J. Phys. Chem. A 2000, 104, 6095−6102 6095

Photophysical Properties of Hydroxy-Substituted Flavothiones

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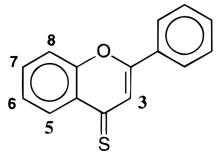
Received: January 6, 2000; In Final Form: March 26, 2000

Flavothione and a number of synthesized hydroxy- (mono- and di-) substituted flavothiones have been thoroughly examined, particularly regarding their absorption, emission, photophysical (triplet yields and lifetimes), and oxygen-photosensitizing characteristics. These were all studied as a function of the nature of the solvent (four), which was particularly critical in terms of aiding in determining the energy and configurational nature of the lowest triplet state as well as the mechanism of intersystem crossing. Theoretical calculations were also performed. Both the location and number of hydroxyl groups have a substantial impact on the nature of the lowest excited triplet state as well as on the relative location of the two lowest excited singlet and triplet states. These in turn affect the magnitude and even the existence of triplet-state occupation as well as the ability to sensitize oxygen (to singlet oxygen). Three groups of compounds exist as characterized by the configurational nature of the triplet and the mechanism of intersystem crossing, or the essential absence of intersystem crossing altogether. The quantum yield of singlet oxygen formation is high for one group where the T(π, π*) state is lowest and generally high in another group where the T(n, π*) state is lowest, except in ethanol where competitive H-atom abstraction occurs. The potential of all hydroxy compounds as photosensitizers is evaluated.

Introduction

We have a continuing interest in the development of photosensitizers that will be effective in a wide variety of systems and circumstances; for example, see refs 1 and 2. Here we report on hydroxy-substituted flavothiones:



This paper presents a comprehensive study of the consequences of mono- (n-OHFT with n = 3, 5, 6, and 7) and bihydroxyl (n,n'-diOHFT with n,n' = 3,6 and 5,7) substitution of flavothione (FT) on the photophysical properties of the resulting flavothiones. The primary aims are to rationalize[1] the effect of the location and number of hydroxyl groups on the energy and orbital origin of the first and the second excited singlet and triplet states,[2] the consequence of the latter on the photophysical properties including the presence of fluorescence, the triplet yields and lifetimes, and the singlet oxygen yields, and[3] how the foregoing depend on the nature of the solvent. Our final goal is to evaluate which compounds and conditions are best suited to produce good photosensitizers.

In addition, it was important that the 5-methoxy-substituted compound was studied regarding the nature of its lowest singlet and triplet excited states and triplet quantum yield to help elucidate the cause of some important changes in photophysical properties of two of the hydroxy-substituted derivatives compared with all others.

Detailed studies of the solution spectroscopy of the parent moiety chromothione[2] as well as FT itself[3] have been reported.

Experimental Section

Substituted flavothiones were synthesized from reaction of the parent hydroxyflavones with either Lawesson's Reagent[2] or diphosphorus pentasulfide[2,4] (Table 1). Flavones were purchased from Merck and Extrasynthesis. Flavothiones were purified by column chromatography or PTLC[5] and then recrystallized from methanol−hexane (1:1). Mass spectroscopy (KRATOS MS 25 RE), ^{1}H NMR (BRUCKER AMX300), FTIR (MATTSON 7000), and UV−vis absorption spectroscopy (OLIS 15) were used to identify the synthesized flavothiones. The crystals were stored in the dark to avoid photodegradation. Melting points were measured with a BUCHI 530 apparatus (Table 1).

Fluorescence and phosphorescence spectra were run in Spex Fluorolog 2021 and were corrected for the instrumental response.

Fluorescence decays were measured with a picosecond-time-resolved fluorimeter earlier described: frequency-doubled emission, λ_{exc} = 400 nm, from a mode-locked Ti−saphire laser (Spectra-Physics), pumped by an argon Ion (Spectra-Physics) laser, repetition rate 800 kHz, pulse width after electronic distortion 28 ps.[6]

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10.1021/jp000084y CCC: $19.00

TABLE 1: Flavothiones: Synthesis Method, Reaction Yield, and Melting Point

compound	reagent	solvent	temp (°C)	irr. time (min)	yield (%)	mp (°C)
FT	R.L.	DME	25	10	76	83−85
	P_2S_5	MeCN	25	10	80	
3-OHFT	P_2S_5	MeCN/Dx (5:2)	80	15	62	64−68
5-OHFT	R.L.	benzene	40	30	6	130−132
6-OHFT	R.L.	benzene	80	60	30	206−208
	P_2S_5	MeCN/THF (1:4)	25	10	21	
7-OHFT	R.L.	THF	65	10	14	191−194
3,6-diOHFT	R.L.	THF	80	40	23	185−187
5,7-diOHFT	R.L.	benzene	40	20	17	216−218
	P_2S_5	MeCN	70	20	35	
3,7-diOHFT	R.L.	benzene/Tol(1:1)	80	15	17	199−202
	P_2S_5	MeCN	40	10	16	
7,8-diOHFT	R.L	Tol	40	30	77	243−246
	P_2S_5	MeCN	25	25	6	

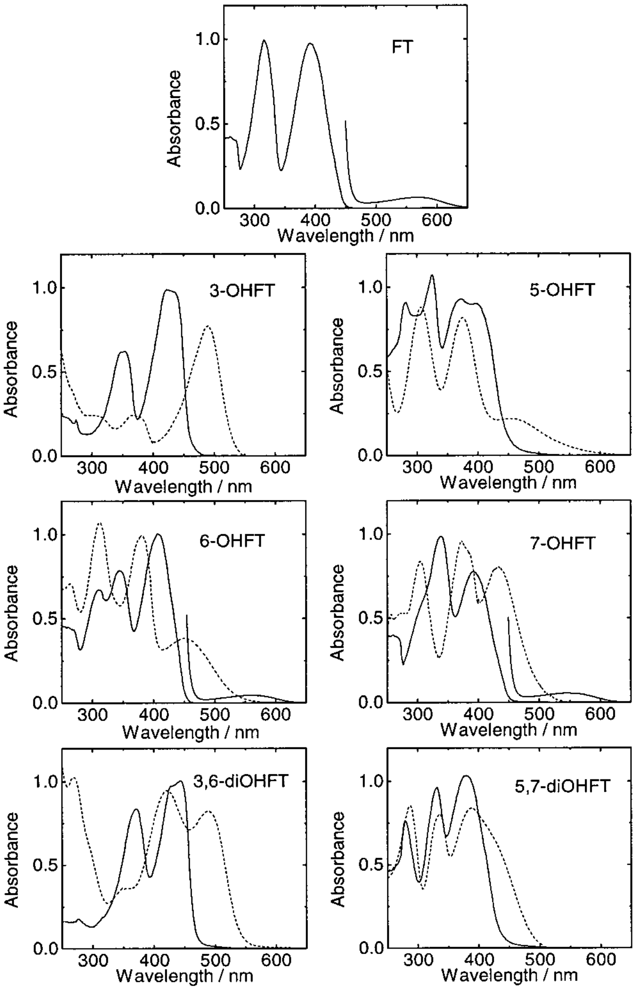

Figure 1. Absorption spectra of flavothiones in benzene (solid line) and in water at pH = 14 (dashed line). Also included are $n \rightarrow \pi^*$ absorption obtained in a 10 cm path cell for FT, 6-OHFT, and 7-OHFT.

Triplet-formation quantum yields and lifetimes were measured with a flash photolysis setup previously described (Nd:YAG Continuum, Surelite II, third harmonics, $\lambda_{exc} = 355$ nm, pulse width ca. 7 ns and energy ≤ 1 mJ pulse^{-1}).[7,8] First-order kinetics was observed for the decay of the lowest triplet state (T−T annihilation was prevented by the low excitation energy). The triplet lifetimes were measured at absorbance ca. 0.2; the concentration effect on triplet lifetime was not investigated. The transient spectra were obtained by monitoring the optical density change at intervals of 5−10 nm over the 300−800 nm range and averaging at least 10 decays at each wavelength.

The product $\epsilon_T\phi_T$ (where ϵ_T and ϕ_T are the triplet extinction coefficient and quantum yield, respectively) was measured at the triplet−triplet absorption maxima. The calibration of the experimental setup was made with an optically matched solution of benzophenone in acetonitrile ($\epsilon_T\phi_T = 6500$ M^{-1}cm^{-1}).[9]

The ϕ_T values in benzene were generally measured by comparing the change of absorbance of β-carotene triplet sensitized by energy transfer from a substrate, ΔA_β, and from benzophenone, ΔA_β(B), used as standard [ϕ_T(B) = 1,[10] under the same experimental conditions (absorbance at 355 nm and concentration of β-carotene)].[11] The efficiency of energy transfer from the substrate, P_{et}, and from benzophenone, P_{et}(B), was also taken into account by measuring the triplet lifetime of the donor in the absence (τ_T) and in the presence (τ_T') of β-carotene:

$$\phi_T = \phi_T(B)\,\frac{\Delta A_\beta}{\Delta A_\beta(B)}\,\frac{P_{et}(B)}{P_{et}} \qquad (1)$$

$$P_{et} = \frac{\tau_T - \tau_T'}{\tau_T} \qquad (2)$$

Only for 5-OHFT and 5,7-diOHFT (and 5-OCH_3FT) were we unable to measure the ϕ_T by energy transfer (see later discussion).

The smaller solubility of β-carotene in other solvents, the reactivity of benzophenone in ethanol (EtOH), and the generally short-lived triplet of flavothiones in EtOH prevented the use of the same method to obtain ϕ_T in EtOH, acetonitrile (MeCN), and the hydrogen-bonding trifluoroethanol (TFE), which is able to destabilize the *n*, π^* states. For these reasons, ϕ_T in the more polar solvents was obtained from the singlet-oxygen quantum yields in air-equilibrated solutions by assuming that the fraction of triplet quenched by molecular oxygen leading to singlet oxygen was independent of the solvent (and taking into account the fraction of triplet quenched by the molecular oxygen in air-equilibrated solutions)

$$(\phi_T)_{EtOH} = \frac{(\phi_\Delta)_{EtOH}}{(S_\Delta)_{benz}}\left(\frac{\tau_T}{\tau_T - \tau_T'}\right)_{EtOH}\left(\frac{\tau_T - \tau_T'}{\tau_T}\right)_{benz} \qquad (3)$$

where $(S_\Delta)_{benz} = (\phi_\Delta/\phi_T)_{benz} \approx 0.6$ was measured in benzene and τ_T and τ_T' correspond to the triplet lifetimes measured, respectively, in Ar-saturated and air-equilibrated solutions.

The ϵ_T values in benzene were obtained from the $\epsilon_T\phi_T/\phi_T$ ratio (the first two terms from the OD change and the denominator from β-carotene sensitization). Furthermore, ϵ_T of FT in acetonitrile was measured by energy transfer from benzophenone at $\lambda_{exc} = 355$ nm; ϵ_T of 6500 M^{-1} cm^{-1} (at 520 nm)[9] was used for the donor (benzophenone).

All measurements were carried out at 22 ± 2 °C; the solutions were saturated by bubbling with argon. The experimental errors on τ_T were estimated to be about ±10% while those on ϵ_T and ϕ_T were ±15%.

The singlet oxygen yields (ϕ_Δ) were determined by measuring the phosphorescence intensity of $O_2(^1\Delta_g)$ with a germanium diode detector in air-equilibrated solutions.[12] The amplified signal extrapolated at zero time was plotted as a function of laser dose. Linear relationships were obtained for all substrates and for phenalenone (in each solvent) used as standard ($\phi_\Delta = 0.97 \pm 0.03$).[13] The ϕ_Δ values were then obtained from the ratio of the slopes of each compound to that of phenalenone

TABLE 2: Experimental Absorption Maxima (λ_{exp}) and Extinction Coefficients (ϵ) of Flavothiones in Benzene and Electronic Transitions Calculated by the INDO/1-CI Semiempirical Method after Geometrical Optimization with INDO/1

						CI	
compound	transition	λ_{exp} (nm)	ϵ (10^4 M^{-1}cm^{-1})	λ_{cal} (nm)	f	%	orbitals
FT	$S_0 \rightarrow S_1$	569	0.005	582	0.000 04	78	$n_{H-1} \rightarrow \pi_L$*
	$S_0 \rightarrow S_2$	392	1.85	381	0.52	91	$\pi_H \rightarrow \pi_L$*
	$S_0 \rightarrow S_3$	319		320	0.089	66	$\pi_H \rightarrow \pi_{L+1}$*
	$S_0 \rightarrow S_4$			305	0.000 90	81	$n_{H-1} \rightarrow \pi_{L+1}$*
	$S_0 \rightarrow S_5$	316	1.89	298	0.45	53	$\pi_{H-2} \rightarrow \pi_L$*
						22	$\pi_H \rightarrow \pi_{L+1}$*
3-OHFT	$S_0 \rightarrow S_1$			574	0.000 01	86	$n_{H-1} \rightarrow \pi_L$*
	$S_0 \rightarrow S_2$	435	2.38	404	0.62	93	$\pi_H \rightarrow \pi_L$*
	$S_0 \rightarrow S_3$	354	1.52	338	0.20	56	$\pi_{H-2} \rightarrow \pi_L$*
						35	$\pi_H \rightarrow \pi_{L+1}$*
	$S_0 \rightarrow S_4$			302	0.42	57	$\pi_H \rightarrow \pi_{L+1}$*
5-OHFT	$S_0 \rightarrow S_1$	608[a]		625	0.000 002	77	$n_H \rightarrow \pi_L$*
	$S_0 \rightarrow S_2$	398	1.36	372	0.53	88	$\pi_{H-1} \rightarrow \pi_L$*
	$S_0 \rightarrow S_3$	326	1.54	340	0.13	74	$\pi_{H-2} \rightarrow \pi_L$*
	$S_0 \rightarrow S_4$			329	0.000 38	83	$n_H \rightarrow \pi_{L+1}$*
	$S_0 \rightarrow S_5$			316	0.018	42	$\pi_{H-1} \rightarrow \pi_{L+1}$*
6-OHFT	$S_0 \rightarrow S_1$	562	0.005	577	0.000 01	79	$n_{H-1} \rightarrow \pi_L$*
	$S_0 \rightarrow S_2$	409	1.80	391	0.48	90	$\pi_H \rightarrow \pi_L$*
	$S_0 \rightarrow S_3$	345	1.42	337	0.16	43	$\pi_{H-2} \rightarrow \pi_L$*
						34	$\pi_H \rightarrow \pi_{L+1}$*
	$S_0 \rightarrow S_4$	311	1.18	313	0.24	35	$\pi_{H-2} \rightarrow \pi_L$*
						49	$\pi_H \rightarrow \pi_{L+1}$*
7-OHFT	$S_0 \rightarrow S_1$	548	0.006	569	0.000 04	77	$n_{H-1} \rightarrow \pi_L$*
	$S_0 \rightarrow S_2$	393	1.51	383	0.42	90	$\pi_H \rightarrow \pi_L$*
	$S_0 \rightarrow S_3$	338	1.93	328	0.43	60	$\pi_{H-2} \rightarrow \pi_L$*
						29	$\pi_H \rightarrow \pi_{L+1}$*
	$S_0 \rightarrow S_4$			303	0.001 3	77	$n_{H-1} \rightarrow \pi_{L+1}$*
	$S_0 \rightarrow S_5$	300(s)	1.00	302	0.39	26	$\pi_{H-2} \rightarrow \pi_L$*
						56	$\pi_H \rightarrow \pi_{L+1}$*
3,6-diOHFT	$S_0 \rightarrow S_1$			569	0.000 08	86	$n_{H-1} \rightarrow \pi_L$*
	$S_0 \rightarrow S_2$	446	1.84	412	0.55	92	$\pi_H \rightarrow \pi_L$*
	$S_0 \rightarrow S_3$	371	1.49	356	0.27	67	$\pi_{H-2} \rightarrow \pi_L$*
	$S_0 \rightarrow S_4$			310	0.24	59	$\pi_H \rightarrow \pi_{L+1}$*
5,7-diOHFT	$S_0 \rightarrow S_1$			651	0.000 08	77	$n_H \rightarrow \pi_L$*
	$S_0 \rightarrow S_2$	381	1.90	378	0.44	88	$\pi_{H-1} \rightarrow \pi_L$*
	$S_0 \rightarrow S_3$	332	1.77	332	0.13	52	$n_H \rightarrow \pi_{L+1}$*
	$S_0 \rightarrow S_4$			330	0.26	44	$\pi_{H-2} \rightarrow \pi_L$*

[a] Value obtained with 5-methoxyflavothione.

multiplied by the known ϕ_Δ of the standard. At least 50 kinetic measurements were averaged for each solution.

Quantum mechanical calculations were performed using the Cerius^2 software. Transition energies and oscillator strengths were obtained by INDO/1-CI calculations, after geometrical optimization with the INDO/1 model.[14] In all cases, electron excitations (singly excited) from the 20 highest occupied to the 20 lowest virtual molecular orbitals were used.

Results

Singlet−Singlet Absorption. Figure 1 shows the absorption spectra of a series of hydroxy-substituted flavothiones in benzene and water (pH = 14). The spectra in benzene are strongly red-shifted with respect to those of the parent flavones, the largest bathochromic shift being observed with 3,6-dihydroxyflavothione. In polar solvents such as methanol, the spectra (not shown) are blue-shifted by 2−4 nm with respect to benzene and an additional absorption band was observed at longer wavelengths. Addition of a drop of concentrated HCl or the use of carefully dried methanol removes this band, while addition of concentrated NaOH increases its intensity at the expenses of the remaining absorption bands. The spectra obtained in water at pH = 14 show this same band, which is assigned to the anionic forms of the compounds. Acidification of the aqueous solutions with HCl induces precipitation.

Figure 1 also shows the red edge of the absorption spectra in benzene, measured in 10 cm optical length cells, for some of the compounds. The $S_0 \rightarrow S_1(n, \pi^*)$ transition is clearly observed for the following compounds: FT, 6-OHFT and 7-OHFT, and 5-OCH$_3$FT. In Table 2, experimental (in benzene) and calculated transition wavelengths are given for the hydroxy compounds.

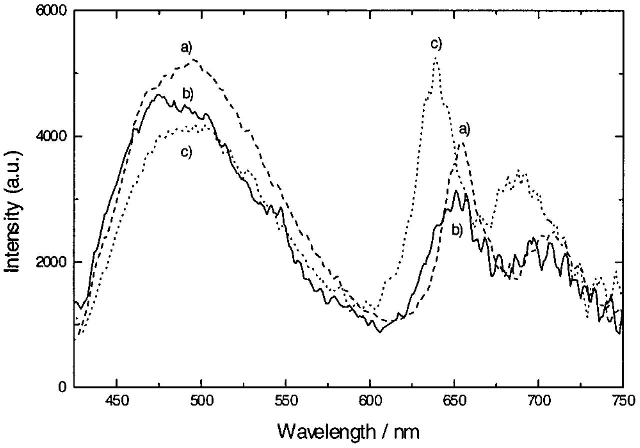

Figure 2. Fluorescence and phosphorescence spectra in 3-methylpentane at 298 K: (a) FT; (b) 6-OHFT; (c) 7-OHFT.

Fluorescence and Phosphorescence. The same three compounds in the series (FT, 6-OHFT, and 7-OHFT) show fluorescence emission from S_2 (Figure 2).

The fluorescence quantum yields (ϕ_f) are low, and the fluorescence decays are single exponential with lifetimes (τ_f)

TABLE 3: Fluorescence Maxima (λ_f), Quantum Yields (ϕ_f), and Lifetimes (τ_f) in 3-Methylpentane (3MeP), Benzene, and Methanol (MeOH) at 293 K, Triplet Energies (E_T) from Phosphorescence Maxima, and Phosphorescence Lifetimes (τ_P) in 3MeP and MeOH at 77 K

compound	solvent	λ_f (nm)	ϕ_f (10^{-4})	τ_f (ps)	E_T (cm^{-1})	τ_p (μs), 77 K
FT	3-MeP	490	5.4	11	15 900	59
	benzene	499	3.8	8.0		
	MeOH	479	1.8	7.3	16 200	49
6-OHFT	3-MeP	484	4.7		16 000	82
	benzene	486	3.7	<3		
	MeOH	470	0.8	3.9	16 400	90
7-OHFT	3-MeP	496	4.4		15 700	58
	benzene	490	2.5	3.6		
	MeOH	480	0.6	3.4	16 750	62

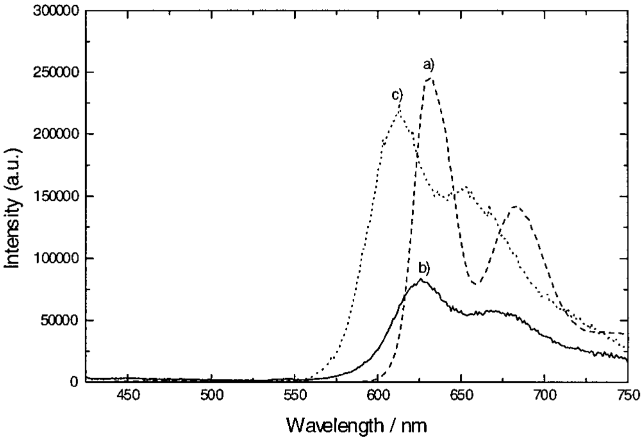

Figure 3. Phosphorescence spectra in 3-methylpentane at 77 K: (a) FT; (b) 6-OHFT; (c) 7-OHFT.

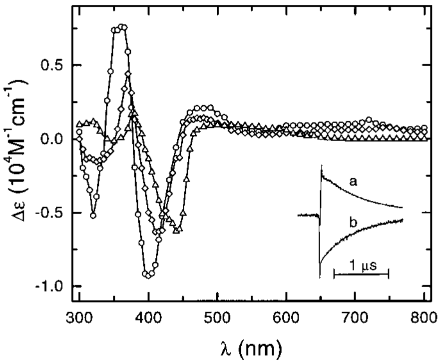

Figure 4. Triplet−triplet spectra of FT (○), 3-OHFT (△), and 6-OHFT (◇) in benzene (λ_{exc} = 355 nm). Inset: Decay and recovery kinetics of FT recorded at 360 (a) and 400 (b) nm.

TABLE 4: Triplet−triplet Absorption Wavelengths (λ_{max}), Triplet Lifetimes (τ_T), and Quantum Yields of Triplet Formation (ϕ_T) of Hydroxyflavothiones, in Benzene, Ethanol (EtOH), Acetonitrile (MeCN), and Trifluoroethanol (TFE) at 295 K

compound	solvent	λ_{max} (nm)	τ_T (μs)	$\epsilon_T{}^a$ (M^{-1} cm^{-1})	ϕ_T	$\phi_T{}^b$
FT	benzene	$\underline{360}$, 480	0.93	7600	1.0	
	EtOH	$\underline{355}$, 480	0.04	7450^c	0.90	1.0
	MeCN	$\underline{350}$, 490	0.31	7300	0.93	1.0
	TFE	$\underline{355}$, 480	0.70	7450^c	0.95	0.86
3-OHFT	benzene	$\underline{320}$, $\underline{380}$, 520	7.1	1700	1.0	
	EtOH	$\underline{370}$, 480	5.9	2350^d		0.95
	MeCN	$\underline{370}$, 480	3.6	3500^d		0.89
	TFE	$\underline{370}$, 510	6.3	3400^d		1.0
6-OHFT	benzene	$\underline{370}$, 480, 750	1.6	4400	0.96	
	EtOH	$\underline{350}$, 465, 670	0.10	4800^d		0.85
	MeCN	$\underline{365}$, 470	0.63	4700^d		0.87
	TFE	$\underline{470}$	0.91	1050^d		0.83
7-OHFT	benzene	$\underline{380}$, 505, 710	1.1	3800	0.98	
	EtOH	e	0.08			0.91
	MeCN	$\underline{370}$, 505	0.38	3900^d		0.73
	TFE	$\underline{515}$	0.45	1400^d		0.70
3,6-diOHFT	benzene	$\underline{325}$, 395, 480, 570	12.0	1250	0.96	
	EtOH	$\underline{300}$, 480, 580	9.1	750^d		0.93
	MeCN	$\underline{470}$, 570	7.1	1540^d		1.0
	TFE	$\underline{490}$, 560	8.1	2500^d		0.95

[a] Extinction coefficient of the underlined maximum. [b] Obtained from the singlet-oxygen quantum yields by assuming the S_Δ measured in benzene (see text). [c] Average of the measured values in benzene and MeCN. [d] Values obtained from the ratio ($\epsilon_T\phi_T/\phi_T$). [e] The triplet−triplet absorption spectrum is strongly hidden by the absorption of a photoproduct and by the recovering of the ground state.

in the picosecond region (Table 3). Both ϕ_f and τ_f are markedly decreased in polar solvents. In methanol, the lifetimes of 6-OHFT and 7-OHFT are shorter than the time resolution of our equipment (~ 3 ps).

The emission spectra of FT, 6-OHFT, and 7-OHFT show a second emission band, phosphorescence, at longer wavelengths at room temperature (Figure 2). The phosphorescence spectra in 3-methylpentane (3MeP) at 77 K, Figure 3, show maxima at about the same wavelengths (~25 nm blue-shifted at 77 K).

Data on the phosphorescence maxima (also considered to be the 0−0 band) and the lifetimes (τ_P) in 3-methylpentane (3MeP) and methanol (MeOH), at 77 K, are also shown in Table 3.

Triplet Properties. Upon laser excitation, $T_1 \rightarrow T_n$ transitions of FT and its hydroxy derivatives were detected, mainly in the 300−550 nm region, just after the laser pulse. The lowest triplet state decayed by first-order kinetics and was quenched efficiently by molecular oxygen ($k_{ox} = 2.5-5 \times 10^9$ M^{-1} s^{-1}) and by β-carotene (low-energy triplet acceptor) to produce singlet oxygen and the T_1 of β-carotene, respectively. Furthermore, the lowest triplet states of FT and hydroxyflavothiones were sensitized by benzophenone, a well-known high-energy triplet donor.

The spectra of three representative hydroxy cases (FT, 3-OHFT, and 6-OHFT) are shown in Figure 4. Since the absorptions of the triplet and of the ground state are in the same spectral region, the triplet transient spectra are strongly affected by the overlap between the $T_1 \rightarrow T_n$ and $S_0 \rightarrow S_2$ absorption bands.

Generally, the lowest triplet state was detected by direct irradiation. Small absorptions due to nontriplet transients (practically unaffected by molecular oxygen) were detected after the triplet decay only for FT and for 5-OHFT in benzene; see further analysis in the Discussion section. Triplet−triplet absorption maxima, triplet lifetimes (τ_T), extinction coefficients for T−T absorptions (ϵ_T), and quantum yields of triplet formation (ϕ_T) in benzene, MeCN, EtOH, and TFE are given in Table 4.

Quenching experiments of the lowest triplet state of FT and 3-OHFT by i-ButOH were performed by recording the triplet lifetime in TFE. The linear plots reported in Figure 5 show the different behavior of the two triplets. In fact, T_1 of FT is quenched by i-ButOH with a quenching rate constant of 2.4×10^5 M^{-1} s^{-1}, while τ_T of 3-OHFT is practically uneffected by i- ButOH in a wide range of concentrations.

The ϕ_T values reported in the sixth column of Table 4 are generally obtained from energy-transfer experiments to β-carotene, while those of the seventh column are obtained via the singlet oxygen quantum yields measured in four solvents. With few exceptions, ϕ_T is close to unity.

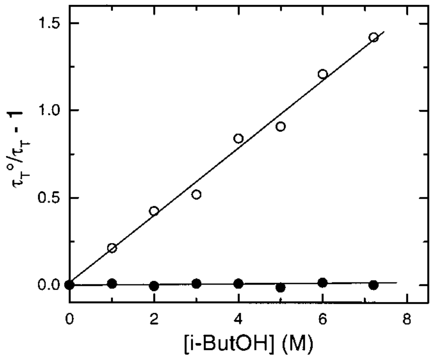

Figure 5. Linear plots for the quenching of triplet lifetimes of (○) FT and (●) 3-OHFT by iso-butanol in TFE.

TABLE 5: Quantum Yield of Singlet-Oxygen Photosensitization (ϕ_Δ) by Flavothiones in Benzene, Acetonitrile (MeCN), Trifluoroethanol (TFE), and Ethanol (EtOH) in Air-Equilibrated Solutions (λ_{exc} = 355 nm)

	ϕ_Δ			
compound	benzene	MeCN	TFE	EtOH
FT	0.64	0.44	0.44	0.14
3-OHFT	0.59	0.62	0.62	0.56
5-OHFT	0.04_5	<0.01	<0.01	<0.01
6-OHFT	0.61	0.48	0.48	0.15
7-OHFT	0.59	0.30	0.30	0.11
3,6-diOHFT	0.61	0.57	0.57	0.56
5,7-diOHFT	0.05	<0.01	<0.01	<0.01

The ϕ_T values of FT in EtOH, MeCN, and TFE (in the 0.90–0.95 range) were obtained from the product $\epsilon_T\phi_T$ and from the ϵ_T's measured by energy transfer from benzophenone (in the case of MeCN) or estimated as the mean values of the ϵ_T's in benzene and MeCN (in the cases of EtOH and TFE).

The triplet yields of the hydroxy derivatives are generally high and close to unity in benzene, except for 5-OHFT and 5,7-diOHFT. There appears to be a very small sensitization of β-carotene and oxygen by the latter compounds, but no oxygen quenchable signal could be clearly seen. Therefore, we estimate the amount of any triplet produced was ≤5%. The 5-OCH_3FT gave a parallel result.

Singlet-Oxygen Quantum Yields. The singlet oxygen yields are given in Table 5 for four different solvents. Note that for all of the hydroxy derivatives, except 5-OHFT and 5,7-diOHFT, the ϕ_Δ's in Bz are similar, ~0.6, and less than ϕ_T. As noted just above, essentially no singlet oxygen was produced either from 5-OHFT and 5,7-diOHFT or from 5-OCH_3FT. Also these values are about the same in acetonitrile and TFE as well, but all except 3-OHFT and 3,6-diOHFT decrease markedly in ETOH. We will consider the reasons for this in the Discussion section.

^{1}H NMR Spectra. Proton NMR spectra of 5-OHFT and 5,7-diOHFT in dimethyl sulfoxide show a sharp signal at 13.6 ppm. This shift at very low field is typical of a hydrogen-bonded hydroxyl proton. The hydroxyl proton for 3-OHFT and 3,6-diOHFT shows a signal at higher field (9.0 ppm), and for 6-OHFT and 7-OHFT no signal is observed.

Theoretical Calculations. The theoretical values of wavelengths and oscillator strengths of the relevant electronic transitions (the lowest five singlet states) for FT and the hydroxyflavothiones are given in Table 3. The contours of the molecular orbitals involved in the electronic transitions of FT are represented in Figure 6; the subscript H stands for the HOMO, H − 1 for next occupied orbital, etc., and L means LUMO, L + 1, the next unoccupied, etc. These contours are not greatly changed by hydroxyl substitution. The calculated values of energy of these molecular orbitals (before CI) are plotted in Figure 7.

Discussion

Singlet Excited States. As can be seen from Table 2, for FT, the energy of $S_1(n, \pi^*)$ with a maximum at 569 nm in benzene (ϵ_{max} = 52 M^{-1} cm^{-1}), and observed by others[3] with a maximum in the vicinity of 588 nm in 3-methylpentane, is well predicted by INDO/1 at 582 nm. The parent, chromothione, exhibits[2] a lowest $S_1(n, \pi^*)$ with a maximum at 561 nm in toluene (ϵ_{max} ~20 M^{-1} cm^{-1}). In any case, for FT the lowest singlet state is (n, π^*). The increase of solvent polarity, polarizability, or hydrogen-donating ability strongly blue-shifts the transition, from 588 nm in 3-MP to 569 nm in benzene and to 508 nm in methanol.

Hydroxyl substitution has a marked influence on the energy of the first three singlet excited states of flavothiones. Generally, it is expected that conjugative-type substitution as OH will blue-shift $S(n, \pi^*)$ transitions and red-shift $S(\pi, \pi^*)$ transitions,[15] and this trend is in fact theoretically predicted for all but the 5-hydroxy- and the 5,7-dihydroxy-substituted flavothiones (Table 2). For both compounds, the $S_2(\pi, \pi^*)$ state is theoretically predicted to be slightly blue-shifted and the $S_1(n, \pi^*)$ state is theoretically predicted to be strongly red-shifted with respect to the unsubstituted flavothione (Table 2). Note the quite acceptable agreement of theory with experiment with respect to these differences (Table 2), namely, the $S_2(\pi, \pi^*)$ state is experimentally red-shifted with respect to FT, for all compounds, except for 5,7-diOHFT where it is blue-shifted. The reason for the exceptional behavior of 5-OHFT and 5,7-diOHFT clearly emerges from observation of Figure 7, where the theoretical energies of the molecular orbitals are plotted: the n molecular orbital (basically a linear combination of the nonbonding orbitals of sulfur) is raised in energy (becomes the HOMO, n_H), and the π is lowered (becomes HOMO − 1, π_{H-1}) in the cases of the 5-hydroxy-substituted compounds. Since the S_2 state is 88% the $\pi_{H-1} \rightarrow \pi_L^*$ configuration and S_1 is 77% the $n_H \rightarrow \pi_L^*$, the $S_1(n, \pi^*)$ is lowered in energy (red-shifted) and the $S_2(\pi, \pi^*)$ is raised (blue-shifted).

The influence of the position of substitution on the extent of energy lowering of the $S_2(\pi, \pi^*)$ and $S_3(\pi, \pi^*)$ states for the remaining compounds is also understood after observation of the contour orbitals in Figure 6. The $S_2(\pi, \pi^*)$ state in FT is a slightly perturbed $\pi_H \rightarrow \pi_L^*$ configuration (Table 2), and the π_H molecular orbital has its largest electron density on carbon 3 (Figure 6). Consequently, the largest bathochromic shifts of the S_2 state are induced by 3-hydroxy substitution.

The $n \rightarrow \pi^*$ transitions are observable as the lowest energy transition in three thiones (FT, 6-OHFT, and 7-OHFT) as well as in 5-OCH_3FT, as noted earlier. In the cases of 3-OHFT and 3,6-diOHFT, even though their oscillator strengths are as high as any of the other four, the $S_0 \rightarrow S_2$ transition is more red-shifted than any of the others and it is very likely that the weak $n \rightarrow \pi^*$ transition is lost in the tail of the much stronger $S_0 \rightarrow S_2$ transition.

Also recall that fluorescence was observed only for the same three thiones (FT, 6-OHFT, and 7-OHFT). The $S_2 - S_1$ energy gap for these is in the 7000–8200 cm^{-1} region, while for 3-hydroxy and 3,6-dihydroxy it is in the 4500–5100 cm^{-1} range. It is likely in the latter two cases that vibrational relaxation is totally or largely competitive with fluorescence from S_2 ($k_{nr} \gg 1 \times 10^{11}$ s^{-1}). In the cases of the 5-hydroxy-substituted flavothiones, the $S_2 - S_1$ energy gap is as great as for the

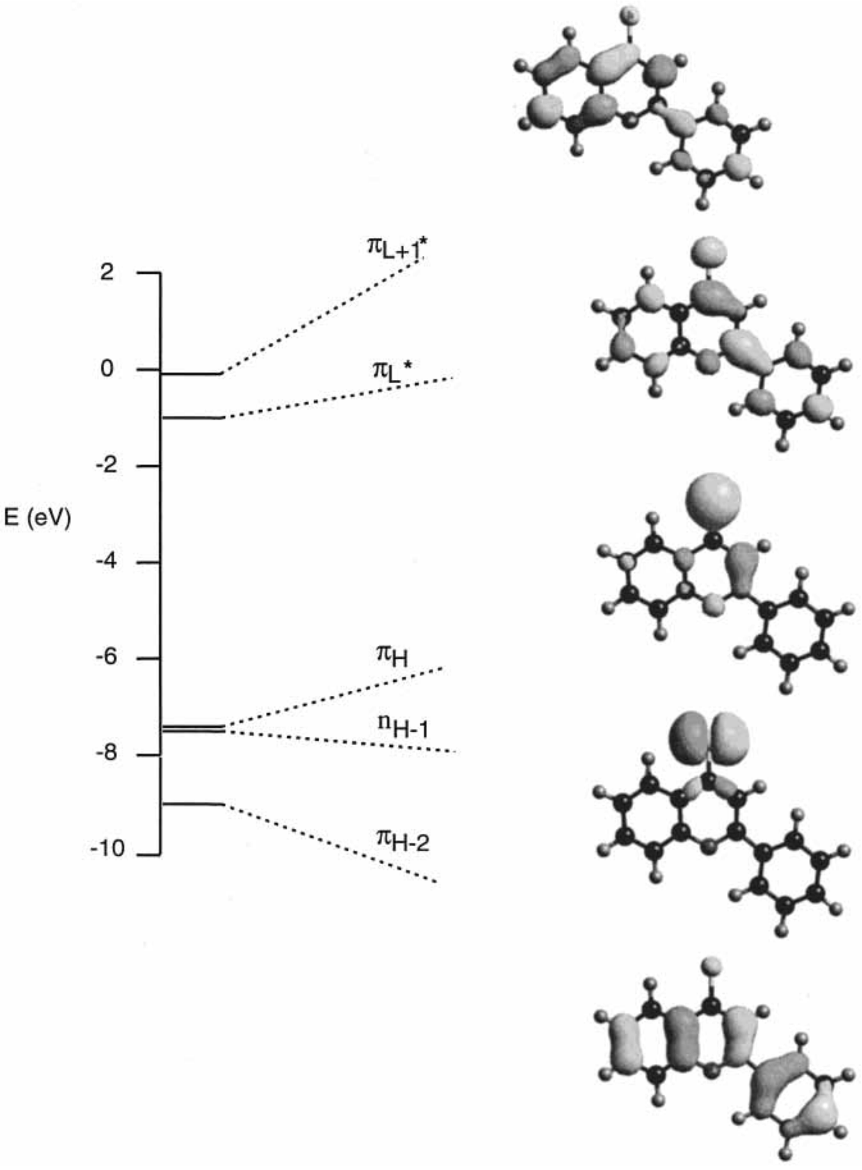

Figure 6. Frontier molecular orbitals of FT calculated by INDO/1.

compounds that fluoresce, but we believe that these may show intramolecular proton transfer (see later discussion), which is known to be fast and dominant over any fluorescence emission process from the original molecule.

Triplet States. On the basis of the triplet lifetimes, the hydroxy compounds can be divided into three groups: *group 1* (FT, 6-OHFT, and 7-OHFT), having a τ_T shorter than 2 μs in benzene, which is further markedly reduced to less than 100 ns in EtOH, *group 2* (3-OHFT and 3,6-diOHFT), with τ_T longer than 6 μs even in EtOH, and group 3 (5-OHFT and 5,7-diOHFT), showing essentially no triplet-state occupation.

Group 1. First, in the case of FT the spectra in benzene, EtOH, MeCN, and TFE all look similar (Figure 4). In benzene, there is one first-order decay of 1 μs, which is similar to those in MeCN and TFE. However, in EtOH, while the decay at 480 nm is single exponential with a decay time equal to 0.04 μs, the 350 nm (decay) and 400 nm (recovery) regions are double exponential: a common decay time of 0.04 μs and a longer one (40 μs at 350 nm and 200 μs at 400 nm). On the basis of the well-known H-atom abstraction capability of thiones in alcohols, but not in benzene, MeCN, or TFE, the single lifetime in the latter three solvents is assigned as that of the nonreacting triplet (not including ground-state quenching). The decay time in EtOH is assigned as essentially the reciprocal rate constant for hydrogen-atom abstraction from EtOH by the FT triplet to produce the FT−H$^\bullet$ radical. All the foregoing is further verified by the fact that the decay time of FT in TFE is strongly quenched by i-ButOH (see Figure 5). The 0.04 μs decay time is unaffected by the presence of oxygen, while the other component at 350 nm (decay time of 40 μs) is shortened to 6 μs. Additional information to be published elsewhere utilizing steady-state photochemical data obtained in several solvents[16] clearly indicate that the foregoing result is a consequence of the fact that the FT−H$^\bullet$ radical reacts with oxygen to form the flavone. The longer 200 μs decay time, seen as a component in the recover (at 400 nm), could then be the result of reforming the ground state via radical−radical reactions outside the solvent cage. On the basis of all of the foregoing, the lowest triplet state is assigned as n, π^*.

In the case of 6-OHFT and 7-OHFT, the decay and recovery decay times in benzene are the same (∼1.6 μs) and essentially the same as FT. In EtOH, the foregoing are shortened to ∼0.1 μs, quite similar to that of FT in EtOH. On the basis of the parallel behavior *and* lifetimes of 6-OHFT and 7-OHFT compared to FT in the same solvents, we assign the lowest triplet state of these two compounds to be n, π^*. An appropriate

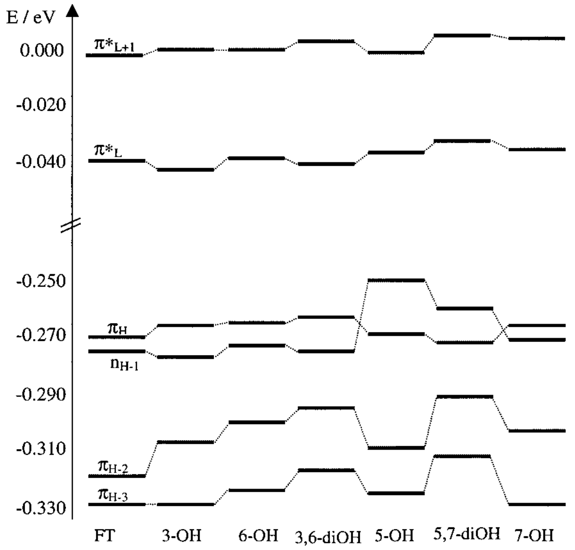

Figure 7. Energies before configuration interaction of the frontier molecular orbitals in the series of hydroxy-substituted flavothione.

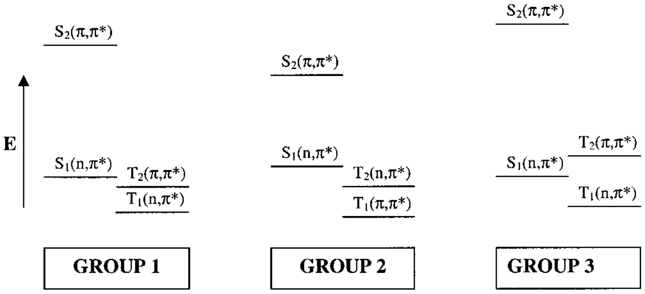

Figure 8. Schematic energy level diagram for the ordering of the two lowest excited singlet and triplet states for group 1, 2, and 3 type compounds (see text for components of each group).

schematic energy level diagram for group 1 compounds, based on calculations, absorption, flash, and phosphorescence data, as well as the S(n, π*) − T(n, π*) [~1000 cm^{-1}] and S(π, π*) − T(π, π*) [~8200 cm^{-1}] energy separations determined from FT and 3-methoxyflavothione respectively, appears in Figure 8.

Group 2. In these cases (the 3-OH substituted cases), the lifetimes of decay and recovery in benzene are 7 and 12 μs, very much longer (7−12-fold) than for the compounds in group 1. Furthermore, in EtOH, the lifetimes are essentially the same (6 and 9 μs). Thus, no quenching process is present; that is, it is clear that H-abstraction does not occur. Therefore, we assign the transient as a triplet and the lowest triplet state as π, π* based on the longer lifetimes and lack of H-atom abstraction in EtOH. Since recovery is not quite to the baseline, some small amount of net photochemistry does occur. An appropriate schematic energy level diagram based on parallel information as obtained for group 1 is given in Figure 8.

Group 3. For the compounds in this group, essentially no triplet transients were detected upon laser excitation in any solvent. At first we believed that the reason for this result for the two hydroxy compounds was an intramolecular proton transfer in the singlet manifold competing with triplet formation. With this occurring, which is typically fast ($k \geq 10^{10}$ s^{-1}), this process would compete with k_{ISC} and very significantly lower ϕ_T, as is observed. Recall that ground-state H-bonding occurs, as determined by NMR, and fluorescence is absent. Also no phosphorescence is observed for the 5-OH-substituted cases. To attempt to verify this idea, we synthesized the 5-OCH$_3$FT. However, as is the case for 5-OHFT, essentially no triplet was observed and no phosphorescence was observed. Thus, the two situations look similar but, of course, no proton transfer can occur in the 5-OCH$_3$FT case. Therefore, there must be another reason for the results concerning the ϕ_T of the 5-OCH$_3$FT (very small or absent), and that must originate from the relative relationship of the energy locations of the lowest two excited singlet and triplet states; see below.

An appropriate energy level diagram based on parallel information obtained for group 3 is given in Figure 8. Note that intersystem crossing from either S$_1$(n, π*) to T$_1$(n, π*) or S$_2$(π, π*) to T$_2$(π, π*) is configurationally forbidden.[17] Moreover, the energy separation of S$_1$ and S$_2$ is large (~10 000 cm^{-1}), and any second-order contribution via vibronic coupling would be negligible. A parallel energy level diagram would also be appropriate for the 5-hydroxy- and 5,7-dihydroxy compounds.

Finally, recall that fluorescence is observed from 5-OCH$_3$-FT but not from either of the two hydroxy derivatives. We believe that this could originate from a fast proton transfer from −OH to the S atom in the hydroxy-substituted cases, which could quench any potential fluorescence.

Mechanisms of Intersystem Crossing. For group 1 compounds (FT, 6-OHFT, and 7-OHFT), the lowest excited singlet and triplet states are n, π*. The mechanism of intersystem crossing is then proposed to be

$$S_1(n, \pi^*) \xrightarrow{SO} T_2(\pi, \pi^*) \xrightarrow{vib} T_1(n, \pi^*)$$

where SO signifies occurrence via the spin−orbital coupling operator and vib signifies occurrence via the vibronic operator.

For group 2 compounds (3-OHFT and 3,6-diOHFT), clear observation of an $n \rightarrow \pi^*$ transition was not possible. Nonetheless, theory (Table 2) predicts the lowest singlet as n, π*. We, perhaps, can have more faith than usual in the calculation since the predicted and experimental values for FT and 6-OHFT (and possibly 7-OHFT) are in very good agreement. Thus, we assign the lowest energy singlet as n, π*, and the lowest energy triplet is π, π*, vide infra. Thus, the mechanism proposed for intersystem crossing is

$$S_1(n, \pi^*) \xrightarrow{SO} T_1(\pi, \pi^*)$$

For group 3 recall that essentially no intersystem crossing occurs.

Singlet-Oxygen Yields. All of the flavothiones in benzene, acetonitrile, and trifluoroethanol show about the same ϕ_Δ (0.6) except for 5-OHFT and 5,7-diOHFT. In ETOH, however, the group 1 compounds undergo a marked decrease in ϕ_Δ. We have assigned the lowest triplet state in these compounds as n, π*. In the 3-substituted compounds (group 2) it is π, π*. Therefore, the reason for the foregoing changes in ϕ_Δ in ETOH for compounds in group 1 is that a competitive H-atom abstraction reaction occurs from T(n, π*), simultaneously with energy transfer to oxygen, thus lowering the ϕ_Δ (Table 5) and shortening the τ_T (Table 4). For 3-OHFT and 3,6-diOHFT, since the lowest triplet state is π, π*, relatively insignificant H-atom abstraction is expected in general, and therefore, the ϕ_Δ remains high even in ETOH, comparable to the three other solvents (Table 5), and τ_T is not shortened (Table 4).

Conclusions

The addition of OH groups to FT generally causes varying degrees of red-shift of the first strong absorption band, the S$_0$

→ S_2 transition, with 3,6-diOH substitution causing the largest red-shift. The exception is 5,7-diOHFT.

The position of OH substitution and the number of OH groups added can cause some dramatic differences in photophysical behavior. These are caused by the relative state order changes of the two lowest excited singlet and triplet states.

The 6-OH or 7-OH substitution (group 1) on FT does not change the nature of the lowest triplet state (n, π^*) or its lifetime compared with FT. In all solvents these have a high ϕ_T (0.8−1, including FT). The ϕ_Δ is generally in the region of ∼0.6 in benzene and decreases somewhat to 0.5 for 6-OH and still lowers for 7-OH (0.3) in MeCN and TFE. There is a large decrease in ϕ_Δ in ETOH, as there is for τ_T, and these are caused by an H-atom abstraction reaction from the solvent (triplet n, π^* state lowest) competing with energy transfer to oxygen.

The 3-OH or 3,6-diOH substitution (group 2) results in a change in the configurational nature of the lowest triplet state from n, π^* (FT, 6-OHFT, and 7-OHFT, group 1) to π, π^*. Also, ϕ_T does not change from ∼1. Moreover, ϕ_Δ is ∼0.6 (as for FT, 6-OHFT, and 7-OHFT) in all solvents *including* ETOH, which is in marked contrast to the large decrease found for FT, 6-OHFT, and 7-OHFT. The latter result is explained by the fact that there is essentially no H-atom abstraction reaction competing with energy transfer to oxygen for the 3-substituted cases because of the change in the configurational origin of the lowest triplet state.

The 5-OH or 5,7-diOH substitution results in a very marked change in the photophysical (and ϕ_Δ) properties compared to all other hydroxy substitutions. Essentially no triplet occupation is observed by direct excitation, and ϕ_Δ is essentially zero in all solvents (four considered). Also, no fluorescence or phosphorescence are observed. The 5-methoxy compound also showed parallel behavior but did exhibit a weak fluorescence. The results are understandable on the basis of the energy relationship among the two lowest singlet and triplet states of n, π^* and π, π^* character, as well some proton transfer from S_2 for the hydroxy derivatives.

It is highly probable that the 5-OH and 5,7-diOH compounds would not be good photosensitizers since they show no triplet-state occupation and a quenched excited singlet-state behavior. On the other hand, the best compounds as potential photosensitizers via a singlet-oxygen mechanism (or possibly an electron-transfer process as well) would be those of group 2, the 3-OH and 3,6-diOH. The group 1 compounds could also be potential photosensitizers via a H-atom extraction mechanism or act via formation of singlet oxygen in an environment where H-atom abstraction was relatively noncompetitive (as in a benzene-like, acetonitrile-like, or perhaps a water environment).

Acknowledgment. This work was supported by the Fundação para o Desenvolvimento da Ciência e Tecnologia (FCT), Portugal, Grant PRAXIS/2/2.1/QUI/324/94. R.S.B. acknowledges the G.G.P. XXI/BCC/3638/96 grant, J.C.L. and I.A. are grateful for the PRAXIS 4/4.1/BPD/3410 and PRAXIS 4/4.1/BD/3589 grants, respectively. F.E., F.O., and G.G.A. are thankful for the grant from the Italian Consiglio Nazionale delle Ricerche and for the support of the National Project by the Ministero dell'Università e della Ricerca Scientifica e Tecnologica and the University of Perugia.

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