CYCLIC VOLTAMMETRIC INVESTIGATIONS ON COPPER (II) AMINO ACID AND DIPEPTIDE SYSTEMS

A THESIS
SUBMITTED FOR THE DEGREE OF
DOCTOR OF PHILOSOPHY

BY

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I N D I A

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To

My Father

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STATEMENT

I hereby declare that the matter embodied in this thesis is the result of investigations carried out by me in School of Chemistry, University of Hyderabad, Hyderabad, India, under the supervision of Dr. P.S. Zacharias.

In keeping with the general practice of reporting scientific observations, due acknowledgement has been made wherever the work described is based on the findings of other investigators.

George Thomas

Hyderabad
June 1984

CERTIFICATE

This is to certify that the work presented in the thesis entitled 'Cyclic Voltammetric Investigations on Copper(II) Amino Acid and Dipeptide Systems' has been carried out by Mr. George Thomas under my supervision and the same has not been submitted elsewhere for a degree.

P. S Zacharias

Hyderabad
June 1984

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Finally, I wish to thank my wife, Lucy, for her understanding, patience and love which added to my pleasure in working.

Hyderabad
June 1984

George Thomas

Preface

Complexes of amino acids and oligo peptides are involved in the exchange and transport mechanisms of trace metals, especially of Cu(II) in human systems. The involvement of Cu(II) ions in many of the biological redox processes is established. Studies on complexes of amino acids or small peptides can provide useful informations regarding the redox process in these systems. Although the chemistry of Cu(II) amino acid and peptide complexes is fairly well known, very little is known about their redox properties particularly of amino acid and dipeptide complexes. The first chapter reviews the known chemistry of Cu(II) amino acid and peptide complexes. The known chemistry of these systems is related to the formation, stability and spectral properties of them in aqueous media under various pH conditions. This chapter also briefly discusses the basic principles of cyclic voltammetry which is the major technique employed for the study of the reduction of these Cu(II) systems.

In chapter two the results of the cyclic voltammetric studies on the reduction of Cu(II) amino acid complexes is presented and discussed. The solid amino acid copper(II)

complexes have the general composition $CuA_2.nH_2O$. Cyclic voltammetric studies are carried out in aqueous media at HMDE with $NaClO_4$ as supporting electrolyte at neutral pH. Cyclic voltammetric behaviour for all the Cu(II) amino acid complexes is similar. The basic features of the voltammogram are the following. In the first forward scan, a cathodic peak C, at about -0.290 V is observed. During the reverse scan one anodic peak D at about -0.230 V and another peak B at about +0.03 V are observed. From the second scan onwards an additional cathodic peak A at about -0.03 V develops. From the experimental evidences, the electrode mechanism has been identified and is briefly discussed. The reduction of the starting CuA_2 complexes proceeds through a one-electron step generating intermediate Cu(I) complexes. A fraction of the Cu(I) species gets reoxidized to the corresponding Cu(II) complexes in the reverse scan resulting in the anodic peak D, about 60 mV anodic to peak C. The other fraction of Cu(I) species undergoes chemical transformations eventually generating Cu^o at the electrode. The generated Cu^o gets oxidized to Cu(II) ions at more anodic potentials giving rise to the anodic peak B. During the second scan these Cu(II) ions get reduced to Cu^o, to give the peak A. The presence of the Cu(I) species is established by trapping the short-lived Cu(I) species using bathocuproin reagent.

The observed CV pattern is consistent with an ECE mechanism for the electrode process in which E_2^o is anodic to E_1^o and $n_2 = 2$ while $n_1 = 1$, both electron-transfers being nearly reversible. Cyclic voltammetric response of these complexes at various pH values is also recorded. The major changes in the CV profile on decreasing the pH are (i) the peak heights of C and D decrease while that of A and B increase; (ii) at intermediate pH values most of the complexes exhibit a broad peak which becomes narrow on further decrease of pH; (iii) peak A appears from the first scan itself. The reasons for these observations are discussed at length.

The results on the reduction of Cu(II) dipeptide complexes is presented and discussed in chapter 3. The general features of the cyclic voltammograms are roughly the same as those of Cu(II) amino acid complexes, i.e., the redox couples C-D and A-B are observed. For these systems too the reduction proceeds through a one-electron process generating the Cu(I) dipeptide complexes. The transient existence of this short-lived species is further established by trapping the intermediate Cu(I) species using bathocuproin reagent. The overall electrode process belongs to the category of ECE mechanism where E_2^o is anodic to E_1^o; $n_2 = 2$ and $n_1 = 1$. The second electron-transfer is nearly reversible while the first one is

quasi-reversible. The protonation studies carried out on the system revealed the existence of protonated species and free Cu(II) ions at low pH conditions. These results are also discussed in detail.

Chapter 4 contains the results of the cyclic voltammetric studies on the reduction of mixed ligand complexes of Cu(II) containing amino acids and peptides. Three major types of mixed (ternary) systems are investigated. They are: (i) mixed amino acid complexes of the type Cu(A)(B) where A and B are two different amino acids with oppositely charged side chains (ii) systems containing one amino acid and 2,2'-bipyridyl ligand (iii) histidine containing mixed amino acid and dipeptide systems. The CV response of the Cu(A)(B) type complexes and the conclusions derived are very similar to that of binary Cu(II) amino acid complexes discussed earlier.

The mixed ligand Cu(II) complexes of amino acids containing bipyridyl are of the composition Cu(BPY)(A). The CV response of these complexes also happens to be similar to the binary Cu(II) amino acid complexes. However, the CV shows that the intermediate Cu(I)(BPY)(A) complexes are more stable than the $Cu(I)(BPY)_2$ complexes. The protonation studies revealed that the

end-product of protonation is not free Cu(II) ions as observed in the case of other systems, but is the 1:1 complex, $Cu(BPY)(H_2O)_2$, which undergoes a one-electron reversible reduction.

The studies on the reduction of histidine containing mixed Cu(II) amino acid and dipeptide complexes form the last part of the chapter. The CV response of these complexes are studied in solution by mixing the components in 1:1:1 ratio and adjusting the pH to the required value. The histidine containing Cu(II) amino acid complexes have only a sharp cathodic peak at about -0.350 V and is identified as due to an adsorption-controlled process. In the case of histidine containing Cu(II) dipeptide complexes the CV response is almost similar to that of binary dipeptide complexes. But the first reduction of the complexes occurs at much more negative potentials than the binary dipeptide complexes. At faster scan rates an adsorption wave is apparent at about 50-60 mV cathodic to the first reduction peak.

CHAPTER 1

INTRODUCTION

1.1 Metal Ions in Biological Systems

The importance of metal ions in the vital functions of living organisms has become apparent in recent years. This has resulted in considerable research activities in the synthesis, study of formation, stability, structure, and reactivity of biologically important metal ion-containing compounds of low and high molecular weight. While new model complexes are being synthesised, known complexes are extensively used to understand more about the complicated biological systems.

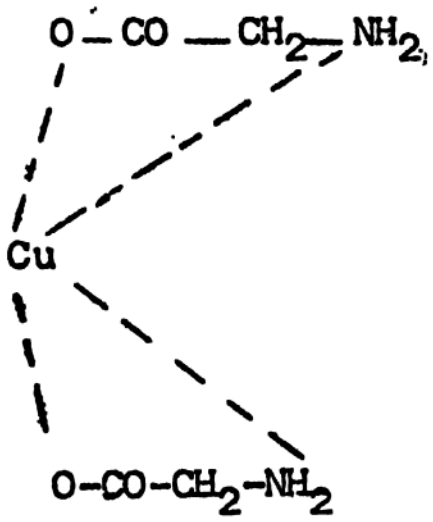
Of the metal ions which are essential for growth and development of living organisms, copper has been recognised at an early stage because of the isolation of

coloured copper proteins. However, the isolation of Cu(II) amino acid complexes from normal human serum by Sarkar and co-workers[1] in 1966 has given a tremendous boost to the synthesis and study of model Cu(II) complexes. The low molecular weight Cu(II) amino acid complexes are considered to be involved in the transport of Cu(II) ions between blood and the tissues.[2] In addition, several ternary histidine Cu(II) amino acid complexes were detected along with $Cu(L\text{-}His)_2$, in human serum.[1] One of these complexes, L-His Cu(II) L-Thr, was actually isolated, crystallized and its X-ray structure was determined.[3] Subsequently several ternary copper(II) amino acid complexes were also synthesised and their solution equilibria studied.[4-6] The Cu(II) amino acid and peptide complexes, both binary and ternary have been used as potential model systems for enzyme-metal-substrate (EMS) complexes. In the following sections a brief report of the known chemistry of Cu(II) amino acid and peptide complexes will be presented.

1.2 Binary Copper(II) Amino Acid Complexes

Copper(II) amino acid complexes were first reported in 1854 by Gossmann.[7] Ley, a few decades later[8] studied the physical properties of the metal complexes of glycine

and other amino acids and found them non-conducting. From various data available, he formulated the structure of the glycine complex as in 1 and referred it as an 'innere-Komplexsalz'. The major part of the known chemistry of Cu(II) amino acid complexes has been worked out during the last three decades.[9-14]

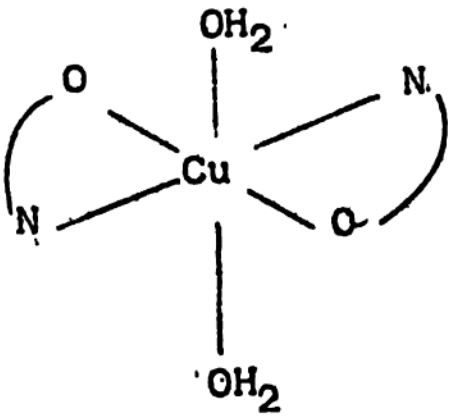


1

A. Structure, Stability and pH-Dependence of Copper(II) Amino Acid Complexes

Structural aspects of a great number of copper(II) amino acid complexes in the solid state are understood from X-ray crystallographic measurements.[15] In contrast, precise knowledge of their solution chemistry is lacking. Several workers have used techniques like ^{13}C NMR [16,17],

ESR[18], UV, visible, CD,[19-21] and potentiometric titrations[22,23] to study the structure, stability and pH-dependence of these complexes. It is now accepted that bidentate amino acids coordinate to copper(II) through nitrogen (amino) and oxygen (carboxyl) donor atoms. They form mainly two types of complexes viz. 1:1, $[CuA]^+$ and 1:2 $[CuA_2]$ depending upon the pH conditions. In normal conditions, in the pH range 4.5-5.8, the 1:1 complexes predominate while the 1:2 complexes predominate in the pH region, 6.2-7.5.[16-23] The structures of bidentate amino acid complexes of Cu(II) at neutral pH, is shown in structures 2-6.

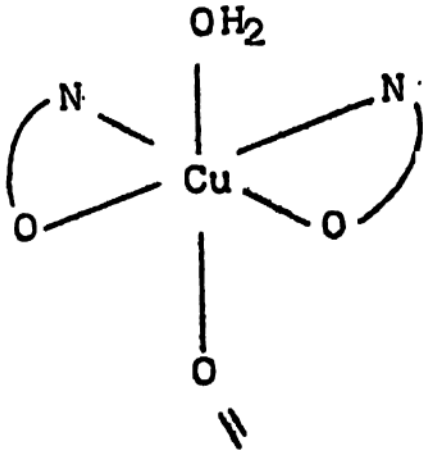


2

$Cu(L\text{-}Pro)_2.2H_2O$[9]

$Cu(L\text{-}His)_2(NO_3)_2.2H_2O$[9]

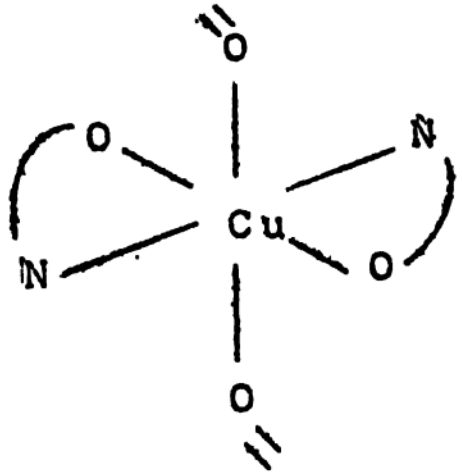
$Cu(\beta\text{-}Ala)_2.2H_2O$[9]



3

$Cu(Gly)_2.H_2O$[9]

$Cu(Glu)_2.H_2O$[9]

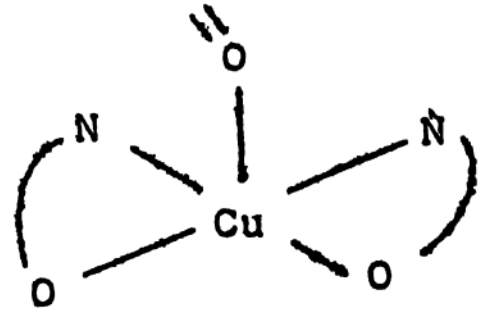


TRANS

4

$Cu(L\text{-}Ala)_2$[9]

$Cu(Met)_2$[24]



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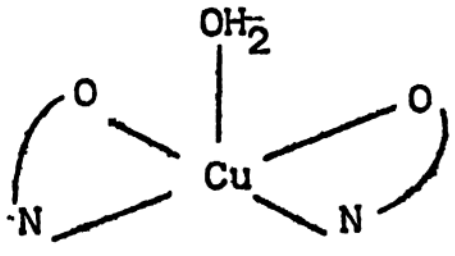
$Cu(L\text{-}Ala)_2$[9]



CIS

6

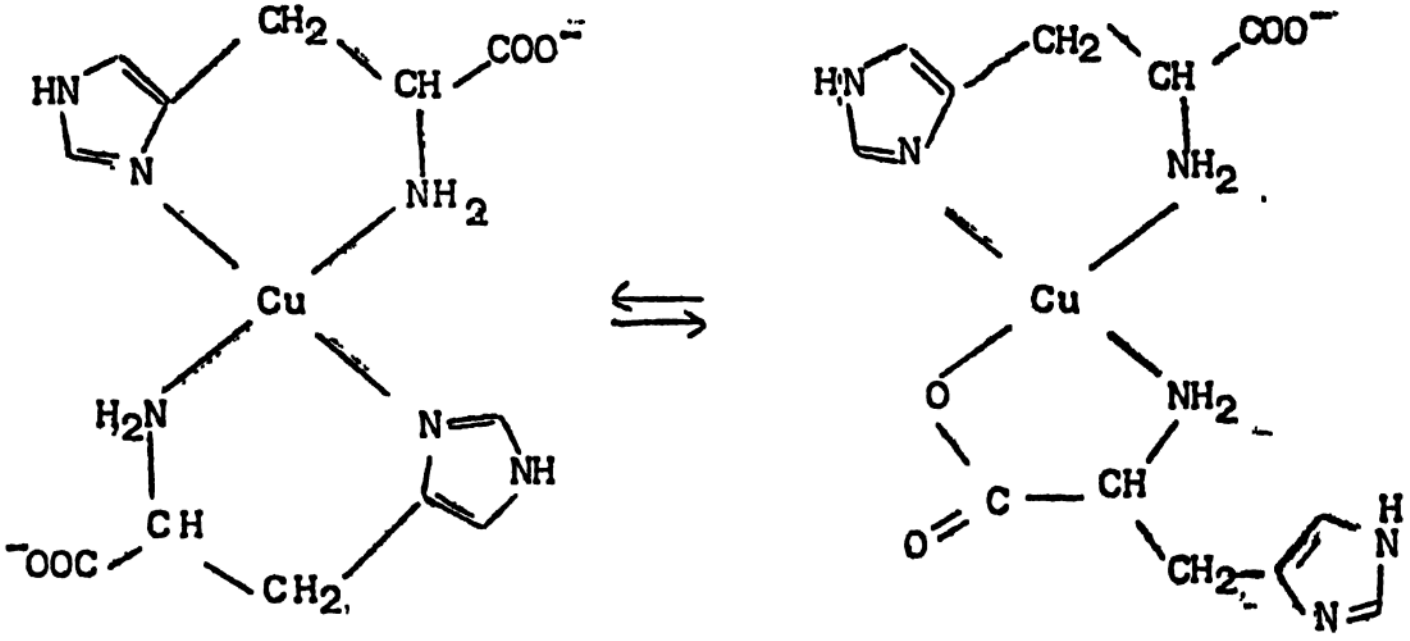
$Cu(L\text{-}Ile)_2 \cdot H_2O$[25]

$Cu(L\text{-}Ser)_2 \cdot H_2O$[26]

In all these complexes, a square-planar arrangement of the donor atoms around the metal ion is maintained with the metal ion slightly displaced towards the axial position. The properties of the above type complexes are decided mainly by the four in-plane donor atoms. The nature of the donor atoms on the apical positions O(water) or O(carboxyl) has very little influence.[9]

The structure of Cu(II) histidine complexes are more complicated. In spite of the large amount of work done on these systems, the structure of Cu(II) L-histidine complexes is not completely elucidated. The main controversy centers around the structure of the physiologically important[27] $Cu(L\text{-}His)_2$ complex. All attempts to crystallize this complex has failed. Recently many models of its structure have appeared in literature.[28-31] Camerman and co-workers[21] in 1978, after considering all the available informations and on the basis of the crystal structure data of Cu(D-His) (L-His) complex and that of $Cu(DL\text{-}His)_2$, proposed a structure for $Cu(L\text{-}His)_2$ where the two histidines coordinate by imidazole and amino nitrogens, trans to each other in a square-planar arrangement with two water molecules coordinating at the axial positions. Later, Goodman and co-workers[18] from ESR studies suggested the existence of an equilibrium between structures having

three and four nitrogen atoms coordinated to Cu(II) as shown in 7 and 8.



7 ⇌ 8

Although the word 'stability' has been used to describe a number of chemical properties of a complex, its use in this section will be restricted to the equilibrium constant for a reaction of the metal ion and the ligands to form the complex. Over the years many workers[10-13] have studied the stability of Cu(II) amino acid and peptide complexes using different methods. In its simplest reaction Cu(II) forms two types of complexes with amino acids, $[CuA]^+$ and $[CuA_2]$, and the overall stability constant, K_s sometimes called β, can be represented as in equation 1.1.

$$K_s = K_1K_2 = \frac{[MA_2]}{[M]\,[A^-]^2} \qquad \ldots 1.1$$

where

$$K_1 = \frac{[MA^+]}{[M]\,[A^-]}, \quad K_2 = \frac{[MA_2]}{[MA^+]\,[A^-]}$$

Hence the overall dissociation constant, K_d, can be represented by equation 1.2.

$$K_d = 1/K_s \qquad \ldots 1.2$$

In the study of reaction mechanisms and thermodynamic properties, the principal use of formation-constants is to calculate the concentration of each species at equilibrium under given conditions. A detailed review of the stability constants of Cu(II) amino acid complexes has appeared recently.[22] Kruck and Sarkar[20] from analytical potentiometric titration technique calculated the stability constants for the various complexes formed between Cu(II) and L-histidine over a wide range of pH (2-11) and deduced the structures for them. It is generally observed that complexes formed by 5-membered rings are more stable than that formed by 6-membered rings.[32] In comparing the stabilities of many bidentate amino acids, it is apparent that L-proline forms the strongest complex with Cu(II).[9]

The pH dependence of Cu(II) amino acid complexes can easily be monitored from its absorption spectra. The Cu(II) amino acid complexes have an absorption maximum at about 625 nm in the near neutral pH values. As early as in 1932, Borsook and Thaimann[33] showed that lowering of the pH of the Cu(II) amino acid complex solution, resulted in a red shift of the absorption band with a decrease in the ϵ value, whereas an increase in the pH values from the neutral pH, increases the ϵ value with virtually no shift in the band position in the pH range 6-8.5. At very low pH values (0.5-2.5) the absorption curve corresponds to that of cupric acetate in alcohol. The pH dependency of Cu(II) amino acid complexes have been studied extensively by many workers.[17-20,23,32,34] Depending on the metal-to-ligand ratio and the pH conditions, complexes of various stoichiometries are possible. Bidentate amino acids generally forms complexes like $[CuA]^+$, $[CuA_2]$ in the pH range 3-10. Sarkar and co-workers[20] observed species like $[Cu\ H_{-1}\ A_2]$ and $[Cu\ H_{-2}\ A_2]$ in addition to the other two common forms, in the Cu(II) L-serine system at high pH values. For Cu(II) L-histidine system, species like $[CuHA]^+$, $[CuA]$, $[CuH_2A_2]^{2+}$, $[CuHA_2]$, $[CuA_2]^{2-}$, $[CuH_{-1}A_2]^-$, $[CuH_{-1}A]$, and $[Cu_2H_{-2}A_2]$ are observed in solution at various experimental conditions,[20] where H_3A refers to L-histidine.

From ESR hyperfine A values of Cu(II) amino acid complexes, Goodman and co-workers[18] detected the formation of 1:1 $[CuA]^+$ and 1:2 $[CuA_2]$ complexes in solution. Using the nitrogen super-hyperfine splittings, it has been shown that 1:2 complexes exist in solution as an equilibrium mixture of cis and trans isomers. ESR spectroscopy was used to investigate Cu(II) L-histidine complexes too in solution.[18,35] Isotropic g and A values were found to be in the ranges 2.144-2.157 and 5.5-6.5 mT respectively for $[CuA]^+$ and 2.118-2.127 and 6.4-7.1 mT respectively for $[CuA_2]$ complexes.

B. Optical Properties

The absorption bands observed for Cu(II) amino acid and peptide complexes can be broadly divided into three types: (i) transitions occurring predominantly due to ligand which can be modified by complexation (ii) Charge-transfer (CT) transitions that involve both ligand and metal ion; and (iii) d-d transitions occurring mainly on the metal ion.

(i) Ligand Transitions. - Amino acids bearing aliphatic side chains display only end-absorption at wave lengths longer than 200 nm. But most of the L-amino acids display

a positive CD band at 210 nm, where the absorption displays only weak bands.[36] Side chains of aromatic amino acids exhibit absorption bands in the 220-300 nm region. Upon chelation, the CD magnitudes of the aromatic transitions undergo enhancement due to a reduction in the number of allowable conformers.[36]

(ii) Charge-Transfer Transitions. - The charge transfer (CT) transitions are associated with the transfer of charge to or from ligand to metal ion. Most of the CT absorption bands are of high intensity ($\epsilon > 10^3$) and generally occur in the UV region. Ligand to metal ion electron-transfer transitions are very common. Cu(II) amino acid complexes show a strong CT band near 220 nm which gives rise to oppositely signed CD peaks at 220 nm and 255 nm.[36] It has been suggested that both the carboxylate and amino groups are responsible for the CT band occurring from 230-250 nm.[37]

(iii) d-d Transitions. - The d-d transitions of transition metal ion complexes provide one of the best methods to study its optical activity. Since Cu(II) complexes of amino acids and small peptides form predominantly, distorted octahedral complexes, the discussion on d-d transitions is confined to octahedral geometry in this section. In

octahedral complexes these transitions are often electric-dipole forbidden and hence exhibit only weak absorption bands but the CD response is usually very strong. Cu(II) complexes of amino acids and peptides exhibit d-d transitions in the visible region between 620-650 nm (ϵ 50-100) and in the range 590-620 nm, the Cu(II) L-amino acid complexes give two CD extrema (both negative) and another one at about 700 nm (positive)[19] in addition to a small positive CD band at about 730 nm ($\Delta\epsilon_{\sim}$+ 0.05).[38] The hexadecant rule formulated to explain the sign identity observed in the case of Cu(II) amino acid complexes is applicable to the ligand field bands where vicinal effects of substituents generate optical activity. The three CD extrema are assaigned to the three possible transitions[19] which are in the order of increasing energy as,

$$^{B}_{1g} \longrightarrow \; ^{A}_{1g} < ^{B}_{1g} \longrightarrow \; ^{B}_{2g} < ^{B}_{1g} \longrightarrow \; ^{E}_{g}.$$

1.3 Binary Copper(II) Dipeptide Complexes

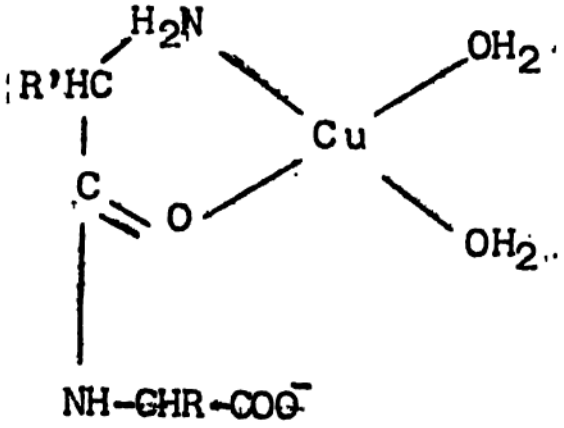
Since the isolation of blue copper proteins, the interest in the chemistry of dipeptide and oligopeptide Cu(II) complexes has been growing. The role of copper in these proteins is not yet fully understood even though the geometry of the copper centre, the oxidation state

of the metal, the nature of the ligand groups coordinated to the metal etc. are found to be important. Physical measurements on redox copper proteins suggest that Cu(II) atoms may have a distorted tetrahedral structure. Proteins also enforce upon itself certain conformational restrictions which helps the metal ion to carry out its highly specific functions. Attempts to understand the role of copper in proteins by studying the low molecular weight model complexes which can mimic the active site of proteins have been partly successful. Even though it is difficult to derive any one-to-one correspondence to the properties of these systems with that of high molecular weight biological systems, such studied have provided important informations regarding the geometry and other properties of the metal binding sites in metal protein systems. Enormous amount of work has been done, particularly on Cu(II) di and other small peptide complexes. Several recent reviews are available on this topic.[9,11,13,15,39-41]

A. Structure, Stability, and pH-Dependence

The structure of Cu(II) di and other small peptide complexes discussed in literature is based on X-ray crystal and solution studies. It is now generally accepted that complex formation with Cu(II) and peptides start with

α-amino group and forms chelates involving the α-amino nitrogen and the oxygen of the neighbouring amide group (9). These complexes get deprotonated at the amide group in the pH range, 4-7, and chelates are formed then by the coordination of the amide nitrogen,[42] α-amino nitrogen and the carboxyl oxygen atoms. These two situations are shown in structures 9 and 10.



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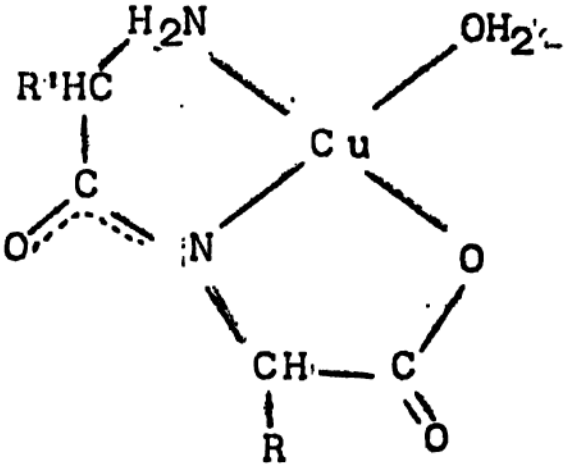
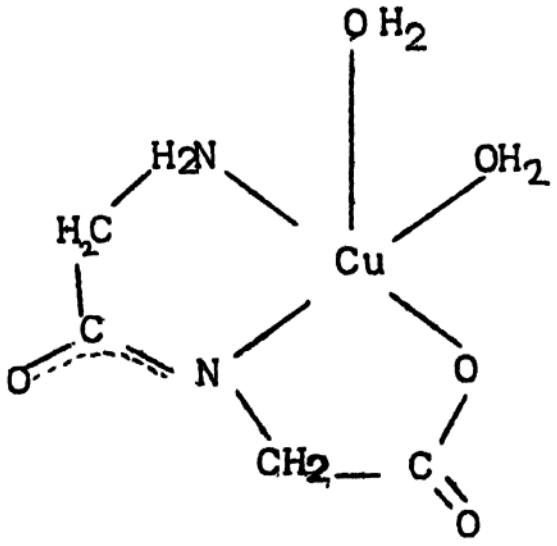


10

Cu(II) diglycine complex has been subjected to extensive studies by many workers. At neutral pH, the diglycine coordinate to Cu(II) via α-amino nitrogen, deprotonated peptide nitrogen and the carboxyl oxygen atom. The structure of this complex is shown in 11.

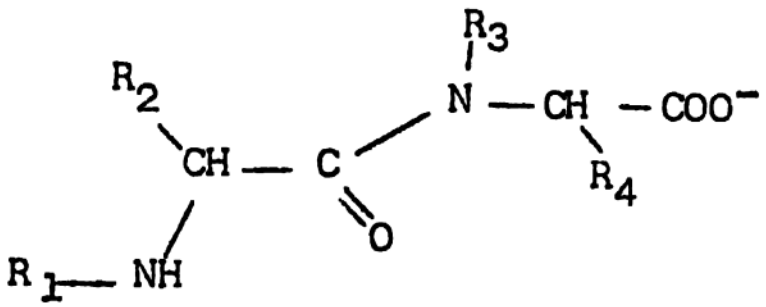


11

Originally deduced from potentiometric titrations,[43] this structure which exists in neutral aqueous solutions[44,45] has been confirmed by optical[19] and calorimetric[46] studies in solution and by X-ray study[47] in crystal where the fourth position is occupied by water molecule. Another water molecule in apical position completes the square-pyramidal geometry. Due to the planar amide bond the

terdentate glygly assumes only the planar conformation about the metal ion. Most of the Cu(II) dipeptide complexes have a square planar arrangement, often distorted, in the plane of the metal ion. Normally the additional ligands in the axial positions will be O (water) or O (carboxyl) atoms, completing the generally observed square-pyramidal geometry.[48-51] In histidine containing dipeptide complexes the imidazole nitrogen coordinates instead of carboxyl oxygen.[48,52]

The stability of Cu(II) oligopeptide complexes are significantly greater than that of simple amine complexes due to the formation of stable 5-membered chelate rings resulting from the peptide coordination. The stability of the peptide complexes is remarkably affected by sterric interactions.[53] A general structure of the dipeptide is shown in 12. A bulky group neighbouring the amino group has a much stronger influence on the stability of the complexes than the same group near to the amide group. It is noted that a substituent at R_3 or R_4 positions [glycyl(N-or α-alkyl) glycinates] does not have any influence on the stability of $[CuLH]^+$ complexes but the stability depends only on the basicity of the amino group. On the other hand a substituent at R_1 or R_2 positions [(N- or α-alkyl glycyl) glycinates] decrease the stability of $[CuLH]^+$ complexes considerably. However, alkyl



12

substituents at R_1 and R_2 facilitates ionization of the amide proton and hence CuL type compounds are more stable than the compounds from unsubstituted dipeptides.[53] Stability constants for various copper(II) dipeptide complexes have been determined by Sigel using analytical potentiometric titrations.[42,53] A weak coordination by hydroxy or thio groups in the side chains to Cu(II) is also observed.[48]

pH dependence of Cu(II) peptide complexes have been studied using various techniques.[54-57] It is generally accepted that simple aliphatic dipeptides form complexes with Cu(II) in various stoichiometries like $[CuLH]^+$, $[CuL]$, $[CuL(OH)]^-$ and $[Cu_2L_2H]^-$ etc. (H_2L = H_3N^+-CHR-CO-NH-CHR'-COO^-; R,R' = alkyl). As mentioned earlier the dipeptides at low pH values ($\sim$ 4) coordinate to Cu(II) through terminal amino group and the amide oxygen. The ionization of the peptide nitrogen occurs in the pH range 4-7 and the chelates undergo structural rearrangement as in equation 1.3.

$$\underline{9} \rightleftharpoons \underline{10} + H^+ \qquad \ldots 1.3$$

At higher pH conditions (>8) the formation of $[CuL(OH)]^-$, is postulated. This results from the deprotonation of the equitorial water molecules of the complex. The formation of the dimer species, $[Cu_2L_2(OH)]^-$ have been reported by many workers[58,59] but its structure is not well established. Species distribution curves over a wide range of pH values are available in literature.[57-60]

B. Optical Properties

The general observations made earlier in the case of Cu(II) amino acid complexes are valid for copper(II)

peptide complexes also. A comprehensive discussion of optical properties of Cu(II) dipeptide complexes is given by Martin.[11] Kober and Haw[61] were the first to seek a correlation between colour and the coordination of Cu(II) peptide complexes and to classify the complexes on the basis of the region of absorption in optical spectra. Brill and co-workers[62] classified the visible band (550-730 nm) of Cu(II) peptide complexes as due to d-d transitions. The spectral shift to shorter wave length corresponds to an increase in the ligand field strength observed when donor oxygen atoms of carboxyl groups or of water molecules are replaced by negatively charged donor nitrogen atoms of deprotonated peptide groups. For the N donors commonly found in proteins, the order of ligand field effects is N (imidazole) < N (peptide) < N (amino).[9] Sportelli and co-workers[52] studied the influence of aromatic amino acid residues in small peptides on their complexation behaviour to Cu(II). Characteristic absorption bands have been established for complexes in which the α-NH_2 and subsequent peptide nitrogen atoms are involved. The order is α-NH_2 group with one peptide amide nitrogen has λ_{max} 660 nm; α-NH_2 with two peptide amide nitrogen has λ_{max} 575 nm; α-NH_2 group with three peptide amide nitrogens has λ_{max} 515 nm.[63-65]

The CD of Cu(II) dipeptide complexes is found to be an additive property of independent contributions from amino- and carboxyl-terminal amino acid residues.[19] The negative sign observed in the case of L-amino acids persists in the glycyl-L-amino acid dipeptide complexes and bulky amino acid side chains yield a net positive sign in the L-amino acyl-glycyl dipeptides.

C. ESR Studies

ESR technique has been extensively used to study the Cu(II) peptide complexes both in solution and in solid state. It is found that in frozen solutions of Cu(II) complexes of small dipeptides, the ESR spectra are very close to axial symmetry.[41] At pH above 12, the bulky side chains cause a change in ESR symmetry from axial to orthorhombic. This is characterized by a decrease in $A_{\parallel}$ values. The changes depend on the side chain bulkiness.[41] Sportelli and co-workers[52] observed, from ESR studies, that in histidine containing small dipeptide Cu(II) complexes, the imidazole nitrogen coordinate to Cu(II) in the equitorial plane in Gly His, Gly His Gly type peptides whereas it coordinates in the axial plane in Gly Gly His type peptides. But the exact nature of the observed influence of the aromatic residues in the

side chains on the coordination behaviour is not established so far. Viola and co-workers[60] used ESR to investigate the solution structure of Cu(II) carnosine complex and proposed a monomeric structure in solution at room temperature with the β-amino nitrogen, deprotonated peptide nitrogen, α-carboxyl oxygen and a water molecule in the equitorial plane of the Cu(II) and the imidazole nitrogen as the fifth coordinating atom in the axial position. More recently Kittl and co-workers[56] reported the formation constants for various complexes formed between Cu(II) and a series of aliphatic dipeptides in solution over a wide range of pH values using ESR. Although the accuracy is lower than that using the potentiometric titrations, the ESR method provides some advantages. If two complexes show similar species distribution and pH-dependence, for example, $[CuL_2H_2] \leftrightarrow [CuL]$ or $[CuL_2]^{2-} \leftrightarrow [CuL(OH)]^-$, these species cannot be distinguished by means of potentiometric titrations, whereas ESR method allows their detection. The formation of different species like $[CuLH]^+$, $[CuL]$, $[CuL(OH)]^-$ and $[Cu_2L_2(OH)]^-$ in solution with 1:1 metal-to-ligand ratio have been detected by ESR. These experiments excluded the possibility of the formation of species like $[CuL_2]^{2-}$ and $[CuL_2H_2]$ (H_2L = H_3^+N-CHR-CO-NH-CHR'-COO^-, R,R' = alkyl). Size of the side group influences the coupling constant A, but a direct correlation between these two is yet to develop.[56]

1.4 Ternary Copper(II) Complexes

Mixed chelates are common in biological fluids and they play an important role in biological processes. The detection and isolation of ternary Cu(II) amino acid complexes from normal human serum had triggered the interest in ternary Cu(II) amino acid and peptide complexes.[12,13]

Mixed chelation here means the coordination of the same metal ion by at least one amino acid or peptide (A) and a bidentate, or polydentate ligand (B), which may or may not be an amino acid or peptide. This gives the general formula Cu(A)(B), for the Cu(II) ternary complexes. A quantitative expression for the interactions between metal ions and ligands, A and B can be given by the equilibrium (overall stability) constant of the reaction in which the ternary complex is formed. The direct detection of mixed ligand complexes with amino acids or peptides is difficult as their properties are not very different from those of the corresponding binary species. Because of this reason potentiometry and absorption spectrometry are of limited use. The best of the available methods seems to be the chromatographic methods.[13]

A. Structure and Stability

It is well understood that mixed chelates are much more stable than the corresponding parent complexes. Martin and co-workers[12] have collected and presented all data available upto 1971 relating to stability of mixed chelates. It is reported, that in general, mixed chelation is not possible when the parent complexes are of different geometrical configurations.[66] Ternary Cu(II) amino acid complexes have structures very similar to that of binary complexes. They prefer square-planar geometry with weak axial ligation. Not many ternary Cu(II) complexes are isolated and structure studied. The crystal structure of Cu(L-His)(L-Thr).H_2O was determined[3] first and later on, the crystal structure of Cu(L-His)(L-Asn)[67] was determined. These two complexes and the Cu(L-His)(L-Gln) together form a group in which histidine is terdentate unlike in other histidine containing ternary Cu(II) amino acid complexes,[68,69] where it is bidentate. In Cu(L-His)(L-Thr) complex, the distance of the O (carboxyl) histidine to Cu(II) is 2.58 Å, as compared with 1.97 Å for the O(carboxyl) of threonine; moreover the O(carboxyl) atom of histidine is in an irregular axial position. This latter bond is not strong and in aqueous solution it may be replaced by water molecules.[13]

B. Optical Properties

Optical properties of mixed ligand complexes are very similar to that of binary species. Nevertheless many workers have used absorption spectroscopy and CD values to study mixed ligand complexes.[70-75] It is observed that in most of the ternary copper(II) complexes, the absorption maxima shift to shorter wave length region by about 20 nm (610-620 nm) compared to binary complexes with almost the same ϵ values as that of binary complexes. In neutral solution a good number of ternary systems exhibit positive CD extrema at 620-630 nm with a CD magnitude enhancement, which is attributed to the increased rigidity due to intramolecular ligand-ligand interaction.[70,71] In case of histidine-containing ternary amino acid Cu(II) complexes the above effects are more pronounced and understood to be due to the presence of a π-acceptor (imidazole) and a π-donor (carboxyl) group in the Cu(II) coordination plane. This is also true in the case of ternary systems involving coordinated aromatic nitrogens.[71,76]

1.5. Copper Amino Acid (A) Amino Acid (B) Type Ternary Complexes

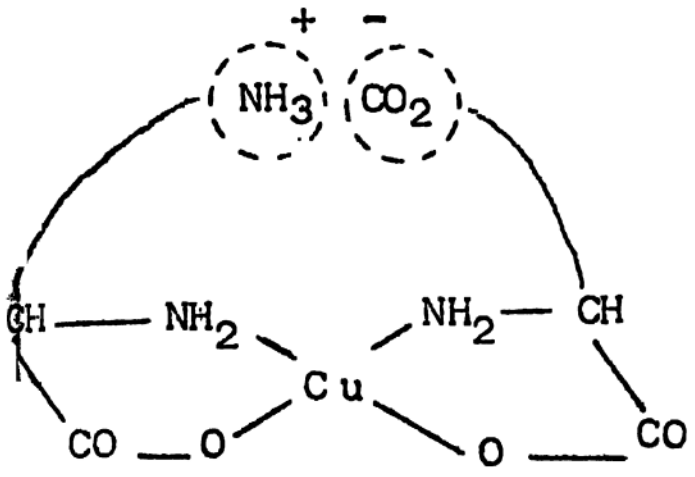
This group of complexes belong to the most studied group of mixed amino acid copper(II) complexes. When both A and B are simple amino acids, only one type of

mixed chelate is formed. On the other hand if A and/or B contains a functional side chain, different modes of coordination is possible. Histidine-containing ternary Cu(II) amino acid complexes behave in a different manner from other mixed complexes and are generally studied as a separate group.[35] Ternary Cu(II) complexes containing amino acids with oppositely charged side chains belongs to another group which has an enhanced stability due to intramolecular ligand-ligand interactions.[70-75] The existence of aromatic side chains increase the stability of ternary Cu(II) amino acid complexes due to 'back-coordination-stabilization effect', where electrons of the t_{2g} orbitals of Cu(II) are directed to the vacant π-orbitals of the ligand. The 'mixed chelation stabilization effect' reaches its maximum in the Cu(L-His)(L-Thr) system.[5] The added stability is explained as due to the π-acceptor property of the imidazole ring in the histidine residue.[74,77]

It is well known that histidine itself can give mixed coordination in its binary Cu(II) complexes. Histidine behaves in an 'ambidentate' fashion where it coordinates to Cu(II) through its amino N and imidazole N donor atoms in a 'histamine-like' manner or through its amino N and carboxyl O in a 'glycine-like' manner. Histidine coordination stabilizes the +2 oxidation state of

Cu(II). For example, when solutions of cystein and Cu(II) are mixed, the Cu(II) gets reduced to Cu(I) with the precipitation of cystine. This does not happen in presence of histidine in the reaction mixture at alkaline pH values where it forms ternary complexes.[12]

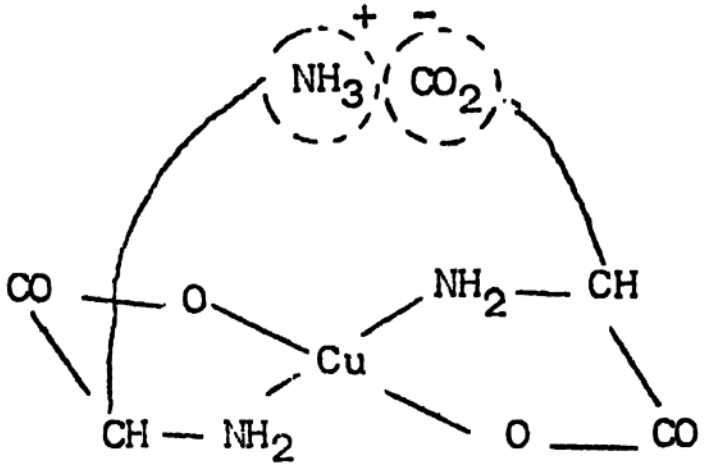
Investigations are being carried out to establish correlation between the stability and the intramolecular ligand-ligand interactions. This interest has been mainly due tc two observations (i) in enzyme-metal-subrate (EMS) complexes formed in enzymatic reactions involving metal ions, non-covalent interaction between enzyme and substrate molecules around the central metal ion are essential for efficiency and . . specificity of the reactions.[72] (ii) in human serum, three histidine-containing ternary complexes are formed in preference to other ternary complexes. On the basis of experimental evidence, electrostatic interactions have been inferred to exist between the oppositely charged side chains of the amino acids in Cu(A)(B), where A and B refer to acidic and basic amino acids respectively. For such systems both cis and trans isomers are observed (13 and 14). Partial optical resolution of racemic amino acids DL-A and DL-B via the ternary Cu(II) complex formation with L-B and L-A respectively, has shown that the D-enantiomers of the racemic amino



Cu(D-A)(L-B)

CIS

13



Cu(L-A)(L-B)

TRANS

14

acids are preferably incorporated into the complexes with L-enantiomers supporting the stereo selectivity due to ligand-ligand interactions.[78,79] An intramolecular interaction like the one discussed above can considerably enhance the stability of ternary complexes.

1.6 Copper Amino Acid Non-amino Acid Type Ternary Complexes

The copper(II) 2,2'-bipyridyl-glycine system is one among the well studied complexes in this group. Stability enhancement on mixed chelation is observed by potentiometric studies.[80,81] It is found that $Cu(BPY)^{2+}$ complexes show preferential binding abilities towards incoming donor atoms on mixed complex formation. It binds ligands with oxygen donors more firmly than those with nitrogen donors. For 'mixed' N/O ligands like glycinates the stability is midway between those observed for pure O and N ligands.[13] Several explanations have been given for this. The increased stability is closely related to the aromatic system of the N ligand. The same conclusions are also valid for peptide and bipyridyl ligands in the Cu(II) in ternary systems. The influence of side chain bulky groups of the peptide on the stability of ternary complexes of (Bpy) and Cu(II) peptide ternary complexes is the same as in the case of binary Cu(II) peptide complexes. In the case of ternary BPY Cu peptide complexes only two coordination positions are available for the peptide to bind. At low pH, the amino nitrogen and the amide oxygen coordinate to Cu(II). This structure has more stability than the corresponding binary peptide complex. On raising the pH to about 8, the deprotonated amide nitrogen coordinates along with the amino nitrogen.

1.7 Copper-Amino Acid-Dipeptide Type Ternary Complexes

Investigations on mixed ligand complexes of Cu(II), containing peptides as a ligand are of considerable interest because in proteins, the deprotonated peptide group is one of the binding group for Cu(II). Sarkar and co-workers[20,31,53,65,82] reported detailed studies involving binary and mixed ligand complexes of peptides and amino acids. Nair and co-workers[77] studied the histidine Cu(II) dipeptide ternary system in perchlorate media and observed three complexes of stoichiometry, [CuABH], [CuAB], and [$CuABH_{-1}$], in addition to the usual binary complexes (A = histidine, B = dipeptide). In [CuABH] species, the proton is attached to the amino group of the histidine residue. At pH values about 8.0, the predominant species is [$CuABH_{-1}$] species, which accommodate over 60% of the total copper concentration. In this species, the dipeptide (B) acts as a bidentate ligand, coordinating copper through the deprotonated amide nitrogen and the amino nitrogen, while the histidine acts as a terdentate ligand. The high stability of these mixed ligand complexes, is attributed to the π-acceptor property of the imidazole ring of the histidine. An alkyl substituent on the glycine-end of the dipeptide has no influence on the stability of [CuAB] species and their formation depends solely on

the basicity of the terminal amino group. But such a substituent on the glycyl residue decreases their stability due to steric effects.[77]

The results outlined so far are indicative of the vast amount of work done and the interest in the Cu(II) amino acid and peptide complexes. However, the redox properties of these systems have not been investigated systematically except for a few polarographic studies on some of the complexes. The usefulness of any model systems depends partially on its redox properties and hence it was decided to investigate systematically this property for the amino acid, dipeptide and mixed amino acid copper(II) complexes. The technique used for this purpose is cyclic voltammetry (CV). Since this is a relatively new technique, a brief introduction to the principles of CV will be included in the following section. The subsequent chapters will discuss the cyclic voltammetric results on the reduction of Cu(II) amino acid, dipeptide, and mixed amino acid complexes.

1.8 Cyclic Voltammetry

Cyclic voltammetry (CV) is the most versatile technique for the study of electroactive species. Its versatility combined with the ease of measurement

has been primarily responsible for its extensive use in electrochemistry, inorganic chemistry, organic chemistry and biochemistry. It has been successfully used in the study of biosynthetic reaction pathways,[83] in the study of electrochemically generated free radicals,[84] in the evaluation of effects of ligands on the oxidation/reduction potential of the central metal ion in complexes and multinuclear clusters, in electron transfer process in inorganic complexes,[85] in the study of systems used in solar energy conversion,[86] and in model studies of enzymatic catalysis.[87] In addition, electrochemical methodology has been used as a novel means of introducing functional groups and removing blocking agents[88] and in the selection of proper oxidising agents to put the metal complex in an intermediate oxidation state.[89] The effectiveness of this technique results from its capability for rapidly observing the redox behaviour over a wide potential range. In spite of its wide usage and the availability of several texts that deal with its theory and practice,[89-95] this technique is not well understood in comparison with other instrumental methods. Since this technique has been extensively used in the investigations summarised in this dissertation, a very brief discussion on the principles of CV will be presented here.

A. Fundamental Principles

The theory and principles involved in linear and cyclic voltammetry have been mathematically worked out by Nicholson and Shain through a series of publications.[96-98] Subsequently the scope of the treatment has been widened by several group of workers,[98-105] to include the large number of electron-transfer reactions. The attempt here will be confined to present a very simple, non-mathematical treatment of the basic principles of cyclic voltammetry.

CV consists of cycling the potential of an electrode, which is immersed in an unstirred solution, and measuring the resulting current. The potential of this working electrode (WE) is controlled versus a reference electrode such as a saturated calomel electrode (SCE) or a silver/silver chloride electrode (Ag/AgCl). The controlling potential applied across these two electrodes can be considered, an excitation signal. This excitation signal for CV is a linear potential scan with a triangular wave form and is shown in Fig.1.1. This triangular potential excitation signal sweeps the potential of the electrode between two values called the switching potentials. The excitation signal shown in Fig.1.1 causes the potential first to scan negatively from +0.80 to -0.20 V at which

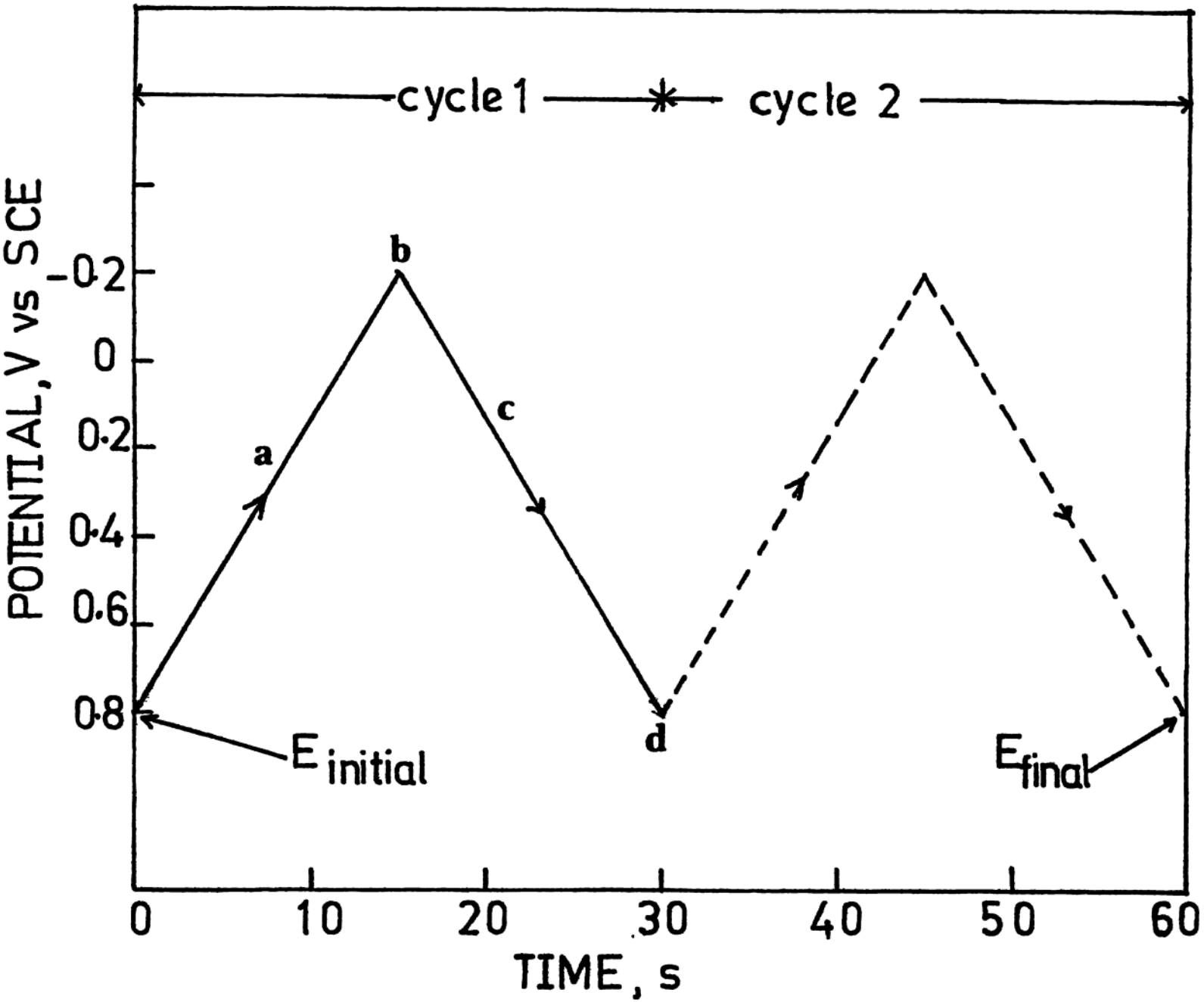


Fig.1.1 Typical excitation signal for cyclic voltammetry - a triangular potential wave form with switching potentials at 0.8 V and -0.2 V versus SCE.

point the scan direction is reversed, resulting in a positive scan, back to the original potential of +0.80 V. The second cycle is indicated by the dashed line. Single or multiple cycles are used in experiments. Often there is very little difference in the current response between the first and successive scans. However, changes that appear on repetitive cycles provide important informations about reaction mechanisms. In linear voltammetry, potential scan, only in one direction is accomplished. Presently available instruments have the facility for varying switching potentials and scan rates.

A cyclic voltammogram is obtained by measuring (plotting) the current at the working electrode during the potential scan against the applied potential thus displaying current (vertical axis) versus potential (horizontal axis). Because the potential varies linearly with time, the horizontal axis can also be considered as a time axis. A typical cyclic voltammogram is shown in Fig.1.2, for $K_3Fe(CN)_6$ (recorded with a platinum working electrode and KNO_3 as supporting electrolyte). In Fig.1.2 the forward scan starts at a potential +0.8 V (a) and at about + 0.3 V (b) a cathodic current is indicated. This is due to the electrode process,

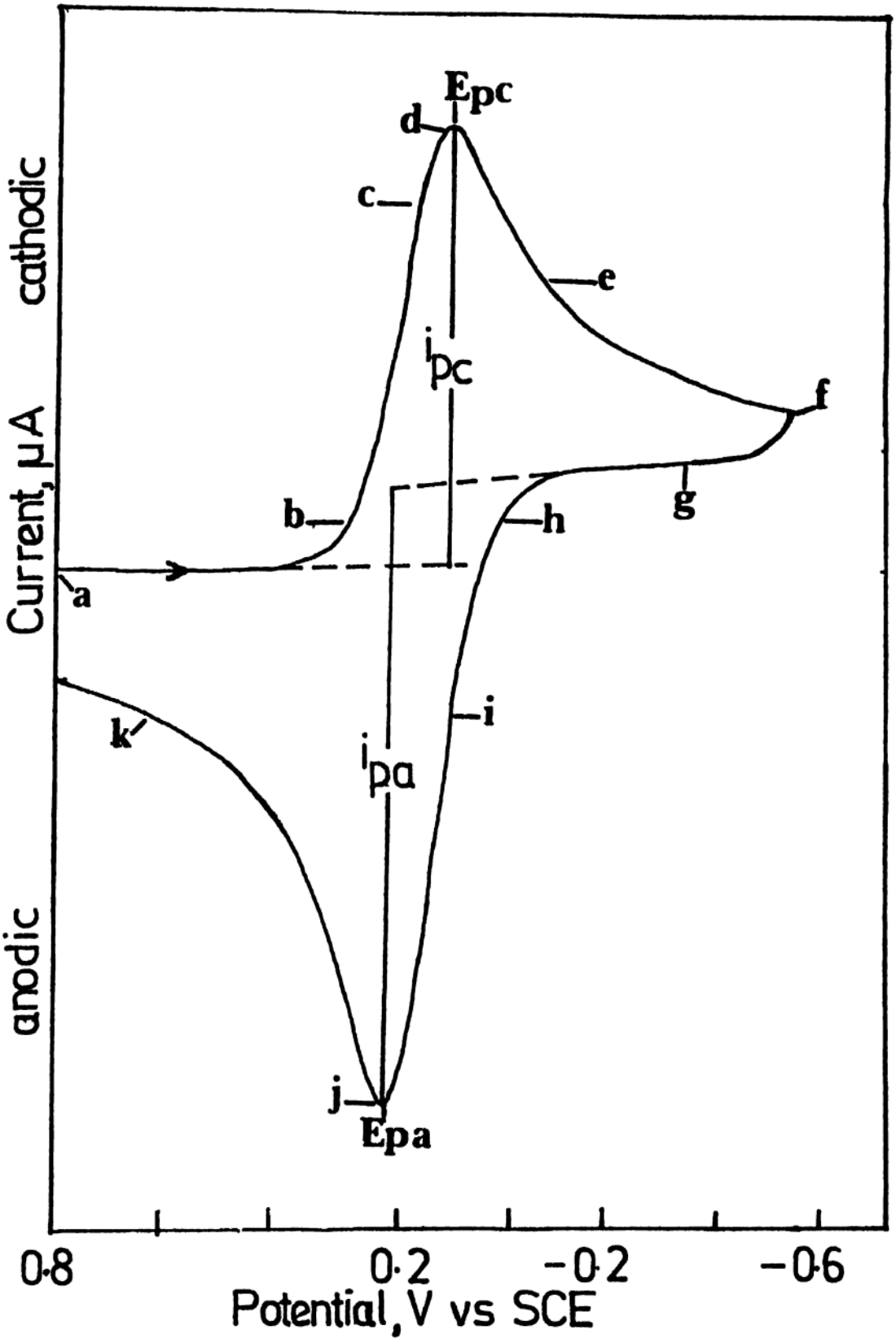


Fig.1.2 Cyclic voltammogram of $K_3Fe(CN)_6$ in KNO_3.
Scan rate 0.05 Vs^{-1} at platinum-disc electrode.

$$Fe^{III}(CN)_6^{3-} + e \longrightarrow Fe^{II}(CN)_6^{4-} \qquad \ldots 1.4$$

This means that the electrode is now a sufficiently strong reductant to reduce $Fe(III)(CN)_6^{3-}$. The cathodic current increases rapidly (b $\longrightarrow$ d) until the concentration of $Fe(III)(CN)_6^{3-}$ at the electrode surface is considerably decreased, causing the current to peak (d). The current then decays (d $\longrightarrow$ f) as the solution surrounding the electrode is depleted of $Fe(III)(CN)_6^{3-}$ due to its electrolytic conversion to $Fe(II)(CN)_6^{4-}$. The scan direction is switched to positive at $\sim$ -0.50 V (f) for the reverse scan. At about (h) the electrode becomes an oxidant and the process,

$$Fe^{II}(CN)_6^{4-} \longrightarrow Fe^{III}(CN)_6^{3-} + e \qquad \ldots 1.5$$

follows generating anodic current (i $\longrightarrow$ k). The anodic current increases rapidly until the surface concentration of $Fe(II)(CN)_6^{4-}$ is decreased causing the current to peak (j). The current then decays (j $\longrightarrow$ k) as the solution surrounding the electrode is depleted of $Fe(II)(CN)_6^{4-}$. The first cycle is completed when the potential reaches + 0.80 V.

The Nernst equation for the reversible $Fe(III)(CN)_6^{3-}$ / $Fe(II)(CN)_6^{4-}$ system is of the form,

$$E = E^{o'} + \frac{0.059}{1} \log \frac{[Fe^{III}(CN)_6^{3-}]}{[Fe^{II}(CN)_6^{4-}]} \quad \ldots 1.6$$

where $E^{o'}$ is the formal reduction potential of the couple and 1 corresponds to the number of electrons involved. An initial value of E which is sufficiently positive of $E^{o'}$ maintains a ratio in which $Fe(III)(CN)_6^{3-}$ predominates. As E is scanned negatively, conversion of $Fe(III)(CN)_6^{3-}$ to $Fe(II)(CN)_6^{4-}$ by reduction is necessary for satisfaction of the Nernst equation.

B. Formal Reduction Potential

The important parameters of a cyclic voltammogram are the magnitudes of anodic peak current (i_{pa}), cathodic peak current (i_{pc}), the anodic peak potential (E_{pa}) and the cathodic peak potential (E_{pc}). These parameters are shown in Fig.1.2. A redox couple in which both species rapidly exchange electrons with the working electrode is an electrochemically reversible couple. The formal reduction potential ($E^{o'}$) for a reversible couple is centred between E_{pa} and E_{pc} and can be expressed as

$$E^{o'} = \frac{E_{pa} + E_{pc}}{2} \quad \ldots 1.7$$

The number of electrons(n) transferred in the electrode reaction for a reversible couple can be determined from the separation between the peak potentials.

$$\triangle E_p = E_{pa} - E_{pc} \simeq \frac{0.059}{n} V \qquad \ldots 1.8$$

Thus for a one-electron process, $\triangle E_p = 0.059$ V. This relationship can be used to evaluate n.

By reversible, it is meant that the reaction is fast enough to maintain the concentrations of the oxidised and reduced forms in equilibrium with each other at the electrode surface. The proper equilibrium ratio at a given potential is determined by the Nernst equation,

$$E = E^{o'} + \frac{RT}{nF} \ln \left[\frac{[O]}{[R]}\right] \qquad \ldots 1.9$$

where O is the oxidised form and R is the reduced form.

Another characteristic of the reversible systems is the dependence of the peak heights on the square root of the scan rate, described by the Randles-Sevcik equation[96,106]

$$i_p = (2.69 \times 10^5) n^{3/2} A D^{1/2} C \nu^{1/2} \qquad \ldots 1.10$$

where i_p is peak current in amperes, n is the number of electrons, electrode area A is in cm^2, D is diffusion coefficient in cm^2/sec, C is the bulk concentration in mol/cm^3 and ν is the scan rate in V/sec. Hence i_p

increases with $\nu^{1/2}$ and is directly proportional to concentration. The values of i_{pa} and i_{pc} should be identical for a simple reversible couple. That is,

$$\frac{i_{pa}}{i_{pc}} = 1 \qquad \ldots 1.11$$

However, the ratio of peak currents can be significantly influenced by chemical reactions coupled to the electrode process.[91]

In some cases the broad point at the top of the current peak makes it difficult to determine the exact position of the peak. In these, it is possible to read the potential where the current is half the value of the peak current. This half-peak potential is related to the polarographic $E_{1/2}$ value by the relation,

$$E_{p/2} = E_{1/2} \pm \frac{28.0}{n} \text{ mV} \qquad \ldots 1.12$$

the sign is positive for a reduction process.

The current depends on two steps in the overall process the movement of electroactive material to the electrode surface and the electron transfer reaction. The electron transfer rate constant for a reduction process is a function of potential and can be described by the relation,

$$k_f = k^o \exp\left[\frac{-\alpha n F}{RT}(E-E^{o'})\right] \qquad \ldots 1.13$$

where k^o (cm/sec) is the standard heterogeneous electron-transfer rate constant. Its value depends on the reaction between the particular compound and the electrode surface used. The number of electrons transferred per molecule is n, F is the Faraday, R is the gas content, T is the Kelvin temperature, $E^{o'}$ is the formal reduction potential, and α is the transfer coefficient. The term α arises because, only a fraction of the energy that is put into the system (as applied potential) lowers the activation energy barrier. Its value varies from zero to unity, often is$\sim$ 0.5.

It has been mentioned already that for a reversible system, the reaction is fast enough at the electrode surface. Many systems look reversible when the voltage is scanned slowly but at higher scan rates $\triangle E_p$ appears greater than $\frac{58}{n}$ mV. It has been pointed out that any deviations from reversible behaviour will be imperceptible if the value of k^o is greater than the numerical value of $0.3\,\nu^{1/2}$. Therefore electron-transfer reactions with rate constants greater than 10 cm/sec will be reversible even in very fast experiments.

Redox couples where the peaks shift farther apart with increasing scan rate is classified as quasi-reversible systems. Also the peak current for a quasi-reversible system is not proportional to $\upsilon^{1/2}$ except when the peaks are widely separated that the system behaves as totally irreversible. In such totally irreversible cases, the peaks are so widely separated ($k^{o} \lesssim 2 \times 10^{5}\, \upsilon^{1/2}$ cm/sec) that no parts of the two peaks overlap on the potential axis. Slow electron-transfer at the electrode surface is responsible for the increase in peak separation. There are also reactions that yield products that cannot be recycled electrochemically to give back the original reactants (due to bond breaking, loss of substituents to solution etc.). Such chemically irreversible reactions have no return peak. For irreversible systems, the equations 1.7, 1.8, 1.10, 1.11 and 1.12 are not applicable.

C. Coupled Chemical Reactions

Most of the metal complexes, inorganic ions, and a few organic compounds undergo electron-transfer reactions without making or breaking covalent bonds. However, a large number of electrochemical reactions involve an electron-transfer step which leads to a species which rapidly reacts with components of the medium through

chemical reactions. CV has turned out to be a powerful technique for the qualitative diagnosis of these homogeneous chemical reactions that are coupled to the electrode reactions. There are several types of reactions involving coupled chemical reactions and are discussed in detail elsewhere.[91,96,99-103]

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CHAPTER 2

REDUCTION OF SOME COPPER(II) AMINO ACID COMPLEXES. ELECTROCHEMICAL IDENTIFICATION OF INTERMEDIATE COPPER(I) AMINO ACID COMPLEXES*

2.1 Abstract

The reduction of Cu(II) amino acid complexes is studied by cyclic voltammetry in aqueous media. The first step is a one-electron reduction of the starting complex generating the corresponding Cu(I) complexes. A fraction of the Cu(I) species is re-oxidised to the copper(II) complexes. The remaining fraction of the Cu(I) complexes undergo chemical reactions generating Cu^{o} at

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*Part of the work discussed in this chapter is submitted for publication in Polyhedron.

the mercury electrode. The transient existence of Cu(I) amino acid complexes is established by trapping this short-lived intermediate Cu(I) species using bathocuproin reagent. The generated Cu^{o} undergoes a two-electron oxidation to Cu(II) during the reverse scan at less cathodic potentials which subsequently gets reduced to Cu^{o} during the second scan. The observed CV pattern corresponds to an ECE mechanism for the electrode process in which the E_2^o is anodic to E_1^o and $n_1 = 1$ while $n_2 = 2$, both the electron-transfer steps being nearly reversible. The pH dependence of the CV response of the title complexes showed the generation of different protonated species. The entire process of protonation is found to be reversible.

2.2 Introduction

The involvement of the transition metal ion, Cu(II), in many of the biological redox processes is well established.[1-3] In many such systems it is found that Cu(II) is bound by one or more histidine nitrogens in a distorted tetrahedral structure. In certain blue copper proteins, for example, Rhus Vernicifera Laccase, the type 2 copper(II) is found to undergo a one-electron reduction at a potential about, 0.4 V versus NHE.[4] Type 2 copper(II) has a square-planar geometry and exhibit considerable

variability in redox behaviour. The ESR and optical parameters of these are very similar to that of simple copper(II) amino acid complexes. It has also been suggested that Cu(II) transport between blood and the tissues is mediated by amino acids.[5] The isolation of Cu(II) amino acid complexes from normal human serum has further strengthened this hypothesis.[6]

The exact mechanism by which the Cu(II) fulfills its task in redox proteins and the mechanism of Cu(II) transport to tissues is not yet clear. Several workers have studied the different aspects of Cu(II) amino acid complexes by spectral[7,8] analytical[9,10] structural[11-15] and other methods.[16-19] In spite of the large amount of informations provided by X-ray analysis, there still exists some uncertainty as to which of the coordination sites of potentially terdentate ligands are involved in the binding of Cu(II) ions in solution. In bidentate amino acids, the 1:1 (CuA) and 1:2 (CuA_2) type complexes are formed depending on the pH conditions and the metal-to-ligand ratio. It is generally agreed that at near neutral pH values the CuA_2 type complexes are formed by the coordination of amino nitrogen and carboxyl oxygen of the two coordinated amino acids in a cis conformation in most cases.[15] But in the case of histidine containing Cu(II) complexes, the coordination pattern is different

(Ch. 1)

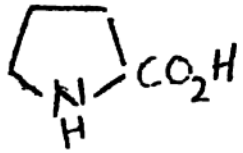
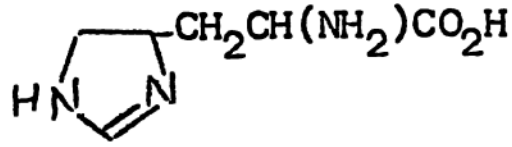
Even though the involvement of Cu(II) in biological redox processes is established, there is hardly any report on the redox behaviour of low molecular weight model complexes of amino acids. This chapter presents our results on the redox behaviour of Cu(II) amino acid complexes studied by cyclic voltammetry.

2.3 Results and Discussion

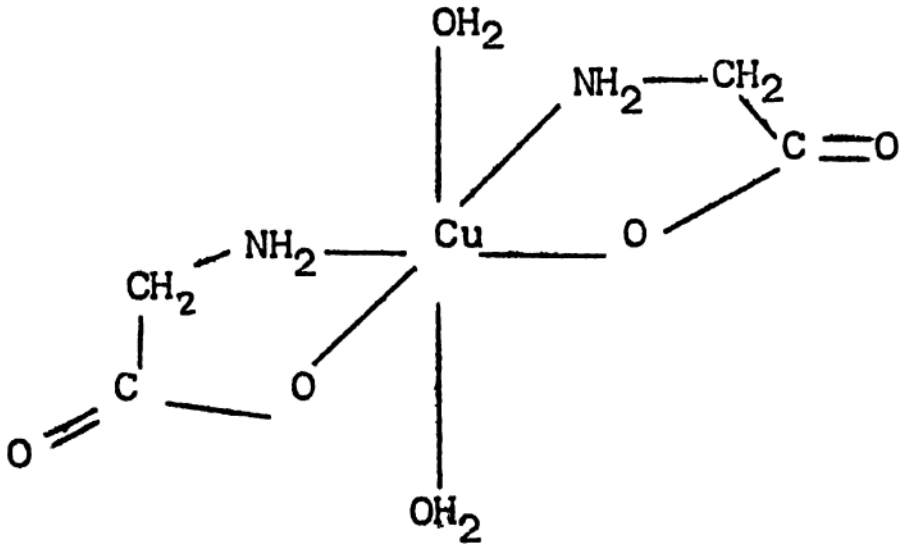
A. Synthesis and Characterization of Complexes

The copper(II) amino acid complexes were prepared by a general procedure, the details of which are given in the experimental section. The amino acids used in the preparation of the complexes, their structure, the abbreviations used in the present discussion and the abbreviation for the complexes are given in Table 2.1. In most cases the complexes are represented by the more common form of notation, for example, $Cu(Gly)_2$. However, whenever there is a need to stress on the oxidation state of the metal ion, another notation, for example $Cu(II)Gly_2$, is used. The complexes were characterized by metal, CHN analysis and spectral methods. Their IR and visible spectra were compared with those reported for Cu(II) amino acid complexes. From these analytical results the identity

Table 2.1. List of amino acids used in the present study and their copper(II) complexes.

Name	Systematic name	Formula	Abbreviation for amino acid	Abbreviation for the copper(II) complex.
Glycine	Amino acetic acid	$CH_2(NH_2)CO_2H$	Gly	$Cu(Gly)_2$
Alanine	α-Amino propionic acid	$CH_3CH(NH_2)CO_2H$	Ala	$Cu(Ala)_2$
Isoleucine	α-Amino-β-methyl-n-valeric acid	$CH_3CH_2CH(CH_3)CH(NH_2)CO_2H$	Ile	$Cu(Ile)_2$
Serine	α-Amino-β-hydroxy-propionic acid	$HOCH_2CH(NH_2)CO_2H$	Ser	$Cu(Ser)_2$
Proline	Pyrrolidine-α-carboxylic acid		Pro	$Cu(Pro)_2$
Ornithine	α,δ-Diamino-n-valeric acid	$H_2NCH_2CH_2CH_2CH(NH_2)CO_2H$	Orn	$Cu(Orn)_2$
Arginine	α-Amino-δ-guanidino-n-valeric acid	$NH = C(NH_2)NH(CH_2)_3CH(NH_2)CO_2H$	Arg	$Cu(Arg)_2$
Lysine	α,ε-Diaminocaproic acid	$H_2N(CH_2)_4CH(NH_2)CO_2H$		
Histidine	α-Amino-β-imidazole propionic acid		His	$Cu(His)_2$

of the complexes were established. The amino acid complexes have the general composition, $Cu(II)A_2.nH_2O$, where n = 1 or 2 depending on the complex and A denotes the amino acid anion. The complexes are neutral and the number shown on the metal ion is only to indicate its oxidation state. The general bonding pattern of these amino acids with respect to glycine is shown in structure 2.1 for ready reference.



2.1

B. Cyclic Voltammetric Response at Neutral pH

The cyclic voltammograms (CV) of the complexes were recorded in aqueous solution in the potential range +0.1 to -0.6 V versus SCE at 25°C under nitrogen atmosphere.

The concentration of the complexes were in the range of 1 mM. Purified $NaClO_4$ (0.1 M) was used as the supporting electrolyte. The dissolution pH of the complexes were about 7.5 and were not adjusted for carrying out the cyclic voltammetric experiments unless specified. Hanging Mercury Drop Electrode (HMDE) of definite area was used as the working electrode. Details of the experimental set up is given in the experimental section.

Cyclic voltammetric behaviour of all these complexes are very similar. Hence the CV response of $Cu(Gly)_2$ is taken as a representative case to discuss their behaviour. Its cyclic voltammogram at scan rate, 0.02 Vs^{-1}, is shown in Fig.2.1 and the voltammograms at various scan rates are shown in Fig.2.2. Cyclic voltammetric data for all complexes at various scan rates are tabulated in Table 2.2. In this table E_p,A; E_p,B; E_p,C and E_p,D represent the peak potentials of peaks A, B, C, and D respectively. $(E_p,A + E_p,B)/2$ represents the mid-potential of A-B couple, $E^{o'}$,C-D represents the formal electrode potential of C-D couple and $\Delta E_{p,C-D}$ denotes the separation between the peaks C and D. There are no peaks in the positive potential range for the complexes. The ligands in the absence of metal ions do not show any voltammetric peaks in the potential range +1.0 to -1.0 V.

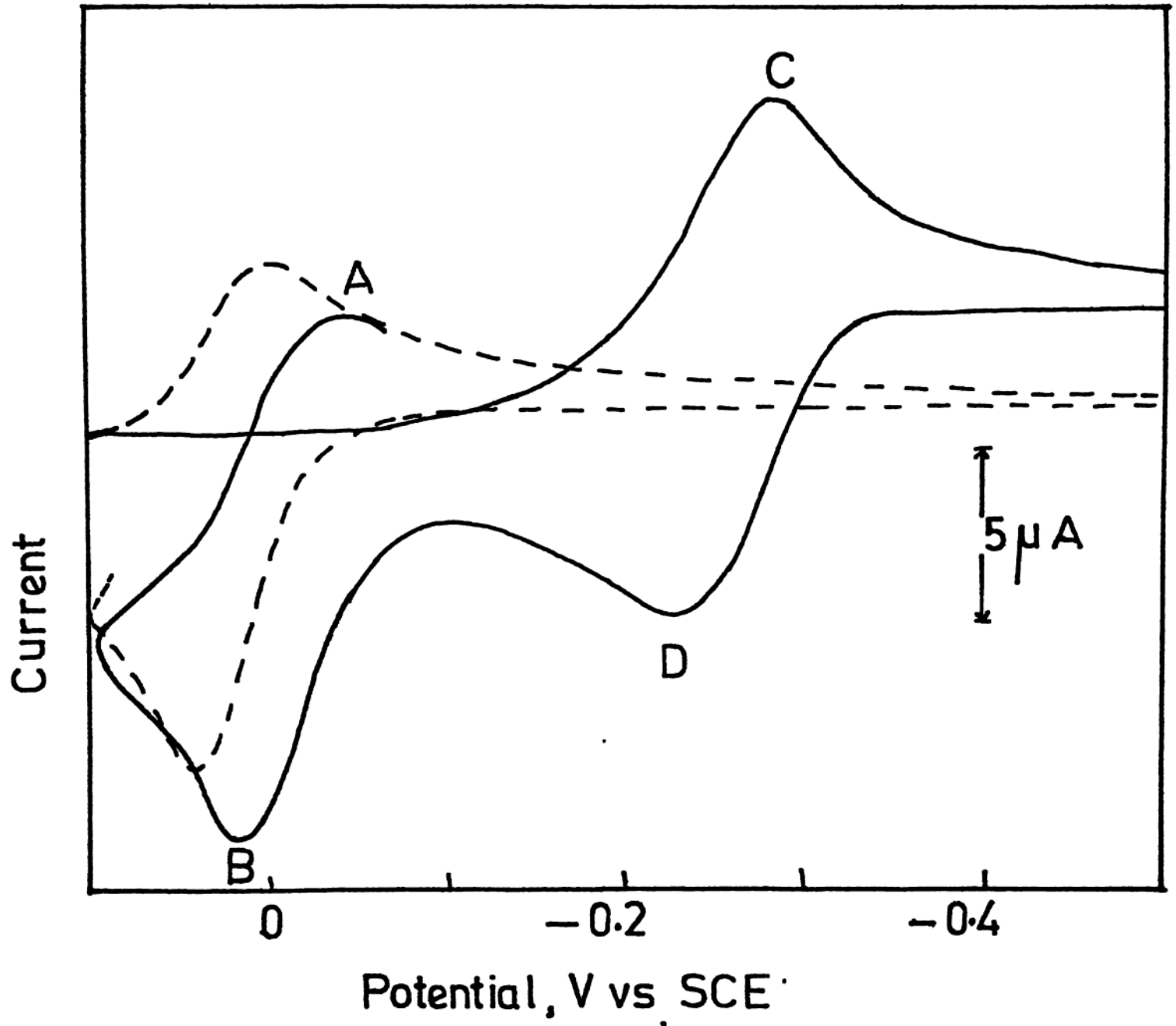


Fig.2.1 Cyclic voltammograms at 0.02 Vs^{-1}; ——, $Cu(Gly)_2$; - - - -, $Cu(ClO_4)_2.6H_2O$

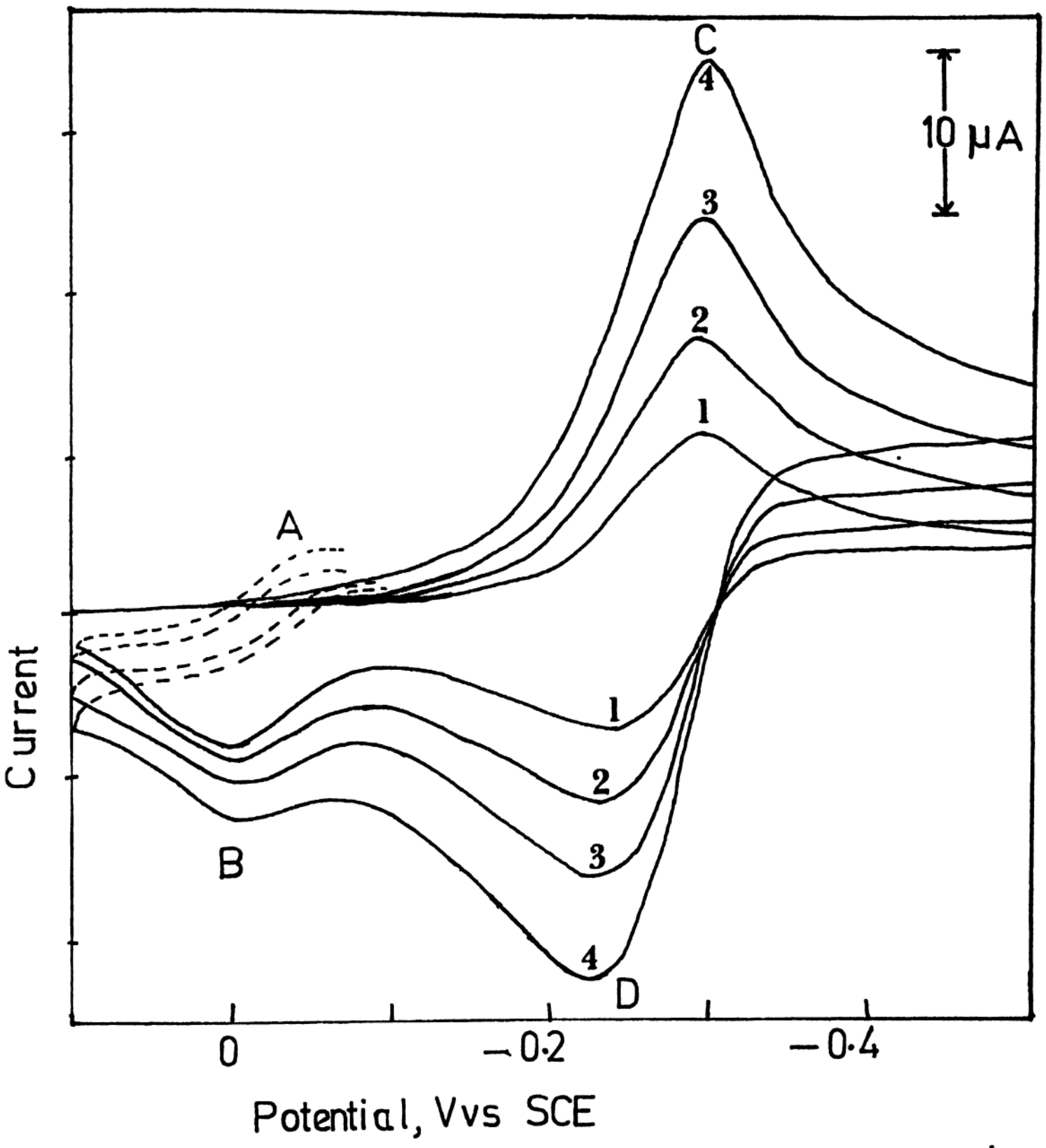


Fig.2.2 Cyclic voltammograms of $Cu(Gly)_2$ at various scan rates. 1, 0.02 Vs^{-1}; 2, 0.05 Vs^{-1}; 3, 0.10 Vs^{-1}; 4, 0.20 Vs^{-1}.

Complex (pH and conc.)	Scan rate (Vs^{-1})	E_p,A[b] (V)	E_p,B (V)	$\frac{E_p,A+E_p,B}{2}$ (V)	E_p,C (V)	E_p,D (V)	$\Delta E_p,C-D$ (V)	$E^{o'}C-D$
$Cu(Gly)_2$	0.02	-0.030	0.020	-0.005	-0.290	-0.230	0.060	-0.260
(pH=7.30;	0.05	-0.030	0.020	-0.005	-0.290	-0.230	0.060	-0.260
c=1.02)	0.08	-0.030	0.020	-0.005	-0.290	-0.230	0.060	-0.260
	0.10	-0.035	0.025	-0.005	-0.290	-0.230	0.060	-0.265
	0.20	-0.040	0.025	-0.007	-0.300	-0.220	0.080	-0.260
$Cu(L-Ala)_2$	0.02	-0.030	0.025	-0.003	-0.280	-0.220	0.060	-0.250
(pH 7.52;	0.05	-0.035	0.025	-0.005	-0.290	-0.220	0.070	-0.255
c=1.09)	0.08	-0.035	0.020	-0.007	-0.290	-0.220	0.070	-0.255
	0.10	-0.040	0.020	-0.010	-0.290	-0.215	0.075	-0.253
	0.20	-[c]	-[c]	-	-0.295	-0.215	0.080	-0.255
$Cu(L-Ile)_2$	0.02	-0.030	0.025	-0.003	-0.270	-0.210	0.060	-0.240
(pH=7.68;	0.05	-0.035	0.025	-0.005	-0.270	-0.200	0.070	-0.235
c=1.00)	0.08	-0.040	0.020	-0.010	-0.225	-0.210	0.065	-0.243
	0.10	-0.040	0.020	-0.010	-0.275	-0.190	0.085	-0.233
	0.20	-[c]	-[c]	-	-0.275	-0.190	0.085	-0.233

contd

$Cu(L\text{-}Pro)_2$	0.02	-0.030	0.045	0.007	-0.295	-0.230	0.065	-0.263
(pH=7.55;	0.05	-0.030	0.045	0.007	-0.300	-0.220	0.080	-0.260
c=1.07)	0.08	-0.030	0.040	0.005	-0.305	-0.220	0.065	-0.263
	0.10	-0.030	0.040	0.005	-0.305	-0.215	0.090	-0.260
	0.20	-[c]	-[c]	-	-0.310	-0.215	0.095	-0.263
$Cu(L\text{-}Ser)_2$	0.02	-0.025	0.035	-0.005	-0.265	-0.200	0.065	-0.233
(pH=7.42;	0.05	-0.030	0.020	-0.005	-0.270	-0.200	0.070	-0.236
c=1.16)	0.08	-0.040	0.020	-0.010	-0.275	-0.200	0.075	-0.236
	0.10	-0.040	0.020	-0.010	-0.275	-0.195	0.080	-0.235
	0.20	-[c]	-[c]	-	-0.275	-0.190	0.085	-0.233
$Cu(L\text{-}Orn)_2$	0.02	-0.040	0.025	-0.007	-0.240	-0.165	0.075	-0.203
(pH=7.71;	0.05	-0.040	0.020	-0.010	-0.245	-0.160	0.085	-0.203
c=1.00)	0.10	-[c]	-[c]	-	-0.245	-0.160	0.085	-0.203
	0.20	-[c]	-[c]	-	-0.245	-0.150	0.095	-0.197
$Cu(L\text{-}Lys)_2$	0.02	-0.040	0.030	-0.005	-0.245	-0.175	0.070	-0.210
(pH=7.74;	0.05	-[c]	0.030	-	-0.245	-0.170	0.075	-0.208
c=1.12)	0.10	-[c]	-[c]	-	-0.250	-0.170	0.080	-0.210

contd.

Table 2.2 contd.

$Cu(L\text{-}Arg)_2$	0.02	-0.040	0.025	-0.007	-0.235	-0.165	0.070	-0.200
(pH=8.30;	0.05	d	d	-	-0.235	-0.150	0.085	-0.193
c= *)	0.10	d	d	-	-0.235	-0.150	0.085	-0.193
	0.20	d	d	-	-0.240	-0.150	0.090	-0.195
$Cu(L\text{-}His)_2$	0.01	-[c]	-[c]	-	-0.400	-0.270	0.130	-0.335
(pH=6.42;	0.02	-[c]	-[c]	-	-0.400	-0.265	0.135	-0.333
c=1.48)	0.05	-[c]	-[c]	-	-0.405	-0.255	0.140	-0.330
	0.10	-[c]	-[c]	-	-0.410	-0.235	0.160	-0.323

a A linear dependence on concentration of peak potentials of peaks C and D is observed below 1 mM concentration of the samples. The origin of this is not clear.

b Peak observed only in the second and subsequent scans.

c Broad peak, difficult to measure precisely.

d Peak not observed.

* Studied in situ (approx. concentration 1 mM).

From an examination of the cyclic voltammograms the following observations are made for discussing the CV results.

(i) During the first forward sweep, only one cathodic peak C is observed at about -0.260 V.

(ii) On the reverse scan a well-defined anodic peak D at about -0.200 V and another anodic peak B at more anodic potentials, about +0.04 V are observed.

(iii) During the second forward scan a new cathodic peak A at about -0.05 V appears.

(iv) An increase in the scan rate decreases the peak heights of A and B. But such an influence is not observed for peaks C and D. From Fig.2.2 this may not be very clear. The reason for this is given later. However this is more clear in Fig.2.3 where $i_p/\upsilon^{1/2}$ value is plotted against scan rate. This observation is true for all other copper(II) amino acid complexes. The $i_p/\upsilon^{1/2}$ values for all complexes are presented in Tables 2.3-2.7.

(v) On holding the potential at a point well past the peak C for a few minutes and on resuming the scan, the peaks A and B increase in heights drastically (Fig.2.4a) while the peaks C and D retrace. The same effect is

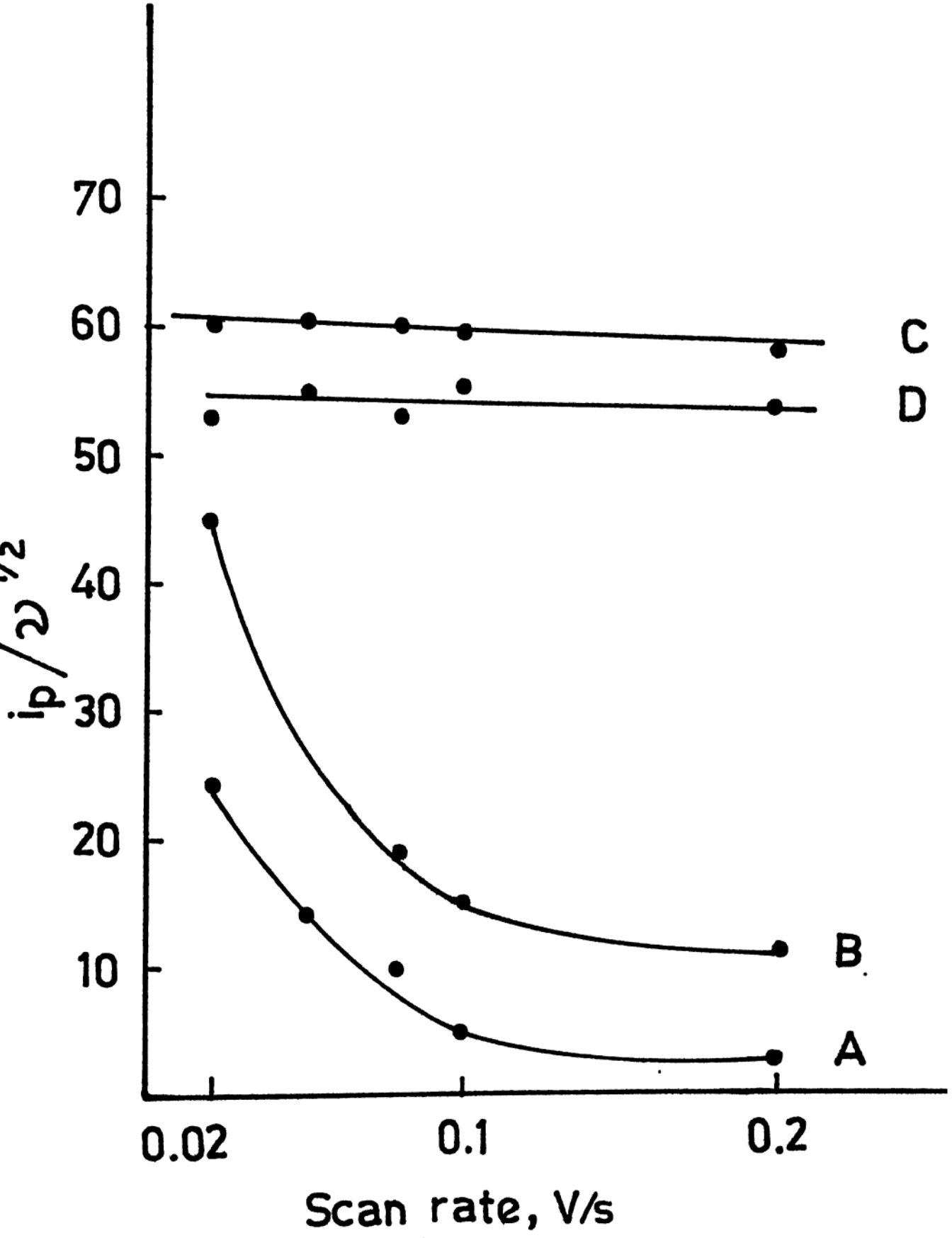


Fig.2.3- Plot of $i_p/\nu^{1/2}$ versus scan rate for various peaks.

A, peak A; B, peak B; C, peak C; D, peak D.

Table 2.3. Current function values at various scan rates.[a] $Cu(Gly)_2$; pH=7.75.

Sweep rate (Vs^{-1})	$i_p,A/\upsilon^{1/2}$[b]	$i_p,B/\upsilon^{1/2}$	$i_p,C/\upsilon^{1/2}$	$i_p,D/\upsilon^{1/2}$	$i_p,D/i_p,C$
0.02	21.21	38.83	81.32	77.78	0.956
0.05	15.65	20.12	78.26	73.79	0.942
0.08	8.84	14.14	81.32	67.18	0.826
0.10	4.74	12.65	79.06	72.73	0.920
0.20	3.35	8.94	76.06	71.55	0.941

a. Current function values are expressed in $\mu A(V/s)^{-1/2}$.

b. Peak observed only in the second scan.

Table 2.4. Current function values at various scan rates[a] $Cu(L\text{-}Ala)_2$ pH=7.48.

Scan rate (Vs^{-1})	$i_p,A/\upsilon^{1/2}$[b]	$i_p,B/\upsilon^{1/2}$	$i_p,C/\upsilon^{1/2}$	$i_p,D/\upsilon^{1/2}$	$i_p,D/i_p,C$
0.02	21.21	53.03	102.53	102.53	1.00
0.05	18.78	35.77	98.38	93.91	0.954
0.08	14.14	28.28	95.45	77.78	0.814
0.10	12.65	25.30	94.86	88.54	0.933
0.20	6.71	8.94	71.55	76.02	1.06

a. Current function values are expressed in $\mu A(V/s)^{-1/2}$.

b. Peak appears only in the second scan.

Table 2.5. Current function values at various scan rates: $Cu(Ser)_2$[a].

Sweep rate (Vs^{-1})	$i_p,A/\nu^{1/2}$[b]	$i_p,B/\nu^{1/2}$	$i_p,C/\nu^{1/2}$	$i_p,D/\nu^{1/2}$	$i_p,D/i_p,C$
0.02	24.74	45 96	60.10	53.03	0.88
0.05	14.31	26.88	60.37	55.90	0.92
0.08	10.23	19.44	60.10	53.03	0.88
0.10	4.74	19.11	60.08	56.92	0.94
0.20	2.68	11.18	58.13	53.66	0.92

a Current function values expressed in $\mu A(V/s)^{-1/2}$.

b Peak appears only in the second scan.

Table 2.6. Current function values at various scan rates.[a] $Cu(Pro)_2$; pH = 7.32.

Sweep rate (Vs^{-1})	$i_p,A/\upsilon^{1/2}$[b]	$i_p,B/\upsilon^{1/2}$	$i_p,C/\upsilon^{1/2}$	$i_p,D/\upsilon^{1/2}$	$i_p,D/i_p,C$
0.02	15.56	42.43	49.50	42.43	0.857
0.05	9.38	22.36	46.96	40.25	0.857
0.08	7.07	15.91	44.19	37.12	0.840
0.10	6.32	8.39	50.60	42.69	0.843
0.20	4.47	4.47	46.96	39.20	0.830

a. Current function values expressed in $\mu A(V/s)^{-1/2}$.

b. Peak observed only in the second scan.

Table 2.7. Current function values at various scan rates[a]
$Cu(Ile)_2$ pH = 7.23.

Scan rate (Vs^{-1})	$i_p,A/\upsilon^{1/2}$[b]	$i_p,B/\upsilon^{1/2}$	$i_p,C/\upsilon^{1/2}$	$i_p,D/\upsilon^{1/2}$	$i_p,D/i_p,C$
0.02	12.37	22.98	28.28	26.51	0.937
0.05	6.71	8.94	27.95	84.59	0.880
0.08	5.30	6.18	28.28	22.98	0.812
0.10	2.37	6.32	28.46	25.29	0.888
0.20	1.12	4.47	27.95	23.47	0.840

a Current function values expressed in $\mu A(V/s)^{-1/2}$.

b Peak observed only in the second scan.

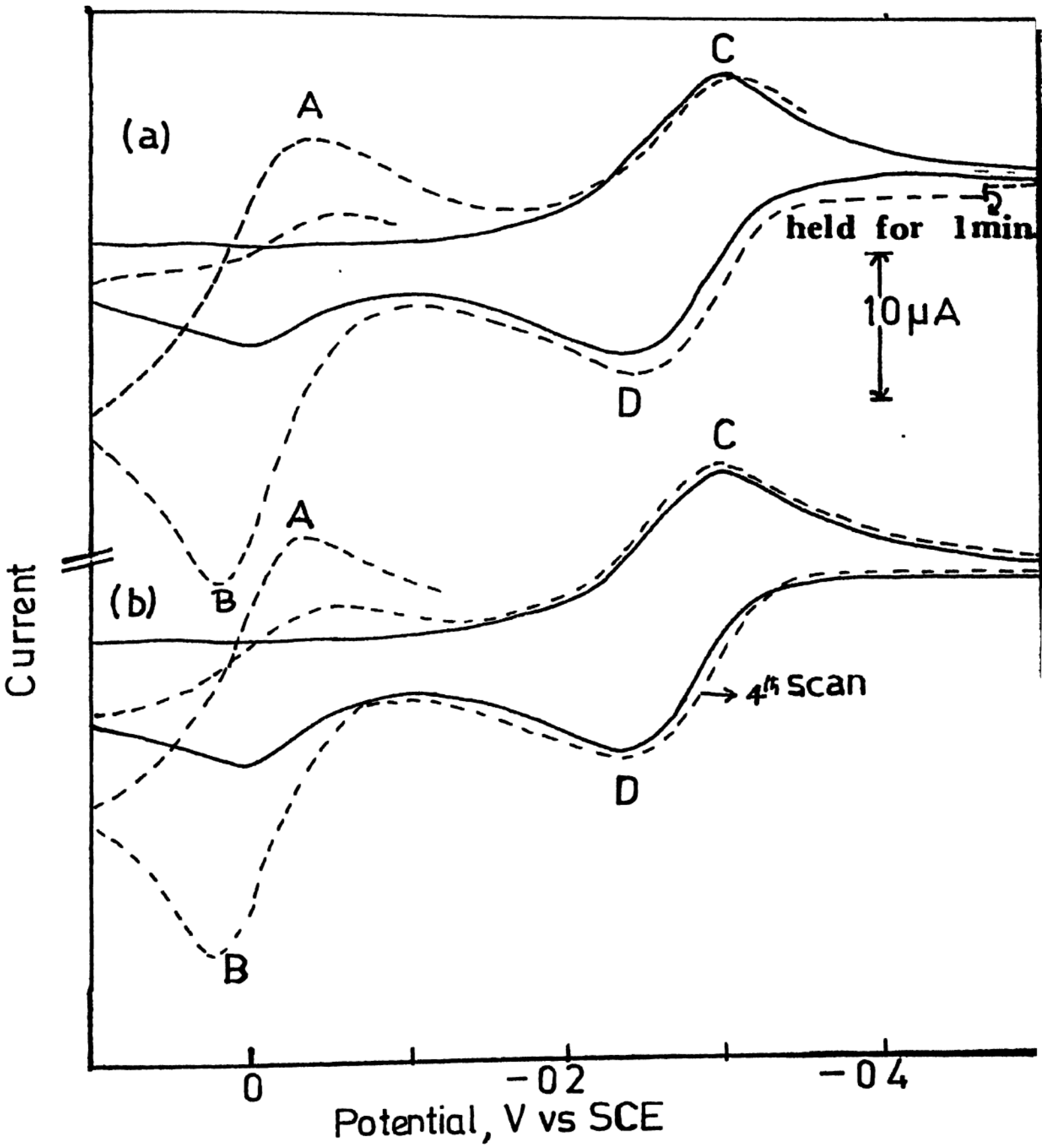


Fig. 2.4. Cyclic voltammograms of $Cu(Gly)_2$ at scan rate 0.02 Vs^{-1}. (a) Effect of holding the potential during scan (b) Effect of multiscan on various peaks: ——, 1st scan; -----, 4th scan.

observed on multiscans in the full potential range as seen in Fig.2.4b.

(vi) If the scan is reversed after peak A in the second and subsequent scans, the peak heights of A and B decrease considerably.

(vii) No voltammetric response is observed if the potential is scanned only upto -0.150 V, that is, before the peak C appears.

Before proceeding to examine the CV results, it appears necessary to briefly comment on the observation that, as the scan rate increases peak heights of A and B decrease while those of C and D remain the same. (cf. Figs.2.2 and 2.3). From Fig.2.2 it appears contrary, i.e., as the scan rate increases the peak heights of C and D increase while those of A and B remain the same. This apparent discrepancy is because of the following reason. For any reversible electron-transfer process, the peak heights increase as the scan rate increases, i.e., $i_p \alpha \nu^{1/2}$. Hence both of C-D couple and A-B couple should have shown increased peak heights as the scan rate increases. Since there is an inherent decrease in peak heights of A and B this cancels the increase in peak heights expected for them.

This results in rather static situation for the A-B peaks. On the other hand for the C-D peaks there is no such nullifying effects to the increase in heights due to increase in scan rates. Hence when $i_p/\nu^{1/2}$ is plotted against scan rate, the actual effect of scan rate on peak heights of various peaks, becomes clear (Fig.2.3).

It is well established that Cu(II) amino acid complexes retain their structure on dissolution in water at neutral pH values.[15,20,21] Therefore no structural change is expected on dissolution of these complexes and the CV recorded in aqueous solution corresponds to the starting $Cu(II)A_2$ species. Then, peak C can arise either due to a one-electron reduction,

$$Cu(II) \xrightarrow{1e} Cu(I) \qquad \ldots 2.1$$

or due to a two-electron reduction,

$$Cu(II) \xrightarrow{2e} Cu^{o} \qquad \ldots 2.2$$

of the starting $Cu(II)A_2$ complex. If the peak C originates due to the process in equation 2.2, it is difficult to explain the appearance of the well-defined anodic peak D about 60 mV anodic to peak C for the following reasons. A two-electron reduction of the Cu(II) species will result in the formation of Cu^{o} which at the mercury electrode

will form amalgam, Cu(Hg). In such a case, on the reverse scan the Cu^{o} will get oxidised at a much less negative potential than the peak D. Even if it is assumed that an undissociated Cu^{o} complex gets re-oxidised giving the peak D, then the peak separation of C and D can be only about 30 mV. This follows from the simple consideration that for a reversible electron-transfer process, the peak separations are of the order 60/n mV where n is the number of electrons involved. On the other hand if peak C is the result of a one-electron reduction of $Cu(II)A_2$ complex, the generated Cu(I) species can undergo re-oxidation giving the peak D. In that case peak separation between C and D should be 60 mV. The ΔE_p,C-D shown in Table 2.2 are in the range 60-80 mV, supporting this assumption. This also shows that the electron-transfer is nearly reversible.

The appearance and other characteristics of peaks B and A indicate the presence of a new redox couple which is dependent on the process responsible for peaks C and D. Observations, (vi) and (vii) bring out the fact that the first electron-transfer is a pre-requisite for the observation of peaks A and B. This together with the observation in (v) shows that as the time scale of the experiment increases, more and more of the species responsible for

peaks A and B are generated. On comparing the peaks A and B with the CV of free Cu(II) ions under identical experimental conditions (Fig.2.1), it can be seen that the A,B peaks correspond to the redox peaks of free Cu(II) ions. It should be pointed out at this stage that there is a few mV difference between the peak positions of free Cu(II) ions and the Cu(II) ions generated electrochemically. Also the ΔE_p,A-B of free Cu(II) ions is about 40 mV while that of the electro-generated Cu(II) ion is about 50 mV. These minor deviations can be rationalized in the following way. In free Cu(II) ions there is no influence of any ligands other than water molecules in the electron-transfer process. However, the free Cu(II) ions generated electrochemically are under the influence (interaction) of amino acid ligands present in solution. These weak interactions can influence the peak potentials marginally, as observed in the present case. For free Cu(II) ions it has been established that the redox couple originates from a reversible two-electron transfer process as in equation 2.3.

$$Cu(II) \underset{}{\overset{2e}{\rightleftharpoons}} Cu^{o} \qquad \ldots\ 2.3$$

This means that peak B is a result of the oxidation of the electrogenerated Cu^{o}. Then the question is how the

Cu^{o} gets generated at the electrode? The possibility is that the Cu^{o} can be the result of the decomposition and/or disproportionation of a fraction of the Cu(I) species. Thus the overall electrode process can be represented by the following sequences.

$$Cu(II)A_2 \xrightleftharpoons{1e} [Cu(I)A_2] \quad \ldots 2.4$$

$$[Cu(I)A_2] \xrightarrow[\text{reactions}]{\text{chemical}} Cu^{o}_{(Hg)} + 2A \quad \ldots 2.5$$

$$Cu^{o}_{(Hg)} \xrightleftharpoons{2e} Cu^{2+}_{(aq)} \quad \ldots 2.6$$

The electrode reaction in equation 2.4 is responsible for the C-D couple while equation 2.6 is responsible for the appearance of the peaks A and B. Equation 2.5 represents the interposed chemical reaction step between the two electron-transfer (equations 2.4 and 2.6) steps. Thus the overall electrode process is of the ECE type.[22] The absence of peak A in the first forward scan shows the absence of free Cu(II) ions in solution to start with. Its presence in the second scan confirms that the peak A appears only after the Cu^{2+} generation through the sequences 2.4 to 2.6.

The equation 2.4 which generates the Cu(I) amino acid complexes represents the most important step in the overall process. Cu(I) complexes are generally stable only in tetrahedral or pseudo-tetrahedral geometries. The Cu(I) species generated electrochemically by the one-electron reduction is expected to have a geometry distorted towards tetrahedral since the parent $Cu(II)A_2$ complexes posses a tetragonally distorted octahedral geometry.[11-15] The extent of stability of the Cu(I) species depends on the degree of tetragonality. The transient existence of Cu(I) complexes demonstrated by the voltammetric pattern shows that it has a life-time long enough to be detected by cyclic voltammetry. It is to be noted that it is for the first time that Cu(I) amino acid complexes is electrochemically detected during the reduction of Cu(II) amino acid complexes.

The process represented in equation 2.5 may be the net result of a few reactions taking place in steps as,

$$Cu(I)A_2 \rightleftharpoons Cu^{1+} + 2A \quad \ldots 2.7$$

$$2\,Cu^{1+} \rightleftharpoons Cu^{o} + Cu^{2+} \quad \ldots 2.8$$

In aqueous solution the disproportionation reaction of Cu(I) ions is quite possible in the absence of other

chelating agents which can stabilise the Cu(I) ions. Under the cathodic potential prevailing in the experimental conditions, the predominant species in equation 2.8 will be Cu^{o} which at the mercury electrode will exist as amalgam.[23] The Cu^{o} thus generated can get oxidised by a two-electron process to Cu(II) ions giving rise to the peak B and the subsequent reduction of Cu(II) ions to Cu^{o} gives rise to the peak A, representing the process in equation 2.6. The current function values $(i_p/\nu^{1/2})$ for the peaks A and B decreases with an increase in scan rate while that of peaks C and D remain constant (Fig.2.3, Tables 2.3-2.7). This indicates that at higher scan rates the chemical reaction represented in equation 2.5 generates only less amounts of Cu^{o} due to lack of time which in turn decreases the $i_p/\nu^{1/2}$ value. But on the other hand, scan rates should not have any effect on peaks C and D and it is found to be so. Giving more time after the generation of Cu(I) species helps to increase the concentration of Cu^{o} which will reflect in the increased peak heights of B and A. The peaks C and D remain unchanged on holding the potential at a point well past peak C while peaks A and B increase considerably. Such a situation can occur only when a steady concentration of the $Cu(I)A_2$ species is maintained at the electrode, irrespective of its decomposition. This means that the

electron-transfer process in equation 2.4 is faster than the chemical reactions in equation 2.5. On the other hand, if the process in equation 2.5 is faster than that in equation 2.4, then with the decrease in scan rate or with the increase in the time-lag for the backward scan, the peak D would have diminished due to the lesser amount of the reduced species, $Cu(I)A_2$ available for re-oxidation.[24]

C. Trapping of the Intermediate Cu(I) Species

Further support to the assumption, that Cu(II) amino acid complexes undergo a reversible one-electron reduction has been obtained by chemically trapping the Cu(I) intermediate and investigating it. It is known that bathocuproin (2,9-dimethyl-4,7-diphenyl-1,10-phenanthroline) is a reagent which is highly specific towards Cu(I) ions in forming stable complexes. The Cu(I) bathocuproin complex has a characteristic high intensity band at 479 nm (ϵ = 14,150 $dm^3\ mol^{-1}\ cm^{-1}$) which can be used for the detection of the complex formation.[25-27]

A controlled potential electrolysis of the Cu(II) amino acid complex solution over mercury pool electrode in the presence of bathocuproin reagent was carried out to trap the Cu(I) species generated by the reduction of

the complex. The experimental details are discussed in the experimental section. As the electrolysis of the well stirred mixture of the complex and the reagent solutions proceeds, the Cu(II) complexes get reduced to Cu(I) species and the reagent forms its Cu(I) complex. This is evident from the increase in the intensity of the colour of the solution. The organic layer containing the reagent and the Cu(I) bathocuproin complex was separated from the colourless aqueous layer after the completion of the electrolysis. This solution gave a strong absorption band at 480 nm. In another experiment the absorption spectrum of the reagent solution was recorded after prolonged agitation of the reagent with the Cu(II) amino acid complex solution without electrolysis and found that the band at 480 nm is absent. On the other hand, the reagent instantaneously forms a deep yellow colour on mixing with the chemically generated (experimental section) Cu(I) ions which also gave a strong absorption band at 480 nm. The CV of the Cu(I) bathocuproin complex generated by electrolytic method, recorded at HMDE (TEAP as supporting electrolyte) compares very well with that of the Cu(I) bathocuproin complex prepared from chemically obtained Cu(I) ions as shown in Fig.2.5. These results conclusively establish the generation of Cu(I) species as an intermediate in the electrode process and proves

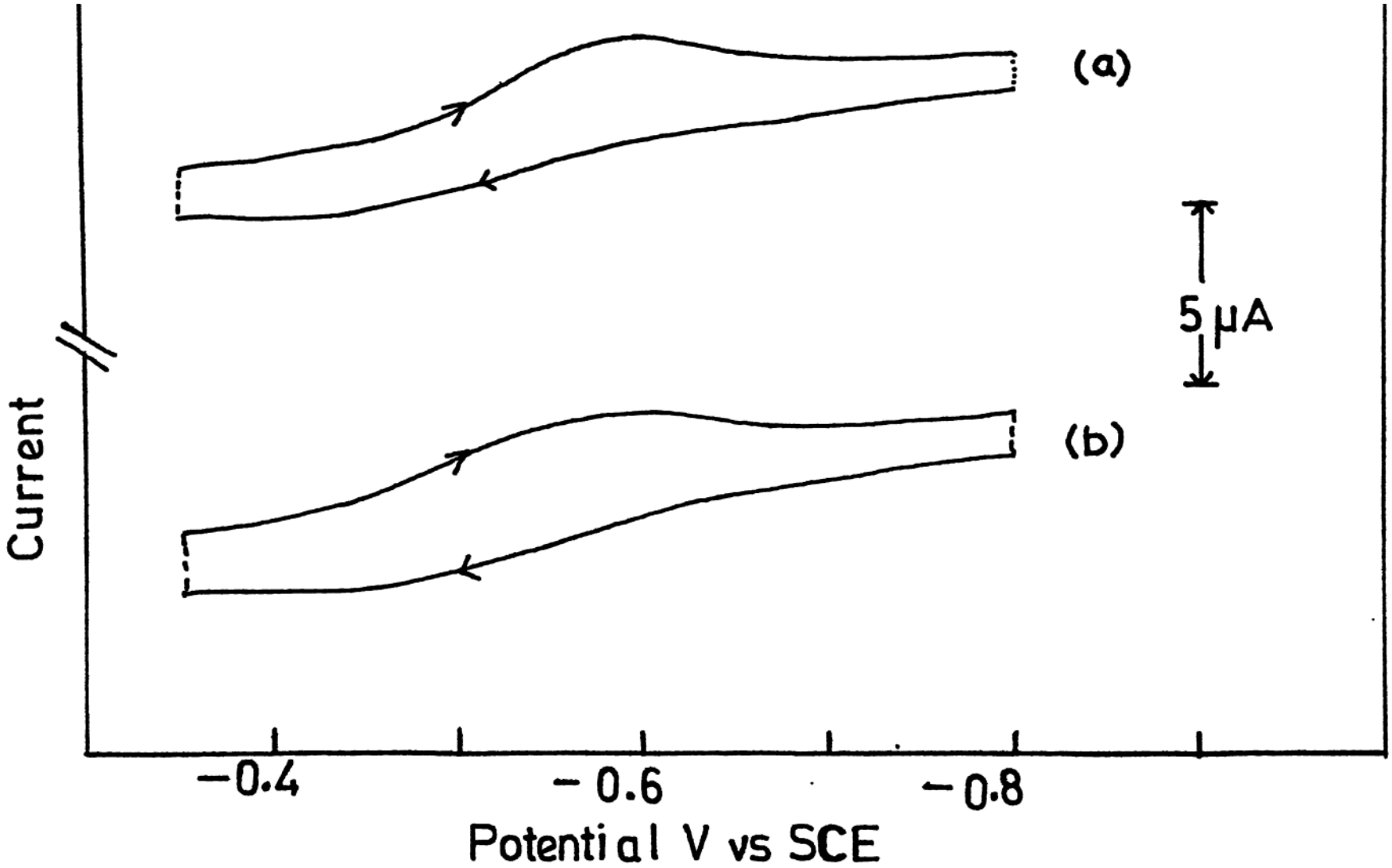


Fig.2.5 Cyclic voltammograms of copper(I) bathocuproin complexes generated (a) chemically from copper(II) ions. (b) electrochemically from copper(II)complexes.

that the first reduction of the $Cu(II)A_2$ complex does proceed by a one-electron process as depicted in equation 2.4.

D. Controlled Potential Coulometry

Generally, controlled potential coulometry (CPC) is used to determine the number of electrons involved in an electron-transfer reaction. Hence this technique was tried to determine the number of electrons(n) involved in the reduction of $Cu(II)A_2$ complexes. CPC was carried out at a potential -0.6 V vs SCE over mercury pool electrode. Details are given in the experimental section. CPC yielded coulombs which corresponded to, n = 2. This value does not agree with the experimental evidence presented earlier for a one-electron reduction. This apparent discrepancy arises because of the following reason. Under the continued influence of the cathodic potential the Cu(I) species generated by the first electron-transfer undergo further reduction to Cu^{o} which immediately forms amalgam with the mercury. Thus the overall process involve two electrons. The absence of Cu(II) ions in the electrolysed solution is confirmed by the disappearance of copper ESR signals and the absorption band during the course of electrolysis.

E. Cyclic Voltammetric Response at Low pH Conditions: Protonation

It is known that Cu(II) amino acid complexes have different modes of coordination in aqueous solution at various pH values. This has been studied by different methods.[7,9,10,16,17] So it is obvious that by decreasing the pH from neutral value, it is possible to generate species other than CuA_2. If this happens, the CV profile discussed in the earlier sections resulting from the reduction of CuA_2 has to change on decreasing the pH. A series of experiments were conducted with a view to study the effect of pH on the CV pattern of the amino acid complexes. Since decrease of pH means protonation of the amino acid moiety, these experiments will be referred to as 'protonation' studies. In this section the results of the studies on the reduction of these amino acid complexes by CV at various pH values will be presented and discussed.

A gradual decrease of the pH of the aqueous solution of the complexes is achieved by the step-wise addition of small amounts (μl) of a protonic acid, $HClO_4$. Cyclic voltammograms were also recorded at various acid pH values using buffer solutions. The buffer solutions used were the following:

for pH 4.0-5.5 acetate buffer; for pH 5.5-7.0 HEPES buffer and for pH 8.0-90 borate buffer. HEPES refers to 2-hydroxy-ethylpiperazine-N'-2-ethane-sulphonic acid, HO-CH_2-Ch_2-N⟨ ⟩N-$CH_2CH_3SO_3^-$. The cyclic voltammograms recorded in the buffer solutions were highly complicated and it is suspected that the presence of buffer anions are responsible for this. Hence the first method of using acid for adjusting the pH was preferred. The addition of acid or base to adjust the pH to generate different species from copper(II) amino acid and peptide complexes is a common practice even in potentiometric studies. It is found that all the Cu(II) amino acid complexes behave in a similar manner. The cyclic voltammetric response of $Cu(Gly)_2$ at different pH conditions is shown in Fig.2.6 as a representative case. The CV data on protonation for all the complexes are given in Tables 2.8-2.12.

The major changes occurring in the CV pattern on protonation are the following. (i) Heights of peaks C and D decrease gradually and finally at low pH values they completely disappear. (ii) Peak A begins to appear in the first scan itself. (iii) In the initial stages of pH decrease, the peak A broadens but on further decrease of pH the broadening disappears. (iv) Peaks A and B increase in heights as pH is decreased.

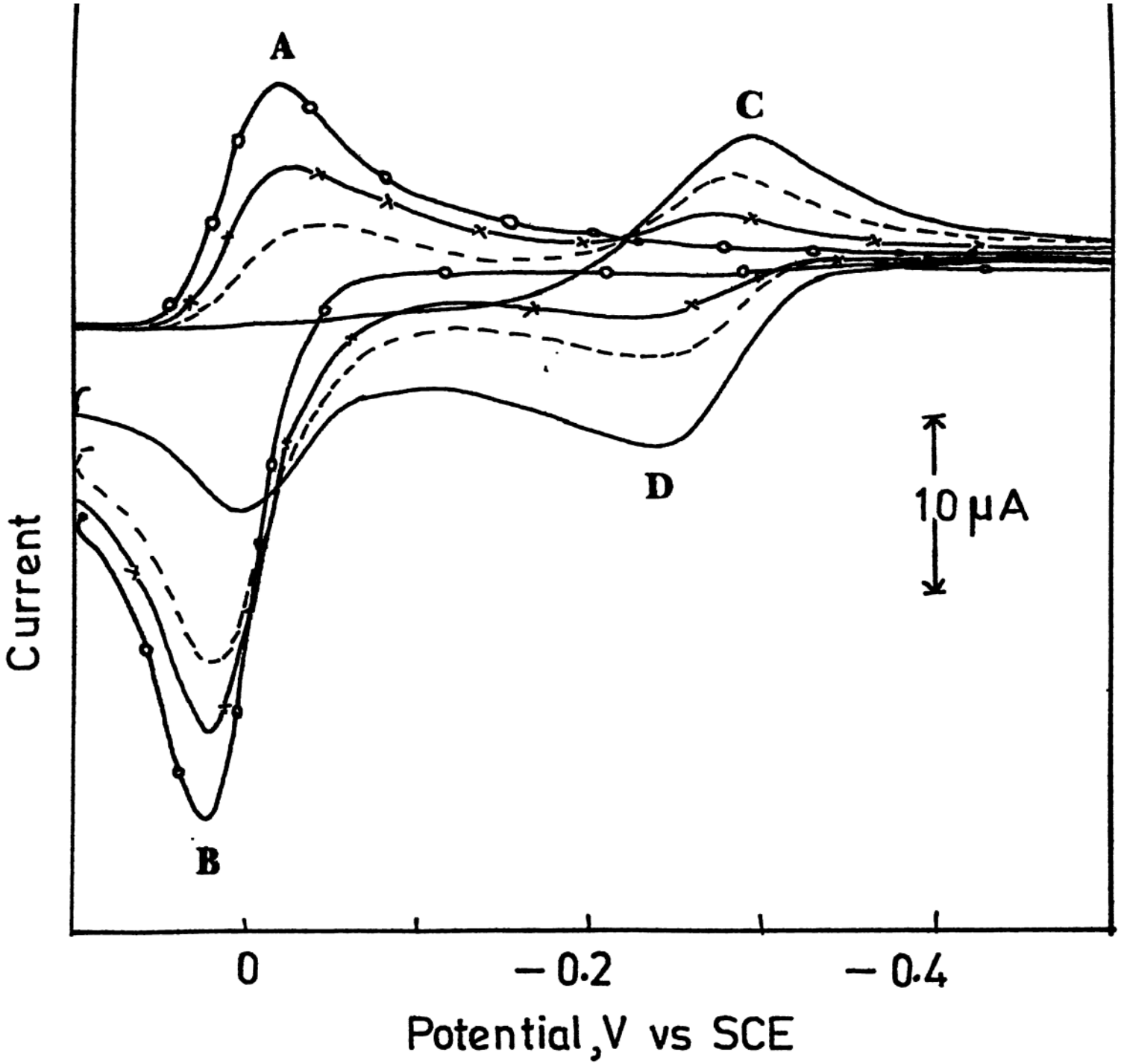


Fig.2.6 Cyclic voltammograms of $Cu(Gly)_2$ at different pH values. ——, 7.25;

ıble 2.8. Cyclic voltammetric data on protonation of $Cu(Gly)_2$[a].

	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	$E^{o'}_{C-D}$	i_p,A/$\upsilon^{1/2}$	i_p,B/$\upsilon^{1/2}$	i_p,C/$\upsilon^{1/2}$	i_p,D/$\upsilon^{1/2}$
	0.02	-[b]	0.020	-0.280	-0.220	-0.250	-[b]	38.89	81.32	77.78
.40	0.05	-[b]	0.020	-0.280	-0.215	-0.248	-[b]	20.12	78.26	73.79
	0.10	-[b]	0.020	-0.285	-0.215	-0.250	-[b]	12.65	79.06	72.73
	0.02	-0.050[c]	0.020	-0.275	-0.220	-0.248	7.07	63.64	74.25	70.71
.25	0.05	-0.060[c]	0.020	-0.280	-0.220	-0.250	8.95	35.78	76.03	71.55
	0.10	-0.050[c]	0.020	-0.280	-0.210	-0.245	9.49	25.30	69.57	69.57
	0.02	-0.050[c]	0.030	-0.275	-0.225	-0.250	16.14	84.85	67.18	60.10
.37	0.05	-0.060[c]	0.025	-0.275	-0.215	-0.245	20.36	49.19	67.08	64.85
	0.10	-0.050[c]	0.025	-0.275	-0.215	-0.245	18.97	37.95	63.26	60.08

contd.

able 2.8 contd.

	0.02	-0.050[c]	0.030	-0.275	-0.220	-0.248	24.82	91.92	63.64	56.57
.81	0.05	-0.050[c]	0.030	-0.275	-0.215	-0.245	26.02	62.61	62.61	53.57
	0.10	-0.050[c]	0.030	-0.275	-0.215	-0.245	25.30	44.27	60.08	56.92
	0.02	-0.035[c]	0.040	-0.270	-0.220	-0.245	42.43	141.42	49.50	38.89
.16	0.05	-0.030[c]	0.040	-0.270	-0.215	-0.243	40.25	93.91	44.72	35.78
	0.10	-0.040[c]	0.040	-0.270	-0.215	-0.243	38.45	72.73	41.11	37.95
	0.02	-0.015	0.045	-0.260	-0.205	-0.233	70.71	178.78	24.75	17.68
.33	0.05	-0.015	0.040	-0.260	-0.205	-0.233	67.08	129.69	31.30	17.89
	0.10	-0.015	0.040	-0.265	-0.205	-0.233	68.41	110.68	25.30	18.97
	0.02	-0.005	0.045	_[b]	_[b]	-	100.53	222.74	_[b]	_[b]
:53	0.05	-0.010	0.045	_[b]	_[b]	-	98.39	161.00	_[b]	_[b]
	0.10	-0.010	0.045	_[b]	_[b]	-	98.31	139.14	_[b]	_[b]

Potentials are expressed in volts and current functions in $\mu A(V/s)^{-1/2}$.

Peak is absent in the first scan.

Broad peak

Table 2.9 Cyclic voltammetric data on protonation of $Cu(L\text{-}Pro)_2$.

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	$E^{o'}_{C-D}$	i_p,A/$\nu^{1/2}$	i_p,B/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$	i_p,D/$\nu^{1/2}$
	0.02	-[b]	0.020	-0.295	-0.230	-0.263	-[b]	45.96	49.50	38.89
7.40	0.05	-[b]	0.020	-0.290	-0.220	-0.255	-[b]	22.36	51.43	40.25
	0.10	-[b]	0.020	-0.290	-0.220	-0.255	-[b]	17.39	53.76	17.39
	0.02	-0.065[c]	0.025	-0.290	-0.230	-0.260	10.61	56.57	42.43	35.36
6.40	0.05	-0.065[c]	0.025	-0.285	-0.225	-0.255	11.18	38.01	46.96	33.54
	0.10	-0.065[c]	0.030	-0.290	-0.220	-0.255	11.08	28.46	44.27	33.20
	0.02	-0.060[c]	0.030	-0.290	-0.230	-0.260	17.68	67.18	38.89	24.75
6.13	0.05	-0.050[c]	0.030	-0.290	-0.230	-0.260	17.89	46.96	40.25	26.83
	0.10	-0.050[c]	0.030	-0.290	-0.230	-0.260	17.39	41.11	37.95	26.88

contd.

Table 2.9 contd.

	0.02	-0.010	0.035	-0.280	-0.220	-0.250	35.36	106.07	28.28	17.68
5.14	0.05	[-0.025 -0.065]	0.035	-0.280	-0.220	-0.250	31.30	67.08	20.12	15.65
	0.10	[-0.020 -0.070]	0.035	-0.285	-0.220	-0.253	31.62	61.66	23.72	14.23
	0.02	-0.010	0.040	-[b]	-[b]	-	67.18	148.49	-[b]	-[b]
4.21	0.05	-0.010	0.040	-[b]	-[b]	-	67.08	109.57	-[b]	-[b]
	0.10	-0.020	0.035	-[b]	-[b]	-	63.25	94.87	-[b]	-[b]
	0.02	-0.005	0.040	-[b]	-[b]	-	74.25	162.63	-[b]	-[b]
3.83	0.05	-0.005	0.040	-[b]	-[b]	-	71.55	111.51	-[b]	-[b]
	0.10	-0.010	0.035	-[b]	-[b]	-	66.41	99.61	-[b]	-[b]

a Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b Peak absent in the first scan.

c Broad peak.

Table 2.10. Cyclic voltammetric data on protonation of $Cu(L\text{-}Ser)_2$[a].

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	$E^{o'}_{C-D}$	i_p,A/$\upsilon^{1/2}$	i_p,B/$\upsilon^{1/2}$	i_p,C/$\upsilon^{1/2}$	i_p,D/$\upsilon^{1/2}$
	0.02	-[b]	0.015	-0.275	-0.220	-0.247	-[b]	38.89	67.18	63.64
7.45	0.05	-[b]	0.010	-0.280	-0.210	-0.245	-[b]	22.36	67.08	62.61
	0.10	-[b]	0.000	-0.280	-0.205	-0.242	-[b]	12.65	63.25	60.92
	0.02	-0.070[c]	0.010	-0.275	-0.220	-0.247	10.61	60.10	60.10	56.57
6.71	0.05	-0.070[c]	0.005	-0.275	-0.210	-0.247	8.94	35.78	60.37	58.14
	0.10	-0.060[c]	0.000	-0.280	-0.210	-0.245	9.49	18.97	56.92	50.60
	0.02	-0.060[c]	0.015	-0.270	-0.220	-0.245	17.68	74.25	56.57	53.03
6.31	0.05	-0.060[c]	0.010	-0.275	-0.215	-0.245	17.89	46.96	53.67	49.19
	0.10	-0.060[c]	0.010	-0.275	-0.210	-0.243	18.97	41.11	52.68	47.43

contd.

Table 2.10 contd.

	0.02	-0.055[c]	0.020	-0.265	-0.220	-0.243	35.36	98.99	45.56	35.36
4.93	0.05	-0.040[c]	0.020	-0.270	-0.220	-0.245	35.78	71.55	42.25	35.78
	0.10	-0.050[c]	0.020	-0.270	-0.215	-0.243	31.62	60.08	44.27	31.62
	0.02	-0.035[c]	0.020	-0.265	-0.220	-0.242	45.96	109.60	42.43	28.28
4.63	0.05	-0.035[c]	0.020	-0.265	-0.220	-0.242	44.72	73.79	38.54	29.07
	0.10	-0.040[c]	0.020	-0.270	-0.220	-0.245	37.95	69.57	38.62	28.46
	0.02	0.020	0.030	-[b]	-[b]	-	84.85	205.06	-[b]	-[b]
4.18	0.05	-0.025	0.030	-[b]	-[b]	-	80.50	138.64	-[b]	-[b]
	0.10	-0.030	0.030	-[b]	-[b]	-	75.89	120.17	-[b]	-[b]

a Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b Peak absent in the first scan.

c Broad peak.

Table 2.11. Cyclic voltammetric data on protonation of $Cu(L\text{-}Ile)_2$[a].

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	$E^{o'}_{C-D}$	i_p,A/$\nu^{1/2}$	i_p,B/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$	i_p,D/$\nu^{1/2}$
	0.02	-[b]	0.010	-0.270	-0.210	-0.240	-[b]	21.21	28.28	24.74
7.05	0.05	-[b]	0.010	-0.270	-0.205	-0.238	-[b]	10.06	27.95	25.71
	0.10	-[b]	0.010	-0.270	-0.205	-0.238	-[b]	7.90	28.46	25.29
	0.02	-0.060[c]	0.025	-0.260	-0.200	-0.230	8.83	35.35	21.21	17.67
6.20	0.05	-0.055[c]	0.015	-0.270	-0.205	-0.238	8.94	38.89	20.12	17.88
	0.10	-0.055[c]	0.015	-0.270	-0.205	-0.238	9.48	17.39	26.87	18.97
	0.02	-0.045[c]	0.020	-0.260	-0.200	-0.230	15.90	45.96	17.67	14.14
5.60	0.05	-0.045[c]	0.020	-0.260	-0.200	-0.230	15.65	32.42	16.77	13.41
	0.10	-0.045[c]	0.020	-0.260	-0.200	-0.230	15.81	28.46	15.81	12.64

contd.

Table 2.11 contd.

	0.02	-0.020	0.025	-[b]	-[b]	-	40.65	99.45	-[b]	-[b]
4.08	0.05	-0.025	0.025	-[b]	-[b]	-	40.24	71.55	-[b]	-[b]
	0.10	-0.025	0.025	-[b]	-[b]	-	37.94	58.50	-[b]	-[b]
	0.02	-0.015	0.030	-[b]	-[b]	-	42.42	95.45	-[b]	-[b]
3.65	0.05	-0.010	0.030	-[b]	-[b]	-	42.48	67.08	-[b]	-[b]
	0.10	-0.020	0.030	-[b]	-[b]	-	39.52	60.08	-[b]	-[b]

a Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b Peak absent in the first scan.

c Broad peak.

Table 2.12. Cyclic voltammetric data on protonation of $Cu(L\text{-}Ala)_2$[a].

pH	Scan rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	$E^{o'}_{C-D}$	i_p,A/$\nu^{1/2}$	i_p,B/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$	i_p,D/$\nu^{1/2}$
	0.02	-[b]	0.025	-0.275	-0.215	-0.245	-[b]	49.49	98.99	98.99
7.52	0.05	-[b]	0.020	-0.280	-0.210	-0.245	-[b]	31.30	93.91	89.44
	0.10	-[b]	0.020	-0.280	-0.205	-0.242	-[b]	18.97	94.86	88.54
	0.02	-0.050[c]	0.035	-0.275	-0.215	-0.245	10.60	77.78	84.85	84.85
6.67	0.05	-0.050[c]	0.025	-0.275	-0.210	-0.243	13.41	44.72	89.44	84.97
	0.10	-0.050[c]	0.025	-0.280	-0.205	-0.243	12.64	34.78	85.38	82.21
	0.02	-0.045[c]	0.025	-0.275	-0.220	-0.247	21.21	81.31	84.85	81.31
6.52	0.05	-0.045[c]	0.030	-0.275	-0.210	-0.243	22.36	58.13	80.49	76.28
	0.10	-0.045[c]	0.030	-0.275	-0.210	-0.243	18.97	44.27	79.05	75.89

contd.

	0.02	-0.035[c]	0.040	-0.270	-0.220	-0.245	35.35	120.20	74.24	65.17
5.86	0.05	-0.050[c]	0.035	-0.270	-0.215	-0.245	35.77	80.49	76.02	62.60
	0.10	-0.040[c]	0.045	-0.275	-0.210	-0.245	34.78	66.40	69.57	62.20
	0.02	-0.030[c]	0.045	-0.270	-0.220	-0.245	56.56	162.63	56.56	45.96
5.06	0.05	-0.030[c]	0.045	-0.270	-0.215	-0.243	49.19	107.33	58.13	44.19
	0.10	-0.040[c]	0.045	-0.270	-0.210	-0.240	50.19	94.86	50.59	44.17
	0.02	-0.005	0.050	-0.260	-0.210	-0.230	77.78	197.98	42.42	29.78
4.44	0.05	-0.010	0.050	-0.260	-0.210	-0.230	71.55	134.16	35.77	30.28
	0.10	-0.010	0.050	-0.260	-0.210	-0.230	69.57	117.00	34.78	28.46
	0.02	0.000	0.050	-[b]	-[b]	-	113.13	268.70	-[b]	-[b]
4.02	0.05	-0.005	0.050	-[b]	-[b]	-	107.33	187.82	-[b]	-[b]
	0.10	-0.010	0.055	-[b]	-[b]	-	104.35	161.27	-[b]	-[b]
	0.02	0.000	0.050	-[b]	-[b]	-	121.28	283.28	-[b]	-[b]
3.85	0.05	-0.010	0.050	-[b]	-[b]	-	120.74	205.71	-[b]	-[b]
	0.10	-0.010	0.055	-[b]	-[b]	-	120.16	179.26	-[b]	-[b]

a Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.
b Peak absent in the first scan.
c Broad peak.

It is now recognised that as the pH decreases from neutral values the Cu(II)A_2 complex gets protonated leading to other forms like the 1:1 species (CuA) and free Cu(II) ions.[7,9,10,16,17] Thus as the pH decreases the CuA and subsequently free Cu(II) ions become the major species in solution. A detailed discussion on the chemistry of protonation is given in chapter 1. The gradual decrease in peak heights of C and D with the decrease of pH, indicates the depletion of the starting CuA_2 species on protonation. At low pH values, when complete protonation results only in free Cu(II) ions, the peaks C and D should completely disappear. This is observed. Since, on decreasing the pH from neutral values more and more of Cu(II) ions are generated, the peaks A and B should increase in heights and gradually should become the only peaks in the CV. These changes are clearly observable in Fig.2.6. It is also to be noted that the cathodic peak A appears from the first scan itself on protonation. This is possible since on decreasing the pH, free copper(II) ions are liberated in solution which can show the characteristic cathodic peak. At neutral pH conditions free Cu(II) ions had to be generated electrochemically, before the cathodic peak A appears from the second scan onwards.

It is also possible that the protonated species, CuA undergoes a one-electron reduction at a potential less cathodic to peak C. Thus it appears that the 'peak-broadening' observed for the peak A on decreasing the pH may be due to the result of the overlap of the reduction wave of CuA with that of free Cu(II) ions liberated from the Cu(II) complexes. If so, the 'peak-broadening' should disappear at very low pH values as the complete protonation of the Cu(II) complexes leaves only free Cu(II) ions in solution. This indeed is observed at a pH about 3.5. These features are very clearly observed in the case of protonation of $Cu(Pro)_2$ and the 'peak-broadening' is shown in Fig.2.7. The broad peak gets split into two overlapping waves at intermediate pH values and at higher scan rates. On decreasing the pH further, the shoulder at more negative potential disappears and the peak becomes a sharp single peak. This single cathodic peak represents the reduction peak of free Cu(II) ions which is the only species at low pH conditions. The anodic peak B, which corresponds to the $Cu^{o} \xrightarrow{2e}$ Cu(II) process, complete the A-B couple at low pH values.

Extensive investigations were also carried out on the reversibility of the protonation process of the Cu(II) amino acid complexes, by raising the pH of the

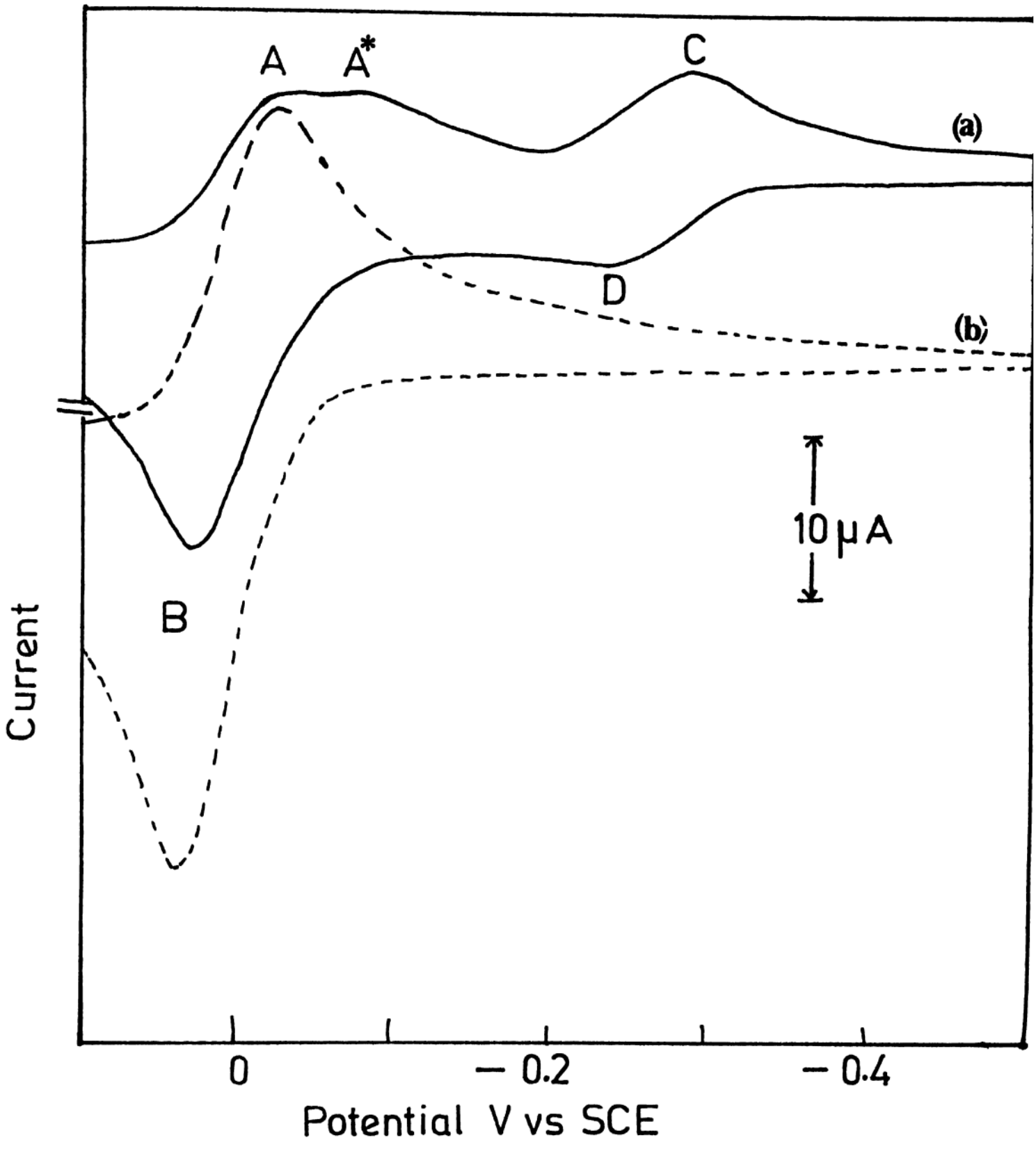


Fig.2.7 Cyclic voltammograms of $Cu(L\text{-}Pro)_2$ at scan rate 0.10 Vs^{-1}
(a) pH = 5.14; (b) pH = 4.21

test solution by the step-wise addition of NaOH to the sample solution from a pH about 3.0. CV were recorded at every stage till the pH reached about 7.0. The results are shown in Fig.2.8. Cyclic voltammetric data on deprotonation of few sample systems are presented in Tables 2.13-2.15. In tables 2.8 to 2.15, it is seen that, in a given pH as the scan rate increases, the value for $i_p,B/\upsilon^{1/2}$ decreases while $i_p,A/\upsilon^{1/2}$, $i_p,C/\upsilon^{1/2}$ and $i_p,D/\upsilon^{1/2}$ remain constant. This results because of the amalgam formation of Cu^o in mercury.[28] These results clearly show that the entire process of protonation is reversible. The cycle, protonation-deprotonation, can be repeated several times.

Thus it can be seen that the protonation studies have corroborated the conclusions presented for the cyclic voltammetric behaviour of Cu(II) amino acid complexes in neutral pH conditions. It has already been mentioned that all the Cu(II) amino acid complexes listed in this chapter behave in a similar fashion both in neutral and low pH conditions. The amino acids used in complex formation have been selected because several of them have additional coordination sites and side chains of varying lengths and bulkiness. It was also the purpose to see the effects of these parameters on the complex formation and subsequently on the reduction pattern. The results presented and

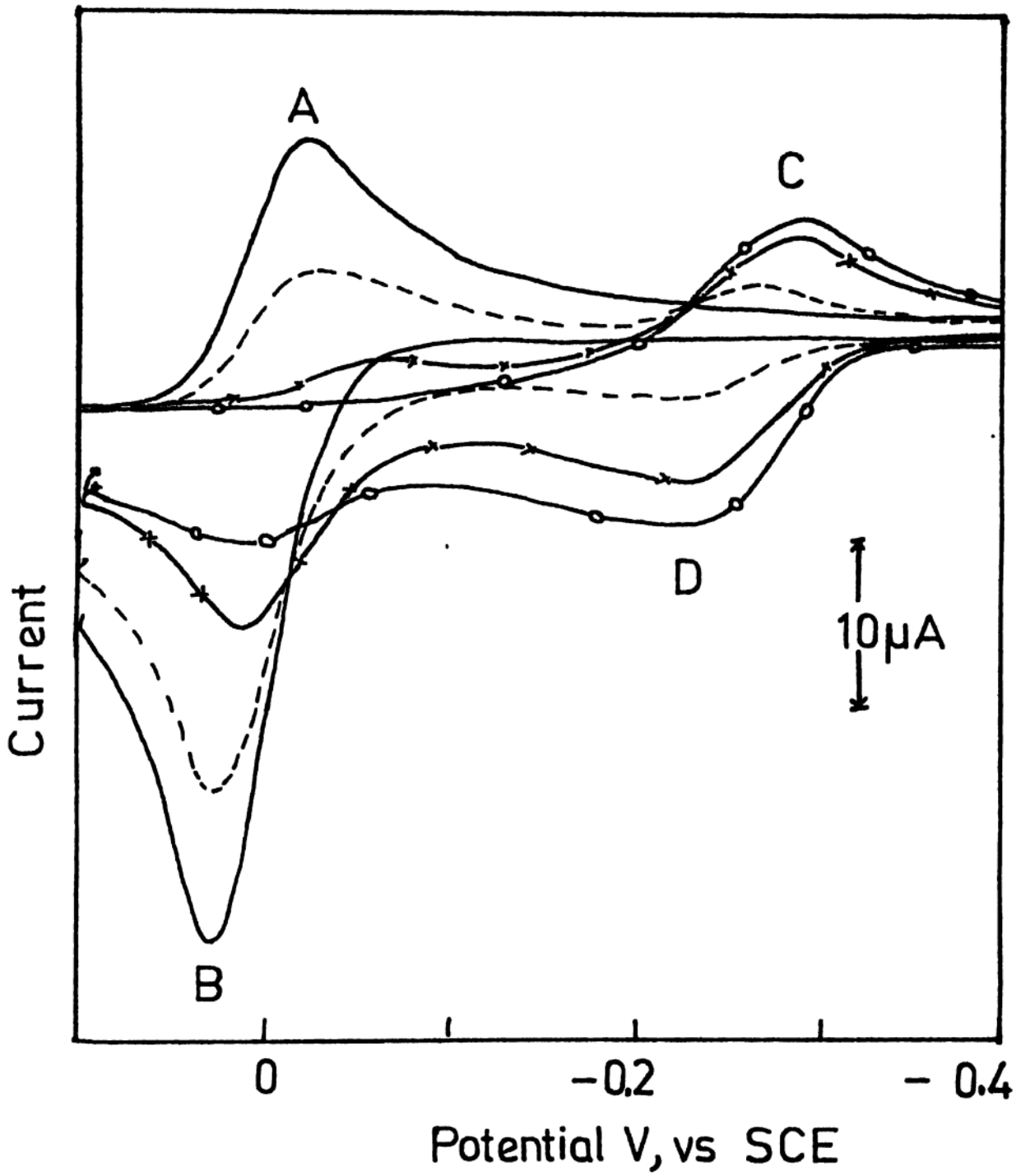


Fig.2.8 Cyclic voltammograms of $Cu(Gly)_2$ on deprotonation at scan rate 0.02 Vs^{-1}. ——, pH 3.53; ----, 4.44; —x—x—, 5.89 —o—o—, 7.42.

Table 2.13. Cyclic voltammetric data on deprotonation of $Cu(Gly)_2$[a].

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	$E^{o'}_{C-D}$	i_p,A/$\upsilon^{1/2}$	i_p,B/$\upsilon^{1/2}$	i_p,C/$\upsilon^{1/2}$	i_p,D/$\upsilon^{1/2}$
	0.02	-0.005	0.045	-[b]	-[b]	-	100.53	222.74	-[b]	-[b]
3.53	0.05	-0.010	0.045	-[b]	-[b]	-	98.39	161.00	-[b]	-[b]
	0.10	-0.010	0.045	-[b]	-[b]	-	98.31	139.14	-[b]	-[b]
	0.02	-0.015[c]	0.040	-0.255	-0.200	-0.228	53.03	148.49	35.36	21.21
4.44	0.05	-0.025[c]	0.030	-0.260	-0.200	-0.230	53.67	98.39	35.78	26.83
	0.10	-0.025[c]	0.030	-0.260	-0.200	-0.230	50.60	82.22	31.62	15.81
	0.02	-0.040[c]	0.030	-0.260	-0.200	-0.230	35.36	106.07	49.50	35.36
5.37	0.05	-0.040[c]	0.030	-0.265	-0.205	-0.235	35.78	71.55	44.72	35.78
	0.10	-0.050[c]	0.030	-0.265	-0.205	-0.235	39.53	60.08	41.11	31.62

contd.

Table 2.13 contd.

	0.02	-0.050[c]	0.025	-0.260	-0.200	-0.230	17.68	77.78	56.57	49.50
5.89	0.05	-0.050[c]	0.020	-0.275	-0.215	-0.247	17.89	53.67	53.67	49.19
	0.10	-0.053[c]	0.020	-0.275	-0.215	-0.245	18.97	34.79	53.76	47.43
	0.02	-[b]	-[b]	-0.280	-0.220	-0.250	-[b]	35.36	74.25	63.67
7.75	0.05	-[b]	-[b]	-0.280	-0.220	-0.250	-[b]	-[b]	62.61	62.61
	0.10	-[b]	-[b]	-0.285	-0.225	-0.255	-[b]	-[b]	69.57	60.08

Potentials are expressed in volts and current faction values in $\mu A(V/s)^{-1/2}$.

For peak absent in the first scan.

Broad peak.

Table 2.14. Cyclic voltammetric data on deprotonation of $Cu(L\text{-}Ala)_2$[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	$E^{o'}_{C-D}$	i_p,A/$\nu^{1/2}$	i_p,B/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$	i_p,D/$\nu^{1/2}$
	0.02	-0.000	0.050	-[b]	-[b]	-	121.28	283.88	-[b]	-[b]
3.85	0.05	-0.010	0.050	-[b]	-[b]	-	120.74	205.71	-[b]	-[b]
	0.1[illegible]	-0.010	0.055	-[b]	-[b]	-	120.16	170.76	-[b]	-[b]
	0.02	-0.005	0.045	-0.250	-0.200	-0.225	95.46	226.27	38.89	17.68
4.26	0.05	-0.010	0.045	-0.255	-0.200	-0.228	89.44	161.00	26.83	18.83
	0.10	-0.020	0.040	-0.255	-0.200	-0.228	85.89	139.14	40.28	18.97
	0.02	-0.020[c]	0.045	-0.265	-0.215	-0.240	63.64	190.92	63.64	45.96
4.65	0.05	-0.030[c]	0.040	-0.270	-0.215	-0.243	62.61	107.33	62.61	44.72
	0.10	-0.035[c]	0.035	-0.275	-0.215	-0.245	60.08	94.87	69.57	44.27

contd.

Table 2.14 contd.

	0.02	-0.040[c]	0.035	-0.275	-0.215	-0.245	46.26	148.26	72.46	55.26
5.29	0.05	-0.045[c]	0.030	-0.275	-0.215	-0.245	44.72	98.39	71.84	53.67
	0.10	-0.050[c]	0.030	-0.275	-0.215	-0.245	44.27	72.73	74.28	56.92
	0.02	-0.050[c]	0.030	-0.275	-0.215	-0.245	-[b]	58.14	98.99	91.92
7.11	0.05	-0.060[c]	0.020	-0.275	-0.215	-0.245	-[b]	44.72	98.39	89.44
	0.10	-0.060[c]	0.020	-0.280	-0.220	-0.250	-[b]	34.79	88.54	88.54

a Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b Peak absent in the first scan

c Broad peak,

Table 2.15. Cyclic voltammetric data on deprotonation of $Cu(Ser)_2$[a].

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	$E^{o'}_{C-D}$	i_p,A/$\nu^{1/2}$	i_p,B/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$	i_p,D/$\nu^{1/2}$
	0.02	-0.020	0.030	-[b]	-[b]	-	84.85	205.06	-[b]	-[b]
4.18	0.05	-0.025	0.030	-[b]	-[b]	-	80.50	138.64	-[b]	-[b]
	0.10	-0.030	0.030	-[b]	-[b]	-	75.89	120.17	-[b]	-[b]
	0.02	-0.035[c]	0.025	-0.265	-0.220	-0.243	49.50	127.28	35.36	28.28
4.60	0.05	-0.035[c]	0.025	-0.265	-0.210	-0.238	46.96	84.97	31.30	26.83
	0.10	-0.040[c]	0.025	-0.265	-0.210	-0.238	44.27	72.73	31.62	25.30
	0.02	-0.050[c]	0.025	-0.275	-0.220	-0.248	28.28	88.39	49.50	42.43
5.43	0.05	-0.050[c]	0.010	-0.275	-0.220	-0.248	26.83	58.14	49.19	44.72
	0.10	-0.050[c]	0.010	-0.275	-0.210	-0.245	25.30	44.27	47.43	44.27

contd.

Table 2.15 contd.

	0.02	-0.050[c]	0.010	-0.280	-0.220	-0.250	10.61	60.10	60.10	56.57
6.53	0.05	-0.060[c]	0.005	-0.280	-0.220	-0.250	8.94	35.78	55.90	58.14
	0.10	-0.060[c]	0.005	-0.280	-0.220	-0.250	9.49	22.14	56.92	50.60

a Potentials expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b Peak absent in the first scan.

c Broad peak.

discussed in earlier sections show that these influences have only marginal effects on the cyclic voltammetric response. It can only mean that in all these complexes the basic coordination sites to the metal ion remains the same, that is to say that amino nitrogen and carboxyl oxygen are the primary coordination sites in almost all amino acids. In CuA_2 type complexes it essentially guarantees CuN_2O_2 type coordination environment. These amino acid complexes are tetragonally distorted from planar configuration. This has been primarily responsible for stabilizing the intermediate copper(I) species generated electrochemically. It should be noted that it is for the first time, to our knowledge, that Cu(I) amino acid complexes have been detected and identified electrochemically as an intermediate species in the electrochemical reduction of copper(II) amino acid complexes.

2.4 Experimental

A. Materials

Glycine, L-ornithine, L-lysine, L-asparagine and bathocuproin reagent etc., were obtained from Sigma Chemicals Co., U.S.A., and were used as such. All other amino acids were purchased from BDH, India. Basic copper

carbonate was used after estimating the copper content gravimetrically.[25] A.R. quality of sodium perchlorate was used. Purified and distilled (twice) mercury was used throughout the study.

B. Preparation of Cu(II) Amino Acid Complexes

The Cu(II) amino acid complexes were prepared by employing a general procedure reported elsewhere.[29] Stoichiometric amounts of the amino acid (2 mM) was added to a suspension of basic copper carbonate (1 mM) in water with stirring. The mixture was heated to 50°C and stirred for 1-2 hrs. The resulting blue solution filtered to remove the unreacted copper carbonate. The pH of the solution was adjusted to about 7.5 and the solution was slowly evaporated to small volume. It was kept at room temperature for 3-4 days, whereupon blue crystals started depositing. The crystals were filtered, washed with water and dried in vacuum at 100-110°C. Their purity was checked by analytical and spectral methods.

Copper content of the amino acid complexes were estimated gravimetrically as cuprous thiocyanate, after decomposing the complexes with conc. HNO_3.

C. Intermediate Trapping

The intermediate Cu(I) species generated in the electrode process was trapped by using bathocuproin reagent. A few mls of bathocuproin reagent solution in amyl alcohol was mixed well with the aqueous solution of the sample in the electrochemical cell by stirring. While oxygen free nitrogen was passing through the solution it was subjected to controlled potential electrolysis at a potential -0.600 V vs SCE. Mercury-pool electrode was used as working electrode. At the end of the electrolysis, the deep yellow organic layer was separated, washed several times with water and its absorption spectra was recorded in the region 200-800 nm. The CV of this solution was also recorded after adding TEAP in acetone as supporting electrolyte.

An authentic sample of the Cu(I) bathocuproin complex was prepared as follows: A solution of bathocuproin reagent in amyl alcohol was added to an aqueous solution of the freshly prepared Cu(I) ions* and stirred for about 10-15 minutes. The resulted deep yellow alcoholic layer was separated and washed with water several times. The electronic spectra and cyclic voltammogram of this solution was recorded as usual.

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*Cu(I) ions were prepared by reducing a Cu(II) solution by sodium metabisulphite.

D. Cyclic Voltammetry

Cyclic voltammograms were recorded using PAR 370 electrochemistry system which includes the 174 polarographic analyzer, 175 universal programmer, RE 0074 X-Y recorder, and a 377 A cell system with a metrohm E410 handing mercury drop electrode (HMDE) as working electrode. A platinum-wire auxiliary electrode and a saturated calomel electrode (SCE) as reference electrode complete the electrode assembly. The SCE was checked for its potential and necessary corrections were applied wherever necessary before reporting the potentials of the samples.

About 5 ml of the test solution (1 mM) containing 0.1 M $NaClO_4$ as supporting electrolyte was transferred to the cell and deareated by passing oxygen free nitrogen for 10-15 minutes. The potentials were scanned in the range +0.1 to -0.6 V vs SCE. CV at different scan rates were recorded for both single and multiscans. Each time a fresh drop of mercury of same size was used. Throughout the experiment the solution was kept under nitrogen blanket.

E. Controlled Potential Coulometry

Controlled potential coulometry (CPC) was carried out using PAR 173 potentiostat/galvanostat along with a

179 digital coulometer. Mercury (distilled) pool electrode was used as working electrode. Saturated calomel electrode and a platinum-wire electrode (AE) complete the circuit. 2-3 ml of the test solution (1-2 mM) containing 1 mM $NaClO_4$ was taken over mercury in the cell and the solution deareated by passing nitrogen. A constant potential of -0.6 V vs SCE was applied to the solution. The completeness of the electrolysis was monitored from an X-Y recorder which plots i-t curve and the digital coulometer reads the coulombs accumulated. The experiment was repeated 3-4 times and the average of the values were taken. A similar experiment was conducted with a blank solution with the same supporting electrolyte concentration. The amount of charge accumulated for the reduction of the sample was obtained by subtracting that for the blank solution from the first reading.

2.5 References

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CHAPTER 3

CYCLIC VOLTAMMETRIC STUDIES OF SOME COPPER(II) DIPEPTIDE COMPLEXES. ELECTROCHEMICAL EVIDENCE FOR COPPER(I) DIPEPTIDE COMPLEXES IN AQUEOUS MEDIA*

3.1 Abstract

The reduction of Cu(II) dipeptide complexes is studied by cyclic voltammetry in aqueous media at HMDE. It proceeds through a one-electron process generating intermediate Cu(I) dipeptide complexes, which is found to be short-lived. A fraction of it undergoes chemical reactions eventually generating Cu^{o} at the mercury electrode. The other fraction of the Cu(I) species is reoxidised to the Cu(II) complexes. The generated Cu^{o} undergoes a

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*Part of the work discussed in this chapter will appear in Polyhedron, 1984, 3, 0000.

two-electron oxidation at more anodic potentials than the Cu(I) species. The transient existence of the Cu(I) complexes is established by trapping the intermediate Cu(I) species using bathocuproin reagent. It is expected that this Cu(I) species will also have the same coordination sites of the parent complex, that is, the deprotonated peptide nitrogen, amino nitrogen and the carboxyl oxygen. The CV pattern corresponds to an ECE mechanism for the reduction of the title complexes where E_2^o is anodic to E_1^o and $n_1 = 1$ and $n_2 = 2$. The pH dependence of the Cu(II) complexes is also investigated by cyclic voltammetry. The changes in the coordination pattern on protonation of the complexes are reflected in the CV response. The process of protonation is found to be quite reversible.

3.2 Introduction

During the last two decades intensive investigations have been carried out on the copper(II) di and oligo peptide complexes to elucidate and interpret the thermodynamic and structural characteristics of them.[1-4] In peptides, and probably also in proteins, the deprotonated amide group is one of the important binding sites for the coordination of Cu(II) ions.[5] Dipeptides coordinate to copper(II) at physiological pH, using deprotonated amide

nitrogen, amino nitrogen and the carboxyl oxygen.[6-8] The involvement of copper(II) in redox processes in biological systems, where copper is bound by histidyl and probably deprotonated peptide-nitrogens has received considerable attention. Copper(I) complexes involving deprotonated peptide nitrogen coordination are of much current interest.[9-12] In contrast with Cu(II) and Cu(III) ions there is very little evidence of deprotonated-peptide nitrogen coordination in Cu(I) complexes. Many workers have tried to detect Cu(I) deprotonated peptide-nitrogen coordination by using potentiometric titration,[9] NMR,[10-12] etc., methods. However, the bulk of the data in literature indicate that such a coordination is very rare and does not occur readily. Very recently Margerum and co-workers[13] observed copper(I) ternary diglycine complex with dmp (2,9-dimethyl-1,10-phenanthroline) by cyclic voltammetry. However, no Cu(I) species could be detected during the cyclic voltammetric studies of the reduction of parent Cu(II) triglycine complexes.[13] Our studies on the reduction of copper(II) amino acid complexes by cyclic voltammetry, discussed in chapter 2, showed the generation of copper(I) complexes as an intermediate in aqueous media. This has been the driving force to explore the reduction of copper(II) peptide complexes for a possible Cu(I) species as an intermediate in these complexes. We were

also interested in studying the effect of changing coordination sites and possibly the geometry of copper complexes on redox behaviour. With these intentions the reduction of a large number of copper(II) dipeptide complexes by cyclic voltammetry have been examined in aqueous media both at near neutral and at low pH conditions. Our results are presented and discussed in this chapter.

3.3 Results and Discussion

A. Synthesis and Characterization of Complexes

The copper(II) dipeptide complexes were prepared by a general procedure, the details of which are given in the experimental section. The dipeptides used in the preparation of the complexes, their structure and the abbreviations used in the present discussion are given in Table 3.1. The identity of the complexes was established by elemental analysis and spectral methods. These results are tabulated in Table 3.2. The complexes have the general formula, Cu(II)L where L represents the dipeptide dianion. The complexes are all of 1:1 composition and are neutral. The coordination sites are the amino and the deprotonated amide nitrogens and the carboxyl oxygen. Two more coordinated water molecules makes the 'geometry' square pyramidal.

Table 3.1. List of the dipeptides used for complex formation.

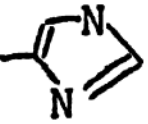
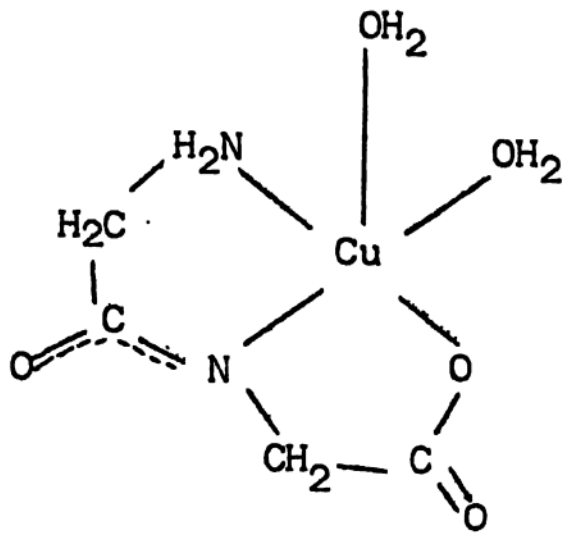
Name	Abbreviation	Structure	Abbreviation of the copper complex
Glycyl-L-Serine	Gly Ser	$H_2N-CH_2-CO-NH-CH(CH_2OH)-COO^-$	Cu(Gly Ser)
Glycyl-Glycyl	Gly Gly	$H_2N-CH_2-CO-NH-CH_2-COO^-$	Cu(Gly Gly)
Glycyl-L-Phenylalanine	Gly Phe	$H_2N-CH_2-CO-NH-CH(CH_2\phi)-COO^-$	Cu(Gly Phe)
Glycyl-L-Isoleucine	Gly Ile	$H_2N-CH_2-CO-NH-CH[CH(CH_3)CH_2CH_3]-COO^-$	Cu(Gly Ile)
Glycyl-L-Tryptophan	Gly Trp	$H_2N-CH_2-CO-NH-CH(CH_2-$  $)-COO^-$	Cu(Gly Trp)
Glycyl-L-Threonine	Gly Thr	$H_2N-CH_2-CO-NH-CH\ CH(CH_3)OH\cdot-COO^-$	Cu(Gly Thr)
Glycyl-L-Tyrosine	Gly Tyr	$H_2N-CH_2-CO-NH-CH(CH_2-\phi\ OH)-COO^-$	Cu(Gly Tyr)
Glycyl-L-Methionine	Gly Met	$H_2N-CH_2-CO-NH-CH(CH_2SCH_3)-COO^-$	Cu(Gly Met)
Glycyl-L-Histidine	Gly His	$H_2N-CH_2-CO-NH-CH(CH_2-$  $)-COO^-$	Cu(Gly His)
L-Methionyl-Glycine	Met Gly	$H_2N-CH(CH_2CH_2SCH_3)-CO-NH-CH_2-COO^-$	Cu(Met Gly)

Table 3.2. Spectral and analytical data for copper(II) dipeptide complexes.

Compound	pH	Visible spectra obsd λ_{max}	Visible spectra obsd ϵ	% C obsd	% C calcd	% H obsd	% H calcd	% N obsd	% N calcd
Cu(Gly Ser)	7.15	634	101	24.08	24.84	4.29	4.14	11.65	11.59
Cu(Gly Gly)	6.60	640	110	24.77	24.80	3.70	3.10	14.50	14.47
Cu(Gly Phe)	7.12	630	62	-	-	-	-	-	-
Cu(Gly Ile)	6.28	634	96	35.93	35.89	5.32	5.93	10.96	10.47
Cu(Gly Trp)	6.25	626	96	41.58	41.54	5.02	4.79	11.18	11.19
Cu(Gly Thr)	6.73	632	84	27.92	28.18	4.46	4.70	10.82	10.96
Cu(Gly Tyr)	6.34	632	64	-	-	-	-	-	-
Cu(Gly Met)	6.55	634	69	31.02	31.40	4.60	4.49	14.46	10.47
Cu(Gly His)	7.38	624	57	31.25	31.02	4.04	4.52	18.31	18.09
Cu(Met Gly)	6.62	634	86	31.74	31.40	4.68	4.49	11.12	10.47

The structure of a typical dipeptide complex, Cu(Gly Gly) is shown in 3.1.



3.1

The redox behaviour of these complexes was examined by cyclic voltammetry and in the following sections the results are presented and discussed.

B. Cyclic Voltammetric Response at Neutral pH

Cyclic voltammograms were recorded in the potential range +0.1 to -0.6 V versus SCE at 25°C in aqueous solution (1×10^{-3}M) at the dissolution pH of about 7. The pH was not adjusted after dissolving the samples in water. Sodium perchlorate (1 mM) was used as supporting electrolyte. Hanging Mercury Drop Electrode (HMDE) of definite area

was used as the working electrode. The details of the experimental set up is given in the experimental section in chapter 2. All complexes show reasonably similar CV response on reduction. There are no peaks in the positive potential range for any of the copper(II) dipeptide complexes under the experimental conditions. None of the ligands in the absence of metal ions show any peak in the potential range, +1.0 to -1.0 V versus SCE. The voltammograms of Cu(Gly Ser) and Cu(Gly Gly) are shown in Fig.3.1 as examples of the general behaviour. In Cu(Gly Ser) the separation between C and D is much smaller than that in Cu(Gly Gly). The CV profile of the rest of the Cu(II) dipeptide complexes are similar to Cu(Gly Gly) complex. The relevant CV data for all complexes are given in Table 3.3 The parameters in this table have the usual significance.

The general features of the voltammetric response for these complexes are summarised below.

(i) During the first forward scan only one cathodic peak C is observed at about -0.300 V.

(ii) In the reverse scan an anodic peak D at $\sim$ -0.18 V appears along with another anodic peak B at about +0.03 V.

(iii) A new cathodic peak A appears during the second scan at $\sim$ -0.03 V.

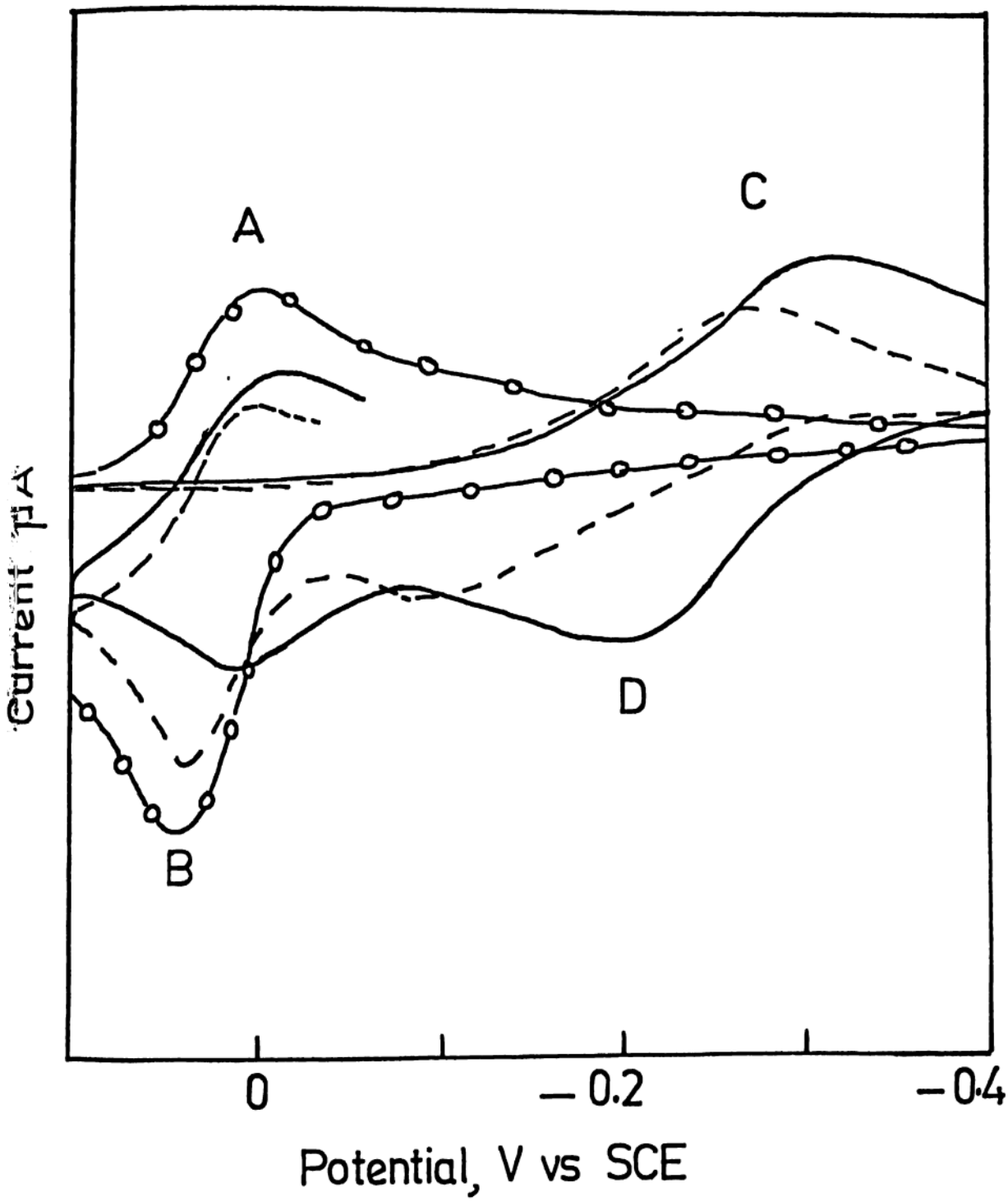


Fig.3.1 Cyclic voltammograms of: ——, Cu(Gly Ser); – – – , Cu(Gly Gly); —o—o— , $Cu(ClO_4)_2.6H_2O$.

Table 3.3. Cyclic voltammetric data on copper(II) dipeptide complexes in aqueous media.[a]

Complex (pH; Conc.)	Sweep rate (Vs^{-1})	E_p,A [b]	E_p,B	E_p,C	E_p,D	$\Delta E_p,C-D$	$\frac{E_p,C+E_p,D}{2}$
Cu(Gly Ser)	0.01	-0.030	0.030	-0.310	-0.205	0.105	-0.258
(pH=7.53;	0.02	-0.040	0.025	-0.310	-0.195	0.115	-0.253
1.02 mM)	0.04	-0.040	0.020	-0.320	-0.190	0.130	-0.255
	0.08	-0.050	0.015	-0.325	-0.185	0.140	-0.255
	0.10	-0.050[c]	0.015	-0.320	-0.185	0.135	-0.253
	0.14	-0.050[c]	0.010	-0.330	-0.180	0.150	-0.255
	0.18	-0.050[c]	0.010	-0.335	-0.175	0.160	-0.255
	0.20	-0.050[c]	0.010	-0.335	-0.175	0.160	-0.255
Cu(Gly Gly)	0.01	-0.010	0.035	-0.325	-0.150[c]	0.175	-0.238
(pH=7.38;	0.02	-0.010	0.040	-0.335	-0.130[c]	0.205	-0.233
1.11 mM)	0.04	-0.010	0.040	-0.345	-0.120[c]	0.225	-0.233
	0.08	-0.010	0.040	-0.350	-0.115[c]	0.235	-0.233
	0.10	-0.010	0.040	-0.355	-0.110[c]	0.245	-0.233

contd.

Table 3.3 contd.

Cu(Gly Phe)	0.01	-0.015	0.040	-0.290	-0.150	0.140	-0.220
(pH=6.67;	0.02	-0.015	0.040	-0.295	-0.150	0.145	-0.223
1.22 mM)	0.04	-0.020	0.045	-0.300	-0.140	0.160	-0.220
	0.08	-0.025	0.045	-0.310	-0.120[c]	0.190	-0.215
	0.10	-0.030	0.040	-0.310	-0.120[c]	0.190	-0.215
	0.20	d	d	-0.315	d	-	-
Cu(Gly Ile)	0.01	-0.010	0.040	-0.300	-0.110	0.190	-0.205
(pH=6.86;	0.02	-0.015	0.045	-0.305	-0.100[c]	0.205	-0.203
1.08 mM)	0.04	-0.020	0.050	-0.310	-0.070	0.240	-0.190
	0.10	d	d	-0.330	-0.050	0.280	-0.190
	0.20	d	d	-0.340	c	-	-
Cu(Gly Trp)	0.01	-0.010	0.040	-0.330	c	-	-
(pH=7.32;	0.02	-0.010	0.040	-0.340	c	-	-
1.02 mM)	0.10	d	d	-0.365	c	-	-

contd.

Table 3.3 contd

Cu(Gly Thr)	0.01	-0.010	0.040	-0.370	-0.115	0.255	-0.243
(pH=7.22;	0.02	-0.015	0.040	-0.370	-0.110	0.260	-0.240
1.25 mM)	0.04	-0.010	0.040	-0.360	-0.100	0.260	-0.230
	0.10	d	d	-0.355	-0.090	0.265	-0.223
Cu(Gly Tyr)	0.01	-0.015	0.040	-0.310	-0.120	0.190	-0.215
(pH=7.39;	0.02	-0.010	0.040	-0.315	-0.120	0.195	-0.218
1.15 mM)	0.04	-0.020	0.040	-0.325	-0.110	0.215	-0.218
	0.10	d	d	-0.325	-0.080	0.245	-0.203
	0.20	d	d	-0.335	-0.070	0.265	-0.203
Cu(Gly Met)	0.01	-0.010	0.035	-0.300	-0.140	0.160	-0.220
(pH=6.80;	0.02	-0.010	0.035	-0.305	-0.130	0.175	-0.218
1.25 mM)	0.05	-0.015	0.040	-0.310	-0.125	0.185	-0.218
	0.10	d	d	-0.315	-0.115[c]	0.200	-0.215

contd.

Table 3.3 contd.

Cu(Gly His)	0.01	-0.010	0.040	-0.380	-0.120	0.260	-0.250
(pH=7.21;	0.02	-0.015	0.040	-0.385	-0.110	0.275	-0.248
0.95 mM)	0.05	-0.020	0.040	-0.390	-0.100 -0.310	0.290 0.080	-
	0.10	d	d	-0.400	-0.310 -0.090	0.310 0.090	-
Cu(Met Gly)	0.02	-0.010	0.040	-0.305	-0.130	0.175	-0.218
(pH=6.80;	0.05	-0.010	0.040	-0.315	-0.120	0.195	-0.218
1.10 mM)	0.10	-0.015	0.040	-0.320	-0.115	0.205	-0.218

a. Potentials are expressed in volts.

b. Peak appears only in the second and subsequent scans.

c. Broad peak.

d. Peak absent.

(iv) On increasing the scan rate the peak heights of A and B decrease considerably while peaks C and D increase only proportional to $\nu^{1/2}$. This is shown in Fig.3.2. This phenomena becomes clear when plotting the $i_p/\nu^{1/2}$ values against scan rate for various peaks. The plots are shown in Fig.3.3 and the data are given in Table 3.4. Similar observations were made in the case of copper(II) amino acid complexes. The reason for this is discussed in chapter 2.

(v) On holding the potential at a point well-past the peak C for a few minutes and on resuming the scan, peaks A and B increase in heights drastically whereas the peaks C and D retrace. A similar effect is observed on multiscans in the full potential range (+0.1 to -0.6 V).

(vi) If the potential scan is reversed after the peak A in the second and subsequent scans, the heights of A and B decrease considerably.

(vii) Peaks A and B do not appear if the potential scan is reversed before the appearance of peak C in the first scan itself.

These observations are very similar to those in the case of copper(II) amino acid complexes discussed in chapter 2. Hence, the pattern of discussion and conclusion bear very close similarity.

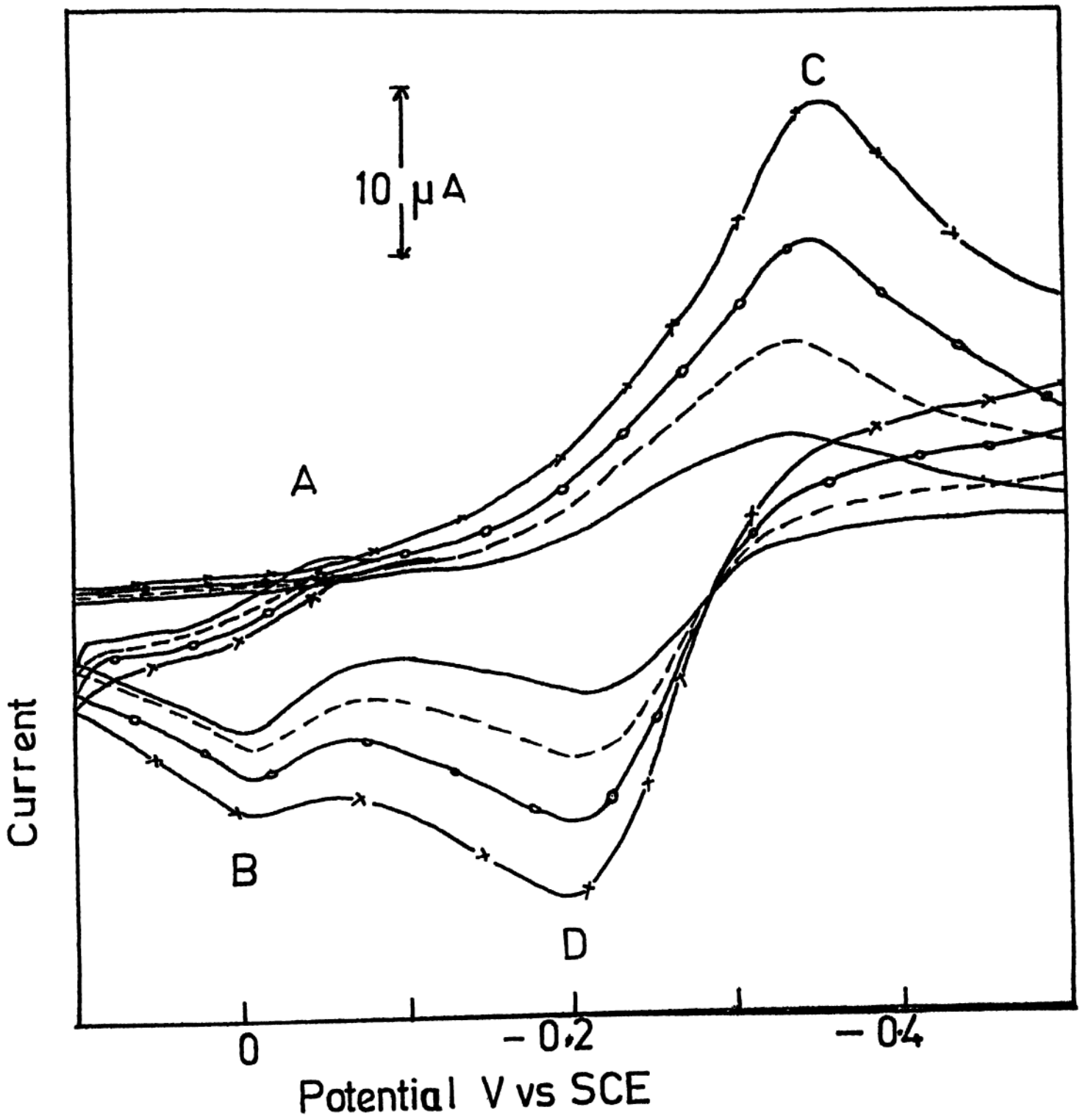


Fig.3.2 Cyclic voltammograms of Cu(Gly Ser) at various scan rates: ——— , 0.02 Vs^{-1} — — — , 0.05 Vs^{-1}; —o—o— , 0.10 Vs^{-1}; —*—*— , 0.20 Vs^{-1}.

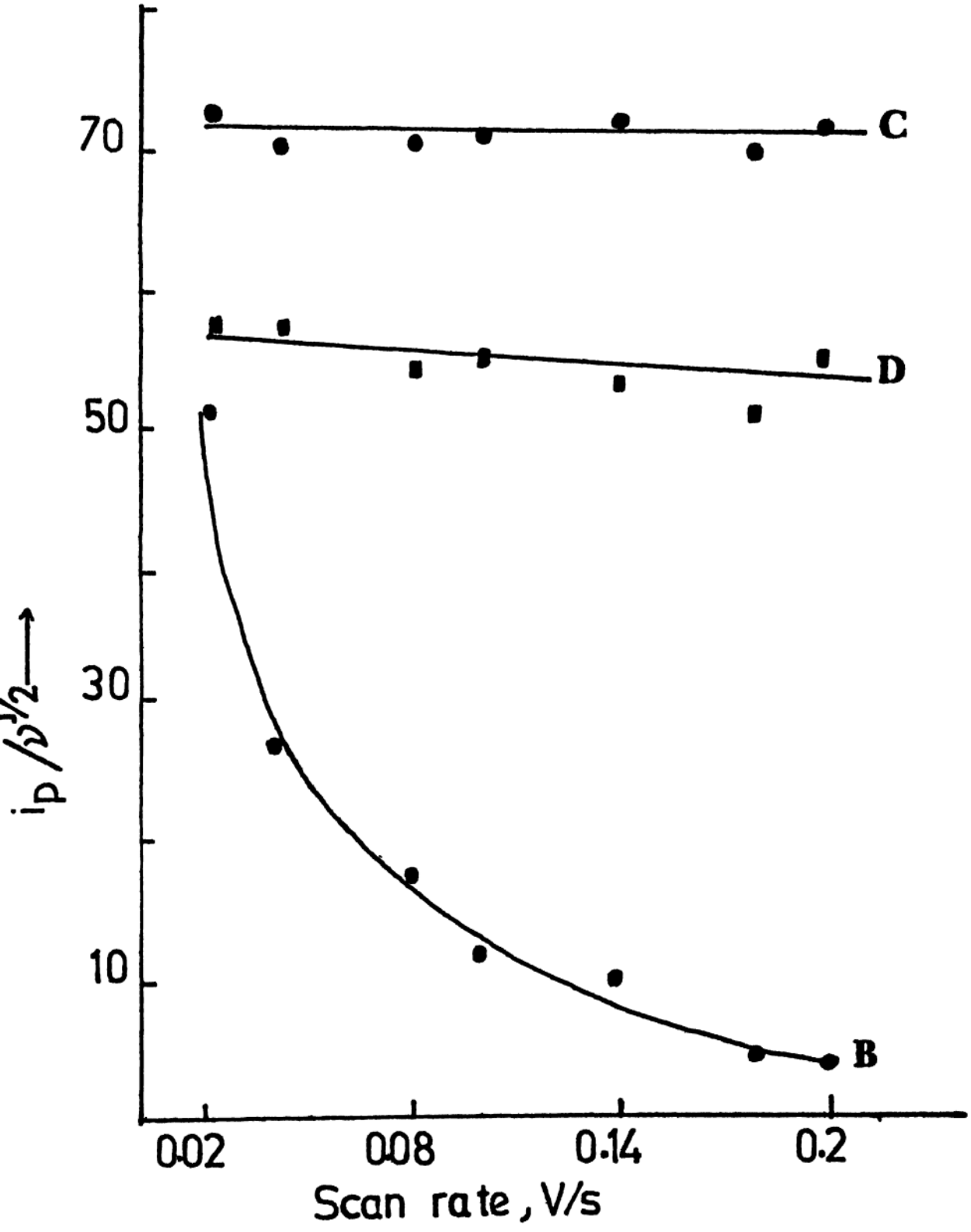


Fig.3.3 Plots of $i_p/\nu^{1/2}$ values against scan rate for various peaks in the voltammogram of Cu(Gly Ser). C - peak C; D - peak D; B - peak B

Table 3.4. Current function values at various scan rates for Cu(Gly Ser)[a]

Scan rate (Vs^{-1})	$i_p,B/\nu^{1/2}$	$i_p,C/\nu^{1/2}$	$i_p,D/\nu^{1/2}$	$i_p,D/i_p,C$
0.01	87.50	75.00	65.00	0.87
0.02	51.26	72.48	65.41	0.90
0.04	27.50	70.00	60.00	0.86
0.08	17.68	70.71	54.80	0.78
0.10	12.65	71.15	55.34	0.78
0.14	10.69	72.16	53.45	0.74
0.18	4.71	70.71	51.85	0.73
0.20	4.41	71.55	55.90	0.78

a. Current function values are expressed in $\mu A(V/s)^{-1/2}$. and the data is collected from aqueous solution of the sample at pH 7.53 and at HMDE (A = 0.02 cm^2).

It is well documented that in Cu(II) dipeptide complexes, the dipeptides coordinate in a terdentate manner using its amino and deprotonated peptide nitrogens and carboxyl oxygen. Two water molecules complete the coordination sites of copper(II).[8] It is also established that the complexes retain their structure on dissolution in aqueous media at neutral pH conditions.[8b,14] Hence the CV recorded in aqueous solutions at about pH 7 corresponds solely to the species of composition, Cu(II)L. Then, peak C of the CV shown in Fig.3.1 can arise due to two possibilities, either due to a one-electron reduction or due to a two-electron reduction of Cu(II)L. These two possibilities are shown in equations 3.1 and 3.2.

$$Cu(II) \xrightarrow{1e} Cu(I) \qquad \ldots 3.1$$

$$Cu(II) \xrightarrow{2e} Cu^{o} \qquad \ldots 3.2$$

If peak C is the result of the two-electron process, then the appearance of peak D cannot be easily explained. On the other hand if the reduction occurs by the process in equation 3.1, peak D can originate from the re-oxidation of the reduced Cu(I) species. It should be noted here that although the general profile of peaks C and D appears to be same for the amino acid and dipeptide complexes,

the peak separation between C and D for amino acid complexes is about 70 mV while that of the dipeptide complexes is about 100-200 mV. Hence for the dipeptide complexes this electron-transfer at best can be regarded as quasi-reversible. The nature and position of peaks A and B are very similar to those observed in the case of Cu(II) amino acid complexes (Ch.2) and compares very well with those of free copper(II) ions (Fig.3.1). The observations (iii) to (v) and (vii) indicate that the first reduction of the starting complex Cu(II)L, is a pre-requisite for the generation of the redox couple A-B. This will imply that the electrogenerated Cu(I)L complex, being an unstable species,[13] undergoes decomposition and/or disproportionation generating Cu^{o} at the electrode. In that case peak B results from a two-electron oxidation of Cu^{o} to Cu(II) ions and peak A in the subsequent scans corresponds to the reduction of Cu(II) ions to Cu^{o}. A comparison of the CV of free copper(II) ions with that of peaks A and B as shown in Fig.3.1, confirms that the peak A and B originate from the Cu^{o}/Cu(II) redox system. The absence of peak A in the first scan shows the absence of free copper(II) ions in solution to start with. Thus the overall electrode process can be represented by equations 3.3-3.5.

$$Cu(II)L \xrightleftharpoons{1e} [Cu(I)L] \quad \ldots 3.3$$

$$[Cu(I)L] \xrightarrow{\text{chemical reactions}} Cu^{o}_{(Hg)} + L^{2-} \quad \ldots 3.4$$

$$Cu^{o}_{(Hg)} \xrightleftharpoons{2e} Cu^{2+}_{(aq)} \quad \ldots 3.5$$

The electrode reactions in equation 3.3 are responsible for the C-D couple while those in equation 3.5 accounts for the A-B couple.

Equation 3.3 which generates the copper(I) dipeptide complexes, represents the most important step in the overall process. Cu(I) complexes are generally stable only in tetrahedral or pseudo-tetrahedral geometries.[15] The Cu(I) dipeptide complexes generated by the reduction can be expected to have a geometry distorted towards tetrahedral, since the parent copper(II) complexes possess a distorted square pyramidal geometry.[8] The extent of stability of the Cu(I) species depends on the degree of tetragonality. The transient existence of Cu(I) dipeptide complexes demonstrated by the CV pattern (peak D) shows that it has a life-time long enough to be detected by cyclic voltammetry. The intermediate Cu(I) dipeptide complexes can be expected to retain the basic coordination pattern of the parent complexes, that is, the deprotonated

peptide nitrogen, the amino nitrogen and the carboxyl oxygen. Coordination from deprotonated peptide nitrogen to Cu(I) is not very common and the Cu(I) intermediate decomposes in course of time. It is thus the time factor of the experiment that is crucial for the detection of this species. The present work is one of the few examples where Cu(I) dipeptide complexes are generated and identified electrochemically.

The CV response of most of the Cu(II) dipeptide complexes are slightly different from that of the Cu(Gly Ser) complex in the sense that the anodic peak D develops clearly only at scan rates higher than 100 mV/s. This implies that the Cu(I) intermediate species of these complexes have a shorter life-time than that of Cu(Gly Ser) complex. For them the peak D is at about 200 mV anodic to peak C showing the process in equation 3.3 is quasi-reversible. In Cu(Gly His) complex, the peak C appears at much more negative potentials (Table 3.3). For glycyl-L-histidine ligand, imidazole nitrogen becomes a coordination site instead of the carboxyl oxygen as in other cases. This change in coordination pattern should account for the shift in the position of peak C. The CV response for Cu(Gly Ser) is very similar to that of Cu(II) amino acid complexes discussed in chapter 2.

The Cu(I) species generated as an intermediate is shown to have only a short life-time.[13] Hence the fraction of the intermediate, which has not been re-oxidised to the parent complex undergoes decomposition ultimately generating Cu^{o} at the electrode. The equation 3.4 may involve the steps as in 3.6 and 3.7.

$$[Cu(I)L] \rightleftharpoons Cu^{1+} + L^{2-} \quad \ldots 3.6$$

$$2\ Cu^{1+} \rightleftharpoons Cu^{o} + Cu^{2+} \quad \ldots 3.7$$

Under the negative potential prevailing in the experimental conditions, the major species in equation 3.7 will be Cu^{o} and it is continuously removed from the reaction medium as mercury amalgam.[16] The observation that $i_p/\vartheta^{1/2}$ values decrease with increase in scan rates for peaks A and B shows that at rapid scan rates the Cu^{o} generation by the interposed chemical reaction decreases. It is also observed that peaks C and D do not get affected by holding the potential at -0.500 V whereas peaks A and B increase considerably. Such a situation can arise only when a steady concentration of the reduced species [Cu(I)L] is maintained at the electrode. This happens only if the process in equation 3.3 is faster than that in 3.4. On the other hand if the process in equation 3.4 is faster

than that in 3.3, then peak D would have diminished at slow scan rates as observed by others,[13,17] or would have disappeared on holding the potential at -0.500 V.

Reaction sequences 3.3 - 3.5 is consistent with ECE mechanism,[18] which has the following characteristics: E_2^o is anodic to E_1^o, $n_1 = 1$, $n_2 = 2$ and the first electron-transfer process is quasi-reversible while the second is reversible.

The effect of ligand concentration on the cyclic voltammetric response of Cu(Gly Gly) complex was examined. The peak A originates from the first scan itself and shifts cathodically with the increase in the glycyl-glycine concentration. The relevant data are presented in Table 3.5. Similar CV behaviour in the case of copper(II) triglycine system has been reported by Margerum and co-workers.[13] This has been explained as due to the catalytic decomposition of Cu(II) peptide complexes to free Cu(II) ions in the presence of excess ligand. A similar reasoning holds good for the present systems too. That is to say that glycyl glycine acts as a proton donor, catalysing the decomposition of the dipeptide complex to free copper ions. The presence of free copper(II) ions in solution is responsible for the appearance of peak A in the

Table 3.5. Effect of ligand (conc) on the CV of Cu(Gly Gly) in aqueous media.

Concn. of the complex ($x10^{-3}M$)	Concn. of the ligand (M)	pH	Sweep rate (Vs^{-1})	E_p,A [a]	E_p,B	$\frac{E_p,A+E_p,B}{2}$	E_p,C	E_p,D	$\Delta E_p,C-D$	$E^{o'}_{C-D}$
1	0.0	6.42	0.02	+0.005	0.040	+0.018	-0.310	-0.135	0.175	-0.223
			0.05	0.000	0.035	+0.018	-0.320	-0.100	0.220	-0.220
			0.10	0.000	0.040	+0.020	-0.330	-0.100	0.230	-0.215
			0.15	-0.005	0.050	+0.023	-0.335	-0.090[b]	0.245	-0.213
1	0.05	5.70	0.02	-0.090	-0.005	-0.048	-0.230	-0.110	0.120	-0.170
			0.05	-0.090	-0.010	-0.050	-0.240	-0.105	0.135	-0.173
			0.10	-0.095	-0.015	-0.055	-0.250	-0.100	0.150	-0.175
			0.15	-0.095	-0.020	-0.058	-0.260	c	-	-
1	0.10	5.79	0.02	-0.110	-0.025	-0.068	-0.205	-0.110[b]	0.095	-0.157
			0.05	-0.115	-1.030	-0.073	-0.225	c	-	-
			0.10	-0.115	-0.035	-0.073	-0.230	c	-	-
			0.15	-0.120	-0.040	-0.080	-0.235	c	-	-

contd.

Table 3.5 contd.

			0.02	-0.135	-0.045	-0.090	-0.210	c	-	-
			0.05	-0.130	-0.045	-0.088	-0.210	c	-	-
1	0.15	5.76	0.10	-0.130	-0.050	-0.090	-0.225	c	-	-
			0.15	-0.130	-0.050	-0.090	-0.230	c	-	-
			0.02	-0.140	-0.060	-0.100	-0.200	c	-	-
			0.05	-0.135	-0.060	-0.098	-0.205	c	-	-
1	0.20	5.72	0.10	-0.130	-0.060	-0.095	-0.215	c	-	-
			0.15	-0.135	-0.065	-0.100	-0.220	c	-	-
			0.02	-0.150[b]	-0.070	-0.110	-0.170[b]	c	-	-
			0.05	-0.140	-0.065	-0.103	-0.190[b]	c	-	-
1.	0.25	5.80	0.10	-0.140	-0.070	-0.105	-0.210	c	-	-
			0.15	-0.140	-0.070	-0.105	-0.215	c	-	-
			0.02	-0.160[b]	-0.065	-0.113	c	c	-	-
			0.05	-0.150[b]	-0.075	-0.113	-0.190[b]	c	-	-
1	0.30	5.86	0.10	-0.150	-0.075	-0.113	-0.200	c	-	-
			0.15	-0.145	-0.075	-0.110	-0.210	c	-	-

contd.

Table 3.5 contd.

1	0.35	5.85	0.02	-0.160[b]	-0.075	-0.118	c	c	-	-
			0.05	-0.160[b]	-0.075	-0.118	c	c	-	-
			0.10	-0.155	-0.080	-0.118	-0.210	c	-	-
			0.15	-0.150	-0.075	-0.113	-0.210	c	-	-
1	0.40	5.85	0.02	-0.160[b]	-0.085	-0.123	c	c	-	-
			0.05	-0.160[b]	-0.080	-0.123	c	c	-	-
			0.10	-0.150	-0.080	-0.125	-0.210	c	-	-
			0.15	-0.150	-0.080	-0.125	-0.210	c	-	-

a. Peak appears in the first scan itself.

b. Broad peak.

c. Peak absent.

first scan itself. A similar situation is observed when pH of the solution is lowered by the addition of an acid, which is discussed under 'protonation'.

C. Trapping of the Intermediate Cu(I) Species

In the case of Cu(II) dipeptide complexes also the generation of Cu(I) dipeptide complexes was established by trapping the short-lived intermediate species using bathocuproin reagent.[19-21] The electrochemically generated Cu(I) bathocuproin complex is identical with the chemically generated bathocuproin complex in its spectral and cyclic voltammetric characteristics. The details of the preparation of these complexes are given in the experimental section. Both types of Cu(I) bathocuproin complexes gave an irreversible reduction peak at -0.598 V in amyl alcohol/TEAP medium at HMDE. The formation of the Cu(I) bathocuproin complex by electrochemical means from the Cu(II) complex indicate the generation of Cu(I) species during the electrode process. These observations are very similar to those observed in the case of Cu(II) amino acid complexes discussed in the previous chapter. These experiments give additional support to the formulation of Cu(I) species as an intermediate in the electrode process.

D. Protonation

It has been shown by different methods that in aqueous media Cu(II) can form complexes with dipeptides in different stoichiometries depending on the pH conditions and the metal-to-ligand ratio.[22] A decrease in the pH from the neutral value should result in the conversion of Cu(II)L complex into other forms of different stoichiometry. Since the reduction potentials of any metal complex is directly related to the coordination pattern around the metal ion, any change in its coordination sphere should reflect in its cyclic voltammetric response. Hence it was decided to study the pH dependence of cyclic voltammograms of all the Cu(II) dipeptide complexes at various scan rates. The results are presented and discussed in comparison with the CV observed at neutral pH values. Our emphasis was more on the acid pH ranges and hence for convenience this section will be referred as 'protonation'.

A gradual decrease in the pH of the solution was achieved by the step-wise addition of small amounts (µl) of $HClO_4$, to the aqueous solution of the Cu(II) dipeptide complexes. As in the case of Cu(II) amino acid complexes, discussed in chapter 2, in the present case also we observed

severe complications in the CV response from buffer anions when buffer solution was used to obtain the required pH values. To avoid these complications addition of acid or base to lower or raise the pH of the test solutions was preferred. All dipeptide complexes behaved in a similar fashion on protonation. Cyclic voltammograms of Cu(Gly Ser) at various pH values are shown in Fig.3.4 and those of Cu(Gly Gly) are shown in Fig.3.5. The CV data on protonation for some of the complexes are given in Tables 3.6-3.11. The parameters in these tables have the usual significance.

The major changes on the CV profile occurring on protonation are the following. (i) Height of peak C and D decrease gradually and finally disappear at low pH values. (ii) Peak A begins to appear in the first scan itself as a broad peak. (iii) Peaks A and B increase in heights accompanied by a decrease in 'peak-broadening' of peak A as the pH decrease. (iv) Peaks A and B shift anodically as pH decreases and at very low pH values (<3.0) the peaks A and B become very similar to the corresponding peaks of free Cu(II) ions. (v) At intermediate pH values (5.3-4.1) an overlapping cathodic peak A^*, close to A, is apparent in Cu(Gly Ser) complex (Fig.3.4).

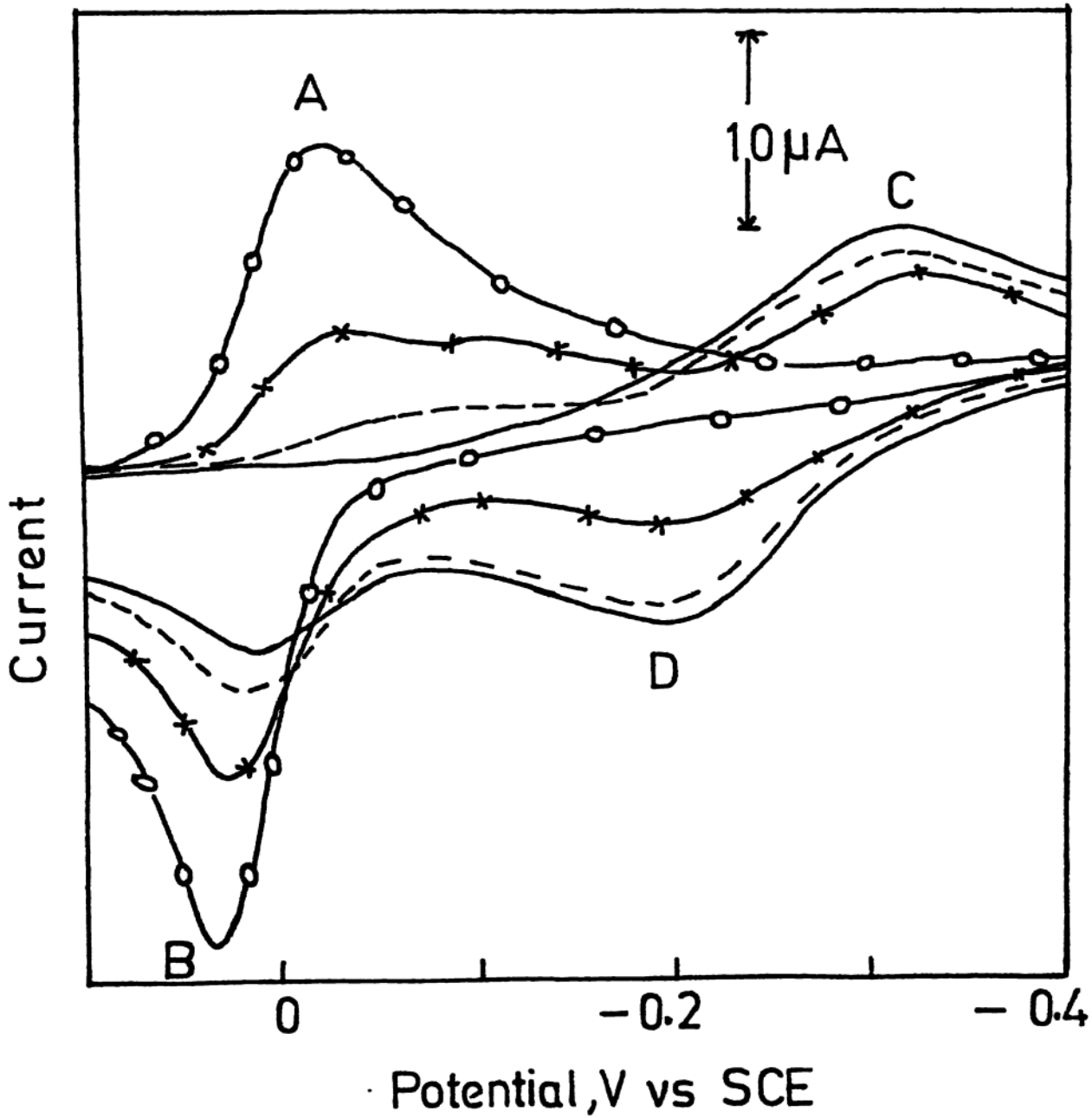


Fig.3.4 Cyclic voltammetric response on protonation of Cu(Gly Ser).
———, pH = 7.30; — — — pH = 6.38; —×—×— , pH = 4.81;
——o——o—— , pH = 3.77.

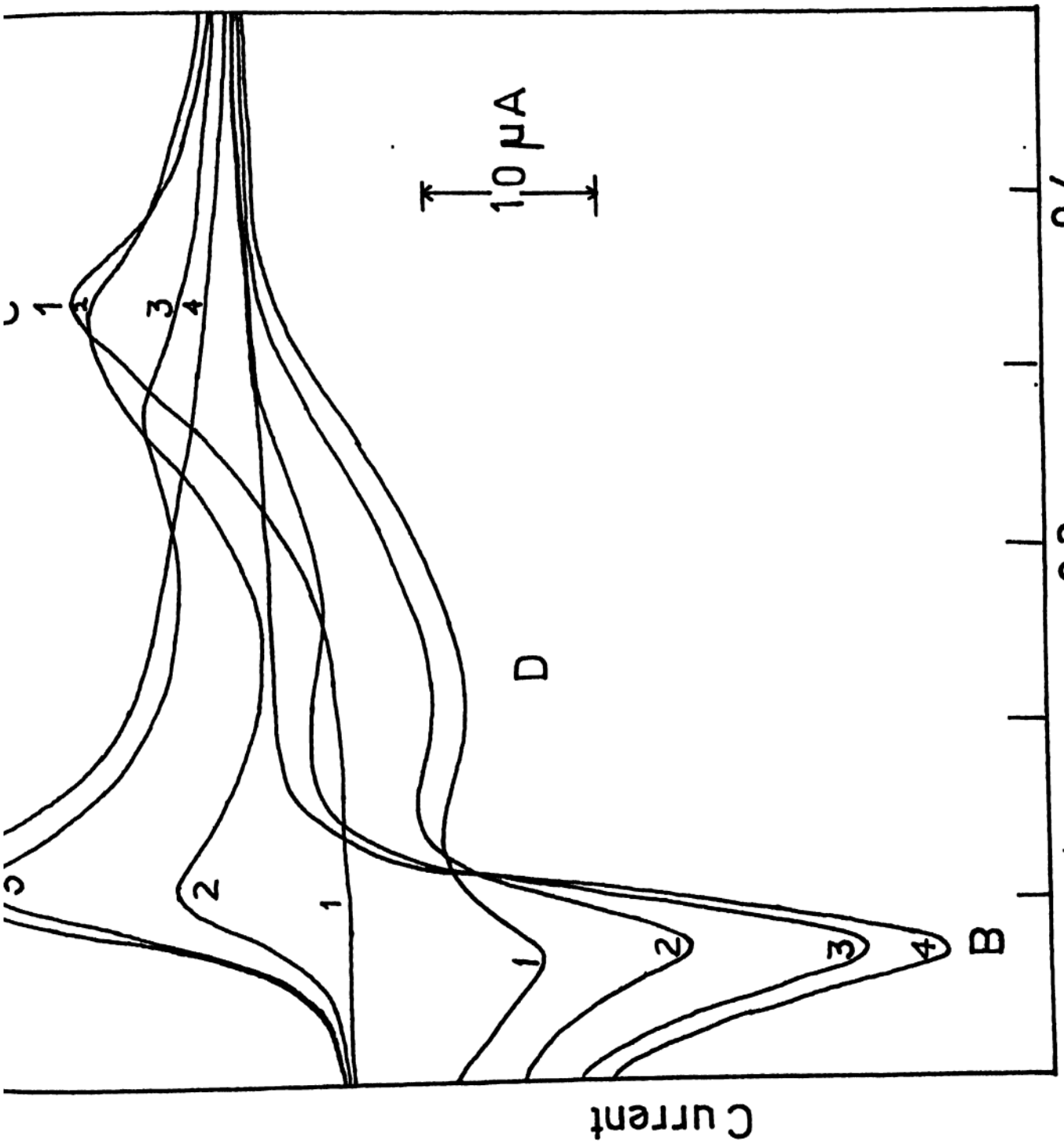


Fig.3.5 Cyclic voltammetric response on protonation of Cu(Gly Gly).

Table 3.6. Cyclic voltammetric data on protonation of Cu(Gly Ser) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	$E^{o'}_{C-D}$	i_p,A/$\nu^{1/2}$	i_p,B/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$	i_p,D/$\nu^{1/2}$
	0.02	c	0.015	-0.310	-0.205	-0.258	c	67.18	84.85	77.78
6.70	0.05	c	0.015	-0.320	-0.200	-0.260	c	56.28	82.73	80.50
	0.10	c	0.010	-0.320	-0.200	-0.260	c	48.13	82.22	81.23
	0.02	-0.070	0.020	-0.310	-0.205	-0.258	14.14	88.39	77.78	74.75
6.51	0.05	-0.080	0.015	-0.320	-0.200	-0.260	13.42	44.72	76.03	71.55
	0.10	-0.090	0.015	-0.315	-0.200	-0.258	15.81	38.11	75.89	69.57
	0.02	-0.070[b]	0.020	-0.310	-0.205	-0.258	21.21	95.46	70.71	71.71
6.38	0.05	-0.090[b]	0.020	-0.315	-0.195	-0.255	17.89	62.61	71.55	67.08
	0.10	-0.100[b]	0.025	-0.315	-0.190	-0.253	22.14	50.60	67.08	63.25

contd.

Table 3.6 contd.

	0.02	-0.050[b]	0.025	-0.305	-0.200	-0.253	35.36	127.28	63.64	56.57
5.32	0.05	[-0.040 / -0.095]	0.025	-0.315	-0.195	-0.260	35.78	89.44	55.90	51.43
	0.10	{-0.000 / -0.110}	0.030	-0.315	-0.190	-0.253	31.62	72.73	50.60	50.60
	0.02	-0.030[b]	0.030	-0.305	-0.200	-0.253	56.57	162.63	56.57	42.43
4.81	0.05	{-0.035 / -0.110}	0.030	-0.315	-0.190	-0.253	51.43	116.28	53.67	40.25
	0.10	{-0.035 / -0.110}	0.030	-0.315	-0.190	-0.253	41.11	101.19	44.24	37.95
	0.02	-0.015	0.030	-0.300	b	-	88.39	219.20	21.21	10.61
4.23	0.05	{-0.015 / -0.110}	0.030	-0.310	b	-	80.50	152.05	20.12	13.42
	0.10	{-0.015 / -0.110}	0.030	-0.310	b	-	75.89	139.14	20.30	12.23
	0.02	-0.010	0.035	c	c	-	113.14	261.63	c	c
3.25	0.05	-0.010	0.035	c	c	-	107.33	196.77	c	c
	0.10	-0.010	0.040	c	c	-	101.19	158.11	c	c

a Potentials expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.
b Broad peak.
c Peak absent in the first scan.

Table 3.7. Cyclic voltammetric data on protonation of Cu(Gly Gly) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D[b]	$E^{o'}_{,C-D}$	i_p,A/$\vartheta^{1/2}$	i_p,C/$\vartheta^{1/2}$
	0.02	c	0.030	-0.325	-0.150	-0.238	-	70.71
6.60	0.05	c	0.030	-0.330	-0.150	-0.240	-	71.55
	0.10	c	0.035	-0.340	-0.150	-0.245	-	70.76
	0.02	-0.010	0.030	-0.325	-0.150	-0.238	14.14	63.63
5.73	0.05	-0.015	0.030	-0.325	-0.150	-0.238	13.34	62.60
	0.10	-0.020	0.035	-0.340	-0.140	-0.240	14.23	63.25
	0.02	-0.010	0.030	-0.300	-0.140	-0.220	42.42	53.03
5.14	0.05	-0.015	0.030	-0.300	-0.140	-0.220	43.26	54.25
	0.10	-0.020	0.035	-0.310	-0.140	-0.225	42.85	52.45

contd.

Table 3.7 contd.

	0.02	-0.010[b]	0.030	-0.280	-0.150	-0.215	95.45	28.28
4.84	0.05	-0.015[b]	0.030	-0.290	-0.140	-0.215	96.25	23.38
	0.10	-0.020[b]	0.035	-0.310	-0.140	-0.225	93.45	27.21
	0.02	-0.010[b]	0.030	c	c	-	113.13	-
4.45	0.05	-0.015[b]	0.030	c	c	-	107.33	-
	0.10	-0.020[b]	0.035	c	c	-	105.42	-
	0.02	-0.010	0.030	c	c	-	120.20	-
4.13	0.05	-0.010	0.030	c	c	-	116.27	-
	0.10	-0.010	0.035	c	c	-	118.45	-

a. Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b. Broad peak.

c. Peak not observed in the first scan.

Table 3.8. Cyclic voltammetric data on protonation of Cu(Gly Phe) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	$E^{o'}_{C-D}$	i_p,A/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$
	0.02	c	c	-0.290	-0.150	-0.220	c	60.10
9.14	0.05	c	c	-0.300	-0.140	-0.220	c	60.37
	0.10	c	c	-0.310	-0.135	-0.223	c	63.25
	0.02	c	c	-0.280	-0.130	-0.205	c	56.57
8.02	0.05	c	c	-0.290	-0.140	-0.215	c	60.42
	0.10	c	c	-0.300	-0.120	-0.210	c	63.25
	0.02	c	0.030	-0.275	-0.130	-0.203	c	56.57
6.84	0.05	c	0.040	-0.280	-0.110	-0.195	c	58.14
	0.10	c	c	-0.290	-0.100	-0.195	c	56.92
	0.02	-0.050	0.030	-0.270	-0.120	-0.195	14.14	56.57
6.28	0.05	-0.050	0.040	-0.280	-0.110	-0.195	12.28	52.38
	0.10	c	c	-0.290	-0.100	-0.195	13.28	56.92

contd.

Table 3.8 contd.

	0.02	-0.030[b]	0.035	-0.250	-0.130[b]	-0.190	49.50	24.75
4.83	0.05	-0.035[b]	0.040	-0.240	-0.120[b]	-0.180	40.26	18.78
	0.10	-0.050[b]	0.050	-0.250	-0.100[b]	-0.175	42.69	11.07
	0.02	-0.020 -0.080	0.035	c	c	-	56.57	c
4.39	0.05	-0.020 -0.080	0.040	c	c	-	55.90	c
	0.10	-0.050[b]	0.050	c	c	-	56.92	c
	0.02	-0.020	0.035	c	c	-	56.86	c
3.80	0.05	-0.020	0.040	c	c	-	54.28	c
	0.10	-0.030	0.050	c	c	-	56.52	c

a. Potentials expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b. Broad peak.

c Peak absent in the first scan.

Table 3.9. Cyclic voltammetric data on protonation of Cu(Gly Thr) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D[b]	$E^{o'}_{C-D}$	i_p,A/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$
	0.02	c	0.040	-0.355	-0.150	-0.253	c	60.10
6.60	0.05	c	0.040	-0.340	-0.150	-0.248	c	58.20
	0.10	c	0.035	-0.355	-0.140	-0.248	c	59.16
	0.02	-0.005	0.030	-0.320	-0.130	-0.225	10.61	42.43
5.88	0.05	-0.010	0.030	-0.335	-0.120	-0.228	9.17	50.09
	0.10	-0.010	0.030	-0.340	-0.110	-0.225	12.65	41.11
	0.02	-0.005[b]	0.030	-0.320	-0.120	-0.220	21.21	35.36
5.55	0.05	-0.010[b]	0.035	-0.320	-0.120	-0.220	22.36	35.78
	0.10	-0.010[b]	0.040	-0.330	-0.110	-0.220	22.14	34.62
	0.02	-0.005[b]	0.030	-0.290	-0.120	-0.205	35.36	31.82
5.26	0.05	-0.010[b]	0.030	-0.305	-0.120	-0.208	31.30	31.30
	0.10	-0.010[b]	0.030	-0.305	-0.105	-0.205	34.79	34.79

contd.

Table 3.9 contd.

	0.02	-0.005	0.030	-0.280	-0.120	-0.200	49.50	28.28
5.06	0.05	-0.010	0.030	-0.295	-0.120	-0.208	46.96	24.60
	0.10	-0.010	0.035	-0.290	-0.105	-0.198	45.85	31.62
	0.02	-0.005	0.030	-0.240	-0.110	-0.175	63.64	17.68
4.88	0.05	-0.010	0.035	-0.265	-0.110	-0.175	60.37	17.89
	0.10	-0.010	0.035	-0.270	-0.105	-0.178	60.08	18.26
	0.02	-0.005	0.030	c	c	-	77.78	c
4.61	0.05	-0.010	0.035	c	c	-	76.03	c
	0.10	-0.010	0.040	c	c	-	74.31	c
	0.02	-0.005	0.035	c	c	-	84.85	c
4.32	0.05	-0.010	0.035	c	c	-	87.21	c
	0.10	-0.010	0.040	c	c	-	33.80	c

a. Potentials expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b. Broad peak.

c. Peak absent in the first scan

Table 3.10. Cyclic voltammetric data for protonation of Cu(Gly Met) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D[b]	$E^{o'}_{C-D}$	i_p,A/$\upsilon^{1/2}$	i_p,C/$\upsilon^{1/2}$
	0.02	c	0.035	-0.305	-0.150	-0.227	-	56.56
6.55	0.05	c	0.035	-0.310	-0.150	-0.230	-	68.60
	0.10	c	0.040	-0.320	-0.150	-0.235	-	56.92
	0.02	-0.010	0.035	-0.290	-0.130	-0.210	15.88	40.24
5.67	0.05	-0.010	0.035	-0.300	-0.150	-0.225	16.76	42.43
	0.10	-0.010	0.040	-0.310	-0.150	-0.230	14.23	44.27
	0.02	-0.010	0.035	-0.275	-0.130	-0.203	34.26	28.23
5.01	0.05	-0.020[b]	0.035	-0.295	-0.150	-0.223	35.77	29.06
	0.10	-0.015[b]	0.040	-0.290	-0.150	-0.220	38.26	27.12

contd.

Table 3.10 contd.

	0.02	-0.010	0.035	-0.260	-0.130	-0.195	48.12	-
4.94	0.05	-0.015[b]	0.035	-0.270	-0.150	-0.210	49.19	-
	0.10	-0.015[b]	0.040	-0.285	-0.150	-0.218	47.35	-
	0.02	-0.010	0.035	c	c	-	70.26	-
4.38	0.05	-0.010	0.035	c	c	-	71.55	-
	0.10	-0.015	0.040	c	c	-	69.57	-
	0.02	-0.010	0.035	c	c	-	75.38	-
3.91	0.05	-0.010	0.035	c	c	-	76.02	-
	0.10	-0.010	0.040	c	c	-	74.26	-

a. Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b. Broad peak.

c. Peak absent in the first scan.

Table 3.11. Cyclic voltammetric data for protonation of Cu(Met Gly) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D[b]	$E^{o'}_{C-D}$	i_p,A/$\gamma^{1/2}$	i_p,C/$\gamma^{1/2}$
	0.02	c	0.040	-0.310	-0.160	-0.235	c	77.78
6.62	0.05	c	0.040	-0.310	-0.160	-0.235	c	76.03
	0.10	c	0.040	-0.320	-0.160	-0.240	c	79.06
	0.02	-0.010	0.035	-0.300	-0.130	-0.215	12.73	63.64
5.48	0.05	-0.010	0.040	-0.300	-0.130	-0.215	13.42	67.08
	0.10	-0.010	0.040	-0.310	-0.140	-0.225	11.49	70.33
	0.02	-0.020	0.040	-0.300	-0.140	-0.220	28.28	60.10
5.17	0.05	-0.020	0.040	-0.300	-0.130	-0.215	26.83	62.61
	0.10	-0.020	0.035	-0.310	-0.120	-0.215	25.30	56.92
	0.02	-0.020	0.040	-0.265[b]	-0.130	-0.198	38.89	49.96
5.09	0.05	-0.025	0.035	-0.270[b]	-0.120	-0.195	40.25	44.25
	0.10	-0.030	0.035	-0.280[b]	-0.120	-0.200	37.95	44.27

contd.

Table 3.11 contd.

	0.02	-0.020	0.035	-0.260[b]	-0.130	-0.195	56.57	38.89
4.93	0.05	-0.025	0.040	-0.260[b]	-0.120	-0.190	53.67	53.67
	0.10	-0.030[b]	0.040	-0.260[b]	-0.120	-0.190	56.52	34.79
	0.02	-0.020	0.035	-0.250	-0.130	-0.190	70.71	21.21
4.28	0.05	-0.025	0.040	-0.250	-0.110	-0.180	71.55	44.72
	0.10	-0.030	0.035	-0.245	-0.110	-0.178	66.41	31.62
	0.02	-0.015	0.035	-0.245	-0.130	-0.187	91.92	14.14
4.17	0.05	-0.020	0.030	-0.245	-0.110	-0.177	86.97	17.89
	0.10	-0.025	0.035	-0.240	-0.115	-	88.54	18.23
	0.02	-0.010	0.035	c	c	-	113.14	c
3.86	0.05	-0.015	0.030	c	c	-	111.80	c
	0.10	-0.015	0.040	c	c	-	109.36	c

a. Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b. Broad peak.

c. Peak absent in the first scan.

Decrease in the peak heights of C and D results because of the decrease in concentration of Cu(II)L species due to its conversion to other forms on protonation. It is well documented that protonation first occurs at the peptide nitrogen followed by a rearrangement which ultimately results in the formation of the protonated species $[Cu(II)LH]^+$. On further protonation it dissociates to free Cu(II) ions. Thus as the pH decreases from neutral value, $[Cu(II)LH]^+$ and Cu(II) ions successively become the major species in solution. (The chemistry of the protonation of the Cu(II) dipeptide complexes is discussed at length in chapter 1) Therefore it is natural to observe a decrease in C, D peak heights as the pH is lowered. It is also possible that the protonated species, $[Cu(II)LH]^+$, can undergo reduction at a potential less cathodic to peak C. The initial 'peak-broadening' observed for the peak A can be due to the overlap of the reduction wave (A) of free Cu(II) ions and the reduction peak (A*) of the $[Cu(II)LH]^+$, when both are relatively at low concentrations. At intermediate pH region (5.3-4.1) the concentration of $[Cu(II)LH]^+$ increases which in turn helps the broad peak A, to get resolved into peaks A and A*. A further decrease in pH catalyses the dissociation of $[Cu(II)LH]^+$ to free Cu(II) ions. This is reflected in the absence of the peak A^* and in the increased peak heights

of A and B at low pH values. In most of the Cu(II) dipeptide complexes, the peak A is observed as a broad peak as in the case of Cu(Gly Gly). But only in Cu(Gly Ser) complex the broad peak A gets resolved into A and A^{*}. The appearance of peak A from the first scan itself, shows the availability of free Cu(II) ions in solution liberated by protonation. In neutral pH conditions they have to be generated electrochemically and hence peak A appears only from the second scan onwards.

The reversibility of protonation is monitored by raising the pH of the solution from a very low value of about 3.0 to about 7.5 by the addition of NaOH and CV were recorded at every stage. As a representative case, cyclic voltammograms of Cu(Gly Ser) on deprotonation is shown in Fig.3.6 and the relevant CV data for some of the complexes are given in Tables 3.12-3.17. As the pH is increased peak A and B decrease in heights and peaks C and D start appearing (reverse of protonation). The entire cycle of protonation-deprotonation can be repeated several times showing the reversibility of protonation.

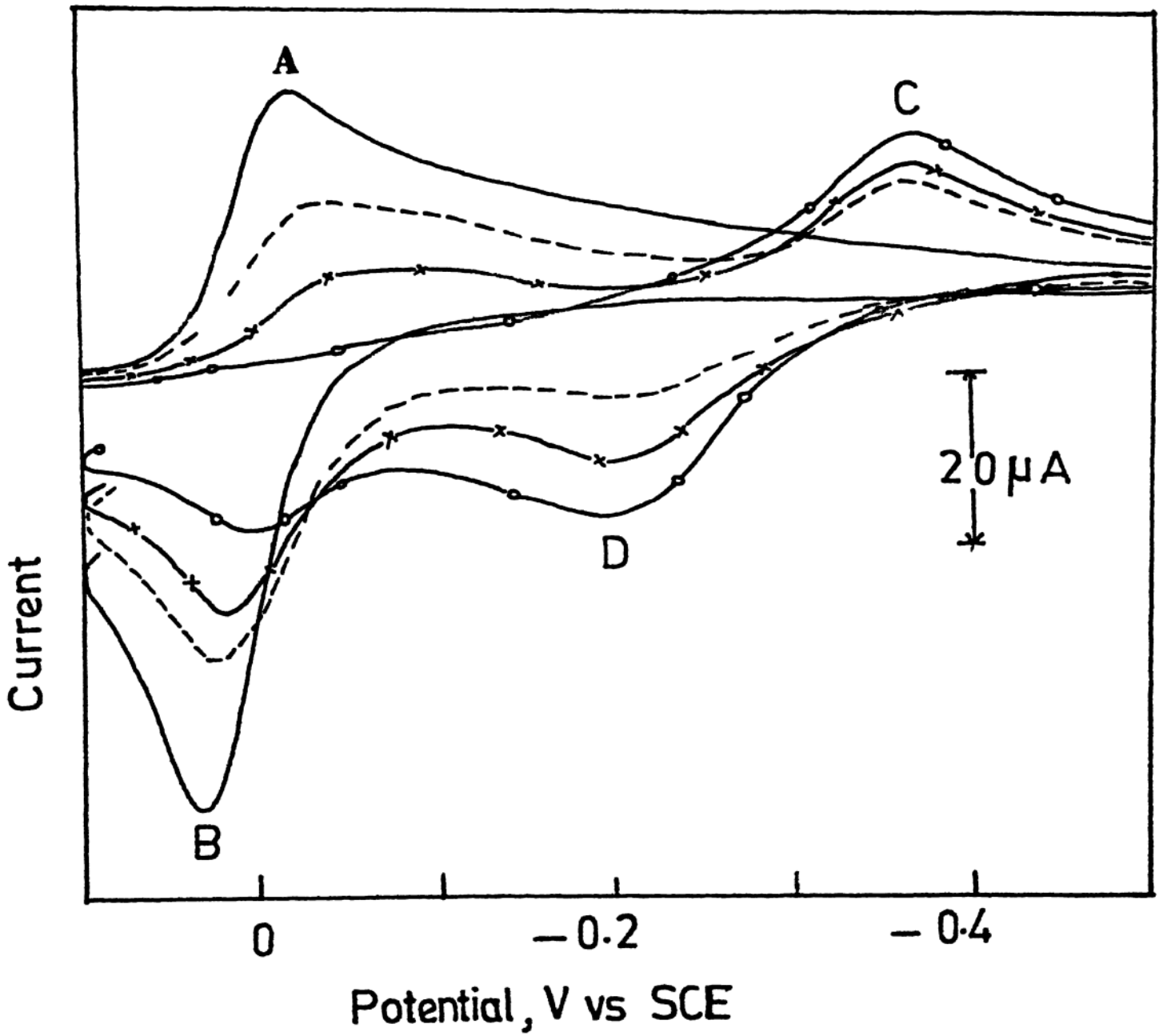


Fig.3.6 Cyclic voltammetric response on deprotonation of Cu(Gly Ser).
——, pH = 3.77; — — —, pH = 4.48; —x—x— pH = 5.24;
—o—o—, pH = 7.32.

Table 3.12. Cyclic voltammetric data on deprotonation of Cu(Gly Ser) in aqueous media[a].

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	$E^{o'}_{C-D}$	i_p,A/$\nu^{1/2}$	i_p,B/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$	i_p,D/$\nu^{1/2}$
	0.02	-0.010	0.035	c	c	-	113.14	261.63	c	c
3.25	0.05	-0.010	0.035	c	c	-	107.33	196.77	c	c
	0.10	-0.010	0.040	c	c	-	101.19	158.11	c	c
	0.02	-0.015	0.030	-0.305	-0.200	-0.253	98.99	247.49	c	c
4.20	0.05	-0.020	0.035	-0.310	-0.205	-0.258	89.44	192.30	13.42	8.94
	0.10	-0.020 -0.120	0.035	-0.305	-0.205	-0.255	88.54	145.46	15.81	9.49
	0.02	-0.030[b]	0.020	-0.310	-0.205	-0.258	67.18	162.63	42.43	35.36
4.48	0.05	-0.040[b]	0.020	-0.310	-0.205	-0.258	82.61	120.75	40.25	33.54
	0.10	-0.035 -0.110	0.020	-0.310	-0.205	-0.258	60.08	107.52	37.96	34.79

contd.

Table 3.12 contd.

	0.02	-0.040[b]	0.020	-0.310	-0.205	-0.258	56.57	155.56	56.57	42.43
4.89	0.05	-0.050[b]	0.020	-0.310	-0.205	-0.258	49.19	111.80	53.67	42.49
	0.10	{-0.050[b] / -0.120}	0.010	-0.310	-0.200	-0.255	44.27	88.54	50.60	41.11
	0.02	-0.050[b]	0.015	-0.310	-0.205	-0.258	42.43	127.28	63.64	56.57
5.24	0.05	-0.060[b]	0.010	-0.310	-0.205	-0.258	40.25	91.68	49.19	49.19
	0.10	{-0.060[b] / -0.110}	0.015	-0.310	-0.200	-0.255	37.55	75.89	56.92	53.76
	0.02	-0.045[b]	0.020	-0.315	-0.205	-0.260	28.28	106.07	63.64	56.57
5.64	0.05	-0.050[b]	0.020	-0.315	-0.205	-0.260	31.30	120.21	58.14	58.14
	0.10	-0.060[b]	0.010	-0.310	-0.200	-0.255	18.97	56.60	69.57	63.25
	0.02	-0.060[b]	0.020	-0.315	-0.205	-0.260	21.21	84.85	91.92	70.71
5.96	0.05	-0.070[b]	0.020	-0.315	-0.205	-0.260	22.36	62.61	80.50	67.08
	0.10	-0.080[b]	0.010	-0.310	-0.200	-0.255	c	50.60	82.22	63.25

contd.

Table 3.12 contd.

	0.02	-0.080[b]	0.010	-0.315	-0.205	-0.260	10.61	74.25	70.71	70.71
6.79	0.05	-0.090[b]	0.010	-0.310	-0.205	-0.257	c	62.61	80.50	67.08
	0.10	-0.090[b]	0.010	-0.305	-0.200	-0.253	c	88.54	82.22	81.23
	0.02	c	0.010	-0.315	-0.200	-0.257	c	49.50	84.85	77.78
7.68	0.05	c	0.010	-0.315	-0.200	-0.257	c	42.49	71.55	71.55
	0.10	c	0.010	-0.300	-0.200	-0.250	c	c	70.95	70.23

a Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b Broad peak

c Peak absent in the first scan.

Table 3.13. Cyclic voltammetric data on deprotonation of Cu(Gly Gly) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	$E^{o'}_{C-D}$[b]	i_p,A/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$
	0.02	-0.010	0.030	c	c	-	120.20	-
4.13	0.05	-0.010	0.030	c	c	-	116.27	-
	0.10	-0.010	0.035	c	c	-	118.45	-
	0.02	-0.010	0.030	c	c	-	113.13	-
4.39	0.05	-0.015	0.030	c	c	-	107.33	-
	0.10	-0.020	0.035	c	c	-		-
	0.02	-0.010	0.030	-0.240	-0.150	-0.185	98.99	7.07
4.54	0.05	-0.015	0.030	-0.240	-0.150	-0.195	93.91	8.94
	0.10	-0.020	0.035	-0.260	-0.150	-0.205		
	0.02	-0.010	0.030	-0.250	-0.150	-0.200	84.85	21.21
4.81	0.05	-0.015	0.030	-0.250	-0.150	-0.200	84.97	20.12
	0.10	-0.020	0.035	-0.290	-0.150	-0.220		

contd.

Table 3.13 contd.

	0.02	-0.010[b]	0.030	-0.295	-0.150	-0.223	56.56	49.49
5.13	0.05	-0.015[b]	0.030	-0.295	-0.150	-0.223	53.65	45.24
	0.10	-0.020[b]	0.035	-0.320	-0.150	-0.235	54.85	47.42
	0.02	-0.010[b]	0.030	-0.320	-0.150	-0.235	35.35	56.56
5.49	0.05	-0.015[b]	0.030	-0.320	-0.150	-0.235	35.12	54.26
	0.10	-0.020[b]	0.035	-0.340	-0.150	-0.245	34.35	55.16
	0.02	c	0.030	-0.330	-0.150	-0.240	-	84.85
6.35	0.05	c	0.030	-0.330	-0.150	-0.240	-	86.21
	0.10	c	0.035	-0.350	-0.150	-0.250	-	82.13

a. Potentials expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b. Broad peak.

c. Peak absent in the first scan.

Table 3.14. Cyclic voltammetric data on deprotonation of Cu(Gly Phe) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	$E^{o'}_{C-D}$	i_p,A/$\upsilon^{1/2}$	i_p,C/$\upsilon^{1/2}$
	0.02	-0.020	0.035	c	c	-	56.86	c
3.80	0.05	-0.020	0.040	c	c	-	54.24	c
	0.10	-0.030	0.050	c	c	-	56.92	c
	0.02	-0.040[b]	0.040	c	c	-	56.57	c
4.61	0.05	-0.050[b]	0.045	c	c	-	53.43	c
	0.10	-0.060[b]	0.050	c	c	-	52.18	c
	0.02	-0.040[b]	0.040	-0.265	-0.130	-0.193	31.82	28.36
5.08	0.05	-0.060[b]	0.045	-0.260	-0.120	-0.190	31.30	26.83
	0.10	-0.060[b]	0.050	-0.265	-0.110[b]	-0.188	31.62	25.30
	0.02	-0.060[b]	0.040	-0.275	-0.140	-0.207	5.26	53.03
6.01	0.05	-0.070[b]	0.045	-0.280	-0.120[b]	-0.200	5.16	51.43
	0.10	-0.080[b]	0.050	-0.285	-0.110[b]	-0.197	4.64	50.60

a. Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b. Broad peak; c. Peak absent in the first scan.

Table 3.15. Cyclic voltammetric data on deprotonation of Cu(Gly Thr) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D [b]	$E^{o'}_{C-D}$	i_p,A/$\vartheta^{1/2}$	i_p,C/$\vartheta^{1/2}$
	0.02	-0.010	0.035	c	c	-	88.39	c
4.32	0.05	-0.015	0.035	c	c	-	89.44	c
	0.10	-0.020	0.040	c	c	-	83.80	c
	0.02	-0.010	0.030	c	c	-	74.25	c
4.69	0.05	-0.015	0.035	c	c	-	71.55	c
	0.10	-0.020	0.040	c	c	-	69.57	c
	0.02	-0.010	0.030	-0.270	-0.130	-0.200	56.57	28.28
4.89	0.05	-0.010	0.035	-0.270	-0.120	-0.195	58.14	35.78
	0.10	-0.020	0.040	-0.275	-0.125	-0.200	55.34	22.14

contd.

Table 3.15 contd.

	0.02	-0.010	0.030	-0.280	-0.110	-0.195	42.43	31.82
5.39	0.05	-0.015	0.030	-0.310	-0.110	-0.210	42.49	29.83
	0.10	-0.020	0.040	-0.310	-0.120	-0.215	41.11	28.46
	0.02	-0.010[b]	0.030	-0.315	-0.120	-0.218	28.28	39.60
5.89	0.05	-0.010[b]	0.030	-0.320	-0.120	-0.220	26.83	38.01
	0.10	-0.025[b]	0.035	-0.320	-0.120	-0.220	22.82	40.21
	0.02	c	0.035	-0.320	-0.160	-0.240	c	49.50
6.24	0.05	c	0.035	-0.320	-0.160	-0.240	c	51.67
	0.10	c	0.035	-0.320	-0.160	-0.240	c	50.26

a. Potentials are expressed in volts and current in $\mu A(V/s)^{-1/2}$.

b. Broad peak

c. Peak absent in the first scan.

Table 3.16. Cyclic voltammetric data on deprotonation of Cu(Gly Met) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D [b]	$E^{o'}_{C-D}$	i_p,A/$\vartheta^{1/2}$	i_p,C/$\vartheta^{1/2}$
	0.02	-0.010	0.035	c	c	-	75.38	-
3.91	0.05	-0.010	0.035	c	c	-	76.02	-
	0.10	-0.010	0.040	c	c	-	74.26	-
	0.02	-0.010	0.035	c	c	-	67.08	-
4.36	0.05	-0.015	0.035	c	c	-	66.28	-
	0.10	-0.020	0.040	c	c	-	66.40	-
	0.02	-0.010	0.035	-0.270	-0.130	-0.200	54.26	13.56
4.94	0.05	-0.015	0.035	-0.280	-0.150	-0.215	53.66	13.41
	0.10	-0.020	0.040	-0.290	-0.150	-0.220	56.92	14.23

contd.

Table 3.16 contd.

	0.02	-0.010[b]	0.035	-0.285	-0.130	-0.207	40.34	24.36
5.10	0.05	-0.015[b]	0.035	-0.290	-0.150	-0.220	42.48	22.36
	0.10	-0.020[b]	0.040	-0.300	-0.150	-0.225	41.10	25.29
	0.02	-0.015[b]	0.035	-0.295	-0.150	-0.223	27.26	39.26
5.60	0.05	-0.020[b]	0.035	-0.300	-0.150	-0.225	26.83	40.24
	0.10	-0.020[b]	0.040	-0.300	-0.150	-0.225	28.46	37.94
	0.02	c	0.035	-0.300	-0.150	-0.225	12.64	48.16
6.34	0.05	c	0.035	-0.300	-0.150	-0.225	13.41	49.19
	0.10	c	0.040	-0.305	-0.150	-0.228	12.64	48.10

a. Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b. Broad peak.

c. Peak absent in the first scan.

Table 3.17. Cyclic voltammetric data for deprotonation of Cu(Met Gly) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D [b]	$E^{o'}_{C-D}$	i_p,A/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$
	0.02	-0.010	0.035	c	c	-	113.14	c
3.86	0.05	-0.015	0.030	c	c	-	111.80	c
	0.10	-0.015	0.040	c	c	-	109.36	c
	0.02	-0.015	0.035	c	c	-	113.14	c
4.33	0.05	-0.015	0.040	c	c	-	110.80	c
	0.10	-0.020	0.045	c	c	-	107.52	c
	0.02	-0.015	0.035	-0.205[b]	-0.130	-0.167	98.99	14.14
4.46	0.05	-0.015	0.040	-0.210	-0.130	-0.170	93.91	20.12
	0.10	-0.020	0.045	-0.220	-0.130	-0.175	88.54	18.97

contd.

Table 3.17 contd.

	0.02	-0.010	0.035	-0.230[b]	-0.130	-0.180	77.78	49.50
4.85	0.05	-0.015	0.040	-0.235	-0.120	-0.178	76.03	35.28
	0.10	-0.020	0.045	-0.245	-0.120	-0.183	75.89	44.27
	0.02	-0.010	0.035	-0.275[b]	-0.130	-0.203	45.96	56.57
5.00	0.05	-0.010	0.040	-0.270	-0.130	-0.200	40.25	58.14
	0.10	-0.020	0.045	-0.285[b]	-0.120	-0.203	44.27	56.52
	0.02	-0.015	0.035	-0.300	-0.160	-0.230	14.14	70.71
5.44	0.05	-0.010	0.040	-0.290	-0.140	-0.215	15.65	67.08
	0.10	-0.020	0.045	-0.310	-0.100	-0.205	12.65	63.25
	0.02	c	0.035	-0.310	-0.170	-0.240	c	91.92
6.99	0.05	c	0.040	-0.310	-0.160	-0.235	c	89.44
	0.10	c	0.040	-0.320	-0.160	-0.240	c	88.54

a. Potentials expressed in volts and current functions in $\mu A(V/s)^{-1/2}$.

b. Broad peak.

c. Peak absent in the first scan.

3.4 Experimental

Chemicals

Cu(II) dipeptide complexes were prepared from basic copper carbonate and the corresponding dipeptides. Basic copper carbonate was used after estimating the copper content gravimetrically.[19] All the dipeptides were obtained from Sigma Chemicals Co., U.S.A. Mercury was purified and distilled. Analar grade sodium perchlorate was used. Doubly distilled water was used throughout the studies.

The Cu(II) dipeptide complexes were prepared by employing a method reported earlier.[8d] Stoichiometric amounts of the dipeptide (1 mM) was added to a suspension of copper carbonate (1 mM) in hot water with stirring. The stirring was continued for 1-2 hrs at 50-60°C and then the mixture was filtered to remove the unreacted copper carbonate. The resulted bluish-violet solution was concentrated by slow evaporation after checking the pH (the pH was adjusted to near neutral values whenever necessary). The concentrated solution after 3-4 days yielded bluish-violet crystals. They were separated by filtration, washed several times with water and dried at 110-110°C in vacuum.

The intermediate Cu(I) species generated during the electrode process of the Cu(II) dipeptide complexes was trapped by using bathocuproin reagent. The details of this experiment is given in chapter 2.

Cyclic voltammetric experiments were carried out in a three-electrode cell with a metrohm E410 hanging mercury drop electrode (HMDE) as working electrode. Other solid electrodes like, platinum-disc or glassy-carbon etc., do not show any CV response for these complexes. Platinum wire was used as auxiliary electrode. All the potentials reported here are referred to saturated calomel electrode (SCE) and are uncorrected for liquid-junction potentials. The details of the experimental set up is given in chapter 2.

3.5 References

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CHAPTER 4

TERNARY COPPER(II) AMINO ACID AND DIPEPTIDE COMPLEXES. REDUCTION IN AQUEOUS MEDIA*

4.1 Abstract

The reduction of three types of ternary Cu(II) complexes is studied by cyclic voltammetry at HMDE in aqueous media. Ternary Cu(II) complexes containing two optically active α-amino acids with oppositely charged groups in their side chains have been studied in detail. Reduction of these complexes proceeds via a one-electron reversible process generating the corresponding Cu(I) species, and is responsible for the C-D couple. Cu^{o} is

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*Part of the work discussed in this chapter will appear in
(i) Transition Met. Chem. (in Press)
(ii) Polyhedron (in press) and
(iii) Indian J. Chem. (in Press).

generated from the unstable Cu(I) species and it undergoes a two-electron oxidation displaying the anodic peak B. The cathodic peak A corresponds to the reduction of the electrogenerated Cu(II) ions and appears from the second scan onwards. The overall electrode process and the CV response of these type of complexes are very similar to those of binary Cu(II) amino acid complexes and belongs to an ECE mechanism. Cyclic voltammetric response on protonation of these complexes is also studied.

The reduction of another type of complexes Cu(BPY)(A) type, where BPY is 2,2'-bipyridyl and A is an amino acid, is also investigated. They also undergo a one-electron reduction generating the Cu(I) species. The stability of the copper(I) species appears to be higher than that of the binary amino acid complexes. The reduction sequence is similar to that of the first type of ternary systems. The CV response on protonation of these ternary complexes is also investigated. Unlike in the case of Cu(II) binary amino acid complexes, these ternary complexes yielded the 1:1 Cu(II) bipyridyl complex, $Cu(BPY)(H_2O)_2$ as the end-product on protonation.

The histidine containing mixed amino acid and dipeptide complexes form the third group of complexes studied in this chapter. The histidine containing ternary

Cu(II) amino acid complexes have totally different cyclic voltammetric behaviour from hitherto discussed pattern. They display a relatively sharp peak at about -0.350 V which is identified as due to an adsorption-controlled process. The presence of histidine moiety in the complex seems to be responsible for the adsorption process. The histidine containing Cu(II) dipeptide complexes undergo a one-electron reduction generating an intermediate Cu(I) species which subsequently decomposes to Cu^{o}. The mechanism of these processes is discussed. The cyclic voltammograms of these complexes are not complicated by adsorption process, even though at fast scan rates an adsorption-controlled wave is observed at 50-80 mV cathodic to the original reduction peak.

4.2 Introduction

In enzyme-metal-substrate (EMS) complexes formed in enzymatic reactions involving metal ions, non-covalent interactions between enzyme and substrate molecules around the central metal ion are essential for the efficiency and specificity of the reactions.[1,2] The existence of such noncovalent interactions prompted the synthesis and study of ternary amino acid copper(II) complexes with intra molecular ligand-ligand interactions.[3-5] The

stability of such ternary complexes is much higher than that of the corresponding binary complexes. Several ternary Cu(II) amino acid complexes with amino acids having oppositely charged side chains are synthesised and characterised both in solution and solid state.[3-6] We have extended our cyclic voltammetric studies on the reduction to these interesting class of compounds in the hope that they might generate more stable Cu(I) species than the binary Cu(II) amino acid complexes. The results are presented and discussed in this chapter.

Ternary complexes of Cu(II), containing bidentate aromatic nitrogen base such as 2,2'-bipyridyl (BPY) and a bidentate oxygen donor ligand have been shown to possess unusual stabilities.[7,8] When the bidentate oxygen donor ligand is replaced by a 'mixed' ligand such as glycine, containing O and N donor atoms, the stability of the resultant ternary complex is reduced but it is still higher than that of the bidentate N donor ligand.[9] Ternary complexes of the type Cu(BPY)(A), where A is an amino acid anion, have been studied extensively by many workers.[10,1] But its redox chemistry is not yet studied well, except for a brief polarographic study on the reduction of these complexes in methanol.[11] In this chapter we discuss our results on the reduction of these complexes.

Mixed amino acid and peptide Cu(II) complexes containing histidine have received considerable attention because of their occurrence and involvement in the transport of Cu(II) ions in biological systems.[12,13] Several ternary amino acid Cu(II) complexes containing histidine have been synthesised and characterized in both solution and in solid state.[14-18] It is observed by many workers that histidine containing ternary Cu(II) complexes of amino acids behave differently from the rest of the ternary complexes in its electronic and ESR spectral aspects and hence are generally discussed as a separate group.[18] Ternary Cu(II) dipeptide complexes containing histidine is also attracted wide attention because it offers a chance to study the peptide-nitrogen coordination to Cu(II) in presence of histidyl group. In many of the biological redox systems, in which Cu(II) is involved, the copper is bound by histidyl-nitrogens and peptide-nitrogens.[19] In this chapter the results of the studies on the reduction of histidine containing ternary Cu(II) amino acid and dipeptide complexes are also discussed.

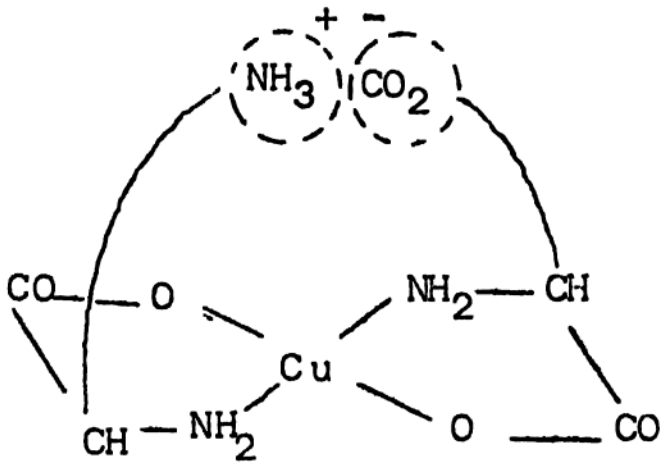
4.3 Results and Discussion

Part-1 : Copper(II) Amino Acid (A) Amino Acid (B) Type

A. Cyclic Voltammograms at Neutral pH Conditions

The mixed amino acid Cu(II) complexes discussed here are of the general formula Cu(A)(B) where A and B

represent two optically active α-amino acids containing oppositely charged groups in their side chains. Their structure is shown in 4.1.



Cu(L-A)(L-B)

TRANS

4.1

The complexes studied are listed in Table 4.1. The complexes are isolated as solids and purified by chromatographic techniques, the details are discussed in the experimental section. Cyclic voltammograms were recorded in the potential range, +0.1 to -0.6 V versus SCE, at HMDE of definite size, in aqueous solutions of the complex (~ 0.6 mM) with $NaClO_4$ as supporting electrolyte at a pH about 7.5. All complexes showed very similar CV response and they closely resemble the CV's of binary Cu(II) amino acid complexes discussed in chapter 2. Hence the discussion on the CV response is in the same lines as that in

Table 4.1. Cyclic voltammetric data on the reduction of ternary copper(II) amino acid complexes in aqueous media.[a]

Compound	Sweep rate (Vs^{-1})	E_p,A[b]	E_p,B	E_p,C	E_p,D	$\Delta E_{p\ C-D}$	$E^{o'}_{C-D}$
Cu(Asp)(Lys)	0.02	-0.035	0.020	-0.265	-0.195	0.070	-0.230
	0.05	-0.040	0.020	-0.265	-0.195	0.070	-0.230
	0.10	-0.040	0.025	-0.270	-0.190	0.080	-0.230
	0.20	c	c	-0.270	-0.185	0.085	-0.228
Cu(Asp)(Arg)	0.01	-0.040	0.020	-0.250	-0.180	0.070	-0.215
	0.02	-0.040	0.020	-0.250	-0.175	0.075	-0.210
	0.05	-0.040	0.010	-0.245	-0.170	0.075	-0.208
	0.10	c	c	-0.250	-0.165	0.085	-0.208
	0.20	c	c	-0.250	-0.160	0.090	-0.205

contd.

Table 4.1 contd.

Cu(Asp)(Orn)	0.02	-0.030	0.025	-0.255	-0.190	0.065	-0.228
	0.05	-0.040	0.025	-0.260	-0.185	0.075	-0.228
	0.10	c	c	-0.260	-0.185	0.075	-0.228
	0.20	c	c	-0.265	-0.180	0.085	-0.228
Cu(Glu)(Lys)	0.01	-0.030	0.030	-0.240	-0.180	0.060	-0.210
	0.02	-0.030	0.025	-0.240	-0.180	0.060	-0.210
	0.05	-0.030	0.020	-0.250	-0.180	0.070	-0.215
	0.10	c	c	-0.250	-0.180	0.070	-0.215
	0.20	c	c	-0.255	-0.175	0.080	-0.210
Cu(Glu)(Arg)	0.01	-0.030	0.030	-0.250	-0.180	0.070	-0.215
	0.02	-0.030	0.025	-0.250	-0.180	0.070	-0.215
	0.05	-0.030	0.025	-0.255	-0.175	0.080	-0.215
	0.10	c	c	-0.255	-0.175	0.080	-0.215
	0.20	c	c	-0.260	-0.170	0.090	-0.215

contd.

Table 4.1 contd.

	0.01	-0.040	0.020	-0.240	-0.160	0.080	-0.200
	0.02	-0.040	0.020	-0.245	-0.160	0.085	-0.202
Cu(Glu)(Orn)	0.05	-0.040	0.015	-0.250	-0.155	0.095	-0.203
	0.10	c	c	-0.250	-0.155	0.095	-0.203
	0.20	c	c	-0.255	-0.155	0.100	-0.205

a Potentials expressed in volts vs SCE; CV recorded at pH about 7.5 for all complexes, in the conc. range 0.62-0.8 mM.

b Peak appears only in the second scan.

c Peak absent.

chapter 2. The relevant CV data for all complexes are given in Table 4.1, the notations have the same meaning as in the tables of the previous chapters. Representative cyclic voltammogram of Cu(Glu)(Arg) is given in Fig.4.1b. None of the amino acid ligands in the absence of metal ions gives any peak in the potential range +1.0 to -1.0 V. There are no peaks for any of the Cu(II) complexes in the positive potential range even at electrodes other than HMDE.

In Fig. 4.1a the cyclic voltammogram of $Cu(Gly)_2$ is reproduced for comparison. The CV behaviour of free Cu(II) ions is given in Fig.4.1b. From a comparison of Figs.4.1a and 4.1b, it becomes clear that the C, D and A,B peaks of ternary complexes have a very close resemblance to those of $Cu(Gly)_2$. It is also clear that peaks A, B of both binary and ternary amino acid complexes are similar to the cathodic and anodic peaks of free Cu(II) ions in position and appearance. The origin of peaks C,D, A, and B are established for $Cu(Gly)_2$ type complexes. A similar reasoning holds for the origin of the peaks of these ternary complexes. The overall electrode process for these ternary amino acid Cu(II) complexes are summarised in equations 4.1 - 4.3.

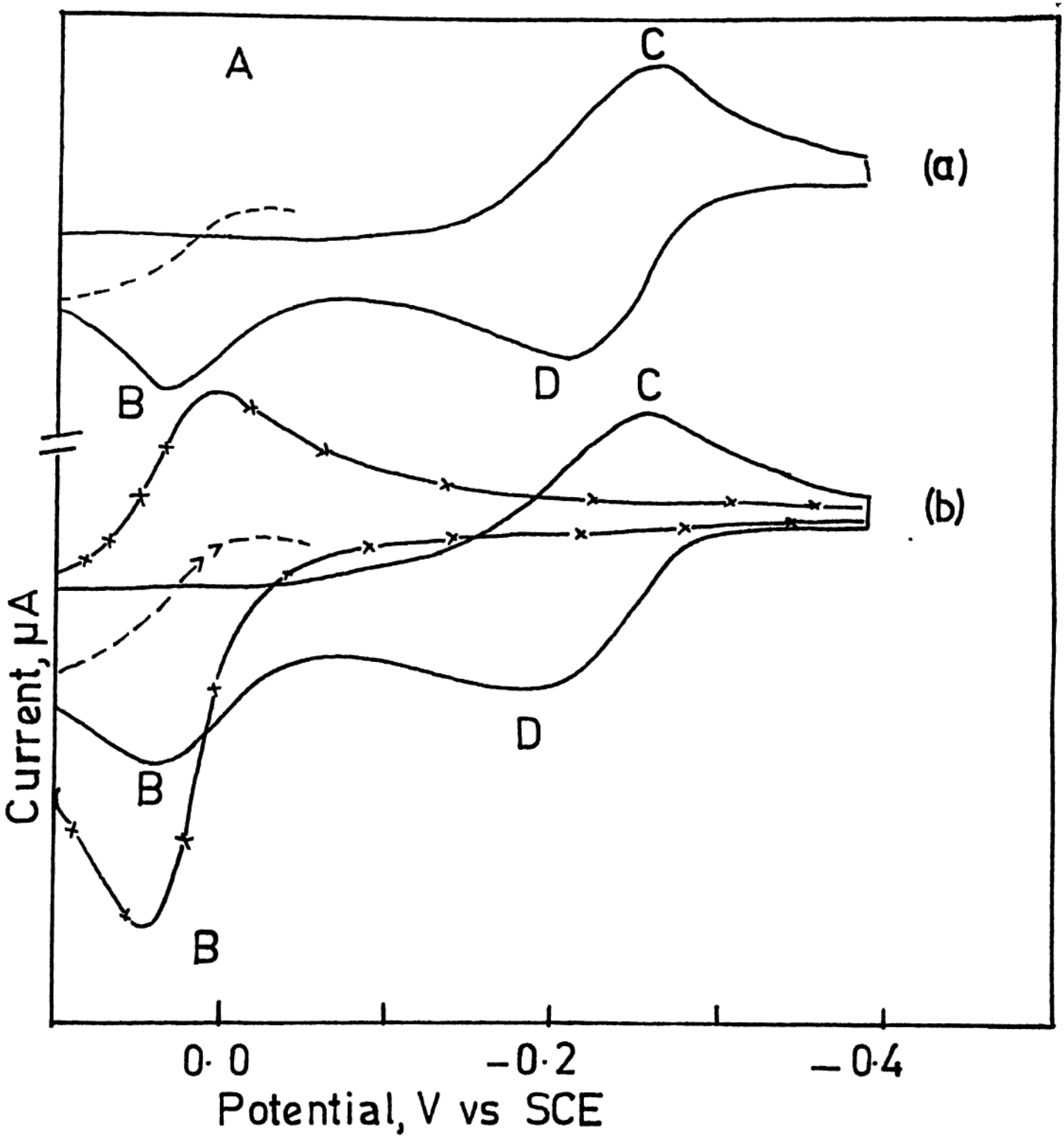


Fig.4.1 (a) Cyclic voltammogram of $Cu(Gly)_2$ at γ = 0.02 Vs^{-1}. (b) Cyclic voltammograms of (——), Cu(Glu)(Arg) at scan rate 0.02 Vs^{-1}; (—x—x—), $Cu(ClO_4)_2.6H_2O$ at scan rate 0.02 Vs^{-1}.

$$Cu(II)(A)(B) \xrightleftharpoons{1e} [Cu(I)(A)(B)] \quad \ldots 4.1$$

$$[Cu(I)(A)(B)] \xrightarrow{\text{chemical reactions}} Cu^{o}_{(Hg)} + A+B \quad \ldots 4.2$$

$$Cu^{o}_{(Hg)} \xrightleftharpoons{2e} Cu^{2+}_{(aq)} \quad \ldots 4.3$$

Equation 4.1 is responsible for the peaks C and D and equation 4.3 is responsible for A,B peaks. The $\triangle E_p$ values between peaks C and D are in the range 70-85 mV and show that the electron-transfer process is nearly reversible. The overall process is consistent with an ECE mechanism,[20] where E^{o}_{2} is anodic to E^{o}_{1}, $n_1 = 1$ and $n_2 = 2$, both the electron-transfer steps being nearly reversible.

Equation 4.1, in the overall electrode process, generates the ternary Cu(I) amino acid complexes. These ternary copper(I) complexes are also short-lived as in the case of the binary complexes (chapter 2). The observed CV response for the ternary complexes shows that the intramolecular ligand-ligand interactions from the side chains of the amino acids do not have any appreciable effect on the stability of the electrogenerated copper(I) species. This is understandable since the stability of Cu(I) species is influenced by the distortion of the basic geometry rather than any other interactions. The identical CV

response for the reduction of binary and ternary amino acid Cu(II) complexes can be rationalised considering the identical in-plane donor atoms, Cu N_2O_2, and the similar coordination geometry for both types of Cu(II) complexes.

B. Cyclic Voltammetric Response at Low pH Values

It has been mentioned and discussed that amino acids can form complexes with Cu(II) in various stoichiometries depending upon the pH conditions, the nature of amino acids, and the metal-to-ligand ratio.[14] The results of the protonation studies on the binary Cu(II) amino acid complexes have prompted us to carry out similar studies on the present ternary systems. CV of all complexes were recorded at different pH values in the acid range. $HClO_4$ is preferred for adjusting the pH values (the reasons for this are mentioned in chapter 2). Cyclic voltammograms of one of the compounds, Cu(Asp)(Arg), at various pH values are shown in Fig. 4.2. The relevant CV data for some of the complexes are given in Tables 4.2 - 4.6. It is seen that as the pH is decreased the peak heights of C and D decrease while that of A and B increase. Also the cathodic peak A begins to appear in the first scan itself. At very low pH values (< 4.5) peaks C and D completely disappear. At intermediate pH values (5.5-4.2) the peaks A and B

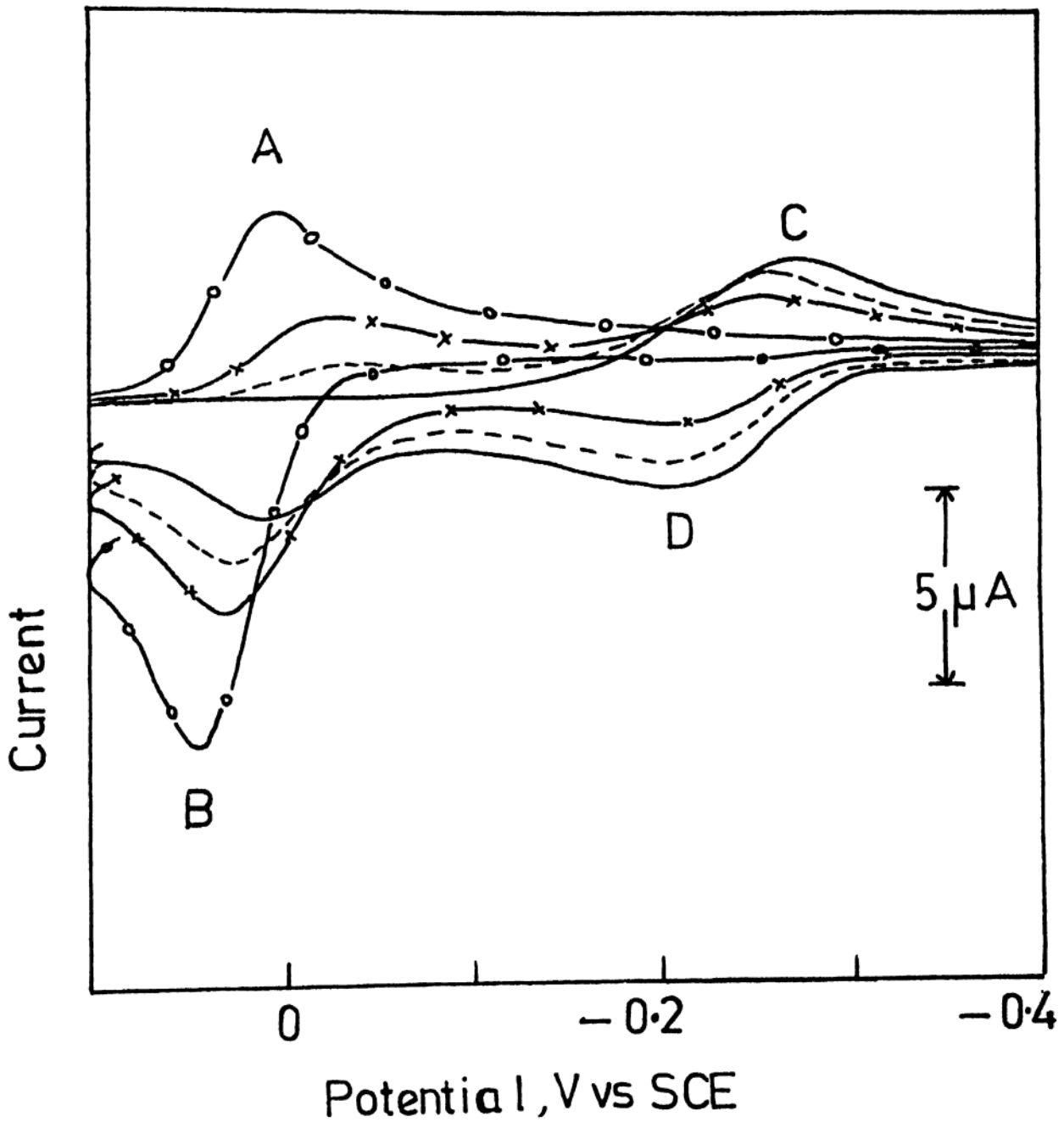


Fig.4.2 Cyclic voltammograms on protonation of Cu(Asp)(Arg) at scan rate, 0.02 Vs^{-1}: (——), pH- 7.10; (—— —), pH - 5.68; (—x—x—), pH = 5.08; (—o—o—), pH 3.05.

Table 4.2. Cyclic voltammetric data on protonation of Cu(Asp)(Lys) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	i_p,A/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$
	0.02	c	0.015	-0.265	-0.190	-	31.82
7.35	0.05	c	0.010	-0.270	-0.200	-	31.30
	0.10	c	0.005	-0.270	-0.200	-	30.28
	0.02	-0.050[b]	0.015	-0.265	-0.190	3.54	28.28
6.47	0.05	-0.055[b]	0.015	-0.265	-0.200	4.47	26.38
	0.10	-0.050[b]	0.010	-0.265	-0.185	4.68	27.82
	0.02	-0.040[b]	0.030	-0.260	-0.190	10.61	21.21
5.92	0.05	-0.040[b]	0.025	-0.260	-0.195	8.94	20.12
	0.10	-0.045[b]	0.025	-0.260	-0.195	9.56	20.85

contd.

Table 4.2 contd.

	0.02	-0.040[b]	0.035	-0.255	-0.190	14.14	17.68
5.53	0.05	-0.040[b]	0.030	-0.255	-0.195	15.65	16.28
	0.10	-0.045[b]	0.035	-0.260	-0.190	15.21	17.12
	0.02	-0.040[b]	0.035	-0.255	-0.185	17.68	10.61
5.22	0.05	-0.040[b]	0.035	-0.255	-0.180	17.89	11.18
	0.10	-0.045[b]	0.035	-0.255	-0.180	16.29	12.26
	0.02	-0.030	0.035	c	c	35.36	-
4.55	0.05	-0.030	0.040	c	c	35.78	-
	0.10	-0.040	0.045	c	c	34.95	-
	0.02	-0.005	0.045	c	c	42.43	-
3.59	0.05	-0.010	0.045	c	c	40.25	-
	0.10	-0.015	0.055	c	c	41.56	-
	0.02	0.000	0.045	c	c	42.45	-
3.25	0.05	-0.005	0.050	c	c	42.49	-
	0.10	-0.010	0.055	c	c	41.94	-

a Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.
b Broad peak
c Peak absent in the first scan.

Table 4.3 contd.

	0.02	-0.030[b]	0.030	-0.260	-0.200	28.34	18.21
4.88	0.05	-0.035[b]	0.030	-0.260	-0.200	31.30	17.89
	0.10	-0.035[b]	0.030	-0.260	-0.200	30.84	19.26
	0.02	-0.025[b]	0.035	-0.250	-0.190	45.56	7.07
4.45	0.05	-0.025[b]	0.035	-0.245	-0.190	40.25	8.94
	0.10	-0.030[b]	0.030	-0.250	-0.190	43.25	9.48
	0.02	-0.020	0.035	c	c	63.64	-
4.17	0.05	-0.025	0.035	c	c	60.67	-
	0.10	-0.025	0.040	c	c	62.15	-
	0.02	.0.000	0.045	c	c	70.71	-
3.50	0.05	-0.005	0.045	c	c	69.32	-
	0.10	-0.010	0.045	c	c	68.39	-

a Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b Broad peak

c Peak absent in the first scan.

Table 4.4. Cyclic voltammetric data on protonation of Cu(Asp)(Orn) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	i_p,A/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$
	0.02	c	0.020	-0.255	-0.190	-	24.27
6.75	0.05	c	0.020	-0.255	-0.185	-	23.36
	0.10	c	0.010	-0.265	-0.180	-	24.35
	0.02	-0.030	0.020	-0.250	-0.190	7.07	19.37
6.26	0.05	-0.040	0.015	-0.255	-0.185	8.94	18.78
	0.10	-0.045	0.010	-0.265	-0.180	7.58	19.12
	0.02	-0.025[b]	0.025	-0.245	-0.190	12.73	14.14
5.69	0.05	-0.040[b]	0.020	-0.250	-0.185	13.42	13.42
	0.10	-0.040[b]	0.025	-0.260	-0.180	12.85	14.64
	0.02	-0.020[b]	0.030	-0.245	-0.190	17.68	8.84
5.36	0.05	-0.030[b]	0.025	-0.250	-0.185	17.83	8.94
	0.10	-0.030[b]	0.035	-0.260	-0.175	16.93	8.26

contd.

Table 4.4 contd.

	0.02	-0.020[b]	0.030	-0.240	-0.185	23.33	3.54
5.08	0.05	-0.030[b]	0.030	-0.245	-0.180	24.64	4.47
	0.10	-0.030[b]	0.035	-0.255	-0.175	23.58	3.92
	0.02	-0.020	0.030	c	c	29.70	-
4.60	0.05	-0.030	0.030	c	c	33.54	-
	0.10	-0.025	0.040	c	c	31.26	-
	0.02	-0.005	0.040	c	c	37.12	-
3.83	0.05	-0.005	0.045	c	c	35.78	-
	0.10	-0.005	0.050	c	c	36.58	-
	0.02	.0.00	0.040	c	c	38.00	-
3.06	0.05	-0.005	0.045	c	c	36.90	-
	0.10	-0.005	0.050	c	c	37.26	-

a Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b Broad peak

c Peak absent in the first scan.

Table 4.5. Cyclic voltammetric data on protonation of Cu(Glu)(Lys) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	i_p,A/$\upsilon^{1/2}$	i_p,C/$\upsilon^{1/2}$
	0.02	c	0.020	-0.240	-0.175	-	33.59
7.85	0.05	c	0.020	-0.240	-0.170	-	33.54
	0.10	c	c	-0.245	-0.170	-	33.26
	0.02	-0.060[b]	0.020	-0.235	-0.180	6.25	26.26
6.84	0.05	-0.060[b]	0.015	-0.240	-0.175	6.71	24.60
	0.10	-0.070[b]	0.015	-0.240	-0.170	6.32	25.30
	0.02	-0.050[b]	0.025	-0.230	-0.180	10.61	18.26
6.24	0.05	-0.055[b]	0.025	-0.235	-0.175	11.18	17.89
	0.10	-0.060[b]	0.020	-0.240	-0.175	11.86	18.97
	0.02	-0.040[b]	0.035	-0.225	-0.175	21.21	10.61
5.34	0.05	-0.045[b]	0.040	-0.225	-0.175	22.36	10.06
	0.10	-0.050[b]	0.040	-0.230	-0.170	21.85	10.26

contd.

Table 4.5 contd.

	0.02	-0.035[b]	0.040	-0.220	-0.170	28.28	5.26
4.99	0.05	-0.035	0.045	-0.220	-0.160	26.83	4.47
	0.10	-0.045	0.045	-0.225	-0.160	27.26	4.74
	0.02	-0.020	0.045	c	c	38.89	-
4.28	0.05	-0.025	0.050	c	c	40.25	-
	0.10	-0.030	0.050	c	c	39.95	-
	0.02	-0.010	0.045	c	c	38.89	-
3.83	0.05	-0.020	0.050	c	c	40.25	-
	0.10	-0.025	0.055	c	c	39.95	-
	0.02	-0.010	0.050	c	c	38.89	-
3.36	0.05	-0.015	0.055	c	c	40.16	-
	0.10	-0.025	0.060	c	c	40.28	-

a Potentials expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b Broad peak

c Peak absent in the first scan.

Table 4.6. Cyclic voltammetric data on protonation of Cu(Glu)(Orn) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	i_p,A/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$
	0.02	c	0.010	-0.240	-0.170	-	28.28
7.17	0.05	c	0.005	-0.240	-0.170	-	26.56
	0.10	c	0.005	-0.250	-0.160	-	25.30
	0.02	c	0.015	-0.235	-0.170	-	24.75
6.66	0.05	c	0.010	-0.240	-0.170	-	23.26
	0.10	c	0.010	-0.250	-0.160	-	22.14
	0.02	-0.040[b]	0.020	-0.230	-0.170	7.07	17.68
6.25	0.05	-0.040[b]	0.020	-0.240	-0.170	6.86	17.26
	0.10	-0.050[b]	0.020	-0.250	-0.160	6.32	18.97
	0.02	-0.030[b]	0.030	-0.230	-0.170[b]	14.14	10.61
5.58	0.05	-0.035[b]	0.025	-0.235	-0.170[b]	13.26	11.58
	0.10	-0.040[b]	0.035	-0.240	-0.160[b]	12.65	11.07

con+d.

Table 4.6 contd.

	0.02	-0.030[b]	0.030	-0.225	-0.165[b]	24.75	3.54
5.18	0.45	-0.030[b]	0.035	-0.230	-0.165[b]	23.56	2.36
	0.10	-0.030[b]	0.040	-0.235	-0.160[b]	22.14	1.58
	0.02	-0.005	0.035	c	c	32.53	-
4.24	0.05	-0.010	0.040	c	c	31.85	-
	0.10	-0.025	0.040	c	c	30.82	-
	0.02	0.000	0.040	c	c	31.64	-
3.75	0.05	-0.005	0.045	c	c	32.63	-
	0.10	-0.010	0.045	c	c	31.62	-
	0.02	0.000	0.040	c	c	35.35	-
3.35	0.05	-0.005	0.045	c	c	34.64	-
	0.10	-0.010	0.050	c	c	35.26	-

a Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b Broad peak

c Peak absent in the first scan.

shift anodically along with a decrease in the 'peak-broadening' of peak A.

On protonation the Cu(A)(B) complexes get converted to other forms preferably the 1:1 complexes, Cu(A) and/or Cu(B).[14] On further protonation they dissociate into free copper(II) ions. The decrease in peak heights and the final disappearance of peaks C and D are due to the depletion of the Cu(A)(B) species on protonation. The binary 1:1 complexes, Cu(A) and Cu(B), may undergo reduction at a potential less cathodic than peak C. Hence the 'peak-broadening observed for peak A at intermediate pH values can be due to the overlap of the reduction waves of these species with that of free Cu(II) ions, when the relative concentration of the latter is very small. A plot of E_p,A and E_p,B against pH for Cu(Asp)(Orn) is shown in Fig.4.3. The plateau (R-S) observed at intermediate pH values should correspond to an enhanced concentration of Cu(A) and Cu(B) species over free Cu(II) ions. The region (P-Q) corresponds to the free copper(II) ions where the concentration of the complexes is practically nil. Fig.4.3 also shows the anodic shift of peak potentials of A and B as the pH decreases. The reversibility of the protonation is examined and the process is found to be reversible. Fig.4.4 shows the cyclic voltammograms of Cu(Asp)(Arg) on increasing pH

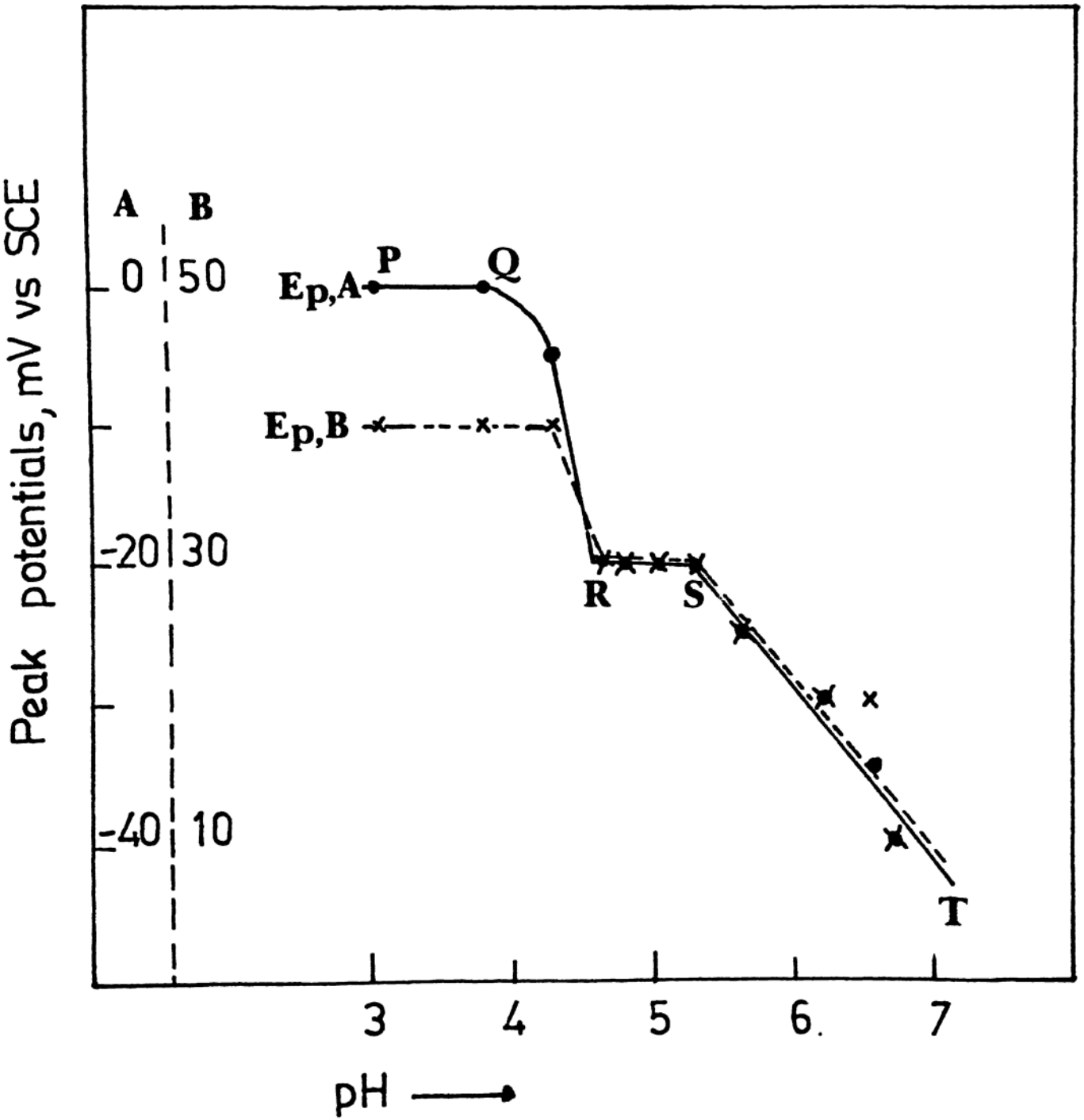


Fig.4.3 Plot of E_p,A and E_pB against pH of Cu(Asp)(Orn); (—●—●—) E_p,A; (—x—x—), E_p,B.

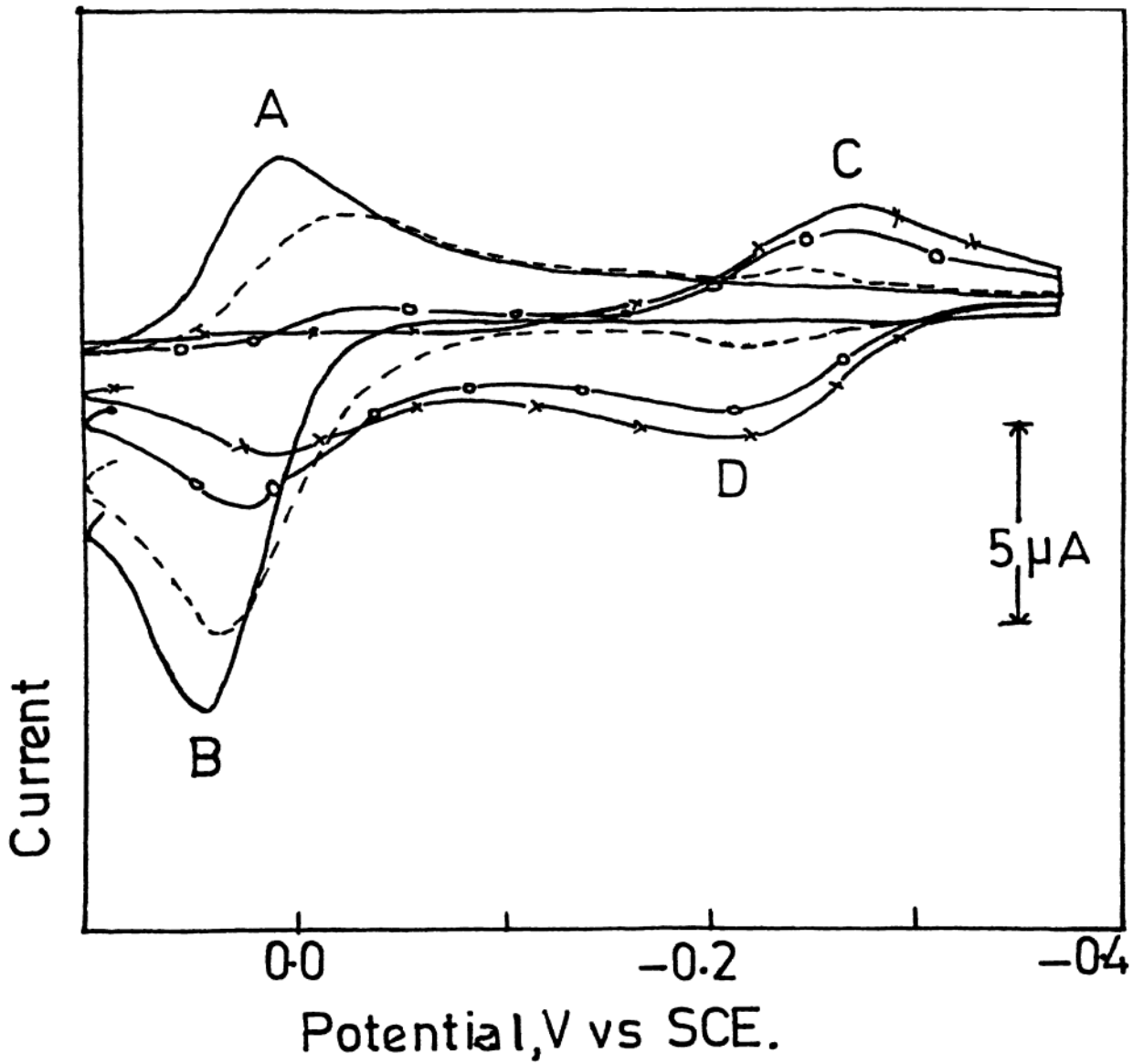


Fig.4.4 Cyclic voltammograms on deprotonation of Cu(Asp)(Arg) at Scan rate 0.02 Vs^{-1}; (——), pH = 3.50; (– – – –), pH = 4.52; (—o—o—), pH = 5.65; (—x—x—), pH = 7.35.

from low values. Corresponding CV data on two sample systems are presented in Tables 4.7 and 4.8. The other complexes follow the same pattern. In all cases the process of protonation is quite reversible.

Part-2 : Copper(II) 2,2'-Bipyridyl (BPY) Amino Acid (A) Type

A. Cyclic Voltammetric Response at Neutral pH

Cyclic voltammograms of ternary copper(II) complexes of amino acids containing 2,2'-bipyridyl, Cu(BPY)(A), were recorded from aqueous solutions of the samples at HMDE in the potential range +0.1 to -0.6 V versus SCE. CV response of all the ternary complexes are very similar and representative cyclic voltammograms of Cu(BPY)(Ala) are shown in Figs. 4.5a and 4.5b. The relevant CV data for all the complexes are given in Table 4.9. The complexes display one reduction peak C at about -0.230 V and on reverse scan an anodic peak D at about -0.150 V appears. At sweep rates slower than 0.01 Vs^{-1}, an additional anodic peak B develops at about +0.02 V and in such cases, a new cathodic peak A, at about -0.03 V appears in the second and subsequent scans. This behaviour is shown in Fig. 4.5b and is very similar to that of binary Cu(II) amino acid complexes discussed in chapter 2.

Table 4.7. Cyclic voltammetric data on deprotonation of Cu(Asp)(Lys) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	i_p,A/$\upsilon^{1/2}$	i_p,C/$\upsilon^{1/2}$
	0.02	0.000	0.045	c	c	42.43	-
3.25	0.05	-0.005	0.050	c	c	42.49	-
	0.10	-0.010	0.055	c	c	41.94	-
	0.02	-0.015	0.040	c	c	38.89	-
4.19	0.05	-0.020	0.040	c	c	38.29	-
	0.10	-0.025	0.050	c	c	37.95	-
	0.02	-0.035	0.030	-0.250	-0.190	24.75	7.07
5.10	0.05	-0.035	0.035	-0.250	-0.190	23.56	7.26
	0.10	-0.030	0.040	-0.255	-0.190	22.14	6.32
	0.02	-0.040[b]	0.030	-0.250	-0.190	17.68	10.61
5.41	0.05	-0.040[b]	0.030	-0.250	-0.190	17.16	10.85
	0.10	-0.045[b]	0.035	-0.255	-0.190	17.39	11.07

contd.

Table 4.7 contd.

	0.02	-0.040[b]	0.030	-0.260	-0.190	14.14	14.14
5.74	0.05	-0.050[b]	0.030	-0.265	-0.185	14.26	14.86
	0.10	-0.055[b]	0.030	-0.265	-0.185	14.23	14.23
	0.02	-0.050	0.025	-0.260	-0.190	10.61	21.21
6.20	0.05	-0.055	0.020	-0.265	-0.185	10.02	21.46
	0.10	-0.060	0.020	-0.265	-0.185	9.49	22.14
	0.02	-0.060	0.020	-0.265	-0.185	3.54	28.28
6.62	0.05	-0.060	0.020	-0.265	-0.185	3.85	26.25
	0.10	-0.060	0.020	-0.265	-0.180	4.74	25.30
	0.02	c	0.010	-0.265	-0.185	-	31.82
7.25	0.05	c	0.020	-0.265	-0.185	-	31.56
	0.10	c	0.015	-0.265	-0.180	-	30.04

a Potentials expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b Broad peak

c Peak absent in the first scan.

Table 4.8. Cyclic voltammetric data on deprotonation of Cu(Asp)(Arg) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	i_p,A/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$
	0.02	0.000	0.045	c	c	70.71	-
3.50	0.05	-0.005	0.045	c	c	68.32	-
	0.10	-0.010	0.045	c	c	68.39	-
	0.02	-0.020	0.040	c	c	57.26	-
4.27	0.05	-0.030	0.030	c	c	58.14	-
	0.10	-0.030	0.045	c	c	58.50	-
	0.02	-0.035[b]	0.035	-0.250[b]	-0.190[b]	48.21	5.62
4.52	0.05	-0.025[b]	0.035	-0.250	-0.190[b]	46.96	4.47
	0.10	-0.030[b]	0.040	-0.240	-0.190[b]	49.02	6.32
	0.02	-0.030[b]	0.035	-0.255	-0.195	28.21	16.26
5.02	0.05	-0.035[b]	0.030	-0.260	-0.200	29.07	17.89
	0.10	-0.040[b]	0.030	-0.260	-0.200	30.04	17.39

contd.

Table 4.8 contd.

	0.02	-0.045[b]	0.025	-0.265	-0.205	14.61	30.84
5.65	0.05	-0.045[b]	0.020	-0.270	-0.210	15.67	31.30
	0.10	-0.050[b]	0.020	-0.270	-0.200	15.81	28.40
	0.02	-0.050	0.020	-0.270	-0.210	7.62	36.26
6.15	0.05	-0.050	0.020	-0.270	-0.210	8.94	35.78
	0.10	-0.050	0.015	-0.275	-0.200	7.91	37.95
	0.02	c	0.015	-0.275	-0.210	-	43.21
6.58	0.05	c	0.015	-0.270	-0.205	-	44.72
	0.10	c	0.015	-0.275	-0.200	-	41.11
	0.02	c	0.010	-0.275	-0.210	-	48.21
7.30	0.05	c	0.010	-0.275	-0.205	-	49.19
	0.10	c	0.010	-0.278	-0.200	-	47.43

a Potentials expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b Broad peak

c Peak absent in the first scan.

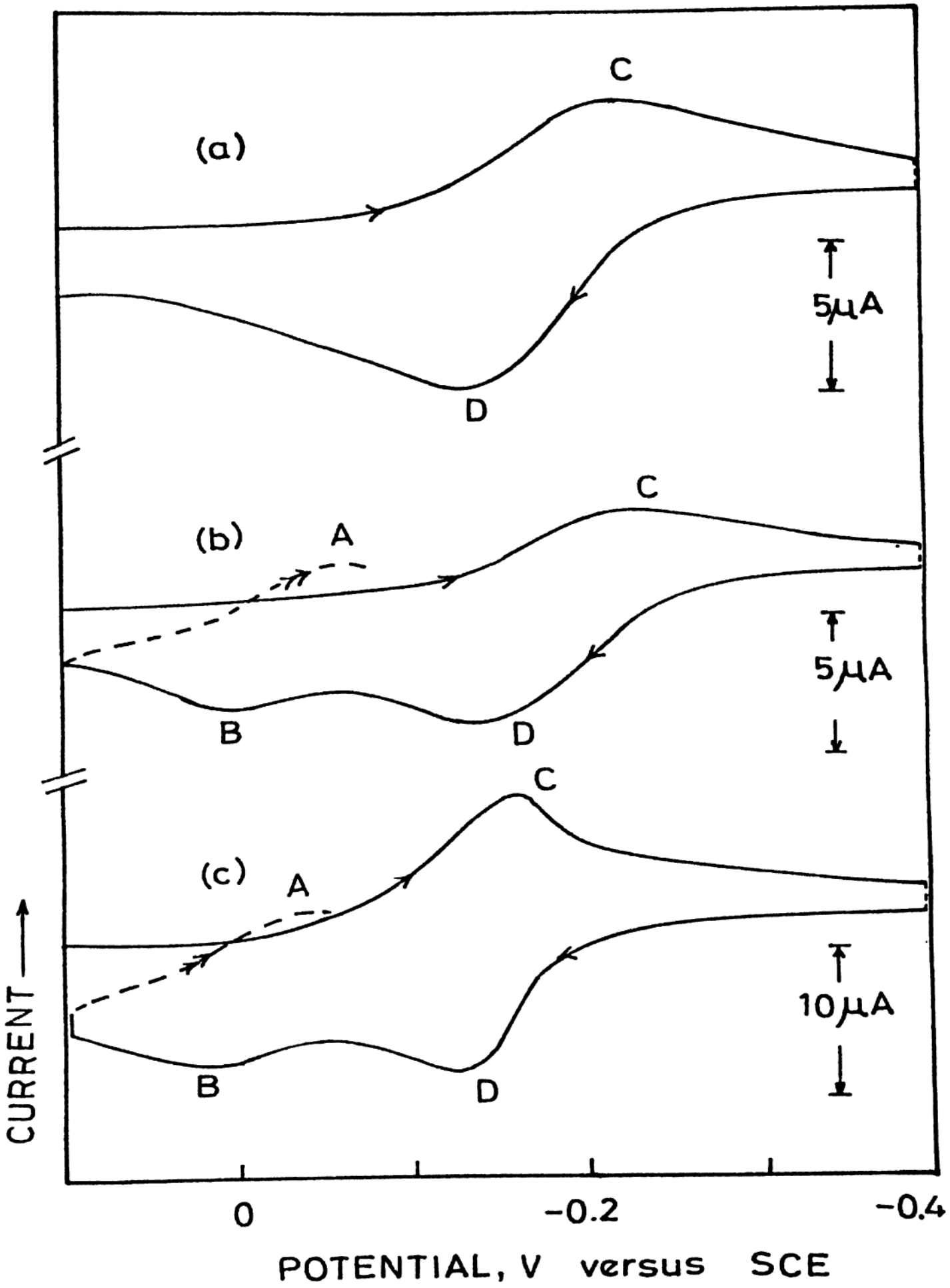


Fig.4.5 Cyclic voltammograms recorded at various scan rates. (a) Cu(BPY)(Ala) at 0.02 Vs^{-1}, (b) Cu(BPY)(Ala) at 0.01 Vs^{-1}, (c) $Cu(BPY)_2$ at 0.02 Vs^{-1}.

Table 4.9. Cyclic voltammetric data on copper(II) mixed amino acid complexes containing bipyridyl in aqueous media.[a]

Compound	Sweep rate (Vs^{-1})	E_p,A[b]	E_p,B	E_p,C	E_p,D	$\Delta E_{p,C-D}$	$E^{o'}_{C-D}$
Cu(BPY)(Gly)	0.01	-0.020	0.040	-0.230	-0.150	0.080	-0.190
	0.02	c	0.030	-0.220	-0.145	0.075	-0.183
	0.05	c	c	-0.220	-0.145	0.075	-0.183
	0.10	c	c	-0.220	-0.135	0.085	-0.178
Cu(BPY)(Ala)	0.01	c	0.020	-0.225	-0.140	0.085	-0.183
	0.02	c	0.020	-0.225	-0.135	0.090	-0.180
	0.05	c	0.020	-0.220	-0.135	0.090	-0.178
	0.10	c	c	-0.220	-0.140	0.080	-0.180
Cu(BPY)(Ile)	0.01	-0.025	0.040	-0.230	-0.150	0.080	-0.190
	0.02	-0.025	0.030	-0.230	-0.150	0.080	-0.190
	0.05	c	c	-0.230	-0.140	0.090	-0.185
	0.10	c	c	-0.225	-0.135	0.090	-0.180

cont'd.

Table 4.9 contd.

Cu(BPY)(Val)	0.01	-0.030	0.040	-0.225	-0.160	0.065	-0.193
	0.02	-0.030	0.030	-0.220	-0.150	0.070	-0.185
	0.05	c	c	-0.220	-0.140	0.080	-0.180
	0.10	c	c	-0.220	-0.140	0.080	-0.180
Cu(BPY)(Ser)	0.02	c	c	-0.225	-0.135	0.090	-0.180
	0.05	c	c	-0.220	-0.140	0.080	-0.180
	0.10	c	c	-0.220	-0.145	0.075	-0.183

a Potentials are expressed in volts.

b Peak appears only in the second scan.

c Peak absent.

The peak C in these ternary complexes results from a one-electron reduction and the peak D from the re-oxidation of the reduced species as in equation 4.4.

$$Cu(II)(BPY)(A) \underset{\text{D}}{\overset{1e}{\rightleftharpoons}} [Cu(I)(BPY)(A)] \quad \ldots 4.4$$

The appearance of the peaks A and B at slow scan rates indicates the generation of Cu^{o} at the electrode. Hence at slow scan rates, the reduced Cu(I) species undergo decomposition and/or disproportionation to generate Cu^{o} at the electrode.

The peaks A and B result from the redox process represented in equation 4.5.

$$Cu^{o}_{(Hg)} \underset{\text{A}}{\overset{2e}{\rightleftharpoons}} Cu^{2+}_{(aq)} \quad \ldots 4.5$$

In Cu(BPY)(A) type complexes, peaks B and A develop only at slow scan rates ($\nu \lesssim 0.01\ Vs^{-1}$), whereas in binary Cu(II) amino acid complexes these peaks are observable even at scan rates as high as $0.1\ Vs^{-1}$. This suggests that the intermediate Cu(I)(BPY)(A) has a longer life-time than the corresponding copper(I) binary amino acid complexes. The binary Cu(II) bipyridyl complex, $Cu(BPY)_2$ gives a CV profile similar to that of binary Cu(II) amino acid

complexes, and is shown in Fig. 4.5c. It also displays peaks corresponding to C-D and A-B couples. The appearance of A and B at normal scan rates, like 0.02 Vs^{-1}, indicates that $Cu(I)(BPY)_2$ intermediate has a shorter lifetime compared to Cu(I)(BPY)(A) complexes mentioned earlier. It is known that ternary bipyridyl Cu(II) complexes, Cu(BPY)(L), where L has both O and N donor atoms ($Cu\ N_3O$ chromophore) is more stable than the binary bipyridyl, $Cu(BPY)_2$ complexes,[8,9] which has a $Cu\ N_4$ chromophore. The observed stability of the intermediate [Cu(I)(BPY)(A)] over the $[Cu(I)(BPY)_2]$ complexes can also be of similar origin.

B. CV Response at Low pH values

To study the effect of changing the pH on these ternary systems cyclic voltammograms were recorded at various pH values. The pH values were adjusted by the addition of $HClO_4$. All complexes behave in a similar fashion on protonation. Cyclic voltammograms at various pH values for one of the complexes, Cu(BPY)(Ser) is shown in Fig.4.6. The relevant CV data for most of the complexes are given in Tables 4.10-4.14. The behaviour of these complexes on protonation is quite different from the systems so far discussed. The major changes are the

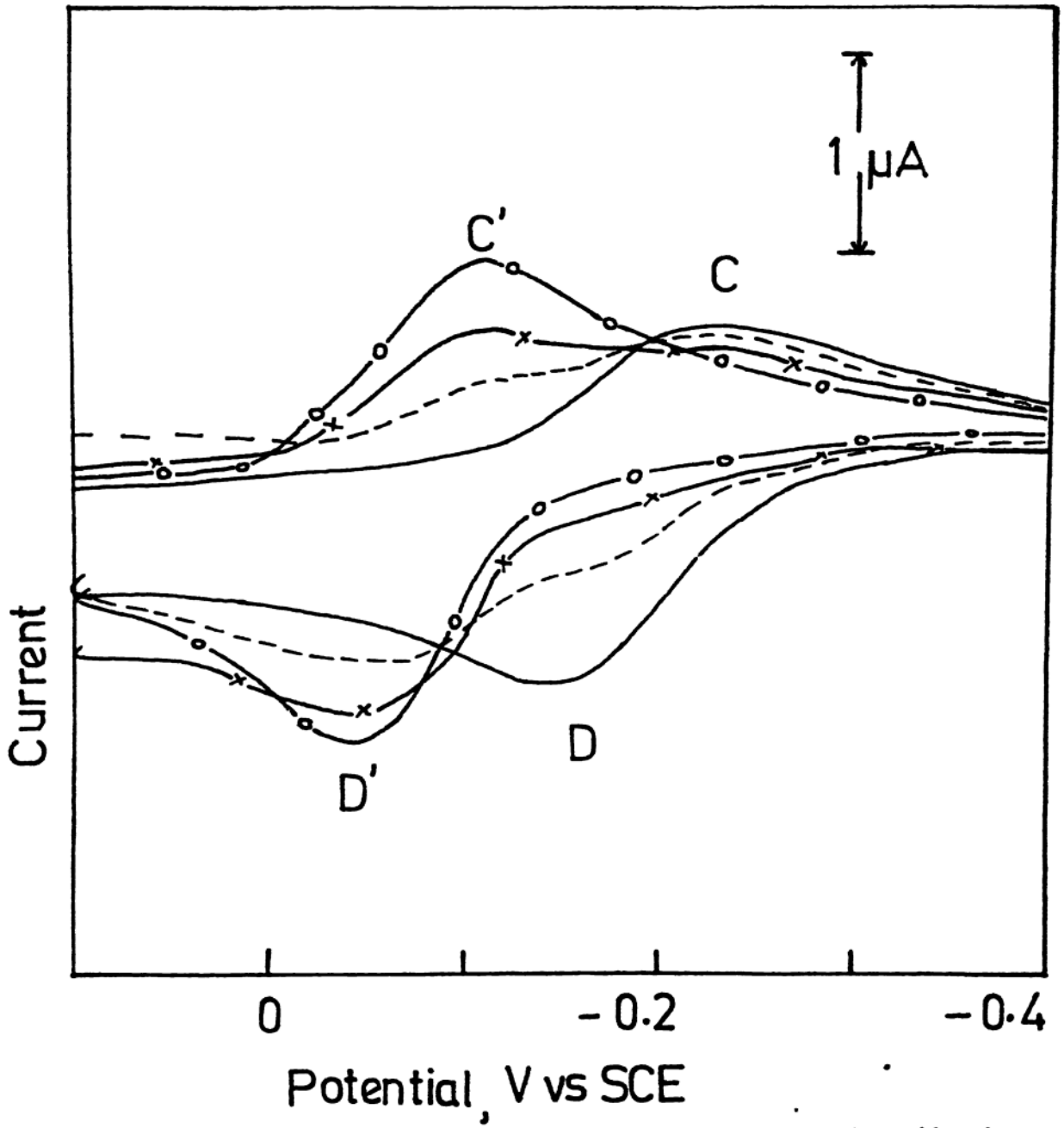


Fig.4.6 Cyclic voltammograms on protonation of Cu(BPY)(Ser) at scan rate, 0.02 Vs^{-1}; (——), pH = 7.50; (– – –),pH = 5.63; (—x—x—), pH = 5.19; (—o—o—), pH = 3.15.

Table 4.10. Cyclic voltammetric data on protonation of Cu(BPY)(Gly) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	i_p,A/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$
	0.02	c	c	-0.225	-0.160	-	8.84
7.46	0.05	c	c	-0.220	-0.160	-	8.21
	0.10	c	c	-0.220	-0.150	-	8.74
	0.02	c	c	-0.220	-0.160	-	8.49
6.76	0.05	c	c	-0.220	-0.150	-	8.39
	0.10	c	c	-0.210	-0.150	-	8.22
	0.02	c	c	-0.220	-0.160	-	7.78
6.18	0.05	c	c	-0.210	-0.150	-	7.56
	0.10	c	c	-0.210	-0.140	-	7.43
	0.02	-0.110[b]	d	-0.220	-0.160	4.24	2.83
5.58	0.05	-0.110[b]	d	-0.210	-0.150	4.32	2.52
	0.10	-0.110[b]	d	-0.210	-0.150	4.43	2.21

contd.

Table 4.10 contd.

	0.02	-0.110	-0.030	-0.240	c	7.07	-
4.55	0.05	-0.110	-0.030	-0.240	c	7.12	-
	0.10	-0.110	-0.030	c	c	6.96	-
	0.02	-0.110	-0.030	c	c	7.78	-
3.73	0.05	-0.110	-0.030	c	c	7.68	-
	0.10	-0.110	-0.030	c	c	7.59	-
	0.02	-0.110	-0.030	c	c	8.56	-
3.03	0.05	-0.110	-0.030	c	c	8.49	-
	0.10	-0.110	-0.040	c	c	8.85	-
	0.02	-0.100	-0.030	c	c	9.16	-
2.80	0.05	-0.100	-0.030	c	c	9.19	-
	0.10	-0.100	-0.040	c	c	9.17	-

a CV data collected from sample solutions of concentration 0.152 mM. Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b Broad peak; c. Peak absent; d. masked by adsorption wave.

Table 4.11. Cyclic voltammetric data on protonation of Cu(BPY)(Ala) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	i_p,A/$\upsilon^{1/2}$	i_p,C/$\upsilon^{1/2}$
	0.02	c	c	-0.210	-0.135	-	6.12
7.60	0.05	c	c	-0.210	-0.130	-	6.52
	0.10	c	c	-0.200	-0.130	-	6.32
	0.02	c	c	-0.210	-0.130	-	5.21
6.48	0.05	c	c	-0.210	-0.130	-	5.25
	0.10	c	c	-0.200	-0.130	-	5.55
	0.02	-0.100[b]	c	-0.200	-0.140	-	5.16
6.15	0.05	c	c	-0.200	-0.140	-	5.20
	0.10	c	c	-0.200	-0.140	-	5.06
	0.02	-0.110	-0.030	c	c	4.26	-
5.03	0.05	-0.110	d	c	c	4.06	-
	0.10	-0.110	d	c	c	4.11	-

contd.

Table 4.11 contd.

	0.02	-0.110	-0.030	c	c	4.50	-
4.50	0.05	-0.110	-0.050	c	c	4.26	-
	0.10	-0.110	-0.050	c	c	4.43	-
	0.02	-0.100	-0.030	c	c	4.68	-
4.15	0.05	-0.110	-0.030	c	c	4.48	-
	0.10	-0.110	-0.040	c	c	4.59	-
	0.02	-0.090	-0.025	c	c	5.16	-
3.98	0.05	-0.090	-0.030	c	c	5.28	-
	0.10	-0.090	-0.030	c	c	5.06	-
	0.02	-0.080	-0.025	c	c	5.45	-
3.76	0.05	-0.080	-0.025	c	c	5.70	-
	0.10	-0.085	-0.025	c	c	5.69	-

a. CV data collected from sample solution of conc 0.262 mM. Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b. Broad peak; c. Peak absent; d. Masked by adsorption wave.

Table 4.12. Cyclic voltammetric data on protonation of Cu(BPY)(Ile) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	i_p,A/$\vartheta^{1/2}$	i_p,C/$\vartheta^{1/2}$
	0.02	c	0.020	-0.225	-0.150	-	8.26
7.80	0.05	c	c	-0.225	-0.140	-	8.15
	0.10	c	c	-0.210	-0.140	-	8.02
	0.02	c	0.020	-0.225	-0.150	-	8.13
7.02	0.05	c	0.020[b]	-0.225	-0.150	-	7.85
	0.10	c	0.020[b]	-0.215	-0.140	-	7.91
	0.02	c	0.020	-0.220	-0.160	-	7.07
6.00	0.05	c	d	-0.210	-0.160	-	7.12
	0.10	c	d	-0.210	-0.150	-	7.42
	0.02	-0.110	-0.030	-0.240	c	7.07	0.92
4.75	0.05	-0.110	-0.030	-0.240	c	7.36	0.81
	0.10	-0.110	d	-0.250	c	7.42	0.89

contd.

Table 4.12 contd.

	0.02	-0.110	-0.030	-0.240	c	7.78	0.75
4.25	0.05	-0.110	-0.030	-0.840	c	7.68	0.81
	0.10	-0.110	-0.040	-0.230	c	7.59	-
	0.02	-0.110	-0.035	c	c	8.49	c
3.79	0.05	-0.110	-0.035	c	c	8.35	-
	0.10	-0.110	-0.040	c	c	8.22	-
	0.02	-0.110	-0.035	c	c	9.21	-
3.26	0.05	-0.110	-0.035	c	c	8.92	-
	0.10	-0.100	-0.035	c	c	8.85	-
	0.02	-0.100	-0.035	c	c	9.90	-
3.06	0.05	-0.100	-0.035	c	c	9.68	-
	0.10	-0.100	-0.035	c	c	9.49	-

a. CV data collected from sample solution of conc 0.225 mM, potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b. Broad peak; c. Peak absent; d. Masked by adsorption wave.

Table 4.13. Cyclic voltammetric data on protonation of Cu(BPY)(Val) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	i_p,A/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$
	0.02	c	c	-0.220	-0.150	-	6.85
7.82	0.05	c	c	-0.210	-0.140	-	6.66
	0.10	c	c	-0.210	-0.140	-	7.12
	0.02	c	0.010[b]	-0.220	-0.150	-	5.89
7.08	0.05	c	0.010[b]	-0.210	-0.140	-	6.27
	0.10	c	c	-0.210	-0.140	-	6.45
	0.02	c	0.010	-0.220	-0.150	-	6.46
6.00	0.05	c	c	-0.210	-0.140	-	6.12
	0.10	c	c	-0.210	-0.140	-	6.32
	0.02	-0.110	d	-0.240	c	4.12	-
5.43	0.05	-0.110	d	-0.240	c	4.23	-
	0.10	-0.110	d	c	c	4.43	-

contd.

Table 4.13 contd.

	0.02	-0.110	-0.030	c	c	5.93	-
4.92	0.05	-0.110	-0.030	c	c	5.85	-
	0.10	-0.110	-0.040	c	c	6.01	-
	0.02	-0.110	-0.030	c	c	6.36	-
4.03	0.05	-0.110	-0.035	c	c	6.52	-
	0.10	-0.110	-0.035	c	c	6.17	-
	0.02	-0.110	-0.050	c	c	7.07	-
3.06	0.05	-0.110	-0.040	c	c	7.50	-
	0.10	-0.110	-0.040	c	c	7.12	-

a. CV data collected from sample solution of conc 0.216 mM potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b. Broad peak; c. Peak absent; d. Masked by adsorption peak.

Table 4.14. Cyclic voltammetric data on protonation of Cu(BPY)(Ser) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	i_p,A/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$
	0.02	c	c	-0.225	-0.140	-	6.36
7.70	0.05	c	c	-0.220	-0.135	-	6.71
	0.10	c	c	-0.225	-0.140	-	6.56
	0.02	c	c	-0.220	-0.140	-	6.00
6.22	0.05	c	c	-0.220	-0.140	-	6.41
	0.10	c	c	-0.220	-0.135	-	6.32
	0.02	-0.100[b]	-0.040[b]	-0.220	-0.140	3.54	2.12
5.63	0.05	-0.090[b]	d	-0.220	-0.140	3.13	2.68
	0.10	-0.100[b]	d	-0.220	-0.150	3.29	2.48
	0.02	-0.110[b]	-0.030[b]	-0.240	c	4.95	-
5.37	0.05	-0.110[b]	d	-0.250	c	4.92	-
	0.10	-0.110[b]	d	-0.230	c	4.68	-

contd.

Table 4.14 contd.

	0.02	-0.110	-0.030[b]	-0.250	c	5.66	-
5.19	0.05	-0.110	d	-0.250	c	5.81	-
	0.10	-0.110	d	-0.240	c	5.72	-
	0.02	-0.110	-0.040	c	c	7.07	-
4.03	0.05	-0.110	-0.050	c	c	7.16	-
	0.10	-0.110	-0.060	c	c	7.31	-
	0.02	-0.110	-0.040	c	c	7.78	-
3.52	0.05	-0.120	-0.055	c	c	7.52	-
	0.10	-0.120	-0.050	c	c	7.62	-
	0.02	-0.110	-0.050	c	c	8.49	-
3.15	0.05	-0.120	-0.050	c	c	8.25	-
	0.10	-0.110	-0.040	c	c	8.52	-

a. CV data collected from solution of conc 0.186 mM. Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b. Peak absent; d. Masked by adsorption wave.

following. As the pH decreases the peaks C and D decrease in heights. Simultaneously a new set of peaks C' and D' starts developing at about -0.110 V and -0.05 V respectively. By the time peak C disappears (at pH about 3.0) the peaks C' and D' develop fully. At intermediate pH values it is possible to observe both the pairs of peaks. The new pair of peaks (C' and D') corresponds to a one-electron reversible ($\triangle E_p \simeq 60$ mV; $i_{pa}/i_{pc} \simeq 1$) redox process. There are no peaks equivalent to A-B couple observed in binary systems. The decrease in heights and the final disappearance of peaks C and D are due to the continuous depletion of the starting complex, Cu(BPY)(A) on protonation. In the case of binary Cu(II) amino acid complexes, they completely dissociate to free Cu(II) ions at very low pH values like 3.2. This was demonstrated by the appearance of the characteristic redox peaks of free Cu(II) ions. But for the bipyridyl ternary systems the peaks C' and D', observed at very low pH values do not correspond to free Cu(II) ions. These facts are clearly observable in Fig.4.7. The binary bipyridyl Cu(II) complexes also show a behaviour similar to that of ternary Cu(BPY)(A) complexes on protonation. These observations show that the end-product of the protonation of the ternary Cu(BPY)(A) and the binary $Cu(BPY)_2$ complexes is the same. The mono bipyridyl Cu(II) complex,[10] $Cu(BPY)(H_2O)_2$, which is stable

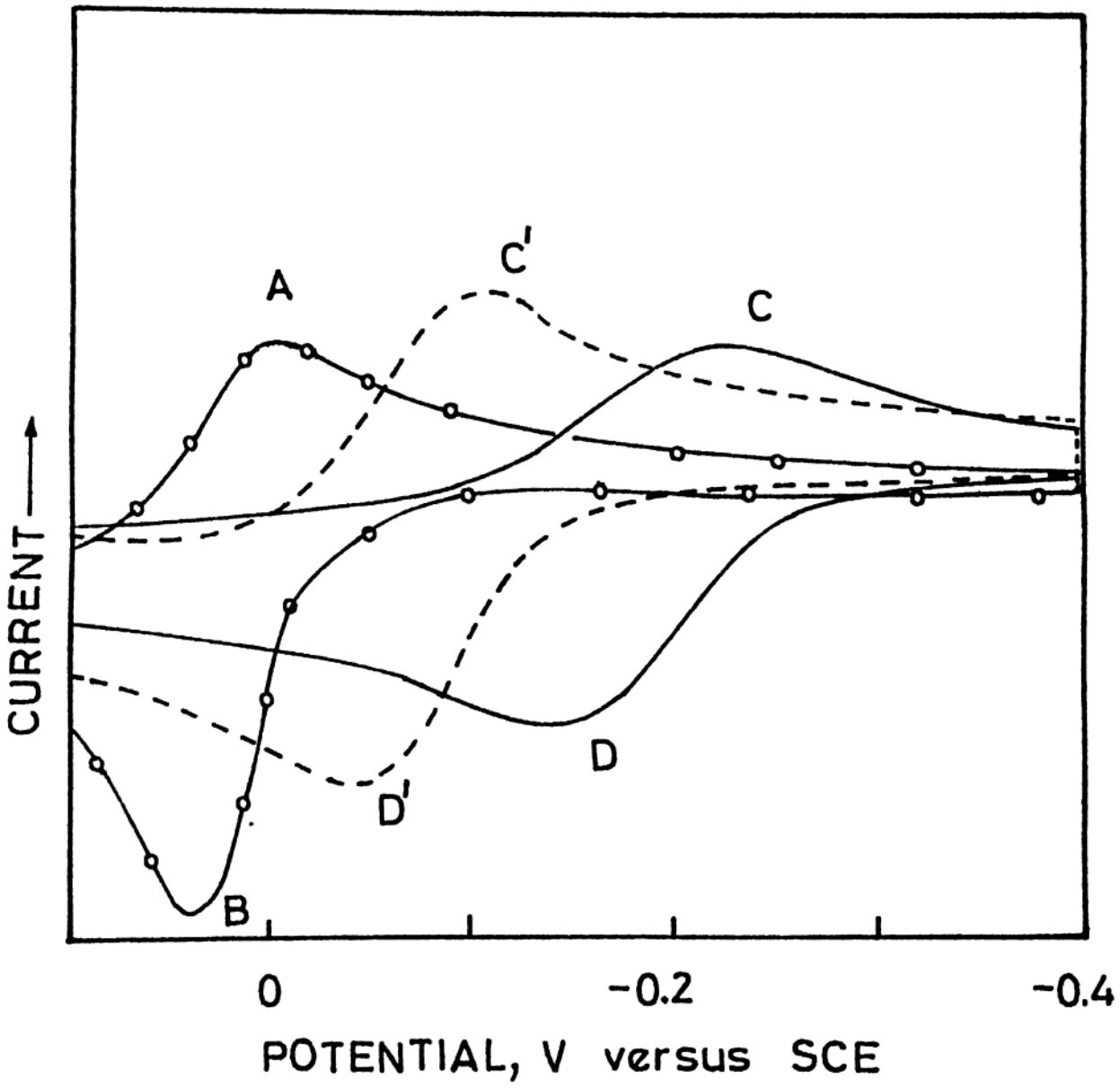


Fig.4.7 Cyclic voltammograms of (——), Cu(BPY)(Ser) at pH = 7.50 (5μA/inch); (— — —), Cu(BPY)(Ser) at pH = 3.12 (5μA/inch); (—o—o—), $Cu(ClO_4)_2$ om aqueous media (10μA/inch).

even at very low pH values can be a definite possibility as the end-product of the protonation of the above complexes. The process of protonation is found to be reversible. CV data for deprotonation of a few complexes is given in Tables 4.15 and 4.16.

Part-3 : Histidine Containing Ternary Copper(II) Complexes

A. Copper(II) Histidine(His) Amino Acid(A) Type

Histidine containing ternary Cu(II) complexes of amino acids were prepared by mixing the components in 1:1:1 ratio and adjusting the pH to the required value. Only for this series of compounds the solid complexes were not isolated but were studied in situ. The formation of the complexes was confirmed by recording their visible spectra. All of the complexes have absorption bands at λ_{max} 610-615 nm.[18] Cyclic voltammograms of these complexes were recorded in aqueous solutions at pH 7.5-8.3 using HMDE and with $NaClO_4$ as supporting electrolyte. Cyclic voltammograms of all these types of complexes are similar and Fig.4.8a presents the sample cyclic voltammograms of the reduction of histidine containing ternary Cu(II) amino acid complex, Cu(His)(Gly). CV data for all complexes are given in Table 4.17.

Table 4.15. Cyclic voltammetric data on deprotonation of Cu(BPY)(Gly) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	i_p,A/$\upsilon^{1/2}$	i_p,C/$\upsilon^{1/2}$
	0.02	-0.090	-0.030	c	c	9.19	-
2.80	0.05	-0.100	-0.030	c	c	9.26	-
	0.10	-0.100	-0.040	c	c	9.17	-
	0.02	-0.100	-0.040	c	c	7.89	-
3.20	0.05	-0.110	-0.040	c	c	8.05	-
	0.10	-0.110	-0.040	c	c	7.59	-
	0.02	-0.110	-0.040	c	c	7.09	-
3.52	0.05	-0.110	-0.040	c	c	7.16	-
	0.10	-0.110	-0.040	c	c	7.27	-
	0.02	-0.110[b]	-0.040	-0.210	c	-	6.56
5.67	0.05	-0.111[b]	-0.040	-0.210	c	-	6.71
	0.10	c	c	-0.210	c	-	6.32

contd.

Table 4.15 contd.

	0.02	c	c	-0.220	-0.160	-	7.63
8.30	0.05	c	c	-0.220	-0.160	-	7.60
	0.10	c	c	-0.220	-0.160	-	7.59
	0.02	c	c	-0.220	-0.160	-	8.16
9.21	0.05	c	c	-0.220	-0.150	-	8.05
	0.10	c	c	-0.220	-0.140	-	8.26

a. CV data collected from sample solutions of conc 0.150 mM. Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b. Broad peak.

c. Peak absent.

Table 4.16. Cyclic voltammetric data on deprotonation of Cu(BPY)(Ser) in aqueous media.[a]

pH	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	i_p,A/$\nu^{1/2}$	i_p,C/$\nu^{1/2}$
	0.02	-0.110	-0.050	c	c	8.49	-
3.15	0.05	-0.120	-0.050	c	c	8.25	-
	0.10	-0.110	-0.040	c	c	8.52	-
	0.02	-0.115	-0.050	c	c	8.24	-
3.40	0.05	-0.115	-0.050	c	c	8.26	-
	0.10	-0.120	-0.040	c	c	8.38	-
	0.02	-0.120	-0.060	c	c	8.02	-
3.63	0.05	-0.120	-0.060	c	c	8.15	-
	0.10	-0.125	-0.055	c	c	8.25	-
	0.02	-0.140[b]	-0.060	c	c	8.12	-
3.88	0.05	-0.135[b]	-0.060	c	c	8.26	-
	0.10	-0.130[b]	-0.060	c	c	8.15	-

contd.

Table 4.16 contd.

	0.02	+0.120	-0.060	-0.210	c	7.42	0.71
4.51	0.05	-0.120	-0.060	-0.220	c	7.33	0.81
	0.10	-0.120	-0.070	-0.210	c	7.59	0.63
	0.02	-0.120	-0.060	-0.240	a	7.01	1.05
5.12	0.05	-0.120	d	-0.240	c	7.02	1.09
	0.10	-0.130	d	-0.240	c	7.21	1.20
	0.02	c	c	-0.220	-0.090	-	7.42
5.85	0.05	c	c	-0.210	-0.100	-	7.41
	0.10	c	c	-0.210	-0.100	-	7.59
	0.02	c	c	-0.220	-0.140	-	7.78
7.13	0.05	c	c	-0.220	-0.140	-	7.82
	0.10	c	c	-0.220	-0.135	-	7.91

a. CV data collected from solution of conc 0.186 mM. Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.

b. Broad peak.

c. Peak absent.

d. Masked by adsorption wave.

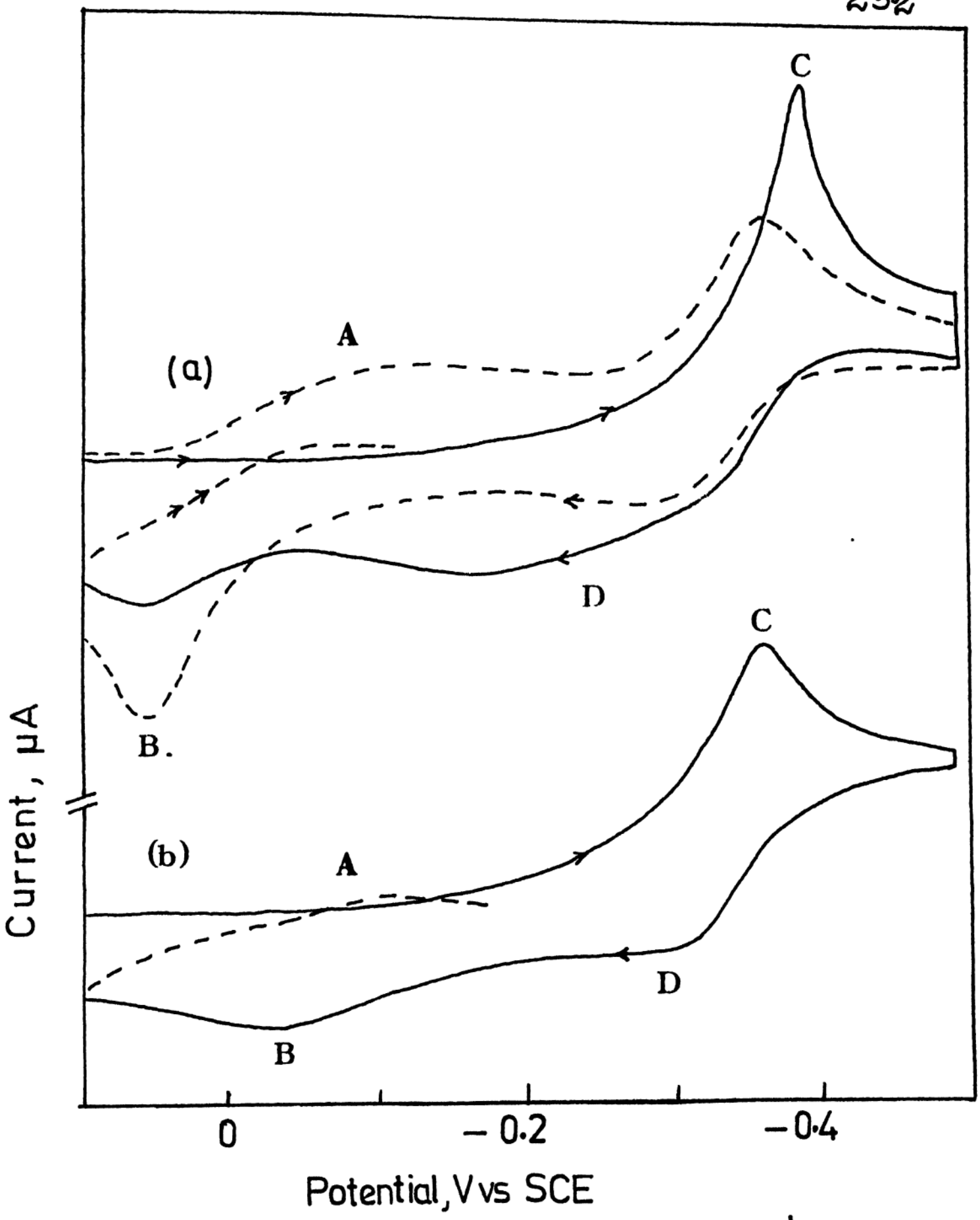


Fig.4.8 Cyclic voltammograms recorded at scan rate 0.02 Vs^{-1}.
(a) (——), Cu(His)(Gly) at pH = 8.2; (– – – –), Cu(His)(Gly) at pH = 5.5, (b) Cu(His)(Gly Gly) at pH – 8.0.

Table 4.17. Cyclic voltammetric data on reduction of histidine containing mixed amino acid copper(II) complexes in aqueous media.[a]

Compound	Sweep rate (Vs^{-1})	E_p,A	E_p,B	E_p,C	E_p,D	$i_p,C/\nu^{1/2}$
Cu(His)(Gly)	0.02	b	b	-0.380	c	84.85
(pH=8.3)	0.05	b	b	-0.390	c	82.73
	0.10	b	b	-0.400	c	82.72
Cu(His)(Ala)	0.02	b	b	-0.410	c	113.13
(pH=8.13)	0.05	b	b	-0.420	c	111.80
	0.10	b	b	-0.435	c	104.35
	0.20	b	b	-0.445	c	105.25
Cu(His)(Ser)	0.02	b	b	-0.375	c	78.48
(pH=8.20)	0.05	b	b	-0.385	c	78.26
	0.10	b	b	-0.395	c	79.05
	0.20	b	b	-0.405	c	78.24

contd.

Table 4.17 contd.

Cu(His)(Thr)	0.02	b	b	-0.365	c	74.24
(pH=8.2)	0.05	b	b	-0.375	c	73.79
	0.10	b	b	-0.385	c	72.73
	0.20	b	b	-0.395	c	73.56
Cu(His)(Gln)	0.02	b	b	-0.370	c	74.24
(pH=8.2)	0.05	b	b	-0.385	c	76.02
	0.10	b	b	-0.390	c	72.73
	0.20	b	b	-0.395	c	75.28
Cu(His)(Asn)*	0.02	b	b	-0.395	c	113.13
(pH=8.1)	0.05	b	b	-0.405	c	107.33
	0.10	b	b	-0.415	c	101.19
	0.20	b	b	-0.420	c	106.25

a. Potentials are expressed in volts and current function values in $\mu A(V/s)^{-1/2}$.
b. Peak absent; c. Very broad, difficult to measure precisely.
* Isolated as crystals.

The CV pattern for the histidine containing ternary complexes shown in Fig.4.8a is quite different from that of the binary and other ternary Cu(II) amino acid complexes. They have only one cathodic peak C in the potential range +0.1 to -0.6 V at pH about 7.8. This cathodic peak does not have the characteristics of a diffusion controlled electron-transfer process for the following reasons: (i) this peak increases in height drastically in the second and subsequent scans,(ii) if the second scan is carried out after holding the potential for a few minutes at a potential well-past the peak C, the increase in peak height is more pronounced,(iii) on diluting the test solution to about 5 times and on recording CV, these ternary systems become closer to simple binary systems in behaviour. These observations imply an adsorption process responsible for the peak C.[21]

The species responsible for the adsorption phenomenon is identified from the following observations. (i) At the pH of the experiment, the species in solution is known to be Cu(His)(A), (ii) The amino acid ligands in the absence of Cu(II) ions do not have any peaks in this potential range. (iii) The addition of stoichiometric amounts of L-histidine to an aqueous solution of $Cu(Gly)_2$ complex immediately changed the CV profile to that of

Cu(His)(Gly) complex. This is due to the rapid formation of the mixed complexes. It is known that stability of the complexes increases tremendously on ternary complex formation. (iv) On decreasing the pH of the solution, the adsorption complications of the peak C decreases and simultaneously the voltammogram begins to look similar to that of the binary complexes. It is known that at pH lower than about 7.0, the stability of the ternary complexes decreases and they get converted into other species predominantly to the binary complexes. These changes are clearly observable in Fig.4.8a. These observations suggest the ternary complex, Cu(His)(A) to be responsible for the adsorption peaks. The CV response of these types of complexes indicate that the diffusion-controlled process is masked by the adsorption phenomena which prevented the further investigations on these types of complexes.

B. Copper(II)-Histidine(His)-Dipeptide Type

The Cu(II) dipeptide complexes, were prepared in situ by mixing the components in water in 1:1:1 ratio and adjusting the pH.[16] The formation of the ternary complexes were confirmed by recording their visible spectra. They all have an absorption band at λ_{max} 610-615 nm.

Figure 4.8b shows the CV behaviour of Cu(His)(Gly Gly), a representative case of these ternary complexes. The CV data for all complexes are presented in Table 4.18. Unlike in the case of histidine containing ternary Cu(II) amino acid complexes, the cathodic peak C is free from adsorption complications. The peak characteristics like, $i_p/\upsilon^{1/2}$ is indopendent of scan rate, constancy of the peak potential with scan rates, and a constant value of about 60 mV for $E_p-E_{p/2}$, indicate that peak C originate from a one-electron reduction of the starting complex, Cu(His)(Gly Gly). The overall appearance of the CV of these complexes is very similar to the voltammograms of the binary complexes shown in Fig.4.1a. The partially developed anodic peak D corresponds to the re-oxidation of the reduced species. The other anodic peak B has the same origin as the peak B of the binary complexes discussed in chapter 2. Thus the overall electrode process can be represented by equations 4.6-4.8.

$$\text{Cu(II)(His)(Dipeptide)} \underset{}{\overset{1e}{\rightleftharpoons}} \text{[Cu(I)(His)(Dipeptide)]} \quad \ldots 4.6$$

$$\text{[Cu(I)(His)(Dipeptide)]} \xrightarrow{\text{chemical reactions}} \text{Cu}^{\circ}_{(Hg)} + \text{His} + \text{Dipeptide} \quad \ldots 4.7$$

$$\text{Cu}^{\circ}_{(Hg)} \overset{2e}{\rightleftharpoons} \text{Cu}^{2+}_{(aq)} \quad \ldots 4.8$$

Table 4.18. Cyclic voltammetric data on reduction of histidine containing ternary copper(II) dipeptide complexes.[a]

Compound	Sweep rate (Vs^{-1})	E_p,A	E_p,B [b]	E_p,C	E_p,D [b]
Cu(His)(Gly Gly)	0.02	-0.080	-0.030	-0.360	-0.285
(pH=7.92)	0.05	-0.080	-0.035	-0.365	-0.270
	0.10	-0.090	-0.040	-0.375	-0.270
	0.20	c	-0.030	-0.370	-0.280
Cu(His)(Gly Thr)	0.02	-0.080	-0.030	-0.365	-0.270
(pH=8.02)	0.05	-0.080	-0.040	-0.370	-0.280
	0.10	-0.090	-0.030	-0.375	-0.285
	0.20	c	-0.030	-0.380	-0.280
Cu(His)(Gly Ser)	0.02	-0.070	-0.035	-0.360	-0.280
(pH=7.82)	0.05	-0.080	-0.030	-0.360	-0 285
	0.10	-0.085	-0.040	-0.360	-0.285
	0.20	c	-0.035	-0.365	-0.280

contd.

Table 4.18 contd.

Cu(His)(Gly Leu)	0.02	-0.080	-0.035	-0.365	-0.280
(pH=7.82)	0.05	-0.090	-0.030	-0.370	-0.285
	0.10	-0.090	-0.035	-0.375	-0.280
	0.20	c	-0.040	-0.385	-0.285
Cu(His)(Gly Phe)	0.02	-0.075	-0.040	-0.355	-0.280
(pH=8.05)	0.05	-0.080	-0.030	-0.355	-0.285
	0.10	-0.080	-0.035	-0.360	-0.275
	0.20	c	-0.035	-0.370	-0.280
Cu(His)(Gly Met)	0.02	-0.080	-0.040	-0.355	-0.280
(pH=7.98)	0.05	-0.085	-0.035	-0.360	-0.285
	0.10	-0.090	-0.035	-0.365	-0.280
	0.20	c	-0.030	-0.370	-0.285

contd.

Table 4.18 contd.

Cu(His)(Gly Tyr)	0.02	-0.080	-0.035	-0.355	-0.280
(pH=8.02)	0.05	-0.080	-0.030	-0.360	-0.280
	0.10	-0.090	-0.040	-0.365	-0.285
	0.20	c	-0.035	-0.370	-0.280
Cu(His)(Gly Glu)	0.02	-0.080	-0.030	-0.360	-0.280
(pH=7.98)	0.05	-0.080	-0.035	-0.365	-0.285
	0.10	-0.090	-0.035	-0.370	-0.275
	0.20	c	-0.030	-0.375	-0.280
Cu(His)(Gly Asp)	0.02	-0.080	-0.030	-0.365	-0.280
(pH=8.04)	0.05	-0.090	-0.035	-0.370	-0.280
	0.10	-0.090	-0.040	-0.375	-0.285
	0.20	c	-0.040	-0.380	-0.280
Cu(His)(Gly His)	0.02	-0.080	-0.030	-0.385	-0.305
(pH=7.80)	0.05	-0.080	-0.040	-0.395	-0.300
	0.10	-0.090	-0.040	-0.405	--
	0.20	c	-0.040	-0.415	--

a. Potentials are expressed in volts; b. Broad peak;
c. Peak absent.

At higher scan rates an additional cathodic peak about 70 mV more negative to peak C is apparent. It has the characteristic features of an adsorption-controlled wave.[21]

It should be noted that the reduction potentials of all of the Cu(His)(Dipeptide) complexes are very close. This implies that the axial coordination by histidyl oxygen possible in some complexes and the alkyl substituents on the glycyl-end of the dipeptides do not have any appreciable influence on the reduction potentials. They are influenced solely by the in-plane coordination pattern in these complexes.

The major observations with regard to the cyclic voltammetric reduction of these ternary systems discussed in this chapter are summarised here. The Cu(A)(B) type ternary systems follow a pattern similar to the binary systems in their reduction as well as protonation processes. The Cu(BPY)(A) type complexes behave in a similar manner to that of binary complexes on reduction. However the end-product of protonation of Cu(BPY)(A) is identified to be $Cu(BPY)(H_2O)_2$. The histidine containing Cu(II) amino acid complexes show only an adsorption-controlled peak in place of peak C of other systems. The histidine containing dipeptide complexes behave similar to binary Cu(II) dipeptide systems.

4.4 Experimental

The ternary Cu(II) complexes of amino acids listed in Table 4.1 were prepared by a reported general procedure.[5b] It is briefly discussed below for the preparation of Cu(Glu)(Arg) complex. $Cu(ClO_4)_2.6H_2O$ (5 mM) L-arginine (5 mM) and L-glutamic acid (5 mM) were dissolved in 30 ml of water and the pH of the resulting solution was adjusted to about 9.0 with NaOH solution. The reaction mixture was stirred for 1-2 hours at room temperature and concentrated in vacuum to a small volume at temperature below 20°C. Addition of methanol to the residue gave blue crystals which were recrystallised from 1:1 aqueous methanol. The pH values of the reaction mixture containing L-ornithine or L-lysine were adjusted to about 7.0.

The complexes thus prepared were further purified by preparative tlc. The sample solutions were loaded on a cellulose (micro crystalline, 300 μ) and the chromatograms were developed by using isopropanol-water-1 M NaOH (60+38+2) solvent system. The developed plates were dried in air and the visualization of different components were achieved by using UV light and chemical methods, such as placing it in iodine-chamber or spraying ninhydrin and heating it to 110°C for 20-25 minutes. The spots due to

the ternary species were identified by comparing with that due to the possible binary complexes and that of the starting components. After the identification of the desired zones they were scrapped off into a beaker and was shaken with water for 5-10 minutes and centrifuged and the clear aqueous solution was evaporated to dryness at low temperature to get the complexes. The authenticity of the complexes were established by comparing their IR with that of the reported complexes.[3b]

Ternary Cu(II)(BPY)(A) type complexes listed in Table 4.9 were prepared by following a reported method.[11] This involves the addition of a solution of $CuSO_4.5H_2O$ (5 mM) in 20 ml of water to an ethanolic solution of 2,2'-bipyridyl (8 mM) in 20 mls of ethanol, with stirring. A blue solid was formed immediately and to this a solution of the amino acid (8 mM) in 10 ml of 0.1 M HCl was added with stirring. 1:1 Ammoniumhydroxide was added to the reaction mixture till a clear blue solution was obtained. The solution was heated to about 50°C and stirred for another 30 minutes and concentrated. The concentrated solution on keeping in the refrigerator for a few days yielded blue needle-shaped crystals which were filtered, washed with cold water followed by ethanol and dried in vacuum at room temperature. The complexes were further

purified by prep. tlc. The procedure adopted was similar to that in the previous case except for that silica gel and n-butanol-water-methanol (20+40+40) solvent system were used instead of cellulose and the other solvent system. The chromatographed samples were used to record CV after establishing its identity and authenticity by spectral methods.

The histidine containing ternary copper(II) complexes were prepared by mixing the components in 1:1:1 ratio in 1 mM concentration and adjusting the pH to 7.5-8.0 to ensure complete formation of the complexes.[3b,4,16] Cyclic voltammograms of these solutions were recorded after checking its authenticity by spectral methods.

The details of the cyclic voltammetric set up used is discussed in detail in chapter 2. The ternary bipyridyl complexes used were of very low concentrations, below 0.5 mM, to avoid adsorption complications.

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VITAE

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Publications

1. 'Oxidation-Reduction of Nitroxyl Radicals: Cyclic Voltammetric Response in Aqueous Media', George Thomas and J.G. Mohanty, Indian J. Chem., 1982, 21A, 451.

2. 'Cyclic Voltammetric Studies of Copper(II) Dipeptide Complexes. Evidence for Copper(I) Dipeptide Complexes in Aqueous Media', George Thomas and P.S. Zacharias, Polyhedron, in press.

3. 'Histidine Containing Mixed Amino Acid and Dipeptide Copper(II) Complexes. Cyclic Voltammetric Studies of the Reduction in Aqueous Media', George Thomas and Panthappally S. Zacharias, Transition Met. Chem., in Press.

4. 'Cyclic Voltammetric Studies of Copper(II) Amino Acid Complexes. Electrochemical Evidence for the Generation of Copper(I) Complexes as an Intermediate', George Thomas and P.S. Zacharias, submitted for publication in Polyhedron.

5. 'Mixed Ligand Copper(II) α-Amino Acid Complexes with Intramolecular Ligand-Ligand Interactions. A Cyclic Voltammetric Studies on the Reduction in Aqueous Media', George Thomas and P.S. Zacharias, *accepted* for publication in Indian J. Chem.

6. 'Mixed Ligand Copper(II) Amino Acid Complexes containing 2,2'-Bipyridyl. Cyclic Voltammetric Studies on the Reduction in Aqueous Media', George Thomas and P.S. Zacharias, Polyhedron, in Press.

Presentations

1. 'Protonation - Deprotonation studies on some Copper(II) Dipeptide complexes by Cyclic Voltammetry', P.S.Zacharias and George Thomas, presented at the All India Conference on 'Recent Trends in Coordination Chemistry' held at Hyderabad - 1981.

2. 'Cyclic Voltammetric Studies of Copper(II) Dipeptide complexes', George Thomas and P.S. Zacharias, presented at the International Symposium on 'Recent Aspects of Electroanalytical Chemistry and Electrochemical Technology' held at Chandigarh, 1982.