

Fig. 1.—Non-contact eutectic fusion of azobenzene (upper, irregular fragment; m.p. 67°) and acenaphthene (lower, elongated rod-like crystal; m.p. 90°) at 50.6°. Eutectic temperature 48.6°. 1, no evidence of fusion after 1 hour at 47.6°; 2 and 3, faint sign of droplets after 18 min. at 49.6°; 4, easily detectable droplets after 30 min. at 50.6°; 5, 6, 7 and 8, progressive accumulation of droplets and melting of solid components at 50.6°.

space the cycle is solid to liquid solution. There is, of course, no thermodynamic difficulty about the formation of a liquid solution from the two pure solids, not in physical contact, at or above the eutectic temperature. The solids are in thermodynamic contact through the vapor phase and the process considered, at constant temperature and virtually constant pressure, is accompanied by a decrease in free energy. The composition of the resulting vapor mixture and, hence, the composition of the liquid, would be expected to be determined by the vapor pressures of the two solids. The composition of the liquid is found to be that

of the eutectic. How the composition of the gas phase is related to that of the liquid cannot be stated. Perhaps data on sublimation pressures which are now in the process of being gathered will throw more light on the picture. Since all eutectic systems are characterized by the fact that the components differ in crystal structure it may well be that all solid eutectics contain inter-crystalline spaces. If this is the case the cycle of eutectic melting may always be solid to vapor to solution.

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[CONTRIBUTION FROM THE SCHOOL OF CHEMISTRY OF THE UNIVERSITY OF MINNESOTA]

## Quenching of the Fluorescence of Solutions of Porphyrins and of Chlorophyll<sup>1</sup>

BY ROBERT LIVINGSTON, LOU THOMPSON AND M. V. RAMARAO

The fluorescence of solutions of mesoporphyrin in benzene is quenched by the following substances: oxygen, 0.0028; chloranil, 0.0034; quinone, 0.0058; trinitrotoluene, 0.0062; *m*-dinitrobenzene, 0.011. The number following each compound is  $[Q]_{1/2}$ , the molarity of the quencher required to reduce the intensity of the fluorescence to one half of its unquenched value. Nine other compounds which were tested proved to be non-quenchers. The quenching efficiency for mesoporphyrin of each of these 14 compounds parallels in general their quenching efficiency for chlorophyll

The quenching of the fluorescence of chlorophyll-a solutions by each of a variety of added substances was measured recently,<sup>2</sup> as part of a program to study the primary act in photochemical processes involving chlorophyll. Most fluorescent solutions which have been studied, are quenched chiefly by reducing agents.<sup>3</sup> All of the compounds which effectively quench the fluorescence of chlorophyll solutions are oxidizing agents. It is of interest to know whether this latter group of quenchers is peculiar to chlorophyll or if it is characteristic of a group of molecules similar in some respects to

chlorophyll. Accordingly, the quenching of solutions of mesoporphyrin and of protoporphyrin has been studied.

### Experimental Methods and Materials

Except for the light filters, the apparatus and routine procedures were similar to those described by Livingston and Ke.<sup>2</sup> The  $\lambda$  5460 Å. line was isolated from the radiation of a GE AH4 mercury arc by means of 2 cm. of 10% copper sulfate, a 4 mm. Corning 3486 and an 8 mm. Corning 5120 filter. The complementary filter, situated in front of the photocell, was a Wratten 88 which transmits light of wave lengths longer than 6000 Å.

The porphyrins used were samples of mesoporphyrin dimethyl ester and of protoporphyrin dimethyl ester,<sup>4</sup> whose final purification was effected chromatographically on activated alumina. The solvent used in these experiments was benzene, which was prepared from "thiophene-free" mate-

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(2) R. Livingston and Chun-Lin Ke, *THIS JOURNAL*, **72**, 909 (1950).

(3) P. Pringsheim, "Fluorescence and Phosphorescence," Interscience Publishers, Inc., New York, N. Y., 1949, pp. 328-335.

(4) We wish to express our thanks to Dr. S. Schwartz of the Medical School of the University of Minnesota who kindly put those samples at our disposal.

TABLE I  
 SUMMARY OF QUENCHING DATA

The number,  $n$ , of quenching experiments performed is given in column four, and the range of concentrations of quenchers in column three.

Quencher	Porphyrin	Limits of [Q]	$n$	$[Q]_{1/2}$	$k_1$	$k_2$	$\frac{k_1(\text{porph})}{k_1(\text{chph})}$
Oxygen	Meso	0.0002-0.0067	8	0.0028	350	0	10.0
Oxygen	Proto	.0003- .0059	8	.0031	320	0	9.1
Chloranil	Meso	.0010- .0201	6	.0034	255	7000	1.5
Quinone	Meso	.0008- .0217	14	.0056	175	0	1.5
Quinone	Proto	.0005- .0300	8	.0058	170	0	1.4
Trinitrotoluene	Meso	.0003- .0237	7	.0062	130	5500	1.2
Trinitrotoluene	Proto	.0005- .0252	7	.0073	115	2500	1.0
<i>m</i> -Dinitrobenzene	Meso	.0014- .0325	9	.0113	75	1500	1.0

rial by treatment with metallic sodium followed by fractional distillation.<sup>5</sup> The reagents used as quenchers were samples which had been purified by Mr. Ke<sup>2</sup> and stored under appropriate conditions.

### Experimental Results

The experimental results are summarized in Tables I and II and in Fig. 1. All of the quenching data can be fitted by a modified Stern-Volmer equation

$$I_0/I = 1 + k_1[Q] + k_2[Q]^2$$

where  $I_0$  is the intensity of the unquenched fluorescence,  $I$  of the quenched fluorescence, and  $[Q]$  is the

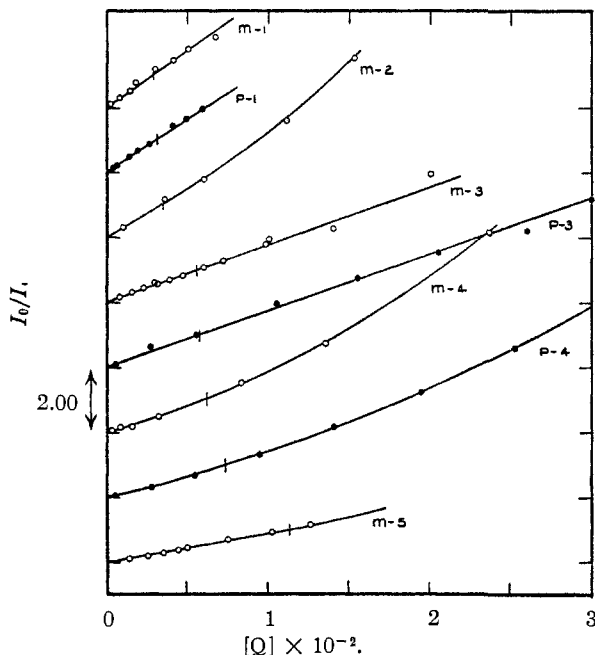


Fig. 1.—Summary of quenching data: m-1, mesoporphyrin and oxygen; p-1 protoporphyrin and oxygen; m-2, mesoporphyrin and chloranil; m-3, mesoporphyrin and quinone; p-3, protoporphyrin and quinone; m-4, mesoporphyrin and trinitrotoluene; p-4, protoporphyrin and trinitrotoluene; m-5, *m*-mesoporphyrin and *m*-dinitrobenzene.

(5) Preliminary measurements, by Dr. W. F. Watson on coproporphyrin and more extensive (unpublished) measurements by Mr. S. Weil on mesoporphyrin, demonstrate that the fluorescence efficiency of porphyrins is the same in dry benzene as it is in benzene containing traces of water, methanol or other "activators" (cf. R. Livingston, W. Watson and J. McArdle, THIS JOURNAL, 71, 1542 (1949)). Accordingly no special care was taken in the present measurements to control the concentrations of "activators" present in the solvent. The solvents were dry by ordinary laboratory standards, but were exposed to laboratory air during the preparation of the solutions.

TABLE II

### EFFECT OF NON-QUENCHERS

Reagent	[Q], $m$	$I_0/I$
Nitropropane	0.13	1.01
Butyl nitrate	.05	1.0
Aniline	.07	1.01
2,6-Diaminopyridine	.19	1.0
<i>t</i> -Hexylmercaptan	.01	1.01
Carbon dioxide	1 atm.	1.0
Carbon monoxide	1 atm.	1.0
Nitrous oxide	1 atm.	1.0

molarity of the quencher. The values of the constants,  $k_1$  and  $k_2$ , were determined empirically. Unlike the chlorophyll data,<sup>2</sup> it is impossible to represent the present results by a formula in which  $k_1^2/8$  is substituted for  $k_2$ . Quenching of the fluorescence of porphyrins by quinone (and probably by oxygen) follows the simple Stern-Volmer law over a wide range of concentrations.

In addition to the quenchers listed in Table I, preliminary measurements were made upon the effect of nitric oxide. Under the present experimental conditions, nitric oxide appears to quench the fluorescence of mesoporphyrin in benzene, obeying a Stern-Volmer equation with a  $k_1$  equal to approximately 55 l./mole. However, this result is open to question, since nitric oxide was also observed to produce marked (reversible and irreversible) color changes in benzene solutions of mesoporphyrin. Preliminary experiments also indicated that phenylhydrazine is a weak quencher for the fluorescence of mesoporphyrin.

Of the twenty-eight substances classed<sup>2</sup> as non-quenchers for chlorophyll, nine were tested in mesoporphyrin solutions. Eight of those, which are listed in Table II, proved to be non-quenchers, or at most very weak quenching agents. The other substance, butyl nitrite, appeared to increase the fluorescence intensity of mesoporphyrin. The increase caused by 0.12  $m$  butyl nitrite was 7% and by 0.18  $m$ , 9%. This effect was not further investigated.

### Discussion

The quenching efficiencies of the fourteen substances tested for porphyrin solutions roughly parallels their efficiencies for chlorophyll solutions. The ratios of the quenching constants,  $k_1$ , for porphyrins to those for chlorophyll are listed in the last column of Table I. With the exception of the value for oxygen, the values of the ratio lie between 1.0 and 1.5. This general similarity in quenching

efficiencies shows that quenching by these agents<sup>1</sup> is not uniquely characteristic of chlorophyll or of chlorins, but is probably a property of all fluorescent porphyrin derivatives and perhaps also of aromatic hydrocarbons. The observed increase in efficiency of quenching for porphyrins may be due in part to the difference in solvent; benzene being used in the present experiments and methanol (or acetone) in the chlorophyll studies.

It is also suggestive that oxygen<sup>6</sup> and nitric oxide<sup>7</sup> which quench the fluorescence of chlorophyll solutions<sup>2</sup> are effective quenchers for solutions of a number of aromatic hydrocarbons. Nitrobenzene which is a moderately strong quencher for

(6) E. J. Bowen and A. H. Williams, *Trans. Faraday Soc.*, **35**, 765 (1939).

(7) H. Weil-Malherbe and J. Weiss, *J. Chem. Soc.*, 541 (1944).

chlorophyll<sup>2</sup> has been reported<sup>8</sup> to quench the fluorescence of rubrene.

The quenching constants,  $k_1$ , for oxygen upon aromatic hydrocarbons, range from 180 to 2400 l./mole. The corresponding values for the porphyrins (350 and 320 l./mole) fall within this range; but, in marked contrast, the value for chlorophyll-a (in ethanol) is tenfold smaller. It is interesting to speculate whether this low quenching efficiency may be a unique property of chlorophyll, possibly related to the very low quantum yield for the autoxidation of alcoholic solutions of chlorophyll.<sup>9</sup>

(8) E. J. Bowen, *Quart. Rev. Chem. Soc. (London)*, **1**, 1 (1947).

(9) J. Knight and R. Livingston, *J. Phys. Colloid Chem.*, **54**, 703 (1950).

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## NOTES

### Acenaphthylene as a Philodienic Compound

BY E. BERGMANN

Four years ago,<sup>1</sup> the philodienic properties of acenaphthylene (I) were reported; a new synthetic route into the fluoranthene series is based on the reaction of (I) with dienes. The investigation has since been continued in this Laboratory; the results are in accord with those published some months ago by Kloetzel and Mertel.<sup>2</sup> Of the additional experimental material which has accumulated, the reaction between (I) and diphenylisobenzofuran (II)<sup>3</sup> may be reported. The normal addition product (III) which was obtained in quantitative yield when the components reacted in xylene solution, could be dehydrated to 7,12-diphenylbenz[K]fluoranthene (IV). (IV) was formed directly, when the components were heated together without solvent at 160°. This substance is the diphenyl derivative of the benz[K]fluoranthene which Orchin and Reggel<sup>4</sup> have obtained by cyclodehydrogenation of 1,2'-dinaphthyl and which Moureu, Chovin and Rivoal,<sup>5</sup> and Orchin and Reggel<sup>6</sup> have synthesized by unambiguous routes. Its ultraviolet spectrum is similar to that of the parent substance,<sup>4</sup> especially as far as the bands of longer wave length are concerned. Indeed, as in the case of 9,10-diphenylanthracene,<sup>7</sup> one could expect, for geometrical reasons, that the phenyl groups would not exert any bathochromic influence on the absorption spectrum.

(1) E. Bergmann, *Nature*, **161**, 889 (1948); *Bull. soc. chim. France*, **19** (1948).

(2) M. C. Kloetzel and H. E. Mertel, *THIS JOURNAL*, **72**, 4786 (1950); see also N. C. Deno, *ibid.*, **72**, 4057 (1950).

(3) For a review of the diene reactions of this compound, see ref. 1.

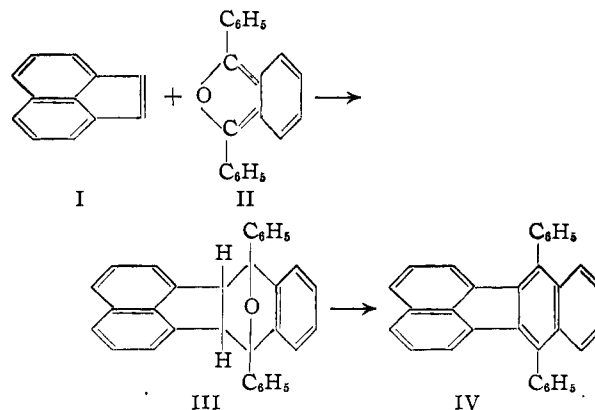
(4) M. Orchin and L. Reggel, *THIS JOURNAL*, **69**, 505 (1947).

(5) H. Moureu, P. Chovin and G. Rivoal, *Compt. rend.*, **223**, 951 (1946); **225**, 501 (1947); *Bull. soc. chim. France*, **99** (1948).

(6) M. Orchin and L. Reggel, *THIS JOURNAL*, **73**, 436 (1951).

(7) R. N. Jones, *ibid.*, **67**, 2021, 2127 (1945).

Benz[K]fluoranthene		IV (in dioxane)	
$\lambda$ (Å.)	log $E_m$	$\lambda$ (Å.)	log $E_m$
4000	4.17	4080	4.23
		3860	4.18
3710	4.13	3660	3.90
3595	3.90	3440	3.90
		3300	4.03
3095	4.83	3100	4.79
3000	4.70	2980	4.63
2850	4.43		
2695	4.37	2700	4.48
2450	4.77	2520	4.80
2175	4.60		



**Adduct (III).**—A mixture of acenaphthylene<sup>2</sup> (I) (0.6 g.) and diphenylisobenzofuran<sup>3</sup> (II) (1.1 g.) was refluxed in xylene (10 cc.) for 7 hours. (Part of the adduct crystallized upon cooling.) Light petroleum (50 cc.) was added, and the precipitate filtered and recrystallized from isoamyl alcohol; yellowish needles with green (in solution green-blue) fluorescence, m.p. 208°; yield, quantitative.

**Anal.** Calcd. for  $C_{22}H_{20}$ : C, 91.0; H, 5.2. Found: C, 91.0, 91.0; H, 5.3, 5.2.

(8) A. Guyot and J. Catel, *Compt. rend.*, **140**, 1348 (1905); *Bull. soc. chim. France*, [3], **38**, 1124 (1906).