CHAPTER 4

POLARON HOPPING IN THREE PROTEINS AND THEIR

CHARGE TRANSFER COMPLEXES
4.1 Introduction

Organic semiconductors have been well-known form a long time [1-3]. The subject has got break through after discovery of metallic conductors [4-6]. However, studies on charge transfer interaction in biomolecules have been very limited [7-17]. Mainly homo-molecular biomolecules are studied [18]. The modes for superconductivity in biomolecules have been proposed [19-21]. Electrical conduction in proteins shows that proteins are semiconductors [18]. In the present work we study infrared spectra of three proteins namely actine, thrombin and γ-globulin and their CTCs with organic acceptors. Recently we studied polaron hopping in some biomolecular solids and their CTCs [22] as well as CTCs of four amino acids [23]. This work is an extension of our work on biomolecular CTCs.

4.2 Experimental details

Three proteins namely actine, thrombin and γ-globulin were obtained from Sigma-Aldrich chemical company and were highly purified. The organic acceptors namely TCNQ (7,7,8,8-tetracyano-p-quinodimethane), TCNE(tetracyano-p-ethylene), chloranil (2,3,5,6-tetrachloro-p-benzoquinone), DDQ (2,3-dichloro-5,6-dicyano-p-
benzoquinone) and KI-I$_2$ were added in equal volume proportions in these three proteins. The mixtures were grinded in an agate mortar with pastle and fine homogeneous powders were prepared. These powders in 5% were again grinded with 95% anhydrous KBr powder which was spectrograde. The mixture was compressed in a manually operated compressing machine in circular die to prepare semitransparent circular pellets. These discs were placed in a dark chamber of spectrophotometer.

The spectra in the range 400-4000cm$^{-1}$ were recorded using GXFTIR single beam spectrophotometer manufactured by Perkin-Emler co. USA, having a resolution of 0.15cm$^{-1}$, a scan range of 15,600-30cm$^{-1}$, a scan time of 20scan/s, and OPD velocity of 0.20cm/s and MIRTGS and FIRTGS detectors. A beam splitter of opt-KBr type was used having a range of 7800-370cm$^{-1}$. The spectra were recorded in purge mode.

4.3 Results and discussion

Recently we studied CT complexes of two proteins namely $\alpha$-keratin and elastin [24, 25]. $\alpha$-keratin is a fibrous protein in which molecules are arranged in a parallel fashion and behaves as a one-dimensional system. Beta density was found revealing hopping mechanisms of atomic vibrations. Fractal dimensionality was found to be
one for this protein. Elastin had two-dimensional structure which was based on planar desmosine molecule as a unit. Gaussian distributions were found common among CTCs of elastin rather than beta density. This revealed two-dimensional delocalization of polarons. Fractal dimensionality was concluded to be two for this protein.

In the present work, three three-dimensional (globular) proteins and their CTCs have been studied. Actin has a rope-like structure similar to fibrous α-keratin and therefore works as a one dimensional system [26]. Monomer molecules are arranged in parallel fashion in acting fibre studies have been carried out on actin structure and function which include immuneflourescence and electron microscopy [27]. Molecular graphics reveal folding of long ribbons. Resistance of actin to cleavage is also a subject of study [28]. Actin-myosin systems show orientation with dielectrophoresis [29]. It has been verified that replacement of ATP with ADP changes dynamics and conformation of actin monomer [30]. Orientation of actine monomer in the F-actin monomer is also studied [27]. Mechanical properties of actin filament network depend on preparation, polymerization conditions and storage [26].

The FTIR spectrum of actin is shown in figure 4.1a. A half-power beta density is seen as a flat peak in 1700-2900cm\(^{-1}\) range. A weak
gaussian in absorption around 600cm\(^{-1}\) is also observed. The gaussian curve is fitted as shown in figure 4.1b.

![Figure 4.1 a The FTIR spectrum of actin only](image1)

![Figure 4.1 b The gaussian distribution in actin](image2)

The FTIR spectra of actin-TCNQ and actin-DDQ are shown in figure 4.2a and figure 4.2b respectively. Actin-TCNQ mainly shows spectrum of TCNQ\(^2-\) ion indicating that it is a highly ionic complex.
TCNQ\(^{2-}\) ions form segregated stacks through the matrix of actin molecules. This type of spectrum is also observed in hemoglobin-TCNQ and myoglobin-TCNQ as well as proteoglycan-TCNQ. The DDQ complex shows a range of nature of transition with 1800 cm\(^{-1}\) as the infrared absorption edge and a gaussian band around 600 cm\(^{-1}\). The nature of transition fits \(A \nu = B(h \nu - E_g)^3\) by plotting \((A \nu)^{1/3}\) vs \(h \nu\) which indicates that it is a forbidden indirect transition with \(E_g \approx 0.25\text{eV}\) as shown in figure 4.3a. Since actin is a macromolecule the transition is found to be indirect. The gaussian band around 600 cm\(^{-1}\) is also fitted by plotting \(\ln A\) vs \((k - k_0)^2\) as a straight line as shown in figure 4.3b.

Figure 4.2 a FTIR spectrum of actin-TCNQ CTC
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Figure 4.2 b FTIR spectrum of actin-DDQ CTC

![FTIR spectrum of actin-DDQ CTC]

Figure 4.3 a The nature of transition in actin-DDQ CTC

![Graph showing the nature of transition]

Figure 4.3 b The gaussian distribution in actin-DDQ CTC

![Graph showing the gaussian distribution]

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The other protein selected for study in the present work is thrombin. Aptamer-thrombin interactions are studied using electrochemical impedance spectroscopy [31]. Aptamers have high specificity and affinity for proteins, peptides and enzymes and are used in biosensors. Amplification of the nano-particles is also used in the determination of thrombin [32]. Thrombin-induced protein phosphorylation in human platelets is also studied [33]. The FTIR spectra of thrombin, thrombin-TCNQ and thrombin-DDQ are shown in figure 4.4 a-c respectively. The spectrum of thrombin shows a half-power beta density in 1625-2925cm\(^{-1}\) range as a flat peak. It also shows a gaussian band around 600cm\(^{-1}\). This property is similar to actin. The beta density is associated with intermolecular polaron hopping at the chain ends where the folded chains of polypeptide end. The amino acid residues are tied by the peptide bonds to form polypeptide chains in a primary or binary structure. The polypeptide chains are folded in three-dimensions to form tertiary or quaternary structure through hydrogen bonding and other molecular forces. Only fractal dimension remains to be one because of primary or secondary structure. Polarons are formed along polypeptide chains and move through the coiled structure by lever mechanism to finally jump from one protein molecule to the neighboring one at the chain ends. The gaussian band
around 600\,cm\(^{-1}\) shows delocalization of charge carriers at low enough energy. Half-power beta density and gaussian bands are fitted as shown in figures 4.5 a-b respectively. Thrombin-TCNQ again reveals the spectrum of TCNQ\(^{2-}\) ions consistent with strongly ionic nature of this charge transfer complex similar to actin-TCNQ. Thrombin-DDQ spectrum (figure 4.4c) is analyzed for nature of transition, half-power beta density and low energy gaussian band which are found. These fits are plotted as shown in figures 4.6a-c respectively. Nature of transition reveals forbidden indirect transition, beta density reveals polaron hopping and gaussian band reveals delocalization of polarons.

Figure 4.4 a The FTIR spectrum of thrombin only
Figure 4.4b The FTIR spectrum of thrombin-TCNQ CTC

Figure 4.4c The FTIR spectrum of thrombin-DDQ CTC

Figure 5.4a Half-power beta density fitted for thrombin
Figure 4.5 b The gaussian distribution in the thrombin only

Figure 4.6 a The nature of transition in thrombin-DDQ CTC

Figure 4.6 b The half-power beta density in thrombin-DDQ CTC
The third protein selected in the present study is γ-globulin is studied for its determination. The interaction of isofraxidin with human γ-globulin has been studied using fluorescence spectroscopy [34]. The binding of barbaloin to human γ-globulin using circular dichroism, FTIR spectroscopy and fluorescence spectroscopy has been studied [35].

The FTIR spectrum of γ-globulin is shown in figure 4.7a. There is half-power beta density between 1800cm$^{-1}$ and 2800cm$^{-1}$ which is fitted as shown in figure 4.7b. A low frequency gaussian bend between 400cm$^{-1}$ and 900cm$^{-1}$ is also evident which is fitted by plotting lnA vs (k-k$_0$)$^2$ as shown in figure 4.7c.
Figure 4.7a The FTIR spectrum of $\gamma$-globulin only

Figure 4.7b The half-power beta density in $\gamma$-globulin CTC

Figure 4.7c The gaussian distribution fitted for $\gamma$-globulin CTC
The FTIR spectra of $\gamma$-globulin–TCNQ, $\gamma$-globulin-TCNE, $\gamma$-globulin-DDQ, $\gamma$-globulin-chloranil and $\gamma$-globulin-I$_2$ are shown in figure 4.8 a-e. $\gamma$-globulin-TCNQ spectrum contains two regions of nature of transition, one between 2000cm$^{-1}$ and 2900cm$^{-1}$ and the other between 1000cm$^{-1}$ and 1600cm$^{-1}$. Both are fitted to be $A\nu = B (\nu - E_g)$ as two-dimensional transition between valence and conduction band. This shows that $\gamma$-globulin-TCNQ is a layered material which is shown in figure 4.9 a and figure 4.9 b respectively. Also a low-frequency gaussian band is found below 900cm$^{-1}$. This is also fitted as shown in figure 4.9 c.

Figure 4.8 a The FTIR spectrum of $\gamma$-globulin-TCNQ CTC
Figure 4.8b The FTIR spectrum of γ-globulin-TCNE CTC

Figure 4.8c The FTIR spectrum of γ-globulin-DDQ CTC

Figure 4.8d The FTIR spectrum of γ-globulin-chloranil CTC
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Figure 4.8a The FTIR spectrum of γ-globulin-iodine CTC

Figure 4.9a The nature of transition in the range of 2000 cm\(^{-1}\)–2900 cm\(^{-1}\) in γ-globulin-TCNQ CTC

Figure 9b The nature of transition in the range of 1000 cm\(^{-1}\)–1600 cm\(^{-1}\) in γ-globulin-TCNQ CTC

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The FTIR spectrum of \( \gamma \)-globulin-TCNE contains one region of nature of transition which is fitted as \( A(h\nu) = B(h\nu - E_g)^3 \). Thus it is a forbidden indirect transition which is shown in figure 4.10 a. There is a weak gaussian band below 800cm\(^{-1}\) which is fitted as shown in figure 4.10b.
The FTIR spectrum of γ-globulin-DDQ contains one region of nature of transition being $A\nu = B (h\nu - E_g)^3$. So again it is a forbidden indirect transition which is shown in figure 4.11a. Intermediate strength gaussian band is also fitted in low frequency range which is shown in figure 4.11b.
The FTIR spectrum of γ-globulin-chloranil contains two regions of nature of transition which are fitted as forbidden indirect transitions corresponding to two band transport as shown in figures 4.12a and 4.12b. Broad gaussian band below 900 cm$^{-1}$ is also fitted as shown in figure 4.12c.
The FTIR spectrum of \( \gamma \)-globulin-iodine contains one region of nature of transition which is fitted as \( A h v = B (h v - E_g)^3 \), a forbidden indirect transition which shown in figure 4.13a. The gaussian band between 400cm\(^{-1}\) and 900cm\(^{-1}\) is also fitted as shown in figure 4.13b.
Band gaps of $\gamma$-globulin CTCs show straight line when plotted against full-widths at half-maximum (FWHM) of beta density peaks as shown in figure 4.14. FWHM increases band gap decreases which shows that as electron-phonon coupling constant increases, band gap decreases.
The total change is found to be of the order of phonon energy or of the order of Urbach tail. This is same behavior as found elsewhere for biomolecular complexes [22].

![Graph showing band gap vs FWHM for γ-globulin CTCs]

**Figure 4.14 γ-globulin CTCs shows straight line graph for band gap vs full width at half maximum**

### 4.4 Conclusion

The solid state spectroscopy of three proteins namely actin, thrombin and γ-globulin and their CTCs reveal polaron hopping due to beta density found common in the FTIR spectra. This also reveals that fractal dimensionality of globular or fibrous protein is unity as the proteins are composed of linear polypeptide chains which are folded. Band gaps vs FWHM of beta density particularly for globulin complexes reveals rectilinear behavior as already found in other biomolecular complexes.
References


34. Lei Rui-Xia, et. Al, Lanzhou University, Natural Sciences, 45, (2009)