## Separation of Inorganic lons by Paper Electrophoresis

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Manuscript received 14 March 1990, revised 3 April 1991. accepted 31 May 1991

**E**LECTROPHORETIC studies of cations have been extensively studied<sup>1</sup> using various electrolyte systems. Most of the studies use simple complexing agents<sup>2</sup>. These studies, however, lead to the possibility of successful separation of binary, ternary or quaternary mixture of cations. We report the electrophoretic migration of 22 cations using complexing agents such as ethylenediamine (en), diethylamine (den) and triethylamine (ten) in potassium hydrogen pthalate buffer system at pH 4.

## Experimental

Chromatographic paper (Whatman-1;  $20 \times 20$  cm) and a Gelman Delux electrophoretic chamber was used for electrophoretic measurements. Constant voltage was supplied by a regulated power supply equipped with timer. An Eil 7405 pH meter was used. Uv lamp (Camage CJ-3600) was used for the detection of spot.

Buffer solution of pH 4 was prepared by mixing potassium hydrogen pthalate and hydrogen chloride. Solutions (0.1 M) of cations were prepared by dissolving their respective chloride salts in distilled water; antimony, bismuth, lead and silver were used as their nitrate salts. The complexing agent

TABLE I - MOVEMENT OF IONS IN ELECTROLYTE SYSTEM							
Cation	Buffer pH $4 \pm 0.1$ Vol. en =4.8 ml dm <sup>-8</sup> T=1h, V=200V Dist. travelled cm	Mobility µ	Buffer pH 4±0.1 Vol. den=5ml dm <sup>-3</sup> T=1h, V=200V Dist. travelled cm	Mobility µ	Buffer pH $4\pm0.1$ Vol. ten = 5 ml dm <sup>-3</sup> T=1h, V=200V Dist. travelled cm	Mobility µ	
Ag <sup>I</sup> Hgili Biuli Cobiv Solili Solili Cobiv Soli Soli Soli Soli Soli Soli Soli Soli	$1 \ 6 \pm 0.5 \\ 6.2 \pm 0.1 \\ 3.1 \pm 0.4 \\ 0.0 \pm 0.0 \\ 5.4 \pm 0.9 \\ 4.3 \pm 0.2 \\ 2.2 \pm 0.9 \\ 0.0 \pm 0.0 \\ 2.1 \pm 0.6 \\ 0 \ 0 \pm 0.0 \\ 5.8 \pm 0.3 \\ 3.6 \pm 0.8 \\ 4.2 \pm 0.2 \\ 4.55 \pm 0.45 \\ 5.1 \pm 0.4^{\circ} \\ 5.1 \pm 0.4^{\circ} \\ 5.1 \pm 0.4^{\circ} \\ 3.6 \pm 0.3 \\ 3.33 \pm 0.2 \\ \end{bmatrix}$	$\begin{array}{c} 4.44\\ 17.22\\ 8.61\\ 0.0\\ 15.13\\ 11.94\\ 6.11\\ 0.0\\ 0\ 0\\ 5.83\\ 0\ 0\\ 16.10\\ 9.63\\ 11.66\\ 12.63\\ 14.16\\ 13.05\\ 10.00\\ 10.27\\ \end{array}$	$1.2 \pm 0.3$ $2.1 \pm 0.4$ $0.0 \pm 0.0$ $0.0 \pm 0.0$ $1.0 \pm 0.5$ $2.8 \pm 0.4$ $0.0 \pm 0.0$ $0.0 \pm 0.0$ $0.0 \pm 0.0$ $0.0 \pm 0.0$ $2.3 \pm 0.3$ $2.1 \pm 0.2$ $3.9 \pm 0.1$ $4.1 \pm 0.5$ $4.2 \pm 0.3$ $3.7 \pm 0.2$ $5.0 \pm 0.4$ $5.1 \pm 0.2$ $5.7 \pm 0.3$ $5.8 \pm 0.2$ $5.0 \pm 0.6i$	3.33 5.93 0.0 2.77 7.77 0.0 0.0 0.0 6 38 5.83 10.83 11.38 11.66 10.27 13.88 14.16 15.83 16.10 13.88	$\begin{array}{c} 0.7 \pm 0.2 \\ 2.1 \pm 0.3 \\ 0.0 \pm 0.0 \\ 0.0 \pm 0.0 \\ 1.2 \pm 0.4 \\ 4.1 \pm 0.3 \\ 0.0 \pm 0.0 \\ 0.0 \pm 0.0 \\ 0.0 \pm 0.0 \\ 2.2 \pm 0.4 \\ 2.3 \pm 0.6 \\ 3.8 \pm 0.1 \\ 3.8 \pm 0.1 \\ 3.8 \pm 0.1 \\ 3.8 \pm 0.1 \\ 3.5 \pm 0.1 \\ 5.8 \pm 0.5 \\ 5.9 \pm 0.6 \\ 6.0 \pm 0.4 \\ 5.7 \pm 0.6 \\ 2.7 \pm 0.5 \end{array}$	$\begin{array}{c} 1.94\\ 5.83\\ 0.0\\ 0.0\\ 3.33\\ 11.38\\ 0.0\\ 0.0\\ 0.0\\ 6.11\\ 6.38\\ 10.55\\ 10.55\\ 11.94\\ 9.72\\ 16\\ 11\\ 16.38\\ 16.66\\ 15.83\\ 7.50\\ \end{array}$	
Na <sup>+</sup> K <sup>+</sup>	3.35±0.2 3.7±0.2 3.8±0.3	0.0 10.27 10.55	5.0±0.5 4.0±0.5 4.1±0.5	13.88 11.11 11 <b>.</b> 38	2.0±0.4 2.2±0.4	5.55 6.11	

TABLE 1-MOVEMENT OF	Ions in	ELECTROLYTE SYSTEM
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in an appropriate amount was then added directly into the buffer solution to get the required concentration.

General method for determination: Buffer solution (400 ml) was added in each compartment of electrophoretic chamber. Chromatographic sheet was first soaked in the buffer-complexing agent mixture and then loaded in the chamber. The sample solution  $(1 \ \mu 1)$  was applied on the sheet with the help of microlitre syringe. The chamber was covered with the glass lid and the constant voltage was supplied for appropriate time. The sheet was then removed, immediately dried and the zone developed with the specific locating reagent/visualised under uv lamp. Colour of the spot was then compared with the standard. The electrophoretic mobility  $(\mu)$  was then calculated by measuring the pathlength.

## **Results and Discussion**

Electrophoretic behaviour of cations on paper in the presence of complexing agents have been investigated to explore an electrolyte system which could be used for their separation in mixtures. Since, electrophoretic migration and stability of the complexes are dependent on the pH of the medium, in order to minimise the expected complications, mixture of complexing agent and buffer is used as electrolyte system instead of the routine procedure of preparing the complexes first and then subjecting the complexes to electrophoresis in the same electrolyte system. The solution of amines, namely en, den and ten have been used as the complexing agents. The experiments were run at pH 4 with

varying field strengths 5, 7.5, 10, 12.5 and 15 V  $cm^{-1}$ .

It was observed that at low field strength zones were broader than at higher field strength. The evaporation rate of the electrolyte system was found to be faster at field strength greater than  $12.5 \text{ V cm}^{-1}$ which caused many complications. Therefore, field strength of 10 V cm<sup>-1</sup> was selected for all studies. Preliminary studies using electrolyte systems such as en+H<sub>2</sub>O, en+HCl (0.1 N)-buffer of pH 4 or 0.1 N HCl indicated that zones under these conditions were elongated. Elongation or tailing was more in the system without en. It was also found that shape of the zones was nearly symmetrical and comparatively of lesser diameter in en+buffer system.

The concentration of 2.0-9.7 ml dm<sup>-3</sup> en was used in the samples run to standardise the conditions. Elongation of zone was observed after electromigration was carried out at concentration greater than 5 ml dm<sup>-3</sup> of buffer for most of the cations. When lower volume of en was used, electromigration was not found to be favourable for effective separation because the distance travelled by the cations became less. Therefore, the optimum volume of en selected for neat experiments was 4.8 ml dm<sup>-3</sup> (0.07 M) of buffer system. The zones for this electrolyte system at field strength 10 V cm<sup>-1</sup> when applied for 1 h, were found to be less than 10 mm in diameter. The results (Table 1) indicate comparatively better electromigration of divalent cations as compared with the monovalent or trivalent one. This may be due to their complex forming tendency with en. The non-mobility of trivalent cation like Fe<sup>III</sup> probably is due to the adsorption of complex (cation-en) which is expected because of the strong interaction between the complex and paper. Based on these conditions and observations, the following separations were feasible :  $Fe^{III}$ ,  $Pb^{II}$ ,  $Sb^{III}$ ,  $Co^{II}$ ,  $Ni^{II}$ ,  $Mg^{II}$ ,  $Cu^{II}$ ;  $Bi^{III}$ ,  $Ag^{I}$ ,  $Cd^{II}$ ,  $Zn^{II}$ ,  $Ca^{II}$ ,  $Hg^{II}$ ;  $Sn^{II}$ ,  $Sb^{III}$ ,  $Cr^{II}$ ,  $Pb^{II}$ ,  $Co^{II}$ ,  $Ni^{II}$ ,  $Mg^{II}$ ,  $Cu^{II}$ ;  $Al^{III}$ ,  $Ag^{I}$ ,  $Cd^{II}$ ,  $Zn^{II}$ ,  $Ca^{II}$ ,  $Hg^{II}$ ;  $Fe^{III}$ ,  $Pb^{II}$ ,  $Co^{II}$ ,  $Ni^{II}$ ,  $Cu^{II}$ ;  $Fe^{III}$ ,  $Al^{III}$ ,  $Sb^{III}$ ,  $Ni^{II}$ ,  $Cr^{II}$ ;  $Fe^{III}$ ,  $Al^{III}$ ,  $Ni^{II}$ ,  $Ca^{II}$ ;  $Fe^{III}$ ,  $Ri^{II}$ ,  $Sr^{II}$ ;  $Ni^{II}$ ,  $Cu^{II}$ ,  $Co^{II}$ ,  $Zn^{II}$ ,  $Fe^{III}$ ;  $Ni^{II}$ ,  $Cd^{II}$ ,  $Cr^{II}$ ,  $Hg^{II}$ .

Paper electrophoresis has also been carried out in den-buffer (0.048 M) and ten-buffer (0.036 M)under similar conditions. However, the separations were not successful. Various reagents such as 8-hydroxyquinol, rubeanic acid, pyrogallol, alizarin red, dithiazone, bromophenol blue and sodium rhodozinate were used for locating the spots which gave coloured complexes after electrophoresis. However, it was found that the use of 8-hydroxyquinol followed by the vision under uv light/amine atmosphere remained the most common and effective way for locating most of the cations.

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