UNIVERSITY OF WISCONSIN LIBRARY
MANUSCRIPT THESES

In presenting this thesis in partial fulfillment of the requirements for an advanced degree at the University of Wisconsin I agree that the Library shall make it freely available for inspection. I further agree that permission for extensive copying or publication of this thesis for scholarly purposes may be granted by my major professor, Dr. Takeru Higuchi, or, in his absence by the Dean of the Graduate School. It is understood that any copying or publication of this thesis for financial gain shall not be permitted without my written permission.

June 3, 1954
Date

Author

FOR BORROWERS OF THIS THESIS

Unpublished theses deposited in the University of Wisconsin Library may be used only with due regard for the rights of the authors. Permission for the extensive copying or publication of this thesis in whole or in part must be obtained in advance in all cases from the major professor named above, or, in his absence, from the Dean of the Graduate School. Extensive copying or publication of this thesis for which there might be financial return to the borrower must have the written permission of the author.

This thesis has been used by the following persons, whose signatures attest their acceptance of the above restrictions. A library which borrows this thesis for the use of its patrons is required to secure the signature of each user.

Name & Address of User	Date Used

THE APPLICATION OF NEWER ANALYTICAL PROCEDURES TO PHARMACEUTICAL ANALYSIS

by

AARON DAVID COOPER

A Thesis Submitted in Partial Fulfillment
of the Requirements for the Degree of

DOCTOR OF PHILOSOPHY

at the

UNIVERSITY OF WISCONSIN

1954

ACKNOWLEDGMENTS

The author wishes to express his sincere appreciation to Dr. Takeru Higuchi for suggesting this study and his guidance in directing it to its present status.

Gratitude is expressed to the American Foundation for Pharmaceutical Education for financial assistance to the author during the course of this study.

Gratitude is also expressed to the Wisconsin Alumni Research Foundation and other organizations which made possible the purchase of specialized apparatus.

TABLE OF CONTENTS

INTRODUCTION

The use of acid-base titrations in nonaqueous solvents is a relatively well established procedure which is employed to a considerable extent as a control method in pharmaceutical industry. Unfortunately, the theoretical aspects underlying the practical applications have been, for the most part, neglected. In the present investigation, a fundamental study of acids, bases, salts, and indicators has been carried out with the hope of clarifying some of the obscure principles.

Because of the size of this research field, the study reported herein is limited to the following three phases of acid-base equilibria in acetic acid:

(1) A potentiometric investigation of relative basicity with emphasis on the development of the relatively more alkaline regions in acetic acid by the use of "stronger" bases.

(2) A spectrophotometric evaluation of indicator constants in terms of the Hammett Acidity Function, aimed at finding indicators which undergo sharp color transitions in the relatively more alkaline regions of acetic acid.

(3) A potentiometric survey of various acid-base indicators to determine their potentialities for titrimetry purposes in acetic acid.

It is the hope of the author, that with the experimental data accumulated and unpublished information available to him, that a clearer understanding of some of the fundamental relationships governing the use of acetic acid as a solvent for analytical purposes may be attained.

A GENERAL REVIEW OF ACID-BASE BEHAVIOR

The theory of electrolytic dissociation as proposed by Arrhenius held a prominent position in the field of acid-base equilibria for over fifty years and was generally accepted by a large percentage of chemists. There were some who realized that this theory was inadequate, being applicable only to aqueous media and of no value when applied to neutralization reactions in other solvents. Unfortunately for these latter few, the inconsistancies which they observed remained unexploited for a number of years until the newer theories of acids and bases were postulated.

Presumably, the work of Kahlenberg (1) in 1902 represents the first significant departure from the Arrhenius theory of electrolytic dissociation. He was able to demonstrate that "ionic" type reactions took place in anhydrous benzene solutions between metallic oleates and dry hydrochloric acid gas, while at the same time the conductivity of the solutions remained virtually the same, both before and after the reaction took place. Kahlenberg concluded that instantaneous chemical reactions could occur in solutions which were excellent insulators, and that the presence of ions was not an absolute necessity. In the following year Vorlander (2), while studying the color changes occurring with indicators of different structural relationships, described the titration of aniline with hydrogen chloride in benzene, using p-dimethylaminoazobenzene ("butter yellow") as the indicator.

The earliest significant work relating to analytical titrations in nonaqueous solvents is ascribed to Folin, Flanders, and Wentworth during the years 1910 to 12 (3-5). These men realized the necessity of using nonassociating solvents to obtain sharp indicator endpoints in the titration of various higher fatty acids with sodium ethoxide. Among the solvents which they employed were chloroform, carbon tetrachloride, benzene, and toluene, while phenolphthalein was used as the indicator. They further showed that the addition of any oxygenated solvents (basic solvents containing unshared electron pairs) to the medium markedly affected the nature of the solution so that sharp endpoints were no longer obtainable; they then proceeded to question the theory of ionization when applied to all solvent systems.

Due to this adherence to the Arrhenius ionization theory, the work of these men (1-5) and others (6-8) was generally overlooked until 1923. In that year, Hantzsch (9) reported a study on the nature of acids and acidic strengths which represented a departure from prevailing theory. At the same time, Brønsted (10) published his preliminary investigation on the protonic theory of acids and bases which was substantiated early the next year in an independent study carried out by Lowry (11). In 1927 Hall and Conant (12,13) published the first of their now famous series of papers dealing with "super acids" and acid-base relationships in nonaqueous solvents.

THE CONCEPTS OF ACIDS AND BASES

Before proceeding to a discussion of acid-base relationships in acetic acid it is desirable to examine briefly the various concepts of acids and bases, along with defining the part which the solvent plays in the development of their intrinsic character. The intriguing question of exactly what constitutes an acid or base has been, and still is, a subject of considerable controversy, with no less than five principle theories of acids and bases having been postulated. In view of this, the following is intended as a brief review of the various acid-base theories along with a classification of the different solvent types and how these affect the intrinsic strength of acids and bases present in them.

The Arrhenius Theory

Following the introduction of the Arrhenius theory of electrolytic dissociation, acids and bases were defined as substances containing hydrogen and hydroxyl ions, respectively, and which undergo dissociation in aqueous solutions to yield the free ions. The term neutralisation constituted an extension of these definitions and represented the reaction between an acid and base in aqueous solution, resulting in the formation of a salt and water according to reaction (1).

$$\underset{\text{(acid)}}{H^+} + \underset{\text{(base)}}{OH^-} \longrightarrow H_2O \qquad (1)$$

This hydrogen-hydroxyl theory provided a useful knowledge of acid-base character at the time of its inception, however its limitations with regard to the solvent water being necessary as the ionising medium were soon recognised by a number of scientists.

The Solvent System Theory (14-17)

The solvent system theory was first employed by Franklin (14,15) in 1905, though not recognised as such, and later extended by himself and Germann (16,17) in the years that followed. Like the Arrhenius theory, it is dependent upon the presence of a solvent, with acids and bases being defined in terms of solvent cation and anion concentrations. Those substances which enhance the cation concentration of the solvent are designated as acids, while those enhancing the solvent anion concentration are bases. The formal analogy to the Arrhenius theory is seen in reactions (2) and (3), the major difference being that ionization is not restricted to aqueous solutions. It is evident from reaction (3), that the addition

In water: $$H_2O + H_2O \rightleftharpoons H_3O^+ + OH^- \qquad (2)$$

In liquid NH_3: $$NH_3 + NH_3 \rightleftharpoons NH_4^+ + NH_2^- \qquad (3)$$

of compounds containing ammonium ions to such a system enhances its acidity, whereas the addition of molecules containing the amide species enhance the basicity. Unfortunately, like the Arrhenius theory, the definition of acids and bases is again dependent upon the presence of a solvent; it represents, however, an advancement in that ionization by solvolysis is not restricted to aqueous solutions.

The Brønsted-Lowry Theory (10,11)

The protonic theory as developed by Brønsted and Lowry, independently of each other, maintains that any substance, to function as an acid, must be capable of furnishing protons, whereas a base must be a species which readily coordinates or accepts available protons. In reaction (4), HA is designated as an acid because of its ability to split into a proton and the conjugate base, A^-, of the acid HA.

$$HA \longrightarrow H^+ + A^- \qquad (4)$$

As the proton cannot exist as a free entity in solution, reaction (4) is unable to proceed to the right unless a proton acceptor is present which intrinsically has a greater affinity for the proton than the conjugate base with which it originally is associated. As a result, the following equilibria must be satisfied in order for a substance to behave explanation of acid-base phenomenon. Unlike other theories, the Lewis definition of an acid or base is not dependent upon the presence of any particular element or solvent, but on the ability of ions or molecules to form coordinate covalent bonds through available unshared electron pairs.

An acid, according to Lewis, is any substance capable of combining with an unshared electron pair to form a coordinate covalent bond, while a base is any substance which possesses the unshared electron pair which is utilized in bond formation. Lewis proposed four fundamental experimental criteria to serve as a basis for his definition of acids and bases. These criteria include neutralization, titration with indicators, displacement, and catalysis, all four of which are discussed in detail by Luder and Zuffanti (18). All four criteria must be exhibited by a molecule or ion in order for the substance to qualify under Lewis' proposed definitions.

Boron triflouride, for example, is a strong Lewis acid because of the electron deficiency in the shell of the boron atom, while ammonia is a typical base because it possesses an unshared electron pair with which boron triflouride can readily combine.

$$F_3B + :NH_3 \longrightarrow F_3B:NH_3 \qquad (8)$$

THE CLASSIFICATION OF SOLVENT TYPES

The various solvents employed for acid-base titrimetry may be classified into four main categories based upon their behavior with regards to proton affinities and how they tend to influence the strength of acids and bases dissolved therein. The following treatment has been compiled from several sources (61,62).

PROTOPHILIC solvents are those possessing unshared electron pairs, thereby having a high proton affinity and being capable of converting a large number of hydrogen compounds into the corresponding onium ions. The amines are typical examples of protophilic solvents, possessing a high electron density on the nitrogen atom, which readily allows coordination with a proton. Protophilic solvents tend to enhance the acidic properties of weakly ionised hydrogen compounds by nearly complete interaction with the acid, resulting in the formation of the onium ion, and increasing the solvent cation concentration as shown in reaction (9).

$$HA + RNH_2 \longrightarrow RNH_3^{+} + A^{-} \qquad (9)$$

AMPHIPROTIC solvents, i.e. water and alcohols, are capable of acting either as acids or bases depending upon environmental conditions. Solvents of this type lie intermediate between the protogenic and protophilic solvents and can generally be characterised by some extent of hydrogen bonding. Water, though an amphiprotic solvent, possesses strong protophilic properties which makes possible nearly complete interaction with strong mineral acids, "leveling" them to the same degree of acidity through formation of the hydronium ion. It is because of this phenomenon, that a solvent such as acetic acid must be employed to differentiate the true acid strengths of the various mineral acids.

APROTIC solvents, i.e. benzene and carbon tetrachloride, are those which neither give up nor coordinate with protons to any measurable extent. These solvents are characterised by the lack of interaction between the solute and solvent, hence all reactions taking place are dependent entirely upon the intrinsic strengths of the respective molecules involved. It is only in a solvent of this type that intrinsic strengths of acids and bases are truly brought forth, as reactions taking place are between solute molecules which are free of solvation effects.

PROTOGENIC solvents are those which readily donate protons to any species possessing an unshared electron pair, thereby resulting in the formation of the corresponding onium ion. As a result of the solvolysis reaction, the basic strength of weak bases is enhanced. This increase in the relative basicity results from an enrichment of the solvent anion concentration, which is the strongest base that can exist in any given solvent. The solvolysis which enhances basic strength may be written as in reaction (10), where the acid HA is the solvent.

$$R_3N: + HA \longrightarrow R_3NH^+ + A^- \qquad (10)$$

The most commonly used protogenic solvent to enhance the basicity of weak amines is acetic acid. Although a number of solvents are superior to acetic acid as a medium for nonaqueous titrations, they do not possess the many inherent advantages present in this solvent (21). Due to the relatively small autoprotolytic constant of acetic acid, the size of the potentiometric "break" is of sufficient magnitude to warrant its use for acid-base titrations. It recently was shown by Pifer and Wollish (22) that the addition of p-dioxane to acetic acid increases the magnitude of the "break" at the potentiometric end-point due to a further decrease in the autoprotolytic constant, thereby increasing the possibilities of microanalytical procedures. On exposure of anhydrous acetic acid to the atmosphere for short time intervals, only traces of moisture are absorbed by the acid, thereby making laboratory handling quite convenient. Furthermore, because of its organic nature, a number of substances which are insoluble in a variety of other solvents are easily dissolved.

Like any other solvent, acetic acid possesses several disadvantages which tend to influence normal acid-base relationships. On dissolving an amine of moderate strength in acetic acid, solvation takes place resulting in the formation of an ionic salt composed of an alkylammonium cation and acetate anion. The low dielectric constant of acetic acid (approximately 6.3 at room temperature) minimizes the extent of dissociation of these oppositely charged ions. These tend to remain in the proximity of each other as neutral ionic combinations being held together by the coulombic forces of attraction. There is some tendency for these ionic combinations to dissociate into their respective ions, and the degree of this "ion-pair" dissociation is responsible for the development of relative basicity in acetic acid. Throughout the discussion which follows, additional information pertaining to the influence of these "ion-pairs" on the properties of the system will be discussed in detail.

Unfortunately at the present time, no acetate reversible electrode has been developed, consequently all measurements in acetic acid are relative rather than absolute, and the determination of exact dissociation constants is not possible. The temperature coefficient of expansion for acetic acid is, furthermore, quite large, thereby producing deviations in concentration with temperature changes.

In addition to its protogenic properties, acetic acid possesses some slight protophilic properties which aids in the differentiation of intrinsic acid strengths. The reaction between the acid and solvent results in the formation of the onium ion, the extent of interaction between the two being dependent upon the proton-donating tendency of the acid (61).

THE THEORETICAL CONSIDERATIONS OF THE ACETIC ACID SYSTEM

POTENTIOMETRIC STUDIES

As previously pointed out, Hall and Conant (12,13) are mainly responsible for the fundamental relationships upon which our present day knowledge of acid-base equilibria in acetic acid are based. Employing a saturated chloranil electrode in conjunction with the standard calomel electrode, they were able to measure apparent differences in hydrogen-ion activity of various acids and bases in acetic acid. With this electrode system, they arbitrarily defined the zero value of their scale as 0.566 volts, with the hope that this value would be shown to be near the zero point of the ordinary pH scale in water. Employing this "zero value", they defined the acidity of an acetic acid solution as $pH^{(HAc)}$, where,

$$pH^{(HAc)} = (0.566 - E) / 0.0591 \qquad (11)$$

E being the observed electromotive force of the cell in question.

Following the development of a satisfactory electrode system, Hall and Werner (21) qualitatively compared the acid strengths of perchloric, sulfuric, and hydrochloric acids and were able to show that perchloric acid is the strongest of the mineral acids when present in a "non-leveling" solvent. They proceeded to employ these acids in the titration of bases of varying strengths and showed the effects of (a) acid strength, (b) the salt formed, (c) the concentration of base present, and (d) the strength of the base on the shape of the titration curve. In addition, they also made a comprehensive comparison with analogous situations in aqueous solutions.

If a series of aqueous solutions of a strong base, differing from each other by a factor of ten-fold, are titrated with a strong acid, the titration curves lie very close to one pH unit apart, are rather similar in shape, and parallel to each other. However, in acetic acid, though the curves for a "strong" base are generally quite similar to those found in water, the effect of a ten-fold dilution is to space the curves ½ pH unit rather than a full unit apart.

As a test for "weak" and "strong" base behavior in various solvents, the curve obtained by plotting the electromotive force against the logarithm X/(1-X), X being the fraction neutralized, follows certain electro-chemical conventions. For weak bases, the curve follows the mass law without correction having a slope of 59 millivolts per unit change in the logarithm of the ratio, whereas strong bases show a marked departure from this slope. Of the several bases investigated in this manner by Hall and Werner (21), the slopes showed a curvature which steadily increased with the strength of the base, and in all cases the slope was less than the theoretical value. It was noted that the curves of the supposedly stronger bases approached the shape of those shown by strong bases in water, hence these workers resorted to a more sensitive test for strong base behavior.

A plot of the logarithm of the unneutralized fraction (1-X) of base against the electromotive force gives, for a strong base in water, a straight line with a slope of 59 millivolts per ten-fold change in concentration. This test when applied to the same bases as above, gave a straight line with a slope of 59 millivolts for the stronger bases, whereas, as the basic strength diminished, the curvature and departure from theoretical slope became more pronounced.

As a final test of dissociation behavior, the potentiometric dilution curves of diethylaniline, pyridine, sodium acetate, o-chloroaniline, and guanidine were experimentally determined and each of the bases exhibited a slope in the vicinity of 0.5, corresponding to a change of 59.1/2 millivolts per ten-fold variation in concentration. As a completely dissociated electrolyte has a slope of 59.1 millivolts for the same concentration change, it is quite obvious that in acetic acid all substances follow the Ostwald law of concentration effect (square root dissociation relationship).

An investigation of 71 different bases in acetic acid led Hall (23) to draw some pertinent conclusions as to the limiting values of basic strength in this solvent. By potentiometric titration with perchloric acid under standardized conditions it was possible to arrange all 71 bases in the order of their strengths by computing the $pH^{(HAc)}$ at which the logarithm of the ratio of the base to salt term vanishes, hence $pH^{(HAc)} = pK'$, the latter being the dissociation constant for the protonated base. A plot of $K_{basicity}^{(HAc)}$* against $K_{basicity}(H_2O)$** gives a curve as shown in Figure 1.

As the basic strength becomes progressively greater in water, it likewise increases in acetic acid up to a certain value beyond which only minor variations in basic strength occur, even though the strength in aqueous solutions is continually increasing. The point at which bases tend to become "strong" in acetic acid occurs at $pK_B = 4.75$, which closely approximates the strength of acetate ion as a base in water.

On the basis of the titration curves in acetic acid (23) and Figure

---

* $K_{basicity}^{(HAc)} = pK' = pH^{(HAc)} + \log R$ at $R = 1$, i.e. at the midpoint of the titration curve where the logarithm of the base to salt ratio (R) vanishes.

** $K_{basicity}^{(H_2O)} = pK_B = \log K_b + pK_w$

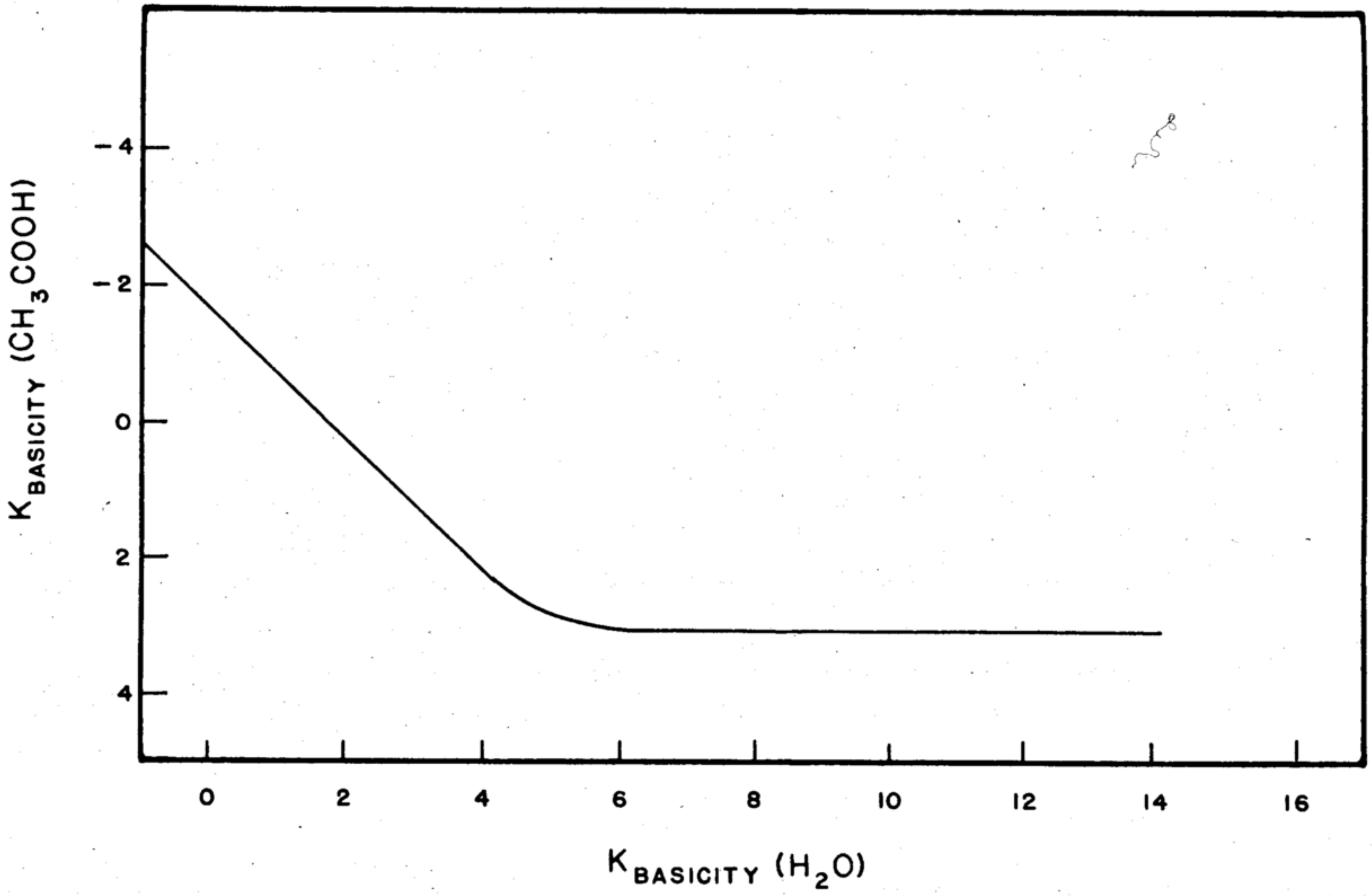


FIG. 1. COMPARATIVE STRENGTH OF BASES IN ACETIC ACID AND WATER (23).

1, the various bases were classified into one of three categories: weak bases, intermediate strength bases, and strong bases. The titration curves of the weaker bases rise rather rapidly at first and continue to rise with increasing acid concentration, though not as sharply. This continual rise may be attributed to the fact that some of these bases are such weak entities that their tendency to react with perchloric acid is diminished to a point where the solution behaves as if just the solvent alone were present.

The curves of the intermediate strength bases are somewhat analogous to those of weak bases in water, however these tend to be somewhat flattened in character. This anomalous behavior has been shown to be caused by changes in activity coefficients during the course of a titration (24) as a result of variations in the "ionic strength."*

The strong base curves are analogous to those for strong bases in water which may be rationalized on the fact that strong bases readily accept a proton in acetic acid, thereby being present almost entirely in the ionized form. The addition of perchloric acid to the system merely serves to substitute perchlorate ion for acetate ion, hence the "ionic strength" remains virtually constant and the proton activity is dependent upon the removal of acetate ion from the system.

CONDUCTOMETRIC STUDIES

Although the work of Hall, Conant, and Werner (12,13,20,21,23,24) pioneered the field of acidity and basicity in acetic acid, these workers

---

* In the light of present thought, the use of the term "ionic strength" is a misnomer since the actual number of free ions present as such is extremely small.

concentrated their efforts primarily on the measurement (potentiometrically) of proton activity in various solutions, and in this manner were able to draw qualitative conclusions as to relative intrinsic strengths.

In order to obtain quantitative data pertaining to the strength of acids and bases, Kolthoff and Willman (25,26) employed conductance measurements in anhydrous acetic acid. On the basis of the conductivity of the respective solutions, the relative intrinsic strengths of several acids and bases were approximated with some degree of accuracy.

As previously pointed out, the ability of an H-acid to donate a proton to a protogenic solvent possessing some protophilic properties is a measure of the intrinsic strength of the acid.

$$HX + CH_3COOH \rightleftharpoons CH_3COOH_2^+ + X^- \qquad (12)$$

If the acid is inherently weak, reaction (12) lies mainly to the left and HX exists predominantly as such in solution. However, if HX is a strong acid, there is a greater tendency for the solvation reaction to proceed to completion and reaction (12) lies predominantly to the right.

It must be pointed out that the dielectric constant of acetic acid is quite low (6.3), hence the ions formed have a tendency to remain as neutral "ion-pairs" or combine as higher neutral ionic aggregations. This "ion-pair" formation was first postulated by Bjerrum (60) and later described by Hantzsch and Voigt (27) in their work dealing with the determination of the true strengths of mineral acids in water-free chloroform. The presence of "ion-pairs" as the normal character of ionized solutes in acetic acid has recently been substantiated by the work of Griswold, Jones, and Birdwhistell (28) who showed that the solubility of potassium bromide in acetic acid is enhanced by the addition of solutes containing a common ion to the system, whereas the addition of solutes lacking a common ion produces very little effect. This they attributed to an exchange reaction between the "ion-pairs" and were able to compute equilibrium constants which remained fairly constant over a wide range of concentrations.

In addition to "ion-pair" formation, the weaker acids are capable of associating themselves with the solvent giving rise to addition compounds which exist as such in solution (25).

A differentiation between "ion-pairs" and neutral combinations is quite important in understanding the changes occurring in the electrical conductivity and basicity of acetic acid solutions into which minute amounts of water are introduced. In the absence of water, the conductance of various acids in acetic acid was shown by Kolthoff and Willman (25) to decrease in the following order:

$$HClO_4 > HBr > H_2SO_4 > HCl > HNO_3$$

At a concentration equivalent to 0.005 molar, the ratio of the conductivities of the above acids are 400:160:30:9:1, respectively, these results being in direct agreement with those reported by Hantzsch and Voigt (27) on the intrinsic strengths of the various acids.

Water, because of its greater protophilic properties compared to acetic acid, interacts with the weaker acids and increases the number of "ion-pairs" present in acetic acid (25). On the other hand, strong acids which have undergone dissociation to some extent in this solvent merely solvate preferentially with the water molecules by means of a transference from the acetic acid to water molecules with the result being very little change (percentage-wise) in the number of "ion-pairs" present. Therefore, the net result is that in the case of a weak acid, because of enhanced dissociation, the conductance will be markedly increased whereas in the case of the strong acid, the effect on the conductance will be very small.

On the addition of water to acetic acid solutions containing various acids, not only is the conductivity of the system altered, but the acidity tends to decrease, the extent of decrease being dependent upon the nature of the acid present (29). As pointed out by Smith and Elliott (29), the changes occurring in acidity are dependent upon the relative basicities of three species: the anion of the acid, the water molecules, and the acetic acid molecules. As the conjugate base (anion) of a weak acid, i.e. hydrochloric acid, is stronger than the conjugate base of a strong acid, i.e. perchloric acid, it is evident that the former will not have as high an affinity as the latter for the basic water molecules. Therefore, the reaction of a weaker acid with water will not proceed to the extent of the stronger acid, and the acidity of solutions containing stronger acids will be decreased to a greater extent than those of the weaker acids. This has been shown to be the case by Smith and Elliott (29) who went further and showed that the basicity constant of water in acetic acid is dependent upon the strength of the acid present, and not upon the water molecules themselves.

The conductance data available from the work of Kolthoff and Willman (25) has been utilized (29) to compute dissociation constant values for several "ion-pair" combinations in acetic acid. By employing a Fuoss and

$$\underset{(\text{"ion-pair"})}{M^+A^-} \rightleftharpoons \underset{(\text{free ions})}{M^+ + A^-} \tag{13}$$

$$K_{\text{dissociation}} = (M^+)(A^-)/(M^+A^-) \tag{14}$$

Kraus (30,31) type plot, it was possible to compute values of the equivalent conductance at infinite dilution and classical dissociation constant for perchloric acid. Using the assumption that the limiting equivalent conductances for the other acids and lithium chloride are nearly the same as perchloric acid, dissociation constants based on that of perchloric acid were calculated. These values* as given by Smith and Elliott (29) are as follows:

Acid or Salt	K_a
perchloric acid	9.0×10^{-7}
hydrobromic acid	1.9×10^{-7}
sulfuric acid	7.4×10^{-9}
hydrochloric acid	5.1×10^{-10}
lithium chloride	7.4×10^{-8}

Quantitative relationships among the various inorganic acetates which behave as bases in acetic acid were also determined by conductometric means. The conductance of equimolar solutions of various acetates in acetic acid as reported by Kolthoff and Willman (26) shows the increasing order of acidity for cations to be: Rb, K, NH_4 (NH_4 = K), Na, Li, Ag, Ba, Sr, Ca, and Mg. A similar study carried out in acetic anhydride by Usanovich and Yatsimirskiĭ (32,33) found the order to be: K, Rb, Ba, Sr, NH_4, Pb, Li, Mn, Ca, Y, Cd, Cu, Mg, Ni, Zn, Al, and Be. Kolthoff and Willman (26) further verified their work by an indicator procedure, measuring the specific conductance of a potassium acetate solution possessing the same color intensity as a fixed concentration of cation acetate, i.e. the concentration of potassium acetate was varied until it produced the same color as that produced by a fixed amount of

---

* It is the opinion of the present author that these dissociation constants should be taken only as a crude approximation, as the assumptions made are not wholly valid.

cation acetate.

In an attempt to correlate some of their data with that of Hall (23), Kolthoff and William investigated the conductance behavior of several organic bases in a similar manner. On the basis of their results, they assumed that the extent of dissociation of the solvation product varies directly with the size of the organic cation. This is in agreement with the dissociation behavior of the inorganic cation acetates, the larger the size of the ion, the greater being the dissociation.

For a given series of salts in acetic acid possessing the same cation, the acid reaction of the solutions tends to decrease in the order: $ClO_4^- > I^- > Br^- > Cl^- > NO_3^-$. Where a common anion is present, the basicity decreases accordingly: $Rb^+ > K^+ \cong NH_4^+ > Na^+ > Li^+$ (26).

COLORIMETRIC STUDIES

The work of Hall and Conant (19,13) provided a means of measuring the relative strengths of acids and bases in acetic acid. In the choice of a zero point for their arbitrary scale (see equation 11), these workers selected 0.566 volts for the following reasons.

> "It seems probable that the strength of anhydro bases should be approximately the same in different solvents and this is the case for urea, dimethylpyrone and acetamide in acetic acid and water as we have defined the pH(HAc) scale. The solvent (acetic acid) half neutralised with a relatively strong base such as pyridine might be expected to have a hydrogen-ion activity of the same order of magnitude as the pK_a of acetic acid dissolved in water. The pH(HAc) of an equimolar mixture of pyridine and pyridine acetate is about 4.4 as compared to the value 4.7 given for the pK_a of acetic acid in water."

In an attempt to correlate acidity measurements in strongly acid solutions with those obtained in dilute aqueous solutions so that the two scales would overlap in regions of moderate acidity, Hammett and Deyrup (34) defined "acidity in terms of a basic indicator," which has since become known as the Hammett Acidity Function, H_0. In measuring the acidity of a solution by means of a simple basic indicator[*], the extent to which any acid present is capable of transferring a proton to a neutral organic base is a measure of the total acid concentration present and differs considerably from the theoretical concept of hydrogen-ion activity.

The acidity function has been defined for use in solvents of high dielectric constant where the protonated form of the indicator base is independent of the anion present; however, as will be pointed out later, there is a useful correlation between H_0 and potentiometric measurements in acetic acid (34,36). The indicators employed are of such type that the addition of a proton results in a change in the visible color intensity due to changes in the ionization ratio, BH^+/B, where B represents the indicator base.

The theoretical derivation of the Hammett Acidity Function is based on the fact that any neutral molecule which has accepted a proton tends to undergo dissociation to some extent according to the following scheme:

$$BH^+ \rightleftharpoons B + H^+ \qquad (15)$$

and

$$pK' = -\log (B)(H^+)/(BH^+) - \log (f_B f_{H^+}/f_{BH^+}) \qquad (16)$$

In dilute aqueous solutions, if pK_A' is known, then the pK' of any other indicator, which overlaps a portion of the acidity range covered by indicator A, may be evaluated in a stepwise procedure. This follows from equation (17) since the indicators are being compared in solutions of

---

[*] A simple basic indicator is defined as being capable of adding only one proton to a neutral molecule, with the resulting protonated form being independent of the conjugate base of the acid.

equal acidity.

$$pK'_B - pK'_A = \log \frac{(B)(AH^+)}{(BH^+)(A)} - \log \frac{f_B f_{AH^+}}{f_{BH^+} f_A} \qquad (17)$$

The fundamental assumption made in the development of the acidity function scale is that the ratio of the activity coefficients, $f_B/_{BH^+}$, is the same for all bases in any given solution, hence the ratio of the activity coefficients in equation (17) vanishes and the following expression remains:

$$pK'_B - pK'_A = -\log (B)(AH^+)/(BH^+)(A) \qquad (18)$$

This assumption has been shown to be valid since the acidity scale is standardized in such a way that it reduces to ordinary pH in dilute aqueous solutions.

Hammett and Deyrup (34) defined the acidity function in the following manner:

$$H_0 = pK' - \log (BH^+)/(B) \qquad (19)$$

By substituting the definition of pK' into equation (19), H_0 may be expressed in its equivalent form showing it to be independent of the nature of the base employed to measure it, provided that the protonated form of the indicator base is independent of the conjugate base of the acid present.

$$H_0 = -\log (a_{H^+})(f_B)/(f_{BH^+}) \qquad (20)$$

By using a series of basic indicators covering a continuous acidity range, and a stepwise application of equation (18), the base strength of any acidic solution relative to water may be obtained. As previously pointed out, it is essential, that the protonated form of the indicator be completely free of the anion in order to obtain a true measure of the ratio BH^+/B, since in the derivation it is assumed that BH^+ is independent of the anion form. In this manner, Hammett and Deyrup (34) were able to compute H_o values for sulfuric acid solutions ranging from 0 to 100 percent in acid concentration. As these results were outside the range of "dilute aqueous solutions," it was not possible to establish the zero point of the H_o scale. This was carried out by Hammett and Paul (35) employing stronger indicator bases in solutions of weaker acidity, and in this manner were able to correct the previous data of Hammett and Deyrup (34).

Hall and Spengeman (36) used the visual colorimetric method developed by Hammett (34,35) to obtain H_o values for acetic acid solutions containing from 0.02 to 50 percent sulfuric acid with the intent of re-interpreting some of the earlier potentiometric data (23,24). Using an indicator whose pK' was obtained by Hammett and Paul (35), they employed five indicators whose pK' values they determined in a stepwise graphical procedure. In order to provide some correlation between the original $pH^{(HAc)}$ scale and H_o, they showed that in dilute solutions of sulfuric acid in acetic acid, the relationship "$H_o = pH^{(HAc)} + 5$", is approximately correct. This was originally suggested by Hammett and Deyrup (34) saying that "In dilute aqueous solution, H_o becomes equal to pH; in acetic acid it is theoretically equal to $pH^{(HAc)}$ of Hall and Conant, but differs by an additive constant from the value actually given by their scale."

Hall and Werner (37) further extended the values of the H_o range in acetic acid into the more positive regions by employing buffer systems of constant solute concentration. Again, indicator constants were determined in the usual stepwise procedure. By the use of sulfuric acid -

urea.hydrogen sulfate buffers, they covered the range -1.6 to -0.38; with urea.hydrogen sulfate - urea, the range -0.17 to 1.93; and with antipyrine.hydrogen sulfate - antipyrine, the range 0.67 to 3.82.

Harbottle (38) applied the acidity function to mixtures of 6 Formal perchloric acid - sodium perchlorate systems using indicators previously evaluated by Hammett. A series of organic acids were prepared by Smith and Elliott (29) who measured the H_o values of their solutions in acetic acid containing 0.12 percent water. On the basis of these measurements they showed that perchloric acid is the strongest of the mineral acids, though incompletely ionized in acetic acid, and that the di- and trisubstituted methane sulfonic acids are somewhat stronger.

Lemaire and Lucas (40) further added to the number of indicators available for acidity measurements in acetic acid and employing these were able to measure the basic strength of several weak bases. They showed that the relationship between the thermodynamic constant, pK_a, of a base and its ionization constant, K_a^s, in acetic acid containing perchloric acid in concentrations up to 0.2 molar is accurately expressed:

$$pK_a - pK_a^s = H_o + \log (H^+) = -3.88 \qquad (21)$$

Employing this expression they measured the basic strength of such colorless weak bases as acetonitrile, dioxane, acetanilide, acetamide, urea, and others.

A list of several indicators in the order of increasing basicity, along with their constants in acetic acid, as determined by various workers is given in Table I. The majority of these indicators are of the aniline type which conform to the Hammett definition, however the color changes which they exhibit are gradual rather than sharp.

As another attempt in the development of an acidity scale as a means

Table I. Indicator Constants in Acetic Acid.

Indicator	pK'	Reference
2,4,6-trinitro-N,N-dimethylaniline	- 4.73	36
2,6-dinitro-4-methylaniline	- 4.45	36
2,4-dichloro-6-nitroaniline	- 3.31	39
p-nitroazobenzene	- 3.29	39
p-nitrodiphenylamine	- 2.51	39
2-nitro-4-chloro-N-methylaniline	- 1.40	40
p-chloro-o-nitoraniline	- 0.94	39
o-nitroaniline	- 0.17	39
2,6-dichloro-4-nitro-N,N-dimethylaniline	- 0.10	40
2,4-dinitro-N,N-diethylaniline	0.30	40
alpha-naphtholbenzein	0.53	29
p-nitro-N,N-dimethylaniline	1.08	37
p-nitroaniline	1.28	37
p-nitro-N,N-diethylaniline	2.10	37
m-nitro-N,N-dimethylaniline	3.14	37
m-nitro-N,N-diethylaniline	3.74	37

of measuring hydrogen-ion activity in extremely acidic solutions, Michaelis and Granick (41) utilized a bivalent, reversible indicator system which was capable of distinguishing three levels of oxidation - reduction. In this system the potentials of the reduced - semi-oxidized (E_1) and semi-oxidized - totally-oxidized (E_2) states were dependent upon the hydrogen-ion concentration and were interrelated by the mean potential (E_m), which represents the normal potential of the reduced - totally-oxidized system.

$$E_m = (E_1 + E_2)/2 \qquad (22)$$

The difference ($E_2 - E_1$) was dependent upon the hydrogen-ion concentration of the system and the spread between these two potentials was employed as a measure of the hydrogen-ion activity at pH values less than zero. The systems were so chosen that in the ordinary acidity ranges, they coincided with pH. The values of E_1 and E_2 were determined by means of potentiometric titration experiments.

The indicators chosen were employed in solutions whose acidity range up to concentrations where there was a change in the slope of the potential curves, E_1 and E_2, resulting from changes in the state of acidic ionization of the indicator forms due to proton attachment. Therefore, indicators which overlapped their ranges were employed to extend into the more acidic regions.

The values obtained by this method were in close agreement with the values obtained with the Hammett Acidity Function (34). A distinct advantage of this method lies in its use of uncharged, singly charged, and doubly charged bases, whereas the Hammett H_o system is derived for uncharged and singly charged indicator bases.

SCOPE OF INVESTIGATION

A considerable amount of work has been done in the relatively acidic regions of the acetic acid solvent system. These studies have been concerned with the development of stronger acids and the use of indicators which show sharp color transitions in the acidic regions. Unfortunately, the relatively alkaline regions of this system have been, for the most part, neglected as a solvent for analytical purposes.

There are a number of methods available for acid-base investigations, however, the present work has been limited to potentiometric and colorimetric studies of the relatively alkaline regions of acetic acid. In carrying out this work, emphasis was placed on the development of stronger bases than those commonly used at present for titration purposes. It was initially desired to locate an effectively totally dissociated base in acetic acid, however it soon became apparent that such an entity would be highly improbable. As a result, attention was then directed towards studying the effects of stereochemical configurations on dissociation behavior in acetic acid, with the aim of locating the strongest base possible.

The measurement of hydrogen-ion activity of a number of bases of varying structural types in acetic acid was carried out by means of a potentiometric dilution procedure. In the choice of organic amines for potentiometric study, particular attention was paid to steric configurations about the nitrogen atom since previous workers (26) have shown that "ion-pair" formation can be reduced by increasing the interionic distance. In comparing relative dissociations of the various bases, potentiometric acidities were equilibrated, and the ratio of the dissociation constants of the bases determined accordingly. Since no absolute values are possible in acetic acid, all measurements made were relative to that of an arbitrary standard.

The constants of several indicators which exhibit their color change in the relatively alkaline regions of acetic acid were evaluated by spectrophotometric procedures. By determining the acidity at which the ratio of the protonated to unprotonated indicator forms was one, it was possible to determine the indicator constant (pK') in terms of the Hammett Acidity Function (H_0). With the aid of these alkaline indicators, it was possible to extend the Hammett Acidity Function scale to the more alkaline regions of the solvent.

In order to survey a number of indicators for their acid-base properties in acetic acid, potentiometric acid-base titrations were performed in the presence of a chosen indicator. The suitability of each indicator was obtained by a comparison of the colorimetric endpoint with that obtained from a graph of the potentiometric titration curve.

EXPERIMENTAL TECHNIQUE

The apparatus and chemicals employed for the various analytical procedures performed in this research study are described below.

PART I: Relative Basicity by Potentiometric Measurements.

APPARATUS. Beckman pH meter, Model G.

Beckman Glass Electrode, No. 1190-80. In order to obtain reproducible results it was found desirable to immerse the electrode in 0.1 normal hydrochloric acid solution for a short time interval after use, and then allow it to soak overnight in distilled water before subsequent usage. By means of this procedure reproducible results were consistently attained.

Beckman Calomel Electrode, No. 4870-71 (Sleeve type). To insure proper response the electrode sleeve was removed and the saturated potassium chloride solution allowed to partially drain out and then replaced with fresh solution. At various intervals, the electrode was completely drained, flushed with distilled water, and then refilled with fresh potassium chloride solution and several potassium chloride crystals to insure proper saturation.

REAGENTS. Glacial acetic acid, c.p. reagent, was refluxed with triacetyl borate according to the procedure of Eichelberger and La Mer (43), and then distilled from an all-glass apparatus. This purified acid was employed in the preparation of all solutions and in potentiometric measurements. During the latter stages of this work, the acetic acid was refluxed with boron acetate obtained from the American Potash and Chemical Corporation, and then distilled in a similar manner.

Sodium salicylate, U. S. P. grade, 0.100 molar in acetic acid.

Potassium biphthalate primary standard (Merck).

Quinaldine red (Eastman Kodak #1361) indicator solution, approximately 0.1 per cent in acetic acid.

Perchloric acid, approximately 0.05 molar, in acetic acid was prepared according to the procedure of Fritz (44). Standardization of the acid was accomplished against potassium biphthalate primary standard with quinaldine red as the indicator.

Stock solutions of the various inorganic acetates, amines, and substituted ammonium acetates, approximately 0.05 molar, were prepared and standardized against perchloric acid with quinaldine red as indicator; where necessary, the weaker inorganic acetate solutions were standardized by potentiometric titrations. The bases investigated included: barium acetate[1], mercuric acetate, lithium acetate[1], magnesium acetate, potassium acetate, sodium acetate[1], strontium acetate[1], ammonium acetate, n-butylamine[2], di-isobutylamine[3], di-n-butylamine[2], tribenzylamine[1,2], tri-n-butylamine[3], tricyclohexylamine (45), triethylamine[2], tri-n-heptylamine[2], tri-n-hexylamine[2], trimethylamine[2], tri-n-octylamine[3], triphenylamine[2], tri-n-propylamine[3], and tetrabutylammonium acetate[1]. These solutions were stored in the reservoirs of automatic burets. The inorganic acetates were of analytical reagent quality dried at suitable temperatures. The nitrogenous compounds were titrated for their molecular weights in acetic acid using perchloric acid titrant and quinaldine red indicator. The results indicated that all the amines were better than 99 per cent.pure. After several recrystallizations the tetrabutylammonium acetate was found to have a molecular weight of 357.0 compared with the

---

[1] These bases were investigated at concentrations greater than 0.05 molar and stock solutions of higher concentration were employed.

[2] These amines were Eastman Kodak quality.

[3] Obtained from Sharples Chemicals Inc., Philadelphia, Pa.

calculated weight of 301.5, corresponding to a purity of 84.50 per cent.

PROCEDURE. A series of dilutions of the various bases in acetic acid were prepared by dilution of the stock solutions to 25.0 milliliters in volumetric flasks. In this manner a series of solutions ranging from 0.0001 to 0.05 molar were obtained for potentiometric investigation. For those substances investigated at concentrations greater than 0.05 molar, dilutions of the more concentrated stock solutions were employed.

Electromotive force measurements in acetic acid are not indicative of true equilibrium voltages, but relative values, hence it was necessary to choose and arbitrary standard with which to compare all potentiometric readings. Sodium salicylate, being the salt of an acid stronger than acetic acid, was chosen as the reference standard with which to compare all EMF measurements. A 0.100 molar acetic acid solution of sodium salicylate used as the reference gave a potential of 376 millivolts, being consistant and reproducible within the sensitivity range of the Model G pH meter. This potential was subtracted from all other measurements, hence all readings are expressed in terms of millivolts relative to the reference solution.

The use of this standard reference solution provided a daily check on the reproducibility of the electrode system. Failure of the system to give the proper reference reading with sodium salicylate solution indicated that the electrodes required some conditioning before actual measurements were begun. Under these circumstances the electrodes were subjected to the treatment discussed above until the proper reference value was reproducible.

All potentiometric measurements were carried out by transferring the proper dilution to a 50-ml. beaker, immersing the electrodes in the solution and recording the EMF readings. It was necessary that both the top and bottom of the calomel sleeve were immersed in the solution to insure reproducibility. Exposure of the electrodes to acetic acid solutions for periods longer than a minute or two resulted in deviations, presumably from a dehydration of the calomel sleeve joint by the acetic acid. Readings were therefore taken as soon as possible and frequently checked by the use of freshly prepared solutions.

After each measurement, the electrodes were rinsed with ethanol, then with water, and finally allowed to soak two minutes in water prior to the next determination. Thorough drying was insured by the use of cheese cloth and lens paper, making certain all the water was removed from the groove of the sleeve electrode.

Results were obtained by plotting the logarithm (molarity x 10^4) against relative millivolt readings and data analyzed on the basis of this chart as shown in Figure 2. The slopes were computed over the region 0.0001 to 0.01 molar where the curves exhibit a marked degree of parallelism.

All experimental measurements are believed accurate to the extent of dilution errors and the sensitivity of the Model G pH meter.

PART II: Determination of Indicator Constants (pK') in Terms of the Hammett Acidity Function, H_o (34).

APPARATUS. Cary Recording Spectrophotometer, Model 11MS.

Quartz absorption cells, 0.1, 1.0, and 10.0 cm.

REAGENTS. Acetic acid, prepared as in Part I.

Antipyrine, N. F. grade, 0.1111 molar in acetic acid.

Pure sulfuric acid, prepared by the "fair and foggy" method of Kunzler (46).

Antipyrine.dihydrogen sulfate, 0.1111 molar in acetic acid, prepared by mixing equal molar quantities of antipyrine and pure sulfuric acid.

Perchloric acid, approximately 0.05 molar, prepared and standardized according to the procedure of Fritz (44).

Tricyclohexylamine, 0.1111 molar in acetic acid.

Sodium acetate, A. R., 0.1111 molar in acetic acid.

Potassium acetate, A. R., 0.1111 molar in acetic acid.

Strontium acetate, A. R., 0.1111 molar in acetic acid.

Acetic acid solutions of the following indicators in concentrations which provided suitable optical density measurements. The indicators were recrystallized from the solvents indicated, dissolved in acetic acid, and filtered before use to insure freedom from undissolved indicator particles.

Quinaldine red (Eastman Kodak #1361) - recrystallized from ethanol.

p-Phenylaminoazobenzene (Eastman Kodak #1714) - recrystallized from an ethanol-acetone mixture.

p-Dimethylaminoazobenzene (Eastman Kodak #338) - recrystallized from Skelly B.

Thymol blue (Eastman Kodak #753) - recrystallized from ethanol.

Benzopurpurin 4B (Eastman Kodak #1071) - employed in a saturated solution in acetic acid.

Orange IV (Eastman Kodak #P 823) - recrystallized from water.

α-Naphtholbenzein (Eastman Kodak #924) - recrystallized from ethanol.

PROCEDURE. Buffer solutions of constant solute concentration and known H_0 values were prepared by mixing the proper volumes of antipyrine and antipyrine.dihydrogen sulfate solutions as described by Hall and Meyer (37). The resulting solutions were 0.1000 molar in total salt concentration after addition of the indicator solution. The final composition of these solutions along with their acidity function values are as follows:

Buffer Code No.	Antipyrine Molarity	Antipyrine.H_2SO_4 Molarity	H_0 Value
A12	–	0.1000	0.67
A14	0.0111	0.0889	2.14
A16	0.0222	0.0778	2.62
A18	0.0444	0.0556	2.96
A20	0.0667	0.0333	3.31
A21	0.0778	0.0222	3.49
A22	0.0889	0.0111	3.82
A23	0.1000	–	–

To each solution a fixed volume of indicator solution was added and the resulting visible absorption spectrum was recorded over the region 325mμ to 700mμ. One-cm. quartz absorption cells were employed with the Cary spectrophotometer.

In order to insure complete conversion of the indicators to their acidic and basic forms, the absorption spectra of A12 and A23 buffers were checked by the use of perchloric acid and tricyclohexylamine solutions, taking into consideration any variations produced by "medium" (47) and/or counterion effects.

The determination of the ionization ratio, BH^+/B, was accomplished by the use of the spectrophotometric data and equation (23), which is

$$H_o = pK' - \log\left[\frac{E_B - E}{E - E_{BH^+}}\right] \qquad (23)$$

similar to that employed in aqueous solutions (47-49). E_B and E_{BH^+} represent the molar extinction coefficients of the base and salt forms, respectively, while E is the molar extinction coefficient at any intermediate acidity where B and BH^+ are present as an equilibrium mixture. The activity coefficient terms for the base and salt forms have been taken as unity since the dye concentrations are present in extremely small amounts. Since the molar dye concentration and cell length are constant for any given set of measurements, optical densities have replaced extinction coefficients in equation (23), hence,

$$H_o = pK' - \log\left[\frac{R' - R}{R - R_o}\right] \qquad (24)$$

where R', R_o, and R are the optical density values corresponding to E_B, E_{BH^+}, and E. Utilizing this data, the values of pK' were computed graphically by plotting H_o against the logarithm of the ionization ratio and locating the value at which the logarithm of BH^+/B for each indicator is zero, and therefore pK' equals H_o.

Quinaldine red was subjected to a thorough spectrophotometric investigation to show the effect of changes in the solution composition on the wave length of the absorption maximum in the alkaline form. Among the factors considered were "ionic strength," indicator concentration, anions, and cations; all of which will be discussed in detail under the section dealing with this indicator. The extent to which quinaldine red adheres to Beer's law was determined by employing 0.1, 1.0, and 10.0 cm. absorption cells, the indicator concentration being reduced by a factor of ten-fold in each instance.

PART III: The Investigation of Indicators for Titration Purposes.

APPARATUS. Beckman pH meter, Model G.

Beckman Glass Electrode, No. 1190-80.

Beckman Calomel Electrode, No. 4970-71 (Sleeve type).

Magnetic stirrer with teflon coated stirring bar.

REAGENTS. Acetic acid, prepared as in Part I.

Perchloric acid, approximately 0.05 molar, prepared and standardized according to the procedure of Fritz (44).

Pure sulfuric acid, prepared by the "fair and foggy" method of Kunzler (46).

Tricyclohexylamine, 0.1111 molar in acetic acid.

Sodium acetate, A. R., 0.1111 molar in acetic acid.

Solutions of the following indicators, in acetic acid, of suitable concentrations so as to render color changes readily visible: quinaldine red, metanil yellow, malachite green oxalate, benzopurpurin 4B, orange IV, propyl red, thymol blue, α-naphtholbenzein, p-phenylaminoazobenzene, p-dimethylaminoazobenzene, meta-cresol purple, formyl violet S4B, brilliant yellow, and naphtol green B. The latter two indicators were used in freshly prepared and filtered solutions.

PROCEDURE. Solutions of perchloric acid in acetic acid were titrated potentiometrically with tricyclohexylamine in the presence of various indicators. Both potentiometric and colorimetric data were recorded at suitable intervals and graphical comparisons between the two endpoints were made, these results being shown in Figures 22 to 33.

Weighed samples of pure sulfuric acid were titrated potentiometrically with (1) sodium acetate and (2) tricyclohexylamine. A comparison of the potentiometric "breaks" are shown in Figure 34, the curves having been shifted vertically so that the half-neutralization points coincide.

RESULTS AND DISCUSSION

PART I: Relative Basicity by Potentiometric Measurements.

According to Hall (23), all amines more protophilic than aniline in water essentially undergo complete conversion to the ammonium form in acetic acid. Because of the low dielectric constant of acetic acid, the resulting "ion-pairs" of the ammonium acetate are unable to undergo dissociation into charged particles, the salt thus remaining intact as neutral undissociated bodies. As basicity in acetic acid is due entirely to the concentration of free acetate ions present in the system, the extent to which these neutral ionic combinations dissociate determines the relative "alkalinity" of the solution.

The results of a limited study on amines carried out by Kolthoff and Willman (26) showed that the dissociation of the "ion-pairs" increased with their ionic diameter. Thus, in accordance with their work, it would be expected that the higher molecular weight tertiary amines would be among the strongest bases, and that basic strength would increase with the length of the alkyl chains. This may not necessarily be true, as will be shown later, since the extent of dissociation may be dependent upon more than just the ionic sizes.

As part of the present search for strongly basic reagents for the acetic acid system, it was felt desirable to conduct a systematic investigation of the relationship between structure and basicity of amines in this solvent. For this purpose, the potentiometric dilution technique previously described and shown to be suitable for the evaluation of relative acidity and basicity was employed. The technique used and some of the underlying theories have been discussed adequately under their respective sections.

The results of determinations made on a number of organic and inorganic bases in acetic acid are given in Tables II, III, and IV, and in Figure 2. Because of the limitations in space and overlapping, the dilution curves for only thirteen of the nineteen bases investigated are shown. It has not been possible to show the remaining dilution curves for tri-n-hexylamine, tri-n-heptylamine, trimethylamine, di-isobutylamine, n-butylamine, and ammonium acetate; these, however, follow a similar pattern and their slopes are recorded in Table V.

The curves shown in Figure 2 can be better understood in light of the following derivation. For any given acetate salt, MAc, there is a dissociative equilibrium present, the extent of which is dependent upon the individual base, and which mathematically is expressed in equation (26). At equal electromotive force potentials, the hydrogen-ion concentrations of any two solutions are identical since potentiometric measurements with the glass electrode are indicative of relative acidities in nonaqueous solvents. Therefore, the acetate ion concentrations of the solutions are also identical as the ion product of hydrogen and acetate ions is a constant similar to the ion product of hydrogen and hydroxyl ions in water. Since the acetate ion concentrations are identical at the same potential, it is possible to equate two different acetate salts against each other by means of equations (27) and (26). In this manner,

$$MAc \rightleftharpoons M^{+} + Ac^{-} \tag{25}$$

$$K = \frac{(M^{+})\ (Ac^{-})}{(MAc)} \tag{26}$$

$$(Ac^{-}) = \left[K(MAc)\right]^{\frac{1}{2}} \tag{27}$$

Table II. Potentiometric Dilution Data for Inorganic Acetates.

	Relative Millivolts*				
MOLARITY	KAc	NaAc	$BaAc_2$	LiAc	$SrAc_2$
0.0002	80^2	87^3	90^2	92	99
0.0004	72^2	79^5	82	83^2	-
0.0006	65^2	73^5	76	78^2	83
0.0008	61^2	72^4	73	75^2	-
0.0010	57^4	-	72^2	71^2	79^2
0.0020	47^4	58^3	62^2	64^2	-
0.0030	42^4	51^5	54	55^2	64
0.0050	36^4	45^4	50	49	57
0.0100	26^2	37^2	40	41^2	49
0.0200	17^2	27^2	31	31	40
0.0400	-	-	-	25^2	-
0.0500	4^2	15	14^2	20^2	23
0.1600	-	-	-16^2	-	-
0.2000	-	-16^2	-20^2	1	- 6
0.3900	-	-	-39^3	-	-
0.4000	-	-31^2	-	-11^2	-18
0.9600	-	-	-66^2	-	-
1.0000	-	-58^2	-34^2	-47	-

---

* Represents the average of the number of determinations as indicated by the individual superscripts. Readings are relative to that shown by a 0.1000 molar solution of sodium salicylate in acetic acid.

Table III. Potentiometric Dilution Data for Organic Amines.

	Relative Millivolts*				
MOLARITY	Tricyclohexyl	Tribenzyl	Triethyl	Tri-n-propyl	Tri-n-butyl
0.0001	61	-	73	75	74
0.0002	45	53	62	63	64
0.0004	36	44	52	53	54
0.0006	30^2	40	47	47^2	51^2
0.0010	24	32	42	42	43
0.0020	15^2	24	32	32^2	35^2
0.0030	10	19	25	26	30
0.0050	3^2	12	21	22	23
0.0100	- 4	4	12	13^2	15^2
0.0200	-10	- 5	2	3	7
0.0500	-15	-11	-13	- 9	- 6
0.1000	-	-19^2	-	-	-
0.2000	-	-29^2	-	-	-
0.5000	-	-47^2	-	-	-

---

* Represents the average of the number of determinations as indicated by the individual superscripts. Readings are relative to that shown by a 0.1000 molar solution of sodium salicylate in acetic acid.

Table III. Potentiometric Dilution Data for Organic Amines (continued).

Relative Millivolts*

MOLARITY	Tri-n-hexyl	Tri-n-heptyl	Tri-n-octyl	Tri-methyl	Di-iso-butyl
0.0001	-	-	82	-	85
0.0002	71	70	70	72	75
0.0004	61	62	61	63	66
0.0006	54	55	54	57	63^{2}
0.0010	47	49	50	50	56
0.0020	39	41	42	42	49^{2}
0.0030	34	36	36	36	43
0.0050	28	30	31	30	37^{2}
0.0100	20	21	22	20	27
0.0200	12	14	12	10	19
0.0500	2	4	2	-9	6
0.1000	-	-	-	-	-
0.2000	-	-	-	-	-
0.5000	-	-	-	-	-

---

* Represents the average of the number of determinations as indicated by the individual superscripts. Readings are relative to that shown by a 0.1000 molar solution of sodium salicylate in acetic acid.

Table III. Potentiometric Dilution Data for Organic Amines (continued).

	Relative Millivolts*			
MOLARITY	Di-n-butyl	n-butyl	Ammonium acetate	Tri-phenyl
0.0001	-	-	-	-
0.0002	80	83	83	-
0.0004	71	73	75	-
0.0006	64	-	70	-
0.0010	59	61	63	-
0.0020	49	52	53	-
0.0030	43	-	48	-
0.0050	38	39	42	-
0.0100	29	33	32	-
0.0200	20	-	22	-
0.0500	7	-	5	-
0.1000	-	-	-	-
0.2000	-	-	-	-
0.5000	-	-	-	-

---

* Represents the average of the number of determinations as indicated by the individual superscripts. Readings are relative to that shown by a 0.1000 molar solution of sodium salicylate in acetic acid.

Table IV. Potentiometric Dilution Data for Tetrabutylammonium Acetate.

MOLARITY	Relative Millivolts*
0.0002	43^{5}
0.0004	34^{4}
0.0006	28^{3}
0.0008	23^{3}
0.0010	21^{5}
0.0020	12^{3}
0.0030	7^{2}
0.0050	2^{3}
0.0100	-6^{6}
0.0200	-12^{4}
0.0340	-17^{2}
0.0500	-19^{2}
0.1350	-27^{2}
0.1690	-30^{2}
0.3390	-42^{2}
0.847	-75^{2}

---

* Represents the average of the number of determinations as indicated by the individual superscripts. Readings are relative to that shown by a 0.1000 molar solution of sodium salicylate in acetic acid.

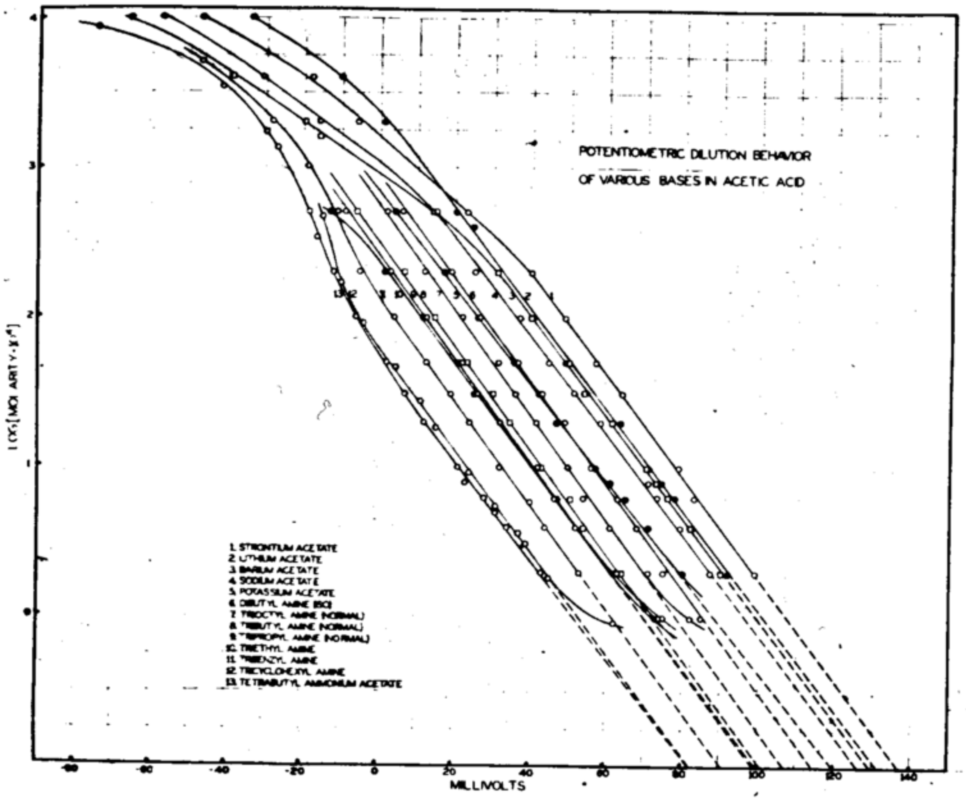


FIGURE 2. THE POTENTIOMETRIC DILUTION BEHAVIOR OF VARIOUS BASES IN ACETIC ACID.

the ratio of the relative dissociations of any two bases in acetic acid

$$\left[K_1(MAc)_1\right]^{\frac{1}{2}} = \left[K_2(MAc)_2\right]^{\frac{1}{2}} \qquad (28)$$

may be computed.

$$\frac{K_1}{K_2} = \frac{(MAc)_2}{(MAc)_1} \qquad (29)$$

By utilizing the concentration range 0.0001 to 0.050 molar, where the dilution curves were parallel to each other, it was possible to equilibrate acetate ion concentrations resulting from dissociation of the "ion-pairs." By the use of equation (29) it was possible to establish Table V showing the relative order of basicity for the nineteen different bases over a 500-fold range in total solute concentration. Also included in Table V are values of the slopes over this limited concentration range. These values indicate how closely the various bases conform to the law of incomplete dissociation, which theoretically requires a slope of 59.1/2 millivolts (0.5) per 10-fold change in total concentration.

The dissociation of strontium acetate was arbitrarily chosen as unity, all other values representing the dissociation of the "ion-pairs" of any given base compared to that of strontium acetate. Because of the low dielectric constant of acetic acid, the dissociation of the second acetate ion of strontium and barium acetates have been neglected as the extent of this dissociation was extremely small, if occurring at all (26).

The marked deviations from linearity shown by the dilution curves at higher solute concentrations may be attributed to changes in the nature of the solvent as the solute concentration was progressively increased. In acetic acid solutions of high solute concentration, the ratio of solvent molecules is not very large as compared with an aqueous solution,

Table V. Relative Dissociation Constants of Various Bases in Acetic Acid and Experimentally Determined Slopes in the Region 0.0001 to 0.05 Molar.

BASE	K_{base}/K_{SrAc_2}	Slope*
Strontium Acetate	1.0	0.50
Lithium Acetate	1.7	0.51
Barium Acetate	1.9	0.50
Sodium Acetate	2.5	0.52
Ammonium Acetate	3.6	0.50
n-Butylamine	4.1	0.50
Di-n-butylamine	4.9	0.50
Potassium Acetate	5.3	0.52
Di-isobutylamine	5.3	0.49
Trimethylamine	8.7	0.51
Tri-n-octylamine	9.2	0.49
Tri-n-heptylamine	9.6	0.49
Tri-n-hexylamine	10.0	0.49
Tri-n-butylamine	15.5	0.49
Tri-n-propylamine	18.6	0.49
Triethylamine	19.5	0.50
Tribenzylamine	34.8	0.49
Tricyclohexylamine	72.5	0.50
Tetrabutylammonium acetate	85.2	0.52

---

* Computed on the basis of the Ostwald Dilution law. A slope of 0.50 corresponds to a 59.1/2 millivolt change per 10-fold change in concentration.

hence as the number of solute molecules is increased, the characteristics of the solvent no longer approach that of the pure form, but are markedly altered and produce the anomalous behaviors observed. In very dilute concentrations, the solutions, tend to approach in the limit, that of pure acetic acid, with the limiting relative electromotive force measurements approaching that of the pure solvent.

Among the inorganic acetates, the results obtained by the present dilution procedure are in close agreement with those obtained by the conductance method of Kolthoff and Willman (26), the degree of dissociation increasing with larger ionic size and decreasing ionic charge. However, the same cannot be said for the organic tertiary amines, the size of the trialkylammonium ion not being as critical as the steric configurations of the alkyl groups.

The actual dependence of the relative basicities of a homologous series of tertiary amines, R_3N, on the length of the R- group is shown in Figure 3, where ammonium acetate, the first member of the series, has arbitrarily been assigned a dissociation value of unity. In examining this diagram it is quite apparent that the relative degrees of dissociation of different "ion-pairs" varies over a 5-fold range. The shape of the curve may be rationalised on the basis that the proton affinities of these bases have little or no influence on the relative degree of dissociation, the latter being influenced mainly by the size and shape of the positively charged cation. As the molecular size of the ammonium ion is increased from the unsubstituted to the trimethyl and triethyl nitrogen, the oppositely charged acetate ion was held off at greater and greater distance, resulting in significant increases in the apparent basicity. An increase in the alkyl chain beyond C_2H_5, however, tends to decrease the apparent basicity, gradually at first, followed by a sharp drop, and

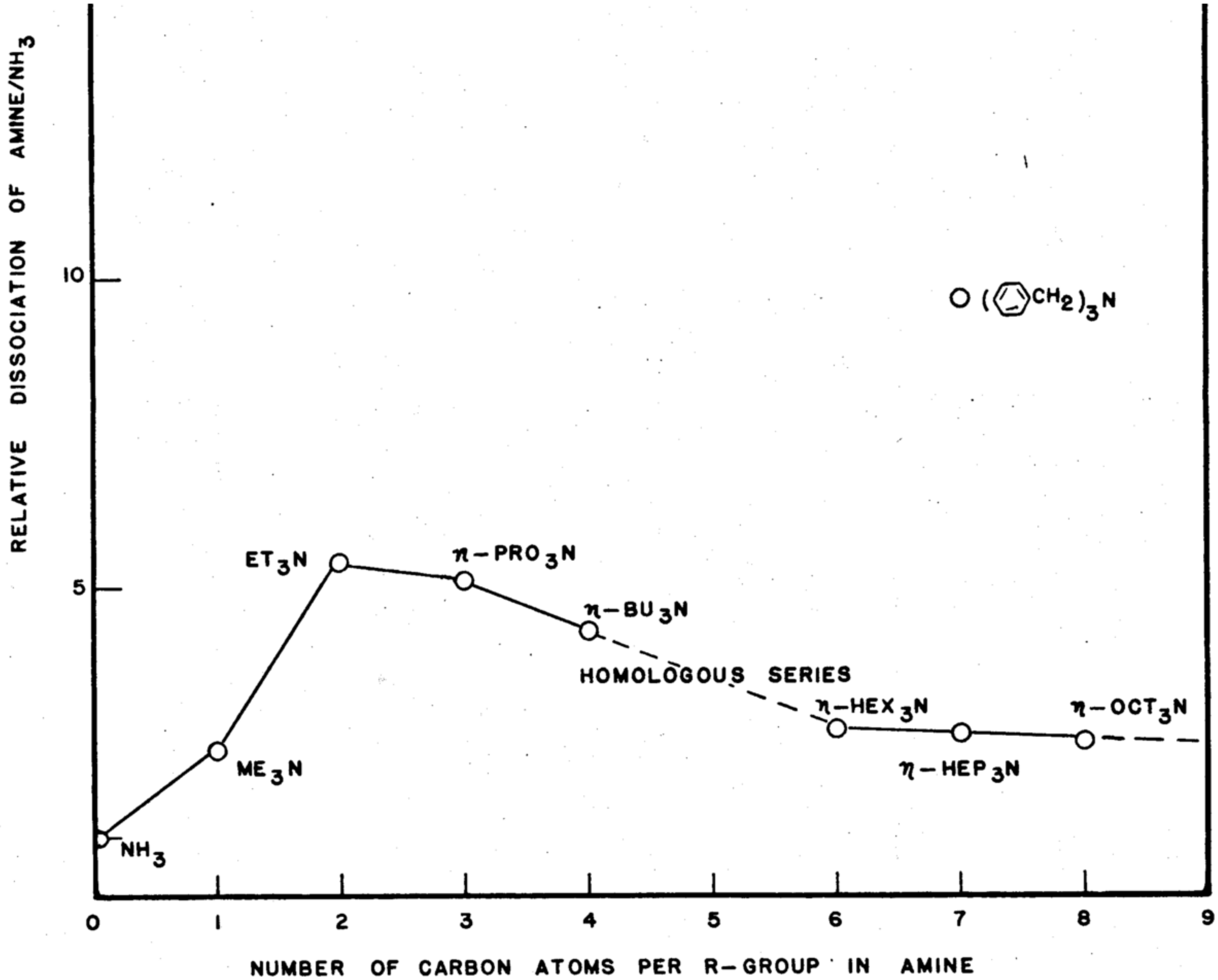


FIG. 3 VARIATIONS IN "ION-PAIR" DISSOCIATION OF ORGANIC AMINES IN ACETIC ACID WITH CHANGES IN CHAIN LENGTH AND CONFIGURATION.

finally a leveling off occurring beyond the n-C_6H_{13} alkyl chain. Consequently a molecule such as the trihexylammonium ion, despite the fact that it possesses a much larger volume than the triethylammonium ion, is a relatively less dissociated base which indicates that factors other than just size effect the extent of "ion-pair" dissociation.

The decrease in apparent basicity as the molecular size is increased beyond the ethyl group is difficult to comprehend; the following two paragraphs, however, are intended as speculation as to possible causes of this decrease.

In a solvent such as water, long chain hydrocarbon molecules tend to curl back on themselves in an effort to avoid the water molecules with which they are miscible only to a limited extent. In doing so, these chains tend to roll up into a somewhat compact mass, the interior of which contains the positive center of the solvation product, along with undissociated anion, the latter being held in the vicinity of the positive nucleus by a combination of coulombic forces and curled up alkyl chains. As the chains become progressively longer, the ability to retain anions within this "web-like" structure is enhanced, thereby decreasing the extent of dissociation of the solvation product. When the same amine is dissolved in an organic solvent such as acetic acid, the extent of repulsion between amine and solvent molecules is somewhat diminished, thus aliphatic chains have a greater tendency to remain stretched out in a long, spindly type of structure. As the length of the alkyl group is increased by the introduction of additional methylene groups, the chain becomes less rigid in character, and is capable of bending about the positive nucleus, thereby entrapping some of the anions within this "web-like" structure. However, in the case of an organic solvent, the amount of anion retained is much less than occurs in water, hence the variations in dissociation in an organic solvent are not as great.

Another possible factor which tends to influence the relative degrees of dissociation of the various amines may be attributed to what has been termed a "methyl effect."* The methyl group, being bulkier than the corresponding methylene group, is capable of affording a substantial increase in protection against a "squeezing in" effect by acetate ions between the alkyl chains. Hence, as the terminal methyl group is moved closer to the nucleus, the resulting dissociation of the solvated amine ("ion-pairs") increases, presumably due to this added shielding furnished by the methyl groups over methylene groups. However, a shortening of the alkyl chain beyond the ethyl radical produces a marked decrease in relative basicity, despite the fact that the nitrogen of trimethylamine has much better shielding than in triethylamine. This is attributed to the relatively small diameter of the trimethylammonium ion, making it possible for the coulombic forces of attraction to play a major part in determining the degree of "ion-pair" separation.

As evident from Figure 3, somewheres between the ethyl and n-propyl groups is located a critical size and shape necessary to provide the maximum physical barrier between the ions, thereby enhancing the dissociation. A branched propyl group or related structure, it would seem, should be capable of promoting much greater "ion-pair" separation than either the ethyl or n-propyl groups can afford. Triphenylamine, which possesses the isopropyl type branching, but a planar configuration, has the proper structural requirements. The three phenyl groups, however, are such excellent electron reservoirs that the nitrogen atom is virtually depleted of its unshared electron pair, thereby being unable to undergo

---

* This term "methyl effect" may be a misnomer.

the solvation which results in the formation of "ion-pairs".

The catalytic hydrogenation of triphenylamine (45) to tricyclohexylamine not only destroys the electron affinity of the phenyl groups, but introduces an electrodotic effect towards the nitrogen atom, thereby restoring basic properties and furnishing a structure with the proper size and steric configuration. Furthermore, the fixed configuration of the cyclohexyl radical prevents a curling up about the nitrogen atom, thereby not inhibiting the dissociation of the "ion-pairs." Confirmation of this enhanced dissociation is shown in Figure 3, tricyclohexylamine being approximately 21-times more dissociated than ammonium acetate. Similarly, tribenzylamine which does not possess quite the protection of tricyclohexylamine due to the planar configuration of the phenyl groups, undergoes approximately 10-times greater dissociation than ammonium acetate.

The tetraalkylammonium acetate bases in acetic acid are analogous in character and strength to tetraalkylammonium hydroxides in water. Both types are unable to undergo self-decomposition by the loss of solvent molecules as is apt to happen with solvated amines, hence both are capable of furnishing a high concentration of basic anion to their respective solvents. Furthermore, steric factors which play a prominent part in acetic acid solutions are greatly increased in a salt of this type, thereby enhancing the amount of "ion-pair" dissociation.

The difference in basicity between a sterically hindered tertiary amine and a tetraalkylammonium acetate is easily explained in terms of shielding about the nitrogen atom. Since the four covalent bonds of a tetraalkylammonium ion are symmetrically distributed about the tetrahedral nitrogen atom, the center of charge is quite deeply buried and the approach of the acetate ion is markedly reduced by the presence of four bulky alkyl groups. The distribution of groups about a tertiary amine which has undergone solvation is such, that even if the four bonds were to be equally distributed spacially, the presence of the proton as one of them would allow angular space into which the anion is capable of entering, reducing the interionic distance, and increasing the coulombic forces of attraction between the ions.

In aqueous solutions, the basicity of a substituted ammonia increases from the mono- to the dialkyl compound, followed by a decrease when a third identical alkyl group is substituted for the remaining hydrogen (50). As an example, in water, the basicity of the ethylamines varies accordingly:

$$NH_3 < C_2H_5NH_2 < (C_2H_5)_2NH >> (C_2H_5)_3N$$

This relationship was confirmed by Brown and Taylor (51), who further extended their work to reference acids other than the proton, i.e. trimethylboron, as a means of determining the relative basicities of an amine series. They concluded that the steric requirements of the ethyl group are quite large, thereby causing a greater dissociation of the addition product between triethylamine and trimethylboron, $(C_2H_5)_3N:B(CH_3)_3$. Davis and Hetzer (52) employed aprotic solvents to show that the strength of acids and bases is determined by the "leveling effect" of the solvent and steric factors associated with the molecule.

As previously shown, steric hindrance plays a prominent part in determining the extent of "ion-pair" dissociation in acetic acid. Table V shows the relative basicities of four different butylamines in acetic acid: n-butylamine, n-dibutylamine, di-isobutylamine, and tri-n-butylamine. The primary and secondary amines undergo relatively the same degree of dissociation for equimolar solutions, whereas the tertiary amine undergoes much greater dissociation. This reflects itself in shielding about the nitrogen nucleus and the ability of the relatively basic acetate ion to penetrate the physical barrier and enter into coulombic attraction with the cation.

In water: $BuNH_2 < Bu_2NH \gg Bu_3N$

In CH_3COOH: $BuNH_2 < Bu_2NH \ll Bu_3N$

From a comparison of experimental data in nonaqueous solvents with literature values for the same bases in water, it can be concluded that the order of strengths of any given group of bases in water is not a good guide to their strengths in a nonaqueous solvent. Analogies are not possible since water and other similar amphiprotic solvents exert a "leveling effect" on bases in the same manner in which they do on acids (53).

Even though molecules such as tetrabutylammonium acetate and tricyclohexylamine are effectively much stronger bases than a large majority of the other organic amines, their effectiveness in enhancing titration "breaks" is determined mainly by the extent to which the "ion-pairs" of the reactants and products are capable of undergoing dissociation. For any given reaction in acetic acid, the mass law may be written as in

$$R_3NHAc + HX \rightleftharpoons R_3NHX + HAc \qquad (30)$$

equations (31) and (32). Substituting for the right-hand members of the

$$K'' = \frac{(R_3NHX)(HAc)}{(R_3NHAc)(HX)} \qquad (31)$$

$$K' = \frac{(R_3NHX)}{(R_3NHAc)(HX)} \qquad (32)$$

expression, their corresponding dissociation equilibrium expressions, then:

$$K' = \frac{(R_3NH^+)(X^-)}{(R_3NH^+)(Ac^-)(H^+)(X^-)} \cdot \frac{(K_{R_3NHAc})(K_{HX})}{(K_{R_3NHX})} \qquad (33)$$

Canceling terms and taking the ion-product of hydrogen and acetate ions into the constant K', then the extent of the reaction may be expressed in terms of the various equilibria present.

$$K = \frac{(K_{R_3NHAc})(K_{HX})}{(K_{R_3NHX})} \qquad (34)$$

It is easily seen that the overall extent of reaction varies directly with the dissociation of the reactants and inversely with the dissociation of the products. Hence, as the interionic distance of the product ions decreases, less "ion-pair" dissociation takes place and the reaction is motivated towards the right side of reaction (30).

Further evidence of this dependency on the extent of "ion-pair" dissociation is borne out in the conductometric work of Higuchi and Rehm (54). In the titration of sulfuric acid with lithium, sodium, and potassium acetates, the specific conductance of the system rose as the first hydrogen was neutralized, the extent of the rise in conductance being determined by the size of the cation ($K^+ > Na^+ > Li^+$) employed. After complete neutralization of the first hydrogen, the conductance then fell, the drop being the largest with the smallest cation (Li^+). The second "break" was much more pronounced with lithium acetate than potassium acetate because of the smaller interionic distance and decreased dissociation. After either neutralization or complexing of the bisulfate ion had taken place, the conductance then increased with further additions of base due to a continual increase in the total ion concentration of the system.

The results obtained by the present potentiometric dilution procedure compare favorably with those obtained by Hall (23) who employed titrimetric methods, as to the relative order of dissociations. The present procedure has the distinct advantage of being free from interfering ions and changes in total solute concentration during the course of the investigation. The quantitative variations between the two sets of data (qualitative relationships being identical) may be attributed to the presence of perchlorate ion in the systems of Hall, as the relative order of basicity was established by $pH^{(HAc)}$ values at the midpoint of the titration where the logarithm of [salt/base] is zero.

On the basis of the results obtained in the present investigation, and as previously noted by Kolthoff and Willman (26), the extent of "ion-pair" dissociation is favored by increases in molecular size. Therefore, it appears that there are several inconsistancies with regards to order of basicity in the data obtained by Hall (23). Ammonium acetate, by Hall's potentiometric evaluation procedure, is regarded as a "stronger" base than either dimethylamine or diethylamine, both of which are substituted ammonias and consequently possess larger molecular sizes than the unsubstituted form. Furthermore, Hall reported tri-n-butylamine and di-n-butylamine to have the same pK' values in acetic acid, whereas in the present investigation, tri-n-butylamine is a much "stronger" base as would be anticipated from the steric factors involved.

PART IIa Determination of Indicator Constants (pK') in Terms of The Hammett Acidity Function, H_o (34).

The use of indicators in acetic acid has been limited essentially to those which show distinguishable color transitions in the moderately and strongly acidic regions of this solvent. Very little attention has been paid to the development and use of indicators which exhibit color changes in the relatively alkaline regions of acetic acid, mainly because of the lack of bases of sufficient strength to produce these changes. It has been pointed out by Davis and Schuhmann (63) that "Before acid-base measurements of organic solutions can be performed as readily as those of aqueous solutions, it will be necessary to provide a series of indicators, ranging from strongly acidic to strongly basic compounds. A further need is for stable, crystalline organic acids and bases sufficiently strong to neutralize weak bases and acids."

In an effort to obtain quantitative data on the behavior of several indicators which exhibit color changes in the relatively alkaline regions of acetic acid, their dissociation constants (pK') in terms of the Hammett Acidity Function (34) have been evaluated using the spectrophotometric procedure previously described. In this manner the regions of indicator color change have been established with the aim of using these values in extending the relatively alkaline regions of the Hammett Acidity Scale in acetic acid, as well as providing for indicators to be used in acid-base titrimetry. The chemistry and observations made on each indicator are discussed in this section.

Before turning to the individual indicators, it may be well to discuss certain aspects of the underlying theories which apply in particular to the method of study employed. In the following discussion, liberalities have been taken in using the terms A12 through A23 and C23. The solutions represented by these code letters have been adequately discussed in the experimental section.

In the present method of evaluating the pK' of various indicators, a plot of H_0 against the logarithm of the ionization ratio, BH^+/B, was made (see equation 19), and the value at which $H_0 = pK'$ graphically computed.

As the acidity function scale was originally established in water-sulfuric acid systems (both solvents possessing high dielectric constants), the salt form of the indicator, BH^+, was fairly independent of the anion present due to the high degree of dissociation occurring. This independence of the cation from anion is not necessarily true in acetic acid because of its low dielectric constant as compared with the sulfuric acid-water system. As a result, deviations in slope from the theoretical value anticipated by equation (19) may be attributed in part, to an equilibrium between the dissociated salt form, BH^+, and the undissociated salt form, BH^+A^-.

Another factor which is encountered in strongly acidic solutions has been termed by Flexser et.al. as the "medium effect" (47). An indicator which behaves properly exhibits a well defined isosbestic point*, all

---

* "If a given solute exists in two forms or reaction products, i.e. B and BH^+, each of which possesses a characteristic absorption which is constant for a series of solutions, both of the forms have equal extinction coefficients at some wave length, if, that is, the absorption curves of the two pure individuals intersect, then likewise all solutions must have the same coefficient at this wave length, because they contain only various mixtures of these individuals. Conversely, if two absorption curves intersect at a point which is not common to all other curves, then it must be true either that more than two forms or reaction products of the solute must be present, or else the change in conditions, for instance, in acidity, which produces variation in the proportion of the two forms also changes their specific absorption by what may be called a medium effect (47)."

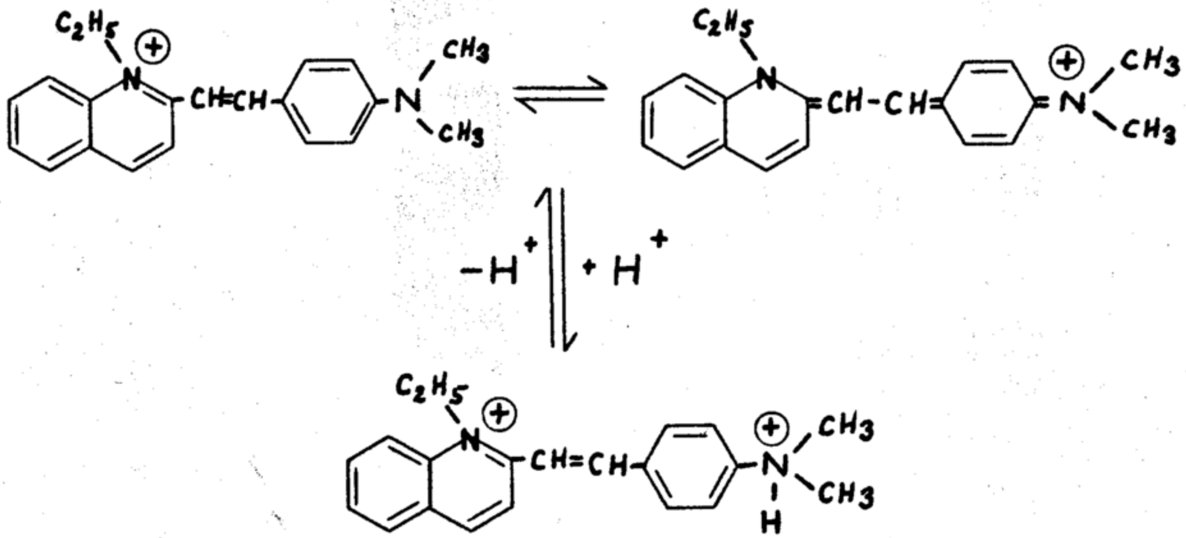
spectrophotometric curves passing through it with only minor variations. In strongly acidic solutions, absorption curves show a tendency to be displaced, the exact reasons for these displacements not being clearly understood. However, they may be attributed to resonance changes within the molecule caused by the high degree of acidity, the presence of counterions, or other unexplainable factors. In correcting for this "medium effect," Flexser, Hammett, and Dingwall (47) assumed that the intensity of the absorption spectrum is unaffected, but that merely a lateral displacement of the curve has taken place. Therefore, the curves may be shifted laterally so that all pass through a common isosbestic point with the intersection of the two curves most closely representing 50 per cent of the indicator in the salt form being chosen as this reference point. In this manner changes in the absorption spectra of the various indicators produced by counterions, stronger acids, and stronger bases have been accounted for and the appropriate corrections applied.

With certain basic type indicators, the absorption intensity (optical density) of the salt form of an indicator, BH^+, differs with the different acid species present. In solutions of all the indicators investigated, except benzopurpurin 4B, perchloric acid merely served to intensify the absorption band without passing through an isosbestic point with the A12 buffer solution (antipyrine.H_2SO_4). Generally the resulting intensity increase was not very significant, hence the acid form of the indicator was taken as being the spectrum produced in the A12 buffer solution. With benzopurpurin 4B, a definite isosbestic point was present and the perchloric acid spectrum was used as the completely acidic form of the indicator.

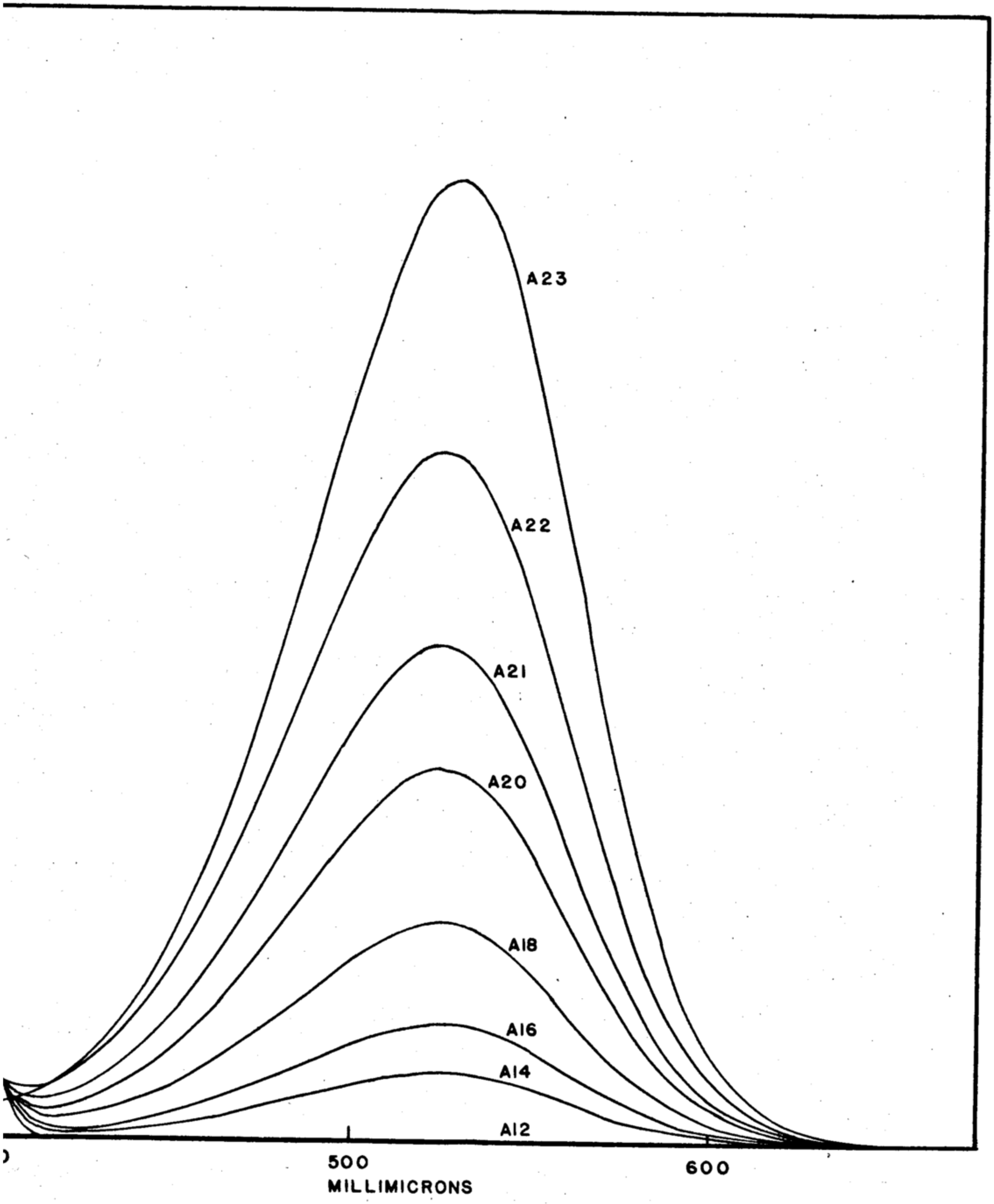
In the relatively more alkaline solutions A23 and C23 (0.1000 molar antipyrine and tricyclohexylamine, respectively), "medium effects" were not visible to any extent. With the exception of thymol blue, which is a strongly alkaline indicator in acetic acid, the absorption intensity of the alkaline form was nearly identical with both A23 and C23 solutions, the observable differences being quite small.

Quinaldine Red

Quinaldine red is a one color indicator possessing a fairly sharp endpoint in both acetic acid and aqueous solutions. In water the color transition takes place over the pH range 1 to 2, whereas in acetic acid, the change occurs in the relatively alkaline regions, its pK' having been determined experimentally as 3.44. The structure of quinaldine red, along with resonance forms are shown below.



The results of the spectrophotometric data for quinaldine red in the acetic acid buffer solutions of Hall and Meyer (37) are shown in Table VI. Figure 4 shows the absorption spectra from which these data were obtained. A careful examination of this diagram showed that all solutions containing sulfuric acid (A12 to A22) had their absorption maxima at the same wave length (525 millimicrons), whereas the pure antipyrine solution had



RPTION SPECTRA OF QUINALDINE RED IN THE BUFFERS OF
AND MEYER (37).

its maximum shifted slightly towards a longer wave length. This shift in maximum was attributed to the counterion or "medium effect." A lateral displacement of the spectrum in the A23 solution so that it passed thru the isosbestic point common to all other solutions, showed the position of the absorption maximum to occur at 525 millimicrons, this corresponding to that of the other absorption spectra. The indicator constant as determined graphically, along with the slope of the ionization ratio curve are shown in Figure 5.

Table VI. Spectrophotometric Data for Quinaldine Red.

Solution	Acid Band 525 mμ O.D.	Log BH^+/B
$HClO_4$*	0.000	-
A12	0.000	-
A14	0.118	1.10
A16	0.200	0.85
A18	0.370	0.53
A20	0.629	0.20
A21	0.838	-0.03
A22	1.165	-0.41
A23	1.620	-
C23**	1.620	-

In order to substantiate the fact that the presence of sulfuric acid

---

* A 0.500 molar solution of perchloric acid in acetic acid.

** C23 is a 0.1000 molar solution of tricyclohexylamine in acetic acid.

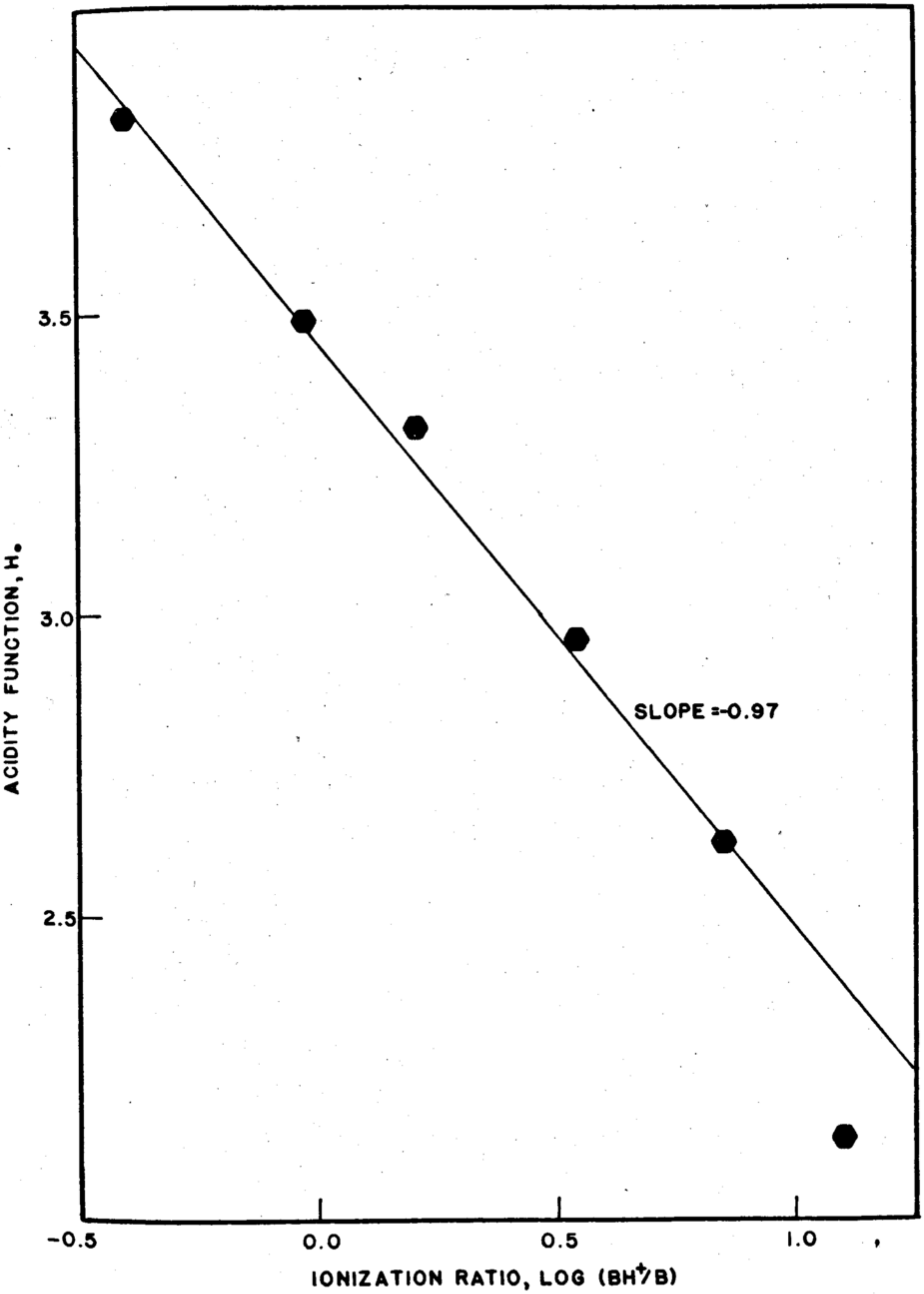


FIG. 5 QUINALDINE RED, $_pK' = +3.44$

molecules caused a shift in the position of the absorption maximum to slightly shorter wave lengths, a series of solutions whose composition ranged between that of A22 and A23 were prepared. The sulfuric acid content of these solutions progressively decreased as shown in Table VII. The acidity function values of these solutions were unnecessary as it was only desired to show the shift in the curve maximum as the sulfuric acid concentration was decreased. In all cases, the solutions were 0.1000 molar in total solute concentration.

Table VII. The Effect of Sulfuric Acid on the Position of the Absorption Maximum of Quinaldine Red.

Solution	Antipyrine Molarity	Antipyrine H_2SO_4: M	Peak at ___ mμ
$A22_2$	0.09555	0.00445	525.0
$A22_3$	0.09666	0.00334	525.9
$A22_4$	0.09777	0.00223	526.7
$A22_5$	0.09888	0.00112	527.4
A23	0.10000	-	528.8

Figure 6 shows the changes in the position of the absorption maxima as the composition of the solutions were varied, the peak wave lengths having been determined from a highly resolved recording of each spectrum. As the position of the absorption maximum of solution $A22_2$ coincided with that of all solutions of greater acidity, the removal of sulfuric acid from the system caused a shift of approximately 3.8 millimicrons in the absorption spectrum.

The influence of five different acetate bases, i.e. 0.1000 molar

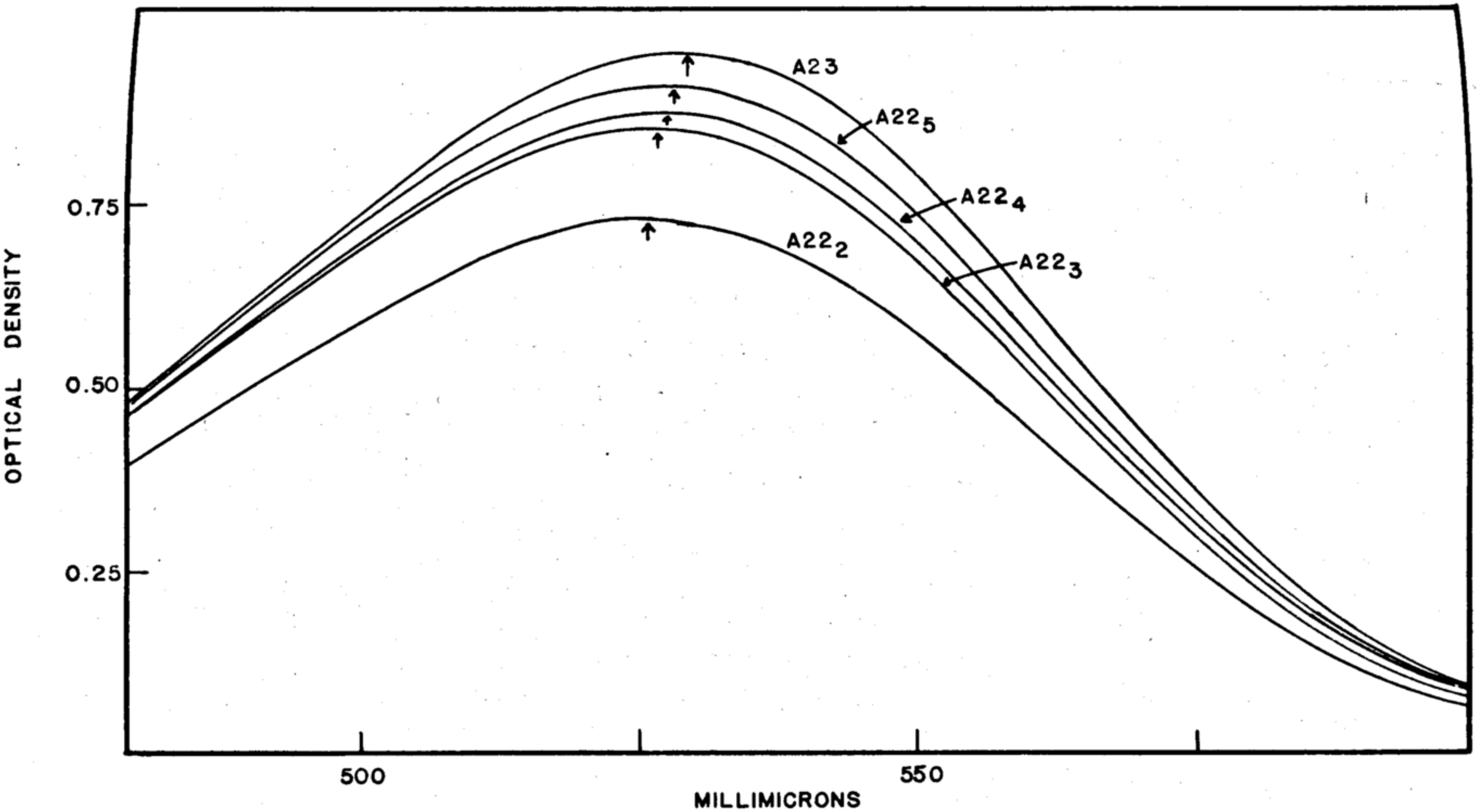


FIG. 6 THE EFFECT OF SULFURIC ACID ON THE ABSORPTION SPECTRA QUINALDINE RED.

solutions of potassium acetate, sodium acetate, strontium acetate, antipyrine, and tricyclohexylamine, on quinaldine red are shown in Figure 7. A highly resolved recording of these spectra showed only minor variations to take place between 527 and 527.5 millimicrons. In each instance the indicator concentration was the same, however the curves were shifted upward 0.05 optical density unit to eliminate overlapping.

The results obtained by varying the cations and anions present in the solution are in agreement with those to be expected of a positively charged indicator in a low dielectric constant solvent. In the presence of an acid, quinaldine red adds a proton on the amino-nitrogen, thereby introducing a second positive charge into the molecule. In accordance with the laws of attraction and repulsion, the presence of different cation bases having a common anion have no effect on the indicator, whereas varying the acidic anions tends to form "ion-pairs" of different combinations with quinaldine red. These different "ion-pair" combinations produce slight shifts in the absorption spectrum of quinaldine red, the extent of the shift being dependent upon the anion present.

The absence of any interaction between quinaldine red and (1) antipyrine or (2) sodium acetate, as well as the uniform changes occurring in the optical density with changing indicator concentrations are shown in Figures 8 and 9. The composition of the various solutions are given in Tables VIII (sodium acetate) and IX (antipyrine).

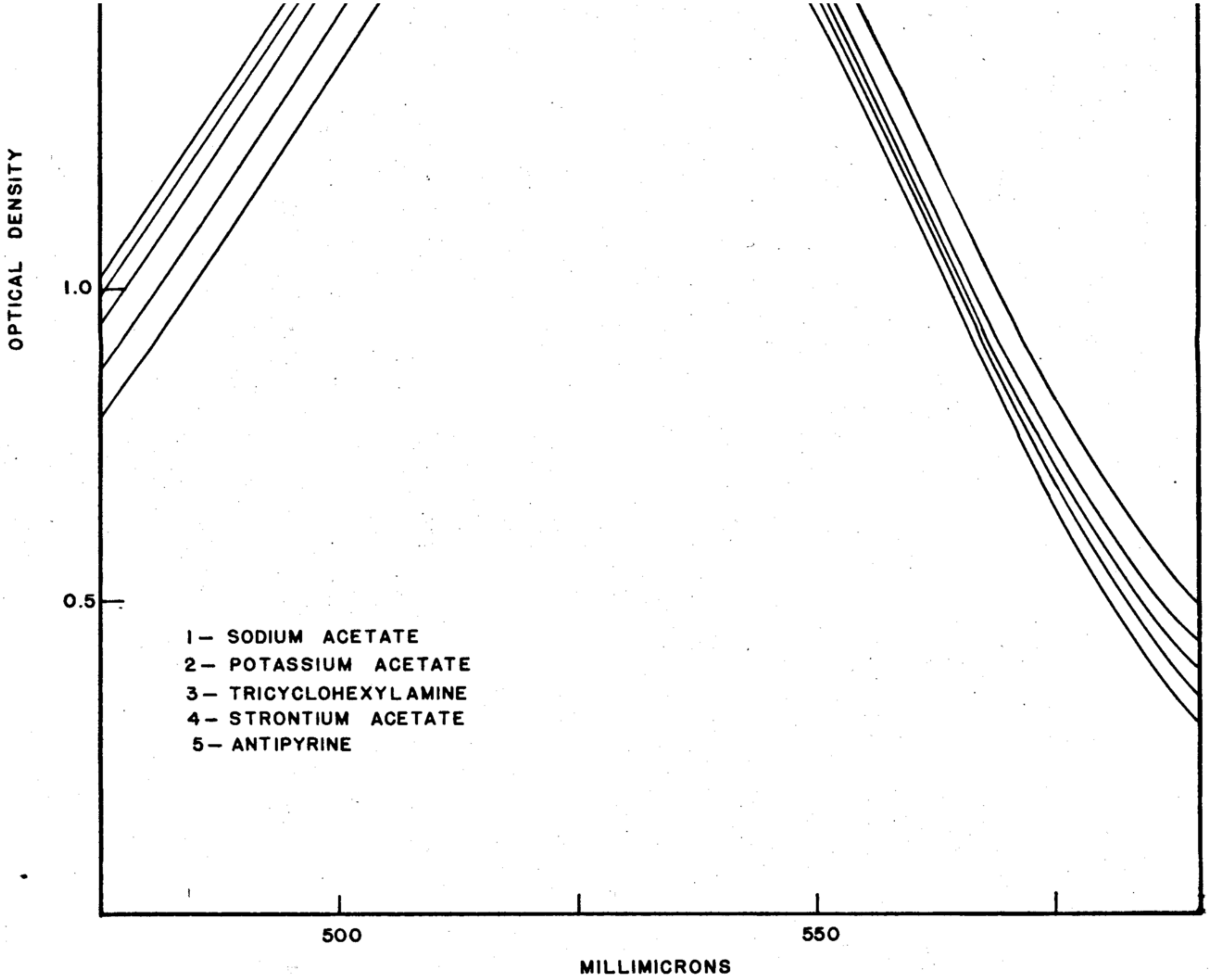


FIG. 7 THE EFFECT OF VARIOUS CATION ACETATE BASES ON THE ABSORPTION SPECTRUM OF QUINALDINE RED.

Table VIII. Changes in Optical Density with Changes in Indicator Concentration at Constant Sodium Acetate Concentration.

Solution	Final Salt Molarity	ml. Indicator Solution	Optical Density
I_1	0.08695	0.0	0.000
I_2	0.08695	0.5	0.417
I_3	0.08695	1.0	0.823
I_4	0.08695	1.5	1.231
I_5	0.08695	2.0	1.640
I_6	0.08695	2.5	2.055

As shown by the last column of Table VIII, the optical density readings were proportional to the indicator concentration, showing that Beer's law was followed over a wide range of indicator concentrations. This was further confirmed by optical density measurements made with 0.1, 1.0, and 10.0 centimeter quartz absorption cells. Each 10-fold increase in cell length was accompanied by a corresponding 10-fold dilution in indicator concentration. In all three cases, the transmission of the solutions were equivalent as required by Beer's law.

$$\text{Optical Density} = \log I_0/I = E'cl \qquad (35)$$

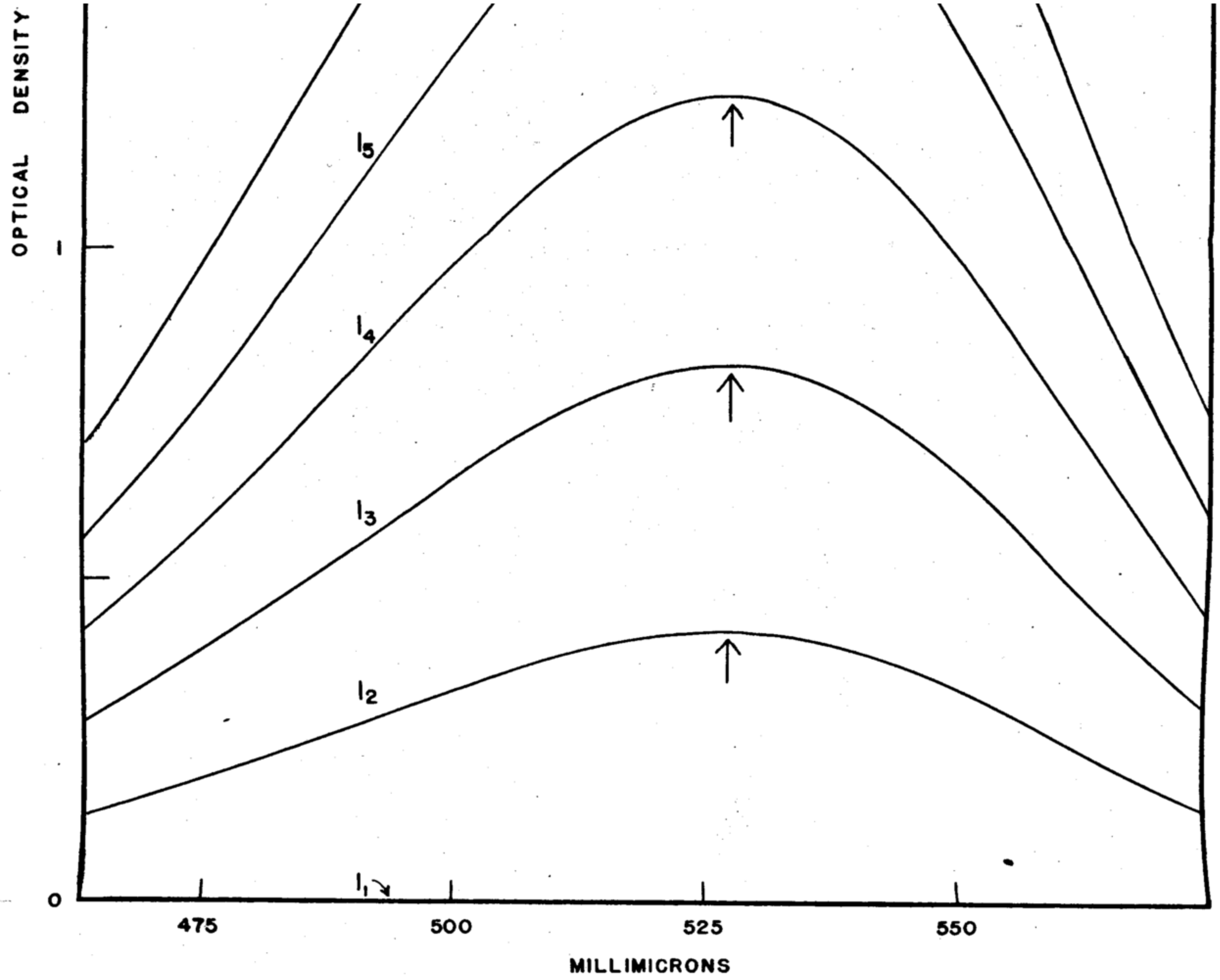


FIG. 8 VARIATIONS IN THE ABSORPTION SPECTRUM WITH CHANGES IN QUINALDINE RED CONCENTRATION AT CONSTANT SODIUM ACETATE CONCENTRATION.

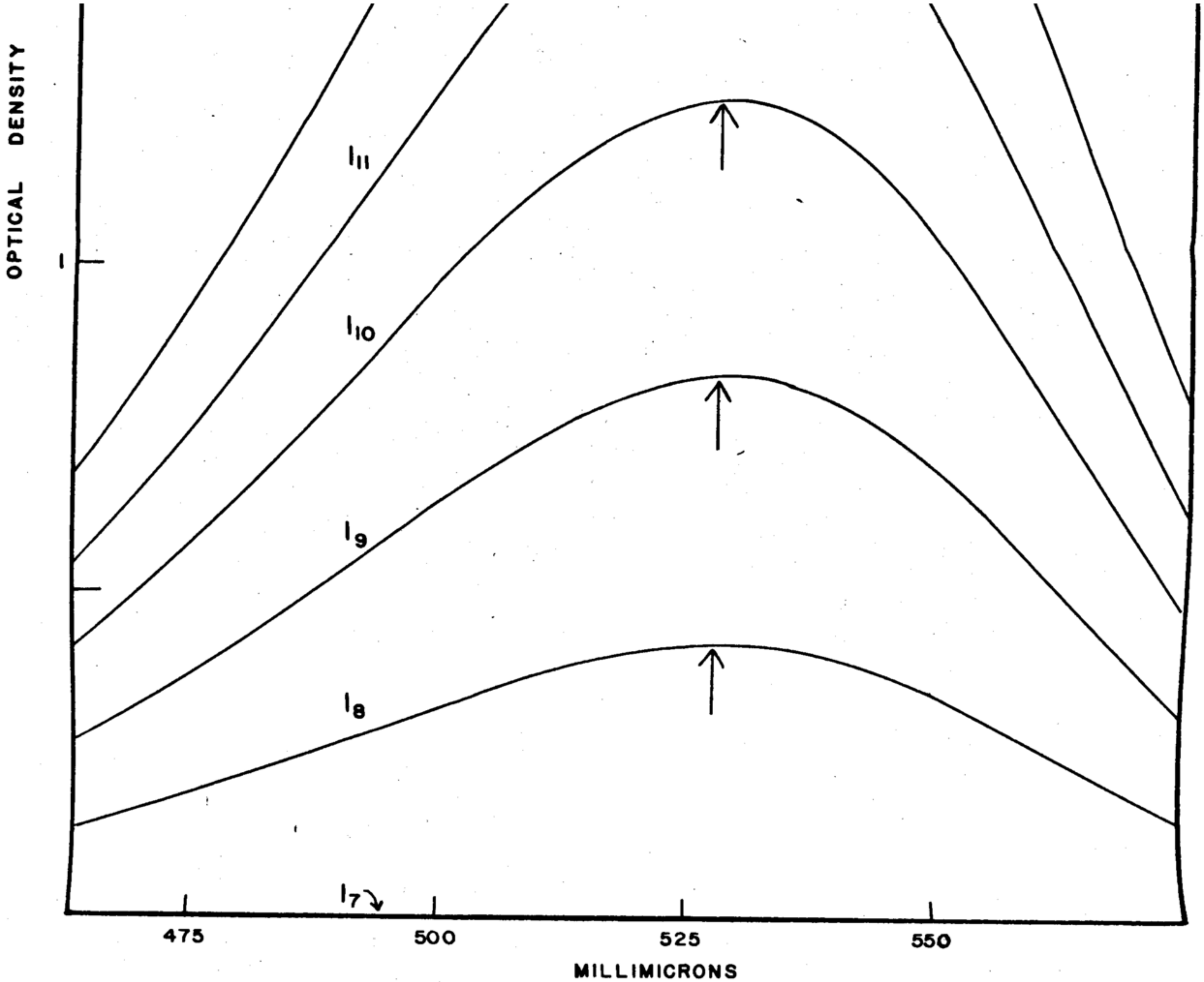


FIG. 9 VARIATIONS IN THE ABSORPTION SPECTRUM WITH CHANGES IN QUINALDINE RED CONCENTRATION AT CONSTANT ANTIPYRINE CONCENTRATION.

Table IX. Changes in Optical Density with Changes in Indicator Concentration at Constant Antipyrine Concentration.

Solution	Final Salt Molarity	ml. Indicator Solution	Optical Density
I_7	0.08695	0.0	0.000
I_8	0.08695	0.5	0.423
I_9	0.08695	1.0	0.840
I_{10}	0.08695	1.5	1.262
I_{11}	0.08695	2.0	1.661
I_{12}	0.08695	2.5	2.090

Variations in total salt concentration ("ionic Strength") produced no significant changes in the absorption spectrum of quinaldine red. Table X shows the composition of the various solutions whose spectra are recorded in Figure 10.

Table X. Changes in Absorption Spectra with Changes in Total Salt Concentration at Constant Indicator Strength.

Solution	Total Salt Molarity	ml. Indicator Solution	Optical Density
B_1	0.10000	1.0	0.950
B_2	0.06667	1.0	0.950
B_3	0.03333	1.0	0.944

The slight variation in solution B_3 as compared to B_1 and B_2 may be attributed to experimental error in adding indicator to the solution, since the variation in optical density is only 0.006 unit and the spectra are identical.

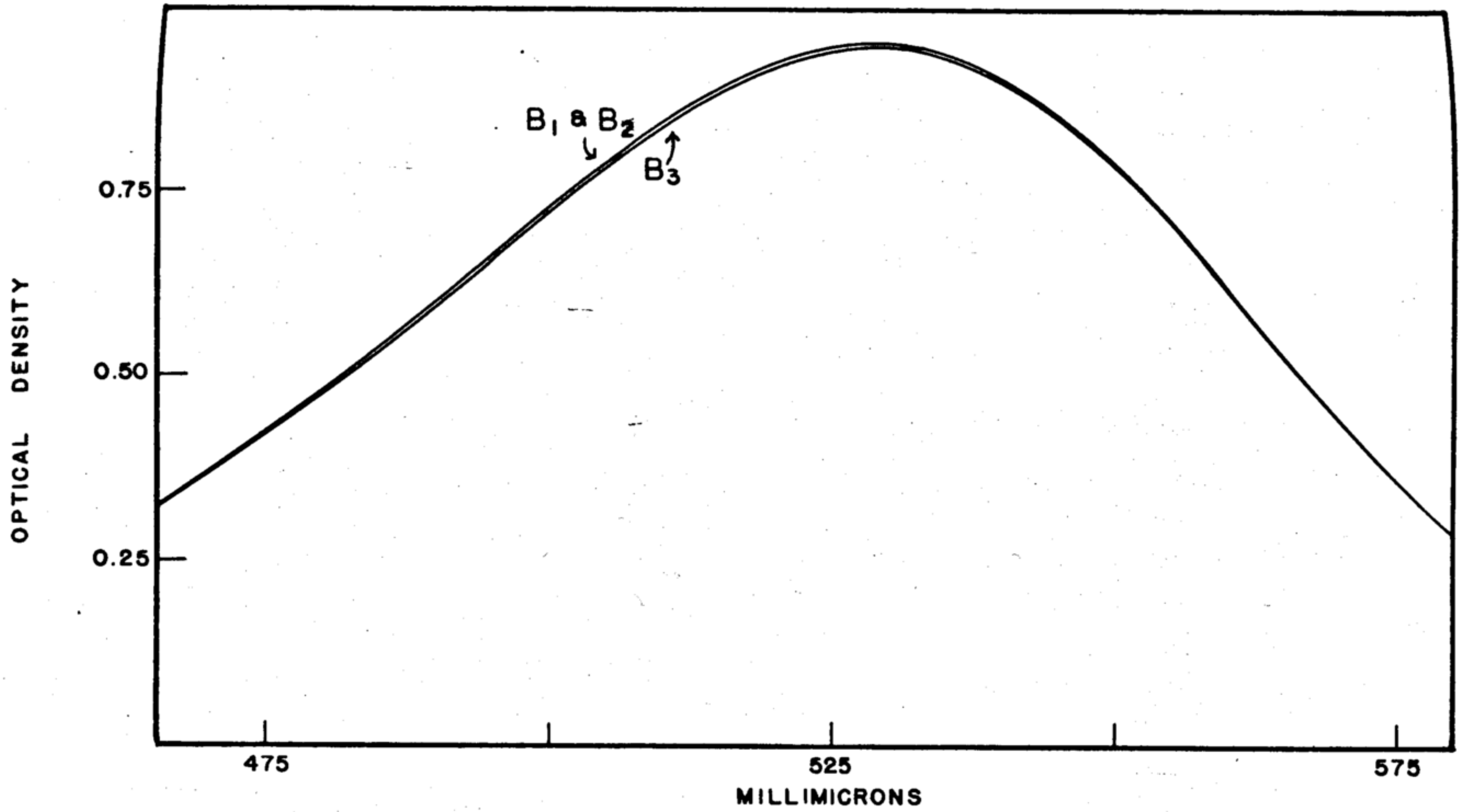


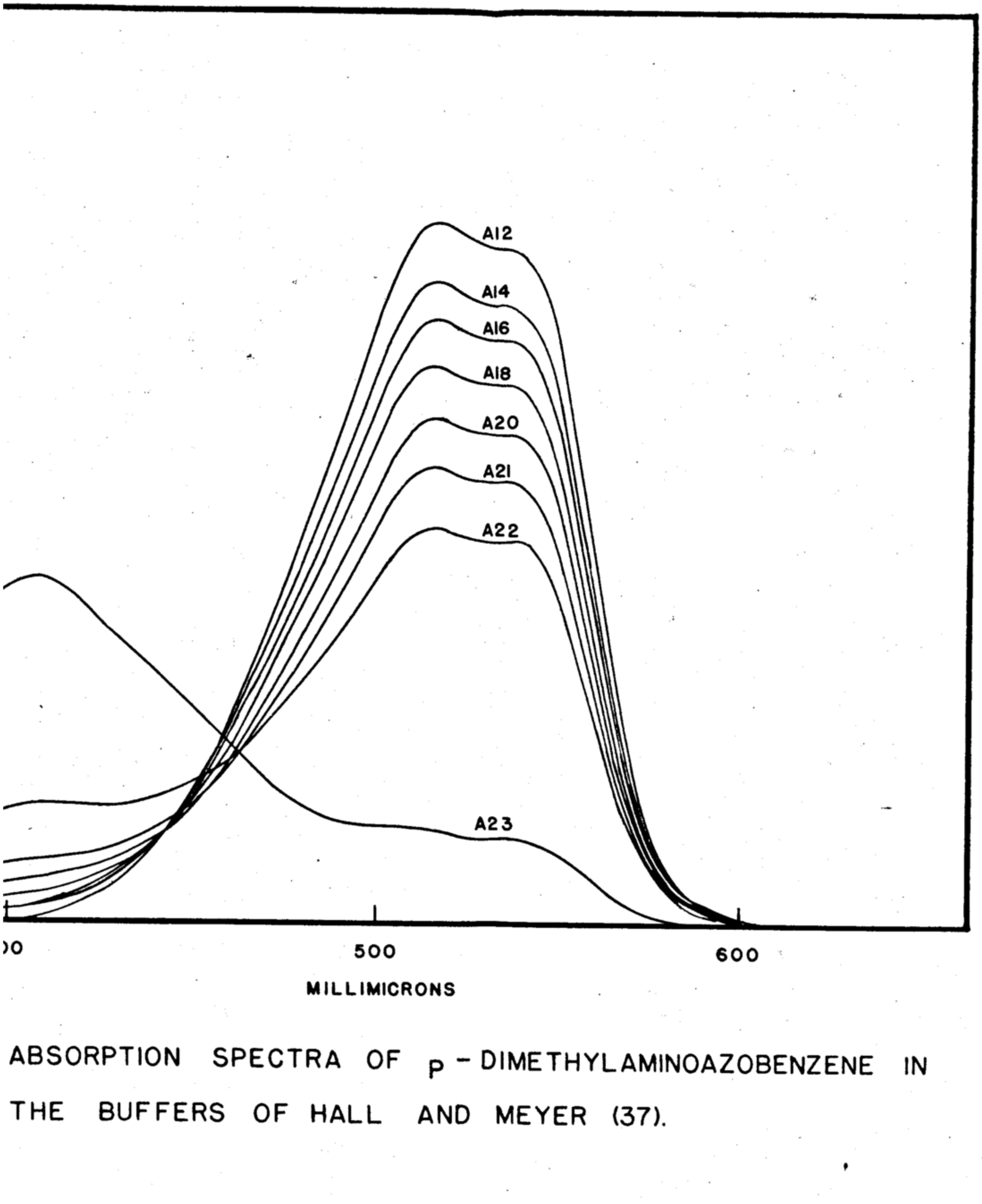
FIG. 10 THE EFFECT OF VARIATIONS IN TOTAL SALT MOLARITY ON THE ABSORPTION INTENSITY OF QUINALDINE RED IN THE BASIC FORM.

With regards to the practical use of quinaldine red as an indicator in acetic acid, Higuchi and Concha (57,58) have shown that it could be used titrimetrically to differentiate between various basic anions. As the color change occurs in the relatively basic regions of the solvent, these workers were able to titrate acetate and chloride ions as bases in the presence of each other.

p-Dimethylaminoazobenzene ("Butter Yellow," Methyl Yellow)

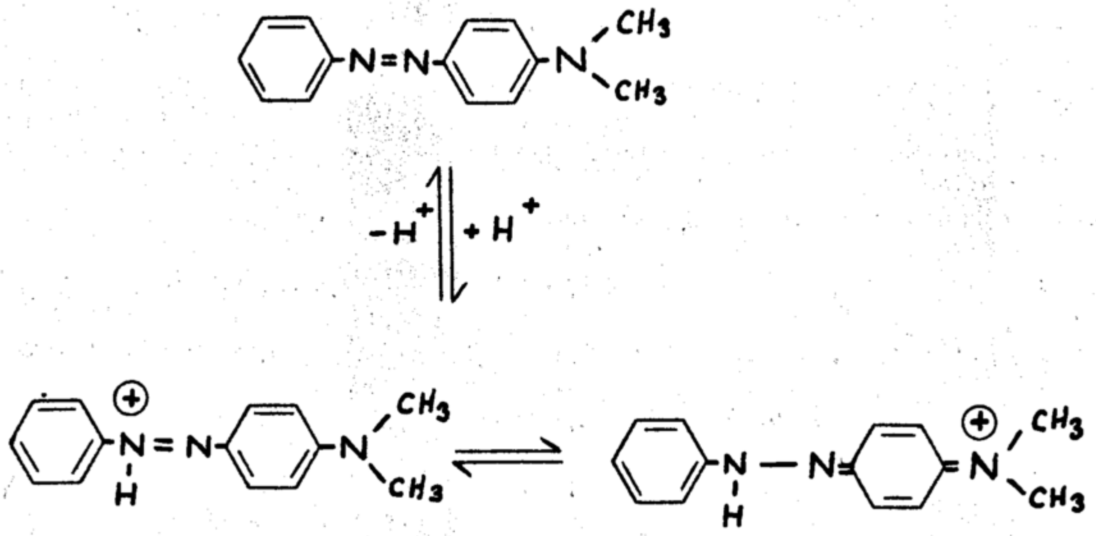
p-Dimethylaminoazobenzene is a two color indicator, being yellow and red in the alkaline and acid forms, respectively. The transition interval in water occurs over the pH range 2.8 to 4.2, whereas the indicator constant in acetic acid has been experimentally determined as 3.85. In acetic acid, the color change is not too sharp, hence its value for titrimetry purposes is limited. The usefulness of this indicator lies in its ability to show color variations in the highly alkaline regions of acetic acid solutions, thereby making possible the establishment of higher H_o values. The absorption spectra for this indicator, as shown in Figure 11, exhibits a strong acid band at 514 millimicrons which has been used to compute the ionization ratio. The graphical evaluation of pK' is shown in Figure 12, the values being obtained from the spectrophotometric data in Table XI.

Methyl yellow is a rather peculiar behaving indicator in acetic acid as evidenced by the slope of -1.81 shown by the ionization ratio curve in Figure 12. This seems to indicate that approximately two protons are being accepted for each molecule of indicator prior to a change in color. The addition of the first proton takes place on the azo-nitrogen farthest removed from the amino-nitrogen, giving rise to the structures shown

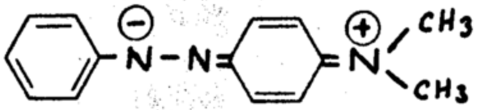


ABSORPTION SPECTRA OF p-DIMETHYLAMINOAZOBENZENE IN THE BUFFERS OF HALL AND MEYER (37).

below.



This represents the predominant form since a consideration of resonance within the uncharged base indicates that the greatest separation of charge occurs between the amine-nitrogen and the most distant azo-nitrogen, thereby favoring addition of the proton on the latter.



Addition of a second proton to p-dimethylaminoazobenzene, in all probability, takes place on the dimethylamino-nitrogen, rather than on the second azo-nitrogen because of the instability of adjacent positive charges. The addition of this second proton produces very little change in the absorption spectrum as no additional resonance structures of importance can be written for it. Hantzsch (59) has shown that various groups on the amine-nitrogen of this indicator, i.e. $-NH_2$, $-N(CH_3)_2$, $-N(C_2H_5)_2$, and $-N(CH_3)_3I$, produced no significant changes in the absorption spectrum, all possessing spectra closely related to azobenzene itself.

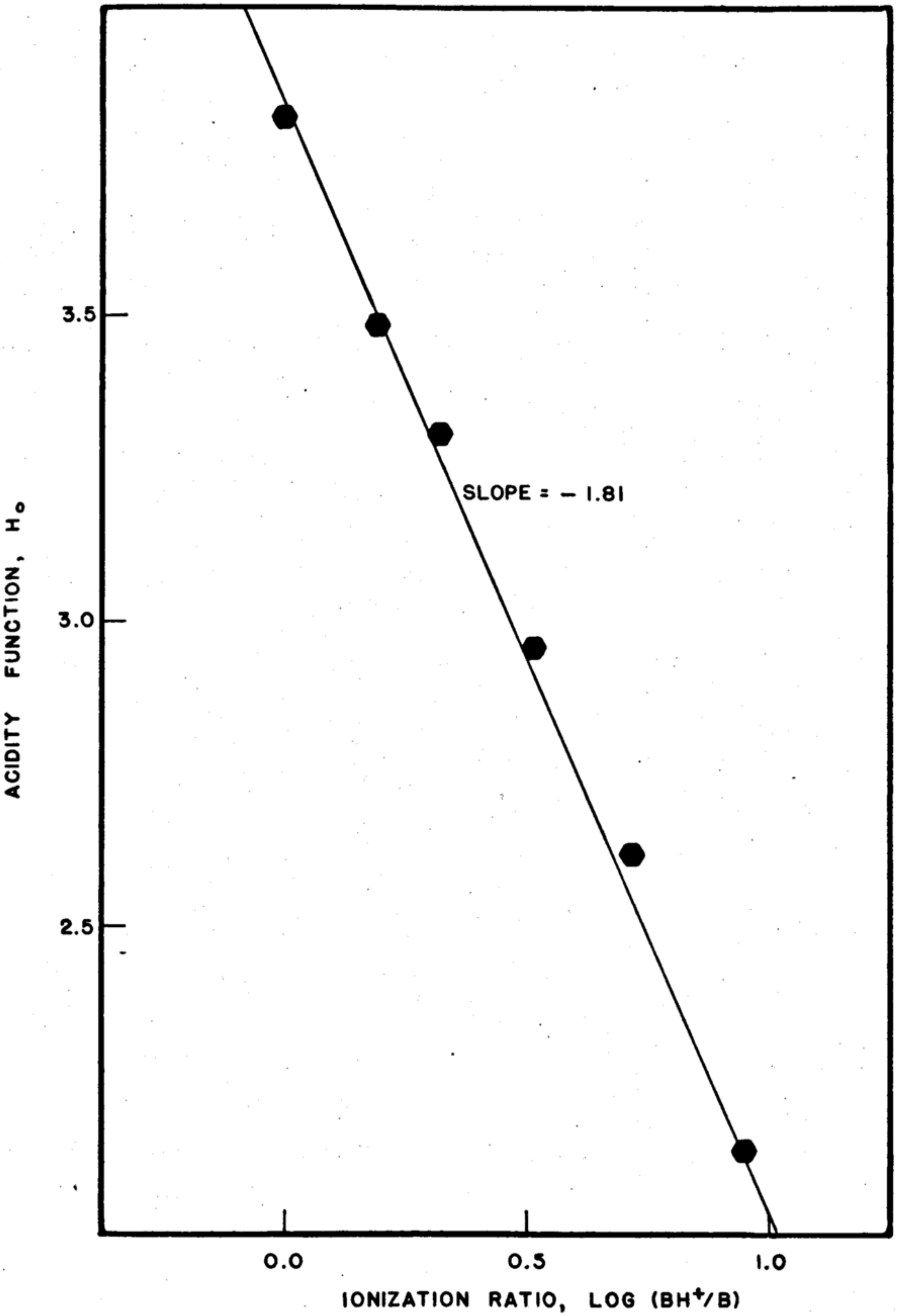
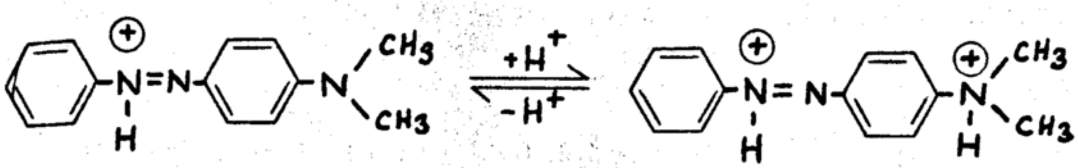


FIG. 12 p − DIMETHYLAMINOAZOBENZENE, $_pK' = 3.85$



In the more acidic buffer solutions, A12 to A21, a common isosbestic point was observed. This point showed a shift towards the longer wave lengths as the solution becomes progressively more alkaline (A22, A23, C23, and a highly concentrated C23 solution) in character, which seems to indicate that the indicator form present is undergoing a change as the relative alkalinity of the solution changes. The optical density measurements of the various alkaline solutions in the region of the absorption maximum remained fairly constant, thereby showing that the indicator was entirely in its alkaline form.

Perchloric acid merely intensified the absorption of the acid form without having any isosbestic point coincident with that of the A12 solution, indicating that a counterion effect may have been present.

Table XI. Spectrophotometric Data for p-Dimethylaminoazobenzene.

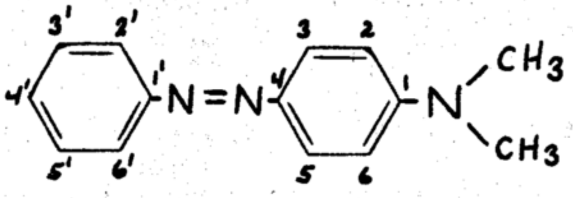
Solution	Acid Band 514 mμ O.D.	Log BH^+/B
$HClO_4$	1.243	-
A12	1.157	-
A14	1.055	0.95
A16	0.996	0.72
A18	0.919	0.51
A20	0.833	0.32
A21	0.752	0.16
A22	0.650	-0.01
A23	0.155	-
C23	0.155	-
C23S*	0.160	-

In order to show the effects of various substituent groups on the dissociation constant (pK') of "butter yellow," several substituted derivatives were spectrophotometrically investigated. The results are given in Table XII. The presence of negative substituents on the ring proper tends to decrease the value of pK' without having much effect on the color properties of the indicator. However, negative substituents not directly on the ring, as in the case of 2'-CF_3- p-dimethylaminoazobenzene, also lower the value of pK', but the color change of the indicator is markedly altered. The presence of an ethyl group as compared to a methyl group

---

* C23S is a highly concentrated solution to tricyclohexylamine.

in the 4' position gives a slightly higher value for the indicator constant, pK'. This is in agreement with theoretical expectations because of the greater inductive effect of the ethyl group.



Ring positions for substituted
"butter yellow" derivatives.

p-Phenylaminoazobenzene (Benzeneazodiphenylamine)

p-Phenylaminoazobenzene, like all the azo dyes, is a two color indicator derived from the parent structure azobenzene. The spectrum of this indicator was quite sharply defined in acetic acid, showing strong acid and alkaline absorption bands at 537 and 405 millimicrons, respectively. In aqueous solutions, this indicator undergoes color change from violet to yellow over the pH range 1.2 to 2.9. In acetic acid, the value of pK' was experimentally determined as 2.79, being considerably less alkaline than "butter yellow."

The absorption spectra for this indicator are shown in Figure 13, the ionization ratio values (Table XIII) having been computed from the acid band at 537 millimicrons. A similar calculation employing the alkaline band at 405 millimicrons gave the same value for pK'. The graphical evaluation of pK' from the acid band at 537 millimicrons is shown in Figure 14.

Table XII. Substituted "Butter Yellow" Derivatives

Derivative[+]	Wave Length mμ	pK'	Ionization Ratio Curve Slope	Acid Color	Alkaline Color
2',5'-di-F-	538	2.56	-1.24	pink	yellow
2,6-di-F-	490	3.03	-0.74	pink	yellow
3',4'-di-F-	509	3.12	-1.36	pink	yellow
2',4',6'-tri-F-	502	3.29	-0.88	straw yellow	bright yellow
3',5'-di-F-	410	3.34	-1.12	pink	yellow
2'-CF_3-	425	3.43	-0.93	colorless	yellow
4'-CH_3-	526	3.70	-1.46	violet	orange
4'-C_2H_5-	526	3.79	-1.40	violet	orange

[+] Gratitude is expressed to Dr. J. A. Miller of the University of Wisconsin Department of Oncology for supplying these dyes.

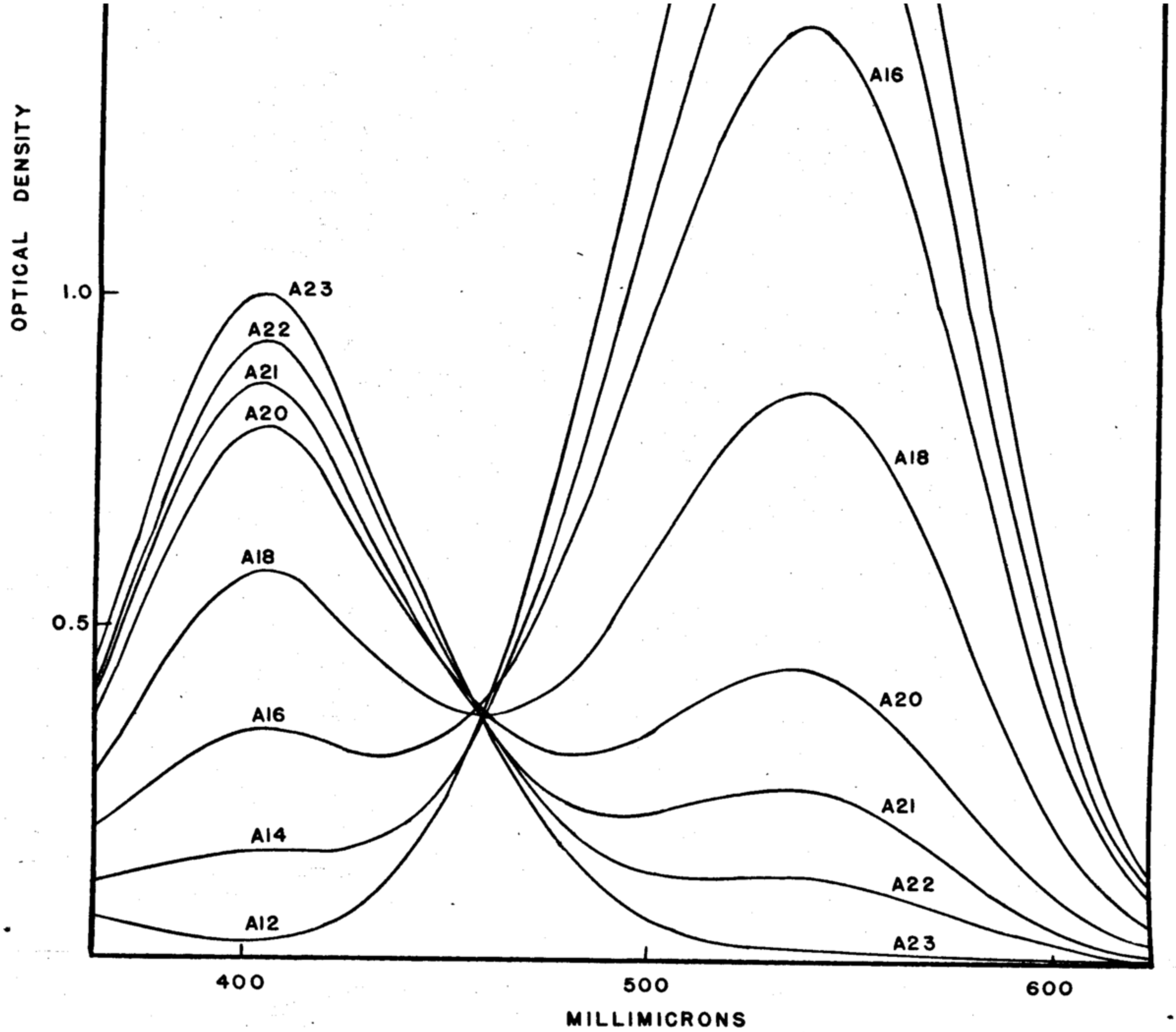
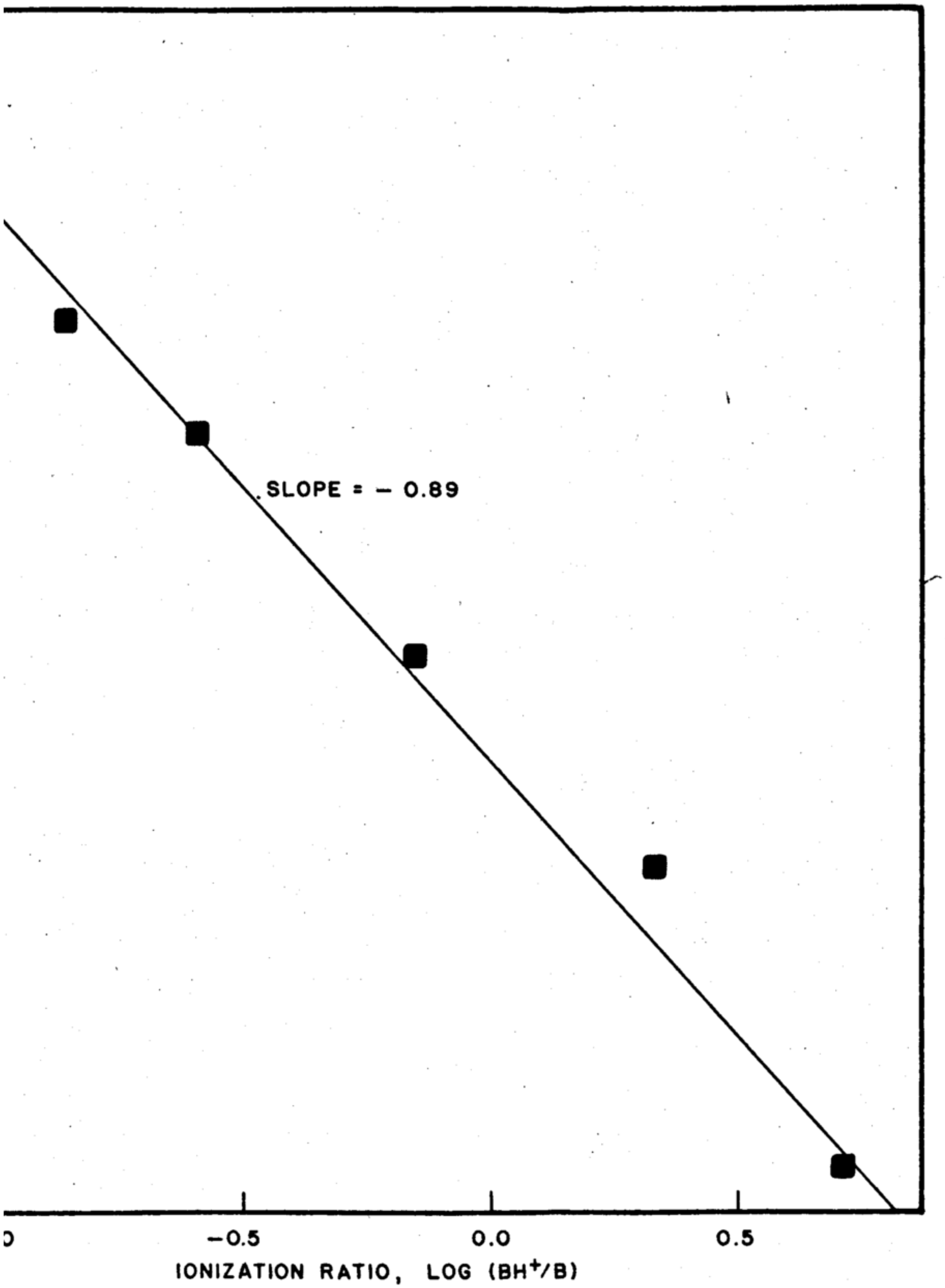
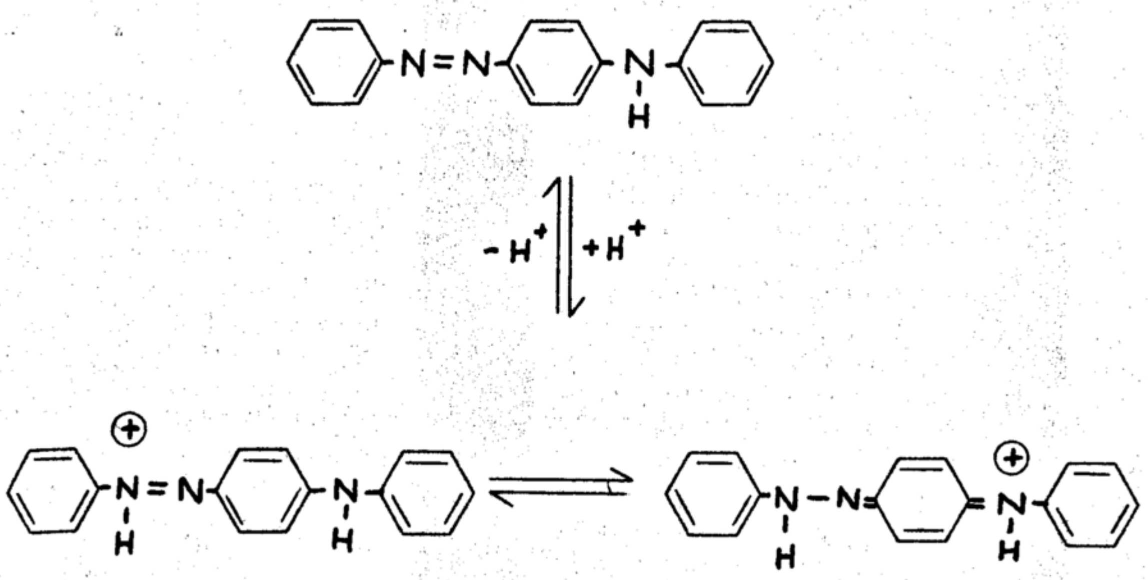


FIG. 13 ABSORPTION SPECTRA OF p-PHENYLAMINOAZOBENZENE IN THE BUFFERS OF HALL AND MEYER (37).



- PHENYLAMINOAZOBENZENE, ${}_{p}K' = +2.79$



As shown above, p-phenylaminoazobenzene behaves as a monoprotic base, the electron affinity of the phenyl group reducing the ability of the amine-nitrogen to add a second proton to the molecule.

Perchloric acid does not have much effect on the absorption spectrum, the band intensity being virtually unchanged, but shifted slightly towards the shorter wave lengths. Likewise, any effects produced by a tricyclohexylamine solution are not noticeable.

The slope of the ionization ratio curve was -0.89 which tends to indicate that the salt form of the indicator, BH^+, may not be entirely independent of the anion, but rather an equilibrium was present between the two forms.

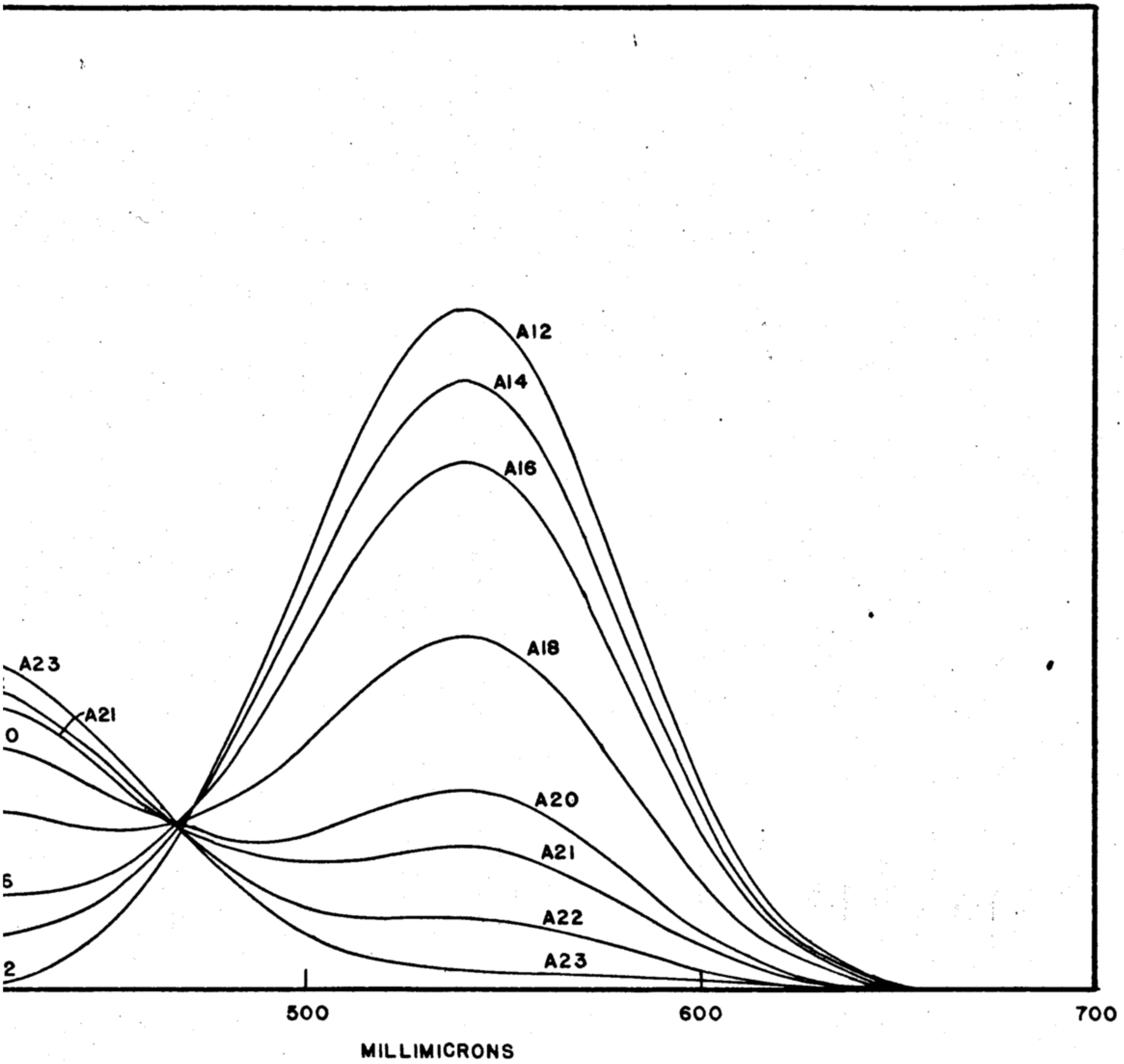
As an indicator, the color change was quite pronounced, making the endpoint very easy to observe.

Table XIII. Spectrophotometric Data for p-Phenylaminoazobenzene.

Solution	Acid Band 537 mμ O.D.	Log HI^+/B
$HClO_4$	2.060	-
A12	2.045	-
A14	1.728	0.71
A16	1.417	0.34
A18	0.863	-0.15
A20	0.441	-0.59
A21	0.262	-0.86
A22	0.130	-1.22
A23	0.021	-
C23	0.015	-

Tropaeolin OO (Orange IV)

Orange IV, like the previous two indicators discussed, is a derivative of azobenzene, exhibiting a color change over the pH region 1.4 to 2.8 in aqueous solutions. As an indicator in acetic acid, its color change was quite sharp, being red and yellow in the acid and alkaline forms, respectively. The experimentally determined value of pK' in acetic acid was 2.94, being graphically evaluated from Figure 16 and the ionization ratio data given in Table XIV.



PTION SPECTRA OF ORANGE IV (TROPAEOLIN OO) IN THE
RS OF HALL AND MEYER (37).

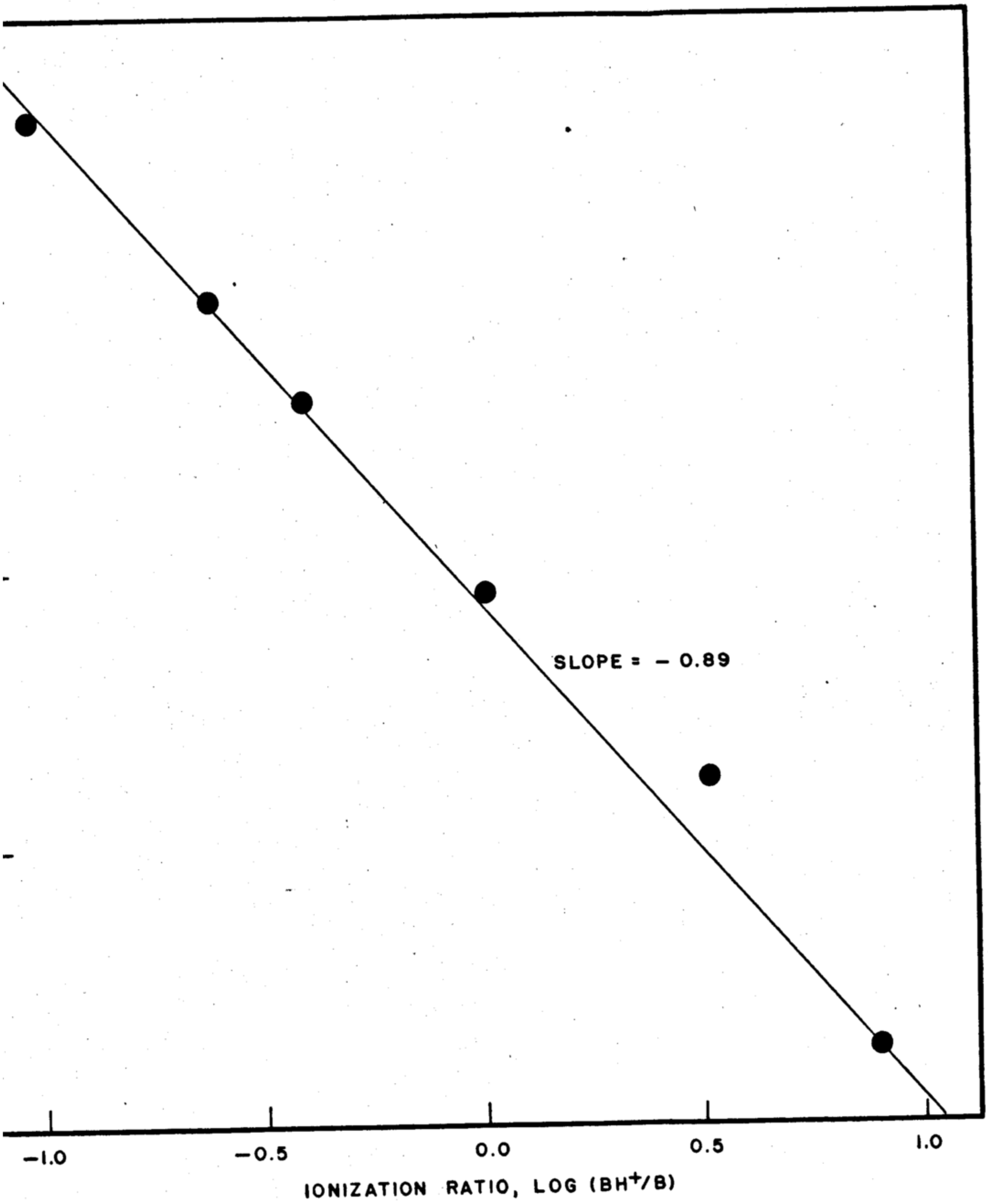
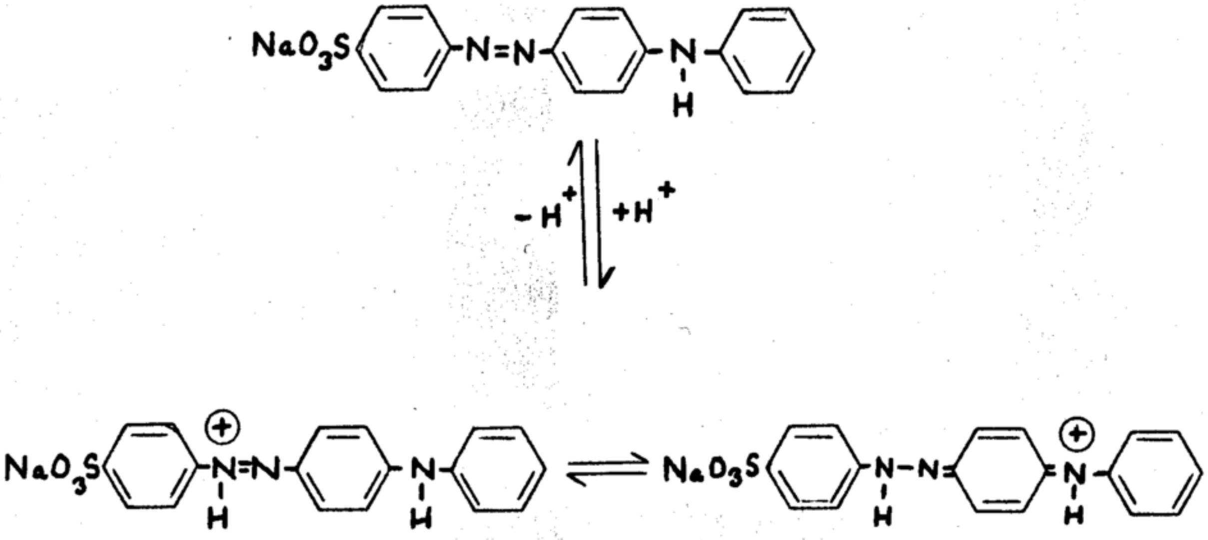


FIG. 16 ORANGE IV (TROPAEOLIN OO), $_{p}K' = +2.94$



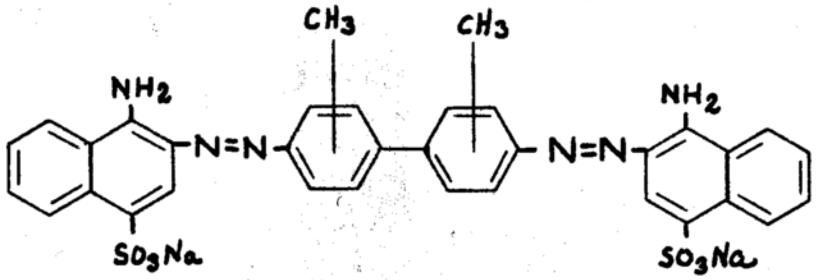
The absorption spectra of the various solutions shown in Figure 15 exhibit a well defined isosbestic point with all the curves passing through it. Both perchloric acid and tricyclohexylamine solutions exert a "medium effect" on this indicator, as would be anticipated, since it is the salt of an azo dye. A solution of perchloric acid shifted the position of the acid band approximately 8 millimicrons towards the longer wave lengths with no change in absorption intensity. Tricyclohexylamine solution also shifted the absorption curve towards the longer wave lengths by approximately 5 millimicrons, however it was accompanied by a slight change in absorption intensity. Consequently, the ionization ratio was computed at 541 millimicrons where absorption by the alkaline form of the indicator was at a minimum.

Table XIV. Spectrophotometric Data for Tropaeolin OO.

Solution	Acid Band 541 mμ O.D.	Log HI^+/B
$HClO_4$	1.049	-
A12	1.041	-
A14	0.931	0.89
A16	0.818	0.52
A18	0.541	0.00
A20	0.308	-0.41
A21	0.220	-0.62
A22	0.118	-1.02
A23	0.032	-
O23	0.021	-

Benzopurpurin 4B

Another azo dye which has shown promise as an indicator in acetic acid titrimetry is benzopurpurin 4B. Differing from congo red only by the introduction of methyl groups on the biphenyl portion of the molecule, this indicator undergoes its color change in the pH region 1.2 to 4.0. The transition from the acidic to basic forms in water is not sharp, hence its value in titrimetry is limited. In acetic acid, the experimentally determined value of the indicator constant, pK', was 3.14, the colored forms of the indicator being sky-blue and pink in acid and alkaline solutions, respectively.



The experimental evaluation of the indicator constant, pK', for benzopurpurin 4B is shown in Figures 17 and 18. The absorption spectra shown in Figure 17 were obtained after allowing a saturated solution of the indicator to equilibrate for three days in acetic acid, before filtering and using*. The absorption intensities (optical densities) and ionization ratio data are given in Table XV.

Due to symmetry in structure, this indicator was expected to add only one proton. This was confirmed by the slope of -1.07 in the ionization ratio curve shown in Figure 18. The addition of the proton takes place, in all probability, on the azo-nitrogen farthest removed from the amine group as in most other azo dyes. The addition of the proton at this point produces the most stable configuration for charge resonance between the benzoid and isomeric quinoid structural forms.

The spectrum in a perchloric acid solution of this indicator was taken as the acid form since it showed a definite isosbestic point with the A12 solution. A solution of tricyclohexylamine also produced a "medium effect" on this indicator, however at 555 millimicrons, where the ionization ratio was computed, the absorption intensities of the A23 and C23 solutions were identical (see Table XV).

---

* The nature of the absorption curves varied with the "age" of the benzopurpurin 4B solution.

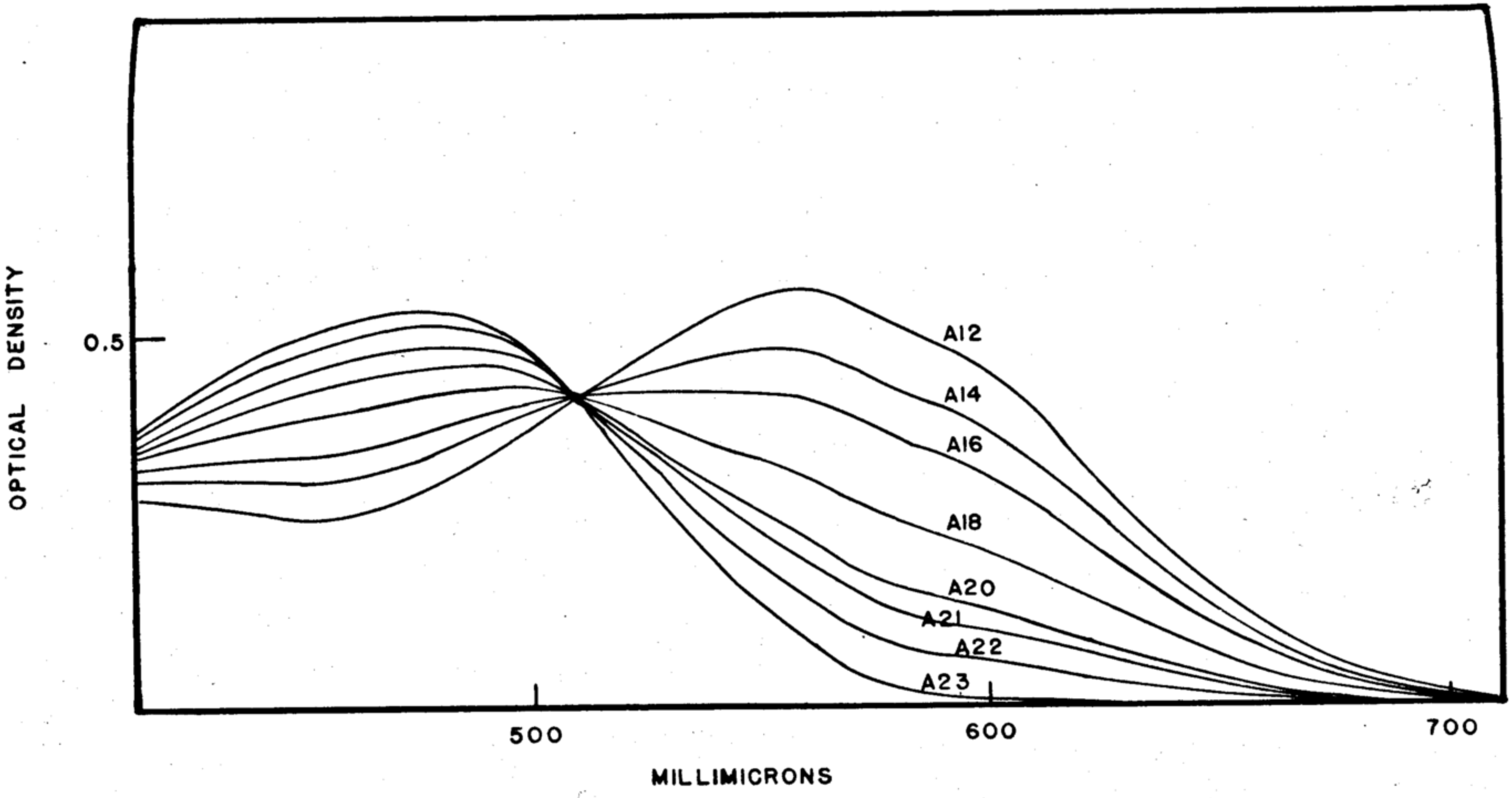


FIG. 17 ABSORPTION SPECTRA OF BENZOPURPURIN 4B IN THE BUFFERS OF HALL AND MEYER (37).

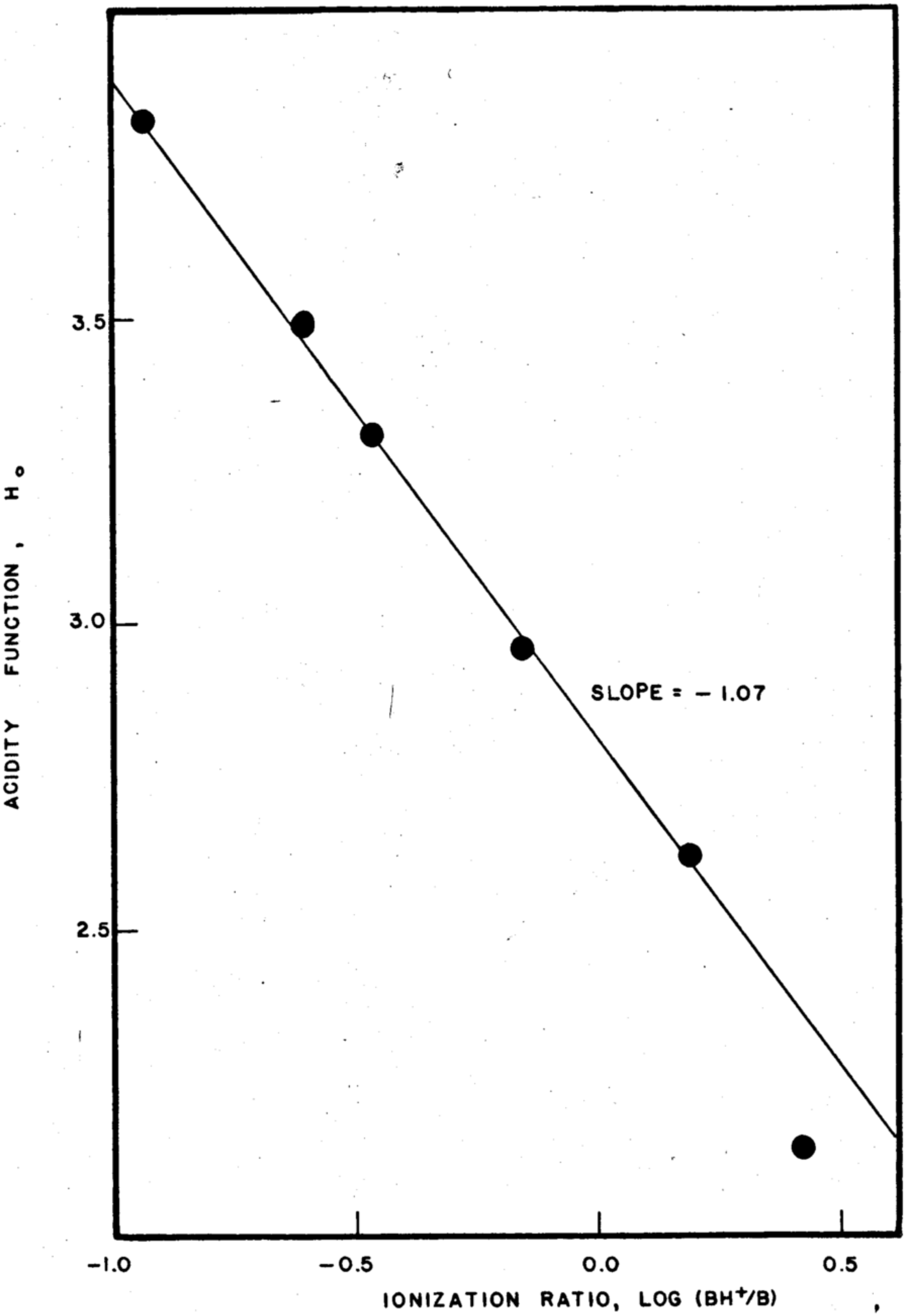


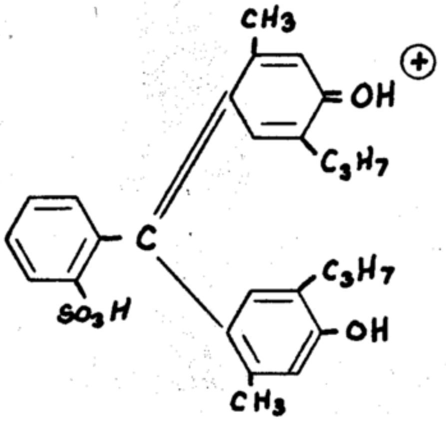
FIG. 18 BENZOPURPURIN 4B, K' = + 2.81

Table XV. Spectrophotometric Data for Benzopurpurin 4B.

Solution	Acid Band 555 mμ O.D.	Log BH^+/B
$HClO_4$	0.620	-
A12	0.557	0.87
A14	0.478	0.42
A16	0.411	0.18
A18	0.310	-0.16
A20	0.228	-0.47
A21	0.199	-0.61
A22	0.150	-0.93
A23	0.095	-
C23	0.095	-

Thymol Blue (Thymosulfonphthalein)

The investigation of thymol blue has been carried out to note the behavior of a protogenic indicator in acetic acid. In aqueous solutions, two color change intervals are observed, the first taking place in the pH range 1.2 to 2.8 and the second occurring between 8.0 and 9.5. In acetic acid, only one color change takes place, the experimentally evaluated indicator constant being 4.24 (Figure 20 and Table XVI). The color transition from the acidic to alkaline forms is not sharp, but merely a continuous color change of increasing or decreasing intensity (see Figure 33). The use of this indicator lies in its ability to distinguish degrees of acidity in the relatively alkaline regions of acetic acid because of its high pK' value, thereby extending H_0 values into the slightly more positive regions of this solvent.



By use of the ionization ratio data obtained by Danguilan (42) for thymol blue in sodium acetate, the value of pK' was computed as 4.25 which is in close agreement with the value of 4.24 using the buffers of Hall and Meyer (37). The sodium acetate concentration at which the logarithm BH^+/B vanished was obtained from a graph of sodium acetate concentration against ionization ratio. This sodium acetate concentration was then converted to its relative electromotive force measurement by means of Figure 2, and using the conversion chart of Danguilan's (42) for H_o into relative millivolts, the value of 4.25 was obtained.

The experimentally determined pK' of 4.24 for thymol blue was used to obtain an approximate H_o value of 4.13 for the antipyrine solution, A23. This calculation was possible since the solutions A23 and C23 were quite different in their absorption intensities, indicating that A23 was insufficiently basic to convert thymol blue to its alkaline form. At 546 millimicrons, where the ionization ratio was computed, absorption by the C23 solution was at a minimum as shown in Table XVI.

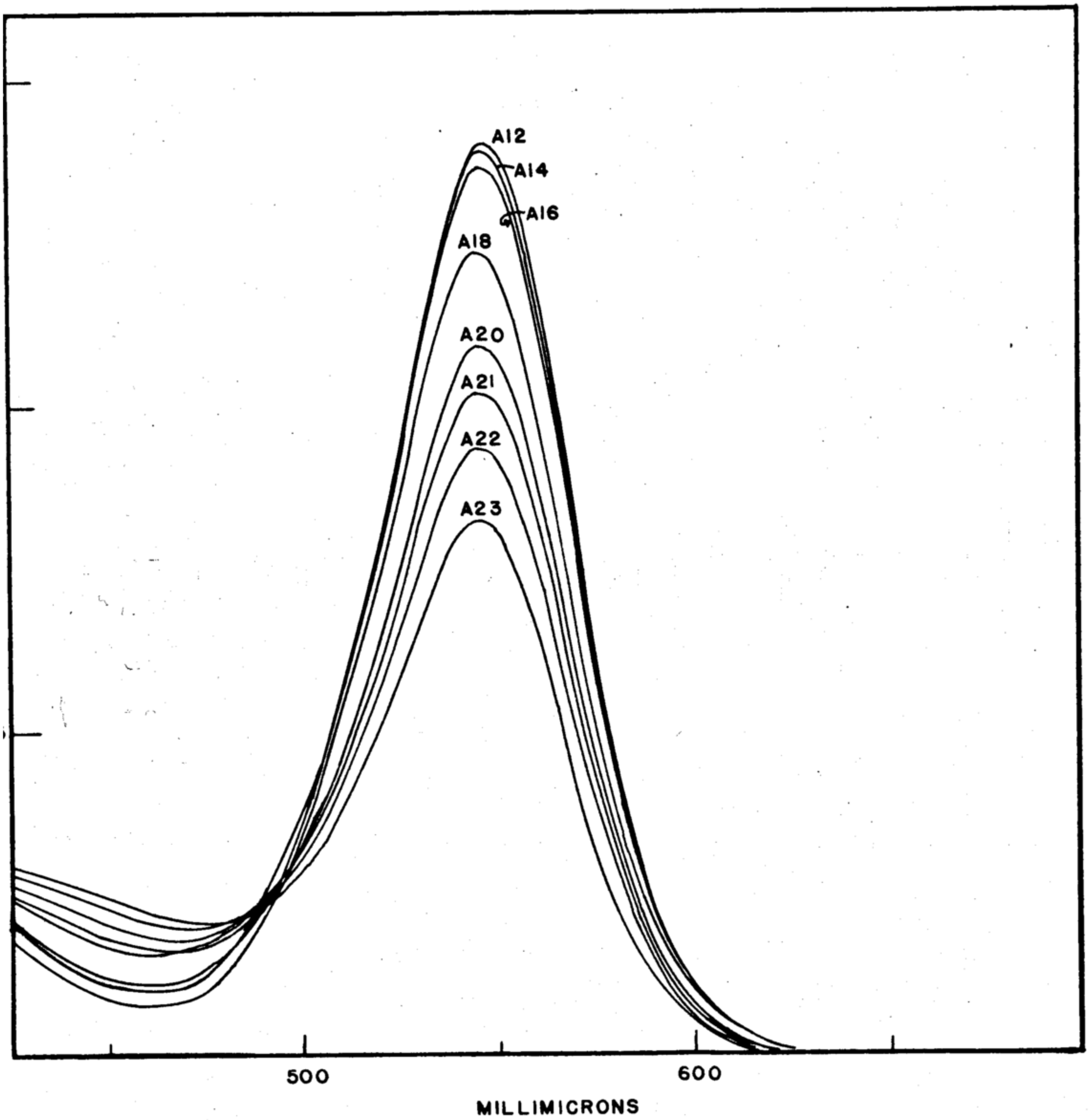


FIG. 19 ABSORPTION SPECTRA OF THYMOL BLUE IN THE BUFFERS OF HALL AND MEYER (37).

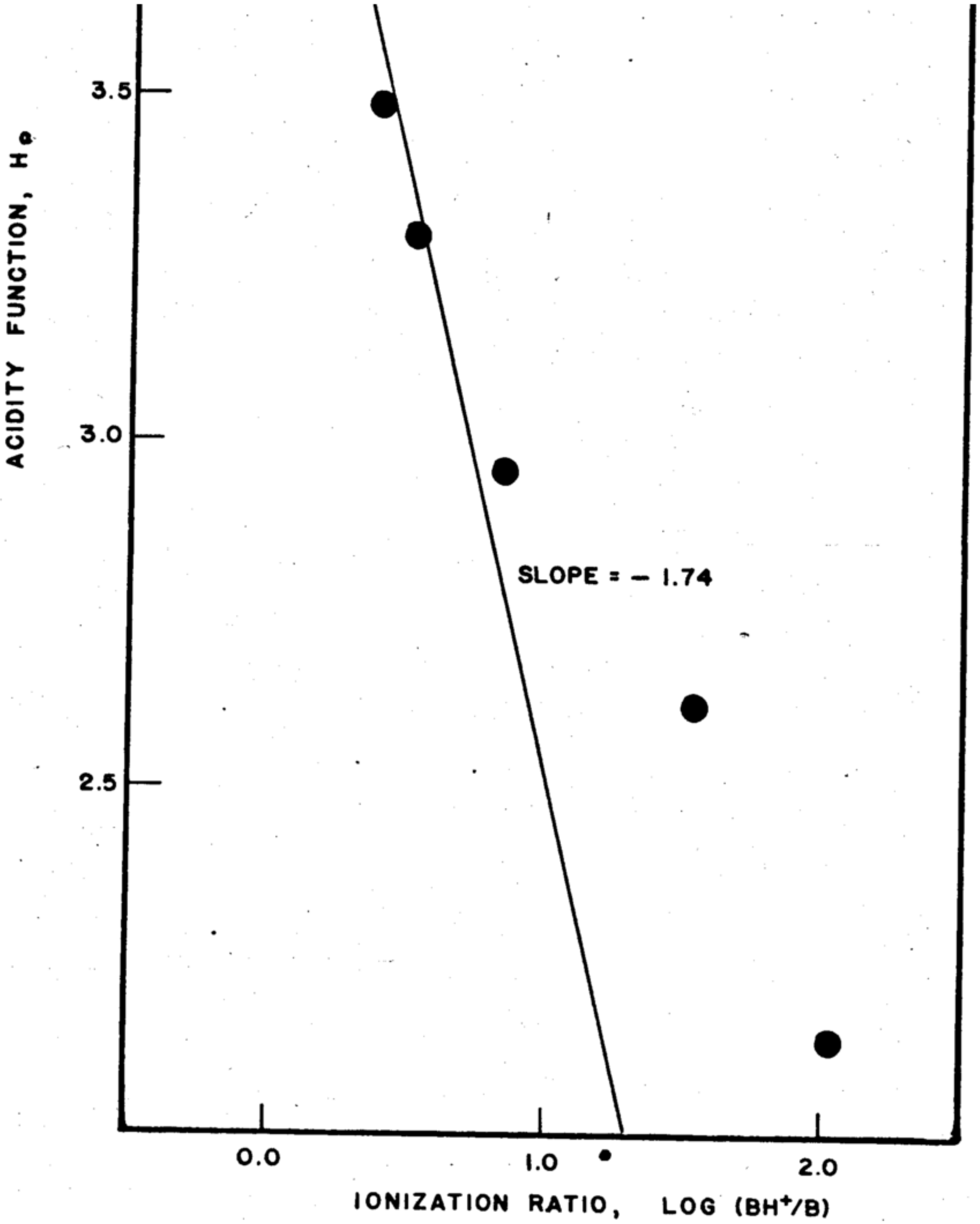


FIG. 20 THYMOL BLUE, $pK' = + 4.24$

Table XVI. Spectrophotometric Data for Thymol Blue.

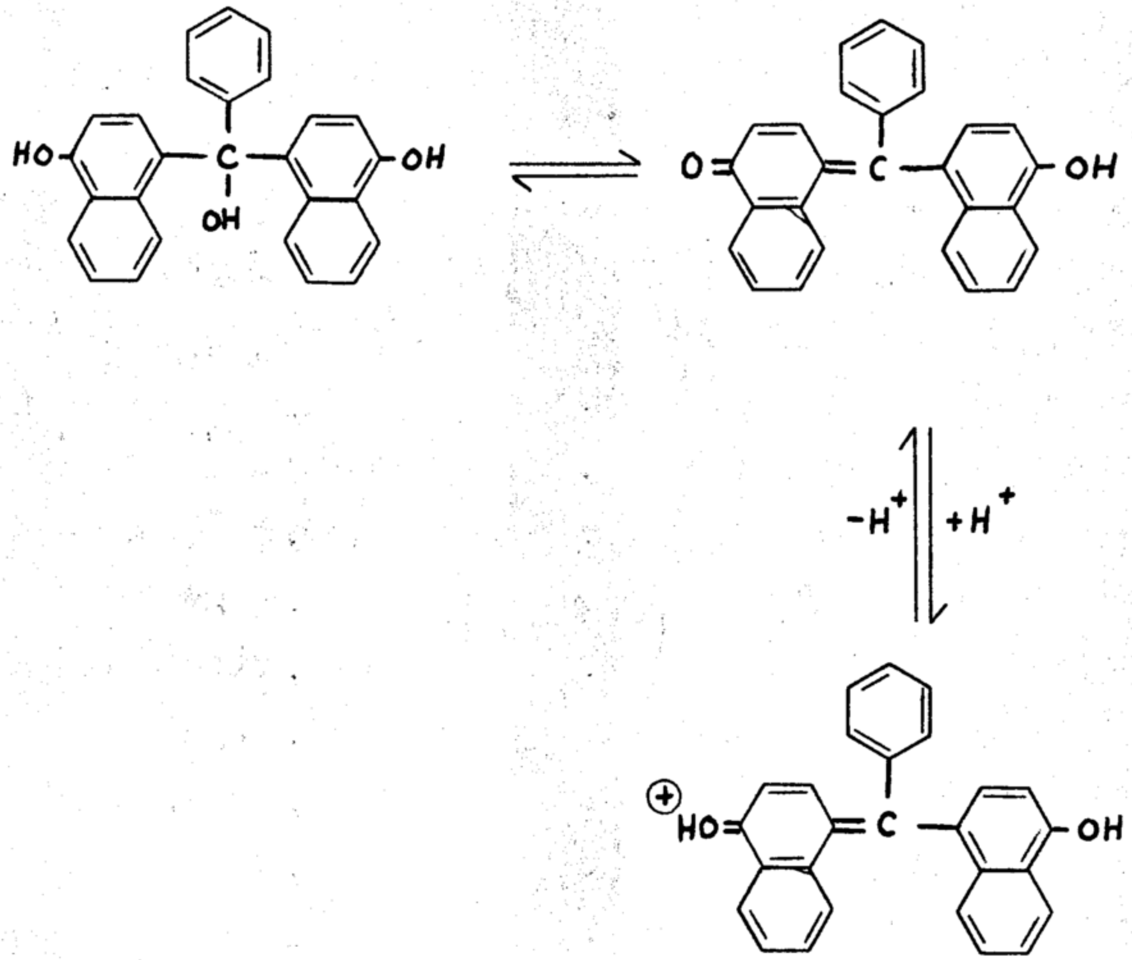
Solution	Acid Band 546 mμ O.D.	Log HI^+/B
$HClO_4$	1.432	-
A12	1.403	-
A14	1.390	2.01
A16	1.365	1.53
A18	1.235	0.84
A20	1.091	0.51
A21	1.019	0.39
A22	0.932	0.26
A23	0.822	-
C23	0.074	-

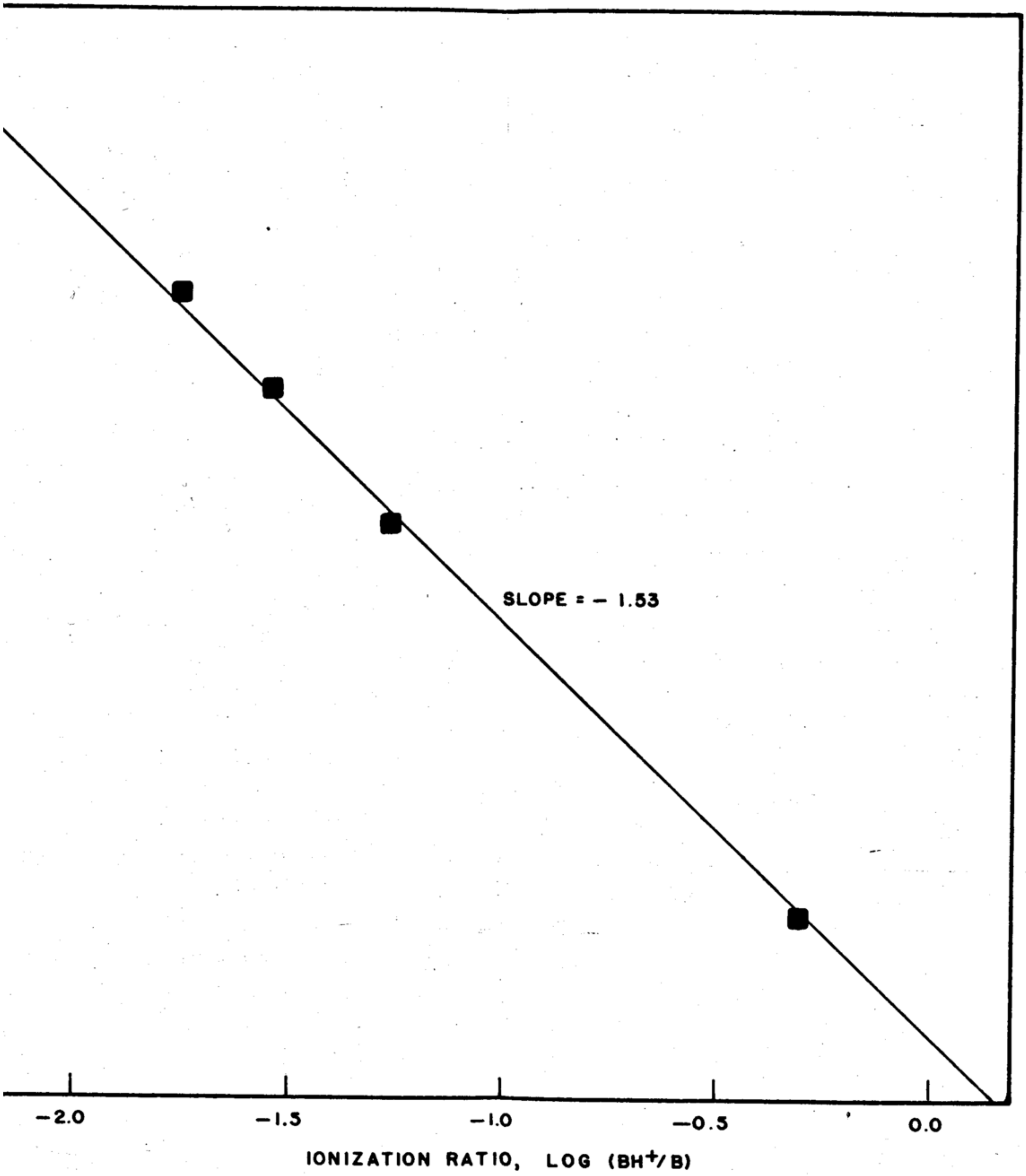
The absorption spectra of thymol blue in the buffers of Hall and Meyer (37) are shown in Figure 19. An examination shows that the curves of solutions A12, A14, and A16 lie fairly close together, hence any slight discrepancies in optical densities are magnified in values of the ionization ratio. For this reason, the absorption spectrum of solution A12 was taken as the acid form of the indicator and the values of the A14 and A16 solutions were neglected in the graphical evaluation of pK' (Figure 20).

Perchloric acid produces a "medium effect" in the absorption curve which is accompanied by a slight increase in band intensity.

Alpha-Naptholbenzein

Unlike the other indicators investigated, α-naphtholbenzein is a triarylcarbinol type dye. In aqueous solutions, two color changes are visible, one at pH values less than one, and the second over the pH range 8.2 to 10. In acetic acid, α-naptholbenzein behaves as a monoprotic indicator, exhibiting only one color change, being yellow in the neutral or alkaline form and green in the acidic form. The experimentally determined pK' value is 0.25, indicating that the color transition takes place in the moderately acidic regions of this solvent.





G. 21 α−NAPHTHOLBENZEIN, $_pK' = +\ 0.25$

The visible absorption spectrum of α-naptholbenzein is quite complex at wave lengths below 555 millimicrons where a well defined isosbestic point is present. At 624 millimicrons, there is a strong acid band which has been used in computing the ionization ratio for this indicator. The perchloric acid and A23 spectra were used as the acid and alkaline forms of the indicator; tricyclohexylamine solution not being necessary, as the indicator requires only a weakly basic solution to convert it entirely into its alkaline form. The spectrophotometric data are given in Table XVII, while in Figure 21, the graphical evaluation of pK' is shown.

Table XVII. Spectrophotometric Data for α-Naphtholbenzein.

Solution	Acid Band 624 mu O.D.	Log BH^+/B
$HClO_4$	1.540	-
A12	0.510	-0.32
A14	0.091	-1.27
A16	0.055	-1.54
A18	0.028	-1.76
A23	0.012	-

PART III: The Investigation of Indicators for Titration Procedures.

In order to evaluate various indicators in acetic acid as replacements for potentiometric procedures, solutions of perchloric acid were titrated with a tricyclohexylamine solution. These titrations were carried out potentiometrically in the presence of an indicator, thereby making possible a comparison of the potentiometric and colorimetric endpoints. In Figures 22 through 33, the color changes taking place along the potentiometric titration curve are shown, and from these comparisons, the suitability of each indicator was ascertained. In Table XVIII, the changes in indicator color at the endpoint have been classified as sharp, faint, or no transition, along with the structural type of each indicator.

Table XVIII. Color Transitions of Nonaqueous Indicators.

A. Sharp Transition

Indicator	Type
Quinaldine Red	Basic: tertiary amine and ethylene bridge
Orange IV	Basic: azo, salt
p-Phenylaminoazobenzene	Basic: azo
Benzopurpurin 4B	Basic: azo, salt
Malachite Green Oxalate	Basic: salt
Metanil Yellow	Basic: azo, salt
Brilliant Yellow	Basic: azo, salt
α-Naphtholbenzein	Triarylcarbinol
Formyl Violet S4B	Basic

B. Faint Transition

Indicator	Type
p-Dimethylaminoazobenzene	Basic azo
Propyl Red	Basic azo
m-Cresol Purple	Acidic

C. No Transition (gradual color fading)

Indicator	Type
Thymol Blue	Sulfonphthalein
p-Sulfo-o-Methoxybenzeneazo-Dimethyl-α-Naphthylamine (Eastman Kodak 1954)	Basic azo
Naphtol Green B	

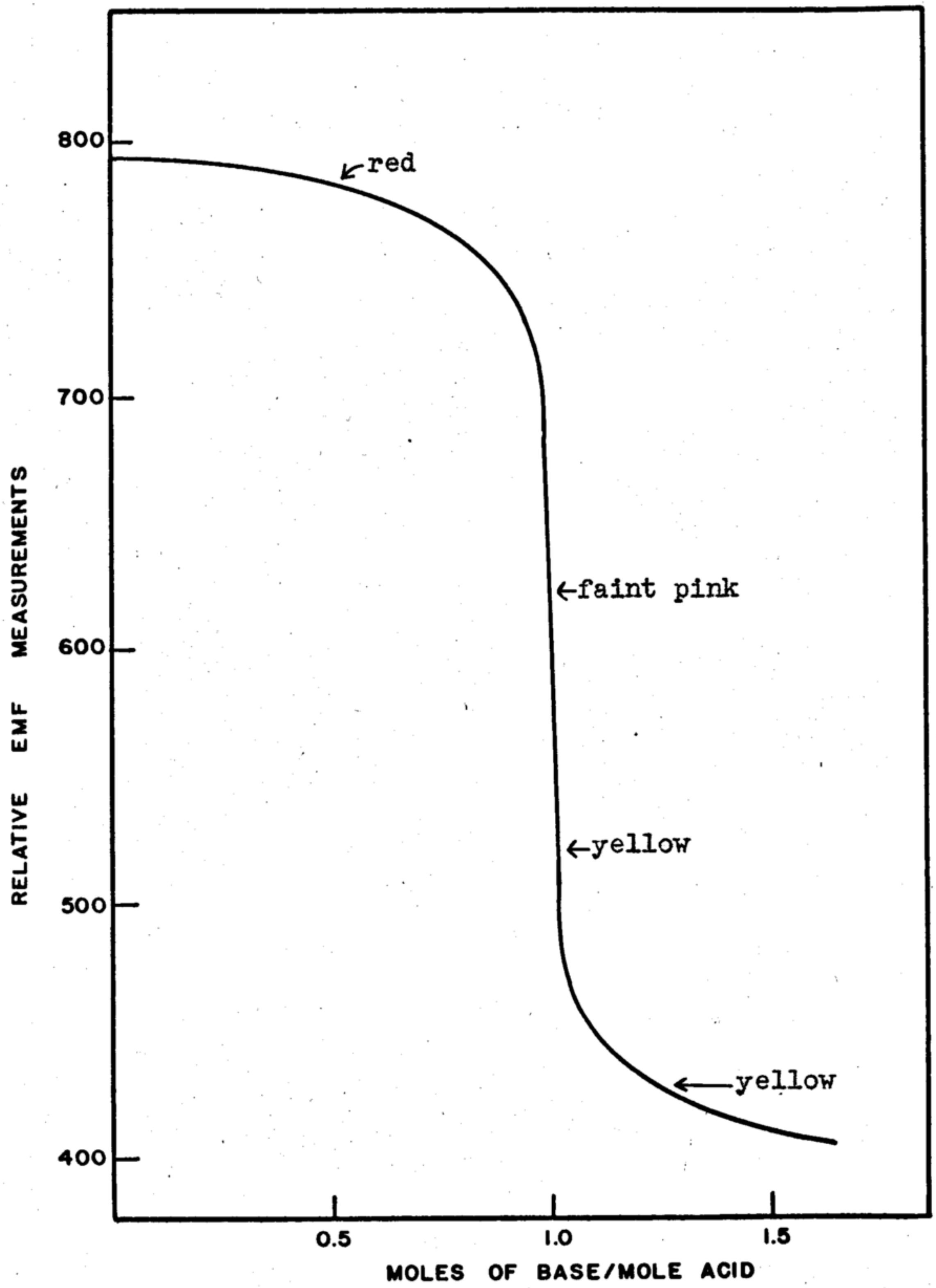


FIGURE 22. POTENTIOMETRIC EVALUATION OF ORANGE IV AS AN INDICATOR IN ACETIC ACID.

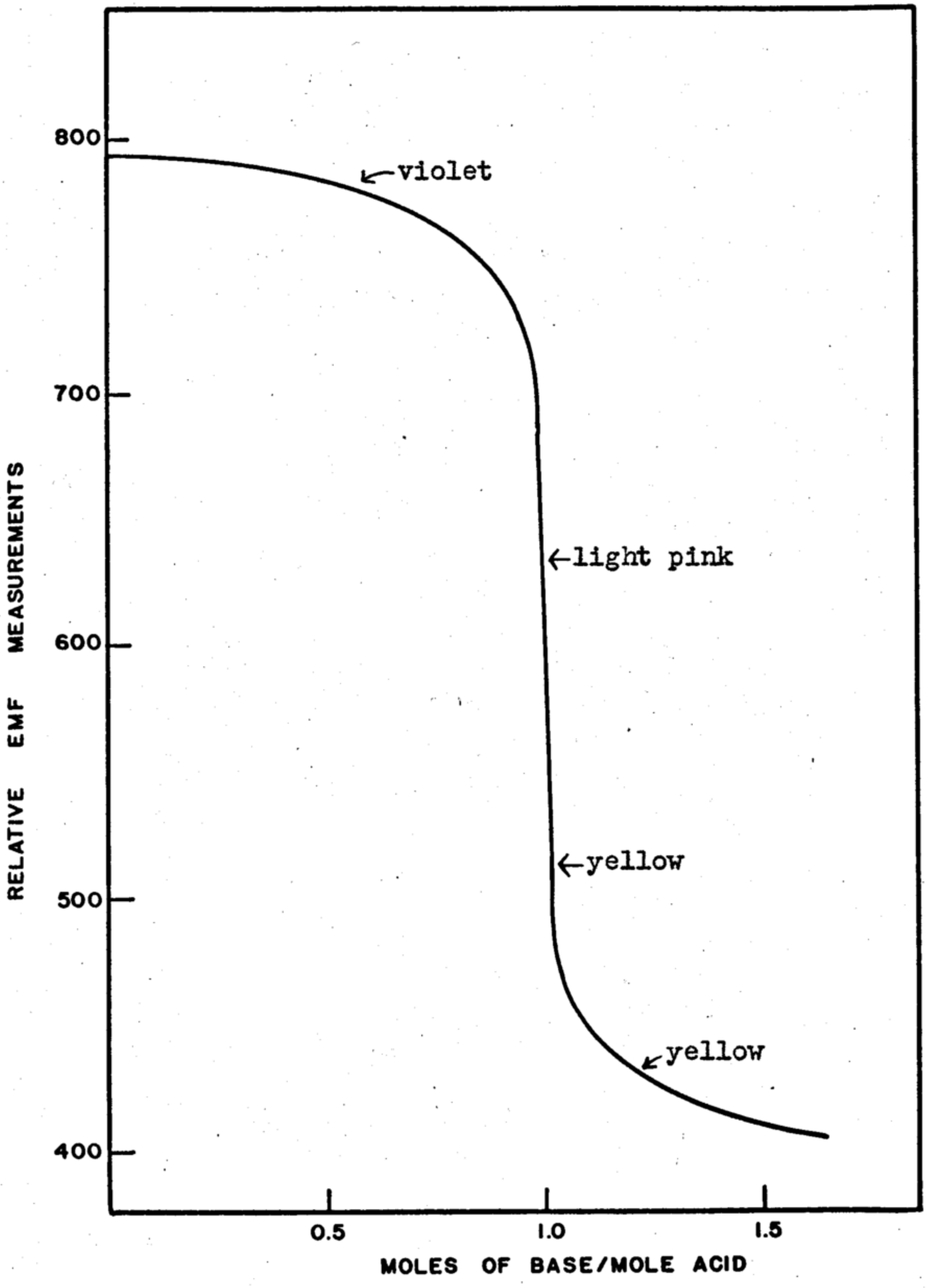


FIGURE 23. POTENTIOMETRIC EVALUATION OF p-PHENYLAMINOAZOBENZENE AS AN INDICATOR IN ACETIC ACID.

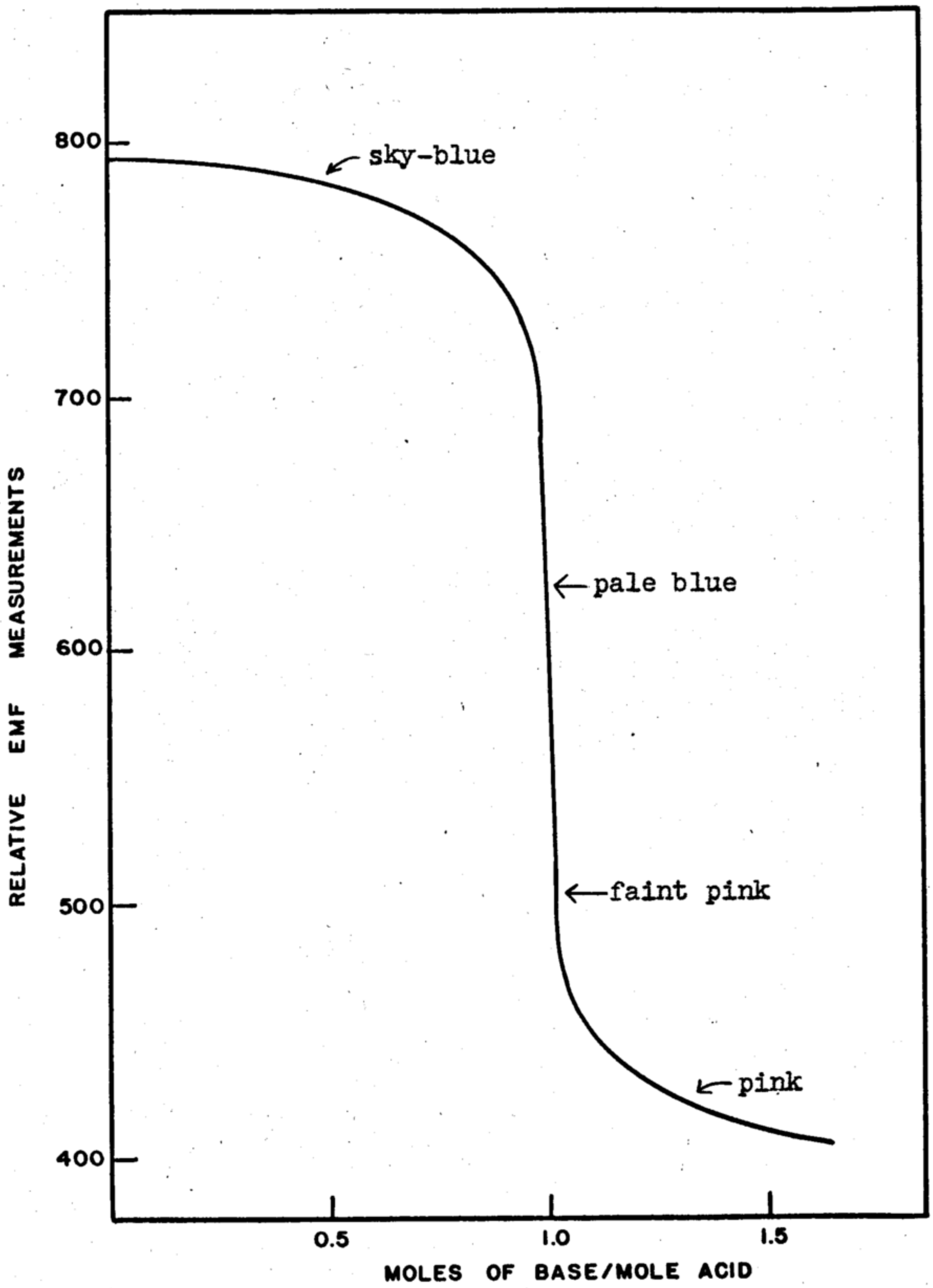


FIGURE 24. POTENTIOMETRIC EVALUATION OF BENZOPURPURIN 4B AS AN INDICATOR IN ACETIC ACID.

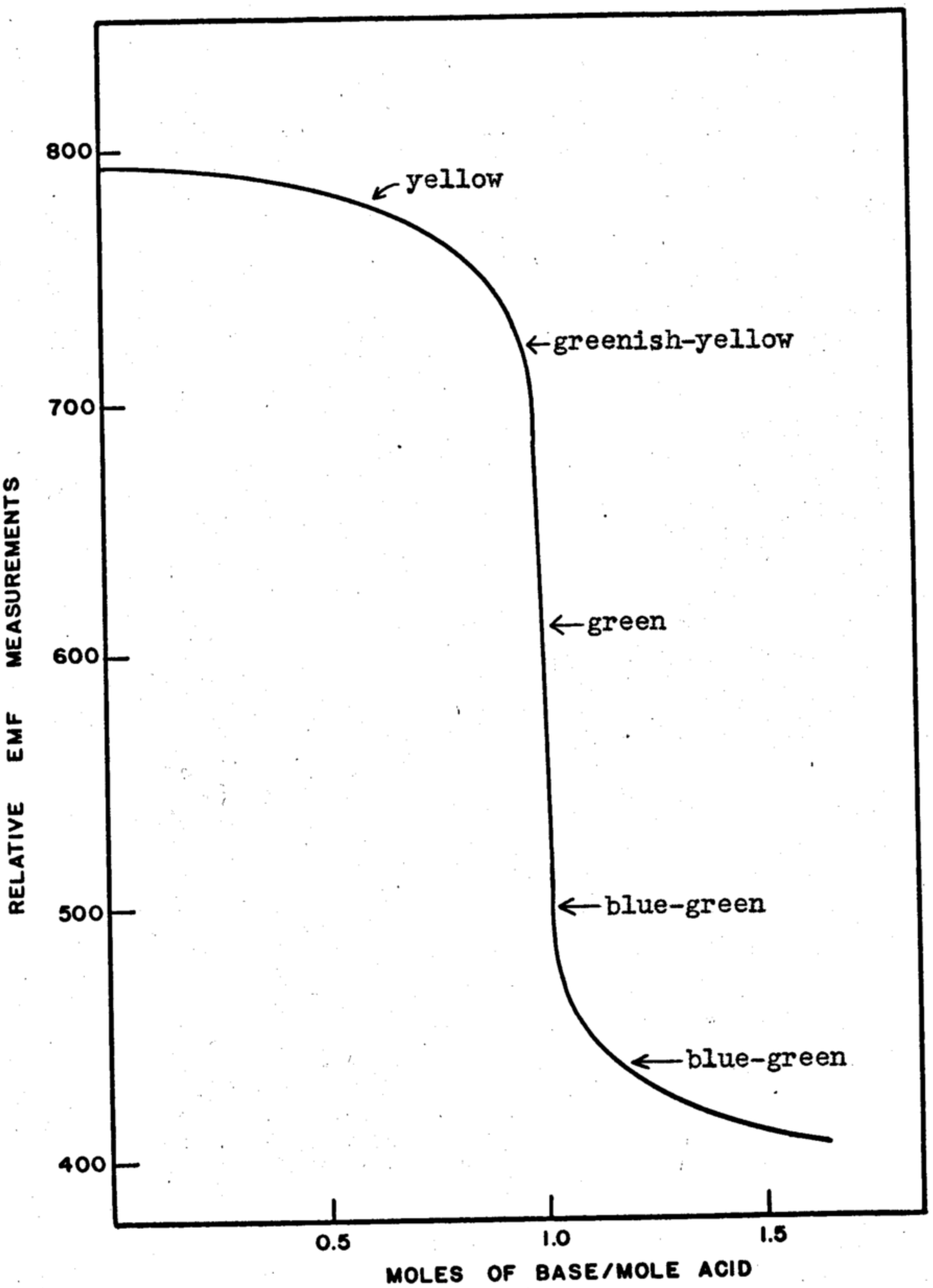


FIGURE 25. POTENTIOMETRIC EVALUATION OF MALACHITE GREEN OXALATE AS AN INDICATOR IN ACETIC ACID.

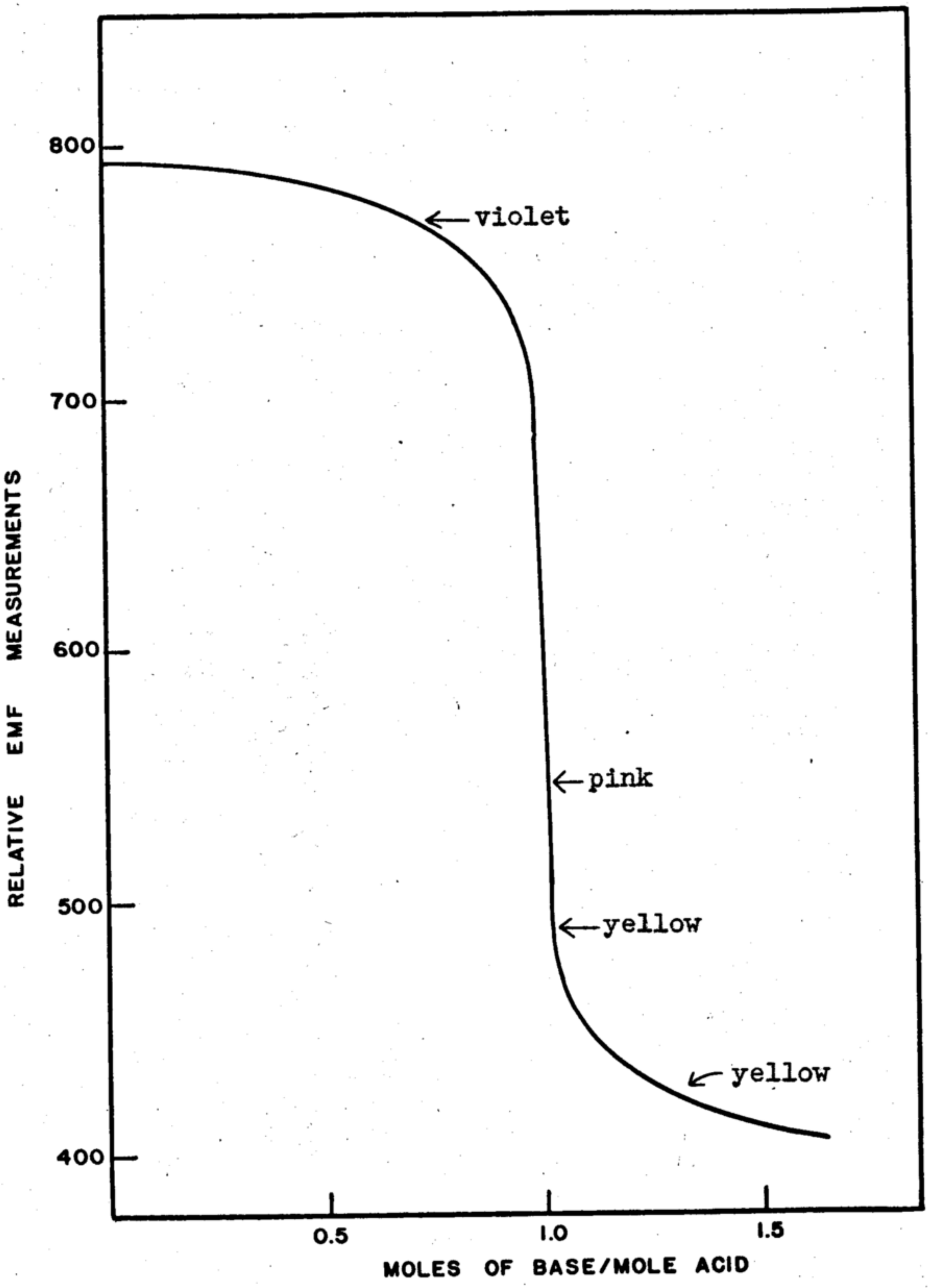


FIGURE 26. POTENTIOMETRIC EVALUATION OF METANIL YELLOW AS AN INDICATOR IN ACETIC ACID.

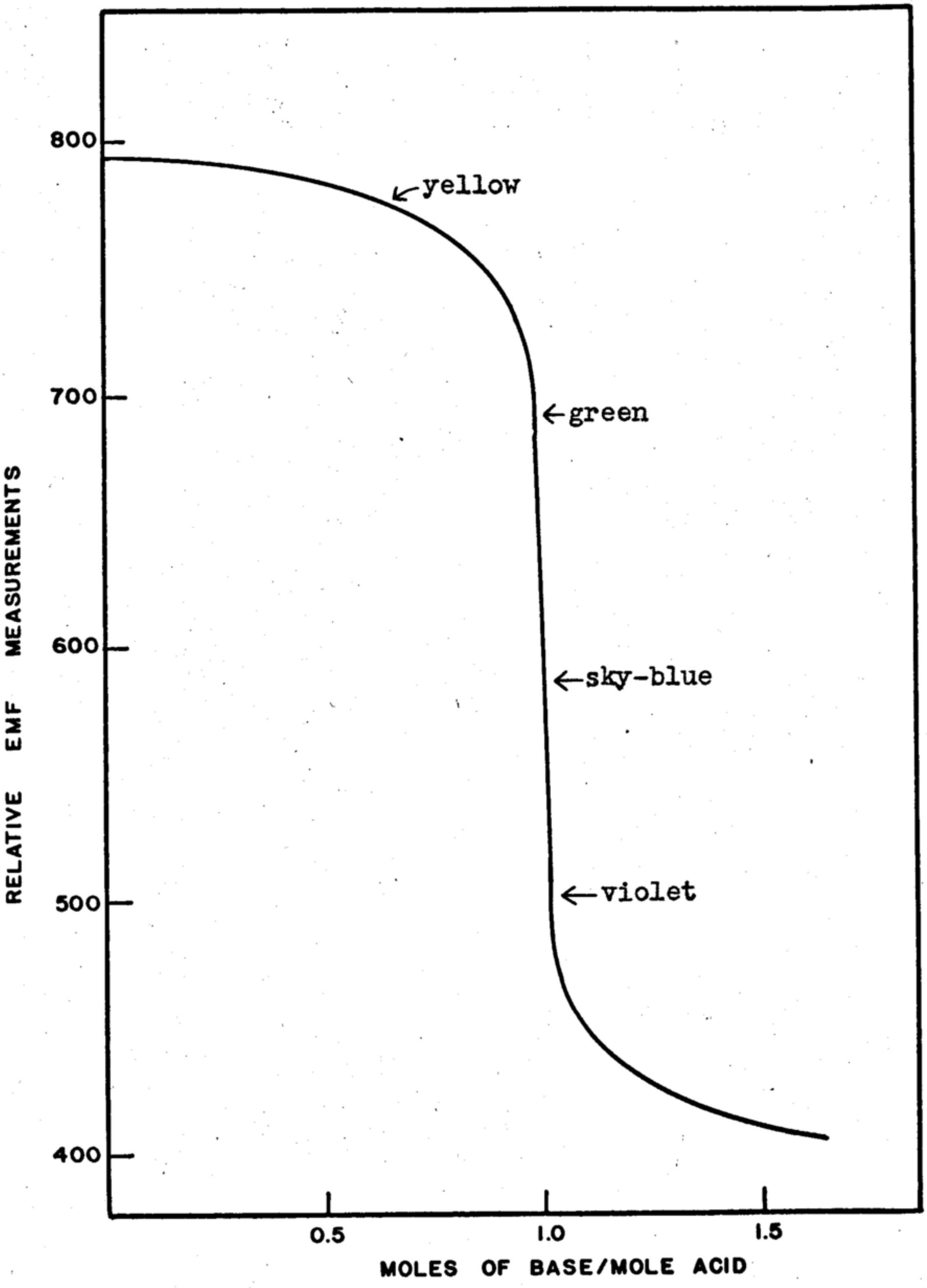


FIGURE 27. POTENTIOMETRIC EVALUATION OF FORMYL VIOLET S4B AS AN INDICATOR IN ACETIC ACID.

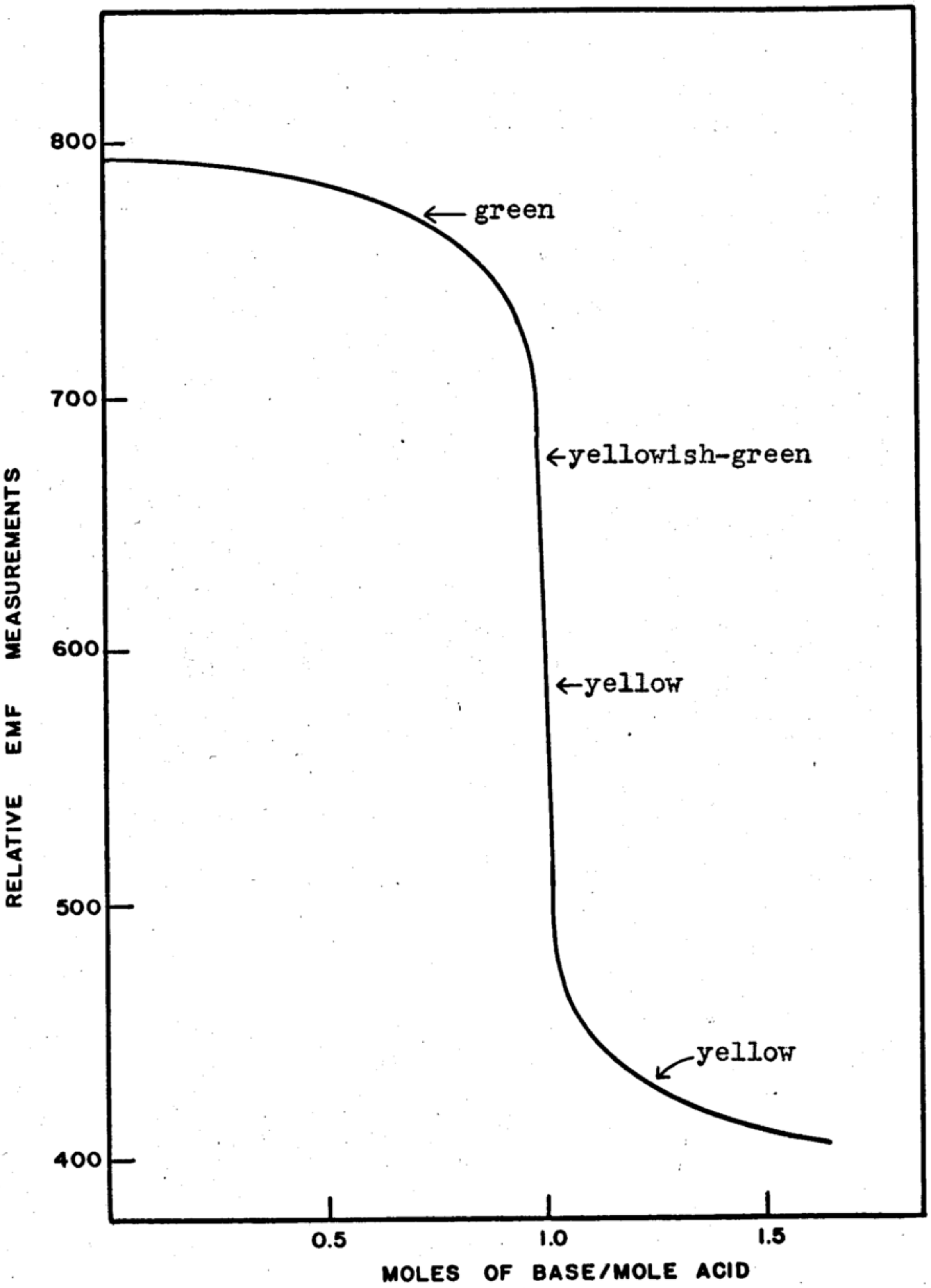


FIGURE 28. POTENTIOMETRIC EVALUATION OF α-NAPHTHOLBENZEIN AS AN INDICATOR IN ACETIC ACID.

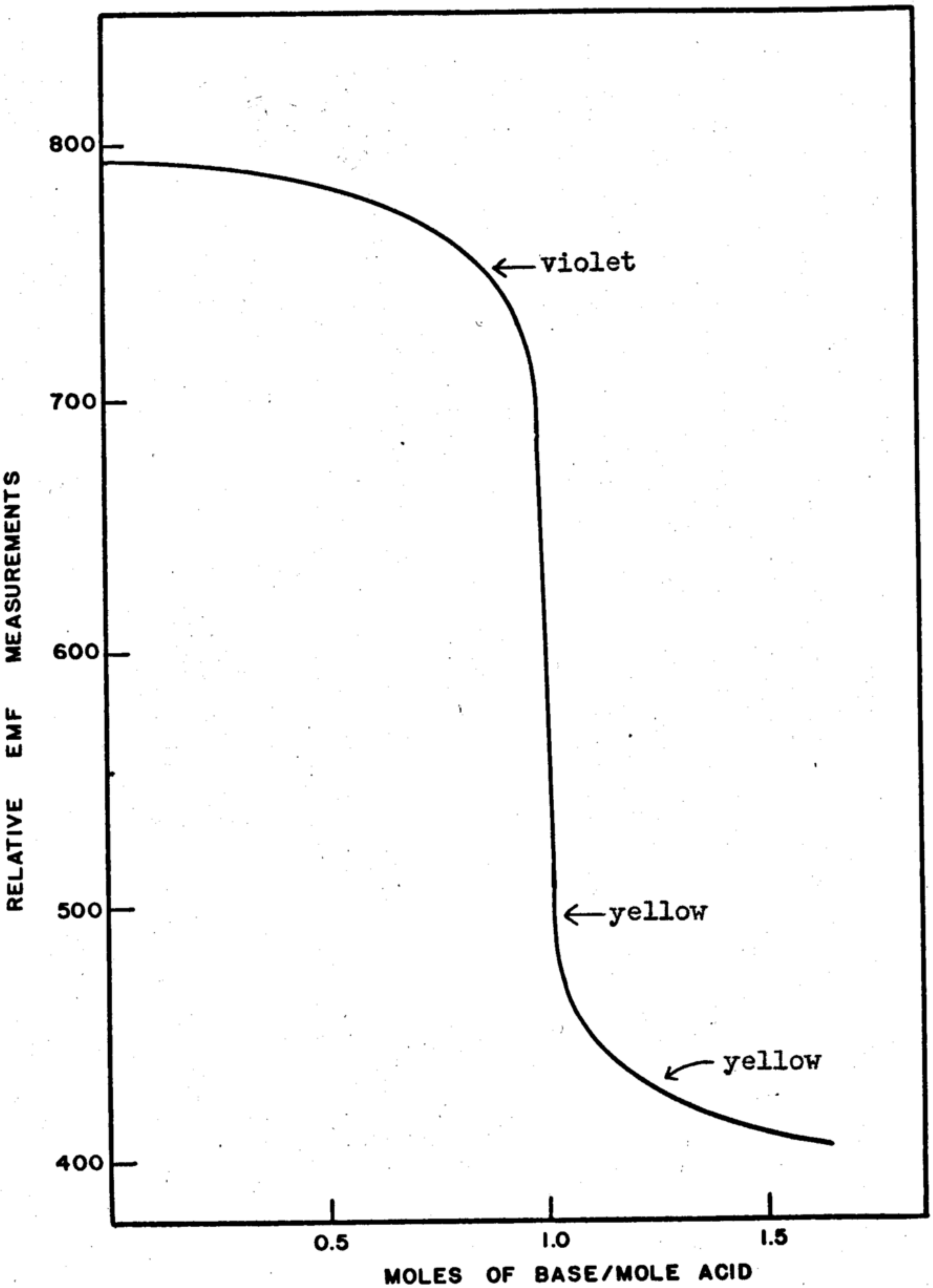


FIGURE 29. POTENTIOMETRIC EVALUATION OF BRILLIANT YELLOW AS AN INDICATOR IN ACETIC ACID.

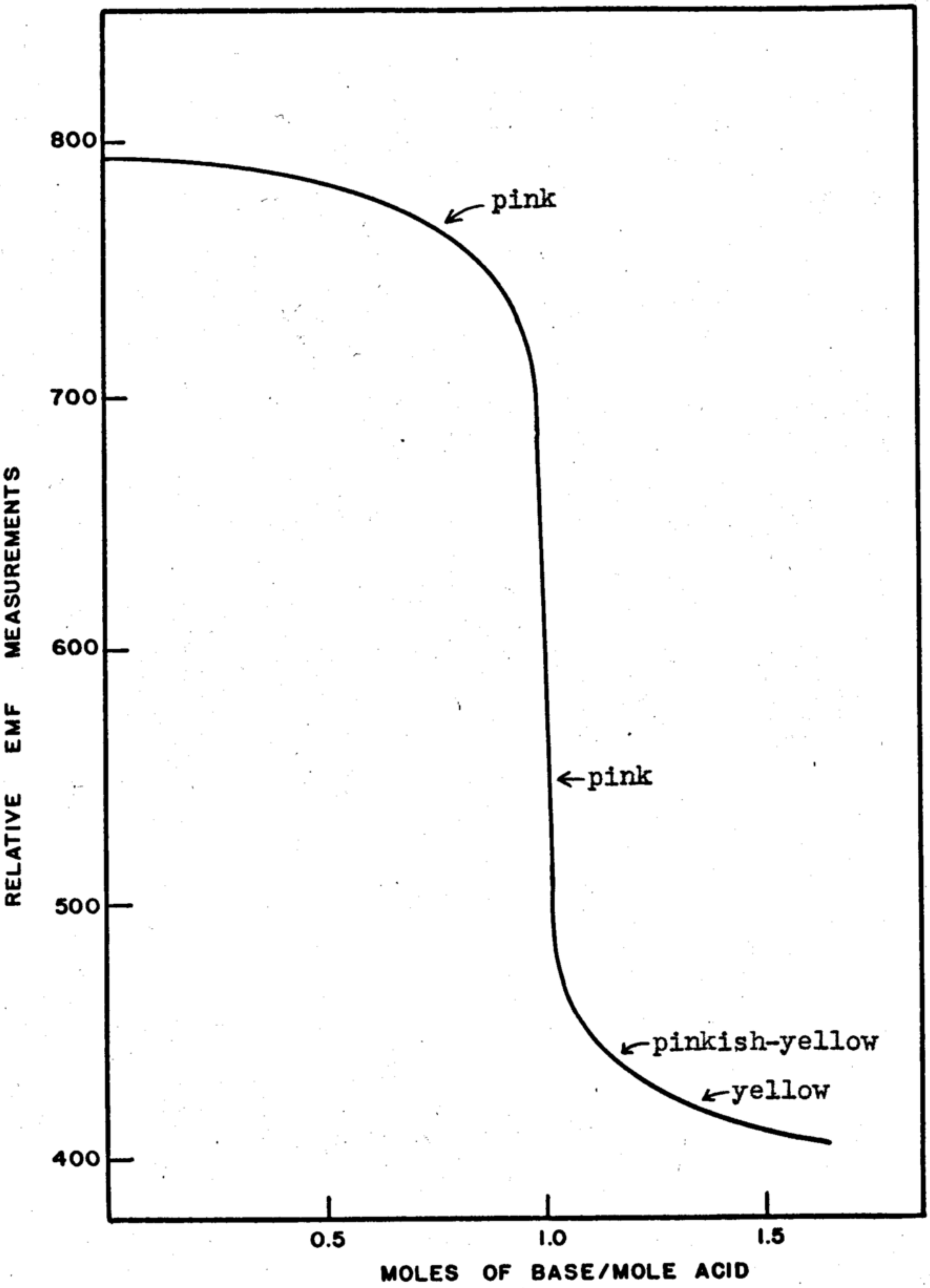


FIGURE 30. POTENTIOMETRIC EVALUATION OF "BUTTER YELLOW" AS AN INDICATOR IN ACETIC ACID.

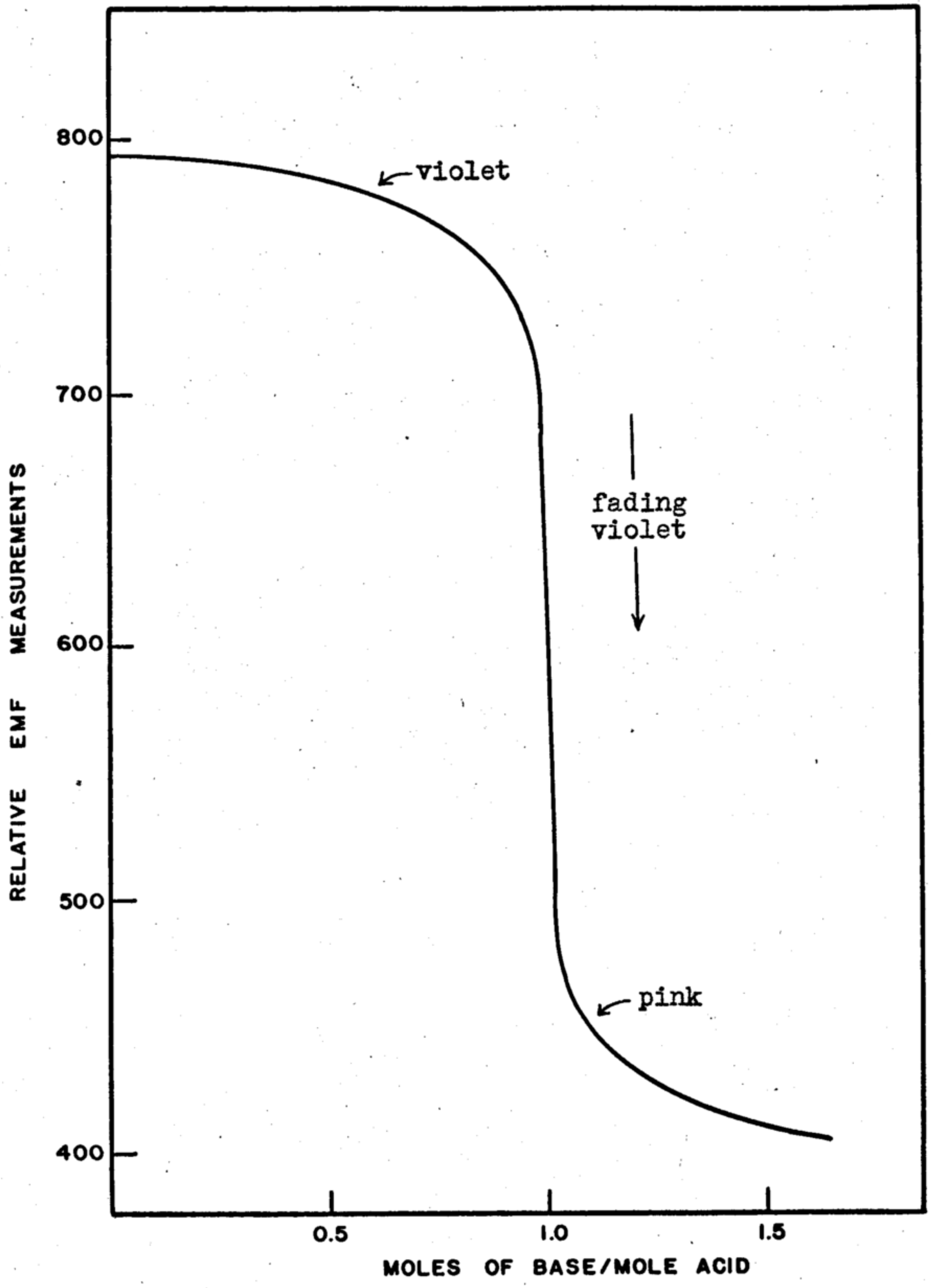


FIGURE 31. POTENTIOMETRIC EVALUATION OF PROPYL RED AS AN INDICATOR IN ACETIC ACID.

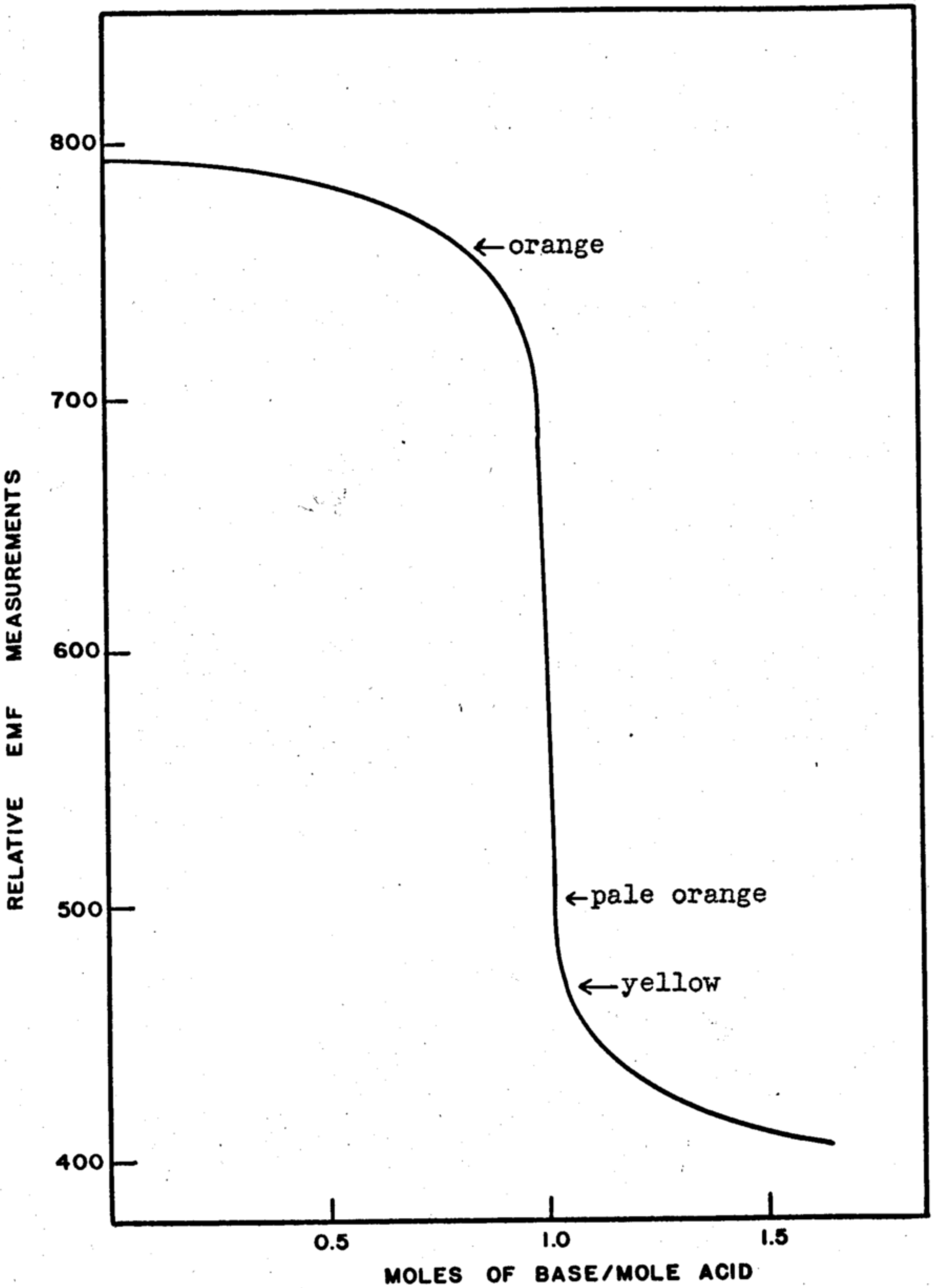


FIGURE 32. POTENTIOMETRIC EVALUATION OF m-CRESOL PURPLE AS AN INDICATOR IN ACETIC ACID.

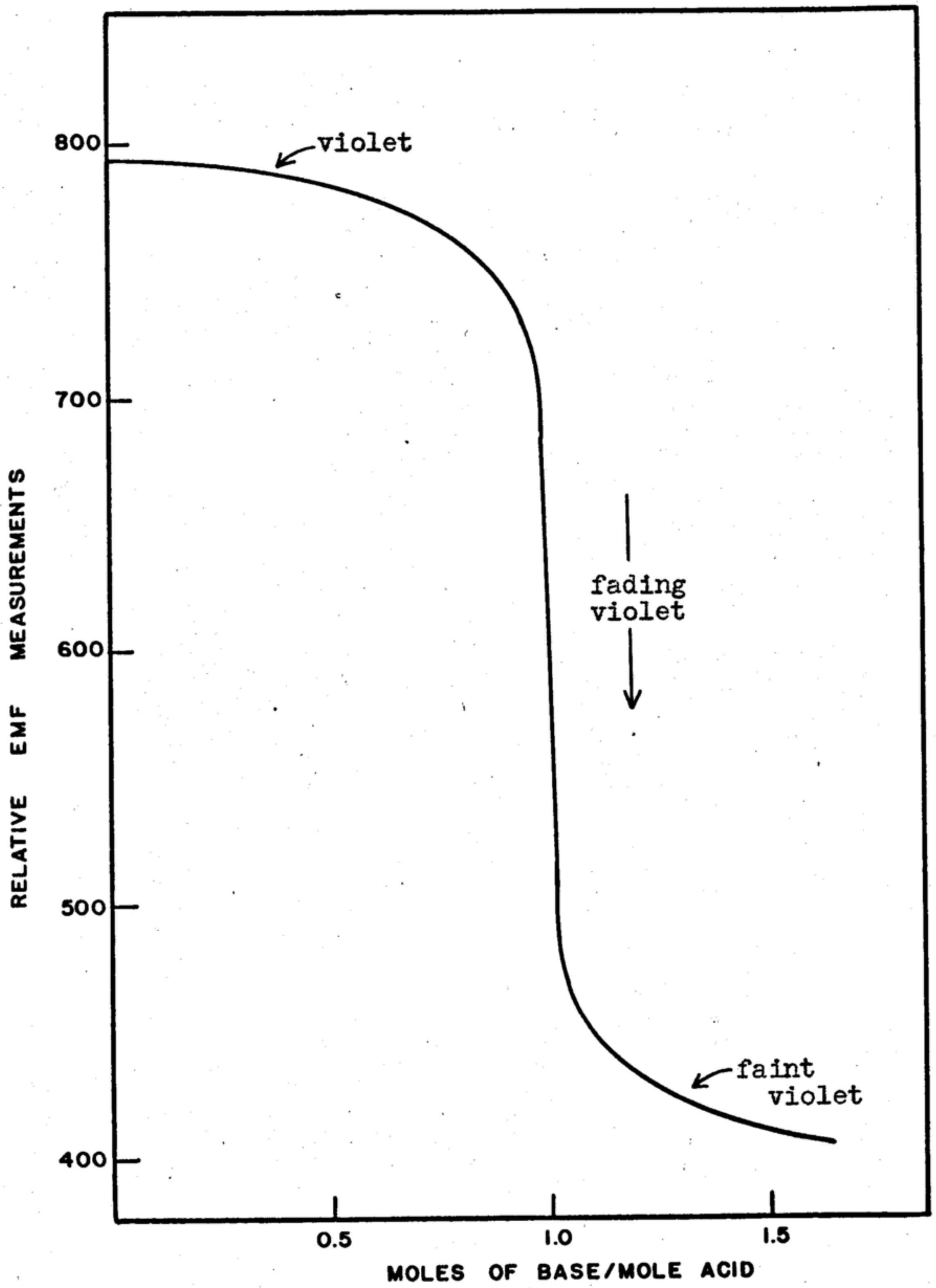


FIGURE 33. POTENTIOMETRIC EVALUATION OF THYMOL BLUE AS AN INDICATOR IN ACETIC ACID.

In the titration of sulfuric acid with sodium acetate and tricyclohexylamine solutions (Figure 34), the potentiometric "break" obtained with the latter is much sharper showing its enhanced value for titration procedures. These titration curves are somewhat analogous to the titration of a moderately strong acid with a weak and strong base, respectively, in aqueous solutions. The curves shown in Figure 34 have been shifted vertically so that the half-neutralization points coincide.

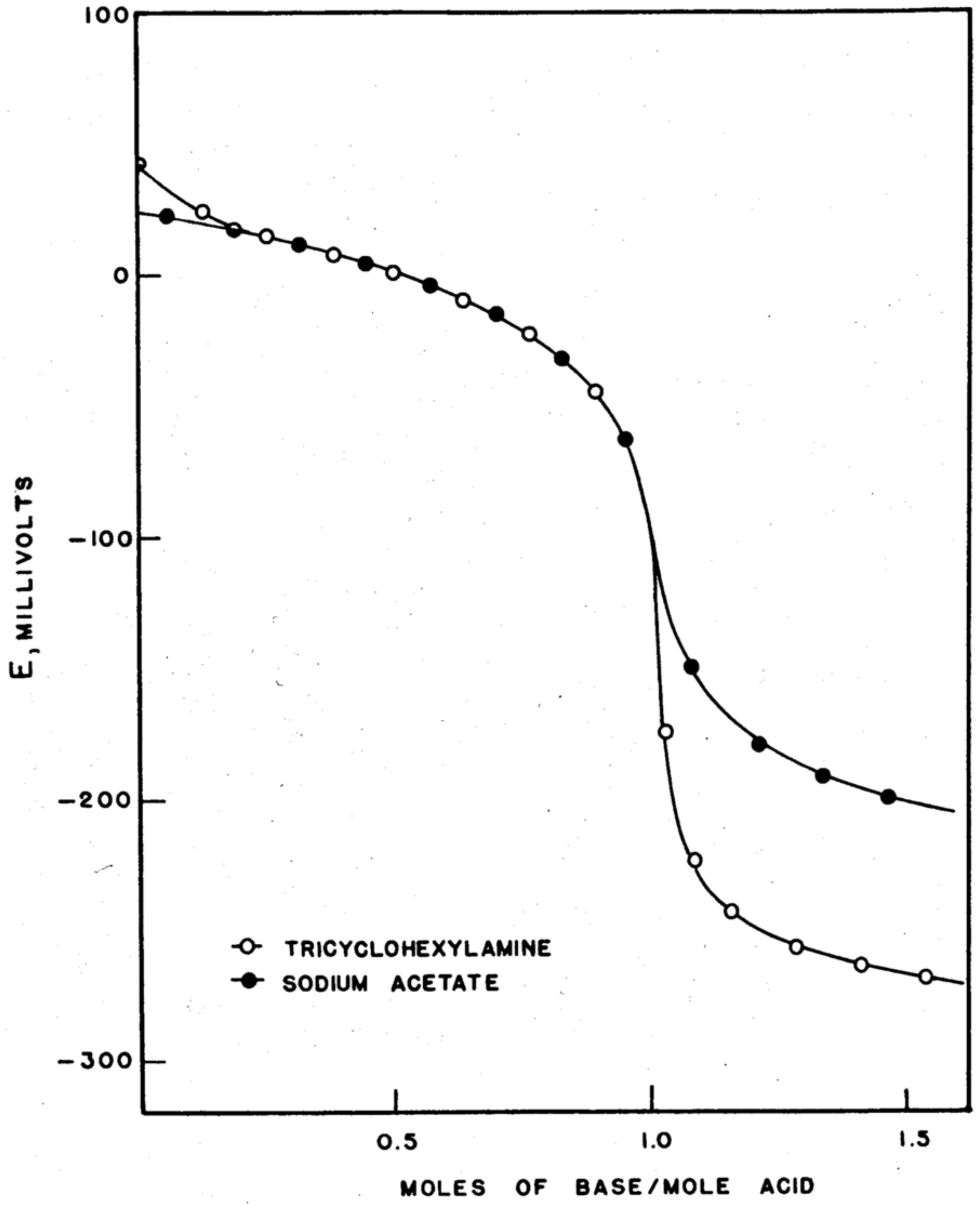


FIG. 34 THE POTENTIOMETRIC TITRATION OF SULFURIC ACID WITH VARIOUS BASES IN ACETIC ACID.

CONCLUSIONS

The interaction between an organic amine and acetic acid results in nearly complete formation of the ammonium salt which remains predominantly in the form of undissociated "ion-pairs" due to the low dielectric constant of the solvent. As the development of relative basicity is dependent upon the extent to which these ammonium acetate molecules dissociate, particular attention has been paid to the influence of steric factors in promoting "ion-pair" separation. On the basis of experimental work performed, and results obtained, the following conclusions have been drawn.

The effects of variation in chain length and arrangement have shown that spacial configurations about the center of positive charge play a predominant part in the extent of "ion-pair" dissociation. In acetic acid, it has been found that the order of basicity of alkyl amines decreases from the tertiary to the secondary to the primary amine. This differs considerably from aqueous solutions, where the secondary amine is the strongest, while the order of strengths of the primary and tertiary amines are dependent upon the individual chain lengths.

In CH_3COOH: $(n\text{-}C_4H_9)_3N > (iso\text{-}C_4H_9)_2NH > (n\text{-}C_4H_9)_2NH >$
$n\text{-}C_4H_9NH_2$

In Water: $(CH_3)_2NH > CH_3NH_2 > (CH_3)_3N$
$(C_2H_5)_2NH > C_2H_5NH_2 = (C_2H_5)_3N$
$(n\text{-}C_3H_7)_2NH > (n\text{-}C_3H_7)_3N > n\text{-}C_3H_7NH_2$

A comparison of the relative basicities for a homologous series of tertiary amines in acetic acid and in water shows a similar pattern to be present in both solvents. In acetic acid and water, triethylammonium acetate undergoes the greatest extent of dissociation. This is in agreement with the data of Brown and Taylor (51), these workers having shown that the steric requirements of the ethyl group are quite large as compared to other alkyl groups.

In CH_3COOH: $(C_2H_5)_3N > (n\text{-}C_3H_7)_3N > (n\text{-}C_4H_9)_3N > (n\text{-}C_6H_{13})_3N >$
$(n\text{-}C_7H_{15})_3N > (n\text{-}C_8H_{17})_3N > (CH_3)_3N > NH_3$

In Water: $(C_2H_5)_3N > (n\text{-}C_3H_7)_3N > (n\text{-}C_4H_9)_3N > (CH_3)_3N > NH_3$

It has been found that an isopropyl type configuration, as is present in tricyclohexylamine, adjacent to the nitrogen atom of a tertiary amine provides a large amount of steric hindrance. This spongy, symmetrical type distribution of atoms about the nitrogen limits the approach of the acetate ion, thereby enhancing "ion-pair" dissociation.

Of all the bases investigated, tetrabutylammonium acetate undergoes the greatest extent of "ion-pair" dissociation in acetic acid. In considering the steric factors involved in this type of structure, it seems highly likely that the tetraalkylammonium acetates may represent the limiting strength of bases in acetic acid. The steric factors which shield the nitrogen atom attain their maximum in a quaternary salt.

It can be concluded from this investigation of steric factors, that the order of strengths of any given group of bases in water is not a good guide to their strengths in acetic acid or other nonaqueous solvents. Water and other similar amphiprotic solvents exert a "leveling effect" on bases in the same manner in which they do on acids (53).

The spectrophotometric evaluation of indicators in acetic acid solutions of known acidity has provided seven indicator constants in terms of the Hammett Acidity Function, H_o (34). The basic indicators quinaldine red, orange IV, p-phenylaminoazobenzene, and benzopurpurin 4B, and the triarylcarbinol α-naphtholbenzein exhibit sharply defined color changes in acetic acid. Of these five indicators, the first four have pK' values in the relatively alkaline regions of the solvent, whereas α-naphtholbenzein undergoes its color change in the moderately acidic regions.

The indicators "butter yellow" and thymol blue have pK' values which extend far into the alkaline regions of acetic acid, however the color changes exhibited are gradual, rather than sharp. Both of these indicators appear to accept two protons prior to the occurrence of a detectable color change.

In general, azo type indicators containing an electrophilic group in their structure exhibit the sharpest color changes in acetic acid. The color changes which occur with acidic indicators, and with azo indicators containing no negative substituents are rather gradual, hence their value in titrimetry is limited.

SUMMARY

The application of nonaqueous titrations in acetic acid is undergoing extensive application in the area of pharmaceutical analysis. Because of the complexity of behavior of various solutes in the acetic acid system, an attempt has been made to clarify some of the obscure relationships involved so as to achieve a clearer understanding of acid-base equilibria in this solvent.

Although a considerable amount of work has been done in the relatively acidic regions of this solvent, very little attention has been paid to the use of the more alkaline regions for analytical purposes. In the present investigation, emphasis has been placed on extending the relatively more alkaline regions of acetic acid by the use of bases stronger than sodium acetate, which is the one most commonly used as a titrant. Along with the development of these stronger bases, a survey was made to uncover suitable indicators for use in the more alkaline regions of the solvent. It was initially desired to locate an effectively totally dissociated base in acetic acid; this however proved unsuccessful, as even the strongest bases follow the Ostwald Dilution law. After extensive investigation, it was evident that steric factors play a prominent role in limiting the relative alkalinity of the solution.

The present study into the more alkaline regions of acetic acid has been carried out by means of potentiometric and colorimetric (indicator) procedures. Unfortunately, potentiometric deviations at the higher solute concentrations raised some doubt as to the validity of measurements at concentrations greater and 0.05 molar.

By means of a potentiometric dilution procedure, amines of differing structural types, i.e. n-alkyl, cyclic, and aromatic, were studied for their relative dissociation behaviors in acetic acid. A careful investigation of the various structural type tertiary amines showed a critical size and shape necessary to obtain the maximum interionic distance. In Figure 1, the variations in "ion-pair" dissociation of the organic amines with changes in chain length and configuration are shown. The dissociation of ammonium acetate has arbitrarily been set as unity, all other dissociations being relative to that of ammonium acetate.

The fact that the maximum degree of "ion-pair" dissociation was obtained with a tetraalkylammonium acetate tends to indicate that these bases, as expected, represent the limiting strengths in acetic acid. This is because the positive charge is deeply buried among the four alkyl groups, thereby limiting the distance of acetate ion approach.

Among the mono-, di-, and trisubstituted ammonia derivatives, the order of basicity in acetic acid differs considerably from that in water. In acetic acid, the order of basicity is tertiary secondary primary; whereas in water, the secondary amine is the strongest, while the order of strengths of the primary and tertiary amines are dependent upon the individual chain lengths.

Spectrophotometric procedures were employed to evaluate dissociation constants (pK') of various acid-base indicators in acetic acid. As the ordinary pH scale is valid only for dilute aqueous solutions, the Hammett Acidity Function (34), H_o, was employed to establish the acidity values of the different acetic acid solutions. The acidity function, as defined in equation (19), makes use of basic indicators (B) as a measure of total

$$H_o = pK' - \log (BH^+/B) \qquad (19)$$

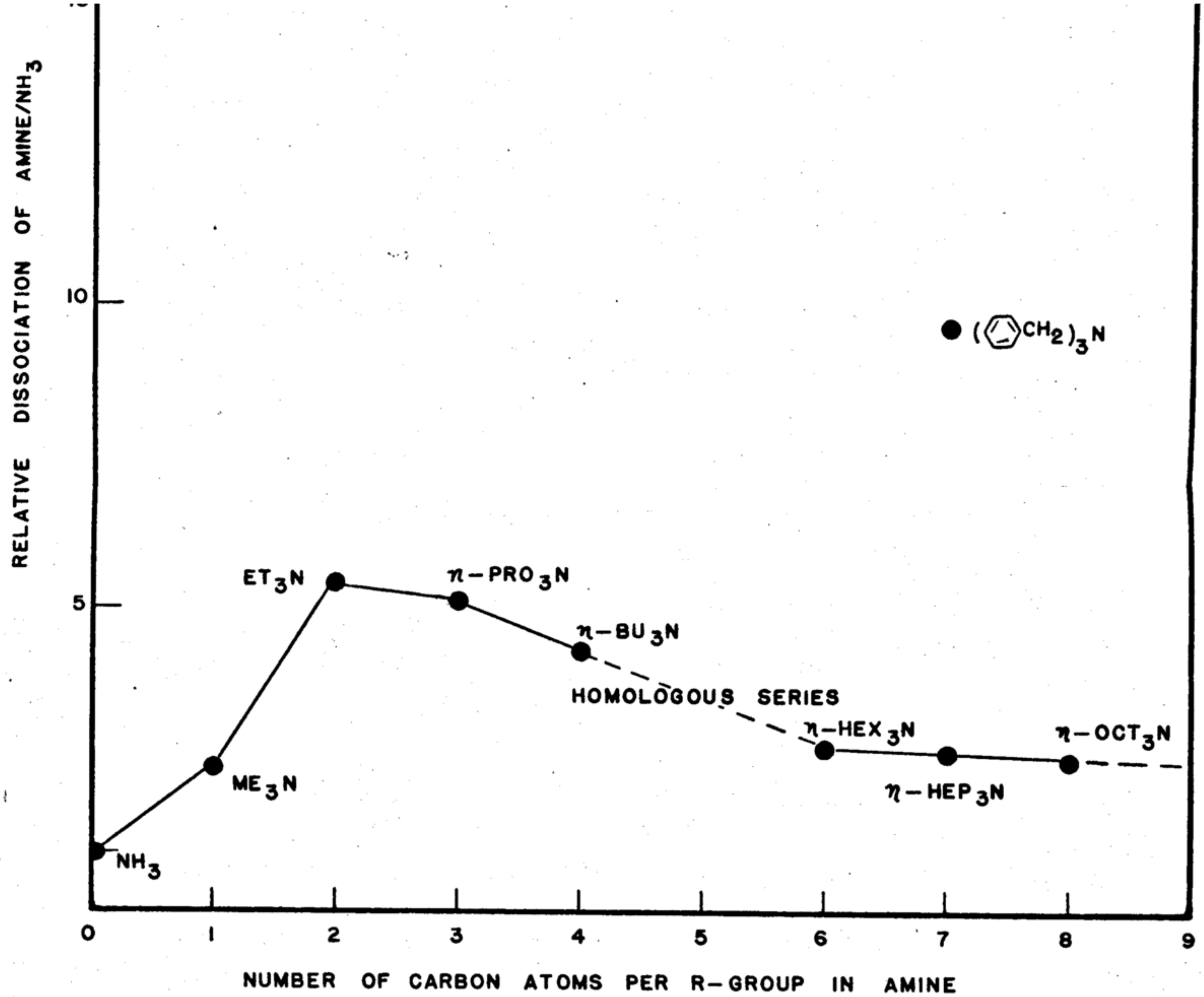


FIG. 1 VARIATIONS IN "ION-PAIR" DISSOCIATION OF ORGANIC AMINES IN ACETIC ACID WITH CHANGES IN CHAIN LENGTH AND CONFIGURATION.

acidity in acidic solvents, relative to the acidity of dilute aqueous solutions. As originally defined, the H_0 function is valid only for solvents of high dielectric constant, however Hall and Spengeman (36) have shown that in dilute solutions, there is a useful correlation between potentiometric and H_0 values in acetic acid.

The constants for the indicators listed below were experimentally determined in acetic acid using solutions of known H_0 value.

Indicator	pK'
Thymol blue	4.24
p-Dimethylaminoazobenzene	3.85
Quinaldine red	3.44
Benzopurpurin 4B	3.24
Orange IV	2.94
p-Phenylaminoazobenzene	2.79
α-Naphtholbenzein	0.25

These indicators are somewhat anomalous in their behavior in acetic acid solutions as compared to their behavior in an aqueous solution. The strongly acidic character and low dielectric constant of the solvent exert a strong influence on the character of the indicator. These influences are manifested in "medium" and counterion effects which result in lateral displacements and intensity changes in the absorption curves. In determining the "true" ionization ratios, BH^+/B, the necessary corrections for these anomalous behaviors were employed.

Fifteen indicators have been evaluated as replacements for potentiometric titration procedures in acetic acid. Of these, the basic and triarylcarbinol types were found to give the sharpest change at the stoichiometric endpoint. With acidic type indicators, the endpoints were very difficult to distinguish.

In summary, these investigations have shown that relative alkalinity in acetic acid is limited to H_0 values in the vicinity of + 5, corresponding to a pH of approximately 5. The indicators found to be most useful in this relatively alkaline range of the solvent were quinaldine red and p-dimethylaminoazobenzene with pK' values of 3.44 and 3.85, respectively. Although the indicator, thymol blue, was found to have a pK' of 4.24, its color change in acetic acid is rather difficult to discern.

BIBLIOGRAPHY

1. L. Kahlenberg, J. Phys. Chem., 6, 1 (1902).
2. D. Vorlander, Ber., 36, 1485 (1903).
3. O. Folin and A. H. Wentworth, J. Biol. Chem., 7, 421 (1910).
4. O. Folin and F. F. Flanders, J. Am. Chem. Soc., 34, 774 (1912).
5. O. Folin and F. F. Flanders, J. Biol. Chem., 11, 257 (1912).
6. A. Hantzsch, Z. physik. Chem., 61, 257 (1908).
7. A. Hantzsch, ibid., 65, 41 (1909).
8. A. Lapworth, J. Chem. Soc., 107, 857 (1915).
9. A. Hantzsch, Z. Elektrochem., 29, 221 (1923).
10. J. N. Brønsted, Rec. trav. chim., 42, 718 (1923).
11. T. M. Lowry, Trans. Faraday Soc., 20, 13 (1924).
12. N. F. Hall and J. B. Conant, J. Am. Chem. Soc., 49, 3047 (1927).
13. J. B. Conant and N. F. Hall, ibid., 49, 3062 (1927).
14. E. C. Franklin, ibid., 46, 2137 (1924).
15. E. C. Franklin, ibid., 27, 820 (1908).
16. A. F. O. Germann, ibid., 47, 2461 (1925).
17. A. F. O. Germann and C. R. Timpany, ibid., 47, 2275 (1925).
18. W. F. Luder and S. Zuffanti, "Electronic Theory of Acids and Bases," New York, J. Wiley and Sons, Inc., 1946, pp. 13-14.
19. M. Usanovich, J. Gen. Chem., U. S. S. R., 9, 182 (1939).
20. N. F. Hall, J. Chem. Ed., 17, 124 (1940).
21. N. F. Hall and T. H. Werner, J. Am. Chem. Soc., 50, 2367 (1928).
22. C. W. Pifer and E. G. Wollish, Anal. Chem., 24, 300 (1952).
23. N. F. Hall, J. Am. Chem. Soc., 52, 5115 (1930).
24. J. B. Conant and T. H. Werner, ibid., 52, 4436 (1930).
25. I. M. Kolthoff and A. Willman, ibid., 56, 1007 (1934).

26. I. M. Kolthoff and A. Willman, ibid., 56, 1014 (1934).

27. A. Hantzsch and W. Voigt, Ber., 62, 975 (1929).

28. E. Griswold, M. M. Jones and R. K. Birdwhistell, J. Am. Chem. Soc., 75, 5701 (1953).

29. T. L. Smith and J. H. Elliott, ibid., 75, 3566 (1953).

30. R. M. Fuoss and C. A. Kraus, ibid., 55, 476 (1933).

31. R. M. Fuoss, ibid., 57, 488 (1935).

32. M. Usanovich and K. Yatsimirskiĭ, Zhur. Obshcheĭ Khim., 11, 954 (1941).

33. M. Usanovich and K. Yatsimirskiĭ, ibid., 11, 959 (1941).

34. L. P. Hammett and A. J. Deyrup, J. Am. Chem. Soc., 54, 2721 (1932).

35. L. P. Hammett and M. A. Paul, ibid., 56, 827 (1934).

36. N. F. Hall and W. F. Spengeman, ibid., 62, 2487 (1940).

37. N. F. Hall and F. Meyer, ibid., 62, 2493 (1940).

38. G. Harbottle, ibid., 73, 4024 (1951).

39. L. P. Hammett, "Physical Organic Chemistry," McGraw-Hill Book Co., New York, N. Y., 1940, p. 266.

40. H. Lemaire and H. J. Lucas, J. Am. Chem. Soc., 73, 5198 (1951).

41. L. Michaelis and S. Granick, ibid., 64, 1861 (1942).

42. M. L. Danguilan, M. S. Thesis, University of Wisconsin Library, Madison, Wisconsin (1953).

43. W. C. Eichelberger and V. K. LaMer, J. Am. Chem. Soc., 55, 3633 (1933).

44. J. S. Fritz, "Acid-Base Titrations in Nonaqueous Solvents," The G. Frederick Smith Chemical Co., Columbus, Ohio, 1952.

45. H. Adkins and H. I. Cramer, J. Am. Chem. Soc., 52, 4349 (1930).

46. J. E. Kunzler, Anal. Chem., 25, 93 (1953).

47. L. A. Flexser, L. P. Hammett and A. Dingwall, J. Am. Chem. Soc., 57, 2103 (1935).

48. A. Thiel, Fortschr. Chem. Physik u. physik. Chem., 18, No. 3, 48 (1924).

49. M. T. Rogers, T. W. Campbell and R. W. Maatman, J. Am. Chem. Soc., 73, 5122 (1951).

50. N. F. Hall and M. R. Sprinkle, ibid., 54, 3469 (1932).

51. H. C. Brown and M. D. Taylor, ibid., 69, 1332 (1947).

52. M. M. Davis and H. B. Hetzer, J. Research Nat'l. Bur. Standards, 46, 496 (1951).

53. M. M. Davis and H. B. Hetzer, ibid., 48, 381 (1952).

54. T. Higuchi and C. Rehm, unpublished data.

55. C. W. Pifer, E. G. Wollish and M. Schmall, Anal. Chem., 26, 215 (1954).

56. J. S. Fritz and R. T. Keen, ibid., 25, 407 (1953).

57. T. Higuchi and J. Concha, J. Am. Pharm. Assoc., Sci. Ed., 40, 173 (1951).

58. T. Higuchi and J. Concha, Science, 113, 210 (1951).

59. A. Hantzsch, Ber., 63, 1760 (1930).

60. N. Bjerrum, Kgl. Danske Videnskab. Selskab., 7, No. 9 (1926).

61. S. Glasstone, "Introduction to Electrochemistry," New York, D. Van Nostrand Co., Inc., 1942, p. 307.

62. L. F. Audrieth and J. Kleinberg, "Non-Aqueous Solvents," New York, J. Wiley and Sons, Inc., 1953, pp. 12-13.

63. M. M. Davis and P. J. Schuhmann, J. Research Nat'l. Bur. Standards, 39, 221 (1947).

Approved by: Takeru Higuchi

Assoc Prof of Pharmacy

June 2nd 1954