substance produced by photochemical action (usually photo-oxidation). These photochemical reactions are usually of the first order, but sometimes of the second order, with respect to the photons. The latter reactions are shown to be due to the action of photons on the phosphorescent state itself. The double phosphorescent state found in some dyes is due to the presence of a pseudo-isomer.

The pattern of the phosphorescence bands is so sharp and characteristic that it will serve well for the identification of substances. The whole structure of the phosphorescent band systems is due to the fall from the phosphorescent state to different vibrational excitations of the ground state. The difference between this structure and that of fluorescence is discussed. The fact that some substances show no phosphorescence is interpreted.

The energy of the phosphorescent state (that of the ground state being taken as zero) is calculated from the position of the band of highest frequency. The effect of band width upon this calculation is discussed. This characteristic energy, for eighty-nine substances, is given in Table II.

The structure of the phosphorescence bands is shown to be important in the study of the vibrational levels of the normal molecule, since it brings into prominence vibrations that are often prohibited in infrared and Raman work. Only two of the many spectra are discussed in this regard: β-chloronaphthalene and benzene. In the latter case it is shown how the identification of the band intervals with known vibrations of normal benzene permits conclusions concerning the size and shape of the phosphorescent state—conclusions that are verified in the theoretical section.

The dependence of the energy of the phosphorescent state upon the presence of auxochromes is discussed. It is also shown that the lifetime of the phosphorescent state is greatly affected by certain substituent groups. The phosphorescence of carbon dioxide, sulfur dioxide, and other inorganic substances is mentioned, but further discussion postponed.

Arguments are advanced for the identification of the phosphorescent state with the triplet, or biradical, state of the molecule, and the distinction between fluorescence and phosporescence is defined. The most conclusive evidence that the phosphorescent state is the triplet state is furnished by study of the energy of that state, for a large number of organic molecules. An equation is set up for the energy of the triplet state, as dependent upon the energy required to break a double bond, upon the repulsive energy, and upon the difference in resonance energy between the normal and the triplet states. The data verify the theory. In particular the cases of anthracene, of biphenyl and its ortho derivatives, and of thiobenzophenone afford remarkable confirmation of the theoretical predictions.

BERKELEY, CALIFORNIA RECEIVED SEPTEMBER 27, 1944

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY OF THE IOWA STATE COLLEGE]

On the Nature of the Starch–Iodine Complex[1]

BY R. E. RUNDLE, JOSEPH F. FOSTER AND R. R. BALDWIN

The nature of the interaction between starch and iodine to form the blue starch–iodine complex has interested chemists since 1812, but has never received adequate explanation.[2] Recently the structure of the complex has been made known, and further pertinent facts concerning complex formation have been revealed by the potentiometric[3] and spectrophotometric[4] titrations of starch and its components with iodine. In this paper an attempt is made to explain the nature of the starch–iodine interaction in the light of these new findings.

It has been found that only the amylose (unbranched) component of starch forms a stable blue complex with iodine,[3] so that the major portion of this paper will be devoted to the amylose-iodine complex. The less stable, red complex formed with amylopectin will, however, receive brief attention.

The structure of the amylose–iodine complex has been established, by optical[5] and X-ray[6] investigations, as a helical amylose chain within which the iodine molecules are arranged parallel to the helix axis (Fig. 1). Freudenberg and associates, who, following Hanes,[7] proposed the above structure of the starch–iodine complex, also attempted to explain the interaction of starch with iodine in terms of the structure of the complex.[8]

(1) Journal Paper No. J-1233 of the Iowa Agricultural Experiment Station. Project 660. Supported in part by a grant from the Corn Industries Research Foundation.

(2) For a review of all but the most recent literature on the starch-iodine complex see G. Barger, "Some Application of Organic Chemistry to Biology and Medicine," McGraw-Hill Book Co., Inc., New York, N. Y., 1930. pp. 127–176.

(3) F. Bates, D. French and R. Rundle, THIS JOURNAL, **65**, 142 (1943).

(4) R. Baldwin, R. Bear and R. Rundle, *ibid.*, **66**, 111 (1944).

(5) R. Rundle and R. Baldwin, *ibid.*, **65**, 554 (1943); R. Rundle and D. French, *ibid.*, **65**, 558 (1943).

(6) R. Rundle and D. French, *ibid.*, **65**, 1707 (1943); R. Rundle and F. Edwards, *ibid.*, **65**, 2200 (1943).

(7) C. S. Hanes, *New Phytologist*, **36**, 101, 189 (1937).

(8) K. Freudenberg, E. Schaaf, G. Dumpert and T. Ploetz, *Naturwissenschaften*, **27**, 850 (1939).

They constructed a space model of an amylose helix assuming a *wannen* form for the pyranose ring of glucose. The resulting helix had a hydrocarbon-like interior with the (OH) groups directed outward from the helix. From this model Freudenberg, *et al.*, proposed that the iodine complex is simply a solution of iodine within the non-polar, hydrocarbon-lined interior of the helix.

In spite of the confirmation of the helical structure of the complex, complex formation appears to involve more than solid solution of iodine in amylose[3]; moreover, the type of interaction proposed by Freudenberg fails to account for the change of color and stability of the complex with change in chain length of the amylose.[3,4] The interpretation of the interaction presented below not only accounts for these features of the amylose–iodine complex but also contains certain points which seem applicable to the many other materials which form highly colored complexes with iodine.

Evidence for Compound Formation.—As shown by the potentiometric titration,[3] when iodine is added to an amylose solution the iodine activity remains very low and nearly constant until one iodine molecule for every six to eight glucose residues in the amylose has been added. (The exact value depends on the iodide activity in the solution.) Solid solution of iodine in amylose should produce amylose–iodine complexes of increasing iodine content and increasing iodine activity as the titration proceeds. It appears, instead, that an amylose–iodine complex of constant iodine activity, and hence probably of constant composition, is formed during the titration. That this is the case was demonstrated by the following experiment.

To a 0.04% amylose solution containing 0.01 *m* potassium iodide was added one-half the iodine needed to reach the end-point of the potentiometric titration. The amylose–iodine complex was precipitated by the addition of excess potassium iodide, and the precipitate was removed by centrifuging. The precipitate contained only half the amylose present in the solution, but it contained all the iodine. So free was the solution from iodine that it was no longer colored, though the amylose concentration was 0.02%, as was easily demonstrated by further titration with iodine.

If the above experiment is interpreted in terms of the amylose–iodine structure it seems necessary to conclude that the escaping tendency of iodine from a partially filled helix is greater than its escaping tendency from a completely filled helix. This is, of course, far from the behavior expected for solution of iodine in amylose.

Amylose Chain Length and Iodine Activity of the Complex.—When amyloses from different plant sources were titrated with iodine the iodine activity during the main part of the titration was a constant and was characteristic of the plant

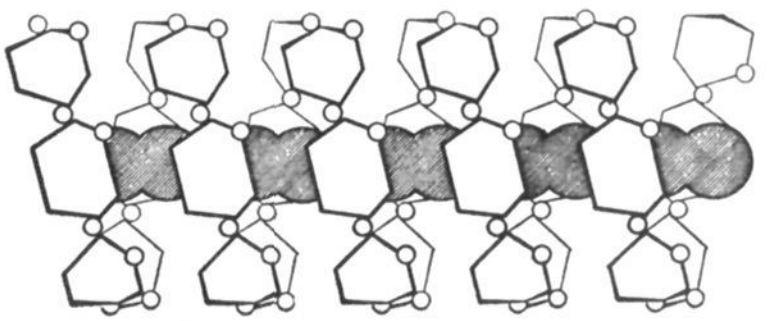

Fig. 1.—Model of an iodine-filled amylose helix.

source of the amylose.[3] These variations were attributed to differences in chain lengths of amyloses from different sources; longer chains apparently produced complexes of lower iodine activity. This has been confirmed by viscosity and osmotic determinations[9] of the molecular sizes of the same amyloses examined by the potentiometric titration. Moreover, it has been shown that a potentiometric titration of a mixture of potato and corn amyloses gave a fairly sharp increase in the iodine activity, characteristic of the difference in iodine activity of the two amylose–iodine complexes, after enough iodine had been added to saturate the potato amylose.[3] It appears that the longer chain (potato) amylose was titrated completely before the shorter chain amylose acquired any iodine.

From the above we conclude not only that a *helix filled with iodine has a lower iodine acivity* than a partially filled helix, but also that the longer the filled helix the lower the iodine activity. Thus it seems that anything which increases the number of iodine molecules arranged linearly within the amylose helix lowers the escaping tendency of iodine from the helix.

Amylose–Iodine Absorption Spectra and Chain Length.—The proposal that iodine in starch is dissolved in a hydrocarbon-like interior of a starch helix is supported by the fact that the colors of starch–iodine solutions and iodine in non-polar solvents are similar.[8] The similarity is not great (Fig. 2), and is somewhat fortuitous; for example, long chain amyloses form iodine complexes which are bluer than solutions of iodine in non-polar solvents, while short chain amyloses form red complexes. It is over only a narrow range of chain lengths that the colors of the two are similar.[4]

More striking than the similarity in color is the difference in molecular extinction coefficients of iodine in polar and nonpolar solvents and iodine in starch solutions. As can be seen in Fig. 2, the molecular extinction coefficient for iodine in starch is an order of magnitude greater than for iodine in water or carbon tetrachloride. The molecular extinction coefficient for pure potato amylose is about 40,000 as compared with 17,000 for the potato starch shown in Fig. 2,[4] so large that it is not well represented on the same graph with iodine in carbon tetrachloride.

(9) J. Foster and R. Hixon, THIS JOURNAL, **65**, 618 (1943); **66**, 557 (1944).

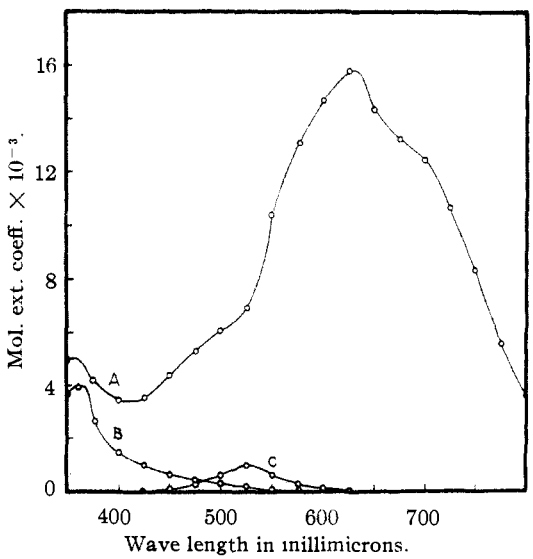

Fig. 2.—Molecular extinction coefficients of: A, 0.00005 molar iodine in 0.01% potato starch solution; B, 0.0001 molar iodine in water; C, 0.001 molar iodine in carbon tetrachloride.

The transmission curves used in obtaining the data for Fig. 2 were determined in the manner described in an earlier paper.[4] As used in Fig. 2 the molecular extinction coefficient K is defined as

$$K = -\log T/cd$$

where T is the fractional transmission, c is the concentration of iodine in moles per liter, and d is the thickness of the absorbing solution.

Comparison of the absorption of iodine in the three solutions was made on the basis of molecular extinction coefficient, since for conveniently measured transmissions different concentrations of iodine were required in the three cases. For solutions which follow Beer's law the molecular extinction coefficient is independent of concentration. Iodine in the above solutions does not follow Beer's law[10]; for solutions of iodine in water and carbon tetrachloride the molecular extinction coefficient increases with iodine concentration, while for iodine in starch and amylose solutions it decreases with iodine concentration at a given starch or amylose concentration. The deviations from Beer's law are, however, not such as to invalidate the comparison of magnitudes shown in Fig. 2 at any obtainable concentrations.

In connection with the alleged similarity of iodine in non-polar solvents to iodine in starch, it is interesting to note that iodine in water has a greater molecular extinction coefficient than iodine in carbon tetrachloride; nevertheless, it is still an order of magnitude less than the molecular extinction coefficient of iodine in starch. Rather than supporting the proposal that iodine in starch is dissolved in a hydrocarbon-like medium the absorption spectra seem to indicate that iodine in amylose is something quite different from iodine in any simple solution.

The molecular extinction coefficient, as well as the wave length of maximum absorption of the amylose–iodine complex, has been found to vary with chain length of the amylose, and hence with the number of iodine molecules per helix.[4] The molecular extinction coefficient increases with increasing chain length, and the wave length of maximum absorption also increases with chain length. In both cases the change appears to grow much less sensitive to changes in chain length as the chain length increases.

Discussion

Though the facts presented here indicate that iodine, instead of forming a solid solution with starch, forms a true compound with the amylose component of starch, the composition of the compound is constant only when formed at a given iodide concentration. It appears that iodide, or probably triiodide ion, can replace iodine in the helix, and the composition of the resultant complex is determined largely by the capacity of the amylose helix.[3,4] Also, there is no reason to believe that the complex is anything other than a molecular complex in which the iodine or triiodide ions are bound to the amylose through secondary chemical interactions. The change of the stability of the amylose–iodine complex and the change in the absorption spectra of the complex with chain length require that the interactions between amylose and iodine and/or iodine and iodine change with chain length. The nature of the iodine molecule and the linear array of iodine molecules in the complex suggest a possible mechanism for the interaction.

Let us first consider the amylose helix. The building unit of the helix is the glucose unit, consisting of a number of dipoles. This unit will have a resultant dipole moment of its own. When these units are arranged into a helix, the components of the dipoles at right angles to the helix axis should oppose each other, so that the net dipole moment normal to the helix axis is negligible or zero. But unless the resultant dipole of each glucose residue is exactly normal to the helix axis the components along the helix axis will add together, so that for a long helix, composed of many glucose residues, the resultant dipole moment should be quite large. It seems to us most probable that there should be a large dipole parallel to the helix axis whether, as Freudenberg, *et al.*, propose, the helix has a hydrocarbon lining, or not.

If an iodine molecule were introduced into the dipolar helix, with the long axis of the iodine molecule parallel to the helix axis, the iodine molecule should acquire an induced dipole. Succeeding iodine molecules introduced into the helix should acquire parallel dipoles, so that the induced dipole interactions should be attractions, and the interactions of the induced dipoles should

(10) A. Stewart and R. Wright, *J. Chem. Soc.*, 111, 183 (1917).

contribute to the strength of each. The magnitude of the induced dipole moment on each iodine molecule should increase with the number of iodine molecules arranged in parallel, and with the strength of the dipole moment on the amylose helix. The latter should increase with length.

The above arguments should require that the escaping tendency of iodine from an amylose helix become less with increasing number of iodines arranged in parallel within the helix and with the length of the helix. This is in accord with the experimental facts outlined above in this paper.

An explanation involving a polar environment for the iodine molecules is not in disagreement with the absorption spectra of the amylose–iodine complex, but rather provides a qualitative explanation for the changes in the spectra with chain length of the amylose. As pointed out above, the blue color of the amylose–iodine complex is no indication that the iodine is in a non-polar medium. Amylose–iodine complexes of short chain lengths are colored red to purple, and even became brown for the very short amylodextrins and for glycogen, where individual branches of the molecule are very short. It appears that isolated iodine molecules in the environment of the interior of the amylose helix produce a reddish-brown color. The absorption shifts to longer wave lengths as the number of interacting dipoles is increased, until for long chain amyloses the absorption is in the red, and the color of the complex is quite blue. The direction of the shift is quite in accord with expectations.

The light absorption of the complex, per iodine molecule, increases with the length of the amylose helix. This seems to require that the field of force on the iodine molecule changes in a systematic fashion with the length of the helix. The picture of complex formation offered above provides for an increased polarity as the helix increases in length due to the increased dipole moment of the helix itself and to the increased iodine–iodine interaction.

Amylopectin behaves with iodine as though solid solution or adsorption of iodine were a true representation of the case.[3] It appears likely that the short branches of the amylopectin molecule have the ability to complex with iodine in the same manner as amylose. Due to the short length of the branches the complex is not so stable, however, and since the unbranched portions are probably not all of the same length the various unbranched portions of the amylopectin form the iodine complex at different iodine activities, corresponding to their lengths. The result is that the iodine activity of the complex increases with iodine added to it as the shorter branches are filled. It has been noted that the most highly branched amylopectins, as determined by end-group assay, give the highest iodine activities for the iodine added in the potentiometric titration.[3]

Many substances are stained blue with iodine.[2] For most of these substances a structure closely analogous to the structure of the amylose–iodine complex is certainly very unlikely. We suggest, however, that many of these colored substances have one thing in common with the starch–iodine complex, *i. e.*, a parallel orientation of iodine molecules in a polar field.

Barger[2] divides the majority of substances which form blue complexes with iodine into two classes: (1) colloids, and (2) crystalline substances of low molecular weight. The colloids he divides into two subdivisions: (a) hydrophilic colloids related to starch, and (b) hydroxides and basic acetates of certain metals. Leaving the hydroxides and basic acetates out of account, there are left two types of substances which stain blue with iodine. In (a) a parallel arrangement of iodines is possible by alignment along the long chains of a macromolecule. How this arrangement is brought about with amylose has been discussed above. The Polaroid Corp. now makes a polarizing sheet from iodine-stained polyvinyl alcohol.[11] In the stretched sheet light with its electric vector parallel to the direction of stretch is preferentially absorbed, so the iodine molecules in the sheet are evidently arranged parallel to the chains of the macromolecule. It is easily demonstrated that a similar orientation of the iodine molecules takes place in cellophane sheets or cellulose fibers stained with iodine, since here too the sheets and fibers become very dichroic, preferentially absorbing light with its electric vector parallel to the cellulose chains.

In his review of crystalline iodine complexes of smaller molecules Barger reports pleochroism for many crystals.[2] Presumably many of the others have received little attention. Pleochroism is evidence of the alignment of iodine molecules in preferred directions, and is in accord with the proposals made here.

Amylose is unique, then, not in staining blue with iodine, but in staining blue in such dilute solutions that molecular dispersion of the amylose seems to be approached. In most, if not all, other blue complexes aggregates of molecules are required, and quite probably the iodine molecules are arranged between neighboring molecules. The amylose–iodine complex is different in that a single molecule envelops a parallel array of iodine molecules. It seems likely that the unique structure of the amylose–iodine complex is the explanation for its unique behavior in solution.

Summary

On the basis of the structure of amylose–iodine and the results of potentiometric and spectrophotometric titrations of starch and its components with iodine, a means of interaction of amylose with iodine is proposed. The success of the proposal

(11) E. H. Land (to Polaroid Corp.), U. S. Patent 2,237,567.

in explaining changes in stability and color of the amylose–iodine complex with changes in amylose chain length is pointed out, and some application of the proposal is made to other blue iodine complexes.

AMES, IOWA RECEIVED SEPTEMBER 18, 1944

[CONTRIBUTION FROM THE WILLIAM G. KERCKHOFF LABORATORIES OF THE BIOLOGICAL SCIENCES, CALIFORNIA INSTITUTE OF TECHNOLOGY]

Thermal Data. XVIII. The Heat Capacity, Heat of Fusion, Entropy and Free Energy of Ethylbenzene

BY GEORGE B. GUTHRIE, JR., RALPH W. SPITZER AND HUGH M. HUFFMAN[1]

Some time ago this Laboratory began a research program involving the study of certain thermal properties of hydrocarbons. Because of the importance of ethylbenzene in hydrocarbon chemistry a study was made of its low temperature thermal properties. In this paper we present the results of this investigation. These data have been utilized to calculate the entropy and free energy of liquid ethylbenzene at 298.16°K.

The Ethylbenzene.—The material used in this investigation was supplied to us in the purified condition by the Shell Development Co. An estimate of the liquid soluble–solid insoluble impurity was made from data obtained by observing the equilibrium temperatures corresponding to known fractions of the material in the solid and liquid form. The impurity estimated from the above data was 0.070 mole per cent.

Experimental.—The experimental method has been described in a recent paper by Ruehrwein and Huffman[2] and only a brief description need be given here. An adiabatic calorimetric system was used in which the material under investigation was contained in a sealed copper calorimeter. A measured quantity of electrical energy was supplied to the calorimeter and at all times during the measurements the temperature of the environment was maintained at that of the calorimeter to prevent heat interchange. The initial and final temperatures of the calorimeter were measured by means of a platinum resistance thermometer. The electrical measurements required for the determination of the energy and of the resistance of the thermometer were made on a White double potentiometer in conjunction with a high sensitivity galvanometer and accurately calibrated resistances. The precision of our measurements is in general better than 0.1% and we believe that above 30°K. the accuracy should be about the same as the precision. The energy measurements were made in terms of the international joule and were converted to the conventional calorie by dividing by 4.1833.

The ethylbenzene was distilled into the calorimeter in an air-free system. The gas space was filled with helium at one atmosphere pressure at room temperature and the calorimeter was then sealed by the application of a drop of soft solder.

The results of the heat capacity measurements are given in Table I. Most of the temperature range was covered at least twice and the results of the measurements in the different series were in excellent agreement. In Table II we have listed the values of the heat capacity at integral temperatures as selected from a smooth curve through all of the data.

(1) Present address, Bureau of Mines, Bartlesville, Oklahoma.

(2) Ruehrwein and Huffman, THIS JOURNAL, **65**, 1620 (1943).

TABLE I

MOLAL HEAT CAPACITY OF ETHYLBENZENE

Molecular weight = 106.160; 0°C. = 273.16°K.

T_M, °K.	ΔT	C_p cal./degree	T, °K.	ΔT	C_p, cal./degree
	Crystals		109.43	6.235	17.960
13.34	1.351	1.013	115.01	16.868	18.604
15.01	1.973	1.380	116.53	7.950	18.749
16.91	1.831	1.846	126.79	12.578	19.859
19.72	4.067	2.585	133.33	19.762	20.629
19.84	4.030	2.638	138.98	11.814	21.222
23.90	4.098	3.809	150.47	11.167	22.482
24.09	4.676	3.859	154.21	22.013	22.952
27.99	4.080	4.965	160.62	9.130	23.677
28.40	3.946	5.086	166.84	3.244	24.622[a]
32.28	4.493	6.150	170.04	3.158	25.464[a]
32.97	5.186	6.333	173.13	3.022	26.926[a]
37.07	5.085	7.374		Liquid	
38.59	4.217	7.728	181.51	9.249	37.648
42.06	4.889	8.513	190.70	9.152	37.936
46.39	9.538	9.375	199.81	9.056	38.257
47.14	5.288	9.524	208.81	8.954	38.626
52.28	4.994	10.480	216.27	9.601	38.982
55.96	9.605	11.120	218.15	9.733	39.048
57.76	5.971	11.436	221.14	17.426	39.211
62.40	3.916	12.177	225.80	9.473	39.446
63.91	6.284	12.410	234.98	10.249	39.989
66.21	3.696	12.755	244.70	10.074	40.532
71.14	6.165	13.445	255.52	11.553	41.246
77.09	5.741	14.272	266.96	11.346	42.016
83.19	6.452	15.028	277.41	9.556	42.787
89.95	7.063	15.874	286.89	9.406	43.504
96.82	6.692	16.617	296.23	9.254	44.275
103.37	6.413	17.318	305.41	9.113	44.994
103.43	5.766	17.320			

[a] These values include premelting.

The Melting Point.—In Table III we have given the equilibrium temperatures corresponding to the known fraction of calorimeter contents in the liquid state.[3] Utilizing these data we have calculated the melting point of the mixture in the calorimeter and also for pure ethylbenzene. The values of the melting point given in the last column of Table III were calculated, by an approximate method, on basis of an impurity of 0.070 mole per cent. From these data we conclude that the melting point

(3) The value given as per cent. liquid is actually the per cent. of the calculated heat of fusion of the contents of the calorimeter but for practical purposes this may be taken as representing the per cent. of the ethylbenzene in the liquid form.