Triplet state properties of flavone in homogeneous and micellar solutions. A laser flash photolysis study

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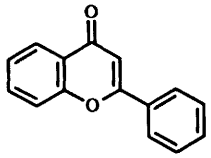
The triplet state properties of flavone (2-phenylchromone) have been investigated by laser flash photolysis. Excitation at 337 nm was carried out by means of a nitrogen laser. The T–T absorption spectrum was determined in homogeneous solvents (benzene, ethanol, acetonitrile) and in micellar solutions of SDS, CTAC and Brij 35. The position of the two maxima at 370 and 620 nm was practically independent of the solvent. The self-quenching rate constants were determined in homogeneous solvents and are in the range $1.8–3.0 \times 10^8$ dm^3 mol^{-1} s^{-1}. The effect of surfactant concentration on the yield and decay of the triplet was investigated. The self-quenching was greatly diminished in the micellar systems. The results could be explained by considering the flavone as totally micellized. The quenching by amines was also studied in the three solvents. The rate constants correlate with the oxidation potential of the amine and a new long-lived transient appears which was attributed to the flavone ketyl radical.

Flavonoids are derivatives of flavone (2-phenylchromone) carrying from two to six hydroxy or alkoxy substituents. The study of the excited state properties of flavonoids has been of great concern during recent years.[1] One of the several reasons for this interest is the hypothesis that flavonoids are involved in the protective mechanism against UV damage in some plants.[2] From the photophysical point of view the major interest is in the intramolecular proton transfer reactions taking place in the excited state. Undoubtedly these processes are related to the photobiological activity of flavonoids.

A large number of papers have been published recently dealing with the photophysics and photochemistry of flavonoids.[3] However the study of flavone itself (**I**) has received much less attention.



I

The photorearrangements of flavone and other related carbonyl compounds have been known for a long time.[4] Irradiation of flavone in the presence of sodium sulfite leads to dimerization of the starting compounds.[5] Hamanoue *et al.* reported that photolysis of flavone affords a cleavage product irrespective of the solvent polarity.[6] More recently, Monici *et al.*[1] investigated the photochemistry of flavone in organic solvents and in micellar systems. The photochemical behaviour was found to be strongly dependent upon the microenvironment. Most of these studies were aimed at the characterization of products under different experimental conditions and less emphasis was given to the mechanistic aspects of the photoreactions. In most cases the flavone triplet state was thought to be involved. The direct observation of the triplet state by laser flash photolysis was reported by Hamanoue *et al.*[6] The T–T absorption spectrum shows bands at 365–370 and 640–650 nm. The exact position of the maximum depends on the solvent. From the solvent polarity effect on the lifetime, a $\pi\pi^*$ character was assigned to the lowest triplet state. The photoreduction by different hydrogen donors was also investigated by Sakaguchi *et al.*[7] in propan-2-ol with the aid of the CIDEP technique. It was proposed that the triplet state reacts with the hydrogen donors forming the flavone ketyl radical when the donor is xanthene of *N*,*N*-diethylaniline, while a benzyl-type radical is formed when tributyltin hydride is employed.

The intersystem crossing of flavone was studied by picosecond absorption spectroscopy and it was found to be a very fast process.[6] It is in accord with the high triplet quantum yield [0.9 (± 20%)] in benzene at room temperature[8] and also with the lack of fluorescence in fluid solution found for chromone, the basic ring of flavone.[9] The reactivity of the triplet towards several substrates was determined by laser flash photolysis in benzene solution at room temperature.[8]

In this paper we present a study of the triplet state properties in polar solvents (ethanol and acetonitrile), a non-polar solvent (benzene) and in micellar solutions by laser flash photolysis. The medium effect on the photoreduction of the triplet state by amines is also investigated.

Experimental

The solvents benzene and acetonitrile (MeCN) were Sintorgan HPLC grade and were used without further purification. Ethanol (EtOH) was distilled twice before use. Flavone was from Sigma and was used as received. The amines (BA = butylamine, i-PA = isopropylamine, DNPA = dipropylamine, DIBA = diisobutylamine, TPA = tripropylamine, TBA = tributylamine, TEOHA = triethanolamine, DMA = *N*,*N*-dimethylaniline) were from Aldrich and they were fractional distilled before use. Cetyltrimethylammonium chloride (CTAC) (Kodak) and sodium dodecyl sulfate (SDS) (BDH) were purified by recrystallization.

For transient absorption determinations a nitrogen laser (Laseroptics), 5 ns FWHM and 5 mJ per pulse, was employed. The laser flash photolysis set-up has been described previously.[10] The signal was acquired by a digitizing scope where it was averaged and then transferred to a computer. The samples were deoxygenated by nitrogen bubbling for at least 15 min. All measurements were carried out at room temperature. The triplet state of ZnTPP (zinc tetraphenylporphyrin) in benzene was used as an actinometer. The triplet yield of ZnTPP was measured at 470 nm immediately after the laser pulse. Values of 7.3×10^4 dm^3 mol^{-1} cm^{-1} and 0.83 were used for ε^T and φ^T ZnTPP, respectively.[11]

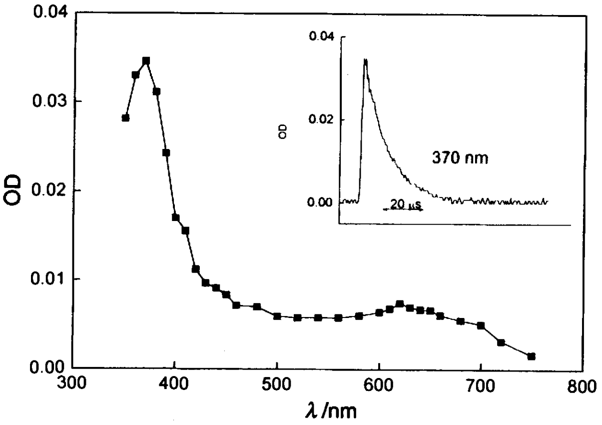


Fig. 1 Transient absorption spectrum of flavone (1×10^{-3} mol dm^{-3}) in benzene immediately after laser excitation at 337 nm. Inset: decay at 370 nm.

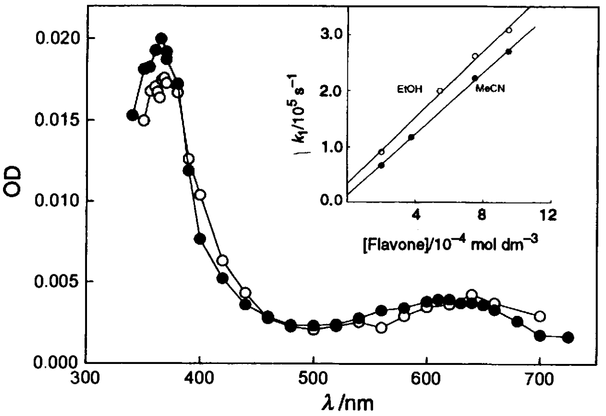


Fig. 2 Transient absorption spectrum of flavone in EtOH (○) and in MeCN (●) taken at 3 µs after the laser pulse. Inset: first-order decay rate constant *vs.* flavone concentration.

Results and discussion

Triplet state in homogeneous solutions

The flavone absorption spectrum in benzene presents a maximum at 292 nm and a shoulder at 310 nm. In the polar solvents, EtOH and MeCN, the maximum remains at the same position while the shoulder is much less noticeable. When flavone is laser flash irradiated at 337 nm in the long-wavelength tail of the absorption spectrum a transient absorption is observed in the UV–VIS region. The transient absorption spectrum in benzene is shown in Fig. 1. In Fig. 2 the spectra are presented in ethanol and acetonitrile. It can be seen that the spectra are practically the same in all the three solvents. It presents a strong peak at 370 nm and a broad band with a maximum at 620–630 nm. These bands coincide with those previously assigned to the triplet state of flavone in the literature.[12] At both wavelengths the absorption decays in a few microseconds with similar first-order kinetics (inset in Fig. 1). The apparent first-order rate constant, k_1, increases with flavone concentration. A plot of k_1 *vs.* concentration can be seen as an inset in Fig. 2. A similar plot is obtained in benzene. From the slope, the second-order rate constant for self-quenching can be obtained and they are collected in Table 1.

The spectrum and the self-quenching rate constant in benzene are in good agreement with those previously reported by Bhattacharyya *et al.*[8]

Table 1 Self-quenching rate constant for flavone triplet

Solvent	$k/10^8$ dm^3 mol^{-1} s^{-1}
Benzene	1.83
Ethanol	2.96
Acetonitrile	2.75

From the initial slope of the plot of the triplet absorbance at 370 nm *versus* the laser intensity, the product $\Phi^T\varepsilon^T$ was determined in benzene, where Φ^T is the intersystem crossing quantum yield and ε^T is the triplet extinction coefficient at 370 nm. Using ZnTPP in benzene as an actinometer, a value of 11 000 ± 1000 dm^3 mol^{-1} cm^{-1} was obtained for $\Phi^T\varepsilon^T$. In order to estimate separately Φ^T and ε^T the energy transfer technique was employed[12] with naphthalene as an energy acceptor. The triplet of flavone is quenched by naphthalene with a rate constant of 3.3×10^9 dm^3 mol^{-1} s^{-1}, which is close to the diffusional limit in benzene, 5.0×10^9 dm^3 mol^{-1} s^{-1}. At the same time as the flavone triplet is being quenched, the typical T–T absorption spectrum of naphthalene replaces the flavone triplet spectrum. From the absorbance at 425 nm, the maximum of the naphthalene triplet, at a naphthalene concentration in which all flavone triplets are quenched, the extinction coefficient at 370 nm, ε_{370}, can be estimated by eqn. (1), where

$$\varepsilon_{370} = \varepsilon_{425}\frac{(OD_{370})}{(OD_{425})} \quad (1)$$

ε_{425} is the molar extinction coefficient of triplet naphthalene at 425 nm in benzene, taken[12] as 13 200 dm^3 mol^{-1} cm^{-1}. OD_{370} is the absorbance of triplet flavone measured immediately after the laser pulse in the absence of naphthalene and OD_{425} is the absorbance of triplet naphthalene measured at a time such that all flavone triplets have been quenched and naphthalene has not begun to decay. In this way ε_{370} was estimated as 11 000 ± 1000 dm^3 mol^{-1} cm^{-1}. Considering the several approximations involved in the method, this can be taken as evidence that the triplet quantum yield of flavone in benzene is near unity. Moreover, both the extinction coefficient and the intersystem crossing quantum yield are within the experimental uncertainty of values determined by pulse radiolysis and energy transfer from benzophenone.[8] In EtOH and MeCN the absorbance at 370 nm immediately after the laser pulse is nearly the same as that for benzene solutions of matched absorbances at 337 nm. Since the spectra are also very similar it can be assumed that the extinction coefficients are alike, and therefore the triplet yield is most likely near unity in the polar solvents.

Triplet state in micellar solutions

Both in SDS and CTAC solutions the transient absorption spectra are practically unchanged from those in homogeneous solvents. However, the triplet lifetimes are at least one order of magnitude longer in micellar solutions (Fig. 3). The longer decay in surfactant solutions may be understood by a compartmentalization effect that inhibits the self-quenching process. In fact the lifetime increases with increasing detergent concentration, both in SDS and CTAC solution. On the other hand, in neutral micellar solution, Brij 35, the lifetimes are only a factor of two longer than in homogeneous media. The triplet yield also increases with the detergent concentration. Both these effects can be seen in Fig. 4 for the case of CTAC. The micellar effect on the triplet yield and lifetime may be explained by the distribution of flavone molecules in the micelles. It is assumed that all the flavone is in the micellar phase and that for those micelles with occupation number greater than one, the intramicellar self-quenching is so fast that no triplet is observed

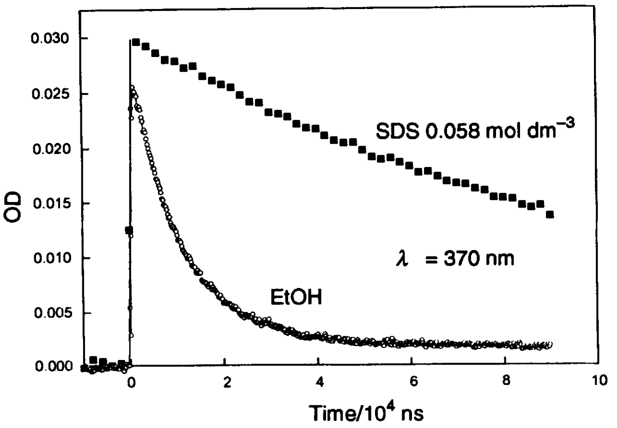


Fig. 3 Triplet decay at 370 nm in EtOH and in SDS micellar solution

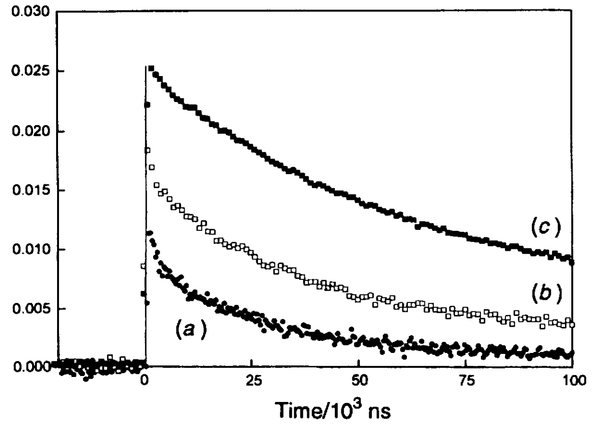


Fig. 4 Effect of CTAC concentration on the triplet yield and decay measured at 370 nm. [CTAC] = 0.015 (■), 0.025 (□) and 0.068 (●) mol dm^{-3}.

in our experimental conditions. This is equivalent to a static quenching of the triplet yield that may be expressed by a relation similar to that for the static fluorescence quenching in micelles,[13] [eqn. (2)] where OD is the observed absorbance of

$$\ln\frac{OD_0}{OD} = \frac{[\mathrm{Fl}]}{[\mathrm{Det}] - cmc} \times N_{ag} \qquad (2)$$

the triplet, OD_0 is the corresponding quantity extrapolated to zero detergent concentration, [Fl] is the flavone concentration and N_{ag} is the micelle's aggregation number. A similar equation was employed by Selwin and Scaiano[14] for the static quenching of the phenanthrene triplet yield by octa-1,3-diene in SDS. A plot according to eqn. (2) for the three surfactants investigated here is shown in Fig. 5. From the slopes, aggregation numbers were determined and they are collected in Table 2. In the case of Brij 35 and SDS there is a reasonable agreement, considering the dispersion of the data, with the values reported in the literature.[13] On the other hand, the result for CTAC is nearly one half of that commonly accepted. This discrepancy may be due to some alteration of the micellar structure caused by the presence of flavone, but in the particular case of CTAC considerable disparity exists in the reported values of the aggregation number. Thus, while numbers in the range 80–115 have been reported,[15] a value of 59 was found by EPR by Kwan *et al.*[16] These results show that the measurement of the triplet yield of totally micellized probes, with an efficient self-quenching process, can be used as a method for determining aggregation numbers.

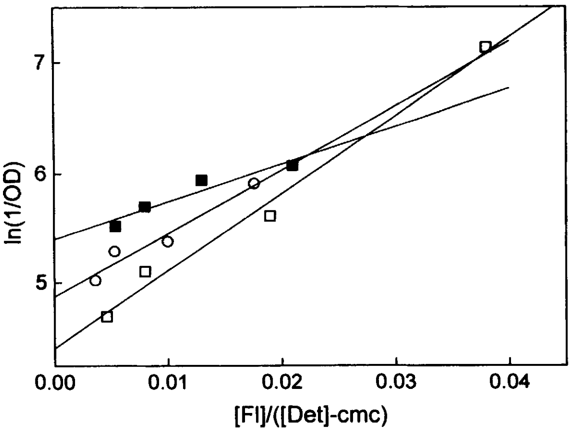


Fig. 5 Plot of the triplet absorption extrapolated at time zero, according to eqn. (2). (■) Brij 35, (○) CTAC, (□) SDS.

The triplet decay as a function of detergent concentration may be explained by a mechanism similar to that proposed for the quenching of triplet states in micellar solutions.[13,14] Flavone is considered to be in equilibrium between the aqueous phase and micelles, and the self-quenching involves a second flavone molecule entering a micelle containing one triplet flavone. With these assumptions the following mechanism can be written where the subscript m or w refers to the flavone

$$\mathrm{Fl_m} \underset{k_+}{\overset{k_-}{\rightleftharpoons}} \mathrm{Fl_w}$$

$$\mathrm{Fl_m} \xrightarrow{h\nu} {}^1\mathrm{Fl*_m} \longrightarrow {}^3\mathrm{Fl*_m}$$

$${}^3\mathrm{Fl*_m} \xrightarrow{k_o} \mathrm{Fl_m}$$

$${}^3\mathrm{Fl*_m} + \mathrm{Fl_w} \xrightarrow{k_+} \text{quenching}$$

incorporated into the micelles or solubilized in water, respectively. The lifetimes as a function of micelle concentration may be written as eqn. (3) where α is the efficiency of the quenching process.

$$\tau^{-1} = k_{obs} = k_o + \alpha k_+[\mathrm{Fl_w}] \qquad (3)$$

The partition constant is given by eqn. (4) where [M] is the

$$K_{eq} = \frac{k_+}{k_-} = \frac{[\mathrm{Fl_m}]}{[\mathrm{Fl_w}][\mathrm{M}]} \qquad (4)$$

concentration of micelles. Since the equilibrium is heavily displaced to the micellar phase, *i.e.* $[\mathrm{Fl_m}] \gg [\mathrm{Fl_w}]$, eqn. (3) may be written as eqn. (5) with $[\mathrm{Fl_T}] = [\mathrm{Fl_m}] + [\mathrm{Fl_w}]$.

$$k_{obs} = k_o + \alpha k_-\frac{[\mathrm{Fl_m}]}{[\mathrm{M}]} \approx k_o + \alpha k_-\frac{[\mathrm{Fl_T}]}{[\mathrm{M}]} \qquad (5)$$

A plot according to eqn. (5) can be seen in Fig. 6. From the slopes, values of k_- may be obtained. The values are collected in Table 2. The values for SDS and CTAC agree with those reported in the literature[13] if a value of 0.01 is taken for α, assumed to be of the same order of magnitude as the ratio of the experimental self-quenching rate constant to the diffusional limit in the homogeneous solvents. It can be seen that the exit rate constant is much higher for Brij 35 micelles. This may be related to the smaller size of the micelles in this case.

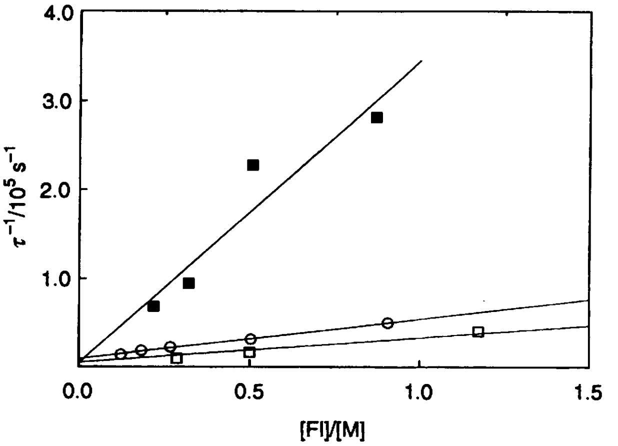


Fig. 6 Experimental first-order decay constants as a function of the ratio of flavone and micelles concentrations. (■) Brij 35, (○) CTAC, (□) SDS.

Table 2 Aggregation number and exit rate constants as determined by self-quenching of triplet flavone

Surfactant	N_{ag}	$\alpha k_{-}/10^4$ s^{-1}
SDS	71	2.8
CTAC	58	4.5
Brij 35	34	34

Table 3 Rate constants for the triplet quenching of flavone amines

		dm^3 mol^{-1} s^{-1}		
Amine	$E_{\frac{1}{2}}$/V[a]	Benzene	EtOH	MeCN
n-BA	1.3	3.1×10^6	—	–
i-PA	1.3	1.3×10^6	2.3×10^7	6.7×10^7
DNPA	0.93	2.7×10^8	2.2×10^8	8.3×10^8
DIBA	0.87	4.9×10^8	2.9×10^8	8.3×10^8
TPA	0.64	3.8×10^8	1.1×10^9	8.4×10^8
TBA	0.62	4.0×10^8	1.1×10^9	1.7×10^9
DMA	0.48	3.4×10^9	3.2×10^9	2.6×10^9

[a] $E_{\frac{1}{2}}$ *vs.* Ag/Ag$^+$ in MeCN from A. J. Bard and H. Lund, *Encyclopedia of the Electrochemistry of the Elements*, Marcel Dekker, New York, 1984, Organic Section, vol. XV.

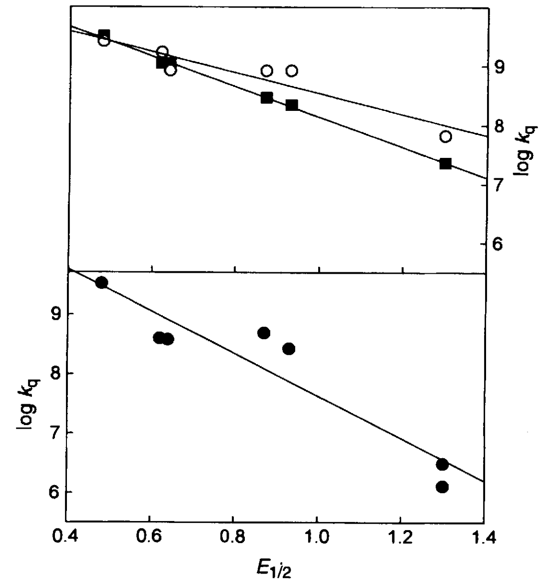


Fig. 7 Quenching rate constants *vs.* oxidation potential of aliphatic amines in EtOH (■), MeCN (○) and benzene (●)

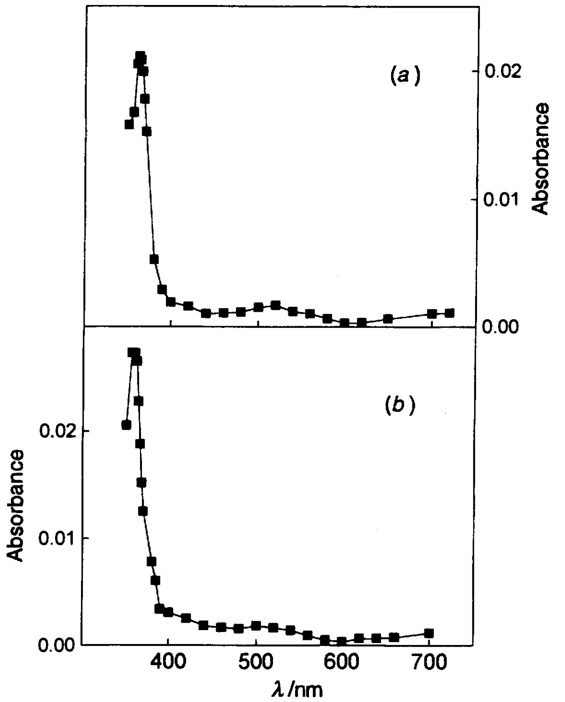
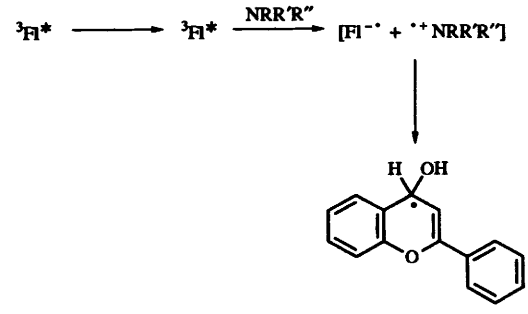


Fig. 8 Transient absorption spectra of flavone in the presence of aliphatic amines. (*a*) 5 × 10^{-4} mol dm^{-3} flavone, 0.14 mol dm^{-3} DNPA in benzene 2 μs after the laser pulse, (*b*) 4 × 10^{-4} mol dm^{-3} flavone, 2.4 × 10^{-4} mol dm^{-3} TPA in EtOH at 40 μs after the laser pulse.

Reductive quenching by amines

Amines are efficient quenchers of the triplet state of flavone. Bimolecular quenching rate constants were determined in MeCN, EtOH and benzene by monitoring the decay at 620 nm as a function of amine concentration. This wavelength was chosen in order to avoid interference by the absorption of the quenching product (*vide infra*). The results are collected in Table 3. The rate constants correlate with the oxidation potential of the amine (Fig. 7). Therefore the quenching process most probably involves a charge transfer process. At the same time as the quenching takes place, the triplet absorption spectrum is replaced by a longer lived transient, with a narrower band for the maximum at 365–370 nm, and a new low intensity band appears at *ca.* 520 nm, (Fig. 8). The same spectrum is obtained at high [Fig. 8(*a*)] or low amine concentration [Fig. 8(*b*)]. This new transient was assigned to the flavone ketyl radical on the following grounds. (*i*) The spectrum is the same for all the amines investigated. (*ii*) The same spectrum is obtained in benzene and in polar solvents. (*iii*) In benzene, where charge separation is most unlikely to be observed in our timescale, the transient species is not expected to be the radical anion of flavone. (*iv*) The same spectrum was reported for the product of the flavone triplet quenching by *p*-methoxyphenol and TEA in benzene solution.[8] The radical formed in the quenching by *N*,*N*-triethylaniline was characterized as the ketyl radical by Sakaguchi *et al.*[7] The radical is supposed to be formed by a fast in-cage proton transfer after the initial electron transfer step (Scheme 1). It is well known that this process occurs in the quenching of triplet carbonyls by aliphatic amines[17] although Mataga and co-workers[18] proposed a direct hydrogen transfer mechanism competing with the two step mechanism of Scheme 1. They conclude that the operation of one or another mechanism depends upon the amine structure.



Scheme 1

With the time resolution of our experiments it is difficult to discriminate between both mechanisms. However, the extremely good correlation observed, especially in ethanol, suggests that in the present case the charge transfer mechanism prevails.

In summary, upon irradiation of flavone in its long wavelength band, the triplet state is formed with a quantum yield of near unity, both in polar and in non-polar solvents. It suffers an efficient self-quenching process that is greatly reduced in micellar solutions. The effect of surfactants on the yield and decay of the triplet can be used to determine micellar parameters. The triplet is also quenched by amines, with the production of the flavone ketyl radical. The quenching rate constants are very similar in polar and in non-polar solvents.

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