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## The Effect of Tetraethylammonium Ion on the Spectra of Certain Sulfonphthalein Indicators

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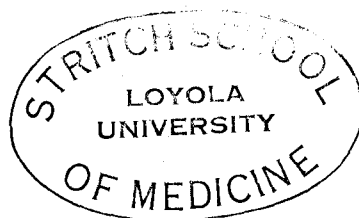
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THE EFFECT OF TETRAETHYLAMMONIUM ION ON THE SPECTRA  
OF CERTAIN SULFONPHTHALEIN INDICATORS



by

Irene E. Mersol

A Thesis submitted to the Faculty of the Graduate School  
of Loyola University in Partial Fulfillment of  
the Requirements for the Degree of  
Master of Science

February

1961

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## LIFE

Irene Emily Mersol was born in Ljubljana, Yugoslavia, April 15, 1935.

She was graduated from Notre Dame Academy, Cleveland, Ohio, June, 1953 and from Ursuline College for Women, Cleveland, Ohio, June, 1957 with the degree of Bachelor of Science.

In September, 1957 she began graduate studies at Loyola University in the Department of Biochemistry holding the positions of research assistant through September, 1959, and teaching assistant from September 1959 to March, 1960.

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## CHAPTER I

### INTRODUCTION

It has been known for many years that the colors of indicators are altered when present in solutions with soaps and detergents. The phenomenon has been explained as due to preferential absorption of one form of the indicator on the detergent micelles, followed by a displacement of the acid-base equilibrium in favor of this form. Hartley (1934) studied effects of anionic, cationic and non-ionic detergents on a large number of colored substances. In his studies he used long-chain quaternary ammonium salts which give soap-like solutions in which aggregates (micelles) of long-chain ions exist and are similar to the ones found in true soaps.

Hartley showed that the greatest color changes occur when the charge on the detergent is opposite to that on the indicator ion. When negatively charged aggregates of long-chain anions are added to a solution of indicator (present in an electrically neutral form) the same effect is observed as if an acid were added; long-chain cations produce the same effect as would the addition of a base. The color change was thought to be due to interaction of electric charges on the indicator and the long-chain ions.

Another displacement was noted with certain indicators



which could not be due to electrostatic forces alone. The effect was most striking in the case of cationic detergents and sulfon-phthalein indicators the color of the solution was not only displaced to the alkaline side, but was found to be qualitatively changed also. It was thought at first that this effect might be due to the presence of more than one color changing compound, but in strongly alkaline solutions where the indicator would exist in one ionic form only, the long-chain cations still had the same qualitative effect. The absorption spectrum of the ions must therefore be different in the presence of the long-chain cation, and chemical combination is suggested.

Hartley (1938) used the above mentioned color change (in brom phenol blue) as the end point for the direct titration of paraffin chain anions against paraffin chain cations (they form an insoluble salt); the alkaline purple color is restored to the solution when the paraffin chain cation is removed in this manner.

Auerbach (1943) and later Ballard et al. (1954) give methods for determining the concentration of long-chain quaternary ammonium salts photometrically, based on the measurement of the indicator extracted into a non-aqueous phase by means of the quaternary ammonium compound. Ballard and coworkers used an excess of brom thymol blue (most color was obtained with this dye) and chloroform as the extracting solvent (the pH was about 7). They found that for higher molecular weight compounds the slopes of the calibration curves expressed as extinction/concentration (mg/ml)

were directly proportional to the number of quaternary nitrogen atoms per molecule and inversely to the molecular weight (or, that one molecule of indicator combines with each nitrogen atom of the quaternary ammonium salt).

Colichman (1947) applied the indicator displacement effect to the direct photometric method of determination of quaternary ammonium salts by using an excess of brom phenol blue in alkaline carbonate solution. The color intensity of the solutions is measured directly (in a colorimeter) without extracting the colored complex with an organic solvent as in the methods mentioned above; In this way difficulties connected with the use of organic solvents in the extraction of color are avoided.

The change in color in brom phenol blue in the presence of some quaternary ammonium salts was studied by Zutrauen and Ter-Minassian (1955) using bromide salts. Solutions of lauryl ( $C_{12}$ ), myristyl ( $C_{14}$ ) and cetyl ( $C_{16}$ ) -trimethylammonium bromides were studied at different concentrations. At first, as the concentration of the cation increases (from  $0.45 \times 10^{-5}$  to  $6.8 \times 10^{-5} M$ ), the optical density at the maximum ( $5920 \text{Å}$ ) decreases; it starts to rise with concentrations greater than  $9.07 \times 10^{-5} M$ , and a new peak appears at about  $6050 \text{Å}$  (the concentration of the dye was the same in all solutions,  $7.5 \times 10^{-6} M$ ). The color change was thought to be caused by the formation of a complex (molecular or micellar) between the indicator ion and the salt. The absorption spectra of brom phenol blue (in the presence of the quaternary ammonium salts)

were studied both with and without potassium chloride (1 M). A minimum in optical density was found in both solutions (with and without electrolyte) at the same concentration of the quaternary ammonium salt when optical densities were plotted against the salt concentration. Since it has been reported (Phillips and Mysels, 1955; Harkins et al. 1946) that the critical micelle concentration (the concentration at which micelles appear) is affected by the presence of electrolytes in solution, the phenomena studied must not be due to micelle formation. In the solutions studied, the critical micelle concentration was found to be at higher salt concentrations than those showing the minimum; using surface tension methods the critical micelle concentrations were found to be at concentrations where the observed optical densities were high and constant. The authors thought that the complex between the indicator and the long-chain salts is thus molecular and that it did not change even when micelles did appear.

Losses of quaternary ammonium compounds due to absorption on glass and concentration in foam were investigated by Fogh et al. (1954) in order to find a colorimetric method for the quantitative microdetermination of quaternary ammonium compounds. All measurements were done in aqueous solutions; along with other data a graph is presented showing a pronounced difference between the absorption spectra of brom cresol purple in absence of salt and in the presence of salt and in the presence of quaternary ammonium salts.

The experimental results discussed so far are not, in

general, applicable to low molecular weight quaternary ammonium salts. Cochin and Woods (1951) published a method for the estimation of tetraethylammonium ion in the plasma and urine of the dog (following an intravenous injection of 20 mg/kg of the compound). A sample of the solution is treated with 10% trichloroacetic acid, centrifuged and the supernatant filtered; a portion of this filtrate is treated with an aqueous solution of brom cresol purple (the indicator concentration being in 100 fold excess of the salt), the indicator-tetraethylammonium ion complex extracted into chloroform, treated with base (0.1 M NaOH) and the alkaline layer read at 5950Å in a Beckman spectrophotometer. The authors found a straight line relationship between the optical density and the concentration of tetraethylammonium ion (5 to 35 micrograms/ml), in both urine and plasma.

Mitchell and Clark (1952) developed a similar method for the determination of low chain quaternary ammonium ions in biological media, using an excess of brom phenol blue and ethylene dichloride as the extracting solvent.

Most of the work described thus far has been done with quaternary ammonium compounds which contain at least one carbon unit of considerable size. In a few experiments it has been shown that the total interaction of tetramethylammonium and tetraethylammonium ions with sulphonphthaleins is of a different order of magnitude than that of the long-chain quaternary cations. No equilibrium constants have been reported for any of these inter-

actions in aqueous solution. Most of the experiments reported involved two-phase systems so that even a qualitative order of reactivity in aqueous solution alone cannot be inferred from the results. The report of Zutrauen and Ter-Minassian (1955) seems to reveal a hitherto unsuspected reaction of brom phenol blue with long-chain quaternary ammonium ions in aqueous solution to form a species which does not absorb light to any appreciable extent in the neighborhood of the absorption of the free dye. These authors state "... (la légère précipitation observée ne serait que partiellement responsable de la décoloration)." So even here, the presence of a second phase prevents one from accepting without question the existence of appreciable complex in solution.

In certain kinetic measurements done in this laboratory (Melchior and Melchior, 1958) a sulfonphthalein dye was used as an indicator of the change of hydrogen ion concentration. A part of the research depended upon changes in the velocity measured caused by the substitution of tetramethylammonium ion for sodium ion. It was noted that the color of unbuffered dye solutions containing sodium ion was different from that of solutions identical except for the substitution of an equivalent amount of tetramethylammonium ion for sodium ion. Quantitative measurement showed that the measurements concerned were not affected appreciably, but it was desirable to determine the extent and nature of the interaction, and to investigate tetraethylammonium ion which might be used similarly.

## CHAPTER II

### EXPERIMENTAL

#### I REAGENTS USED

The water used in the experiments was redistilled in an all glass system. Carbon dioxide was removed by passing nitrogen gas through the solution and the water was then dispensed from a container protected by soda lime.

Other materials used were:

Acetic Acid - Glacial Acetic Acid, Mallinckrodt Analytical Reagent 2504, lot ASP.

Brom Cresol Green - Tetrabromo-m-cresolsulfonphthalein, National Aniline Division 330, lot 16327, three times recrystallized from chloroform and carbon tetrachloride, dried and stored under high vacuum prior to use.

Brom Phenol Blue - Tetrabromophenolsulfonphthalein, Harleco Water soluble 859, lot 34.

Brom Thymol Blue - Dibromothymolsulfonphthalein, Harleco Water soluble 863.

Cetyl Trimethylammonium Bromide - Hexadecyltrimethylammonium Bromide, Eastman Kodak T 5650, lot 7.

Cresol Red - o-cresolsulfonphthalein, Harleco 878, lot 50 and National Aniline Division 232, lot 16402. A one to one mixture of

the two, twice recrystallized from acetone and chloroform, dried and stored under high vacuum prior to use, was used in the preparation of solutions.

Hydrochloric Acid - Concentrated, Mallinckrodt Analytical Reagent 2612, lot GPY. Standardized against standard sodium hydroxide using 1% phenolphthalein as indicator.

Silver Nitrate - Mallinckrodt Analytical Reagent 2169, lot BSY, standardized against standard hydrochloric acid neutralized with sodium hydroxide, with 5% potassium chromate as indicator.

Sodium Bromide - Mallinckrodt Analytical Reagent 0535, lot BMH.

Sodium Hydroxide - Fifty-one per cent, Baker Analytical Reagent, lot 6062. Standardization of dilute aqueous solutions was done against potassium acid phthalate (Mallinckrodt Primary Standard, lot APMA) with 1% phenolphthalein as indicator, following the procedure outlined by the National Bureau of Standards.

Sodium Phosphate - Sodium Monohydrogen Phosphate, Heptahydrate Mallinckrodt Analytical Reagent 7914, lot NHX.

Sodium Phosphate Tribasic - Mallinckrodt Analytical Reagent 7940, lot XHM.

Tetraethylammonium Bromide - Eastman Kodak 1514, lot 21, recrystallized twice from 2-propanol. Solutions were standardized against silver nitrate using 5% potassium chromate as indicator.

Tetramethylammonium Bromide - Eastman Kodak 1521, three times recrystallized, the solutions used standardized with silver nitrate.

## II PREPARATION OF SOLUTIONS FOR SPECTRA

Aliquots of an aqueous solution of indicator were pipetted into volumetric flasks of appropriate size. To these flasks were added measured amounts of the salt solutions (tetraethylammonium bromide or sodium bromide) and acid or base or buffer as appropriate.

The solutions were made to volume with nitrogen purged, glass distilled water, mixed and the absorption spectra determined against water in cells with fused silica windows and equipped with standard taper stoppers. A Beckman DU spectrophotometer with a photomultiplier attachment and temperature controlled cell compartment was used. Tap water was circulated through the lamp housing to prevent overheating of the lamp. The slit width was varied from 0.03 to 0.10 mm. as required to maintain a useful sensitivity. Readings to be compared at a given wave length were made with identical slit width settings.

The pH of the solutions studied was, in general, that at which more than 99% of the indicator will be present as a single ionic species. This calculation was based on the  $pK_a$  of the indicator and the assumption that no complexes are formed between any of the indicator species and positive ions other than hydrogen ions. For example, cresol red has a  $pK_a$  of approximately 8 (7.7 to 8.3 in the range of ionic strengths used). Therefore, at pHs less than 4.7 there will be less than 0.1% of the basic form of the indicator present in solution, while at pHs greater than



11.3 there will be less than 0.1% of the acid form of the indicator present in solution. As can be seen in Table I, cresol red was studied in 0.001 N acetic acid, pH 3.8 and in 0.01 M trisodium phosphate, pH 11.7 (R. G. Bates, F. D. Pinching, and E. R. Smith 1950). Excessively high and excessively low pHs were avoided because of the possibility of slow reactions with or catalysis by high concentrations of hydrogen or hydroxide ion.

TABLE I  
COMPOSITION OF SOLUTIONS USED FOR SPECTRA

<u>Solution</u>	<u>Bromothymol Blue</u>		<u>Cresol Red</u>	
	<u>Acid</u>	<u>Basic</u>	<u>Acid</u>	<u>Basic</u>
DYE: mg/ml	0.020	0.010	0.010	0.005
Molarity	$3.20 \times 10^{-5}$	$1.60 \times 10^{-5}$	$6.66 \times 10^{-6}$	$3.33 \times 10^{-6}$
SALT: when used				
NaBr	0.1500M	0.1500M	0.1652M	0.1652M
$(C_2H_5)_4NBr$	0.1490M	0.1490M	0.1451M	0.1451M
ACID: Acetic	0.001M	-	0.001M	-
BASE: $Na_3PO_4$	-	0.01M	-	0.01M
	<u>Brom Cresol Green</u>		<u>Brom Phenol Blue</u>	
DYE: mg/ml	0.020	0.010	0.015	0.006
Molarity $\times 10^{-6}$	7.29	3.65	9.49	3.80
SALT: when used				
NaBr	0.1652M	0.1652M	0.1500M	0.1500M
$(C_2H_5)_4NBr$	0.1451M	0.1451M	0.1490M	0.1490M
ACID: HCl	0.0105M	-	0.1047M	-
BASE: $Na_2HPO_4$	-	0.01M	-	0.01M

Sager et al. (1948) found that the extent of fading of sulfonphthalein indicators in alkaline solutions depended on the concentration of alkali. The reaction being:



whereby the alkaline form of the indicator ( $R^-$ ) adds a hydroxyl ion to form the colorless carbinol ( $ROH^-$ ). The replacement of a hydrogen atom by halogen increases its rate of reaction with the negative hydroxyl ion. The opposite inductive effect of a methyl or other alkyl group leads to a decrease in the rate of the fading reaction.

These authors found that the time required to decrease the initial color intensity to one half (in strong alkali) was : 20 minutes for brom phenol blue, 30 hours for brom cresol green and cresol red, and 70 days for brom thymol blue. At lower concentrations of alkali the fading reactions were considerably slower (and are not significant under the conditions used in the research reported in this thesis).

The spectra obtained in the work presented here were corrected from the similarly measured absorption spectra of solutions which contained no indicator, but were otherwise identical.

The Tyndall effect was used to determine the presence of a second phase in these solutions. The reason being that if a beam of light is passed through a solution in which particles are present, these will reflect the light and thus introduce an error in the measurements of absorbance. A considerable Tyndall effect was observed in certain indicator solutions containing cetyl

trimethylammonium bromide.

The purity of the various samples of indicator was investigated by descending paper chromatography on acid washed filter paper prepared especially for chromatography (Schleicher and Schuell 589 "Orange Ribbon"). The developed chromatograms were examined in visible light and under illumination with mercury vapor lamps (Mineralite) equipped with filters for the 2537 and the 3660 lines of mercury. Samples which showed more than one component were not considered sufficiently pure for critical experiments.

### III SOLUTIONS USED FOR CHROMATOGRAPHY

A BASIC. The mixture contained concentrated ammonium hydroxide, n-butyl alcohol and water (1:10:9 by volume). The two phases obtained were separated, the lower water layer used only to equilibrate the chamber, while the upper butanol layer was the traveling solvent.

B ACID. The mixture (one phase) consisted of glacial acetic acid, tertiary butyl alcohol and water (1:4:1 by volume).

The dye samples (approximately 10 mg.) were dissolved in a minimum amount of spotting solution (0.1 M aqueous ammonium hydroxide) one hour before application to paper strips. Amounts of four to eight microliters were then applied and the spots dried for one hour. The paper strips were then placed into chromatography jars and allowed to equilibrate for a minimum of twelve hours; the appropriate solvent was added to the solvent troughs

and allowed to descend eight hours in basic and sixteen hours in acid solution. The chromatograms were then removed from the chambers, allowed to air-dry on a stainless steel rack for approximately one hour, and examined for components in ordinary and ultraviolet light.

## CHAPTER III

### RESULTS

When the absorption spectra of several commercial samples of sulfonphthalein dyes were studied, a small but definite difference was found between solutions in aqueous sodium bromide and those in aqueous tetraethylammonium bromide. The following indicator-dyes were used:

O-cresolsulfonphthalein or cresol red,

3', 3'', 5', 5'' -tetrabromophenolsulfonphthalein or brom phenol blue

3', 3'', 5', 5'' -tetrabromo-m-cresolsulfonphthalein or brom cresol green,

3', 3'' -dibromothymolsulfonphthalein or brom thymol blue.

All above dyes show three maxima of absorption in basic and two distinct maxima in acid solution with a third in the form of a shoulder or plateau. The molar absorbances at the positions of maximum absorption in acid and basic solutions and in the presence of the two salts (sodium bromide and tetraethylammonium bromide) are given in Table II. In these comparisons aliquots of the same dye solutions were used in all cases.

Brom phenol blue and brom thymol blue in the presence of either tetraethylammonium or sodium bromide (the salt concentration being kept constant) were found to follow Beer's and Lambert's law in the concentrations between 1.5 to 10 mg/ml of dye in basic and

TABLE II

## THE EFFECT OF SALTS ON THE ABSORPTION MAXIMA OF VARIOUS DYES

Test Conditions: 0.15 Molar salt solutions used

DYE	MAXIMUM		MOLAR ABSORBANCE $\times 10^{-3}$	
	$\lambda$	$\text{cm}^{-1}$	NaBr	$\text{Et}_4\text{NBr}$
CRESOL RED				
Basic:	5720	17483	217.	223.
	3636	27500	21.9	20.7
	2941	34000	42.6	42.0
Acid: (plateau)	4348	23000	77.9	76.1
	3175	31500	20.1	21.1
BROM PHENOL BLUE				
Basic:	5900	16950	157.	159.
	3810	26250	17.4	17.4
	3101	32250	32.8	30.6
Acid: (shoulder)	4348	23000	38.9	33.0
	3279	30500	9.6	8.7
	2759	36250	19.3	17.17
BROM CRESOL GREEN				
Basic:	6154	16250	187.	191.
	4000	25000	37.8	37.8
	3077	32500	53.7	54.8
Acid: (shoulder)	4444	22500	80.2	77.5
	3509	28500	31.0	30.9
	2778	36000	41.1	37.6
BROM THYMOL BLUE				
Basic:	6154	16250	330.	341.
	3940	25380	75.2	75.
	3053	32750	122.	118.0
Acid: (shoulder)	4320	23125	145.	145.
	3333	30000	67.8	71.0
	2797	35750	36.8	81.7

3.75 to 20.0 mg/ml of dye in acid solutions; 10, 20 and 50 mm cells were used in the study of the effect of path length.

The lines obtained when the optical density of the solutions (at the first absorption maximum in acid and base) were plotted against path length and dye concentration all passed through the origin within experimental error.

The commercial dye samples were tested for purity by descending paper chromatography. The results obtained are given in Table III. A number of components was found in all samples except brom thymol blue.

Cresol red and brom cresol green were selected for more careful study. Three times recrystallized brom cresol green shows only a trace of a second component; in samples of cresol red no second component was found on the chromatograms even after the first recrystallization. The latter part of Table III shows the results obtained.

The purified samples of cresol red and brom cresol green were used in all the experiments that follow. No difference was observed between the absorption spectra of the purified and commercial samples.

Figures 1 and 2 show the absorption spectra of cresol red solutions containing 0.5 molar sodium bromide in the presence of base (0.01 M sodium triphosphate) and acid (0.001 M acetic acid). Sodium bromide was chosen because the extinction coefficients of dye solutions without salt and identical solutions except for the

## CHROMATOGRAPHY OF SULFONPHTHALEIN INDICATORS

Substance	Visible Spots		Fluorescent Spots	
	Acid*	Basic	Acid	Basic
<b>COMMERCIAL</b>				
Brom thymol blue	1	1	none	none
Brom phenol blue	2	3	none	3
Brom cresol green	1	2	1	3
Cresol red	1	1	none	1
<b>PURIFIED</b>				
Brom cresol green 1x <sup>a</sup>	1	1	1	1
Brom cresol green 2x	1	1	1 <sub>b</sub>	1 <sub>b</sub>
Brom cresol green 3x	1	1	1	1
Cresol red 1x	1	1	none	none
Cresol red 2x	1	1	none	none

\* "Acid" refers to chromatograms developed with a mixture of one volume of glacial acetic acid, four volumes of tertiary butyl alcohol and one volume of water. "Basic" refers to chromatograms developed with the upper layer of a mixture of one volume of concentrated ammonium hydroxide, ten volumes of normal butyl alcohol and nine volumes of water.

a The numbers refer to the number of times the sample was crystallized.

b The single fluorescent component decreased with each crystallization, it is barely perceptible in this sample.

presence of this salt are essentially the same. By the use of this 'inert' salt one can avoid the possible effect of ionic strength which might enter in if solutions of tetraethylammonium bromide (of a high ionic strength) were compared with a dye solution containing no salt. In all experiments where absorption spectra were compared aliquots of the same dye solution were added to flasks



THE ABSORPTION SPECTRA OF  
CRESOL RED IN 0.5 MOLAR NaBr  
(BASIC)

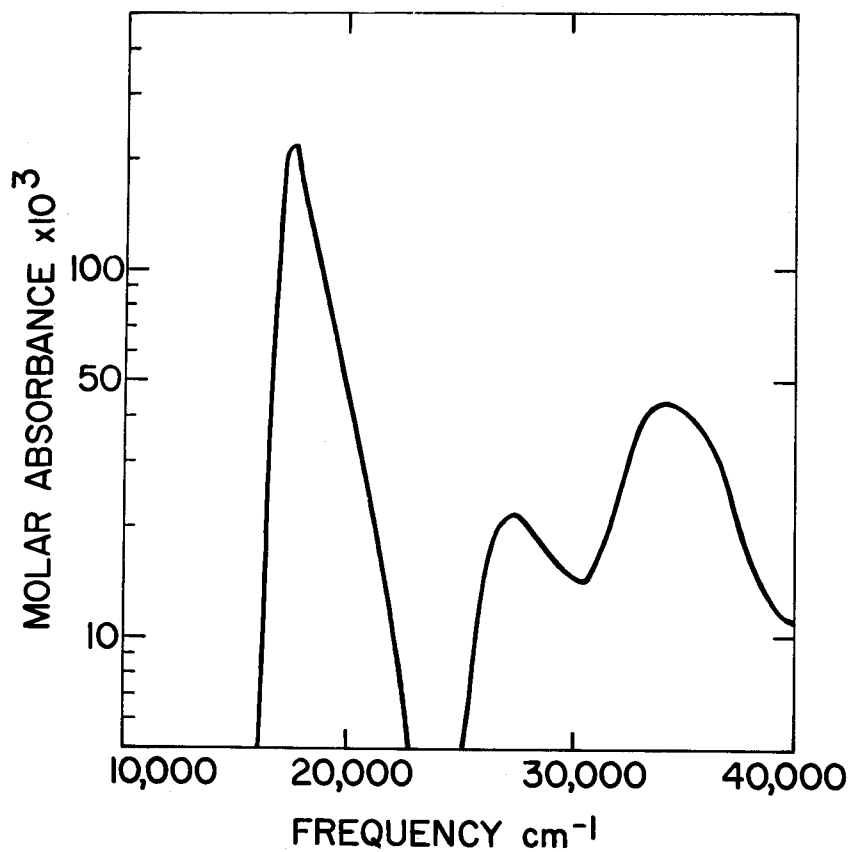


Figure 1

THE ABSORPTION SPECTRA OF  
CRESOL RED IN 0.5 MOLAR NaBr  
(ACID)

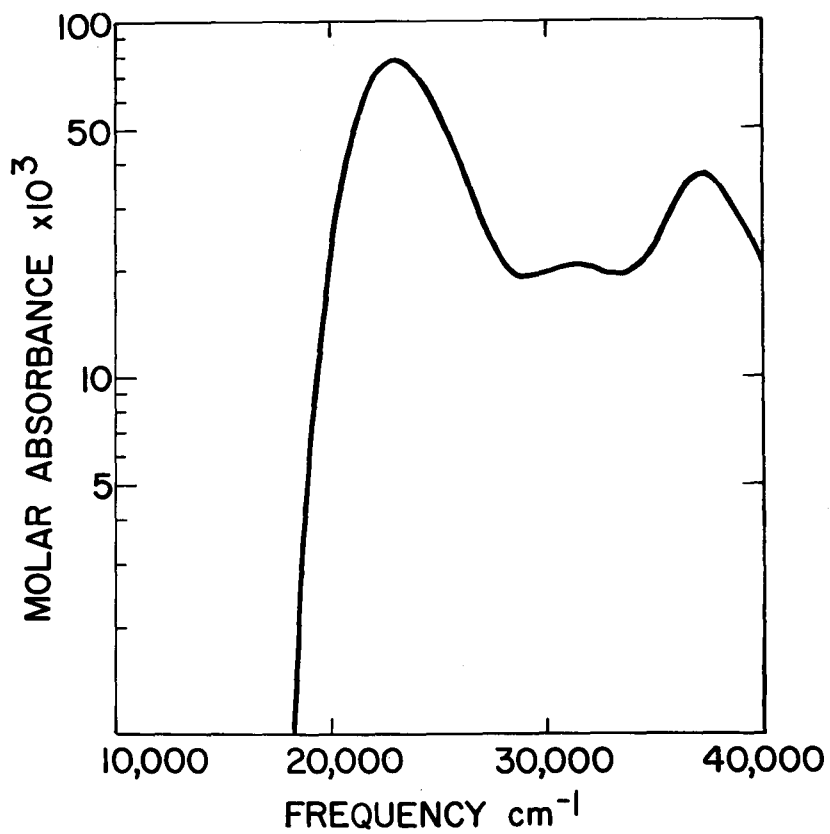


Figure 2

containing tetraethylammonium bromide and sodium bromide of the same concentration.

Absorption spectra of brom cresol green solutions containing 0.5 molar sodium bromide in base (0.01 M sodium triphosphate) and acid (0.01 M hydrochloric acid) are given in Figures 3 and 4 respectively.

The difference between the molar absorbance of dye solutions in the presence of tetraethylammonium bromide and those with sodium bromide is small, and in order to show the differences clearly the absorption of dye solutions containing the latter salt were subtracted from those obtained for dye solutions containing tetraethylammonium bromide and plotted against frequency.

Figure 5 shows such a plot for cresol red in basic 0.5 M salt solutions. In acid solution (Figure 6) the effect of tetraethylammonium bromide on cresol red is considerably smaller and opposite to that in basic solutions.

Similar but less pronounced differences were found with indicator solutions (acidic and basic) containing 0.15 molar salts. Table IV shows the difference in molar absorbance between the two salts at the first maximum of absorption in both acid and base. It is clear that tetraethylammonium ion causes an increased absorption at the long wave length side (low energy) of the first basic band; this is followed by a decrease in absorption on the high energy side of the first acid absorption maxima. Effects on the higher energy absorption bands are not large enough to be well resolved.

THE ABSORPTION SPECTRA OF  
BROM CRESOL GREEN IN 0.5 MOLAR NaBr  
(BASIC)

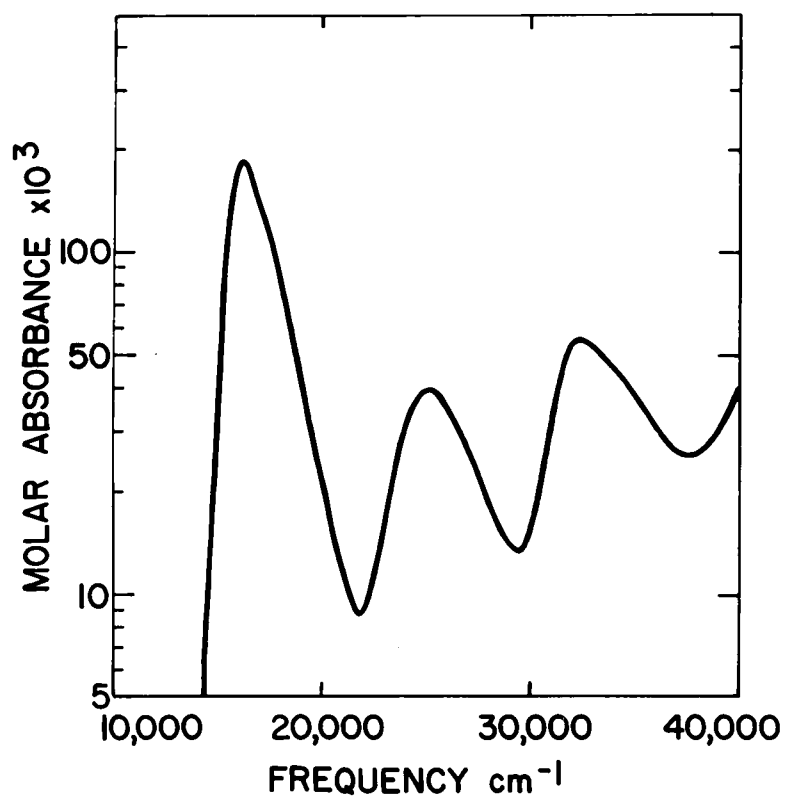


Figure 3

THE ABSORPTION SPECTRA OF  
BROM CRESOL GREEN IN 0.5 MOLAR NaBr  
(ACID)

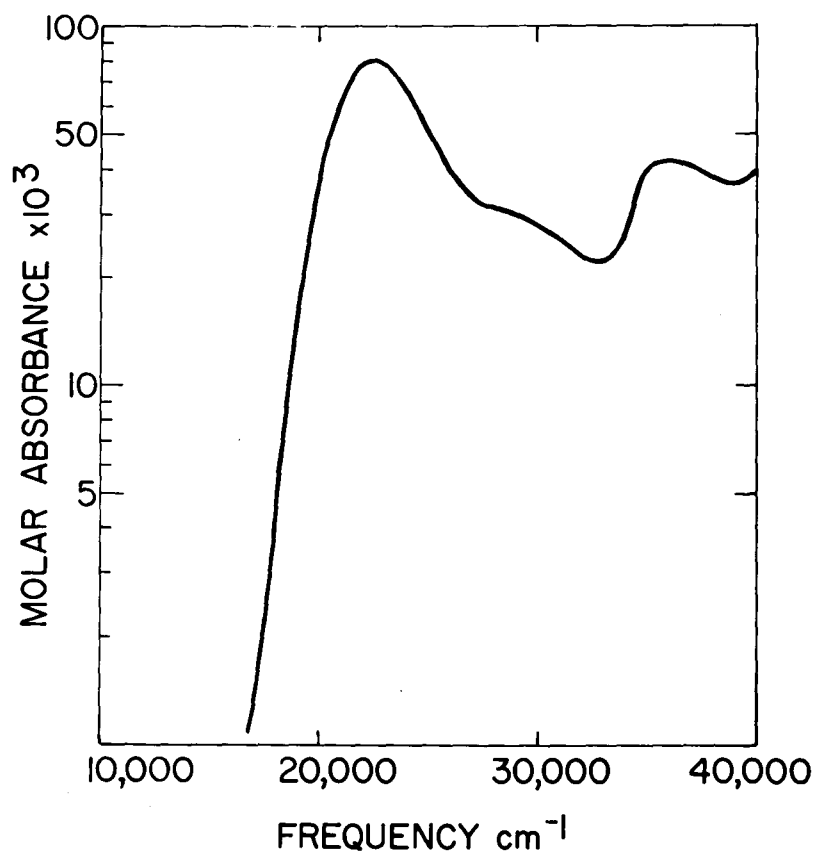


Figure 4

THE EFFECT OF TETRAETHYLAMMONIUM  
ION ON THE ABSORPTION OF LIGHT BY  
BASIC SOLUTIONS OF CRESOL RED

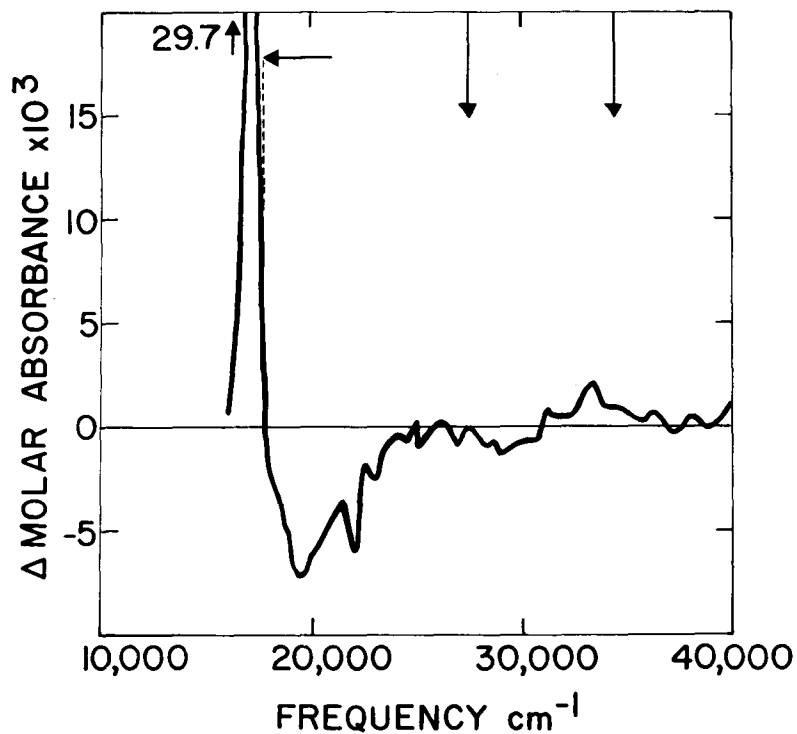


Figure 5

Molar absorbance in 0.5 M  $\text{Et}_4\text{NBr}$  minus molar absorbance in 0.5 M NaBr.

The three arrows on the right indicate the positions of maximum absorption in 0.5 M NaBr.

THE EFFECT OF TETRAETHYLAMMONIUM  
ION ON THE ABSORPTION OF LIGHT BY  
ACID SOLUTIONS OF CRESOL RED

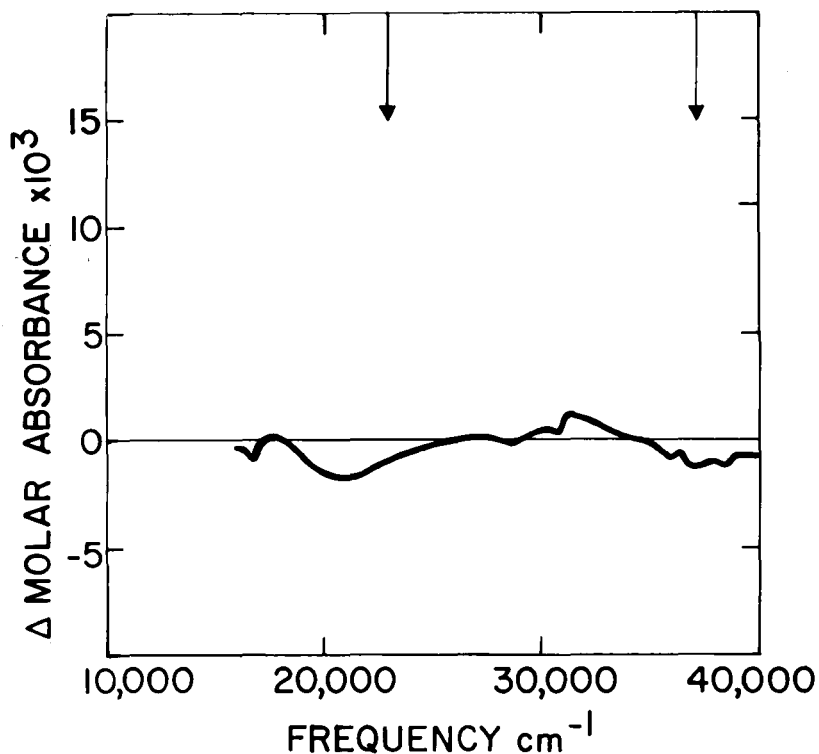


Figure 6

Molar absorbance in 0.5 M  $\text{Et}_4\text{NBr}$  minus molar absorbance in 0.5 M NaBr.

The upper arrows indicate the positions of maximum absorption or shoulders in 0.5 M NaBr.

THE EFFECT OF TETRAETHYLAMMONIUM  
ION ON THE ABSORPTION OF LIGHT BY  
BASIC SOLUTIONS OF BROM CRESOL GREEN

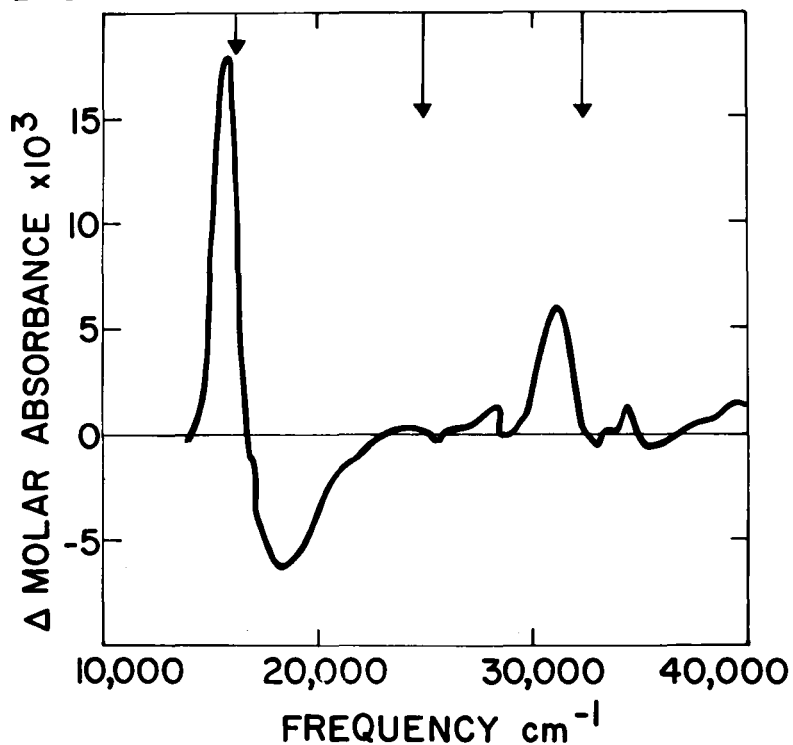


Figure 7

Molar absorbance in 0.5 M  $\text{Et}_4\text{NBr}$  minus molar absorbance in 0.5 M  $\text{NaBr}$ .

The upper arrows indicate the positions of maximum absorption in 0.5 M  $\text{NaBr}$ .



THE EFFECT OF TETRAETHYLAMMONIUM  
ION ON THE ABSORPTION OF LIGHT BY  
ACID SOLUTIONS OF BROM CRESOL GREEN

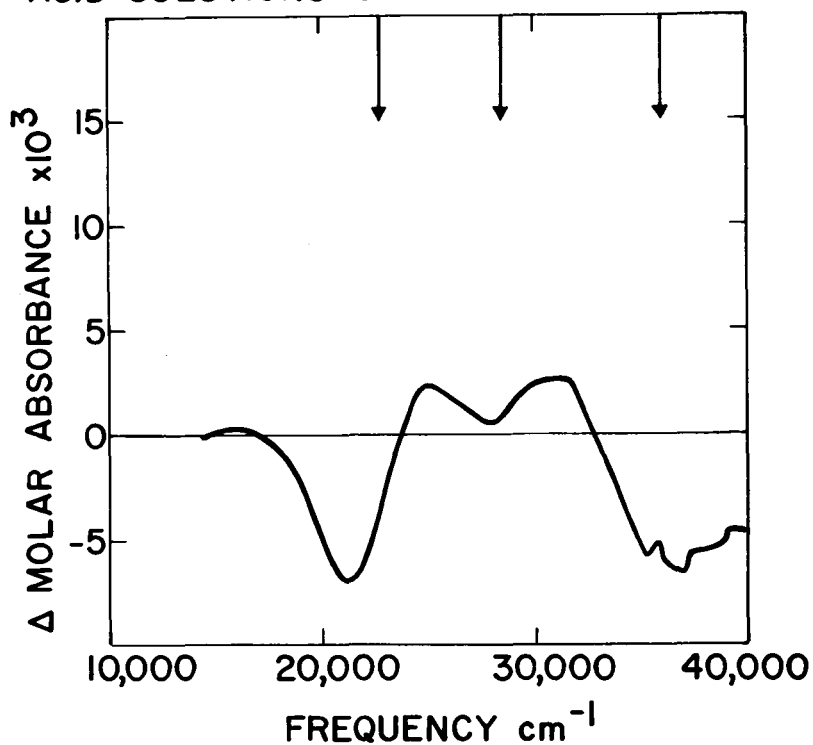


Figure 8

Molar absorbance in 0.5 M  $\text{Et}_4\text{NBr}$  minus molar absorbance in 0.5 M  $\text{NaBr}$ .

The upper arrows indicate the positions of maximum absorption or shoulders in 0.5 M  $\text{NaBr}$ .

Tetraethylammonium ion has a similar effect on the molar absorbance of basic solutions of brom cresol green (see Figure 7).

Figure 8 shows the difference in molar absorbance of the two salts in acid aqueous solutions of brom cresol green. The effects found here are more pronounced than in cresol red but the directions are the same with both dyes. For comparison of the values obtained in 0.15 and 0.50 molar salt solutions see Table IV.

TABLE IV

EFFECT OF SALT CONCENTRATION ON ABSORPTION MAXIMA OF  
BROM CRESOL GREEN AND CRESOL RED

$\Delta$  Molar absorbance  $\times 10^3$

Dye	CRESOL RED		BROM CRESOL GREEN	
	Basic	Acid	Basic	Acid
0.15 M	+5.7	-1.2	+4.1	-2.7
0.50 M	13.2	-1.5	7.6	-4.4

1 unit of molar absorbance: is 0.0034 O.D. units in base  
is 0.007 O.D. units in acid

#### EFFECTS OF TEMPERATURE AND TIME

Some of the earlier measurements with long path length cells were made at "room temperature" which was  $24 \pm 2^\circ\text{C}$ . To determine whether temperature change might have any influence on the results obtained the spectra of several solutions were measured at  $25^\circ\text{C}$  and at  $37^\circ\text{C}$ . The effects are too small to have had a

significant effect on the measurements.

In order to be certain that the solutions were stable during the period of measurement, the absorption spectra of several dye solutions were remeasured after several days. Table V shows that measurable fading occurs in some, but not in all solutions, but the magnitude is not sufficient to cause difficulty in the measurements reported.

TABLE V

EFFECT OF TIME UPON THE FIRST ABSORPTION MAXIMA OF CERTAIN INDICATORS IN PRESENCE OF 0.15 MOLAR SALTS

INDICATOR	Per cent change in four days at $24 \pm 3^{\circ}\text{C}$			
	ACID		BASIC	
	NaBr	$\text{Et}_4\text{NBr}$	NaBr	$\text{Et}_4\text{NBr}$
Cresol red	+0.2	-1.2	+2.0	+0.2
Brom cresol green	-0.1	-0.1	-2.0	+0.2
Brom thymol blue	zero	-1.2	-0.2	+0.2
Brom phenol blue	-4.2	-4.0	-0.4	+1.6

#### SALT CONCENTRATION STUDIES USING BROM CRESOL GREEN

Brom cresol green was chosen for the study of the effect of salt concentration on its absorption spectra. The reason for this choice was the fact that the absorption spectra of the indicator-dye are affected to a considerable extent in both acid and basic solutions.

The differences in optical density between solutions

containing tetraethylammonium bromide and sodium bromide at the first absorption maxima in acid and basic solutions are given in Table VI. Aliquots of the same indicator solution were compared in all cases ( $7.29 \times 10^{-6} \text{M}$  indicator in acid and  $3.65 \times 10^{-6} \text{M}$  indicator in base).

A direct relationship is noted between the salt concentration and the difference in optical density. In order to find the maximum effect further experiments were performed using higher salt concentrations. The effect of tetramethylammonium bromide on the absorption spectra of brom cresol green was investigated at the same time. Molar absorbances in the presence of tetraethylammonium bromide and tetramethylammonium bromide were compared (Table VII) with the values obtained in sodium bromide of the same concentrations.

TABLE VI

SALT CONCENTRATION EFFECTS ON OPTICAL DENSITY AT MAXIMUM  
ONE OF BROM CRESOL GREEN SOLUTIONS

Salt Concentration (molar)	O.D. in $\text{Et}_4\text{NBr}$ minus O.D. in $\text{NaBr}$	
	BASIC $16250\text{cm}^{-1}$	ACID $22500\text{cm}^{-1}$
0.5	0.029	-0.032
0.15	0.015	-0.020
0.05	0.009	- .006
0.01	0.006	-0.007
0.001	0.003	+0.003
$3.8 \times 10^{-6}$	0.004	+0.002
$3.0 \times 10^{-6}$	0.004	+0.005

The differences in molar absorbance between the tetraalkyl bromides and sodium bromide (all 0.8 M) are shown in Figure 9; only the first absorption bands in both basic and acid aqueous solutions of brom cresol green are drawn. The effect of tetramethylammonium bromide is considerably smaller than that of the tetraethylammonium bromide.

Four wave lengths were chosen from the absorption spectra of brom cresol green for a comparison of the effects of the two quaternary ammonium salts: 1) the positions of the first maximum in sodium bromide in both acid and basic solutions, and 2) the positions of maximum difference in the absorption spectra (in both acid and base) caused by the tetraalkylammonium salts. The values obtained are given in Table VII below.

TABLE VII

EFFECT OF SALTS AND SALT CONCENTRATION ON THE OPTICAL  
DENSITY OF BROM CRESOL GREEN SOLUTIONS

Salt concentration (molar)	1) $\Delta$ BASIC 16250cm <sup>-1</sup>		2) $\Delta$ BASIC 15750cm <sup>-1</sup>	
	E	M	E	M
0.80	0.035	0.013	0.078	0.026
0.50	0.024	0.007	0.059	0.013
0.15	0.012	0.001	0.026	0.006
	1) $\Delta$ ACID 22500cm <sup>-1</sup>		2) $\Delta$ <sup>Acid</sup> BASIC 21000cm <sup>-1</sup>	
	E	M	E	M
0.80	-0.040	-0.025	-0.066	-0.032
0.50	-0.032	-0.017	-0.050	-0.023
0.15	-0.018	-0.020	-0.019	-0.015
E = O.D. Et <sub>4</sub> NBr - O.D. NaBr      M = O.D. Me <sub>4</sub> NBr - O.D. NaBr				

# THE EFFECT OF TETRAALKYLAMMONIUM ION ON THE ABSORPTION OF LIGHT BY SOLUTIONS OF BROM CRESOL GREEN

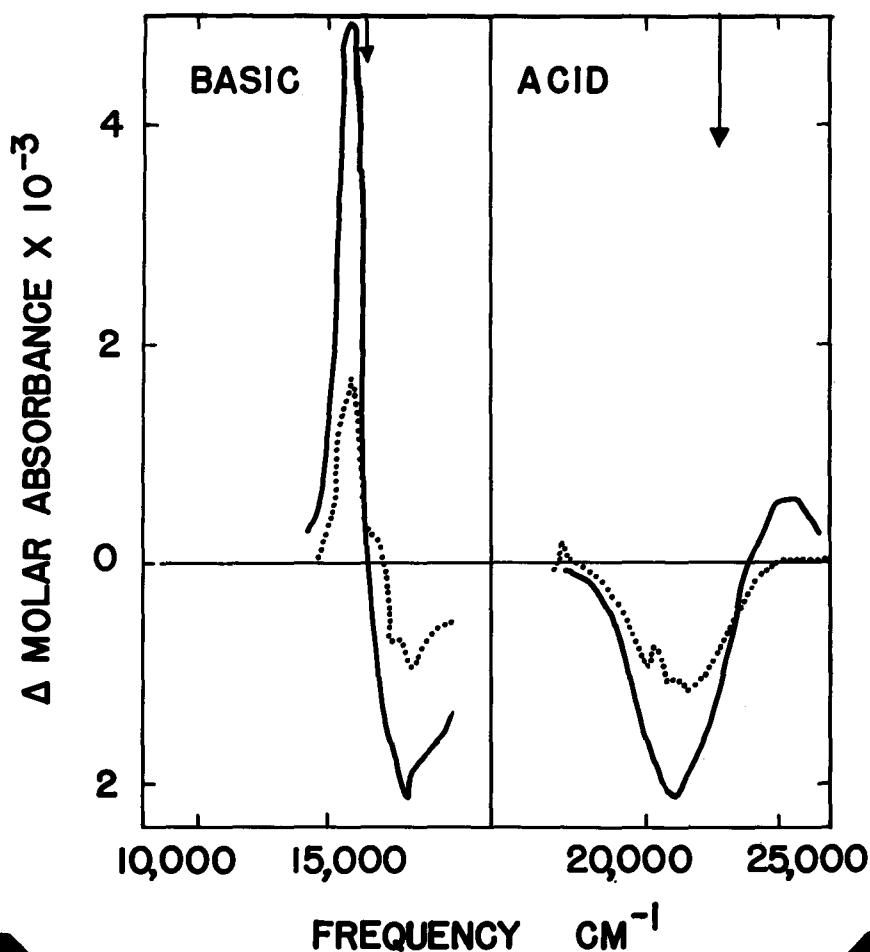


Figure 9

The solutions contain identical aliquots of brom cresol green in 0.01 M  $\text{Na}_3\text{PO}_4$

— Molar absorbance in 0.8 M  $\text{Et}_4\text{NBr}$  minus molar absorbance in 0.8 M  $\text{NaBr}$

- - - Molar absorbance in 0.8 M  $\text{Me}_4\text{NBr}$  minus molar absorbance in 0.8 M  $\text{NaBr}$

The upper arrows indicate the position of maximum absorption in 0.8 M  $\text{NaBr}$

The effect of tetramethylammonium bromide in base is approximately one third of that due to tetraethylammonium bromide; in acid media it is about one half of that observed for tetraethylammonium bromide in the more concentrated solutions (0.8 and 0.5 molar) and approaches the value obtained for the other salt when molar concentrations of 0.15 are studied. A graphical presentation of the data in Table VII is given in Figures 10 and 11.

#### EXPERIMENTS WITH CETYLTRIMETHYLAMMONIUM BROMIDE

It was desirable to have a direct comparison of the effects of a long chain tetraalkylammonium ion (on the absorption of light by solutions of Brom Cresol Green) with those of the symmetrical tetramethylammonium and tetraethylammonium ions. In preliminary experiments with cetyltrimethylammonium bromide in the presence of brom cresol green, precipitates were noted in a number of the mixtures, particularly those which contained relatively small amounts of the quaternary ammonium salt. Closer observation of the various mixtures revealed definite Tyndall effects in others. The Tyndall effect could be much reduced (removed?) by centrifugation in an International Clinical centrifuge for twenty minutes at 3000 RPM. Accordingly, an aliquot of each mixture was centrifuged and the absorption spectrum of the supernatant measured. In no case was a Tyndall effect observed after centrifugation. The absorption spectrum of an uncentrifuged aliquot was also measured when obvious precipitation was not too great, to determine

THE EFFECT OF TETRAALKYLAMMONIUM  
ION ON THE ABSORPTION OF LIGHT BY  
BASIC SOLUTIONS OF BROM CRESOL GREEN

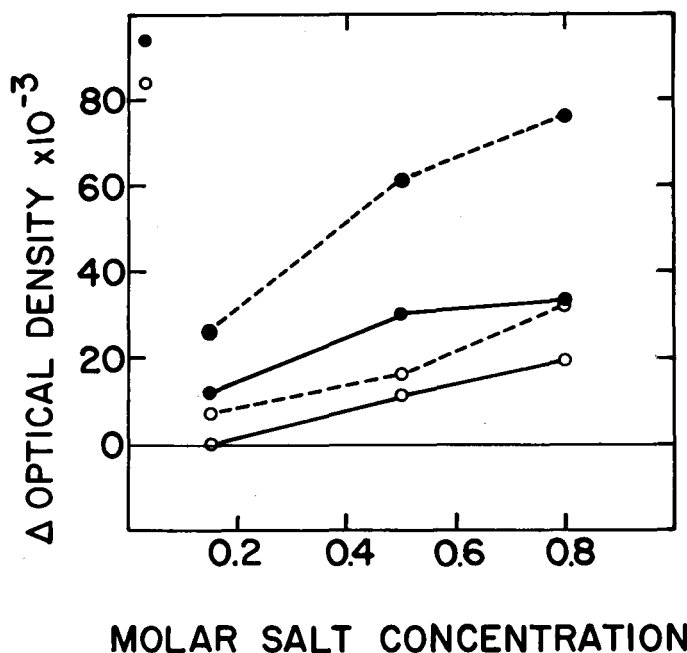


Figure 10

The solutions contained identical aliquots of brom cresol green  
in 0.01 M  $\text{Na}_3\text{PO}_4$

— Density in  $\text{Et}_4\text{NBr}$  minus density in  $\text{NaBr}$  of the same concentration  
o Density in  $\text{Me}_4\text{NBr}$  minus density in  $\text{NaBr}$  of the same concentration

— =  $16,250 \text{ cm}^{-1}$  (6154Å) The frequency of maximum absorption  
in dilute aqueous solutions

- - - =  $15,750 \text{ cm}^{-1}$  (634.9Å) The frequency of maximum effect of  $\text{R}_4\text{N}^+$



# THE EFFECT OF TETRAALKYLAMMONIUM ION ON THE ABSORPTION OF LIGHT BY ACID SOLUTIONS OF BROM CRESOL GREEN

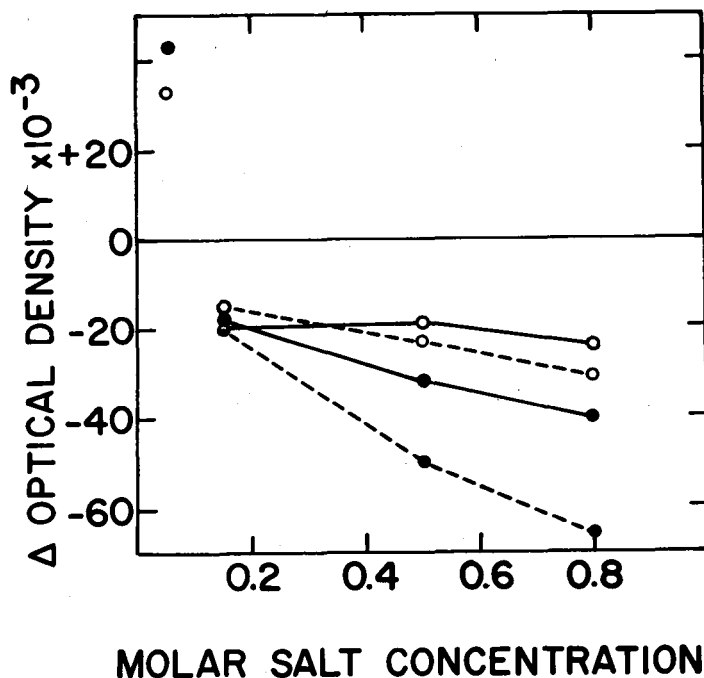


Figure 11

The solutions contained identical aliquots of brom cresol green in 0.01 M HCl

- Density in  $\text{Et}_4\text{NBr}$  minus density in  $\text{NaBr}$  of the same concentration
- o Density in  $\text{Me}_4\text{NBr}$  minus density in  $\text{NaBr}$  of the same concentration
- \_\_\_\_\_ =  $22,500\text{cm}^{-1}$  ( $4444\text{\AA}$ ) the frequency of maximum absorption in dilute aqueous solution
- - - =  $21,000\text{cm}^{-1}$  ( $4762\text{\AA}$ ) the frequency of maximum effect of  $\text{R}_4\text{N}^+$

what observations would have been reported if the precipitate present had been ignored. The results of these experiments are collected in Tables VIII and IX.

TABLE VIII

EFFECTS OF CETYLTRIMETHYLAMMONIUM BROMIDE ON THE ABSORPTION OF LIGHT BY ACID SOLUTIONS OF BROM CRESOL GREEN\*

Quaternary Salt, mols per liter times $10^4$	Initial Mixture Tyndall Effect	Optical Density 4444A <sup>o</sup>	Supernatant After Centrifugation Wavelength of Maximum Absorption	Optical Density at Max.	Optical Density 4444A <sup>o</sup>
zero	none	0.563	4444A <sup>o</sup>	0.563	0.563
0.010	v. small	0.557	4444	0.547	0.547
0.037	medium	0.552	4444	0.498	0.498
0.073	medium	0.513	4444	0.460	0.460
0.10	large	not read	4444	0.392	0.392
0.20	large	not read	4444	0.242	0.242
1.0	large	not read	4396	0.020	0.018
6.0	none	0.400	4211	0.460	0.400
10.	none	0.402	4211	0.475	0.402

\* Indicator  $7.3 \times 10^{-6}$  M. in 0.02M HCl

TABLE IX

EFFECTS OF CETYLTRIMETHYLAMMONIUM BROMIDE ON THE ABSORPTION OF LIGHT BY BASIC SOLUTIONS OF BROM CRESOL GREEN\*

Quaternary Salt, mols per liter times $10^4$	Initial Mixture Tyndall Effect	Optical Density 6154A <sup>o</sup>	Supernatant After Centrifugation Wavelength of Maximum Absorption	Optical Density at Max.	Optical Density 6154A <sup>o</sup>
zero	none	0.673	6154A <sup>o</sup>	0.673	0.673
0.010	none	0.644	6154	0.617	0.617
0.037	none	0.653	6154	0.683	0.683
0.073	none	0.660	6154	0.624	0.624
0.10	small	0.622	6154	0.607	0.607
0.20	medium	0.607	6154	0.546	0.546
1.0	large	0.378	6154	0.076	0.076
6.0	none	0.602	6250	0.618	0.602
10.0	none	0.613	6250	0.634	0.613

\* Indicator  $3.7 \times 10^{-6}$  M. in 0.01M Na<sub>3</sub>PO<sub>4</sub>.

## CHAPTER IV

### DISCUSSION

The data contained in this thesis show clearly that both tetramethylammonium ion and tetraethylammonium ion measurably affect the absorption spectra of various sulfonphthalein indicator ions in aqueous solution. The effects are small compared with the total effect of tetraalkylammonium ions which contain longer carbon chains, but are not complicated by the appearance of a second phase in the system, and may therefore be more revealing if adequately studied.

A simple explanation of the observed changes in the absorption of these indicator ions in the presence of tetraalkylammonium ions is that a complex is formed between the negative indicator ion and the positive tetraalkylammonium ion. Benesi and Hildebrand (1949) developed a spectrophotometric method for the detection of complexes and for the measurement of the equilibrium constant of complex formation. McConnell and Davidson (1950) applied this method to the detection of metal-ligand complexes of the type MA. They used the expression:

$$\left( \frac{ab}{(D - D')} \right) = \frac{a}{(E_1 - E_0)} + \frac{1}{(E_1 - E_0) K_1}$$

where a and b are the total concentrations of the ligand and the metal respectively. D is the optical density of the solution

containing both metal and ligand,  $D'$  is the optical density of a solution which contains the same amount of metal ion per liter, but no ligand, and  $K_1$  is the equilibrium constant for the formation of the complex:

$$K_1 = \frac{(MA)}{(M)(A)} \quad \checkmark$$

where the bracketed quantities indicate concentrations at a given ionic strength. The quantities  $E_0$  and  $E_1$  are the molar extinction coefficients of the metal ion and the complex. If the system is such that the only light absorbing species present are the metal ion and the complex indicated, a plot of  $ab/(D - D')$  versus  $\textcircled{a}$  will be linear. If the experimental results so plotted are linear,  $E_1 - E_0$  can be determined from the slope of the line, and  $K_1$  from the y intercept and the slope of the line.

This method was applied to the interpretation of the measurements made on solutions of brom cresol green with varying concentrations of tetraethylammonium bromide. In this case "b" represents the total concentration of indicator species, and "a" that of tetraethylammonium ion. Figure 12 shows the graphs of the data for measurements at  $4444\text{\AA}$  for the acid species and at  $6154\text{\AA}$  for the basic species. It is clear from this graph that only one complex is formed in each case. Table X shows the equilibrium constants derived from these and similar plots of measurements made on the same solutions at different wavelengths. It is clear that the numerical evaluation of these constants is not as precise as would be desirable, and it is necessary to determine the reason for

TABLE X

## EQUILIBRIUM CONSTANTS OF

## BROM CRESOL GREEN-TETRAETHYLAMMONIUM ION COMPLEX FORMATION

Tests Conditions: 0.8 M salt solutions used

## BASIC

Wavelength	$\Delta$ Molar Absorbance	$K_{eq}$	Average
6667	8.8 x $10^3$	0.30	
6349	21.0 x $10^3$	0.94	
6154	10.0 x $10^3$	0.91	0.71

## ACID

4762	9.1 x $10^3$	0.94	
4546	7.3 x $10^3$	0.57	
4444	5.5 x $10^3$	0.35	0.62

this lack of precision. In Figure 12, the experimental points for the measurements lie on straight lines. However, the function plotted ;  $ab/(D - D')$  is extremely sensitive to small errors in the measurement of both D and D', because  $(D - D')$  is so small. This graph also shows the effects of errors of 0.002 density units, and of 0.005 density units upon the values of the function plotted in this case. The limits shown are those of the worst conditions--the indicated errors add to increase (or decrease) the factor  $(D - D')$  by twice the assumed error. It is clear from this graph that no interpretation of the data could be made if an error of 0.005 O.D. units had been made. This justifies the care with which the solutions were prepared from identical aliquots of the same solution of indicator. It is clear from this graph that the linear character of the plot would not be changed greatly by errors as large as \_ 0.004 O.D. units in the density difference factor--and

DETERMINATION OF THE EQUILIBRIUM CONSTANT  
OF THE 1:1 COMPLEX BETWEEN BROM CRESOL GREEN  
AND TETRAETHYLAMMONIUM ION

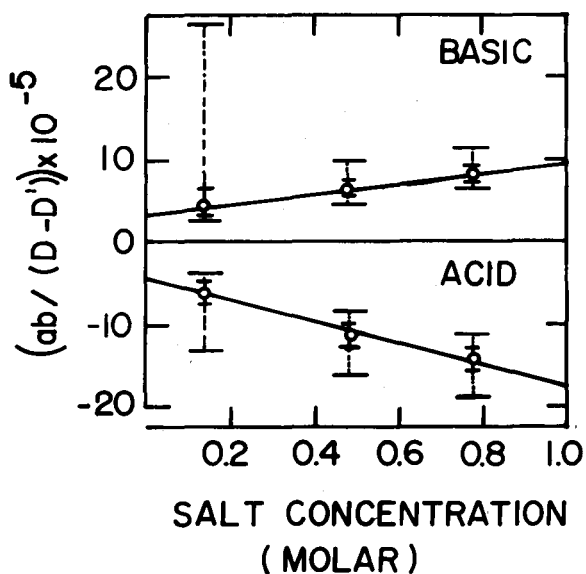


Figure 12

o observed value

— assumed error of  $\pm 0.002$  optical density units

- - - assumed error of  $\pm 0.005$  optical density units

observations at 4444Å (acid) and 6154Å (basic)

it is equally clear from the same Figure that the actual errors were smaller and, in fact, approach the limit of resolution of the spectrophotometer itself. In spite of this care, however, the measured density differences are so small that the exact value of the equilibrium constant remains inaccessible. This is a property of the molecular system, not of the method of measurement.

The conclusions which can be drawn from this analysis are not without interest. First, there is no doubt that a one to one complex is formed between tetraethylammonium ion and brom cresol green in aqueous solution. Complexes involving quaternary ammonium ions are not often encountered, and in many systems these ions are used as reference "non complexing" positive ions (Melchior, 1954). If both the demonstrated complexes are "ion pairs", which seems probable, the statement of Popov and Humphrey (1959) "-- it seems that ion-pair dissociation or association does not seem to influence the absorption spectra of solutions to any significant degree, provided that the attraction is purely electrostatic." must be considered. This statement is based upon measurements made on solutions of tetraphenylarsonium chloride in solvents of varying dielectric constants--in some of which the dissociation of the salt was small. The molar absorptivities of solutions of tetraphenylarsonium chloride at 2650Å changed from 2780 in water to 2850 in 70% dioxane to 3040 in 95% dioxane-water. It is 2830 in acetonitrile and 3150 in ethylene dichloride. The authors state that "there is no apparent correlation between the values of molar

absorptivity and the ion pair dissociation constant." and conclude that the observed differences are more likely due to changes "in the refractive index of the medium."

The data cited above do show that there is no simple one to one correspondence between the intensity of absorption at a particular wavelength and the formation of ion pairs. However, it does not show that the formation of ion pairs does not affect the absorption spectrum. If anything, it shows the opposite. If we consider the series water, and water-dioxane mixtures, the intensity of absorption increases with the amount of dioxane: 2780, 2850, 3040 as dioxane increases from zero to 70% and then 95%. The amount of change is largest between 70% and 95% -- which is in agreement with the fact that the amount of actual association at  $3 \times 10^{-4}$  molar salt calculated from the constants given in Table VII of Popov and Humphrey is very small in 70% dioxane and very large in 95% dioxane.

It is therefore possible to recognize the interpretive difficulties encountered by these authors, while rejecting their sweeping conclusions concerning the lack of effect of ion pair formation upon the absorption of light by one of the participating ions.

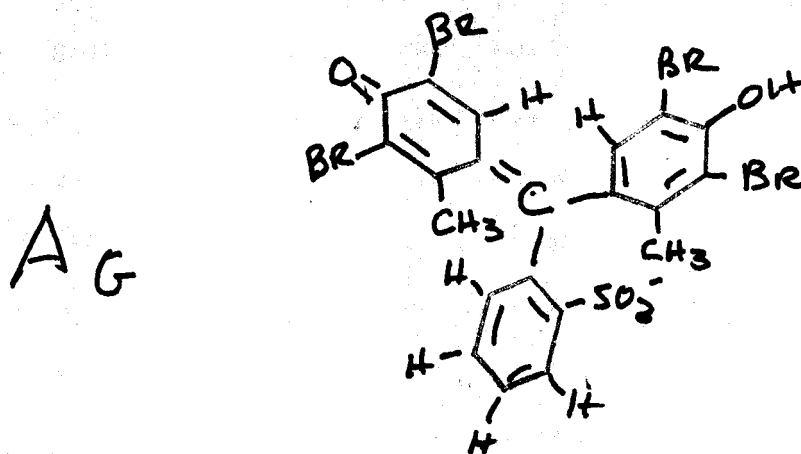
It is necessary to consider how the formation of a complex between tetraethylammonium ion and the two brom cresol green ions studied could affect the absorption spectra as has been observed. First, it is probable that the association is of the



"ion-pair" type, that is, there is no contribution of new covalent bonding. It is recognized that this cannot be proven, but is the summation of present day knowledge of chemical bonding in a large number of compounds.

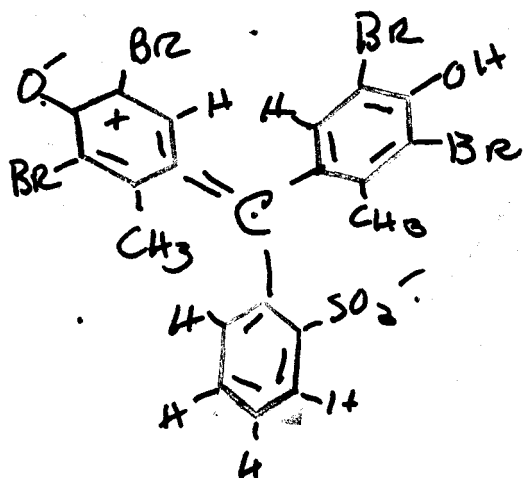
How, then, can the association with this large positive ion affect the absorption of light by either of the brom cresol green ions-- and affect the two quite differently? To discuss this it is necessary to know the structure of the absorbing species and the way in which the absorption is changed by this interaction.

Brom Cresol Green (3', 3'', 5', 5'' -tetrabromo-m-cresol-sulfonphthalein) exists in acid aqueous solution mainly as the hybrid of four Kekule structures of which one is shown below:

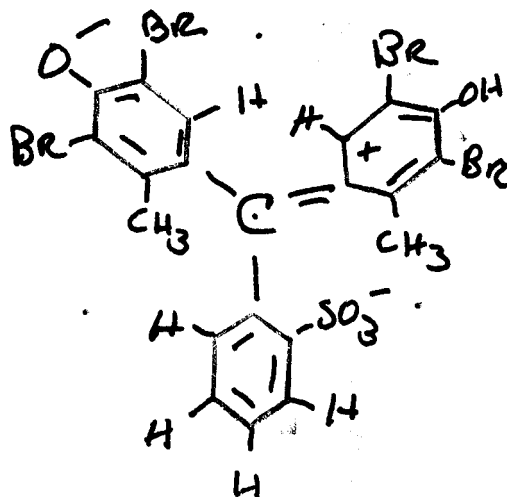


This hybrid is the ground state of the absorption of light by this substance in acid solution. The upper state of each of the absorption bands is less clearly understood, but that of the lowest energy (frequency) is presumably a charge-separation form involving

structures similar to those shown as  $A_u$



$A_u$

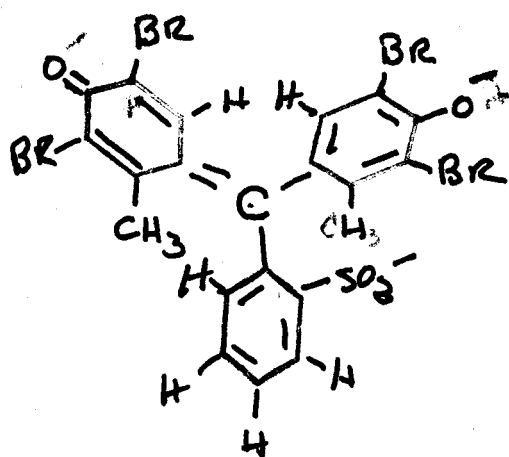


The difference in energy between these two states is measured by the frequency of the light absorbed:  $E_U - E_G = \text{constant times frequency}$ . The negative charge of the acid species is localized in the sulfonate group, and this group is presumably closely approached by the positively charged tetraethylammonium ion. Molecular models of these substances show that close approach to the sulfonate by an ion of this size involves equally close approach to at least one of the nonsulfonated rings of the indicator ion,

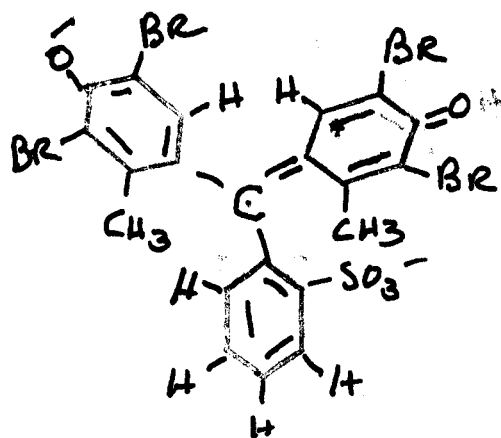
Since the positive center is closer to the benzenoid rings than to the terminal oxygens of these rings, this juxtapositioning would favor forms with negative charges on the benzenoid carbons rather than forms with positive charges. Since the major forms in the upper state involve resonance forms which have positively charged benzenoid forms as major contributors, the

interference of the tetraalkylammonium ion would be more effective in the upper state than in the ground state. That is, the increase in internal energy of the upper state would be greater than that which occurs in the ground state--therefore the value of  $E_U - E_G$  would be increased in the presence of tetraalkylammonium ion and the frequency of the light absorbed would be expected to be higher --which is in accordance with the decreased absorbance at low frequency observed in acid solutions of sulfonphthaleins in the presence of tetraalkylammonium ions.

A similar analysis should hold for the observations in basic solutions. The major resonance forms in the ground state of these solutions have a negatively charged phenolic oxygen: these solutions have a negatively charged phenolic oxygen:



B<sub>G</sub>



In addition to the eight major canonical forms, there are numerous forms in which the negative charge resides on the carbon atoms of the benzene rings--all these forms would be stabilized in the vicinity of a positive charge, therefore their contribution would

be increased and the energy of the hybrid decreased in the presence of tetraalkylammonium ions. The upper state of this type of compound involves a greater oscillation of the negative charge through the two benzenoid structures. The interaction of with a local positive charge would be greater than that of the lower state, consequently the lowering of energy would be greater. The value of  $E_U - E_G$  for the di-negative ion of sulfonphthaleins would be reduced in the presence of tetraalkylammonium ions, therefore the frequency of the light absorbed would be expected to decrease. This is also in accord with the observed increase in the absorption of lower frequency light by these di-negative ions in the presence of tetraalkylammonium ions.

A number of points require consideration. First, why is this effect not obtained with sodium ion? Experimentally the absorption spectra of sulfonphthalein ions in dilute buffer is not significantly changed by the addition of sodium bromide in concentrations from 0.01 to 0.80 molar. The answer is to be found in two factors--that of ion size and the effect of hydration. Sodium ion has a crystal radius of 0.95 to 0.98 Angstrom units, and although the hydrated ion is considerably larger this increase in size is gained at the expense of a considerable decrease in effective charge because of the polarization of the water involved in the hydration shell. Thus, even if sodium ion did form an ion pair with the sulfonate portion of a sulfonphthalein ion, the positive charge near the unsulfonated benzene rings would be effectively nil,

being damped out by the water which intervenes. Tetraethylammonium ion is quite large and the dielectric effect of the hydrocarbon portion is negligible so that the positive charge at the center is not damped out by polarization because there is no intervening water or other large dielectric. Tetramethylammonium ion is somewhat smaller than tetraethylammonium ion, and does not extend so close to the non-sulfonated benzene rings of sulfonphthaleins when it is close to the sulfonate group--in the "ion-pair" position. In this case one might reasonably expect smaller effects of the same kind as those observed in the presence of tetraethylammonium ion. This is what is observed as shown in Figure 9. Because the effects were so much smaller it was not possible to determine an acceptably accurate association constant for the ion-pair complex, but the reduced net effect at equal concentrations of tetraalkylammonium ion is in harmony with prediction--either less complex or a smaller spectral effect of the same amount of complex.

The experiments with cetyltrimethylammonium ion were essentially exploratory, but the results were sufficiently revealing to justify some consideration. The first provable interaction of low concentrations of cetyltrimethylammonium ion with the mononegative ion of brom cresol green was the formation of an insoluble compound. Table VIII shows that a Tyndall effect was observed at concentrations as low as  $1 \times 10^{-6}$  molar quaternary ion in the presence of  $7 \times 10^{-6}$  molar indicator. When the precipitate was removed by centrifugation, the position of maximum absorption was

unchanged in the supernatant solution. At higher concentrations of the quaternary ammonium ion, the solutions remained clear, no precipitate could be separated in the centrifuge, and the absorption maximum was shifted toward higher energies--higher frequencies. There is no doubt that the species present in these solutions involve more than one quaternary ion per indicator ion. The fact that the frequency change is in the same direction as that due to the smaller quaternary ammonium ions studied may indicate that the cause of the spectra shift is basically the same, but a direct comparison is not possible. In basic solutions, precipitates appeared to remove significant amounts of dye only at higher concentrations. This is probably due to the fact that two quaternary ions are required per sulfonphthalein. It may be significant that the same total concentration of cetyltrimethylammonium ion ( $1 \times 10^{-4}M$ ) is required to remove most of the indicator from solution. It is significant that the maximum observed in these solution of the di-negative ion of brom cresol green is not significantly changed until a sufficient excess of quaternary is present to redissolve the two to one compound which can be separated at lower concentrations.

It is not the purpose of this thesis to discuss the effect of the formation of micelles on the absorption spectra of these dyes--in fact, all the measurements reported are below what Zutrauen and Ter Minassian call "la concentration critique micellaire".

## SUMMARY

It has been shown that moderate concentrations of tetraethylammonium ion have a measureable effect on light absorption by the mononegative and the di-negative ions of several representative sulfonphthalein indicators. In the case of brom cresol green, the interaction has been shown to be the formation of one to one complex between the indicator ion and the quaternary ion. It has been shown that the increase in absorption at low frequency (long wavelength) of the di-negative indicator in the presence of the quaternary ion is predictable on the basis of the molecular and energy properties of the system. These same properties provide an equally satisfactory explanation for the opposite effect of tetraethylammonium ion upon the absorption of light by the mononegative ion of the same indicator--decreased absorption at low frequency (long wavelength), as well as for the reduced effects of tetramethylammonium ion and the lack of observable effect of sodium ion. Exploratory experiments with cetyltrimethylammonium ion show that its interactions with brom cresol green are too complex for proper comparison.

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APPROVAL SHEET

The thesis submitted by Irene E. Marsol has been read and approved by three members of the faculty of the Graduate School.

The final copies have been examined by the director of the thesis and the signature which appears below verifies the fact that any necessary changes have been incorporated, and that the thesis is now given final approval with reference to content, form, and mechanical accuracy.

The thesis is therefore accepted in partial fulfillment of the requirements for the Degree of Master of Science.

Jan 25, 1961  
Date

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Signature of Adviser