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

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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 byproduct 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 byproduct (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 Byproducts (D/DBP) Rule.⁶ The European Union also reduced their regulatory value for bromate from 50 to 10 µg/L⁷, 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 a Thermo Scientific[™] Dionex[™] 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 a Dionex 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 EPA 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 Ion chromatography (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 a Dionex IonPac AS9-HC column with a carbonate eluent to determine trace bromate and other disinfection byproduct 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 Dionex 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 Dionex IonPac 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-ug/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

- Thermo Scientific Dionex ICS-2500 Reagent-Free[™] Ion Chromatography (RFIC[™]) system:
 - 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
- Thermo Scientific Dionex EluGen EGC II KOH Cartridge (P/N 058900)
- Thermo Scientific Dionex CR-ATC Continuously Regenerated Anion Trap Column (P/N 060477)
- PC10 Pneumatic Postcolumn Delivery module (P/N 050601)
- PCH-2 Postcolumn Reaction Heater (P/N 039348)
- Knitted Reaction Coil, 500 µL, potted (for PCH-2) (P/N 039349)
- Two 4 L plastic bottle assemblies (for external water mode of suppression)
- Thermo Scientific Dionex Chromeleon[™] Chromatography Workstation

Reagents and Standards

- Deionized water, Type I reagent grade, 18 M Ω -cm resistivity or better
- *o*-Dianisidine, dihydrochloride salt (ODA, 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,, Fluka 71388, 80% pure)
- Sodium bromate (NaBrO₃, EM SX 03785-1)
- Sodium chlorate (NaClO₃, Fluka 71370)

Conditions						
Columns:	Dionex IonPac AS19 Analytical, 4×250 mm (P/N 062885) Dionex IonPac AG19 Guard, 4×50 mm (P/N 062887)					
Eluent:	10 mM KOH from 0–10 min, 10–45 mM from 10–25 min*					
Eluent Source:	Dionex EG50 with Dionex CR-ATC					
Flow Rate:	1.0 mL/min					
Temperature:	30 °C					
Injection:	250 µL					
Detection:	Suppressed conductivity, Thermo Scientific Dionex ASRS [™] ULTRA II Suppressor, 4 mm (P/N 061561), auto-suppression, external water mode, 130 mA current					
Background Conductance:	<1 µ\$					
System Backpressure:	~2200 psi					
Run Time:	30 min					
PCR						
Detection:	Absorbance at 450 nm (tungsten lamp)					
Postcolumn Reagent Flow:	0.54 mL/min					
Postcolumn Hea Temperature:	ater 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. 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 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 dilute to volume with DI water. Prepare a separate secondary stock standard containing bromate only at 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			

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 μ L 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 $0.45 \mu m$ syringe filters, discarding the first 300 μ L of the effluent. To prevent degradation of chlorite or the formation of bromate from hypobromous acid/hypobromite, preserve the samples by adding 50 μ L of EDA preservation solution per 100 mL of sample.

System Preparation and Setup

Prepare the Dionex ASRS ULTRA II suppressor 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 Dionex ASRS ULTRA II suppressor for use in the external water mode by connecting the Regen Out port of the suppressor to the Regen In port of the Dionex CR-ATC column. The Regen In port of the suppressor should connect directly to the external water source. The Regen Out port of the Dionex CR-ATC column is then connected to 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 Dionex EGC II KOH cartridge in the EG50 eluent generator and configure it with the Dionex CR-ATC column according to the Dionex CR-TC Quickstart (Document No. 031911). Use the Dionex Chromeleon system configuration to set up the Dionex EGC II KOH cartridge with the software. Condition the cartridge as directed by the Dionex EGC II Quickstart (Document No. 031909) with 50 mM KOH at 1 mL/min



Figure 1. System configuration for EPA Method 317.0.

for 30 min. Install a 4×50 mm Dionex IonPac 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 Dionex 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 Dionex 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 Dionex 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 (for example, 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.

Results and Discussion

U.S. EPA Method 317.0 specifies the use of a Dionex 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 $0.71 \mu g/L$ for a 225 μL injection by suppressed conductivity and 0.12 $\mu g/L$ by visible absorbance (225 μL injection).



Figure 2. Separation of low ppb DBP anions and bromide using the Dionex IonPac AS19 column.

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 Dionex IonPac AS19 column and detection by suppressed conductivity.¹⁸ In this application note, we examine the feasibility of using the Dionex 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 byproduct 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,

Table 2. Linearity and MDLs for disinfection byproduct anions and bromide.

Analyte	Range (µg/L)	Linearity (r²)	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

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 Dionex IonPac 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 Dionex IonPac 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 POL of 0.5 µg/L, comparable to that reported in Method 317.0 using the Dionex IonPac 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.

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

	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< td=""><td>5.0</td><td>97.0</td><td><mdl< td=""><td>5.0</td><td>94.6</td><td><mdl< td=""><td>5.0</td><td>97.5</td><td><mdl< td=""><td>5.0</td><td>104.5</td></mdl<></td></mdl<></td></mdl<></td></mdl<>	5.0	97.0	<mdl< td=""><td>5.0</td><td>94.6</td><td><mdl< td=""><td>5.0</td><td>97.5</td><td><mdl< td=""><td>5.0</td><td>104.5</td></mdl<></td></mdl<></td></mdl<>	5.0	94.6	<mdl< td=""><td>5.0</td><td>97.5</td><td><mdl< td=""><td>5.0</td><td>104.5</td></mdl<></td></mdl<>	5.0	97.5	<mdl< td=""><td>5.0</td><td>104.5</td></mdl<>	5.0	104.5
Bromate (conductivity)	2.5	3.0	103.3	10.0	10.0	95.7	<mdl< td=""><td>1.01</td><td>10.5</td><td><mdl< td=""><td>1.0</td><td>103.4</td></mdl<></td></mdl<>	1.01	10.5	<mdl< td=""><td>1.0</td><td>103.4</td></mdl<>	1.0	103.4
Bromate (Vis)	2.2	3.0	96.3	10.1	10.0	102.8	<mdl< td=""><td>1.0</td><td>106.9</td><td><mdl< td=""><td>1.0</td><td>97.4</td></mdl<></td></mdl<>	1.0	106.9	<mdl< td=""><td>1.0</td><td>97.4</td></mdl<>	1.0	97.4
Chlorate	64.0	73.0	94.2	<mdl< td=""><td>5.0</td><td>99.0</td><td>1.6</td><td>5.0</td><td>104.0</td><td><mdl< td=""><td>5.0</td><td>103.7</td></mdl<></td></mdl<>	5.0	99.0	1.6	5.0	104.0	<mdl< td=""><td>5.0</td><td>103.7</td></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< td=""><td>5.0</td><td>102.0</td></mdl<>	5.0	102.0

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 the 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 Dionex 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 3. Determination of trace DBP anions and bromide in tap water.



Figure 4. Determination of trace DBP anions and bromide in spiked tap water.

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.

Conclusion

This application note described an IC method that used an electrolytically generated potassium hydroxide eluent combined with a hydroxide-selective Dionex IonPac AS19 column for the determination of trace DBP anions and bromide. This method used suppressed conductivity



Figure 5. Determination of trace DBP anions and bromide in bottled drinking water B.

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 Dionex IonPac AS9-HC column, as described in Method 317.0. However, comparable sensitivities for both the Dionex IonPac 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 Dionex IonPac 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 Dionex IonPac 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.



Figure 6. Determination of trace DBP anions and bromide in spiked bottled drinking water B.

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Suppliers

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