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Determination of analytes at extreme concentration ratios by gradient ion chromatography with solid-phase reaction detection

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ABSTRACT

Conditions for separating anions by gradient ion chromatography are explored where detection is performed with the post-column addition of a solid-phase reagent (SPR). Gradient ion chromatography can be useful when analyzing samples containing trace analytes in the presence of large amounts of a particular anion. However, loss of peak height and area of early eluting anions can occur when there is an extremely high concentration of a second later eluting anion. The distortion appears to result from a moving zone overloading of the separation column. Only early eluting anions are affected. The concentration dependence of the excess anion is studied and various approaches to mitigate the peak distortion are discussed.

INTRODUCTION

Gradient elution ion chromatography is still a new field. Some early limited gradient work was performed [1–4], but the first modern, practical gradient ion chromatography work using membrane suppression and conductivity detection was published by Rocklin and co-workers in 1987 [5].

Gjerde and Benson have recently introduced a new method for conductivity detection in ion chromatography. The method is called suspension detection method or solid-phase reagent (SPR) detection method [6,7]. A colloidal suspension of SPR is added to the eluent stream prior to conductivity detection to produce a low background eluent signal (see experimental section). Jandik and co-workers [8,9] adapted the SPR detection method for gradient ion chromatography. Their work showed that the method works well with a variety of eluents for ion-exchange and ion-interaction chromatography. These include eluents based on borate, hydroxide, and carbonate counter ions.

In this work, gradient ion chromatography and SPR detection is used to exam-

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ine the determination of minor amounts of analytes in the presence of excess concentration of anions. Conditions are described needed to adjust peak windows for the IC-Pak Anion column. Large amounts of chloride in the sample may cause overloading of the column for early eluting peaks. Although the chromatogram may look normal, the overloading can cause a reduction of certain sample peak heights from what they would be without the presence of extreme analyte concentrations.

EXPERIMENTAL

Chemicals for the preparation of standard solutions and eluents were laboratory grade as obtained from a range of suppliers. All chromatographic mobile phases were filtered with a Millipore Solvent Purification Kit prior to use. Only 18 MΩ water obtained from a Milli-Q Laboratory Water System (Millipore, U.S.A.) was utilized in the experiments.

Two versions of polymethacrylate-based anion-exchange columns were used. IC Pak A Anion column (50 × 4.6 mm I.D.) contains a 10-μm packing material. The second column, IC Pak A HR Anion, has the dimensions of 75 × 4.6 mm I.D. and contains 6 μm diameter material. The exchange capacity for both of these columns is 30 μequiv./g. Sarasep AN1, a polystyrene-based anion exchanger, was used for some experiments. This column has dimensions of 250 × 4.6 mm I.D.

The SPR is manufactured by Sarasep, and distributed exclusively by Waters (patents pending). The SPR is obtained as a concentrate (Part No 36546) containing about 12% solids with a cation-exchange capacity not less than 500 milliequivalents per liter of suspension. The SPR concentrate was diluted about tenfold with deionized water prior to use. The diluted suspension did not show any appreciable sedimentation as determined in a 100-ml graduated cylinder filled with 100 ml of SPR over a period of time lasting 8 hours. The conductivity of the SPR suspension (1.2%) did not exceed 10 μS/cm. To eliminate any effects of contaminants introduced by handling or during prolonged storage of SPR suspensions, the manufacturer recommends use of an inline polishing column (SPR-H^+ Polishing column kit, Waters, U.S.A., Part No. 36547.) The column contains a high-capactiy anion exchanger in the OH-form. The polishing column was regenerated every 50 hours with 10 ml of 100 m*M* KOH followed by a rinsing with a least 15 ml of deionized water.

Basic eluents such as sodium carbonate are highly conductive. SPR reacts with the eluent stream to produce low-conducting carbonic acid. At the same time, Na^+ or any other eluent counter ion is taken up by the SPR. Also, sample anions are converted to high-conducting strong acids.

A Water M600 MS and a Waters Action Analyzer IC were used in the work. The pump flow-rate was set at 1 ml/min for all chromatograms. All injections were 100 μl. The eluents were prepared using degassed water and degassed during use by continuous sparging with He. This treatment prevented CO_2 from being absorbed by the basic eluents. Carbonate absorbed in the eluent may contribute to gradient 'ghost' peaks.

Fig. 1 gives details of the instrumental configuration for post-column addition of SPR suspensions. The flow-rate of SPR (0.4 ml/min) is determined by adjustments of pressure from the air supply (12). The reagent flows from the pressurized PTFE vessel (13) through the SPR polishing column (10) and through a check valve (8) to a

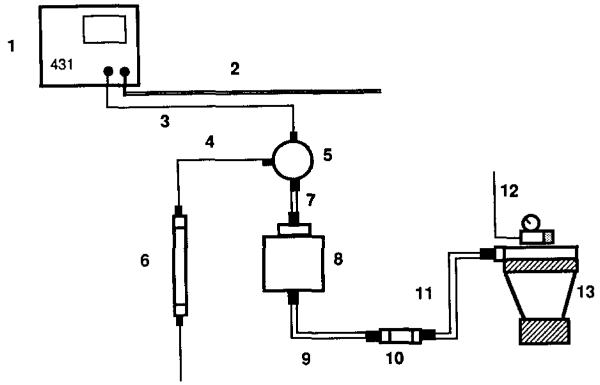

Fig. 1. Detailed diagram of hardware configuration for post-column addition of SPR. 1 = Conductivity detector: Waters 431 detector, four electrode cell design; 2 = waste line: 4 × 0.009 in. stainless connected to 431 + 24 × 1/16 × 0.060 in. PTFE tubing; 3 = tee to 431: 15 × 1/16 × 0.010 in. PTFE to 431 inlet; 4 = column to tee: shortest 1/16 × 0.010 in. PTFE from column to tee; 5 = tee: Unmount tee from check valve block for shortest path length; 6 = analytical column: Waters IC PAK A or IC PAK A HR; 7 = check valve to tee: 2 × 1/8 in. O.D. PTFE; 8 = check valve; 9 = polisher column to check valve; 3 × 1/8 in. O.D. PTFE; 10 = polisher column: 8 × 25 mm containing AG1 × 8, 200 mesh; 11 = reservoir to polisher column: 12 × 1/8 in. O.D. PTFE; 12 = air supply: minimum of 90 p.s.i. compressed air supply; 13 = reservoir for SPR: reconfigure with outlet on left side.

mixing tee (5), where it blends into the eluate from the chromatographic column (6). The check valve prevents any accidental addition of chromatographic phase to the SPR supply inside the vessel. The instrumentation shown in Fig. 2 should be used with SPR within the prescribed range of 0.5–1.5% solids suspension concentration. During more than a 12-month period of evaluation, the authors did not experience any sedimentation or plugging problems.

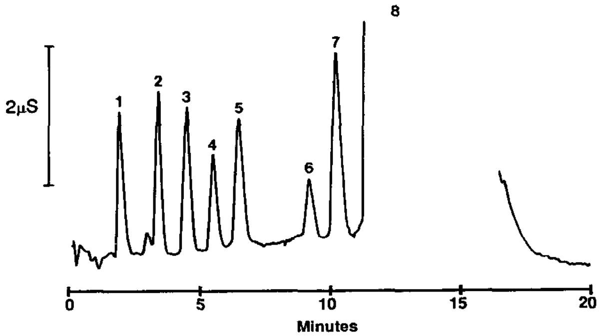

Fig. 2. Separation of traces of inorganic anions in a sample matrix containing high levels of chromate. Column: IC Pak Anion; Eluent: 1 ml/min. A = water, B = 100 mM boric acid adjusted to pH 8.3 with sodium hydroxide. Gradient: 0 to 5 min, 25 to 50% B, and then hold 50% B 5 to 20 min. Peaks: 1 = fluoride (0.5 ppm); 2 = chloride (1 ppm); 3 = nitrite (2 ppm); 4 = bromide (2 ppm); 5 = nitrate (2 ppm); 6 = phosphate (3 ppm); 7 = sulfate (2 ppm); 8 = chromate (500 ppm).

RESULTS AND DISCUSSION

Gradient ion chromatography can be used to analyze samples containing extreme ratios of analyte concentrations. In these samples, gradients may be used to modify the spacing between the peaks in the critical regions of the chromatogram. Generally, it is possible to design separations with maximized peak to peak resolution in those regions where the highly concentrated analytes are expected to elute. In some instances, in order to reduce the total run time, it may be desirable to compress the remaining regions of a chromatogram after the separation of the critical peaks has been achieved.

Equations have been derived that describe the effect of different gradient profiles on sample anion retention [5]. However, there are no set rules for developing the gradients needed in ion chromatography. The gradient used, depends on the column and the desired sample resolution. Method development for gradient ion chromatography is still largely a trial and error method. Dilute monovalent eluents are used to resolve early eluting monovalent sample species. Then, the eluent concentration is increased to elute strongly retained anions. A steeper eluent concentration increase will shift multivalent sample species to shorter retention times faster than monovalent sample species. The eluent pH shift with gradient can also be an important factor in gradient separations. For example a pH change in the gradient can change phosphate from a divalent to a trivalent anion resulting in longer retention.

Samples containing high levels of chromate are common in the electroplating industry as well as in environmental contracting laboratories. Fig. 2 shows an example of a gradient separation of a sample with concentration ratios of the analyte peaks ranging from 1:1000 to 1:166. All peaks between fluoride and sulfate are well resolved. The peak due to an elevated level of chromate elutes within a reasonable time. Under otherwise identical conditions, an isocratic separation require an analysis time in excess of 60 minutes to accomplish a similar resolution of the seven inorganic anions in the presence of the high chromate concentrations. The unidentified peak eluting shortly before chloride is due to carbonate which entered the sample by absorption from the atmosphere.

The chromatogram in Fig. 3 shows the separation of trace amounts of anions in the presence of high levels of phosphate. The phosphate was added to the standard

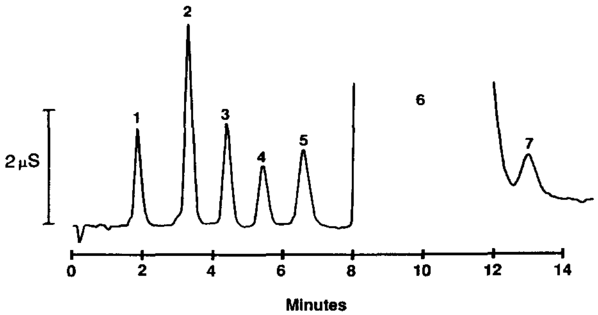

Fig. 3. Chromatographic analysis of low concentrations of anions in the presence of high concentration of phosphate. Conditions are the same as in Fig. 2. Gradient: 0 to 7 min, 50% B, 7 to 20 min 100% B. Peaks 1–7 and concentration as in Fig. 2. except for phosphate (peak 6) which is 500 ppm.

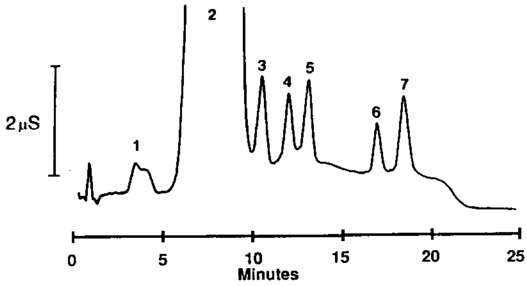

Fig. 4. Separation of low levels of anions in the presence of high concentration of chloride. Column: IC Pak Anion; Eluent: 1 ml/min. A = water, B = 50 m*M* boric acid, pH 8.3. Gradient: 0 to 8 min, 20% B; 8 to 12.5 min, 20 to 100% B; 12.5 ot 20 min, 100% B. Peaks 1–7 and concentrations as in Fig. 2, except for chloride (peak 2) which is 500 ppm.

mixture as dibasic anion. An advantage of the borate mobile phase is its relatively high buffering capacity toward alkaline samples. At pH 8.3, only about 10% of the boric acid is converted to borate. The remaining 90% is available as a buffer preventing large changes of pH due to alkaline samples matrices. Carbonate elutes with chloride under the conditions listed in Fig. 3. To prevent this, eluent A can be changed to either 7.28 m*M* or 10.68 m*M* boric acid adjusted to pH 8.6 with KOH (ref. 8).

Optimization of separation conditions for samples containing elevated levels of chloride was studied next. The chromatogram shown in Fig. 4 shows traces of nitrite, bromide, nitrate, phosphate, and sulfate relatively well resolved from the chloride matrix peak. However, the distorted shape of the fluoride peak is an indication of a strong interference that would be typical for a sample of this type.

The distortion of the fluoride peak was investigated with a series of standard mixtures containing 1 ppm fluoride in the presence of several different levels of chloride. As shown in Fig. 5, the size of the peak for constant levels of fluoride decreases with increasing amounts of chloride. Between 2 and 100 ppm of chloride, the fluoride peak was not yet visibly distorted. Consequently, the interference of chloride at con-

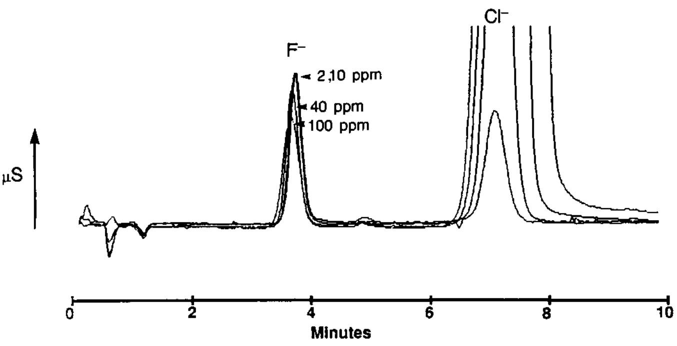

Fig. 5. Decreasing peak heights of 1 ppm fluoride peak with increasing concentrations of chloride (2 to 100 ppm). Column: IC Pak Anion. Isocratic elution with 10 m*M* boric acid, pH 8.3.

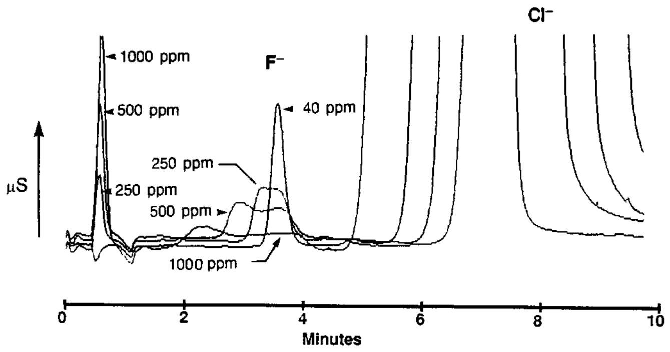

Fig. 6. Distortion of peak shapes for 1 ppm fluoride at elevated levels of chloride (40 to 1000 ppm). Conditions are the same as in Fig. 6.

centrations about 20–100 ppm with the quantitative determination of fluoride could be easily overlooked. At a higher range of chloride concentrations, 250–1000 ppm, the fluoride peak showed distortion that increased with increasing chloride concentration (Fig. 6). At the same time, the peak at the void (retention time = 0.2 min) increased in peak height with increasing chloride concentration. Normally the void peak is negative and is called a water dip. It is highly unusual for this peak to be positive. In an experiment with 2.8 m*M* $NaHCO_3$ and 2.2 m*M* Na_2CO_3 eluent and two IC Pak A columns in series, the void was collected and reinjected onto the ion chromatograph. It was found that virtually all of the lost fluoride and some excess chloride was in the void peak. The peak heights of the other anions —nitrite, bromide, nitrate, phosphate, and sulfate— were not affected. The peak height of fluoride was affected by the presence of up to 2000 ppm of chromate, nitrate, or phosphate. Experiments performed with membrane suppression detection showed the same behavior.

Table I shows that the counter ion of the sample has no effect of the fluoride peak reduction. The rate of peak height reduction with increasing chloride concentra-

TABLE I

PEAK HEIGHT OF FLUORIDE IN THE PRESENCE OF EXCESS KCl, LiCl, AND HCl SAMPLE

5.6 m*M* $NaHCO_3$, 4.4 m*M* Na_2CO_3 isocratic eluent, 1 ml/min with 2 IC Pak A columns in series. All chromatograms and data are from 100-µl injections.

ppm Cl	Peak height (cm)		
	KCl	LiCl	HCl
2	3.55	3.50	3.50
40	3.35	3.40	3.40
100	3.35	3.40	3.35
250	3.35	3.25	3.25
500	2.60	2.70	2.70
1000	2.30	2.30	2.30

tion was the same in all three cases. Therefore, it is unlikely that loss of fluoride is due to protonation of fluoride by the sample matrix.

Next, other early eluting anions were investigated. Fig. 7 shows a plot of normalized peak height of fluoride, propionate, and iodate. The results show that iodate is affected the greatest, then fluoride, and finally propionate. The relative loss of each anion is related to the affinity of the anion for the anion-exchange resin. Anions with the lowest selectivity for the anion-exchange column have the greatest loss of peak height. The elution order of these anions is iodate, fluoride, and propionate. In separate experiments, acetate and formate also showed reduced peak height.

Early eluting peaks by nature compete poorly for the ion-exchange sites. When an injection is made, three different anions are in competition: the early eluting anion, chloride, and the eluent anion. Presumably, changing the eluent type might help. Three different eluents were tried in these experiments: borate, carbonate/bicarbonate, and hydroxide. But when the eluent concentration for each eluent is adjusted so that sample peak elutes at roughly the same time, the peak reduction behaviour is similar for eluent types tested.

Increasing the column capacity may help. In fact, better results by a factor of 2 were obtained when two columns were connected in series. However, it should be

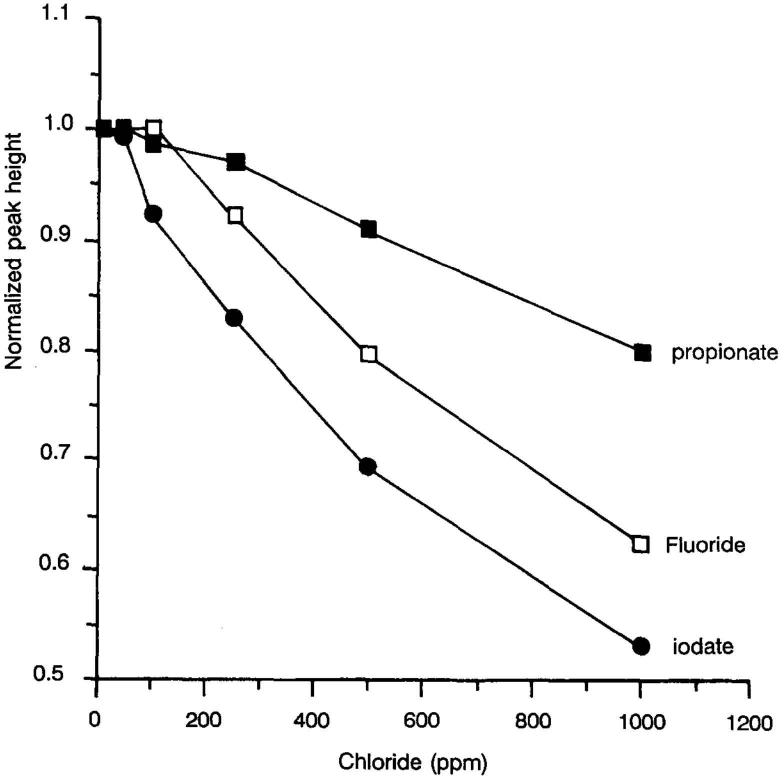

Fig. 7. Plots of normalized peak heights of 30 ppm propionate, 5 ppm fluoride, and 30 ppm iodate as a function of chloride concentration. Column: 2 × IC Pak Anion column in series. Eluent: 2.8 m*M* $NaHCO_3$, 2.2 m*M* Na_2CO_3.

noted that simply increasing the capacity may not be enough. Some commercial columns may have higher capacity, but have poorer selectivity for early eluting anions (indicated by whether these anion elute near the void).

Another method is to use ion-exclusion chromatography to determine anions of weak acids [10]. Trace amounts of these acids can be determined in the presence of large amounts of anions of strong acids. Of course, anions of strong acids cannot be determined unless a coupled system (ion exclusion and ion exchange) is used. Then both types of anions can be determined simultaneously.

An eluent gradient may be used to adjust eluent conditions so that the early eluting anions are better retained by the column. Thus, the early eluting anions do not have to compete with the eluent anions to be retained by the anion-exchange sites. This increases the peak window between chloride and the void peak. Under some experiments performed with a Sarasep AN1 column, a 10-min gradient of 0 to 10 mM KOH gradient produced no loss of 2 ppm fluoride in the presence of 500 ppm of chloride. However, it's impossible to be more than qualitative with recommending gradient conditions. Whether a gradient works depends on the type selectivity, and condition of the anion-exchange column. It may be better to remove chloride with a sample preparation column prior to analysis. In any case, method calibration should be performed with standard solutions that are similar to the sample matrix.

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