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One-Electron Transfer Reactions of 10-Methyl-9,10-dihydroacridine and the Kinetic Isotope Effects for the Intervening Ionization and Protonation of a Carbon Acid

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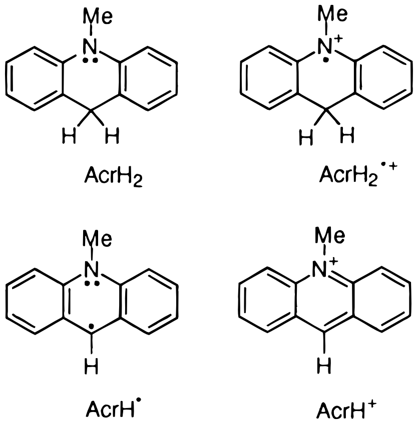
Oxidations of 10-methyl-9,10-dihydroacridine, $AcrH_2$, with transition-metal reagents that require single-electron processes were studied in acetonitrile/water, 20/80 by volume. The reaction proceeds in three stages: (1) electron transfer yielding $AcrH_2^{\bullet +}$, a very rapid step for Ce_{aq}^{4+} and $IrCl_6^{2-}$ but rate-controlling for Fe_{aq}^{3+} (k_{298}/L mol^{-1} s^{-1} = 37.9 at μ = 0.1 M and 128 at μ = 1.0 M); (2) reversible and relatively slow ionization/protonation of $AcrH_2^{\bullet +}/AcrH^{\bullet}$; and (3) the final highly exoergic and diffusion-controlled oxidation of $AcrH^{\bullet}$ to $AcrH^+$, the stable product. The second step corresponds to ionization/protonation at carbon. Four isotopically distinct versions of the reactions could be examined, $AcrL_2$ and L_2O/L_3O^+ (L = H, D) in every combination. The primary, secondary, and solvent isotope effects on the rate constants in each direction are all appreciable, and they were resolved by the indicated experiments.

Introduction

Dihydronicotinamide adenine dinucleotide (NADH) has attracted continuing interest owing to its participation in biological electron-transfer reactions.[1−4] In various enzymatic processes, NADH and its analogues serve as one- and two-electron reductants. Owing to the instability of NADH toward acid-catalyzed hydrolysis,[5] many mechanistic studies have employed 10-methyl-9,10-dihydroacridine ($AcrH_2$, **Chart 1**) and its substituted analogues,[6] because they appear otherwise analogous and yet are stable in acidic aqueous and semiaqueous media.[5] Such reactions include various organic and inorganic oxidizing reagents.[7,8] We have used $AcrH_2$ here, not only because some of the reaction partners require the presence of acid but also even more because the effect of variable acid concentration on the conjugate acid and base forms of the intermediate radical state is of interest.

Chart 1



Single electron transfer, hydride ion transfer, and hydrogen atom transfer mechanisms have all been proposed for biological and artificial redox reactions of NADH and its analogues; indeed, the different partners will surely require different mechanisms. The distinction between electron-transfer and hydride-transfer mechanisms, once a point of confusion, has now been carefully made.[9,10] Hydride transfer often prevails except when the reaction partner is, like $Fe(CN)_6^{3-}$ and $Fe(phen)_3^{3+}$, limited to a 1e mechanism.[8,11] Although all enzy-

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matic reactions of pyridine mononucleotides proceed by hydride transfer, the 1e reactions of NADH model compounds such as $AcrH_2$ are of great interest. The partners in this work were deliberately chosen so that the reactions would always proceed along 1e pathways.

The reaction between several dihydropyridine derivatives and ferricyanide ions in aqueous acetonitrile features two 1e oxidations giving the pyridinium ions; between these steps, the ionization of a proton from a carbon atom occurs. In this sequence, shown in eq 1, the first step is rate-controlling.[7]

$$PyH_2 \underset{Fe(CN)_6^{4-}}{\overset{Fe(CN)_6^{3-}}{\rightleftharpoons}} PyH_2^{\bullet +} \underset{+H^+}{\overset{-H^+}{\rightleftharpoons}} PyH^{\bullet} \xrightarrow{Fe(CN)_6^{3-}} PyH^+ + Fe(CN)_6^{4-} \quad (1)$$

In a 1e pathway involving $AcrH_2$, a dihydroacridine radical cation ($AcrH_2^{\bullet +}$, Chart 1) will be an intermediate. Such a case is the reaction with tris(phenanthroline)iron(3+) ion, for which a similar mechanism, given in eq 2, has been proposed.[8]

$$AcrH_2 \xrightarrow{Fe(phen)_3^{3+}} AcrH_2^{\bullet +} \underset{+H^+}{\overset{-H^+}{\rightleftharpoons}} AcrH^{\bullet} \xrightarrow{Fe(phen)_3^{3+}} AcrH^+ + Fe(CN)_3^{2+} \quad (2)$$

The intermediate cation radical was detected by optical and EPR measurements carried out in acetonitrile solution.[8] The intermediate radical cation was identified by UV−vis and EPR spectroscopy. Also, the pK_a of the radical cation was determined; in CH_3CN, pK_a = 8.1−6.8 (0.03−0.30 M H_2O). In this mechanism, the deprotonation of the radical cation was rate-controlling, both electron-transfer steps being very rapid in comparison.

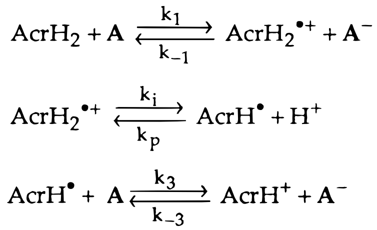
Although the outlines of the mechanism of $AcrH_2$ oxidation in nonaqueous solutions now seem to be well-understood, relatively little is known about the kinetic behavior of dihydroacridines in aqueous and semiaqueous media. It is certainly desirable to obtain more experimental data for these types of $AcrH_2$ reactions, considering the biological importance of aqueous chemistry.

In this study we have employed several metal complexes to develop a unified mechanistic picture of the oxidation of $AcrH_2$ in aqueous acetonitrile. By systematically varying the strength of an inorganic oxidant, it is possible to cover different modes of the oxidation mechanism. A further resolution of the chemistry of the $AcrH_2^{\bullet +}$ radical cation was also sought, including its pK_a, its standard reduction potential, and the kinetics of the acid-ionization reaction. The last feature is particularly interesting in that hydrogen−deuterium kinetic isotope effects could be determined for this relatively slow reacting carbon acid.

Experimental Section

Materials. 10-Methyl-9,10-dihydroacridine ($AcrH_2$) and its 9,9′-dideuterated analogue ($AcrD_2$) were prepared by the reduction with $LiAlH_4$ or $LiAlD_4$, according to the literature procedure.[12] 10-Methylacridinium iodide was prepared by the reaction of acridine with methyl iodide.[13] Lithium perchlorate was obtained from Aldrich and recrystallized three times from water. Other materials were obtained commercially. The purity of the organic materials was checked by ^1H NMR.

Scheme 1



$$AcrH_2 + \mathbf{A} \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} AcrH_2^{\bullet +} + \mathbf{A}^-$$

$$AcrH_2^{\bullet +} \underset{k_p}{\overset{k_i}{\rightleftharpoons}} AcrH^{\bullet} + H^+$$

$$AcrH^{\bullet} + \mathbf{A} \underset{k_{-3}}{\overset{k_3}{\rightleftharpoons}} AcrH^+ + \mathbf{A}^-$$

Kinetic Studies. A solvent mixture consisting of H_2O/CH_3CN (80/20 by volume) was used; the organic component was needed for the solubility of the dihydroacridines. Ionic strength was maintained at 1.0 M with lithium perchlorate. The studies were carried out spectrophotometrically with a Shimadzu UV-3101 instrument. Most of the kinetics required anaerobic stopped-flow (SF) techniques, for which an Applied Photophysics instrument was used, with the temperature controlled at 25.0 ± 0.2 °C. Even then, the initial portion of the fastest experiments was lost in the mixing time, about 2 ms. Concentrations of certain reagents were determined spectrophotometrically from parameters in the literature[14,15] or determined in the course of this work:

	ϵ/L mol^{-1} cm^{-1}
$AcrH_2$	$\epsilon_{285} = 1.32 \times 10^4$
$IrCl_6^{2-}$	$\epsilon_{487} = 4.075 \times 10^3$
$Fe(H_2O)_6^{3+}$	$\epsilon_{240} = 4.16 \times 10^3$
$AcrH^+$	$\epsilon_{358} = 1.88 \times 10^4$
	$\epsilon_{417} = 3.94 \times 10^3$
$AcrH_2^{\bullet +}$	$\epsilon_{640} = 6.44 \times 10^3$

Kinetic traces following the buildup of $AcrH^+$ at 358 or 417 nm, and the loss of $AcrH_2^{\bullet +}$ at 640 nm, were analyzed with the KaleidaGraph program. The program KinSim was used to simulate concentration−time curves for complex schemes, and its relative FitSim to determine one rate constant in a complex scheme, given the values of the other constants and the starting concentrations with global fitting of the data from a family of experiments.

Cyclic Voltammetry. A BAS-100 electrochemical analyzer was used. It was equipped with a three-electrode system, a glassy carbon or platinum disk working electrode, a platinum wire counter electrode, and a saturated calomel electrode (SCE) as reference. The SCE was separated from the bulk of the solution by a fritted-glass bridge of low porosity that contained the solvent and supporting electrolyte, which was *tert*-$Bu_4N^+PF_6^-$ in acetonitrile and sodium perchlorate in aqueous acetonitrile.

Results

General Observations. The family of net reactions being reported in this study consists of the oxidation of 10-methyl-9,10-dihydroacridine to the acridinium ion by metal complexes (denoted as electron acceptors, **A**) that are limited to single-electron steps. The net equation is

$$AcrH_2 + 2\mathbf{A} = AcrH^+ + 2\mathbf{A}^- + H^+ \quad (3)$$

A unified set of reactions is shown in Scheme 1. The first and

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third steps are electron-transfer reactions in the cases examined, with the unusual acid ionization step intervening. The thermodynamics of these steps is determined by the electrode potentials for the half-reactions $AcrH_2^{\bullet+} + e^- = AcrH_2$ ($E_1°$) and $AcrH^+ + e^- = AcrH^\bullet$ ($E_3°$). The value of $E_1°$ was reported as 0.81 V versus SCE in acetonitrile.[8] The same reaction, examined by slow-scan cyclic voltammetry in an argon atmosphere, exhibited an anodic wave with a current maximum of 0.81 V versus SCE in acetonitrile and 0.46 V versus SCE in 20% acetonitrile/80% water, the solvent used in this study. The other couple has $E_3° = -0.43$ V versus SCE in acetonitrile, reflecting the strongly reducing nature of the $AcrH^\bullet$ radical.[8]

Reactions of $AcrH_2$ with Cerium(IV). The reactions of $AcrH_2$ with Ce(IV) as **A** were carried out by use of the SF technique. The system was maintained under an argon atmosphere since the radical intermediates are oxygen-sensitive. The reactions were monitored at 358 and 417 nm, following $AcrH^+$, and at 640 nm, following $AcrH_2^{\bullet+}$. Data taken at two of these wavelengths are shown for a typical experiment in Figure S-1 (Supporting Information).

At all three wavelengths the same kinetics was found. The implication is that the first step (Scheme 1) occurs within the mixing time, and the kinetics is controlled by the last two steps. The rate equation at fixed $[H^+]$ is given by

$$v = \frac{k_i k_3 [AcrH_2^{\bullet+}][Ce_{aq}^{4+}]}{k_p[H^+] + k_3[Ce_{aq}^{4+}]} \tag{4}$$

The kinetic dependence on $[H^+]$ is more complex than implied by this expression, however, since Ce_{aq}^{4+} and $CeOH^{3+}$ are species of appreciable acidity. The acid ionization constants are[16]

$$Ce_{aq}^{4+} \underset{+H^+}{\overset{+H_2O}{\rightleftharpoons}} CeOH^{3+} \underset{+H^+}{\overset{+H_2O}{\rightleftharpoons}} Ce(OH)_2^{2+}$$

$$K_1 = 13 \text{ mol L}^{-1};\ K_2 = 0.3 \text{ mol L}^{-1} \tag{5}$$

We next postulate, as justified later, that only the aquacerium(4+) ion is the reactive partner. The rate law is then

$$k_{obs} = \frac{v}{[AcrH_2^{\bullet+}]} = \frac{k_i k_3 [Ce(IV)]_T}{k_p\left\{[H^+] + K_1 + \frac{K_1K_2}{[H^+]}\right\} + k_3[Ce(IV)]_T} \tag{6}$$

In keeping with this, at low concentrations of cerium(IV), <~0.2 mM, the reaction was first-order with respect to the concentrations of both $AcrH_2^{\bullet+}$ and cerium(IV). Figure 1 shows a plot of the second-order rate constants ($=k_{obs}/[Ce(IV)]_T$) as a function of the acid concentration. With the assumption that the last denominator term of eq 6 is negligible compared to the other terms at low ceric concentrations, the $[H^+]$-dependence of the rate constant is given by

$$k = \frac{k_{obs}}{[Ce(IV)]_T} \cong \frac{k_i k_3/k_p}{[H^+] + K_1 + \frac{K_1K_2}{[H^+]}} \tag{7}$$

A nonlinear least-squares treatment of the data defines the composite rate constant as $k_i k_3/k_p = (3.04 \pm 0.02) \times 10^7\ s^{-1}$. For reasons given in the Discussion, we assign k_3 as the diffusion-controlled value, which we take to be the same as the aqueous value, 7.4×10^9 L mol^{-1} s^{-1}. This analysis yields $pK_a(AcrH_2^{\bullet+}) = -\log(k_i/k_p) = 2.39$.

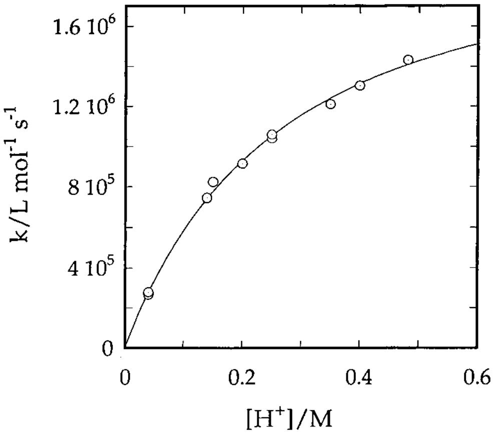

Figure 1. Variation of the second-order rate constant with $[H^+]$ for the reaction between $AcrH_2$ (1.5×10^{-5} M) and Ce_{aq}^{4+} (2×10^{-4} M) at $\mu = 1.0$ M. The line represents the fit to eq 7, with fixed values $K_1 = 13$ mol L^{-1} and $K_2 = 0.3$ mol L^{-1}.

At higher concentrations of cerium(IV), rate saturation was observed, as expected from eq 6. A reciprocal plot of $1/k_{obs}$ versus $1/[Ce(IV)]_T$, according to eq 8, gave $k_i = 721 \pm 29\ s^{-1}$

$$\frac{1}{k_{obs}} = \frac{1}{k_i} + \frac{k_p\left\{[H^+] + K_1 + \frac{K_1K_2}{[H^+]}\right\}}{k_i k_3} \frac{1}{[Ce(IV)]_T} \tag{8}$$

and, with k_3 again taken as 7.4×10^9 L mol^{-1} s^{-1}, $k_p = (1.33 \pm 0.03) \times 10^5$ L mol^{-1} s^{-1}. A global fit to both variables simultaneously was then made with a plot of k_{obs} versus the quotient $\{[H^+] + K_1 + K_1K_2/[H^+]\}/[Ce(IV)]_T$, as presented in Figure 2. This method of analysis gave $k_i = 691 \pm 36\ s^{-1}$ and $k_p = (1.18 \pm 0.11) \times 10^5$ L mol^{-1} s^{-1}. The values are all the same within less than 3 standard deviations. The global fit represents the most general treatment and provides the values that will be cited hereinafter. The more accurate value of pK_a of $AcrH_2^{\bullet+}$ is then 2.23 ± 0.05.

Kinetic Isotope Effects in Cerium(IV) Reactions. The reaction with $AcrD_2$ was carried out in D_2O/CH_3CN, with a variation of [Ce(IV)] at fixed $[D^+] = 0.400$ M. The values ${}^{D}k_i^{D} = 106 \pm 6\ s^{-1}$ and ${}^{D}k_p^{D} = (3.41 \pm 0.58) \times 10^4$ L mol^{-1} s^{-1} were obtained by fitting the data to eq 6, taking as the values of K_1 and K_2 those cited previously.

To bring out certain other features of the kinetic isotope effects, similar experiments were also performed for $AcrD_2$ in H_2O/CH_3CN. Least-squares fitting gave "k_i" $= 115 \pm 3\ s^{-1}$ and "k_p" $= (2.23 \pm 0.25) \times 10^4$ L mol^{-1} s^{-1}. Corresponding data were obtained for $AcrH_2$ in D_2O/CH_3CN, "k_i" $= 688 \pm 22\ s^{-1}$ and "k_p" $= (9.77 \pm 0.67) \times 10^4$ L mol^{-1} s^{-1}. The interpretation of these rate constants in terms of kie's is less straightforward, in that several reactions enter, and the analysis will be deferred to the Discussion.

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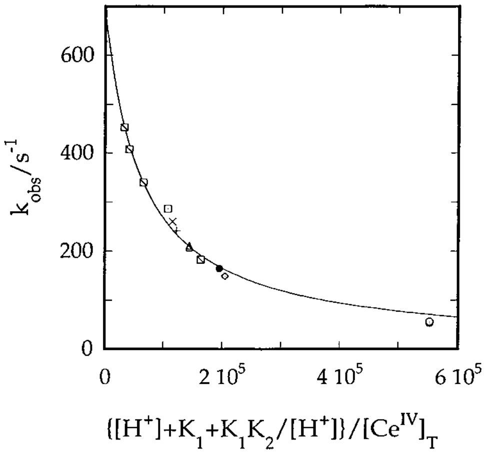

Figure 2. Global fit of rate constants to the variations of cerium(IV) and acid concentration according to eq 6. The different symbols represent values of $[H^+]$ in the range 0.040−0.482 M.

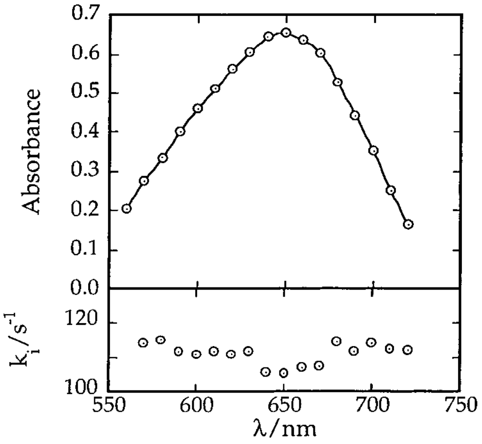

Figure 3. The spectrum of $AcrD_2^{•+}$ is given by the absorbance change in the reaction between 1×10^{-4} M $AcrD_2$ and 4×10^{-3} M Ce(IV) at 0.40 M H_3O^+ and $\mu = 1.0$ M. The lower portion shows the rate constants k_i at the different wavelengths and the small systematic drift of the values. The most reliable rate constants are considered to be those in the center range where the absorbance changes are the largest.

Kinetics at Higher Cerium(IV) Concentration. At a sufficiently high concentration of ceric ion, k_{obs} can be approximated as k_i. Since the protio system has a value of k_i that nearly exceeds the limit of the SF apparatus, experiments were done with $AcrD_2$ in D_2O/CH_3CN (80/20). The radical cation $AcrD_2^{•+}$ was monitored directly in the region 560−720 nm. The rate constants at those wavelengths with the greatest absorbance changes are 106 ± 3 s^{-1}, but values some 1−10% higher were recorded at wavelengths with small absorbance changes. The agreement of this rate constant with those from the fitting procedure at low and variable cerium concentrations serves to substantiate the earlier treatment. This is illustrated by Figure 3.

Reactions of $AcrD_2$ with Hexachloroiridate(IV). The stoichiometry of the reaction with **A** = $IrCl_6^{2-}$ was determined spectrophotometrically under anaerobic conditions. A small excess of $IrCl_6^{2-}$ was allowed to react with $AcrD_2$, the spectrum of the solution being recorded before and after the addition. The excess of $IrCl_6^{2-}$ was assayed at 487 nm. The spectrum of the product, obtained by subtracting the calculated $IrCl_6^{2-}$ absorbance at each wavelength, matched the independent spectrum of $AcrD^+$. The stoichiometry showed a value for $IrCl_6^{2-}/AcrD_2$ of 1.95/1 under air-free conditions. Repetitions under air gave ratios in the range (1.4−1.5)/1, dependent on the concentration of the iridium species taken, indicating partial oxygen scavenging of the intermediate radicals, which is not unexpected.

In one set of experiments, $AcrH_2$ (2 μM) and $IrCl_6^{2-}$ (5 μM) were used in H_2O/CH_3CN. The acid concentrations were 0.2−0.5 M, to provide balanced competition between the k_p and k_3 steps. The rate constants approached or exceeded the limit of the SF instrument, because $k_i^H > k_i^D$, but the values of k_{obs} were ∼200−500 s^{-1}, increasing as $[H^+]$ decreased, all consistent with expectations.

The next set of experiments used $AcrD_2$. The reaction kinetics was studied in H_2O/CH_3CN with these conditions: 0.300−1.00 mM $IrCl_6^{2-}$, 50.0 μM $AcrD_2$, and 0.100 M $HClO_4$ at $\mu = 1$ M. The reaction progress was monitored at 358 ($AcrD^+$) and 650 ($AcrD_2^{•+}$) nm. The reaction followed first-order kinetics at each wavelength, with $k_{obs} = 115 \pm 2$ s^{-1}, independent of concentrations of the iridium complex and acid.

Reaction with Hexaaquairon(III). The reactions of $AcrH_2$ and $AcrD_2$ with $Fe(H_2O)_6^{3+}$ in H_2O/CH_3CN under air-free conditions were monitored with UV−vis and SF techniques at several wavelengths: 300 ($AcrL_2$), 358 ($AcrL^+$), and 640 ($AcrL_2^{•+}$) nm. In a typical experiment excess Fe_{aq}^{3+} (1 mM) was mixed with $AcrL_2$ (50 μM) in a solution with 0.1−0.5 M H^+ at $\mu = 1.0$ M. No absorbance change was noted at 640 nm, indicating that the rate of loss of $AcrL_2^{•+}$ exceeded its rate of formation. The data at 300 and 358 nm followed first-order kinetics, and the same value of k_{obs} was found for a given experiment at each wavelength.

The values of k_{obs} are directly proportional to $[Fe_{aq}^{3+}]$ and independent of $[H^+]$; the slope of this line represents k_1 (Scheme 1). No inhibition by Fe_{aq}^{2+}, which was added up to 1 mM, was observed. At a given ionic strength, the rate constants for $AcrH_2$ and $AcrD_2$ were the same, the values being $k_1 = 37.9 \pm 0.3$ L mol^{-1} s^{-1} ($\mu = 0.1$ M) and 128 ± 1 L mol^{-1} s^{-1} ($\mu = 1.0$ M).

Cyclic Voltammetry. Slow-scan CV experiments were performed. In CH_3CN an anodic wave was observed at 0.81 V versus SCE. The same experiment in H_2O/CH_3CN showed an anodic wave at 0.46 V versus SCE. In neither case was a cathodic wave observed at a scan rate of 3 V/s. These results are depicted in Figure 4.

Discussion

The apparent rate constants determined from the experimental data are summarized in Table 1.

Reactions with Ce(IV). The reaction proceeds by two single-electron pathways with an intervening acid−base step. This reaction network is represented in Chart 1. The first step in the case of Ce(IV) occurs within the mixing time of the SF apparatus; there, a 1e reaction converts dihydroacridine to a transient radical cation, $AcrH_2^{•+}$, that can itself be observed optically (see Figures S-1 and 4) and by EPR.[8] The radical cation is not further oxidized directly, but only after an acid ionization reaction produces the oxidizable radical $AcrH^•$. It proved possible to monitor the latter process by both the loss of $AcrH_2^{•+}$ and the buildup of $AcrH^+$. The data obtained from both methods agreed.

In the analysis of the data it was assumed that the *rate constant* for the reaction of Ce_{aq}^{4+} and $AcrH^•$, k_3, was at the diffusion-controlled limit, even though the actual *rate* of that reaction was competitive with the protonation reaction. The two steps compete because Ce_{aq}^{4+} represents a small proportion of the total cerium(IV) in the acid range studied, Ce_{aq}^{4+} and $CeOH^{3+}$ being quite acidic. The feature of Ce_{aq}^{4+} as the only

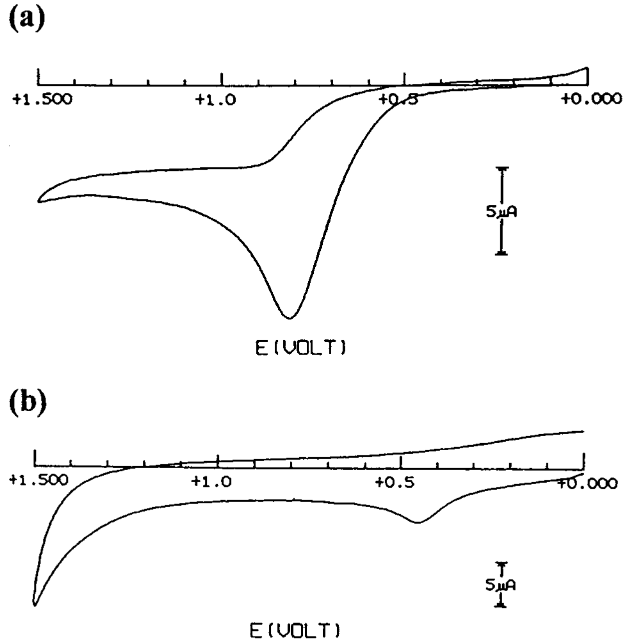

Figure 4. Cyclic voltammograms of $\mathrm{AcrH_2}$ in (a) $\mathrm{CH_3CN}$, and (b) 20% $\mathrm{CH_3CN}$/80% $\mathrm{H_2O}$ at 25 °C.

Table 1. Kinetic Parameters k_{i} and k_{p} Determined in Different Experiments for the Reactions of Ce(IV) and $\mathrm{AcrL_2}$ in $\mathrm{L_2O,L_3O^+/CH_3CN}$ at 25.0 °C

elementary reactions	$k_{\mathrm{i}}/\mathrm{s^{-1}}$	$k_{\mathrm{p}}/10^4\ \mathrm{L\ mol^{-1}\ s^{-1}}$
$\mathrm{AcrH_2^{\bullet+} + H_2O \rightleftarrows AcrH^{\bullet} + H_3O^+}$	691 ± 36	11.8 ± 1.1
$\mathrm{AcrD_2^{\bullet+} + D_2O \rightleftarrows AcrD^{\bullet} + D_3O^+}$	106 ± 6	3.41 ± 0.58
$\mathrm{AcrHD^{\bullet+} + D_2O \rightleftarrows AcrH^{\bullet} + D_3O^+}$	53,[a] 46[b]	(6.34,[c] 5.99[b,c])
$\mathrm{AcrHD^{\bullet+} + H_2O \rightleftarrows AcrD^{\bullet} + H_3O^+}$	345,[a] 402[b]	6.34,[d] 6.72[b,d]

[a] Statistical estimate from line 1 or 2. [b] Computer fitting, allowing for a secondary kie on k_{i} or k_{p}. [c] Calculated value; see Discussion. [d] Estimated error: ±10−15%.

reactant was the intended implication of the notation used in eq 6. As for k_3 itself, we note the large driving force (estimated ΔG° for the third reaction shown in Scheme 1 is ~ -200 kJ $\mathrm{mol^{-1}}$) and the lack of significant reorganization energy between either of the redox partners, $\mathrm{Ce_{aq}^{4+}/Ce^{3+}}$ and $\mathrm{AcrH^{\bullet}/AcrH^+}$. These factors stand in evidence as to the correctness of this assignment. Indeed, the reorganization requirement is just the reason we would advance to explain our finding that neither $\mathrm{CeOH^{3+}}$ nor $\mathrm{Ce(OH)_2^{2+}}$ reacts with $\mathrm{AcrH^{\bullet}}$, nor does $\mathrm{Ce_{aq}^{4+}}$ react with $\mathrm{AcrH_2^{\bullet+}}$.

The analysis of the kinetic data provided no support for the pathway referred to as radical disproportionation, which was reported to be one reaction that occurs in acetonitrile.[8] This step is given by

$$2\mathrm{AcrH_2}^{\bullet+} \xrightarrow{k_{\mathrm{disp}}} \mathrm{AcrH}^+ + \mathrm{AcrH_2} + \mathrm{H}^+ \tag{9}$$

Given the molecular structures involved Chart 1, this equation represents an unlikely elementary reaction. We suggest instead that any second-order pathway is more likely to be electron-transfer; eq 10 appears to be a reasonable alternative.[17] As it

$$\mathrm{Acr}^{\bullet} + \mathrm{AcrH_2}^{\bullet+} \xrightarrow{k_{\mathrm{et}}} \mathrm{AcrH}^+ + \mathrm{AcrH_2} \tag{10}$$

turns out, a second-order decay pathway was not found under the conditions used here. Kinetic simulations with the KinSim program have shown that a pathway second-order with respect to $[\mathrm{AcrH_2^{\bullet+}}]$ cannot contribute to the loss of the radical cation to any significant degree.

Acid Ionization. The value $\mathrm{p}K_{\mathrm{a}} = 2.23$ in the nearly aqueous medium, although close to the first reported value of 2.0 in water itself,[13] is lower than the values reported previously when only a small amount of water was present in acetonitrile: $\mathrm{p}K_{\mathrm{a}} = 8.1$ at 0.03 M $\mathrm{H_2O}$ and $\mathrm{p}K_{\mathrm{a}} = 6.8$ at 0.30 M $\mathrm{H_2O}$, the extremes of the range studied.[8] The linear plot of $\mathrm{p}K_{\mathrm{a}}$ versus log $[\mathrm{H_2O}]$, if extrapolated to 80% water, allows one to estimate a $\mathrm{p}K_{\mathrm{a}}$ of 4.0. Considering the extrapolation involved, the agreement is not unreasonable. The reported effect of water at these low levels is to accelerate k_{i} to a small extent and retard k_{p} strongly. In the present case, k_{i} for $\mathrm{AcrH_2^{\bullet+}}$ rises to 690 $\mathrm{s^{-1}}$ (from 6.4 to 11.5 $\mathrm{s^{-1}}$ at 0.03−0.30 M $\mathrm{H_2O}$) and k_{p} falls to 2.6×10^5 L $\mathrm{mol^{-1}\ s^{-1}}$ (from 8.1×10^8 to 0.7×10^8 L $\mathrm{mol^{-1}\ s^{-1}}$).[8] Both rate constants change significantly, and in opposite directions, as one moves to the medium with 80% water. Considering the $\mathrm{p}K_{\mathrm{a}}$ difference between aqueous and acetonitrile media,[8] the $\mathrm{p}K_{\mathrm{a}}$ value of 0.6 in acetonitrile[17] may be inaccurate, as it would require a negative $\mathrm{p}K_{\mathrm{a}}$ in water.

Kinetic Isotope Effects. The kie's for k_{i} and k_{p} are appreciable. When $\mathrm{AcrL_2}$ was studied in $\mathrm{L_2O}$, the effects could be studied quantitatively. They are represented as follows:

$$\left(\frac{{}^{\mathrm{H}}k_{\mathrm{i}}^{\mathrm{H}}}{{}^{\mathrm{D}}k_{\mathrm{i}}^{\mathrm{D}}}\right) = \frac{691}{106} = 6.52 \pm 0.50$$

$$\left(\frac{{}^{\mathrm{H}}k_{\mathrm{p}}^{\mathrm{H}}}{{}^{\mathrm{D}}k_{\mathrm{p}}^{\mathrm{D}}}\right) = \frac{11.8 \times 10^4}{3.41 \times 10^4} = 3.45 \pm 0.67$$

A second L (H or D), the one not being transferred, is present at position 9. Thus one must consider the possibility of a secondary kie in the analysis of the findings, in which case the above values are composites of secondary and solvent effects. To explore this further, experiments were performed starting with $\mathrm{AcrD_2}$ in $\mathrm{H_2O/CH_3CN}$ with $[\mathrm{H_3O^+}] = 0.300$ M. This situation is more complex than those examined before, owing to the possibility that D and H are interchanged during the course of the reaction. The full set of reactions is displayed in Scheme 2, with a step of negligible importance shown with a dashed arrow. In this notation the rate constants are labeled with the fixed L as a left superscript and the entering L as a right superscript.

We first assumed, neglecting for now any secondary isotope effect on k_{i} (see later), that ${}^{\mathrm{H}}k_{\mathrm{i}}^{\mathrm{D}}$ for $\mathrm{AcrHD^{\bullet+}}$ was the same as the statistically corrected value of ${}^{\mathrm{D}}k_{\mathrm{i}}^{\mathrm{D}}$, or 53 $\mathrm{s^{-1}}$; likewise, we took ${}^{\mathrm{D}}k_{\mathrm{i}}^{\mathrm{H}}$ for $\mathrm{AcrHD^{\bullet+}}$ as 345 $\mathrm{s^{-1}}$. With these and the known rate constants, only one parameter of importance remained, the protonation constant for $\mathrm{AcrD^{\bullet}}$, ${}^{\mathrm{D}}k_{\mathrm{p}}^{\mathrm{H}}$. The data from the family of experiments with varying [Ce(IV)] and fixed $[\mathrm{H_3O^+}] = 0.300$ M were fitted as a group by the FitSim routine. This gave ${}^{\mathrm{D}}k_{\mathrm{p}}^{\mathrm{H}} = (6.34 \pm 0.04) \times 10^4$ L $\mathrm{mol^{-1}\ s^{-1}}$. The agreement between the fit and the experiments is excellent; Figure S-2 (Supporting Information) displays one such trace, showing the buildup of $\mathrm{AcrL^+}$ and the match between experiment and model.

The Case of $\mathrm{AcrH_2}$ in $\mathrm{D_2O/D_3O^+}$. This situation, the fourth and final combination, should be analogous to that in the preceding section. The difficulty is that the time scale is now very short, being governed in large part by $k_{\mathrm{i}}^{\mathrm{H}}$, and data of useful precision could not be obtained.

Analysis of the Kinetic Isotope Effect. On the basis of these determinations, further isotope effects can be calculated. One

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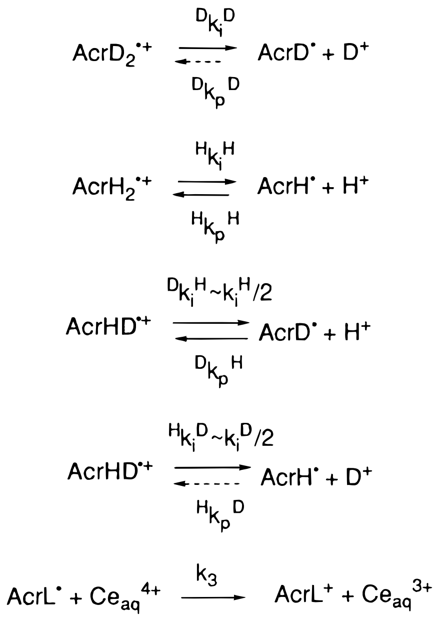
Scheme 2



is the secondary kinetic isotope effect for the L-group remaining. The value is

$$\left(\frac{^Hk_p{}^H}{^Dk_p{}^D}\right) = \frac{11.8 \times 10^4}{6.34 \times 10^4} = 1.86 \pm 0.17$$

For certain practical reasons, it was not feasible to determine accurately the alternative formulation, $^Hk_p{}^H/^Dk_p{}^D$, a second measure of the secondary isotope effect, although logic requires the same value, a point to which we shall return.

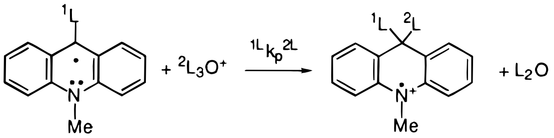
The solvent isotope effect (i.e., the effect of the incoming proton/deuteron) is given by

$$\left(\frac{^Hk_p{}^H}{^Dk_p{}^D}\right) = \frac{6.34 \times 10^4}{3.41 \times 10^4} = 1.86 \pm 0.32$$

where again the alternative measure of the same effect is not readily available. The coincidence of these numbers is fortuitous, but other relations between the component rate constants are required. The second kie is also equal to $^Hk_p{}^H/^Hk_p{}^D$, and the only value that will satisfy all requirements is $^Hk_p{}^D = 6.34 \times 10^4$ L mol^{-1} s^{-1}; it necessarily also satisfies the requirement given in the preceding paragraph. Again, the coincidence with $^Dk_p{}^H$ is largely fortuitous.

A series of FitSim simulations was then performed to assess the extent to which the secondary kie for k_i, or its lack, affects the secondary kie for k_p. Several values of the secondary kie, $^Hk_i{}^H/^Dk_i{}^H$, were assumed, in the range 1.0−1.2, this being suggested by other work,[18−23] and then used to calculate $^Hk_i{}^D$ and $^Dk_i{}^H$. The corresponding value of ($^Hk_p{}^H/^Dk_p{}^H$) for each k_i pair was then obtained by the previously described fitting procedures. The results show that $^Hk_p{}^H/^Dk_p{}^H$ does not change by more than 7% and retains its sign. The error analysis also shows that the secondary isotope effect on k_i is either small or nonexistent. This finding is somewhat surprising, in that small but appreciable α-D secondary isotope effects have been seen in other reactions. A typical effect is the value 1.145 ± 0.009 for hydride abstraction from NADH by 4-cyano-2,6-dinitrobenzene sulfonate.[20] Unusually large α-D secondary isotope effects were found for the reduction of acetone, 1.38, and cyclohexanone, 1.50, by various alcohol dehydrogenases.[24]

In the system at hand, the secondary kie measures the effect of the nontransferred ^1L of the dihydroacridine ring system:

 (11)

Conventional transition-state theory predicts a small inverse kie for the conversion of an sp^2-hybridized C−L bond to an sp^3-like transition state in such reactions.[22,25,26] Hydride transfer to the acridinium cation produces a small normal isotope effect whose magnitude depends on whether H^- or D^- is transferred.[19,21] Nuclear tunneling along the reaction coordinate has been proposed to explain such unusual kinetic behavior in this and several other systems.[21,23] The normal secondary isotope effect on k_i in this work also suggests a possibility of tunneling in the protonation of AcrH$^\bullet$.

A presumption was made earlier that k_i shows no secondary kie; on that basis, we then took $^Hk_i{}^D = 53$ s^{-1} and $^Dk_i{}^H = 345$ s^{-1}. Further calculations and simulations were carried out in which the ratio ($^Hk_i{}^H/^Dk_i{}^H$), which measures this effect, was varied in the range 1.3−0.7. At each point in the calculation, $^Dk_p{}^H$ was obtained from the experimental data. The optimum fit occurred at these values of the secondary isotope effect: $(^Hk_i{}^H/^Dk_i{}^H)^{sec} = 0.86$ and $(^Hk_p{}^H/^Dk_p{}^H)^{sec} = 1.76$. The inverse kie on k_i suggests that the protio form has a contribution from tunneling. Moreover, the normal kie for k_p implicates tunneling in the reverse direction as well.

Reaction with Hexachloroiridate(IV). Again the driving force for the first step of Scheme 1, electron transfer between AcrL$_2$ and $IrCl_6^{2-}$, is only ~0.2 V. Still, the combination of it with the large electron-exchange rate constant for $IrCl_6^{2-}/IrCl_6^{3-}$ [27] was sufficient to make the initial electron-transfer step fast, and only subsequent steps were kinetically important. Since the concentration of the hexachloroiridate ion was not reduced by participation in hydrolytic equilibria, a low concentration of $IrCl_6^{2-}$ suffices to attain the plateau. With AcrD$_2$, $k_i{}^D = 115 \pm 2$ s^{-1}, in close agreement with the value 106 ± 6 s^{-1} for determinations with cerium(IV). Attempts with AcrH$_2$ gave reactions too rapid for accurate determinations (recall here that the actual value of the plateau is measured directly, not by extrapolation, as for cerium(IV)); with micromolar levels of $IrCl_6^{2-}$, adjusted to bring the k_p and k_3 steps into competition, it was possible to observe the expected acid retardation.

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Reaction with Hexaaquairon(III). All three steps in Chart 1 apply, but the first is now rate-controlling and the others are very rapid in comparison. Thus the rate law is simply d[AcrL$^+$]/dt = k_1[AcrL$_2$][Fe(H$_2$O)$_6^{3+}$]. Consistent with that, the calculated value of k_{-1}, from k_1/K_1, is ~9 L mol^{-1} s^{-1} at $\mu = 1.0$ M. In this estimate, K_1 was obtained from $\Delta E°$ under the assumption that in 80% H_2O/CH_3CN the irreversible potential determined by CV is close to the standard potential,[28] which seems to hold for this couple in acetonitrile. The value of k_{-1} makes it clear that both steps following the first are indeed quite rapid relative to it, which means that the first step can properly be assigned as rate-controlling.

The rate constant k_1 remained independent of [H$^+$], 0.1−0.5 M, at constant ionic strength. Therefore Fe(H$_2$O)$_6^{3+}$, not (H$_2$O)$_5$FeOH^{2+}, is the reactant. The clear implication of this is that the k_1 step occurs by outer-sphere electron transfer. We thus eliminate an inner-sphere mechanism, since the literature precedents suggest that the hydroxo complex would then have been far more reactive, and a pH effect manifest. Further support for the assignment of electron transfer as the mechanism of the k_1 step comes from the lack of any kie, as AcrH$_2$ and AcrD$_2$ gave the same value of this rate constant in the Fe(H$_2$O)$_6^{3+}$ reaction.

General Mechanism for the One-Electron Oxidation of Dihydroacridine. The scheme presented earlier, Scheme 2, appears uniformly valid. Strong oxidizing agents such as Ce$_{aq}^{4+}$ and IrCl$_6^{2-}$ that have high electron-exchange rates,[27,29] render the first electron-transfer step irreversible and too rapid for observation. In such cases, therefore, one can study the acid−base reactions of the radical species free of metal ion interference. With Fe(H$_2$O)$_6^{3+}$, a reagent of mild oxidizing strength and low electron-exchange rate,[30,31] the reverse is true: outer-sphere electron transfer in the first step is rate-controlling.

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Supporting Information Available: Two figures showing stopped-flow kinetic data for the reaction between AcrH$_2$ and Ce$_{aq}^{4+}$ and a comparison of the experimental data and the FitSim calculation for one experiment with AcrD$_2$ and Ce(IV) in H$_2$O and H$_3$O$^+$ are presented (2 pages). Ordering information is given on any current masthead page.

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(28) We have assumed that the irreversible potential determined by CV is close to the $E°$. This assumption seems to be valid for the oxidation of AcrH$_2$ in pure acetonitrile (see ref 8).

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