METAL COMPLEXES OF AMINO ACID DERIVATIVES

A thesis presented to the University of Surrey for the degree of Master of Philosophy in the Faculty Science

by

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DEDICATION

To

Almighty Allah

who blessed me with sound health and mind, I dedicate this work.
The author wishes to express his sincere thanks to his supervisors Drs K.B. Nolan and L.F. Larkworthy for their advice, help and continuous encouragement throughout the course of the work. Thanks are also due to other members of the chemistry department, particularly the technical staff who assisted me in my work.

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I am overwhelmed by a feeling of gratitude to all the members of my family, for their cooperation, encouragement and exemplary patience during the period of my absence from home.

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<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Definition</th>
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<tbody>
<tr>
<td>Me</td>
<td>Methyl</td>
</tr>
<tr>
<td>Et</td>
<td>Ethyl</td>
</tr>
<tr>
<td>DMF</td>
<td>Dimethylformamide</td>
</tr>
<tr>
<td>R</td>
<td>Any alkyl group</td>
</tr>
<tr>
<td>M</td>
<td>Metal</td>
</tr>
<tr>
<td>X</td>
<td>Halide</td>
</tr>
<tr>
<td>B.M</td>
<td>Bohr magneton</td>
</tr>
<tr>
<td>L</td>
<td>Ligand</td>
</tr>
<tr>
<td>M.P</td>
<td>Melting point</td>
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<tr>
<td>B.P</td>
<td>Boiling point</td>
</tr>
<tr>
<td>en</td>
<td>Ethylenediamine</td>
</tr>
<tr>
<td>tren</td>
<td>2,2',2''-triaminotriethylamine</td>
</tr>
<tr>
<td>Gly</td>
<td>Glycine</td>
</tr>
<tr>
<td>Gly-Gly</td>
<td>Glycylglycine</td>
</tr>
<tr>
<td>Fig</td>
<td>Figure</td>
</tr>
<tr>
<td>n.m.r</td>
<td>Nuclear magnetic resonance</td>
</tr>
<tr>
<td>i.r</td>
<td>Infrared</td>
</tr>
<tr>
<td>u.v</td>
<td>Ultraviolet</td>
</tr>
<tr>
<td>V</td>
<td>very</td>
</tr>
<tr>
<td>b</td>
<td>broad</td>
</tr>
<tr>
<td>s</td>
<td>strong</td>
</tr>
<tr>
<td>Sh</td>
<td>Shoulder</td>
</tr>
<tr>
<td>m</td>
<td>Medium</td>
</tr>
<tr>
<td>w</td>
<td>Weak</td>
</tr>
</tbody>
</table>
This thesis deals with various aspects of the chemistry of metal complexes with amino acid derivatives and related ligands. The introduction covers background literature on these complexes particularly in relation to their reactions in solution. A number of copper(II) and nickel(II) complexes of the ligand 2-(2'-acetoxyethylamino)ethylamine, \( \text{NH}_2\text{(CH}_2\text{)}_2\text{NH(CH}_2\text{)}_2\text{OCOCH}_3 \), have been synthesised and characterised by spectroscopic and magnetic measurements. The complexes isolated depend on whether or not the non-complexing base, triethylamine, is added to the reaction mixture, which contains the ligand (L) as the dihydrochloride or dihydrobromide. Hence complexes such as \( \text{LH}_2\text{[CuBr}_4\text{]} \) or \( \text{CuLBrg} \) may be obtained, depending on the conditions.

The tetramethylamide of EDTA has been synthesised and its complexes with a number of metal ions (e.g. Cu(II), Ni(II), Co(II), Zn(II), Fe(III), Cd(II)) have been isolated. In these complexes the ligand is either a tetradentate 2N,2O donor or a hexadentate 2N,4O donor.

The metal ion promoted hydrolysis of diimides of EDTA and related compound are also described. In particular the hydrolysis of 1,2-bis(3,5-dioxopiperazin-1-yl)propane in the presence of copper(II) has been investigated in detail. This compound is anticancer drug, razoxane, and its activity is due to the fact that it is metabolised in vivo to a chelating agent which interferes with metalloenzymes necessary for the growth of tumour cells. In the copper(II) promoted hydrolysis of razoxane only one of the imide rings is opened but this produces a chelating agent which, based on its structure, should strongly bind metal ions.
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CHAPTER 1
Introduction
1. Introduction

Metal complexes of ligands with amino acids esters, amides, imides, peptides and polypeptides have been widely studied during the past two decades.\textsuperscript{1-3} Currently the main topic of interest is the metal promoted hydrolysis of organic substrates.\textsuperscript{4} Metal ions have been found to promote the hydrolysis of amino acid esters, amides and anhydrides.\textsuperscript{5-7} In these reactions the metal ion polarises the scissile carbonyl bond by binding the carbonyl oxygen, and makes it more susceptible to attack by external nucleophiles such as hydroxide ion. Alternatively it may facilitate reaction between the carbonyl group and a juxtaposed hydroxy ligand in its coordination sphere. In addition, decreases in the basicity of leaving group resulting from coordination are sometimes responsible for the high reactivity.\textsuperscript{8,9}

Metal ion catalysis in the reaction of acyl derivatives also serves as a model for carboxypeptidase A (CPA), a zinc containing exopeptidase.\textsuperscript{10} Catalysis by CPA has been proposed to involve an anhydride intermediate and the Glu-270 carboxylate of CPA\textsuperscript{11} as the attacking nucleophile. Various shapes of the pH profile of $K_{cat}$ for CPA catalysed hydrolysis of ester substrates have been explained in terms of water attack on the anhydride intermediate.\textsuperscript{12}

Many of the early kinetic studies of the hydrolysis of coordinated substrates were hampered by the difficulty in identifying the hydrolytically most active species in solution. The difficulty arises from the fact that solutions containing metal ions (M\textsuperscript{+}) and ester, amide or peptide ligands (L) may contain a large number of complex species for example $ML^{n+}$, $ML_2^{n+}$, $ML_3^{n+}$ etc. The most abundant
species will depend on the metal to ester ratio, and inevitably there is more than one complex present. In addition the mixed ligand complexes $[M(L)(A)]^{(n-1)+}$, $[M(L)_{2}(A)]^{(n-1)+}$ etc will also form in solution as hydrolysis of the ligand $L$ to $A$ the product proceeds. This problem can be overcome by using a polydentate ligand which forms only a 1:1 complex with metal ions e.g the tetramethyl ester of EDTA (1).\textsuperscript{13-14} This approach has considerably simplified the treatment of kinetic data. The most recent developments in the area have come from the use of inert cobalt (III) systems and it has been possible to resolve many of the mechanistic problems which have arisen from studies of other kinetically labile systems.\textsuperscript{15-16}

\[
\begin{align*}
\text{H}_3\text{CO}_2\text{C} & \equiv \text{H}_2\text{C} \\
\text{CH}_2 & \equiv \text{CH}_2 \\
\text{N} & \equiv \text{N} \\
\text{H}_2\text{C} & \equiv \text{C} \\
\text{N} & \equiv \text{C} \\
\text{M} & \equiv \text{CH}_2 \\
\text{H}_3\text{CO} & \equiv \text{OCH}_3 \\
\end{align*}
\]

(1)

This thesis describes the preparation and characterisation of ester and amide type ligands and their complexes as well as kinetic investigations of hydrolysis reactions. An intramolecular mechanism for the hydrolysis of a ligand in a labile metal complex has been established to the first time.
1. Aminoacid Ester Complexes Containing Coordinated Ester Groups

In the early 1930's the hydrolysis of peptides was found to be subject to metal ion catalysis. In 1952 Kroll discovered that the hydrolysis of amino acid esters was also catalysed by metal ions. Kroll suggested that the complex of ethylglycinate with copper(II), cobalt(II) and manganese(II) underwent relatively rapid base hydrolysis at pH 7, conditions under which hydrolysis of the free ester was unobserved. Kroll explained the catalytic behaviour of the metal ion in terms of its ability to polarise the ester carbonyl group, (2), and consequently to increase its susceptibility to attack by the nucleophilic hydroxide ion. This mechanism is similar to that involved in the acid catalysed hydrolysis of esters.

\[
\begin{align*}
\text{CHR} - C - OR' \\
\text{H}_2\text{N} - C - O - \text{M}
\end{align*}
\]

(2)

Polarisation of the carbonyl group by metal ions has since been verified by infrared spectral studies of metal complexes containing glycine ester and alanine ester ligands. Amino acid ester ligands can be monodentate or bidentate, and examples of complexes containing both modes of coordination have been isolated and their solid state infrared spectra studied. Thus, formation of the chelated ester species leads to a reduction in the carbonyl stretching frequency from 1740 cm\(^{-1}\) (4) to 1600 cm\(^{-1}\) (3) indicating significant polarisation of the carbonyl bond by the metal ion.
(a) Formation and Reactions of Cobalt(III) Complexes of Glycine and Glycine Esters.

Alexander and Bush first described the preparation of cobalt(III) complexes of glycine$^{20}$ and glycine esters.$^{21}$ Complexes of the type (5), $\text{cis} - [\text{Co(en)}_2(\text{NH}_2\text{CH}_2\text{CO}_2\text{R})\text{Cl}]\text{Cl}_2$ were prepared by reacting the appropriate amino acid ester hydrochloride with trans-dichlorobis(ethylenediamine)-cobalt(III)chloride in aqueous solution. In such complexes the free amino acid ester was generated in situ by the presence of a weakly coordinating base such as diethylamine.

\[
\text{Trans} - [\text{Co(en)}_2\text{Cl}_2]\text{Cl} + \text{Cl}^- \text{NH}_2\text{CH}_2\text{CO}_2\text{R} \quad \text{---} \quad \text{cis} - [\text{Co(en)}_2(\text{NH}_2\text{CH}_2\text{CO}_2\text{R})\text{Cl}]\text{Cl}_2 \quad \cdots \quad (1)
\]

\[
(5)
\]
They also studied the mercury(II) promoted hydrolysis of the cis–

\[ [\text{Co(en)}_2\text{Cl(GlyOR)}]^2+ \] ion in acid solution and proposed that a chelated ester species (8) was the reactive intermediate in the \( \text{Hg}^{2+} \) promoted reaction. The \( \text{Hg(II)} \) ion assists removal of the coordinated chloride ion to give a five coordinate species, and the ester carbonyl oxygen competes so effectively with solvent water for the vacant coordination site, that the chelated ester (8) complex is formed.

This complex (8), on standing, undergoes a reaction involving hydrolysis of the highly activated ester group. In acidic solutions (pH<4) the rate expression for the hydrolysis reaction is

\[
\text{Rate} = k[\text{Ester complex}(8)]
\]

indicating that attack by the relatively weak nucleophile, water, occurs. In solution of pH>7 however, hydroxyl ion is the attacking nucleophile and the rate expression

\[
\text{Rate} = k_2[\text{Ester complex}(8)][\text{OH}^-]
\]

is observed for the hydrolysis reaction, which is very rapid under these conditions (reaction (3)).

Metal ion activation of the ester group with respect to attack by nucleophiles has also proved extremely useful synthetically. Thus chelated ester complexes like (8) in nonaqueous media are attacked by aminoacid esters (reaction 4), amines, and alcohols to give complexes containing peptide esters, amides and transesterified ligands respectively.\(^2\text{3}\)

Reaction (4) provides an example of a situation where a metal ion singularly plays the role of two reagents commonly used in peptide synthesis.\(^2\text{4}\) It activates the carbonyl group to nucleophilic attack, fulfilling the role of coupling agents.
such as dicyclohexylcarbodiimide. Secondly it protects the sensitive amino group on the coordinated aminoacid ester and in this way functions like N- protecting groups such as carbobenzoxy and dimedone (5, 5-dimethyl cyclohexane 1,3-dione).

\[
\begin{align*}
\text{(en)}_2 \text{- Co} \begin{array}{c}
\text{N} \text{H}_2 \text{- CH}_2 \text{- CO}_2 \text{H}_5
\end{array} \begin{array}{c}
\text{Cl}
\end{array} \begin{array}{c}
\text{C} \text{O}
\end{array} \text{Cl} \begin{array}{c}
\text{C} \text{O}
\end{array} \begin{array}{c}
\text{N} \text{H}_2 \text{- CH}_2 \text{- CO}_2 \text{H}_5
\end{array}
\end{align*}
\]

\[
\text{Hg(II)} \xrightarrow{-\text{HgCl}^+} \text{slow}
\]

\[
\begin{align*}
\text{(en)}_2 \text{- Co} \begin{array}{c}
\text{N} \text{H}_2 \text{- CH}_2 \text{- CO}_2 \text{H}_5
\end{array} \begin{array}{c}
\text{Cl}
\end{array} \begin{array}{c}
\text{C} \text{O}
\end{array} \begin{array}{c}
\text{N} \text{H}_2 \text{- CH}_2 \text{- CO}_2 \text{H}_5
\end{array} \begin{array}{c}
\text{Cl}
\end{array} \begin{array}{c}
\text{C} \text{O}
\end{array} \begin{array}{c}
\text{N} \text{H}_2 \text{- CH}_2 \text{- CO}_2 \text{H}_5
\end{array}
\end{align*}
\]

\[
\begin{align*}
\text{(en)}_2 \text{- Co} \begin{array}{c}
\text{N} \text{H}_2 \text{- CH}_2 \text{- CO}_2 \text{H}_5
\end{array} \begin{array}{c}
\text{Cl}
\end{array} \begin{array}{c}
\text{C} \text{O}
\end{array} \begin{array}{c}
\text{N} \text{H}_2 \text{- CH}_2 \text{- CO}_2 \text{H}_5
\end{array} \begin{array}{c}
\text{Cl}
\end{array} \begin{array}{c}
\text{C} \text{O}
\end{array} \begin{array}{c}
\text{N} \text{H}_2 \text{- CH}_2 \text{- CO}_2 \text{H}_5
\end{array} \begin{array}{c}
\text{Cl}
\end{array} \begin{array}{c}
\text{C} \text{O}
\end{array} \begin{array}{c}
\text{N} \text{H}_2 \text{- CH}_2 \text{- CO}_2 \text{H}_5
\end{array}
\end{align*}
\]
(b) The Copper (II) Complex of the Tetramethyl Ester of EDTA

Copper (II) forms a 1:1 complex (11) with the tetramethyl ester of EDTA in aqueous solution. This was confirmed by infrared spectroscopy which showed that two of the methoxy carboxyl groups are coordinated to the metal ion. The coordinated ester groups give rise to carbonyl stretching frequencies at 1705 cm$^{-1}$ in the infrared spectrum of the complex, while the remaining two ester groups have carbonyl stretching frequencies at 1730 cm$^{-1}$. 
Since the carbonyl stretching frequencies of the free ligand, EDTA (OMe), occur at 1740 cm\(^{-1}\), it may well be that the latter two ester groups are weakly coordinated in axial positions to the metal ion, in which case the ligand is hexadentate. Otherwise the ligand may be quadridentate with these ester groups remaining uncoordinated and aquo ligands occupying the fifth and sixth coordination sites of the metal ion. Direct coordination to the metal ion results in two
of the ester carbonyl groups in the above complex being highly activated to attack by nucleophiles such as OH, H$_2$O. Consequently the tetraester complex is rapidly base hydrolysed to the corresponding diesterdicarboxylato complex (12), even in aqueous solutions of very low hydroxide ion concentrations (pH<6). Parallel hydrolytic pathways involving attack by the weak nucleophile, H$_2$O, on the coordinated ester groups also occurs. The hydrolysis of the tetraester complex is summarised in reaction (5).

The second order rate constants $k_1$ and $k_2$ for bimolecular hydroxide ion attack on the above substrates are both some $10^6$ times faster than the second order rate constant for hydrolysis of the ester groups in the free ligand EDTA (OMe)$_4$ . Additional evidence for metal ion activation of the ester groups is that attack by H$_2$O, although slow in the case of the complexes (the nucleophilicity of H$_2$O is a factor of $10^{11}$ smaller than the nucleophilicity of OH$^-$ with respect to complexes) is unobservable for the free ligands.
\[
\left[ \begin{array}{c}
\text{H}_3\text{CO}_2 \text{C-} \text{H}_2 \text{C} \\
\text{CH}_2-\text{CH}_2 \\
\text{CH}_2\text{CO}_2 \text{CH}_3
\end{array} \right]^{2+}
\]
\[
\xrightarrow{+\text{OH}^-, k_1}
\]
\[
\left[ \begin{array}{c}
\text{H}_3\text{CO}_2 \text{C-} \text{H}_2 \text{C} \\
\text{CH}_2-\text{CH}_2 \\
\text{CH}_2\text{CO}_2 \text{CH}_3
\end{array} \right]^{+}
\]
\[
\xrightarrow{+\text{OH}^-, k_2}
\]
\[
\left[ \begin{array}{c}
\text{H}_3\text{CO}_2 \text{C-} \text{H}_2 \text{C} \\
\text{CH}_2-\text{CH}_2 \\
\text{CH}_2\text{CO}_2 \text{CH}_3
\end{array} \right]
\]
\[
\xrightarrow{+\text{H}_2\text{O}, -\text{H}^+, k_1'}
\]
\[
\left[ \begin{array}{c}
\text{H}_3\text{CO}_2 \text{C-} \text{H}_2 \text{C} \\
\text{CH}_2-\text{CH}_2 \\
\text{CH}_2\text{CO}_2 \text{CH}_3
\end{array} \right]
\]
\[
\xrightarrow{+\text{H}_2\text{O}, -\text{H}^+, k_2'}
\]
2. Aminoacid Ester Complexes Containing Uncoordinated Ester Groups

A number of divalent metal complexes of ethyl and methyl glycinate, methyl α-alanine and cysteine methyl esters have been characterised and their solid state infrared spectra studied. Many of these complexes are of the type MX₂(NH₂ - CHR - CO₂R') (X = Cl, Br) in which the ester ligand is monodentate and coordinated to the metal via the amino group. Information on these complexes has been confined to their infrared spectra with little attention having been devoted to the kinetics of ester hydrolysis. However, the few available results given on these kinetic studies are inconclusive and conflicting, due in part to the low stability constants of the complexes in aqueous solution.

In the complex cis-[Co(en)₂(glyOR)OH]²⁺ (14), (R = -CH₂(CH₃)₂), which is obtained by Br⁻ removal from cis-[Co(en)₂(glyOR)Br]²⁺ in weakly alkaline solutions, the ester group although not directly coordinated to the metal ion, is nevertheless very sensitive to base hydrolysis. This occurs by a mechanism involving attack by neighbouring coordinated OH⁻ on the carbonyl group (reaction 6).

The rate enhancement for this process over the hydrolysis of the free ester is comparable in magnitude to those earlier encountered for directly coordinated esters. The metal ion presumably contributes to the ease of the reaction by holding the two reactants in juxtaposition. Moreover chelate ring formation helps to stabilise the transition state of the reaction relative to the ground state, thus lowering the energy of activation. These factors lead to an extraordinarily efficient
process in view of the fact that the nucleophilicity of OH\(^-\) is decreased by a factor of at least 10\(^7\) on coordination to the metal ion.

In a related study, the complex \[\text{[Co(NH}_3\text{)]}_6(\text{NH}_2\text{-CH}_2\text{-CO}_2\text{C}_2\text{H}_5})\]\(^{5+}\) was found to undergo intramolecular amidolysis in alkaline solutions.\(^{26}\) The product \[\text{[Co(NH}_3)_4\text{NH}_2\text{-CH}_2\text{-CONH})\]\(^{2+}\), which contains glycine imide chelated through both nitrogen atoms, results from intramolecular attack by coordinated NH\(_3^-\), the conjugate base of an NH\(_3\) ligand, on the ester carbonyl group with concomitant loss of C\(_2\)H\(_5\)OH.

Metal complexes of amino acid esters with two or more nitrogen, sulfur and oxygen donor atoms have been widely investigated in respect of structural and kinetic viewpoints.\(^{17,26-32}\) These complexes are in general thermodynamically more
stable in solution than complexes containing monodentate amino-acid esters and as a result ester hydrolysis reactions in these systems are more amenable to kinetic study.

Methyl 2, 3- diaminopropionate\(^\text{30}\) (15), methyl histidinate (16) and methyl cysteinate\(^\text{27-29}\) (17) form 1:1 or 2:1 complexes with metal ions under appropriate conditions. Infrared spectral measurements provide no evidence for bonding between the metal ion and the ester function in the solid state or in aqueous solution. Methyl cysteinate forms neutral complexes with bivalent metal ions. As expected the neutral bis (methylcysteinate) complex undergoes base hydrolysis reactions more slowly than the analogous positively charged complexes of the other ligands.\(^\text{32}\) The catalytic activity of the various divalent metal ions \(\text{Ni} > \text{Pb} > \text{Zn} > \text{Cd} > \text{Hg}\) correlates quite well with the order of metal nitrogen bond strengths obtained from infrared spectral measurements\(^\text{28,33}\) on the complexes. This rough correlation of hydrolytic reactivity with metal nitrogen bond strength probably implies that the catalytic activity in these neutral complexes is due to the inductive effect of the metal ion acting through the amino nitrogen. Withdrawal of electrons from the acyl carbon atom facilitates nucleophilic attack by hydroxide ion.

A number of kinetic studies at different metal to ligand ratios have been reported for Cu(II) and Ni(II) complexes with L-methyl histidinate,\(^\text{26,29,34}\) and for Cu(II) and Hg(II) complexes with methyl DL-2,3- diaminopropionate.\(^\text{35}\) In the case of the bis- (methyl histidinate) copper (II) complex cation, hydrolysis is 265 times more rapid compared to free ester, even after the statistical advantage is accounted for. The rate enhancement however is less than 4 relative to the monoprotonated ester.\(^\text{36}\)
3. Metal Complexes of Glycinamide and Related Complexes.

The amide group has been widely studied as a monodentate donor in simple species such as acetamide, and dimethylformamide and in cyclic systems such as γ-butyrolactam. More recently, however, much more attention has been given to bidentate and tridentate amide ligands containing nitrogen donor atoms additional to the amide group.

**Glycinamide and Related Complexes**

The copper (II) complex of glycinamide was isolated in 1932. Neutrality was assumed to be achieved by ionization of the amide group of glycinamide. Later this was confirmed by potentiometric titration results, which showed that an aqueous solution mixture of glycinamide and Cu(II), present in a 2:1 molar ratio, consumed two moles of base per mole of metal ion. A similar Cu(II) complex of 1-leucinamide was also isolated.

Several planar 2:1 aminoacid amide complexes of Ni(II) and Pd(II) have also been synthesised and characterised spectroscopically. Pd(II) forms both cis and trans complex with L-alanine and L-leucine amidates but only a trans complex with glycinamidate (18). Ni(II) forms only trans complexes with these ligands. Cationic complexes of Ni(II) with amino-acid amides have also been isolated under neutral conditions.

The effect of Cu(II) on the hydrolysis of glycinamide has been investigated in the pH range 6- 12. In near neutral solutions the metal ion promotes the hydrolysis of the amide, by forming the hydrolytically active cationic complex [Cu...
At high pH, however, this complex is converted to the neutral glycinamidate complex Cu(NH$_2$ - CH$_2$ - CONH)$_2$ which is insensitive to base hydrolysis.

A variety of N, O chelated glycinamide complexes of the type [Co(en)$_2$(NH$_2$ - CH$_2$ - CONRR'')]($\text{NO}_3$)$_2$ ClO$_4$ (R = H, CH$_3$; R' = H, CH$_3$) have been prepared and hydrolysis of the amide ligands investigated in the pH range 8-14.$^{43}$ In the case of the R = R' = H and R = H, R' = CH$_3$ complexes ionization of the amide groups (pKa $\approx$ 11) occurs in this pH range and the kinetic data have been interpreted in terms of equilibria involving the hydrolytically active amide complexes and their amideate conjugate bases, which are hydrolytically inactive. As a result the rate of base hydrolysis shows a first order dependence on [OH$^-\text{]}$ at low pH (where only the conjugate acid is present) but an independence of [OH$^-\text{]}$ at high pH. In the N, N- dimethylglycinamide complex (R = R' = CH$_3$) the absence of an amide proton
rules out the possibility of amide group ionization and first order dependence on \([\text{OH}^-]\) was observed for base hydrolysis of this complex over the entire pH range. In the case of all three amides however the metal ion causes a rate enhancement of about \(10^6\) for base hydrolysis.

The complex cis-[Co(en)_2(NH_2-CH_2-CO NH_2)(OH)]^{2+}, (19), which contains the monodentate glycinamide ligand, has been identified as one of the products of Br^- removal from the complex cis-[Co(en)_2(NH_2-CH_2-CO NH_2)Br]^{2+} in aqueous base.\(^4\) The hydroxy complex undergoes a rapid intramolecular hydrolysis reaction involving attack by coordinated OH^- on the adjacent amide group (reaction 7).
4. Peptide Complexes

A number of discussions are available on the general topic of metal peptide complexes.\(^{45,46}\)

The complex diaquo-glycylglycinato copper (II) hydrate has been crystallized from solution and its crystal structure determined.\(^ {47}\) The peptide molecule behaves as a tridentate chelate via its terminal amino groups, peptide nitrogen atom and carboxyl groups, with a water molecule completing the square planar structure. A much more loosely bound water molecule occurs perpendicular to the plane and the third water molecule links the structure by means of hydrogen bonds.

Nickel (II) also induces the ionization of the amide hydrogen atoms of peptides to give complexes with peptide nitrogen donors.\(^ {48}\) During this ionization Ni (II) complexes of glycyl peptides with three or more residues change colour from blue to yellow corresponding to a transition from a paramagnetic, octahedral to a diamagnetic square planar species. In the case of the glycylglycine complex, however, no such colour change is observed on amide group ionization and the conjugate base was shown to be the octahedral complex sodium bisglycylglycinato-nickel (II) in which the glycylglycinato dianion acts as an N, N, O tridentate ligand. Pd (II) is more effective than either Ni (II) or Cu (II) in promoting peptide group ionization (pKa \(\approx 3.5\)) and complexes of this metal ion containing peptide nitrogen donor atoms are readily obtained.\(^ {49}\)

Oxidation of Co (II) complexes of glycylglycine (Gly - Gly \(H_2\)) led to the isolation of the crystalline complexes \([Co(H_2O)_6]\), \([Co(Gly - Gly)\_2\int_{12}^{17}]\) and \(Ba[Co(Gly - Gly)\_2\int_{12}^{17}]\).\(^ {50}\) All three complexes contain chemically identical \([Co(Gly - Gly)\_2^-] an-\)
ions in which the ligand is tridentate through two nitrogen and one oxygen donor atoms. Acidification of solutions containing \([\text{Co(Gly - Gly)}_2]^-\) leads to the rapid and reversible uptake of the two protons by the peptide oxygen atoms yielding the cationic complex \([\text{Co(Gly - Gly H)}_2]^+\).

5. Metal Ion Promoted Peptide Hydrolysis

It has been found that a number of complexes of the type \([\text{CoN}_4(\text{OH}) (\text{OH}_2)]^{2+}\) (\(\text{N}_4 = \text{a system of four nitrogen donor atoms i.e., 2 en, trien or tren})\) stoichiometrically cleave the N-terminal amino-acid from di- or tri-peptides.\(^{51}\) The proposed mechanism for this reaction involves replacement of the aquo ligand by the peptide (through its amino groups) followed by intramolecular attack by the adjacent hydroxyl ligand on the N-terminal peptide carbonyl group. Evidence for this pathway is provided by the fact that the trans-isomer of the above complex is inactive and N-protected peptides are not hydrolysed.

A number of N, O chelated complexes of the type \([\text{CoN}_4(\text{Gly - Gly H})_2]^{2+}\) have been prepared and their base hydrolysis reaction studied.\(^{43}\) The metal ion causes rate enhancements of \(\sim 10^5\) relative to base hydrolysis of the free ligand. As in the case of Cu (II) the complexes \([\text{CoN}_4(\text{Gly - Gly H})]^{2+}\) obtained by peptide group ionization are insensitive to base hydrolysis.\(^{52}\)
References


CHAPTER 2
Experimental Techniques
1. Spectroscopic Methods

The infrared absorption spectra of the ligands and their complexes were obtained on a Perkin Elmer 577 grating infrared spectrophotometer. Solid samples as nujol mulls were examined using sodium chloride plates in the wavenumber range 625-4000 cm$^{-1}$. The solid state electronic spectra of the complexes were recorded in the wave number range 12,000-50,000 cm$^{-1}$ on a Beckman Acta MIV spectrophotometer fitted with a reflectance accessory using barium sulphate as reference. The electronic solution spectra were recorded in the wavenumber range 300-700 nm on a Pye Unicam SP 8000 u.v-visible recording spectrophotometer. Distilled water was used as the solvent. The $^1$H n.m.r spectra were recorded on a Bruker WH 90 magnetic resonance spectrometer. Melting points were determined on a Kolfer micro hot stage using a standarised thermometer. Microanalyses were performed by the micro analytical service, Chemistry Department, University of Surrey, Guildford.

2. Potentiometric Methods

Proton ionization constants of amide ligands when complexed with copper were obtained by potentiometric titration. The kinetics of base hydrolysis of the imide complexes were investigated by the pH-stat technique. Both of these operations were carried out using a Radiometer pH meter 26, with an Automatic Titrat- or 11, an autoburette unit ABU12 to deliver the titrant and a titrigraph SBR2 to monitor the addition of titrant versus pH (in potentiometric titration) or time (in pH-stat kinetics). A high alkalinity glass electrode was used in conjunction with
a saturated calomel electrode as reference. The electrode system was standardised at 25.0 °C with 0.05M potassium hydrogen phthalate (pH 4.01) and 0.01M sodium tetraborate (borax, pH 9.81) buffers. The reaction solution was contained in a double walled vessel (50cm³) through which water from a constant temperature water bath was circulated and this was fitted with a cover which was designed to accommodate the electrode system, nitrogen inlet and outlet tubes, a titrant inlet tube and stirrer. The whole apparatus was thermostatted at 25.0±0.1 °C. All kinetics studies were carried out at a constant ionic strength of 0.3 by the use of the appropriate concentration of sodium perchlorate.

3. Magnetic Measurements

Magnetic susceptibilities were measured at room temperature by the Gouy method. HgCo(CNS)₄ was used as the susceptibility standard and the measurements were made with a Johnson Matthey JME magnetic susceptibility balance. The mass susceptibility $X_\rho$ was calculated using the equation

$$X_\rho = \frac{C L (R - R_0)}{10^6 m}$$

where $L$ is the sample length (cm)

$m$ is the sample mass (g)

$R_0$ is the reading of empty sample tube

$R$ is the reading of tube with sample

$C$ is the calibration constant.

The molar susceptibility $\chi_m$ of each complex was calculated using the expres-
where $M$ is the molecular weight of the complex. The molar susceptibility of the metal ion $\chi_A$, was calculated by allowing for the additive diamagnetic susceptibility of the rest of the molecule, $(\chi_L)$ viz

$$\chi_A = \chi_m - \chi_L$$  \hspace{1cm} (3)

The effective magnetic moment of the metal ion, $\mu_e$ in Bohr magnetons, was obtained using the expression

$$\mu_e = 2.828 \sqrt{\chi_A T}$$  \hspace{1cm} (4)

where $T$ is the absolute temperature.

4. EDTA Titrations

The relative molecular mass of the complex was obtained by using a specific indicator to obtain the metal percentage. It was assumed that 1 mole of EDTA was equivalent to 1 mole of metal ion.\(^7\)

5. Kinetic Methods

(a) Ester Hydrolysis

The base catalysed hydrolysis of an amino acid ester whether complexed or uncomplexed may be represented by the general equation

$$RCOOR' + OH^- \rightarrow RCO_2^- + R'OH$$  \hspace{1cm} (5)
where $R$ is a metal ion containing group in the case of an ester complex. This reaction is bimolecular and obeys the rate expression

$$Rate = k_2[R - COOR'][OH^-]$$

(6)

If $[OH^-]$ is maintained constant, pseudo first order conditions are satisfied and the rate equation simplifies to

$$Rate = k_{obs}[R - COOR']$$

(7)

where $k_{obs}$ is equal to $k_2[OH^-]$.

During reaction (5), $OH^-$ is consumed and the progress of the reaction may be followed by the pH-stat kinetic method, which monitors the rate of hydroxide ion consumption at constant pH. From the plots of $\ln(V_o - V_t)$ versus time, where $V_o$ is the total volume of alkali consumed and $V_t$ is the volume consumed at time $t$, values of $k_{obs}$ may be obtained and a series of independent values of $k_2$, the second order rate constant may then be calculated.

(b) Hydrolysis of diimides in the presence of Cu(II)

This reaction depends on the metal ion as catalyst (as described in Chapter 5). The hydrolysis of the diimide in the presence of copper(II) was studied at constant pH in the range 4 to 5. The reaction would normally be expected to follow second order kinetics at constant pH.

$$Rate = k_2[Complex][OH^-]$$

(8)

If $[OH^-]$ is constant then the reaction becomes pseudo first order,

$$Rate = k_{obs}[Complex]$$

(9)
where \( k_{ob} \) is the pseudo first order rate constant and is equal to \( k_2[OH^-] \). The data were analyzed by two methods and observed rate constants calculated. These methods are,

1. **The \( V_o \) method;** \( \ln(V_o - V_t) \) versus \( t \) was plotted (see Figure 4 in Chapter 5), the slope of the plot giving \( k_{ob} \). (\( V_o = \)final volume of titre and \( V_t = \) volume of titre at time \( t \)).

2. **Guggenheim method;** \( \ln [V_t + 2t(\frac{1}{2}) - V_t] \) versus \( t \) was plotted, the slope of the plot again giving \( k_{ob} \). Generally method (1) was used when the final volume was known otherwise method (2) was employed.

\[ (t[\frac{1}{2}] = \text{half life of reaction}) \]
References


2. Radiometer Copenhagen, Autotitration Users Handbook.


CHAPTER 3
Metal Complexes of
2-(2’-Acetoxyethylamino) Ethylamine
Dihydrochloride or Dihydrobromide
1. Introduction

Metal complexes of ligands containing ester groups (e.g., amino acid and peptide esters) have recently been extensively studied. Much of this work has dealt with metal-promoted hydrolysis reactions directly or indirectly relevant to those operating at the active sites of metalloenzymes. Enhanced hydrolysis rates may result from (a) polarisation through complexing of the acyl group as for the complex in structure (1).

\[
\left[ \begin{array}{c}
\text{NH}_3 \\
\text{O} \\
\text{NH}_2
\end{array} \right]^3+ \\
\text{CO} \rightleftharpoons \text{O} \rightleftharpoons \text{CH} \\
\text{NH}_2
\]

(1)

(b) Juxtaposition of the acyl group and hydroxide ion in the coordination sphere of the metal ion as in the complex having structure (2).

\[
\left[ \begin{array}{c}
\text{NH}_2 \rightleftharpoons \text{CH} \rightleftharpoons \text{O} \\
\text{NH}_2 \rightleftharpoons \text{CH} \rightleftharpoons \text{NH}_2
\end{array} \right]^2+
\]

(2)

(c) Generation of better leaving groups through complex formation as in structure (3).

In order to investigate the last of these factors, it was decided to synthesise the...
title ligand (4) and during the course of the work a number of its complexes were isolated. In this chapter the synthesis and characterization of the new ligand and some of its metal complexes are reported.

\[
\begin{align*}
\text{(3)}
\end{align*}
\]

2. Experimental

(a) Preparation of 2-(2'-Acetoxyethylamino)ethylamine Dihydrochloride, \( \text{NH}_2(\text{CH}_2)_2\text{NH}(\text{CH}_2)_2\text{OCOCH}_3\cdot2\text{HCl}, (L.2\text{HCl}) \)

2-(2'-Aminoethylamino) ethanol, \( \text{NH}_2(\text{CH}_2)_2\text{NH}(\text{CH}_2)_2\text{OH} \) (From Aldrich and used without further purification) (10.0 g, 0.1 mole) was dissolved in glacial acetic acid (100 cm\(^3\)) which had previously been saturated with dry HCl gas. Further HCl gas was passed through the solution over a 10 minute period during
which time precipitation of the amine dihydrochloride occured. The resulting suspension was treated with acetyl chloride (25cm³, 0.35 mole) and refluxed until the solid had completely dissolved. On cooling a white crystalline material was obtained. This was washed with ethanol, then ether, and air dried. The product was recrystallised from ethanol; yield 9.5g(44.0%). m.p 171-173C. Mass spectrum m/e=147 (molecular ion having last 2HCl); "H n.m.r in D₂O 2.25δ (singlet,3-CH₃), 3.35 δ (broad,6,-CH₂ adjacent to protonated nitrogen atoms), 4.45 δ (triplet,2,-CH₂O). The i.r spectrum (Figure 2) of the product showed a ν(NH₃,NH₂) at 2200-3300cm⁻¹; ν(NH₂) at 2010cm⁻¹; and ν(C=O) at 1740cm⁻¹ which is typical of an aliphatic ester. Microanalysis; found C, 32.76; H, 7.33; N, 12.78%. C₆H₁₆N₂O₂Cl₂; requires C, 32.87; H, 7.30; N, 12.78%

(b) Preparation of 2-(2'-Acetoxyethylamino)ethylamine Dihydrobromide,

NH₂(CH₂)₃NH(CH₂)₂OCOCH₃.2HBr (L₂HBr)

The ligand dihydrobromide L₂HBr was prepared by dissolving L₂HCl(2.0g, 9x10⁻⁵mole) in 48% aqueous hydrogen bromide solution (5cm³). Addition of ethanol(150cm³) resulted in precipitation of L₂HBr. The product was collected by filtration, washed with ethanol, then ether and dried in a vacuum oven at 40°C. Yield 1.6g (46%) m.p 150-152°C. The i.r spectrum of the product showed ν(NH₃,NH₂) at 2200-3300cm⁻¹, ν(NH₂) at 2010cm⁻¹ and ν(C=O) at 1740cm⁻¹ which is typical of an aliphatic ester. Microanalysis; found C, 32.76; H, 7.33; N, 12.78%.
FIG. 2. I.R. spectrum of 2-(2-Acetoxyethylamino)ethylamine Dihydrochloride.
(c) Preparation of Metal Complexes of 2-(2'-Acetoxyethylamino)ethylamine Dihydrochloride

(i) \((\text{LH.HCl})_2 [\text{CuCl}_4]\). To a refluxing solution of \(\text{L.2HCl} (1.0\text{g}\ 4.5\times10^{-3}\text{mole})\) in ethanol \((100\text{cm}^3)\) was added dropwise with stirring to a solution of \(\text{CuCl}_2.2\text{H}_2\text{O} (1.5\text{g},9\times10^{-3}\text{mole})\) in ethanol \((25\text{cm}^3)\). During the addition the colour of the solution first became yellow, then green and a yellow precipitate was deposited. The mixture was stirred for 30 minutes at room temperature, filtered and the precipitate washed with ethanol and ether. The product was dried in a vacuum oven at 40°C. Yield 1.1g\((42\%)\).

(ii) \(\text{LH}_2[\text{CuBr}_4]\). This bronze coloured complex was obtained by the method used for the complex in (i) except that \(\text{CuBr}_2 (2.0\text{g},9\times10^{-3}\text{mole})\) was used instead of \(\text{CuCl}_2.2\text{H}_2\text{O}\). Yield 1.4g\((46\%)\).

(iii) \(\text{CuLCl}_2\). A stirred suspension of \(\text{L.2HCl} (1.0\text{g},4.5\times10^{-3}\text{mole})\) in absolute ethanol \((30\text{cm}^3)\) was treated dropwise with neat triethylamine \((1.25\text{cm}^3,9\times10^{-3}\text{mole})\). The resulting solution was added dropwise with the stirring to a solution of \(\text{Cu(NO}_3)_2.3\text{H}_2\text{O} (2.17\text{g}, 9\times10^{-3}\text{mole})\) in ethanol \((25\text{cm}^3)\) at room temperature. After a few minutes a blue precipitate was obtained. This was filtered, washed with ethanol, then ether and finally air dried. Yield 0.54g; \((43\%)\).

(iv) \(\text{CuLClBr.H}_2\text{O}\). This was prepared by the method used for complex (iii) except that \(\text{CuBr}_2 (2.0\text{g},9\times10^{-3}\text{mole})\) was used instead of \(\text{Cu(NO}_3)_2.3\text{H}_2\text{O}\). The green precipitate was collected by suction filtration, washed with ethanol, then ether and dried in a vacuum oven at 40°C. Yield 0.6g \((37\%)\).

(v) \(\text{NiLCl}_2.1/2\text{CH}_3\text{OH}\). This green complex was prepared by the method used for
complex(iii) except that NiCl$_2$ (1.2g,9×10$^{-3}$ mole) in methanol (20cm$^3$) was used instead of Cu(NO$_3$)$_2$.3H$_2$O. Yield 0.4g (31%).

(d) Preparation of Metal Complexes of 2-(2'-Acetoxyethylamino) ethylamine Dihydrobromide

(i) CuLBr$_2$: A stirred suspension of L.2HBr (1.0g,3×10$^{-3}$ mole) in absolute ethanol (20cm$^3$) was treated dropwise with neat triethylamine (1.3cm$^3$,9×10$^{-3}$ mole). The resulting solution was added dropwise with stirring to a solution of CuBr$_2$ (2.0g, 9×10$^{-3}$ mol) in ethanol (20cm$^3$) at room temperature. The green precipitate which formed was collected by suction filtration washed with ethanol and dried in a vacuum oven at 40°C. Yield 0.48g (40%).

(ii) NiLBrg. 1/4C$_2$H$_5$OH: This green complex was prepared by the method used for complex d(i) except that NiBr$_2$.3H$_2$O (1.31g,4×10$^{-3}$ mole) was used instead of CuBr$_2$. Yield 0.4g (33%).

Microanalytical, spectroscopic and magnetic moment data for all the complexes are given in the Tables 1-5.
Table 1. Analytical Data for Metal Complexes of 2-(2'-Acetoxyethylamino) ethylamine Dihydrochloride or Dihydrobromide

\[ \text{C}_6\text{H}_{14}\text{N}_2\text{O}_2\text{Cl}_2 \text{ or C}_6\text{H}_{14}\text{N}_2\text{O}_2\text{Br}_2 \]

<table>
<thead>
<tr>
<th>Complex</th>
<th>Calc. %</th>
<th>Found %</th>
</tr>
</thead>
<tbody>
<tr>
<td>((\text{LH.HCl})_2 \cdot \text{CuCl}_4)</td>
<td>C 25.15  H 5.59  N 9.78</td>
<td>C 25.18  H 5.55  N 9.56</td>
</tr>
<tr>
<td>(\text{LH}_2 \cdot \text{CuBr}_4)</td>
<td>C 21.26  H 4.43  N 8.27</td>
<td>C 20.66  H 4.57  N 7.87</td>
</tr>
<tr>
<td>(\text{CuLCl}_2)</td>
<td>C 25.66  H 4.99  N 9.98</td>
<td>C 25.96  H 4.97  N 9.94</td>
</tr>
<tr>
<td>(\text{CuLClBr}\cdot\text{H}_2\text{O})</td>
<td>C 21.00  H 4.66  N 8.16</td>
<td>C 21.39  H 4.13  N 8.19</td>
</tr>
<tr>
<td>(\text{NiLCl}_2\cdot\frac{1}{2} \text{CH}_3\text{OH})</td>
<td>C 26.74  H 5.48  N 9.60</td>
<td>C 26.54  H 5.60  N 9.99</td>
</tr>
<tr>
<td>(\text{CuLBr}_2)</td>
<td>C 19.50  H 3.79  N 7.58</td>
<td>C 20.08  H 3.71  N 7.58</td>
</tr>
<tr>
<td>(\text{NiLBr}_2\cdot\frac{1}{4} \text{C}_2\text{H}_5\text{OH})</td>
<td>C 20.74  H 4.12  N 7.45</td>
<td>C 20.73  H 4.57  N 7.93</td>
</tr>
</tbody>
</table>
Table 2. Relative Molecular Mass and the Percentage of Metal in Complexes of 2-(2'-Acetoxyethylamino)ethylamine Dihydrochloride or Dihydrobromide

<table>
<thead>
<tr>
<th>Complex</th>
<th>Relative Molecular mass calc</th>
<th>Calculated % metal</th>
<th>Relative Molecular mass found</th>
<th>% Metal found</th>
</tr>
</thead>
<tbody>
<tr>
<td>(LH.HCl)₂ [CuCl₄]</td>
<td>572.5</td>
<td>11.09</td>
<td>576.35</td>
<td>11.02</td>
</tr>
<tr>
<td>(C₁₂H₂₂N₄O₄Cl₅Cu)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>LH₂ [CuBr₄]</td>
<td>677.1</td>
<td>9.38</td>
<td>683.3</td>
<td>9.29</td>
</tr>
<tr>
<td>(C₁₂H₂₀O₄N₄CuBr₄)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CuLCl₂</td>
<td>280.5</td>
<td>22.64</td>
<td>281.76</td>
<td>22.54</td>
</tr>
<tr>
<td>(C₆H₁₄N₂O₂Cl₂Cu)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CuLClBr.H₂O</td>
<td>342.9</td>
<td>18.52</td>
<td>345.68</td>
<td>18.14</td>
</tr>
<tr>
<td>(C₆H₁₆N₂O₃CuClBr)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NiLCl₂.1/2 CH₃OH</td>
<td>291.7</td>
<td>20.13</td>
<td>290.52</td>
<td>20.54</td>
</tr>
<tr>
<td>(C₆H₁₄N₂O₂Cl₂Ni)1/2 MeOH</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CuLBr₂</td>
<td>369.3</td>
<td>17.19</td>
<td>370.5</td>
<td>17.29</td>
</tr>
<tr>
<td>(C₆H₁₄N₂O₂Br₂Cu)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NiBr₂.1/4 C₂H₅OH</td>
<td>376.01</td>
<td>15.61</td>
<td>380.00</td>
<td>15.45</td>
</tr>
<tr>
<td>(C₆H₁₄N₂O₂NiBr₂)1/4 EtOH</td>
<td></td>
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</table>
Table 3. Salient I.R. Spectral Bands of Metal Complexes of
2(2'Acetoxyethylamine)ethylamine

Dihydrochloride or Dihydrobromide (cm$^{-1}$)

<table>
<thead>
<tr>
<th>Compound</th>
<th>$\nu$(C=O) ester/cm$^{-1}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>$L\ 2HCl$</td>
<td>1740$^*$</td>
</tr>
<tr>
<td>$(LH.HCl)_2\ [CuCl_4]$</td>
<td>1740, 1755$^m$</td>
</tr>
<tr>
<td>$(LH_2)\ [CuBr_4]$</td>
<td>1735$^*$</td>
</tr>
<tr>
<td>$CuLCl_2$</td>
<td>1730$^m$, 1745$^h$</td>
</tr>
<tr>
<td>$CuLClBr_2H_2O$</td>
<td>1737$^m$</td>
</tr>
<tr>
<td>$NiLCl_2\ {\text{1/2 CH}}_3 OH$</td>
<td>1730$^&quot;$</td>
</tr>
<tr>
<td>$CuLBr_2$</td>
<td>1740$^m$</td>
</tr>
<tr>
<td>$NiLBr_2\ {1/4 C}_2\text{H}_6\text{OH}$</td>
<td>1730$^m$</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Compound</th>
<th>Amine stretching bands/cm$^{-1}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>$L\ 2HCl$</td>
<td>2200 - 3,300$^b, m$</td>
</tr>
<tr>
<td>$(LH.HCl)_2\ [CuCl_4]$</td>
<td>2415, 2480, 2570, 2610</td>
</tr>
<tr>
<td>$(LH_2)\ [CuBr_4]$</td>
<td>2400, 2700$^b, m$</td>
</tr>
<tr>
<td>$CuLCl_2$</td>
<td>3195, 3260, 3335$^m$</td>
</tr>
<tr>
<td>$CuLClBr_2H_2O$</td>
<td>3185, 3235, 3320$^m$</td>
</tr>
<tr>
<td></td>
<td>3500$^b$</td>
</tr>
<tr>
<td>$NiLCl_2\ {1/2 CH}_3 OH$</td>
<td>3165, 3245, 3295$^m$</td>
</tr>
<tr>
<td></td>
<td>3500$^b$</td>
</tr>
<tr>
<td>$CuLBr_2$</td>
<td>3185, 3235, 3330</td>
</tr>
<tr>
<td>$NiLBr_2\ {1/4 C}_2\text{H}_6\text{OH}$</td>
<td>3160, 3240, 3300</td>
</tr>
<tr>
<td></td>
<td>3500$^b$</td>
</tr>
</tbody>
</table>
Table 4. Room Temperature Magnetic Moments of Metal Complexes of 2-(2'-Acetoxyethylamino)ethylamine Dihydrochloride or Dihydrobromide

<table>
<thead>
<tr>
<th>Complex</th>
<th>$\chi_g$ ($cm^{-1}$T)</th>
<th>$\chi_m$ ($cm^{-1}$T)</th>
<th>$\chi_A$ ($cm^{-1}$T)</th>
<th>$\mu_e$ B.M.</th>
</tr>
</thead>
<tbody>
<tr>
<td>$(LH.HCl)_2 [CuCl_4]$</td>
<td>$3 \times 10^{-6}$</td>
<td>$1.746 \times 10^{-5}$</td>
<td>$1.424 \times 10^{-5}$</td>
<td>1.82</td>
</tr>
<tr>
<td>$LH_2 [CuBr_4]$</td>
<td>$2.671 \times 10^{-6}$</td>
<td>$1.856 \times 10^{-5}$</td>
<td>$1.525 \times 10^{-5}$</td>
<td>1.89</td>
</tr>
<tr>
<td>CuLCl$_2$</td>
<td>$6.049 \times 10^{-6}$</td>
<td>$1.697 \times 10^{-5}$</td>
<td>$1.562 \times 10^{-5}$</td>
<td>1.92</td>
</tr>
<tr>
<td>CuLClBr.$H_2$O</td>
<td>$3.884 \times 10^{-6}$</td>
<td>$1.434 \times 10^{-5}$</td>
<td>$1.278 \times 10^{-5}$</td>
<td>1.73</td>
</tr>
<tr>
<td>NiLCl$_2$.1/2 CH$_3$OH</td>
<td>$1.446 \times 10^{-6}$</td>
<td>$4.219 \times 10^{-5}$</td>
<td>$4.073 \times 10^{-5}$</td>
<td>3.08</td>
</tr>
<tr>
<td>CuLBr$_2$</td>
<td>$4.233 \times 10^{-6}$</td>
<td>$1.563 \times 10^{-5}$</td>
<td>$1.371 \times 10^{-5}$</td>
<td>1.78</td>
</tr>
<tr>
<td>NiLBr$_2$.1/4 C$_2$H$_6$OH</td>
<td>$1.132 \times 10^{-6}$</td>
<td>$4.258 \times 10^{-5}$</td>
<td>$4.09 \times 10^{-5}$</td>
<td>3.09</td>
</tr>
</tbody>
</table>

Table 5. Reflectance Spectral Data for Metal Complexes of 2-(2'-Acetoxyethylamino)ethylamine Dihydrochloride or Dihydrobromide

<table>
<thead>
<tr>
<th>Complex</th>
<th>$\lambda_{max}$/nm</th>
</tr>
</thead>
<tbody>
<tr>
<td>$(LH.HCl)_2 [CuCl_4]$</td>
<td>400 775</td>
</tr>
<tr>
<td>$LH_2 [CuBr_4]$</td>
<td>417 770</td>
</tr>
<tr>
<td>CuLCl$_2$</td>
<td>350 700</td>
</tr>
<tr>
<td>CuLClBr.$H_2$O</td>
<td>385 690</td>
</tr>
<tr>
<td>NiLCl$_2$.1/2 CH$_3$OH</td>
<td>375 580 700</td>
</tr>
<tr>
<td>CuLBr$_2$</td>
<td>400 695</td>
</tr>
<tr>
<td>NiLBr$_2$.1/4 C$_2$H$_6$OH</td>
<td>390 675</td>
</tr>
</tbody>
</table>
3. Results and Discussion

The addition of acetyl chloride to a solution of 2-(2'-aminoethylamino) ethanol in HCl/acetic acid results in acetylation of the hydroxy group and gives after refluxing L.2HCl. (4)

\[
\left[ \text{NH}_3 \text{CH}_2 \text{CH}_2^+ \text{NH}_2 \text{CH}_2 \text{CH}_2 \text{OCOC}_2 \right] 2\text{Cl}
\]  

(4)

This was characterised by C, H, N, microanalysis, (Table 1) and by mass, \(^1\)H n.m.r (Figure 1) and i.r spectroscopy (Figure 2, Table 2). The mass spectrum shows m/e at 147 (base peak) corresponding to the molecular ion having lost two molecules of HCl. The \(^1\)H n.m.r spectrum in D\(_2\)O solution displayed signals at 2.25 (singlet), 2.53 (broad) and 4.44 (triplet) which integrated in the ratio 3:6:2. The first of these may be assigned to the CH\(_3\) group, the second to a superimposition of signals due to the CH\(_2\) groups adjacent to the protonated nitrogen atoms (6H) and the most downfield signal to the CH\(_2\) group directly attached to the CH\(_2\)CO\(_2\) group. The i.r. spectrum of L.2HCl is ill defined in the 2,200-3,300 cm\(^{-1}\) region due to the presence of intense broad overlapping \(\nu_{\text{sym}}\) and \(\nu_{\text{asym}}\) NH\(_3\) and NH\(_2\) bands. The medium band at \(\sim\) 2000 cm\(^{-1}\) is characteristic of the N-H stretching band of secondary amines. The \(\nu(C=O)\) absorption is found at 1740cm\(^{-1}\), which is typical of aliphatic esters (Figure 2, Table 2).

The products obtained as a result of adding CuX\(_2\) or NiX\(_2\) (X=Cl, Br) to L.2HCl or L.2HBr in ethanol or methanol depend on whether or not the noncomplexing base, triethylamine is added to the reaction mixture. The addition of CuX\(_2\) (X=Cl, Br) to a solution of L.2HCl in ethanol gave salts containing the complex
anions \([\text{Cu Cl}_4]^{2-}\) and \([\text{Cu Br}_4]^{2-}\). The presence of the conjugate acid of L as a cation in these salts is confirmed by the i.r spectra which show symmetric and asymmetric N-H stretching bands due to the protonated primary and secondary amino groups in the region 2000-3300 cm\(^{-1}\). Complex salts containing the tetrahalocuprate (II) anion have been widely investigated\(^{11}\) and the geometry of the anion varies between tetrahedral and square planar (most are flattened tetrahedral). This depends on a range of factors such as crystal packing forces, hydrogen bonding to cation, ligand-ligand repulsion and many other factors.

The addition of ligand to metal ion solutions in the presence of the non-complexing base triethylamine produces complexes in which the ligand is coordinated to the metal ion. Evidence from infrared spectroscopy suggests that the ester groups are either uncoordinated or weakly coordinated to the metal ion in these complexes since the positions of CO stretching bands are in similar positions to those in the free ligands. On this evidence the ligands should be bidentate and the structures of the complexes may be written as in structure (5). The relative molecular mass of these complexes (Table 5) were determined by EDTA titration\(^{12}\) and the percentage of metal in the complexes was in close agreement with the calculated value. The i.r spectra of all the complexes are very similar. All have \(\nu(C=O)\) bands at 1730-1740 cm\(^{-1}\) and N-H stretching bands at 2200-3400 cm\(^{-1}\). In the hydrated complexes OH stretching bands occur at 3500 cm\(^{-1}\) (broad). The reflectance spectra of the copper(II) complexes all have two bands in the region 350-850 nm (Table 4). The copper(II) halide complexes are blue, or green.\(^{13}\) The blue or green colours are due to the presence of an absorption band in the region 600-900 nm of the spectrum.
The room temperature magnetic moments of the copper(II) complexes (1.73-1.92 B.M) are close to the spin only value (1.73 B.M) and consistent with square planar, square pyramidal or octahedral geometries.

The reflectance spectra of the nickel(II) complexes have three bands in the region 350-850nm. The room temperature magnetic moments of the nickel(II) complexes (3.09-3.08) are close to the spin only value (2.87 B.M) for octahedral nickel(II).
References


Chapter 4
Metal Complexes of EDTA Derivatives.
Preparation of the Tetramethyl amide of EDTA and its Metal Complexes.
1. Introduction

Despite the wide use of EDTA as an analytical reagent for metal ions, very little attention has been given to its derivatives such as esters, amides and imides and their interaction with metal ions. A few examples of such derivatives, e.g., the tetramethyl\(^1,2\) and tetraethyl\(^3\) esters and the tetramide\(^4\) have been reported. The Cu(II) promoted hydrolysis of some of these ligands has been studied.\(^1\)

In this chapter the syntheses, characterisation and properties of the new compound EDTA (NHMe\(^4\)) and its metal complexes are described.

2. Experimental

(a) Preparation of the Tetramethyl Ester of EDTA

\[
\begin{align*}
\text{HC} & \quad \text{CH} \\
\text{2} & \quad 2 \\
(\text{HCOCHC})_2 & \quad N\left(\text{CHCOCH}\right)_2, \quad \text{EDTA(O\text{Me}})_4 \\
\end{align*}
\]

This compound was prepared by a previously reported method\(^1-3\).

A suspension of ethylenediaminetetraacetic acid, H\(_4\)EDTA (30.0 g, 0.102 mol) in dry methanol (140 cm\(^3\), 0.2 mol) was cooled in an ice bath and thionyl chloride (5 cm\(^3\), 0.2 mol) added dropwise with stirring over two hours, keeping the temperature below 10\(^\circ\)C. The suspension was refluxed until all the solid had completely
dissolved. The solution was then basified with a saturated sodium bicarbonate solution and the desired ester was extracted with ether. The ether phase was dried over anhydrous sodium sulphate. The solvent was removed to leave a light yellow, oily residue. Yield 29.1g (81.9%). The i.r spectrum of the product (liquid film) showed a sharp absorption band at 1740 cm\(^{-1}\), which is typical of \(\nu(C=O)\) for an aliphatic ester\(^5\).

(b) Preparation of the Tetramide of EDTA, EDTA \((NH_2)\)_4

This compound was prepared by a previously reported method.\(^4\)

(c) Preparation of the tetramethyl amide of EDTA, EDTA \((NHMe)\)_4

Tetramethylethylenediaminetetra acetate, EDTA(OMe)_4 (6.0 g, 0.0172 mol) prepared as previously described and dry methanol (50 cm\(^3\)) were stirred at room temperature. To this methyl-amine (30% in ethanol, 37 cm\(^3\)) was added dropwise. The resulting solution, which turned slightly yellow, was stirred for an hour at room temperature, then refluxed for three hours, cooled and the solvent removed on the rotary evaporator, leaving a solid residue. Acetone was added and the suspension stirred for 1 hour at room temperature. The product was collected by filtration, recrystallized from acetone and dried in a vacuum oven at 40°C for three hours. Yield 5.68g (96%), m.p. 184-86°C. The i.r spectrum (Table 3) of the product (Nujol mull) showed absorptions bands at 3340 cm\(^{-1}\) (sharp) (NH stretching), 1640 cm\(^{-1}\) (amide I band) and 1530 cm\(^{-1}\) (amide II band)\(^5\).

Microanalysis: found C, 48.25; H, 8.44, N, 24.11%. \(C_{14}H_{28}N_6O_4\), requires C, 48.83; H, 8.14; N, 24.42%. \(^1\)H n.m.r (Table 1) spectrum in D\(_2\)O showed signals at \(\delta 2.63\) (singlet, 4H, CH\(_2\)), 2.74\(^6\) (singlet, 12H, CH\(_3\)) and 2.23\(^7\) (singlet, 8H, CH\(_2\)).
### Table 1. NMR spectral Data of the tetramethylamide of EDTA, $(C_{14}H_{28}N_8O_4)$

<table>
<thead>
<tr>
<th>Chemical Shift ppm</th>
<th>Multiplicity</th>
<th>Relative Integration</th>
<th>Assignment</th>
</tr>
</thead>
<tbody>
<tr>
<td>2.23</td>
<td>singlet</td>
<td>8H</td>
<td>a</td>
</tr>
<tr>
<td>2.63</td>
<td>singlet</td>
<td>4H</td>
<td>b</td>
</tr>
<tr>
<td>2.74</td>
<td>singlet</td>
<td>12H</td>
<td>c</td>
</tr>
</tbody>
</table>

(d) Preparation of Metal Complexes of EDTA (NHMe)$_4$

A stirred solution of the metal salt ($2 \times 10^{-3}$ mol) in dry methanol (10 cm$^3$) was added dropwise to a stirred solution of EDTA (NHMe)$_4$ ($1.45 \times 10^{-3}$ mol) also in dry methanol (10 cm$^3$) at room temperature. In many cases the desired complexes crystallized from solution on standing, otherwise crystallization was induced by the addition of ether. Yields 50-90%. Elemental, spectral (i.r and reflectance), magnetic and EDTA titration data for these complexes are presented in...
Table 2. Elemental analysis for metal complexes of the Tetramethylamide of EDTA, \( (C_{14}H_{28}N_6O_4) \)

<table>
<thead>
<tr>
<th>Complex</th>
<th>Calculated %</th>
<th>Found %</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>C</td>
<td>H</td>
</tr>
<tr>
<td>( [CuL(H_2O)]_2[CuCl_4]H_2O )</td>
<td>25.18</td>
<td>5.10</td>
</tr>
<tr>
<td>( [Cu_2L_2Br_2][CuBr_4]2CH_3OH )</td>
<td>25.31</td>
<td>4.50</td>
</tr>
<tr>
<td>( CuL(NO_3)_2 )</td>
<td>31.60</td>
<td>5.26</td>
</tr>
<tr>
<td>( [CoL][CoCl_4] )</td>
<td>27.82</td>
<td>4.64</td>
</tr>
<tr>
<td>( [CoL(H_2O)]_2Br_2 )</td>
<td>28.06</td>
<td>5.34</td>
</tr>
<tr>
<td>( CoL(NO_3)_2 )</td>
<td>31.88</td>
<td>5.31</td>
</tr>
<tr>
<td>( NiL(NO_3)_2\cdot1/2H_2O )</td>
<td>31.36</td>
<td>5.41</td>
</tr>
<tr>
<td>( NiLCl_2.5H_2O )</td>
<td>29.80</td>
<td>6.74</td>
</tr>
<tr>
<td>( NiLB_2\cdot3H_2O )</td>
<td>27.25</td>
<td>5.51</td>
</tr>
<tr>
<td>( NiL_2 )</td>
<td>25.59</td>
<td>4.26</td>
</tr>
<tr>
<td>( [CdL][CdI_4]\cdot(CdI_2)_2L )</td>
<td>15.60</td>
<td>2.60</td>
</tr>
<tr>
<td>( FeLCl_2.2H_2O )</td>
<td>30.97</td>
<td>5.90</td>
</tr>
<tr>
<td>( FeL(NO_3)_2.3H_2O )</td>
<td>26.25</td>
<td>5.31</td>
</tr>
<tr>
<td>( FeLSO_4.6H_2O )</td>
<td>27.82</td>
<td>6.62</td>
</tr>
<tr>
<td>( ZnLSO_4.2H_2O )</td>
<td>23.90</td>
<td>4.55</td>
</tr>
</tbody>
</table>
Table 3. Spectra for metal complexes of the Tetramethylamide of EDTA, \((C_{14}H_{28}N_6O_4)\).

<table>
<thead>
<tr>
<th>Compound</th>
<th>Amide I (\nu(C=O)), Amide II, (\delta(NH)) bands/cm(^1)</th>
<th>(\nu(NH)), (\nu(OH)) cm(^1)</th>
</tr>
</thead>
<tbody>
<tr>
<td>EDTA((NHMe))(_4)</td>
<td>1640 1530</td>
<td>3340</td>
</tr>
<tr>
<td>([CuL(H_2O)]_2[CuCl_4]H_2O)</td>
<td>1620 1570</td>
<td>3100,3270 3480,3500</td>
</tr>
<tr>
<td>([Cu_2L_2Br_2][CuBr_4]2CH_3OH)</td>
<td>1630 1560</td>
<td>3100,3270 3500</td>
</tr>
<tr>
<td>CuL((NO_3))_2</td>
<td>1640 1570</td>
<td>3100,3260</td>
</tr>
<tr>
<td>[CoL][CoCl_4]</td>
<td>1640 1580</td>
<td>3100,3270 3500</td>
</tr>
<tr>
<td>[CoL(H_2O)]_2Br_2</td>
<td>1640 1570</td>
<td>3100,3240 3500</td>
</tr>
<tr>
<td>CoL((NO_3))_2</td>
<td>1640 1590</td>
<td>3100,3250</td>
</tr>
<tr>
<td>NiL((NO_3))_2(1/2H_2O)</td>
<td>1635 1580</td>
<td>3095,3230 3500</td>
</tr>
<tr>
<td>NiLCl_5H_2O</td>
<td>1635 1580</td>
<td>3100,3240 3500</td>
</tr>
<tr>
<td>NiLB_3H_2O</td>
<td>1635 1580</td>
<td>3095,3230 3500</td>
</tr>
<tr>
<td>NiL</td>
<td>1630 1580</td>
<td>3100,3240</td>
</tr>
<tr>
<td>[CdL]([CdI_2]_2L)</td>
<td>1640 1565</td>
<td>3110, 3300,3360,3470</td>
</tr>
<tr>
<td>FeLCl_32H_2O</td>
<td>1650 1570</td>
<td>3240 3440,3540</td>
</tr>
<tr>
<td>FeL((NO_3))_3H_2O</td>
<td>1650 1570</td>
<td>3100,3270 3500</td>
</tr>
<tr>
<td>FeLSO_4(6H_2O)</td>
<td>1640 1580</td>
<td>3100,3260 3500</td>
</tr>
<tr>
<td>ZnLSO_4(2H_2O)</td>
<td>1640 1570</td>
<td>3100,3240,3500</td>
</tr>
</tbody>
</table>

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Table 4. Reflectance spectra for metal complexes of the
Tetramethylamid of EDTA

<table>
<thead>
<tr>
<th>Complex</th>
<th>$\lambda_m/nm$</th>
</tr>
</thead>
<tbody>
<tr>
<td>$[CuL(H_2O)_2][CuCl_4H_2O]$</td>
<td>375</td>
</tr>
<tr>
<td>$[Cu_2L_2Br_2][CuBr_4]2CH_3OH$</td>
<td>375 525 610sh</td>
</tr>
<tr>
<td>$CuL(NO_3)_2$</td>
<td>765</td>
</tr>
<tr>
<td>$[CoL][CoCl_4]$</td>
<td>530,630,665,695</td>
</tr>
<tr>
<td>$[CoL(H_2O)_2]Br_2$</td>
<td>525,645,665,700</td>
</tr>
<tr>
<td>$CoL(NO_3)_2$</td>
<td>465 625</td>
</tr>
<tr>
<td>$NiL(NO_3)_2 \cdot 1/2 H_2O$</td>
<td>400 605</td>
</tr>
<tr>
<td>$NiLCl_25H_2O$</td>
<td>390 650</td>
</tr>
<tr>
<td>$NiLBr_23H_2O$</td>
<td>395 640</td>
</tr>
<tr>
<td>$NiLI_2$</td>
<td>385 605</td>
</tr>
<tr>
<td>$FeLCl_22H_2O$</td>
<td>410 550</td>
</tr>
<tr>
<td>$FeL(NO_3)_93H_2O$</td>
<td>400 485</td>
</tr>
<tr>
<td>$FeLSO_46H_2O$</td>
<td>400</td>
</tr>
</tbody>
</table>
Table 5. Room magnetic moments of metal complexes
of the tetramethylamide of EDTA

<table>
<thead>
<tr>
<th>Complex</th>
<th>$X_a$</th>
<th>$X_m$</th>
<th>$XA$</th>
<th>$\mu_{c,BM}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>$[CuL(H_2O)]_2[CuCl_4]H_2O$</td>
<td>3.71310^6</td>
<td>2.47710^3</td>
<td>2.14510^3</td>
<td>2.24</td>
</tr>
<tr>
<td>$[Cu_2L_2Br_2]_2[BrCl_4]_2CH_3OH$</td>
<td>2.14710^6</td>
<td>3.05210^3</td>
<td>2.37510^3</td>
<td>2.35</td>
</tr>
<tr>
<td>CuL(NO$_3$)$_2$</td>
<td>3.08210^6</td>
<td>1.63810^3</td>
<td>1.38710^3</td>
<td>1.80</td>
</tr>
<tr>
<td>[CoL][CoCl$_4$]</td>
<td>1.37210^6</td>
<td>8.28610^3</td>
<td>7.99210^3</td>
<td>4.32</td>
</tr>
<tr>
<td>CoL(H$_2$O)$_2$Br$_2$</td>
<td>1.34910^5</td>
<td>8.07810^3</td>
<td>7.77810^3</td>
<td>4.30</td>
</tr>
<tr>
<td>CoL(NO$_3$)$_2$</td>
<td>1.60210^5</td>
<td>8.44110^3</td>
<td>8.19010^3</td>
<td>4.37</td>
</tr>
<tr>
<td>NiL(NO$_3$)$_2$·1/2 H$_2$O</td>
<td>8.96410^6</td>
<td>4.77210^3</td>
<td>4.50910^3</td>
<td>3.24</td>
</tr>
<tr>
<td>NiLCl$_5$H$_2$O</td>
<td>8.72110^6</td>
<td>4.91610^3</td>
<td>4.58810^3</td>
<td>3.27</td>
</tr>
<tr>
<td>NiLBr$_5$H$_2$O</td>
<td>8.85710^6</td>
<td>4.84410^3</td>
<td>4.53110^3</td>
<td>3.25</td>
</tr>
<tr>
<td>NiL$_2$</td>
<td>7.39710^6</td>
<td>4.85610^3</td>
<td>4.55410^3</td>
<td>3.26</td>
</tr>
<tr>
<td>FeLCl$_2$H$_2$O</td>
<td>2.35310^5</td>
<td>0.0127</td>
<td>0.0125</td>
<td>5.40</td>
</tr>
<tr>
<td>FeL(NO$_3$)$_3$H$_2$O</td>
<td>1.89710^5</td>
<td>0.0121</td>
<td>0.0118</td>
<td>5.26</td>
</tr>
<tr>
<td>FeL$_3$O$_4$H$_2$O</td>
<td>2.03610^5</td>
<td>0.0122</td>
<td>0.01196</td>
<td>5.28</td>
</tr>
</tbody>
</table>
Table 6. Relative molecular mass and the percentage of metal in the complexes of the tetramethylamide of EDTA

<table>
<thead>
<tr>
<th>Complex</th>
<th>Relative molecular mass Calc</th>
<th>Calculated % M</th>
<th>% M found</th>
<th>Relative molecular mass found</th>
</tr>
</thead>
<tbody>
<tr>
<td>[CuL(H$_2$O)$_2$][CuCl$_4$]H$_2$O</td>
<td>667.1</td>
<td>9.51</td>
<td>9.43</td>
<td>673</td>
</tr>
<tr>
<td>Cu$_2$L$_2$Br$_2$[CuBr$_4$]2C$_3$H$_5$OH</td>
<td>1421.9</td>
<td>4.46</td>
<td>4.47</td>
<td>1417.65</td>
</tr>
<tr>
<td>CuL(NO$_3$)$_2$</td>
<td>531.5</td>
<td>11.94</td>
<td>12.01</td>
<td>528.42</td>
</tr>
<tr>
<td>CuL(CuCl$_4$)</td>
<td>603.8</td>
<td>9.75</td>
<td>9.66</td>
<td>609.7</td>
</tr>
<tr>
<td>CuL(H$_2$O)$_2$Br$_2$</td>
<td>598.5</td>
<td>9.84</td>
<td>9.90</td>
<td>595.24</td>
</tr>
<tr>
<td>CuL(NO$_3$)$_2$</td>
<td>526.9</td>
<td>11.18</td>
<td>11.03</td>
<td>534.04</td>
</tr>
<tr>
<td>CuL(H$_2$O)$_2$Cl$_2$</td>
<td>535.7</td>
<td>10.95</td>
<td>10.96</td>
<td>535.03</td>
</tr>
<tr>
<td>CuL(NO$_3$)$_2$.1/2$H_2$O</td>
<td>563.69</td>
<td>10.41</td>
<td>10.42</td>
<td>563.16</td>
</tr>
<tr>
<td>NiL(NO$_3$)$_2$.1/2$H_2$O</td>
<td>616.49</td>
<td>9.52</td>
<td>9.47</td>
<td>619.41</td>
</tr>
<tr>
<td>NiLCl$_3$.H$_2$O</td>
<td>656.49</td>
<td>8.33</td>
<td>8.91</td>
<td>658.48</td>
</tr>
<tr>
<td>EiL[Cl$_2$]$_2$L</td>
<td>1076.42</td>
<td>10.44</td>
<td>10.41</td>
<td>1079.52</td>
</tr>
<tr>
<td>FeLCl$_3$.2H$_2$O</td>
<td>542.34</td>
<td>10.30</td>
<td>10.36</td>
<td>538.5</td>
</tr>
<tr>
<td>FeL(NO$_3$)$_2$.6H$_2$O</td>
<td>639.84</td>
<td>8.73</td>
<td>8.61</td>
<td>648.5</td>
</tr>
<tr>
<td>FeL$O_4$.6H$_2$O</td>
<td>603.84</td>
<td>9.25</td>
<td>8.92</td>
<td>625.8</td>
</tr>
<tr>
<td>ZnLSO$_4$.2H$_2$O</td>
<td>541.38</td>
<td>12.07</td>
<td>11.83</td>
<td>552.5</td>
</tr>
</tbody>
</table>
Tables 2-6. The complexes have the general formula $M(C_{14}H_{28}N_6O_4)X_n$. solvent

($M = Cu^{II}, Co^{II}, Ni^{II}, Cd^{II}, Fe^{II}, Fe^{III}: X = Cl, Br, I, NO_3, SO_4$)

(e) Potentiometric Titration of the Cu^{II}-EDTA $(NHMe)_4$ Complex

The ionization constant of the ligand when complexed with copper was obtained by potentiometric titration. This operation was carried out using a Radiometer pH meter 26 fitted with an automatic titrator 11 and an autoburette unit ABU12 to deliver the titrant. A high alkalinity glass electrode was used in conjunction with a saturated calomel electrode as reference. The electrode system was standardized at 25°C with 0.05M potassium hydrogen phthalate (pH = 4.01) and 0.01M disodium tetra borate (borax pH = 9.81) buffers. The reaction solution was contained in a 150 cm$^3$ vessel, fitted with a cover which was designed to accommodate the electrode system, nitrogen inlet and outlet tubes, a titrant inlet tube and stirrer. The whole apparatus was thermostatted at 25°C.

The tetramethylamide of EDTA, EDTA$(NHMe)_4$ (0.1738, $5 \times 10^{-4}$ mol) was added to a solution of copper nitrate (25 cm$^3$, $2 \times 10^{-5}$ mol) and placed in the vessel and stirred. At the beginning of the reaction, the colour of the solution was light green. The starting pH was noted and its change was monitored as a 0.05M solution of NaOH was added. The resulting potentiometric titration curve is shown in Fig 2 and the data presented in Table 7. The pK$_a$ values were obtained using a MINIQUAD computer program. The equilibria corresponding to the pK$_a$ values obtained are represented in equations (1) and (2). The site of coordination changes from amide oxygen to nitrogen as a result of ionization.
THE TITRATION CURVE OF THE TETRAMETHYLAMINE OF EDTA AND COPPER NITRATE WITH 0.05M NaOH AT 25°C.

Fig 2. VOLUME OF 0.05M NaOH ADDED (ml)
Table 7. Potentiometric titration data. the Cu^{II} - EDTA(NHMe)_4

(210^3M, 25.0cm^3) complex vs. 0.05 M NaOH at 25 °C

<table>
<thead>
<tr>
<th>Volume of 0.05 M NaOH added</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0 cm^3</td>
<td>4.49</td>
</tr>
<tr>
<td>0.1 cm^3</td>
<td>5.73</td>
</tr>
<tr>
<td>0.2 cm^3</td>
<td>6.09</td>
</tr>
<tr>
<td>0.3 cm^3</td>
<td>6.39</td>
</tr>
<tr>
<td>0.4 cm^3</td>
<td>6.53</td>
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<tr>
<td>0.5 cm^3</td>
<td>6.70</td>
</tr>
<tr>
<td>0.603 cm^3</td>
<td>6.88</td>
</tr>
<tr>
<td>0.7 cm^3</td>
<td>7.04</td>
</tr>
<tr>
<td>0.803 cm^3</td>
<td>7.23</td>
</tr>
<tr>
<td>0.9 cm^3</td>
<td>7.43</td>
</tr>
<tr>
<td>1.001 cm^3</td>
<td>7.66</td>
</tr>
<tr>
<td>1.1 cm^3</td>
<td>7.90</td>
</tr>
<tr>
<td>1.201 cm^3</td>
<td>8.13</td>
</tr>
<tr>
<td>1.3 cm^3</td>
<td>8.32</td>
</tr>
<tr>
<td>1.4 cm^3</td>
<td>8.50</td>
</tr>
<tr>
<td>1.5 cm^3</td>
<td>8.67</td>
</tr>
<tr>
<td>1.6 cm^3</td>
<td>8.84</td>
</tr>
<tr>
<td>1.7 cm^3</td>
<td>9.01</td>
</tr>
<tr>
<td>1.8 cm^3</td>
<td>9.21</td>
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<tr>
<td>1.9 cm^3</td>
<td>9.44</td>
</tr>
<tr>
<td>2.002 cm^3</td>
<td>9.78</td>
</tr>
<tr>
<td>2.1 cm^3</td>
<td>10.22</td>
</tr>
<tr>
<td>2.51 cm^3</td>
<td>11.19</td>
</tr>
</tbody>
</table>

pKa values obtained using a MINIQUAD computer program.

(i) 8.64 st. deviation = 0.00423

(ii) 6.71 st. deviation = 0.00094
\[ \begin{align*}
\text{(a)} & \quad \text{MeHNOCH}_2 \text{N}_2 \text{CHCONHMe}^2+ \\
\text{(b)} & \quad \text{MeHNOCH}_2 \text{N}_2 \text{CHCONHMe}^+ \\
\text{(c)} & \quad \text{MeHNOCH}_2 \text{N}_2 \text{CHCONHMe}^+ \\
\end{align*} \]

\[ \begin{align*}
\text{(a)} & \quad \text{MeHNOCH}_2 \text{N}_2 \text{CHCONHMe}^2+ \\
\text{(b)} & \quad \text{MeHNOCH}_2 \text{N}_2 \text{CHCONHMe}^+ \\
\text{(c)} & \quad \text{MeHNOCH}_2 \text{N}_2 \text{CHCONHMe}^+ \\
\end{align*} \]

\[ \begin{align*}
\text{(a)} & \quad \text{MeHNOCH}_2 \text{N}_2 \text{CHCONHMe}^2+ \\
\text{(b)} & \quad \text{MeHNOCH}_2 \text{N}_2 \text{CHCONHMe}^+ \\
\text{(c)} & \quad \text{MeHNOCH}_2 \text{N}_2 \text{CHCONHMe}^+ \\
\end{align*} \]
3. Results and Discussion

(a) The Tetramethylamide of EDTA from Spectral Data

The addition of methylamine to a solution of the tetramethylester of EDTA in methanol gives after refluxing the tetramethylamide of EDTA. This was characterized by C, H, N microanalysis and by $^1$H n.m.r spectroscopy (Table I). The i.r spectrum (Table 3) of the ligand shows an absorption band at 3340 cm$^{-1}$ which may be assigned to an NH stretching vibration. The ligand EDTA(NHMe)$_4$ also shows a broad band at 1640 cm$^{-1}$ shouldered by a weaker absorption band at 1530 cm$^{-1}$. These are the amide I, $\nu$(CO) and amide II, $\nu$(NH$_2$) bands respectively.\(^5\) The former band is normally the more intense of the two. The broadness of these bands is probably due to the presence of extensive hydrogen bonding. In the ligand, intramolecular and intermolecular hydrogen bonding is possible with the amide and amine nitrogen atoms, as well as the amide oxygen atoms all potential hydrogen acceptors. As well as band broadening, hydrogen bonding also causes shifts to lower wave number in the absorption under discussion.\(^5\)

(b) Potentiometric Titration of the Cu(II) EDTA (NHMe)$_4$ Complex

Aqueous solutions containing equimolar amounts of EDTA(NHMe)$_4$ and Cu(II) are diacidic and consume two moles of alkali per mole of metal ion (or ligand) in a potentiometric titration. This is consistent with the generally observed acidic behaviour of an amide group coordinated to Cu(II) (as discussed in Chapter I). The site of coordination normally changes from amide oxygen to nitrogen as a result of ionization. The ionizations relevant to base consumptions
are represented in equation (1). The resulting potentiometric curve and data are
given in Table 7 and Fig. 2. The pKₐ values were obtained using a MINIQUAD
Computer program.⁷ Two values were found in the normal pH range implying that
two ionizations occur in this range. The ionization constant pK₁ = 6.71 ± 0.01
and, pK₂ = 8.64 ± 0.01 were obtained. These values compare satisfactorily with
the ionization constants of the amide groups in the related bisglycinamidocopper
(II) complex cation for which pKₐ values of 6.91 and 8.12 have been reported
(equations 3, 4).⁸
Amide groups in Cu(II) peptide complexes are generally more acidic than the EDTA (NHMe)₄ complex just discussed.⁸⁻¹¹ The potentiometric evidence suggests that in aqueous solution of pH<4, the Cu(II)-EDTA (NHMe)₄ complex has structure (a) (equation 1). The conjugate base of this complex has structure (b) and reaches its maximum concentration in solution at pH≈ 7.5. At pH>10 the main species in solution has structure (c). In all cases aquo ligands are probably weakly bonded in axial positions to the metal ion.

(c) Structures of the Metal Complexes of EDTA (NHMe)₄ Inferred From Spectral and Magnetic Data

The products were obtained as a result of adding MX₂ or M'X₃ (where M = Cu²⁺, Co²⁺, Ni²⁺, Cd²⁺, Zn, Fe²⁺ and X = Cl, Br, I, NO₃, SO₄, M' = Fe²⁺, X = Cl or NO₃) dissolved in methanol to the tetramethylamine of EDTA also in methanol. Four of the products contain complex anions, e.g, [CuCl₄]²⁻, [CuBr₄]²⁻, [CoCl₄]²⁻ and [CdI₄]²⁻ in addition to the complex cations containing EDTA (NHMe)₄ ligands. The presence of the ligand (L) in these complexes is confirmed by the i.r spectra. When the ligand forms a complex with the metal, some of the hydrogen bonds are broken. Metal ions however also cause shifts to lower frequencies in the stretching bands described above. Therefore the position of the absorptions in the complexes relative to the free ligand are difficult to predict, since this largely depends on the relative effects of metal ions and hydrogen bonding in lowering band frequencies. The i.r spectra of all the complexes consist of intense broad absorptions in the region 3000-3500 cm⁻¹ and 1500-1700 cm⁻¹. The former region contains in addition to NH₂ stretching bands, absorptions due to lattice and coordinated water molecules.¹² The 1500-1700 cm⁻¹ region of the i.r spectra of the complexes contains a number of absorptions due to amide I and
II bands (Table 3).

**Copper (II) Complexes.**

The reflectance spectra of the Cu(II) complexes all have two bands in the region 350-750 nm (Table 4) the latter tailing into the near infrared region. This is a characteristic feature of many known Jahn Teller distorted six coordinated Cu(II) complexes. The main band is resolved into at least three components ascribable to \(d_{x^2-y^2} \rightarrow d_{xy}, d_{x^2-y^2} \rightarrow d_{z^2}, \) and \(d_{x^2-y^2} \rightarrow d_{xz}, d_{yz} \) 'hole' transitions. The energy level diagram for the 3d orbitals of Cu(II) in a distorted Oh field is shown in Fig.3. The transitions just mentioned involve the 'hole' in the \(d_{x^2-y^2} \) orbital.

In the case of the chloride complex a likely structure is \([CuL(H_2O)_2][CuCl_4]H_2O\) in which the cation may be a distorted octahedral Cu\(^{II}\) species in which L behaves as a tetradentate ligand (Fig.4) with two aquo ligands completing the octahedral coordination.

In the case of the bromide complex \([Cu_2L_2Br_2]CuBr_4\cdot2\text{CH}_3\text{OH}\) two possibilities exist:

(a) in one of these the cation is binuclear containing L or Br as bridging ligands.

(b) the other is the salt \([CuL_2]Cu_2Br_6\) in which the cation is a 2:1 complex (L is bidentate) and the anion is the well known \([Cu_2Br_6]^{2-}\).

The room temperature magnetic moments of the Cu(II) complexes (~ 1.8 B.M) are consistent with square, planar, square pyramidal or octahedral geometries, but give little further information other than to indicate they are magnet-
Nickel (II) Complexes

There are two bands in the region 350-750 nm for all the reflectance spectra of the nickel (II) complexes NiEDTA(NHMe)$_4$ X$_2.n$H$_2$O (X=Cl, Br, I, NO$_3$; n=1/2, 3, 5). There is another band at longer wavelength outside this scale. The three band spectrum is consistent with the presence of an octahedral complex of Ni(II) showing characteristic $^3A_2g \rightarrow ^3T_{2g}$, $^3A_2g \rightarrow ^3T_{1g}$($F$) and $^3A_2g \rightarrow ^3T_{1g}$($P$) transi-
tions which are generally observed. The energy levels involved in these transitions are shown in the partial energy diagram for Ni(II) in an octahedral environment (Fig.5). Only triplet states and the low energy singlet state derived from the Russell Saunders ¹D state of the free ion are represented in the diagram given below. The reflectance spectra (Table 4) of the complexes NiL(NO₃)₂1/2 H₂O and NiL₂ are identical which suggest an identical ligand environment. A postulated structure is given in Fig 6.

The visible bands for NiLCl₂.5H₂O and NiLBr₂.3H₂O occur at longer wavelength than those for the above two complexes suggesting that in these cases one or both of the weak field halide ligand may be coordinated. Alternatively these complexes may be formulated as [NiL(H₂O)₂]Cl₂.3H₂O and [NiL(H₂O)₂]Br₂.3H₂O in which the amide is present as a 2N (amine), 20 tetradentate ligand with two aquo ligands completing the octahedral coordination.

The room temperature magnetic moments (3.24-3.27 B.M) (Table 5) are also consistent with octahedral stereochemistry.¹⁴ In an octahedral field, the metal ion has two unpaired electrons and its ground state is a spin triplet. The spin only formula predicts a value of 2.8 B.M for the magnetic moment but the observed value generally exceeds this value due to a partial orbital contribution to the total magnetic moment.¹⁴ The i.r spectra (Table 3) of all the reported Ni(II) complexes are virtually identical. All have amide I bands at around 1630 cm⁻¹ and amide II bands of about one third the intensity at 1580 cm⁻¹. The NH stretching bands in all cases occur at ~3240 and 3100 cm⁻¹. In the hydrated complexes a (broad) OH stretching band occur at 3500 cm⁻¹. The relative molecular masses of these complexes (Table 6) were determined by EDTA titration, the percentage of metal in the complex was in close agreement with the calculated value.
Fig. 6 Partial Energy Level Diagram for Ni(II) in an Octahedral Field.

\[
\begin{align*}
\text{NHCH}_3 \\
\text{C} = & \text{O} \\
\text{HC}_2 & \text{CH}_2 \text{CH}_2
\end{align*}
\]

\[
\begin{align*}
\text{Ni} \\
\text{H}_{\text{CH}_2} & \text{C} = \text{O} \\
\text{O} & \text{C} \text{NHCH}_3
\end{align*}
\]

\[
\begin{align*}
\text{X}_2
\end{align*}
\]

\[
\begin{align*}
\text{X} = \text{NO}_3, 1
\end{align*}
\]

(6)
Cobalt(II) Complexes.

The reflectance spectra of the cobalt(II) complexes (Table 4) all have three bands in the region 350-750 nm. The three band spectrum is consistent with the presence of octahedral ($O_h$) high spin Co complexes for which three spin allowed transitions are expected. These are the $^4T_{1g}(F) \rightarrow ^4T_{2g}(v_1)$, $^4T_{1g}(F) \rightarrow ^4A_{2g}(v_2)$ and $^4T_{1g}(F) \rightarrow ^4T_{1g}(P) (v_3)$. The energy levels involved in these transitions are shown in the partial energy level diagram for a high spin octahedral Co(II) complex (Fig. 7) for $O_h$ symmetry. The first band is usually weaker than the third and the second is often unobserved. However a marked increase in the intensity of the $v_1$ and $v_2$ bands has been reported for some Co(II) complexes which deviate from $O_h$ symmetry. The observed room temperature magnetic

![Partial Energy Level Diagram for a High-spin Octahedral Co(II) Complex.](image)
magnitudes (4.30-4.37 B.M) (Table 5) for [CoL]CoCl₄, [CoL(H₂O)₂]Br₂ and CoL(NO₃)₂ are significantly outside the range of values normally observed for O₅ Co(II) complexes (4.7-5.2 B.M). In high spin octahedral complexes the ground state is ⁴T₁g and observed magnetic moments are much higher than spin only values (3.89 B.M) due to a considerable orbital contribution. In fields of less than O₅ symmetry the ground state is split with concomitant reduction in the orbital contribution.

![Diagram 8](image8)

![Diagram 9](image9)
The relative molecular masses of these complexes (Table 6) were determined by EDTA titration. The percentage of metal in the complex was in close agreement with the calculated value. The i.r spectra (Table 3) of all the Co(II) complexes have $\nu(\text{C}=\text{O})$ amide I and $\nu(\text{NH}_2)$ amide II bands at (1640 cm$^{-1}$) and (1570-1590 cm$^{-1}$) respectively. The NH stretching bands occur at (3100-3130 cm$^{-1}$) and (3240-3270 cm$^{-1}$) and in the hydrated complex the $\nu(\text{OH})$ stretching bands occur at 3500 cm$^{-1}$ broad).

Iron (II) Complexes.

The reflectance spectrum of the complex [FeL(H$_2$O)$_2$]SO$_4$.4H$_2$O (Table 4) contains a band at 400 nm which appears to be split. In a high spin octahedral field the ground state of Fe(II) is $^6T_{2g}$ and the 400 nm band is due to the spin allowed transition to the $^6E_g$ state, the degeneracy of which should be lifted by the Jahn-Teller effect.$^{12}$ The observed room temperature magnetic moment (5.29 B.M) is typical of a magnetically dilute Fe(II) complex in a high spin $O_h$ field.$^{16}$ On this basis and on the basis of spectral data it would appear that the amide ligand may be tetridentate or hexadentate in the above complex with four or six lattice waters.
Iron (III) Complexes.

The reflectance spectra of the Fe(III) complexes (Table 4) contain two bands in the region 350-750 nm. There is another band at longer wavelength outside this scale and this corresponds to the presence of octahedral complexes of Fe(III). Less is known about Fe(III) spectra due to the much greater tendency of the trivalent ion to have charge transfer bands in the near ultraviolet region. The observed room temperature magnetic moment are 5.26 and 5.40 B.M for FeL(NO$_3$)$_3$.3H$_2$O and FeLCl$_3$.2H$_2$O respectively. The i.r spectra of these complexes have $\nu$(C=O) amide I band and $\nu$(NH$_2$) amide two bands at 1640-1650 cm$^{-1}$ and 1570-1580 cm$^{-1}$ respectively. The NH stretching bands occur at 3100-3440 cm$^{-1}$ and the OH stretching bands occur at 3500 cm$^{-1}$ (broad).

![Chemical structure](image)

(11)
Cadmium (II) Complex.

The product obtained as a result of adding CdI₂ solution in methanol to the ligand solution in methanol gives a salt containing the complex anion \([\text{CdI}_4]^{-}\). The relative molecular mass of the complex (Table 6) was determined by EDTA titration. The percentage of metal in the complex was in close agreement with the calculated value. The i.r spectra (Table 3) of the Cd(II) complex has \(\nu(C=O)\) amide I and \(\nu(NH_2)\) amide II bands at 1640 cm\(^{-1}\) and 1565 cm\(^{-1}\) respectively.

\[
[\text{CdL}][\text{CdI}_4]
\]

Zinc (II) Complex.

The i.r spectrum of Zn(II) complex has \(\nu(C=O)\) amide I band and a \(\nu(NH_2)\) amide II band which occur at (1640 cm\(^{-1}\), 1570 cm\(^{-1}\)) respectively. The NH stretching bands occur at (3100, 3240 cm\(^{-1}\)) and OH (stretching) band occurs at 3500 cm\(^{-1}\) (broad). From the spectral data, the ligands act as tetradentate to the Zn(II) metal ion, in which the metal cation may be octahedral with two aquo ligands completing octahedral coordination\(^{14}\).
References


CHAPTER 5

Metal Complexes of Amino Acid Derivatives.
The Copper(II) Promoted Hydrolysis of
1,2-Bis(3,5-dioxopiperazin-1-yl) Propane
(BDOPPP).
1. Introduction

Metal ions play an important and versatile role in biological and medicinal chemistry, for instance iron in oxygenation and redox haemoproteins, zinc in hydrolytic metalloenzymes, platinum complexes, such as cis-platin, in anticancer therapy. There is also a group of dioxopiperazine drugs with anticancer properties which depend on the metal chelating properties of their hydrolysed derivatives. This study deals with the copper(II) promoted hydrolysis of 1,2-bis(3,5-dioxopiperazin-1-yl) propane (BDOPPP) which is a precursor of razoxane, (1), a bisdioxopiperazine anti-tumor drug. Scheme 1 shows BDOPPP (1) and its metabolic product, in vivo (2).

![Scheme 1](image-url)

Scheme 1
Razoxane is a member of a family of drugs developed by the Imperial Cancer Research Fund laboratories (ICRF) after it had been noted by Furst that many anti-tumor drugs could act as chelating agents. The first compound to show activity was the bis cyclic imide of EDTA (ICRF 154), where activity was noted in experimental tumors but not in clinical trials. This was attributed to the poor absorption of the relatively insoluble agent and resulted in the development of substituted bisdioxopiperazanes (3).

![Chemical Structure of ICRF 154](image)

Studies have been reported which correlate activity to structure and to potential lipophilicity (Figure 1). In both cases activity was determined by dosage required (M) to reduce the colony forming ability of mouse L cells by 50%, while potential lipophilicity was determined by taking the log of the partition coefficient (p) of the substituted bisdioxopiperazine in an octanol-water system (Table 1).

Hence it can be seen that BDOPPP (ICRF 159) exhibits sufficient lipophilicity to enter the body, but would have to be hydrolysed to the active form in vivo. This reaction would depend on metal ion catalysis.

A similar role for metal ions in the promoted hydrolysis of amino acid esters, amides and peptides has been well documented. For instance the base hydrolysis of ethyl glycinate is enhanced 1.3x10^5 fold in the presence of Copper(II). Corresponding information for metal promoted imide hydrolysis has not been reported,
Table 1. ID₅₀ and log p values for substituted dioxopiperazines

<table>
<thead>
<tr>
<th>ICRF No.</th>
<th>R</th>
<th>R'</th>
<th>ID₅₀</th>
<th>log p</th>
</tr>
</thead>
<tbody>
<tr>
<td>154</td>
<td>H</td>
<td>H</td>
<td>7.300</td>
<td>-2.34</td>
</tr>
<tr>
<td>159</td>
<td>CH₃</td>
<td>H</td>
<td>3.000</td>
<td>-1.05</td>
</tr>
<tr>
<td>192</td>
<td>C₂H₆</td>
<td>H</td>
<td>720.000</td>
<td>-2.12</td>
</tr>
<tr>
<td>193</td>
<td>CH</td>
<td>CH₃</td>
<td>0.090</td>
<td>-0.78</td>
</tr>
<tr>
<td>197</td>
<td>cb</td>
<td>cb</td>
<td>55.000</td>
<td>-1.51</td>
</tr>
<tr>
<td>202</td>
<td>CH₃</td>
<td>C₂H₆</td>
<td>0.045</td>
<td>-0.37</td>
</tr>
</tbody>
</table>

cb = cyclobutane

hence the current study. BDOPP should complex to copper(II) as in structure (4) in view of the high affinity of this ion for N donors. However in an aqueous system it would be expected to undergo metal promoted base hydrolysis as shown in Scheme 2. Since the opening of the imide rings generates good complex-
ing carboxylate sites, the coordination around the cooper(II) ion should occur as shown.

\[
\begin{align*}
\text{Scheme 2}
\end{align*}
\]

Hence it can be seen that the hydrolysis of BDOPPP in the presence of copper(II) produces \(H^+\). This makes the pH stat method an excellent method for following the kinetics of this reaction.

2. Experimental

Preparation of BDOPPP

Propane-1,2-diaminotetraacetic acid (10g, 0.033 mole) and formamide (40
cm³, 1.005 moles) were heated together under nitrogen in a three-necked quick fit flask at a reduced pressure of 200mm Hg and a temperature of 110-120°C for 1 hour. The temperature was then raised to 150-155°C for a total of five hours using an oil bath. A solution formed after 30 minutes. After 5 hours heating the mixture was cooled to room temperature and kept overnight. The crystalline product was collected by filtration, washed with cold methanol, then ether and air dried. Yield=3.4g (38.5%) m.p=236-238 °C (lit value 237-239°C). Microanalysis for BDOPPP (ICRF 159) (1); found C,49.01 H, 6.07, N, 20.59%. C₁₁ H₁₅ N₂ O₄ requires C, 49.25, H, 5.97, N, 20.89%.

Copper nitrate solutions were prepared at the concentrations as given in Table 2. Ionic strength (I) was maintained constant by the use of the appropriate concentration of sodium perchlorate as given by equation (1).³

\[ I = 0.5 \sum (c^2z^2) \]

where \( z = \) charge and \( c = \) concentration.

Table 2. Concentrations of reaction solutions (M)

<table>
<thead>
<tr>
<th>Conc. of Cu(NO₃)₂ (M)</th>
<th>Conc. of NaClO₄ (M)</th>
<th>Ionic strength</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.1</td>
<td>0.00</td>
<td>0.3</td>
</tr>
<tr>
<td>0.075</td>
<td>0.075</td>
<td>0.3</td>
</tr>
<tr>
<td>0.05</td>
<td>0.15</td>
<td>0.3</td>
</tr>
<tr>
<td>0.025</td>
<td>0.225</td>
<td>0.3</td>
</tr>
<tr>
<td>0.0125</td>
<td>0.2625</td>
<td>0.3</td>
</tr>
</tbody>
</table>

Kinetics measurements were performed using the pH stat method.⁵ This
involved titrating BDOPPP (2×10⁻⁴ moles, 0.0536g) dissolved in DMF/H₂O (10:25cm³, Analar grade DMF) with sodium hydroxide (0.1M) in the presence of varying concentrations of copper(II) (8.9×10⁻³ — 0.071 M). The concentrations given, account for the dilution due to the DMF which was necessary to dissolve the imide. Reaction temperature was maintained at 25°C ± 0.1°C by carrying out the reaction in a double walled vessel (50ml) through which water from a constant temperature water bath was passed. Titrations were performed using an autotitrination assembly consisting of a Radiometer Autoburette ABU12+TTA60 and recorded on a REA160 Titrigraph module. This titration assembly was also used to carry out an acid/base titration on the diimide. In this case no copper(II) was employed and for the acid titration HCl (0.1 M) was used.

3. Results and Discussion

BDOPPP is a diacid base where the two tertiary amine nitrogens may be protonated according to the equilibria in scheme 3.

The plot of the titration with acid is shown in Figure 2. From this data a pKa value was found using the MINIQUAD program. Only one value was found in the normal pH range implying that only one nitrogen is protonated in this range. The value found was pKa=2.68 (t=25+/−0.1°C ) and this corresponds to the pKa value of the conjugate acid (6).

An i.r spectrum was obtained (see Spectrum 1) as a nujol mull using a Perkin Elmer Model 577. Assignments were made as shown in Table 3.

The position of the carbonyl stretch is unusual, but Conley suggests this is typical of a cyclic ring. This feature could be investigated in the products of the
Fig 2. Plot of pH vs volume for acid titration of BDOPPP.
hydrolysis, if they could be isolated from solution. The C-N doublet at 1175 and 1148 cm$^{-1}$ is characteristic of tertiary amines$^{10}$.

The hydrolysis of BDOPPP in the presence of copper(II) was studied at constant pH in the pH range 4 to 5. An excess of copper(II) was used to ensure that only mono-ligation occurred and that no 2:1 complexes were formed. The reaction in scheme 2 would normally be expected to follow second order kinetics, equation (2).

$$Rate = k_2[\text{complex}(4)][OH^-]$$  \hspace{1cm} (2)

At constant pH, $[OH^-]$ is constant and the reaction becomes pseudo first order.

$$Rate = k_{obs}[\text{complex}(4)]$$  \hspace{1cm} (3)

where $k_{obs}$ is the pseudo first order rate constant and is equal to $k_2[OH^-]$. A typical
Table 3. I.R Assignments for BDOPP (1)

<table>
<thead>
<tr>
<th>Assignment</th>
<th>Frequency (cm⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>N-H (stretch)</td>
<td>3405 (w), 3215 (w), 3098 (w)</td>
</tr>
<tr>
<td>C=0 (stretch cyclic ring)</td>
<td>1715 (s,b)</td>
</tr>
<tr>
<td>N-H (amide III band)</td>
<td>1306 (w,sh), 1285 (w,b)</td>
</tr>
<tr>
<td>C-N (stretch, aromatic character)</td>
<td>1360 (w,sp), 1334 (w,sp)</td>
</tr>
<tr>
<td>C-N (stretch, aliphatic from amine)</td>
<td>1175, 1148 (w,d)</td>
</tr>
</tbody>
</table>

Abbreviation: w = weak, s= strong, b = broad, sh = shoulder, sp = sharp, d = doublet.

The kinetic plot from the REA 160 Titrigraph, displaying volume of base added vs time, is shown in Figure 3. The reaction is clearly first order. The data were analysed by two methods and observed rate constants calculated. These methods were:

1. The Vₐ method: ln(Vₐ-Vₜ) vs t was plotted, the slope of the plot giving kₜₐₜ, where Vₐ = final volume of titre, and Vₜ = volume titre at time t.
2. The Guggenheim method: ln (Vₜ+2[t/1/2]-Vₜ) versus t was plotted. Again the slope of the plot gives kₜₐₜ, where t[1/2] = half life of reaction.

Generally method (1) was used when the Vₐ volume was known, otherwise method (2) was employed. Tables 4-6 give values of kₜₐₜ against [Cu²⁺] at fixed pH.

By using high concentrations of copper(II) it was hoped to push the complex formation equilibrium completely to the right, i.e towards the product complex.
Table 4. $K_{obs}$ vs [Cu$^{2+}$] at pH 4.02

<table>
<thead>
<tr>
<th>[Cu$^{2+}$] M</th>
<th>$K_{obs}$ (min$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0178</td>
<td>0.015</td>
</tr>
<tr>
<td>0.0357</td>
<td>0.036</td>
</tr>
<tr>
<td>0.0536</td>
<td>0.059</td>
</tr>
<tr>
<td>0.0714</td>
<td>0.082</td>
</tr>
</tbody>
</table>

Table 5. $K_{obs}$ vs [Cu$^{2+}$] at pH 4.51

<table>
<thead>
<tr>
<th>[Cu$^{2+}$] M</th>
<th>$K_{obs}$ (min$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0178</td>
<td>0.052</td>
</tr>
<tr>
<td>0.0357</td>
<td>0.092</td>
</tr>
<tr>
<td>0.0536</td>
<td>0.158</td>
</tr>
<tr>
<td>0.0714</td>
<td>0.290</td>
</tr>
</tbody>
</table>

Table 6. $K_{obs}$ vs [Cu$^{2+}$] at pH 5.01

<table>
<thead>
<tr>
<th>[Cu$^{2+}$] M</th>
<th>$K_{obs}$ (min$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0089</td>
<td>0.059</td>
</tr>
<tr>
<td>0.0178</td>
<td>0.175</td>
</tr>
<tr>
<td>0.0357</td>
<td>0.349</td>
</tr>
</tbody>
</table>

(4). If this occurred the reaction rate would be independent of [Cu$^{2+}$]. However the dependence of $k_{obs}$ on [Cu$^{2+}$] implies that this has not been accomplished.

The chelation between $[\text{Cu(H}_2\text{O)}_6]^{2+}$ and BDOPPP and the subsequent hydrolysis can be represented according to scheme 4.
Fig. 3 BDOPPP monitored by pH-stat
The rate of the reaction is given by

\[
Rate = k_2 [OH^-]^{41}\n\]

\[
= k_2 K[OH^-][Cu^{2+}][BDOPPP] \n\]

\[
= k_{obs}[BDOPPP] \n\]

at constant pH and with Cu(II) in vast excess, where \( k_{obs} = k_2 K[OH^-][Cu^{2+}] \).
Reaction scheme 4 shows that the hydrolysis of BDOPPP should be dependent on $[\text{OH}^-]$ concentration. This dependence (Figure 5) appears to be first order in $[\text{OH}^-]$. The constants $k_2 K[Cu^{2+}] (= k_2)$ are calculated from the slope of the plot (Figure 4) of $k_{obs}$ (ordinate) vs $[\text{OH}^-]$ concentration. These are:

$$k_2 = 1.7 \times 10^8 M^{-1} min^{-1} (0.0178 M[Cu^{2+}])$$

$$k_2 = 3.3 \times 10^9 M^{-1} min^{-1} (0.0357 M[Cu^{2+}])$$

From these constants values of $K_2 K(M^{-2} min^{-1}) = 9.55 \times 10^9$ and $9.24 \times 10^9$ were calculated.

The hydrolysis of the free ligand BDOPPP studied at pH 9.00 and 25±0.1°C also by the pH stat method, was very slow (Figure 5) and approximate rate constants were calculated from the estimated half lives. A summary of the results of the hydrolysis of BDOPPP are presented below

$$\text{half life(min)} = 518.5$$

$$k_{obs} (min^{-1}) = 1.336 \times 10^{-3}$$

$$k_2 (M^{-1} min^{-1}) = 133.6$$

In the presence of copper(II) it is clear that a very large rate enhancement is observed for the hydrolysis of BDOPPP. At $[Cu^{2+}] = 0.0357$ M the rate enhancement is $2.47 \times 10^6$, while at $[Cu^{2+}] = 0.0178$ M the rate enhancement is $1.27 \times 10^6$. Obviously the rate enhancement is increasing with increasing $[Cu^{2+}]$. This is because of incomplete complex formation under our experimental conditions and the rate enhancement when the complex is fully formed would be much higher than $2.47 \times 10^6$. However, this would require concentrations of Cu(II) that were too high to achieve.
It was necessary to ensure that during all of the previous reactions DMF was not hydrolysed as shown in Scheme 5.

Initial investigation using the pH stat method at 25+/−0.1°C for solution of Cu²⁺ (0.0714M) in DMF/H₂O (10:25) revealed no measureable hydrolysis at pH 4.01. This is to be expected on the basis of the work by Langois and Broche¹² who studied the base hydrolysis of DMF in the temperature range 70-90°C with varying concentration of base. By using published value of Eₐₑₓ, 14.9 kcal mol⁻¹ and a pre-exponential factor A, of 1.44×10⁷ and applying the Arrhenius Equation,¹⁹

\[
\log K = \log A - \frac{E_{act}}{2.3RT}
\]  

the second order rate constant for the base hydrolysis of DMF at 25°C is 

\[k_2=0.01M^{-1}\text{min}^{-1}\]. Hence in the range pH 4 to pH 5 the observed rate constant for this reaction will be in the order of 1—10×10⁻¹²min⁻². More recent studies¹⁴ have shown the DMF hydrolysis may also exhibit rate enhancement by metal ion promotion. Using the reported enhancement factor of 1×10⁴ as a guide still leaves an observed rate constant for DMF hydrolysis which will have a negligible effect on the result of our work.

Metal ion promoted hydrolysis of amino-acid esters and peptides has been a subject of interest since the early 1930's when the phenomenon was first noted with peptides. However the discovery by Kroll¹⁶ in 1952 that the hydrolysis of α-amino
Fig 4. Plot of $[\text{OH}]$ vs $K_{\text{obs}}$
VOLUME OF TITRANT (ARBITARY VALUE)

Fig. 5 pH-stat monitored BDOPPP hydrolysis.
acid esters was also catalysed by metal ions renewed interest in this area. Many of these reactions provide simple models for more complex metalloenzymes such as carboxypeptidase A, leucine aminopeptidase and glycyglycine dipeptidase.\textsuperscript{16}

One of the largest problems in studying these reactions has been the determination of the binding sites of the ligand to the metal and the actual species involved in the hydrolytic reaction. This has been largely overcome by the use of polydentate ligands where the formation constants are higher. The development of the pH stat method has further simplified the problems of interpreting the data by removing the need for a buffer system to maintain constant \([\text{OH}^-]\), thus removing possible metal ion-buffer interactions.

In the case of labile metal ions ambiguities still exist regarding mechanisms and this is illustrated by the copper(II) catalysed hydrolysis of ethyl glycinate as shown in scheme 6\textsuperscript{16} (the intermolecular pathway) and scheme 7\textsuperscript{17} (the intramolecular pathway). Previously these mechanistic pathways have only been resolved used kinetically inert systems, such as cobalt (III).

Scheme 6 involves chelation of the amino acid ester to the metal cation, followed by nucleophilic attack by hydroxide ion on the carbonyl group. The rate enhancement occur because the electron withdrawing effect of the metal in such a chelate increases the susceptibility of the carbonyl group to attack by nucleophiles.

The second mechanism does not involve chelation. In this mechanism a hydroxy ligand (conjugate base of an aquo ligand) attacks the uncomplexed carbonyl group intramolecularly, the large rate enhancement is achieved by the juxtapositioning of the two reaction sites. In both cases the reactions obey second order kinetics:

\[
\text{Rate} = k_2 [\text{reacting complex}] [\text{OH}^-]
\]  

(5)
where $k_2$ for scheme 6 is as indicated and $k_2$ for scheme 7 is equal to $k_1K$ where these are as indicated. It has been shown\(^\text{18}\) that rate enhancements of up to $10^{11}$ fold are achieved with the intramolecular pathway, while those for the intramolecular pathway tend to be in the order of $<10^7$.

The rate enhancements observed in the reaction studied in this work tended to indicate that the intramolecular mechanism was the more likely. The feasibility of the two pathways were investigated by the use of CPK molecular models (Figure 6-8). The models revealed that chelation of the carbonyl group to the copper(II) ion was rendered impossible because of steric contraints and the intramolecular mechanism such as that in scheme 6 is therefore impossible. However the models show that the intermolecular attack by coordinated hydroxide ligand on the un-
The complexed carbonyl group is quite feasible and that a pathway as shown in scheme 7 is applicable. Because of geometric factors we here have a system for which only one of the mechanisms can operate, the first time this has been accomplished using a labile complex.

The models also explain why only sufficient hydroxide ion is consumed in the pH stat experiments to hydrolyse one and not two imide rings, as shown in the proposed pathway, scheme 8.
Scheme 8
The hydrolysis of one ring makes available two coordination sites, i.e. the -CO₂ and -CONH₂ groups. When the first ring is hydrolysed the coordination around the metal ion changes as shown and the influence of the metal ion, i.e. its catalytic effect, on the second imide ring is removed. The use of molecular models show that complexing of the metal ion to the amino N, -CO₂ and -CONH₂ groups as shown in (XIX) makes coordination of the other amino group highly unfavourable although a weak interaction with the metal, particularly in view of the John Teller effect might be permitted. This interaction however does not result in any noticeable catalysis.
Fig 6. This shows that the copper centre (grey sphere) cannot complex with the carbonyl oxygen (red).
Fig 7. This shows the feasibility of coordinated hydroxide attacking carbonyl carbon.
Fig 8. The hydrolysed imide with one intact ring removed from the metal coordination sphere.
4. Summary

BDOPPP has been shown to undergo a rate enhancement of approximately $1 - 2.5 \times 10^6$ fold in the presence of copper(II) at a range of values of pH 4-5 and a temperature of 25°C. An intermolecular mechanism for the hydrolysis has been proposed.
References


