Short Communication

Ionophoretic studies on mixed complexes

Metal–nitrilotriacetate–penicillamine system

B. B. TIWARI*, R. K. P. SINGH and K. L. YADAVA

Electrochemical Laboratories, Department of Chemistry, University of Allahabad, Allahabad-211 002 (India)

(First received April 3rd, 1990; revised manuscript received November 29th, 1990)

ABSTRACT

   A method involving the use of paper electrophoresis is described for the study of equilibria in mixed-ligand complex systems in solution. The technique is based on the movement of a spot of metal ion under an electric field with the complexants added to the background electrolyte at pH 8.5. The stability constants of the complexes were found to be 5.28, 5.15 and 4.97 (log K values) for Hg(II), Be(II) and Ni(II) complexes, respectively, at $\mu = 0.1$ and 35°C.

INTRODUCTION

   The properties and chemical reactions of naturally occurring D-penicillamine have already been subjected to wide investigation. The most important reactions are processes involving participation of the mercapto group, and the main biochemical aspects have been reviewed by Jocelyn [1]. The complex chemical significance of these amino acids is determined by the mercapto sulphur donor atom, which is very soft in character. Great biological importance is attached to the study of metal–sulphur bonds formed in such processes, primarily in the non-haeme iron proteins [2] and the blue copper proteins [3]. The significance of this amino acid is enhanced by the fact that it displays independent therapeutic activity. Its most valuable use is for the treatment of Wilson's disease, caused by accumulation of copper. As a consequence of its property of forming stable complexes, it can also be employed to advantage for the elimination of other heavy metals (*e.g*, Pb, Hg) from the organism. It has been utilized in connection with rheumatoid arthritis and neonatal jaundice. Sorensen [4] has demonstrated the anti-inflammatory activity of copper—D-penicillamine.
   Czakis-Sulikowska [5] studied the formation of mixed halide complexes of Hg(II). We previously described [6,7] a method for the study of mixed complexes. This

work is an extension of the technique and reports our observations on mixed systems, *viz.*, Hg(II)–, Be(II)– and Ni(II)–nitrilotriacetate (NTA)–penicillamine.

EXPERIMENTAL

Instruments

A Systronics Model 604 electrophoresis equipment system was used, which has a built-in power supply (a.c.–d.c.) that is fed directly to paper electrophoretic tanks. In order to maintain the temperature constant, two hollow metallic plates coated with thin plastic paper on the outer surface were used for sandwiching paper strips and thermostated water (35°C) was circulated through them. pH meaurements were made with an Elico Model L_{1-10} pH meter using-glass electrode.

Chemicals

Mercury(II), beryllium(II) and nickel(II) perchlorate solutions were prepared by precipitating the corresponding carbonates from solutions of the nitrates (analytical-reagent grade) with sodium hydrogencarbonate, washing the precipitates thoroughly with boiling water and treating them with a suitable amount of 1% perchloric acid. The resulting mixtures were heated to boiling on a water-bath and then filtered. The solutions were standardized and diluted to $5.0 \cdot 10^{-3}$ M.

Metal spots were detected on the paper using hydrogen sulphide [for Hg(II)], aluminon [for Be(II)] and dimethylglyoxime [for Ni(II)]. Silver nitrate in alkaline acetone was used for glucose.

Background electrolyte

The background electrolyte in the study of binary complexes consisted of 0.1 M perchloric acid and $1.0 \cdot 10^{-2}$ M penicillamine. It was maintained at pH 8.5 by addition of sodium hydroxide. Each solution was standardized as usual. Stock solutions of 9.0 M perchloric acid, 2.0 M sodium hydroxide and 0.5 M penicillamine were prepared from AnalaR reagents (BDH, Poole, U.K.). A 0.01 M nitrolotriacetic acid solution was prepared from a sample obtained from E. Merck (Darmstadt, Germany).

Procedure

The hollow base plate in the instrument was made horizontal with a spirit level. A 150-ml volume of background electrolyte was placed in each tank of the electrophoretic apparatus. Paper strips (Whatman No. 1, 30 × 1 cm) in duplicate were spotted with metal ions and glucose in the centre with a micropipette and were subsequently placed on the base plate and sandwiched under the upper hollow metallic plate with the ends of the strips immersed in the tank solutions on both sides. Then a potential difference of 200 V was applied between the tank solutions and electrophoresis was carried out for 60 min. Subsequently the strips were removed and the spots were detected. The averages for duplicate strips were noted for calculations and movement of the glucose spot was used as a correction factor. The mobilities were calculated by dividing the movement by the potential gradient and expressed in cm^2 V^{-1} min^{-1}.

RESULTS AND DISCUSSION

Metal–penicillamine binary system

The plot of ionophoretic mobility of a metal spot against pH gives a curve with a number of plateaus as shown in Fig. 1. A plateau is obviously an indication of a pH range where the speed is virtually constant and indicates the formation of a certain complex species. The first corresponds to a region in which metal ions are uncomplexed. Fig. 1 reveals a second plateau in each instance with zero mobility, indicating the formation of neutral 1:1 complexes. With a further increase in pH, the mobility decreases, giving rise to a third plateau in a negative region, indicating an anionic nature of the metal complex. Prominent liganding properties of unprotonated anionic species of penicillamine ruling out the involvement of a zwitterion have been reported [8]. Further increase in pH has no effect on the mobility of metal ions.

In view of the above observations, the complexation of metal ions with the penicillamine anion $[L^{2-}]$ may be represented by

$$M^{2+} + L^{2-} \underset{K_2}{\overset{K_1}{\rightleftharpoons}} ML \tag{1}$$

$$ML + L^{2-} \rightleftharpoons ML_2^{2-} \tag{2}$$

where M represents Ni, Be and Hg and ML and ML_2^{2-} are their complexes with penicillamine.

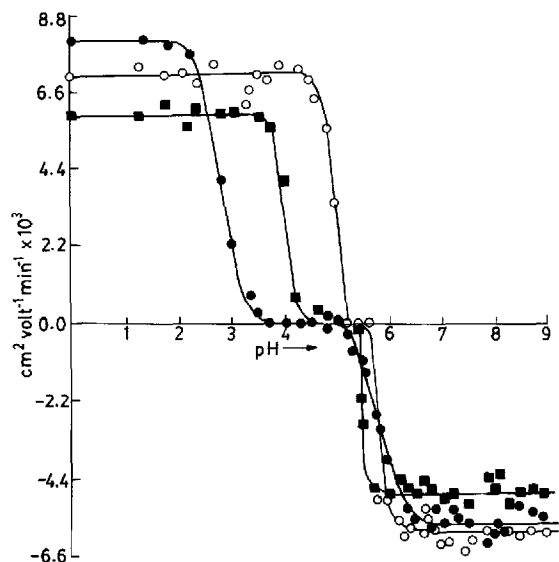The metal spot on the paper is thus a conglomeration of uncomplexed metal

Fig. 1. Mobility curve for M–penicillamine systems. $\bigcirc$ = Ni(II)–penicillamine; $\blacksquare$ = Be(II)–penicillamine; $\bullet$ = Hg(II)–penicillamine.

ions, a 1:1 complex and a 1:2 complex. This spot is moving under the influence of the electric field, and its overall mobility, U, can be represented by the equation

$$U = \sum_n u_n f_n \tag{3}$$

where u_n and f_n are the mobility and mole fraction, respectively, of particular complex species. On taking into consideration the different equilibria, this equation is transformed into

$$U = \frac{u_0 + u_1 K_1 \ [L^{2-}] + u_2 K_1 K_2 \ [L^{2-}]^2}{1 + K_1 \ [L^{2-}] + K_1 K_2 \ [L^{2-}]^2} \tag{4}$$

where u_0, u_1 and u_2 are mobility of the uncomplexed metal ion, the 1:1 metal complex and the 1:2 metal complex respectively.

Eqn. 4 was used for the determination of the stability constants of metal ions with penicillamine. For calculating the first stability constant, K_1, the region between the first and second plateaus is pertinent. The overall mobility U will be equal to the arithmetic mean of the mobility of the uncomplexed metal ion, u_0, and that of the first complex, u_1, at a pH where, $K_1 = 1/[L^{2-}]$. With the help of the dissociation constants of penicillamine ($k_1 = 10^{1.90}$, $k_2 = 10^{7.85}$, $k_3 = {}^{10.55}$) [9,10], shown in Fig. 2, the concentration of the penicillamine anion $[L^{2-}]$ is determined for the pH using the following equation, from which K_1 can be calculated:

$$[L^{2-}] = \frac{[L_T]}{1 + \dfrac{[H]}{k_3} + \dfrac{[H]^2}{k_2 k_3} + \dfrac{[H]^3}{k_1 k_2 k_3}} \tag{5}$$

where $[L_T]$ = total concentration.

The stability constant of the second complex, K_2 can be calculated by taking into consideration the region between the second and third plateaus of the mobility curve. These calculated values are given in Table I.
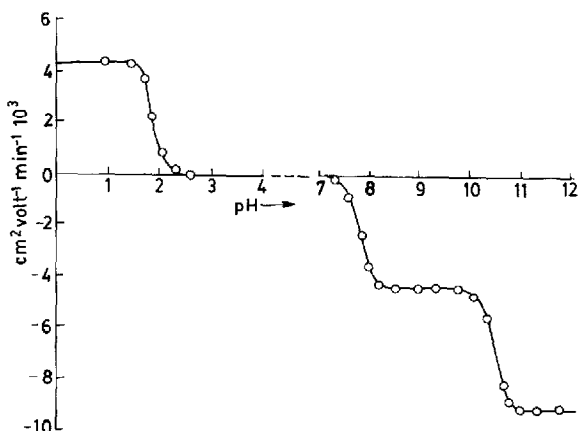

Fig. 2. Mobility curve for H^+–penicillamine.

TABLE 1

STABILITY CONSTANTS OF SOME BINARY AND TERNARY COMPLEXES OF Hg(II), Be(II) AND Ni(II)

Ionic strength, $\mu = 0.1$; temperature $= 35°C$. NTA anion $= N(CH_2COO)_3^{3-}$. Penicillamine anion $= (CH_3)_2C(S^-)CH(NH_2)COO^-$.

Metal ions	Calculated values of stability constants[a]				Literature values of stability constants[a]			
	Log $K_{1_{ML}}^M$	Log $K_{2_{ML_2}}^M$	Log K_{M-NTA}^M	Log $K_{M-NTA-L}^{M-NTA}$	Log $K_{1_{ML}}^M$	Log $K_{2_{ML_2}}^M$	Log K_{M-NTA}^M	Log $K_{M-NTA-L}^{M-NTA}$
Hg(II)	14.95	23.85	13.30	5.28	17.50[10] 16.15[10] 16.10[11]	23.5[10]	14.6[12]	— —
Be(II)	12.50	22.70	7.10	5.15	—	—	7.10[11] 7.11[13]	—
Ni(II)	10.53	19.43	11.49	4.97	11.40[10] 11.11[10] 10.75[10] 10.63[9] 10.63[11] 10.75[11]	22.30[10] 21.79[10] 22.89[10] 23.02[9] 23.37[11] —	10.80[12] 11.90[12]	—

[a] $K_{1_{ML}}^M = \dfrac{[ML]}{[M][L]}$; $K_{2_{ML_2}}^M = \dfrac{[ML_2]}{[ML][L]}$; $K_{M-NTA}^M = \dfrac{[M-NTA]}{[M][NTA]}$; $K_{M-NTA-L}^{M-NTA} = \dfrac{[M-NTA-L]}{[M-NTA][L]}$.

Metal–NTA–binary system

The overall mobilities of metal spots in the presence of NTA at different pH values are shown in Fig. 3. The mobility of the last plateau in the case of Hg(II), Be(II) and Ni(II) is negative, showing the anionic nature of Hg(II)–, Be(II)– and Ni(II)–NTA complexes. Hence only one NTA anion, $(NTA)^{-3}$, is assumed to combine with metal ions to give 1:1 complexes, which is in conformity with the findings of others [14]. The stability constants of complexes with NTA were calculated as described in the preceding paragraph. The calculated values are given in Table 1.

Metal–NTA–penicillamine ternary system

The study of this system was made at pH 8.5. It was observed from the mobility curves for the metal–penicillamine and M–NTA binary systems that binary complexes are formed at a pH < 8.5. Hence it was considered appropriate to study the transformation of M–NTA complexes into M–NTA–penicillamine complexes at pH. 8.5 in order to avoid any side interactions.

The plot of mobility against logarithm of concentration of added penicillamine gives a curve (Fig. 4) containing two plateaus, one at the beginning and the other at the end. The mobility of the range of the first plateau corresponds to the mobilities of 1:1 M–NTA complexes, as also observed in the study of M–NTA complexes. The mobility of the last plateau is more negative than the mobility of a pure M–NTA complex. Further, as the mobility of the last plateau does not agree with the mobility of 1:1 and 1:2 metal–penicillamine complexes (observed in the study of binary M–NTA systems),
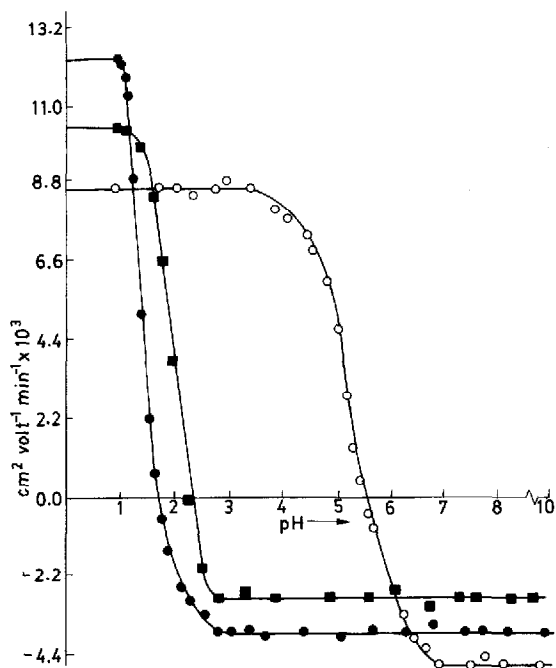

Fig. 3. Mobility curve for M–NTA systems. ■ = Ni(II)–NTA; ● = Hg(II)–NTA; ○ = Be(II)–NTA.
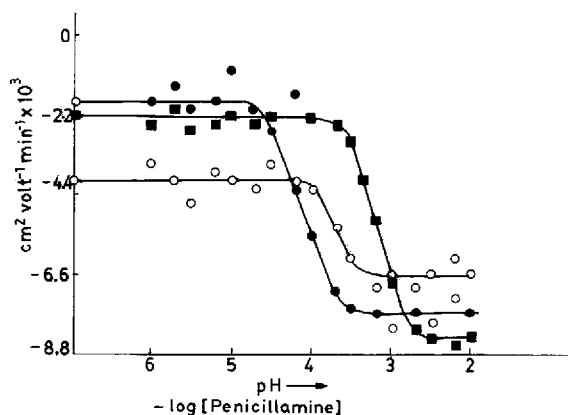
Fig. 4. Mobility curves for M–NTA–penicillamine systems. $\bigcirc$ = Be(II)–NTA–penicillamine; $\bullet$ = Hg(II)–NTA–penicillamine; $\blacksquare$ = Ni(II)–NTA–penicillamine.

it is inferred that the moiety in the last plateau is due to coordination of the penicillamine anion with a 1:1 M–NTA moiety, resulting in the formation of a 1:1:1 M–NTA–penicillamine mixed complex:

$$M\text{–}NTA^- + L^{2-} \underset{}{\overset{K'}{\rightleftharpoons}} M\text{–}NTA\text{–}L^{3-} \tag{6}$$

In the present electrophoretic study, the transformation of a simple complex into a mixed complex took place, the overall mobility being given by

$$U = u_0 f_{\text{M-NTA}} + u_1 f_{\text{M-NTA-L}} \tag{7}$$

where u_0, u_1 and $f_{\text{M-NTA}}, f_{\text{M-NTA-L}}$ are the mobilities and mole fractions of the M–NTA and M–NTA–L complexes, respectively. The above on adding the values of mole fractions, this equation becomes

$$U = \frac{u_0 + u_1 K' \, [L^{2-}]}{1 + K' \, [L^{2-}]} \tag{8}$$

where u_0 and u_1 are the mobilities in the regions of the two plateaus of the curve. The concentration of penicillamine at which the overall mobility is the mean of the mobilities of the two plateaus is determined from Fig. 4. The concentration of the penicillamine anion at pH 8.5 for this penicillamine concentration was calculated. K' is obviously equal to $1/[L^{2-}]$. All these values of K' are given in Table I.

REFERENCES

1 C. P. Jocelyn, *Biochemistry of the SH Group*, Academic Press, London and New York, 1972.
2 R. G. Moore and P. J. R. Williams, *Coord. Chem. Rev.*, 18 (1976) 125.
3 A. J. Fee, *Struct. Bonding*, 23 (1975).
4 R. J. Sorensen, *J. Med. Chem.*, 19 (1976) 135.

5 M. Czakis-Sulikowska, *Zesz. Nauk, Politech. Lodz, Chem.*, (1965) 23; *C.A.*, 65 (1966) 1746e.
6 P. C. Yadava, A. K. Ghose, K. L. Yadava and A. K. Dey, *J. Chromatogr.*, 9 (1976) 416.
7 J. R. Yadava, J. K. Sirkar and K. L. Yadava, *Electrochim. Acta*, 26 (1981) 391.
8 J. R. Blackburn and M. M. Jones, *J. Inorg. Nucl. Chem.*, 35 (1973) 1605.
9 D. D. Perrin, *Stability Constants of Metal Ion Complexes, Part B (Organic Ligands) (IUPAC Chemical Series*, No. 22), Pergamon Press, Oxford, 1979.
10 A. E. Martell and R. M. Smith, *Critical Stability Constants, Vol. 1 (Amino Acids)*, Plenum Press, New York and London, 1977.
11 L. G. Sillen and A. E. Martell, *Stability Constants of Metal Ion Complexes (Special Publication*, No. 17), Chemical Society, London, 1964.
12 D. D. Perrin, *Stability Constants of Metal Ion Complexes, Part B (Organic Ligands) (IUPAC Chemical Series*, No. 22), Pergamon Press, Oxford, 1979, p. 417.
13 A. E. Martell and R. M. Smith, *Critical Stability Constants, Vol. 1 (Amino Acids)*, Plenum Press, New York and London, 1977, p. 139.
14 N. G. Elenkova and R. A. Tesoneva, *J. Inorg. Nucl. Chem.*, 35 (1973) 841.