# Kinetics of the Formation and Dissociation of Pentacyanoferrate(II) Complexes of Cysteine, Penicillamine, Glutathione, and 2-Mercaptoethylamine †

By **Donal H. Macartney** and **Alexander McAuley,*** Department of Chemistry, University of Victoria, Victoria, B.C., Canada V8W 2Y2

The results of a kinetic study of the formation and dissociation of pentacyanoferrate(II) complexes of cysteine (cys), glutathione (glut), penicillamine (pen), and 2-mercaptoethylamine (mea) are reported. Formation rate measurements have been made in the range pH 1—10 and the observed acid dependences are attributed to reaction of various ionic forms of the ligands related by protonation equilibria. For the neutral ligands the rate constants for formation ($k_L$) of complexes with cys, glut, pen, and mea are 330, 370, 350, and 200 $dm^3\ mol^{-1}\ s^{-1}$, respectively, at 25 °C and $I$ = 0.50 mol $dm^{-3}$ ($Na[ClO_4]$). A correlation between the formation rate constants and the ligand charge type is observed; deviations from this relationship are discussed in terms of the location of the charges in the ligand. Rates of ligand release have been measured over the range pH 2.5—11, in the presence of an excess of pyrazine, and a hydrogen-ion dependence observed for each ligand. A decrease in the dissociation rate with increasing pH is attributed to the reduced reactivity of $[Fe(CN)_5(^-SR)]^{n-}$ with respect to its conjugate acid $[Fe(CN)_5(HSR)]^{(n-1)-}$. At pH 5 the rate constants for dissociation ($k_{-L}$) of cys, glut$^-$, pen, and Hmea$^+$ from $[Fe(CN)_5L]^{n-}$ are 0.104, 0.055, 1.34, and 0.102 $s^{-1}$, respectively. The large value of $k_{-L}$ for penicillamine is associated with the steric effects of the methyl groups.

AMINO-ACIDS and peptides containing sulphur have been shown to be important in many biological systems.[1] Thiolate is a functional group frequently associated with these ligands which can also bind to metal ions in metalloproteins. Two important features of a sulphur-containing compound of this type are the ability of the sulphur centre to compete with other functional groups for the metal-binding sites and the effect of the metal ion on the ligand once co-ordinated. With the pentacyanoferrate(II) ion there is only a single site for ligand co-ordination and this can be used to investigate the binding of specific functional groups.

The reactions of the aquapentacyanoferrate(II) with a number of nitrogen heterocycles,[2] amines,[3] and sulphur [4,5] donor ligands have been studied previously. Systems investigated include the biologically important ligands imidazole,[6] histidine,[6] glycine,[6] and isonicotinohydrazide.[7] The importance of protonated and deprotonated ligand forms and the effect of these features on the reaction rates has also been described.[8-11]

In this paper the kinetics of the complexation of $[Fe(CN)_5(OH_2)]^{3-}$ with cysteine (cys), glutathione (γ-glutamylcysteinylglycine)(glut), penicillamine (3-mercaptovaline) (pen), and 2-mercaptoethylamine (mea) are reported. Specific rate constants for complexes of these ligands (and their conjugate acids and bases) were determined by measuring the formation rates over the range pH 1—10. The effect of the ligand charge on the reaction rate is discussed in terms of an ion-pair dissociative mechanism.

We also report the results of a kinetic study of the release of the ligands from $[Fe(CN)_5L]^{n-}$. The complete dissociation of the ligand from the metal centre can be accomplished by the addition of an excess of a strongly co-ordinating ligand such as pyrazine (pyz). The rates of dissociation were measured in the range pH 2.5—11 where both protonated and deprotonated forms of the co-ordinated ligands may exist.

This work also provides information concerning the acid-dissociation constants of the co-ordinated ligands and the effect of protonation on the kinetics of ligand substitution in these systems.

## EXPERIMENTAL

*Reagents.*—L-Glutathione, DL-penicillamine, and 2-mercaptoethylamine (Koch-Light) and L-cysteine (Sigma) were used without further purification. Sodium amminepentacyanoferrate(II), $Na_3[Fe(CN)_5(NH_3)]\cdot 3H_2O$, was prepared from $Na_3[Fe(CN)_5(NO)]$ by a standard procedure [12] and recrystallized several times from concentrated ammonia.[5] Solutions of aquapentacyanoferrate(II) were produced by dissolving solid $Na_3[Fe(CN)_5(NH_3)]\cdot 3H_2O$ in nitrogen-saturated distilled water. Low concentrations ($2 \times 10^{-5}$—$4 \times 10^{-5}$ mol $dm^{-3}$) of the complex were used to prevent dimerization.[13] Solutions of pentacyano(ligand)ferrate(II) were prepared by adding an excess of the ligand ($10^{-3}$ mol $dm^{-3}$) to a deoxygenated solution of aquated $[Fe(CN)_5(NH_3)]^{3-}$ ($5 \times 10^{-5}$—$10 \times 10^{-5}$ mol $dm^{-3}$). Pyrazine (Aldrich) solutions were prepared in deoxygenated distilled water. The visible spectra of the pentacyanoferrate(II) complexes were measured with a Beckman DU-8 computing spectrophotometer.

Kinetic studies were made using a stopped-flow apparatus described previously.[14] The apparatus was thermostatted to ±0.05 °C in the temperature range 8—30 °C. The pH of the reaction mixture was controlled using phthalate, phosphate, and borate buffers and appropriate amounts of $HClO_4$ at low pH. Solution acidities between pH 2 and 11 were measured using a Radiometer 26 instrument with a Radiometer GK2311C combination electrode. All solutions were deoxygenated using nitrogen and maintained at an ionic strength of 0.50 mol $dm^{-3}$ with sodium perchlorate.

The formation reactions were monitored at 440 nm, the $\lambda_{max.}$ of $[Fe(CN)_5(OH_2)]^{3-}$, while the dissociation reactions were followed at 454 nm, the $\lambda_{max.}$ of $[Fe(CN)_5(pyz)]^{3-}$

† Penicillamine = 3-mercaptovaline, glutathione = γ-glutamylcysteinylglycine.

in the visible region. Pseudo-first-order conditions were employed with the ligand in excess. Absorbance data derived from photomultiplier output were collected by a PCM-12 mini-computer. Plots of $\ln(A_t - A_\infty)$ against time were linear for three or more half-lives. Basic kinetic data are available as Supplementary Publication No. SUP 23080 (16 pp.).*

## RESULTS

The visible spectra of the pentacyanoferrate(II) complexes of cysteine, glutathione, penicillamine, and 2-mercaptoethylamine are shown in Figure 1 together with the spectrum of $[Fe^{II}(CN)_5(OH_2)]^{3-}$. The spectra were measured at pH 5

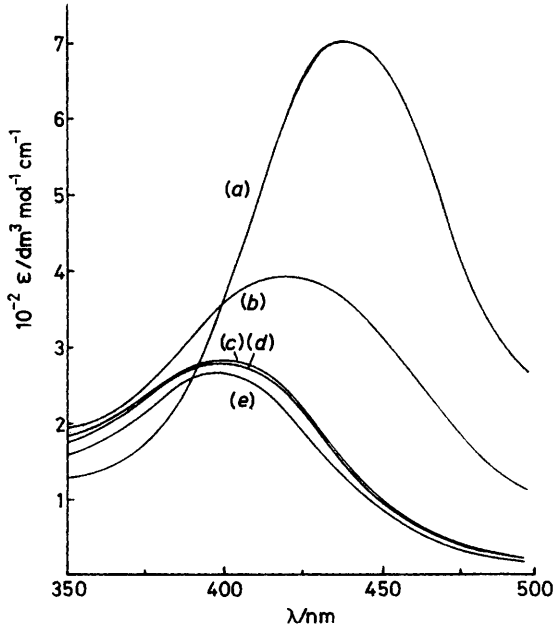



FIGURE 1 Visible spectra of pentacyanoferrate(II) complexes, $[Fe(CN)_5L]^{n-}$, L = $OH_2$ (*a*), pen (*b*), glut$^-$ (*c*), cys (*d*), and Hmea$^+$ (*e*)

plexes with neutral sulphur [5] (*e.g.* thiourea) and saturated nitrogen-donor ligands.[6,8] The spectrum of the penicillamine complex has its maximum at a lower energy. This difference is reflected in other properties as discussed below. The positions and absorption coefficients of the visible peaks are presented in Table 1.

With the ligands used in this study, co-ordination to the metal centre could occur either through the sulphur of the thiol group or the nitrogen of the amine groups. Evidence from spectroscopic studies suggests that the former is the case. Under the conditions (pH 5—6) studied, the ligand is principally (>90%) in the form where the thiol group remains protonated.[15,16] We have no evidence for any dissociation of the hydrogen ion on complex formation and consider the thiol to be co-ordinated as a neutral ligand (RSH). This is in contrast to the complexing of RSH to cation centres (*e.g.* $Fe^{3+}$) [14] where there is evidence for co-ordination *via* $RS^-$. Attempts to prepare pentacyanoferrate(II) complexes with ethanolamine and serine, the oxygen analogues of mercaptoethylamine and cysteine, were unsuccessful until the pH was raised above the $pK_a$ of the amine groups.[15,16] Similar results have been observed [8] with $H_2en^{2+}$ (en = ethylenediamine) indicating that a quaternary ammonium group will not co-ordinate to $[Fe(CN)_5]^{3-}$ until it is deprotonated.

The similarity of the spectrum of the pentacyanoferrate(II) complex of glutathione to those of mercaptoethylamine and cysteine at pH 5 suggests that this ligand is also sulphur bonded. Additional evidence in the case of penicillamine comes from the spectrum of the pentacyanoferrate(III) complex. The charge-transfer peak at 680 nm is near to those found for sulphur-donor ligands such as thiourea (600 nm) [17] and thiomalic acid (675 nm).[18] Reduction of $[Fe^{III}(CN)_5(pen)]^{2-}$ with ascorbic acid produced $[Fe^{II}(CN)_5(pen)]^{3-}$ with no evidence in the stopped-flow range for isomerization of co-ordination sites. At high pH, there is the possibility of co-ordination either through $RS^-$ or $NH_2$ donor groups, however no unusual behaviour was observed to suggest substantially different rates of release of $RS^-$ and $NH_2R'$. Further kinetic and spectroscopic studies are in progress to confirm these observations.

TABLE 1

Properties of the sulphur-containing ligands $HSCR^1{}_2CR^2R^3$ and their pentacyanoferrate(II) complexes

| Ligand | Substituents [a] R¹ | R² | R³ | $pK_a$ values of ligand [b] | $\lambda_{max}$ [b,c]/nm of $[Fe(CN)_5L]^{n-}$ |
|---|---|---|---|---|---|
| Cysteine | H | $COO^-$ | $NH_3^+$ | 1.80 ($H_3L^+ \rightleftharpoons H_2L + H^+$)<br>8.10 ($H_2L \rightleftharpoons HL^- + H^+$)<br>10.07 ($HL^- \rightleftharpoons L^{2-} + H^+$) | 402 (275) |
| Penicillamine | $CH_3$ | $COO^-$ | $NH_3^+$ | 1.85 ($H_3L^+ \rightleftharpoons H_2L + H^+$)<br>7.80 ($H_2L \rightleftharpoons HL^- + H^+$)<br>10.45 ($HL^- \rightleftharpoons L^{2-} + H^+$) | 421 (360) |
| Mercaptoethylamine | H | H | $NH_3^+$ | 8.26 ($H_2L^+ \rightleftharpoons HL + H^+$)<br>10.80 ($HL \rightleftharpoons L^- + H^+$) | 402 (265) |
| Glutathione | H | $CONHCH_2COO^-$ | $NHCO(CH_2)_2CH(NH_3^+)COO^-$ | 1.95 ($H_4L^+ \rightleftharpoons H_3L + H^+$)<br>3.40 ($H_3L \rightleftharpoons H_2L^- + H^+$)<br>8.55 ($H_2L^- \rightleftharpoons HL^{2-} + H^+$)<br>9.40 ($HL^{2-} \rightleftharpoons L^{3-} + H^+$) | 402 (280) |

[a] pH 5.0. [b] $I$ = 0.50 mol dm$^{-3}$. [c] ε/dm³ mol$^{-1}$ cm$^{-1}$ given in parentheses.

in a solution containing an excess of ligand. The absorbance maxima of the cys, glut, and mea complexes are nearly identical to one another and are similar in position and absorption coefficients to other pentacyanoferrate(II) com-

The kinetics of the formation reactions of $[Fe(CN)_5(OH_2)]^{3-}$ with the ligands cys, glut, mea, and pen were investigated using the stopped-flow technique. The re-

$$[Fe(CN)_5(OH_2)]^{3-} + L \underset{k_{-L}}{\overset{k_L}{\rightleftharpoons}} [Fe(CN)_5L]^{3-} + H_2O \quad (1)$$

* For details see Notices to Authors No. 7, *J. Chem. Soc., Dalton Trans.*, 1980, Index issue.

actions were studied under pseudo-first-order conditions with excess of ligand by following the decay of $[Fe(CN)_5(OH_2)]^{3-}$ at 440 nm. The rate of decay may be expressed in the form (2), good first-order behaviour being observed

$$-d[Fe(CN)_5(OH_2)^{3-}]/dt = k_{obs.}[Fe(CN)_5(OH_2)^{3-}] \quad (2)$$

to three or more half-lives. Plots of $k_{obs.}$ against ligand concentration, shown in Figure 2, are linear with positive

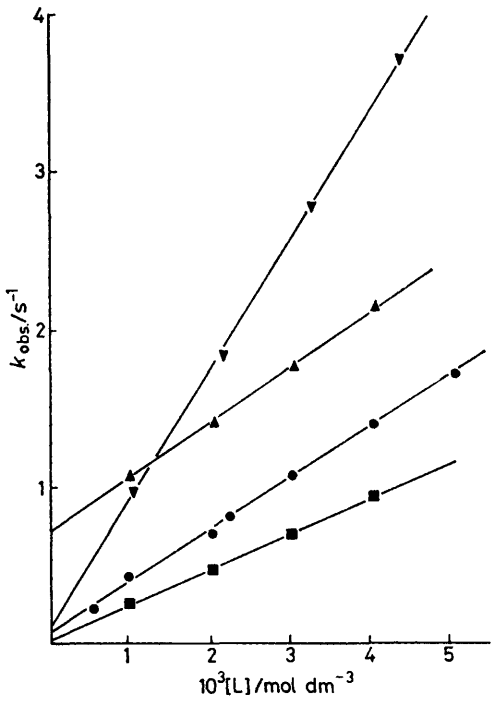



FIGURE 2 Variation of $k_{obs.}$ with ligand concentration for the formation reactions of $[Fe(CN)_5L]^{n-}$ with L at 25 °C, pH 6, and $I$ = 0.50 mol $dm^{-3}$ ($Na[ClO_4]$). L = cys (●), $glut^-$ (■), pen (▲), or $Hmea^+$ (▼)

intercepts. These observations are consistent with equation (1) where $k_{obs.}$ may be expressed as in (3). The term

$$k_{obs.} = k_L[L] + k_{-L} \quad (3)$$

$k_{-L}$ is the rate constant for the dissociation of L from $[Fe^{II}(CN)_5L]^{n-}$. The kinetics of these dissociation reactions are more conveniently studied in the presence of an excess of a strongly co-ordinating ligand such as pyrazine. Investigations of this nature were undertaken and the results are presented and discussed below.

pH *Dependence of* $k_L$.—The complex-formation reactions were investigated over the range pH 1—10. The rates of formation at various hydrogen-ion concentrations at 25 °C are listed in Table 2. In order to analyze the rate dependence on acid concentration in these reactions a knowledge is necessary of the metal and ligand species present in solution at the various pH values used. The ligands under consideration display several acid-dissociation equilibria within the pH range, associated with the deprotonation of the functional groups (COOH, SH, and $NH_3^+$) present.[15,16] The macroscopic $pK_a$ values for the four ligands are listed in Table 1. They are for an ionic strength of 0.50 mol $dm^{-3}$ and were calculated by interpolations from literature values.[19-22] For simple amines the $pK_a$ increases with increasing ionic strength ($I$), while with aminocarboxylic acids there is a decrease in the $pK_a$ with increasing $I$ to a minimum at $I \approx 0.5$ mol $dm^{-3}$ followed by an increase as $I$ continues to rise.[15] The cysteine system has been studied in some detail [21] and a similar approach has been used for other ligands in this study.

TABLE 2

Variation of the formation rate constants with pH at 25 °C and $I$ = 0.50 mol $dm^{-3}$ ($Na[ClO_4]$)

| Ligand | pH | $k_L/dm^3\ mol^{-1}\ s^{-1}$ | pH | $k_L/dm^3\ mol^{-1}\ s^{-1}$ |
|---|---|---|---|---|
| Cysteine | 1.30 | 14 | 7.31 | 277 |
| | 2.40 | 166 | 8.05 | 203 |
| | 2.79 | 240 | 8.60 | 128 |
| | 3.65 | 310 | 8.71 | 108 |
| | 5.02 | 331 | 9.19 | 83 |
| | 6.55 | 329 | | |
| Glutathione | 1.62 | 58 | 5.59 | 223 |
| | 2.46 | 225 | 5.79 | 231 |
| | 2.92 | 255 | 6.93 | 218 |
| | 3.49 | 266 | 8.00 | 178 |
| | 3.66 | 260 | 8.55 | 103 |
| | 4.22 | 237 | 9.71 | 43 |
| Penicillamine | 2.03 | 142 | 5.75 | 354 |
| | 2.78 | 246 | 7.20 | 316 |
| | 3.68 | 308 | 7.99 | 199 |
| | 4.80 | 347 | 8.76 | 101 |
| | 5.18 | 349 | 9.34 | 52 |
| Mercaptoethylamine | 1.28 | 51 | 6.72 | 792 |
| | 1.87 | 109 | 6.77 | 782 |
| | 2.40 | 435 | 7.24 | 721 |
| | 3.37 | 754 | 7.93 | 562 |
| | 4.38 | 836 | 8.78 | 274 |
| | 5.85 | 853 | 9.75 | 148 |

The lower $pK_a$ values (1.8—3.4) correspond [20] to the carboxyl groups while the higher values have been assigned to the thiol (7.8—8.6) and amine (9.4—10.8) groups. At neutral pH (5—7) the ligands cys, $glut^-$, and pen exist in the zwitterionic form $[HS{-}R(NH_3^+)COO^-]$. At low pH the $[Fe(CN)_5(OH_2)]^{3-}$ ion may also be protonated, at one of the cyanide ligands. The acid-dissociation constant ($K_m$) for this process has been determined by Malin and Koch [23] to be $2.35 \times 10^{-3}$ mol $dm^{-3}$ at 25 °C.

The reaction scheme in equations (4)—(11) may be written, involving the pH-related forms of the metal complex and the ligands (Table 1). The nature of the products

$$[HFe(CN)_5(OH_2)]^{2-} \overset{K_m}{\rightleftharpoons} [Fe(CN)_5(OH_2)]^{3-} + H^+ \quad (4)$$

$$H_4L^+ \overset{K_1}{\rightleftharpoons} H_3L + H^+ \quad (5)$$

$$H_3L \overset{K_2}{\rightleftharpoons} H_2L^- + H^+ \quad (6)$$

$$H_2L^- \overset{K_3}{\rightleftharpoons} HL^{2-} + H^+ \quad (7)$$

$$[Fe(CN)_5(OH_2)]^{3-} + H_4L^+ \overset{k_1}{\rightleftharpoons} \text{products} \quad (8)$$

$$[Fe(CN)_5(OH_2)]^{3-} + H_3L \overset{k_2}{\rightleftharpoons} \text{products} \quad (9)$$

$$[Fe(CN)_5(OH_2)]^{3-} + H_2L^- \overset{k_3}{\rightleftharpoons} \text{products} \quad (10)$$

$$[Fe(CN)_5(OH_2)]^{3-} + HL^{2-} \overset{k_4}{\rightleftharpoons} \text{products} \quad (11)$$

$[Fe(CN)_5L]^{n-}$ will depend on the specific ligand acid-dissociation constants, some of which have been determined by studies of the pH-dependent dissociation reactions of $[Fe(CN)_5L]^{n-}$ (see below).

The second-order rate constant, $k_L$, may be expressed in terms of the specific rate constants and acid-dissociation constants in the general form (12). The rate constants

$$k_L = \frac{\{(k_1[H^+]/K_1) + k_2 + (k_3K_2/[H^+]) + (k_4K_2K_3/[H^+]^2)\}}{\{1 + ([H^+]/K_1) + (K_2/[H^+]) + (K_2K_3/[H^+]^2)\}(1 + [H^+]/K_m)} \quad (12)$$

associated with individual reaction steps for each complex are presented in Table 3. The pH profiles of the formation

TABLE 3

Second-order rate constants ($dm^3$ $mol^{-1}$ $s^{-1}$) for the formation of $[Fe(CN)_5L]^{n-}$ at 25 °C and $I$ = 0.50 mol $dm^{-3}$ ($Na[ClO_4]$)

| Ligand | $k_1$ | $k_2$ | $k_3$ | $k_4$ |
|---|---|---|---|---|
| Cysteine | 900 | 330 | 70 | |
| Glutathione | 950 | 370 | 220 | 35 |
| Penicillamine | 850 | 350 | 70 | |
| Mercaptoethylamine | 850 | 200 | 30 | |

rates are shown in Figure 3 for cys and mea and in Figure 4 for pen and glut. These plots are basically bell-shaped

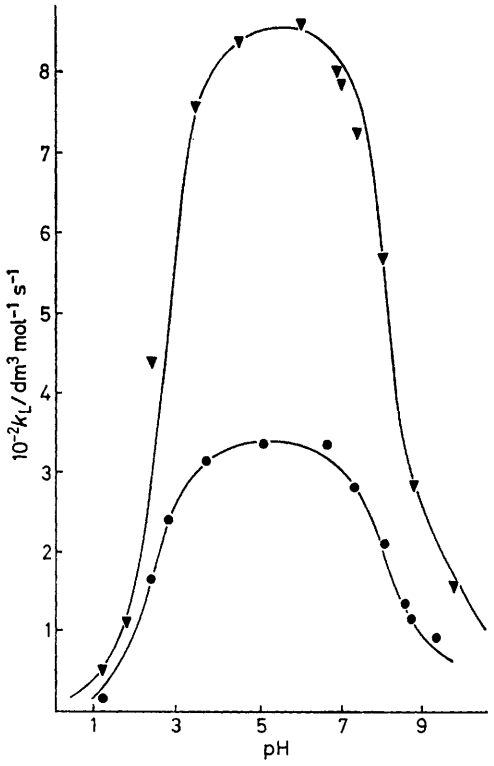



FIGURE 3 pH Profile of the rate constants for the formation of $[Fe(CN)_5L]^{n-}$ at 25 °C where L = cys (●) or mea (▼). The solid curves are derived from equation (12)

curves with a plateau in the range pH 4—6. At higher acidities the rate decreases as the iron(II) complex is in the less reactive form, $[HFe(CN)_5(OH_2)]^{2-}$. A marked decrease in the lability of the aquo-ligand in $[HFe(CN)_5(OH_2)]^{2-}$ had previously been observed in the reactions with neutral thioureas [24] ($k \leqslant 1$ $dm^3$ $mol^{-1}$ $s^{-1}$) and the *N*-methylpyrazinium cation [23] ($k = 23 \pm 42$ $dm^3$ $mol^{-1}$ $s^{-1}$).

The reaction scheme presented above does not include terms for the reaction pathways involving $[HFe(CN)_5(OH_2)]^{2-}$ with $H_3L$ and $H_4L^+$ where specific rate constants have been estimated to be *ca.* 1 and *ca.* 5—10 $dm^3$ $mol^{-1}$ $s^{-1}$ respectively. The plateau region corresponds to the reaction of the $[Fe(CN)_5(OH_2)]^{3-}$ ion with the neutral (zwitterionic) forms of cys and pen. The rate constants for the overall neutral ligands ($k_2$) are in the range 200—370 $dm^3$ $mol^{-1}$ $s^{-1}$, consistent with the values observed [2-5] for a variety of uncharged ligands (*e.g.* imidazole or thiourea). The values are higher, however, than those obtained for glycinate or other anionic ligands ($NO_2^-$ and $CN^-$).[11,13]

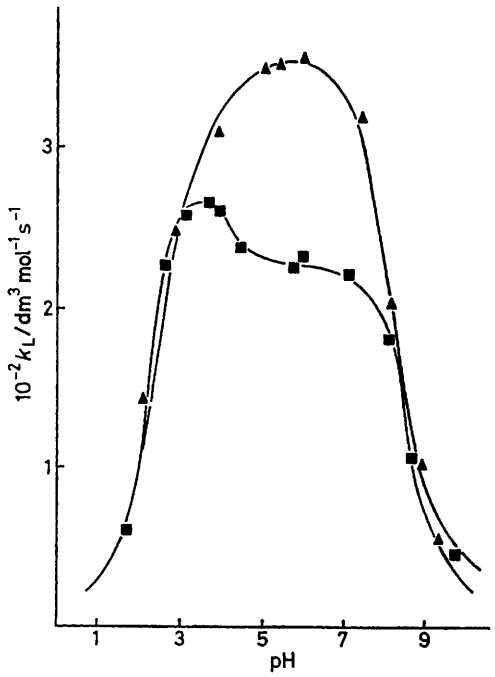



FIGURE 4 pH Profile of the rate constants for the formation of $[Fe(CN)_5L]^{n-}$ at 25 °C where L = pen (▲) or glut (■). Solid curves as in Figure 3

The fact that the observed rate constants are similar to those for neutral centres is seen as supportive evidence for co-ordination of the RSH moiety. Up to pH 8, mercaptoethylamine exists as a cation, $HSCH_2CH_2NH_3^+$. The rate constant for $Hmea^+$ (850 $dm^3$ $mol^{-1}$ $s^{-1}$) is very similar to the value found for the *N*-methylpyrazinium ion (920 $dm^3$ $mol^{-1}$ $s^{-1}$) [2] at the same ionic strength (0.50 mol $dm^{-3}$).

The pH–rate profile (Figure 4) for glutathione which has two carboxylate groups is also a bell-shaped curve except that a second maximum is observed at pH 3.3. The rate terms involved are for reaction of $glut^+$ ($k_1$) and glut ($k_2$). At neutral pH the tripeptide exists primarily in an anionic form with a deprotonated carboxylate group in the glutamyl residue together with a glycine residue in the zwitterionic form (Table 1). The rate constants for $glut^-$ and $glut^{2-}$ are abnormally large for ligands of this charge type.[19] This may be attributed to the remote location of the charges with respect to the metal centre.

Table 4 lists the rate constants, $k_L$, associated (pH 5.5—6.0) with the formation reactions of the ligands cys, $glut^-$, pen, and $Hmea^+$ at several temperatures. Also presented are the formation rate constants for $Hmea^+$ at various ionic strengths ($I$). The decrease in $k_L$ with increasing ionic strength is expected in the reaction of a positively charged ligand with the $[Fe(CN)_5(OH_2)]^{3-}$ ion. Similar changes in $k_L$ with $I$ have been observed for the *N*-methylpyrazinium ion [2] and other cationic ligands.[25] The activation para-

TABLE 4

Formation rate constants and activation parameters for L = cys, glut$^-$, pen, and Hmea$^+$. The ionic strength ($I$) is 0.50 mol dm$^{-3}$ (Na[ClO$_4$]) unless otherwise indicated

| Ligand | $\theta_c$ / °C | $I$ / mol dm$^{-3}$ | $k_L$ / dm$^3$ mol$^{-1}$ s$^{-1}$ | $\Delta H^{\ddagger}$ [a] | $\Delta S^{\ddagger}$ [b] |
|---|---|---|---|---|---|
| Cysteine (cys) | 10.4 | | 83 | 16.6 | 8.8 |
| | 10.9 | | 91 | | |
| | 15.6 | | 128 | | |
| | 20.7 | | 211 | | |
| | 25.0 | | 325 | | |
| Glutathione (glut$^-$) | 11.3 | | 51 | 18.2 | 13 |
| | 15.9 | | 75 | | |
| | 19.1 | | 132 | | |
| | 22.0 | | 172 | | |
| | 25.0 | | 219 | | |
| Penicillamine (pen) | 10.8 | | 104 | 14.1 | 0.4 |
| | 14.8 | | 142 | | |
| | 19.5 | | 221 | | |
| | 25.0 | | 354 | | |
| Mercaptoethylamine (Hmea$^+$) | 12.0 | | 263 | 14.7 | 4.3 |
| | 15.5 | | 367 | | |
| | 19.7 | | 546 | | |
| | 25.0 | 0.010 | 2 310 | | |
| | 25.0 | 0.050 | 1 830 | | |
| | 25.0 | 0.100 | 1 370 | | |
| | 25.0 | 0.250 | 1 120 | | |
| | 25.0 | 0.500 | 859 | | |
| | 25.0 | 1.00 | 610 | | |

[a] ±0.5 kcal mol$^{-1}$. [b] ±2.0 cal K$^{-1}$ mol$^{-1}$.

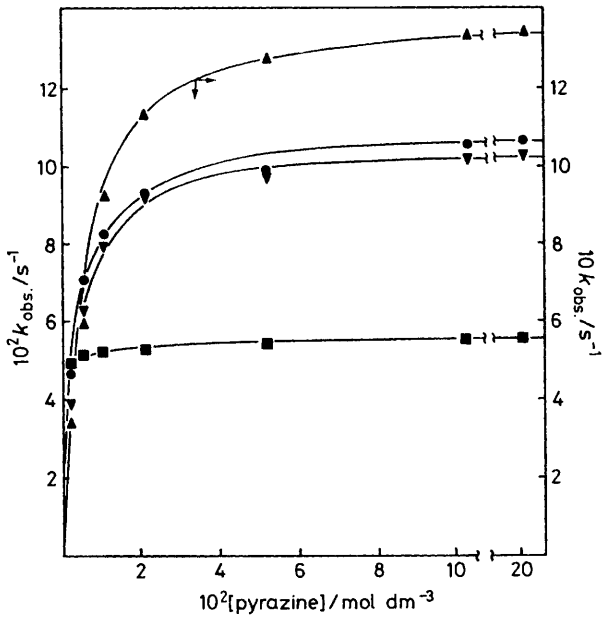



FIGURE 5 Dependence of the observed rate constant for the dissociation of [Fe$^{II}$(CN)$_5$L]$^{n-}$ on pyrazine concentration at 25 °C and pH 5. L = cys (●), glut$^-$ (■), Hmea$^+$ (▼), or pen (▲)

meters for the formation reactions are in agreement with parameters reported for other substitution reactions of [Fe(CN)$_5$(OH$_2$)]$^{3-}$.[2-7]

*Dissociation Reactions.*—Kinetic and thermodynamic evidence from previous studies [2,5,8] suggest that the mechanism for the release of L from [Fe(CN)$_5$L]$^{n-}$ complexes is dissociative in nature [equations (13)—(15)]. In the system

$$[Fe(CN)_5L]^{3-} \overset{k_{-L}}{\rightleftharpoons} [Fe(CN)_5]^{3-} + L \quad (13)$$

$$[Fe(CN)_5]^{3-} + H_2O \rightleftharpoons [Fe(CN)_5(OH_2)]^{3-} \quad (14)$$

$$[Fe(CN)_5]^{3-} + L' \overset{k_{L'}}{\longrightarrow} [Fe(CN)_5L']^{3-} \quad (15)$$

under investigation, L = cys, glut, mea, or pen and L′ is pyrazine. The observed first-order rate constant is given by the expression (16).

$$k_{obs.} = k_{-L}k_{L'}[L']/(k_L[L] + k_{L'}[L']) \quad (16)$$

At high concentrations of pyrazine (0.20 mol dm$^{-3}$) $k_{obs.}$ approaches $k_{-L}$, the limiting rate of dissociation of L from the pentacyanoferrate(II) complex, as shown in Figure 5. The values of $k_{-L}$ for cys, glut$^-$, and Hmea$^+$ are similar to those reported for thiourea and its derivatives ($k_{-L}$ = 5 × 10$^{-2}$—10 × 10$^{-2}$ s$^{-1}$).[24] For penicillamine, however, the corresponding value of 1.34 s$^{-1}$ is one of the largest reported. The dissociation rate constants derived from the intercepts (pH 6) in the complex-formation plots (Figure 2) are in excellent agreement with the more accurately determined values obtained by the competition method.

The rates of dissociation of all the ligands from [Fe$^{II}$(CN)$_5$L]$^{n-}$ were found to be dependent on the pH of the reaction solution. The limiting rate constants, measured over the range pH 2.5—11 at 25 °C, are presented in Table 5. In each case the rate decreases with increasing pH with the most dramatic fall occurring in the pH range 6.5—8.0. The pH profiles for $k_{-L}$ are shown in Figures 6 (cys and pen) and 7 (glut and mea).

A reaction scheme involving HS–R and $^-$SR may be written as in equations (17)—(19) where $K_5$ is the acid

TABLE 5

Variation of $k_{-L}$ with pH for [Fe$^{II}$(CN)$_5$L]$^{n-}$ where L = cys, glut, pen, and mea at 25.0 °C and $I$ = 0.50 mol dm$^{-3}$ (Na[ClO$_4$])

| Ligand | pH | $10^2k_{-L}$/s$^{-1}$ | pH | $10^2k_{-L}$/s$^{-1}$ |
|---|---|---|---|---|
| Cysteine | 2.50 | 10.1 | 7.55 | 5.20 |
| | 3.64 | 10.2 | 7.87 | 3.49 |
| | 4.76 | 10.5 | 8.08 | 1.87 |
| | 5.68 | 9.85 | 8.78 | 1.24 |
| | 6.45 | 8.90 | 9.59 | 0.99 |
| | 7.22 | 6.65 | 10.96 | 0.61 |
| Glutathione | 3.41 | 4.98 | 7.35 | 3.89 |
| | 3.80 | 5.10 | 7.89 | 2.68 |
| | 4.16 | 5.66 | 8.67 | 2.09 |
| | 5.57 | 5.31 | 8.87 | 1.89 |
| | 6.33 | 4.83 | 9.47 | 1.25 |
| | 7.00 | 4.76 | 10.46 | 1.19 |
| | | $k_{-L}$/s$^{-1}$ | | $k_{-L}$/s$^{-1}$ |
| Penicillamine | 3.62 | 1.23 | 7.26 | 0.622 |
| | 4.37 | 1.32 | 7.72 | 0.283 |
| | 5.15 | 1.33 | 8.21 | 0.131 |
| | 5.72 | 1.29 | 8.58 | 0.060 |
| | 6.41 | 1.18 | 9.66 | 0.026 |
| | 6.87 | 0.995 | | |
| | | $10^2k_{-L}$/s$^{-1}$ | | $10^2k_{-L}$/s$^{-1}$ |
| Mercaptoethylamine | 2.45 | 9.90 | 8.06 | 6.22 |
| | 3.80 | 10.3 | 8.43 | 3.46 |
| | 5.50 | 10.1 | 8.80 | 1.67 |
| | 7.00 | 9.30 | 9.72 | 0.96 |
| | 7.20 | 8.98 | 10.65 | 0.42 |
| | 7.59 | 8.20 | | |

$$[Fe(CN)_5(HSR)]^{n-} \overset{K_a}{\rightleftharpoons} [Fe(CN)_5(^-SR)]^{(n+1)-} + H^+ \quad (17)$$

$$[Fe(CN)_5(HSR)]^{n-} \xrightarrow{k_5} [Fe(CN)_5]^{3-} + RSH \quad (18)$$

$$[Fe(CN)_5(^-SR)]^{(n+1)-} \xrightarrow{k_6} [Fe(CN)_5]^{3-} + RS^- \quad (19)$$

dissociation constant of the co-ordinated thiol. The rate

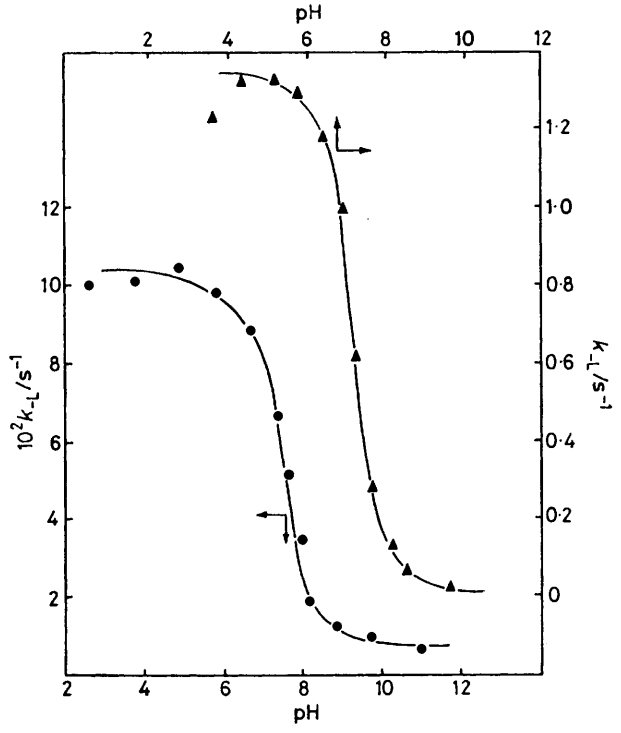



FIGURE 6 pH Profiles of the rate constants, $k_{-L}$, for the dissociation of pentacyanoferrate(II) complexes of cysteine (●) and penicillamine (▲) at 25 °C. The solid lines were calculated from equation (20) using data from Table 7

constant, $k_{-L}$, is of the form (20). Using the limiting rates

$$k_{-L} = (k_5[H^+] + k_6K_5)/([H^+] + K_5) \quad (20)$$

at high and low pH (Figures 6 and 7) it is possible to calculate the $pK_a$ of the co-ordinated thiol. Values are approximate (to 0.2 units) but all four ligands show only a slight decrease in acid-dissociation constants on co-ordination (0.2—0.9 $pK$ units). Using the parameters from Table 6 in equation (20), calculated pH profiles of the dissociation rates were constructed and are shown in Figures 6 and 7. Good

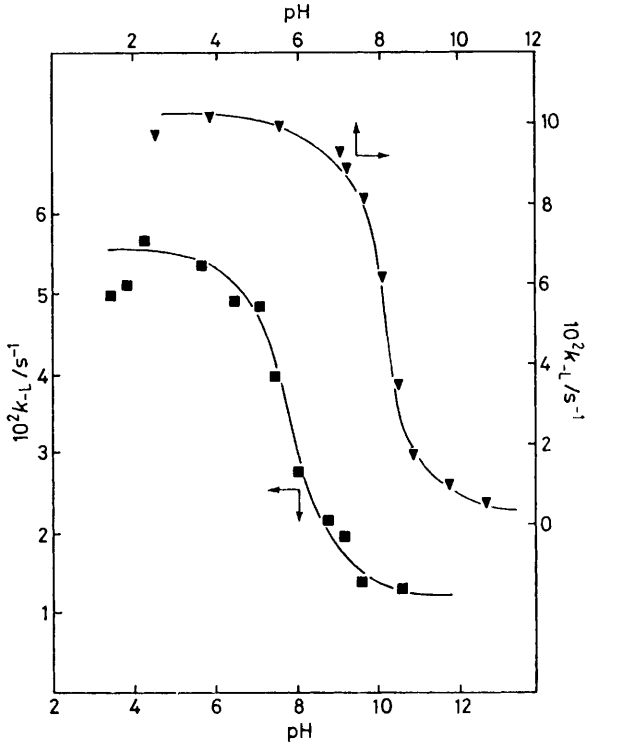



FIGURE 7 pH Profiles of the rate constants, $k_{-L}$, for the dissociation of pentacyanoferrate(II) complexes of glutathione (■) and mercaptoethylamine (▼) at 25 °C. Solid lines as in Figure 6

agreement with the experimental points is observed in each case. Below pH 4 the rate tended to decrease with increasing acidity. At low pH protonation of the carboxyl groups of the ligand and/or the cyanide ligands may occur. Decreased lability of L from the protonated $[HFe(CN)_5L]^{2-}$ has been described above and observed in other cases where L is $H_2O$.[23,24] The limiting dissociation rate constants for the pentacyanoferrate(II) complexes of cys, glut$^-$, pen, and Hmea$^+$ at several temperatures are presented in Table 7 together with the corresponding activation parameters.

TABLE 6

Rate and acid-dissociation parameters for the release of HL and L from pentacyanoferrate(II) complexes

| Ligand [a] | $10^2k_{-L}$/dm$^3$ mol$^{-1}$ s$^{-1}$ $HL^{n+}$ | $10^2k_{-L}$/dm$^3$ mol$^{-1}$ s$^{-1}$ $L^{(n-1)+}$ | $k_{-HL}/k_{-L}$ | $pK_L$ [b] | $pK_{ML}$ [b] | Ref. |
|---|---|---|---|---|---|---|
| cys | 10.4 | 0.90 | 12 | 8.1 | 7.3 | c |
| glut$^-$ | 5.5 | 1.1 | 5.0 | 8.6 | 7.7 | c |
| pen | 134 | 2.0 | 67 | 7.8 | 7.2 | c |
| Hmea$^+$ | 10.2 | 0.50 | 20 | 8.3 | 8.1 | c |
| Hen$^+$ | 1.04 | 0.515 | 2.0 | 9.9 | 10.0 | 8 |
| Hhd$^+$ | 0.53 | 0.41 | 1.3 | 10.8 | 10.8 | 10 |
| Hnic | 0.142 | 0.048 | 3.0 | 3.1 | 3.3 | 9 |
| Hinh | 0.073 | 0.026 | 2.8 | 10.5 | 10.4 | 9 |
| HCN | 0.97 | 0.040 | 24 | | | 11 |
| $HSO_3^-$ | 0.063 | 0.0057 | 11 | | | 11 |

[a] en = Ethylenediamine, hd = hexane-1,6-diamine, Hnic = nicotinic acid, and Hinh = isonicotinohydrazide. [b] L = Free ligand, ML = co-ordinated ligand. [c] This work.

TABLE 7

Dissociation rate constants and activation parameters for $[Fe^{II}(CN)_5L]^{n-}$ where L = cys, glut$^-$, pen, and Hmea$^+$ and $I$ = 0.50 mol dm$^{-3}$ ($Na[ClO_4]$)

| Ligand | $\theta_c$/°C | $10^2k_{-L}$/s$^{-1}$ | $\Delta H^{\ddagger}$ [a] | $\Delta S^{\ddagger}$ [b] |
|---|---|---|---|---|
| Cysteine | 8.6 | 1.38 | 18.3 | −1.6 |
| (pH 5.2) | 11.4 | 3.62 | | |
| | 15.9 | 5.39 | | |
| | 20.3 | 7.89 | | |
| | 25.0 | 10.5 | | |
| Glutathione | 11.2 | 1.33 | 18.2 | −3.5 |
| (pH 7.0) | 14.7 | 1.75 | | |
| | 19.1 | 2.62 | | |
| | 25.0 | 4.76 | | |
| | 29.2 | 7.69 | | |
| Penicillamine | 8.6 | 15.0 | 21.9 | 15.6 |
| (pH 4.8) | 15.1 | 37.5 | | |
| | 20.2 | 76.2 | | |
| | 25.0 | 133 | | |
| | 30.4 | 231 | | |
| Mercaptoethylamine | 11.3 | 1.98 | 20.2 | 4.3 |
| (pH 7.0) | 14.7 | 2.83 | | |
| | 19.1 | 4.98 | | |
| | 25.0 | 9.30 | | |
| | 29.2 | 15.3 | | |

[a] ±0.5 kcal mol$^{-1}$. [b] ±2 cal K$^{-1}$ mol$^{-1}$.

## DISCUSSION

The variation in the rate constants in Table 3 indicates that the rate of substitution of $[Fe(CN)_5(OH_2)]^{3-}$ ion is dependent on the charge of the entering ligand. This dependence on ligand charge and the insensitivity of the rate to variations in the ligand type (*N- vs. S*-donor, *etc.*) is consistent with an ion-pair dissociative mechanism,[26] equations (21)—(23) where $k_L = K_{os}k_o$. The rate-

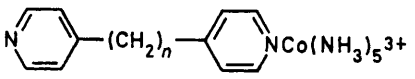
$$[Fe(CN)_5(OH_2)]^{3-} + L^{n+} \overset{K_{os}}{\rightleftharpoons} [Fe(CN)_5(OH_2), L]^{(3-n)-} \quad (21)$$

$$[Fe(CN)_5(OH_2), L]^{(3-n)-} \xrightarrow{k_o} [Fe(CN)_5, L]^{(3-n)-} + H_2O \quad (22)$$

$$[Fe(CN)_5, L]^{(3-n)-} \xrightarrow{fast} [Fe(CN)_5L]^{(3-n)-} \quad (23)$$

determining loss of co-ordinated water is preceded by an ion-pair formation where $K_{os}$ is dependent on the charge of L and the ionic strength of the reaction medium. The rate constant $k_L$ may be shown to be related to the charge product, $Z_MZ_L$,[27] and a reasonable correlation between $Z_MZ_L$ and log $k_L$ has been observed for these and other ligands. The data are, however, only approximate owing to uncertainties in the calculated values of the outer-sphere ion-pair association constant and to the accumulation of data at differing $I$. Deviations from the correlation occur for anionic ligands such as histidinate ($k$ = 91 dm$^3$ mol$^{-1}$ s$^{-1}$) [6] and glut$^-$ ($k$ = 220 dm$^3$ mol$^{-1}$ s$^{-1}$) which exhibit higher values than those normally observed for simple anions (*e.g.* for $CN^-$, $k$ = 38 dm$^3$ mol$^{-1}$ s$^{-1}$).[28] The rate constants for these ligands are close to the range for neutral species (200—400 dm$^3$ mol$^{-1}$ s$^{-1}$) suggesting that the negative charge of the carboxylate group has little effect on the rate when located in a position remote from the future co-ordination site. Similar results were obtained by Cassatt and Wilkins [29] in the reaction of the nickel(II) ion with amino-acids and peptides. A more extreme example of this deviation is seen with the ligand $[(NC)_5Fe(pyz)Ru(NH_3)_4(pyz)]^-$ where the formation rate constant is 1 190 dm$^3$ mol$^{-1}$ s$^{-1}$ ($I$ = 0.20 mol dm$^{-3}$).[25] The $[Fe(CN)_5]^{3-}$ moiety only partially reduces the charge on the ruthenium centre which is closer to the eventual co-ordination site (pyrazine).

This phenomenon is also evident in the variation of rate constants [30] for a series of bipyridyl ligands (L), see below: when $n$ = 0, $k$ = 5 500; $n$ = 1, $k$ = 3 900;




L

$n$ = 2, $k$ = 4 500; and $n$ = 3, $k$ = 2 200 dm$^3$ mol$^{-1}$ s$^{-1}$. These observations illustrate the importance of the distribution of charges in the entering ligand on the determination of the rate of substitution of $[Fe(CN)_5(OH_2)]^{3-}$. In the present study, at higher pH values there is a marked decrease in formation rate with all of the thiol ligands. In the range pH 7—9 the $RS^-$ ion will be formed and on electrostatic grounds lower reaction rates might be expected.

In the dissociation reactions of $[Fe^{II}(CN)_5L]^{n-}$ the nature of the leaving ligand has been shown to be of major importance in the rate of release of L. The effect of protonation (at a more remote site in the ligand) on the dissociation rate has been reported for a series of terminal aliphatic diamines.[10] As the $NH_3^+$ group becomes further separated from the $[Fe(CN)_5]^{3-}$ centre, $k_{-HL}$ decreases and the difference between $k_{-HL}$ and $k_{-L}$ diminishes. Table 6 lists parameters for ethylenediamine and hexane-1,6-diamine. Similar values of $k_{-HL}/k_{-L}$ were observed in the cases of nicotinic acid and isonicotinohydrazide.[9] The results of the present study indicate a similar trend with ratios of the rate constants in the same order. It is also of interest to note that p$K_a$ values for co-ordinated and free ligands are closely similar for both sulphur- and nitrogen-donor systems.

An interesting result from this study is the very large value of $k_{-L}$ for penicillamine (1.34 s$^{-1}$). The two methyl groups on the carbon adjacent to sulphur in the ligand have a steric destabilizing effect on the $[Fe(CN)_5(pen)]^{3-}$ complex. The decreased stability is evident in the equilibrium constant (2.61 × 10$^2$ dm$^3$ mol$^{-1}$) compared to that of the cysteine complex (3.17 × 10$^3$ dm$^3$ mol$^{-1}$), in which the two methyl groups are replaced by hydrogen atoms. The thermodynamic parameters indicate that these steric effects reduce the stability of the penicillamine complex by *ca.* 1.6 kcal mol$^{-1}$.* Similar effects have been observed in pentacyanoferrate(II) complexes of histidine, methylimidazoles, and cyanopyridines.

The penicillamine complex exhibits a peak in the visible region at a lower energy than found for the other three complexes. A relationship between the rates of

* Throughout this paper: 1 cal = 4.184 J.

dissociation of ligands from pentacyanoferrate(II) complexes and their $^1A_1$–$^1E(1)$ *d–d* transitions has been demonstrated by Toma *et al.*[6] This correlation may be used to predict the stability of various $[Fe(CN)_5L]^{n-}$ complexes, based on the position of L in the spectrochemical series. The lower energy of the *d–d* transition in the penicillamine complex compared to those for the other ligands is reflected in the larger rate of dissociation.

The decrease in the rate of ligand release as the pH is raised is accompanied by a shift in the $\lambda_{max.}$ of $[Fe^{II}(CN)_5L]^{n-}$. At pH 9.5 the visible peak has moved 10—20 nm higher in energy relative to that observed at pH 5, corresponding to an increase in the stability of the complex.

One of us (D. H. M.) thanks the University of Victoria for a Graduate Fellowship. This work was supported by NSERC (Canada).

[0/1311 *Received*, 20*th August*, 1980]

REFERENCES

[1] H. Sigel (editor), ' Metal Ions in Biological Systems,' Marcel Dekker, New York, 1979, vol. 9.
[2] H. E. Toma and J. M. Malin, *Inorg. Chem.*, 1973, **12**, 2080.
[3] N. E. Katz, M. A. Blesa, J. A. Olabe, and P. J. Aymonino, *Inorg. Chim. Acta*, 1978, **27**, L65.
[4] H. E. Toma, J. M. Malin, and E. Giesbrecht, *Inorg. Chem.*, 1973, **12**, 2084.
[5] D. H. Macartney and A. McAuley, *Inorg. Chem.*, 1980, **20**, 748.
[6] H. E. Toma, J. M. Martins, and E. Giesbrecht, *J. Chem. Soc., Dalton Trans.*, 1978, 1610.
[7] M. A. Blesa, I. A. Funai, P. J. Morando, J. A. Olabe, P. J. Aymonino, and G. Ellenreider, *J. Chem. Soc., Dalton Trans.*, 1977, 845.
[8] M. A. Blesa, J. A. Olabe, and P. J. Aymonino, *J. Chem. Soc., Dalton Trans.*, 1976, 1196.
[9] M. A. Blesa, I. A. Funai, P. J. Morando, and J. A. Olabe, *J. Chem. Soc., Dalton Trans.*, 1977, 2092.
[10] N. E. Katz, M. A. Blesa, J. A. Olabe, and P. J. Aymonino, *J. Chem. Soc., Dalton Trans.*, 1978, 1603.
[11] J. Legros, *J. Chim. Phys. Phys.-Chim. Biol.*, 1964, **61**, 909, 923.
[12] G. Brauer, ' Handbook of Preparative Inorganic Chemistry,' 2nd edn., Academic Press, New York, 1965, vol. 2, p. 1511.
[13] A. R. Garafalo and G. Davies, *Inorg. Chem.*, 1976, **15**, 1787.
[14] K. J. Ellis and A. McAuley, *J. Chem. Soc., Dalton Trans.*, 1973, 1533.
[15] A. E. Martell and R. M. Smith, ' Critical Stability Constants,' Plenum Press, New York, 1974, vol. 1.
[16] R. M. Smith and A. E. Martell, ' Critical Stability Constants,' Plenum Press, New York, 1975, vol. 2.
[17] D. H. Macartney and A. McAuley, *Can. J. Chem.*, 1981, **59**, 132.
[18] D. H. Macartney and A. McAuley, unpublished work.
[19] N. C. Li and R. A. Manning, *J. Am. Chem. Soc.*, 1955, **77**, 2895.
[20] M. Friedman, J. F. Cavins, and J. S. Wall, *J. Am. Chem. Soc.*, 1965, **87**, 3672.
[21] R. D. Graham, D. R. Williams, and P. A. Yeo, *J. Chem. Soc., Perkin Trans. 2*, 1972, 1876.
[22] D. L. Rabenstein, *J. Am. Chem. Soc.*, 1973, **95**, 2797.
[23] J. M. Malin and R. C. Koch, *Inorg. Chem.*, 1978, **17**, 752.
[24] D. H. Macartney and A. McAuley, *Inorg. Chem.*, 1979, **18**, 2891.
[25] H. E. Toma, *J. Coord. Chem.*, 1978, **7**, 231.
[26] M. Eigen, *Z. Phys. Chem.* (*Frankfurt*), 1954, **1**, 176.
[27] K. Kustin and J. Swinehart, *Prog. Inorg. Chem.*, 1970, **13**, 107.
[28] Z. Bradić, M. Pribanić, and S. Ašperger, *J. Chem. Soc., Dalton Trans.*, 1975, 353.
[29] J. C. Cassatt and R. G. Wilkins, *J. Am. Chem. Soc.*, 1968, **90**, 6045.
[30] J. Jwo, P. L. Gaus, and A. Haim, *J. Am. Chem. Soc.*, 1979, **101**, 6189.