FLUORESCENCE, PHOSPHORESCENCE AND BASICITY IN THE FIRST EXCITED TRIPLET OF 2-METHYLHARMINE AND HARMALINE

A. OLBA TORRENT, F. TOMAS VERT, I. ZABALA SANCHEZ and P. MEDINA CASAMAYOR

Departamento de Química Física, Facultad de Ciencias Químicas, Universidad de Valencia, Burjasot, Valencia (Spain)

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Summary

The fluorescence and phosphorescence spectra of 2-methylharmine and harmaline were investigated at 77 K. Two species, the cation and the neutral molecule, are characterized in both the fluorescence and the phosphorescence spectra depending on the solvent used. Fluorescence and phosphorescence spectra have also been recorded at room temperature for ionic species adsorbed on cellulose.

The pK_a values for the first excited triplet state equilibria have been calculated by applying the Förster–Weller relation. It is found that the nitrogen of the pyrrole ring of 2-methylharmine is more acid and the nitrogen of the pyridinic ring of harmaline is more basic in the first excited triplet than in the ground state.

1. Introduction

The molecules studied, 2-methylharmine and harmaline (Fig. 1), have been reported to be hallucinogens of considerable pharmacological interest [1 - 3].

The fluorescence of these derivatives in aqueous solutions at 25 °C has been reported for a wide range of pH values [4, 5]. The pK_a values for the acid–base equilibria (Fig. 2) on the first excited singlet state were reported from these studies.

It was established that the emission spectra are strongly dependent on the temperature and the phase in which the molecules are dissolved or dispersed. In the present work we have studied the emission spectra of the cationic (BH^+) and the neutral (B) species of 2-methylharmine and harmaline in solid phases at 77 K, and the basicity of these molecules in the first triplet state.

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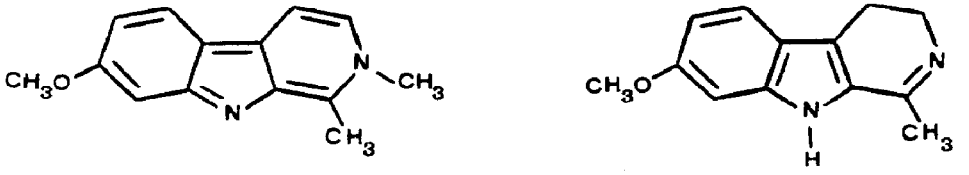
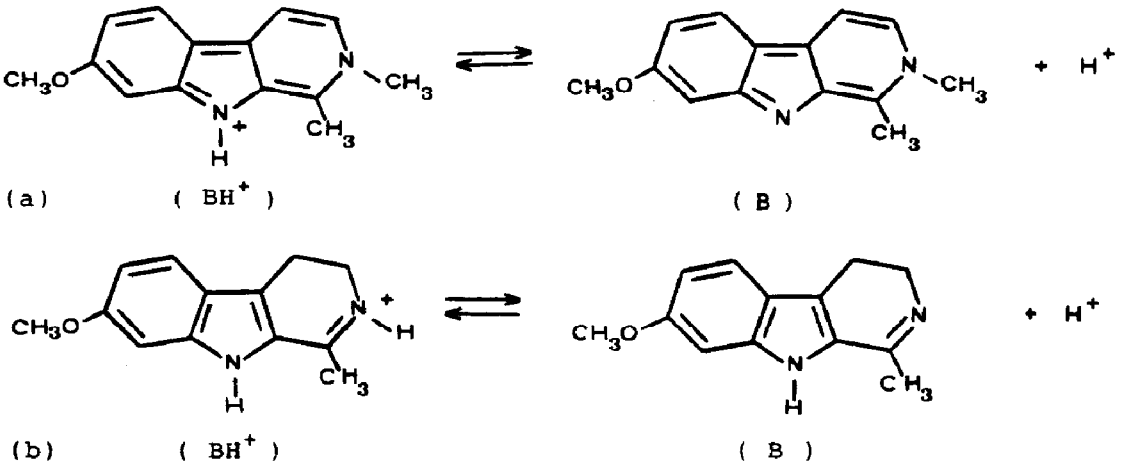


(a) (b)

Fig. 1. Molecular structures: (a) 2-methylharmine and (b) harmaline.



(a) (BH^+) (B) + H^+

(b) (BH^+) (B) + H^+

Fig. 2. Acid–base equilibria: (a) 2-methylharmine and (b) harmaline.

The pK_a values for the acid–base equilibria on the first triplet state (T_1) can be derived from the phosphorescence spectra of the acid (BH^+) and the basic (B) forms by means of the Förster–Weller method [6]. The values estimated in this way can indicate whether the basicity of the compounds studied lies near the basicity of the first excited singlet (S_1) or that of the ground state (S_0) and they can also indicate the influence of the site of protonation on the acid–base properties in the first excited triplet state.

It has been reported that the pK_a values of related pyridinic derivatives increase by 5 - 7 units on electronic excitation [7], and the acidity of pyrrole derivatives increases in the excited state [8].

2. Experimental details

2.1. Materials

Harmaline, harmaline hydrochloride and 2-methylharmine were obtained from the Sigma Chemical Company. The solvents used in these studies were 1:100 (by volume) H_2SO_4–ethanol and ethanol–NaOH (0.08 M) mixtures. Uvasol grade ethanol, analytical grade sulphuric acid and sodium hydroxide were obtained from Merck.

The concentrations of the samples were 10^{-4} M before freezing.

2.2. Apparatus

Uncorrected excitation, fluorescence and phosphorescence spectra were recorded using a Perkin–Elmer MPF-44A spectrofluorometer equipped with a phosphorescence accessory. The spectra at 77 K were recorded in transparent matrices produced by freezing the system to liquid nitrogen temperature. The emission for ionic species in the adsorbed phase at room temperature was recorded as described previously [9].

The absorption spectra at room temperature were recorded using a Cary 219 spectrophotometer.

All the experiments at room temperature were carried out using thermostatted solutions at a temperature of 25 ± 0.1 °C.

3. Results and discussion

3.1. Electronic absorption spectra

The absorption spectra of the H_2SO_4–ethanol solutions at room temperature were very similar to those recorded on aqueous media at pH values near 1 [4, 5] with small displacements to the red.

The ethanol–NaOH solutions show absorption spectra very similar to those obtained for aqueous solutions at pH 12.9 [4, 5]. This behaviour indicates that the molecules studied are in the BH^+ and the B forms in H_2SO_4–ethanol and ethanol–NaOH solutions respectively.

Harmaline is similar to the aromatic β-carbolines [4] in showing a hypsochromic shift corresponding to the transformation from the cation to the neutral species. However, this shift is bathochromic in 2-methylharmine because the protonation site is at the nitrogen in the pyrrole ring.

The absorption maxima for solutions of each species are summarized in Table 1.

TABLE 1

Frequencies of the lowest absorption maxima

	$\bar{\nu}_{max}(BH^+)$ (cm^{-1})	$\bar{\nu}_{max}(B)$ (cm^{-1})
2-Methylharmine	27472.5	24691.3
Harmaline	26455.0	29940.1

3.2. Fluorescence spectra

The fluorescence spectra of the molecules studies are shown in Figs. 3 and 4. The spectra recorded for H_2SO_4–ethanol solutions correspond to the species BH^+ in both molecules. The spectra for ethanol–NaOH mixtures correspond to the neutral B species for 2-methylharmine and show the emission to the neutral B species and the cationic BH^+ species for harmaline. This

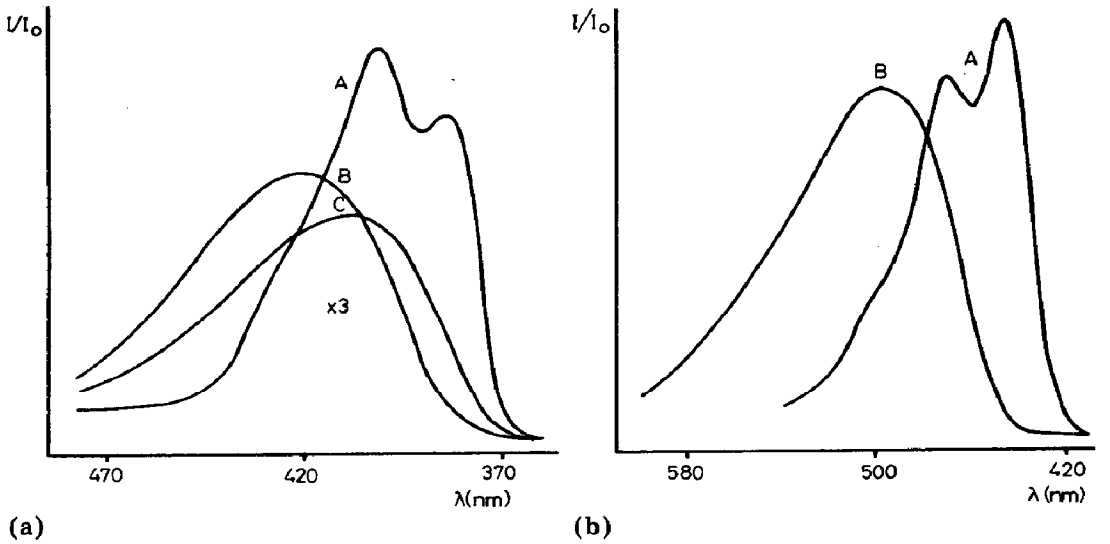


Fig. 3. Fluorescence spectra of 2-methylharmine. (a) In H_2SO_4–ethanol solutions: curve A, at 77 K; curve B, at 298 K; curve C, in the dispersed phase at room temperature (λ_{ex} = 330 nm). (b) In NaOH–ethanol solutions: curve A, at 77 K; curve B, at 298 K (λ_{ex} = 330 nm).

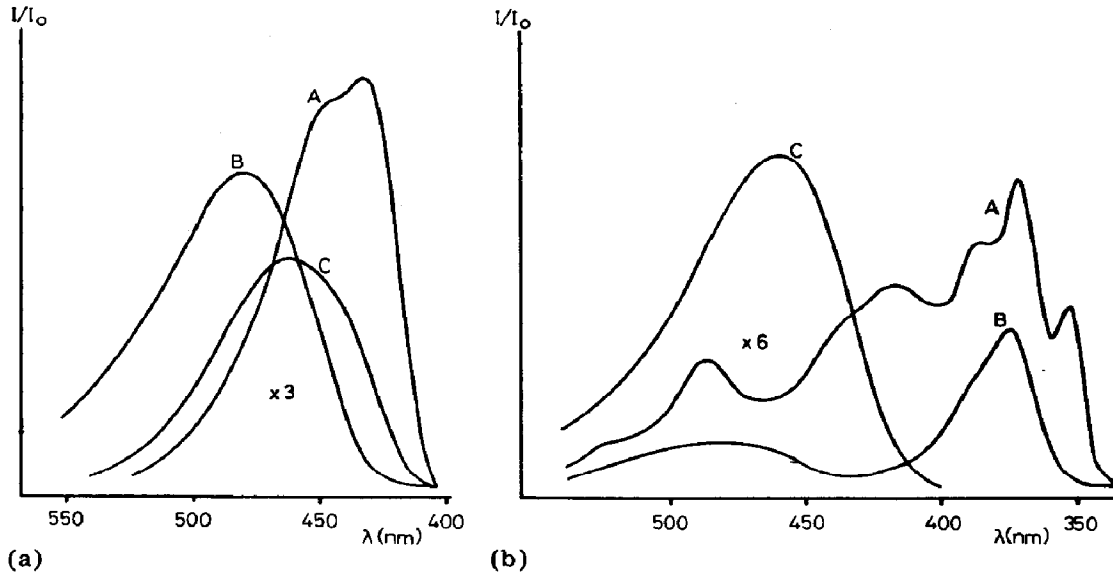


Fig. 4. Fluorescence spectra of harmaline. (a) In H_2SO_4–ethanol solutions: curve A, at 77 K; curve B, at 298 K; curve C, in the dispersed phase at room temperature (λ_{ex} = 380 nm). (b) In NaOH–ethanol solutions: curve A, at 77 K; curve B, at 298 K; curve C, in the dispersed phase at room temperature (λ_{ex} = 320 nm).

result suggests that the protonation of the neutral species in the excited state is very rapid for harmaline in ethanol–NaOH solutions.

In order to confirm this assignment we have carried out additional experiments in the adsorbed phase. It is known that ionic molecular species

adsorb strongly on cellulose surfaces [9, 10]. By exciting at $\lambda = 380$ nm (where the BH^+ species fundamentally absorbs) the spectrum was recorded. The emission of the BH^+ species can be seen. This spectrum is also seen on exciting at $\lambda = 320$ nm (where the BH^+ and the B species absorb) (Fig. 4). These spectra are the same as those obtained for harmaline in H_2SO_4–ethanol in the adsorbed phase.

At room temperature the fluorescence spectra are similar to those recorded in aqueous media [4, 5].

The resolution of the spectra is better at 77 K than at room temperature and the former are blue shifted.

The modifications of the molecular environment in the solid phase at 77 K relative to that in the liquid phase at room temperature are the origin of the differences in the fluorescence spectra as has also been suggested elsewhere [11]. Thus the rigidity of the vitreous phase retards the free relaxation of the molecular environment. Because the relaxation is restricted, the solvated form at 77 K is more energetic than at room temperature in the liquid phase where the solvent rearrangement is free, and consequently the fluorescence at 77 K should occur at shorter wavelengths. The improved resolution of the fluorescence spectra is due to the rigidity and the low temperature.

The maxima corresponding to 0–0 transitions for the molecules studied are summarized in Table 2 together with those reported for the spectra at 25 °C.

TABLE 2

Frequencies of the lowest fluorescence maxima

	$\bar{\nu}_{0-0}(BH^+)$ (cm^{-1})		$\bar{\nu}_{0-0}(B)$ (cm^{-1})		$\Delta\bar{\nu}_{0-0}$ (cm^{-1})	
	77 K	*298 K*	*77 K*	*298 K*	*77 K*	*298 K*
2-Methylharmine	25906.7	23809.5	22727.2	20202.0	−3179.5	−3607.5
Harmaline	23255.8	20920.5	28735.6	26666.6	5479.8	5746.1

3.3. Phosphorescence spectra

The phosphorescence spectra of the acid (in H_2SO_4–ethanol solution) and the neutral (in ethanol–NaOH solution) forms of 2-methylharmine and harmaline are shown in Figs. 5 and 6. The frequencies of these maxima are summarized in Table 3.

The phosphorescence emission obtained for the molecules studied corresponds to the species which are dominant in the ground state.

In contrast with the fluorescence emission of harmaline in ethanol–NaOH solution, the phosphorescence of the BH^+ species does not appear in this solvent. Therefore in the first excited triplet state the protonation of the neutral species does not occur during the lifetime of the excited state and the acid–base equilibrium is not established.

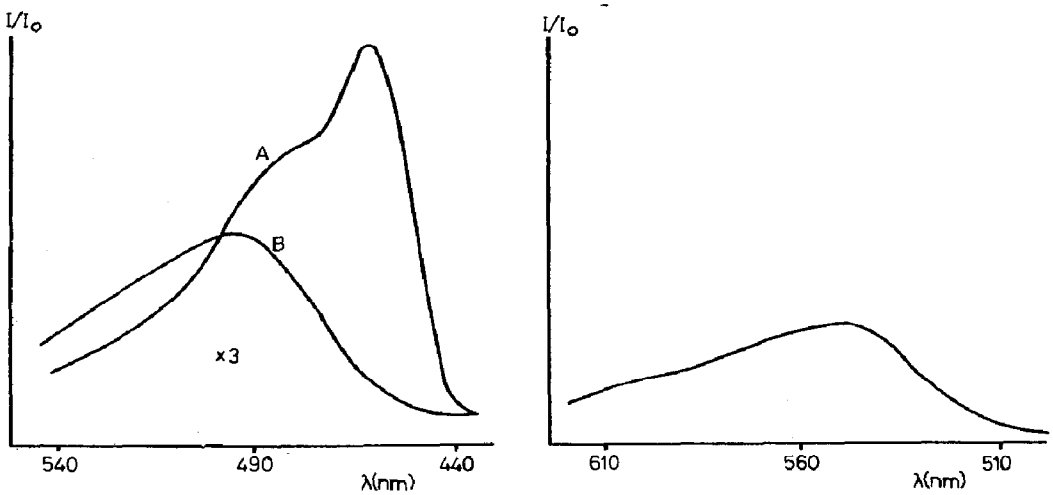


Fig. 5. Phosphorescence spectra of 2-methylharmine. (a) In H_2SO_4–ethanol solutions: curve A, at 77 K; curve B, in the dispersed phase at room temperature (λ_{ex} = 330 nm). (b) In NaOH–ethanol solution (λ_{ex} = 400 nm).

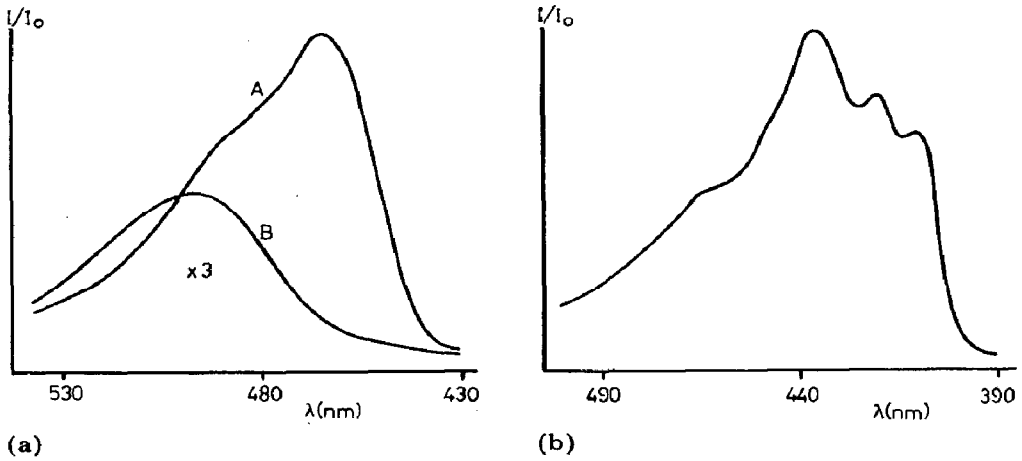


Fig. 6. Phosphorescence spectra of harmaline. (a) In H_2SO_4–ethanol solutions: curve A, at 77 K; curve B, in the dispersed phase at room temperature (λ_{ex} = 380 nm). (b) In NaOH–ethanol solution (λ_{ex} = 320 nm).

TABLE 3

Frequencies of the lowest phosphorescence maxima at 77 K

	$\bar{\nu}_{0-0}(BH^+)$ (cm^{-1})	$\bar{\nu}_{0-0}(B)$ (cm^{-1})	$\Delta\bar{\nu}_{0-0}$ (cm^{-1})
2-Methylharmine	21739.1	18181.8	−3557.3
Harmaline	21459.2	24390.2	2931.0

The phosphorescence occurs at low frequencies for the acid species in harmaline and for the neutral species in 2-methylharmine. Moreover, the phosphorescence for the basic (B) form of harmaline is more intense and better resolved than the same species of 2-methylharmine.

It is known that the phosphorescence of some compounds can be recorded at room temperature if they can be adsorbed on a dry inert surface [12]. The non-radiative processes are restricted and the radiative emission can be observed. However, not all the substances can be adsorbed satisfactorily. We have observed that the ionic substances are adsorbed well by cellulose, but that the neutral species are not adsorbed significantly or that the neutral adsorbed molecules, if any, do not exhibit phosphorescence.

One of the advantages of this technique is that the phosphorescence spectra of ionic species can be recorded at room temperature, thus avoiding the problem of finding a suitable solvent for low temperature measurements.

The intensity of phosphorescence in the adsorbed phase is small, and it is necessary to increase the slit width of the spectrofluorometer with a corresponding loss of resolution. The phosphorescence spectra of the cationic species are shown in Figs. 5 and 6. They were recorded using the chopper in order to eliminate the fluorescence emission. In addition, the phosphorescence band in the adsorbed phase is red shifted with respect to the band recorded at 77 K.

3.4. Basicity of the first excited triplet T_1

The pK_a values for the first triplet excited state were derived by means of the Förster expression [6]:

$$pK_a(T_1) = pK_a(S_0) + \frac{0.625}{T} \Delta\bar{\nu}_{0-0}$$

where $\Delta\bar{\nu}_{0-0}$ (cm^{-1}) is the difference between the 0–0 band energies of the phosphorescence spectra of the basic species (B) and the protonated species (BH^+), or, in a more general case, between the molecular species on each side of the equilibrium and T is the absolute temperature. The results obtained are only estimated because of the inaccuracies in locating the 0–0 transitions.

The differences between the first excited states are shown in Table 4. The values given were derived from the 0–0 transitions of the fluorescence

TABLE 4

Energy differences between the lowest electronic states

	$\Delta E(S_0 - S_1)$ (kcal mol^{-1})		$\Delta E(S_0 - T_1)$ (kcal mol^{-1})		$\Delta E(S_1 - T_1)$ (kcal mol^{-1})	
	BH^+	B	BH^+	B	BH^+	B
2-Methylharmine	74.1	65.0	62.1	52.0	12.0	13.0
Harmaline	66.5	82.2	61.4	69.7	5.1	12.4

TABLE 5

Excited state pK_a values estimated from emission data at 77 K

	$pK_a(S_0)$[a]	$pK_a(S_1)$	$pK_a(T_1)$
2-Methylharmine	11.5	4.9	4.0
Harmaline	10.0	21.5	16.2

[a]See refs. 4 and 5.

and phosphorescence spectra at 77 K and must be considered only estimated because of the inaccuracies in locating the maxima. It is found that the T_1 states are about 5 - 13 kcal mol^{-1} lower in energy than the S_1 states.

The $pK_a(T_1)$ values estimated are given in Table 5. It can be seen that the basicity of the T_1 state of harmaline is stronger than that of the ground state and is nearer to that of the S_1 state. For 2-methylharmine, because the site of protonation is different, the basicity of the T_1 state is lower than that of the ground state and is very similar to that of the first excited singlet.

In contrast with reports in the literature establishing that the basicities of the first triplet excited states are nearer to those of the ground states than those of the first singlet states [13 - 15], the basicities of the first triplet states of the molecules studied are quite different from those of the ground states but similar to those of the first singlet states.

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