H] + K'}{K'}\right)$$

$$\frac{\text{slope}}{\text{intercept}} = K_{app} = \frac{(K_4k_3/k_{13})([H] + K')([H] + K_2)}{[H]K_2} \qquad (7)$$

C. Reaction with Both Species of Buffer. Consider two reaction paths involving interaction of X with buffer components BH and B

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[25] J. F. Coetzee, G. P. Cunningham, D. K. McGuire, and G. R. Padmanabhan, *Anal. Chem.*, **34**, 1139 (1962).

$$X + BH \overset{K_4}{\rightleftharpoons} X' \qquad X + B \overset{K_5}{\rightleftharpoons} X''$$

$$X' \xrightarrow{k_{13}} P_1 \qquad X'' \xrightarrow{k_{14}} P_1$$

Assuming that the ratio of the reactivity of species B to that of species BH is given by

$$e = \frac{k_{14}/K_5}{k_{13}/K_4}$$

it may be shown that

$$K_{\text{app}} = \frac{(K_4k_3/k_{13})([H] + K')([H] + K_2)}{[H]([H] + eK_2)} \qquad (11)$$

When $e = 0$ (*i.e.*, species B is unreactive), eq. 11 reduces to eq. 6. When $e = 1$ (*i.e.*, both buffer species equally reactive), eq. 11 reduces to eq. 10.

Acknowledgment. We are grateful to the National Institutes of Health for financial support (Grant No. AM-04288) and to Dr. Josef Rudinger for helpful discussion. During this investigation, B. A. C. was supported by a training grant (2G-53) of the U. S. Public Health Service (1963–1964) and was a Predoctoral Fellow of the National Institute of General Medical Sciences, U. S. Public Health Service (1964–1965).

Reaction of Acyclic α-Halo Sulfones with Base.[1a] Stereochemistry of the Intermediate Episulfones

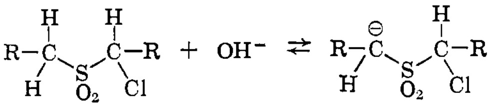
Norman P. Neureiter[1b]

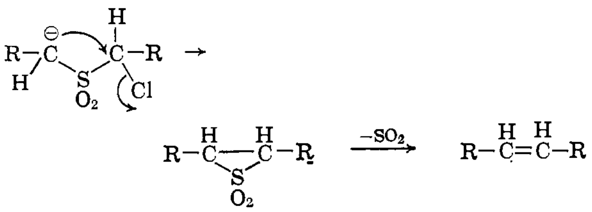
Contribution from the Esso Research and Engineering Company, Baytown Research and Development Division, Baytown, Texas. *Received July 19, 1965*

Abstract: The reaction of α-halo sulfones with base to give olefins has been investigated with a variety of acyclic sulfones and a variety of bases. The *cis/trans* ratios of the product olefins have permitted conclusions to be drawn about the reaction mechanism. The internal displacement of halide from the halo sulfone anion appears to favor formation of the *cis* episulfones which then give *cis* olefins. The magnitude and nature of this "*cis* effect" is briefly discussed. The postulated intermediates, *cis*- and *trans*-2-butene episulfones, have been synthesized by an independent route and their reactions have been examined.

The intriguing transformation of α-halo sulfones into olefins upon treatment with dilute alkali, first reported in 1940 by Ramberg and Bäcklund,[2] has been of both synthetic and mechanistic interest. Although the reaction has not found broad use in olefin synthesis, there appeared recently two reports of several practical synthetic schemes involving α-halo sulfones.[3,4] A salient feature of the reaction is its general applicability. With very few exceptions, notably in systems leading to small ring olefins,[5] the reaction is general for molecules containing the structural elements of a sulfonyl group, an α-halogen, and at least one α′-hydrogen atom. The double bond in the product cleanly and unequivocally supplants the sulfonyl group in the starting material.

A mechanistic study by Bordwell and Cooper[6] was interpreted to be consistent with the sequence shown in eq. 1 and 2. The reaction was first order in both hydroxide ion and in sulfone. The data pointed to rapid equilibrium between the sulfone and its anion, rate-controlling elimination of halide from the anion, and rapid loss of sulfur dioxide from the episulfone intermediate, either thermally or with participation of hydroxide ion.[7]


(1)


(2)

Ramberg and Bäcklund[2] had concluded from simple physical data that the products from α-bromoethyl ethyl sulfone and ethyl α-bromopropyl sulfone were mainly *cis*-2-butene and *cis*-2-pentene, respectively. Bordwell and Cooper, on the other hand, recovered a 94% yield of *trans*-stilbene from the reaction of α-chlorobenzyl benzyl sulfone.[6] The stereochemistry of the reaction required clarification. At this point the present work was begun.

Stereochemical Results. When α-chloroethyl ethyl sulfone was gently refluxed with 2 *N* potassium hydroxide solution, the sole olefinic product (76% yield)

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[1] (a) These results were communicated in preliminary form by N. P. Neureiter and F. G. Bordwell, *J. Am. Chem. Soc.*, **85**, 1209 (1963). (b) c/o American Embassy, Bonn/Bad Godesberg, Germany. Reprints available from Esso Research and Engineering. Company.

[2] (a) L. Ramberg and B. Bäcklund, *Arkiv Kemi, Mineral. Geol.*, A13, No. 27 (1940); (b) see also B. Bäcklund, Thesis, Uppsala, 1945.

[3] N. P. Neureiter, *J. Org. Chem.*, 30, 1313 (1965).

[4] L. A. Paquette, *J. Am. Chem. Soc.*, **86**, 4383 (1964).

[5] See, for example, the unsuccessful attempt of I. Mischon to prepare cyclobutene through this reaction, Ph.D. Thesis, Technische Hochschule zu Karlsruhe, Germany, 1955.

[6] F. G. Bordwell and G. D. Cooper, *J. Am. Chem. Soc.*, 73, 5187 (1951).

[7] There recently have appeared two papers on the mechanism of this reaction by L. A. Paquette. His evidence also supports the internal displacement reaction and formation of the episulfone intermediate: *ibid.*, **86**, 4085 (1964). He has also presented evidence against the possibility of a carbene mechanism in that instance: *ibid.*, **86**, 4089 (1964).