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Research Article

Separation of cations by capillary electrophoresis: The effects of temperature and voltage

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ABSTRACT

Cation analysis plays a crucial role in various industries, including food, agriculture, and environmental monitoring. This study explored capillary electrophoresis for the separation of ammonium, potassium, sodium, calcium, magnesium cations, focusing on the effects of temperature (20-30°C) and voltage (20-30 kV) on peak and separation parameters: migration time, plate number, response factor, tailing factor, resolution, and selectivity. The shortest migration times for cations (from ammonium to magnesium) were observed at 30°C-30 kV (from 4.27 to 7.11 min), while the longest times (from 7.45-12.9 min) occurred at 20°C-20 kV. The temperature significantly interacted with voltage on the migration of cations (p<0.05). The highest plate number was observed for magnesium (36800), followed by calcium (32031), potassium (14712), ammonium (6927), and sodium (5060). Temperature increases significantly (p<0.05) improved sodium's plate number (from 2560 to 5060) but reduced it for other cations. Both temperature and voltage significantly (p<0.05) enhanced peak symmetry for sodium, as indicated by increase in its tailing factor (from 0.54 to 0.58 and from 0.54 to 0.57, respectively). Furthermore, the temperature and voltage showed a significant (p<0.05) interaction on the tailing factors of ammonium and potassium. Resolution between sodium and calcium was suboptimal (<1) at 20°C-30 kV but reached to 2.19 at 30°C-30 kV. These findings demonstrate that higher temperature and voltage conditions enhance separation efficiency, particularly for sodium, by improving plate number, peak symmetry, and resolution. This study presents a comprehensive quantitative comparison and offers specific capillary electrophoresis conditions to facilitate further research and method validation for cation analyses.

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INTRODUCTION

The cations of sodium, potassium, calcium and magnesium are essential minerals that play crucial roles in human health. High intake of sodium is linked to hypertension while low levels of calcium can affect the bones [1]. The most common reduction of sodium is by substituting with potassium, hence providing health benefits such as blood pressure

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reduction, decreased cardiovascular disease, and kidney injury [2]. Calcium is essential in the maintenance of bone health and cellular functions, while magnesium is essential in enzyme activity and energy metabolism. Ammonium is a cation derived from ammonia (NH₃), a common compound in soil and water. It plays a crucial role in the nitrogen cycle and is available to plants for nutrition [3]. In water, the cations also play an important role in taste, hardness, and its suitability for drinking, agriculture and industrial applications [4]. Similarly, in many countries around the world, including Türkiye, labelling regulations require very explicit reporting on mineral cation content in foodstuffs and beverages; these further applications the need for appropriate and reliable analytical techniques [5]. Sodium adsorption ratio (SAR) calculated from the relative concentration of sodium to calcium and magnesium is widely used to assess the suitability of water for agricultural irrigation since it provides an indication of the potential impact of sodium on the structure and permeability of the soil [6]. Sodium and potassium are essential in formulating intravenous fluids and medications to ensure isotonicity. Calcium and magnesium are widely used in pharmaceutical industries as supplements to address deficiencies and as antacids acid, respectively [7]. The knowledge of the behavior and concentration of the cations present in foodstuffs, determination of water quality, industrial processing, and monitoring would be significant in applications. The precision of cation analysis, hence, is principle in ensuring product quality and consumer health, apart from being an inevitable tool in the enforcement of regulatory requirements.

The most common techniques followed for the determination of cations in such natural samples are atomic absorption spectroscopy (AAS), flame photometry, and ion chromatography [8, 9, 10]. AAS generally offers lower detection limits and can analyze a wider range of elements than flame photometry. While flame photometry is easier to operate, making it preferred choice for analysis of major cations [11]. Ion chromatography (IC) also separates cations based on their interaction with a stationary phase and a mobile phase [8]. While these methods are effective, they are costly, require extensive maintenance, and may need larger sample volumes. Therefore, an alternative simple method which can determine the cations in natural samples has been tried to be obtained by capillary electrophoresis (CE). CE is a powerful analytical technique widely used for the separation of ions with many advantages compared to other analytical techniques. CE has even produced higher separation efficiency compared to AAS and IC [12, 13] in testing of water samples while consuming lower sample volumes [8, 14, 15]. However, gaps remain regarding the effects of temperature and voltage on peak and separation parameters during the analysis of capillary electrophoresis. The effects of temperature and voltage in CE can be related to two phenomena: joule heating and electroosmotic flow (EOF). Although, voltage increases the EOF and accelerates the migration, applying excessive voltage may negatively

affect both the resolution of analytes and the efficiency of the capillary due to the heating effect [16, 17]. An increase in temperature lowers the viscosity of the buffer, which in turn increases the EOF and ion mobility, causing the analyte to migrate faster. However, an excessively high temperature can clearly accelerate EOF, leading to a loss of resolution and selectivity [18, 19]. Knox and McCormack [20] stated that Joule heating in capillaries increases molecular diffusion resulting in a smaller than expected rise in the plate number, causing peak broadening, and decreasing in separation efficiency. Besides, Joule heating decreases resolution owing to the increased thermal diffusion which makes the sample broaden with the consequence of reducing separation efficiency [21]. A low resolution between the Mg²⁺ and Ca2+ cations had been observed in works dealing with the analysis of cations by CE, with applied voltages of 15 kV and 20 kV [15, 22]. The application of a voltage of 30 kV resulted in low resolution for the Na⁺ and Mg²⁺ cations [23]. Despite its potential, the interactive effects of temperature and voltage on the peak and separation characteristics of cations are not well known for multi-cation analysis.

In this study, it was aims to determine the temperature (20-30°C) and voltage (20-30 kV) that can improve migration time, plate number, peak symmetry, resolution, and selectivity in cation analysis by CE. By addressing these operational conditions, the findings are expected to improve the applicability of CE in industries where precise cation analysis is critical for quality control, and regulatory compliance.

MATERIALS AND METHODS

Standards

The cation standard test sample containing 100 ppm each of ammonium ($\mathrm{NH_4^+}$), potassium ($\mathrm{K^+}$), sodium ($\mathrm{Na^+}$), calcium ($\mathrm{Ca^{2+}}$), and magnesium ($\mathrm{Mg^{2+}}$) was used (5064-8205, Agilent Technologies, Santa Clara, CA, USA). Buffer solution (5064-8203, Agilent Technologies) was filtered using 0.22 µm syringe filter and degassed for 5 min using an ultrasonic bath (621.06.003, ISOLAB GmbH, Germany) prior to runs. The pH values were 2.8 for the cation standard and 3.2 for the buffer, respectively. The water used to CE analysis was purified by passage through an Ultra-Pure Milli-Q plus water system from Millipore (Millipore, Milford, MA, USA). A standard NaOH solution (1 N, 5062-8576, Agilent Technologies) was used for conditioning the capillary.

Instrumentation

Separation of cations was carried out using a capillary electrophoresis system (Agilent G7100) coupled with a real-time UV-Visible diode-array detector (190 to 600 nm) and interfaced with ChemStation software (Agilent Technologies, Santa Clara, CA, USA). The separation capillary used was a standard bare fused silica with 80.5 cm

total length and an internal diameter of 50 μ m (G1600-62211, Agilent Technologies). The distance from the point of injection to the deuterium lamp on-column detection was 72 cm.

Procedures

The cation standard sample was diluted to 25 ppm with MilliQ water and filtered using a 0.22 µm PTFE syringe filter. Separation of cations by CE was performed according to the procedure described by Varden and Bou-Abdallah [24]. Prior to use, the capillary was conditioned with 0.1 M NaOH for 10 min, ultra-pure water for 10 min, and running buffer for 15 min as recommended by the manufacturer. Between each run the capillary was purged with buffer for 5 min. The buffer was replaced after every change in operation conditions. The cations were separated under constant voltage (positive polarity) and temperature of the capillary were as follows: 20 kV and 20°C, 20 kV and 30°C, 30 kV and 20°C, 30 kV and 30°C, respectively. These ranges were chosen to evaluate their effects on the resolution and migration of cations. The voltage directly affects the electrophoretic migration of analytes; higher voltage generally leads to faster migration and/or shorter analysis times, but with a potential decrease in resolution. Similarly, the temperature of the capillary affects the buffer viscosity and ion mobility; higher temperature generally reduces viscosity and increases ion separation. Also, the parameters were determined by previous studies [14, 22-25], ensuring compatibility with the CE system.

Samples were injected by electro-migration at 5 kV for 5 s. The detector was set at UV signal of 280 nm (bandwidth 20 nm), with an indirect reference signal of 214 nm (bandwidth 10 nm), a response time of 2.0 s, and a sampling rate of 2.5 Hz. Total run time was 15 minute. Each sample was run at least three times. At the end of each day's analyses, the capillary was washed with purified water for 15 min and then flushed with air for 3 min.

Peak and Separation Characteristics

Migration time (Mt) refers to the amount of time it takes for an analyte to migrate from the point of injection to the detector. The plate number (N), also known as the number of theoretical plates, was calculated using the following formula:

$$N = 5.54 \left(\frac{Mt}{W_{1/2}} \right)^2$$
 (Eq. 2.1)

Where:

Mt is the migration time.

 $W_{1/2}$ is the width of the peak at half-height.

The response factor (Rf) is calculated as the ratio of the peak area to its corresponding known concentration (25 ppm). Tailing factor (Tf) was calculated at 5% of the peak height using the following formula:

Tf =
$$\frac{W_{0.05}}{2f}$$
 (Eq. 2.2)

Where:

 $W_{0.05}$ is the width of the peak.

f is the distance from the peak apex to the leading edge of the peak.

Selectivity (a) is defined as the ratio of the effective electrophoretic mobilities of two analytes. It is calculated as the ratio of the migration times for two adjacent peaks [26]. Resolution (Rs) was calculated using the following formula:

Rs =
$$\frac{2\left(M_{t2} - M_{t1}\right)}{W_{b1} + W_{b2}}$$
 (Eq. 2.3)

Where:

Mt₁ and Mt₂ are the migration times of the first and second peaks, respectively.

 W_{b1} and W_{b2} are the baseline widths of the first and second peaks, respectively.

Statistical Analysis

Peak and separation characteristics were subjected to a (2x2)x3 factorial analysis of variance with fixed factors of 2 voltage levels (20 kV and 30 kV) and 2 temperature levels (20°C and 30°C) in a completely randomized design. The general linear model is as follows:

$$Y_{ijk} = \mu + \alpha_i + \beta_j + (\alpha\beta)_{ij} + e_{ijk}$$

Where.

Y_{iik}, recorded the value of separation parameters

μ, mean of the population

 α_i , the effect of ith voltage group i= 1-2

 β_i , the effect of jth temperature group j= 1-2

 $\alpha\beta_{ij},$ the effect of relationship between voltage and temperature

e_{iik}, error term

The parameters of migration time and tailing factor for which the interaction between temperature and voltage was found to be significant, it was illustrated using an interaction plot. Differences between the mean values were analyzed using one-way ANOVA and Duncan's multiple comparison test. All statistical analyses and interaction plots were performed using the SPSS package program (Version 22, IBM Statistics, USA).

RESULTS AND DISCUSSION

Peak Characteristics

The electropherogram of cations separated by CE under different temperature and voltage conditions was shown in Fig. 1. The effect of temperature and voltage applied to the separation of cations on migration time of cations was shown in Fig. 2. Ammonium was the first migrated cation, followed by K^+ , Na^+ , Ca^{2+} and Mg^{2+} . The longest and the shortest migration times of cations were at $20^{\circ}\text{C}-20$ kV (from 7.45 to 12.9) and $30^{\circ}\text{C}-30$ kV (from 4.27 to 7.11) conditions, respectively (Fig. 1 and 2). This can also be attributed to the lower viscosity of the buffer at higher temperatures, which enhances the electroosmotic flow (EOF) and the mobility of cations [14, 19].

The shorter migration times have been previously reported for the same cations separated by CE [15, 27]. However, Opekar and Tuma [28] reported higher migration times for cations than those observed in the present study. It was also reported that cations in drinking water [29] and milk [30] migrated in less than 5 minutes at 25 kV and 20 kV, respectively. These differences could be attributed to the column and/or buffer used in the separation of cations by CE. Rizelio et al. [15] separated cations in honey using a 48.5 cm length capillary under 15 kV applied voltage within 1.5 minutes.

As shown in Figure 2, the temperature interacted with voltage to affect the migration time of all cations (P<0.05). This finding is consistent with the statement of Rathore [19], who reported that the degree of Mt shortening with increasing temperature is greater at higher voltages. It can also be stated that the increase in voltage also shortens the Mt even more as the temperature increases (Fig. 2). As expected, voltage increased the EOF of cations, while temperature reduced the time cations spent in the capillary column. The EOF refers to the bulk flow of the electrolyte solution in the capillary, which is driven by the applied electric field [21]. Increasing the voltage increases the electric field strength, which generally leads to faster migration times for cations. Higher temperatures can increase the kinetic energy of the ions, which may enhance their mobility. Similarly, Cheng et al. [31] reported that the migration times of cations decreases as the voltage increased from 18 kV to 30 kV.

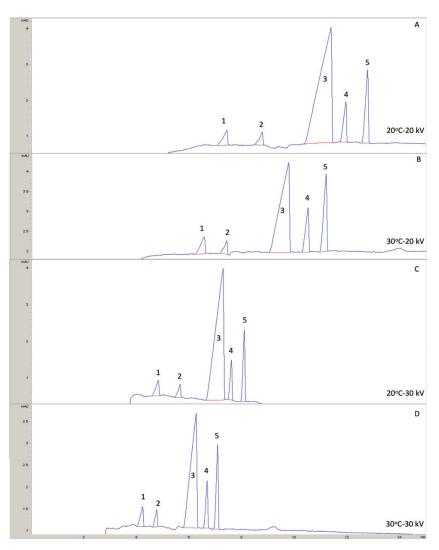


Figure 1. Electropherogram of cations (1: NH₄⁺, 2: K⁺, 3: Na⁺, 4: Ca²⁺, 5: Mg²⁺) separated by capillary electrophoresis under 20°C-20 kV (A), 30°C-20 kV (B), 20°C-30 kV (C), and 30°C-30 kV (D).

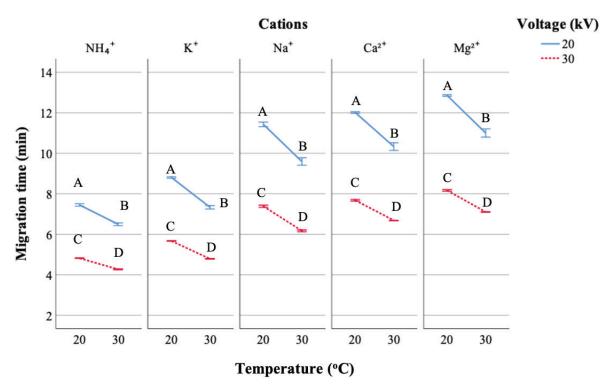


Figure 2. The effect of interaction between temperature and voltage on the migration time of cations. Error bars indicate standard errors (n=3). A-C Different capital letters indicate significant differences between samples (p<0.05).

The plate number of cations separated by CE under different temperature and voltage were given in Table 1. The plate number (N), also referred to as the number of theoretical plates, serves as a key parameter for assessing efficiency of the chromatographic separation [32]. The highest N value was observed for Mg²⁺ (36800), followed by Ca²⁺ (32031), K⁺ (14712), NH₄⁺ (6927), and Na⁺ (5060). Opekar and Tuma [28] observed a similar ranking for cations (Mg²⁺>Ca²⁺>K⁺>NH₄⁺) with respect to plate number. The plate numbers of K⁺, Ca²⁺ and Mg²⁺ cations separated by CE at 25 kV and 25°C were determined as 5500, 37000 and 42000, respectively [33]. Shi and Fritz [34] stated that the plate number of cation peaks depends on the concentration injected to CE and determined the N value of 100 ppm Zn²⁺ to be lower than 100000. As shown in Table 1, temperature

had a significant (p<0.05) effect on the plate number of all cations. An increase in temperature significantly (p<0.05) increased the N value for Na⁺ (from 2560 to 5060), while it significantly (p<0.05) decreased the N values for the other cations (Table 1). The plate numbers for Ca²⁺ and Mg²⁺ were higher when analyzed at 20 kV compared to 30 kV at both temperatures (Table 1). This can be attributed to the 'Joule heating' as a result of high voltage, as reported by Copper [16]. This could be attributed to increase in temperature reduced the viscosity of the buffer, thereby enhancing the mobility and separation of Na⁺ more significantly than the others. As regard to other cations, factors such as increased diffusion or thermal broadening may have counteracted the advantages of reduced viscosity [19, 20].

Table 1. Plate number of cations separated by capillary electrophoresis under varying temperatures and voltages

Cations	Factors of temperature (T) and voltage (V)				Significance		
	20°C-20 kV	20°C-30 kV	30°C-20 kV	30°C-30 kV	Т	V	TxV
NH ₄ ⁺	9337±1313	7688±631	6807±1148	7046±626	*	ns	ns
ζ+	19719±1572	17610±964	15118±2348	14305±1484	**	ns	ns
Na+	3063±1815	2077±351	3995±1025	6124±2735	*	ns	ns
Ca^{2+}	45957±2247	44255±1469	33491±1461	30570±633	***	*	ns
Mg^{2+}	57452±136	51770±880	39075±1791	34525±233	***	***	ns

The data were expressed as mean±standard deviation (n=3). *p<0.05, **p<0.01, ***p<0.001, ns: not significant (p>0.05)

The response factor of cations separated by CE under different temperature and voltage were given in Table 2. A higher response factor indicates that the detector generates a stronger response for each unit of the standard analyte [35]. The response factor of Na⁺ was 5 to 24 times higher than those of the other cations (Table 2). Lee and Lin [36] also reported that Na+ has a higher peak height than other cations separated by capillary electrophoresis at 25°C and 25 kV. Therefore, Na+ can be determined with better sensitivity by capillary electrophoresis compared to other cations. This also means that analytes with higher response factors naturally exhibit higher calibration slopes. Fung and Lau [14] also determined a higher calibration slope for Na⁺ among the 13 cations analyzed by CE. Since K⁺ has the lowest response factor among the cations (Table 2), its detection limit is expected to be higher than those of the others, which is consistent with the findings of Suarez-Luque et al. [37] and Abe et al. [33].

Among the cations, the response factor for Na⁺ was the only one to decrease significantly (P<0.05) with increasing temperature, regardless of whether the voltage was 20 kV or 30 kV (Table 2). With respect to voltage, no significant difference was observed in the response factors, except for Na⁺ (Table 2). This situation may be due to changes in the plate number of Na⁺ peak. The plate number of the Na⁺ peak was highest at 30°C with 30 kV applied, whereas other cations exhibited the lowest plate numbers under the same conditions. In this context, it appears that the response factor may be negatively affected by the plate number of peak. When the plate number is low, it indicates broader peaks, which can lead to a lower response factor. As plate number is a measure of column efficiency, a higher plate number indicates better separation, reducing peak overlap and improving resolution [38].

The effect of temperature and voltage applied for the separation of cations on tailing factor of cations was shown in Fig. 3. Tailing factor (Tf) of a chromatographic peak is calculated to assess the shape of the peak, particularly how symmetrical it is. An ideal chromatographic peak is characterized as a 'Gaussian peak'. For the perfect peak symmetry, the tailing factor (Tf) should be '1'. As the Tf value deviates negatively from '1', the peak's fronting increases, while

positive deviations indicate increased tailing [35, 39]. All analyzed cations showed fronting with Tf values less than '1'. It is difficult to find peak tailing factor data for cations analyzed by CE in the literature. The cation with the highest fronting was Na+, which exhibited the lowest Tf value (Fig 3). The temperature and voltage interacted to influence the Tf of NH₄⁺ and K⁺, which migrate faster in the capillary than the other cations (Fig 1). This can be attributed to the smaller size and higher electrophoretic mobilities of NH₄⁺ and K⁺ ions, as well as the fact that temperature and voltage accelerate these cations more than the others [20]. As shown in Fig. 3, the tendency for temperature to increase Tf value depends on the applied voltage. The temperature increased Tf significantly only when a voltage of 30 kV was applied to separate the NH₄⁺, K⁺ cations. Thus, applying 30 kV instead of 20 kV for cation separation increased the Tf value, resulting in more symmetrical peaks. Also, the significant interaction between temperature and voltage means that their combined effect was greater than the sum of their individual effects on peak symmetry of NH₄+, K+ cations. This highlights the need to optimize both parameters simultaneously to maximize separation efficiency and peak quality. In applications where precise quantification is required, such as in quality control or environmental monitoring, symmetric peaks ensure accurate analysis.

This could be attributed to the higher voltage application increasing the number of plates. Changes in the tailing factor were consistent with the plate number. In fact, a statistically significant positive correlation (r=0.654, p<0.001) has been identified between the plate number of peaks and the tailing factor. Dyson [38] also noted that tailing negatively affects the plate number by causing peak broadening, thereby reducing separation efficiency.

Separation Characteristics

The separation characteristics of selectivity and resolution determined in cations were shown in Table 3 and Table 4, respectively. As stated by Dursun and Güler [35], selectivity (α), also known as the separation factor or relative retention, is used to describe the ability to distinguish between two analytes based on their electrophoretic mobilities. Since selectivity is defined as the ratio of migration

Table 2. Response factor of cations separated by capillary electrophoresis under varying temperatures and voltages

Cations	Factors of Temperature (T) and Voltage (V)						Significance		
	20°C-20 kV	20°C-30 kV	30°C-20 kV	30°C-30 kV	T	V	TxV		
NH ₄ ⁺	0.20±0.02	0.13±0.01	0.21±0.04	0.13±0.00	ns	***	ns		
$K^{\scriptscriptstyle +}$	0.14 ± 0.01	0.10 ± 0.01	0.13 ± 0.02	0.08 ± 0.01	ns	***	ns		
Na^{+}	4.34±2.52	3.59 ± 0.74	1.95±0.25	0.94±0.64	*	ns	ns		
Ca^{2+}	0.36 ± 0.02	0.23 ± 0.01	0.36 ± 0.01	0.22±0.01	ns	***	ns		
Mg^{2+}	0.62 ± 0.03	$0.40 {\pm} 0.00$	0.63 ± 0.00	$0.40 {\pm} 0.01$	ns	***	ns		

The data were expressed as mean±standard deviation (n=3). *P<0.05, ***P<0.001, ns: not significant (P>0.05)

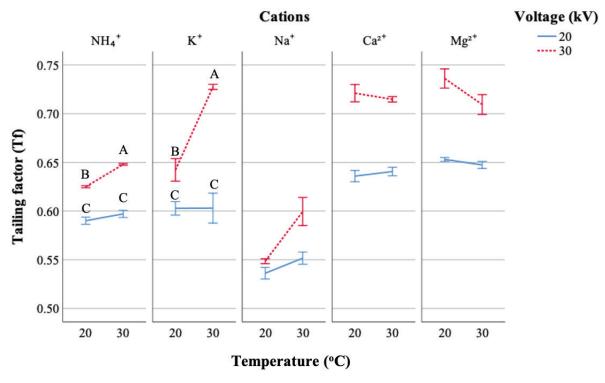


Figure 3. The effect of interaction between temperature and voltage on the tailing factor of cations. Error bars indicate standard errors (n=3). $^{A-C}$ Different capital letters indicate significant differences between samples (p<0.05).

times for two adjacent peaks, the smallest possible value is '1' [26]. The α values of cations were found higher than '1' (Table 3).

The α values alone may not be sufficient for evaluating the separation, as they are calculated solely from migration times. Resolution, being a function of the retention factor, separation factor, and theoretical plate number, serves as a good indicator of how well chromatographic peaks are separated [35]. An increase in temperature has been found to enhances the resolution of $K^+\text{-Na}^+$ and $Na^+\text{-Ca}^{2+}$ (from 4.40 to 5.45 and from 0.94 to 1.98, respectively) but decrease the resolution of $NH_4^+\text{-}K^+$ and $Ca^{2+}\text{-}Mg^{2+}$ (from 4.59 to 2.94 and from 3.59 to 2.86). The better resolution of Na^+ from the preceding K^+ and following Ca^{2+} cations (Table 4), particularly at higher temperatures, was consistent with the

increased plate number (Table 1) and tailing factor (Fig. 3). There was no significant difference in the resolution of Na⁺-Ca²⁺ separated under 20 kV and 30 kV (Table 4). With the increase in voltage from 20 kV to 30 kV, the resolution of NH₄⁺-K⁺ (from 3.93 to 3.60) and Ca²⁺-Mg²⁺ (from 3.41 to 3.04) decreased significantly (p<0.01).

While an Rs value of '1' is acceptable, an Rs value exceeding '1.5' signifies better separation, with consecutive peaks fully separated from each other [26, 40]. As shown in Table 4, good resolution was obtained except for Na⁺-Ca²⁺ separated at 20°C both at 20 kV (1.13) and 30 kV (0.74). Better resolution was determined for Ca²⁺- Mg²⁺ and NH₄⁺- K⁺ compared to cations separated at 20 kV with a 50 cm capillary [22] and 30 kV with a 55 cm capillary [23]. This is most likely due to the longer capillary (80.5 cm total length)

Table 3. Selectivity of cations separated by capillary electrophoresis under varying temperatures and voltages

Cations	Parameters of Temperature (T) and Voltage (V)				Significance		
	20°C-20 kV	20°C-30 kV	30°C-20 kV	30°C-30 kV	T	V	TxV
NH ₄ +- K+	1.18±0.01	1.18±0.00	1.13±0.00	1.12±0.00	***	ns	ns
K+- Na+	1.30±0.03	1.30 ± 0.01	1.31±0.02	1.29 ± 0.02	ns	ns	ns
Na+- Ca2+	1.05±0.02	1.04 ± 0.00	1.08 ± 0.00	1.08 ± 0.01	***	ns	ns
Ca ²⁺ - Mg ²⁺	1.07±0.00	1.06±0.00	1.07±0.00	1.06±0.00	ns	**	ns

The data were expressed as mean \pm standard deviation (n=3). **p<0.01, ***p<0.001, ns: not significant (p>0.05)

Cations	Parameters of Temperature (T) and Voltage (V)				Significance		
	20°C-20 kV	20°C-30 kV	30°C-20 kV	30°C-30 kV	T	\mathbf{V}	TxV
NH ₄ +- K+	4.82±0.17	4.35±0.06	3.03±0.27	2.85±0.09	***	**	ns
K+- Na+	4.64±0.69	4.17±0.14	5.27±0.11	5.64±0.74	**	ns	ns
Na+- Ca2+	1.13±0.59	0.74 ± 0.13	1.76±0.21	2.19±0.72	**	ns	ns
Ca ²⁺ - Mg ²⁺	3.83±0.16	3.35±0.07	2.99±0.04	2.73±0.07	***	***	ns

Table 4. Resolution of cations separated by capillary electrophoresis under varying temperatures and voltages

The data were expressed as mean±standard deviation (n=3). **p<0.01, ***p<0.001, ns: not significant (p>0.05).

used in the present study. It was observed that Na $^+$ and Ca $^{2+}$ had Rs values above '1.5' when analyzed at 30°C. This was compatible with the increased number of theoretical plates for the Na $^+$ peak as temperature increased (Table 1). The Rs values determined for NH $_4^+$ (1.57), K $^+$ (5.64), and Na $^+$ (5.23) were reported by Neaga et al. [41].

CONCLUSION

To our knowledge, this is the first report on the peak and separation characteristics of cations including analysis by capillary electrophoresis at varying temperatures and voltages. The findings demonstrate that an increase in both temperature and voltage significantly reduced the migration times of cations. The efficiency of separation varied among the cations, with magnesium exhibiting the highest plate number. Especially at 30°C, the increase in the plate number of the sodium peak resulted in better separation from the preceding potassium and following calcium peaks, thus enhancing resolution. The interaction between temperature and voltage resulted in a better symmetry for the ammonium and potassium peaks, emphasizing the importance of optimizing both parameters to obtain better results in terms of efficiency. The selectivity and resolution of cations analyzed, particularly at 30°C, indicated better separation ability. Overall, these findings provide valuable insights for improving analytical methods in capillary electrophoresis, potentially enhancing selectivity of cation separation.

Our findings provide a robust method for detecting cations, which can significantly enhance routine quality control practices in the food, agricultural, chemical and medicinal industries. In conclusion, the operating at 30°C with a higher voltage in the separation of cations by capillary electrophoresis could be recommended, especially as it increases the plate number and improves peak symmetry for sodium, thereby enhancing the resolution and providing a shorter analysis time.

AUTHORSHIP CONTRIBUTIONS

Authors equally contributed to this work.

DATA AVAILABILITY STATEMENT

The authors confirm that the data that supports the findings of this study are available within the article. Raw data that support the finding of this study are available from the corresponding author, upon reasonable request.

CONFLICT OF INTEREST

The author declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

ETHICS

There are no ethical issues with the publication of this manuscript.

STATEMENT ON THE USE OF ARTIFICIAL INTELLIGENCE

Artificial intelligence was not used in the preparation of the article.

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