

## The crystal structure of (L-proline-L-alanine) copper (II) · 2.5 H<sub>2</sub>O

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**Abstract.** The crystal and molecular structure of (L-proline-L-alanine) Cu · 2.5 H<sub>2</sub>O has been determined by the heavy atom method with 583 visually estimated reflexions and refined by full matrix least squares to  $R = 0.101$ . The crystals are monoclinic with  $a = 16.47(3)$ ,  $b = 7.74(2)$ ,  $c = 9.51(3)$  Å and  $\beta = 109.7(5)^\circ$ ; space group C2;  $Z = 4$ . The copper coordination is nearly square pyramidal with one water in the basal plane and another occupying the apical together with the amino N, peptide N and the carboxyl O of the dipeptide in the coordination. The Cu-peptide-Cu linkage is *via* the hydrogen bonding between the water in the basal plane and the carbonyl O atom of the proline residue. One water is on the 2-fold axis.

**Keywords.** Cu coordination in (L-proline L-alanine) Cu(II)2.5 H<sub>2</sub>O.

### 1. Introduction

This paper on the structure determination is third in the series on Cu complexes of biomolecules, the first being bis (L-threonine) Cu (ii) H<sub>2</sub>O and the second, (L-valine-L-tyrosine) Cu (ii) 4·H<sub>2</sub>O-CULVAT (Amirthalingam and Muralidharan 1975, 1976). Here the interest is to find out the nature of Cu coordination in biological system and to investigate the geometry of the dipeptide ligand. It will also be interesting to find out the conditions under which water molecules prefer to go into coordination with Cu.

### 2. Experimental

The compound (L-proline-L-alanine) Cu (ii) 2.5 H<sub>2</sub>O, CUPRAL was prepared by reacting freshly prepared Cu(OH)<sub>2</sub> with the dipeptide (pH 7.6) and crystallined from aqueous solution as stable blue plates. X-ray diffraction studies with Cu K<sub>α</sub> showed that the crystals are monoclinic with  $a = 16.47(3)$ ,  $b = 7.74(2)$ ,  $c = 9.51(3)$  Å and  $\beta = 109.7(5)^\circ$ . The space group was fixed as C2 (rejecting the Cm or C2/m) since the ligand is optically active. (The systematic absences are  $hkl$ ,  $h+k = 2n \pm 1$ ).  $Z = 4$  of molecular formula Cu (C<sub>8</sub>H<sub>13</sub>N<sub>2</sub>O<sub>3</sub>) · 2.5 H<sub>2</sub>O ( $\rho_0 = 1.70$   $\rho_c = 1.71$  g·cm<sup>-3</sup>) With the crystal mounted along  $b$  and using Cu K<sub>α</sub> radiation, intensities for 583 reflexions were collected and visually estimated. Attempts to get fifth and the higher layer reflexions failed because they were extremely weak and diffuse. No absorption correction was made in view of the small size of the crystal used. The mean

cross sections was 0.27 mm. Scaling was carried out using Wilson's method together with the common reflexions obtained from  $h k 0$  and  $0 k l$  data.

### 3. Structure determination

The Cu position ( $x$  and  $z$ ) was fixed from the Harker section at  $y = 0$ . The  $y$  coordinate was taken as zero. The resulting Fourier map contained many spurious peaks in addition to the duplication due to the false mirror. Assuming a trial model of nearly square planar coordination of Cu, four more atoms were fixed and by iterative Fourier methods, the complete structure was solved.

At this stage, it became clear that one water molecule is in the basal plane while the other occupied the apical. Also there was a water O on the 2-fold axis itself. The structural parameters including scale were refined by full matrix least squares, using Cruickshank's weighting scheme with  $A = 5.0$  and  $C = 0.01$ . H atoms were ignored. Anisotropic thermal parameters were used only for the Cu atom. It was found that the  $C^\gamma$  of the pyrrolidine ring was highly disordered. In the final stages, Fourier and difference Fourier maps (computed without the contribution of this atom C(2)) showed a highly elongated, ellipsoidal electron density for this atom, indicating that the disorder is rather a thermal one as opposed to statistical disorder. Hence a sterically allowed position for this atom was assumed and only its temperature factor was refined in the final stages and its final value happens to be  $11.0 \text{ \AA}^2$  (table 1). The final  $R$  for all observed reflexions is 0.10 1.<sup>†</sup>

**Table 1.** Final positional ( $\times 10^4$ ) and thermal parameters of the atoms with e s d 's in parenthesis.

	$x$	$y$	$z$	$B$
Cu	1941 (1)	0	1935 (1)	—
O(1)	2360 (7)	4981 (15)	2011 (8)	4.8 (4)
O(2)	2952 (6)	-1041 (14)	1522 (7)	3.4 (5)
O(3)	4170 (6)	-431 (14)	1170 (7)	3.1 (4)
O(4)W	1493 (5)	-2289 (13)	2062 (13)	2.8 (3)
O(5)W	1075 (5)	841 (14)	-811 (7)	3.5 (4)
O(6)W	0	6708 (15)	0	3.8 (4)
N(1)	1076 (4)	1347 (11)	2461 (6)	2.5 (3)
N(2)	2631 (4)	2075 (11)	2170 (6)	2.0 (3)
C(1)	973 (5)	914 (12)	3916 (7)	3.5 (4)
C(2)	1501	2361	4941	11.0 (18)
C(3)	1371 (6)	3980 (12)	3941 (7)	4.6 (7)
C(4)	1240 (6)	3232 (12)	2372 (8)	4.3 (7)
C(5)	2120 (4)	3483 (11)	2140 (6)	1.2 (3)
C(6)	3400 (4)	2042 (11)	1752 (7)	3.4 (4)
C(7)	3543 (5)	82 (12)	1461 (7)	3.2 (5)
C(8)	4180 (6)	2571 (13)	3165 (8)	4.8 (8)

Anisotropic temperature factor for Cu of the form

$$\exp [-(\beta_{11}h^2 + \beta_{22}k^2 + \beta_{33}l^2 + 2\beta_{12}hk + 2\beta_{23}kl + 2\beta_{13}hl)]$$

$$\begin{array}{ccc} \beta_{11} & \beta_{22} & \beta_{33} \\ 0.0027 (1) & 0.0101 (21) & 0.0096 (3) \\ -0.0018 (7) & 0.0064 (3) & -0.0005 (13) \end{array}$$

<sup>†</sup>A list of  $F_o$  and  $F_c$  can be obtained from the authors on request.

The final positional and thermal parameters are given in table 1. The form factors for Cu<sup>2+</sup>, N, C and O were taken from *International Tables for X-ray crystallography* (1962). Anomalous dispersion correction for Cu ( $\Delta f' = -2.1 \epsilon$   $\Delta f'' = 0.7$ ) was used in the refinement. The structure projected down *c*-axis is shown in figure 1 and bond angles and lengths in figure 2 (a, b).

#### 4. Description and discussion of the structure

The dipeptide acts as a tridentate ligand, with amino N, peptide N and carboxyl O (of the alanine residue) coordinating to Cu. The fourth basal as well as the apical positions are occupied by water molecules. The Cu-peptide-Cu linkage is *via* the hydrogen bonding between the water in the basal plane (O (4)) and the carbonyl O atom (O (1)) of the proline residue. There is a network of hydrogen bonds

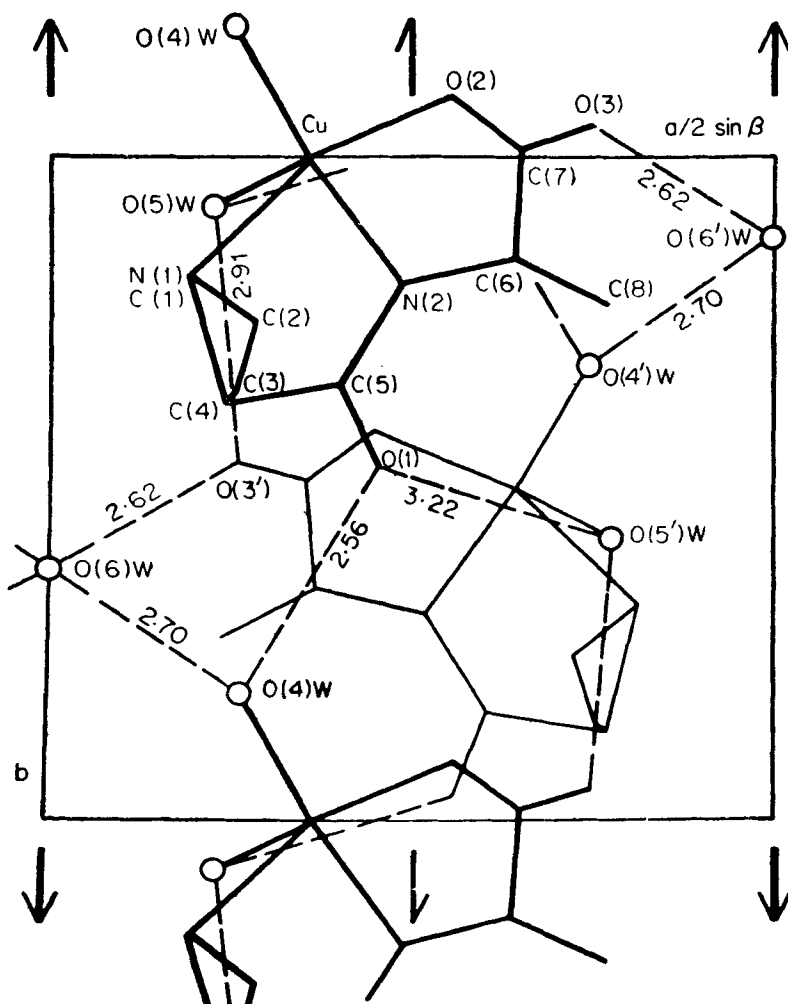


Figure 1. Structure projected down *c*. Broken lines indicate possible hydrogen bonds.



between water molecules and the carboxyl O atoms of alanine and O atom of proline residue.

The Cu (ii) coordination (figure 3) is square pyramidal. The bond lengths and angles around the metal atom are quite normal and are very similar to those found in CULVAT and other similar Cu (ii) complexes (Freeman 1966, 1967). The apical water is 2.61 Å away. The basal plane (plane C) is slightly distorted square planar and Cu is displaced from this plane by 0.122 Å towards the apical water molecule (table 2).

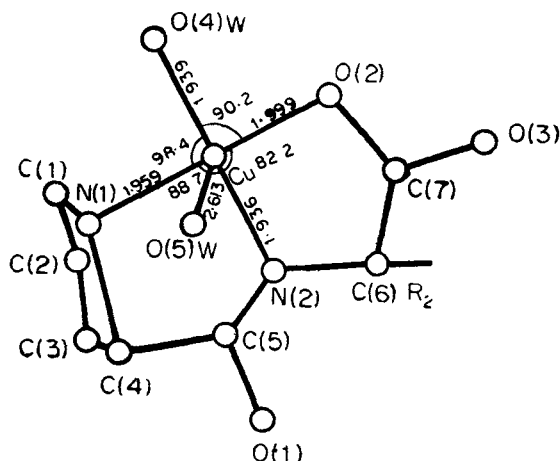


Figure 3. Copper coordination.

Table 2. The least squares planes and deviations (Å) of the atoms with e s d's in parenthesis. The equation of the plane is  $ax + by + cz + d = 0$ .

(i) Amide group (plane A)			
C(4) + 0.006 (2)	*N(1) - 0.065 (2)	a = 0.107	
C(5) - 0.019 (3)	*Cu - 0.447 (3)	b = 0.082	
N(2) + 0.007 (2)	*C(6) - 0.218 (3)	c = 0.991	
O(1) + 0.007 (2)		d = -2.446	
(ii) Carboxyl group (plane B)			
C(6) - 0.001 (2)	*N(2) + 0.120 (3)	a = 0.174	
C(7) + 0.003 (2)	*Cu - 0.056 (2)	b = -0.133	
O(2) - 0.001 (2)		c = 0.976	
O(3) - 0.001 (2)		d = -2.202	
(iii) Basal plane (plane C)			
N(1) - 0.066 (2)	*Cu - 0.122 (3)	a = 0.176	
N(2) + 0.076 (2)	*O(5)W - 2.708 (3)	b = -0.120	
O(2) - 0.074 (2)		c = 0.977	
O(4)W + 0.065 (2)		d = -2.274	
(iv) Pyrrolidine ring plane (plane D)			
C(3) + 0.025 (2)	*C(2) + 0.516 (4)	a = 0.916	
C(4) - 0.043 (2)		b = -0.230	
N(1) + 0.045 (2)		c = 0.326	
C(1) - 0.026 (2)		d = -1.351	
Angles between planes			
A—B 13.0 (5)	B—C 0.7 (5)	A—C 12.3 (5)	

\*Atom not included in plane calculation.

The bond lengths and angles of the ligand (figure 2) are quite normal and comparable to CULVAT and similar structures (Freeman 1966, 1967). The deviations of the N atoms, N(1),  $-0.065$  and N(2),  $0.120$  Å from the best planes *A* and *B* are comparable with values obtained in Cu-chelates of L-tyrosine (Tatsch and van der Helm 1969). The pyrrolidine ring is in an envelope form with C(2) (C<sup>γ</sup>) displaced by  $0.516$  Å (table 2). As mentioned earlier the C<sup>γ</sup> appears to be thermally disordered. On the other hand a statistical disorder of C<sup>γ</sup> is found in L-leucyl L-prolyl-L-glycine (Leung and Marsh 1958). The torsion angles listed in table 3 for the dipeptide are similar to those in CULVAT. The C<sup>α</sup>-C<sup>β</sup> bonds of both the proline and alanine residue are parallel to each other.

The possible hydrogen bonding in the structure are listed in table 4. The water molecule on the 2-fold axis acts both as donor and acceptor, while the water molecules in the coordination sphere of Cu act only as donors. The hydrogen bonding distances from the apical water,  $2.91$ ,  $3.09$  and  $3.22$  Å are rather large compared to those from water O(4),  $2.56$  and  $2.70$  Å. A similar behaviour of water molecules is found in Cu(gg)·2H<sub>2</sub>O (Kistenmacher and Szalda 1975).

It is interesting to closely examine these particular type of structures and find out whether one can arrive at some condition for the water molecule to coordinate with the metal Cu. Normally water molecules which act both as donors and acceptors in hydrogen bonding will link with the carboxyl O and carbonyl O atom and to a

Table 3. Torsion angles (e.s.d.'s in parenthesis)

Description	Value
N(1) C(4) C(5) N(2)	+ 5.0 (8)°
N(1) C(4) C(5) O(1)	- 179.0 (10)°
C(4) C(5) N(2) C(6)	- 170.1 (8)°
N(2) C(6) C(7) O(2)	- 5.1 (8)°
N(2) C(6) C(7) O(3)	+ 175.1 (10)°
C(5) N(2) C(6) C(7)	+ 161.2 (10)°
C(5) N(2) C(6) C(8)	- 87.2 (8)°
N(1) C(4) C(3) C(2)	- 13.9 (8)°
C(4) C(3) C(2) C(1)	+ 30.2 (10)°
C(3) C(2) C(1) N(1)	- 34.6 (8)°

Table 4. Intermolecular contacts—possible hydrogen bonds.

O(6).....O(4) <i>a</i>	2.70(2) Å
O(1).....O(4) <i>a</i>	2.56(2)
O(6).....O(3) <i>b</i>	2.62(2)
O(2).....O(5) <i>c</i>	3.09(2)
O(5).....O(1) <i>c</i>	3.22(2)
O(3).....O(5) <i>c</i>	2.91(2)

Key to symmetry operations.

- (a)  $x, 1 + y, z$   
 (b)  $-\frac{1}{2} + x, \frac{1}{2} + y, z$   
 (c)  $\frac{1}{2} + x, -\frac{1}{2} + y, -z$ .

lesser extent to the terminal NH<sub>2</sub> group and also to donors like — OH, etc. of the hydrophilic side chains. In bis (L-threonine) Cu·H<sub>2</sub>O, for example the one water present in the lattice is fully occupied as discussed above. In CULVAT, there are 4 water molecules, but none of them coordinate with Cu, since the hydrogen bonding scheme is already fully exhausted as discussed above. In CUPRAL, there are 2.5 water molecules, of which 2 are found in the coordination sphere. Here the dipeptide has only non-functional side chains, and as such the hydrogen bonding scheme is not exhausted at all so that the water molecules are forced to coordinate with Cu. The above argument is again borne out in the cases of Cu(ala)<sub>2</sub>·6H<sub>2</sub>O and Cu(gg)·3H<sub>2</sub>O where water molecules are found in the coordination sphere. (Freeman 1966). Hence it appears that the absence of hydrophilic side chain could be a deciding factor in pushing the water molecules into the coordination sphere of Cu.

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