

Metal Complexes of Some Peptide Derivatives: Part V—Complex Formation of Copper(II) with N-Salicyloyl Derivatives of Some Amino Acids

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Complex formation of Cu(II) with N-salicyloyl derivatives of some amino acids has been studied by equilibrium method at 25, 30, 35 and 45°C in 50% v/v aq. ethanol and at $\mu = 0.2 M$ (NaNO₃). Thermodynamic parameters associated with complexation equilibria have been correlated with the structure and bonding of the complexes.

Interactions of transition metal ions with amino acids and small peptide derivatives offer simple models for metal-protein interactions and enzymes¹⁻⁵. In neutral solution protonation and metallation with the amide (—CONH—) bond occur at the amide carbonyl oxygen atom. Metallation at the amide nitrogen occurs only through substitution of the amide proton provided there is a possibility of chelation of the metal ions^{2,5,6}. With a view to understanding the effect of an adjacent phenolic OH group on the coordinating properties of the amide bond we had earlier studied the complexation equilibria of Cu(II) with N-salicyloylglycine (SG)⁷, and N-salicyloyl *dl*- α -alanine (SA)⁸ at 30°C in 50% v/v aq. ethanol medium at a constant ionic strength ($\mu = 0.2 M$ NaNO₃). Complexation equilibria of Cu(II) with N-salicyloyl- β -alanine (SB) and N-salicyloyl-*dl*-methionine (SM), in the same solvent medium have been studied. Complexation equilibria of the Cu(II)-SG and Cu(II)-SA system have also been studied at 25°, 35° and 45°C; the data for these systems at 30°C were obtained from literature^{7,8}. Some solid complexes of Cu(II) have also been isolated and characterised on the basis of IR and UV-VIS spectral data.

Materials and Methods

All the ligands were prepared as described earlier^{7,8} and their purities were checked by elemental analyses, m.ps, UV and IR spectra. All the other reagents used were of AR quality.

Bright green crystalline Cu(II) complexes, Cu(HL-H).2H₂O, (where H₂L = SB and SM) were obtained by treating the monosodium salts (NaHL) of the respective ligands with Cu(II) sulphate in aqueous solution as described earlier^{7,8}. These complexes on acetylation furnished the monoacetyl derivatives, Cu(CH₃COL-H).2H₂O.

The procedure for the pH-metric study was the same as described earlier^{7,8}. Preliminary values of the equilibrium constants were evaluated by the procedure of Irving and Rossotti⁹ and the values were finally refined employing the computer programme¹⁰ SCOGS.

Results and Discussion

Characterisation of solid complexes

Cu(HL-H).2H₂O and Cu(CH₃COL-H).2H₂O
(H₂L = SB, SM)

The IR spectra of ligands in KBr exhibited ν COOH modes in the region 1740-1720 cm⁻¹. These modes were absent in the spectra of Cu(HL-H).2H₂O or Cu(CH₃COL-H).2H₂O. However, the spectra of the complexes displayed two new bands at 1540-1530 and 1370-1330 cm⁻¹ assigned to ν asCOO⁻ and ν sCOO⁻ respectively, suggesting Cu²⁺ — ⁻OOC bonding in the complexes¹¹. The amide I and II bands in these complexes were observed at 1600-1590 and 1450-1440 cm⁻¹ in comparison to those at 1660-1600 (br), 1550-1540 and 1300-1280 cm⁻¹ respectively due to amide I, II and III bands for the free ligands. Such shifts of the amide bands indicated bonding of deprotonated amide nitrogen^{3,4,12} to Cu(II) in these complexes. Both the series of complexes exhibited ν (O-H) mode in the region 3400-3300 cm⁻¹ which, however, could not be considered as a diagnostic for the free phenolic OH group, as similar bands were expected to appear due to H₂O molecules. Formation of Cu(CH₃COL-H).2H₂O through acetylation of Cu(HL-H).2H₂O by the procedure usually applied for acetylation of phenols indicated the presence of free phenolic OH group in the parent Cu(HL-H).2H₂O complexes. Analytical data of Cu(CH₃COL-H).2H₂O also supported substitution

Table 1—Proton-Ligand Constants at Different Temperatures and Associated Thermodynamic Parameters at 25°C

Ligand	Constant ^c	[Solvent = 50% (v/v) aq. ethanol; $\mu = 0.2 M(\text{NaNO}_3)$]				ΔG^a	ΔH^a	ΔS^b
		25°	30°	35°	45 ± 0.1°C			
SG	$pK_{\text{H}_2\text{L}}^{\text{H}}$	4.00	3.98 ⁷	3.96	3.91	−23.0	−8.4	49
	$pK_{\text{HL}}^{\text{H}}$	8.86	8.83 ⁷	8.79	8.74	−50.8	−12.8	128
SA	$pK_{\text{H}_2\text{L}}^{\text{H}}$	4.22	4.18 ⁸	4.15	4.10	−24.2	−9.6	49
	$pK_{\text{HL}}^{\text{H}}$	9.36	9.30 ⁸	9.25	9.16	−53.7	−12.8	137
SB	$pK_{\text{H}_2\text{L}}^{\text{H}}$	4.82	4.80	4.77	4.73	−27.6	−8.5	64
	$pK_{\text{HL}}^{\text{H}}$	9.02	8.98	8.96	8.90	−51.7	−13.0	130
SM	$pK_{\text{H}_2\text{L}}^{\text{H}}$	4.05	4.04	4.02	3.98	−23.2	−8.7	49
	$pK_{\text{HL}}^{\text{H}}$	8.78	8.75	8.72	8.66	−50.4	−19.2	105

(a): kJ mol^{-1} (b): $\text{JK}^{-1}\text{mol}^{-1}$; (c): error limits for constants, ± 0.05 in log scale; for ΔG , ΔH , $\pm 0.5 \text{ kJ mol}^{-1}$ and for ΔS , $\pm 3 \text{ JK}^{-1}\text{mol}^{-1}$.

of one hydrogen atom in $\text{Cu}(\text{HL-H})\cdot 2\text{H}_2\text{O}$, preferably phenolic proton by an acetyl group. This was supported by the fact that while $\text{Cu}(\text{HL-H})\cdot 2\text{H}_2\text{O}$ gave intense violet colour with neutralised FeCl_3 solution, the corresponding acetyl derivatives did not give any colour with this reagent.

Electronic spectra of both $\text{Cu}(\text{HL-H})\cdot 2\text{H}_2\text{O}$ and $\text{Cu}(\text{CH}_3\text{COL-H})\cdot 2\text{H}_2\text{O}$ were almost superimposable and maximum absorption in the region 610–600 nm were indicative of coordination of $\text{Cu}(\text{II})$ by deprotonated amide nitrogen and carboxylate oxygen atoms in a square planar $\text{Cu}(\text{N},\text{O})(\text{H}_2\text{O})_2$ geometry without having any apical coordination^{5,13} as might be provided by the phenolic oxygen atom. As the ligand ion, $(\text{CH}_3\text{COL-H})^{2-}$ could provide only bidentate (N,O) chelation via the deprotonated amide nitrogen and the carboxylate oxygen atoms, it may, therefore, be inferred that in $\text{Cu}(\text{HL-H})\cdot 2\text{H}_2\text{O}$ the ligand $(\text{HL-H})^{2-}$ is (N,O) bidentate leaving the phenolic OH intact. The possibility of coordination of SCH_3 moiety of SM ligand was also ruled out because the electronic spectra of $\text{Cu}(\text{II})$ complexes of SM were superimposable with those of SG, SA and SB. Room temperature magnetic moment values of all the $\text{Cu}(\text{II})$ complexes were in the range 1.76–1.84 B.M. as expected for one unpaired electron and indicated the absence of any $\text{Cu}\cdots\text{Cu}$ interaction in these complexes.

Proton-ligand and $\text{Cu}(\text{II})$ -ligand equilibria

The ligands SB and SM like SG and SA could also be titrated as biprotic acids (H_2L), deprotonation occurring from $-\text{COOH}$ and phenolic $-\text{OH}$ groups in successive steps giving two well separated buffer regions in the pH ranges 2.5–6.5 and 8–12.

That the phenolic OH and not the amide ($-\text{CONH}-$) group was deprotonated in the second step, was confirmed by the monoprotic nature of benzoylglycine ($\text{C}_6\text{H}_5\text{CONHCH}_2\text{COOH}$), which gave a single buffer region (pH 2.5–6.5) in the entire pH range (2–12) studied. Increase of $K_{\text{COOH}}^{\text{H}}$ and K_{OH}^{H} values (Table 1) with increase in temperature indicated that COOH and OH are involved in H-bonding in the ligands. The H-bonding breaks down at higher temperature thus accounting for enhanced acidity. Positive entropy changes (ΔS) associated with proton-ligand equilibria indicate increased flexibility of the deprotonated forms of the ligands due to the absence of H-bonding. Enthalpy changes (ΔH) accompanying deprotonation of phenolic OH are found to be roughly 1.5 times higher than the corresponding values for the deprotonation of $-\text{COOH}$, probably due to the labilizing influence of $\text{C}=\text{O}$ moiety of $-\text{COOH}$ function on $\text{O}-\text{H}$ bond. Higher acidities (K_{OH}^{H}) of phenolic OH of the ligands over that of salicylic acid may be due to stabilization of the L^{2-} anion by H-bonding involving amide proton.

Buffer regions (pH 2.5–6.5) are observed⁷ at lower pH values in the presence of $\text{Cu}(\text{II})$ while the regions pH 8–12 do not show any change. Complexation with $\text{Cu}(\text{II})$, in solution, therefore, occurs only via deprotonation of $-\text{COOH}$. Four protons per $\text{Cu}(\text{II})$ are released in the pH range 2.5–6.5, where hydrolysis of the Cu^{2+} (aq.) ion does not occur. Mole ratio study of $\text{Cu}(\text{II})$ - H_2L mixtures at pH values corresponding to $\bar{n} \geq 4$, by spectrophotometric method

⁷ $\bar{n}$: Average number of protons released due to the reaction of $\text{Cu}(\text{II})$ with the ligands, H_2L .

Table 2—Formation Constants^a of Cu(II) Complexes at Different Temperatures[Solvent = 50% (v/v) aq. ethanol; $\mu = 0.2 M$ (NaNO₃)]

System	Temp. ($\pm 0.1^\circ\text{C}$)	$pK_{\text{Cu} + \text{H}_2\text{L}}^{2\text{H}}$	$pK_{\text{Cu}(\text{HL-H}) + \text{H}_2\text{L}}^{2\text{H}}$	$pK_{\text{Cu}(\text{HL-H})_2}^{\text{H}}$	$pK_{\text{Cu}(\text{L-H})(\text{HL-H})}^{\text{H}}$
Cu(II) + SG	25	6.15	6.40	8.54	10.02
	30 (ref. 7)	6.06	6.32	8.43	9.98
	35	5.97	6.24	8.32	9.94
	45	5.81	6.10	8.14	9.82
Cu(II) + SA	25	6.52	6.90	8.77	10.53
	30 (ref. 8)	6.42	6.79	8.66	10.49
	35	6.33	6.68	8.55	10.45
	45	6.18	6.51	8.39	10.34
Cu(II) + SB	25	7.96	9.14	<i>b</i>	<i>b</i>
	30	7.87	9.03	<i>b</i>	<i>b</i>
	35	7.80	8.92	<i>b</i>	<i>b</i>
	45	7.66	8.73	<i>b</i>	<i>b</i>
Cu(II) + SM	25	6.39	6.59	8.40	10.35
	30	6.34	6.54	8.35	10.32
	35	6.29	6.49	8.31	10.29
	45	6.18	6.38	8.23	10.23

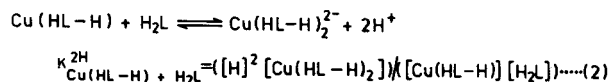
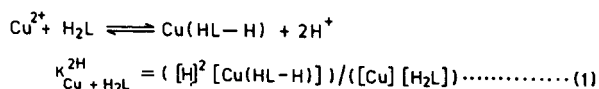
(a) Error limits for constants, ± 0.05 in log scale; (b) constants could not be determined due to precipitation.Table 3—Thermodynamic Parameters^a Associated with Formation of Cu(II) Complexes at $25 \pm 0.1^\circ\text{C}$ [Solvent = 50% (v/v) aq. ethanol, $\mu = 0.2 M$ (NaNO₃)]

Parameter	Cu(II)-SG	Cu(II)-SA	Cu(II)-SB	Cu(II)-SM
$\text{Cu}^{2+} + \text{L}^{2-} \rightleftharpoons \text{Cu}(\text{HL-H})$				
$\log K_{\text{Cu}(\text{HL-H})}^{\text{Cu}}$	6.71	7.05	5.88	6.44
$\Delta G (\text{kJmol}^{-1})$	-38.5	-37.0	-33.7	-36.9
$\Delta H (\text{kJmol}^{-1})$	12.8	3.0	9.6	2.0
$\Delta S (\text{JK}^{-1}\text{mol}^{-1})$	172	146	145	131
$\text{Cu}(\text{HL-H}) + \text{L}^{2-} \rightleftharpoons \text{Cu}(\text{HL-H})_2^{2-}$				
$\log K_{\text{Cu}(\text{HL-H})_2}^{\text{Cu}(\text{HL-H})}$	6.46	6.67	4.70	6.24
$\Delta G (\text{kJmol}^{-1})$	-37.0	-40.4	-27.0	-35.8
$\Delta H (\text{kJmol}^{-1})$	8.0	8.8	9.5	2.0
$\Delta S (\text{JK}^{-1}\text{mol}^{-1})$	151	158	123	127
$\text{Cu}(\text{HL-H})_2^{2-} \rightleftharpoons \text{Cu}(\text{L-H})(\text{HL-H})^{3-} + \text{H}^+$				
$pK_{\text{Cu}(\text{HL-H})_2}^{\text{H}}$	8.54	8.77	<i>b</i>	8.40
$\Delta G (\text{kJmol}^{-1})$	-49.0	-50.3	<i>b</i>	-48.2
$\Delta H (\text{kJmol}^{-1})$	-38.5	-36.6	<i>b</i>	-16.5
$\Delta S (\text{JK}^{-1}\text{mol}^{-1})$	35(40)	46(49)	<i>b</i>	106(49)
$\text{Cu}(\text{L-H})(\text{HL-H})^{3-} \rightleftharpoons \text{Cu}(\text{L-H})_2^{4-} + \text{H}^+$				
$pK_{\text{Cu}(\text{L-H})(\text{HL-H})}^{\text{H}}$	10.02	10.53	<i>b</i>	10.35
$\Delta G (\text{kJmol}^{-1})$	-57.4	-60.4	<i>b</i>	-59.3
$\Delta H (\text{kJmol}^{-1})$	-19.2	-18.3	<i>b</i>	-11.5
$\Delta S (\text{JK}^{-1}\text{mol}^{-1})$	128(128)	141(137)	<i>b</i>	160(105)

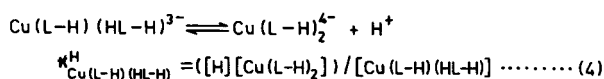
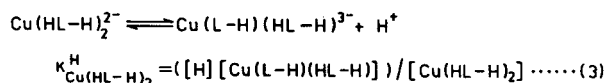
(a) Error limits: for the constants ± 0.05 in the log scale; for ΔG and ΔH , $\pm 0.5 \text{ kJmol}^{-1}$; and for ΔS , $\pm 3 \text{ JK}^{-1}\text{mol}^{-1}$. (b) Could not be determined due to precipitation. Values in the parentheses are for the deprotonation equilibria of the free ligand.

at 610-600 nm established the 1:2 (Cu:ligand) stoichiometry. Therefore, hydroxo complexes and complexes higher than 1:2 were omitted in calculating the Cu(II)-ligand constants. Formation of Cu(II)-

$2\text{H}_2\text{L}$ complexes can, therefore, be accounted for by assuming the substitution of $-\text{COOH}$ and amide ($-\text{CONH}-$) protons of the ligands by Cu(II) (Eqs 1 and 2):



The higher pH buffer region (pH 8-12) arose because of the deprotonation of phenolic OH of the free ligand, HL⁻ ions, together with the deprotonation of uncoordinated phenolic OH in Cu(HL-H)₂²⁻ complexes (Eqs 3 and 4):



Charges were dropped from these expressions for clarity. The (HL-H)₂²⁻ ions in the complexes stand for the ligand species which have lost their carboxylic (-COOH) and amide (-CONH-) protons while still retaining the phenolic OH protons and the (L-H)³⁻ ions stand for the fully deprotonated ligand species^{5,13}. Equilibria (3) and (4) could not be followed for Cu(II)-SB system due to precipitation at pH ≥ 9.5. Equilibrium constants for equilibria (1-4) are presented in Table 2.

Stability constants $K_{\text{Cu}(\text{HL}-\text{H})}^{\text{Cu}}$ and $K_{\text{Cu}(\text{HL}-\text{H})_2}^{\text{Cu}(\text{HL}-\text{H})}$ (Table 3) were calculated with the help of the relations (5) and (6) respectively.

$$\log K_{\text{Cu}(\text{HL}-\text{H})}^{\text{Cu}} = \log K_{\text{Cu}+\text{H}_2\text{L}}^{2\text{H}} + pK_{\text{COOH}}^{\text{H}} + pK_{\text{OH}}^{\text{H}} \dots\dots\dots (5)$$

$$\log K_{\text{Cu}(\text{HL}-\text{H})_2}^{\text{Cu}(\text{HL}-\text{H})} = \log K_{\text{Cu}(\text{HL}-\text{H})+\text{H}_2\text{L}}^{2\text{H}} + pK_{\text{COOH}}^{\text{H}} + pK_{\text{OH}}^{\text{H}} \dots\dots\dots (6)$$

Formation of Cu(HL-H) and Cu(HL-H)₂²⁻ complexes through metal-promoted deprotonation of the amide bond was accompanied by positive enthalpy change (Table 3). Positive entropy changes accompanying the formation of Cu(HL-H) and Cu(HL-H)₂²⁻ complexes suggested their chelated structures. The $\bar{n}(\text{pH})$ plot corresponding to the two series of equilibria, (1), (2) and (3), (4) showed a sharp increase from $\bar{n}=0$ to $\bar{n}=4$ (pH 2.5-6.5) and a flat rise in the region from $\bar{n}=4$ to $\bar{n}=6$ (pH 8-12). Absorption maxima λ_{max} of 1:5 Cu(II):H₂L mixtures at pH values corresponding to $\bar{n}=0, 2, 4$ and 6 were 730, 650, 620-610 and 620-610 nm respectively for all the systems. This blue shift of 90 nm in the ab-

sorption maxima indicated a change from an octahedral Cu(O)₆ system in Cu²⁺ (aq.) ion to a square planar Cu(N,O)₂ system in Cu(HL-H)₂²⁻ complexes. The constancy of λ_{max} in going from $\bar{n}=4$ to $\bar{n}=6$ indicates the retention of square planar Cu(N,O)₂ geometry even in the fully deprotonated Cu(L-H)₂⁴⁻ complexes. The observed λ_{max} values 620-610 nm for the Cu(HL-H)₂²⁻ and Cu(L-H)₂⁴⁻ complexes are quite close within experimental errors, to the calculated values⁵ (605 nm) for square planar Cu(N,O)₂ systems, where Cu(II) is coordinated to two deprotonated amide nitrogen and two carboxylate oxygen atoms. This observation further rules out the possibility of apical coordination of Cu(II) by phenolic oxygen in the Cu(L-H)(HL-H)³⁻ and Cu(L-H)₂⁴⁻ complexes. However, the possibility of chelation *via* hydrogen bonding of the phenolic proton with the amide carbonyl oxygen in the Cu(HL-H)₂²⁻ complexes cannot be ruled out. Comparable magnitudes of $pK_{\text{Cu}(\text{HL}-\text{H})}^{\text{H}}$ and $pK_{\text{HL}}^{\text{H}}$ values for all these systems indicate deprotonation equilibria (3) and (4) of Cu(HL-H)₂²⁻ complexes to be analogous to deprotonation of the phenolic moieties of the free ligands (HL⁻) species. With the exception of Cu(II)-SM system, even the entropy changes (ΔS) corresponding to deprotonation of Cu(HL-H)₂²⁻ complexes (Eqs 3 and 4) are comparable (Tables 1 and 3) with those of the free phenolic OH of the free ligand ion (HL⁻). Positive ΔS values for these equilibria are the result of breakdown of H-bonding in uncoordinated phenolic OH with C=O of the amide group.

Comparable magnitudes of formation constants (Tables 2 and 3) and identical λ_{max} values of Cu(II) complexes derived from SG, SA and SB with those derived from SM which has an extra binding site, viz. the SCH₃ moiety, suggest similar type of coordination, viz. (N,O) bidentate chelation by all the four ligands. The SCH₃ moiety of SM was not probably associated to Cu(II) in the complexes. Comparatively higher entropy change associated with deprotonation equilibria (3) and (4) of Cu(II)-SM system may be due to release of some sort of a hydrophobic strain involving -CH₂CH₂SCH₃ moiety. Stabilities of Cu(II) complexes follow the ligand order: SA > SG > SM > SB. The lowest stabilities of Cu(II)-SB complexes are probably the result of angle strain in the six-membered chelate rings without having a double bond¹⁴ as against five-membered chelate rings in the complexes derived from the other three ligands. Electron releasing effect of CH₃ group at α -position to amide and COOH moieties in SA ligand provides additional basicity to the donor atoms and consequently Cu(II) complexes with SA ligands are more stable than those with SG. Similar trends in stability have also been observed for Cu(II)

complexes with glycylglycinate¹⁵ and glycyl-*dl*- α -alaninate¹⁶. The bidentate (N,O) chelated square planar $\text{Cu}(\text{HL-H})_2^{2-}$ complexes are predominant ($\approx 95\%$) in $\text{Cu}(\text{II})$ containing species in the region of biological pH.

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