that each mode of tetrahedral intermediate–catalyst interaction found in imidate hydrolysis will have its quantitative counterpart in the related acyl transfer reactions. Use of this reasoning led to the verifiable prediction that the alcoholysis of 4-hydroxybutyranilide should be subject to bifunctional catalysis by certain general acid–base catalysts.[3,4] Finally, we have omitted from consideration the most general and at the same time most complex situation, that involving simultaneously three tetrahedral intermediates, TH_2^+, TH, and T^-, each giving rise to different products on breakdown and each subject to particular catalytic influences. Clearly, a thorough analysis of the general case should provide the framework for the description of many acyl transfer reactions.

Conclusions

Two qualitative observations derived from the study of the products of hydrolysis of imidate esters have provided strong support for the existence of tetrahedral intermediates in such reactions: (1) the effect of pH variation on the nature of the products of hydrolysis is not related to the effect of pH on the over-all reaction rate; (2) at constant pH, catalysts alter the yields of hydrolysis products without affecting the over-all rate of hydrolysis. In the present paper, it has been shown that a number of additional quantitative conclusions may be stated concerning the effects of pH and general acid–base catalysts on the rates and mechanisms of certain acyl transfer reactions. It appears that many aspects of the chemistry of the elusive addition intermediates formed at the acyl carbonyl group will be amenable to study by these indirect methods.

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The Acid-Catalyzed Hydrolysis of a Series of Phosphoramidates[1]

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Contribution from the Southeast Water Laboratory, Athens, Georgia 30601, and the Copolymer Rubber and Chemical Corporation, Baton Rouge, Louisiana 70821. *Received August 14, 1967*

Abstract: The acid-catalyzed hydrolysis of a series of six 2,4-dichlorophenyl methyl N-alkylphosphoramidates (I) has been studied. Most of the reactions were carried out in 25% dioxane in water (v/v) in the presence of 0.19 *M* HCl, with the concentration of phosphoramidate being 0.002 *M*. The reactions were followed kinetically by ultraviolet spectrophotometry and were found to be first order in phosphoramidate. Product determination experiments showed that the hydrolysis involved P–N bond cleavage with negligible liberation of 2,4-dichlorophenol or alcohol. The product of acid hydrolysis is thus the appropriate alkylamine salt of 2,4-dichlorophenyl methyl phosphoric acid (II). Activation energies and entropies were calculated, the highly negative value of the latter showing the reaction to be bimolecular. The hydrolysis is first order in hydronium ion as shown by pH dependence studies. The phosphoramidates hydrolyzed faster by a factor of 2 in D_2O than in H_2O, indicating a fast prior protonation of the substrate. A small increase in the rate of hydrolysis of the methyl phosphoramidate was observed with increasing percentage of water in the solvent at the same pH. Linear free-energy relationships were used to determine the effect of the N-alkyl substituent on the rate of hydrolysis. It was concluded that both steric and polar substituent effects operate, with an increase in bulk of the substituent hindering the reaction slightly while electron withdrawal aids the reaction. Possible mechanisms are proposed for the acid hydrolysis of the phosphoramidate diesters.

Phosphoramidates are important as pesticides and several of them, including schradan (octamethylpyrophosphoramide), Ruelene (4-*t*-butyl-2-chlorophenyl methyl N-methylphosphoramidate), and Zytron (2,4-dichlorophenyl methyl N-isopropylphosphoramidothiolate), are now on the market. There is evidence that diesters of phosphoramidic acid should act as phosphorylating agents in nucleotide synthesis,[3] and these compounds have been found useful as chemical reagents for the synthesis of unsaturated organic nitrogen compounds.[4]

Phosphoramidates are esters of phosphoramidic acid, $(HO)_2P(O)NH_2$. Much of the chemical interest in this class of compounds is due to the presence of the phosphorus-to-nitrogen bond and its susceptibility to cleavage in aqueous acidic solution. (Hydrolysis in alkaline solution leads principally to P–O bond cleavage.[5]) Although there is recorded a relatively large

(1) (a) Presented in part at the 18th Southeastern Regional Meeting of the American Chemical Society, Louisville, Ky., Oct 1966. (b) Mention of products and manufacturers in this publication is for identification only and does not imply endorsement by the Federal Water Pollution Control Administration or the United States Department of the Interior.

(2) (a) To whom inquiries should be addressed: Federal Water Pollution Control Administration, Southeast Water Laboratory, Athens, Ga. 30601. (b) This paper is taken from the Ph.D. dissertation of A. W. Garrison, presented to Emory University, Atlanta, Ga. Part of the work was accomplished at Emory University and part at the Southeast Water Laboratory. (c) U. S. Department of Health, Education, and Welfare trainee, 1963–1965.

(3) A. R. Todd, *Proc. Chem. Soc.*, 199 (1962).

(4) W. S. Wadsworth and W. D. Emmons, *J. Org. Chem.*, **29**, 2816 (1964).

(5) J. E. Berger and E. Wittner, *J. Phys. Chem.*, 70, 1025 (1966).

Table I. Rate Constants (Min^{-1}) for the Hydrolysis of 0.002 *M* 2,4-Dichlorophenyl Methyl N-Alkylphosphoramidates in Aqueous Solutions of 0.19 *M* HCl and 25% Dioxane

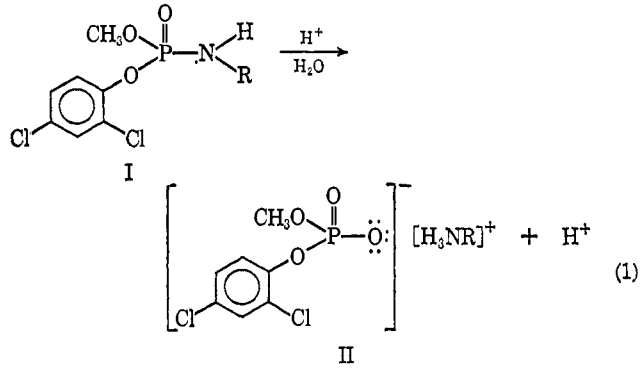
Temp, °C	Methyl	Ethyl	*n*-Butyl	Isopropyl	*sec*-Butyl	*t*-Butyl
15	0.0232 ± 0.0006					
25	0.0455 ± 0.0003	0.0216 ± 0.0003	0.0130 ± 0.0003			0.0008[a]
35	0.0759 ± 0.0018	0.0372 ± 0.0003	0.0225 ± 0.0006	0.00695 ± 0.00010	0.00310 ± 0.00003	
45	0.141 ± 0.004	0.0617 ± 0.0002	0.0374 ± 0.0004	0.0121 ± 0.0001	0.00577 ± 0.00004	
55		0.0975 ± 0.0027	0.0630 ± 0.0011	0.0226 ± 0.0021	0.0111 ± 0.0005	
65				0.0378 ± 0.0007	0.0188 ± 0.0005	0.0074 ± 0.0015
70						0.00865 ± 0.00015
75						0.0113 ± 0.0003

[a] Initial reaction rate only. Not used in determination of activation energy.

amount of work on the cleavage of the P–N bond under acid catalysis in such compounds as phosphoramidic acid and N-substituted phosphoramidic acids and in such commercially important pesticides as schradan, $(Me_2N)_2P(O)\cdot OP(O)(NMe_2)_2$, and tabun, $(Me_2N)(EtO)$-$PO\cdot CN$, only two publications deal with the kinetics of acid hydrolysis of phosphoramidates.[6,7] These papers report a study of the acid-catalyzed hydrolysis of four dimethyl N,N-dialkylphosphoramidates. No structure–reactivity correlation is made; no solvent effects are studied; and no mechanistic proposals are made in these reports. Furthermore, the compounds concerned are dimethyl esters and do not involve an aromatic substituent.

The present work is an attempt to determine the mechanism of hydrolysis of phosphoramidate diesters in acid solution. It is essentially a study of the kinetics of this reaction, using a series of 2,4-dichlorophenyl methyl N-alkylphosphoramidates (I). This particular series was chosen because of its resemblance to several organophosphorus compounds of pesticidal importance which also contain the chlorinated phenyl moiety. It was decided to study enough members of the series to determine whether a valid structure–reactivity correlation exists, while at the same time restricting the changes in structure to variations in the alkyl group.

The over-all reaction was assumed to be as follows.



R = methyl, ethyl, isopropyl, butyl, *sec*-butyl, or *t*-butyl

Results

The progress of each reaction was followed by measuring the increase in ultraviolet absorbance at 275.5 mμ with time. This increase was due to the formation of an alkylamine salt of 2,4-dichlorophenyl methyl phosphoric acid (II), as shown by the spectral characteristics of a pure sample of one of these compounds. The methylamine salt II (R = methyl) had an absorptivity of 905 (in an aqueous solution of 25% dioxane and 0.19 *M* HCl) at 276.5 mμ while the reactant, the corresponding methylphosphoramidate I (R = methyl), had an absorptivity of 725 at 275 mμ. The kinetics of the reactions were followed at 275.5 mμ since this wavelength gave the greatest change in absorbance during the reaction. Further proof that II is the product of the phosphoramidate hydrolysis is given below.

Plots of time *vs.* an absorbance function according to the method of Guggenheim[8] gave straight lines for all six phosphoramidates of the series under all conditions studied. The hydrolyses reactions are thus pseudo first order in phosphoramidate, being that an excess of water and of hydrochloric acid was always present. One of the kinetic plots for the methyl derivative is shown in Figure 1. Table I gives the rate constants (k), calcu-

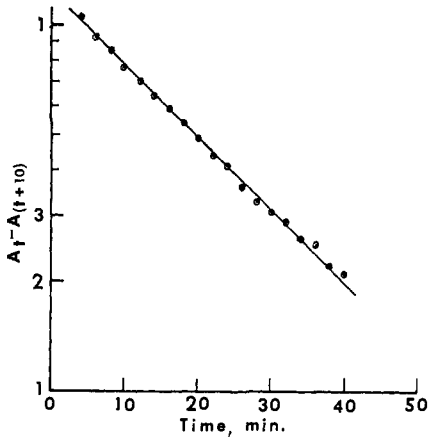


Figure 1. Kinetic plot for the acid-catalyzed hydrolysis of 0.002 *M* 2,4-dichlorophenyl methyl N-methylphosphoramidate in aqueous solution of 0.19 *N* HCl and 25% dioxane at 25°: A = phosphoramidate absorbance.

lated from the slopes of the kinetic plots, for each phosphoramidate at each temperature involved. These reactions were carried out in aqueous solutions of 0.19 *M* HCl containing approximately 25% dioxane to increase solubility of the phosphoramidates, which were present in 0.002 *M* concentration.

A. Product Determination. As shown in reaction 1, the product of the hydrolysis was assumed to be the alkylamine salt of 2,4-dichlorophenyl methyl phos-

(6) M. Sélim and T. N. Thanh, *Compt. Rend.*, **250**, 2377 (1960).
(7) T. N. Thanh and M. Sélim, *ibid.*, **250**, 2724 (1960).
(8) A. A. Frost and R. G. Pearson, "Kinetics and Mechanism," 2nd ed, John Wiley and Sons, Inc., New York, N. Y., 1961, p 49.

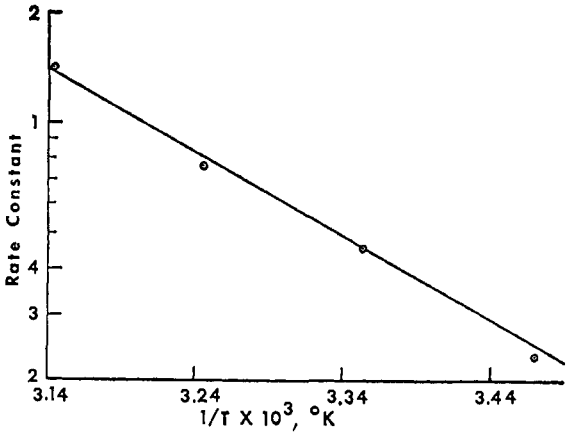


Figure 2. Arrhenius plot for the acid-catalyzed hydrolysis of 2,4-dichlorophenyl methyl N-methylphosphoramidate.

phoric acid. This was shown to be true by chemical analysis of the acidic reaction solutions after they were allowed to stand a sufficient time for complete hydrolysis. The solutions were made basic and the liberated amines were removed by distillation, collected, and titrated with standard acid. Table II summarizes

Table II. Determination of Amines Produced by Acid-Catalyzed Hydrolysis of N-Substituted Phosphoramidates (I)

Substituent	No. of samples analyzed	Amine,[a] mol	Calcd % completion
Methyl	1	0.00021	100
Ethyl	3	0.00019	95
Isopropyl	4	0.00018	90
Butyl	1	0.00017	85
sec-Butyl	2	0.00018	90
t-Butyl	1	0.00019	95

[a] Average mole of amine collected from 10 ml of the corresponding 0.002 *M* phosphoramidate solution after hydrolysis.

the analytical results and gives the calculated per cent of completion of each hydrolysis reaction based on the amine determination.

Spectrophotometric analysis also gave evidence of the character of the reaction product. This evidence, which is described in the Experimental Section, also serves to establish the validity of the spectrophotometric kinetic technique.

A 0.002 *M* solution of the pure methylamine salt of 2,4-dichlorophenyl methyl phosphoric acid (II, R = methyl),[9] the reaction product of the methyl phosphoramidate hydrolysis, showed no change in absorption at 275.5 mμ in aqueous acid solution for 45 min at 25°. This shows that no measurable consecutive reaction took place after the hydrolysis of the phosphoramidate P–N bond.

It is conceivable that hydrolysis of the phosphoramidates could have produced 2,4-dichlorophenol by P–O bond cleavage. An analytical method was developed to determine the amount of 2,4-dichlorophenol in the solutions of the hydrolyzed phosphoramidates. On the basis of these analyses, the amount of 2,4-dichlorophenol in the phosphoramidate solutions after hydrolysis was insignificant.

It is also possible that bond cleavage could have occurred at the C–N bond to produce the corresponding alcohol. However, analysis by nmr showed no methanol or 2-propanol in solution after hydrolysis of the methyl- or of the isopropylphosphoramidate, and it was assumed that no significant C–N bond cleavage occurred during hydrolysis of any of the derivatives studied.

B. Effect of Temperature on Reaction Rate. Arrhenius plots for all of the phosphoramidates, using the reaction rate constants and corresponding temperatures given in Table I, were linear. An example is given in Figure 2. The activation energies for the hydrolysis of each derivative, calculated from the Arrhenius plot slopes, are presented in Table III. The

Table III. Activation Parameters for the Acid-Catalyzed Hydrolysis of 2,4-Dichlorophenyl Methyl N-Alkylphosphoramidates (I)

N-Alkyl substituent	E_a, kcal/mol	$\Delta S^{\pm}$, cal/(mol deg)	$\Delta F^{\pm}$, kcal/mol	$\Delta H^{\pm}$, kcal/mol
Methyl	10.4	−31	19	9.81
Ethyl	10.1	−34	20	9.51
Butyl	10.1	−35	20	9.51
Isopropyl	12.1	−30	20	11.5
sec-Butyl	12.5	−31	21	11.9
t-Butyl	13.2	−31	22	12.6

corresponding entropies of activation, calculated according to the transition-state theory, and enthalpies and free energies of activation, calculated from standard thermodynamic relationships, are also given in Table III.

C. Solvent Effects. Since the presence of dioxane in the aqueous solution was necessary to keep the phosphoramidates dissolved, the effect of the variation of the percentage of dioxane in the solvent was studied. This was done for the methyl derivative in five different solutions. Table IV gives the kinetic results for these

Table IV. The Effect of Solvent Changes on the Rate of Hydrolysis of 2,4-Dichlorophenyl Methyl N-Methylphosphoramidate at 25° in 0.19 *M* HCl

Dioxane,[a] %	$[H_2O]$, *M*	Dielectric[b] constant	k, min^{-1}	k_2, l./(mol min)
10.0	50.0	71.2	0.127[c]	0.0025
14.5	47.5	67.2	0.085 ± 0.001	0.0018
24.5	42.0	58.4	0.0455 ± 0.0003	0.0011
49.5	28.0	36.3	0.0103 ± 0.0001	0.0004
74.0	14.5	14.8	0.0061 ± 0.0009	0.0004

[a] Per cent dioxane in water, v/v. [b] From a plot of vol % dioxane *vs. D* prepared from data of King and Josephs in A. A. Frost and R. G. Pearson, "Kinetics and Mechanism," John Wiley and Sons, Inc., New York, N. Y., 1953, p 135. [c] Only one run.

hydrolyses, all of which were carried out at 25° at the same calculated pH (0.70–0.72) and at a phosphoramidate concentration of 0.002 *M*. The second-order rate constants, k_2, were obtained by dividing the experimentally observed first-order constants by the concentration of water. It is seen that an increase in the dielectric constant of the solvent from about 15 to 71

(9) By courtesy of Dr. E. H. Blair, Edgar C. Britton Research Laboratory, The Dow Chemical Company.

results in a sixfold increase in k_2. As supplemental evidence for the solvent effect, the reaction of the isopropylphosphoramidate under conditions identical with those of the methyl derivative was followed in 10 and 24.5% dioxane. The ratio of first-order rate constants for the two solutions was 2.6. The ratio of first-order rate constants for the methyl derivative in the same two solvents was 2.8. This shows the validity of extrapolating solvent effects from one member of the phosphoramidate series to another.

D. The Kinetic Effect of Deuterium Oxide. The hydrolyses of the methyl- and of the isopropylphosphoramidate were followed in D_2O solutions containing 0.19 *M* HCl and 25% dioxane. The ratio of the rate in D_2O as compared with that in H_2O, all other variables being the same for both reactions, was 2.08 for the methylphosphoramidate at 25° and 1.64 for the isopropylphosphoramidate at 45°. These data are summarized in Table V.

Table V. The Kinetic Effect of Deuterium Oxide on the Rate of Acid Hydrolysis of Two 2,4-Dichlorophenyl Methyl N-Alkylphosphoramidates (I)

N-Alkyl substituent	k, min^{-1}		k_D/k_H
	D_2O	H_2O	
Methyl (25°)	0.0946 ± 0.001	0.0455 ± 0.0003	2.08
Isopropyl (45°)	0.0199 ± 0.0002	0.0121 ± 0.0001	1.64

E. Effect of Ionic Strength. For the majority of kinetic studies discussed in this work, the concentration of hydrochloric acid was 0.19 *M*, and the ionic strength was therefore 0.19. Two reactions of the methylphosphoramidate were run at increased ionic strengths to determine if there was any effect on the velocity, enough potassium chloride being added to achieve the desired strengths. The ionic strengths and observed reaction rate constants were as follows: 0.19 μ, k = 0.0455 min^{-1}; 0.30 μ, k = 0.0465 min^{-1}, 0.40 μ, k = 0.0407 min^{-1}. As shown by these experimental results, no meaningful effect on reaction rate was observed by varying the ionic strength over this relatively narrow range.

F. pH Dependence Studies. The hydrolysis of 2,4-dichlorophenyl methyl N-methylphosphoramidate was studied in solutions of six different pH values ranging from 0.75 to 3.3 in order to determine the pH dependence of the reaction. It was assumed that the other five phosphoramidates would show the same dependence on the pH of the medium. In all solutions the concentration of methylphosphoramidate was 0.002 *M* and the solvent was 25% dioxane by volume. All reactions were carried out either in large concentrations of acid (0.19 *M*) or in buffered solutions so that any change in acid concentration as the reactions progressed would be negligible. Table VI gives the observed rate constant at each pH value studied. In a plot of this data (pH *vs.* log k), all points fall reasonably close to a straight line with a slope of −1.0. No attempt was made to hold the ionic strength constant for the various pH values. The ionic strength at pH 0.75 was 0.19, while the ionic strength at pH 1.48, 1.78, and 2.27 was constant at 0.05. However, the data given above (section E) show that changes in ionic strength over this narrow range probably have a negligible effect on the reaction rate.

Table VI. The Effect of pH on the Rate of Hydrolysis of 2,4-Dichlorophenyl Methyl N-Methylphosphoramidate at 65.5°

pH	k, min^{-1}
0.75	0.373
1.48	0.108 ± 0.001
1.78	0.045 ± 0.002
2.27	0.010
2.65	0.0077 ± 0.0006
3.32	0.0013

G. The Taft Correlations. The Taft equation[10]

$$\log (k/k_0) = \sigma^*\rho^* \quad (2)$$

gave a fair linear correlation between structures of the phosphoramidates and their rates of hydrolysis, the slope of the line, ρ^*, being equal to +8.3. This value is of too high a magnitude to be accepted without question, however. The largest ρ^* value recorded in Taft's tables is 4.68.[10] A plot of the same rate constants *vs.* E_s, Taft's steric substituent constant, was made using Taft's linear steric energy relationship[10]

$$\log (k/k_0) = \delta E_s \quad (3)$$

Linear correlation was not good.

An equation[10] (4) combining both polar and steric effects gives a reasonably good correlation between reaction rate and structure for the phosphoramidate hydrolyses. The plot of the data according to this

$$\log (k/k_0) = \sigma^*\rho^* + \delta E_s \quad (4)$$

equation is shown in Figure 3, and Table VII gives the values of σ^*, E_s, and k for each substituent. In this equation, k_0, σ^*, and ρ^* mean the same as in the Taft equation (2), while E_s is Taft's steric substituent constant and δ is the steric reaction constant. The value of δ was approximated to be +0.6 by trying various numbers for δ and plotting log $(k/k_0) - \delta E_s$ against σ^* for each trial until the most linear relationship was obtained. The slope of this line (Figure 3) was then found to be $\rho^* = +3.6$ by the least-squares method. The standard deviation of experimental measurements from the regression line of Figure 3 is 0.21. The correlation coefficient is 0.97. All reaction solutions were 0.002 *M* in phosphoramidate, 25% in dioxane, and 0.19

Table VII. Rate Constants and σ^* and E_s Values for the Taft Equation as Applied to the Acid-Catalyzed Hydrolysis of 2,4-Dichlorophenyl Methyl N-Alkylphosphoramidates at 25°

N-Alkyl substituent	σ^* value[a]	E_s value[a]	k, min^{-1} (25°)
Methyl	0.000	0.00	0.0455
Ethyl	−0.100	−0.07	0.0216
Butyl	−0.130	−0.39	0.0130
Isopropyl	−0.190	−0.47	0.00355[b]
sec-Butyl	−0.210	−1.13	0.00151[b]
t-Butyl	−0.300	−1.54	0.00046[b]

[a] Reference 10. [b] Calculated for 25° by extrapolation of the corresponding Arrhenius plot.

(10) R. W. Taft, Jr., in "Steric Effects in Organic Chemistry," M. S. Newman, Ed., John Wiley and Sons, Inc., London, 1956, Chapter 13.

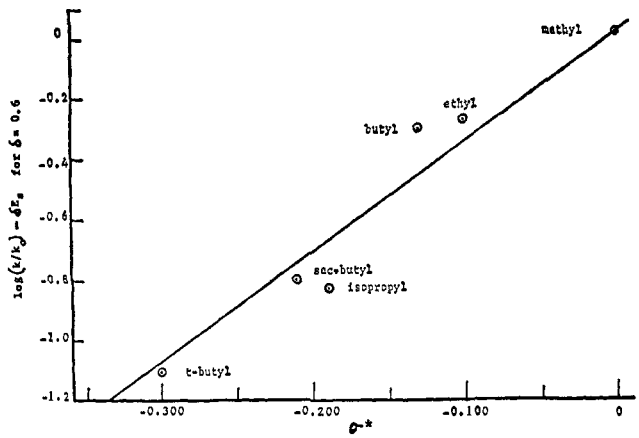


Figure 3. Plot of log $(k/k_0) = \sigma^*\rho^* + \delta E_s$ for the acid-catalyzed hydrolysis of a series of 2,4-dichlorophenyl methyl N-alkylphosphoramidates at 25°.

M in HCl. The reference rate constant, k_0, is k for the methyl derivative, 0.0455 min^{-1}.

Experimental Section

A. **Materials.** The six phosphoramidates were prepared by the method of Blair.[11] The starting material for all six compounds, 2,4-dichlorophenyl phosphorodichloridate, was prepared as described by Gefter.[12] The identities of the synthesized phosphoramidates were established by elemental analyses and by their nmr and ir spectra. They are all white crystals or colorless viscous liquids with a musty, garlic-like odor and are stable in air at room temperature. Distilled water was from the ordinary laboratory supply with no further purification. The hydrochloric acid was concentrated reagent grade. The dioxane used was Fisher reagent grade. The deuterium oxide was from Columbia Organic Chemicals Co., Columbia, S. C., and was stated to be 99.77% pure.

To check the effect of the purity of the dioxane on the kinetics of the reactions, a portion of the commercial reagent grade dioxane was purified according to a prescribed technique[13] and was then used as the solvent in a kinetic run instead of the commercial material (unpurified). The rate constant obtained in this run, using the methylphosphoramidate, was 0.0450 min^{-1} at 25°. The effect of the purity of the laboratory distilled water was checked by using redistilled water as the solvent in another kinetic run, a rate constant of 0.0458 min^{-1} being observed. These results were compared with the rate constant obtained for the methylphosphoramidate using the commerical dioxane and the laboratory supply of distilled water, this constant being 0.0455 min^{-1} (Table I). Considering these results, it was decided to use the commercial reagent grade dioxane and the laboratory distilled water as solvents without further purification.

B. **Equipment.** A Beckman DU spectrophotometer (Model 2400) was used to follow the kinetics. This instrument was equipped with a 2180 Dual Thermospacer Set (Beckman Instrument Co.) which served as a constant-temperature cell compartment. Water at a constant temperature was circulated through this compartment by use of a Bronwill constant-temperature circulator (Bronwill Scientific, Rochester, N. Y. 14601).

C. **Stock Solutions.** A solution of approximately 25% by volume dioxane was made by mixing 250 ml of reagent grade dioxane with 750 ml of distilled water. A stock solution of 0.38 M HCl was prepared by dissolving the calculated amount of concentrated acid in the necessary volume of 25% dioxane solution, titrating with standardized sodium hydroxide, and adjusting to 0.38 M. All phosphoramidate stock solutions were 0.004 M. The proper amount of the compound was dissolved in 100 ml of the 25% dioxane solution prepared above. These phosphoramidate solutions were stable over a period of several days as shown by the fact that occasional measurements showed little change in ultraviolet absorption at 275.5 mμ.

D. **Kinetic Methods.** For a typical kinetic run, the hydrolysis solution was prepared by mixing 5 ml of the 0.004 M phosphoramidate stock solution (in 25% dioxane) with 5 ml of the 0.38 M HCl stock solution (in 24% dioxane). This gave a solution of 0.002 M phosphoramidate and 0.19 M HCl (pH 0.75) in 24.5% dioxane by volume. For runs at temperatures of 35° or higher, the phosphoramidate and HCl stock solutions and the mixing pipet were brought to reaction temperature before preparing the hydrolysis solution.

A portion of the hydrolysis solution, as prepared above, was transferred to a 1-cm, glass-stoppered quartz cuvette and placed in the instrument cell compartment. Absorbance readings were taken manually at certain time intervals (from the time of mixing the hydrolysis solution) at a constant wavelength of 275.5 mμ against a reference of 0.002 M phosphoramidate in 25% dioxane with no HCl. The temperature was maintained constant to within ±0.1° (within ±0.2° at temperatures of 45° or higher). After each run, the cuvettes were rinsed thoroughly with 25% dioxane in water.

The prearranged time intervals for readings were closer together at the beginning of the reaction, 1 or 2 min, and gradually became longer as the reaction progressed. Most of the faster reactions were followed to between 80 and 90% of completion, while the slower ones were usually followed to between 65 and 80%. The data were plotted according to the method of Guggenheim,[8] which eliminates the need for determining A_∞. Duplicate runs were usually made using the same stock solutions. The great majority of the kinetic plots were linear and rate constants calculated from duplicate runs were usually reproducible to a range of less than 5% of the average value of the two runs, as may be seen in Table I. The rate constants for reactions at 15, 25, and 35° were usually reproducible to within 2%, while those at high temperatures, or for the very slow reactions, sometimes varied in the order of 10%.

Kinetic runs to determine the solvent dependence of the reaction were followed as described above, the per cent of dioxane in the hydrolysis solution and in the reference solution being varied as indicated in Table IV. Kinetic experiments to determine the effects of ionic strength were also carried out as described above except that the required amounts of potassium chloride (electrode grade) to give the desired ionic strengths were added to the hydrolysis solutions just before addition of the HCl. These reactions were 0.002 M in phosphoramidate, 0.19 M in HCl, and 24.5% in dioxane.

E. **pH Dependence Studies (Table VI).** These reactions were followed as described above. The concentration of the methylphosphoramidate was 0.002 M and the solvent was 25% dioxane by volume in all reactions. The reactions were run at 65.5° except for the pH 0.75 and 2.27 runs which were at 25°. The k values for the latter two runs at 65.5° were extrapolated from existing data. The reaction at pH 2.27 was followed for only 1 hr, so the calculated k is only for the initial reaction. This reaction and the reaction at pH 3.32 were run only one time; all other reactions were run in duplicate. The pH of each hydrolysis solution was measured just after mixing the solution and again at the end of the reaction, both times after the solution was cooled to room temperature, and the pH of each reaction was considered to be the average of these two measurements. The pH meter was calibrated with a pH 2.00 buffer at room temperature. The hydrolysis solutions were prepared by mixing 5 ml of 0.004 M methylphosphoramidate in 25% dioxane with 5 ml of the appropriate buffer made up in 25% dioxane. The buffering agents for the various pH ranges were as follows: pH 0.75, no buffer, this reaction run in 0.19 M HCl at 25° as usual; pH 1.48–2.27, KCl and HCl as required; pH 2.65 and 3.32, tartaric acid and potassium sodium tartrate as required.

F. **Kinetic Isotope Effects.** Stock solutions and hydrolysis solutions were prepared as described under C and D above, using deuterium oxide rather than H_2O. The H_2O in the concentrated hydrochloric acid diluted the D_2O so that the total aqueous portion of the final hydrolysis solution was about 98.5% D_2O and 1.5% H_2O. The reactions were followed in the usual way and were run in duplicate.

G. **Product Determination. a. Amine Analysis.** The final hydrolysis solutions were analyzed after reaction for total amines present in the form of amine salts. Known amounts of 0.002 M solutions of each of the phosphoramidate derivatives in 25% dioxane and 0.19 M HCl were prepared and allowed to stand a sufficient time for complete hydrolysis. The solutions were then transferred to a conventional distillation apparatus, made basic with 10% NaOH, and heated to 100° with an oil bath. The evolved

(11) C. L. Moyle, and L. L. Wade, U. S. Patent 2,944,937 (July 12, 1960).

(12) E. L. Gefter, "Organophosphorus Monomers and Polymers," Associated Technical Services, Inc., Glen Ridge, N. J., 1962, pp 88–89.

(13) A. Weissberger, Ed., "Techniques of Organic Chemistry," Vol. VII, 2nd ed, Interscience Publishers, Inc., New York, N. Y., 1955, p 371.

amines were collected under water. The amine solutions thus obtained were titrated with HCl by the method of Siggia.[14] The results, reported in Table II, include corrections for the blanks described in the following paragraph.

To ascertain that the analytical process did not itself produce any amine from the phosphoramidates, fresh solutions of 100 ml each of 0.002 *M* methyl-, ethyl-, isopropyl-, and *sec*-butylphosphoramidates in 25% dioxane were prepared but were not treated with acid. These solutions were subjected to the same process of being made basic, distillation, and titration as described above. These nonhydrolyzed samples served as blanks for the hydrolyzed samples.

b. Estimation of 2,4-Dichlorophenol. To determine the amount of 2,4-dichlorophenol present in the hydrolyzate from each phosphoramidate, a solution of 0.002 *M* phosphoramidate in 0.2 *M* HCl and 25% dioxane was prepared in 100 ml quantity for each member of the series and allowed to stand long enough for complete hydrolysis. The solutions were extracted with a total of 60 ml of chloroform, the extracts were dried over sodium sulfate, and 3 ml of each extract was made to volume with chloroform in a 200-ml volumetric flask. The absorbance of each solution was read on the Beckman DU spectrophotometer at 284 mμ in a 1-cm quartz cell. The amount of 2,4-dichlorophenol in each solution was then obtained by comparing this absorbance reading to a standard curve. No significant amount of the phenol was found in any of the hydrolyzed phosphoramidate solutions.

c. Alcohol Determination. A 0.02 *M* solution of the methylphosphoramidate in 25% dioxane and 0.19 *M* HCl was prepared in an nmr tube and allowed to stand a sufficient time for complete hydrolysis as indicated by the presence of a quartet at τ 7.44 (J = 6.5 cps) due to the $CH_3NH_3^+$ cation. No signal was observed at τ 6.67, the frequency of absorption of the methyl protons in a reference solution of 0.2 *M* methanol in 25% dioxane and 0.19 *M* HCl. A similar experiment was carried out with the isopropylphosphoramidate. The doublet due to the isopropyl methyl protons shifted from τ 8.85 (J = 6.5 cps) to 8.70 (J = 6.5 cps) during the course of the hydrolysis, while the absorption of the isopropyl methyl protons in a reference solution of isopropyl alcohol was at τ 8.88 (J = 6.0 cps).

d. Spectrophotometric Evidence. Spectrophotometric analysis also gave evidence of the character of the reaction products. The absorbance of a 0.002 *M* solution of the pure, synthetically prepared methylamine salt of 2,4-dichlorophenyl methyl phosphoric acid (II, R = methyl),[9] the expected hydrolysis product of the methylphosphoramidate, was 1.79 at 275.5 mμ. This was in 0.19 *M* hydrochloric acid in aqueous 25% dioxane solution, the same reaction medium used in making the kinetic measurements. This value, minus the absorbance of 0.002 *M* 2,4-dichlorophenyl methyl methylphosphoramidate in 25% dioxane solution at 275.5 mμ was 0.34 absorbance unit, the calculated ΔA. This was the calculated increase in absorbance at 275.5 mμ from the beginning of the hydrolysis to the end. The observed absorbance change during the actual kinetic runs, $A_\infty - A_0$, averaged 0.344 absorbance unit for the methylphosphoramidate. Since the hydrolysis products from the various phosphoramidates differ only in the alkyl moiety of the alkylamine cation, the absorptivity of each of the hydrolysis products should be the same. Therefore, the absorbance value of the methylamine salt given above (1.79) was used in calculating the ΔA value for each phosphoramidate hydrolysis. These calculated ΔA values agreed closely with the observed values, as shown in Table VIII, and this substantiates the over-all hydrolysis as proceeding according to reaction 1.

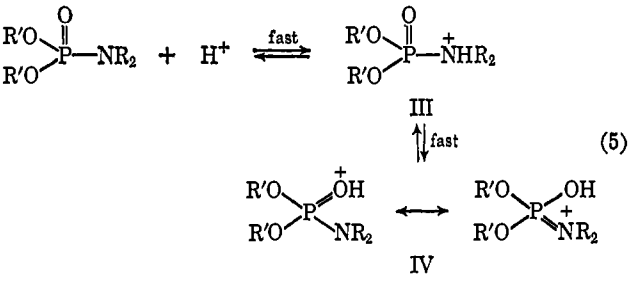
Table VIII. Calculated and Observed Absorbance Changes at 275.5 mμ during the Hydrolysis of the N-Alkylphosphoramidates

N-Alkyl substituent	Obsd ΔA ($A_\infty - A_0$)	Calcd ΔA
Methyl	0.344	0.34
Ethyl	0.330	0.32
Isopropyl	0.265	0.27
Butyl	0.269	...
sec-Butyl	0.250	0.26
t-Butyl	0.179	0.19

(14) S. Siggia, "Quantitative Organic Analysis via Functional Groups," 3rd ed, John Wiley and Sons, Inc., New York, N. Y., 1963, p 423.

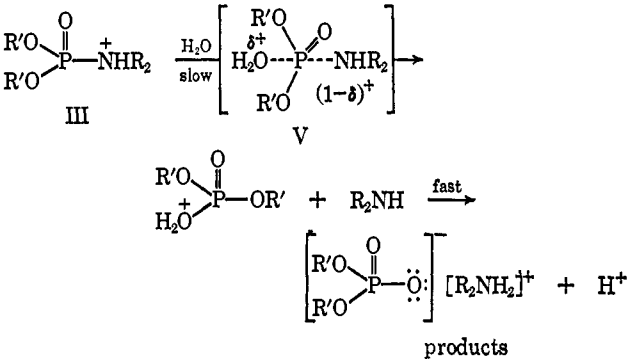
Discussion

As suggested by a survey of the chemical literature, the following four tentative mechanistic possibilities for the hydrolysis of phosphoramidate diesters in acid solution are advanced. These mechanisms are analogous to proposed mechanisms for acid hydrolysis of other phosphoramidic-type compounds[15] and of amides.[16-18] In each case, initial protonation of the nitrogen or the phosphoryl oxygen, according to the following equilibrium, is assumed (R = hydrogen or an alkyl group while R′ = an alkyl or aryl group).

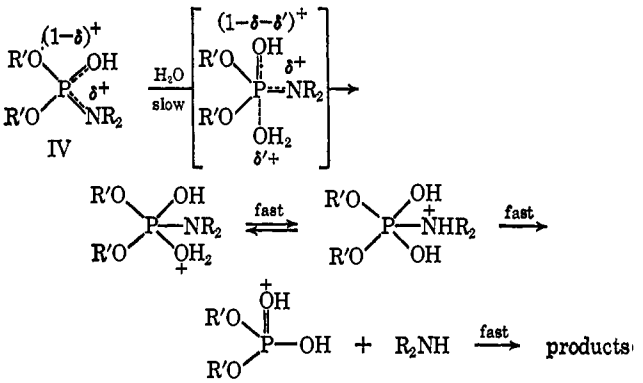


(5)

A. Direct substitution of water involving a rate-determining one-step process in which the P–N bond is being broken at the same time the water molecule is attacking the phosphorus atom.



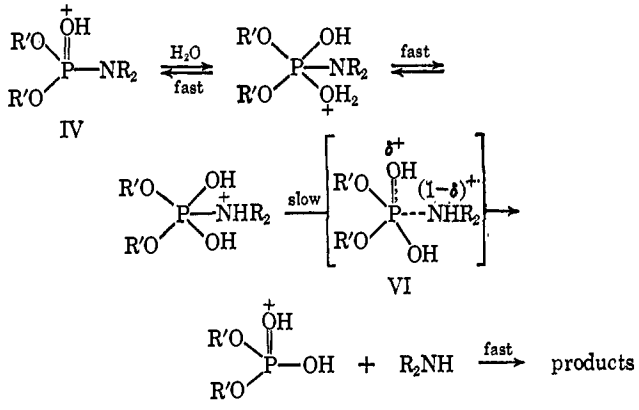
B. A two-step, addition–elimination sequence in which the slow step is addition of water to the phosphoryl group *via* attack at phosphorus. A subsequent rapid step involves unimolecular cleavage of the P–N bond.



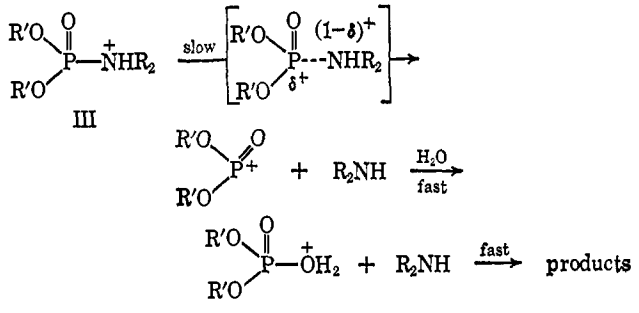
C. The same as B except that the addition of water is fast, the rate-determining step being subsequent unimolecular cleavage of the P–N bond to produce the

(15) J. R. Cox, Jr., and O. B. Ramsay, *Chem. Rev.*, **64**, 317 (1964).
(16) J. Hine, "Physical Organic Chemistry," 2nd ed, McGraw-Hill Book Co., Inc., New York, N. Y., 1962, pp 111, 291.
(17) J. Koskikallio, *Acta Chem. Scand.*, **18**, 1831 (1964).
(18) E. Whalley, *Advan. Phys. Org. Chem.*, **2**, 144 (1964).

phosphoric acid diester and the free amine. Evidence has been invoked for a similar mechanism in the acid-catalyzed hydrolysis of ethylene phosphoric acid.[19]



D. Unimolecular P–N bond cleavage followed by rapid reaction with water. This has been postulated to occur in the case of phosphoramidic acid in acid solution[20] and has been considered as a possibility in the acid-catalyzed hydrolysis of amides.[18]



Two other pathways were eliminated by the fact that no alcohol reaction products could be found after hydrolysis. These were (1) attack of water at the amino carbon atom with SN2 inversion and C–N bond cleavage to produce a more simple phosphoramidate and an alcohol, and (2) unimolecular C–N bond cleavage to form a simpler phosphoramidate and an alcohol. In both cases, the simpler phosphoramidate produced would have been unsubstituted at nitrogen and would have hydrolyzed rapidly to give products not distinguishable from those predicted by mechanisms A–D by the analytical methods described in the Experimental Section.

The observed reaction products resulting from P–N bond cleavage (the amine salts of 2,4-dichlorophenyl methyl phosphoric acid (II)) and the lack of observation of any significant P–O bond cleavage or of any alcohol products are consistent with each of the mechanisms A–D. Phosphorus-to-nitrogen bond cleavage was also observed by Sélim and Thanh[6,7] in their study of the acid hydrolysis of some dimethyl N,N-dialkylphosphoramidates, and by Heath and Casapieri[21] and Montgomery and Turnbull.[22]

A. The Kinetic Expression. The reactions were pseudo first order for each of the phosphoramidates under all conditions studied. The hydrolyses were carried out at a constant pH (from the beginning to the end of the reaction), so that the first-order character of the hydrogen ion concentration was not observed. The concentration of water, of course, was in large excess at all times. The observed rate expression is, therefore, rate = k[phosphoramidate], where k is the observed rate constant. Since the hydrolysis is first order in hydrogen ion, as discussed below, and assuming the order in water to be one, the true kinetic expression for the reaction is

$$\text{rate} = k'[H^+][H_2O][\text{phosphoramidate}]$$

That the reaction is first order in phosphoramidate is to be expected, and is borne out by previous work, including that of Sélim and Thanh[6,7] and Larsson.[23] The acid-catalyzed hydrolysis of phosphoramidic acid and the N-substituted phosphoramidic acids are also first order.[20,24]

The proof for first-order participation of the hydrogen ion (hydronium ion) is presented by the data of Table VI. It is known that the value of the slope of the plot of log k *vs.* pH should theoretically be -1 for such participation,[25] and this was, in fact, observed. Specific hydronium ion catalysis would be expected for these reactions *a priori* based on analogy with the acid hydrolysis of amides[18,25] and of various types of phosphoramidates.[20–23] Sélim and Thanh[6,7] found a linear relationship between the log of the rate constant and the pH of the reaction medium, showing the acid hydrolysis of a series of dimethyl N,N-dialkylphosphoramidates to be first order in hydronium ion.

Each of the tentative mechanisms A–D presented above involves first-order phosphoramidate participation as well as specific hydronium ion catalysis. A fast equilibrium step in which the phosphoramidate is protonated is followed by a slower step involving either attack of water on the protonated species or unimolecular decomposition of the protonated species.

B. Deuterium Isotope Effects. As shown in Table V, the replacement of H_2O with D_2O as the solvent for the phosphoramidate hydrolysis resulted in an approximately twofold increase in rate.

This rate increase is to be expected for most acid-catalyzed reactions, with a few exceptions.[26] If there is a rapid preequilibrium in which the reactant is converted to its conjugate acid followed by a second, rate-controlling step, as is the case with specific acid catalysis, the reaction will be fastest in the heavy water. This is assuming the rate-controlling step gives rise to no large counter-isotope effect. The enhancement is usually in the order of 1.3 to 3.0. For example, the specific acid-catalyzed hydrolysis of acetamide, a similar reaction to the one discussed, has a value of $k_D/k_H = 1.45$ in 0.1 N HCl.[26] Halmann, Lapidot, and Samuel[20] found the isotope effect in the hydrolysis of phosphoramidic acid to be $k_D/k_H = 1.4$. Although the rate of reaction in D_2O was determined for only two of the

(19) P. C. Haake and F. H. Westheimer *J. Am. Chem. Soc.*, **83**, 1102 (1961).
(20) M. Halmann, A. Lapidot, and D. Samuel, *J. Chem. Soc.*, 1299 (1963).
(21) D. F. Heath and P. Casapieri, *Trans. Faraday Soc.*, **47**, 1093 (1951).
(22) H. A. C. Montgomery and J. H. Turnbull, *J. Chem. Soc.*, 1963 (1958).
(23) L. Larsson, *Acta Chem. Scand.*, 7, 306 (1953).
(24) J. D. Chanley and E. Feageson, *J. Am. Chem. Soc.*, **85**, 1181 (1963).
(25) P. G. Ashmore, "Catalysis and Inhibition of Chemical Reactions," Butterworth and Co., London, 1963, p 30.
(26) K. Wiberg, *Chem. Rev.*, **55**, 718 (1955).

compounds in this phosphoramidate series, it is assumed that all six derivatives would show that $k_D > k_H$ and that the hydrolysis of the phosphoramidate diesters in acid solution proceeds by specific acid catalysis.

C. Solvent Effects. The fact that the calculated second-order rate constants, k_2 (Table IV), increase with increasing concentration of water is due to either of two possible causes: (1) the reaction is not first order in water as assumed in the calculations, or (2) there is a solvent effect due to some change in the medium, such as a variation in the dielectric constant. If the order in water is assumed to be two, the calculated third-order rate constants are neither constant throughout the range nor do they show a trend in any direction. It is more reasonable, then, to assume that the reaction is first order in water and that a medium effect is being observed. Table IV shows that there is only a sixfold increase in the second-order reaction rate constant as the dielectric constant of the solvent increases from about 15 to 71. The direction of this trend is not as expected. Each of the mechanisms A–D involves a dispersal of charge in the transition state of the rate-determining step relative to the reactants. Thus, an increase in polarity of the solvent should favor the reactants and decrease the reaction rate. (The solvent polarity should have little influence on the rapid prior protonation step of the reaction.) An analogous discrepancy was observed by Koskikallio,[17] who studied the acid hydrolysis of acetamide in a wide range of dioxane–water mixtures.

The *small magnitude* of the observed solvent effect is evidence that a transition state of considerably different polarity, relative to the reactants, is unlikely for the acid-catalyzed phosphoramidate hydrolysis.[27] This is consistent with the negligible ionic strength effects observed and with each of the tentative mechanistic possibilities A–D proposed above.

D. Entropies of Activation. The energies and entropies of activation for the acid-catalyzed hydrolyses of the 2,4-dichlorophenyl methyl alkylphosphoramidates are presented in Table III. The values of E_a range from 10.1 to 13.2 kcal/mol, while $\Delta S^{\pm}$ ranges from -30 to -35 eu.

It is generally found that bimolecular specifically acid-catalyzed reactions have large negative entropies of activation while unimolecular acid-catalyzed reactions have small activation entropies of either sign.[28,29] Koskikallio[17] found $\Delta S^{\pm}$ to be -19 eu for the acid-catalyzed hydrolysis of acetamide in water containing 0.1 *N* $HClO_4$ and reported $\Delta S^{\pm}$ to be -18.2 eu for acetamide in water containing 0.1 *N* HCl. He gave these values as evidence for a bimolecular reaction. Schaleger and Long reported the hydrolysis to proceed by a bimolecular mechanism and to have $\Delta S^{\pm} = -37$ eu in 1 *M* HCl.[28] It is informative to calculate the entropies of activation from the activation energies and rate constants found by Sélim and Thanh[7] in their study of the acid-catalyzed hydrolysis of a series of dimethyl N,N-dialkylphosphoramidates. Their series of compounds is similar to those studied here, and the activation parameters should be similar. The $\Delta S^{\pm}$ values calculated from their data range from -32 to -36 eu and the energies of activation range from 10.0 to 13.7 kcal/mol.

It therefore seems reasonable to conclude that the large negative entropies of activation determined in this work are very good evidence for a bimolecular rate-determining step in the acid-catalyzed hydrolysis of the phosphoramidates. This is sufficient to all but eliminate mechanism D from the list of tentative possibilities presented earlier, as this involves a unimolecular decomposition in the rate-determining step. Also, mechanism C should not involve a large negative entropy of activation.

E. Substituent Effects. Equation 4, advanced by Taft, has found only limited application. However, a plot of this equation (Figure 3) shows as good or better correlation between reactivities of the phosphoramidates and their structures than does eq 2. The value of ρ^* in eq 4 was calculated to be $+3.6$, indicating that the polar character of the N-alkyl group produces the predominant substituent effect. The steric character of the group has only a small effect on the reactivity, as is shown by the small value of the steric reaction constant, δ $+0.6$. The positive value of ρ^* shows that the reaction is facilitated by electron-withdrawing substituents, while the positive δ value indicates that an increase in bulk of the alkyl substituent sterically retards the rate of hydrolysis to a slight extent. Although a more precise fit of the data to the straight line of Figure 3 would be desirable, there is no question about the over-all qualitative value of the plot. The proposed transition state for each of the mechanisms A–D involves a decrease in positive charge at the nitrogen atom relative to its charge in the ground state of the molecule. This means that electron attraction by the substituent on the nitrogen would facilitate the reaction, in agreement with the Taft correlation (Figure 3). Of course, the magnitude of ρ^* should be different for each type of transition state since they all involve a different degree of increase in negativity at nitrogen. For example, in mechanism B, since the P–N bond is broken subsequent to the rate-determining step, the decrease in positive charge on nitrogen on going from reactant to transition state is small and a ρ^* value as large as that observed might not be expected.

It is obvious that in either the nitrogen (III) or oxygen (IV) protonated forms of the prior protonation equilibrium (reaction 5) before the rate-determining step, electron-donating substituents on nitrogen will favor the protonated phosphoramidate by partial stabilization of the positive charge. The polar effects in this step will thus tend to cancel some of the opposite but larger effects in the rate-determining step. In fact, in mechanism B, the over-all effect might be the enhancement of rate by electron donors.

All the proposed mechanisms are not in accord with the observed steric effect. Taking only a qualitative approach, it is observed experimentally that an increase in bulk of the substituent on nitrogen hinders the reaction. This is consistent with all the bimolecular mechanisms presented above (A, B, and C), where either direct displacement by water or addition of water to the phosphoryl group results in an increase of the coordination number of phosphorus from four to five in going from the ground state to the transition state. This steric

(27) E. A. Gould, "Mechanisms and Structure in Organic Chemistry," Henry Holt and Co., New York, N. Y., 1959, p 253.
(28) L. L. Schaleger and F. A. Long, *Advan. Phys. Org. Chem.*, **1**, 1 (1963).
(29) E. Whalley, *ibid.*, **2**, 136 (1964).

crowding should cause a small reduction in rate with increase in size of the substituent on nitrogen.[30] On the other hand, in the proposed unimolecular reaction mechanism D, a larger alkyl group on nitrogen would facilitate the reaction by relief of steric strain around phosphorus or carbon in going from the ground state to the transition state. Steric effects of the alkyl group in mechanism C would be difficult to analyze.

Steric effects in the prior equilibrium step should not be important, since the protonated and unprotonated phosphoramidates differ only by the presence or absence of a proton.

F. The Site of Initial Protonation. The site of protonation of the phosphoramidate to form the conjugate acid is still in question, with no direct experimental proof to distinguish between phosphoryl oxygen (IV, reaction 5) and nitrogen (III) protonation. Several amides have been shown to be protonated at the oxygen in strong acid, but with some small degree of N-protonation in aqueous acid.[31] This has been discussed by other workers also.[16-18] In most of the previous work involving acid hydrolysis of phosphoramidates of various types, nitrogen protonation is assumed, but no proof is given.

Some indirect evidence is available to show that protonation of the phosphoramidates probably involves mainly the oxygen atom of the phosphoryl group. First, the phosphoryl group is more polar than the carbonyl group; *i.e.*, oxygen has a considerable partial negative charge. In addition, the partially positive phosphorus should have a marked tendency to form a p_π–d_π bond with the nitrogen in phosphoramidates.[32] (This reduces the positive charge on phosphorus and is the main reason for the unreactivity of phosphoramidates to basic hydrolysis.[33]) This would tend to stabilize by resonance the oxygen protonated form of the phosphoramidate (IV, reaction 5), whereas no such stabilization is possible in the nitrogen-protonated form III. The nitrogen atom would tend to be less basic because of this π-bond formation. This is experimentally indicated by the ease of exchange of the nitrogen proton (in the neutral form of the phosphoramidates in neutral solution) for deuterons from D_2O as observed by nmr.[34] This facile exchangeability was shown by Traylor and Westheimer[35] to also exist in phosphoramidochloridates.

(30) Reference 10, p 629.
(31) G. Fraenkel and C. Franconi, *J. Am. Chem. Soc.*, **82**, 4478 (1960).
(32) R. F. Hudson and L. Keay, *J. Chem. Soc.*, 1859 (1960).
(33) J. R. Cox, Jr., and O. B. Ramsay, *Chem. Rev.*, **64**, 323 (1964).
(34) The nitrogen proton absorption produced a multiplet in the nmr spectrum of each phosphoramidate (in $CDCl_3$) at between τ 5.03 and 4.74. This multiplet disappeared in each case upon shaking the solution of the sample with about 0.2 ml of D_2O.
(35) P. S. Traylor and F. H. Westheimer, *J. Am. Chem. Soc.*, **87**, 553 (1965).

The above evidence and analogies lead to the belief that O protonation in the phosphoramidates in probably even more important than in the amides. However, the protonation of nitrogen is possible, at least to some extent. Therefore, it is postulated here than an equilibrium occurs between the unprotonated and the two protonated forms, III and IV, as shown by reaction 5. This same type of equilibrium was proposed by Koskikallio[17] to be involved in the acid hydrolysis of acetamide.

Any of the proposed mechanisms could be accommodated by this equilibrium since either form III or form IV would react with water.

Conclusions

In view of the above discussion, it is seen that mechanism A of the list of tentative mechanistic proposals is consistent with all the experimental evidence presented, while mechanism D can be eliminated since it is not consistent with either the large negative entropies of activation or the observed steric retardation effect. The other two pathways are possible, although pathway C is considered less likely than A because of a possible lack of consistency with the large negative entropies of activation, while pathway B is not completely consistent with the observed polar effects and is probably even less likely than C. It would be difficult to distinguish experimentally between mechanisms A and B, but pathways A and C could be distinguished by oxygen exchange experiments.[36,37]

The mechanistic process for all organophosphorus bimolecular displacements is usually pictured as the more simple direct displacement. However, an analogy with carbon ester and amide hydrolysis, in which addition–elimination mechanisms often are involved, and the fact that phosphorus can expand its octet to allow formation of the addition intermediate, demand that both methods of attack be considered as possibilities.

The proposal of the existence of both mechanisms A and C follows naturally after the assumption of the protonation equilibrium involving both forms III and IV (reaction 5). Protonated species III would tend to be attacked by water by the direct displacement mechanism A since the transition state resulting from *addition* of water to III would be of much higher energy than transition state V. By the same reasoning, the oxygen-protonated species IV would call for the addition mechanism C involving transition state VI rather than reacting by the high-energy transition state of a direct displacement process.

(36) J. R. Cox, Jr., and O. B. Ramsay, *Chem. Rev.*, **64**, 320 (1964).
(37) I. Dostrovsky and M. Halmann, *J. Chem. Soc.*, 1004 (1956).