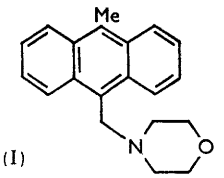
745. *Fluorescence and Ionisation Rate of an Acid.*

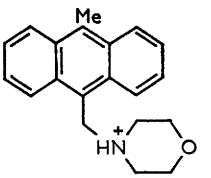
By W. S. METCALF.

If the fluorescence of an acid differs from that of its conjugate base, the fluorescence changes with the hydrogen-ion concentration. If ionisation is much slower than the emission of fluorescence, the acid and base absorb and emit light independently, except that they compete for the exciting light. If ionisation is not much slower than emission, the intensity and half-life of the fluorescence change quite differently with hydrogen-ion concentration. The rate constant for the ionisation of the acid form of 9-methyl-10-morpholinomethylanthracene is found in this way not to exceed 10^6 sec.$^{-1}$.

WELLER [1,2] has shown that the rate constants describing the ionisation of the excited form of β-naphthol can be deduced from the variation of its fluorescence with pH, and Kokubun [3] has studied acridone in acid solutions.



(I)



(II)

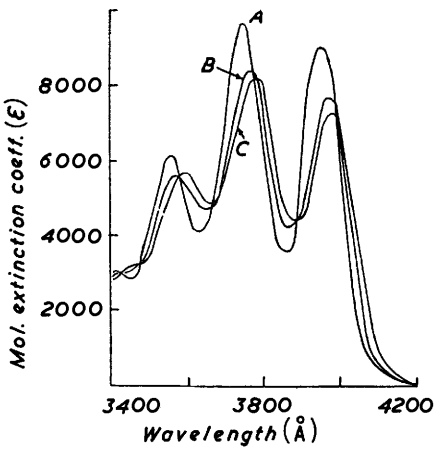
9-Methyl-10-morpholinomethylanthracene (I) was made by us for another investigation with the intention that the acid–base system in the molecule should be insulated by the

[1] Weller, *Z. Elektrochem.*, 1952, **56**, 662.
[2] Weller, *Z. phys. Chem.* (*Frankfurt*), 1955, **3**, 238; 1958, **17**, 224.
[3] Kokubun, *Z. Elektrochem.*, 1958, **62**, 599.

methylene group from the site of the optical transition in the aromatic system. The absorption spectra of the acidic and the basic form are similar, but not identical (Fig. 1). The acid fluoresced strongly but, unexpectedly, the base fluoresced only very faintly. The fluorescence of anthracene is weakly quenched by amines; in this base the amino-group is favourably placed to quench the fluorescence rapidly.

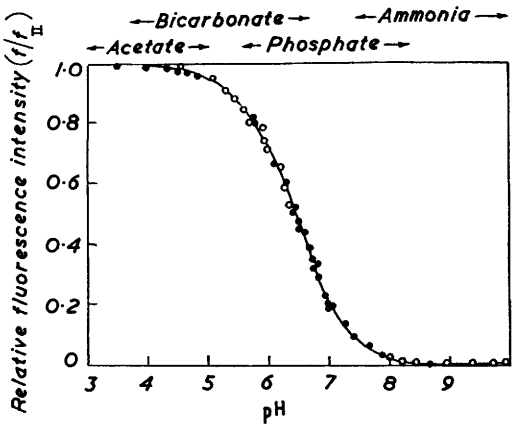
It seemed that a study of the fluorescence of buffered solutions of compound (I) might yield a value for the rate at which optically excited ions (II) lose protons to give the base (I), because such a process would provide a route other than fluorescence by which excited molecules could disappear. To this end the relative fluorescence intensities of solutions of the same stoicheiometric concentration (10^{-5}M) of (I) in acetate, bicarbonate, phosphate, and ammonia buffers were measured, with the result shown in Fig. 2.

FIG. 1. *Absorption spectrum of* 9-*methyl*-10-*morpholinomethylanthracene.*

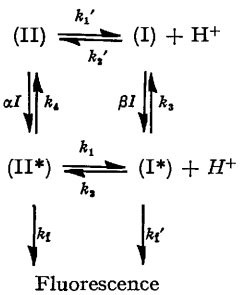


A, *In* 0·01N-NaOH.
B, *In buffer of* pH 6·73.
C, *In* 0·01N-HCl.

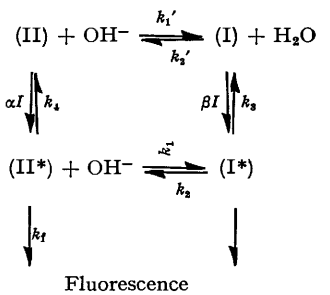
FIG. 2.



There are at least two schemes (as shown) containing known or plausible reactions which lead to the observed effect of hydrogen-ion concentration on fluorescence intensity, and there is an ambiguity in each which can be resolved by measuring the average lifetime of the excited molecules. α is the fraction of the incident light I which is absorbed by (II)



Scheme I



Scheme II

and β that which is absorbed by (I); k_4 and k_3 describe the internal degradation of energy in (II*) and (I*).

For scheme I

$$d[II^*]/dt = \alpha I - (k_f + k_1 + k_4)[II^*] + k_2[H^+][I^*] \quad . \quad . \quad . \quad (1)$$

$$d[I^*]/dt = \beta I + k_1[II^*] - (k_f' + k_2[H^+] + k_3)[I^*] \quad . \quad . \quad . \quad (2)$$

and the intensity of fluorescence

$$f = k_f[\mathrm{II}^*] + k_f'[\mathrm{I}^*] \quad . \quad . \quad . \quad . \quad . \quad . \quad . \quad . \quad (3)$$

If scheme II is adopted, k_1 and $k_2[\mathrm{H}^+]$ are replaced by $k_1[\mathrm{OH}^-]$ and k_2 respectively, throughout this paper.

The equations (1), (2), (3) can be solved [4] for f. When I is constant, the result is most readily expressed as the quantum efficiency $E = f/I$, which is found to be

$$E = \alpha E_{\mathrm{II}}\left\{\frac{1 + k_2[\mathrm{H}^+]\tau_{\mathrm{I}}(1 + \beta/\alpha)}{1 + k_1\tau_{\mathrm{II}} + k_2[\mathrm{H}^+]\tau_{\mathrm{I}}}\right\} + \beta E_{\mathrm{I}}\left\{\frac{1 + k_1\tau_{\mathrm{II}}(1 + \alpha/\beta)}{1 + k_1\tau_{\mathrm{II}} + k_2[\mathrm{H}^+]\tau_{\mathrm{I}}}\right\} \quad . \quad . \quad (4)$$

E_{II}, E_{I} are the quantum efficiencies of (II) and (I) given by $k_f/(k_f + k_4)$ and $k_f'(k_f' + k_3)$, respectively. τ_{II}, τ_{I} are given by $1/(k_f + k_4)$ and $1/(k_f' + k_3)$, and are the average lifetimes of (II*) and (I*) in acid and alkali, respectively.

When the emission of fluorescence is fast compared with the acid–base reactions the terms in braces in (4) become unity, giving

$$E = \alpha E_{\mathrm{II}} + \beta E_{\mathrm{I}} \quad . \quad . \quad . \quad . \quad . \quad . \quad . \quad . \quad (5)$$

The fluorescence from each species depends only on the fraction of the exciting light it appropriates and the efficiency with which it is converted into fluorescence,

$$\alpha = [\mathrm{II}]\varepsilon_{\mathrm{II}}/([\mathrm{II}]\varepsilon_{\mathrm{II}} + [\mathrm{I}]\varepsilon_{\mathrm{I}}) \quad \text{and} \quad \alpha + \beta = 1$$

where the molar extinction coefficients $\varepsilon_{\mathrm{II}}$, ε_{I} refer to the frequency of the exciting light (3650 Å). Therefore equation (5) may be rearranged to

$$\left\{1 - \frac{f}{f_{\mathrm{II}}}\right\} \Big/ \left\{\frac{f}{f_{\mathrm{II}}} - \frac{f_{\mathrm{I}}}{f_{\mathrm{II}}}\right\} = \frac{K}{[\mathrm{H}^+]} \quad . \quad . \quad . \quad . \quad . \quad . \quad (6)$$

where $K = [\mathrm{I}][\mathrm{H}^+]/[\mathrm{II}] = k_1'/k_2'$ and is the ionisation constant of the unexcited acid (II).

As the curve of Fig. 2 shows, this equation describes the experimental results correctly if $K = 10^{-6\cdot6}$ mole l.$^{-1}$ and $f_{\mathrm{I}}/f_{\mathrm{II}} = 0\cdot012$. Now $10^{-6\cdot6}$ mole l.$^{-1}$ is an unusually high value for the ionisation constant of the acid form of a tertiary alkylamine. Moreover, the experimental results in Fig. 2 are accounted for equally well by the supposition that the emission of fluorescence is slow compared with the acid–base reactions. In this case equation (4) becomes

$$\frac{f}{f_{\mathrm{II}}} = \left\{\frac{1 + \dfrac{K}{[\mathrm{H}^+]} \cdot \dfrac{\varepsilon_{\mathrm{I}}}{\varepsilon_{\mathrm{II}}}}{1 + \dfrac{K}{[\mathrm{H}^+]}}\right\} \times \frac{1 + \dfrac{E_{\mathrm{I}}}{E_{\mathrm{II}}} \cdot \dfrac{\tau_{\mathrm{II}}}{\tau_{\mathrm{I}}} \cdot \dfrac{K^*}{[\mathrm{H}^+]}}{1 + \dfrac{\tau_{\mathrm{II}}}{\tau_{\mathrm{I}}} \cdot \dfrac{K^*}{[\mathrm{H}^+]}} \quad . \quad . \quad . \quad . \quad . \quad (7)$$

where $K^* = k_1/k_2$, which is the ionisation constant of the excited acid (II*).

The molar extinction coefficients $\varepsilon_{\mathrm{II}}$ and ε_{I} of acid and base are so nearly equal at the wavelength of the exciting light (3650 Å) that the term in braces is indistinguishable from unity. The rest can be arranged in the form

$$\left\{1 - \frac{f}{f_{\mathrm{II}}}\right\} \Big/ \left\{\frac{f}{f_{\mathrm{II}}} - \frac{f_{\mathrm{I}}}{f_{\mathrm{II}}}\right\} = \frac{\tau_{\mathrm{II}}}{\tau_{\mathrm{I}}} \cdot \frac{K^*}{[\mathrm{H}^+]} \quad . \quad . \quad . \quad . \quad . \quad (8)$$

which also describes accurately the data in Fig. 2 if $K^* = 10^{-8\cdot5}$ mole l.$^{-1}$ and $\tau_{\mathrm{I}}/\tau_{\mathrm{II}} = 0\cdot012$. The value $10^{-8\cdot5}$ mole l.$^{-1}$ is near that expected for the ionisation constant of the acid form of a tertiary alkylamine. The optical excitation is not expected to affect the ionisation constant noticeably in this compound.

[4] Metcalf, preceding paper.

However, measurement of the phase delay of the fluorescence when the exciting light is modulated at 5 Mc./sec. shows that emission *is* fast compared with the acid–base reactions, and that the first interpretation requiring $K = 10^{-6\cdot6}$ mole l.$^{-1}$ is the correct one.

When the exciting light is modulated, its intensity can be described by a Fourier series,

$$I = I_1 + I_2 \sin \omega t + I_3 \sin 2\omega t + \quad . \quad . \quad . \quad . \quad . \quad . \quad (9)$$

The solution [4] of equations (1), (2), (3), (9) is:

$$f = f_1 + f_2 \sin (\omega t - \phi_2) + f_3 \sin (2\omega t - \phi_3) \quad . \quad . \quad . \quad . \quad (10)$$

where $f_1 = EI_1$ and E is given by equation (4), and tan ϕ_2, which is also a measurable quantity,[5] is given by

$$\tan \phi_2 = \omega(\omega^2 S + PR - QS)/[QR + \omega^2 (PS - R)] \quad . \quad . \quad . \quad (11)$$

P, Q, R, S are functions of the rate constants, the concentrations, and the extinction coefficients, as follows:

$$P = k_f + k_4 + k_f' + k_3 + k_1 + k_2[H^+]$$

$$Q = (k_f + k_4 + k_1)(k_f' + k_3 + k_2[H^+]) - k_1 k_2[H^+]$$

$$R = k_f\{\alpha(k_f' + k_3 + k_2[H^+]) + k_2[H^+]\} + k_f'[\alpha k_1 + \beta(k_f + k_4 + k_1)]$$

$$S = k_f\alpha + k_f'\beta$$

The expression for tan ϕ_2 can be greatly simplified by the following approximations, justifiable for the present compound:

$$\alpha/\beta = [II]/[I] \qquad k_1/k_2 = k_1'/k_2'; \qquad k_f = k_f' \text{ (ref. 6)}$$

The expression that results is still too complex to quote in a form whose properties are obvious. Insertion of numerical values shows that if the rate of transformation of the excited acid to the base is greater than 2% of the rate of emission of fluorescence, then the average life of the excited acid molecules will be measurably reduced in the pH region near 6·6 where the fluorescence falls. Experiment shows that this is not the case: tan ϕ_2 remains constant at a value corresponding to $\tau_{II} = 1\cdot40 \times 10^{-8}$ sec. as the pH is increased until f/f_{II} has fallen to 0·2, beyond which point tan ϕ_2 can no longer be measured accurately. The conclusion is that k_1 is less than $0\cdot02/\tau_{II}$, that is, less than $1\cdot4 \times 10^6$ sec., and that the ionisation constant of the unexcited acid is $10^{-6\cdot6}$ mole l.$^{-1}$.

A direct determination of the ionisation constant K by conventional methods failed, because the base, whose solubility in water is only 4×10^{-5}M, does not equilibrate quickly with its solution in a buffer, and it is absorbed slowly on glass. The ultraviolet absorption spectrum of a solution buffered at $[H^+] = 10^{-6\cdot73}$ mole l.$^{-1}$ showed both acid and base forms to be present, but did not allow the calculation of K because the spectra of the two forms are very similar, and the measurements were affected by fluorescence reaching the photocell of the Beckman recording spectrophotometer (see Fig. 1).

For scheme I, $K = 10^{-6\cdot6}$ mole l.$^{-1}$, $\tau_{II} = 1\cdot4 \times 10^{-8}$ sec., $k_1 < 10^{6\cdot1}$ sec.$^{-1}$. If, as is expected, $K = K^*$, where the asterisk refers to the excited species, then $k_2 < 10^{12\cdot7}$ l. mole^{-1} sec.$^{-1}$, and if, as is also expected [6] from the similarity of the absorption spectra of (I) and (II), $k_f = k_f'$, then $\tau_I = 0\cdot012\tau_{II} = 1\cdot7 \times 10^{-10}$ sec.$^{-1}$.

For scheme II, $K = 10^{-6\cdot6}$ mole l.$^{-1}$, $\tau_{II} = 1\cdot4 \times 10^{-8}$ sec., $k_2 < 10^{6\cdot2}$ sec.$^{-1}$, and with the approximations above, $k_1 < 10^{13\cdot6}$ l. mole^{-1} sec.$^{-1}$ and $\tau_I = 1\cdot7 \times 10^{-10}$ sec.

The large ionisation constant, $10^{-6\cdot6}$ mole l.$^{-1}$, compared with that of other aliphatic

[5] Bailey and Rollefson, *J. Chem. Phys.*, 1953, **21**, 1315.
[6] Lewis and Kasha, *J. Amer. Chem. Soc.*, 1945, **67**, 995.

tertiary amines is noteworthy. The rate of proton loss is not anomalous compared with that of the oxygen and carbon acids.[7]

Experimental

9-Methyl-10-morpholinomethylanthracene, m. p. 137—138°, is made from morpholine and 9-chloromethyl-10-methylanthracene,[8] and crystallises from ethyl acetate and from ether (Found: C, 82·8; H, 7·3; N, 4·7. $C_{20}H_{21}ON$ requires C, 82·5; H, 7·3; N, 4·8%).

The solutions, freed from oxygen, were buffered by acetic acid and sodium acetate, by carbon dioxide (saturated at 1 atm.) and sodium hydrogen carbonate, by disodium hydrogen phosphate and hydrochloric acid, or by ammonia and hydrochloric acid.

As the free base is absorbed slowly on glass, a concentrated solution of the alkaline buffer component was added from a micrometer syringe to a 10^{-5}M-solution of the fluorescent compound in the dilute acid component of the buffer. Correction was made for the consequent small dilution of the fluorescent compound. The concentration of hydrogen ions was calculated from the concentrations of the various species present and their activity coefficients f_z given by $\log f_z = -0{\cdot}50\, z^2\sqrt{\mu}/(1 + \sqrt{\mu})$. The ionic strength μ was near 0·01M. The ionisation constants of the buffer acids recorded by Robinson and Stokes [9] were adjusted to 20°. The quenching of fluorescence by the components of the buffer was not detectable at the concentrations used.

The photometer for measuring relative fluorescence intensities (f/f_{II}) has been described.[10] The inequality of the dark currents of the photocells is corrected more easily by light from a very small neon lamp whose distance from the photocells is variable, than by adjusting the current in a hot filament lamp as described earlier.

The fluorometer used to measure tan ϕ_2 is based on that of Bailey and Rollefson.[5] Details will appear elsewhere. Lifetimes are obtained directly from a nomogram prepared from the equations derived by Bailey and Rollefson. Our measurement of the average life of excited acridone ($1{\cdot}45 \times 10^{-8}$ sec.) differs from that of Bailey and Rollefson ($1{\cdot}59 \times 10^{-8}$) by an amount much greater than the sensitivity of the apparatus (1×10^{-10} sec.). Sally G. Page has shown that the measured mean life of excited molecules of anthracene (2×10^{-4}M to 4×10^{-5}M in " AnalaR " benzene), of 9,10-diphenylanthracene (10^{-5}M to 4×10^{-5}M in ethanol), of acridone (saturated in water), and of quinine sulphate (2×10^{-6}M to 2×10^{-5}M in 0·01M-nitric acid), is constant over the range of concentrations quoted. Oxygen must be removed from these solutions, except in the case of quinine sulphate. These compounds were easily purified, and purification did not change the lifetime observed. The mean lives, measured on our apparatus, are $3{\cdot}9 \times 10^{-9}$, $8{\cdot}8 \times 10^{-9}$, $14{\cdot}5 \times 10^{-9}$, and $20{\cdot}5 \times 10^{-9}$ sec., respectively. Measurements in other laboratories differ among themselves and from ours by more than the expected error. Until agreement is secured among several laboratories as to the precise values of these lifetimes, all measurements could, with advantage, be accompanied by a measurement on one of these substances of similar lifetime. Accordingly, $\tau_{II} = (1/\omega)\tan\phi_2 = 1{\cdot}40 \times 10^{-8}$ sec. at 20°. For acridone under similar conditions of absorption

$$\tau = 1{\cdot}45 \times 10^{-8} \text{ sec.}, \ \omega = 2\pi\nu, \text{ where } \nu = 5{\cdot}000 \times 10^{6} \text{ c./sec.}$$

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Christchurch, New Zealand. [*Received, December 21st*, 1959.]

[7] Bell, *Quart. Rev.*, 1959, **13**, 169.
[8] Badger and Pearce, *J.*, 1950, 2314.
[9] Robinson and Stokes, " Electrolyte Solutions," Butterworths, London, 1955.
[10] Melhuish and Metcalf, *J.*, 1954, 976.