

Equilibria and kinetics for *pH*-dependent axial ligation of alkyl(aquo) cobaloximes with aromatic and aliphatic N-donor ligands

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Abstract. Equilibria and kinetics of the reaction of bromomethyl(aquo) cobaloxime with histamine, histidine, glycine and ethyl glycine ester and iodomethyl(aquo) cobaloxime with cyanide, imidazole and substituted imidazoles were studied as a function of *pH* at 25°C, 1.0 M ionic strength (KCl) by spectrophotometry technique. The rate of substitution of H₂O varies with the *pKa* of the incoming ligand, thus establishing the existence of nucleophilic participation of the ligand in the transition state. Dissociation kinetic reactions were also studied as a function of *pH*. Binding and kinetic data were interpreted based on the basicity, steric crowd of the entering ligand and HSAB principle. To compare the rate constants of the entering ligands *pH* independent second-order rate constants were calculated.

Keywords. Alkylcobaloximes; histamine; histidine; imidazoles; equilibrium constants.

1. Introduction

The key step in the mechanism of action of many enzymes, which require Vit-B₁₂ coenzyme, is generally accepted as the homolytic cleavage of the Co–C bond^{1–3}. It is widely believed that structural and conformational changes in coenzyme B₁₂ lead to acceleration in Co–C bond cleavage rates^{4–6}. The axial ligation reactions of metallo-porphyrin ions in aqueous solution are dependent upon the particular metal ion^{7–12}, equatorial ligands¹³ and the axial ligands^{14–18}. The study of simple models of the B₁₂ coenzyme, such as the cobaloximes, RCo(DH)₂L, where L = neutral ligand and R = alkyl group, has furnished a significant amount of data^{19,20} that have provided a foundation for understanding the behaviour of cobalamins²¹. These cobaloximes have been the subject of extensive kinetic and mechanistic studies^{22,23}. This activity has been motivated by the possibility that axial base release may be involved in biological mechanisms. Contrary to this, as models for coenzyme B₁₂, cobaloximes can be faulted on a number of counts including electrochemical²⁴, kinetic^{25,26} and structural properties^{20,27,28}. The (DH)₂ equatorial ligand system is not as electron donating as the corrin in coenzyme B₁₂ or the

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schiff-base equatorial ligands of other B₁₂ models²⁹. Compared to both cobalamins and other model systems, cobaloximes have stronger Co–C bonds²⁹ and shorter Co–L (L = pyridine or substituted pyridines) bonds²⁰. Eldik *et al*³⁰ studied the ligand substitution reactions of *trans*-[Co(en)₂Me(H₂O)]²⁺ a simple model for coenzyme B₁₂, with cyanide and imidazole as entering ligands and found that these ligands displace the coordinated water molecule *trans* to the methyl group and form the six coordinate complex. There is a need to study ligand substitution reactions *trans* to the axial alkyl ligand in coenzyme B₁₂ and various model complexes. Since it is known that methyl cobaloximes and coenzyme B₁₂ undergo substitution of their axial benzimidazole ligand with a protein histidine residues during complexation to the enzyme methionine synthase and methyl malonyl coenzyme A mutase, respectively^{31,32}.

Since binding of cobaloximes with amino acids, imidazoles and histamine are more closely related to the structural and bonding characteristics of corrin systems involved in biological mechanisms, we decided to explore the kinetics and equilibria of the axial ligation of the alkyl(aquo)cobaloximes with the aromatic ligands imidazole, substituted imidazoles, histamine, histidine and aliphatic ligands (glycine, ethyl glycine ester).

2. Materials and methods

Histamine (histamine dihydrochloride), histidine (histidine monohydrochloride), glycine, ethyl glycine ester were obtained from Sigma and imidazole, 1-methyl imidazole, 2-methyl imidazole, 2-ethyl imidazole, 1,2-di methyl imidazole were obtained from Acros. KCl, HPLC grade methanol, acetic acid, HCl, phosphoric acid, formic acid were obtained from Fluka. Dipotassium hydrogen phosphate, potassium dihydrogen phosphate, potassium phosphate, *tris*(hydroxymethyl)aminomethane (Tris), sodium acetate, potassium hydroxide were obtained from Acros. Double-distilled, deionized water was used throughout.

To maintain appropriate pH 0.2M buffers of HCl (0–1.5 pH), KH₂PO₄ and H₃PO₄ (2.0 pH), HCOOH and KOH (2.5–3.0 pH), CH₃COOH and CH₃COONa (3.5–5.5 pH), K₂HPO₄ and KH₂PO₄ (6.0–8.0 pH), Tris and HCl (8.5–9.0 pH), K₂HPO₄ and K₃PO₄ (9.5–11.5 pH) were used.

Alkyl(aquo) cobaloximes were prepared by modified procedure of Brown *et al*³³. All manipulations were performed under minimal illuminations due to photolability of the carbon–cobalt bond¹⁴. These alkyl(aquo) cobaloximes are photolabile, particularly in solution. Soluble in alcohols and DMSO, less so in chloroform or water and virtually insoluble in ether and hydrocarbon solvents.

pH values were determined with a Digisun digital pH meter equipped with a combined glass electrode. The electrode was standardized at two pH values (pH = 4 and 9.2) with standard buffer solutions. UV and visible spectra were recorded on a Hitachi U-3410, the sample compartment of which is provided with a thermostat and the concentrations of bromomethyl(aquo) cobaloximes (0.00125 M) was fixed at 436 nm and iodomethyl(aquo) cobaloximes (0.001 M) was fixed at 442 nm. For axial ligation single wavelength measurements were made on an Elico single beam spectrophotometer SL 171 model. The sample compartment of which was thermostated at 25 ± 0.1°C.

3. Results and discussion

3.1 Determination of dissociation constants of the ligands

Values for the pK_a of the conjugate acid of ligands are obtained by potentiometric titration at $25 \pm 0.1^\circ\text{C}$. Values of pK_a 's are obtained by a linear least-squares fit of the data to (1) below, derived from (2), where \mathbf{a}_L is the fraction of the total ligand present as the free base (or unprotonated) species as shown in (3).

$$pH = pK_a + \log [(\mathbf{a}_L)/(1-\mathbf{a}_L)], \quad (1)$$

$$K_a = [L^-] [H^+]/[HL], \quad (2)$$

$$\mathbf{a}_L = K_a/(K_a + [H^+]). \quad (3)$$

K_a is the dissociation constant of the ligand.

3.2 Determination of equilibrium constants

Apparent equilibrium constants (K_{app} values, see (4) below) for the axial ligation of alkyl(aquo) cobaloximes (scheme 1) were determined by spectrophotometric measurements. Solutions containing $\text{RCo}(\text{DH})_2(\text{OH}_2)$, an appropriate buffer (0.2 M) to maintain pH, KCl to maintain ionic strength (1.0 M) and varying concentrations of ligand are taken in 3 mm cuvettes and allowed to equilibrate in a thermostated holder at $25 \pm 0.1^\circ\text{C}$ for 15 min prior to addition of cobaloxime.

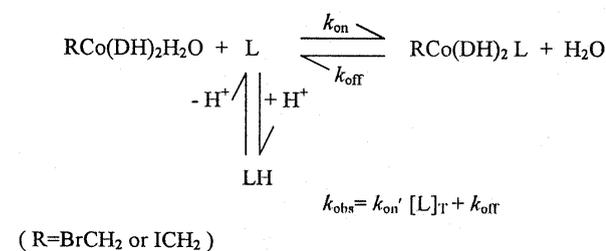
$$K_{app} = \frac{[\text{RCo}(\text{DH})_2\text{L}]}{[\text{RCo}(\text{DH})_2\text{H}_2\text{O}][\text{L}]_{free}}. \quad (4)$$

Final absorbance readings are taken after equilibrium is established as indicated by the time independence of the readings.

For such experimental setups, at a given pH, (5) is applied as follows

$$\Delta A = \Delta A_{max} [L]_f / (1/K_{app} + [L]_f), \quad (5)$$

where ΔA is the difference in absorbance between solutions containing cobaloxime and added ligand (L) and solutions containing only cobaloxime at the same concentration, ΔA_{max} is the maximum absorbance change thus obtained at high $[L]$, and $[L]_f$ is the



Scheme 1.

equilibrium concentration of the ligand in both ionization states. The data are analysed by a least-squares fit to the rearranged form of (5) to give

$$\Delta A = \Delta A_{\max} - \{1/K_{\text{app}} (\Delta A/[L]_f)\}, \quad (6)$$

$$[L]_f = [L]_{T-} - (C_T \Delta A/\Delta A_{\max}). \quad (7)$$

$[L]_f$ is calculated from (7) using the measured value of ΔA_{\max} , $[L]_{T-}$ is the total concentration of added ligand and C_T is the total concentration of cobaloxime. Values of K_{app} are obtained from the least-squares fit of (6) i.e., the plot of ΔA vs $\Delta A/[L]_f$ and the slope is $-1/K_{\text{app}}$.

The values for the equilibrium constants for axial ligation with respect to unprotonated ligand are calculated from the relation $K_{\text{eq}} = K_{\text{app}}/\mathbf{a}_L$, where \mathbf{a}_L is calculated from (3).

3.3 Determination of ligation rates (k_{on})

For each ligand L , at various pH values, first-order rate constants (k_{obs}) are determined from the absorbance measurements at the same wavelength used for K_{app} determinations under pseudo-first order condition with L being, at least in 10-fold excess over cobaloxime concentration.

Reaction progress is monitored by measurements of the change in the absorbance upon addition of alkyl(aquo) cobaloxime to a 3 ml cuvette, which contain KCl to maintain unit ionic strength, necessary buffer (0.2 M) to maintain pH and ligand in the thermostated ($25 \pm 0.1^\circ\text{C}$) cell compartment of Elico SL171 model. First order rate constants (k_{obs}) are obtained by least-squares fits of the data to (8) below

$$\ln(A_t - A_{\infty}) = k_{\text{obs}}t, \quad (8)$$

where A_t is the absorbance at time t and A_{∞} is the final absorbance.

Second-order rate constants, k_{on} , at a given pH for a given ligand are obtained from the slopes of least-squares fits of the data,

$$k_{\text{obs}} = k_{\text{on}}' [L]_{T-} + k_{\text{off}}, \quad (9)$$

where $[L]_{T-}$ is the total concentration of L present. Values of k_{on} , the pH independent second-order ligation rate constant are calculated from $k_{\text{on}} = k_{\text{on}}'/\mathbf{a}_L$, where \mathbf{a}_L is defined above.

3.4 Determination of k_{off}

Ligand dissociation rate constants, k_{off} (scheme 1), are measured spectrophotometrically by addition of a small volume of a solution containing preformed $\text{RCo}(\text{DH})_2\text{L}$ to cuvettes containing KCl buffer (0.2 M) in the thermostated ($25 \pm 0.1^\circ\text{C}$) cell compartment of the spectrophotometer.

Absorbance is continuously monitored at the same wavelength (436 nm or 442 nm) used for K_{app} and k_{obs} measurements. Triplicate measurements are made at each pH and first-order rate constants, k_{off} , are determined as above (8). In all cases, the ligand dissociation proceeds to $\geq 99\%$ completion at both pH s. All plots of (8) are satisfactorily

linear (correlation coefficients ≥ 0.998). All determinations were averaged to obtain a final value of k_{off} .

Imidazole, substituted imidazoles, histamine, histidine, glycine, ethyl glycine ester undergo protonation of N-atom with acid dissociation constants, pK_a in the range of 6–10. The values of the equilibrium constant K_{app} for the reaction of the glycine, ethyl glycine ester, histidine and histamine with bromomethyl cobaloximes and K_{app} values for the reaction of imidazole and substituted imidazoles with iodomethyl cobaloximes are given in table 1. Logarithmic plots of $\log K_{\text{app}}$ vs pH are shown in figure 1 which indicates that as the pH increases the K_{app} increases and the affinity for ligands increases in the order Glyest < Gly < Hisamn < Hisdn for bromomethyl(aquo) cobaloxime and 2Etimd < 1,2-diMeimd < 2-Meimd < Imd < 1-Meimd \ll CN^- for iodomethyl (aquo) cobaloxime. If we compare the pH dependent binding plots of glycine and ethyl glycine ester in both cases K_{app} increases with increase in pH and after certain pH they become pH independent, glycine shows pH dependence up to 10 pH and later becomes pH independent, whereas ethyl glycine ester binding is pH dependent up to 8 pH and later becomes pH independent. The binding of histidine to bromomethyl(aquo) cobaloxime has been shown in figure 2.

The equilibrium constants for the ligation of $\text{ICH}_2\text{Co}(\text{DH})_2\text{OH}_2$ by imidazole, substituted imidazoles and CN^- is also dependent upon the pK_a values of the ligands. In case of imidazole and 1-meimidazole the pH dependent binding constants are measured from pH 5.0 to 8.5, which demonstrate the pH dependent and pH independent binding of these ligands to $\text{ICH}_2\text{Co}(\text{DH})_2\text{OH}_2$, whereas in case of 2-Meimd, 2-Etimd and 1,2 Dimeimd, the binding constants cannot be measured below pH 6.5 as they bind weakly to $\text{Co}(\text{III})$ of cobaloxime. If we compare the binding constants of various ligands with $\text{ICH}_2\text{Co}(\text{DH})_2\text{OH}_2$ they are in the order $K_{\text{CN}^-} \gg K_{1\text{-Meimd}} > K_{\text{Imd}} > K_{2\text{-Meimd}} > K_{1,2\text{-Dimeimd}} > K_{2\text{-Etimd}}$. Though 2-Meimd, 1,2Dimeimd and 2-Etimd are more basic than

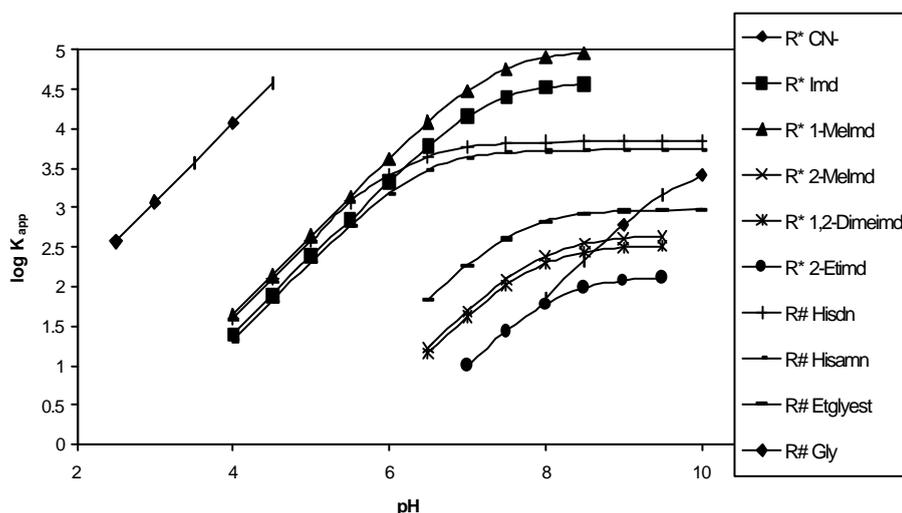


Figure 1. Dependence of $\log K_{\text{app}}$ on pH for the axial ligation of $\text{RCo}(\text{DH})_2\text{OH}_2$ by different ligands at 25°C (R^* – ligation with ICH_2 complex; $\text{R}^\#$ – ligation with BrCH_2 complex).

Table 1. Formation constants ($\log k$) for the axial ligation of $\text{RCo}(\text{DH})_2\text{OH}_2$ by L at 25°C.

L	pH																K_{eq}
	2.5	3.0	3.5	4.0	4.5	5.0	5.5	6.0	6.5	7.0	7.5	8.0	8.5	9.0	9.5	10.0	
<i>R=CH₂Br</i>																	
Hisdn	-	-	-	1.60	2.10	2.58	3.08	3.41	3.65	3.77	3.81	3.83	3.84	3.84	3.84	-	6900
Hisamm	-	-	-	1.32	1.82	2.31	2.78	3.18	3.48	3.64	3.70	3.72	3.73	3.74	3.74	-	5480
EtGlyest	-	-	-	-	-	-	-	-	1.83	2.27	2.61	2.83	2.93	2.96	2.97	2.98	958
Gly	-	-	-	-	-	-	-	-	-	-	-	-	2.33	2.79	3.16	3.41	3979
<i>R=CH₂I</i>																	
CN-	2.57	3.07	3.57	4.07	4.57	-	-	-	-	-	-	-	-	-	-	-	13.6×10^8
1-Meimd	-	-	-	1.64	2.14	2.64	3.13	3.62	4.08	4.48	4.76	4.90	4.96	-	-	-	98400
Imd	-	-	-	1.39	1.89	2.64	2.84	3.33	3.78	4.15	4.40	4.52	4.57	-	-	-	39300
2-Meimd	-	-	-	-	-	-	-	-	1.22	1.69	2.09	2.39	2.54	2.61	2.63	-	440
1,2-Dimeimd	-	-	-	-	-	-	-	-	1.51	1.61	2.01	2.29	2.43	2.49	2.51	-	336
2-Etimd	-	-	-	-	-	-	-	-	-	0.99	1.43	1.77	1.98	2.08	2.11	-	135

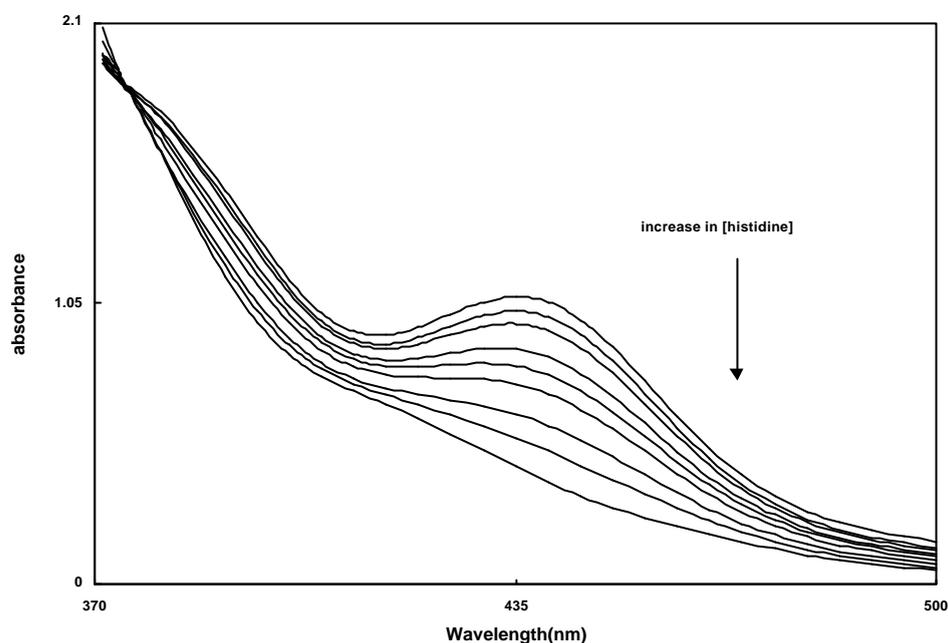


Figure 2. Binding of $\text{BrCH}_2\text{Co}(\text{DH})_2\text{OH}_2$ with varying concentrations of histidine at $\text{pH} = 7.5$ and 25°C , isosbestic point = 380 nm .

Hisdn and Imd, they form less stable complexes. This is due to steric hindrance caused by the methyl or ethyl at the C_2 of imidazole. Similar trends are observed³⁴ in the study of $[\text{CNC}o(\text{DH})_2\text{L}]$ (where $L = 2$ -substituted imidazoles) and in the binding of $P(n\text{-but})_3$ to cobaloximes³⁵. Though the P in $P(n\text{-but})_3$ is soft and more basic than imidazoles it binds weakly, this indicates that steric hindrance plays a dominant role.

In the case of histamine and histidine there is no increase in K_{app} at the pH above the pK_a of the ligand. This clearly indicates that in these ligands the binding is through the endocyclic nitrogen. If it binds through NH_2 group at higher pH , there should be an increase in K_{app} even at higher pH . With histidine, the coordination is through the nitrogen of the imidazole ring, though there is a possibility of COO^- and NH_2 coordination, the NH_2 is mostly protonated below 8.0 pH , hence not available for binding.

A soft or class b character has been assigned to cobaloximes (III)³⁶ and is consistent with the observed greater ligand affinity of cyanide, imidazole^{37,38}, histidine or histamine than the hard glycine or ethyl glycine ester. Furthermore, softness appears to be related to the ability of a cobalt complex to stabilize a $\text{Co}-\text{C}$ bond. $\text{Co}(\text{III})$ to ligand σ bonding is used to explain the reverse order for the dependence of ligation strength upon ligand basicity. The order of $\text{RCo}(\text{DH})_2\text{L}$ stability is attributed to the ability of imidazoles or histidine or histamine to accept electrons into higher energy unfilled σ^* anti bonding orbitals through $d\sigma \rightarrow p\sigma$ back bonding, whereas primary amine (glycine or ethyl glycine ester) cannot accept electrons in either fashion. The reverse order for the dependence of $\text{RCo}(\text{DH})_2\text{L}$ stability on ligand basicity among two series of ligands, aromatic (histamine, histidine, imidazole and substituted imidazoles) and aliphatic (glycine and ethyl glycine ester) is not unexpected based on the following reasons.

- (1) An increase in basicity is associated with increased ability for σ donation for example glycine form more stable complexes than ethyl glycine ester, since glycine is more basic (pK_a 9.74) than ethyl glycine ester (pK_a 7.62).
- (2) An increase in basicity is associated with decreased ability for the aromatic ligands to function as π acceptors.

The values of $K_{\text{Hisdn}} > K_{\text{Hisamn}}$, though histamine is slightly more basic than histidine. Histidine and histamine bind to Co(III), via $N \rightarrow \text{Co(III)}$ donor as well as $\text{Co(III)} \rightarrow N \pi$ bond. Histidine is a better π acceptor than histamine, hence histidine forms more stable complexes than histamine.

The plot of pseudo first-order rate constant k_{obs} against histidine, histamine or imidazole concentration is linear with a very small intercept, which may indicate that a small dissociation is accompanied by the complex formation (figure 3). This appears to be more likely at lower pH (i.e. much below the pK_a of imidazole, histidine or histamine) this is probably due to the protonation of ligand. The kinetic studies cannot be taken at high pH by conventional methods due to fast reactions. This is supposed by the observed high binding constant values at high pH . In case of histidine and histamine, as the pH is increased the rate of formation of complex increases. In case of histamine there is not much change in the k_{obs} even the pH is increased up to 7.0 pH . In both the cases, as the pH is decreased from 4.0 pH initially the rate of dissociation is constant but after reaching 2.5 pH there is a sudden increase in the dissociation rate constant. That means the bound histamine or histidine comes out from the complex at lower pH easily. This supports the very low binding constant at lower pH and high binding constant at higher pH (table 1).

The plots of k_{obs} vs concentration of glycine and ethyl glycine ester give straight lines with non-zero intercepts. The rate of dissociation (k_{off}) increases with decreasing pH (figure 4). For glycine the plot of k_{obs} vs pH increases with pH linearly. Whereas in case of ethyl glycine ester it is sigmoidal that is from pH 7 to 8 it increases slowly and then increases suddenly from 8 to 8.5, after which it is steady and there is no change in the k_{obs} with increase in pH . This can be explained that at high pH it reaches saturation, it means there is no effect of pH on the rate of formation (table 2).

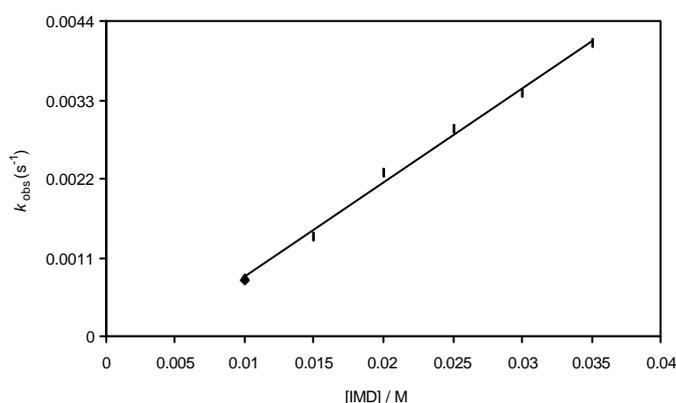


Figure 3. Dependence of [IMD] on pseudo first-order rate constants, k_{obs} , for the formation of $\text{ICH}_2\text{Co}(\text{DH})_2$ IMD at $pH = 5$ and 25°C , the gradient $k_{\text{on}}' = 0.132 \text{ dm}^3 \text{ mol}^{-1} \text{ s}^{-1}$.

Table 2. Kinetic data for the axial ligation of bromomethyl(aquo)cobaloxime by different ligands at 25°C.

pH	$k_{\text{obs}} (\text{s}^{-1})$					$k_{\text{off}} (\text{s}^{-1})$					$k_{\text{obs}} (\text{s}^{-1})$					
	Histidine ($\times 10^3$)	His ($\times 10^3$)	Ethyl glycine ester ($\times 10^2$)	Glycine ($\times 10^2$)	pH	Histidine ($\times 10^2$)	His ($\times 10^3$)	C:L*	Histidine ($\times 10^3$)	His ($\times 10^3$)	Ethyl glycine ester ($\times 10^2$)	Gly ($\times 10^3$)	Histidine ($\times 10^3$)	His ($\times 10^3$)	Ethyl glycine ester ($\times 10^2$)	Gly ($\times 10^3$)
4.5	0.90	0.60	—	—	1.5	8.10	9.50	1:10	1.00	0.76	6.26	1.04	1.00	0.76	6.26	1.04
5.0	1.00	0.76	—	—	—	—	—	1:15	1.80	1.33	7.30	1.25	1.80	1.33	7.30	1.25
5.5	1.80	1.60	—	—	2.0	5.00	5.30	1:20	2.40	1.76	7.92	1.42	2.40	1.76	7.92	1.42
6.0	2.90	2.20	—	—	—	—	—	1:25	3.06	2.20	8.45	1.64	3.06	2.20	8.45	1.64
6.5	4.00	3.80	—	—	2.5	3.20	1.80	1:30	3.84	2.60	9.16	1.78	3.84	2.60	9.16	1.78
7.0	5.08	4.76	0.63	—	—	—	—	1:35	—	—	9.87	2.02	—	—	9.87	2.02
7.5	6.60	6.10	0.73	—	3.0	1.10	1.10	—	—	—	—	—	—	—	—	—
8.0	9.50	6.13	0.89	1.04	—	—	—	k_{on}' (s^{-1})	0.11	0.07	0.1	0.03	0.11	0.07	0.1	0.03
8.5	—	—	1.50	2.91	3.5	1.02	0.90	—	—	—	—	—	—	—	—	—
9.0	—	—	1.64	5.40	—	—	—	α	0.06	0.04	0.193	0.02	0.06	0.04	0.193	0.02
9.5	—	—	1.65	7.40	4.0	0.890	0.65	k_{on} (dm^3 mol^{-1} s^{-1})	2.00	1.93	0.57	1.71	2.00	1.93	0.57	1.71
10.0	—	—	—	9.90	—	—	—	—	—	—	—	—	—	—	—	—
10.5	—	—	—	12.8	—	—	—	—	—	—	—	—	—	—	—	—

*Ratio of $[\text{BrCH}_2\text{Co}(\text{DH})_2\text{OH}_2]$ (1.25×10^{-3}) M and $[\text{L}]\text{M}$.

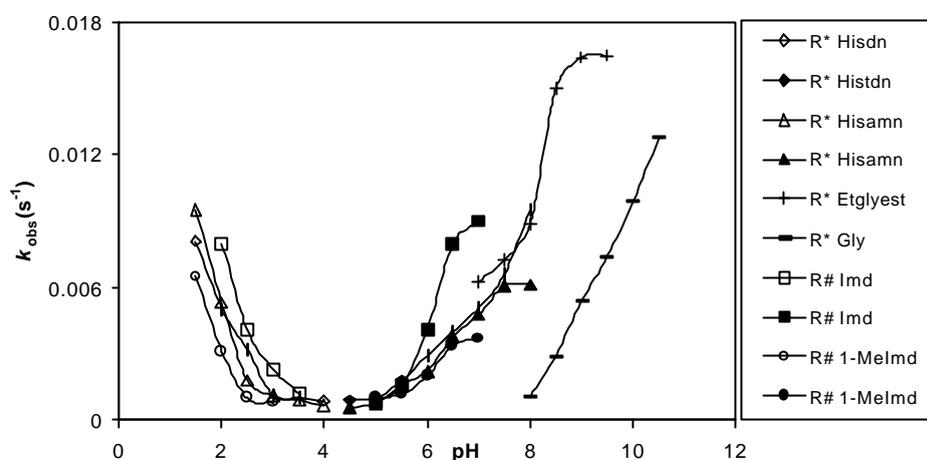


Figure 4. Dependence of k_{obs} on pH for the axial ligation of $[\text{RCo}(\text{DH})_2\text{OH}_2]$ and dependence of k_{off} on pH for the dissociation of L from $[\text{RCo}(\text{DH})_2L]$ complex at 25°C . ($\text{R}^* - \text{BrCH}_2$; $\text{R}^\# - \text{ICH}_2$).

Figure 4 shows the pseudo first-order rate constants for the formation (k_{obs}) and dissociation (k_{off}) as a function of pH (table 3). For Imd k_{obs} increases slowly up to 3.5 pH and then there is a sharp rise. For CN^- there is a slow increase between 1.0 and 1.5 pH then rises sharply between 2.0 and 3.0 pH . Later it is steady and there is not much increase in k_{obs} with increase in pH . These kinetic data are supported by binding data. The rate of dissociation of Imd and CN^- *trans* to the $[\text{ICH}_2\text{Co}(\text{DH})_2L]$ complex increases with decrease in pH . Imidazole can be removed completely at pH 2.0 whereas CN^- is removed at 0.0 pH . This also supports the fact that CN^- binds more strongly than imidazole.

The kinetics of substitution of the axial base in alkylcobaloximes and related cobalt complexes has been studied under a variety of conditions^{39,40}. In none of the studies was the mechanism established conclusively although in all cases strong evidence was provided that the intimate mechanism is dissociative (Id or D).

In coordinating solvents ‘pentacoordinate’ species are formed involving pentacoordinate alkylcobalt complexes and solvent. In view of the evidence presented above for the existence of pentacoordinate alkylcobaloximes and the ligation kinetic studies of others, both on alkyl cobalt complexes with other equatorial ligand system⁴¹ and on cobaloxime(III) complexes^{42,43}, an $\text{S}_{\text{N}}1$ mechanism appears to be operative.

The small dependence of k_{on} upon ligand basicity within each series of ligands is clearly related to the fact that while the reacting complex is a soft acid the ligand is hard. The rate constants are better correlated with the relative softness of the ligand among the ligands we have studied.

The small difference in the rate of ligand substitution despite large differences in the stabilities of the $\text{Co}(\text{III})$ complexes⁴⁴ and aquo cobalamine have been taken to indicate the lack of significant activation of the transition state by the incoming ligand and, conversely, domination of the transition state activation by the leaving ligand (i.e., a dissociative interchange mechanism Id)⁴⁵. The stability of pentacoordinate alkyl cobalt complexes and the evidence that both the dominant soft $\text{Co}(\text{III})$ complexes,

Table 3. Kinetic data for the axial ligation of iodomethyl(aquo)cobaloxime by different ligands at 25°C.

pH	$k_{\text{obs}} (\text{s}^{-1})$				$k_{\text{off}} (\text{s}^{-1})$				$k_{\text{obs}} (\text{s}^{-1})$					
	Imd ($\times 10^3$)	1-Me Imd ($\times 10^3$)	CN ⁻ ($\times 10^2$)	pH	CN ⁻ ($\times 10^2$)	Imd ($\times 10^3$)	1-Me Imd ($\times 10^3$)	C:L*	Imd ($\times 10^3$)	1-Me Imd ($\times 10^3$)	CN ⁻	Imd ($\times 10^3$)	1-Me Imd ($\times 10^3$)	CN ⁻
1.5	-	-	1.42	0.0	1.17	-	-	1:10	0.8	0.7	0.01	-	-	-
2.0	-	-	1.78	-	-	-	-	-	-	-	-	-	-	-
2.5	-	-	2.98	0.5	1.01	-	-	1:15	1.4	1.1	0.02	-	-	-
3.0	-	-	3.46	-	-	-	-	-	-	-	-	-	-	-
3.5	-	-	3.5	1.0	0.75	-	-	1:20	2.3	1.5	0.02	-	-	-
4.0	-	-	-	-	-	-	-	-	-	-	-	-	-	-
4.5	-	-	-	1.5	-	-	6.50	1:25	2.9	1.9	0.03	-	-	-
5.0	0.8	1.0	-	-	-	-	-	-	-	-	-	-	-	-
5.5	1.6	1.2	-	2.0	-	80.0	3.1	1:30	3.4	2.3	0.03	-	-	-
6.0	4.1	2.0	-	-	-	-	-	-	-	-	-	-	-	-
6.5	8.0	3.4	-	2.5	-	4.1	1.0	1:35	4.1	2.7	0.04	-	-	-
7.0	9.0	3.7	-	-	-	-	-	-	-	-	-	-	-	-
				3.0	-	2.2	0.8	k_{on}' (s^{-1})	0.132	0.8	1.034			
				3.5	-	1.2	-	α	0.006	0.001	2.8×10^{-6}			
								k_{on} ($\text{dm}^3 \text{mol}^{-1} \text{s}^{-1}$)	23.07	567.15	3.8×10^7			

*Ratio of $[\text{ICH}_2\text{Co}(\text{DH})_2\text{OH}_2]$ (1.00×10^{-3}) M and $[\text{L}]\text{M}$.

$[\text{Co}(\text{CN})_5\text{H}_2\text{O}]^{2-}$ and $[\text{Co}(\text{NH}_3)_5\text{SO}_3]^+$, undergo SN^1 ligand substitution reactions^{46,47}, clearly favor this mechanism for the ligation reaction of $\text{BrCH}_2\text{Co}(\text{DH})_2\text{OH}_2$. The coordination between the softness of a cobalt(III) complex and the stability of its pentacoordinate species permits SN^1 mechanism for ligand substitution⁴⁸.

To compare the rate constants of the various ligands for the formation of complex with $\text{BrCH}_2\text{Co}(\text{DH})_2\text{OH}_2$ and $\text{ICH}_2\text{Co}(\text{DH})_2\text{OH}_2$, we have calculated the second order rate constant, k'_{on} from the slopes of the pseudo first-order rate constants as a function of concentration of the ligand. Since this is also pH -dependent for better comparison we have calculated k_{on} , the pH independent second-order rate constant. The order of k_{on} is as follows: $\text{CN}^- \gg 1\text{-Meimd}^{28} > \text{Imd} > \text{Hisdn} > \text{Hisamn} > \text{Gly} > \text{Etglyest}$. This is in accordance with the basicity order of the ligands. Though the basicity of glycine and ethyl glycine ester are larger than imidazole, histidine or histamine k_{on} are much smaller. But within glycine and ethyl glycine ester again they follow the basicity order, k_{on} of glycine > ethyl glycine ester. This can be explained based on \mathbf{p} bonding and HSAB principle.

4. Conclusions

In the ligation reaction of $\text{BrCH}_2\text{Co}(\text{DH})_2\text{OH}_2$ and $\text{ICH}_2\text{Co}(\text{DH})_2\text{OH}_2$ the \mathbf{p} accepting ligands (cyanide, imidazole, histidine or histamine) react more rapidly than the purely \mathbf{s} donors (glycine or ethyl glycine ester). The greater reactivities of the cyanide, imidazole, histidine and histamine compound to glycine or ethyl glycine ester are discussed based on the basicity, $\mathbf{d-p-p}$ back bonding and HSAB principle. From these studies we also found that there is severe steric strain between substituent at the C_2 of a coordinated imidazole and the cobaloxime.

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References

1. Toraya T, Krodel E, Mildran A S and Abeles R H 1979 *Biochemistry* **18** 417
2. Abeles R H and Dolphin D 1976 *Acc. Chem. Res.* **9** 114
3. Halpern J 1974 *Ann. NY. Acad. Sci.* **2** 239
4. Dolphin D (ed.) 1982 *B₁₂* (New York: Wiley) vol. 2
5. Pratt J M 1985 *Chem. Soc. Rev.* 161
6. Hay B P and Finke R G 1987 *J. Am. Chem. Soc.* **109** 8012
7. Ashley K R 1976 *J. Inorg. Nucl. Chem.* **39** 357
8. Ashley K R and Leipoldt G J 1981 *Inorg. Chem.* **20** 2326
9. Leipoldt G J, Van Eldik R and Kelm H 1983 *Inorg. Chem.* **22** 4146
10. Fleischer E B and KrishnaMurthy M 1971 *J. Am. Chem. Soc.* **93** 3784
11. KrishnaMurthy M 1977 *Inorg. Chim. Acta* **25** 215
12. Ashley K R, Shyu S and Leipoldt G J 1980 *Inorg. Chem.* **19** 1613
13. Choo P L, Mulichak A M, Jones R W Jr., Bacon J W and Pett V B 1990 *Inorg. Chim. Acta* **171** 183
14. Brown K L and Kallen R G 1972 *J. Am. Chem. Soc.* **94** 1894
15. Brown K L, Lyles D, Pencovici M and Kallen R G 1975 *J. Am. Chem. Soc.* **97** 7338
16. Brown K L and Satyanarayana S 1992 *Inorg. Chim. Acta* **201** 113
17. Hirota S, Kosugi E, Marzilli L G and Yamauchi O 1998 *Inorg. Chim. Acta* **275** 90

18. Rajeshwar Rao A, Sridhar V and Satyanarayana S 1999 *Proc. Natl. Acad. Sci. India* **A69** 1
19. Randaccio L, Bresciani-Pahor N, Zangardo E and Marzilli L G 1989 *Chem. Soc. Rev.* **16** 229
20. Bresciani-Pahor N, Forcolin M, Marzilli L G, Randaccio L, Summers M F and Toscano P J 1985 *Coord. Chem. Rev.* **63** 1
21. Kim S H, Chen H L, Feilchenfeild N and Halpern J 1988 *J. Am. Chem. Soc.* **110** 3120
22. Crumbliss A L and Wilmarth W K 1970 *J. Am. Chem. Soc.* **92** 2593
23. Trogler W C, Stewart R C and Marzilli L G 1974 *J. Am. Chem. Soc.* **96** 3641
24. Elliot C M, Hershenhart E, Finke R G, Smith B L 1981 *J. Am. Chem. Soc.* **103** 5558
25. Parker W O Jr, Bresciani-Pahor N, Zangrando E, Randaccio L and Marzilli L G 1985 *Inorg. Chem.* **24** 3908
26. Reenstra W W and Jencks W P 1979 *J. Am. Chem. Soc.* **101** 5780
27. Summers M F, Marzilli L G, Bresciani-Pahor N and Randaccio L 1984 *J. Am. Chem. Soc.* **106** 4478.
28. Rossi M, Glusker J P, Randaccio L, Summers M F, Toscano P J and Marzilli L G 1985 *J. Am. Chem. Soc.* **107** 1729
29. Halpern J 1983 *J. Pure Appl. Chem.* **55** 3677
30. Mohamed S A Hamza, Carlos Ducker-Benfer and Rudi Van Eldik 2000 *Inorg. Chem.* **39** 3777
31. Drennan C L, Huang S, Drummond J T, Mathews R G and Ludwig M C 1994 *Sciences* **206** 2669
32. Manica F, Keep N H, Nakagawa A, Leadlay P F, Mc Swerney S, Ramussen B, Bosake P, Diat O and Evans P R 1996 *Structure* **4** 229
33. Brown K L 1986 *Organometallic syntheses* (eds) R B King and J J Eisch (Amsterdam: Elsevier) vol 3, p 186
34. Marques H M, Egan T J, Marsh J H, Mellor J R and Munro O Q 1989 *Inorg. Chim. Acta* **166** 249
35. Frederick R J and Ronald C K 1975 *J. Am. Chem. Soc.* **97** 5820
36. Hague D N and Halpern J 1967 *J. Inorg. Chem.* **6** 2059
37. Sridhar V and Satyanarayana S 2000 *Proc. Indian Acad. Sci. (Chem. Sci.)* **112** 579
38. Sridhar V and Satyanarayana S 2001 *Indian J. Chem* **A40** 165
39. Poon C K 1973 *Coord. Chem. Rev.* **10** 1
40. Herlinger A W and Brown T L 1972 *J. Am. Chem. Soc.* **94** 388
41. Costa G, Mestroni G, Tazher G, Goddall D M and Hill H A O 1970 *Chem. Commun.* 34
42. Earley J E and Zimmerman J G 1972 *Inorg. Nucl. Chem. Lett.* **8** 687
43. Zsako J, Finta Z and Varhelyi C S 1972 *J. Inorg. Nucl. Chem.* **34** 2887
44. Eigen M and Wilkins R G 1965 *Adv. Chem. Ser. No. 4*
45. Longford D H and Gray H B 1965 *Ligand substitution processes* (ed.) A Benjamin (New York:)
46. Haim A J, Grassi R J and Wilmarth W K 1965 *Adv. Chem. Ser. No. 49*
47. Halpern J, Palmer R A and Blakley L M 1966 *J. Am. Chem. Soc.* **88** 2877
48. Firth R A, Hill H A O, Pratt J M, Thorp R G and Williams R J 1969 *J. Chem. Soc. A* 381