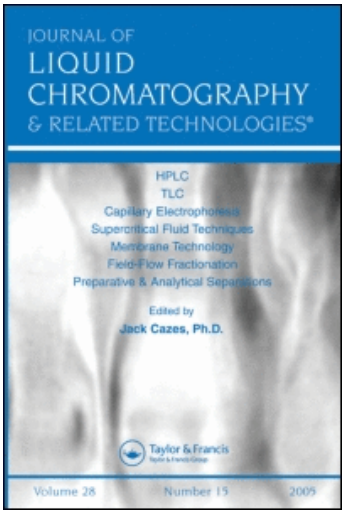

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CAPILLARY ELECTROPHORESIS OF ALKALI AND ALKALINE-EARTH CATIONS WITH IMIDAZOLE OR BENZYLAMINE BUFFERS

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Abstract

The separation of alkali, alkaline earth, and ammonium cations in several samples of water was achieved by capillary electrophoresis with indirect UV detection . A solution of imidazole (10^{-2} M, pH 4.5) was used as buffer to resolve a mixture of six cations (K^+, Na^+, Ca^{2+}, Ba^{2+}, Li^+ and Mg^{2+}) by capillary electrophoresis at 214 nm in less than 10 min. The addition of potassium cation to the running buffer has an influence on the resolution of Ca^{2+}/Na^+ and Na^+/Mg^{2+} peaks. A linear relationship between the corrected peak area and concentration was obtained in the 1-10 ppm range for these cations using a hydrodynamic injector. This electrophoretic system permitted the separation of these inorganic cations at a 50 ppb-level concentration with an hydrodynamic injection, thus making it possible to quantitatively determine their presence in mineral waters by capillary electrophoresis.

At pH 4.5, potassium and ammonium unfortunately have identical ionic mobilities causing them to comigrate in an imidazole buffer. Using an alkaline solution of benzylamine (10^{-2} M, pH 9) as carrier electrolyte, their separation can be successfully achieved with excellent resolution at 204 nm.

The analyses of tap water and several mineral waters have been achieved by capillary electrophoresis.

INTRODUCTION

Capillary electrophoresis, a recent analytical technique, is currently undergoing rapid development owing to its efficiency, high resolution, relative simplicity, UV detection with low wavelengths (190 nm), speed and automatization of separations, and low buffer consumption. Compared to liquid phase chromatography, one disadvantage, seems to be not only a rather weak reproducibility of the electroosmotic flow assuring the migration times, but also a lower concentration sensitivity. However, this technique is available not only for the separation of inorganic and organic anions and cations but also for the separation of ionizable and neutral organic molecules [1].

Until today, capillary electrophoresis had been studied more for the analysis of anions than of cations. Nevertheless, the analytical approach is similar, in that indirect detection is required because of the transparency of inorganic ions in the UV region (except for a few inorganic anions such as nitrate and bromide), and the operating conditions such as the electroosmotic flow must be in the same direction as the electrophoretic mobility of the ions analyzed in order to minimize their migration time. Table I shows some of the articles relative to the separation of cations by capillary electrophoresis [2-13].

Several types of detectors have been utilized for the separation of cations with the two major ones being the indirect UV detector and indirect fluorimetric detector. Initially, Aguilar et al. [2] separated the cations Fe^{2+}, Cu^{2+} and Zn^{2+} in the form of their cyano complexes in capillary electrophoresis; a phosphate buffer (20 mM) was used at pH 7 and cation detection was carried out at 214 nm. Then Foret et al. [5] separated fourteen lanthanides by capillary electrophoresis using an electrophoretic buffer consisting of an α-hydroxylisobutyric acid as complexing agent and creatinin as the indirect UV detector marker; in fact, the similarity between the electrophoretic mobilities of alkali and alkaline-earth, rare-earth and metal cations requires the addition of a water-soluble complexing reagent to the electrolyte. The electrophoretic mobility of each cation thus decreases following their complexation in-situ by α-hydroxylisobutyric acid and selectivity is largely improved. The factors affecting the separation of such cations were studied by Weston et al. [6,7]; the complexing agent was also α-hydroxylisobutyric acid and the compound allowing indirect UV detection was called UV Cat 1, a proprietary reagent developed at Waters. Very recently, Beck et al. [9] firstly proposed a nice alternative way for the separation of several alkali and alkaline earth cations by using an imidazole-based electrophoretic buffer and indirect UV detection at 214 nm. Finally, Swaile and Sepaniak [12] used a laser-based fluorimetric detector to detect cations Ca^{2+}, Mg^{2+} and Zn^{2+}, complexed by 8-hydroxyquinoline-5-sulfonic acid; this technique was applied to the detection of Ca(II) and Mg(II) in blood serum.

In order to use indirect photometric detection and to obtain symmetrical peaks, a UV absorbing cationic compound having similar electrophoretic mobility to that of the cations analyzed was chosen as main constituent of the buffer; several organic bases were considered among which was an aromatic amino (benzylamine) or a heterocyclic azote (imidazole).

Table I. A few examples of some inorganic cation separations by capillary electrophoresis.

Cations	Detection	Electrolyte	pH	Time	Sensitivity	Ref.
Zn^{2+}, Cu^{2+} Fe^{2+}	Direct UV 214 nm	phosphate 20 mM	7,0	5 min	-	2
$Au(CN)_2^-$ $Ag(CN)_2^-$	Direct UV 214 nm	$NaHCO_3$ 10 mM	9,6	-	2,82 µg/mL Au 3,48 µg/mL Ag	3
Al^{3+}, AlF^{2+}, AlF_2^+	Indirect UV 214 nm	imidazole 5 mM	3,5	5 min	10^{-5} M	4
lanthanides	Indirect UV 220 nm	acetate 30 mM - creatinine- HIBA 4 mM	4,8	5 min	10^{-7} M	5
alkali alkaline-earth lanthanides	Indirect UV 214 nm	UV Cat-1 5 mM HIBA 6,5 mM	4,4	10 min	0,05-0,1µg/mL 0,1-0,2 µg/mL	6,7
lanthanides	Indirect UV 214 nm	benzylamine 9 mM HIBA 4 mM CH_3COOH 20 mM	4,6	8 min	-	8
K^+, Na^+, Ba^{2+} Ca^{2+}, Mg^{2+}, Li^+	Indirect UV 214 nm	imidazole 5 mM	4,4	10 min	0,1 µg/mL	9
K^+, Ca^{2+}, Na^+, Mg^{2+}	Indirect UV 214 nm	UV Cat-1 5 mM- HIBA 6,5 -4,0 mM	4,0	10 min	5 µg/mL	10
alkali alkaline-earth	Indirect Fluorimetry	quinine sulfate H_2SO_4 0,58 mN	3,7	5 min	0,1-0,5fmol	11
Mg^{2+}, Ca^{2+} Zn^{2+}	Fluorimetry	HQS^- 2,5 mM λ_{exc} = 325 nm λ_{emis}= 425 nm	8,0	10 min	0,05-0,6 µg/mL	12
alkali alkaline-earth	Indirect Fluorimetry λ_{exc} = 251 nm λ_{emis}= 345 nm	Ce(III) 0,5 mM- 18-crown-6 2.5 mM	-	7 min	$1\text{-}3.10^{-6}$ M	13
K^+, Na^+, Rb^+, Li^+	conductimetry	MES 20 mM - histidine	6,0	8 min	2.10^{-6} M	14

HIBA: α-hydroxyisobutyric acid
HQS^-: 8-hydroxyquinoline-5-sulfonic acid
UVCat 1: UV background-providing component of the electrolyte (Waters product)

Several examples of cation separations are presented (K^+, Na^+, Ca^{2+}, Ba^{2+}, Li^+, Mg^{2+}, and NH_4^+) as well as their quantitative analyses.

EXPERIMENTAL

Capillary Electrophoresis Materiels

Different capillary electrophoresis systems were employed for this study; one was used in manual mode (Europhor), and the others in automatic mode with a thermostated capillary column (Spectra, Beckman).

Separations carried out on a Spectraphoresis 1000 apparatus (Spectra-Physics, San Jose, CA, USA) used a capillary column whose dimensions were 70 cm (or 44) total length, 63 cm (or 37) from the point of injection to the detector cell, and 50 μm I.D. Separations were achieved at an adjustable temperature (25-40°C) under positive power supply (15-30 kV) with a few tenths of microampers. Detector time constant was set at 0.1 s. The hydrodynamic injection was achieved in a short time (0.5 or 1 s). Data was kept in an IBM PS/2 computer model 70 386 using data acquisition software and electrophoretic data manipulation. This apparatus also contained a variable wavelength detector which allowed the recording of the UV-visible spectrum of the migrating compound.

Separations carried out on a P/ACE 2100 apparatus (Beckman Instruments Inc., Fullerton, CA, U.S.A.) equipped with a UV detector with wavelength filters (190, 200, 214, 254, 260, 280 nm). Fused silica capillaries (Beckman Intruments Inc.) of 75 μm I.D. x 375 μm O.D. and 57 cm long (50 cm to the detector) were used. The part of the capillary where separation takes place was kept at a constant temperature by immersion in a cooling liquid circulating in the cartridge with a detection aperture of 100 μm x 800 μm. The solutes were injected at the anode end of the capillary in the hydrodynamic mode by azote superpressure (0.5 psi). Data were collected using a IWT data acquisition system. Detector time constant was 0.1 s and data acquisition rate was 20 Hz.

Electrophoretic separations carried out on a Prime Vision apparatus (Europhor, Toulouse, France) utilized a fused-silica capillary tube with the following dimensions: total length, 96 cm; injector-detector length, 65 cm; I.D., 75 μm. Positive power supply (15 to 25 kV) delivered by a high-voltage generator (Prime Vision V) was applied between the two electrodes. The hydrodynamic injection times ranged between 1 and 10 s. The UV-visible detector (Prime Vision II) has variable wavelengths. A small section of polyimide was removed in order to carry out on-column UV detection with an optical length close to 75 μm; the detection wavelength was set at 214 and 204 nm for the imidazole and benzylamine buffers, respectively. The electropherograms were recorded on a Shimadzu C-R 5A integrator (Kyoto, Japan).

The pH of each solution was verified on a Beckman pH meter (Model ϕ10, Fullerton, CA, USA). The capillary tubing of fused silica was conditioned daily by rinsing with a solution of sodium hydroxide 1 N (10 min), then water (10 min), and finally with the

electrophoretic buffer (15 min). Between two consecutive analyses, the capillary was rinsed with water (3 min) then with the electrophoretic buffer (5 min) to improve reproducibility of the electroosmotic flow and migration times of the solutes.

Reagents and Products

All chemical products used were of analytical quality. Imidazole (99% purity) was obtained from Sigma (St Louis, MO, USA) whereas the tetrabutylammonium bromide (TBABr) (99% purity) and benzylamine (99% purity) were purchased from Aldrich (Milwaukee, WI, USA). The water used in the preparation of buffers as well as that necessary for dilutions was of HPLC quality (Fisons, Farmitalia, Milan, Italy). The electrophoretic buffer pH was adjusted with acetic acid 1M (Carlo Erba, Milan, Italy). Finally, each buffer or rinsing solution was filtered before use through a membrane filter having a diameter of 25 mm and porosity of 0.2 μm (Whatman, Maidstone, Great Britain).

RESULTS AND DISCUSSION

The electrophoretic buffer used for the separation of inorganic cations must contain an organic cation (stable within the 4-9 pH range to resolve cations K^+ and NH_4^+, particularly), which has an electrophoretic mobility close to that of the inorganic cations to be separated in order to obtain symmetrical and efficient electrophoretic peaks, and also an intense chromophore group in the UV region.

The imidazole (or 1,3-diaza-2,4-cyclopentadienyl ring) is a heterocyclic azote (pKa_1 = 6.9 and pKa_2 = 14.5) whose UV absorption spectrum maximum at pH 4.5 was set at 211 nm (Fig. 1). Moreover, the imidazole cation appears to have an electrophoretic mobility close to that of the calcium and sodium cations [9]. By using an imidazole buffer concentration of 10^{-2} M and a pH adjusted to 4.5, a mixture of five inorganic cations (K^+, Ba^{2+}, Ca^{2+}, Na^+ and Li^+) was nicely separated in 5 min (Fig. 2); separation voltage was equal to +20 kV (current 12 μA). The sample injection was carried out at the anodic end of the capillary and detection at the cathodic end; the electroosmotic flow (at 10.3 min) was in the same direction as the electrophoretic mobility of the cation. The number of theoretical plates was fairly high (from 377 000 for Ba^{2+}, 231 000 for Ca^{2+}, 257 000 for Na^+ to 102 000 for K^+) as mentionned in Table II. The migration order of Na^+ relative to Ca^{2+} and Mg^{2+} differs from that reported previously [9] but the buffer has not exactly the same composition. Besides, as already been reported, the co-ion buffer concentration may influence the migration of the analytes.

Addition of a Cation to the Electrolyte

A clear separation improvement of the sodium-magnesium cations was observed when the K^+ cation was added to the electrophoretic buffer (Fig. 3). Fig. 4 shows the

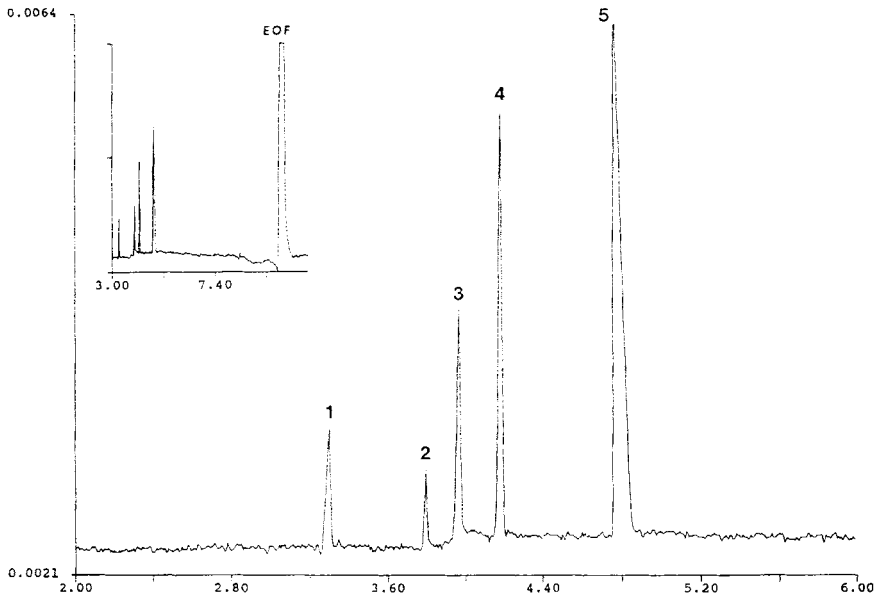


Figure 1. On-line UV spectra of selected buffers in the separation of cations by capillary electrophoresis.
1) imidazole (at pH 4.5); 2) benzylamine (at pH 9).

influence of the K^+ ions added to the buffer on the resolution of the Ca^{2+}/Na^+ and Na^+/Mg^{2+} peaks. The resolution of the Ca^{2+}/Na^+ peaks decreases for increasing potassium concentration added to the imidazole buffer and becomes smaller than 2.0 at potassium concentration greater than 10^{-3} M. At the opposite, the resolution of the Na^+/Mg^{2+} peaks increases for increasing potassium concentration in the running buffer. For a concentration of K_2SO_4 close to 0.5 mM, the resolution of Na^+-Mg^{2+} was greatly improved, and the separation of six cations was completed in less than 5 min (see Fig. 5a). However, the addition of a potassium cation to the buffer disturb indirect UV detection of the potassium cation, generally present in various samples (mineral water), by causing continually decreasing sensitivity loss for K^+ cation (Figs. 3a-e) and even a negative peak (Figs. 3d-e). An alternate solution would be to add to the electrophoretic buffer a cation not present in the sample to be analyzed; for example, the tetrabutylammonium cation added to the buffer at a concentration of 10^{-3} M avoided this detection drawback by maintaining the same resolution of cations Ca^{2+}, Na^+ and a slightly higher resolution of cations Na^+, Mg^{2+} (see Fig. 5b).

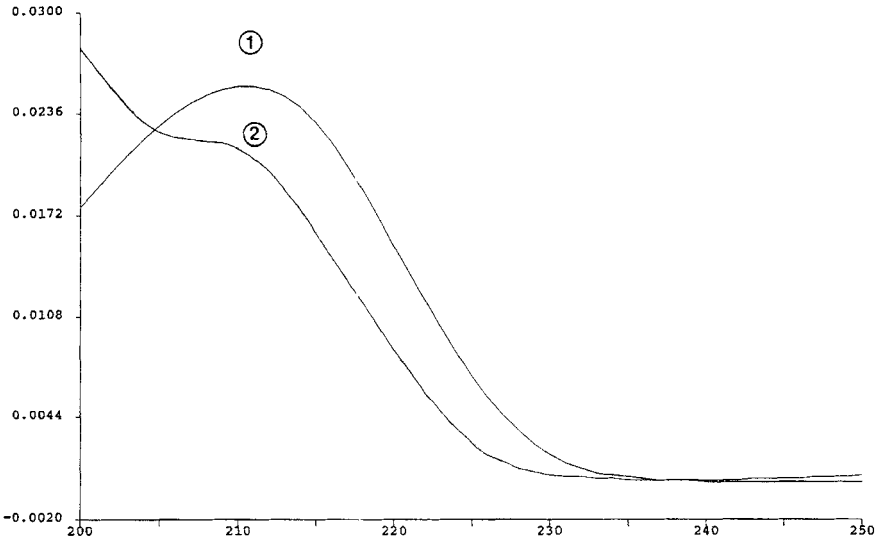


Figure 2. Separation of a standard mixture of five inorganic cations by capillary electrophoresis with imidazole as carrier electrolyte.
Buffer, 10^{-2} M imidazole (pH 4.5); applied voltage, + 19.8 kV; thermostatted capillary dimensions, 70 cm x 50 µm I.D.; wavelength detection, 214 nm; current, 12 µA; hydrodynamic injection, 5 s; temperature, 25°C; cation concentration, 4 ppm. Identification: 1) K^+; 2) Ba^{2+}; 3) Ca^{2+}; 4) Na^+; 5) Li^+.

Table II. Electrophoretic parameters of cations.
Applied voltage, + 19.8 kV; field strength, 283.5 v/cm; electroosmotic flow, $3.59.10^{-4}$ $cm^2/V^{-1}.s^{-1}$; experimental conditions: as the Figure 1.

cation	efficiency	asymetry factor	electrophoretic mobility (x 10^4 $cm^2 \cdot V^{-1} \cdot s^{-1}$)
K^+	102 000	0.29	7.63
Ba^{2+}	377 000	0.79	6.18
Ca^{2+}	231 000	0.46	5.74
Na^+	257 000	0.53	5.25
Li^+	60 000	5.31	4.17

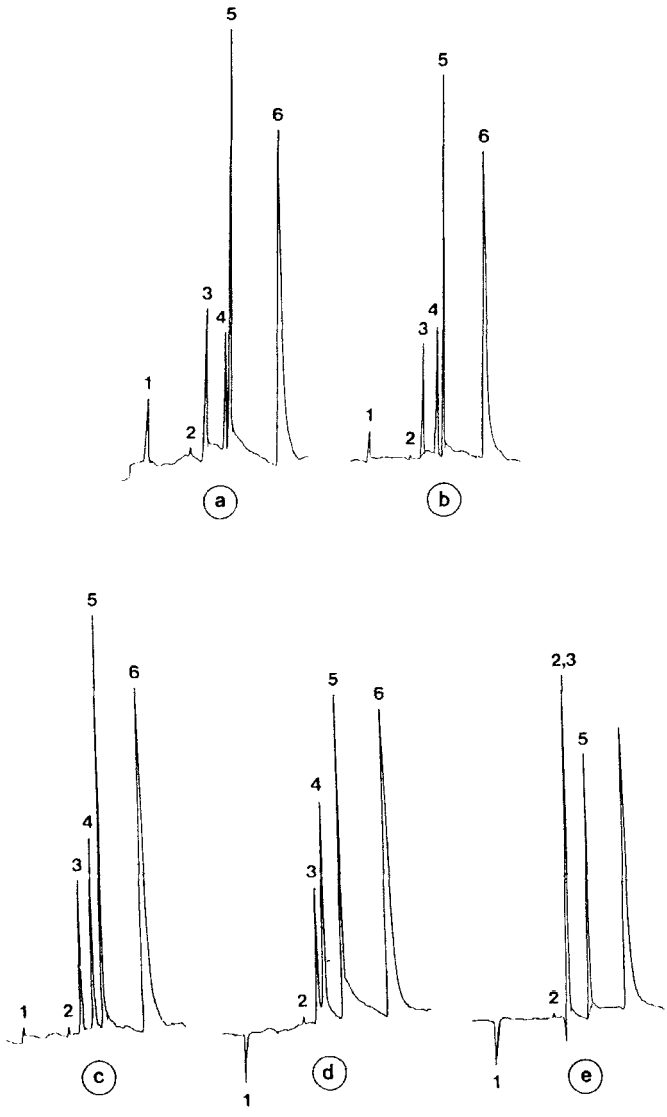


Figure 3. Addition of K^+ ions to the imidazole buffer.

Applied voltage, + 15 kV; capillary dimensions; 65 cm x 75 μm I.D.; wavelength detection, 214 nm; temperature 22 °C. Identification: 1) K^+; 2) Ba^{2+}; 3) Ca^{2+}; 4) Na^+; 5) Mg^{2+}; 6) Li^+.

Concentration of K^+ cation added to the imidazole buffer (10^{-2} M, pH 4.5): a) 0 mM; b) 0.25 mM; c) 0.5 mM; d) 1 mM; e) 1.5 mM

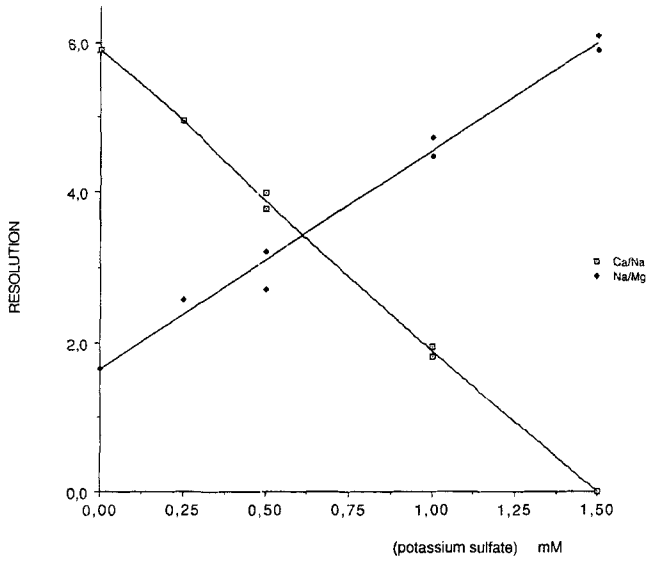
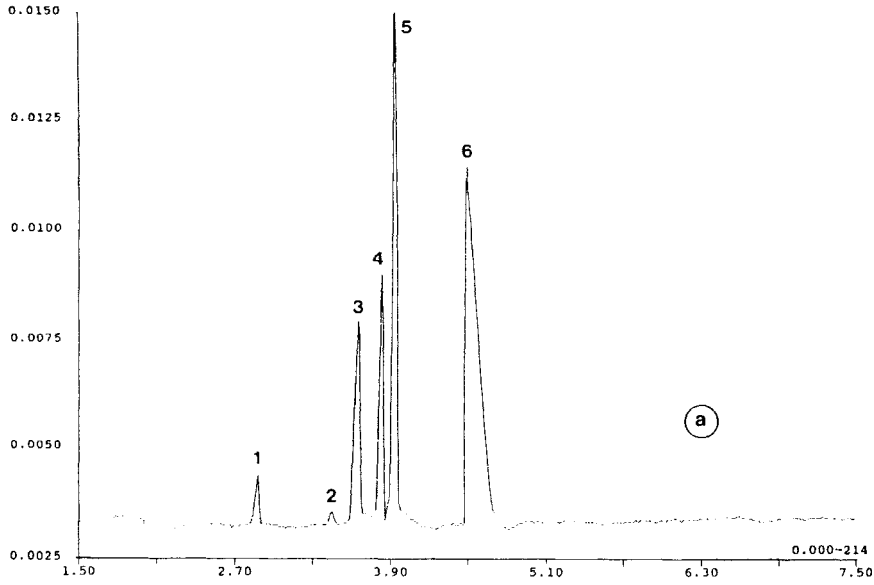


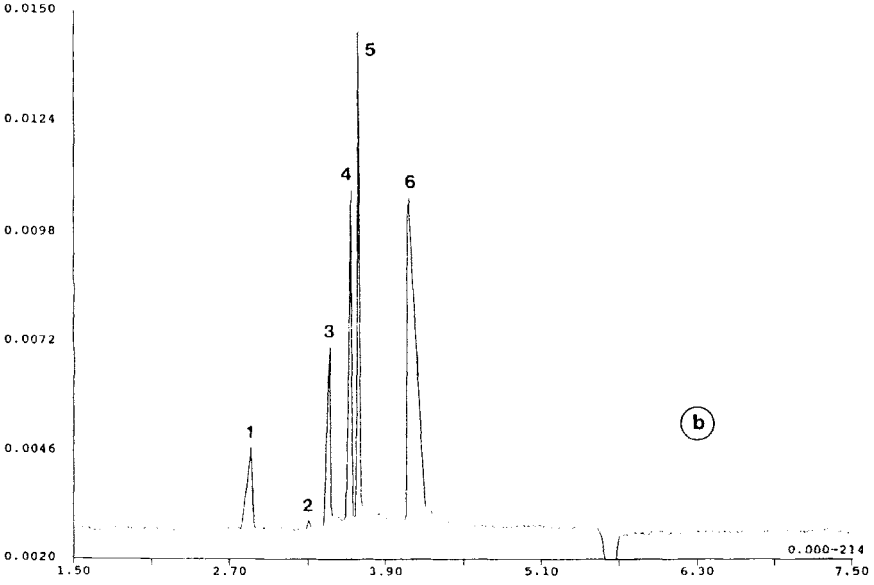
Figure 4. Influence of K_2SO_4 concentration added to the imidazole running buffer on the electrophoretic resolution of the Ca^{2+}/Na^+ and Na^+/Mg^{2+} peaks.
Buffer, imidazole 10^{-2} M + K_2SO_4, pH 4.5. Capillary dimensions: 65 cm x 75 µm I.D. UV detection at 214 nm; hydrodynamic injection time, 3s. a) Ca^{2+}/Na^+; b) Na^+/Mg^{2+}.

Some of the experiments must be repeat with coated capillary with lower pH buffer or different inorganic and organic cations as buffers additives to gain additional insights into the role of cation/silica interactions.

Separation of Potassium and Ammonium Using Benzylamine Carrier

The simultaneous separation of ammonium and potassium cations is very useful in the analysis of food products or water. Unfortunately, these two cations have nearly identical electrophoretic mobilities (at 25°C, 73.5 and 73.4 mho-cm^2/eq for K^+ and NH_4^+, respectively), bringing about their comigration with the imidazole buffer (pH 4.5). In an alkaline medium, their separation is successful by partially transforming the ammonium cation to NH_3 (pKa = 9.25). Under these electrophoretic alkaline conditions, the mobility of the NH_4^+ cation decreases and the potassium cation migrates faster than the ammonium; on the other hand, the electroosmotic flow increases bringing about a shorter analysis time (see Fig. 6). The selected buffer chosen was benzylamine (pKa = 9.3) which has an high absorbance in the 200-204 nm UV range (Fig. 1b); its concentration was 10^{-2} M with the pH adjusted to 9.





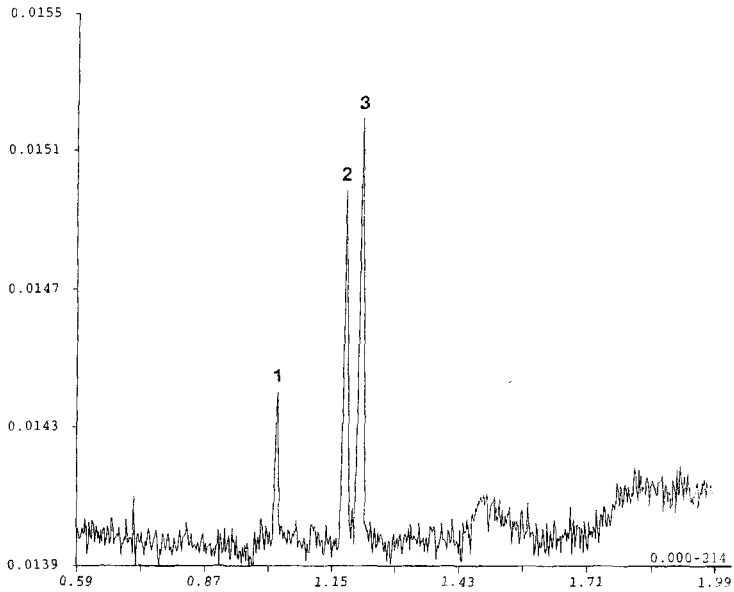


Figure 6. Separation of potassium and ammonium cations by capillary electrophoresis with benzylamine as carrier electrolyte.
Applied voltage, + 20 kV; thermostatted capillary dimensions, 44 cm x 50 μm I.D.; wavelength detection, 204 nm; temperature, 25°C; hydrodynamic injection time, 2 s; buffer; 10^{-2} M benzylamine (pH 9); solute concentration, 5 ppm. Cations: 1) K^+; 2) NH_4^+; 3) Na^+.

Determination of Electroosmotic Flow

The electroosmotic flow may be graphically determined by plotting for each cation the reciprocal migration time $t_m(i)$ versus the equivalent conductance limit λ_i^∞. Indeed, the migration speed of cation i in the capillary zone electrophoresis is expressed by the

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Figure 5. Addition of K_2SO_4 or TBABr to the imidazole buffer
a) Buffer, 10^{-2} M imidazole + K_2SO_4 5 10^{-4} M (pH 4.5); applied voltage, + 25 kV; thermostatted capillary dimensions, 70 cm x 50 μm I.D.; wavelength detection, 214 nm; electromigration injection, 5 s at 10 kV; solute concentration, 10 ppm.
b) Buffer, 10^{-2} M imidazole + tetrabutylammonium bromide (TBABr) 1.10^{-3} M (pH 4.5); applied voltage, + 25 kV; thermostatted capillary dimensions, 70 cm x 50 μm I.D.; wavelength detection, 214 nm; electromigration injection, 5 s at 10 kV; solute concentration, 10 ppm. Cations: 1) K^+; 2) Ba^{2+}; 3) Ca^{2+}; 4) Na^+; 5) Mg^{2+}; 6) Li^+.

following equation:

$$v_m(i)=\left[m_{ep}(i)+m_{eo}\right].E \quad (1)$$

where $m_{ep}(i)$ is the electrophoretic mobility of cation i, m_{eo} the electroosmotic mobility, and E is the electric field. The reciprocal migration time may be expressed as a function of the electrophoretic mobility $m_{ep}(i)$ following equation 2:

$$\frac{1}{t_m(i)}=\frac{1}{t_0}+\frac{m_{ep}(i).V}{L_t.L_d} \quad (2)$$

where L_t represents the total column length, L_d the column length from the injector to the detector, V the electric voltage, t_o the migration time of a neutral compound, and $t_m(i)$ the migration time of cation i. Moreover, the electrophoretic mobility depends on the ionic strength I of the medium according to the Debye-Hückel theoretical equation 3:

$$m_{ep}(i)=m_{ep}^{\infty}(i)-\left(0.23Z_i^2.m_{ep}^{\infty}(i)+31.4.10^{-5}Z_i\right).I^{\frac{1}{2}} \quad (3)$$

where $m_{ep}^{\infty}(i)$ is the electrophoretic mobility of cation i at infinite dilution and Z_i its electrical charge. Suppose that only the strength of Coulomb and Stokes is exerted on cation i, then its equivalent conductance limit λ_i^{∞} is directly proportional to the electrophoretic mobility according to the Nernst-Einstein relationship 4:

$$\lambda_i^{\infty}=Fm_{ep}^{\infty}(i) \quad (4)$$

where F is the Faraday constant. For each cation i, we may assume that $m_{ep}(i)$ was close to $m_{ep}^{\infty}(i)$ due to the low ionic strength (I = 0.00965) of the imidazole buffer (10^{-2} M, pH 4.5). Thus, the value of time t_o, and consequently, of the electroosmotic flow can be determined by plotting the graph $1/t_m(i) = f(\lambda_i^{\infty}.V/F.L_d.L_t)$.

$$\frac{1}{t_m(i)}=\frac{1}{t_0}+\frac{\lambda_i^{\infty}.V}{F.L_d.L_t} \quad (5)$$

According to the experimental data related to the Fig.2 , the reciprocal migration time varies linearly with the quantity, $\lambda_i^{\infty}.V/F.\ L_d\ .\ L_t$., (see Fig. 7); the experimental slope is equal to 0.95 (close to theoretical value unity) and the time t_o is found to be equal to 9.9 min from the original ordinate value. The equivalent conductance limits λ_i^{∞} of alkali and alkaline-earth cations are sufficiently different for their separation to be achieved in free solution [15] as shown in Fig. 2. Migration times decrease with the equivalent conductance limit, according to the following decreasing migration time order for the cations: potassium (λ_{K^+} = 73.5 mho-

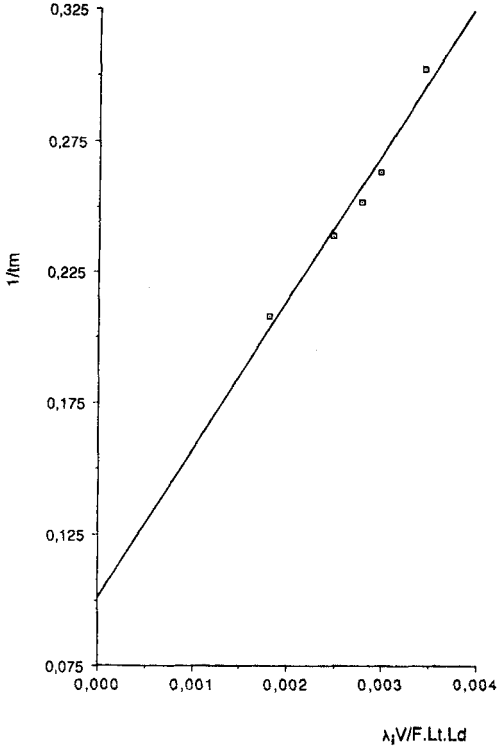

Figure 7. Determination of the electroosmotic flow plotted against the inversed migration time $[t_m(i)]$ of each cation as a function of $\lambda_i^{\infty} V/FL_dL_t$, where λ_i^{∞} is the equivalent conductance limit (mho-cm^2/eq) of the cation i, L_t the total column length (cm), L_d the column length from the injector to the detector (cm), V the electrical voltage, and $t_m(i)$ the migration time of the cation (i).

Buffer, imidazole 10^{-2} M, pH 4.5. Capillary dimensions: 70 cm x 50 μm I.D; UV detection, 214 nm; hydrodynamic injection, 3 s; applied voltage, + 24.8 kV; temperature, 40°C; concentration of cations, 10 ppm.

cm^2), baryum ($\lambda_{Ba^{2+}}$ = 63.6), calcium ($\lambda_{Ca^{2+}}$ = 59.5), magnesium ($\lambda_{Mg^{2+}}$ = 53.1), sodium (λ_{Na^+} = 50.1), and finally lithium (λ_{Li^+} = 38.7). However, magnesium having the slowest migration time compared to that of sodium is contradictory to the order of their respective equivalent conductance limits unless we suppose a possible complexation of these cations by imidazole or a strong interaction of magnesium with the silica [11].

Dependence of Efficiency versus Injection Time and Solute Concentration

As would be expected for a capillary separation technique, the capacity of capillary electrophoresis is limited. The efficiency of the electrophoretic peaks was determined for these cations analyzed in different experimental conditions with a no-thermostatted capillary. The experimental influence of the injected cation concentration and of the hydrodynamic injection time on the number of theoretical plates, N, were studied with the imidazole buffer (10^{-2} M, pH 4.5).

Fig.8a shows the plots of the relative number of theoretical plates N/N_{max} as a function of cation concentration for sodium and lithium (N/N_{max} is the highest number observed for a concentration of 1 mg/L). The separation voltage (+12 kV) and the hydrodynamic injection time (2 s) were kept constant while the cation concentration varied froml 1 μg/mL to 10 μg/mL. The highest efficiency (200 300 for sodium and 177 100 for lithium) was observed at the lowest injected concentration. A notable decrease in efficiency was observed with the concentration (Fig.8), as already mentionned by ROW [16]; thus, there were a decrease in the number of theoretical plates for sodium peak (from 200 300 to 110 200) and for lithium peak (177 100 to 55 000) when the cation concentration rises up from 1 to 10 μg/mL

Just as, with a constant concentration, an increasing injected volume brings about a loss in efficiency of the electrophoretic peaks (see Fig. 8b); thus there was a 47% decrease in the number of theoretical plates for sodium peaks (from 175 000 to 93 000) when the injection time was increased from 1 to 5 s for a concentration of 1 ppm. This study confirms the rather limited column capacity in free zone electrophoresis.

Quantitation

By using hydrodynamic injection, the sample curves linking the corrected surface variations of the electrophoretic peaks to the analyzed cation concentrations are plotted for cations K^+ and NH_4^+, Na^+, Ca^{2+} and Mg^{2+} (Figs. 9a, 9b). The calibration curves were determined with either an imidazole buffer 10^{-2} M, pH 4.5 (experimental conditions: voltage, +18 kV; hydrodynamic injection time, 3 s), or with a benzylamine buffer 10^{-2} M, pH 9.0 (experimental conditions: voltage, +24 kV; hydrodynamic injection, 10 s). The linear

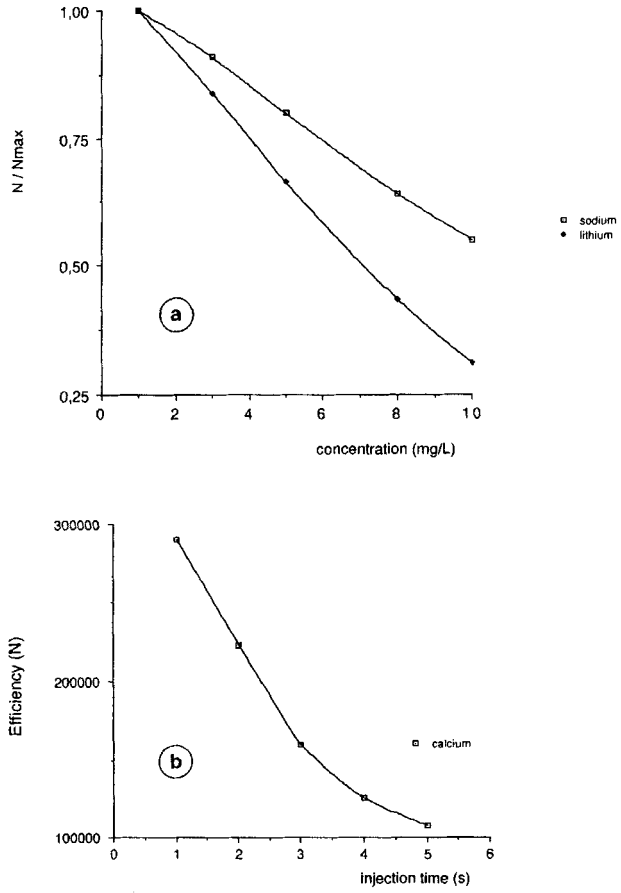

Figure 8. Effect of injected solute amount on peak efficiency.

a) at constant hydrodynamic injection time (2 s);

b) at constant solute concentration (10 ppm).

Buffer, imidazole 10^{-2} M (pH 4.5); capillary, 65 cm x 75 μm I.D; UV detection at 214 nm; hydrodynamic injection, 3 s; applied voltage, + 18 kV.

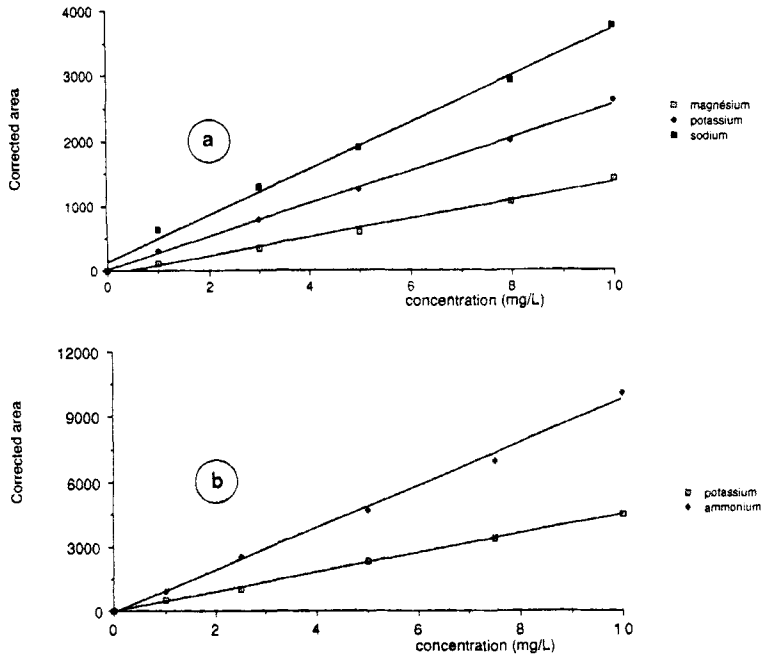

Figure 9. Relationships between concentrations (ppm) of the cations and corrected peak areas (arbitrary units).
a) buffer, imidazole 10^{-2} M (pH 4.5); capillary, 65 cm x 75 μm I.D.; UV detection at 214 nm; hydrodynamic injection, 3 s; applied voltage, + 18 kV. b) buffer, benzylamine 10^{-2} M (pH 9.0); capillary 65 cm x 75 μm I.D.; detection at 204 nm; hydrodynamic injection, 10 s; applied voltage, + 24 kV.

regression coefficients were equal to 0.999 for K^+ and NH_4^+. Using a longer hydrodinamic injection time, the separation of a 50 ppb-level cation mixture may be achieved without any resolution loss (Fig.10).

Analysis of mineral Waters

Fig. 11a shows the capillary electrophoresis separation of alkali and alkaline-earth cations found in tap water using an imidazole buffer (10^{-2} M, pH 4.5). The electrophoretic peaks correspond to potassium, sodium, calcium and magnesium, respectively. Generally, the alkali cations Na^+, Ca^{2+}, and Mg^{2+} present in mineral waters are of very different quantities

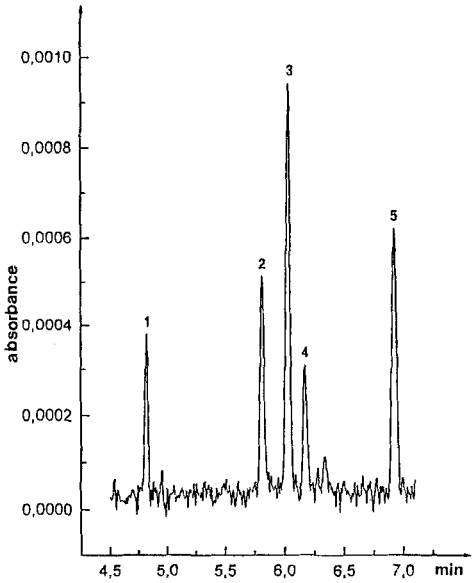

Figure 10. Separation of sub-ppm level cation mixtures by capillary electrophoresis. Buffer, 10^{-2} M imidazole (pH 4.5); applied voltage, + 10 kV; thermostatted capillary dimensions, 57 cm x 75 μm I.D.; wavelength detection, 214 nm; current, 7 μA; hydrodynamic injection, 20 s (except for 5 s for 4 ppm); temperature, 25°C. Identification: 1) K^+; 2) Ca^{2+}; 3) Na^+; 4) Li^+.
Cation concentration: 50 ppb.

which make the quantitative analysis less accurate, especially in the case of the sodium cation. An alternative solution to maintain the resolution would consist by adding TBABr (Fig. 11b) or methanol to decrease the EOF value (Fig. 11c) to the electrolyte; the resolution of the Ca^{2+}/Na^+ and Ca^{2+}/Mg^{2+} peaks is improved at the small expense of an increase in analysis time. In particularly, Fig. 11c shows the analysis of mineral water by capillary electrophoresis using an imidazole buffer (10^{-2} M, pH 4.5) with 20% CH_3OH; the concentration ratio was nearly 70 between Ca^{2+} and Na^+, and 12 between Na^+ and Mg^{2+}.

CONCLUSION

The separation of alkali (sodium, potassium) and alkaline-earth (magnesium, calcium, barium) cations and also ammonium cation was achieved by capillary electrophoresis

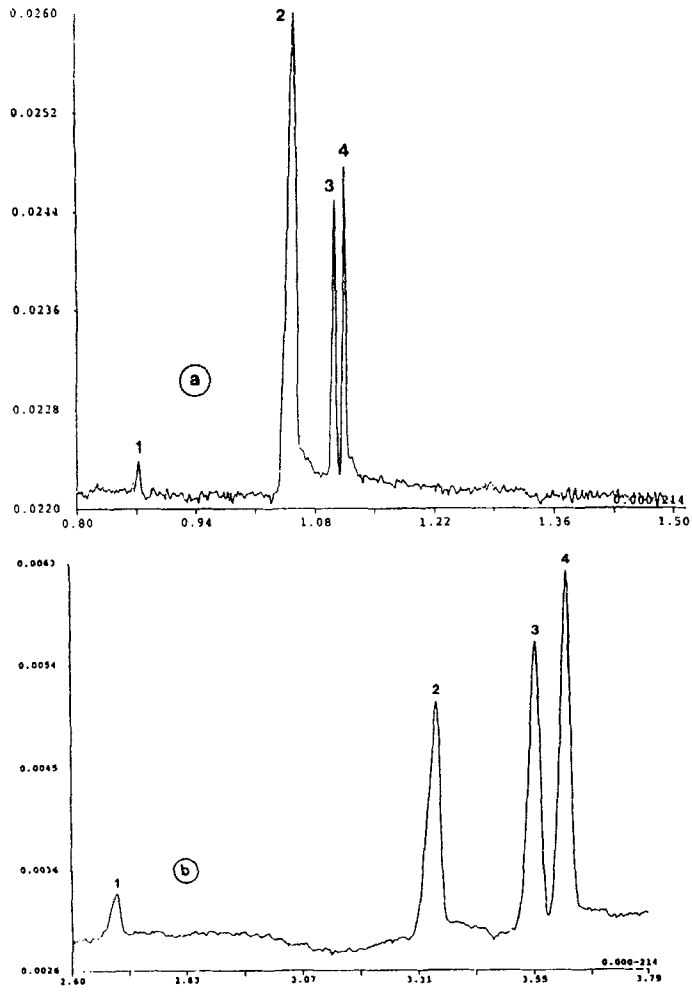

Figure 11 Separation of inorganic cations by CE in several waters.

a) Tap water; applied voltage, + 30 kV; thermostatted capillary dimensions 44 cm x 50 µm I.D.; wavelength detection, 214 nm; hydrodynamic injection time, 0.5 s; buffer 10^{-2} M imidazole (pH 4.5).

b) Mineral water; buffer, 10^{-2} M imidazole + TBABr 5.10^{-3} M (pH 4.5); applied voltage, 10 kV; capillary dimensions, 44 cm x 50 µm I.D.; wavelength detection, 214 nm; hydrodynamic injection time, 1 s.

c) Mineral water; buffer: 10^{-2} M imidazole + TBABr 10^{-3} M (pH 4.5) + 20% methanol; applied voltage, 8 kV; capillary dimensions, 44 cm x 50 µm I.D.; wavelength detection, 214 nm; hydrodynamic injection time, 1 s; temperature, 40°C.

Cations: 1) K^+; 2) Ca^{2+}; 3 Na^+; 4) Mg^{2+}.

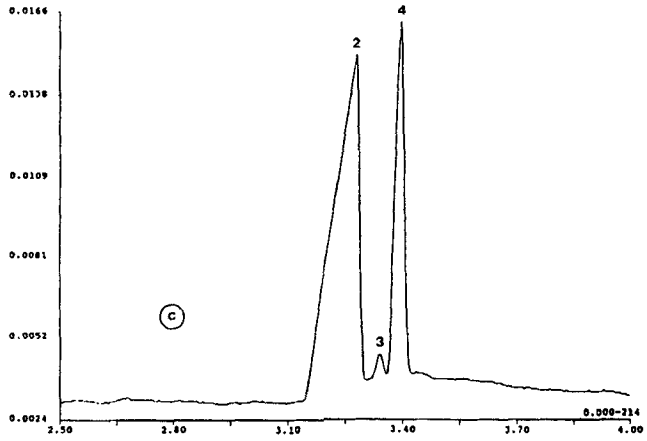

Figure 11 (continued)

using imidazole- or benzylamine-based electrophoretic buffers (pH 4.5 or 9.0, respectively) allowing indirect UV detection with carrier electrolytes.

Using an imidazole buffer (10^{-2} M, pH 4.5), the separation of cations K^+, Na^+, Ca^{2+}, Mg^{2+}, Ba^{2+} and Li^+ was carried out in a few minutes with excellent resolution (in particular for the Na^+/Ca^{2+} couple).

The separation of cations K^+ and NH_4^+ is possible using an alkaline electrophoretic buffer (pH 9.0) in order to decrease the electrophoretic mobility of the ammonium cation, hence differentiating it from the potassium cation.

The advantage of capillary electrophoresis over other separation techniques comes from its simple equipment, the use of a constant composition buffer and analysis speed compatible with routine analyses (especially for mineral waters).

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