

STUDIES ON COMPLEXATION IN SOLUTION WITH A PAPER ELECTROPHORETIC TECHNIQUE [THE SYSTEM Cr(III), Cu (II), Ni(II) and Zn(II) -ALIZARIN – NITRILOTRIACETIC ACID]

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ABSTRACT

In this work, the complexation of a biologically active ligand alizarin and nitrilotriacetic acid with Cu(II), Zn(II), Ni(II), and Cr(III) metal is reported in the solution phase using paper electrophoretic technique to determine the corresponding stability constant of the complexes. This method is based on the movement of a spot of metal ion in an electric field with the complexants added in the background electrolyte at pH 8.5. The concentration of primary ligand alizarin was kept constant, while that of the secondary ligand NTA was varied. A plot of log[NTA] versus mobility was used to obtain information on the mixed complexes and to calculate stability constants. The stability constant of mixed complexes of metal (II)/(III)-alizarin-NTA have been found to be $10^{5.40}$, $10^{5.10}$, $10^{5.00}$ and $10^{4.70}$ for Cr(III), Cu (II), Ni(II) and Zn(II) respectively at 35°C and ionic strength $I=0.1 \text{ mole dm}^{-3}$ (HClO₄).

Keywords: Binary and ternary metal complexes, Overall mobility, paper electrophoresis, stability constants, metal- alizarin system.

I.INTRODUCTION

The study of mixed ligand complexes has received a great importance because of their wide applications in various fields of chemical activity and more particularly because of their presence in biological systems ^[1]. Due to growing interest of metal complexes in textile industry, alizarin is taken into account along with NTA, in present study. Alizarin, which consists of 1,2-dihydroxyanthraquinone is the common madder of anthraquinoid pigment group. It has been used for textiles, food, chemical analysis and medicine ^{[2] - [3]}. The color of alizarin is light brown and dissociates low in water. It is the root of Rubiatictorum plant ^[6]. Alizarin and its derivatives form stable metal complexes ^{[7] - [8]}. Transition metals are particularly suitable for this purpose because they can adopt a wide variety of coordination number, geometries, and oxidation states in comparison with other main group elements. Moreover, transition elements chosen i.e. Cu(II), Zn(II), Ni(II) and Cr(III) are more or less biologically essential elements and more than a dozen biological activities depend on them without which life

cannot be sustained. ^{[9] - [14]}. On reviewing the literature, it is proved that metal complexes exhibit greater biological activity than original compound ^[15]. In order to form new metal complexes which would be helpful for scientist and researchers in the field of medicine, biological fields, industries etc., the complexes must fulfill some requirements like water solubility, thermodynamic stability etc. In this context, stability constants were determined in given work and comparison was drawn between different mixed metal ligand complexes. Traditionally, in order to study the complexation of ligand with metallic cations, potentiometry, paper electrophoresis, UV-Vis. and mass spectroscopy can be used. ^{[16] - [18]} In this study, paper electrophoresis was chosen because of its simplicity, low cost, and availability in most laboratories. The factors affecting the mobility of ions like variation in temperature during the experiment, capillary flow on paper, electro osmosis and adsorption are the usual drawbacks of paper electrophoretic technique. But the technique here is almost free from these factors.

Kiso ^[19] has done a comprehensive study on paper electrophoretic migration of metal complexes. Few reports are available on metal-alizarin complexes, but no reports are available on metal-alizarin-NTA mixed complexes. In view of this, an attempt was made to establish the optimum conditions for metal(II)/(III)-alizarin-NTA complex formation.

II. EXPERIMENTAL

2.1 Instruments:

An electrophoretic apparatus (systronics type 604, India) was used together with the various accessories supplied with the instrument. The apparatus consisted of polyvinyl chloride of the cassette type and chromatogram could be run simultaneously on eight paper strips. The power unit capable of yielding a maximum of 300V. In each case electrophoresis was carried out for 1 hr. at 220V and at 35°C. Whatman no.1 paper strips were used with an applied voltage gradient of 9V Cm⁻¹. pH measurements were made with Elico digital pH meter having glass electrode assembly working on 220 volts/ 50 cycles stabilized AC main was used.

2.2 Chemicals

Zn(II), Cu(II), Ni(II) and Cr(III) perchlorates were prepared by precipitating the corresponding carbonates from solutions of nitrate (Anala R grade) with the solution of sodium carbonate, washing the precipitates thoroughly with boiling water and dissolving in a suitable amount of perchloric acid. The resulting solutions were heated to boiling on a water bath and then filtered. The solutions were standardized and diluted with distilled water. As required Anala R/ BDH grade alizarin, NaOH, HClO₄, NTA and specific color developing reagents were used for different metal ions of binary and ternary system sets for specific color development. 1-(2- pyridylazo)-2 naphthol(PAN), 0.1% (w/v) in ethanol was used for detecting all the metal ions. A saturated aqueous solution (0.9ml) of silver nitrate was diluted with acetone to 20ml. Glucose was detected by spraying with this solution and then with 2% ethanolic sodium hydroxide, forming a black spot.



2.3 Background electrolyte

Stock solution of 5.0 M perchloric acid was prepared by its 70% solution (Anala R grade) 2.0M sodium hydroxide (Anala R grade), 0.5M alizarin and 0.5M NTA (AnalaR grade) were prepared. Each solution was standardized as usual.

The background electrolytes used in the study of binary complexes were 0.1M perchloric acid and 0.01M alizarin.

For the study of mixed systems, the background electrolyte used consisted of 0.1M perchloric acid, 0.01M alizarin and varying amounts of 0.01M NTA. The mixed system was maintained at pH 8.5 by the addition of sodium hydroxide.

2.4 Procedure

The midpoint of each paper was marked and moistened with the background electrolyte. Excess electrolyte was allowed to drain off and papers were mounted on the plate of electrophoretic chamber. A spot of the metal solution was then applied at the marked point with the help of a fine glass capillary. For recording observations of particular metal ion two strips were spotted with the metal ion solution along with additional two spotted with glucose. The electrode compartment was filled with the equal volumes of the desired background electrolyte solutions. The lid of the chamber was then placed; it was left for 10 minutes to ensure wetting of strips. Subsequently a direct 220 volts' potential was applied between the electrodes. Electrophoresis was carried out for 60 minutes after which these strips were removed from the tanks with a glass rod 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 the migration towards anode) to obtain correct path length. Migration towards anode and cathode were designated by negative and positive signs respectively. The actual distance of the sample spot was measured after taking into account the distance travelled by the reference glucose spot. The distance recorded in the duplicated differed within $\pm 5\%$ and the average of two distances was noted. The mobilities were then calculated and expressed in $10^3 \text{cm}^2 \text{v}^{-1} \text{min}^{-1}$.

2.4.1. Mixed Complexes.

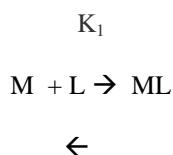
Paper strips in duplicate were marked with metal ions along with an additional two marked with glucose. After soaking the paper strips with the BGE, electrophoresis was carried out for 1 h at 220 V potential differences as in the case of binary complexes. For subsequent observations, NTA solution, maintained at pH 8.5 with NaOH, was recorded. The mobilities of metal ions were recorded versus $\log[\text{NTA}]$ and is shown in Figure3.

III RESULTS AND DISCUSSION

3.1 Metal(II/III)-alizarin Binary System.



The ionophoretic mobility of the metal spot against pH gives a curve with a number of plateaus as is shown in 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 corresponds to a region in which metal ions are uncomplexed. In this region of low pH, the concentration of the alizarin is at a maximum, and this species is non complexing. Beyond this range, metal ion spots have progressively decreasing mobility, and complexation of metal ions should be taking place with anionic species of alizarin whose concentration increases progressively with an increase of pH. Figure 1 shows two plateaus in all four Zn(II), Ni(II), Cu(II) and Cr(III), metal cations with the alizarin anion. It is therefore assumed that the anionic species alizarin has complexed with the metal ions to form different complexes. Figure.1 discloses that Cu(II), Ni(II), Zn(II), and Cr(III) ions form their first complex movement toward the negative electrode. With a further increase of pH, mobility in case of all the four metal ions decreases giving rise to a second plateau with zero mobility that indicates its neutral nature. The second plateau in each case is due to a (1:1) metal-ligand complex. A further increase of pH has no effect on the mobility of metal ions, which indicates no further interaction between the metal ions and ligands. It is significant that these studies give clear evidence of the complexation of the anionic species of alizarin with metal ions forming binary complexes of 1:1 in composition. In general, the complexation of metal ions with alizarin anion may be represented in general as



The metal ion moving under the influence of electrical field, the overall mobility is given by the equation.

$$u = \sum u_n f_n$$

Where, u_n and f_n are mobility and mole fraction of a particular complex species. This equation is transformed into the following form. On taking into consideration of different equilibria.

$$u = \frac{u_0 + u_1 K_1 [L] + u_2 K_1 K_2 [L]^2 + \dots}{1 + K_1 [L] + K_1 K_2 [L]^2 + \dots}$$

Where K 's ($K_1 K_2$) are the stability constants of complexes and $[L]$ is concentration of alizarin. u 's (u_0, u_1, u_2) are the ionic mobilities of different species of the metal ions, which can be accessed from the plateaus of the Fig1. In the region between first and second plateau, the system contains, mixture of free metal ion and 1:1 complex. The existence of 1:2 complex can be excluded and hence the third term in the numerator and the denominator of the above equation can be neglected. U would be equal to $(u_0 + u_{1/2})$ provided $K_1 [L] = 1$.

The pH corresponding to the average value of u_0 and u_1 is found from the fig.1 and with the knowledge of the dissociation constant of alizarin (pKa 6.94), the concentration of alizarin ion at this pH is calculated. Its reciprocal gives the stability constant K_1 of the 1:1 complex



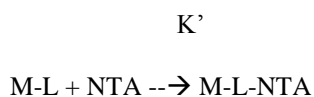
The concentration of chelating alizarin anion [L] is determined as

$$[L] = \frac{[L]_{tot}}{1 + [H]/K_1 + [H]^2/K_1 K_2}$$

Where $[L]_{tot}$ is the total concentration of the ligand alizarin (0.01M) and K_1 and K_2 are first and second dissociation constants of the alizarin respectively.

3.2 Metal(II/III)-alizarin-NTA Mixed System

Mixed complexes are formed due to attachment of two different ligating species to a central metal ion. As follows from the mobility curve of the metal(II)-alizarin complexes that are formed at $pH < 8.5$. It was therefore considered necessary to study the transformation of metal(II)-alizarin binary complexes into metal(II)-alizarin-NTA mixed complexes at $pH 8.5$ to avoid any side interaction. The plot of mobility against log of concentration of added NTA gives a curve (Figure 3) containing two plateaus, one in the beginning and the other at the end. The mobility in the range of the first plateau corresponds with the mobility of the metal(II)-alizarin complex. Further, the mobility in the last plateau does not coincide with the mobility of 1:1 metal(II)-alizarin complexes. It is thought that the mobility in the last plateau is due to coordination of the NTA anion to the 1:1 metal(II)-alizarin complex resulting in the formation of a 1:1:1 metal(II)-alizarin-NTA mixed complex as



For M-L-NTA complexes the stability constant K' is calculated by using modified equation.

$$u = \frac{u_0 + u_1 K' [NTA]}{1 + K' [NTA]}$$

Here u_0 and u_1 are the mobilities of M-L and M-L-NTA complexes. From the fig.3 concentration of NTA at which overall mobility as the mean of the mobilities of the two plateaus is determined by using the equation.

$$[L] = \frac{[L]_{tot}}{1 + [H]/K_1}$$

The concentration of nitrilotriacetic acid anion at $pH 8.5$ is calculated. K' is obviously equal to $1/[NTA]$.



IV. FIGURES AND TABLES

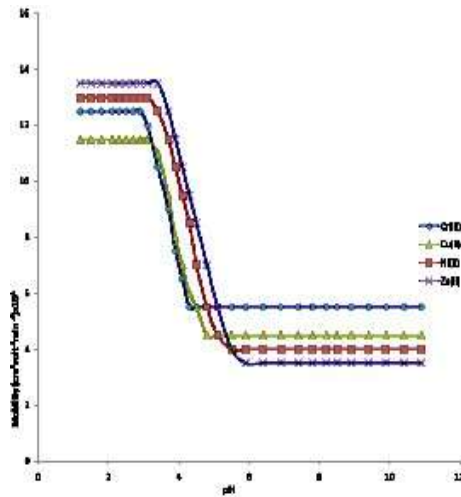


Fig. 1 Mobility curves for metal(II)/metal(III)- alizarin systems = Cr(III) -aliz -- =Cu(II)-alizarin; - - - - = Ni(II) -alizarin; --♦-- =Zn(II)-alizarin. The paper strips were spotted with 0.1 μl of sample solution and glucose (for making osmotic correction)

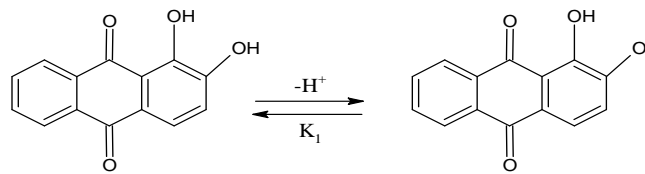


Figure 2. Mode of dissociation of pure alizarin.

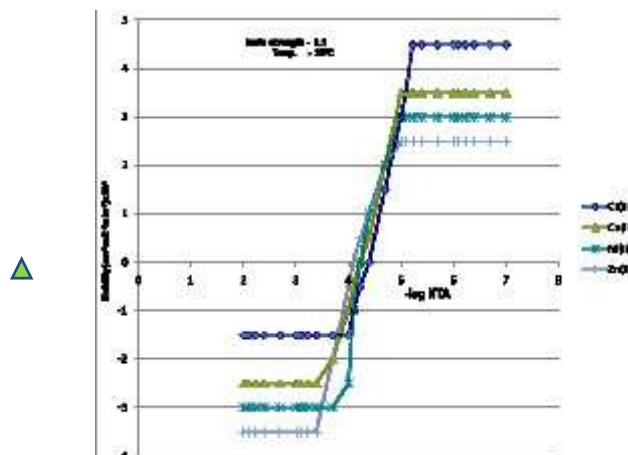


Fig.3 ♦ =Cr(III)-alz-NTA; ▼ =Cu(II)-alizarin-NTA; —x— = Ni(II) – alizarin -NTA;

—+—Zn(II)-alizarin- NTA The background electrolyte = 0.1M perchloric acid,

0.01M alizarin and varying amounts of 0.01M NTA

. The mixed system was maintained at pH 8.5

by the addition of sodium hydroxide.

TABLE. 1

Stability Constants of Binary Complexes of Cr(III), Cu(II), Ni(II) and Zn(II) with Alizarin and Mixed Complexes of Cr(III), Cu(II), Ni(II) and Zn(II) with alizarin and Nitrilotriacetic acid

Temp. 35⁰C Ionic Strength – 0.1M (HClO₄)

metal ions.	Complexes	Stability constants	Calculated Values	Literature values
Cr(III)	Cr(III)-Alz 1:1	K	5.1405	-
	Cr(III)-Alz-NTA 1:1:1	K ₁	5.3979	-
Cu(II)	Cu(II)-Alz 1:1	K	4.8407	5.1 ^[20]
	Cu(II)-Alz-NTA 1:1:1	K ₁	5.0969	-
Ni(II)	Ni(II)-Alz 1:1	K	4.4415	4.1 ^[20]
	Ni(II) 1:1:1	K ₁	5.0000	-
Zn(II)	Zn(II)-Alz 1:1	K	4.1433	-
	Zn(II)-Alz-NTA 1:1:1	K ₁	4.6990	-

The calculated values of K₁ are given in table 1. It is observed from Tables 1 and 2 that the order of stability constants, viz., $Cr^{+++} > Cu^{++} > Ni^{++} > Zn^{++}$ is same for metal(II/III)-Alizarin, metal(II/III)- nitrilotriacetic acid binary, and metal(II)- alizarin-nitrilotriacetic acid ternary complexes, but the corresponding stability constant values are higher in the mixed complexes. Therefore, the mixed complexes are considered to be more stable than binary complexes of metal cations with alizarin and nitrilotriacetic acid.^[21] It is also observed from Tables 1 and 2 that the stability constant values 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. No comparison can be made for the stability constant values of mixed ligand complexes because they are reported for the first time.

TABLE. 2

Table 2. Stability Constants of Binary Complexes of Chromium(III), Copper(II), Nickel(II) and Zinc(II) with Nitrilotriacetic Acid

Temp. 35°C Ionic Strength – 0.1M (HClO₄)

Metal ions	Complex 1:1	Stability const.	Calculated values	Lit. val .
Cr(III)	Cr(III)-NTA	K	4.5	-
Cu(II)	Cu (II)-NTA	K	3.5	-
Ni(II)	Ni(II)-NTA	K	3.0	-
Zn(II)	Zn(II)-NTA	K	2.5	-

V. CONCLUSION

Metal complexes of biologically active ligand alizarin have several applications including textile industries and medical domain where chelators are applied as detoxifiers to cure metal poisoning or as markers for special examinations. Furthermore, it could be shown that the dark storage of samples was crucial for the achievement of useful results. It reflects that a new strategy or new route can be developed to synthesize new metal complexes for diverse applications. If new chelators should be used it will be very helpful to have information about their one of the very important chemical property called stability constant of the metal ligand complex.

The present paper technique is limited to charged species, yet it is an alternate, and simple technique to be used.

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