Bioinorganic and Bioorganic Chemistry

Studies on the Formation of Complexes between Mn(II), Co(II), Ni(II) and Cu(II) Cations and Diphosphate, Adenosine Monophosphoric Acid and Adenosine Diphosphoric Acid Anions in Water–Tetramethyl Ammonium Bromide 0.2 *M* Medium

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The equilibria of the formation of complexes between the metallic cations Mn^{2+}, Co^{2+}, Ni^{2+} and Cu^{2+} and the anionic ligands diphosphate ($HP_2O_7^{3-}$, $P_2O_7^{4-}$) adenosine monophosphoric (AMP^{2-}) and adenosine diphosphoric ($HADP^{2-}$, ADP^{3-}) have been studied, in a water–TMA^+Br^- medium, by variable pH potentiometry and by pH-stat potentiometry [1, 2].

This work allows us essentially to admit the existence of four types of complexes [MHL, $M(HL)_2$, ML, ML_2] and to determine the values of their corresponding formation constants [1, 3–8]. The thermodynamic quantities corresponding to the formation of complexes have been determined by Van t'Hoff rule and by pH-stat calorimetry [9]. The values obtained by these two different methods are quite consistent and point out the importance of the variations of entropy (ΔS) in the formation of complexes (*e.g.* Table I).

With regard to the variations of entropy, the results show that the values of ΔS are important and increasing with temperature. This phenomenon can be explained by a reorganization of the molecules of the water of the medium between the metallic cations, the ligands and the resulting complexes, consequently to the important disappearance of electric charges.

TABLE I. Comparison between the Results Obtained by Potentiometry and by Calorimetry, TMA^+Br^- 0.2 *M* Medium, pH = 7, at 25 °C.

Complexes	ΔH(Kcal/mol)		log *K*		ΔG(Kcal/mol)		ΔS(cal/mol)	
	pot.	cal.	pot.	cal.	pot.	cal.	pot.	cal.
CoAMP	–0.1	–0.18	2.34	2.37	–3.20	–3.22	10.40	10.18
NiAMP	–2.1	–2.46	2.46	2.48	–3.36	–3.38	3.5	3.05

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The Formation and Structure of Cu(II)–Clupeine Z Complexes

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The interaction of metal ions with DNA and the role it plays in biological processes has been a field of permanent interest and the object of many studies [1]. Recently, transition metal ions, including that of copper, have been reported to occur naturally in deoxyribunucleoproteins [2]. Although the interaction of Cu(II) with DNA has been thoroughly investigated [3], the specific binding ability of Cu(II) towards basic proteins at physiological pH has been overlooked. In this report we present the results of an investigation aiming to characterize complex formation between Cu(II) and one component of the protamine associated with DNA in herring sperm nuclei, namely, Clupeine Z. This protein contains 32 aminoacid residues, two-thirds of which are arginine [4].

At [Clu]/[Cu] = 1 titration curves show that at pH 6.6 two protons per Cu(II) are released. A first complex(I) is formed at this stage, the CD spectrum of which displays two negative bands at 660 and 275 nm and a positive one at 315 nm. The last is compatible with peptide involvement in coordination, while the negative peak at 275 nm is characteristic of metal binding to the terminal amino-nitrogen [5]. A second complex(II) is formed at pH 8.5 when two additional protons are removed. Its CD spectrum presents two bands: a negative one at 550 nm and a positive one at 255 nm. The first is consistent with binding to amino nitrogens of lateral chains [6]. Figure 1 illustrates the species distribution in this system obtained by treatment of titration data. The equilibrium constants defined by

$$K_I = \frac{[\text{Complex I}]\,[\text{H}^+]^2}{[\text{Cu}]\,[\text{Clu}]} \quad \text{and}$$

$$K_{II} = \frac{[\text{Complex II}]\,[\text{H}^+]^4}{[\text{Cu}]\,[\text{Clu}]}$$

where [Clu] = fully protonated [Clu] concentration, have been evaluated as: $K_I = 10^{-9}$ mol l^{-1}, and $K_{II} = 10^{-23}$ mol^3 l^{-3}.

The proposed structures of both complexes are shown in Fig. 2.

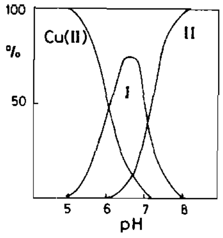

Fig. 1. Species distribution in Cu(II)–CluZ system as a function of pH.

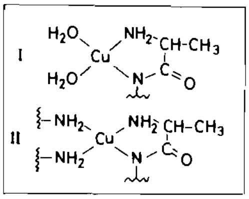

Fig. 2. Proposed structures of complexes I and II.

When [Clu]/[Cu] = 1/2, at pH 6.6 there is only one Cu(II) bound per protein molecule (site 1) and two protons are released. Spectral data are consistent with complex I formation as previously. From pH 6.6 to 8.5 the second Cu(II) bounds to the protein and four additional protons are neutralized. The spectral patterns suggest that in addition to complex II which is formed at site 1, another complex(III) is formed at site 2. In this complex two amino nitrogens of lateral chains and two oxygens of water molecules lie at the corners of the coordination square.

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Thermodynamics of Mixed Complexes of Cu(II) and Zn(II) with ATP and Some Aromatic Aminoacids

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Up to now mainly simple complexes of divalent metal ions with nucleotides have been studied in solution [1–4]; it is only for a few years that the stoichiometry, the formation constants and the structure of mixed complexes of M(II)–*NT* (*NT* = nucleotide) with biofunctional ligands have been reported [4–6]. By means of spectroscopic investigations two different types of ternary complexes have been investigated; these complexes can be represented as *L*–*NT*–M(II) and *L*–M(II)–*NT* (*L* = biogenic amines, aminoacids, *etc*...) [7]. In such mixed complexes an interesting 'secondary' bonding, due to formation of stacking adduct between the base of the nucleotide and the 'aromatic' moiety of other ligand, has been described [6]. Recently [8, 9], we have determined, by direct calorimetry, the effect of stacking interaction on the thermodynamic parameters concerning the formation of copper(II) and zinc(II) mixed com-