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# An ultraviolet absorption and fluorescence study of 1,10-phenanthroline and related compounds in aqueous solution

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# AN ULTRAVIOLET ABSORPTION AND FLUORESCENCE STUDY OF 1,10-PHENANTHROLINE AND RELATED COMPOUNDS IN AQUEOUS SOLUTION

by

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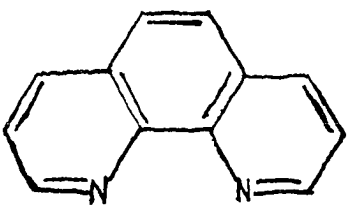
# I. LITERATURE REVIEW AND THEORY

## A. 1,10-Phenanthroline

### 1. Introduction

1,10-Phenanthroline and its derivatives have long been used as analytical reagents and have been extensively studied for this purpose. The structure of 1,10-phenanthroline is depicted below.



For convenience, this compound will be abbreviated 1,10-phen throughout the remainder of this text. Since the work presented in this thesis is not directly concerned with the application of 1,10-phen as an analytical reagent, no review of the extensive literature on this topic will be given. The interested reader is referred to a recent, comprehensive book on this topic by Schilt (65). The work presented in this thesis will be concerned with the acid-base chemistry of 1,10-phen in the ground and excited states, and the compound will principally be studied by ultraviolet absorption and fluorescence spectroscopy. The review of the literature will cover only those areas pertinent to the above mentioned topics.

### 2. Acid-base chemistry

Lee, Koltoff, and Leussing (44) have extensively investigated the reaction of 1,10-phen with the hydrogen ion. A pontentiometric titration

of 1,10-phen with hydrochloric acid indicated a 1:1 ratio of 1,10-phen to hydrochloric acid, while little change in potential was noted in the 1:2 region. They also performed a conductimetric titration which again indicated only a 1:1 adduct. Lee et al. concluded that, while 1,10-phen has two basic nitrogens, it behaves as a monoacidic base. They estimated the distance between the nitrogen atoms to be 2.5 Å and concluded that electrostatic and steric factors prevented the addition of a second hydrogen ion. It was later shown by Margerum et al. (47) from uv absorption spectra that a second proton can be added, however, this does not occur appreciably in solutions less acidic than 1M $HClO_4$. Therefore, the 1:2 protonated complex is of little or no concern in most situations.

It is convenient to represent the basicity of nitrogen heterocycles by the acid dissociation constant, $Ka_1$, defined by

$$Ka_1 = \frac{[H^+]\,[B]}{[HB^+]} \tag{1}$$

and

$$pKa_1 = pH + \log \frac{[HB^+]}{[B]} \tag{2}$$

Similarly, the expression for the second protonation is given by

$$Ka_2 = \frac{[HB^+]\,[H^+]}{[H_2B^{++}]} \tag{3}$$

and

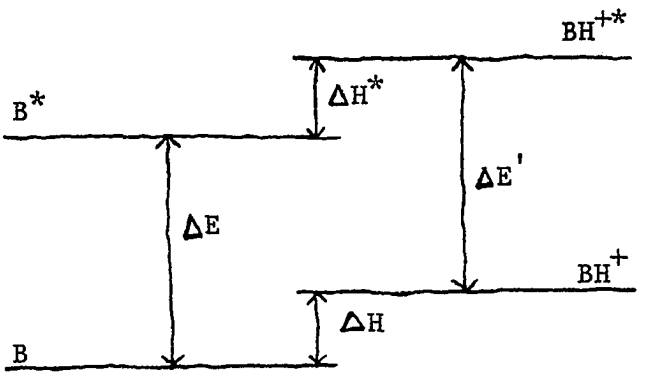
$$pKa_2 = pH + \log \frac{[H_2B^{++}]}{[HB^+]} \tag{4}$$

The $pKa_1$ for 1,10-phen has been measured many times by almost every conceivable technique. The results of some of these determinations are shown

in Table 1, which also lists several values of $pKa_2$. For the most part, the methods used are straightforward and the reader is referred to the references or any good advanced analytical textbook for the details.

Considerable work has been done on the determination of $pKa_1$ values for the substituted 1,10-phenanthrolines. Schilt and Smith (64) have determined the $pKa_1$ values for some forty substituted 1,10-phenanthrolines by pH measurements in dioxane-water mixtures and then extrapolating the values obtained to zero dioxane concentration. Brandt and Gullstrom (13) have applied the same technique to determine many of the values of the 5-substituted derivitives.

Excited state $pKa_1$ values can be determined from spectra by a method suggested by Weller (77). The ground and excited states of a base and its conjugate acid are depicted below.




B and $B^*$ = the ground state and the excited state of the base, respectively.

$BH^+$ and $BH^{+*}$ = the ground state and the excited state of the conjugate acid, respectively.

$\Delta E$ and $\Delta E'$ = difference in energy between the ground state and the excited state.

Table 1. The $pKa_1$ and $pKa_2$ values of 1,10-phenanthroline

| | $T^oC$ | $\mu$ | Method | Reference |
|---|---|---|---|---|
| $pKa_1$ | | | | |
| 4.77 | 25 | 0.0 | pH measurement with glass electrode | (44) |
| 4.86 | 25 | 0.003 | pH measurement with glass electrode | (64) |
| 4.92 | 25 | 0.1 | pH measurement with glass electrode | (79) |
| 4.857 | 25 | 0.1 | pH measurement with glass electrode | (56) |
| 4.95 | 20 | 0.1 | pH measurement with glass electrode | (1) |
| 4.92 | 20 | 0.2 | pH measurement with glass electrode | (39) |
| 4.96 | 25 | 0.001 | conductance measurement | (44) |
| 4.93 | 25 | 0.1 | absorption spectra of solutions of known pH | (15) |
| 4.85 | 20 | * | absorption spectra of solutions of known pH | (59) |
| 5.46 | 23 | 0.02 | absorption spectra of solutions of known pH | (45) |
| $pKa_2$ | | | | |
| -1.6 | 23 | | absorption spectra of solutions of known $H_o$** | (45) |
| -1.6(HCl) | * | | absorption spectra of solutions of known $H_o$** | (48) |
| -1.7($HClO_4$) | * | | absorption spectra of solutions of known $H_o$** | (48) |

* Not specified.

**$H_o$ = Hammet acidity function.

From the diagram,

$$\Delta E + \Delta H^{*} = \Delta E' + \Delta H \qquad (5)$$

Then

$$\Delta H - \Delta H^{*} = \Delta E - \Delta E' = (\Delta G - T\Delta S) - (\Delta G^{*} - T\Delta S^{*}). \qquad (6)$$

Assuming that $\Delta S = \Delta S^{*}$, the above expressions may be manipulated to give

$$pKa_1 - pKa_1^{*} = \frac{\Delta E - \Delta E'}{2.3RT} . \qquad (7)$$

The energy difference between the ground state and the excited state is estimated from spectra. Using the above method, Langmuir (42) obtained a value of 9.92 for $pKa_1^{*}$ for the lowest energy singlet state of 1,10-phen. The molecule is seen to be considerably more basic in the excited state, which is a general rule for nitrogen heterocycles. Jones (35) redetermined $pKa_1^{*}$ using an improved method for estimating the energy difference between the ground state and the excited state and obtained a value of 10.0. Using phosphorescence spectra, it is possible to determine $pKa_1^{*}$ for the lowest energy triplet state. Brinen et al. (14) have obtained a value of 6.7 for 1,10-phen. Again, 1,10-phen is seen to be more basic in an excited state than in the ground state.

The thermodynamic quantities, $\Delta G$, $\Delta H$, and $\Delta S$, have been determined by a variety of techniques for the neutralization reaction of 1,10-phen. Näsänen and Uusitalo (56) have measured the $pKa_1$ at 0°, 25°, and 50°C. From a plot of $pKa_1$ vs. $\frac{1}{T}$, $\Delta H$ and $\Delta S$ were found to be -3.5 Kcal mole$^{-1}$ and 10.2 e.u. Perkampus and Köhler (59) employed a spectrophotometric technique to obtain the $pKa_1$ as a function of temperature and found $\Delta G$ to be -6.23 Kcal mole$^{-1}$, and $T\Delta S$ to be 2.76 Kcal mole$^{-1}$ at 20°C. Using the same technique, Lahiri and Aditya (41) found $\Delta G$ = -6.9 Kcal mole$^{-1}$,

$\Delta H$ = -4.07 Kcal mole$^{-1}$, and $\Delta S$ = 9.5 e.u. Direct calorimetric measurements of the neutralization of 1,10-phen with 0.5 M $HNO_3$ in 1M $NaNO_3$ have been made by Kul'ba and Makashev (40). They found a $\Delta H$ = -4.6 Kcal mole$^{-1}$, $\Delta G$ = -6.4 Kcal mole$^{-1}$, and $\Delta S$ = 8 e.u.

Grimes (32) is apparently the first to suggest the possibility that more than one molecule of 1,10-phen associates with the hydronium ion. Considerable evidence was cited for the existence of poly(1,10-phen)-hydrogen(I) species. In solutions containing concentrations of acid and 1,10-phen greater than 0.02 M, larger concentrations of free hydrogen ion were present than were predicted from the $pKa_1$ value determined in dilute solution, which could be accounted for by the existence of the poly(1,10-phen)hydrogen(I) species. A ten-fold increase in the solubility of 1,10-phen over that expected for the formation of a simple conjugate acid was noted in solutions of increasing acid strength, which again could be accounted for by considering more than one molecule of 1,10-phen associated with each hydronium ion. In addition, a conductometric titration of a concentrated solution of 1,10-phen gave breaks at 1,10-phen to hydrochloric acid ratios of 2:1 and 1:1. Grimes (32) developed a Ag/Ag $(1,10\text{-phen})_2NO_3$ electrode that responds to the activity of 1,10-phen in much the same manner that a Ag/AgCl electrode responds to the chloride ion. Using this electrode, Grimes (32) determined the overall formation constants for these adducts defined by equation (20),

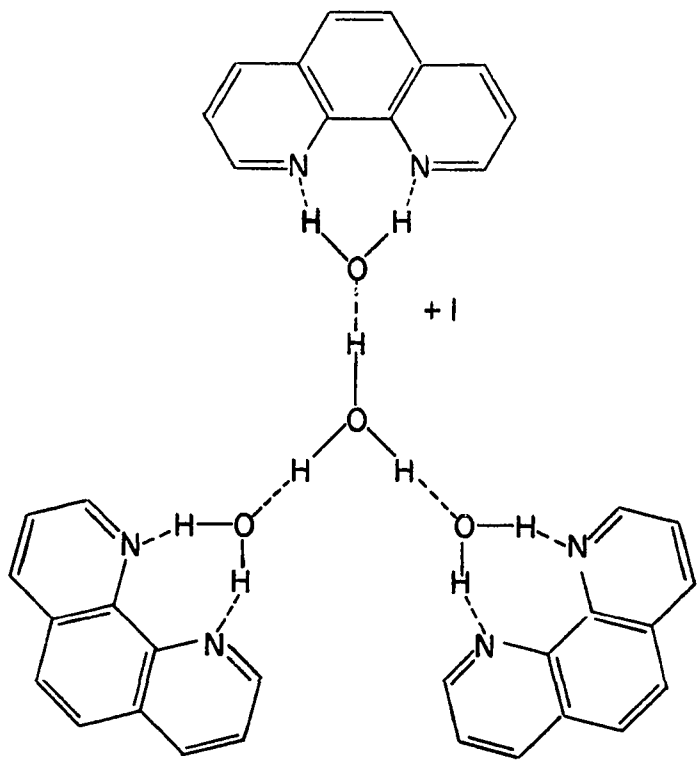
$$\beta_n = \frac{[HB_n^+]}{[H^+][B]^n}$$

where B represents the basic form of 1,10-phen. These are 5.05, 8.4, and

10.3 for the logarithm of $\beta_1$, $\beta_2$, and $\beta_3$, respectively. The constants indicate that these adducts do not form appreciably in dilute solutions, which may account for their not having been previously recognized. The constants for the poly(1,10-phen)hydrogen(I) adducts have been determined by several others, who perfected and further utilized the Ag/Ag(1,10-phen)$_2$-$NO_3$ electrode. Fullerton (31) obtained values of 5.27, 8.10, and 12.07 for the logarithm of $\beta_1$, $\beta_2$, and $\beta_3$, respectively. Fahsel and Banks (21), however, have published the best constants to date. Utilizing computer techniques and carefully controlling the ionic strength, they obtained values of 5.11, 7.22, and 9.03 for the logarithm of $\beta_1$, $\beta_2$, and $\beta_3$, respectively. Based on current concepts of the hydronium ion in aqueous solutions, Fahsel and Banks (21) proposed the structure depicted on the following page for the 3:1 adduct.

Langmuir (42) was the first to study the fluorescence spectrum of 1,10-phen in concentrated solutions. Langmuir reported a new band in the fluorescence spectrum with a dependence on pH and total 1,10-phen concentration that suggested the band was due to the poly(1,10-phen)hydrogen(I) species. The band appeared at 500 $m\mu$, which is shifted long wavelength to the fluorescence of the basic and monoprotonated form of 1,10-phen. Attempts to purify the 1,10-phen by recrystallization did not alter the intensity of the 500 $m\mu$ band, which ruled out the possibility of the fluorescence being due to an impurity. An increase in the molar absorptivity of the tailing edge of the long wavelength uv absorption band in concentrated solutions was also noted.

Jones (35) sought to further associate the appearance of the 500 $m\mu$ fluorescence with the poly(1,10-phen)hydrogen(I) aggregates. The


N
N
H
H
O
+1
H
O
H
H
N
H
O
O
H
N
H
H
N
N

intensity of the 500 $m_\mu$ fluorescence band was due to the poly(1,10-phen)-hydrogen(I) species. In addition, Jones (35) found that an identical 500 $m_\mu$ fluorescence band could be generated in concentrated 1,10-phen solutions in the presence of the ammonium ion at a pH of 8.3. At this pH no poly(1,10-phen)hydrogen(I) species exist in the ground state, and an explanation was sought which would account for the appearance of the 500 $m_\mu$ fluorescence in basic solution. The following experimental facts were noted by Jones (35).

1. The molar absorbtivity of the longwavelength uv absorption band is lower for a $1.2 \times 10^{-2}$ M 1,10-phen solution at a pH of 13.3 than it is for a $1.2 \times 10^{-3}$ M 1,10-phen solution at the same pH.
2. The intensity of the fluorescent band at 500 $m_\mu$ increases with increasing 1,10-phen concentration.
3. The intensity of the 500 $m_\mu$ fluorescence is similarly dependent on ammonium ion concentration.
4. The percent quenching of the 1,10-phen fluorescence by the ammonium ion increases with increasing 1,10-phen concentration.

Based on these observations, Jones (35) proposed the following mechanism to account for the appearance of the 500 $m_\mu$ fluorescence in basic solution.

$$
\begin{aligned}
&\text{(a)}\quad B + B \longrightarrow (BB) \\
&\text{(b)}\quad B + h\nu \longrightarrow B^* \\
&\text{(c)}\quad (BB) + h\nu \longrightarrow (BB)^* \\
&\text{(d)}\quad (BB)^* \longrightarrow (BB) + h\nu' \\
&\text{(e)}\quad B^* \longrightarrow B + h\nu' \\
&\text{(f)}\quad (BB)^* + H_2O \longrightarrow (BBH^+) + OH^- \\
&\text{(g)}\quad (BBH^+)^* \longrightarrow (BB) + H^+ + h\nu''
\end{aligned}
\qquad (8)
$$

B = 1,10-phen.
$h\nu'$ = 365 $m_\mu$ fluorescence.
$h\nu''$ = 500 $m_\mu$ fluorescence.

Jones proposed a ground state dimeric species to account for fact one. The formation of the dimer is seen in step (a) of the proposed mechanism. The dimer is seen to be responsible for the 500 $m_\mu$ fluorescence in steps (f) and (g). The formation of the dimer depends on 1,10-phen concentration, accounting for fact two. Fact three is accounted for by the catalytic effect of the ammonium ion on step (f). Weller (76) has shown the ammonium ion to be an effective catalyst in excited state protonations. The dimer is seen to fluoresce with the same spectral distribution as 1,10-phen--steps (d) and (e). Since the dimer is dependent on 1,10-phen concentration, the quenching of the 365 $m_\mu$ fluorescence by the ammonium ion would also show a concentration dependence, accounting for fact four. Both the work of Jones (35) and Langmuir (42) will require some critical comment which will be discussed in detail in the main body of the thesis.

## B. Fluorescence Quenching

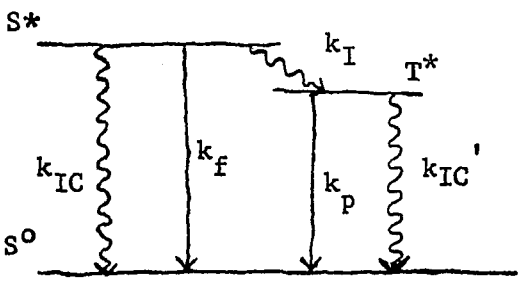
### 1. Introduction

Because of a growing interest in the chemistry of excited states and the increasing availability of good instrumentation, the fluorescence literature has been growing exponentially. It would not be practical to review the entire fluorescence literature or to give a complete description of fluorescence processes. The reader is referred to several excellent books on the topic for an introduction to this field. The books by Pringsheim (61) and Förster (25) are old but are considered classics in the field. A more recent work by Hercules (34) is also an excellent

introduction. Only the portions of the fluorescence literature which are pertinent to the development of this thesis will be reviewed. These are fluorescent quenching processes and excited state aggregation. In addition, the discussion will pertain principally to absorption and fluorescence in aromatic hydrocarbons.

## 2. Intrinsic quenching

A large part of the information contained in the following discussion is taken from Hercules (34). When a typical aromatic hydrocarbon in the ground state is excited by absorption of a photon, it will rapidly undergo conversion to the lowest vibrational level of the excited singlet state of lowest energy, regardless of the singlet state to which it was originally excited. This is due to the efficiency of vibrational relaxation in solution and to the high degree of vibrational coupling between excited singlet states. The latter fact is due to the relatively small energy separation between excited singlet states. This is a general rule for aromatic hydrocarbons in solution at moderate temperatures, and there are few exceptions. This process occurs in approximately $10^{-13}$ to $10^{-11}$ seconds. Once a molecule arrives in the lowest energy excited state, there are several possibilities for returning to the ground state. The simplified diagram depicted below will serve to identify these processes.



$S^o$ = ground state.

$S^*$ = excited singlet state of lowest energy.

$T^*$ = excited triplet state of lowest energy.

$k_I$ = rate constant for intersystem crossing from the singlet to the triplet state.

$k_f$ and $k_p$ = rate constants for the fluorescence and phosphorescence transitions, respectively.

$k_{IC}$ and $k_{IC}'$ = rate constants for internal conversion for the singlet and triplet states, respectively.

The fluorescence transition occurs from the $S^*$ state to the $S^o$ state by the emission of a photon. The $S^*$ state generally has a natural lifetime of $10^{-9}$ to $10^{-7}$ seconds, which imposes certain kinetic restrictions on the deactivation of this state. If a process is to compete with the fluorescence process, it must have a rate which is greater than or equal to the spontaneous decay of this state.

While the processes competing with the fluorescence transition are subject to environmental perturbations, they are considered intrinsic quenching processes because they are principally a function of the electronic structure of the molecule. The following processes will fall under that classification.

The intersystem crossing process, denoted by $k_I$ in the diagram, is a radiationless process which occurs by vibrational coupling. This is a spin forbidden process which occurs in approximately $10^{-8}$ to $10^{-7}$ seconds. In general, the efficiency of the process increases with decreasing energy difference between the singlet state and the triplet state. The rate of this process is seen to be of the same order as the fluorescence process and it competes effectively with it. Internal conversion is a

process whereby a molecule in an excited state returns to the ground state without the emission of a photon. The energy is presumably converted to heat. In general, internal conversion is a difficult quantity to measure and the process is little understood.

The $T^* \longrightarrow S^o$ transition is the phosphorescence transition. At room temperature this transition is not usually observed. The characteristically long lifetime of the $T^*$ state, due to the forbiddenness of the $T^* \longrightarrow S^o$ transition, leads to a high probability of collisional deactivation in solution at room temperature. For this reason, phosphorescence is usually observable only at low temperatures in rigid media.

The following discussion will consider processes competing with the fluorescence process which are a function of the environment of the molecule. For this reason, these processes will fall under the general classification of extrinsic quenching.

3. Solvent quenching

It is a well known fact that the fluorescence quantum efficiency of an aromatic hydrocarbon is highly dependent on the solvent in which it is dissolved. Because solvent quenching is of no particular importance to the development of this thesis, no lengthy discussion will be given of this topic. The reader is referred to the review by Van Duuren (72) for a more complete discussion.

Bowen et al. (10, 12) have extensively studied solvent quenching and concluded that it occurs through the formation of an intermediate encounter complex. They propose the following mechanism,

$$\begin{aligned} &\text{(a)} \quad A + h\nu \longrightarrow A^* \\ &\text{(b)} \quad A^* + S \longrightarrow (A^-S^+) \longrightarrow A + S \end{aligned} \qquad (9)$$

where S is a solvent molecule and $A^*$ is a potentially fluorescent molecule. The intermediate, $(A^- S^+)$, is seen to be charge transfer in nature. One can see that greater solvent quenching is predicted for solvents of Lewis base character.

Solvents containing "heavy-atoms" can also quench fluorescence by promoting intersystem crossing, which leads to increased phosphorescence at the expense of fluorescence (49). The mechanism for this process is also believed to involve charge transfer (50). It is evident that solvents such as carbon tetrabromide and ethyl iodide are probably not suitable for sensitive fluorescence studies, but their use may well be advantageous in a phosphorescence study.

Fluorescent molecules which are capable of hydrogen bonding are subject to another kind of solvent effect. The relative fluorescence yields of quinoline in water, ethanol, and benzene are 1000:30:1, respectively (72). This is due to a reordering of the electronic states in the molecule (34). The fluorescence transition in benzene is $\pi^* \longrightarrow n$, where n designates the nonbonding electrons on the nitrogen. The long lifetimes and smaller singlet-triplet energy split associated with this transition results in an increased probability of intersystem crossing with a resulting decrease in fluorescence yield. In water and ethanol, the electrons on the nitrogen are strongly solvated, which raises the energy of the $\pi^* \longrightarrow n$ transition so that the lowest energy transition is now $\pi^* \longrightarrow \pi$. The $\pi^* \longrightarrow \pi$ transition is a fully allowed transition, and results in high fluorescence yields in water and ethanol.

The addition of a foreign solute to a solution containing a fluorescent molecule can lead to several types of quenching which may be

divided into two classifications, collisional and noncollisional.

4. Noncollisional quenching

Energy transfer between a potentially fluorescent molecule in an excited state with another molecule which is at a distance of 50 to 100 Å is known as noncollisional quenching. The process is a resonance energy transfer and is not to be confused with the trivial process of absorption of the fluorescence of one molecule by another. The process was apparently first studied and interpreted by Förster (24), who investigated the quenching of the trypaflavine fluorescence by rhodamine B. The transfer of the excitation energy was estimated to occur over a distance of 70 Å and was found to be independent of the viscosity of the medium. The latter fact serves to distinguish this quenching mode from collisional quenching. Watson and Livingston (75) were also early to recognize this quenching mode in chlorophyll and give an excellent discussion of it in their paper.

5. Collisional quenching

Collisional quenching is a diffusion controlled process which depends on the contact between a potentially fluorescent molecule in an excited state and the quencher. This process has been shown to obey the Stern-Volmer law (67),

$$\frac{I^o}{I} = 1 + k_1 [Q] \qquad (10)$$

where $I^o$ is the fluorescence intensity in the absence of the quencher, I is the intensity at quencher concentration Q, and $k_1$ is the quenching constant. While this simple equation adequately describes the quenching process mathematically, the physical process is quite complicated.

Approximately 50-100 Kcal $mole^{-1}$ are being dissipated and a simple collision does not adequately account for this energy loss (34). The quenching is believed to occur either through an enhancement of intersystem crossing or through electron transfer. Umberger and LaMer (71) have derived from diffusion theory the following expression for the quenching constant in solution,

$$k_1 = \frac{8RT}{3000\eta}\sqrt{1 + \frac{2r}{\tau^\circ D}}\ \tau^\circ \qquad (11)$$

where,

T = absolute temperature,
R = gas constant in ergs $deg^{-1}$ $mole^{-1}$,
r = radius of the molecules in cm.,
$\eta$ = viscosity in poise,
$\tau^\circ$ = mean lifetime of the excited state in the absence of Q in sec,
D = diffusion coefficient in $\frac{cm^2}{sec}$.

The equation as written assumes the quencher and fluorescer are uncharged and of equal molecular size. In addition, the assumption is made that every collision is effective which is generally not true. The reader is referred to the original paper for details on these points. The importance of this equation is that it indicates the important variables in collisional quenching. Debois and Van Hemert (20) have used this equation to estimate the mean lifetime of some fourteen organic compounds dissolved in n-hexane with reasonable success. Their work indicates that to a first approximation the quenching constant can be estimated by the term

$$\frac{8RT}{3000\eta}\ \tau^\circ .$$

The effect of ionic strength on the quenching constant has been studied by Stoughton and Rollefson (70). The results of this study

indicate that the dependence of the quenching constant on ionic strength is in the direction and of the order of magnitude predicted for an ordinary bimolecular reaction. Deviations from the Stern-Volmer law have been studied by Boaz and Rollefson (9). Positive deviations are attributed to the presence of a quencher molecule in the same solvent cage as the fluorescing molecule in the ground state, increasing the probability of quenching. Quenching equations were developed which account for this behavior. It is evident from this work (9) and the work of Umberger and LaMer (71) that conclusions drawn from considerations of the Stern-Volmer law are valid only at low quenching.

The effect of more than one quencher on the fluorescence intensity has been studied by Stevens and Dubois (68). The quenching law for two quenchers is given by,

$$\frac{I^o}{I} = 1 + k_1 [Q_1] + k_2 [Q_2] , \qquad (12)$$

where the notation is the same as that stated for the Stern-Volmer law.

It is common for the quantum efficiency of many aromatics to decrease in solutions more concentrated than $10^{-4}$ to $10^{-3}$ $\underline{M}$ (33). This is often due to collisions between a potentially fluorescent molecule and a molecule of the same kind in the ground state (self-quenching). Collisional self-quenching that is diffusion controlled obeys the Stern-Volmer law cited above (33). Melhuish (53) has studied the effect of self-quenching on quantum efficiencies and has determined the self-quenching constants for a large number of aromatics from the Stern-Volmer law. The analytical implications of self-quenching are that the use of

fluorescence as a method is restricted to dilute solutions. As the collision between an excited molecule and one of the same kind in the ground state can lead to the formation of dimeric species which are important to the development of this thesis, this topic will be discussed in detail in the following section.

## C. Ground and Excited State Aggregation in Aromatic Organic Molecules

### 1. Excited state aggregates

The collision between an aromatic molecule in an excited state and a molecule of the same kind in the ground state can result in several different kinds of spectral behavior depending on the strength of the interaction. If the strength of the interaction is particularly strong, a stable dimer may form. The absorption spectrum of the monomer decreases with time with a concurrent appearance of a dimer spectrum. The fluorescence spectrum is unchanged, but concentration quenching occurs. This type of behavior is typical of anthracene (11, 19).

Another type of behavior, which appears to be more common, is the formation of a dimer which dissociates upon loss of the excitation energy. This type of dimer is known as an excimer (excited state dimer) and was first studied by Förster and Kasper (29). A review of the work of Förster and Kasper (29) will serve to characterize the spectral behavior exhibited by excimers. As the molar concentration of pyrene is increased above $10^{-4}$ M, a structureless, blue fluorescence band appears, which is red shifted with respect to the violet, structured band characteristic of monomeric pyrene. The intensity of the blue emission is

proportional to the square of the pyrene concentration, and the appearance of the blue emission is accompanied by concentration quenching of the monomer fluorescence. The absorption spectrum is independent of concentration, indicating that the species responsible for the emission exists only in the excited state. A mechanism consistent with these data is depicted below:

$$\begin{aligned} &\text{(a)}\quad M + h\nu \longrightarrow M^* \\ &\text{(b)}\quad M^* + M \longrightarrow D_2^* \\ &\text{(c)}\quad M^* \longrightarrow M + h\nu' \\ &\text{(d)}\quad D^* \longrightarrow M + M + h\nu'' \end{aligned} \qquad (13)$$

where,

$M$ = ground state monomer,
$M^*$ = excited state monomer,
$D^*$ = excimer (excited state dimer),
$h\nu'$ = monomer fluorescence, and
$h\nu''$ = excimer fluorescence.

Since the work of Förster and Kasper (29) in 1955, excimer fluorescence has been detected for a large number of aromatics, including many common ones such as benzene, toluene, and naphthalene. Birks and Christophorou (6, 7) have compiled a listing of a large number of compounds exhibiting excimer fluorescence. The lack of an excimer fluorescence band does not necessarily indicate that an excimer does not exist. The excimer may thermally dissociate before emission takes place. In this case, lowering the temperature will allow the fluorescence process to compete with dissociation, resulting in the appearance of the excimer fluorescence band. This behavior is exhibited by naphthalene (28) and phenanthrene (4).

An additional characteristic of excimer fluorescence has been pointed out by Birks et al. (8), which has become known as the 6000 $cm^{-1}$ rule. This rule is the experimental observation that the energy separation

between the 0-0 band of the monomer fluorescence (highest energy transition) and the peak of the excimer fluorescence is approximately 6000 $cm^{-1}$ for all known excimers. Various attempts have been made to explain the 6000 $cm^{-1}$ rule, but none have been entirely satisfactory. Birks et al. (8) have postulated that the 6000 $cm^{-1}$ rule indicates a common type of excimer interaction, probably charge transfer. Using a simple Huckel molecular orbital treatment, Azumi and Azumi (3) have indicated that the 6,000 $cm^{-1}$ phenomenon could arise if the intermolecular distance between monomers is constant. This treatment assumes the structure of the dimers is a "sandwich" configuration with the planes of the two monomers parallel. It should be pointed out that excimers are generally believed to be "sandwich" dimers.

The theoretical approaches to the bonding in excimers originally proceeded along two different lines. The first was by Ferguson (22) in 1958, who indicated that charge transfer is the predominate source of bonding energy in excimers. This proposal was based on energy considerations and the lack of vibrational structure in excimer fluorescence, even at low temperatures. Förster (26, 27) later proposed a molecular exciton theory, which accounts for the bonding energy by a resonance interaction of the transition moments of the two monomers (dipole-dipole interaction). Azumi and McGlynn (4) have pointed out that neither the charge transfer theory nor the molecular exciton theory adequately accounts for excimer bonding energies. Based on a correlation between the predicted charge transfer bonding energy and excimer fluorescence energy, they concluded that charge transfer is important in excimer bonding, but the energies of excimer formation predicted by charge transfer are

too great. Negative results were obtained in an attempt to correlate the energies of excimer formation predicted from the molecular exciton theory. Azumi and McGlynn (4) conclude that both charge transfer and exciton states must be included in meaningful calculations and proposed calculations based on the configuration interaction of charge transfer and exciton states. Subsequent calculations by Azumi, Armstrong, and McGlynn (2) indicate that this hypothesis is generally correct. The calculations reproduce experimental energies of excimer formation at appropriate interplanar distances.

The calculations in this treatment are prohibitive, however, which limits the usefulness of them. In addition, certain inconsistencies arise as pointed out by Azumi and Azumi (3) and implied by Ferguson (23). Nevertheless, they are important in understanding the nature of the forces involved. It is perhaps sufficient comment that a simple and admittedly inexact molecular orbital treatment by Azumi and Azumi (3) yields results which are very close to the more exact configuration interaction treatment.

## 2. Ground state aggregation

The ground state aggregation of aromatic organic compounds in solution is a rather common phenomenon. The formation of dimeric, trimeric, and higher polymeric species of dyes is well-known. Among the more common dyes reported to form dimers in solution are Thionine and methylene blue (62), fluorescein (43), and rhodamine B (30). The bonding in the dimeric species is generally believed to be due to London dispersion forces (27, 62). As London dispersion forces are a function of the polarizability of

a molecule, it can be seen why dyes are more susceptible to ground state dimerization than are aromatics such as benzene and naphthalene. Upon dimerization, the absorption spectra of dyes generally show a splitting of the intense long wavelength band into a short wavelength and a long-wavelength component with respect to the monomer band. The dimers of dyes, with a few exceptions, are non-fluorescent (43, 30), which results in concentration quenching of the fluorescence. McRae and Kasha (51) have discussed this point and have attributed the non-fluorescence of these dimers to increased intersystem crossing.

The dimerization of molecules capable of hydrogen bonding to each other, such as carboxylic acids, is well-known and requires no detailed discussion. The interested reader is referred to the discussion by Pauling (58). The current theory accounting for the change in absorption spectra upon dimerization is the molecular exciton theory, which has been reviewed by Kasha (37). The theory is general in that it applied to dimers, trimers, nonconjugated polymers, and crystals. Moreover, it is applicable to a variety of bonding situations, i.e., it applied hydrogen bonded aggregates as well as dispersion force dimers. It should be emphasized, however, that the theory applies only to loosely bonded aggregates, i.e., the electrons associated with each monomeric unit remain localized after dimerization and excitation. The molecular exciton model is based on a resonance interaction between excited states, in other words, a delocalization of the excitation energy between two monomeric units of a dimer. The strength of the exciton interaction in the dimer depends on the magnitude of the transition dipoles and their geometric relationship in the monomeric units, and inversely on the cube

of the intermolecular distance. For strongly coupled exciton states, expected for intense transitions and short intermolecular distances, the theory predicts complete splitting of the monomeric absorption band. In the case of weak coupling, the spectral band shape remains intact and second-order effects, such as increases and decreases in band intensity, are observed.

The dye molecules discussed previously will serve to illustrate the application of the theory. For a dye molecule in a "sandwich" configuration, the transition dipoles can be aligned repulsive, ↑ ↑ , and attractive, ↑ ↓ . This results in two exciton states, one of high energy (dipoles aligned repulsive), and one of low energy (dipoles aligned attractive). Thus one would predict the spectrum of the monomer band to be split into a high energy and a low energy component corresponding to transitions to the two exciton states, which is experimentally observed. Other geometric configurations are discussed in the review by Kasha (37). Mention should be made of the fact that the transition to one of the states is forbidden which may result in one of the bands not appearing in the spectrum or appearing with low intensity.

# II. PURPOSE

The usefulness of 1,10-phen as a colorimetric reagent has been clearly demonstrated and is so well-known that it requires no further comment. More recently, 1,10-phen has been applied to fluorescence analysis. Veening and Brandt (73) have developed a sensitive and selective method for ruthenium based on the fluorescence of the tris(5,methyl-1,10-phen)ruthenium(II) complex. Fluorescence methods using 1,10-phen and its derivatives have also been developed for some of the rare earths (66) and copper (35). In addition, iridium and osmium (36) have been reported to form fluorescent complexes with 1,10-phen. It is evident that more analytical applications involving 1,10-phen are forthcoming, and that a study of the spectral behavior of 1,10-phen would be a valuable and indespensable addition to this development.

Because the spectral behavior of 1,10-phen has previously been investigated by Jones (35) and Langmuir (42), some justification for reopening this investigation is required. The following points are evident upon reviewing the previous studies on this compound:

1. The studies by Jones (35) and Langmuir (42) employed apparent fluorescence spectra, i.e., the spectra were not corrected for the varying response to intensity with wavelength of the photomultiplier tube, gratings, and other instrumental variables. Depending on the spectral region and the instrument used, an apparent spectrum can be a seriously distorted version of the true fluorescence spectrum. While apparent spectra may be of analytical value, theoretical interpretations of them are

obviously not meaningful.

2. As the absorption spectrum serves to define the chemistry of the ground state of a molecule, it is a necessary and logical starting point for fluorescence investigations. The necessary absorption data for the poly(1,10-phen)hydrogen(I) system are lacking. The data available are, at best, fragmentary.
3. The mechanism proposed by Jones (35) to account for the appearance of poly(1,10-phen)hydrogen(I) fluorescence spectra in basic solution requires a ground state dimer, indicated by changes in the absorption spectrum, which fluoresces at the same spectral distribution as the monomer. This implies strong coupling in the ground state dimer and very weak coupling in the excited state dimer. This behavior is opposite that expected for a compound similar to 1,10-phen. Several more likely explanations for the behavior of 1,10-phen were not pursued by Jones (35).

In light of the above points, it is evident that a reinvestigation of the poly(1,10-phen)hydrogen(I) system is warranted.

While the spectral behavior of aromatic hydrocarbon has been extensively investigated, very little has been done with their heterocyclic analogs. For example, the possibility of excimer fluorescence in nitrogen heterocycles in solution has not been investigated. It is hoped that an investigation of 1,10-phen and related compounds would be a valuable addition to the excited state chemistry of nitrogen heterocycles.

In summary, this work will encompass an uv absorption and fluorescence study of 1,10-phen and related compounds in the hope that this study will contribute both to the development of analytical reagents for

use in fluorescence analysis and to a better understanding of excited state chemistry.

# III. AN INVESTIGATION OF THE GROUND STATE AGGREGATION OF 1,10-PHENANTHROLINE IN BASIC SOLUTION

## A. Introduction

The purpose of these experiments is to determine whether or not the ground state dimer proposed by Jones exists and, if so, to determine its formation constant. The evidence presented by Jones (35) consists principally in a decrease in the molar absorptivity of the long wavelength transition of a $1.2 \times 10^{-2}$ M 1,10-phen solution when compared to a solution $1.2 \times 10^{-3}$ M in 1,10-phen. The evidence presented by Jones (35) is considered inadequate because it does not indicate the behavior of 1,10-phen in the intermediate concentrations where the existence of the dimer was invoked to account for the unusual quenching behavior exhibited by the ammonium ion on the 1,10-phen fluorescence. Obviously, if the dimer is to be invoked to account for the unusual fluorescence quenching behavior, its existence must be demonstrated at the same concentrations at which the unusual quenching behavior is exhibited.

In addition, vapor pressure osmometry will be employed to determine whether 1,10-phen dimerizes at higher concentrations than can be obtained in aqueous solution where the solubility is rather low ($\sim$0.016 M). The method takes advantage of the fact that a solution exposed to a vapor of the pure solvent will attain a higher temperature, owing to the exothermic condensation of the solvent on the solution. The rate of condensation is proportional to the molal concentration of the solute in the solvent and essentially measures the vapor pressure lowering of the solvent by the solute. The method is convenient for determining molecular

weights and aggregation in organic solvents. A pair of matched thermistors, one containing solution and the other containing solvent, are employed to determine the temperature change by measuring the change in resistance. The experimental details and thermodynamic considerations of the technique have been discussed by Meeks and Goldfarb (52). The interested reader is referred to this paper for a more complete discussion.

## B. Experimental

### 1. UV absorption measurements

Absorption data were taken with the Carey Model 14 and Carey Model 16 spectrophotometers. The high molar absorptivity of 1,10-phen, coupled with the relatively high concentrations used, required very short path lengths. This was accomplished by inserting a 0.995-cm or a 0.990-cm quartz slug in a 1-cm quartz cell.

### 2. Vapor pressure measurements

Vapor pressure measurements were obtained with the Thomas Isothermal Molecular Weight Apparatus, Model 12, following directions supplied by the manufacturer. The temperature of the compartment was thermostated at 30.4°C.

### 3. Reagents

1,10-phen was obtained from the Aldrich Chemical Co. The 1,10-phen used for the vapor pressure measurements was titrated by conductometric titration with standard hydrochloric acid and found to be 100% pure within the experimental error of the titration. The 1,10-phen used for the

absorption measurements was recrystallized from hot water as the monohydrate and air-dried at room temperature. The water content was determined by thermogravimetric analysis. Bibenzyl was obtained from Eastman Organic Chemicals and recrystallized from ethanol. The product melted at 51 to 52°C. The literature value is 52.5°C. The benzene was Spectro Grade, obtained from Eastman Organic Chemicals and dried over sodium metal.

4. Solution preparations

The solutions for both the uv absorption data and the vapor pressure measurements were prepared by making appropriate dilutions from concentrated stock solutions. The pH of the aqueous solutions was adjusted to 12 by adding an aliquot of KOH. At this pH all of the 1,10-phen is in the basic form.

5. Miscellaneous

Class A glassware was used throughout these experiments. The temperature of the aqueous solutions was 23° to 25°C.

## C. Results and Discussion

The results of the uv absorption study in basic solution are shown in Figures 1 and 2. The wavelengths chosen in relationship to the uv spectrum of 1,10-phen are indicated in Figure 8. The absorbances at all wavelengths are seen to comply with Beer's law up to about $1 \times 10^{-2}$ M. Above $1 \times 10^{-2}$ M, a slight deviation is seen in all of the plots. In addition, the complete spectra for these solutions do not indicate any shifts in wavelength up to $1 \times 10^{-2}$ M. This fact and the compliance with

Figure 1. Beer's law plots of 1,10-phenanthroline in basic solution

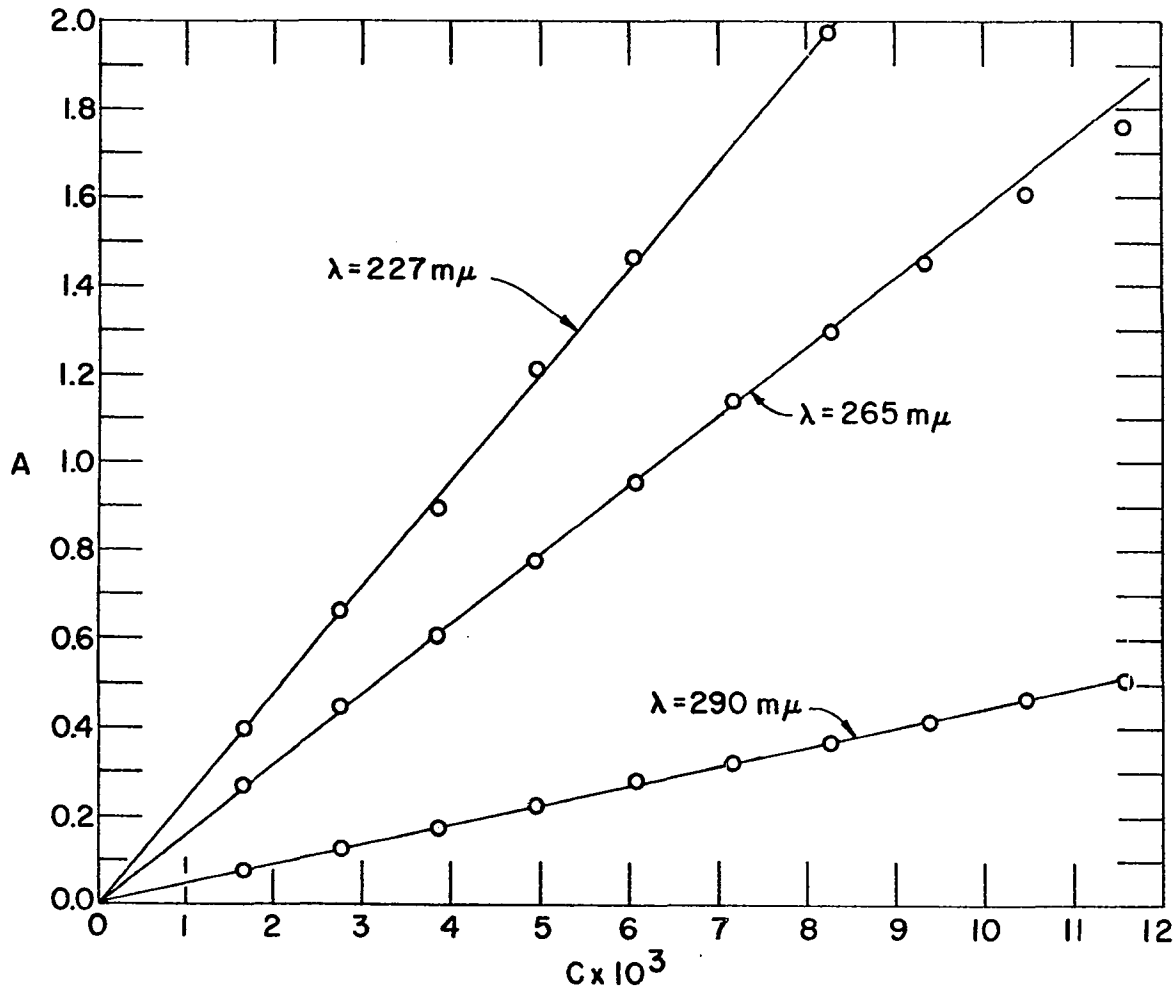
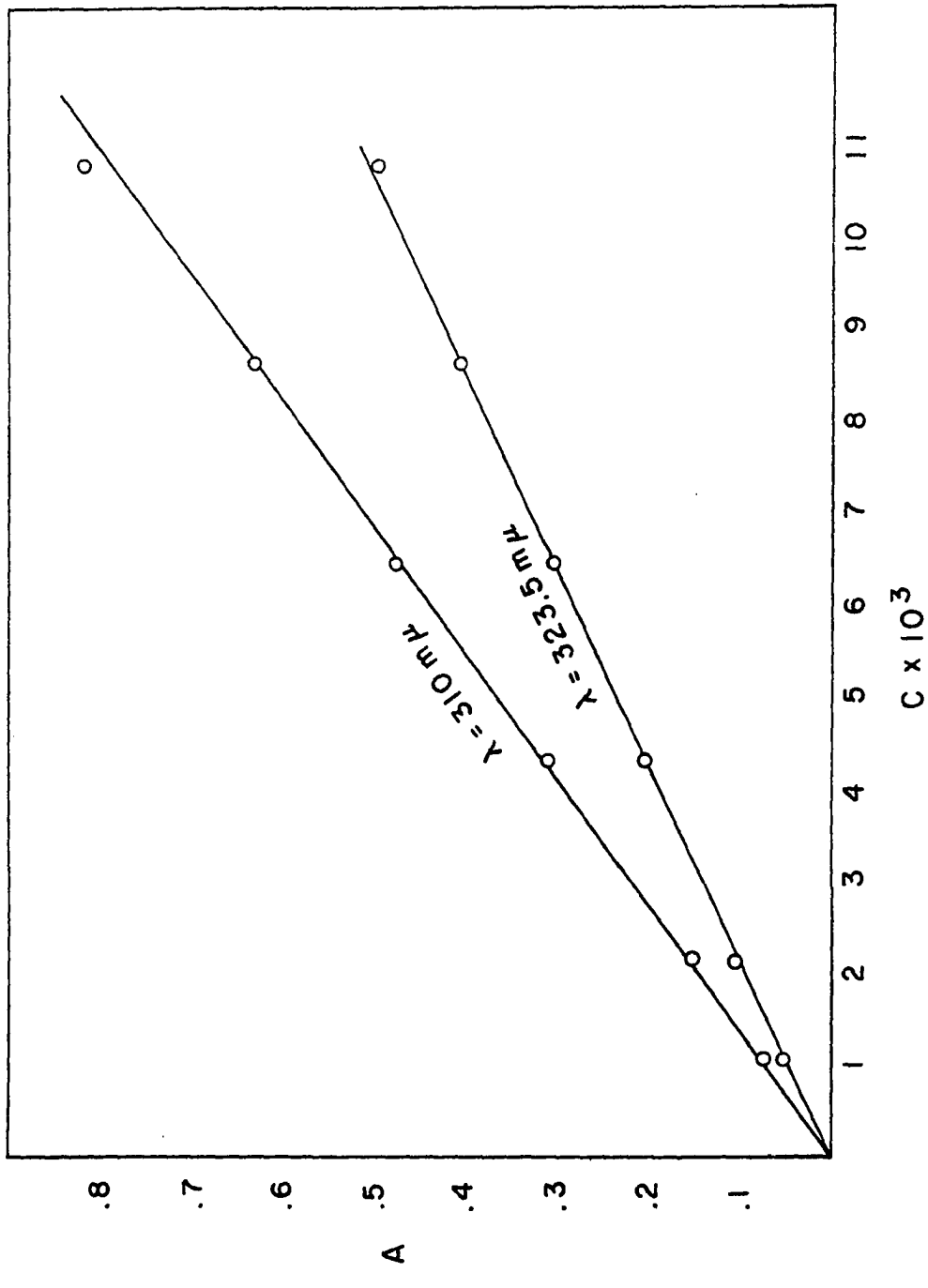

2.0
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
A
λ=227mμ
λ=265 mμ
λ=290 mμ
0
1
2
3
4
5
6
7
8
9
10
11
12
C x 10^3

Figure 2. Beer's law plots of 1,10-phenanthroline in basic solution

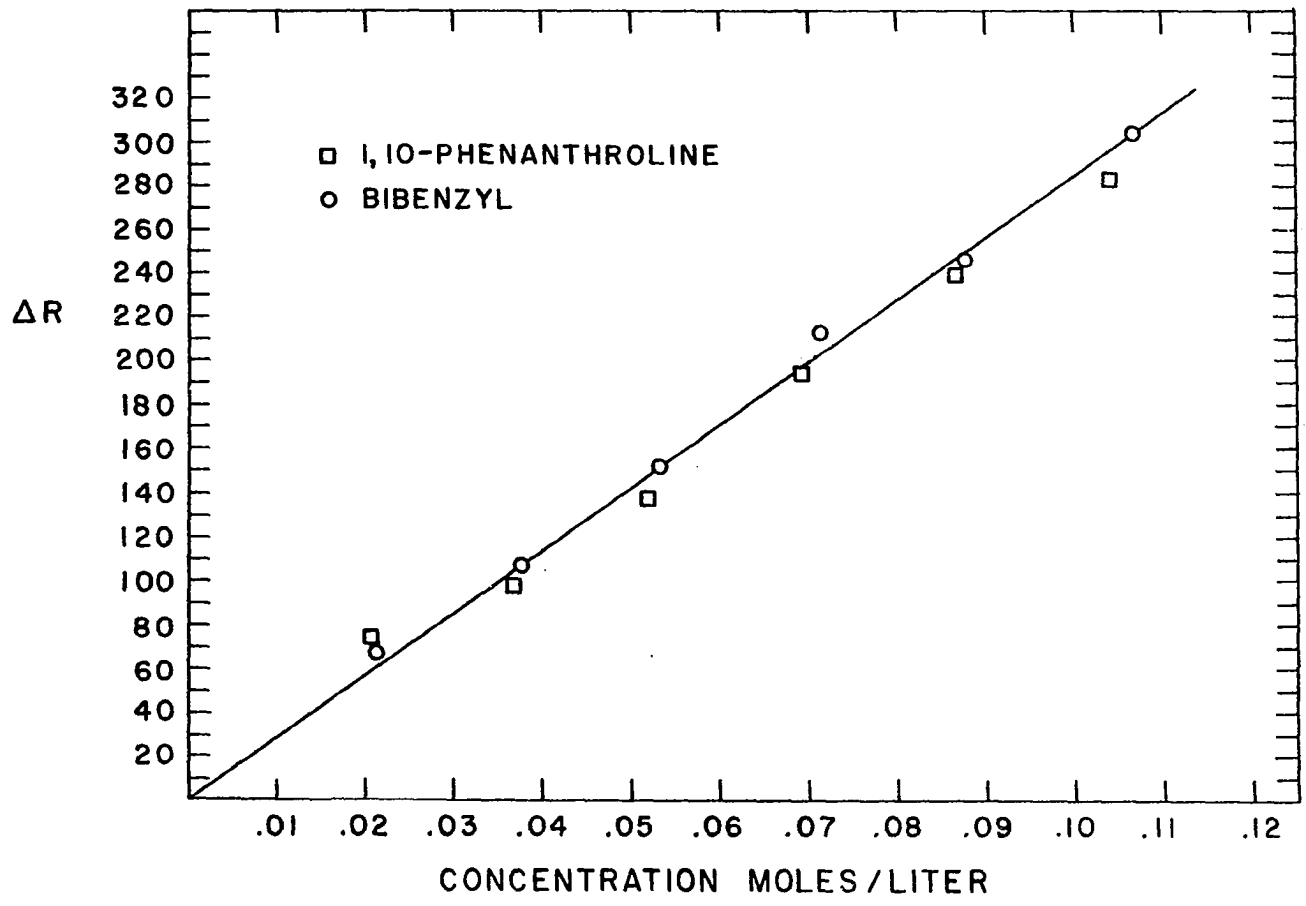

λ = 310 mμ
λ = 323.5 mμ
A
C x 10^3
.1
.2
.3
.4
.5
.6
.7
.8
1
2
3
4
5
6
7
8
9
10
11

Beer's law are taken to indicate that no dimer exists in aqueous solution in this region. Above $1 \times 10^{-2}$ M there is some evidence for the aggregation proposed by Jones (35), which is perhaps not unexpected as $1.0 \times 10^{-2}$ M is beginning to approach the molar solubility of 1,10-phen in water ($\sim 1.6 \times 10^{-2}$ M). It is not possible from the data taken to define the nature of the aggregation. It may be in the form of dimers, trimers, higher order aggregates, or, for that matter, absorption of the compound on the walls of the quartz cell, which is not uncommon for aromatics in solution. The concentration range over which the deviation occurs is small and the spectral shifts insignificant, all of which would make it difficult to investigate the system, and the results would be of little significance.

The results of the vapor pressure lowering by 1,10-phen in benzene are shown in Figure 3. Bibenzyl is used as a standard because it is expected to be monomeric in benzene, which is indicated in the work of Neumayer (57). The $\Delta R$ is a relative change in the resistance of the thermistor containing the solution and is proportional to the vapor pressure lowering of the solvent by the solute. These data indicate there is no inherent tendency for 1,10-phen to dimerize even at concentrations an order of magnitude greater than those employed in aqueous solution. These data also tend to indicate that the deviations from Beer's law are related to the approaching solubility limit of 1,10-phen in aqueous solution. Similar experiments in isopropanol also indicated that dimers are not formed in this solvent.

It is evident that the dimer proposed by Jones (35) does not exist at concentrations that would account for the quenching behavior exhibited

Figure 3. $\Delta R$ as a function of bibenzyl and 1,10-phenanthroline concentration


ΔR
320
300
280
260
240
220
200
180
160
140
120
100
80
60
40
20
□ 1,10-PHENANTHROLINE
○ BIBENZYL
.01
.02
.03
.04
.05
.06
.07
.08
.09
.10
.11
.12
CONCENTRATION MOLES/LITER

by the ammonium ion on the 1,10-phen fluorescence or the appearance of the 500 $m_\mu$ fluorescence in basic solution. The increase in percentage quenching by the ammonium ion and the appearance of the 500 $m_\mu$ fluorescence begin at $1 \times 10^{-3}$ M, and there is clearly no evidence for dimerization at this concentration. An alternate explanation will be required which is taken up in Chapter V of this thesis. The investigation of the poly(1,10-phen)hydrogen(I) system is also simplified by these data, because no concern need be given for dimeric species in an already complicated system.

# IV. AN UV ABSORPTION AND FLUORESCENCE STUDY OF THE POLY(1,10-PHENANTHROLINE)HYDROGEN(I) AGGREGATES

## A. Introduction

This work will entail a fluorescence and uv absorption study of the poly(1,10-phen)hydrogen(I) system reported by Fahsel and Banks (21). The objectives of this study are to obtain spectral evidence for these species and to illucidate the nature of the bonding involved. As fluorescence spectra reflect the chemistry of the lowest energy excited state and absorption spectra reflect the chemistry of the ground state, two proposals are required because the chemistry of these states is generally quite different.

## B. Experimental

### 1. Absorption spectra

Absorption spectra were obtained with a Carey Model 14 spectrophotometer. Various path lengths were achieved by inserting an appropriate quartz slug in a 1-cm quartz cell. The spectrophotometric titration was accomplished with a Beckman Model DU spectrophotometer in a 180-ml tall-form beaker.

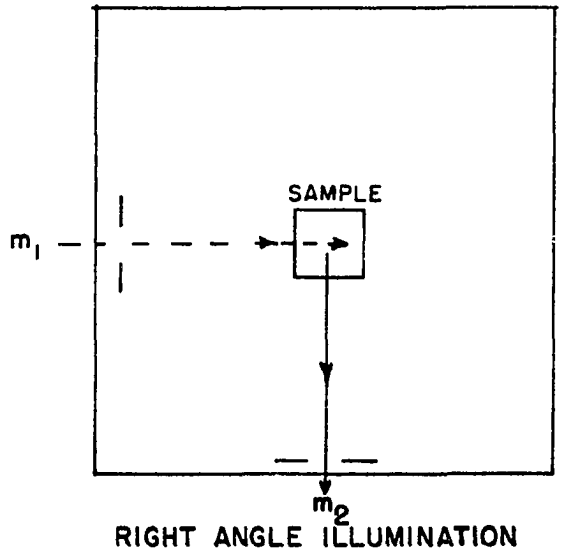
### 2. Fluorescence spectra

Fluorescence spectra were obtained with an Aminco-Bowman Spectrophotofluorimeter equipped with a Mosely X-Y recorder and an RCA 1P28 photomultiplier tube. Slit arrangement 3 was used for all fluorescence spectra.

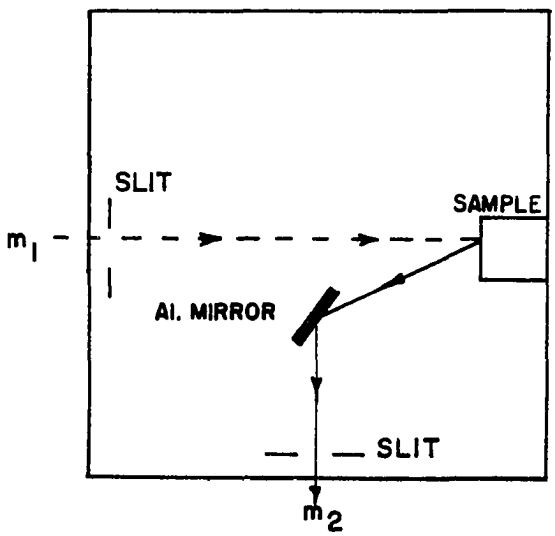
The two methods of illumination used in this work are illustrated in Figure 4. Frontal illumination was used in these experiments. The short path length used for the concentrated spectra was achieved by inserting a slug in a 1-cm cell. This gave a path length of $1.5 \times 10^{-3}$ cm.

All spectra were corrected for the varying response to intensity with wavelength of the photomultiplier tube, gratings, and mirrors. The method of Melhuish (53) was used to calibrate the instrument. An excellent discussion of the practical aspects of this method has been given by Chen (17). The method employs rhodamine B as a quantum counter to obtain the spectral distribution of the excitation source, which is a xenon arc. A concentrated solution of rhodamine B in ethlene glycol (3 gms/liter) is placed in the frontal illumination apparatus and its fluorescence at 640 $m_{\mu}$ is monitored as a function of the excitation wavelength. Because the quantum yield of rhodamine B is independent of the excitation wavelength and all of the intensity is absorbed in a thin layer at the surface of the cell, the fluorescence is proportional to the number of quanta incident on the solution. The fluorescence therefore reflects the relative spectral distribution of the xenon arc. The emission of the arc is then reflected into the emission monochrometer and photomultiplier tube at 10 $m_{\mu}$ intervals with a mirror. After correcting for the reflectance of the mirror, the relative intensity of the arc is divided by the response of the emission monochrometer and phototube. This gives the correction factor for a given wavelength. The reflectance data for the mirror were determined by Mr. Bruno Schmidt of this Laboratory. The spectra were also corrected for the absorbance of a glass filter which was used to reduce scattered light from the emission

Figure 4. Methods of illumination for obtaining fluorescence spectra. $m_1$ is the excitation monochrometer. $m_2$ is the emission monochrometer




RIGHT ANGLE ILLUMINATION




FRONTAL ILLUMINATION

unit at the shorter wavelengths.

3. Solution preparation

Solutions of 1,10-phen were prepared by an appropriate dilution from a stock solution. The variation of pH at a constant 1,10-phen concentration was accomplished by the addition of concentrated $H_2SO_4$ and KOH to a 1,10-phen solution with a micropipet or a capillary tube, withdrawing aliquots at appropriate pH values. The volumes of acid and base required are negligible with respect to the total volume. The pH measurements were made with a Beckman Expanded Scale pH Meter. This method allowed for the adjustment of pH without the addition of salts and buffers which might affect the fluorescence spectra. Because unbuffered solutions are prone to change pH on standing, the solutions were used as soon as possible after preparation.

The HCl used for the spectrophotometric titration was standardized with $Na_2B_4O_7 \cdot 10H_2O$, prepared by the method suggested by Vogel (74).

4. Reagents

The 1,10-phen used for the concentrated absorption spectra was obtained from Aldrich Chemical Co. and analyzed by conductometric titration. The material was found to be 100% pure within the experimental error of the titration. Subsequent uv absorption and fluorescence spectra of this material confirmed the analysis. The 1,10-phen used for the dilute solution absorption spectra was obtained from the Aldrich Chemical Co. and recrystallized from $H_2O$. The water content of the air-dried product was determined by thermogravimetric analysis. Rhodamine B was obtained from Allied Chemical and Dye Corp. and recrystallized from $H_2O$.

5. Miscellaneous

The computer program used to calculate the formation constants was written with the help of R. Bowers of this Laboratory. The temperature of the solutions was 23° to 25°C in all experiments. Class A glassware was used in all experiments.

## C. Results and Discussion

The constants of Fahsel and Banks (21) were used to compute the concentrations of the various poly(1,10-phen)hydrogen(I) species as a function of pH and total 1,10-phen concentration. The computer drawn plots are shown in Figures 5, 6, and 7. The poly(1,10-phen)hydrogen(I) species are seen to exist at total 1,10-phen concentrations greater than approximately $1 \times 10^{-3}$ M and in the pH range of 3.5 to 7.

The absorption spectra of dilute solutions of 1,10-phen as a function of pH are shown in Figure 8. This concentration of 1,10-phen would correspond to the behavior predicted by the formation constants in Figure 5. The isosbestic points at 267 $m_\mu$ and in the 290 $m_\mu$ region are indicative of two species present, the basic form and its conjugate acid, which correlates with the behavior predicted by the formation constants. The absorption spectra for the concentrated solutions are shown in Figure 9. These spectra correspond to the behavior predicted by the formation constants in Figure 7. Significant concentrations of the poly-(1,10-phen)hydrogen(I) species are seen to exist under these conditions. The most noticeable effect the poly(1,10-phen)hydrogen(I) species have on the absorption spectra is the loss of the isosbestic points as predicted by the formation constants. Overall, the effect of these species

Figure 5. The 1,10-phenanthroline system as a function of pH. □ = basic form. O = acidic form

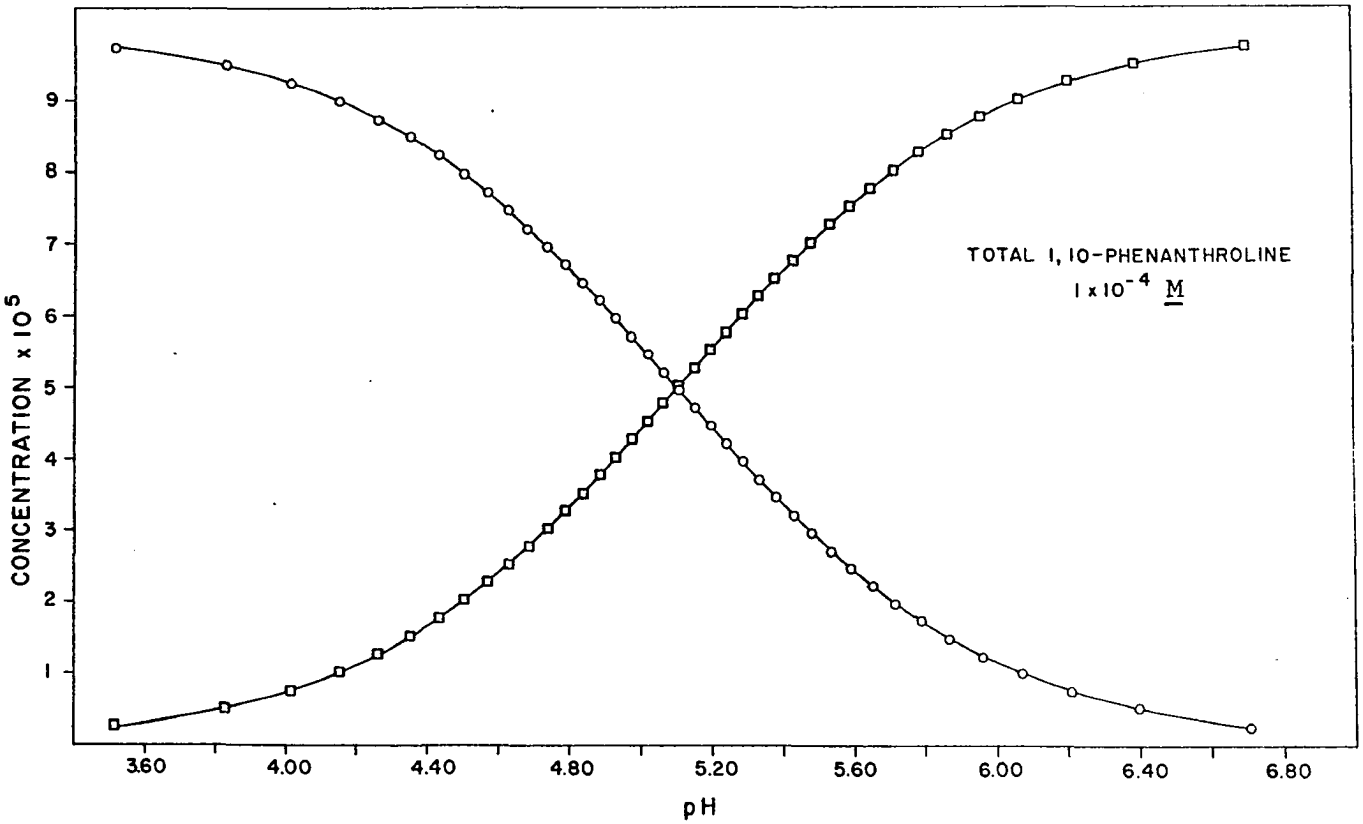

CONCENTRATION x 10^5
9
8
7
6
5
4
3
2
1
3.60
4.00
4.40
4.80
5.20
5.60
6.00
6.40
6.80
pH
TOTAL 1, 10-PHENANTHROLINE
1 x 10^-4 M

Figure 6. The 1,10-phenanthroline system as a function of pH. X = basic form. O = acidic form. $\Delta$ = bis(1,10-phenanthroline)hydrogen(I). + = tris(1,10-phenanthroline)-hydrogen(I)

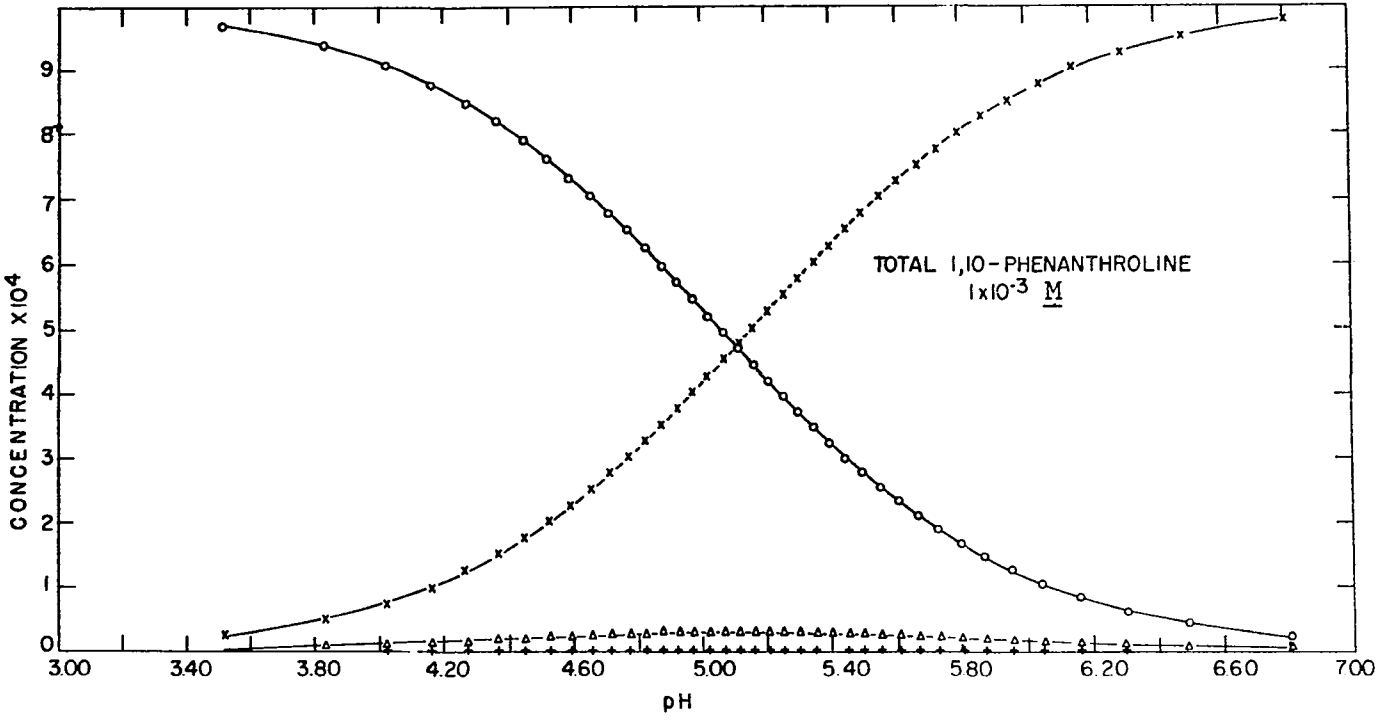

TOTAL 1,10-PHENANTHROLINE
1x10-3 M
CONCENTRATION X10⁴
pH
0
1
2
3
4
5
6
7
8
9
3.00
3.40
3.80
4.20
4.60
5.00
5.40
5.80
6.20
6.60
7.00

Figure 7. The 1,10-phenanthroline system as a function of pH. X = basic form. 0 = acidic form. Δ = bis(1,10-phenanthroline)hydrogen(I). □ = tris(1,10-phenanthroline)-hydrogen(I)

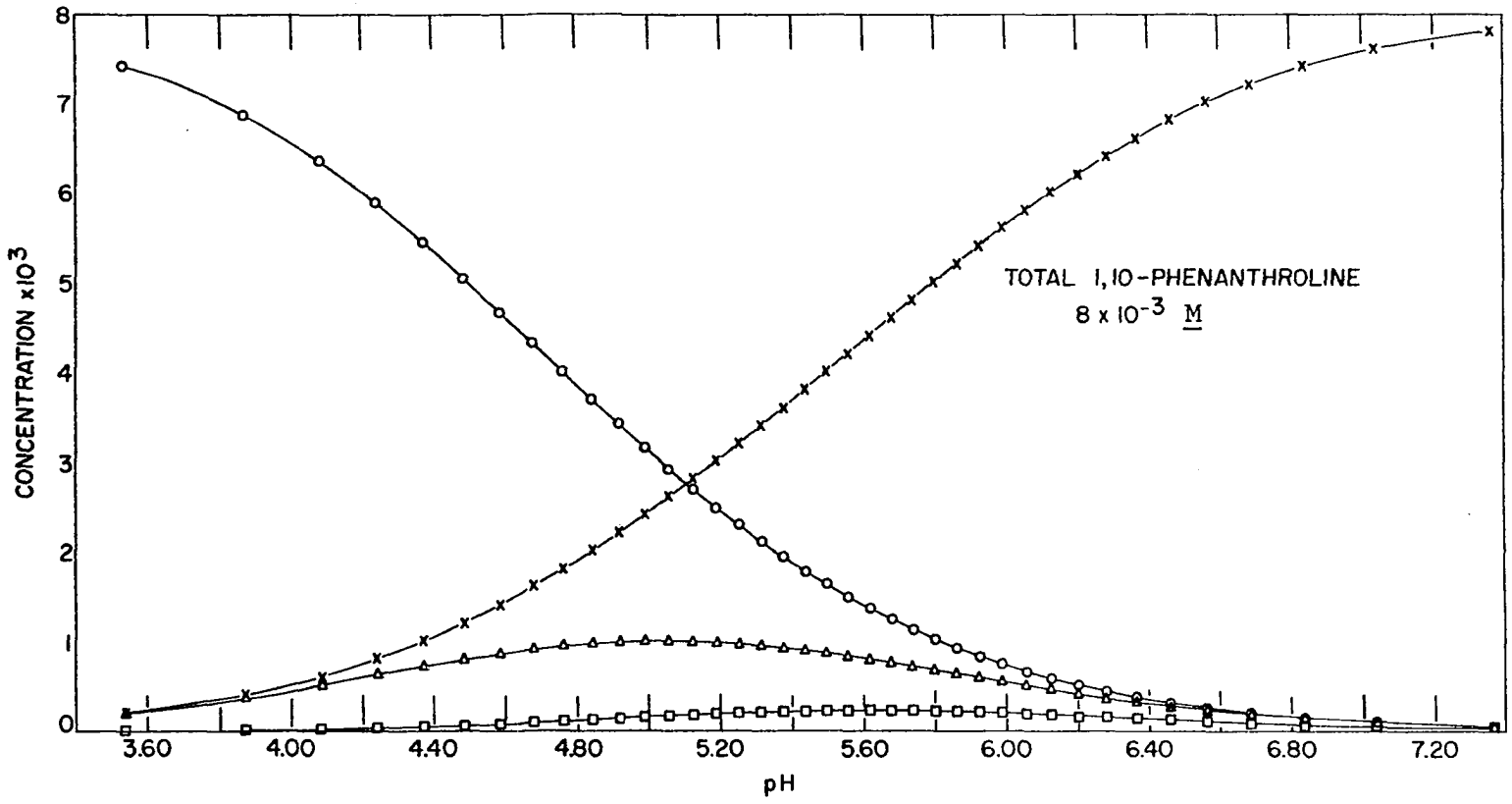

CONCENTRATION x10³
8
7
6
5
4
3
2
1
0
3.60
4.00
4.40
4.80
5.20
5.60
6.00
6.40
6.80
7.20
pH
TOTAL 1,10-PHENANTHROLINE
8 x 10⁻³ M

Figure 8. The uv absorption spectra of 1,10-phenanthroline as a function of pH. The total concentration of 1,10-phenanthroline is $2.55 \times 10^{-4}$ M

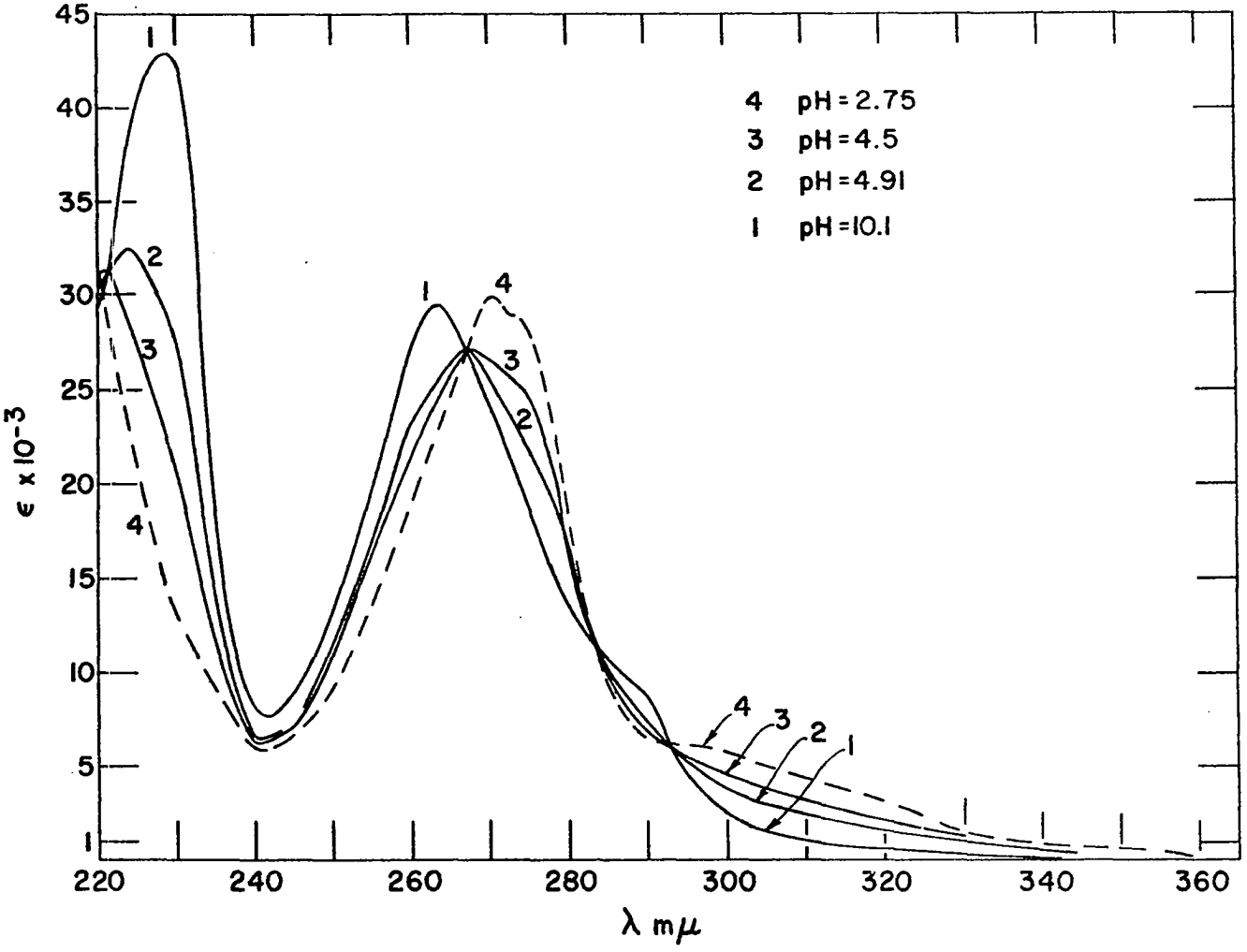
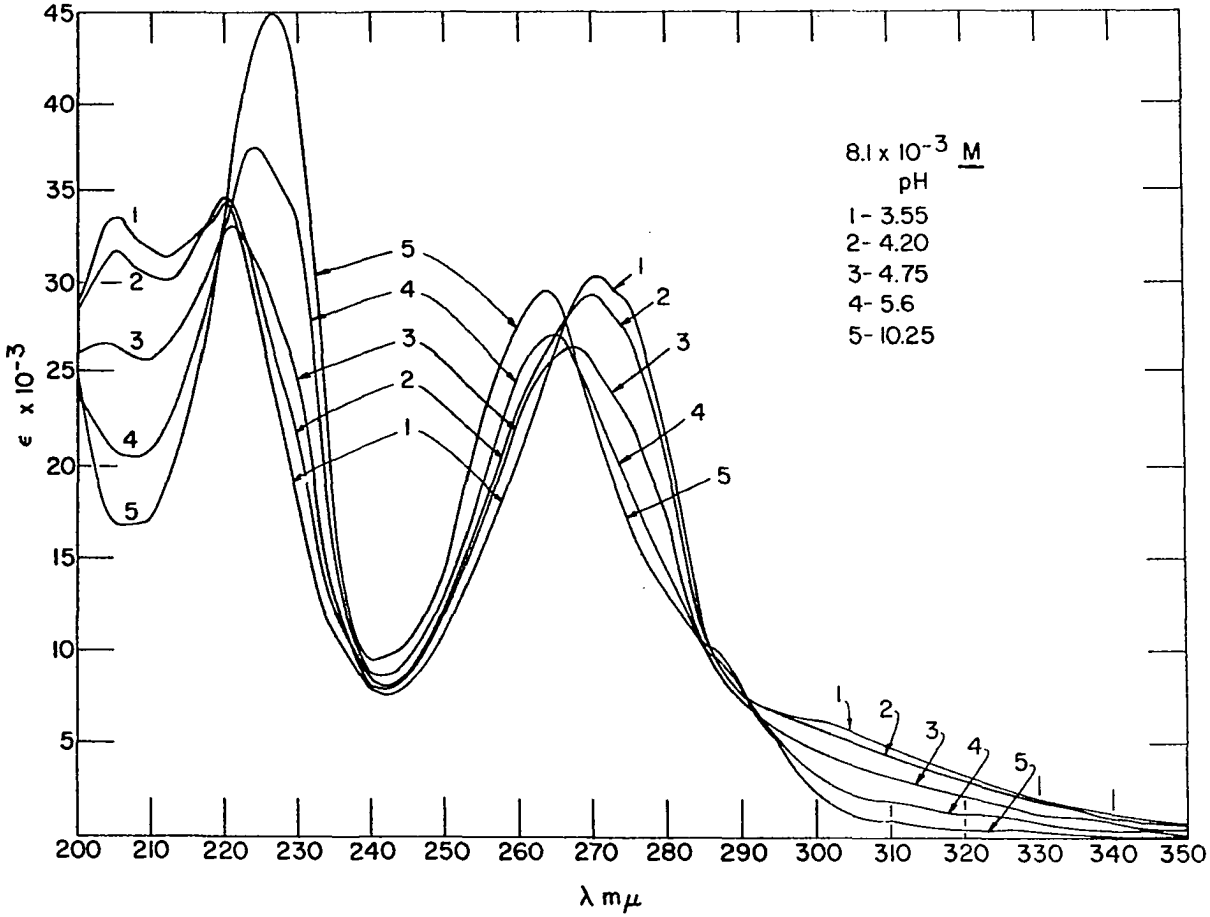

4 pH = 2.75
3 pH = 4.5
2 pH = 4.91
1 pH = 10.1
ε x 10⁻³
λ mμ
45
40
35
30
25
20
15
10
5
1
220
240
260
280
300
320
340
360

Figure 9. The uv absorption spectra of 1,10-phenanthroline as a function of pH. The total 1,10-phenanthroline concentration is $8.1 \times 10^{-3}$ M

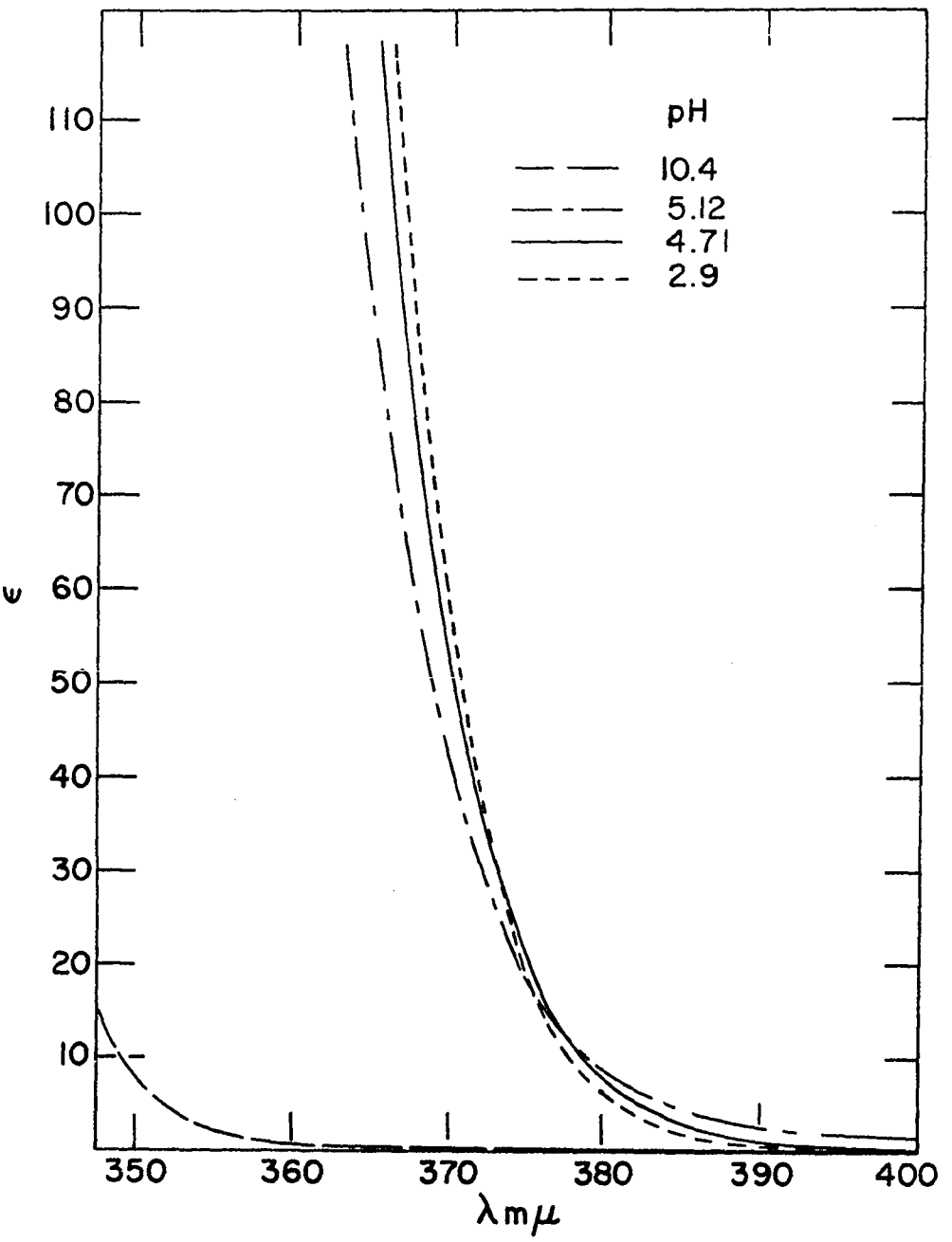

8.1 x 10⁻³ M
pH
1 - 3.55
2 - 4.20
3 - 4.75
4 - 5.6
5 - 10.25
ε x 10⁻³
λ mμ
45
40
35
30
25
20
15
10
5
200
210
220
230
240
250
260
270
280
290
300
310
320
330
340
350

on the absorption spectra of 1,10-phen is small. No band splitting or significant spectral shifts are observed and, as in the dilute solution spectra, the longest wavelength transition is exhibited by the acidic form. This is also indicated in Figure 10, which are the spectra of concentrated solutions obtained with a longer path length to exaggerate the tailing edge.

In the formalism of the exciton theory, this spectral behavior would best be described as the weak coupling case. This is consistent with the structure proposed by Fahsel and Banks (21) which places 1,10-phen molecules in the secondary hydration sphere of the hydronium ion. A structure with 1,10-phen molecules occupying the primary hydration sphere would seem unlikely for steric reasons. Since the structure proposed by Fahsel and Banks places emphasis on hydrogen bonding in the aggregates, the phenomenon of aggregate formation ought to be entirely general, i.e. nitrogen heterocycle capable of forming a reasonably strong hydrogen bond with water ought to form aggregates of the type described for 1,10-phen. This topic will be pursued in other sections of the thesis.

As indicated previously, there is an increase in tailing in the absorption spectra containing the poly(1,10-phen)hydrogen(I) aggregates, which is shown in Figure 10. This is probably due to band broadening in the spectra of the aggregates. At 385 $m_{\mu}$ the molar absorptivity of the solutions containing aggregates is seen to become larger than that of the acidic form. As this appeared to be the most distinctive feature of the aggregate spectrum, an attempt was made to obtain spectral evidence for the poly(1,10-phen)hydrogen(I) species by a spectrophotometric titration. A 0.0138 M 1,10-phen solution was titrated with a 0.3418 N

Figure 10. The uv absorption spectra of 1,10-phenanthroline as a function of pH. The total 1,10-phenanthroline concentration is $8 \times 10^{-3}$ M


pH
10.4
5.12
4.71
2.9
110
100
90
80
70
60
50
40
30
20
10
ε
350
360
370
380
390
400
λmμ

HCl solution at 385 $m\mu$. The results of this titration are shown in Figure 11. The extrapolated straight line portions of the titration curve indicate species at approximately 2.5:1 and 1:1 1,10-phen to HCl ratios. The lack of distinct breaks at 3:1 and 2:1 are attributed to the coexistence of these species over a fairly wide pH range as indicated in Figure 7 and to the relatively small formation constants for these aggregates. In addition to providing spectral evidence for the poly-(1,10-phen)hydrogen(I) aggregates, the titration curve indicates that the spectral behavior that has been discussed is related to these species.

The fluorescence spectra of dilute solutions of 1,10-phen as a function of pH are seen in Figure 12. At this pH only the basic form and its conjugate acid are predicted by the formation constants as indicated in Figure 5. The spectra are consistent with this prediction as only two bands are seen, the basic form at 365 $m\mu$ and its conjugate acid at 415 $m\mu$. The spectra for the concentrated solutions, corresponding to the situation described by the formation constants in Figure 7, are shown in Figure 13. A new band is seen in these spectra at approximately 490 $m\mu$, along with the loss of the isosbestic point. The dependence on pH and total 1,10-phen concentration suggest that this band is due to the poly(1,10-phen)hydrogen(I) aggregates. Because no band shifts of this magnitude are seen in the absorption spectra, it must be concluded that the aggregates undergo a rearrangement after excitation. Red-shifted bands of this type are common for aromatic hydrocarbons and are characteristic of excimer species. In the case of loosely bound aggregates, such as the poly(1,10-phen)hydrogen(I) species, all that would be required for excimer fluorescence would be for two of the 1,10-phen molecules in

Figure 11. A spectrophotometric titration of 1,10-phenanthroline at 385 $m\mu$. The absorbances are corrected for dilution by the titrant

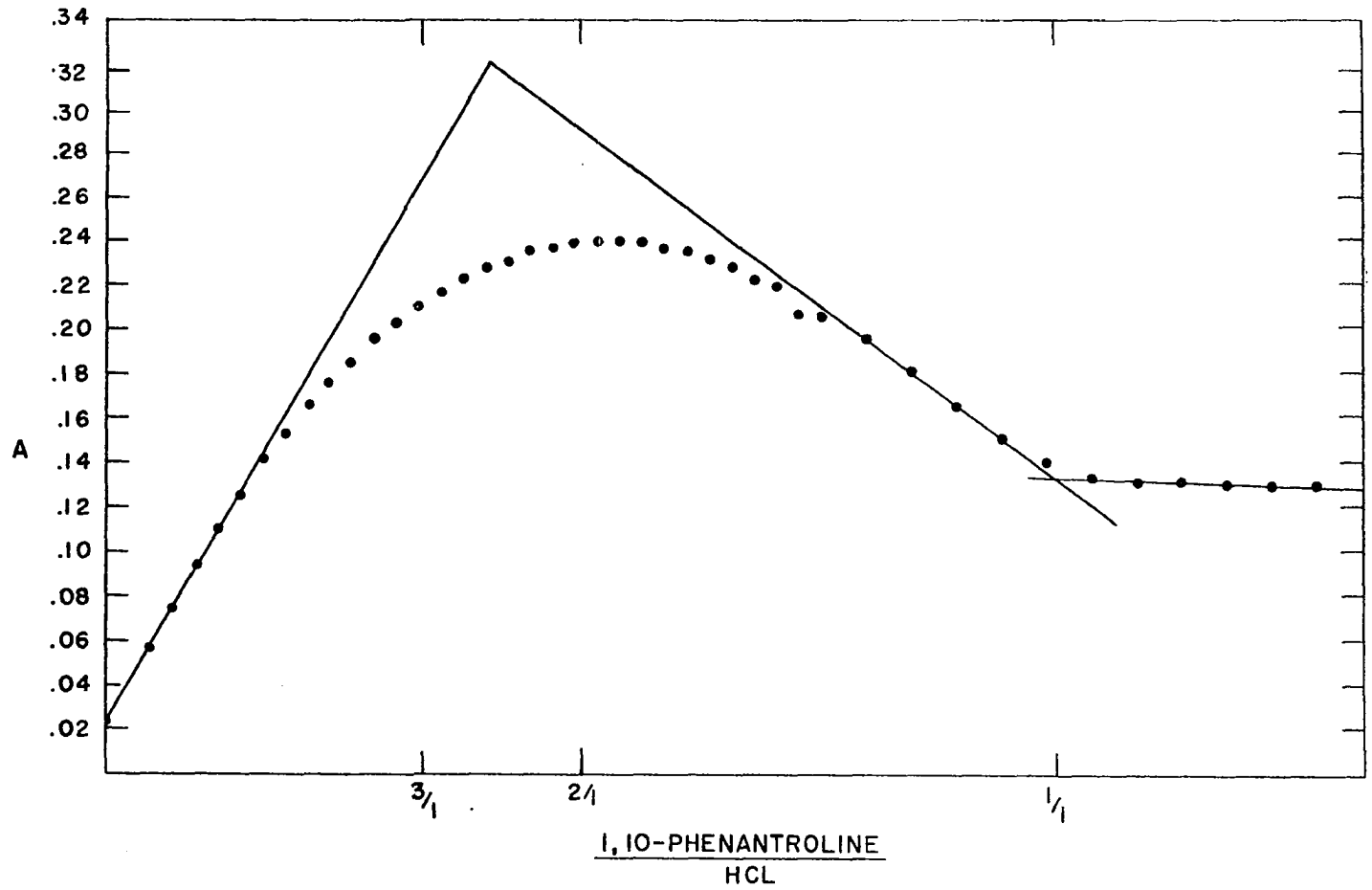

.34
.32
.30
.28
.26
.24
.22
.20
.18
.16
.14
.12
.10
.08
.06
.04
.02
A
3/1
2/1
1/1
1, 10-PHENANTROLINE
HCL

Figure 12. The fluorescence spectra of 1,10-phenanthroline as a function of pH. The total 1,10-phenanthroline concentration is $1.37 \times 10^{-4}$ M. Excitation wavelength--297 $m\mu$. Method of illumination--frontal

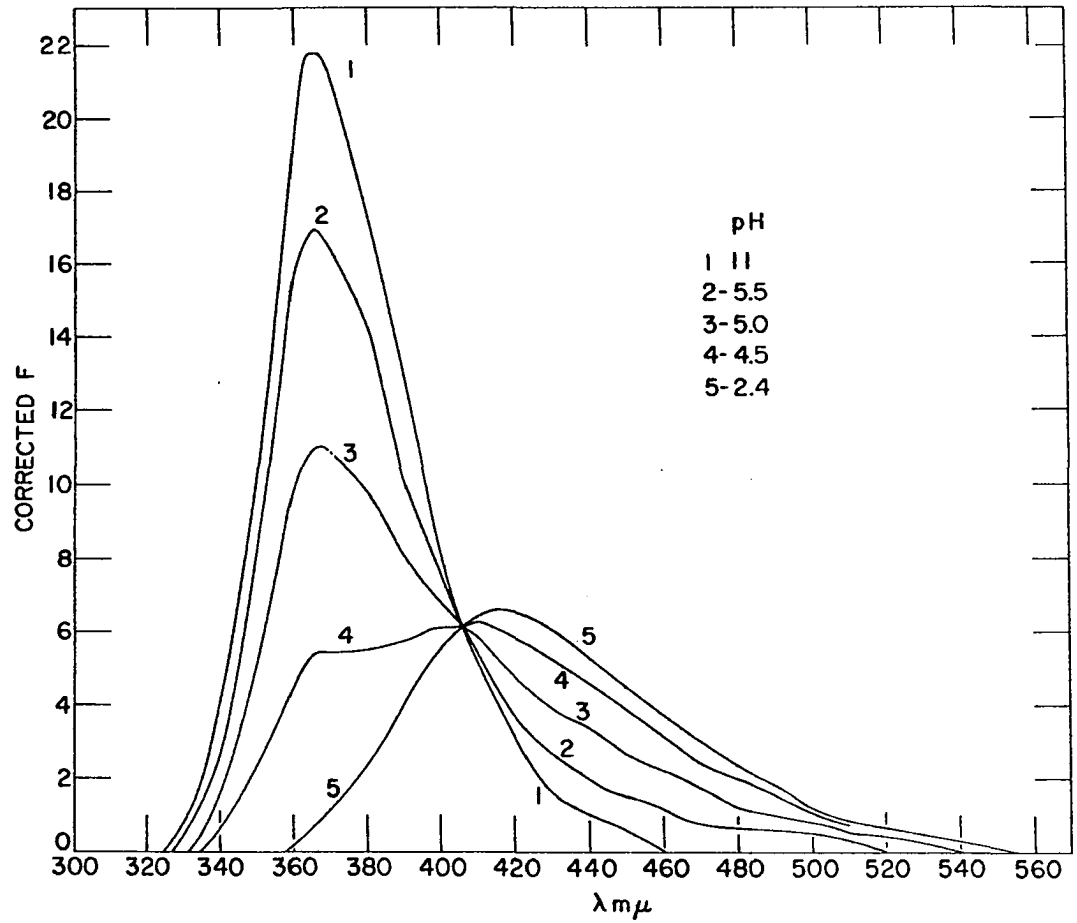
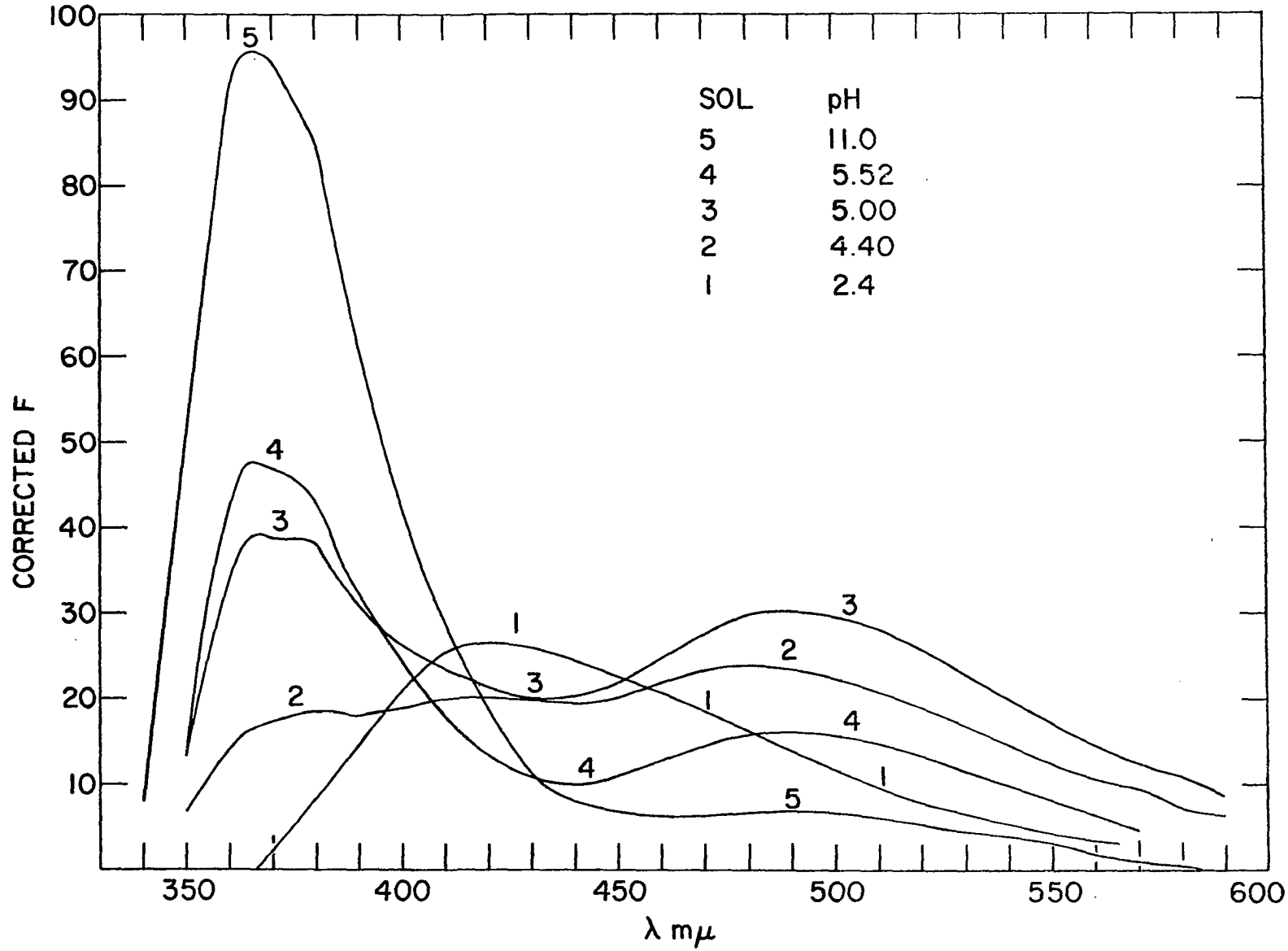

CORRECTED F
λmμ
pH
1 11
2-5.5
3-5.0
4-4.5
5-2.4
0
2
4
6
8
10
12
14
16
18
20
22
300
320
340
360
380
400
420
440
460
480
500
520
540
560
1
2
3
4
5

Figure 13. The fluorescence spectra of 1,10-phenanthroline as a function of pH. The total 1,10-phenanthroline concentration is $8.22 \times 10^{-3}$ M. Excitation wavelength--297 $m\mu$. Method of illumination--frontal


SOL pH
5 11.0
4 5.52
3 5.00
2 4.40
1 2.4
CORRECTED F
λ mμ
100
90
80
70
60
50
40
30
20
10
350
400
450
500
550
600

the aggregate to rearrange into a "sandwich" configuration with the planes of the molecules parallel. The bonding for an excimer type of species is expected to involve charge transfer and molecular exciton states.

The 490 $m_\mu$ fluorescence is red-shifted 6302 $cm^{-1}$ from the 0-0 band of the monoprotonated 1,10-phen at 26,700 $cm^{-1}$ (35), which is also indicative of an excimer type of interaction. While the 6000 $cm^{-1}$ rule would not strictly apply in this case, it ought to give a rough indication of the approximate energy that excimer fluorescence would be expected for 1,10-phen. The 6302 $cm^{-1}$ shift noted here is well within the limitations of the 6000 $cm^{-1}$ rule and the error in determining the 0-0 band.

Spectrum 5 of Figure 13 indicates some unusual behavior in that the 490 $m_\mu$ fluorescence is seen at low intensity and no poly(1,10-phen)-hydrogen(I) aggregates are predicted at this pH. An explanation of this behavior will require considerable data and will be the topic of the next section of this thesis. The explanation of this behavior will provide further evidence that the 490 $m_\mu$ fluorescence is due to an excimer type of species.

It should be noted here that excimer fluorescence has not been previously reported for nitrogen heterocycles in solution. The molecular orbital calculations of Azumi and Azumi (3), however, indicate that there are no theoretical reasons for not expecting this behavior in nitrogen heterocycles. The results of this work would lend some validity to the predictions of Azumi and Azumi.

# V. A STUDY OF THE QUENCHING OF THE FLUORESCENCE OF 1,10-PHENANTHROLINE IN BASIC SOLUTION

## A. Purpose

The purpose of this work will be to explain the appearance of the 490 $m_{\mu}$ fluorescence band in basic solution. The increase in percentage quenching of the fluorescence of the free-base 1,10-phen in concentrated solution by the ammonium ion, which is coincident with the appearances of the 490 $m_{\mu}$ fluorescence band, will also be investigated. As mentioned previously, this problem has been investigated by Jones (35), who proposed a ground state dimer to account for this behavior. It has been shown previously in this work that the ground state dimer does not exist over the concentration range that would account for this behavior. It is evident that a new approach is required. In addition, the quenching of the 1,10-phen fluorescence by quenchers other than ammonia buffers will be studied. It is hoped that this work will contribute in a general sense to a better understanding of fluorescence quenching processes.

## B. Experimental

### 1. Fluorescence measurements

Fluorescence data were taken on an Aminco-Bowman Spectrophotofluorimeter equipped with a RCA IP28 photomiltiplier tube. Slit arrangement 3 and the frontal illumination apparatus were used for all experiments. Fluorescence spectra were corrected by the method previously described in this thesis. The self-quenching of the 1,10-phen

fluorescence was determined by two different methods. One method is based on the fact that the fluorescence is a linear function of concentration if the absorbances of the solutions are less than 0.05 (33). The deviations from a linear response may be attributed to self-quenching. Using a path length of $1.5 \times 10^{-3}$ cm and an exciting wavelength of 297 $m_{\mu}$, the absorbance of 1,10-phen in basic solution is less than 0.05 if the 1,10-phen concentration is less than $8 \times 10^{-3}$ M. The path length of $1.5 \times 10^{-3}$ cm was achieved by inserting a quartz slug in a 1-cm cell using the frontal illumination apparatus. Under these conditions, the fluorescence intensity was low with respect to instrumental noise and scatter from the excitation source, making the method inherently inaccurate. A more accurate method is to use a solution of very high absorbance in the frontal illumination apparatus. Under these conditions, all of the exciting light is absorbed in the path length sampled by the emission monochrometer. The fluorescence is therefore the maximum obtainable and is independent of concentration. This is experimentally observed at a 1,10-phen concentration of approximately $2 \times 10^{-4}$ M at an excitation wavelength of 265 $m_{\mu}$ in basic solution. The fluorescence remains nearly constant to about $1 \times 10^{-3}$ M, indicating that the fluorescence is independent of concentration and all of the exciting light is being absorbed. Decreases in fluorescence above this concentration are attributed to self-quenching. This method will be referred to as the method of totally absorbing solutions throughout the remainder of this thesis.

2. Preparation of solutions

All 1,10-phen solutions were prepared by appropriate dilutions from concentrated stock solutions. In all solutions in which an ammonia buffer was not employed, the pH was adjusted to approximately 12 by the addition of an aliquot of KOH.

The viscosities of the solutions in Figure 16 were obtained by adding ethylene glycol in appropriate amounts. The viscosities were estimated from the data given in the International Critical Tables.

3. Reagents

The 1,10-phenanthroline was obtained from the Aldrich Chem. Co., recrystallized from $H_2O$, and analyzed for water content by thermogravimetric analysis. Ethylene glycol was distilled twice and checked for fluorescent impurities. None were found. The potassium halide salts were Baker "Analyzed" reagents and were used as is. All other materials were reagent grade.

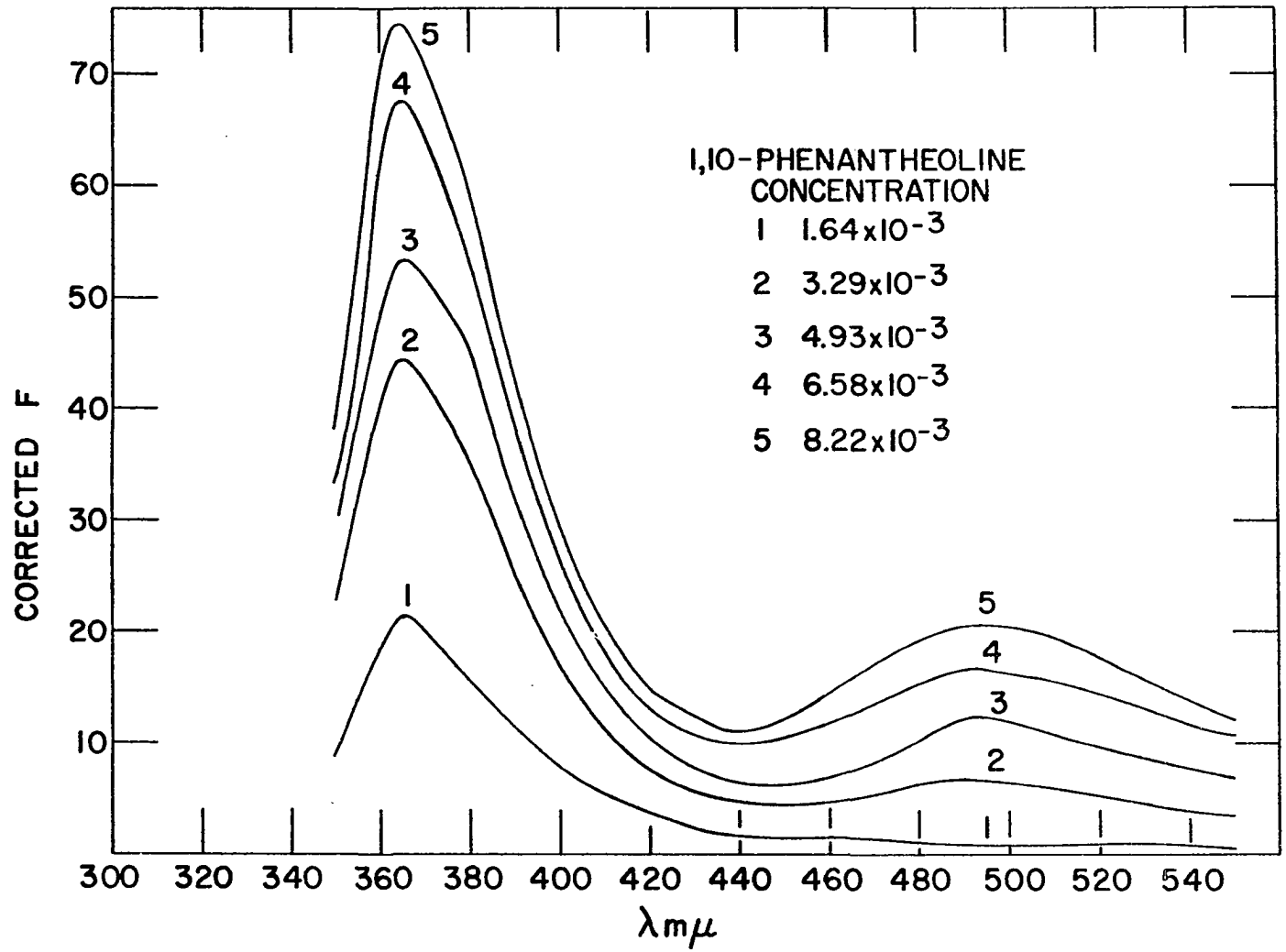
4. Miscellaneous

The temperature was 23° to 25°C for all solutions. Class A glassware was used in all experiments.

## C. Results and Discussion

The appearance of the 490 $m_\mu$ fluorescence band in basic solution as a function of 1,10-phen concentration is seen in Figure 14. The fluorescence band is seen to be identical to that exhibited by the poly(1,10-phen)-hydrogen(I) aggregates. The purpose of the ammonium ion is to function as a catalyst for the excited state protonation. The catalytic effect

Figure 14. The fluorescence spectra of 1,10-phenanthroline in the presence of an ammonium sulfate buffer as a function of 1,10-phenanthroline concentration. Excitation wavelength--297 $m_{\mu}$. Method of illumination--frontal. Ph--8.35. Concentration of $NH_4^+$ = 0.43 M. Path length--1.5 x $10^{-3}$ cm


1,10-PHENANTHEOLINE
CONCENTRATION
1 1.64x10-3
2 3.29x10-3
3 4.93x10-3
4 6.58x10-3
5 8.22x10-3
CORRECTED F
70
60
50
40
30
20
10
300 320 340 360 380 400 420 440 460 480 500 520 540
λmμ
1
2
3
4
5

of the ammonium ion on excited state protonations will require some further explanation. Nitrogen cycles are more basic in the excited state than in the ground state. It is expected that one would see the transformation of the acidic form to the basic form in the fluorescence spectrum in the pH region close to the excited state $pKa_1$. It has been noted, however, that many nitrogen heterocycles, including 1,10-phen, exhibit the transformation in the fluorescence spectrum close to the ground state $pKa_1$. This is attributed to the fact that the fluorescence process occurs at a rate greater than the hydrolysis reaction. Weller (76) was the first to demonstrate that the ammonium ion can act as a catalyst for the excited state hydrolysis reaction of acridine. Subsequent to Weller's work, this phenomenon has been demonstrated for 5,6-benzoquinoline (55), and Jones (35) has shown that the ammonium ion is functioning similarly in this system. The dependence of the 490 $m_\mu$ fluorescence on 1,10-phen concentration is indicative of more than one 1,10-phen associated with the species giving rise to the 490 $m_\mu$ fluorescence band. This is also verified by the fact that the same fluorescence band is observed for the poly(1,10-phen)hydrogen(I) aggregates. If more than one 1,10-phen is associated with the appearance of the 490 $m_\mu$ fluorescence band in basic solution, and it has been shown in this work that the appearance of this fluorescence band is not due to ground state aggregation, then the aggregation must occur after excitation. There are two possible mechanisms by which this might occur. One would involve excited state protonation followed by a reaction with another 1,10-phen as indicated below.

$$P^* + H_2O \longrightarrow HP^{*+} + OH^- \qquad (a)$$
$$P + HP^{*+} \longrightarrow HP_2^{*+} \qquad (b) \qquad (14)$$
$$HP_2^{*+} \longrightarrow h\nu\ (490\ m\mu) + H^+ + 2P \qquad (c)$$

P represents 1,10-phen and the asterisk indicates an excited state. The mechanism is seen to involve $HP^{*+}$, which is known to be fluorescent and should exhibit the fluorescence of the acidic form of 1,10-phen as indicated in Figure 12. In dilute basic solution, the acidic form of the 1,10-phen fluorescence does not appear, nor does the addition of as much as 0.78 M $NH_4^+$ catalyze the excited state protonation. This indicates that the reaction,

$$P^* + H_2O \longrightarrow HP^{*+} + OH^- \qquad (14a)$$

does not occur even in the presence of significant amounts of the catalyst. If this reaction does not occur, then the mechanism must involve the reaction,

$$P^* + P \longrightarrow P_2^* \qquad (15a)$$

This mechanism is seen to involve a reaction with the species giving the fluorescence of the basic form of 1,10-phen, which may be verified by demonstrating the well-known phenomenon of self-quenching.

Self-quenching was verified experimentally by two methods. One method employed the use of very short path lengths and is based on the deviation from an expected linear fluorescence response. The results of this study indicate self-quenching beginning at approximately the same 1,10-phen concentration at which the 490 $m\mu$ fluorescence band appears, verifying equation (15a). This method, however, required stringent control of excitation wavelength and path length, and the fluorescence

intensity was low with respect to noise and scatter, making it inherently inaccurate. An alternate method was employed, which is based on the fact that the fluorescence intensity is independent of concentration if all of the exciting radiation is absorbed near the surface of the cell and frontal illumination is used. The results of the self-quenching experiments using this method, both in the absence and the presence of the ammonium ion, are seen in Figure 15. The Stern-Volmer self-quenching constants, defined by equation (10), are 32.8, and 60 in the presence of the ammonium ion. The increase in self-quenching in the presence of the ammonium sulfate buffer will be discussed subsequently, but first it is necessary to demonstrate that the self-quenching is due to a diffusion rate-controlled collision between an excited and an unexcited 1,10-phen, rather than self-quenching by a resonance transfer of excitation energy discussed previously in this thesis. This may be accomplished by demonstrating a reciprocal dependence on viscosity for the self-quenching constant. The dependence of the self-quenching constant on viscosity is shown in Figure 16. The self-quenching constants were determined by the method of totally absorbing solutions discussed above and in the experimental section. The results are clearly in line with a quenching mechanism described by equation (15), and this equation must be the first step leading to the formation of the species responsible for the 490 $m_{\mu}$ fluorescence band.

In order to explain the increase in self-quenching in the presence of the ammonium sulfate buffer, the self-quenching was considered to be reversible. Quenching equations were developed to describe a situation which involves reversible and irreversible self-quenching in the presence of another quencher which are shown in detail in the Appendix, along with

Figure 15. The self-quenching of 1,10-phenanthroline in basic solution. Excitation wavelength--265 $m_{\mu}$. Fluorescence wavelength--365 $m_{\mu}$. Method of illumination--frontal (totally absorbing solutions). pH of the $(NH_4)_2SO_4$ buffer = 8.25. Concentration of the $NH_4^+$ = 0.88 M. $I^o$ and I are the fluorescence intensities in the absence and presence of self-quenching, respectively

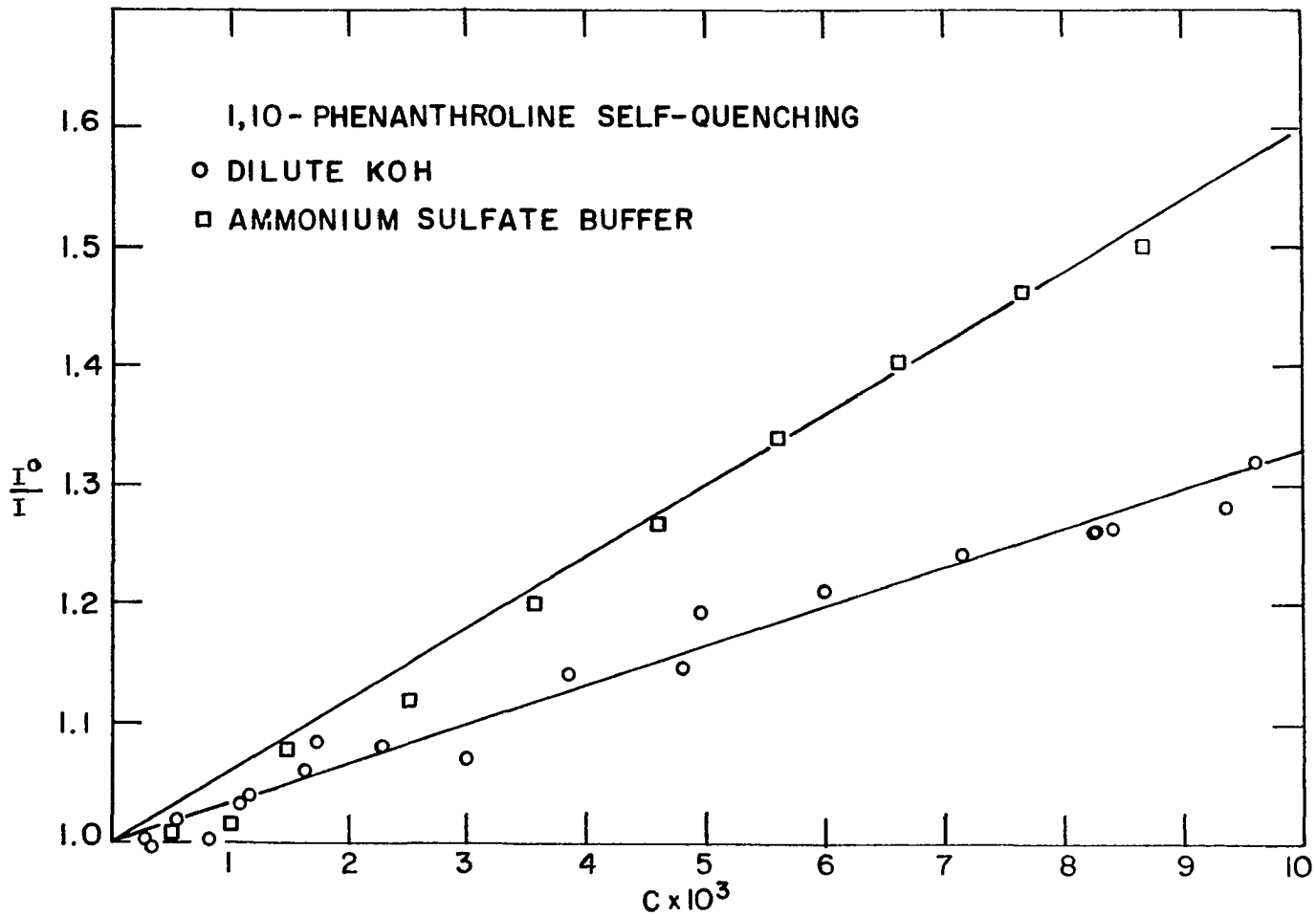
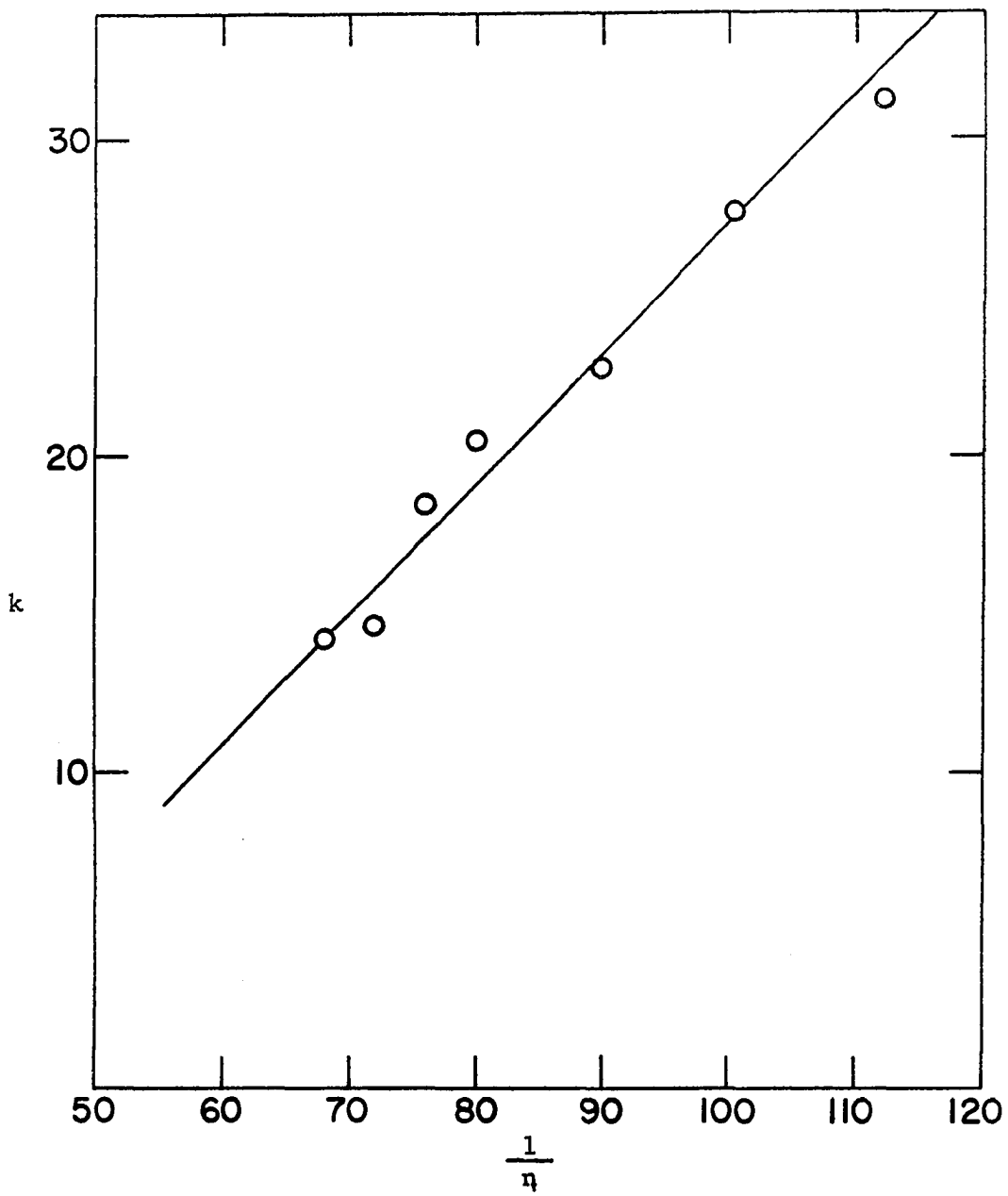

1,10-PHENANTHROLINE SELF-QUENCHING
o DILUTE KOH
□ AMMONIUM SULFATE BUFFER
1.6
1.5
1.4
1.3
1.2
1.1
1.0
$\frac{I^o}{I}$
1
2
3
4
5
6
7
8
9
10
C x 10³

Figure 16. The dependence of the self-quenching constant, k, on viscosity. $\eta$ is in poise


30
20
10
k
50
60
70
80
90
100
110
120
1/η

the notation used so as not to encumber the discussion. The expression for irreversible self-quenching is given by,

$$\frac{I^o}{I} = 1 + \frac{k_1}{k_{I1} + k_f} [M]_2 \qquad (15b)$$

which is equation (8A) at zero Q .

In the presence of another quencher at some constant concentration, the quenching equation is given by,

$$\frac{I^o}{I} = 1 + \frac{k_1}{k_{I1} + k_f + k_3 [Q]} [M]_2 \qquad (8A)$$

The quenching constant is seen to be always smaller in the presence of Q, reflecting the competition between self-quenching and the quencher, Q, for the potentially fluorescent species. It is apparent that this expression does not fit the case in question, as the self-quenching increased in the presence of the ammonium sulfate buffer. For the reversible case, the expression is given by,

$$\frac{I^o}{I} = 1 + \frac{k_1}{k_f + k_{I1} + k_3 [Q]} \left[1 - \frac{k_2}{k_2 + k_{I2} + k_4 [Q]}\right] [M]_2 \qquad (17A)$$

The self-quenching constant is seen to consist of two terms, the term seen in the irreversible case and the bracketed term which reflects the reversibility of the self-quenching. The bracketed term is seen to increase the self-quenching constant, while a decrease is noted in the other term. Therefore, self-quenching in the reversible case may be greater or less than it is in the absence of Q, depending on the relative value of the constants, $k_3$ and $k_4$. It is concluded that the self-quenching of 1,10-phen is reversible. It should be emphasized that in the above equations the terms involving Q are strictly constant, i.e., the equations

were derived for a constant quencher concentration. Equations will subsequently be presented in which Q is a variable.

The reversibility of excited state dimerization has also been proposed by Birks et al. (8) and Förster (28) to account for the temperature dependence of the ratio of eximer to monomer fluorescence. Williams (78) has also proposed a reversible dimerization to account for lifetime measurements which indicate a component in the fluorescence spectrum of phenanthrene with a lifetime $10^4$ times greater than expected. The component with the long lifetime is attributed to the fluorescence derived from the dissociation of the excited dimer into an unexcited and an excited molecule, the excited molecule being responsible for the fluorescence with the long lifetime. Also, Ludwig and Amata (46b) have recently proposed a similar scheme to account for the quenching of the benzene fluorescence by $CCl_4$.

The experimental data discussed above may be summarized as follows:

1. The appearance of the 490 $m_\mu$ fluorescence in basic solution is dependent on 1,10-phen concentration, indicating that two 1,10-phen molecules are involved in the species responsible for this fluorescence.

2. The 490 $m_\mu$ fluorescence is also exhibited by the poly-(1,10-phen)hydrogen(I) aggregates, and Jones (35) has shown that the appearance of this fluorescence band is catalyzed by the addition of the ammonium ion. Both facts indicate that a hydrogen ion is involved.

3. The free base 1,10-phen does not protonate in moderately basic solutions in the excited state, even though it is predicted

by the excited state $pKa_1$, nor does the protonation occur in the presence of an ammonium ion catalyst.

4. The appearance of the 490 $m_\mu$ fluorescence in basic solution is coincident with the appearance of reversible self-quenching.

A mechanism consistent with these facts is,

$$\begin{array}{ccccc} P + P^* & \rightleftharpoons & P_2^* + H_2O & \longrightarrow & P_2H^* + OH^- \\ \downarrow & & & & \downarrow \\ h\nu\ (365\ m_\mu) + P & & & & h\nu\ (490\ m_\mu) + P + P + H_2O \end{array} \qquad (16)$$

where,

P = 1,10-phen in the ground state,
$P^*$ = 1,10-phen in an excited state, and
$P_2^*$ = an excited state dimer.

The only feature of this mechanism that has not been experimentally verified is the basicity of the excited state dimer, $P_2^*$. However, Jones (35) and Langmuir (42) have shown that 1,10-phen is more basic in the excited state and it is perhaps not unexpected that a dimer derived from 1,10-phen would also be more basic in the excited state. $P_2^*$ would appear from these data to be non-fluorescent and the protonated form fluorescent. This suggests a situation similar to that exhibited by quinoline (34), where the relative energy of the n and the $\pi$ states determines the quantum efficiency. Without experimental verifaction, this proposal is offered as no more than speculation.

In order to further substantiate the reversible self-quenching behavior, the quenching by potassium halide salts and an ammonium chloride buffer was studied in dilute and concentrated 1,10-phen solutions. It was hoped that those data would supplement the data available in the

literature on quenching processes. In addition, quenching equations will be presented which are more general than those previously presented, in that the quencher, Q, will be treated as a variable.

The quenching data for the ammonium chloride buffer are seen in Figure 17. The quenching of the 1,10-phen fluorescence in dilute solutions is seen to obey the Stern-Volmer law with a quenching constant of 0.9. The quenching in concentrated solutions involves both the quenching of the free-base 1,10-phen and self-quenching. The quenching equation, developed in the Appendix, for the irreversible case is given by,

$$\frac{I^o}{I} = 1 + \frac{k_1}{k_f + k_{I1}} [M]_2 + \frac{k_3}{k_f + k_{I1}} [Q] \qquad (11A)$$

The broken line is the quenching predicted by equation (11A) for the ammonia buffer at this 1,10-phen concentration. The experimentally observed quenching is seen to be greater than predicted, which may be accounted for again by considering the self-quenching process as being reversible. The equation for the reversible case is given by,

$$\frac{I^o}{I} = 1 + \frac{k_3}{k_f + k_{I1}} [Q]_N + \frac{k_1}{k_f + k_{I1}} \left[ 1 - \frac{k_2}{k_{I2} + k_2 + k_4 [Q]_N} \right] [M]_2 \qquad (18A)$$

This equation predicts greater quenching by the ammonium chloride buffer than in the irreversible case by increasing the apparent self-quenching. The increase in apparent self-quenching is due to the competition of the quencher with the dissociation of the excited dimer. Similar behavior was noted for the halide ions. The quenching of the 1,10-phen fluorescence in dilute basic solution by KCl, KBr and KI is shown in Figure 18. The Stern-Volmer quenching constants are 39.5, 11.63, and 0.0 for KI,

Figure 17. The quenching of the 1,10-phenanthroline fluorescence by an ammonium chloride buffer. Excitation wavelength--265 $m_{\mu}$. Fluorescence wavelength--365 $m_{\mu}$. Method of illumination--frontal (totally absorbing solutions). pH of the buffer = 8.45. $I^{o}$ and I are the fluorescence intensities in the absence and presence of the quencher, respectively

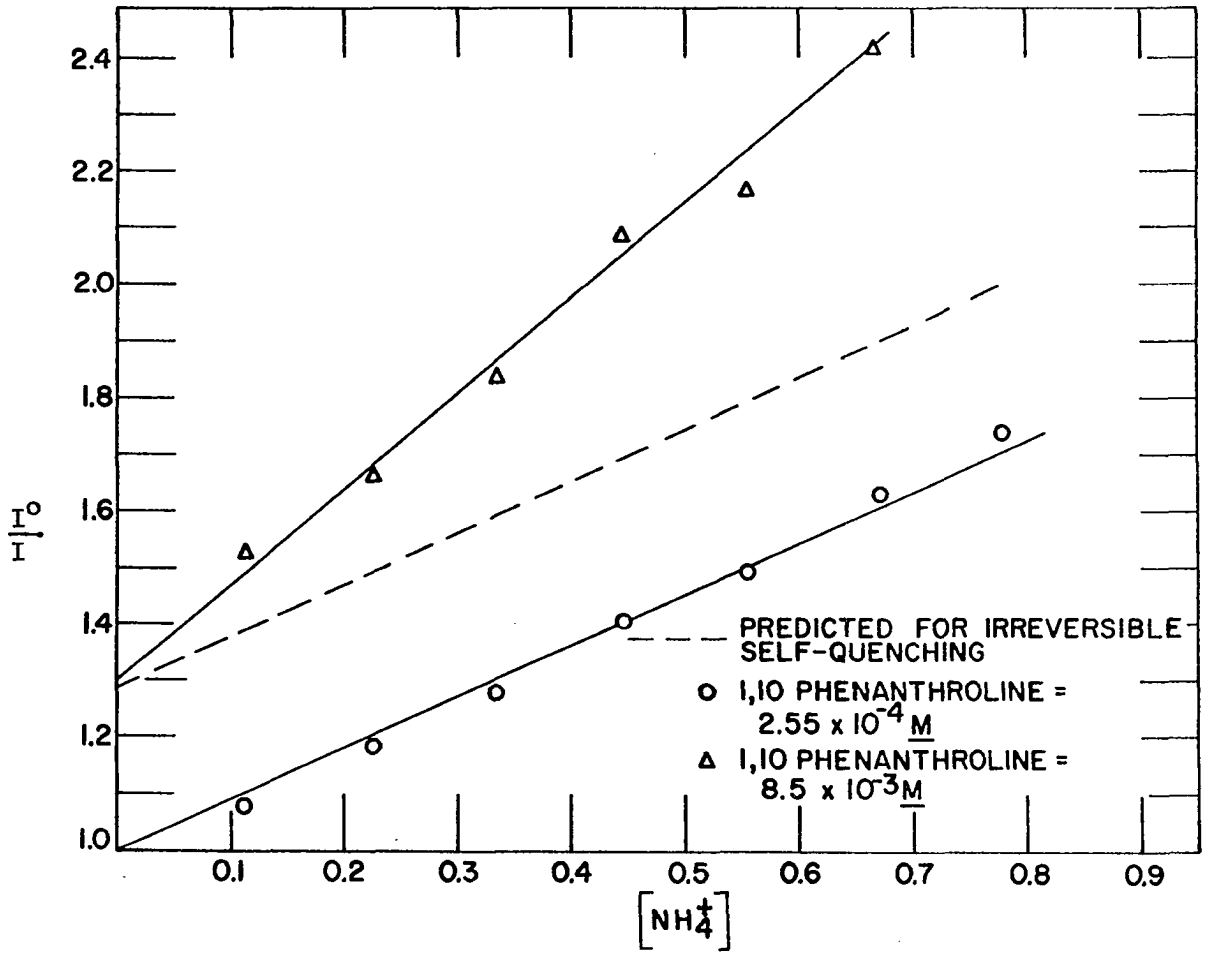
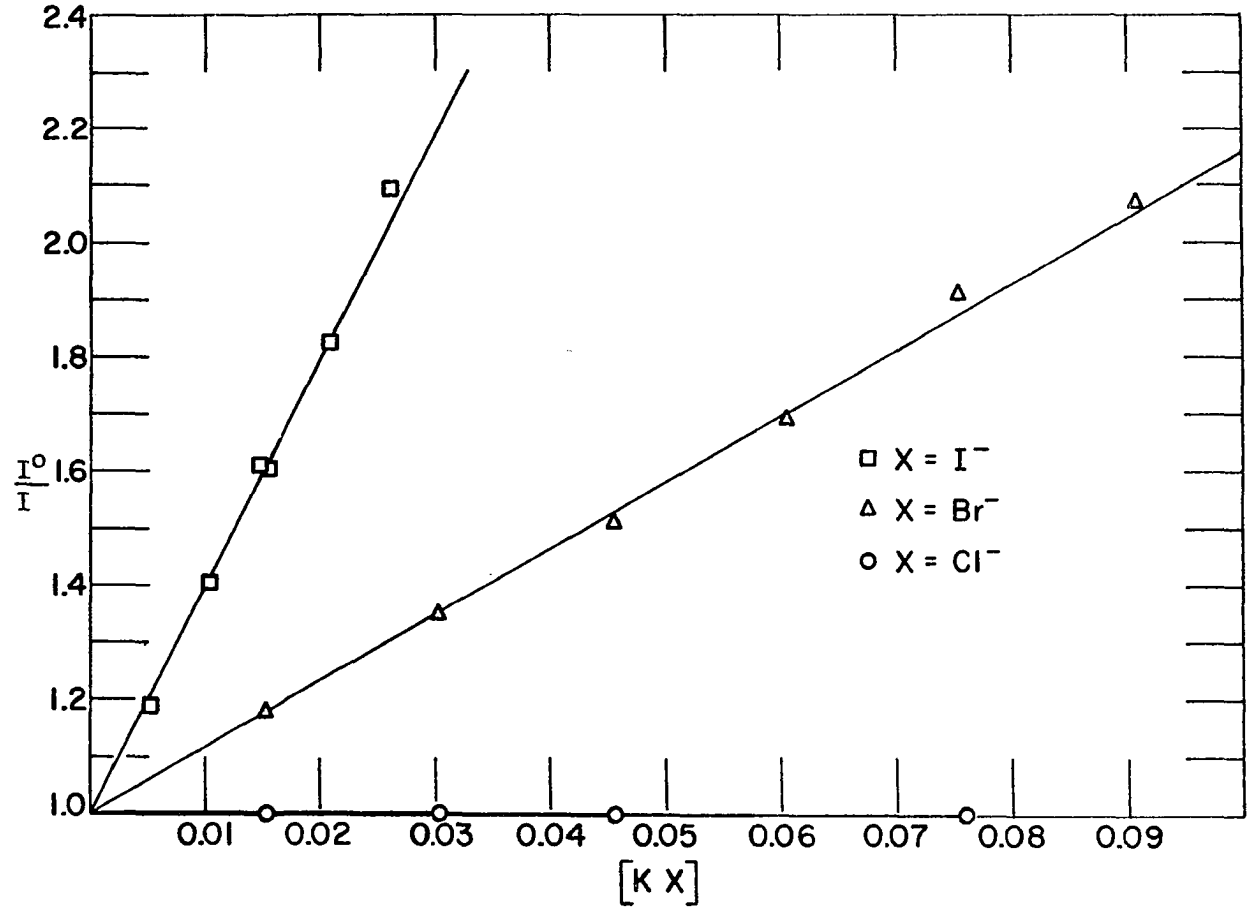

2.4
2.2
2.0
1.8
1.6
1.4
1.2
1.0
$\frac{I^o}{I}$
0.1
0.2
0.3
0.4
0.5
0.6
0.7
0.8
0.9
$[NH_4^+]$
PREDICTED FOR IRREVERSIBLE SELF-QUENCHING
1,10 PHENANTHROLINE = $2.55 \times 10^{-4}$ M
1,10 PHENANTHROLINE = $8.5 \times 10^{-3}$ M

Figure 18. The quenching of the 1,10-phenanthroline fluorescence in dilute solution by potassium halide salts. Excitation wavelength--265 $m_{\mu}$. Fluorescence wavelength--365 $m_{\mu}$. Method of illumination--frontal (totally absorbing solutions). pH = 12. Concentration of the 1,10-phenanthroline = $2.55 \times 10^{-4}$ M. $I^{o}$ and I are the fluorescence intensities in the absence and presence of the quencher, respectively

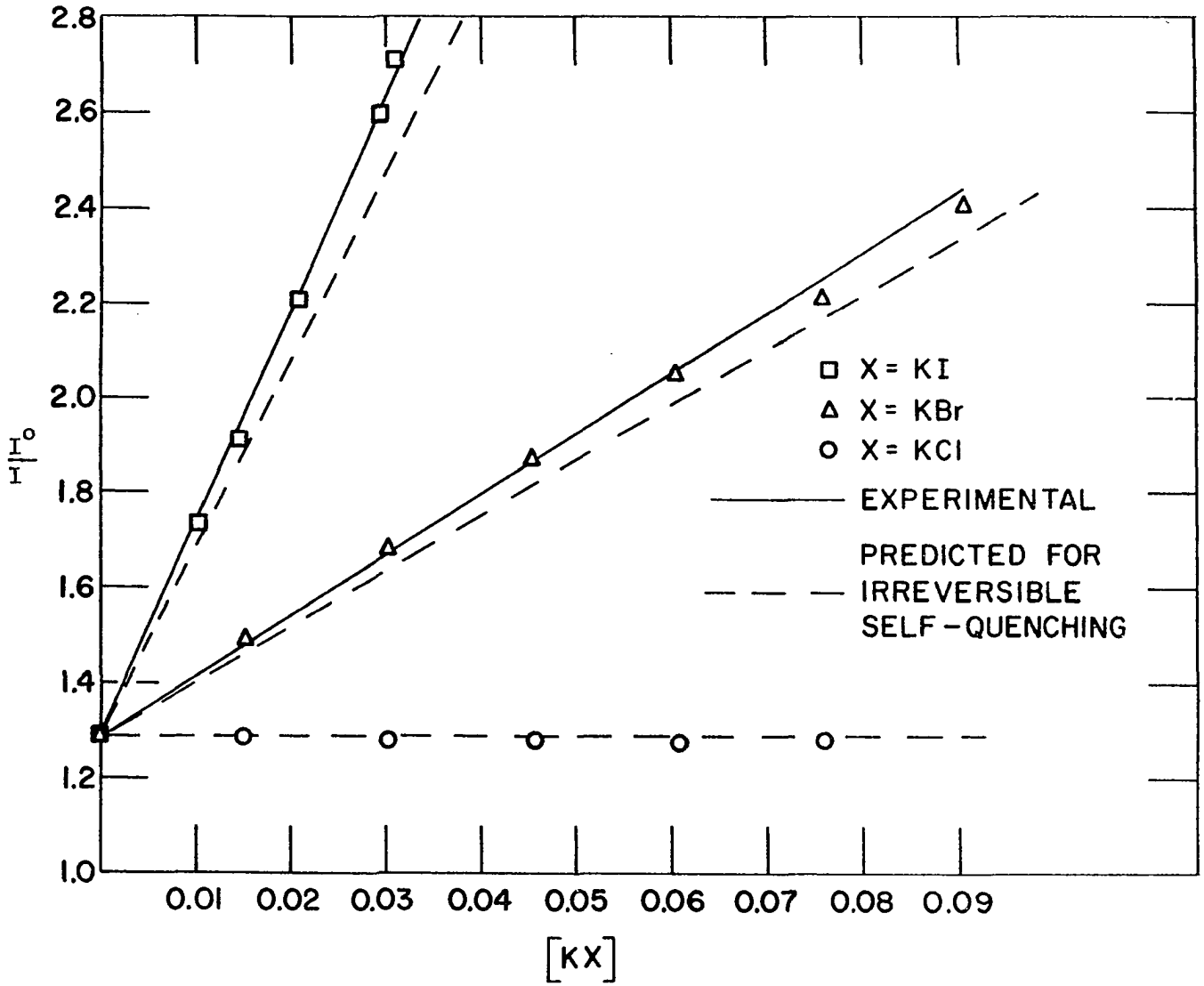

2.4
2.2
2.0
1.8
1.6
1.4
1.2
1.0
$\frac{I^o}{I}$
0.01
0.02
0.03
0.04
0.05
0.06
0.07
0.08
0.09
[K X]
□ X = I⁻
△ X = Br⁻
○ X = Cl⁻

KBr and KCl, respectively. Using these constants and the experimentally observed self-quenching of a $8.55 \times 10^{-3}$ M 1,10-phen solution, the quenching behavior was predicted for the irreversible case from equation (11A) for the halide ions. The predicted and the experimentally observed quenching of the fluorescence of an $8.55 \times 10^{-3}$ M solution of 1,10-phen are shown in Figure 19 for the halide ions. The observed quenching is again seen to be greater than predicted for irreversible self-quenching, except for the $Cl^-$ ion which is apparently a non-quencher for both 1,10-phen and the excited dimer. These data indicate that equation (18A) is applicable in this case.

The quenching constants for the halide ions are seen to increase with increasing ease of oxidation of the halide ion in Figure 18. This would imply that the quenching mechanism probably involves an encounter complex of charge transfer character with the halide ion donating an electron to the 1,10-phen. The quenching of the free-base 1,10-phen fluorescence by the ammonium chloride buffer is more difficult to explain. The quenching of the excited dimer by the ammonium ion involves protonation leading to a fluorescent species which does not occur with the free-base 1,10-phen in dilute solutions. It is also difficult to imagine a charge transfer mechanism involving the ammonium ion. The chloride ion is strictly a non-quencher, which leaves ammonia by a process of elimination. Ammonia would seem to be somewhat of a possibility as the quencher. The verification of this is left as an area for future research.

The quenching of the 1,10-phen fluorescence in basic dilute solution by the iodide ion was found to exhibit significant deviations from the

Figure 19. The quenching of the 1,10-phenanthroline fluorescence in concentrated solution by potassium halide salts. Excitation wavelength--265 $m_{\mu}$. Fluorescence wavelength--365 $m_{\mu}$. Method of illumination--frontal (totally absorbing solutions). pH = 12. Concentration of the 1,10-phenanthroline = $8.55 \times 10^{-3}$ M. $I^{o}$ and I are the fluorescence intensities in the absence and presence of the quencher, respectively


2.8
2.6
2.4
2.2
2.0
1.8
1.6
1.4
1.2
1.0
I°/I
0.01
0.02
0.03
0.04
0.05
0.06
0.07
0.08
0.09
[KX]
X = KI
X = KBr
X = KCl
EXPERIMENTAL
PREDICTED FOR IRREVERSIBLE SELF-QUENCHING

Stern-Volmer quenching law at iodide concentrations greater than 0.03 M. This is shown in Figure 20. This behavior has been reported in the literature by Boaz and Rollefson (9) for the quenching of the acridone fluorescence by the iodide ion. The behavior was attributed to the quencher being in the same solvent cage as the potentially fluorescent molecule in the ground state. A quenching equation was derived by them to account for this behavior and is given by,

$$\frac{I^o}{I} = 1 + k_1 [Q] + k_2 [Q]^2 \qquad (17)$$

where the notation is the same as that used previously. The 1,10-phen quenching was tested for conformity to this equation by plotting $\frac{\frac{I^o}{I} - 1}{[KI]}$ vs [KI] as shown in Figure 19. The data in Figure 19 are seen to conform to this quenching equation. It is concluded that the deviations from the Stern-Volmer law in this experiment are also due to the iodide ion being in the solvent cage of 1,10-phen in the ground state.

Figure 20. The quenching of the 1,10-phenanthroline fluorescence in dilute solution by the iodide ion. Excitation wavelength--265 $m\mu$. Fluorescence wavelength--365 $m\mu$. Method of illumination--frontal (totally absorbing solutions). pH = 12. Concentration of the 1,10-phenanthroline = $2.55 \times 10^{-4}$ M. $I^o$ and I are the fluorescence intensities in the absence and presence of the quencher, respectively

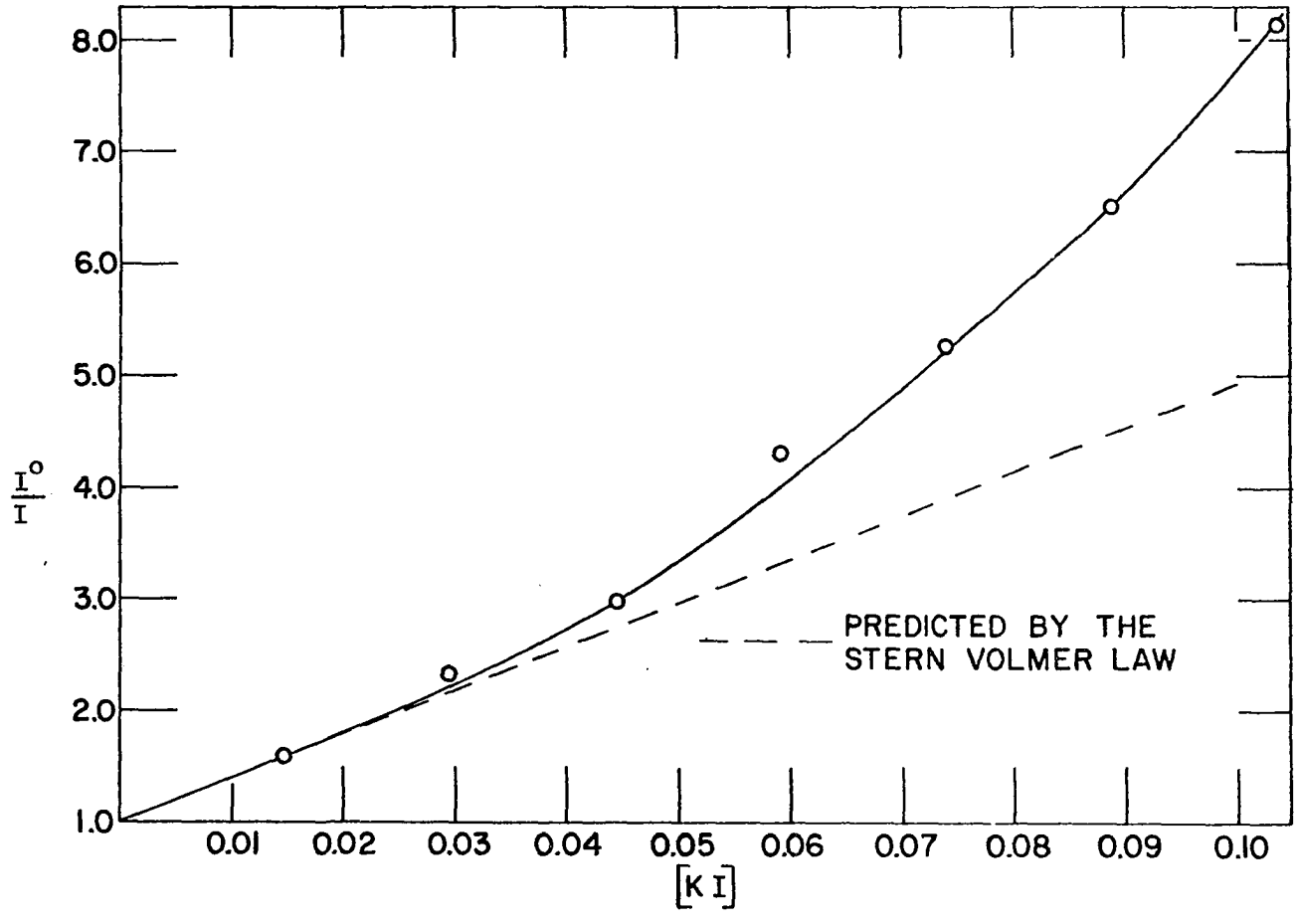
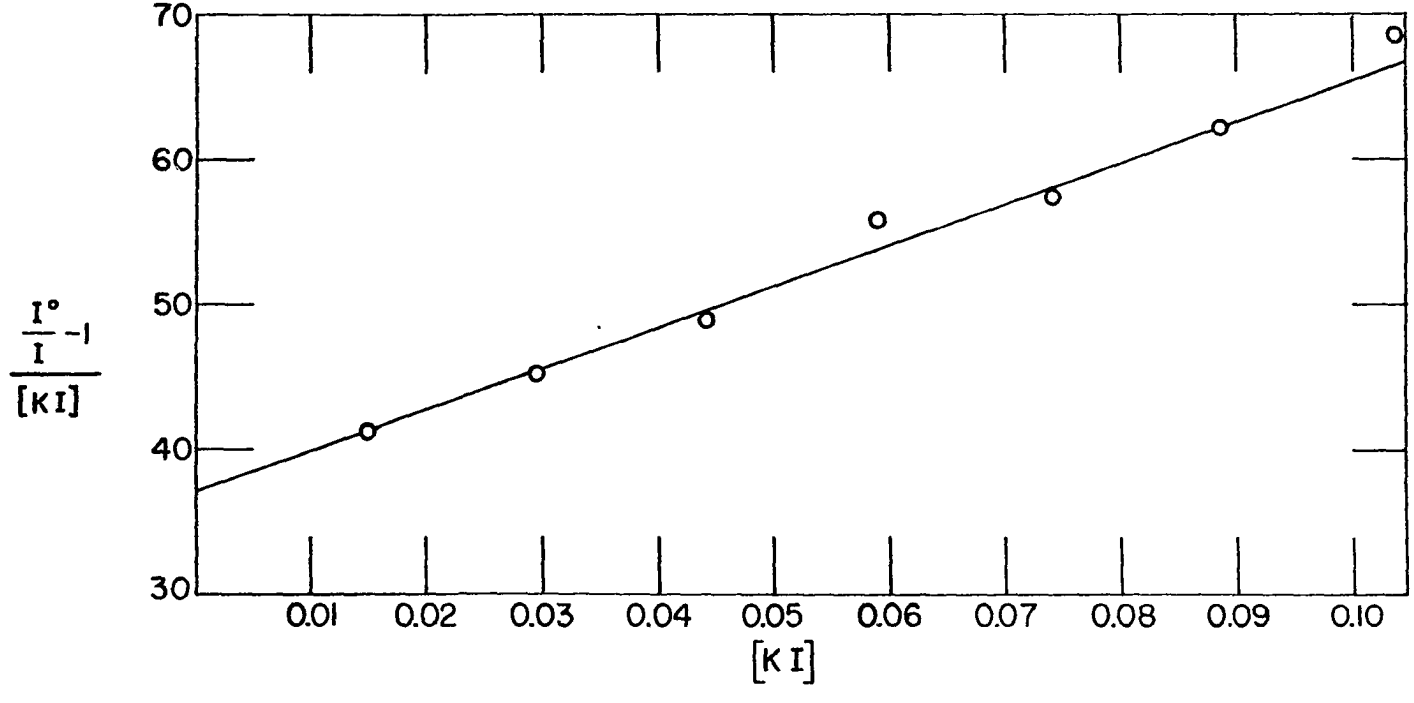

8.0
7.0
6.0
5.0
4.0
3.0
2.0
1.0
$\frac{I^o}{I}$
0.01
0.02
0.03
0.04
0.05
0.06
0.07
0.08
0.09
0.10
[K I]
PREDICTED BY THE
STERN VOLMER LAW

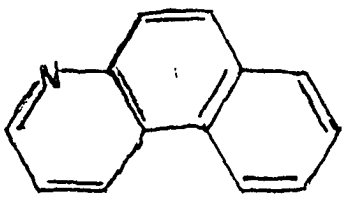
Figure 21. Plot of $\frac{I^o}{I} - 1$ vs. $[KI]$ from the data given in Figure 20


70
60
50
40
30
$\frac{I^\circ}{I} - 1$
[KI]
0.01
0.02
0.03
0.04
0.05
0.06
0.07
0.08
0.09
0.10
[KI]

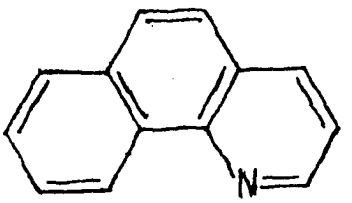
# VI. AN UV ABSORPTION AND FLUORESCENCE STUDY OF 5,6-BENZOQUINOLINE AND 7,8-BENZOQUINOLINE

## A. Introduction and Literature

A description of the fluorescence and uv absorption behavior of 1,10-phen has been presented in some detail in this thesis. The implications of this research are that some of the behavior exhibited by 1,10-phen ought to be exhibited by similar molecules. To test this hypothesis, 5,6-benzoquinoline and 7,8-benzoquinoline were chosen for study and will be investigated to determine whether they form hydrogen bonded aggregates with the hydronium ion in the ground state and excimer species in the excited state. The structures of these two compounds are depicted below.



5,6-benzoquinoline



7,8-benzoquinoline

The spectral properties of 5,6-benzoquinoline and 7,8-benzoquinoline in aqueous solutions have been the subject of some investigation. The absorption spectra of these compounds have been studied as a function of pH by Perkampus and Kohler (59) and they reported pKa values of 4.15 and 5.00 for 7,8-benzoquinoline and 5,6-benzoquinoline, respectively. Nakamizo (55) has reported the fluorescence and absorption spectra of 5,6-benzoquinoline in aqueous solution. The ground and excited state pKa values were found to be 5.03 and 10.95, respectively. Ground and

excited state pKa values have also been determined by Kilimov et al. (38) for 5,6-benzoquinoline and 7,8-benzoquinoline, but the details were unavailable. Perkampus and Kortüm (60) have reported excimer fluorescence at 454 $m_{\mu}$ for 5,6-benzoquinoline and 7,8-benzoquinoline in the solid state at low temperatures. While it is not strictly valid to compare results obtained under these conditions to those obtained in this study, the appearance of excimer fluorescence for these compounds in solution would not be unexpected.

## B. Experimental

### 1. Absorption and fluorescence data

The instruments, cells, methods of illumination, and techniques for obtaining corrected fluorescence spectra have been previously described in this thesis and will not be repeated here.

### 2. Solubility data

Saturated solutions were prepared by placing the solid and either standardized acid or base in 15-ml test tubes and shaking them for 48 hours with a Burrell wrist-action shaker. The equilibration time was determined by monitoring the uv absorption spectrum for 1 week. Equilibrium was reached in approximately 48 hours. The solubilities were determined by an analysis of an aliquot from each of the saturated solutions. The analyses were accomplished by measuring the uv absorption spectra after appropriate dilution and pH adjustment. The molar absorptivities used in these analyses were determined by measuring the absorbance of several standard solutions, prepared by weighing out the solid material

and dissolving it to an appropriate volume.

The saturated solutions which were used in the determination of the overall formation constant for the bis(5,6-benzoquinoline)hydrogen(I) complex were prepared by equilibrating various samples of the solid with acid solutions in volumetric flasks. The solutions were stirred with a magnetic stirrer for 48 hours. After a sufficient cooling and settling period, the solutions were filtered.

3. Solution preparation

5,6-benzoquinoline and 7,8-benzoquinoline solutions were prepared by an appropriate dilution from a stock solution. Standard acids were analyzed by the method previously described in this thesis. The pH values for the dilute solution spectra were obtained by the same method used for 1,10-phen.

4. Reagents

7,8-benzoquinoline was obtained from Aldrich Chemical Co. and recrystallized from ether at dry-ice-acetone temperatures. The resulting product melted sharply at 50°C. Aldrich Chemical Co.'s purissimum-grade 5,6-benzoquinoline was used as received except as noted in the discussion. All other chemicals were reagent grade.

5. Miscellaneous

The temperature was 23° to 25°C for all of the experiments. Class A glassware was used for all experiments.

## C. Results and Discussion

To determine whether 5,6-benzoquinoline and 7,8-benzoquinoline form aggregates in which more than one base is associated with the hydronium ion, a solubility study was employed. The solubility of a sparingly soluble ligand in the presence of a metal ion is given by (63),

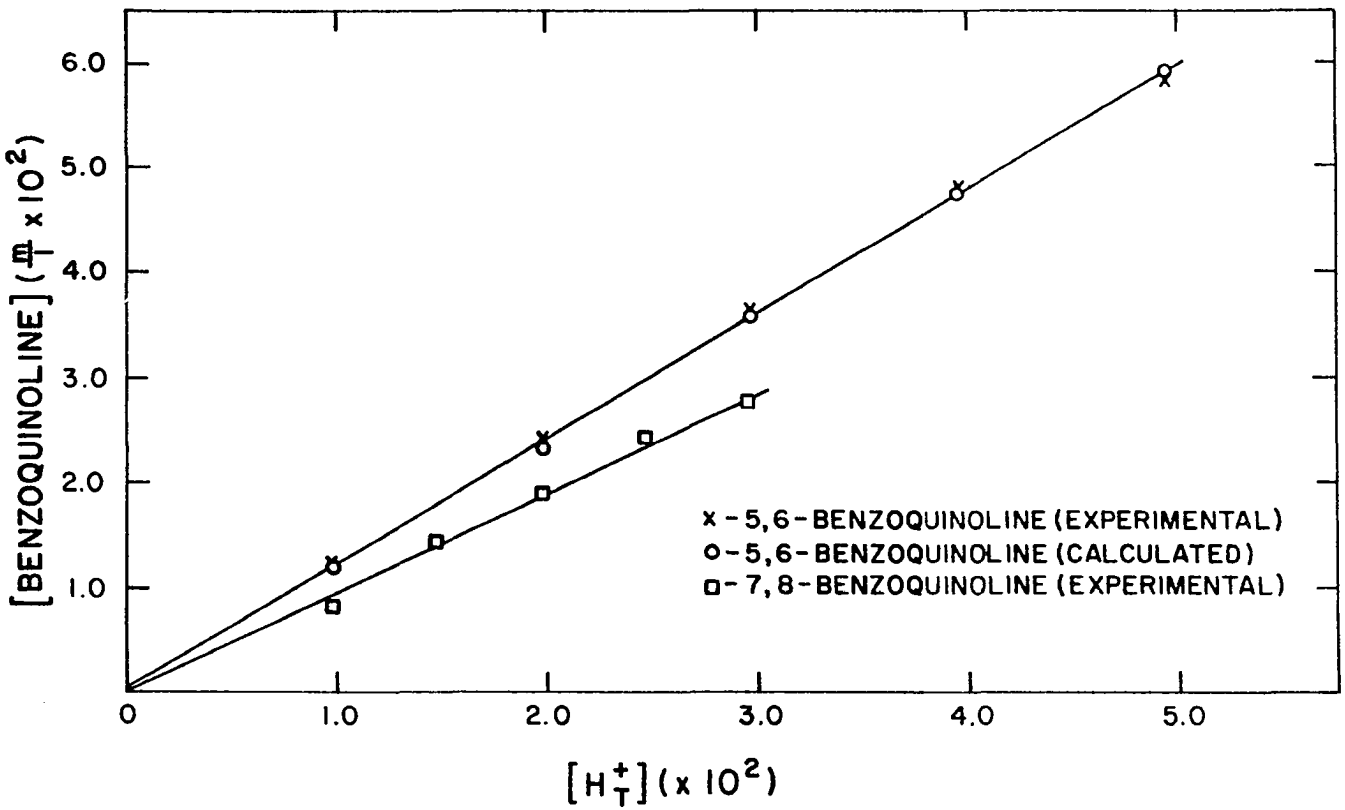
$$S_L = S_L^o + \sum_{n=1}^{n} n [ML_n] \qquad (18)$$

where $S_L$ is the total or analytical solubility of the ligand and $S_L^o$ is the solubility of the ligand in the absence of the metal ion. For the purposes of this study, the hydrogen ion can be considered as the metal ion and the nitrogen heterocycle as the ligand. The mean ligand number is given by,

$$\bar{n} = \frac{S_L - S_L^o}{[M]} \qquad (19)$$

and it is evident that a plot of $S_L$ vs. hydrogen ion concentration will be linear with a slope of $\bar{n}$. The solubility data for 5,6-benzoquinoline and 7,8-benzoquinoline are shown in Figure 22. The slopes are linear for both compounds, indicating that the above equations are valid for this system. 7,8-Benzoquinoline gave a slope of one, indicating no aggregates are formed with this compound. 5,6-Benzoquinoline gave a slope of 1.2, however, which is interpreted to indicate aggregate formation. With a mean ligand number of 1.2, it is reasonable to assume that the highest order complex formed in appreciable quantities is the bis-(5,6-benzoquinoline)hydrogen(I) complex. Expanding equation (18) in

Figure 22. The solubility of 5,6-benzoquinoline and 7,8-benzoquinoline as a function of the concentration of acid


[BENZOQUINOLINE] ($\frac{m}{l}$ x 10$^2$)
6.0
5.0
4.0
3.0
2.0
1.0
0
1.0
2.0
3.0
4.0
5.0
[$H_T^+$] (x 10$^2$)
x - 5,6-BENZOQUINOLINE (EXPERIMENTAL)
o - 5,6-BENZOQUINOLINE (CALCULATED)
□ - 7,8-BENZOQUINOLINE (EXPERIMENTAL)

terms of the overall formation constants, defined by,

$$\beta_n = \frac{[HB_n]}{[H^+]\,[B]^n} \quad , \tag{20}$$

gives,

$$S_L = [B] + \beta_1 [B]\,[H^+] + 2\beta_2 [B]^2\,[H^+] \tag{21}$$

where B is 5,6-benzoquinoline. It is evident that a pH measurement of a saturated solution and the solubility, $S_L$, are sufficient data to calculate $\beta_2$, if $\beta_1$ and the solubility of B are known. $\beta_1$ is seen to be the reciprocal of the acid dissociation constant, which has been reported by Nakamizo (55) to be $1.071 \times 10^5$. The solubility and pH of two saturated solutions of 5,6-benzoquinoline (0.0015 and 0.0025 M in $H_2SO_4$) were determined along with the solubility of B in basic solution, which was found to be $3.95 \times 10^{-4}$ M. Using this information and the acid dissociation constant of Nakamizo (55), $\beta_2$ was calculated and found to be $7.34 \times 10^8$. Using this value and a charge balance equation, the solubilities were calculated and compared with the experimentally determined solubilities of 5,6-benzoquinoline. These are shown in Figure 22. While this value of $\beta_2$ is presented as no more than an estimate, the agreement between experimental and calculated data is quite good.

Aggregate formation with these two compounds seems to correlate with the basicity of the two molecules. The $pKa_1$ values of 5,6-benzoquinoline and 7,8-benzoquinoline have been reported to be 5.03 (55) and 4.15 (59), respectively. The more basic 5,6-benzoquinoline is seen to form an aggregate while the less basic 7,8-benzoquinoline does not. Comparisons of this kind with solubility data are possible only if the

free-base solubilities are similar. Thus 1,10-phen with a $pKa_1$ value very close to that of 5,6-benzoquinoline, forms aggregates to a much greater extent in saturated solutions than 5,6-benzoquinoline does because of the greater aqueous solubility of the free-base 1,10-phen, which is nearly two orders of magnitude greater than that of 5,6-benzoquinoline in aqueous media.

In order to study the absorption and fluorescence spectra of aggregates, it is first necessary to study the spectra of very dilute solutions to establish the spectrum of the free-base and its conjugate acid. These spectra then serve as a reference to determine the effect of aggregation on spectral properties. The absorption spectra of dilute solutions are also necessary to establish excitation wavelengths for fluorescence measurements. The dilute solution absorption spectra of 5,6-benzoquinoline and 7,8-benzoquinoline as a function of pH are shown in Figures 23 and 24, respectively. As expected, the spectra of the two compounds are similar. In comparison with the 1,10-phen dilute solution spectra, more fine structure is evident and the molar absorptivity is increased in the long wavelength transitions. The latter fact is of some consequence in the interpretation of the quantum efficiencies of these compounds and will be referred to when this topic is discussed. The dilute solution fluorescence spectra are seen in Figures 25 and 26 for 5,6-benzoquinoline and 7,8-benzoquinoline, respectively. Using the method of Weller (76), discussed in detail previously in this thesis, the excited state pKa values were estimated for both compounds using the dilute solution absorption and fluorescence spectra and the ground state pKa values cited above. The excited state pKa values are 10 and 11 for

Figure 23. The uv absorption spectra of 5,6-benzoquinoline as a function of pH. The concentration of the 5,6-benzoquinoline is $6.67 \times 10^{-5}$ M

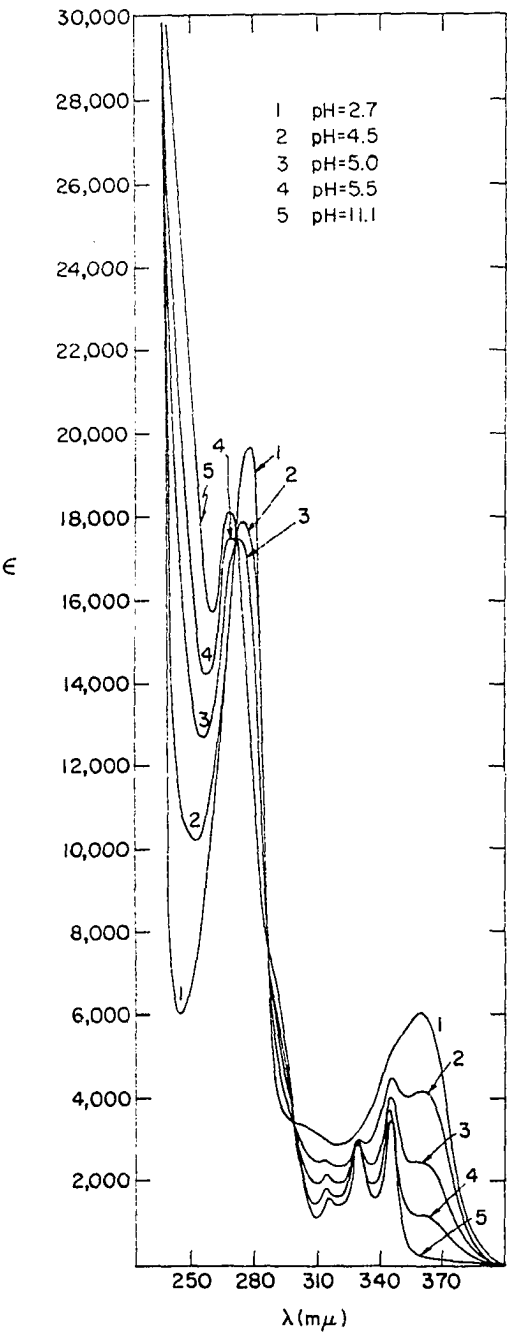

30,000
28,000
26,000
24,000
22,000
20,000
18,000
16,000
14,000
12,000
10,000
8,000
6,000
4,000
2,000
ε
1 pH=2.7
2 pH=4.5
3 pH=5.0
4 pH=5.5
5 pH=11.1
250
280
310
340
370
λ(mμ)

Figure 24. The uv absorption spectra of 7,8-benzoquinoline as a function of pH. The concentration of the 7,8-benzoquinoline is $5.18 \times 10^{-5}$ M

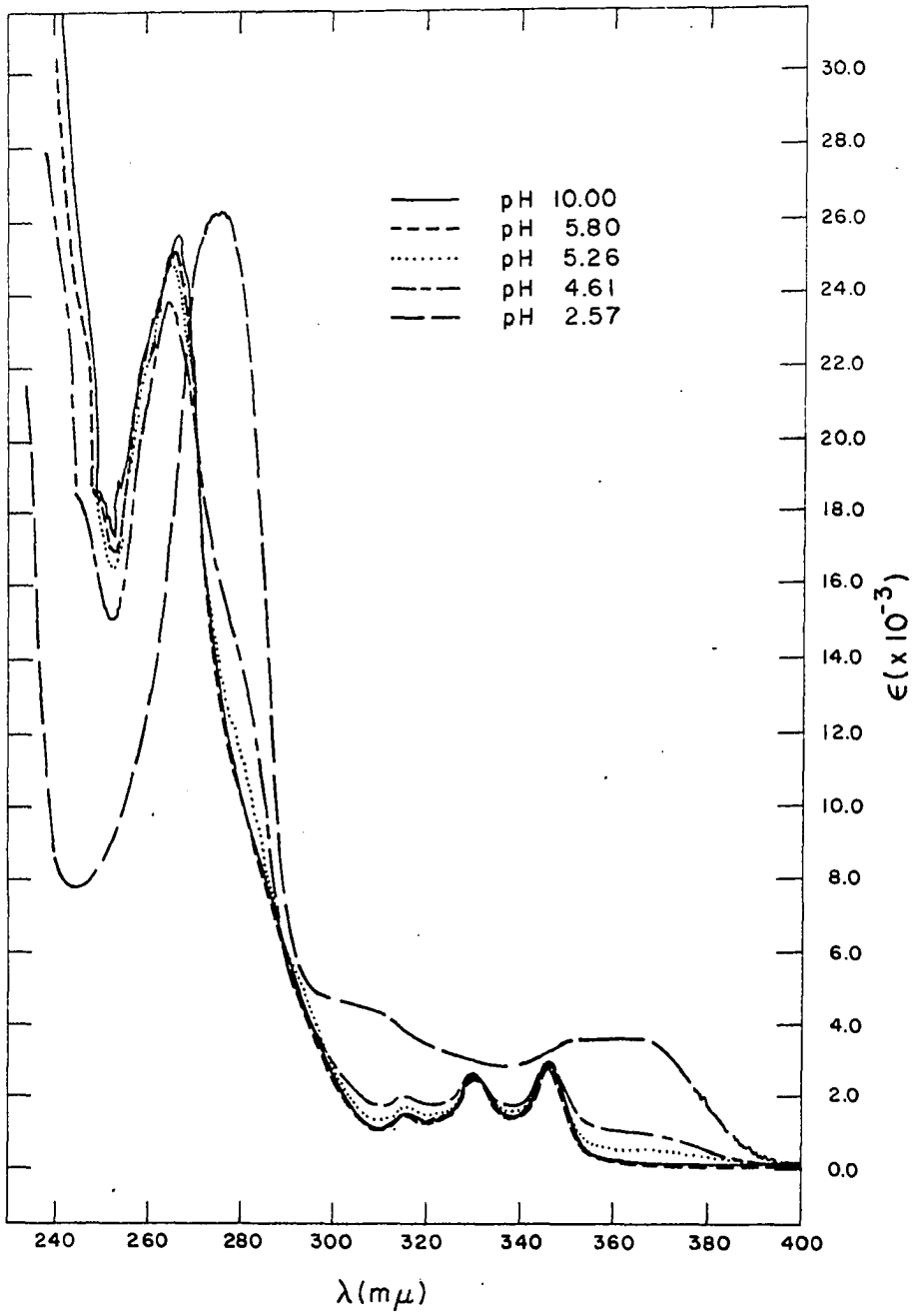

pH 10.00
pH 5.80
pH 5.26
pH 4.61
pH 2.57
30.0
28.0
26.0
24.0
22.0
20.0
18.0
16.0
14.0
12.0
10.0
8.0
6.0
4.0
2.0
0.0
ε(x 10⁻³)
240
260
280
300
320
340
360
380
400
λ(mμ)

Figure 25. The fluorescence spectra of 5,6-benzoquinoline as a function of pH. Excitation wavelength--310 $m_{\mu}$. Concentration of the 5,6-benzoquinoline--2.47 x $10^{-5}$ M. Method of illumination--right angle with a 4-mm cylindrical cell

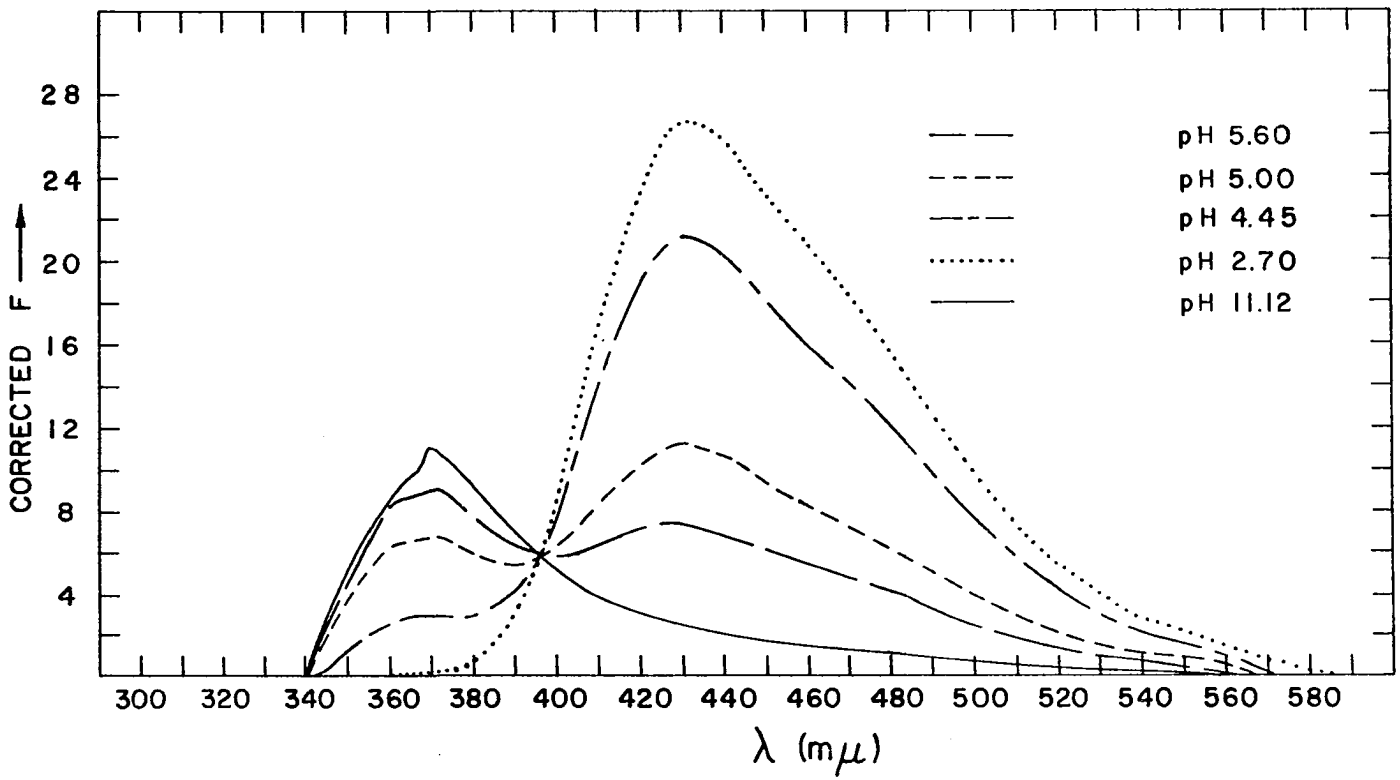
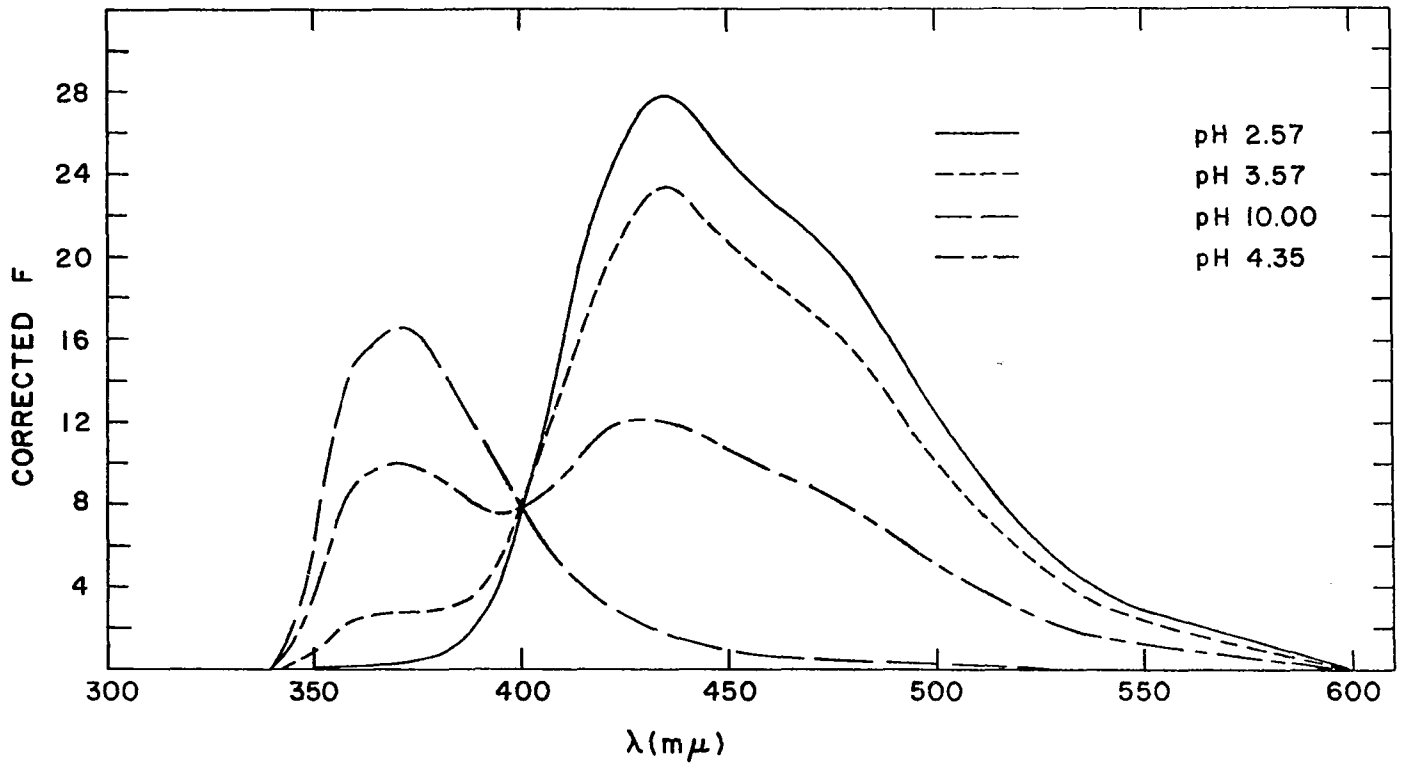

CORRECTED F
28
24
20
16
12
8
4
300 320 340 360 380 400 420 440 460 480 500 520 540 560 580
λ (mμ)
pH 5.60
pH 5.00
pH 4.45
pH 2.70
pH 11.12

Figure 26. The fluorescence spectra of 7,8-benzoquinoline as a function of pH. Excitation wavelength--310 $m_{\mu}$. Concentration of the 7,8-benzoquinoline--5.18 x $10^{-5}$ M. Method of illumination--right angle with a 4-mm cylindrical cell

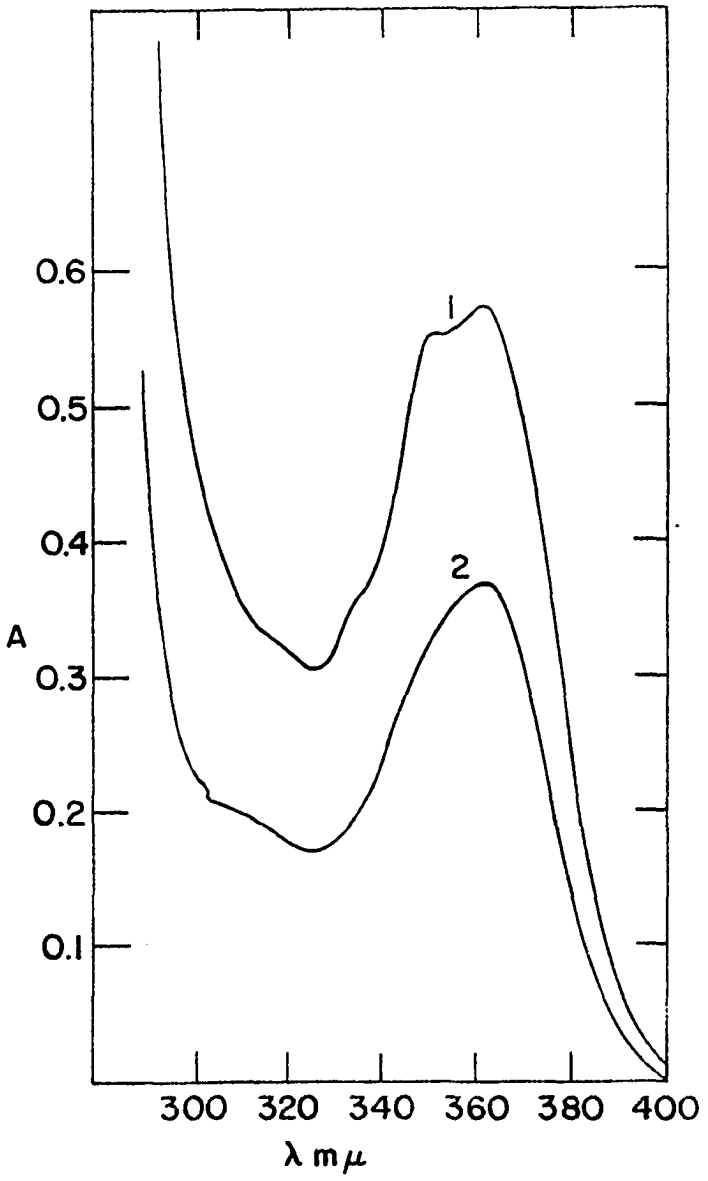

CORRECTED F
28
24
20
16
12
8
4
300
350
400
450
500
550
600
λ(mμ)
pH 2.57
pH 3.57
pH 10.00
pH 4.35

7,8-benzoquinoline and 5,6-benzoquinoline, respectively. The dilute solution fluorescence spectra for these compounds indicate the change from the acidic to the basic form close to the ground state pKa values. This is attributed, as in the case of 1,10-phen, to the fluorescence rate being much greater than the hydrolysis rate of the excited state. This would result in the molecule being in the same form that it was in the ground state when the fluorescence process occurs.

The absorption spectrum of a solution containing the aggregate of 5,6-benzoquinoline, prepared by saturating a .0025 M HCl solution with 5,6-benzoquinoline, showed little difference from the acid form, as indicated in Figure 27. Some caution is required in the interpretation of this spectrum due to the small amount of aggregate present relative to the amount of the acidic form present. It would appear, however, that the spectrum of the aggregate does not differ appreciably from the free-base and the acidic form of 5,6-benzoquinoline. Such spectral behavior would be consistent with an aggregate with the emphasis on hydrogen bonding similar to that proposed by Fahsel and Banks (21) for the poly-(1,10-phen)hydrogen(I) aggregates.

Attempts to obtain a fluorescence spectrum of the bis(5,6-benzo-quinoline)hydrogen(I) aggregate were unsuccessful. A new fluorescence band appeared at 500 $m_{\mu}$ in solutions concentrated enough to contain the aggregate. This band could not be attributed to the aggregate alone because the same band appeared in acid solution where only the acid form exists. The appearance of this band, along with a substantial overlapping band due to the acid form, made it difficult to discern the effect of the aggregate on the spectrum. The 500 $m_{\mu}$ band indicated an

Figure 27. Spectrum of a solution containing the bis(5,6-benzoquinoline)hydrogen(I) aggregate and the acid form of 5,6-benzoquinoline. 1--solution containing the aggregate. 2--simple acidic form


0.6
0.5
0.4
A
0.3
0.2
0.1
1
2
300
320
340
360
380
400
λ mμ

excimer fluorescence which warranted further investigation.

The appearance of the 500 $m_\mu$ band of 5,6-benzoquinoline in 0.05 M $H_2SO_4$ is seen in Figure 28. The solutions are totally absorbing under these conditions, which, in the absence of self-quenching, would give a fluorescence yield independent of concentration. The decrease in the yield of the fluorescence of the acid form is accompanied by an increase in the 500 $m_\mu$ fluorescence. The 500 $m_\mu$ band is shifted 5706 $cm^{-1}$ from the 0-0 band of the acid form of 5,6-benzoquinoline at 25706 $cm^{-1}$. This is within the limitations of the 6000 $cm^{-1}$ rule and further implicates excimer fluorescence. Similar behavior was exhibited by 7,8-benzoquinoline in acid solutions. The appearance of the long wavelength band as a function of 7,8-benzoquinoline concentration in 0.05 M $H_2SO_4$ is seen in Figure 29. The band for 7,8-benzoquinoline appears at 510 $m_\mu$ which is 5981 $cm^{-1}$ from the 0-0 band of the acid form at 25,588 $cm^{-1}$. This is interpreted as an indication of excimer fluorescence.

In order to definitely establish that these long wavelength bands are due to excimer species, it is necessary to demonstrate that no changes occur in the ground state and that the bands are not due to an impurity in the materials used. Absorption spectra for 5,6-benzoquinoline and 7,8-benzoquinoline were obtained in acidic media over the concentration range excimer fluorescence is exhibited. No changes in the spectra were noted. Beer's law plots for the two compounds in acid media are shown in Figure 30. The absorption data indicate no ground state aggregation for the acid form of these compounds. In addition, repeated recrystallization of 7,8-benzoquinoline and 5,6-benzoquinoline did not alter the fluorescence spectra in concentrated solutions. It must be concluded

Figure 28. The fluorescence spectra of 5,6-benzoquinoline in 0.05 M $H_2SO_4$ as a function of 5,6-benzoquinoline concentration. Excitation wavelength--300 $m\mu$. Method of illumination--frontal (totally absorbing solutions)

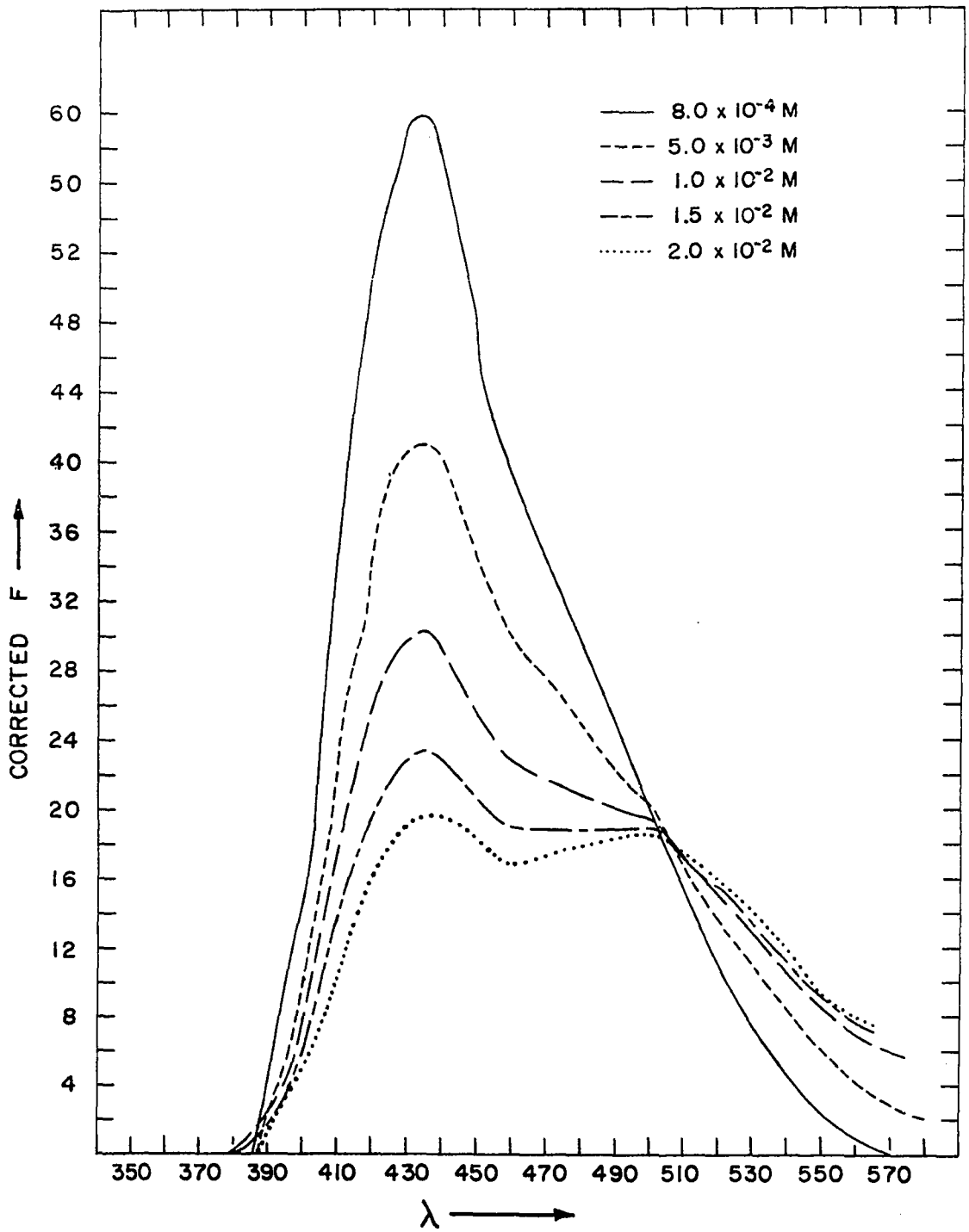

8.0 x 10⁻⁴ M
5.0 x 10⁻³ M
1.0 x 10⁻² M
1.5 x 10⁻² M
2.0 x 10⁻² M
CORRECTED F
60
50
52
48
44
40
36
32
28
24
20
16
12
8
4
350 370 390 410 430 450 470 490 510 530 550 570
λ

Figure 29. The fluorescence spectra of 7,8-benzoquinoline in 0.05 M $H_2SO_4$ as a function of 7,8-benzoquinoline concentration. Excitation wavelength--300 $m_\mu$. Method of illumination--frontal (totally absorbing solutions)

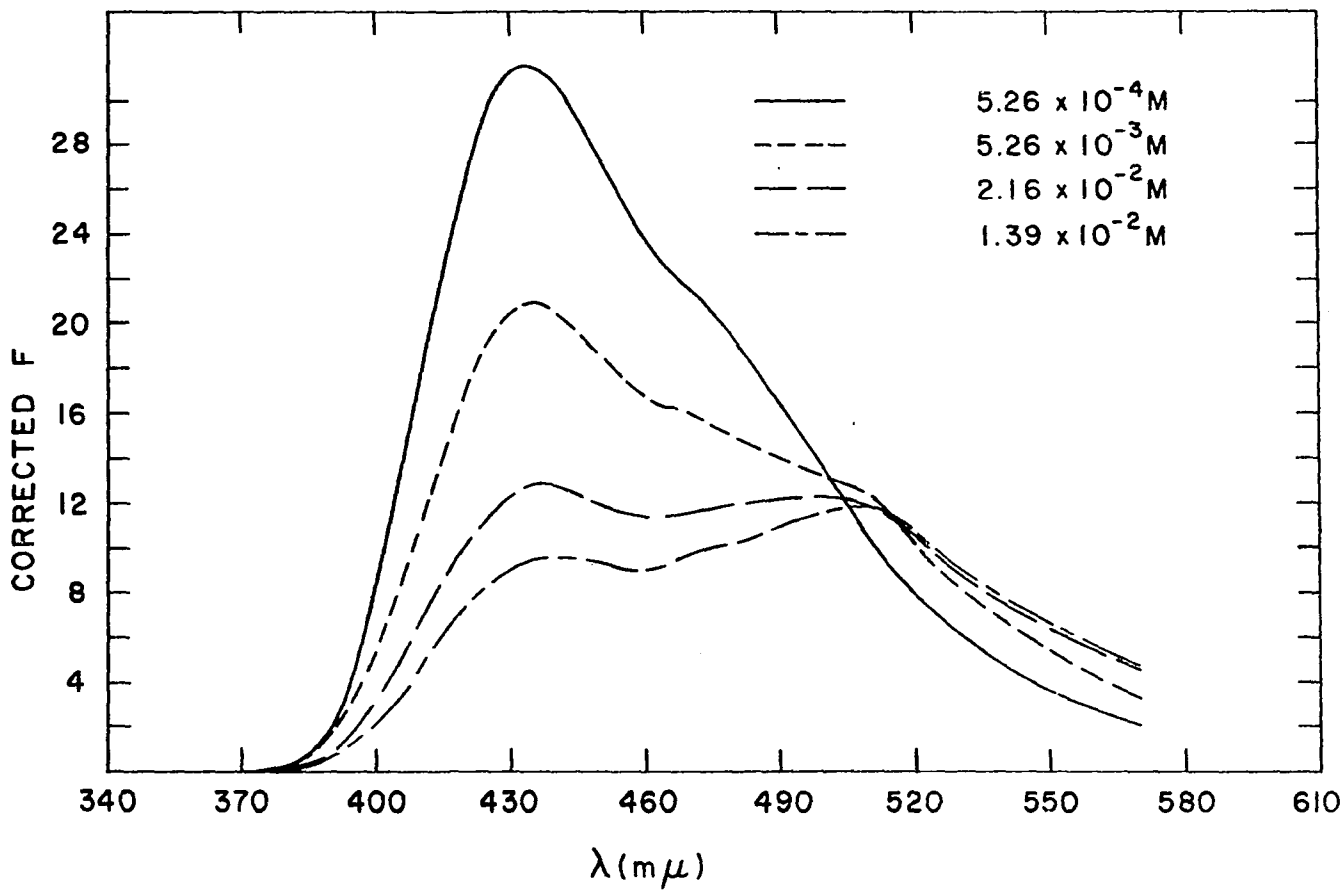
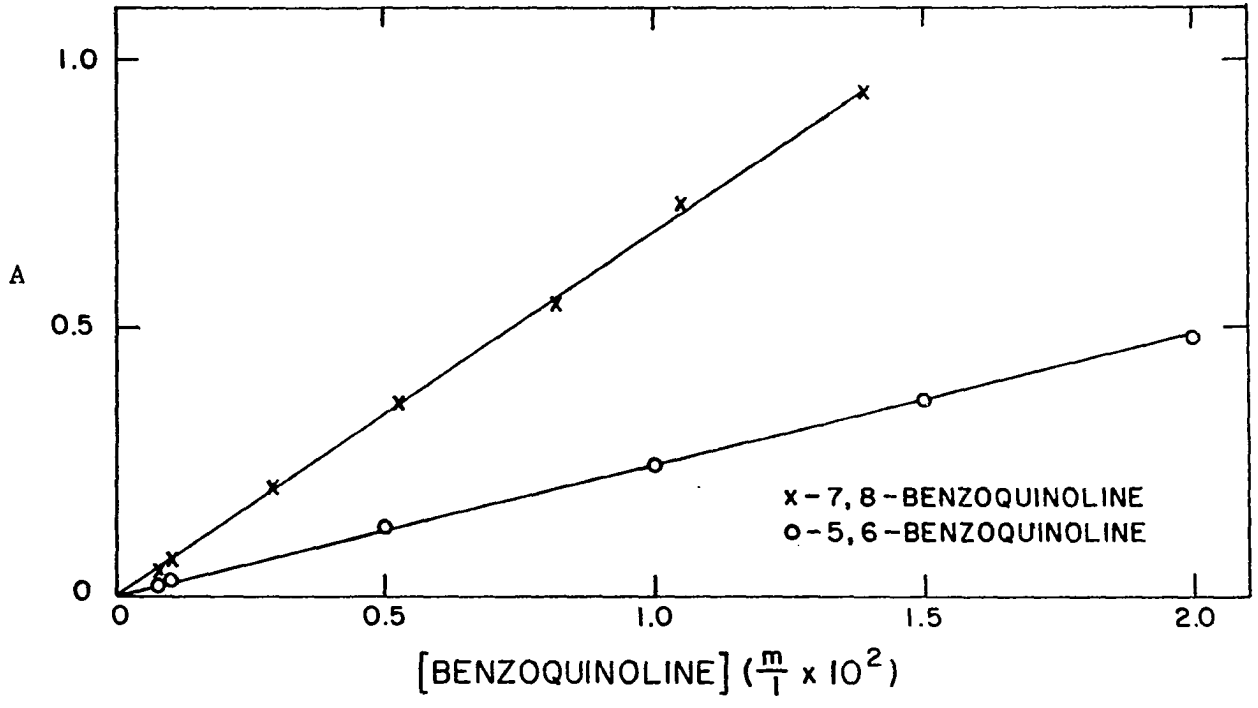

5.26 x 10^-4 M
5.26 x 10^-3 M
2.16 x 10^-2 M
1.39 x 10^-2 M
CORRECTED F
28
24
20
16
12
8
4
340
370
400
430
460
490
520
550
580
610
λ(mμ)

Figure 30. Beer's law plots for 5,6-benzoquinoline and 7,8-benzoquinoline in 0.05 M $H_2SO_4$. Wavelength for 5,6-benzoquinoline--360 $m\mu$. Wavelength for 7,8-benzoquinoline--360 $m\mu$. Path length--0.02 cm for 7,8-benzoquinoline. Path length $\sim$ 0.005 cm for 5,6-benzoquinoline


1.0
0.5
0
A
0
0.5
1.0
1.5
2.0
x - 7, 8 - BENZOQUINOLINE
o - 5, 6 - BENZOQUINOLINE
[BENZOQUINOLINE] ($\frac{m}{l}$ x $10^2$)

from these data that the long wavelength bands are due to excimer species.

The self-quenching plots for 5,6-benzoquinoline and 7,8-benzoquinoline, determined by the method of totally absorbing solutions, are shown in Figure 31. The plots of the two compounds are remarkably similar and are seen to obey the Stern-Volmer law. The Stern-Volmer quenching constant is 113.6 for both compounds. The following mechanism is proposed to account for these data,

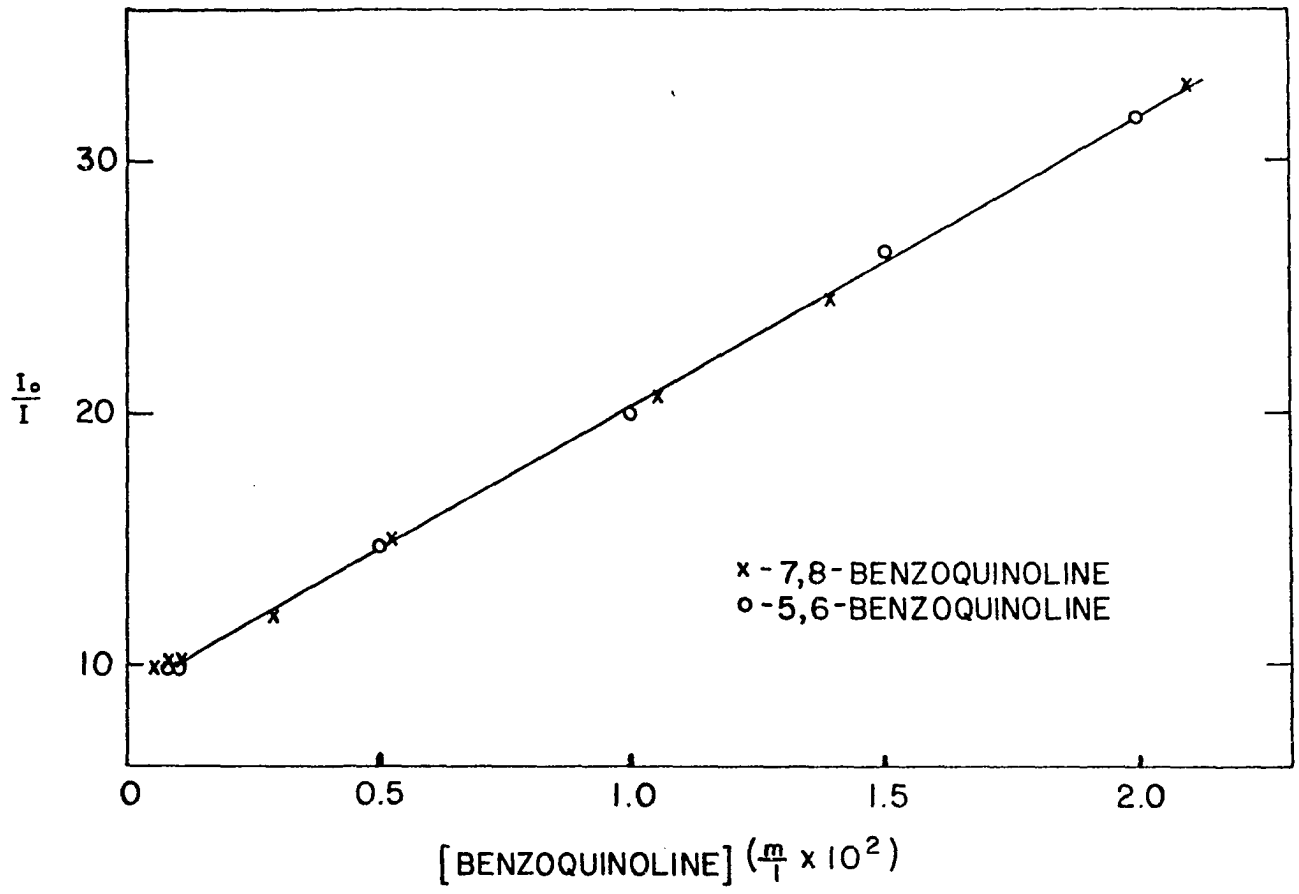
$$\begin{array}{ccccc} MH^{+*} & + & MH^{+} & \rightleftharpoons & DH_2^{+2*} \\ \downarrow & & & & \downarrow \\ h\nu & + & MH^{+} & & h\nu' + MH^{+} + MH^{+} \end{array} \qquad (22)$$

where,

$MH^{+}$ = the acidic form of 5,6-benzoquinoline or 7,8-benzoquinoline,
$D^{+2*}$ = an excited state dimer (excimer),
$h\nu$ = the fluorescence of the acidic form of 5,6-benzoquinoline and 7,8-benzoquinoline,
$h\nu'$ = excimer fluorescence for 5,6-benzoquinoline or 7,8-benzoquinoline,
* = a molecule in an excited state.

The relatively high values of the self-quenching constants indicate that the dissociation of the excimer species is not as significant as it is in the 1,10-phen system. Apparently the fluorescence of the excimer provides a rapid and efficient pathway for the molecule to return to the ground which competes favorably with the dissociation of the excimer.

Figure 31. The self-quenching of 5,6-benzoquinoline and 7,8-benzoquinoline in 0.05 M $H_2SO_4$. Excitation wavelength--300 $m\mu$. Fluorescence wavelength--430 $m\mu$. Method of illumination--frontal (totally absorbing solutions)


30
20
10
$\frac{I_o}{I}$
0
0.5
1.0
1.5
2.0
[BENZOQUINOLINE] ($\frac{m}{l}$ x $10^2$)
x - 7,8-BENZOQUINOLINE
o - 5,6-BENZOQUINOLINE

# VII. THE REACTION OF 2,2'-BIPYRIDINE WITH THE HYDROGEN ION

## A. Introduction

As 2,2'-bipyridine contains the same functional group as 1,10-phen, it would seem reasonable that it might exhibit some of the same ground state behavior, i.e., it ought to form aggregates in which more than one molecule of 2,2'-bipyridine is bonded to the hydronium ion. The existence of such species would be of some importance in that any accurate determination of stability constants of 2,2'-bipyridine metal complexes would require a knowledge of the conditions under which these species form. In addition, the existence of these species would contribute to a general understanding of aggregation with the hydronium ion. The purpose of this investigation will be to determine whether such aggregates exist and to attempt to describe the bonding in them.

## B. Experimental

### 1. Solubility data

The solubilities were determined by the same method used for 5,6-benzoquinoline and 7,8-benzoquinoline. Equilibration time was determined by monitoring the uv absorption spectrum of a solution in contact with the solid for one week and was found to be 48 hours. Agitation of the solution was accomplished with a Burrell wrist-action shaker. The saturated solutions were analyzed for 2,2'-bipyridine by measuring uv absorption spectra of these solutions after appropriate dilution and pH adjustment.

2. Absorption spectra

Absorption spectra were obtained with a Carey Model 14 spectrophotometer. Because of the high concentration of the solution and the high molar absorptivity of 2,2'-bipyridine, it was necessary to use very short path lengths to obtain the spectrum of the aggregate in Figure 33. This was accomplished by pressing a drop of the solution between two quartz plates to obtain a thin film of the solution. The spectrophotometric titration was accomplished with the Beckman Model DU spectrophotometer using a 180-ml tall-form beaker.

3. Reagents

2,2'-biypridine was obtained from Aldrich Chemical Co. and used as received for the solubility study. Melting point for this material is 69 to 70°C. The literature value is 70°C. The 2,2'-biypyridine used for the absorption data was recrystallized from ether at dry-ice acetone temperatures and gave a melting point of 70°C. All other chemicals were reagent grade.

4. Miscellaneous

The temperature for these experiments was 23° to 25°C. Class A glassware was used in all experiments.

## C. Results and Discussion

A solubility study was employed to determine whether 2,2'-bipyridine forms aggregates with the hydronium ion in concentrated solutions. The details of using solubilities to determine whether or not aggregates exist have been discussed in Part VI of this thesis. The solubility of

2,2'-bipyridine was measured as a function of HCl concentration from 0.01 M to 0.05 M. A linear relationship between solubility and HCl concentration was obtained with a slope of 1.45, indicating that 2,2'-bipyridine forms aggregates with the hydronium ion. These data are shown in Figure 32. A spectrum of a solution containing aggregates, prepared by saturating a 0.00989 M HCl solution with 2,2'-bipyridine, was compared to the spectrum of a dilute solution at the same pH. These data are shown in Figure 33. The spectrum containing aggregates is shifted slightly toward the acid form. In general, the spectral differences are small when compared to the spectra of the acidic and basic forms, which is indicative of a complex similar to that proposed for 1,10-phen and 5,6-benzoquinoline. This structure for the aggregate places 2,2'-bipyridine molecules in the secondary hydration sphere of the hydronium ion.

A concentrated solution of 2,2'-bipyridine (0.035 M) was titrated spectrophotometrically to determine the composition of the aggregate. The titration is shown in Figure 34. An extrapolation of the linear portions of the titration curve gave a break at a ratio 2.5 to 1 2,2'-bipyridine to HCl. The broken line in Figure 34 is the approximate behavior expected if no aggregates formed. The 2.5:1 ratio is interpreted as an indication that both the 3:1 and 2:1 aggregates co-exist over a reasonably wide pH range. The determination of the stability constants for this system is left as an area for future research as the objectives of this research were to indicate that aggregation with the hydronium ion is a general phenomenon with nitrogen heterocycles.

Figure 32. The molar solubility of 2,2'-bipyridine as a function of the total molar concentration of HCl

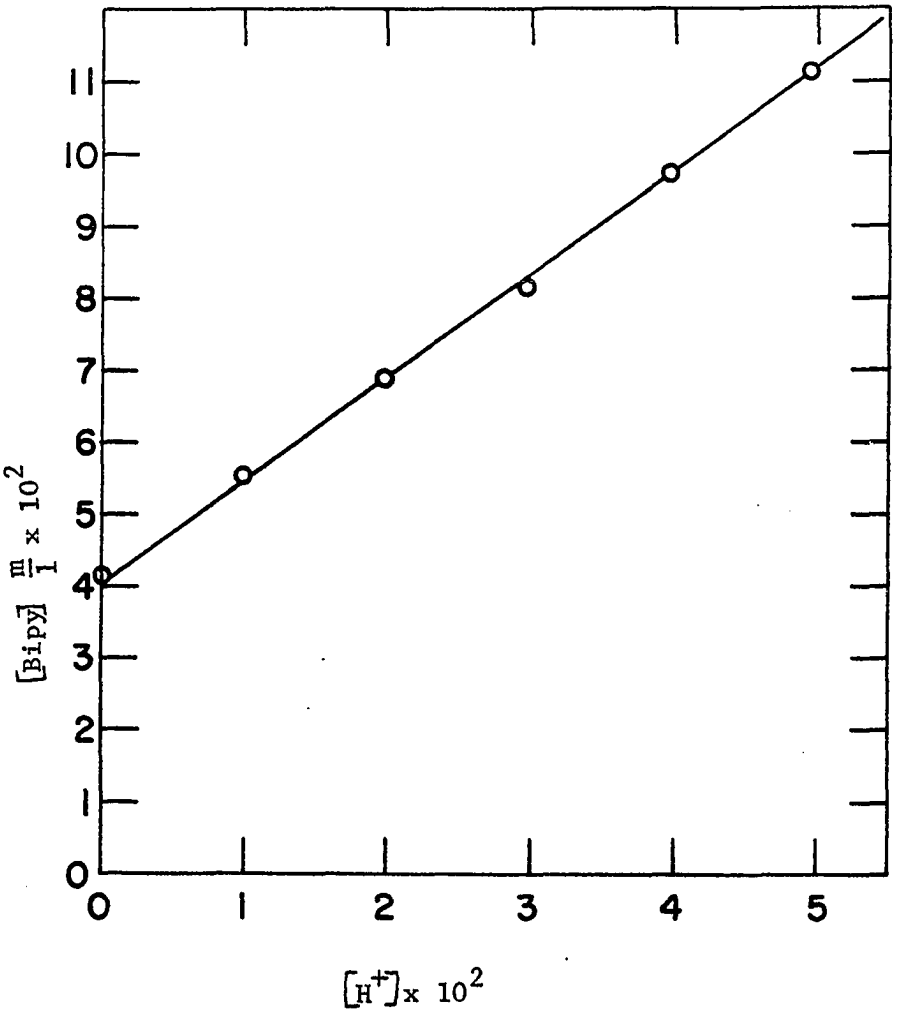

11
10
9
8
7
6
5
4
3
2
1
0
0
1
2
3
4
5
[Bipy] $\frac{m}{l}$ x $10^2$
$[H^+]$x $10^2$

Figure 33. The absorption spectra of 2,2'-bipyridine in aqueous solution. The total concentration of 2,2'-bipyridine in the dilute solutions is $1.023 \times 10^{-4}$ M and approximately 0.054 M in the concentrated solution

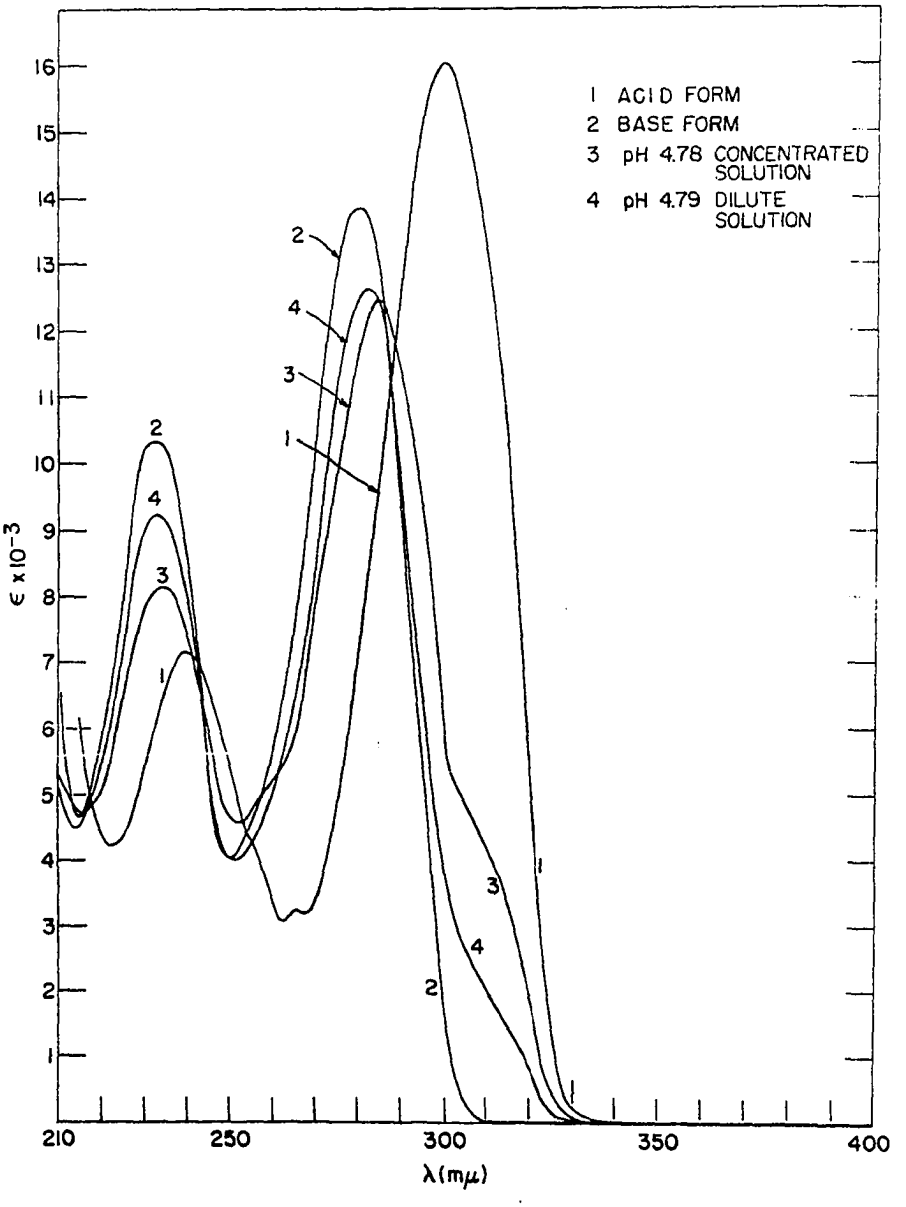
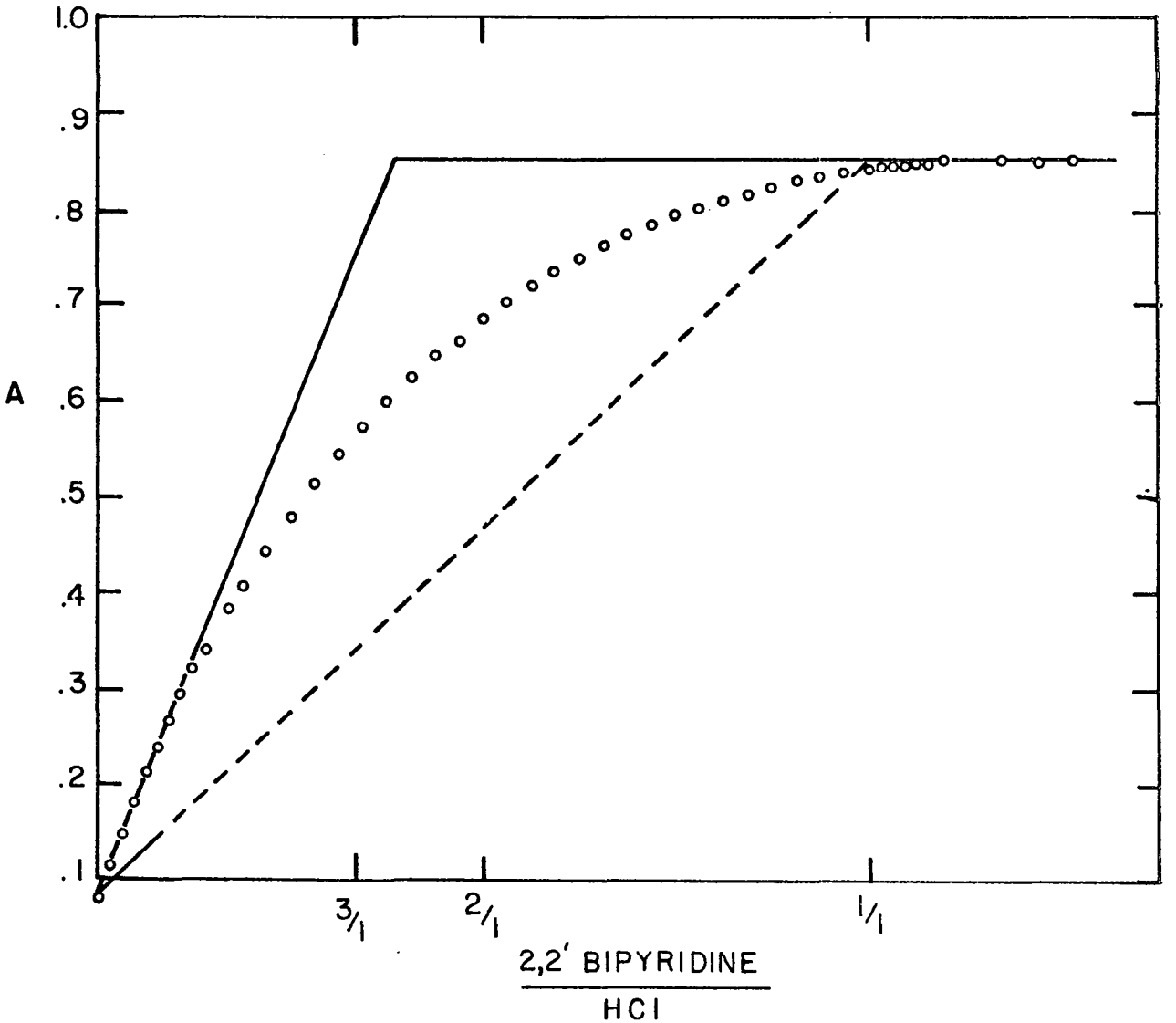

1 ACID FORM
2 BASE FORM
3 pH 4.78 CONCENTRATED SOLUTION
4 pH 4.79 DILUTE SOLUTION
ε x 10⁻³
λ(mμ)
210
250
300
350
400

Figure 34. A spectrophotometric titration of 2,2'-bipyridine with HCl at 330 $m\mu$. The absorbances are corrected for dilution by the titrant


1.0
.9
.8
.7
A .6
.5
.4
.3
.2
.1
3/1
2/1
1/1
2,2' BIPYRIDINE
HCl

# VIII. THE QUANTUM EFFICIENCIES OF 1,10-PHENANTHROLINE, 5,6-BENZOQUINOLINE, AND 7,8-BENZOQUINOLINE

## A. Introduction

The fluorescence quantum efficiency, which is the number of quanta emitted divided by the number of quanta absorbed, is an important and fundamental measurement in fluorescence spectroscopy. In fluorescence spectroscopy the fluorescence quantum efficiency is the equivalent of the molar absorptivity in absorption spectroscopy. In analytical applications, the quantum efficiency serves to define detection limits and the extent to which a molecule may interfere with a determination. Quantum efficiencies are also instructive in revealing the nature of the fluorescence process and the processes competing with it. The quantum efficiency of 1,10-phen has not been reported in aqueous solutions, and this study would not be complete without this determination. The quantum efficiency of 5,6-benzoquinoline has been reported by Nakamizo (55), using anthracene as the standard. Kilimov (38) has determined the quantum efficiency for both 5,6-benzoquinoline and 7,8-benzoquinoline, but the details were unavailable.

## B. Experimental

### 1. Determination of the quantum efficiencies

A comparative method was employed to determine the quantum efficiencies. The method is based on the assumption that the integrated area of a corrected fluorescence spectrum is proportional to the total number of quanta emitted. Comparing the integrated area of the corrected

fluorescence spectrum with that of a standard, whose quantum efficiency is known, allows for the calculation of the quantum efficiency after correction for the differences in light absorbed for the two compounds. A simple formula which takes all of this into account is given by (18),

$$\frac{Q_1}{Q_2} = \frac{\phi_2}{\phi_1} \times \frac{F_1}{F_2} \times \frac{A_2}{A_1} \qquad (23)$$

where Q is the quantum efficiency, F is the integrated area, A is the absorbance of the solution at the excitation wavelength, and $\phi$ is the relative photon output at the excitation wavelength. Quinine was used as the standard for these determinations because it has been extensively studied for this purpose and is generally conceded to be an excellent standard. Chen (18) has recently given an excellent evaluation of quinine for this purpose.

2. Preparation of solutions

All solutions were prepared by the dilution of concentrated stock solutions prepared by dissolving a weighed amount of the compounds. The quinine sulfate was dissolved in 0.05 M $H_2SO_4$. The concentration of the quinine was determined by measuring uv absorption spectra of the solution and using the molar absorbitivities reported by Chen (18). The pH adjustment of the solutions of the nitrogen heterocycles was accomplished by the addition of micro-amounts of concentrated $H_2SO_4$ or KOH, using a Beckman Expanded Scale pH meter to determine the final pH.

3. Reagents

1,10-Phen, 5,6-benzoquinoline, and 7,8-benzoquinoline were obtained

from the Aldrich Chemical Co. 1,10-Phen was recrystallized from $H_2O$ and analyzed for water content by thermogravimetry. Purissimum grade 5,6-benzoquinoline was used as received. 7,8-Benzoquinoline was recrystallized from ether at dry-ice acetone temperatures. The resulting product melted sharply at 50°C. NF-grade quinine sulfate was obtained from Mallencknodt Chemical Co. and was recrystallized twice from hot water. All other chemicals were reagent grade.

4. Fluorescence and absorption measurements

The methods for obtaining corrected fluorescence spectra have previously been described in this thesis and will not be repeated here. Right angle illumination was used with 4-mm cylindrical quartz cells. Absorption spectra were obtained with instruments previously described in this thesis.

5. Miscellaneous

The temperature was 25°C in all determinations. Class A glassware was used throughout.

## C. Results and Discussion

The quantum efficiencies determined in this work are shown in Table 2 along with the values for 5,6-benzoquinoline reported by Nakamizo (55).

The absorbances for all determinations are less than 0.05, so that the fluorescence response is linear with concentration and self-absorption of the fluorescence is minimized. A value of 0.505 was used in the calculations for the quantum efficiency of quinine sulfate in accordance

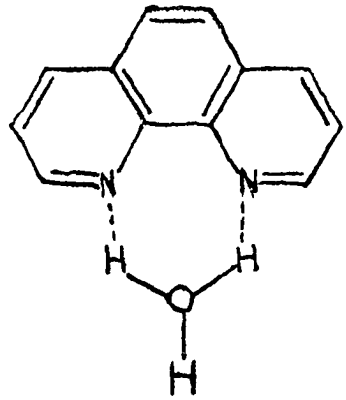
Table 2. The quantum efficiencies of 1,10-phenanthroline, 5,6-benzoquinoline, and 7,8-benzoquinoline

| | Excitation wavelength ($m\mu$) | Concentration $\frac{M}{l}$ | Form | Quantum efficiency |
|---|---|---|---|---|
| 1,10-phen | 310 | $1.98 \times 10^{-5}$ | Acidic | 0.007 |
| | 300 | $1.98 \times 10^{-5}$ | Basic | 0.017 |
| 5,6-benzoquinoline | 310 | $3.2 \times 10^{-5}$ | Acidic | 0.49 |
| | 310 | $3.2 \times 10^{-5}$ | Basic | 0.41 |
| | Not reported | $10^{-5}$ | Acidic | 0.43* |
| | Not reported | $10^{-5}$ | Basic | 0.41* |
| 7,8-benzoquinoline | 310 | $2.59 \times 10^{-5}$ | Acidic | 0.308 |
| | 310 | $2.59 \times 10^{-5}$ | Basic | 0.351 |

*Values reported by Nakamizo (55).

with the data of Chen (18). The agreement between the values for 5,6-benzoquinoline in this work and those of Nakamizo (55) is quite good in view of the error expected in the determination of quantum efficiencies. The most striking feature of these determinations is the low values obtained from the forms of 1,10-phen in comparison to the values for 7,8-benzoquinoline. This is also reflected in the low molar absorptivity of 1,10-phen at long wavelengths compared to 7,8-benzoquinoline. This suggests that the lowest energy transitions in 1,10-phen perhaps is $\pi^* \longrightarrow n$, which is the fluorescence transition. This could occur if only one of the nitrogens in 1,10-phen is strongly solvated, leaving the n state of the other nitrogen higher in energy than the $\pi$ states.

Charton (16) has proposed this to account for the compliance of the $pKa_1$ values of the substituted 1,10-phenanthrolines to the extended Hammet equation. This is in contrast to the usual concept depicted below.



The data of Langmuir (42) lend some support to these proposals. Langmuir found that the long wavelength absorption band of 1,10-phen in cyclohexane shifts toward shorter wavelengths with the addition of glacial acetic acid, a proton donor. This is characteristic of a $\pi^* \longrightarrow n$ transition. In addition, 1,10-phen was found to be non-fluorescent in the absence of the proton donor and fluorescent in the presence of it, giving the fluorescence spectrum of the acid form. These data indicate that in the absence of solvation of the nitrogens by a proton donor, the lowest energy transition is $\pi^* \longrightarrow n$.

Story (69) has recently determined that the relative quantum efficiency of 1,10-phen is approximately twenty times greater in the tris(1,10-phen)zinc(II) complex than in the acidic form. In this complex the n states of both nitrogens are expected to be lower in energy than the $\pi$ states. The lowest energy transition would then be $\pi^* \longrightarrow \pi$,

a fully allowed transition. These data are also consistent with the above proposals.

It is expected then that the quantum efficiency of the diprotonated form of 1,10-phen would verify the above proposals, because in this species both nitrogens are presumably protonated. Jones (35) has measured the intensity of the 1,10-phen fluorescence over a wide range of acid concentrations. These intensities may be taken as a rough indication of the quantum efficiency of the diprotonated form of 1,10-phen relative to the acid form. Upon initial formation of the diprotonated form, the relative quantum efficiency decreases and then increases with increasing acid concentration to about twenty times that of the simple acid form in 70% perchloric acid. Jones (35) showed, however, that water acts as a quencher for the diprotonated form by correlating the fluorescence intensity with the decreasing water concentration in the concentrated acid. While one is tempted to conclude that the increased quantum efficiency in solutions of concentrated acid is due to the solvation of both nitrogens, it is evident that the quenching by water also affects the quantum efficiency in this system. In short, the formation of the diprotonated specie involves going to acid concentrations which result in a change of the character of the solvent system, making it difficult to interpret the data.

It is evident that these data are not conclusive. A more sophisticated approach is required which will distinguish between the basic nature of the transition and various solvent effects. Polarized fluorescence in an appropriate solvent would seem to be a good approach to this problem. It is interesting to note that the other compounds reported

to exhibit $\pi^* \longrightarrow n$ fluorescence, the diazines (5) and 5,6-diaza-phenanthrene (46), are also nitrogen heterocycles containing two nitrogens.

# IX. SUGGESTIONS FOR FUTURE WORK

The aggregation of 1,10-phen, 5,6-benzoquinoline, and 2,2'-bipyridine with the hydronium ion would seem to indicate that this is a rather general phenomenon with nitrogen heterocycles. A general study of this phenomenon with a variety of nitrogen heterocycles would be instructive in further elucidating the chemistry of the hydronium ion. In addition, the stability constants for the 2,2'-bipyridine aggregates ought to be determined in view of the analytical importance of this compound.

It is also evident that excimer fluorescence is probably possible for most nitrogen heterocycles whose parent hydrocarbons exhibit excimer fluorescence. Organic solvents, in which these compounds are more soluble than they are in aqueous solution, are suggested for future studies. In addition the solvents should allow for a variation of temperature over a wide range. The temperature dependence of the ratio of excimer fluorescence to monomer fluorescence is most helpful in characterizing these systems.

Finally the development of 1,10-phen as a reagent for the fluorescence analysis of trace amounts of metals should be undertaken. The platinum metals as well as zinc and cadmium would be good prospects.

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# XII. APPENDIX

The purpose of these derivations is to obtain expressions for fluorescence self-quenching in competition with another quencher. Two cases will be considered, reversible self-quenching and irreversible self-quenching.

## A. Irreversible Self-Quenching

The derivation will relate to the following situation in concentrated solutions:

$$[M]_2 + [Q] \xleftarrow{k_3[Q]} [M^*]_2 + [M]_2 \xrightarrow{k_1} [D^*] \longrightarrow [M]_2 + [M]_2$$

$$[M^*]_2 \xrightarrow{k_f} h\nu + [M]_2 \qquad [M^*]_2 \xrightarrow{k_{I1}} [M]_2 \tag{1A}$$

where,

$[M]_2$ = a monomeric species with the subscript indicating a given concentration,

$[D^*]$ = an excited state dimeric species,

$[Q]$ = a quencher at concentration Q,

$k_{I1}$ = the rate constant for intersystem crossing, internal conversion, and solvent quenching,

$k_3$ = the rate constant for the fluorescence quenching by $[Q]$,

$k_1$ = the rate constant for self-quenching, and

$k_f$ = the fluorescence rate constant.

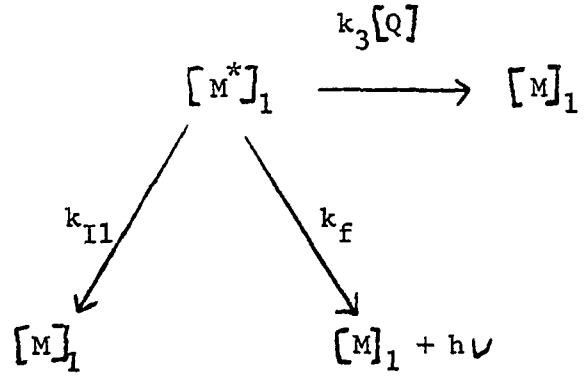
An asterisk denotes the molecule is in an excited state. Since the fluorescence is constant with time, the steady state approximation may be made.

$$\frac{d[M^*]_2}{dt} = 0 = I_a - k_{I1}[M^*]_2 - k_f[M^*]_2 - k_1[M^*]_2[M]_2 - k_3[M^*]_2[Q] \tag{2A},$$

where $I_a$ is the rate of formation of $[M^*]_2$. The rate of formation is a function of the emission rate of the excitation unit and the absorbance of the solution. Using the method of totally absorbing solutions, discussed in detail in the Experimental Section, $I_a$ is independent of the concentration if the excitation wavelength is held constant. Rearranging and factoring out $[M^*]_2$ gives,

$$I_a = [M^*]_2 \left\{ k_{I1} + k_f + k_1[M]_2 + k_3[Q] \right\}. \tag{3A}$$

A term is required for the fluorescence intensity in the absence of quenching. The case where the quencher, $[Q]$, is present in the absence of self-quenching at a constant concentration is described by the following:




(4A)

The notation is the same as that used in (1A). Applying the steady state approximation gives,

$$0 = \frac{d[M^*]_1}{dt} = I_a - k_{I1}[M^*]_1 - k_f[M^*]_1 - k_3[M^*]_1[Q].$$

$I_a$ is equal to the $I_a$ used in (2A), as these equations pertain to totally absorbing solutions using the frontal illumination apparatus. This condition will be true for the remainder of these derivations. Rearranging and factoring gives,

$$I_a = [M^*]_1 \left\{ k_{I1} + k_f + k_3[Q] \right\}. \tag{5A}$$

Dividing (3A) by (5A) gives,

$$\frac{I_a}{\bar{I}_a} = \frac{M^*_2 \left\{ k_{I1} + k_f + k_1[M]_2 + k_3[Q] \right\}}{M^*_1 \left\{ k_{I1} + k_f + k_3[Q] \right\}} , \tag{6A}$$

which upon rearrangement and dividing through gives,

$$\frac{[M^*]_1}{[M^*]_2} = 1 + \frac{k_1[M]_2}{k_{I1} + k_f + k_3[Q]} . \tag{7A}$$

Since $I^o = k_f[M^*]_1$ and $I = k_f[M^*]_2$, $\frac{[M^*]_1}{[M^*]_2} = \frac{I^o}{I}$, where $I^o$ is the fluorescence intensity in the absence of self-quenching and I is the intensity at concentration $[M]_2$. Making the appropriate substitution gives,

$$\frac{I^o}{I} = 1 + \frac{k_1[M]_2}{k_{I1} + k_f + k_3[Q]} \tag{8A}$$

The following gives the expression for the fluorescence intensity in the absence of self-quenching and the quencher, Q:

$$\begin{array}{l} [M^*]_1 \xrightarrow{k_{I1}} [M]_1 \\ \quad \downarrow k_f \\ [M]_1 + h\nu \end{array} \tag{9A}$$

As in the above derivations,

$$\frac{d[M^*]_1}{dt} = 0 = I_a - k_f[M^*]_1 - k_{I1}[M^*]_1 \tag{10A}$$

Rearranging and dividing (3A) by (10A) yields after the substitution for $\frac{[M^*]_1}{[M^*]_2}$,

$$\frac{I^o}{I} = 1 + \frac{k_1}{k_f + k_{I1}}[M]_2 + \frac{k_3}{k_f + k_{I1}}[Q]_N , \tag{11A}$$

where $[Q]_N$ denotes that this is a variable in this expression.

## B. Reversible Self-Quenching

In order to conserve space, the same notation will be used as above, and the derivations will be given in less detail.

Reversible self-quenching in the presence of a competing quencher, Q , is described by the following expression:

$$\begin{array}{ccccccc} [M]_2 & \xleftarrow{k_{I1}} & [M^*]_2 + [M]_2 & \underset{k_2}{\overset{k_1}{\rightleftharpoons}} & [D^*] & \xrightarrow{k_{I2}} & [M]_2 + [M]_2 \\ & \swarrow^{k_f} \quad \searrow_{k_3[Q]} & & & \searrow_{k_4[Q]} & & \\ h\nu + [M]_2 & & [M]_2 & & [M]_2 + [M]_2 & & \end{array} \quad , \tag{12A}$$

where,

$[D^*]$ = an excited state dimer,

$k_2$ = rate constant for the dissociation of the excited dimer, $D^*$ ,

$k_{I2}$ = rate constant for intersystem crossing, internal conversion, and solvent quenching, and

$k_4$ = rate constant for the quenching of the excited dimer, $[D^*]$ by the quencher, $[Q]$.

All other constants have been previously defined. If the excited dimer, $[D^*]$, is fluorescent, an additional term must be included to account for this. Using the steady state approximation,

$$0 = \frac{d[M^*]_2}{dt} = I_a - k_f[M^*]_2 - k_3[Q][M^*]_2 - k_{I1}[M^*]_2 - k_1[M^*]_2[M]_2 + k_2[D^*] \tag{13A}$$

and,

$$\frac{d\,D^*}{dt} = 0 = k_1[M^*]_2[M]_2 - k_{I2}[D^*] - k_4[Q][D^*] - k_2[D^*] \tag{14A}$$

Solving for $[D^*]$ in (14A) gives,

$$D^* = \frac{k_1 [M^*]_2 [M]_2}{k_{I2} + k_2 + k_4 [Q]} \qquad (15A)$$

Substituting (15A) and (14A) and factoring gives,

$$I_a = [M^*]_2 \left\{ k_f + k_{I1} + k_3 [Q] + k_1 \left[ 1 - \frac{k_2}{k_{I2} + k_2 + k_4 Q} \right] [M]_2 . \right\} \qquad (16A)$$

A dilute solution in the presence of a constant amount of quencher, Q, is described by equation (5A). Dividing (16A) by (5A) and substituting for $\frac{[M^*]_1}{[M^*]_2}$ gives,

$$\frac{I^o}{I} = 1 + \frac{k_1}{k_f + k_{I1} + k_3 [Q]} \left[ 1 - \frac{k_2}{k_2 + k_{I2} + k_4 [Q]} \right] [M]_2 \quad . \qquad (17A)$$

The terms involving [Q] are constants in (17A). A dilute solution in the absence of both self-quenching and the quencher, [Q], is described by equation (10A). Rearranging (10A) and dividing (16A) by (10A) gives after substitution for $\frac{[M^*]_1}{[M^*]_2}$,

$$\frac{I^o}{I} = 1 + \frac{k_3}{k_f + k_{I1}} [Q]_N + \frac{k_1}{k_f + k_{I1}} \left[ 1 - \frac{k_2}{k_{I2} + k_2 + k_4 [Q]_N} \right] [M]_2 \quad , \qquad (18A)$$

where $[Q]_N$ is a variable.