Environmental Water Applications Notebook

Anions • Cations • Bromate • Haloacetics Acids • Disinfection Byproducts

Thermo SCIENTIFIC

Table of Contents

Introduction to Environmental Water Analysis

Everyone in the global community is impacted by the quality of water resources. The water we drink must be free from harmful chemicals to ensure good health. The purity of ground and surface waters in our environment is critical to ensuring sustainable use. The water discharged by municipal wastewater treatment plants and industrial facilities must be monitored to ensure strict compliance with environmental guidelines. Process waters must be kept clean from contaminants to ensure product quality and acceptable exposure levels.

Thermo Fisher Scientific is committed to enhancing the quality of our global water resources. As innovation leaders in ion and liquid chromatography, our analytical instruments are used by government and industry to provide solutions for environmental water testing for a wide range of regulated and emerging inorganic elements and organic compounds.

As pioneers of suppression technology, we started a revolution in ion chromatography (IC) that increased the sensitivity and accuracy of ion determination. As constant innovators, we developed Reagent-Free™ (RFIC™) systems that set a new benchmark for ion analysis. Today, RFIC systems with eluent generation and eluent regeneration provide the ultimate in sensitivity and ease of use.

We also have a full high-performance liquid chromatography (HPLC) product line for the analysis of organic contaminants, from nano- to preparative-scale separation capabilities, including ultra HPLC (UHPLC).

In fact, we are the only separations science company that provides instrumentation, columns, and applications perfectly suited for both inorganic and organic contaminants.

THERMO SCIENTIFIC AND DIONEX INTEGRATED SYSTEMS

Dionex Products are now a part of the Thermo Scientific brand, creating exciting new possibilities for scientific analysis. Now, leading capabilities in LC, IC, and sample preparation are together in one portfolio with those in mass spectrometry (MS). Combining Dionex's leadership in chromatography with Thermo Scientific's leadership position in mass spec, a new range of powerful and simplified workflow solutions now becomes possible.

For more information on how the new line-up of Thermo Scientific products can expand your capabilities and provide the tools for new possibilities, choose one of our integrated solutions:

- Ion Chromatography and Mass Spectrometry
- Liquid Chromatography and Mass Spectrometry
- Sample Preparation and Mass Spectrometry

GROUND AND SURFACE WATER

Surface water is the largest source of fresh water used for human consumption. The U.S. Geological Survey implemented the National Water-Quality Assessment (NAWQA) Program in 1991 to develop long-term data on streams, rivers, groundwater, and aquatic systems. The data support national, regional, state, and local policies and decisions related to water-quality management. The NAWQA program is designed to answer the following questions:

- What is the condition of our nation's streams, rivers, and groundwater?
- How are these conditions changing over time?
- How do natural features and human activities affect these conditions, and where are those effects most pronounced?

Thermo Scientific has codeveloped several methods with the U.S. EPA Office of Ground Water and Drinking Water. This collaboration has strengthened with the development of unique technology, including electrolytic suppression and RFIC with eluent generation or regeneration.

DRINKING AND BOTTLED WATER

Currently, less than 1% of the planet's water is available for human consumption—making this valuable resource even more important. With surface water contamination and groundwater resources overexploited, the need for effective water analysis and monitoring has never been higher.

Regulatory agencies around the world have developed standards for water analysis and have provided guidance on water disinfection to assure drinking water quality. Thermo Scientific provides a variety of solutions for inorganic and organic drinking water contaminants.

WASTEWATER

Wastewater includes liquid waste from residences, industry, and agriculture, comprising a wide range of potential contaminants and concentrations. Industries discharge a variety of pollutants in their wastewater, including heavy metals, organic toxins, oils, nutrients, and solids, all of which endanger ecosystems and pose a threat to human health. In some areas, treated wastewater is recycled for irrigation purposes and even as drinking water. This reuse of water is gaining closer scrutiny as demand increases for water resources.

Treating and recycling wastewater requires careful analysis and monitoring, including the determination of low-level contaminants such as pharmaceuticals and personal care products (PCPs). Dionex HPLC and IC instruments are well suited to determine a wide range of nonpolar, polar, and ionic contaminants.

FAST WATER ANALYSIS

High-Throughput Solutions for Inorganic and Organic Contaminant Analyses

The Challenge:

Emerging contaminants, stricter regulations, growing municipalities and industries—all increase analytical laboratories' workloads, requiring processing of more samples and performing more tests in less and less time.

We have developed new technologies and methods to help labs and businesses increase their productivity and throughput for the analysis of inorganic and organic contaminants in a variety of water matrices.

Columns

Thermo Scientific Dionex IonPac Fast IC columns for anions, organic acids, oxyhalides, cations, and amines use the same proven chemistry in shorter column formats, decreasing run times by as much as three times while still retaining sufficient resolution.

Thermo Scientific Acclaim columns for organic contaminants use smaller particles that allow higher flow rates at standard pressures and compatibility with higher pressure systems. When used with the Thermo Scientific Dionex UltiMate 3000 rapid separation LC (RSLC) systems, these columns provide separation times as much as 30 times faster than standard columns and systems.

Inorganic Contaminants

The Thermo Scientific Dionex ICS-5000 capillary RFIC system provides IC on demand, reducing equilibration times and calibration requirements that save labor and increase throughput. The innovative Thermo Scientific Dionex IC Cube module, with half the connections of a standard IC configuration, makes plumbing and reconfiguring the system easier. Capillary Fast IC and monolith columns combine the speed of Fast IC with the convenience of IC whenever you need it—on demand. The simultaneous injection, sample, and standard preparation features of the Thermo Scientific Dionex AS-AP Autosampler, along with its AutoDilution capability, increase throughput, reduce manual labor, and decrease delays from out-of-range samples.

Organic Contaminants

UltiMate™ 3000 HPLC and RSLC systems are all UHPLC+ focused, enabling faster separations at standard HPLC system prices. From the economical Basic Automated system to the ×2 Dual RSLC system for high throughput, automated sample preparation, sample concentration, and matrix elimination, Thermo Scientific has the system to fit your needs and budget.

Thermo Scientific Dionex Chromeleon Chromatography Data System software version 7.1 streamlines your path from samples to results. eWorkflows guide the operator through a minimal number of choices needed to run that workflow, making configuration of even the most complex multidimensional analysis easy. Data analysis tools help users process chromatograms with minimal effort, report templates and audit trails, and help ensure regulatory compliance, and System Wellness tools increase up time.

Thermo Scientific is committed to enhancing the quality of our global water resources. Our analytical instruments are used by government and industry labs globally to provide services for environmental water testing for a wide range of regulated and emerging inorganic elements and organic compounds.

Analysis of Anions

Environmental Water Applications Notebook

Monitoring Inorganic Anions and Cations During Desalination

INTRODUCTION

As of 2009, there were 14,450 desalination plants worldwide producing more than 60 million cubic meters of water a day.[1] Because of the growing demand for water and the limited supply of fresh water, desalination increasingly is being used to produce potable and irrigation water from salty or brackish water. The global market for desalination to generate supplies of potable water is projected to grow at an annual rate of 10% over the next 10 years. Seawater desalination is a $10 billion industry today and is forecasted to reach $16 billion in 2020.[1]

A wide variety of desalination techniques are currently available and more are being developed. Most use distillation or membrane techniques. The performance of desalination processes is evaluated by monitoring the common anions and cations in the feed, intermediate, and final water product. For the final drinking water product, ion chromatography (IC) is approved for monitoring primary and secondary anions according to U.S. Environmental Protection Agency (EPA) method 300.0,[2] and Federal and State regulatory agencies ensure that U.S. National Primary and Secondary Drinking Water Standards are met.

Common cations, though not considered contaminants, are monitored and reported by many public water suppliers in the United States. Cations, particularly calcium and magnesium, are measured to determine water hardness. In addition to calcium and magnesium, ammonium is also measured and regulated in public water supplies in EU countries and Japan. During desalination, the levels of divalent cations affect performance of membrane processes like reverse osmosis (RO).[3] High levels of calcium or magnesium result in frequent fouling of the membranes, which is highly undesirable. Therefore, it is critical to monitor anions and cations at all stages of desalination.

Another challenge for the desalination of seawater is the removal of boron, which is typically found at concentrations of 4.5 mg/L. World Health Organization 2008 guidelines suggest a concentration of 0.5 mg/L,[4] whereas the U.S. EPA recommends a maximum lifetime exposure of 0.6 mg/L.[5] Depending on pH levels, boron can exist in ionic and non-ionic forms. Above pH 8, the removal efficiency using RO is enhanced due to the formation of borate. RO membranes remove ions better than non-ionic forms of the same compounds. This suggests that raising the pH may improve the removal efficiency of boron. However, raising the pH too high results in the formation of scales formed by the precipitation of carbonate salts of calcium and magnesium, which can disrupt membrane performance. In addition to monitoring the pH, it is important to know the concentration of scale-forming divalent cations calcium and magnesium in order to maintain optimal RO membrane performance.

Compared to traditional sources of water, desalination is an energy-intensive process that requires expensive infrastructure. The potential benefits of desalination are constantly being evaluated because of the high economic and environmental costs. Hence, efficient water monitoring techniques are needed to understand the robustness of desalination processes.

This work describes an IC method using a Dionex ICS-3000 system with IonPac® AS18 anion-exchange and CS12A cation-exchange columns, electrolytically generated hydroxide and methanesulfonic acid eluents, and suppressed conductivity detection to simultaneously measure the common anions and cations in water samples obtained from desalination processes. This method uses a 2 mm column format for anion separations, a 3 mm column format for cation separations, and electrolytically generated eluents that require only the addition of deionized water for continuous operation. The linearity, method detection limits (MDLs), precision, and recovery of anions and cations in saline and drinking water matrices for this method are discussed here. This IC method supports all the monitoring needs of a desalination facility because it can measure anions and cations in diverse matrices ranging from seawater to drinking water.

EQUIPMENT

Dionex ICS-3000 Reagent-Free™ Ion Chromatography system* with eluent generation (RFIC-EG™) including:

DP Dual Pump module

EG Eluent Generator module

DC Detector/Chromatography module (single- or dual-temperature zone configuration)

AS Autosampler (with Simultaneous Injection Upgrade Kit, Dionex P/N 063742)

EluGen EGC II KOH cartridge (Dionex P/N 058900)

Continuously-Regenerated Anion Trap Column, CR-ATC II (Dionex P/N 060477)

EluGen EGC II MSA cartridge (Dionex P/N 058902)

Continuously-Regenerated Cation Trap Column, CR-CTC II (Dionex P/N 066262)

Chromeleon® 6.8 or 7 Chromatography Workstation

Polystyrene Autoselect™ vials with caps and septa, 10 mL (Dionex P/N 055058)

Nalgene® 125 mL HDPE narrow mouth bottles (VWR P/N 16057-062)

Nalgene 250 mL HDPE narrow mouth bottles (VWR P/N 16057-109)

Nalgene 250 mL 0.2 µm nylon filter units (VWR P/N 28199-371)

Nalgene 1000 mL 0.2 µm nylon filter units (VWR P/N 28198-514)

*The applications described here can run on any Dionex RFIC system. The applications also can run with manually prepared eluents on any Dionex IC system.

REAGENTS AND STANDARDS

Deionized water, Type I reagent grade, 18 MΩ-cm resistivity or better, filtered through a 0.2 µm filter immediately before use

Fluoride Standard 1000 mg/L (Dionex P/N 037158)

Chloride Standard 1000 mg/L (Dionex P/N 037159)

Nitrite Standard 1000 mg/L (UltraScientific P/N ICC-007)

Bromide Standard 1000 mg/L (UltraScientific P/N ICC-001)

Sulfate Standard 1000 mg/L (UltraSceintific P/N ICC-006)

Nitrate Standard 1000 mg/L (UltraScientific P/N ICC-004)

Phosphate Standard 1000 mg/L (UltraScientific P/N ICC-005)

Lithium Standard 1000 mg/L (UltraScientific P/N ICC-104)

Sodium Standard 1000 mg/L (UltraScientific P/N ICC-107)

Ammonium Standard 1000 mg/L (UltraScientific P/N ICC-101)

Potassium Standard 1000 mg/L (UltraScientific P/N ICC-106)

Magnesium Standard 1000 mg/L (UltraScientific P/N ICC-105)

Calcium Standard 1000 mg/L (UltraScientific P/N ICC-104)

Sodium Chloride (J.T. Baker P/N 4058-05)

Sodium Sulfate (VWR P/N EM-SX0760-1)

Sodium Nitrite (JT Baker P/N 1-3780)

Sodium Bromide (Aldrich P/N 31050-6)

Sodium Nitrate (Baker P/N 3770-05)

Potassium Phosphate Monobasic (Fisher P/N P286-1)

Lithium Chloride (Fisher, P/N L-121-100)

Ammonium Chloride (Sigma A-5666)

Potassium Chloride (Mallinckrodt P/N 6858)
Magnesium Chloride Hexahydrate (BDH P/N 0244 5009)
Calcium Chloride Dihydrate (Fisher P/N C-79)
Combined Six Cation Standard-II (Dionex P/N 046070)
Combined Seven Anion Standard (Dionex P/N 66933)

CONDITIONS

Anion Determinations

Columns:	IonPac AG18, 2 × 50 mm IonPac AS18, 2 × 250 mm
Eluent:	22 mM KOH from 0–7 min, 22–40 mM KOH from 7–8 min, 40 mM KOH from 8–18 min*
Eluent Source:	EGC II KOH with CR-ATC
Injection Volume:	4 µL
Flow Rate:	0.25 mL/min
Detection:	Suppressed conductivity, ASRS® 300, 2 mm, recycle mode, suppressor current 15 mA
Background Conductance:	<1 µS

Cation Determinations

Columns:	IonPac CG12A-5 µm, 3 × 30 mm IonPac CS12A-5 µm, 3 × 150 mm
Eluent:	20 mM MSA
Eluent Source:	EGC II MSA with CR-CTC
Injection Volume:	10 µL
Flow Rate:	0.50 mL/min
Detection:	Suppressed conductivity, CSRS® 300, 2 mm, recycle mode, suppressor current 30 mA
Background Conductance:	<0.5 µS

Both Anion and Cation Determinations

Temperature:	30 °C (column and detector compartment)
Noise:	~0.5–1.0 nS (conductivity)
System Backpressure:	~2500 psi
Run Time:	20 min (including column equilibration time)

*The column equilibrates for 2 min at 22 mM KOH prior to injection.

PREPARATION OF SOLUTIONS AND REAGENTS

Eluent Solutions

Generate potassium hydroxide (KOH) and methanesulfonic acid (MSA) eluents online by pumping high-quality degassed, deionized (DI) water through the EGC II KOH and MSA cartridges, respectively. Chromeleon Chromatography Data System (CDS) software tracks the amount of KOH and MSA used and calculates the remaining lifetime. Although electrolytic eluent generation delivers the best performance, manually prepared eluents may be used, if needed.

Stock Standard Solution

Certified standard solutions can be purchased from Dionex or other commercial sources. When commercial standards are not available, 1000 mg/L stock standard solutions can be prepared by dissolving appropriate amounts of the required analyte in DI water in a plastic volumetric flask (Table 1). Store in plastic containers at 4 ºC. Stock standards are stable for at least 3 months.

Working Standard Solutions

Prepare composite working standards at lower analyte concentrations by diluting appropriate volumes of the 1000 mg/L stock with DI water. Prepare working standards containing < 100 mg/L anions or cations daily. Store standard solutions at < 6 °C when not in use.

Table 1. Mass of Compound Required to Prepare 1L of 1000 mg/L Stock Standard Solutions

Analyte	Compound	Amount (g)
Fluoride	Sodium fluoride (NaF)	2.210
Chloride	Sodium chloride (NaCl)	1.648
Nitrite	Sodium nitrite ($NaNO_2$-N)	4.926
Bromide	Sodium bromide (NaBr)	1.288
Nitrate	Sodium nitrate ($NaNO_3$-N)	6.068
Sulfate	Sodium sulfate (Na_2SO_4)	1.479
Phosphate	Potassium phosphate monobasic (KH_2PO_4-P)	4.394
Lithium	Lithium chloride (LiCl)	6.108
Sodium	Sodium chloride (NaCl)	2.542
Ammonium	Ammonium chloride (NH_4Cl)	2.965
Potassium	Potassium chloride (KCl)	1.907
Magnesium	Magnesium chloride hexahydrate ($MgCl_2 \cdot 6H_2O$)	8.365
Calcium	Calcium chloride dihydrate ($CaCl_2 \cdot 2H_2O$)	3.668

SAMPLE PREPARATION

Artificial Seawater

Prepare simulated seawater by diluting the salts listed in Table 2 into 1 L of DI water following the method of Kester et al.[6] with the exclusion of strontium chloride. This yields a solution with approximately 3.5% salinity.

Table 2. Salts Added to Form Simulated Seawater (1L)

Compound	Amount (g)
Sodium chloride	2.393
Sodium sulfate	4.008
Potassium chloride	0.677
Sodium bicarbonate	0.196
Potassium bromide	0.098
Boric acid	0.026
Sodium fluoride	0.003

Commercial Aquarium Sea Salt

Follow package directions (1/2 cup of salt per gallon of DI water) to prepare commercially available synthetic sea salt, creating a solution of approximately 3.5% salinity. Prepare a 1 L portion with 30 g of aquarium salt. (A sea salt density of approximately 2.2 g/cm^3 was used to convert the preparation directions to metric units.[7])

Seawater (From California's San Francisco Bay)

Collect surface seawater in a 250 mL HDPE Nalgene bottle that has been precleaned before sample collection. Store the sample on ice until it can be filter sterilized through a 250 mL, 0.2 µm nylon filter unit. After filtration, store the sample at < 6 °C.

Filter all samples through a 0.2 µm nylon filter unit before injection.

SYSTEM PREPARATION AND CONFIGURATION

Configure the autosampler (AS) for simultaneous injection into the anion and cation detection systems. In the simultaneous mode, the AS delivers sample to two independent IC systems. The sample is injected simultaneously and equally to both systems (two injection valves are required). Dual analyses can be performed with only one sample. A 5 or 10 mL syringe and an 8.5 mL sampling needle assembly are required for simultaneous injections. Full-loop injections are required for this mode.

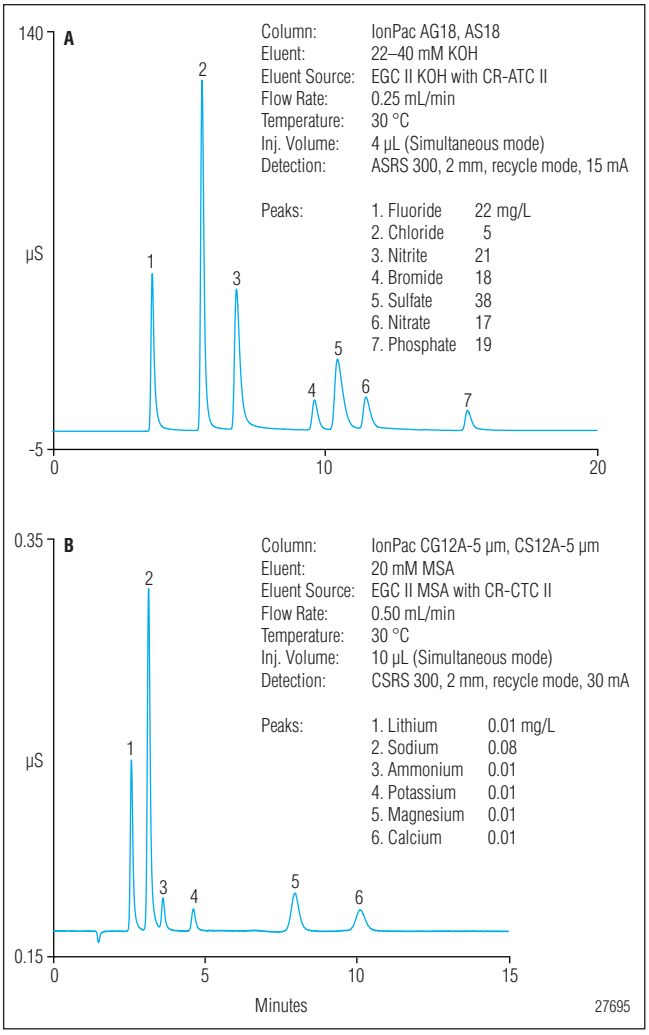

Figure 1. Separation of common A) anions using the IonPac AS18 column and B) cations using the IonPac CS12A column.

Using Chromeleon software, configure the two chromatography systems and the AS into a single timebase and assign each system a unique device name and channel. Use one control panel to monitor and control both systems and all samples in one sequence.

The system also may be configured for sequential injection into the two IC systems. In the sequential option, the sample is delivered to the first system, flow is rerouted (diverted), and then sample is delivered to the second system.[8]

RESULTS AND DISCUSSION

Using the IonPac CS12A and AS18 columns, the common anions and cations were easily resolved in 20 min (Figure 1 A and B). Note that this method provided good resolution between sodium and ammonium, the two analytes that can be challenging to resolve, especially

Table 3. Linear Range, MDLs, and Precisions for Anions and Cations							
Analyte	**Range (mg/L)**	**Corr. Coeff. (r^2)**	**MDL Standard (µg/L)**	**MDL (µg/L)[a]**	**QCS (mg/L)**	**Retention Time Precision (RSD)[b]**	**Peak Area Precision (RSD)[b]**
Lithium	0.02–16	0.9999	1	0.08	1	0.07	0.20
Sodium	0.10–100	0.9999	4	0.13	4	<0.01	0.29
Ammonium[c]	0.01–8	0.9997	5	0.10	5	<0.01	0.97
Potassium	0.02–16	0.9997	10	0.10	10	0.03	0.61
Magnesium	0.02–80	0.9998	5	0.53	5	0.03	0.09
Calcium	0.02–80	0.9999	5	0.36	5	0.04	0.15
Fluoride	0.08–100	0.9996	10	0.62	2	0.05	0.05
Chloride	0.24–300	0.9999	10	0.66	20	0.02	0.04
Nitrite (-N)	0.08–100	0.9994	20 (67 as NO_2^-)	0.51 (-N)	2 (6.7 as NO_2^-)	0.03	0.07
Bromide	0.08–100	0.9994	25	0.46	2	0.02	0.05
Sulfate	0.16–201	0.9994	20	0.67	60	0.01	0.07
Nitrate (-N)	0.02–22	0.9999	27 (120 as NO_3^-)	0.24 (-N)	2.3 (10 as NO_3^-)	0.02	0.08
Phosphate (-P)	0.03–33	0.9999	23 (70 as PO_4^{3-})	0.15 (-P)	0.7 (2 as PO_4^{3-})	0.04	0.14

[a]MDL = (t) × (S) where t = Student's *t* value for a 99% confidence level and a standard deviation estimate with n-1 degrees of freedom [t = 3.14 for seven replicates of the MDL Standard], and S = standard deviation of the replicate analysis

[b]Relative standard deviation, n = 7

[c]Quadratic fit

when one is in a large excess relative to the other. This method also achieved good retention time for fluoride, which was well resolved from the void volume.

Table 3 summarizes the calibration data, the method detection limits (MDLs), retention time, and peak area precisions for the common anions and cations. MDLs and precision data were obtained from seven replicate injections of the MDL and QCS standards, respectively, prepared in DI water. Anion and cation MDL standards were prepared at concentrations of 3–5× the estimated method detection limits.

Correlation coefficient values obtained from the calibration plots were between 0.9994 and 0.9999. The calibration curves were linear for all anions and cations except ammonium. Analytes that form weak acids or bases in the suppressor are known to exhibit nonlinear behavior. A quadratic curve-fitting function was used for ammonium. The retention time precision ranged from < 0.01–0.07%, and the peak area precision ranged from 0.04–0.97%. The high retention time precisions are attributed to consistent generation of high-purity KOH and MSA using the eluent generator module and the respective continuously regenerated trap columns (CR-ATC and CR-CTC).

Table 4. Between-Day (n=3) Retention Time (RT) and Peak Area Precisions (Triplicate Injections of Spiked Sample)									
Cations					**Anions**				
San Francisco Bay Water									
Analyte	**RT (min)**	**RT Precision (RSD)**	**Peak Area (µS*min)**	**Peak Area Precision (RSD)**	**Analyte**	**RT (min)**	**RT Precision (RSD)**	**Peak Area (µS*min)**	**Peak Area Precision (RSD)**
Lithium	2.57	<0.01	0.72	0.13	Fluoride	3.63	<0.01	0.39	0.57
Sodium	3.19	0.06	18.4	0.07	Chloride	5.45	0.00	40.3	0.31
Ammonium	3.61	0.02	0.32	0.20	Nitrite	6.77	0.02	0.61	0.64
Potassium	4.59	0.03	0.51	0.40	Bromide	9.63	0.03	0.14	0.82
Magnesium	7.87	0.02	4.31	0.12	Sulfate	10.53	0.01	5.14	0.20
Calcium	9.96	0.02	0.91	0.38	Nitrate	11.57	0.01	0.26	0.63
					Phosphate	15.44	0.03	0.09	0.65
Commercial Aquarium Sea Salt									
Lithium	2.57	0.05	0.72	0.15	Fluoride	3.63	0.05	0.37	0.52
Sodium	3.19	0.02	18.4	0.05	Chloride	5.45	0.02	40.2	0.18
Ammonium	3.61	<0.01	0.29	0.54	Nitrite	6.77	0.02	0.63	0.79
Potassium	4.59	0.03	0.52	0.30	Bromide	9.63	0.01	0.14	0.63
Magnesium	7.91	0.02	4.32	0.09	Sulfate	10.54	0.03	5.11	0.26
Calcium	10.08	0.01	0.88	0.41	Nitrate	11.57	0.01	0.26	0.68
					Phosphate	15.45	0.02	0.09	1.01
Artificial Seawater									
Lithium	2.57	0.05	0.71	0.22	Fluoride	3.63	< 0.01	0.37	0.39
Sodium	3.20	0.04	21.2	0.05	Chloride	5.45	0.01	45.83	0.22
Ammonium	3.61	<0.01	0.31	0.52	Nitrite	6.77	0.02	0.62	0.61
Potassium	4.59	0.03	0.58	0.37	Bromide	9.63	0.01	0.15	0.62
Magnesium	7.91	0.01	4.56	0.36	Sulfate	10.52	0.02	3.92	0.27
Calcium	10.07	0.03	0.94	0.50	Nitrate	11.57	0.01	0.27	0.24
					Phosphate	15.44	0.02	0.09	0.55

Method performance was evaluated by measuring recoveries in samples of spiked saline (Table 4) and drinking water (Table 5). Samples were spiked with analytes at a level that was 50–100% of the amount determined in the original sample. The between-day precision for anions and cations in the spiked samples ranged from < 0.01–1.6% over three days.

Table 5. Between-Day (n=3) Retention Time (RT) and Peak Area Precisions (Triplicate Injections of Spiked Sample)									
Cations					**Anions**				
Tap Water									
Analyte	**RT (min)**	**RT Precision (RSD)**	**Peak Area (µS*min)**	**Peak Area Precision (RSD)**	**Analyte**	**RT (min)**	**RT Precision (RSD)**	**Peak Area (µS*min)**	**Peak Area Precision (RSD)**
Lithium	2.57	0.05	0.75	0.13	Fluoride	3.63	0.04	0.82	0.34
Sodium	3.16	0.04	8.34	0.10	Chloride	5.47	0.01	7.47	0.32
Ammonium	3.61	0.02	0.48	0.14	Nitrite	6.78	0.01	0.29	0.46
Potassium	4.59	<0.01	0.27	0.15	Bromide	9.64	0.02	0.12	0.76
Magnesium	7.91	0.02	4.11	0.19	Sulfate	10.53	0.03	5.13	0.34
Calcium	9.97	0.01	6.57	0.18	Nitrate	11.56	0.01	0.31	0.66
					Phosphate	15.44	0.01	0.09	0.89
Bottled Mineral Water									
Lithium	2.57	0.03	0.75	0.13	Fluoride	3.63	0.04	0.90	0.49
Sodium	3.16	<0.01	8.57	0.07	Chloride	5.47	0.01	6.12	0.33
Ammonium	3.61	0.04	0.29	0.54	Nitrite	6.76	0.02	0.22	0.65
Potassium	4.59	0.01	0.61	0.09	Bromide	9.61	0.01	0.12	0.76
Magnesium	7.80	0.02	11.4	0.12	Sulfate	10.44	0.03	8.52	0.50
Calcium	9.73	0.01	18.5	0.21	Nitrate	11.52	0.02	0.74	0.34
					Phosphate	15.51	0.03	0.09	0.53

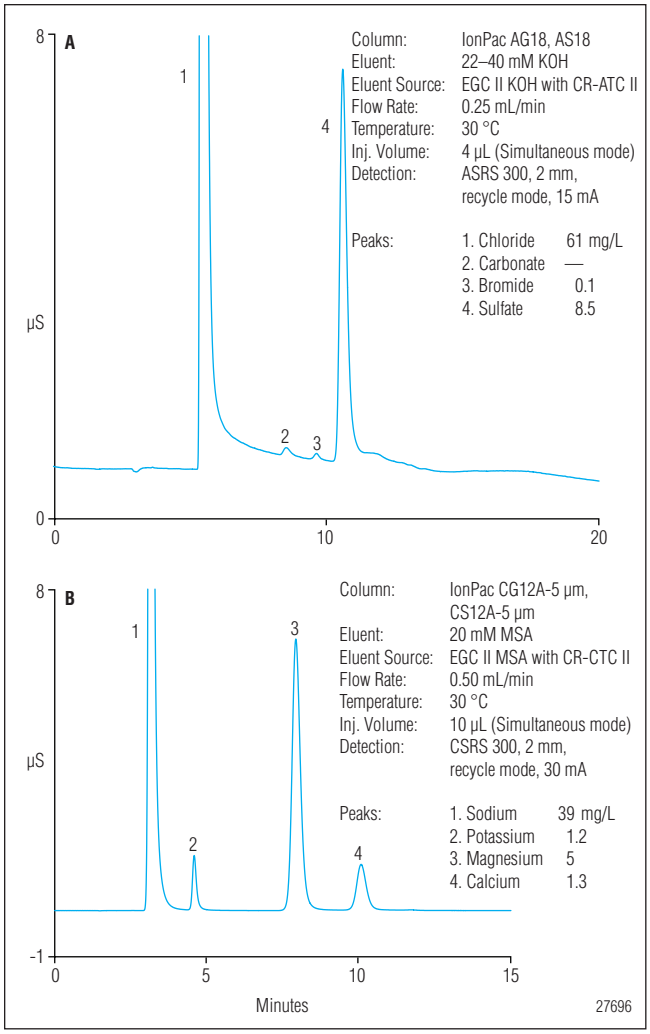

Figure 2. San Francisco, CA bay water: determination of common A) inorganic anions using the IonPac AS18 column and B) cations using the IonPac CS12A column.

Figure 2 shows the separation of A) anions and B) cations in water from California's San Francisco Bay. The bay water sample is representative of the typical feed water into a desalination plant. The bay water sample was diluted 200-fold so that measured levels were within the calibrated range. The major inorganic anions in bay water are chloride and sulfate, and the major inorganic cations are sodium, potassium, magnesium, and calcium.

Table 6. Inorganic Anion and Cation Recoveries										
Analyte	**San Francisco Bay Water**		**Artificial Seawater**		**Commercial Aquarium Sea Salt**		**Bottled Mineral Water**		**Tap Water**	
	Amount Added (mg/L)	**Recovery* (%)**	**Amount Added (mg/L)**	**Recovery* (%)**	**Amount Added (mg/L)**	**Recovery* (%)**	**Amount Added (mg/L)**	**Recovery* (%)**	**Amount Added (mg/L)**	**Recovery* (%)**
Lithium	1	93.5	1	92.4	1	98.1	1.0	96.9	1	97.2
Sodium	40	89.9	40	95.3	40	90.9	20.1	93.1	20	96.6
Ammonium	1	108.3	1	105.7	1	105.9	1.0	97.9	1	99.9
Potassium	2	94.5	2	96.7	2	99.6	1.0	94.9	1	95.7
Magnesium	5	97.2	5	97.4	5	97.6	5.0	94.6	5	97.1
Calcium	2	88.4	2	83.7	2	82.8	15.0	95.7	15	85.6
Fluoride	1	109.6	1	107.0	1	106.9	1	98.1	1	98.7
Chloride	74	87.2	74	92.7	74	89.7	16	84.2	16	85.9
Nitrite (-N)	1	100.7	1	101.7	1	105.3	1	37.2	1	41.5
Bromide	1	84.4	1	83.6	1	84.1	1	87.9	1	88.4
Sulfate	16	81.2	16	83.9	16	84.9	16	93.9	16	84.4
Nitrate (-N)	0.2	98.4	0.2	103.7	0.2	100.0	0.2	100.9	0.2	101.3
Phosphate (-P)	0.3	88.0	0.3	84.3	0.3	86.6	0.3	88.4	0.3	85.0

*Average over 3 days

As seen in Figure 2 and Table 6, all anions and cations were well resolved and had acceptable recoveries (80–120%) using the criteria outlined in U.S. EPA Method 300.0.

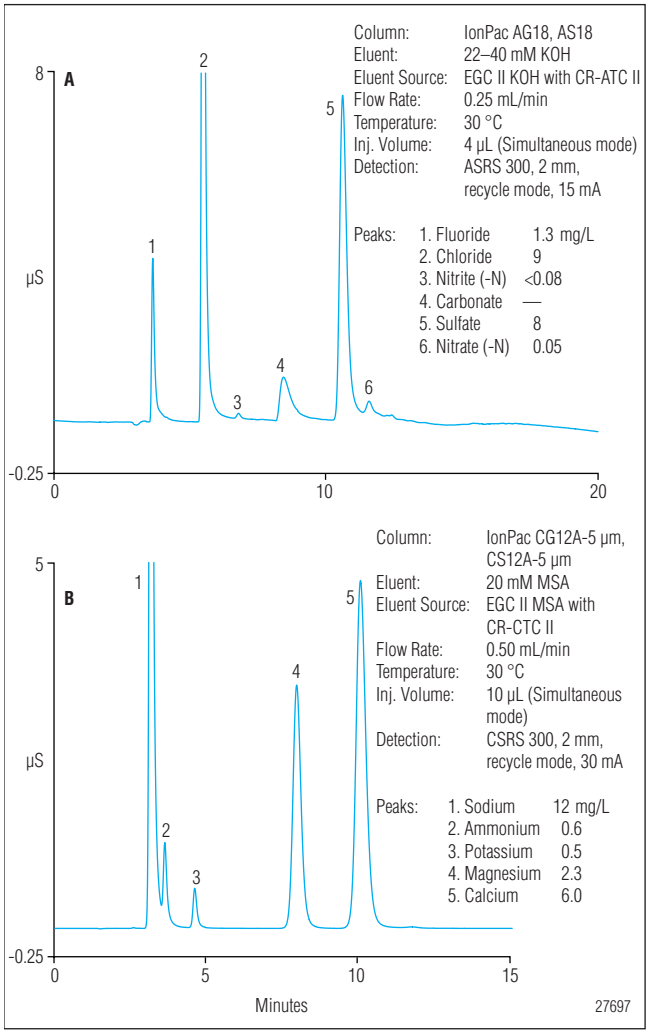

Figure 3. Sunnyvale, CA tap water: determination of common A) inorganic anions using the IonPac AS18 column and B) cations using the IonPac CS12A column.

Figure 3 (A and B) shows the chromatogram for all anions and cations in Sunnyvale, CA drinking water. Tap water samples have fluoride, chloride, and sulfate as the predominant inorganic anions. Table 6 lists the recoveries of anions and cations in the drinking water matrices. All anions and cations were well resolved and, with the exception of nitrite, had acceptable recoveries (80–120%). The low recovery of nitrite can be attributed to biological activity in these samples (which is minimal in the high saline matrices) and the instability of nitrite in oxidizing environments, such as chlorinated water or other oxidizing disinfectants in drinking water.

In summary, the current methods using the IonPac AS18 and CS12A columns provide acceptable recoveries for anions and cations in both saline and drinking water matrices. This work shows methods that can be used for diverse matrices that are typically encountered in a desalination plant.

CONCLUSION

IonPac AS18 and CS12A columns with electrolytically generated hydroxide and MSA eluents can simultaneously determine anions and cations in saline and drinking water matrices. The capacities of the IonPac AS18 and CS12A columns allow sample analysis with minimal sample pretreatment. The RFIC-EG system allows continuous operation of the instrument with minimal maintenance. Only water for eluent generation and suppressor regeneration must be added to keep the instrument running for sample analysis. Additionally, the smaller column format generates less waste and uses less eluent, saving both time and money. The methods were shown to be accurate by the good recovery of anions and cations in a wide variety of samples including natural and artificial seawater and drinking water. These methods are robust for all ion-monitoring needs of a typical desalination facility and support a varying range of matrices from seawater to drinking water.

SUPPLIERS

Fisher Scientific, 2000 Park Lane Drive, Pittsburgh, PA 15275, U.S.A. Tel: 800.766.7000. www.fishersci.com

VWR, 1310 Goshen Parkway, West Chester, PA 19380, U.S.A. Tel: 800-932-5000. www.vwr.com

Sigma-Aldrich Chemical Co., P.O. Box 2060, Milwaukee, WI 53201, U.S.A. Tel: 800-558-9160. www.sigmaaldrich.com

ULTRA Scientific, 250 Smith Street, N. Kingstown, RI 02852. U.S.A. Tel : 800-338-1754. www.ultrasci.com

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Determination of Total Phosphorus in Wastewater Using Caro's Reagent and Ion Chromatography

INTRODUCTION

Phosphorus in the environment is beneficial for many biological processes, but too much phosphorus can create an imbalance in the ecosystem. Human activity can increase the concentration of phosphorus in the environment in many ways, particularly with the use of phosphorus-containing fertilizers and laundry detergents. Agricultural runoff is a major cause of increased phosphorus in both natural and manmade waterways. Phosphate stimulates the growth of plankton and aquatic plants, favoring some fast-growing species over others, which may have important roles in the ecosystem. Excess growth of fast-growing plants consumes large amounts of dissolved oxygen, which can suffocate fish. Excessive plant growth also blocks sunlight for bottom-dwelling species, compromising their health and prevalence, another unwanted change in the ecosystem. Monitoring the concentration of phosphorus in the environment is a good indicator because higher than normal concentrations lead to environmental problems.

Converting organic and inorganic phosphorus to orthophosphate allows phosphorus to be determined by ion chromatography (IC) with suppressed conductivity detection. Masson et al. demonstrated that soluble phosphate can be determined in soil water extracts in the µg/L concentration range and with higher accuracy than by a colorometric method.[1] In their sample analysis, Masson et al. used water to extract inorganic phosphate from the soil sample.

In the study presented here, the authors treat the wastewater sample with potassium peroxymonosulfate (Caro's reagent) and heat to convert all the phosphorus to orthophosphate. The orthophosphate is determined by ion chromatography (IC). The authors describe two IC methods to determine phosphate after treating a wastewater sample with Caro's reagent. One IC method uses a classic carbonate/bicarbonate eluent system with a column designed to determine the common inorganic anions with a carbonate/bicarbonate eluent. The second method uses a hydroxide eluent prepared by an eluent generator with a column designed to determine the common inorganic anions with a hydroxide eluent. Further, the second method uses a Reagent-Free™ IC (RFIC™) system and delivers all its benefits, including excellent retention time reproducibility for accurate peak identification, and time and labor savings by eliminating eluent preparation and associated errors.

Here, two IC methods are presented for the determination of phosphorus content in wastewater. The sample's organic and inorganic phosphorus are converted to orthophosphate by treatment with Caro's reagent before analysis by IC.

EQUIPMENT

Dionex ICS-3000 system including*:

DP Dual Pump

DC Detector/Chromatography module with dual temperature zone equipped with 6-port injection valve

EG Eluent Generator module

AS Autosampler

Peristaltic Pump with CRD 300 (4 mm) (P/N 064905)

Chromeleon® Chromatography Data System software Version 6.80 SR7

*For analysis using Condition A (see below), any Dionex IC system can be used. Condition B requires the use of a Dionex RFIC system.

REAGENTS AND STANDARDS

Deionized water (DI), Type I reagent-grade, 18 MΩ-cm resistivity or better

Sodium carbonate (Na_2CO_3, Ajax)

Sodium hydrogen carbonate ($NaHCO_3$, Ajax)

Sodium fluoride (NaF, Fluka)

Sodium chloride (NaCl, Fluka)

Sodium nitrite ($NaNO_2$, Fluka)

Sodium bromide (NaBr, Fluka)

Sodium nitrate ($NaNO_3$, Fluka)

Sodium sulfate (Na_2SO_4, Fluka)

Potassium dihydrogen orthophosphate (KH_2PO_4, Fluka)

8 mol/L Potassium hydroxide solution, 8 N (KOH, KANTO)

400 g/L Sodium hydroxide solution (NaOH, KANTO)

OXONE®, monopersulfate compound, Sigma-Aldrich (P/N 228036)

AS22 Sodium carbonate/bicarbonate concentrate (Dionex, P/N 063965)

PREPARATION OF SOLUTIONS AND REAGENTS

Eluent Solution

Condition A

Eluent (4.5 mM Na_2CO_3/1.4 mM $NaHCO_3$)

Dilute 10 mL AS22 sodium carbonate/bicarbonate concentrate to 1 L in a 1 L volumetric flask with DI water and mix.

CRD-300 Regenerant (200 mM NaOH)

Dilute 20 mL of 400 g/L sodium hydroxide solution to 1 L in a 1 L volumetric flask with DI water and mix.

Condition B

The eluent generator (EG) produces the eluent using the EluGen EGC II KOH cartridge and deionized water (18 MΩ-cm resistivity or better) supplied by the pump. The eluent concentration is controlled by Chromeleon software. The EluGen cartridge requires at least 14 MPa (2000 psi) of system backpressure, which ensures optimal removal of electrolysis gas from the eluent produced by the cartridge. See the *ICS-3000 Ion Chromatography System Operator's Manual* (Dionex Document No. 065031-04) for instructions on adding backpressure.

Caro's Reagent

Dissolve 0.5 g potassium monopersulfate triple salt ($2KHSO_4*KHSO_4*K_2SO_4$) in a 100 mL volumetric flask with DI water and dilute to volume.

Standard Solutions

1000 mg/L Stock Standard Solution

Dissolve accurately weighed salts (Table 1) in separate 100 mL volumetric flasks with DI water and dilute to volume.

Table 1. Amounts of Compounds Used to Prepare 100 mL of 1000 mg/L Standards

Ion	Compound	Weight (g)
Fluoride	Sodium fluoride (NaF)	0.221
Chloride	Sodium chloride (NaCl)	0.165
Nitrite	Sodium nitrite ($NaNO_2$)	0.150
Bromide	Sodium bromide (NaBr)	0.129
Nitrate	Sodium nitrate ($NaNO_3$)	0.137
Sulfate	Sodium sulfate (Na_2SO_4)	0.148
Phosphate*	Potassium dihydrogen orthophosphate (KH_2PO_4)	0.430

*The solution is 1000 mg/L phosphorus

10 mg/L Mixed Anion Stock Standard Solution

Pipette 1 mL each of 1000 mg/L fluoride, chloride, nitrite, bromide, nitrate, sulfate, and phosphorus standards into a 100 mL volumetric flask and bring to volume with DI water.

Working Standard Solutions for Condition A

The working standard solutions for analysis by Condition A are prepared as follows. Pipette the listed volume of 10 mg/L anion stock standard solution into 50 mL beakers (Table 2). Add 20 mL DI water to each beaker. Pipette 2 mL of Caro's reagent into each beaker. Boil the standard solutions using a hot plate for 30 min and then let them cool to room temperature. Transfer these standard solutions into 50 mL volumetric flasks and dilute to volume with Condition A eluent.

Working Standard Solutions for Condition B

The working standard solutions for analysis by condition B are prepared without adding Caro's reagent. Pipette the listed volumes of 10 mg/L anion stock standard solution into 50 mL volumetric flasks (Table 2). Dilute to volume with DI water.

Table 2. Volumes of 10 mg/L Stock Anion Standard Solution for Condition A and Condition B Working Standards

Concentration of Phosphorus (µg/L)	Volume of 10 mg/L Mixed Anion Stock Standard Solution (mL)	Volume of Caro's Reagent (only for Condition A) (mL)	Final Volume (mL)
20	0.1	2	50
40	0.2	2	50
80	0.4	2	50
160	0.8	2	50
320	1.6	2	50

SAMPLE PREPARATION

Filter the wastewater sample with qualitative 2, 110 mm filter paper (Whatman, Catalog No. 1002 110).

Sample Preparation for Condition A

Accurately pipette 20 mL filtered wastewater sample into a 50 mL glass beaker. Add 2 mL Caro's reagent and mix thoroughly. Cover the beaker with a watch glass. Boil the sample by using a hot plate for 30 min and then let it cool to room temperature. Transfer this sample into a 50 mL volumetric flask and dilute to volume with Condition A eluent.

Sample Preparation for Condition B

Prepare as for Condition A but after transferring the sample to a 50 mL volumetric flask, add 5 µL 8 N potassium hydroxide and bring to volume with DI water.

CHROMATOGRAPHIC CONDITIONS

Condition A

Column:	IonPac® AS22 (4 × 250 mm) (P/N 064141)
Guard:	IonPac AG22 (4 × 50 mm) (P/N 064139)
Eluent:	4.5 mM Na_2CO_3/ 1.4 mM of $NaHCO_3$
Flow Rate:	1.2 mL/min
Inj. Volume:	20 µL
Column Oven:	35 °C
Pressure:	~1800 psi
Detection:	Suppressed conductivity ASRS® 300, 4 mm (P/N 064554), recycle mode CRD 300, 4 mm (P/N 064637), External chemical mode (200 mM NaOH)
Suppressor Current:	45 mA

Condition B

Column:	IonPac AS18 (4 × 250 mm) (P/N 060549)
Guard:	IonPac AG18 (4 × 50 mm) (P/N 060511)
Eluent Source:	KOH produced by an EG equipped with a EGC II KOH cartridge (P/N 058900) and CR-ATC (P/N 060477)
Gradient Steps:	15 mM from –7 to 8 min and 15 to 45 mM from 8 to 20 min
Flow Rate:	1.2 mL/min
Inj. Volume:	20 µL
Column Oven:	5 °C
Pressure:	~2200 psi
Detection:	Suppressed conductivity ASRS 300, 4 mm (P/N 064554), external water mode
Suppressor Current:	170 mA

RESULTS AND DISCUSSION

Chromatography

Two sets of chromatographic conditions were used in this application. The first set of conditions used a carbonate/bicarbonate eluent. The eluent was prepared manually. This first set of chromatographic conditions was referred to as Condition A. The second set used a hydroxide eluent. The eluent was automatically generated by the eluent generator module using deionized water and an eluent generator cartridge. This second set of chromatographic conditions was referred to as Condition B. This application note presents a comparison of Conditions A and B. Condition B was a little more sensitive than Condition A, as shown by the Caro's reagent analysis. Caro's reagent was prepared and the analysis performed at the same time using both conditions (Figures 1 and 2). Condition B easily detected phosphate in Caro's reagent (Figure 2) while Condition A did not (Figure 1). Phosphate was observed by Condition A at about 10 min but it was difficult to integrate.

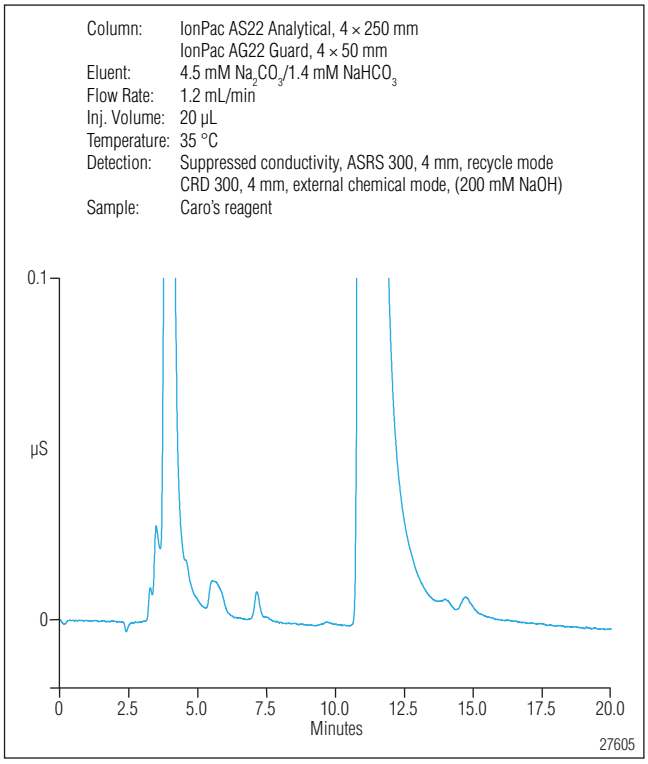

Figure 1. Chromatogram of Caro's reagent (Condition A).

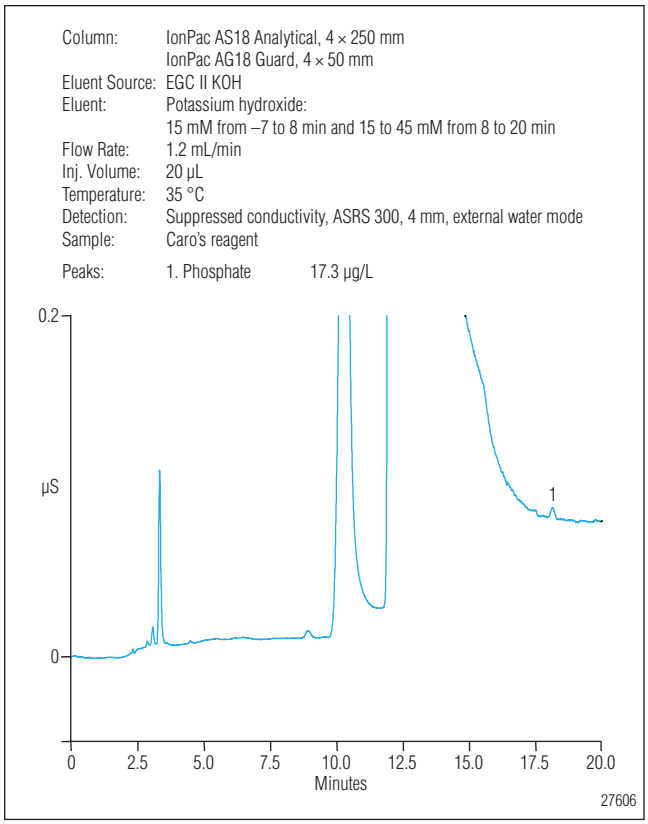

Figure 2. Chromatogram of Caro's reagent (Condition B).

Method Calibrations

Figures 3 and 4 show the chromatography of the calibration standards by Conditions A and B. Phosphate was present in Caro's reagent and difficult to detect by Condition A. To correct the measurement of phosphate by Condition A, the same amount of Caro's reagent was added to the calibration standard solutions. The calibration standards for Condition B were prepared in DI water and the method blank, in which phosphate was detected, was subtracted from the result of sample analysis. The concentrations of the phosphorus standard solutions were the same for both sets of conditions. Table 2 shows the concentrations of the calibration standards for both sets of conditions. Table 3 shows that the calibration is linear in the range tested for both sets of conditions.

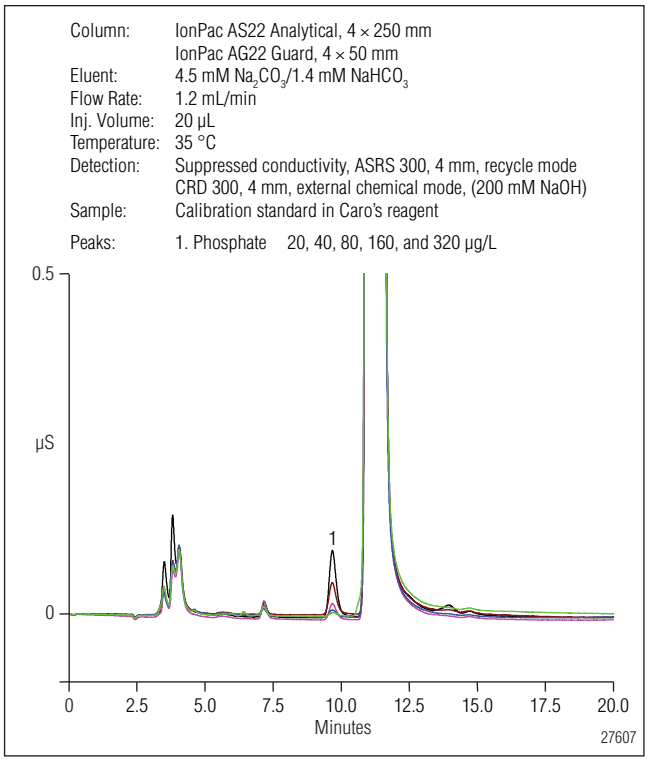

Figure 3. Overlay of chromatograms of five concentrations of phosphate standard for method calibration (Condition A).

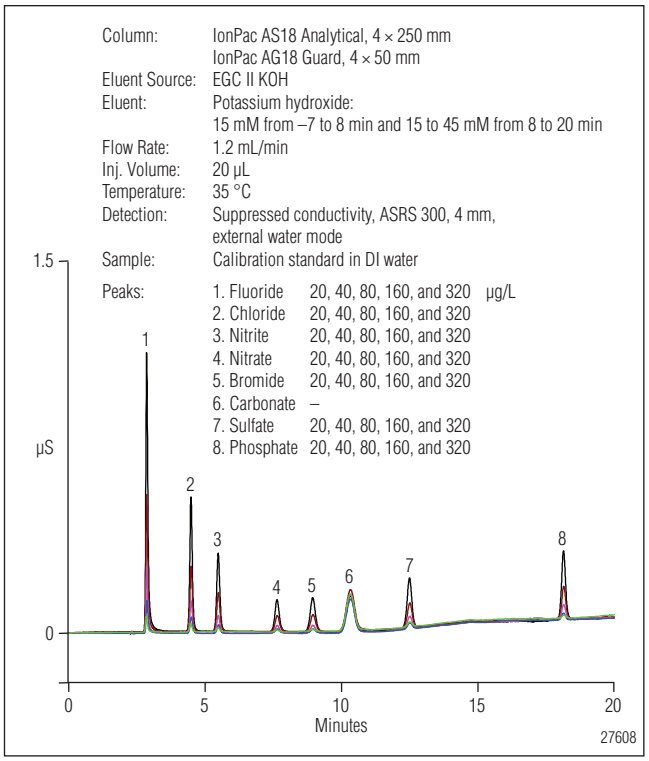

Figure 4. Overlay of chromatograms of five concentrations of the mixed anion standard, including phosphate, for method calibration (Condition B).

Method Detection Limits

As noted earlier, phosphorus was found in Caro's reagent, but it was not integrated when Condition A was used. The method detection limit (MDL) for Condition A was estimated from seven sample injections by using the standard deviation of found phosphorus and the Student's *t* value for the 99% confidence level (Table 4, column 3). This calculation estimated a value of 5.63 µg/L, which was lower than the amount of phosphorus in Caro's reagent determined by Condition B.

Table 3. Calibration Results

Chromatographic Condition	Name	% r^2	Offset (µS*min)	Slope (µS*min) / µg/L
A	Phosphorus	99.9659	–0.0001	0.0001
B	Phosphorus	99.8822	0.0008	0.0001

Table 4. Found Concentration of Phosphorus in Caro's Reagent, Sample, and Spiked Sample

Injection No.	Found Concentration by Condition A (µg/L)			Found Concentration by Condition B (µg/L)		
	Caro's Reagent	Sample	Spiked Sample	Caro's Reagent	Sample	Spiked Sample
1	—	199	252	18.5	220	272
2	—	204	253	15.4	223	272
3	—	202	250	16.7	226	278
4	—	202	253	17.0	221	273
5	—	200	252	19.2	219	275
6	—	200	253	16.9	224	274
7	—	204	250	17.3	223	273
Average	—	202	252	17.3	222	274
%RSD	—	0.89	0.54	7.04	1.09	0.73

An analysis of Caro's reagent by Condition B found 17.3 µg/L phosphorus, which was not reliably determined by Condition A. The method detection limit for Condition B was estimated from seven injections of the 10 µg/L mixed anion standard that contained phosphate. Chromatography of the 10 µg/L of mixed anion standard is shown in Figure 5. This analysis estimated a MDL of 2.39 µg/L. For either set of chromatographic conditions, the amount of phosphorus in Caro's reagent set a practical limit on the MDL for this analysis. Samples submitted for this analysis were also likely to have phosphorus concentrations well above the MDL for either set of chromatographic conditions, thus making both suitable for this application.

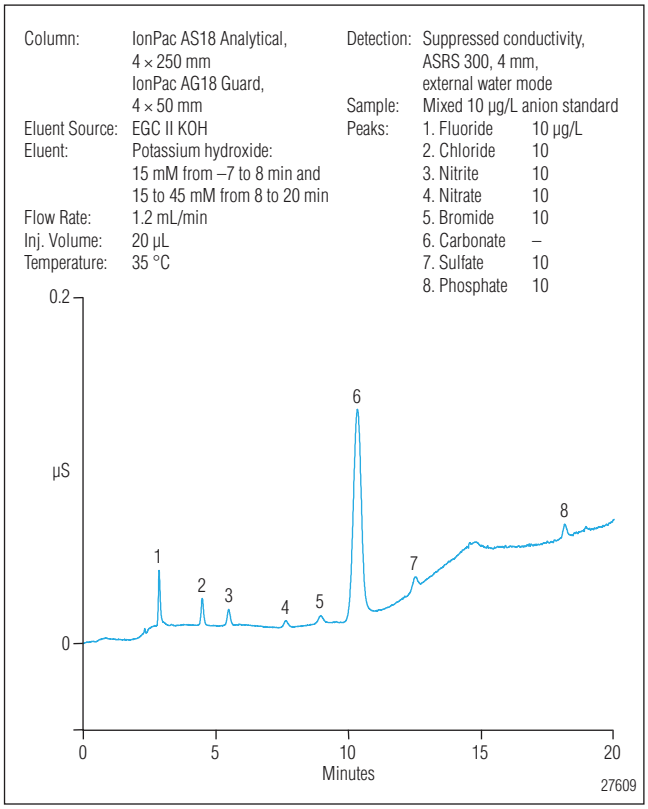

Figure 5. Chromatogram of a 10 µg/L mixed anion standard in DI water (Condition B).

Sample Analysis

A wastewater sample was collected in Bangkok, Thailand. To compare the total phosphorus measurement using Conditions A and B, the same wastewater sample was prepared by following the sample preparations for Conditions A and B as described in the Sample Preparation section. Seven injections were made for both sample preparations and a method blank for Condition B. The average concentrations of phosphorus in the wastewater determined by Conditions A and B were 202 µg/L and 222 µg/L, respectively. The average amount of phosphorus in the method blank (Condition B) was 17.3 µg/L. Subtracting the method blank for Condition B yielded a value of 205 µg/L, which was very close to that determined by Condition A. Table 4 shows the data from the phosphate determination of the wastewater sample using both analysis conditions; Figures 6 and 7 show the chromatography.

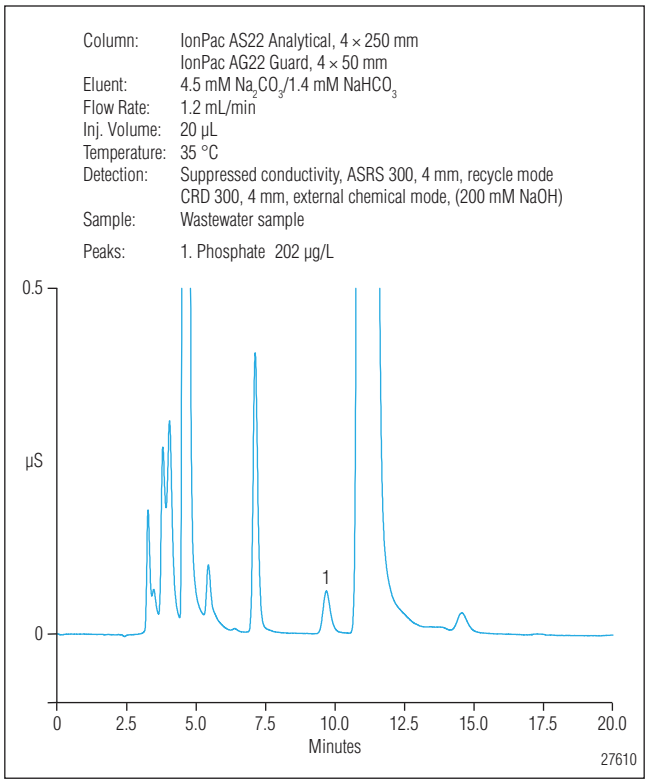

Figure 6. Chromatogram of wastewater sample (Condition A).

Table 5. Recovery and Corrected Sample Concentration Results							
Analyte	Condition	Spiked Concentration	Average Found Concentration (µg/L)			Recovery (%)	Corrected Sample Concentration (µg/L)
			Caro's Reagent	Sample	Spiked Sample		
Phosphorus	A	50	—	202	252	100.05	202
	B	50	17.3	222	274	100.54	205

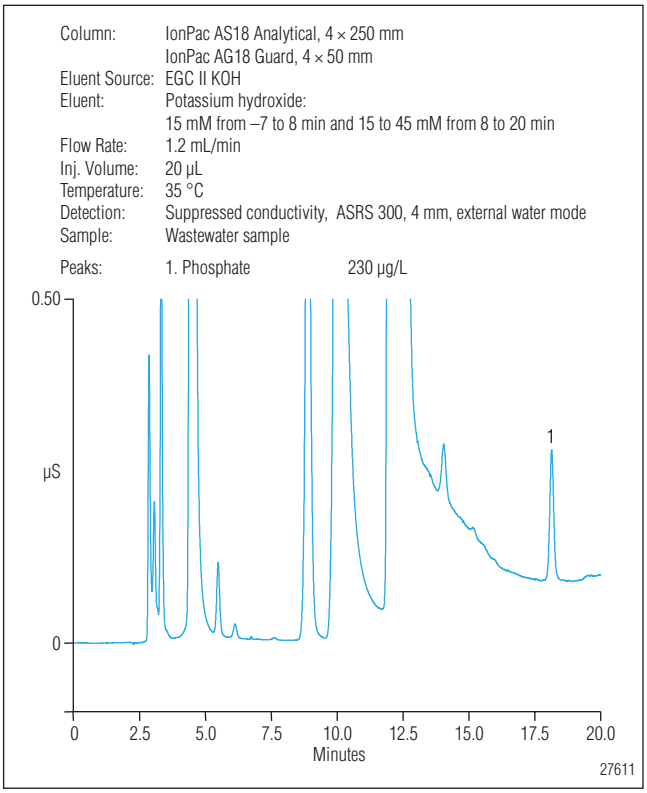

Figure 7. Chromatogram of wastewater sample (Condition B).

To evaluate the recovery of both conditions, the same wastewater sample was spiked with the 10 mg/L mixed anions stock standard solution before the sample preparation. This yielded 50 µg/L of spiked phosphorus after sample preparation. Both sets of conditions exhibited excellent recovery, with values of 100.05% and 100.54% for Conditions A and B, respectively. Tables 4 and 5 show the recovery data.

SUMMARY

Both IC methods presented in this application note can be used to make an accurate measurement of the total phosphorus content of a wastewater sample after treatment with Caro's reagent. The analyst has a choice of a method with simple eluent preparation or a method that requires no eluent preparation and has the high reproducibility of a RFIC system.

REFERENCE

1. 1. Masson, P.; Morel, C.; Martin, E.; Oberson, A.; Friesen, D. *Commun. Soil Sci. Plant Anal.* **2001,** *32,* 2241–2253.

Determination of Total Phosphorus Using Two-Dimensional Ion Chromatography

INTRODUCTION

The determination of phosphorus is prescribed by the U.S. EPA (methods 365.1–365.5). These colorimetric methods use an autoanalyzer or spectrophotometer. Total phosphorus determination is described as a subset of these methods and includes oxidative decomposition of organic phosphates in the sample by heating with persulfate (methods 365.1–365.3, 365.5) or acidic mercuric sulfate (method 365.4) to transform all the phosphates to orthophosphate. The method depends on a color reaction between orthophosphate, ammonium molybdate, and antimony potassium tartrate to form an antimony-phosphomolybdate complex. This complex is subsequently reduced by ascorbic acid to form a blue-colored complex, whose concentration is determined by visible absorption spectroscopy.

Ion chromatography (IC) with suppressed conductivity detection can be used to determine phosphate in the presence of other ions. IC offers a valuable secondary analysis method for total phosphate and serves as a confirmatory technique. IC is preferrable to colorimetry due to the elimination of a very toxic reagent, antimony potassium tartrate. However, the presence of a large amount of sulfate from the persulfate digestion and the inherent complexity of some samples (for example, soil and wastewater) makes IC difficult to perform at the required detection limit of 10 µg/L with a direct injection of the sample and single-dimensional analysis.

A simple, two-dimensional analytical technique is presented for the analysis of total phosphorus. A sample is injected onto an IonPac® AS11-HC column. The phosphate-containing fraction of the separated sample is then directed to an anion concentrator column where the phosphate and other anions in that fraction are trapped. The majority of the sample goes directly to waste while the phosphate and a smaller amount of interfering anions are collected on the concentrator column. The concentrator column is then placed back in line with the original separator column and the trapped anions it contains are reseparated on the column. This process removes the majority of the matrix ions. The sensitivity and accuracy of the analysis is improved since the phosphate analyte now forms a sharper peak with better signal-to-noise ratio and better separation from matrix interferences.

EXPERIMENTAL

Instrumentation

Dionex ICS-3000 Reagent-Free™ Ion Chromatography system with Eluent Generation (RFIC-EG™ system) consisting of:

SP Single pump (P/N 061706)

EG Eluent generator module (P/N 061714)

DC Detector compartment module with conductivity detector (P/N 063772 and P/N 061716)

Automation Manager with 10-port valve (P/N 061734 and P/N 061962)

TAC-ULP1 (5 × 23 mm) concentrator column (P/N 061400)

Conditions

Columns:	IonPac AG11-HC, 4 mm (P/N 052962) and IonPac AS11-HC, 4 mm (P/N 052960)
Eluent:	Electrolytically generated KOH gradient; EGC (P/N 058900), 1 mL/min; 20 mM (0 to 15 min), 40 mM (20 min), 20 mM (20.1 min)
Inj. Volume:	250 µL
Temperature:	30 °C (column compartment) 30 °C (detector compartment) 35 °C (conductivity detector)
Detection:	Suppressed conductivity, ASRS® 300 suppressor (4 mm) (P/N 064554), 100 mA

This application can also be conveniently performed on an ICS-2100 system (P/N 069576) equipped with an auxiliary 10-port valve (P/N 069473).

RESULTS AND DISCUSSION

Water samples were prepared by heating in a laboratory microwave oven with persulfate. Samples were injected onto an IonPac AS11-HC column, which was selected because it exhibits the high capacity needed for this highly concentrated sample. The initial separation is carried out isocratically with 20 mM potassium hydroxide to retain the phosphate on the column and elute most of the matrix ions away from the phosphate. A full-scale chromatogram (Figure 1) shows that the phosphate peak, approximately 25 nS tall, is not observable on this scale. The amount of phosphate in the sample is approximately 1 part per million of the total anion content, with the vast majority of the remainder being sulfate. Figure 2 shows the expanded scale chromatogram, where the phosphate peak is now observable and elutes at about 13.4 min on the tail of the huge sulfate peak. The phosphate peak is also broad, with a peak width at half

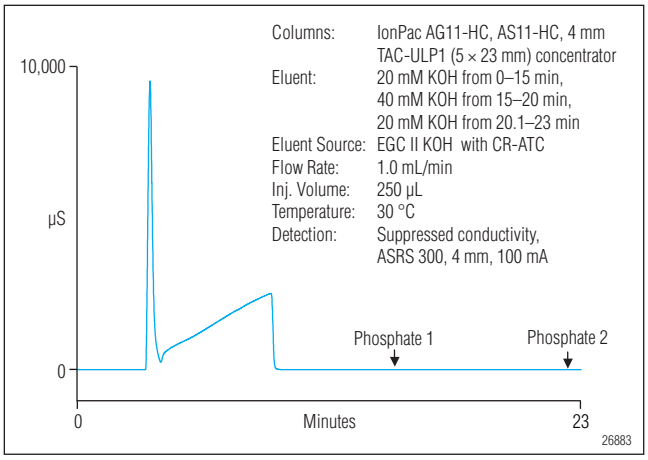

Figure 1. Full-scale chromatogram of digested wastewater showing approximate retention times of the two phosphate peaks.

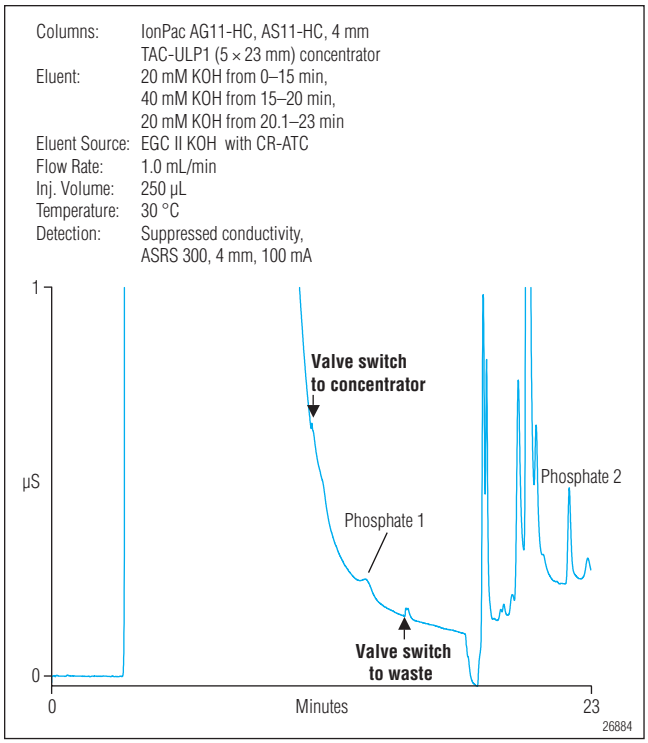

Figure 2. Expanded-scale chromatogram of wastewater digest.

height of 0.4 min. If the analysis were carried out using only this separation, quantitative analysis of the phosphate peak on the tail of the sulfate peak would result in lower analytical sensitivity due to the problems of detecting and integrating a small peak on the tail of a very large peak.

The auxiliary valve was switched to collect column effluent onto the concentrator column during the retention time period of 11 to 15 min, as noted in Figures 2 and 3. This traps the phosphate peak and a relatively small amount of other anions, and directs them back to the concentrator column. It is important to note that the sample has passed through the suppressor before being

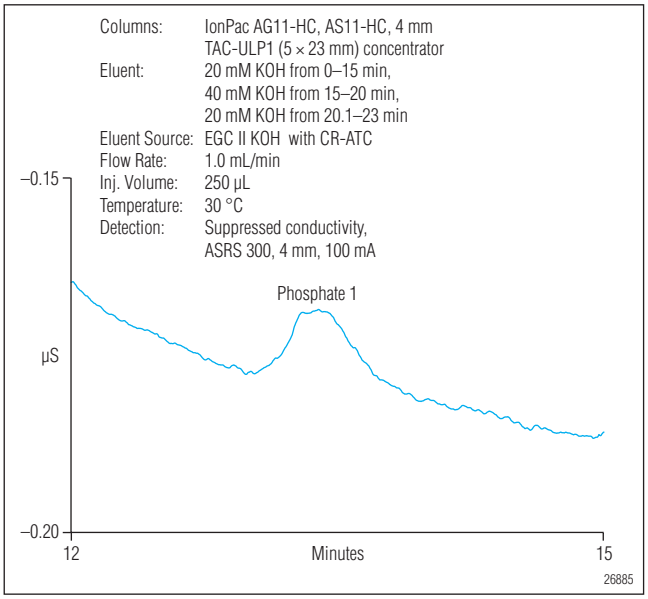

Figure 3. First phosphate peak, 10 µg/L standard.

collected on the concentrator. The suppressor converts the hydroxide eluent to water, thus enabling the concentrator column to capture the anions in the sample in a narrow band, as there is no eluent to disperse them off the concentrator column.

At 15 min retention time, the auxiliary valve was switched back to its original configuration, which now positions the concentrator column before the analytical column (Figure 4.) At this point, an elution gradient is also started. Gradient elution has the effect of eluting the trapped anions from the concentrator column in a narrow band. The increasing ionic strength also reduces the elution time of the phosphate during this second pass to half of what it was on the first pass through the column.

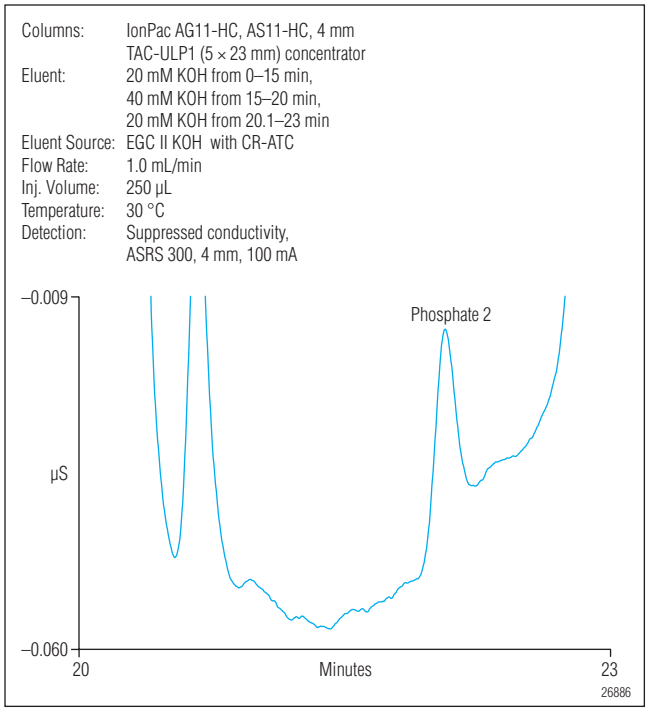

Figure 4. Second phosphate peak, 10 µg/L, same chromatographic run as Figure 3. Note improved peak height and peak shape and resulting improvement in signal-to-noise ratio.

These effects together produce a second phosphate peak, which is now isolated from contaminants and is about one-third narrower than the original peak. This sharper peak results in an improvement of signal-to-noise by about three-fold.

A calibration curve from 10 to 30 µg/L is seen in Figure 5. Plumbing diagrams showing the various valve

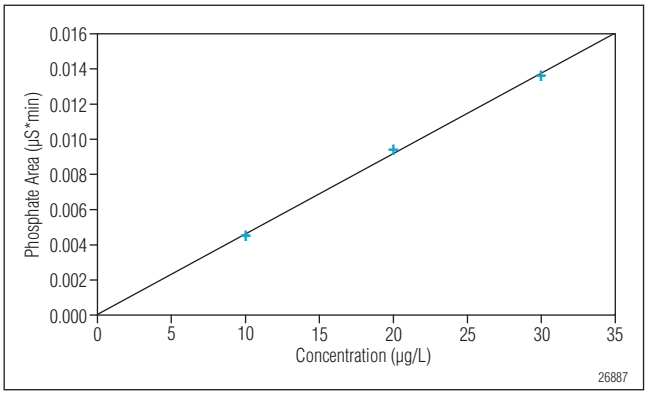

Figure 5. Calibration curve (10 µg/L to 30 µg/L) using second phosphate peak.

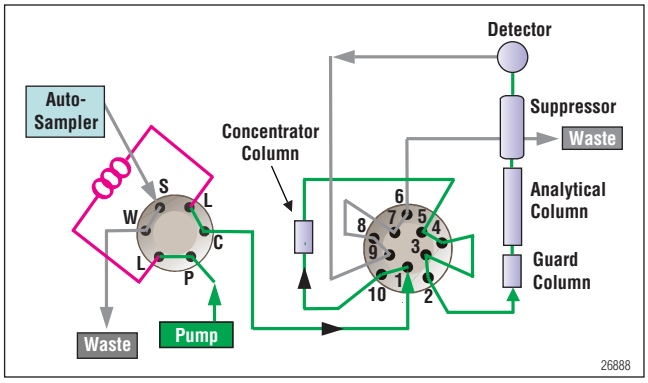

Figure 6. Inject sample.

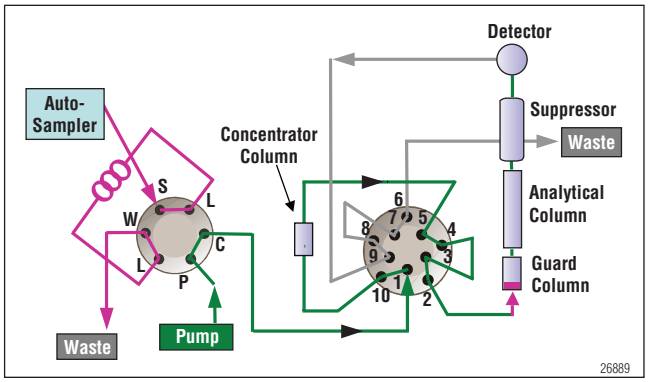

Figure 7. Analyze sample.

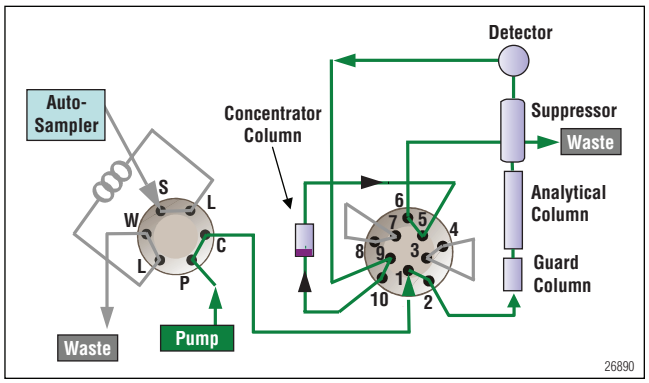

Figure 8. Collect phosphate-containing band on concentrator column.

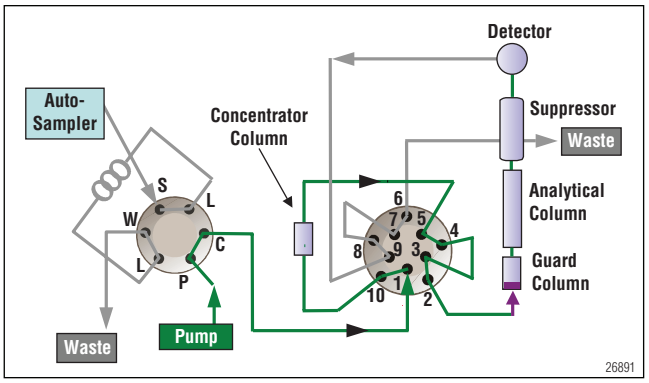

Figure 9. Reanalyze phosphate heart-cut with gradient elution.

positions and flows at different stages in the analysis are seen in Figures 6–9.

CONCLUSION

In this application note, a two-dimensional separation technique to isolate phosphate from a huge excess of sulfate is demonstrated. Compared to the first phosphate peak, the second peak is approximately three times taller than the peak resulting from the first pass through the column, thus improving signal-to-noise ratio and detectability by a factor of three (Figures 3 and 4). The technique depends on the flexibility of the application of an auxiliary high-pressure switching valve, along with a concentrator column, to recycle trapped analyte for a second separation run on the analytical column.

This approach provides improved quantitation of the phosphate using only one column. The use of a single column eliminates the need for a second analytical pump and thus, reduces instrument complexity and cost. Integration of the auxiliary sample preparation valve in the ion chromatograph also helps reduce the complexity of software control and hardware management.

Determination of Common Anions and Organic Acids Using Ion Chromatography-Mass Spectrometry

INTRODUCTION

Mass spectrometry (MS) as a detector for ion chromatography (IC) has gained popularity recently due to the increasing demand for sensitivity, selectivity, confirmation of identity, and structural interpretation.[1-8] Compared to other commercially available MS detectors, the MSQ Plus™ detector offers substantially improved performance for low-molecular weight analytes, which covers most analytes for small-molecule IC applications.

This application note (AN) demonstrates the IC-MS method using the MSQ Plus detector for the determination of five common anions and selected organic acids. Method performance with respect to calibration range, reproducibility, and method detection limits are also presented.

EXPERIMENTAL

Instrumentation

System:	Dionex ICS-2000 RFIC™ system
Columns:	IonPac® AS20 (2.1 × 250 mm) with AG20 (2.1 × 50 mm) CR-ATC Continuously Regenerated Anion-Trap Column
Mobile Phase:	28 mM hydroxide generated from EGC II KOH cartridge
Flow Rate:	0.25 mL/min
Injection Volume:	5 µL
Detection:	Suppressed conductivity (external water at 0.50 mL/min) MSQ Plus single quadrupole mass spectrometer
Ionization Interface:	Electrospray ionization (ESI)
Desolvation Solvent:	0.22 mL/min acetonitrile delivered by an AXP-MS auxiliary pump
MS Detection Mode:	Negative Selected Ion Monitoring (SIM) Scan details shown in Table 1.
Needle Voltage:	1.5 kV
Nebulizer Gas:	Nitrogen at 85 psi
Probe Temperature:	450 ºC

Software

Instrument control, data acquisition, processing, and report generation were accomplished through the Chromeleon® Chromatography Data System (version 6.8 SR6).

Reagents and Standards

All stock standard solutions for individual analytes were prepared in deionized water (D.I. water, 18.2 MΩ-cm^{-1}, Millipore). Fluoride and nitrate standards were prepared by diluting standard solutions obtained from Ultra Scientific (P/N ICC-003) and Dionex (P/N 060254) respectively. Chloride, sulfate, and phosphate standards were prepared by dissolving potassium salts in D.I. water (pure chemicals obtained from Mallinckrodt and Fisher). Standard solutions of organic acids were prepared by dissolving each chemical (in organic or salt form) in D.I. water: pyruvic acid, sodium salt (Sigma, P2256), α-ketoglutaric acid, monopotassium salt (Sigma, K2000), and tartaric acid (Aldrich, T400). Methanol (CH_3OH) and acetonitrile (CH_3CN) were purchased from Burdick & Jackson (HPLC/UV grade, Honeywell, Muskegon, MI).

The mixed standard solution, including each of the listed analytes at 5.0 ppm, was prepared by diluting each of the stock standard solutions in D.I. water. Calibration standards were prepared by series dilution to 1000 ppb, 500 ppb, 200 ppb, 100 ppb, 50 ppb, 20 ppb, 10 ppb, 5 ppb, and 2 ppb.

RESULTS AND DISCUSSION

The optimization of the acquisition parameters of the MSQ Plus spectrometer for IC analysis of common anions and selected organic acids is discussed in detail elsewhere.[9] MSQ parameters, including probe temperature, nebulizer gas flow, assistant makeup flow rate, needle voltage, and cone voltage were optimized using Response Surface Methodology (RSM) with Central Composite Design (CCD). Compared to the one-parameter-at-a-time approach for optimization, RSM with CCD includes the effects from interactions between parameters (ignored by a single parameter approach) and also minimizes the numbers of experiments required to reach the optimum system performance. The optimum conditions determined for the MSQ Plus spectrometer in these experiments are listed in the instrumentation section and the analyte individualized cone voltages for SIM scans are listed in Table 1.

Table 1. MS SIM Scans*

Analyte	Adduct	Observed (*m/z*)	Cone Voltage (V)
Fluoride	$[F(HF)]^-$	39.1	88
Chloride	$[Cl]^-$	35.1	26
Nitrate	$[NO_3]^-$	62.1	73
Sulfate	$[HSO_4]^-$	97.1	45
Phosphate	$[H_2PO_4]^-$	97.1	69
Pyruvate	$[CH_3COCOO]^-$	87.1	52
α-Ketoglutarate	$[HOOCCO(CH_2)_2COO]^-$	145.2	50
Tartrate	$[HOOC(CH_2OH)_2COO]^-$	149.2	37

* Use 0.5 span and 0.1 s dwell time for all SIM scans.

It is important to note that the SIM ion selected for fluoride is 39.1 *m/z,* which is associated with the fluoride adduct $[F(HF)]^-$. Representative conductivity and SIM chromatograms are shown in Figure 1 (1 ppm with 5 µL). Compared to nonspeciating conductivity detection, each analyte can be quantified very specifically using MS SIM detection, shown as one peak in SIM channel for each target analyte. The deprotonated sulfate $[HSO_4]^-$ and phosphate $[H_2PO_4]^-$ ions which have the same mass-to-charge ratio (*m/z*) at 97 are shown as two peaks in SIM channel 97, and can also be accurately quantified because they are chromatographically separated. The differentiation of sulfate from phosphate can be confirmed by the isotope peak 99.1 ($[H^{34}SO_4]^-$) at ~5% relative peak intensity of peak 97.

Calibration curves were generated using concentrations from low ppb to 1.0 ppm, and method detection limits (MDL) were calculated by $MDL = S \times t_{99\%,\ n=7}$ where S is the standard deviation and t is the Student's t at 99% confidence interval. Seven replicate injections of a calibration standard at 20 ppb were performed to calculate the MDL (100 ppb was used to calculate MDL for fluoride). Calibration range, coefficient of determination, and MDL values are summarized in Table 2. Figure 2 shows the calibration curve for tartrate from 2 ppb to 1000 ppb and the graphic insert shows the linear curve at the lower levels.

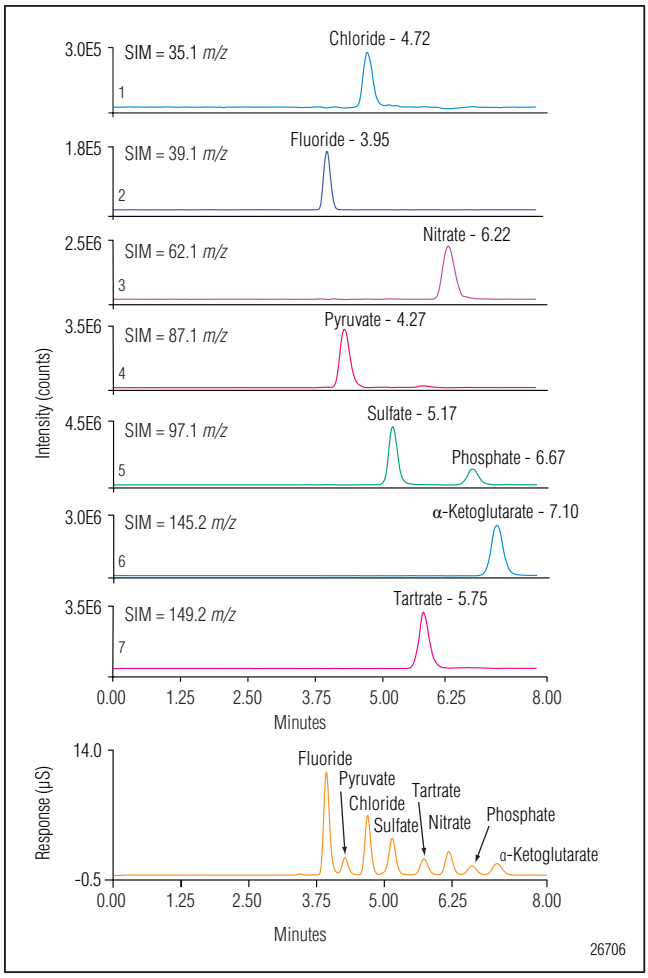

Figure 1. Conductivity and SIM chromatograms of five common anions and three selected organic acids.

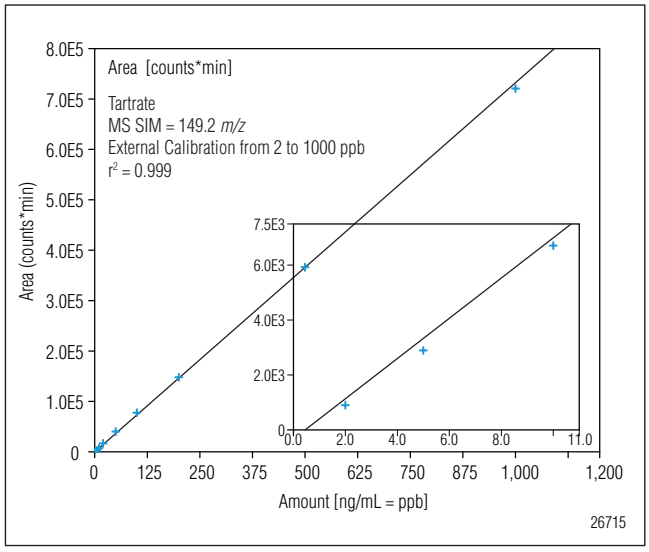

Figure 2. Calibration curve of tartrate at concentrations from 2 ppb to 1000 ppb.

Table 2. Calibration Range, Coefficient of Determination, and MDLs

Analyte	Calibration Range (ppb)		Calibration Type	Weighting	r^2	%RSD*	MDL (ppb)
Fluoride	100	1000	Linear	1/X	0.997	9.35	29.4
Chloride	2	1000	Cubic	1/X	0.980	5.16	3.2
Nitrate	2	1000	Cubic	1/X	0.992	4.05	2.5
Sulfate	2	1000	Linear	1/X	0.998	5.75	3.6
Phosphate	5	1000	Linear	1/X	1.000	7.78	4.9
Pyruvate	2	1000	Quadratic	1/X	0.999	5.06	3.2
α-Ketoglutarate	5	1000	Linear	1/X	0.998	4.98	3.1
Tartrate	2	1000	Linear	1/X	0.999	5.20	3.3

* %RSD was calculated from 7 replicate injections of a calibration standard at 20 ppb except for fluoride (100 ppb).

After calibration, this method was used for quantification of the target analytes in commercially bottled drinking water. A bottled water sample was injected directly without sample preparation. Figure 3 shows the overlay of SIM chromatograms. Four anions were detected in the bottled water sample: 713 ppb chloride, 496 ppb sulfate, 143 ppb nitrate, and 181 ppb phosphate.

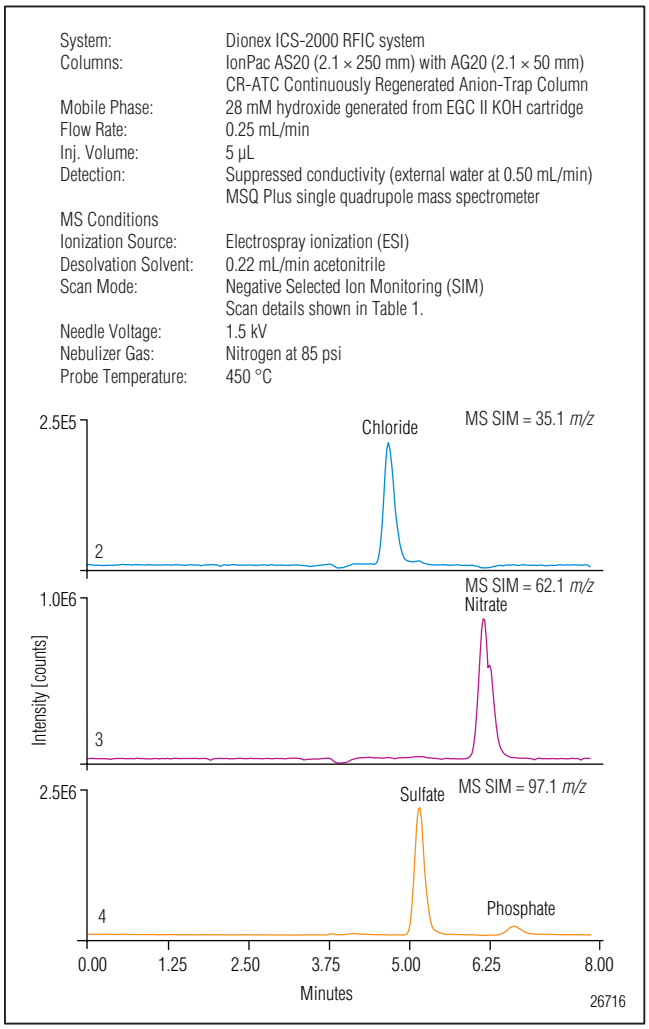

Figure 3. Anions in a bottled water sample.

CONCLUSION

This application note demonstrates the use of the IC-MS method for the determination of five common anions and three organic acids at parts per billion levels. With the use of very specific MS SIM detection, ppb level quantification can be performed with greater confidence on real world samples, such as bottled drinking water. The quantification results from MS were without prior sample preparation or employing preconcentration, which may be required with other detection methods.

Note that the optimum settings and responses may vary on different instruments; optimization of MSQ Plus source conditions and acquisition parameters is highly recommended for best results.

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Determination of Iodide in Seawater and Other Saline Matrices Using a Reagent-Free Ion Chromatography System with Suppressed Conductivity and UV Detections

INTRODUCTION

Iodine is an essential nutrient found in seawater, seafood, and iodine-enriched food, such as iodized table salt. The most common forms of iodine in diet are iodide and iodate, with additional iodo-organic compounds providing a small fraction of bioavailable iodine. Iodine deficiency affects thyroid hormone production and leads to developmental diseases, goiter, and paralysis.[1] Table salt is routinely iodized to prevent such diseases and promote public health. Iodization levels vary by country and range from 5–100 mg iodine per kg of salt.[2] In the United States, Canada, and most northern European countries, potassium iodide is frequently used to iodize salt. In tropical countries, potassium iodate is preferred due its greater stability under humid conditions.[3]

Because iodide is an essential source of iodine, there is need to determine iodide in a variety of matrices. Seawater is a large natural source of iodide, typically containing 50–60 µg/L iodide with a wide range of observed concentrations. For example, iodide amounts can range from less than 1 µg/L to greater than 60 µg/L, with the measured concentrations dependent on water depth, oxygen concentration, and the biological mediation of the iodide/iodate equilibrium.[4] Determination of iodide in seawater and other saline matrices by ion chromatography is challenging due to high sample chloride concentrations. In seawater, the matrix is further complicated by high concentrations of carbonate and sulfate. This high ionic strength matrix makes direct analysis of samples containing iodide difficult.

In this application note (AN), a RFIC-EG™ system with UV detection is used to determine iodide in seawater, synthetic sea salt, and iodized table salt. Iodide is separated from other matrix anions on the IonPac® AS20 column, which is a high-capacity column designed for the separation of polarizable anions. The unique selectivity of this column makes it ideal for separating hydrophobic anions in matrices with high concentrations of chloride, sulfate, and carbonate. The high capacity of the IonPac AS20 column allows the determination of iodide in saline matrices with no sample pretreatment. Furthermore, by detecting iodide by UV absorbance, other anions, such as chloride and carbonate, are not detected and do not interfere with iodide determination. This proposed method is further simplified by using a 2 mm column format and an in-line high-pressure hydroxide eluent generator requiring only the addition of deionized water for continuous operation. In this AN, the linearity, detection limits, precision, and recovery of iodide in saline matrices for the proposed method are demonstrated.

EQUIPMENT

Dionex ICS-3000 Reagent-Free™ Ion Chromatography (RFIC-EG) system consisting of:

SP Single Pump or DP Dual Pump module

EG Eluent Generator module

DC Detector/Chromatography module (single or dual temperature zone configuration)

AS Autosampler

ICS-3000 VWD UV-vis Absorbance Detector (P/N 064654, 4 wavelength or P/N 064377 single wavelength) with a PEEK™ semi-micro flow cell (PN 6074.0300)

EluGen® EGC II KOH cartridge (Dionex P/N 058900)

Continuously-Regenerated Anion Trap Column, CR-ATC II (Dionex P/N 060477)

Chromeleon® 6.8 Chromatography Data System

Polystyrene AutoSelect™ vials with caps and septa, 10 mL (Dionex P/N 055058)

Nalgene® 125 mL HDPE narrow mouth bottles (VWR P/N 16057-062)

Nalgene 250 mL HDPE narrow mouth bottles (VWR P/N 16057-109)

Nalgene 250 mL 0.2 µm nylon filter units (VWR P/N 28199-371)

Nalgene 1000 mL 0.2 µm nylon filter units (VWR P/N 28198-514)

REAGENTS AND STANDARDS

Deionized water, Type I reagent grade, 18 MΩ-cm resistivity or better

Sodium chloride (JT Baker P/N 4058-05)

Sodium sulfate (VWR, P/N EM-SX0760-1)

Potassium chloride (Mallinckrodt P/N 6858)

Sodium bicarbonate (VWR, P/N EM-SX0320-1)

Potassium bromide (JT Baker P/N 2998-01)

Sodium fluoride (Fisher P/N S-299)

Boric acid (JT Baker P/N 0084-01)

Potassium iodide (VWR, P/N VW5225-1)

CONDITIONS

Columns:	IonPac AG20, 2 × 50 mm IonPac AS20, 2 × 250 mm
Eluent:	13 mM KOH from 0–10 min, 13–45 mM KOH from 10–15 min, 45 mM KOH from 15–25 min*
Eluent Source:	EGC II KOH with CR-ATC
Flow Rate:	0.25 mL/min
Temperature:	30 °C (column & detector compartment)
Inj. Volume:	10 µL
Detection:	A) Suppressed conductivity, ASRS® 300 (2 mm), external water mode, 28 mA suppressor current B) UV, 223 nm
Background Conductance:	< 1 µS
Noise:	~0.5–1.0 nS (conductivity) ~0.05–0.10 mAU (UV)
System Backpressure:	~2600 psi

* The column equilibrates for 10 min at 13 mM KOH prior to injection.

PREPARATION OF SOLUTIONS AND REAGENTS

Eluent Solution

Generate the potassium hydroxide (KOH) eluent online by pumping high-quality, degassed, deionized water through the EGC II KOH cartridge. Chromeleon software will track the amount of KOH used and calculate the remaining lifetime. To minimize the baseline shift and background noise, manual eluent preparation is not recommended.

Stock Standard Solution

A stock solution of 1000 mg/L of potassium iodide (KI) was prepared by dissolving 131 mg in 100 mL (100.00 g) of DI water. The solution was stored in Nalgene HDPE bottles at < 6 °C.

Standard Solutions

Intermediate stock solutions of 1 mg/L were prepared gravimetrically by pipetting 0.100 mL (0.100 g) of a 1000 mg/L KI standard into a 125 mL HDPE bottle and diluting to a total volume of 100 mL (100.0 g). Calibration standards between 10 µg/L and 250 µg/L were prepared by appropriate dilution of the intermediate stock solution with DI water. The standard solutions were stored at <6 °C when not in use.

SAMPLE PREPARATION

Simulated Seawater

Simulated seawater was prepared by dissolving the salts listed in Table 1 into 1 L of DI water, following the method of Kester et al., with the exclusion of magnesium chloride, calcium chloride, and strontium chloride.[5] These salts were not used in the simulated seawater because they add only a small amount of additional chloride to the matrix and require preparation by drying, dissolution, and volumetric addition to the other salts. This yields a solution with a salinity of approximately 3.5%.

Table 1: Salts Added to Form Simulated Seawater (1 L)

Reagent	Amount added (mg)
Sodium Chloride	23900
Sodium Sulfate	400
Potassium Chloride	680
Sodium Bicarbonate	198
Potassium Bromide	95.4
Boric Acid	27.1
Sodium Fluoride	4.0

Synthetic Sea Salt

Commercially available synthetic sea salt was prepared by following package directions (1/2 cup of salt per gallon of deionized water) to prepare a solution of approximately 3.5% salinity. A 1 L portion was prepared with 30 g of aquarium salt. A sea salt density of approximately 2.2 g/cm^3 was used to convert the preparation directions to metric units.[6]

Fresh Iodized Table Salt

Solutions of table salt were initially prepared at a salinity level similar to seawater. These solutions contained iodide in amounts that exceeded the standard curve and were further diluted to generate solutions of approximately 100 ppb iodide. This was done by serial dilution as follows: Dissolve 1.92 g of table salt in 100 mL of deionized water. Then, dilute 6.0 mL (6.0 g) of this solution in 57.5 g of DI water.

Seawater Collected at Half Moon Bay and Pacifica, CA

Surface seawater was collected in a 250 mL HDPE Nalgene bottle that had been cleaned prior to sample collection. The sample was stored on ice until it could be filter-sterilized through a 250 mL, 0.2 µm nylon filter unit. After filtration the sample was stored at <6 °C.

A 75 µg/L iodide standard was filtered by the same method and analyzed to determine iodide loss during the filtration step. The iodide concentration determined in the filtered standard was equivalent to an unfiltered standard, within the error of the method.

Precautions

Samples should be prepared for analysis within 24 h to prevent loss of iodide by oxidation. Additionally, all solutions should be stored at <6 °C and the autosampler should be thermostatically controlled to <10 °C.

The external water for regenerating the suppressor should be set to a flow rate of 2–2.5 mL/min. The recycle mode of suppressor regeneration is not feasible for this method due to the high salt concentrations that can dramatically shorten the working lifetime of the ASRS and the CR-ATC.

The method performance will critically depend on the noise observed in the UV detector. To reduce UV noise, the ASRS performance must be optimized.This can monitored by the observed noise in the conductivity detection channel. We recommend that the flow rate of the external water be confirmed daily to ensure that the ASRS is consistently regenerated.

If conductivity detection is not used to analyze samples, the suppressor must still be in place to maintain an acceptable UV baseline. Without suppression, the hydroxide eluent will absorb in the UV and contribute to a high background, preventing detection of iodide.

Because these samples contain high concentrations of anions, carryover was observed when the syringe flush volume was less than 1000 µL. A syringe flush volume of 1000 µL or greater is strongly recommended between samples.

RESULTS AND DISCUSSION

In preliminary experiments, the IonPac AS16, AS20, and AS24 columns were evaluated for the determination of iodide in seawater; however, the AS20 column was found to be superior for this application. This study found that while the capacity of the AS24 column is nearly double that of the AS20 and more than three times that of the A16, the retention time of iodide was 10 min longer on the AS24 compared to the other columns. Compared to the AS16 column, the AS20 column has a higher capacity and better resolution between chloride, carbonate, sulfate, and iodide. The AS20 column provides a good compromise for shorter retention times, good sensitivity, and high capacity that allow the separation of iodide in a saline matrix without dilution.

Figure 1 shows the separation of an iodide standard spiked in simulated seawater. The high concentrations of chloride, sulfate, and carbonate in this matrix make quantification of iodide difficult by conductivity detection. As shown, the iodide peak is obscured by the carbonate and sulfate peaks (Figure 1A). Iodide absorbs in the UV at 223 nm while the other anions at high concentrations remain undetected. Iodide can easily be detected by UV absorbance without interference from sulfate or carbonate, as shown in Figure 1B.

Linear range, limit of quantitation, limit of detection

To determine the linearity of the method, iodide calibration standards in deionized water were injected in triplicate, covering the average concentration range of iodide in seawater. Both detection methods showed similar linearity; 0.9967 and 0.9962 for conductivity and UV detection, respectively. The LOD and LOQ were confirmed by injections of iodide standards prepared at concentrations estimated to give peak heights that are 3 times and 10 times the noise, respectively. Table 2 summarizes the linearity, LOD, LOQ, retention time, and peak area precisions. The observed values will vary depending on the performance of the suppressor which affects the background and noise observed in the UV detector.

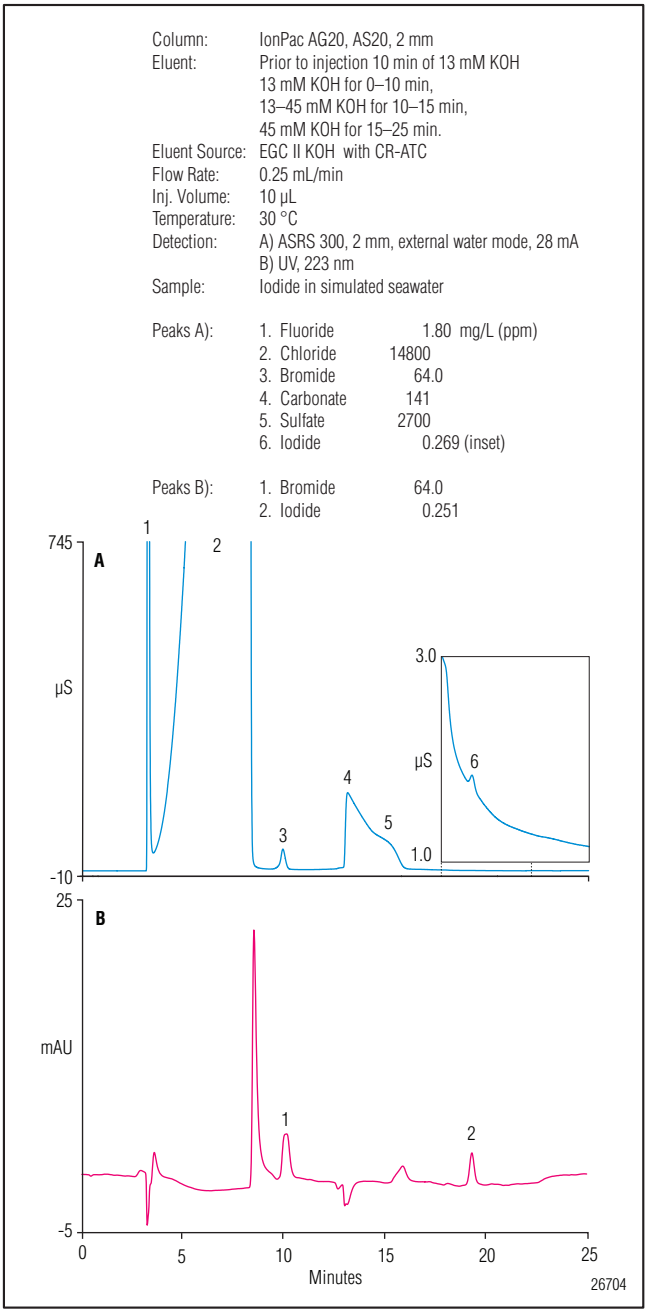

Figure 1. Determination of iodide in simulated seawater on the IonPac AS20 column.

Table 2. Linearity, LOD, LOQ, and Precision for Iodide Determination by UV and CD Detection Methods							
Detection	**Retention Time (min)**	**Range (µg/L)**	**Correlation Coefficient (r^2)**	**LOD* (µg/L)**	**LOQ* (µg/L)**	**Retention Time Precision (RSD)****	**Peak Area Precision (RSD)****
Conductivity	19.33	50–250	0.9967	15	50	0.013	1.67
UV	19.43	50–250	0.9962	15	50	0.032	1.34

**LOQ and LOD are highly dependent on the noise the day they are measured. The LOD and LOQ are concentrations that resulted in peaks during three days of testing that were an average of 3X and 10X the noise respectively.*
*** Seven injections of 100 µg/L iodide standard.*

Accuracy and Precision

The method performance was initially evaluated with seven replicate injections of a 100 µg/L iodide standard. The calculated retention time and peak area precisions were ≤0.03% and ≤1.7%, respectively. Two samples were analyzed for iodide: a synthetic sea salt and an iodized table salt. Freshly prepared table salt contained 92.4 µg/L of iodide, or 46 µg/g of iodide in the dry salt. Figure 2 shows the determination of iodide in table salt using the AS20 column and conductivity and UV detections. The concentrations of chloride, carbonate, and sulfate in this sample are less than in the simulated seawater and, therefore, iodide is easily quantified using both conductivity and UV detections. The precision for triplicate injections of these samples was equivalent to or better than that observed for the standards (Table 3). Figure 3 shows the separation of iodide in a sample of synthetic sea salt. When prepared as described in the sample preparation section, synthetic sea salt contained 108 µg/L of iodide in solution, which is 3.6 µg/g of iodide in the dry salt. When compared to the table salt (Fig. 2A), the iodide is difficult to determine using conductivity detection due to the high concentrations of sulfate and carbonate. However, the iodide peak is clearly resolved and easily quantified using UV detection (Figure 3B).

Table 3. Determination of Iodide in Saline Samples with UV Detection, Triplicate Injections					
Sample	Retention Time (min)	Retention Time Precision (RSD)	Peak Area Precision (RSD)	Amount Found (µg/L)	Amount in Dry Salt (µg/g)
Synthetic Sea Salt (30.4 mg/mL)	19.36	0.05	0.524	108 ± 0.6	3.6 ± 0.02
Fresh Table Salt (1.98 mg/mL)	19.43	<0.01	1.237	92.4 ± 1.2	46 ± 0.7

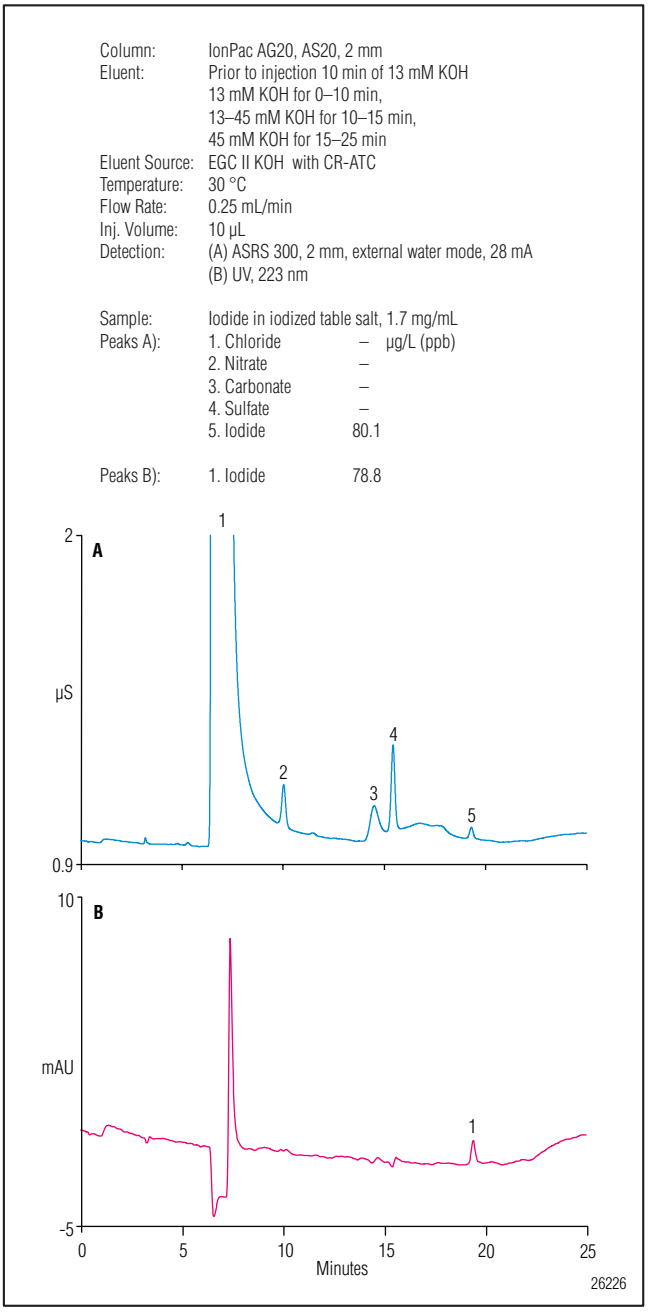

Figure 2. Determination of iodide in iodized table salt.

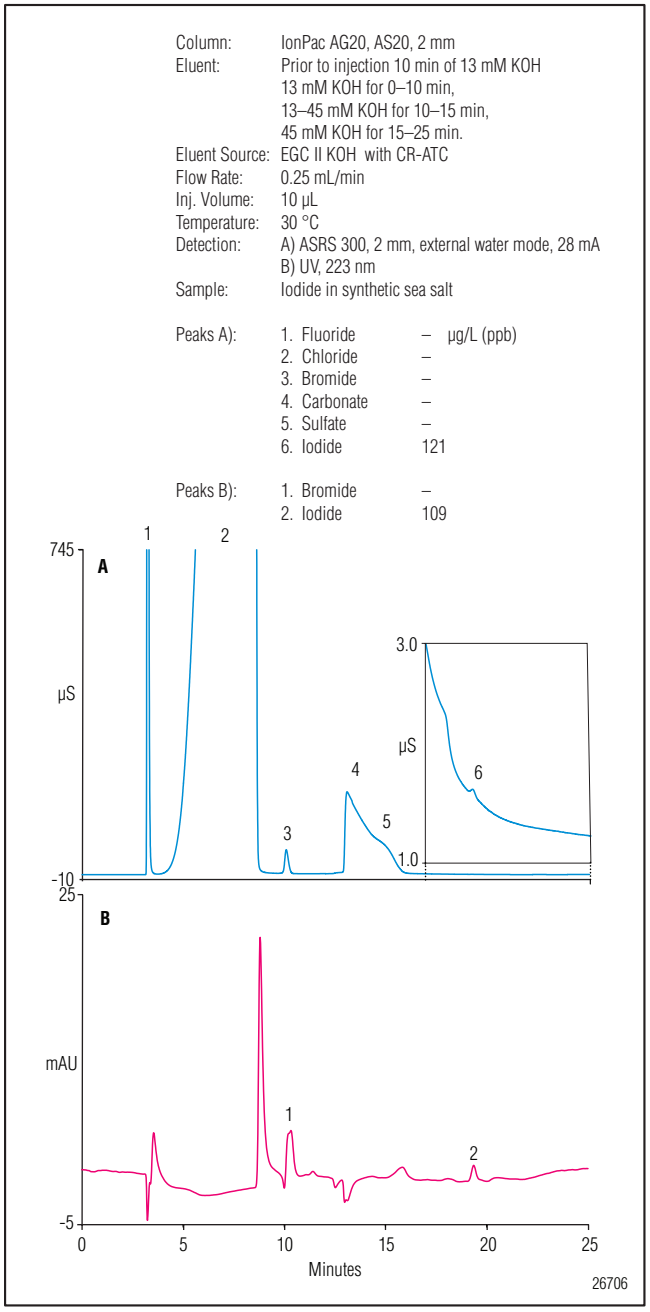

Figure 3. Determination of iodide in synthetic sea salt.

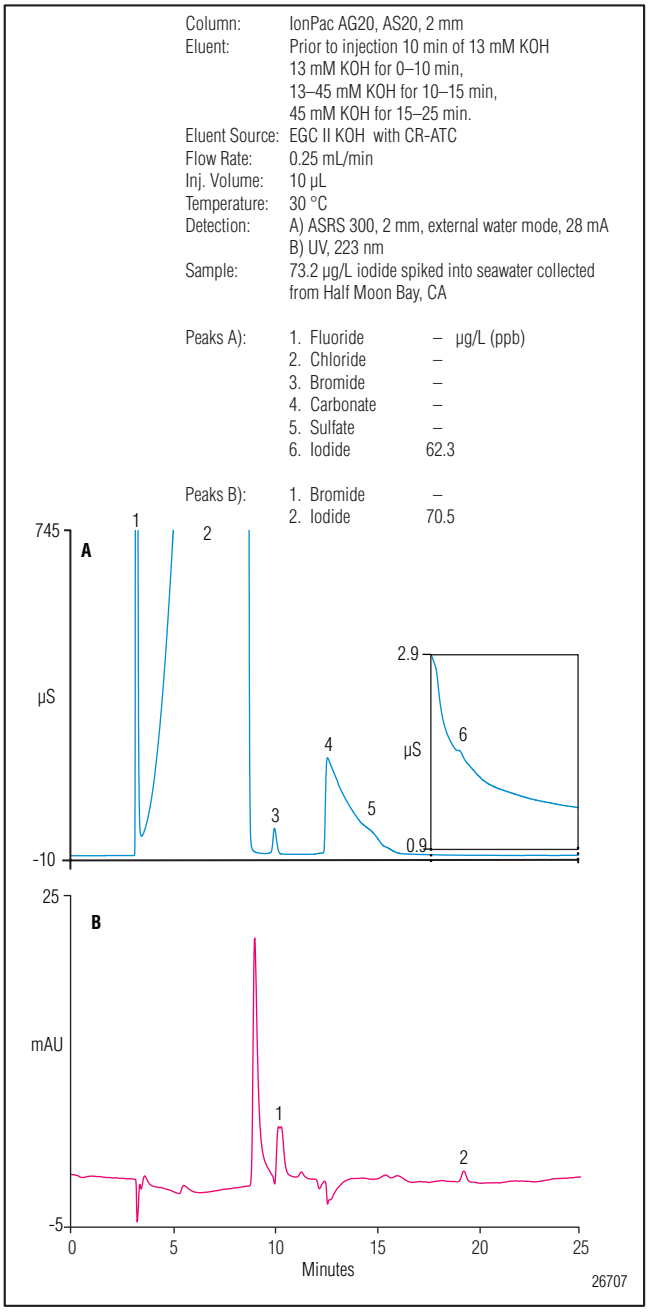

Figure 4. Determination of iodide in seawater collected at Half Moon Bay, CA.

To evaluate accuracy, recoveries were determined in five samples spiked with iodide. Recoveries using UV detection are excellent, ranging from 94–103% in saline matrices (Table 4). The salinity of the matrices is highly variable and these recoveries suggest that the method is accurate. Figures 4A and 5A show the separation of iodide spiked into natural seawater collected in Half Moon Bay, CA and Pacifica, CA, respectively.

In both samples, the iodide is very difficult to determine by conductivity detection and recoveries are low (77–85%). Sulfate and carbonate in the sample obscure iodide and make determination by suppressed conductivity difficult, leading to the observed poor recoveries. As shown in Figures 4B and 5B, iodide is easily determined by UV detection, resulting in improved recoveries.

Table 4. Recoveries of Iodide from Saline Samples, UV Detection

Sample	Amount Found in Spiked Sample (µg/L)	Amount Added (µg/L)	Recovery (%)*
Simulated Sea Water	48.2 ± 2.0	49.5	97 ± 4.1
	142 ± 1.2	141	101 ± 0.9
	252 ± 1.5	244	103 ± 0.6
Synthetic Sea Salt	201 ± 0.8	96.6	94 ± 0.4
Table Salt, fresh	189 ± 1.0	99.1	99 ± 0.5
Seawater, Half Moon Bay, CA	70.5 ± 0.8	73.2	96 ± 1.2
Seawater, Pacifica, CA	72.4 ± 0.7	74.6	97 ± 1.0

** Recovery of iodide was determined by triplicate spiked sample injections immediately following triplicate unspiked sample injections.*

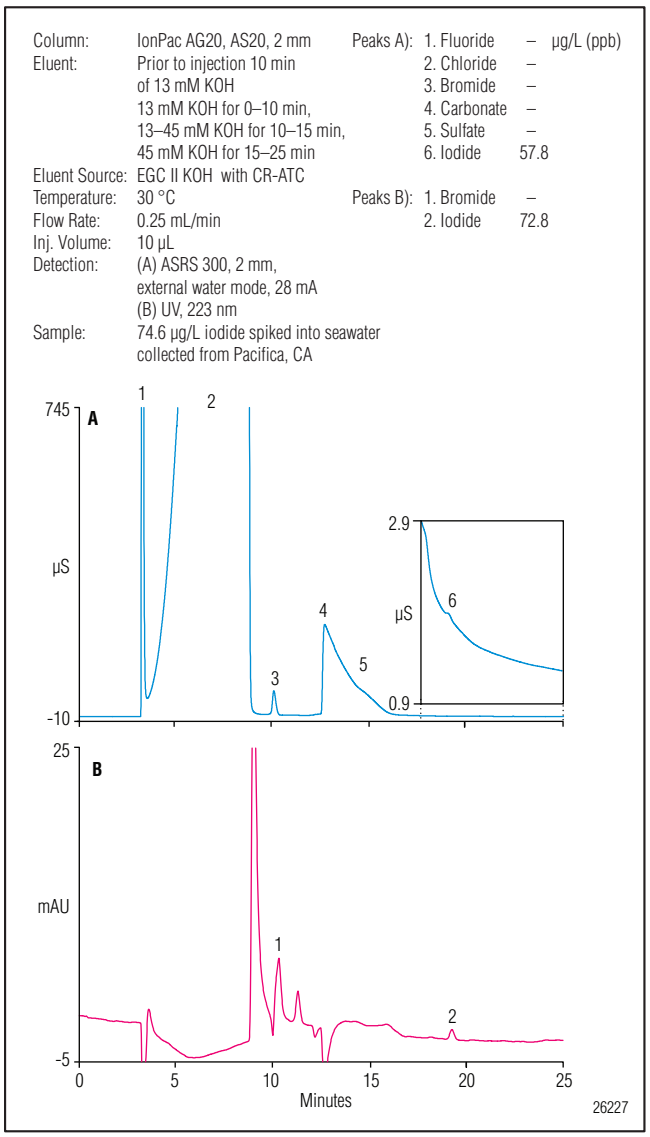

Figure 5. Determination of iodide in seawater collected at Pacifica, CA.

CONCLUSION

This application note describes the use of the IonPac AS20 column with electrolytically generated hydroxide eluent to determine iodide in saline matrices. The high capacity of the AS20 column allows sample analysis without dilution or sample pretreatment. The method was shown to be accurate by recovering iodide in a variety of samples including natural seawater and iodized table salt. UV detection simplifies integration of the iodide peak and improves specificity of the method in comparison to conductivity detection. The use of a RFIC-EG system allows continuous operation of the instrument with minimal maintenance. Only water for eluent generation and suppressor regeneration needs to be added to keep the instrument prepared for analyzing samples. Additionally, the 2 mm column format generates less waste and uses less eluent, saving both time and money.

LIST OF SUPPLIERS

Fisher Scientific
2000 Park Lane Drive
Pittsburgh, PA 15275, USA
Tel: 800-766-7000
www.fischersci.com

VWR
1310 Goshen Parkway
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Tel: 800-932-5000.
www.vwr.com

Sigma-Aldrich Chemical Co.
P.O. Box 2060
Milwaukee, WI 53201, USA
Tel: 800-558-9160.
www.sigma-aldrich.com

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Determination of Inorganic Anions in Environmental Waters Using a Hydroxide-Selective Column

INTRODUCTION

Ion chromatography (IC) is now a well-established and accepted technique for the monitoring of inorganic anions in environmental waters, such as surface, ground, and drinking waters. In the U.S., water quality is legislated through the Safe Drinking Water Act (SDWA) and the Clean Water Act (CWA). The goal of the CWA is to reduce the discharge of pollutants into waters, whereas the SDWA ensures the integrity and safety of drinking waters.[1,2] Primary and secondary drinking water standards have been adopted in the U.S. for certain inorganic anions. The U.S. National Primary Drinking Water Standards (NPDWS) include fluoride, nitrite, and nitrate. A maximum contaminant level for each of these anions is specified in the NPDWS as the regulatory standard for minimizing potential health effects arising from their ingestion in drinking water.[3] Other common inorganic anions, such as fluoride, chloride, and sulfate, are considered secondary contaminants and are regulated under the U.S. National Secondary Drinking Water Standards, which are guidelines regarding taste, color, odor, and certain aesthetic effects.[4]

IC has been approved for the compliance monitoring of primary and secondary inorganic anions in drinking water since the mid-1980s, as described in U.S. EPA Method 300.0.[5] In 1992, the U.S. EPA-EMSL (Cincinnati) laboratory recommended promulgation of U.S. EPA Method 300.0 for compliance monitoring in all U.S. EPA regions for the analysis of inorganic anions in wastewater under the National Pollution Discharge Elimination System program.[2]

Many other industrialized countries have similar health and environmental standards and a considerable number of regulatory IC methods have been published worldwide (e.g., in Germany, France, Italy, and Japan) for the analysis of anions in drinking water. In addition, many standard organizations, including ISO, ASTM, and AWWA, have validated and published IC methods for the analysis of inorganic anions in drinking water, groundwater, and wastewater.[6,7]

U.S. EPA Method 300.0 (Part A) describes the use of a Dionex IonPac® AS4A anion-exchange column using a carbonate/bicarbonate eluent and suppressed conductivity detection for the determination of inorganic anions in environmental waters, such as drinking water, wastewater (mixed domestic and industrial), groundwater, and aqueous solid extracts. However, the method allows for alternative columns, eluents, suppression devices, and detectors to be used—provided that equivalent or better performance for the method is obtained and that the quality assurance requirements are met, including an initial demonstration of capability.

Traditionally, columns designed for use with carbonate/bicarbonate eluents have been used for determining inorganic anions in environmental samples. Columns that use hydroxide eluents (i.e., hydroxide-selective columns) have not been as widely used for routine analysis of inorganic anions in environmental waters due to the lack of appropriate selectivity and difficulty in preparing contaminant-free hydroxide eluents. The introduction of automated, electrolytic eluent generation has eliminated the difficulty in preparing hydroxide eluents. A hydroxide-selective column, the Dionex IonPac AS18, was developed to determine inorganic anions in environmental waters. In this application note, we describe the use of automated eluent generation, combined with a high-capacity, hydroxide-selective, anion-exchange column—the IonPac AS18—for the determination of inorganic anions in environmental waters. The linear range, method detection limits, and recovery of fortified sample matrices are described.

EQUIPMENT

A Dionex ICS-2000 Reagent-Free™ Ion Chromatography (RFIC) System was used in this work. The ICS-2000 is an integrated ion chromatograph and consists of:

- Eluent Generator
- Column Heater
- Pump Degas
- EluGen EGC-KOH Cartridge (Dionex P/N 058900)
- CR-ATC (Dionex P/N 060477)

AS50 Autosampler

Chromeleon® 6.5 Chromatography Workstation

This application note is also applicable to other RFIC systems.

REAGENTS AND STANDARDS

Deionized water, Type I reagent-grade, 18 MΩ-cm resistivity or better

Sodium and potassium salts, ACS reagent-grade or better, for preparing anion standards (VWR or other)

Fluoride standard 1000 mg/L, 100 mL (Dionex P/N 037158)

Chloride standard 1000 mg/L, 100 mL (Dionex P/N 037159)

Sulfate standard 1000 mg/L, 100 mL (Dionex P/N 037160)

Bromide standard 1000 mg/L, 100 mL (Ultra Scientific, VWR P/N ICC-001)

CONDITIONS

Columns:	IonPac AS18 Analytical, (4 × 250 mm P/N 060549) IonPac AG18 Guard, (4 × 50 mm P/N 060551)
Eluent:	22–40 mM KOH from 7–8 min
Eluent Source:	ICS-2000 with CR-ATC
Flow Rate:	1.0 mL/min
Temperature:	30 °C
Injection:	25 µL
Detection:	Suppressed conductivity, ASRS® ULTRA, 4 mm (P/N 053947) AutoSuppression® Recycle Mode 100 mA current
System Backpressure:	~2500 psi
Run Time:	20 min

PREPARATION OF SOLUTIONS AND REAGENTS

Stock Standard Solutions

Stock Anion Standard Solutions (1000 mg/L)

For several of the analytes of interest, 1000 mg/L standard solutions are available from Dionex and other commercial sources. When commercial standards are not available, 1000 mg/L standards can be prepared by dissolving the appropriate amounts of the required analytes in 1000 mL of deionized water according to Table 1. Stock standard solutions for nitrite and nitrate were prepared in concentration units as nitrite-*N* and nitrate-*N*, whereas phosphate was prepared in concentration units as phosphate-*P* as specified in U.S. EPA Method 300.0. Standards are stable for at least one month when stored at 4 °C.

Table 1. Preparation of Stock Standard Solutions

Anion	Compound	Amount (g)
Fluoride	Sodium fluoride (NaF)	2.210
Chloride	Sodium chloride (NaCl)	1.648
Nitrite	Sodium nitrite ($NaNO_2$-*N*)	4.926
Bromide	Sodium bromide (NaBr)	1.288
Nitrate	Sodium nitrate ($NaNO_3$-*N*)	6.068
Phosphate	Potassium phosphate, monobasic (KH_2PO_4-*P*)	4.394
Sulfate	Sodium sulfate (Na_2SO_4)	1.479

Working Standard Solutions

Composite working standard solutions at lower analyte concentrations are prepared from the 1000 mg/L stock solutions. Working standards containing less than 100 mg/L anions should be prepared daily. Seven levels of calibration standards were used in this study to cover the expected concentrations found in environmental samples. Table 2 shows the anion standard concentrations used to calculate the method detection limits (MDLs) and the concentration of the quality control standard (QCS) used to determine retention time stability and peak area precision. Table 3 shows the linear concentration range investigated for each inorganic anion.

Table 2. Concentration of MDLs and QCS Standards

Analyte	MDL Calculation Standard (µg/L)	QCS Used for RSD Calculation (mg/L)
Fluoride	10	2
Chloride	10	20
Nitrite-*N*	6.1 (20 as NO_2)	2
Bromide	25	2
Nitrate-*N*	6.8 (30 as NO_3)	10
Phosphate-*P*	23 (70 as PO_4)	2
Sulfate	20	60

SAMPLE PREPARATION

All samples were filtered through an appropriate 0.45-µm syringe filter, discarding the first 300 µL of the effluent. The only exception was the domestic wastewater, which was filtered through a 0.20-µm syringe filter before injection into the IC. However, to prolong column lifetimes, some domestic wastewater samples may require pretreatment with a C18 cartridge to remove hydrophobic organic material.[8,9]

RESULTS AND DISCUSSION

Although U.S. EPA Method 300.0 (Part A) specifies the use of an IonPac AS4A column, section 6.2.2.1 states that, "An optional column may be used if comparable resolution of peaks is obtained and the requirements of Section 9.2 can be met." Section 9.4.6 further states that, "In recognition of the rapid advances occurring in chromatography, the analyst is permitted certain options, such as the use of different columns and/or eluents to improve the separations or lower the cost of measurements." Each time such modifications to the method are made, the analyst is required to repeat the procedure in Section 9.2 of the method. Section 9.2 discusses the quality control parameters, including the initial demonstration of performance, linear calibration range, quality control samples, and determination of MDLs. Based on this information, the analyst may substitute a column, such as the IonPac AS18, in place of the AS4A, as well as the use of a different eluent, such as hydroxide in place of carbonate/bicarbonate, as in the case of the AS18 hydroxide-selective column.

The IonPac AS18 is a latex agglomerated column with a 7.5-µm-diameter macroporous resin bead consisting of ethylvinylbenzene (EVB) cross-linked with 55% divinylbenzene (DVB), which makes the column 100% solvent compatible. The outer layer consists of 65 nm latex functionalized with alkanol quaternary ammonium groups. The net result is a column with a high-capacity, improved efficiency, and greater selectivity toward hydroxide eluents than the AS4A column.

The IonPac AS18 has a significantly higher capacity (285 µeq/column compared to 20 µeq/column for the AS4A). This higher capacity allows improved resolution between chloride and nitrite and the ability to better tolerate high-ionic-strength matrices without column overloading, which is important in the environmental industry—particularly for the analysis of wastewater samples. Comparison of chromatograms (Figure 1) obtained with the AS18 and AS4A columns reveals noticeable differences in selectivities. Hydroxide-selective stationary phases typically give a greater retention of weakly retained analytes, such as fluoride and acetate, and only moderate retention of divalent hydrophilic anions, such as sulfate.[10] This greater retention is evident from the separation using the AS18 column (Figure 1B) where fluoride is well resolved from the void volume, whereas fluoride is not completely resolved from the void volume using the AS4A column (Figure 1A). Additionally, sulfate elutes between bromide and nitrate on the AS18, whereas on the AS4A sulfate is the last eluting peak, which is typical for a column using carbonate eluents. Finally, phosphate elutes after sulfate on the AS18 column when using the hydroxide eluent conditions in Figure 1B. The higher eluent pH, compared to the AS4A, results in a greater charge on the polyprotic acid species, therefore increasing its retention.

Traditionally, common inorganic anions have not been determined using hydroxide eluents due to the lack of a suitable hydroxide-selective column and the difficulty in preparing contaminant-free hydroxide eluents. Additional precautions must be taken when preparing hydroxide eluents to minimize contamination by carbonate, which can cause a significant baseline shift during a hydroxide gradient and variation in retention times.[11] Therefore, eluents are best prepared from fresh 50% (w/w) sodium hydroxide aqueous solution rather than pellets, because the pellets are normally coated with a layer of carbonate formed when CO_2 from the atmosphere

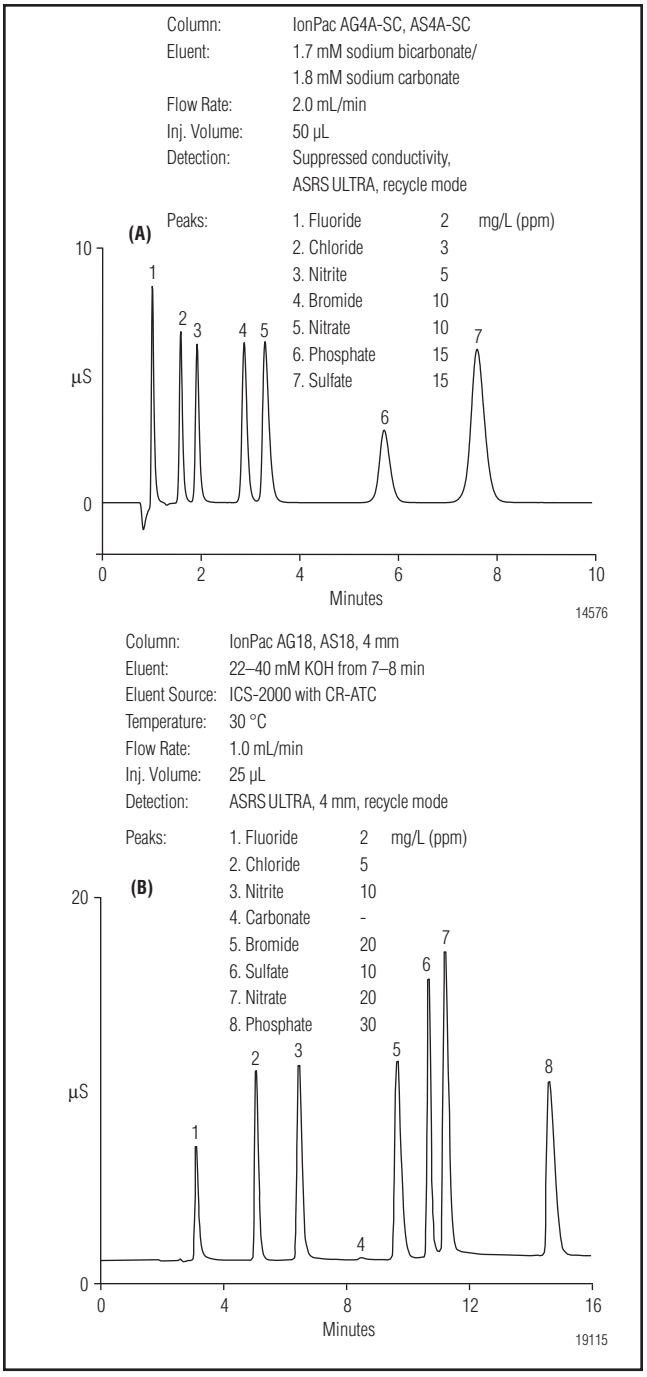

Figure 1. *Separation of common inorganic anions using the IonPac AS4A-SC column (A) and the IonPac AS18 column (B).*

is absorbed onto the pellet surface. The hydroxide solution should be weighed and quickly transferred to a container with an appropriate volume of degassed water and then pressurized with helium. Use of an anion-exchange trap column can reduce carbonate contamination in the eluent. However, a moderate baseline rise is still observed during hydroxide gradient analysis.

Table 3. Linearity, MDLs, and Retention Time and Peak Area Precisions Obtained Using the IonPac AS18 Column[a]

Analyte	Range (mg/L)	Linearity (r^2)	Calculated MDL[b] (µg/L)	Retention Time Precision (RSD[c]) %	Peak Area Precision (RSD) %
Fluoride	0.1–100	0.9991	2.3	0.13	0.27
Chloride	0.2–200	0.9999	2.5	0.09	0.19
Nitrite-*N*	0.1–100	0.9992	1.6 (5.3 as NO_2)	0.06	0.25
Bromide	0.1–100	0.9999	5.7	<0.05	0.73
Nitrate-*N*	0.1–100	0.9999	1.6 (7.1 as NO_3)	<0.05	0.19
Phosphate-*P*	0.1–100	0.9999	5.3 (16.3 as PO_4)	<0.05	0.63
Sulfate	0.2–200	0.9998	5.1	<0.05	0.19

[a] Dionex ICS-2000 Reagent-Free IC System

[b] MDL = $\sigma t_{S,99}$ where $t_{S,99}$ = 3.14 for n = 7

[c] RSD = Relative Standard Deviation, n = 7

To overcome the difficulties typically encountered when preparing hydroxide eluents, an electrolytic eluent generation device has been developed that automates the production of high-purity, carbonate-free potassium hydroxide eluents. This device essentially eliminates the adsorption of carbon dioxide in the hydroxide eluent that can result in undesirable baseline shifts and irreproducible retention times, and therefore compromise the integrity of the analytical results. The replacement of a conventional anion-exchange trap column with a Continuously Regenerated Anion Trap Column (CR-ATC)—for removal of carbonate and other anionic contaminants from the source water—is strongly recommended when using hydroxide eluents. The CR-ATC minimizes baseline shifts, improves retention time stability, and improves detection limits.[12] In addition, the CR-ATC offers several advantages over conventional anion trap columns. The CR-ATC eliminates the need for off-line chemical regeneration of the trap, allowing continuous operation and fast IC system start-up after shutdown.

The quality control section of U.S. EPA Method 300.0 (Section 9.0) requires a demonstration of linearity, MDLs, and acceptable instrument performance by the analysis of a QCS prior to performing analyses using the method. The method linearity using the IonPac AS18 was determined over a seven-point calibration range. MDLs for each of the anions in U.S. EPA Method 300.0 Part A were determined by performing seven replicate injections of deionized water, fortified at a concentration of three to five times the estimated instrument detection limits. Table 2 shows the standards used to calculate the MDLs and concentrations of the QCS. Table 3 shows the linear concentration ranges investigated, the coefficients of determination (r^2), and calculated MDLs for each target anion that was performed on the IonPac AS18 column using electrolytic generation of potassium hydroxide with an ICS-2000 system. Retention time and peak area precisions were determined from seven replicate injections of a QCS prepared in deionized water (Table 3). The high retention time stability can be attributed to the consistent generation of high-purity potassium hydroxide using the ICS-2000 system.

Table 4. Anion Recoveries for Spiked Water Samples Obtained Using the IonPac AS18 Column						
	Drinking Water		Raw Water		Surface Water	
Anion	Amount Added (mg/L)	Recovery (%)	Amount Added (m/gL)	Recovery (%)	Amount Added (mg/L)	Recovery (%)
Fluoride	1	115.5	1	99.2	1	103.4
Chloride	40	96.9	30	93.8	30	100.3
Nitrite-*N*	1	103.8	2	106.4	2	115.1
Bromide	1	102.2	2	105.3	2	100.3
Nitrate-*N*	5	107.7	5	94.9	5	101.5
Phosphate-*P*	5	102.8	10	92.5	10	93.4
Sulfate	60	97.0	40	98.8	80	97.0
	Domestic Wastewater		Industrial Wastewater		Well Water	
Anion	Amount Added (mg/L)	Recovery (%)	Amount Added (mg/L)	Recovery (%)	Amount Added (mg/L)	Recovery (%)
Fluoride	1	114.5	1	103.1	1	96.9
Chloride	60	101.1	30	94.8	40	99.0
Nitrite-*N*	2	119.9	2	103.5	2	101.1
Bromide	2	106.0	2	104.7	2	102.5
Nitrate-*N*	5	101.8	5	95.1	5	95.0
Phosphate-*P*	20	101.4	5	91.9	5	88.1
Sulfate	56	101.0	80	94.9	50	103.3

The data in Table 3 represent the typical results expected when using the IonPac AS18 for routine analyses of common inorganic anions with U.S. EPA Method 300.0. These results demonstrate that the IonPac AS18 and electrolytically generated hydroxide eluent "improves the separations" as required in Section 9.4.6. The routine use of hydroxide eluents has the potential to further improve the performance of other existing IC methods and applications where carbonate/bicarbonate eluents have commonly been used. The advantages of using hydroxide eluents for IC are improved linearity, lower background conductivity, and improved MDLs when compared to "conventional" IC columns such as the IonPac AS4A that use carbonate/bicarbonate eluents. The use of electrolytically generated potassium hydroxide eluent further increases method automation. Water is the only solution required to operate the system because the hydroxide eluent is electrolytically generated on-line, the CR-ATC requires no off-line regeneration using chemical reagents, and the ASRS electrolyt-ically generates the hydronium ion used for suppression.

The performance of the AS18 was also evaluated through a single-operator precision and bias study using spiked water samples of various origins. Table 4 shows typical recovery results for single-operator data obtained using the IonPac AS18 column for common inorganic anions spiked into drinking water, raw (unfinished) drinking water, and other environmental water matrices.

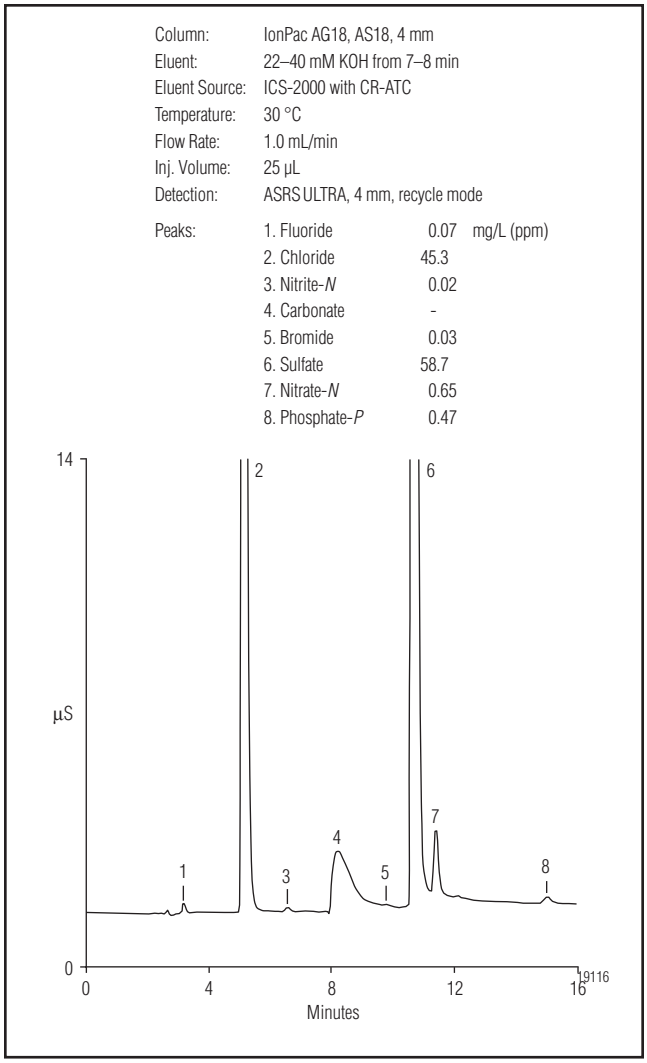

Figure 2. *Determination of inorganic anions in Sunnyvale, CA, drinking water using the IonPac AS18 column.*

Figure 2 shows a chromatogram of inorganic anions in Sunnyvale, California, drinking water. As Table 4 shows, all inorganic anions demonstrated acceptable recoveries (i.e., 80–120%) using the criteria outlined in U.S. EPA Method 300.0. Figure 3A shows a chromatogram of surface water obtained from a lake in Northern California. Figure 3B shows the same surface water sample spiked with 1–80 mg/L of the target

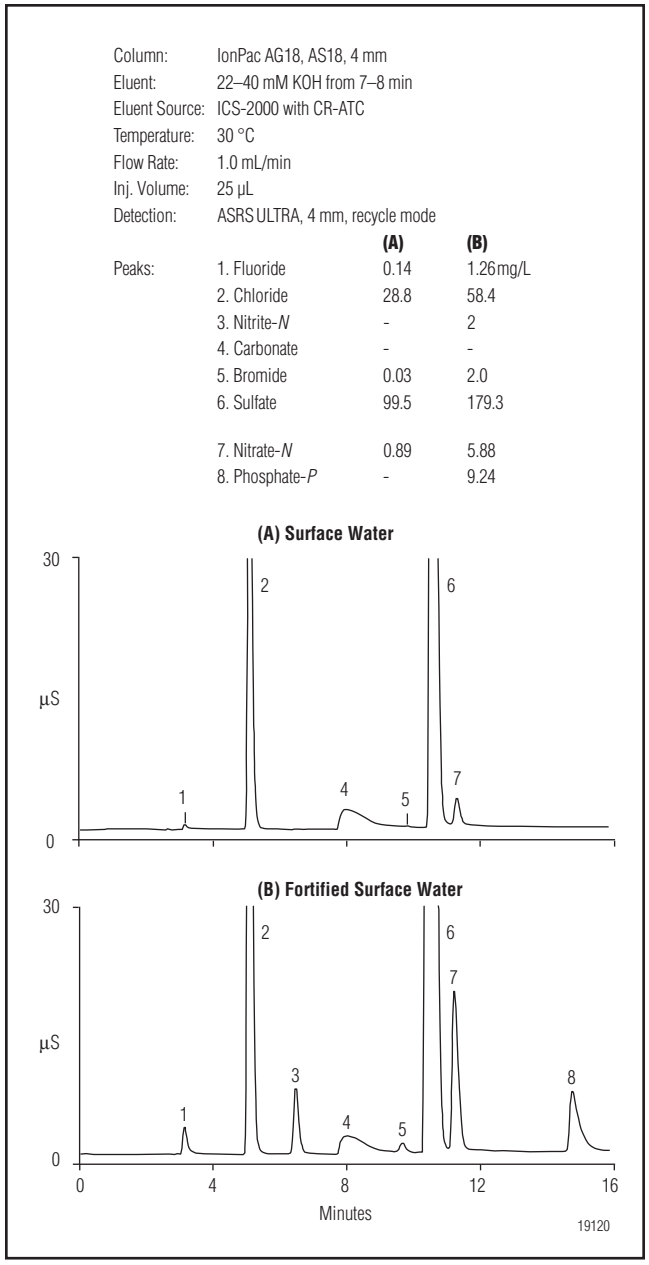

Figure 3. *Determination of inorganic anions in (A) surface water and (B) fortified surface water using the IonPac AS18 column.*

inorganic anions. All peaks were well resolved and recoveries were within the method's required limits (see Table 4, surface water percent recovery). Despite the high concentration of sulfate present, there was no interference with the relatively low concentration of nitrate.

Figure 4 shows a chromatogram of inorganic anions in a more complex matrix, a domestic wastewater sample obtained from a septic sewage system. This chromatogram demonstrates that a high concentration of sulfate (>200 mg/L) can be accurately quantified with excellent peak efficiency and no column overloading. In fact, U.S. EPA Method 300.0 recommends a maximum calibration concentration point of 95 mg/L sulfate and diluting the sample into the working range if the concentration exceeds 95 mg/L. Therefore, the improved linearity obtained by using hydroxide eluents, and the higher capacity of the AS18 column with a calibration range of 0.2–200 mg/L (see Table 3) for sulfate, can improve sample throughput by reducing the need to dilute and reanalyze high-ionic-strength samples.

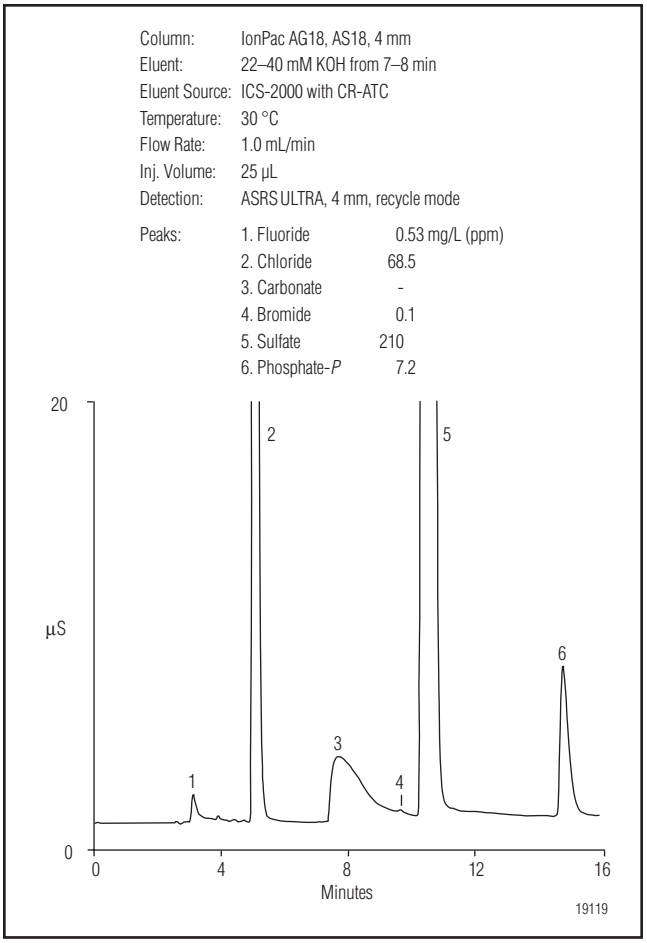

Figure 4. *Determination of inorganic anions in domestic wastewater using the IonPac AS18 column.*

SUMMARY

The use of a Reagent-Free ion chromatograph with an IonPac AS18 column and electrolytic eluent generation is an improved approach to the routine determination of inorganic anions in environmental waters. The AS18 provides improved retention for fluoride from the column void volume, overall improved selectivity, and a significantly higher capacity compared to the AS4A column specified in U.S. EPA Method 300.0. Quantitative recoveries were obtained for all common inorganic anions spiked into typical environmental waters using the AS18 column. In addition, electrolytic generation of potassium hydroxide eliminates the need to manually prepare eluents, increasing the level of automation, ease of use of the IC system, and data reproducibility. This approach to U.S. EPA Method 300.0 allows improved method performance for resolution, linearity, precision, and MDLs. The use of hydroxide eluents in U.S. EPA Method 300.0 and 300.1 has been determined by the U.S. EPA Office of Water to be acceptable for compliance monitoring under the CWA and SDWA.[13]

SUPPLIERS

Fisher Scientific, 2000 Park Lane, Pittsburgh, PA, 15275-1126 USA, Tel: 1-800-766-7000, www.fishersci.com

VWR Scientific, 1310 Goshen Parkway, West Chester, PA 19380 USA, Tel: 1-800-932-5000, www.vwrsp.com.

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13. U.S. Environmental Protection Agency, Office of Water; November 19, 2002.

Fast Analysis of Anions in Drinking Water by Ion Chromatography

INTRODUCTION

The U.S. National Primary Drinking Water Standards specify a Maximum Contaminant Level (MCL) for a number of inorganic anions, including fluoride, nitrite, and nitrate. The MCLs are specified to minimize potential health effects arising from ingestion of these anions in drinking water. Other common anions, such as chloride and sulfate, are considered secondary contaminants and guidelines exist regarding taste, odor, color, and certain aesthetic effects. U.S. EPA Method 300.0[1] describes the use of ion chromatography (IC) with a Dionex IonPac® AS4A anion exchange column, a carbonate/bicarbonate eluent, and suppressed conductivity detection for the determination of these inorganic anions in environmental waters, such as drinking water, wastewater, and aqueous soil extracts. The scope of the method allows optional columns and suppression devices to be used provided that comparable resolution of peaks is obtained and the method quality control requirements can be met.

In this paper, we describe the use of the IonPac AS14A anion exchange column[2] with a new Atlas™ Electrolytic Suppressor (AES™)[3] for the routine high-throughput determination of common inorganic anions in drinking water matrices. The IonPac AS14A provides greater speed and efficiency, ruggedness equivalent to the AS4A-SC column, improved separation of fluoride from the void volume (water dip), and better overall separation selectivity. The AES is a continuously electrolytically regenerated suppressor based on the MonoDisc™ suppression technology. The Atlas electrolytic suppressor offers lower baseline noise and improved ruggedness and reliability. The analytical throughput, potential interferences, linear range, method detection limits, system stability, and analyte recoveries obtained using the AS14A column with the Atlas suppressor for drinking water are described in this Application Note.

EQUIPMENT

Dionex DX-600 ion chromatography system configured for Atlas anion suppression consisting of:

- GS50 or GP50 Gradient Pump
- ED50A Electrochemical Detector or CD25A Conductivity Detector
- AS40 Automated Sampler with 0.5-mL sample vials
- LC30 Chromatography Oven with a rear-loading valve

PeakNet® Chromatography Workstation

REAGENTS AND STANDARDS

Deionized water (DI H_2O), Type I reagent grade, 18 MΩ-cm resistance or better

AS14A Eluent Concentrate (100X), P/N 056937

All anion standards were 99% ACS reagent grade or better:

- Sodium fluoride, CAS 7681-49-4 (Fisher Scientific or other)
- Sodium chloride, CAS 7647-14-5 (J.T. Baker or other)
- Sodium nitrite, CAS 5347-50-0 (Fisher Scientific or other)
- Sodium bromide, CAS 7647-15-6 (EM Sciences or other)
- Sodium nitrate, CAS 7631-99-4 (Aldrich Chemical Company or other)
- Potassium phosphate, monobasic, anhydrous, CAS 7778-77-0 (Sigma Chemical Company or other)
- Sodium sulfate, anhydrous, CAS 7757-82-6 (EM Sciences or other)

CONDITIONS

Columns:	IonPac AS14A Analytical, 3 x 150 mm, 5-µm particle diameter (P/N 056901)
	IonPac AG14A Guard, 3 x 30 mm, 5-µm particle diameter (P/N 056899)
Eluent:	8.0 mM Sodium carbonate and 1.0 mM sodium bicarbonate
Temperature:	30 ˚C
Run Time:	6 min
Flow Rate:	0.8 mL/min
Sample Volume:	25 µL
Detection:	Suppressed conductivity, Atlas AAES™ (P/N 056116), recycle mode, 45 mA
System Backpressure:	2610–2890 psi (18–20 MPa)
Background Conductance:	23–25 µS

PREPARATION OF SOLUTIONS AND REAGENTS

Standard Solution

Starting Anion Standard Solution (10,000 mg/L)

Sodium fluoride—Formula weight = 42.00, anionic mass percent = 45.26%. Dissolve 2.209 g of sodium fluoride solid in 100 mL deionized water.

Sodium chloride—Formula weight = 58.45, anionic mass percent = 60.67%. Dissolve 1.648 g of sodium chloride solid in 100 mL deionized water.

Sodium nitrite—Formula weight = 69.00, anionic mass percent = 66.68%. Dissolve 1.500 g of sodium nitrite solid in 100 mL deionized water.

Sodium bromide—Formula weight = 102.91, anionic mass percent = 77.66%. Dissolve 1.288 g of sodium bromide solid in 100 mL deionized water.

Sodium nitrate—Formula weight = 85.01, anionic mass percent = 72.96%. Dissolve 1.371 g of sodium nitrate solid in 100 mL deionized water.

Potassium phosphate (monobasic)—Formula weight = 136.09, anionic mass percent = 71.27%. Dissolve 1.403 g of potassium phosphate solid in 100 mL deionized water.

Sodium sulfate—Formula weight = 142.06, anionic mass percent = 83.82%. Dissolve 1.193 g of sodium sulfate solid in 100 mL deionized water.

Stock Anion Standard Solution (1000 mg/L)

Dilute each 10,000 mg/L starting anion solution 10-fold in deionized water.

Working Standard Solutions

Dilute 1000 mg/L stock anion standard solutions together as required with deionized water to prepare the appropriate working standard mixtures. The five levels of working standards used in this study for calibration and quality checks are presented in Table 1. These concentration ranges were chosen to bracket the concentrations typical for drinking water samples. The intermediate standard (level 3) was used as a quality check and to evaluate long-term response stability.

LABORATORY FORTIFIED BLANK (LFB) AND MATRIX (LFM)

Dilute 1000 mg/L standard solutions together as required with deionized water to prepare 20X fortification concentrate (see Table 1). The 20X fortification concentrate (5.0 mL) was diluted in deionized labora-

tory water (95.0 mL) to produce the LFB, and 5.0 mL was added to 95.0 mL of Sunnyvale, California drinking water to make the LFM. The concentrations of LFB and LFM are given in Table 1. The LFB and LFM are used to calculate the spike recovery of anions from deionized water and drinking water.

Table 1 Anion Standards and Controls

	Anion Standards,* mg/L (ppm) Levels					LFB*	LFM**
	1	2	3	4	5	DI Water Blank Fortification (ppm)	Matrix Fortification (ppm)
Fluoride	0.1	0.5	2.5	5	10	1	1
Chloride	0.5	5	25	50	100	10	10
Nitrite	0.1	1	5	10	20	2	2
Bromide	0.1	1	5	10	20	2	2
Nitrate	0.1	1	5	10	20	5	5
Phosphate	0.2	1.5	7.5	15	30	10	10
Sulfate	0.5	5	25	50	100	20	20

25-µL Injections
LFB = Laboratory fortified blank
LFM = Laboratory fortified matrix
* Anion standards and LFBs were prepared in laboratory water.
** LFMs were prepared in tap water collected from Sunnyvale, California.

Eluent Solution

8.0 mM Sodium Carbonate/1.0 mM Sodium Bicarbonate

Weigh 1980 g deionized water into an eluent bottle. Degas water for approximately 20 min. Carefully add 20.0 mL of the AS14A Eluent Concentrate (100X) to the degassed water. Mix and then quickly transfer the eluent bottle to the instrument and pressurize the bottle with helium at 8 psi (0.055 MPa). For experiments involving stability studies longer than 1 day, the eluent was prepared as described above but added to the previous eluent.

RESULTS AND DISCUSSION

The use of the IonPac AS14A (3 mm) column at 0.8 mL/min can reduce the run time to 6 minutes (Figure 1) from the usual 10 min for the 0.5 mL/min flow rates. The run times for other IonPac columns commonly used for drinking water analysis are presented in Table 2. Actual run times are typically longer because additional time is needed to fill the injection loop of the autosampler prior to injection and to download data. This additional time varies with the autosampler used. The run times can be reduced by several means: increased eluent strength, increased temperature, and increased flow rate. The AS4A operated at 2 mL/min can reduce the run times to 9 min, but cannot completely resolve the fluoride peak from the water dip. The AS14 column was designed to elute fluoride away from the water dip, but the run times are longer. The AS14A (3 mm) column was designed to elute fluoride from the water dip and also reduce the run times. The performance of this column is maintained

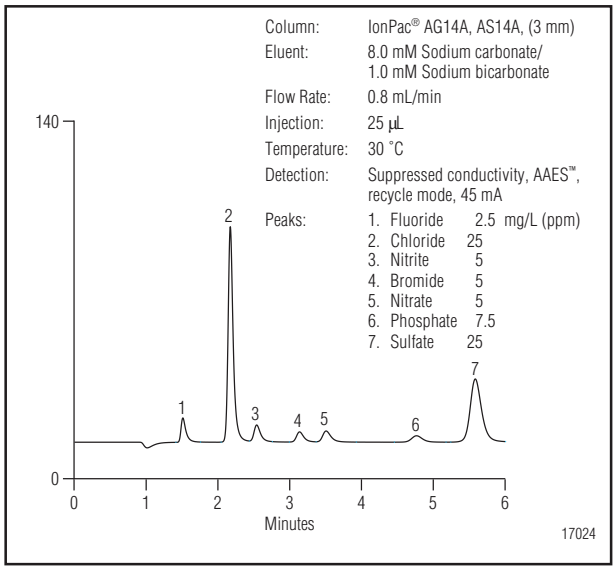

Figure 1. Seven common anion standards using the AS14A (3 mm) at 0.8 mL/min with the AAES suppressor.

Table 2 Comparison of Run Times and Throughput for Dionex IonPac Columns Recommended for Drinking Water Analysis

Column Set	Flow Rate (mL/min)	Sodium Carbonate/ Sodium Bicarbonate Eluent Concentration (mM)	Run Time (min)*	Actual Run Time (min)**	No. of Inject. per 24 h
AS4A/ AG4A (4 mm)	2.0	1.8 mM/1.7 mM	9	10.1	143
AS14/ AG14 (4 mm)	1.2	3.5 mM/1.0 mM	14	15.1	95
	1.2	4.8 mM/0.6 mM	11	12.1	119
	1.5	4.8 mM/0.6 mM	9	10.1	143
	2.0	4.8 mM/0.6 mM	7	8.1	178
AS14A/ AG14A (3mm)	0.5	8.0 mM/1.0 mM	10	11.1	130
	0.8	8.0 mM/1.0 mM	6	7.1	203

* Time from injection to end of shoulder of last peak (sulfate) plus ~1 min
** Actual time per injection, which includes AS40 autosampler loading, injection, and data transfer.

even at higher flow rates, making it well suited for high-throughput water analysis. Using the AS40 autosampler, each injection was calculated to take 7.1 minutes of actual time, and therefore 203 injections were possible over 24 h, surpassing the throughput of any other IonPac column (see Table 2).

The Atlas suppressor is appropriate for this application. Although the Atlas has lower capacity than the ASRS® and AMMS® membrane-based suppressors, the relatively low ionic strength of the carbonate eluent used in this application is well within its operating range. The advantage of Atlas suppression is that baseline noise can be minimized relative to the membrane suppression. In the Recycle mode, the Atlas suppressor provides both low noise and the convenience of long-term maintenance-free operation. An equilibrated system will produce peak-to-peak noise between 0.50–3.5 nS for this application. Figure 2 compares baseline noise between ASRS-ULTRA and Atlas suppressors. The magnitude of the baseline noise can be further reduced by using the External Water mode of suppression as in membrane-based suppressors. All results presented in this Note used the Recycle mode. Both the ASRS-ULTRA and the Atlas suppressor can be used for this high throughput application using the AS14A at higher flow rates, but the Atlas generally produces lower noise and thus lower detection limits.

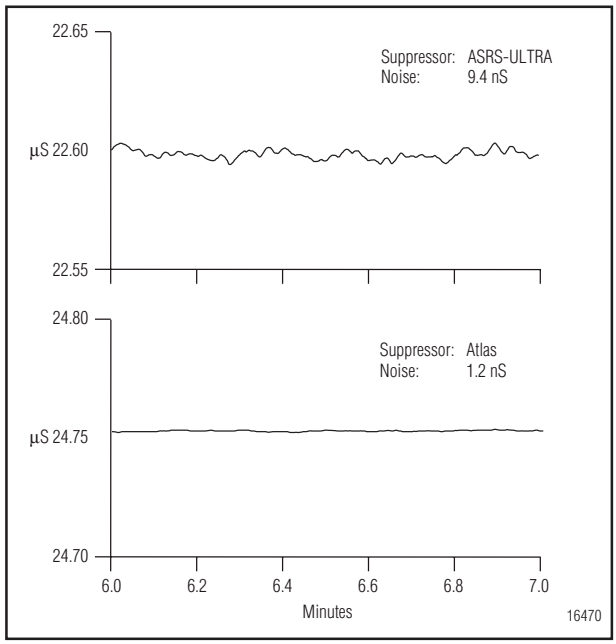

Figure 2. Baseline noise for ASRS and Atlas suppressors using the IonPac AS14A (3 mm) at 0.8 mL/min (8 mM Sodium carbonate and 1 mM sodium bicarbonate eluent).

Because the Atlas suppressor produces lower noise, the limits of detection (LOD) and quantitation (LOQ) are reduced. Table 3 compares the estimated LODs and LOQs of seven anions for both ASRS-ULTRA and Atlas suppressors. The LOD is defined in this Note as the corresponding concentration of each respective standard anion peak height that is equivalent to three times the average of 40 one-minute interval peak-to-peak noise measure. The LOQ is ten times this average. The LOD for the Atlas ranged from 0.8–10 ppb, and from 6–74 ppb for the ASRS-ULTRA. In this study, the Atlas had about a seven times lower detection limit for each anion. Noise values will vary with each suppressor and will affect this comparison. In general, Atlas suppressors should yield lower detection limits than ASRS suppressors.

Table 3 Lower Limits of Detection and Quantification Using the AS14A (3 mm) at 0.8 mL/min with ASRS and Atlas Suppression

	Lower Limits of Detection (LOD)* (ppb)		Lower Limits of Quantification (LOQ)** (ppb)	
	ASRS	Atlas	ASRS	Atlas
Fluoride	6	0.8	20	3
Chloride	7	1	24	3
Nitrite	20	3	68	9
Bromide	29	4	95	13
Nitrate	24	4	82	12
Phosphate	74	10	245	32
Sulfate	25	3	83	10

* LOD based on 3 times the peak-to-peak noise.

** LOQ based on 10 times the peak-to-peak noise.

The lower detection limits using Atlas suppression permit the detection of trace anions in drinking water. Figure 3 shows the separation of anions in Sunnyvale, CA drinking water using the AS14A (3 mm) column at 0.8 mL/min with Atlas suppression. Bromide was measured at a concentration of 20 ppb in this drinking water and the LOD for this ion was 4 ppb, which is above the measured concentration of this drinking water sample based on the Atlas results. The measure of bromide at this concentration is often difficult to achieve by other methods.

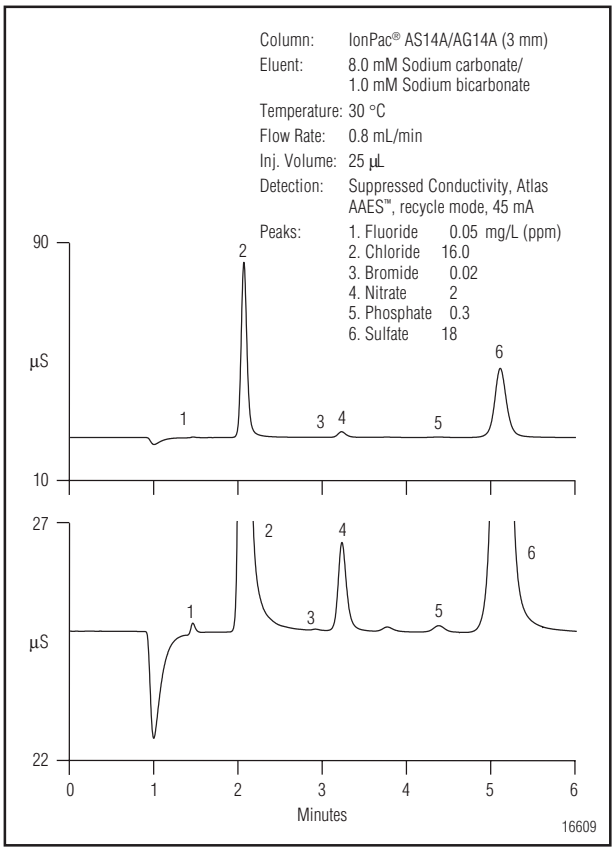

Figure 3. Anions in Sunnyvale, CA drinking water in August separated using the IonPac® AS14A (3 mm) at 0.8 mL/min.

Peak area precision generally improves at higher concentrations. Sunnyvale drinking water was analyzed sequentially for over 6 days in groups of 20 injections interspersed with duplicate quality control samples (blank, level 3 standard, LFB, LFM). The measured anion concentrations for the 940 injections of drinking water sample were plotted with the calculated RSDs (Figures 4 and 5). Chloride (17 ppm) and sulfate (19 ppm) concentrations were measured with high precision (1.2 and 1.1% RSD, respectively). At low to trace level concentrations, high stability was also observed. Nitrate (1.9 ppm) was 1.2% RSD, phosphate (0.33 ppm) was 4.9% RSD, fluoride (0.047 ppm) was 5.5%, and bromide (0.021 ppm) was 14% RSD. This measured concentration of bromide (21 ppb) was only slightly higher than the lower limit of quantification (13 ppb) for this anion using the Atlas (Table 3), thus the precision was poor. When ASRS-ULTRA suppression is used under these conditions, the precision decreases for anions close in concentration to the LOD (Table 4).

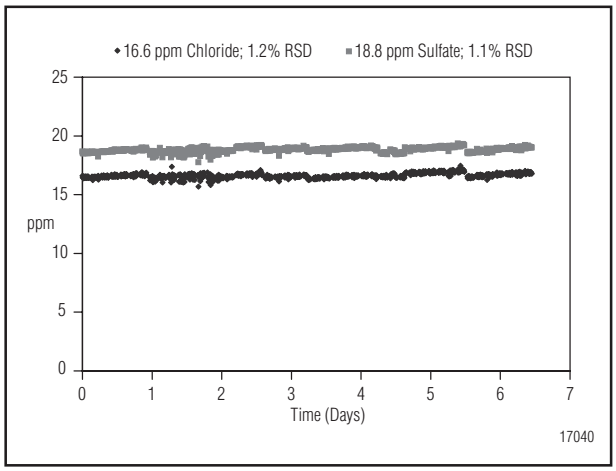

Figure 4. Stability of high level anions in Sunnyvale, CA drinking water using the AS14A (3 mm) at 0.8 mL/min with the Atlas suppressor.

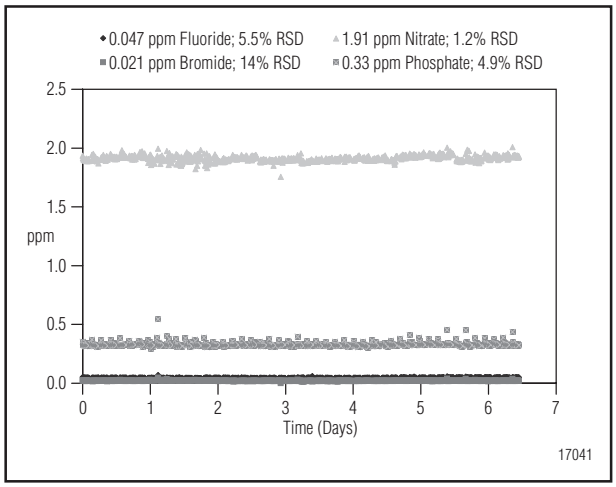

Figure 5. Stability of low level anions in Sunnyvale, CA drinking water using the AS14A (3 mm) at 0.8 mL/min with the Atlas suppressor.

Table 4 Peak Area Precision for the Same Sunnyvale Water Sample Over 3 Days Using ASRS and Atlas Suppressors

	Measured Concentration (ppm)	RSD	
		ASRS %	Atlas %
Fluoride	0.05	21.0	5.1
Chloride	17	1.0	1.1
Nitrite	< 0.003	ND	ND
Bromide	0.02	ND	14.0
Nitrate	2	1.2	1.2
Phosphate	0.3	7.3	5.0
Sulfate	19	0.9	1.1

ND = Not Detected (< 50% of the injections below the detection limit)
IonPac® AS14A and AG14A (3 mm) at 0.8 mL/min, 25 µL injections

The r^2 values ranged from 0.997 to 1.000 for the seven anions (Figure 6). Linearity extends above the concentrations selected for this study and presented in this Application Note. These concentrations used for calibration were designed to appropriately encompass the concentrations typically observed in drinking water samples. The response factors for each anion were calculated from the ratio of peak area to ppm concentration for a 25-µL injection. The response for each anion remained stable over 6 days of continuous operation (Figure 7), with RSDs ranging from 1.0 to 2.3%.

The recovery of anions from either deionized water (LFB) or drinking water (LFM) was calculated using the calibration curve generated at the beginning of the study and remained high over 6 days of continuous operation. The mean % recovery ranged from 96–103% for the LFB, and 92–102% for the LFM (Table 5). At no time did the percent recovery for the LFB drop below the 90% threshold for U.S. EPA Method 300.0, nor did the recovery from LFM drop below the 80% threshold. The retention times remained stable for all anions over 6 days of continuous operation, with RSDs ranging from 0.4–0.9%. At 4.5 days, a slight shift in retention time was observed corresponding in time with the replenishment of an eluent bottle, indicating some interday variance in retention times might occur from minor variations in eluent preparation. The optimized settings for the retention time acceptance window in the PeakNet software and the high level of separation of anions prevented misidentification of peaks.

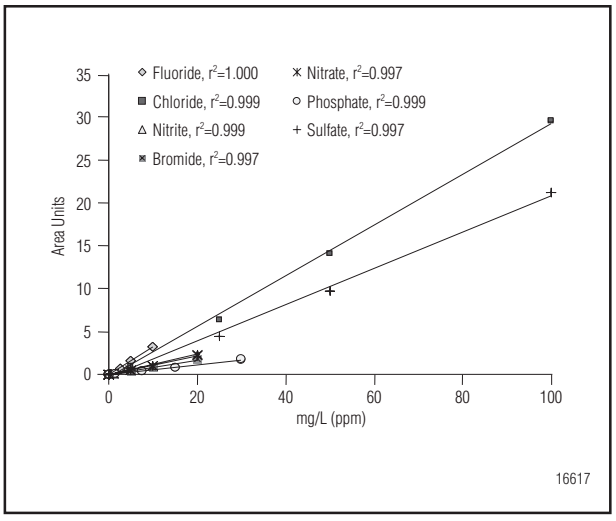

Figure 6. Calibration curves for common anions using the AS14A (3 mm) at 0.8 mL/min with the Atlas suppressor.

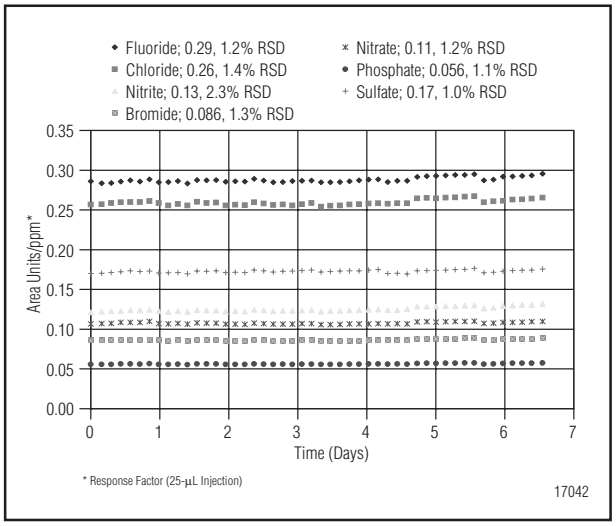

Figure 7. Stability of peak area response factors over 6 days using the AS14A (3 mm) at 0.8 mL/min with the Atlas suppressor.

Table 5 Recoveries of Common Anions From Deionized Lab Water (LFB) and Drinking Water Sample Matrix (LFM) Over 6 Days Using the AS14A (3 mm) at 0.8 mL/min with the Atlas Suppressor

	Recovery			
	LFB		LFM	
Anion	Mean%	RSD%	Mean%	RSD%
Fluoride	96	1.1	97	1.2
Chloride	99	1.4	98	2.8
Nitrite	98	2.5	92	3.2
Bromide	98	1.3	98	1.4
Nitrate	102	1.0	97	1.2
Phosphate	98	1.0	99	1.0
Sulfate	103	0.9	102	1.7

This method is useful for a variety of drinking water samples. Figures 8 and 9 show the separation of anions from fluorinated drinking water (Palo Alto, CA) and from Sierra Nevada mountain water low in ions (Twain Harte, CA), respectively. Even at high fluoride levels, this peak continues to elute out of the water dip using the AS14A (3 mm) column at 0.8 mL/min. When concentrations of ions become low, the use of Atlas suppression makes the detection of trace ions possible and with greater precision and accuracy. Figures 3 and 10 show water collected from the same Sunnyvale source during different seasons (August and February, respectively). Table 6 summarizes the measured results for the water samples tested and shows that this method is capable of monitoring seasonal changes in drinking water anions.

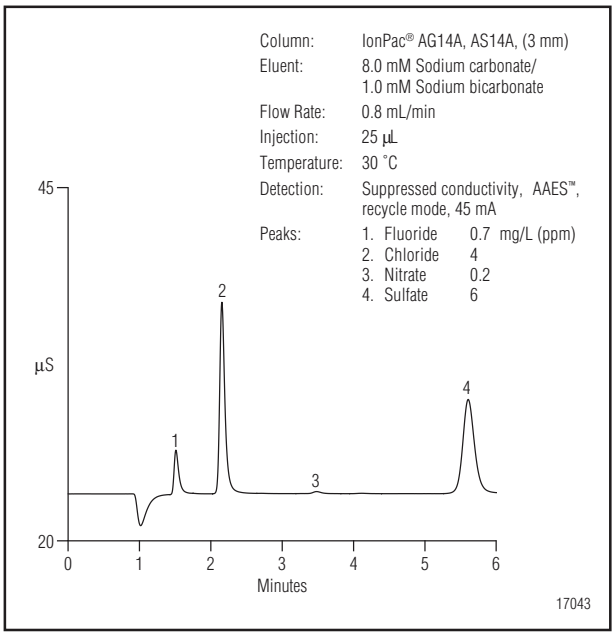

Figure 8. Anions in Palo Alto, CA drinking water separated using the IonPac AS14A (3 mm) at 0.8 mL/min.

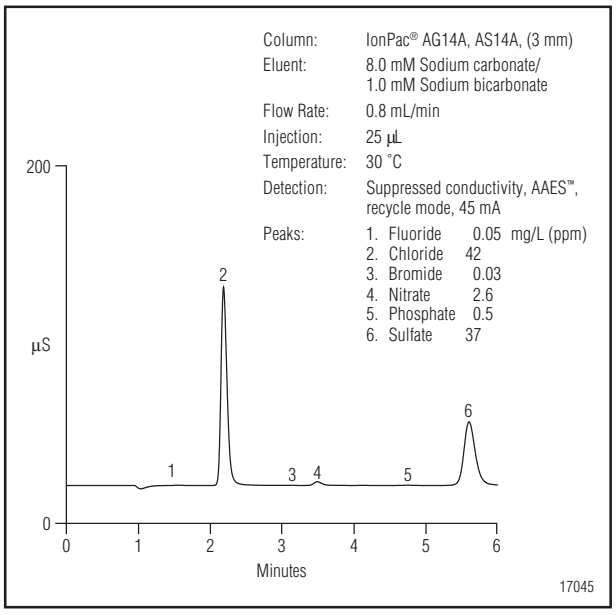

Figure 10. Anions in Sunnyvale, CA drinking water in February separated using the IonPac AS14A (3 mm) at 0.8 mL/min.

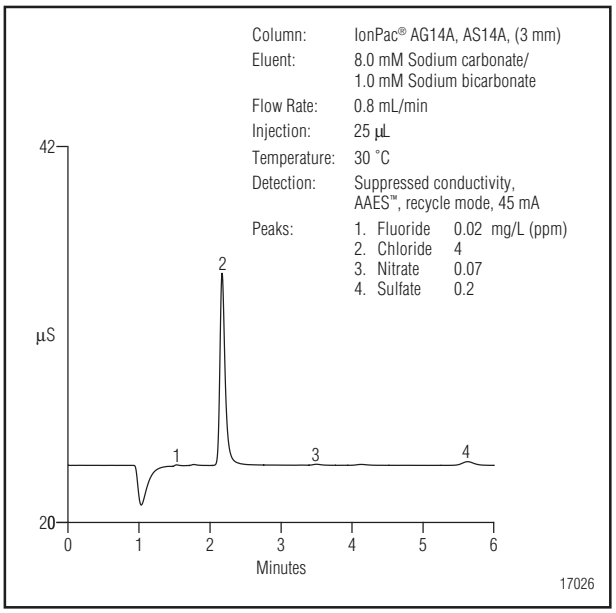

Figure 9. Anions in Twain Harte, CA drinking water separated using the IonPac AS14A (3 mm) at 0.8 mL/min.

Table 6 Summary of Drinking Water Samples Using AS14A (3 mm) at 0.8 mL/min with the Atlas Suppressor

	Sunnyvale 8/8/00 (ppm)	Sunnyvale 2/22/01 (ppm)	Palo Alto 2/21/01 (ppm)	Twain Harte 2/20/01 (ppm)
Fluoride	0.047	0.052	0.728	0.016
Chloride	16.6	42.1	4.1	3.5
Nitrite	< 0.003	< 0.003	< 0.003	< 0.003
Bromide	0.021	0.033	<0.004	<0.004
Nitrate	1.91	2.60	0.17	0.066
Phosphate	0.328	0.483	<0.010	<0.010
Sulfate	18.8	36.8	6.45	0.228

SUMMARY

The IonPac AS14A (3 mm) used at a faster flow rate reduces run times and increases sample throughput. The increase in flow rate from 0.5 to 0.8 mL/min increased the number of injections per day by 56%. The Atlas suppressor lowers baseline noise, which lowers detection limits and therefore improves the detection of trace-level ions. Good long-term performance was realized using the AS14A with Atlas suppression at 0.8 mL/min.

REFERENCES

1. U.S. EPA Method 300.0, "The Determination of Inorganic Anions in Water by Ion Chromatography", United States Environmental Protection Agency, Office of Research and Development, Environmental Monitoring Systems Laboratory, Cincinnati, OH 45268.
2. Jackson, P.E.; Donovan, B; Pohl, C.A.; Kiser, R.E. "A New Block-Grafted Anion Exchange Phase for Environmental Water Analysis Using Ion Chromatography." *J. Chromatog.*, **2001**, *920*, 51–60.
3. Small, H.; Riviello, J. "Electrically Polarized Ion-Exchange Beds in Ion Chromatography: Ion Reflux." *Anal. Chem.* **1998**, *70 (11)*, 2205–2212.

SUPPLIERS

Aldrich Chemical Co., 1001 West Saint Paul Avenue, P.O. Box 355, Milwaukee, Wisconsin 53233, USA. Tel: 1-800-558-9160. www.sigma-aldrich.com

EM Science, 480 South Democrat Road, Gibbstown, NJ 08027, USA. Tel: 1-800-222-0342. www.emscience.com

Fisher Scientific, 2000 Park Lane, Pittsburgh, PA, 15275-1126, USA. Tel: 1-800-766-7000. www.fishersci.com

J.T. Baker Inc., 222 Red School Lane, Phillipsburg, NJ 08865, USA. Tel: 1-800-582-2537. www.jtbaker.com

Sigma Chemical Co., P.O. Box 952968, St. Louis, MO 63195-2968, USA. Tel: 1-800-521-8956. www.sigma-aldrich.com

Determination of Inorganic Anions in Wastewater by Ion Chromatography

INTRODUCTION

The determination of common inorganic anions in environmental waters, such as wastewater and drinking, ground, and surface waters, is one of the most important applications of ion chromatography (IC) worldwide. Water quality in the U.S. is legislated through the Safe Drinking Water Act (SDWA) and the Clean Water Act (CWA). The SDWA ensures the integrity and saftey of U.S. drinking water, and the goal of the CWA is to reduce the discharge of pollutants into U.S. waters.[1,2] Ion chromatography has been approved for compliance monitoring of these common inorganic anions in U.S. drinking water since the mid-1980s, as described in U.S. EPA Method 300.0.[3] This method received technical approval in 1992 and interim regulatory approval in 1995 for the analysis of inorganic anions in wastewater under the National Pollution Discharge Elimination System (NPDES) permits program.[2]

Many other industrialized countries have similar health and environmental standards and a considerable number of regulatory IC methods have been published worldwide (e.g., in Germany, France, Italy, and Japan) for the determiniation of anions in wastewater. In addition, many standards organizations (including ISO, ASTM, and AWWA) have validated IC methods for the analysis of inorganic anions in wastewater.[4,5] This Application Note describes the determination of inorganic anions in wastewater and other environmental waters using conditions that are consistent with those in U.S. EPA Method 300.0.[3] The use of two optional columns, the IonPac® AS14 and IonPac AS9-HC, is also discussed.

EQUIPMENT

Dionex DX-120 and DX-500 Ion Chromatography Systems were used for this work. The DX-120 is a dedicated ion chromatograph; the DX-500 is a modular system, which in this case consisted of:

GP50 Gradient Pump

CD20 Conductivity Detector

AD20 UV/Vis Detector

LC20 Chromatography Enclosure with rear-loading injection valve

AS40 Automated Samplers (5-mL vials) and a PeakNet™ Chromatography Workstation were used with both systems.

REAGENTS AND STANDARDS

Deionized water, Type I reagent grade, 18 MΩ-cm resistance or better

0.18 M Sodium carbonate/0.17 M Sodium bicarbonate (Dionex IonPac AS4A Eluent Concentrate, P/N 39513)

0.35 M Sodium carbonate/0.1 M Sodium bicarbonate (Dionex IonPac AS14 Eluent Concentrate, P/N 53560)

0.5 M Sodium carbonate (Dionex P/N 37162)

Sodium and potassium salts, ACS reagent grade, for preparing anion standards (VWR or other)

Fluoride standard 1000 mg/L, 100 mL (Dionex P/N 37158)

Chloride standard 1000 mg/L, 100 mL (Dionex P/N 37159)

Sulfate standard 1000 mg/L, 100 mL (Dionex P/N 37160)

Nitrate standard 1000 mg/L, 100 mL (Ultra Scientific, VWR P/N ICC-004)

Nitrite standard 1000 mg/L, 100 mL (Ultra Scientific, VWR P/N ICC-007)

Phosphate standard 1000 mg/L, 100 mL (Ultra Scientific, VWR P/N ICC-005)

Bromide standard 1000 mg/L, 100 mL (Ultra Scientific, VWR P/N ICC-001)

CONDITIONS

Part A

Columns:	IonPac AG4A-SC, 4 x 50 mm (P/N 43175) IonPac AS4A-SC, 4 x 250 mm (P/N 43174)
Eluent:	1.8 mM Sodium carbonate/ 1.7 mM Sodium bicarbonate
Run Time:	< 8 min
Flow Rate:	2.0 mL/min
Injection Volume:	50 µL
Detection:	Suppressed conductivity, ASRS®-ULTRA (4 mm) in recycle mode, 50 mA current
System Backpressure:	~ 1000 psi
Background Conductance:	~ 14 µS

Part B

Columns:	IonPac AG14, 4 x 50 mm (P/N 46134) IonPac AS14, 4 x 250 mm (P/N 46124)
Eluent:	3.5 mM Sodium carbonate/ 1.0 mM Sodium bicarbonate
Run Time:	< 14 min
Flow Rate:	1.2 mL/min
Injection Volume:	50 µL
Detection:	Suppressed conductivity, ASRS-ULTRA (4 mm) in recycle mode, 100 mA current
System Backpressure:	~ 1600 psi
Background Conductance:	~ 17 µS

Part C

Columns:	IonPac AG9-HC, 4 x 50 mm (P/N 51761) IonPac AS9-HC, 4 x 250 mm (P/N 51786)
Eluent:	9.0 mM Sodium carbonate
Run Time:	< 24 min
Flow Rate:	1.0 mL/min
Injection Volume:	50 µL
Detection:	Suppressed conductivity, ASRS-ULTRA (4 mm) in recycle mode, 100 mA current
System Backpressure:	~ 2200 psi
Background Conductance:	~ 22 µS

PREPARATION OF SOLUTIONS AND REAGENTS

Stock Standard Solutions

Stock Anion Standard Solutions (1000 mg/L)

For several of the analytes of interest, 1000 mg/L standard solutions are available from Dionex and other commercial sources. When commercial standards are not available, 1000 mg/L standards can be prepared by dissolving the appropriate amounts of the corresponding mass in 1000 mL of deionized water according to Table 1. Standards are stable for at least one month when stored at 4 °C.

Table 1 Masses of Compounds Used to Prepare 1 L of 1000 mg/L Anion Standards

Anion	Compound	Mass (g)
Fluoride	Sodium fluoride (NaF)	2.210
Chloride	Sodium chloride (NaCl)	1.648
Nitrite	Sodium nitrite ($NaNO_2$)	1.499
Bromide	Sodium bromide (NaBr)	1.288
Nitrate	Sodium nitrate ($NaNO_3$)	1.371
o-Phosphate	Potassium phosphate, monobasic (KH_2PO_4)	1.433
Sulfate	Sodium sulfate (Na_2SO_4)	1.522

Working Standard Solutions

Composite working standards at lower analyte concentrations are prepared from the 1000 mg/L standards described above. Working standards containing less than 100 mg/L anions should be prepared daily. Table 2 shows the linear concentration range investigated for each anion, as well as the concentration of the standard used to calculate the method detection limits (MDLs) and the

concentration of the quality control sample (QCS) used to determine the retention time and peak area precision.

Table 2 Concentration of Linearity, MDL, and Reproducibility Standards

Anion	Seven-Point Calibration Range (mg/L)	MDL Calculation Standard (mg/L)	QCS Standard for RSD Calculation (mg/L)
Fluoride	0.1–100	0.025	2
Chloride	0.2–200	0.010	20
Nitrite	0.1–100	0.025	2
Bromide	0.1–100	0.050	2
Nitrate	0.1–100	0.045	10
o-Phosphate	0.1–100	0.045	2
Sulfate	0.2–200	0.050	60

Eluent Solutions

For the IonPac AS4A-SC and AS14 columns, dilute 20 mL of the appropriate eluent concentrate to 2.0 L with deionized water. For the IonPac AS9-HC column, dilute 36 mL of the 0.5 M sodium carbonate concentrate to 2.0 L with deionized water. Transfer to a 2-L eluent container and pressurize the container with helium at 8 psi.

SAMPLE PREPARATION

All samples were filtered through appropriate 0.45-µm syringe filters, discarding the first 300 µL of the effluent to waste, as specified in Section 4.4 of EPA Method 300.0.[3] The domestic wastewater sample was treated with a C_{18} Sep-Pak cartridge (Waters Corporation, Milford, MA) to remove hydrophobic organic material in order to prolong column lifetimes.[6] The C_{18} cartridge was preconditioned with 5 mL of methanol followed by 5 mL of deionized water. The sample (5 mL) was then passed through the cartridge, with the first 1 mL of the effluent being discarded. Aqueous soil extracts were prepared by the extraction of 3.0 g of soil in 30 mL of deionized water in an ultrasonic bath for 30 min followed by filtration with a 0.45-µm filter.

RESULTS AND DISCUSSION

A variety of methods have been used for the analysis of inorganic anions, including traditional spectroscopic techniques such as colorimetry; wet chemical methods such as gravimetric analysis, turbidimetry, and titrimetry; and electrochemical techniques such as ion selective electrodes (ISEs) and amperometric titrations.[4] However, many of these methods are not specific and suffer from interferences or limited sensitivity; they also can be labor-intensive and are often difficult to automate. Because many of the individual test procedures described above can be replaced by one chromatographic separation, IC was quickly accepted by regulatory agencies worldwide for the determination of anions in wastewater and other environmental waters.

IonPac AS4A-SC Column

U.S. EPA Method 300.0(A) specifies the use of an IonPac AS4A anion-exchange column with an eluent of 1.8 mM sodium carbonate/1.7 mM sodium bicarbonate for the separation of common anions.[3] The method specifies the use of an AMMS® (Anion MicroMembrane Suppressor) operated in the chemical regeneration mode; however, an ASRS (Anion Self-Regenerating Suppressor) provides equivalent method performance with added convenience. Conductivity is used as a bulk property detector for the measurement of inorganic anions.

Figure 1 shows a typical chromatogram of a standard containing low-ppm levels of common inorganic anions separated using an IonPac AS4A-SC column as described in Part A of the "Conditions" section. The pellicular AS4A-SC column has an outer layer of latex with selectivity similar to

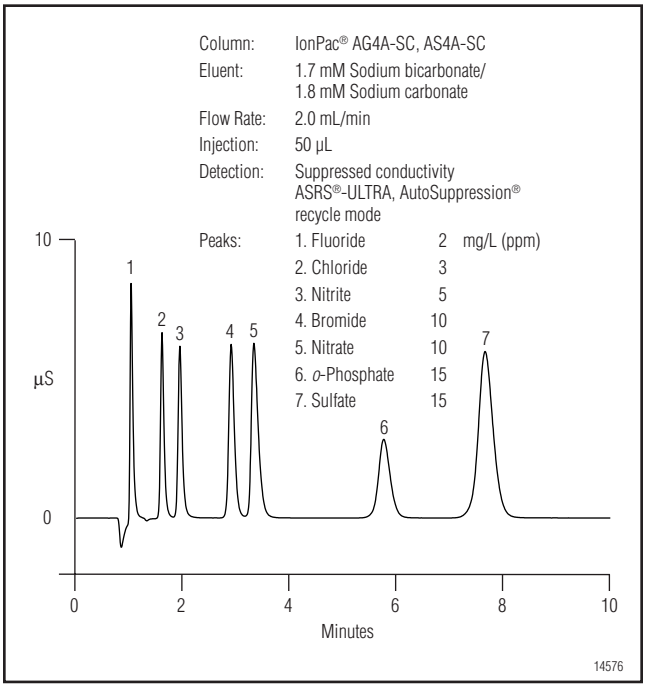

Figure 1. Separation of a low-ppm inorganic anion standard using an IonPac AS4A-SC column.

that of the AS4A, although the substrate of the AS4A-SC is ethylvinylbenzene (EVB) crosslinked with 55% divinylbenzene (DVB), which makes the column 100% solvent-compatible. All the anions are well resolved within a total run time of less than 8 min. The method linearity was determined for the inorganic anions over a seven-point calibration range. MDLs were calculated for each of the anions according to the procedure described in U.S. EPA Method 300.0.[3] The MDLs are estimated by injecting seven replicates of reagent water fortified at a concentration of 3 to 5 times the estimated instrument detection limit. The MDL is then calculated as (t) x (SD) where t = Student's t value for a 99% confidence level and a standard deviation estimate with n - 1 degrees of freedom (t = 3.14 for seven replicates) and SD = standard deviation of the replicate analysis.

Table 3 shows the concentration ranges investigated, the resulting linear coefficients of determination (r^2), and the calculated MDL for each anion. The retention time and peak area precision (expressed as % RSD) were determined from seven replicate injections of a quality control sample, as described in Table 2. Table 3 also shows typical retention time and peak area precision data that can be obtained for inorganic anions using the IonPac AS4A-SC column with a DX-120 system.

The performance of methods used for environmental analysis are typically validated through single- and multi-operator precision and bias studies on spiked samples. Table 4 shows typical recovery results for single-operator data obtained using the IonPac AS4A-SC column for common anions spiked into industrial wastewater, domestic wastewater, and other environmental water matrices. The samples were spiked with the analytes at approximately the same levels as specified in U.S. EPA Method 300.0.[3]

Figure 2A shows a chromatogram of inorganic anions in industrial wastewater from a chemical manufacturing plant obtained using the IonPac AS4A-SC column. Figure 2B shows the same wastewater sample spiked with 1–40 mg/L of inorganic anions. All peaks are well resolved in the spiked sample and acceptable recoveries (i.e., 80–120%) were obtained for all anions in this relatively simple matrix. Figure 3 shows a chromatogram of inorganic anions in a more complex matrix, domestic wastewater from a septic sewage system. In general, Table 4 shows that acceptable recovery data was obtained for the inorganic anions in most matrices. The one exception was the domestic wastewater sample, shown in Figure 3, where a recovery of < 60% was obtained for fluoride under these conditions. Fluoride concentrations of < 1.5 mg/L are subject to interference from mg/L levels of small organic acids, such as formate and acetate, when

Table 3 Linearity, MDL, Retention Time, and Peak Area Precision Obtained Using the IonPac AS4A-SC Column[a]

Anion	Range (mg/L)	Linearity (r^2)	Calculated MDL[b] (µg/L)	Retention Time Precision (% RSD[c])	Area Precision (% RSD)
Fluoride	0.1–100	0.9971	5.9	0.48%	0.67%
Chloride	0.2–200	0.9996	2.3	0.30%	0.47%
Nitrite	0.1–100	0.9997	5.7 (1.8 as NO_2-N)	< 0.05%	0.53%
Bromide	0.1–100	0.9967	9.7	< 0.05%	0.13%
Nitrate	0.1–100	0.9969	6.2 (1.4 as NO_3-N)	0.40%	0.17%
o-Phosphate	0.1–100	0.9967	17.8 (5.8 as PO_4-P)	0.30%	0.35%
Sulfate	0.2–200	0.9975	6.7	< 0.05%	0.14%

[a] Dionex DX-120 system
[b] MDL = $\sigma^* t_{s,99}$ where $t_{s,99}$ = 3.14 for n = 7
[c] RSD = Relative standard deviation, n = 7

Table 4 Anion Recoveries for Spiked Water Samples Obtained Using the IonPac AS4A-SC Column

Anion	Drinking Water		Raw Water		Surface Water	
	Amt. Added (mg/L)	Recovery (%)	Amt. Added (mg/L)	Recovery (%)	Amt. Added (mg/L)	Recovery (%)
Fluoride	1	93.9	1	96.5	1	109.0
Chloride	10	97.4	20	83.2	40	81.4
Nitrite	2	91.6	2	102.1	4	105.0
Bromide	2	98.7	2	96.7	2	101.0
Nitrate	5	92.4	5	94.4	10	96.7
o-Phosphate	10	95.0	10	95.4	10	107.9
Sulfate	20	97.5	40	106.8	40	106.4

Anion	Domestic Wastewater		Industrial Wastewater		Soil Extract	
	Amt. Added (mg/L)	Recovery (%)	Amt. Added (mg/L)	Recovery (%)	Amt. Added (mg/L)	Recovery (%)
Fluoride	1	57.0	1	88.0	2	99.0
Chloride	20	82.7	20	100.8	5	100.2
Nitrite	2	217.0*	2	98.0	2	102.5
Bromide	2	86.5	2	92.0	2	91.0
Nitrate	5	6.8*	5	96.2	5	90.2
o-Phosphate	20	101.6	20	98.8	20	111.7
Sulfate	40	90.6	40	105.9	20	96.6

*Sample stored for longer than recommended holding time; inappropriate recovery due to microbial action.

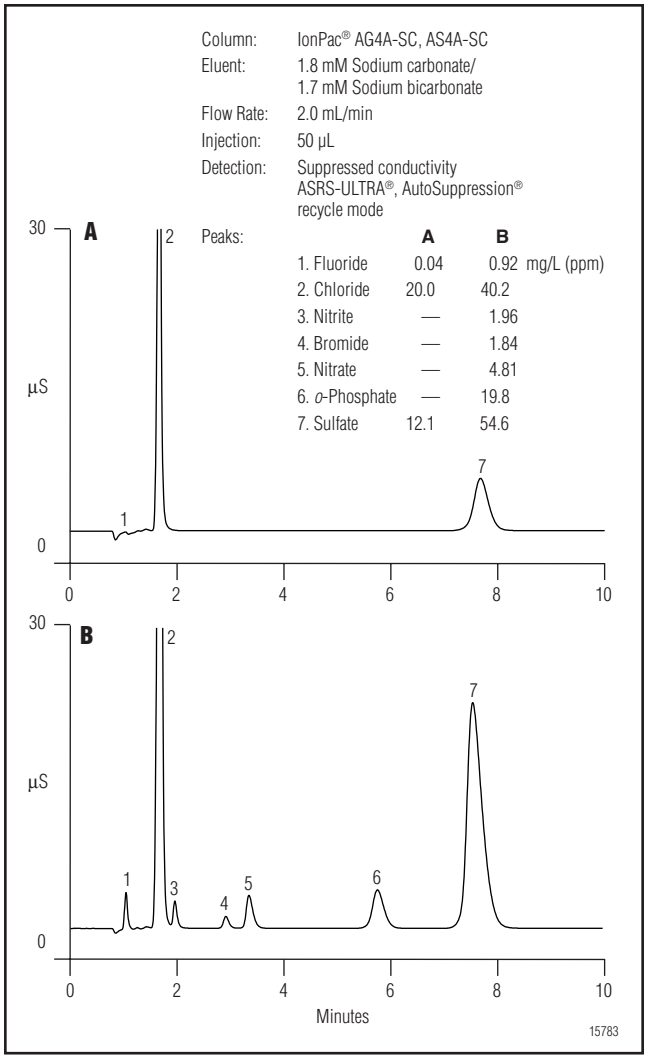

Figure 2. Determination of inorganic anions in A) industrial wastewater and B) spiked industrial wastewater using an IonPac AS4A-SC column.

using the AS4A column.[3] Consequently, this column is not recommended for the analysis of fluoride in complex samples that contain small organic acids.

In this sample, the recoveries for nitrite and nitrate were also not as expected. At the time of these analyses, the sample had been stored (at 4 °C) for longer that the two-day recommended holding time for nitrite/nitrate.[3] In this case, the unexpected recoveries were due to the presence of nitrifying/denitrifying microbes in the sample rather than any chromatographic resolution problems.

IonPac AS14 Column

While U.S. EPA Method 300.0 specifies the use of an IonPac AS4A column, Section 6.2.2.1 states that "an optional column may be used if comparable resolution of peaks is obtained and the quality control requirements of Section 9.2 can be met." The IonPac AS14 column

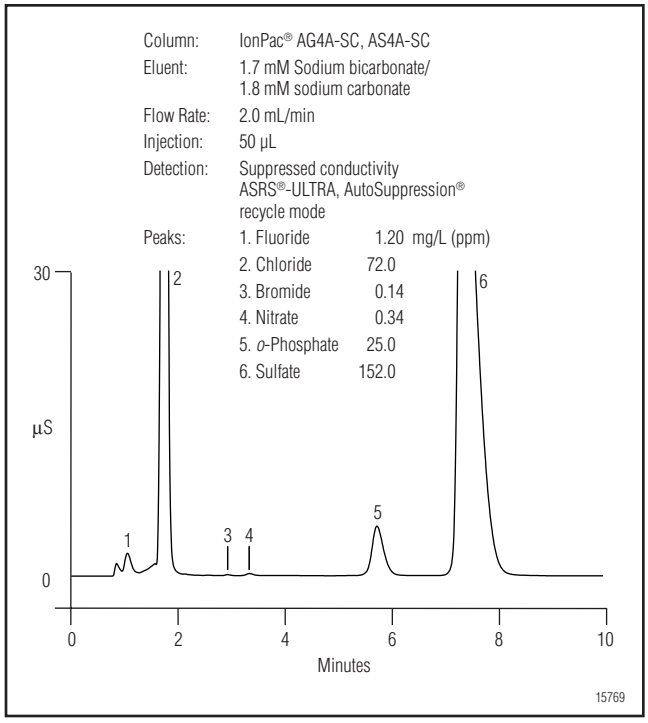

Figure 3. Determination of anions in domestic wastewater using an IonPac AS4A-SC column.

is packed with a methacrylate-based functional group grafted onto the surface of a macroporous resin consisting of EVB crosslinked with 55% DVB. The AS14 column provides complete resolution of fluoride from formate and acetate in addition to improved resolution of fluoride from the column void peak.

The improved selectivity and higher capacity of the AS14 column (65 µeq/column compared to 20 µeq/column for the AS4A) also allows improved resolution of chloride and nitrite, which is important in environmental water analysis. One drawback of using the grafted, higher capacity AS14 column is lower peak efficiencies than those obtained using the latex-agglomerated AS4A-SC column. Figure 4 shows a typical chromatogram of a standard containing low-ppm levels of common anions separated using the IonPac AS14 column as described in Part B of the "Conditions" section. Fluoride is clearly resolved from the column void volume and the overall selectivity is improved compared to the chromatogram shown in Figure 1, although the total run time is increased to 14 min.

The method linearity using the AS14 column was again determined over a seven-point calibration range and the MDLs were calculated according to U.S. EPA Method 300.0.[3] Table 5 shows the concentration ranges investigated, the resulting linear coefficients of determination (r^2), and typical calculated MDLs for

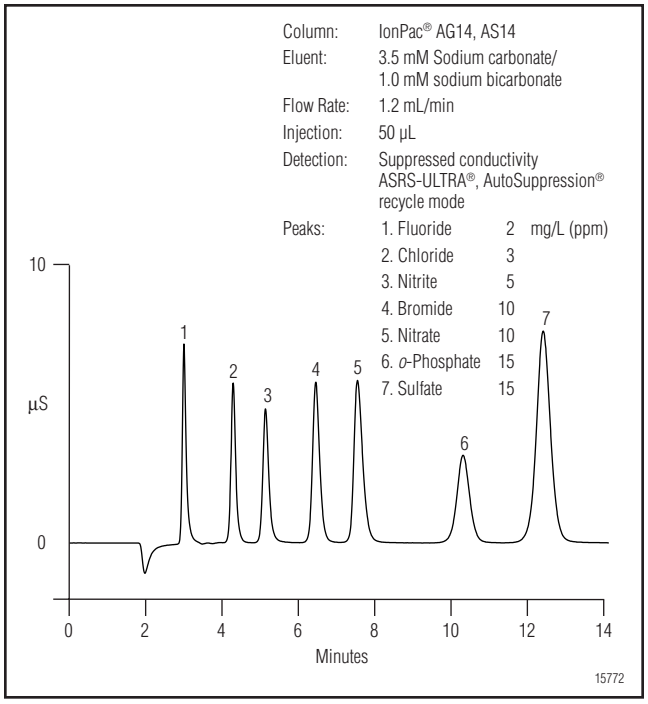

Figure 4. Separation of a low-ppm inorganic anion standard using an IonPac AS14 column.

each anion. The retention time and peak area precision (expressed as % RSD) were determined from seven replicate injections of the quality control sample. Table 5 also shows typical retention time and peak area precision data that can be obtained for inorganic anions using the IonPac AS14 column with a DX-500 system. The performance of the AS14 method was also validated through single-operator precision and bias studies on spiked samples. Table 6 shows typical recovery results for single-operator data obtained using the IonPac AS14 column for common anions spiked into industrial wastewater, domestic wastewater, and other environmental water matrices.

Figure 5 shows a chromatogram of inorganic anions in the same domestic wastewater sample shown in Figure 3, obtained using the IonPac AS14 column. In this case, fluoride clearly is well resolved from the column void volume and also from the later eluting (acetate) peak. Table 6 shows that acceptable recovery data (i.e., 80–120%) was obtained for the inorganic anions in all matrices when using the AS14 column, with the exception of nitrite and nitrate in the same domestic wastewater sample. However, this was again due to the presence of nitrifying/denitrifying microbes rather than any chromatographic resolution problems, although the sample had now been held for a different length of time than when the analysis was first performed using the AS4A-SC column.

Table 5 Linearity, MDL, Retention Time, and Peak Area Precision Obtained Using the IonPac AS14 Column[a]

Anion	Range (mg/L)	Linearity (r^2)	Calculated MDL[b] (µg/L)	Retention Time Precision (% RSD[c])	Area Precision (% RSD)
Fluoride	0.1–100	0.9980	3.5	0.23%	0.17%
Chloride	0.2–200	0.9995	2.9	0.41%	0.51%
Nitrite	0.1–100	0.9997	6.5 (2.0 as NO_2-N)	0.40%	0.37%
Bromide	0.1–100	0.9976	7.8	0.56%	0.51%
Nitrate	0.1–100	0.9970	7.7 (1.7 as NO_3-N)	0.66%	0.54%
o-Phosphate	0.1–100	0.9963	20.2 (6.6 as PO_4-P)	0.15%	0.57%
Sulfate	0.2–200	0.9973	8.2	0.15%	0.59%

[a] Dionex DX-500 system

[b] MDL = $\sigma * t_{s,99}$ where $t_{s,99} = 3.14$ for n = 7

[c] RSD = Relative standard deviation, n = 7

Table 6 Anion Recoveries for Spiked Water Samples Obtained Using the IonPac AS14 Column

Anion	Drinking Water		Raw Water		Surface Water	
	Amt. Added (mg/L)	Recovery (%)	Amt. Added (mg/L)	Recovery (%)	Amt. Added (mg/L)	Recovery (%)
Fluoride	1	91.5	1	85.1	1	101.0
Chloride	10	94.6	20	84.0	40	83.6
Nitrite	2	103.1	2	92.0	4	100.2
Bromide	2	96.1	2	95.6	2	93.3
Nitrate	5	87.2	5	89.4	10	93.2
o-Phosphate	10	93.8	10	94.2	10	106.4
Sulfate	20	96.1	40	106.6	40	106.1
Anion	**Domestic Wastewater**		**Industrial Wastewater**		**Soil Extract**	
	Amt. Added (mg/L)	Recovery (%)	Amt. Added (mg/L)	Recovery (%)	Amt. Added (mg/L)	Recovery (%)
Fluoride	1	90.8	1	90.1	2	101.1
Chloride	20	87.3	20	96.7	5	96.7
Nitrite	2	0.0*	2	98.2	2	89.3
Bromide	2	96.8	2	96.2	2	89.9
Nitrate	5	15.3*	5	95.1	5	92.8
o-Phosphate	20	94.3	20	95.9	20	111.0
Sulfate	40	91.5	40	102.0	20	94.7

*Sample stored for longer than recommended holding time; inappropriate recovery due to microbial action.

IonPac AS9-HC Column

The IonPac AS14 column provides suitable performance for the determination of anions in the majority of wastewater samples; however, very high ionic strength wastewaters are best analyzed using a still higher capacity column. The pellicular stationary phases typically used in IC have a monolayer of fully functionalized latex particles that are electrostatically attached to a surface-functionalized, microporous core particle. The ion-exchange capacity of the resin can be increased by using a larger diameter latex, although this approach ultimately results in decreased efficiency when producing high capacity columns.[7] The AS9-HC column uses a superporous (2000-Å pore size) resin consisting of EVB crosslinked with 55% DVB. This core particle allows the methacrylate-based latex layer to be thinly

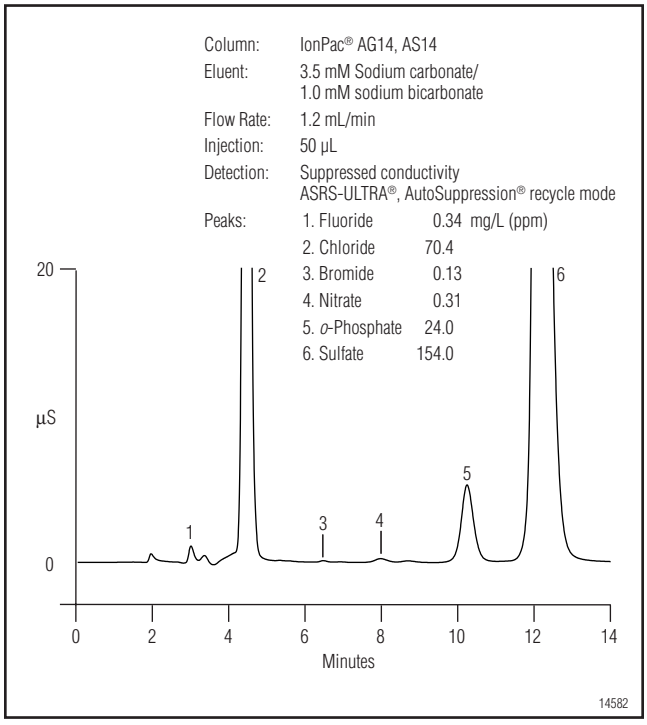

Figure 5. Determination of inorganic anions in domestic wastewater using an IonPac AS14 column.

coated on both the exterior and interior surfaces of the resin and provides a simple way to produce a column with higher capacity (190 µeq/column) using a standard diameter latex while maintaining high chromatographic efficiency.[8]

Figure 6 shows a typical chromatogram of a standard containing low-ppm levels of common anions separated using the IonPac AS9-HC column as described in Part C of the "Conditions" section. Fluoride is again clearly resolved from the void volume and the overall peak resolution is further improved compared to both the AS4A-SC and AS14 columns, hence the AS9-HC column is ideal for the analysis of samples containing dissimilar levels of inorganic anions. The disadvantages of using the high capacity AS9-HC column are that the total run time increases to 24 min and the peak response is reduced when compared to that obtained using the lower capacity AS4A-SC and AS14 columns.

The method linearity using the AS9-HC column was again determined over a seven-point calibration range and the MDLs were calculated according to U.S. EPA Method 300.0.[3] Table 7 shows the concentration ranges investigated, the resulting linear coefficients of determination (r^2), and typical calculated MDLs for each anion. The retention time and peak area precision (expressed as % RSD) were determined from seven replicate injections of the quality control sample. Table 7 also shows typical retention time and peak area precision data that can be obtained for inorganic anions using the IonPac AS9-HC column with a DX-500 system.

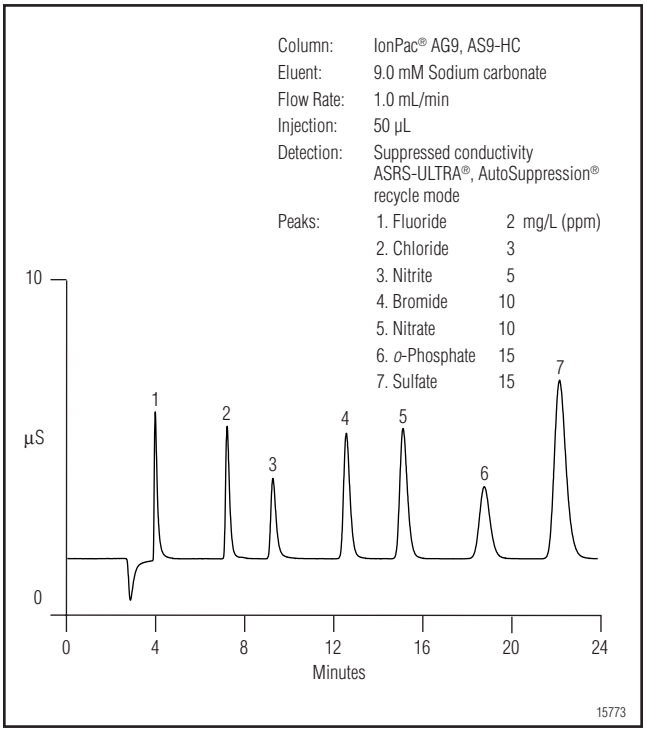

Figure 6. Separation of a low-ppm inorganic anion standard using an IonPac AS9-HC column.

The performance of the AS9-HC method was also validated through single-operator precision and bias studies on spiked samples. Table 8 shows typical recovery results for single-operator data obtained using the IonPac AS9-HC column for common anions spiked into industrial wastewater, domestic wastewater, and other environmental water matrices. Table 8 shows that acceptable recovery data (i.e., 80–120%) was also obtained for the inorganic anions in all matrices when using the AS9-HC column.

Figure 7 shows a chromatogram of 0.2 ppm nitrite spiked into ASTM substitute wastewater (which had been modified to contain elevated levels of chloride and lower nitrite levels) obtained with the AS9-HC column and dual detection using suppressed conductivity and UV/Vis absorption at 214 nm.[9] These chromatograms show the benefit of increased column capacity as chloride and nitrite can be determined at ratios up to 10,000:1 with the AS9-HC column when using conductivity detection. The use of direct UV detection at 214 nm is selective for UV-absorbing anions such as nitrite, bromide, and nitrate and allows the determination of nitrite in the presence of still higher levels of chloride. Table 9 shows the maximum chloride:nitrite ratios that can be analyzed with the AS14 and AS9-HC columns using conductivity and direct UV detection. Note that the ratio is stated with nitrite expressed as both NO_2 and NO_2-N.

Table 7 Linearity, MDL, Retention Time, and Peak Area Precision Obtained Using the IonPac AS9-HC Column[a]

Anion	Range (mg/L)	Linearity (r^2)	Calculated MDL[b] (µg/L)	Retention Time Precision (% RSD[c])	Area Precision (% RSD)
Fluoride	0.1–100	0.9980	7.4	0.36%	0.58%
Chloride	0.2–200	0.9989	5.4	0.22%	0.27%
Nitrite	0.1–100	0.9994	6.2 (1.9 as NO_2-N)	< 0.05%	0.74%
Bromide	0.1–100	0.9968	12.1	0.12%	0.56%
Nitrate	0.1–100	0.9962	10.7 (2.4 as NO_3-N)	0.13%	0.23%
o-Phosphate	0.1–100	0.9972	31.1 (10.2 as PO_4-P)	0.08%	0.75%
Sulfate	0.2–200	0.9967	14.4	0.08%	0.43%

[a] Dionex DX-500 system
[b] MDL = $\sigma * t_{s,99}$ where $t_{s,99}$ = 3.14 for n = 7
[c] RSD = Relative standard deviation, n = 7

Table 8 Anion Recoveries for Spiked Water Samples Obtained Using the IonPac AS9-HC Column

Anion	Drinking Water		Raw Water		Surface Water	
	Amt. Added (mg/L)	Recovery (%)	Amt. Added (mg/L)	Recovery (%)	Amt. Added (mg/L)	Recovery (%)
Fluoride	1	94.8	1	93.3	1	108.3
Chloride	10	96.3	20	85.1	40	81.4
Nitrite	2	95.5	2	96.5	4	106.9
Bromide	2	94.4	2	94.0	2	99.4
Nitrate	5	118.1	15	85.6	10	99.0
o-Phosphate	10	97.4	10	101.9	10	102.1
Sulfate	20	101.9	40	113.2	40	108.9
Anion	**Domestic Wastewater**		**Industrial Wastewater**		**Soil Extract**	
	Amt. Added (mg/L)	Recovery (%)	Amt. Added (mg/L)	Recovery (%)	Amt. Added (mg/L)	Recovery (%)
Fluoride	1	106.8	1	103.6	1	82.5
Chloride	20	86.7	20	104.6	50	86.1
Nitrite	2	102.6	2	103.1	5	118.1
Bromide	2	99.6	2	99.3	2	100.6
Nitrate	5	100.8	5	101.6	5	118.5
o-Phosphate	20	103.5	20	104.9	10	84.3
Sulfate	40	81.4	40	107.5	40	110.5

SUMMARY

The IonPac AS4A-SC column provides suitable performance for the determination of inorganic anions in low ionic strength wastewater samples and similar matrices, such as drinking, raw, and surface waters, as outlined in U.S. EPA Method 300.0. Low levels of fluoride are subject to interference from mg/L levels of small organic acids with the AS4A column, hence this column is not recommended for the analysis of fluoride in more complex wastewater samples that may contain small organic acids.

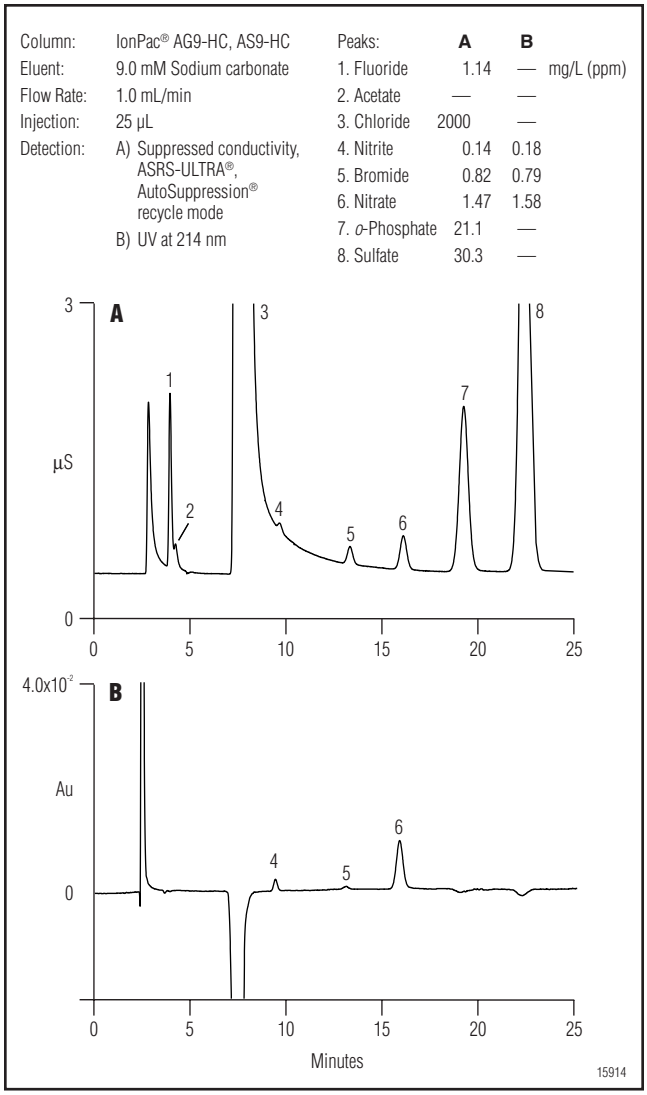

Figure 7. Determination of low levels of nitrite in wastewater containing elevated levels of chloride using the AS9-HC column with dual A) suppressed conductivity and B) UV/Vis detection.

Table 9 Maximum Cl:NO_2 Ratios at Which Nitrite Can Be Quantified Using the IonPac AS14 and AS9-HC Columns[a]

Column and Detection Mode	Cl:NO_2	Cl:NO_2-N
AS14; Conductivity	100:0.1 mg/L (1,000:1)	100:0.034 mg/L (3285:1)
AS14; UV (214 nm)	500:0.03 mg/L (16,667:1)	500:0.009 mg/L (54,761:1)
AS9-HC; Conductivity	2000:0.2 mg/L (10,000:1)	2000:0.068 mg/L (32,850:1)
AS9-HC; UV (214 nm)	5000:0.045 mg/L (111,000:1)	5000:0.014 mg/L (365,000:1)

[a] Dionex DX-500 system with a 25-µL injection

The IonPac AS14 column provides improved fluoride resolution from the column void peak and complete resolution of fluoride from formate and acetate. The improved selectivity and higher capacity make the AS14 column the most appropriate choice for the routine determination of inorganic anions in typical, moderate ionic strength domestic and industrial wastewater samples. The IonPac AS9-HC column has a significantly higher capacity than the AS4A-SC and AS14 columns, so total run times are longer and peak response is somewhat reduced; however, this column is ideal for the determination of inorganic anions in high ionic strength wastewater samples. This column can be used to determine nitrite in a 10,000-fold excess of chloride when using conductivity detection, while direct UV detection allows the determination of nitrite in the presence of still higher levels of chloride.

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SUPPLIERS

Fisher Scientific, 711 Forbes Avenue, Pittsburgh, PA 15219-4785, USA. Tel: 1-800-766-7000.

VWR Scientific, P.O. Box 7900, San Francisco, CA 94120, USA. Tel: 1-800-932-5000.

Waters Corporation, 34 Maple Street, Milford, MA 01757 USA. Tel: 1-800-252-4752.

Determination of Nitrite and Nitrate in Drinking Water Using Ion Chromatography with Direct UV Detection

INTRODUCTION

The ion chromatographic analysis of nitrite and nitrate in drinking water is accomplished using direct UV detection of the analytes. The method is free from most ionic interferences due to the specificity of UV detection. The method is applicable to all drinking water samples. Bromide may also be separated from other ions and detected using this method.

The method of chemically suppressed conductivity detection of nitrite and nitrate in drinking water (Dionex Application Update #131) is an alternative to this method. Note that if the two methods are combined, chemical suppression will yield additional benefits in the determination of nitrite and nitrate with UV detection. The use of a suppressor (AMMS-II, Dionex P/N 043074) between the column and the detector cell reduces background absorbance and eliminates negative peaks associated with chloride and sulfate in this method (see Fig. 3).

RECOMMENDED EQUIPMENT

Dionex Ion Chromatograph with a UV/Visible absorbance detector

REAGENT AND STANDARD PREPARATION

Sodium carbonate / sodium bicarbonate eluent concentrate (P/N 039513)

Sodium Nitrite, ACS Grade

Sodium Nitrate, ACS Grade

Eluent

To prepare 1.0 L of eluent (1.8 mM sodium carbonate, 1.7 mM sodium bicarbonate), dilute 10.0 mL of eluent concentrate to 1000 mL with deionized water.

Stock Standards

1000 ppm Nitrite: Dissolve 1.499 g $NaNO_2$ in 1.0 L of deionized water

1000 ppm Nitrate: Dissolve 1.371 g $NaNO_3$ in 1.0 L of deionized water

Working Standards

Dilute the stock standards to concentration levels that bracket the concentration level of interest. Prepare working standards from the stock standard just prior to analysis.

CONDITIONS

Column:	IonPac® AS9 analytical column with AG9 guard
Eluent:	1.8 mM Na_2CO_3 / 1.7 mM $NaHCO_3$
Flow Rate:	2.0 mL/min
Sample Volume:	25 µL
Detection:	UV at 210 nm, 0.2 AUFS

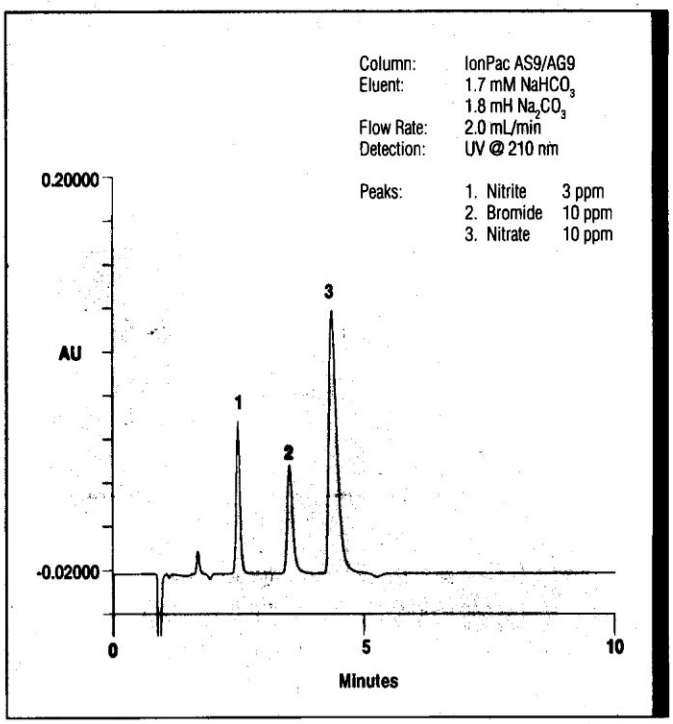

FIGURE 1. DETERMINATION OF NITRITE AND NITRATE WITH DIRECT UV DETECTION

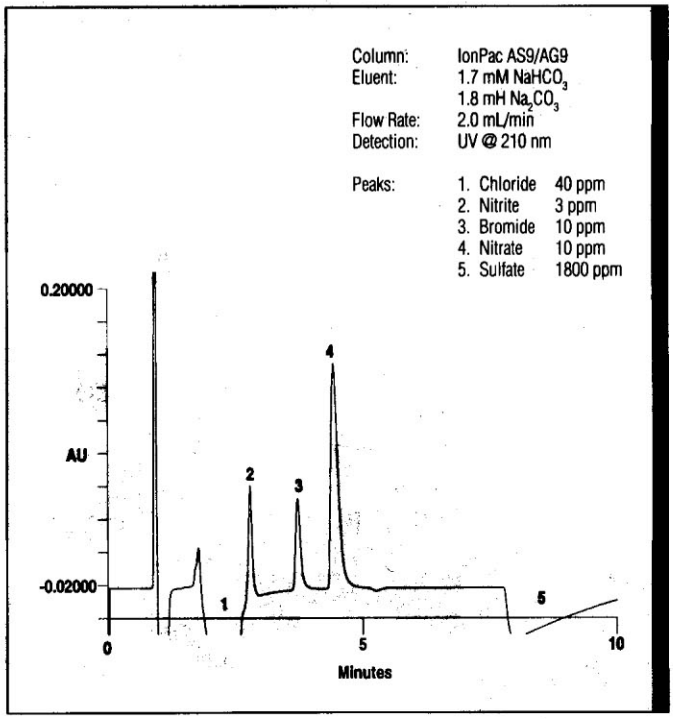

FIGURE 2. DIRECT UV DETECTION OF NITRITE AND NITRATE IN DRINKING WATER (PRESERVED WITH SULFURIC ACID*)

*Presently, the US EPA is evaluating the suitability of acid preservation as described in EPA/570/9-90/008

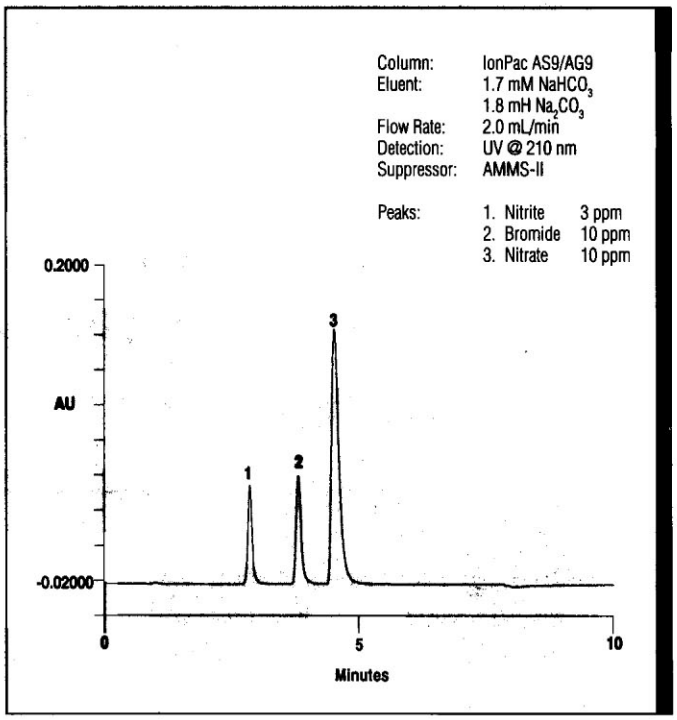

FIGURE 3. DIRECT UV DETECTION OF NITRITE AND NITRATE IN DRINKING WATER (PRESERVED WITH SULFURIC ACID*) WITH CHEMICAL SUPPRESSION OF ELUENT

PERFORMANCE CHARACTERISTICS

The detection limit in drinking water samples using a 25-µL loop is 10 ppb for nitrite and 15 ppb for nitrate. This corresponds to 3.0-ppb nitrogen as nitrite, and 3.5-ppb nitrogen as nitrate. The method is linear for nitrite over the range 10 ppb to 50 ppm. It is linear for nitrate over the range of 15 ppb to 75 ppm.

Determination of Anions in Municipal Drinking Water by Fast IC Using an Hydroxide Eluent

The determination of inorganic anions (fluoride, chloride, nitrite, sulfate, bromide, nitrate, and phosphate) in municipal drinking water is one of the most important ion chromatography (IC) applications worldwide. In the United States (U.S.), water integrity is legislated through the Safe Drinking Water Act (SDWA), which ensures water quality and safety.[1] Other industrialized countries have similar regulations and, therefore, similar analytical needs.

Since the 1980s, with the approval of the U.S. Environmental Protection Agency (EPA) Method 300.0 (Part A), Dionex IC methods have been used for compliance testing of inorganic anion determinations. In 1993 and 1997, IC methods using Thermo Scientific Dionex IonPac™ AS4A and Dionex IonPac AS9-HC anion-exchange columns were specified in U.S. EPA Methods 300.0 (Part A) and 300.1 (Part A).[2,3] As advancements in column technology continued, new columns were proposed, such as the Dionex IonPac AS18 column in Dionex Application Note (AN) 154 in 2003.[4] However, both methods have run times exceeding 15 min.

In this study, mg/L concentrations of inorganic anions in a municipal drinking water sample were separated using a 2 × 150 mm, IonPac AS18-Fast anion-exchange column designed for fast separations using electrolytically generated hydroxide eluents. The Thermo Scientific Dionex ICS-5000 Reagent-Free™ IC (RFIC™) instrument was selected for this method to demonstrate the latest instrument technology. These results, shown in Figure 1, demonstrated the separation of mg/L concentrations of six anions in a municipal drinking water sample using hydroxide eluent at 0.45 mL/min. All anions were eluted from the column within 5 min and detected by suppressed conductivity detection.

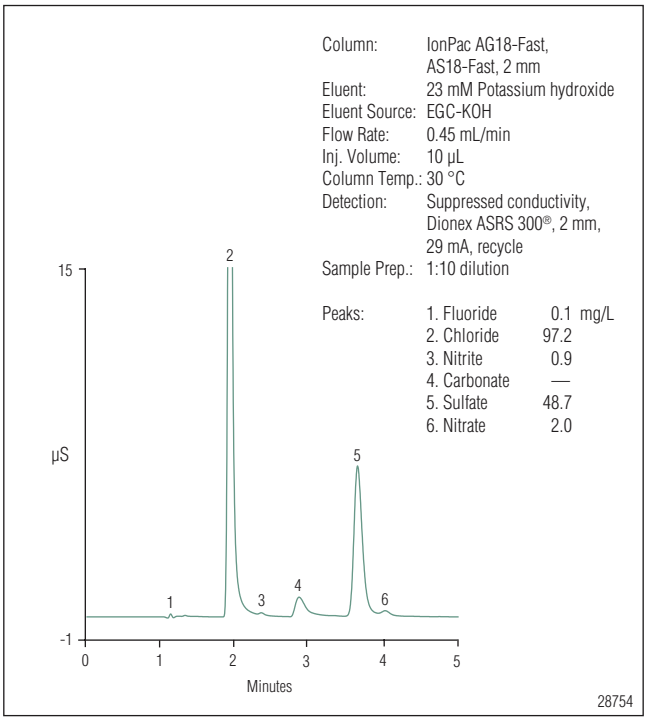

Figure 1. Determination of anions in a municipal drinking water sample by Fast IC on an IonPac AS18-Fast column using an RFIC system.

This method has the advantage of a fast separation which reduces cycle time, thereby resulting in a three-fold improvement in sample analyses over the methods specified in U.S. EPA Methods 300.0 and 300.1, saving time, money, and labor. The eluent is electrolytically generated in-line using just deionized water, which provides precise and accurate eluent concentration without additional time and labor spent on eluent preparation. The ICS-5000 system has the advantages of ease-of-use and flexibility to analyze at capillary, microbore, or standard flow rates. In this method, the ICS-5000 system was used at microbore flow rates to provide inorganic anions determinations needed for environmental compliance testing. Anion determinations using an electrolytically generated hydroxide eluent are thoroughly discussed in Dionex AN 154.[4]

CONDITIONS

An ICS-5000 RFIC system including a Thermo Scientific Dionex AS-AP Autosampler and Thermo Scientific Dionex Chromeleon™ Chromatography Data System software were used for all analyses. The chromatography conditions are listed in Figure 1.

SAMPLE PREPARATION

The municipal drinking water sample was filtered with a 0.45 µm IC syringe filter prior to analysis.

REFERENCES

1. *National Primary Drinking Water Regulations;* Code of Federal Regulations, 40 CFR, Part 141; U.S. Environmental Protection Agency: Cincinnati, OH, 1998.
2. *The Determination of Inorganic Anions in Water by Ion Chromatography*; Method 300.0, rev 2.1; U.S. Environmental Protection Agency: Cincinnati, OH, 1993.
3. *The Determination of Inorganic Anions in Water by Ion Chromatography*; Method 301.0, rev 1.0; U.S. Environmental Protection Agency: Cincinnati, Ohio, 1997.
4. Thermo Fisher Scientific, *Determination of Inorganic Anions in Environmental Waters Using a Hydroxide-Selective Column.* Application Note 154, LPN 1539, 2003, Sunnyvale, CA.

Determination of Nitrite and Nitrate in Drinking Water Using Chemically Suppressed Ion Chromatography

INTRODUCTION

Ion chromatography provides a convenient method for the determination of common inorganic anions in drinking water, including nitrite and nitrate. Using chemically suppressed ion chromatography, trace nitrite and nitrate are accurately and rapidly determined in drinking water. This includes water samples that have been preserved with sulfuric acid. The method will tolerate chloride levels up to 150 ppm and sulfuric acid concentrations as high as 0.5%. For most drinking water samples, the addition of 0.1% by volume sulfuric acid will acidify the sample to pH <2. This ion chromatographic method, using an IonPac® AS9, will tolerate levels of sulfate up to five times that amount.

Figure 1 shows a chromatogram of drinking water spiked to 3.3 ppm nitrite and 22.0 ppm nitrate. The sample is preserved with sulfuric acid to pH <2.

RECOMMENDED EQUIPMENT

Any Dionex Ion Chromatograph with a conductivity detector.

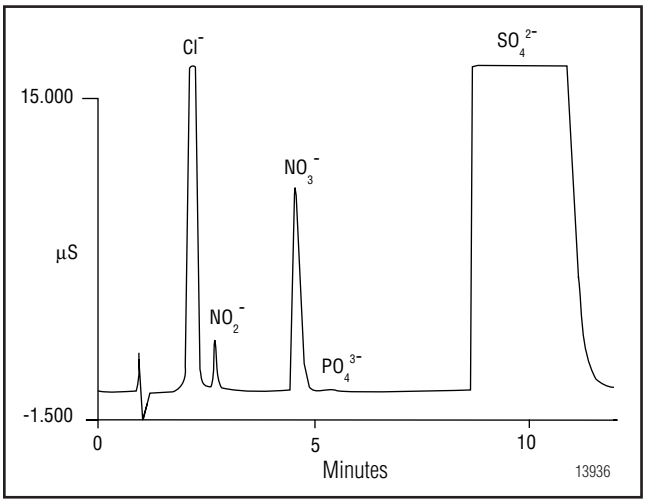

Figure 1 *Nitrite and nitrate in acid preserved drinking water.*

CONDITIONS

Column:	IonPac AS9 analytical with IonPac AG9 guard
Eluent:	1.8 mM Sodium carbonate 1.7 mM Sodium bicarbonate
Flow Rate:	2.0 mL/min
Suppressor:	Anion MicroMembrane™ Suppressor (AMMS®)
Regenerant:	25 mN Sulfuric acid
Regenerant Flow Rate:	6 mL/min using AutoRegen® accessory
Sample Volume:	25 µL
Detection:	Suppressed conductivity, 10 µS full scale

REAGENT AND STANDARD PREPARATION

Sodium carbonate/sodium bicarbonate concentrate (P/N 39513)

Suppressor regenerant concentrate (4 pack, P/N 37164)

Sodium nitrite, ACS Grade

Sodium nitrate, ACS Grade

ELUENT

To prepare 1.0-L of eluent, dilute 10.0 mL of eluent concentrate to 1000 mL with deionized water.

REGENERANT

To prepare 1.0-L of regenerant, dilute regenerant concentrate to 1000 mL with deionized water as directed on the regenerant concentrate label.

STOCK STANDARDS

1000 ppm Nitrite – Dissolve 1.499 g of sodium nitrite in 1.0 L of deionized water. 1000 ppm Nitrate – Dissolve 1.371 g sodium nitrate in 1.0 L of deionized water.

WORKING STANDARDS

Dilute the stock standards to concentration levels that bracket the concentration level of interest. Prepare working standards fresh from the stock standard prior to analysis.

PERFORMANCE CHARACTERISTICS

Detection limits in drinking water samples using a 25-µL loop are 10 ppb for nitrite and nitrate. This corresponds to 3.0 ppb nitrogen as nitrite, and 2.3 ppb nitrogen as nitrate. The method is linear for nitrite over the range 10 ppb to 50 ppm. It is linear for nitrate over the range of 20 ppb to 100 ppm.

Cost-Effective Determination of Inorganic Anions and Cations in Municipal Drinking Water Using Capillary Ion Chromatography

INTRODUCTION

The determination of common inorganic anions and cations in drinking water is one of the most important applications of ion chromatography (IC) worldwide. IC has been approved for compliance monitoring of inorganic anions in United States (U.S.) drinking water since the mid-1980s, as described in U.S. Environmental Protection Agency (EPA) Method 300.0.[1] Many other industrialized countries have similar health and environmental standards and a considerable number of regulatory IC methods have been published worldwide (e.g., in Germany, France, Italy, and Japan). In addition, many standards organizations, including the International Organization for Standardization (ISO), American Society for Testing and Materials (ASTM), and American Water Works Association (AWWA), have validated IC methods for the determination of inorganic anions in drinking water.[2,3] The concentration of some anions in drinking water are regulated due to their toxic effects. For example, high levels of fluoride can cause skeletal and dental fluorosis, and nitrite and nitrate can cause methemoglobulinemia, which can be fatal to infants. Other common anions, such as chloride and sulfate, are considered secondary contaminants and can affect odor, color, and certain aesthetic characteristics in drinking water.

IC methods for dissolved alkali and alkaline earth metals and ammonia in drinking water are also important. Drinking water is frequently monitored for the presence of sodium under the U.S. EPA Safe Drinking Water Act. Ammonium is commonly a required target analyte for wastewater discharge permits, and is monitored in process wastewaters.

This study describes the determination of inorganic anions and cations in drinking water using the Thermo Scientific Dionex ICS-5000 capillary IC system. Scaling down from standard bore to capillary scale brings many benefits to IC analysts. Capillary Thermo Scientific Dionex Reagent-Free™ IC systems deliver fast turnaround from sample submission to results by reducing eluent preparation, system startup, and equilibration times. Perhaps most importantly, the system can be left on and ready for analysis at any time because of its low consumption of eluent (15 mL of source water a day). Having the system always on and ready for analysis significantly streamlines the workflow in IC. An always on system maintains stability and requires less frequent calibrations. The amount of waste generated is significantly decreased and the eluent generation cartridge producing the eluent lasts 18 months under continuous operation mode, which translates into reduced overall cost of ownership.

Figure 1 shows the determination of inorganic anions in drinking water using capillary IC. The inorganic anions were separated on the Thermo Scientific Dionex IonPac® AS19 capillary column and detected by suppressed conductivity detection. All anions were separated and eluted within 13 min. The relative standard deviation of peak area for each analyte was 0.6% when 60 injections were evaluated within 24 h.

Figure 2 shows the determination of inorganic cations in drinking water using capillary IC. The inorganic cations were separated on the IonPac CS12A capillary column and detected by suppressed conductivity detection. All cations of interests were determined within 12 min.

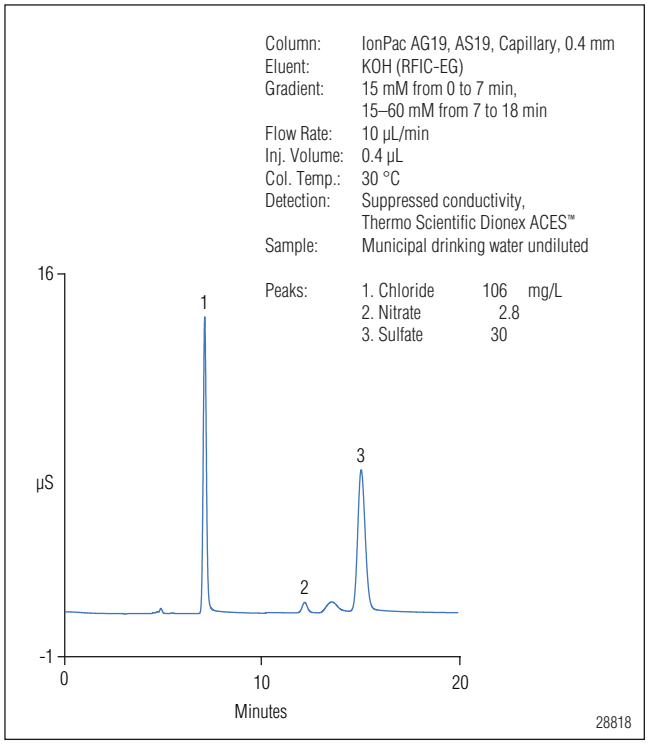

Figure 1. Anions in municipal drinking water on the IonPac AS19 capillary column.

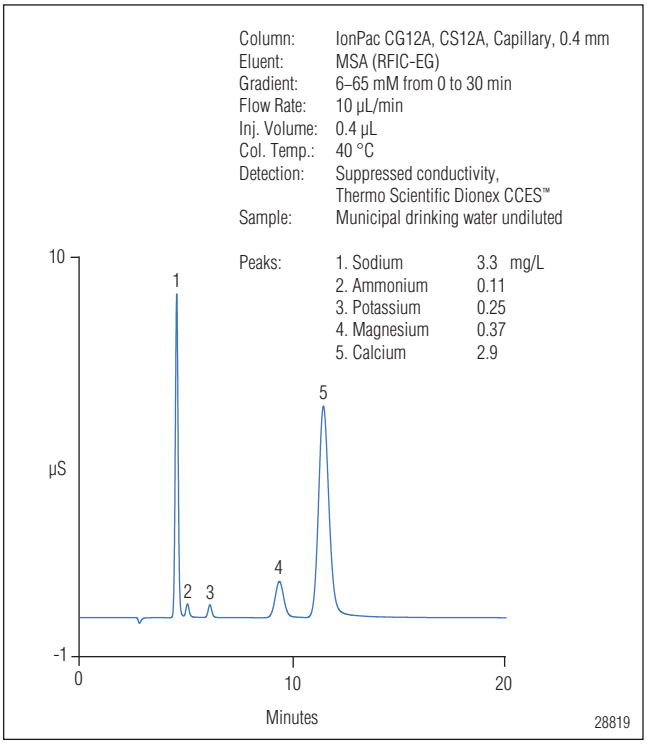

Figure 2. Cations in municipal drinking water on the IonPac CS12A capillary column.

CONDITIONS

The Dionex ICS-5000 capillary system, Thermo Scientific Dionex AS-AP Autosampler, and Thermo Scientific Dionex Chromeleon® Chromatography Data System software are used in this experiment. All experimental parameters are listed in Figures 1 and 2.

SAMPLE PREPARATION

Analyze municipal drinking water by capillary IC without sample pretreatment.

CONCLUSION

The introduction of the capillary Reagent-Free IC systems redefine the IC workflow for determination of inorganic anions and cations, providing enhanced mass sensitivity and ease of use.[4] These systems are a great solution for routine characterization of water samples with the always on, always ready capability simplifying the overall IC workflow.

REFERENCES

1. *The Determination of Inorganic Anions in Water by Ion Chromatography*; Method 300.0, Revision 2.1; U.S. Environmental Protection Agency: Cincinnati, OH, 1993.
2. Greenberg, A. E.; Clesceri, L. S.; Eaton, A. D., Eds.; *Standard Methods for the Examination of Water and Wastewater*, 18th ed.; American Public Health Association: Washington, DC, 1992.
3. *Standard Test Methods for Anions in Water by Chemically Suppressed Ion Chromatography*; D4327-97, Vol. 11.01; American Society for Testing and Materials; West Conshohocken, PA; 1999; pp 420–427.
4. Dionex Corporation, *Mass Sensitivity of Capillary IC Systems Explained.* Technical Note 90, LPN 2649, Sunnyvale, CA, 2011.

Municipal Drinking Water Analysis by Fast IC

INTRODUCTION

The determination of inorganic anions (fluoride, chloride, nitrite, sulfate, bromide, nitrate, and phosphate) in municipal drinking water is one of the most important ion chromatography (IC) applications worldwide. In the United States, water integrity is legislated through the Safe Drinking Water Act (SDWA), which ensures water quality and safety.[1] Other industrialized countries have similar regulations and, therefore, have similar analytical needs.

Since the 1980s, with the approval of EPA Method 300.0 (Part A), Dionex IC methods have been used for compliance testing of inorganic anions. In 1993 and 1997, Dionex IC methods using IonPac® AS4A and IonPac AS9-HC anion-exchange columns were specified in Methods 300.0 (Part A) and 300.1 (Part A).[2,3] As advancements in column technology continued, new columns were proposed, such as the IonPac AS14 column in AN 133 in 2004.[4] However, both methods have run times of more than 14 min.

In this study, mg/L concentrations of inorganic anions were separated on a 2 × 150 mm, IonPac AS22-Fast anion-exchange column designed for fast separations using carbonate eluents. Eluents were prepared by diluting the IonPac AS22 Reagent Concentrate for ease of use and to minimize eluent preparation errors. The results demonstrate the separation of mg/L concentrations of seven anions in a municipal drinking water sample using carbonate eluents at 0.5 mL/min (Figure 1). All anions were eluted from the column within 5 min and detected by suppressed conductivity detection with the ASRS® 300 Anion Self-Regenerating Suppressor.®

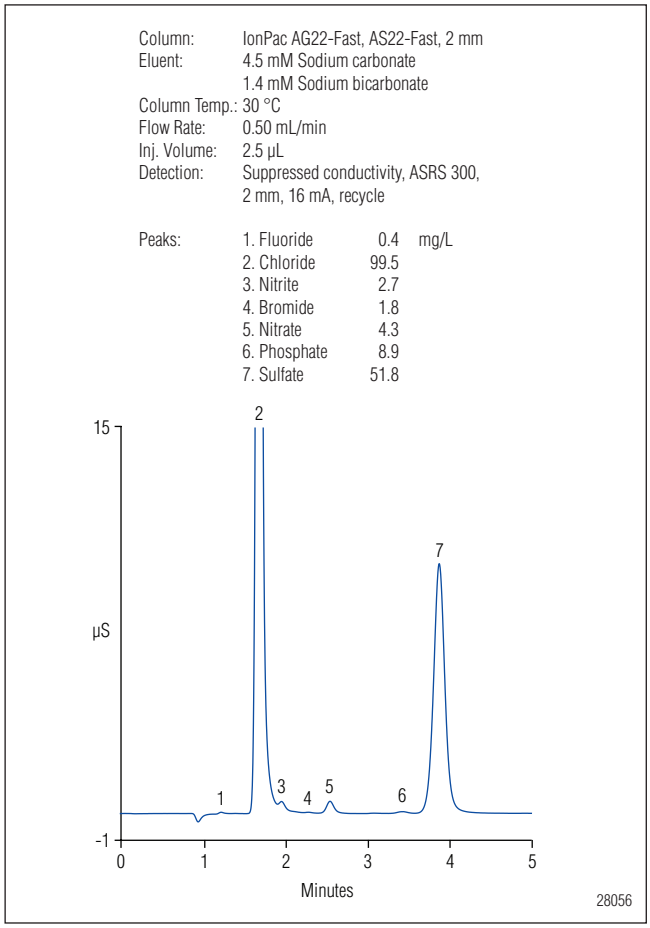

Figure 1. Determination of anions in a municipal drinking water sample by Fast IC on the IonPac AS22-Fast column.

This suppressor uses electrolytic suppression to provide low background noise and improve the accuracy of the results. This means faster equilibration times without regenerant preparation, which saves time and money.

CONDITIONS

A Dionex ICS-1100 Integrated IC system with electrolytic suppression, an AS Autosampler, and Chromeleon® Chromatography Data System (CDS) software were used for the analyses. The chromatography conditions are listed in Figure 1.

SAMPLE PREPARATION

The municipal drinking water sample was filtered with a 0.45 µm, IC syringe filter prior to analysis.

CONCLUSION

This method using the IonPac AS22-Fast column provides an equivalent method—using the latest column technology—to the EPA 300.0 (Part A) and 300.1 (Part A) approved methods while providing a 5 min run time that reduces cycle time, lowering the overall cost, saving time, and increasing the sample throughput. This method uses the ICS-1100 Integrated IC System but can be performed easily on any other Dionex IC system, including Reagent-Free™ IC (RFIC™) with eluent regeneration and eluent generation IC systems. Anion determinations in municipal drinking water using carbonate eluents are thoroughly discussed in AN 133.[4]

REFERENCES

1. Fed. Regist. **2010**; Vol. 75, No. 184.
2. *The Determination of Inorganic Anions in Water by Ion Chromatography*; Method 300.0, Revision 2.1; U.S. Environmental Protection Agency; Cincinnati, OH, 1993.
3. *The Determination of Inorganic Anions in Water by Ion Chromatography*; Method 301.0, Revision 1.0; U.S. Environmental Protection Agency; Cincinnati, OH, 1997.
4. *Dionex Corporation, Determination of Inorganic Anions in Drinking Water by Ion Chromatography*. Application Note 133, LPN 1192, Sunnyvale, CA, 2004.

Analysis of Cations

Environmental Water Applications Notebook

Monitoring Inorganic Anions and Cations During Desalination

INTRODUCTION

As of 2009, there were 14,450 desalination plants worldwide producing more than 60 million cubic meters of water a day.[1] Because of the growing demand for water and the limited supply of fresh water, desalination increasingly is being used to produce potable and irrigation water from salty or brackish water. The global market for desalination to generate supplies of potable water is projected to grow at an annual rate of 10% over the next 10 years. Seawater desalination is a $10 billion industry today and is forecasted to reach $16 billion in 2020.[1]

A wide variety of desalination techniques are currently available and more are being developed. Most use distillation or membrane techniques. The performance of desalination processes is evaluated by monitoring the common anions and cations in the feed, intermediate, and final water product. For the final drinking water product, ion chromatography (IC) is approved for monitoring primary and secondary anions according to U.S. Environmental Protection Agency (EPA) method 300.0,[2] and Federal and State regulatory agencies ensure that U.S. National Primary and Secondary Drinking Water Standards are met.

Common cations, though not considered contaminants, are monitored and reported by many public water suppliers in the United States. Cations, particularly calcium and magnesium, are measured to determine water hardness. In addition to calcium and magnesium, ammonium is also measured and regulated in public water supplies in EU countries and Japan. During desalination, the levels of divalent cations affect performance of membrane processes like reverse osmosis (RO).[3] High levels of calcium or magnesium result in frequent fouling of the membranes, which is highly undesirable. Therefore, it is critical to monitor anions and cations at all stages of desalination.

Another challenge for the desalination of seawater is the removal of boron, which is typically found at concentrations of 4.5 mg/L. World Health Organization 2008 guidelines suggest a concentration of 0.5 mg/L,[4] whereas the U.S. EPA recommends a maximum lifetime exposure of 0.6 mg/L.[5] Depending on pH levels, boron can exist in ionic and non-ionic forms. Above pH 8, the removal efficiency using RO is enhanced due to the formation of borate. RO membranes remove ions better than non-ionic forms of the same compounds. This suggests that raising the pH may improve the removal efficiency of boron. However, raising the pH too high results in the formation of scales formed by the precipitation of carbonate salts of calcium and magnesium, which can disrupt membrane performance. In addition to monitoring the pH, it is important to know the concentration of scale-forming divalent cations calcium and magnesium in order to maintain optimal RO membrane performance.

Compared to traditional sources of water, desalination is an energy-intensive process that requires expensive infrastructure. The potential benefits of desalination are constantly being evaluated because of the high economic and environmental costs. Hence, efficient water monitoring techniques are needed to understand the robustness of desalination processes.

This work describes an IC method using a Dionex ICS-3000 system with IonPac® AS18 anion-exchange and CS12A cation-exchange columns, electrolytically generated hydroxide and methanesulfonic acid eluents, and suppressed conductivity detection to simultaneously measure the common anions and cations in water samples obtained from desalination processes. This method uses a 2 mm column format for anion separations, a 3 mm column format for cation separations, and electrolytically generated eluents that require only the addition of deionized water for continuous operation. The linearity, method detection limits (MDLs), precision, and recovery of anions and cations in saline and drinking water matrices for this method are discussed here. This IC method supports all the monitoring needs of a desalination facility because it can measure anions and cations in diverse matrices ranging from seawater to drinking water.

EQUIPMENT

Dionex ICS-3000 Reagent-Free™ Ion Chromatography system* with eluent generation (RFIC-EG™) including:

DP Dual Pump module

EG Eluent Generator module

DC Detector/Chromatography module (single- or dual-temperature zone configuration)

AS Autosampler (with Simultaneous Injection Upgrade Kit, Dionex P/N 063742)

EluGen EGC II KOH cartridge (Dionex P/N 058900)

Continuously-Regenerated Anion Trap Column, CR-ATC II (Dionex P/N 060477)

EluGen EGC II MSA cartridge (Dionex P/N 058902)

Continuously-Regenerated Cation Trap Column, CR-CTC II (Dionex P/N 066262)

Chromeleon® 6.8 or 7 Chromatography Workstation

Polystyrene Autoselect™ vials with caps and septa, 10 mL (Dionex P/N 055058)

Nalgene® 125 mL HDPE narrow mouth bottles (VWR P/N 16057-062)

Nalgene 250 mL HDPE narrow mouth bottles (VWR P/N 16057-109)

Nalgene 250 mL 0.2 µm nylon filter units (VWR P/N 28199-371)

Nalgene 1000 mL 0.2 µm nylon filter units (VWR P/N 28198-514)

*The applications described here can run on any Dionex RFIC system. The applications also can run with manually prepared eluents on any Dionex IC system.

REAGENTS AND STANDARDS

Deionized water, Type I reagent grade, 18 MΩ-cm resistivity or better, filtered through a 0.2 µm filter immediately before use

Fluoride Standard 1000 mg/L (Dionex P/N 037158)

Chloride Standard 1000 mg/L (Dionex P/N 037159)

Nitrite Standard 1000 mg/L (UltraScientific P/N ICC-007)

Bromide Standard 1000 mg/L (UltraScientific P/N ICC-001)

Sulfate Standard 1000 mg/L (UltraSceintific P/N ICC-006)

Nitrate Standard 1000 mg/L (UltraScientific P/N ICC-004)

Phosphate Standard 1000 mg/L (UltraScientific P/N ICC-005)

Lithium Standard 1000 mg/L (UltraScientific P/N ICC-104)

Sodium Standard 1000 mg/L (UltraScientific P/N ICC-107)

Ammonium Standard 1000 mg/L (UltraScientific P/N ICC-101)

Potassium Standard 1000 mg/L (UltraScientific P/N ICC-106)

Magnesium Standard 1000 mg/L (UltraScientific P/N ICC-105)

Calcium Standard 1000 mg/L (UltraScientific P/N ICC-104)

Sodium Chloride (J.T. Baker P/N 4058-05)

Sodium Sulfate (VWR P/N EM-SX0760-1)

Sodium Nitrite (JT Baker P/N 1-3780)

Sodium Bromide (Aldrich P/N 31050-6)

Sodium Nitrate (Baker P/N 3770-05)

Potassium Phosphate Monobasic (Fisher P/N P286-1)

Lithium Chloride (Fisher, P/N L-121-100)

Ammonium Chloride (Sigma A-5666)

Potassium Chloride (Mallinckrodt P/N 6858)
Magnesium Chloride Hexahydrate (BDH P/N 0244 5009)
Calcium Chloride Dihydrate (Fisher P/N C-79)
Combined Six Cation Standard-II (Dionex P/N 046070)
Combined Seven Anion Standard (Dionex P/N 66933)

CONDITIONS

Anion Determinations

Columns:	IonPac AG18, 2 × 50 mm IonPac AS18, 2 × 250 mm
Eluent:	22 mM KOH from 0–7 min, 22–40 mM KOH from 7–8 min, 40 mM KOH from 8–18 min*
Eluent Source:	EGC II KOH with CR-ATC
Injection Volume:	4 µL
Flow Rate:	0.25 mL/min
Detection:	Suppressed conductivity, ASRS® 300, 2 mm, recycle mode, suppressor current 15 mA
Background Conductance:	<1 µS

Cation Determinations

Columns:	IonPac CG12A-5 µm, 3 × 30 mm IonPac CS12A-5 µm, 3 × 150 mm
Eluent:	20 mM MSA
Eluent Source:	EGC II MSA with CR-CTC
Injection Volume:	10 µL
Flow Rate:	0.50 mL/min
Detection:	Suppressed conductivity, CSRS® 300, 2 mm, recycle mode, suppressor current 30 mA
Background Conductance:	<0.5 µS

Both Anion and Cation Determinations

Temperature:	30 °C (column and detector compartment)
Noise:	~0.5–1.0 nS (conductivity)
System Backpressure:	~2500 psi
Run Time:	20 min (including column equilibration time)

*The column equilibrates for 2 min at 22 mM KOH prior to injection.

PREPARATION OF SOLUTIONS AND REAGENTS

Eluent Solutions

Generate potassium hydroxide (KOH) and methanesulfonic acid (MSA) eluents online by pumping high-quality degassed, deionized (DI) water through the EGC II KOH and MSA cartridges, respectively. Chromeleon Chromatography Data System (CDS) software tracks the amount of KOH and MSA used and calculates the remaining lifetime. Although electrolytic eluent generation delivers the best performance, manually prepared eluents may be used, if needed.

Stock Standard Solution

Certified standard solutions can be purchased from Dionex or other commercial sources. When commercial standards are not available, 1000 mg/L stock standard solutions can be prepared by dissolving appropriate amounts of the required analyte in DI water in a plastic volumetric flask (Table 1). Store in plastic containers at 4 ºC. Stock standards are stable for at least 3 months.

Working Standard Solutions

Prepare composite working standards at lower analyte concentrations by diluting appropriate volumes of the 1000 mg/L stock with DI water. Prepare working standards containing < 100 mg/L anions or cations daily. Store standard solutions at < 6 °C when not in use.

Table 1. Mass of Compound Required to Prepare 1L of 1000 mg/L Stock Standard Solutions

Analyte	Compound	Amount (g)
Fluoride	Sodium fluoride (NaF)	2.210
Chloride	Sodium chloride (NaCl)	1.648
Nitrite	Sodium nitrite ($NaNO_2$-N)	4.926
Bromide	Sodium bromide (NaBr)	1.288
Nitrate	Sodium nitrate ($NaNO_3$-N)	6.068
Sulfate	Sodium sulfate (Na_2SO_4)	1.479
Phosphate	Potassium phosphate monobasic (KH_2PO_4-P)	4.394
Lithium	Lithium chloride (LiCl)	6.108
Sodium	Sodium chloride (NaCl)	2.542
Ammonium	Ammonium chloride (NH_4Cl)	2.965
Potassium	Potassium chloride (KCl)	1.907
Magnesium	Magnesium chloride hexahydrate ($MgCl_2 \cdot 6H_2O$)	8.365
Calcium	Calcium chloride dihydrate ($CaCl_2 \cdot 2H_2O$)	3.668

SAMPLE PREPARATION

Artificial Seawater

Prepare simulated seawater by diluting the salts listed in Table 2 into 1 L of DI water following the method of Kester et al.[6] with the exclusion of strontium chloride. This yields a solution with approximately 3.5% salinity.

Table 2. Salts Added to Form Simulated Seawater (1L)

Compound	Amount (g)
Sodium chloride	2.393
Sodium sulfate	4.008
Potassium chloride	0.677
Sodium bicarbonate	0.196
Potassium bromide	0.098
Boric acid	0.026
Sodium fluoride	0.003

Commercial Aquarium Sea Salt

Follow package directions (1/2 cup of salt per gallon of DI water) to prepare commercially available synthetic sea salt, creating a solution of approximately 3.5% salinity. Prepare a 1 L portion with 30 g of aquarium salt. (A sea salt density of approximately 2.2 g/cm^3 was used to convert the preparation directions to metric units.[7])

Seawater (From California's San Francisco Bay)

Collect surface seawater in a 250 mL HDPE Nalgene bottle that has been precleaned before sample collection. Store the sample on ice until it can be filter sterilized through a 250 mL, 0.2 µm nylon filter unit. After filtration, store the sample at < 6 °C.

Filter all samples through a 0.2 µm nylon filter unit before injection.

SYSTEM PREPARATION AND CONFIGURATION

Configure the autosampler (AS) for simultaneous injection into the anion and cation detection systems. In the simultaneous mode, the AS delivers sample to two independent IC systems. The sample is injected simultaneously and equally to both systems (two injection valves are required). Dual analyses can be performed with only one sample. A 5 or 10 mL syringe and an 8.5 mL sampling needle assembly are required for simultaneous injections. Full-loop injections are required for this mode.

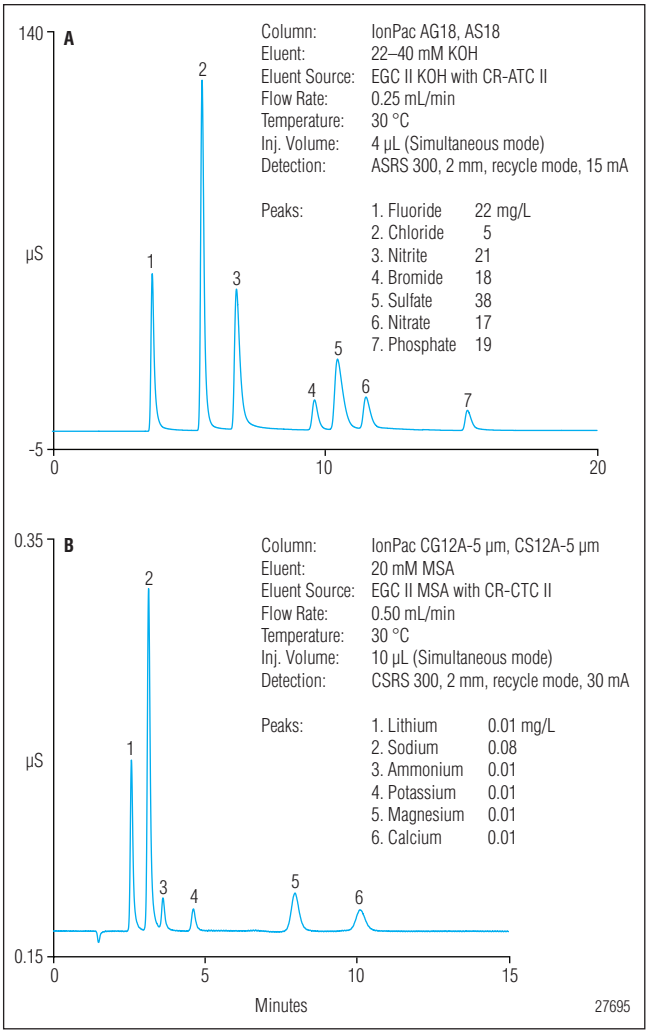

Figure 1. Separation of common A) anions using the IonPac AS18 column and B) cations using the IonPac CS12A column.

Using Chromeleon software, configure the two chromatography systems and the AS into a single timebase and assign each system a unique device name and channel. Use one control panel to monitor and control both systems and all samples in one sequence.

The system also may be configured for sequential injection into the two IC systems. In the sequential option, the sample is delivered to the first system, flow is rerouted (diverted), and then sample is delivered to the second system.[8]

RESULTS AND DISCUSSION

Using the IonPac CS12A and AS18 columns, the common anions and cations were easily resolved in 20 min (Figure 1 A and B). Note that this method provided good resolution between sodium and ammonium, the two analytes that can be challenging to resolve, especially

Table 3. Linear Range, MDLs, and Precisions for Anions and Cations							
Analyte	**Range (mg/L)**	**Corr. Coeff. (r^2)**	**MDL Standard (µg/L)**	**MDL (µg/L)[a]**	**QCS (mg/L)**	**Retention Time Precision (RSD)[b]**	**Peak Area Precision (RSD)[b]**
Lithium	0.02–16	0.9999	1	0.08	1	0.07	0.20
Sodium	0.10–100	0.9999	4	0.13	4	<0.01	0.29
Ammonium[c]	0.01–8	0.9997	5	0.10	5	<0.01	0.97
Potassium	0.02–16	0.9997	10	0.10	10	0.03	0.61
Magnesium	0.02–80	0.9998	5	0.53	5	0.03	0.09
Calcium	0.02–80	0.9999	5	0.36	5	0.04	0.15
Fluoride	0.08–100	0.9996	10	0.62	2	0.05	0.05
Chloride	0.24–300	0.9999	10	0.66	20	0.02	0.04
Nitrite (-N)	0.08–100	0.9994	20 (67 as NO_2^-)	0.51 (-N)	2 (6.7 as NO_2^-)	0.03	0.07
Bromide	0.08–100	0.9994	25	0.46	2	0.02	0.05
Sulfate	0.16–201	0.9994	20	0.67	60	0.01	0.07
Nitrate (-N)	0.02–22	0.9999	27 (120 as NO_3^-)	0.24 (-N)	2.3 (10 as NO_3^-)	0.02	0.08
Phosphate (-P)	0.03–33	0.9999	23 (70 as PO_4^{3-})	0.15 (-P)	0.7 (2 as PO_4^{3-})	0.04	0.14

[a]MDL = (t) × (S) where t = Student's t value for a 99% confidence level and a standard deviation estimate with n-1 degrees of freedom [t = 3.14 for seven replicates of the MDL Standard], and S = standard deviation of the replicate analysis

[b]Relative standard deviation, n = 7

[c]Quadratic fit

when one is in a large excess relative to the other. This method also achieved good retention time for fluoride, which was well resolved from the void volume.

Table 3 summarizes the calibration data, the method detection limits (MDLs), retention time, and peak area precisions for the common anions and cations. MDLs and precision data were obtained from seven replicate injections of the MDL and QCS standards, respectively, prepared in DI water. Anion and cation MDL standards were prepared at concentrations of 3–5× the estimated method detection limits.

Correlation coefficient values obtained from the calibration plots were between 0.9994 and 0.9999. The calibration curves were linear for all anions and cations except ammonium. Analytes that form weak acids or bases in the suppressor are known to exhibit nonlinear behavior. A quadratic curve-fitting function was used for ammonium. The retention time precision ranged from < 0.01–0.07%, and the peak area precision ranged from 0.04–0.97%. The high retention time precisions are attributed to consistent generation of high-purity KOH and MSA using the eluent generator module and the respective continuously regenerated trap columns (CR-ATC and CR-CTC).

Table 4. Between-Day (n=3) Retention Time (RT) and Peak Area Precisions (Triplicate Injections of Spiked Sample)									
Cations					**Anions**				
San Francisco Bay Water									
Analyte	**RT (min)**	**RT Precision (RSD)**	**Peak Area (µS*min)**	**Peak Area Precision (RSD)**	**Analyte**	**RT (min)**	**RT Precision (RSD)**	**Peak Area (µS*min)**	**Peak Area Precision (RSD)**
Lithium	2.57	<0.01	0.72	0.13	Fluoride	3.63	<0.01	0.39	0.57
Sodium	3.19	0.06	18.4	0.07	Chloride	5.45	0.00	40.3	0.31
Ammonium	3.61	0.02	0.32	0.20	Nitrite	6.77	0.02	0.61	0.64
Potassium	4.59	0.03	0.51	0.40	Bromide	9.63	0.03	0.14	0.82
Magnesium	7.87	0.02	4.31	0.12	Sulfate	10.53	0.01	5.14	0.20
Calcium	9.96	0.02	0.91	0.38	Nitrate	11.57	0.01	0.26	0.63
					Phosphate	15.44	0.03	0.09	0.65
Commercial Aquarium Sea Salt									
Lithium	2.57	0.05	0.72	0.15	Fluoride	3.63	0.05	0.37	0.52
Sodium	3.19	0.02	18.4	0.05	Chloride	5.45	0.02	40.2	0.18
Ammonium	3.61	<0.01	0.29	0.54	Nitrite	6.77	0.02	0.63	0.79
Potassium	4.59	0.03	0.52	0.30	Bromide	9.63	0.01	0.14	0.63
Magnesium	7.91	0.02	4.32	0.09	Sulfate	10.54	0.03	5.11	0.26
Calcium	10.08	0.01	0.88	0.41	Nitrate	11.57	0.01	0.26	0.68
					Phosphate	15.45	0.02	0.09	1.01
Artificial Seawater									
Lithium	2.57	0.05	0.71	0.22	Fluoride	3.63	< 0.01	0.37	0.39
Sodium	3.20	0.04	21.2	0.05	Chloride	5.45	0.01	45.83	0.22
Ammonium	3.61	<0.01	0.31	0.52	Nitrite	6.77	0.02	0.62	0.61
Potassium	4.59	0.03	0.58	0.37	Bromide	9.63	0.01	0.15	0.62
Magnesium	7.91	0.01	4.56	0.36	Sulfate	10.52	0.02	3.92	0.27
Calcium	10.07	0.03	0.94	0.50	Nitrate	11.57	0.01	0.27	0.24
					Phosphate	15.44	0.02	0.09	0.55

Method performance was evaluated by measuring recoveries in samples of spiked saline (Table 4) and drinking water (Table 5). Samples were spiked with analytes at a level that was 50–100% of the amount determined in the original sample. The between-day precision for anions and cations in the spiked samples ranged from < 0.01–1.6% over three days.

Table 5. Between-Day (n=3) Retention Time (RT) and Peak Area Precisions (Triplicate Injections of Spiked Sample)									
Cations					**Anions**				
Tap Water									
Analyte	**RT (min)**	**RT Precision (RSD)**	**Peak Area (µS*min)**	**Peak Area Precision (RSD)**	**Analyte**	**RT (min)**	**RT Precision (RSD)**	**Peak Area (µS*min)**	**Peak Area Precision (RSD)**
Lithium	2.57	0.05	0.75	0.13	Fluoride	3.63	0.04	0.82	0.34
Sodium	3.16	0.04	8.34	0.10	Chloride	5.47	0.01	7.47	0.32
Ammonium	3.61	0.02	0.48	0.14	Nitrite	6.78	0.01	0.29	0.46
Potassium	4.59	<0.01	0.27	0.15	Bromide	9.64	0.02	0.12	0.76
Magnesium	7.91	0.02	4.11	0.19	Sulfate	10.53	0.03	5.13	0.34
Calcium	9.97	0.01	6.57	0.18	Nitrate	11.56	0.01	0.31	0.66
					Phosphate	15.44	0.01	0.09	0.89
Bottled Mineral Water									
Lithium	2.57	0.03	0.75	0.13	Fluoride	3.63	0.04	0.90	0.49
Sodium	3.16	<0.01	8.57	0.07	Chloride	5.47	0.01	6.12	0.33
Ammonium	3.61	0.04	0.29	0.54	Nitrite	6.76	0.02	0.22	0.65
Potassium	4.59	0.01	0.61	0.09	Bromide	9.61	0.01	0.12	0.76
Magnesium	7.80	0.02	11.4	0.12	Sulfate	10.44	0.03	8.52	0.50
Calcium	9.73	0.01	18.5	0.21	Nitrate	11.52	0.02	0.74	0.34
					Phosphate	15.51	0.03	0.09	0.53

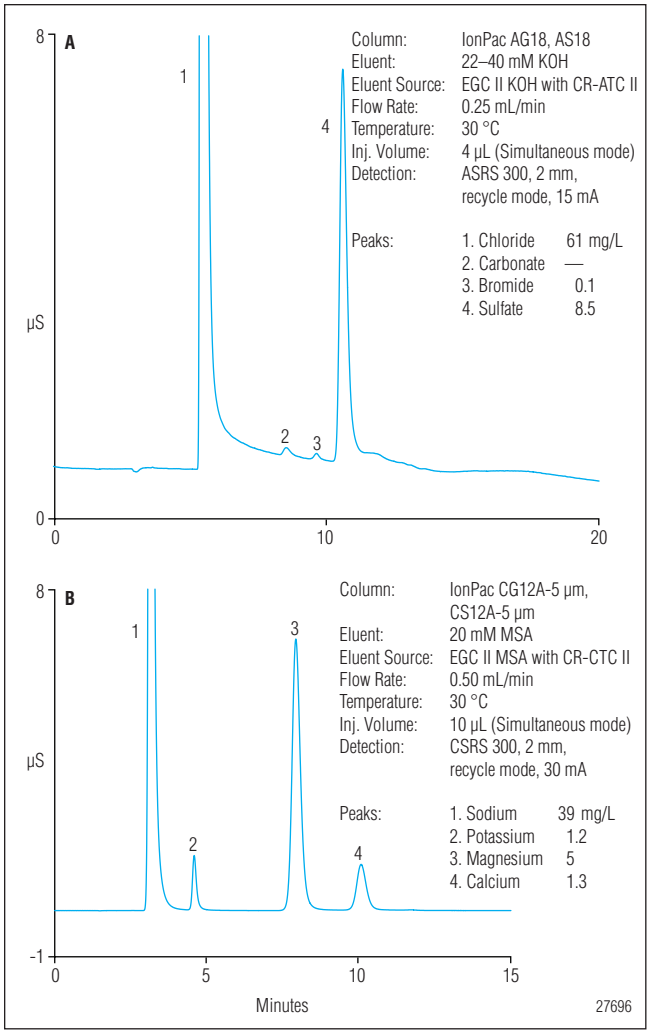

Figure 2. San Francisco, CA bay water: determination of common A) inorganic anions using the IonPac AS18 column and B) cations using the IonPac CS12A column.

Figure 2 shows the separation of A) anions and B) cations in water from California's San Francisco Bay. The bay water sample is representative of the typical feed water into a desalination plant. The bay water sample was diluted 200-fold so that measured levels were within the calibrated range. The major inorganic anions in bay water are chloride and sulfate, and the major inorganic cations are sodium, potassium, magnesium, and calcium.

Table 6. Inorganic Anion and Cation Recoveries										
Analyte	**San Francisco Bay Water**		**Artificial Seawater**		**Commercial Aquarium Sea Salt**		**Bottled Mineral Water**		**Tap Water**	
	Amount Added (mg/L)	**Recovery* (%)**	**Amount Added (mg/L)**	**Recovery* (%)**	**Amount Added (mg/L)**	**Recovery* (%)**	**Amount Added (mg/L)**	**Recovery* (%)**	**Amount Added (mg/L)**	**Recovery* (%)**
Lithium	1	93.5	1	92.4	1	98.1	1.0	96.9	1	97.2
Sodium	40	89.9	40	95.3	40	90.9	20.1	93.1	20	96.6
Ammonium	1	108.3	1	105.7	1	105.9	1.0	97.9	1	99.9
Potassium	2	94.5	2	96.7	2	99.6	1.0	94.9	1	95.7
Magnesium	5	97.2	5	97.4	5	97.6	5.0	94.6	5	97.1
Calcium	2	88.4	2	83.7	2	82.8	15.0	95.7	15	85.6
Fluoride	1	109.6	1	107.0	1	106.9	1	98.1	1	98.7
Chloride	74	87.2	74	92.7	74	89.7	16	84.2	16	85.9
Nitrite (-N)	1	100.7	1	101.7	1	105.3	1	37.2	1	41.5
Bromide	1	84.4	1	83.6	1	84.1	1	87.9	1	88.4
Sulfate	16	81.2	16	83.9	16	84.9	16	93.9	16	84.4
Nitrate (-N)	0.2	98.4	0.2	103.7	0.2	100.0	0.2	100.9	0.2	101.3
Phosphate (-P)	0.3	88.0	0.3	84.3	0.3	86.6	0.3	88.4	0.3	85.0

*Average over 3 days

As seen in Figure 2 and Table 6, all anions and cations were well resolved and had acceptable recoveries (80–120%) using the criteria outlined in U.S. EPA Method 300.0.

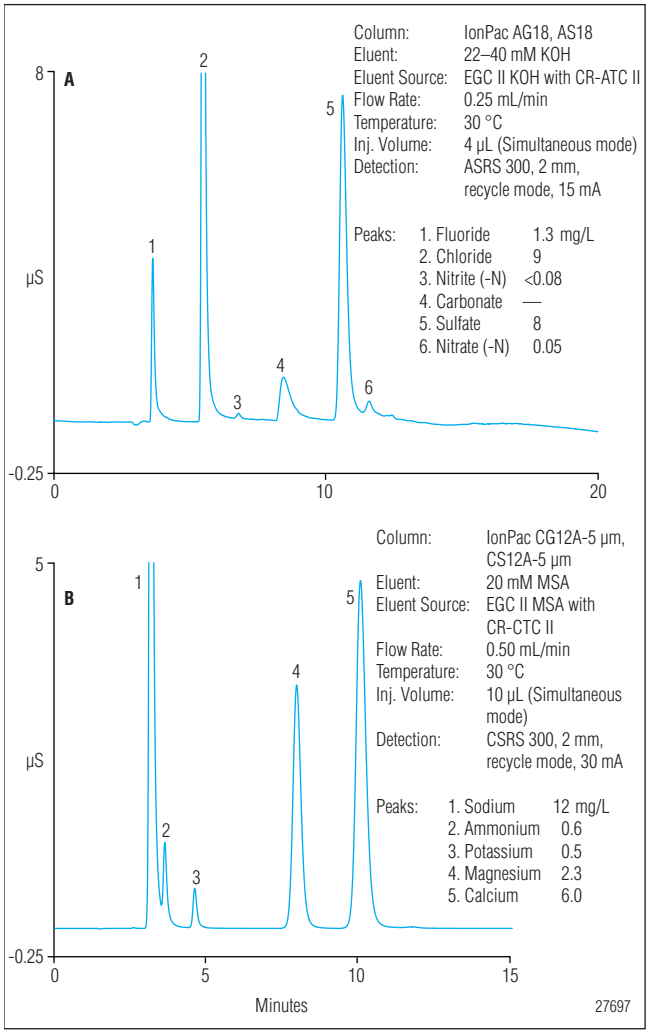

Figure 3. Sunnyvale, CA tap water: determination of common A) inorganic anions using the IonPac AS18 column and B) cations using the IonPac CS12A column.

Figure 3 (A and B) shows the chromatogram for all anions and cations in Sunnyvale, CA drinking water. Tap water samples have fluoride, chloride, and sulfate as the predominant inorganic anions. Table 6 lists the recoveries of anions and cations in the drinking water matrices. All anions and cations were well resolved and, with the exception of nitrite, had acceptable recoveries (80–120%). The low recovery of nitrite can be attributed to biological activity in these samples (which is minimal in the high saline matrices) and the instability of nitrite in oxidizing environments, such as chlorinated water or other oxidizing disinfectants in drinking water.

In summary, the current methods using the IonPac AS18 and CS12A columns provide acceptable recoveries for anions and cations in both saline and drinking water matrices. This work shows methods that can be used for diverse matrices that are typically encountered in a desalination plant.

CONCLUSION

IonPac AS18 and CS12A columns with electrolytically generated hydroxide and MSA eluents can simultaneously determine anions and cations in saline and drinking water matrices. The capacities of the IonPac AS18 and CS12A columns allow sample analysis with minimal sample pretreatment. The RFIC-EG system allows continuous operation of the instrument with minimal maintenance. Only water for eluent generation and suppressor regeneration must be added to keep the instrument running for sample analysis. Additionally, the smaller column format generates less waste and uses less eluent, saving both time and money. The methods were shown to be accurate by the good recovery of anions and cations in a wide variety of samples including natural and artificial seawater and drinking water. These methods are robust for all ion-monitoring needs of a typical desalination facility and support a varying range of matrices from seawater to drinking water.

SUPPLIERS

Fisher Scientific, 2000 Park Lane Drive, Pittsburgh, PA 15275, U.S.A. Tel: 800.766.7000. www.fishersci.com

VWR, 1310 Goshen Parkway, West Chester, PA 19380, U.S.A. Tel: 800-932-5000. www.vwr.com

Sigma-Aldrich Chemical Co., P.O. Box 2060, Milwaukee, WI 53201, U.S.A. Tel: 800-558-9160. www.sigmaaldrich.com

ULTRA Scientific, 250 Smith Street, N. Kingstown, RI 02852. U.S.A. Tel : 800-338-1754. www.ultrasci.com

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Determination of Inorganic Cations and Ammonium in Environmental Waters by Ion Chromatography Using the IonPac® CS16 Column

INTRODUCTION

The common alkali and alkaline earth cations are not considered primary drinking water contaminants in the U.S.; however they are monitored and reported by many public water suppliers here and are regulated in the EU and Japan. Calcium and magnesium are also routinely measured to determine water hardness, an important parameter for corrosion control.

Ammonia is a colorless, pungent gas. It is highly soluble in water, where it exists in equilibrium between a molecular form associated with water and the ionized form (the ammonium cation, NH_4^+). The extent of its toxicity to aquatic life depends upon the extent of dissociation, which in turn depends upon temperature and pH. Ammonia can enter environmental waters as a product of anaerobic decomposition of nitrogen-containing compounds or from waste streams containing ammonia. Ammonium cation is routinely measured in the U.S. for wastewater discharge compliance monitoring and in the EU and Japan in both wastewater and drinking water.[1]

Alkali and alkaline earth cations are commonly determined by using spectroscopic techniques such as AAS or ICP, but ammonium cation in the same sample must be measured separately by a wet chemical technique such as titrimetry, colorimetry (Nesslerization, phenate, or automated phenate methods), or ammonia-selective electrode.[2] Furthermore, the latter two methods may also require a separate distillation step before ammonia can be determined in wastewater. Ion chromatography (IC) in a single run can determine ammonium plus all the important inorganic cations, including lithium, sodium, potassium, magnesium, and calcium.

The IonPac CS16 is a high-capacity cation exchange column with 100% solvent compatibility and medium hydrophobicity. The high capacity of 8400 µeq/column is achieved by using a smaller bead diameter (5 µm), a higher density of grafted carboxylic acid cation exchange groups, and a larger column format. The higher capacity improves performance for trace-level determinations of cations in high ionic strength matrices by extending the linear range and resolving higher concentration ratios of sodium and ammonium.

This Application Note describes the use of ion chromatography with a Dionex IonPac CS16 cation exchange column, an electrolytically generated methanesulfonic acid (MSA) eluent, and suppressed conductivity detection to determine dissolved alkali and alkaline earth cations and ammonium in drinking water, wastewater, and aqueous soil extracts. This Note discusses the linear range, method detection limits, and analyte recoveries obtained with the IonPac CS16 and evaluates the effect of potential interferences on method performance during the analysis of typical environmental samples.

EQUIPMENT

Dionex DX-600 chromatography system consisting of:

GP50 Gradient Pump with vacuum degas option

EG40 Eluent Generator

EluGen EGC-MSA cartridge (Dionex P/N 053922)

ED50A Electrochemical Detector with conductivity cell and DS3 Detector Stabilizer

AS50 Automated Sampler with thermal compartment

PeakNet® 6.1 Chromatography Workstation

Syringe filters (IC Acrodisk, Gelman P/N 4483 or Anotop IC, Whatman P/N 6809 9232)

REAGENTS AND STANDARDS

Deionized water, Type I reagent grade, 17.8 MΩ-cm resistivity or better

Lithium standard, 1000 mg/L (Ultra Scientific; VWR P/N ULICC 104)

Sodium standard, 1000 mg/L (Ultra Scientific; VWR P/N ULICC 107)

Ammonium standard, 1000 mg/L (Ultra Scientific; VWR P/N ULICC 101)

Potassium standard, 1000 mg/L (Ultra Scientific; VWR P/N ULICC 106)

Magnesium standard, 1000 mg/L (Ultra Scientific; VWR P/N ULICC 105)

Calcium standard, 1000 mg/L (Ultra Scientific; VWR P/N ULICC 103)

Lithium chloride (LiCl; Fisher L-121-100)

Sodium chloride (NaCl; Fisher S-271)

Ammonium chloride (NH_4Cl; Sigma A-5666)

Potassium chloride, (KCl; Sigma P-3911)

Magnesium chloride hexahydrate ($MgCl_2 \cdot 6H_2O$; Aldrich 24,696-4)

Calcium chloride dihydrate ($CaCl_2 \cdot 2H_2O$; Fisher C79-500)

Combined Six Cation Standard-II (Dionex P/N 046070)

CONDITIONS

Columns:	IonPac CS16 Analytical, 5 x 250 mm (Dionex P/N 079805) IonPac CG16 Guard, 5 x 50 mm (Dionex P/N 057574)
Eluent:	26 mM MSA
Eluent Source:	EG40
Flow Rate:	1.5 mL/min
Temperature:	30 °C
Injection:	10 µL
Detection:	Suppressed conductivity, CSRS®-ULTRA (4 mm), Autosuppression Recycle mode, current setting 100 mA
Background:	< 1 µS
Noise:	~0.2 nS peak-to-peak
Backpressure:	~2300 psi
Run Time:	30 min

PREPARATION OF SOLUTIONS AND REAGENTS

Reagent Water

Type I reagent grade distilled or deionized water with a specific resistance of 17.8 MΩ-cm or greater, filtered through a 0.2-µm filter immediately before use.

Eluent Solution

Generate 26 mM MSA eluent on-line by pumping reagent water through the EG40/EGC-MSA. PeakNet software tracks the amount of MSA used and calculates the remaining lifetime.

Alternatively, prepare 26 mM MSA by diluting 65 mL of 0.4 N Methanesulfonic Acid Eluent Concentrate (Dionex P/N 057562) to 1.0 L with reagent water. Degas the eluent by sonicating under vacuum for 10 minutes or by sparging with helium. Store the eluent in plastic labware.

Or, prepare a 1.0 N MSA stock solution. Carefully add 96.10 g of methanesulfonic acid (MSA, > 99%, Dionex P/N 033478) to a 1-L volumetric flask containing about 500 mL of deionized water. Dilute to the mark and mix thoroughly. Prepare 26 mM MSA by diluting 26 mL of the 1.0 N MSA stock solution to 1.0 L with reagent water. Degas the eluent. Store the eluent in plastic labware.

Stock Standard Solutions

Purchase certified solutions or prepare 1000 mg/L stock standard solutions of each of the cations of interest. Dry reagent-grade salts to a constant weight and accurately weigh the amounts given in Table 1. Dissolve in reagent water in a 100-mL plastic volumetric flask. Dilute to volume with reagent water. Store in plastic containers at 4 °C. Stock standards are stable for at least three months.

Table 1 Mass of Compound Required to Prepare 100 mL of 1 g/L Solution of Cation

Cation	Compound	Mass (g)
Li^+	LiCl	0.6108
Na^+	NaCl	0.2542
NH_4^+	NH_4Cl	0.2965
K^+	KCl	0.1907
Mg^{+2}	$MgCl_2 \bullet 6H_20$	0.8365
Ca^{+2}	$CaCl_2 \bullet 2H_20$	0.3668

Working Standard Solutions

Prepare composite working standards at lower concentrations by diluting appropriate volumes of the 1000 mg/L stock standards with reagent water. Prepare working standards daily if they contain less than 100 mg/L of the cations.

SAMPLE PREPARATION

Filter all water samples through a 0.45-µm IC syringe filter. Discard the first 300 µL of filtrate and filter the remainder directly into a clean plastic autosampler vial.

Prepare aqueous soil extracts by adding 30 mL of either deionized water or 26 mM MSA to 3.0 g of soil. Extract in an ultrasonic bath for 30 minutes and filter through a 0.45-µm IC syringe filter as above.

SYSTEM PREPARATION AND SETUP

Prepare the CSRS-ULTRA for use by hydrating the eluent chamber. Use a disposable plastic syringe to slowly push approximately 3 mL of 200 mN NaOH through the Eluent Out port and 5 mL of 200 mN NaOH through the Regen In port. Allow the suppressor to sit for approximately 20 minutes to fully hydrate the suppressor screens and membranes. (For more information on CSRS operation, see the *Installation and Troubleshooting Instructions for the CSRS-ULTRA,* Document No. 031370-05).

Install the EG40, connect it to the system, and configure it with the PeakNet chromatography data system. Condition the EluGen MSA cartridge as directed in the EG40 manual by running a gradient from 1 to 60 mN MSA in 20 minutes, then 60 mN for 40 minutes at 1 mL/min. (For instructions on EG40 installation and use, see the *Operator's Manual for the EG40 Eluent Generator System,* Document No. 031373).

Remove the backpressure tubing temporarily installed during conditioning of the EluGen cartridge. Install a 5 x 50 mm IonPac CG16 and a 5 x 250 mm IonPac CS16 column. Make sure that the system pressure displayed by the pump is at least 2000 psi when 26 mM MSA is delivered at 1.5 mL/min, because the EG40 high pressure degas tubing assembly requires at least 2000 psi (14 MPa) of backpressure to efficiently remove hydrolysis gas from the eluent. If necessary, install backpressure coils supplied with the EG40 ship kit to bring the system pressure to between 2000 and 2800 psi. Because the system pressure can rise over time, occasional trimming of the backpressure coil may be necessary to maintain system pressure under 3000 psi. Do not exceed 3000 psi.

The CS16 storage solution is 30 mM MSA; before use, equilibrate the column with 26 mM MSA eluent for 60 minutes. Prior to sample analysis, analyze a system blank of reagent water. An equilibrated system has a background signal of less than 1 µS, and peak-to-peak noise should be about 0.2 nS. There should be no peaks eluting at the same retention time as the cations of interest.

Prepare a 500X dilution of the Six Cation Standard-II (Dionex P/N 046070) and make a 10-µL full loop injection. The column is equilibrated when two consecutive injections of standard produce the same retention times. Confirm that the resulting chromatogram resembles the chromatogram in Figure 1A.

Peak area precision and accuracy depend on autosampler performance. Replace the water in the flush reservoir daily with freshly filtered and degassed reagent water. Inspect the AS50 daily for bubbles in the sample syringe or its tubing. Purge to remove any bubbles by following the instructions in the AS50 manual.

The precision and accuracy of the AS50 will vary depending on the mode of injection. The most accurate and precise injections can be made with a calibrated sample loop in the full loop injection mode. To conserve sample, use one of the partial loop injection modes. Refer to the AS50 reference manual for a complete discussion of the different injection modes.

Make sure that the correct sample loop size and sample syringe volume are entered in the AS50 plumbing configuration screen.

RESULTS AND DISCUSSION

Three chromatograms of a cation standard performed at a constant 30 °C are overlayed in Figure 1A, demonstrating the good retention time reproducibility that results from temperature control. On the CS16 column, the retention time of cations will vary somewhat with temperature. This variability can be exploited to optimize selectivity among analytes, but maintaining a constant temperature ensures the best possible retention time reproducibility. If run at ambient temperature, this method will provide good selectivity between the cations, as shown in Figure 1B. In Figure 1B, a cation standard was run once at ambient temperature and then two more times as the temperature was increased to 27 °C. Note that the retention time of potassium is especially variable. Retention time variability could lead to misidentified peaks if the sample contains amines or other unknown compounds that elute near the standard cations. Use a thermal compartment for the best retention time reproducibility. Under these conditions lithium, sodium, ammonium, potassium, magnesium, and calcium are baseline resolved within 25 minutes.

Table 2 summarizes the calibration data and method detection limits (MDLs) obtained for the six cations. The high capacity of the CS16 column results in a calibration curve that is linear over three orders of magnitude for most of the cations, except for ammonium. The nonlinear dependence of peak height (or area) on amount is common for analytes that form weak acids or weak bases in the suppressor.[3,4] A quadratic curve fitting function extends the calibration curve for ammonium to a concentration of 40 mg/L.

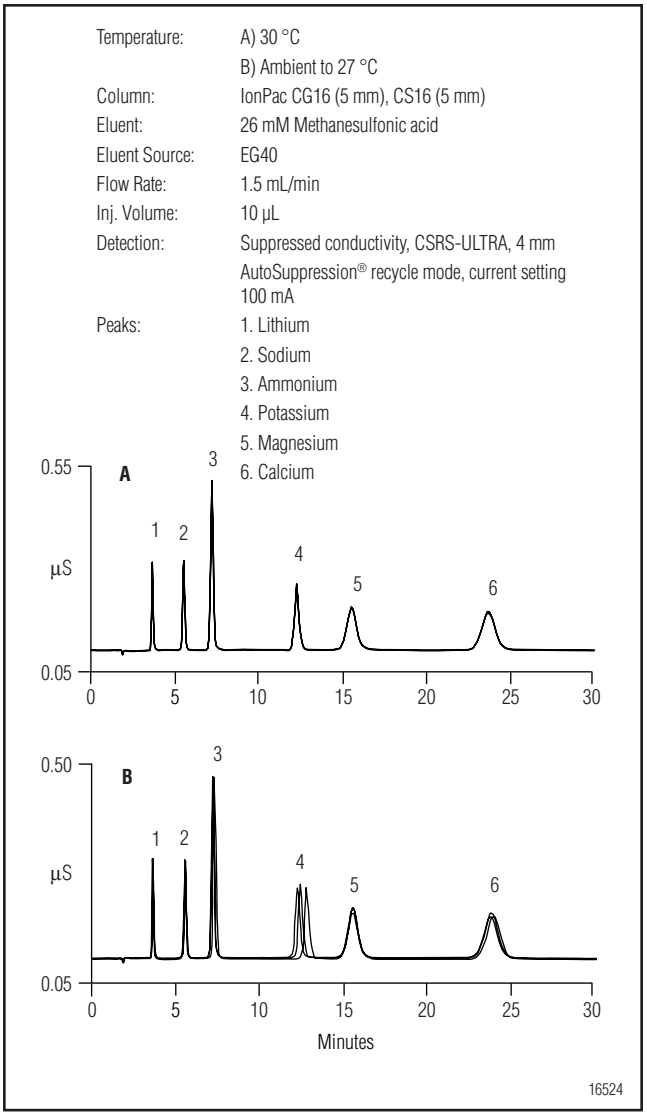

Figure 1. Temperature control improves RT reproducibility.

Table 2 Linear Range and MDLs for Cations and Ammonium

Cation	Range (mg/L)	Linearity (r^2)	Calculated MDL[a] (µg/L)	MDL Standard (µg/L)
Li^+	0.05–80	0.9999	0.19	1
Na^+	0.1–1000	0.9999	1.81	4
NH_4^+[b]	0.05–40	0.9993	1.23	5
K^+	0.05–80	0.9999	2.64	10
Mg^{+2}	0.05–80	0.9999	1.00	5
Ca^{+2}	0.05–80	0.9998	1.09	5

[a] MDL = (t) x (S) Where t = Student's t value for a 99% confidence level and a standard deviation estimate with n-1 degrees of freedom [t = 3.14 for seven replicates of the MDL Standard], and S = standard deviation of the replicate analysis.

[b] Quadratic fit

The method detection limit (MDL) is defined as the minimum concentration of an analyte that can be identified, measured, and reported with 99% confidence that the analyte concentration is greater than zero. To establish the single operator, within-day MDL for each analyte, make seven replicate injections of reagent water fortified with each analyte at a concentration of three to five times the estimated instrument detection limit. Perform all the calculations defined in the method and report the concentration values in the appropriate units.[5]

Suppressed conductivity detection allows detection down to the low µg/L level if 10 µL of sample is injected. Depending on the ionic strength and nature of the sample matrix, you may be able to achieve lower detection limits by injecting more sample.

The EG40 conveniently generates a high purity MSA eluent with a very stable composition. The EG40 increases the level of automation for this application while providing results comparable to a manually prepared eluent. This is reflected in the retention time (t_R) and peak area reproducibility summarized in Table 3 for seven injections of a QCS standard.

A typical way to validate the performance of methods used for environmental analysis is through precision and bias studies on spiked samples.[6] We evaluated the performance of this method in a similar way. First we determined the levels of the inorganic cations and ammonium in various environmental water samples. Then we spiked the samples with the analytes at a level that was 50–100% of the amount in the native sample. Table 4 summarizes the spike recovery of inorganic cations from various environmental water matrices. This method using the IonPac CS16 column provides acceptable recovery (i.e., 80–120%) of the inorganic cations from all matrices. Because the CS16 is a high capacity column, environmental samples with a wide range of ionic strength can be analyzed without interference from the matrix, as illustrated in Figure 2.

Table 3 Precision for Cations and Ammonium

Cation	QCS Standard (mg/L)	t_R Precision (%RSD)[a]	Area Precision (%RSD)[a]
Li^+	1	0.08	0.93
Na^+	4	0.09	0.97
NH_4^+	5	0.10	0.83
K^+	10	0.15	0.99
Mg^{+2}	5	0.19	0.93
Ca^{+2}	5	0.22	1.15

[a] Relative standard deviation, n = 7

Table 4 Inorganic Cation Recoveries Using the IonPac CS16

Cation	Amount Added (mg/L)	Recovery (%)
Reagent Water		
Lithium	2	100
Sodium	2	97
Ammonium	2	107
Potassium	2	82
Magnesium	2	86
Calcium	2	82
Domestic Wastewater		
Lithium	2	109
Sodium	50	97
Ammonium	50	107
Potassium	20	94
Magnesium	20	105
Calcium	2	101
Drinking Water		
Lithium	1	103
Sodium	20	101
Ammonium	1	105
Potassium	1	84
Magnesium	30	100
Calcium	20	105
ASTM Wastewater		
Lithium	2	99
Sodium	100	95
Ammonium	2	109
Potassium	20	95
Magnesium	3	95
Calcium	2	86

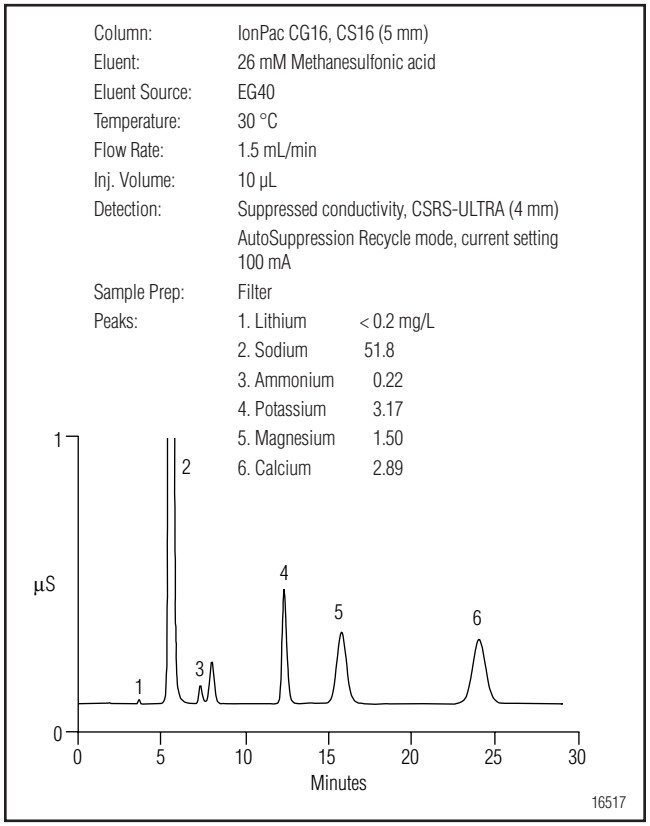

Figure 2. Determination of inorganic cations and ammonium in industrial wastewater with the IonPac CS16 column.

The IonPac CS16 is a high capacity cation exchange column that replaces the CS15 column for disparate concentration ratios of ammonium and sodium in diverse sample matrices. The CS16 is ideal for the determination of low concentrations of ammonium in environmental waters. It provides improved resolution of sodium from ammonium and alkanolamines, even for samples high in ionic strength. Figure 3 illustrates the determination of trace level ammonium in the presence of high sodium.

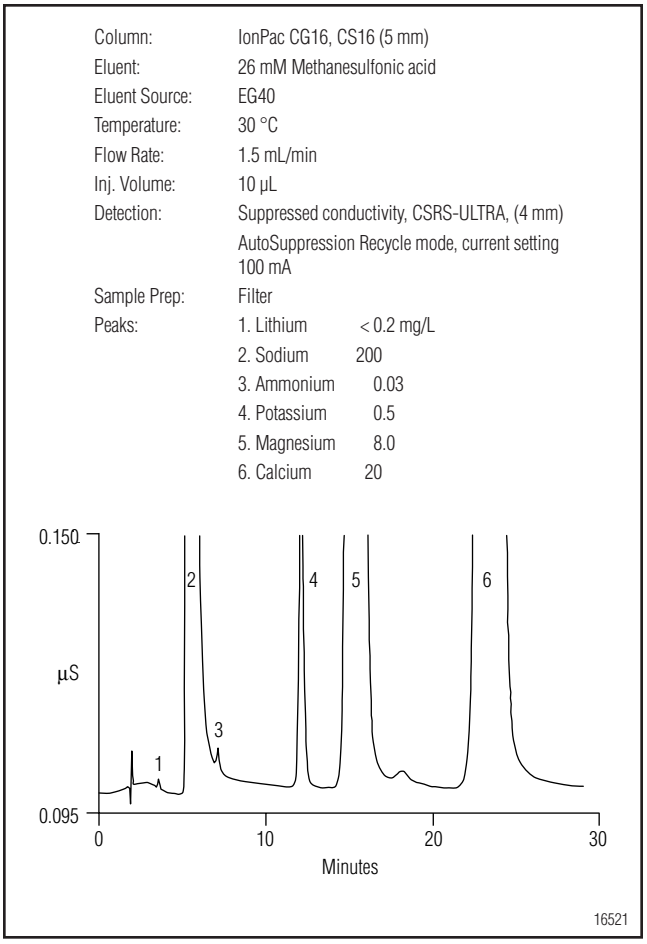

Figure 3. Resolution of trace ammonium from high sodium with the IonPac CS16 column.

The IonPac CS16 packing is compatible with acidic eluents and samples. Acid digests, acid-preserved samples, or acidic soil extracts that contain up to 100 mM hydronium ion can be injected without pH adjustment. Figure 4 shows this for an acidic soil extract.

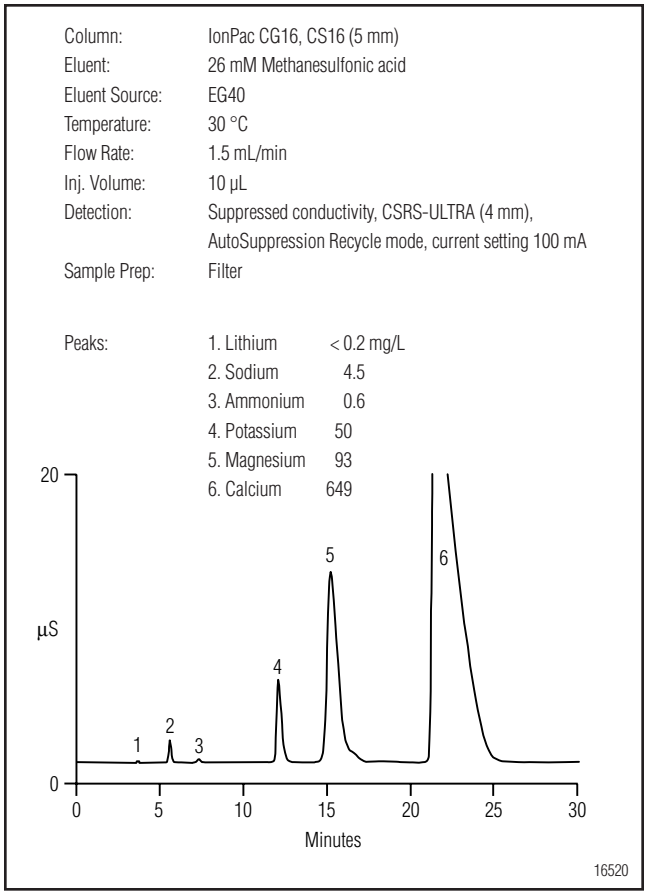

Figure 4. Determination of inorganic cations and ammonium in acidic soil extract with the IonPac CS16 column.

Strongly retained compounds from injected environmental water samples can accumulate on the column and degrade its performance. Signs of a fouled column include loss of capacity, loss of resolution, shortened retention times, higher noise and background, spurious peaks, and peak tailing. The CS16 column can be flushed with up to 100% acetonitrile to help remove contaminants from the column. (For more information on column troubleshooting and cleanup, see the Installation Instructions and Troubleshooting Guide for the IonPac CS16 Analytical Column, Document No. 031725).

SUMMARY

This application uses the IonPac CS16 column with a 26 mM MSA eluent and suppressed conductivity detection to determine inorganic cations and ammonium at concentrations ranging from 0.1–80 mg/L. The high capacity of the IonPac CS16 column enables the analysis of a wide range of environmental waters, and also resolves trace ammonium in the presence of a 10,000-fold higher concentration of sodium. The retention time precision is 0.2% RSD or less, and the peak area precision is 1% RSD or less, for standards in the low mg/L range.

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2. "Standard Methods for the Analysis of Water and Wastewater", 18th Edition, p. 4-75 to 4-85. American Public Health Association, Washington, DC, 1992.
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6. "The Determination of Inorganic Anions in Water by Ion Chromatography"; U.S. Environmental Protection Agency Method 300.0; Cincinnati, Ohio, 1993.

SUPPLIERS

VWR Scientific Products, 3745 Bayshore Blvd., Brisbane, CA 94005, USA. Tel: 800-932-5000. www.vwrsp.com.

Fisher Scientific, 711 Forbes Ave., Pittsburgh, PA 15219-4785, USA. Tel: 800-766-7000. www.fisherscie.com

Sigma Chemical Co., P.O. Box 14508, St. Louis, MO 63178 USA. Tel: 800-325-3010. www.sigma-aldrich.com.

Determination of Calcium and Magnesium in Brine

INTRODUCTION

To prevent membrane poisoning, new membrane technology in chlor-alkali cells requires feed brine that is relatively free of alkaline earth metals. Membrane manufacturers recommend that saturated brines be purified to a total hardness below 50 µg/L (ppb) to extend membrane life and ensure electrical current efficiency in the cell.

Industrial laboratories have been concerned with the determination of calcium and magnesium in brine for a number of years. For example, investigators at Vulcan Chemical developed an IC method for the determination of alkaline earth metals in high-purity brine in the 1980s. The method in this Application Note can be considered an extension of previous work at Vulcan Chemical.[1]

This Application Note describes a method for determining low µg/L amounts of calcium and magnesium in a 30% sodium chloride brine by ion chromatography. A MetPac™ column, which selectively retains calcium and magnesium and has a low affinity for sodium, is used to concentrate calcium and magnesium from the brine. The concentrated calcium and magnesium are separated on an IonPac® CS12A cation-exchange column and detected by suppressed conductivity detection.

EQUIPMENT

Dionex DX-500 Ion Chromatography system consisting of:

- GP40 Gradient Pump
- CD20 Conductivity Detector
- LC20 Chromatography Enclosure equipped with a rear-loading Rheodyne injection valve
- Rinsing Pump, DQP (P/N 35250)
- LC10 Chromatography Organizer equipped with a rear-loading Rheodyne injection valve

4-L Plastic bottle (for 1 mM hydrochloric acid rinsing solution)

PeakNet Chromatography Workstation

REAGENTS AND STANDARDS

Deionized water (DI H_2O), Type I reagent grade, 18 MΩ-cm resistance or better

> 99% Methanesulfonic acid (Fluka or equivalent)

Hydrochloric acid, ultrapure reagent (J.T. Baker ULTREX® II, 36.9% or equivalent)

Sodium hydroxide, 50% (w/w) aqueous solution (Fisher Scientific or equivalent)

Brine (30% sodium chloride solution, kindly provided by Dr. David Hildebrand, Vulcan Chemical, Wichita, Kansas, USA)

CONDITIONS

Trap Column (for contaminants in the rinse solution):	TMC-1 Concentrator Column (P/N 49000)
Concentrator Column:	MetPac CC-1 Concentrator Column (P/N 39567)
Analytical Columns:	IonPac CS12A Analytical, 4 x 250 mm (P/N 46073)
	IonPac CG12A Guard, 4 x 50 mm (P/N 46074)
Eluent:	A: 20 mM Methanesulfonic acid
Eluent Flow Rate:	1 mL/min
Rinsing Reagent:	1 mM Hydrochloric acid

Rinsing Flow Rate:	2 mL/min
Rinse Time:	20 min
Total Run Time:	35 min
Sample Volume:	100 µL
Detection:	Suppressed conductivity, CSRS®-II (4 mm), AutoSuppression® recycle mode
System Backpressure:	1000–1500 psi (6.9–10.3 MPa)
Background:	0.3–3 µS

Time Functions Program

Time (min)	A(%)	Valve 1[1]	Valve 2[2]	Remarks
Initial	100	load	inject	
0.0	100	load	load	Fill sample loop
1.0	100	inject	load	Sample to MetPac
20.0	100	load	inject	Begin sampling[3]
35.0	100	load	inject	Finish sampling

[1] Valve 1 is used for loading the concentrator column (MetPac).

[2] Valve 2 is used for eluting and separating compounds of interest on the CS12A analytical column.

[3] Begin sampling refers to data collection (the MetPac column is switched in-line with the CG12A and CS12A columns.)

PREPARATION OF SOLUTIONS AND REAGENTS

Standard Solutions

Stock magnesium standard solution (1000 mg/L)
(VWR Scientific)

Stock calcium standard solution (1000 mg/L)
(VWR Scientific)

Calibration Standard Solutions

Appropriate calibration standards are prepared from 1000 mg/L standards specified above. Select a range similar to the expected analyte concentrations in the samples. All standards should be prepared in brine.

Eluent Solutions

Stock eluent solution: 1 M Methanesulfonic acid

Weigh 934.5 g of deionized water (Type I reagent grade, 18 MΩ-cm resistance or better) into an eluent bottle. Degas for approximately 5 minutes. Tare the bottle and carefully add 65.5 mL of > 99% methanesulfonic acid directly to the bottle.

Working eluent solution: 20 mM Methanesulfonic acid

Weigh 980.0 g of deionized water (Type I reagent grade, 18 MΩ-cm resistance or better) into an eluent bottle. Degas for approximately 5 minutes. Add 20 mL of 1 M methanesulfonic acid to the bottle.

Stock rinsing solution: 1 M Hydrochloric acid

Weigh 909.70 g of deionized water (Type I reagent grade, 18 MΩ-cm resistance or better) into an eluent bottle. Degas for approximately 5 minutes. Tare the bottle and carefully add 90.3 mL of ultrapure reagent grade hydrochloric acid directly to the bottle.

Working rinse solution: 1 mM Hydrochloric acid

Weigh 999.0 g of deionized water (Type I reagent grade, 18 MΩ-cm resistance or better) into an eluent bottle. Degas for approximately 5 minutes. Add 1 mL of 1 M hydrochloric acid directly to the bottle.

Stock sample pH adjustment solution: 500 mM Sodium hydroxide

Weigh 960.0 g of deionized water (Type I reagent grade, 18 MΩ-cm resistance or better) into an eluent bottle. Degas for approximately 5 minutes. Tare the bottle and carefully add 40.0 g of 50% sodium hydroxide directly to the bottle.

Standards and Sample Preparation

Add 0.2 mL of 500 mM sodium hydroxide solution to 9.8 mL of sample (brine).

The final concentration of sodium hydroxide is 10 mM.

SYSTEM OPERATION

System configuration and operation parameters for this application are outlined in previously published documents.[2,3]

The sensitive analysis of calcium and magnesium in brine is accomplished in four steps:

1. Fill the sample loop
2. Load the concentrator column
3. Eliminate the sodium matrix
4. Separate calcium and magnesium

Figure 1 shows how the system performs these tasks. In Figure 1A, the system is in the standby mode, ready for sample analysis. In Figure 1B, Valve 2 is switched to the LOAD position and the sample is loaded into the 100 µL sample loop on Valve 1.

Sample Handling

The black rubber plunger in disposable plastic syringes can be a source of contamination. To minimize the introduction of contamination, pull rather than push the sample into the loop, as shown in Figure 1B. Be sure to pull slowly so that bubbles are not introduced. The loop should be overfilled by at least 3 times its capacity (> 300 µL) to ensure reproducible results. It is important to have the TMC column in line between the DQP pump and Rheodyne Valve 1 (Figure 1A) to remove the significant amount of calcium and magnesium present in the 1 mM hydrochloric acid solution. The contamination level of calcium and magnesium in the 1 mM hydrochloric acid solution depends on the reagent purity and grade of DI water. A water blank should be run before analyzing

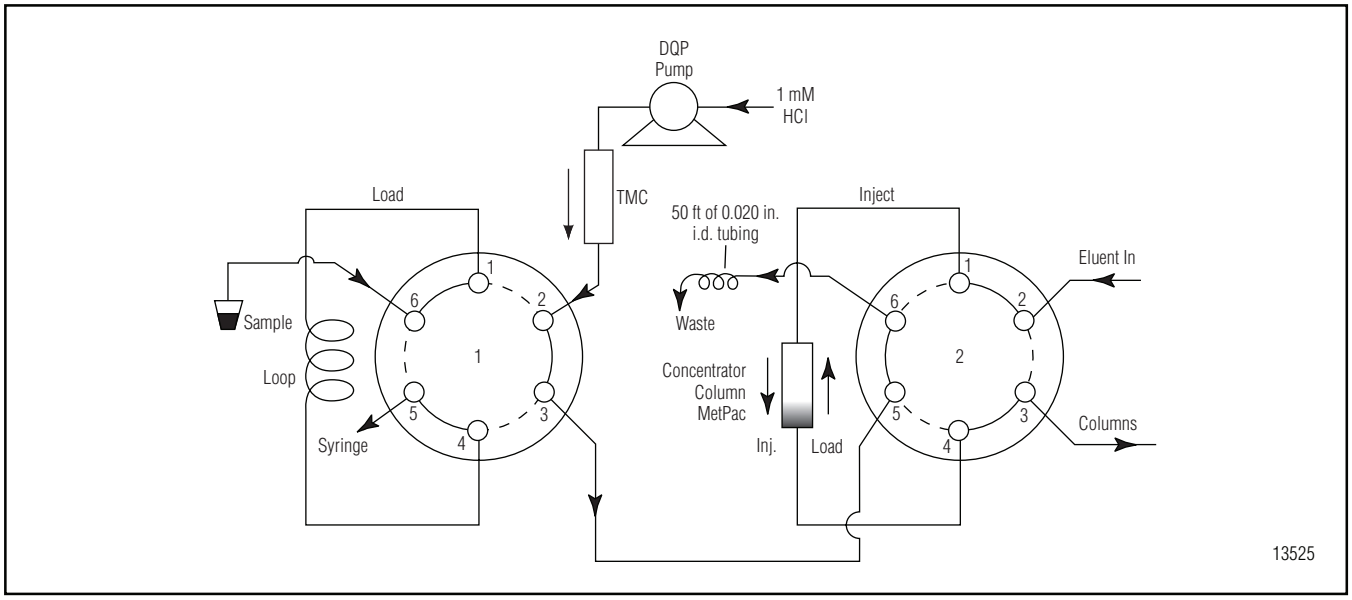

Figure 1A *Initial conditions*

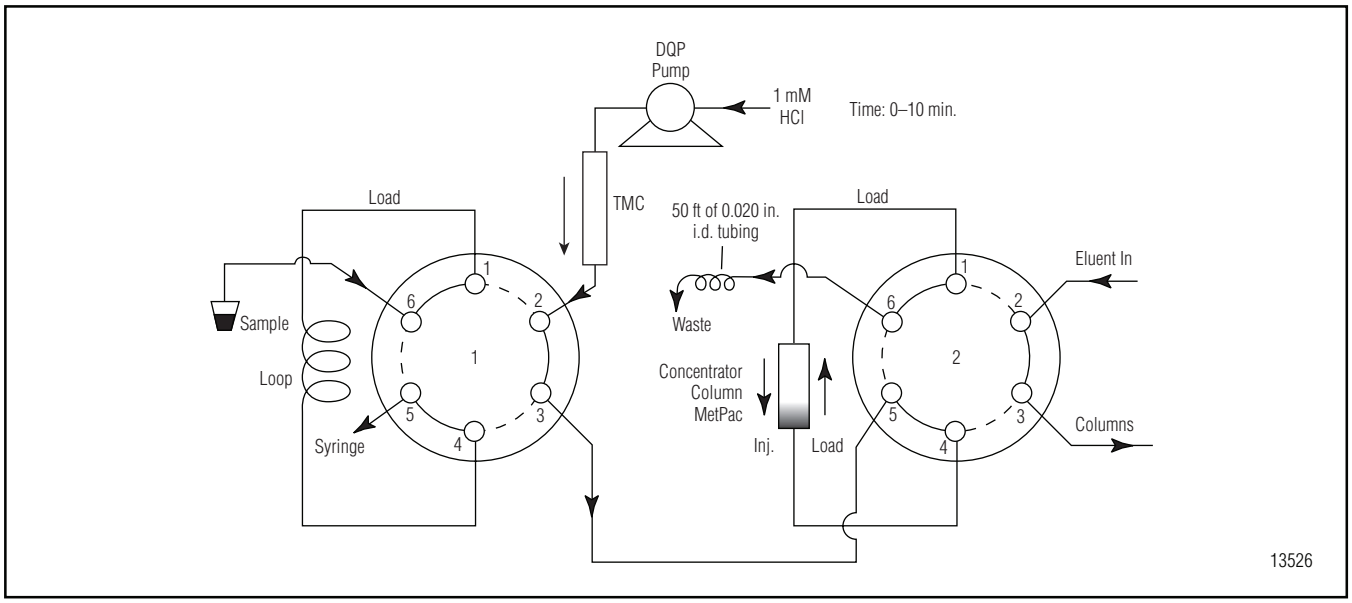

Figure 1B *Loading the sample loop*

samples. If there has been a significant increase in magnesium and calcium, the capacity of the TMC is exhausted. To clean the TMC, flush with approximately 25–30 mL of 1.5 M hydrochloric acid.

Matrix Elimination

After the sample loop is filled, 1 mM hydrochloric acid from the DQP transfers the sample out of the loop and onto the MetPac column in the opposite direction of the eluent (Figure 1C). Calcium and magnesium are retained on the concentrator column. The sodium matrix is removed from the MetPac column with 1 mM hydrochloric acid from the DQP flowing at 2 mL/min for 20 minutes (DQP head pressure should be at least 100 psi). Finally, activating Valve 2 to the INJECT position switches the MetPac column in-line with the eluent stream and the analytical columns (Figure 1D). Calcium and magnesium are then eluted from the MetPac column in the reverse

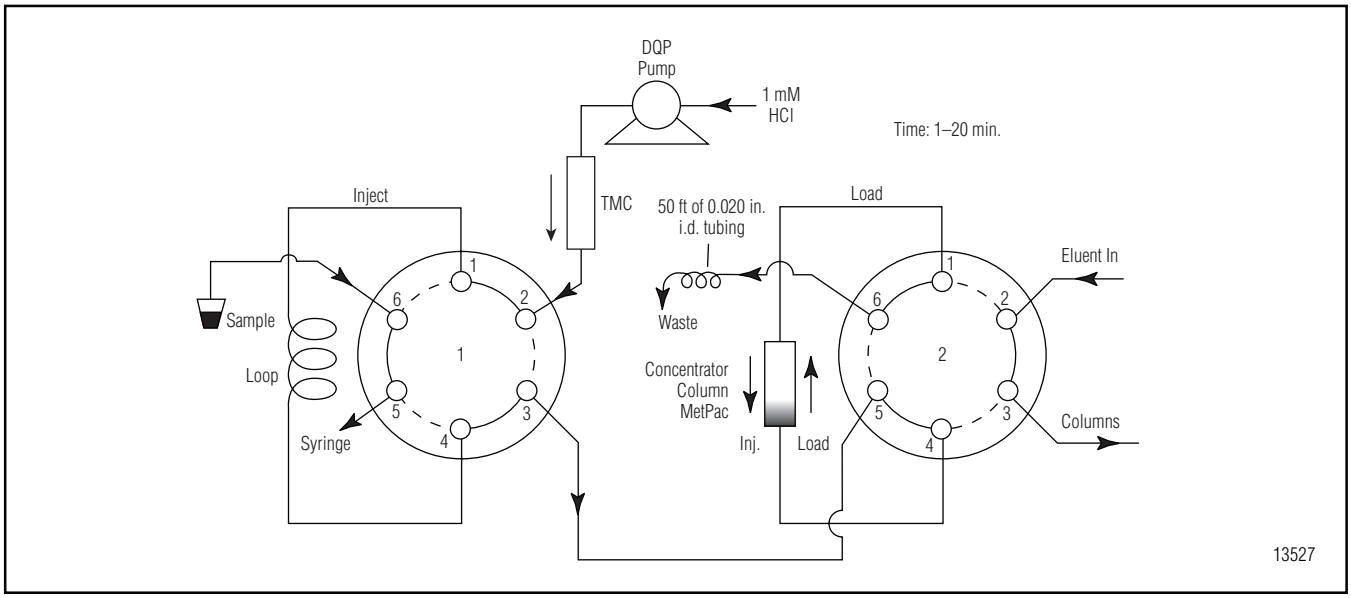

Figure 1C *Loading the concentrator column and eliminating the matrix*

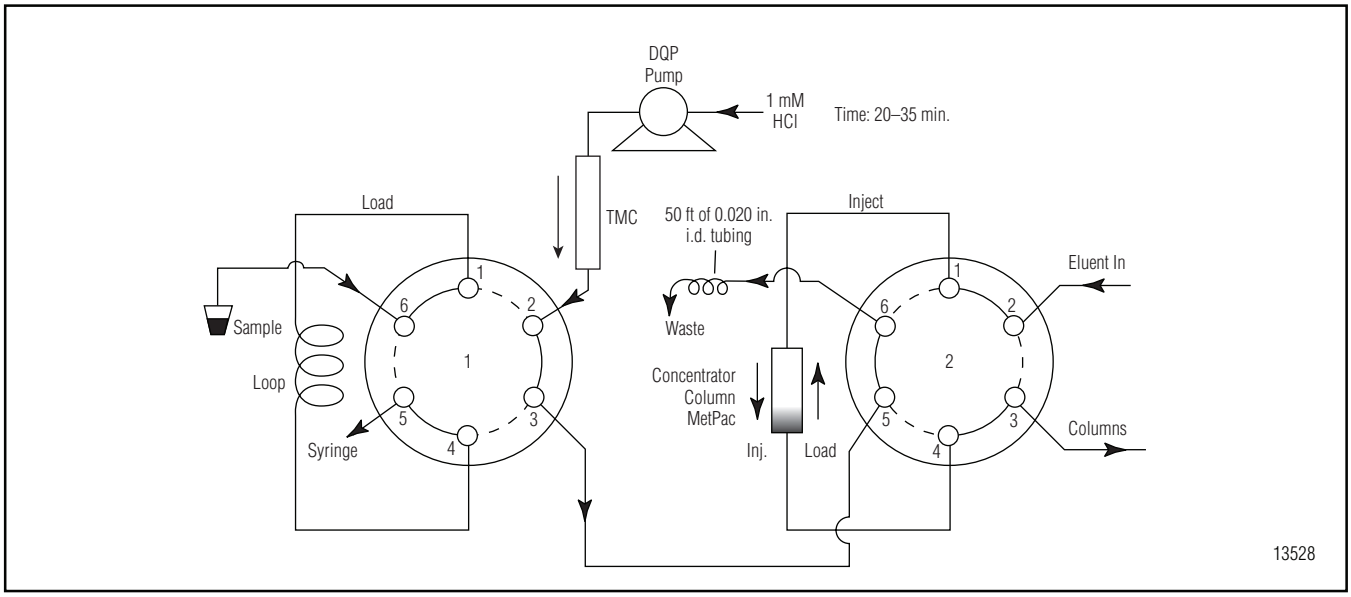

Figure 1D *Chromatography of retained Calcium and Magnesium*

direction of the concentration step and separated on the CG12A and CS12A columns. Special care should be taken to minimize contamination. The deionized water used for preparing the rinse solution, eluent, and sodium hydroxide solution should be free of measurable levels of ionic impurities, organics, microorganisms, and particulate matter larger than 0.2 µm in diameter.

RESULTS AND DISCUSSION

For the best performance at low µg/L levels, it is critical that baseline noise be kept to a minimum. An equilibrated system will demonstrate a conductivity background between 0.3–3 µS. Peak-to-peak noise is typically 10 nS and system backpressure is 1000–1500 psi (6.9–10.3 MPa). A system blank is determined by using deionized water as the sample. This blank establishes the baseline and confirms the lack of contamination in the system. A small amount of sodium contamination in the system does not interfere with calcium and magnesium detection.

In these experiments, purified brine from a major chlor-alkali producer was used. Brine prepared with commercially available sodium chloride (VWR, reagent grade) contained over 100 µg/L of calcium and magnesium. All standards must be prepared in brine. In the method described here, trace levels of magnesium and calcium in brine are concentrated on a MetPac column, which is more selective for divalent than monovalent cations. For optimum calcium and magnesium recovery, the sample pH must be adjusted to 11.5. At high pH values, the MetPac selectivity for divalent over monovalent cations is increased. At lower pH values, calcium and magnesium recoveries are poor and are not linear within the concentration range used for this analysis. Calcium and magnesium recoveries were not increased with sample pH values higher than 11.5. Figures 2 and 3 show that the detection of calcium and magnesium is linear in the low µg/L range. Even at pH 11.5, the MetPac column binds some sodium. To remove sodium, the MetPac is rinsed with 1 mM hydrochloric acid. Higher concentrations of hydrochloric acid cause a reduction in calcium and magnesium recovery, while lower hydrochloric acid concentrations increase the rinse time required for rugged chromatography. Despite washing the MetPac column for 20 minutes to eliminate the matrix, detectable levels of sodium elute from the MetPac to the CS12A column (see Figure 4). However, this level of sodium does not interfere with the detection of low concentrations of calcium and magnesium (see Figure 5). Shorter wash times (less than

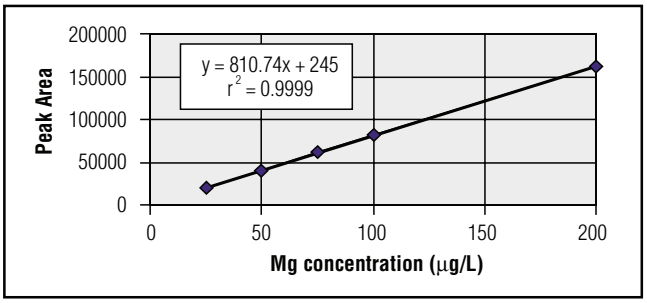

Figure 2 *Magnesium calibration in brine*

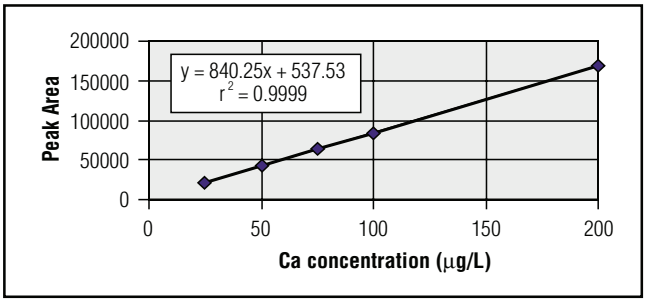

Figure 3 *Calcium calibration in brine*

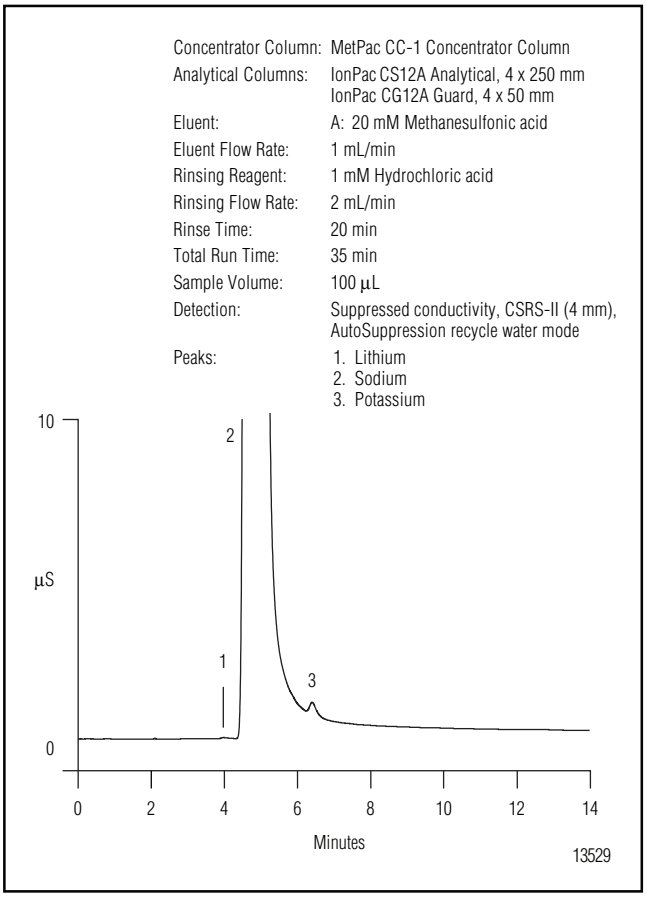

Figure 4 *Brine blank*

20 minutes) were investigated and did not adequately eliminate the sodium interference. The injection volume was optimized at 100 µL. A larger injection volume may exceed the MetPac column capacity and cause irreproducible results. The data in Table 1 show that 5 µg/L of calcium and magnesium can be easily quantified in brine.

SUMMARY

The method outlined in this Application Note accurately quantifies low µg/L amounts of calcium and magnesium in brine by using an on-line matrix elimination technique.

REFERENCES

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2. Dionex Corporation, "Determination of Trace Anions in Isopropyl Alcohol", Application Note 85.
3. Kaiser, E.; Wojtusik, M. J. *J. Chromatogr. A.* **1994**, *671*, 253–258.

LIST OF SUPPLIERS

J.T. Baker Incorporated, 222 Red School Lane, Phillipsburg, New Jersey, 08865, USA. Tel.: 1-800-582-2537.

VWR Scientific, P.O. Box 7900, San Francisco, California, 94120, USA. Tel.: 1-800-932-5000.

Fluka Chemika-BioChemika, Fluka Chemie AG, Industriestrasse 25, CH-9471, Buchs, Switzerland. Tel.: 081 755 25 11.

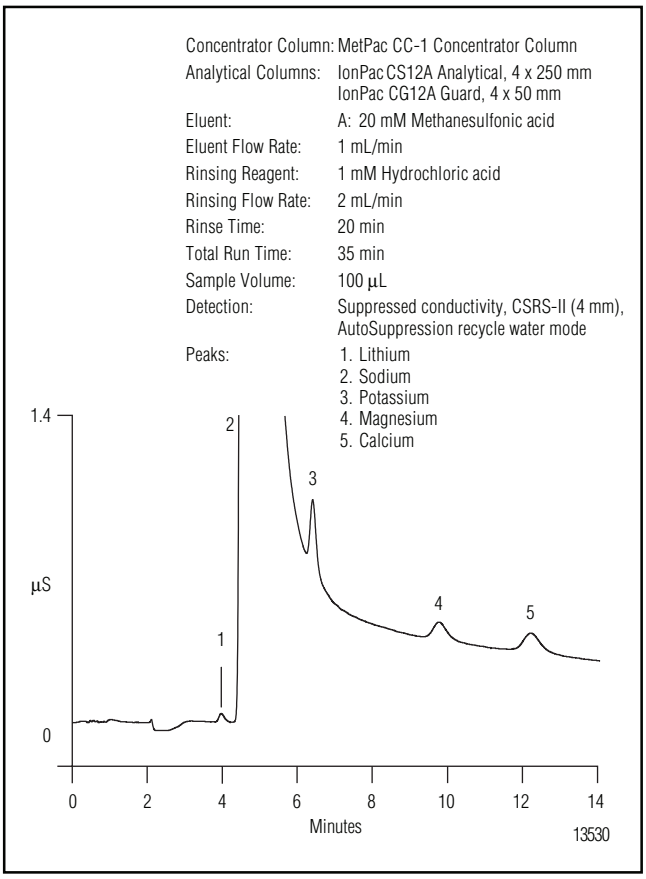

Figure 5 *25 ppb of Calcium and Magnesium in brine*

Table 1 Area counts and RSD of 5 ppb calcium and magnesium in brine

Injection #	5 ppb Magnesium (area count)	5 ppb Calcium (area count)
1	3748	4309
2	3773	4355
3	3709	4368
4	3817	4410
5	3856	4588
6	3826	4471
7	3921	4572
Average	3807	4439
RSD	1.86	2.46

Determination of Trace Cations in Power Plant Waters Containing Morpholine

INTRODUCTION

Morpholine and ammonium are used as additives in power plant waters. Morpholine acts as a corrosion inhibitor, whereas ammonium is used to control pH. In this matrix, it is critical to determine the presence of inorganic cation contaminants. This method uses the IonPac CS14 column to quantify trace concentrations of lithium, sodium, potassium, magnesium, and calcium in the presence of high levels of ammonium and morpholine. Acetonitrile can be added to the eluent to improve peak shape and optimize resolution for some of the cations of interest.

EQUIPMENT

Dionex Chromatography system comprising:
Advanced Gradient Pump (AGP)
Liquid Chromatography Module (LCM-3), equipped with Model 9126-038 Rheodyne Injector or equivalent
Sample Preparation Module (SPM) [for 8200 Process Analyzer]
Conductivity Detector Module (CDM-3)
Sample Loading Pump, RP-1

REAGENTS AND STANDARDS

Deionized water (DI H_2O), Type I reagent grade, 17.8MW / cm resistance or better. Methanesulfonic Acid (MSA) (+99% pure) Acetonitrile (ACN) HPLC grade

CONDITIONS

Columns:	(2) IonPac CG14 (2-mm) guard columns, one used as a guard column, the other as a concentrator column (1) IonPac CS14 (2-mm) analytical column
Eluent :	8 mM Methanesulfonic acid or 8 mM Methanesulfonic acid in 5% acetonitrile (v/v)
Eluent Flow Rate:	0.25 mL/min
Rinsing Flow Rate:	1.0 mL/min
Sample Volume:	1.0 mL
Detection:	Suppressed Conductivity
Suppressor:	CSRS-I (2-mm), AutoSuppression, Recycle Mode (without acetonitrile); External Water Mode (with acetonitrile)

Pump programs:

1. 8 mM methanesulfonic acid

Time	E1	V5	V6	Remarks
0.0	100	1	0	Prime RP-1 with sample
5.0	100	0	0	Load sample to CG-14
6.0	100	0	0	Inject

2. 8 mM methanesulfonic acid with 5% acetonitrile

Time	E1	V5	V6	Remarks(rinse)
0.0	100	1	0	Prime RP-1 with sample
5.0	100	0	0	Load sample to CG-14
6.0	100	0	1	Rinse CG-14 with w/ 5% ACN
8.0	100	1	1	Inject

Timed Events Programs (for operation of 8200 Process Analyzer):

1.) 8 mM methanesulfonic acid

Step	Time	Description
Initial	-	ACI SPM valve 3 OFF
Initial	-	ACI RP-1 OFF
1	0.3	ACI RP-1 ON
2	4.0	AGP Run Gradient Clock
3	9.9	CDM-3 AutoOffset ON
4	10.0	ACI SPM valve 3 OFF
4	10.0	ACI RP-1 OFF
4	10.0	Start Sampling

2.) 8 mM methanesulfonic acid with 5% acetonitrile

Step	Time	Description
Initial	-	ACI SPM valve 3 OFF
Initial	-	ACI RP-1 OFF
1	0.3	ACI RP-1 ON
2	4.0	AGP Run Gradient Clock
3	11.9	CDM-3 AutoOffset ON
4	12.0	ACI SPM valve 3 OFF
4	12.0	ACI RP-1 OFF
4	12.0	Start Sampling

note: ACI = Advanced Computer

PREPARATION OF SOLUTIONS AND REAGENTS

1 M Methanesulfonic acid (MSA) Eluent Concentrate

Weigh 96.10 g of methanesulfonic acid (MSA). Carefully add this amount to a 1-L volumetric flask containing about 500 mL of deionized water. Dilute to the mark and mix thoroughly.

8 mM Methanesulfonic acid (MSA)

Pipette 8.0 mL of the 1.0 M MSA eluent concentrate into a 1-L volumetric flask. Dilute to 1 L using deionized water. Degas the eluent.

8 mM Methanesulfonic acid (MSA) / 5% Acetonitrile

Pipette 8.0 mL of the 1.0 M MSA eluent concentrate into a 1-L volumetric flask. Dilute to approximately 800 mL using deionized water. Degas the eluent. Add 50 mL of acetonitrile and mix until all components are in solution. Dilute to a final volume of 1.0 L using deionized water.

5% Acetonitrile for Rinsing

Add 50 mL of acetonitrile to approximately 800 mL using deionized water in a 1-L volumetric flask and mix until all components are in solution. Dilute to a final volume of 1.0 L using deionized water.

Stock standard solution (1000 mg/L)

Lithium (Li^+) 1000 mg/L: Dissolve 6.108 g of lithium chloride (LiCl) in deionized water and dilute to 1.000 liter.

Sodium (Na^+) 1000 mg/L: Dissolve 2.542 g of sodium chloride (NaCl) in deionized water and dilute to 1.000 liter.

Ammonium (NH_4^+) 1000 mg/L: Dissolve 2.965 g of ammonium chloride (NH4Cl), in deionized water and dilute to 1.000 liter.

Potassium (K^+) 1000 mg/L: Dissolve 1.907 g of potassium chloride (KCl) in deionized water and dilute to 1.000 liter.

Morpholine (tetrahydro-1,4-oxazine [C_4H_9NO])

1000 mg/L: Pipet 1.00 mL of morpholine into 800 mL of deionized water and dilute to 1.000 liter.

Magnesium (Mg_2^+) 1000 mg/L: Dissolve 8.365 g of magnesium chloride, hexa- hydrate MgCl2.6H2O in deionized water and dilute to 1.000 liter.

Calcium (Ca_2^+) 1000 mg/L: Dissolve 3.668 g of calcium chloride, dihydrate $CaCl_2.2H_2O$ in deionized water and dilute to 1.000 liter.

Calibration

Intermediate standards (low mg/L) are prepared by appropriate dilutions of the stock solutions. Calibration standards (mg/L) are prepared by further diluting the intermediate standards. Prepare a minimum of three concentration levels to bracket the expected concentrations of the sample of interest.

DISCUSSION AND RESULTS

Trace cations in a power plant morpholine matrix are determined by concentrating 1 mL of sample on a 2-mm CG14 concentrator column. No sample pre treatment is necessary. The 2-mm column and suppressor system used in this application has advantages over a 4-mm system: lower eluent flow rates, higher suppression capacity, and less waste generation.

Two different eluents can be used, 8 mM MSA with or without 5% acetonitrile. The benefit of having organic solvent in the eluent results in improved morpholine peak shape, better separation between morpholine and magnesium, and increased response for the divalent cations. The CSRS is operated in the external water mode with 5% acetonitrile in the eluent. When using the eluent containing acetonitrile, the sample is loaded onto the CG14 concentrator column and then rinsed with 2 mL of 5% acetonitrile. This process replaces the aqueous mobile phase in the concentrator with one more nearly matched to the eluent while still retaining the cations of interest. The concentrator column is then switched in line with the eluent stream and the analytical columns. The cations of interest are eluted from the concentrator and separated on the guard and analytical columns. Figures 1 - 4 illustrate how the system performs these tasks. When 8 mM MSA without acetonitrile is used as the eluent, the CSRS is operated in the recycle mode and the step represented by Figure 3 is omitted.

To validate this method, precision and linearity were determined for the case with 5% acetonitrile in the eluent. A multilevel calibration based on the values listed in Table 1 yielded good r^2 values over a wide range. A sample with analyte concentrations within the calibration range showed acceptable precision for both concentration and retention time. Figure 5 shows a representative chromatogram of the sample and Table 2 summarizes the precision results for 21 replicates. For comparison, Figure 6 shows the same sample run with 8 mM MSA and the CSRS in the recycle mode. The maximum sample volume that can be loaded for this method without significant deviation from linearity is 50 mg/L morpholine. A system blank is determined by running deionized water as a sample. Figure 7 illustrates a chromatogram of a blank for the 5% acetonitrile method.

This method using the IonPac CS14 is a useful analytical procedure to determine trace cations in power plant waters containing high morpholine and ammonium. Reproducibility and linearity are within acceptable limits. This method is applicable to on-line and grab sample analysis. The addition of acetonitrile to the eluent improves morpholine peak shape and resolution, permitting quantitative analysis of morpholine.

Table 1. Multilevel Calibration for Trace Cations in Morpholine Mix for 8 mM MSA w/ 5% ACN

Cation	Level 1	Level 2	Level 3	Level 4	r^2
Lithium	0.125	0.375	1.25	3.75	0.99766
Sodium	0.5	1.5	5.0	15.0	0.99825
Ammonium	37.5	112.5	375.0	1125.0	0.99495
Potassium	0.5	1.5	5.0	15.0	0.99690
Morpholine	500.0	1500.0	5000.0	15000.0	0.99478
Magnesium	0.5	1.5	5.0	15.0	0.98745
Calcium	2.5	7.5	25.0	75.0	0.99166

Concentration in mg/L

Table 2. Reproducibility for Trace Cations in Morpholine Mix for 8 mM MSA w/ 5% ACN

Cation	Conc (mg/L)	RSD, Conc (%)	Retention Time (min)	RSD Retention Time (%)
Lithium	0.5	4.2	4.30	0.3
Sodium	2.0	3.8	4.72	0.3
Ammonium	150.0	3.8	5.53	0.3
Potassium	2.0	3.7	6.78	0.8
Morpholine	2000.0	3.0	8.15	0.6
Magnesium	2.0	2.0	11.16	0.3
Calcium	10.0	2.7	12.93	0.4

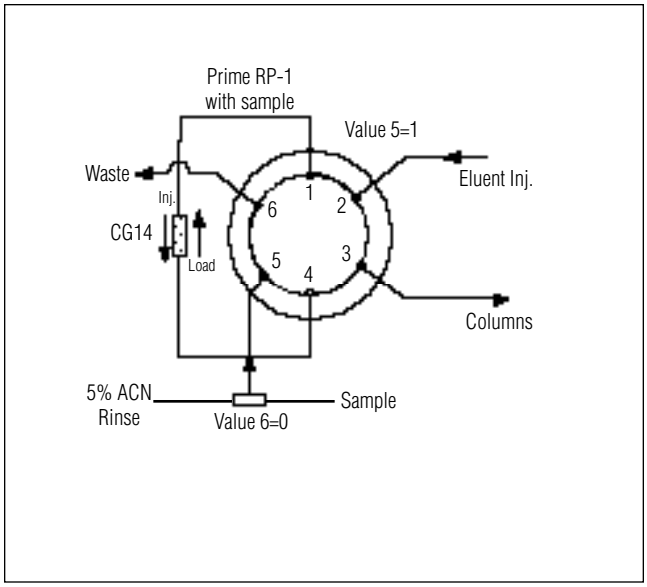

Figure 1. *Prime RP-1 with sample*

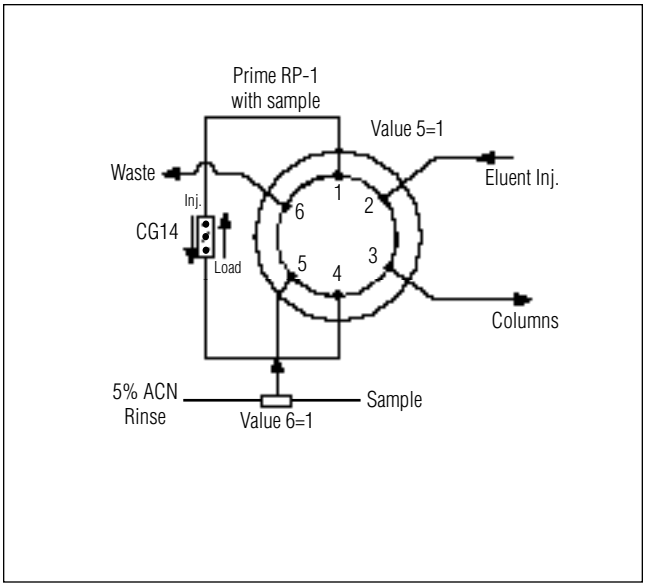

Figure 3. *Rinse CG14 with 5% ACN*

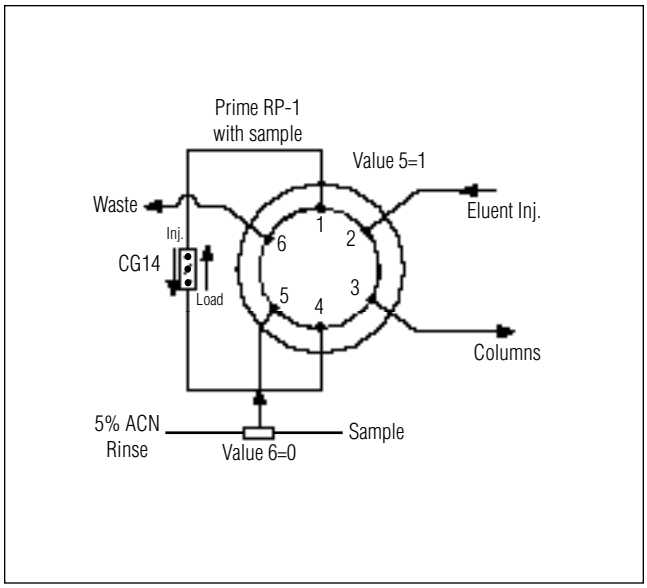

Figure 2. *Load sample to CG14*

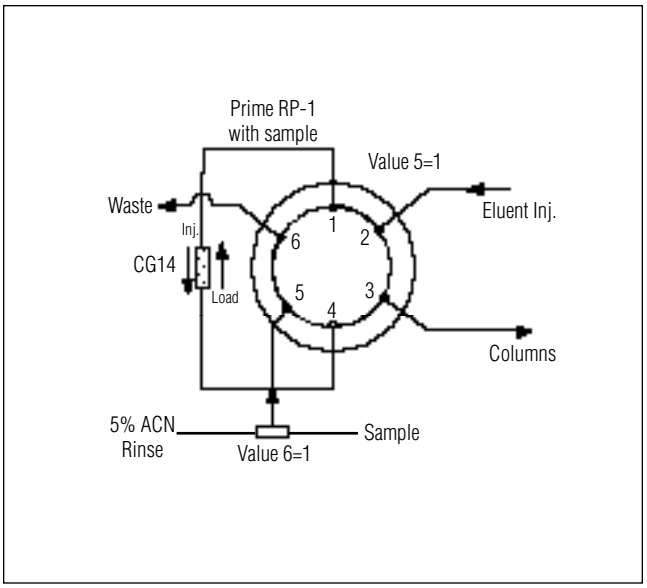

Figure 4. *Schematic of IC system: chromatographing the retained ions*

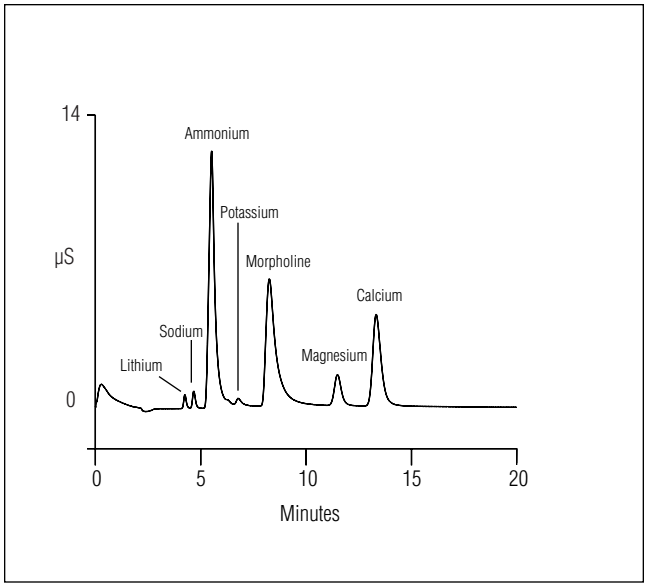

Figure 5. *Trace cations in Morpholine mix: Method 2*

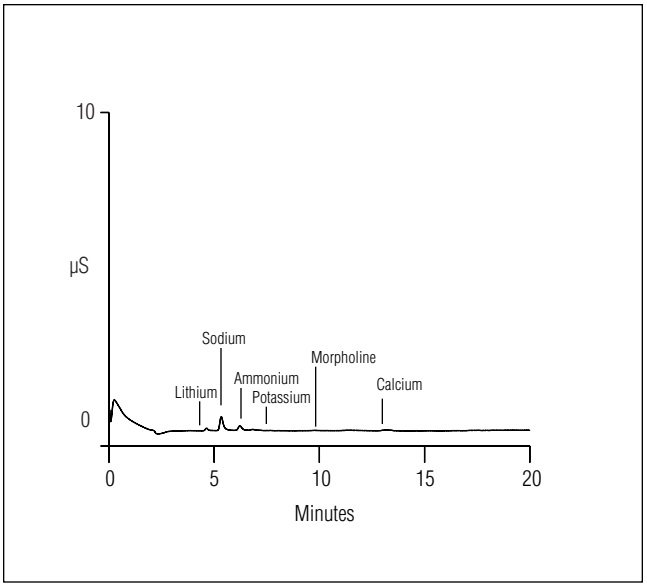

Figure 6. *Trace cations in Morpholine mix: method 1*

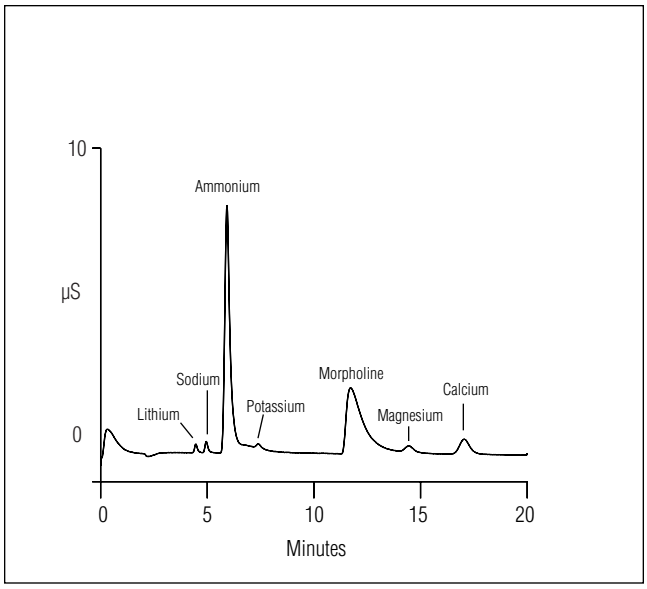

Figure 7. *Typical blank chromatograms: Method 2*

PRECAUTIONS

Several factors can affect the success of this method. Morpholine can decompose to a variety of compounds such as: formic acid, methylamine, ethylamine, and glycolic acid. The presence of amines could potentially interfere with the separation of the analytes of interest. In addition, it is important to minimize contamination by using the highest quality deionized water and using special care when handling chemicals and instrumentation.

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Bostic, D.; Burns, G.; Harvey, S. J. Chromatogr. **1992**, 602, 163-171

Determination of Diethanolamine and Triethanolamine in Surface Finishing, Wastewater and Scrubber Solutions

INTRODUCTION

Alkanolamines are important in the chemical and pharmaceutical industries for production of emulsifying agents and the manufacturing of laundry additives and dyes. The analysis of alkanolamines is also important in metal surface finishing and in wastewater effluents. They are commonly used in acid gas removal systems (scrubbers) in both oil refineries and natural gas plants. Hydrogen sulfide and carbon dioxide are two of the primary acid gases formed in a refinery. When the gases are dissolved in an aqueous medium, they dissociate to form weak acids. Amines, weak bases, combine chemically with the weak acids to form salts, thus removing the acid gases from the process stream. When the amine solution becomes overloaded with salts, the efficiency of the scrubbing process is adversely affected. Thus, continuous monitoring of the amine solution can improve amine makeup, improve final product performance, and decrease system maintenance, Fig. 1. Monitoring soil and water samples in and around a refinery can identify sources of scrubber leaks before any serious losses or environmental contamination occurs, Fig. 2. Alkanolamines are also used in surface finishing as shown in Figure 3 to control the etching process of aluminum and aluminum alloys in the aerospace industry. [1, 2, 3, 4]

Several alternatives are currently available for the determination of alkanolamines, including wet chemistry, gas chromatography and traditional high performance liquid chromatography. However, these methods are time consuming, with sample preparation and analysis times as long as two hours. Because these compounds lack natural chromophores or fluorophores, derivatization of the alkanolamines is required prior to detection. As a result, the data collected by these alternate methods are prone to numerous matrix interferences and shortened column life. Ion chromatography works particularly well for separations of alkanolamines. The sensitivity by conductivity detection, however, is poor because of the low equivalent conductance of alkanolamines.

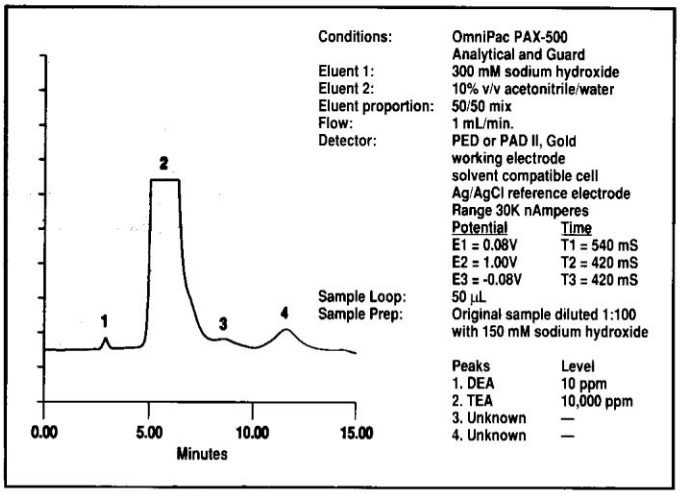

Figure 1. Diethanolamine Determination in an Oil Refinery Triethanolamine Scrubber Solution

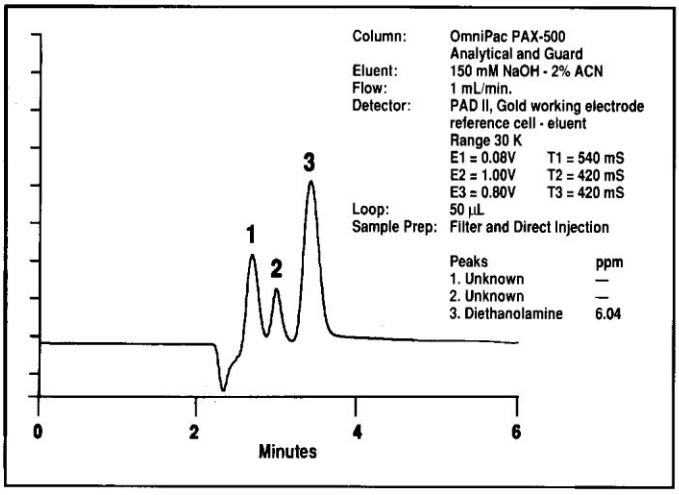

Figure 2. Oil Refinery Pond Water for Diethanolamine

RECOMMENDED EQUIPMENT

Dionex Series 4500i with a Pulsed Amperometric Detector (PAD II) or a Pulsed Electrochemical Detector (PED)

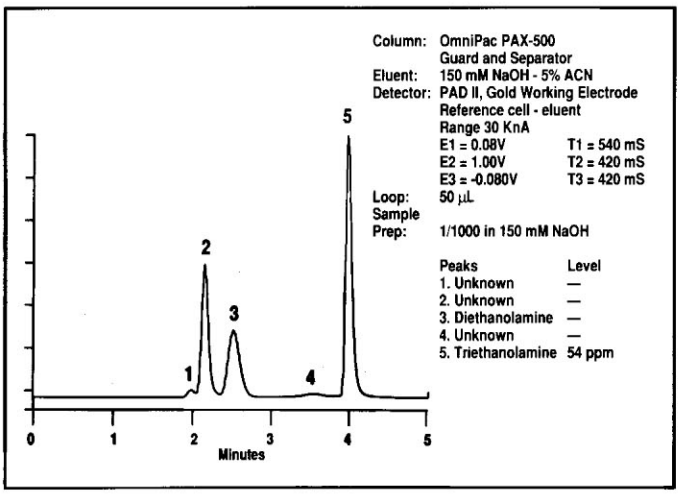

Figure 3. TEA Determination in an Operational Alkaline Etch Solution

PREPARATION OF REAGENTS AND STANDARDS

Columns:	OmniPac® PAX-500 Analytical and Guard
Eluent 1:	300mM sodium hydroxide
Eluent 2:	10% v/v acetonitrile/water
Eluent Proportion:	50% E1/50% E2
Flow:	1 mL/min.
Detector:	PED or PAD II, Gold working electrode Solvent compatible cell Ag/AgCl Reference electrode Range 30K nA

Potential	Time
E1 = 0.08V	T1 = 540 mS
E2 = 1.00V	T2 = 420 mS
E3 = -0.08V	T3 = 420 mS

Sample Loop:	50 µL
Sample Prep:	Dilute in 150 mM sodium hydroxide

It is recommended that all standards and samples be prepared and stored in 150mM sodium hydroxide. Alkanolamines are not stable at low pH and quickly degrade. Standards and samples prepared in deionized water also degrade but more slowly.

RESULTS AND DISCUSSION

Determination of total alkanolamines in this method is accomplished using liquid chromatography on Dionex OmniPac columns and Pulsed Amperometric Detection (PAD). The blended isocratic eluent is 150mM sodium hydroxide solution containing 5% acetonitrile. The sodium hydroxide in the eluent maintains a high pH thus suppressing ionization of the alkanolamines. Under these conditions the alkanolamines are retained and separated by a reversed phase mechanism on the OmniPac PAX-500 analytical column. The acetonitrile present in the eluent controls the retention of the alkanolamines. Increasing the acetonitrile concentration reduces the retention times.

Standards were prepared in 150 mM sodium hydroxide at the 0.1, 1, 10, 50, 75, and 100 ppm levels. Linearity over the range of 0.1 to 100 ppm using a 50 µL sample exhibited a coefficient of determination (r^2) greater than 0.998 for each alkanolamine. Precision, expressed as percent relative standard deviation (%RSD) for 374 replicate analyses of the alkanolamines at the 70 ppm level was better than 3% in an operational alkaline etch solution. The method detection limit by direct injection using a 50 µL loop has been shown to be 10 parts-per-billion in an alkaline etch solution. Peak areas were used in preference to peak height for quantification of the alkanolamines.

PRECAUTIONS

The eluent used in this method contains sodium hydroxide and acetonitrile. Acetonitrile in a high pH solution decomposes to weak organic acids and other compounds. These decomposition products, when present, interfere with the electrochemical detector's response. The acetonitrile decomposition reaction is not rapid, and small amounts of the breakdown components have no immediate effects upon the analysis. To avoid this decomposition reaction, on line low pressure mixing using the Dionex Gradient Pump is recommended to blend separate sodium hydroxide and acetonitrile solutions together.

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2. Burwell, K.F.; Dubek, D.J.; Sigmund, P.W., *Hydrocarbon Processing*, March 1982, 108-116.
3. Keaton, M.M.; Bourke, M.J., *Hydrocarbon Processing*, August 1983.
4. Campbell, D.C.; Carson, S.; Heberling, S.; Bramer, D.V., Improved Separation and Detection of Alkanolamines using Liquid Chromatography and Electrochemical Detection, Submitted to: *Journal of Chromatography.*

Cost-Effective Determination of Inorganic Anions and Cations in Municipal Drinking Water Using Capillary Ion Chromatography

INTRODUCTION

The determination of common inorganic anions and cations in drinking water is one of the most important applications of ion chromatography (IC) worldwide. IC has been approved for compliance monitoring of inorganic anions in United States (U.S.) drinking water since the mid-1980s, as described in U.S. Environmental Protection Agency (EPA) Method 300.0.[1] Many other industrialized countries have similar health and environmental standards and a considerable number of regulatory IC methods have been published worldwide (e.g., in Germany, France, Italy, and Japan). In addition, many standards organizations, including the International Organization for Standardization (ISO), American Society for Testing and Materials (ASTM), and American Water Works Association (AWWA), have validated IC methods for the determination of inorganic anions in drinking water.[2,3] The concentration of some anions in drinking water are regulated due to their toxic effects. For example, high levels of fluoride can cause skeletal and dental fluorosis, and nitrite and nitrate can cause methemoglobulinemia, which can be fatal to infants. Other common anions, such as chloride and sulfate, are considered secondary contaminants and can affect odor, color, and certain aesthetic characteristics in drinking water.

IC methods for dissolved alkali and alkaline earth metals and ammonia in drinking water are also important. Drinking water is frequently monitored for the presence of sodium under the U.S. EPA Safe Drinking Water Act. Ammonium is commonly a required target analyte for wastewater discharge permits, and is monitored in process wastewaters.

This study describes the determination of inorganic anions and cations in drinking water using the Thermo Scientific Dionex ICS-5000 capillary IC system. Scaling down from standard bore to capillary scale brings many benefits to IC analysts. Capillary Thermo Scientific Dionex Reagent-Free™ IC systems deliver fast turnaround from sample submission to results by reducing eluent preparation, system startup, and equilibration times. Perhaps most importantly, the system can be left on and ready for analysis at any time because of its low consumption of eluent (15 mL of source water a day). Having the system always on and ready for analysis significantly streamlines the workflow in IC. An always on system maintains stability and requires less frequent calibrations. The amount of waste generated is significantly decreased and the eluent generation cartridge producing the eluent lasts 18 months under continuous operation mode, which translates into reduced overall cost of ownership.

Figure 1 shows the determination of inorganic anions in drinking water using capillary IC. The inorganic anions were separated on the Thermo Scientific Dionex IonPac® AS19 capillary column and detected by suppressed conductivity detection. All anions were separated and eluted within 13 min. The relative standard deviation of peak area for each analyte was 0.6% when 60 injections were evaluated within 24 h.

Figure 2 shows the determination of inorganic cations in drinking water using capillary IC. The inorganic cations were separated on the IonPac CS12A capillary column and detected by suppressed conductivity detection. All cations of interests were determined within 12 min.

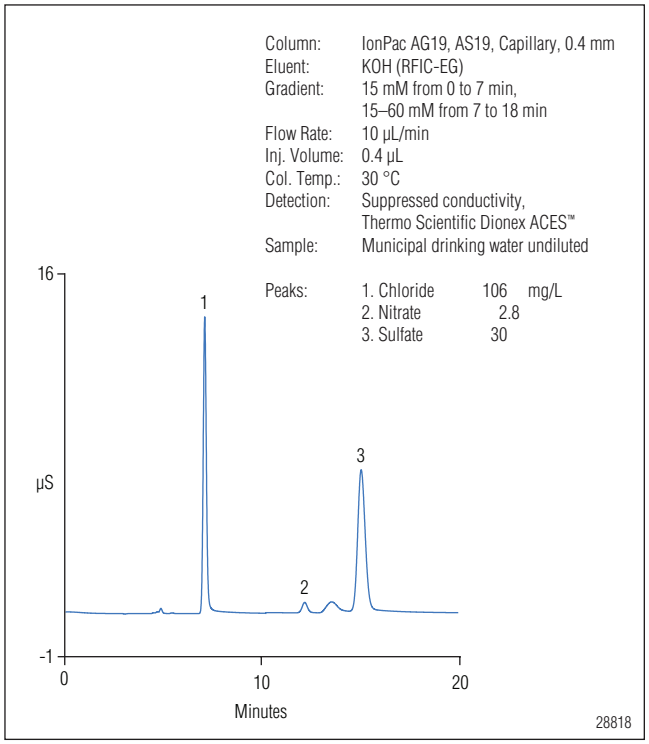

Figure 1. Anions in municipal drinking water on the IonPac AS19 capillary column.

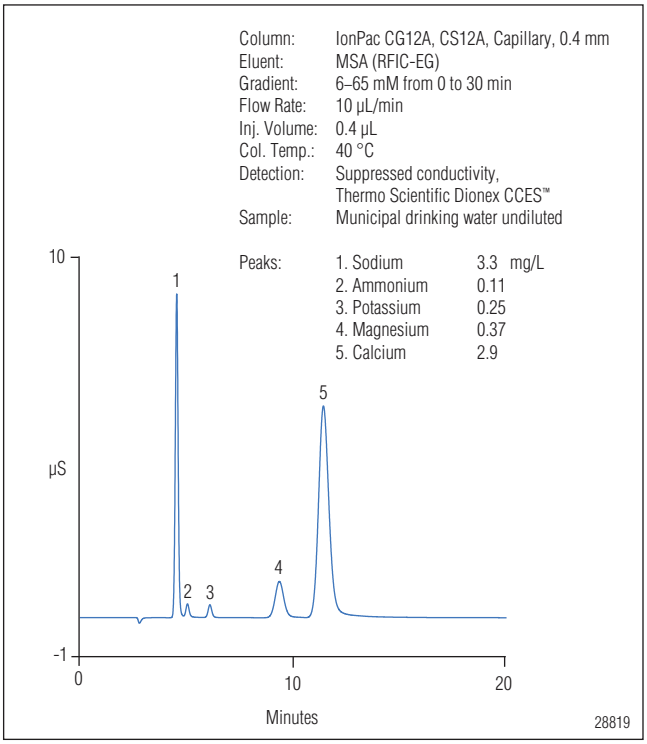

Figure 2. Cations in municipal drinking water on the IonPac CS12A capillary column.

CONDITIONS

The Dionex ICS-5000 capillary system, Thermo Scientific Dionex AS-AP Autosampler, and Thermo Scientific Dionex Chromeleon® Chromatography Data System software are used in this experiment. All experimental parameters are listed in Figures 1 and 2.

SAMPLE PREPARATION

Analyze municipal drinking water by capillary IC without sample pretreatment.

CONCLUSION

The introduction of the capillary Reagent-Free IC systems redefine the IC workflow for determination of inorganic anions and cations, providing enhanced mass sensitivity and ease of use.[4] These systems are a great solution for routine characterization of water samples with the always on, always ready capability simplifying the overall IC workflow.

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1. *The Determination of Inorganic Anions in Water by Ion Chromatography*; Method 300.0, Revision 2.1; U.S. Environmental Protection Agency: Cincinnati, OH, 1993.
2. Greenberg, A. E.; Clesceri, L. S.; Eaton, A. D., Eds.; *Standard Methods for the Examination of Water and Wastewater*, 18th ed.; American Public Health Association: Washington, DC, 1992.
3. *Standard Test Methods for Anions in Water by Chemically Suppressed Ion Chromatography*; D4327-97, Vol. 11.01; American Society for Testing and Materials; West Conshohocken, PA; 1999; pp 420–427.
4. Dionex Corporation, *Mass Sensitivity of Capillary IC Systems Explained.* Technical Note 90, LPN 2649, Sunnyvale, CA, 2011.

Analysis of Bromate, Haloacetic Acids, and Disinfection Byproducts

Environmental Water Applications Notebook

Determination of Bromate in Bottled Mineral Water Using the CRD 300 Carbonate Removal Device

INTRODUCTION

Drinking and bottled waters are commonly disinfected with ozone. Ozone is highly effective and, unlike many other disinfectants, does not remain in the water or change its taste. Unfortunately, when bromide is present in water, it is converted to bromate by the ozone treatment. Bromate is recognized as a potential human carcinogen, which has led to the regulation of its concentration in drinking and bottled water. Major regulatory bodies worldwide (e.g., U.S. EPA and the European Commission) have set a maximum allowable bromate concentration in drinking water of 10 µg/L.[1] In Europe, the limit was lowered to 3 µg/L for bottled natural mineral and spring waters disinfected by ozonation.[2]

Over the past two decades, Dionex has led the effort in developing sensitive and robust ion chromatography (IC) methods for determining bromate and other oxyhalides (e.g., chlorite and chlorate). U.S. EPA Method 300.0 (B) and 300.1 (B) used the IonPac® AS9-SC and IonPac AS9-HC columns, respectively, along with suppressed conductivity detection for bromate, chlorite, and chlorate determinations in drinking water. In 1997, Dionex introduced the AS9-HC column to allow the direct injection of 250 µL of drinking water to easily meet the 10 µg/L regulatory requirement. This method was documented in Dionex Application Note 81 (AN 81).[3] Since then, Dionex has developed a number of products and techniques, and worked with regulatory agencies and international standards organizations to improve the sensitivity and ruggedness of bromate determinations as well as the types of samples that can be directly injected.

Dionex products were instrumental in the development of the postcolumn derivatization techniques in U.S. EPA methods 317.0 and 326.0. These methods used the AS9-HC and Dionex suppression technology for conductivity detection of oxyhalides combined with postcolumn addition and absorbance detection for enhanced determination of bromate. EPA Methods 317.0 and 326.0 are documented in AN 136 and AN 149.[4,5] To improve the sensitivity for bromate using direct injection, Dionex developed the IonPac AS19 column. This column was designed for use with hydroxide eluents rather than the carbonate eluents used with the AS9-HC. Hydroxide eluents offer improved sensitivity for suppressed conductivity detection as compared to carbonate eluents. This improved sensitivity was documented in AN 167.[6] Hydroxide eluents are also advantageous because they can be generated easily using an eluent generator as part of a Reagent-Free™ IC (RFIC™) system. RFIC systems improve reproducibility and simplify analysis. The AS19 separation can also replace the AS9-HC separation in EPA Methods 317.0 and 326.0, which is documented in AN 168 and AN 171.[7,8] The AS19 was also used with an isocratic hydroxide eluent rather than the typical gradient for analysis of drinking water for bromate.[9] This method, presented in Application Update 154 (AU 154), cannot determine all the common inorganic anions in a single injection like the gradient method in AN 167. For determination of sub-µg/L concentrations of bromate in drinking water and higher ionic strength matrices without postcolumn

derivatization, Dionex developed a two-dimensional IC technique (AN 187) that uses an AS19 column in the first dimension, and an AS24 column, developed specifically for determining haloacetic acids and bromate by IC-MS and IC-MS/MS, in the second dimension.[10]

Dionex AN 184 showed that the AS19 method in AN 167 could be used to meet the 3 µg/L European limit for bromate in natural mineral and spring waters disinfected by ozonation.[11] The same application note compared the AS19 chromatography to chromatography with the AS23, a column that uses carbonate eluents and was designed to replace the AS9-HC. The AS23 has a higher capacity than the AS9-HC, and a different selectivity for the carbonate ion so that it is less likely to interfere with bromate determinations. AN 184 showed that poorer sensitivity associated with using carbonate eluents when compared to hydroxide eluents made the AS23 performance inferior to that of the AS19.

The present application note describes the use of a carbonate removal device, the CRD 300, to remove the majority of carbonate from the eluent and allow hydroxide-like performance and detection sensitivity. This device was used with the IonPac AS23 to determine bromate in a bottled mineral water samples. Detection sensitivity when using the CRD 300 was improved compared to chromatography without the CRD 300. Scientists responsible for water analysis can choose the column and eluent chemistry that best meets their needs to reliably determine bromate at concentrations below the common 10 µg/L regulatory limit.

EQUIPMENT

Dionex ICS-2000 Reagent-Free Ion Chromatography System* equipped with the following for carbonate/bicarbonate eluent generation:

EluGen® EGC II K_2CO_3 cartridge (P/N 058904)

EPM Electrolytic pH Modifier (P/N 063175)

EGC Carbonate Mixer (P/N 061686)

CRD 300 Carbonate Removal Device (4 mm) with VC Vacuum Pump (P/N 068474)

Chromeleon® 6.8 Chromatography Management Software

*This application can be run on any Dionex system equipped for carbonate/bicarbonate eluent generation.
Alternately, this application can be run with a manually prepared carbonate/bicarbonate eluent.

REAGENTS AND STANDARDS

Deionized water, type I reagent grade, 18 MΩ-cm resistivity or better

Sodium chlorite, 80% ($NaClO_2$, Fluka)

Potassium bromate ($KBrO_3$, Fluka)

Sodium chlorate ($NaClO_3$, Fluka)

Individual stock standards of fluoride, chloride, and sulfate, 1000 mg/L each (Merck)

PREPARATION OF SOLUTIONS AND REAGENTS

Carbonate Eluent Generation

The Eluent Generator (EG) produces the eluent using the EluGen EGC II K_2CO_3 cartridge, Electrolytic pH Modifier, EGC Carbonate Mixer, and deionized water supplied by the pump. The eluent concentration is controlled by the Chromeleon software. Backpressure tubing must be added to achieve 2300–2500 psi backpressure that will allow the EG degasser to function properly. See the ICS-2000 Operator's Manual Section 2.4.4, "Eluent Generator" for instructions on adding backpressure.

To set up the EGC II K_2CO_3, see the EGC II K_2CO_3 cartridge, Electrolytic pH Modifier, and EGC Carbonate Mixer Product Manual (Doc. No. 065075) for more information.

Manual Eluent Preparation

From Eluent Concentrate

Prepare 1 L of eluent by adding 10 mL of the Dionex IonPac AS23 Eluent Concentrate (P/N 064161) to a 1 L volumetric flask. Bring to volume with DI water and mix thoroughly.

From Manually Prepared Stock Solutions

Stock Carbonate/Bicarbonate Eluent Preparation

1.0 M Na_2CO_3 and 1.0 M $NaHCO_3$

Weigh 10.596 g sodium carbonate and 8.400 g sodium bicarbonate into separate 100 mL volumetric flasks. Bring each to volume with DI water.

IonPac AS23 Eluent (4.5 mM Na_2CO_3 /0.8 mM $NaHCO_3$)

For 1L, prepare by adding 4.5 mL of 1.0 M Na_2CO_3 and 0.8 mL of 1.0 M $NaHCO_3$ to a 1L volumetric flask, bring to volume with DI water, and mix thoroughly.

Stock Standard Solutions

Prepare 1000 mg/L stock standard solutions of fluoride, chloride, sulfate, chlorite, bromate, and chlorate by weighing 0.221 g, 0.165 g, 0.148 g, 0.168 g, 0.131 g, and 0.128 g, respectively, into separate 100 mL volumetric flasks. Bring each to volume with DI water.

Secondary Standards

The stock standards are used to prepare the 1000 µg/L secondary standards of chlorite, bromate, and chlorate. Take a defined volume of the stock standard and dilute it 1 to 1000 with DI water (e.g., dilute 100 µL to 100 mL in a 100 mL volumetric flask). Use these standards to prepare the working standards and to spike the bottled mineral water sample.

Working Standards

Prepare the standards for calibration and MDL studies by mixing defined volumes of the 1000 mg/L stock standard solutions of fluoride, chloride, and sulfate and the 1000 µg/L secondary standards of chlorite, bromate, and chlorate. For example, to prepare the working standard containing 0.5 mg/L fluoride, 50 mg/L chloride, 100 mg/L sulfate, and 40 µg/L of each of the oxyhalides, add 0.05 mL of the fluoride stock standard, 5 mL of the chloride stock standard, 10 mL of the sulfate stock standard, and 4 mL of each oxyhalide secondary standard to a 100 mL volumetric flask and bring to volume.

Sample

The bottled mineral water sample was purchased from a local market in Bangkok, Thailand and was bottled at its source in the mountains of Thailand. The label reported the presence of fluoride, chloride, sulfate, and bicarbonate, but not their concentrations.

CRD 300 IN VACUUM MODE SETUP

The CRD 300 in vacuum mode uses a vacuum pump to evacuate the regenerant chamber of the CRD 300 so that CO_2 gas is literally sucked out of the eluent. A bleed tube feeds a trickle of fresh air into the regenerant chamber to constantly sweep out the CO_2 gas. To operate the CRD 300 in vacuum mode, mount the CRD 300 directly on top of the suppressor and plumb the eluent from the Eluent Out of the suppressor to the Eluent In of the CRD 300. The Eluent Out of the CRD 300 is connected to the conductivity cell In and conductivity cell Out goes to waste if the system is running in external water mode. If the system is operated in recycle mode, connect conductivity cell Out to the suppressor Regen In. Connect the vacuum tubing to the vacuum port of the vacuum pump and to the ballast bottle. Connect a length of 1/8" Teflon® tubing from the ballast bottle to the Regen Out of the CRD 300. Make sure the third port on the ballast bottle is closed and air tight. Connect 15 cm of red (0.005" i.d.) PEEK™ tubing to the Regen In of the CRD 300; this is the air bleed assembly. Begin eluent flow before beginning vacuum operation. When eluent flow is established, turn on the vacuum pump. The background conductivity should drop almost immediately. When the eluent pump is turned off, immediately turn off the vacuum pump. Avoid operating the vacuum pump while eluent flow is stopped. A TTL can be wired to automate stopping the vacuum pump.

CONDITIONS

Condition A (Eluent Generation and CRD 300)

Column:	IonPac AS23 (4 × 250 mm) (P/N 064149) IonPac AG23 (4 × 50 mm) (P/N 064147)
Eluent:	EGC II K_2CO_3 (P/N 058904) EPM (P/N 063175) 4.5 mM K_2CO_3/0.8 mM $KHCO_3$
Flow Rate:	1.0 mL/min
Inj. Volume:	250 µL
Temperature:	30 °C
Suppressor:	Suppressed conductivity, ASRS® 300, 4 mm (P/N 064554), external water mode, 25 mA CRD 300, 4 mm, (P/N 064637) vacuum mode
Background:	< 1.5 µS
Noise:	~ 0.3 nS
Back Pressure:	~2200 psi

Condition B (Manual Eluent Preparation and no CRD 300)

Column:	IonPac AS23 (4 × 250 mm) (P/N 064149) IonPac AG23 (4 × 50 mm) (P/N 064147)
Eluent:	4.5 mM Na_2CO_3/0.8 mM $NaHCO_3$
Flow Rate:	1.0 mL/min
Inj. Volume:	250 µL
Column Temp:	30 °C
Suppressor:	Suppressed conductivity, ASRS 300, 4 mm (P/N 064554), external water mode, 25 mA
Background:	17-19 µS
Noise:	~ 3.0 nS
Back Pressure:	~1800 psi

RESULTS AND DISCUSSION

Chromatography

Bromate, chlorite, and chlorate were resolved from seven common inorganic anions using an IonPac AS23 column under its recommended eluent conditions (4.5 mM Na_2CO_3/0.8 mM $NaHCO_3$). Chromatogram B in Figure 1 shows this separation. The background conductivity after suppression using the carbonate eluent is between 18 and 19 µS. The higher the background, the higher the noise, and this results in a lower signal-to-noise ratio (i.e., lower sensitivity). The background of the suppressed hydroxide eluent used for the IonPac AS19 column is < 1 µS. In order for the carbonate eluent system of the AS23 to approach the detection limits delivered by the hydroxide eluent system of the AS19, the background must be reduced. The CRD 300 was designed to remove carbonate from the eluent (after suppression) and thereby reduce the background to improve detection limits. Chromatogram A shows the same AS23 separation as B using a CRD 300. Note that the background has been reduced to about 1 µS, the injection dip at about 2 min is greatly reduced in size, and there is a noticeable improvement in analyte sensitivity. Throughout this application note, we compare the determination of bromate, chlorite, and chlorate with the AS23 and suppressed conductivity, both with and without the CRD 300.

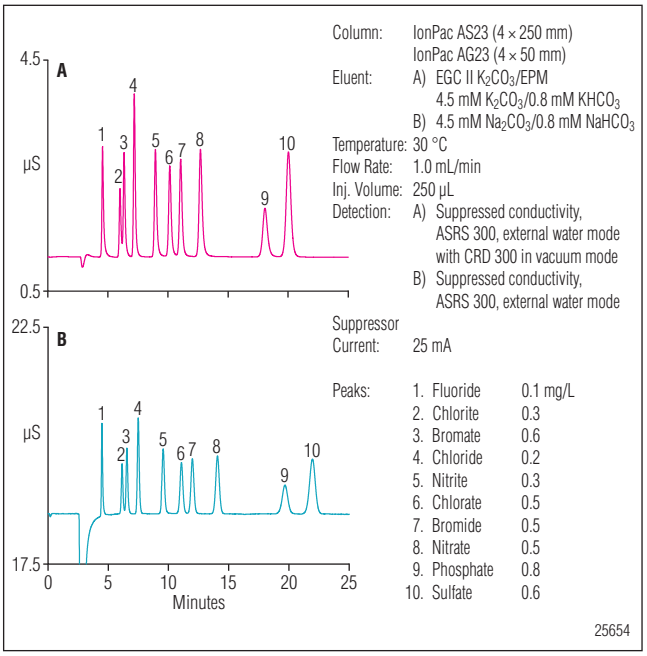

Figure 1. Chromatography of a mixed anion standard A) with a CRD 300 and electrolytically prepared eluent, and B) without a CRD 300 and with manually prepared eluent.

Figure 2 shows single injections from the MDL determinations of bromate, chlorite, and chlorate with and without the CRD 300. Fluoride, (0.5 mg/L), chloride (50 mg/L), and sulfate (100 mg/L) were added to the MDL standards to simulate the ionic strength of bottled water samples. Due to the higher background and noise of the system without the CRD 300 (Chromatogram B, Figure 2), higher analyte concentrations were used for the MDL test compared to the system with the CRD 300. Table 1 shows the results of the MDL determination. For all three oxyhalide analytes, the MDL is lower for the system with the CRD 300. The MDL values without the CRD 300 are similar to those determined with the AS23 in AN 184. The values when using the CRD 300, though lower than without, are not as low as those determined with the AS19 and hydroxide eluent in AN 184.

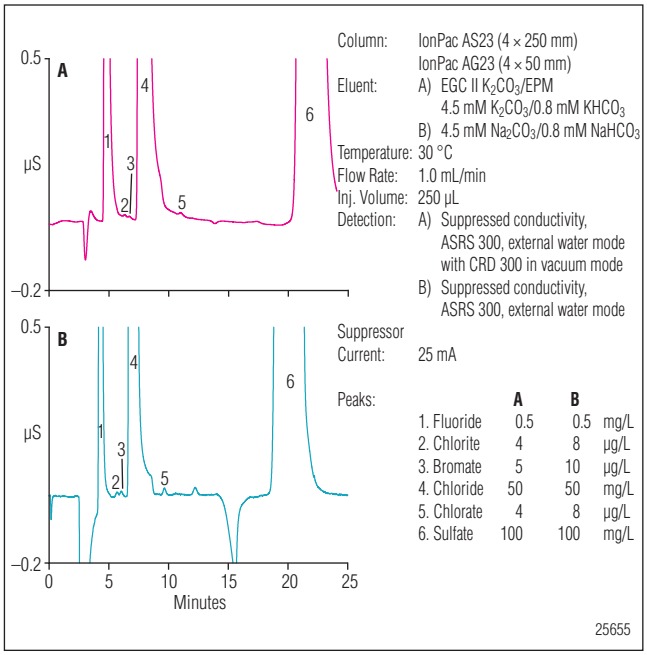

Figure 2. Example chromatograms from the MDL determination A) with a CRD 300, and B) without a CRD 300.

Table 1. MDL Determinations of Chlorite, Bromate, and Chlorate with and without a CRD 300

Injection No.	Height (µS)					
	With CRD 300			Without CRD 300		
	Chlorite	Bromate	Chlorate	Chlorite	Bromate	Chlorate
	4 µg/L	5 µg/L	4 µg/L	8 µg/L	10 µg/L	8 µg/L
1	0.0057	0.0041	0.0076	0.0099	0.0121	0.0189
2	0.0051	0.0042	0.0071	0.0114	0.0128	0.0199
3	0.0053	0.0042	0.0065	0.0093	0.0115	0.0204
4	0.056	0.0043	0.0074	0.0105	0.0132	0.0215
5	0.059	0.0047	0.0074	0.0111	0.0133	0.0205
6	0.0061	0.0045	0.0077	0.0103	0.0125	0.0201
7	0.0057	0.0042	0.0076	0.0114	0.0111	0.0199
Average	**0.0056**	**0.0043**	**0.0073**	**0.0105**	**0.0124**	**0.0202**
RSD	**5.97**	**5.04**	**5.42**	**7.41**	**6.67**	**3.94**
MDL (µg/L)	**0.75**	**0.79**	**0.68**	**1.86**	**2.10**	**0.99**

Another calibration was performed for both systems using consistent concentrations of fluoride, chloride, and sulfate (0.5 mg/L, 2 mg/L, and 10 mg/L, respectively) in standards with three levels of chlorite, bromate, and chlorate concentrations; 10, 20, and 40 µg/L. Overlays of three calibration standards are shown in Figure 3 and the results are in Table 2. The calibration data are equivalent.

Both systems were used to analyze a bottled mineral water sample from the mountains of Thailand. Figure 4 shows the analysis of this sample and Table 3 reports the results of the analysis. The sample had just over 10 µg/L bromate and 1–2 µg/L chlorate, suggesting a second disinfection process besides ozonation was used. Due to the noise of the system without the CRD 300, the chlorate peak could not be identified with confidence. To evaluate accuracy, known amounts of bromate, chlorite, and chlorate were spiked into the bottled mineral water sample. Figure 5 shows the chromatography from this study and Table 4 shows that all analytes were recovered at >85%. In this experiment, the recovery was better for the system with the CRD 300.

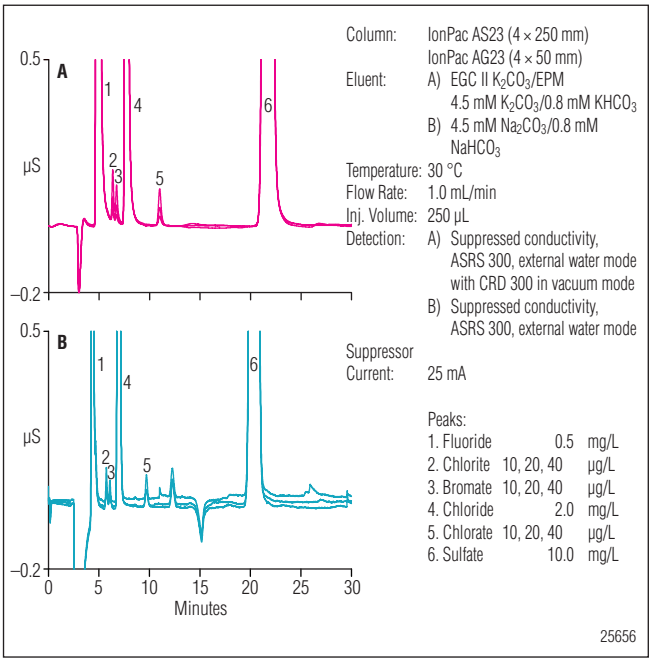

Figure 3. Overlay of chromatograms of three concentration levels of chlorite, bromate, and chlorate in a mixed anion standard A) with a CRD 300, and B) without a CRD 300.

Table 2. Chromeleon Calibration Report for Chlorite, Bromate, and Chlorate with and without a CRD 300

Peak Name	Points	R-Square (%)	
		With CRD 300	Without CRD 300
Chlorite	3	99.9961	99.9748
Bromate	3	100.0000	99.9986
Chlorate	3	99.9995	99.9637

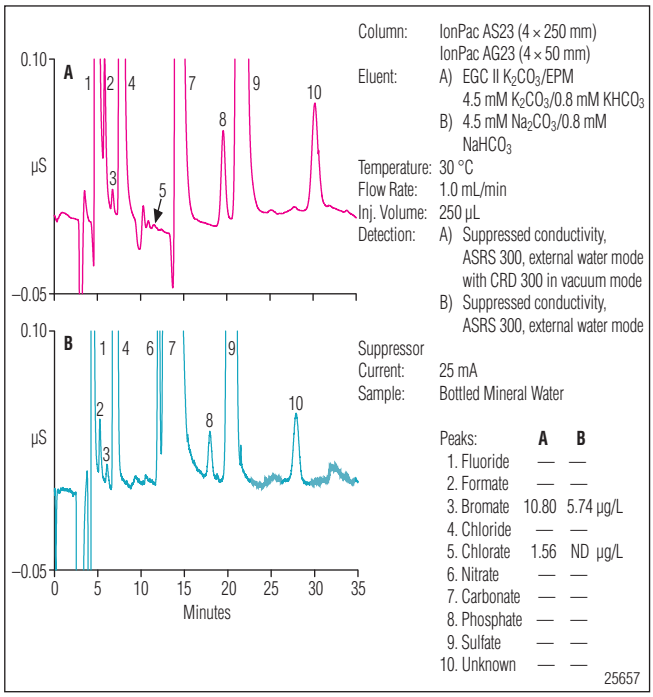

Figure 4. Chromatography of a bottled mineral water sample A) with a CRD 300, and B) without a CRD 300.

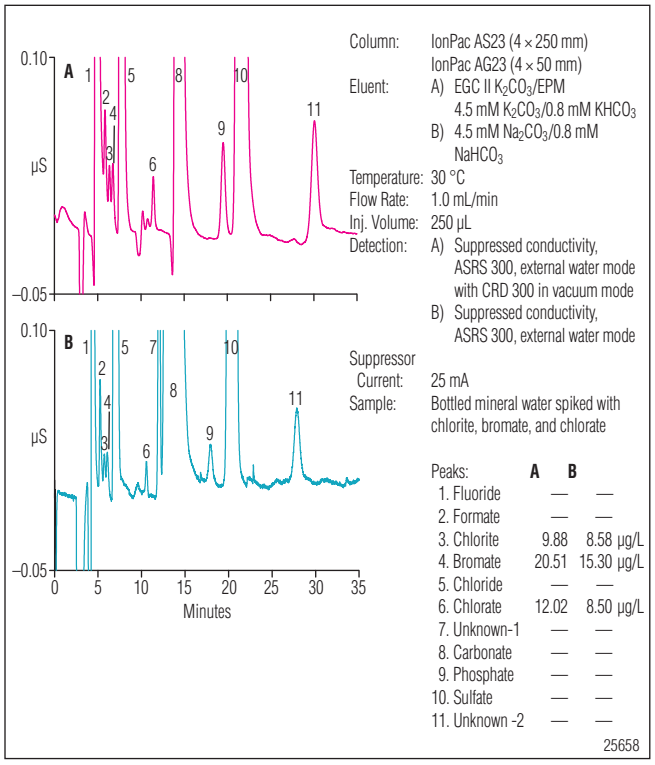

Figure 5. Chromatography of a bottled mineral water sample spiked with chlorite, bromate, and chlorate (10 µg/L each) A) with a CRD 300, and B) without a CRD 300.

Table 3. Determination of Bromate and Chlorate in a Bottled Mineral Water Sample with and without a CRD 300

Injection No.	With CRD 300 (µg/L)		Without CRD 300 (µg/L)	
	Bromate	Chlorate	Bromate	Chlorate
1	11.0	1.52	5.33	ND
2	10.9	1.55	6.23	ND
3	10.9	1.35	5.02	ND
4	10.1	1.91	6.25	ND
5	11.3	1.48	5.89	ND
Average	10.8	1.56	5.74	—
RSD	4.34	13.42	9.61	—

Table 4. Spike Recovery of Chlorite, Bromate, and Chlorate with and without a CRD 300

	With CRD 300			Without CRD 300		
	Chlorite	Bromate	Chlorate	Chlorite	Bromate	Chlorate
Sample	ND[a]	10.83	1.56	ND[a]	5.74	ND[a]
Spike	10	10	10	10	10	10
Measured[b] Amount	9.88	20.51	12.02	8.58	15.30	8.50
RSD	2.39	1.60	2.45	2.39	1.60	2.45
Recovery(%)	98.8	98.5	104	85.8	97.2	85.0

[a] ND = Not Detected

[b] The average of five injections

SUMMARY

This application note shows that using the CRD 300 with the IonPac AS23, bromate can be determined in bottled mineral water at concentrations < 5 µg/L. The method sensitivity for bromate and other oxyhalides approaches that of the hydroxide eluent system featured in Dionex Application Note 184.

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Determination of Sub-µg/L Bromate in Municipal and Natural Mineral Waters Using Preconcentration with Two-Dimensional Ion Chromatography and Suppressed Conductivity Detection

INTRODUCTION

Ozone is a powerful drinking water disinfectant that is effective in treating chlorine resistant organisms, such as *Cryptosporidia*.[1] For bottled water, ozonation is generally preferred over other available disinfection treatment methods because it does not leave a taste or residual disinfectant, due to the short lifetime of ozone.[2,3] Ozone also improves the quality of finished drinking water, by reducing filtered water turbidity and decreasing the formation of many halogenated disinfection by-products. However, ozonation of drinking water containing bromide can result in the formation of the disinfection by-product bromate, a potential human carcinogen even at low µg/L concentrations.[4] The U.S. EPA and European Commission have established a regulatory maximum contaminant level (MCL) of 10 µg/L bromate in drinking waters.[5,6] In the U.S., bottled water is considered a food product and is therefore regulated by the U.S. Food and Drug Administration (FDA) under the Federal Food, Drug, and Cosmetic Act.[7] In 2001, the U.S. FDA also established an MCL of 10 µg/L bromate in bottled drinking water.[7] More recently, the European Commission set a lower MCL of 3 µg/L bromate for natural mineral waters and spring waters treated by ozonation.[8] However, these limits were based on the feasibility of detection and removal, even though studies suggest concentrations lower than 1 µg/L pose increased lifetime cancer risks.[4,9]

Published EPA methods for determining low concentrations of bromate in drinking waters using direct injection have focused primarily on using columns designed specifically for carbonate eluents combined with suppressed conductivity detection or postcolumn reaction followed by UV/Vis detection.[10-12] Dionex Application Note 167 demonstrated that the use of a high-capacity hydroxide-selective IonPac® AS19 column, an electrolytically generated hydroxide eluent, a large loop injection, and suppressed conductivity detection can significantly reduce the bromate detection limit from 1.4 µg/L, reported in EPA Method 300.1, to 0.34 µg/L.[13] In addition, the use of a hydroxide eluent produced a bromate MDL of <0.2 µg/L for absorbance detection after postcolumn addition using EPA Methods 317.0 and 326.0.[14,15]

However, determining low concentrations of bromate in high ionic strength matrices using suppressed conductivity detection is subject to potential interferences and loss of sensitivity. Although postcolumn reaction methods do not generally suffer from interferences by common anions, column overloading with high ionic strength samples can still cause peak broadening and an associated loss of response. In particular, natural mineral waters typically contain elevated levels of common anions that can significantly exceed the concentrations present in most municipal drinking water samples, presenting an additional challenge for the currently available methods to determine <1 µg/L bromate. Natural mineral waters previously have been analyzed for trace concentrations of bromate using the the IonPac AS19 column with a hydroxide eluent or the IonPac AS23 column and a carbonate/bicarbonate eluent using a large loop injection and suppressed conductivity detection.[16]

In this application note, we demonstrate the use of a two-dimensional (2-D) ion chromatography (IC) system for the determination of trace concentrations of bromate in municipal and natural mineral waters with high ionic strength matrices. The first dimension uses a high capacity

4-mm IonPac AS19 column to resolve the bromate from the matrix ions. The matrix ions are diverted to waste while a 2 mL plug (cut volume) containing the bromate is transferred to the second dimension for analysis. Bromate is well resolved in the second dimension using a 2-mm IonPac AS24 column. This method is fully automated using an ICS-3000 Reagent-Free™ IC (RFIC™) system. In addition, this 2-D IC method achieves bromate detection limits equivalent to or better than postcolumn addition methods. The 2-D IC method avoids the cost and disposal of the chemicals required for postcolumn configurations and simplifies the experimental setup. Additionally, it avoids potential column overload during analysis of high ionic strength matrices.

EQUIPMENT

Dionex ICS-3000 Reagent-Free Ion Chromatography system consisting of:
- DP Dual Pump module
- EG Eluent Generator module with a dual setup
- DC Detector/Chromatography module
- (single or dual temperature zone configuration)

AS Autosampler with a 5 mL syringe (P/N 053915), 8.2 mL sampling needle assembly (P/N 061267)

Two EluGen® EGC II KOH cartridges (P/N 058900)

Two Continuously-Regenerated Anion Trap Columns, CR-ATC (P/N 060477)

Four 4-L plastic bottle assemblies for external water mode of operation

Chromeleon® 6.8 Chromatography Management Software

REAGENTS AND STANDARDS

Deionized water, Type I reagent grade, 18 MΩ-cm resistivity or better

Bromate standard (1000 mg/L, Ultra Scientific, VWR P/N ULICC-010)

Sodium bromate ($NaBrO_3$) (EM Science SX0385-1)

Sodium chloride (NaCl) (J.T. Baker; VWR P/N JT3625-1)

Sodium nitrate ($NaNO_3$) (Fisher Scientific S343-500)

Sodium bicarbonate ($NaHCO_3$) (EM Science SX0320-1)

Sodium sulfate (Na_2SO_4) (Aldrich 29,931-3)

Sodium phosphate, dibasic, anhydrous (Na_2HPO_4) (JT Baker 4062-1)

CONDITIONS

First Dimension

Columns:	IonPac AG19 guard, 4 × 50 mm (P/N 062887) IonPac AS19 analytical, 4 × 250 mm (P/N 062885)
Eluent:	10 mM potassium hydroxide 0–12 min,[a] step to 65 mM at 12 min, 65 mM 12–35 min[b]
Eluent Source:	EGC II KOH with CR-ATC
Flow Rate:	1 mL/min
Temperature:	30 °C (lower compartment) 30 °C (upper compartment)
Injection Vol:	1000 µL
Detection:	Suppressed conductivity, ASRS® ULTRA II (4 mm), AutoSuppression® external water mode (flow rate: 3–5 mL/min) Current setting: 161 mA
System Backpressure:	~2300 psi
Expected Background Conductance:	<0.5 µS
Noise:	~1–2 nS/min peak-to-peak
Run Time:	35 min

[a]The step change described here should occur after the valve on system #2 has switched from the load to inject position.
[b]The method equilibrates for 5 min at 10 mM KOH prior to injection.

Second Dimension

Columns:	IonPac AG24 guard, 2 × 50 mm (P/N 064151) IonPac AS24 analytical, 2 × 250 mm (P/N 064153)
Eluent:	10 mM potassium hydroxide 0–24 min, step to 65 mM at 24 min, 65 mM 24–35 min[b]
Eluent Source:	EGC II KOH with CR-ATC
Flow Rate:	0.25 mL/min
Temperature:	30 °C (lower compartment) 30 °C (upper compartment)
Cut Volume:	2 mL (on the concentrator column)
Concentrator:	TAC-ULP1, 5 × 23 mm (P/N 061400)

Detection:	Suppressed conductivity, ASRS ULTRA II (2 mm), AutoSuppression external water mode (flow rate: 1–3 mL/min) Current setting: 41 mA
System Backpressure:	~2400 psi
Expected Background Conductance:	<0.8 µS
Noise:	~2–3 nS/min peak-to-peak
Run Time:	35 min

[b]*The method equilibrates for 5 min at 10 mM KOH prior to injection.*

PREPARATION OF SOLUTIONS AND STANDARDS

Stock Bromate Standard Solution

Dissolve 0.1180 g sodium bromate in 100 mL of deionized water for a 1000 mg/L standard solution. This standard is stable for at least six months when stored at 4 °C.

Bromate Primary Dilution Standard

To prepare a 10 mg/L bromate solution, add 1 mL of the 1000 mg/L stock standard to a 100 mL volumetric flask. Bring to volume with deionized water. When stored at 4 °C, the resulting solution is stable for at least one month.

Bromate Secondary Dilution Standard

To prepare a 1 mg/L bromate solution, add 10 mL of the primary dilution standard to a 100 mL volumetric flask. Bring to volume with deionized water. When stored at 4 °C, the resulting solution is stable for at least one month.

Bromate Calibration Standards

To prepare bromate calibration standards at concentrations of 0.15, 0.25, 0.50, 1, 3, 5, 10, and 15 µg/L, add the appropriate volumes of the bromate secondary dilution standard to separate 100 mL volumetric flasks. Bring to volume with deionized water.

Common Anion Stock Solutions

Prepare 1000 mg/L each of chloride, nitrate as N, bicarbonate, sulfate, and phosphate as P. Dissolve 0.1649 g sodium chloride in deionized water and dilute to 100 mL. Dissolve 0.6068 g sodium nitrate in deionized water and dilute to 100 mL. Dissolve 0.1377 g sodium bicarbonate in deionized water and dilute to 100 mL. Dissolve 0.1479 g sodium sulfate in deionized water and dilute to 100 mL. Dissolve 0.4583 g anhydrous sodium phosphate, dibasic in deionized water and dilute to 100 mL.

Laboratory Synthetic Sample Matrix (LSSM)

The LSSM contains 100 mg/L each of chloride, bicarbonate, and sulfate and 10 mg/L each of nitrate-N and phosphate-P. Prepare this solution by adding 10 mL each of chloride, bicarbonate, and sulfate from their respective 1000 mg/L stock solutions to a 100 mL volumetric flask. Add 1 mL each of nitrate-N and phosphate-P from their respective 1000 mg/L stock solutions to the volumetric flask containing chloride, bicarbonate, and sulfate. To fortify this solution with 0.5 µg/L or 5 µg/L bromate add 0.05 mL or 0.5 mL, respectively of the 1 mg/L bromate secondary dilution standard to the volumetric flask and bring to volume with deionized water.

SYSTEM PREPARATION AND SETUP

Install an EGC II KOH cartridge for each system channel. Install backpressure tubing temporarily in place of the columns on both system channels to produce a total backpressure of 2000–2500 psi at a flow rate of 1 mL/min. Condition the cartridges by setting the KOH concentration to 50 mM at 1 mL/min for 30 min. After completing the conditioning process, disconnect the backpressure tubing. Install a CR-ATC between the EGC II KOH cartridge and the EGC degas. Hydrate the CR-ATC prior to use by following the instructions outlined in the EluGen Cartridge Quickstart Guide (Document No. 065037-02). Figure 1 shows a detailed schematic diagram of the system setup.

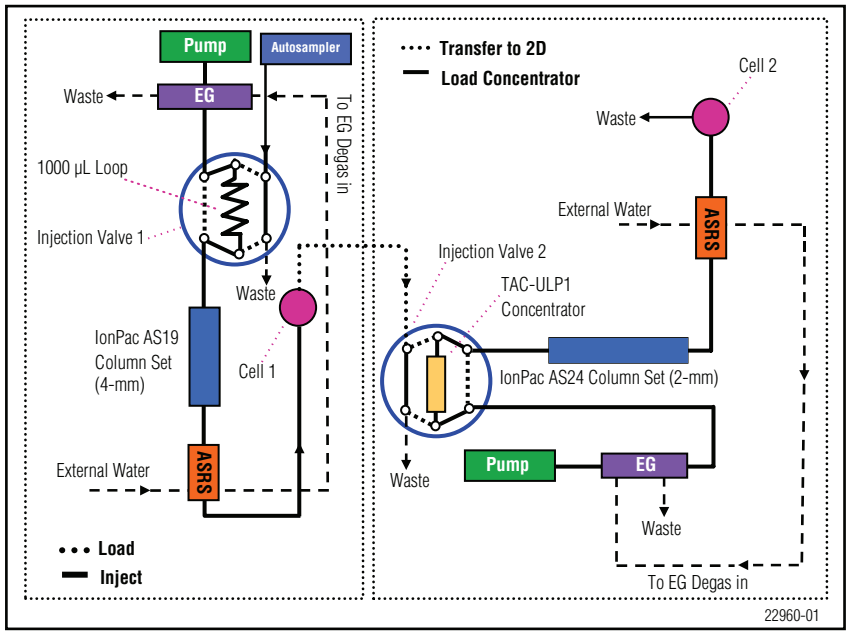

Figure 1. Schematic diagram of an ICS-3000 two-dimensional ion-chromatography system for the determination of trace concentrations of bromate.

Install an 8.2 mL sampling needle assembly (P/N 061267) is also required for operation. The largest injection possible with a 5 mL syringe installed on the AS Autosampler is 4000 µL. To inject 1000 µL, select the normal mode from the front panel of the autosampler. The normal mode will allow the autosampler to flush the sample loop prior to injection. Enter the correct Sample Loop Size and Sample Syringe Volume in the AS Plumbing Configuration screen. Instruct the AS to inject 1000 µL with the Chromeleon software.

Prepare a 1000 µL sample loop by measuring approximately 86.4 in. of 0.030 in. i.d. tubing. To verify the volume of the loop, first weigh the empty tubing. Fill the tube with deionized water then reweigh the filled tube and calculate the volume. The total sample volume should be 1000 µL ± 5%. Install the sample loop on Injection Valve 1 of the DC-3000.

Because the two dimensions are working as one system, a second timebase does not need to be created. However, to allow independent control of the DC-3000 injection valves in the timebase, the DC settings in the Chromeleon system configuration must be changed. To modify this configuration, go to the DC High Pressure Valves tab in the system configuration, double-click InjectValve_2, and change controlled by AS to DC.

Install a 4 × 50 mm IonPac AG19 and a 4 × 250 mm IonPac AS19 column on system #1 in the lower compartment of the DC. Install a 2 × 50 mm IonPac AG24 and a 2 × 250 mm IonPac AS24 column on system #2. Connect a piece of 0.01 in. i.d. PEEK™ tubing from the Cell Out on system #1 to the Sample Inlet Port on Injection Valve #2. The length of this tubing should be kept to a minimum. Install a 5 × 23 mm TAC-ULP1 concentrator in place of the sample loop on system #2. The direction of sample loading on the TAC-ULP1 should be in the opposite direction of the analytical flow.

Make sure the pressure for both systems is from 2200–2500 psi using the operating conditions described earlier to allow the degas assembly to effectively remove electrolysis gases from the eluent. If necessary, install additional backpressure tubing between the degas assembly and the injection valve to achieve the recommended pressure setting. Monitor the pressure periodically as it can gradually rise over time. To reduce pressure, trim the backpressure tubing.

Hydrate the ASRS ULTRA II suppressors prior to installation using a disposable plastic syringe. Push 3 mL of degassed deionized water through the Eluent Out port and 5 mL of degassed deionized water through the Regen In port. Allow the suppressors to stand for 20 min to fully hydrate the suppressor screens and membranes. Before installing the suppressors, rinse the analytical column with 65 mM KOH while diverting to waste. Install the ASRS ULTRA II for use in the external water mode

by connecting the Regen Out of the suppressor to the Regen In of the CR-ATC and connect the Regen In of the suppressor to the external water source. The Regen Out of the CR-ATC is connected to the Regen In of the EG degasser.

Equilibrate the AS19 and AS24 columns with 65 mM KOH at their respective flow rates, shown in the Conditions section, for approximately 60 min. Analyze a matrix blank by injecting 1000 µL deionized water using 8–10 min as the default cut time. An equilibrated system has background conductances of < 0.5 µS and < 0.8 µS for the AS19 and AS24 columns, respectively. Determine the final cut time (preconcentration time) for the second dimension, as described in the next section, before injecting a bromate standard.

Determining the Cut Time for the Second Dimension

Because there may be slight variations in system plumbing, column capacity, and tubing lengths, individual laboratories should first determine the optimum cut time (from the first dimension) before determining bromate in the second dimension. To determine the cut time for analysis in the second dimension, we recommend performing duplicate 1000 µL injections of 15 µg/L bromate prepared in deionized water and 15 µg/L bromate prepared in a LSSM containing 100 mg/L each of chloride, sulfate and bicarbonate and 10 mg/L each of nitrate-N and phosphate-P.

For this application, it is important that valve #2 on the second dimension remains in the inject position during this time to avoid any baseline disturbances that may occur in the first dimension. This can be accomplished by placing a semicolon (";") before the DC inject commands in the Chromeleon program.

Determine the start time for placing valve #2 in the load position by subtracting 1 min from the retention time (RT) of bromate in the LSSM sample (in this experiment, bromate RT = 9 min – 1 min = 8 min). Determine the time for switching valve #2 to the inject position by adding 0.2 min to the time when the bromate peak in deionized water returns to the baseline (in this experiment, bromate returned to baseline = 9.8 min + 0.2 min = 10.0 min). The chromatograms used for the present experiment are shown in Figures 2A and 2B.

Note: it is important to verify the retention time of bromate on the AS19 column weekly to ensure good trapping efficiency on the TAC-ULP1 concentrator.

After the cut time has been established, enable valve #2 by removing the semicolons prior to the DC inject commands in the Chromeleon program. Perform duplicate injections of a 5 µg/L bromate standard to verify that nearly identical bromate retention times are achieved

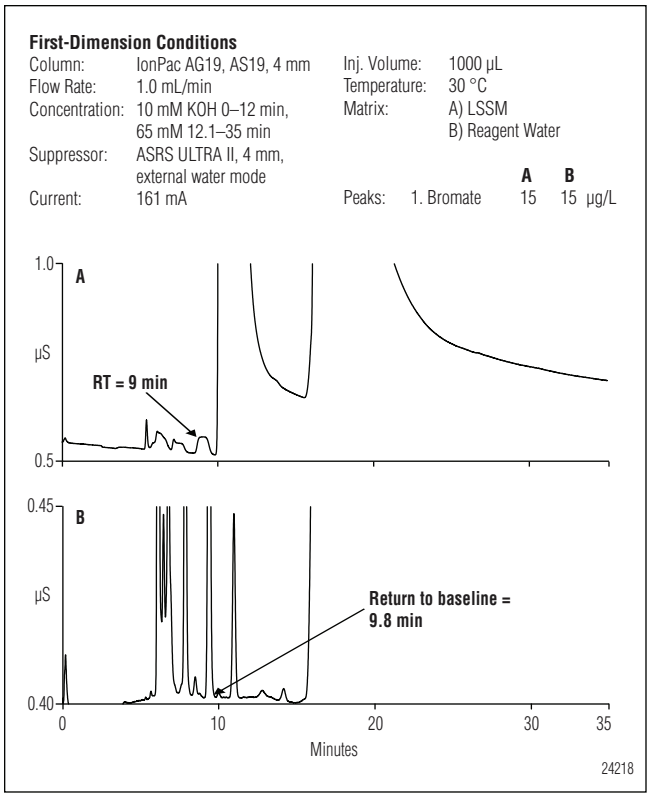

Figure 2. Determination of the cut window in the first dimension.

on the AS24 column in the second dimension. Figure 3 shows an example chromatogram of 5 µg/L bromate separated on the AS24 column in the second dimension.

RESULTS AND DISCUSSION

The bromate in the second dimension was calibrated by injecting a 1000 µL water blank and a duplicate injection of eight calibration standards in the first dimension to cover the desired concentration range. The peak area response generated by the calibration standards was tabulated against the bromate concentration using a quadratic regression curve. Table 1 summarizes the calibration data obtained from injecting standards in the range of 0.15-15 µg/L bromate. The accuracy of the calibration curve was verified by injecting a 5 µg/L bromate standard prepared from a second source, producing a calculated recovery of 97.1%.

Table 1. Calibration Data and Method Detection Limits for Bromate

Analyte	Range (µg/L)	Linearity[a] (r^2)	MDL Standard (µg/L)	SD (µg/L)	Calculated MDL (µg/L)
Bromate	0.15–15	0.9995	0.20	0.012	0.036

[a] Quadratic fit

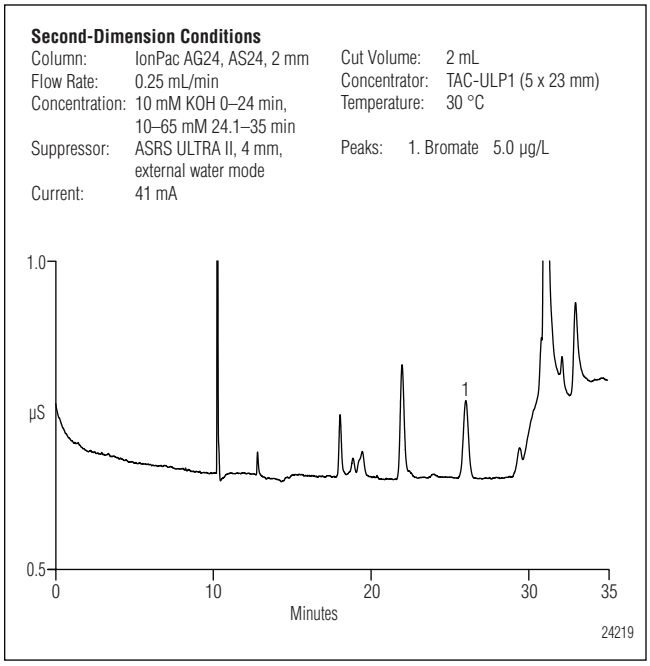

Figure 3. Chromatogram of a 5 µg/L bromate standard with the IonPac AS24 column in the second dimension.

During the initial development of the 2-D method for determining trace concentrations of bromate, the lowest concentration minimum reporting level (LCMRL) was determined. The U.S. EPA has developed a statistical approach for determining a single-laboratory LCMRL using linear regression and prediction intervals and has found this to be a more systematic procedure for determining the minimum reporting level (MRL).[17] The MRL is the lowest analyte concentration that demonstrates known quantitative quality, whereas the LCMRL is the lowest true concentration at which the future recovery is predicted to fall with a 99% confidence between 50% and 150% recovery. The LCMRL can be used to determine the MRL for a particular analyte by either using a multiplying factor or by combining results from a multi-laboratory study. Although the EPA encourages all laboratories to determine the LCMRL to aid in evaluating the performance of spiked recoveries at or below the MRL, it does not mandate LCMRL determinations.

In this study, the LCMRL was determined to be 0.15 µg/L bromate by preparing and analyzing seven individual replicate injections of 0.15, 0.20, 0.30, 0.40, and 0.50 µg/L bromate. The data from these replicate injections were then inserted into the statistical program provided on the EPA website (http://www.epa.gov/OGWDW/methods/sourcalt.html) to determine the LCMRL. The target MRL for this application was established at 0.50 µg/L bromate, which is just over three times the calculated LCMRL.

EPA Method 314.2, also using 2-D IC, does not require the determination of the detection limit for validation of the method. However, some laboratories may require this determination due to the various regulatory bodies associated with compliance monitoring. The limit of detection (LOD) of the 2-D method was determined for bromate by performing seven replicates of reagent water fortified with 0.20 µg/L bromate and using the following equation:

$$LOD = St_{(n-1,\ 1-\alpha=0.99)}$$

where:

$t_{(n-1,\ 1-\alpha=0.99)}$ = student's *t*-value for a 99% confidence level with n - 1 (t = 3.14 for seven replicate injections)

n=number of replicates

S=standard deviation of replicate analyses

A calculated LOD of 0.036 µg/L bromate was determined as shown in Table 1. The bromate LOD using 2-D IC is significantly lower than the bromate detection limits of 0.12 µg/L and 0.17 µg/L reported in EPA Methods 317.0 and 326.0, using postcolumn addition and absorbance detection.[11,12]

The performance of the 2-D bromate method was evaluated through a single-operator precision and accuracy study using fortified municipal and natural mineral water samples. The recovery of bromate was evaluated by analyzing eight different matrices, including reagent water, LSSM, two municipal drinking waters from different sources, and four natural mineral waters from different countries (France, Japan, Switzerland, United Kingdom). Each sample was fortified with 0.5 and 5 µg/L bromate. To ensure the accuracy of the calibration curve, quality control standards prepared at 0.5, 5, and 15 µg/L bromate were analyzed at the beginning, middle, and end of each sample analysis batch.

Table 2 summarizes the performance of the method for determining trace concentrations of bromate in municipal drinking waters using 2-D IC. As shown, trace concentrations of bromate were detected in both municipal drinking water samples, well below the current regulatory limit of 10 µg/L. The most likely source of bromate in these samples is the hypochlorite solution used for disinfection treatment, as ozonation is not used at either of the tested drinking water treatment facilities.

For the municipal drinking waters fortified with 0.5 µg/L bromate, the calculated recoveries were between 98 and 99%. For the same samples fortified with 5 µg/L bromate, the recovery was approximately 105%. Figure 4 shows chromatograms of drinking water B, unfortified and fortified with 0.5 µg/L bromate, using the combined IonPac AS19/AS24 columns with 2-D IC. As shown, bromate is well-resolved from any potential inference on the IonPac AS24 column, and therefore produces an excellent recovery of 98.7%.

Table 2. Bromate Recoveries from Fortified Reagent Water, LSSM, and Municipal Drinking Water Matrices

Matrix	Amount Found (µg/L)	Amount Added (µg/L)	Replicates	Average Recovery (%)	Peak Area Precision (RSD)
Reagent Water	—	0.5	7	101.5	1.98
		5.0	7	105.6	0.66
LSSM[a]	—	0.5	7	96.1	5.75
		5.0	7	106.7	1.66
Drinking Water A	0.45	0.5	7	98.2	6.06
		5.0	7	104.5	1.71
Drinking Water B	1.19	0.5	7	98.7	2.51
		5.0	7	105.6	1.91

[a] LSSM = Laboratory Synthetic Sample Matrix containing 100 mg/L each of chloride, sulfate, and bicarbonate and 10 mg/L each of nitrate-N and phosphate-P

Some drinking water samples may contain elevated concentrations of chloride, sulfate, and bicarbonate that can increase peak broadening of bromate and therefore lower recovery. To determine whether the 2-D IC method can analyze these types of samples, a LSSM was prepared and fortified with 0.5 and 5 µg/L bromate. The excellent recoveries shown in Table 2 for this sample and system configuration indicate that bromate was not influenced by the increased concentrations of common anions.

In general, the ionic strength of natural mineral waters significantly exceeds the concentrations found in typical municipal drinking water samples. Determining low concentrations of bromate in these matrices using currently available methods is a challenging analytical problem because of column overloading. In some cases, sample dilution is required, increasing the MRL in proportion to the dilution factor. Alternatively, samples can be treated with OnGuard® cartridges to remove most of the chloride, carbonate, and sulfate in the sample. However, this requires additional time and increases the cost of each analysis. One of the primary advantages of 2-D IC is that most samples can be injected directly without any sample pretreatment, thereby simplifying analysis.

In this study, four different natural mineral water samples were analyzed for bromate. Table 3 summarizes the ionic properties of three of the investigated samples according to the manufacturers' specifications. As shown, the ionic strength of the mineral waters analyzed in this study varied significantly. None of the bottled minerals waters indicated that ozonation was used as a disinfection treatment method; therefore the detection of bromate was not anticipated.

The samples were fortified with 0.5 and 5 µg/L bromate to evaluate the accuracy of the 2-D method for determining bromate in natural mineral waters (Table 4). As shown, recoveries were in the range of 95-105% with peak area precisions for seven replicate injections in the range of 0.6-7.2%. Previously, a 250 µL direct injection of mineral water C was analyzed with the IonPac AS19 column followed by suppressed conductivity detection.[16] Using the method parameters described in Application Note 184 required a 1:5 sample dilution to reduce the 1445 mg/L sulfate in the sample to avoid column overloading. This would result in an increase in the MRL by a factor of five. However, because the 2-D method removes most of the interfering matrix ions in the first dimension, the sample can be injected directly without any sample preparation steps, thereby maintaining the method's MRL of 0.5 µg/L bromate. However, the increased sulfate concentration did shift the retention time of bromate in the first dimension, resulting in a lower bromate recovery than expected. To improve the bromate recovery, the cut window was increased from 8-10 min to 7-10 min to account for the shift in retention time on the AS19 column. This minimal change in cut time resulted in a significant improvement in recovery from <50% to ~96%. Figure 5 shows example chromatograms of unfortified and fortified mineral water C. As shown in these chromatograms, a significant

Table 3. Concentrations (mg/L) of Cations and Anions of the Investigated Mineral Water Samples

Mineral Water	Na^+	K^+	Mg^{2+}	Ca^{2+}	F^-	Cl^-	NO_3^-	HCO_3^-	SO_4^{2-}
A	5.5	0.7	9.5	50.8	—[a]	5.9	<3	190	5.8
B	9.0	0.6	3.4	147.3	0.12	21.5	18	390	33
C	4.2	—[a]	117	510	1.8	3.0	<0.1	278	1445

[a] Not specified

Table 4. Bromate Recoveries from Fortified Natural Mineral Water Samples

Matrix	Amount Found (µg/L)	Amount Added (µg/L)	Replicates	Average Recovery (%)	Peak Area Precision (RSD)
Mineral Water A	<MDL[b]	0.5	7	95.2	3.37
		5.0	7	103.9	1.22
Mineral Water B	<MDL	0.5	7	95.2	5.85
		5.0	7	105.5	0.62
Mineral Water C[a]	<MDL	0.5	7	95.6	7.23
		5.0	7	103.8	1.22
Mineral Water D	<MDL	0.5	7	96.5	4.00
		5.0	7	103.8	1.35

[a] Cut time changed from 8–10 min to 7–10 min due to the increased amount of sulfate (1445 mg/L) that shifted retention times on the first dimension column

[b] <MDL = less than the method detection limit

amount of chloride was transferred to the AS24 column due to the shift in retention times on the AS19 column. The IonPac AS24 is a high-capacity anion-exchange column that provides an excellent bromate/chloride resolution as demonstrated in this example and thereby allowed good quantification of bromate from the sample.

CONCLUSION

This application note describes a 2-D IC system for the determination of ≥0.5 µg/L bromate in municipal and natural mineral waters. The method provides an improvement to existing EPA methods for bromate by providing lower detection limits and improved recoveries of bromate in high ionic strength matrices. In addition, samples can be injected directly without requiring the use of OnGuard cartridges, sample dilution, or sample degassing for carbonate removal prior to analysis. The elimination of time-consuming off-line sample preparation improves consistency between different analysts and laboratories. The method also allows the determination of trace concentrations of bromate in a wide range of sample matrices.

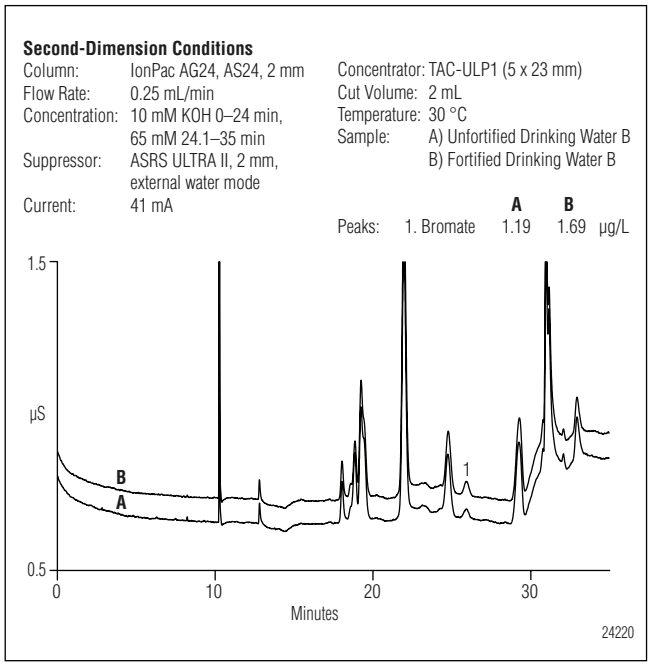

Figure 4. Chromatogram of (A) drinking water B and (B) drinking water B fortified with 0.5 µg/L bromate.

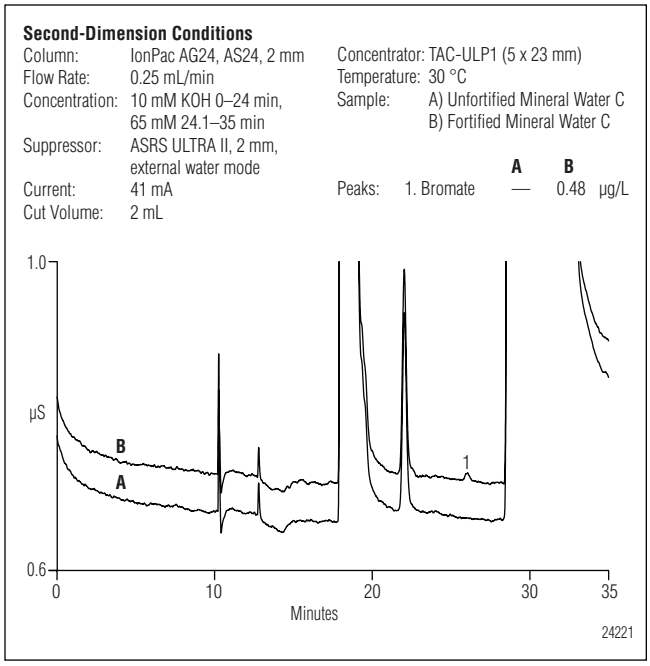

Figure 5. Chromatogram of (A) mineral water C and (B) mineral water C fortified with 0.5 µg/L bromate.

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Determination of Trace Concentrations of Chlorite, Bromate, and Chlorate in Bottled Natural Mineral Waters

INTRODUCTION

Bottled water has been one of the fastest growing beverage markets in the last five to ten years. Global consumption approached 41 billion gallons in 2004, an increase of 6.5% from 2003.[1] The bottled water industry markets to health conscious consumers as an alternative not only to tap water, but also to carbonated soft drinks and juice drinks.[1] Regardless of whether the water is delivered from a local municipality or is prepackaged in a bottle, the consumption of safe and reliable drinking water is essential to maintain a healthy lifestyle.

Bottled water must be disinfected to remove pathogenic microorganisms and ensure it is safe for human consumption. Water companies prefer ozone as a disinfectant because it is one of the most effective treatments available, it does not leave a taste, and there is no residual disinfectant in the bottled water.[2,3] Some bottlers, however, use ultraviolet light or chlorine dioxide as alternative treatment methods.[2] Reactions between disinfectants and natural organic and inorganic matter in the source water can result in the production of undesirable disinfection byproducts (DBPs), such as chlorite, bromate, and trihalomethanes, that are potentially harmful to humans.[4] Bromate, for example, can be formed by ozonation of water containing naturally occurring bromide, or may be present as an impurity in sodium hypochlorite used for treatment.[5]

Results from toxicological studies led the International Agency for Research on Cancer to conclude that bromate is a potential human carcinogen, even at low µg/L (ppb) concentrations.[6] The World Health Organization (WHO) estimated excess lifetime cancer risks of 10^{-4}, 10^{-5}, and 10^{-6} for drinking water containing bromate at 20, 2, and 0.2 µg/L, respectively.[9] The U.S. EPA,[7] European Commission,[8] and the WHO[9] set a maximum permissible limit of 10 µg/L bromate in tap water. The U.S. FDA[10] adopted the same regulatory limit for bottled water. In Europe, natural mineral waters and spring waters treated by ozonation have a maximum permissible limit of 3 µg/L bromate.[11]

Traditionally, ion chromatography (IC) with suppressed conductivity detection has been used for determination of bromate and other DBPs in drinking water, as described in EPA Method 300.1.[12] This method describes the use of a high-capacity IonPac AS9-HC column with a carbonate eluent and large loop injection to achieve a method detection limit (MDL) of 1.4 µg/L bromate. In early 2006, the U.S. EPA enacted stage 2 of the disinfectants/disinfection byproducts (D/DBP) rule, maintaining the maximum permissible limit for bromate but adding three additional analytical methods to further improve the selectivity and sensitivity for bromate.[13] U.S. EPA Methods 317.0 and 326.0 combine suppressed conductivity detection and absorbance detection after postcolumn addition to achieve bromate MDLs less than 0.2 µg/L.[14,15] IC coupled to inductively coupled plasma mass spectrometry has also been demonstrated for the determination of low concentrations of bromate in environmental waters, permitting a bromate MDL of 0.3 µg/L.[16]

A high-capacity IonPac® AS19 column with an electrolytically generated hydroxide eluent, large loop injection, and suppressed conductivity detection can achieve a calculated bromate MDL of 0.34 µg/L.[17] Absorbance detection after postcolumn addition can reduce this MDL to less than 0.2 µg/L, using EPA Methods 317.0 and 326.0.[18,19] In this application note, we compare the IonPac AS19 using an electrolytically generated hydroxide eluent to the IonPac AS23 column using an electrolytically generated carbonate/bicarbonate eluent for the determination of chlorite, bromate, and chlorate in natural mineral waters. We compare the linearity, method detection limits, precisions, and recovery for three mineral waters obtained from three European countries to determine whether these columns have the sensitivity required to meet current EPA and EU requirements.

EQUIPMENT

A Dionex ICS-2000 Reagent-Free™ Ion Chromatography (RFIC™) system was used in this work. The ICS-2000 is an integrated ion chromatograph and consists of:

- Eluent generator
- Pump with in-line vacuum degas
- Column heater
- Hydroxide system:
  - EluGen® EGC II KOH cartridge (Dionex P/N 058900)
- CR-ATC (Dionex P/N 060477)
- Carbonate system:
  - EluGen EGC II K_2CO_3 cartridge (Dionex P/N 058904)
- EPM Electrolytic pH Modifier to generate the carbonate/bicarbonate eluent (Dionex P/N 063175)
- EGC Carbonate Mixer (Dionex P/N 061686)
- Two 4-L plastic bottle assemblies (for external water mode of suppression)
- AS Autosampler
- Chromeleon® Chromatography Management Software

REAGENTS AND STANDARDS

Deionized water, type I reagent grade, 18 M -cm resistivity or better

Sodium chlorite ($NaClO_2$, Fluka 71388, 80% pure)

Sodium bromate ($NaBrO_3$, EM SX 03785-1)

Sodium chlorate ($NaClO_3$, Fluka 71370)

CONDITIONS

Columns:	(A) IonPac AS19 Analytical, 4 x 250 mm (Dionex P/N 062885) IonPac AG19 Guard, 4 x 50 mm (Dionex P/N 062887)
	(B) IonPac AS23 Analytical, 4 x 250 mm (Dionex P/N 064149) IonPac AG23 Guard, 4 x 50 mm (Dionex P/N 064147)
Eluent:	(A) 10 mM KOH from 0–10 min, 10–45 mM from 10–25 min, 45 mM from 25–30 min*
	(B) 4.5 mM K_2CO_3/0.8 mM $KHCO_3$
Eluent Source:	(A) EGC II KOH with CR-ATC
	(B) EGC II K_2CO_3 with EPM
Flow Rate:	1.0 mL/min
Temperature:	30 °C
Injection:	250 µL
Detection:	(A) Suppressed conductivity, ASRS® ULTRA II, 4 mm (Dionex P/N 061561) AutoSuppression® recycle mode 130 mA current
	(B) Suppressed conductivity, ASRS ULTRA II, 4 mm AutoSuppression external water mode 25 mA current
CRD:	(A) 4-mm format (P/N 062983)
Background Conductance:	(A) <1 µS
	(B) 18–20 µS
System Backpressure:	~2200 psi
Run Time:	30 min

*Method returns to 10 mM KOH for 3 min prior to injection.

PREPARATION OF SOLUTIONS AND REAGENTS

Eluent Solution for the AS23 Column

4.5 mM Carbonate/0.8 mM Bicarbonate

Generate the carbonate/bicarbonate eluent on-line by pumping high quality deionized water (18 M -cm resistivity or better) through the EluGen EGC II K_2CO_3 Cartridge and EPM. Chromeleon will track the amount of eluent used and calculate the remaining lifetime.

Alternatively, prepare the eluent solution by adding 10 mL of the AS23 Eluent Concentrate (Dionex P/N 064161) to a 1-L volumetric flask containing

approximately 700 mL of degassed deionized water. Bring to volume and mix thoroughly. The 0.45 M sodium carbonate/0.08 M sodium bicarbonate concentrate can also be prepared from the salts by combining 47.7 g sodium carbonate (MW=106 g/mole) and 6.72 g sodium bicarbonate (MW=84 g/mole) in a 1-L volumetric flask containing approximately 700 mL of degassed deionized water. Bring to volume and mix thoroughly.

Stock Standard Solutions

Prepare 1000 mg/L stock standard solutions of chlorite, bromate, and chlorate by dissolving 0.1676 g, 0.1180 g, and 0.1275 g, respectively, of the corresponding sodium salts in separate 100 mL volumetric flasks of DI water.

Calibration Standard Solutions

Prepare a secondary stock solution containing 1 mg/L each of chlorite and chlorate and a separate secondary stock solution containing 1 mg/L bromate by performing the appropriate dilutions of the 1000 mg/L stock standards. Calibration standards can then be prepared from the secondary solutions using the appropriate dilutions. Dilute working standards should be prepared monthly, except those that contain chlorite, which must be prepared every two weeks, or sooner if evidence of degradation is indicated by repeated QC failures. Concentration ranges used in this application note are shown in Table 1.

SAMPLE PREPARATION

For the present analysis, mineral waters B and C were degassed for 10–15 min under vacuum due to an excess amount of bicarbonate in the samples. Increased amounts of bicarbonate in the sample can produce shifts in retention times as shown in Figures 1A and 1B. In addition, due to the presence of significantly high concentrations of sulfate in mineral water C, the sample was diluted 1:5 with DI water prior to analysis.

Table 1. Calibration Data, Retention Time Precisions, Peak Area Precisions, and Method Detection Limits For DBP Anions

Analyte	Range (µg/L)	Linearity (r^2)	Retention Time[a] RSD (%)	Peak Area RSD (%)	MDL Standard (µg/L)	Calculated MDL (µg/L)
IonPac AS19 Column						
Chlorite	2-50	0.9999	0.04	1.20	1.0	0.18
Bromate	1-25	0.9995	0.03	1.40	2.0	0.31
Chlorate	2-50	0.9999	0.01	0.54	1.0	0.28
IonPac AS23 Column						
Chlorite	10-50	0.9999	0.07	2.20	5.0	1.02
Bromate	5-25	0.9998	0.07	2.63	5.0	1.63
Chlorate	10-50	0.9998	0.11	2.48	9.0	2.05

[a]RSD= relative standard deviation, n = 10 for a standard consisting of 10 ppb bromate and 20 ppb each of chlorite and chlorate.

RESULTS AND DISCUSSION

The IonPac AS23 is a high-capacity anion-exchange column specifically designed to be used with carbonate /bicarbonate eluent for the determination of the trace DBPs, chlorite, bromate, and chlorate, together with common inorganic anions, including bromide (precursor to bromate), in drinking waters. To simplify the method and avoid manual eluent preparation, this column can be used with electrolytically generated potassium carbonate that is modified by an Electrolytic pH Modifier (EPM) to automatically generate the carbonate/bicarbonate eluent that is required for analyte separation. The IonPac AS23 column was developed using a unique polymer technology to achieve a capacity of 320 µeq/column, higher than the IonPac AS9-HC column (190 µeq/column) described in EPA Method 300.1. The combination of an optimized selectivity for DBP anions, high anion exchange capacity, and improved selectivity of carbonate from inorganic anions and oxyhalides, makes this column an ideal replacement for the AS9-HC column.

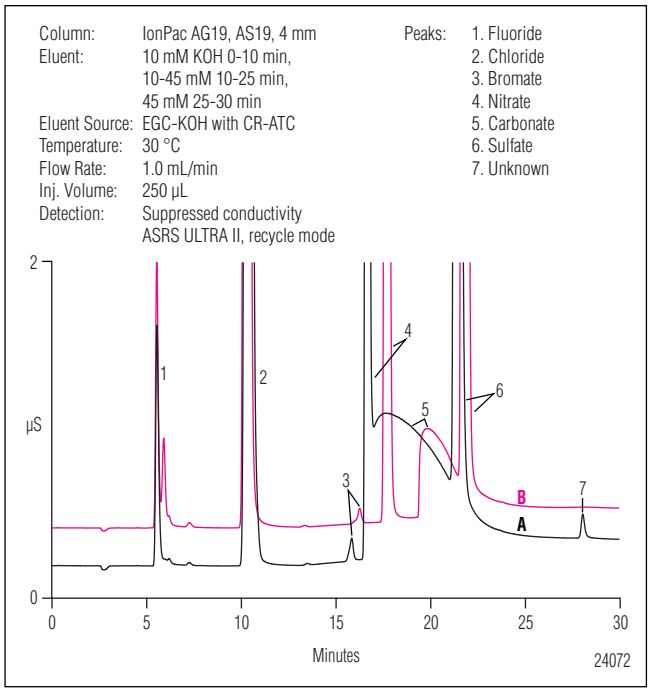

Figure 1. Comparison of mineral water B A) before vacuum degas and B) after vacuum degas.

In this application, we compare the IonPac AS23 column to the hydroxide-selective IonPac AS19 column for the determination of trace DBP anions in natural mineral waters. Figure 2 compares the separation for chlorite, bromate, and chlorate on the IonPac AS19 and AS23 columns. As shown, both columns provide good selectivity for the target DBP anions.

The linear calibration ranges, MDLs, and quality control standard (QCS) performances were evaluated for the hydroxide and carbonate eluent systems. The hydroxide eluent system was calibrated using four increasing concentrations of chlorite and chlorate (2-50 µg/L) and five increasing concentrations of bromate (1-25 µg/L). For the carbonate-based system, chlorite and chlorate were calibrated from 10-50 µg/L whereas bromate was calibrated from 5-25 µg/L using three different concentrations. Each system produced a linear response in its respective range with a correlation coefficient greater than 0.999. The improved sensitivity of the hydroxide eluent system, however, allowed a lower minimum reporting limit (MRL) than the carbonate-based system. The MDLs for the target DBPs were determined for each system by performing seven replicate injections of reagent water fortified with the calibration standards at concentrations of three to five times the estimated instrument detection limits.

Table 1 compares the calibration data, retention time and peak area precisions for a QCS, and MDLs for the IonPac AS19 with an electrolytically generated hydroxide eluent to the IonPac AS23 with an electrolytically generated carbonate/bicarbonate eluent. The calculated MDL of bromate with the IonPac AS19 column was 0.31 µg/L compared to 1.63 µg/L using the IonPac AS23 column. This demonstrates that hydroxide eluents improve the sensitivity for bromate compared to carbonate-based eluents and are therefore more suitable to meet the current European regulatory requirement of 3 µg/L bromate in natural mineral waters. Either the AS19 or AS23 based IC systems are capable of measuring the 10 µg/L requirement of bromate for tap water or U.S. bottled water according to the regulations established by the U.S. EPA, U.S. FDA, WHO, and European Commission.

In the U.S., mineral water is defined as water that contains no less than 250 ppm total dissolved solids (TDS) and that originates from a geologically and

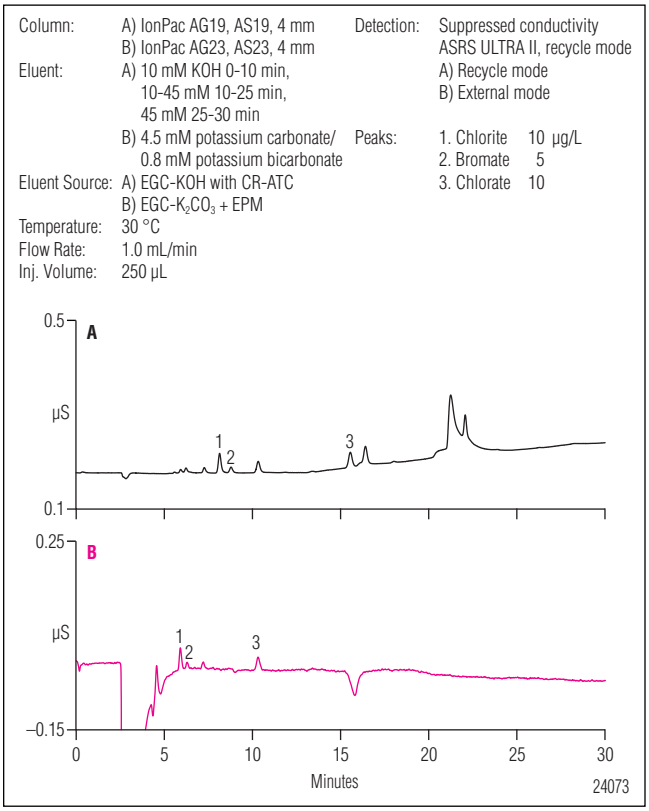

Figure 2. Separation of disinfection byproducts using the A) IonPac AS19 column and B) IonPac AS23 column.

physically protected underground water source. Mineral content must be maintained at a constant level and no minerals may be added to the water.[18] In Europe, mineral water is defined as microbiologically wholesome water, originating from an underground water table or deposit and emerging from a spring tapped at one or more natural or bored exits. It can contain less than 50 ppm TDS.[19] The total mineral content of the waters can vary significantly, with higher mineral concentrations generally appearing in Russia, the Baltic States, and Germany. The differences between regions are most likely a result of differences in the overall compositions of the waters and the geological locations.[20]

In this application, three natural mineral waters from different European countries with TDSs that varied significantly from 136 to 2359 ppm were evaluated. The properties of the investigated water samples are summarized in Table 2. As shown, the ionic strength of mineral water C is significantly higher than observed in typical drinking waters. The absence of bromate in the bottled mineral waters analyzed indicated that ozonation was not used for disinfection.

Tables 3 and 4 summarize typical recoveries for single-operator data obtained using the IonPac AS19 and AS23 columns, respectively, for trace concentrations of DBP anions in three European natural mineral water samples. As shown, chlorite and bromate were not detected in any of the samples analyzed,whereas only a trace concentration of chlorate was detected in mineral water A. To determine the accuracy of the method, the samples were spiked with 5 µg/L bromate and 10 µg/L each of chlorite and chlorate. Calculated recoveries for the spiked mineral water samples were in the range of 86–97% and 84–111% using the IonPac AS19 and AS23 columns, respectively. The analyte recoveries using either a hydroxide or carbonate/bicarbonate eluent were within the acceptable range of 75–125% according to the criteria described in EPA Method 300.1. Figure 3 compares chromatograms of mineral water A using the IonPac AS19 and AS23 columns. Figure 4 shows the same chromatograms spiked with 5 µg/L bromate and 10 µg/L each of chlorite and chlorate, which resulted in good recoveries for both eluents. Although bromide was not quantified in this study, the estimated concentrations were approximately 16 µg/L in mineral waters A and B and 2 µg/L in mineral water C. Therefore, ozonation of mineral waters A and B could potentially produce bromate. To demonstrate the applicability of detecting bromate at concentrations significantly less than the 3 µg/L European regulatory limit for ozonated mineral waters, mineral water A was spiked with 0.5 µg/L bromate (Figure 5). As shown, bromate can be observed easily at this concentration, with good peak-to-peak baseline noise of 0.3–0.5 nS.

Table 2. Concentrations in mg/L of Cations and Anions in the Investigated Mineral Water Samples

Mineral water	Na^+	K^+	Mg^{2+}	Ca^{2+}	F^-	Cl^-	NO_3^-	HCO_3^-	SO_4^{2-}
A	11.8	6.2	8	11.5	–[a]	13.5	6.3	71	8.1
B	4.5	0.5	8	32.0	–[a]	5.0	< 2	133	7.0
C	4.2	–[a]	117	510	1.8	3.0	< 0.1	278	1445

[a]Not specified.

Table 3. Recoveries of Disinfection Byproduct Anions in Natural Mineral Waters Using the IonPac AS19 Column

Mineral water	Analyte	Amount found (µg/L)	Amount added (µg/L)	Recovery (%)
A	Chlorite	<MDL	10	87.7
	Bromate	<MDL	5.0	96.0
	Chlorate	4.4	10	91.1
B	Chlorite	<MDL	10	86.4
	Bromate	<MDL	5.0	97.4
	Chlorate	<MDL	10	90.7
C	Chlorite	<MDL	10	87.6
	Bromate	<MDL	5.0	94.7
	Chlorate	<MDL	10	92.8

Table 4. Recoveries of Disinfection Byproduct Anions in Natural Mineral Waters Using the IonPac AS23 Column

Mineral water	Analyte	Amount found (µg/L)	Amount added (µg/L)	Recovery (%)
A	Chlorite	<MDL	10	107.6
	Bromate	<MDL	5.0	91.2
	Chlorate	4.6	10	99.3
B	Chlorite	<MDL	10	110.6
	Bromate	<MDL	5.0	93.5
	Chlorate	<MDL	10	92.9
C	Chlorite	<MDL	10	104.3
	Bromate	<MDL	5.0	83.9
	Chlorate	<MDL	10	102.6

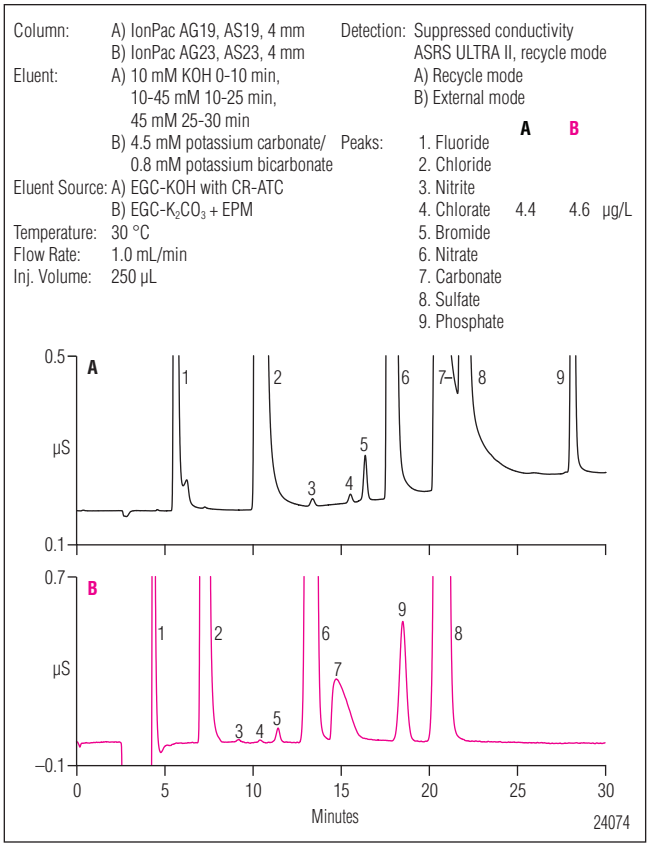

Figure 3. Comparison of the A) IonPac AS19 and B) IonPac AS23 columns for the separation of DPB anions in mineral water A.

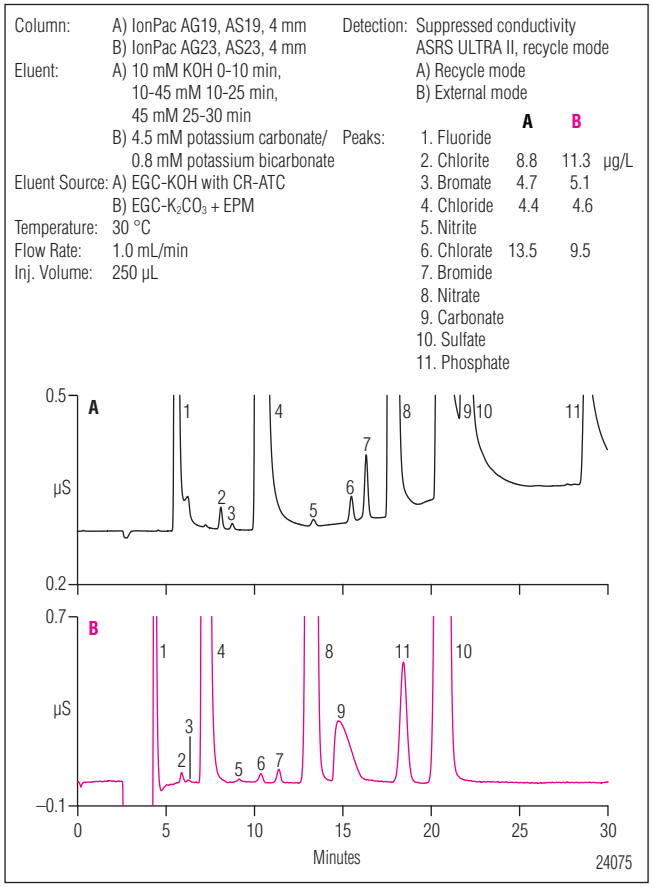

Figure 4. Comparison of the A) IonPac AS19 and B) IonPac AS23 columns for the separation of trace concentrations of common anions and DPB anions spiked in mineral water A.

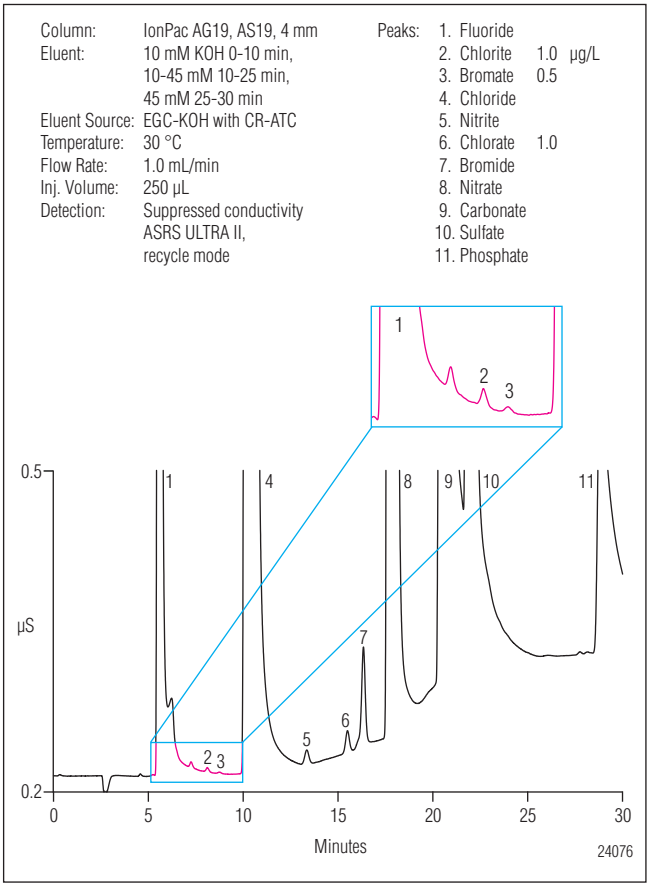

Figure 5. Chromatogram of mineral water A spiked with 1 µg/L each chlorite and chlorate and 0.5 µg/L bromate.

CONCLUSION

The IonPac AS19 column using an electrolytically generated hydroxide eluent was compared to the AS23 column using an electrolytically generated carbonate/bicarbonate eluent for the determination of trace concentrations of DBP anions in natural mineral waters. The improved sensitivity using a hydroxide eluent allowed the detection of lower concentrations of bromate, a potential human carcinogen, in drinking waters. Therefore, the IonPac AS19 with an electrolytically generated hydroxide eluent is recommended for laboratories that must comply with EU Directive 2003/40/EC, which permits a maximum of 3 µg/L bromate in mineral waters treated with ozone. The use of either the IonPac AS19 column with a hydroxide eluent or IonPac AS23 column with a carbonate/bicarbonate eluent provides the required sensitivity to meet the maximum permissible limit of 10 µg/L bromate currently required by most regulatory agencies. Both columns demonstrated good resolution between bromate and chloride and comparable recovery for mineral water samples spiked with known concentrations of chlorite, bromate, and chlorate. In addition, hydroxide or carbonate/bicarbonate eluents can be generated on-line from deionized water, freeing the operator from manually preparing eluents. This increases the automation, ease-of-use, andreproducibility between analysts and laboratories.

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Determination of Disinfection Byproduct Anions and Bromide in Drinking Water Using a Reagent-Free Ion Chromatography System Followed by Postcolumn Addition of an Acidified On-Line Generated Reagent for Trace Bromate Analysis

INTRODUCTION

Public drinking water municipalities routinely disinfect their water supplies to protect the public from potentially dangerous microorganisms. Chlorine dioxide, chloramine, and ozone are common disinfection treatments used to treat public water supplies.[1] These treatments produce byproducts that expose the public to potentially harmful chemicals. For example, the use of chlorine dioxide for disinfection treatment can generate the oxyhalide disinfection byproducts (DBPs) chlorite and chlorate, whereas the use of chloramine can produce chlorate.[2] Although ozonation of water supplies is a particularly effective disinfection treatment, bromate may be generated if the source water contains elevated levels of naturally occurring bromide. Bromate has been identified as an animal carcinogen and a potential human carcinogen by the International Agency for Research on Cancer.[3]

The USEPA has estimated a potential cancer risk of 1 in 104 for a lifetime exposure to drinking water containing 5 µg/L bromate and a potential risk of 1 in 105 for 0.5 µg/L bromate.[4] The USEPA promulgated the Stage 1 Disinfectants/Disinfection Byproducts (D/DBP) Rule in 1998 that established a maximum contaminant level (MCL) for bromate at 10 µg/L and an MCL for chlorite of 1000 µg/L.[5] At the same time, the USEPA set a maximum contaminant goal of zero for bromate. In an EU (European Union) directive, the EU also proposed a regulatory value of 10 µg/L bromate that must be met within 10 years after entry into the EU.[6] The World Health Organization has reduced their bromate guideline from 25 µg/L to a provisional value of 10 µg/L.[7]

Considerable efforts have focused on developing improved analytical methods for determining trace concentrations of inorganic DBPs in drinking water to meet current regulatory requirements. Traditionally, ion chromatography (IC) with suppressed conductivity detection has been used to determine chlorite, bromate, and chlorate in environmental waters, as described in Method 300.0 (B).[8] This method describes the use of an IonPac® AS9-SC column with a reported method detection limit (MDL) of 20 µg/L bromate. Method 300.1 (B) was published in the Stage 1 D/DBP Rule as an update to Method 300.0 to further reduce the bromate MDL from 20 to 1.4 µg/L.[9] Method 300.1 describes the use of an IonPac AS9-HC column with a carbonate eluent and a large volume injection followed by suppressed conductivity detection. The bromate detection limit can be reduced to <1 µg/L by using preconcentration after sample pretreatment.[10,11]

Postcolumn derivatization methods can also be used to quantify bromate at sub-µg/L concentrations. The Stage 2 D/DBP Rule published two methods that combine Method 300.1 (B) with a postcolumn reagent (PCR) to further improve the sensitivity of bromate determinations in environmental waters.[12] EPA Method 317.0 combines suppressed conductivity and the postcolumn addition of *o*-dianisidine (ODA) followed by visible detection to achieve a bromate MDL of 0.1 µg/L with a practical quantitation limit (PQL) of 0.5 µg/L.[3,13] However, the ODA PCR is a potential human carcinogen.[14] Therefore, EPA Method 326.0 was developed as an alternative to Method 317.0. Method 326.0 uses a postcolumn reaction that generates hydroiodic acid (HI) in situ, from an excess of potassium iodide (KI), that combines with bromate from the column effluent to form the triiodide anion (I_3^-) that is detected by absorbance at 352 nm.[15]

Most published EPA methods specify the use of an IonPac AS9-HC column and a 9 mM sodium carbonate eluent for determining DBP anions in drinking water. A hydroxide-selective column had not been used for this application due to the lack of a suitably selective column for the target DBP anions, chlorite, bromate, and chlorate. The introduction of the IonPac AS19, a hydroxide-selective column, not only improved the selectivity for disinfection byproducts, but also provided the typical advantages observed when using hydroxide eluent for trace applications, such as lower baseline noise and improved sensitivity. For example, the use of the AS19 column combined with electrolytically-generated potassium hydroxide eluent resulted in a bromate MDL approximately three times lower than with the IonPac AS9-HC column and carbonate eluent.[16,17] The AS19 can also be substituted for the AS9-HC in EPA method 317.0.18. In this application note, we demonstrate the performance of the AS19 column for EPA Method 326.0. This method allows quantification of bromate to 1 µg/L by suppressed conductivity detection with a hydroxide eluent and 0.5 µg/L using postcolumn reaction with UV detection. The linearity, method detection limits, and quantification of the target DBP anions and bromide in municipal and bottled drinking waters are discussed.

EQUIPMENT

A Dionex ICS-3000 Reagent-Free Ion Chromatography System (RFIC) consisting of:

- DP Dual Pump or SP Single Pump
- DC Dual Compartment with a CD conductivity detector and an Automation manager (PN 061962) equipped with a RCH-1 Postcolumn Reaction Heater (Dionex P/N 061747)
- VWD UV/Vis Absorbance Detector with a PEEK® analytical flow cell (PN 6074.0200)
- AS Autosampler
- EG Eluent Generator with an EluGen® EGC II KOH Cartridge (Dionex P/N 058900)
- Continuously-Regenerated Anion Trap Column, CR-ATC (Dionex P/N 060477)
- PC10 Postcolumn Pneumatic Delivery Module (Dionex P/N 050601)
- Knitted Reaction Coil, 500 µL, potted (for RCH-1) (Dionex P/N 039349)
- PEEK Mixing Tee (Dionex P/N 048227)
- Four 4-L plastic bottle assemblies (Dionex P/N 063292)
- Three bottles for external water mode of suppression
- One bottle for 0.3 N sulfuric acid for the online conversion of KI to I_3^-.

Chromeleon® Chromatography Management Software

Polystyrene Autoselect™ vials with caps and septa, 10 mL (Dionex P/N 055058)

Nalgene Filter Unit, 0.2 µm nylon membrane, 1000 mL (VWR P/N 28198-514)

REAGENTS AND STANDARDS

Deionized water, Type I reagent grade, 18 MW-cm resistivity or better

Potassium Iodide (KI) (VWR P/N BDH0264-500g)

Ammonium Molybdate Tetrahydrate $[(NH_4)_6Mo_7O_{24}•4H_2O]$ (Sigma-Aldrich, A7302)

Ethylenediamine (EDA) (Aldrich, 24,072-9)

Sulfuric Acid, 36 N (J.T. Baker INSTRA-ANALYZED 9673-33)

Bromide Standard, 1000 mg/L, 100 mL (Ultra Scientific, VWR P/N ICC-001)

Sodium Chlorite ($NaClO_2$) (Fluka 71388, 80% pure)

Bromate Standard, 1000 mg/L, 100 mL (Ultra Scientific, VWR P/N ICC-010)

Sodium Bromate ($NaBrO_3$) (EM SX 03785-1)

Sodium Chlorate ($NaClO_3$) (Aldrich, 24,414-7)

DL-Malic Acid, Disodium salt (Sigma-Aldrich, M6773)

CONDITIONS

Columns:	IonPac AS19 Analytical, 4 × 250 mm (Dionex P/N 062885) IonPac AG19 Guard, 4 × 50 mm (Dionex P/N 062887)
Eluent:	10 mM KOH from 0–10 min, 10–45 mM from 10–25 min, 45 mM from 25-30 min*
Eluent Source:	EGC II KOH with CR-ATC
Flow Rate:	1.0 mL/min
Temperature:	30 °C
Inj. Volume:	250 µL
Detection:	Suppressed conductivity, ASRS® 300, 4 mm (Dionex P/N 064554) AutoSuppression® external water mode 112 mA current
Background Conductance:	<1 µS
Noise:	~1 nS
System Backpressure:	~2400 psi
Run Time:	30 min

Postcolumn Reaction Conditions

UV Detection:	Absorbance at 352 nm (deuterium lamp)
PCR Flow:	0.26 M potassium iodide at 0.3 mL/min
AMMS III:	0.3 N sulfuric acid at 2.5 mL/min
Postcolumn Heater Temp:	80 °C
UV Noise:	<0.1 mAU

*Method returns to 10 mM KOH for 5 min prior to injection.

PREPARATION OF SOLUTIONS AND REAGENTS

Deionized Water Preparation

Deionized water should be degassed prior to use in the RFIC system. The presence of oxygen in the system will affect the baseline in the postcolumn system and it must be removed. Water can be degassed by filtering it through a 1 L, 0.2 µm nylon filter unit (Nalgene) and then sonicating the solution while it is under vacuum for 15 min. For larger volumes of water, a vacuum-safe glass container may be used by applying vacuum to the container while sonicating for 15 min.

Ethylenediamine (EDA) Preservation Solution

Dilute 2.8 mL of 99% EDA to 25 mL with DI water according to Section 7.1.3 in EPA Method 326.0 to prepare a 100 mg/mL solution. Use 50 µL of 100 mg/mL EDA per 100 mL of standard or sample so the final EDA concentration is 50 mg/L. Store this solution at <6 °C and prepare fresh monthly.

Sulfuric Acid Solution (0.3 N)

Add 33.3 mL of concentrated sulfuric acid to ~ 1000 mL of DI water in a 2 L glass volumetric flask. Dilute the solution to 2 L with DI water. Transfer this solution (0.6 N sulfuric acid) to a 4 L plastic eluent bottle assembly. Fill the volumetric flask with an additional 2 L of DI water and add this water to the 4 L plastic eluent bottle to form a 0.3 N sulfuric acid solution.

Ammonium Molybdate Solution (2.0 mM)

Add 0.247 g of ammonium molybdate $[(NH_4)_6Mo_7O_{24}•4H_2O]$ to about 50 mL DI water in a 100-mL volumetric flask according to Section 7.1.4 in EPA Method 326.0. Dissolve and bring to volume with DI water. This solution is stored in an opaque plastic bottle at <6 °C and prepared fresh monthly.

Postcolumn Reagent (PCR) (0.26 M KI with 43 µM Ammonium Molybdate Tetrahydrate)

The PCR is prepared by adding 43.1 g of potassium iodide (KI) to a 1 L volumetric flask containing approximately 500 mL DI water and mixing to completely dissolve the solid. Dilute to volume with DI water and mix. Filter and degas this solution by vacuum filtration through a 0.2 µm nylon filter unit and add 215 µL of 2.0 mM ammonium molybdate solution. Immediately place the solution in the PC-10 reagent delivery vessel and pressurize with helium. Protect the PC-10 from light by covering with aluminum foil. If properly protected from light, this reagent is stable for 24 h.

Stock Standard Solutions

Prepare 1000 mg/L stock standard solutions by dissolving the corresponding mass of the salt in 100 mL DI water (Table 1). Alternatively, commercially available 1000 mg/L standards may be used. Stock standards for most anions listed in Table 1 are stable for at least six months when stored at <6 °C. Chlorite is only stable for two weeks when stored at <6 °C and protected from light.

Prepare a secondary stock standard containing 5 mg/L each of chlorite, chlorate, and bromide by combining 0.5 mL of each anion in a 100 mL volumetric flask and diluting to volume with DI water. Prepare a separate secondary stock standard containing 1 mg/L of bromate only by adding 0.1 mL of the 1000 mg/L bromate stock to a 100 mL volumetric flask and dilute to volume with DI water.

Table 1. Mass of Compounds Used to Prepare Stock Standard Solutions

Analyte	Compound	Amount (g)
Chlorite	Sodium chlorite ($NaClO_2$), 80%	0.1676
Bromate	Sodium bromate ($NaBrO_3$)	0.1180
Chlorate	Sodium chlorate ($NaClO_3$)	0.1275
Bromide	Sodium bromide (NaBr)	0.1288

Working Standard Solutions

Prepare dilute working standards by performing appropriate dilutions of the secondary stock solutions with deionized water containing EDA at a final concentration of 50 mg/mL. Dilute working standards should be prepared monthly, except those that contain chlorite which must be prepared every two weeks, or earlier if evidence of degradation is indicated by repeated QC failures as discussed in Method 326. Store all working standard solutions at <6 °C.

Surrogate (Sodium Malate) Stock Solution

Prepare a 1000 mg/L solution of malate by dissolving 135 mg of sodium malate in 100 mL of DI water. Add 100 µL of this solution to 100 mL of sample to spike the sample with 1 mg/L of surrogate.

SAMPLE PREPARATION

Filter samples, as necessary, through a 0.45 µm syringe filter, discarding the first 300 µL of the effluent. To prevent degradation of chlorite or the formation of bromate from hypobromous acid/hypobromite, preserve the samples by adding 50 µL of EDA preservation solution per 100 mL of sample. If a sample contains an excess amount of chlorite then the chlorite removal procedure described in Section 11.1.4.1 in Method 326.0 must be followed and the sample must then be reanalyzed for bromate. The holding time for preserved samples stored at <6 °C is 28 days for bromate, chlorate, and bromide and 14 days for chlorite.

Use of dichloroacetate (DCA) or trichloroacetate (TCA) as a surrogate is not recommended. Instead, add 100 µL of a 1000 mg/L malate solution to 100 mL of sample to obtain 1 mg/L of the malate surrogate.

SYSTEM PREPARATION AND SETUP

Prepare the ASRS 300 (Dionex P/N 064554) for use by hydrating the suppressor. Use a disposable plastic syringe and push approximately 3 mL of degassed DI water through the Eluent Out port and 5 mL of degassed DI water through the Regen In port. Allow the suppressor to stand for approximately 20 min to fully hydrate the suppressor screens and membranes. Install the ASRS 300 for use in the external water mode by connecting the Regen Out of the suppressor to the Regen In of the CR-ATC. The Regen In of the suppressor should connect directly to the external water source. The Regen Out of the CR-ATC is then connected to the SRS Waste In of the EG degasser. This configuration allows the eluent out of the analytical column to be connected to the conductivity detector after the suppressor and then to the mixing tee of the PCR system. Adjust the head pressure on the external water to achieve a total flow of 4-6 mL/min. Depending

on the backpressure of the installed components, the pressure on the external water should fall between 7–10 psi. Lower noise will be achieved if the total external water flow rate is as close to 6 mL/min as possible.

Prepare the AMMS 300 (P/N 064558) for use by hydrating the suppressor. Use a disposable plastic syringe and push approximately 3 mL of 0.3 N sulfuric acid through the Eluent Out port and 5 mL of 0.3 N sulfuric acid through the Regen In port. Allow the suppressor to stand for approximately 20 min to fully hydrate the suppressor screens and membranes. Install the suppressor in the chemical regeneration mode. Adjust the pressure on the 0.3 N sulfuric acid reservoir to deliver a flow rate of 2–3 mL/min. The pressure needed will be ~10–15 psi if an approximately 45 cm piece of 0.010" ID PEEK tubing is connected to the end of the tubing attached to the AMMS 300 Regen Out port.

Install the EGC II KOH cartridge in the EG and configure it with the CR-ATC according to the CR-TC Quickstart (LPN 031911). Use the Chromeleon system configuration to set up the EGC II KOH cartridge with the software. Condition the cartridge as directed by the EGC II Quickstart (LPN 031909) with 50 mM KOH at 1 mL/min for 30 min. Install a 4 × 50 mm AG19 and 4 × 250 mm AS19 column. Make sure the pressure displayed by the pump is at an optimal pressure of ~2300 psi when 45 mM KOH is delivered at 1 mL/min. This allows the EG degas assembly to effectively remove hydrolysis gases from the eluent. If necessary, install additional backpressure tubing to adjust the pressure to 2300 ± 200 psi.

Configure the ICS-3000 with the PCR system as shown in Figure 1. Orange PEEK tubing (Dionex P/N 042855, 0.020" ID) should be used between the PC-10 and the AMMS and between the AMMS and the mixing tee. The orange PEEK line from the AMMS should join the mixing tee directly opposite the black PEEK line from the conductivity detector. Black PEEK tubing (Dionex P/N 042690, 0.010" ID) should be used from the postcolumn reactor to the UV flow cell. The waste line from the UV flow cell should be made of a length of green PEEK tubing (Dionex P/N 044777, 0.030" ID) that is directed to a waste container. If noise above 0.1 mAU is consistently observed after system equilibration, a short piece of black PEEK tubing can be inserted between the cell and the waste line tubing to reduce trapped bubbles and therefore noise in the cell.

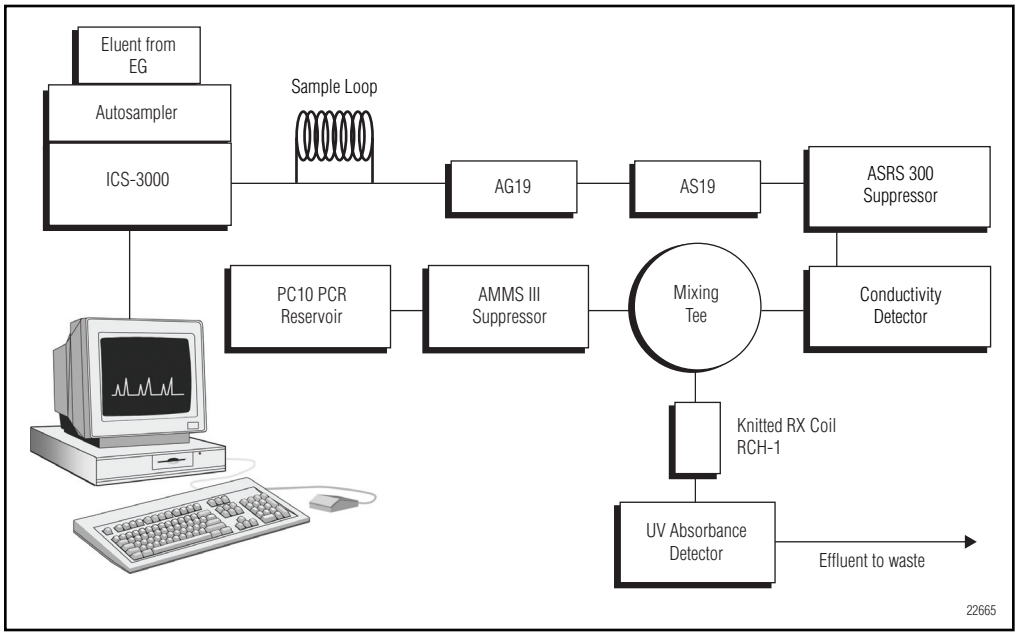

Figure 1. IC system configuration for EPA Method 326.0.

The PCR flow rate for this application was determined based on the analytical to PCR flow rate ratio provided in EPA Method 326.0. For our system, this resulted in the use of a 0.3 mL/min PCR flow rate. Set the temperature on the RCH-1 to 80 °C and the wavelength on the VWD to 352 nm. Allow both the suppressed conductivity and visible detection baselines to stabilize. Measure the PCR flow rate by collecting the combined effluent from the IC pump and PCR system in a pre-weighed vial for at least 5 min. After 5 min, weigh the collected solution. The mass of the solution is divided by the collection time (e.g., 5 min) to determine the total flow rate of the system. The PCR flow rate is the difference between the measured total flow rate and the flow rate delivered by the IC pump. Adjust the pressure of the postcolumn delivery module (PC10) and measure the flow rate again until the correct flow rate of 0.3 mL/min is achieved. Acceptable values for this flow rate range between 0.28 mL/min to 0.33 mL/min. The delivery pressure can range between 26-50 psi. If the pressure needed to reach 0.3 ml/min of PCR is greater that 50 psi, check the system for leaks or possible restrictions in flow such as crimped or clogged tubing. It is critical to confirm the flow rate daily, in addition to whenever the PCR is changed or if the quality control standard deviates from the EPA's acceptance criteria. Prior to analyzing any samples, inject 250 µL of DI water using the described method. This is the method blank. No peaks should elute at the same retention times as the target analytes. An equilibrated system has a suppressed background conductance <1 µS and peak-to-peak noise of ~1–2 nS per min and a UV peak-to-peak noise of <0.1 mAU per min. For the initial setup of the system it may take overnight for the conductivity detection to equilibrate and 2 days for the postcolumn UV detection to achieve a noise level of <0.1 mAU. For systems that are being restarted after a short shutdown, such as overnight or for a weekend, this equilibration will take 2–3 hours.

SYSTEM SHUTDOWN

If the system needs to be shut down, such as for a weekend, the following steps should be taken to protect the system components and ensure a smooth startup when the system is needed.

1: While the PCR and IC eluent are flowing, wearing latex or other suitable protective gloves and safety glasses, remove the line at the mixing tee that leads back to the AMMS Eluent Out port and plug this port on the tee. Turn off the pressure at the PC10. Allow eluent flow (10 mM KOH at 1 mL/min) for 30 min through the system with the Reaction Coil Heater (RCH) at 80 °C.

2: While the system is flushing with eluent, remove the PCR from the PC10 and wash the reservoir with DI water. Fill the reservoir with DI water and install in the PC10. Remove the line from the Eluent In port of the AMMS and flush this line with DI water from the PC10. Re-install this line to the Eluent In port of the AMMS and flush the AMMS with DI water.

3: At the mixing tee, disconnect the tubing leading from the Cell Out of the conductivity detector to the mixing tee. Connect this line to a separate waste line of green PEEK tubing. The conductivity detection portion of the system is now separate from the postcolumn detection portion of the system and can be shut down. The pressure for the external water can be shut off and the water reservoirs vented to stop flow. If the conductivity system will be shut down for several days, remove the Eluent In line from the ASRS and plug the port on the ASRS to protect the suppressor.

   Turn on the PC10 using the same pressure that was used for delivering the PCR. Flow through the AMMS should be approximately 1–1.5 mL/min. Connect the line from the Eluent Out port of the AMMS to the mixing tee. Flow acidified DI water from the AMMS through the RCH at 80 °C. Allow this to flow for 30 min to remove the residual hydroxide eluent from the UV cell.

4: Turn off the UV lamp and turn off the RCH. Allow the RCH to cool to <50 °C and then turn off the PC10 pressure. Turn off pressure to the external H_2SO_4 and vent the H_2SO_4 reservoir.

PRECAUTIONS

- When the application is set up for the first time, and whenever an AMMS 300 is replaced, it will require 2–3 days of flow for the PCR system to equilibrate and the noise on the VWD to fall below 0.1 mAU. The detection of 0.5 µg/L of bromate should not be attempted until after the PCR system has equilibrated. After this initial equilibration, the system should equilibrate and be ready to run samples in 2–3 h after a shutdown.
- The noise and drift present in the system will be highly dependent on maintaining consistent flow rates of the PCR, the external water, and the 0.3 N sulfuric acid. A high quality dual-stage regulator is highly recommended between the source gas (Helium or Nitrogen of grade 4.6 or better) and the regulators for the individual pressurized bottles. Use of a house compressed gas system is not recommended.
- Movement of the waste line leading from the UV flow cell will impact the noise observed in the UV. Be sure to secure this line so that it is not located where it can be disturbed.
- Daily checks of the pressures and flows for all pneumatically fed solutions are recommended. If the pressure of the PC10 needed to deliver 0.3 mL/min of PCR continually increases, the UV flow cell should be backflushed with eluent to remove any potential particulates that may have passed through the filtering process. To backflush the cell, remove the PCR line from the mixing tee and plug the tee at that port. Reverse the flow into the UV cell. Allow the eluent to flow for 1-2 min. Reconnect the line to the cell inlet and reconnect the waste line to the cell outlet.
- The presence of oxygen in the eluent will increase the background observed in the VWD detector. It is strongly recommended that the DI water be thoroughly degassed by vacuum and sonication prior to use.
- Filtration of the potassium iodide through a Nalgene 0.2 µm nylon filter unit is strongly recommended to remove insoluble material. The first time a filter unit is used, the membrane will turn yellow. This discoloration in the membrane will not affect the PCR that has been filtered and the PCR can be used for bromate analysis. If desired, the filter unit can be reused if promptly rinsed with DI water and an additional 1000 mL of DI water is filtered to clean the nylon membrane. Successive filtration with this filter unit will not further discolor the membrane.

RESULTS AND DISCUSSION

USEPA Method 326.0 specifies the use of an IonPac AS9-HC column with a 9 mM sodium carbonate eluent for the determination of chlorite, chlorate, and bromide by suppressed conductivity detection and bromate by suppressed conductivity and UV absorbance detection after postcolumn reaction with acidified potassium iodide.[15] Method 326.0 reports a bromate detection limit of 1.2 µg/L for a 225 µL injection by suppressed conductivity and 0.17 µg/L by UV absorbance (225 µL injection). Previously, we demonstrated that the bromate detection limit by suppressed conductivity can be reduced further to 0.34 µg/L using an electrolytically generated hydroxide eluent and a novel hydroxide-selective IonPac AS19 column.[16] Furthermore, we demonstrated that suppressed conductivity detection and postcolumn reaction with o-dianisidine may be used with an electrolytically generated hydroxide eluent and the AS19 column to achieve a bromate detection limit by visible detection equivalent to that reported in Method 317.0.13 In this application note, we examine the feasibility of using the IonPac AS19 column with the combination of suppressed conductivity detection and a postcolumn reaction system for UV absorbance detection. The use of a suitable hydroxide-selective column for this application allows for lower detection limits for the target disinfection byproduct anions by suppressed conductivity detection while still providing the improved sensitivity and selectivity for bromate obtained by the postcolumn reaction system.

Figure 2 shows chromatograms of 1 µg/L bromate and 10 µg/L each of chlorite, chlorate, and bromide. The top chromatogram shows the response obtained using suppressed conductivity detection and the bottom chromatogram was obtained using UV detection after postcolumn reaction with acidified KI. Bromate is well-resolved from chlorite. Although bromate is easily detected at this concentration using suppressed conductivity detection, an enhanced response for bromate is observed after postcolumn reaction with acidified KI followed by UV detection.

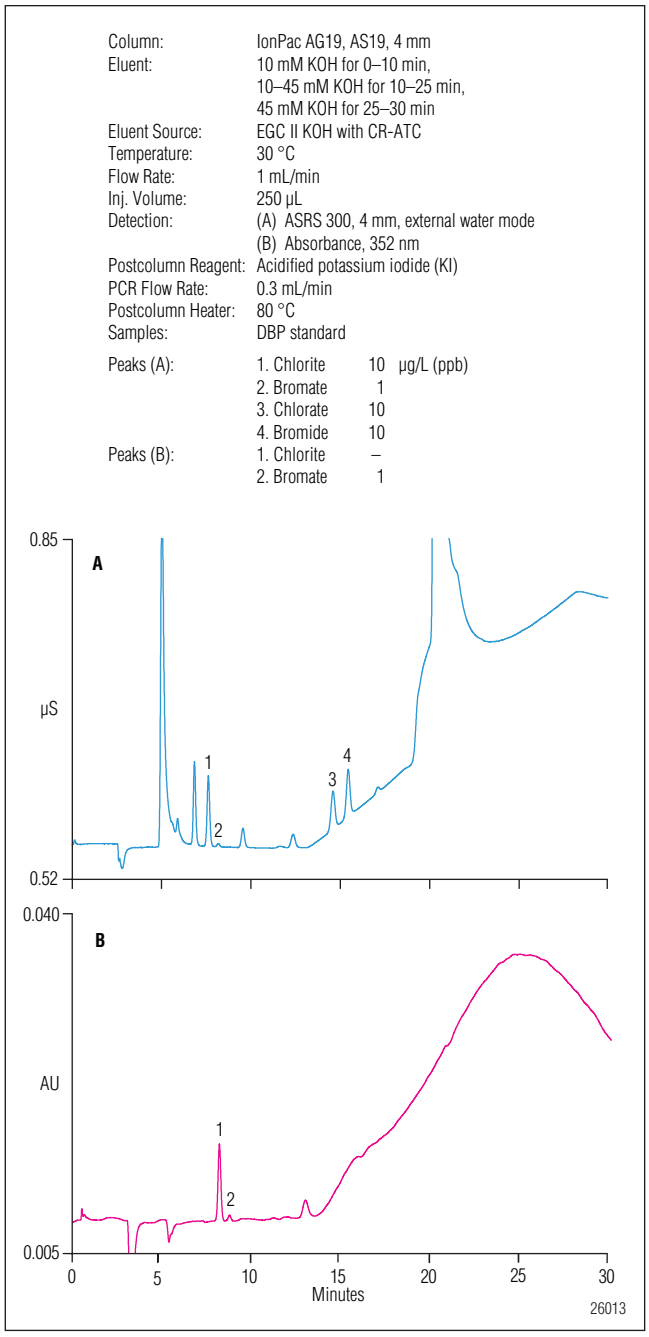

Figure 2. Separation of low ppb DBP anions and bromide on the IonPac AS19 column using suppressed conductivity detection and UV absorbance after PCR with acidified potassium iodide.

Table 2 summarizes the calibration data and method detection limits (MDLs) obtained for the DBP anions and bromide using the AS19 column and an electrolytically generated hydroxide eluent with suppressed conductivity and UV detections. The MDLs for the target analytes were determined by performing seven replicate injections of reagent water fortified at a concentration of three to five times the estimated instrument detection limit.[15] The calculated MDLs for bromate using suppressed conductivity detection followed by postcolumn reaction and UV detection were 0.12 µg/L and 0.18 µg/L, respectively. During the determination of the MDLs, the noise observed in the suppressed conductivity detection channel was unusually low, at 0.3-0.5 nS. Additionally, these were determined by injection of standards with minimal interference from other components that may be present in samples. The determined detection limit of 0.12 µg/L should not be expected in environmental samples. This method allows quantification of bromate to 1 µg/L using suppressed conductivity and 0.5 µg/L with UV detection using an AS19 column with an electrolytically generated potassium hydroxide eluent. Therefore, bromate was calibrated from 1–50 µg/L with suppressed conductivity and 0.5–15 µg/L using UV detection. Chlorite, chlorate, and bromide were each calibrated from 5–500 µg/L. These calibration ranges are expected to cover the typical concentrations found in environmental samples.

Table 2. Linearity and MDLs for DBP Anions and Bromide

Analyte	Range (µg/L)	Linearity (r^2)	MDL Standard (µg/L)	Calculated MDL (µg/L)
Chlorite	5–500	0.9993	0.6	0.33
Bromate (conductivity)	1–50	0.9997	0.5	0.12
Bromate (UV)	0.5–15	0.9999	0.5	0.17
Chlorate	5–500	0.9991	1.0	0.40
Bromide	5–500	0.9991	1.9	0.29

EPA Method 326.0 requires an initial demonstration of capability to characterize the instrument and laboratory performance of the method prior to performing sample analyses, as described in Section 9.2.15 An initial demonstration of precision, accuracy, and analysis of a quality control sample (QCS) are part of the criteria used for this characterization. For evaluating the precision and accuracy of the conductivity detector, Method 326.0 recommends using 20 µg/L each of the four target anions. However, because an electrolytically generated hydroxide eluent improves the overall sensitivity of the method, we determined that 5 µg/L bromate and 10 µg/L each of chlorite, chlorate, and bromide standards were suitable for characterizing the instrument and laboratory performance. For the absorbance detector, 2 µg/L bromate was used. EPA Method 326.0 considers an RSD ≤20% and an average recovery of ±15% to be acceptable performance. The precision of our replicate analyses was <5.8% RSD and the accuracy was 92–102%, well within EPA's acceptance criteria. A QCS should be analyzed after the calibration curves are initially established, on a quarterly basis, or as required to meet data quality needs. All QCS analyses in our experiments met the EPA's ±15% recovery criteria.

Table 3 summarizes the method's performance for the determination of trace DBP anions and bromide in municipal and bottled drinking water samples. For samples fortified with low concentrations of the target analytes, recoveries ranged from 90–112%, well within the 75–125% acceptance criteria of EPA Method 326.0. Figures 3–6 illustrate the performance for the determination of DBP anions and bromide in municipal tap

Table 3. Recoveries of Trace DBP Anions Spiked into Water Samples

Analyte	Amount Found (µg/L)	Amount Added (µg/L)	Recovery (%)
Tap Water A			
Chlorite	4.6	6.9	95.9
Bromate (conductivity)	0.32	1.0	95.5
Bromate (UV/Vis)	0.35	1.0	98.1
Chlorate	74.7	80.1	97.5
Bromide	34.6	39.9	95.4
Tap Water B			
Chlorite	< MDL	4.6	108.0
Bromate (conductivity)	2.4	3.0	102.8
Bromate (UV/Vis)	2.8	3.0	94.7
Chlorate	62.4	69.7	96.7
Bromide	17.5	19.9	92.3
Bottled Water A-1			
Chlorite	< MDL	4.9	105.3
Bromate (conductivity)	9.5	9.7	101.1
Bromate (UV/Vis)	10.8	9.7	97.3
Chlorate	< MDL	6.2	99.8
Bromide	19.0	19.9	95.0
Bottled Water A-2			
Chlorite	<MDL	6.4	95.9
Bromate (conductivity)	8.7	9.7	95.7
Bromate (UV/Vis)	8.5	9.7	98.4
Chlorate	< MDL	6.4	107.6
Bromide	3.2	6.4	111.8
Bottled Water B			
Chlorite	< MDL	4.9	108.3
Bromate (conductivity)	< MDL	1.0	102.4
Bromate (UV/Vis)	< MDL	1.0	104.5
Chlorate	< MDL	5.2	101.5
Bromide	10.4	9.9	90.8

Column:	IonPac AG19, AS19, 4 mm
Eluent:	10 mM KOH for 0–10 min, 10-45 mM KOH for 10–25 min, 45 mM KOH for 25–30 min
Eluent Source:	EGC II KOH with CR-ATC
Temperature:	30 °C
Flow Rate:	1 mL/min
Inj. Volume:	250 µL
Detection:	(A) ASRS 300, 4 mm, external water mode (B) Absorbance, 352 nm
Postcolumn Reagent:	Acidified potassium iodide (KI)
PCR Flow Rate:	0.3 mL/min
Postcolumn Heater:	80 °C
Peaks (A):	1. Unknown – µg/L (ppb) 2. Bromate 2.4 3. Chlorate 62.4 4. Bromide 17.5
Peaks (B):	1. Unknown – 2. Bromate 2.8

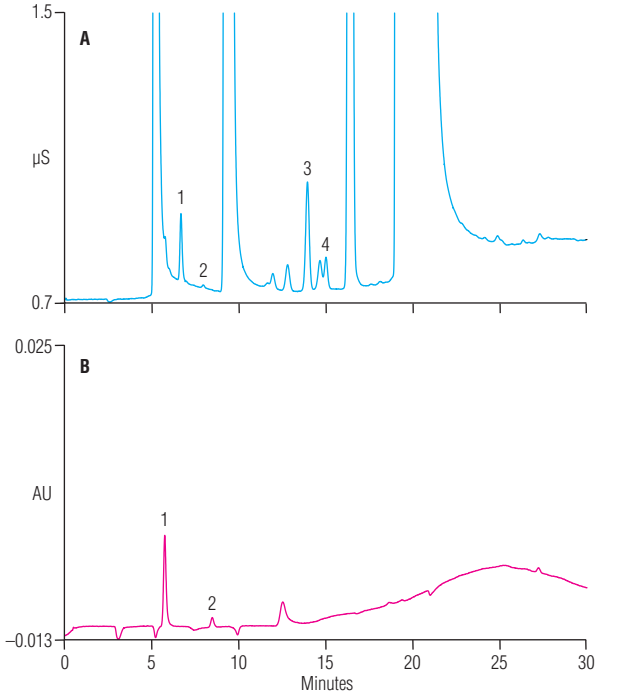

Figure 3. Determination of trace DBP anions and bromide in tap water B using suppressed conductivity detection and UV absorbance after PCR with acidified iodide.

Column:	IonPac AG19, AS19, 4 mm
Eluent:	10 mM KOH for 0–10 min, 10–45 mM KOH for 10–25 min, 45 mM KOH for 25–30 min
Eluent Source:	EGC II KOH with CR-ATC
Temperature:	30 °C
Flow Rate:	1 mL/min
Inj. Volume:	250 µL
Detection:	(A) ASRS 300, 4 mm, external water mode (B) Absorbance, 352 nm
Postcolumn Reagent:	Acidified potassium iodide (KI)
PCR Flow Rate:	0.3 mL/min
Postcolumn Heater:	80 °C
Peaks (A):	1. Chlorite 6.3 µg/L (ppb) 2. Bromate 5.5 3. Chlorate 133.3 4. Unknown – 5. Bromide 36.4
Peaks (B)	1. Chlorite – 2. Bromate 5.6

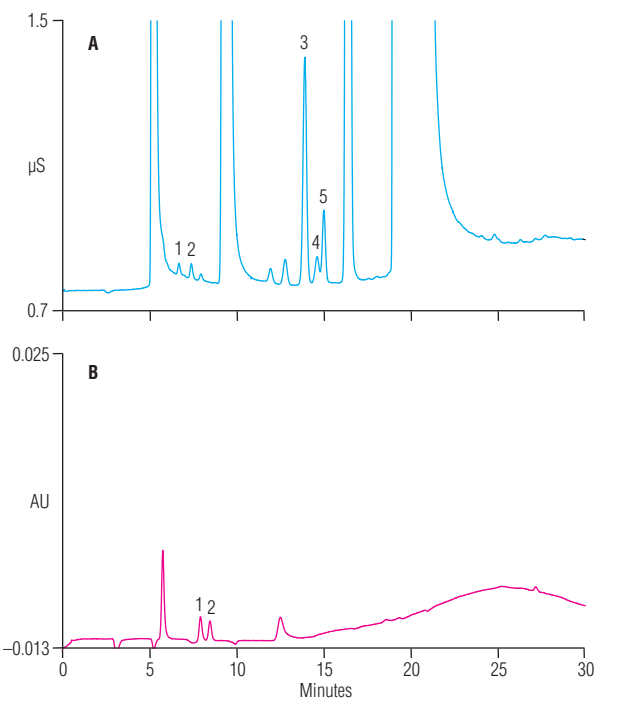

Figure 4. Determination of DBP anions spiked into tap water B using suppressed conductivity detection and UV absorbance after PCR with acidified potassium iodide.

waters and bottled drinking waters using the IonPac AS19 column. Figure 3 shows chromatograms of a 250 µL injection of Tap Water B using suppressed conductivity and UV detection at 352 nm after postcolumn reaction with acidified KI. Bromate, chlorate, and bromide were detected in the tap water. Bromide was not completely resolved from the earlier eluting unknown analyte. However, fortification of the sample with 20 µg/L bromide still produced good recovery of 92%. Bromate is clearly visible at approximately 3 µg/L with the absorbance detector; however, this bromate concentration was also easily determined using suppressed conductivity detection with the AS19 column. Figure 4 shows the same tap water sample spiked with chlorite, bromate, chlorate, and bromide at concentrations ranging from 3–70 µg/L. Analyte recoveries for this sample ranged from 92–108%.

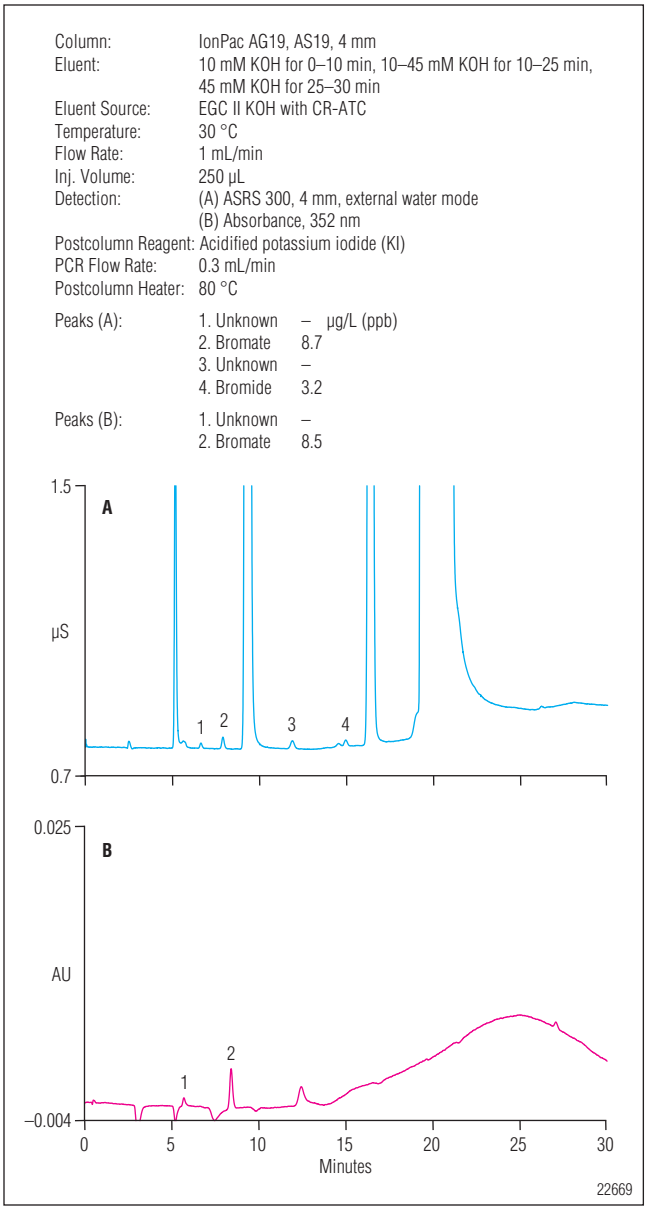

Figure 5. Determination of DBP anions and bromide in bottled water A-2 using suppressed conductivity detection and UV absorbance after PCR with acidified potassium iodide.

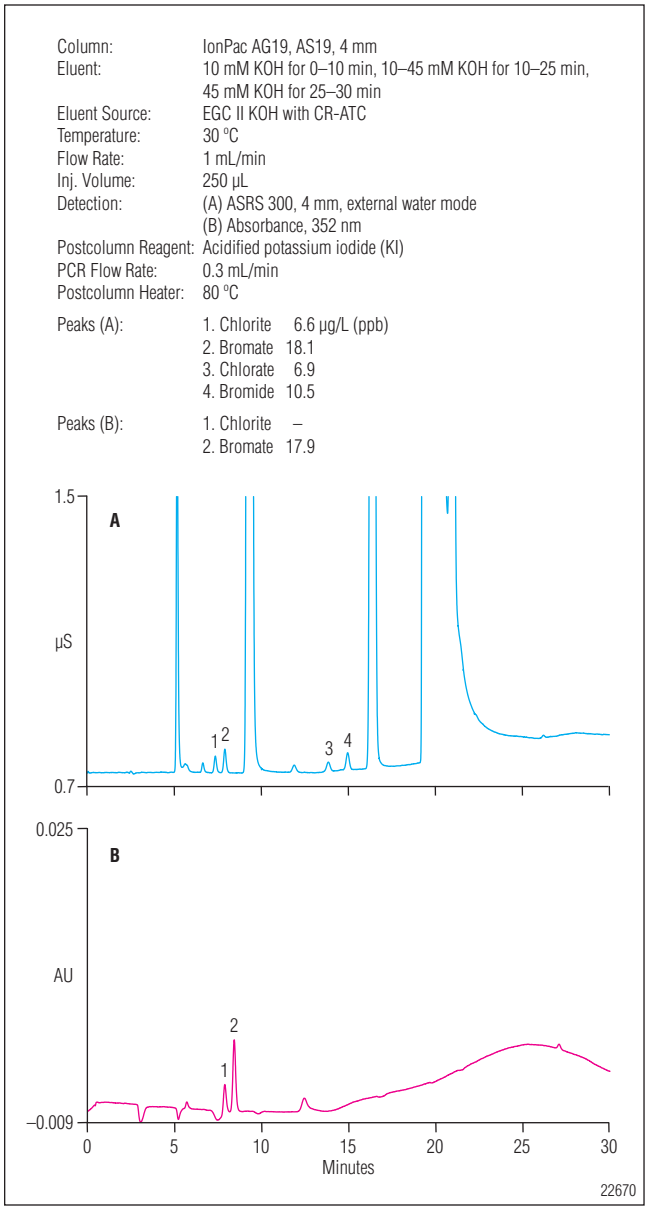

Figure 6. Determination of DBP anions and bromide in spiked bottled water A-2 using suppressed conductivity detection and UV absorbance after PCR with acidified potassium iodide.

Bottled Water A-2 is the same brand of bottled water product as A-1, except it was purchased approximately seven months later. The initial bromate concentration detected in A-1 was at the current EPA regulatory limit of 10 µg/L. However, the bromate concentration found in the second purchase (A-2) was ~8.7 µg/L, slightly below the regulatory limit. Figure 5 shows chromatograms of the ozonated bottled drinking water A-2 containing 8.7 µg/L bromate and 3.2 µg/L bromide. The top chromatogram (Figure 5A) shows the response of the target analytes obtained by suppressed conductivity detection; the bottom chromatogram (Figure 5B) was obtained by UV detection after postcolumn reaction with acidified KI. The bromate response is easily observed on both detector channels; however, the response using UV detection is enhanced compared to the conductivity detector. Figure 6 shows the same bottled drinking water sample spiked with 6–10 µg/L of the target DBP anions and bromide.

EPA 326.0 stipulates use of a surrogate to be added to a sample before filtration and other processing to monitor method performance. This surrogate should: 1) chemically resemble the target analytes, 2) be commercially available at a defined purity, 3) be stable in solution when properly stored, 4) be extremely unlikely to be found in the sample, and, 5) not coelute with the analytes of interest. The choice of surrogate used can be made by the analyst, but data must be maintained to show that the surrogate used meets the requirements above. The recommended surrogate in EPA 326.0 is DCA. This surrogate interferes with quantification when using a hydroxide eluent. Trichloroacetic acid (TCA) has been suggested as a replacement for DCA as a surrogate. For samples with high amounts of carbonate, the carbonate interferes with determination of TCA, making it a poor surrogate for this method. To minimize this peak overlap between carbonate and the surrogate, sodium malate was investigated. None of these options for use as a surrogate are detected by UV using the PCR. Malate is not typically present in drinking water samples, and it is also well-separated from the analytes of interest and from the peak resulting from carbonate without obscuring other peaks. Figure 7 shows the resolution possible when analyzing a municipal tap water sample containing chlorate, bromate, and bromide. The malate peak is separated from the carbonate peak, and is suitable for use as an internal surrogate. Eight sequential injectons of 1 mg/L malate in a DBP standard showed an RSD of 0.73 for peak area and an RSD of 0.01 for retention time. Malate is an appropriate surrogate for samples with high carbonate concentrations when using these conditions.

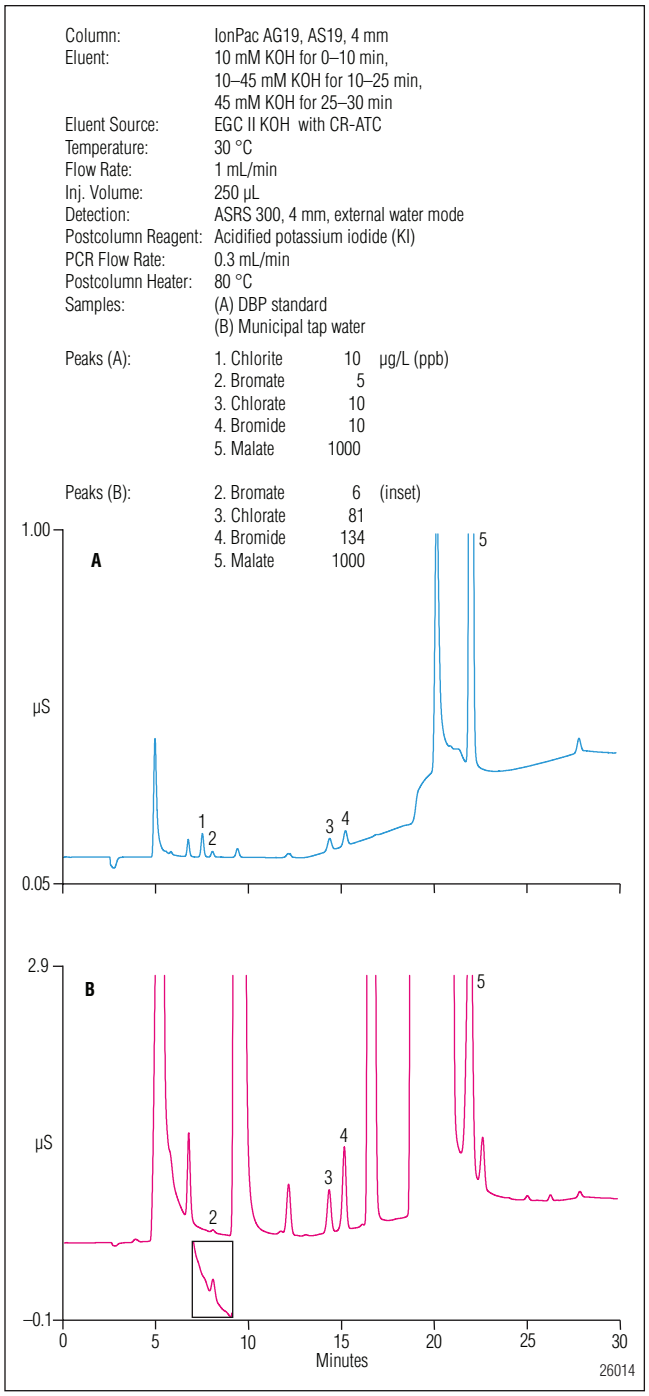

Figure 7. Determination of DBP in water using malate as a surrogate.

RUGGEDNESS

In order to test the method for ruggedness it was repeated for several weeks. With a range of PC10 delivery pressures between 26–54 psi and PCR flow rates ranging between 0.28–0.33 mL/min the peak area determined for a 1.0 ppb bromate standard had an RSD of 9.2 over 21 business days of analysis with a peak retention time RSD of 0.89. During this time period, two AMMS suppressors and two different flow cells were used to evaluate the effect of changes to the system on the response. Aside from the equilibration time required when changing an AMMS, no significant effect of the individual suppressor or flow cell was observed.

Low area responses in individual injections during this 21 day period were observed when the PC10 pressure required to deliver 0.3 mL/min of PCR increased when no other changes were made. For this reason, it is recommended that any increase in the PC10 delivery pressure from the pressure after initial system equilibrium, or any decrease in PCR flow be immediately investigated and corrected. Backpressure increases from the UV flow cell can be corrected by flushing the cell as described in the precautions section. Restrictions in tubing for the PCR will lead to poor results due to potential clogging and eventual changes in the flow of the reagent. Replace any crimped PEEK tubing to ensure consistent flow rates of all solutions.

Changes in the flow rate of the external water or the 0.3 N sulfuric acid will also change the baseline observed in the UV detection channel. Changes in the delivery pressures, and therefore flow rates, of these reagents will lead to baseline drift in the UV detection channel. As long as these flow rates are constant, a stable baseline will be achieved. If baseline drift is observed, confirm that no tubing is crimped or blocked before making changes to the consumables on the ICS-3000.

CONCLUSION

This application note describes an IC method using an electrolytically generated potassium hydroxide eluent combined with a hydroxide-selective IonPac AS19 column for determination of trace DBP anions and bromide using suppressed conductivity detection, followed by postcolumn addition of acidified KI with UV detection. The postcolumn reaction improves the selectivity and sensitivity for the determination of bromate in environmental water samples. The use of a hydroxide eluent improved the sensitivity for bromate using suppressed conductivity and UV detection compared to using a 9 mM carbonate eluent with the AS9-HC column, as described in Method 326.0. Furthermore, the use of postcolumn addition and UV detection with the AS19 column allowed quantification of bromate from 0.5–15 µg/L without compromising the detection of chlorite, bromate, chlorate, and bromide. However, the significant improvement in bromate detection by suppressed conductivity using electrolytically generated hydroxide eluent may eliminate the need for postcolumnn reaction for some environmental samples. Finally, this application demonstrates that the hydroxide-selective AS19 column combined with a hydroxide eluent can be successfully used in place of the AS9-HC column for compliance monitoring by USEPA Method 326.0.

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18. *Determination of Trace Concentrations of Disinfection Byproduct Anions and Bromide in Drinking Water Using Reagent-Free™ Ion Chromatography Followed by Postcolumn Addition of o-Dianisidine for Trace Bromate Analysis.* Application Note 168 (LPN 1706); Dionex Corporation, Sunnyvale, CA, 2005.

SUPPLIERS

Sigma-Aldrich Chemical Co., P.O. Box 2060, Milwaukee, WI, 53201, USA, Tel: 800-558-9160. www.sigma-aldrich.com.

Fluka Biochemika, 1001 West St. Paul Avenue, P.O. Box 2060, Milwaukee, WI, 53201, USA. Tel: 800-558-9160. www.sigma-aldrich.com

VWR, 1310 Goshen Parkway, West Chester, PA, 19380 USA. Tel: 800-932-5000. www.vwr.com

Determination of Trace Concentrations of Disinfection By-Product Anions and Bromide in Drinking Water Using Reagent-Free™ Ion Chromatography Followed by Postcolumn Addition of *o*-Dianisidine for Trace Bromate Analysis

INTRODUCTION

To ensure that public water systems (PWSs) are free from potentially dangerous microbes, the water is often disinfected before entering a community's distribution system.[1] The most common disinfectants are chlorine, chloramine, chlorine dioxide, and ozone. Many PWSs have converted from using chlorination to chloramination because chlorine treatment produces potential human carcinogens, such as trihalomethanes, that pose human health risks. However, chloramine can produce the by-product chlorate, whereas chlorine dioxide disinfection can generate both chlorite and chlorate.[2] Ozone is a particularly effective disinfection treatment that can alleviate most of the taste and odor issues often present in chlorinated water. However, ozonation of source water containing naturally occurring bromide can produce the disinfection by-product (DBP) bromate, a suspected human carcinogen. The World Health Organization (WHO) has estimated an excess lifetime cancer risk of 10^{-5} for drinking water containing 3 µg/L bromate.[3] In the U.S., the lifetime cancer risk was estimated to be 10^{-4} for drinking water containing 5 µg/L bromate with a potential 10^{-5} risk at 0.5 µg/L.[4]

The U.S. Environmental Protection Agency (EPA) requires that PWSs serving 100,000 or more connections report the concentration of target microorganisms present, the removal process used, and the concentration of DBPs present in their water.[5] In 1998, the EPA established a maximum contaminant level (MCL) of 10 µg/L bromate and 1000 µg/L chlorite in drinking water under the Stage 1 Disinfectants/Disinfection By-Products (D/DBP) Rule.[6] The European Union also reduced their regulatory value for bromate from 50 to 10 µg/L[7], and the WHO recently established the same provisional guideline of 10 µg/L bromate as technological advances allowed the determination of lower bromate concentrations.[8]

Traditionally, ion chromatography (IC) has been used to determine bromate and other oxyhalides in environmental waters as described in U.S. EPA Method 300.0 Part B.[9] This method uses an IonPac AS9-SC column with a reported method detection limit (MDL) of 20 µg/L bromate. EPA Method 300.1 was promulgated under the Stage 1 D/DBP Rule as an update to Method 300.0 to further reduce the bromate MDL to 1.4 µg/L. Method 300.1 uses an IonPac AS9-HC column, a high-capacity anion-exchange column, with a carbonate eluent and a large-loop injection followed by suppressed conductivity detection.[10] The MDL for bromate can be reduced to <1 µg/L using sample pretreatment followed by preconcentration.[11,12] However, this approach adds considerable complexity and cost to the analysis.

The U.S. EPA proposed the Stage 2 D/DBP Rule in 2003. Although no changes were made to the MCLs for bromate or chlorite, two postcolumn derivatization methods were promulgated to improve the selectivity and sensitivity for bromate.[13] U.S. EPA Method 317.0 is an extension of Method 300.1 B that combines suppressed conductivity detection and postcolumn addition of o-diansidine (ODA) followed by visible detection to achieve a bromate MDL of 0.1 µg/L with a practical quantitation limit (PQL) of 0.5 µg/L.[4,14] Bromate has also been detected by postcolumn reaction (PCR) with excess iodide under acidic conditions, as described in Method 326.0. The formation of the triiodide ion is detected spectrophotometrically at 352 nm, allowing an MDL <0.2 µg/L bromate using a large-injection volume.[15] IC coupled to mass spectrometry or inductively coupled plasma mass spectrometry has also been used to determine bromate in environmental waters, permitting bromate MDLs of 0.5 and 0.8 µg/L, respectively.[16,17]

Most promulgated EPA methods reported using an IonPac AS9-HC column with a carbonate eluent to determine trace bromate and other disinfection by-product anions in drinking waters. However, hydroxide eluents provide considerably lower suppressed background conductivity, lower noise, and therefore lower detection limits than carbonate eluents. An electrolytically generated hydroxide eluent combined with a hydroxide-selective IonPac AS19 column reduced the bromate MDL by more than 50% compared to using a carbonate eluent.[18] In this application note, we demonstrate the performance of the hydroxide-selective AS19 column for U.S. EPA Method 317.0. This method combines the advantages of a hydroxide eluent using suppressed conductivity detection with postcolumn addition to further improve the quantification of sub-µg/L bromate. The linearity, method detection limits, and the quantification of chlorite, bromate, chlorate, and bromide in municipal and bottled drinking waters are discussed.

EQUIPMENT

A Dionex ICS-2500 Reagent-Free Ion Chromatography system (RFIC™) consisting of:

- GP50 Gradient Pump with vacuum degas option
- CD25A Conductivity Detector
- AD25 UV-Vis Absorbance Detector with 10-mm cell
- AS50 Thermal Compartment with conductivity cell
- AS50 Autosampler
- EG50 Eluent Generator

EluGen® EGC-KOH Cartridge (Dionex P/N 058900)

Continuously Regenerated Anion Trap Column, CR-ATC (Dionex P/N 060477)

PC10 Pneumatic Postcolumn Delivery Module (Dionex P/N 050601)

PCH-2 Postcolumn Reaction Heater (Dionex P/N 039348)

Knitted Reaction Coil, 500 µL, potted (for PCH-2) (Dionex P/N 039349)

Two 4-L plastic bottle assemblies (for external water mode of suppression)

Chromeleon® Chromatography Workstation

REAGENTS AND STANDARDS

Deionized water, Type I reagent-grade, 18 MΩ-cm resistivity or better

o-Dianisidine, dihydrochloride salt (ODA, Sigma-Aldrich D-3252)

Ethylenediamine (EDA, Sigma-Aldrich E-1521)

Nitric Acid (70%, J. T. Baker INSTRA-ANALYZED 9598-00)

Methanol (spectrophotometric grade, Sigma-Aldrich M-3641)

Potassium bromide (KBr, J. T. Baker 2998)

Bromide standard 1000 mg/L, 100 mL (ULTRA Scientific, VWR P/N ICC-001)

Sodium Chlorite ($NaClO_2$, Fluka 71388, 80% pure)

Sodium Bromate ($NaBrO_3$, EM SX 03785-1)

Sodium Chlorate ($NaClO_3$, Fluka 71370)

CONDITIONS

Columns:	IonPac® AS19 Analytical, 4 × 250 mm (Dionex P/N 062885)
	IonPac AG19 Guard, 4 × 50 mm (Dionex P/N 062887)
Eluent:	10 mM KOH from 0–10 min, 10–45 mM from 10–25 min*
Eluent Source:	EG50 with CR-ATC
Flow Rate:	1.0 mL/min
Temperature:	30 °C
Injection:	250 µL
Detection:	Suppressed conductivity, ASRS® ULTRA II, 4 mm (Dionex P/N 061561), AutoSuppression® external water mode, 130 mA current
Background Conductance:	<1 µS
System Backpressure:	~2200 psi
Run Time:	30 min

PCR

Detection:	Absorbance at 450 nm (tungsten lamp)
Postcolumn Reagent Flow:	0.54 mL/min
Postcolumn Heater Temperature:	60 °C

*Method returns to 10 mM KOH for 3 min prior to injection

PREPARATION OF SOLUTIONS AND REAGENTS

Postcolumn Reagent

Add 40 mL of 70% nitric acid to about 300 mL of deionized (DI) water in a 500-mL volumetric flask. Add 2.5 g potassium bromide and stir to dissolve. Dissolve 250 mg *o*-dianisidine • 2HCl in 100 mL methanol, add to the nitric acid/KBr solution, and bring to volume with DI water. Allow the solution to stand overnight until the slight champagne color fades. Then filter through a 0.45-µm filter before use.

Stock Standard Solutions

Prepare 1000 mg/L stock standard solutions by dissolving the corresponding mass of the salt in 100 mL DI water (Table 1). Stock standards for most anions listed in Table 1 are stable for at least 6 months when stored at <6 °C. Chlorite is only stable for two weeks when stored at <6 °C and protected from light.

TABLE 1. MASSES OF COMPOUNDS USED TO PREPARE 100 mL OF 1000 mg/L ANION STANDARDS

Analyte	Compound	Amount (g)
Chlorite	Sodium chlorite ($NaClO_2$), 80%	0.1676
Bromate	Sodium bromate ($NaBrO_3$)	0.1180
Chlorate	Sodium chlorate ($NaClO_3$)	0.1275
Bromide	Sodium bromide (NaBr)	0.1288

Prepare a secondary stock standard containing 5 mg/L each of chlorite, chlorate, and bromide by combining 0.5 mL of each anion in a 100-mL volumetric flask and dilute to volume with DI water. Prepare a separate secondary stock standard containing bromate only at 1 mg/L by adding 0.1 mL of the 1000-mg/L bromate stock to a 100-mL volumetric flask and dilute to volume with DI water.

Working Standard Solutions

Prepare dilute working standards by performing appropriate dilutions of the secondary stock solutions as necessary. Dilute working standards should be prepared monthly, except those that contain chlorite, which must be prepared every two weeks or sooner if degradation is indicated by repeated quality check failures.

Preservation Solution

Dilute 2.8 mL of 99% ethylenediamine (EDA) to 25 mL with DI water according to Section 7.4 in EPA Method 317.0 to prepare a 100-mg/mL EDA solution. Use 50 µL of 100-mg/mL EDA per 100 mL of standard or sample so the final EDA concentration is 50 mg/L. Prepare fresh monthly.

SAMPLE PREPARATION

Filter samples, as necessary, through single-use 0.45-µm syringe filters, discarding the first 300 µL of the effluent. To prevent degradation of chlorite or the formation of bromate from hypobromous acid/hypobromite, preserve the samples by adding 50 µL of EDA preservation solution per 100 mL of sample.

SYSTEM PREPARATION AND SETUP

Prepare the ASRS ULTRA II for use by hydrating the suppressor. Use a disposable plastic syringe and push approximately 3 mL of degassed DI water through the "Eluent Out" port and 5 mL of degassed DI water through the "Regen In" port. Allow the suppressor to stand for approximately 20 min to fully hydrate the suppressor screens and membranes. Install the ASRS ULTRA II for use in the external water mode by connecting the "Regen Out" of the suppressor to the "Regen In" of the CR-ATC. The "Regen In" of the suppressor should connect directly to the external water source. The "Regen Out" of the CR-ATC is then connected to the "SRS Waste In" of the EG50 degasser. This configuration allows the eluent out of the analytical column to be connected to the mixing tee of the PCR system.

Install the EGC II KOH cartridge in the EG50 and configure it with the CR-ATC according to the *CR-TC Quickstart* (Document No. 031911). Use the Chromeleon system configuration to set up the EGC II KOH cartridge with the software. Condition the cartridge as directed by the EGC II Quickstart (Document No. 031909) with 50 mM KOH at 1 mL/min for 30 min. Install a 4 × 50 mm AG19 and 4 × 250 mm AS19 column. Make sure the pressure displayed by the pump is at an optimal pressure of ~2300 psi when 45 mM KOH is delivered at 1 mL/min. This setting allows the EG50 degas assembly to effectively remove hydrolysis gases from the eluent. If necessary, install additional backpressure tubing to adjust the pressure to 2300 ± 200 psi.

Configure the ICS-2500 with the PCR system as shown in Figure 1. To maintain a 1-mL/min analytical flow rate, the PCR flow rate was determined based on the analytical to PCR flow rate ratio provided in EPA Method 317.0. For the ICS-2500 system, this ratio resulted in the use of 0.54 mL/min PCR flow rate. Set the temperature on the PCH-2 to 60 °C and the wavelength on the AD25 to 450 nm. Measure the PCR flow rate at the operating parameters by collecting the combined effluent from the IC pump and PCR system in a 10-mL graduated cylinder for at least 5 min. The PCR flow rate is the difference between the total flow rate and that of the IC pump divided by the amount of time used for collection (e.g., 5 min). Adjust the pressure of the postcolumn delivery module (PC10) and measure the flow rate again until the correct flow rate of 0.54 mL/min is achieved. Confirm the flow rate daily, whenever the PCR is changed, and if the quality control standard deviates from the EPA's acceptance criteria. Allow both the suppressed conductivity and visible detection baselines to equilibrate. Prior to analyzing any samples, inject 250 µL of DI water using the described method. This is the method blank. No peaks should elute at the same retention time as the target analytes. An equilibrated system has a suppressed background conductance <1 µS and peak-to-peak noise of ~1–2 nS/min.

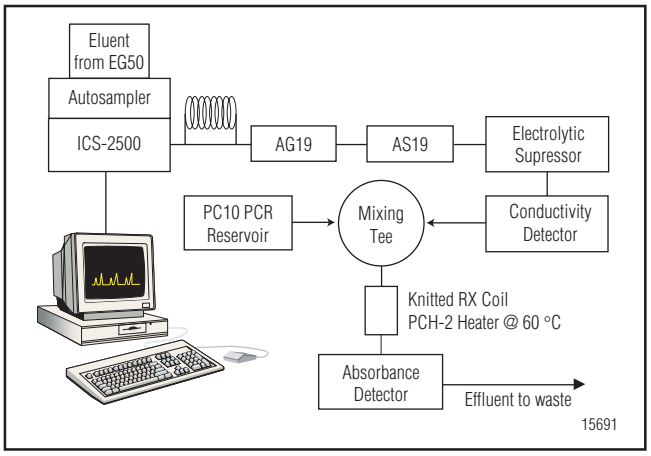

Figure 1. System Configuration for EPA Method 317.0

RESULTS AND DISCUSSION

U.S. EPA Method 317.0 specifies the use of an IonPac AS9-HC column with a 9 mM sodium carbonate eluent for the determination of chlorite, chlorate, and bromide by suppressed conductivity detection and bromate by suppressed conductivity and visible detection after postcolumn reaction with o-dianisidine (ODA).[14] This method reports a bromate detection limit of 0.71 µg/L for a 225-µL injection by suppressed conductivity and 0.12 µg/L by visible absorbance (225-µL injection). Previously, we demonstrated that the bromate detection limit can be reduced further to 0.34 µg/L using an electrolytically generated hydroxide eluent with a novel hydroxide-selective IonPac AS19 column and detection by suppressed conductivity.[18] In this application note, we examine the feasibility of using the IonPac AS19 column with the combination of suppressed conductivity detection and a postcolumn reaction system for visible absorbance detection. The use of a

suitable hydroxide-selective column for this application allows for lower detection limits for the target disinfection by-product anions by suppressed conductivity detection while still providing the improved sensitivity and selectivity for bromate obtained by the PCR.

A calibration curve was established for determining the target analytes, chlorite, bromate, chlorate, and bromide by conductivity detection. In this application, chlorite, chlorate, and bromide were calibrated from 5–500 µg/L, as suggested by Method 317.0. This calibration range is expected to cover the concentrations found in typical environmental samples. However, in field samples, bromate is usually present at significantly lower concentrations than other inorganic DBP anions. The improved sensitivity obtained using an electrolytically generated hydroxide eluent allowed a lower PQL of 1 µg/L bromate compared to 5 µg/L using the AS9-HC column with a carbonate eluent. Therefore, bromate was calibrated from 1–40 µg/L, which is expected to cover concentrations found in most environmental samples. According to Method 317.0, the linear range should not cover more than two orders of magnitude in concentration. Because our linear range extended two orders of magnitude in concentration, seven calibration standards were used. Bromate was calibrated over the range 0.5–15 µg/L with the PCR system. Although this calibration is less than two orders of magnitude, Method 317.0 still recommends using at least five calibration standards for the absorbance detector. Table 2 summarizes the calibration data and method detection limits (MDLs) obtained for the DBP anions and bromide using the AS19 column. The MDLs for the target analytes were determined by performing seven replicate injections of reagent water fortified at a concentration of three to five times the estimated instrument detection limit.[14] The use of a PCR system and visible detection with a hydroxide-selective column provides a bromate PQL of 0.5 µg/L, comparable to that reported in Method 317.0 using the AS9-HC column. Also, the addition of a PCR system did not compromise the sensitivity obtained by suppressed conductivity detection using a hydroxide eluent. Figure 2 shows chromatograms of the target DBP anions containing 5 µg/L bromate and 10 µg/L each of chlorite, chlorate, and bromide using suppressed conductivity (Figure 2A) and visible detection (Figure 2B) following postcolumn addition of ODA. Notice the enhanced response for bromate on the absorbance detector compared to the conductivity detector.

TABLE 2. LINEARITY AND MDLs FOR DISINFECTION BY-PRODUCT ANIONS AND BROMIDE

Analyte	Range (µg/L)	Linearity (r^2)	MDL Standard (µg/L)	Calculated MDL (µg/L)
Chlorite	5–500	0.9982	1.0	0.26
Bromate (conductivity)	1–40	0.9997	1.5	0.32
Bromate (Vis)	0.5–15	0.9996	0.5	0.14
Chlorate	5–500	0.9999	1.3	0.38
Bromide	5–500	0.9997	2.0	0.52

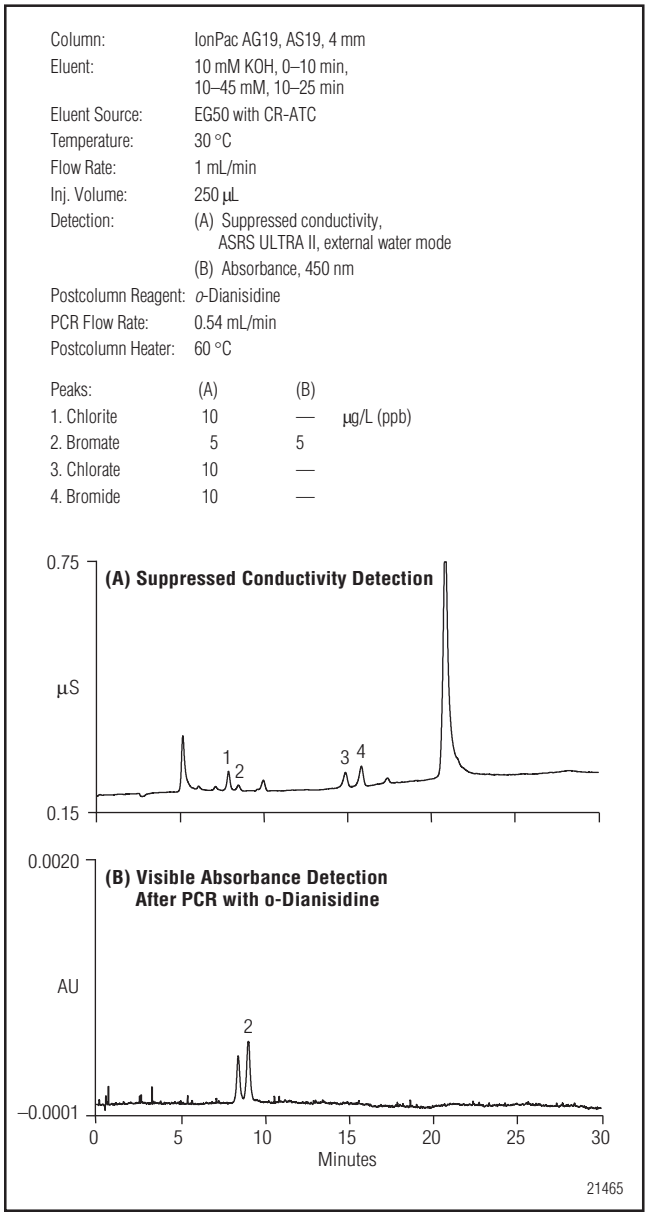

Figure 2. Separation of low ppb DBP anions and bromide using the IonPac AS19 column

TABLE 3. RECOVERIES OF TRACE DBP ANIONS AND BROMIDE SPIKED INTO WATER SAMPLES

	Tap water			Bottled water A			Bottled water B			Surface water		
Analyte	**Amount found (µg/L)**	**Amount added (µg/L)**	**Recovery (%)**	**Amount found (µg/L)**	**Amount added (µg/L)**	**Recovery (%)**	**Amount found (µg/L)**	**Amount added (µg/L)**	**Recovery (%)**	**Amount found (µg/L)**	**Amount added (µg/L)**	**Recovery (%)**
Chlorite	<MDL	5.0	97.0	<MDL	5.0	94.6	<MDL	5.0	97.5	<MDL	5.0	104.5
Bromate (conductivity)	2.5	3.0	103.3	10.0	10.0	95.7	<MDL	1.0	110.5	<MDL	1.0	103.4
Bromate (Vis)	2.2	3.0	96.3	10.1	10.0	102.8	<MDL	1.0	106.9	<MDL	1.0	97.4
Chlorate	64.0	73.0	94.2	<MDL	5.0	99.0	1.6	5.0	104.0	<MDL	5.0	103.7
Bromide	19.0	20.0	98.1	18.0	20.0	97.5	0.9	5.0	111.5	<MDL	5.0	102.0

EPA Method 317.0 requires an initial demonstration of capability to characterize the instrument and laboratory performance of the method prior to performing sample analyses, as described in Section 9.2.[14] An initial demonstration of precision, accuracy, and analysis of a quality control sample (QCS) are part of the criteria used for this characterization. For evaluating the precision and accuracy of the conductivity detector, Method 317.0 recommends using 20 µg/L each of the four target DBP anions. However, because the use of an electrolytically generated hydroxide eluent improves the overall sensitivity of the method, we determined that the use of 5 µg/L bromate and 10 µg/L each of chlorite, chlorate, and bromide was suitable for characterizing the instrument and laboratory performance. For the absorbance detector, a recommended concentration of 2 µg/L bromate was used. EPA Method 317.0 considers a %RSD <20% and an average recovery of ±15% to be acceptable performance. The precision of our replicate analyses was <4.5% and the recovery was 94–103%, well within EPA's acceptance criteria. A QCS should be analyzed after the calibration curves are initially established, on a quarterly basis, or as required to meet data quality objectives. All QCS analyses in our experiments met the EPA's ±20% recovery criteria.

The analyte recoveries for the target DBP anions and bromide were assessed by fortifying known amounts of the anions into the field samples. The concentrations were fortified at concentrations equal to or greater than the native concentrations. Table 3 summarizes the recovery data for the analysis of drinking water, surface water, and bottled drinking water samples. As shown, analyte recoveries were in the range of 94–112%, well within the 75–125% acceptance criteria of Method 317.0. Figures 3–6 illustrate the performance for the determination of DBP anions and bromide in municipal tap water and bottled drinking water using the IonPac AS19 column. Figure 3 shows chromatograms of a 250-µL injection of tap water using suppressed conductivity and visible detection at 450 nm after postcolumn addition of ODA. In this sample, bromate, chlorate, and bromide were detected in the tap water. Bromate is clearly visible at about 2 µg/L with the absorbance detector. However, this bromate concentration can also be easily determined using suppressed conductivity detection with the method parameters described in this application document. Figure 4 shows the same tap water sample spiked with chlorite, bromate, chlorate, and bromide at concentrations ranging from 3–73 µg/L. Analyte recoveries for this sample ranged from 94–103%.

Figure 5 shows chromatograms of bottled drinking water B using suppressed conductivity and visible detection after postcolumn reaction with ODA. The conductivity detector observed only trace amounts of chlorate and bromide in the sample. However, no bromate was found with either of the detection methods, which is in agreement with the manufacturer who did not report using any ozonation as a disinfection treatment for this bottled water product. Figure 6 shows the same sample spiked with 1–5 µg/L of the target DBP anions. The average recoveries of the spiked sample were 97–112%, well within the acceptance criteria.

Column:	IonPac AG19, AS19, 4 mm
Eluent:	10 mM KOH, 0–10 min, 10–45 mM, 10–25 min
Eluent Source:	EG50 with CR-ATC
Temperature:	30 °C
Flow Rate:	1 mL/min
Inj. Volume:	250 µL
Detection:	(A) Suppressed conductivity, ASRS ULTRA II, external water mode
	(B) Absorbance, 450 nm
Postcolumn Reagent:	*o*-Dianisidine
PCR Flow Rate:	0.54 mL/min
Postcolumn Heater:	60 °C

Peaks:	(A)	(B)	
2. Bromate	2.5	2.2	µg/L (ppb)
3. Chlorate	64	—	
4. Bromide	19	—	

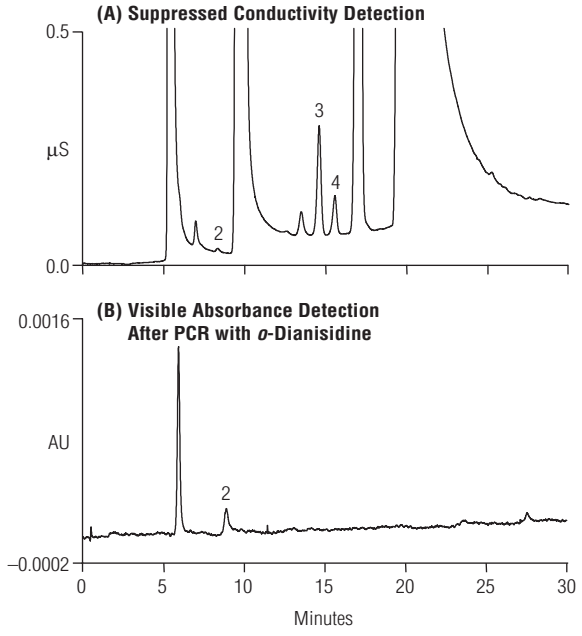

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Figure 3. Determination of trace DBP anions and bromide in tap water.

Column:	IonPac AG19, AS19, 4 mm
Eluent:	10 mM KOH, 0–10 min, 10–45 mM, 10–25 min
Eluent Source:	EG50 with CR-ATC
Temperature:	30 °C
Flow Rate:	1 mL/min
Inj. Volume:	250 µL
Detection:	(A) Suppressed conductivity, ASRS ULTRA II, external water mode
	(B) Absorbance, 450 nm
Postcolumn Reagent:	*o*-Dianisidine
PCR Flow Rate:	0.54 mL/min
Postcolumn Heater:	60 °C

Peaks:	(A)	(B)	
1. Chlorite	4.9	—	µg/L (ppb)
2. Bromate	5.6	5.1	
3. Chlorate	133	—	
4. Bromide	38.4	—	

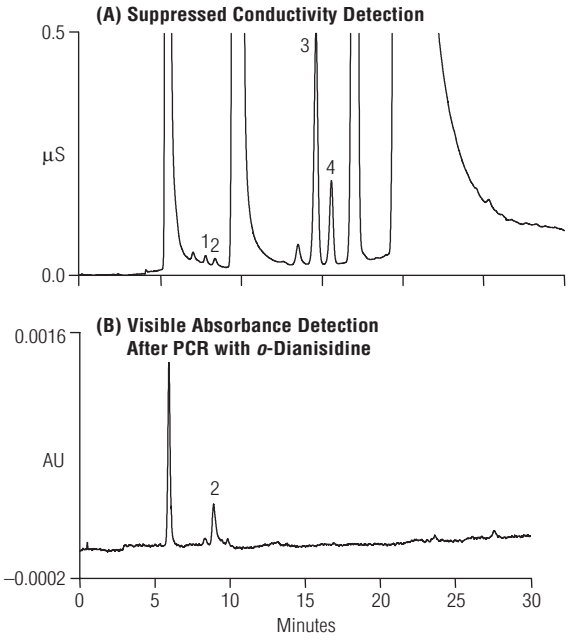

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Figure 4. Determination of trace DBP anions and bromide in spiked tap water.

CONCLUSION

This application note described an IC method that used an electrolytically generated potassium hydroxide eluent combined with a hydroxide-selective IonPac AS19 column for the determination of trace DBP anions and bromide. This method used suppressed conductivity detection followed by postcolumn addition of ODA with visible detection to improve the selectivity and sensitivity for the determination of bromate in environmental waters. The use of a hydroxide eluent improved the sensitivity for bromate using suppressed conductivity, compared to a 9 mM carbonate eluent used with the AS9-HC column, as described in Method 317.0. However, comparable sensitivities for both the AS19 and AS9-HC columns were observed using postcolumn addition of ODA and visible detection. The use of postcolumn addition and visible detection with the AS19 column allowed quantification of bromate from 0.5–15 µg/L without compromising the suppressed conductivity detection of chlorite, bromate, chlorate, and bromide. This application document demonstrates that the hydroxide-selective AS19 column combined with a hydroxide eluent can be successfully used in place of the AS9-HC column for compliance monitoring by U.S. EPA Method 317.0.

Column:	IonPac AG19, AS19, 4 mm
Eluent:	10 mM KOH, 0–10 min, 10–45 mM, 10–25 min
Eluent Source:	EG50 with CR-ATC
Temperature:	30 °C
Flow Rate:	1 mL/min
Inj. Volume:	250 µL
Detection:	(A) Suppressed conductivity, ASRS ULTRA II, external water mode
	(B) Absorbance, 450 nm
Postcolumn Reagent:	*o*-Dianisidine
PCR Flow Rate:	0.54 mL/min
Postcolumn Heater:	60 °C

Peaks:	(A)	(B)	
3. Chlorate	1.6	—	µg/L (ppb)
4. Bromide	0.9	—	

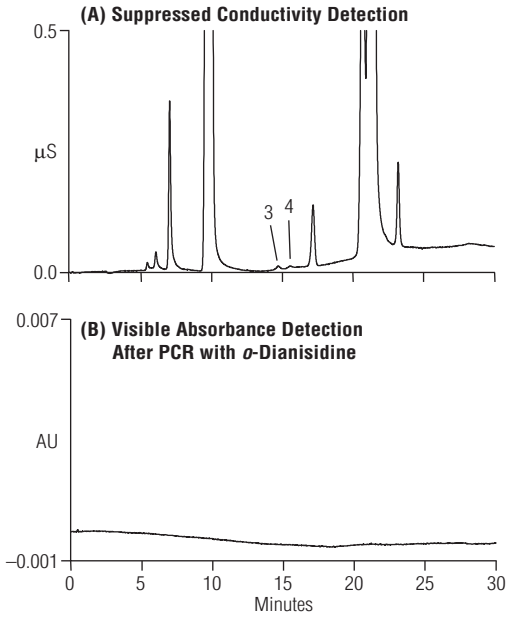

Figure 5. Determination of trace DBP anions and bromide in bottled drinking water B.

Column:	IonPac AG19, AS19, 4 mm
Eluent:	10 mM KOH, 0–10 min, 10–45 mM, 10–25 min
Eluent Source:	EG50 with CR-ATC
Temperature:	30 °C
Flow Rate:	1 mL/min
Inj. Volume:	250 µL
Detection:	(A) Suppressed conductivity, ASRS ULTRA II, external water mode
	(B) Absorbance, 450 nm
Postcolumn Reagent:	*o*-Dianisidine
PCR Flow Rate:	0.54 mL/min
Postcolumn Heater:	60 °C

Peaks:	(A)	(B)	
1. Chlorite	5.1	—	µg/L (ppb)
2. Bromate	1.0	1.1	
3. Chlorate	5.0	—	
4. Bromide	5.0	—	

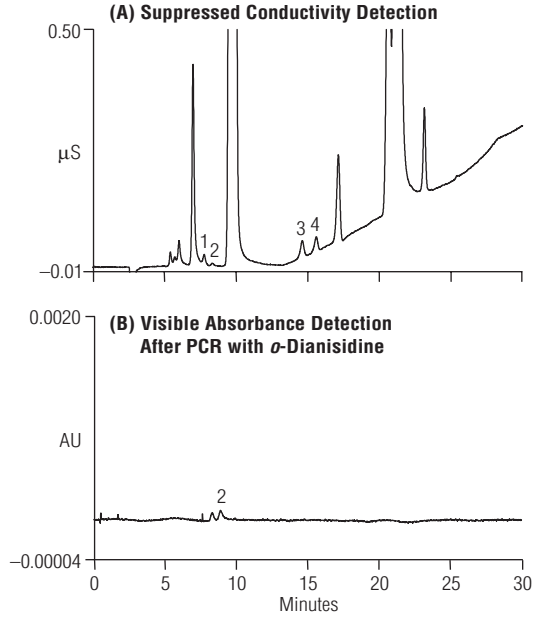

Figure 6. Determination of trace DBP anions and bromide in spiked bottled drinking water B.

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17. U.S. EPA Method 321.8; U.S. Environmental Protection Agency, Cincinnati, OH, 1997.
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SUPPLIERS

Sigma-Aldrich Chemical Co., P.O. Box 2060, Milwaukee, WI 53201, USA, Tel.: 800-558-9160, www.sigmaaldrich.com

Fluka, P.O. Box 2060, Milwaukee, WI 53201, USA, Tel.: 800-558-9160, www.sigmaaldrich.com

Determination of Chlorite, Bromate, Bromide, and Chlorate in Drinking Water by Ion Chromatography with an On-Line-Generated Postcolumn Reagent for Sub-µg/L Bromate Analysis

INTRODUCTION

Public water suppliers treat drinking water with disinfectants to protect public health and give drinking water a pleasant taste and odor. Unfortunately, some of the chemical disinfectants or by-products of the disinfection process are themselves harmful. For example, chlorine dioxide generates the inorganic oxyhalide disinfection by-products (DBPs) chlorite and chlorate; hypochlorite treatment may also generate the DBP chlorate;[1] and ozonating source water that contains elevated levels of natural bromide can produce the DBP bromate.[2] Both the World Health Organization (WHO) and the U.S. Environmental Protection Agency (EPA) have listed bromate as a potential carcinogen at the low-µg/L level.[3]

EPA's Stage 1 Disinfectants/Disinfection By-Products rule (D/DBP) specifies a maximum contaminant level (MCL) of 10 µg/L for bromate, an MCL of 1000 µg/L[4] for chlorite, and prescribes EPA Method 300.1[5] for compliance monitoring of bromate and chlorite in drinking water. It is expected that when the EPA promulgates Stage 2 of the D/DBP rule, the MCL for bromate will remain at 10 µg/L and the EPA will propose additional methods for compliance monitoring to add flexibility and improved performance. Until then, the EPA is evaluating new methods with improved performance for D/DBP monitoring, including EPA Method 317.0 (IC-PCR, Dionex Application Note 136), EPA Method 321.8 (IC/ICP-MS), and EPA Method 326.0 (IC-PCR).[6–8]

This application note describes an improved ion chromatography (IC) method to quantify oxyhalide DBP anions and bromide at low concentration levels in reagent water, bottled water, and finished drinking water using an approach that is technically equivalent to U.S. EPA Method 326.0. The oxyhalide anions chlorite, chlorate, bromide, and bromate are separated on an IonPac® AS9-HC column and measured by using suppressed conductivity detection (as in EPA Method 300.1), followed by postcolumn reaction (PCR) to enhance detection of bromate. Sensitivity for bromate is improved by more than a factor of 10 through the use of a postcolumn reaction in which hydroiodic acid (HI) generated *in situ* from potassium iodide (KI) reacts with bromate in the column effluent to form the triiodide anion (I_3^-) as shown in the following set of reactions:[9]

$$BrO_3^- + 3I^- + 3H^+ \rightleftharpoons 3HOI + Br^-$$
$$3HOI + 3I^- + 3H^+ \rightleftharpoons 3I_2 + 3H_2O$$
$$3I_2 + 3I^- \rightleftharpoons 3I_3^-$$

Triiodide is then detected by its strong absorbance at 352 nm.

Because the HI PCR reagent is generated on-line and used immediately, reagent purity and stability should be more easily ensured than in EPA Method 317.0. It is also advantageous from a safety and exposures standpoint to use the *in situ* generated HI versus the toxic *o*-dianisidine (ODA) PCR reagent employed in Method 317.0.

Method 326.0 allows for the determination of all three key oxyhalide anions and bromide at low-µg/L levels using conductivity detection. Bromate can be quantified down to 0.5 µg/L using PCR with UV absorbance detection. Although Method 326.0 is not yet promulgated by the U.S. EPA Office of Ground Water and Drinking Water, the conductivity portion of the method has been determined acceptable for compliance monitoring for the oxyhalide DBPs and bromide.

EQUIPMENT

A Dionex DX-600 ion chromatographic system consisting of:
- GP50 Gradient Pump with Vacuum Degas Option
- ED50A Conductivity Detector with AS50 Conductivity Cell (P/N 55400)
- AD25 UV/Vis Absorbance Detector with 10-mm Cell
- AS50 Automated Sampler with Thermal Compartment
- PC10 Pneumatic Postcolumn Delivery Module (P/N 50601)
- Anion MicroMembrane™ (AMMS®) III Suppressor

PCH-2 Reaction Heater (P/N 39348)

Knitted Reaction Coil, 500 µL, Potted (for PCH-2) (P/N 39349)

Two 4-L plastic bottle assemblies (for external water mode suppression)

Chromeleon® Chromatography Workstation

REAGENTS AND STANDARDS

Deionized water, Type I reagent-grade, 18 MΩ-cm resistivity or better

0.5 M sodium carbonate (Na_2CO_3) Anion Eluent Concentrate (Dionex P/N 37162)

Potassium iodide (KI) (Sigma P-8256) or (Fisher P-410)

Ammonium molybdate tetrahydrate [$(NH_4)_6 Mo_7O_{24}•4H_2O$] (Aldrich 22,136-6)

Iron (II) sulfate heptahydrate ($FeSO_4•7H_2O$) (Aldrich 21,542-2)

Ethylenediamine (EDA) (Alfa Products 11932)

Dichloroacetic acid (DCAA) (Fluka 35810)

Sulfuric acid, (18M) (J.T. Baker INSTRA-ANALYZED 9673-33)

Nitric acid, (70%) (J.T. Baker INSTRA-ANALYZED 9598-00)

Bromate standard, 1000 mg/L, $NaBrO_3$ in H_2O (SPEX CertiPrep AS-$BRO_3$9-2Y)

Bromide standard, 1000 mg/L, NaBr in H_2O (ULTRA Scientific ICC-001)

Chlorate standard, 1000 mg/L, $NaClO_3$ in H_2O (SPEX CertiPrep AS-$CLO_3$9-2Y)

Chlorite standard, 1000 mg/L, $NaClO_2$ in H_2O (SPEX CertiPrep AS-$CLO_2$9-2Y)

Sodium bromide (NaBr) (Aldrich 31,050-6)

Sodium bromate ($NaBrO_3$) (EM SX 03785-1)

Sodium chlorate ($NaClO_3$) (Fluka 71370)

Sodium chlorite ($NaClO_2$) (Fluka 71388, ~80% pure)

CONDITIONS

Columns:	Dionex IonPac AG9-HC, 50 × 4 mm i.d. Guard Column (Dionex P/N 51791) Dionex IonPac AS9-HC, 250 × 4 mm i.d. Analytical Column (Dionex P/N 51786)
Eluent:	9.0 mM sodium carbonate (Na_2CO_3)
Flow Rate:	1.3 mL/min
Temperature:	30 µC
Sample Volume:	225 µL
Detection:	Suppressed Conductivity, Anion Atlas® Electrolytic Suppressor (AAES™) (P/N 056116) AutoSuppression® external water mode, 78 mA Temperature compensation, 1.7%/°C
Expected Background:	~23–26 ⊠S
Expected Backpressure:	~2400 psi
Run Time:	20 min

PCR

Detection:	Absorbance at 352 nm
PCR Reagent Flow:	0.26 M potassium iodide at 0.4 mL/min
AMMS III:	0.3 N sulfuric acid at 2.5 mL/min
Postcolumn Heater Temp:	80 °C

PREPARATION OF SOLUTIONS AND REAGENTS

Reagent Water

Distilled or deionized water 18 MΩ-cm or better, free of the anions of interest, and filtered through a 0.2-micron filter.

Eluent Solution

9 mM sodium carbonate

Dilute 36 mL of 0.5 M sodium carbonate concentrate to 2 L with deionized water. Unless the in-line degas option is being used, sparge eluent prior to use with helium or sonicate under vacuum for 10 min.

Ethylenediamine (EDA) Preservative Solution

Dilute 2.8 mL of ethylenediamine (99%) to 25 mL with reagent water. Prepare the solution fresh monthly.

Ferrous Iron Solution [1000 mg/L Fe (II)]

Add 0.124 g of ferrous sulfate heptahydrate ($FeSO_4 \cdot 7H_2O$) to about 15 mL of reagent water containing 6 ≈L concentrated nitric acid in a 25-mL volumetric flask. Dissolve and bring to volume with reagent water (final pH ~2). Prepare fresh every two days.

Sulfuric Acid Solution (0.5 N)

Dilute 1.4 mL of concentrated sulfuric acid to 100 mL with reagent water.

Ammonium Molybdate Solution (2.0 mM)

Add 0.247 g of ammonium molybdate tetrahydrate [$(NH_4)_6\ Mo_7O_{24} \cdot 4H_2O$)] to about 50 mL of reagent water in a 100-mL volumetric flask. Dissolve and bring to volume with reagent water. Store in an opaque plastic bottle and prepare fresh monthly.

Postcolumn Reagent

Add 43.1 g of potassium iodide to about 500 mL of reagent water in a 1-L volumetric flask and mix to dissolve. Add 215 µL of the ammonium molybdate solution. Bring to volume with reagent water and mix. Remove dissolved gasses by sparging with helium or by sonicating under vacuum for 20 min. Immediately place it in the PC-10 reagent delivery vessel and blanket with helium. Protect from light by covering the PC-10 module with aluminum foil. The reagent is stable for 24 h under these conditions.

Stock Standard Solutions

Purchase certified solutions or prepare stock standard solutions by dissolving the corresponding mass of the salt for each of the anions of interest (see Table 1) in deionized water and dilute to 100 mL.

Prepare a mixed anion calibration stock standard at 20 mg/L by combining 2 mL each of the bromide, chlorite, and chlorate stock standards in a 100 mL volumetric flask. Mix and bring to volume with reagent water. These standards are stable for at least one month when stored at less than 6 °C.

Because bromate decomposes in the presence of chlorite, prepare a bromate-only calibration stock standard at 5 mg/L by adding 0.5 mL of the bromate stock standard to a 100-mL volumetric flask and bringing to volume with reagent water. This standard is stable for two weeks when stored at less than 6 °C.

Working Standard Solutions

Use deionized water to prepare appropriate dilutions of the calibration stock standards as needed. Prepare mixed calibration standards containing all four anions fresh each day as needed.

SAMPLE PREPARATION

Sparge the water samples taken from a treatment plant employing chlorine dioxide or ozone with an inert gas (e.g., nitrogen, argon, or helium) for 5 min. Add 1.00 mL of EDA preservation solution per 1 L of sample to prevent conversion of residual hypochlorite or hypobromite to chlorate or bromate. This solution also prevents metal-catalyzed conversion of chlorite to chlorate. Samples preserved in this manner are stable for at least 14 days when stored in amber bottles at 4 °C.[10]

Table 1. Masses of Compounds Used to Prepare 100 mL of 1000-mg/L Anion Standards

Anion	Compound	Mass (g)
BrO_3^-	Sodium bromate ($NaBrO_3$)	0.1180
Br^-	Sodium bromide (NaBr)	0.1288
ClO_3^-	Sodium chlorate ($NaClO_3$)	0.1275
ClO_2^-	Sodium chlorite ($NaClO_2$)	0.1344*

* Mass of pure (>99%) sodium chlorite. For accurate results, determine the exact purity of $NaClO_2$ by using the iodometric titration procedure[14] and adjust the mass of the compound used accordingly. For example, for technical-grade sodium chlorite (80% pure) use (0.1344 g)(100%/80%) = 0.1680 g.

Most samples preserved as above can be filtered through a 0.45-micron filter (Gelman IC Acrodisk P/N 4485 or equivalent) and directly injected onto the ion chromatograph. However, each sample that contains excess chlorite must be treated to remove chlorite and then reanalyzed for bromate, because elevated levels of chlorite can interfere with the bromate quantification by PCR.

The treatment procedure to remove chlorite requires two portions of the water sample. Place one 10-mL aliquot of the sample into a 20-mL microbeaker. Place a second 10-mL aliquot into a second 20-mL beaker. Fortify one aliquot of the sample with bromate at a level approximating the native concentration of bromate in the untreated sample. This laboratory-fortified matrix (LFM) will indicate correct performance of the chlorite removal step. Acidify both aliquots with 33 µL of 0.5 N sulfuric acid solution and confirm the final pH (5–6) with pH test strips. Add 40 µL of ferrous iron solution, mix, and allow to react for 10 min. Filter the treated samples through a 0.45-micron nylon filter to remove precipitated ferric hydroxide. Then pass the solution through a hydronium-form, cation-exchange cartridge (Dionex OnGuard® H, P/N 39596) to remove excess soluble iron. The treated samples must be analyzed within 30 h.[11]

SYSTEM PREPARATION AND SETUP

Configure the IC with the AG9/AS9-HC columns and PCR system as depicted in Figure 1 and as described in the PC10 postcolumn delivery system installation instructions. Verify that the pump flow rate is within specifications and recalibrate if necessary. A GP50 should deliver water at 1.0 0.005 mL/min against a constant backpressure of 2000 psi. Verify that the UV absorbance detector wavelength accuracy is within specifications. Recalibrate if necessary. It is good practice to periodically record the visible lamp output (i.e., the reference cell current in nA) and elapsed time to assist in potential troubleshooting. Consult the pump and detector manuals for procedural details.

Install a 1-mL sample syringe and set the AS50 syringe speed to 4 or 5 to make fast large-loop injections. Install a calibrated 225-µL sample loop made from 111 cm of 0.02-in. i.d. PEEK tubing. Enter the correct sample "Loop Size" and "Sample Syringe Volume" in the AS50 Plumbing Configuration screen.

Prepare the AAES for use by hydrating the eluent chamber. Use a disposable plastic syringe to slowly push approximately 3 mL of DI water through both the "Eluent In" port and "Regen In" port. Allow the suppressor to sit for approximately 20 min to fully hydrate the suppressor monodisks and membranes. Because the effluent from the conductivity detector cell will undergo a postcolumn reaction, install the AAES in the external water mode by following the *Installation Instructions and Troubleshooting Guide for the Anion Atlas Electrolytic Suppressor* (Document No. 031770). Make sure that the pressure downstream from the Atlas suppressor does not exceed the recommended operating pressure of 20–100 psi. Use 0.02-in. i.d. PEEK tubing from the Atlas suppressor to the mixing tee, to the PCR coil, to the absorbance detector, and to waste, and keep it as short as is practical to minimize backpressure. Adjust the head pressure on the external water reservoir to deliver a flow rate of 5–10 mL/min (~10–15 psi). Use an AAES current of 78 mA.

Prepare the AMMS III (P/N 56750) for use by hydrating the eluent chamber. Use a disposable plastic syringe to slowly push approximately 3 mL of 0.2 N sulfuric acid through the "Eluent Out" port and 5 mL of 0.2 N sulfuric acid through the "Regen In" port. Allow the suppressor to sit for approximately 20 min to fully hydrate the suppressor screens and membranes. Install the AMMS III in the chemical regeneration mode by following the *Installation Instructions and Troubleshooting Guide for the Anion Micromembrane Suppressor* (Document No. 031727). Adjust the head pressure on the 0.3 N sulfuric acid reservoir to deliver a flow rate of 2–3 mL/min (~10–15 psi if a short piece of 0.01-in. i.d. PEEK tubing is connected to the AMMS III "Regen Out" port and trimmed accordingly).

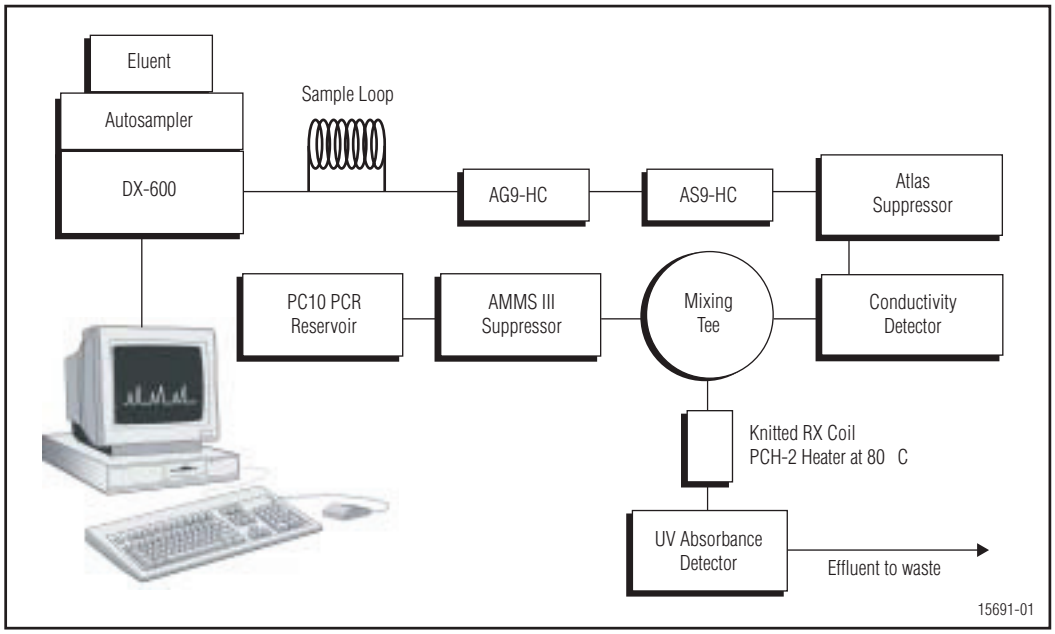

Figure 1. *IC system configuration for EPA Method 326.0.*

Pump the eluent at 1.3 mL/min and set the PC10 pneumatic pressure to 70 psi. To measure the PCR flow rate, collect the effluent from the detector (i.e., the total flow from the IC pump and PCR module) in a 10-mL graduated cylinder for 5 min. The PCR flow rate is the difference between the total flow rate and that of the IC pump. Adjust the air pressure of the PC10 postcolumn delivery module and remeasure the flow rate until the correct PCR flow rate of 0.4 mL/min is established. Variations in the PCR flow rate affect the postcolumn reaction time, pH, dilution, mixing rate, and ratio of the reactants. Stable day-to-day results depend on a well-controlled PCR flow rate. Confirm this flow rate on a daily basis and whenever detector response for a calibration check standard deviates beyond quality control acceptance criteria.

The storage solution 10 mM $NaHCO_3$ is shipped with the AS9-HC. After equilibrating the column with 9.0 mM carbonate eluent for 20 min, analyze a system blank of reagent water. An equilibrated system has a background conductance ~26 µS, with the peak-to-peak noise typically 1–2 nS per min. The background absorbance at 352 nm should be less than 200 mAU with peak-to-peak noise of less than 50 µAU per min. There should be no peaks eluting within the retention time window of the bromate anion. The column is equilibrated when two consecutive injections of a standard produce the same retention time for bromate.

RESULTS AND DISCUSSION

Figure 2 shows the chromatograms of a mixed anion standard containing 5 µg/L each of chlorite, bromate, bromide, and chlorate. The top trace (A) was obtained with the conductivity detector and the bottom trace (B) was obtained with the UV/Vis absorbance detector after postcolumn reaction with acidified KI. The bromate peak is baseline resolved from chlorite on both detector channels. However, the response on the absorbance detector after PCR with acidified KI is significantly enhanced compared to the response obtained on the conductivity detector.

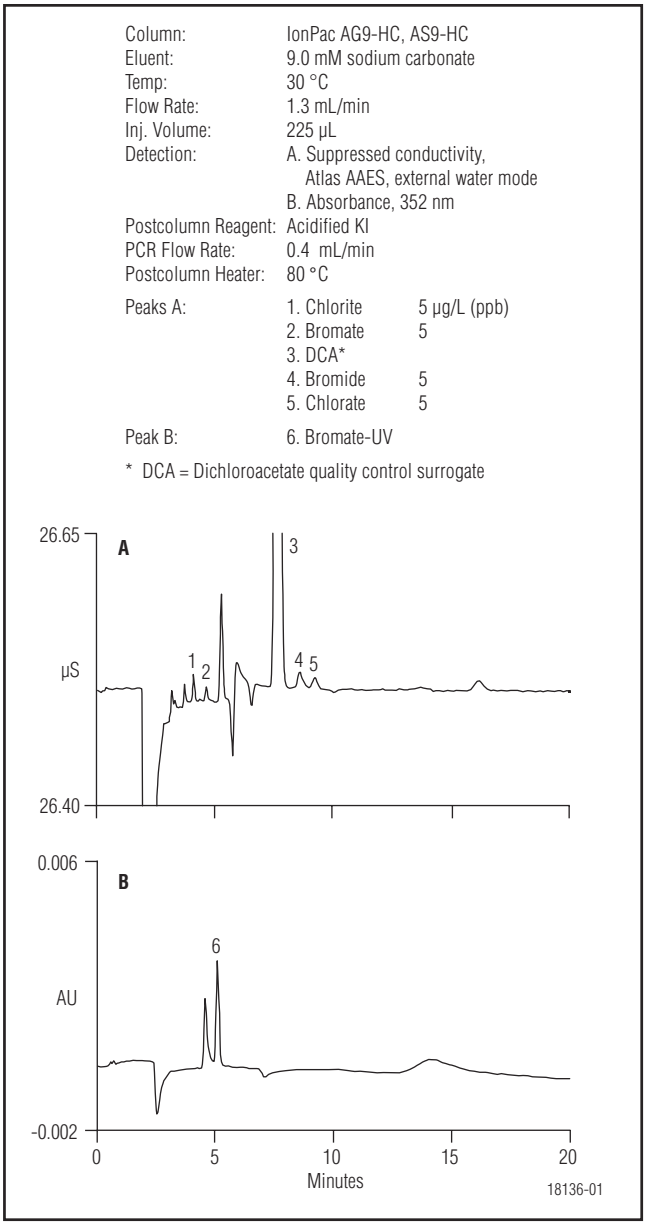

Figure 2. *Separation of a low-ppb inorganic anion standard using an IonPac AS9-HC column; (A) suppressed conductivity detection and (B) UV absorbance detection after PCR with acidified KI.*

Table 2 summarizes the calibration data and method detection limits (MDLs) obtained for the oxyhalide DBP anions and bromide using dual conductivity and UV detection. The MDL for each analyte was established by making eight replicate injections of a reagent

Table 2. Linear Ranges and MDLs for Oxyhalides and Bromide

Solute	Range (µg/L)	r^2	MDL Standard (µg/L)	Calculated MDL*(µg/L)
Chlorite	5.0–1000	0.9999	5.0	1.10
Bromate-conductivity	5.0–1000	0.9994	5.0	0.82
Bromide	5.0–1000	1.0000	5.0	1.10
Chlorate	5.0–1000	0.9999	5.0	0.85
Bromate-UV	0.5–15	0.9999	0.5	0.06

* The MDLs were calculated as MDL = *(t)* x (S) Where *t* = Student's *t* value for a 99% confidence level and a standard deviation estimate with n – 1 degrees of freedom (*t* = 3.00 for eight replicates of the MDL Standard), and S = standard deviation of the replicate analysis.

water blank fortified at a concentration of 3–5 times the estimated instrument detection limit.[12] The use of PCR addition and UV detection allows quantification of bromate down to 0.5 µg/L, without compromising the detection limits obtained with suppressed conductivity detection for the other anions of interest. Electronic smoothing (Olympic, 25 points, 5 sec, 1 iteration) of the UV signal was used to improve the calculated MDL for bromate.[13]

Figures 3–6 illustrate the method's performance for the determination of inorganic oxyhalide DBP anions and bromide in drinking water and bottled water samples. Figure 3 shows the chromatograms from a direct injection of drinking water (from Sunnyvale, CA). The top trace (A) was obtained with the conductivity detector and the bottom trace (B) was obtained with the UV/Vis absorbance detector after postcolumn reaction with acidified KI. Chlorite, bromate, bromide, and chlorate were all observed in the drinking water sample. The target analyte anions were well resolved from the sample matrix. The bromide was probably present in the source water. During ozonation, some of the bromide can convert to bromate. Chlorate can enter the water both as a source water contaminant and as a disinfection byproduct from the use of hypochlorite. Chlorite is a residual from treatment with chlorine dioxide.

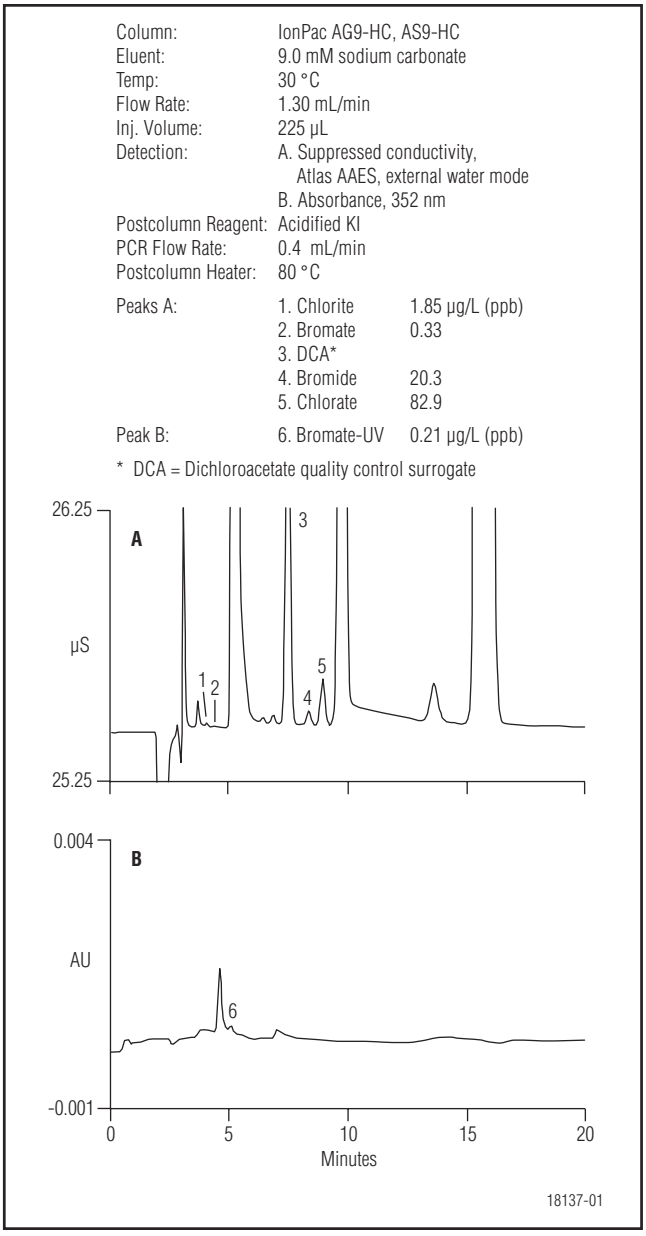

Figure 3. *Determination of DBP anions in Sunnyvale, CA drinking water; (A) suppressed conductivity detection and (B) UV absorbance detection after PCR with acidified KI.*

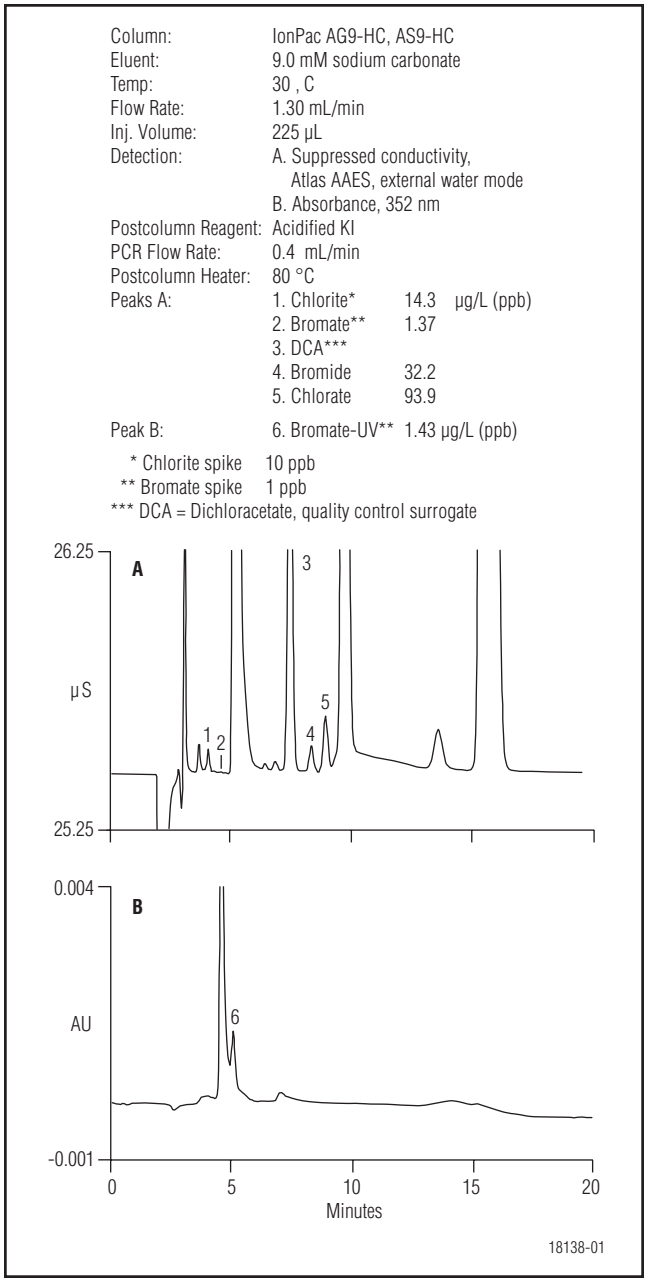

Figure 4. *Determination of DBP anions in spiked Sunnyvale, CA drinking water; (A) suppressed conductivity detection and (B) UV absorbance detection after PCR with acidified KI.*

Figure 4 shows chromatograms of the same drinking water sample spiked with bromate at 1 µg/L, and with chlorite, bromide, and chlorate at 10 µg/L. The top trace (A) was obtained with the conductivity detector and the bottom trace (B) was obtained with the UV/Vis absorbance detector after postcolumn reaction with acidified KI. The benefits of PCR with UV detection for bromate determination can clearly be seen in Figure 4 (B), where the bromate peak response is significantly enhanced compared to the conductivity detector. No response is observed for the large chloride peak that elutes immediately after bromate. Table 3 shows that quantitative recoveries were obtained for the oxyhalide anions and the bromide spiked into drinking water. In addition, quantitative recoveries were obtained for the oxyhalide anions and bromide spiked into the simulated high-ionic-strength water that contained elevated levels of the common matrix anions: chloride, carbonate, sulfate, nitrate, and phosphate. The use of PCR with UV/Vis detection allows the quantification of bromate down to 0.5 µg/L in the presence of 100 mg/L chloride (a 200,000 fold excess) with no sample pretreatment.

Table 3. Anion Recoveries for Spiked Water Samples

	Tap Water		High-Ionic-Strength Water	
Anion*	**Amount Added (µg/L)**	**Recovery**	**Amount Added (µg/L)**	**Recovery**
Chlorite	10	114%	100	97%
Bromate-conductivity	1	107%	10	98%
Bromide	10	98%	100	105%
Chlorate	10	113%	100	99%
Bromate-UV	1	124%	10	65%***
Bromate-UV**			1.0	106%

*Data were obtained from multianalyte spikes into Sunnyvale, CA tapwater and high-ionic-strength water (HIW) containing 100 mg/L chloride, 100 mg/L carbonate, 100 mg/L sulfate, 10 mg/L nitrate-N, and 10 mg/L phosphate-P.

** Bromate only (1.0 µg/L) was added to an HIW sample to determine low-level recovery for this anion using UV detection.

*** Bromate recovery was reduced by chlorite interference.

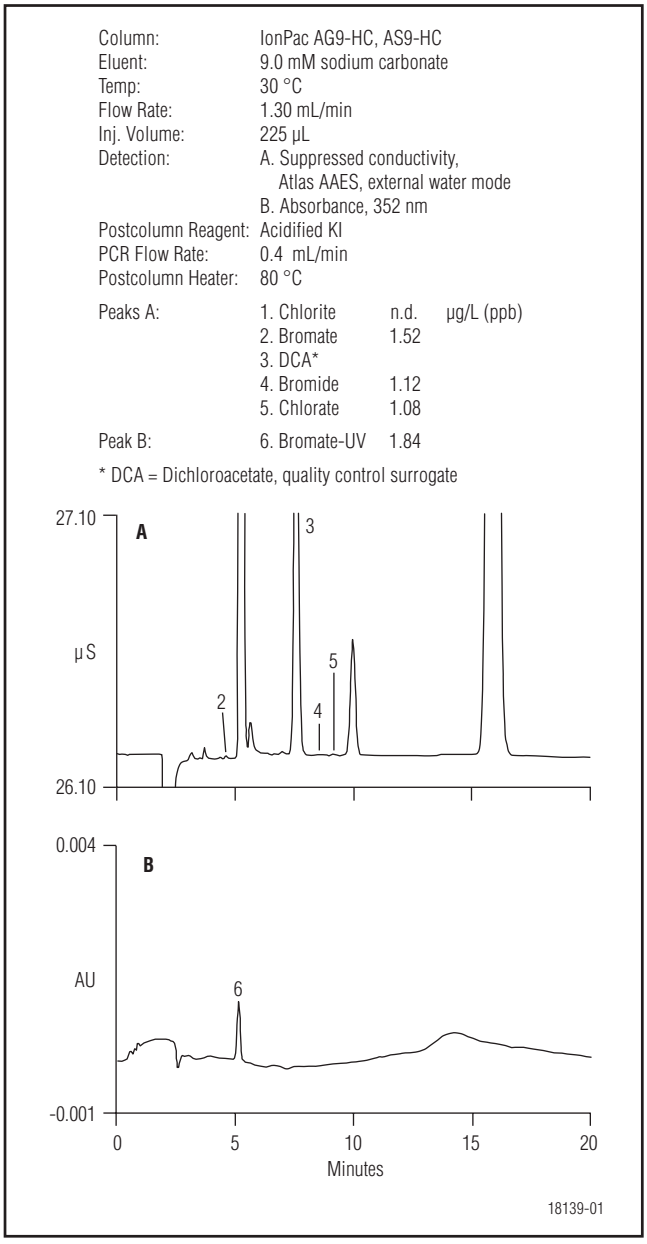

Figure 5. *Determination of DBP anions in bottled water; (A) suppressed conductivity detection and (B) UV absorbance detection after PCR with acidified KI.*

Figure 5 shows the chromatograms from a direct injection of bottled water. The top trace (A) was obtained with the conductivity detector, and the bottom trace (B) was obtained with the UV/Vis absorbance detector. The bottle label read: "Prepared using filtration, reverse osmosis, deionization, and ozonation". The DBP precursor bromide and the DBP bromate were both observed in the bottled water sample.

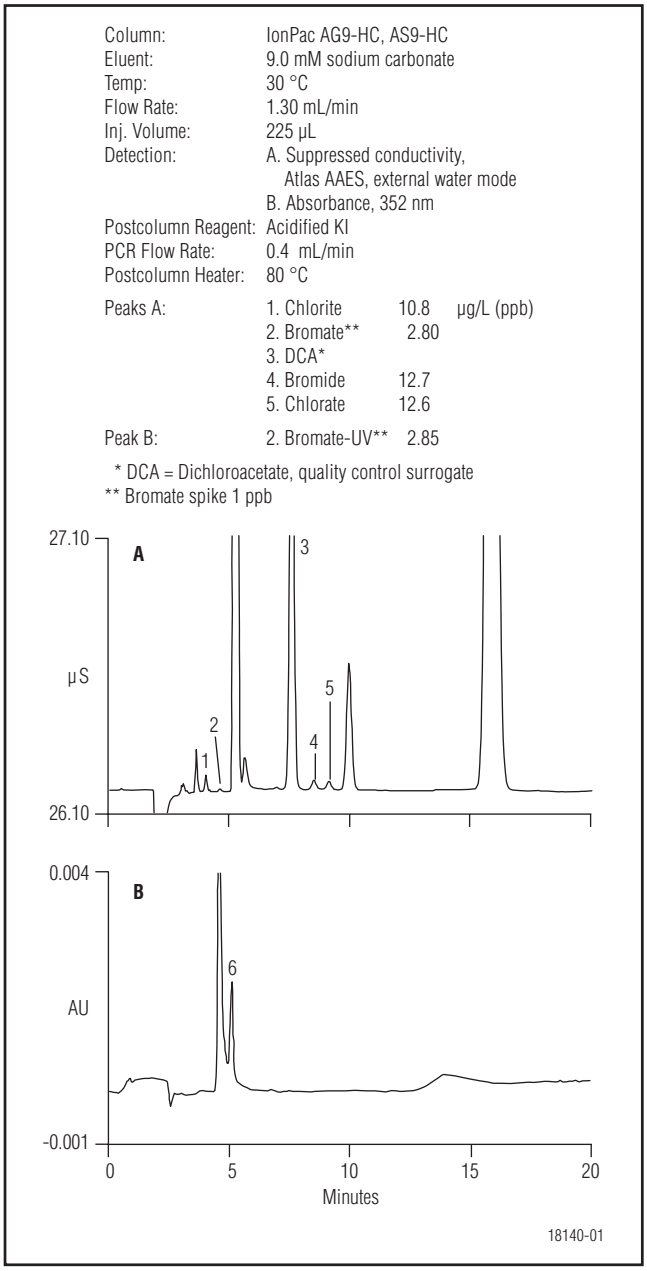

Figure 6. *Determination of DBP anions in spiked bottled water; (A) suppressed conductivity detection and (B) UV absorbance detection after PCR with acidified KI.*

Figure 6 shows the chromatograms of the same bottled water sample spiked with bromate at 1.0 µg/L, and with chlorite, bromide, and chlorate at 10 µg/L. The top trace (A) was obtained with the conductivity detector, and the bottom trace (B) was obtained with the UV/Vis absorbance detector after postcolumn reaction with acidified KI. Quantitative recoveries were obtained for all the added oxyhalide anions and bromide.

REMOVAL OF CHLORITE INTERFERENCE

When chlorine dioxide is used to disinfect drinking water, the DBP anion chlorite is found in the finished drinking water. Chlorite, like bromate, reacts with acidified KI and produces a response at 352 nm. High chlorite levels can interfere with quantification of bromate at low concentrations. The interference from chlorite can be minimized by reducing the chlorite with ferrous sulfate, as described in the "Sample Preparation" section. To evaluate the ferrous sulfate treatment, we analyzed a series of simulated chlorine dioxide-treated tap waters (STWs) spiked with varying levels of bromate. After determining the bromate level in each STW, we prepared the corresponding laboratory-fortified matrices (LFMs) by spiking each STW sample with an amount of bromate equal to 50–100% of the observed level. We then treated each STW and its corresponding LFM with ferrous sulfate and reanalyzed. The results, summarized in Table 4 and Figure 7, show that acceptable recoveries of bromate are obtained after such treatment. This treatment approach is recommended when analysis of low-level bromate is required in chlorine dioxide-treated drinking waters.

SUMMARY

The IC method described in this application note uses an IonPac AS9-HC column and suppressed conductivity detection, followed by postcolumn addition of acidified KI with UV detection, specifically for enhanced bromate response to determine all key oxyhalide anions and bromide at low-µg/L levels in drinking and bottled waters. The postcolumn addition and UV detection allows quantification of bromate at 0.5–15 µg/L without compromising the suppressed conductivity detection of chlorite, bromide, and chlorate. Conductivity detection is recommended for the quantification of bromate at 15–50 µg/L.

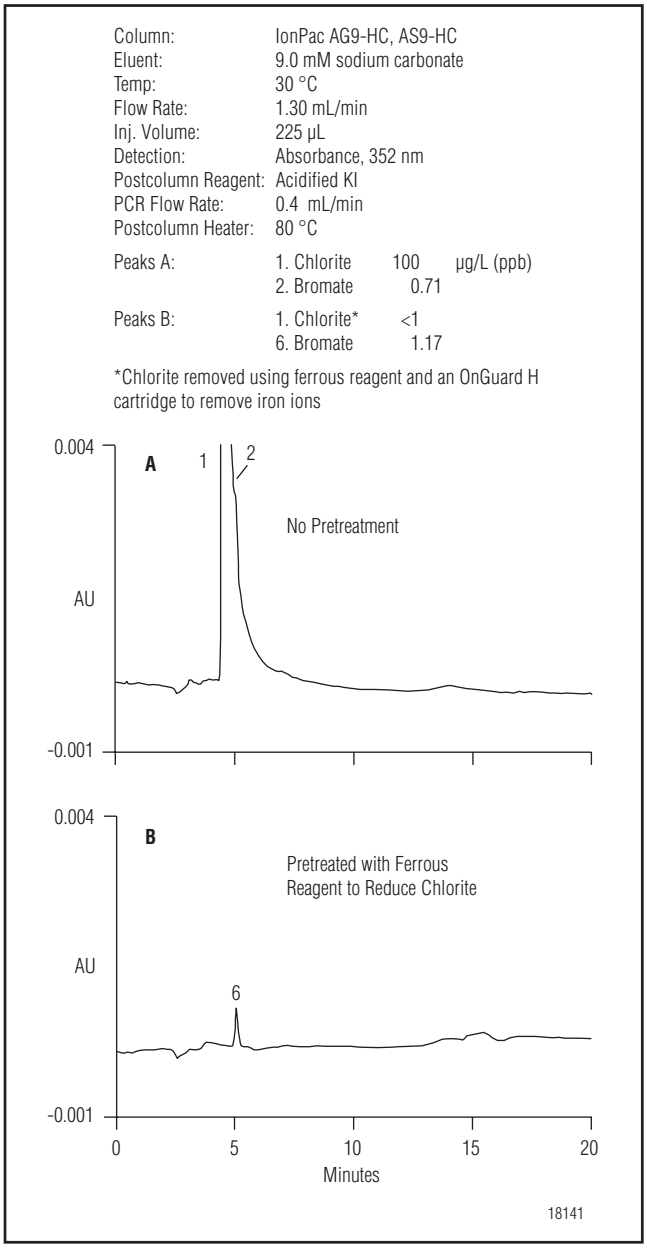

Figure 7. *Determination of DBP anions in simulated chlorine dioxide-treated water (STW). (A) Untreated STW, UV absorbance detection after PCR with acidified KI, and (B) STW after treatment with ferrous sulfate to remove chlorite, UV absorbance detection after PCR with acidified KI.*

Table 4. Bromate Recovery from Simulated Chlorine Dioxide-Treated Waters (STW)*						
	Spiked STW Fe (II) Treated			Laboratory Fortified Matrix Fe (II) Treated		
Sample	Amount Added (µg/L)	Amount Found (µg/L)	Recovery	Amount Added (µg/L)	Amount Found (µg/L)	Recovery
STW	0	0.19		0.5	0.61	84%
STW-1	0.5	0.70	102%	0.5	1.20	100%
STW-2	1.0	1.17	98%	1.0	2.24	107%
STW-3	2.0	2.18	100%	2.0	4.33	108%
STW-4	5.0	5.22	101%	5.0	10.24	100%

* Chlorite present at 100 g/L.

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13. Schibler, J. A. *Am. Lab.* **1997,** 63.
14. Method 4500-C102.C. In *Standard Methods for the Examination of Water and Wastewater,* 18th Ed.; Greenberg, A. E.; Clesceri, L. S.; Eaton, A. D. (Eds.); APHA: Washington, DC, 1992.

SUPPLIERS

Aldrich Chemical Co., P.O. Box 2060, Milwaukee, WI 53201 USA, Tel: 800-558-9160, www.aldrich.sial.com.

Alfa Products, 30 Bond St., Ward Hill, MA 01835 USA, Tel.: 800-343-0660, info@alfa.com.

EM Science, P.O. Box 70, Gibbstown, NJ 08027 USA, Tel: 800-222-0342, www.emscience.com.

Fisher Scientific, 2000 Park Lane, Pittsburgh, PA 15275-1126 USA, Tel: 800-766-7000, www.fishersci.com.

Fluka, Box 2060, Milwaukee, WI 53201 USA, Tel: 800-558-9160, www.sigma-aldrich.com.

J. T. Baker, 222 Red School Lane, Phillipsburg, NJ 08865 USA. Tel.: 800-582-2537, www.jtbaker.com (order from VWR).

Sigma Chemical Co., P.O. Box 14508, St. Louis, MO 63178 USA, Tel: 800-325-3010, www.sigma-aldrich.com.

SPEX CertiPrep, Inc., 203 Norcross Ave., Metuchen, NJ 08840 USA, Tel.: 800-LAB-SPEX, www.spexcsp.com (order from Fisher).

ULTRA Scientific (order from VWR).

VWR Scientific Products, 3745 Bayshore Blvd., Brisbane, CA 94005, USA, Tel.: 800-932-5000, www.vwrsp.com.

Determination of Inorganic Oxyhalide Disinfection Byproduct Anions and Bromide in Drinking Water Using Ion Chromatography with the Addition of a Postcolumn Reagent for Trace Bromate Analysis

INTRODUCTION

The chlorination of drinking water can produce trihalomethanes and other suspected carcinogenic disinfection byproducts (DBPs) that endanger human health.[1] Unfortunately, common alternatives to chlorination also can produce harmful DBPs. The use of chlorine dioxide for the disinfection of drinking water generates the inorganic oxyhalide DBPs chlorite and chlorate, and the presence of chlorate has been reported in waters treated with hypochlorite.[2] Ozonation, an increasingly prevalent and effective disinfection technique, produces bromate as a DBP anion if the source water contains naturally occurring bromide.[3] Bromate has been judged by both the World Health Organization (WHO) and the U.S. Environmental Protection Agency (EPA) as a potential carcinogen, even at very low µg/L levels. The U.S. EPA has estimated a potential cancer risk equivalent to 1 in 10^4 for a lifetime exposure to drinking water containing bromate at 5 µg/L.[4]

The U.S. EPA has recently issued new rules that require public water supplies to control previously unregulated microbes (e.g., cryptosporidium and giardia) and cancer-causing DBPs in finished drinking water. The Stage 1 D/DBP Rule specifies a Maximum Contaminant Level (MCL) for bromate of 10 µg/L and an MCL for chlorite of 1000 µg/L.[5] The EPA intends to convene Stage 2 of the D/DBP Rule in the near future, while both Germany and Japan are considering regulatory limits for inorganic DBPs.[6]

The recent efforts by global regulatory agencies to monitor levels and establish regulatory limits has generated considerable interest in the development of improved analytical methods for the determination of trace level inorganic oxyhalide DBPs. The determination of bromate and other inorganic DBPs traditionally has been accomplished by ion chromatography (IC) using an IonPac® AS9-SC anion-exchange column with a carbonate/bicarbonate eluent and suppressed conductivity detection, as described in U.S. EPA Method 300.0 (B).[7] EPA Method 300.1 was published as an update to Method 300.0 in 1997. Method 300.1 specifies the use of an IonPac AS9-HC column and suppressed conductivity detection for the determination of bromate, bromide, chlorite, and chlorate at low µg/L levels by direct injection.[8] The detection limit for bromate determined by IC with suppressed conductivity detection can be further reduced to 1 µg/L by using preconcentration after appropriate sample cleanup.[1]

Postcolumn derivatization can also be used to improve detection limits when using IC for inorganic DBP analysis. The use of IC with dual postcolumn addition of hydrochloric acid and then chlorpromazine can achieve a method detection limit (MDL) for bromate of 0.49 µg/L.[9] Iodate, chlorite, and bromate have been detected by using a postcolumn reaction with excess bromide under acidic conditions. The tribromide ion formed can be detected spectrophotometrically at

267 nm, allowing an MDL of less than 0.5 µg/L for bromate with a large-volume injection.[10] Sub-µg/L MDLs for bromate have also been reported by workers using other postcolumn reagents, such as fuchsin or excess iodide under acidic conditions.[11,12] In addition to postcolumn reaction (PCR) methods, electrospray tandem mass spectrometry (MS-MS) and inductively coupled plasma mass spectrometry (ICP-MS) have been used as specific detection techniques for the ion chromatographic analysis of bromate. The use of electrospray MS-MS detection can achieve an MDL for bromate of approximately 0.1 µg/L; the use of ICP-MS detection has been reported to permit an MDL for bromate of 0.8 µg/L.[13,14]

This Application Note describes an improved IC method to quantify low levels of oxyhalide DBP anions and bromide in reagent water, bottled water, and finished drinking water. The method uses an IonPac AS9-HC column and suppressed conductivity detection, followed by postcolumn addition of *o*-dianisidine (ODA) to enhance visible absorbance detection of the bromate ion. This method allows quantification of all the key oxyhalide anions and bromide at low µg/L levels by using conductivity detection, and the postcolumn addition of ODA followed by visible detection allows quantification of bromate down to 0.5 µg/L. This method requires only a single postcolumn reagent delivered pneumatically with conventional postcolumn instrumentation.[2] The approach described in this Application Note is technically equivalent to that described in U.S. EPA Method 317.0 titled "Determination of Inorganic Oxyhalide Disinfection By-Products in Drinking Water Using Ion Chromatography with the Addition of a Postcolumn Reagent for Trace Bromate Analysis".[15]

EQUIPMENT

Dionex DX-500 ion chromatographic system consisting of:
- GP50 Gradient Pump with vacuum degas option
- ED40 Conductivity Detector with DS3 Detector Cell
- AD20 UV/Vis Absorbance Detector with 10-mm cell
- AS50 Autosampler

PC10 Pneumatic Postcolumn Delivery Module (P/N 50601)

PCH-2 Postcolumn Reaction Heater (P/N 39348)

Knitted Reaction Coil, 500 µL, potted (for PCH-2) (P/N 39349)

Two 4-L plastic bottle assemblies (for external water mode suppression)

PeakNet® 5.1 Chromatography Workstation

REAGENTS AND STANDARDS

Deionized water, Type I reagent grade, 18 MΩ-cm resistivity or better

0.5 M Carbonate Anion Eluent Concentrate (Dionex P/N 37162)

o-Dianisidine, dihydrochloride salt (ODA; Sigma D-3252)

Iron (II) sulfate heptahydrate ($Fe_2SO_4 \cdot 7H_2O$; Aldrich 21,542-2)

Ethylenediamine (EDA; Sigma E-1521)

Nitric acid, (70%; J.T. Baker INSTRA-ANALYZED 9598-00)

Methanol (spectrophotometric grade; Sigma M-3641)

Potassium bromide (KBr; J.T. Baker 2998)

Sodium bromide (NaBr; Aldrich 31,050-6)

Sodium bromate ($NaBrO_3$; EM SX 03785-1)

Sodium chlorate ($NaClO_3$; Fluka 71370)

Sodium chlorite ($NaClO_2$; Fluka 71388, ~80% pure)

Bromate standard, 1000 mg/L, $NaBrO_3$ in H_2O (SPEX CertiPrep AS-BRO39-2Y)

Bromide standard, 1000 mg/L, NaBr in H_2O (SPEX CertiPrep AS-BR9-2Y)

Chlorate standard, 1000 mg/L, $NaClO_3$ in H_2O (SPEX CertiPrep AS-CLO39-2Y)

Chlorite standard, 1000 mg/L, $NaClO_2$ in H_2O (SPEX CertiPrep AS-CLO29-2Y)

CONDITIONS

Columns:	Dionex AG9-HC, 50 x 4 mm ID guard column (P/N 51791) Dionex AS9-HC, 250 x 4 mm ID analytical column (P/N 51786)
Eluent:	9.0 mM Sodium carbonate (Na_2CO_3)
Flow Rate:	1.3 mL/min
Temperature:	Ambient
Sample Volume:	225 µL
Detection:	Suppressed conductivity: ASRS®-ULTRA (P/N 53946), AutoSuppression® external water mode, 100 mA current, DS3 Cell (P/N 44130), 35 °C, 1.7%/°C.
Background Conductance:	~24 µS
System Backpressure:	~2300 psi
Run Time:	25 min

PCR

Detection:	Absorbance at 450 nm (tungsten lamp)
Postcolumn Reagent Flow:	0.7 mL/min
Postcolumn Heater Temp.:	60 °C

PREPARATION OF SOLUTIONS AND REAGENTS

Reagent Water

Distilled or deionized water, 18 MΩ-cm or better, free of the anions of interest and filtered through a 0.2-µm filter.

Eluent Solution (9 mM Sodium Carbonate)

Dilute 18 mL of 0.5 M sodium carbonate concentrate to 1 L with deionized water. Unless the in-line degas option is being used, sparge eluent prior to use with helium or sonicate under vacuum for 10 min.

Postcolumn Reagent

Add 40 mL of 70% nitric acid to about 300 mL reagent water in a 500-mL volumetric flask. Add 2.5 g KBr and stir to dissolve. Dissolve 250 mg of *o*-dianisidine • 2 HCl in 100 mL methanol and add to the nitric acid/KBr solution. Bring to volume with reagent water. Prepare in advance, set aside overnight until the slight champagne color fades, and filter through a 0.45-µm filter. Discard any PCR reagent that is not colorless or nearly colorless after sitting overnight. The reagent is stable for one month when stored at room temperature.

Stock Standard Solutions

Purchase certified solutions or prepare stock standard solutions by dissolving the corresponding mass of the salt for each of the anions of interest (see Table 1) in reagent water and dilute to 100 mL.

Table 1 Masses of Compounds Used to Prepare 100 mL of 1000 mg/L Anion Standards

Anion	Compound	Mass (g)
BrO_3^-	Sodium bromate ($NaBrO_3$)	0.1180
Br^-	Sodium bromide (NaBr)	0.1288
ClO_3^-	Sodium chlorate ($NaClO_3$)	0.1275
ClO_2^-	Sodium chlorite ($NaClO_2$)	0.1680*

*Because sodium chlorite is usually available only as an 80% technical grade salt, the 80% purity is accounted for in the 0.1680 g mass cited above. If an alternate purity is used, make an appropriate adjustment in the mass of salt used after determining the exact percentage of $NaClO_2$, which can be done using an iodometric titration procedure.[16]

Prepare a mixed anion calibration stock standard at 20 mg/L by combining 2 mL of each of the bromide, chlorite, and chlorate stock standards in a 100-mL volumetric flask. Mix and bring to volume with reagent water. These standards are stable for at least 1 month when stored at < 6 ºC.

Because bromate decomposes in the presence of chlorite, prepare a bromate-only calibration stock standard at 5 mg/L by adding 0.5 mL of the bromate stock standard to a 100-mL volumetric flask and bringing to volume with reagent water. This standard is stable for 2 weeks when stored at < 6 °C.

Working Standard Solutions

Use reagent water to prepare appropriate dilutions of the calibration stock standards as needed.

Ethylenediamine (EDA) Preservative Solution

Dilute 2.8 mL of ethylenediamine (99%) to 25 mL with reagent water. Prepare fresh monthly.

Ferrous Iron Solution [1000 mg/L Fe (II)]

Add 6 µL concentrated nitric acid to about 15 mL reagent water in a 25 mL volumetric flask. Add 0.124 g ferrous sulfate heptahydrate ($FeSO_4 \cdot 7H_2O$), dissolve, and bring to volume with reagent water (final pH ~ 2). Prepare fresh every 2 days.

Sulfuric Acid Solution (0.5 N)

Dilute 1.4 mL of concentrated sulfuric acid to 100 mL with reagent water.

SAMPLE PREPARATION

When taking a sample from a treatment plant that uses chlorine dioxide or ozone, the sample must be sparged immediately with an inert gas (e.g., nitrogen, argon, or helium) for 5 min. Add 1.00 mL of EDA Preservative Solution per 1.0 L of sample to prevent conversion of residual hypochlorite or hypobromite to chlorate or bromate. This also prevents metal-catalyzed conversion of chlorite to chlorate. The samples preserved in this manner are stable for at least 14 days when stored in amber glass bottles at 4 °C.[17]

After appropriate preservation, most samples can be filtered through a 0.45-µm filter and directly injected onto the ion chromatograph. However, each sample that contains excess chlorite must be treated to remove chlorite and then reanalyzed for bromate, because

elevated levels of chlorite can interfere with the quantification of bromate by PCR.

The treatment procedure to remove chlorite requires two portions of sample. Place two 10-mL aliquots of the sample into separate 20-mL beakers. Fortify one aliquot with bromate at a level approximating the native concentration of bromate in the untreated sample. This laboratory fortified matrix (LFM) will indicate correct performance of the chlorite removal step. Acidify both aliquots with 33 µL of sulfuric acid reagent and confirm the final pH (5–6) with pH test strips. Add 40 µL of ferrous iron solution, mix, and allow to react for 10 min. Filter the treated samples through a 0.45-µm nylon filter to remove precipitated ferric hydroxide, and then pass the solution through a hydronium form cation-exchange cartridge (Dionex OnGuard®-H, P/N 39596) to remove excess soluble iron. The treated samples must be analyzed within 30 h.

SYSTEM PREPARATION AND SET-UP

Configure the IC with the PCR system as depicted in Figure 1. Determine the PCR flow rate by collecting the combined effluent from the IC pump and the PCR module in a 10-mL graduated cylinder for 1 min. The PCR flow rate is the difference between the total flow rate and that of the IC pump. Adjust the air pressure of the postcolumn delivery module (PC10) and remeasure the flow rate until the correct flow rate of 0.7 mL/min is established. Confirm this flow rate on a weekly basis or whenever detector response for a calibration check standard deviates beyond quality control acceptance criteria.

To determine target anions at trace concentrations, it is essential to have low baseline noise. Minimize baseline noise by taking the following steps during system set-up. Install the ASRS-ULTRA in the external water mode rather than the recycle mode. Prior to sample analysis, determine a system blank by analyzing 225 µL of deionized water using the method described above. An equilibrated system has a background conductance of ~ 24 µS, peak-to-peak noise of ~ 5 nS per minute, and no peaks eluting at the same retention time as the anions of interest.

RESULTS AND DISCUSSION

Figure 2 shows the chromatograms of a mixed anion standard containing 10 µg/L bromate and 15 µg/L each of chlorite, bromide, and chlorate obtained by using dual A) suppressed conductivity and B) UV/Vis absorbance after postcolumn reaction with ODA. The bromate peak is baseline-resolved from chlorite on both detector channels; however, it shows a significantly enhanced

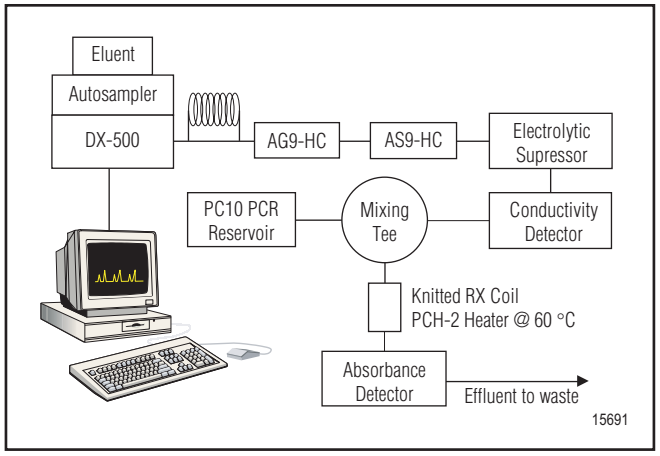

Figure 1. IC system configuration for EPA Method 317.0.

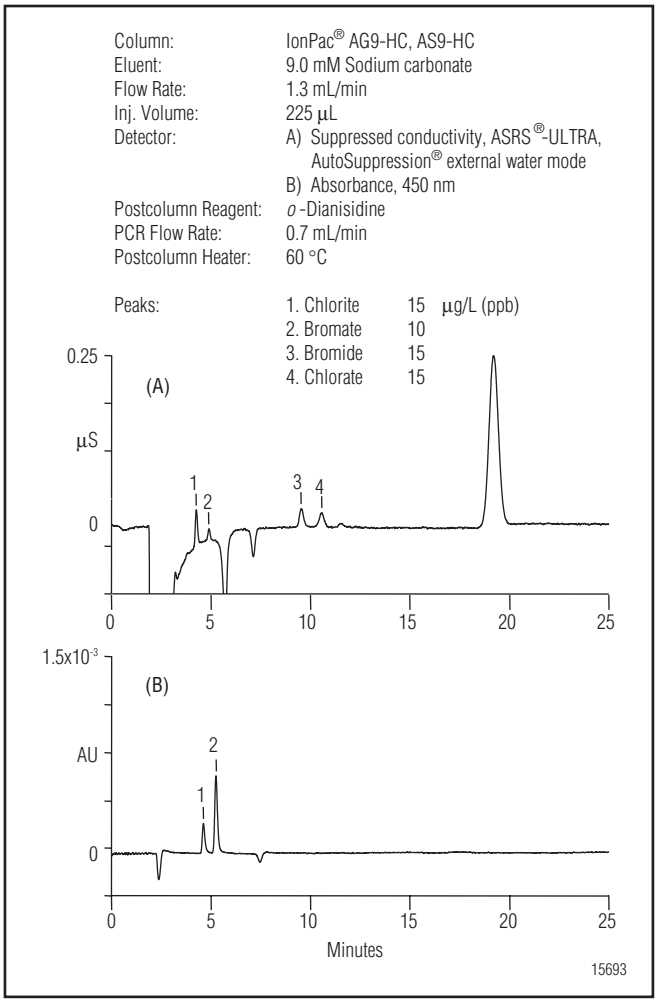

Figure 2. Separation of a low-ppb DBP anion standard using an IonPac AS9-HC column: A) suppressed conductivity detection and B) visible absorbance detection after PCR with o-dianisidine.

response on the absorbance detector after PCR with ODA compared to the response obtained on the conductivity detector.

Table 2 summarizes the calibration data and method detection limits (MDLs) obtained for the oxyhalide DBP anions and bromide using dual conductivity and PCR detection. The MDL for each analyte was established by making seven replicate injections of a reagent water blank fortified at a concentration of 3 to 5 times the estimated instrument detection limit.[2] The use of PCR addition and UV/Vis detection allows quantification of bromate down to 0.5 µg/L without compromising detection limits obtained with suppressed conductivity detection for the other anions of interest.[6] Note that the use of electronic smoothing (Olympic, 25 points, 5 sec, 1 iteration) of the UV/Vis signal improves the calculated MDL for bromate.[2] Figure 3 demonstrates the effect of smoothing on the performance of the PCR detection for a 1.0 µg/L bromate standard. No significant loss of peak response is observed after smoothing, although baseline noise is reduced by a factor of approximately 2x, which results in a similar improvement in the detection limit (Table 2).

Table 2 Linear Ranges and MDLs for Oxyhalides and Bromide

Solute	Range (µg/L)	r^2	MDL Standard (µg/L)	Calculated MDL* (µg/L)
Chlorite	5.0–500	0.9999	5.0	1.80
Bromate–conductivity	5.0–50	0.9986	5.0	1.22
Bromide	5.0–500	0.9999	5.0	1.90
Chlorate	5.0–500	0.9999	5.0	1.85
Bromate–UV/Vis (smoothed)	0.5–15	0.9986	1.0	0.09
Bromate–UV/Vis (no smoothing)	0.5–15	0.9986	1.0	0.19

*MDL = (*t*) x (S) Where *t* = student's *t* value for a 99% confidence level and a standard deviation estimate with n - 1 degrees of freedom [*t* = 3.14 for seven replicates of the MDL standard], and S = standard deviation of the replicate analysis.

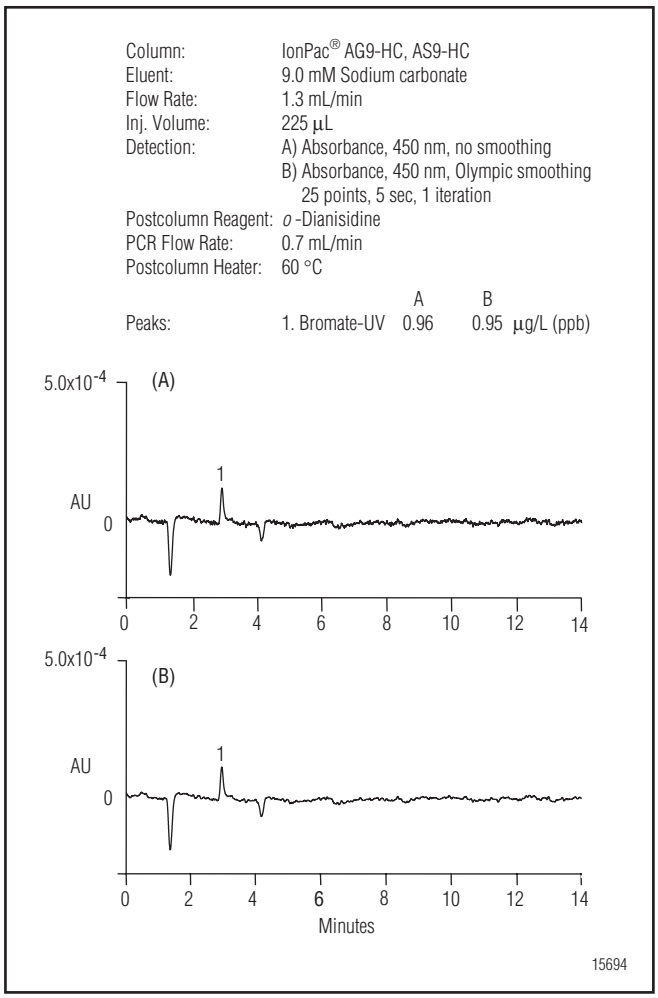

Figure 3. Effect of smoothing on bromate determination: A) unsmoothed data and B) smoothed data (Olympic, 25 points, 5 sec, 1 iteration).

Figures 4–7 illustrate the performance of the method for the determination of inorganic oxyhalide DBP anions and bromide in drinking and bottled water samples. Figure 4 shows the chromatograms from a direct injection of drinking water (from Sunnyvale, California) obtained by using dual A) suppressed conductivity and B) UV/Vis absorbance after postcolumn reaction with ODA. Neither chlorite nor bromate are observed in the drinking water sample; however, bromide and chlorate (frequently observed as a disinfection byproduct from the use of hypochlorite) are well resolved from the sample matrix.

Figure 5 shows the chromatograms of the same drinking water sample spiked with chlorite, bromate, bromide, and chlorate at levels of 108, 11.3, 36, and 72 µg/L, respectively. The chromatograms were obtained using, in series, dual A) suppressed conductivity and B) UV/Vis absorbance after postcolumn reaction with ODA. Quantitative recoveries were obtained for all anions, as shown in Table 3. The benefits of PCR with UV/Vis detection for bromate determination can clearly be seen in Figure 5B: bromate peak response is significantly enhanced compared to the response on the conductivity detector and no response is observed for the large peak from about 20 µg/L chloride that elutes immediately after bromate. The use of PCR with UV/Vis detection allows the quantification of bromate down to 0.5 µg/L in the presence of 200 mg/L chloride (a 400,000-fold excess) with no sample pretreatment.

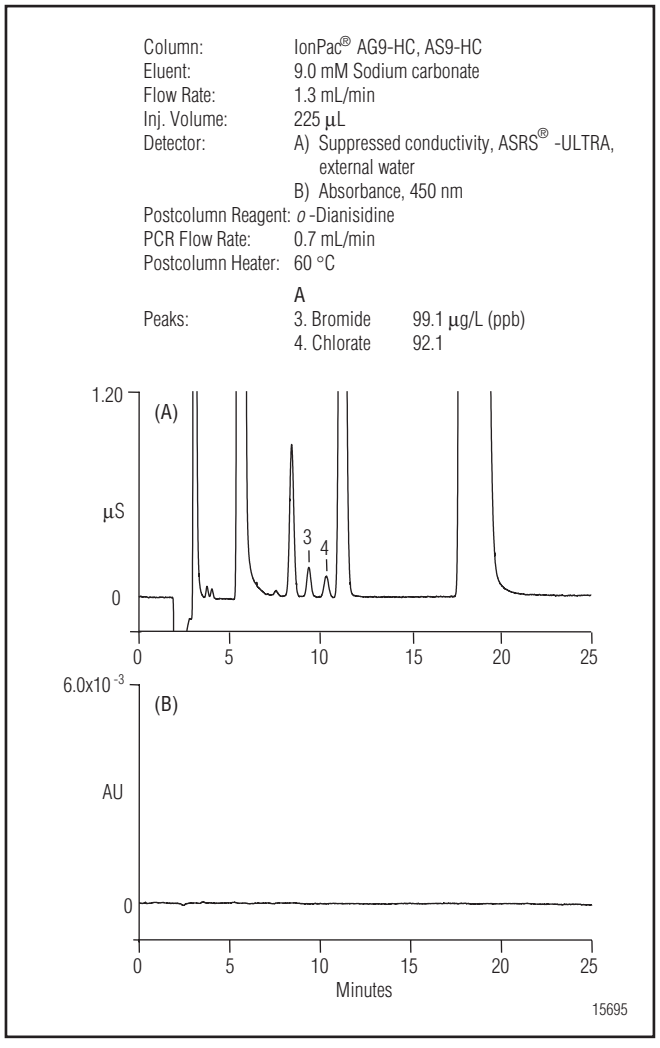

Figure 4. Determination of DBP anions in tap water: A) suppressed conductivity detection and B) visible absorbance detection after PCR with o*-dianisidine.*

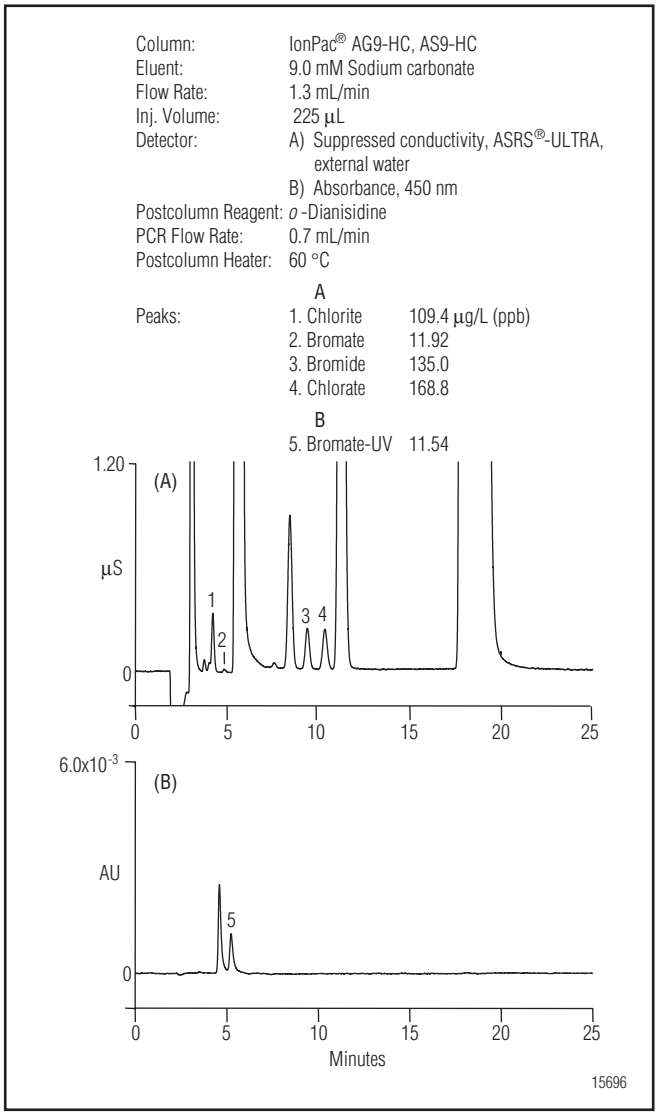

Figure 5. Determination of DBP anions in spiked tap water: A) suppressed conductivity detection and B) visible absorbance detection after PCR with o*-dianisidine.*

Figure 6 shows the chromatograms from a direct injection of bottled spring water obtained using, in series, dual A) suppressed conductivity and B) UV/Vis absorbance after postcolumn reaction with ODA. In this instance, both bromate and bromide are observed in the bottled water sample. Bromate, which is formed during ozonation of source water containing bromide, is present at about 2 µg/L and can clearly be seen in the UV/Vis chromatogram, although no peak is evident on the conductivity detector. Figure 7 shows the chromatograms of the same bottled water sample spiked with chlorite, bromate, bromide, and chlorate at levels of 126, 13.2, 42, and 84 µg/L, respectively. These chromatograms were obtained by using, in series, dual A) suppressed conductivity and B) UV/Vis absorbance after postcolumn reaction with ODA. Table 3 shows that quantitative recoveries were again obtained for all anions. Table 3 also shows the recoveries obtained for bromate spiked into the same drinking and bottled water samples at a lower concentration of 2.2 µg/L when using UV/Vis absorbance after postcolumn reaction with ODA. This method permits quantitative recoveries (80–120%) for bromate at levels down to 1 µg/L when using PCR and UV/Vis detection.

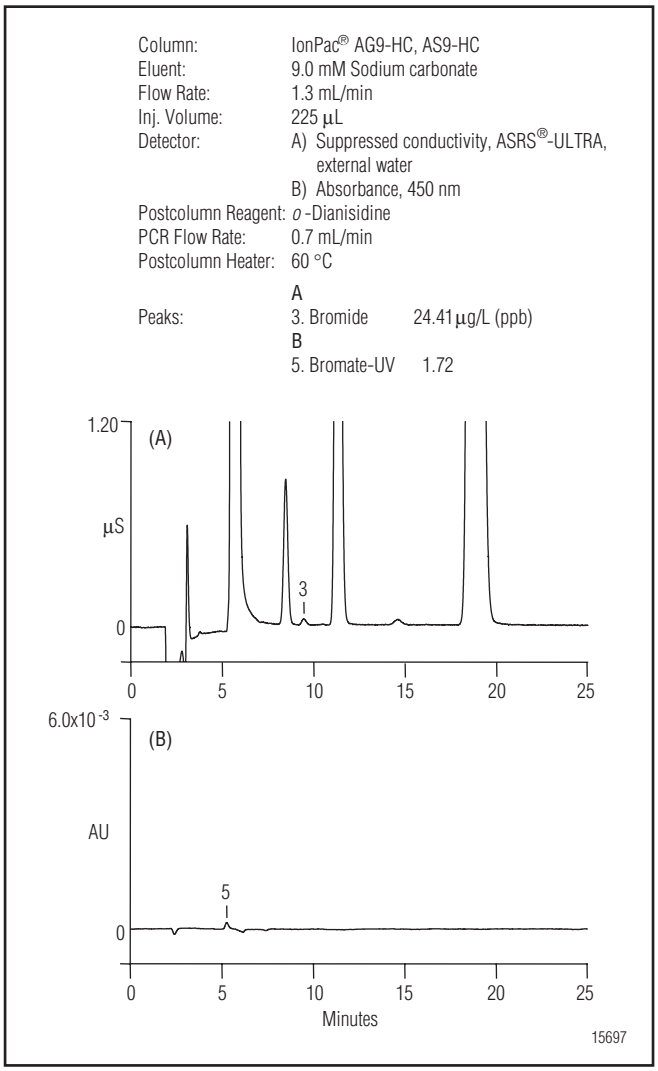

Figure 6. Determination of DBP anions in bottled water: A) suppressed conductivity detection and B) visible absorbance detection after PCR with o*-dianisidine.*

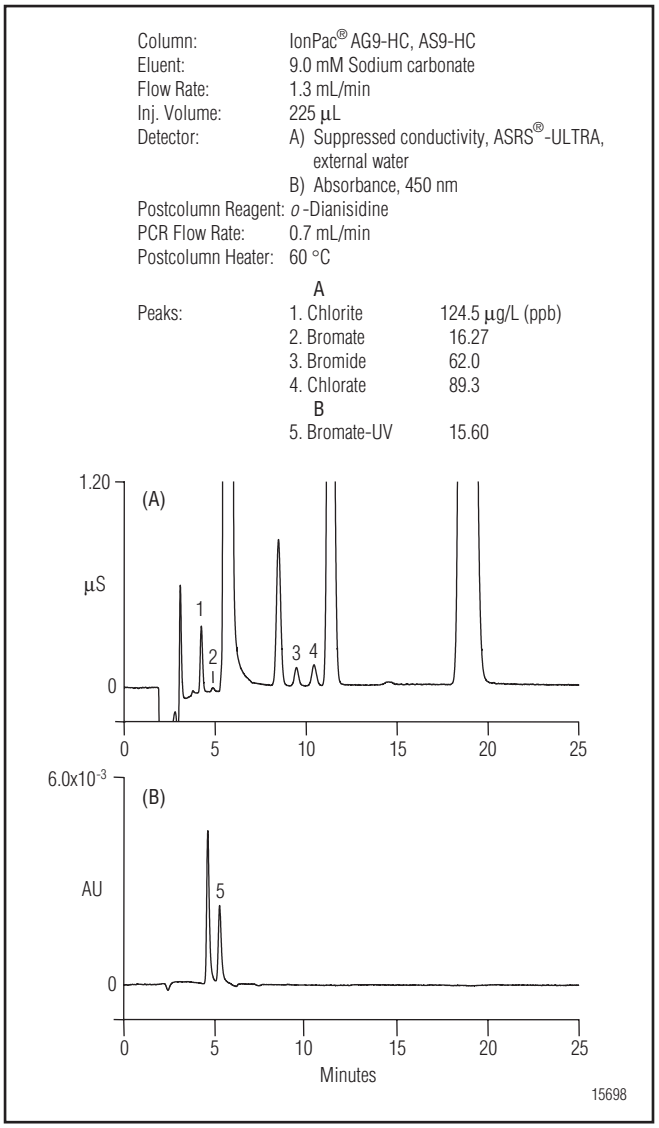

Figure 7. Determination of DBP anions in spiked bottled water: A) suppressed conductivity detection and B) visible absorbance detection after PCR with o*-dianisidine.*

Table 3 Anion Recoveries for Spiked Water Samples

	Tap Water		Bottled Water	
Anion*	**Amount Added (μg/L)**	**Recovery**	**Amount Added (μg/L)**	**Recovery**
Chlorite	108	104%	126	104%
Bromate–conductivity	11.3	105%	13.2	105%
Bromide	36.0	100%	42	100%
Chlorate	72	107%	84	107%
Bromate–UV/Vis	11.3	102%	13.2	102%
Bromate–UV/Vis**	2.2	91%	2.2	96%

*Data were obtained from multi-analyte spikes into tap and bottled water samples.

**Bromate only (2.2 μg/L) was added to tap and bottled water samples to determine low level recovery for this anion using UV/Vis detection.

Table 4 Bromate Recovery from Simulated Chlorine Dioxide Treated Waters (STW)*

	Spiked STW Fe (II)-Treated		Laboratory Fortified Matrix Fe (II)-Treated	
	Amount Added (μg/L)	**Recovery**	**Amount Added (μg/L)**	**Recovery**
STW	0	ND	2.0	90%
STW-1	1.75	74%	2.0	78%
STW-2	2.15	80%	4.0	75%
STW-3	4.61	76%	6.0	82%
STW-4	5.14	80%	8.0	75%

* Chlorite present at 100 μg/L

REMOVAL OF CHLORITE INTERFERENCE

When chlorine dioxide is used to disinfect drinking water, the DBP anion chlorite is found in the finished drinking water. Chlorite, like bromate, reacts with *o*-dianisidine to form a complex that absorbs at 450 nm. High chlorite levels can interfere with quantification of low-level bromate.[2] One approach to minimize the interference from chlorite is to remove the chlorite by reduction with ferrous sulfate, as described in the "Sample Preparation" section. This treatment was evaluated by applying it to a series of simulated chlorine dioxide-treated tap waters, which had been spiked with varying levels of bromate, and the corresponding LFMs. The results, summarized in Table 4, show that acceptable recoveries of bromate are obtained after such treatment. This treatment approach is recommended when analysis of low-level bromate is required in chlorine dioxide-treated drinking waters.

SUMMARY

The IC method described in this Application Note, which uses an IonPac AS9-HC column and suppressed conductivity detection, followed by postcolumn addition of *o*-dianisidine with UV/Vis detection specifically to enhance bromate response, allows the determination of all the key oxyhalide anions and bromide at low μg/L levels in drinking and bottled waters. The use of postcolumn addition and UV/Vis detection allows the quantification of bromate in the range of 0.5-15 μg/L without compromising the suppressed conductivity detection of chlorite, bromide, and chlorate. Conductivity detection is recommended for the quantification of bromate in the range of 15–50 μg/L.

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17. Hautman, D.P.; Bolyard, M. *J. Chromatogr.* **1992,** *602,* 65.

SUPPLIERS

Aldrich Chemical Co., P.O. Box 2060, Milwaukee, WI 53201, USA. Tel: 800-558-9160. www.sigma-aldrich.com.

Fluka, P.O. Box 2060, Milwaukee, WI 53201, USA. Tel: 800-558-9160. www.sigma-aldrich.com.

Pierce Chemical Co., 3747 North Meridian Road, P.O. Box 117, Rockford, IL 61105, USA. Tel: 800-874-3723. www.piercenet.com.

Sigma Chemical Co., P.O. Box 14508, St. Louis, MO 63178, USA. Tel: 800-325-3010. www.sigma-aldrich.com.

SPEX CertiPrep, Inc., 203 Norcross Ave., Metuchen, NJ 08840, USA. Tel.:800-522-7739. www.spexcsp.com.

VWR Scientific Products, 3745 Bayshore Blvd., Brisbane, CA 94005, USA. Tel: 800-932-5000. www.vwrsp.com.

Trace Level Determination of Bromate in Ozonated Drinking Water Using Ion Chromatography

INTRODUCTION

During the 1970s it was discovered that the chlorination of drinking water produced carcinogens, such as the trihalomethanes. Since then environmental regulatory agencies, as well as drinking water treatment technologists, have been aggressively researching alternative disinfection methods that minimize the production of byproducts with significant health risks. Ozonation has emerged as one of the most promising alternatives to chlorination. Ozonation, however, tends to oxidize bromide to bromate, which presents a potential problem since bromide is naturally present in source waters. Bromate has been judged by both the U.S. EPA and the World Health Organization as a potential carcinogen, even at the low µg/L level. Many regulatory agencies prefer to regulate potential carcinogens to the 10^{-5} health risk level or lower.* The U.S. EPA has recommended that bromate in ozonated water be controlled to < 10 µg/L while further health risk studies are underway. Accordingly, analytical methods must be found to quantify bromate at these levels, so as to aid in researching ozonation process design options to minimize this contaminant.

The following equations show the pathway by which bromide (Br^-) is oxidized by ozone to bromate (BrO_3^-) through the intermediate formation of hypobromite (OBr^-). These equations also show that ozone does not oxidize hypobromous acid (HOBr) to bromate. Since increased acid (H_3O^+) will favor the formation of hypobromous acid, this suggests that ozonation at a low pH will tend to minimize bromate formation (see Figure 1).

$$Br^- + O_3 + H_2O \longrightarrow HOBr + O_2 + OH^- \quad \{1\}$$
$$HOBr + H_2O \longrightarrow H_3O^+ + OBr^-$$
$$OBr^- + 2O_3 \longrightarrow BrO_3^- + 2O_2$$
$$HOBr + O_3 \longrightarrow \text{No Reaction}^1$$

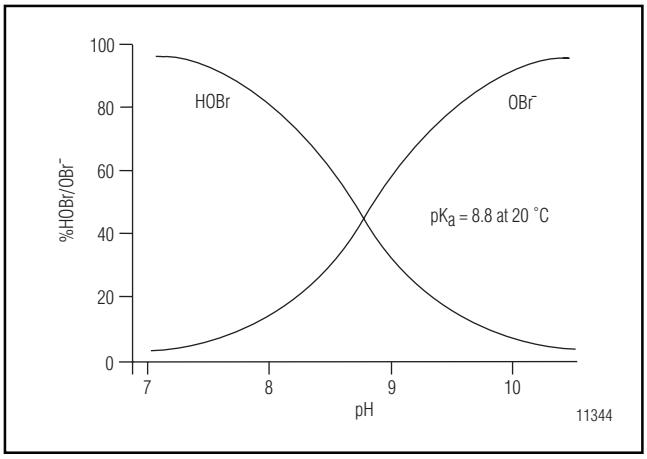

Figure 1 *A decrease in pH favors the formation of hypobromous acid.*

The final concentration of bromate is dependent on the concentration of bromide in the source water, ozone concentration, and duration of contact. Currently, the separation of bromate in a drinking water matrix is accomplished by using direct injection ion chromatography (IC) with suppressed conductivity detection.

Table 1 shows the method detection limits (MDLs) for bromate and other anions of interest that were achieved by U.S. EPA researchers with a 200-µL injected sample on a Dionex IonPac® AS9-SC column using a borate-based eluent. The detection limit for bromate using this methodology is 7.3 µg/L.[2] Injecting a larger sample impairs chromatographic efficiency and does not significantly improve MDLs. The disadvantage to this method is that the amount of bromate present in a typical ozonated water sample is near or below the current detection limit.

* Probable increase in deaths due to a cancer, 10^{-5} = 1 in 100,000 people.

This application note reports the development of a modified IC method that significantly improves the method detection limits for bromate by sample preconcentration. This method is consistent with the proposed ASTM method for bromate.[3]

In this method, the sample is first preserved by sparging to remove reactive gases, such as chlorine dioxide or ozone. Ethylenediamine is then added to convert any hypobromite to the corresponding bromamines, thus preventing their ongoing conversion to bromate. The preserved sample is then spiked with a magnesium chloride and a sodium carbonate reagent. This sample is then passed through three treatment cartridges in the following sequence: OnGuard-Ba, OnGuard-Ag, and OnGuard-H. This treatment reduces the concentration of sulfate, chloride, carbonate, and metals. The magnesium and carbonate ions are added to ensure reliable sulfate reduction, while maintaining high recovery of bromate.

The treated sample is then loaded into a large sample loop (e.g., 2–5 mL) and the anions remaining in the sample, including bromate, are concentrated on a high capacity concentrator column. A weak borate eluent is then used to elute the concentrated anions through the analytical column set where they are separated and through the suppressed conductivity detector where they are quantified. After bromate is eluted, a strong borate eluent is used to purge the columns of remaining ions prior to analysis of the next sample.

Table 1 Method detection limits using the AS9-SC column

Anion	Spiking Conc. µg/L	Stats MDL[a] µg/L	Noise MDL[b] µg/L	Conservative MDL, µg/L
ClO_2^-	10.0	3.4	2.9	3.4
ClO_3^-	25.0	5.2	9.4	9.4
BrO_3^-	10.0	7.3	5.9	7.3
Br^-	10.0	3.9	8.3	8.3

[a] MDL = SD (t_3)
[b] MDL = 3 x noise
Conditions: 9 mM NaOH, 36 mM Boric acid, 1.0 mL/min, 200 µL injection

EQUIPMENT

Dionex DX 500 IC system consisting of:
- GP40 Gradient Pump
- LC20 Chromatography Enclosure with Second Channel Option
- CD20 Conductivity Detector with DS3 Detection Stabilizer
- EO1 Eluent Organizer
- DXP Sample Delivery Pump
- AS40 Automated Sampler
- AC2 Power Control Accessory

PeakNet Chromatography Workstation

REAGENTS AND STANDARDS

Deionized water, 17.8 MΩ-cm resistance or better (Type I reagent grade)

Boric acid, >99% pure (Aldrich, Milwaukee, Wisconsin, USA)

Sodium hydroxide, 50% (w/w) (Fisher Scientific, Pittsburgh, Pennsylvania, USA)

Potassium bromate (Fluka Chemie AG, Buchs, Switzerland)

Magnesium chloride (Aldrich, Milwaukee, Wisconsin, USA)

Sodium carbonate (Aldrich, Milwaukee, Wisconsin, USA)

Ethylenediamine, 99% (Aldrich, Milwaukee, Wisconsin, USA)

SAMPLE PRETREATMENT CARTRIDGES

OnGuard™-Ba Cartridges

OnGuard-Ag Cartridges

OnGuard-H Cartridges

CONDITIONS

Columns: IonPac AS9-SC Analytical, 4-mm i.d.
IonPac AG9-SC Guard, 4-mm i.d.
IonPac AG10 Guard, 4-mm i.d., or TAC-LP1 (Concentrator Column)

Metal Trap Column: MetPac™ CC-1

Eluent A: 40 mM Boric acid/20 mM Sodium hydroxide

Eluent B: 200 mM Boric acid/100 mM Sodium hydroxide

Gradient:

Time (min)	Eluent A (%)	Eluent B (%)	Valve A (setting)	Valve B (setting)
0.00	100	0	Load	Inject
1.50	100	0	Inject	Load
6.50	100	0	Load	Inject
12.49	100	0	Load	Inject
12.50	0	100	Load	Inject
17.49	0	100	Load	Inject
17.50	100	0	Load	Inject

Flow Rate: 2 mL/min
Injection Volume*: 5 mL (max.)
Concentrator Pump
Flow Rate: 2 mL/min
Detection: Suppressed conductivity
Suppressor: ASRS AutoSuppression, external water mode

Note: Loop volume should be checked by filling loop with water and determining actual volume by weight on an analytical balance.

*Use 0.037-inch i.d. tubing for sample loop (1 cm = 6.9 µL)

PREPARATION OF SOLUTIONS AND REAGENTS

Standard Solutions

Bromate (BrO_3^-) 1000 mg/L

Dissolve 1.31 g of potassium bromate ($KBrO_3$) in water and dilute to 1.00 L.

Preservation Solution

Ethylenediamine Preservation Solution (45 g/L)

Dilute 10 mL of ethylenediamine (99%) to 200 mL with water. Use 1 mL of this solution per liter of sample.

OnGuard-Ba Activating Reagents

0.50 M ($MgCl_2$)

Dissolve 48 g of magnesium chloride in water and dilute to 1 L.

0.17 M (Na_2CO_3)

Dissolve 18 g of sodium carbonate in water and dilute to 1 L.

Chromatography Eluent (A)

40 mM Boric Acid / 20 mM Sodium Hydroxide

Dissolve 2.47 g boric acid in 990 mL of water, add 1.6 g of 50% sodium hydroxide, and dilute to 1.00 L. Transfer this solution to an eluent container and vacuum degas for 10 minutes.

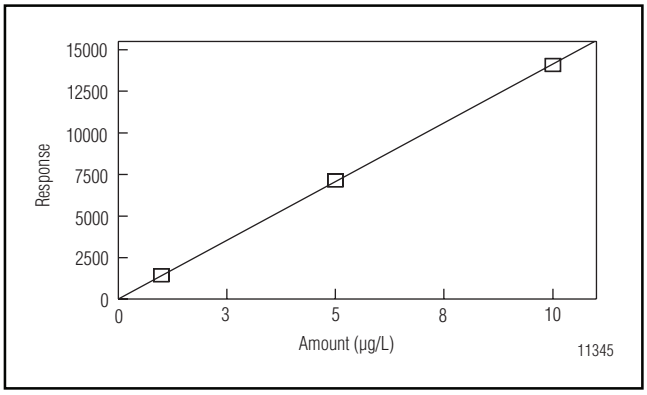

Figure 2 *Linearity plot for bromate at µg/L levels.*

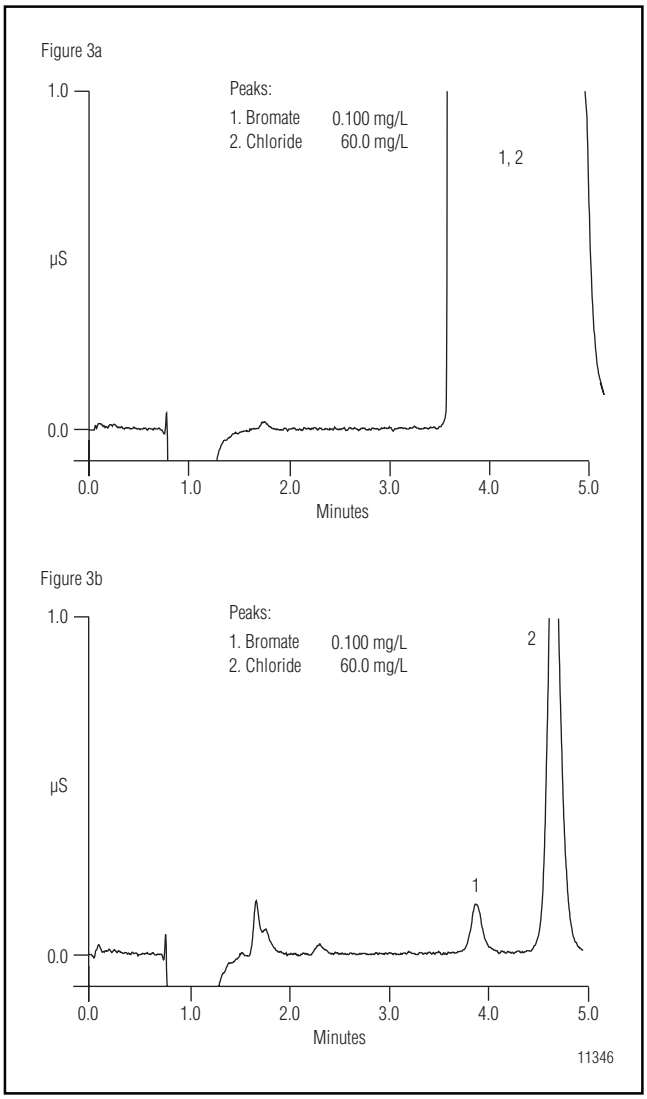

Figure 3a *An untreated sample in which chloride coelutes with bromate.*

Figure 3b *A sample pretreated with OnGuard-Ag in which bromate is resolved from chloride.*

Purge Eluent (B)

200 mM Boric Acid /100 mM Sodium Hydroxide

Dissolve 12.36 g of boric acid in 900 mL of water, add 8.0 g of 50% sodium hydroxide and dilute to 1.00 L. Transfer this solution to an eluent container and vacuum degas for 10 minutes.

Note: Care must be taken to minimize air contact with hydroxide reagent as absorbed carbon dioxide will change eluent characteristics. Keep eluent containers pressurized with an inert gas to prevent atmospheric carbon dioxide from entering.

Preparation of Calibration Standards

Calibration Standards

Prepare calibration standards at a minimum of three concentrations in deionized water from the stock standard solution. The lowest concentration of the bromate standard should slightly exceed 1 µg/L, the method detection limit (MDL). The other concentrations of the calibration curve should correspond to the expected range of concentrations found in the samples of interest. A typical calibration curve is shown in Figure 2.

SAMPLE PREPARATION

Samples should be sparged for 5 minutes to remove any reactive gases. Next, preserve samples with ethylenediamine to prevent oxidation of chlorite or formation of bromate from hypobromite by adding 1 mL of ethylenediamine preservation solution per liter of sample. Now add the OnGuard-Ba activating reagents: 1 mL of the 0.5 M magnesium chloride reagent and 1 mL of the sodium carbonate reagent to 100 mL of sample. All samples should be filtered through a 0.45-µm filter prior to injection. With high levels of sulfate, chloride, and carbonate in the sample matrix, the exchange sites on the AG10/AS9-SC columns are overloaded and bromate cannot be detected as a separate peak (Figure 3a). Sulfate is removed by passing the sample through the Dionex OnGuard-Ba cartridge. This cartridge removes sulfate by forming the precipitate barium sulfate.

Chloride is removed by passing the sample through the Dionex OnGuard-Ag cartridge. Chloride precipitates as silver chloride. Next, the sample is passed through the Dionex OnGuard-H cartridge. It minimizes the carbonate in the sample by converting it to carbonic acid, which is removed by sparging the sample with helium for 2–3 minutes.

Table 2 Determination of bromate in drinking water, 5 mL preconcentrated

Sample	Bromate Present (µg/L)	Bromate Added (µg/L)	Bromate Found[a] (µg/L)	n	SD (µg/L)	MDL[b] (µg/L)
Raw Water A	ND[c]	1.0	1.1	7	0.09	0.3
	ND	5.0	5.1	6	0.29	0.9
	ND	10.0	10.0	7	0.58	1.7
Raw Water B	1.1	0.0	1.1	7	0.04	0.1
Raw Water B	1.1	1.0	1.2	7	0.11	0.3
(Ozonated)	1.1	5.0	4.7	7	0.70	2.1
	1.1	10.0	10.0	5	1.52	5.1

[a] Reference to 10 µg/L fortification of matrix

[b] MDL = SD x (t_s) 99%

[c] ND = Not Detected (< 0.1 µg/L)

DISCUSSION AND RESULTS

Traditionally, a cation resin in the barium-form has been used to remove sulfate from the sample matrices by forming a barium sulfate precipitate ($K_{sp} = 1.1 \times 10^{-10}$). Using this method, however, the sulfate removal varies considerably. Our studies indicate that for consistent sulfate removal, a sample must have a sufficient amount of a divalent cation to displace the divalent barium from the resin so that it can react with sulfate.[4] Some samples have sufficient calcium and magnesium to initiate the barium displacement; whereas, others do not contain sufficient levels of calcium and magnesium. Therefore, to ensure consistent sulfate removal a divalent cation such as magnesium must be added. It has been determined that at least 120 mg/L in magnesium from magnesium chloride will provide sufficient barium displacement from a cation resin in the barium form (OnGuard-Ba) for removal of sulfate up to 500 mg/L. Furthermore, it has been found that a minimum level of carbonate is required to ensure high bromate recovery when sulfate is being removed by the OnGuard-Ba cartridge. The excess chloride can be removed with OnGuard-Ag treatment, which is also required for removing chloride that is normally present in the sample.

The OnGuard-Ag cartridge packing is a silver form, high capacity, strong acid, cation exchange resin that is designed to remove chloride from the sample matrices. The cartridge capacity is 1.5–1.8 meq per cartridge. By treating the sample with the OnGuard-Ag cartridge, the chloride level is reduced to approximately 0.4 mg/L; this level is sufficient to resolve bromate from chloride (see Figure 3b).

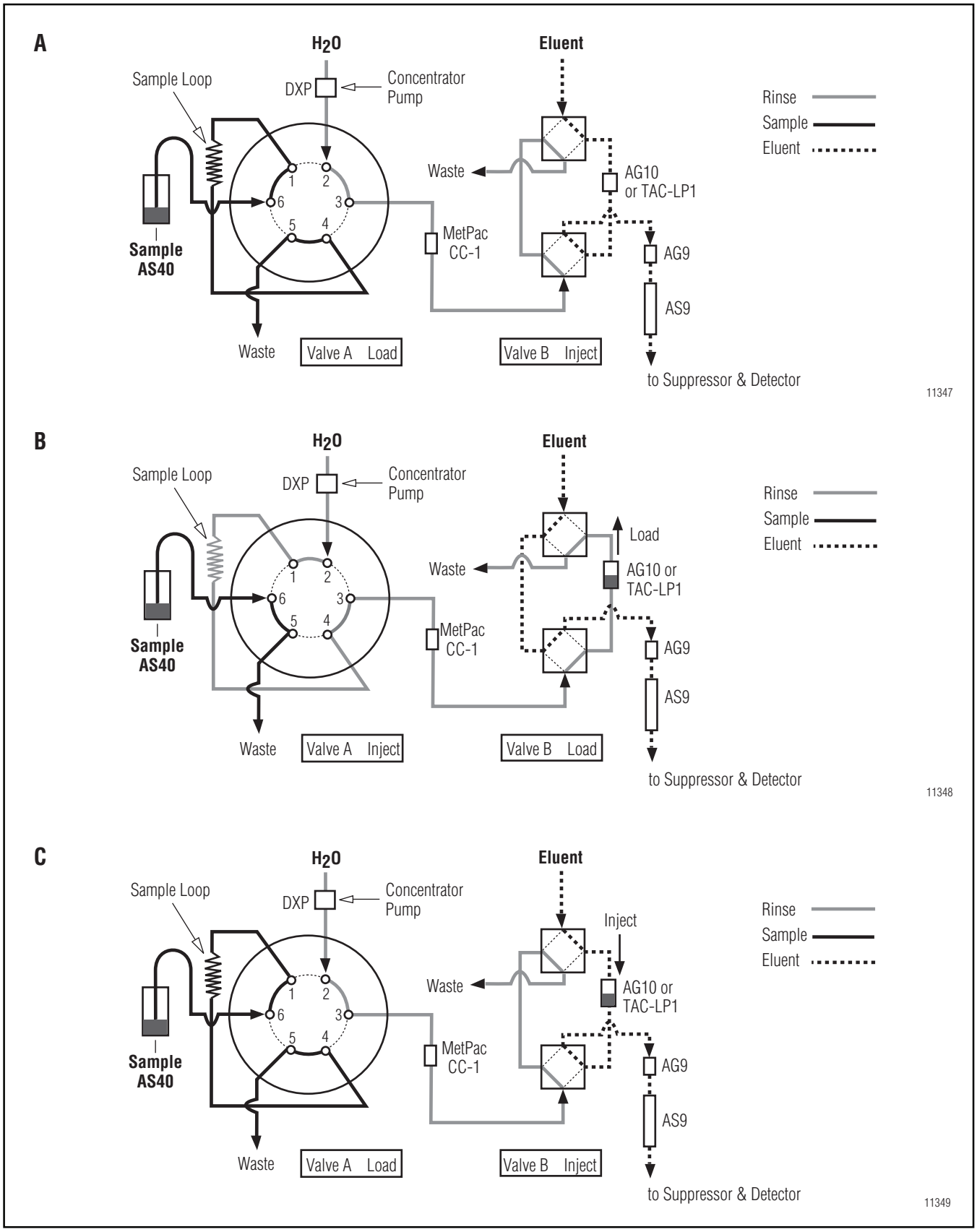

Figure 4 *Preconcentration suppressed IC system configuration. (A) sample loaded into loop; (B) sample washed onto concentrator column; (C) retained anions eluted to AG9/AS9-SC analytical column set.*

The lower sulfate and chloride levels allow for a larger sample volume to be concentrated, which improves the bromate response. The OnGuard-H sample pretreatment cartridge is a hydrogen-form cation resin. Its use followed by helium sparging minimizes the carbonate in the sample, which further improves preconcentration efficiency. The use of the OnGuard-Ag leaches a small amount of silver from the cartridge into the sample matrix. The accumulation of silver on the analytical column and concentrator column will affect column performance over time. The OnGuard-H cartridge also removes metal ions such as silver. To further avoid metal contamination, a Dionex MetPac CC-1 column is installed between the two injection valves (see Figure 4). The MetPac CC-1 metal chelating column not only removes the silver, but it also removes other metal cations that may foul the analytical column.

The determination of bromate utilizing this method is a three step process as illustrated in Figures 4a–4c: Step 1 loads the sample loop, Step 2 washes the sample onto the concentrator, Step 3 separates the anions of interest on the analytical column. Figure 4a illustrates the sample being loaded into the sample loop using an autosampler. During this first step, the GP40 pumps Eluent A to the AS9-SC column. After the loop is filled, the DXP Pump is turned "ON" and it washes the sample from the sample loop onto the concentrator column using deionized water (see Figure 4b). The sample loop is then rinsed 2.5 times its volume to ensure that all of the sample is transfered onto the concentrator. The concentrator column strongly retains anionic species such as bromate, chloride, and sulfate. Figure 4c shows the concentrator column being switched in-line with the IonPac AG9/AS9-SC columns. At this step, the retained anions are eluted to the analytical column. After the chloride elution, the remaining anions are purged off the analytical column using the purge eluent.

After purging for 5 minutes, the AG9/AS9-SC columns are equilibrated with the chromatography eluent for 7–10 minutes. The equilibration time is placed at the beginning of the analysis sequence, during which the sample loop is being filled and the sample is flushed onto the concentrator column. The total analysis time for this method is 25 minutes. Table 2 lists the bromate MDLs that have been achieved when preconcentrating raw water samples, obtained before and after ozonation.

Using this method, bromate (at the low µg/L level) can be measured in a matrix containing as much as 200 mg/L of chloride, carbonate, and sulfate as shown in Figure 5. Figure 6 shows the analysis of an ozonated drinking water sample spiked with 1 µg/L bromate.

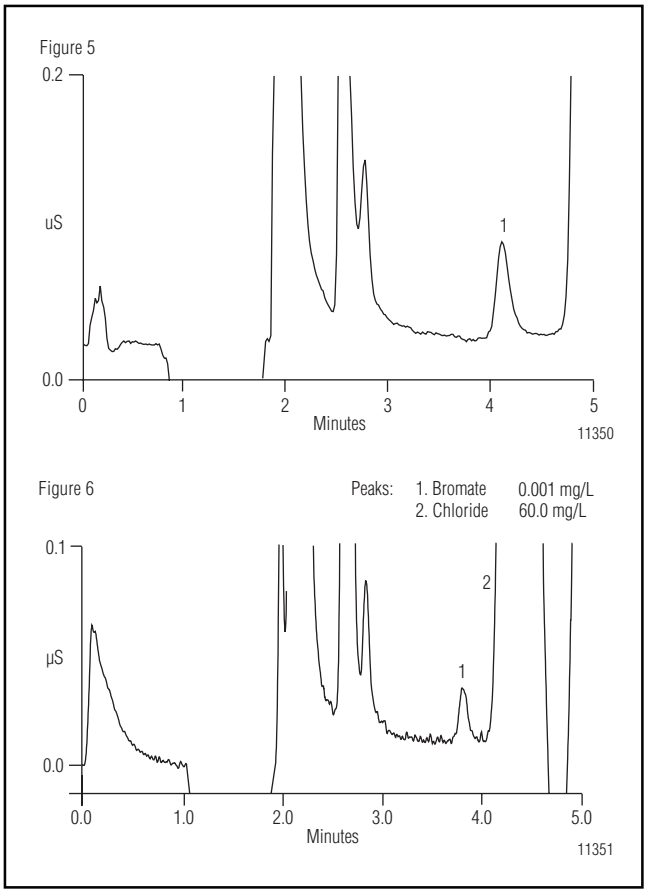

Figure 5 *Bromate in the presence of 200 mg/L of chloride, carbonate, and sulfate.*

Figure 6 *An ozonated drinking water sample is spiked with 1.0 µg/L of bromate.*

REFERENCES

1. Haag, W. R.; Hoigne, J. *Environmental Science and Technology,* **1983***, 17*, 261.
2. Hautman, D. P.; Bolyard, M. *J. Chromatogr,* **1991**, *602*, 7.
3. American Society for Testing and Materials. Proposed ASTM Method "Determination of Oxyhalides and Bromide in Water by Chemically Suppressed Ion Chromatography," under review by the ASTM D-19 Committee on Water.
4. Patent filed.

Determination of Haloacetic Acids in Water Using IC-ESI-MS/MS

INTRODUCTION

This method allows separation and detection of sub-µg/L levels of nine haloacetic acids (HAAs) in high-ionic strength matrices. Using this method, the analytes are separated from chloride, sulfate, nitrate, bromide and bicarbonate, and detected using a triple quadrupole mass spectrometer with an electrospray interface. Quantification is achieved using internal standards.

Haloacetic acids occur in drinking water during the disinfection process, as a result of the reaction between chlorine and natural organic materials, such as humic and fulvic acids.[1,2] The iodoacids (e.g. iodoacetic acid) are much less stable and are not included in this analysis. When bromide is present in the water, bromoacetic acids and mixed chloro- and bromoacetic acids can also be generated. Haloacetic acids have been linked to possible health threats to human health. Monitoring for monochloroacetic acid (MCAA), monobromoacetic acid (MBAA), dichloroacetic acid (DCAA), trichloroacetic acid (TCAA) and dibromoacetic acid (DBAA) has been in effect since they were first regulated under the Stage I Disinfection Byproducts (DBP) Rule, Dec. 16, 1998, with a Minimum Contamination Level (MCL) set at 60 µg/L. Stage II DBP Rule, Jan. 4, 2006, maintained the MCL, but also instituted minimum reporting limits (MRL) requirements of 2 µg/L for MCAA and 1 µg/L for the other HAAs. The remaining four HAAs that may be present in drinking water are: chlorobromoacetic acid (CBAA), chlorodibromoacetic acid (CDBAA), dichlorobromoacetic acid (DCBAA), and tribromoacetic acid (TBAA).

The determination of the chloro-, bromo-, and mixed haloacetic acids in waters destined for human consumption, including drinking water and swimming pool water, has been reported using a variety of analytical techniques.[3] USEPA Methods 552.2 and 552.3 use acidic methanol derivatization followed by gas chromatography with electron capture detection.[4] This method is both labor-intensive and time-consuming. Bruzzoniti[5] recently published a table summarizing the existing IC columns and methods used for HAAs analysis, including this method, which uses the IonPac® AS24 column. Asami[6] used offline sample pretreatment with external standard calibration and MS/MS detection for calibration of haloacetic acids and oxyhalides, using perchlorate as an internal standard. Only the method using the IonPac AS24 method addresses the issue of high-ionic strength matrices. All the IC methods take advantage of the low pKa values of HAAs (~ 0.7–2.8) by using anion-exchange separation mode. Hydroxide-based eluents are used in conjunction with chemical suppression, so the background signal entering the mass spectrometer is as low as that of water. When mass spectrometric detection is used, the matrix ions are typically diverted to waste during the analytical run to avoid contamination of the detector. USEPA Method, 332.0[7] uses the same configuration as discussed in this paper for the determination of perchlorate in drinking water; namely, ion chromatography with matrix diversion and detection using suppressed conductivity followed by electrospray mass spectrometry. The selectivity of the

analytical column in a method using matrix diversion must be designed such that the matrix ions are sufficiently resolved from target analytes.

Four internal standards are used in our method for the nine target analytes. These were chosen because they elute throughout the chromatographic run, thus allowing easy tracking of close-eluting analytes. Stuber and Reemtsma[8] discuss the challenges of quantification using LC-ESI-MS in the presence of significant matrix effects, and provide some guidance for using internal standards. Most of the currently published work describing various analytical approaches for determination of HAAs, however, does not adequately address the challenges of very high-ionic strength matrices, or the need for internal standards to obtain accurate and precise quantification.

Figure 1 shows the chromatogram of the nine standards, the two matrix diversion windows, and the general form of the KOH gradient. The three periods noted in the figure correspond to three periods for data collection in the Analyst method program. Figure 2 shows the general instrumentation schematic.

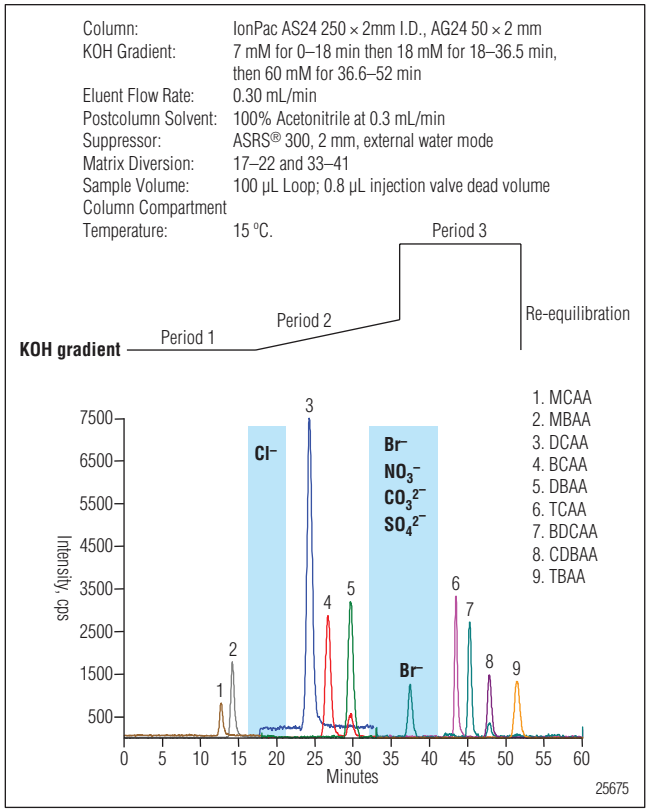

Figure 1. Chromatogram produced by chromatography conditions and mass spectrometer conditions provided in Table 1. The shaded areas show the time windows for matrix diversion to waste and the matrix ions that elute in those windows. The time windows for data collection in Periods 1, 2, and 3 are indicated.

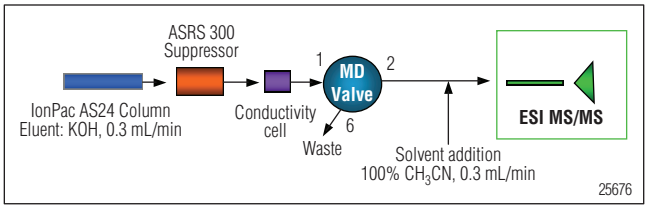

Figure 2. ICS-3000 system schematic, showing matrix diversion and mass spectrometric detection.

RECOMMENDED EQUIPMENT

ICS3000 Chromatography System

DP Dual Pump module (or SP and an AXP)

DP1 Analytical Pump

DP2 pump is used to deliver post-suppressor acetonitrile

DC Dual-Zone Chromatography Module

CD Conductivity Detector

AS Autosampler with sample tray cooling

EG Eluent Generator

Mixing Tee (Upchurch, part number U-466)

Mass Spectrometer

ABI-Sciex (Toronto, Canada) API2000™ Triple Quadrupole Mass Spectrometer with electrospray interface capable of negative ion detection, or equivalent

Nitrogen and Zero Air supplies as specified by MS manufacturer

25-pin relay cable connecting mass spectrometer to DP pump module; pins 19 (red) and 7 (black) in the DP connector

Software

Dionex DCMSLink™ 2.0 software or higher

ABI Sciex Analyst software (version 1.4.2 or higher)

XCalibur 2.0 or higher

Standards

Deionized water: 18 MΩ or better

Acetonitrile (HPLC grade)

Analyte standard mix (1000 µg/mL) of the nine native HAAs; monochloroacetic acid (MCAA), monobromoacetic acid (MBAA), dichloroacetic acid (DCAA), trichloroacetic acid (TCAA), dibromoacetic acid (DBAA), chlorobromoacetic acid (CBAA), chlorodibromoacetic acid (CDBAA),

dichlorobromoacetic acid (DCBAA), and tribromoacetic acid (TBAA) purchased from Restek (P/N31896). The internal standards from Dionex are: monochloroacetic acid-2-13C (1000 µg/mL, P/N 069406), monobromoacetic acid-1-13C (1000 µg/mL, P/N 069407), dichloroacetic acid-2-13C (1000 µg/mL, P/N 069408), and trichloroacetic acid-2-13C (1000 µg/mL, P/N 069409). Standards are dissolved in methyl tert-butyl ether (MtBE). A working standard mixture of the four internal standards was prepared in deionized water. All standard solutions were kept refrigerated at 4 °C when not in use. Standards in the 2-5 µg/L range are stable for 14 days when stored at 4 °C with PTFE/silicone septa. Because the standards are purchased in MtBE, which has limited solubility in water (~ 5%), not more than ~ 0.5% of MtBE is added when making the mixtures, relative to the total water volume.

Sample Preparation

Samples were collected in amber glass bottles with PTFE-lined screw caps. Crystalline or granular ammonium chloride is added to the sample containers to produce a final concentration of 100 mg/L of ammonium chloride. The preservation requirements are exactly the same as those described in EPA Method 552.3.

CONDITIONS

Chromatography Conditions

Column: IonPac AS24 250 × 2mm I.D., IonPac AG24 50 × 2mm

KOH Gradient:

Time	KOH (mM)
-7.0	7
0.0	7
18.0	7
36.5	18
36.6	60
52.0	60

Eluent Flow Rate: 0.30 mL/min

Postcolumn Solvent: 100% Acetonitrile at 0.3 mL/min

Suppressor: ASRS 300 (2 mm) External water mode

Anion Trap: CR-ATC (2-mm)

Matrix Diversion: 17-22 and 33-41

Sample Volume: 100 µL sample loop

Column Compartment Temperature: 15 ºC

Autosampler Temperature: 8 °C

Detector Compartment Temperature: 30 °C

Mass Spectrometric Conditions: Tables 1-5

Table 1. API2000 Conditions

Analyte	KOH Gradient	Transition	Source-Dependent Parameters	Declustering Potential (V)	Focusing Potential (v)	Collision Energy (eV)	Entrance Potential (V)	Collsion Cell Entrance Potential (V)	Collision Cell Exit Potential (V)	Dwell Time (mSec)
MCAA MCAA-21-13C	7 mM 0 – 18 min	92.9/34.9 93/34.9	Curtain 20 CAD 2 Ionspray -4500 Temp 475 °C GS1/GS2 90/90	-20	-300	-12	-10	-12	-6	600 each
MBAA MBAA-1-13C		137/78.8 138/78.8		-11	-350	-12	-7	-10	-14	600 each
Dalapon	18 mM 18 – 36.5 min.	141/97	Curtain 25 CAD 4 Ionspray -4500 Temp 475 °C GS1/GS2 90/90	-13	-350	-11	-8	-13	-6	500
DCAA DCAA-2-13C		127/82.9 128/84		-11	-320	-12	-6.5	-12	-6	500 each
BCAA		170.8/78.8		-16	-300	-28	-6	-14	-8	500
DBAA		214.7/170.7		-11	-350	-12	-4.5	-15	-10	500
TCAA TCAA-2-13C	60 mM 36.6 – 52 min	161/116.9 162/118	Curtain 25 CAD 4 Ionspray -4500 Temp 475 °C GS1/GS2 90/90	-6	-290	-7	-7	-13.7	-13.7	400 each
BDCAA		207/81 or 79/79		-12	-300	-6	-4	-15	-14	400
CDBAA		207/78.8		-11	-300	-20	-4	-15	-6	400
TBAA		250.75/78.8		15	-350	-28	-5	-12	-12	400

Table 2. API3200 Conditions									
Analyte	**KOH Gradient**	**Transition**	**Source-Dependent Parameters**	**Declustering Potential (V)**	**Collision Energy (eV)**	**Entrance Potential (V)**	**Collsion Cell Entrance Potential (V)**	**Collision Cell Exit Potential (V)**	**Dwell Time (mSec)**
MCAA MCAA-2-13C	7 mM 0 – 18 min	92.9/34.9	Curtain 30 CAD 2 Ionspray -4500 Temp 500 °C GS1/GS2 70/70	-15	-3	-5	-17	-5	600 each
MBAA MBAA-1-13C		137/78.8		-14	-7	-8	-20	-1.5	600 each
Dalapon	18 mM 18 – 36.5 min.	141/97	Curtain 30 CAD 3 Ionspray -4500 Temp 500 °C GS1/GS2 70/70	-17	-5	-5	-11	-1	500
DCAA DCAA-2-13C		127/82.9		-15	-3	-5	-17	-1	500 each
BCAA		170.8/78.8		-26	-4	-8	-32	-1.5	500
DBAA		214.7/170.7		-22	-3.5	-22.8	-18	-1.5	500
TCAA TCAA-2-13C	60 mM 36.6 – 52 min	161/116.9	Curtain 30 CAD 3 Ionspray -4500 Temp 250 °C GS1/GS2 70/70	-12	-3	-6	-19	-1	400 each
BDCAA		207/81 or 79/79		-85	-4	-15.1	-10	-1.5	400
CDBAA		207/78.8		-12	-3	-16.8	-6	-14	400
TBAA		250.75/78.8		-13	-2.5	-13	-32	-1.5	400

Table 3. API4000 Conditions								
Analyte	**KOH Gradient**	**Transition**	**Source-Dependent Parameters**	**Declustering Potential (V)**	**Collision Energy (eV)**	**Entrance Potential (V)**	**Collision Cell Exit Potential (V)**	**Dwell Time (mSec)**
MCAA MCAA-2-13C	7 mM 0 – 18 min	92.9/34.9	Curtain 20 CAD 2 Ionspray -4000 Temp 500 °C GS1/GS2 50/50	-25	-15	-2	-3	600 each
MBAA MBAA-1-13C		137/78.8		-25	-15	-2	-3	600 each
Dalapon	18 mM 18 – 36.5 min.	141/97	Curtain 20 CAD 8 Ionspray -4300 Temp 500 °C GS1/GS2 50/50	-33	-15	-4	-13	500
DCAA DCAA-2-13C		127/82.9		-43	-31	-5	-2	500 each
BCAA		170.8/78.8		-31	-17	-5	-9	500
DBAA		214.7/170.7		-21	-11	-4	-5	500
TCAA TCAA-2-13C	60 mM 36.6 – 52 min	161/116.9	Curtain 20 CAD 10 Ionspray -4200 Temp 250 °C GS1/GS2 50/50	-35	-22	-4.5	-12	400 each
BDCAA		206.8/81		-35	-22	-4.5	-12	400
CDBAA		206.8/81		-35	-22	-4.5	-12	400
TBAA		250.9/78.8		-30	-34	-4	-12	400

Table 4. Thermo Quantum Access Conditions								
Analyte	**Q1/Q3**	**CE**	**Tube Lens**	**Cap Temp (°C)**	**Sheath Gas/Aux Gas**	**Ion Sweep**	**Skimmer Offset (V)**	**Scan Time/ Section (s)**
MCAA MCAA-2-13C	93/35.6 94/35.6	10	26	270	40/15	0.1	0	1.25
MBAA MBAA-1-13C	137/79.1 138/79.1	12	33	270	40/15	0.1	0	1.25
DCAA DCAA-2-13C	127/83.2 128/84	11	26	270	40/15	0.1	0	1.25
DBAA	214.8/79.2	24	33	270	40/15	0.1	0	1.25
BCAA	171/79.2	35	44	270	40/15	0.1	0	1.25
TCAA TCAA-2-13C	161.1/117.1 162/118	10	69	270	40/15	0.1	0	1.6
BDCAA	79/79	15	30	270	40/15	0.1	0	1.6
CDBAA	206.7/79.1	15	30	270	40/15	0.1	0	2.5
TBAA	250.7/79.1	25	26	270	40/15	0.1	0	2.5

Table 5. Waters Quattro Premier Parameters					
Analyte	**Transition**	**Dwell (sec)**	**Cone, V**	**Extractor/RF Lens/Source Block Temp (V/V/oC)**	**Collision Energy, V**
MCAA MCAA-2-13C	92/35 93/93	1.0 0.5	15	-3/-0.5/120	8
MBAA MBAA-1-13C	136.9/78.9	0.5	15	-3/-0.5/120	10
DAL	140.9/97	0.5	18	-3/-0.5/120	8
DCAA DCAA-2-13C	126.9/83	0.5	17	-3/-0.5/120	10
BCAA	172.9/128.9	0.5	17	-3/-0.5/120	10
DBAA	216.8/172.84	0.5	18	-3/-0.5/120	12
TCAA TCAA-2-13C	160.9/116.9 162.9/118.9	0.5 0.5	16	-3/-0.5/120	8
BDCAA	162.9/80.9	1.0	25	-3/-0.5/120	10
CDBAA	206.8/78.9	1.0	28	-3/-0.5/120	10
TBAA	250.8/78.9	1.0	28	-3/-0.5/120	12

Other Conditions:
Desolvation Gas: 350 °C @ 940 L/hr
Capillary: -2.8V
Collision Pressure: 5.5×10^{-3} (0.15 flow @ 7 psig)
Cone Flow: 100 L/hr,
ACN Flow Rate: 0.2 mL/min

RESULTS AND DISCUSSION OF THE METHOD

Separation

One of the most important features of this method is the ability to quantify the HAAs in the presence of matrices of high-ionic strength. Initially, the separation was achieved using the IonPac AS20 (250 × 2 mm I.D., 78 µEq/column). Reduced peak height, lower peak efficiencies, and shifting retention times were observed when the matrix composition exceeded 100 mg/L chloride and sulfate. The IonPac AS24 column (250 × 2 mm I.D.) 140 µEq/column provides approximately twice the anion-exchange capacity as the AS20.9 This improved capacity is required for this application, where the concentration of common matrix ions can be as high as 250 mg/L chloride, 250 mg/L sulfate, 150 mg/L bicarbonate, and 30 mg/L nitrate. The high capacity of the column insures that the ion-exchange sites are not consumed with matrix ions during the separation.

Figure 1 shows separation of nine HAA standards and the time windows for the common matrix ions. The common ions shown are separated from the HAAs, and this separation allows time for the diversion of these ions to waste using a three-way valve before they can enter the mass spectrometer.

Solvent Addition

Addition of 0.3 mL/min of 100% acetonitrile after the IC suppressor and before the ESI inlet improves sensitivity from twofold to 10-fold depending on the analyte and condition of the mass spectrometer. The flow of solvent to the mass spectrometer continues during matrix diversion for stability of the electrospray.

Temperature

The retention times for the HAAs increase as column temperature increases. In addition, some of the HAAs–most notably the brominated species–are less stable at higher temperatures and high pH. The autosampler temperature was set to 8 °C and the column compartment temperature to 15 °C to maximumize analyte and internal standard stability and retention time reproducibility. In addition, stable retention times are critical to maintain the times for the matrix diversion windows.

The ESI source temperature was optimized for maximum sensitivity of all analytes. With this method, trisubstituted HAAs are more susceptible to source temperature changes, and the best sensitivity was achieved at the minimum temperature needed for desolvation in the electrospray interface.

Matrix Diversion

The IonPac AS24 column manual contains a procedure for setting the correct matrix diversion window times and method parameters.

Internal Standards and Calibration

As is common, the ratios of peak areas for analytes and internal standards versus analyte concentration are used to produce the calibration plots. Internal standards were chosen that elute in each of the three sections of the gradient method due to changes in the background and eluent composition over the course of the run. Several choices for Multiple Reaction Monitoring (MRM) transitions were available due to the presence of Cl and Br isotopes. MCAA-2-13C (*m/z* 94 > 35), MBAA-1-13C (*m/z* 138 > 79), DCAA-2-13C [1] (*m/z* 128 > 84), and TCAA-2-13C 13C (*m/z* 162 > 118) were chosen because they exhibited low background and good sensitivity. Other choices may be appropriate depending on sample matrix.

Referring to Figure 1, Period 1 uses 7 mM KOH eluent and the analytes are MCAA and MBAA. Chloride elutes at the end of this region, so a matrix diversion window separates this first section of the gradient from the second section. The brominated acetic acids–especially MBAA–are known to be susceptible to decomposition at elevated temperature and pH, so stable-labeled MBAA-1-13C is used for accurate tracking of the MBAA analyte. MCAA-2-13C is also used as an internal standard in the first section of the chromatogram for the quantification of MCAA. The stable-labeled internal standard for Period 2 of the gradient is DCAA-2-13C. In this section, the KOH concentration ramps to 18 mM and the analytes are the dihaloacetic acids, including DCAA, BCAA, and DBAA. Period 2 ends with the diversion of sulfate, nitrate, bromide, and bicarbonate to waste. The concentration of KOH eluent is increased to 60 mM in Period 3 of the gradient and the trihaloacetic acids TCAA, BDCAA, DBCAA and TBAA elute. The internal standard for this section is TCAA-2-13C.

The system was calibrated using a mixture of nine haloacetic acids at levels of 0.25, 1.0, 2.5, 5.0, 10.0, and 20.0 µg/L, with the four isotopically labeled internal standards at 3.0 µg/L added to each sample and standard. A relative response ratio was generated to produce the calibration plots. A linear fit was used with 1/x weighting. Correlation coefficients in deionized water were 0.998 or better.

Precursor and Product Ions

Precursor ions are generally the result of deprotonation $(M\text{-}H)^-$ of the organic acid. Because the target species all have halide substituents, there are multiple choices for possible transitions. The specific transitions are: MCAA (92.9 > 34.9), MBAA (137 > 78.8), DCAA (127 > 82.9), BCAA (170.8 > 78.7), DBAA (214.7 > 170.7), TCAA (161 > 116.9), BDCAA (207 > 81 or 79 > 79), CDBAA (207 > 78.8), and TBAA (250.7 > 78.8). The trivalent compounds BDCAA and CDBAA are difficult to optimize, and BDCAA often fragments to *m/z* 79 in Q1, so the best sensitivity can usually be found at 79 > 79, although other transitions can be used if they provide adequate sensitivity. The MS/MS voltages are relatively low, suggesting the general fragility of these analytes. Tables 1–5 provide working conditions for five different mass spectrometers tested with this method.

Analytical Results

Table 6 shows linearity in deionized water and a matrix composed of 250 mg/L chloride, 250 mg/L sulfate, 30 mg/L nitrate, and 150 mg/L bicarbonate. The fitting method was linear with 1/x weighting using Analyst software. At the maximum matrix concentrations (250 mg/L chloride, 250 mg/L sulfate, 150 mg/L bicarbonate and 30 mg/L nitrate) linear range is 0.5-10 µg/L with $r^2 = 0.997$ or better. Minimum detection limits (MDLs) were calculated using the Student's *t*-test calculation with seven injections. The MDL values were 0.1–1.0 µg/L for the nine HAAs in the high-level matrix. DCAA showed the highest sensitivity, and the trivalent mixed acids BDCAA and CDBAA showed the least sensitivity.

Calibration check standards (CCS) were placed in each sequence at approximately every 10 sample injections at levels of 0.5 and 5.0 µg/L, and at the end of every sequence. The recovery of each CCS was 95–105% in every instance. In addition, the sample was spiked with 2.5 µg/L of the native calibration mixture to calculate percent recovery.

Figure 3 shows the extracted ion currents for Periods 1 and 2, and Figure 4 shows the extracted ion current for Period 3 from a water sample with high-ionic strength. This sample is from within the pressure zone of a southwest public water utility whose source is primarily surface water. The chloride concentration of the sample was 170 mg/L and the sulfate concentration was 215 mg/L. Concentrations were determined by ion chromatography, and the sample was not diluted before analysis. The monosubstituted and disubstituted halogenated analytes found in Period 1 and 2 are: MCAA (1.2 µg/L), MBAA (0.8 µg/L), DCAA (6.1 µg/L), BCAA (5.8 µg/L), and DBAA (2.9 µg/L). Figure 4 includes the trisubstituted HAAs for the sample. The analytes found in this sample are: TCAA (1.6 µg/L), BDCAA (4.3 µg/L), CDBAA (3.8 µg/L), andTBAA (0.7 µg/L) (See Table 7). These chromatograms were processed using Gaussian smoothing for 10 cycles. Some analytes can be found at several MRM transitions. Analytes that are seen on two MRM transitions used in the method are indicated with arrows. These results were compared to amounts quantified using Method 552.2 and amounts range from 65–130% of that method. (See Table 8).

Figure 4B shows the brominated species in the sample which experienced some degradation in Q1. An unidentified brominated compound elutes just prior to TCAA in the sample; this explains the sharp front on the TCAA peak. The 251 > 79 transistion is the most sensitive for quantification of CDBAA, although, with optimized tuning, monitoring mass 79 (79 > 79) and transistion *m/z* 207 > 81 was useful. The *m/z* 207 ion is the nominal mass for BDCAA and the decarboxylated CDBAA. As the number of bromide substitutions increases, the parent ion becomes less stable. The MRM for TBAA is *m/z* 251 > 79 where the *m/z* 251 ion is the result of decarboxylation of the parent ion. With the isotopes and the presence of the multiple halogens, there are several possibilities for MRM transitions.

Table 6. Linearity and MDL in Deionized Water and Matrix

Analyte	ISTD 5 µg/L	R^2 (Calibration Range 0.250-20 µg/L) DIW/Matrix	MDL µg/L/%RSD (n=7, 1 µg/L)	DI water MDL µg/L/%RSD (n=7, 1 µg/L) In Matrix
MCAA	MCAA-1-13C	0.9997/0.9989	0.51/3.5	0.44/14.7
MBAA	MBAA-1-13C	0.9999/0.9990	0.08/3.6	0.13/4.2
DCAA	DCAA-2-13C	0.9999/0.9991	0.39/2.0	0.10/3.3
BCAA	DCAA-2-13C	0.9999/0.9992	0.20/0.8	0.10/0.8
DBAA	DCAA-2-13C	0.9999/0.9993	0.16/5.5	0.33/10.8
TCAA	TCAA-2-13C	0.9999/0.9993	0.24/0.5	0.09/0.3
BDCAA	TCAA-2-13C	0.9991/0.9991	0.26/5.0	0.64/18.9
CDBAA	TCAA-2-13C	0.9992/0.9994	0.38/5.5	0.52/16.4
TBAA	TCAA-2-13C	0.9994/0.9998	0.26/9.2	0.36/9.9

Table 7. Summary of IC-ESI-MS/MS Analytical Results for Real Samples										
Sample	**Cl^--SO_4^{2-} (mg/L)**	**MCAA IC/MSMS (µg/L) %Spike Rec**	**MBAA IC/MSMS (µg/L) %Spike Rec**	**DCAA IC/MSMS (µg/L) %Spike Rec**	**BCAA IC/MSMS (µg/L) %Spike Rec**	**DBAA IC/MSMS (µg/L) %Spike Rec**	**TCAA IC/MSMS (µg/L) %Spike Rec**	**BDCAA* IC/MSMS (µg/L) %Spike Rec**	**CDBAA IC/MSMS (µg/L) %Spike Rec**	**TBAA IC/MSMS (µg/L) %Spike Rec**
Treated Reservoir Water	163 243	1.11 93%	1.08 103%	15.1 72%	8.5 76%	3.72 84%	5.85 80%	7.13 104%	4.75 92%	1.07 106%
Sample M	93 237	2.31 118%	1.16 106%	15.0 56%	9.4 65%	4.40 80%	6.2 70%	7.49 99%	5.12 72%	1.19 125%
Sample O	170 215	1.21 116%	0.82 105%	6.11 96%	5.83 94%	2.93 98%	1.59 91%	4.27 92%	3.85 100%	0.76 95%

Table 8. Summary of Method 552.2 Results for Real Samples										
Sample	**Cl^--SO_4^{2-} (mg/L)**	**MCAA (µg/L) 552.2 %Rec**	**MBAA (µg/L) 552.2 %Rec**	**DCAA (µg/L) 552.2 %Rec**	**BCAA (µg/L) 552.2 %Rec**	**DBAA (µg/L) 552.2 %Rec**	**TCAA (µg/L) 552.2 %Rec**	**BDCAA (µg/L) 552.2 %Rec**	**CDBAA (µg/L) 552.2 %Rec**	**TBAA (µg/L) 552.2 %Rec**
Treated Reservoir Water	163 243	1.31 85%	0.95 113%	17.33 87%	10.53 81%	4.74 78%	7.81 75%	7.75 104%	6.39 74%	Na
Sample M	93 237	2.12 109%	0.89 130%	16.33 92%	9.86 95%	4.44 100%	7.09 87%	7.03 106%	6.03 85%	Na
Sample O	170 215	1.33 91%	0.64 128%	6.23 98%	6.54 89%	3.43 85%	2.24 71%	4.32 99%	5.95 65%	Na

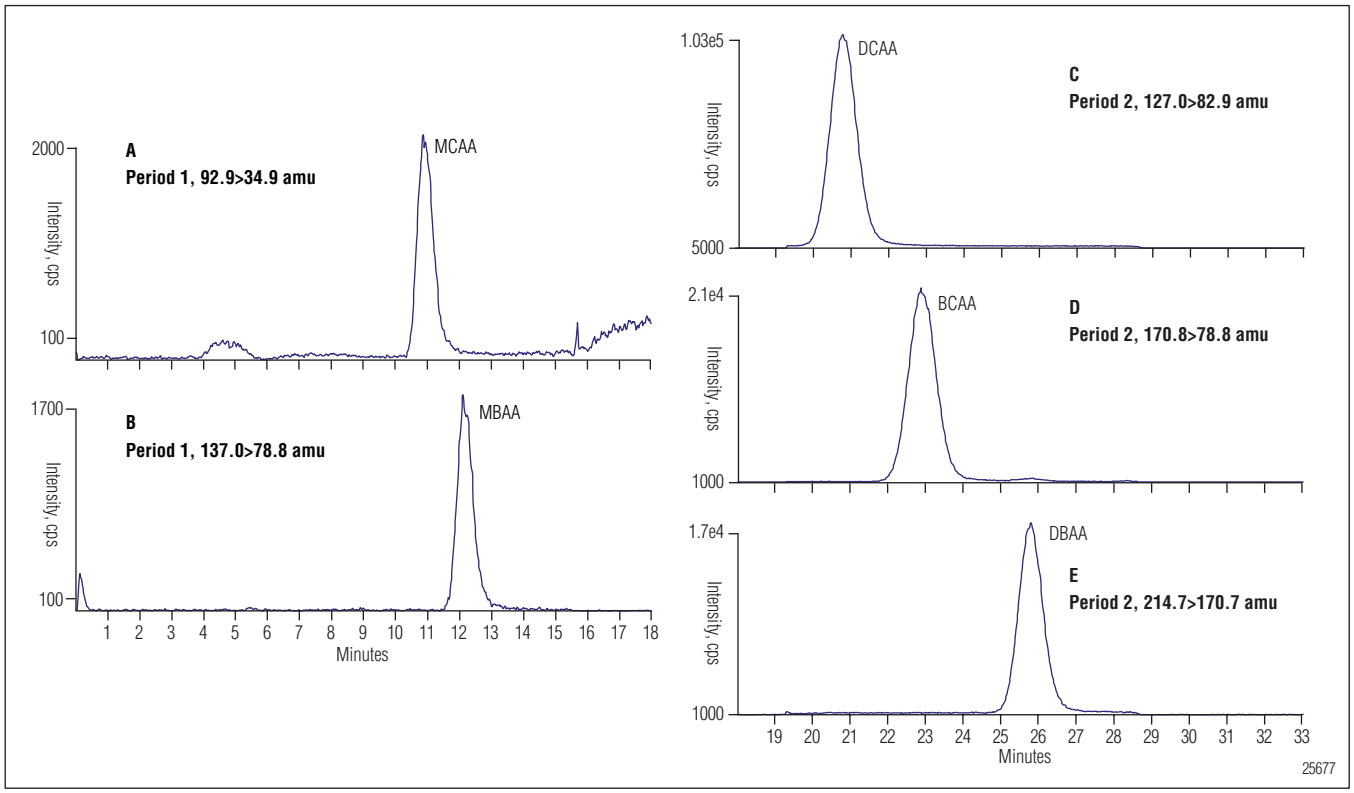

Figure 3. Extracted ion currents for Periods 1 and 2 for the treated water reservoir sample. A, monochloroacetic acid, 1.2 µg/L found; B, monobromoacetic acid, 0.82 µg/L found; C, dichloroacetic acid, 6.1 µg/L found; D, bromochloroacetic acid, 5.8 µg/L found; E, dibromo-acetic acid, 2.9 µg/L found. MRMs are as indicated. For conditions, see Figure 1.

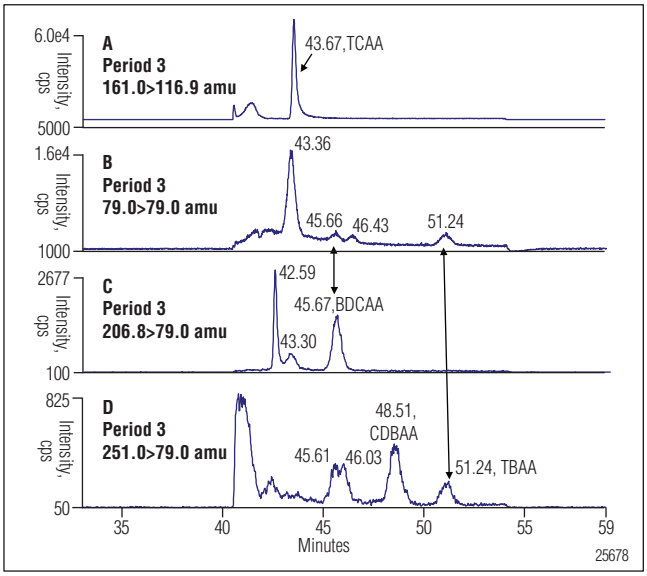

Figure 4 shows the extracted ion chromatograms for the indicated transistions of a water sample with high-ionic strength. A, 1.6 µg/L trichloroacetic acid found; B, bromide fragments; C, 4.3 µg/L bromodichloroacetic acid found; D, 3.8 µg/L chlorodibromoacetic acid and 0.7 µg/L tribromoacetic acid found

Conclusion

This application note describes a method for the determination of haloacetic acids without sample preparation. Using IC-MS/MS with the RFIC™ system and matrix diversion, this method provides sub-µg/L level detection of nine haloacetic acid compounds with direct injection of a sample with high-ionic strength. The parameters used in this method were used in the development of EPA method 557 for determination of haloacetic acids, bromide, and Dalapon, a general-use pesticide.

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Column Selection Guide

Environmental Water Applications Notebook

Column Selection Guide

Silica Columns

			Reversed-Phase (RP)						Mixed-Mode			HILIC		Application-Specific						Example Applications
			Acclaim 120 C18	Acclaim 120 C8	Acclaim 300 C18	Acclaim Polar Advantage (PA)	Acclaim Polar Advantage II (PA2)	Acclaim Phenyl-1	Acclaim Trinity P1	Acclaim Mixed-Mode WAX-1	Acclaim Mixed-Mode WCX-1	Acclaim Mixed-Mode HILIC-1	Acclaim HILIC-10	Acclaim Organic Acid	Acclaim Surfactant	Acclaim Explosives E1	Acclaim Explosives E2	Acclaim Carbamate	
General Applications	*Neutral Molecules*	High hydrophobicity	√	√	√	√	√	√	√	√	√	√	√						*Fat-soluble vitamins, PAHs, glycerides*
		Intermediate hydrophobicity	√	√	√	√	√	√	√	√	√	√							*Steroids, phthalates, phenolics*
		Low hydrophobicity	√			√	√					√	√						*Acetaminophen, urea, polyethylene glycols*
	Anionic Molecules	High hydrophobicity	√	√	√	√	√	√	√	√	√	√							*NSAIDs, phospholipids*
		Intermediate hydrophobicity	√	√	√	√	√	√	√	√		√							*Asprin, alkyl acids, aromatic acids*
		Low hydrophobicity				√			√	√		√	√						*Small organic acids, e.g. acetic acids*
	Cationic Molecules	High hydrophobicity	√	√	√	√	√	√		√	√	√							*Antidepressants*
		Intermediate hydrophobicity	√	√	√	√	√	√	√		√	√							*Beta blockers, benzidines, alkaloids*
		Low hydrophobicity	√			√			√		√	√	√						*Antacids, pseudoephedrine, amino sugars*
	Amphoteric/ Zwitterionic Molecules	High hydrophobicity	√	√	√	√	√	√	√	√	√	√							*Phospholipids*
		Intermediate hydrophobicity	√	√	√	√	√	√			√								*Amphoteric surfactants, peptides*
		Low hydrophobicity				√	√		√	√	√	√	√						*Amino acids, aspartame, small peptides*
	Mixtures of Neutral, Anionic, Cationic Molecules	Neutrals and acids	√			√	√		√	√									*Artificial sweeteners*
		Neutrals and bases	√			√	√		√		√								*Cough syrup*
		Acids and bases				√			√										*Drug active ingredient with counterion*
		Neutrals, acids, and bases				√			√										*Combination pain relievers*
Specific Applications	*Surfactants*	Anionic	√	√	√	√	√								√				*SDS, LAS, laureth sulfates*
		Cationic													√				*Quats, benzylalkonium in medicines*
		Nonionic	√	√	√	√	√					√			√				*Triton X-100 in washing tank*
		Amphoteric	√	√	√	√	√								√				*Cocoamidopropyl betaine*
		Hydrotropes													√				*Xylenesulfonates in handsoap*
		Surfactant blends													√				*Noionic and anionic surfactants*
	Organic Acids	Hydrophobic							√	√				√					*Aromatic acids, fatty acids*
		Hydrophilic							√	√				√					*Organic acids in soft drinks, pharmaceuticals*
	Environmental Contaminants	Explosives														√	√		*U.S. EPA Method 8330, 8330B*
		Carbonyl compounds															√		*U.S. EPA 1667, 555, OT-11; CA CARB 1004*
		Phenols	√			√													*Compounds regulated by U.S. EPA 604*
		Chlorinated/Phenoxy acids				√													*U.S. EPA Method 555*
		Triazines	√			√													*Compounds regulated by U.S. EPA 619*
		Nitrosamines				√													*Compounds regulated by U.S. EPA 8270*
		Benzidines	√			√													*U.S. EPA Method 605*
		Perfluorinated acids				√													*Dionex TN73*
		Microcystins	√																*ISO 20179*
		Isocyanates					√					√							*U.S. OSHA Methods 42, 47*
		Carbamate insecticides																√	*U.S. EPA Method 531.2*
	Vitamins	Water-soluble vitamins				√	√		√										*Vitamins in dietary supplements*
		Fat-soluble vitamins	√	√	√	√	√	√		√									*Vitamin pills*
	Pharmaceutical Counterions	Anions							√	√									*Inorgaic anions and organic acids in drugs*
		Cations							√		√								*Inorgaic cations and organic bases in drugs*
		Mixture of Anions and Cations							√										*Screening of pharmaceutical counterions*
		API and counterions							√										*Naproxen Na^+ salt, metformin Cl^- salt, etc.*

Polymer Columns

		IonPac AS23	IonPac AS22	IonPac AS22-Fast	IonPac AS14/A	IonPac AS12A	IonPac AS9/HC/SC	IonPac AS4A/SC	IonSwift MAX-100	IonPac AS24	IonPac AS21	IonPac AS20	IonPac AS19	IonPac AS18	IonPac AS18-Fast	IonPac AS17-C	IonPac AS16	IonPac AS15	IonPac AS11(-HC)	IonPac AS10	IonPac AS7	IonPac AS5	IonPac Fast Anion IIIA	OmniPac PAX-100	OmniPac PAX-500	IonPac CS18	IonPac CS17	IonPac CS16	IonPac CS15	IonPac CS14	IonPac CS12A	IonPac CS11	IonPac CS10	IonPac CS5A	OmniPac PCX-100	OmniPac PCX-500	AminoPac PA10	AminoPac PA1	CarboPac PA200	CarboPac PA100	CarboPac PA20	CarboPac PA10	CarboPac PA1	CarboPac MA1	DNAPac PA200	DNAPac PA100	ProPac WAX/SAX	ProPac WCX/SCX	ProPac IMAC	ProPac HIC	ProPac PA1	ProSwift	IonPac ICE-AS6	IonPac ICE-AS1	IonPac ICE-Borate	IonPac NS1
ANIONS	Inorganic Anions	√	√	√	√	√	√	√	√	√		√	√	√	√	√		√	√	√																																						
	Oxyhalides	√				√	√			√			√																																											
	Bromate	√					√			√			√																																											
	Perchlorate										√	√					√																																							
	Organic Acids								√							√		√	√	√																																				
	Phosphoric/Citric Acids																						√																																
	Poly/High-Valence Anions								√			√					√		√		√	√																																
	Hydrophobic Anions								√			√					√		√																																				
	Hydrophobic/Halogenated Anions								√			√							√					√																														
	Anionic Neutral Molecules									√	√	√	√												√																														
CATIONS	Inorganic Cations																										√	√	√	√	√	√	√	√																						
	Sodium/Ammonium																												√	√		√																						
	Amines/Polyvalent Amines																										√	√																										
	Aliphatic/Aromatic Amines																											√				√	√																				
	Alkanol/Ethhanolamines																										√	√		√																								
	Biogenic Amines																										√	√																										
	Transition/Lanthanide Metals																																	√																				
	Hydrophobic Cations																										√	√			√				√																				
	Cationic Neutral Molecules																																			√																				
BIO-MOLECULES	Amino Acids																																				√	√																		
	Phosphorylated Amino Acids																																				√																		
	Amino Sugars																																				√		√	√	√	√												
	Oligoscharides																																						√	√			√	√										
	Mono-/Di-Saccharides																																								√	√	√	√									
	Glycoproteins																																												√	√	√							
	Alditols/Aldoses mono/di Saccharides																																								√	√	√	√									
	ds Nucleic Acids																																						√	√												
	Single-Stranded Oligonucleotides																																						√	√												√
	Peptides																																												√	√	√	√						
	Proteins																																												√	√	√	√	√	√				
	Metal-binding Proteins																																												√									
	Monoclonal antibodies																																								√	√				√			√	√	√	√	√	√	√	√
ORGANIC MOLECULES	Aliphatic Organic Acids																																																					√	√		√
	Alcohols																																																					√	√		
	Borate																																																						√		
	Large Molecules, Anions																																																						√	
	Small Molecules																																																						
	Small Molecules/LC-MS																																																						
	Polar/Non-Polar Small Molecules																																																					
	Hydrophobic/Aliphatic Organic Acids																																																				
	Surfactant Formulations																																																				√
	Explosives/EPA 8330																																																				
MODE	Anion Exchange / Carbonate	√	√	√	√	√	√	√																																													
	Anion Exchange / Hydroxide								√	√	√	√	√	√	√	√	√	√	√	√	√	√	√																																
	Cation Exchange																										√	√	√	√	√	√	√	√																				
	Multi-Mode																								√	√										√	√																
	Affinity																																																					
	Ion Exclusion																																																				
	Reversed Phase																																																				√
	Anion Exchange/Other																																				√	√														

Column Specifications

IC Anion Columns

Column	Format	Primary Eluent	Application	Particle Diameter	Substrate Crosslinking	Latex Diameter	Latex Crosslinking	Capacity (per column)	Functional Group	Hydrophobicity
IonPac AS24	2 × 250 mm	Hydroxide	Recommended column for haloacetic acids prior to MS or MS/MS detection	7 µm	55%	-	-	140 µeq	Alkanol quaternary ammonium	Ultralow
IonPac AS23	2 × 250 mm 4 × 250 mm	Carbonate	Recommended column for inorganic anions and oxyhalides. Trace bromate in drinking water.	6 µm	55%	-	-	80 µeq 320 µeq	Alkyl quaternary ammonium	Ultralow
IonPac AS22	2 × 250 mm 4 × 250 mm	Carbonate	Recommended column for fast analysis of common inorganic anions.	6.5 µm	55%	-	-	52.5 µeq 210 µeq	Alkyl quaternary ammonium	Ultralow
IonPac AS21	2 × 250 mm	Hydroxide	Recommended column for trace perchlorate prior to MS or MS/MS detection	7.0 µm	55%	-	-	45 µeq	Alkanol quaternary ammonium	Ultralow
IonPac AS20	2 × 250 mm 4 × 250 mm	Hydroxide	Recommended column for trace perchlorate prior to suppressed conductivity detection.	7.5 µm	55%	-	-	77.5 µeq 310 µeq	Alkanol quaternary ammonium	Ultralow
IonPac AS19	2 × 250 mm 4 × 250 mm	Hydroxide	Recommended column for inorganic anions and oxyhalides. Trace bromate in drinking water.	7.5 µm	55%	-	-	60 µeq 350 µeq	Alkanol quaternary ammonium	Low
IonPac AS18	2 × 250 mm 4 × 250 mm	Hydroxide	Recommended column for the analysis of common inorganic anions.	7.5 µm	55%	65 nm	8%	75 µeq 285 µeq	Alkanol quaternary ammonium	Low
IonPac AS17-C	2 × 250 mm 4 × 250 mm	Hydroxide	Trace anions in HPW matrices. Carboxylated resin, no sulfate blank. Low capacity for fast analysis of common inorganic anions using gradient elution with the Eluent Generator.	10.5 µm	55%	75 nm	6%	7.5 µeq 30 µeq	Alkanol quaternary ammonium	Low
IonPac AS16	2 × 250 mm 4 × 250 mm	Hydroxide	High capacity for hydrophobic anions including iodide, thiocyanate, thiosulfate, and perchlorate. Polyvalent anions including: polyphosphates and polycarboxylates	9 µm	55%	80 nm	1%	42.5 µeq 170 µeq	Alkanol quaternary ammonium	Ultralow
IonPac AS15	2 × 250 mm 4 × 250 mm	Hydroxide	High capacity for trace analysis of inorganic anions and low molecular weight organic acids in high purity water matrices.	9 µm	55%	-	-	56.25 µeq 225 µeq	Alkanol quaternary ammonium	Medium-High
IonPac AS15-5mm	3 × 150 mm	Hydroxide	Fast run, high capacity for trace analysis of inorganic anions and low molecular weight organic acids in high purity water matrices.	5 µm	55%	-	-	70 µeq	Alkanol quaternary ammonium	Medium-High
IonPac AS14A-5 µm	3 × 150 mm	Carbonate	Recommended column for fast analysis of common inorganic anions.	5 µm	55%	-	-	40 ueq	Alkyl quaternary ammonium	Medium
IonPac AS14A	4 × 250 mm	Carbonate	For analysis of common inorganic anions.	7 µm	55%	-	-	120 µeq	Alkyl quaternary ammonium	Medium
IonPac AS14	2 × 250 mm 4 × 250 mm	Carbonate	Moderate capacity for fast analysis of common inorganic anions.	9 µm	55%	-	-	16 µeq 65 µeq	Alkyl quaternary ammonium	Medium-High

Column	Format	Primary Eluent	Application	Particle Diameter	Substrate Crosslinking	Latex Diameter	Latex Crosslinking	Capacity (per column)	Functional Group	Hydrophobicity
IonPac AS12A	2 × 200 mm 4 × 200 mm	Carbonate	Moderate capacity for analysis of inorganic anions and oxyhalides. Trace chloride and sulfate in high carbonate matrices.	9 µm	55%	140 nm	0.20%	13 µeq 52 µeq	Alkyl quaternary ammonium	Medium
IonPac AS11-HC	2 × 250 mm 4 × 250 mm	Hydroxide	High capacity for the determination of organic acids and inorganic anions in uncharacterized samples.	9 µm	55%	70 nm	6%	72.5 µeq 290 µeq	Alkanol quaternary ammonium	Medium-Low
IonPac AS11	2 × 250 mm 4 × 250 mm	Hydroxide	Low capacity for fast profiling of organic acids and inorganic anions in well-characterized samples.	13 µm	55%	85 nm	6%	11 µeq 45 µeq	Alkanol quaternary ammonium	Very Low
IonPac AS10	2 × 250 mm 4 × 250 mm	Hydroxide	High capacity for the analysis of inorganic anions and organic acids in high nitrate samples.	8.5 µm	55%	65 nm	5%	42.5 µeq 170 µeq	Alkyl quaternary ammonium	Low
IonPac AS9-HC	2 × 250 mm 4 × 250 mm	Carbonate	High-capacity column for inorganic anions and oxyhalides. Trace bromate in drinking water.	9 µm	55%	90 nm	18%	48 µeq 190 µeq	Alkyl quaternary ammonium	Medium-Low
IonPac AS9-SC	4 × 250 mm	Carbonate	Low capacity for fast analysis of inorganic anions and oxyhalides. Specified column in US EPA Method 300.0 (B).	13 µm	55%	110 nm	20%	30-35 µeq	Alkyl quaternary ammonium	Medium-Low
IonPac AS4A-SC	2 × 250 mm 4 × 250 mm	Carbonate	Low capacity for fast analysis of common inorganic anions. Specified column in U.S. EPA Method 300.0 (A).	13 µm	55%	160 nm	0.50%	5 µeq 20 µeq	Alkanol quaternary ammonium	Medium-Low
IonPac Fast Anion IIIA	3 × 250 mm	Hydroxide	Recommended column for phosphoric and citric acids in cola soft drinks.	7.5 µm	55%	-	-	55 µeq	Alkanol quaternary ammonium	Ultralow
IonPac AS7	4 × 250 mm	Specialty Eluents	Polyvalent anions including chelating agents, polyphosphates and polyphosphonates. Cyanide, sulfide, hexavalent chromium, and arsenic speciation.	10 µm	2%	530 nm	5%	100 µeq	Alkyl quaternary ammonium	Medium-High
IonPac AS5A	4 × 150 mm	Hydroxide	Low capacity for fast profiling of organic acids and inorganic anions in well-characterized samples.	5 µm	2%	60 nm	4%	35 µeq	Alkanol quaternary ammonium	Low
IonPac AS5	4 × 250 mm	Hydroxide	Metal-EDTA complexes, metal-cyanide complexes, and oxyanions.	15 µm	2%	120 nm	1%	20 µeq	Alkanol quaternary ammonium	Low

IC Cation Columns

Column	Format	Primary Eluent	Application	Particle Diameter	Substrate Crosslinking	Latex Diameter	Latex Crosslinking	Capacity (per column)	Functional Group	Hydrophobicity
IonPac CS18	2 × 250 mm	MSA	Recommended column for polar amines (alkanolamines and methylamines) and moderately hydrophobic and polyvalent amines (biogenic and diamines). Nonsuppressed mode when extended calibration linearity for ammonium and weak bases is required	6 µm	55%	-	-	0.29 µeq	Carboxylic acid	Medium
IonPac CS17	2 × 250 mm 4 × 250 mm	MSA	Recommended column for hydrophobic and polyvalent amines (biogenic amines and diamines)	7 µm	55%	-	-	0.363 µeq 1.45 µeq	Carboxylic acid	Very Low
IonPac CS16	3 × 250 mm 5 × 250 mm	MSA	Recommended column for disparate concentration ratios of adjacent-eluting cations such as sodium and ammonium. Can be used for alkylamines and alkanolamines.	5 µm	55%	-	-	3.0 µeq 8.4 µeq	Carboxylic acid	Medium
IonPac CS15	2 × 250 mm 4 × 250 mm	MSA	Disparate concentration ratios of ammonium and sodium. Trace ethanolamine in high-ammonium or high- potassium concentrations. Alkanolamines.	8.5 µm	55%	-	-	0.7 µeq 2.8 µeq	Carboxylic acid/ phosphonic acid/ crown ether	Medium
IonPac CS14	2 × 250 mm 4 × 250 mm	MSA	Aliphatic amines, aromatic amines, and polyamines plus mono- and divalent cations.	8.5 µm	55%	-	-	0.325 µeq 1.3 µeq	Carboxylic acid	Low
IonPac CS12A-MS	2 × 100 mm	MSA	IC-MS screening column for fast elution and low flow rates required for interfacing with IC-MS	8.5 µm	55%	-	-	0.28 µeq	Carboxylic acid/ phosphonic acid	Medium
IonPac CS12A-5 µm	3 × 150 mm	MSA	Recommended column for high efficiency and fast analysis (3 min) of mono- and divalent cations.	5 µm	55%	-	-	0.94 µeq	Carboxylic acid/ phosphonic acid	Medium
IonPac CS12A	2 × 250 mm 4 × 250 mm	MSA	Recommended column for the separation of mono- and divalent cations. Manganese morpholine, alkylamines, and aromatic amines.	8.5 µm	55%	-	-	0.7 µeq 2.8 µeq	Carboxylic acid/ phosphonic acid	Medium
IonPac CS11	2 × 250 mm	HCl + DAP	Separation of mono- and divalent cations. Ethanolamines if divalent cations are not present.	8 µm	55%	200 nm	5%	0.035 µeq	Sulfonic acid	Medium
IonPac CS10	4 × 250 mm	HCl + DAP	Separation of mono- and divalent cations.	8.5 µm	55%	200 nm	5%	0.08 µeq	Sulfonic acid	Medium
IonPac CS5A	2 × 250 mm 4 × 250 mm	Pyridine dicarboxylic acid	Recommended column for transition and lanthanide metals analysis. Aluminum analysis.	9 µm	55%	140 nm 75 nm	10% 20%	0.02 µeq/ 0.005 µeq 0.04 µeq/ 0.01 µeq	Sulfonic acid/ alkanol quaternary ammonium	-

Transferring HPLC Methods to UHPLC

Environmental Water Applications Notebook

Easy Method Transfer from HPLC to RSLC with the Dionex Method Speed-Up Calculator

INTRODUCTION

The goal of every chromatographic optimization is a method that sufficiently resolves all peaks of interest in as short a time as possible. The evolution of packing materials and instrument performance has extended chromatographic separations to new limits: ultrahigh-performance liquid chromatography (UHPLC).

The new Dionex UltiMate® 3000 Rapid Separation LC (RSLC) system is ideal for ultrafast, high-resolution LC. The RSLC system was designed for ultrafast separations with flow rates up to 5 mL/min at pressures up to 800 bar (11,600 psi) for the entire flow-rate range. This industry-leading flow-pressure footprint ensures the highest flexibility possible; from conventional to ultrahigh-resolution to ultrahigh-speed methods. The RSLC system, with autosampler cycle times of only 15 seconds, oven temperatures up to 110 °C, and data collection rates up to 100 Hz (even when acquiring UV-Vis spectra), sets the standard for UHPLC performance. Acclaim® RSLC columns with a 2.2 µm particle size complete the RSLC dimension.

A successful transfer from an HPLC method to an RSLC method requires recalculation of the chromatographic parameters. Underlying chromatographic principles have to be considered to find the appropriate parameters for a method transfer. With the Method Speed-up Calculator, Dionex offers an electronic tool that streamlines the process of optimum method transfer. This technical note describes the theory behind the Method Speed-Up Calculator and the application of this interactive, multi-language tool, illustrated with an exemplary method transfer from a conventional LC separation to an RSLC separation. You may obtain a copy of this calculator from your Dionex representative.

METHOD SPEED-UP STRATEGY

The purpose of method speed-up is to achieve sufficient resolution in the shortest possible time. The strategy is to maintain the resolving power of the application by using shorter columns packed with smaller particles. The theory for this approach is based on chromatographic mechanisms, found in almost every chromatography text book. The following fundamental chromatographic equations are applied by the Method Speed-Up Calculator for the method transfer from conventional to ultrafast methods.

The separation efficiency of a method is stated by the peak capacity *P*, which describes the number of peaks that can be resolved in a given time period. The peak capacity is defined by the run time divided by the average peak width. Hence, a small peak width is essential for a fast method with high separation efficiency. The peak width is proportional to the inverse square root of the number of theoretical plates N generated by the column. Taking into account the length of the column, its efficiency can also be expressed by the height equivalent to a theoretical plate *H*. The relationship between plate height H and plate number *N* of a column with the length L is given by Formula 1.

Formula 1: $$N = \frac{L}{H}$$

Low height equivalents will therefore generate a high number of theoretical plates, and hence small peak width for high peak capacity is gained. Which factors define *H*? For an answer, the processes inside the column have to be considered, which are expressed by the Van Deemter equation (Formula 2).

Formula 2: $$H = A + \frac{B}{u} + C \cdot u$$

The Eddy diffusion *A* describes the mobile phase movement along different random paths through the stationary phase, resulting in broadening of the analyte band. The longitudinal diffusion of the analyte against the flow rate is expressed by the term *B*. Term *C* describes the resistance of the analyte to mass transfer into the pores of the stationary phase. This results in higher band broadening with increasing velocity of the mobile phase. The well-known Van Deemter plots of plate height *H* against the linear velocity of the mobile phase are useful in determining the optimum mobile phase flow rate for highest column efficiency with lowest plate heights. A simplification of the Van Deemter equation, according to Halász[1] (Formula 3), describes the relationship between column efficiency (expressed in plate height *H*), particle size d_p (in µm) and velocity of mobile phase *u* (in mm/s):

Formula 3: $$H = 2 \cdot d_p + \frac{6}{u} + \frac{d_p^2 \cdot u}{20}$$

The plots of plate height H against velocity u depending on the particle sizes dp of the stationary phase (see Figure 1, top) demonstrate visually the key function of small particle sizes in the method speed-up strategy: The smaller the particles, the smaller the plate height and therefore the better the separation efficiency. An efficiency equivalent to larger particle columns can be achieved by using shorter columns and therefore shorter run times.

Another benefit with use of smaller particles is shown for the 2 µm particles in Figure 1: Due to improved mass transfer with small particle packings, further acceleration of mobile phases beyond the optimal flow rate with minimal change in the plate height is possible.

Optimum flow rates and minimum achievable plate heights can be calculated by setting the first derivative of the Halász equation to zero. The optimal linear velocity (in mm/s) is then calculated by Formula 4.

Formula 4: $$u_{opt} = \sqrt{\frac{B}{C}} = \frac{10.95}{d_p}$$

The minimum achievable plate height as a function of particle size is calculated by insertion of Formula 4 in Formula 3, resulting in Formula 5.

Formula 5: $$H_{min} \approx 3 \cdot d_p$$

Chromatographers typically prefer resolution over theoretical plates as a measure of the separation quality. The achievable resolution *R* of a method is directly proportional to the square root of the theoretical plate number as can be seen in Formula 6. *k* is the retention factor of the analyte and *k* the selectivity.

Formula 6: $$R = \frac{1}{4} \cdot \sqrt{N} \cdot \frac{k_2}{1 + k_2} \cdot \frac{\alpha - 1}{\alpha}$$

If the column length is kept constant and the particle size is decreased, the resolution of the analytes improves. Figure 1, bottom, demonstrates this effect using 5 µm and 2 µm particles.

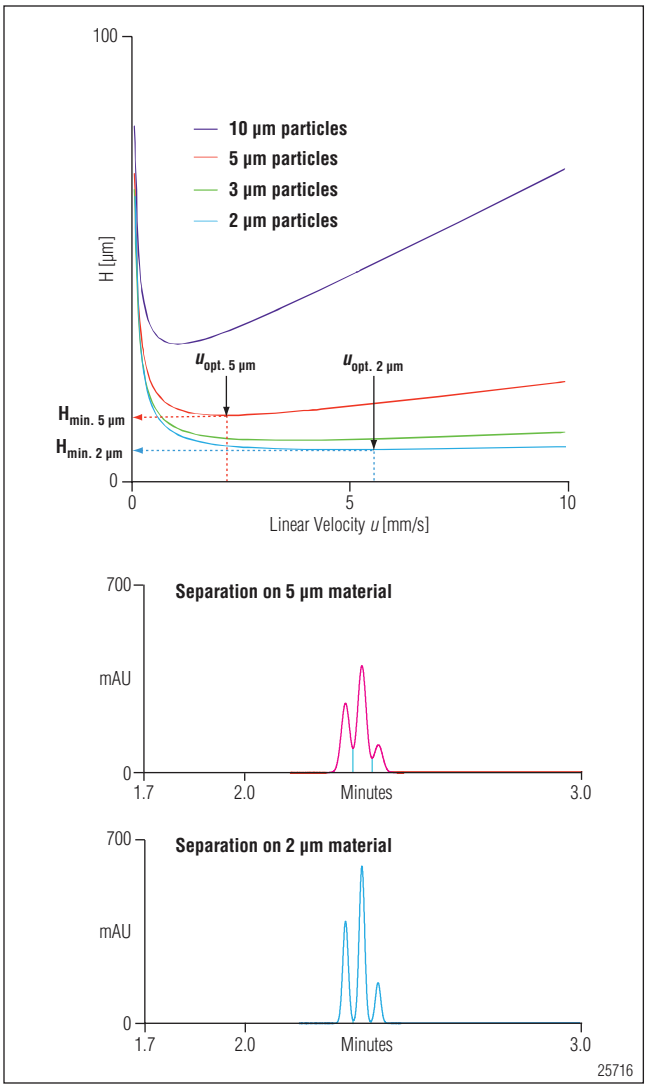

Figure 1. Smaller particles provide more theoretical plates and more resolution, demonstrated by the improved separation of three peaks (bottom) and smaller minimum plate heights H *in the Van Deemter plot (top). At linear velocities higher than uopt,* H *increases more slowly when using smaller particles, allowing higher flow rates and therefore faster separations while keeping separation efficiency almost constant. The speed-up potential of small particles is revealed by the Van Deemter plots (top) of plate height* H *against linear velocity u of mobile phase: Reducing the particle size allows higher flow rates and shorter columns because of the decreased minimum plate height and increased optimum velocity. Consequently, smaller peak width and improved resolution are the result (bottom).*

When transferring a gradient method, the scaling of the gradient profile to the new column format and flow rate has to be considered to maintain the separation performance. The theoretical background was introduced by L. Snyder[2] and is known as the gradient volume principle. The gradient volume is defined as the mobile phase volume that flows through the column at a defined gradient time t_G. Analytes are considered to elute at constant eluent composition. Keeping the ratio between the gradient volume and the column volume constant therefore results in a correct gradient transfer to a different column format.

Taking into account the changed flow rates F and column volume (with diameter d_c and length L), the gradient time intervals t_G of the new methods are calculated with Formula 7.

Formula 7: $$t_{G,new} = t_{G,old} \cdot \frac{F_{old}}{F_{new}} \cdot \frac{L_{new}}{L_{old}} \cdot \left(\frac{d_{c,new}}{d_{c,old}} \right)^2$$

An easy transfer of method parameters can be achieved by using the Dionex Method Speed-Up Calculator (Figure 2), which incorporates all the overwhelming theory and makes manual calculations unnecessary. This technical note describes the easy method transfer of an example separation applying the calculator. Just some prerequisites described in the following section have to be taken into account.

PREREQUISITES

The Method Speed-Up Calculator is a universal tool and not specific for Dionex products. Nevertheless, some prerequisites have to be considered for a successful method transfer, which is demonstrated in this technical note by the separation of seven soft drink additives.

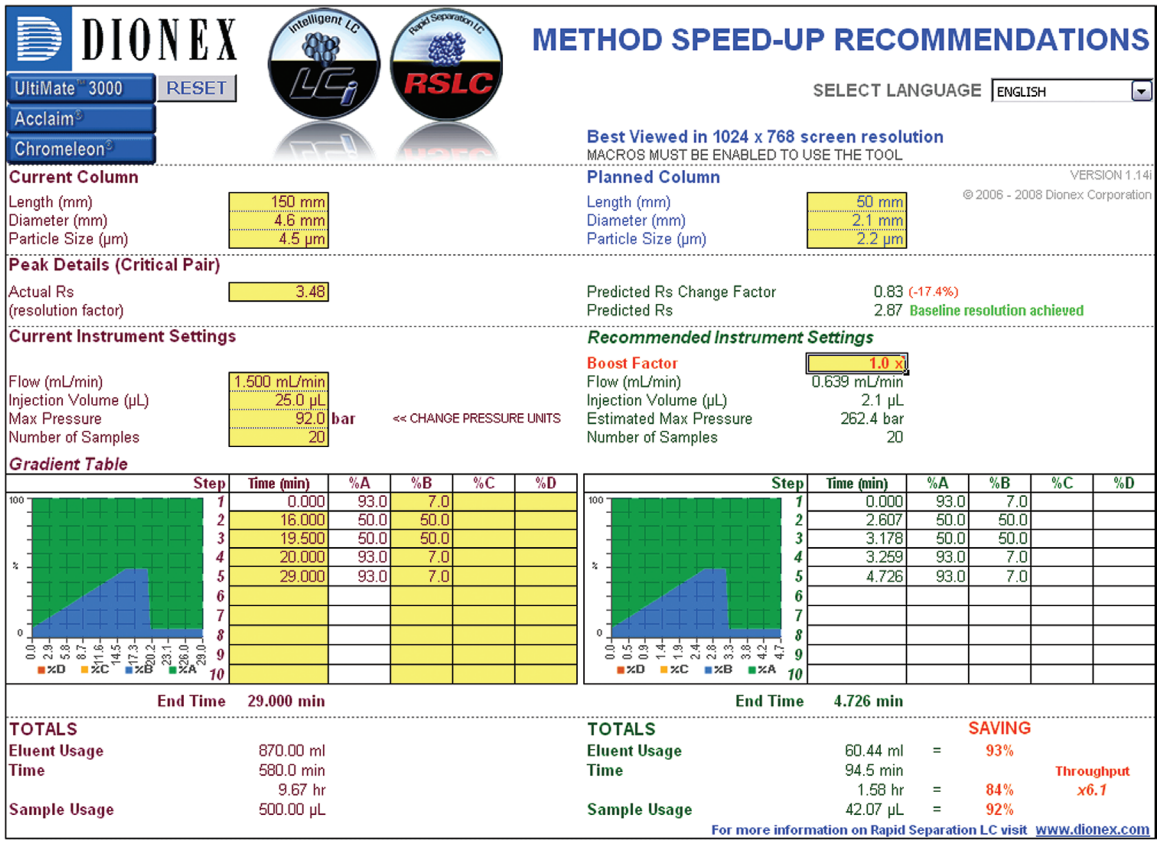

Figure 2. The Dionex Method Speed-Up Calculator transfers a conventional (current) HPLC method to a new (planned) RSLC method.

Column Dimension

First, the transfer of a conventional method to an RSLC method requires the selection of an adequate column filled with smaller particles. The RSLC method is predicted best if the selectivity of the stationary phase is maintained. Therefore, a column from the same manufacturer and with nominally identical surface modification is favoured for an exact method transfer. If this is not possible, a column with the same nominal stationary phase is the best choice. The separation is made faster by using shorter columns, but the column should still offer sufficient column efficiency to allow at least a baseline separation of analytes. Table 1 gives an overview of the theoretical plates expected by different column length and particle diameter size combinations using Dionex Acclaim column particle sizes. Note that column manufacturers typically fill columns designated 5 µm with particle sizes 4–5 µm. Dionex Acclaim 5 µm columns are actually filled with 4.5 µm particles. This is reflected in the table.

Table 1. Theoretical Plates Depending on Column Length and Particle Diameter (Calculated Using Formula 5)

	Theoretical Plates N		
Particle size	4.5 µm	3 µm	2.2 µm
Column length: 250 mm	18518	27778	37879
150 mm	11111	16667	22727
100 mm	7407	11111	15152
75 mm	5555	8333	11364
50 mm	3703	5556	7576

If the resolution of the original separation is higher than required, columns can be shortened. Keeping the column length constant while using smaller particles improves the resolution. Reducing the column diameter does not shorten the analysis time but decreases mobile phase consumption and sample volume. Taking into account an elevated temperature, smaller column inner diameters reduce the risk of thermal mismatch.

System Requirements

Smaller particles generate higher backpressure. The linear velocity of the mobile phase has to be increased while decreasing the particle size to work within the Van Deemter optimum. The UltiMate 3000 RSLC system perfectly supports this approach with its high maximum operation pressure of 800 bar (11,600 psi). This maximum pressure is constant over the entire flow rate range of up to 5 mL/min, providing additional potential to speed up applications even further by increasing the flow rate.

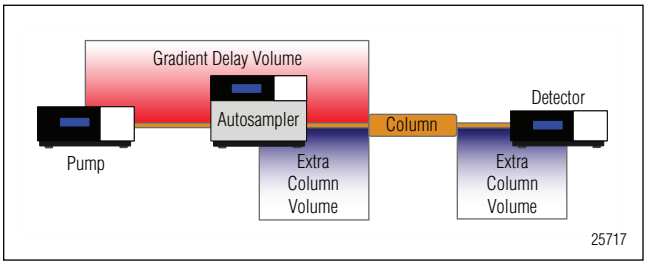

Figure 3. Gradient delay volume and extra column volume of an HPLC system. Both play an important role in method speed-up.

For fast gradient methods, the gradient delay volume (GDV) plays a crucial role. The GDV is defined as the volume between the first point of mixing and the head of the column. The GDV becomes increasingly important with fast, steep gradients and low flow rate applications as it affects the time taken for the gradient to reach the head of the column. The larger the GDV, the longer the initial isocratic hold at the beginning of the separation. Typically, this leads to later peak elution times than calculated. Early eluting peaks are affected most. In addition, the GDV increases the time needed for the equilibration time at the end of a sample and therefore increases the total cycle time. A general rule is to keep the gradient steepness and the ratio of GDV to column volume constant when transferring a standard method into a fast LC method. This will maintain the selectivity of the original method.[3]

The GDV can be adjusted to the column volume by installing appropriate mixer kits to the RSLC pump (see Table 2), which contributes most to the GDV. Typically, 100 µL or 200 µL mixers are good starting points when operating a small volume column in an RSLC system.

Another option is to switch the sample loop of the split-loop autosampler out of the flow path. The GDV is then reduced by the sample loop volume in the so-called bypass mode. The GDV of a standard sample loop of the RSLC autosampler is 150 µL, the micro injection loop has a 50 µL GDV.

Table 2. Mixer Kits Available for UltiMate 3000 RSLC System to Adapt GDV of Pump

Mixer Kit	GDV pump
Mixer kit 6040.5000	35 µL
Static mixer kit 6040.5100	100 µL
Static mixer kit 6040.5150	200 µL

Besides the gradient delay volume, the extra column volume is an important parameter for fast LC methods. The extra column volume is the volume in the system through which the sample passes and hence contributes to the band broadening of the analyte peak (Figure 3). The extra column volume of an optimized LC system should be below $^{1}/_{10}$th of the peak volume. Therefore the length and inner diameter of the tubing connections from injector to column and column to detector should be as small as possible. Special care has to be taken while installing the fittings to avoid dead volumes. In addition, the volume of the flow cell has to be adapted to the peak volumes eluting from the RSLC column. If possible, the flow cell detection volume should not exceed $^{1}/_{10}$th of the peak volume.

Detector Settings

When transferring a conventional method to an RSLC method, the detector settings have a significant impact on the detector performance. The data collection rate and time constant have to be adapted to the narrower peak shapes. In general, each peak should be defined by at least 30 data points. The data collection rate and time constant settings are typically interrelated to optimize the amount of data points per peak and reduce short-term noise while still maintaining peak height, symmetry, and resolution.

The Chromeleon® Chromatography Management Software has a wizard to automatically calculate the best settings, based on the input of the minimum peak width at half height of the chromatogram. This width is best determined by running the application once at maximum data rate and shortest time constant. The obtained peak width may then be entered into the wizard for optimization of the detection settings. Refer to the detector operation manual for further details.

METHOD SPEED-UP USING THE CALCULATOR

Separation Example

Separation was performed on an UltiMate 3000 RSLC system consisting of a HPG-3200RS Binary Rapid Separation Pump, a WPS-3000RS Rapid Separation Well Plate Sampler with analytical sample loop (100 µL), a TCC-3000RS Rapid Separation Thermostatted Column Compartment with precolumn heater (2 µL), and a VWD-3400RS Variable Wavelength Detector with semi-micro flow cell (2.5 µL). Chromeleon Chromatography Management Software (version 6.80, SR5) was used for both controlling the instrument and reporting the data. The modules were connected with stainless steel micro capillaries, 0.01" ID, $^{1}/_{16}$" OD when applying the conventional LC method, 0.007" and 0.005" ID, $^{1}/_{16}$" OD when applying the RSLC methods. A standard mixture of seven common soft drink additives was separated by gradient elution at 45 °C on two different columns:

- Conventional HPLC Column: Acclaim 120, C18, 5 µm, 4.6 × 150 mm column, (P/N 059148)
- Rapid Separation Column: Acclaim RSLC 120, C18, 2.2 µm, 2.1 × 50 mm column (P/N 068981).

The UV absorbance wavelength at 210 nm was recorded at 5 Hz using the 4.6 × 150 mm column and at 25 Hz and 50 Hz using the 2.1 × 50 mm column. Further method details such as flow rate, injection volume, and gradient table of conventional and RSLC methods are described in the following section. The parameters for the method transfer were calculated with the Dionex Method Speed-Up Calculator (version 1.14i).

The conventional separation of seven soft drink additives is shown in Figure 4A. With the Method Speed-Up Calculator, the method was transferred successfully to RSLC methods (Figure 4B and C) at two different flow rates. The easy method transfer with this universal tool is described below.

Column Selection for Appropriate Resolution

The column for method speed-up must provide sufficient efficiency to resolve the most critical pairs. In this example, separating peaks 5 and 6 is most challenging. A first selection of the planned column dimensions can be made by considering the theoretical plates according to Table 1. The 4.6 × 150 mm, 5 µm column is actually filled with 4.5 µm particles. Therefore, it provides 11,111 theoretical plates. On this column, the

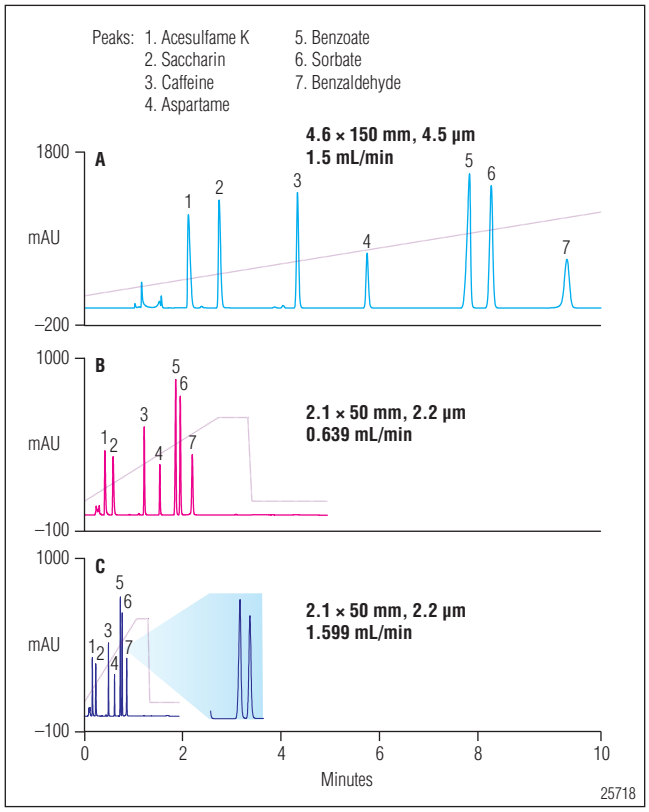

Figure 4. Method transfer with the Method Speed-Up Calculator from A) a conventional LC separation on an Acclaim 5 µm particle column, to B) and C) RSLC separations on an Acclaim 2.2 µm particle column.

resolution is $R_{(5,6)}$=3.48. This resolution is sufficiently high to select a fast LC column with fewer theoretical plates for the speed up. Therefore, a 2.1 × 50 mm, 2.2 µm column with 7579 plates was selected.

The first values to be entered into the yellow field of the Method Speed-Up Calculator are the current column dimension, planned column dimension, and the resolution of the critical pair. To obtain the most accurate method transfer, use the particle sizes listed in the manufacturer's column specifications sheet instead of the nominal size, which may be different. Dionex Acclaim columns with a nominal particle size of 5 µm are actually filled with 4.5 µm particles, and this value should be used to achieve a precise method transfer calculation. This has a positive impact on the performance and pressure predictions for the planned column. Based on the assumption of unchanged stationary phase chemistry, the calculator then predicts the resolution provided by the new method (Figure 5).

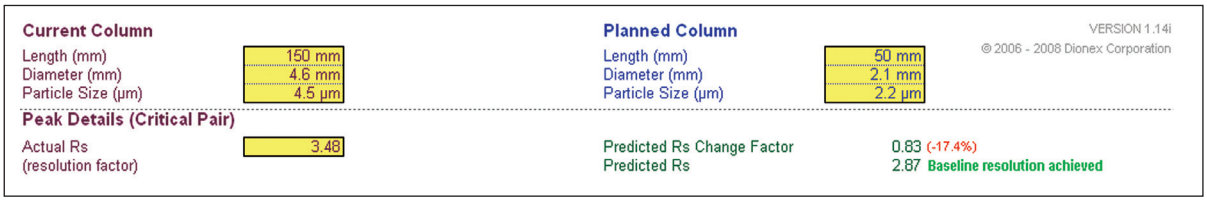

Figure 5. Column selection considering the resolution of the critical pair.

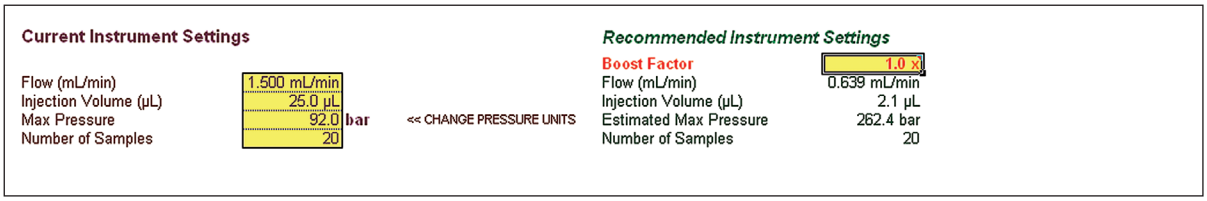

Figure 6. The flow rate, injection volume and backpressure of the current method are scaled to the new column dimension.

In the example in Figure 5, the predicted resolution between benzoate and sorbate is 2.87. With a resolution of $R \geq 1.5$, the message "Baseline resolution achieved" pops up. This indicates that a successful method transfer with enough resolution is possible with the planned column. If R is smaller than 1.5, the red warning "Baseline is not resolved" appears. Note that the resolution calculation is performed only if the boost factor BF is 1, otherwise it is disabled. The function of the boost factor is described in the Adjust Flow Rate section.

Instrument Settings

The next section of the Method Speed-Up Calculator considers basic instrument settings. These are flow rate, injection volume, and system backpressure of the current method (Figure 6). In addition to these values, the detector settings have to be considered as described in the earlier section "Detector Settings". Furthermore, the throughput gain with the new method can be calculated if the number of samples to be run is entered.

Adjust Flow Rate

As explained by Van Deemter theory, smaller particle phases need higher linear velocities to provide optimal separation efficiency. Consequently, the Dionex Method Speed-Up Calculator automatically optimizes the linear velocity by the ratio of particle sizes of the current and planned method. In addition, the new flow rate is scaled to the change of column cross section if the column inner diameter changed. This keeps the linear velocity of the mobile phase constant. A boost factor (*BF*) can be entered to multiply the flow rate for a further decrease in separation time. If the calculated resolution with $BF=1$ predicts sufficient separation, the method can be accelerated by increasing the boost factor and therefore increasing the flow rate. Figure 1 shows that applying linear velocities beyond the optimum is no problem with smaller particle phases, as they do not significantly loose plates in this region. Note that the resolution calculation of the Method Speed-Up Calculator is disabled for $BF \neq 1$.

For the separation at hand, the flow rate is scaled from 1.5 mL/min to 0.639 mL/min when changing from an Acclaim 4.6 × 150 mm, 4.5 µm column to a 2.1 × 50 mm, 2.2 µm column (see Figure 6), adapting the linear velocity to the column dimensions and the particle size. The predicted resolution between peak 5 and 6 for the planned column is $R=2.87$. The actual resolution achieved is $R=2.91$, almost as calculated (chromatogram B in Figure 4).

A Boost Factor of 2.5 was entered for further acceleration of the method (Figure 7). The method was then performed with a flow rate of 1.599 mL/min, and resolution of the critical pair was still sufficient at $R=2.56$ (see zoom in chromatogram C in Figure 4).

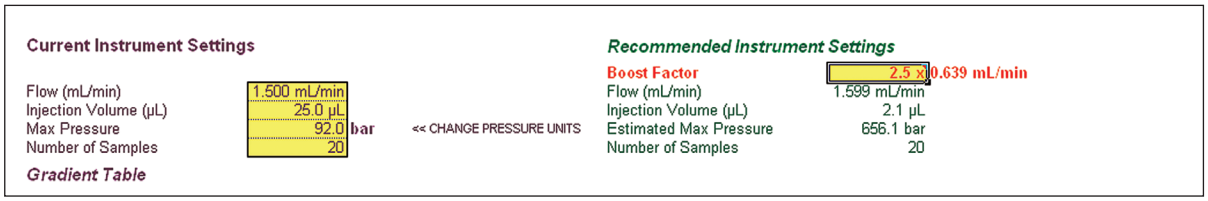

Figure 7. The new flow rate is further accelerated by applying the Boost Factor of 2.5.

Scale Injection Volume

The injection volume has to be adapted to the new column dimension to achieve similar peak heights by equivalent mass loading. Therefore the injection plug has to be scaled to the change of column cross section. In addition, shorter columns with smaller particles cause a reduced zone dilution. Consequently, sharper peaks compared to longer columns are expected. The new injection volume $V_{inj,new}$ is then calculated by Formula 8, taking a changed cross section and reduced band broadening by changed particle diameter into account.

Formula 8: $$V_{inj,new} = V_{inj,old} \cdot \left(\frac{d_{c,new}}{d_{c,old}}\right)^2 \cdot \sqrt{\frac{L_{new} \cdot d_{p,new}}{L_{old} \cdot d_{p,old}}}$$

Generally, it is recommended that a smaller flow cell be used with the RSLC method to minimize the extra column volume. Also, the difference in path length of different flow cell sizes has to be taken into account while scaling the injection volume. In the example of the soft drink analysis, the injection volume is scaled from 25 µL to 2.1 µL when replacing the Acclaim 4.6 × 150 mm, 4.5 µm column with a 2.1 × 50 mm, 2.2 µm column (see Figure 6).

Predicted Backpressure

Speeding-up the current method by decreasing particle size and column diameter and increasing flow rate means elevating the maximum generated backpressure. The pressure drop across a column can be approximated by the Kozeny-Carman formula.[4] The pressure drop of the new method is predicted by the calculator considering changes in column cross section, flow rate, and particle size and is multiplied by the boost factor. The viscosity of mobile phase is considered constant during method transfer. The calculated pressure is only an approximation and does not take into account nominal and actual particle size distribution depending on column manufacturer. If the predicted maximum pressure is above 800 bar (11,600 psi) the warning "Exceeds pressure limit RSLC" is shown, indicating the upper pressure limit of the UltiMate 3000 RSLC system. However, in the case the method is transferred to a third party system, its pressure specification has to be considered.

In the example of the soft drink analysis, the actual pressure increases from 92 bar to 182 bar with *BF*=1 on the 2.1 × 50 mm column, and to 460 bar for the RSLC method with *BF*=2.5. The pressures predicted by the Method Speed-Up Calculator are 262 bar and 656 bar, respectively. The pressure calculation takes into account the change of the size of the column packing material. In a speed up situation, the pressure is also influenced by other factors such as particle size distribution, system fluidics pressure, change of flow cell, etc. When multiplication factors such as the boost factor are used, the difference between calculated and real pressure is pronounced. The pressure calculation is meant to give an orientation, what flow rates might be feasible on the planned column. However, it should be confirmed by applying the flow on the column.

Adapt Gradient Table

The gradient profile has to be adapted to the changed column dimensions and flow rate following the gradient-volume principle. The gradient steps of the current method are entered into the yellow fields of the gradient table. The calculator then scales the gradient step intervals appropriately and creates the gradient table of the new method.

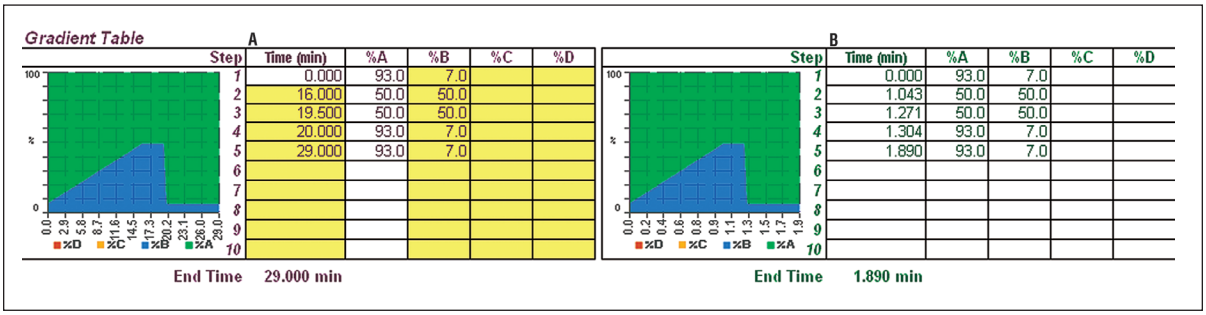

Figure 8. The gradient table of the current method (A) is adapted to the boosted method (B) according to the gradient-volume principle.

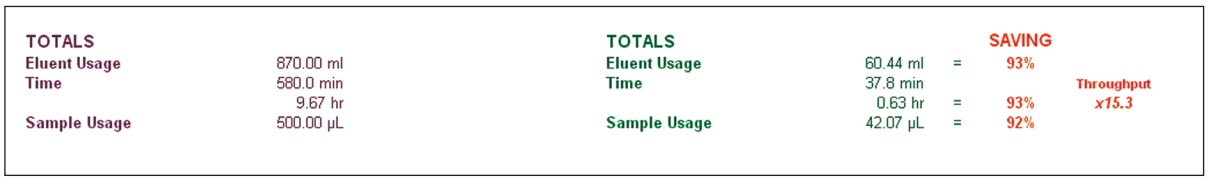

Figure 9. The absolute values for analysis time, eluent usage, and sample usage of the current (purple) and planned (green) method are calculated by the Method Speed-Up Calculator. The savings of eluent, sample, and time due to the method transfer are highlighted.

The adapted gradient table for the soft drink analysis while using a boost factor *BF*=1 is shown in Figure 8. According to the gradient-volume principle, the total run time is reduced from 29.0 min to 4.95 min by taking into account the changed column volume from a 4.6 × 150 mm, 5 µm (4.5 µm particles entered) to a 2.1 × 50 mm, 2.2 µm column and the flow rate reduction from 1.5 mL/min to 0.639 mL/min. The separation time was further reduced to 1.89 min by using boost factor *BF*=2.5. Gradient time steps were adapted accordingly. The comparison of the peak elution order displayed in Figure 4 shows that the separation performance of the gradient was maintained during method transfer.

Consumption and Savings

Why speed-up methods? To separate analyte peaks faster and at the same time reduce the mobile phase and sample volume consumption. Those three advantages of a method speed-up are indicated in the Method Speed-Up Calculator sheet right below the gradient table. The absolute values for the time, eluent, and sample usage are calculated taking the numbers of samples entered into the current instrument settings section of the calculation sheet into account (see Figure 6).

Regarding the soft drink analysis example, geometrical scaling of the method from the conventional column to the RSLC method means saving 93% of eluent and 92% of sample. The sample throughput increases 6.1-fold using *BF*=1. The higher flow rate at *BF*=2.5 results in a 15.3-fold increased throughput compared to the conventional LC method (Figure 9).

CONCLUSION

Fast method development or increased sample throughput are major challenges of most analytical laboratories. A systematic method speed-up is accomplished by reducing the particle size, shortening the column length, and increasing the linear velocity of the mobile phase. The Dionex Method Speed-Up Calculator automatically applies these rules and scales the conventional LC parameters to the conditions of the RSLC method. The interactive electronic tool is universally applicable. New instrument settings are predicted and gradient tables are adapted for optimum performance for the new method. The benefit of the method transfer is summarized by the integrated calculation of savings in time, eluent and sample. In addition, users can benefit from getting results earlier and thereby reducing the time to market. The Dionex Method Speed-Up Calculator is part of Dionex's total RSLC solution, which further consists of the industry leading UltiMate 3000 RSLC system, powerful Chromeleon Chromatography Management Software, and high-efficiency Acclaim RSLC columns.

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