Evidence for a Solvent-Induced Change in the Rate-Determining Step in the Water-Catalyzed Hydrolysis of the Dimethylimmonium Ion of Benzophenone

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The first order rate constants (k_1) and the kinetic solvent isotope effects (KSIE) for the hydrolysis of the dimethylimmonium ion of benzophenone (2) have been determined in mixed water–acetonitrile solvents varying between pure water and 90% acetonitrile–10% water by volume. Between 0 and 50% acetonitrile the k_1 increases from $(1409 \pm 6) \times 10^{-6}$ s^{-1} to $(3487 \pm 42) \times 10^{-6}$ s^{-1} and the KSIE decreases from 2.19 to 1.97. Between 50 and 90% added acetonitrile the k_1 decreases from $(3487 \pm 42) \times 10^{-6}$ s^{-1} to $(1662 \pm 8) \times 10^{-6}$ s^{-1} and KSIE increases from 1.97 to 2.32. The $\Delta G^{\ddagger}$ remains almost constant while $\Delta H^{\ddagger}$ and $\Delta S^{\ddagger}$ decrease regularly with the increase of acetonitrile in the solvent mixture. The proton inventory studies generate downwardly bowed k_n vs n plots. However, the degree of bowing decreases with the increase of acetonitrile in the solvent mixture. The results are interpreted by invoking a mechanism that involves a rate-limiting conversion of the polarized substrate (A′) into a protonated carbinolamine (B′) via a cyclic transition state (4) at lower volume percent, 0–50%, of acetonitrile and two rate-limiting steps, the conversion of A′ into a carbinolamine (B) via an acyclic transition state and the conversion of B into zwitterion (C) via a cyclic transition state at higher volume percent, 50–90%, of acetonitrile. These latter processes occur at comparable rates.

The kinetic parameters that are observed for the water-catalyzed hydrolysis of reactive acyl[2] and imine[3] compounds have often been regarded as implying substantial involvement of two water molecules in the transition state (1). One water molecule is presumably covalently bound to the carbonyl or imine carbon and the other acts as a base to abstract a proton from the first. For many such reactions it is of interest to know whether or not there are other water molecules, in addition to the two water molecules firmly bound, involved with the substrate in the transition state. Kinetic data are often inadequate in obtaining this information since water is usually present in great excess.

Kinetic solvent isotope effects in protium oxide–deuterium oxide mixtures are an extremely sensitive probe for acquiring this information, which is certainly of considerable interest in enzyme-catalyzed reactions. Furthermore, comparison between entropies of activation for similar transition states can also provide insight into solvent reorganization and specific interactions between solvent and transition state during a reaction.

As a part of our continuing effort in this direction, we chose to study the solvent isotope effect for the neutral water-catalyzed hydrolysis of the dimethylimmonium ion of benzophenone (2) in various protium oxide–deuterium oxide mixtures containing 0, 25, 50, 75, and 90 volume % acetonitrile and to determine the activation parameters of this reaction in these solvent mixtures. A comparison of the results with the corresponding results for the neutral hydrolysis of 1-acetylimidazolium ion (3a) and 1-acetyl-
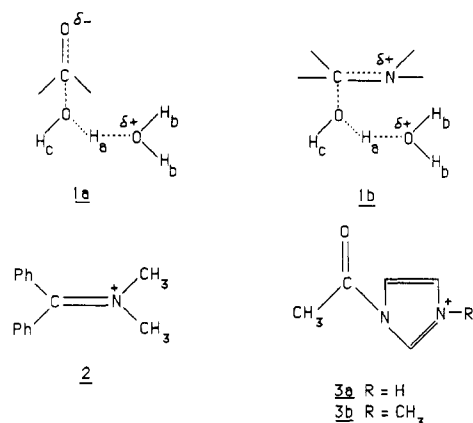
3-*N*-methylimidazolium ion (3b) reveals that binding of water molecules in excess of those in 1b is required in order to account for the results observed in solvent mixtures containing in excess of 50 volume % water. Furthermore, the comparison reveals that there is a change in the rate-determining step when the concentration of water in the solvent mixture is decreased below 50%.

Results

The neutral water-catalyzed hydrolysis of 2 was studied in water–acetonitrile mixtures containing different volume percentages of protium oxide or deuterium oxide at pH(D) 3.0 at 25 °C. The reaction rate was found to increase with the decrease in water concentration down to about 50% and then to decrease with a further decrease in water concentration (Figure 1). This bell-shaped water concentration versus rate profile is unusual.

The hydrolysis of 2 was also studied in protium oxide–deuterium oxide mixtures of atom fraction of deuterium n equal to 0, 0.25, 0.50, 0.70, and 1.0 in solvent systems containing 0, 25, 50, 75, and 90% by volume acetonitrile. Table I shows the dependence of the observed first-order rate constants on the isotopic composition of the solvent in these acetonitrile–water solvent systems.

The first-order rate constants for the reaction were also determined in protium oxide–acetonitrile mixtures containing 0, 25, 50, 75, and 90% volume acetonitrile at different temperatures in the range 25–50 °C (Table II).[16] The values of the activation parameters derived from these

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Table I. First-Order Rate Constants ($10^6 k_n$, s^{-1}) for the Water-Catalyzed Hydrolysis of 2 in H$_2$O–D$_2$O Mixtures of Atom Fraction of Deuterium n in Acetonitrile–Water Mixtures at 50.00 ± 0.05 °C[a]

atom fraction of deuterium (n)	volume percentage acetonitrile				
	0	25	50	75	90
0.00[b]	1409 ± 6[c]	2764 ± 9	3487 ± 42	2861 ± 14	1662 ± 8
0.25[b]	1179 ± 8	2330 ± 31	3012 ± 9	2441 ± 17	1414 ± 17
0.50[b]	978 ± 8	1987 ± 1	2555 ± 6	2093 ± 25	1209 ± 16
0.75[b]	801 ± 4	1644 ± 89	2121 ± 1	1771 ± 12	964 ± 9
1.00[b]	644 ± 3	1336 ± 3	1769 ± 5	1424 ± 24	749 ± 7

[a] The pH(D) was maintained at 3.0 by the addition of HCl or DCl. Ionic strength was maintained at 0.025 M by the addition of potassium chloride. [b] The actual values of the atom fraction of deuterium were determined by Josef Nemeth[15] and were 25% acetonitrile, 0.000, 0.247, 0.494, 0.741, and 0.988; 50% acetonitrile, 0.000, 0.246, 0.491, 0.737, and 0.982; 75% acetonitrile, 0.000, 0.244, 0.487, 0.731, and 0.974; 90% acetonitrile, 0.000, 0.243, 0.485, 0.728, and 0.970. [c] The rate constants in 0% acetonitrile were taken from ref 8 for comparison. All reported rate constants represent the average of at least three runs. Error limits are standard deviations.

Table III. Thermodynamic Activation Parameters for the Water-Catalyzed Hydrolysis of 2 as a Function of the Volume Percent Acetonitrile in Acetonitrile–Water Mixtures[a]

volume % H$_2$O–CH$_3$CN	$\Delta H^{\ddagger}$,[b] kcal/mol	$-\Delta S^{\ddagger}$,[c] eu	$\Delta G^{\ddagger}$,[d] kcal/mol
100–0	15.20	24.50 ± 0.01	22.90 ± 0.20
75–25	14.16	26.64 ± 0.04	22.55 ± 0.19
50–50	13.16	29.25 ± 0.06	22.38 ± 0.24
25–75	11.94	33.77 ± 0.03	22.45 ± 0.24
10–90	11.23	36.67 ± 0.06	22.79 ± 0.26

[a] The pH of the solutions was maintained at 3.0 by the addition of HCl. The ionic strength was maintained at 0.025 M by the addition of potassium chloride. [b] Calculated from kinetic rate constants measured at 33.65, 38.97, 44.50, and 50.02 °C. [c] $\Delta S^{\ddagger}$ is the mean of the $\Delta S^{\ddagger}$ values calculated at each of the above temperatures. [d] $\Delta G^{\ddagger}$ is the mean of the $\Delta G^{\ddagger}$ values calculated at each of the above temperatures.

Figure 2. Dependence of the free energy of activation of the water-catalyzed hydrolysis of 2 (circles), 3a (squares), and 3b (triangles) upon the percentage of acetonitrile in acetonitrile–water mixtures. The data for 3a and 3b are taken from ref 14 while those for 2 are from Table III.

Scheme I

Figure 1. Dependence of the observed first-order rate constants for the water-catalyzed hydrolysis of 2 upon the volume percentage of acetonitrile in acetonitrile–L$_2$O (L = H or D) mixtures of atom fraction of deuterium n = 0 (circles), 0.5 (squares), and 1.0 (triangles). The data are from Table I.

data are shown in Table III.

Discussion

Several interesting features are apparent from the data. The rate of hydrolysis of 2 increases with the increase in acetonitrile in the solvent mixture up to 50% and then decreases with any further increase in the volume percent acetonitrile. This type of bell-shaped curve is frequently observed in pH–rate profiles. In such cases, it is frequently interpreted as due to the change in rate-determining step in the reaction.[4] In the present case, we can also invoke such a concept to account for the observed bell-shaped rate versus water concentration curve (Figure 1). The general mechanism (Scheme I) for the hydrolysis of imines[5] as

formulated by Jencks in his pioneering efforts is one in which the rate-determining step is either step 1 or step 3, depending upon the acidity of the medium. Step 2 is intramolecular and fast under all conditions.

The present investigation was carried out in the pH-independent region (i.e. at pH 3) established by the earlier workers.[3a,b,6] The reaction is the neutral water-catalyzed hydrolysis. This is identical to step 1 of Scheme I when B is a water molecule and the transition state would be 1b. If we consider step 1 as rate-determining in the hydrolysis of 2 in solvent mixtures containing 50 or more percent of water, what other step is the rate-determining step in solvent mixtures with less than 50% water? It cannot be step 3 because the pH of the medium is maintained at 3.0 in all investigations.[5] It may be step 2 in Jencks' scheme for the reaction. The bell-shaped rate–water concentration plot (Figure 1) may also arise if two factors, which have opposite effects on the reaction rate, are operating to different extents in solvent mixtures containing different concentrations of water.

The variation in $\Delta G^{\ddagger}$ with the change in the concentration of water, observed in the hydrolysis of 2, is relatively small when compared with that observed in the hydrolysis of 3a and 3b (Figure 2). Inspection of the

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Table IV. Values of the Curvature Parameter, γ, and Isotopic Fractionation Factor ϕ_a for Transition State 5 and ϕ_a and ϕ_b for Transition State 1b or 6 Calculated for the Water-Catalyzed Hydrolysis of 2 in Acetonitrile–Water Mixtures

volume % water–CH$_3$CN	k_{H_2O}/k_{D_2O}	γ^a	5 ϕ_a	1b or 6	
				ϕ_a	ϕ_b
100–0	2.19	0.33 ± 0.04	0.768^b	0.673^{c,d}	0.824^{c,d}
75–25	2.07	0.40 ± 0.19	0.768	0.677	0.845
50–50	1.97	0.47 ± 0.14	0.764	0.650	0.884
25–75	2.01	0.55 ± 0.22	0.742	0.609	0.904
10–90	2.22	0.85 ± 0.13	0.657	0.484	0.969

^{a} The curvature parameter, γ, was calculated by using the computer program GAMISO1 provided by Albery.[7] ^{b} Values of ϕ_a for 5 were calculated from the γ values by using eq 2. ^{c} Values of ϕ_a and ϕ_b for 1b and 6 were calculated by using the equation $k_n = k_0 (1 - n + n\phi_a)(1 - n + n\phi_b)^2$. ^{d} It was necessary to use a value of $\gamma = 0.37$, the upper limit based on the error, in this particular case since $\gamma = 0.33$ gave undefined values.

variation of ΔH^* and ΔS^* for the hydrolysis of 2 reveals the existence of a compensatory effect. Surprisingly, the plot of $(\delta\Delta H^* - T\delta\Delta S^*)$ versus the percentage of water in the solvent is also bell-shaped. This clearly identifies two factors, enthalpy and entropy barriers, as being responsible for the observed variation of the rate with water concentration. However, it does not rule out the possibility of a change in the rate-determining step.

Another interesting result demonstrated by the data is the shape of the proton inventory plots (i.e. the plots of the rate constants, k_n, versus atom fraction of deuterium n in the solvent mixture). Although the plots (not shown) are all bowl-shaped, the extent of bowing is different for different solvent mixtures and it generally decreases with the decrease in water concentration in the solvent mixture as evident from the increase in the value of the curvature parameter γ (Table IV) calculated from eq 1. The in-

$$\gamma = 8 \ln [(k_{0.5}/k_0)/(k_1/k_0)^{1/2}]/[\ln (k_1/k_0)^2] \quad (1)$$

crease in the γ value, i.e. the decrease in the bowing, indicates the involvement of another rate-determining transition state with fewer or more protons contributing to the isotope effect than in the first transition state[7,13] or a decrease in the number of protons involved in the transition state. Since the increase is really significant below 50% water, it is clear that the second transition state comes into predominance only at lower water concentrations.

Another interesting feature is revealed (Table IV) by the values of the isotopic fractionation factors (ϕ_a and ϕ_b) of the H$_a$ and H$_b$ protons calculated on the assumption that the reaction proceeds through the transition state 1b. The fractionation factor ϕ_a of the proton (H$_a$) in flight remains virtually constant while that of the H$_b$ protons undergoes a very small increase for the reactions carried out in solvents with greater than 50% water. But, at low concentrations of water we observe a decrease in ϕ_a and an increase in ϕ_b. This situation is far from reality. This

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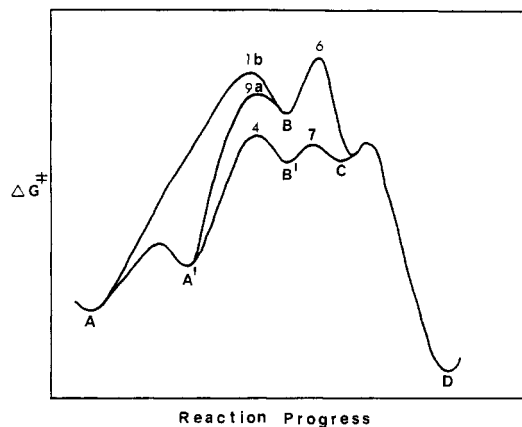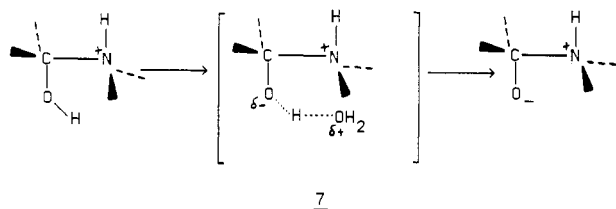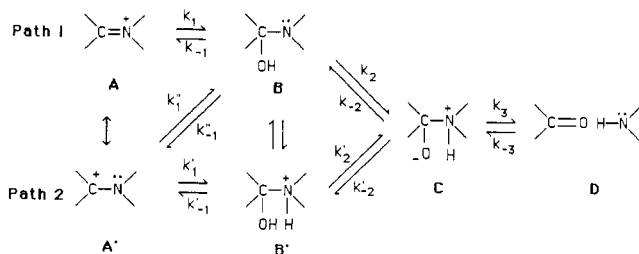(16) See paragraph at the end of paper regarding supplementary material.

Scheme II

6

probably arises because we treat two consecutive transition states as a single transition state for the purpose of calculating ϕ values.

Still another unusual feature revealed by the proton inventory data is the change in kinetic solvent isotope effect as a function of solvent composition. The effect decreases with the decrease of water concentration down to 50% and then increases with any further decrease in water concentration. It is perhaps significant in the present study that the kinetic solvent isotope effects for 3a and 3b appear to decrease continuously with the decrease in water concentration.[21,m]

The kinetic features observed in the present work may be accounted for if we make the following assumptions: (a) The neutral water-catalyzed hydrolysis of 2 occurs via a cyclic addition of water, as in 4, rather than by the acyclic process (1b) proposed in earlier works. The fourth-order dependence on water concentration that is observed in the hydrolysis of 2 in dilute formic acid, in fact, led us and Jencks to favor an acyclic transition state (5).[8,9] However, as Jencks warned, we should not attach to much importance to this order dependence, especially when we observe a bell-shaped rate versus water concentration plot.

4   5
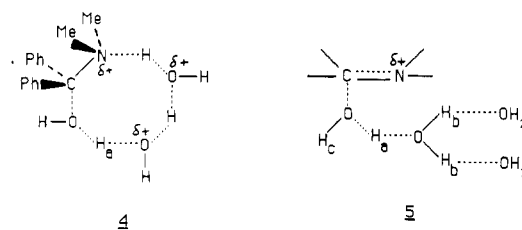
A factor that may seem to argue against 4 is the unfavorable nonbonded interaction between the two phenyl groups and two methyl groups in 4. If the system avoids this interaction by proceeding through 5 or 1b, it has to face it in the next step (i.e. in the formation of zwitterion (Scheme II) from the carbinolamine), which is considered to be fast and intramolecular in nature.

The Gross–Butler equation corresponding to transition state 4 is eq 2. This involves three protons with equal fractionation factors. The value of $\phi_a = 0.768$ reproduces

$$k_n = k_0(1 - n + n\phi_a)^3 \quad (2)$$

the rate constants observed in the present study. This lends support to the transition state 4. Furthermore, the presence of the water point well below the Brønsted line for catalysis of the reaction by carboxylate ions and oxygen bases suggests that the water-catalyzed reaction occurs by a process that is different from that catalyzed by the above bases.[6] The bases presumably replace the basic water molecule in 1b.

The cyclic process is possible if the carbon–nitrogen π-bond of 2 is completely polarized and the nitrogen is sp^3

Figure 3. Reaction progress diagram (see Scheme IV) for immonium ion hydrolysis.

Scheme III

7

Scheme IV

hybridized. This mean that the increase in acetonitrile concentration increases the rate by increasing the polarization of 2.

(b) The formation of the zwitterion from the carbinolamine occurs by an acyclic process if the addition of water to the immonium ion occurs by a cyclic process or vice versa. This assumption is necessary and justifiable since protonation at the nitrogen determines the cyclic or acyclic formation of the zwitterion (Schemes II and III).

Incorporation of these two assumptions into the Jencks' scheme (i.e. Scheme I) for the neutral hydrolysis of the immonium ion leads to Scheme IV. The schematic reaction progress diagram for this scheme is shown in Figure 3. The transition states of the k_1 and k_2 steps in path 1 are **1b** and **6**, respectively, and those of the k'_1 and k'_2 steps in path 2 are **4** and **7**, respectively. Path 1 is a higher energy path than path 2 for several reasons.

The transition state (**1b**) of the k_1 step in path 1 has higher free energy than the transition state **4** of the k'_1 step in path 2, in part, because of solvation effects.

In **1b**, the solvation at one end (i.e. at nitrogen) of the molecule interferes with solvation at the other end (**8a**) (i.e. at carbon) and destabilizes **1b**. But in **4**, the solvation at opposite ends of the molecule is reinforcing (as shown in **8b**) and stabilizes **4**.[10]

Also, in **1b**, the nitrogen undergoes rehybridization to sp^3. Thus, either the lone pair developing on nitrogen will be antiperiplanar to the newly forming C–O bond (**9a**) or the developing geometry will produce unfavorable non-

bonded interactions between the phenyl and methyl groups (**9b**). Either way, **1b** is destabilized. In **4**, the first effect is absent and the second effect can be assumed to be compensated by the favorable reinforcement of solvation mentioned above.
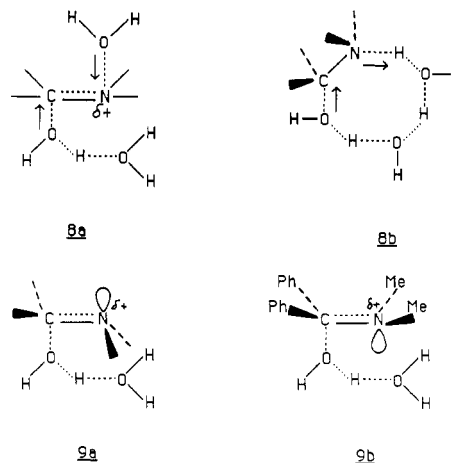
Although these reasons indicate that transition state **4** has less free energy than **1b**, it is clear that the energy difference may be very small. Hence, it is mainly the greater ΔG^* of formation of **1b** from the low energy A, rather than the effects mentioned above, that favors the formation of B' from the high energy species A'. For this reason, the formation of B from A' is only slightly less favorable than the formation of B' from A'.

The carbinolamine (B) has higher energy than its protonated form (B') since solvation at the nitrogen in B is less effective than in B'. This also favors path 2. The concentration of B will also be very low since it reverts to A more easily than does B' due to the fact that the lone pair on nitrogen is antiperiplanar to the C–O bond in B.

Lastly, the transition state (**6**) of the k_2 step in path 1 of Scheme IV is of higher energy than that (**7**) of the k'_2 step in path 2 because the unfavorable nonbonded interactions between the phenyl and methyl groups are absent in **7**, the solvation at nitrogen in **7** is greater than in **6**, and a lone pair of the hydroxyl oxygen in **7** is antiperiplanar to the C–N bond. The latter effect lengthens the C–N bond. This effect, in turn, enhances proton abstraction from the hydroxyl group in B'. But in **6** none of the lone pairs on the O–H oxygen can be antiperiplanar to the C–N bond due to the planar requirement.[11]

For the reasons cited above, the reaction is thought to proceed through path 2 of Scheme IV when water is present in high concentration. If we assume the steady state formation of B', the observed rate constant of the reaction is shown in eq 3.

$$k_{obsd} = k'_1 k'_2/(k'_{-1} + k'_2) = k'_1/[1 + (k'_{-1}/k'_2)] \quad (3)$$

At high concentrations of water, k'_2 is large and k'_{-1} is small for the reasons given earlier, and eq 3 reduces to eq 4. The corresponding kinetic solvent isotope effect is given by eq 5. The increase in the concentration of added

$$k_{obsd} = k'_1 \quad (4)$$

$$k_{H_2O}/k_{D_2O} = k'_1(H_2O)/k'_1(D_2O) \quad (5)$$

acetonitrile increases the polarization of A into A'. Hence, the rate acceleration observed on increasing the acetonitrile concentration at higher concentrations of water is due solely to the increase of polarization of A. Since this is a preactivation process, this is consistent with the observed constancy of ϕ_a values (Table IV) calculated on the basis of transition state **4** for reactions in 100, 75, and 50%

water. Since the formation of A′ involves the breaking of the carbon–nitrogen π-bond, this breaking is part of the reaction coordinate motion and acetonitrile enhances this conversion by replacing water solvation. This shows up in a greater decrease in $\Delta H^{\ddagger}$ than in $\Delta S^{\ddagger}$. This situation mimics the selective desolvation by an enzyme of a portion of a substrate not identical with the site of the reaction, leading to enhanced reactivity of the part remaining in water. Since protium oxide is a better base but a weaker acid than deuterium oxide,[12] the resonance between A and A′ is slightly more shifted toward A′ in deuterium oxide than in protium oxide due to favorable solvation at A′ as shown in 10. Since increasing concentrations of acetonitrile enhance the desolvation of 10a in deuterium oxide more than in protium oxide, $k(H_2O)/k(D_2O)$ decreases with the increasing concentration of acetonitrile.
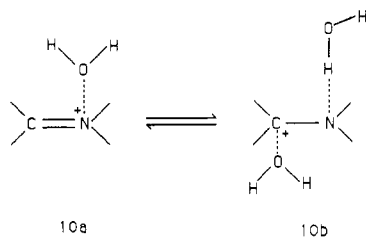
An increase in the concentration of acetonitrile above 50% in water–acetonitrile mixtures significantly decreases the three-dimensional network of hydrogen-bonded water molecules and reduces the role of water as a solvent. Under such conditions, the entropy barrier for the formation of 4 increases more than the decrease in the enthalpy barrier because the formation of 4 involves the transfer of three water molecules from less structured bulk solvent to a more structured transition state. Since the stability difference between the transition states 4 and 1b, as noted earlier, is small, this greater increase in the entropy barrier favors the formation of B rather than B′ from A′, and the reaction occurs through the acyclic transition state 9a, which is similar to 1b. The formation of C from B occurs through the cyclic transition state 6 as shown in Scheme II. This, again, is a high energy path because of promotion of significant solvent structuring at the transition state in the low structured solvent. As a result, the reaction that occurs along A → A′ → B → C → D has both k''_1 and k_2 steps of comparable rates or a k_2 step slower than k''_1. Either way, the reaction involves two consecutive transition states 9a and 6, of steps k''_1 and k_2, respectively. This is evident from the increase in the value of the curvature parameter observed in 25% and 10% water. The Gross–Butler equation for the solvent isotope effect of this reaction with two consecutive transition states is shown in eq 6. Here x and y are the numbers of protons in the

$$k_n = k_0 \frac{(1 + k''_{-1,0}/k_{2,0})}{[(1 - n + n\phi_a)^{-x} + (k''_{-1,0}/k_{2,0})(1 - n + n\phi_b)^{-y}]} \quad (6)$$

transition states 9a and 6 respectively, ϕ_a and ϕ_b are the fractionation factors of the proton in 9a and 6, respectively, and $k''_{-1,0}$ and $k_{2,0}$ are the rate constants of the steps k''_{-1} and k_2 (Scheme IV), respectively, in a solvent with $n = 0$.[13] Though the dependence of the solvent isotope effect on

n is complex, it is readily understandable from eq 6 that the increase in the $k''_{-1,0}/k_{2,0}$ value results in an increase in the solvent isotope effect and a decrease in the value of curvature parameter γ. This can be verified by substituting hypothetical values of ϕ_a, ϕ_b, x, and y into the equation. This suggests that the increase in the solvent isotope effect and the decrease in the γ value observed with the increase of acetonitrile in the solvent mixtures containing less than 50% water is due to the existence of the consecutive transition states in the reaction path. Furthermore, it indicates that the value of $k''_{-1,0}/k_{2,0}$ increases with the decrease in water concentration. This means the decrease in water concentration increases k''_{-1} or decreases k_2, or both, at lower concentrations of water. In the present case, since reversion of B to A′ is favored stereoelectronically by the lone pair on nitrogen (9a) and the conversion of B into C through transition state 6 is progressively inhibited by the increasing entropy barrier, the increase in $k''_{-1,0}/k_{2,0}$ value is the result of the decrease of $k_{2,0}$ and increase of $k''_{-1,0}$. Under these conditions eq 3 becomes eq 7. This accounts for the decrease in rate observed with the decrease of water at lower concentrations of water.

$$k_{obsd} = k''_1/(1 + k''_{-1}/k_2) \quad (7)$$

The results and conclusions of the present study, especially those at low concentrations of water, are very important in understanding many biologically important reactions, such as aldolization, decarboxylation, and the visual process that involve the formation and hydrolysis of imines since these are enzymatic reactions, and the interior of many enzymes is at least partially nonaqueous in character.

Experimental Section

Materials. The iodide salt of the dimethylimmonium ion of benzophenone (2) was prepared as described previously.[8] Deuterium oxide (99.8 atom % deuterium; Aldrich) and deuterium chloride (20% solution in D_2O; Aldrich) were used as obtained. Potassium chloride was oven-dried. Acetonitrile was distilled from phosphorous pentoxide and stored under nitrogen.

Solutions. Solvent mixtures containing the desired volume percentage acetonitrile in protium oxide or deuterium oxide were adjusted to pH(D) 3.0 by the addition of HCl or DCl as appropriate. The definition of "pH" in such a medium is a problem. However, we found that the pH-independent region remains almost the same at different volume percent of acetonitrile. Appropriate volumes of these stock solutions were used to prepare the protium oxide–deuterium oxide mixtures.

Kinetics. The hydrolysis reactions were monitored at 300 nm by following the absorbance change with a Cary 118C UV–vis spectrophotometer equipped with a constant-temperature cell compartment. Reactions were initiated by injections of 25 μL of a 1.5×10^{-2} M solution of 2 in acetonitrile into 3.0 mL of the reaction solution. Rate constants were calculated by using a nonlinear least-squares computer program.