Steric Control of the Excited-State Intramolecular Proton Transfer in 3-Hydroxyquinolones: Steady-State and Time-Resolved Fluorescence Study

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3-Hydroxyquinolones (3HQs), similarly to their 3-hydroxychromone analogs, undergo excited state intramolecular proton transfer (ESIPT) resulting in dual emission. In the ground state, 2-phenyl-3HQ derivatives are not flat due to a steric hindrance between the 2-phenyl group and the 3-OH group that participates in the ESIPT reaction. To study the effect of this steric hindrance on the ESIPT reaction, a number of 3HQ derivatives have been synthesized and characterized in different organic solvents by steady-state and time-resolved fluorescence techniques. According to our results, 2-phenyl-3HQ derivatives undergo much faster ESIPT (by nearly 1 order of magnitude) than their 2-methyl-3HQ analogs. Moreover, 1-methyl-2-phenyl-3HQ having a strongly twisted 2-phenyl group undergoes a two- to three-fold slower ESIPT compared to 2-phenyl-3HQ. These results suggest that the flatter conformation of 2-phenyl-3HQ, which allows a close proximity of the 2-phenyl and 3-OH groups, favors a fast ESIPT reaction. The absorption and fluorescence spectra of the 3HQ derivatives additionally confirm that the steric rather than the electronic effect of the 2-phenyl group is responsible for the faster ESIPT reaction. Based on the spectroscopic studies and quantum chemical calculations, we suggest that the 2-phenyl group decreases the rotational freedom of its proximal 3-OH group in the more planar conformation of 2-phenyl-3HQ. As a result, the conformations of 3HQ, where the 3-OH group orients to form an intramolecular H-bond with the 4-carbonyl group, are favored over those with a disrupted intramolecular H-bond. Therefore, the 2-phenyl group sterically favors the intramolecular H-bond and thus accelerates the ESIPT reaction. This conclusion provides a new understanding of the ESIPT process in 3-hydroxyquinolones and related systems and suggests new possibilities for the design of ESIPT based molecular sensors and switchers.

1. Introduction

The mechanism of the excited-state intramolecular proton transfer (ESIPT) in 3-hydroxyflavones (3HFs) is a matter of debate since its introduction by Sengupta and Kasha.[1,2] These authors suggested that ESIPT occurs without energetic barrier so that it is essentially a tunneling process. Moreover, they proposed a critical role for the motion of the 2-phenyl group in the ESIPT reaction. This interpretation was critically revised by Woolfe and Thistlethwaite who claimed that ESIPT in apolar solvents exhibits a high activation barrier but does not depend much on the phenyl motion.[3] The role of the 2-phenyl ring in 3HF was further investigated by Barbara et al. who showed that modification of the phenyl group with a methyl group in its ortho position decreases the intensity of the tautomer emission.[4] A similar effect was observed later with 2-benzofuranyl derivatives.[5] These results point out that a planar configuration of 3HF is required for fast ESIPT reaction.[4,6] The observed phenomenon was explained by the electronic effect of the 2-phenyl group on 3-hydroxychromone moiety. Thus, this planar configuration and, therefore, the optimized conjugation of the phenyl group with the 3-hydroxychromone ring increases the basicity of the 4-carbonyl group and thus strengthens the intramolecular H-bond. This in turn accelerates the ESIPT reaction in line with the very fast ESIPT kinetics (30 fs) recently measured for 3HF in aprotic solvents.[7,8]

Recently, a new class of dyes, 2-aryl-3-hydroxy-4(*1H*)-quinolones (3HQs), which are structural analogs of 3HFs, was introduced[9,10] (Chart 1). The 3HQ dyes exhibit also two emission bands due to ESIPT.[11] Because 3HFs already found a large range of applications as fluorescent probes in biology,[12−17] the 3HQ dyes form a new promising class of dyes, especially due to their higher photostability as compared to the 3HF analogs.[18] However, the development of improved 3HQ dyes for particular applications requires a better understanding of their ESIPT mechanism. In this respect, we have recently shown that the dual emission of 3HQs is highly sensitive to solvent properties, especially polarity and H-bonding basicity. In addition, the introduction of a methyl group at the nitrogen heteroatom was found to increase dramatically the N* emission, so that both bands are of comparable intensities in a number of organic solvents.[11,18]

This strong effect of the N-methyl group, which actually does not change considerably other spectroscopic properties, could be mainly due to changes of the orientation of the 2-phenyl group with respect to the 3HQ heterocycle. A somewhat similar

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CHART 1: Structures of 3HF and 3HQ Derivatives.

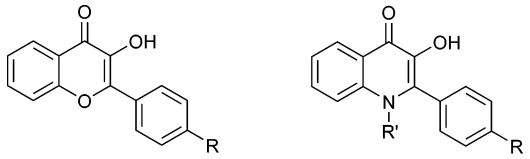
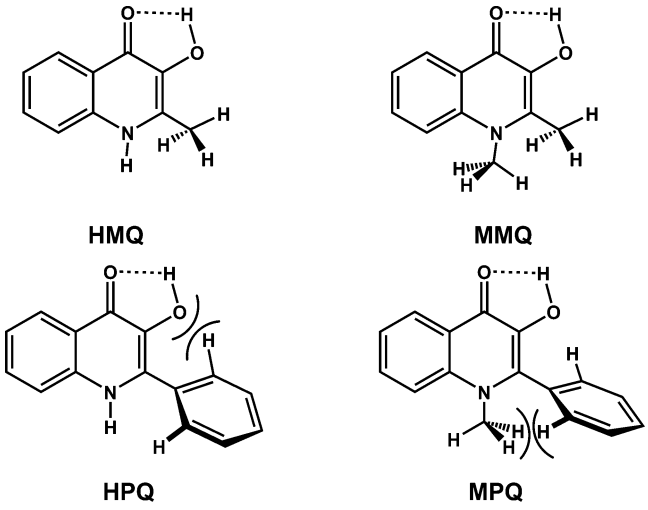


CHART 2: Structures of Studied 3HQs.



effect was observed with ortho-substituted 3HF,[4] but in both cases the mechanism of the effect of the phenyl group remains unclear. To further understand this phenomenon, we synthesized substituted 3HQs without 2-phenyl group and compared their properties with 2-phenyl-substituted derivatives (Chart 2). Our results show that the steric interaction of 2-phenyl and 3-OH groups is of key importance. Indeed, 2-phenyl-3-hydroxy-4(*1H*)-quinolone (HPQ), the 2-phenyl-3HQ with the most flat structure, undergoes the fastest ESIPT, while the 2-methyl-3-hydroxy-4(*1H*)-quinolone (HMQ) and 1-methyl-2-methyl-3-hydroxy-4(*1H*)-quinolone (MMQ) dyes without 2-phenyl group exhibit the slowest ESIPT. These results suggest that the flat structure of HPQ freezes the rotation of the 3-OH group, stabilizing the intramolecular H-bond and thus accelerating the ESIPT reaction.

2. Materials and Methods

HPQ, 1-methyl-2-phenyl-3-hydroxy-4(*1H*)-quinolone (MPQ), HMQ, and MMQ were synthesized by condensing the corresponding anthranilic acid ester in polyphosphoric acid as previously described.[10,11] All the solvents and chemicals were purchased from Aldrich. The solvents were of spectroscopic grade. Absorption spectra were recorded on a Cary 4 spectrophotometer (Varian) and fluorescence spectra on a FluoroMax 3.0 (Jobin Yvon, Horiba) spectrofluorometer. For the steady-state fluorescence measurements, the excitation wavelength was 360 nm. Fluorescence quantum yields were determined by taking quinine sulfate in 0.5 M sulfuric acid (quantum yield, $\varphi = 0.577$)[19] as a reference.

Time-resolved fluorescence measurements were performed with the time-correlated, single-photon counting technique using the frequency-doubled output of a Ti-Sapphire laser (Tsunami, Spectra Physics), pumped by a Millenia X laser (Tsunami, Spectra Physics).[20] The excitation wavelength was set at 320 nm. The fluorescence decays were collected at the magic angle (54.7°) of the emission polarizer. The single-photon events were detected with a microchannel plate Hamamatsu R3809U photomultiplier coupled to a Philips 6954 pulse preamplifier and were recorded on a multichannel analyzer (Ortec 7100) calibrated at 25.5 ps/channel. The instrumental response function was recorded with a polished aluminum reflector, and its full-width at half-maximum was 50 ps. Time-resolved decays were analyzed both by the iterative reconvolution method and the maximum entropy method (MEM).[21] The goodness of the fit was evaluated from the χ^2 values, the plots of the residuals, and the autocorrelation function. Quantum chemical calculations of 3HQs were performed with the AM1 semiempirical method[22] using the MOPAC 6.0 program. To estimate the mean dihedral angle between 2-phenyl and 3HQ rings, we first calculated with AM1 method the dependence of energy versus the dihedral angle and then applied Boltzmann statistics for room temperature to calculate the probability of each conformation.

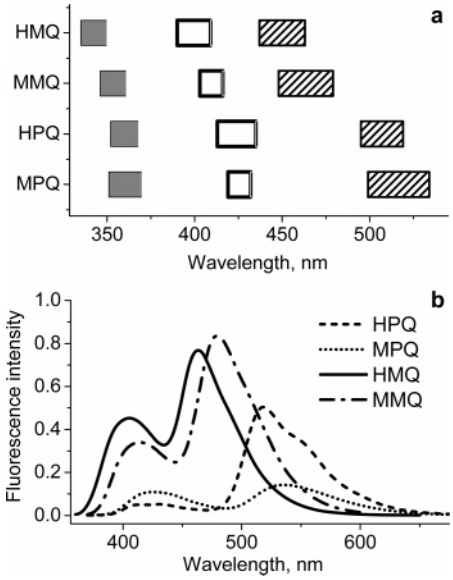


Figure 1. (a) Ranges of absorption and fluorescence maxima of 3HQs in the studied organic solvents. (b) Fluorescence spectra of 3HQs in DMF. The excitation wavelength was 360 nm.

3. Results and Discussion

3.1. Spectroscopic Properties of 3HQ Dyes. Absorption spectra of the four studied dyes in different organic solvents are composed of a single band with the maximum slightly varying with the dye structure. In 2-methyl-3HQ, introduction of a N-methyl group (MMQ) results in a ca. 10 nm red shift of the absorption maximum (Figure 1, Table 1), which is probably related to the electron donor property of the methyl group.

In contrast, the N-methyl group does not shift the absorption maximum of 2-phenyl-3HQ. This absence of red shift is probably related to the decrease of planarity of the 3HQ molecule in the presence of the N-methyl group[18] that compensates the red shift effect of this group. Indeed, a similar decrease of the planarity of a 3HF molecule by modification of its 2-phenyl ring with an ortho-methyl group was previously shown to shift the absorption band by ca. 20 nm to the blue,[4] due to the decrease of the electronic conjugation between the 2-phenyl and chromone rings.

The fluorescence spectra of all dyes in the studied solvents show two emission bands, which according to our previous studies[11,18] can be assigned to the emission of the normal- (N*) and tautomeric- (T*) excited states. For all the dyes, the excitation spectra recorded at these two emission band maxima are very close to the corresponding absorption spectrum, indicating that both emission forms originate from the same

TABLE 1: Spectroscopic Properties of 3HQs in Different Organic Solvents[a]

solvent	3HQ	λabs nm	λN* nm	λT* nm	I_{N*}/I_{T*}	Φ
chloroform	MMQ	360	405	466	0.07	0.37
	MPQ	368	418	518	0.07	0.23
	HMQ	343	390	453	0.08	0.41
	HPQ	364	422	503	0.02	0.44
ethyl acetate	MMQ	354	403	475	0.07	0.42
	MPQ	368	420	532	0.09	0.15
	HMQ	343	394	459	0.09	0.44
	HPQ	363	403	513	0.03	0.36
acetonitrile	MMQ	352	403	471	0.11	0.45
	MPQ	366	419	525	0.10	0.12
	HMQ	341	397	456	0.12	0.49
	HPQ	362	424	510	0.02	0.32
ethanol	MMQ	353	403	470	0.26	0.54
	MPQ	366	424	523	0.51	0.13
	HMQ	340	390	448	0.18	0.63
	HPQ	362	425	514	0.09	0.36
methanol	MMQ	352	404	467	0.45	0.52
	MPQ	364	423	519	1.01	0.13
	HMQ	340	396	448	0.37	0.54
	HPQ	361	425	512	0.34	0.35
DMF	MMQ	361	413	478	0.52	0.72
	MPQ	368	426	536	0.77	0.19
	HMQ	348	406	463	0.59	0.71
	HPQ	367	432	519	0.16	0.39
DMSO	MMQ	360	416	479	0.92	0.80
	MPQ	370	432	534	2.00	0.32
	HMQ	350	409	463	1.05	0.86
	HPQ	368	435	519	0.34	0.51
H_2O (phosphate	MMQ	346		448		0.61
buffer pH 7)	MPQ	351	422	499	0.72	0.13
	HMQ	335		437		0.54
	HPQ	352	413	495	0.12	0.45

[a] λ_{abs}: position of absorption maxima. λ_{N*} and λ_{T*}: position of fluorescence maxima of N* and T* forms. I_{N*}/I_{T*}: ratio of the intensities of the two emission bands at their peak maxima. φ: fluorescence quantum yield.

ground-state species. The position of these two emission bands varies with the substituents. Introduction of a N-methyl group in 2-methyl-3HQ shifts the emission maxima to a small extent. These shifts are close to those observed in the absorption spectra, so that the Stokes shifts of the N* and T* bands remain unchanged. Meantime, introduction of a 2-phenyl group shifts the N* and T* bands to a different extent. While the N* band shifts slightly to the red similarly to the absorption band, the T* band is strongly red-shifted. As a result, the Stokes shift of the T* band of MPQ is 13 nm (1380 cm^{-1}) larger than that of MMQ. A similar shift is observed when HPQ is compared with HMQ. This significant increase in the Stokes shift of the T* band on introduction of the 2-phenyl group can be assigned to the flattening of the HPQ and MPQ molecules in the T* state. Indeed, it was previously shown that the 2-phenyl-3HQ dyes are not planar in their ground states,[18] so that the 2-phenyl group is tilted with respect to the quinolone heterocycle. As a consequence, a flattening of the molecule in the excited state would decrease the energy of the S_1 state (due to increased electronic conjugation) and increase the energy of the nonrelaxed S_0 state due to the unfavored flat conformation in the ground state. Therefore, this flattening of the dye in the excited state leads to the observed larger Stokes shift for HPQ and MPQ. Because this phenomenon is only observed to the T* emission band, we conclude that the studied 2-phenyl-3HQs are probably more flat in the T* state than in the N* state. The importance of this conclusion becomes clearer when the intensity ratio of the two emission bands as well as their time-resolved decays are analyzed.

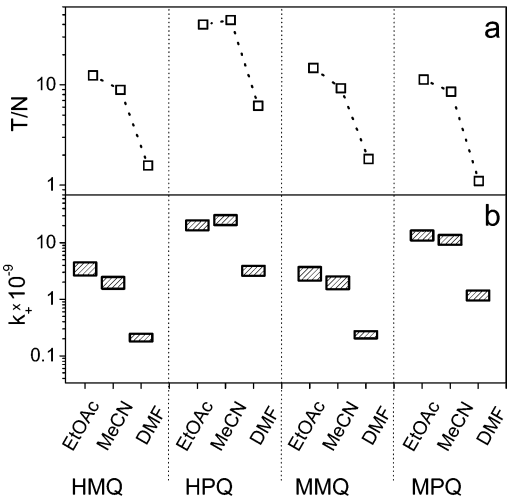


Figure 2. Intensity ratio of T* to N* bands (a) and the ESIPT rate constants (b) of the four 3HQs in ethyl acetate (EtOAc), acetonitrile (MeCN) and dimethylformamide (DMF).

Substitution of the 2-methyl group in HMQ by a 2-phenyl group in HPQ strongly increases the relative intensity of the T* emission, that is, the I_{T*}/I_{N*} ratio (Figure 2a). Further attachment of the N-methyl group in MPQ decreases the I_{T*}/I_{N*} ratio back to the values of the nonsubstituted dye HMQ. Meanwhile, introduction of the N-methyl group in the dye without 2-phenyl group (MMQ) does not change significantly the relative intensities of the two emission bands (Figure 2a, Table 1). Thus, we conclude that introduction of the 2-phenyl group favors the T* emission of the 3HQ dyes. In contrast, the N-methyl group switches off this effect, likely by twisting the phenyl ring out of the quinolone plane, due to steric hindrance. This conclusion is substantiated by the lower quantum yield of MPQ in respect with HPQ, in line with a less planar conformation in MPQ with a lower conjugation of the aromatic units. Similar changes in the intensity ratio were previously reported for 3HFs when the 2-phenyl group is methylated in the sterically hindered ortho position.[4] Thus, it appears that the more planar conformation of HPQ favors the T* emission.

Noticeably, in contrast to the absorption and emission maxima that do not change considerably with the solvent, the intensity ratios vary strongly, especially in solvents of different basicity. For instance, the relative intensity of the T* emission in dimethylformamide (DMF) is much lower than that in acetonitrile, a solvent of similar polarity but much lower basicity. This strong effect of solvent basicity is in line with our recent studies with other 3HQ derivatives and suggests that it is a general property of 3HQ dyes.[23]

3.2. Time-Resolved Fluorescence Measurements. To analyze whether the changes in the I_{N*}/I_{T*} ratios are connected with the rates of the ESIPT reaction, the time-resolved decays of both emission bands of the 3HQ dyes were measured in different solvents. In both HMQ and HPQ dyes, the fluorescence decays of the N* band are clearly faster than those of the T* band (Figure 3). Moreover, the T* decay curves contain a raising component, which indicates that the T* species is formed due to an excited-state process. The fluorescence decay times and the corresponding pre-exponential coefficients of the four studied dyes in various organic solvents are given in Table 2.

The identical short-lived decay times τ_1 for the N* and T* forms and their negative amplitudes α_1 for the T* form confirm that the T* state is produced from the N* state through an ESIPT reaction.[20] The long-lived lifetime τ_2 is systematically observed for the T* emission, but for the N* band, it is either not observed

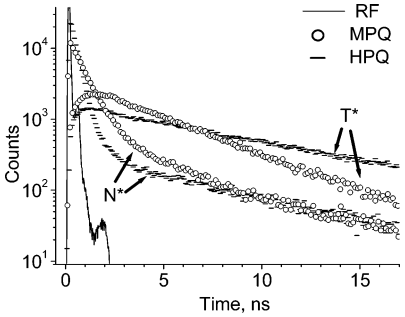


Figure 3. Fluorescence decays of N* and T* forms of 3HQs in DMF. The decays of the N* and T* forms were recorded at 420 and 540 nm, respectively. The response function (RF) of the instrument is also presented. The excitation wavelength was 320 nm.

or shows a small amplitude (Table 2). According to previous studies,[20,24] these results suggest an irreversible ESIPT reaction for all four compounds in the studied solvents. HMQ and MMQ dyes show relatively high τ_2 values in line with their high-fluorescence quantum yields (Tables 1 and 2). Within this couple of compounds, the N-methyl group does not significantly affect the emission kinetics. Introduction of the 2-phenyl group decreases considerably τ_2 and this decrease is especially strong in the MPQ dye, which contains the N-methyl group. The decrease in τ_2 correlates well with the decrease in the quantum yield (Tables 1 and 2), suggesting that the 2-phenyl group probably due to its rotational motion opens new deactivation channels in the dye. This effect appears especially strong when the phenyl ring is twisted with respect to the quinolone heterocycle due to the steric hindrance of the N-methyl group.

The effect of substituents is even stronger on the short-lived decay time τ_1, which is related to the kinetics of the ESIPT reaction (Table 2). Indeed, τ_1 is the longest in 3HQs without 2-phenyl group and does not depend much on the N-methyl group in this case. Introduction of the 2-phenyl group decreases τ_1 up to 12-fold in the absence of the N-methyl group and up to 5-fold in the presence of the N-methyl group (Table 2). To evaluate quantitatively the effect of the substituents on the ESIPT kinetics, we used the model of irreversible phototautomerization,[24] which is in line with the observed low amplitudes of τ_2 at the N* band. This model allows us to neglect the backward kinetic rate constant (k_-) and to estimate the ESIPT rate constants k_+ of the four 3HQs.[20,23]

Through the use of this model, the quantum yields of the N* and T* bands (Q_{N^*} and Q_{T^*}) can be expressed as

$$Q_{N^*} = k_R^{N^*} \tau_1 \quad (1)$$

$$Q_{T^*} = k_R^{T^*} \tau_2 (k_+ \tau_1) \quad (2)$$

where τ_1 and τ_2 are the short-lived and long-lived decay times, respectively, given by the following

$$\tau_1 = 1/(k_R^{N^*} + k_{NR}^{N^*} + k_+); \tau_2 = 1/(k_R^{T^*} + k_{NR}^{T^*})$$

where $k_R^{N^*}$, $k_R^{T^*}$, $k_{NR}^{N^*}$, $k_{NR}^{T^*}$ are the radiative and nonradiative rate constants of the N* and T* forms, respectively, and k_+ is the forward rate constant of ESIPT. Calculation of Q_{N^*} from the steady-state fluorescence spectra (Table 1) and τ_1 from time-resolved measurements (Table 2) allows us to obtain the values of the radiative decay constant $k_R^{N^*}$ from eq 1 for the four 3HQs in different solvents (Table 2).

The value of $k_R^{N^*}$ for HMQ and MMQ is similar in different solvents and is not sensitive to the introduction of the N-methyl group. In contrast, introduction of the 2-phenyl group increases $k_R^{N^*}$ probably by increasing the size and thus the oscillator strength of the fluorophore. Subtraction of $k_R^{N^*}$ from $1/\tau_1$ gives the sum of the nonradiative and ESIPT rate constants, $k_{NR}^{N^*} + k_+$ (Table 2). To estimate the contribution of the forward ESIPT rate constant k_+ to this sum, we used the following approach.

The ratio of eqs 1 and 2 gives us a simplified expression of k_+ as

$$k_+ = \frac{k_R^{N^*}}{k_R^{T^*}} \frac{Q_{T^*}}{Q_{N^*}} \frac{1}{\tau_2} \quad (3)$$

Because the $k_R^{N^*}$ value does not vary significantly with the solvent, we can reasonably assume that the radiative rate constant of the T* state, $k_R^{T^*}$, shows a similar behavior. As a consequence, the ratio $k_R^{N^*}/k_R^{T^*}$ can be considered as constant in different solvents. Thus, k_+ is proportional to the relative intensities of the N* and T* bands and to $1/\tau_2$. Consequently, dividing eq 3 in one solvent by that in another solvent allows us to estimate the relative changes of k_+ from solvent to solvent as

$$\frac{k_+^1}{k_+^2} = a \quad (4)$$

where k_+^1 and k_+^2 are the forward ESIPT rate constants in the first and the second solvent, respectively, calculated from eq 3.

The values of k_+ can be directly obtained from the experimentally obtained sum $k_{NR}^{N^*} + k_+$ in the case if $k_+ \gg k_{NR}^{N^*}$. To check this last assumption, we calculated the ratio of $k_{NR}^{N^*} + k_+$ for a pair of solvents as

$$\frac{k_+^1 + k_{NR}^{N^*1}}{k_+^2 + k_{NR}^{N^*2}} = b \quad (5)$$

where $k_{NR}^{N^*1}$ and $k_{NR}^{N^*2}$ are the nonradiative rate constants for the N* form in the first and the second solvent, respectively.

Then, from the combination of eqs 4 to 5, we can express the $k_+/k_{NR}^{N^*}$ ratio for a particular solvent as

$$\frac{k_+^1}{k_{NR}^{N^*1}} = \frac{a - b\dfrac{k_{NR}^{N^*2}}{k_{NR}^{N^*1}}}{b - a} \quad (6)$$

Considering a pair of solvents with a low value of b, like DMF−ethyl acetate or DMF−acetonitrile and assuming that $k_{NR}^{N^*1}$ and $k_{NR}^{N^*2}$ values are of the same order (which is in line with the values of the fluorescence quantum yields), we can neglect $b\, k_{NR}^{N^*2}/k_{NR}^{N^*1}$ and express the ratio of the rate constants as

$$k_+^1/k_{NR}^{N^*1} \approx \frac{a}{b - a} \quad (7)$$

Using eq 7, we estimated the $k_+/k_{NR}^{N^*1}$ ratio for all four 3HQs in the three studied solvents. This ratio always exceeds 2.5, so that $k_+ > 0.7(k_{NR}^{N^*} + k_+)$. Therefore, the value of k_+ varies between $0.7(k_{NR}^{N^*} + k_+)$ and $k_{NR}^{N^*} + k_+$, which allows us to estimate k_+ for all four compounds and calculate the error of this estimation (Table 2). When different compounds are compared in the same solvent, it appears that the 2-phenyl group increases the ESIPT rate constant dramatically (Table 2). The

TABLE 2: Time-Resolved Fluorescence Parameters of the Two Emission Bands of 3HQs in Organic Solvents

		τ_1[a], ns (α_1[c])		τ_2, ns (α_2[c])						
dye	solvent	N*	T*	N*	T*	Q_{N^*}[a]	Q_{T^*}[a]	$k_R^{N^*}$[b]	$k_+ + k_{NR}^{N^*}$[b]	k_+[b]
HPQ	EtOAc	0.044 (1.00)	0.041 (−0.57)		6.80 (0.43)	0.0056	0.36	0.13	23.1	20 ± 3
	MeCN	0.032 (0.98)	0.049 (−0.48)	0.032 (0.02)	7.30 (0.52)	0.0044	0.30	0.13	28.5	24 ± 4
	DMF	0.283 (1.00)	0.258 (−0.41)		8.28 (0.59)	0.061	0.40	0.24	3.61	3.1 ± 0.6
MPQ	EtOAc	0.063 (1.00)	0.080 (−0.57)		4.23 (0.43)	0.0093	0.15	0.14	15.2	13 ± 2
	MeCN	0.079 (0.98)	0.073 (−0.50)	4.20 (0.02)	4.20 (0.50)	0.011	0.14	0.15	12.7	11 ± 2
	DMF	0.700 (0.9)	0.700 (−0.49)	2.85 (0.1)	3.64 (0.51)	0.088	0.13	0.13	1.32	1.1 ± 0.2
HMQ	EtOAc	0.250 (1.00)	0.264 (−0.53)		8.4 (0.47)	0.027	0.41	0.13	4.64	3.9 ± 0.7
	MeCN	0.392 (0.95)	0.340 (−0.42)	8.6 (0.05)	8.6 (0.58)	0.035	0.46	0.09	2.54	2.2 ± 0.4
	DMF	2.73 (0.99)	2.90 (−0.43)	8.2 (0.01)	12.4 (0.57)	0.25	0.47	0.08	0.25	0.21 ± 0.04
MMQ	EtOAc	0.207 (1.00)	0.231 (−0.40)		8.1 (0.60)	0.021	0.40	0.08	3.84	3.3 ± 0.6
	MeCN	0.383 (1.00)	0.335 (−0.40)		8.8 (0.60)	0.032	0.42	0.09	2.62	2.2 ± 0.4
	DMF	2.90 (0.78)	3.36 (−0.74)	6.5 (0.22)	11.8 (0.74)	0.22	0.50	0.08	0.28	0.24 ± 0.04

[a] τ_1, τ_2 (ns) are the short-lived and long-lived decay times respectively, α_1, α_2 are the relative amplitudes; Q_{N^*},Q_{T^*} are the quantum yields of the N* and T* forms, respectively; $k_R^{N^*}$, $k_{NR}^{N^*}$ ($\times 10^{-9}$ s^{-1}) are, respectively, the radiative and nonradiative rate constants of the N* form; k_+ ($\times 10^{-9}$ s^{-1}) is the forward rate constant of the ESIPT reaction. [b]The error for all values is ±10% due to the precision of the measurements. [c]α_1 and α_2 values were normalized according to $|\alpha_1| + |\alpha_2| = 1$.

highest k_+ value is observed for HPQ. Introduction of the N-methyl group in MPQ results in a 2-fold decrease of k_+ as compared to HPQ. Thus, our data directly evidence that the 2-phenyl group favors the ESIPT reaction, while twisting this group from the quinolone plane by the N-methyl group decreases this effect.

Besides the structure of the 3HQ dye, the nature of the solvent also influences the ESIPT kinetics. Indeed, the ESIPT rate is much lower in DMF than in acetonitrile and ethyl acetate. Taking into account that DMF and acetonitrile are of close polarity but differ by their basicity, we conclude that the ESIPT rate slows down in basic solvents. This conclusion is in line with recent time-resolved studies[23,25] and suggests that basic solvents perturb the intramolecular hydrogen bond in 3HQs through the formation of an intermolecular H-bond of the dye 3-OH group with the solvent. This intermolecular H-bond blocks or slows down the ESIPT reaction.

Importantly, the variations of the k_+ values correlate well with the variations of the relative intensity of the T* emission (Figure 2), suggesting that the ESIPT kinetics is responsible for the strong variations of the intensity ratios. Noticeably, a deviation is observed for the MPQ dye, which exhibits a somewhat lower relative intensity of the T* emission. This may be due to an increase of the nonradiative deactivation rate from the T* state, which is supported by the observed lower quantum yield of MPQ (Table 1).

3.3. Mechanism of the Substituent Effect on the ESIPT Rate. Both electronic and steric effects can be considered to explain the increase of the ESIPT rate constant by the 2-phenyl group in 3HQ. In previous studies, a planar conformation was shown to be required in 2-phenyl-3HF for a fast ESIPT process.[4] This was explained by the electronic effect of the phenyl group, which in a planar conformation increases the basicity of the 4-carbonyl group and thus accelerates the ESIPT. However, this explanation does not consider that the phenyl group can also decrease the acidicity of the 3-OH group and thus, slow down the ESIPT process. Moreover, modification of the 2-phenyl ring by electron donor groups that further increase the basicity of the carbonyl oxygen of 3HFs dramatically slows down the ESIPT reaction, further questioning the interpretation based on the electronic effect.[20] In 3HQs, the electronic effects of the 2-phenyl group also do not explain the variations of the ESIPT rate. Indeed, according to our recent studies, an analog of MPQ containing an electron donor methoxy group on the 2-phenyl ring shows an ESIPT rate close to that of MPQ in the same solvents. This indicates that the electron donor properties of the 2-phenyl group do not strongly affect the ESIPT kinetics. Moreover, when 2-methyl and 2-phenyl-subsituted 3HQ are compared, it appears that the introduction of the 2-phenyl group does not modify the Stokes shift of the N* band (Figure 1a). This is likely because the 2-phenyl group is out of the quinolone plane, so that the aromatic moieties probably form a large dihedral angle that prevents electronic conjugation between them in both ground and excited states. In contrast, the T* state of HPQ shows a much larger Stokes shift than its 2-methyl analog MPQ (Figure 1a, Table 1), suggesting a more planar configuration for the T* state of HPQ (Table 2). Addition of the N-methyl group that decreases the planarity of the MPQ dye does not affect the positions of the absorption and fluorescence bands but strongly modulates the ESIPT rate (Figures 1 and 2). Thus, the orientation of the 2-phenyl group with respect to the

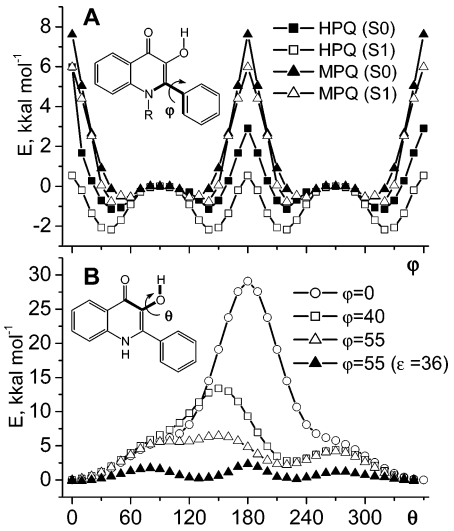
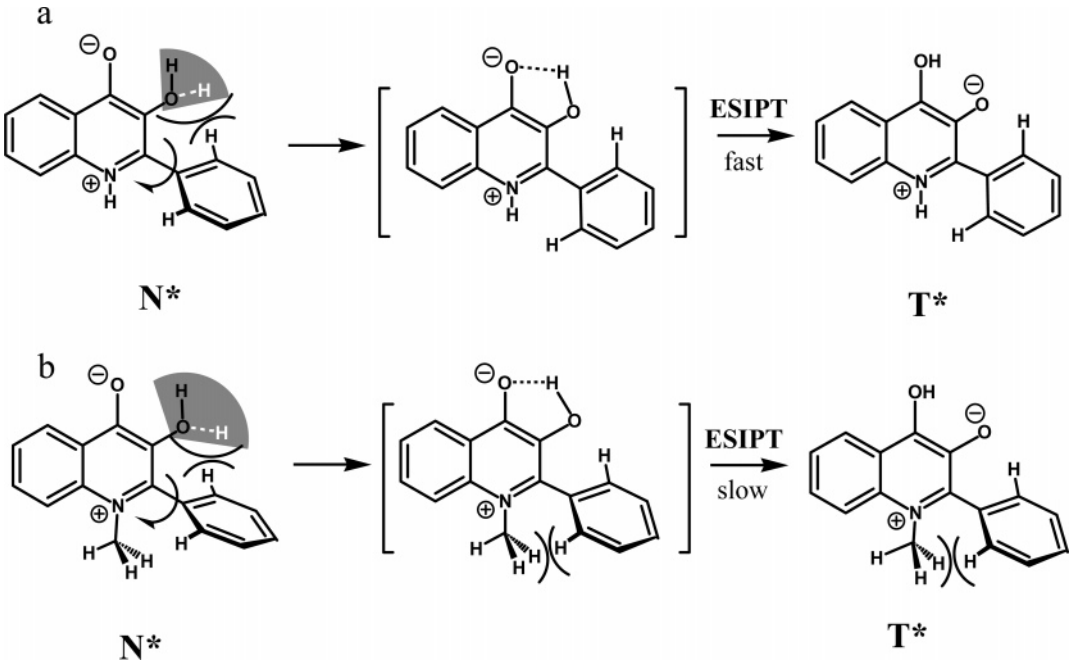


Figure 4. Calculated energies of the 3HQs in the ground (S_0) and Franck−Condon excited (S_1) states at different torsional angles of the 2-phenyl (φ) and 3-OH (θ) groups. (A) Calculated energies of HPQ and MPQ as a function of φ. (B) Calculated energies of HPQ in the S_1 Franck−Condon state as a function of θ for different fixed values of φ. Calculations were done with the AM1 method in vacuum and in acetonitrile ($\epsilon = 36$).

SCHEME 1: (a) The Interaction between the 3-Hydroxy Group and the 2-Phenyl Moiety in N−H Substituted 3HQs Limits Considerably the Rotational Motion of the Hydroxyl Group; (b) the Steric Effect of the Proximal *N*-Methyl Group Decreases the Planarity of the Dye, Enabling a Larger Rotational Freedom of the 3-OH Group.



3HQ plane affects directly the ESIPT kinetics, and this effect does not seem to correlate with the energies of the electronic transitions.

To understand better the observed phenomena, we performed quantum chemical calculations of the energies of the 2-phenyl-3HQ dyes in both their ground state and excited state as a function of the 2-phenyl orientation using the AM1 method.[22] In the ground state, a flat conformation is not favorable for both HPQ and MPQ dyes, so that the mean angle between the 2-phenyl and quinolone rings is 49 and 61°, respectively (Figure 4). In the excited state, the planar conformation becomes slightly more favorable so that the corresponding angle decreases to 38 and 54°, respectively. Thus, according to our calculations, both HPQ and MPQ are not flat, while the excited state favors some flattening. Moreover, the twist between 2-phenyl and 3HQ rings is larger in the MPQ molecule probably due to the steric effect of the N-methyl group. Evidently, HPQ is not flat due to the steric interaction of the 2-phenyl group with the 3-OH group. This interaction may in turn affect the rotational motion of the 3-OH group. We analyzed the motion of the 3-OH group in the excited state of HPQ with the AM1 method, and we found that its rotational freedom depends strongly on the orientation of the 2-phenyl ring. Thus, in the case of the relatively flat conformation of HPQ (with the φ angle between the 2-phenyl and quinolone rings being lower than 40°) the rotation of 3-OH is restricted, favoring the H-bonding of the 3-OH group with the 4-carbonyl group. When φ is increased to 55°, the energy barrier of rotation of the 3-OH group decreases significantly, thus favoring the disruption of the intramolecular H-bond. Because the mean φ angle in MPQ in the excited state is close to 55°, the intramolecular H-bond is likely disrupted in this dye. If the solvent (acetonitrile, $\epsilon = 36$) is taken into account in the calculations (at $\varphi = 55°$), the energy barrier of the 3-OH group rotation is almost absent, so that at room temperature a large population of the dye with disrupted intramolecular H-bond is expected.

These calculations allow us to conclude that in the excited state flattening of the HPQ molecule restricts the rotational motion of the 3-OH group so that its proton is forced to be directed toward the 4-carbonyl group (Scheme 1a). This in turn stabilizes the intramolecular hydrogen bond and thus increases the ESIPT rate constant.

In contrast, introduction of the N-methyl group twists the 2-phenyl moiety out of the quinolone plane and thus increases the rotational freedom of the 3-OH group (Scheme 1b). As a consequence, it weakens the intramolecular H-bond and decreases the ESIPT rate. The largest rotational freedom is probably observed for 3HQs without 2-phenyl group, which show the lowest rates of ESIPT reaction (Table 2). In 3HFs, the ortho-methyl group in 2-phenyl, which also decreases significantly the planarity of the fluorophore, was also reported to decrease the strength of the intramolecular H-bond between 3-OH and 4-carbonyl groups, so that a fraction of the molecules exists without intramolecular H-bond.[4] These data strongly support our hypothesis that the 3-OH group in the nonplanar species of 3HQs (and 3HFs) exhibits higher rotational freedom and thus lower binding to the 4-carbonyl group. In basic solvents, which can form H-bond with 3-OH group, the decrease in the ESIPT rate is especially strong, so that for 2-methyl-3HQ dyes in DMF we observe an extremely slow ESIPT process (Table 2, Figure 2b). Such unusually slow ESIPT is likely the consequence of the large rotational freedom of the 3-OH group that allows the basic solvent to disrupt the intramolecular H-bond and thus to slow down the ESIPT reaction.

4. Conclusions

Our results show that substitution of 2-methyl with 2-phenyl group in 3HQs accelerates significantly the ESIPT reaction, while twisting of the 2-phenyl group out of the 3HQ plane by a proximal substituent slows down the ESIPT reaction. These data together with the absorption and fluorescence spectra of the dyes suggest that the ESIPT reaction in 2-phenyl-3HQ is accompanied by a flattening of the molecule (i.e., a decrease in the dihedral angle between the 2-phenyl and 3-hydroxyquinolone rings). On the basis of our data, we hypothesize that the steric

influence of the phenyl ring on the ESIPT reaction dominates over its electronic influence. According to our model, the 2-phenyl group being close to the plane of the 3HQ heterocycle sterically limits the rotational motion of the 3-OH group, stabilizing the intramolecular H-bond of the 3-OH group with the 4-carbonyl group and thus accelerating the ESIPT reaction.

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Supporting Information Available: All the recorded fluorescence decay curves (PDF). This material is available free of charge via the Internet at http://pubs.acs.org.

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