Hydrogen Bonding Effect on the Fluorescence of π-Electron System. III. Fluorescence Quenching of Some Nitrogen Heterocycles Caused by Hydrogen Bond Formation*

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In II[1] of this series, we have interpreted the inner quenching of fluorescence of some nitrogen heterocycles as due to the interaction between *n*- and π-electrons, on the basis of the fact that the enhancements of fluorescence yields of these molecules caused by hydrogen bonding is greater, the stronger the donating powers of proton donors used. The fluorescence yields of acridine hydrogen bonded with ethanol, β-phenylethylalcohol and benzylalcohol are almost the same. We can observe, however, no enhancement of fluorescence yield, when acridine is hydrogen bonded with phenol which is capable of making hydrogen bond intimately connected with π-electron system in contrast to molecules such as benzylalcohol and β-phenylethylalcohol. Actually, we have observed a remarkable fluorescence quenching by phenol when 3,6-diaminoacridine (DAA) and 3,6-bisdimethylaminoacridine (BDAA) which are able to fluoresce fairly well even in non-polar solvent, are used as fluorescer and proton acceptor.

These facts indicate that the interaction between π-electron systems via the hydrogen bond may lead to the non-radiative degradation of excited state. To elucidate the problem further, we have used various proton donors such as naphthols, phenol, aniline and pyrrole, the hydrogen bonding by which are intimately related with π-electron system.

For all of these donors, strong or moderate decrease of fluorescence intensity was observed. Moreover, the effect of viscosity on the quenching was studied by changing the hydrocarbon solvents from *n*-hexane to liquid paraffin, and no viscosity dependence of quenching was observed. In the present paper, we will report these facts with some discussions on the mechanism of fluorescence quenching caused by the interaction between π-electron systems of fluorescer and of quencher molecules.

Experimental

Apparatus.—Absorption spectra were measured with Beckman spectrophotometer model DU. Fluorometer was the same as described previously[2]. A high pressure mercury lamp with appropriate filters was used as an exciting light source. Matsuda color filter UV-DI was used for all samples as an exciting light filter.

For the fluorescent light, the light filter used for acridine was the same as before[2], and for DAA and BDAA, Matsuda color filter VY—I was used.

Reagents.—Acridine, DAA[3] and BDAA[3] were the same sample as used previously. *n*-Hexane and benzene were the same as reported before[1]. Cyclohexane and decaline were shaken with fuming sulfuric acid diluted with conc. sulfuric acid and distilled carefully. Liquid paraffin was purified through the column of activated alumina for removing the fluorescing impurity. Extra pure grade phenol was distilled carefully before use. Aniline was purified by the method of Keyes and Hildebrand[4], and stored in vacuo. Commercial pyrrole was distilled carefully and stored in vacuo.

Experimental Results

A. Change of Absorption Spectrum by

* I and II of this series: N. Mataga, Y. Kaifu and M. Koizumi, This Bulletin, **29**, 115 (1956), and N. Mataga and S. Tsuno, ibid., **30**, 368 (1957).

1) N. Mataga and S. Tsuno, loc. cit.

2) N. Mataga, Y. Kaifu and M. Koizumi, This Bulletin **29**, 373 (1956).

3) N. Mataga, ibid., **30**, 375 (1957).

4) D. B. Keyes and J. H. Hildebrand, *J. Am. Chem. Soc.*, **39**, 2126 (1917).

Hydrogen Bonding.—According to Zanker[5], the lowest excited singlet states of DAA and BDAA are ^1L_a as it is the case for acridine. When the proton donors are added to the solutions of these nitrogen heterocycles in non-polar solvents, the absorption spectra show characteristic changes due to the hydrogen bonding equilibrium. In all these molecules, hydrogen bonding induces the red shift of ^1L_a band. Some examples of these spectral changes caused by hydrogen bonding are shown in Figs. 1 and 2.

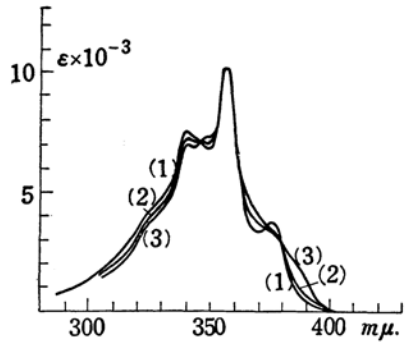

Fig. 1. Change of absorption spectrum of acridine in benzene by added phenol. Concentration of acridine: 5.00×10^{-5} mole/l. Concentration of phenol: (1) 0, (2) 7.12×10^{-3} mole/l., (3) 3.56×10^{-2} mole/l.

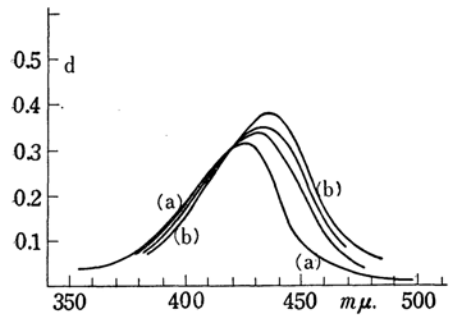

Fig. 2. Change of absorption spectrum of BDAA in benzene by added phenol. Concentration of BDAA: 1.36×10^{-5} mole/l. Cell length: 1 cm. Concentration of added phenol: (a) 0, (b) 9.4×10^{-3} mole/l.

5) V. Zanker, *Z. physikal. Chem.*, N. F., **2**, 52 (1954).

* The concentration ranges of donors used for the calculation of equilibrium constants, are as follows.
phenol and naphthols: 10^{-4}~10^{-2} mol./l., where nearly 100% monomer exists. [a]
(a) R. Mecke, *Disc. Faraday Soc.*, No. **9**, 161 (1950).
aniline: 10^{-2}~10^{-1} mol./l., where nearly 100% monomer exists.[b]
pyrrole: 5×10^{-2}~4×10^{-1} mol./l., where 90~70% monomer exists.[b]
(b) N. Fuson, M. L. Josien, R. L. Powell and E. Utterback, *J. Chem. Phys.*, **20**, 145 (1952).

From these spectral changes, we can evaluate the equilibrium constant of hydrogen bond formation in the ground state, using Eq. (1)[1]*.

$$\frac{1-(d_0/d)}{[D]} = -K+\left(\frac{\varepsilon_c}{\varepsilon_a}\right)K(d_0/d) \qquad (1)$$

Some examples of analyses using this formula were given in Figs. 3 and 4.

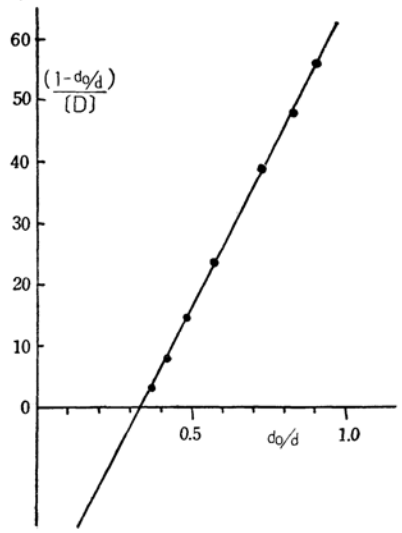

Fig. 3. $(1-d_0/d)/[D]\sim d_0/d$ relation for acridine-phenol-benzene system at 386 mμ.

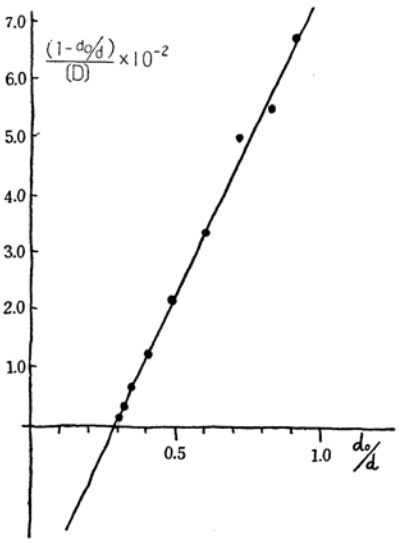

Fig. 4. $(1-d_0/d)/[D]\sim d_0/d$ relation for BDAA-phenol-benzene system at 448 mμ.

B. Effect of Hydrogen Bonding on the Fluorescence Intensity.—Acridine is non-fluorscent in non-polar solvent, and no enhancement of fluorescence intensity was

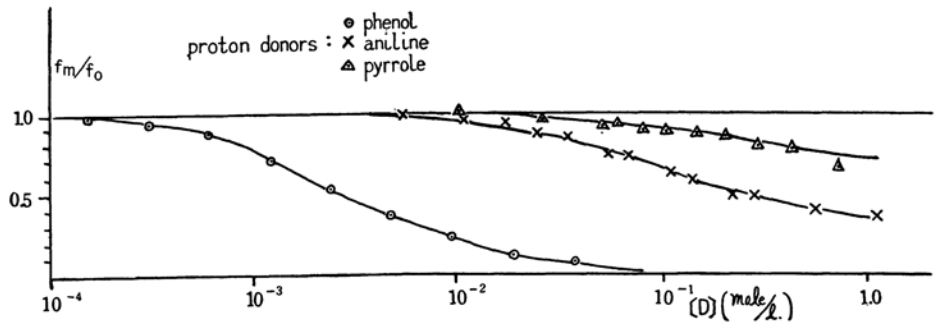

Fig. 5. Changes of relative fluorescence intensity of BDAA in benzene by added various proton donors.

observed when it is hydrogen bonded with phenol.

DAA and BDAA are fluorescent even in a non-polar solvent, and decrease in fluorescence intensity is observed, when various proton donors such as phenol, naphthols, aniline and pyrrole are added in the solution. Some examples of quenching curves for BDAA in benzene are given in Fig. 5.

The observed changes of fluorescence intensity may most probably be attributed to the fact that the free and the hydrogen bonded molecules have different efficiency of fluorescence and that the concentration of these molecular species are determined by association equilibrium. Then, from these changes of fluorescence intensity, we can evaluate the equilibrium constant of hydrogen bond formation and the ratio of the quantum yields of fluorescences of hydrogen bonded and free molecules, using the following formulas, (2) and (3)[1]*.

$$\frac{1-(f_0/f_m)}{[D]} = -K+\alpha\left(\frac{\varepsilon_c}{\varepsilon_a}\right)K(f_0/f_m) \qquad (2)$$

$$\frac{1-(f_0/f_m)(d_m/d_0)}{[D]} = -K'+\alpha\left(\frac{d_m}{d_0}\right)K'(f_0/f_m) \qquad (3)$$

Owing to a very small difference of extinction coefficients of free and hydrogen bonded molecules at the wave length of exciting light, (2) and (3) practically coincide in their forms and it has been established that the experimental results can be reproduced by either Eq. (2) or (3) satisfactorily for all cases studied, the equilibrium constants evaluated by Eqs. (2) and (3) being paractically the same. Some examples of these analysis are given in Figs. 6 and 7. Further, the equilibrium constant obtained in this way from fluorescence intensity measurement agrees with that evaluated from the change in absorption spectrum.

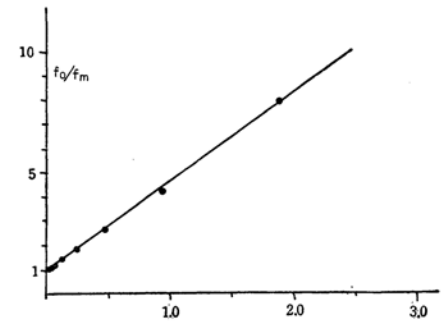

Fig. 6. $f_0/f_m \sim [D]$ relation for BDAA-phenol-benzene system[a]
(a) Conditions, $\alpha=0$ in Eq. (2) or $\alpha=0$, $d_m=d_0$ in Eq. (3), are satisfied in this case.

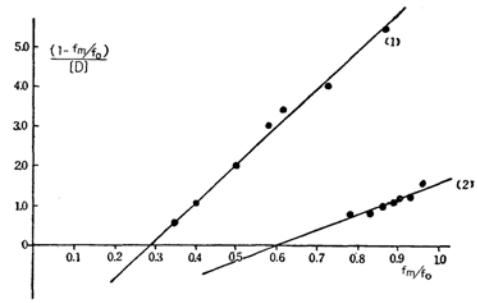

Fig. 7. $\frac{(1-f_m/f_0)}{[D]} \sim f_m/f_0$ relations for BDAA-aniline-benzene (1) and BDAA pyrrole-benzene (2) systems[b].
(b) (Conditions $d_m=d_0$ in Eq. (3) or $\varepsilon_c=\varepsilon_a$ in Eq. (2), are satisfied in these cases.)

Therefore, the association equilibrium in the excited state is practically the same as that in the ground state.

C. Effect of Solvent Viscosity on the Fluorescence Quenching.—If the diffusion controlled dynamical processes in the excited state such as the energy dissipation due to dissociation of hydrogen bond or

TABLE I

acceptor	donor	solvent	T	K_T	ϕ_c/ϕ_0
acridine	phenol	benzene	287	32	
BDAA	α-naphthol	〃	306	5.7×10^2	~0
	β-naphthol	〃	304	2.9×10^2	~0
	phenol	〃	300	3.7×10^2	~0
	aniline	〃	300	9.6	0.29
	pyrrole	〃	300	3.8	0.59
	phenol	n-hexane	294	1.3×10^3	0.02
	〃	cyclohexane	298	1.4×10^3	0.01
	〃	decalin	298	0.9×10^3	0.06
	〃	liquid paraffin	297	1.4×10^3	~0
DAA	phenol	benzene	286	6.3×10^2	~0
	aniline	〃	287	20	0.29

rotation around the hydrogen bond[6] are concerned with the non-radiative degradation of excited state, the ratio of the fluorescence yields of hydrogen bonded and free molecules, ϕ/ϕ_0, may show some dependence on the solvent viscosity. From such a viewpoint, we have examined the effect of solvent viscosity on the fluorescence quenching of BDAA by phenol, changing the solvent from n-hexane to liquid paraffin. The results obtained are shown in Fig. 8 and Table I.

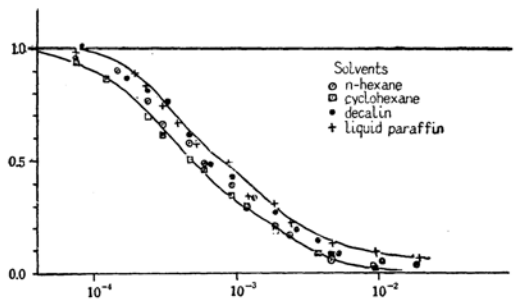

Fig. 8. Changes in relative fluorescence intensity of BDAA in various solvents of different viscosities by added phenol.

No viscosity dependence has been observed and the fluorescence is almost completely quenched by hydrogen bonding with phenol, in all the solvents examined. Therefore, no dynamical mechanism, in which the molecule moves as a whole, may prevail in the quenching of the present type.

Discussion

The results obtained are collected in the Table I. When hydrogen bond is isolated from π-electron system of donor molecule by σ-bond such as the case for benzylalcohol and β-phenylethylalcohol, the enhancement of fluorescence yield by the mechanism described previously[1], occurs. However, when hydrogen bond is intimately related with π-electron system, i. e. the conjugation of hydrogen bond with π-electron system is possible, such as the case for phenol, naphthols, aniline and pyrrole, the decrease in fluorescence yield was observed. Moreover, from the independence of quenching of the viscosity of the solvent, it is clear that any dynamical process in the excited state, in which the quencher molecule moves as a whole, does not concern the degradation of excited state. Therefore, it is most likely that the cause of non-radiative degradation may be ascribed to the interaction between π-electron systems via the hydrogen bond, i. e. the delocalization of π-electron through the hydrogen bond, in the excited state. An analogous mechanism in which the charge transfer interaction between excited anthracene and solvent molecule leads to the fluoresecnce quenching, was proposed by Bowen and West[7]. From the stand-point mentioned above, a merely electrostatic model of the hydrogen bond may not be appropriate.

A charge transfer model of hydrogen bond using molecular orbital and perturbation theory, was proposed by Nukasawa, et al.[8] and supported by Tsubomura on the basis of his detailed calculation[9] for the system O—H···O and also from the result of his infrared measurement[10]. From the viewpoint of the quenching phenomenon with which the present paper

6) G. Oster and Y. Nishijima, *J. Am. Chem. Soc.*, **78**, 1581 (1956).

7) E. J. Bowen and K. West, *J. Chem. Soc.*, **1955**, 4394.

8) K. Nukasawa, J. Tanaka and S. Nagakura, *J. Phys. Soc. Japan*, **8**, 792 (1953).

9) H. Tsubomura, This Bulletin, **27**, 445 (1954).

10) H. Tsubomura, *J. Chem. Phys.*, **23**, 2130 (1955); ibid., **24**, 927 (1956).

is concerned, the charge transfer model of hydrogen bond is more favored than the merely electrostatic one, especially in the excited state.

A similar phenomenon was also observed for the fluorescence quenching of naphthols and naphthylamines due to hydrogen bonding with pyridine, and this type of non-radiative degradation of the excited state due to the delocalization of π-electron through the hydrogen bond, may form a group in the quenching phenomena by complex formation.

Summary

1. Hydrogen bonding effect on the fluorescence of acridine, 3, 6-diaminoacridine and 3, 6-bisdimethylaminoacridine was studied in non-polar solvent added with proton donors such as phenol, naphthols, aniline and pyrrole.

2. Their fluorescence yields are decreased when hydrogen bonded with these proton donors, in which the hydrogen bond is capable of conjugation with π-electron system, in contrast to the case of donors such as benzylalcohol and β-phenylethylalcohol in which the hydrogen bond is isolated by σ-bond from π-electron system, and hydrogen bonding with which leads to the enhancement of fluorescence yield.

3. No viscosity dependence of quenching has been observed. From this fact participation of diffussion controlled dynamical process in the non-radiative degradation of excited state is unlikely.

4. The cause of non-radiative degradation of excited state by hydrogen bonding was ascribed to the delocalization of π-electrons through the hydrogen bond, especially in the excited state.

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