CHAPTER III

REFRACTOMETRIC AND DIFFERENTIAL REFRACTOMETRIC STUDIES ON THE CHARGE TRANSFER INTERACTION & ION PAIR FORMATION OF BIOMOLECULES WITH SOME $\pi$- AND $\sigma$-ACCEPTORS

3.1 INTRODUCTION

In recent years, the chemistry of molecular complexes and ion-pairs have experienced an impressive renaissance. The stability of molecular complexes which mainly occurs through interaction between donor and acceptor may play an important role in understanding several fundamental mechanism involved in the chemical initiation of some vital diseases in the biological systems, the conformation of proteins and nucleic acids, the role of drug interaction, the mechanism of toxicity as well as carcinogenicity of pesticides. The formation of molecular complexes between carcinogen and

*A part of this work has been published in the Indian Journal of Pure & Applied Physics, 18, 504-9 (1980), and Monsch. Fur. Chemi. (in press)
biomolecules have been mentioned in the literature, but a detailed investigations on these interactions has not yet been explored. Thus, in order to study the role of molecular complexes in the chemical initiation of cancer on molecular level, the interaction of some biomolecules (glycine, alanine, valine, leucine, tryptophan, tyrosin, phenylalanine, indole, 2-methyl-indole, 3-methyl-indole and genetic bases) with some $\pi$-acceptor (chloranil and DDT) and $\sigma$-acceptor (iodine) have been studied in aqueous and non-aqueous solvents. These molecules have been chosen because of their following importance. The aminoacids are the unit of proteins. Indoles are the basic component of several biomolecules. Genetic bases are the unit of RNA and DNA. DDT acts as a carcinogen and benzene and naphthalene are the unit of the polycyclic aromatic hydrocarbons which are reported to be electron acceptor and donor and also the potent carcinogens. Iodine plays an important role in chemical carcinogenesis.

3.2 EXPERIMENTAL

The donors (glycine, alanine, valine, leucine, tryptophan, tyrosin and phenylalanine) were obtained from Fluka AG and have been used as such. AR grade indoles (indole, 2-me-indole and 3-me-indole) have been obtained from Fluka AG and have been recrystallized as reported earlier. Benzene and naphthalene (AR grade BDH) were purified as reported (Chapter II, Sec.2.2.3).
AR grade chloranil supplied from E. Merck was recrystallised from the standard solution of benzene. Reagent grade iodine (BDH) was used after resublimation. DDT (Aldrich Chemical, USA) was of 98.9% purity and was used as such. Ethanol was used after distilling thrice. Analytical grade carbon tetrachloride was kept over anhydrous calcium chloride for several days and it was distilled before use. Dioxane (AR grade), THF, CHCl₃ and CH₃CN (AR grade) have been purified by recommended procedures.17

Stock solution of donors and acceptor were prepared by weighing on an analytical balance and then diluted to the required volume in volumetric flasks in appropriate solvents. These were subsequently diluted with the same solvent to get the test samples.

The refractive indices have been measured by Bausch & Lomb refractometer with an accuracy of ±0.0002 at 30°C. These were measured for following four sets of solution:

(i) the refractive indices of donor solution (n_D),
(ii) the refractive indices of acceptor solution (n_A),
(iii) the refractive indices of mixed solutions (n) of donor and acceptor, and
(iv) the refractive indices of molecular complex and binary mixture of THF-dioxane solutions.
3.3 DATA ANALYSIS

The $K_1$ and extent of electronic polarization ($\lambda$) have been calculated by using Eqs. (3.1 - 3.4) recently developed by Sahai et al.\textsuperscript{18-20}

\begin{align*}
\delta \varphi / C_D^0 &= \left\{ K_1 C_A^0 / \lambda (1 + K_1 C_A^0) \right\} - \left\{ K_1 \delta \varphi / (1 + K_1 C_A^0)^2 \right\} \\
\Delta \varphi_a / C_D^0 &= \left\{ K_1 C_A^0 / \lambda (1 + K_1 C_A^0) \right\} - \left\{ K_1 \Delta \varphi_a / (1 + K_1 C_A^0)^2 \right\} \\
\Delta \varphi_d / C_D^0 &= \left\{ K_1 C_A^0 / \lambda (1 + K_1 C_A^0) \right\} - \left\{ K_1 \Delta \varphi_d / (1 + K_1 C_A^0)^2 \right\} \\
\Delta \Omega_{CD_A} / C_D^0 &= \left\{ K_1 C_A^0 / \lambda (1 + K_1 C_A^0) \right\} - \left\{ K_1 \Delta \Omega_{CD_A} / (1 + K_1 C_A^0)^2 \right\}
\end{align*}

where the notations have their usual meanings and their method of calculations are the same as reported in Chapter II. As expected from Eqs. (3.1 - 3.4), the plots of $\delta \varphi$ versus $\delta \varphi / C_D^0$, $\Delta \varphi_a$ versus $\Delta \varphi_a / C_D^0$, $\Delta \varphi_d$ versus $\Delta \varphi_d / C_D^0$ and $\Delta \Omega_{CD_A}$ versus $\Delta \Omega_{CD_A} / C_D^0$ were linear with a slope $= -K_1 / (1 + K_1 C_A^0)^2$, and intercept $= K_1 C_A^0 / \lambda (1 + K_1 C_A^0)$ (Figs. 3.1 - 3.5). Refractometric and differential refractometric titration techniques have indicated 1:1 stoichiometry of these complexes (Figs. 3.6-3.9).

The $K_1$ has also been calculated using Yoshida and Osawa's\textsuperscript{21} Equation (3.5):
FIG. 3.1 Plots of $\delta \phi$ versus $\delta \phi/c_D$ for Molecular Complexes of Aromatic Aminoacids with Chloranil in 50% Aqueous Ethanol.
FIG. 3.2 Plots of $\Delta \phi_a$ versus $\Delta \phi_a / C_D^0$ for Molecular Complexes of Aminoacids with Chloranil in 50% Aqueous Ethanol.
FIG. 3.3 Plots of $\Delta \Omega c_{DA}$ versus $\Delta \Omega c_{DA} / c_D^0$ for Tryptophan-Chloranil (○○○), Tyrosin-Chloranil (○○○) and Phenylalanine-Chloranil (□□□) Complexes in 50% Aqueous Ethanol.
FIG. 3.4 Plots of $\delta \phi$ versus $\delta \phi/c^0$ for Molecular Complexes of Some Aliphatic Aminoacids with Chloranil in 50% Aqueous Ethanol.
FIG. 3.5 Plots of $\Delta \Omega_{\text{CDA}}$ versus $\Delta \Omega_{\text{CDA}}/C_D^0$ for Molecular Complexes of Some Aliphatic Aminoacids with Chloranil in 50% Aqueous Ethanol.
FIG. 3.6 Molar Ratio Plots for the Molecular Complexes of Some Aromatic Aminoacid with Chloranil in 50% Aqueous Ethanol
FIG. 3.7 Molar Ratio Plots for Molecular Complexes of Some Aromatic Aminoacid with Chloranil in 50% Aqueous Ethanol.
FIG. 3.8 Molar Ratio Plots for the Tryptophan-Chloranil (●●●), Tyrosine-Chloranil (○○○) and Phenylalanine-Chloranil (ΔΔΔ) Complexes in 50% Aqueous Ethanol.
FIG. 3.9 Molar Ratio Plots of Molecular Complexes of Some Aliphatic Aminoacids with Chloranil in 50% Aqueous Ethanol.
\[ K_1 = 2 \sqrt{k} \left\{ \int k(C + C') - (C + kC') \right\}^2 / (C - kC')^2 \] .. (3.5)

where \( C \) and \( C' \) are the maximum concentrations of both the systems and \( k \) is the maximum deviation from the additive line when molar ratio of solutes is plotted against square of refractive indices \( (n^2) \). \( K_1 \) has also been calculated by modified Yoshida and Osawa's method as suggested by Sahai and Singh\(^{22}\) in which instead a plot of \( n^2 \) versus molar ratio of solutes, \( \Delta \Omega_{CDA} \), the refraction per cm\(^3\) due to charge-transfer complex is plotted against molar ratio of solutes. \( \Delta \varphi_a \) and \( \Delta \varphi_d \) have also been used to calculate \( K_1 \) by Yoshida and Osawa's method. Few representative plots of molar ratio of solutes versus \( \Delta \Omega_{CDA} \) or \( \Delta \varphi_a \) or \( \Delta \varphi_d \) are shown in Figs. (3.10 and 3.11).

The equilibrium constant \( (K) \) and solvation number \( (n) \) of the two distinct species of intimate and solvent-separated ion-pairs coexisting in equilibrium, have been calculated\(^{23,24}\) by using Eqs. (3.6) & (3.7):

\[ \log \varphi_R = n \log[S] + \log K \] .. (3.6)

\[ \log \Delta \varphi_R = n \log[S] + \log K \] .. (3.7)

where \( \varphi_R \) and \( \Delta \varphi_R \) have their usual meanings and they have been calculated as reported in Chapter II (cf. Sec. 2.2.5, p. 65 and Sec. 2.2.6, p. 70).
FIG. 3.10 A Plot of Refraction per cm$^3$ due to Charge-Transfer $\Delta \Omega \ S_{DA}$ versus Molar Ratio of Solutes Indicating 1:1 Stoichiometry of Tryptophan-Chloranil Complex in 50% Aqueous Ethanol.
FIG. 3.11 A Plot of Refraction per cm$^3$ due to Charge-Transfer $\Delta \Omega C_{DA}$ versus Molar Ratio of Solutes Indicating 1:1 Stoichiometry of Alanine-Chloranil Complex in 50% Aqueous Ethanol.
The stoichiometry of these complexes have been determined by plotting \( \Delta \theta_c A \) (or \( \Delta / \Delta \theta_{DA} C_A \)) against \( X_D \) (mole fraction of donor). The maxima occur at \( X_D = n/(1 + n) \) (Fig. 3.12). After determining \( n \), the \( K_n \) has been calculated by using Eq. (3.8), recently developed by Sahai and Singh, \(^{25}\)

\[
K_n = (\Delta \theta_D / \Delta \theta_{DA}) / (C_D^o)^n (1 - \Delta \theta_D / \Delta \theta_{DA})^{n+1}
\]  

(3.8)

where \( C_D^o = X_D C_T = n/(1 + n C_T) \). After determining the \( K_1 \) from different equations and different types of Job plots, the percentage of contribution of donor (\( S_{Ad} \)) and acceptor (\( S_{Aa} \)) in total 30% solute aggregation have been calculated using Eqs. (3.9 & 3.10), respectively.

\[
S_{Ad} = 30 \Delta K_a / \Delta K_{da}
\]

(3.9)

\[
S_{Aa} = 30 \Delta K_d / \Delta K_{da}
\]

(1.10)

where the values of \( \Delta K_{da} \), \( \Delta K_a \) and \( \Delta K_d \) have been calculated by the equations reported in Chapter II (Sec. 2.2.3, p. 62).

3.4 RESULTS AND DISCUSSION

3.4.1 Interaction of Aminoacids with Chloranil

On mixing the solution of aminoacids (aliphatic and aromatic) with a solution of chloranil in the 50% aqueous
FIG. 3.12 Plots of $X_D$ versus $\phi / \phi_D C_A$ for Molecular Complexes of Some Aliphatic Aminoacids with Chloranil in 50% Aqueous Ethanol.
ethanol, an appreciable increase in refractive index was observed with the increase in donor concentration and keeping the acceptor concentration constant. At this stage, there may be doubt that this relative increase in refractive index may be due to the increase in donor concentration and not due to the charge transfer interaction. This problem has been solved by taking the refractive index of donor and acceptor solution separately. Again, on mixing the equal amount of donor and acceptor solution, the refractive index increases appreciably more than that of separate component. This appreciable increase is due to the charge-transfer interaction and not due to the donor concentration. Thus, this relative increase in refractive index of solution may be interpreted due to the charge-transfer from lone pair (n) of amino group (-NH₂) of amino acids to π* orbital of chloranil.***

The $K_1$ and extent of electronic polarization ($\rho$) calculated for the interaction of amino acids with chloranil from Eqs. (3.1 - 3.4) are listed in Table 3.1. From this table, it is evident that $K_1$ calculated for aliphatic aminoacid-chloranil complexes from Eqs. (3.1 - 3.4) are almost the same. For these

***Spectrophotometric results have indicated that the donation is taking place from the lone pair of amino group and not from the π-electron pool of benzene nucleus of aromatic amino-acids. But above technique can only provide information regarding the charge-transfer, not the probable site of donation. On the basis of equilibrium constant data, one may simply propose the probable site of interaction.
Table 3.1. Equilibrium Constant ($K_1$), Extent of Electronic Polarization ($\alpha$) and Solute Aggregation Data for 1:1 Molecular Complexes of Some Amino Acids with Chloranil in 50% aqueous Ethanol at 30°C.

<table>
<thead>
<tr>
<th>Aminoacid</th>
<th>Equilibrium Constant ($K_1$) 1 mol(^{-1}) Spectrophotometric Method</th>
<th>Extent of Electronic Polarization $\alpha \times 10^5$</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>$K_1$  T°C</td>
<td>From Eq. 3.1</td>
</tr>
<tr>
<td>Phenylalanine</td>
<td>151±4  167±5  160±5  131±2</td>
<td>3.81  4.09  3.84  2.97</td>
</tr>
<tr>
<td>Tyrosin</td>
<td>167±5  173±5  170±5  142±2</td>
<td>3.93  3.98  3.68  3.52</td>
</tr>
<tr>
<td>Tryptophan</td>
<td>176(15)  26  222±5  208±5  200±5  156±3</td>
<td>5.04  4.63  4.52  3.58</td>
</tr>
<tr>
<td>Valine</td>
<td>275±10  265±10  255±10  240±5</td>
<td>4.65  6.29  6.12  5.71</td>
</tr>
<tr>
<td>Leucine</td>
<td>224(15)  20  300±10  272±10  265±10  258±10</td>
<td>5.20  6.57  6.40  6.28</td>
</tr>
<tr>
<td>Glycine</td>
<td>298(15)  20  320±10  300±10  280±10  275±10</td>
<td>5.79  7.28  7.22  6.77</td>
</tr>
<tr>
<td>Alanine</td>
<td>318(15)  20  400±15  375±15  360±15  310±15</td>
<td>7.46  9.26  9.20  7.91</td>
</tr>
</tbody>
</table>

Number in parentheses indicates the reference number.
systems, the concentration of solutes was kept low \((10^{-3} \text{ M})\). Therefore, the possibility of solute aggregation becomes low. The difference noted in \(K_1\)'s values obtained from these equations may be due to the betterment of respective equations. \(K_1\) calculated from Eq. (3.1), in which refraction per \(\text{cm}^3\) due to solvent was considered, show maximum percentage of error. But for the same system if Eq. (3.2) in which the refraction per \(\text{cm}^3\) due to solution (donor + acceptor) and acceptor was considered, the percentage of error is less. The value of \(K_1\) calculated from Eq. (3.3) was noted to be the same as was calculated from Eq. (3.2). This indicates that the contribution of donor or acceptor in solute aggregation is almost negligible. The best values have been obtained when Eq. (3.4) has been used in which differential refractometric method has been applied to evaluate \(\Delta \mathcal{R}_{CD,A}\), the refraction per \(\text{cm}^3\) due to charge-transfer.

The \(K_1\) calculated from Yoshida and Osawa's method\(^{21}\) and by applying \(\Delta \mathcal{R}_a\) or \(\Delta \mathcal{R}_d\) or \(\Delta \mathcal{R}_{CD,A}\) versus molar ratio of solutes and from the plot of \(n^2\) versus molar ratio of solutes listed in Table 3.2, show slight difference. This may again be interpreted due to the betterment of the differential refractometric method. We could not get the different values for the maximum deviation from the base line \((k)\), when plotted \(\Delta \mathcal{R}_a\) versus molar ratio of solutes. Thus, it was impossible to calculate the different \(K_1\) values through this plot. This is in parallel agreement with our earlier
Table 3.2. Equilibrium Constant ($K_1$) and Extent of Polarization ($\alpha$) Data for Molecular Complexes of Aminoacids with Chloranil Obtained from Different Methods in 50% Aqueous Ethanol at 30°C.

<table>
<thead>
<tr>
<th>Aminoacid</th>
<th>Equilibrium Constant ($K_1$) l.mol⁻¹</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1</td>
</tr>
<tr>
<td>Phenylalanine</td>
<td>157±5</td>
</tr>
<tr>
<td>Tyrozin</td>
<td>191±8</td>
</tr>
<tr>
<td>Tryptophan</td>
<td>208±10</td>
</tr>
<tr>
<td>Valine</td>
<td>268±10</td>
</tr>
<tr>
<td>Leucine</td>
<td>295±10</td>
</tr>
<tr>
<td>Glycine</td>
<td>295±10</td>
</tr>
<tr>
<td>Alanine</td>
<td>310±10</td>
</tr>
</tbody>
</table>

1. From the plot of $n^2$ versus molar ratio of solutes
2. From the plot of $\Delta \alpha$ versus molar ratio of solutes
3. From the plot of $\Delta \alpha$ versus molar ratio of solutes
4. From the plot of $\Delta \alpha$ versus molar ratio of solutes
5. From Eq. 3.8.
observations\textsuperscript{19} (Ch.II, Sec. 2.2.3, p. 57). From Tables 3.1 & 3.2, it is also evident that the contribution of donor is less than that of acceptor in total 30\% solute aggregation. This is also in parallel agreement with our earlier observation.\textsuperscript{19}

The refractive index of donor as well as that of chloranil in 50\% aqueous ethanol increases with concentration. The plot of $\Delta n$, the difference in refractive index of calculated and observed values, against $C$, concentration of the complex, is linear indicating that there is no interaction between the complex and individual species. Since the complex is generally more polar than the components, the extent of electronic polarization or refractive index increases and the deviation depends upon the extent of interaction between donor and acceptor.\textsuperscript{27-30}

Therefore, stronger is the complex, larger is the deviation in the $n$ values. In present cases, the deviation is neither too large nor too small which indicates that the interaction is of moderate type. This is in consistent with the $K_1$ values of these systems. From Tables (3.1) and (3.2), it is evident that the interaction is stronger in alanine than other aliphatic aminoacids. The better donor ability of alanine may be due to the inductive effect of $-CH_3$ groups which may make the $-N:$ atom more basic than that of others. The $K_1$ obtained from Eq. (3.1) and Yoshida and Osawa's plot show greater difference.* This

*In Eq. (3.1), $\Delta \phi$ the difference in the refraction per cm$^3$ of solution and solvent and in Yoshida and Osawa's method, square of refractive index ($n^2$) has been plotted against molar ratio of solutes. Therefore, the greater difference in $K_1$ values calculated from both the methods, may be interpreted due to solute-solvent interaction.
clearly indicates that the solvent in these systems can act as electron donor. This is in agreement with the observations of spectrophotometric investigations, in which water and ethanol have been reported to be the electron donor. From Table (3.1), it is also clear that the interaction is stronger in case of tryptophan than tyrosin and phenylalanine. Also tyrosin shows strong interaction than phenylalanine. From theoretical calculation of Pullman, it is also evident that tryptophan having an indole nucleus should behave as a better electron donor.

The extent of electronic polarization \( (\alpha^2) \) obtained from refractometric methods were found linear with \( K_1 \). From Figs. (3.10) & (3.11), it is evident that as soon as the complex formation takes place, the electronic polarizability increases than that of additive values. But it has been reported that the dipole moment of the complex always depends upon the total polarizability of the complex. In calculations of dipole moment of the complex so far, the electronic polarizability is neglected on the grounds that the electronic polarizability of the complex is equal to the sum of the electronic polarizability of individual molecules. But from this study, it is concluded that, in order to get the exact dipole moment of the complex, the electronic polarizability could not be neglected.

3.4.2 Amino acids - Iodine Systems

When excess of amino acids are added to the aqueous solution of iodine, bleaching occurs which takes several minutes
for tryptophan and several hours for other aminoacids. The similar observation was noticed previously by Slifkin. During the bleaching process, the tryptophan-iodine system shows the increase in refractive index and for the same system when bleaching process is over, the decrease in refractive index was noticed. For other aminoacids, during bleaching and after bleaching process is over, the decrease in refractive index was observed. From this observation, it is clear that in case of tryptophan-iodine complex, outer complex (stage I) is more dominating than other aminoacid-iodine complexes. Since in other aminoacids, a direct decrease in refractive index was observed, therefore in these cases, the lifetime of stage I is very low. As soon as the outer complex is formed, the system becomes tight indicating the increase in refractive index. But after bleaching the transformation of outer complex into inner complex occurs (stages II and III) and the system becomes less tight showing the decrease in refractive index. In case of other aminoacids, the outer complex stage was not detected, because directly the decrease in refractive index was noted which is due to the immediate transformation of outer to inner complex. From these observations, a mechanism of transformation of these complexes shown in Scheme I, may be proposed:
This mechanism of interaction gives further support to Slifkin's proposal given earlier for the interaction of aminoacids with iodine. Further, a binary mixture of dioxane and tetrahydrofuran (THF) have been used to monitor the solvent-separation of an intimate ion-pair. A plot of $n^2$ and $\Delta \rho_{CS}$, the refraction per cm$^3$ due to solvent-separated ion-pair species, versus mole fraction of THF (Fig. 3.13) is a sigmoid type curve with two plateaus, one around 0.2 and other around 0.6 mole fraction of THF. This may be due to the coexistence of two distinct species in equilibrium:

$$AAI^+, I^-_3 + n\text{THF} \rightleftharpoons K \rightleftharpoons AAI^+/I^-_3$$

intimate ion-pair  solvent-separated ion-pair
FIG. 3.13 Square of Refractive Index ($n^2$) due to Intimate and Solvent-Separated Ion-pair Formation of Some Aromatic Aminoacid-I$_2$ Complexes as a Function of THF in the Mixture Dioxane-THF.
Similar results for \( \text{Ph}_3^M\text{-I}_2 \) (where \( M = N, P, \text{As} \& \text{Sb} \)) complexes have also been obtained by Sahai et al. using NMR, \(^{35}\) refractometric and conductometric methods. \(^{18}\) A representative plot of \( \log \Delta \varphi_R \text{ versus } \log[\text{THF}] \) are shown in Figure (3.14). The \( K \) and \( n \) for these systems have been calculated using Eqs. (3.6) and (3.7) and these data are recorded in Table 3.3. From this table, it is evident that a single THF molecule has managed to intercalate itself between anion and cation.

### 3.4.3 Interaction of Indoles with Iodine

Foster and Hanson\(^{36}\) have shown that a mixed solution of indoles and iodine in dichloromethane have an absorption maximum at 367 nm and as the triiodide ion band also absorbs in this region, therefore, no definite conclusion about the charge-transfer band was made. They have also reported that some irreversible changes occur, which may be due to the chemical reaction between indoles and iodine. Since the indoles are the basic components of several biomolecules\(^{37}\) and the radicals of these biomolecules have noted to play a dominant role in cancer induction,\(^ {38}\) therefore, the above noted observations have initiated us to study the interaction of indoles with iodine using refractometric and differential refractometric techniques.

The \( K_1 \) calculated from Eqs. (3.1 - 3.4) are listed in Table (3.4). From this table, it is clear that \( K_1 \) calculated from these equations are almost the same (except indole-iodine
FIG. 3.14 Refractometric Solvent-Separated Ion-pair Formation of Some $\text{AAI}^+, \text{I}_3^+$ (AA = Aminoacids) as a Function of THF in a Mixture of Dioxane-THF
Table 3.3. Solvation number (n) and Equilibrium Constant (K) Data for the Intimate and Solvent-Separated Ion-pairs in the System:

\[ \text{AAI}^+ + I^- + n\text{THF} \xrightleftharpoons{K} \text{AAI}_2^+/2I^- \]

\( \text{AA} = \text{Aminoacid} \)

<table>
<thead>
<tr>
<th>AA</th>
<th>Solvation number (n)</th>
<th>Equilibrium constant (K)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Refractometric Method</td>
<td>Differential Refractometric Method</td>
</tr>
<tr>
<td>Phenylalanine</td>
<td>1.10</td>
<td>1.05</td>
</tr>
<tr>
<td>Tyrosin</td>
<td>1.20</td>
<td>1.05</td>
</tr>
<tr>
<td>Tryptophan</td>
<td>0.85</td>
<td>1.00</td>
</tr>
<tr>
<td>Valine</td>
<td>0.80</td>
<td>0.96</td>
</tr>
<tr>
<td>Leucine</td>
<td>0.92</td>
<td>0.98</td>
</tr>
<tr>
<td>Glycine</td>
<td>1.15</td>
<td>1.05</td>
</tr>
<tr>
<td>Alanine</td>
<td>1.15</td>
<td>1.05</td>
</tr>
</tbody>
</table>
**Table 3.4.** Equilibrium Constant ($K_1$), Extent of Polarization ($\phi$) & Solute Aggregation ($SA_a$, $SA_d$) Data for 1:1 Molecular Complexes of Indoles with Iodine in Carbon Tetrachloride at 30°C.

<table>
<thead>
<tr>
<th>Complex</th>
<th>Equilibrium Constant ($K_1$) 1 mol$^{-1}$</th>
<th>Extent of electronic polarization $\times 10^5$</th>
<th>$SA_a$</th>
<th>$SA_d$</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>From Eq.3.1 From Eq.3.2 From Eq.3.3 From Eq.3.4</td>
<td>From Eq.3.1 From Eq.3.2 From Eq.3.3 From Eq.3.4</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Indole-iodine</td>
<td>110  88  72  53</td>
<td>4.10  3.4  3.1  2.6</td>
<td>15.67  14.32</td>
<td></td>
</tr>
<tr>
<td>2-Me-indole-iodine</td>
<td>200  189  181  170</td>
<td>5.34  5.19  5.10  4.45</td>
<td></td>
<td></td>
</tr>
<tr>
<td>3-Me-indole-iodine</td>
<td>250  224  215  200</td>
<td>6.20  5.85  5.60  5.29</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
complex *). Their percentage of deviation from the differential refractometric values are under the experimental error (10-15%).† Since the order of $K_1$ values for these complexes are same as was found for aminoacid-chloranil systems, therefore, their discussion will also be of same type. The $K_1$ values obtained from Yoshida and Osawa's method and Sahai et al. plots are recorded in Table 3.5. In this case, also the order of $K_1$ values is same as has been obtained by Eqs. (3.1 - 3.4). Consequently, the basis of its discussion will also be the same as given earlier (cf. Chapter II, Sec. 2.2.3). From Table 3.5, it is evident that $Δ\phi_{DA}, Δ\phi_a$ and $Δ\phi_d$ maxima are less in CHCl$_3$ than in CCl$_4$. This lowering in maxima was found to be the maximum in the case of 3-me-indole-iodine system, though its equilibrium constant is higher than 2-me-indole-iodine and indole-iodine complexes. According to previous observations, the deviation from the base line ($k$) always depends upon the extent of interaction. If the complex is strong, the deviation in $Δ\phi_a$ or $Δ\phi_d$ or $Δ\phi_{DA}$ values should be large and the reverse was found to be true for weak complexes. But for indoles-

* In order to get appropriate variation in scale due to limited accuracy of the instrument and low $K_1$ values, the solute concentration was raised ($10^{-2}$ M). At such a higher concentration, the solute aggregation may occur which may prevent to give the reliable value of $K_1$. Therefore, $K_1$ calculated from Eq. (3.1) shows maximum deviation with respect to Eq. (3.4).

†Since the spectrophotometric values of $K_1$ are not available and it has been proved that differential refractometric values were more reliable than refractometric values, therefore, a comparison with differential refractometric values has been made in the present method.
Table 3.5. Equilibrium Constant ($K_1$) and Solute Aggregation Data for 1:1 Molecular Complexes Obtained on the Basis of Yoshida and Osawa's Equation in Carbon-Tetrachloride at 30°C

<table>
<thead>
<tr>
<th>Complex</th>
<th>Equilibrium Constant ($K_1$) 1.mol⁻¹</th>
<th>$\Delta \varphi_a$ maxima</th>
<th>$\Delta \varphi_d$ maxima</th>
<th>$\Delta \Omega_{CDA}$ maxima</th>
<th>$S_{Aa}$</th>
<th>$S_{Ad}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Indole-iodine</td>
<td></td>
<td>90</td>
<td>0.69</td>
<td>59</td>
<td>49</td>
<td>0.60</td>
</tr>
<tr>
<td>2-Me-indole-iodine</td>
<td></td>
<td>191</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>165</td>
<td>0.82</td>
<td></td>
<td></td>
</tr>
<tr>
<td>3-Me-indole-iodine</td>
<td></td>
<td>246</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1. Calculated from the plot of $n^2$ versus molar ratio of solutes
2. Calculated from the plot of $\Delta \varphi_a$ versus molar ratio of solutes
3. Calculated from the plot of $\Delta \varphi_d$ versus molar ratio of solutes
4. Calculated from the plot of $\Delta \Omega_{CDA}$ versus molar ratio of solutes.
5. Calculated from Equation (3.8).
iodine systems the deviation from the ideal behaviour (being $K_1$ higher and maxima low) may be interpreted as follows:

In refractometric measurements, the refraction per cm$^3$ of the complex always depends upon the concentration of donor (D) and acceptor (A). Since, the decrease in $\Delta \varphi_a$ or $\Delta \varphi_d$ or $\Delta \Omega_{DA}$ maxima were noted, it means, the concentration of the complex is low in the solution. This may be due to the transformation of outer complex to inner complex (Scheme II). As in solution, the concentration of ionic species (inner complex) becomes higher, the low concentration of outer complex is obvious which indicates the lowering of $\Delta \varphi_a$ or $\Delta \varphi_d$ or $\Delta \Omega_{DA}$ maxima values even in the presence of strong interaction.

The values of $\Delta \varphi_a$, $\Delta \varphi_d$ and $\Delta \Omega_{DA}$ were found to be least in 3-me-indole-iodine complex which shows that the formation of inner complex is more facilitated in this case than the others. From $K_1$ values of this system, it is also evident that 3-me-indole should behave as noted above. If the lifetime of outer complex is less and directly inner complex is formed, a negative deviation may be obtained as was noted for triethylamine-iodine system.$^{19}$

We have attempted to calculate equilibrium constant and stoichiometry of these complexes in CHCl$_3$. By applying Job method, 1:2 stoichiometry was found. Therefore, Eqs. (3.1 - 3.4), Yoshida and Osawa's$^{21}$ Eq. (3.5) and modified methods of Sahai et al.$^{19,22}$ could not be used to calculate equilibrium
constant of these complexes. At this stage, only Eq. (3.8) has been used to evaluate $K_2$ of these systems in CHCl$_3$ (Table 3.6). $\Delta \phi_a$, $\Delta \phi_d$ and $\Delta \Omega_{CDa}$ maxima obtained in CHCl$_3$ were found in the same order as were found in CCl$_4$. Thus, on the basis of this observation a mechanism (similar to aminoacid-iodine systems) of transformation of outer complex to inner complex has been proposed (Scheme II):

**SCHEME II**

$$
\text{IN} + I_2 \rightleftharpoons \text{IN} \cdot I - I \quad \text{(stage I)}
$$

(Outer complex)

$$
\text{IN} \cdot I - I + I_2 \rightleftharpoons \text{IN} \cdot I^+ \cdot I^- \cdot I - I \quad \text{(stage II)}
$$

(Transition state)

$$
\text{IN} \cdot I^+ \cdot I^- \cdot I - I \rightleftharpoons \text{IN} \cdot I^+ , I_3^- \quad \text{(stage III)}
$$

(Inner complex)

$\text{IN} = \text{Indoles}$

Equations (3.1 - 3.4) have also been used to study the ion-pair formation of these systems. Though, from these equations, the refractometric maxima ($\Delta \phi_a$, $\Delta \phi_d$ and $\Delta \Omega_{CDa}$) have not been observed, but a parameter $C$, the extent of electronic polarization, has been calculated. Normally, the $C$ value increases with the increase of $K_1$ of the complex at the same concentration of the reactants. In 2-me-indole-iodine and
In indole-iodine system, the $\alpha$ value also noted to be high. This abnormal increase in $\alpha$ values may be interpreted due to the formation of inner complex. These values are linear with $K_1$ values which indicate that the higher the equilibrium constant of the system it will facilitate more to form inner complex than the system having low equilibrium constant values. This observation is in parallel agreement with the observation made through $\Delta \phi_a$, $\Delta \phi_d$ and $\Delta \Omega_{CDA}$ maxima, obtained from different plots.

A plot of $\Delta \Omega_{C_{ss}}$, the refraction per cm$^3$ due to solvent-separated ion-pair species, versus mole fraction of THF (Fig. 3.15) is a sigmoid type curve with two plateaus, one around 0.2 and the other around 0.6 mole fraction of THF. This may again be interpreted due to the coexistence of two distinct species in equilibrium:

$$\text{IN I}^+ + \text{I}_3^- + n\text{THF} \xleftrightarrow{K} \text{IN I}^+ // \text{I}_3^-$$

intimate ion-pair

solvent-separated ion-pair

Similar results for Ph$_3^M$-I$_2$ and aminoacids-iodine complexes have also been obtained by Sahai et al. The $K$ and $n$ for this equilibrium have been calculated by Eqs. (3.6 & 3.7) and these data are recorded in Table (3.7) which suggest that a single THF molecule has managed to intercalate itself between anion and cation.
Table 3.6. Equilibrium Constant \((K_2)\) Obtained from Sahai and Singh's Method\(^\text{25}\) in Chloroform at 30°C.

<table>
<thead>
<tr>
<th>Complex</th>
<th>Equilibrium Constant ((K_2)) 1.(\text{mol}^{-1})</th>
</tr>
</thead>
<tbody>
<tr>
<td>Indole-iodine</td>
<td>780.00</td>
</tr>
<tr>
<td>2-Me-indole-iodine</td>
<td>1120.20</td>
</tr>
<tr>
<td>3-Me-indole-iodine</td>
<td>1440.40</td>
</tr>
</tbody>
</table>

Table 3.7. Solvation Number and Equilibrium Constant Data of the Intimate and Solvent-Separated Ion-pair in the System:

\[
\text{InI}^+, I_3^- + n\text{THF} \rightleftharpoons K \xrightarrow{\text{K}} \text{InI}^+/I_3^- 
\]

<table>
<thead>
<tr>
<th>In</th>
<th>Solvation Number ((n))</th>
<th>Equilibrium Constant ((K) 1.\text{mol}^{-1})</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Refractometric Method</td>
<td>Differential Refractometric Method</td>
</tr>
<tr>
<td>Indole</td>
<td>0.83 ± 0.05</td>
<td>0.96 ± 0.05</td>
</tr>
<tr>
<td>2-Me-indole</td>
<td>0.88 ± 0.05</td>
<td>1.02 ± 0.05</td>
</tr>
<tr>
<td>3-Me-indole</td>
<td>0.92 ± 0.05</td>
<td>1.10 ± 0.05</td>
</tr>
</tbody>
</table>

In = Indoles.
FIG. 3.15  Square of Refractive Index ($n^2$) (0-0-0), Refraction per cm$^3$ due to Solvent-Separated ($\Delta \Omega C_{ss}$) (000) Ion-pair Formation of 3-Methyl Indole-Iodine Complex as a Function of THF in a Mixture of Dioxane-THF. A Variation of $n^2$ for Binary Mixture of Dioxane-THF alone (-----)
3.4.4 Interaction of DDT with Indoles

It has been reported that DDT can act as a carcinogen. \(^{11,14}\) This initiated us to study the interaction of DDT with some compounds of biological interest (indoles).

The \(K_1\) and extent of electronic polarization (\(\alpha_L\)) calculated from Eqs. (3.1 - 3.4) and Fig. (3.16) for these complexes are recorded in Table 3.8. In order to get appropriate variation in scale due to limited accuracy of the instrument and low \(K_1\) values, the solute concentration was raised \((0.1 - 1.0\ M)\). At such higher concentration, the solute aggregation occurs which prevents to get the reliable value of \(K_1\). Their order of variation in \(K_1\) values are the same as reported in previous sections of this chapter. \(K_1\) has also been calculated by Yoshida and Osawa's\(^{21}\) (Fig. 3.17) and Sahai et al.\(^{19,22}\) methods (Fig. 3.18) and these are reported in Table 3.9. From Tables 3.8 & 3.9, it is clear that contribution in solute aggregation of acceptor is more than that of donor. This is in good agreement with our earlier observation (cf. Ch. II, Sec. 2.2.3, p. 63). In these complexes, a small but positive deviation at 1:1 molar ratio of solutes was noted (Fig. 3.18). This indicates that these complexes are weak which is in parallel agreement with those of earlier studies (cf. Ch. II, Sec. 2.2.3, p. 57).

DDT has three probable sites of interaction. Apart from the electron deficient chlorophenyl rings and trichloromethyl group, the benzhydryl system can also act as the site of
FIG. 3.16 Plots of $\Delta \Omega C_{DA}$/C_D versus $\Delta \Omega C_{DA}$/C_D for Molecular Complexes of DDT with Some Compound in CCl₄.
Table 3.8. Equilibrium Constant \( (K_1) \) Extent of Electronic Polarization \( (\alpha) \) and Solute Aggregation Data for 1:1 Molecular Complexes of Some Donors with DDT in Carbon Tetrachloride at 30°C

<table>
<thead>
<tr>
<th>Donor</th>
<th>( K_1 ) (l. mol(^{-1}))</th>
<th>Refractometric Methods</th>
<th>N.M.R. (ref.)</th>
<th>Extent of Polarization ( \alpha \times 10^2 )</th>
<th>Solute aggregation</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Eq. 3.1</td>
<td>Eq. 3.2</td>
<td>Eq. 3.3</td>
<td>Eq. 3.4</td>
<td>( K_1 )</td>
</tr>
<tr>
<td>Indole</td>
<td>0.98±</td>
<td>0.88±</td>
<td>0.83±</td>
<td>0.62±</td>
<td>0.308±</td>
</tr>
<tr>
<td></td>
<td>0.04±</td>
<td>0.03±</td>
<td>0.03±</td>
<td>0.02±</td>
<td>0.010±</td>
</tr>
<tr>
<td>2-Methyl-indole</td>
<td>1.06±</td>
<td>0.98±</td>
<td>0.98±</td>
<td>0.68±</td>
<td>0.330±</td>
</tr>
<tr>
<td></td>
<td>0.05±</td>
<td>0.03±</td>
<td>0.03±</td>
<td>0.03±</td>
<td>0.013±</td>
</tr>
<tr>
<td>3-Methyl-indole</td>
<td>1.12±</td>
<td>0.04±</td>
<td>0.96±</td>
<td>0.75±</td>
<td>3.80±</td>
</tr>
<tr>
<td></td>
<td>0.08±</td>
<td>0.08±</td>
<td>0.04±</td>
<td>0.04±</td>
<td>0.024±</td>
</tr>
<tr>
<td>Naphthalene</td>
<td>0.53±</td>
<td>0.48±</td>
<td>0.46±</td>
<td>0.32±</td>
<td>1.29±</td>
</tr>
<tr>
<td></td>
<td>0.02±</td>
<td>0.02±</td>
<td>0.02±</td>
<td>0.02±</td>
<td>0.05±</td>
</tr>
<tr>
<td>Benzene</td>
<td>0.63±</td>
<td>0.59±</td>
<td>0.56±</td>
<td>0.48±</td>
<td>1.68±</td>
</tr>
</tbody>
</table>
FIG. 3.17 A plot of Refraction per cm$^3$ due to Charge-Transfer $\Delta \Omega C_{DA}$ versus Molar Ratio of Solutes Indicating 1:1 Stoichiometry of Indole-Chloranil Complex in CCl$_4$
FIG. 3.18 A Plot of $X_D$ versus $\frac{\phi_D}{\Delta \phi_{DA}^o}$ Indicating 1:1 Stoichiometry of 3-Methyl-Indole-DDT Complex in $CCl_4$. 
Table 3.9. Equilibrium Constant ($K_1$) and Solute Aggregation Data for 1:1 Molecular Complexes of Some Donors with DDT, Obtained on the Basis of Yoshida and Osawa's Equation in Carbon Tetrachloride at 30°C.

<table>
<thead>
<tr>
<th>Donor</th>
<th>$K_1$ (1. mol⁻¹)</th>
<th>Refractometric Methods</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>SAₐ</th>
<th>SAₐ</th>
</tr>
</thead>
<tbody>
<tr>
<td>Indole</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>8.82</td>
<td>21.18</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>1.02±</td>
<td>0.92±</td>
<td>0.88±</td>
<td>0.68±</td>
<td>0.71±</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>0.05</td>
<td>0.05</td>
<td>0.04</td>
<td>0.02</td>
<td>0.03</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2-Methyl indole</td>
<td></td>
<td></td>
<td>1.10±</td>
<td>0.96±</td>
<td>0.83±</td>
<td>0.71±</td>
<td>0.82±</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>0.05</td>
<td>0.04</td>
<td>0.06</td>
<td>0.03</td>
<td>0.04</td>
<td></td>
<td></td>
</tr>
<tr>
<td>3-Methyl indole</td>
<td></td>
<td></td>
<td>1.18±</td>
<td>1.02±</td>
<td>0.94±</td>
<td>0.82±</td>
<td>0.86±</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>0.08</td>
<td>0.08</td>
<td>0.06</td>
<td>0.04</td>
<td>0.04</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Naphthalene</td>
<td></td>
<td></td>
<td>0.66±</td>
<td>0.58±</td>
<td>0.51±</td>
<td>0.42±</td>
<td>0.45±</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>0.03</td>
<td>0.02</td>
<td>0.02</td>
<td>0.02</td>
<td>0.02</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Benzene</td>
<td></td>
<td></td>
<td>0.60±</td>
<td>0.55±</td>
<td>0.50±</td>
<td>0.46±</td>
<td>0.42±</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>0.02</td>
<td>0.04</td>
<td>0.02</td>
<td>0.02</td>
<td>0.02</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1. Calculated from the plot of $n^2$ versus molar ratio of solutes
2. Calculated from the plot of $\Delta \phi_a$ versus molar ratio of solutes
3. Calculated from the plot of $\Delta \phi_d$ versus molar ratio of solutes
4. Calculated from the plot of $\triangle \phi_{CD}$ versus molar ratio of solutes
5. Calculated from Eq. (3.8).
interaction through hydrogen bonding. Electron withdrawal by the trichloromethyl group results in increase in polarity of the C-H bond involving the benzhydryl protons. The partial positive charge on the proton might make it possible to participate in hydrogen bond type association with the compounds having lone pair (n) electrons on an oxygen or nitrogen. In the present case, this type of possibility may be but it has been noted that hydrogen bonding between two components lead to a greater change in refractive index, \( \Delta \phi_a \), or \( \Delta \phi_d \) or \( \Delta \phi_{\text{DA}} \). We are unable to get a greater change in these values but a small change was noted. Consequently, it can be interpreted that the donation is taking place from the \( \pi \)-electron pool of indoles and not from the lone pair of nitrogen atom. This is in good agreement with those of Sahai obtained through NMR measurements. We are unable to locate the actual site of \( \pi \)-electron donation of indoles through these techniques. MO calculations on indoles and substituted indoles using the frontier-electron density principle support the suggestion of Szent-Gyorgyi et al.\textsuperscript{40} regarding somewhat localized \( \pi \)-charge transfer interaction involving C-2—C-3 atoms of indoles. The low \( K_1 \) values in the present systems are in good agreement with this hypothesis. An increase in the \( K_1 \) values has been observed in 2 or 3-methyl-indole. Highest value of \( K_1 \) obtained in the case of 3-methyl-indole is indicative of the most effective increase in electron density, and hence a formal negative charge by methylation at 3-position. This methylation makes the
3-position more basic which increases the donor capability of the 3-methyl-indole than others.

It has been reported that with the non-polar aromatics, \( \pi \)-electron pool of DDT acts as acceptor instead a benzhydryl trichloromethyl group. Through refractometric measurements, it is difficult to specify the site of interaction in the cases of benzene-DDT and naphthalene-DDT systems. As the \( K_1 \) calculated from this method are in good agreement with that of spectroscopic method\(^{11} \) through which it has already been proved that \( \pi \)-electron pool of DDT acts as an electron acceptor. Thus on the basis of these \( K_1 \) data, it may be visualized that \( \pi - \sigma^* \) type of interaction occurs in case of benzene-DDT and naphthalene-DDT systems.

3.4.5 Interaction of Genetic Bases with Iodine

The equilibrium constant for the charge-transfer interaction of genetic bases with iodine could not be calculated due to the unavailability of concentration of iodine used for study. When an aqueous solution of iodine is mixed with the aqueous solution of genetic bases, a decrease in refractive index was noted. While studying the charge-transfer interaction of other cases (aminoacids with chloranil), an increase in refractive index was observed. This decrease in refractive index of these complexes may be interpreted due to the formation of ion-pair. The immediate transformation of outer complex to inner complex may be due to the high dielectric constant of
the media and a mechanism for this transformation may be proposed (Scheme III):

**SCHEME III**

\[ \text{GB} + I_2 \rightleftharpoons \text{GB.I-I} \quad \text{(stage I)} \]

\[ \text{GB.I-I} + I_2 \rightleftharpoons \text{GB.I}^+ + I_3^- \quad \text{(stage II)} \]

GB = Genetic bases.

In the above Scheme III, the stage I was not detected because the direct decrease in refractive index was observed. This mechanism of interaction finds further support from the spectrophotometric method\(^{37}\) proposed by Slifkin. Further a binary mixture of dioxane and tetrahydrofuran have been used to monitor the solvent separation of an intimate ion-pair. A plot of \(\Delta \Omega_{CS}^{SS}\) versus mole fraction of THF is shown in Fig. (3.19). From this figure, it is evident that as soon as the solvation of THF molecule occurs, the appreciable increase in refraction per cm\(^3\) was observed. A sigmoid type of curves with two plateaus, one around 0.4 and the other around 0.9 mole fraction of THF was observed which may be explained due to the coexistence of two distinct species in equilibrium:

\[ \text{GB.I}^+ + I_3^- + n\text{THF} \rightleftharpoons K \rightleftharpoons \text{GB.I}^+ \| I_3^- \]

intimate ion-pair

solvent-separated ion-pair
FIG. 3.19 Refraction per cm$^3$ due to Solvent-Separated Ion-pair, $\Delta \Omega_{CS}$ of Molecular Complexes of Genetic Bases with Iodine as a Function of THF in Mixture of dioxane-THF
The $K$ and $n$ for the above mentioned ion-pair equilibria, calculated from Eqs. (3.6) and (3.7) are listed in Table 3.10. It is also clear from this table that a single THF molecule has managed itself to intercalate between anion and cation.

The present investigation is in support of the view of the recently developed, "Electronic Theory of Cancer", proposed by Szent-Gyorgyi and also the theme of his paper, "Proteins, Regulation and Cancer" in which he has proposed that the life is based on the two miracles. Miracle one was the creation and folding of protein molecule. Miracle two was the transformation of this molecule into a highly reactive radical. The cancer cell seems to be unable to perform miracle two. On the basis of our present investigation, it seems that miracle two is not performed by the cell due to the charge-transfer complexation of protein with the carcinogen.

An increasing number of evidence has been accumulated supporting the view that chemical carcinogenesis results from the reaction of the carcinogen or a reactive metabolite with cellular macromolecules. Although no definite evidence has been obtained as to whether the polycyclic aromatic hydrocarbon carcinogen manifests its activity as itself or through an reactive metabolite of the polycyclic aromatic hydrocarbon in carcinogenesis. Thus, Morreal and Ts'0 et al. have shown that polycyclic aromatic hydrocarbon carcinogens bind covalently to DNA or RNA bases in vitro, only when such activating agents as hydrogen peroxide or iodine were added. But
Table 3.10. Solvation Number (n) and Equilibrium Constant (K) Data for the Intimate and Solvent-Separated Ion-pairs in the System:

\[ \text{GB}^+, \text{I}_3^- + n\text{THF} \xrightleftharpoons[K]{\text{nTHF}} A\text{A}^+/\text{I}_3^- \]

GB = Genetic Bases

<table>
<thead>
<tr>
<th>GB</th>
<th>Solvation number (n)</th>
<th>Equilibrium constant (K) 1. mol(^{-1})</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Refractometric Method</td>
<td>Refractometric Method</td>
</tr>
<tr>
<td>Adenine</td>
<td>0.90 ± 0.005</td>
<td>1.108 ± 0.05</td>
</tr>
<tr>
<td>Guanine</td>
<td>0.95 ± 0.005</td>
<td>0.900 ± 0.05</td>
</tr>
<tr>
<td>Cytosine</td>
<td>1.10 ± 0.010</td>
<td>1.080 ± 0.05</td>
</tr>
<tr>
<td>Thymine</td>
<td>1.15 ± 0.010</td>
<td>1.120 ± 0.05</td>
</tr>
<tr>
<td>Uracil</td>
<td>1.20 ± 0.005</td>
<td>1.140 ± 0.10</td>
</tr>
</tbody>
</table>
recently Nagata et al.\textsuperscript{52} have demonstrated that the formation of cation radical of 3,4-benzopyrene may not be accelerated by iodine. Obviously, the carcinogenicity of polycyclic aromatic hydrocarbon through cation radical hypothesis may be disproved. From our present observation of the charge-transfer interaction of biomolecules with iodine and consequently formation of ion-pairs of these systems, it looks, as soon as iodine is added with the carcinogen, instead forming the cation radical of polycyclic aromatic hydrocarbon, iodine starts interacting with biomolecules. This interaction may lead some disorder in genetic bases as well as in amino acids which subsequently may be helpful in producing mutagenesis instead of carcinogenesis. Since, mutagenesis is a prereaction of carcinogenesis, therefore, in long term this mutagenesis transforms into carcinogenesis. This is an support of the observation in which it has been noted that 3,4-benzopyrene may produce greater tumors in presence of iodine than 3,4-benzopyrene alone. On molecular level, thus it may be said that the molecular interaction and subsequent transformation into ion-pairs may produce the mutagenesis but in long term in carcinogenesis.
REFERENCES


36. R. Foster and P. Hanson, Tetrahedron, 21, 255 (1965).


