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Electrostatic Acceleration of Enolization in Cationic Ketones

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Abstract: Rate constants for the water, acetate, and hydroxide ion-catalyzed enolizations of the cationic ketones 2-acetyl-1-methylpyridinium ion (**3**) and 1-methyl-8-oxo-5,6,7,8-tetrahydroquinolinium ion (**4**) have been measured at 25 °C and compared with those reported for the enolization of 2-acetyl-3,4-dimethylthiazolium ion (**2**) (Halkides, C. J.; Frey, P. A.; Tobin, J. B. *J. Am. Chem. Soc.* **1993** *115*, 3332). For **3**, k_{H_2O}, k_{OAc}, and k_{OH} are 1.32 ± 0.21 s^{-1}, (2.82 ± 0.95) × 10^{-2} M^{-1} s^{-1}, and 187 ± 50 M^{-1} s^{-1}, respectively. The corresponding values for **4** are 9.17 ± 0.24) × 10^{-2} s^{-1}, (4.32 ± 0.18) × 10^{-4} M^{-1} s^{-1} and 10.9 ± 0.2 M^{-1} s^{-1}, respectively. The values of pK_a for **3** and **4** are 11.13 and 11.90, respectively, at 25 °C. The hydration equilibrium constant K_h for **3** is 0.084 ± 0.004 at 25 °C. The correlation of hydration constants for ketones with σ^* (Greenzaid, P.; Luz, Z.; Samuel, D. *J. Am. Chem. Soc.* **1967** *89*, 749) allow the hydration constants for **2** and **3** to be used in the estimation of σ^* values for the substituents. The σ^* values are 1.68 and 1.02 for 2-(*N*-methylthiazolium) and 2-(*N*-methylpyridinium), respectively. Use of the σ^* values to estimate the inductive effects of these substituents on the pK_as of **2** and **3** allows the inductive effects to be separated from the through-space electrostatic effects. The inductive effects are estimated to contribute 4.2- and 1.9-log units to lowering the pK_as of **2** and **3**, respectively. From this and the measured pK_a of 11.1 for **3**, the through-space electrostatic contribution to lowering the pK_a is 6.3-log units, or 8.6 kcal mol^{-1} in free energy of enolate stabilization. Assuming the same through-space effect for **2**, its pK_a is estimated to be 8.8. Comparisons of these enolization rates and pK_as with those for ordinary methyl and benzyl ketones indicate that the 2-(*N*-methylpyridinium) substituent in **3** stabilizes the enolate by 11.2 kcal/mol in free energy, and about 8.6 kcal/mol of this can be attributed to through-space electrostatic stabilization. The through-space electrostatic component accounts for a 330-fold enhancement in k_{OH} for **3** compared with a typical methyl ketone (pK_a = 19.3 for acetone). The value of k_{OH} for **2** is 1.1 × 10^6 times that for a typical methyl ketone (pK_a = 19.3) and 70 times that for a neutral methyl ketone exhibiting the pK_a of 8.8 estimated for **2**. The through-space electrostatic effects on the enolate of **2** and the transition state for its formation account for a 2.3 × 10^4-fold enhancement in the enolization rate. The remaining 47-fold rate enhancement for **2** is attributed to inductive effects. The through-space electrostatic effects have important implications for enzymatic catalysis of enolization.

I. Introduction

The α-deprotonation of carbonyl compounds to produce enolate ions has been the topic of many investigations.[1] Correlations of base-catalyzed enolization rate constants (k_B^E) with pK_a values are nearly linear, while displaying a slight curvature owing to a large Marcus intrinsic barrier, $\Delta G_o^{\ddagger}$. However, rate constants that deviate from this curve are not fully understood. In this context, electrostatic rate enhancement has recently been emphasized, its significance being that enolate stabilization provided by electrostatic interactions may be important for enzymatic catalysis of enolization.[2,3]

We have reported the rapid rates of enolization of the methyl ketones 2-acetylthiamine pyrophosphate (**1**) and 2-acetyl-3,4-dimethylthiazolium ion (**2**).[3] The rate constant for hydroxide ion-catalyzed enolization of **1** was found to be >10^6 times that

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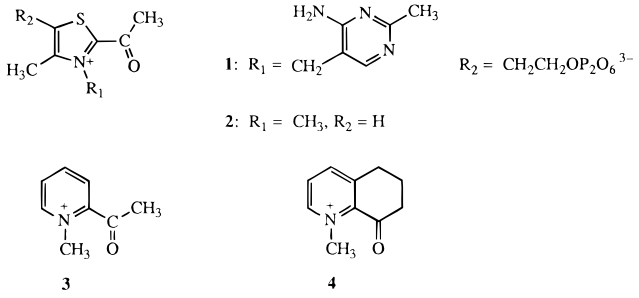

for acetone. The major factor in these enhanced enolization rates has been suggested to be electrophilic stabilization of the enolates by the positively charged thiazolium ring. Electrophilic effects on enolization in cationic ketones have not been separated into through-space and through-bond, inductive components. The through-space component should be especially relevant to enzymatic catalysis of enolization because of the possibility that enolate ions at enzymatic active sites can be stabilized by the side chains of basic amino acids.

Electrophilic contributions to the enolization of cationic ketones can be further understood through knowledge of the thermodynamic acidity constants for their ionizations. However, it is not feasible to measure pK_a values for enolizations of acylthiazolium ions such as **1** and **2** because they rapidly undergo deacylation in basic solutions.[4,5] The 2-acylpyridinium ions **3** and **4** also undergo hydroxide ion-catalyzed deacylations, but at slower rates than 2-acylthiazolium ions. Therefore, pK_as and enolization rates for these compounds can be measured. Intramolecular electrostatic effects may be important in the enolization of 2-acylpyridinium ions. In fact, very fast base-catalyzed enolization of **3** has been reported.[6]

In the present paper we have extended our study of cationic ketones to 2-acetyl-1-methylpyridinium ion (**3**) and 1-methyl-8-oxo-5,6,7,8-tetrahydroquinolinium ion (**4**). The rates of enolate ion formation have been obtained by direct measurements in aqueous hydroxide solutions and by iodine scavenging in acetate buffers. These results along with pK_a determinations in aqueous base have been used to evaluate the influence of electrostatic stabilization on the ionizations of these ketones, and the results have been compared with those for 2-acetyl-3,4-dimethylthiazolium ion **2**. The electrophilic accelerations of enolization by the positively charged nitrogen atoms in **2** and **3** have been separated into inductive and through-space electrostatic components.

II. Experimental Section

Materials. Reagents used in this study were purchased from the Aldrich Chemical Co. Piperidine, 1-methylpiperidine, and ethanolamine were distilled before being used to prepare buffers.

2-Acetyl-1-methylpyridinium Iodide (3). 2-Acetylpyridine (2.00 g, 16.5 mmol) was refluxed in excess methyl iodide (2.43 g, 17.1 mmol) for 24 h.[6] Removal of methyl iodide under vacuum gave light green crystals, which were recrystallized three times from ethanol to give 0.94 g of yellow crystals (22% yield), mp 160−161 °C (lit.[6] mp 160−162 °C): ^{1}H NMR (CD_3CN) 500 MHz) δ 2.76 (s, 3H), 4.36 (s, 3H) 8.10 (t, 1H), 8.38 (d, 1H), 8.65 (t, 1H),8.77 (d, 1H). Anal. Calcd for $C_8H_{10}INO$ C, 36.53; H, 3.83; N, 5.32. Found C, 36.45; H, 3.86; N, 5.17.

1-Methyl-8-oxo-5,6,7,8-tetrahydroquinolinium Iodide (4). The procedure of Thummel et al.[7] was followed with slight variations. Reaction of 2,3-cyclohexenopyridine (13.1 g, 98 mmol) with benzaldehyde (14.1 g, 108 mmol) in the presence of acetic anhydride (17.7 g) at 170 °C gave 8-benzylidene-5,6,7,8-tetrahydroquinoline, which was purified by distillation (bp 119−228 °C at 10 Torr) to give 13.2 g of a viscous liquid. This became a yellow solid upon standing (66% yield) mp 59−62 °C (lit.[7] 62−64 °C). Ozonolysis of this compound (4.50 g, 20.5 mmol) was performed in 100 mL of methanol using a T-23 Welsbach Ozonator. Purification by column chromatography (silica gel; CH_2Cl_2) gave 0.98 g of a yellow solid (32% yield) the identity of which was verified by its ^{1}H NMR spectrum.

Methylation of 8-oxo-5,6,7,8-tetrahydroquinolone (0.23 g, 1.6 mmol) was carried out by stirring with trimethyloxonium tetrafluoroborate (0.21 g, 1.6 mmol) in nitromethane. Following the removal of nitromethane under vacuum, the product was dissolved in water and passed through an anion exchange column (Bio-Rad AG-1 × 8) in the iodide form. Recrystallization from ethanol/diethyl ether gave 0.16 g of dark orange crystals (34% yield), mp 168−171 °C: ^{1}H NMR (CD_3CN, 500 MHz) δ 2.21 (p, 2H), 2.89 (t, 2H), 3.21 (t, 2H), 4.46 (s, 3H), 8.03 (t, 1H) 8.51 (d, 1H), 8.69 (d, 1H); ^{13}C NMR (DMSO-d_6, 127 MHz) δ 20.6, 28.5, 49.3,129.1, 140.9, 145.9, 147.5. Anal. Calcd for $C_{10}H_{12}INO$: C, 41.54; H, 4.18; N, 4.85. Found: C, 41.19; H, 4.24; N, 4.79.

Hydration Equilibrium. The ratios of ketone/hydrate for **3** and **4** in D_2O were measured by ^{1}H NMR spectroscopy (500 MHz). Solutions of the ketones were allowed to equilibrate for 30 min in 0.010 M $DClO_4$ + 0.090 M $NaClO_4$ at 25 °C before recording their NMR spectra.

Kinetics of Hydroxide Catalyzed Ionization. The rates of ionization of **3** and **4** in aqueous sodium hydroxide solutions were determined spectrophotometrically by monitoring the increases in absorbance at 350 and 400 nm. In all the kinetic solutions, the ionic strength was maintained at 0.10 by the presence of the appropriate concentration of $NaClO_4$. Measurements were made at 25.0 ± 0.1 °C using a Hewlett Packard 9153C Diode Array UV−vis spectrophotometer for **4** and a stopped-flow spectrophotometer (Update Instruments System 7-RS) for the faster ionization reactions of **3**.

Kinetic runs for the ionization of **3** in NaOH solution were conducted by first allowing the reactants to equilibrate to 25 °C in the mixing syringes of the stopped-flow spectrophotometer. Ionizations were started by the 1:1 mixing of an aqueous solution of 2-acetyl-1-methylpyridinium ion with an aqueous sodium hydroxide solution so that the concentration of substrate following mixing was 4×10^{-4} M. The ionization followed first-order kinetics accurately for more than five half-lives. Rate constants were calculated by fitting the data to the exponential form of the first-order rate equation.

Products from the reaction of 2-acetyl-1-methylpyridinium ion in NaOH solution were examined in two time course experiments by ^{1}H NMR spectroscopy. The reactions were started by rapidly dissolving 5 mg of **3** in 9.0 mL of 0.1 M NaOH. One reaction was stopped after 5 s and the other after 4 min by adding 9.0 mL of 0.1 M HCl and quickly mixing. Following the removal of water by lyophilization, samples were dissolved in DMSO-d_6, filtered, and their NMR spectra recorded.

For the ionization of **4** in NaOH solution monitored by UV spectrophotometry, reactions were started by the addition of an aliquot of a stock solution of ketone in acetonitrile to 3.00 mL of NaOH solution ([ketone] = 4.4×10^{-4} M and acetonitrile was <1% by volume). In these kinetic runs, however, the exponential increase in absorbance at 400 nm was accompanied by a process that brings about a decay in absorbance. In some cases, the rates had to be corrected for the disappearance of the enolate ion. First-order rate constants were calculated either by using a fit to a simple exponential function or by using the equation $A = A_f k/(l - k)[\exp(-kt) - \exp(-lt)]$,[8] where A_f is the total absorbance change due to enolate ion and k and l are the rate constants for the formation and decay of enolate ion, respectively.

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Kinetics of Iodination. Rates of iodination were measured in aqueous acetate buffer solutions by monitoring the disappearance of I_3^- at 354 nm. Iodine was generated by the acid-catalyzed reaction of KIO_3 with KI. The iodination reactions were done under zeroth-order conditions for **3** and **4**.

The first-order reaction of iodine with 2-acetyl-1-methylpyridinium iodide was carried out as follows: To 1.00 mL of aqueous buffer solution containing 9×10^{-5} M I_2 was added 3.3 μL of 0.027 M substrate stock solution in acetonitrile. The disappearance of iodine in buffer solution in the absence of substrate corresponded to only a small portion of the rate measured in the presence of the substrate. Corrected absorbance decays were obtained by subtracting the background decays from the total decay curve. The resulting decay curves fitted accurately to an exponential function to give the observed first-order rate constants.

Iodinations of **3** and **4** were performed in acetate buffer solutions under zeroth-order conditions. To 3.00 mL of acetate buffer was added 12.0 μL of substrate solution in acetonitrile. The mixture was allowed to equilibrate to 25 °C ([ketone] = 2−0.8 mM). The concentration of 2-acetyl-1-methylpyridinium ion was measured by recording the absorbance of the substrate solution at 310 nm, where the effective molar extinction coefficient is 106.6 ([ketone] = A_{310}/ϵ_{310}). The kinetic runs were initiated by addition of 8.0 or 10.0 μL of 1.0010 mM I_2/0.100 M NaI to the ketone solution, and the absorbances at 354 nm were recorded as a function of time. Traces of decreasing absorption versus time for the first 30% of I_2 consumption were linear. The calculated gradients, v, were converted into observed first-order rate constants using the equation $k_{obs} = v/\epsilon_{354}$[ketone], and the effective extinction coefficient, $\epsilon_{354} = 9.14 \times 10^3$ M^{-1} (for $[I_2] = 9 \times 10-5$ M and $[I^-] = 9.7 \times 10^{-4}$ M in acetate buffer solution at pH 4.2 to 5.0 at 25 °C).

pK_a Determinations. The carbon acidities of **3** and **4** were determined at 25 °C in NaOH solutions and buffer solutions (piperidine, 1-methylpiperidine, and ethanolamine) at an ionic strength of 0.10 by use of the Hewlett-Packard 9153C spectrophotometer. The pHs of buffer solutions were measured by use of a Radiometer PHM 26 pH meter.

The final absorbance values A_f at 350 nm (**3**) and 400 nm (**4**) following the formation of enolate ions were used to construct titration curves. Final absorbance values for **4** were determined as described in the kinetics section. After dissolving **3** in aqueous solution, the absorbance values at 350 nm were recorded one minute from the mixing time (low pH) or by extrapolation of the exponential absorbance decay to the time of mixing (high pH).

The expression for the final absorbance values is given by the sum of the absorption contributions from each of the species in solution $A_f = \epsilon_K[K] + \epsilon_E[E] + \epsilon_h[h] + \epsilon_{h^-}[h^-]$: ϵ_X is the extinction coefficient for a given species, X = K (ketone), E (enolate ion), h (hydrate), h^- (hydrate anion). The pH-dependent absorbance in terms of total substrate concentration, $[S]_{tot}$ is

$$A_f = \frac{\{(\epsilon_h + \epsilon_h K_h)[H^+] + (\epsilon_E K_a + \epsilon_{h^-} K_h K_a^h)\}[S]_{tot}}{(1 + K_h)[H^+] + K_a + K_h K_a^h} = (A[H^+] + B)[S]_{tot}/(K[H^+] + C)$$

For **3** the value of K on the right side of the equation was set at $K = 1 + K_h = 1.084$. For **4** the value of K was set at 1.00 because the hydration of this ketone is negligible. Curve fits for both substates were done according to the above equation, and the acid dissociation constants were obtained using the kinetically determined value of K_Z ($=K_h K_a^h$) and $K_a = C - K_h K_a^h$ (see Results).

III. Results

Hydration Equilibrium. The hydration equilibrium constants of **3** and **4** were determined in aqueous solutions at 25 °C (ionic strength 0.10) by measuring the proportions of hydrate and ketone at equilibrium. A spectrophotometric determination of K_h by semicarbazide scavenging[9] was not possible because the ketones reacted much too slowly, so we measured K_h by ^{1}H NMR spectroscopy. The hydrate of **4** was not observed in the NMR spectrum in D_2O, so that $K_h < 0.001$. Signals assigned to the hydrate and ketone of **3** appeared in an equilibrated solution of the ketone in D_2O, and the integrals of these signals were used to calculate $(K_h)_{D2O} = 0.098$. The equilibrium constant in D_2O was corrected to K_h in H_2O by use of the equation $(K_h)_{D_2O}/(K_h)_{H_2O} = \Phi^2 = 1.08$.[9,10] The corrected value of $(K_h)_{H_2O}$ was 0.084 ± 0.004.

Enolate Ion Formation. Reaction of the cationic ketones **3** and **4** with aqueous NaOH leads to the appearance of chromophores at 350 and 400 nm, respectively. The absorption maxima appear at longer wavelengths than those characterizing enolates of alkylsubstituted monocarbonyl compounds (220−260 nm).[1a] However, the λ_{max} values for the enolates of **3** and **4** are similar to those of the enolates of the related aromatic compounds 3-(phenylacetyl)pyridinium ion (360 nm) and 4-(phenylacetyl)pyridinium ion (446 nm), respectively.[11]

Consideration has been given to whether the chromophores that develop quickly upon mixing **3** or **4** with hydroxide are attributable to pseudobase addition compounds rather than to enolate ions. These chromophores are most consistent with enolates for the following reasons: (a) The spectral changes are reversed upon neutralization, consistent with an acid/base equilibration. (b) The positions of the absorption maxima are similar to those for other, structurally related aromatic enolates.[11] (c) An NMR experiment with **4** in D_2O confirmed exchange of the methylene protons adjacent to the carbonyl group occurs at a rate comparable with development of the chromophore. Ketone **4** was dissolved in 0.1 M NaOD, stirred for 2 min at room temperature, and quenched with 1 equiv of DCl. Solvent was removed, and **4** was dissolved in CD_3CN for ^{1}H NMR analysis, which showed >99% loss of the protons in the α-CH_2 group (normally a 2H triplet at 2.89 ppm). A lower limit for the exchange rate was estimated by considering the two-stage exchange reaction: $-CH_2- \rightarrow -CHD- \rightarrow -CD_2-$ governed by rate constants k_1 and k_2, respectively. Setting $k_1 = k$ and $k_2 = k/2$, assuming a negligible secondary isotope effect, the total proton content for the two-proton exchange is given by $[H] = [H_0] \exp(-kt/2)$.[12] An estimate of the minimum exchange rate constant leading to 99% exchange, which corresponds to at least 7 half-lives in 120 s, is $k \geq 0.08$ s^{-1}, and the enolization rate constant is $2k \geq 0.15$ s^{-1} for **4** in 0.1 M NaOD. The minimum enolization rate constant as estimated by NMR is consistent with the spectrophotometrically measured value of 0.3 s^{-1} in 0.1 M NaOH.

Over a longer time interval, the chromophores attributed to enolate ions decreased in intensity. *N*-Methylpyridinium ion was identified by ^{1}H NMR as the product arising from this absorbance decay in the reaction of **3** in NaOH. We propose that the disappearance of the enolate ions derived from **3** and **4** results from deacylation by way of the hydrate anion species, as shown in eq 1 for the reaction of **3**. This deacylation is analogous to the well-known deacylation of 2-acetyl-3,4-dimethylthiazolium ions,[5] but it takes place on a much slower time scale, allowing enolization to be observed spectrophotometrically at high pH values. In this sense, the deacylations of **3** and **4** validates the 2-acylpyridinium ions as analogues of 2-acetylthiazolium ions.

Kinetics of Enolate Ion Formation. The time dependencies

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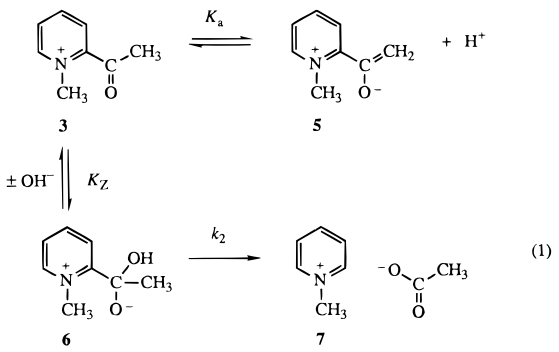


3 **5** **6** **7**

of the increases in absorption accompanying enolate ion formation in aqueous sodium hydroxide solutions were measured by stopped-flow spectrophotometry in the case of **3** and conventional spectrophotometry for **4** at 25 °C and ionic strength 0.10. The rates of enolate formation observed at 350 nm for **3** were first order in ketone concentration for more than 98% of the reaction. Observed first-order rate constants were obtained in NaOH solutions over the concentration range 0.004−0.10 M. The increase in absorbance at 400 nm corresponding to the ionization of **4** was followed by its decay. In hydroxide solutions of 0.8−9.9 mM, the decay of the band at 400 nm was slow relative to its formation; therefore, first-order rate constants were evaluated using a single exponential function. At base concentrations of 20−99 mM, however, the secondary hydrolysis reaction interfered, and the corrected rate constants for enolate ion formation were determined as described in the Experimental Section.

The first-order rate constants for ionizations of **3** and **4** increase with the hydroxide concentration at the lower end of the concentration range and, as shown in Figures 1 and 2, reach limiting values at higher concentrations. This behavior is consistent with an ionization reaction in which there is a kinetically controlled conjugate base, Z^-, formed with an equilibrium constant of K_Z. In the case of the reaction of **3** according to eq 1, Z^- is the hydrate anion and K_Z is the equilibrium constant for its formation. The applicable rate law is given by eq 2,

$$k_{obs} = (k_{OH}[OH^-] + k_{H_2O})/(1 + K_Z[OH^-]/K_w) \qquad (2)$$

where K_w is the ion product of water and k_{H_2O} is the rate constant for uncatalyzed enolization. The constants k_{OH}, k_{H_2O}, and K_Z were evaluated for **3** and **4** by fitting data to eq 2. Because these ketones are in equilibrium with their hydrate forms, the rate constants are multiplied by the correction factor ($K_h + 1$) to convert to the "true" rate constants, which are given in Table 1.

The hydrate anion **6** (eq 1) and its analogue derived from **4** are the most obvious species that can be postulated for the structure of an anionic decomposition intermediate Z^-. The formation of hydrate anions has also been postulated to explain saturation kinetics in the deprotonation reactions of 1-methyl-3-acetylpyridinium and 4-(arylacetyl)pyridinium ions in basic aqueous solutions.[11] The equilibrium constant K_Z is, therefore, related to the acid dissociation constant for the hydrate, K_a^h: $K_Z = K_h K_a^h$.

Rate constants for enolate ion formation in acetic acid/acetate buffers were measured indirectly by iodine scavenging. Rate measurements were made in a series of solutions at constant ionic strength and buffer ratio while varying the buffer concentration. Rates of enolization were measured in a series of experiments at four buffer ratios: [HOAc]/[AcO$^-$] = 0.49, 0.97, 1.38, and 1.92. The enolizations were zero order with respect to iodine in at least the first portion of the reaction (up to 50%). This behavior was observed previously for 2-acetyl-1-methylpyridinium ion by Cox,[6] who also demonstrated that the rates were independent of the halogen used.

The rates of enolization were found to be proportional to the buffer concentration. Therefore, the data were fitted to eq 3 to obtain the buffer catalytic coefficients k_{cat}. For each data set at a given buffer ratio, k_{cat} was then fitted to eq 4, in which k_{HA}

$$k_{obs} = k_o + k_{cat}[buffer] \qquad (3)$$

$$k_{cat} = k_{HA} + (k_B - k_{HA})f_B \qquad (4)$$

and k_B are the buffer-acid and buffer-base catalytic coefficients, respectively, and f_B is the fraction of buffer in the basic form (f_B = [AcO$^-$]/([HOAc] + [AcO$^-$]). Linear plots of k_{cat} versus [buffer] with zero intercepts showed that the reaction is subject to general base catalysis but not general acid catalysis.

Plots of the intercepts at zero buffer concentration (k_o) versus hydroxide concentration were made in order to determine the rate constants k_{H_2O} and k_{OH} by fitting to eq 5. For both ketones

$$k_o = k_{H_2O} + k_{OH}[OH^-] \qquad (5)$$

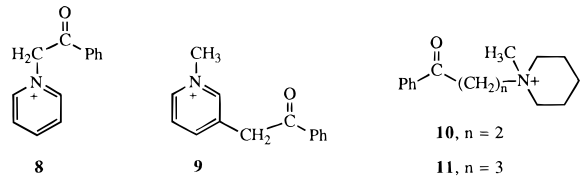
the slope was zero within error, indicating that the effect of hydroxide ion in acetate buffer solutions was small. Upper limits of k_{OH} were found to be 6.3×10^3 and 1.7×10^2 M^{-1} s^{-1} for **3** and **4**, respectively.

The rate constants determined in acetate buffer studies were the catalytic coefficients referring to the unit concentration of an equilibrium mixture of ketone and its hydrate. They were converted to true catalytic coefficients based on unit ketone concentration through multiplication by ($1 + K_h$), Where K_h = 0.084 and 0 for **3** and **4**, respectively (see below). The rate constants corrected for hydration are given in Table 1. The rate constant for the enolization of **3** by acetate agrees with the previously determined value of 8.3×10^{-3} M^{-1} s^{-1}.[6]

pK_a Determination. The ketone acidity constants for **3** and **4** in aqueous solution were determined spectrophotometrically. The final absorbance values following enolate ion formation in kinetic runs (350 nm for **3** and 400 nm for **4**) were used to construct the titration curves in Figures 3 and **4** according to the procedure described in the Experimental section. The calculated values of pK_a for compounds **3** and **4** are given in Table 1.

IV. Discussion

When compared with acetone, **2** and **3** undergo hydroxide-catalyzed ionization about 10^6 and 1.7×10^3 times times faster, respectively, and their rates of base-catalyzed enolization are also orders of magnitude faster than that of acetaldehyde. Cationic ketones **8** and **9** also ionize very fast,[13] and the ketones **10** and **11** undergo hydroxide ion-catalyzed enolization faster



8 **9** **10**, n = 2 **11**, n = 3

than their unmethylated forms by factors of 2600 and 15, respectively. Fast enolizations of these compounds have been

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Table 1. Summary of Rate and Equilibrium Constants for the Interconversions of 2-Acetyl-3,4-dimethylthiazolium and 2-Acetyl-1-methylpyridinium Ions in Aqueous Solution[a]

parameter/compd	**2**[3]	**3**	**4**
k_{OH} (M^{-1} s^{-1})	$(1.20 \pm 0.20) \times 10^{5}$ [b]	$(1.87 \pm 0.50) \times 10^{2}$ [b]	10.9 ± 0.2
k_{H_2O} (s^{-1})		1.32 ± 0.21	$(9.17 \pm 0.24) \times 10^{-2}$
k_{OAc} (M^{-1} s^{-1})	$(7.63 \pm 0.95) \times 10^{-3}$ [b]	$(2.82 \pm 0.95) \times 10^{-2}$ [b]	$(4.32 \pm 0.18) \times 10^{-4}$
pK_a	8.8[c]	11.13 ± 0.05	11.90 ± 0.03
pK_Z ($-\log K_h K_a^h$)		12.53 ± 0.12	12.32 ± 0.04
K_h	1.10 ± 0.10	0.084 ± 0.04	<0.001

[a] Ionic strength = 0.10, 25 °C. [b] Corrected for hydration equilibrium. [c] Estimated as described in the text.

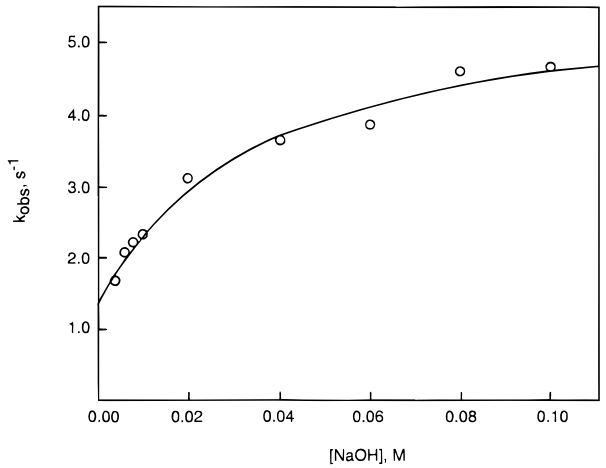


Figure 1. Dependence of ionization rate on NaOH concentration for 2-acetyl-1-methylpyridinium ion **3**. The first order rate constants are plotted versus the concentration of aqueous NaOH for the ionization of 2-acetyl-1-methylpyridinium ion **3**. Each point represents the average of 5 to 7 rate measurements.

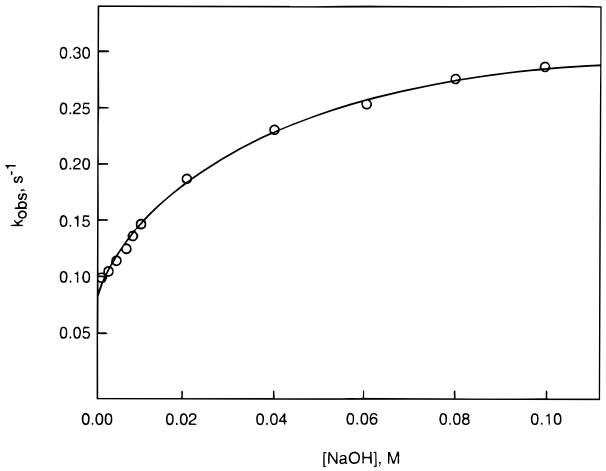


Figure 2. Dependence of ionization rate on NaOH concentration for 1-methyl-5,6,7,8-tetrahdroquinolinium ion (**4**). The first-order rate constants are plotted versus the concentration of aqueous NaOH for the ionization of 1-methyl-5,6,7,8-tetrahdroquinolinium ion (**4**). Each point represents the average of 6 rate measurements.

attributed to electrostatic stabilization of the developing negative charge on the carbonyl oxygen by the positive charge on nitrogen in the transition state for enolate formation.[14] The electrophilic effects of positively charged nitrogen in the enolization of these molecules can be conceptualized as the product of two factors, the inductive effects of the electropositive substituents and the through-space electrostatic effects of the positive charges. Each of these effects can be manifested in both the ground state for the enolate ion and the transition state for its formation. Inductive and through-space effects are not

(14) Cox, B. G.; De Maria, P; Fini, A. *J. Chem. Soc., Perkin Trans. 2* **1984**, *44*, 3308.

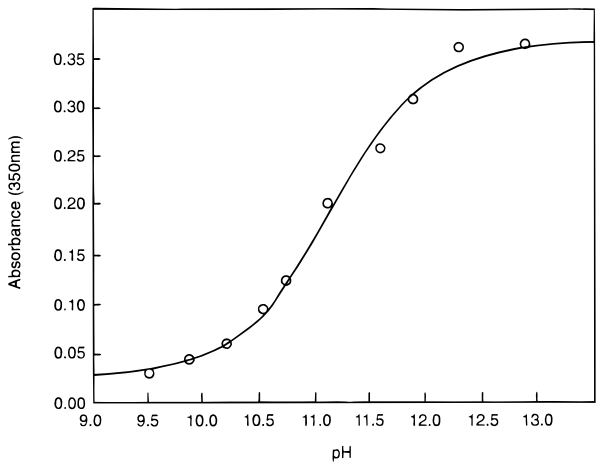


Figure 3. Titration curve for the ionization of 2-acetyl-1-methylpyridinium ion (**3**) in aqueous NaOH. Absorbance at 350 nm is plotted against pH. Each point represents the average of five to six determinations.

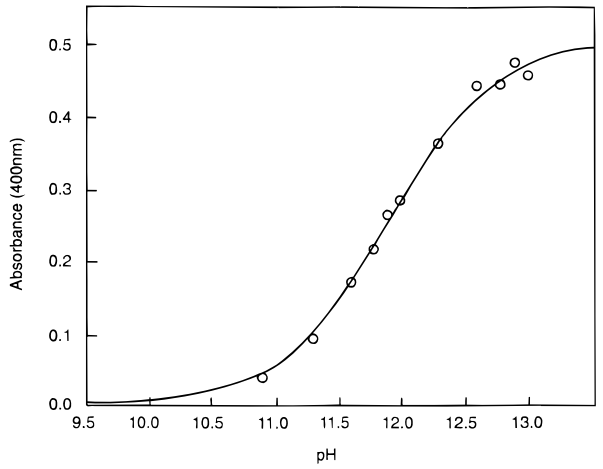


Figure 4. Titration curve for the ionization of 1-methyl-8-oxo-5,6,7,8-tetrahydroquinolinium ion (**4**) in aqueous NaOH. Absorbance at 400 nm is plotted against pH. Each point represents the average of six determinations.

easily separated and have not been for **8**−**11**. In the following discussion, we shall attempt to delineate the inductive and through-space electrostatic effects of the 2-(*N*-methylpyridinium) and 2-(*N*-methylthiazolium) substituents on the enolizations of **2** and **3**. The through-space electrostatic components are of special interest for understanding enzymatic enolization, in which positively charged side chains of basic amino acid residues may facilitate the formation of enolate ions by stabilizing them in their ground states and in the transition states for their formation.

Kinetics of Enolate Ion Formation. Rates of base-catalyzed enolate ion formation vary with the structure of the carbonyl molecule. In general, the more acidic the molecule the faster the ionization reaction. Kresge and Keeffe have constructed a

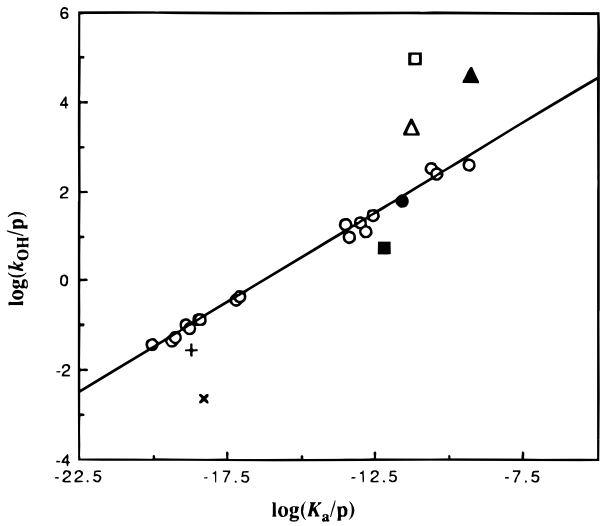


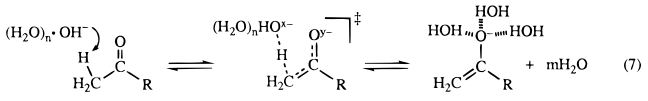
Figure 5. Correlation of k_{OH} for enolization with pK_a values of ketones. Relationship between the rate constants for hydroxide-catalyzed enolate ion formation, logk_{OH}/p, and carbon acid ionization constants, log(K_a/p), for carbonyl molecules at 25 °C. Symbols: (open circles) 17 methyl and benzyl ketones; the correlation line is given by eq 6[1a]; (filled triangle) 2-acetyl-3,4-dimethylthiazolium ion (**2**);[3] (filled circle) 2-acetyl-1-methylpyridinium ion (**3**); (filled square) 1-methyl-8-oxo-5,6,7,8-tetrahydroquinolinium ion; (open square) α-(1-pyridinium)acetophenone (**8**);[14] (open triangle) α-3-(1-methyl-pyridinium)acetophenone (**9**);[14] (×) isobutyrophenone;[16] (+) cyclohexanone.[17]

logarithmic correlation of rate constants for hydroxide-catalyzed enolate formation with pK_a values for monocarbonyl compounds in water at 25 °C.[1a] A good linear relationship has been found for 17 benzyl and methyl ketones,[15] as shown by the open circles in Figure 5. The data were fitted to eq 6, where p is the number

$$\log(k_{OH}/p) + (0.40 \pm 0.01)\log(K_a/p) + (6.47 \pm 0.13) \quad (6)$$

of chemically equivalent acidic protons in the carbonyl molecule. Cationic ketones **8** and **9** and other selected ketones[16,17] are included in Figure 5 to illustrate deviations from the overall correlation. The 2-acetyl-3,4-dimethylthiazolium ion (**2**) is also included in Figure 5 as a positive deviation, based on its ionization rate[3] and its estimated pK_a (see below). Positive deviations have been attributed to intramolecular electrostatic acceleration.[1a,18]

Electrostatic Acceleration. The central fact about enolate ion formation is that it is very slow when compared with the dissociation of protons from "normal acids" of comparable pK_a. The most probable reason for the slow rate is suggested by the structural formulas in eq 7, which illustrates the fact that the negative charge generated by abstraction of a proton from the α-carbon resides mainly on the enolate-oxygen atom. The attacking hydroxide ion must be solvated in the ground state, whereas the enolate oxygen must be solvated both in the



transition state and the final ground state for the enolate. The two solvation sites are more remote from each other than in the case of a "normal acid", in which the negative charge is substantially borne by the atom from which the proton has been abstracted. In enolate ion formation, therefore, substantial reorganization of the solvent must take place in the transition state, and this slows the reaction.[19] In a series of ketones closely related in structure, as represented by the 17 open circles for benzyl and methyl ketones in Figure 5, the asymmetries of the solvation requirements are similar, so that the ionization rates are correlated by the line. In cases in which requirements for solvation differ from those of ordinary benzyl and methyl ketones, deviations from the correlation line will be observed. Such deviations have been referred to as transition state imbalance or imperfect synchronization.[19]

The fact that values of log(k_{OH}/p) for cationic ketones **2**, **8**, and **9** lie well above the line in Figure 5 suggests that stabilizing interactions appear in their enolization transition states that are more pronounced than in the enolate ions. The positive deviations correspond to 70-, 920-, and 30-fold enhancements in rates for ketones **2**, **8**, and **9**, respectively, relative to the rates that would coresond to their pK_a values. The ionization rate for 2-acetyl-1-methylpyridinium ion **3** is well correlated to its acidity. In the case of the cyclic ketone 1-methyl-8-oxo-5,6,7,8-tetrahydroquinolinium ion (**4**), the carbonyl group appears to be in position for an electrostatic interaction with the enolate oxygen; however, this ketone ionizes more slowly than expected from its pK_a. This may to be due to the fact that it is a cyclic ketone that is ring-fused to the pyridinium ion, which may impose conformational constraints on the enolization process. Through-space electrostatic stabilization that appears to be made possible by its structure does not overcome the internal conformational constraints. Examples from the literature of other negative deviations include isobutyrophenone[16] and cyclohexanone,[17] which also fall below the line in Figure 5. The second α-alkyl substituent in isobutyrophenone is thought to destabilize the negative charge on the incipient enolate ion, bringing about a transition state imbalance. This effect together with a probable steric retardation on the deprotonation causes the slower rate.

Estimation of pK_a for 2-Acetyl-3,4-dimethylthiazolium Ion. 2-Acetyl-3,4-dimethylthiazolium ion (**2**) ionizes extremely rapidly, and 2-acetylthiamine pyrophosphate (**1**) ionizes about 4 times faster.[3] The ionization rates for these compounds can be compared with those of other methyl ketones if the ionization constant for enolate formation can be evaluated. The pK_as of **1** and **2** cannot be measured directly owing to their rapid hydrolysis in basic solutions. However, the acidity of **2** may be estimated by comparing its ketonic properties with those of **3** described in this paper.

The inductive effect of the 2-(*N*-methylthiazolium) substituent on the carbonyl group of **2** has been recognized as a factor in its favorable hydration equilibrium.[5] In a series of methyl ketones, the hydration constants are related to the polar substituent parameter σ^* by eq 8.[20a] Hydration does not entail

$$-\log(1/K_h) = (1.70 \pm 0.07)\Sigma\sigma^* - (2.81 \pm 0.31) \quad (8)$$

an alteration in electrostatic charge, so that the effects of the

(15) (a) Chiang, Y.; Kresge, A. J.; Tang, Y. S.; Wirz, J. *J. Am. Chem. Soc.* **1984**, *106*, 460. (b) Jones, J. R.; Mark, R. E.; Subba Rao, S. C. *Trans. Faraday Soc.* **1967**, *63*, 111. (c) Guthrie, J. P.; Cossar, J.; Klym, A. *Can. J. Chem.* **1987**, *65*, 2154. (d) Keeffe, J. R.; Kresge, A. J.; Toullec, J. *Can. J. Chem.* **1986**, *64*, 1224. (e) Chiang, Y.; Kresge, A. J.; Krough, E. T. *J. Am. Chem. Soc.* **1988**, *110*, 2600. (f) Chiang, Y.; Hojatti, M.; Keeffe, J. R.; Kresge, A. J.; Schepp, N. P.; Wirz, J. *J. Am. Chem. Soc.* **1987**, *109*, 4000. (g) Chiang, Y.; Kresge, A. J.; Walsh, P. A.; Yin, Y *J. Chem. Soc., Chem. Commun.* **1989**, 869. (h) Yin, Y. Ph. D. Dissertation, Department of Chemistry, University of Toronto, 1988. (i) Pollack, R. M.; Mack, J. P. G.; Eldin, S. *J. Am. Chem. Soc.* **1989**, *111*, 6419.

(16) Pruszynski, P.; Chiang, Y.; Kresge, A. J.; Schepp, N. P.; Walsh, P. A. *J. Phys. Chem.* **1986**, *90*, 3760.

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(18) Kirby, A. *Adv. Phys. Org. Chem.* **1980**, *17*, 183.

(19) (a) Bernasconi, C. F. *Pure Appl. Chem.* **1982**, *54*, 2335. (b) Bernasconi, C. F. *Acc. Chem. Res.* **1987**, *20*, 301.

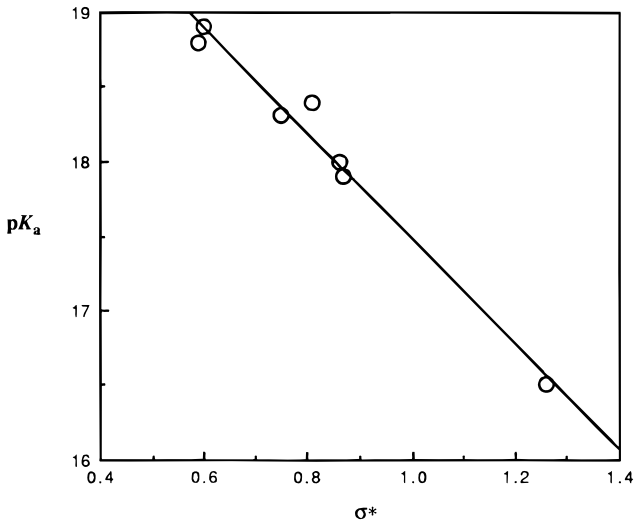

Figure 6. Correlation of pK_a with σ^* in phenyl-substituted acetophenones. Literature values of pK_a and σ^* for their phenyl substituents[21] are plotted for the compounds *p*-methoxyacetophenone, *p*-methylacetophenone, acetophenone, *p*-fluoroacetophenone, *p*-chloroacetophenone, *p*-bromoacetophenone, and *p*-nitroacetophenone. The correlation line is given by p$K_a = (-2.13 \pm 1.05)\sigma^* + 19.79$.

2-(*N*-methylthiazolium) and 2-(*N*-methylpyridinium) substituents on the hydration of **2** and **3** should be attributed to their inductive effects. On the basis of the published value of K_h for **2**[3] and the present evaluation of K_h for **3**, the values of σ^* calculated from eq 8 are 1.68 and 1.02 for **2** and **3**, respectively. These values of σ^* can be used to estimate the inductive effects of the 2-(*N*-methylthiazolium) and 2-(*N*-methylpyridinium) substituents on the pK_as of **2** and **3**.[20b]

A linear correlation between pK_a and σ^* values of α-groups in a series of phenyl-substituted acetophenones is shown in Figure 6, which is plotted using literature data.[21] This correlation should pertain to **2** and **3** because all of the data are for methyl ketones bonded to aromatic rings through sp^2 carbons. Therefore, the σ^* values for **2** and **3** can be used with the correlation line in Figure 6 to determine what the pK_as of **2** and **3** would be based soley on the inductive effects of 2-(*N*-methylthiazolium) and 2-(*N*-methylpyridinium) substituents. This operation gives values of 15.1 and 17.4 as the pK_as for **2** and **3**, respectively. The measured pK_a for **3** (11.13,Table 1) is 6.3-log units lower than that predicted by the inductive effect. The additional acid strengthening should be attributed to through-space electrostatic stabilization of the enolate ion by the cationic nitrogen. On structural grounds, we believe that the same electrostatic effect should increase the acidity of of the 2-acetyl-3,4-dimethylthiazolium ion (**2**). We therefore estimate the pK_a of 2-acetyl-3,4-dimethylthiazolium ion as 15.1 − 6.3, or 8.8.[22]

Table 2. Carbonyl Stretching Frequencies of Acylpyridine and Acylthiazole Compounds[a]

	ν (cm^{-1})		$\Delta\nu^b$ (cm^{-1})	
ketone	CH_3CN	D_2O	CH_3CN	D_2O
2-acetylpyridine	1701.4	1691.8		
$N^+-D\ Cl^-$ [c]	1720.7	1716.9	19.3	25.1
$N^+-CH_3\ I^-$ (**3**)		1715.1		23.3
$N-Cu^{2+}$ [d]		1686.9		−4.9
3-acetylpyridine	1692.8	1685.5		
$N^+-D\ Cl^-$ [c]	1711.1	1708.8	18.3	23.3
$N^+-CH_3\ I^-$		1705.1		23.3
8-oxy-5,6,7,8-tetrahydroquinoline	1703.4			
$N^+-CH_3\ I^-$ (**3**)	1716.9		13.5	
2-acetyl-4-methylthiazole	1689.9			
$N^+-CH_3\ I^-$ (**2**)	1716.9		27.0	

[a] Concentration = 0.02 M. [b] $\nu_{(N-\text{substituted ketone})} - \nu_{(\text{neutral ketone})}$. [c] Ketone dissolved in 0.10 M DCl/D_2O. [d] Ketone dissolved in 0.20 M $CuCl_2$/D_2O.

The pK_a of **2** is remarkably low. We know of no other methyl ketone that is so acidic. Other ketones that are comparably acidic contain two or more activating functional groups on the ionizing carbon atom, one example being acetylacetone (pK_a = 9).

Spectroscopic Properties of Cationic Ketones. The inductive acid-strengthening effect of the 2-(*N*-methylthiazolium) substituent in **2** is larger than that of the 2-(*N*-methylpyridinium) substituent in **3**. Examination of the spectroscopic properties of the ketonic groups can give information about whether the acid strengthening features of the substituents may be attributed in part to differential destabilization of the keto forms. The carbonyl stretching frequency is sensitive to the electronic structure and is, therefore, sensitive to substituent inductive effects. The carbonyl stretching frequencies are given in Table 2. The frequency for 3-acetylpyridine is 1680−1690 cm^{-1}, and for 2-acetylpyridine, it is 1690−1701 cm^{-1}, depending on the solvent. A slight shift of ν to lower frequency upon changing from organic solvent to D_2O may result from the increase in polarity and/or the increase in hydrogen bonding.

Methylation and deuteration of the ring nitrogens lead to 10−27 cm^{-1} shifts in the C═O frequency for the ketones. It is significant that the carbonyl stretching frequencies for the cationic forms are relatively unaffected by solvent. The explanation of this in the case of 3-acetylpyridine has been suggested to be that resonance forms carrying positive charge on ring carbons make a significant contribution to the electronic structures of aromatic ketones.[23] In the quaternary system, resonance forms that tend to increase positive charge on the positively charged ring are disfavored, so that the contributions of these forms are less important than in the uncharged parent ketones. The $\nu_{C=O}$ for the *N*-methylated forms are much more like unperturbed carbonyl stretches (1700−1720 cm^{-1}). As shown in the bottom line of Table 2, the effect of the quaternary nitrogen on $\nu_{C=O}$ in the thiazolium system is substantially more pronounced than in the pyridinium system. This can be correlated with the greater inductive effect in the thiazolium system. The greater electrophilicity of the 2-(*N*-methylthiazo-

(20) (a) Greenzaid, P.; Luz, Z.; Samuel, D. *J. Am. Chem. Soc.* **1967**, *89*, 749. (b) A similar analysis could be applied to 2-acetylthiamine pyrophosphate **1**, for which values of $(K_h)_{D_2O}$ are available.[4a] However it is questionable whether the hydration of **1** can be correlated with that of **2** or other ordinary ketones because of the steric interference with hydration that can be expected on the part of the *N*-pyrimidinyl group on the thiazolium ring of **1**. The pyrimidinyl group lies near the ketonic function, and for this reason the major form of **1** in aqueous solution is the internally cyclized carbinolamine form, in which the pyrimidinyl amino group forms an adduct with the acetyl carbonyl group.[4a]

(21) The σ^* values are from: Perrin, D. D.; Dempsey, B.; Sergeant, E. P. *pK$_a$ Prediction for Organic Acids and Bases*; Chapman and Hall: New York, 1981. The pK_a values are from: Keeffe, J. R.; Kresge, A. J. In *The Chemistry of Enols*; Rappoport, Z., Ed.; Wiley: New York, 1990.

(22) A much less reliable estimate of pK_a for **2** could be obtained by placing its value for $\log(k_{OH}/3)$ on the correlation line in Figure 5. The resulting value of 4.2 is an unreasonably low pK_a for a methyl ketone, 4.6-log units lower than the much more reasonable estimate of 8.8 found above and shown in Table 1. This supports our conclusion that **2** enolizes much more rapidly than can be accounted for by its pK_a, and it must result from an internal lowering of the transition state energy for enolization. The most obvious source of this is an electrostatic interaction of the positively charged ring with the developing negative charge in the transition state for enolization.

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lium) group may be due to its lesser aromaticity relative to the 2-(*N*-methylpyridinium) group, which should allow the positive charge to be more highly dispersed in the pyridinium ring. A lesser dispersal of positive charge in the thiazolium ring would tend to make carbon-2 more electropositive than it is in the pyridinium ring.

Through-Space Electrostatic Acceleration of Enolization. The through-space electrostatic effects of quaternary nitrogen on the ionization rates of **2** and **3** can be conceptually separated into two categories: (1) electrostatic stabilization of the enolate ion in its ground state, which manifests itself as increased carbon acidity (decreased pK_a), and (2) additional electrostatic stabilization of the incipient enolate oxyanion in the transition state, which is manifested in a positive deviation from the correlation line in Figure 5. The through-space electrostatic effect on the carbon acidities of **2** and **3** is here estimated as corresponding to a 6.3-log unit downward perturbation of pK_a, and it corresponds to a 330-fold acceleration of the enolization rate for both compounds. The additional through-space electrostatic stabilization of the transition state in the case of **2** leads to an additional 70-fold rate enhancement. The latter effect may be attributed to a decreased requirement for solvent reorganization in the transition state owing to electrostatic stabilization. The overall through-space electrostatic acceleration for **2** is, therefore, (330)(70) or 2.3×10^4.[24]

The inductive effects of the 2-(*N*-methylthiazolium) and 2-(*N*-methylpyridinium) substituents on enolization rates can also be estimated. Using acetone (pK_a = 19.3) as a standard methyl ketone, and ignoring for the moment the through-space electrostatic contributions to the acidities, the electron withdrawing properties of the 2-(*N*-methylthiazolium) and 2-(*N*-methylpyridinium) substituents would have the effect of lowering the pK_a values to 15.1 and 17.4 for **2** and **3**, respectively. On the basis of eq 6, the values of k_{OH} for methyl ketones exhibiting these pK_as will be 46- and 5.7-fold higher, respectively, than that for acetone. The inductive effect of the 2-(*N*-methylthiazolium) group on the rate is larger than for the 2-(*N*-methylpyridinium) group, and both are smaller than the through-space acceleration.

Implications of Electrostatic Acceleration for Enzymatic Enolizations. Enzymes catalyze enolizations of aldehydes, ketones, thio esters, and carboxylic acids at very fast rates (ms) at pH 7. Solvent reorganization should not retard enzymatic enolization, because solvation of the enolate can be expected to be provided by the enzymatic binding site.[19b] However, high pK_a values remain as significant barriers. Two major classes of interactions between carbonyl compounds and enzymatic sites have been proposed to contribute to overcoming these barriers, electrostatic acceleration by metal ion cofactors or basic amino acid groups in the active site,[2] and low-barrier hydrogen bonds formed between substrate carbonyl groups and electrophilic groups in the active site.[25] We have here described the fast enolization of two cationic ketones in which hydrogen bonding and metal coordination can play no role.

The present results show that purely electrostatic, through-space stabilization of the enolate ions and the transition states for enolate formation for **2** and **3** in aqueous solutions are significant. Analogous electrostatic effects in enzymatic active sites can be expected to be at least comparable, and even larger when the effective dielectric constants are smaller than that of water. Cationic groups of enzymes that may participate in enolization include the basic groups of amino acid side chains such as lysine, arginine, and histidine. All of these groups can contribute hydrogen bonds as well as positive charges toward stabilizing enolates and transition states, and the hydrogen bonds will further enhance enolization rates. Metal ions can be as effective or more effective than basic groups. Low-barrier hydrogen bonds may be brought into play, especially in sites lacking metal ions or cationic amino acids, such as the enolization site of triose phosphate isomerase.

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(24) A reviewer has suggested that a part of the inductive effect may arise from a field or through-space effect. We have here taken the inductive effect to be that exerted by the electropositivity of the substituent carbon, carbon-2 of **2** or **3**, on the methyl ketone groups, whatever the origins of electropositivity. In **2** and **3**, the sources of electropositivity are the sp^2 character of carbon-2 and electron withdrawal by the quaternary nitrogen to which it is bonded. In these cases, the most direct and simple medium for electropositivity to be transmitted from the quaternary nitrogen to carbon-2 is through the covalent linkage between them. It is difficult to imagine a nonbonded field between nitrogen-1 and carbon-2 that would be a better conduit for electron withdrawal than the bond linking them.

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