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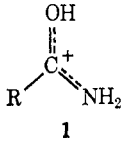
Position of Protonation and Mechanism of Hydrolysis of Simple Amides

Sir:

The nmr spectra of simple amides dissolved in very acidic solvents such as 100% H_2SO_4[1] or FSO_3H[2,3] provide strong evidence that the principal conjugate acids of these substances in these media are the oxygen-protonated species, **1**. In less acidic solvents, rapid exchange of the added proton prevents a similar positive structural assignment; nevertheless, it has been widely assumed that protonation occurs chiefly on oxygen under these conditions as well.[4]

Quite recently, however, this assumption was questioned, and an argument was advanced favoring predominant formation of the nitrogen-protonated species, **2**, in dilute or moderately concentrated aqueous acids.[3,5]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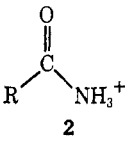
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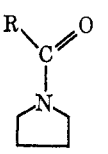
This proposal has aroused considerable interest and has generated a number of counter arguments.[6] None of these rebuttals, however, is free of unproven assumptions, and the issue therefore still remains unsettled. We wish here to describe new experimental work which provides an especially clear answer to the question of the position of protonation of amides in dilute and moderately concentrated aqueous acids and has an important bearing on the mechanism of acid-catalyzed amide hydrolysis as well.

Our study is based upon the well-known ring-size effect of five- and six-membered cyclic structures commonly called I strain: five-membered rings resist a change in hybridization of one of the ring atoms from sp^2 to sp^3, whereas six-membered rings favor such a process.[7] We applied this effect to the present problem by examining the behavior of amides in which the nitrogen atom is incorporated in five- and six-membered rings, **3** and **4**.[8] Since the amide group is planar, or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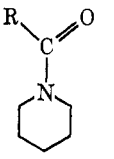
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very nearly so,[10] the nitrogen atom in unprotonated amides is sp^2 hybridized. Protonation on nitrogen makes this atom tetravalent and changes its hybridization to sp^3; *N*-acylpyrrolidines, **3**, should therefore be weaker nitrogen bases than *N*-acylpiperidines, **4**. Protonation on oxygen, on the other hand, simply reinforces the conjugation which gives the nitrogen atom in the free bases sp^2 hybridization, and O protonation should therefore show no ring-size effect. These phenomena may be seen, for example, in aniline derivatives with planar nitrogen such as *N*-(*p*-nitrophenyl)pyrrolidine ($pK_a = -1.1$)[11] and *N*-(*p*-nitrophenyl)piperidine ($pK_a = +1.8$);[11] here the five-membered cyclic amine is nearly three orders of magnitude less basic than its six-membered homolog. On the other hand, the basic strengths of the parent amines, pyrrolidine ($pK_a = 11.31$)[12a] and piperidine ($pK_a = 11.12$),[12b] are nearly identical, for here the nitrogen atom is already pyramidal in the free amine and no change occurs upon protonation.

We examined the effect of ring size upon basicity in two series of substrates, the acetamides, **3** and **4** R = CH_3, and the *p*-nitrobenzamides, **3** and **4** R = *p*-$NO_2C_6H_4$. Basic strengths were measured in dilute to moderately concentrated aqueous $HClO_4$ by standard indicator methods, making use of the decrease in uv absorption which these amides show upon protonation. Ratios of concentrations of protonated to unprotonated amide, C_{BH^+}/C_B, were determined at a number of points over the range $C_{BH^+}/C_B \simeq 0.1$–10; these values were then extrapolated down to the point where acidity functions cease to deviate from C_{H^+}, *i.e.*, down

Table I. Basicities and Rates of Hydrolysis of Cyclic Amides

Substrate	pK_a[a]	10^6k,[b] sec^{-1}
N-Acetylpyrrolidine	0.08 ± 0.02	8.5 ± 0.9
N-Acetylpiperidine	0.04 ± 0.02	43.9 ± 0.2
N-(*p*-nitrobenzoyl)pyrrolidine	−1.97 ± 0.03	
N-(*p*-nitrobenzoyl)piperidine	−2.20 ± 0.03	

[a] Measured in aqueous $HClO_4$ at 25°. [b] Measured in 20 wt % aqueous $HClO_4$ at 81.9°.

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(2) R. J. Gillespie and T. Birchall, *Can. J. Chem.*, **41**, 148 (1963); T. Birchall and R. J. Gillespie, *ibid.*, **41**, 2642 (1963).
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(5) M. Liler, "Reaction Mechanisms in Sulfuric Acid," Academic Press, New York, N. Y., 1971, p 106; *J. Chem. Soc., Chem. Commun.*, 527 (1972).
(6) H. Benderly and K. Rosenheck, *J. Chem. Soc., Chem. Commun.*, 179 (1972); C. R. Smith and K. Yates, *Can. J. Chem.*, **50**, 771 (1972); R. B. Martin, *J. Chem. Soc., Chem. Commun.*, 793 (1972); R. B. Martin, and W. C. Hutton, *J. Amer. Chem. Soc.*, **94**, 4752 (1972); A. C. Hopkinson and I. G. Csizmadia, *Can. J. Chem.*, **51**, 1432 (1973). See also A. R. Fehrst, *J. Amer. Chem. Soc.*, **93**, 3504 (1971); R. S. Molday and R. G. Kallen, *ibid.*, **94**, 6739 (1972).
(7) H. C. Brown, R. S. Fletcher, and R. B. Johannesen, *J. Amer. Chem. Soc.*, **73**, 212 (1951); H. C. Brown, J. H. Brewster, and H. Shechter, *J. Amer. Chem. Soc.*, **76**, 467 (1954).
(8) The protonation of cyclic amides in which the carbonyl group as well as the nitrogen atom is in the ring, *i.e.*, lactams, had been studied before.[9] In these substances, however, two ring atoms are initially sp^2; simple I-strain concepts therefore do not apply, and comparison with the behavior of polymethylenimines, as was made in that work, is not completely valid.
(9) R. Huisgen, H. Brade, H. Walz, and I. Glogger, *Chem. Ber.*, **90**, 1437 (1957).
(10) F. K. Winkler and J. D. Dunitz, *J. Mol. Biol.*, **59**, 169 (1971).
(11) J. W. Eastes, M. H. Aldridge, R. R. Minesinger, and M. J. Kamlet, *J. Org. Chem.*, **36**, 3847 (1971).
(12) R. M. Izatt and J. J. Christensen, "Handbook of Biochemistry," 2nd ed, Chemical Rubber Co., Cleveland, Ohio: (a) p J-152; (b) p J-141.

to $H = -\log C_{H^+} = +1.00$, and pK_a's were calculated. The results (Table I) show that the two acetamides have basic strengths which are identical within the (rather small) experimental error and that the acidity constants of the *p*-nitrobenzamides differ by less than a factor of 2. These effects are clearly incompatible with N protonation, but they are just what is expected for O protonation.

The two cyclic acetamides were also used to determine the effect of ring size upon the rate of acid-catalyzed amide hydrolysis. The reactions were monitored by nmr spectroscopy, using the change in acetyl group proton signal which occurs as the amides are converted into acetic acid. The results are summarized in Table I.

These rate measurements were made in 20% aqueous $HClO_4$ where both amides are essentially completely protonated; the initial states of the reactions are therefore the O-protonated conjugate acids with fully sp^2 hybridized nitrogen atoms. The fact that the five-membered ring substrate reacts more slowly than its six-membered homolog indicates that a change in hybridization occurs as hydrolysis takes place. The rate factor, however, is only 5, which is much less than the approximately thousandfold difference shown by the *N*-(*p*-nitrophenyl)amines for the full hybridization change, $sp^2 \rightarrow sp^3$,[11] and that suggests that the change in hybridization is only partly complete at the hydrolysis transition state. This is incompatible with either of the two reaction mechanisms commonly advanced for amide hydrolysis *via* N-protonated species: (1) rate-determining formation of a tetrahedral intermediate from the N-protonated cation or (2) direct displacement of nitrogen by water in this ion.[13] In both of these mechanisms, the N-protonated cation is formed, and the change to sp^3 hybridization is thus complete, *before* the rate-determining step. A small rate factor, on the other hand, *is* compatible with rate-determining formation of a tetrahedral intermediate from the O-protonated cation. In such a mechanism, conjugation between the nitrogen atom and the carbonyl group is being destroyed in the rate-determining step; this converts the nitrogen atom from an amide nitrogen into an amine nitrogen, which changes its hybridization from sp^2 to sp^3. This change, however, can be no more than partly accomplished at the rate-determining transition state, and that leads to a rate factor considerably less than the full hybridization change effect.

The results of this investigation thus favor O-protonated species, and discredit N-protonated conjugate acids, not only as the principal products of the equilibrium protonation of amides in dilute and moderately concentrated aqueous acids but also as essential intermediates in acid-catalyzed amide hydrolysis.

Acknowledgment. We are grateful to the National Science Foundation for financial support of this work and to Professor B. M. Wepster for correspondence concerning the ring-size effect.

(13) For a particularly good recent discussion of the various mechanisms for acid-catalyzed amide hydrolysis, see C. R. Smith and K. Yates, *J. Amer. Chem. Soc.*, **94**, 8811 (1972).

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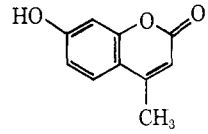
Excited-State Reactions of a Laser Dye. Evidence for a Two-Step Phototautomerism in 7-Hydroxy-4-methylcoumarin

Sir:

Previously[1] we have shown that 7-hydroxy-4-methylcoumarin (I) exhibits a variety of fluorescence spectra



I

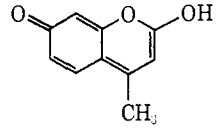
depending on the acidity of the solution being tested. These observations led to the construction of a tunable dye laser in which laser action from the near uv (390 nm) to the yellow (600 nm) was obtained from a single solution of I which was pumped by a nitrogen laser. Three different fluorescent species were identified: the "neutral" form I* which has excitation and emission maxima at 320 and 387 nm, respectively; the "basic" form (anion) II*, λ_{max}^{excit} 364 nm, λ_{max}^{emiss} 445 nm; and the "acid" form III*, λ_{max}^{excit} 320 nm, λ_{max}^{emiss} 485 nm. The formation of the "acid" form originally was attributed to an excited state protonation reaction in which the product appeared to have no corresponding stable ground state.[1,2]

$$I \xrightarrow{h\nu} I^*$$

$$I^* + H^+ \longrightarrow (I\cdots H^+)^*$$

$$(I\cdots H^+)^* \longrightarrow I + H^+ + h\nu\ (485\ nm)$$

Subsequent experiments[3] on the time-resolved spectroscopy of stimulated fluorescence from the "acid" form III* furnished additional evidence that an excited state reaction was occurring. Independently, Nakashima, *et al.*,[4] and Yakatan, *et al.*,[5] have suggested that the "acid" form III is a phototautomer of I.



III

We have now further elucidated the nature and formation of the excited states, I*, II*, and III*, by the use of gain spectroscopy.[6] (The gain spectrum of a fluorescent molecule differs from the fluorescence spectrum which would be measured in an ordinary spectrophotofluorometer, in that it represents the efficiency of *stimulated* fluorescence rather than spontaneous fluorescence over the wavelength range.) In addition to supporting the assignment of III as a phototautomer,[4,5] we have been able to demonstrate that the phototautomerization

(1) C. V. Shank, A. Dienes, A. M. Trozzolo, and J. Myer, *Appl. Phys. Lett.*, **16**, 405 (1970).
(2) The instability of the ground state which is meant to be conveyed here is that the "equilibrium" concentration of the ground state is smaller than can be detected by absorption measurements.
(3) A. Dienes, C. V. Shank, and A. M. Trozzolo, *Appl. Phys. Lett.*, **17**, 189 (1970).
(4) M. Nakashima, J. A. Sousa, and R. C. Clapp, *Nature (London)*, **235**, 16 (1972).
(5) G. J. Yakatan, R. J. Juneau, and S. G. Schulman, *Anal. Chem.*, **44**, 1044 (1972).
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