Capillary electrophoretic analysis of inorganic anions in atmospheric hailstone samples

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Abstract

A new application of capillary electrophoresis (CE) for measuring inorganic anions in hailstones was carried out. Five hailstone specimens were collected from large blocks of ice that fell in January 2000 in some provinces of Spain. Sample handling and preparation procedures were performed with care. CE analysis of anions was carried out using indirect UV detection at 254 nm with a negative power supply (-15 kV) and hydrostatic injection (10 cm for 30 s) at 35 °C. Anion separation was completed in less than 4 min. The working electrolyte consisted of 4.7 mM sodium chromate, 4.0 mM OFM-OH (tetradecyltrimethylammonium hydroxide), 10 mM CHES [2-(N-cyclohexylamino)ethanesulfonic acid], and 0.1 mM calcium gluconate (pH 9.1). Good repeatability of migration times after eight injections ($<0.7\%$ RSD), adequate linearity responses ($r^2 > 0.9$) as well as satisfactory detection limits (<0.35 ppm) were achieved. The analytical results provided by CE were compared with those obtained by traditional wet-chemical (WCH) methods. Most of the results obtained by CE were consistent with those of WCH, except for one sample. © 2002 Elsevier Science B.V. All rights reserved.

Keywords: Hailstones; Inorganic anions

1. Introduction

Devastating natural hazards related to the occurrence of urban hailstorms have been reported throughout history [1–3]. Blocks of ice, called hailstones, are formed in the atmosphere by complex heterogeneous nucleation processes at low atmospheric temperatures [4,5]. The hailstones that have fallen at several locations of the Earth have shown a wide variability in shape, size and weight [3–5]. The magnitude of these physical properties, together with the possible intervals of recurrence, cause hailstones to become a serious risk of damage for people, buildings, and aircraft engines [2,6].

From 10 to 17 January 2000, numerous unusually large hailstones (weighing from 300 g to 4 kg) fell in some provinces of Spain under atypical atmospheric conditions [3,4,7]. Five hailstone specimens were carefully collected and preserved under refrigeration. Geochemical studies based on the chemical and isotopic composition of these samples are currently being carried out to determine their origin and structure and also to define the nucleation mechanisms and atmospheric processes that enabled the

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hailstones to be formed [3,4]. The determination of the isotopic and chemical composition of these samples was defined as a fundamental analytical task. Measurements of oxygen ($\delta^{18}O$) and deuterium (δ^2H) showed a notable isotopic variability in some samples [3]. These isotopic data showed that these samples would be expected to reveal a complex chemical composition. The use of reliable and highly sensitive analytical techniques was therefore required for performing the chemical analyses.

For inorganic cations and metal ions, atomic absorption spectrometry and inductively coupled plasma mass-spectrometry techniques were employed, respectively [3]. For the analysis of anions, traditional wet-chemical (WCH) methods were used initially. Unfortunately, the results provided by these techniques were not totally satisfactory due to the poor sensitivity exhibited for various anions. Since the concentration of some anions in hailstones was sub-ppm and the available amount of sample was limited, a low-cost and multi-elemental technique, with low detection limits and a small consumption of sample, was needed. Inorganic anions can be measured either by ion chromatography (IC) or capillary electrophoresis (CE) [8–10]. Such multi-elemental techniques have been defined appropriately as complementary analytical tools rather than competitive [9]. Today, IC dominates in most chemical laboratories involved in the analysis of anions using conductivity detection [10,11]. The advances achieved in IC would seem to show that this technique is the most reliable and sensitive method for measuring anions. However, for the analysis of sample volumes smaller than 100 μL, IC is not recommended as a suitable technique [12,13]. This technical feature constitutes a serious disadvantage in analyses where the total amount of sample available is rather limited.

CE is an emerging analytical technique exhibiting low sample requirements, low detection limits, high resolution, and rapid separation [12,14,15]. CE uses differences in the electrophoretic mobilities of the analytes to provide an efficient separation [14–16]. The measurement of indirect absorbance is usually the detection mode preferred in CE for analyzing inorganic and low-molecular-mass anions [8,15–18]. Hydrostatic and electrokinetic injection have been used successfully for analyzing anions at low con-

centrations [8]. The advances in CE have made it a mature and well-established technique for the analysis of anions in various matrices (e.g. Refs. [8,14–17]). Numerous CE methodologies for the analysis of anions in different environmental water samples have been developed [12,18–21]. However, for the analysis of anions in atypical atmospheric materials, such as hailstones, ice crystals, ice cores, snow, raindrops or fog drops, the use of CE has seldom appeared in the literature (e.g. Refs. [12,13,19]). In order to extend the range of application of CE and to demonstrate its high sensitivity as well as its sample and time saving capabilities, further work is currently being carried out in different laboratories. The present work reports a new evaluation of CE for performing an efficient separation and quantification of anions in unusual hailstone samples. This work describes a simple procedure for studying the indirect UV detection and behavior of analytes in a background electrolyte of sodium chromate at different temperatures. The present CE methodology was applied according to the sample and analysis requirements and was evaluated in terms of precision and accuracy. A comparison between the analytical results provided by CE and those obtained by WCH is also presented.

2. Experimental equipment

2.1. Capillary electrophoresis instrumentation

The CE instrument used was a Quanta 4000 (Waters, Milford, MA, USA). The configuration included a negative power supply (-15 kV), an indirect UV detection system, and a conventional fused-silica capillary (60 cm total length×75 μm I.D.×375 μm O.D.) with the detection window placed 7.5 cm from the receiving electrolyte end to the detector cell. The UV detection system was set at a wavelength of 254 nm using a mercury lamp. The capillary was maintained at a temperature of 35 °C. All samples were introduced into the capillary by hydrostatic injection (by elevating the sample 10 cm for 30 s). Instrument operation, data acquisition, and peak integration processes were controlled by a Waters 820 Workstation. Electropherogram data were logged at a transfer rate of 20 points per second

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using the Millenium 2000 software. Each day before commencing the experiments, the instrument was pre-conditioned by flushing for 10 min with 0.1 *M* NaOH followed by 10 min with Milli-Q water and then stabilized by flushing for 10 min with the working electrolyte. Before each run, the capillary was rinsed with Milli-Q water and equilibrated with the running electrolyte for 2 min in order to minimize the variability of the migration times of each analyte.

2.2. Chemical reagents and procedures

All chemical reagents were of analytical grade. Electrolyte and standard anion solutions were prepared with Milli-Q water (18 mΩ cm) produced by a purification system (Millipore, Barcelona, Spain). The working electrolyte was prepared from a mixture of 4.7 m*M* sodium chromate, 4.0 m*M* OFM-OH (tetradecyltrimethylammonium hydroxide) obtained from Waters, 10 m*M* CHES [2-(*N*-cyclohexyl-amino)ethanesulfonic acid], and 0.1 m*M* calcium gluconate (pH 9.1). Fresh working electrolyte was prepared daily, filtered through a 0.45 μm membrane filter (Millipore), and degassed. Stock solutions of inorganic anions at a concentration of 1000 mg L^{-1} were prepared by dissolving dried sodium or potassium salts following the standard procedures suggested by the American Society for Testing and Materials (ASTM) [11]. Standard working solutions were prepared weekly by diluting the stock solutions with Milli-Q water.

2.3. Sample collection and handling

Five hailstones that fell in several provinces of Spain were carefully collected in polypropylene

containers previously washed with Milli-Q water [3,4]. Information concerning hailstone sampling and some physical features of the samples is summarized in Table 1. The hailstone samples were immediately stored under refrigeration at approximately −20 °C to avoid loss of material by evaporation, as well as to prevent possible contamination on the hailstone surface by water-steam condensation, or by the absorption of carbon dioxide from the environment. Small ice cores were carefully extracted from the inner section of each hailstone by a drilling process using a clean stainless steel micro-drill. The ice cores were placed in aseptic bottles where the cored samples were finally melted. The liquid samples obtained were stored at ~5 °C and filtered through a filter membrane (0.2 μm) before injection into the CE instrument in order to prevent fouling problems.

3. Results and discussion

3.1. Electrolyte selection and separation of anions

High-mobility electrolytes containing sodium chromate as chromophoric ion, together with indirect UV detection, have been tested successfully for CE analysis of inorganic anions [18,22]. Three electrolytes based on different sodium chromate compositions were recently evaluated for measuring major/trace anions in hydrothermal fluids [8]. From this experimental work, a mixture of 4.7 m*M* sodium chromate, 4.0 m*M* OFM-OH, 10 m*M* CHES and 0.1 m*M* calcium gluconate (pH 9.1) was chosen as the most suitable electrolyte to achieve the complete separation of anions in less than 6 min [8]. Major anions (Cl^-, SO_4^{2-}, HPO_4^{2-}, and HCO_3^-) were measured successfully using hydrostatic injection (10

Table 1
Sampling information and physical features of the hailstone samples collected

Province of Spain	Sample name	Sampling date	Hailstone size, length×width ×height (m)	Hailstone mass (kg)
Tocina (Sevilla)	TO	10 January 2000	0.15×0.15×0.10	>2.0
Alcudia (Valencia)	AL	12 January 2000	0.13×0.08×0.05	0.6
Enguera (Valencia)	ENG	15 January 2000	0.13×0.08×0.06	~1.0
Chilches (Castellón)	CHIL	15 January 2000	0.20×0.26×0.18	~4.0
Algemesí (Valencia)	ALG	17 January 2000	0.12×0.09×0.05	0.8

cm for 30 s) at 25 °C. Trace anions (F^-, Br^-, and NO_3^-) were quantified effectively using electrokinetic injection (4 kV, 10 s) at 15 °C.

With these electrophoretic conditions, the only technical disadvantage was the lengthy analysis time. In the present work, the reduction of the time parameter was therefore defined as an important experimental target to be achieved. From a theoretical point of view, a reduction in the analysis time can be obtained either by increasing the applied voltage, by reducing the capillary length, or by increasing the capillary temperature [23]. In this study, the capillary temperature was examined as a key operating parameter for obtaining a reduction in the analysis time. Capillary temperature has been identified as an effective operating parameter for improving both the speed and the separation efficiency of ions [8,24]. As the capillary temperature is increased, the electrolyte viscosity decreases with a consequent increase in the electroosmotic flow, which finally induces a reduction in the migration times of the analytes [23].

With this expected behavior in mind, the capillary was thermostatted at two temperatures, 25 and 35 °C, which were maintained by a heating device installed in the instrument. Capillary temperatures lower than 25 °C were not considered since these would produce an increase in the migration time. Temperatures greater than 35 °C were not tested because they could result in problems of zone spreading, sample decomposition, or boiling of the electrolyte [23]. As expected, variation of the temperature between 25 and 35 °C produced a significant effect on the time without affecting the anion separation. When the capillary temperature was set at 25 °C, the separation was completed in less than 6 min, the same time reported by Santoyo et al. [8]. An electropherogram obtained with the capillary temperature set at 35 °C is shown in Fig. 1. A significant reduction in the analysis time and good efficiency of separation were obtained. The separation of anions was achieved within 4 min (~3.8 min), a considerably shorter time than that obtained at 25 °C (~6 min). Symmetrical and narrow peak shapes of most anions were observed, except for the hydrogen carbonate (HCO_3^-) asymmetrical peak, which showed a slight tail. This problem is usually attributed to electrostacking, which occurs with hydrostatic injection because the
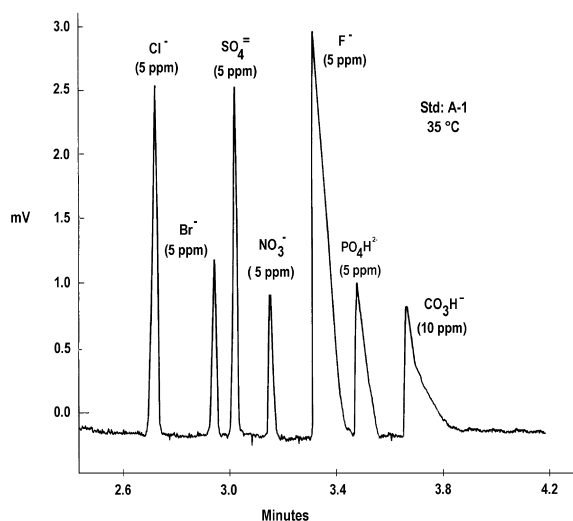

Fig. 1. Electropherogram of a standard mixture of seven inorganic anions. Indirect UV detection at 254 nm with a Hg lamp. BGE; 4.7 mM sodium chromate, 4.0 mM OFM-OH, 10 mM CHES, and 0.1 mM calcium gluconate (pH 9.1). Capillary temperature, 35 °C; separation voltage, −15 kV; hydrostatic injection (10 cm height for 30 s). Other operating conditions are summarized in Sections 2.1 and 3.1.

ionic strength of the sample is lower than that of the electrolyte [18]. All these results suggest that a capillary temperature of 35 °C and the electrophoretic conditions described can be used to obtain a considerable reduction in the analysis time, efficient separation, and reliable quantification of the inorganic anions present in hailstones.

3.2. Repeatability of the CE method, linearity, accuracy, and limits of detection

Eight injections of a standard mixture (A-12) containing Cl^- (0.5 ppm), Br^- (0.5 ppm), SO_4^{2-} (0.5 ppm), NO_3^- (ppm), F^- (0.1 ppm), HPO_4^{2-} (0.5 ppm), and HCO_3^- (15 ppm) were used to evaluate the repeatability using hydrostatic injection at 35 °C. A schematic diagram showing eight overlapped electropherograms obtained during these assays is presented in Fig. 2. The repeatability of migration time, peak area, and peak height was computed and expressed as a percentage of the relative standard deviation (RSD) (Table 2). An average RSD of about 0.64% was obtained for the migration times.
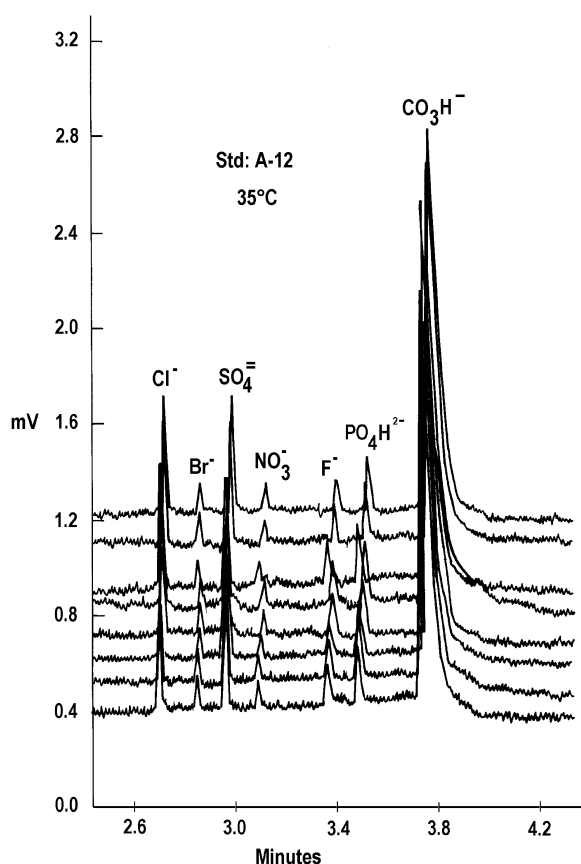
Fig. 2. Schematic diagram showing repeatability results for eight injections of a standard solution of anions (A-12) at 35 °C. The composition of the anion standard is presented in Section 3.2. Superposed electropherograms show all inorganic anions to be well resolved. Operating conditions are summarized in Fig. 1.

Table 2
Repeatability tests based on eight injections of a standard solution of anions: A-12[a]

Anion	LOD (ppm)	RSD (%)		
		Migration time	Peak area	Peak height
Cl^-	0.12	0.52	1.50	0.89
Br^-	0.10	0.69	3.01	1.26
SO_4^{2-}	0.12	0.59	1.74	0.92
NO_3^-	0.12	0.62	3.02	2.27
F^-	0.06	0.65	1.50	2.75
HPO_4^{2-}	0.15	0.72	3.70	1.85
HCO_3^-	0.32	0.70	1.16	1.90

[a] The composition of the standard mixture of anions is presented in Section 3.2.

The repeatability of the peak area ranged from 1.20 to 3.69% for all inorganic anions. Peak height repeatability also varied from 0.89 to 2.75%. The linearity response of the CE method was also evaluated. Five concentrations of each anion with three replicates of each calibration point were used in a weighted linear regression according to the algorithm proposed by Bevington [25]. A linear relationship between peak area and concentration was confirmed for all the anions. Satisfactory linearity results were inferred from both the correlation factors ($r^2 > 0.9$) and the high degree of probability [$P_c(r,n)$] found for these calibration runs (Table 3).

In the absence of an appropriate certified reference material, the accuracy of these analyses was computed from the *y*-intercept of a linear plot, using an external standard with known concentration, and following a well-known statistical procedure very frequently applied in CE [26]. In all linear plots, the origin of the axis was found within the 95% confidence interval, which indicated a reasonable accuracy for the analysis method. The limit of detection (LOD) of each anion, defined as the measurable concentration of a particular ion giving a peak three times as high as the background noise (N), was also estimated (Table 2). Such LODs were calculated by consecutive dilution of a standard mixture. Although the LODs obtained in this work were slightly high, they were quite suitable for our application to most of the hailstone samples.

3.3. Quantitative CE analysis and comparison with the results obtained from WCH methods

Five samples obtained from melted hailstones (TO, AL, ENG, CHIL, and ALG) were analyzed by CE. In order to prevent both overloading effects in the ion zone and co-migration problems, after each sample injection the capillary was washed for 2 min with Milli-Q water and then for 2 min with electrolyte. A typical electropherogram showing the anion separation in a hailstone sample (AL) is presented in Fig. 3. An efficient separation and good detection sensitivity of all anions were obtained. It is important to note that, in some sample electropherograms, e.g. CHIL, trace concentrations of Br^-, NO_3^-, and F^- were poorly detected, and therefore reported as being below their LODs. Such

Table 3
Evaluation of linearity using standard mixtures of anions with five concentrations and three replicates

Anion	Concentration range (ppm)	Linearity	r^2	$P_c(r,n)$
Chloride, Cl^-	5–40	$y = 628.1x + 810.9$	0.996	0.0003
Bromide, Br^-	0.1–0.3[a]	$y = 9523.3x - 96.8$	0.987	0.0017
Sulfate, SO_4^{2-}	1–40	$y = 540.1x - 376.2$	0.997	0.0002
Nitrate, NO_3^-	1–20	$y = 463.5x - 57.8$	0.967	0.0071
Fluoride, F^-	0.1–3	$y = 2007.3x + 100$	0.999	0.00003
Phosphate, HPO_4^{2-}	0.1–5	$y = 1280x - 1470$	0.978	0.0039
Hydrogen carbonate, HCO_3^-	10–80	$y = 421.9x + 191.7$	0.993	0.0007

[a] For the Br^- analyses of the AL sample, the calibration curve ranged from 0.1 to 3.0 ppm.

lower responses could be improved by changing the sample introduction with electrokinetic injection. However, since the amount of sample available was limited, it was not actually possible to optimize the experiment.

All the quantitative analyses were performed using calibration curves for each anion detected. Most of

the analyzed hailstones showed variability in their chemical composition pattern (Table 4). The composition of major anions in most of the samples (Table 4) was typified by moderate concentrations of Cl^-, SO_4^{2-}, and HCO_3^-, intermediate concentrations of NO_3^-, and lower concentrations of Br^- (1.8 ppm or below the LOD), F^- (<0.69 ppm), and HPO_4^{2-}
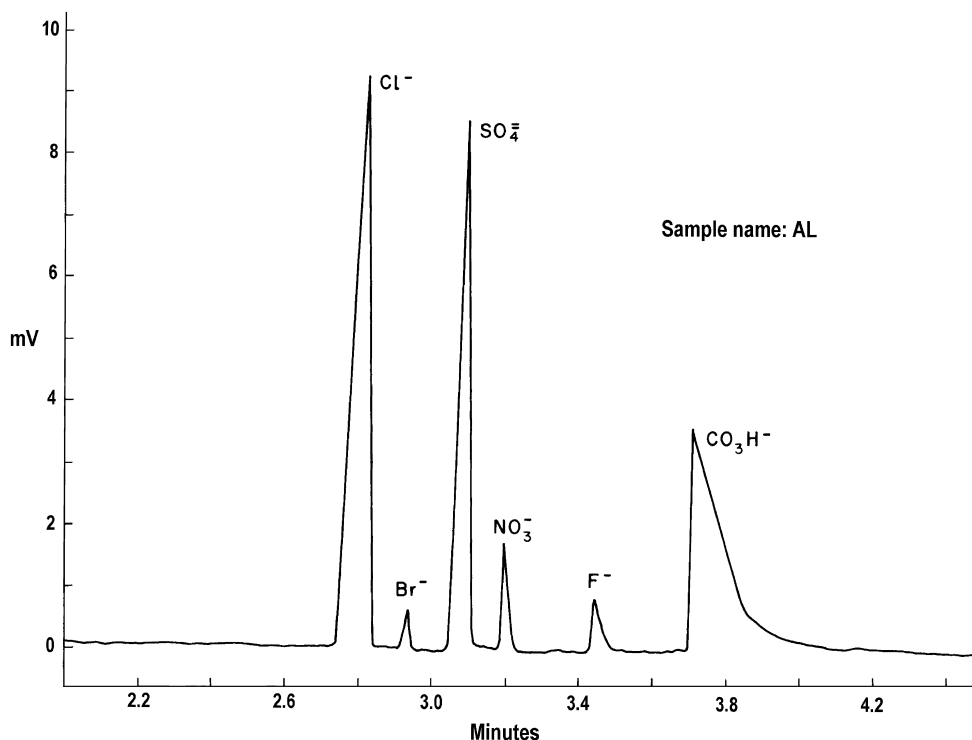

Fig. 3. Electropherogram of hailstone sample AL. Operating electrophoretic conditions are summarized in Fig. 1.

Table 4
Capillary electrophoresis results obtained during the analysis for inorganic anions in hailstone samples collected in provinces of Spain

Anion	Concentration ($\pm\sigma_c$) (ppm)				
	AL	CHIL	ALG	TO	ENG
Cl^-	39.0 (±0.6)	152 (±2)	41.0 (±0.6)	17.0 (±0.2)	15.0 (±0.2)
Br^-	1.8 (±0.1)	<LOD	n.d.	n.d.	n.d.
SO_4^{2-}	30.0 (±0.5)	16.0 (±0.3)	7.0 (±0.1)	18.0 (±0.3)	15.0 (±0.2)
NO_3^-	4.0 (±0.1)	0.60 (±0.02)	0.6 (±0.02)	2.0 (±0.1)	11.0 (±0.3)
F^-	0.69 (±0.01)	<LOD	0.23 (±0.003)	0.59 (±0.01)	<LOD
HPO_4^{2-}	n.d.	39.3 (±0.5)	0.24 (±0.01)	0.20 (±0.01)	12.6 (±0.1)
HCO_3^-	24.1 (±0.3)	39.3 (±0.4)	21.5 (±0.2)	20.0 (±0.2)	81.0 (±0.9)

<LOD, below the limit of detection; n.d., not detected; σ_c, average concentration error.

(<0.24 ppm). The concentration of the major anions was calculated as the mean value from three injections; the average precision errors were less than 1.5, 1.7, and 1.2% for Cl^-, SO_4^{2-}, and HCO_3^-, respectively.

To verify the chemical compositions quantified by CE, analyses of the hailstones for some anions (Cl^-, SO_4^{2-}, and NO_3^-) using traditional WCH methods were also performed. For the WCH methods, the molecular UV–Vis spectrophotometry technique (Alliance analyzer with continuous flow) was used. SO_4^{2-} was measured by a precipitation reaction with barium chloride and blue methylthymol. The Cl^- content was quantified after reaction with mercury thiocyanate in the presence of ferrous ion. NO_3^- was determined by chemical reduction to nitrite with a Cu–Cd reducer column. The analytical errors of the WCH methods for measuring SO_4^{2-}, Cl^-, and NO_3^- were generally ±2, ±1, and $\pm1\%$, respectively. These analytical results were compared with the concentrations reported by CE. Confidence limits or error bands of 10 and 20% were used to evaluate the existence of significant differences among the comparisons. Most of the results obtained by CE were consistent with those of WCH, except for all the anion concentrations corresponding to the CHIL sample, which exceeded the method difference of 20%. A possible explanation for this significant difference could be due to either: (i) a sampling error during the extraction of the core from the hailstone specimen; (ii) the analysis of different strata of the core sampled, which was shown to have a notable heterogeneity from the isotopic point of view [3]; or (iii) matrix interference of the sample, combined with the poor sensitivity detection of the WCH method.

4. Conclusions

A new application of CE for determining inorganic anions in atmospheric hailstone samples has been demonstrated. An electrolyte consisting of 4.7 m*M* sodium chromate, 4.0 m*M* OFM-OH, 10 m*M* CHES, and 0.1 m*M* calcium gluconate (pH 9.1) was found to be suitable for quantifying all the anions present in the samples. A significant reduction in the analysis time was obtained when the capillary temperature was set at 35 °C. Under such electrophoretic conditions, the separation of all anions was completed efficiently in less than 4 min. The experimental application performed in this work not only confirms the sensitivity and selectivity capabilities of the CE analytical technique for analyzing inorganic anions, but also makes an original contribution to the environmental sciences by providing a new reliable analytical tool to be used in future investigations.

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