The Acetylpyridinium Ion Intermediate in Pyridine-Catalyzed Hydrolysis and Acyl Transfer Reactions of Acetic Anhydride. Observation, Kinetics, Structure–Reactivity Correlations, and Effects of Concentrated Salt Solutions[1]

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Abstract: The formation and hydrolysis of the acetylpyridinium ion intermediate (λ_{max} 272 nm, ϵ *ca.* 4400; 225 nm, ϵ *ca.* 7000) in the pyridine-catalyzed hydrolysis of acetic anhydride have been observed directly. A kinetic treatment is described for dealing with this two-step reaction, in which a significant amount of intermediate accumulates in a reversible first step. Identical rate constants (84 ± 2 M^{-1} sec^{-1}) for acetylpyridinium ion formation and for the pyridine-catalyzed acetylation of 10^{-3} M toluidine and anisidine by acetic anhydride establish that these transacylation reactions proceed through the same intermediate and that formation of the intermediate is rate determining. Rate and equilibrium constants for the formation and hydrolysis of the N-acetyl derivatives of 4-methylpyridine, 3,4-lutidine, and 4-methoxypyridine are also reported. The hydrolysis of these compounds is subject to general base catalysis by acetate ion and by the corresponding pyridine. Structure–reactivity correlations give β values (based on the pK of the pyridine) of 0.9 and 1.6 for the rate and equilibrium constants, respectively, for acetylpyridinium ion formation from acetic anhydride and −0.5 and −0.7 for the rate constants for the reactions of substituted acetylpyridinium ions with water and acetate, respectively. Acetyl-4-methoxypyridinium and acetylimidazolium ions show deviations from these plots which are attributed to resonance stabilization. Concentrated sodium perchlorate causes a striking decrease in the rate of hydrolysis of acetylpyridinium ion (700-fold at 9 M) but *increases* the rate of reaction of pyridine with acetic anhydride.

It is generally believed that the pyridine-catalyzed hydrolysis of acetic anhydride proceeds through the intermediate formation of the N-acetylpyridinium ion (eq 1). Pyridine-catalyzed hydrolyses of other acti-

$$\mathrm{Py} + \mathrm{Ac_2O} \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} \mathrm{AcO^-} + \mathrm{AcPy^+} \xrightarrow{k_2} \mathrm{Py} + \mathrm{AcOH} \quad (1)$$

$$+\mathrm{HY} \downarrow k_N$$

$$\mathrm{AcY}$$

vated acyl compounds with good leaving groups are thought to proceed through the same intermediate and other pyridine-catalyzed acylation reactions presumably involve the same mechanism, with another nucleophile, HY, replacing water as the eventual acyl group acceptor (eq 1, k_N). Pyridine is an efficient catalyst for these reactions because it is a highly effective nucleophile for acyl compounds with a good leaving group and because the presumed intermediate acylated tertiary amine cannot lose a proton to give a resonance-stabilized amide and is, therefore, highly reactive toward water and other nucleophiles.

The evidence for the mechanism of eq 1 is kinetic: (a) the pyridine-catalyzed hydrolysis of acetic anhydride is orders of magnitude faster than the general base catalyzed hydrolysis of acetic anhydride by acetate ion and must, therefore, proceed by a different mechanism;[2,3] (b) the reaction is inhibited by acetate ion, which would be expected to react with acetylpyridinium ion to regenerate starting materials (eq 1, k_{-1});[4] (c) pyridine catalyzes a rapid exchange of labeled acetate ion into acetic anhydride, which can be explained by the same back reaction;[5] and (d) the steric hindrance introduced by a 2-methyl substituent causes a decrease of over 100-fold in the catalytic efficiency of pyridine.[4] Inhibition of the pyridine-catalyzed solvolysis of phenyl acetates by the leaving phenolate ion and of acyl fluoride by fluoride ion provides evidence for an acetylpyridinium ion intermediate in these reactions.[6-8] However, attempts to observe directly the formation of acetylpyridinium ion from acetic anhydride have been unsuccessful and it has been generally believed that it is too unstable for detection.[2,9]

The phosphorylpyridinium ion has been observed as an intermediate in pyridine-catalyzed hydrolysis and phosphoryl transfer reactions of phosphoramidate,[10] and a rough estimate of the free energy of hydrolysis of the acetylpyridinium ion relative to that of acetic anhydride,[11] based on the free energy of hydrolysis and pK of acetylimidazole and the dependence of the free energy of hydrolysis of acyl compounds on the acidity of the leaving group,[12,13] suggested that this intermediate would accumulate in detectable concentrations during the pyridine-catalyzed hydrolysis of acetic anhydride in water, a solvent in which the equilibrium of the first step of eq 1 should be relatively favorable. It has been

(1) Supported by grants from the National Science Foundation (GB 5648) and the National Institute of Child Health and Human Development of the National Institutes of Health (HD 01247).
(2) V. Gold and E. G. Jefferson, *J. Chem. Soc.*, 1409 (1953).
(3) M. Kilpatrick, Jr., *J. Amer. Chem. Soc.*, **50**, 2891 (1928).
(4) A. R. Butler and V. Gold, *J. Chem. Soc.*, 4362 (1961).
(5) C. A. Bunton, N. A. Fuller, S. G. Perry, and V. J. Shiner, *Tetrahedron Lett.*, 458 (1961).
(6) W. P. Jencks and M. Gilchrist, *J. Amer. Chem. Soc.*, **90**, 2622 (1968).
(7) C. A. Bunton and J. H. Fendler, *J. Org. Chem.*, **31**, 2307 (1966).
(8) W. R. Ali, A. Kirkien-Konasiewicz, and A. Maccoll, *Chem. Ind. (London)*, 809 (1964).
(9) D. E. Koshland, Jr., *J. Amer. Chem. Soc.*, 74, 2286 (1952).
(10) W. P. Jencks and M. Gilchrist, *ibid.*, **87**, 3199 (1965).
(11) W. P. Jencks, F. Barley, R. Barnett, and M. Gilchrist, *ibid.*, **88**, 4464 (1966).
(12) J. Gerstein and W. P. Jencks, *ibid.*, **86**, 4655 (1964).
(13) W. P. Jencks and J. Carriuolo, *J. Biol. Chem.*, **234**, 1272 (1959).

reported in a preliminary communication that the formation and decomposition of the acetylpyridinium ion can, in fact, be observed spectrophotometrically in the course of this reaction.[14] We describe here the details of these experiments, proof that the same compound is an intermediate in the pyridine-catalyzed acetylation of anilines, structure–reactivity data for the formation and reactions of a series of substituted acylpyridinium ions, and a relatively simple method for dealing with the kinetics of a system, such as that of eq 1, in which an intermediate accumulates at a significant concentration.

Kinetic Treatment

The following method of analyzing the non-steady-state system of consecutive reactions, first step reversible, does not appear to be of common knowledge, although this type of kinetics is by no means rare, and the principle for its treatment is known.[15] For example, it has been recently stated in a similar situation[16] that "if k_1, k_{-1}, and k_2 do not differ greatly from one another, there is no simple integrated solution or quantities which may be plotted to solve for k_1 and k_{-1}," and indeed, we know of no such method available in the common textbooks and reviews. It is hoped, therefore, that the following method will be of general interest.

The intermediate B in the reaction sequence

$$A \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} B \xrightarrow{k_2} C \qquad (2)$$

has a time dependence given by

$$B = \frac{A_I^0 k_1}{\lambda_1 - \lambda_2}(e^{-\lambda_2 t} - e^{-\lambda_1 t}) \qquad (3)$$

or, in terms of ultraviolet absorption

$$A_B = \frac{\epsilon A_I^0 k_1}{\lambda_1 - \lambda_2}(e^{-\lambda_2 t} - e^{-\lambda_1 t}) \qquad (4)$$

where A_I^0 is the initial concentration of A, t represents time, A_B is the absorption of B, ϵ is its extinction coefficient, and λ_1 and λ_2 are kinetic constants which define the shape of the curve for the appearance and disappearance of B, according to eq 5 and 6. These equations were obtained by the integration of the differential equations by the usual method.[17]

$$\lambda_1 = {}^1/_2(k_1 + k_{-1} + k_2 + [(k_1 + k_{-1} + k_2)^2 - 4k_1k_2]^{1/2}) \qquad (5)$$

$$\lambda_2 = {}^1/_2(k_1 + k_{-1} + k_2 - [(k_1 + k_{-1} + k_2)^2 - 4k_1k_2]^{1/2}) \qquad (6)$$

The pyridine-catalyzed hydrolysis of acetic anhydride below pH 7 may be described by the scheme

$$Ac_2O \underset{k_{-1}[AcO^-]}{\overset{k_1[Py]}{\rightleftharpoons}} AcPy^+ \xrightarrow{k_0 + k_{2_{py}}[Py] + k_{2_{AcO^-}}[AcO^-]} Py + AcO^- \qquad (7)$$

Under experimental conditions in which the concentrations of pyridine and acetate are much larger than that of acetic anhydride, the individual steps are pseudo-first-order and the reaction may be treated according to eq 2, in which "k_1" = k_1[Py], "k_{-1}" = k_{-1}[AcO$^-$], and "k_2" = $k_0 + k_{2_{py}}$[Py] + $k_{2_{AcO^-}}$[AcO$^-$] The values of λ_1 and λ_2 are obtained from the ascending and descending parts of the curve for the appearance and disappearance of B, as described in the Experimental Section. The rate constants may be obtained simply from the λ's by the following manipulations. The addition of eq 5 and 6 gives

$$\lambda_1 + \lambda_2 = k_0 + (k_1 + k_{2_{py}})[Py] + (k_{-1} + k_{2_{AcO^-}})[AcO^-] \qquad (8)$$

thence a plot of $\lambda_1 + \lambda_2$ against [Py] for constant [AcO$^-$] and varying [Py] gives a slope of $(k_1 + k_{2_{py}})$ with intercept $k_0 + (k_{-1} + k_{2_{AcO^-}})$[AcO$^-$]. Similarly a plot of $\lambda_1 + \lambda_2$ against [AcO$^-$] at constant [Py] gives a slope of $(k_{-1} + k_{2_{AcO^-}})$ and intercept $k_0 + (k_1 + k_{2_{py}})$[Py]. Examples of such plots for the reaction of acetic anhydride with pyridine at varying pyridine and acetate concentrations are shown in Figures 1 and 2, respectively.

The product of eq 5 and 6 gives

$$\lambda_1\lambda_2 = k_1[Py](k_0 + k_{2_{py}}[Py] + k_{2_{AcO^-}}[AcO^-]) \qquad (9)$$

so that a plot of $\lambda_1\lambda_2$/[Py] against [Py] at constant [AcO$^-$] gives a slope of $k_1k_{2_{py}}$ and intercept $k_1k_{2_{AcO^-}}$[AcO$^-$] + k_1k_0. For constant [Py] and varying [AcO$^-$] the plot of $\lambda_1\lambda_2$/[Py] against [AcO$^-$] gives a slope of $k_1k_{2_{AcO^-}}$ and intercept $k_1k_{2_{py}}$[Py] + k_1k_0.

Thus, the values of k_0, $(k_1 + k_{2_{py}})$, and $(k_{-1} + k_{2_{AcO^-}})$ are obtained from eq 8, and the values of k_1k_0, $k_1k_{2_{AcO^-}}$, and $k_1k_{2_{py}}$ from eq 9. As k_0 is known, k_1 is found from k_1k_0 which then gives $k_{2_{AcO^-}}$ and $k_{2_{py}}$, to finally give all the individual rate constants.

Experimental Section

Materials. Commercial acetic anhydride, pyridine, 4-methylpyridine, and 3,4-lutidine were redistilled. The latter two amines were converted to the hydrochlorides with hydrogen chloride in ether followed by recrystallization from ethanol (without this procedure opalescent aqueous solutions were formed when a solution of the pyridine was acidified). 4-Methoxypyridine was prepared by a known procedure[18] modified slightly by fractional distillation at reduced pressure to minimize decomposition that causes a yellow discoloration, bp 56–56.5° (3.6 mm) (lit.[18] 191–191.5° (760 mm)). *p*-Toluidine and *p*-anisidine were sublimed. Inorganic salts were used without further purification, except for filtration of solutions. Glass-distilled water was deaerated with an aspirator prior to use.

Kinetic Methods. All runs were carried out at 25.0 ± 0.1° and an ionic strength maintained at 1.0 with potassium chloride, unless otherwise noted. Spectra were recorded on a Cary-14 and kinetic runs were performed using a Gilford 2000 recording spectrophotometer and stopped-flow apparatus. The conditions for the kinetic experiments are summarized in Table I.

In the stopped-flow experiments, a pyridine buffer, containing any other nucleophilic reagents (such as sodium acetate or aniline) and made up to 1 *M* in potassium chloride, was placed in a 1.0-ml syringe. A solution of acetic anhydride in acetonitrile (25–100 μl) was added to 20 ml of 1 *M* potassium chloride at 25°, and this was used to fill the second 1.0-ml syringe. The run was generally carried out exactly 30 sec after the addition of acetic anhydride to the salt solution. The dead time of the apparatus is 3–4 msec.

The acetylpyridinium ion was generally monitored at 280 nm. The reactions were carried out in the presence of a large excess of acetate, so that changes in acetate concentrations were negligible. The absorbance followed the characteristic curve for the appearance and decay of an intermediate (eq 4). Conditions were chosen such that λ_2 was sufficiently less than λ_1 (at least five–ten times) so that its decay (λ_2) could be plotted as a simple semilogarithmic function by using the portion of the curve after 8–10 half-lives of

(14) A. R. Fersht and W. P. Jencks, *J. Amer. Chem. Soc.*, 91, 2125 (1969).
(15) See, for example, B. H. Havsteen, *J. Biol. Chem.*, 242, 769 (1967).
(16) M. A. Suwyn and R. E. Hamm, *Inorg. Chem.*, 6, 2150 (1967).
(17) A. A. Frost and R. G. Pearson, "Kinetics and Mechanism," 2nd ed, Wiley, New York, N. Y., 1961, p 173.
(18) L. Haitinger and A. Lieben, *Monatsh. Chem.*, 6, 279 (1885).

Table I. Experimental Conditions for the Determination of the Rates of Reactions of Acetic Anhydride with Pyridines and Acylpyridinium Hydrolysis and Acetolysis at 25°[a]

Pyridine	Concentration of pyridine, *M*	Fraction base	Concentration of acetate, *M*	Fraction base	No. of runs
Pyridine	0.20	0.5	$(1.1–3.3) \times 10^{-2}$	0.89	11
Pyridine	0.12	0.5	$4.5 \times 10^{-3} - 5.6 \times 10^{-2}$	0.89	26
Pyridine	0.08–0.16	0.5	1.12×10^{-2}	0.89	11
Pyridine	0.04–0.24	0.5	4.5×10^{-3}	0.89	24
Pyridine	0.022–0.11	0.89	4.04×10^{-3}	0.99	20
Pyridine[b]	0.022–0.11[c]	0.89			15
Pyridine[b]	0.011–0.08[d]	0.89			10
Pyridine	0.030–0.4[e]	0.5			18
Pyridine	0.050–0.3[f]	0.5			16
4-Methyl-[g,h]	0.10	0.5	0–0.07	0.98	28
4-Methyl-[h]	0.18–0.66	0.33			27
4-Methyl-	0.04–0.10	0.50			11
3,4-Dimethyl-[h]	0.01–0.05	0.5			10
3,4-Dimethyl-[g]	0.03	0.47	0.02–0.14	0.99	10
4-Methoxy-[h]	0.01–0.125	0.5			14
4-Methoxy-[g]	0.02	0.5	0.1–0.4	0.99	14

[a] Maintained at ionic strength 1.0 with potassium chloride unless otherwise noted. [b] Determination of second-order rate constant for pyridine attack on acetic anhydride by aniline trapping. [c] 10^{-3} *M p*-anisidine added. [d] 10^{-3} *M p*-toluidine added. [e] Ionic strength of 4.0 maintained with sodium perchlorate. [f] Ionic strength of 6.0 maintained with sodium perchlorate. [g] Determination of the nucleophilic and general base catalysis rate constants for acetate with the acylpyridinium. [h] Determination of the rate constants for attack of pyridine on acetic anhydride, and the spontaneous and pyridine general base catalyzed hydrolysis of the acylpyridinium.

the "formation" reaction (*i.e.*, *ca.* $6.9/\lambda_1$ sec). The value of λ_2 was determined from the slope of the semilogarithmic plot and the preexponential term $\epsilon A_1^{\circ} k_1/(\lambda_1 - \lambda_2)$ from the intercept at $t = 0$.

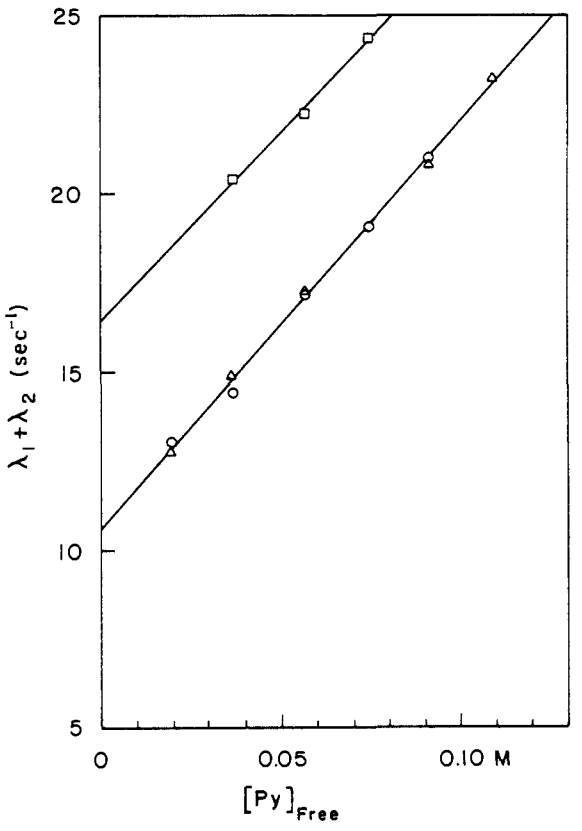


Figure 1. Plot of $(\lambda_1 + \lambda_2)$ against pyridine concentration for the pyridine-catalyzed hydrolysis of acetic anhydride (pyridine corrected for apparent association using K_{ass} of 0.5; see Results): (a) 4×10^{-3} *M* acetate, 2×10^{-4} *M* acetic anhydride, pH 5.5 (△), pH 6.5 (○); (b) 10^{-2} *M* acetate, 5×10^{-4} *M* acetic anhydride, pH 5.5 (□).

The values were then fed back into eq 4 to give λ_1 by a second semilogarithmic plot of

$$-\lambda_1 t = \ln\left(e^{-\lambda_2 t} - \frac{A_B(\lambda_1 - \lambda_2)}{\epsilon A_1^0 k_1}\right)$$

This second calculation was generally performed on an IBM 1130 computer. Rate constants were generally reproducible to ±5%, unless noted otherwise.

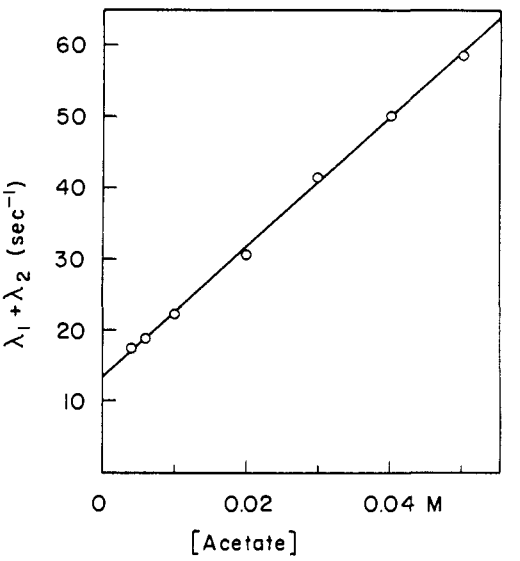


Figure 2. Plot of $(\lambda_1 + \lambda_2)$ against acetate concentration for the pyridine-catalyzed hydrolysis of acetic anhydride at pH 5.5, with 0.12 *M* pyridine buffer (50% free base) and 5×10^{-4} *M* acetic anhydride.

Acetyl-4-methylpyridinium, acetyl-3,4-lutidinium, and acetyl-4-methoxypyridinium ions were monitored at 280, 280, and 275 nm, respectively. The magnitude of the rate constants for these compounds is such that the reactions were ordinarily carried out with $1.0–2.5 \times 10^{-4}$ *M* acetic anhydride and without added acetate under conditions such that λ_1 = "k_1" and λ_2 = "k_2"; *i.e.*, the observed rise and fall in absorbance directly reflect the rate constants for the formation and hydrolysis of the acetylpyridinium ions. The values of λ_1 and λ_2 were evaluated as described above.

Pseudo-first-order rate constants for the reactions with substituted anilines were measured directly from the decrease in absorbance of the aniline.

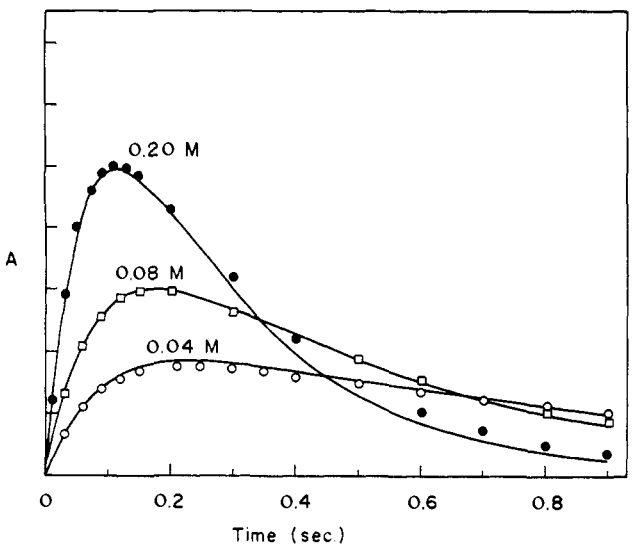


Figure 3. Plots of absorbance at 280 nm (arbitrary units) against time for acetylpyridinium ion appearance and disappearance in the presence of 4×10^{-3} *M* acetate at 25° and ionic strength 1.0 in 50% free base pyridine buffers at the indicated concentrations of total pyridine. The points are experimental and the solid lines are calculated from eq 4 and the derived rate constants.

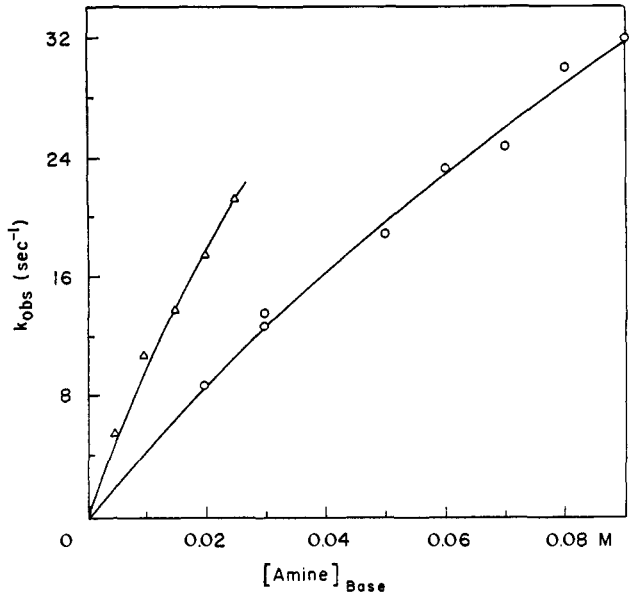


Figure 4. Plots of k_{obsd} against amine concentration, at 25° and ionic strength 1.0, for the attack of 4-methylpyridine at 33 and 50% base (○) and 3,4-lutidine, 50% base (△), on acetic anhydride. Solid curves are calculated based on $k_{ass} = 3.0$, $k_1 = 490\ M^{-1}$ sec^{-1} for the former, and $K_{ass} = 10.0$, $k_1 = 1160\ M^{-1}$ sec^{-1} for the latter amine.

Measurements of pH were carried out with a Radiometer Model 4 pH meter thermostated at 25°.

Kinetic experiments in concentrated sodium perchlorate solutions were carried out by the addition of solid acetylpyridinium chloride[19] to an acid solution or by generation of acetylpyridinium ion *in situ* as follows: 25 μl of acetic anhydride solution in acetonitrile was added to a solution of pyridine buffer–sodium perchlorate in a 1-cm cuvette. The solution was stirred and immediately quenched by the addition of 5 *M* hydrochloric acid to give a final acid concentration of approximately 0.16 *M*. The addition of acid supresses any hydroxide, pyridine, or acetate reaction. The rate of acetylpyridinium ion disappearance was followed at 280 nm and was found to be insensitive to the acid concentration in this range. Some experiments were carried out with the stopped-flow apparatus in 4 and 6 *M* sodium perchlorate containing pyridine buffer, 50% free base. The concentrated salt results in a separation of the rate constants for the formation and breakdown of the acetylpyridinium ion, so that the kinetics can be analyzed in the same way as for the other acetylpyridinium ions and the addition of acetate ion is not required (at low salt concentrations a higher initial concentration of acetic anhydride is required because the rapid hydrolysis results in a low concentration and absorbance of the acetylpyridinium ion).

Activity Coefficients. The effect of sodium perchlorate on the activity coefficient of pyridine was determined by spectrophotometric measurements of the distribution of 0.002 *M* pyridine between hexane and water. The hexane–water distribution ratio was found to be 0.55, 0.55, 0.56, 0.57, 0.59, 0.67, 0.91, and 1.34 in 0, 0.2, 0.4, 0.6, 1.0, 2.0, 4.0, and 6.0 *M* sodium perchlorate, respectively; in 1 *M* potassium chloride it was found to be 0.76.

Absorption Spectrum. The molar extinction coefficient at 280 nm of the acetylpyridinium ion in 1 *M* potassium chloride was determined to be 3240 by substitution of the rate constants into eq 4. The same value was obtained in 4 and 6 *M* sodium perchlorate solution. The rest of the spectrum, at >270.5 nm, was obtained by comparing the maximum absorbance at a given wavelength with that at 280 nm in a series of runs with reaction mixtures of identical composition.

In 9 *M* sodium perchlorate the rate of hydrolysis is sufficiently slow that the spectrum of acetylpyridinium ion can be measured directly. A solution of acetic anhydride was added to a cuvette containing pyridine buffer and sodium perchlorate and quickly acidified. The blank was an identical solution in which the reaction had been allowed to proceed to completion. The difference spectrum was immediately recorded with a Cary 14 spectrophotometer, first with decreasing and then with increasing wavelength, and was corrected for the small amount of hydrolysis which had occurred during the run. The concentration of acetylpyridinium ion was determined from its extinction at 280 nm and this quantity was used to correct the observed spectrum for the decrease in pyridinium ion concentration compared to the blank (caused by acetylation of pyridine).

Results

Kinetics. Typical experimental results showing the appearance and disappearance of the absorption of acetylpyridinium ion at 280 nm at different concentrations of pyridine are shown in Figure 3. The points represent experimental values and the solid lines are theoretical curves calculated from the derived rate constants and eq 4. The procedure for evaluation of the kinetic constants from the kinetic curves has been described above and representative plots of the data are shown in Figures 1 and 2.

The pseudo-first-order rate constants for attack of 4-methylpyridine and 3,4-lutidine on acetic anhydride to give the corresponding acylpyridinium ions are plotted against amine concentration in Figure 4. The plots show the characteristic downward curvature observed in other reactions of pyridines that can be accounted for by an activity coefficient effect or a self-association which becomes significant at higher amine concentrations.[20,21] The effect is more important for methyl-substituted pyridines and the data can be satisfactorily described by apparent association constants of 0, 0.5,[20] 3.0,[20] and 10 M^{-1} for 4-methoxypyridine, pyridine, 4-methylpyridine, and 3,4-lutidine, respectively. The solid lines in Figure 4 have been calculated as described previously[20] using these association constants.

(19) A. K. Sheinkman, S. L. Portnova, Yu. N. Sheinker, and A. N. Kost, *Dokl. Akad. Nauk SSSR*, 157, 1416 (1964); *Dokl. Chem.*, 817 (1964).

(20) A. J. Kirby and W. P. Jencks, *J. Amer. Chem. Soc.*, 87, 3209 (1965).

(21) (a) R. J. L. Andon, J. D. Cox, and E. F. G. Herington, *J. Chem. Soc.*, 3188 (1954); (b) N. Ibl, G. Dändliker, and G. Trümpler, *Helv. Chim. Acta*, 37, 1661 (1954).

Table II. Rate Constants for the Nucleophilic Attack of Pyridines on Acetic Anhydride[a]

Amine	pK_a	k_1, M^{-1} sec^{-1}
Pyridine	5.51	84
4-Methylpyridine	6.33	490
3,4-Lutidine	6.79	1160
4-Methoxypyridine	6.82	935

[a] At 25° and ionic strength 1.0, maintained with potassium chloride.

the pyridine reaction (k_{2py}) represents general base catalysis by a substituted pyridine of the hydrolysis of the corresponding acylpyridinium ion. Rate constants were evaluated according to the procedure described above and illustrated in Figures 1–4 and from experiments carried out under the conditions described in Table I. The rate constants for reactions of pyridines have been corrected for any significant association or activity coefficient effect under the conditions of the experiments;

Table III. Summary of Rate Constants for the Reactions of Nucleophilic and General Base Reagents with Acetylpyridinium (AP^+), Acetyl-4-methylpyridinium ($AMeP^+$), Acetyl-3,4-lutdinium (AL^+), and Acetyl-4-methoxypyridinium ($AMeOP^+$) Ions[a]

Compd	pK_a	AP^+	$AMeP^+$	AL^+	$AMeOP^+$
Water (k_0)[b]	−1.75	6.9	2.30	1.50	0.40
Acetate ($k_{2_{AcO^-}}$) (general base)[f]	4.61[c]		(0.7)[d]	3.06	1.1
Acetate (k_{-1})[f] (nucleophilic)[f]	4.61[c]	910	262	120	30
Pyridine (k_{2py})[f]	5.51[c]	18 ± 6			
4-Methylpyridine[f]	6.33[e]		10		
3,4-Lutidine[f]	6.79[e]			18	
4-Methoxypyridine[f]	6.82[e]				2.8

[a] At 25° and ionic strength 1.0, maintained with potassium chloride. [b] Pseudo-first-order rate constant for the pH-independent hydrolysis; k_0, sec^{-1}. [c] From ref 6. [d] Very approximate value. [e] This study, from measurements of the pH of partially neutralized solutions. [f] k_2, M^{-1} sec^{-1}.

The kinetic constants for the reactions of substituted pyridines with acetic anhydride and for the nucleophilic and general base catalyzed reactions with the substituted acylpyridinium ions are summarized in Tables II and

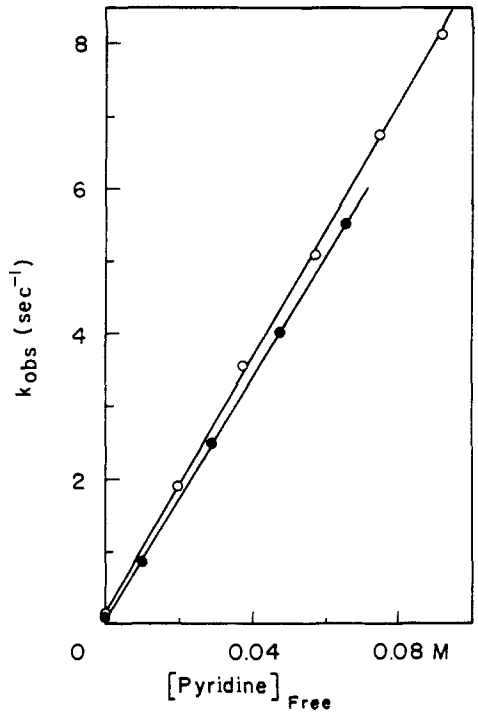


Figure 5. Plot of k_{obsd} against pyridine concentration (corrected for association) for the disappearance of 10^{-3} M p-toluidine (●, 300 nm) and 10^{-3} M p-anisidine (○, 296.5 nm) at pH 6.5, 25°, and ionic strength 1.0, with 10^{-4} M acetic anhydride.

III. The acetate reactions in Table III are for general base catalyzed hydrolysis ($k_{2_{AcO^-}}$) and for the nucleophilic reaction to regenerate starting material (k_{-1}); this correction is only a few per cent at the lower concentrations and does not depend on the assumed mechanism of the effect.

Nucleophilic Catalysis of Transacylation. The rate constants for the pyridine-catalyzed acetylation of p-toluidine and p-anisidine by acetic anhydride are shown as a function of pyridine concentration in Figure 5. The pseudo-first-order rate constants for disappearance of the substituted aniline increase linearly with pyridine concentration and give second-order rate constants of 82 and 86 M^{-1} sec^{-1} for the reactions with toluidine and anisidine, respectively. These values are identical, within experimental error, to the value of k_1 = 84 M^{-1} sec^{-1} which was determined directly for the reaction of pyridine with acetic anhydride to form acetylpyridinium ion (Table II). Furthermore, the reaction rate is independent of the concentration of the acyl acceptor; identical rate constants were obtained at 10^{-3} and 5×10^{-4} M p-anisidine. These results establish that this pyridine-catalyzed acyl transfer reaction proceeds according to eq 10 and 11 with rate-determining formation

$$\mathrm{Py} + \mathrm{Ac_2O} \xrightarrow[k_1]{\text{slow}} \mathrm{AcPy^+} \xrightarrow[k_N[\mathrm{ArNH_2}]]{\text{fast}} \mathrm{AcNHAr} \qquad (10)$$

$$\frac{-\mathrm{d[ArNH_2]}}{\mathrm{d}t} = k_1[\mathrm{Py}][\mathrm{Ac_2O}] \qquad (11)$$

of the acylpyridinium ion followed by a rapid reaction with the acyl acceptor, under the conditions of these experiments.

Reactions in Concentrated Salt Solutions. The rate of hydrolysis of acetylpyridinium ion generated from acetic anhydride and pyridine decreases drastically in concentrated sodium perchlorate solutions. There is a linear relationship between log k and the salt concentration (Figure 6, open circles). The same rate constants are found with acetylpyridinium chloride, synthesized[19] from acetyl chloride and pyridine at −60° (solid circles), confirming the identity of the intermedi-

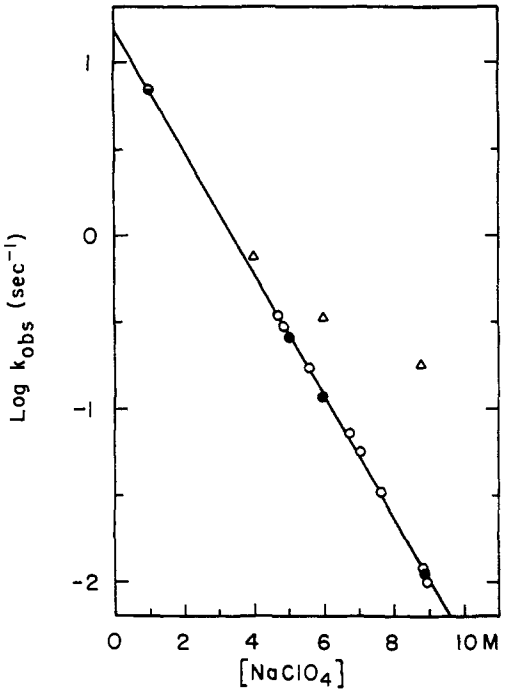


Figure 6. Hydrolysis of acetylpyridinium in concentrated salt solutions at 25°: (a) prepared *in situ* in $NaClO_4$; i, acid conditions (○); ii, in pyridine buffer (50% free base) extrapolated to zero buffer concentration (△); (b) solid acetylpyridinium chloride added to acidified $NaClO_4$ (●); (c) hydrolysis in ionic strength 1.0 solutions with added KCl instead of $NaClO_4$ in pyridine buffer (50% free base, pH 5.5), extrapolated to zero buffer concentration (◒).

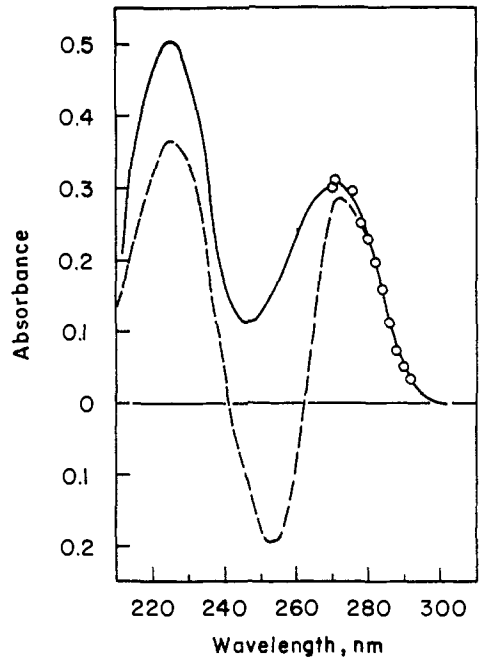


Figure 7. Spectrum of acetylpyridinium ion: ----, observed difference spectrum of equimolar acetylpyridinium and pyridinium, 6.87×10^{-5} *M* in acidified 9 *M* $NaClO_4$; ——, same, corrected for pyridinium ion absorption; ○, spectrum of acetylpyridinium, 6.87×10^{-5} *M* in 1 *M* KCl, pH 5.5, determined kinetically.

ate. Experiments in pyridine buffers (triangles) indicate that a hydroxide ion induced hydrolysis becomes significant in the presence of sodium perchlorate, although it is not significant at the same pH in 1 *M* potassium chloride; *i.e.*, sodium perchlorate causes an increase in the ratio k_{OH^-}/k_0.

The second-order rate constant for the reaction of acetic anhydride with pyridine was found to *increase* in the presence of sodium perchlorate; rate constants of 136 M^{-1} sec^{-1} in 4.0 *M* and 190 M^{-1} sec^{-1} in 6.0 *M* sodium perchlorate represent more than a twofold rate increase compared to the value of 84 M^{-1} sec^{-1} in 1 *M* potassium chloride. Sodium perchlorate was found to have only a small salting out effect on pyridine, insufficient to account for this rate increase, with a Setschenow constant $k_s = 0.03$, based on distribution measurements in the range of 0–1.0 *M* salt concentration. The activity coefficient ratios for pyridine in 4.0 and 6.0 *M* sodium perchlorate, relative to that in 1 *M* potassium chloride, are 1.21 and 1.77, respectively.

Absorption Spectrum of the Acetylpyridinium Ion. By taking advantage of the slow hydrolysis rate in concentrated salt solutions, it is possible to measure the spectrum of the acetylpyridinium ion directly in 9 *M* sodium perchlorate. The dashed and solid lines in Figure 7 show such a spectrum before and after correction for the change in pyridine–pyridinium ion concentration which is caused by acetylation of 1 equiv of pyridine. The absorption of the acetylpyridinium ion intermediate at a number of different wavelengths was measured in a series of identical kinetic experiments and the resulting spectrum above 270.5 nm was normalized with a molar extinction coefficient of 3240 at 280 nm to give the partial spectrum shown by the open circles in Figure 7. It is apparent that there is good agreement between the results of the kinetic and the direct measurements and that salt has little effect on the absorption spectrum. The acetylpyridinium ion has absorption maxima at 272 (ϵ *ca.* 4.4×10^3) and 225 nm (ϵ *ca.* 7×10^3).

Discussion

Mechanism. Four lines of evidence were obtained in this work that establish the intermediacy of the acetylpyridinium ion in the pyridine-catalyzed hydrolysis and acyl transfer reactions of acetic anhydride.

(1) The observation by ultraviolet spectroscopy of the formation and decay of an intermediate in the course of the pyridine-catalyzed hydrolysis of acetic anhydride provides direct confirmation of the earlier kinetic evidence for an acetylpyridinium ion in this reaction. Increasing pyridine concentration causes an increased rate and extent of formation of the intermediate and increasing acetate concentration decreases the amount of intermediate formation by increasing the back reaction to re-form acetic anhydride. At a given concentration of amine, larger concentrations of intermediate are formed more rapidly with the more basic methyl- and methoxy-substituted pyridines. Hydrolysis of the acylpyridinium ions is subject to general base catalysis by acetate and by the corresponding pyridine; the pyridine is a more effective catalyst than the less basic acetate ion in each case (Table III). The rate constants for this catalysis can only be measured accurately for the reactions with the substituted pyridines because of the rapid rate of the uncatalyzed hydrolysis and back-reaction of the unsubstituted acetylpyridinium ion. However, the finding that the ratio of hydrolysis to the exchange of labeled acetate into acetic anhydride in the

presence of pyridine is independent of acetate concentration in the range 0.5–1.0 *M* shows that the hydrolysis of the acetylpyridinium ion must also be catalyzed by acetate.[5]

Koshland has estimated the half-life of acetylpyridinium ion in water to be <1 sec[9] and this is borne out now in a measured half-life of some 100 msec.

(2) The acetylpyridinium ion intermediate exhibits the characteristic absorption spectrum of a six-membered aromatic ring with an acetyl substituent, such as acetophenone or a substituted pyridine. Its absorption maxima at 272 (ϵ *ca.* 4400) and 225 nm (ϵ *ca.* 7000) may be compared with those of 3-acetylpyridinium chloride in ethanol at 269 (ϵ 3900) and 224 nm (ϵ 5800),[22] for example.[23] The same absorption spectrum (above 270.5 nm) and extinction coefficient were obtained from kinetic experiments in 1 *M* potassium chloride, carried out at a series of different wavelengths, and by direct observation in concentrated sodium perchlorate solution.

(3) The rate of hydrolysis of synthetic acetylpyridinium ion can be studied in concentrated sodium perchlorate solutions and was found to be the same as that of the intermediate formed from acetic anhydride.

(4) The fact that the rate of the pyridine-catalyzed acetylation of substituted anilines by acetic anhydride is independent of the nature and the concentration of the aniline demonstrates that these acyl transfer reactions proceed through the rate-determining formation of the acetylpyridinium ion intermediate, followed by a fast reaction with the aniline (eq 10). The high selectivity of the acetylpyridinium ion, in spite of its high reactivity, is demonstrated by the fact that anilines at concentrations on the order of 10^{-3} *M* compete effectively with 55 *M* water for reaction with this intermediate. The identity of the rate constants for acetylpyridinium ion formation obtained from the kinetics of the hydrolysis reaction and from the pyridine-catalyzed acetylation of two substituted anilines (eq 10) provides a confirmation of the accuracy of the methods used to evaluate the rate constants for pyridine-catalyzed hydrolysis of acetic anhydride and even stronger evidence for a common intermediate in the hydrolysis and transacylation reactions.

The remarkable efficacy of pyridines as catalysts for transacylation reactions of acetic anhydride is a consequence not only of their relatively unhindered tertiary amine structure which, as mentioned above, cannot lose a proton to form a stable amide upon acylation, but also of the selectivity of the reactive acetylpyridinium ion intermediates toward nucleophiles and of the peculiar dependence of the rate constants for the reaction of nucleophilic reagents with highly reactive acylating agents on the p*K* of the nucleophile. The reactivity of substituted pyridines with acetic anhydride shows a large dependence upon their basicity (Table II) with a β value of 0.87, similar to that for other reactive acyl donors, in a plot of log *k* against p*K*. However, more basic amines exhibit a marked leveling off of this dependence of nucleophilic reactivity upon basicity; for primary amino acids there is a much smaller dependence upon basicity with a β value of roughly 0.25.[24] A similar leveling off is seen with other highly reactive acyl donors and indicates that the transition state is reached at an early point along the reaction coordinate with the more basic amines.[6] The pyridines fall just at the break point of this structure–reactivity correlation, so that they take maximal advantage of the dependence of reactivity upon basicity and still produce a highly reactive (and selective) acylating agent upon acylation. At a relatively low pH value, at which the alkaline hydrolysis of acetic anhydride will be minimal, the pyridines are much more reactive than more basic amines which are mostly in the unreactive, protonated form. It is of interest that the pyridines are appreciably more reactive than the more basic imidazole molecule toward acetic anhydride and other highly reactive acylating agents; imidazole is the more reactive nucleophile toward less reactive acylating agents, such as phenyl acetate.[6,25]

Kinetics. The kinetic treatment of the reactions of acetic anhydride with substituted pyridines more basic than pyridine itself is straightforward, because the formation of the substituted acetylpyridinium intermediate is fast and its hydrolysis is slow relative to acetylpyridinium ion. Consequently, the reactions can be studied in the absence of added acetate ion, the formation and hydrolysis reactions are moderately well separated, and a near-maximal amount of intermediate is formed at high nucleophile concentrations. Under these conditions the initial rise in absorption reflects the rate constant for intermediate formation and the subsequent fall reflects hydrolysis of the intermediate. By varying the concentration of the appropriate pyridine the spontaneous rate and pyridine general base catalyzed rate of hydrolysis may be readily obtained, and from the initial parts of the absorbance *vs.* time changes the second-order rate constants for the attack of the pyridine on acetic anhydride are also readily obtained.

The kinetic treatment is more complex for the pyridine reaction because the lower nucleophilic reactivity of pyridine and the higher reactivity of the acetylpyridinium ion lead to a situation in which there is overlapping of the rate constants and the rise and fall in absorption do not directly reflect the rate constants for the formation and hydrolysis, respectively, of the intermediate. Because of this high reactivity, the amount of acetate released in the initial reaction is sufficient, even at concentrations of 2–5 $\times$ 10^{-4} *M*, to react significantly with the intermediate to regenerate acetic anhydride and cause a departure from pseudo-first-order kinetics, so that these experiments were carried out in the presence of an excess of added acetate in order that the acetate concentration and the rate of the back reaction would remain constant. It should be noted that the equation for the time course of the concentration of an intermediate formed in an irreversible first-order process, *e.g.*

$$A_B = \frac{\epsilon A_1^0 k_1}{k_2 - k_1} (e^{-k_1 t} - e^{-k_2 t})$$

is symmetrical with respect to the interchange of k_1 and

(22) M. L. Swain, A. Eisner, C. F. Woodward, and B. A. Brice, *J. Amer. Chem. Soc.*, 71, 1341 (1949).

(23) G. A. Olah and P. J. Szilagyi (*ibid.*, 91, 2949 (1969)) have recently reported the nmr spectrum of acetylpyridinium ion in "magic acid."

(24) D. M. Brouwer, M. J. Vlugt, and E. Havinga, *Proc. Kon. Ned. Akad. Wetensch. Ser. B*, 61, 141 (1958).

(25) D. M. Brouwer, M. J. Vlugt, and E. Havinga, *ibid.*, 60, 275 (1957).

k_2 *apart from* the amplitude or preexponential term. Thus, unless the extinction coefficient, ϵ, is known, solution of the equation for k_1 and k_2 does not unambiguously assign the rate constants to the formation and decomposition reactions, respectively. To unambiguously assign the values, either the extinction coefficient must be known or chemical evidence must be invoked, such as the dependence of k_1 and k_2 upon the concentrations of the reactants. For acetylpyridinium formation, for example, the value of k_1 must be strongly dependent on pyridine concentration and approach zero at zero pyridine concentration, whereas the k_2 term is relatively insensitive to pyridine concentration. At low concentrations of pyridine, hydrolysis of the intermediate is considerably faster than its formation, so that the observed absorbance changes are small and are directly proportional to the pyridine concentration. Under these conditions the decay of the absorption reflects primarily the rate constant for *formation* of the intermediate and the rise in absorption reflects primarily the rate constants for *decomposition* of the intermediate. The experimental results, particularly the agreement of the rate constant for acetylpyridinium ion formation obtained from the kinetics of the directly measured appearance and hydrolysis of the intermediate with that obtained from the pyridine-catalyzed acetylation of anilines, indicate that the treatment described above provides a satisfactory method for extracting the individual rate constants from the observed time dependence of the absorbance changes.

The Effect of Concentrated Sodium Perchlorate. The striking inhibition of the hydrolysis of acetylpyridinium ion by concentrated sodium perchlorate is similar to that observed previously for the pH-independent hydrolysis of other acyl compounds, such as acetylimidazolium ion[26] and acetic and succinic anhydrides.[27,28] The rate decrease is even larger than that for acetylimidazolium ion, amounting to 700-fold going from 1 *M* potassium chloride to 9 *M* sodium perchlorate. In marked contrast to the hydrolysis reactions, the rate of the nucleophilic reaction of pyridine with acetic anhydride is *increased* by sodium perchlorate; in 4 *M* sodium perchlorate the rate is increased 1.6-fold, compared to a tenfold inhibition of the spontaneous hydrolysis of acetic anhydride at this salt concentration.

The salt effects may be described formally in terms of activity coefficients and salting out constants of the reactants and transition states according to the Brønsted–Bjerrum equation

$$\frac{k_{salt}}{k_{H_2O}} = \frac{f_A f_B}{f^{\pm}}$$

(in which the activity coefficients refer to the reactants A and B and the transition state) and the related Setschenow equations

$$\log k_{salt}/k_{H_2O} = k_s^{rate}[salt]$$
$$k_s^{rate} = k_s^{A} + k_s^{B} - k_s^{\pm}$$

in which the salting out constants k_s are measures of the change in free energy of the reactants and transition state caused by the salt. The decrease in the logarithm of the hydrolysis rate of acetylpyridinium ion is linear with respect to salt concentration (Figure 6) with a slope of -0.35, compared to -0.30 for acetylimidazolium ion and -0.24 for acetic and succinic anhydrides. The activity coefficients of acetylpyridinium and acetylimidazolium ions are not easily measured, but acetic anhydride is salted in by sodium perchlorate[26] with a value of $k_s = -0.1$; the value of k_s for pyridine is 0.03. Thus, the $k_s^{\pm}$ for acetic anhydride hydrolysis is $-0.1 - (-0.24) = 0.14$ (following the usual practice of neglecting the effect of salt on the unknown number of water molecules involved in the reaction). The $k_s^{\pm}$ value for the reaction of pyridine with acetic anhydride is $0.03 - 0.1 - 0.07 = -0.14$, which is equal but *opposite* to that for hydrolysis. The effects of sodium perchlorate on the activity coefficients and free energies of the transition states for the two reactions are, therefore, also almost equal and opposite.

We will defer a detailed discussion of these remarkable salt effects for a later publication and only wish to point out here that (a) although it is certain that the decrease in the activity of water caused by concentrated salt solutions contributes to the inhibition of hydrolysis and to the opposite effects of salts on the reactions with water and with pyridine, the inhibition of hydrolytic reactions of this kind by concentrated salt solutions cannot be accounted for by effects on water activity alone,[26,27] and (b) salt effects on the transition state of the pyridine–acetic anhydride reaction, in which there is a large development of positive charge on the attacking amine, may be similar to those on the reacting acyl compounds.

Structure–Reactivity Relationships. The equilibrium constants for acetylpyridinium ion formation from acetic anhydride and substituted pyridines were calculated from the rate constants for the reactions in the two directions and are summarized in Table IV, along with the corresponding value for the reaction with imidazole. The free energies of hydrolysis were calculated from the known free energy of hydrolysis of

Table IV. Summary of Free Energies of Hyrolysis and Equilibrium Constants for Acetyl Transfer of Acetylpyridinium Ions at 25°, Ionic Strength 1.0

Compd	K^a $\frac{[AcPy^+][AcO^-]}{[Py][Ac_2O]}$	$-\Delta F°$ [b] kcal/mol	$-\Delta F_{pH7}°$ [c] kcal/mol
Acetylpyridinium	0.092	18.31	23.63
Acetyl-4-methylpyridinium	1.86	17.67	21.98
Acetyl-3,4-lutidinium	9.67	17.33	21.17
Acetyl-4-methoxypyridinium	31.2	16.65	20.48
Acetylimidazolium[d]	2950	14.05	14.33

[a] Equilibrium constants for the acetyl transfer reaction, calculated from the kinetic data of Tables II and III. [b] Standard free energy of hydrolysis based on a standard state of 1 *M* concentrations of the reactants and the conjugate acid forms of the products and an activity of pure water of 1.0 [convention I of W. P. Jencks, S. Cordes, and J. Carriuolo, *J. Biol. Chem.*, **235**, 3608 (1960)]. [c] Standard free energy of hydrolysis at pH 7.0 based on a standard state of 1 *M* total stoichiometric reactants and on an activity of pure water of 1.0 (W. P. Jencks, S. Cordes, and J. Carriuolo, *ibid.*, **235**, 3608 (1960), convention III). In all cases pK_a' values at 25° and ionic strength 1.0 were used. [d] Data from ref 11 and 12 and a pK_a of 3.86 for acetylimidazolium (D. G. Oakenfull, unpublished results).

(26) S. Marburg and W. P. Jencks, *J. Amer. Chem. Soc.*, **84**, 232 (1962).
(27) C. A. Bunton, N. A. Fuller, S. G. Perry, and I. H. Pitman, *J. Chem. Soc.*, 4478 (1962).
(28) C. A. Bunton, J. H. Fendler, N. A. Fuller, S. Perry, and J. Rocek, *ibid.*, 5361 (1963).

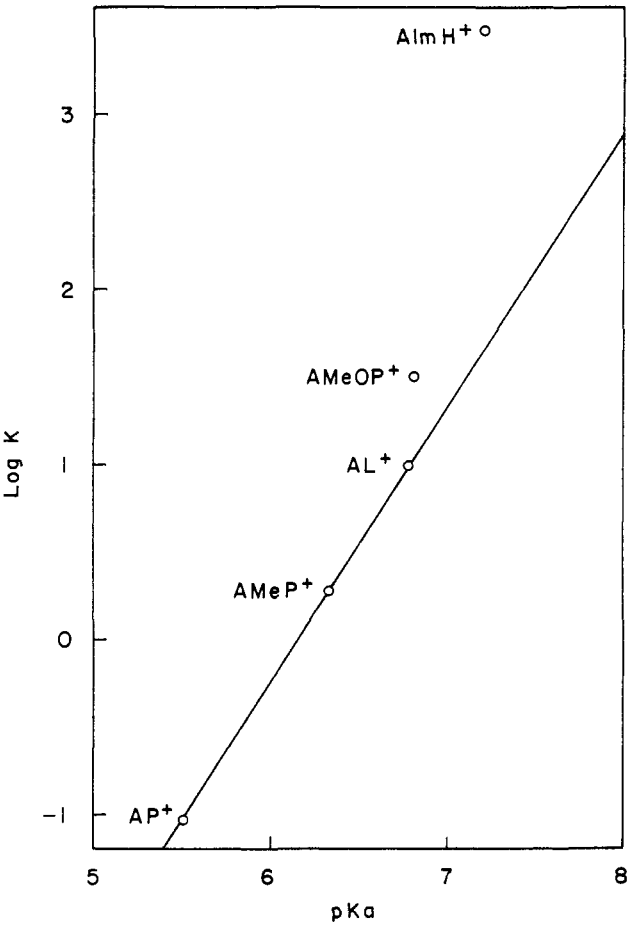


Figure 8. The dependence of the equilibrium constants for the formation of substituted acetylpyridinium and acetylimidazolium ions, $K = [AcN^+<][AcO^-]/[N<][Ac_2O]$, upon basicity of the parent amine. The line has a slope of 1.58.

acetic anhydride[11] and are expressed in Table IV for the ionic species of the reaction shown in eq 12 and for

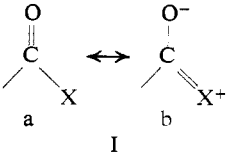
$$AcPy^+ + H_2O \rightleftharpoons AcOH + HPy^+ \qquad (12)$$

hydrolysis at pH 7.0. For acetyl-4-methylpyridinium ion the equilibrium constant for the reaction with *p*-nitrophenol was estimated from the rate constant for the reaction of 4-methylpyridine with *p*-nitrophenyl acetate, the ratio of the rate constants for the reactions of water and *p*-nitrophenolate ions with acetyl-4-methylpyridinium ion,[29] and the rate constant for the hydrolysis of this compound. Based on the known free energy of hydrolysis of *p*-nitrophenyl acetate[12] the free energy of hydrolysis of acetyl-4-methylpyridinium ion was calculated to be −17.4 kcal/mol, in reasonable agreement with the value of −17.67 calculated from the reaction with acetic anhydride.

The free energies of hydrolysis of the acetylpyridinium ion, expressed according to eq 12, are larger than that of acetic anhydride (−15.7 kcal/mol); *i.e.*, an acylated tertiary amine, in which the normal amide resonance does not occur, is less stable than the acylated oxygen of acetic anhydride, which has a similar p*K* but still presumably provides some resonance stabilization to the acyl group.

(29) A. R. Fersht and W. P. Jencks, *J. Amer. Chem. Soc.*, **92**, 5442 (1970).

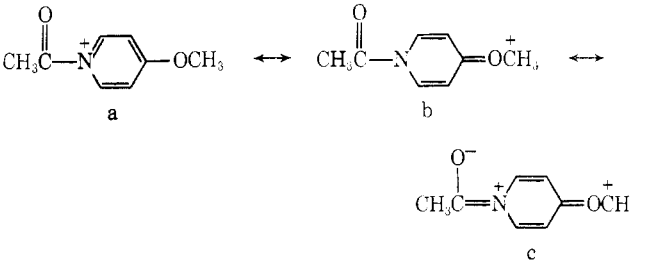
A logarithmic plot of the equilibrium constants for the reactions of pyridine and methyl-substituted pyridines with acetic anhydride to form the corresponding acylpyridinium ions against the basicity of the pyridine has a slope, β, of 1.6 (Figure 8). The corresponding reactions with 4-methoxypyridine and imidazole show positive deviations from this plot, indicating that the products of these reactions are more stable than expected from their basicity. The slope of 1.6 is very similar to the value of 1.7 for the analogous reaction of a series of phenolate ions with acetylimidazolium ion[12] and indicates that the stability of acylpyridinium ions and of phenyl acetates is affected in almost the same way by substituents on the atom to which the acyl group is attached. In both reactions, the substituent sensitivity for addition of the acyl group is greater than for addition of a proton; *i.e.*, the effective electronegativity of the acyl group is greater than that of the proton. A similar result is found for a series of acetanilides.[30] The result with the pyridine compounds is of interest in that it shows that electron donation by resonance to the carbonyl group from the atom which is acylated (I) is not required in order for this high



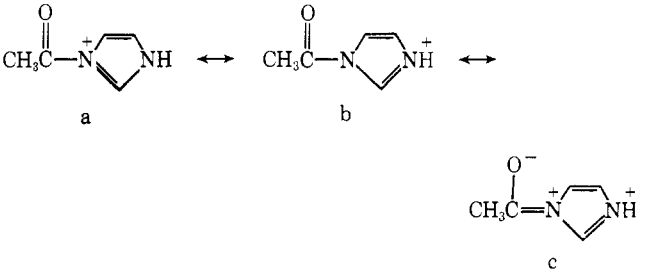
I

sensitivity and effective electronegativity to exist.

The positive deviations of the acetyl derivatives of 4-methoxypyridine and imidazole from the basicity relationship of Figure 8 suggest that these compounds have additional stabilization by resonance electron donation to the carbonyl group from a *p*-methoxy group or imidazole nitrogen (II and III). These deviations are also evident in the free energies of hydrolysis expressed according to eq 12 and indicate that this resonance stabilization is more important for the acyl com-



II



III

(30) W. P. Jencks, B. Schaffhausen, K. Tornheim, and H. White, in preparation.

pounds than for the addition of a proton to the basic nitrogen atom, for which the contributing resonance forms IIc and IIIc are not available. The extra stabilization attributed to this resonance amounts to about 0.7 kcal for the 4-methoxypyridinium ion and 2.7 kcal for acetylimidazolium ion.

Satisfactory linear free-energy correlations of reaction rate with the basicity of the leaving group exist for the nucleophilic reactions of acetate and water with methyl-substituted and unsubstituted acetylpyridinium ions, but acetyl-4-methoxypyridinium and acetylimidazolium ions deviate from this relationship, as in the equilibrium correlations (Figure 9). The slope of -0.7 for the correlation line of the acetate reaction is large and indicates a high degree of selectivity of the nucleophilic reagent toward even such highly reactive acylating agents. We interpret the smaller slope of -0.5 for the water reaction as an indication that this reaction may be less selective, even though slower, because general base catalysis converts the water molecule to an incipient hydroxide ion with a lower selectivity approaching the still lower selectivity of hydroxide ion as a nucleophile.[6,31] This relatively low selectivity of the water reaction provides an explanation for the surprising fact, which has been noted previously,[32] that the ratio of the second-order rate constants for the acetate and water reactions is larger for the more reactive acetylpyridinium ion (k_{AcO^-}/k_{H_2O} = 7300) than for acetylimidazolium ion (k_{AcO^-}/k_{H_2O} = 29), so that if only these reactions are compared the acetylpyridinium ion appears to be more selective than the acetylimidazolium ion. This relatively low reactivity of acetylpyridinium ion toward water is still another factor which makes pyridine an effective catalyst for acyl transfer reactions.

Because of its similarity to the hydroxide reaction, the possibility of charge delocalization by general base catalysis, and its uncertain mechanism, we believe that the water reaction is not the ideal choice for a "standard" reaction of an uncharged nucleophile. Indeed, if the data of a recent study of electrostatic effects in nucleophilic reactions with charged acyl compounds[33] are plotted against the rate constants for reaction with methoxyamine or ethylenediamine, instead of water, as a "standard," significant electrostatic effects are seen for anionic nucleophiles, including hydroxide and trifluoroethoxide ions. The net charge of the transition state for the amine reactions is zero and there is relatively little delocalization of charge from the reaction center; the similar β values for attacking and leaving groups suggest that the changes in the charge density on the attacking and leaving groups are similar.[6,29]

The negative deviations of acetyl-4-methoxypyridinium and acetylimidazolium ions from the correlations of Figure 9 reflect the special resonance stabilization of the starting materials which has been noted above and suggest that a large part of this stabilization is lost in the transition state because of bond formation to the carbonyl group, more in the case of the acetate than the water reaction. The same conclusion may be reached by correlations with the Hammett substituent

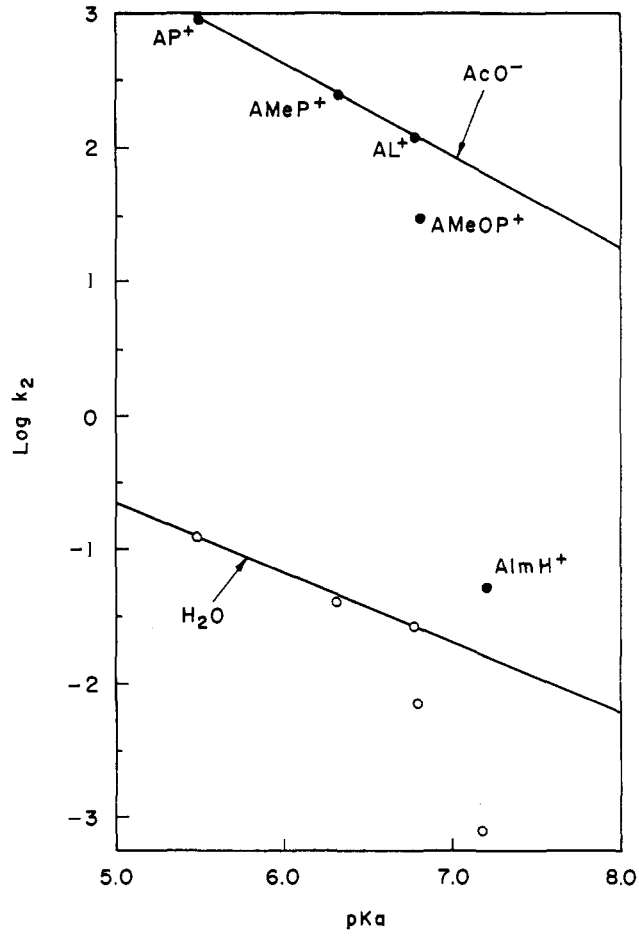


Figure 9. The dependence of the rate constants for the reactions with acetate (●) and water (○) of a series of acetylpyridinium and acetylimidazolium ions on the basicity of the leaving amine. The lines have slopes of 0.68 and 0.50.

constants σ and σ^+. Although the basicity of pyridines follows σ more closely than σ^+, the reactivity and stability of the acylpyridinium ions are better correlated by σ^+, as expected if electron donation by resonance is important in the latter series. The unexpectedly high rates of hydrolysis of benzoylimidazole and trimethylacetylimidazole and their conjugate acids probably reflect decreases in resonance stabilization caused by these bulky acyl groups.[26,34]

The rate constants for general base catalysis of the hydrolysis of acylpyridinium and acetylimidazolium ions by water, acetate, and the parent amine give Brønsted plots with slopes, β, of 0.30–0.35. This value of β is smaller than that of 0.47 for the general base catalyzed hydrolysis of ethyl dichloroacetate,[35] but is similar to the value of 0.32 for the more reactive phenyl dichloroacetate,[36] indicating that the value of β shows the expected decrease as the pK of the leaving group decreases.

Gold and Jefferson[2] have postulated that the high reactivity of acetylpyridinium ion is due to a favorable entropy term, the destruction of the positive charge on hydrolysis leading to a decrease of order in the solvent. However, we believe that the high reactivity is due to an

(31) B. M. Anderson, E. Cordes, and W. P. Jencks, *J. Biol. Chem.*, 236, 455 (1961); B. D. Batts and V. Gold, *J. Chem. Soc. A*, 984 (1969).

(32) S. L. Johnson, *J. Phys. Chem.*, 67, 495 (1963). The ratios given here are based on second-order rate constants for the nucleophilic reactions with acetate ion[11] and with water.[13]

(33) B. Holmquist and T. C. Bruice, *J. Amer. Chem. Soc.*, 91, 2982, 2985 (1969).

(34) H. A. Staab, *Chem. Ber.*, 89, 2088 (1956); J. A. Fee and T. H. Fife, *J. Org. Chem.*, 31, 2343 (1966).

(35) W. P. Jencks and J. Carriuolo, *J. Amer. Chem. Soc.*, 83, 1743 (1961).

(36) A. R. Fersht and A. J. Kirby, *ibid.*, 89, 4853 (1967).

electronic destabilization of the acetylpyridinium ion, *i.e.*, an internal energy effect rather than a solvation effect, in the two following ways.

(a) The positive charge on the nitrogen atom destabilizes the partial positive charge on the sp^2 carbonyl carbon atom. The very large sensitivity of the thermodynamic and kinetic stability of the acylpyridinium ions to the polar character of this atom, as measured by the basicity of the parent pyridine, supports this assertion.

(b) In spite of the existence of some resonance stabilization by conjugation with the aromatic ring, the loss of the normal resonance stabilization of amides by electron donation from the amide nitrogen atom (I) causes a marked destabilization of acetylpyridinium ions. It has been estimated that the resonance structure Ib contributes about 40% to the structure and some 20 kcal/mol of resonance stabilization to simple aliphatic amides.[37] Solvent structure effects are unlikely to give rise to large free-energy changes because of the tendency for mutual compensation of entropy and enthalpy changes upon solvent rearrangement.[38]

Acknowledgment. We are indebted to Mrs. Karin Salvesen for carrying out the activity coefficient measurements.

(37) P. Haake, W. B. Miller, and D. A. Tyssee, *J. Amer. Chem. Soc.*, **86**, 3577 (1964);
L. C. Pauling, "Nature of the Chemical Bond," 3rd ed, Cornell University Press, Ithaca, N. Y., 1960, p 197.

(38) See, for example, L. G. Hepler, *J. Amer. Chem. Soc.*, **85**, 3089 (1963).

Reactions of Nucleophilic Reagents with Acylating Agents of Extreme Reactivity and Unreactivity. Correlation of β Values for Attacking and Leaving Group Variation[1]

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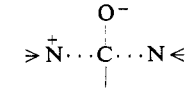
Abstract: Rate constants are reported for the reactions of a series of nucleophilic reagents with substituted N-acetylpyridinium ions. Logarithmic plots of reaction rate against the pK of the nucleophile for oxygen nucleophiles are curved and for nitrogen nucleophiles show an initial slope of $\sim$0.9, followed by a leveling off. Nevertheless, a high degree of selectivity with respect to factors other than basicity is maintained for reactions with rate constants of up to 3.3×10^8 M^{-1} sec^{-1}. Rate constants for nucleophilic reactions with methyl acetate, trifluoroethyl acetate, and acetic acid, calculated from the equilibrium constants and the rate constants for the reverse reaction, show a large dependence on basicity with β values of $\sim$1.5, compared to 1.7 for the complete reaction. A similar β value is obtained for leaving group variation in the reactions of unreactive esters with 4-methylpyridine and *p*-nitrophenolate ion. Acyl transfer reactions exhibit a rough correlation of β values for the nucleophile with the (negative) β values for the leaving group. Proton transfer from the nucleophile has a large effect on the rate for reactions with acetate esters less reactive than phenyl acetate.

We report here a study of structure–reactivity relationships for the reactions of a series of substituted N-acylpyridinium ions with nitrogen and oxygen nucleophilic reagents. These and other results have made possible the correlation of rate constants for reactions of nucleophiles with acyl compounds over a wide range of reaction rates, from 10^{-20} to 10^8 M^{-1} sec^{-1}. The experiments were carried out for the following three reasons.

(1) We wished to determine whether such extremely reactive *acylating agents* as the acetylpyridinium ions display a selectivity toward nucleophiles similar to that exhibited by less reactive acyl compounds; *i.e.*, to what extent do the transition states for reactions of these compounds occur early along the reaction coordinate (a "Hammond postulate" [2] type of effect) and what is the nature of any resulting change in the nature of the transition state?

(2) We wished to determine whether these compounds exhibit a sharp leveling off in their sensitivity toward the basicity of the *nucleophile* with strongly basic nucleophiles, as is observed with oxygen esters with good leaving groups.[3]

(3) We hoped to obtain further information about the nature of transition states and structure–reactivity correlations for acyl transfer reactions in general by examining a series of symmetrical reactions with the same entering and leaving atom. If proton transfer does not occur, the reactions of nitrogen bases with acylpyridinium ions have a symmetry, analogous to that in the reactions of esters with alkoxide ions,[3] such that with attacking and leaving groups of similar pK the transition state should be almost symmetrical.



Although there is no question but that tetrahedral ad-

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(2) G. S. Hammond, *J. Amer. Chem. Soc.*, 77, 334 (1955); J. E. Leffler, *Science*, 117, 340 (1953).

(3) W. P. Jencks and M. Gilchrist, *J. Amer. Chem. Soc.*, **90**, 2622 (1968).