ISOTACHOPHORESIS OF METAL–NEUTRAL LIGAND COMPLEXES: 18-CROWN-6 ETHER COMPLEXES WITH ALKALI METALS

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SUMMARY

Results of isotachophoretic separations of alkali metals using aqueous electrolytes with added 18-crown-6 ether are presented. Complexation of these cations by crown ether leads to an improved ability to vary effective mobilities. A simplified theory for 1:1 complexes is presented and good agreement is seen between calculated and observed step heights. The possibility of determining complex formation constants by isotachophoresis is demonstrated.

INTRODUCTION

Metal–ligand complex formation is important in isotachophoresis as a method of varying the effective mobilities of sample ions. Kaniansky and Everaerts[1] showed the utility of adding Ca^{2+} to the leading electrolyte for improving carboxylic acid separations through complexation. Complex formation also is useful for improving metal separations, where effective mobilities are less sensitive to pH. An example is the separation of lanthanoids using 2-hydroxyisobutyric acid as a complexing counter-ion[2].

One class of ions whose mobilities are difficult to modify is alkali metals. Beckers and Everaerts successfully separated NH_4^+ and K^+ using methanol as a solvent[3]. However, the ability to vary mobilities of alkali metals in aqueous electrolytes is desirable. While these metals form weak complexes with acidic ligands, they form moderate complexes with neutral ionophores such as crown ethers. Use of crown ethers offers the potential for improved control over alkali metal separations. In this paper we present the results of isotachophoresis of mixtures of alkali metals using 18-crown-6 ether in the leading electrolyte.

The theory of complexation equilibria in the isotachophoretic steady state has been treated extensively by Hirokawa and Kiso[4,5]. Their computer simulations allow prediction of steady state concentrations, effective mobilities and conductances in complexed zones. Also, complex stability constants can be determined from isotachophoresis data. However, the equations presented by Hirokawa and Kiso are not applicable directly to neutral ionophore equilibria. We present here a simplified theory for alkali metal–crown ether complexes in isotachophoresis using 1:1 leading electrolytes.

THEORETICAL

The leading electrolyte is a non-complexed, 1:1 salt, L^+Y^-, at a concentration c_L with neutral ligand C added at a concentration $\bar{c}_C$. The primary assumption of the model is that there is no mass transport of ligand; *i.e.* the total ligand concentration in zone of sample M^+ also is $\bar{c}_C$. Formation of the complex in the metal zone, (M^+ + C ↔ MC^+) is governed by

$$K = c_{MC} / c_M c_C \tag{1}$$

where c_{MC}, c_M and c_C are the concentrations of complex, free metal and free ligand, respectively. This formation constant is equivalent to the thermodynamic constant since activity coefficients for metal and complex cancel. By substituting expressions for total metal concentration, $\bar{c}_M = c_M + c_{MC}$, and total ligand concentration, $\bar{c}_C = c_C + c_{MC}$, an equation for free metal concentration can be obtained.

$$Kc_M^2 + c_M (1 + K\bar{c}_C - K\bar{c}_M) - \bar{c}_M = 0 \tag{2}$$

The total metal concentration in the steady state is determined by the Kohlrausch regulating function[6] and is dependent on leading ion mobility and leading zone specific conductance

$$\bar{c}_M = \bar{\lambda}_M c_L (\lambda_L + \lambda_y^L)/\lambda_L(\bar{\lambda}_M + \lambda_y^M) \tag{3}$$

where c_L and λ_L are the concentration and single ion conductance of the leading ion, λ_y^L and λ_y^M are the single ion conductances of the counter-ion in the leading and metal zone, and $\bar{\lambda}_M$ is the effective single ion conductance of the metal

$$\bar{\lambda}_M = (\lambda_M c_M + \lambda_{MC} c_{MC})/\bar{c}_M \tag{4}$$

Substitution of eqn. 4 into eqn. 3 yields an expression for the total metal concentration in the steady state

$$a_1 \bar{c}_M^2 + (a_2 c_M - a_3 c_L) \bar{c}_M + a_4 c_M c_L = 0 \tag{5}$$

where $a_1 = \lambda_L (\lambda_y^M + \lambda_{MC})$, $a_2 = \lambda_L (\lambda_M - \lambda_{MC})$, $a_3 = \lambda_{MC} (\lambda_L + \lambda_y^L)$ and $a_4 = (\lambda_{MC} - \lambda_M) (\lambda_L + \lambda_y^L)$. The single ion conductances at finite ionic strength can be obtained from the Onsager equation

$$\lambda = \lambda° - (A \lambda° + B) c^{\frac{1}{2}} \tag{6}$$

where $c = \bar{c}_M$ for λ_M, λ_{MC} and λ_y^M, $c = c_L$ for λ_L and λ_y^L, and $A = 0.23$ and $B = 60.65$ for water at 25°C[7].

Eqns. 2, 5 and 6 can be solved iteratively for given values of c_L, K, $\bar{c}_C$, and $\lambda°$. Initially $\bar{c}_M$ and c_M are set equal to c_L. Single ion conductances then are corrected to finite ionic strength using eqn. 6. $\bar{c}_M$ is found from eqn. 5 and c_M from eqn. 2. The procedure is repeated until successive values of $\bar{c}_M$ agree within 0.1%. Specific con-

ductivities, k, are calculated for the leading L and metal M zones. A step height ratio is calculated as $R = k_L/k_M = c_L (\lambda_L + \lambda_y^L)/(\lambda_M c_M + \lambda_{MC} c_{MC} + \lambda_y^M \bar{c}_M)$.

The effect of metal–neutral ionophore complexation is shown in Fig. 1. Numerical solutions of eqn. 2–6 are plotted for the specific case $\lambda_M^o = \lambda_L^o = \lambda_y^o = 5 \lambda_{MC}^o$ and $c_L = 10$ mM. For formation constants of 10 or greater, addition of complexing agent to the leading electrolyte at concentrations above 1 mM causes significant changes in the relative step height of the metal zone. In the limit that $K\bar{c}_C \gg 1$, the relative step height is constant at a value dependent on λ_{MC}. The greatest effect on step height will come from large ionophores which have the smallest values of λ_{MC}^o. Since 18-crown-6 ether has stability constants between 5 and 100 and $\lambda_M^o/\lambda_{MC}^o$ ratios between 2 and 3 with many alkali metals, addition of this ligand to leading electrolytes should provide a convenient method of varying effective mobilities of these metals in cationic isotachophoresis.
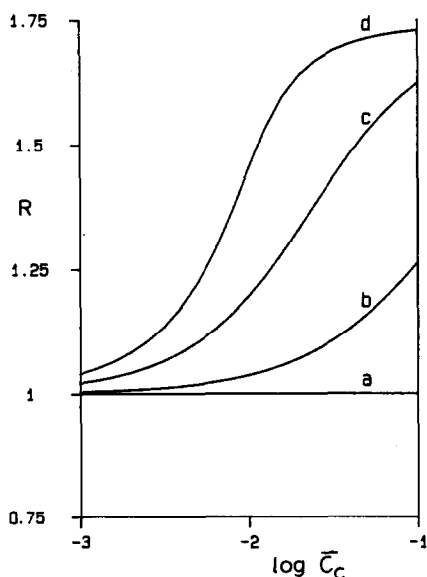

Fig. 1. Dependence of relative step height, R, on crown ether concentration $\bar{c}_C$ (molar). $\lambda_M^o = \lambda_L^o = \lambda_y^o = 5 \lambda_{MC}^o$; $c_L = 0.01$ M; (a) $K = 0$, (b) $K = 10$, (c) $K = 100$, and (d) $K = 1000$ l/mole.

EXPERIMENTAL

Isotachophoresis was performed on an LKB (Bromma, Sweden) 2127 Tachophor equipped with a 2127-140 conductivity detector and a 20-cm long capillary with an I.D. of 0.8 mm. Leading electrolytes were 10 mM hydrochloric acid with varying amounts of 18-crown-6 ether added. The terminating electrolytes were 10 mM tris(hydroxymethyl)aminomethane (THAM) adjusted to pH 8.5 with 0.1 M hydrochloric acid, or 10 mM tetrabutylammonium chloride. Initial currents of 300 μA were reduced to 50 μA prior to detection.

Hydrochloric acid and chloride salts of potassium, sodium, ammonium, lithium and rubidium were obtained from Fisher (Pittsburgh, PA, U.S.A.). Cesium chlo-

ride was obtained from J. T. Baker (Phillipsburg, NJ, U.S.A.). THAM and 18-crown-6 were obtained from Sigma (St. Louis, MO, U.S.A.) and tetramethylammonium (TMA) and tetrabutylammonium (TBA) chlorides were obtained from Eastman (Rochester, NY, U.S.A.). All chemicals were reagent grade and were used without further purification.

RESULTS AND DISCUSSION

Prediction of effective mobilities

To test the validity of the model derived above, measurements of the relative step height of K^+ were performed using leading hydrochloric acid electrolytes containing various amounts of 18-crown-6. In order to obtain precise relative step height measurements, TMA was used as an internal standard. Measured step heights of the leader, h_L, metal, h_M and standard, h_s can be converted into a relative step height by

$$R_M = R_L + [(h_M - h_L)/(h_s - h_L)] (R_s - R_L) \qquad (7)$$

The R_s value for TMA was calculated from eqns. 5 and 6 and was found to be 8.23 using $\lambda_L^\circ = 349.8$, $\lambda_Y^\circ = 76.35$, $\lambda_M^\circ = 46.6$ S cm²/equiv. and $c_L = 10$ mM.

Table I gives the results of experimentally observed step heights and those calculated from eqns. 2, 5 and 6 along with calculated total metal concentration, single ion conductance and metal zone conductance for K^+ at various added 18-crown-6 concentrations. Good agreement is seen between predicted and observed step heights for this simple model. Using 100 mM 18-crown-6, the mobility of the potassium zone is to low for it to move in the isotachophoretic steady state and its step height is reported as greater than that of the terminator, THAM. For crown ether concentrations between 1 and 30 mM, the average relative difference between observed and calculated step heights is 1.0%. This accuracy is sufficient for predicting the effect of neutral ligand addition on isotachophoresis separations when stability constants and complex mobilities are known.

TABLE I

CALCULATED AND OBSERVED RELATIVE STEP HEIGHTS AND CALCULATED CONCENTRATIONS AND CONDUCTANCES FOR K^+/18-CROWN-6 COMPLEX

Leading electrolyte = HCl; $c_L = 10$ mM; $K = 107$ l/mole, $\lambda_L^\circ = 349.8$, $\lambda_Y^\circ = 76.35$, $\lambda_M^\circ = 73.5$, $\lambda_{MC}^\circ = 25.3$ S cm²/equiv.[8]. $\lambda_L = 335.7$; $\lambda_Y^L = 68.5$; $k_L = 4042$ μS/cm.

$\bar{c}_C$ (mM)	$\bar{c}_M$ (mM)	λ_M (S cm²/equiv.)	k_M (μS/cm)	R	
				Calc.	Obs.
0	5.90	67.5	813	4.97	4.77
1	5.76	64.6	778	5.20	5.14
3	5.50	59.3	714	5.66	5.59
10	4.77	46.5	560	7.22	7.27
30	3.83	33.3	401	10.1	10.0
100	3.17	25.7	310	13.1	>12.6

Separation of alkali metals

The effect of 18-crown-6 on the separation of mixtures of alkali metals and TMA is shown in Fig. 2. With no added crown ether (Fig. 2a), K^+, NH_4^+, Cs^+ and Rb^+ have similar mobilities and are poorly separated. Only Na^+ and Li^+ are well separated from the other alkali metals. Addition of 3 mM 18-crown-6 to the leading electrolyte (Fig. 2b) results in decreased K^+ mobility and a well separated K^+ zone. NH_4^+, Cs^+, and Rb^+ remain in a mixed zone. Increasing the crown ether concentration to 30 mM causes dramatic changes in the separation pattern (Fig. 2c). Complexation of NH_4^+ and Rb^+ gives separation of these ions from Cs^+. However, K^+ mobility has decreased to where it forms a mixed zone with Li^+.

Fig. 3 plots experimental relative step heights calculated from eqn. 7 *vs.* crown ether concentration in the leading electrolyte. Inspection of the plot indicates that a
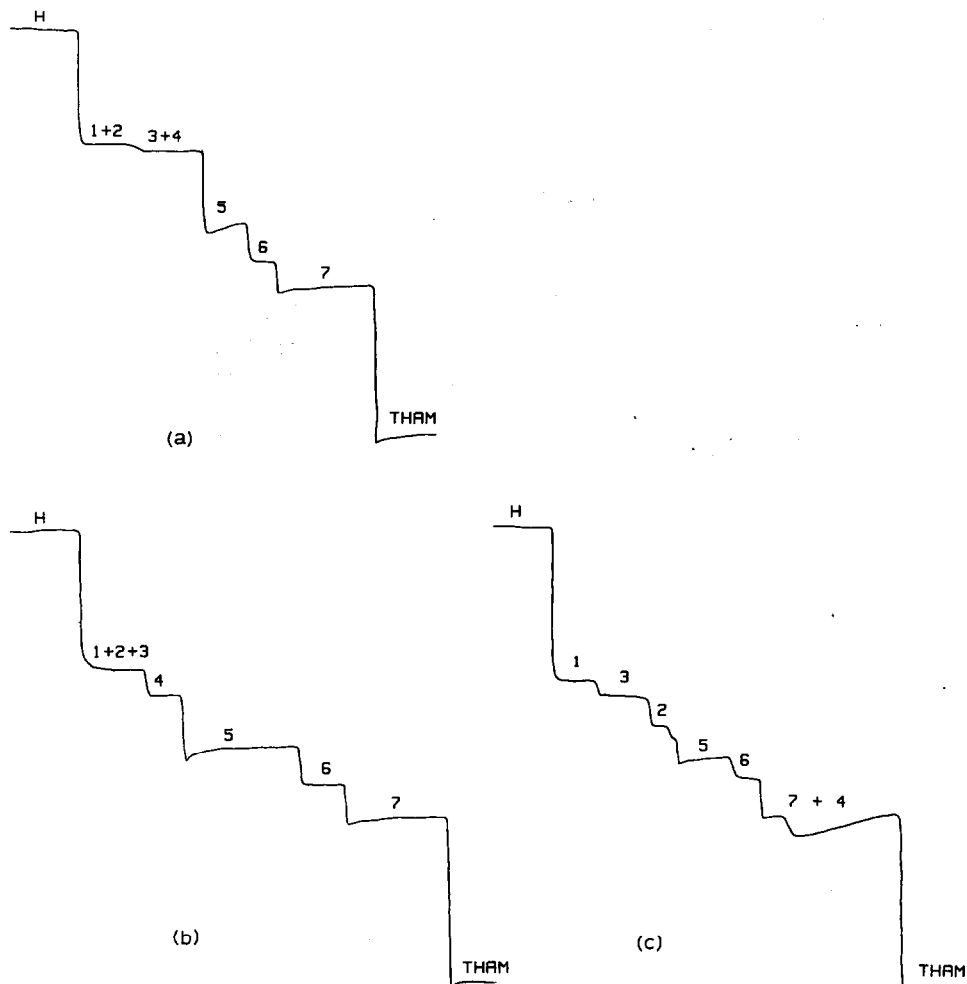

Fig. 2. Isotachopherograms of (1) Cs^+, (2) Rb^+, (3) NH_4^+, (4) K^+, (5) Na^+, (6) TMA and (7) Li^+. Leading electrolyte: 10 mM HCl plus (a) 0 mM, (b) 3 mM and (c) 30 mM 18-crown-6. Terminating electrolyte: 10 mM THAM, pH 8.5. Detection current, 50 μA.
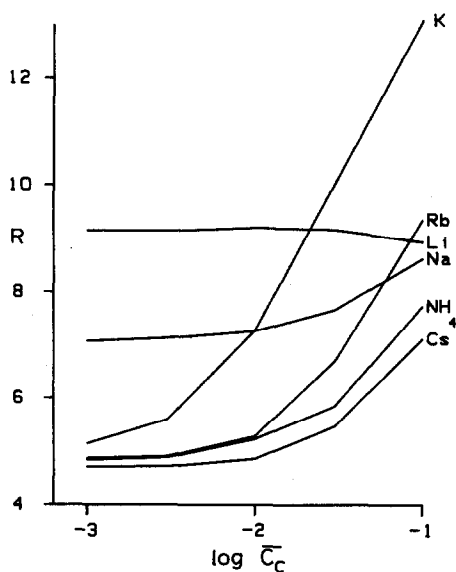
Fig. 3. Dependence of experimental relative step height, R, on 18-crown-6 concentration, $\bar{c}_C$, for alkali metals. Conditions are the same as Fig. 2. Curves are not smooth since step heights were measured at discrete crown concentrations.

crown ether concentration of 50 mM should give a reasonable separation of the alkali metals. This separation is shown in Fig. 4. All five alkali metals and NH$_4^+$ are well separated. Using this electrolyte, TMA will form a mixed zone with Na$^+$.

In our laboratories, we routinely use 10 mM hydrochloric acid plus 3 mM 18-crown-6 as a leading electrolyte for K$^+$ determinations. An example is monitoring
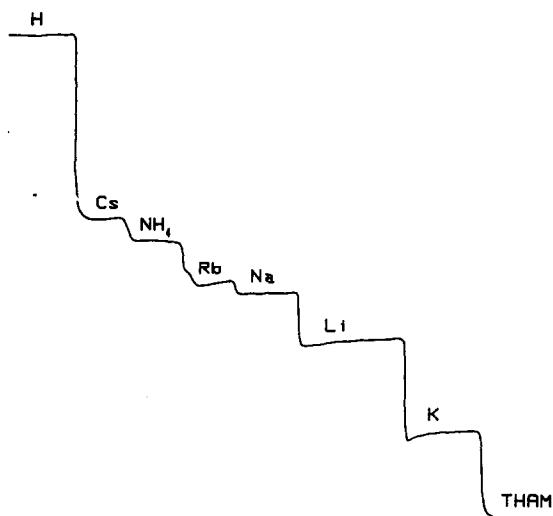

Fig. 4. Separation of alkali metals using 10 mM HCl plus 50 mM 18-crown-6 leading electrolyte. Other conditions are the same as Fig. 2.
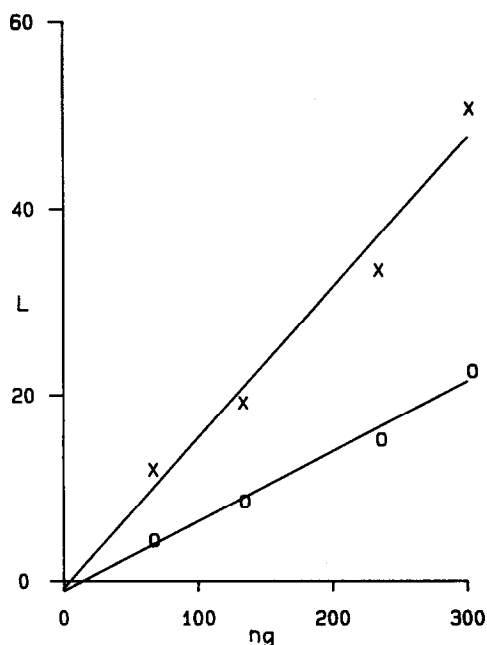
Fig. 5. Zone length, L, in seconds $vs.$ nanograms injected for ($\times$) K^+ and (0) lysine. Leading electrolyte: 10 mM HCl plus 3 mM 18-crown-6. Terminating electrolyte: 10 mM TBA chloride. Detection current, 50 μA.
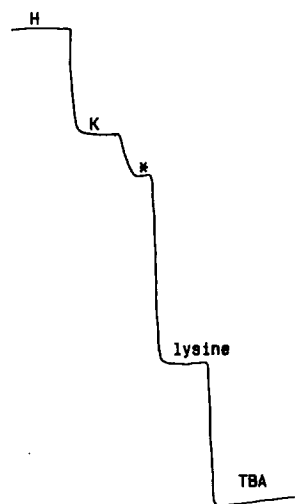
Fig. 6. Separation of K^+ and lysine using electrolytes described in Fig. 5. * zone is electrolyte impurity.

the neutralization of lysine with potassium hydroxide to form potassium lysinate. For this analysis, TBA must be used as the terminating ion in order for lysine to migrate isotachophoretically. K^+ / lysine mole ratios and any NH_4^+ impurity can be determined simultaneously. Calibration curves of zone length $vs.$ nanograms injected for K^+ and lysine are shown in Fig. 5. Zone lengths were measured using an automated data acquisition system reported previously[9]. A typical sample injection is shown in Fig. 6. For this particular sample, lysine was found to be 89% neutralized and no NH_4^+ was detected.

TABLE II

FORMATION CONSTANTS OF 18-CROWN-6 COMPLEXES DETERMINED BY ISOTACHOPHORESIS

		K^+	NH_4^+	Na^+
K (ITP)	(n = 4)	107 ± 7	11.0 ± 0.1	3.3 ± 0.4
K (Lit.)	Ref. 11*	107 ± 25	17.0 ± 2.3	6.3 ± 1.5
	Ref. 12**	115 ± 10	12.6 ± 2.9	<2
	Ref. 13**	111.6 ± 0.4	–	6.6 ± 0.3

* Calorimetry.
** Potentiometry.

Determination of complex formation constants

Variation of K when calculating R from eqns. 2, 5 and 6 to obtain a least squares fit with experimental step heights allows extraction of formation constants from isotachophoretic data. A "brute-force" fit was obtained by testing log K values of 0 and 3. Goodness of fit, F, of the experimental and calculated R values was found using

$$F = \Sigma \ W_i \ (R_i^{\text{calc}} - R_i^{\text{obs}})^2 \tag{8}$$

where weighting factors W were proportional to experimental step heights. After the initial K values were tested, further K values were selected by the method of bisection.

Table II presents the results of fitting data for K^+, NH_4^+, and Na^+ to best K values. Average K values and standard deviations are from four sets of R *vs.* $\bar{c}_C$ data at five crown concentrations. Parameters were the same as in Table I except $\lambda_M^0 = 50.1$ S cm^2/equiv. for Na^+. Equal values for complex mobilities could be used since the three cation complexes have nearly identical Stokes radii[10]. Agreement between formation constants determined by isotachophoresis and previously reported values is seen. Relative standard deviations of 5–10% compare favorably with the precision of the literature methods. The advantage of the ITP method is that the formation constants for several metals can be determined simultaneously.

Constants obtained isotachophoretically are slightly lower than the literature values. The model does not account for Joule heating in the capillary which will alter the values of λ^0 and K governing the steady state.

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