1961

Reactions of iodine and iodide ions in the presence and absence of polysaccharides

John Albert Effenberger
Iowa State University

Recommended Citation

Effenberger, John Albert, "Reactions of iodine and iodide ions in the presence and absence of polysaccharides " (1961). *Retrospective Theses and Dissertations*. 2431.
https://lib.dr.iastate.edu/rtd/2431

REACTIONS OF IODINE AND IODIDE IONS IN THE PRESENCE AND ABSENCE OF POLYSACCHARIDES

by

John Albert Effenberger

A Dissertation Submitted to the
Graduate Faculty in Partial Fulfillment of
The Requirements for the Degree of
DOCTOR OF PHILOSOPHY

Major Subject: Physical Chemistry

Approved:

Signature was redacted for privacy.

In Charge of Major Work

Signature was redacted for privacy.

Head of Major Department

Signature was redacted for privacy.

Dean of Graduate College

Iowa State University
Of Science and Technology
Ames, Iowa

1961

TABLE OF CONTENTS

Page

INTRODUCTION

That iodine and iodide ions interact with each other in aqueous solutions has been known for more than a century, and systematic studies of the reaction have appeared in the literature from time to time during the past sixty years. In spite of this, it is not yet certain that the most simple interaction in these solutions has been satisfactorily described from a quantitative point of view. This reaction leads to the formation of the triiodide ion, I_3^-, which establishes an equilibrium with free iodine and iodide ion according to the equation:

$$I_2\ (aq) + I^-\ (aq) \rightleftharpoons I_3^-\ (aq)\ . \qquad \text{(Eq. 1)}$$

The earliest investigators were quantitatively accurate in their descriptions of this reaction for only the most dilute solutions of iodine in dilute aqueous solutions of iodide ion. The apparent variation in the formation constant of the triiodide ion in more concentrated solutions of reactants seemed rather puzzling, and is still an enigma only partially resolved.

The first explanation offered to account for the variation was that in moderately concentrated solutions, the interactions between iodine and iodide ions were not confined to the triiodide stage, but that higher order complex ions were produced in solution. The experimental data elaborating upon this suggestion are inconsistent in part, and in general relate to the solid state rather than to aqueous solutions. Actual investigation of the species which exist in solutions has been somewhat neglected due to the tendency to assume that they contain the same complexes found in the solid state.

One of the difficulties which is encountered in an investigation of these solutions is the regrettable lack of knowledge concerning activity coefficients for those complexes which may be present. This fact was recognized in a primitive way by even the earliest investigators who continually referred to the necessity for assuming equal degrees of dissociation for the iodide and polyiodide salts. Indeed, at one time an attempt was made to explain the behavior of concentrated solutions of iodine in iodide in terms of merely activity corrections. There is, however, ample evidence at the present time to show that the observed effects would be exceedingly difficult to handle in such a manner.

An even more fundamental difficulty is encountered, however, inasmuch as the identities of those complexes other than triiodide which might exist in aqueous solutions are unknown. Until the nature of these complexes is established, very little progress can be made toward a quantitatively satisfactory description of such solutions. In the light of a few excellent accounts of pentaiodides, heptaiodides, and even enneaiodides in the literature, it has been tacitly assumed that the ions which exist in solution are of the same general type, namely I^-_{2n+1}. However, there have been occasional references to the possible existence of ions of the type, $I^=_{2n+2}$, as well. It is the primary purpose of the present research to identify those ions of either type which may be present in moderately concentrated solutions of iodide ion over the entire solubility range of iodine.

One of the most fascinating polyiodide-containing systems is the so-called starch-iodine complex. The reaction of starch with iodine to form an intensely colored product has actually been under investigation even longer than the reaction of iodine with iodide in the absence of starch. Nonetheless, due to the scant knowledge of the true nature of either starch or aqueous solutions containing iodine and iodide, there is still very little that may be said with certainty about the nature of the starch-iodine reaction. The literature is replete with fragmentary and contradictory accounts of the reaction. Since the starch-iodine reaction appears to lead to the formation of polyiodides of a distinctly different nature from those formed in aqueous solutions of iodine and iodide alone, as judged by the markedly different light absorption in the visible region of the spectrum, it is also the objective of the present investigation to elaborate upon these differences.

REVIEW OF LITERATURE

Formation of Polyiodides

Shortly after the discovery of iodine in 1811 by Courtois, the observation was made by Gay-Lussac (1) that the presence of KI greatly enhanced the solubility of I_2 in aqueous solutions. Due to the marked color change which accompanied this phenomenon, he suggested that a real chemical reaction had occurred between I_2 and KI in solution.

For the next fifty years, this observation remained in obscurity; however, in 1860, the question was first raised by Baudrimont (2) as to whether or not a "true chemical bonding" existed between the I_2 and KI in aqueous solutions. He had found it possible to completely extract I_2 from its solutions in aqueous KI by merely shaking with carbon disulfide. Thus, he concluded that no "true chemical bonding" existed. The same conclusion was reached by Dossioss and Weith (3) who showed that bubbling air through a solution of I_2 in KI resulted in complete removal of the I_2.

The entire question took a new turn when in 1877 Johnson (4) was able to crystallize a material from exceedingly concentrated solutions of I_2 in aqueous KI which satisfied the empirical formula, KI_3. The volume changes which occurred upon dissolving the material suggested that it was not merely KI in loose combination with I_2, but was a true triiodide salt. An effort was made by Wells and Wheeler (5) in 1892 to prepare triiodides of potassium, cesium, and rubidium. Some physical properties including the melting points, solubility characteristics, and descriptions of crystal types were tabulated by these investigators. They also prepared a few of the mixed trihalide salts and the pentaiodide of cesium.

In spite of the unquestionable existence of polyiodides in the solid state, the existence of an I_2 - KI compound in solution could still be questioned. Le Blanc and Noyes (6) in 1890 performed a series of cryoscopic measurements upon solutions of KI containing various amounts of dissolved I_2. The observed failure of substantial amounts of I_2 to depress the freezing point to the extent expected if no interaction had occurred led clearly to the conclusion that the solute was associated to some degree. These investigators also performed a conductometric experiment which led them to believe that there existed in solution an equilibrium between free I_2, free I^-, and some unidentified complex containing both I_2 and I^-.

Confident of the existence of triiodide in the solid state, Jakowkin (7) set out to prove its existence in solution. He devised a technique for examining the equilibrium between I_2 and I^- to form I_3^- in aqueous solutions. His now well known distribution technique consisted of allowing iodine to distribute itself between an aqueous phase containing KI and a second liquid phase in which I_2 was soluble, but I^- and

I_3^- were not. This allowed the determination of the free I_2 in the aqueous phase at equilibrium since it is related to the experimentally determined concentration in the second liquid phase through a distribution coefficient. Jakowhin used carbon disulfide as his immiscible liquid phase and determined a value for the dissociation constant of KI_3 of 1.67×10^{-3} at 25°C. In a later study, Jakowkin (8) discovered a systematic variation of the distribution coefficient with I_2 concentration. Attributing this to iodine-organic solvent interaction, he proceeded to recalculate the dissociation constant for more appropriate values of the distribution coefficient. The new value was 1.4×10^{-3} which is not unlike the value obtained in recent more critical determinations.

However, this value seemed to be a true constant only for rather dilute solutions of I_2 and KI. The value of the equilibrium constant appeared to vary systematically with the I_2 concentration, and although the effect was barely noticeable at the lowest I^- concentrations, it did become increasingly apparent at higher I^- concentrations. It was easily observed in solutions only 0.1 <u>M</u> in KI. Jakowkin attributed this behavior to the formation of higher polyiodides at higher concentrations and even suggested that $I_4^=$ might be the next ion of significance.

In studying the effect that the cation had on the equilibrium constant, Jakowkin (9) found that the value for the constant was essentially the same with Li^+, Na^+, K^+, and Ba^{++} indicating that the equilibrium really did involve an I_3^- ion rather than its undissociated salt. A somewhat lower value was found for HI_3.

Several investigations, notably those of Noyes and Seidensticker (10) and Fedotieff (11), showed that in dilute solutions of I^- the solubility of I_2 could be explained on the assumption that only I_3^- and no higher complex was formed. Korenmann (12) has also shown this in a very recent investigation of the polyhalide salts. Korenmann obtained a value for the dissociation constant of I_3^- of 1.33×10^{-3} at 25°C. At moderate I^- concentrations, however, the solubility of I_2 is considerably more than may be accounted for by the free I_2 and I_3^- only.

Following the suggestion that the higher polyiodides must surely exist, attempts were made to prepare these compounds. It had been reported by Johnson (4) that KI_3 was extremely difficult to obtain in the solid state due to its hygroscopic nature. It could be expected that crystallization of higher polyiodides would be an even more formidable task.

Abegg and Hamburger (13) made a phase study of $KI - I_2$ mixtures in saturated aqueous solutions which had been equilibrated with benzene. Only the I_2 was soluble in the benzene phase and consequently they were able to follow the free iodine concentration of the aqueous phase by means of the distribution coefficient. According to the phase rule, as long as only two solid phases are present, the proportions of these solids may be varied continuously without changing the composition of the saturated solution. However, if sufficient excess of one of the solids has been added to react with all of the other and causes its dissolution, the system will gain a degree of freedom. Thus, the composition of the solution will vary until it becomes saturated with another I_2 - KI compound. One may ascertain the composition of the solids present when new phases appear by simply following the composition of the saturated solution. Abegg and Hamburger did not find KI_3, but reported a KI_7. Also reported were RbI_3, CsI_3, RbI_7, CsI_7, RbI_9, and CsI_9. Evaluation of this work is difficult since the data on the composition of the solid phases which led to their conclusions were not published. Furthermore, the actual variation in I_2 content of the benzene phase was rather small making it difficult to detect real changes in I_2 concentration. Linhart (14) later examined the solubility data of Abegg and Hamburger and found it grossly inconsistent.

However, another phase analysis of the KI - I_2 system was made by Foote and Chalker (15) in 1908 in which they analyzed both the solid and aqueous phases for I_2 and KI content. This investigation confirmed the existence of KI_3 as reported by Johnson (4) and of KI_7 as reported by Abegg and crystallographic I - I distances is given by Slater (20). A comprehensive discussion of the crystalline polyhalide ions has been given by Rundle (21) in an effort to correlate the available crystal data with molecular orbital calculations.

The pentaiodide of tetramethylammonium has been investigated by Hach and Rundle (22), and even the enneaiodide of tetramethylammonium has been subjected to crystallographic analysis by Rundle et al. (23). A crystalline compound containing the $I_8^=$ ion has also been studied by Havinga et al. (24) by means of x-ray analysis.

At about the same time that research was in progress to identify the anomalous behavior of concentrated I_2 - KI solutions with the probable existence of higher polyiodides, Lewis (25) published his now well known ideas with regard to the concept of activities. Certain investigators, notably Parsons and Corliss (26) and Parsons and Whittemore (27), then sought to explain the behavior of such solutions on the basis of activity effects alone, and even denied the existence of triiodide ion.

Dawson (28) had shown that HI_3 behaves as a strong electrolyte in the course of his determination of the dissociation constant for triiodide ion at 13.5°C. It seemed, therefore, that there could be no serious error in assuming the activity coefficients of HI_3 and HI to be equal, at least in dilute solutions. Bray and McKay (29) concluded from their investi-

Hamburger (13). They also reported the existence of the solid compounds RbI_3, CsI_3, and CsI_5. They were, however, unable to confirm the higher Rb^+ and Cs^+ polyiodides which had been previously reported by Abegg and Hamburger (13).

Evidence was also presented in 1902 by Dawson and Gawler (16) for the existence of higher polyiodides in nitrobenzene solutions. It was shown that although KI is itself insoluble in nitrobenzene, it may be extracted from its aqueous solutions by a nitrobenzene solution containing I_2. The ratio of I_2 to KI in saturated nitrobenzene solutions approached a value of four. Thus, if the existence of only one complex were to be assumed, this ratio would require it to be the enneaiodide. At any rate, no matter how many complexes are present, complexes other than those containing iodine and iodide in a 1:1 ratio would be required to explain the observed ratio. Similar results were reported by Dawson and Goodson (17) using the iodides of NH_4^+, Na^+, Li^+, Rb^+, Cs^+, Ba^{++}, and Sr^{++}. They were, however, unable to obtain any solid enneaiodides.

This early confusion as to the existence of the solid polyiodides has long been dispelled, however, and several excellent crystallographic studies by means of x-ray analysis of crystalline polyiodides are available. The triiodides of ammonium and tetraphenylarsonium have been investigated by Mooney (18, 19) and a theoretical discussion of the various gations of the triiodide equilibrium that no assumptions regarding the relative values of these activity coefficients could yield a satisfactory account for the behavior of more concentrated solutions. Furthermore, Washburn and Strachan (30) performed an experiment in which activity corrections were made from conductivity data. The results indicated that the variation of the triiodide dissociation constant could not be a simple activity effect. Thus, it seemed more realistic to pursue the initial suggestion that higher polyiodides must exist in aqueous solutions.

Linhart (31) made an attempt to describe the behavior of relatively concentrated solutions of KI saturated with I_2 in terms of two complex ions, I_3^- and $I_{14}^=$. Although this description of the saturated solutions is in fair agreement with the experimental data, it is not unlikely that as good or better agreement might be obtained assuming the existence of a greater number of smaller complex ions. Furthermore, it seems more realistic to assume that ions considerably more simple than $I_{14}^=$ are more likely to exist in these solutions. It is of some interest to note, however, that an ion of the type $I_{2n+2}^=$ was suggested by Linhart.

It is also of some interest to note that an experiment performed by Laurie (32) as long ago as 1909 had led him to believe that even in solutions up to 1 M in KI and saturated with I_2, triiodide is the only complex present. A closer examination, however, reveals that the proper interpretation would be that in such solutions a large proportion of the complex ions do contain I_2 and I^- in a 1:1 ratio. One should note that this would include $I_6^=$ as well as I_3^-.

Davies and Gwynne (33) have recently attempted to describe the behavior of moderately concentrated solutions of KI containing I_2 by assuming alternatively the existence of I_5^- or $I_6^=$ in addition to I_3^-. They have shown that $I_6^=$ is more consistent with their data, but have not explicitly eliminated the possibility that I_5^- or even other complex ions might be present. These same authors performed a series of investigations of the temperature dependence of the triiodide formation constant, and have shown that the interaction energy of I_2 and I^- is consistent with an ion-induced dipole type reaction.

In addition to the liquid-liquid distribution technique introduced by Jakowkin, another distribution technique was used by Jones and Kaplan (34). This technique involves the equilibration of iodine between two aqueous phases via the vapor phase in a specially designed equilibrator. One of the aqueous phases is iodide free; thus, the free iodine activity may be obtained by titration of this phase. The dissociation constant found for the triiodide ion by this procedure is in fair agreement with that found by Jakowkin's method.

The absorption spectrum of an I_2 - KI solution offers strong evidence for the existence of a complex in solution. The absorption spectrum of aqueous KI - I_2 solutions is characterized by two prominent maxima at about 2900 Å and 3500 Å. The intensity of the maxima depend upon both the I_2 and the KI concentrations. Neither maximum is present in I_2 or KI solutions alone. Consequently, this constitutes strong evidence that an absorbing species other than I_2 or KI is present in these solutions. The spectra of various triiodide salts have been examined by Allsopp (35) who reported that all of the salts examined possessed the two maxima at 2900 Å and 3500 Å with about the same molar absorbancy indices. It would seem plausible, therefore, to assume that these maxima are actually characteristic of the production of I_3^- from I_2 and I^- in aqueous solutions. It has also been suggested by the same author that the 6000 cm^{-1} frequency separation of these two maxima is consistent with the production of both an excited and normal iodine atom in the $^2P_{1/2}$ and $^2P_{3/2}$ states in the absorption process. Evidence that an I - I^- configuration was required to obtain these maxima was also obtained. This is entirely consistent with the view that an ion-induced dipole reaction occurs to produce a triiodide ion.

An observation due to Winther (36) and disputed by Forster (37), the significance of which has been long neglected, is that although the relative intensities of the 2900 Å and 3500 Å maxima remain constant over a wide range of

KI concentrations, at a sufficiently high KI concentration the 3500 Å maximum increases while the 2900 Å maximum decreases in intensity upon the further addition of KI. Although this observation has not been explained in the literature, it is the contention of the present author that this may be construed as evidence for the existence of $I_4^=$. Such interpretation may also be placed upon the curious reversal of the relative intensities of these two bands mentioned by Symons and Doyle (38) in their discussion of color centers in alkali halide crystals. Studies by Awtrey and Connick (39) made use of the molar absorbancy index of I_3^- at 3520 Å to determine the dissociation constant of the triiodide ion.

The only investigation of the kinetics of the triiodide equilibrium is due to Myers (40) who performed an experiment based upon the fact that the nuclear magnetic resonance absorption line of iodide ions is broadened by the presence of I_2 molecules. The broadening is interpreted as being due to the diminution of the phase-memory time of the nuclei in the iodide ions. The strong quadrupolar coupling in the triiodide ion is responsible for this diminution. From the degree of broadening caused by various I_2 concentrations, it is possible to calculate the average lifetime of an iodide ion, and from this the rate constant for the reaction. Assuming a simple bimolecular process, the rate constant for the formation of triiodide is 4.1×10^{10} l $mole^{-1}sec^{-1}$. The rate constant for the dissociation of triiodide is 7.6×10^7 sec^{-1}. These studies were performed at 35°C.

The most extensive studies of the temperature dependence of the value for the triiodide dissociation constant are due to Davies and Gwynne (33). However, the value for ΔH of the reaction found by these authors does not agree with that of Awtrey and Connick (39); furthermore, an unusually large ΔC_p for the reaction was observed. The results of these and other investigations of the dissociation constant for triiodide and its temperature dependence are tabulated in Table 1 and Table 2.

Starch-Iodine Reaction

It is well known that aqueous solutions of iodine are capable of assuming a deep blue color in the presence of starch. The complex which is recognized as being responsible for this pronounced absorption of red light has been the subject of sporadically intense investigation for nearly one hundred and fifty years. Nonetheless, as in the case of the formation of polyiodine complexes in the absence of starch, the nature of the reaction which leads to the formation of this blue complex is rather imperfectly understood.

The apparent contradictions and confusion in the earliest literature on the starch-iodine reaction may be attributed to

Table 1. Significant values for the formation constant, K_3, of triiodide ion

K_3	t(°C)	Reference	Remarks
1406	0.0	34	Vapor phase distribution; hydrolysis[a]
1408	0.0	41	Hydrolysis repressed
1529[b]	1.0	39	Spectrophotometric determination
985	13.5	28	Conductometric; hydrolysis repressed
909[b]	16.1	39	Spectrophotometric determination
715	25.0	8	Early value; hydrolysis[a]
769	25.0	30	Dissociation of HI_3
714	25.0	34	Vapor phase distribution; hydrolysis[a]
752	25.0	12	Solubility data on dilute I^- solutions
768	25.0	33	Extrapolated value
571[b]	33.4	39	Spectrophotometric determination
590	38.38	33	Extrapolated value
481[b]	39.2	39	Spectrophotometric determination
490	49.65	33	Extrapolated value
411	63.05	33	Extrapolated value

[a] No attempt was made to prevent the formation of I^- by hydrolysis of I_2.

[b] These values are based upon an assumed value of 714 at 25°C.

Table 2. Summary of values for ΔH_3 obtained from temperature variation of K_3[a]

ΔH_3 (cal)	t(^oC)[b]	Reference	Remarks
5100	--	39	Based upon assumed K_3 at 25^oC[c]
4304	12.5	34	From K_3 at 0^oC and 25^oC
3645	31.69	33	From K_3 at 25^oC and 38.38^oC
3272	44.01	33	From K_3 at 38.38^oC and 49.65^oC
2815	56.35	33	From K_3 at 49.65^oC and 63.05^oC

[a] The enthalpy and equilibrium constant are for the formation of triiodide ion from aqueous I_2 and aqueous I^-.

[b] The temperature given is the mean of the temperatures for which K_3 has been determined.

[c] This is an average value for ΔH_3 over the temperature range 0.0^oC to 39.2^oC.

the hopelessly inadequate state of knowledge concerning the nature of starch itself at the time. It is really only in relatively modern times that starch has been identified as a mixture of two essentially different kinds of high polymeric substances whose affinities for iodine differ markedly. The mixture itself is not a simple one inasmuch as its components constitute a polydisperse system of such a nature that the iodine binding capacity of these components is profoundly dependent upon the degree of polymerization (DP).

Of equally important significance is the role which molecular geometry plays in this rather unique type of complex. It has been the result of attention to such details as these and to the recognition of the importance of the slight but non-negligible hydrolysis of iodine in aqueous solutions that new and challenging concepts have been introduced in the elucidation of the starch-iodine system.

Observations by Rundle and Baldwin (42) on the dichroism of flow exhibited by solutions of the complex when subjected to a velocity gradient suggested that the complex might have a helical configuration; subsequent interpretation of x-ray data on the complex in the solid state by Rundle and French (43) left little doubt but that at least in certain crystalline modifications, an amylose helix enclosing a "polyiodine core" must be present.

Following the development by Schoch (44) of a fractionation technique by means of which starch could be separated into amylose, a linear polymer, and amylopectin, a highly branched polymer, it became possible to accumulate more meaningful quantitative data on the starch-iodine reaction. It was discovered by Baldwin <u>et al</u>. (45) in 1944 that it was really the amylose which reacted with iodine to form an intensely blue complex. The reaction of iodine with amylopectin led to the formation of a "plum-colored" complex. In 1948, Swanson (46) confirmed this observation and in addition showed that the color of even the amylose-iodine complex was a sensitive function of the degree of polymerization of the amylose, changing from an intense blue to deep red as the degree of polymerization decreased.

It was also shown by Baldwin <u>et al</u>. (45) by spectrophotometric titration that higher iodine activities were required to saturate the complex at higher iodide ion concentrations. This suggested to these authors that iodide ion might conceivably be an integral part of the polyiodine core. The fact that crystalline amylose could absorb iodine vapors to form the complex, however, made the exact role of iodide ion uncertain. Very recent evidence has been produced by Thoma and French (47) to demonstrate that iodide ion is absolutely essential for the formation of the blue complex in aqueous solutions. These investigators showed that when the hydrolysis of iodine was completely repressed, the only absorption in the visible spectrum of an amylose-iodine solution was that due to the iodine itself.

A potentiometric technique was successfully adapted to the study of the starch-iodine reaction by Bates <u>et al</u>. (48). A later modification due to Gilbert and Marriott (49) enabled the utilization of differential titration at exceedingly low concentrations of iodine. It was concluded from the data of Gilbert and Marriott that the formation of the blue complex was actually a stepwise reaction in which the ratio of bound conclusions. Nonetheless, the concept of a series of different kinds of polyiodine cores within a helix is one to be seriously considered.

Due to the eminently successful efforts of Thoma et al. (51) to fractionate the amylose series of dextrins into individual members, it became possible to explore the iodine binding capacities of the individual polysaccharides from a DP of 4 up to a DP of 18 (Thoma and French, 52). It was shown that even the very low molecular weight members did form a complex with triiodide ion. However, only maltononaose and higher members appeared to enhance appreciably the visible spectrum of the triiodide ion. Spectrophotometric evidence was also offered to indicate that some complex was formed which exhibited a maximum in absorption at wavelengths intermediate between that due to either triiodide or iodine itself. The potentiometric data presented by these authors also indicated a loop-helix transition in the conformation of the polysaccharide-triiodide complex between DP 6 and DP 7.

The significance of the present research with regard to the nature of the starch-iodine reaction will be discussed in a later section.

iodine to bound iodide was initially 2:1 and later changed to 3:2. These investigators suggested that the blue complex was actually one in which an amylose helix enclosed a linear polyiodine core whose basic repeating unit was $I_8^=$.

A most important discovery was made by Mould and Synge (50) in the course of examining the electrophoretic properties of the amylose-iodine system. These authors succeeded in separating a solution containing iodine and amylose into three components of distinctly different anionic mobility and color by means of electrophoresis in both an agar bed and a cell packed with tiny glass beads. These three fractions were colored blue, red, and orange respectively. The molecular size of the amylose in each of these three fractions was determined by means of electrokinetic ultrafiltration, and it developed that the amylose which gave a blue complex was in the DP range 40-130, the amylose which gave a red complex was in the DP range 25-40, and the amylose which gave an orange complex was in the DP range 10-25. Using the differential potentiometric technique of Gilbert and Marriot, these investigators concluded that the blue complex contained an $I_8^=$ polyiodine core as previously reported. However, the red complex appeared to contain an I_5^- polyiodine core. Due to the serious experimental uncertainties encountered in iodine titrations at the exceedingly low iodine concentrations employed, however, it is rather difficult to evaluate such

EQUILIBRIA IN AQUEOUS SOLUTIONS OF IODINE AND IODIDE

Hydrolysis of Iodine

The absorption spectrum of a freshly prepared, neutral, aqueous solution of iodine is characterized by maxima at 4600 A, 2880 Å, and 3520 Å. However, if the water used as a solvent is initially adjusted to a pH of 2 with either sulfuric or iodic acid, the only observable maximum is that which occurs at 4600 Å. Such behavior is most simply understood in terms of the hydrolysis of aqueous iodine according to the equations:

$$I_2(aq) + HOH \rightleftharpoons H_2IO^+ + I^- \qquad K = 1.2 \times 10^{-11}$$

$$I_2(aq) + HOH \rightleftharpoons HIO + I^- + H^+ \qquad K = 5.4 \times 10^{-13}$$

$$3I_2(aq) + 3HOH \rightleftharpoons IO_3^- + 5I^- + 6H^+ \qquad K = 7.1 \times 10^{-48}$$

The iodide ion thus produced in the hydrolysis then becomes available to interact with the aqueous iodine. It is this interaction which gives rise to the additional maxima in the ultraviolet absorption spectrum of aqueous iodine. The extent to which hydrolysis occurs and iodide ion is produced depends upon the pH of the solution since H^+ is one of the products of hydrolysis. Thus, in the presence of added acid, the additional maxima are not observed. The amount of iodide ion produced in neutral solutions, however, is not negligible as may be calculated from the equilibrium constants which are due to Allen and Keefer (53). In any quantitative investigations, the effects of hydrolysis can only be ignored when they have been minimized by the addition of a suitable acid. Ideally, it would be desirable to determine experimentally the iodide ion activity in aqueous solutions containing both iodine and iodide if at all possible.

Triiodide Ion Formation

The primary reaction between I_2 and I^- in aqueous solutions results in the formation of the triiodide ion, I_3^-. This complex ion is very likely due to an ion-induced dipole type of interaction between the reacting species. An equilibrium is rapidly established according to the equation

$$I_2(aq) + I^-(aq) \rightleftharpoons I_3^-(aq)$$

The thermodynamic equilibrium constant, K_3, for the formation of the triiodide ion may be expressed in terms of the molar concentrations of the species involved and the corresponding molar activity coefficients.

$$K_3 = \frac{(I_3^-)}{(I_2)(I^-)} \cdot \frac{f_{I_3^-}}{f_{I_2}\, f_{I^-}}$$

Collecting the activity coefficients into a single factor, F_3, the formation constant may be represented as follows

$$K_3 = \frac{(I_3^-)}{(I_2)(I^-)} F_3 \qquad \text{(Eq. 2)}$$

At low ionic strength, f_{I_2}, is independent of ionic strength and may be taken as unity (33); furthermore, at any given ionic strength, f_{I^-} and $f_{I_3^-}$ may be expected to be very nearly equal. Consequently, in dilute solutions of low ionic strength, one may assume as an excellent first approximation that F_3 is very nearly unity. Thus, the evaluation of K_3 is reduced to an estimation of the molar concentrations of the species involved at equilibrium.

It is convenient for the purpose of describing equilibria between I_2, I^-, and complex ions containing these species to distinguish between I_2 molecules which are intimately associated with I^- ions in a complex, and those which are not so involved. The former may be considered bound iodine, I_{2_b}, while the latter may be thought of as free iodine, I_2. The molar concentrations of these different kinds of iodine may be expressed as $(I_2)_b$ and (I_2) respectively. The sum of the free and the bound iodine concentrations is the total iodine molarity, $(I_2)_t$. Analogous quantities for expressing I^- concentrations are (I^-), $(I^-)_b$, and $(I^-)_t$ for the free, bound, and total iodide molarities respectively.

The most readily available quantities experimentally are $(I_2)_t$, (I_2), and $(I^-)_t$. In the event that the reaction between I_2 and I^- leads to the formation of I_3^- only, stoichiometry requires that $(I_2)_b$ and $(I^-)_b$ be identical; and since the difference between $(I_2)_t$ and (I_2) gives $(I_2)_b$, K_3 may be readily calculated from the following equation

$$K_3 = \left[(I_2)_t - (I_2)\right] / (I_2)\left[(I^-)_t - (I_2)_t + (I_2)\right] . \quad \text{(Eq. 3)}$$

It is this equation which has been most frequently used to evaluate K_3.

Polyiodide Ion Formation

It is well known that the numerical value of the formation constant for I_3^- as determined by means of Eq. 3 is in fact a true constant only at the lowest concentrations of I_2 and I^-. At higher I^- concentrations in particular, the "constant" so determined is observed to increase rapidly with increasing I_2 concentration. One may express Eq. 3 in terms of the concentrations of bound I_2, free I_2, and free I^- as in Eq. 4. It is then readily observed that the calculated value of K_3 is directly proportional to the concentration of the bound I_2.

$$K_3 = (I_2)_b / (I_2)(I^-) \quad \text{(Eq. 4)}$$

Thus, the steady increase in the calculated value for K_3 may be attributed to a corresponding increase in the concentration of bound I_2. This of course suggests that complex ion formation is not confined to the triiodide stage, but that additional complex ions containing I_2 and I^- are formed in aqueous solution.

This explanation has received virtually universal acceptance for two reasons. Firstly, it is a recognized fact that at sufficiently high total iodide concentration, it is possible to dissolve more than an equimolar amount of iodine. Thus, polyiodides of a type other than I_3^-, which contains iodine and iodide in equimolar quantities, would be required to satisfy the solubility data in terms of complex ion formation. Secondly, several polyiodides of the type I_{2n+1}^- such as I_5^-, I_7^-, and I_9^- have actually been crystallized from aqueous solution and subjected to x-ray analysis. Thus, the actual existence of such polyiodides in solution would appear to be but a logical extension of well established conclusions regarding the solid state.

However, attempts by Davies and Gwynne (33) to evaluate the importance of I_5^- as a polyiodide in solutions of moderate iodide concentration have led to the somewhat unexpected conclusion that a polyiodide such as $I_6^=$ would explain their observations in a much more satisfactory manner. It is therefore apparent that one cannot eliminate the possibility that polyiodides of the type $I_{2n+2}^=$ might also exist in aqueous solutions. In any event, it does seem likely that some type of polyiodide in addition to triiodide would be required to provide a mechanism for the observed increase in iodine solubility at moderately high iodide concentrations.

THEORY

Polyiodide Formation Function

One may write the following generalized equation to represent the various interactions between I_2 and I^- to form complex polyiodide ions in aqueous solutions.

$$n\ I_2 + m\ I^- \rightleftharpoons I_{2n+m}^{-m} \qquad \text{(Eq. 5)}$$

If one neglects activity coefficients or assumes that under certain conditions such corrections will simply constitute a constant multiplicative correction factor to the thermodynamic equilibrium constant, one may write the following equation for the non-thermodynamic equilibrium constant governing the above reaction.

$$K_{2n+m} = (I_{2n+m}^{-m}) \ / \ (I_2)^n (I^-)^m \qquad \text{(Eq. 6)}^a$$

Thus, the equilibrium constants for the various reactions are defined simply by the specification of particular values of n and m. Evidence has been cited for the possible existence of ions of the type I_{2n+1}^- and $I_{2n+2}^=$ in aqueous solutions. It would, therefore, seem appropriate to establish a model which included the possible existence of I_3^-, $I_4^=$, I_5^-, $I_6^=$, I_7^-, $I_8^=$, and I_9^-. Since the species $I_6^=$ is representative of the generalized $(I_3^-)_n$ ion as well as the $I_{2n+2}^=$ ion type,

---

[a] In all expressions of equilibria, the ( ) are used to indicate the molar concentration of the enclosed species.

it would seem logical to include $(I_3^-)_3$, i.e. I_9^{-3}, in the model as well as the previously mentioned ions.

The individual equilibrium constants are defined, therefore, by the following equation set.[a]

$$K_3 = (I_3^-) / (I_2)(I^-) \qquad K_7 = (I_7^-) / (I_2)^3(I^-)$$

$$K_4 = (I_4^{=}) / (I_2)(I^-)^2 \qquad K_8 = (I_8^{=}) / (I_2)^3(I^-)^2$$

$$K_5 = (I_5^-) / (I_2)^2(I^-) \qquad \bar{K}_9 = (I_9^-) / (I_2)^4(I^-)$$

$$K_6 = (I_6^{=}) / (I_2)^2(I^-)^2 \qquad K_9 = (I_9^{-3}) / (I_2)^3(I^-)^3$$

(Eq. 7)

It has already been mentioned that K_3 as expressed in Eq. 4 can only be expected to remain a true equilibrium constant if $(I_2)_b$ and (I_3^-) are identical. Consequently, if it is true that at higher concentrations of I^- the $(I_2)_b$ includes I_2 in the form of polyiodides other than I_3^-, one would expect the calculated value of K_3 to rise. Inasmuch as the quantity defined by Eq. 4 is in fact a measure of the extent to which further complex ion formation occurs, it is useful to re-define this quantity. Let us, therefore, define this quantity as the polyiodide formation function, Φ, according to the following equation.

$$\Phi = (I_2)_b / (I_2)(I^-) \qquad \text{(Eq. 8)}$$

---

[a] Since both I_9^- and I_9^{-3} have the same index for K, $2n+m = 9$, the former is symbolized as $\bar{K}_9$ and the latter as simply K_9 to avoid ambiguity.

In view of the fact that the $(I_2)_b$ may be expressed in terms of the molar concentrations of the complex ions containing I_2, as in Eq. 9, it is possible to relate Φ to the equilibrium constants defined by Eq. 7.

$$(I_2)_b = n \sum_n \sum_m (I_{2n+m}^{-m}) \qquad \text{(Eq. 9)}$$

By combining Eq. 6 and Eq. 9, and substituting into Eq. 8 the expression thus obtained for $(I_2)_b$ as a function of the molar concentrations of reactants and equilibrium constants, one obtains the following expression for the polyiodide formation function in terms of measurable quantities, (I_2) and (I^-), and the various equilibrium constants. It is, therefore, possible in principle to obtain estimates of these constants by curve-fitting.

$$\Phi = \sum_n \sum_m n\, K_{2n+m}\, (I_2)^{n-1} (I^-)^{m-1} \qquad \text{(Eq. 10)}$$

It is useful to examine Eq. 10 in its expanded form in order to observe certain useful properties of this function.

$$\begin{aligned}\Phi = {} & K_3 + K_4(I^-) + 2\, K_5(I_2) + 2\, K_6(I_2)(I^-) + 3\, K_7(I_2)^2 \\ & + 3\, K_8(I_2)^2(I^-) + 4\, \overline{K}_9(I_2)^3 + 3\, K_9(I_2)^2(I^-)^2\end{aligned} \qquad \text{(Eq. 11)}$$

One may examine the values approached by Φ as (I_2), (I^-), or both approach zero since such values are experimentally accessible. These values may be expressed by the following equations.

$$\Phi_{(I_2)\rightarrow 0} = K_3 + K_4(I^-) \qquad \text{(Eq. 12)}$$

$$\Phi_{(I^-)\rightarrow 0} = K_3 + 2\,K_5(I_2) + 3\,K_7(I_2)^2 + 4\,\overline{K}_9(I_2)^3 \qquad \text{(Eq. 13)}$$

$$\Phi_{(I_2),(I^-)\rightarrow 0} = K_3 \qquad \text{(Eq. 14)}$$

The relationships expressed in Eq. 12, Eq. 13, and Eq. 14 will allow a simplification of the model in terms of the ionic species which must be considered if such simplification is at all possible. Thus, for example, one would include $I_4^=$ in a model only if $\Phi_{(I_2)\rightarrow 0}$ did in fact prove to be a linear function of (I^-) as predicted by Eq. 12. The usefulness of these properties of Φ in obtaining preliminary estimates of the various constants by graphical means is another attractive feature of this function.

Distribution Law

If to a system of two immiscible liquids a third substance is added which is itself soluble in both liquids, the substance is found to distribute itself between the two liquid phases in a definite manner. An exact mathematical expression of this statement may be derived from a consideration of the requirement that the chemical potential, U, of a substance distributed between two phases must be identical in each phase at equilibrium. Thus, in the first liquid phase, the chemical potential may be expressed by

$$U_1 = U_1^o + RT \ln a_1 \qquad \text{(Eq. 15)}$$

and in the second liquid phase, the chemical potential is similarly given by

$$U_2 = U_2^o + RT \ln a_2 \qquad \text{(Eq. 16)}$$

where U_1^o and U_2^o are the chemical potentials of the solute in the standard state and are, therefore, constants at any given temperature. The activities of the solute in the two phases are given by a_1 and a_2 respectively. Since at equilibrium $U_1 = U_2$, it follows that at any given temperature and pressure

$$a_1 \;/\; a_2 = \text{constant} \qquad \text{(Eq. 17)}$$

This exact expression of the distribution law may be put into an approximate form for practical purposes. If the solution behaves ideally in the sense that either Henry's Law or Raoult's Law may be applied, then the activities may be replaced by the appropriate mole fractions. Furthermore, if the solutions are dilute, the ratio of the mole fractions may be approximated by the ratio of the concentrations expressed in moles per liter of solution. Thus, the distribution law may be expressed as in Eq. 18.

$$c_1 \;/\; c_2 = \text{constant} = K \qquad \text{(Eq. 18)}$$

This form of the distribution law is based upon the assumptions of dilute solution and-ideal behavior; thus, as one of the phases becomes non-ideal in this respect, the experimental value of K will be observed to change. If one knows the value of K at any given concentration of solute in one of the phases, therefore, it is possible to calculate the concentration of the solute in the other phase at equilibrium.

Thus the distribution law may be applied to the partitioning of I_2 between an aqueous phase containing I^- and a CCl_4 phase in which the I_2 is also soluble. Since I^- and polyiodide complex ions are not soluble in CCl_4, one may estimate the free I_2 concentration in the aqueous phase from knowledge of the distribution coefficient of I_2 for this system even though considerable I_2 may be present in the form of complex ions.

Potentiometry

The reduction occurring at the reversible iodine electrode may be written as

$$I_2 + 2\ e^- \rightleftharpoons 2\ I^- \qquad \text{(Eq. 19)}$$

The electrode potential, E, due to this process may be expressed by the Nernst equation as follows.

$$E = E^o - \frac{RT}{2F} \ln\ (I^-)^2\ f_{I^-}^2\ /\ (I_2)\ f_{I_2} \qquad \text{(Eq. 20)}$$

where, R is the universal gas constant,

T is the absolute temperature,

F is the Faraday constant,

E° is the standard reduction potential of the aqueous iodine electrode,

f_{I^-} is the molar activity coefficient of I^-, and

f_{I_2} is the molar activity coefficient of I_2.

For a concentration cell consisting of two iodine electrodes immersed in solutions brought into electrical contact by means of a salt bridge, the standard reduction potential is exactly cancelled by the standard oxidation potential in the expression for the emf of such a cell. If in addition, one may assume that the molar activity coefficients for the I_2 and I^- are the same in each half-cell, these factors will also disappear from the expression for the emf due to cancellation. Thus, the emf of such a cell may be given as a simple function of the molar concentrations of the I_2 and I^- in the two half-cells as follows.

$$E = \frac{RT}{2F} \ln (I_2)_1 \, (I^-)_2^2 \, / \, (I^-)_1^2 \, (I_2)_2 \qquad \text{(Eq. 21)}$$

where the subscripts refer to the different half-cells.

If the I_2 and I^- concentrations are known for one of the half-cells, a reference half-cell, but are unknown in the other half-cell, Eq. 21 may be rewritten as follows:

$$E = \frac{1}{w} \log Q + \frac{1}{w} \log (I^-)^2 \, / \, (I_2) \qquad \text{(Eq. 22)}$$

where, $\frac{1}{w} = 2.303 \, \frac{RT}{2F}$,

$Q = (I_2) \, / \, (I^-)^2$ in the reference half-cell, and

(I^-) and (I_2) are the iodide and iodine molarities in the unknown half-cell.

Solving Eq. 22 for (I^-), one obtains

$$(I^-) = 10^{1/2\ wE\ -\ \log Q\ -\ \log (I_2)} \qquad \text{(Eq. 23)}$$

It is, therefore, evident that from a measurement of E for any solution containing I_2 and I^- in equilibrium against a reference half-cell, it is possible to estimate (I^-) if one has an independent estimate of (I_2). Inasmuch as (I_2) may be estimated by suitable application of the distribution law, the estimation of (I^-) follows directly upon the measurement of the emf of such solutions against an appropriate reference solution.

MATERIALS AND METHODS

Determination of (I_2)

The concentration of free iodine in an aqueous solution containing an appreciable amount of iodide ion cannot in general be determined by direct titration if there has been considerable incorporation of iodine into polyiodide ions which are in rapid, reversible equilibrium with the free iodine. Thus, one must have recourse to some indirect method for the determination of free iodine in such systems.

One such indirect method is the distribution of iodine between the phase of interest, and a second immiscible liquid phase which may then be used as a reference phase. In order to apply such a technique, it is necessary to have some knowledge of the appropriate distribution coefficient.

Since the aqueous phase of interest will in general contain iodide and polyiodide salts, it is essential that the distribution coefficient used to relate the iodine concentration in one phase to that in the other be one which takes into account the presence of such salts. Furthermore, the hydrolysis of iodine must be taken into account or minimized during the determination of the distribution coefficient.

Inasmuch as the aqueous phases of interest contain dissolved salts at a constant ionic strength of 2.00, the appropriate distribution coefficient would appear to be one between a reference phase such as CCl_4 and an aqueous phase containing an inert salt at an ionic strength of 2.00. By an inert salt is meant one which does not undergo specific interactions with iodine. The use of KNO_3 as a suitable inert salt is suggested by the fact that the solubility of iodine in 2 M KNO_3 when corrected for the volume occupied by the salt and cationic hydration is essentially identical with the solubility of iodine in water alone. Since KNO_3 is used as a diluent salt for the maintenance of constant ionic strength in the polyiodide solutions of interest, it is of course desirable to use this same diluent salt for maintenance of constant ionic strength in the determination of the distribution coefficient. The use of KNO_3 as a diluent salt in the polyiodide containing solutions is dictated by the fact that it is a 1-1 electrolyte as is KI, the iodide salt used in these solutions, and by the desirability of introducing as few different kinds of cations as possible. The presence of dilute sulfuric acid in the aqueous phase during the determination of the distribution coefficient is also desirable to repress the hydrolysis of iodine.

The appropriate distribution coefficient, therefore, is that for iodine distributed between CCl_4 and an aqueous phase containing 2.00 M KNO_3 and .001 M H_2SO_4. The experimental determination of this distribution coefficient as a function of iodine concentration in the CCl_4 phase is discussed in a later section.

Determination of (I^-)

In order to make use of the potentiometric method to determine the (I^-) by means of Eq. 23 , one must first establish the fact that a reference electrode such as the one described does indeed exist. The use of such a concentration cell under the conditions of constant ionic strength also gives some justification to the cancellation of activity coefficients required in the derivation of Eq. 21.

In order to use an iodine electrode as a reference electrode, it would seem that some estimation of the polyiodide formation function is necessary in order to obtain an estimate of (I_2) and (I^-); thus, it would appear that one needs Φ to obtain (I^-) and vice versa. Such a situation might be met by an iterative estimation of the quantities involved; however, a more direct alternative is available.

Consider the following definition of K*,

$$K^* = (I_2)_b \ / \ (I_2) \ \left[(I^-)_t - (I_2)_b\right] \qquad \text{(Eq. 24)}$$

From the definition of Φ in Eq. 8, it is clear that

$$\frac{K^*}{\Phi} = (I^-) \ / \ (I^-)_t - (I_2)_b \qquad \text{(Eq. 25)}$$

Thus, to the extent that $(I^-)_t - (I_2)_b$ is an estimate of (I^-), K* and Φ estimate the same thing. It is evident also that as $(I_2)_b$ approaches zero, (I^-) and $(I^-)_t$ become more and more identical; thus, as (I_2) approaches zero, $(I_2)_b$ must also approach zero and K* must approach Φ. Since the estimation of K* involves only the determination of (I_2), $(I^-)_t$, and (I_2), it is possible to obtain a reliable estimate of Φ at (I_2) = zero by a graphical plot of K* against (I_2). Furthermore, a value for K* at very low (I_2) may be obtained directly from such a graph and could be reasonably expected to be a reliable estimate of Φ at this I_2 concentration. Thus, in principle one may use such a solution as a reference solution for the potentiometric determination of (I^-) in more concentrated solutions.

The actual experimental determination of (I^-) by such means is described in a later section.

Determination of $(I_2)_b$

The determination of $(I_2)_b$ proceeds immediately from the following identity.

$$(I_2)_b = (I_2)_t - (I_2) \qquad \text{(Eq. 26)}$$

Since one may obtain an estimate of (I_2) from distribution methods, the estimation of $(I_2)_b$ follows immediately upon the determination of $(I_2)_t$. This quantity may be obtained simply by the titration of the aqueous phase with a reagent such as sodium thiosulphate which reacts quantitatively with I_2. The experimental determination of this quantity is discussed in a later section.

Multiple Regression Analysis

The expanded form of the polyiodide formation function given in Eq. 11 suggests at once that a general multiple regression analysis of Φ as a function of the equilibrium constants for polyiodide formation and precise values of (I_2) and (I^-) would provide a powerful method for the estimation of these equilibrium constants. In addition, the usual statistical procedures may be used to determine whether or not any given estimate of an equilibrium constant is significantly different from zero. In other words, it is possible to determine simultaneously whether or not any of the polyiodides included in the model exist, and the formation constants for those which appear to exist.

In order to elaborate upon the applicability of multiple regression analysis, it is useful to make the following identifications with the quantities appearing in Eq. 11.

$$\begin{array}{lll}
\text{Let} \quad K_3 = A_0 & 3\ K_7 = A_4 & \\
K_4 = A_1 & 3\ K_8 = A_5 & \\
2\ K_5 = A_2 & 3\ K_9 = A_6 & \\
2\ K_6 = A_3 & 4\ \bar{K}_9 = A_7 & \\
(I^-) = x_1 & (I_2) = x_2 & \text{(Eq. 27)} \\
(I^-)(I_2) = x_3 & (I_2)^2 = x_4 & \\
(I^-)(I_2)^2 = x_5 & (I^-)^2(I_2)^2 = x_6 & \\
(I_2)^3 = x_7 & &
\end{array}$$

By making the above identifications, it is obvious that Eq. 11 may be expressed as follows.

$$\Phi = A_o + A_i\ x_1 + A_2\ x_2 + A_3\ x_3 + A_4\ x_4 + A_5\ x_5 + A_6\ x_6 + A_7\ x_7 \qquad \text{(Eq. 28)}$$

This expression may be more conveniently expressed as

$$\Phi = A_o + \sum_i A_i\ x_i \qquad \text{(Eq. 29)}$$

where $i = 1, 2, \ldots, 7$.

Furthermore, if one makes several observations of Φ and the x_i, say n observations, one would have the set of equations,

$$\Phi_j = A_{oj} + \sum_i A_i\ x_{ij} \qquad \text{(Eq. 30)}$$

where $j = 1, 2, \ldots, n$. In this form, one may immediately recognize the general linear hypothesis where the x_{ij} are constants and the A_i are regression coefficients to be estimated from observations of the Φ_j and x_{ij}.

Of course, it is true that the x_{ij} are not constants which may be fixed at a definite set of values during the experiment; however, one may conceptually consider the x_{ij} to be experimentally determinable with such a high degree of precision that repetition would lead to exactly the same value for the x_{ij}. This is an approximation which may only be approached and never reached experimentally, but it is nonetheless extremely useful to explore the consequences of such an approximation.

Low Molecular Weight Amylose

The low molecular weight amylose used in the electrophoretic fractionation experiments was obtained by the acid hydrolysis of Superlose, a commercially available amylose obtained from Stein and Hall Co. The Superlose was dispersed in 9 M HCl at ice temperatures, and the hydrolysis was allowed to proceed until the hydrolysate gave a plum-colored complex upon the addition of aqueous iodine. The hydrolysis was then arrested by neutralization with anhydrous NaAc and the salts were then removed from the hydrolysate by repeated dialysis and vacuum concentration at room temperature. The low molecular weight amylose was recovered from solution by precipitation with acetone at room temperature.

Fractionation of Low Molecular Weight Amylose

The separation of amylose-polyiodide complexes into three fractions with different iodine-staining properties was achieved through an adaptation of the procedure developed by Mould and Synge (50). A Beckman Model CP hanging-curtain electrophoresis apparatus was employed in conjunction with a glass curtain fabricated from Whatman GF/B glass fiber paper.

The supporting electrolyte employed was an acetate buffer of pH 4.8, ionic strength .04, and contained .002 M KI and

.001 $\underline{M}$ I_2. The solution which was fractionated consisted of a 0.08% solution of low molecular weight amylose in the supporting electrolyte. Upon impressing a potential difference of 230 v. across the curtain, the applied solution was separated into three fractions of distinctly different color. Blue, red, and orange-yellow bands were readily observed to move down the curtain with the flow of the buffer. The blue and red fractions were actually separated by about 2 cm at the point of collection while the bands themselves were about 4 cm in width at this point. No actual "window" could be observed between the red and orange-yellow fractions. These fractions were collected and their absorption spectra determined. The spectra are discussed in a later section.

EXPERIMENTAL

Distribution Coefficient

The distribution coefficient, K_d, for I_2 between CCl_4 and aqueous 2 M KNO_3 containing .001 M H_2SO_4 was determined at 25 °C by the equilibration of 100 cc of CCl_4 and 100 cc of the acidified 2 M KNO_3 with varying amounts of added I_2. The reagents used throughout were Mallinckrodt analytical reagent grade chemicals. These were used without further purification except for the CCl_4 which was redistilled immediately prior to use. The water used in the aqueous phase was obtained by redistillation of distilled water from an alkaline $KMnO_4$ solution.

The two liquids were introduced into 250 cc glass-stoppered flasks and solid I_2 was added. From preliminary data, it was possible to add an amount of I_2 sufficient for the approximate establishment of any desired I_2 concentration in the CCl_4 phase. Thus, several determinations of K_d were made at different I_2 concentrations between 4×10^{-3} M and 1.1×10^{-1} M in the CCl_4 phase. This covered a concentration range from very dilute solutions to saturated solutions of I_2.

After introduction of the I_2 into the flasks, they were stoppered and subjected to the action of a wrist-action shaker until the I_2 had dissolved completely in all solutions less than saturated with respect to I_2. The flasks were then placed in a water bath thermostated at 25 ± .02 °C and brought to equilibrium at this temperature. It was determined that even after 2 weeks of equilibration, during which time the flasks were frequently agitated for extended periods of time by a wrist-action shaker, some of the more concentrated solutions had not yet achieved equilibrium. However, after 3 weeks one could not detect any further changes in the ratio of I_2 concentrations in the two phases.

The I_2 concentration in the liquid phases was determined by titration of aliquots withdrawn by means of a volumetric pipette with standardized solutions of sodium thiosulphate. The normality of the thiosulphate solution used in titration was chosen so that the titrating volume would be in the range 20 to 30 cc whenever possible. All standard solutions were frequently standardized against a primary standard which consisted of a carefully prepared solution of KIO_3 which was allowed to liberate I_2 from KI in acid solutions. Standardization was achieved by direct titration of the I_2 liberated with the sodium thiosulphate solutions.

The aliquots withdrawn from the sample flasks were immediately pipetted into titration flasks containing a small volume of water to which about .5 g of KI had been added. The presence of KI helped to minimize the loss of I_2 by volatilization during the titrations and, in the case of the CCl_4 phases, it also increased greatly the rate at which the I_2 could be extracted into the aqueous phase for titration. A magnetic stirrer was also used to speed up the rate of extraction of I_2 into the aqueous phase by providing a mechanism for the intimate mixing and constant renewal of surfaces between the phases.

The I_2 was titrated directly in the aqueous phase with a standardized solution of sodium thiosulphate. Duplicate titrations agreed to within 0.10 cc. The concentration of I_2 in either phase was then calculated from the following equation.

$$(I_2) = \underline{N} \times V_t \, / \, 2 \, V_s \qquad \text{(Eq. 31)}$$

where, $\underline{N}$ = the normality of the titrating solution

V_t = the average titrating volume

V_s = the volume of the sample aliquot

(I) = the molar concentration of (I_2) in the sample.

The distribution coefficient, K_d, was then calculated from the following definition.

$$K_d = (I_2)_c \, / \, (I_2)_a \qquad \text{(Eq. 32)}$$

where, $(I_2)_c$ is the molar concentration of I_2 in the CCl_4 phase,

$(I_2)_a$ is the molar concentration of I_2 in the aqueous phase.

Inasmuch as K_d is defined in terms of molar concentrations, one might expect that K_d will be a function of the I_2

concentration. Thus, K_d has been determined for several values of $(I_2)_c$. The results of this determination are tabulated in Table 3 and displayed graphically in Fig. 1.

The solubility of I_2 in 2 M KNO_3 as determined in this experiment is in excellent agreement with the value obtained by Kiss and Urmanczy (54), and if corrected for the volume occupied by the KNO_3 and for cationic hydration is very nearly identical to the solubility of I_2 in water alone. Thus, it would appear that the KNO_3 is essentially an inert diluent in the system.

Estimation of Q for the Reference Electrode

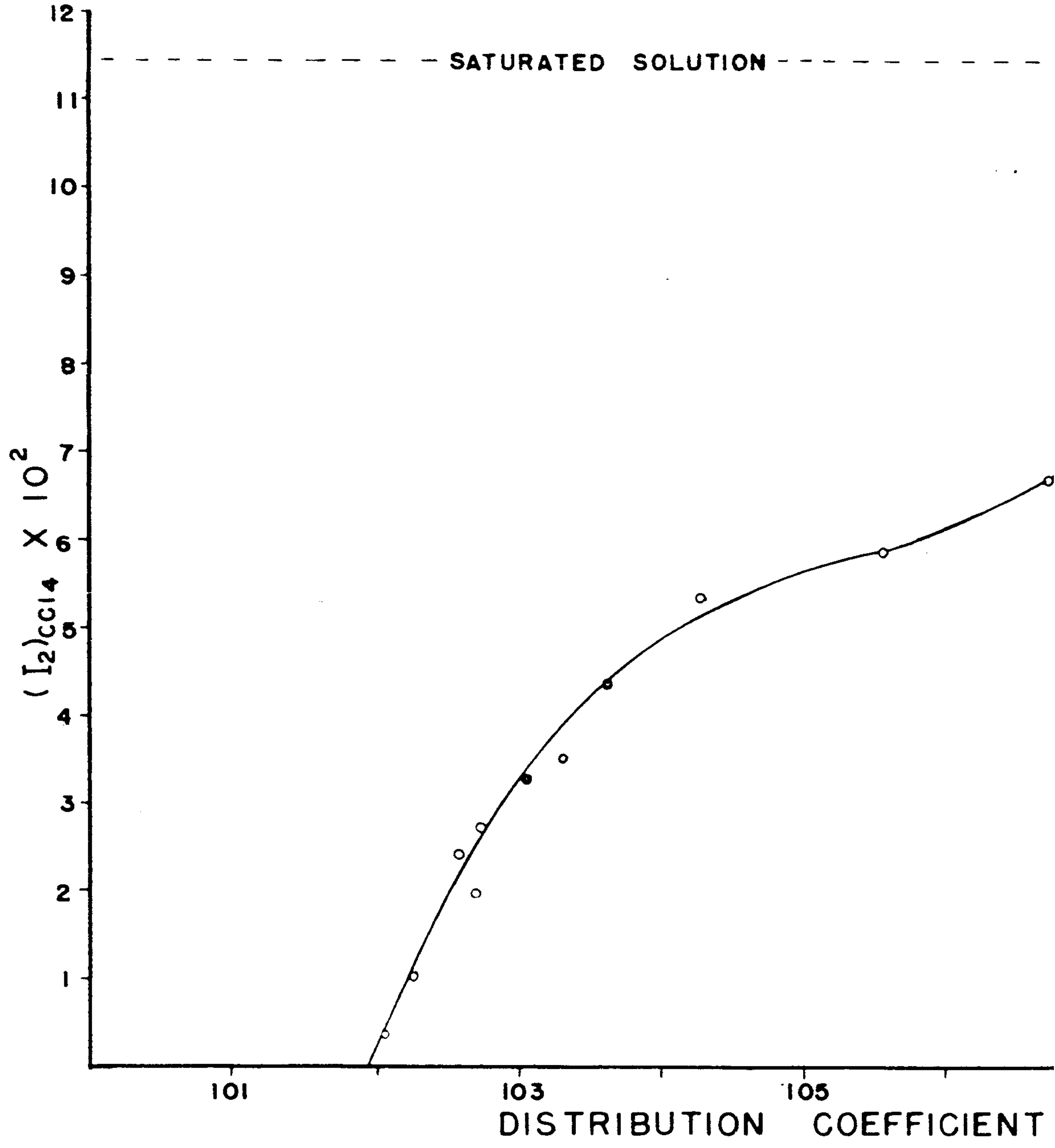
It has been pointed out that in order to establish a suitable reference electrode for use in a concentration cell, one must obtain some estimate of Φ in the reference solution. The most direct approach to this problem has been outlined previously and involves the determination of K* as defined in Eq. 24. In order to obtain values for K* experimentally, one must have estimates of $(I^-)_t$, $(I_2)_b$, and (I_2).

It is clear from Eq. 12 that if the formation of $I_4^=$ did in fact occur in aqueous solutions, the value of Φ approached as (I_2) approaches zero should be a linear function of (I^-). Since K* is an approximation to Φ in the same limit, it is to be expected that the same functional relationship would exist between k* and (I^-). It was necessary, therefore, to

Table 3. Distribution coefficient for I_2 between CCl_4 and aqueous 2 $\underline{M}$ KNO_3 containing .001 $\underline{M}$ H_2SO_4 at 25 ± 0.02 °C

$(I_2)_{CCl_4} \times 10^2$	$(I_2)_{aq} \times 10^4$	K_d
0.3913	0.3833	102.0_8
1.5364	1.5020	102.2_9
1.9811	1.9283	102.7_4
2.3857	2.3249	102.6_1
2.7104	2.6377	102.7_6
3.1492	3.0563	103.0_3
3.5011	3.3885	103.3_2
4.3479	4.1954	103.6_3
5.1699	5.0121	104.7_5
5.8621	5.5533	105.5_6
6.6538	6.2316	106.7_8
7.1386	6.6505	107.3_3
8.4866	7.8308	108.3_7
8.9857	8.3038	108.2_1
9.6909	8.9132	108.7_2
10.6859	9.8426	108.5_7
11.4863	10.5435	108.9_4

Fig. 1. Distribution coefficient, K_d, of I_2 between CCl_4 and an aqueous 2 M KNO_3 solution containing 0.001 M H_2SO_4 at 25 ± .02 °C


SATURATED SOLUTION
$(I_2)_{CCl_4} \times 10^2$
12
11
10
9
8
7
6
5
4
3
2
1
101
103
105
DISTRIBUTION COEFFICIENT

determine K* at several concentrations of KI in order to allow for this possibility.

Several aqueous solutions were prepared containing varying amounts of KI from .05 M up to about 1.0 M. Sufficient KNO_3 was added to these solutions to bring them all to an ionic strength of 2.00. Equilibration of 100 cc of each of the aqueous solutions with 100 cc of CCl_4 and varying amounts of I_2 was performed in the same manner described for the determination of the distribution coefficient. Mallinckrodt analytical reagent grade chemicals were used throughout these experiments without further purification.

The concentration of KI in the aqueous phase of these solutions was determined by titration with standard $AgNO_3$ solutions using eosin as an indicator. These titrations were performed in subdued light since the precipitated AgI is quite photosensitive. The molar concentration of the KI solutions so determined was in extremely good agreement with the molar concentration calculated from the weight of KI used to prepare the solution.

The molar concentration of the free I_2 in the aqueous phase was determined from the experimentally obtained molar concentration of I_2 in the CCl_4 phase and the corresponding distribution coefficient obtained from Fig. 1. The molar concentration of the I_2 in the CCl_4 phase was obtained in the manner described for the determination of the distribution

Table 4. K* as a function of (I_2) for several KI concentrations at 25 ± .02 °C

$(I_2)_t \times 10^2$	$(I_2)_c \times 10^2$	$(I_2)_a \times 10^4$	$(I_2)_b \times 10^2$	K*
$(I^-)_t$ = .049494, ionic strength = 2.00				
0.11385	0.31405	0.30789	0.11077	743.5
0.11737	0.32336	0.31702	0.11420	745.0
0.30717	0.88070	0.86174	0.29855	744.9
0.31677	0.91027	0.89068	0.30786	744.8
0.49791	1.48252	1.44849	0.48343	747.3
0.51788	1.54737	1.51184	0.50276	747.9
0.66764	2.06213	2.00987	0.64754	748.9
0.64760	1.98678	1.93832	0.62821	750.0
0.84194	2.69913	2.62433	0.81570	751.9
0.83340	2.67359	2.60077	0.80739	749.5
1.00574	3.34691	3.24628	0.97318	753.9
1.02032	3.39982	3.29600	0.98736	756.1
1.16273	4.01633	3.88051	1.12393	757.1
1.18930	4.15277	4.00074	1.14929	756.0
1.28990	4.60075	4.42593	1.24558	759.8
$(I^-)_t$ = .098988, ionic strength = 2.00				
0.26516	0.36586	0.35869	0.26157	756.7
0.23266	0.32165	0.31534	0.22950	752.7
0.98436	1.47028	1.43582	0.97000	756.6
1.23348	1.88723	1.84030	1.21251	758.5
1.52656	2.40872	2.34539	1.49711	759.8
1.63920	2.61735	2.54606	1.61379	765.0
2.06554	3.46838	3.35920	2.03194	766.9
2.22211	3.80751	3.67875	2.18532	770.1
2.56040	4.55019	4.37939	2.51661	778.4
2.59760	4.65391	4.47707	2.55283	776.2
2.98750	5.65242	5.38839	2.93361	781.6
3.07775	5.92488	5.61600	3.02159	782.3
3.37340	6.84499	6.39420	3.30946	785.5
3.46503	7.08556	6.60966	3.39893	791.1
3.75373	8.03264	7.42045	3.67952	797.3

coefficient. Duplicate titrations agreed to within 0.1 cc in every instance.

The total I_2 concentration in the aqueous phase was also determined as described in the section on the determination of the distribution coefficient. From the identity expressed in Eq. 23, the $(I_2)_b$ was obtained from the difference between $(I_2)_t$ and (I_2). Having thus determined the values for $(I^-)_t$, $(I_2)_t$ and (I_2), it was possible to calculate K* by means of Eq. 24. The results of these determinations and calculations are tabulated in Table 4.

In order to use this data to provide a suitable description of a solution which may be used as a reference electrode standard, it is sufficient to decide upon an arbitrary but low free I_2 concentration for the reference solution. One may then obtain a fairly precise estimate of K*, and therefore Φ, directly from Fig. 2. It is then possible to calculate the amount of I_2 which would have to be dissolved in the solutions of KI and KNO_3 to achieve this concentration. Since the value of K* changes only very slightly with (I_2) at very low concentrations, only a close approximation to the calculated amount of total I_2 required need be achieved. The exact concentration of $(I_2)_t$ in these solutions could be obtained by titration.

Such a procedure was adopted by choosing a target concentration of 1×10^{-5} M free I_2 for the reference solutions.

Table 4. (Continued)

$(I_2)_t \times 10^2$	$(I_2)_c \times 10^2$	$(I_2)_a \times 10^4$	$(I_2)_b \times 10^2$	K*
		$(I^-)_t = 0.29698$, ionic strength = 2.00		
0.67598	0.30597	0.29601	0.67300	778.1
0.62598	0.28241	0.27687	0.62311	774.1
2.15748	0.98615	0.98644	2.14761	790.3
1.99217	0.93165	0.91159	1.98306	785.0
3.72557	1.80627	1.76135	3.70795	810.0
3.44694	1.68700	1.62634	3.43068	803.1
5.23600	2.66798	2.59531	5.21005	819.8
4.85662	2.43977	2.37563	4.83287	818.2
6.49264	3.43180	3.32378	6.45940	836.3
6.73179	3.60412	3.48729	6.69691	835.0
7.77146	4.29968	4.14627	7.72999	848.7
8.21468	4.58581	4.41367	8.17054	860.0
9.17594	5.35181	5.11157	9.12482	867.8
9.64638	5.70414	5.43769	9.59200	877.4
10.39808	6.38422	5.99739	10.33811	890.5
		$(I^-)_t = 0.49453$, ionic strength = 2.00		
1.12066	0.29442	0.28865	1.11778	800.5
1.14497	0.30065	0 29476	1.14202	802.0
3.62947	0.97772	0 95620	3.72903	826.0
3.73883	1.00487	0.98276	3.72900	829.9
6.10086	1.68470	1.64522	6.08441	852.7
6.38687	1.75499	1.71302	6.36974	863.1
8.44692	2.38828	2.32775	8.42351	881.9
8.99475	2.57422	2.50532	8.96970	884.4
10.77261	3.15425	3.06088	10 74200	906.6
11.52169	3.40060	3.29675	11.48873	918.0
13.10848	3.99168	3.85670	13.06991	931.4
13.51312	4.11387	3.92092	13.47341	943.0
15.35072	4.88118	4.68219	15.30390	957.1
15.68364	4.96872	4.76616	15.63598	970.1
17.63804	5.82398	5.52036	17.58289	999.4

Table 4. (Continued)

$(I_2)_t \times 10^2$	$(I_2)_c \times 10^2$	$(I_2)_a \times 10^4$	$(I_2)_b \times 10^2$	K*
		$(I^-)_t = 0.69290$, ionic strength = 2.00		
1.70439	0.31122	0.30511	1.70134	825.0
1.56640	0.28437	0.27879	1.56361	828.1
6.01052	1.11076	1.08685	5.99965	872.2
6.52801	1.20613	1.17901	6.51622	880.4
10.26567	1.92030	1.87255	10.24695	926.8
9.79773	1.84591	1.80089	9.77972	912.5
14.29975	2.78955	2.71093	14.27264	957.0
13.22279	2.54111	2.47310	13.19805	951.4
18.68416	3.70064	3.63818	18.64778	1012.1
17.49657	3.48713	3.37900	17.46258	997.1
22.93066	4.86840	4.67249	22.88384	1055.4
23.76258	5.00016	4.79632	23.71462	1084.9
26.86910	5.91360	5.60797	26.81302	1125.6
27.73548	6.22175	5.86681	27.69681	1135.0
31.13002	7.34319	6.82769	31.06174	1190.1
		$(I^-)_t = 0.84077$, ionic strength = 2.00		
2.14320	0.31250	0.30637	2.14014	851.9
1.94735	0.28617	0.28056	1.94455	848.3
6.91579	1.01555	0.99320	6.90586	901.0
7.39765	1.08431	1.06045	7.38692	908.3
11.81030	1.75355	1.71445	11.79318	952.7
13.20202	1.96002	1.91128	13.18291	972.9
16.46480	2.48510	2.41976	16.43060	1003.8
15.74794	2.37825	2.31572	15.72478	993.4
21.61418	3.29733	3.19974	21.58218	1079.3
21.98866	3.41613	3.31181	21.95556	1067.2
25.74380	4.07725	3.93940	25.70441	1117.8
24.74949	4.20459	4.05652	24.70893	1126.0
31.16160	5.20725	4.98540	31.11175	1178.3
32.80650	5.53302	5.27708	32.75403	1209.4
35.26454	6.08608	5.75518	35.20699	1245.4

Table 4. (Continued)

$(I_2)_t \times 10^2$	$(I_2)_c \times 10^2$	$(I_2)_a \times 10^4$	$(I_2)_b \times 10^2$	K*
	$(I^-)_t = 0.98216$, ionic strength = 2.00			
2.53566	0.30631	0.30030	2.53266	881.4
6.06853	0.73654	0.72104	6.06132	912.2
7.91390	0.96181	0.94111	7.90449	930.0
9.41285	1.13497	1.10935	9.40175	953.7
10.57514	1.28406	1.26519	10.56253	960.0
13.83796	1.67552	1.63465	13.82161	1001.9
14.79017	1.80395	1.75995	14.77257	1005.9
19.00264	2.33126	2.27108	18.97993	1054.7
19.80901	2.42079	2.35715	19.78544	1070.2
24.20448	2.99787	2.91197	24.17536	1121.3
25.87337	3.22495	3.12950	25.84208	1141.0
32.01387	3.99825	3.86304	31.33053	1212.6
31.94875	4.11807	3.97497	31.97524	1214.4
39.15974	5.30992	5.07641	39.10898	1303.4
40.70396	5.51408	5.25902	40.65137	1342.8

The actual concentration of total iodine added was determined by direct titration with standard sodium thiosulphate. The KI solutions employed were the same solutions used in the determination of K* at the various total KI concentrations.

The actual concentration of free I_2 and free I^- were then calculated from the concentration of total KI, total I_2, and the appropriate value for K* obtained from Fig. 2. From these calculated values of (I_2) and (I^-), the quantity, Q, defined in Eq. 22 was readily computed. The results of these computations and the data pertaining to them are tabulated in Table 5.

Fig. 2. K^* as a function of the free iodine concentration in moles/liter for solutions of ionic strength 2.00 at 25 ± .02 °C

A = 0.049898 M KI

B = 0.098988 M KI

C = 0.29696 M KI

D = 0.49453 M KI

E = 0.69290 M KI

F = 0.84077 M KI

G = 0.98216 M KI

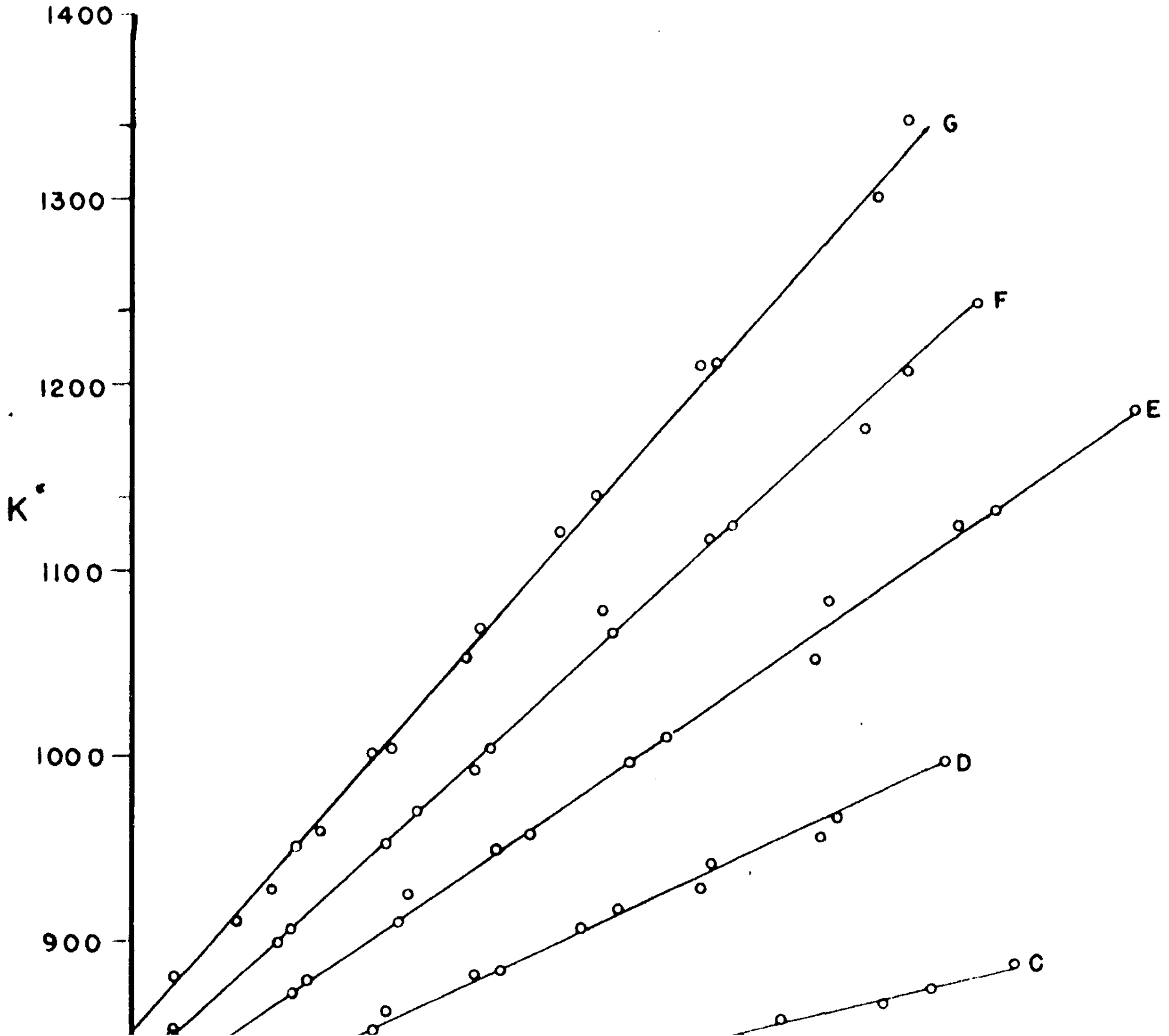

1400
1300
1200
1100
1000
900
K'
G
F
E
D
C

Table 5. Composition of reference electrode solutions[a]

$(I^-)_t$	$(I_2)_t \times 10^4$	K*[b]	$(I_2)_c \times 10^4$	$(I_2) \times 10^5$	(I^-)	$(I^-)^2/(I_2)$
.049494	83.069	743	8.0834	2.2345	.048686	106.079
.098988	7.9071	760	7.8005	1.066	.098208	904.767
.29696	24.033	772	23.9282	1.052	.29457	8248.24
.49453	39.826	798	39.7246	1.015	.49056	23709.3
.69290	58.093	817	57.9897	1.033	.68710	45702.5
.84077	70.752	837	70.6506	1.012	.83371	68683.0
.98216	83.912	853	83.8115	1.009	.97378	93978.9

[a] All solutions contain sufficient KNO_3 to establish an ionic strength of 2.00.

[b] Obtained from Fig. 2 for $(I_2) = 1 \times 10^{-5}$ <u>M</u>.

Determination of the Polyiodide Formation Function

It was necessary to determine $(I_2)_b$, (I_2), and (I^-) in order to make use of Eq. 8 to calculate Φ, the polyiodide formation function. These determinations were conducted in solutions of varying total KI and total I_2 concentration in order to accumulate appropriate data for the multiple regression analysis previously described.

The same method was employed for the determination of $(I_2)_b$ and (I_2) as that described for the determination of K*. Since the determination of K* as a function of (I_2) allowed the estimation of Φ in dilute solutions of I_2 in aqueous KI, it was possible to prepare solutions of KI and I_2 suitable for use as reference electrode electrolytes for the potentiometric determination of (I^-) (see preceding section).

The properties of the standard cell solutions are described in Table 5. It should be noted that a different standard solution was prepared for the several different total KI concentrations encountered in these investigations. The ionic strength of all solutions was adjusted to 2.00. These measures were taken to minimize any effects which might be due to an imbalance in the salt content of the two half-cells under observation.

The actual cell assembly consisted of two glass electrode vessels of 50 cc capacity, one for the reference electrode and one for the working electrode. The electrodes themselves were Pt button electrodes sealed in glass obtained from Beckman Instruments Inc. Electrical contact between the two half-cells was made through a salt bridge containing 2 $\underline{M}$ KNO_3. Actual contact between the bridge solution and the cell electrolyte was made through asbestos fibers sealed into the ends of the bridge. The electrode and bridge lead entered the cell vessel through a rubber stopper which fit tightly into the top of the vessel. The cell vessels were thermostated in a water-bath maintained at 25 ± .02 °C. during measurements. The potential difference between the two Pt electrodes was obtained by means of a Leeds & Northrup Type K potentiometer in conjunction with a ballistic type galvanometer which served as a null detector.

The actual measurements were obtained by placing 40 cc of the appropriate reference solution in one electrode vessel and 40 cc of the solution under investigation in the other electrode vessel. Both the reference solution and the solution under investigation had been thermostated at the temperature of measurement prior to transfer to the electrode vessels which were also immersed in the same thermostat. The emf of the cell thus obtained was reproducible to within 0.1 mv at low (I_2) and within 0.5 mv at high (I_2). The measurements of emf were made just before the titration data was obtained on the solutions under investigation and the CCl_4 phase with which they had been equilibrated. The (I^-) was calculated by means of Eq. 25 from the value of $\sim$ for the reference solution, (I_2), and the measured emf. It was then possible to calculate Φ from Eq. 8. The data for the solutions investigated in this manner are tabulated in Tables 6 through 12 along with the calculated values of .

Spectrophotometry

Aqueous solutions of I_2 and KI

The spectrophotometric behavior of solutions of I_2 in KI was investigated at high concentrations of KI in an attempt to corroborate the suggestion from potentiometric measurements that polyiodides of higher order in I^- exist in such solutions.

Several solutions containing the same concentration of total I_2 but varying amounts of total KI were prepared by volumetric dilution of a stock solution of I_2 in very dilute KI with a concentrated stock KI solution. The total KI concentrations investigated ranged from .01 to 3.6 *M*. The concentration of the stock I_2 solution was determined by titration with a standard solution of sodium thiosulphate and that of the stock KI solution was calculated from the weight of KI used in preparation of the solution.

A Cary recording spectrophotometer was used to obtain the absorption spectra of these solutions. Due the the extremely high absorbancies encountered in the ultraviolet spectra of such solutions, it was found necessary to use 1 cm

Table 7. Data for computation of the polyiodide formation function

$(I_2)_t \times 10^2$	$(I_2)_c \times 10^2$	$(I_2)_a \times 10^4$	$(I_2)_b \times 10^2$	emf (v)	(I^-)	Φ
$(I^-)_t$ = .098988, ionic strength = 2.00						
0.26516	0.36586	0.35869	0.26157	0.01551	.097806	745.6
0.23266	0.32165	0.31534	0.22950	0.01342	.097001	750.3
0.98436	1.47028	1.43582	0.97000	0.03602	.088686	761.6
1.23548	1.88723	1.84030	1.21251	0.04017	.085426	771.3
1.52656	2.40872	2.34539	1.49711	0.04425	.082278	775.8
1.63925	2.61735	2.54606	1.61379	0.04566	.081143	781.1
2.06554	3.46838	3.35920	2.03194	0.05030	.077804	777.5
2.22211	3.80751	3.67875	2.18532	0.05200	.076208	779.5
2.56040	4.55019	4.37939	2.51661	0.05520	.073409	783.0
2.59760	4.65391	4.47707	2.55283	0.05558	.073134	779.7
2.98750	5.65242	5.38839	2.93361	0.05953	.068797	791.4
3.07775	5.92488	5.61600	3.02159	0.06045	.067766	793.9
3.37340	6.84499	6.39420	3.30946	0.06331	.064689	800.1
3.46503	7.08556	6.60966	3.39893	0.06410	.063779	806.3
3.75373	8.03264	7.42045	3.67952	0.06656	.061166	810.7
3.82051	8.27470	7.64760	3.74403	0.06739	.060356	811.1
4.05019	9.10694	8.37805	3.96641	0.06973	.057672	820.9
4.12882	9.33351	8.58649	4.04296	0.07035	.056994	826.1
4.44220	10.54592	9.68404	4.35454	0.07262	.055436	811.1
4.52101	10.87323	9.96630	4.42135	0.07325	.054847	808.8
4.67242	11.45760	10.49711	4.56745	0.07438	.053867	807.8
4.56845	11.04171	10.11608	4.46729	0.07361	.054499	810.3
4.66928	11.45800	10.49748	4.56431	0.07470	.053201	817.3
4.66645	11.45700	10.49656	4.56149	0.07431	.054013	804.6

Table 6. Data for computation of the polyiodide formation function (temperature = 25 ± .02 °C)

$(I_2)_t \times 10^2$	$(I_2)_c \times 10^2$	$(I_2)_a \times 10^4$	$(I_2)_b \times 10^2$	emf (v)	(I^-)	Φ
		$(I^-)_t = 0.04949$, ionic strength = 2.00				
0.11385	0.31405	0.30789	0.11077	0.00432	.048303	744.8
0.11737	0.32336	0.31702	0.11420	0.00439	.048881	737.0
0.30717	0.88070	0.86174	0.29855	0.01838	.046749	741.1
0.31677	0.91027	0.89068	0.30786	0.01892	.046538	742.7
0.49791	1.48252	1.44849	0.48343	0 02620	.044703	746.6
0.51788	1.54737	1.51184	0.50276	0.02678	.044650	744.8
0.66764	2.06213	2.00987	0.64754	0.03127	.043226	745.3
0.64760	1.98678	1.93832	0.62821	0.03087	.043116	751.7
0.84194	2.69913	2.62433	0.81570	0.03556	.041796	743.7
0.83340	2.67359	2.60077	0.80739	0.03551	.041690	744.6
1.00564	3.34691	3.24628	0.97318	0.03930	.040198	745.8
1.02032	3.39982	3.29600	0.98736	0.03950	.040181	745.5
1.16273	4.01633	3.88051	1.12393	0.04249	.038808	746.3
1.18930	4.15277	4.00074	1.14929	0.04300	.038630	743.6
1.28990	4.60075	4.42593	1.24558	0.04505	.037514	750.2
1.32903	4.78631	4.59780	1.28305	0 04591	.036977	754.7
1.43087	5.30648	5.06827	1.38019	0.04789	.035942	757.7
1.43657	5.49116	5.23466	1.41422	0.04861	.035518	760.6
1.55801	6.02942	5.70158	1.50099	0.05054	.034386	765.6
1.59332	6.26231	5.90227	1.53430	0.05137	.033873	767.4
1.67383	6.72531	6.29710	1.61086	0.05255	.033417	765.5
1.72156	7.00172	6.53145	1.65624	0.05345	.032861	771.7
1.80643	7.56874	7.02760	1.73615	0.05507	.032004	771.9
1.85366	7.88763	7.30336	1.78062	0.05546	.032134	758.7

Table 8. Data for computation of the polyiodide formation function

$(I_2)_t \times 10^2$	$(I_2)_c \times 10^2$	$(I_2)_a \times 10^4$	$(I_2)_b \times 10^2$	emf (v)	(I^-)	Φ
			$(I^-)_t = 0.29698$, ionic strength = 2.00			
0.67598	0.30397	0.29801	0.67300	0.01380	.28973	779.5
0.67598	0.28241	0.27687	0.62311	0.01260	.28927	778.0
2.15748	0.98615	0.98644	2.14761	0.03035	.27676	786.7
1.99217	0.93165	0.91159	1.98306	0.02897	.28074	774.9
3.72557	1.80627	1.73135	3.70795	0.03950	.25901	812.8
3.44694	1.66700	1.62634	3.43068	0.03869	.25686	821.2
5.23600	2.66798	2.59531	5.21005	0.04589	.24516	818.8
4.85662	2.43977	2.37563	4.83287	0.04492	.24928	816.2
6.49264	3.43180	3.32378	6.43940	0.05043	.23250	835.9
6.73179	3.60412	3.48729	6.69691	0.05150	.22843	840.7
7.77146	4.29968	4.14627	7.72999	0.05475	.21943	849.6
8.21468	4.58581	4.41367	8.17054	0.05591	.21645	855.3
9.17594	5.35181	5.11157	9.12482	0.05885	.20775	859.3
9.64638	5.70414	5.43769	9.59200	0.06058	.20032	880.6
10.39808	6.38422	5.99739	10.33811	0.06308	.19086	903.2
10.83359	6.83354	6.38350	10.76975	0.06443	.18683	903.0
11.43296	7.31808	6.83930	11.36457	0.06598	.18206	912.7
12.05723	8.00118	7.39480	11.98328	0.06800	.17500	926.0
12.61568	8.43674	7.77938	12.53789	0.06937	.17017	947.1
13.18389	9.13614	8.40491	13.09984	0.07127	.16427	948.8
13.69984	9.65888	8.88990	13.61094	0.07273	.15960	959.3
13.97612	10.03379	9.21800	13.88394	0.07351	.15767	955.3
14.51296	10.71840	9.82889	14.41467	0.07520	.15244	962.1
14.90092	11.17828	10.24121	14.79851	0.07589	.15148	953.9

Table 9. Data for computation of the polyiodide formation function

$(I_2)_t \times 10^2$	$(I_2)_c \times 10^2$	$(I_2)_a \times 10^4$	$(I_2)_b \times 10^2$	emf (v)	(I^-)	Φ
			$(I^-)_t = 0.49453$, ionic strength = 2.00			
1.12066	0.29442	0.28865	1.11778	0.01353	.48285	802.0
1.14497	0.30065	0.29476	1.14202	0.01400	.48474	799.3
3.62947	0.97772	0.95620	3.72903	0.03040	.46893	835.2
3.73883	1.00487	0.98276	3.72900	0.03088	.45880	827.0
6.10086	1.68470	1.64552	6.08441	0.03890	.43443	851.3
6.38687	1.75499	1.71302	6.36974	0.03990	.42637	872.9
8.44692	2.38828	2.32775	8.42881	0.04489	.40927	884.2
8.99475	2.57422	2.50532	8.96970	0.04631	.40176	891.1
10.77261	3.15425	3.06088	10.74200	0.04963	.39030	899.2
11.52169	3.40060	3.29675	11.48873	0.05101	.38381	908.0
13.10846	3.99168	3.85670	13.00991	0.05417	.36708	923.2
13.51312	4.11387	3.92092	13.47341	0.05472	.36475	930.2
15.35072	4.88118	4.68219	15.30390	0.05810	.34709	941.7
15.68364	4.96872	4.76616	15.63598	0.05863	.34303	956.4
17.63804	5.82398	5.52036	17.58289	0.06210	.32253	987.8
18.23702	6.16543	5.81919	18.17883	0.06340	.31481	992.3
19.79884	6.91152	6.45936	19.73425	0.06578	.30336	1007.1
20.43377	7.18749	6.69226	20.36684	0.06671	.29678	1025.5
21.97512	7.98336	7.38516	21.90127	0.06910	.28405	1044.0
22.29657	8.18647	7.58709	22.22070	0.06992	.27888	1050.2
24.15140	9.28928	8.54970	24.06590	0.07297	.26290	1070.7
24.57742	9.51742	8.74361	24.48998	0.07360	.25942	1079.7
25.69072	10.44736	9.58033	25.59492	0.07615	.24599	1086.1
26.64400	10.88944	9.98573	26.54414	0.07727	.24033	1106.1

Table 10. Data for computation of the polyiodide formation function

$(I_2)_t \times 10^2$	$(I_2)_c \times 10^2$	$(I_2)_a \times 10^4$	$(I_2)_b \times 10^2$	emf (v)	(I^-)	Φ
			$(I^-)_t = 0.69290$, ionic strength = 2.00			
1.70459	0.31122	0.30511	1.70134	0 01392	.67993	820.1
1.56640	0.28437	0.27879	1.56361	0.01357	.67362	832.6
6.01052	1.11076	1.08685	5.99965	0.03213	.63064	875.3
6.52801	1.20613	1.17901	6.51622	0.03375	.62395	885.8
10.26567	1.92030	1.87255	10.24695	0.04121	.58815	930.4
9.79773	1.84591	1.80089	9.77972	0.04061	.59040	919.8
14.29975	2.78956	2.71093	14.27264	0.04745	.55505	948.5
13.22279	2.54111	2.47310	13.19805	0.04600	.56092	951.4
18.68416	3.70064	3.63818	18.64778	0.05310	.51606	993.2
17.49637	3.48713	3.37900	17.46258	0.05171	.52499	984.4
22.93056	4.86640	4.67249	22.88384	0.05849	.47414	1032.9
23.76258	5.00016	4.79632	23.71462	0.05899	.47112	1049.5
26.86910	5.91360	5.60797	26.81302	0.06323	.43192	1107.0
27.75548	6.22175	5.86681	27.69681	0.06398	.42906	1100.3
31.13002	7.34318	6.82769	31.06174	0 06757	.40249	1130.3
31.69656	7.51395	6.97350	31.62682	0.06818	.39722	1141.8
34.39584	8.51066	7.84750	34.31737	0.07202	.36287	1205.1
35.52316	8.94102	8.22919	35.44087	0.07300	.35769	1204.0
38.60143	10.25024	9.40820	38.50735	0.07712	.32575	1256.4
37.43152	9.73735	8.94566	37.34206	0.07511	.33958	1228.3
40.99482	10.51160	10.43417	40.88970	0.08016	.30480	1285.7
39.57924	10.64021	9.75718	39.48167	0.07798	.32084	1261.2
40.95773	11.28776	10.34625	40.85427	0.07971	.30888	1278.4
40.41812	11.03892	10.11817	40.31694	0.07904	.31352	1270.9

Table 12. Data for computation of the polyiodide formation function

$(I_3)_t \times 10^2$	$(I_2)_c \times 10^2$	$(I_2)_a \times 10^4$	$(I_2)_b \times 10^2$	emf (v)	(I^-)	Φ
$(I^-)_t = 0.98216$, ionic strength = 2.00						
2.53566	0.30631	0.30030	2.53266	0.01450	.95534	882.7
6.06853	0.73654	0.72104	6.06132	0.02598	.92377	910.0
7.91390	0.93181	0.94111	7.90449	0.03009	.91295	920.0
9.41285	1.13497	1.10995	9.40165	0.03307	.89144	950.2
10.57514	1.28406	1.25519	10.56253	0.03477	.88725	946.4
13.83796	1.67552	1.63465	13.82161	0.03937	.84651	998.9
14.79017	1.80395	1.75995	14.77257	0.04050	.84056	998.6
19.00264	2.33126	2.27108	18.97993	0.04465	.81241	1026.7
19.80901	2.42079	2.35715	19.78544	0.04558	.78910	1063.7
24.20448	2.99757	2.91197	24.17536	0.04975	.75428	1100.7
25.87337	3.22493	3.12950	25.84208	0.05111	.74162	1113.4
32.01387	3.99825	3.86304	31.33053	0.05553	.69372	1163.1
31.94873	4.11807	3.97497	31.97524	0.05613	.68746	1170.1
39.15974	5.30992	5.07641	39.10898	0.06173	.62471	1233.2
40.70396	5.51408	5.25902	40.65137	0.06288	.60801	1271.3
46.74240	6.66758	6.24014	46.68000	0.06754	.56242	1354.1
47.59872	6.93702	6.47412	47.53398	0.06850	.54204	1354.5
54.48467	8.49658	7.83456	54.40633	0.07442	.47355	1466.4
59.07720	9.60980	8.83235	58.98888	0.07801	.43723	1527.5
59.87833	9.95172	9.13840	59.78695	0.07935	.42213	1549.8
64.66032	11.45760	10.49711	64.55535	0.08320	.38945	1579.1
64.11156	11.27881	10.33805	64.00818	0.08288	.39134	1582.1

Table 11. Data for computation of the polyiodide formation function

$(I_2)_t \times 10^2$	$(I_2)_c \times 10^2$	$(I_2)_a \times 10^4$	$(I_2)_b \times 10^2$	emf (v)	(I^-)	Φ
			$(I^-)_t = 0.84077$, ionic strength = 2.00			
2.14320	0.31250	0.30637	2.14014	0.01443	.82066	851.2
1.95735	0.2[illegible]617	0.28056	1.95455	0.01340	.82394	845.5
6.91579	1.01555	0.99320	6.90586	0.03156	.76447	909.5
7.39753	1.08431	1.06045	7.38692	0.03222	.76994	904.7
11.81030	1.75355	1.71245	11.79318	0.03931	.72482	950.1
13.20202	1.96002	1.91128	13.18291	0.04173	.71382	966.3
16.45480	2.48510	2.41976	16.43060	0 04583	.68471	991.7
15.74794	2.37825	2.31572	15.72478	0 04500	.69182	981.5
21.61418	3.29733	3.19974	21.58218	0.05146	.63240	1066.6
21.98868	3.41613	3.31181	21.95556	0.05198	.63048	1051.5
25.74380	4.07728	3.93940	25.70441	0.05555	.59842	1090.4
24.74949	4.20459	4.05652	24.70893	0.05606	.59530	1023.2
31.16160	5.20725	4.98540	31.11175	0.06068	.55132	1131.9
32.80680	5.53302	5.27708	32.75403	0.06208	.53713	1155.6
35.26454	6.08608	5.75516	35.20699	0.06539		
35.93054	6.28644	5.91943	35.87135	0.06521	.50363	1203.2
39.93878	7.19488	6.70226	39.87176	0.06860	.46964	1266.7
41.67736	7.80516	7.22700	41.60509	0.07068	.44976	1280.0
44.01723	8.50313	7.84060	43.93882	0.07317	.42519	1318.0
48.28317	9.80672	9.00939	48.19307	0.07740	.38658	1383.7
48.83559	10.0078	9.18343	48.74376	0.07799	.38143	1391.5
52.50730	11.43296	10.47930	52.40250	0.08186	.34994	1429.0
52.62173	11.39848	10.44774	52.52128	0.08180	.35076	1433.2

quartz cells with matched quartz spacers of 9.5 mm width. The path length traversed in the solution itself was thus reduced to .05 cm. A solution of exactly the same KI concentration as that of the sample under investigation was used as a blank. The actual absorption due to the KI was negligible in these solutions in the spectral range of interest except for the most concentrated KI solutions. Even in these cells, however, the absorbancy due to the KI was less than 1% of the observed absorbancy at 2880 Å. No attempt was made to maintain constant ionic strength in these solutions in view of the fact that solutions up to 3.6 _M_ in KI were to be examined. The spectra so obtained are displayed in Figs. 3 and 4. The data is tabulated in Table 13.

Amylose-polyiodide complexes

The spectra of the three different amylose-polyiodide complexes isolated by means of the electrophoretic technique described in an earlier section were investigated for comparison with the spectra of polyiodides in aqueous solution containing no complexing agent of the amylose type.

The absorbancy of these fractions as isolated from the electrophoretic apparatus was sufficiently high to require the use of quartz spacers in 1 cm quartz cells. A water blank was used in each instance inasmuch as no suitable reagent blank could be prepared due to the lack of concentration data

Fig. 3. Spectrophotometric behavior of a solution containing 4.963×10^{-4} M I_2 as a function of the total I^- concentration

A = 0.101 M KI

B = 0.501 M KI

C = 1.001 M KI

D = 1.601 M KI

E = 2.001 M KI

F = 2.401 M KI

G = 3.001 M KI

H = 3.601 M KI

Quartz cells and spacers were used (path length, 0.05 cm). The blank contained the same concentration of KI without I_2.

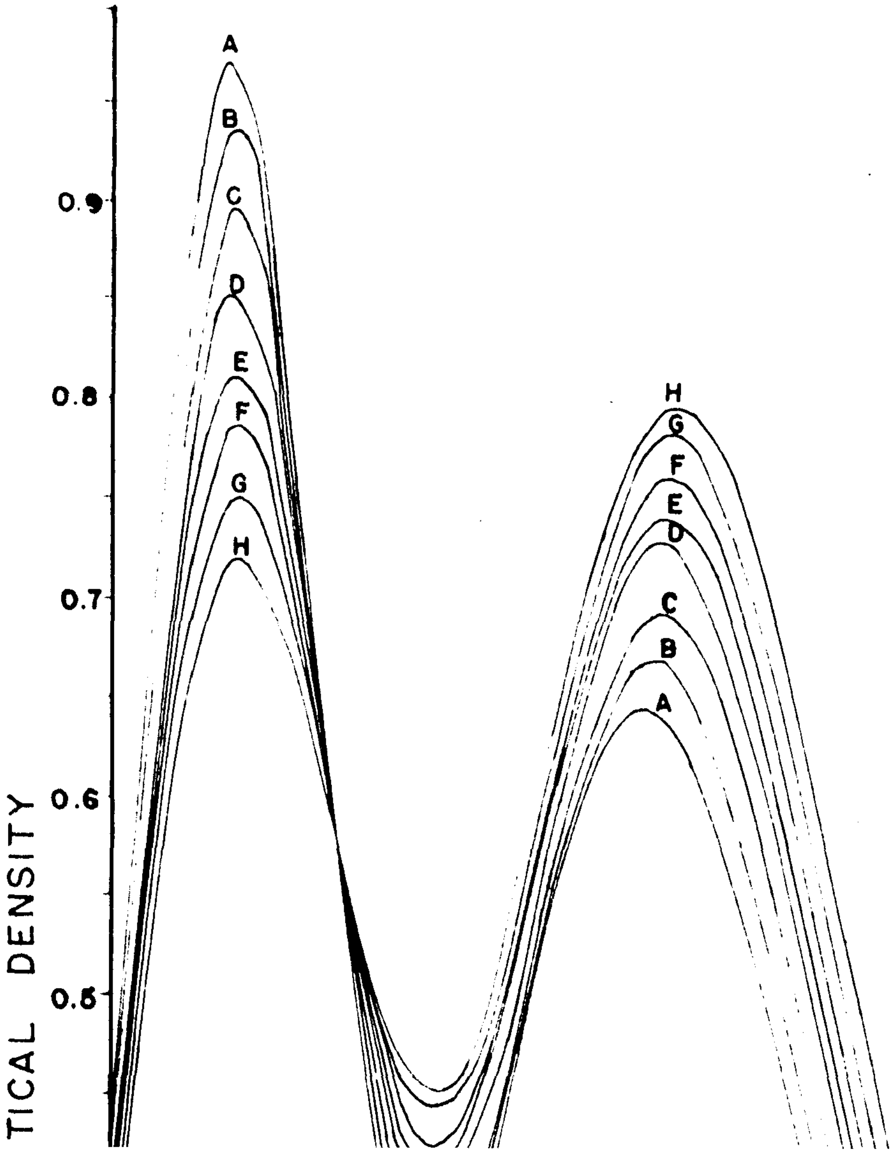

TICAL DENSITY
0.5
0.6
0.7
0.8
0.9
A
B
C
D
E
F
G
H
H
G
F
E
D
C
B
A

Fig. 4. Spectrophotometric behavior of a solution containing 4.070×10^{-4} M I_2 as a function of the total I^- concentration

A = 0.101 M KI
B = 1.001 M KI
C = 1.601 M KI
D = 2.001 M KI
E = 3.001 M KI
F = 3.601 M KI

Quartz cells and spacers were used (path length, 0.05 cm). The blank contained the same concentration of KI without I_2.

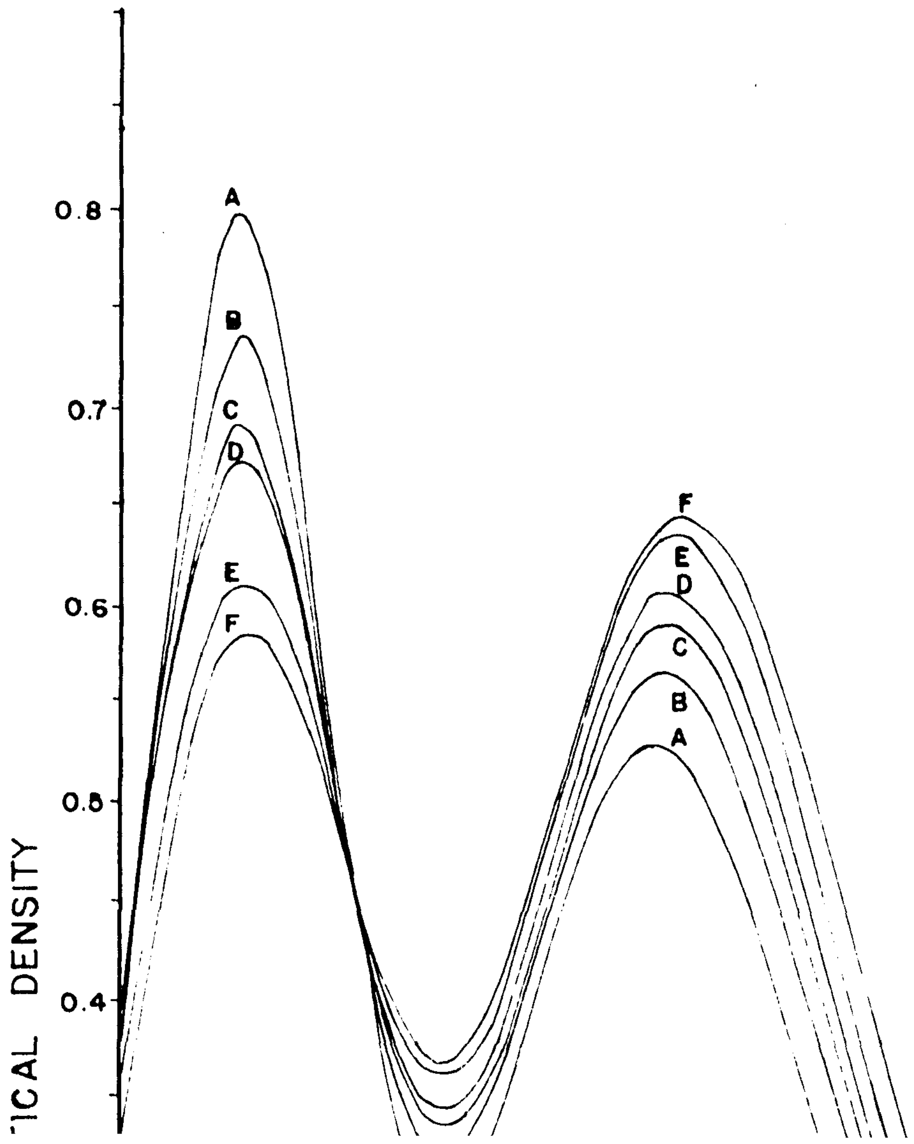

TICAL DENSITY
0.8
0.7
0.6
0.5
0.4
A
B
C
D
E
F
F
E
D
C
B
A

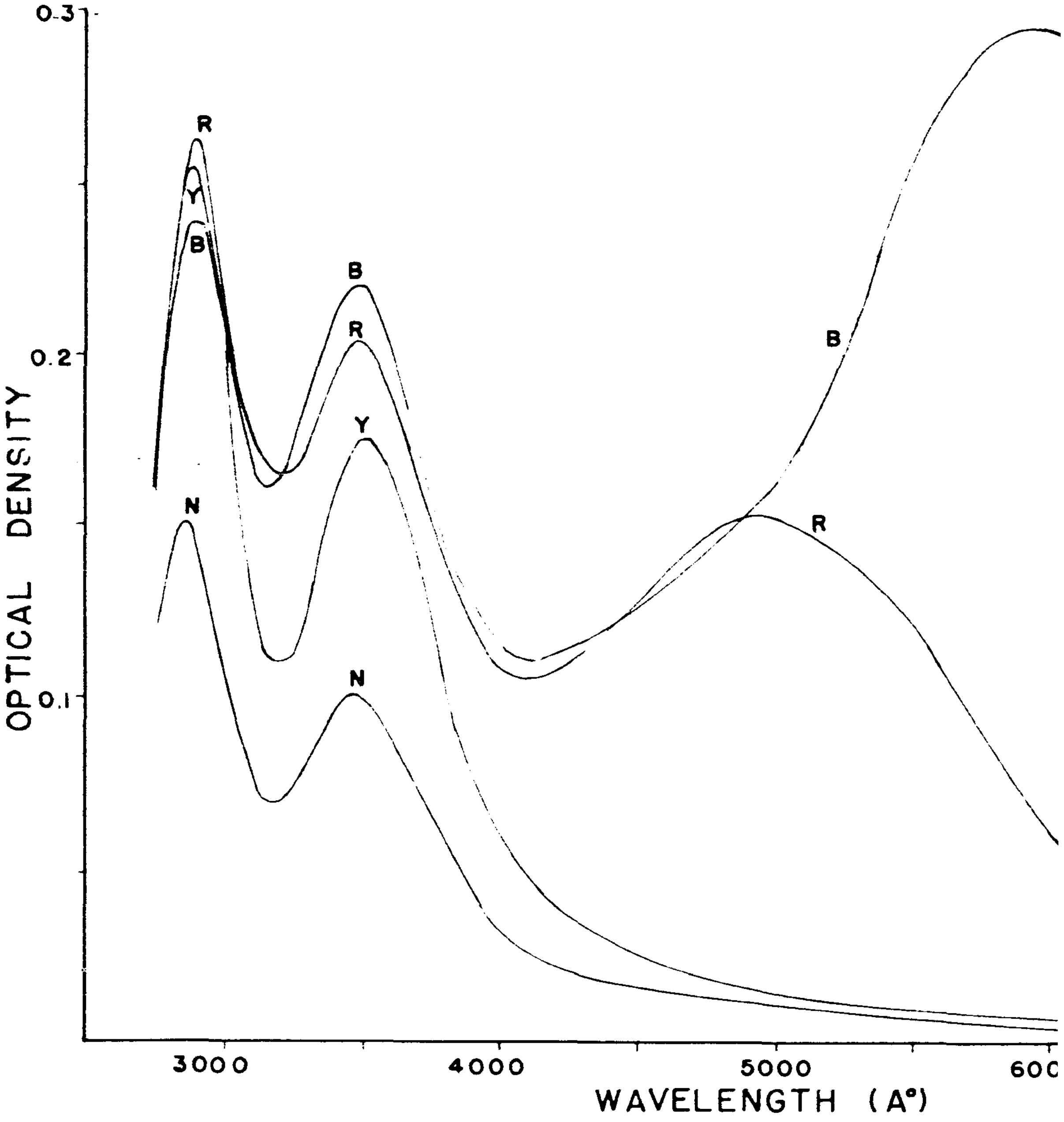
Table 13. Spectrophotometric behavior of aqueous solutions of I_2 and KI

$(I_2)_t = 4.963 \times 10^{-4}$ <u>M</u>			$(I_2)_t = 4.070 \times 10^{-4}$ <u>M</u>		
$(I^-)_t$	A_{2880}[a]	A_{3550}[b]	$(I^-)_t$	A_{2880}[a]	A_{3550}[b]
.011	.850	.558	.011	.695	.455
.051	.958	.630	.051	.787	.518
.101	.973	.645	.101	.799	.530
.251	.964	.657	.251	.792	.537
.501	.938	.671	.501	.774	.551
1.001	.896	.698	1.001	.734	.569
1.601	.850	.730	1.601	.691	.580
2.401	.789	.759	2.001	.668	.592
3.001	.753	.781	3.001	.613	.638
3.601	.718	.800	3.601	.585	.646

[a] Absorbancy at 2880 Å.

[b] Absorbancy at 3550 Å.

for these fractions. A comparison of the qualitative aspects of these spectra with those of aqueous polyiodide ion spectra was a major objective in this investigation. These spectra are displayed in Fig. 5. The significance of these spectra will be discussed in a later section.


0.3
0.2
0.1
OPTICAL DENSITY
3000
4000
5000
600
WAVELENGTH (A°)
R
Y
B
B
R
Y
N
N
B
R

RESULTS AND DISCUSSION

Elimination of Parameters from the Regression Model

Allowance is made in the regression model as set forth by Eq. 25 for the possible existence of eight different polyiodide ions in the systems under investigation. Since it is quite likely that certain of these ionic species do not actually occur in these systems, it would greatly simplify the mathematical analysis of the data if some evidence could be brought forth for the elimination of some of these ions from consideration. Similarly, it would be of great value if evidence could be obtained which might be construed as strongly indicative of the presence of any of these polyiodides in the systems studied. The properties of the polyiodide formation function emphasized by Eqs. 12 and 13 suggest that a preliminary qualitative examination of Φ in the limiting conditions of zero concentration of the reactants as a function of (I^-) and (I_2) should shed some light on this subject.

If $I_4^=$ formation actually occurs in the systems under study, Eq. 12 predicts that $\Phi_{(I_2)=0}$ should be a linear function of (I^-). Since K* is a good approximation to Φ in this same limit, it is to be expected that the same functional relationship would exist between **K*** and (I^-) at zero I_2 concentration. The value of K* at zero I_2 concentration was obtained by extrapolation of the observed values of K* as a function of (I_2). These extrapolated values were obtained from Fig. 2 and are tabulated in Table 14. The functional dependence of $K^*_{(I_2)=0}$ upon (I^-) is illustrated graphically in Fig. 3. This obvious linear dependence in accordance with the prediction of Eq. 12 is taken as strong evidence for the formation of $I_4^=$ in these solutions, and for the inclusion of this term in the regression model.

Table 14. Extrapolated values for K^* at $(I_2) = 0$ as a function of (I^-)

$K^*_{(I_2)=0}$	(I^-)
743	.049494
750	.098988
770	.29696
791	.49453
812	.69290
831	.84077
846	.98216

In view of the fact that the functional relationship predicted by Eq. 12 is so exactly realized, the simple linear regression suggested by this relationship was performed in order to obtain estimates of K_3 and K_4. The value of K_3 obtained in this manner as an estimate of the intercept in

Fig. 6. Limiting values of K* at zero I_2 concentration as a function of the KI concentration in moles/liter

These values were obtained by extrapolation from the data of Fig. 2, and also represent the theoretical values for Φ, the polyiodide formation function, at zero I_2 concentration.

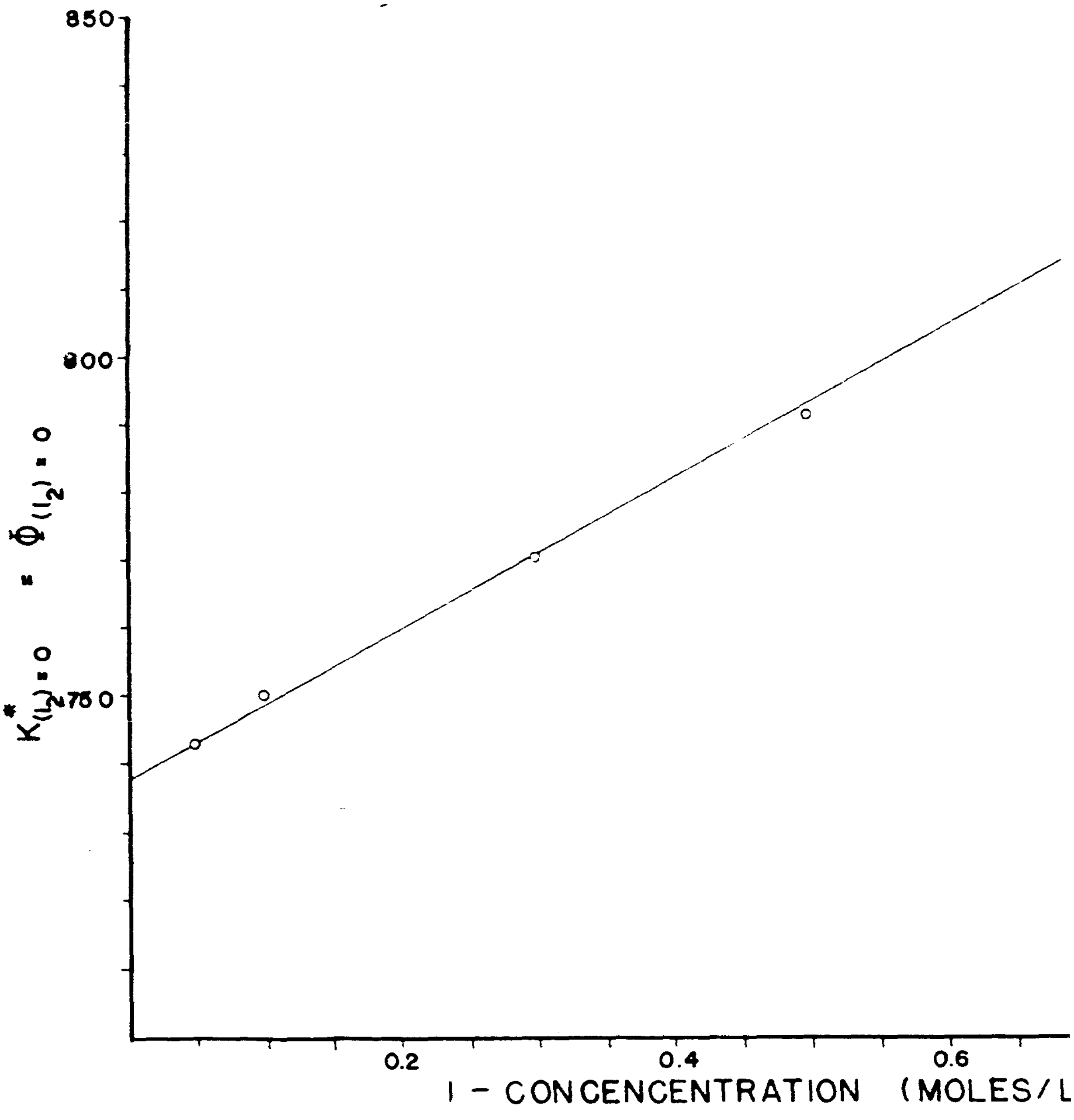

850
800
750
$K^*_{(I_2)=0} = \Phi_{(I_2)=0}$
0.2
0.4
0.6
I - CONCENCENTRATION (MOLES/L

Fig. 6 is 737.81 ± 0.97. The value of K_4 from this regression as estimated by the slope of the line in Fig. 6 is 109.49 ± 0.81.

It should also be noted that if polyiodide ions such as I_5^-, I_7^-, I_9^-, or any ion of the type I_{2n+1}^-, other than I_3^-, are formed in the systems investigated, it is predicted by Eq. 13 that $\Phi_{(I^-)=0}$ would be a function of (I_2). The data of Tables 6 through 12 has been graphically displayed in Fig. 7. One of the outstanding features of this graph is the fact that the value of Φ at zero I^- concentration appears to be independent of the I_2 concentration. The contours of constant I_2 concentration were obtained by interpolation along the experimentally determined contours. The significance of this independence of $\Phi_{(I^-)=0}$ and (I_2) is that no significant contribution to polyiodide formation in the systems under investigation is made by an ion of the type I_{2n+1}^- other than I_3^-.

Regression Models

Full model

In view of the fact that three of the eight parameters of the regression model as set forth by Eq. 28 may be excluded by consideration of the qualitative properties of Φ, the original model may be reduced to the following five parameter model.

Fig. 7. Polyiodide formation function as a function of the free I^- concentration in moles/liter

Contours of constant free I_2 were obtained by extrapolation from the data of Tables 6 through 12.

A = 0.49898 M KI

B = 0.098988 M KI

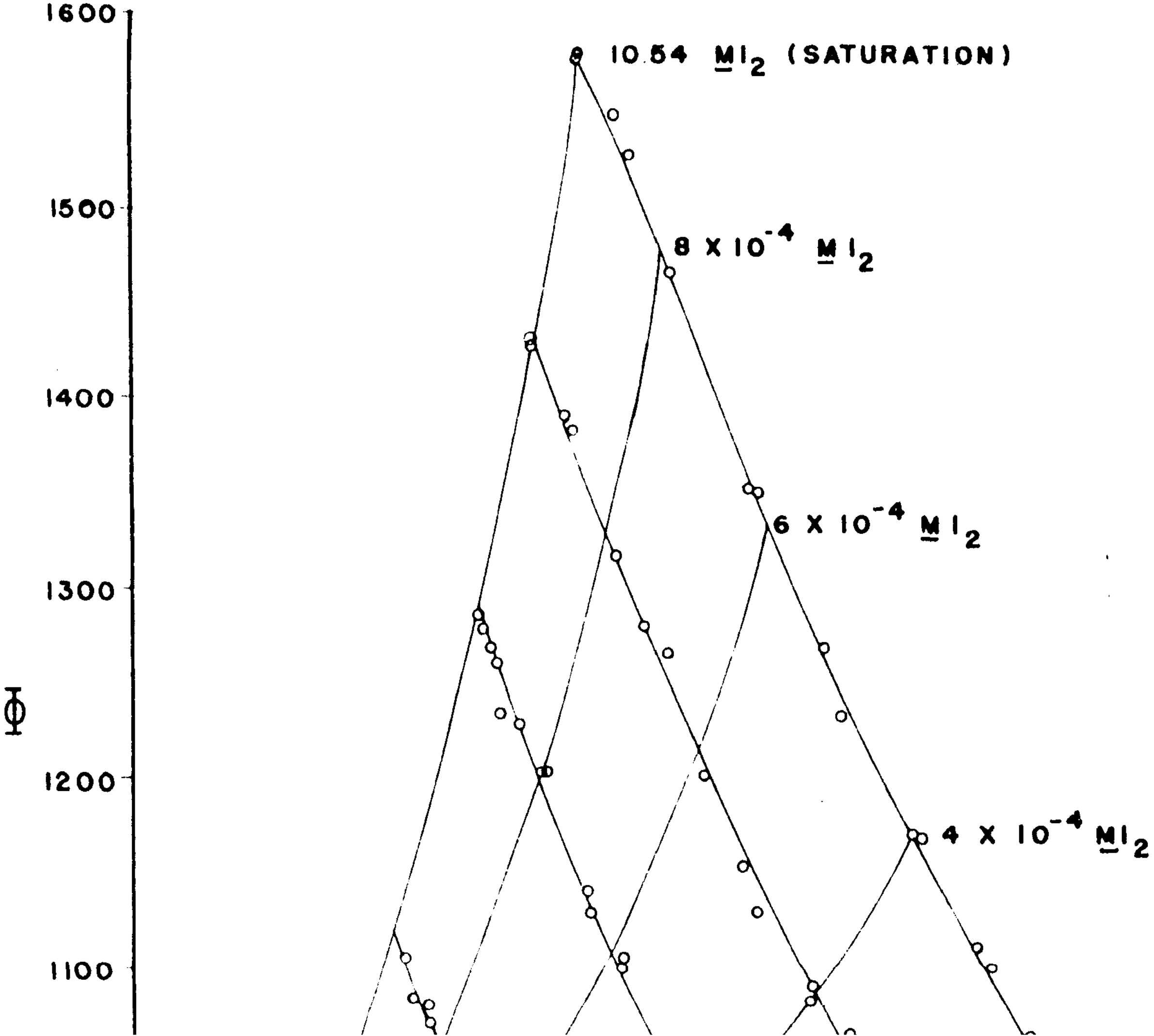
C = 0.29696 M KI

D = 0.49453 M KI

E = 0.69290 M KI

F = 0.84077 M KI

G = 0.98216 M KI


1600
1500
1400
1300
1200
1100
Φ
10.54 $\underline{M}$ I_2 (SATURATION)
8 X 10^{-4} $\underline{M}$ I_2
6 X 10^{-4} $\underline{M}$ I_2
4 X 10^{-4} $\underline{M}$ I_2

$$\Phi = A_0 + A_1 x_1 + A_2 x_2 + A_3 x_3 + A_4 x_4 \qquad \text{(Eq. 31)}$$

where $A_0 = K_3$, $A_1 = K_4$, $A_2 = 2K_6$, $A_3 = 3K_8$, $A_4 = 3K_9$

$x_1 = (I^-)$, $x_2 = (I_2)(I^-)$, $x_3 = (I_2)^2(I^-)$, and

$x_4 = (I_2)^2(I^-)^2$

Since several observations of Φ are actually obtained experimentally, one may represent Eq. 31 in the more general form,

$$\Phi_j = A_{0j} + \sum_i A_i x_{ij} \qquad \text{(Eq. 32)}$$

where $i = 1, 2, 3$, and 4

$j = 1, 2, 3, \ldots, n$, where n is the number of observations on Φ.

The theory of multiple regression used in this analysis may be found in Kempthorne (55). In summary, it may be simply stated that the best, linear, unbiased estimators for the parameters of a model such as the one expressed by Eq. 32 may be obtained by a straightforward application of matrix methods. The variance of these parameters may also be obtained from the variance-covariance matrix elements and the basic estimate of variance.

The actual computations were conducted with the aid of the IBM 650 computer. The estimated values for the regression coefficients together with their estimated variances are tabulated in Table 12. The variance-covariance matrix for this model is given in Table 13. The estimates for the

Table 18. Variance-covariance matrix for full regression model

2.906081 x 10^{-1}	-8.712229 x 10^{2}	7.529440 x 10^{5}	-3.417302 x 10^{5}
- .712229 x 10^{2}	8.009475 x 10^{6}	-2.758863 x 10^{9}	-1.560387 x 10^{10}
7.529440 x 10^{5}	-2.758863 x 10^{9}	7.706015 x 10^{12}	-1.209925 x 10^{13}
-3.417302 x 10^{6}	1.560387 x 10^{10}	-1.209925 x 10^{13}	6.861863 x 10^{13}

Table 16. Estimated regression coefficients for full and restricted models[a]

Full model: $A_0 = 745.016$ $V(A_0) = 10.7704$

$A_1 = 137.378$ $V(A_1) = 72.8777$

$A_2 = 3.8672 \times 10^5$ $V(A_2) = 29.0974 \times 10^8$

$A_3 = 4.8641 \times 10^8$ $V(A_3) = 27.9986 \times 10^{14}$

$A_4 = 2.3518 \times 10^9$ $V(A_4) = 24.9283 \times 10^{15}$

Model excluding $I_8^=$ (A_3):

$A_0 = 754.757$

$A_1 = 89.8611$

$A_2 = 5.60865 \times 10^5$

$A_4 = 3.11849 \times 10^9$

Model excluding I_9^{-3} (A_4):

$A_0 = 717.116$

$A_1 = 149.090$

$A_2 = 8.62989 \times 10^5$

$A_3 = 9.01083 \times 10^8$

---

[a] The variance of these estimates is given only for the full model, since tests of significance indicated that the restricted models were not as good as the full model in fitting the data.

formation constants of the polyiodide ions associated with these regression coefficients are tabulated as Table 19.

Restricted models

It is informative to eliminate certain of the parameters from the regression model, and to perform an analysis of the data excluding this term from the model. It is possible by such technique to perform a statistical test which will ascertain whether or not the parameter excluded actually contributed significantly to a reduction of the sums of squares due to deviations about regression. From a physical point of view, such a procedure is actually testing whether or not the formation constant for any given polyiodide ion, as estimated by regression technique, is significantly different from zero. Consequently, this procedure allows one to ascertain whether or not the existence of any given polyiodide ion is required to explain the experimental observations.

Since independent evidence for the existence of I_3^- and $I_4^=$ exists and the investigations of Davies and Gwynne (33) point strongly to the existence of $I_6^=$, only two restricted models were considered. One such model excluded $I_8^=$ from consideration, and the other excluded I_9^{-3} from consideration. The variance-covariance matrices associated with these restricted models are given in Tables 17 and 18. The estimated regression coefficients from these models appear in Table 15.

Table 17. Variance-covariance matrix for model excluding $I_5^{=}$ (A_3)

1.270570×10^{-1}	-4.016882×10^{2}	8.404707×10^{5}
-4.016882×10^{2}	7.021760×10^{6}	-1.793858×10^{10}
6.404712×10^{5}	-1.793858×10^{10}	4.962154×10^{13}

Table 18. Variance-covariance matrix for model excluding I_9^{-3} (A_4)

1.989042×10^{-1}	-7.389721×10^{2}	6.926881×10^{5}
-7.389721×10^{2}	5.312461×10^{6}	-5.157582×10^{9}
6.926881×10^{5}	-5.157582×10^{9}	5.572601×10^{12}

The corresponding estimates of the formation constants are tabulated in Table 19.

Tests of Significance

The regression analysis of this data may be summarized in the analysis of variance which appears in Table 20. In order to determine whether or not either of the restricted models is virtually equivalent to the full model in accounting for the deviations about regression, it is simply required to examine the expected mean squares which are tabulated. The theory of such testing is discussed by Kempthorne (55). The essential point to realize is that if either of the

Table 15. Estimated polyiodide formation constants for full and restricted models[a]

Full model:	$K_3 = 745.016 \pm 3.282$
	$K_4 = 137.378 \pm 8.537$
	$K_6 = 1.9336 \times 10^5 \pm 0.2697 \times 10^5$
	$K_8 = 1.6214 \times 10^8 \pm 0.1764 \times 10^8$
	$K_9 = 7.3592 \times 10^8 \pm 0.5263 \times 10^8$
Model excluding $I_8^=$:	$K_3 = 754.756$
	$K_4 = 89.851$
	$K_6 = 2.8043 \times 10^5$
	$K_9 = 1.0385 \times 10^9$
Model excluding I_9^{-3}:	$K_3 = 717.116$
	$K_4 = 149.090$
	$K_6 = 4.2648 \times 10^5$
	$K_8 = 3.0036 \times 10^8$

[a] These formation constants are for polyiodide formation at 25 ± .02 °C and at an ionic strength of 2.00.

restricted regression models is equally as good as the full model in fitting the data, then the single degree of freedom due to the fitting of any one parameter after fitting the remaining parameters is associated with a mean square which should simply be an estimate of error. Thus, one should be able to perform a statistical test of significance for this

Table 20. Analysis of variance for full and restricted regression models

Source	Degrees of freedom	Sum of squares	Mean square
Regression on A_1, A_2, A_3, and A_4	4	6.747123×10^6	
Regression on A_1, A_2, and A_3	3	6.716421×10^6	
Regression on A_3 after fitting A_1, A_2, and A_4	1	3.0702×10^4	3.0702×10^4
Regression on A_1, A_2, and A_4	3	6.666496×10^6	
Regression on A_4 after fitting A_1, A_2, and A_3	1	8.0627×10^4	8.0627×10^4
Deviation about regression	160	5.81260×10^4	363.288
Total	164	6.805249×10^6	

single orthogonal degree of freedom.

The mean square due to deviations about regression in the full model and the mean square due to the fitting of some additional parameter after fitting the restricted model would both be estimates of error if the additional parameter was actually not significantly different from zero. The ratio of two independent estimates of error is an F statistic. It is possible to test the hypothesis that the parameter excluded in the restricted model is not significantly different from zero by means of the usual F test. The statistical criterion to be used is $F_{1,160}$ since the number of degrees associated with the error estimates are 1 and 160 respectively.

The F ratio calculated for a test of significance of A_3 is 54.4, and for the significance of A_4 the F ratio is 222. In view of the fact that $F_{1,160}$ is only 6.805 even at the 1% level, these calculated F values are both highly significant and indicate that the restricted models are not nearly as good as the full model in accounting for the data. Thus, there is no evidence that $I_8^{=}$ and I_9^{-3} do not exist, but are polyiodides whose existence in the systems studied is rather likely.

Spectrophotometric Evidence for $I_4^=$ Formation

The spectrophotometric behavior of solutions of I_2 in rather concentrated I^- was investigated in an effort to obtain some corroborative evidence for the existence of polyiodides of higher molecularity in I^- than I_3^-. The exceedingly high molar absorbancies of such solutions precluded the attainment of all but the most moderate concentrations of total I_2. As may be seen in Figs. 3 and 4, even when the total I_2 concentration is of the order of 10^{-4} and the length of the path traversed by the light is reduced to .05 cm, the absorbancy is nearly unity. It was still possible under these conditions to hope to gain some evidence for the formation of $I_4^=$.

The most salient feature of aqueous solutions of I_2 and I^- is the appearance of two prominent maxima in the ultraviolet spectrum at about 2900 Å and 3500 Å, neither of which is present in aqueous solutions of I_2 alone. The relative intensities of these maxima have been observed by numerous investigators to remain very nearly constant. The maxima have been universally attributed to the I_3^- ion. Indeed, the determination of the equilibrium constant for I_3^- formation has been performed at several temperatures by Awtrey and Connick (39) by making use of the extinction coefficient of I_3^- at 2900 Å.

The I^- concentrations investigated in the present work covered a far greater range than has usually been subjected to investigation. Although no new spectral maxima or substantial shifts in the position of maxima were observed, a most unexpected behavior of the usual ultraviolet maxima was observed. It is evident from the data of Table 13 that the two maxima which have long been attributed to I_3^- alone cannot in fact belong to only one ionic species under the conditions of these experiments. It is immediately obvious that although the 2880 Å absorbancy rises to a maximum and then sharply decreases as the I^- concentration is increased, the 3550 Å maximum rises rapidly at first and then more slowly as the I^- concentration increases. Nonetheless, the higher wavelength maximum continues to increase even when the lower wavelength maximum is rapidly decreasing. This is not the behavior one would observe if a single absorbing species were responsible for both maxima.

The behavior of these spectra as illustrated in Figs. 3 and 4 is reminiscent of the spectra obtained from dyes which undergo color changes at various values of pH due to the interconversion of an acidic and basic form. One may even note the presence of an isosbestic point at about 3050 Å. It is, therefore, suggested by analogy that the interconversion of I_3^- and an ion containing one more I^- component should account for these observations. Thus, the data was quantitatively analyzed to obtain an estimate of K_4', the equilibrium constant for the reaction

$$I_3^- + I^- \rightleftharpoons I_4^= \qquad \text{(Eq. 33)}$$

The absorbancy, A, at any given wavelength of a solution in which I_3^- and $I_4^=$ are the only absorbing species may be expressed as follows:

$$A = d\ e_3\ (I_3^-) + d\ e_4\ (I_4^=) \qquad \text{(Eq. 34)}$$

where d is the path length of the incident light through the solution

e_3 is the molar absorbancy index of I_3^- at this wavelength

e_4 is the molar absorbancy index of $I_4^=$ at this wavelength

and the parentheses indicate molar concentrations.

In solutions containing very little free I_2, the total I_2 concentration may be expressed as the sum of the concentrations of I_3^- and $I_4^=$. If $(I_2)_t$ is held constant, one may write

$$(I_2)_t = (I_3^-) + (I_4^=) = c \qquad \text{(Eq. 35)}$$

Eliminating $(I_4^=)$ through the equilibrium constant for the reaction expressed in Eq. 33, and solving for (I_3^-), one obtains

$$(I_3^-) = c \ / \left[1 + K_4^{'}\ (I^-)\right] \qquad \text{(Eq. 36)}$$

Since

$$(I_4^=) = K_4^{'}\ (I_3^-)\ (I^-) \qquad \text{(Eq. 37)}$$

then

$$(I_4^=) = K_4^{'}\ c\ (I^-) \ / \left[1 + K_4^{'}\ (I^-)\right] \qquad \text{(Eq. 38)}$$

Upon substitution for (I_3^-) and $(I_4^=)$ from Eqs. 36 and 38 into Eq. 34, one obtains

$$A = \left[d\ e_3\ c + d\ e_4\ c\ K_4'\ (I^-)\right] /\ 1 + K_4'\ (I^-) \qquad \text{(Eq. 39)}$$

The quantity, $d\ e_3\ c$, contains only known parameters and experimentally determinable quantities and may be defined as D.

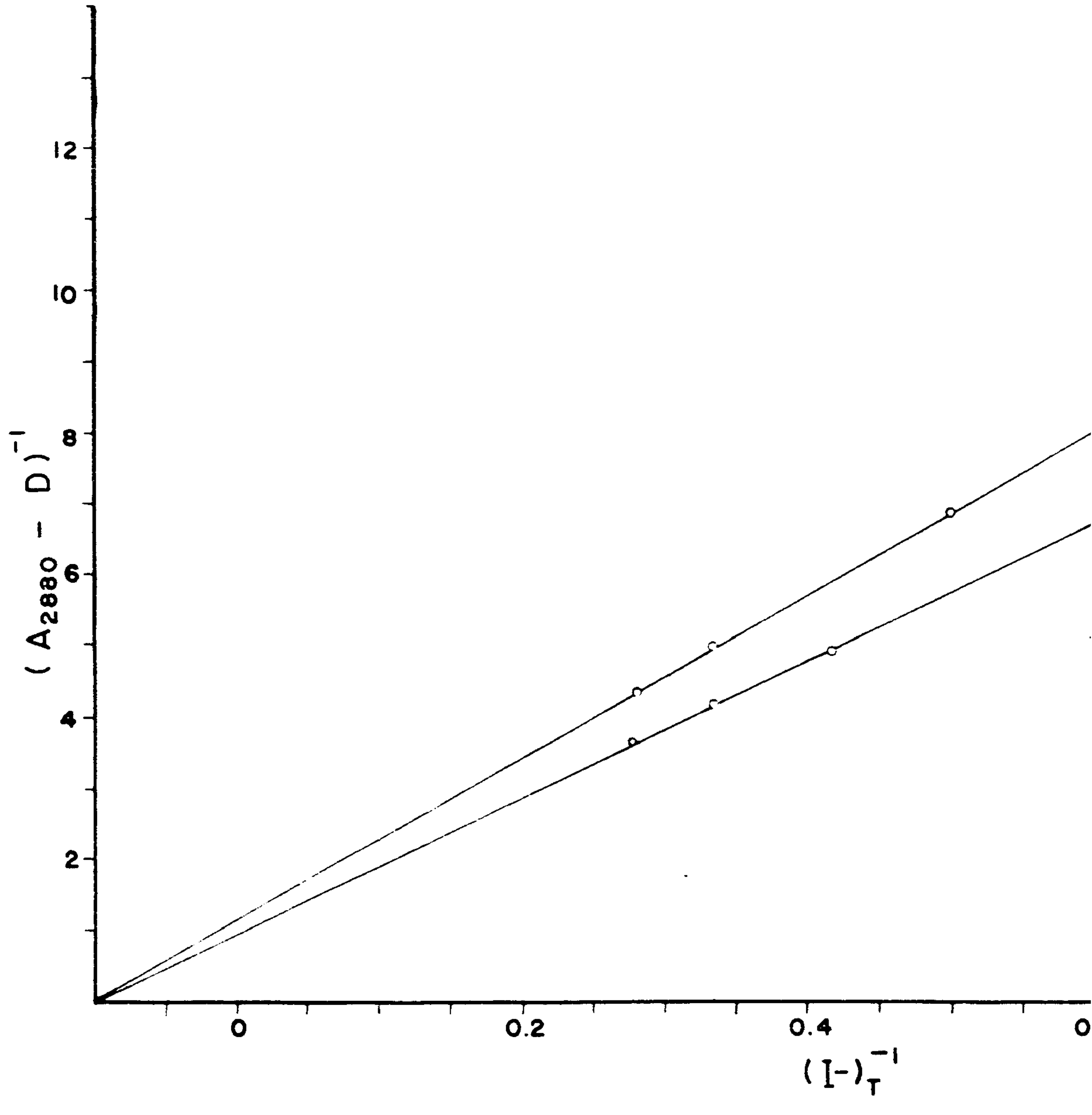
One may rearrange Eq. 39 to obtain the following form which may be readily analyzed due to the linear relationship suggested.

$$\frac{1}{A - D} = \frac{1}{D + d\ e_4\ c} + \frac{1}{K_4'\ (D + d\ e_4\ c)\ (I^-)} \qquad \text{(Eq. 40)}$$

It is evident that a linear relationship should exist between $1 / (A - D)$ and $1 / (I^-)$.

If $(I^-)_t$ is very much larger than $(I_2)_t$, one may approximate (I^-) by $(I^-)_t$. It should be noted that the ratio of the intercept to the slope is actually an estimate of K_4'. The linearity of the experimentally obtained data for solutions equal to or greater than 1.00 $\underline{M}$ in KI is shown in Fig. 8 for two different I_2 concentrations. A linear regression analysis of $1 / (A - D)$ on (I^-) for these same solutions was performed using the observed absorbancy at 2880 Å and a molar absorbancy index of 4.0×10^4 for I_3^-. This data was taken from Table 15. The estimates obtained for K_4' by this analysis were 0.107 and 0.100 for the two different total I_2 concentrations. This agreement is considered to be rather good.

Fig. 8. Reciprocal of (A_{2880} - D) as a function of the reciprocal of the total I^- concentrations for solutions in which the total I^- concentration is approximately equal to the free I^- concentration (the intercept on the abscissa is an estimate of $-K_4'$)


$(A_{2880} - D)^{-1}$
12
10
8
6
4
2
0
0.2
0.4
0
$(I^-)_T^{-1}$

The value of K_4' calculated from the ratio of K_4 to K_3 obtained by the regression analysis of the polyiodide formation function is 0.184. These values agree in order of magnitude, and in view of the fact that the conditions in the solutions used to obtain the two estimates were quite different, this discrepancy is not considered large.

Amylose-polyiodide Complexes

The most prominent feature of the absorption spectra of the various complexes separated by electrokinetic means is the appearance of very pronounced visible maxima. The blue complex is characterized by a maximum at 5900 Å and the red complex is characterized by a maximum at 5120 Å. In the case of the yellow-orange complex, absorption of light in the near ultraviolet region of the visible spectrum is also somewhat higher than that of an $I_2 - I^-$ solution containing no amylose.

The fact that the spectra of aqueous solutions of polyiodides are so dissimilar to those obtained in the presence of amylose is taken as strong evidence that the type of polyiodide ion which constitutes the core of the amylose iodine complex is of an entirely different nature from the polyiodide ions encountered in free solution.

It is entirely possible that the polyiodide ions in solution do not possess the same spectral characteristics as those in the amylose helix simply because they are not constrained to form a linear array as must those in the helix. It is also possible that the formation of I_5^-, I_7^-, and I_9^-, although of little importance in the absence of amylose, may be of considerable importance in the formation of a helical complex with amylose.

SUMMARY

1. The distribution coefficient for I_2 between CCl_4 and 2.00 M KNO_3 has been determined at 25° C. The KNO_3 appears to act merely as an inert diluent in this system.
2. The formation of various polyiodide ions in aqueous solutions at moderate concentrations of KI and over the entire solubility range of I_2 has been investigated. A qualitative examination of the data seems to indicate that the formation of ions such as I_5^-, I_7^-, and I_9^- is of no great importance in these systems. The rather surprising observation was made that ions such as $I_4^=$, $I_6^=$, $I_8^=$, and I_9^- appear to explain the data satisfactorily in terms of specific interactions.
3. A multiple regression analysis of the data led to the estimation of formation constants for I_3^-, $I_4^=$, $I_6^=$, $I_8^=$, and I_9^{-3} at 25° C in solutions of ionic strength 2.00.
4. Spectrophotometric evidence has been obtained for the formation of $I_4^=$ from I_3^- and I^- in aqueous solutions. An estimate of the extent to which this reaction occurs was in fair agreement with that obtained in the regression analysis.
5. Fractionation of amylose-polyiodide complexes was achieved by means of electrophoresis on a hanging glass curtain. This technique appears to be a good method for preparing small quantities of homogeneous low molecular weight

amylose.

6. The polyiodides formed in aqueous solution in the absence of amylose appear to be of an entirely different nature from those formed in the presence of amylose.

BIBLIOGRAPHY

1. Gay-Lussac, J. L., Ann. de chemie, 91, 72 (1814)
2. Baudrimont, E., Compt. rend., 51, 527 (1860)
3. Dossios, L. and Weith, W., Z. für Chemie, 5, 380 (1869)
4. Johnson, G. S., J. Chem. Soc., 249 (1877)
5. Wells, H. L. and Wheeler, H. L., Z. anorg. Chem., 1, 442 (1892)
6. Le Blanc, M. and Noyes, A. A., Z. physik. Chem., 6, 401 (1890)
7. Jakowkin, A. A., Z. physik. Chem., 13, 539 (1894)
8. Jakowkin, A. A., Z. physik. Chem., 18, 585 (1895)
9. Jakowkin, A. A., Z. physik. Chem., 20, 19 (1896)
10. Noyes, A. A. and Seidensticker, J., Z. physik. Chem., 27, 357 (1898)
11. Fedotieff, P. P., Z. anorg. Chem., 69, 32 (1911)
12. Korenmann, I. M., J. Gen. Chem. (USSR), 17, 1608 (1947)
13. Abegg, R. and Hamburger, A., Z. anorg. Chem., 50, 426 (1906)
14. Linhart, G. A., J. Am. Chem. Soc., 40, 162 (1918)
15. Foote, H. W. and Chalker, W. C., Am. Chem. J., 39, 561 (1908)
16. Dawson, H. M. and Gawler, R., J. Chem. Soc., 524 (1902)
17. Dawson, H. M. and Goodson, E., J. Chem. Soc., 796 (1904)
18. Mooney, R. C. L., Z. Kristallogr., 90, 143 (1935)
19. Mooney, R. C. L., Acta Cryst., 12, 187 (1959)
20. Slater, J. C., Acta Cryst., 12, 197 (1959)

21. Rundle, R. E. On the Polyhalides. (Typewritten) Dept. of Chemistry, Iowa State University of Science and Technology, Ames, Iowa. 1961.

22. Rundle, R. E. and Hach, R. J., J. Am. Chem. Soc., 73, 4321 (1951)

23. Rundle, R. E., James, W. J., Hach, R. J. and French, D., Acta Cryst., 8, 814 (1955)

24. Havinga, E. E., Boswijk, K. H., and Wiebenga, E. H., Acta Cryst., 7, 487 (1954)

25. Lewis, G. N., Proc. Am. Acad., 43, 259 (1907)

26. Parsons, C. L. and Corliss, H. P., J. Am. Chem. Soc., 32, 1367 (1910)

27. Parsons, C. L. and Whittemore, C. F., J. Am. Chem. Soc., 33, 1934 (1911)

28. Dawson, H. M., J. Chem. Soc., 238 (1901)

29. Bray, W. C. and McKay, G. M. J., J. Am. Chem. Soc., 32, 914 (1910)

30. Washburn, E. W. and Strachan, E. K., J. Am. Chem. Soc., 35, 691 (1913)

31. Linhart, G. A., J. Am. Chem. Soc., 40, 158 (1918)

32. Laurie, A. P., Z. physik. Chem., 67, 627 (1900)

33. Davies, M. and Gwynne, E., J. Am. Chem. Soc., 74, 2748 (1952)

34. Jones, G. and Kaplan, B. B., J. Am. Chem. Soc., 50, 1845 (1928)

35. Allsopp, C. B., Proc. Roy. Soc. London, A. 158, 167 (1937)

36. Wintner, C., Z. physik. Chem., 108, 236 (1924)

37. Forster, E. O. The Influence of Iodide Ion, of Amylose, and Certain Other Polysaccharides on the Spectrum of Iodine in Aqueous Solutions. Unpublished Ph.D. Dissertation. New York, Columbia University Library. (1951)

38. Symons, M. C. R. and Doyle, W. T., Q. Revs., 14, 62 (1960)

39. Awtrey, A. D. and Connick, R. E., J. Am. Chem. Soc., 73, 1842 (1951)

40. Myers, O. E., J. Chem. Phys., 28, 1027 (1958)

41. Jones, G. and Hartmann, H. L., J. Am. Chem. Soc., 37, 241 (1915)

42. Rundle, R. E. and Baldwin, R. R., J. Am. Chem. Soc., 65, 554 (1943)

43. Rundle, R. E. and French, D., J. Am. Chem. Soc., 65, 558 (1943)

44. Schoch, T. J., Adv. in Carbohydrate Chem., 1, 247 (1945)

45. Baldwin, R. R., Bear, R. S., and Rundle, R. E., J. Am. Chem. Soc., 66, 111 (1944)

46. Swanson, M. A., J. Biol. Chem., 172, 825 (1948)

47. Thoma, J. A. and French, D., J. Am. Chem. Soc., 82, 4144 (1960)

48. Bates, F. L., French, D., and Rundle, R. E., J. Am. Chem. Soc., 65, 142 (1943)

49. Gilbert, G. A. and Marriott, J. V. R., Trans. Farad. Soc., 44, 84 (1948)

50. Mould, D. L. and Synge, R. L. M., J. Biol. Chem., 58, 585 (1954)

51. Thoma, J. A., Wright, H., and French, D., Arch. Biochem. Biophys., 85, 452 (1959)

52. Thoma, J. A. and French, D. The Starch-Iodine-Iodide Interaction. Part II. Potentiometric Investigations. Manuscript accepted for publication, J. Am. Chem. Soc. (*ca*. 1961)

53. Allen, T. A. and Keefer, R. M., J. Am. Chem. Soc., 77, 2957 (1955)

54. Kiss, A. and Urmanczy, A., Z. anorg. allgem. Chem., 202, 172 (1931)

55. Kempthorne, O. Design and Analysis of Experiments. Wiley. New York (1952)

ACKNOWLEDGEMENTS

The author wishes to express his deep appreciation to Dr. Dexter French for his counsel, guidance, and patience throughout the course of this investigation, and for the many stimulating discussions concerning the interpretation of phenomenon in solution.