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Capillary ion electrophoresis, an environmental method for the determination of anions in water

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ABSTRACT

Capillary ion electrophoresis has recently been introduced as a new separations technique for the analysis of inorganic anions. Among its many attributes are rapid, highly efficient separations with different selectivities (compared to ion chromatography), simplicity, and economy.

This paper demonstrates the ability of capillary ion electrophoresis to analyze primary and secondary anionic contaminants as well as other ions of environmental concern in drinking water, groundwater, and wastewater. Analysis time is less than five minutes. A comparison of the data to ion chromatography shows excellent correlation.

INTRODUCTION

The United States Environmental Protection Agency (EPA) has established analytical methods for inorganic contaminants using ion chromatography (IC) [1]. However, there is an emerging technology that offers considerable advantages. Capillary ion electrophoresis (CIE) (Waters' trade name: Capillary Ion Analysis, CIA) offers significant improvement over IC in efficiency and analysis time. The unique selectivity of CIE provides an alternative solution to coelution problems that occur with IC in real samples [2].

CIE has recently been introduced as a new separation technique for the analysis of inorganic and organic ions [3]. CIE is a branch of capillary electrophoresis (CE) which is optimized for the rapid analysis of low-molecular-mass anions and cations. It separates ions according to their mobility in electrolytic solutions [4–7]. The technique has been successfully applied to the analysis of a variety of anionic solutes in several complex sample matrices [8,9].

CE instrumentation has few moving parts and uses a low cost, easily replaceable open tubular capillary instead of packed chromatography column. No equilibration time is required for the capillary. Simply fill the capillary with electrolyte and run. Sample preparation is minimal because there is no chromatography column to be protected from extraneous materials in the sample. Analyses are completed in less than five minutes.

The scale of electrolyte consumption is at least an order of magnitude smaller than liquid or ion chromatography eluent consumption. High sensitivity is achieved while analyzing only nanoliters of sample, using only a few microliters of electrolyte in the capillary. This ability to analyze complex samples without producing any significant volume of additional waste has caught the attention of analysts who are becoming involved in the mixed waste program.

In this paper a test method using CIE has been developed for the analysis of anions in aqueous matrices. The test method was applied to samples of drinking water, groundwater, and wastewater for the analysis of bromide, chloride, fluoride, nitrate, nitrite, phosphate, and sulfate. The results obtained are compared to data from the same samples analyzed by IC.

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EXPERIMENTAL

Instrumentation

The capillary electrophoresis system employed was the Waters (Waters Chromatography Division of Millipore, Milford, MA, USA) Quanta 4000. The configuration for performing anion analysis included a negative power supply (0–30 kV), a mercury lamp for indirect UV detection at 254 nm, and a Waters Accu-Sep polyimide-coated fused-silica capillary (60 cm × 75 μm I.D.). A hydrostatic sample injection mode (10 cm for 30 s) was used in this work. Data acquisition was performed with a Waters 860 data station. Detector time constant was set at 0.1 s and data acquisition rate was 20 points/s.

The ion chromatograph used in this study was a Waters Action Analyzer with a 700 WISP Satelite auto sampler and a Model 431 conductivity detector. The Waters 860 data station was also employed for data acquisition. The data acquisition rate was 1 point/s. The analytical column used was a Waters IC-Pak Anion HC (150 × 4.6 mm I.D.) methacrylate-based anion exchanger.

Reagents

Water (18 MΩ) (Millipore, Bedford, MA, USA) was used to prepare all solutions. The chromate electrolyte was prepared from a concentrate containing 100 m*M* sodium chromate tetrahydrate (Malinkrodt, Paris, KY, USA; analytical-reagent grade) and 0.0056 m*M* sulfuric acid (J. T. Baker, Phillipsburg, NJ, USA; Ultrex grade) Electroosmotic flow (EOF) modifer for reversal of the direction of EOF was a 20 m*M* concentrate (CIA-Pak OFM anion BT) obtained from Waters. The working electrolyte consisted of 4 m*M* chromate–0.3 m*M* OFM anion BT, pH 8.1. CIA methods are covered under U.S. Patents: 5 104 506 and 5 128 055.

When using the Waters MilliTrap H^+ membrane cartridge the modified borate–gluconate eluent is recommended [10]. The eluent was prepared from analytical-grade chemicals, boric acid, gluconic acid, and lithium hydroxide monohydrate obtained from Aldrich, Milwaukee, WI, USA. Glycerin and HPLC-grade acetonitrile were of obtained from J. T. Baker. Eluents and working electrolytes were prepared fresh daily, filtered and degassed with a Millipore solvent clarification kit prior to use.

The standard mixtures were prepared by diluting 1000 ppm stock solutions containing a single anion. Particulates from samples were removed with disposable prerinsed Millex-GV 0.22 μm filters (Millipore). The samples were diluted with water or run neat.

RESULTS AND DISCUSSION

Anion analysis

In CIE, a free zone separation occurs as the ions migrate according to their mobility in the carrier electrolyte when an electric field is applied through the capillary. The electrolyte co-ion is selected to have a different UV absorbance but similar mobility to the analytes of interest to permit differential UV detection with optimal peak symmetry. Detection sensitivity can be maximized by selecting a background ion that has both maximum molar absorptivity and matching ionic mobility [6]. For anion analysis an electroosmotic flow modifier (OFM) is added to the electrolyte to ensure the bulk flow of the electrolyte, electroosmotic flow (EOF), is directed towards the detector. Anions move in the same direction as the EOF towards the anode or co-EOF. This increases the net velocity of the anions through the capillary and creates a fast separation. Water from the sample has no net charge and therefore migrates in the same direction at the rate of the EOF. Note that cations such as those found in groundwater samples (sodium, potassium, calcium, and magnesium) migrate in the opposite direction (counter-EOF) towards the cathode.

Electropherogram is the proper name to describe the chart generated by CIE although the separation does resemble a chromatogram. The analytes each have a migration time which is the time it takes the analyte to migrate from the point of injection to the detector portion of the capillary. Identification and quantitation is performed by an external standard. Fig. 1 shows the separation of a seven-anion standard by CIE consisting of inorganic anions commonly monitored in water. The concentration of each anion in the standard is in the low ppm range. Due to the different separation mechanism of CIE, bromide is the first to migrate followed by chloride, sulfate, nitrite, nitrate, fluoride, and phosphate. The analysis time is under 5 min.

Peak shape is optimized by closely matching the mobility of the carrier electrolyte ions with that of

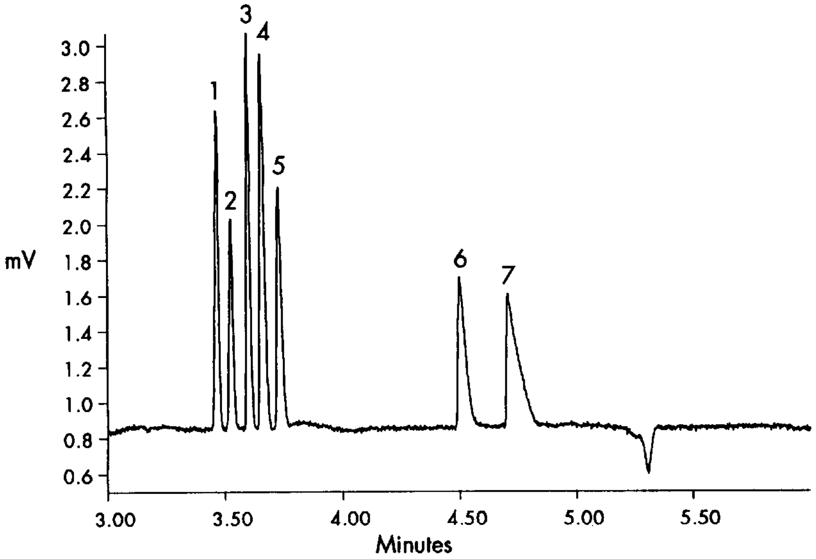


Fig. 1. Electropherogram of seven-anion standard. CIE test method conditions: Fused-silica 60 cm × 75 μm I.D. capillary; voltage 15 kV (negative); 4 m*M* chromate–0.3 m*M* CIA-Pak OFM Anion BT (patented) at pH 8.1; indirect UV detection at 254 nm; hydrostatic injection (30 s at 10 cm). Peaks: 1 = bromide (4 ppm); 2 = chloride (2 ppm); 3 = sulfate; (4 ppm); 4 = nitrite (4 ppm); 5 = nitrate (4 ppm); 6 = fluoride (1 ppm); 7 = phosphate (4 ppm).

the analyte ions. When analyte anions have lower mobility than the electrolyte co-ion , the peaks will tail; when the analyte ion mobility is higher, the peaks will front [11]. For any set of ionic mobilities, peak asymmetry increases with increasing concentration of the analyte ion in the migrating zone [12].

Selectivity is based on the charge and size of the ion. The greater the charge to mass ratio (more conductive) the faster the analyte migrates. The inherent speed of CIE enables high peak capacity in a very short time frame with separation efficiencies reaching over 500 000 theoretical plates. Electrolyte parameters that can change or optimize selectivity include pH, ionic strength, EOF modifiers, complexing agents, and organic solvents.

Drinking water analysis

The CIE test method was first successfully applied to drinking water Fig. 2 shows the electrophoretic separations of two drinking water samples.The upper electropherogram contains a separation of chloride, sulfate, nitrate, and carbonate of a sample taken from a well. The first three components are well resolved for easy identification and quantitation. Carbonate, a weak acid, exhibits low mobility at the electrolyte pH 8.1 and elutes well after the strong acid anions. At pH 8.1, carbonate exists as bicarbonate (HCO_3^-) and exhibits a gradually increasing mobility (shorter migration time) as the pH of the electrolyte increases. As it converts to its dibasic form (CO_3^{2-}) at pH 12 it migrates just after nitrate [4].

The peak shape of carbonate can be attributed to the electrostacking condition [6,13]. According to the electrostacking condition, the samples zones in hydrostatic injections must have lower ionic strength than the carrier electrolyte. The net effect is an accumulation of sample ions inside a very narrow zone at the sample-carrier electrolyte boundary. This electrostacking occurs before the migration of the analyte ion zone through the bulk of the carrier electrolyte solution. The result is narrow, symmetrical peaks. The excessive amount of carbonate contained in the water samples creates the adverse effect; a broad, asymmetrical peak. A simple solution is to dilute the sample in water (18 MΩ).

The lower electropherogram in Fig. 2 displays a drinking water sample obtained from a household

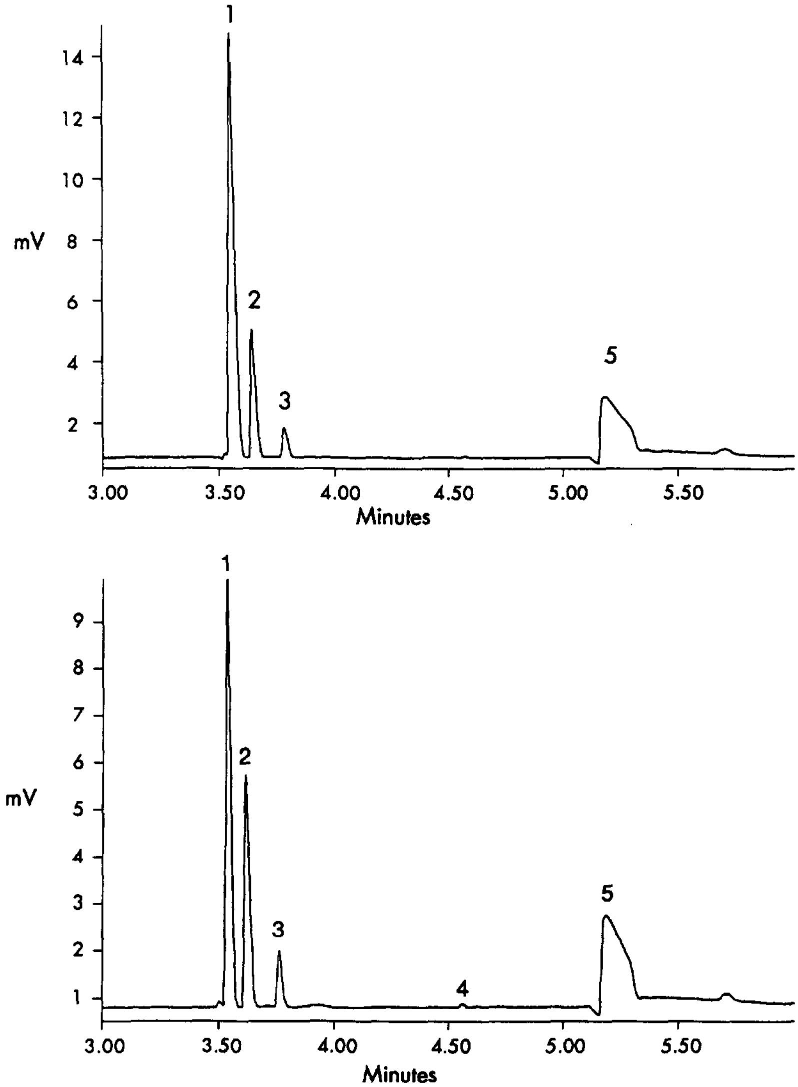


Fig. 2. Analysis of anions in drinking water. Same CIE test method conditions as in Fig. 1. Upper electropherogram: Wellwater neat sample. Peaks: 1 = chloride (36.5 ppm); 2 = sulfate (11.4 ppm); 3 = nitrate (3.2 ppm); 5 = carbonate (not quantitated). Lower electropherogram: Tapwater neat sample. Peaks: 1 = chloride (20.0 ppm); 2 = sulfate (14.1 ppm); 3 = nitrate (3.5 ppm); 4 = fluoride (0.06 ppm); 5 = carbonate (not quantitated).

tap. Four components of interest were found along with ubiquitous carbonate. Note the high sensitivity for fluoride and its migration time. Using an IC anion-exchange column, early eluting fluoride is found close to the void volume that contains cations, neutral species and the classic water dip. This can make detection and quantitation of fluoride difficult. With CIE, fluoride migrates in the latter portion of the electropherogram well resolved from any other components. High sensitivity for fluoride is achieved due to the "lightness" of the ion. Principles of differential UV detection revolve around displacement of the electrolyte co-ion in this case chromate, when the analyte ions are UV transparent. It

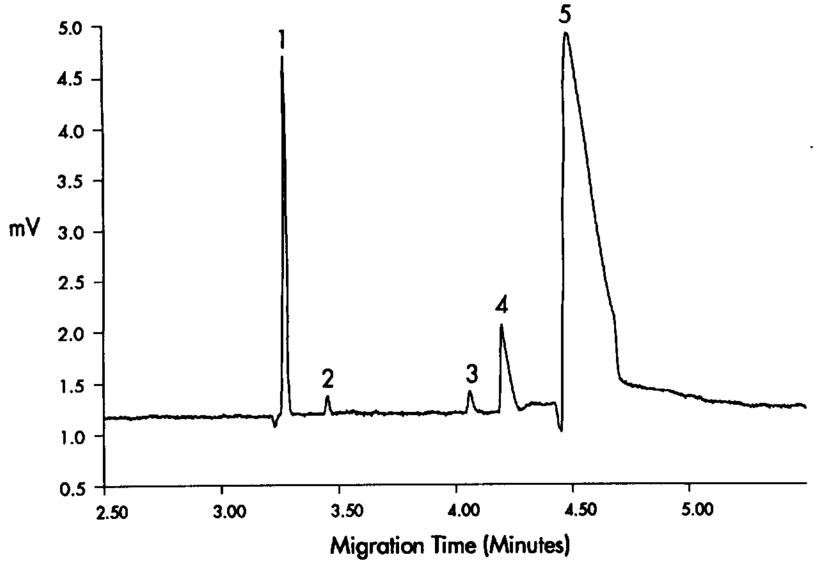


Fig. 3. Electropherogram of EPA groundwater sample No. 1. Same CIE test method conditions. The sample was diluted 1:10 with water. Original sample concentrations reported. Peaks: 1 = chloride (50.3 ppm); 2 = nitrate (4.4 ppm); 3 = fluoride (1.8 ppm); 4 = phosphate (26.1 ppm); 5 = carbonate (not quantitated).

takes two monovalent fluoride (F$^-$) ions to displace one divalent chromate (CrO$_4^{2-}$) ion. Since fluoride is a "light" ion there are more molecules per unit of concentration placed into the capillary. For example using the atomic masses of fluoride (18.9) and chloride (35.5), 1 ppm of fluoride = 0.05 mequiv./l and 1 ppm of chloride = 0.03 mequiv./l. Therefore mass to mass, fluoride generates a greater response than chloride.

Groundwater analysis

The characteristics of CIE are extremely advantageous for the analysis of anions in groundwater. An example of the power of this technique is demonstrated by the analysis of groundwater samples taken by the EPA from a hazardous waste site. Figs. 3–5 show electropherograms of groundwater samples taken from different locations at the site.

EPA groundwater sample No. 1 (Fig. 3) displays the separation of four anions of interest, chloride, nitrate, fluoride, and phosphate, in the presence of an excessive level of carbonate. Due to the unique selectivity of CIE, the large amount of carbonate does not interfere with the analysis. Cations contained in the sample do not participate in the separation. The water peak is not seen in the separation because it does not reach the detector until 8 min. These features make identification and quantitation of anions in groundwater easy and simple. Sample preparation simply involved diluting the sample in deionized water and filtration to remove particules.

The electropherogram of EPA groundwater sample No. 2 (Fig. 4) shows an expanded view of a separation of anions in a high carbonate sample matrix. Three components of interest, chloride, sulfate, and fluoride, were identified and quantitated. By taking a closer look at the electropherogram, a fluoride peak can easily be distinguished. It is an example of the high sensitivity of CIE in real samples. The solution concentration of fluoride detected is 60 ppb based on a 1:10 dilution of the sample. This amount of fluoride would be impossible to detect in the analysis of this groundwater sample by IC. It is a classic example of coelution problems that exist under normal isocratic IC conditions as described earlier.

Fig. 5 demonstrates the ability of CIE to detect anions of interest in EPA groundwater sample No. 3 in the presence of high sulfate. The sample was

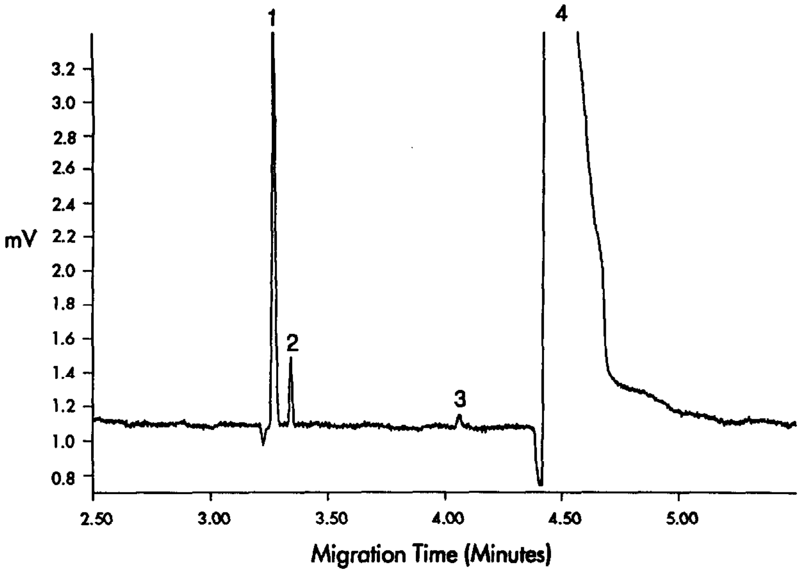
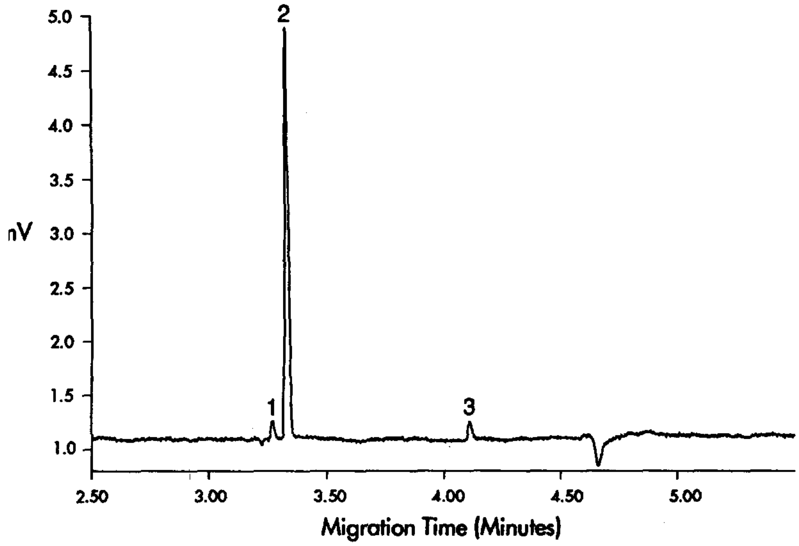


Fig. 4. Expanded view of the electropherogram of EPA groundwater sample No. 2. Same CIE test method conditions. The sample was iltued 1:10, with water. Original sample concentrations reported. Peaks: 1 = chloride (32.8 ppm); 2 = sulfate (6.1 ppm); 3 = fluoride 0.6 ppm); 4 = carbonate (not quantitated).

iluted 1:1000 to maintain optimum peak shape. A mall amount of chloride is separated and detected rom a large amount of sulfate. Phosphate was also dentified in the sample. The solution concentrations displayed in the electropherogram for chloride and phosphate are 271 ppb and 330 ppb, respectively.



ig. 5. Electropherogram of EPA groundwater sample No. 3. Same CIE test method conditions. The sample was diluted 1:1000 with ater. Original sample concentrations reported. Peaks: 1 = chloride (271.0 ppm); 2 = sulfate (8165.8 ppm); 3 = phosphate (330.3 pm).

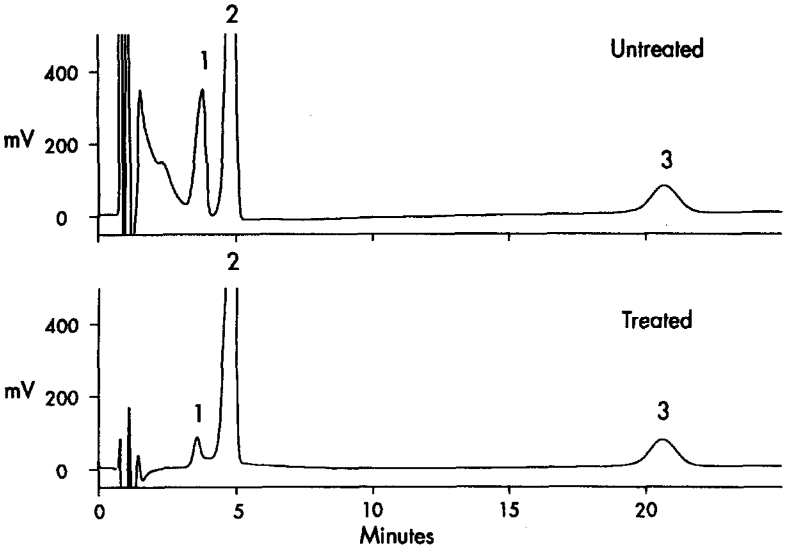


Fig. 6. Anion analysis of industrial wastewater by ion chromatography using Waters Method A-1000. Conditions, column: Waters IC-Pak Anion HC, eluent: modified borate–gluconate, flow-rate: 2.0 ml/min, detection: conductivity. Upper chromatogram; untreated wastewater sample. Lower chromatogram; same wastewater sample after treatment with Waters Millitrap H^+ membrane cartridge. Peaks: 1 = carbonate (not quantitated); 2 = chloride (83.2 ppm); 3 = sulfate (23.8 ppm).

Wastewater analysis

The selectivity for anion separations using CIE differs significantly from that obtained using conventional anion-exchange columns in IC, where retention is largely a function of valence state or affinity to the anion-exchange sites. In IC, fluoride elutes first followed by the monovalents chloride, nitrite, bromide and nitrate, then the divalents hydrogenphosphate and sulfate. Analysis time typically ranges from 10 to 20 min. An example of the selectivity of IC is demonstrated by the analysis of an industrial wastewater sample using Waters Method A-1000: conductivity detection of anions using single column ion chromatography [10]. The upper chromatogram in Fig. 6 contains the separation of the untreated wastewater sample. With an anion-exchange separation; cations, neutral organic species, and water; all elute at the void volume of the column. Short-chained monocarboxylic acids and weakly retained anions, such as fluoride and methanesulfonate, are all eluted early and tend to be poorly resolved in many instances. The presence of elevated carbonate and/or high levels of sample cations may further complicate the early portion of the chromatogram. The use of gradient or coupled IC systems can often overcome these resolution problems. However, both of these approaches are more complex than simple isocratic methods.

Another possible solution is sample pre-treatment, that is removal of interferences before analysis. The lower chromatogram in Fig. 6 represents the separation of the same wastewater sample after passing it through a Millitrap H^+ membrane cartridge. The sample preparation device removes cations, reduces the amount of carbonate and neutralizes high pH samples. This "cleans up" the early portion of the chromatogram allowing better detection and quantitation of the anions of interest. In this sample chloride and sulfate were identified.

Anion analysis of wastewater using CIE exhibits several significant advantages. First, the cations migrate in the opposite direction so they do not interfere in the separation. Second, the strong acid anions (inorganics) *i.e.*, bromide, chloride, sulfate, nitrite, nitrate, fluoride, and phosphate, have high charge-to-ionic radii ratios so they are the first to migrate through the capillary. Third, organic acids commonly found in wastewater such as formate, acetate, and propionate migrate later and are significanly removed form their migration time band. Since their pK_a values range from 3 to 5, they are fully ionized at pH 8.1. However, the organic acids have a smaller charge to mass ratio (less conductive) and therefore lower mobility. Fourth, carbonate also a weak acid, migrates among the short chain carboxylic acids. Fifth, neutral components like water, are carried along by the EOF and have appreciably longer migration times. Since the capillary is an open tube, the analysis is complete when the last peak of interest passes through the detector. The remaining slower migrating anions and neutral species can be simply purged from the capillary with an automated vacuum purge once the desired separation is obtained. This also refills the capillary with fresh electrolyte for the next sample. Thus analysis times are typically less than five minutes.

These attributes are confirmed from the analysis of the same "untreated" industrial wastewater sample by CIE. Fig. 7 contains an expanded view of the electrophoretic separation identifying chloride and sulfate at the same concentrations determined by IC in Fig. 6. Taking a closer look at the electropherogram, a fluoride peak is easily detected and quantitated. Fluoride was not detected in the wastewater sample by IC even after sample pre-treatment with a Millitrap membrane cartridge.

Analysis of a power plant wastewater effluent by CIE is described in Fig. 8. Chloride, sulfate, and nitrate were detected in the presence of carbonate. Based on a 1:10 dilution of the sample the nitrate peak represents a solution concentration of 220 ppb.

Under the electrophoretic conditions described in his study, the capillary electrophoresis system displays linearity over three orders of magnitude. The reproducibility of the system is excellent. The % relative standard deviation (R.S.D.) of migration times from 15 consecutive hydrostatic injections of a seven-anion standard is less than 0.5%. The R.S.D. of peak areas from 4 consecutive hydrostatic injections of a seven-anion standard is less than 1.4%.

Comparison of the data

Four water samples were analyzed by both IC and CIE. The results are summarized in Table I. Columns 2 and 3 contain the original concentrations in ppm of each anion detected in the samples by IC and CIE, respectively. The concentrations for

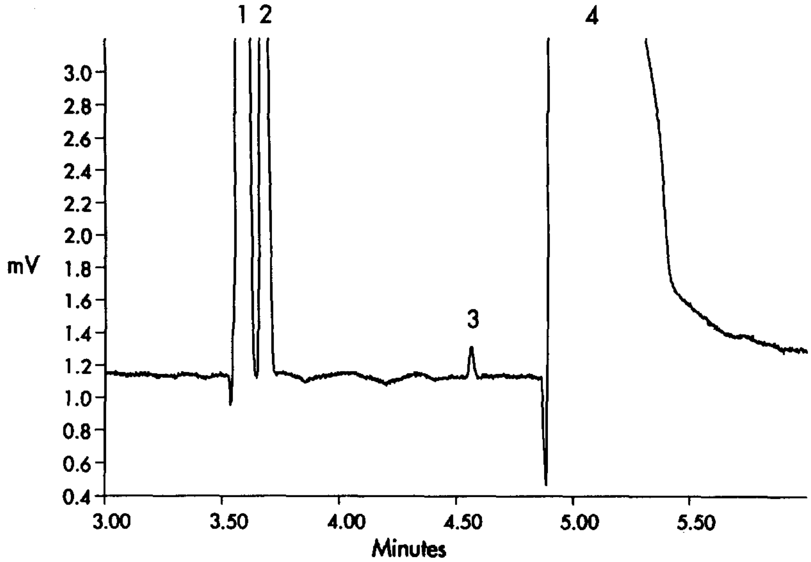


Fig. 7. Anion analysis of industrial wastewater by CIE. Same CIE test method conditions. Electropherogram of the same untreated wastewater sample described in Fig. 6. Peaks: 1 = chloride (83.0 ppm); 2 = sulfate (23.1 ppm); 3 = fluoride (0.13 ppm); 4 = carbonate (not quantitated).

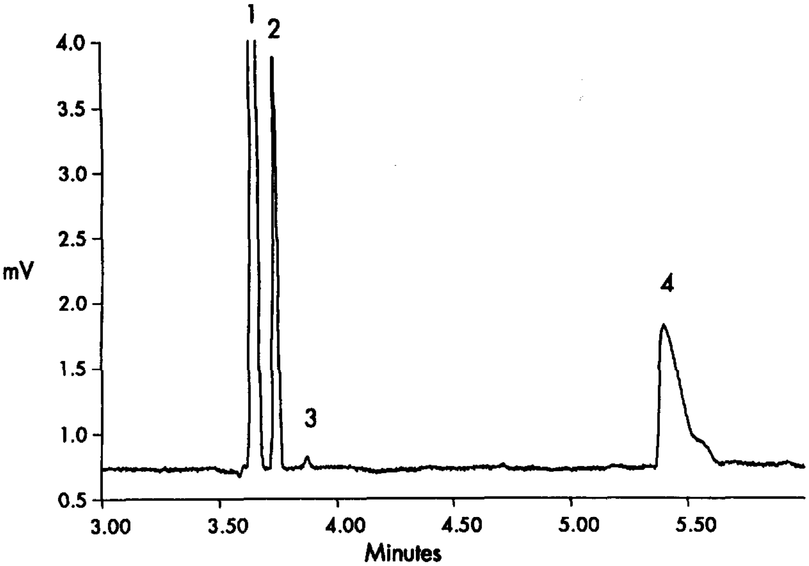


Fig. 8. Electropherogram of power plant wastewater effluent. Same CIE test method conditions. Sample was diluted 1:10 with water. Original sample concentrations reported. Peaks: 1 = chloride (199.8 ppm); 2 = sulfate (76.8 ppm); 3 = nitrate (2.2 ppm); 4 = carbonate (not quantitated).

chloride, sulfate, and nitrate determined by both analytical techniques show excellent correlation. Column 4 displays a simple ratio calculated by taking the CIE concentration in column 2 and dividing it by the corresponding IC concentration in column 2. A value of 1.000 is the ideal number. The actual ratios ranged from 0.937 to 1.041. Note that fluoride detected in two of the samples by CIE was not detected by the IC method.

TABLE I

DATA COMPARISON OF 4 WATER SAMPLES

	IC (ppm)	CIE (ppm)	CIE/IC
Tapwater			
Chloride	20.222	20.035	0.991
Sulfate	14.772	14.044	0.951
Nitrate	3.551	3.531	0.994
Fluoride	Not detected	0.062	–
Wellwater			
Chloride	37.650	36.476	0.969
Sulfate	11.951	11.432	0.956
Nitrate	3.165	3.179	1.004
Industrial wastewater			
Chloride	83.148	83.025	0.998
Sulfate	23.831	23.065	0.967
Fluoride	Not detected	0.132	–
Power plant wastewater			
Chloride	191.834	199.768	1.041
Sulfate	79.882	76.748	0.961
Nitrate	2.384	2.231	0.937

CONCLUSIONS

Capillary ion electrophoresis is a powerful separation technique which offers many advantages for the analysis of inorganic and organic acid anions in aqueous matrices. Rapid, highly efficient separations with different selectivities compared to ion chromatography are obtained. The matrix independent separation requires minimal sample preparation. Only nanoliters of sample volume and small amounts of electrolyte are needed to perform the analysis. This low reagent consumption minimizes the waste that is produced. The instrumentation is simple with low maintenance and very economical to operate. More important the capillary is not a chemical product and is a fraction of the cost of an IC column.

Due to these attributes, this new environmental method has been submitted to the ASTM committee D-19 on Water and subcommittee D19.05 on Inorganics in Water for review. Further investigation of CIE for environmental analysis is proceeding.

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