Interligand Interaction in Mixed Amino Acid Complexes

Vadaparti Manjula and Pabitra K. Bhattacharya
Department of Chemistry, Faculty of Science, M.S. University of Baroda, Baroda 290 002, India

The formation constants of the ternary complexes [CuL(L')], where L and L' are tryptophan (Try), tyrosine (Tyr), phenylalanine (Phe), or 3,4-dihydroxyphenylalanine (L-dopa), have been determined potentiometrically in aqueous media at 25 °C and I = 0.2 mol dm^{-3} using the SCOGS computer program. The ternary complexes are found to be stable due to intramolecular interligand interactions between the aromatic side groups of the amino acids.

Metalloenzymes, involving metal ion, enzyme, and substrate, are essentially ternary complexes. Investigations of the stability of ternary complexes may therefore help towards understanding the driving forces which lead to the formation of such complexes in biological systems. The synthetic models cannot, however, compete in efficiency, since, in metalloenzymes, there are intramolecular interactions between the non-co-ordinated groups in the enzyme and the substrate, which are co-ordinated to the metal ion. Hence a study of intramolecular interactions in model ternary complexes has invited the attention of many chemists. The interactions can be of two types.[1-3] A rigid interaction between a co-ordinated aromatic diamine and the non-co-ordinated base of a nucleotide, as in [Cu–bipy–ATP][4,5] (bipy = 2,2'-bipyridine ATP = adenosine triphosphate), or a flexible interaction between the non-co-ordinated side group of an amino acid and the base of ATP or non-co-ordinated side group of another amino acid.

In the complex [Cu–ATP–Try][6] (Try = tryptophan) an aromatic ring-stacking interaction has been observed between the purine moiety of ATP and the indole of tryptophan. In binary complexes of amino acids with side groups such as tyrosine, tryptophan, and phenylalanine, it is observed that log K_2 is higher than expected from statistical and electrostatistical considerations.[3] This increase in stability is attributed to an intramolecular ring-stacking interaction. In mixed amino acid ternary complexes such as copper–phenylalanine–norvalinate[7] a flexible hydrophobic interaction has been shown. Interligand interaction has also been proposed in [Cu–Phe–Tyr][1] (Phe = phenylalanine, Tyr = tyrosine) complexes due to non-co-ordinated side groups.

In the present paper this hydrophobic interligand interaction and its effect on the stability of the ternary complex has been studied using the ternary complexes [CuL(L')], where L = L' = tryptophan (Try), tyrosine (Tyr), or phenylalanine (Phe), in aqueous media. The study has been further extended to the case of ternary complexes involving 3,4-dihydroxyphenylalanine (L-dopa). The latter is important as a neurotransmitter[8] in biochemical processes and is used for therapeutic purposes in cases of Parkinson's disease.[9-11] It is also interesting because of its ambidentate nature,[8,12,13] and so it was of interest to study mixed amino acid complexes involving L-dopa.

Experimental

All the reagents were of AnalaR grade and the titrations were carried out in aqueous media using an Orion ionalyzer/901 with an accuracy of pH of ±0.001. The temperature was maintained at 25 °C using a thermostatically controlled water bath.

Formation constants for the ternary complexes were determined by titrations of aqueous solutions (50 cm^3) of the reactants (metal and ligand concentrations of 0.002 and 0.004 mol dm^{-3}, respectively) in the ratios Cu^{2+}:L:L' = 1:1:1 and 1:2:2, against carbonate-free standard sodium hydroxide, and using the computer program SCOGS.[14] Each set of titrations was repeated to check the reproducibility of the data.

Titrations of acid + ligand and acid + Cu + ligand (with ratios of metal and ligand concentrations of 1:2 and 1:3) were carried out and the data used for computer calculation of the proton–ligand formation constants for the ligands and metal–ligand formation constants for [CuL] and [CuL_2]. These refined values were used as constants in computer calculations of the formation constants for [CuL(L')]. The values of the proton–ligand formation constants and binary complex formation constants are presented in Table 1. Values of the formation constants of the ternary complexes are presented in Table 2. The values obtained using different concentrations and different ratios of metal ion and ligands agree within ±0.01, which is the accuracy of the values reported.

Results and Discussion

The stability of ternary complexes can be quantified by calculating the values of Δ log K.[15] Δ log K values in the present study have an accuracy of ±0.02.

$$\begin{aligned}\Delta \log K &= \log K^{ML}_{ML(L')} - \log K^{M}_{ML'} \\ &= \log K^{ML'}_{ML(L')} - \log K^{M}_{ML} \\ &= \log K^{M}_{ML(L')} - \log K^{M}_{ML} - \log K^{M}_{ML'}\end{aligned}$$

In the case of [CuL(L')] complexes where L = alanine and L' = phenylalanine, Δ log K was found to be negative.[16] This is because of the electrostatic repulsion between the alanine monoanion and the phenylalanine monoanion. However, it is observed that in the present complexes, of the type [CuL(L')], where L and L' are phenylalanine, tryptophan, and tyrosine, Δ log K is less negative. This is because the amino acids are co-ordinating from the amino carboxylate end and there is a hydrophobic interaction with the free phenyl, indole, or hydroxyphenyl group. Δ log K is less negative in [Cu–Try–Tyr] than in [Cu–Try–Phe]. This may be because of hydrogen bonding between the indole moiety of tryptophan and the hydroxyphenyl OH of tyrosine.

The ligand L-dopa is ambidentate in nature. In the case of Cu^{II} complexes it has been observed that at low pH (pH 2—5) dopa co-ordinates from the amino carboxylate end and above pH 6 both amino carboxylate and pyrocatecholate are involved in binding with Cu^{II}, resulting in polymeric species. In the ternary complexes it is also observed that on titration of a 1:1:1 mixture of Cu + amino acid + L-dopa with sodium hydroxide a buffer region is observed up to pH 5, corresponding to two moles of added alkali, indicating amino acid co-ordination for both amino acids. However, at a higher pH (6.5) a second buffer region starts, showing that L-dopa co-ordination changes to the catechol site, liberating two protons. However, computer calculations considering both types of co-ordination of L-dopa

Table 1. Proton–ligand formation constants of tryptophan (Try), tyrosine (Tyr), phenylalanine (Phe), and 3,4-dihydroxyphenylalanine (L-dopa), and binary complex formation constants of copper(II), with standard deviation (σ) in parentheses

Ligand	K_1^H	K_2^H	Copper(II) complexes: $\log K_{CuL}^{Cu}$	$\log K_{CuL_2}^{CuL}$
Tryptophan (Try)	9.44	1.99	7.98	7.38
	(0.07)	(0.01)	(0.06)	(0.05)
Tyrosine (Tyr)	9.07	1.89	7.20	6.81
	(0.01)	(0.01)	(0.04)	(0.04)
Phenylalanine (Phe)	9.21	1.92	7.49	6.65
	(0.01)	(0.02)	(0.05)	(0.04)
3,4-Dihydroxyphenylalanine (L-dopa)	8.60	2.20	6.94	5.65
	(0.01)	(0.01)	(0.04)	(0.04)

Table 2. Mixed-ligand stability constants [$\log K(CuLL')$] and $\Delta \log K$ in aqueous medium and $I = 0.2$ mol dm^{-3} $NaClO_4$ at 25 °C, with standard deviations (σ) in parentheses

	$\log K_{Cu}^{CuL(L')}$	$\Delta \log K$
[Cu–Try–Tyr]	14.29	−0.89
	(0.001)	
[Cu–Try–Phe]	14.49	−0.98
	(0.001)	
[Cu–L-dopa–Try]	15.35	+0.45
	(0.008)	
[Cu–L-dopa–Tyr]	14.42	+0.28
	(0.001)	
[Cu–L-dopa–Phe]	14.29	−0.24
	(0.002)	

do not give refinement of these results. Hence the calculations have been confined to pH 5, considering only the species with amino acid co-ordination.

It is observed that $\Delta \log K$ in the case of L-dopa-containing complexes is less negative, or more positive, than in the case of other mixed amino acid complexes. $\Delta \log K$ becomes more positive in the order [Cu–L-dopa–Phe] < [Cu–L-dopa–Tyr] < [Cu–L-dopa–Try]. This can be explained as being due to intramolecular hydrogen bonding between the non-co-ordinating groups. The hydrogen bonding between the indole moiety of tryptophan and the dihydroxyphenyl of L-dopa is more than that between the hydroxyphenyl of tyrosine and dihydroxyphenyl of L-dopa.

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References

1 H. Sigel, H. Tribolet, and K. H. Scheller, *Inorg. Chim. Acta*, 1985, **100**, 151.
2 R. Frieden, *J. Chem. Educ.*, 1973, **52**, 754.
3 A. Odani and O. Yamuchi, *Inorg. Chim. Acta*, 1984, **93**, 13.
4 C. F. Naumann, B. Prijs, and H. Sigel, *Eur. J. Biochem.*, 1974, **41**, 209.
5 P. Chaudhari and H. Sigel, *J. Am. Chem. Soc.*, 1974, **96**, 2750.
6 J. B. Orenberg, B. E. Fischer, and H. Sigel, *J. Inorg. Nucl. Chem.*, 1980, **48**, 785.
7 A. Gergely, I. Sovago, I. Nagypal, and R. Kiraly, *Inorg. Chim. Acta*, 1972, **6**, 435.
8 K. S. Rajan, S. Mainer, and J. M. Davis, *Bioinorg. Chem.*, 1978, **9**, 187.
9 K. S. Rajan, S. Mainer, and J. M. Davis, *J. Inorg. Nucl. Chem.*, 1975, **9**, 289.
10 T. Kiss and A. Gergely, *Inorg. Chim. Acta*, 1983, **78**, 247.
11 J. B. Summer and K. Myback, 'The Enzymes,' Academic Press, New York, 1951, vol. 2, part 1, p. 489.
12 K. S. Rajan, A. A. Manian, J. M. Davis, and H. Dekimejian, *Brain Res.*, 1976, **107**, 31.
13 R. K. Bogges and R. B. Martin, *J. Am. Chem. Soc.*, 1975, **97**, 3076.
14 (*a*) J. G. Saycee, *Talanta*, 1968, **15**, 1397; (*b*) *ibid.*, 1971, **18**, 653; (*c*) J. G. Saycee and V. S. Sharna, *ibid.*, 1972, **19**, 831.
15 H. Sigel, *Chimia*, 1967, **21**, 489.
16 B. E. Fischer and H. Sigel, *J. Am. Chem. Soc.*, 1980, **102**, 299.

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