Competition between Dipolar Relaxation and Double Proton Transfer in the Electronic Spectroscopy of Pyrroloquinolines†

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Received: October 14, 1998; In Final Form: December 15, 1998

The molecule 1-*H*-pyrrolo[3,2-*h*]quinoline (1-HPQ) (alternatively pyrido[3,2-*g*]indole) and its 1-methyl derivative (1-MPQ) are shown to exhibit a pronounced dipolar-relaxation red shift of their principal fluorescence in protic solvents. For 1-HPQ, the red shift is $\Delta\nu = 2232$ cm^{-1}, comparing methanol with methylcyclohexane solutions (λ_{max} 363−395 nm for 1-HPQ, 298 K). For 1-HPQ a second, weaker long-wavelength fluorescence is observed at λ_{max} 565 nm in methanol at 298 K, a shift of 7617 cm^{-1} from the fluorescence band maximum of the normal tautomer (at 395 nm); this is shown to be a proton-transfer tautomer fluorescence. The dual fluorescence of 1-HPQ in protic solvents is evidently the result of an unusual competition between the competing pathways of dipolar relaxation and excited state proton transfer. Two models are discussed for the excitation dynamics, leading to the conclusion that two populations of solvated 1-HPQ molecules exist in protic solvents, one promoting excited state double proton transfer catalyzed by a solvent bridge, and the other permitting primarily dipolar relaxation.

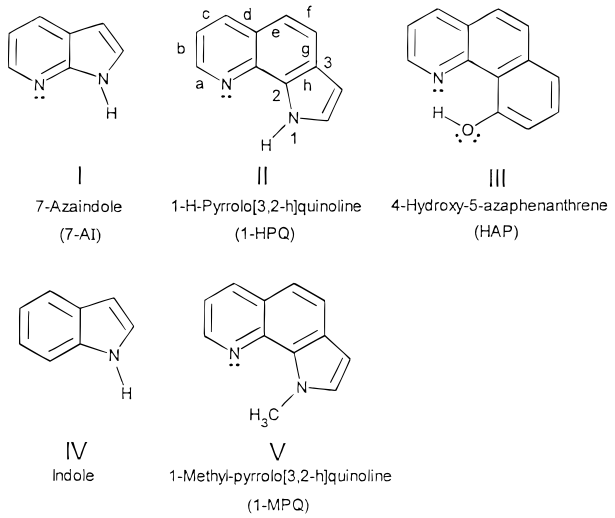
1. Introduction: Excitation Competition Mechanisms in Heteroaromatics

Current research in this laboratory on a pyrroloquinoline molecule has revealed interesting variations in the intricate mechanisms of competing excitation pathways involving solvent dipolar relaxation upon solute molecule excitation and proton-transfer tautomerization. The pyrroloquinoline studied, 1-HPQ (II) (1-*H*-pyrrolo[3,2-*h*]quinoline) (alternatively, pyrido[3,2-*g*]-indole) can be considered to be an electronic structural and behavioral intermediate between the well-studied molecule 7-azaindole[1−3] (7-AI) (I) and the recently studied 4-hydroxy-5-azaphenanthrene[4−6] (HAP) (III) (Chart 1).

The *7-azaindole* molecule (I) features several defining spectral characteristics. The lowest near-UV absorption band yields a strong violet fluorescent band in dilute solution in hydrocarbon solvents, which in protic solvents becomes strongly red-shifted (4300 and 5770 cm^{-1} in 2-propanol and water, respectively) and broadened.[1,7] This parallels the behavior of the precursor molecule indole (IV), for which the large red shift in protic solvents has been attributed to a solvent−solute exciplex formation.[8,9] In contrast, more recent studies have adduced the solvent dipolar-relaxation effect[10,11] as the origin of the protic solvent red shift of the fluorescence band of indole. A second defining spectral behavior of 7-AI is its exhibiting an *excited state double proton transfer* (ESDPT) as the mechanism of a phototautomerization, yielding a green-fluorescing transient tautomer species.[1] This tautomerization cannot occur in the isolated single molecule, as the pyrrolic proton does not overlap with the n(sp^2) lone pair orbital of the aza-N; this lone pair orbital is a coplanar σ-type orbital and is axially directed away from the pyrido ring center. For an efficient excited-state proton transfer, electronic and other characteristics being favorable, a preformed H-bonded cyclic structure would be necessary for possible proton transfer (PT) to be observed. Consequently, it has been shown[1] that ESDPT can be observed in 7-AI upon dimerization in more concentrated (10^{-3} M) hydrocarbon solution, or it can occur in protic solvents such as alcohols, which may form an H-bonded bridge[1] (from N−H to the aza-N lone pair). These are considered to be catalyzed biprotonic phototautomerizations. Petrich et al.[12,13] have adopted our model[1,3] for differential proton transfer in various protic solvents. Their introduction of the related azatryptophan has led to the

CHART 1

STRUCTURES AND NOMENCLATURE OF SOME N-HETEROCYCLICS



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† This paper is dedicated to the memory of Professor Bryan E. Kohler whose perception of the fundamental significance of an apparent energy gap in the spectra of polyenes led to his remarkable development of the subject up to the visual pigments.

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10.1021/jp9840679 CCC: $18.00
Published on Web 02/11/1999

development of a powerful luminescence probe for protein studies. In the case of water as a solvent the PT in 7-AI cannot be observed, unless very dilute water (e.g., in dioxane) is used, with the purpose of breaking up the long H-bonded water chains so that single or double water molecules can form an H-bonded bridge between the aza-N and the pyrrolo-H atom. Thus, a concerted double proton transfer in the excited state is facilitated in this fashion.[3] Chapman and Maroncelli[14] believe they have detected a proton transfer for 7-AI in liquid water by an anomalous mean decay time observed in the long-wavelength tail of the normal tautomer fluorescence.

The *4-hydroxy-5-azaphenanthrene* (III) stands in sharp contrast to 7-AI regarding *excited-state intramolecular proton transfer* (ESIPT). In this case the six-membered ring H-bond is so strong between the protic hydroxy-H atom and the aza-N atom that water as solvent does not interfere. Thus proton transfer (PT) is observable even in liquid water as the solvent.[4−6] This is an unusual case, as in other circumstances water molecules can offer strong interference to ESIPT. For example, in the much weaker five-membered ring H-bond in the 3-hydroxyflavonols,[15] the proton-transfer tautomerization is completely blocked in liquid water. In both 7-AI and HAP the characteristic PT fluorescence (room-temperature solution) is observed in the green region of the spectrum, representing the typical large PT shift of 7000 to 10 000 cm^{-1} measured from the normal molecular absorption onset of each species. In HAP no normal tautomer fluorescence is observed as a consequence of the very efficient conversion to the PT tautomer in the excited state.[4−6]

The 1-*H-pyrrolo[*3,2-*h]quinoline* molecule (II) can be considered as the 7-AI modified by a benzo ring spacer, separating the pyrido ring from the pyrrolo ring. It is obvious that the geometry of this 1-HPQ molecule does not admit the possibility of an internal H-bond from the pyrrolo-H to the aza-N atom. Thus, we anticipate that as in the case of 7-AI, a protic-bridge catalysis will be involved if ESDPT can be observed, and as in the case of 7-AI, if H_2O is to be a bridging molecule, it would have to be strongly diluted by an inert or facilitating solvent to permit one or two H_2O molecule H-bonded bridges. In such a case, involving one- or two-molecule alcohol bridges, a diffusion-controlled mechanism will arise to permit a double-H-bond bridge to form. The reason for this is the necessity of solvent rearrangement[15] to form H-bonded bridges (between the −N−H proton donor and the aza-N proton acceptor) in place of the separate H-bonded chains that would exist commonly in the protic solvation of the hetero groups. A more complex solvation mechanism involving solvent rearrangement around the intramolecular H-bonding and PT-loci in a solute molecule was introduced by Woolfe and Thistlethwaite.[16] They suggested that the growth lifetime (69 ± 8 ps) for the excited-state tautomer of 3-hydroxyflavone (3-HF) originated in methanol solvent *rearrangement* to a closed H-bonded cyclic structure facilitating (double) proton-transfer involving 3-HF and methanol. A related observation was made by Moog and Maroncelli on 7-azaindole in alcohol solutions,[17] who presented a definitive study of the dynamics of proton transfer in 7-AI for a long series of aliphatic alcohols.

Such a diffusion-controlled process should be subject to the viscosity-dependent potential barrier visualized in the Dellinger−Kasha solvent cage model.[18−20] Parallel to the 7-AI case, if the excited-state dipole moment vector is rotated within the molecular framework relative to the ground-state dipole moment vector, a large dipolar solvent relaxation could also be observable. This could occur for both the normal fluorescence and the proton-transfer fluorescence. The 1-HPQ molecule, because of the delicate catalytic origin of its proton-transfer fluorescence, would offer the opportunity of separating the dipolar (protic) solvent cage relaxation from the protic-catalyzed PT fluorescence. In addition, the methylated 1-MPQ molecule (V) offers the opportunity to exclude ESDPT as a test of the assignment of such PT fluorescence, if observable, in 1-HPQ.

In this paper we shall investigate the normal absorption and fluorescence spectra of 1-HPQ at 298 K and at 77 K in rigid glass solvents, the effect of protic solvent dipolar relaxation on the fluorescence spectrum, the phosphorescence spectrum, and the possibility of a catalytically induced excited state double proton transfer. Our purpose is to elucidate the photophysical and photochemical behavior of the 1-*H*-pyrrolo[3,2-*h*]quinoline molecule. Spectroscopic models will be presented to illustrate alternate competitive pathways of the excitation mechanisms. In addition, we shall review the various chemical and biomedical applications now being made with 1-HPQ.

2. Experimental Section

Absorption Measurements. Absorption spectra were recorded on a Shimadzu UV-2100 spectrophotometer using quartz cuvettes of 1 cm path length, or as mentioned otherwise in the text and figure captions. Both 1-HPQ and 1-MPQ show an excellent Lambert−Beer behavior for the concentrations assayed in this paper (1.2×10^{-5} to 1.2×10^{-3} M), and therefore there was no evidence of aggregation effects in the solvents used.

Fluorescence and Phosphorescence Measurements. Fluorescence spectra and lifetimes were obtained from a phase-modulation Fluorolog-2 lifetime spectrofluorometer (SPEX). The lifetimes were obtained relative to glycogen scattering solutions.[21] Decisions on the validity of the lifetimes rested on examination of the statistics of a fit (a plot of the residual deviations with frequency) and the reduced χ^2 values. The reduced χ^2 values obtained in our experiments were all close to unity. All of the solutions were stirred during the lifetime measurements. Low-temperature emission studies were carried out by employing a liquid nitrogen accessory to cool the samples to temperatures approaching 77 K. Phosphorescence spectra were recorded using a SPEX FluoroMax spectrophotometer. All the solvents used were spectrophotometric grade and were used as supplied.

General Methods. Melting points were measured in a Thomas-Hoover capillary-melting point apparatus and are uncorrected. Nuclear magnetic resonance spectra were obtained on a Varian Gemini 300 MHz NMR spectrometer at standard conditions. Chemical shifts and coupling constants were measured in deuteriochloroform referred to TMS (tetramethylsilane) as the internal standard.

Synthesis. The molecule 1-*H*-pyrrolo[3,2-*h*]quinoline (II) (1-HPQ) was readily prepared in 50% overall yield from 8-aminoquinoline by a straightforward three-step sequence similar to the procedure previously reported by Sergeeva et al.[22] The melting point was determined to be 99−101 °C.

The molecule 1-methyl-pyrrolo[3,2-*h*]quinoline (V) (1-MPQ) was prepared in 92% yield (after column chromatography) from 1-*H*-pyrrolo[3,2-*h*]quinoline, under mild reaction conditions, short reaction time, and easy workup. The procedure used was reported previously by Kikugawa et al.[23] for the synthesis of *N*-alkylindoles. The melting point was determined to be 58−60 °C.

Nuclear Magnetic Resonance Data. The purity of both compounds was checked by NMR and melting point measurements and was determined to be higher than 99%.

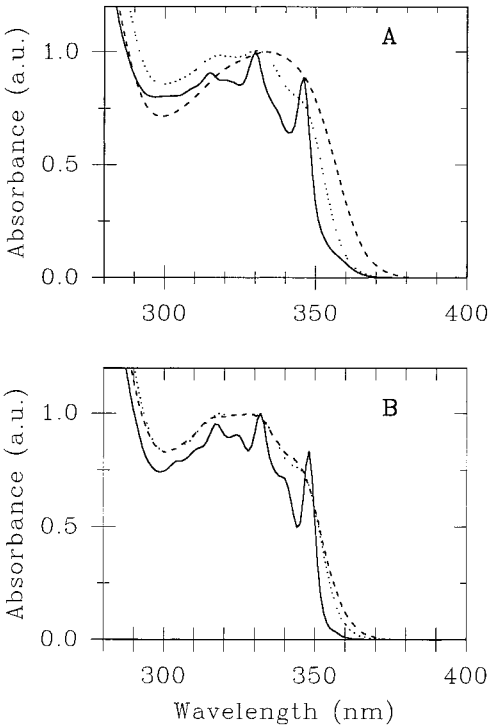


Figure 1. Near-ultraviolet absorption spectra of 1-HPQ (II) (A) and 1-MPQ (V) B) in *n*-pentane (solid line), dioxane (dot line), and methanol solutions (dash line) at 298 K.

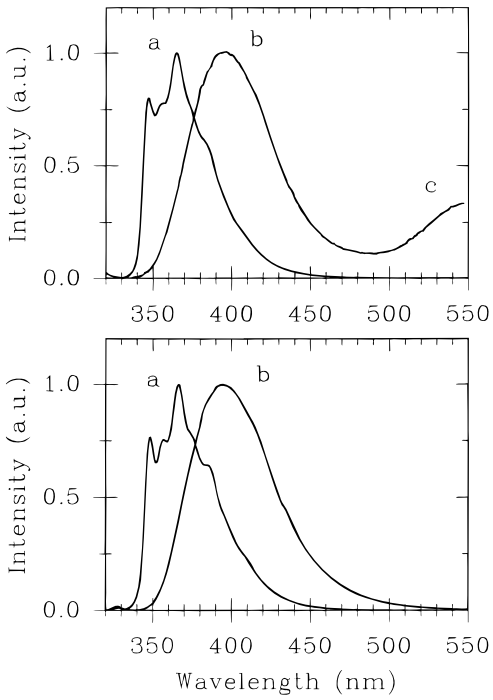


Figure 2. Dipolar-relaxation shift of the fluorescence spectra of 1-HPQ (II) (upper) and 1-MPQ (V) (lower) at 298 K. Curves a and b represent the fluorescence spectra of the normal tautomer in methylcyclohexane solution and methanol solution, respectively. Curve c represents the onset of proton-transfer fluorescence of 1-HPQ in methanol solution (cf. Figures 4 and 5).

Some selected ^{1}H ($CDCl_3$) NMR spectroscopic data for 1-HPQ are 12.47 (br s, 1H, NH), 8.96 (dd, 1H, J = 4.2 and 1.8, H-8), 8.30 (dd, 1H, J = 8.4 and 1.8, H-6), 7.90 (d, 1H, J = 8.7, H-5), 7.52 (d, 1H, J = 8.7, H-4), 7.44 (dd, 1H, J = 8.4 and 4.2, H-7), 7.43 (dd, 1H, J = 2.7 and 2.7, H-2), and 6.80 (dd, 1H, J = 2.4 and 2.4, H-3). Chemical shifts are given in ppm, and coupling constants, J, in Hz (s = singlet and d = doublet).

Some selected ^{1}H ($CDCl_3$) NMR spectroscopic data for 1-MPQ are 8.84 (dd, 1H, J = 4.5 and 1.8, H-8), 8.18 (dd, 1H, J = 8.1 and 1.8, H-6), 7.74 (d, 1H, J = 9.0, H-5), 7.40 (d, 1H, J = 8.14, H-4), 7.33 (dd, 1-H, J = 8.1 and 4.5, H-7), 7.16 (d, 1H, J = 2.7, H-2), 6.63 (d, 1H, J = 2.7, H-3), 4.53 (s, 3H, CH_3−N).

Theoretical Calculations.[24] The ground-state dipole moment was calculated by B3LYP Hybrid Density Functional Theory[25,26] in conjunction with a 6-31G** basis set (with full optimization of the geometry). The first excited-state dipole moment has been calculated at the CIS/6-31G** level[27] using the B3LYP/6-31G** optimized ground-state geometry. The calculations have been undertaken with the Gaussian 94 program.[28]

3. Assignment and Solvent Perturbations of the First Absorption Band of the Pyrroloquinolines

In Figure 1A,B we display the absorption spectra[29] of 1-*H*-pyrrolo[3,2-*h*]quinoline (II) (1-HPQ) and 1-methyl-pyrrolo[3,2-*h*]quinoline (V) (1-MPQ) in different solvents (*n*-pentane, dioxane, and methanol). For both 1-HPQ and 1-MPQ, there is a loss of observable vibronic structure upon increasing the polarity of the medium, and a small red shift is observed in the absorption maxima. It is noticeable in Figure 1A,B for 1-HPQ especially that the long-wavelength tail of the absorption band is broader in the protic solvent (methanol), in addition to an average shift of the center of gravity of the band to the red. The spectra for 1-MPQ are significantly less perturbed. These observations should be understandable in terms of the nature of the electronic transitions involved.

The long-wavelength absorption band of 1-HPQ (and 1-MPQ) in the range 300−350 nm is certainly assignable to a $\pi^* \leftarrow \pi$ transition, with a molar absorption coefficient of 3800 M^{-1} cm^{-1} at 330 nm.

The 1-HPQ molecule exhibits strong H-bonding effects[30−33] and is extremely sensitive to the presence of water. The water molecule acts as a powerful perturbing agent on the spectroscopy of 1-HPQ, acting as a solvent yielding the largest spectral perturbations. Comparing the first absorption band for 1-HPQ in dioxane, in very dilute H_2O in dioxane, and in liquid H_2O as solvent with the analogous spectra for 1-MPQ, the methylated molecule is observed to be less sensitive to H-bonding perturbation. The absorption spectra of the water solutions of 1-HPQ and 1-MPQ show additional absorption in the region 370−450 nm attributable to the formation of ionic species of 1-HPQ and 1-MPQ. The 1-HPQ molecule is so sensitive to water that in a cyclohexane dried over freshly extruded sodium threads a slight spectral broadening was still apparent owing to minute traces of water present in the solution. The special H-bond complexing avidity of 1-HPQ appears to be involved in its biomedical applications, as summarized in section 8.

4. Solvent Cage Dipole Relaxation Effects on the Fluorescence Spectra of Pyrroloquinolines

The pyrroloquinolines 1-HPQ (II) and 1-MPQ (V) reveal striking wavelength shifts in their fluorescence spectra in polar solvents vs nonpolar solvents. Figure 2 (upper) shows the methylcyclohexane solution spectrum, band a of 1-HPQ, and the methanol solution spectrum, bands b and c. The shift (a → b) of the F−C maximum representing the $^1L_b \rightarrow {}^1A$ transition[34] (363 to 395) equals 2232 cm^{-1}. For 1-MPQ the shift (a → b) (366 to 394 nm) equals 1940 cm^{-1}. (The unique band c for

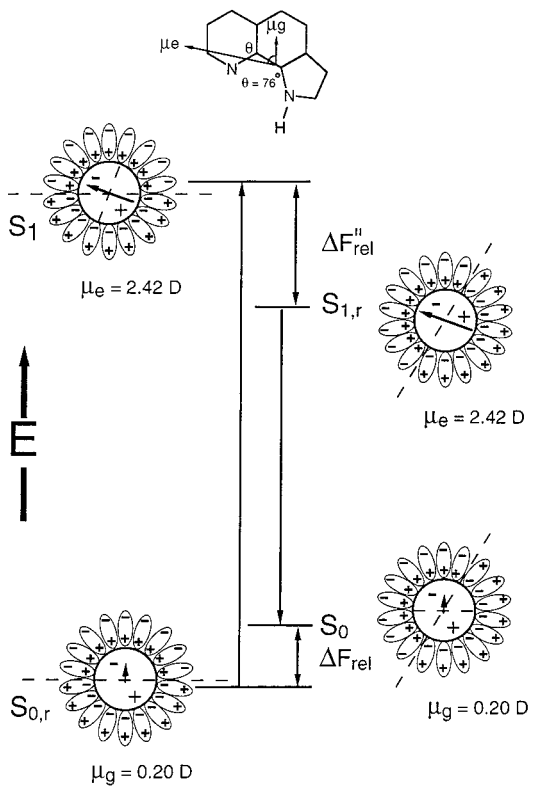


Figure 3. Schematic depiction of the dipolar-relaxation mechanism. Vector in inner circle represents the permanent dipole of the solute. Outside the solute contour is a representation of the array of solvent dipoles. $S_{0,r}$ and $S_{1,r}$ represent the equilibrium relaxed solute/solvent array, S_0 and S_1 represent the nonequilibrium array. The dashed lines are used as guidelines so that the reorientation of the dipole solvent cage can be understood.

1-HPQ will be discussed in section 5 as the double PT fluorescence.) These are substantial shifts in the dipolar solvents and are clearly the result of solvent cage dipolar relaxation caused by changes in orientation of the internal dipole moment of the solute molecule upon $^1L_b \leftarrow {}^1A$ excitation.

Solvent cage dielectric relaxation has been surveyed[35] qualitatively as representing (a) the electronic polarization solvent cage, yielding instantaneous response under solute electronic state excitation, (b) the random dipole solvent cage, immobilizable by freezing to the glass state, and (c) the ordered dipole solvent cage, exhibiting reequilibration to a new electronic state configuration, with a solvent dipole relaxation time delay. Case c applies to a dipolar solvent in a liquid state and represents the effect observed for 1-HPQ and 1-MPQ in methanol solvent at 298 K (Figure 2). The schematic diagram given in Figure 3 illustrates a solute molecule dipole that changes orientation upon, e.g., S_0 to S_1 electronic excitation, surrounded by the first shell of solvent cage dipoles. The dipolar field orientation of the medium cannot change instantly during the ultrarapid electronic excitation process and equilibrates during the dipolar relaxation time to the equilibrated excited-state $S_{1,r}$. Upon fluorescence emission, the dipolar solvent cage field must again equilibrate to the S_0 solute dipolar intramolecular orientation, yielding a second solvent dipolar relaxation. The nonequilibrium free energies of solvent dipolar relaxation are $\Delta F_{rel}''$ and ΔF_{rel} respectively; these values appear in the mechanism pathways diagrams in section 7. The dipole moment calculations for 1-HPQ (cf. Experimental Section and Figure 3) show that the dipole moment magnitude for the ground electronic state (μ_g) is 0.20 D, whereas for the first singlet excited state the dipole moment (μ_e) magnitude has a value of 2.42 D. Besides that great dipole moment change in magnitude, the dipole moment reorientation is even more relevant, it being a 76° dipole moment reorientation change for the $S_o \rightarrow S_1$ electronic transition. In principle, the dipolar-relaxation spectral shifts can be clocked by fast-time transient spectroscopy. There are notable contrasts between the dipolar solvent cage effects observed for 1-HPQ (II) and 1-MPQ (V) and those observed for indole (IV) and 7-azaindole (I) (Chart 1). First, we note that the former pair exhibits dipolar solvent shifts of ca. 2000 cm^{-1}, whereas the latter pair shows much larger shifts between 4000 and 6000 cm^{-1}. Second, in the case of the indoles a large quenching of the fluorescence is observed accompanying the dipolar solvent shift effect, attributed to electron-transfer quenching in addition to solvent−solute exciplex formation.[7−9] Such a direct quenching effect is absent for 1-MPQ. In fact, by adding traces of water to a dioxane solution of 1-MPQ, there is a small increase in the absolute fluorescence intensity vs that for the pure dioxane solution of 1-MPQ. We conclude that the dipolar solvent effect on the fluorescence spectrum of 1-MPQ is a pure solvent cage dipolar relaxation effect, devoid of any exciplex or electron-transfer quenching.

TABLE 1: Luminescence Data[a] for Pyrroloquinolines (Pyridoindoles)

	solvent					
	cyclohexane			methanol		
species	φ_F[b]	τ_F (ns)	$10^{-7}k_r$ (s^{-1})	φ_F[b]	τ_F (ns)	$10^{-7}k_r$ (s^{-1})
1-HPQ (II)	0.16	4.22	3.8	0.0004	0.13	3.1×10^6 [c]
1-MPQ (V)	0.25	4.58	5.5	0.16	5.53	2.9×10^7

[a] These data are for the normal tautomer fluorescence $S_1 \rightarrow S_0$. The data for the PT tautomer fluorescence $S_1' \rightarrow S_0'$ for 1-HPQ in methanol are $\tau_{F'} = 0.26$ ns and $\varphi_{F'} < 0.0003$. All data for 298 K. [b] Method of Meech and Phillips,[36] using 2-aminopyridine in aqueous sulfuric acid 0.05 N as standard, with $\varphi_F = 0.66$, corrected for refractive indices of the solvents used. [c] A 10-fold decrease of the radiative rate of 1-HPQ (verified by independent results from two different instruments) is explained because methanol is hydrogen-bonded to 1-HPQ, thereby implying different solute solvation, and thus, a perturbed excited electronic state compared with that in the case 1-HPQ in cyclohexane.

The fluorescence quantum yields (Table 1) for 1-MPQ in cyclohexane and methanol are 0.25 and 0.16, respectively. However, for 1-HPQ the quantum yield is significantly lower in methanol (0.0004) but not in cyclohexane (0.16). Thus, excitation of the 1-HPQ normal tautomer gives rise to the proton-transfer tautomer fluorescence band (Figure 2, upper, band c), competing with the normal tautomer fluorescence (cf. section 5) in methanol solution. However, 1-HPQ does suffer a larger quantum yield loss of this normal tautomer fluorescence than can be accounted for by the formation of the PT fluorescence. This competitive extra pathway will be discussed in section 5. It is also noteworthy that 1-MPQ has no PT fluorescence (cf. Figure 4) in methanol or in water, as expected.

5. Excited-State Double Proton Transfer in Pyrroloquinolines

In studying the solvent cage dipolar-relaxation spectra of the pyrroloquinolines (II and IV) it was observed that the methanol solution fluorescence spectrum of 1-HPQ (II) exhibited a second fluorescence band centered on ca. 570 nm in the green region. Figure 4 presents the fluorescence emission curves of 1-HPQ (II) (solid curve) and 1-MPQ (V) (dashed curve) in methanol solvent at 298 K. (The methyl of 1-MPQ prevents the double proton transfer[1] that can be catalyzed by the H-bonded proton-

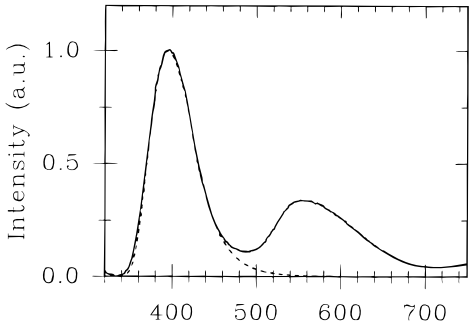
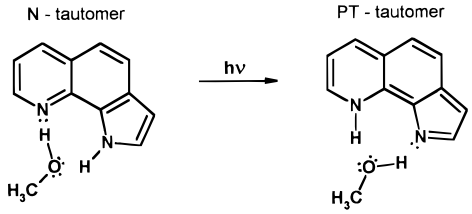


Figure 4. Normalized fluorescence spectra of 1-HPQ (II) (solid line) and 1-MPQ (V) (dashed line) in methanol solution at 298 K, showing the normal tautomer fluorescence at ~400 nm and the proton-transfer tautomer fluorescence at ~550 nm.

SCHEME 1

METHYL ALCOHOL CYCLIC COMPLEX WITH 1-HPQ FOR CATALYZED ESDPT



transfer bridge[37] offered by the methanol molecule, Scheme 1.) In the excited state in such systems the aza-N becomes more proton-attracting, and the pyrrolo-N−H becomes more proton-donating and through the action of the inductive effect the concerted double proton transfer can occur. The schematic double-well potential for the phototautomerization involved in double proton transfer is presented in section 7 (Figures 8 and 9). As in the case of 7-AI (I), the 1-HPQ (II) molecule requires a catalytic transfer via a one-molecule H-bonded proton-donor−acceptor bridge, or a two or more molecule proton-transfer relay.[37] If, however, separate H-bonding molecules occupy the aza-N and pyrrole-N−H sites, or separate chains of such molecules, the double proton transfer would be diminished or blocked.

Figure 5 illustrates the effect of progressive addition of methanol to the dioxane solution of 1-HPQ at 298 K: (a) the (dipolar relaxation shifted) normal tautomer fluorescence at 381 nm progressively diminishes in intensity, and (b) the double-proton-transfer fluorescence intensity at ca. 570 nm progressively increases. However, as stated in section 4, the appearance of the double-proton-transfer fluorescence does not make up for the much larger quantum yield loss of the normal 1-HPQ fluorescence. Because we believe that exciplex formation and consequent electron-transfer quenching interaction with the solvent are eliminated by the study of 1-MPQ, another radiationless process must be involved. The only one that would seem to be available is the pseudo-Jahn−Teller effect,[38] which would provide a radiationless pathway to the ground state. We do not have enough information on states near the S_1' double-proton transfer excited state to definitively establish this pathway, but transient spectroscopy techniques in the phototautomer system could disclose the possibility.

It is interesting that water addition (up to 6.8×10^{-3} M) to dioxane solution of 1-HPQ develops weakly the double-proton-transfer fluorescence. As more water is added, ionic species of 1-HPQ are generated and emit fluorescence in the 480 nm region, interfering with the neutral molecule fluorescence.[24]

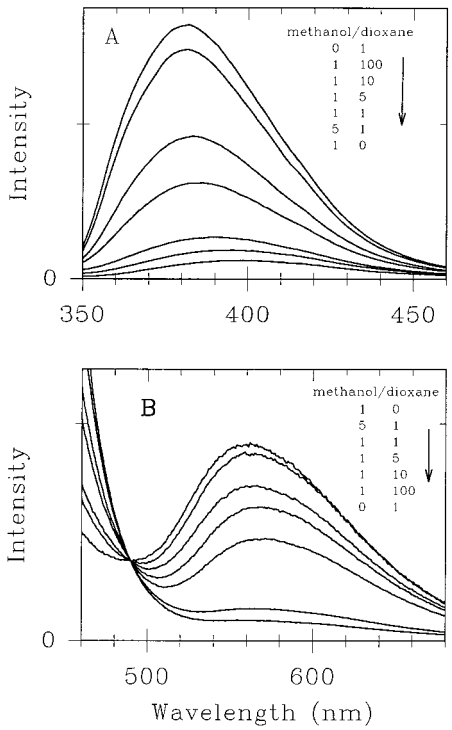


Figure 5. Solvent effects of methanol addition to dioxane solutions of 1-HPQ (II) at 298 K: (A) quenching of normal tautomer emission and (B) catalyzed induction of double-proton-transfer tautomer emission spectra for methanol/dioxane (v/v) solutions.

However, in the excited-state proton-transfer fluorescence region the results are not simple like those illustrated in Figure 5 for methanol addition to 1-HPQ. The additional complexity of HOH self-aggregation complicates a simple relationship for the spectroscopic changes. These results on H_2O-catalyzed proton-transfer parallel those observed[3] in 7-AI, again demonstrating the breaking up of the H-bonded chain of water to permit H-bonding bridges.

In the protic solvent-bridge catalyzed ESDPT it has been recognized[1,15,16] that an energy barrier exists for the bridged relay[37] proton transfer arising from the necessity of solvent H-bonding chain rearrangement to form the H-bonding bridge, in the present case from the pyrrolo-N−H to the aza-N of 1-HPQ. Such a barrier was pictured in Figure 8 of the paper on 3-hydroxyflavone[15] in the consideration of solvent-catalyzed proton transfer (although in that case nonassisted proton transfer commonly is also observable in nonprotic solvents). This barrier represents the diffusion-controlled solvent molecule rearrangement in the catalyzed ESDPT and will be elaborated in section 7 on Excitation Mechanisms.

6. Low-Temperature Glass Solvent Cage Spectroscopy of Pyrroloquinolines

There are three discrete phenomena in the electronic excitation of the pyrroloquinolines 1-HPQ and 1-MPQ that are controllable by solvent cage microviscosity from the range of room-temperature liquid solvents to low-temperature rigid glass solvents. *First*, we recognize that the *dipolar solvent cage relaxation* has a finite time to reach a new equilibrium orientation if the solute molecule excited state electric dipole moment is reoriented internally within its molecular framework. *Second*, in the special case of ESDPT, which requires catalysis of double proton transfer, the *protic solvent rearrangement required for the H-bond bridge formation* from the pyrrolo-N−H to the distal aza-N will introduce a time modulation and

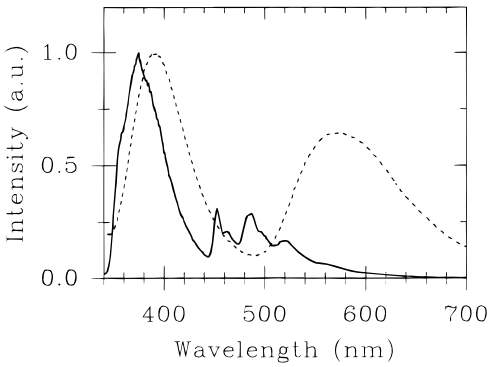


Figure 6. Emission spectra of 1-HPQ (II) in ethanol at 298 K (dashed line) and at 77 K (solid line). Shown are the normal tautomer fluorescence at ~370 nm and phosphorescence origin onset at ~450 nm, at 77 K. The normal tautomer and ESDPT tautomer fluorescence maxima are at 390 and 570 nm (298 K), respectively.

will also be sensitive to the solvent cage microviscosity. *Third*, the metastable *lowest triplet state emission* would be observable as a phosphorescence by the absence of dynamical quenching processes in the rigid glass solvent.

Three of the spectroscopic consequences of the effects alluded to above for 1-HPQ are illustrated in Figure 6. The dashed curve shows the luminescence emission observable at 298 K in ethanol solvent. Centered on 395 nm is seen the dipole-relaxation shifted normal tautomer fluorescence band, and centered on 570 nm is the ESDPT-catalyzed proton-transfer tautomer fluorescence. At 77 K in ethanol rigid glass solvent, the normal fluorescence is blue-shifted close to its position in a hydrocarbon solvent, as the solvent cage dipole relaxation is here inhibited. Simultaneously, the ESDPT phenomenon is completely blocked, with no evidence for the proton-transfer tautomer fluorescence in the 500−700 nm region for the rigid glass solution at 77 K. The $T_1 \rightarrow S_0$ phosphorescence appears conspicuously with a 0,0 resolved band at 452 nm.

The molecule 1-MPQ in methanol at 298 and 77 K (Figure 7, lower) shows the effect of the rigid glass solvent cage inhibition of the solvent dipole relaxation at a larger dispersion display, the diffuse spectrum (normal tautomer fluorescence; cf. Figure 4) being centered on 395 nm for the 298 K solution spectrum. At 77 K the fluorescence spectrum is blue-shifted strongly to its position in a hydrocarbon solution at 298 K, except for a small polarizability shift. The fluorescence spectra (unique normal tautomer) of 1-MPQ in methylcyclohexane solvent at 298 and 77 K are given in Figure 7 (upper).

7. Excitation Mechanisms in the Competition between Dipolar Relaxation and Proton Transfer in Pyrroloquinolines

In this section we shall discuss alternative pathways in the competition between dipolar solvent cage relaxation responding to electronic excitation dipole moment reorientation in the 1-HPQ (II) and 1-MPQ (V) molecules, versus the excited state double-proton-transfer reaction and its spectroscopic effect.

In both dipolar solvent cage relaxation and the solvent reorientation for H-bonding bridge formation required for the protic-solvent catalysis of ESDPT, a time-dependent diffusion-controlled process is involved. It is not obvious or simply correct that a potential barrier of time-dependent origin and character can be added to the equilibrium molecular potential function of a molecule. For this reason a Born−Oppenheimer spectroscopic model for the solvent cage was developed[18,19] in contrast to the classical Franck−Rabinowitch solvent cage. In this model

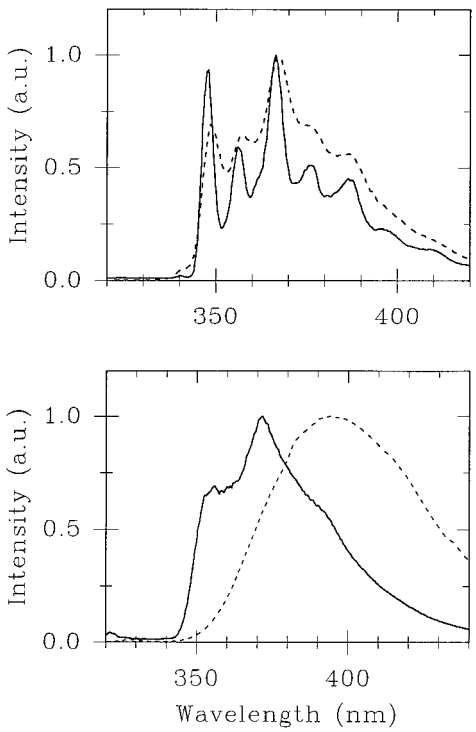


Figure 7. (Upper) fluorescence spectra of 1-MPQ (V) at 298 K (dash line) and at 77 K (solid line) in methylcyclohexane. (Lower) fluorescence spectra of 1-MPQ (V) at 298 K (dash line) and at 77 K (solid line) in methanol.

a criterion was developed on the basis of the inequality of the rates of an ultrarapid process (e.g., solvent cage molecule vibrational motion) vs a much slower process (e.g., solute molecule vibrational motion at the top of a potential barrier, wherein the restoring force is $\partial V/\partial Q = 0$), or any other applicable inequality. In other language, a rapid vibrational motion of the solute may lead to a solvent cage barrier "collision" as if the barrier is immobile, even though on a long time scale the barrier is mobile. The inequality condition may be taken in either sense for rate of solute molecule vibrations vs the rate of the solvent cage molecule vibrational motion, depending on where in the solute molecule motions an intramolecular potential may be horizontal or exhibit a barrier, or wherein the Born−Oppenheimer approximation is applicable otherwise.

If a solute/cage rate of motion inequality is evident, the wave functions for the electronic system (ψ_a), the solute vibration (θ) and the solvent cage vibrational relaxation (ξ) may be written as a simple product (B−O separability), and the solvent cage perturbation can be added[18,19,39] to the solute molecule intrinsic potential $V_{\text{molec-matrix}} = V_{\text{solute}} + V_{\text{interaction}}$.

Thus, in going from a mobile liquid with a very high molecular relaxation rate to the case wherein the microviscosity increases and the solvent cage relaxation rate slows down, the solvent cage barrier will increase in height and width, until ultimately in the glass state an impenetrable infinite solvent cage barrier may result,[39] thereby blocking phenomena dependent on solvent cage relaxation, as illustrated by the experimental results of section 6.

We now consider the competitive pathways available to 1-HPQ as a molecule exhibiting both dipolar-relaxation fluorescence and intramolecular proton-transfer fluorescence spectra. The choice of pathway depends on the relative rates of dipolar solvent cage relaxation as the origin of time-dependent spectral shifts in excited-state fluorescence, vs the solvent rearrangement

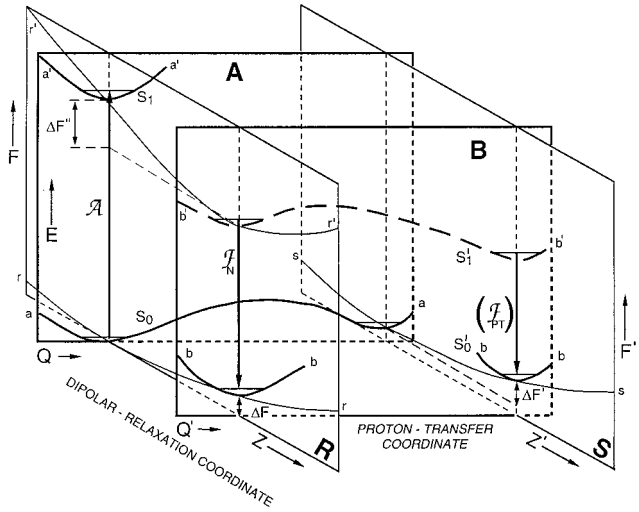


Figure 8. Schematic representation of the competition in spectroscopy of pyrroloquinoline (II) between dipolar relaxation (coordinate Z,Z′ in planes R,S) and H-bond-complex-catalyzed double proton transfer (ESDPT) (coordinates Q,Q′ in planes A,B). Spectra observed are absorption *A* (Figure 1) and the fluorescence F_N (cf. Figure 9).

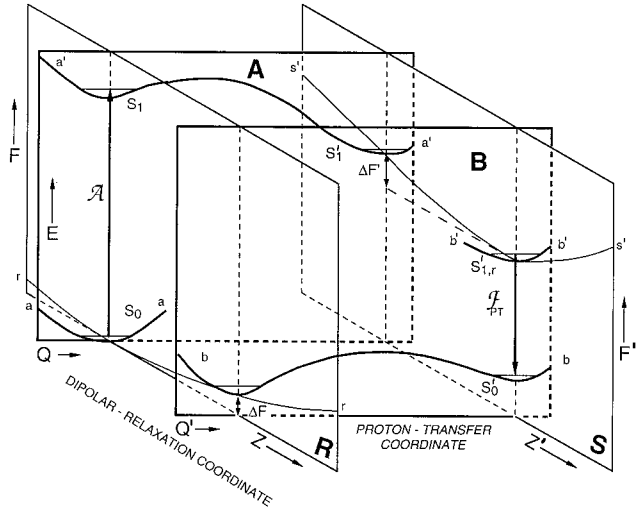
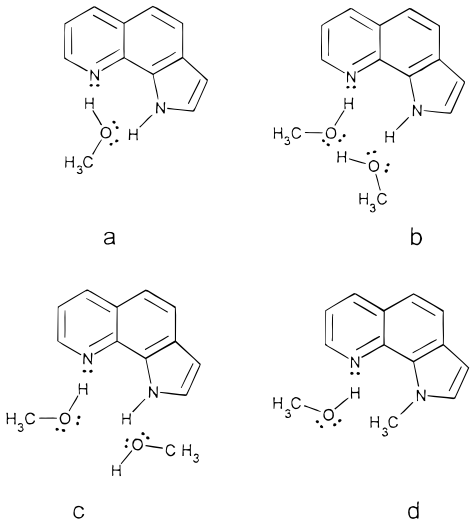


Figure 9. Schematic representation of the ultrafast ESIPT observable for intrinsically H-bonded molecules, illustrating concealed dipolar relaxation. Spectra observed are normal tautomer absorption and proton-transfer tautomer emission.

time-dependent catalysis, or a preformed doubly-H-bonded solvate mode of double-proton transfer. The multicoordinate diagrams of Figures 8 and 9 separate the dipolar solvent cage relaxation steps from the excited state (double) proton-transfer steps. Note that the dipolar relaxation potential curves indicate zero barrier between the initial coordinate system Q and the relaxed coordinate system Q′ (for the $S_0 \rightarrow S_1$ excitation step (cf. Figure 4). This would apply to our case of the methanol solvent at 298 K in which the dipolar relaxation rate is so fast that fluorescence emission is seen for the normal tautomer form of 1-HPQ after the relaxation step. Thus, Figure 8 fits the case at hand. However, after solvent cage dipolar relaxation Figure 8 would pose a competition between the normal tautomer fluorescence and the excited-state double-proton-transfer reaction. This is not at all likely, as an intrinsic intramolecular proton transfer normally far outpaces (by several orders of magnitude) the decay rate of normal tautomer species fluorescence. Therefore, the ESDPT step is shown dashed, as improbable. Thus after fluorescence by the solvent-relaxed normal tautomer S_1 (relax.), a back dipolar relaxation to the initial S_0 (relax.)

SCHEME 2

H-BONDED METHANOL BRIDGES TO 1-HPQ



can take place. As the spectra of Figure 6 indicate, however, *both normal tautomer dipolar-relaxation fluorescence and proton-transfer tautomer fluorescence* are observed at 298 K in methanol solution. An alternative excitation pathway is traced in Figure 9. Here it is assumed that the state S_1 (unrelax.) upon being excited is so rapidly converted by ESDPT that the state S_1' (unrelax.) is produced before solvent dipolar relaxation could take place, outpacing the latter at state S_1. Subsequently, a dipolar relaxation from S_1' (unrelax.) $\sim\sim>$ S_1' (relax.) could take place before the PT-tautomer fluorescence could be observed. In the ground state of the tautomer the S_0' (unrelax.) could undergo rapid reverse DPT, reaching state S_0 (unrelax.), which would then undergo reverse dipolar relaxation S_0 (unrelax.) $\sim\sim>$ S_0 (relax.).

All of the above discussion is contingent upon the relative rates of ESDPT in the solvated molecule vs the solvent dipolar relaxation rate. Waluk et al.[40] recently measured the ESDPT rate for a molecule (dipyrido[2,3-*a*:3′,2′-*i*]carbazole) closely related to 1-HPQ, measuring <30 ps. The molecule reported by Waluk et al. contains a pair of quinolines arrayed in a bilateral symmetry around a central pyrrole ring. This is a slow rate for excited-state proton transfer and might in fact not be fast enough to outpace the solvent dipolar relaxation. In such a case, Figure 8 would apply, and no ESDPT could be observed.

We are thus left with a dilemma. As model *A* we assume that our solution of 1-HPQ consists of two main solute molecular species, that is, solute 1-HPQ (V) molecules with a preformed H-bonded bridge as in Scheme 2a (one methanol bridge) and 2b (a two methanol bridge). Such a model would permit a normal tautomer fluorescence and a subsequent dipolar relaxation to be observed according to the scheme in Figure 9. A temperature dependence would shift the equilibrium.

As model B we assume that no preformed cyclical H-bonded solvates exist, but instead we would require that a species such as Scheme 2c could rearrange with a delay time to form species such as Scheme 2a or 2b. This could then allow a longer ESDPT time, as Waluk et al.[40] observed. Model B could also explain the disappearances of ESDPT in methanol glass solutions at 77 K (Figure 6, solid). The rise time of <30 ps observed by Waluk et al. places such a proton-transfer rate in close competition to dipolar-relaxation rates in liquid protic solvents. Thus, such a

competition would require a hybrid mechanism combining the schemes of Figure 8 and 9.

Thus, it is possible for the ESDPT to involve two different mechanisms and rates, a subpicosecond rise-time rate (direct ESDPT) for a simple preformed cyclic complex like the species in Scheme 2a, and a slow rise-time rate arising from the solvent rearrangement required to change from a species in Scheme 2c to those species of Scheme 2a or 2b. Future work with femtosecond laser techniques should give information to clear up this complex set of interwoven mechanisms.

8. Biochemical and Chemical Applications of Pyrroloquinolines

The 1-*H*-pyrrolo[3,2-*h*]quinoline molecule 1-HPQ (II) has been cited in numerous places in the literature in connection with potentially valuable chemical and biomedical applications. It is thus of substantial interest to understand the electronic structural characteristics and consequent physical and chemical behavior of 1-HPQ in order to derive the fullest understanding of its properties. The propensity of 1-HPQ to form intermolecular H-bonds has been reported in the literature.[30−33] In addition, it is known to form a cyclic dimer connected by strong H-bonds.[33] The H-bonding tendency of 1-HPQ figures prominently in its special chemical and physiological properties. In chemical applications, some derivatives of 1-MPQ have been demonstrated to be good stabilizers for polymers.[41] The molecule 1-HPQ has been proposed as a host molecule in molecular recognition ab initio studies of ring motions.[42]

Among the biomedical reports are its proposed use as a potential anticancer drug.[43] The 1-HPQ molecule exhibits a tuberculostatic activity[44] at 4 μg/mL against tubercle strain H-37. The molecule has also been tested as an antimalarial drug.[45]

The multiple H-bonding possibilities of the 1-*H*-pyrrolo[3,2-*h*]quinoline (also known as pyrido[3,2-*g*]indole, either as single molecular species or as a building block for cavity-shaped hosts, has led to a broad spectrum of biomedical and enzyme−substrate model studies. The urea skeleton as a subunit occurs in a wide variety of molecules, including alkyl-substituted urea, imidazolidone, methyl biotin, and barbital as guest components.

The binding of urea by the pyrroloquinoline (pyridoindole) unit has led to research on urea transport through supported liquid membranes and has suggested their use as anti-uremic agents. This research generally involves molecular ensembles consisting of symmetrical arrays of a pair of pyrido[3,2-*g*]indole subunits.[46−49] These structures then offer an H-bonding complexation cavity in which the guest molecule (e.g., an alkylurea) can be held by as many as four H-bonds, with very high binding constants, as demonstrated by Hegde et al.[47,48] In another extensive series of research by the Hamilton group,[50] the incorporation of such pyrido[3,2-*g*]indole groups into specially designed macrocycle cavity molecules offers as many as six H-bonds for guest molecule binding, as exemplified in their applications to the barbiturates.

Conclusion

The dipolar-relaxation red shift of the fluorescence observed in methanol solutions for both 1-HPQ and its methyl derivatives 1-MPQ offers potential insight into the dynamics of the excitation mechanism. The unusual case of the excited-state double proton transfer (ESDPT) observed simultaneously for the 1-HPQ molecule with its apparent failure to outpace the dipolar-relaxation rate lends additional interest to the competitive pathways. Both the solvent dipolar relaxation and the solvent rearrangement pictured for the cyclical H-bonded complex formation in the ESDPT case are subject to solvent cage perturbations. Differential modulation of their corresponding potentials by such effects will offer a control of the excitation dynamics and the pathway of excitation.

The many studies that have been made of the pyrroloquinoline (pyridoindole) framework as a building block for site-specific interactions in enzyme modeling and its numerous applications make extensions of mechanistic studies on the chemical physics of the associated electronic processes of increasing interest.

Acknowledgment. The authors acknowledge with thanks Prof. J. Catalan for allowing us to publish in advance some theoretical data on the 1-HPQ molecule.

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