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STUDIES ON MERCURY(II), NICKEL(II) AND LEAD(II) BIOLOGICALLY IMPORTANT BINARY COMPLEXES WITH α-AMINOBUTYRIC ACID IN SOLUTION

Brij Bhushan Tewari[1*]

ABSTRACT

Complexation reactions of α -aminobutyric acid with mercury(II), nickel(II) and lead(II) have been studied in solution phase using a paper ionophoretic technique. This method is based on movement of a spot of metal ion in an electric field at various pHs of background electrolyte. A plot of pH versus mobility was used to obtain information for binary complexes and to calculate its stability constants. The stability constants of the ML^+ and ML_2 binary complexes of metal(II) α-aminobutyric acid have been found to be (8.59 ± 0.01; 6.93 ± 0.05), (6.83 ± 0.03; 5.38 ± 0.07) and (4.42 ± 0.02; 2.72 ± 0.03) (logarithm stability constant values) for the mercury(II), nickel(II) and lead(II) complexes, respectively at ionic strength 0.1 M ($HClO_4$) and a temperature of 35° C.

Key words: Ionophoretic technique, mercury(II) complexes, nickel(II) complexes, lead(II) complexes,α-aminobutyric acid, stability constant.

INTRODUCTION

For a mononuclear binary complex, if a central atom (central group) M (the 'metal') and a ligand L have been defined, then in the following expressions K_n, is the stepwise formation constant, and β_n is the cumulative formation constant for the complex ML_n. They can both be referred to as stability constants (stepwise and cumulative)[1].

$$K_n = K\ (ML_{n-1} + L = ML_n)$$

$$\beta_n = K\ (M + nL = ML_n)$$

Data on the complexation of biologically significant metal ions and bioactive ligands α-aminobutyric acid give insight into many physicochemical processes. Nickel classified as beneficial metal and mercury as well as lead as toxic metal, respectively. Mercury(II), nickel(II) and lead(II) are well known for their medicinal applications and toxicity[2-7]. α-Aminobutyric acid ($C_4H_9NO_2$) is a naturally occurring amino acid, which do not occur in proteins. It is found in animal and plant tissues. It has significant application in biological systems[8-10].

The paper electrophoretic technique usually suffers from a number of defects. Temperature during electrophoresis, capillary flow on paper, electro-osmosis and adsorption affect the mobility of charged moieties[11]. The present technique is almost free from these destroying factors and very convenient in use. It gives results in fair agreement with accepted literature values.

Communications[12, 13] from our laboratory described a new method for the study of metal complexes. A search of the literature indicated few reports on Ni(II) α-aminobutyric acid

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[1*] Department of Chemistry, Faculty of Natural Sciences, University of Guyana, PO Box: 101110, Georgetown, Guyana, Tel: 592-222-4926, Fax: 592-222- 3596 , E-mail: brijtew@yahoo.com

binary complexes and no report on Hg(II) / Pb(II) α-aminobutyric acid complexes. In view of this, an attempt was made to establish the optimum conditions for metal(II) α-aminobutyric acid complex formation. In addition, the present paper describes a paper electrophoretic method for the determination of the stability constants of these complexes.

EXPERIMENTAL

Instruments

A Systronics (Naroda, India) paper electrophoresis equipment horizontal – cum vertical type, model 604, has been used. The apparatus consisted of a PVC moulded double tank vessel. In our laboratory significant change to the instrument has been made. Two hollow rectangular plates covered with thin poly (vinyl chloride) sheets have been used through which thermostated water is run for controlling the temperature. The tanks were closed with a transparent PVC moulded lid. The whole assembly is tight, which prevents moisture changes, which may upset the equilibria in a paper strip. This assembly design thus keeps to a minimum the disturbing effects of evaporation from the unwanted liquid flow in the paper. Each electrolyte tank contains a separate electrode chamber. The auxiliary unit is specially designed to operate either in voltage mode or in current mode.

An Elico (Hyderabad, India) model $L_{1\text{-}10}$ having glass and calomel electrodes assembly and working on 220 Volts/50 Hz established a. c. mains, was employed for pH measurements.

Chemicals

Mercury(II), nickel(II) and lead(II) perchlorate solutions were prepared by preliminary precipitation of metal carbonates from a 0.1 M solution of sodium carbonate (AnalaR grade, BDH, Poole, UK). The precipitates were thoroughly washed with boiling water and treated with calculated amounts of 1 % perchloric acid. The resulting mixture was heated to boiling on a water bath and then filtered. The metal content of the filtrates were determined and final concentration was kept at 0.005 M[14, 15]. The position of the Ni^{2+} spots on the paper at the end of the experiment was detected using ammonical dimethylglyoxime (DMG), that of Pb^{2+} detected by 0.1 % solution of 1-(2-pyridylazo) – 2- naphthol (PAN) (Merck, Darmstadt, Germany) in ethanol, that of Hg^{2+} detected using hydrogen sulphide in water. The 0.005 M glucose (BDH, AnalaR) solution was prepared in water and used as an indicator for the correction due to electroosmosis. A saturated aqueous solution (0.9 mL) of silver nitrate was diluted with acetone to 20 mL. Glucose was detected by spraying with this silver nitrate solution and then with 2 % ethanolic solution of sodium hydroxide, when a black spot was formed.

Background electrolyte(BGE)

Stock solution of 5.0 M perchloric acid was prepared from its 70 % solution (SDS, AnalaR grade). 2.0 M sodium hydroxide and 0.5 M α-aminobutyric acid (BDH, Poole, UK) solutions were prepared. The background electrolyte used in the study of binary complexes were 0.1 M perchloric acid and 0.1 M α-aminobutyric acid. The system was maintained at various pHs by the addition of sodium hydroxide.

Procedure

Whatman No. 1 filter paper for chromatography was used for the purpose of electrophoresis. For recording observation of particular metal ion, two strips were spotted with the metal ion solution along with additional two spotted with glucose using 1.0 l pipette and then mounted on the insulated plate. Each of the two electrolyte vessels were filled with 150 ml of BGE solutions containing 0.1 M perchloric acid and 0.01 M α-aminobutyric acid. The paper became moistened with the background electrolyte solutions due to diffusion. The second insulated plate was placed on paper strips and then thermostated water (35° C) was circulated into the plates to keep the temperature constant. The lid was then placed on the instrument to make it air tight. It was left for 10 minutes to insure wetting of strips. Subsequently a direct 240 volts potential was applied between the electrodes. Electrophoresis was carried for 60 minutes after which these strips were removed from the tank and dried. The metal ion and glucose spots were detected by specific reagents. The leading and tailing edge were measured from the marked centre point and the mean were taken. The distance moved by glucose was subtracted (in case of migration toward anode) to obtain correct path length. Migration towards anode and cathode were designated by negative and positive signs, respectively.

Electrophoretic observations on metal ions were recorded at various pH values of the background electrolyte, the ionic strength being maintained at 0.1 M. The observed mobility of migrant was calculated by using the formula.

$$U = \frac{d}{X \cdot t}$$

After applying the correction factor the observed mobility is given as

$$U = \frac{d \pm d_G}{X \cdot t}$$

where U = mobility of metal ion / complex ion; d = mean of duplicate distance travelled by metal ion / complex ion; d_G = mean of duplicate distance travelled by glucose spot; X = field strength (7.5 V/cm); t = time for electrophoresis. The speed of the metal ions / complex ions are reported with pH values. A plot of mobility against pH curve for metal(II) α-aminobutyric acid is shown in figure 1.

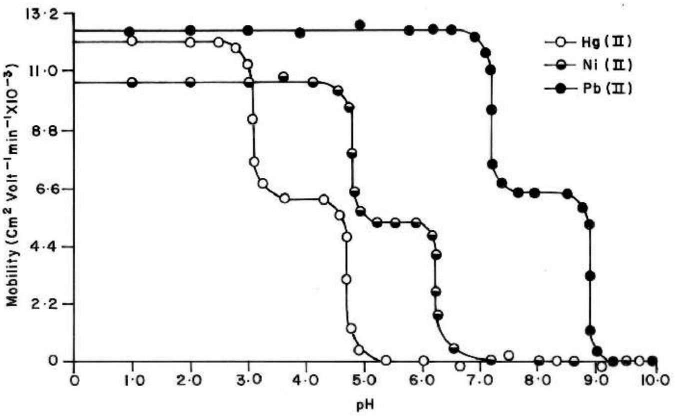


Figure 1. Mobility curve for the metal(II) – α-aminobutyric acid systems. —○— = Hg(II) – α-aminobutyric acid; —⊖— = Ni(II) – α -aminobutyric acid, —●— = Pb(II) – α-aminobutyric acid. Background electrolyte = 0.1 M perchloric acid and 0.01 M α-aminobutyric acid. pH was maintained by addition of sodium hydroxide. Concentration of Hg^{2+}, Ni^{2+} and Pb^{2+} = 0.005 M. Ionic strength = 0.1 M. Temperature = 35° C. The paper strips were spotted with 0.1µl of sample solutions and glucose (for making osmotic corrections).

RESULTS

The plot of overall electrophoretic speed of metal spot against pH gives a curve with a number of plateaus (figure 1). A constant speed over a range of pH is possible only when a particular complex species is overwhelmingly formed. Thus every plateau is indicative of formation of a certain complex species. The first one in the beginning corresponds to a region in which metal ions are uncomplexed. This region of low pH, concentration of CH_3 CH_2 CH (NH_3^+) COOH species of α-aminobutyric acid is maximum and this species is non-complexing. Beyond this range, metal ion spots have progressively decreasing mobility, complexation of metal ions should be taking place with anionic species of α-aminobutyric acid whose concentration increases progressively with the increase of pH. Figure 1 shows three plateaus with Hg(II), Ni(II) and Pb(II) metal ions. Hence all three metal ions form two complexes with α-aminobutyric acid anion. It is therefore assumed that the anionic species CH_3 CH_2 CH (NH_2) COO^- of α-aminobutyric acid has complexed with the metal ions to form different complexes.

Figure 1 reveals that Hg^{2+}, Ni^{2+} and Pb^{2+} metal ions form their first complex movements towards negative electrode. Hence, one CH_3 CH_2 CH (NH_2) COO^- must have combined with Hg^{2+}, Ni^{2+} and Pb^{2+} metal ions to give $[Hg\ \{CH_3\ CH_2\ CH\ (NH_2)\ COO\}]^+$, $[Ni\ \{CH_3\ CH_2\ CH\ (NH_2)\ COO\}]^+$ and $[Pb\{CH_3\ CH_2\ CH\ (NH_2)\ COO\}]^+$ complex cations, respectively. With further increase of pH, mobility in all three metal ions decreases giving rise to third plateau with zero mobility indicates its neutral nature.

The third plateau in each case is due to (1:2) metal – ligand complex. Hence, two CH_3 CH_2 CH (NH_2) COO^- must have combined with Hg^{2+}, Ni^{2+} and Pb^{2+} to give $[Hg\ \{CH_3\ CH_2\ CH\ (NH_2)\ COO\}_2]$, $[Ni\ \{CH_3\ CH_2\ CH\ (NH_2)\ COO\}_2]$ and $[Pb\{CH_3\ CH_2\ CH\ (NH_2)\ COO\}_2]$ neutral metal complexes. In view of the above observation, the complexation of metal ions with α-aminobutyric acid anion may be represented as:

$$M^{2+} + L^- \overset{K_1}{\rightleftarrows} ML^+ \qquad (1)$$

$$ML^+ + L^- \overset{K_2}{\rightleftarrows} ML_2 \qquad (2)$$

where M^{2+} is Hg^{2+} , Ni^{2+} and Pb^{2+} metal cation; $[L^-]$ is the α-aminobutyric acid anion; K_1 and K_2 first and second stability constants, respectively.

The metal spot on the paper is thus a combination of uncomplexed metal ions, 1:1 and 1:2 metal complexes. The spot is moving under the influence of electric field and the overall mobility U is given by equation (3)[16].

$$U = \frac{\begin{array}{c} u_{0,0}\ \beta_{0,0} + u_{1,0}\ \beta_{1,0}\ [L] + u_{2,0}\ \beta_{2,0}\ [L]^2 + \text{--} \\ + u_{1,1}\ \beta_{1,1}\ [HL] + u_{2,1}\ \beta_{2,1}\ [HL]^2 + \text{--} \\ + u_{2,1}\ \beta_{2,1}\ [H_2L] + u_{2,2}\ \beta_{2,2}\ [H_2L]^2 + \text{--} \end{array}}{\begin{array}{c} \beta_{0,0} + \beta_{1,0}\ [L] + \beta_{2,0}\ [L]^2 + \beta_{1,1}\ [HL] \\ + \beta_{2,1}\ [HL]^2 + \beta_{2,1}\ [H_2L] \\ + \beta_{2,2}\ [H_2L]^2 + \text{---------} \end{array}} \qquad (3)$$

where $u_{0,0}$ equals speed of uncomplexed metal ion, $u_{1,0}$ equals speed of complex formed by the combination of one unprotonated anionic ligand with metal ion and $u_{x\cdot p}$ equals speed of the metal complex formed by the combination of x anions containing, p, protons each. β's equals overall stability constant of the different metal complexes formed in the interaction. On taking into consideration different equilibrium above equation transformed into following useful form

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

where u_0, u_1 and u_2 equals mobilities of uncomplexed metal ion, 1:1 metal complex and 1:2 metal complex, respectively. Equation (4) has been used for calculating stability constant of complexes metal ions with α-aminobutyric acid. For calculating first stability constant, K_1, the region between first and second plateau is pertinent. The overall mobility, U, will be equal to the arithmetic mean of mobility of uncomplexed metal ion, u_0 and that of the first complex u_1, at a pH where $K_1 = 1/ CH_3 CH_2 CH (NH_2) COO^-$. The dissociation constant of pure α-aminobutyric acid ($K_1 = 10^{2.30}$; $K_2 = 10^{9.63}$) is determined from same paper electrophoretic technique. The dissociation of pure α-aminobutyric acid can be represented as:

$$CH_3 CH_2 CH (NH_3^+) COOH$$

$$K_1 \downarrow\uparrow -H^+$$

$$CH_3 CH_2 CH (NH_3^+) COO^-$$

$$K_2 \downarrow\uparrow -H^+$$

$$CH_3 CH_2 CH (NH_2) COO^-$$

With the help of dissociation constants, the concentration of α-aminobutyric acid anion $[L^-]$ is determined for the pH, using following equation.

$$[L^-] = \frac{[L_T]}{1 + [H] / K_1 + [H]^2 / K_1 K_2} \qquad (5)$$

where $[L_T]$ equals total concentration of α-aminobutyric acid (0.01 M); K_1 and K_2 equals first and second dissociation constants of pure α-aminobutyric acid. By knowing the concentration of α-aminobutyric acid anion $[L^-]$, K_1 can be calculated ($K_1 = 1/[L^-]$). The stability constant, K_2 of second complex can be calculated by taking into consideration, the region between second and third plateau of mobility curve. These calculated values are given in table 1.

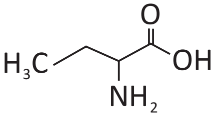
Table 1. Stability constants of binary complexes of Hg(II), Ni(II) and Pb(II) with α-aminobutyric acid.

Metal ion	Complexes	Stability constant	Logarithm stability Constant value *
Mercury(II)	ML^+	K_1	
	ML_2	K_2	8.59 ± 0.01
Nickel(II)	ML^+	K_1	6.93 ± 0.05
			6.83 ± 0.03
	ML_2	K_2	(5.37 ± 0.02)[19]
			5.38 ± 0.07
			(4.41 ± 0.04)[19]
Lead(II)	ML^+	K_1	4.42 ± 0.02
	ML_2	K_2	2.72 ± 0.03

Ionic strength = 0.1 M; temperature = 35° C; M = metal cations (Hg^{2+}, Ni^{2+}, Pb^{2+}); ligand = -aminobutyric acid;α -aminobutyric acid anion = $CH_3CH_2CH(NH_2)COO^-$
*Literature values are given in parentheses.

DISCUSSION

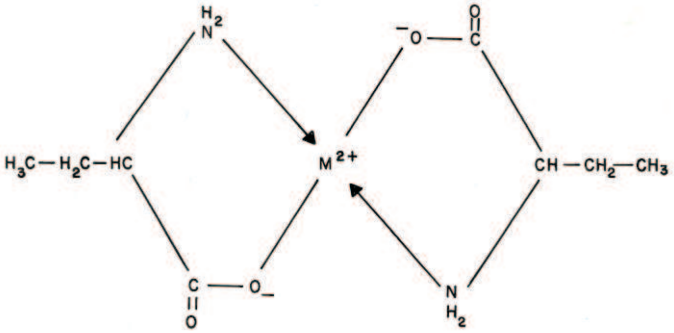
It is observed from table 1 that stability constants are approximately similar to literature values. The slight deviation in the values obtained from different sources is mainly due to the difference in temperature, ionic strength and experimental conditions used by different workers. The stability constants metal complexes can be very easily calculated by this technique. The present technique is limited to charged species and the precision of the method is not as high as other physicochemical methods. However, uncertainty in the result is ± 5 %. It is not felt that it can replace the most reliable methods although it is a new approach worth developing. The molecular structure of α-aminobutyric acid is given below.



It is also observed from table 1 that first and second stability constants of ML and ML_2 complexes followed the order.

mercury(II) > nickel(II) > lead(II)

The values of K_2 are found to be lower in comparison in K_1 in each case, this may be due to the decrease in coordinating tendency of ligand with higher state of aggregation[17,18]. High stability constant values of the mercury(II) α-aminobutyric acid complexes indicate strong bonding between the mercury(II) cation and the α-aminobutyric acid anion. Whilst low stability constant values of lead(II) α-aminobutyric acid complexes indicate weak bonding between the lead(II) cation and -aminobutyric acid anion. The proposed structure for the ML_2 complexes may be given as:



CONCLUDING REMARKS

Following conclusions can be shown from the present study.

- Mercury(II), nickel(II) and lead(II) are significant for biological systems but as such they are toxic, the α-aminobutyric acid may be used to reduce the level of these metal ions in the biological systems.
- Mercury(II) α -aminobutyric acid and lead (II) α-aminobutyric acid couples are found to have maximum and minimum stability constant values, respectively.
- ML_2 binary complexes are found to have low stability constant values and less stable in comparison to ML^+ complexes.
- The present ionophoretic technique is very helpful in finding that complex system is formed or not, and if formed its stability constants can also be determined.
- Stability constants of metal complexes can be very easily calculated by this technique, of the present paper electrophoretic technique has significant advantages over the other physicochemical methods reported in chemical literature for the determination of stability constants of metal complexes.

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